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Evaluation of the Antinutrients, Amino Acid Profile and Physicochemical Properties of *Hura crepitan* Seed

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Authors' contributions

This work was carried out in collaboration among all authors. Authors JNN and NCO conceptualized and designed the study. Author IAO wrote the protocols and initial draft of the manuscript. Author AFO performed the statistical analysis and interpretation of the data. Author EN did the literature searches, involved in collection of data and managed the analyses of the study. All authors read and approved the final manuscript.

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ABSTRACT

The Objectives of the Study: To evaluate the Antinutritional components, Amino acid profiles and the physic-chemical properties of *Hura crepitan* (Sand box) seed.
Design of the Study: This study was structured to fit into using a combination of T-test and one way Analysis of Variance (ANOVA) to evaluate the data obtained from the laboratory analysis.
Place and Duration of Study: This research work was done at the Department of Food Science and Technology Laboratory, Federal University of Technology, Owerri, Nigeria, between May 2019 and November 2019.
Methods: The mature dry fruits of the *Hura crepitan* seeds were harvested from the plants on Federal University of Technology Owerri, Imo State campus.

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The *Hura crepitan* seeds were sorted in order to remove dirts and foreign other foreign contaminants. The cleaned seeds were divided into four portions and stored in separate glass containers for further processing. The first portion of the seeds were dried at 60°C in Gallen Kamp moisture extraction oven for 6 hours at and pulverised in a Monilex blended into flour, some seeds were boiled while some portions were roasted and processed into fine flour and subsequently subjected to analysis to evaluate the antinutritional contents, amino acid s as well as determining the physic-chemical properties of the samples.

Results: The results obtained suggested that the anti-nutrients in the raw seed-flour were flavonoids with 17.50%, alkaloid (6.20%), tannin (5.24%), and cyanogenic glycoside (1.76%). Fermentation and moist cooking were found to be more effective in the reduction of the anti-nutrients in the *Hura crepitan* seeds. The amino acid profiles were evaluated, and twenty amino acids were identified in the seed flour. The three major ones implicated were arginine (3.25 g/100 g in cooked and 8.05 g/100 g in fermented), glutamic acid (6.05 g/100 g in cooked and 10.2 g/100 g in fermented) and valine (8.03 g/100 g in raw and 8.58 g/100 g in fermented). The limiting amino acid is methionine with a chemical score of 44.52%. the physicochemical properties of the sandbox seeds evaluated suggested that the free fatty acids values ranged from 3.60% to 6.03% and there were no significant differences (P>0.05) among the samples, the iodine value ranged from 104.94% to 126.90%, the peroxide value for the sample varies between 2.96% to 44.81%.

Conclusion: This study suggested that the *Hura crepitan* seed contains appreciable amounts of essential amino acids as well as having good physicochemical properties while the use of moist heat and/or fermentation can reduce the antinutritional components to the bearest minimum. Hence, can be utilized some areas of food industries where protein (amino acids) are critically required.

Keywords: Sandbox seed; anti-nutrients; amino acids; physicochemical properties; cooking; fermentation.

1. INTRODUCTION

The sandbox tree (*Hura crepitan*), also known as 'possum wood' and 'jabillo' in Spanish is an evergreen tree of the spurge (Euphorbiaceae) family. It is native to tropical regions of North and South America in Amazon Rainforest [1]. It is recognized by the many dark, pointed spines on its brown bark. The *Hura crepitan* tree (plate 1.) is reported to grow up to 30m (100ft) high in the Amazon region of South America with large ovate leaves which are papery thin [2]. This tree was introduced by travelers to Nigeria, though they do not grow up to 30m in height and their leaves are not as wide as observed in the Amazon region. This may be because of climatic conditions and type of soil. This tree grows up to 10m high in Nigeria. They are planted as shade trees along the walkways in church compounds, parks, cities and schools. The fruits are tyre shaped capsules (plate 3), 3 cm to 4 cm in thickness with a diameter of 5 cm to 6 cm, with sixteen carpels arranged radially around the central axis [2]. When ripe and dry, capsules explode to disperse some seeds up to 14 metres away. Hence, the name 'Dynamite' tree, because of the explosive sound of the ripe dry fruit as it splits into segments. The seeds are circular and flattened about 2 cm in diameter with smooth brown colour. This tree offers a wide range of uses, for instance, the plant secretes yellowish milky latex used by Amerindians to produce poisoned darts [3,4]. The *Hura crepitan* has been found to contain certain nutrients ranging from amino acids (protein), ash (minerals), lipids in the form of oil, etc; thus, the seed has also been said to contain some antinutrients in varying degrees such as phytates, alkaloid, saponins, flavonoids, etc. [1]. However, alongside the nutritional contents, it also offers other economic benefits including ornamental purposes. The *Hura crepitan* which contains various nutyrients as well as phytochemicals may also provide essential oils and pigments that could be used in foods/animal feeds industries, chemical industries, pharmaceutical industries and cosmetic industries. In Nigeria, presently, the *Hura crepitan* tree is seen as an ornamental tree used to provide shade along walkways and within large premises such is its present relevance within the campus of the Federal University of Technology, Owerri (FUTO).

Plate 1. Friuts of *Hura crepitan* Tree

Plate 2. Seed of *Hura crepitan*

Plate 3. Matured dried friut of *Hura crepitan*

The use of wild plants and seeds in different localities provide optimum source of nutrients especially in times of food scarcity [5]. Yet there is only scanty information on the nutritional and anti-nutritional qualities of some of these wild plants and seeds. Studies on such plants and seeds may help in identifying the values and potentials of the plant or seed. Also without investigation into the anti-nutritional profiles of the seed or plant, the use of such materials in pharmaceutical or food preparations could have prolonged side effects that may occasionally be fatal. *Hura crepitan* seed is one of such seed that need to be studied. The objective of this work is therefore to evaluate the antinutritional, amino-acid profile, and phytochemical analysis of sand box (*Hura crepitan*) seeds.

It is envisaged that a good nutrient-balance, in the seed flour with regards to protein and amino acids will reveal the potential of the seed for inclusion in human and animal diet. The study is aimed at carrying out a research on *Hura crepitan* seed with the hope of acquiring information on the seed and consequently its potential uses. Therefore the study will investigate the aninutritional composition, amino acid profile and phytochemical analysis of the *Hura crepitan* seed flour. Thus, it is envisaged that this study when completed may expose another oil seed with a good potential for food and industrial uses. Also Raw Materials Research and Development Council (RMRDC) have been fostering researches which will lead to development of our raw materials, oil seeds inclusive.

2. MATERIALS AND METHODS

2.1 Collection of Raw Material

The mature dry intact (indehiscent) fruits of the sand box (*Hura crepitan*) seeds were obtained from some sand box plants around the Federal

University of Technology Owerri (FUTO) campus. These hard fruits were broken with to obtain the brown coloured seeds contained in hollow chambers of the pods, with each chamber housing a seed and up to fourteen seeds from a single fruit. Wholesome seeds released from dehisced fruits, were also picked-up from the ground around the trees. Both the harvested and picked seeds were combined to form the raw materials used for this research work. The brown seeds were dehulled with hammer to obtain the milky coloured cotyledons.

2.2 Processing of the *Hura Crepitan* Seeds

The seeds from *Hura crepitan* treewere sorted to remove dirts and foreign materials. The cleaned seeds were divided into four portions and put in separate glass containers with loose covers for further processing. The seeds in the first container (E1) were dried in Gallen Kamp moisture extraction oven for 6 hours at 60°C and pulverised in a Monilex blender (model, chemistry and Norris Ltd Chemfold, England) into flour. The seeds in the second container (E2) were boiled for 2 hours. After cooking, the boiled seeds were put in a tray, and then dried in a Gallen Kamp oven for 24 hours at 65°C. The dried seeds were also pulverised in a Monilex blender into a fine flour. The seeds in the third container (E3) were roasted at 70°C for 50 min. The roasted seeds were also pulverised in a Monilex blender into a fine flour. The seeds in the fourth container (E4) were boiled for two hours until the cotyledons became very soft. The water was discarded and the cooked seeds were wrapped in plantain leaves and put in a basket. They were left to ferment for three days. At the end of three days, the fermented seeds were dried in the Gallen Kamp moisture extraction oven at 60°C for 6 hours and ground into flour using the Monilex blender (model, chemistry and Norris Ltd Chemfold, England). All the samples were stored in well labelled air tight glass containers and kept as stock in the freezer (°C).

2.3 Determination of Anti-nutrient in *Hura crepitan* Seed Flour

2.3.1 Determination of alkaloid content

The method used for this determination was the one adopted by [6]. Five grams of the prepared sample were weighed into a beaker of 250 ml capacity with addition of 200 ml of 20% ethanolic acetic acid and the mixture was allowed to stay for four hours for complete reaction, then following filtration. The filtrate was concentrated in a hot-water bath to one quarter of its initial volume. Concentrated ammonium hydroxide solution was added drop-wise to the extract until precipitation was complete. The whole solution was allowed to settle under gravity and the precipitate was collected by a method of filtration. A filter paper of known weight and contents were dried in a moisture extraction oven set at 70°C for 6 hours. The dried material was then cooled over a desiccator and subsequently weighed.

$$\%\ Alkaloid = \frac{w_3 - w_2}{w_1} \times \frac{100}{1}$$

Where,

W_3 = Weight of filter paper and alkaloids
W_2 = Weight of filter paper
W_1 = Weight of sample

2.3.2 Determination of tannin content

Tannin was determined by the method of [7] using Unico UV-2102 Spectrophotometer. Five hundred milligrams of the sample was weighed into a 100 ml plastic bottle and 50 ml of distilled water was added mixed thoroughly for 1 hour using a mechanical shaker. The sample was subsequently filtered into a 50 ml volumetric flask and made up to mark with distilled water. Then 5 ml of the filtrate was transfered into a tube and mixed with 3 ml of 0.1m FeCl in 0.1N HCl and 0.008M Potassium ferocyanide (KCN) solution. The absorbance of the sample was measured spectrophotometrically at 120nm wavelengths for 10 minutes. A blank sample was prepared and the colour was developed and read at the same wavelength. A standard was prepared using tannin acid to get 100ppm and measured. The total tannin content was calculated using the formular below:

$$mg/cm^3 Tannin = \frac{Absorbance of sample}{Absorbance of Standard} \times \frac{Conc.\ of Standard}{1}$$

2.3.3 Determination of saponin content

The saponin content was determined by the method of [8]. 20 g of the test sample was dispersed in 200 ml of 20% ethanol to form a suspension. The suspension was heated over a hot water bath raised at 55°C for 4 hours with continuous stirring. Then resultant mixture was filtered and the residues were re-extracted with another 200 ml of 20% ethanol by volume and the combined extract was concentrated to 10 ml,

over a hot water bath heated to 90°C. The concentrated mixture was subsequently transferred into a 250 ml separating funnel with the addition of a solution of diethylether. The sample was rigorously shaken and allowed to settle under gravity. The aqueous layer was recovered and the other layer was discarded. The purification process was done by the incorporation of 60 ml of n-butanol into the aqueous phase (layer) and the mixture was shaken, then allowed to separate. The extract was washed twice with 10 ml of 5% aqueous sodium chloride in a separating funnel. The twice washed extract was poured into a pre-weighed beaker and evaporated to dryness on a boiling water bath. The resultant extract was cooled and subsequently reweighed. The percentage saponin was calculated using the following formular:

$$\%\, Saponin = \frac{w_3 - w_2}{w_1} \times \frac{100}{1}$$

Where,

W_3 = Weight empty of beaker + Sample
W_2 = Weight of empty beaker
W_1 = Weight of sample
100 =Scaling factor (in percentage)

2.3.4 Theflavonoid content

The method described by [6] was used for this determination. One hundred grams (100 g) of the sample was extracted with 100 ml of 80% aqueous methanol at room temperature repeatedly. The entire mixture was filtered through Whatman filter paper (125mm). The filtrate was later transferred into a crucible and evaporated to dryness over a hot water bath and weighed.

$$\%\, Flavonoid = \frac{w_3 - w_2}{w_1} \times \frac{100}{1}$$

Where,

W_3 = Weight of beaker and sample
W_2 = Weight of empty beaker
W_1 = Weight of test sample
100 = Scaling factor to convert to percentage

2.3.5 Evaluation of cyanogenic glycoside content

The method use3d for the determination of the Hydrogen cyanide (HCN) was a method adopted by [8]. One gram of the flour sample was placed in a round bottomed flask of 250 ml capacity. A 200 ml of deionized water was added into the flask while it was allowed to stand for 2 h. The entire mixture was subjected to distillation and about 150 ml of the mixture was distilled into a 250 ml Erlenmyer flask containing 20 mls of 2.5% (W/V) Sodium hydroxide (NaOH) solution. About 0.5 g of Tannic acid (anti-foaming agent) was added to the sample mixture. 100 ml of the distillate was subsequently measured into a 250 mls Erlenmeyer flask with the addition of 8.0 mls of 6.0M NH_4OH solution, followed by 2.0 mls of 5.0% Potassium iodide (KI) solution. The contents of the flask was rigorously mixed, and titrated with a solution of 0.02N Silver Nitrate ($AgNO_3$) until the mixture was clear with no evidence of turbidity.

The Cyanogenic glycoside (HCN) content was obtained using the following formular:

$$X = T \times \frac{108}{1} \times \frac{N}{1000} \times \frac{Ve}{Va} \times \frac{100}{1}$$

Where,

X = % HCN
T = Titre value
108 = Equivalent weight of $AgNO_3$
N = Normality of $AgNO_3$ solution

1000 = Scaling factor to obtain silver nitrate content of 1.0 mls of silver nitrate solution

Ve = Extract volume
Va = Aliquot volume titrated
100 = Scaling factor to convert value to percentage

2.3.6 Evaluation of the oxalate content

The method of stipulated by [9] was used for this assay. A Five grams of the test sample was transferred into a 100cm^3 Erlenmeyer flask with the addition of 20cm^3 of 0.3N hydrochloric acid solution. The entire mixturein the flask were stirred using a magnetic stirrer for I hour at 50°C following filtration. This process was repeated two times. The filtrates were combined and the volume was raised to 100cm^3 with deionized water. A 20cm^3 of the filtrate was transferred into a 100cm^3 beaker and 3-5 drops of phenolphthalein indicator was added. Subsequently, 5.0N ammonium hydroxide was added in drops till the reaction mixture was alkaline. Glacial ethanoic acid was added in drop wise till the pink colouration disappeared and few more drops were added subsequently to acidify

the mixture. A 5cm^3 of solution of 5.0% Calcium chloride was added to the mixture while to stand for three hour to effect complete reaction. The mixture was centrifuged at 300 r.p.m for 15 minutes. The residue was washed three times with hot water by centrifugal means. The residue was subsequently dissolved in 5cm^3 of 3.0N Sulphuric acid solution (H_2SO_4) with warming at 70 - 80°C. The resulting mixture was titrated with freshly prepared 0.01N potassium permanganate solution till a permanent pink colouration that lasted for about 30 secs was obtained. A blank titration was performed using the same volume of 3.0N tetraoxosulphate (vi) acid as that used in dissolving the oxalate residue of the sample was carried out.

$$\%\ oxalate = \frac{x}{1} \times \frac{1}{1000} \times \frac{100a}{1} \times \frac{100b}{5}$$

Where,

X = weight of oxalate e obtained by multiplying molar mass and molarity of oxalate
1000 = reference volume for molar concentration
100a = total volume of extract
100b = scaling factor to convert to percentage
5 = weight of sample taken for analysis

2.3.7 Determination phenol content of *Hura crepitan*

The percentage Phenol contents oif the *Hura crepitan* were determined using a mthod described by [10]. Two hundred milligrams of the *Hura crepitan* seed flour was weighed into a 100cm^3 Erlenmeyer flask followed by the addition of 10 ml of methanol solutioin. The content of the flask was gently stirred and continuously for an hour until a homogenous mixture is achieved. However, the mixture was subsequently filtered using whatman filter paper. Three hundred microliters (300µl) of filterate was transfered into a test tube with the addition of 1.50mlk of folin-ciocalteau solution which has been diluted ten times. Also 1.2 ml of 7.5% sodium carbonate solution was added to the filtrate. While allowing the mixture to stand for 30 minutes. The absorbance of the mixture was measured spectrophotometrically using a uv spectrophotometer at 765nm and a blank was also prepared using 300µl of distilled water.

Serial dilutions covering 0-60µg / ml standard solution were prepared. The ent process as done with the sample was repeated. The absorbance of the working standard was measured at the same wavelength and with these a calibration curve was obtained. With the aid of the calibration curve the concentration of phenol in the sample was obtained.

2.3.8 The phytate content of the *Hura crepitan* seed flour

The method of described by [11] was adopted for this determination. Ten grams of the sample was measured and placed in a thimble. The fat content was extracted by soxhlet method using hexane. The defatted sample was dried in an moisture extraction oven at 70°C and thereafter, was transferred into a desiccator to cool down to room temperatur. Furthermore, 2.0 g of the defatted sample was weighed into a 100.0cm^3 Erlenmeyer flask with addition of 50cm^3 of 0.18M trichloroacetic acid solution. The mixture was stirred on a magnetic stirrer for one hour. The reaction mixture was subjected to centrifugation for ten minutes at 300r.p.m. A 10cm^3 of the supernatant was added to 4.0cm^3 of 0.036M iron (III) chloride solution in a boiling water bath for 45 minutes. The resulting mixture was centrifuged and the resulting ferric phythate was collected. The phythate residue was washed twice with 20.0cm^3 of 0.18M trichloroacetic acid solution and also twice with 30.0cm^3 of portable water. Then 5.0cm^3 of 1.5M sodium hydroxide solution was added to the residue. Into the reaction mixture, 25.0cm^3 of water was added and the mixture was heated over a boiling water bath till the coagulation of ferric hydroxide was achieved. The mixture was then centrifuged and the ferric hydroxide was collected after washing with water. The ferric hydroxide that was obtained was dissolved in 40.0cm^3 of 3.2M trioxonitrate (v) acid. With distilled water, the volume of the mixture was raised to 100.0cm^3, and was used in the determination of iron content which was needed to analyse the phytic acid value in the sample.

Furthermore, three clean test tubes were brought and 2.0cm^3 of the phytate solution was pipetted into one test tube, into another test tube, 2.0cm^3 of distilled water was pipetted into the tube and 2.0cm^3 of standard iron solution was measured into the third vessel. One drop of concentrated hydrochloric acid and one drop of thioglycollic acid solution were added into each of the tubes. The contents of each tube were mixed rigorously and allowed to stand for 30 minutes. 0.4cm^3 of 50.0% sodium acetate were added to each mixture and were allowed to stand for 10 minutes at room temperature. The absorbance of the contents of each tube was measured

spectophotometrically using a UV/vis spectrometer set at 540nm.

Thus, the iron content was calculated as:

$$ugironper\ 100.0\ g = \frac{Absorbanceofsample \times ConcentrationofStandard}{AbsorbanceofStandard}$$

However, from the iron value, the phytic acid content was obtained using the formular;

$$Phyticacidin\mu g/100gsample = \frac{660.8}{31} \times \frac{6Z}{4}$$

Where,

Z = iron content in the phytate in μg
660.8 = Molecular weight of femic phytate
31 = Molecular weight of phosphorus
6.4 = Mole ratio phosphorus to iron in phytate

2.4 Determination of Amino Acids of *Hura Crepitan* Seed Flour

The amino acid profiles of the *Hura crepitan* was determined by spectorophotometric method with slight modification described by [12]. The underlying principle is that Ninhydrin combines with amino acids to form coloured complexes, the intensity of whose colour depends on the amount of amino acid present.

One gram of ground *Hura crepitan* seed flour was weighed into a stoppered 250 ml conical flask with addition of 100 ml of 6MHCl. The temperature of the mixture was raised by subjecting the sample to heating in an oven for 16 hours at 50°C for hydrolysis. The mixture obtained was filtered through a double layered Whatman No. 42 Filter paper into another 25 ml conical flask.

Thereafter, a 2 ml of the hydrolysate was transfered into a 30 ml test tube and 10 ml of buffered ninhydrin reagent was added. The mixture was heated in a boiling water bath for 15 minutes and subsequently down to room temperature following the addition of 3 ml of 50% ethanol solution immediately. Next, 0-5$\mu g/ml$ of working standard amino acids was prepared from each standard solution of amin acids and these dilutions (working standards) were heated with the buffered ninhydrin reagent as done with the sample hydrolysate above. The absorbance of hydrolyzate of sample and the working standards were measured at their appropriate wavelengths as stipulated in Table 1 of each amino acid.

Using the absorbance of each working standard solution and their corresponding concentration, the calibration curves were plotted through which the slope was determined, hence, the percentage amino acid.

Thus,

$$\%\ Aminoacid = \frac{Absorbanceofsample \times gradientfactor \times Dilutionfactor}{Weightofsample}$$

Table 1. Wavelength of colour developed by each amino acid

Amino acid	Wavelength	Colour
Asparagine	505nm	Light blue
Alanine	620nm	Blue
Aspartic acid	465nm	Purple
Cysteine	600nm	Blue
Glutamic Acid	560nm	Purple
Glycine	525nm	Purple
Histidine	460nm	Purple
Isoleucine	580nm	Light Purple
Leucine	590nm	Purple
Lysine	450nm	Orange yellow
Methionine	525nm	Greenish yellow
Omithine	570nm	Purple
Phenylanine	545nm	Yellow
Proline	470nm	Yellow
Pyrrolysine	455nm	Yellowish
Serine	485nm	Blue
Threonine	615nm	Bluish green
Thyrosine	530nm	Greenish blue
Typtophan	565nm	Yellowish Blue
Valine	490nm	Greenish Blue

2.5 The Physico-chemical Properties of the *Hura crepitan* Seed Oil

2.5.1 Determination of the percentage of oil in *Hura crepitan* (sand box) seed

The oil contents od the different weighted portions of each of the four flour samples (E_1 - E_4) were extracted using N-hexane solvent in a soxhlet apparatus as described in [6]. Each portion of a sample was separately extracted and the percentage oil was calculated. In each case, about 150 ml of the solvent, N-Hexane was transfered into the boiling round bottomed flask placed on a heating mantle. The sample portion to be extracted was placed inside a thimble and placed into the refluxing chamber of the apparatus. The set up containing the solvent was allowed to boil under reflux for 8 hours while washing off the fats in the flour sample. At the end of the process, the solvent was recovered and the remnanants evaporated using a moisture extraction oven. Thereafter, the percentage of oil extracted from each sample was determined respectively using the formular below:

$$\%\ Oil = \frac{Weight of oil}{Weight of sample} \times \frac{100}{1}$$

2.5.2 Determination of the acid value

The method described by [6] was used for this determination. One gram of each treatment oil sample was weighed into a 100 ml beaker and 50 ml of neutral alcohol was poured into the beaker with addition of 3 drops of phenolphthalein indicator into the sample mixture. The solution was subjected to boiling on a steam water bath. Thereafter, the solution was titrated with 0.2N Potassium hydroxide solution until permanent light pink solution was obtained. The burette reading (N) was recorded and the acid value was then calculated as follows:

$$\frac{11.22N}{W}$$

Where,

N = ml of 0.2N Potassium Hydroxide used,
W = weight of the oil sample in g.
11.22 =Equivalent weight of Potassium hydroxide

2.5.3 Determination of the saponification value

The saponification value for the samples was determine by weighing 2 g of each treatment oil sample into a conical flask and in accordance with a method described by [6]. A 25 ml of 0.5N alcoholic potassium hydroxide solution was then added to the solution and the flask was heated to boiling over a water bath, with intermittent shaking of the contents. The temperature of the system was raised gradually and was allowed to boil gently for 60mins and afterwards, was allowed to cool slightly. Few drops of phenolphthalein indicator were added and the sample was then titrated with 0.5NHCl until the pink colour ation of the indicator just disappeared while the burette reading was noted and recorded. The same procedure was used for the blank. The saponification value was thus calculated using the following formular:

$$28.06\frac{(a-b)}{W}$$

Where,

a = 0.5N HCl required by main titration
b = 0.5N HCl required by the blank titration
W = weight of oil
28.06=Equivalent weight of Potassium hydroxide

2.5.4 Determination of iodine value

The iodine value was determined according to the method described by [6]. The oil sample (0.4 g) was weighed into a conical flask and 10 ml of Carbon tetrachloride was added to dissolve the oil. A well-fitting glass stopper was inserted to seal the flask and 20 ml of Wij's solution was added to the flask. The flask was sealed more effectively by moistening the stopper with a minimum quantity of 10% solution of potassium iodide. The content of the flask was vigorously swirled and the flask was placed in a dark chamber at a temperature of 15-20°C for 90 mins. At the end of this period, 15 ml of 10% potassium iodide solution and 100 mls of distilled water were added to the mixture. The content was titrated with 0.1M Sodium - thiosulphate solution until the yellow colour disappeared. Thereafter, few drops of 1% starch indicator were added and the titration continued with adiition of thiosulphate drop-wise until the resulting blue coloration disappeared after vigorous shaking [6]. The same procedure was used for the blank so as to authenticate the process. The iodine value (iv) was calculated with the expression:

$$IV = 1269c(V_1 - V_2)m$$

Where,

c = concentration of sodium thiosulphate used
V_1= volume of sodium thiosulphate used for the blank
V_2= volume of sodium thiosulphate used for the test
m = mass of the sample

2.5.5 Determination of the peroxide value

The methods describe by [6] were used for this determination. One gram of the oil sample was weighed into a clean dry boiling tube and one gram powdered potassium iodide (KI) and 20 ml of a mixture of glacial acetic acid and chloroform was added we added to the oil samples in the boiling tube. The tube was then placed in boiling water so that the liquid boiled within 30 sec. The contents of the tube were poured into a flask containing 20 ml of 5% potassium iodide (KI) solution. The tube was washed out twice with 25 ml of distilled water. A few drops of starch solution were added to the mixture and the later was titrated with 0.002M Sodium - thiosulphate. The same procedure was used for the blank at the same time. The peroxide value was determined as follows:

$$Pv = \frac{SN10^3}{W}$$

Where,

s is the ml of $Na_2S_2O_3$ used,
N =the normality of $Na_2S_2O_3$, and
W = the weight of oil sample (g)

2.5.6 Determination of the refractive index

The Refractive Index was measured at 40°C using a Refractometer. The prism box of a refractometer was opened and a few drops of the oil was placed on the ground surface of the lower prism. It was then closed and the box flattened again while ensuring that the oil did not flow away. The cross wires of the telescope was focused by rotating the eye piece and adjusting the mirror so as to get good illumination for a better focus. By means of the lower knob, the prism box was slowly turned backward and forward until the field of view became coloured fringe. By means of the upper knob, the compensator was rotated until the coloured fringe disappeared and the lighted image showed a sharp edge. The prism box was still rotated until the sharp edge was in coincidence with the intersection of the cross-wires in the telescope. The refractive index of the sample was read off on the scale through the eye-piece and thus, recorded.

2.5.7 Determination of the density

The density was determined by a method of [6]. Some quantities of oil was measured into the cylinder of a known weight and the volume was noted and recorded as (B). Thereafter, the cylinder with the oil was weighed together. The already known weight of the cylinder was subtracted from the weight of the oil and cylinder to obtain the weight of the oil (C). The Density however, was expressed mathematically as follows:

$$\text{Density} = \frac{Mass\ (C)}{Volume\ (B)}$$

Thus, the Specific gravity is the ratio of the density of the oil to the density of equal volume of water.

2.5.8 Determination of smoke point

Two grams of the *Hura crepitan* seed oil was weighed into a cylindrical metal container, attached with 0-100°C mercury glass thermometer. The setup is heated at a controlled rate in a GallenKamp Oven set at 105°C. The temperature at the point at which a thin continuous stream of bluish smoke was first observed was measured by the thermometer and taken as the 'smoke point'.

2.5.9 Determination of the colour of oil

The method of [6] was used for this determination. The colour of the oil was measured by a Lovibond Tintometer using I-inch cell. An oil sample was taken from the *Hura crepitan* seed oil and was properly filtered through a porous membrane (filter paper) so as to remove any foreign impurities, traces of moisture and other extraneous materials. The filtered oil was subsequently transfered into a cleaned and dry I-inch glass cell and was placed in position in the tintometer. The colour was matched with sliding red, yellow and blue colours.

The colour of the oil was reported in terms of Lovibond units using the following expressions:

$$Colourreading - (aY + 5bR) or (aY + 10bR)$$

Colour reading – (aY + 5bR) or (aY + 10bR)

Where,

a =sum total of the various yellow slides (Y) used
b = sum total of the various red (R) slides used
Y + 5R is the mode of expressing the colour of light coloured oils and
Y + 10R is the dark coloured oils.

2.5.10 Determination of fatty acid constituents of *Hura crepitan* seed oil

The spectrophotometric method as described by [13] was applied for this determination. A 2.0 gram of oil extracted from the samples of *Hura crepitan* seeds was placed into a 100 ml conical flask containing 20 ml of Benzene and was mixed vigorously to extract the fatty acids. The mixture was transferred into a separating funnel while 2 ml of 10% Copper acetate solution was added to separate the benzene extract. Subsequently, 0 – 10ppm standard solutions of each fatty acid was prepared from each specific standard fatty acid. The absorbances of the different standard solutions and the sample fatty acids were read on a spectrophotometer at a specified wavelength for each fatty acids as follows Linolenic Acid (680nm); Arachidonic Acid (690nm); Behenic Acid (615nm); Palmitoleic Acid (625nm); : Lauric Acid (671nm); Stearic Acid (650nm); Palmitic Acid (630nm); Oleic Acid (670nm); Linoleic Acid (660nm); Myristic Acid (635nm); Capric Acid (640nm); Caprylic Acid (645nm) Lignoceric Acid (695nm). The calibration curves were plotted using the absorbance of each specific standard fatty acid solutions and their corresponding concentration. From the calibration curve of each fatty acid, its slope (gradient) was calculated and used in determining the percentage fatty acid as follows;

$$\%\ FattyAcid = \frac{Absorbance of oil extract \times GradientFactor \times DilutionFactor}{Weight of Sample taken \times\ 10{,}000}$$

2.5.11 Determination of the storage stability of the *Hura crepitan*seed oil

Two sets of fresh *Hura crepitan* seed oil samples extracted from the raw seed flour were placed in 50 ml translucent bottles, each of 12 in duplicate were separately stored in a dark cupboard at room temperature and light for 6 months. The corresponding changes in the oil samples were determined by the measuring the changes in free fatty acids, peroxide value, colour, and iodine value. The analysis was carried out periodically at 30days interval for the six months.

2.6 Statistical Analysis

The data generated was subjected to three-way analysis of variance using Micro soft excel 2007 software and means were separated using Fisher's Least Significant Difference (LSD) at P<0.05.

3. RESULTS AND DISCUSSION

Table 2 shows that Flavonoids, Alkaloids and Tannins were the predominant antinutrients in the *Hura crepitan*.

3.1 Antinutrients *in Hura crepitan* Seed Flour Samples

The predominant anti-nutrients in *Hura crepitan* seed flour were Flavonoids and Alkaloids with values decreasing from 17.5% (raw flour) to 12.16% (fermented flour) for flavonoids and 6.2% (raw flour) to 5.47% (fermented flour) for alkaloids (Table 2). The levels of oxalic acid and Cyanogenic glycosides ranged from 1.76 (raw flour) to 0.86% (fermented flour) for cyanogenic glycoside and 1.48% (raw flour) to 0.32% (fermented flour) for oxalates. While, there were no significant (P>0.05) differences between Alkaloid values, Flavonoid values and cyanogenic glycoside values of the flour samples, there was a significant (P<0.05) difference between the oxalate values in the raw flour and its value in cooked and fermented flours. Phenols, phytates and saponins were present in the seed flour samples but their levels were below 1.0 mg/100 g in all treatments. The values of the anti-nutrients in the *Hura crepitan* seed sample were low when compared to the values obtained by [14], in their work on *L. siceraracia* seeds'. It was reported that phytate values (10.70 and 20.84 mg/g), oxalate (5.85 and 6.45 mg/g), and tannin (5.41 and 22.92 mg/g) for raw and fermented seed. Nevertheless, the values reported for phytate and tannin contents of *Jatropha curcas* were high, being 3688.42 mg/100 g and 590.00 mg/100 g respectively [15].

Notwithstanding that the values of all anti-nutrients analysed were lower in roasted seed flours than their level in raw seed flours which could be attributed to the extent of heat treatment as most of anti nutrients are heat labile and also volatile upon heating (i.e; easily denatured by heat); the roasting operation had less effect in

Table 2. Mean values of Anti-nutrients (mg/100 g) in *H. crepitan* seed flour sample

	Raw seed flour	Cooked seed flour	Roasted seed flour	Fermented seed flour	
					LSD
Alkaloids	6.20[a]	5.67[a]	6.00[a]	5.47[a]	3.82
Tannin	5.24[b]	2.4[a]	1.98[a]	1.50[a]	1.98
Saponin	0.09[a]	0.08[a]	0.11[a]	0.07[a]	1.01
Flavonoids	17.50[a]	14.50[a]	16.50[a]	12.16[a]	6.54
Cyanogenic-glycoside	1.76[a]	1.12[a]	0.94[a]	0.86[a]	1.09
Oxalates	1.48[b]	0.41[a]	1.02[a,b]	0.32[a]	0.82
Phenols	0.37[a]	0.12[b]	0.19[a,b]	0.09[b]	0.21
Phytate	0.56[a]	0.25[b]	0.32[a,b]	0.12[b]	0.25

Means with similar superscripts in the same row are not significantly {p>0.05} different

degrading (reducing) the anti-nutrients studied. Alkaloids and Flavonoids seemed to be more resistant to each of the treatments among the nutrients studied. The flavonoid content of *Hura crepitan* seed flour are higher than values reported for *Garcinia kola* (1.98 mg/100 g) and *Afraniomum meleguetas* (5.76 mg/100 g) and these seeds are used in herbal medicine for the treatment of intestinal troubles [8].

The fermentation process seemed to be more effective for the reduction or degradation of each of the anti-nutrients studied.

However, antinutrients can be quite harmful if found in large amounts in our foods. Phytic acid can tie up the diet supply of phosphorus, calcium, iron, magnesium and zinc, thereby reducing their bioavailability and increasing the risk of some health problem such as anaemia which emanates from iron deficiency [16]. Tannin is a type of enzyme inhibitor that prevents adequate digestion and can cause protein deficiency and gastrointestinal problems. Oxalates inhibit the absorption of plant aminoacids making the proteins to be of "poor quality". Saponins affect the gastrointestinal lining, contributing to 'leaky gut sydrome' and auto immune disorders. They are particularly resistant to digestion by humans and have the ability to enter the bloodstream and trigger immune responses [16]. Cyanogenic glycoside is an effective cytochrome oxidase inhibitor, which interfers with aerobic respiratory systems [17]. Thus fermenting the seed would go a long way in solving the problems of nutrient malabsorption and toxicological effects of the seeds. Generally, these antinutrients bind with the minerals to form complexes these by inhibiting nutrients absorption and bioavailabilities.

3.2 Amino Acid Profile of the *Hura crepitan*

Twenty amino acids namely, alanine, arginine, aspartic acid, cysteine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, threonine, tryptophan, tyrosine, ornithine, cystine, serine and valine were identified in *Hura crepitan* seed flour samples (Table 3). Eleven of these acids (including arginine required by infants) are essential for human nutrition. With regards to the amount of the amino acid in the raw seed sample (before processing), only three of them namely arginine (7.22 g/100 g), glutamic acid (9.87 g/100 g) and valine (8.03 g/100 g) could be regarded as major ones in the seed flour. These three also had values that ranged from 3.25 g/100 g (arginine) to 6.05 g/100 (glutamic acid) in cooked seed flour, 7.28 g/100 g (arginine) to 10.05 g/100 g (glutamic acid) in roasted seed flour and 8.05 g/100 g (arginine) to 10.21 g/100 g (glutamic acid) in fermented seed flour. Both roasted and fermented seed flours had valine values of 8.15 g/100 g and 8.58 g/100 g respectively as compared to a value of 8.03 g/100 g in raw seed flour. Besides arginine (essentially for infants) and valine, onlyleucine, phenylalanine and tyrosine had values up to 3.0 g/100 g in the treatment samples (other than cooked). The cooked samples had the lowest values for all amino acids studied. This observation implied that those amino acids were soluble in water and thus suffered leaching into cook water. Roasted samples had relatively (differences not significant at p>0.05) higher values for all amino acids identified than the raw samples. This could be attributed to higher dry matter resulting from moisture loss (8.0% moisture) in roasted sample as compared to 10% moisture in raw seed samples. Fermented seed samples also had relatively higher values for all amino acids

identified in the seed than the values for the other treatment samples, though there were no significant ($p>0.05$) differences between the values of each amino acid in the samples. Similar increase in the level of amino acids with fermentation have been reported in other seeds like African locust bean [18]. Generally, with regards to nutrition for all ages, the most predominant essential amino acid in *Hura crepitan* is valine with a value of not less than 8.09/100 g that resisted roasting and fermentation effect. Glutamic acid is the most predominant amino-acid with an approximate value of 10.0 g/100 g. Furthermore, roasting and fermentation however increases the amino acid contents of *Hura crepitan* seed flour with corresponding decrease in the anti nutritional factors, this they do by drastically eliminating the anti nutrients which binds with those nutrients to form complexes, hence, reducing their quantity and/or bioavailability of the potential nutrients [18]. Roasting thus deals with heat input and are capable of disintegrating most antinutrients owing to the fact that some of them are labile/sensitive) to heat.

The nutritive value of plant protein quality is usually assessed by comparing its essential amino acids content with reference standards for ideal protein quality set by the World Health Organization [19] which is based on the amino acid requirements for children aged 2-5 years. Therefore, our results showed that *Hura crepitan* seed contain all the essential amino acids needed with phenyalamine and valine having above the 100% relative chemical score of 134.64% and 204.29% respectively. The limiting amino acid is methionine with 44.55% chemical score. The seed has relatively high level of essential and non essential amino acids which makes it a healthy food for human and animal nutrition.

Amino acids are considered very crucial for human health, since they contribute considerably to the health of the human nervous system, hormone production, and muscular structure. In addition, they are needed for vital organs and cellular structure. Deficiency of the essential amino acids may cause hormonal imbalance, lack of concentration, irritability and even depression [20]. The anti oxidant activity of these amino acids suggests a disease preventive role as exemplified by arginine which is beneficial for prevention of cardiovascular disease [14]. Valine, leucine and isoleucine are involved with muscle strength, endurance and muscle stamina. During period of valine deficiency, all of the other amino acids are less well absorbed by gastrointestinal tract [20].

Table 4. revealed that the acid values ranged from 5.055% (oil from cooked sample) to 9.12% (oil from fermented flour sample), the iodine value ranged from 120.5 (cooked) to 171.32% (roasted sample), the peroxide values ranged from 0.72 $meqO_2/kg$ to 2.96 $meqO_2/kg$; the saponification values ranged from 99.58 mg KOH/g (cooked) to 116.4 mgKOH/g (roasted).

3.3 Chemical Properties of *Hura crepitan* Seed Oil

The acid values of the different flour samples studied ranged from 5.05% (cooked sample) to 9.12% (fermented flour). The raw seed flour had an acid value of 7.63% (Table 4). This observation implied that heating effected evaporization of some volatile acids in the samples and fermentation expectedly produced more acids. The acid values of *Hura crepitan* seed oil were higher than that of groundnut oil (5.99%) as reported by [21] and soya bean oil (4.28%) as reported by [22] High acid level indicates that the oil may likely undergo oxidation. The acid value which is twice the free fatty acid value are used to describe how many fatty acids are cleaved from their parent molecules which are triglycerides or phospholipids. However, the cleavage of a free fatty acid from a parent molecule is a hydrolytic break down process and is often used in whole biological systems as an indication of stress [23].

The mean scores for the saponification values of the oil samples ranged from 99.58 mg KOH/g (cooked flour sample) to 116.4 mgKOH/g (roasted flour sample). These values for the oil samples were lower than the range of 188 mgKOH/g to 196 mgKOH/g for most oil of plant origin [24]. Besides the saponification value reported for palm oil is 200 mgKOH/g while the value for groundnut is 193 mgKOH/g and coconut oil value is 257 mgKOH/g [24]. The saponification values of *Hura crepitan* seed oil are close to 116.00 mgKOH/g obtained by [24] for *Afzelia africana* seed oil, *Persea gratesima* (106.60 mgKOH/g).

The saponification values obtained in *Hura crepitan* seed oil suggested that it can be blended with oils of higher saponification values such as coconut oil in the production of soap. This idea can be justified owing to the fact that

Hura crepitan seed oil is relatively high 14.67 g/100 g) in lauric acid as observed in coconut oil also.

Inclusively, the iodine values of seed flour oil samples of *Hura crepitan* ranged from 120.5 I_2g/100 g (cooked sample) to 171.32 I_2g/100 g (roasted sample) but statistically there were no significant ($p>0.05$) differences between the iodine values of the samples. Thus, the iodine values of oil increases with corresponding increase in the degree of saturation of the oil. This observation suggested that cooking might have aided leaching of unsaturated fatty acids, while roasting aid their retention in the oil.

The iodine value range of 120.5 - 171.32 puts the *Hura crepitan* oil in the semi-drying range and it can thus be used in the surface coatings industry to modify alkyl resins [24]. Nevertheless, the iodine value of *Hura crepitan* seed oil can be compared to the values reported for soybean (117-143) and rapeseed oil (94-120) but higher than that of coconut oil (10.0) [25]. The iodine value measures the number of double bonds present in the oil; thus, the degree of saturation f the oil. However, it is not a measure of quality but is an indicator of oil composition.

The peroxide values of the seed flour oil samples ranged from 0.72 meq O_2/kg (roasted seed flour oil) to 2.96 meqO_2/kg (raw seed flour oil). There was a significant ($p<0.05$) difference between the peroxide value (0.72 meqO_2/kg of oil) of the roasted flour oil and those of the other samples. The lowest peroxide value for the oil sample was observed in roasted seed flour oil. This implied that oil oxidation was lower in roasted seeds. The peroxide values of 0.72 meqO_2/kg to 2.96 meqO_2/kg obtained for *Hura crepitan* seed oil are comparable to the 1.50 meqO_2/kg reported for groundnut oil and 2.5 meqO_2/kg reported for cotton seed oil by [26]. The low value indicates that the oil can resist lopolytic hydrolysis and oxidative deterioration. The perioxide value measures hydroperoxide products. It is a good indicator of the primary oxidation products of the oils [23].

Table 4 shows that the refractive index values ranged from 1.410 to 1.457, specific gravity values ranged from 0.890 to 0.905 and smoke point ranged from 209°C to 224°C.

3.4 Physical Properties of *Hura Crepitan* Seed Oils Samples

According to the treatments applied on the *Hura crepitan* seeds, and as as recorded in Table 5, the oil yield ranged from 31.87% (in cooked seeds) to 38.13% (roasted seeds). The raw seed had an oil yield of 35.80% implying that some oil was lost during the cooking in the cook-water. Roasted seeds had relatively higher value (38.13%) of oil yield because roasting affected some drying and loss of moisture in the seeds thereby altering the proportion of oil among the components. However, it can be observed that fermented seed flour freely released its oil during the extraction process. Notwithstanding, there were no significant ($p>0.05$) differences between the values of oil yield of the different treatment samples.

The refractive index value (1.410-1.457) compared favourably with some non-conventional and conventional oil seeds. *Buturospermum parkii* (Shea butter tree) (1.453), *Lophira lanceolata* (Meni oil tree, “Okopia” in Igbo language) (1.459), *Sterculia setegera* (Tropical chestnuts) (1.465), *Detarium microcarpum* (sweet dattock or tallow tree) (1.465), *Blighia sapida* (ackee apple or isin in Yoruba) (1.449), *Scterocarya birrea* (marula ‘hard nut’) (1.422) as reported by [26]. *Cocos nucifera* (1.465) and *Colocynthis citrillus* (coconut) (1.466) [25], *Cucumeropsis efula* (1.470) and *Prunus amygdalus*(almund) (1.470) [27], *Arachis hypogea* (1.449) as reported by [25]. The refractive index showed that the oil contained some double bonds in its fatty acid composition. The refractive index increases as the double bond increases [26].

The specific gravity of the oil was also comparable with other oils, *Afzelia africana* (0.890-0.910), [28]. Specific gravity values of 0.740, 0.924 and 0.926 were reported for custard seed [29], *Nicotiana tabacum L*. [26] and tropical almond [30] respectively. Another study reported 0.88 and 0.93 for soxhlet and mechanically extracted *Cocos nucifera* seed oils respectively and 0.90 and 0.92 for soxhlet and mechanically extracted *Colocynthis citrillus* seed oils respectively [25].

Table 3. Mean values for the amino acid of *H. crepitan* seed flour sample (g/100 g protein)

	Raw seed flour	Cooked seed flour	Roasted seed flour	Fermented seed flour	FAO/WHO UNU reference value	% chemical score (fermented)
Alanine	2.61[a]	1.84[a]	2.69[a]	2.78[a]		
Arginine	7.22[a]	3.25[a]	7.28[a]	8.05[a]		
Aspartic acid	2.87[a]	1.79[a]	3.04[a]	3.26[a]		
Cysteine	2.03[a]	1.54[a]	2.07[a]	2.18[a]		
Glutamic acid	9.87[a]	6.05[a]	10.05[a]	10.21[a]		
Glycine	2.95[a]	1.38[a]	3.21[a]	3.65[a]		
Histidine	2.57[a]	1.27[a]	2.69[a]	2.87[a]	3.4	84.41
Isoleucine	1.69[a]	1.15[a]	1.81[a]	1.95[a]	4.2	46.43
Leucine	3.21[a]	2.75[a]	4.08[a]	4.14[a]	4.2	98.57
Lysine	1.93[a]	1.69[a]	2.01[a]	2.08[a]	4.2	49.52
Methionine	0.78[a]	0.45[a]	0.93[a]	0.98[a]	2.2	44.55
Phenylalanine	3.66[a]	1.33[a]	3.72[a]	3.77[a]	2.8	134.64
Proline	2.59[a]	1.12[a]	2.71[a]	2.74[a]		
Threonine	1.47[a]	1.04[a]	1.62[a]	1.68[a]	2.8	60.00
Trytophan	1.16[a]	1.19[a]	1.23[a]	1.29[a]		
Tyrosine	3.14[a]	1.63[a]	3.21[a]	3.26[a]		
Ornithine	0.25[a]	0.14[a]	0.32[a]	0.35[a]		
Cystine	1.95[a]	1.39[a]	2.09[a]	2.12[a]		
Serine	0.72[a]	0.43[a]	0.88[a]	0.97[a]		
Valine	8.03[a]	4.66[a]	8.15[a]	8.58[a]	4.2	204.29

Means with similar superscripts in the same row are not significantly {p>0.05} different

Table 4. Mean values of the chemical properties of *H. crepitan* seed oil

	Oil from raw seed	Oil from cooked seed	Oil from roasted seed	Oil from fermented seed	
					LSD
Acid value %	7.63[a]	5.05[a]	6.31[a]	9.12[a]	6.02
Saponification value (mg KOH/g)	107.99[a]	99.58[a]	116.4[a]	105.19[a]	20.51
Iodine Value (I_2g/100 g)	126.9[a]	120.5[a]	171.32[a]	152.28[a]	55.98
Peroxide Value (meqO_2/kg of oil)	2.96[a]	2.54[a,b]	0.72[a]	2.94[b]	2.01
Free fatty acid (%)	3.6[a]	2.54[a]	3.15[a]	4.56[a]	3.92

Means with similar superscripts in the same row are not significantly {p>0.05} different

Table 5. Mean values of the Physical properties of *H. Crepitan* seed oil

	Oil from raw seed			Oil from cooked seed		Oil from roasted seed		Oil from fermented seed	
Percentage of oil yield %	35.80[a]			31.87[a]		36.13[a]		37.93[a]	
Refractive Index	1.46[a]			1.44[a]		1.46[a]		1.41[a]	
Specific gravity	0.89[a]			0.90[a]		0.91[a]		0.90[a]	
Smoke Point (^{0}C)	224[a]			216[a]		209[a]		218[a]	
Colour	R	2.00	Golden	4.60	Dark Yellow	2.50	Golden yellow	10.00	Brown
	Y	20.00	Yellow	20.00		20.00		23.00	
	B	0.00		0.00		0.00		19.00	
	N	0.00		0.00		0.00		0.00	
State at (28 – 31)^{0}C	liquid			Liquid		Liquid		liquid	

Means with similar superscripts in the same row are not significantly {p>0.05} different

It can however be seen that the fermentation treatment had adverse effect on the extracted oil more than other treatments. For example, the oil samples extracted from the raw seed and roasted seeds had golden yellow colour, the oil extracted from the fermented seed flour had a brown colour, which implies some higher degree of oxidation than in other samples. The oil from the cooked seed flour was yellow in colour but not as bright and attractive or very appealing as the raw and roasted seed oil samples. Notwithstanding, all the oils were liquid at 28°C-31°C with no significant ($p>0.05$) differences. Their refractive index values were: (1.410- 1.457), specific gravity values (0.890 - 0.905) and smoke point (209°C - 224°C).

However, all the physical properties observed for the oils were in agreement with those recommended by [31] and as such, indicate their non toxicity and edibility.

4. CONCLUSION AND RECOMMENDATION

The results of this study suggested that the *Hura crepitan* seeds high level of proteins in the form of amino acids. The high amino acid profile of *Hura crepitan* seed suggested that the seed could be potentially useful as a good source of food supplement for human nutrition especially for infants and children where protein-energy and malnutrition have continued to hamper growth and development. Likewise, the *Hura crepitan* seed flour could be used for the formulation of animal feeds so as to curb nutrient deficiency in relation to proteins, lipids and minerals. The study also suggested that *Hura crepitan* seed contains some anti-nutritive factors ranging from alkaloids, cyanogenic-glycoside, saponins, oxalates, tannins, flavonoids, phenols and phytates. Flavonoids, alkaloids and tannins were the predominant anti-nutrients. The fermentation process was found to be more effective for the reduction or degradation of each of the anti-nutrients under survey. It is therefore recommended that the *Hura crepitan* seed be incorporated in food processing industries since it contains high amino acid constituents while the anti-nutrients can be degraded using fermentation process. Hence, the oil or the flour should be refined for edibility since they exhibits good physical and chemical properties.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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