



SCIENCEDOMAIN international www.sciencedomain.org

Assessment of the Protein Quality and Mineral Bioavailability of *Dacryodes edulis* Seed and Seed Coat Mixture

C. U. Ogunka-Nnoka¹, P. U. Amadi^{1*}, P. C. Ogbonna¹ and P. O. Ogbegbor¹

¹Department of Biochemistry, University of Port Harcourt, Choba, Rivers State, Nigeria.

Authors' contributions

This work was carried out in collaboration between all authors. Author CUON designed the study, Author PUA performed the statistical analysis, wrote the protocol, wrote the first draft of the manuscript and managed the literature searches. Authors PUA and PCO managed the analyses of the study. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JSRR/2017/32405 <u>Editor(s)</u>: (1) Fu-Feng Liu, Department of Biochemical Engineering, School of Chemical Engineering and Technology, Tianjin University, Tianjin, P.R. China. <u>Reviewers</u>: (1) Sanjay Mishra, IFTM University, India. (2) Dra Fabiana América Silva Dantas de Souza, Federal Rural University of Pernambuco, Brazil and University of Pernambuco (Campus Mata Norte), Brazil. Complete Peer review History: <u>http://www.sciencedomain.org/review-history/18786</u>

Original Research Article

Received 24th February 2017 Accepted 17th March 2017 Published 25th April 2017

ABSTRACT

Aim: This study was carried out to evaluate the amino acid composition, protein quality, and mineral content of *Dacryodes edulis* seed and seed coat mixture.

Methodology: Ethanol extract of the sample was obtained and analyzed using HPLC amino acid auto analyzer for the evaluation of amino acids, and an atomic absorption spectrophotometer for the mineral content analysis.

Results: Leucine (7.68 g/100 g), lysine (6.44 g/100 g) and arginine (5.08 g/100 g) were the highest occurring essential amino acids, while glutamic acid (15.06 g/100 g), aspartic acid (11.73 g/100 g) and proline (4.09 g/100 g) were the highest occurring non-essential amino acids. Results for the amino acid groups followed the order; Total non-essential amino acid (TNEAA) > Total essential amino acid (TEAA) with His > Total essential amino acid (TEAA) without His > Total essential amino acid (TAAA) > Total branched chain amino acid (TBCAA) > Total basic amino acid (TBAA) > Total aromatic amino acid (TAAA) > Total sulphur amino acid (TSAA). For the protein quality indices, the predicted protein efficiency ratios (P-PERs I, II, and III) were 2.63, 2.65, and 2.06 respectively, while the Essential amino acid index (EAAI) and Leu/IIe were 1.51 and 1.89 respectively. Only

tryptophan exceeded the reference scoring pattern. The result of elemental analysis showed calcium, phosphorus, magnesium, and zinc as the four highest occurring minerals, while only the Ca/P, Ca/K, Ca/Mg, and [Ca]/[Phy] results fell within their reference values. **Conclusion:** This study has shown that *D. edulis* seed and seed coat mixture lacks majority of the recommended mineral proportions, and requires additional supplementation to equal the amino acid scoring patterns, but can be harnessed to ameliorate tryptophan related deficiencies.

Keywords: Dacryodes edulis; amino acids; protein quality; minerals; phytate.

1. INTRODUCTION

Dacrvodes edulis is an evergreen shade loving species of tropical humid non-flooded forest [1] indigenous to countries like Liberia, Nigeria, and Cameroon [2]. The plant belongs to the Burseracea family possessing numerous nutritional and medicinal benefits [3], where the seeds are utilized for the propagation of its fruits. The fruit is made up of a seed enclosed by a seed coat and then a pulpy pericarp. The pulp is the edible portion of the fruit that can be consumed cooked, raw, grilled in an oven, or roasted in hot ash. In Nigeria, the pulp can be eaten alone or with fresh maize. The nutritional importance of the fruit has been mainly due to the content of the pulp which is rich in amino acids, vitamin C, and fatty acids [4]. Both the seed and fruit are rich in proteins, fats and oil, and vitamins and minerals which makes it an important source of nutrition, capable of promoting its commercialization [5] and to possibly strengthen food security as well as reduce poverty. The fruits are also utilized for the production of edible oils used for cosmetic, nutritional, and pharmaceutical purposes [6,7,8]. The seeds of D. edulis contains about 18-70% oil which are mainly arachidonic acid along with other fatty acids [9], making the seed comparable to other popularly consumed seed sources of edible oils [10]. The applicability of the seed as a food supplement has also been investigated [11]. Since the seed and seed coat are usually discarded after consumption of the pulp, its utilization for various nutritional and medicinal purposes will neither threaten food supply nor cause food shortage. Besides this, due to worldwide need for cheap sources of nutrients especially proteins for low income groups in underdeveloped and developing countries, efforts are now vastly tailored towards the development of low cost proteins of plant origin [12]. Protein and calorie malnutrition is regarded as one of the major challenges faced by developing countries more especially for infants and pregnant women. In this case, the availability of plant sources of proteins becomes vital due to

high costs of animal sources of proteins. Notwithstanding that conventional legumes have been very helpful sources of proteins in these countries, their production is also presently insufficient to carter for the teeming population as well as provide for animal feed [13]. Hence, the search for unconventional leguminous sources of proteins remains a continuous activity. Recent studies on D. edulis fruit have mostly accounted for the production and characterization of the seed oil [14,10] whereas the mineral, amino acid, and protein quality evaluations are yet to be extensively studied. In fact, as of the time of this present study, no study has been carried out on the nutritional assessment of the mixture of D. edulis seed and seed coat. It is on this basis that this study was undertaken to provide information on the amino acid composition, protein quality, and mineral content of D. edulis seed and seed coat mixture.

2. MATERIALS AND METHODS

2.1 Sample Collection and Identification

The seed and seed coat of *Dacryodes edulis* used for the study were obtained from Choba Market in Obio-akpor L.G.A, River State Nigeria and identified at the Department of Plant Science and Biotechnology, University of Port Harcourt, Choba, Rivers State, Nigeria.

2.2 Sample Preparation

The seed and seed coat were washed thoroughly and allowed to dry at room temperature. The dried seed and seed coat (200 g) were oven dried at a temperature of 60°c between 3 to 4 days, ground together to fine powder with an electric grinder, packaged in air tight glass jar, and stored at room temperature to prevent moisture and dust, until analysis were carried out.

2.3 Preparation of Extract

The powdered seed and seed coat mixture (100 g) was introduced into a soxhlet extractor

with a reflux condenser attached, and extracted with 500ml of ethanol for 12 hrs. The ethanol solvent was removed by placing the extract on a rotary evaporator, thus concentrating the extract.

2.4 Amino Acid Determination

The amino acid analysis was carried out using a HPLC amino acid analyzer (Sykam-S7130) according to the method of Ijarotimi and Olopade [15]. Hydrolysates of the sample were obtained following the procedure of Moore and Stein [16]. The sample (200 mg) was transferred into a hydrolysis tube containing 5 ml of 6 N HCL, and afterwards tightly closed and incubated for 24 h at 110°C. After incubation and filtration, the filtrate (200 ml) was evaporated to dryness at 140°C for one hour and diluted with 1 ml of 0.12 N, citrate buffers (pH 2.2) similar to the amino acid standards. Then, the sample hydrolysate (150 µl) was injected in a cation separation column at 130°C. Ninhydrin solution and an eluent buffer (containing solvent A, pH 3.45 and solvent B, pH 10.85) were delivered simultaneously into the high temperature 16m length reactor coil at a flow rate of 0.7 ml/min. The buffer/ninhydrin mixture was heated in the reactor for 2 minutes at 130°C and the mixture were detected at wavelengths of 570 nm and 440 nm on a dual channel photometer. The amino acid composition was calculated from the areas of standards obtained from the integrator and expressed as percentages of the total protein.

To determine the tryptophan content, sample aliquot (100 mg) was dispersed into glass ampoules together with 1 ml 5 M NaOH. The ampoules were flame sealed and incubated at 110°C for 18 hours. The tryptophan contents of the alkaline hydrolysates were determined calorimetrically using the methods of Spies and Chambers [17] as modified by Rama Rao et al. [18].

2.5 Determination of Protein Quality Parameters

The method of Adeyeye [19] was applied for the determination of TAA (Total Amino Acid), TEAA/TAA (ratio of total essential amino acid to total amino acid), TNEA/TAA (ratio of total non-essential amino acid to total amino acid), TArAA (Total aromatic amino acid), TBAA (Total basic amino acid), TAAA (Total acidic amino acid), and TSAA (Total sulfur-containing amino acid).

The regression equation methods of Chavan et al. [20] was followed for the determination of the predicted PER values of *D. edulis* seed and seed coat mixture;

- I. PER = 0.684 +0.456(Leu) 0.047(Pro)
- II. PER = -0.468 + 0.454(Leu) 0.105(Tyr)
- III. PER = -1.816 + 0.435(Met) + 0.780(Leu) + 0.211(His) 0.944(Tyr).

The essential amino acid index (EAAI) was calculated following the method of Steinke et al. [21], as shown below;

EAAI=

 $9\sqrt{\frac{mg \ of \ Lys \ in \ 1g \ of \ test \ protein \times 8 \ essential \ amino \ acids + His}{mg \ of \ Lys \ in \ 1g \ of \ Reference \ protein}}}$

The amino acid scores were calculated as the ratio of 1 mg of amino acid per g of test protein to 1 mg of amino acid per g of standard protein $\times 100$.

The reference pattern of essential amino acid requirements (g/100 g of protein) as provided by Adeyeye et al. [22] was used as the standard.

2.6 Mineral Content, Mineral Ratio and Phytate Determination

Wet digestion of samples (5 ml) using a mixture of concentrated HNO₃ and 60% (v/v) HClO₄ was carried out according to the method of AOAC [23] where the organic matter in the sample was digested and afterwards diluted to a final volume of 25 ml with deionized distilled water. The levels of Na, K, Ca, Fe, Mg, P, Mn, Cu, and Zn in the samples were thus evaluated using an atomic absorption spectrophotometer (Buck Scientific model 210 VGP) and flame photometer (Jenway model). The sulphate contents of the food samples were determined turbidimetrically according to AOAC [24]. The chloride level was determined titrimetrically using the method of AOAC, [24]. The mineral ratios were determined as described by David [25], while the phytate content was evaluated using the method of Schroeder et al. [26]. The method of Igwe et al. [27] was employed for the calculation of [phytate]/[Zn], and [Ca]/[phytate], as described below:

[phytate]/[Zn] = [phytate (mg/100 g) / 660] /[zinc (mg/100 g) / 65.38] [Ca]/[phytate] = [calcium (mg/100 g) / 40.08] / [phytate (mg/100 g) / 660]

3. RESULTS AND DISCUSSION

The essential and non essential amino acid constituent of D. edulis seed and seed coat mixture are presented in Table 1. Leucine, lysine, and arginine were the highest occurring essential amino acids recorded in Table 1. The occurrence of leucine in this present study was in line with the reports of Olaofe and Akintayo, [28] that observed leucine as the most concentrated amino acid in Nigerian plant products. Ogungbenle and Oladipe [29] recorded much higher contents of leucine, lysine, and arginine from Nicker beans. Aremu et al. [30] reported lower contents of these amino acids in cranberry beans and cowpea. Leucine is a branched chain amino acid that induces the activation of mTOR as well as promotes the stimulation of muscle protein synthesis [31]. Fontana et al. [32] have reported that adiposity results from the decreased dietary intake of leucine. Smriga et al. [33] have hypothesized that lysine is an anxiolytic agent through the reduction of stress markers like cortisol, as well as reduces anxiety induced diarrhea. Further, Chen et al. [34] have shown that immunodeficiency results from deficiency of lysine, while rich dietary sources of lysine have been found helpful against incidence of stroke. as well as normalizes blood pressure [35]. Arginine has well known roles in hormone release, wound healing, cell division and other immune functions [36,37,38]. The phenylalanine, valine, threonine, and isoleucine concentration of D. edulis seed and seed coat mixture recorded in Table 1 were 4.69, 4.55, 4.28, and 4.06 g/100 g respectively. Luffa cylindrica seed [39] produced lower phenylalanine and valine contents when compared to the results of this study for D. edulis seed and seed coat mixture. However, E. gigas [29] provided much higher phenylalanine and valine content than that of D. edulis seed and seed coat mixture evaluated in this study. Seeds from Bixa orellana and Hibiscus esculentus were reported by Glew et al. [40] to contain higher threonine content than D. edulis seed and seed coat mixture shown in this study. From the reports of Adeyeye, [19] cooked and roasted seeds of Arachis hypogea produced lower isoleucine contents while the raw samples of the seed had comparable threonine content when compared to those for D. edulis seed and seed coat mixture presented in this study. Phenylalanine is the precursor for the synthesis of catecholamines, while valine promotes insulin sensitivity and as well, essential for the selfrenewal of hematopoetic stem cell [41]. Titchenal et al. [42] observed that deficiency of threonine

leads to neurologic dysfunction and lameness while Nishimura et al. [43] reported that supplementation with isoleucine causes a reduction in lipid mass and reduces weight gain. Histidine, methionine and tryptophan were the least occurring amino acid in D. edulis seed and seed coat mixture, with tryptophan, the limiting amino acid. The occurrence of histidine shown in this study was in similar proportions to defatted walnut [44] but higher than those for the endosperm and sarcotesta portions of Carica papaya seeds [45]. The methionine contents of D. edulis seed and seed coat mixture reported in this study were higher than the findings of Bolanle et al. [46] for P. guineense and B. eurycoma, while the amount of tryptophan recorded in *D. edulis* seed and seed coat mixture were comparable to that of Bombax constatum, but higher than that of C. papaya seeds [45]. Bolanle et al. [46] observed that histidine is a precursor for histamine necessary for inflammation while methionine is required for choline synthesis that forms lecithin and other phospholipids in the body. Tryptophan, in addition to its importance in protein synthesis, serves as a precursor for niacin, and serotonin. Glutamic acid (15.06 g/100 g), and aspartic acid (11.73 g/100 g), were among the highest occurring non essential amino acids in the sample. Both the glutamic acid and aspartic acid of the sample evaluated in this present study were comparable to that of Adansonia digitata [40], but higher than the aspartic and glutamic acids contents of the legumes; Brachystegia eurycoma and Piper guineense [46]. Proline, serine, and alanine contents of D. edulis seed and seed coat mixture were 4.09, 3.83, and 3.83 g/100 g respectively (Table 1). These values were lower than those for Pumpkin and Tiger nut seeds [47]. Tyrosine, glycine and cysteine were the least constituent amino acids of D. edulis seed and seed coat mixture. Notwithstanding the level of occurrence of these non-essential amino acids, it is expected that in addition to food sources, the amount synthesized by the body can provide sufficiently for the dietary needs of the amino acids.

The total amino acid, amino acid groups, percentage compositions and protein quality indices of *D. edulis* seed and seed coat mixture were shown in Table 2. The total amino acid (88.41) was comprised of 46.58 g/100 g (52.69%) total non-essential amino acid (TNEAA), 41.83 g/100 g (47.31%) total essential amino acid (TEAA) with His, and 39.11 g/100 g (44.23%) total essential amino acid (TEAA)

Ogunka-Nnoka et al.; JSRR, 14(1): 1-11, 2017; Article no.JSRR.32405

Table 1. Amino acid composition (g/100 g) of

without His. These percentage compositions clearly imply that the amount of total essential amino acids were lower than that of non essential amino acids. Also, it shows that histidine had a percentage composition of 3.08%. The total amino acid contents in this study was lower than those reported for Mucana puriens seed [48], and Lophira lanceolata seeds [49], but higher than fermented cocoa nibs (70.8 g/100 g) and B. sapida (48.13 g/100 g) [50]. In addition, the report for TNEAA, TEAA with and without His, for B. eurycoma and P. guineense [46] were lower than those reported for of D. edulis seed and seed coat mixture. The total essential amino acid without His (39.11 g/100g) met the FAO requirements (35.0 g/100 g) for essential amino acids without Histidine [51]. Furthermore, the occurrence of TArAA (9.19 g/100 g) in this study was ideal for infants according to the reports of Adeyeye et al. [50] while the TSAA (2.64 g/100 g) recorded in this study fell below that (5.8 g/100 g) for Pre-school children [51]. The TAAA (27.69 g/100 g) was higher than the TBAA (14.24 g/100 g) indicating that the protein is probably acidic. The TBCAA (16.29) was higher than TBAA, TArAA and TSAA. Monirujjaman and Ferdouse, [52] posited that BCAAs participates in skeletal muscle degradation and protein synthesis, and together with ArAAs may influence an individual's behavior through the influence on the synthesis of some neurotransmitters [53,54]. FAO/WHO, recommends the predicted protein [55] efficiency ratio (PER) as one of the quality parameters used for protein evaluation. P-PERs values were higher than that of sorghum and millet ogi (0.27 and 1.62 respectively) [56], and pigeon pea (1.82) and cowpea (1.21) [57]. The EAAI is tool useful for the protein quality evaluation of food formulations, notwithstanding that the application of various processing methods limits its usefulness in assessing differences in protein quality [58]. The EAAI recorded in this study (1.51) was higher than that for soy flour [58], raw and cooked groundnut seeds [19], and B. eurycoma, and P. guineense [46]. The Leu/Ile balance is a useful indicator in the regulation of niacin and tryptophan metabolism [59]. The Leu/Ile (1.89) recorded in this study shows higher leucine bioavailability, thus indicating that this sample can be useful during leucine related deficiency diseases. The values recorded for TEAA (+His) /TAA (0.47) and TEAA (-His) /TAA (0.44) shows that His contribution to the TEAA is minor, while the TBAA/TAAA of <1 shows the acidity of the protein content of the sample under study.

D. edulis seed and seed coat mixture		
Amino acid	Value (g/100 g)	
Essential amino acids		
Lysine	6.44±0.13	
Histidine	2.72±0.17	
Arginine	5.08±0.11	
Methionine	1.25±0.12	
Threonine	4.28±0.04	
Isoleucine	4.06±0.05	
Valine	4.55±0.02	
Leucine	7.68±0.04	
Tryptophan	1.08±0.05	
Phenylalanine	4.69±0.45	
Non-essential amino acids		
Alanine	3.83±0.01	
Aspartic acid	11.73±0.59	
Cysteine	1.39±0.11	
Glutamic acid	15.06±0.61	
Glycine	3.23±0.12	
Proline	4.09±0.06	
Tyrosine	3.42±0.13	
Serine	3.83±0.05	

Values are means and standard deviations of triplicate determinations

The amino acid scores of *D. edulis* seed and seed coat mixture are shown in Table 3. Phenylalanine + tyrosine produced the highest score followed by lysine. Only the tryptophan scores (1.08) of *D. edulis* seed and seed coat exceeded the scoring pattern (1.0). Amino acid score provides information on the levels of individual amino acids in a food relative to the need for that amino acid. Methionine + cysteine were observed to be the limiting amino acids with a score of 0.75. This implies that the sample is to be consumed for 4.67 times to attain the levels of the scoring pattern for Methionine + cysteine.

The mineral composition of *D. edulis* seed and seed coat mixture is shown in Table 4. The result show that calcium (152.23 mg/100 g) is the most abundant element found in D. edulis seed and seed coat mixture, followed by phosphorus (50.53 mg/100 g), and magnesium (19.11 mg/100 g). Seeds of Cucurbita pepo [60] contained much lower calcium content, comparable phosphorus content, but a higher magnesium content, when compared to the result of this present study for D. edulis seed and seed coat mixture. The reports of Agomuo et al. [61] showed higher Ca and Mg contents of both Treculia africana (breadfruit) and Citrullus vulgaris (melon seeds) than those for D. edulis seed and seed coat mixture presented in this study. Calcium plays central roles for the normal

functioning of muscles and nerves, while deficiency of magnesium leads to serious muscle twisting that could cause convulsion and possibly death [62]. Phosphorus occurs in inorganic form in phospholipids, ATP, nucleic acids etc. Sodium (7.68 mg/100 g) and potassium (4.67 mg/100 g) were the least occurring macrominerals in this study. Bambara groundnut [63] and cowpea [64] had comparable sodium contents with those for D. edulis seed and seed coat mixture presented in this study, however, the sodium content of red kidney bean [65] pinto bean [66], and Pigeon pea [67] were much higher than that of D. edulis seed and seed coat mixture recorded in this study. The potassium content of cowpea, melon, and groundnut [68] were similar to that of D. edulis seed and seed coat mixture obtained in this study. Sodium and potassium are important for the maintenance of acid-base balance. While sodium is the principal cation in extracellular fluids, potassium is the principal cation in intracellular fluids. The result further indicated that zinc (10.76 mg/100 g) and iron (6.97 mg/100 g) were the most abundant micro-minerals evaluated in D. edulis seed and seed coat mixture presented in Table 4. The zinc and iron content of soya bean reported by Oluyemi et al. [68] were much lower than the findings of this study for *D. edulis* seed and seed coat mixture. Also, the zinc and iron content of African breadfruit [69] was lower than those for D. edulis seed and seed coat mixture in this study. Zinc is found as a cofactor of many dehydrogenases and polymerases, and is useful during the breakdown of macronutrients and cell replication [70]. McDonald et al. [71] posited that iron deficiency leads to anemia, while in children, the deficiency causes learning disabilities. The manganese content of D. edulis seed and seed coat mixture in this study (0.88 mf/100 g) was lower than that of Sphenostylis stenocarpa [72] but higher than that of Cucurbita pepo [60]. According to Chandra, [73] manganese is an essential constituent of enzymes involve in metabolism of pyruvate, formation of urea, and biosynthesis of connective tissue. Copper occurs in the blood plasma in the form of erythrocuprin, and functions during the onset of haemopoiesis.

Table 2. Amino acid groups (g/100 g) and percentage contents, and protein quality parameters in seed and coat of *D. edulis* mixture

Parameters		Value (g/100 g)
Amino acid groups		
Total amino acid (TAA)		88.41
Total non-essential amino acid (TNEAA)		46.58
Total essential amino acid (TEAA) with His		41.83
Total essential amino acid (TEAA) without His		39.11
Total aromatic amino acid (TArAA)		9.19
Total basic amino acid (TBAA)		14.24
Total acidic amino acid (TAAA)		27.69
Total sulphur amino acid (TSAA)		2.64
Total branched chain amino acid (TBCAA)		16.29
% Amino acid contents		
%TNEAA		52.69
%TEAA(with His)		47.31
%TEAA(without His)		44.23
%TArAA		10.39
%TBAA		16.11
%TAAA		31.31
%TSAA		2.98
%TBCAA		18.42
Protein quality indices		
Predicted protein efficiency ratios (P-PERs)	I	2.63
	II	2.65
	III	2.06
Essential amino acid index (EAAI)		1.51
Leu/lle		1.89
TEAA (+His) /TAA		0.47
TEAA (-His) /TAA		0.44
ΤΒΑΑ/ΤΑΑΑ		0.51

The copper content of D. edulis seed and seed coat mixture in this study was found higher than Dacryodes edulis pulp [74] and melon [75] but lower than cowpea [76] and soya bean [77]. The cadmium and lead content of D. edulis seed and seed coat mixture in this study were 0.03 mg/100 g and 0.58 mg/100 g respectively. The cadmium content was comparable to that of Allanblackia floribunda and Garcinia kola [78], while the lead content were higher than that of Garcinia kola, but lower than those for Poga oleosa and Allanblackia floribunda. Asagba and Eriyamremu [79] found that cadmium inhibits some antioxidant enzymes which imply that its occurrence in food materials should be at its barest minimum. Also, some studies have identified а relationship between lead consumption and reproductive and hypothalamicpituitary unit toxicities [80] thus requiring its absence from food materials.

Table 3. Amino acids scores (g/100 g protein) of the seed and coat of *D. edulis* mixture

Amino acids	Scoring pattern	Value
Isoleucine	4.0	1.02
Leucine	7.0	1.09
Lysine	5.5	1.17
Methionine +cysteine	3.5	0.75
Phenylalanine + tyrosine	6.0	1.35
Threonine	4.0	1.07
Tryptophan	1.0	1.08
Valine	5.0	0.91

Provisional amino acid scoring pattern (g/100 g of protein) [22]

The calculated mineral ratios and phytatemineral ratios are shown in Table 5. The Na/K ratio was found to be 1.64. Monitoring the Na/K ratio is important for the prevention of high blood pressure, which in food substances, a value <1 is recommended. This implies that the consumption of D. edulis seed and seed coat mixture might not be suitable for the management of hypertension. The Ca/P (3.04), Ca/K (32.59), and Ca/Mg ratio in this study exceeded the minimum ratio required for adequate intestinal absorption of Ca. This suggests that the consumption of D. edulis seed and seed coat mixture might promote strong bone formation. The Zn/Cu ratio presented in Table 5 is (107.6). A high Zn/Cu (> 16) may lead to poor copper absorption which can alter the oxidant/antioxidant balance [81]. The Fe/Cu ratio (69.7) in this study exceeded the normal range of 0.2-1.6. Elevated Fe/Cu ratio can cause excessive production of free radicals that could cause the impairment of normal

cellular activity (David, 2010). The phytate composition (20.43 mg/100 g) of the sample under study was lower than those for *Daniellia oliveri* kernel seed [82] and *Jatropha curcas* kernel seed [83]. Phytates binds to divalent cations thus preventing their absorption and utilization by the body. The [Phy]/[Zn] (0.19) indicates a possible reduced bioavailability of zinc since it was less than 1 while the result obtained for [Ca]/[Phy] content was in line with the reference value of \geq 6 by Adeyeye et al. [22] indicating that the calcium bioavailability was not impaired.

Table 4. Phytate and mineral composition (mg/kg) and mineral ratios of seed and coat of *D. edulis* mixture

Parameters	Value (mg/100 g)
Phytate	20.43±4.07
Macro minerals	
Sodium (Na)	7.68±0.21
Potassium (K)	4.67±0.25
Calcium (Ca)	152.23±10.37
Magnesium (Mg)	19.11±0.75
Phosphorus (P)	50.53±6.34
Micro minerals	
Manganese (Mn)	0.88±0.12
Copper (Cu)	0.10±0.03
Zinc (Zn)	10.76±0.5
Cadmium (Cd)	0.03±0.01
Lead (Pb)	0.58±0.04
Iron (Fe)	6.97±0.01

Values are expressed as means and standard deviations of triplicate determinations

Table 5. Calculated mineral and phytatemineral ratios of *D. edulis* seed and seed coat mixture

Parameter	Ratio
Na/K	1.64
Ca/P	3.04
Ca/K	32.59
Zn/Cu	107.6
Ca/Mg	7.96
Fe/Cu	69.7
[Phy]/[Zn]	0.19
[Ca]/[Phy]	1220.70

4. CONCLUSION

This study has shown that *D. edulis* seed and seed coat mixture contains a higher proportion of non essential amino acid to essential amino acids, with glutamate and aspartate the most predominant, and only tryptophan exceeded the reference scoring pattern. Though calcium,

magnesium and zinc were the most predominant minerals, *D. edulis* seed and seed coat mixture lacks majority of the recommended mineral proportions, requires additional supplementation to equal the amino acid scoring patterns, but can be harnessed to ameliorate tryptophan related deficiencies.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

- Leaky RRB, Atangana AR, Kengnni E, Waruhiu AN, Usoro C, Anegbe PO, Tchoundjeu Z. Domestication of Dacryodes edulis in West and Central Africa: Characterization of genetic variation Trees. Livelihood. 2002;12:57-71.
- Anegbeh PO, Ukafor V, Usoro C, Tchoundjeu Z, Leakey RRB, Schreckenberg K. Domestication of *Dacryodes edulis*: Phenotypic variation of fruit traits from 100 trees in south eastern Nigeria. New Forests. 2005;29:149-160.
- Ujowundu CO, Kalu FN, OE Okafor Agha NC, Alisi CS, Nwaoguikpe RN. Evaluation of the chemical composition of *Dacryodes edulis* (G Don) seeds Int. J. Biol. Chem. Sci. 2010;4(4):1225-1233.
- 4. Leakey RRB. Potential for novel food products from Agroforestry trees: A review. Food Chem. 1999;66:1-14.
- Ogunmoyole T, Kade IJ, Johnson OD, Makun OJ. Effect of boiling on the phytochemical constituents and antioxidant properties of African pear *Dacryodes edulis* seeds *in vitro*. African Journal of Biochemistry Research. 2012;6(8):105-114.
- Kapseu C, Avouampo E. Djeumako B. Oil extraction from *Dacryodes edulis* (G Don). H J Lam fruit Forests, Trees and Livelihoods. 2002;12:97-104.
- Mbofung CMF, Silou T, Mouragadja I. Chemical characterization of Safou (*Dacryodes edulis*) and evaluation of its potential as an ingredient in nutritious biscuits. Forests, Trees and Livelihoods. 2002;12:105-118.
- Kalenda DT, Missang CE, Kinkela T, Krebs HC, Renard CM. New developments in the chemical characterization of the fruit of *Dacryodes edulis* (G Don). H J Lam

Forests, Trees and Livelihoods. 2002;12: 119-124.

- Ajayi IA, Adesanwo O. Comparative study of the mineral element and fatty acid composition of *Dacryodes edulis* pulp and seed. World J Agric Sci. 2009;5:279-283.
- 10. Arisa NU, Lazarus A. Production and refining of *Dacryodes edulis* "native pear" seeds oil Afri J Biotechnol. 2008;7(9): 1344-1346.
- Obasi NBB, Okolie NP. Nutritional constituents of the seeds of the Africa pear (*Dacryodes edulis*). Food Chemistry. 1993; 46:297-299.
- Fatoumata T, Tidjani A, Camel L, Guo-Wei L, Yong-Hui S. Extraction, characterization, nutritional and functional properties of Roselle (*Hibiscus sabdariffa*) seed proteins. Songklanakarin J Sci Technol 2013;35(2):159-166.
- Siddhuraju P, Becker K. Antioxidant properties of various solvent extracts of total phenolic constituents from three different agro-climatic origins of drumstick tree (*Moringa oleifera* Lam). Journal of Agricultural and Food Chemistry. 2003;51: 2144–2155.
- Ikhuoria EU, Maliki M. Characterization of Avocado pear (*Persea americana*) and African pear (*Dacryodes edulis*) extracts. African Journal of Biotechniology. 2007;6 (7):950-952.
- Ijarotimi OS, Olopade AJ. Determination of amino acid content and protein quality of complementary food produced from locally available food materials in Ondo State, Nigeria. Mal J Nutr. 2009;15(1):87-95.
- Moore S, Stein WH. Chromatographic amino acids determination by the use ofautomatic recording equipment. Methods Enzymol. 1963;6:819-831.
- 17. Spies JR, Chamber DC. Chemical determination of tryptophan in proteins. Anal Chem. 1949;21:1249-1266.
- Rama Rao MV, Tara MR, Krishnan CK. Colour imetric estimation of tryptophan content of pulses. Journal of Food Science and Technology, (Mysore). 1974;11:13-216.
- Adeyeye EI. Effect of cooking and roasting on the amino acid composition of raw groundnut (*Arachis hypogaea*) seeds. Acta Sci Pol Technol Aliment. 2010;9(2):201-216.
- 20. Chavan UD, Mckenzie DB, Shahidi F. Functional properties of protein isolates

from beach pea (*Lathyrus maritius* L). Food Chemistry. 2001;74:177-187.

- 21. Steinke FH, Prescher EE, Hopkins DT. Nutritional evaluation (PER) of isolated soybean protein and combinations of food proteins. J Food Sci. 1980;45:323-327.
- 22. Adeyeye EI, Orisakeye OT, Oyarekua MA. Composition, mineral safety index, calcium, zinc and phytate interrelationships in four fast-foods consumed in Nigeria. Bull Chem Soc Ethiop. 2012;26(1):43-54.
- 23. AOAC. Official methods of analysis 15th edn, Washington DC, USA Association of official analytical chemists inch 400-2200 Wilson Boalevard, Arlinton Virginia USA. 1990;2:910-92.
- 24. AOAC. Standard official methods of analysis of the association of analytical chemists 14 Edn, Association of Analytic Chemists, Washington DC; 1984.
- 25. David LW. HTMA mineral ratios, a brief discussion of their clinical importance. Trace Elements Newsletter. 2010;21:1.
- 26. Schroeder WA, Kay LM, Mills RS. Anal Chem. 1990;22:760.
- Igwe CU, Ibegbulem CO, Nwaogu LA, Ujowundu CO, Okwu GN. Calcium, Zinc and phytate interrelationships in four lesser-known African seeds processed into food condiments. J Adv. Chem. 2013;4(2): 288-292.
- Olaofe O, Akintayo ET. Prediction of isoelectric points of legume and oilseed proteins from their amino acid compounds. J Techno-Sci. 2000;4:49-53.
- 29. Ogungbenle HN, Oyadipe OT. Compositional and amino acid profile of Nicker Bean (*Entada gigas*). Seeds British Biotechnology Journal. 2015;6(2):43-50.
- Aremu MO, Olaofe O, Akintayo TE. A comparative study on the chemical and amino acid composition of some Nigerian under-utilized legume flours. Pakistan Journal of Nutrition. 2006b;5(1):34-38.
- Etzel MR. Manufacture and use of dairy protein fractions. The Journal of Nutrition 2004;134(4):996S–1002S. PMID 15051860
- Fontana L, Cummings NE, Arriola A, Sebastian I, Neuman JC, Kasza I, Schmidt BA, Cava E, Spelta F, Tosti V. Decreased consumption of branched-chain amino acids improves metabolic health. Cell Reports. 2016;16:520–30.
- 33. Smriga M, Kameishi M, Uneyama H Torii K. Dietary L-lysine deficiency increases

stress-induced anxiety and fecal excretion in rats. J Nutr. 2002;132(12):3744-6.

- Chen C, Sander JE, Dale NM. The effect of dietary lysine deficiency on the immune response to Newcastle disease vaccination in chickens. Avian Dis. 2003;47(4):1346– 51.
- 35. Flodin NW. The metabolic roles, pharmacology, and toxicology of lysine. J Am Coll Nutr. 1997;16(1):7-21.
- Tapiero H, Mathé G, Couvreur P, Tew KD. "L-Arginine" (review). Biomedicine & Pharmacotherapy. 2002;56 (9):439–445.
- Witte MB, Barbul A. Arginine physiology and its implication for wound healing (review). Wound Repair and Regeneration. 2003;11(6):419–23.
- Stechmiller JK, Childress B, Cowan L. Arginine supplementation and wound healing (review). Nutrition in Clinical Practice. 2005;20(1):52–61.
- Oyetayo FL, Ojo BA. Food value and phytochemical composition of *Luffa cylindrica* seed flour. American Journal of Biochemistry. 2012;2(6):98-103.
- Glew RH, Dorothy JV, Cassius L, Louis EG, Garrett CS, Andrzej P, Mark M. Amino acid, fatty acid, and mineral composition of 24 indigenous plants of Burkina Faso. Journal of Food Composition and Analysis. 1997;10:205–217.
- Taya Y, Ota Y, Wilkinson AC, Kanazawa A, Watarai H, Kasai M, Nakauchi H, Yamazaki S. Depleting dietary valine permits nonmyeloablative mouse hematopoetic stem cell transplantation. Science. 2016;354(6316):1152–1155.
- 42. Titchenal CA, Rogers QR, Indrieri RJ, Morris JG. Threonine imbalance, deficiency and neurologic dysfunction in the kitten. J Nutr. 1980;110(12):2444-59.
- Nishimura J, Masaki T, Arakawa M, Seike M, Yoshimatsu H. Isoleucine prevents the accumulation of tissue triglycerides and upregulates the expression of PPARγ and uncoupling protein in diet-induced obese mice. Journal of Nutrition. 2010;140(3): 496–500.
- 44. Xiaoying M, Yufei H. Composition, structure and functional properties of protein concentrates and isolates produced from walnut (*Juglans regia* L). Int J Mol Sci. 2012;13(2):1561-1581.
- 45. Passera C, Spettoli P. Chemical composition of papaya seeds. Plant Food Hum Nutr. 1981;31(1):77-83.

- 46. Bolanle AO, Akomolafe SF, Adefioye A. Proximate analysis, mineral contents, amino acid composition, anti-nutrients and phytochemical screening of *Brachystegia eurycoma* Harms and *Pipper guineense* Schum and Thonn. American Journal of Food and Nutrition. 2014;2(1):11-17.
- 47. Glew RH, Glew RS, Chuang L-T, Huang Y-S, Millson M, Constans D, Vanderjagt DJ. Amino acid, mineral and fatty acid Content of pumpkin seeds (*Cucurbita* spp) and *Cyperus esculentus* nuts in the Republic of Niger. Plant Foods for Human Nutrition. 2006;61:51–56.
- Fathima KR, Soris PT, Mohan VR. Nutritional and anti-nutritional assessment of *Mucana puriens* (L) DC var puriens an underuntilized tribal pulse. Advances in Bioresearch. 2010;1(2):79-89.
- 49. Idouraine A, Kohlhepp EA, Weber CW, Warid WA, Martinez-Tellez J. Nutrient constituent from eight lines of naked squash (*Cucurbilta pepo L*). Journal of Agricultural Food Chemistry. 1996;44:721-724.
- 50. Adeyeye EI, Akinyeye RO, Ogunlade I, Olaofe O, Boluwade JO. Effect of farm and industrial processing on the amino acid profile of cocoa beans. Food Chemistry Journal. 2010;118:337-363.
- 51. FAO/WHO/UNU: Energy and Protein requirements Technical report series. Geneva. 1985;275:204.
- 52. Monirujjaman M, Ferdouse A. Metabolic and physiological roles of branched-chain amino acids. Advances in Molecular Biology. 2014;6. (Article ID 364976). Available:<u>http://dxdoiorg/101155/2014/364</u> 976
- Skeie B, Kvetan V, Gil KM, Rothkopf MM, Newsholme EA, Askanazi J. Branch-chain amino acids: Their metabolism and clinical utility. Critical Care Medicine. 1990;18(5): 549–571.
- 54. Fernstrom JD. Branched-chain amino acids and brain function. Journal of Nutrition. 2005;(135supplement)6:1539S–1546S.
- 55. FAO/WHO. Protein quality evaluation Report of Joint FAO/WHO Expert Consultation FAO Food and Nutrition Paper 51 FAO Rome; 1991.
- Oyarekua MA, Eleyinmi AF. Comparative evaluation of the nutritional quality of corn, sorghum and millet ogi prepared by modified traditional technique. Food Agric Environ. 2004;2(2):94-99.

- 57. Salunkhe DK, Kadam SS. Handbook of world food legumes, nutritional chemistry, processing technology and utilisation boca raton. CRC Press Florida; 1989.
- 58. Nielson SS. Introduction to the chemical analysis of foods. CBS Publishers and Distributors, New Delhi; 2002.
- 59. Ghafoorunissa S, Narasinga Rao BS. Effect of leucine on enzymes of the tryptophanniacin metabolic pathway in rat liver and kidney. Biochem J. 1973;134: 425-430.
- Elinge CM, Muhammad A, Atiku FA, Itodo AU, Peni IJ, Sanni OM, Mbongo AN. Proximate, mineral and anti-nutrient composition of pumpkin (*Cucurbita pepo* L) seeds extract. International Journal of Plant Research, 2012;2(5):146-150.
- 61. Agomuo EN, Amadi PU, Iheka CU, Duru MK. Impact of different processing methods on the micronutrient properties of selected legumes consumed in Eastern Nigeria. International Journal of Biological Research. 2017;5(1):10-14.
- 62. Hegarty V. Decisions in nutrition 5th edition Time mirrowmosby London. 1988;80-132.
- 63. Aremu MO, Olaofe O, Akintayo ET. Chemical composition and physiochemical characteristics of two varieties of bambara groundnut (*Vigna subterrenea*) flours. Journal of Applied Science. 2006a;6(9): 1900-1903.
- 64. Aremu MO, Olaofe O, Akintayo TE. A comparative study on the chemical and amino acid composition of some Nigerian under-utilized legume flours. Pakistan Journal of Nutrition. 2006b;5(1):34-38.
- 65. Audu SS, Aremu MO. Effect of processing on chemical composition of red kidney bean (*Phaseolus vulgaris*) flour. Pakistan Journal of Nutrition. 2011a;10(11):1069– 1075.
- Audu SS, Aremu MO. Nutritional composition of raw and processed pinto bean (*Phaseoulus vulgaris* L) grown in Nigeria. Journal of Food Agriculture & Environment. 2011b;9(3&4):72–80.
- 67. Arawande JO, Borokini FB. Comparative study on chemical composition and functional properties of three Nigerian legumes (jack beans, pigeon pea and cowpea) JETEAS. 2010;1(1):89–95.
- Oluyemi EA, Akinlua AA, Adenuga AA Odebajo MB. Mineral contents of some commonly consumed Nigerian foods. European J of Scientific Research. 2005; 6(2):11–15.

- 69. Olapade AA, Umeonuorah UC. Mineral, vitamin and antinutritional content of African breadfruit (*Treculia africana*) seeds processed with Alum and Trona. IOSR Journal of Environmental Science, Toxicology and Food Technology (IOSR-JESTFT). 2013;5(5):71-78.
- Arinola OG. Essential trace elements and metal binding proteins in Nigerian consumers of alcoholic beverages. Pakistan Journal of Nutrition. 2008;7(6): 763-765.
- 71. McDonald A, Edwards RA, Greenhulgh FD, Morgan CA. Animal nutrition prentices Hall, London. 1995;101-122.
- 72. Ndidi US, Ndidi CU, Olagunju A, Muhammad A, Billy FG, Okpe O. Proximate, antinutrients and mineral composition of raw and processed (boiled and roasted) *Sphenostylis stenocarpa* seeds from Southern Kaduna, Northwest Nigeria. ISRN Nutrition. 2014;9. (Article ID 280837).

Available:<u>http://dxdoiorg/101155/2014/280</u> 837

- 73. Chandra RK, Micro-nutrients and immune functions: An overview Annal New York Academic Science. 1990;587:9-16.
- 74. Amadi BA, Arukwe U, Duru MKC, Amadi CT, Adindu EA, Egejuru L, Odika PC. Phytonutrients and antinutrients screening of *D. edulis* fruits at different maturation stages. Journal of Natural Product Plant Resource. 2012;2(2):310-313.
- 75. Lawal OU. Effect of storage on the nutrient composition and the mycobiota of sundried water melon seeds (*Citrullus lanatus*). Journal of Microbiology, Biotechnology and Food Sciences. 2011;1(3):267-276.
- 76. Chinma CE, Alemede IC, Emelife IG. Physicochemical and functional properties

of some Nigerian cowpea varieties. Pakistan Journal of Nutrition. 2008;7(1): 186-190.

- Odumodu CU. Nutrients and anti-nutrients content of dehulled soybean continental. Journal of Food Science and Technology. 2010;4:38–45.
- Dike MC, Asuquo ME. Proximate, phytochemical and mineral compositions of seeds of Allanblackia floribunda, Garcinia kola and Poga oleosa from Nigerian rainforest. African Journal of Biotechnology. 2012;11(50):11096-11098.
- 79. Asagba SO, Eriyamremu GE. Oral cadmium exposure alters haematological and liver function parameters of rats fed a Nigerian like diet. J Nutr Envt Med. 2007; 16(1-3):267-274.
- Bonde JP, Joffe M, Apostoli P, Dale A, Kiss P, Spano M. Sperm count and chromatin structure in men exposed to inorganic lead: Lowest adverse effect levels. Occup Environ Med. 2002;59(4): 234-242.
- Zorica H, Aida C, Aldina K, Mirzeta S. Zinc/copper ratio in the medicinal plants. 17th International Research/Expert Conference Trends in the Development of Machinery and Associated Technology. TMT, Istanbul, Turkey; 2013.
- Hassan LG, Dangoggo SM, Umar KJ, Sadu I, Folorunsho FA. Proximate, minerals and anti-nutritional factors of *Daniellia oliveri* seed kernel. Chem class Journal. 2008;5:31-36.
- Azza AA, Ferial MA. Nutritional quality of Jatropha curcas seeds and effect of some physical and chemical treatments on their anti-nutritional factors. African Journal of Food Science. 2010;4(3):93-103.

© 2017 Ogunka-Nnoka et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history: The peer review history for this paper can be accessed here: http://sciencedomain.org/review-history/18786