

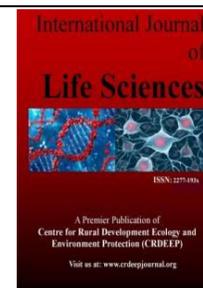
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Full Length Research Paper

Physical and Nutritional Properties of *Coula edulis*

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ABSTRACT

The physical, proximate, phytochemical contents, amino acid and micronutrient profile of *Dacryodes edulis* were investigated. The unshelled nuts have mean values of 3.64cm and 3.05cm for length and diameter respectively, while the shelled nut have mean values of 2.23cm and 1.92cm for length and diameter. The percent edible portion was 25% while the percent shell 75% of the whole nut, indicating a high shell content. The mean moisture, ash protein, fat Dietary fibre and carbohydrate contents were 14.61, 6.09, 4.6, 24.31, 4.27 and 45.67% respectively. The results show that sodium has the highest concentration 512.1mg/100g while copper recorded the lowest concentration with value 0.6983mg/100g. Vitamin E had the highest concentration (8.46mg/100g) and supplied 56% of the recommended dietary allowance (RDA) for a 100g sample, while vitamin K had the lowest concentration of 0.003184mg/100g and supplied 3% of RDA/100g sample. Vitamin E had the highest concentration (8.46mg/100g) and supplied 56% of the recommended dietary allowance (RDA) for a 100g sample, while vitamin K had the lowest concentration of 0.003184mg/100g and supplied 3% of RDA/100g sample. Vitamin C has the highest concentration 34.99mg/100g among the water-soluble vitamins. A 100g of *Coula edulis* would supply 39% RDA of vitamin C. The tannin content averaged 0.72%, while saponin was 0.18%. Alkaloid and phytate were 1.07%, 0.28% respectively. The highest scoring essential amino acid is leucine (7.10g/100gprotein) which supplied 101% of the recommended dietary allowance (RDA), while the limiting amino acid is methionine (1.5g/100gprotein) and had 43% amino acid chemical score.

Introduction

Almost all health problems experienced globally today are directly or indirectly related to nutrition, either as a causative, preventive or corrective factor. Therefore malnutrition or poor nutrition (which includes undernutrition, over nutrition and micronutrient deficiencies) is always a major contributor to low productivity through poor physical and mental health. Several efforts have been made by both governmental and nongovernmental agencies towards the alleviation of all forms of malnutrition, such as mandatory food fortification, nutrient supplementation, nutritional education, food diversification and more recently, biofortification. Food diversification is the option that has the potential to reduce malnutrition on a long term sustainable basis.

But it has received little attention due to overdependence on few popular refined starchy staples while most indigenous species remain underutilized and in some cases nearing extinction eg. *Treculia africana*, *Dacryodes edulis*, *Dioscorea spp*, *Xanthosoma spp*, *Coula edulis*, etc These foods have played important roles in the provision of adequate nutrition especially micronutrient supply for the indigenous peoples living in South

eastern Nigeria over generations. *Coula edulis* (African walnut) belongs to the family Olacaceae which comprises about 250 species (Mabberley, 1997). It is a medium sized, evergreen tree growing to a height of 25- 38 m with dense crown that can cast a deep shade. (Alan, 1999).

It is referred to as 'udi' in igbo, 'ekon' in Ibibio and Efik, 'Ivianledge' in Edo and 'asala' in Yoruba. The nuts serve as good source of nutrients for the local populace (Ekop and Eddy, 2005). The fleshy fruit is tasty but covered with a hard thick shell that makes the nut difficult to extract since the thick shell must first be cracked open.

Presently, *Coula edulis* is not commonly cultivated in homesteads and plantations and its potentials have not been well exploited. There are no known commercial products from them and the tree nut is nearing extinction since little is known about it by many. The main objective of this study is to evaluate the physical and nutritional characteristics of *Coula edulis*. The data obtained in this study will be useful in the finding the right application of the nut in commercial food products.

Materials and Methods

Materials procurement

Coula edulis used in this present study was obtained from Ini local government area of Akwa Ibom State, Nigeria. They were cleaned to remove extraneous materials. The edible meat within the shell was obtained by cracking open the shell with a small hammer.

Physical Examination

One hundred grams of the nuts (shelled and unshelled) were weighed using an electronic balance, The number of nuts contained in each 100 g portion was counted and recorded. The mean weight of individual nut was then calculated by dividing 100g by the number of nuts. The length and diameter of the nuts and the shell thickness were measured using vernier callipers. The shelled nuts were dried at 60°C for 30 min in a dry air oven, milled with a locally made attrition mill and used for subsequent analysis

Proximate Analysis

The moisture, fat, protein, dietary fibre, ash and carbohydrate were measured using AOAC (2010) method.

Determination of mineral profile

The method described by Onwuka (2005) was used in the determination of mineral content of the flour sample of *Coula edulis*. The milled sample (0.5g) was weighed into a pre -acid rinsed digest tube. 10ml of 6M HCl was added and heated to dryness in a water bath. The residue was dissolved in a solution of 10ml of 6M HNO₃ warmed on a water bath and filtered using a Whatman filter paper into a 100cm³ calibrated flasks. The filter paper was washed with distilled water and the filtrate was made up to the 100cm³ mark. The digest was used for determination of calcium and magnesium, potassium and sodium by the flame photometry method. Other metals such as Cu, Fe, Zn, Mn were determined using the atomic absorption spectrophotometric method.

Determination of Ascorbic Acid (Vitamin C)

The method described by Uzomah *et al.*, (2002) was used. Two grams (2g) of the sample was accurately weighed and dispersed in 350ml of 5% meta-phosphoric acid (MPA) for 3min. A 200ml centrifuge cup was filled with the solution and centrifuged for 15min at 2500rpm. Twenty five milliliter (25ml) of the aliquots was pipetted into a 50ml beaker. One milliliter (1.0ml) of 50% sulphuric acid was added to the beaker (to reduce the pH to 0.6) and 2.9ml formaldehyde was added. Then 2 x 10ml of the aliquots was pipetted into a 25ml test tube and allowed to stay for 8min before titrating with the dye (2,6-dichlorophenol-indophenol). The procedure was repeated on 25ml of 5% MPA to determine the dye blank.

Calculation:

$$\text{Ascorbic acid (mg/100g)} = \frac{T_1 \times F \times 28.9 \times S \times 100}{10 \times 25 \times W}$$

Where; T₁ = volume (ml) of dye corresponding to the ascorbic acid content in 10ml aliquot.

F = dye factors for dye used

28.9 = volume (ml) of extract (25ml) sulphuric acid and formaldehyde at pH 0.6

S = total volume (ml) of extract (vol. of extract, 350ml) plus moisture content of sample

W = weight (g) of sample taken

Determination of B-Complex Vitamins

The method described by Augustine, (1985) was used in the determination of B-complex vitamins; thiamine (B₁), Riboflavin(B₂), Niacin(B₃), pantothenic acid (B₅), pyridoxine (B₆), folic acid (B₉), cyanocobalamin (B₁₂).

Procedure:

The sample was homogenized using a mixer blender, and 2.5g was weighed into 10ml volumetric flask and 5ml of the buffer solution was added. The mixture was shaken for 5min using mechanical shaker/sonicator. The solution was made up to mark with buffer, filtered and injected into HPLC (HPLC/W₁/08). The standards of each vitamin were prepared and injected into the HPLC column. The absorbance of the standards and sample were read using UV detectors at 275nm wavelength. The calibration curve was plotted using concentration versus absorbance.

$$\text{Concentration mg/l} = \frac{\text{mm/ml (from calibrated curve)} \times 1000 \times \text{dilution}}{\text{Weight of sample}}$$

Determination of Carotenoids

Method described by A.O.A.C, (2005) was used for the determination of carotenoid content. Two grams of sample was weighed accurately and placed in a high speed blender. Forty milliliters (40ml) of acetone, 60ml of hexane and 0.1g MgCO₃ were added and blended for 5 min. The solution was allowed to settle and decanted into a separator. The residue obtained was washed with 2 x 25ml portions of acetone, then with 25ml hexane and the extracts were combined. The combined extracts were washed free of acetone with 5 x 100ml portions of water and the upper layer transferred to a 100ml volumetric flask containing 9ml acetone and then diluted to volume with hexane. Absorbance of solution was determined at 436nm using a calibrated spectrophotometer. The β-carotene concentration was obtained with the formula;

$$C = \frac{A \times 454 \times 2.2}{196 \times L \times W}$$

Where C = carotene concentration (μg/g)

A = absorbance of sample

L = cell length in cm

W = g product/ml final dilution

$$\text{Vitamin A activity in Retinol Equivalent (RE)} = \frac{\text{carotenoid concentration (}\mu\text{g/g)}}{6}$$

Determination of Vitamin D

One gram (1.0g) of the sample was weighed into a 100ml beaker. Twenty milliliter (20ml) of petroleum ether was added and the mixture was thoroughly shaken for 5min to obtain a homogenous solution. The resulting mixture was allowed to stand for 1h with intermittent shaking at 10 min intervals. After that, the mixture was centrifuged for 15min at 2500rpm and 3ml was transferred into triplicate tubes and evaporated to dryness. Two milliliters (2.0ml) of alcohol was added followed by 0.5ml of 0.1% pyrogallol in ethanol, with the addition of 4 drops of 10% aluminum chloride in ethanol and the mixture was heated in a water bath for four minutes and allowed to cool. Then 4.5ml of ethanol was finally added, mixed and absorbance was read at 470nm. A blank sample was prepared and its absorbance read at

the same wavelength. Standard vitamin D was used to obtain the calibration curve.

$$\text{Vitamin D } (\mu\text{g}/100\text{g}) = \frac{\text{Absorbance of sample} \times \text{gradient factor} \times \text{dilution factor}}{\text{Weight of sample}}$$

Determination of Vitamin E

One gram (1g) of sample was weighed into a 250ml conical flask fitted with a reflux condenser. 10ml of absolute alcohol and 20ml of 1M alcoholic sulphuric acid were added. The condenser and flask were wrapped in Aluminum foil and refluxed for 45min and cooled for 15min. Fifty milliliters (50ml) of distilled water was added to the mixture and transferred to a 250ml separating funnel covered with aluminum foil. The unsaponifiable matters in the mixture were extracted with 5 x 30ml dimethylether. The combined extracts were washed free of acid and dry evaporated at a low temperature and the residues obtained were immediately dissolved in 10ml absolute alcohol. Aliquots of solutions of the sample and standards (0.3-3.0mg vitamin E) were transferred to a 20ml volumetric flask, 5ml absolute alcohol added, followed by a careful addition of 1.0ml concentrated HNO₃. The flasks were placed on a water bath at 90°C for exactly 3min from the time the alcohol begins to boil. It was rapidly cooled under running water and adjusted to volume with absolute alcohol. The absorbance was determined at 470nm against a blank containing 5ml absolute alcohol and 1ml concentrated HNO₃ treated in a similar manner.

$$\text{Vitamin E } (\mu\text{g}/100\text{g}) = \frac{\text{Absorbance of sample} \times \text{gradient factor} \times \text{dilution factor}}{\text{Weight of sample}}$$

Determination of Vitamin K

Vitamin K (2-methyl-1, 4-napthoquinone), known as menadione was determined as follows. Five grams (5g) of sample were weighed into a 250ml beaker and 30ml of butyl alcohol was added. The mixture was thoroughly shaken to obtain a homogenous solution. The resulting mixture was filtered through a whatman No. 42 filter paper into a 100ml volumetric flask and made up to mark with butyl alcohol. 10ml aliquot of the filtrate was pipette into a 30ml centrifuge tube and 3 drops of 2, 4-dinitrophenyl hydrazine was added to develop the blue color which will subsequently change to bluish green upon addition of 3ml of alcoholic ammonia. Standard solutions of vitamin K (0-20 µg /ml) were prepared and treated as sample to obtain a gradient factor. The absorbance of standards and sample were read on a spectronic 21D spectrophotometer at a wave length of 480nm.

Vitamin K in µg/100g is calculated using the formula;

$$\text{Vitamin K } (\mu\text{g}/100\text{g}) = \frac{\text{Absorbance of sample} \times \text{gradient factor} \times \text{dilution factor}}{\text{Weight of sample}}$$

Determination of Percentage Recommended dietary Allowance (RDA) of the Vitamins

The percentage RDA supplied by each of the vitamins was calculated as follows;

$$\% \text{RDA} = \frac{\text{Amount of vitamin in the sample}}{\text{Recommended dietary Allowance of the same vitamin}} \times \frac{100}{1}$$

Determination of phytochemicals

Alkaloid Determination

This was done by the alkaline precipitation gravimetric method described by Harborne, (1998). 2g of the sample was dispersed in 10% acetic acid solution in ethanol to form a ratio of 1:10(10%). The mixture was allowed to stand for 4h at 28°C. It was later filtered through whatman No.42 filter paper, the filtrate was concentrated to one quarter of its original volume by evaporation and treated with drop wise addition of concentrated aqueous NH₄OH until the alkaloid was precipitated. The alkaloid precipitated was received in a weighed filter paper, washed with 1% ammonia solution dried in the oven at 80°C. Alkaloid content was calculated and expressed as a percentage of the weight of the sample analyzed.

Tannins Determination

The method of Swain (1979) was used for the determination of tannin content of the *Coula edulis* flour. 0.2g of the sample was measured into a 50ml beaker. 20ml of 50% methanol was added and covered with paraffin and placed in a water bath at 77-80°C for 1h, and stirred with a glass rod to prevent lumping. The extract was quantitatively filtered using a double layered whatman No.1 filter paper into a 100ml volumetric flask using 50% methanol to rinse. This was made up to the mark with distilled water and thoroughly mixed. 1ml of sample extract was pipetted into 50ml volumetric flask, 20ml distilled water, 2.5ml folin Denis reagent and 10ml of 17%Na₂CO₃ were added and mixed properly. The mixture was made up to the mark with distilled water, mixed well and allowed to stand for 20min when bluish-green coloration developed. Standard tannic acid solutions of range 0-10ppm were treated similarly as 1ml of sample above. The absorbance of the tannic acid standard solutions as well as samples was read after color development on a spectronic 21D spectrophotometer at a wave length of 760nm.

Percentage Tannin was calculated using the formula;

$$\text{Tannin } (\%) = \frac{\text{Absorbance of sample} \times \text{average gradient} \times \text{dilution factor}}{\text{Weight of sample} \times 10,000}$$

Saponin Determination

The spectrophotometric method of Brunner (1984) was used for saponin analysis. One gramme (1.0g) of finely ground sample was weighed into a 250ml beaker and 100ml isobutyl alcohol was added. The mixture was shaken on a UDY shaker for 5h to ensure uniform mixing. Thereafter, the mixture was filtered through a whatman No.1 filter paper into 100ml beaker and 20ml of 40% saturated solution of magnesium carbonate added. The mixture obtained with saturated MgCO₃ was again filtered through a whatman No.1 filter paper to obtain a clear colorless solution. One milliliter (1.0ml) of the colorless solution was pipette into 50ml volumetric flask and 2ml of 5%FeCl₃ solution was added and made up to mark with distilled water. It was allowed to stand for 30min for blood red color to develop. 0-10ppm standard saponin solution were prepared from saponin stock solutions were treated similarly with 2ml of 5%FeCl₃ solution as done for 1.0ml sample above. The absorbance of the sample as well as the standard solutions were read after color development on a spectronic 21D spectrophotometer at a wavelength of 380nm.

Percentage Saponin was calculated using the formula;

$$\text{Saponin (\%)} = \frac{\text{Absorbance of sample} \times \text{average gradient} \times \text{dilution factor}}{\text{Weight of sample} \times 10,000}$$

Phytate Determination

This was determined according to the method described by Oberlease *et al.*, (1973). The sample was first extracted with 0.2N HCl, Then, 0.5ml of the extract solution was pipetted into a test tube fitted with a ground glass stopper. One milliliter (1.0ml) of ferric solution was added and the tube heated in a boiling water bath for 30min ensuring that the first 5min, that the tube remains well stoppered, cooled in ice water for 15min and allowed to adjust to room temperature. The content of the tube was mixed and centrifuged for 30min at 300rpm. One milliliter (1.0ml) of the supernatant was transferred to another test tube and 1.5ml of 2, 2 bipyridine solution was added into the test tube. The absorbance was measured at 519nm against distilled water. The method was used for reference solution as a substitute for the sample solution. The preparation of the calibration curve was carried out by plotting the concentrations of the reference solution against their corresponding absorbance. The absorbance of the test sample was used to obtain the concentration from the calibration curve.

Determination of Amino Acid Profile

The amino acid profile in the known sample was determined using method describe by Benitez (1992). The sample was dried to a constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Technicon Sequential Multi – sample amino acid analyzer (TSM); Model DNA 0209. These procedures are described as follows:

Defatting of sample

The sample was defatted using chloroform/methanol mixture of ratio 2:1. Four grams (4g) of the sample was put in extraction thimble and extracted for 15h in soxhlet extraction apparatus as described by A.O.A.C., (2000).

Nitrogen determination

A small amount (200mg) of the ground sample was weighed, wrapped in whatman filter paper (No.1) and put in the kjeldahl digestion flask. Concentrated sulphuric acid (10ml) was added. Catalyst mixture (0.5g) containing Sodium sulphate (Na₂SO₄), Copper sulphate (CuSO₄) and Selenium oxide (SeO₂) in the ratio of 10.5:1 was added into the flask to facilitate digestion. Four pieces of anti – bumping granules were added. The flask was then put in kjeldhal digestion apparatus for 3h until the liquid turned light green. The digested sample was cooled and diluted with distilled water to 100ml in standard volumetric flask. Aliquot (10ml) of the diluted solution with 10ml of 45% sodium hydroxide was put into the Markham distillation apparatus and distilled into 10ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until about 70ml of distillate was collected. The distillate was then titrated with standardize 0.01N hydrochloric acid to grey colour.

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

Where: a = Titer value of the digested sample
b = Titer value of blank sample

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V= volume after dilution (100ml)
W= Weight of dried sample (mg)
C= Aliquot of the sample used (10ml)
14= Nitrogen constant in mg

Hydrolysis of the sample

Fifty milligram (50mg) of the defatted sample was weighed into glass ampoule. Seven milliliters (7.0ml) of 6N HCl was added and oxygen was expelled by passing nitrogen into the ampoule (this is to avoid possible oxidation of some amino acids during hydrolysis e.g. methionine and cystine). The glass ampoule was then sealed with bunsen burner flame and put in an oven preset at 105°C±5⁰C for 22h. The ampoule was allowed to cool before being broken opened at the tip and the content was filtered to remove the unwanted materials. It should be noted that tryptophan is destroyed by 6N HCl during hydrolysis. The filtrate was then evaporated to dryness at 40°C under vacuum in a rotary evaporator. The residue was dissolved with 5ml to acetate buffer (pH 2.0) and stored in plastic specimen bottles, which were kept in the freezer.

Loading of the Hydrolysate into TSM Analyzer

The amount loaded was between 5 to 10 micro liters. This was dispensed into the cartridge of the analyzer. The TSM analyzer is designed to separate and analyze free acidic, neutral and basic amino acids of the hydrolysate. The period of an analysis lasted for 76 min. The amino acids present in the samples were identified by matching their peak retention time in the chromatogram with those of the peaks of standard mixture of amino acids with norleucine as internal standard.

Method of calculating amino acid values from the chromatogram peaks

The net height of each peak produced by the chart recorder of TSM (each representing an amino acid) was measured. The half height of the peak on the chart was found and width of the peak on the half height was accurately measured and recorded. Approximately area of each peak was then obtained by multiplying the height with the width at half-height. The norleucine equivalent (NE) for each amino acid in the standard mixture was calculated using the formula:

$$NE = \frac{\text{Area of Norleucine peak}}{\text{Area of each amino acid}}$$

A constant S was calculated for each amino acid in the standard mixture.

$$\text{Where } S_{\text{std}} = NE_{\text{std}} \times \text{molecular weight} \times \mu \text{ MAA}_{\text{std}}$$

Finally, the amount of each amino acid present in the sample was calculated in g/16gN or g/100g protein using the following formula:

$$\text{Concentration (g/100g protein)} = \frac{NH \times W \times NH / 2 \times S_{\text{std}} \times C}{\text{Dilution} \times 16 \div NH \times W \text{ (nleu)}}$$

Where C = Sample wt (g) x N% x 10 x vol. loaded
Where NH = Net Height
W = Width @ half height
Nle = Norleucine.

Results and Discussion

Physical Properties of Coula Edulis

The results of the physical properties of *Coula edulis* (weight, length, diameter, thickness) are shown on Table 1. The average weight of unshelled nut was 17.12g, while the shelled nut (the meat) has a mean value of 4.23g. This means that the percent edible portion is 25% while the percent shell 75%. The high shell

content suggests that it can be commercially utilized for other non-food by-products. The unshelled nuts have mean values of 3.64cm and 3.05cm for length and diameter respectively, while the shelled nut have mean values of 2.23cm and 1.92cm for length and diameter. The average shell thickness was 0.43cm. The number of shelled nuts required to produce a 100g is 26 shelled nuts.

Table 1: Physical properties of *Coula edulis*

Physical Property	Mean value + Standard Deviation
Weight of Unshelled nut (g)	17.21±2.60
Weight of shelled nut (g)	4.23±0.56
% edible portion	25
% shell	75
Length of unshelled nut (cm)	3.64±0.13
Length of Shelled nut (cm)	2.23±0.08
Diameter of unshelled nut (cm)	3.05±0.18
Diameter of shelled nut (cm)	0.43±0.09
Shell thickness (cm)	0.43±0.09
Number of nuts per 100g	26±0.63

Axial dimension (length, diameter, thickness) of nuts are useful in sizing, sorting and other separation process design. It is of paramount importance in determining the aperture size of the shelling machine screen. The seeds axial dimensions are required in the design of equipment for removal of nut shell. It is also important in the design or fabrication of suitable screw press oil extraction machine (Salawu *et al.*, 2013). The knowledge of the physical properties of agricultural products is usually very essential for the design of suitable machine and equipment for the production, handling, processing and storage of these products (Idowu *et al.*, 2012). The number of nuts per 100g is a good index for determining the yield of the *Coula edulis*. From the result, 1kg of the edible portion can be obtained from 260 seeds.



Plate 2: Shelled *Coula edulis* nut.



Plate 1: Unshelled *Coula edulis* nuts

Proximate composition of Coula edulis

The proximate composition of *Coula edulis* is shown on Table 2. The moisture content of the nut was 14.61%. The moisture content of tree nuts required for shelf stability ranges from (2 – 20%) and at an optimum relative humidity of 55 –70%. Packaging in moisture – proof containers is recommended, and the lower the temperature of storage (0 -10⁰C), the longer the storage life (Kader *et al.*, 2014). Thus, the low moisture content of the seed is advantageous in improving shelf life.

Table 2: Proximate composition of *Coula edulis*

Parameters	% Composition
Moisture content	14.61±0.029
Ash content	6.09±0.065
Crude protein	4.60±0.025
Crude fat	24.31±0.014
Total dietary fiber (TDF)	4.72±0.082
Soluble dietary fiber (SDF)	1.04±0.016
Insoluble dietary fiber (IDF)	3.68±0.025
Carbohydrate	45.67±0.079
Total energy value (kcal)	419.87±0.392

The ash content of the *Coula edulis* sample was 6.09% and is higher than the values reported for cashew nut (2.4%) by Nandi (1998) and that of chest nut (2.2%) by (<http://foodscience/nuts>). Since the ash content of a sample is a reflection of the amount of minerals it contains, *Coula edulis* is therefore very rich in mineral and can contribute meaningfully to the micro-nutrient needs of children as well as adults.

The result showed that *Coula edulis* contains 4.60% protein. This value is lower than the values for walnut (15.23%) and hazel nut (14.95%) reported by USDA, (2010). However, the protein value compares favorably with and even higher than the value (3.8%) reported for lychee nut (<http://foodscience/nuts>). The nutritional composition of foods generally differ significantly depending upon variety and geographical origin (Ajah and Madubuike, 1997), storage and method of processing/sample preparation (Aremu, 1993). Thus *Coula edulis* can contribute significantly to the daily protein requirement (10-30g) of individuals stipulated by USDA, (2010).

The fat content of *Coula edulis* obtained was 24.31%. The value obtained is lower than the values reported for roasted groundnut (44%), coconut (32%) and cashew nut (48%) but much higher than that of some cereals like rice; brown (2%), sorghum (17.7%) and oat (9%) as reported by FAO, (1998). Fats are essential in diets for energy and they increase the palatability of foods by absorbing and retaining their flavours. Fats are also vital in the structural and biological functioning of the cells and help in the transport of nutritionally essential fat – soluble vitamins (Aiyesanmi and Oguntokun, 1996).

The percentage mean value of the soluble fiber in *Coula edulis* is 1.04% while the insoluble dietary fiber is 3.68%. The mean total dietary fiber content of *Coula edulis* was 4.72% which is lower than the values reported for macadamia nut (8.68g/100g), hazel nut (9.78g/100g) and walnut 96.78g/100g) by USDA, (2010), but higher than the values reported from most other nuts (1.64 - 4.3%) (<http://foodsciencewikispaces.com/nuts>). An adequate

intake of fiber is important to facilitate and regulate intestinal transit, thus preventing constipation (Plessi *et al.*, 1999). The soluble fiber helps lower blood sugar and blood cholesterol by dissolving in water and forming a gummy substance that bind cholesterol and carbohydrates in the intestine (NHWC, 2002).

The amount of carbohydrate obtained by difference (45.67%) was higher than the value obtained for macadamia nut (13.8%) by USDA (2010), roasted groundnut (24%), coconut (15%) and cashew nut (19%) by Enwere (1998). The high carbohydrate content of *Coula edulis* makes it a good quality food. Carbohydrates play a significant role providing energy for metabolism for living organisms (Onyeka, 2008).

Coula edulis nuts are very high in energy. Hundred grams (100g) of the nut provides 419.87kcal of energy and thus supplies a good percentage of the recommended dietary allowance (1300kcal-3000kcal) of different age groups (FNB, 2004).

Mineral Profile of Coula edulis

The mineral composition of *Coula edulis* is as shown on Table 3. The results show that sodium has the highest concentration 512.1mg/100g while copper recorded the lowest concentration with value 0.6983mg/100g. The next abundant mineral element is potassium which was 409.7mg/100g, followed by calcium and magnesium having the same value of 130.4mg/100g. Manganese averaged 1.4432mg/100g, a value adequate for the supply of the recommended daily intake of children one to three years of age(1.2mg) but provides a percentage of the RDA for other age groups from 4 – 70+ years (1.5 -2.6mg) as shown by NHWC, (2002). Female sex hormones and thyroid hormones are dependent on manganese to work properly. Copper supplied 78% of the RDA/100g, zinc supplied 13% RDA/100g. *Coula edulis* is also rich in iron (6.6108 mg/100g) supplying 83% of the RDA. This blood building element is very essential in human and animal consumption. Mineral elements aid biochemical functions necessary for growth, development and overall health and they help in enzyme function (NHWC, 2002).

Table 3:Mineral profile of *Coula edulis*

Mineral	Composition mg/100g	RDA (mg)	%RDA(USDA (2010)
Calcium	130.4	1000	13
Magnesium	130.4	400	33
Potassium	409.7	4700	9
Sodium	512.1	1500	34
Manganese	1.4432	2.3	63
Copper	0.6983	0.9	78
Zinc	1.3967	11	13
Iron	6.6108	8	83

RDA: Recommended dietary allowance

Vitamin profile of Coula edulis

The vitamin content of the *Coula edulis* are shown on Table 4. Vitamin E had the highest concentration (8.46mg/100g) among the fat soluble vitamins and supplied 56% of the recommended dietary allowance (RDA) in 100g of sample, while vitamin K had the lowest concentration of 0.003184mg/100g and supplied 3% of RDA per 100g of sample. Vitamin A (retinol equivalent)

concentration averaged 0.054mg/100g and provides 6% RDA while vitamin D supplies 67% RDA with concentration 0.01mg/100g. Vitamin D is very essential for its help in calcium absorption and also in its critical role in regulating how much calcium stays in the blood (NHWC, 2002). Vitamin E has a beneficial effect on fertility, aids in the absorption of iron and proper maintenance of cell membrane.

Table 4: Vitamin Profile of *Coula edulis*

Vitamin	Composition (mg/100g)	RDA (mg)	%RDA USDA (2010)
Vitamin A (RE)	0.054	0.9	6
Cholecalciferol (vit D)	0.01	0.015	67
Tocopherol (vit E)	8.460	15	56
Phylloquinone (vit K)	0.003184	0.12	3
Ascorbic acid (vit C)	34.9926	90	39
Thiamine (vit B1)	0.3706	1.2	31
Riboflavin (vit B2)	0.4358	1.3	34
Niacin (vit B3)	0.7756	16	5
Pantothenic acid (vit B5)	0.1229	5	3
Pyridoxine (vit B6)	1.3278	1.3	102
Folic acid (vit B9)	0.2859	0.4	72
Cyanocobalamin(vit B12)	0.0005819	0.0024	24

RE: Retinol equivalent

Vitamin C has the highest concentration 34.99mg/100g among the water-soluble vitamins. A 100g of *Coula edulis* would supply 39% of the RDA for vitamin C. Vitamin C acts as a strong antioxidant, protecting other vitamins from harmful oxidation, prevents cell damage and may also stop the production of cancer-causing nitrosamines in the stomach (NHWC, 2002). The next abundant of the water-soluble vitamins is vitamin B6 (0.4358mg/100g), vitamin B1 (0.3706mg/100g), vitamin B9 (0.2859mg/100g) and vitamin B12 having the lowest concentration (0.0005819mg/100g). Vitamin B6 provides more than the recommended dietary allowance having 102% of RDA/100g and is the highest supplied water-soluble vitamins, followed by vitamin B9 (72%RDA/100g), while pantothenic acid (vitamin B5) supplied the least percentage of the RDA/100g (3%). Vitamin B6 (pyridoxine) helps the body to maintain normal homo-cystein levels that can lower the risk of heart disease and also helps in protein metabolism (NHWC, 2002). Generally, the water soluble vitamins are non-toxic when consumed in excess

because their solubility in water and water-like fluids which facilitates their excretion through urine and sweat. On the other hand, their inability to be stored accounts for their relatively fast development of deficiency symptoms (Okaka *et al.*, 2002). Therefore, regular intake of these nutrients is essential for a health.

Phytochemicals in *Coula edulis*

The results of some phytochemical in *Coula edulis* are shown on Table 5. Phytochemicals are chemical compounds that occur naturally in plants. They are substances that may have biological significance for example carotenoids or flavonoids, but are not established as essential nutrients and they work to halt the factors that are adverse to health in humans such as cancer, high cholesterol levels and aging (NHWC, 2002). The tannin content averaged 0.72%, while saponin was 0.18%. Alkaloid and phytate contents were 1.07% and 0.28% respectively.

Table 5: Phytochemicals in *Coula edulis*

Phytochemical	% composition
Tannin	0.72±0.071
Saponin	0.18±0.051
Alkaloid	1.07±0.041
Phytate	0.28±0.043

Data are means ± SD of triplicate determinations

Tannins are plant polyphenols which have ability to form complexes with metal ions and with macro-molecules such as proteins and polysaccharides and adversely affects protein digestibility (De-Bruyne *et al.*, 1999). Though tannins have traditionally been considered anti-nutritional but it is now known that their beneficial or anti-nutritional properties depend upon their chemical structure and dosage (Muller-Harvey and McAllan, 1992). For example, earlier researchers have shown that products containing chestnuts tannins at low dosages (0.15-0.2%) in the diet are beneficial (Schiavone *et al.*, 2008).

Saponin binds with bile salts and cholesterol in the digestive tract, which prevents cholesterol from being reabsorbed into the body, have antioxidant properties by reacting with cholesterol rich membranes of cancer cells, preventing the cells from growing. Studies have shown that saponins inhibits tumour cell growth and reduced tumour cell activity in a dose dependent manner. The higher the concentration of saponins, the lower the

tumour cell growth and activity (Sung *et al.*, 1995). Thus saponin helps to protect the human body against cancers, and also lower cholesterol levels, decrease blood lipids, lower cancer risks, and lower blood glucose response (Shi *et al.*, 2004).

Phytates are antioxidant compounds found in whole grains, legumes, nuts and seeds. Phytate has a strong binding affinity to minerals such as calcium, magnesium, iron, copper and zinc which results in precipitation, making the minerals unavailable for absorption in the intestines and is common in the hulls of nuts, seeds and grains (Munir and Joseph, 2009). Though phytate acts as an anti-nutrient with regards to iron by its ability to sequester and trap iron, it exhibits anti-cancer properties, reduce cholesterol and triglycerides and positively impacts the glycemic response of certain foods (Graf *et al.*, 1990). A phytic acid intake of 4.00-9.00mg/100g reduces iron absorption by 4-5 folds in humans (Hurrell *et al.*, 1992). Alkaloid is a nitrogenous basic compounds found in plants, typically insoluble in water and

physiologically active such as morphine, strychnine, quinine, nicotine and caffeine (Collins discovery Encyclopedia, 2005). The lower limit of nicotine causing fatal outcomes in man is 500-1000mg (6.5-13mg/kg) while that of caffeine LD₅₀ is 127mg/kg for mice (Mayer, 2014). Alkaloids are very useful in prolonging the action of several hormones and acting as stimulant as reported by Okwu, (2004).

Osagie (1998) reported that simple boiling, cooking and soaking can reduce the concentration of anti-nutrients in food stuffs. However many of the anti-nutrients in foods are under study for their potential health benefits (Dinkova and Kostov, 2012).

Amino Acid Profile of *Coula edulis*

Protein quality is an attribute of protein that speaks of its amino acid composition, digestion and utilization. The efficiency with which a protein is used for growth or maintenance is a measure of its quality (Onyeka, 2008). The essential amino acid profile is shown on Tables 6 while the non-essential amino acid profile is shown on Table 7. The highest scoring essential amino acid is leucine (7.10g/100gprotein) which supplied 101% of the recommended dietary allowance (RDA), while the limiting amino acid is methionine (1.5g/100gprotein) and had 43% amino acid chemical score. The second most abundant essential amino acid is valine (3.56g/100gprotein) followed by lysine (3.54g/100gprotein), phenylalanine (3.52g/100gprotein), isoleucine (3.0g/100gprotein), threonine (2.27g/100gprotein) and the least methionine (1.50g/100gprotein). Isoleucine has a chemical score of 75%, lysine 64%, phenylalanine 59%, threonine 57% and valine 72%.

This implies that the consumption of 100g of edible portion of the nut (about 26 nuts) a day will provide a high percentage of the essential amino acids for a healthy individual. There is however the need for supplementation with other cereal food products especially in those areas where protein energy malnutrition (PEM) has continued to pose challenges. Chronic malnutrition mostly occurs due to poor feeding practices and low quality protein commonly associated with plant –based single diets (Badamosi *et al*; 1995). However, combination of plant foods from different botanical origin will ensure adequate supply of the essential amino acid since they tend to complement each other.

The nonessential amino acid value (Table 7) is highest for glutamic acid (12.86g/100gprotein) and lowest for cysteine (1.24g/100gprotein). The next abundant of the non essential amino acids is aspartic acid (9.66g/100gprotein). Arginine averaged (6.55g/100gprotein) which is an amino acid, amongst other functions is the source from which nitric oxide is derived; this oxide acts as an endothelial vasodilator and has an antiplatelet action. The arginine content of nuts is from 2g-3g/100g of food (Brown and Hu, 2000). Alanine is (3.56g/100g), serine (3.55g/100g), glycine (3.43g/100g), proline (3.02g/100g), tyrosine (2.81g/100g) and histidine (2.29g/100g).

The nutritional value of protein is a combination of two factors; total essential amino acid content of the protein and protein digestibility which is an indicator of the availability of the essential and non-essential amino acids in the protein. Also the amino acids composition is an important aspect in defining the characteristics of the protein (www.fnrc.nal.usda.gov/dietary-guidance).

Table 6: Essential Amino acid Profile of *Coula edulis*

Amino acid	Composition (%/100g prtein)	Amino acid content of Reference protein FAO/WHO (1991)	Chemical score
Isoleucine	3.00	4.00	75
Leucine	7.10	7.00	101
Lysine	3.54	5.50	64
Methionine	1.50	3.50	43
Phenylalanine	3.52	6.00	59
Threonine	2.27	4.00	57
Valine	3.56	5.00	71
Tryptophan	Not determined	1.00	

Data are means of triplicate determinations

Table 7: Non essential amino acid profile of *Coula edulis*

Amino acid	Composition g/100gprotein
Histidine	2.29±0.008
Arginine	6.55±0.016
Aspartic acid	9.66±0.016
Serine	3.55±0.008
Glutamic acid	12.86±0.016
Proline	3.02±0.008
Glycine	3.43±0.022
Alanine	3.56±0.008
Cystine	1.24±0.016
Tyrosine	2.81±0.022

Data are means ± SD of triplicate determinations

Conclusion

The result of the physical analysis of the *Coula edulis* has revealed its dimensions which is importance in the fabrication of suitable machines for commercial and industrial processing of the nut. It also helps in estimating the yield which is of paramount importance for its optimal utilization in the food industry for maximization of profit. This work has revealed the nutritional composition of the nut which has provided information for nutritional guidance and counseling for the health sector as well as its potentials for use for both human consumption and raw material for the commercial food industry. *Coula edulis* is a good source of carbohydrate (45.67%), fat (24.31%), dietary fibre (4.72%) and protein (4.60%). The chemical score of some of the essential amino acids; isoleucine (75%), leucine (101%), lysine (64%) and valine (71%) supplied more than 50% of the recommended dietary allowance. The limiting amino acid was methionine with 43% amino acid score. The micronutrients, Ca, Mg, K, Na, Mn, Cu, Zn, Fe and vitamin profile such as cholecalciferol (67%RDA/100g), pyridoxine (102%RDA/100g), folic acid (72%RDA/100g) and tocopherol (56%RDA/100g) detected in the flour sample revealed the nutritional benefits of the nuts for human consumption. The results of the phytochemical composition of the flour sample shows that it contains tannin, saponin, alkaloid and phytate. And each of these are known for various protective effects, antioxidant properties and also exhibit antimicrobial properties

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