

PROXIMATE AND ANTI-PROXIMATE INVESTIGATION OF AFRICAN ELEMI (*CANARIUM SCHWEINFURTHII*)

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ABSTRACT

An investigation into the proximate and anti-proximate composition of African elemi (*Canarium schweinfurthii*) were carried out to ascertain their importance in dietary and in Plant Science and Biotechnology. The laboratory work took place at the Department of Plant Science and Biotechnology, Imo State University, Owerri, Nigeria. The results showed that the moisture content of raw fruit pulp sample AE₁ was (22.50±0.97), this was higher than (18.11±0.36) observed in AE₂. The Ash content reported were (15.10±0.28) for AE₁ and (11.25±0.07) for AE₂. The Crude fibre content obtained were (6.61±0.01) for AE₁ and (11.47±0.47) for AE₂. The fat content of AE₁ was (8.87±0.12) and AE₂ was (9.88±0.68). The Protein content of AE₁ was 18.20±0.00 and 20.65

Introduction

Ayoade *et al.*, (2017) reported that indigenous fruit plays a vital role in the livelihoods of many rural communities in Nigeria, especially those living in the drylands. Anyalogbu *et al.*, (2017) noted that *Canarium schweinfurthii* fruits are among the widely consumed as traditional snacks in East, West and Central Africa. *Canarium schweinfurthii* (African elemi) of Burseraceae family is a perennial plant which is widely distributed in the East, West and Central Africa (Orwa *et al.*, 2009). Burkil (1994) and Orwa *et al.*, (2009) observed that, the plant is found

± 0.00 for AE_2 . The percentage Carbohydrate content of AE_1 was 27.69 ± 1.90 and AE_2 was 30.96 ± 1.01 . There was significant differences ($P > 0.05$) between the proximate samples. The Magnesium value of (13.71 ± 0.020 for AE_1 was higher compared to (11.67 ± 0.03) of AE_2 obtained. The Iron content of AE_1 (1.03 ± 0.04) is higher than (0.62 ± 0.05) of AE_2 . The values obtained for Sodium and Potassium in this research was (6.54 ± 0.04 and 4.97 ± 0.04) for AE_1 and (5.95 ± 0.03 and 4.33 ± 0.04) for AE_2 respectively. Calcium was found to be (7.55 ± 0.04) for AE_1 and (6.78 ± 0.05) for AE_2 . Phosphorous values were 5.89 ± 0.00 for AE_1 and 5.45 ± 0.04 for AE_2 . There was significant differences ($P > 0.05$) between the mineral contents of the samples. The vitamin A content value obtained for AE_1 (53.15 ± 0.73) was higher than the values obtained for AE_2 (30.90 ± 0.65). Vitamins C content value for raw fruit pulp (AE_1) (13.11 ± 0.930) was higher than the values obtained for the blanched fruit pulp AE_2 (10.65 ± 0.44) and Vitamin E content, AE_1 (4.20 ± 0.00) and AE_2 (3.29 ± 0.45). There was significant differences ($P > 0.05$) between the vitamins content of the samples. The flavonoid content of the raw African elemi fruit pulp AE_1 (3.52 ± 0.02) was decreased by blanching to (1.54 ± 0.05) of AE_2 . The phytate content of the raw fruit AE_1 (4.0 ± 0.08) was reduced by blanching to (3.19 ± 0.08) in AE_2 . The phenol content of AE_1 (1.22 ± 0.04) was higher than the value (1.12 ± 0.02) of AE_2 . The saponin content in sample AE_1 and AE_2 were (3.43 ± 0.04) and (3.02 ± 0.10) respectively. Alkaloids content in raw fruit pulp AE_1 was (4.07 ± 0.03) but it was decreased by blanching to (2.27 ± 0.11) AE_2 . Cyanogenic glucoside content was observed in the raw fruit samples AE_1 to be (5.24 ± 0.07) but it decreased by blanching to (4.57 ± 0.55) for AE_2 . There was significant difference between ($P > 0.05$) the samples. Therefore the fruit has low anti-nutritional factors and could be harnessed as sources of nutrients and major contributors to the diet.

Keywords: Proximate, Anti-Proximate, African Elemi (*Canarium Schweinfurthii*), Diet, Investigation.

In parts of middle-belt, South-east and South-west regions of Nigeria. Fruiting periods are different in these regions; therefore availability of the fruit is possible throughout the year. Dawang *et al.*, (2016) stated that the local names for the African elemi include *ube mgba* (Igbo), *ako* or *origbo* (Yoruba), *atili* (Hausa) and *oda* (Idoma). Orwa *et al.*, (2009) and Dongmo *et al.*, (2010) stipulated that different parts of the plant such as leaves, barks, roots and fruits have been used variously as medicine, food, ornaments and fuel.

Ngbede *et al.*, (2008) speculated that the fruit of African elemi is dark brown purplish plum like containing a hard shaped trigonous seed. They stated further that the

purplish ripe fruit are common in the forest zone while the dark brown fruit are common in the savannah region, with a 2.5 – 3.5cm long and ellipsoid, it is made up of three layers which are the epicarp (outer layer), mesocarp (the pulp) and endocarp (the seed), (Olawale, 2012). The pulp is traditionally eaten raw, cooked or softened in warm water, which serves as a traditional snacks or condiment for preparing dish in Nigeria (Anyalogbu et al., 2017; Abayeh, 2000). The fruit looks like the smaller species of African pear (*Dacryodes edulis*) that can be collected and soften immediately by putting it in the mouth or by roasting, frying or soaking in hot water for few minutes usually eaten with roast or boiled corn, but it is not African pear (Olatunji, 2005), while Okpala (2016) observed that African elemi is more harder and requires intense process to get softened. Ayoade *et al.*, (2015) reported that *Canarium schweinfurthii* is one of those plants usually used by indigenous people for its nutritional value; the nutrient composition of the fruit makes it good for use, as nutritional and therapeutically valuable for human. Maduelosi *et al.*, (2015) stated that African elemi produces its fruits during the rainy season between the months of April to September. Ayoade *et al.*, (2017) highlighted that the fruit store best under cold storage thus preventing moisture loss that may result in shrinkage of the fruit surface reducing its aesthetic value and also prevent microbial growth.

Canarium schweinfurthii fruit are hypoallergenic fruits that are eaten by all ages both children and adult since they do not cause any allergic reaction as their pulp has a desirable sweet but not too sugary taste similar to that of avocado pear (Agu *et al.*, 2008). The pulp is of oily consistency and edible (Agu *et al.*, 2008).

Today, medicinal plants are generally embraced by most population in the world especially in the developing countries because they have been discovered to hide the best medicine that nature can offer to mankind. They are also cheaper and more accessible than the conventional orthodox medicine. The African elemi tree is medicinally, pharmaceutical and economically beneficial to human beings. Different parts of the plant such as the leaves, barks, roots and fruits have been used variously as medicine, food, ornaments and fuel. Currently, it is believed that the fresh pulp of this fruit helps in curing chronic eye problems. There is need to investigate the possible significance effect in reducing diseases and to ascertain the dietary potentials of the plant. Results from the study will hopefully serve as guidelines for proper placement of the plant medicinally, pharmaceutically and economically in Southeast Nigeria. The study would not only be useful to Nigeria but also to other African countries that are involved in the production, consumption and exportation of plant produce.

African elemi is one of the indigenous fruits that is gradually going extinct because there is poor utilization of the fruits. African elemi have no industrial uses and there is no

technical know-how on how to preserve them as a result all the harvested fruits are either consumed fast or lost due to post harvest spoilage. Currently there is a growing realization among nutrition experts that fruit should no longer be considered a luxury but a necessity, since they are essentially good for maintenance of health. The prevalence of diet related chronic disease is on the increase in both developed and developing countries and increased consumption of fruits and vegetables of which African elemi fruit is one of them, has been reported to help in alleviating some nutritional and health problems like, diabetes, cardiovascular conditions, osteoporosis, cancer. In southern Senegal it is used by traditional healers as a remedy for diabetes mellitus while in Congo, central African republic and tropical countries it is used to cure fever, serves as a stimulant, emollient, in post partum pain, constipation, malaria, diarrhea, sexual infections and rheumatism, anemia, eye diseases, cardiovascular condition, tooth ache and so there is need to increase the awareness of the benefits of this indigenous fruit to people. The main objective of this research was to determine the proximate and anti-proximate content of African elemi fruit pulp. Specific Objectives are: (a) to investigate the moisture content, ash content, protein content, crude fat, crude fiber and carbohydrate content of African elemi fruit pulp, (b) to determine the Sodium, Potassium, magnesium, Iron, Phosphorous and Calcium, (c) to evaluate the Vitamin A, C and E content of African elemi fruit pulp and (d) to ascertain the saponins, flavonoid, alkaloids, cyanogenic glycoside, phytic acid and phenols content of African elemi fruit pulp.

MATERIALS AND METHODS

Area of Study

The research was carried out at the Plant Science and Biotechnology Laboratory, Department of Plant Science and Biotechnology, Faculty of Biological Sciences, Imo State University, Owerri, Nigeria. Imo State University is sited at latitude 5°10' N and 6° 0' N and Longitude 4° 3.5' E and 7° 0' N in Nigeria.

Sample Procurement

African elemi fruit used for the study was purchased from fruits and vegetable dealers at Ekeonuwa market in Owerri, Imo State.

Processing of the Samples

The ripe *Canarium* fruit was sorted by hand-picking to remove the bad ones and thoroughly washed with clean water to remove dirt adhering to the surface of the fruit. The method enunciated by Nyam *et al.*, (2018) and Ayoade *et al.*, (2017) was used. About

500g of the clean fruit were air dried for 30 minutes, and then split open with a sharp knife to remove the pulp from the seed. The pulp was blended into a powdered form, packaged and labeled AE1 and stored at a temperature of 4°C prior to analysis. Accepted eating tenderness was obtained from another 500g of the raw sample of the fruit by soaking in warm water at 65°C for about 20 minutes to soften the seed coat for easy removal. The seed pulp was removed using sterilized knife and was grounded using Marlex Excella Mixer / Grinding machine. The ground sample was packed in screwed-caped air tight plastic and will be refrigerated at 4°C prior analysis and will be labeled AE2.

Proximate Composition Analysis

Proximate analysis of samples was carried out in triplicate determination using standard methods (AOAC, 2010).

Moisture content

The standard method of the AOAC (2010) was used. Two gram-portions (2g) of each flour samples were weighed into previously dry crucibles with lid. The crucibles with samples were then dried in an oven at 105°C for 24 hours. The crucibles with contents were cooled in desiccators and weighed, then were put back into the oven and the operation was repeated until a constant weight was obtained. The loss in weight obtained represents the moisture content and was calculated with the following formula:

$$\% \text{ Moisture content} = \frac{W_1 - W_2}{W_1} \times \frac{100}{1} \dots\dots\dots \text{Equation 1}$$

Where:

W_1 = will be the weight of sample before drying,

W_2 = will be the weight of sample after drying.

Ash content

The ash content was determined by the furnace incineration method described by AOAC (2010). Two grams (2g) portion of each sample were weighed into previously and dried porcelain crucibles. The crucibles and sample were placed in the muffled furnace at 550°C for 4 hours (until a grey ash will be left). The ash left in the crucible were cooled in desiccators and reweighed. The ash content were calculated as:

$$\% \text{ Ash} = \frac{W_2 - W_1}{\text{Weight of sample}} \times \frac{100}{1} \dots\dots\dots \text{Equation 2}$$

Where :

W_1 = will be the weight of empty crucible

W_2 = will be the weight of the crucible and ash.

Crude Protein content

The crude protein was determined using the micro-Kjedahl technique (AOAC, 2010). One gram (1g) portion of each sample was weighed into filter paper and add into the dry digestion Kjeldahl flask, followed by 0.12g of Copper Sulphate (CuSO_4). Two and half gram (2.5g) of Sodium Sulphate (Na_2SO_4); and 2.50ml of concentrated Sulphuric acid (H_2SO_4) were added with 3g selenium catalyst and a few anti-bumping chips. It was then heated in a flame. It was then heated in a flame chamber until the solution becomes clear (colourless). The solution was cooled to room temperature after which 80ml of distilled water were added. Then, 50ml of 2% boric acid was placed in the receiving flask under the condenser with two drops of methyl red indicator added. The digestion flask was heated until 100ml distillate was collected. And 10ml of the distillate was titrated with 0.649M H_2SO_4 to get pink colour. The same procedure was carried out on the blank.

The amount of Nitrogen was calculated:

Weight of sample = 1.0g

Volume of H_2SO_4 required for the titration = 2.50ml

Normality of H_2SO_4 = N

Nitrogen % of sample (% N)

$$= \frac{\text{Titer} - \text{Blank} \times \text{Normality of acid} \times \text{N Factor}}{\text{Weight of sample}} \dots\dots\dots \text{Equation 3}$$

$$\text{Crude protein} = \% \text{ N} \times 6.25$$

Determination of Crude Fat content

The standard method of the AOAC (2010) was used. Two hundred and fifty ml (250ml) extraction flask was washed, dried in the oven, cooled in desiccators and weighed. The Soxhlet extractor was filtered with a reflux condenser and 3g of the sample was weighed into a filter paper, folded and transferred into a 50mm X 10mm extraction thimble and plugged lightly with a cotton wool. The thimble was placed in the extraction barrel and petroleum ether added until it siphoned over once in the flask directly below it. The flask was then heated and refluxed for seven hours. The thimble was then removed from the barrel and the solvent distilled off until the extraction flask was almost dry. The flask containing the fat was dried in the oven to a constant weight at 40°C to evaporate the solvent completely, then cooled in desiccators and weighed. The difference in weights obtained was used to calculate as the % crude fat.

$$\% \text{ Crude fat} = \frac{W_2 - W_1}{\text{Wt of sample}} \times \frac{100}{1} \dots\dots\dots \text{Equation 4}$$

Where:

W_1 = weight of empty flask

W_2 = the weight of sample with extracted fat

Wt = weight of sample

Determination of Crude fiber

The AOAC (2010) method was used, twenty milliliters (20ml) of 0.1M H_2SO_4 were poured into a 100ml conical flask containing 2g of the defatted sample. The flask was fitted to a reflux condenser. Another 180ml of boiling dilute Sulphuric acid was added into this flask and heat applied so that the entire liquid started boiling in 1min. the refluxing was continued for 30 minutes with occasional shaking to bring down any particles attached to the side. A funnel was -fitted with a filter paper (Whatman 54) and boiling water poured into the funnel and allowed to stand until the funnel become hot. The water was then drained by suction. Then the hot acid mixture was poured immediately into a shallow layer of hot water into the funnel. The suction was adjusted so that the filtration of the bulk of 200ml (20ml + 180ml) was completed within 10 minutes. The residue in the filter paper was washed with boiling water until the washing was acid free. Again the residue was washed back into the original flask containing 200ml of hot 0.3M NaOH. The liquid was boiled for 30 minutes and filtered taking the same precautions as before. Again the residue in the filter was washed with boiling water, then with 1% HCl and finally with boiling water until the washings was free from acid. The residue was then washed with ethanol. The insoluble matter was carefully transferred into a dry and weighed crucible by means of a spatula, taking care not to dislodge any fibre from the filter paper. The crucible was dried in the oven at 70°C for 2 hours, cooled in desiccators and weighed. The residue was ashed into the furnace at 550°C for 3 hours, cooled in desiccators and weighed.

Calculation:

Weight loss during ignition is the weight of the fibre

$$\% \text{ Fibre} = \frac{\text{Weight of fibre}}{2} \times \frac{100}{1} \dots\dots\dots \text{Equation 5}$$

Where 2 is the original weight of defatted sample

Total Carbohydrate

The standard method of the AOAC (2010) was used. This was determined by the differences between the whole sample and the sum of the liquid, ash, protein and crude fibre compositions of the sample.

% Carbohydrate = 100 – (% protein + % fat + % Ash + % Crude fibre).

Determination of Saponins

This was done by the double solvent extraction gravimetric method (Harbone, 1973). Five grams (5g) of the sample was mixed with 50ml of 20% aqueous ethanol solution and incubated for 12 hours at a temperature of 55°C with constant agitation. After that, the mixture was filtered through Whatman No 42 grades of fibre paper. The residue was re-extracted with 50ml of the ethanol solution for 30 minutes and the extracts weighed together. The combined extract was reduced to about 40ml by evaporation and then transferred to a separating funnel and equal volume (40ml) of diethyl ether was added to it. After mixing well, there was a partition and the other layer was discarded while the aqueous layer was reserved. This aqueous layer was re-extracted with the ether after which its pH was reduced to 4.5 with drop wise addition of dilute NaOH solution.

Saponin in the extract was taken up in successive extraction with 60ml and 30ml portion of normal butanol. The combine extract (ppt) was washed with 5% NaCl solution and evaporated to dryness in a previously weighed evaporating dish. The saponin was then dried in the oven at 60°C (to remove any residual solvent) cooled in a desiccators and re-weighed. The saponin was determined and calculated as a percentage of the original samples.

$$\% \text{ Saponin} = \frac{W_2 - W_1}{W} \times 100$$

Where, W, weight of sample to be used

W₁ = weight of empty evaporation dish

W₂ = weight of dish + saponin extract

Determination of Alkaloids

The alkaline precipitation gravimetric method (Harbone, 1973) was used. Five grams (5g) of the sample was dispersed in 100ml of 10% acetic acid in ethanol solution. The mixture was shaken well and allowed to stand for 4 hours at room temperature and shaken every 30 minutes. At the end of this period, the mixture was filtered through Whatman No 42 grade of filter paper. The filtrate extract was concentrated by evaporation, to a quarter of its original volume; the extract was treated with drop wise addition of concentrated NH₃ solution to precipitate the alkaloid. The dilution was done until the NH₃ was in excess. The alkaloid precipitate was removed by filtration using weighed Whatman No 42 filter paper. The paper was dried at 60°C and re-weighed after

cooling in desiccators. The weight of alkaloid was determined and expressed as a percentage of the sample.

$$\% \text{ Alkaloid} = \frac{W_2 - W_1}{\text{Weight}} \times \frac{100}{1}$$

Where, W1 = weight of empty filter paper

W2 = weight of filter paper + alkaloid precipitate

Determination of Flavonoid

This was determined according to the method of AOAC (2000). Five grams (5g) of sample was boiled in 50ml of 2M HCl solution for 30 minutes under reflux. It was allowed to cool and then be filtered through Whatman No 42 filter paper. A measured volume of the extract was treated with equal volume of ethyl acetate starting with drop. The flavonoid precipitated was recovered by filtration using weighed filter paper. The resulting weight difference gives the weight of flavonoid in the sample.

Calculation:

$$\text{Flavonoid (\%)} = \frac{\text{weight of precipitate}}{\text{Initial weight of sample}} \times \frac{100}{1}$$

Determination of Cyanogenic glucoside

This was determined by alkaline pikrate colourimeter method by Balagopalan *et al.*, (1988). One gram (1g) of the sample was dispersed in 50ml of distilled water in a 25ml conical flask. An alkaline pikrate paper was hung over the sample mixture and the blank in their respective flasks. The set up was incubated overnight and each pikrate paper was eluted into a 60ml of distilled water. A standard cyanide solution was prepared and diluted to a required concentrate. The abundance of the eluted sample solution and that of the standard was measured spectrophotometrically at 540nm wavelength with the reagent blank at zero. The cyanide content was determined by the formula shown below:

$$\text{HCN mg/kg} = \frac{1000 \times \text{au} \times \text{C} \times \text{D}}{\text{W} \times \text{as}}$$

Where, W = weight of sample to be analyzed

au = absorbance of test sample

as = absorbance of standard HCN solution

D = Dilution factor where applicable

C = concentration of the standard in mg/dl

Oxalate Determination

This was carried out according to AOAC (1990). Two grams (2g) of the sample was weighed out and extracted thrice at 50°C stirred for 1 hour with 20ml of 0.3M HCl. The combined extract was diluted to 100ml with distilled water and will be used for total oxalate estimation. The oxalate was estimated by pipetting about 5ml of the extract which was made alkaline with 1ml of 5M Ammonium Hydroxide. About 3 drops of Phenolphthalein was added to the extract and acetic acid was added in drops. Also about 1ml of 5% CaCl (aq) was then added to the mixture and allowed to stand for 2 hours after which it was centrifuged at 3,000 rpm for 15mins. The supernatants were discarded and the precipitates washed three times with hot water, thoroughly mixed and centrifuged each time. In the test tube, 2ml of 3M H₂SO₄ was added and the precipitate was dissolved by warming in water bath at 75°C. The content of the test tube was then titrated with freshly prepared 0.01M KMnO₄ at room temperature until the first pink colour appeared throughout the solution. This was then warmed at 75°C and the titration continued until the pink colour persisted.

$$\% \text{ Oxalate} = \frac{V_t}{W_s} \times V_{me} \times \text{Titre}$$

Where, V_t = Total volume of titrate = 100

W_s + Weight of the sample = 2g

V_{me} = Volume - mass equivalent (i.e, 1cm³ of 0.05M KMnO₄ is equivalent to 0.00225g Anhydrous Oxalate acid)

Phytate Determination

This was carried out according to AOAC (1990). Two gram (2g) of the sample was weighed into a test tube. About 10ml of distilled water was added. The sample was extracted using 2ml of 0.2M HCl (aq). About 0.5ml of the extract was pipette into a test tube fitted with glass stopper. Then, 1ml of the solution was added in the tube and covered with stopper. The tube was heated in a boiling water bath for 30mins and the tube was covered very well with the stopper for the first 15mins. Then the test tube containing the solution was cooled in ice water for 15mins and allowed to adjust to room temperature. Then the content of the test tube was mixed very well and centrifuged for 30mins. About 1ml of the supernatant was transferred into another test tube and about 1.5ml of the solution was added. The absorbance at 420nm against distilled water was measured.

$$\% \text{ Phytate} = \frac{A_u}{A_s} \times \frac{C}{W} \times \frac{100}{V_A} \times V_f$$

A_u = Absorbance of test sample

As = Absorbance of standard solution

C = Concentration of standard solution

W = Weight of sample used

Vf = Total volume of extract

Va = Volume of extract

Determination of Phenols

The total phenol was determined by the folin's spectrophotometer, the method described by AOAC (2000). The phenol was extracted in 200mg of the sample with 10ml concentration methanol and the mixture shaken for 30mins at room temperature and centrifuged at 500mg for 15mins and the supernatant decanted and was used for spectrophotometric determination of polyphenol. A portion (1ml) of the extract from each sample was treated with equal volume of folin-Dennis reagent followed by the addition of 2ml of Na₂CO₃ solution; meanwhile standard phenol solution was precipitated and diluted accordingly. 1ml of the standard solution was also treated with folin-Dennis reagent and Na₂CO₃ solution. The absorbance was measured in a spectrophotometer at 560nm; measurement was made with a reagent blank at zero. The phenol content was calculated using the formulae below:

$$\% \text{ Phenol} = \frac{100 * \text{Au} * \text{C} * \text{Vf} * \text{D}}{\text{W} * \text{As} * \text{Va}}$$

Where

W = Weight of the sample

Au = Absorbance of the test sample

As = Absorbance of standard phenol solution

C = Concentration of the standard phenol solution

Vf = Total extract volume

Va = Volume of extract analyzed

D = Dilution factor where applicable.

Determination of Vitamin A

The method according to Delia, (2004) procedure was used. Two grams (2g) of the sample was weighed into a beaker and 10ml of distilled water was added into the beaker and it was shaken carefully to form paste. Twenty five (25ml) of alcoholic KOH solution was added and heated in a water bath for 1hr with frequent shaking. The mixture was cooled rapidly with the addition of thirty (30ml) of water and transferred into a separating funnel. The extraction was carried out three (3) times with 250ml of

Chloroform. Two grams (2g) of Anhydrous Na_2SO_4 was added to the extract to remove any trace of water. The mixture was then filtered with 100ml volumetric flask and made up with water.

Formula : Vitamin A (ug/100g) or (mg/kg) = $\frac{\text{Absorbance of sample} \times \text{Dilution factor}}{\text{Weight of sample.}}$

NOTE: Beta carotene (mg/100ml) = Vitamin A x 6.

Determination of Vitamin C

Vitamin C in the sample was determined using the method of the Association of Vitamin Chemists as described by Kirk and Sawyer (1991). Five grams (5g) of the sample was dispersed in 50ml of EDTAITCA solution and homogenized. The homogenate was filtered; Whatman No 42 filter paper and more of the extractant was used to wash the residue in the filter paper until 50ml filtrate was obtained. A 20ml portion of the filter was measured into a conical flask and 10ml of 30% KT solution was added to it mixed well and then followed by 1 % starch solution. The mixture was titrated against 0.01m CuSO_4 solution. A reagent blank was also titrated. The Vitamin C content was calculated based on the relationship that 1m 10.01m CuSO_4 = 0.88mg Vitamin C.

Therefore Vitamin C mg/100g = $\frac{100}{W} \times 0.88 \times \frac{(T-B)}{V_a} \times V_t$

Where:

W = Weight of sample

T = Time value of sample

B = Time value of blank

Vt = Total extract volume

Va = Volume of extract of extract titrated.

Determination of Vitamin E

The spectrophotometric method of the Association of Vitamin Chemist described by Pearson (1976) was used. 1g of each sample was mixed with 10ml of absolute alcohol and 20ml of molar alcoholic sulphuric acid solution. The mixture was boiled under reflux, under reduced light (aluminium wrapped container) for 45 minutes. 50ml of distilled water was added and the mixture transferred to a separation funnel using an additional 50ml of distilled water to wash out. The unsaponifiable matter was extracted with 5 portion of 30ml diethyleth. The combined extract was washed free of acid by using several portion of distilled water and dried over Sodium Sulphate in a desiccator.

Thereafter, the extract was evaporated until the solvent was gone. The extract was then re-dissolved in 10ml of absolute ethanol standard. Vitamin E in solution was prepared and diluted. 2ml of the extract solution and 2ml of standard solution as well as 2ml of distilled water was dispersed into 3 different test tubes to serve as sample standard and blanks respectively. 5ml of absolute alcohol was added to each test tube followed by 1ml of Conc. HNO_3 which was added with great caution. The test tubes were placed in a water bath at 90°C until the alcohol boils. Boiling was allowed for 3mins only. The volume of the content of each tube was made up to 20ml with absolute alcohol. The absorbance of each was measured at 470nm with the blank at zero. The vitamin E content was calculated as shown below:

$$\text{Vitamin E (1u/100g)} = \text{Au/As} \times \text{C} \times \text{Vf/Va} \times 100/\text{w}$$

Where:

Au = Absorbance of sample

As = Absorbance of standard Vitamin E solution

C = Concentration of standard Vitamin E solution (1u/ml)

Vf = Total extract volume

Va = Volume of aliquote of extract analyzed

W = Weight of sample to be used.

Mineral Analysis

The method described by Onwuka (2005) was used in the determination of mineral content of sample. Half gram (0.5g) of the dried flour sample was weighed into a pre-acid rinsed digest tube. 10cm^3 of 6M HCl was added and heated to dryness in a water bath. The residue was dissolved in a mixture of 10cm^3 of 6M HNO_3 acid, warmed o a water bath and filtered using a Whatman filter paper into 100cm^3 calibrated flasks. The filter paper was washed with distilled water and the filtrate diluted with distilled water and made up to the 100cm^3 mark. The digest was for the determination of Calcium, Sodium and Potassium by the flame photometry method. The heavy metals such as Na, Fe, K, P, Ca and Mg were determined using the atomic absorption spectrophotometer method.

Statistical Analysis

Data was subjected to descriptive and Chi-square tests using Statistical Package for Social Sciences (SPSS) version 20.0 USA Inc (2000).

RESULTS

Table 1 shows the proximate composition of raw (AE_1) and blanched (AE_2) of African elemi fruit pulp. The moisture content of raw African elemi fruit pulp AE_1 (22.5 ± 0.97)

was higher than that of blanched fruit pulp AE₂ (18.11±0.36). There was significant difference (P>0.05) between the samples. The Ash content of the raw sample AE₁ (15.10±0.28) was higher than that of blanched African elemi fruit pulp AE₂ (11.25±0.07). There was a significance difference (P>0.05) between the samples. Crude fibre content of AE₂ (11.47±0.47) was higher than that of AE₁ (8.87±0.12) indicating a significant difference (P>0.05) between the samples. The study observed a significant difference (P>0.05) in the Protein content of AE₁ (18.20±0.0) and AE₂ (20.65±0.0) while Carbohydrate content of AE₂ (30.96±1.01) was higher than that of AE₁ (27.69±1.9) and there was a significant difference (P>0.05) between the samples.

Table 1: Proximate Composition of Raw and Blanched African elemi fruit pulp

Nutrient %	AE1	AE2	P Value	T Value	Remark
Moisture	22.5±0.97	18.11±0.36	4.39	0.134	S
Ash	15.10±0.28	11.25±0.07	3.85	0.025	S
Crude fibre	6.61±0.01	11.47±0.47	4.86	0.06	S
Fat	8.87±0.12	9.88±0.68	1.01	0.236	S
Protein	18.20±0.0	20.65±0.0	2.45	0.00	S
Carbohydrate	27.69±1.9	30.96±1.01	3.27	0.124	S

Value given are Mean ± Standard Deviation of duplicate determination (significant confidence level 99%); AE₁ = Raw African elemi fruit pulp; AE₂ = Blanched African elemi fruit pulp; S = Statistically significant; NS = Not Statistically significant.

The Table 2 indicates the Vitamin content of raw and blanched African elemi fruit pulp. The raw African elemi fruit pulp AE₁ contains (53.15±0.73) of Vitamin A while the blanched fruit pulp AE₂ contains (30.90±0.65). there was a significant difference (P>0.05) between the samples. The Vitamin C content of AE₁ (13.11±0.93) was higher than AE₂ (10.65±0.44) and there was a significant difference (P>0.05) between the samples. The study observed a significant difference (P>0.05) in the Vitamin E content of AE₁ (4.21±0.0) and AE₂ (3.29±0.45).

Table 2: Vitamins Content of Raw and Blanched Africa elemi fruit pulp

Vitamins (Mg/100g)	AE1	AE2	P Value	T Value	Remark
Vitamin A	53.15±0.73	30.90±0.65	22.25	0.019	S
Vitamin C	13.11±0.93	10.65±0.44	2.46	0.089	S
Vitamin E	4.21±0.0	3.29±0.45	0.92	0.209	S

Values given are Mean $\pm$ Standard Deviation of duplicate determination (Significant confidence level 95%); AE₁ = Raw African elemi fruit pulp; AE₂ = Blanched African elemi fruit pulp; S = Statistically significant; NS = Not Statistically significant.

Table 3 shows the result of the Mineral contents of raw and blanched African elemi fruit pulp samples. The Magnesium content of the raw fruit pulp AE₁ (13.71 ± 0.02) was higher than that of the blanched fruit pulp AE₂ (11.67 ± 0.03) which there was a significant difference ($P > 0.05$) between the samples. The Iron content of the samples was (1.03 ± 0.04) for AE₁ and (0.62 ± 0.05) for AE₂ showing a significant difference ($P > 0.05$). The Sodium content of AE₁ (6.54 ± 0.04) was higher than that of AE₂ (5.95 ± 0.03) and there was a significant difference ($P > 0.05$) between the samples.

Potassium content of AE₁ (4.97 ± 0.04) and AE₂ (4.33 ± 0.04) was significantly different ($P > 0.05$) between the samples. The Calcium content of AE₁ (7.55 ± 0.04) was significantly higher than that of AE₂ (6.78 ± 0.05). There was a significant difference ($P > 0.05$) between the samples and Phosphorous (5.89 ± 0.00) for AE₁ and (5.45 ± 0.04) for AE₂ shows a significant difference ($P > 0.05$).

Table 3: Mineral Contents of Raw and Blanched African elemi fruit pulp

Minerals (Mg/100g)	AE ₁	AE ₂	P Value	T Value	Remark
Magnesium	13.71 ± 0.02	11.67 ± 0.03	2.04	0.004	S
Iron	1.03 ± 0.04	0.62 ± 0.05	0.41	0.02	S
Sodium	6.54 ± 0.04	5.95 ± 0.03	0.59	0.02	S
Potassium	4.97 ± 0.04	4.33 ± 0.04	0.64	0.00	S
Calcium	7.55 ± 0.04	6.78 ± 0.05	0.77	0.01	S
Phosphorous	5.89 ± 0.00	5.45 ± 0.04	0.44	0.004	S

Value given are Mean $\pm$ Standard Deviation of duplicate determination (significant confidence level 95%); AE₁ = Raw African elemi fruit pulp; AE₂ = Blanched African elemi fruit pulp; S = Statistically significant; NS = Not Statistically significant.

The table 4 shows the result of the Anti-nutrient content of raw AE₁ and blanched AE₂ African elemi fruit pulp. AE₁ was higher in flavonoids (3.52 ± 0.02), phytate (4.0 ± 0.08), phenol (1.22 ± 0.04), saponin (3.43 ± 0.04), alkaloids (4.07 ± 0.03), and cyanogenic glucoside (5.24 ± 0.07) than in AE₂ with flavonoid (1.54 ± 0.05), phytate (3.19 ± 0.08), phenol (1.12 ± 0.02), saponin (3.02 ± 0.10), and alkaloid (4.57 ± 0.55). The study showed a significant difference ($P > 0.05$) between the samples while a non significant difference ($P > 0.05$) was observed in phenol.

Table 4: Anti-nutrient content of Raw and Blanched African elemi fruit pulp

Anti-nutrient	AE ₁	AE ₂	P Value	T Value	Remark
Flavonoids (%)	3.52±0.02	1.54±0.05	1.98	0.06	S
Phytate (%)	4.0±0.08	3.19±0.08	0.81	0.094	S
Phenol (%)	1.22±0.04	1.12±0.02	0.10	0.30	NS
Saponin (%)	3.43±0.04	3.02±0.10	0.41	0.07	S
Alkaloids (%)	4.07±0.03	2.27±0.11	1.80	0.018	S
Cyanogenic Glucoside (Mg/100g)	5.24±0.07	4.57±0.55	0.67	0.301	S

Values given are Mean± Standard Deviation of duplicate determination (significant confidence level 95%); AE₁ = Raw African elemi fruit pulp; AE₂ = Blanched African elemi fruit pulp; S = Statistically significant; NS = Not Statistically significant.

DISCUSSION

Proximate Composition of Raw and Blanched African Elemi Fruit Pulp

The result of proximate composition of *Canarium* fruit are shown in Table 1. Moisture content is a measure of the water content in the fruit samples, moderate moisture content indicates that it can be stored for a long time without the development of moulds (Umar *et al.*, 2007). The moisture content of raw fruit pulp sample AE₁ was (22.50±0.97), this was higher than (18.11±0.36) observed in AE₂. There was significant difference (P>0.05) between the samples. The moisture content in both AE₁ and AE₂ were lower than the values (30.21±1.03) reported by (Ayoade *et al.*, 2017) and 25.62% reported by Maduelosi and Angaye (2015). The Ash content reported in this study (15.10±0.28) for AE₁ and (11.25±0.07) for AE₂ were higher to the values 3.31% reported by Maduelosi and Angaye (2015), (1.86±0.01) reported by Ayoade *et al.*, (2017). The Ash content of a biological material is an analytical term which refers to the inorganic residue that remains after the organic matter has been burnt away (Maduelosi and Angaye, 2015). Ash content gives an idea of the amount of mineral element present in the sample while the organic matter gives an estimate of Protein, Lipid (Fats), Carbohydrate and Nucleic acid content of the sample (Onwuka, 2005). The Crude fibre content obtained for raw *Canarium* fruit (AE₁) was (6.61±0.01) and blanched *Canarium* fruit pulp (AE₂) was (11.47±0.47). This value is lower than 17.90% of African pear reported by Stace (1990) and higher than (3.19±0.02) reported by Ayoade *et al.*, (2017). The recommended dietary allowance (RDA) of fibre for children, adults, pregnant and breast feeding mothers are 19 - 25%, 21 - 38%, 28% and 29% respectively (Ayoade *et al.*, 2017), therefore *Canarium* fruit is not a good source of dietary fibre for humans however, it could be recommended for low fibre feed formulation or supplement (Ayoade *et al.*, 2017). The fat content of AE₁ (8.87±0.12) and AE₂ (9.88±0.68) was lower

than the values (34.83 ± 0.05) reported by (Ayoade *et al.*, 2017). The Fat content of *Canarium* fruits makes it suitable as a source of edible Oils and could be applicable in high Lipid foods such as margarine (Ayoade *et al.*, 2017). The Protein content of 18.20 ± 0.00 (AE₁) and 20.65 ± 0.00 for (AE₂) are closely related to that in protein rich food such as beans, cowpea, pigeon peas, melon and pumpkin which ranges between 23.1 – 33.00% (Olaofe, 1994). The percentage Carbohydrate content (27.69 ± 1.90) in sample AE₁ and (30.96 ± 1.01) in sample AE₂ falls within the acceptable values for edible fruit (Kirk and Sawyer, 1991).

Vitamin Content of Raw and Blanched African Elemi Fruit Pulp

The result of vitamin content of the raw fruit pulp AE₁ and blanched fruit pulp AE₂ was shown in Table 2. The raw fruits (AE₁) had the highest vitamin A, C and E content. There was significant difference ($P > 0.05$) between the samples. The vitamin A content value obtained for AE₁ (53.15 ± 0.73) was higher than the values obtained for AE₂ (30.90 ± 0.65). Vitamin A (B- carotene equivalent) are fat soluble vitamins and was the reason they are not leached into the water used in processing, but losses are mostly as a result of thermal destruction and oxidation (Fellows, 2000). The vitamin A in the fruit pulp helps improve eye vision (Onwordi, 2014).

Vitamin C content value for raw fruit pulp (AE₁) (13.11 ± 0.930) was higher than the values obtained for the blanched fruit pulp AE₂ (10.65 ± 0.44) and there was significant difference ($P > 0.05$) between the samples. Vitamin C is a water soluble vitamin and heat labile, thus losses in the vitamin content might have occurred during blanching leading to lower vitamin C value in blanched fruit pulp. Vitamin C is anti-scurvy, it helps in the development of tissues, bones and also helps the body resist infection (Olusanya, 2008). Vitamin C also reduces age related diseases, if *Canarium* fruit is consumed regularly it shows the risk of macular degeneration in aged people (Perry *et al.*, 2000).

Vitamin E content, AE₁ (4.20 ± 0.00) has high vitamin E content (tocopherol) are fat soluble vitamin and losses in vitamin content are as a result of thermal destruction and oxidation (Fellow, 2000). Vitamin E is useful for skin and hair care (Onwordi, 2014). Vitamin A (Carotenoids), C (Ascorbic acid) and E (Tocopherol) are antioxidants that have been associated with prevention of nutritionally associated disease such as cancer, coronary heart and obesity (Larauri *et al.*, 1996).

Mineral Content of Raw (AE₁) and Blanched (AE₂) African elemi fruit pulp

Table 3 shows the mineral content of raw AE₁ and blanched fruit pulp of African elemi AE₂. Magnesium is an element in connection with circulatory disease and calcium metabolism in bone (Ishida *et al.*, 2000); it is also involved in bone mineralization, the

building of protein enzyme action, normal muscular contraction and transmission of nerve impulses (Ayoade *et al.*, 2017). The Magnesium value of (13.71 ± 0.020 for AE₁) was higher compared to (11.67 ± 0.03) of AE₂ obtained in this study. The value obtained for Magnesium in the study was lower compared to (24.00 ± 0.02) value reported by Ayoade *et al.*, (2017).

Iron is essential micronutrient for haemoglobin formation, normal functioning of central nervous system (CNS) and in oxidation of Carbohydrate, Protein and Fats (Ayoade *et al.*, 2017). The Iron content of AE₁ (1.03 ± 0.04) is higher than (0.62 ± 0.05) of AE₂ reported in this research, and there was significant difference ($P > 0.05$) between the samples. The raw fruit pulp (AE₁) had appreciable amount of Iron, its consumption could be encouraged for menstruating and lactating women (Ayoade *et al.*, 2017).

Sodium content in combination with Potassium was involved in proper acid-base balance, maintenance and nerve transmission in the body system (Ayoade *et al.*, 2017). The values obtained for Sodium and Potassium in this research was (6.54 ± 0.04 and 4.97 ± 0.04) for AE₁ and (5.95 ± 0.03 and 4.33 ± 0.04) for AE₂ respectively. There was significant difference ($P > 0.05$) between the samples. The variation of Sodium to Potassium in this research was of significant importance particularly to hypertensive patient (Umar *et al.*, 2007). A high intake of Potassium has been reported to protect against increasing blood pressure and other cardiovascular risks (Tene *et al.*, 2016).

Calcium which forms component of bones and teeth necessary for blood clotting and muscle contraction was found to be (7.55 ± 0.04) for AE₁ and (6.78 ± 0.05) for AE₂ and there was significant difference ($P > 0.05$) between the samples.

Phosphorous is related to Calcium for bone, teeth, muscle growth and maintenance of health (Umar *et al.*, 2007). 5.89 ± 0.00 for AE₁ and 5.45 ± 0.04 for AE₂ obtained for Phosphorous indicate that there was significant difference ($P > 0.05$) between the samples. The availability of Calcium in the body depends on Calcium to Phosphorous ratio and the presence of anti-nutritional factors (Ayoade *et al.*, 2017). For the absorption of Calcium in the intestine the ratio of 1.1 is required from both Calcium and Phosphorous (Umar *et al.*, 2007).

Anti-nutritional Composition of Raw (AE1) and Blanched (AE2) African elemi fruit pulp

The result in Table 4 shows that the anti-nutrient concentrations in the fruit pulp were affected by the processing method. The flavonoid content of the raw African elemi fruit pulp AE₁ (3.52 ± 0.02) was decreased by blanching to (1.54 ± 0.05) of AE₂. Flavonoid has protective effects including anti-inflammatory, anti-oxidant, anti-viral and anti-carcinogenic properties (Omodamiro *et al.*, 2016; Anyalogbu *et al.*, 2017). At very high concentrations they manifest a myriad of anti-nutrient activities, they chelate metals

such as Iron, Copper and Zinc and decrease their absorption (Karamac, 2009), and they also inhibit digestive enzymes and may also precipitate proteins (Ademezyke *et al.*, 2017). The flavonoid content in both AE₁ and AE₂ (3.52 ± 0.02 and 1.54 ± 0.05) is not considered toxic to the consumer, this is because the USDA estimates that in the U.S. daily total flavonoid consumption by the average adult is approximately 250-275mg (Whfood.org; 2016) and the value obtained in this research was lower than the value (14.98 ± 2.91) AE raw reported by Anyalogbu *et al.*, (2017).

The Phytate or Phytic acid is the storage form of Phosphorous an important mineral used in the production of energy as well as the formation of structural elements like cell membrane (Jacela *et al.*, 2010). But at high level they bind to essential mineral such as Iron, Zinc, Calcium and Magnesium in the digestive tract and inhibit their absorption by the body (Weaver and Kannan, 2002). The phytate content of the raw fruit AE₁ (4.0 ± 0.08) was reduced by blanching to (3.19 ± 0.08) in AE₂. Phytate can also act as anti-oxidant exhibit anti-cancer properties and may have a positive impact on cholesterol and blood sugar (Onomi *et al.*, 2004). Both samples are below the tolerable limit for phytate and so could be considered safe for human consumption since the tolerable limit is 20.00-25.00mg/100g (Weaver and Kannan, 2002).

The phenol content of AE₁ (1.22 ± 0.04) was higher than the value (1.12 ± 0.02) of AE₂. Although polyphenol are anti-oxidant compounds, phenol itself is inactive as an anti-oxidant but thought to have medicinal properties (Landete, 2013; Haslam *et al.*, 1989). Phenol may form complexes with essential amino acids, enzymes and other proteins and nutrient and also form insoluble iron-phenol complex in the gastrointestinal tract and thus makes the iron unavailable for absorption (Shahidi and Naczki, 1992). The two samples are considered safe for consumption as the value obtained is below the tolerable limits of its anti-nutrients content (IPCS, 1994).

The saponin content in sample AE₁ and AE₂ were (3.43 ± 0.04) and (3.02 ± 0.10) respectively. There was significant difference between ($P > 0.05$) the samples. Saponin exhibits anti-nutrient factors or toxicity when consumed in large amounts, involving possible liver damage, gastric pain, diarrhea or other adverse effects (Quillaja, 2018). The natural tendency of saponins to ward off microbes makes them good candidates for treatment of fungal infections and so serves as natural antibiotics helping the body to fight infection and microbial invasion (Okwu, 2004). The result of the research shows there is a relatively low presence of saponins in both samples.

Alkaloids content in raw fruit pulp AE₁ was (4.07 ± 0.03) but it was decreased by blanching to (2.27 ± 0.11) AE₂. High level of Alkaloids exerts toxicity and adverse effects to humans especially in physiological and neurological activities (Anyalogu *et al.*, 2017). Lower dose of alkaloids have a wide range of pharmacological activities including anti

malaria antiasthma, anticancer analgesic and anti bacterial (Kittakoop *et al.*, 2014; Raymond *et al.*, 2010 and Cushnie *et al.*, 2014). The Alkaloid content in the fruit samples is safe for consumption.

Cyanogenic glucoside content was observed in the raw fruit samples AE₁ to be (5.24±0.07) but it decreased by blanching to (4.57±0.55) for AE₂. The observed decrease with processing may be due to vaporization of free cyanide by heat (Anyalogbu *et al.*, 2017). The toxicity of cyanogenic glucoside is associated with their ability to be hydrolysed either spontaneously or in the presence of enzyme to produce cyanide as end product of their hydrolysis (Islamiyat *et al.*, 2016). Toxicity may result in a cute poisoning and has also been implicated in the etiology of several chronic diseases (Cressey and Sunders, 2012). The concentration of cyanogenic glucoside in both samples was below the lethal dose of 50 – 60g/kg (Inuwa *et al.*, 2011) thus, the fruits are free of HCN toxicity risk.

CONCLUSION

The *Canarium* fruit pulp both raw and processed contains appreciable levels of protein and carbohydrate. The research further revealed that the fruit contain appreciable levels of vitamin A and C and nutritionally valuable minerals such as Magnesium, Iron, Sodium, Potassium, Calcium and Phosphorous. Additionally and more importantly, certain anti-nutritive agent was discovered like phytate, flavonids, oxalate, phenol, saponin, alkaloids and cyanogenic glucoside which are toxic and interfere with digestion and absorption, but all are below the toxic level or acceptable daily intake, therefore the fruit has low anti-nutritional factors and could be harnessed as sources of nutrients and major contributors to the diet.

RECOMMENDATION

Based on the findings from the study, further research should be carried out in the effective utilization of *Canarium* fruits not only as a fruit to be eaten but also as a raw material in production of edible oil. Also *Canarium schweinfurthi* should be planted on a large scale to ensure its sustainable utilization. Awareness also has to be created through nutrition education so that people will understand fully the wonderful benefits of this fruits especially those in urban areas.

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