INTERNATIONAL JOURNAL OF INNOVATIVE RESEARCH & DEVELOPMENT

ISSN 2278 – 0211 (Online)

Proximate Composition and Ascorbic Acid Content of *Chrysophyllum albidum* (African Star Apple) Seeds

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Abstract:
The chemical evaluation of Chrysophyllum albidum seeds was carried out to ascertain the proximate composition, ascorbic acid content and its mineral content using standard analytical methods. The result of the proximate composition was shown to be moisture (14.00±0.22%), ash (4.00±0.10%), fat (25.58±0.51%), fibre (16.00±0.13%), protein (25.76±0.45%) and carbohydrate (28.66±0.01%). The ascorbic acid content was found to be 5.16%. The mineral content of the seed was found to be Calcium (18.04±0.05mg/g), Iron (33.20±0.12mg/g), Potassium (2.50±0.02mg/g), Magnesium (0.16±0.01mg/g), Sodium (0.15±0. 02mg/g), phosphorus (9.80±0.10mg/g), and zinc (34.00±0.14mg/g). This study revealed that Chrysophyllum albidum seeds have low moisture content, high fat, fibre, ash and protein content with relatively low amount of ascorbic acid and a good source of iron, zinc, phosphorus, calcium and lipid. Hence, it is a viable source for vegetable oil, mineral supplements and vitamin C.

Keywords: *proximate, composition, Ascorbic acid, mineral, African Star Apple*

1. Introduction

Chrysophyllum albidum popularly known as African star apple is a member of the plant family known as sapotaceae. It has a milky sweet pulp (which is usually eaten) that houses the seeds (which are not edible) in Nigeria. The plant pulp and Peel have been reported to be excellent sources of energy in animal and human diets as it is rich in digestible proteins and carbohydrates (Edem and Dosunmu, 2011)with good array of minerals and amino acids (Del Rosario et al.,1998).

The fruit pulp has been found to have the highest content of ascorbic acids (Asenjo, 1946). This acid is a sugar acid with antioxidant properties and it plays a role in the synthesis of collagen and elastin (collagen and to a large extent elastin is a significant component of blood vessels, bones, joints, teeth, gums and all connective tissue in the body). The fruit is known to contain about 90% anarcardic acid. This acid is very useful industrially in protecting wood and as a source of resin (Amusa et at., 2003).

This present study has the sole aim of presenting *Chrysophyllum albidum* seeds obtained from botanical garden in University of Calabar, Calabar Cross River State with the view to promoting this non-conventional, unexploited seeds of the plant as a good source of crude fat, protein, fibre, vegetable oil and minerals (amongst other non-conventional oil seeds).

2. Materials and Methods

2.1. Sample Collection and Preparation

Fruits of *Chrysophyllum albidum* were obtained from the botanical garden in the University of Calabar, Calabar Cross River State of Nigeria and was authenticated by an Herbarium, Botany Department of University of Calabar. The seeds were then removed from the fruits, cracked and place on the asbestos pad and dried in an oven at a temperature of 60-70^{0}c. They were brought out and turned over for air circulation. The dried seeds were ground into powdered form and stored in an air tight container for further analysis.

2.2. Proximate Analysis

Proximate analysis involves the determination of moisture, ash, crude fibre, crude protein, crude fat and carbohydrates (Verma, 2010). Carbohydrates were calculated while ash, moisture, fats, fibre, and crude protein were determined using methods described by (AOAC 2000). All determinations were carried out in triplicate. The procedures are as follows:

2.3. Moisture Content

The samples were weighed (5g) into pre-weighed beaker and placed in an oven for about six (6) hours at a temperature of 100^{o}C to a constant weight. The loss in weight was expressed as a percentage of the initial weight. Thus, the difference in weight indicates the amount of water contained in the sample (AOAC 2000).

2.4. Ash Content

5.0g sample in a pre-weighed label crucible was placed in the muffle furnace at a temperature of about 500^{o}C for 20 minutes. The furnace was allowed to cool before removing the crucible with its content. The crucible was later cooled in desiccator and reweighed to get the ash content (AOAC 2000).

2.5. Crude Fat Content

5.0g sample in a soxhlet extractor thimble was wrapped with a filter paper and plugged tightly with a filter paper and with cotton wool. 150ml of petroleum ether (bpt 60 – 80^{o}C) was poured into 300ml round bottom flask containing anti-bombings and the soxhlet extractor assembled. The samples were extracted for about 4hrs until the soxhlet become colourless. The extracts were poured into a dried pre-weighed beaker and the thimble rinsed with a little quantity of petroleum ether back into the beaker. The beaker was heated on a steam bath to drive off the solvent. The extracted lipid left in the beaker was dried in desiccators and weighed (AOAC 2000).

2.6. Crude Fibre Content

5.0g sample was put in a pre-weighed beaker. 50ml of 1.25% H_2SO_4 solution was added and made up to 200ml with distilled water and stirred. The mixture was heated with continuous stirring for thirty (30) minutes and allowed to cool and settle. Distilled water was added and allowed to settled then decanted, decantation was repeated for six (6) times consecutively to make the mixture acid free. 50ml of 1.25% NaOH was added to 200ml distilled water in a beaker and heated for thirty (30) minutes with continuous stirring. It was cooled and settled. Distilled water was added and decanted for six (6) times consecutively. The mixture was filtered and kept for about forty-five (45) minutes for water to drain completely and the weight taken (AOAC 2000).

2.7. Carbohydrate Content

This was obtained by taking each percentage value of protein, fat, fibre and ash content from the total dry matter.

% carbohydrate = 100 – (% fat + % fibre + ash + % protein)

2.8. Crude Protein (Modified Kjeldahl Method)

The analysis was carried out in three (3) stages, these were;

- The digestion stages
- The distillation stages
- The titration stages

Digestion Stage: 5g sample was put in a 250ml kjeldahl flask. 2g each of the kjeldahl catalysts (Copper Sulphate and Sodium Sulphate) were weighed into the kjeldahl flasks. Anti-bumping granules were added and 30ml of concentrated Sulphuric acid was also added to the flask. The digestion flask was then placed on the heating mantle for an hour before being transferred to electric stove. The digestion process proceeds with occasional swirling until a clear solution was obtained. The clear solution was transferred into a 100ml standard flask and made up to the mark with distilled water.

Distillation Stage: 10ml of the digest was measured into the micro distillation apparatus. 12.5ml of 1.25% NaOH was also added to the flask. A condenser was connected from the distillation apparatus to a volumetric flask containing 10ml of 5% boric acid and 2 drops of double indicator (methyl red and methyl blue). The distillate was collected in a flask and then titrated with 0.1M standard hydrochloric acid until a pale pink colour end point was obtained.

$$\% \text{ Nitrogen} = \frac{ml of HCl(sample) - ml of HCl(blank) - molarity of HCl}{weight of sample \times ml of digest \times 1000}$$

% Protein = % Nitrogen × Protein factor (AOAC 2000).

2.9. Determination of Ascorbic Acid Content. (Redox Titration Using Iodine Solution).

The solution of iodine 5.0 × 10^{-3}M was prepared. 5g of the sample was made into a paste by further grinding with mortar and pestle. 100ml of distilled water was then added to the paste. The solution was filtered. 10ml of the filtrate was

pipetted into a conical flask. It was then titrated with the iodine solution until a dark blue-black colour was obtained. The titration was repeated with further aliquots (10ml) of sample solution until concordant results were obtained.

3. Digestion of Sample

The powdered samples were weighed (5g) into a conical flask. 25ml of concentrated nitric acid was added to the sample. The conical flask with its content was gently heated (50-70^{0}c) on a hot plate until the colour changed from brown to colourless. The digest was made up to 100ml with deionise water. Appropriate dilutions were made for each element. All determinations were carried out in triplicate.

3.1. Determination of Sodium and Potassium using Flame Photometer

The recommended standard method of (AOAC 2000) was used for the analysis. Standard solution and aliquots of diluted clear digest were used for flame photometry.

3.2. Determination of Ca, Mg, P, Fe and Zn Using Atomic Absorption Spectrophotometer

The method of (AOAC 2000) was used in these determinations. Standard solutions were prepared for each element. Aliquots of the diluted clear digest were used for atomic absorption spectrophotometry.

4. Result and Discussion

Seed	Proximate Composition(%)	Elemental Composition(Mg/100g)	Ascorbic acid (%)
	Moisture Ash Fat Fibre Protein Carbohydrate	Ca Fe K Mg Na P Zn	
Chrysophyllum albidum	14.00±0.22 4.00±0.10 25.58±0.51 16.00±0.13 25.76±0.45 28.66±0.01	18.04±0.015 33.20±0.12 2.50±0.02 0.16±0.01 0.15±0.02 9.80±0.10 34.00±0.14	5.16
Chrysophyllum cainato	56.04±1.64 0.84±0.25 15.81±0.53 4.19±0.46 4.63±0.12 78.49±0.04	- - - - - - -	
African Oil bean	6.54±0.01 1.94±0.01 44.24±0.10 2.56±0.02 34.42±0.02 10.30±0.02	- - - - - - -	
Cowpea	5.20±0.01 2.48±0.01 41.84±0.04 2.37±0.00 21.46±0.04 28.05±0.06	2.10±0.21 10.10±0.90 7.33±0.34 1.94±0.93 0.8.±0.69 - 0.06±0.03	
Locust bean	4.81±0.02 4.81±0.04 49.20±0.04 4.66±0.01 33.50±0.12 -	1.47033 50.67 - 30.60 - - -	
Watermelon Lagenaria	3.81±0.00 2.48±0.01 41.84±0.04 2.37±0.00 21.46±0.04 28.05± 0.06	- 2.73 0.22 - 0.08 0.22 0.81	66.88
Data are : Mean ±	Standard deviation of triplicate determinations.		

Table 1: Results of Proximate Composition, Ascorbic Acid Content and Elemental Composition of Chysophyllum albidum seeds compared with other commonly known seeds (adopted from other published work)

5. Discussion

Table 1 shows the proximate composition of *Chrysophyllum albidum* seed.From the result obtained, moisture content was found to be 14.00±0.22%. This value is lower than 56.04±1.64 for *Chrysophyllum cainito* seed (Oranusi et al., 2015). This connote that this seed will have relatively long shelf life. This is because the moisture content of seeds, fruits and vegetable is indicative of their shelf life. The higher the moisture content, the more susceptible the seed, fruits and vegetable is to microbial attack. Ash was found to be 4.00±0.10%. This is higher than 2.6% for African oil bean (Odoemelam, 2005), 2.48±0.01 for both cowpea and watermelon (Inobeme et al., 2014)(Anthony and Egbuonu 2015). This evidently shows that Chrysophyllum albidum seed has a high mineral content. Crude fat was found to be 25.58±0.51%. This is quite high. Seeds with high lipid content are usually compared with those of soybean, locust bean and cotton seeds. These are commercially exploited and are classified as oil seeds (Ayodele et al., 2000). This makes *Chrysophyllum albidum* seed a good source of lipid for both commercial and industrial use. The fibre content of the seed was found to be 16.00±0.13%. This value is higher than 2.37±0.00 for both locust bean seed and water melon seed (Anthony and Egbuonu 2015) (Inobeme et al., 2015). Fibre helps in the maintenance of human health and has been known to reduce cholesterol level in the blood. The protein content was found to be 25.76±0.45%. This value is higher than 21.46±0.04 for cowpea (Inobeme et al., 2000) and 21.46±0.04 for watermelon seed (Inobeme et al., 2000). The results however, suggest that *Chrysophyllum albidum* seed could be used as an alternative source of protein supplement. More so, carbohydrate was found to be 28.66±0.01%. This is lower than 78.49±0.04 for *Chrysophyllum cainato*(Oranusi et al., 2015). However, this seed is not a good source of carbohydrate.

The ascorbic acid content was found to be 5.16% as shown in Table 1. This value is relatively low compared to 66.88% for *Lagenaria* seed (Chinyere et at., 2009). However, this acid has medical, biological and physiological uses. Medically, the fact that it is an anti-scurvy acid its supplementation reduces the risk of myocardial infarction, stroke,

cardiovascular mortality, or all- cause mortality (Ye et al., 2013). Biologically, vitamin C is essential to a healthy diet as well as being a highly effective in humans, acting to minimize oxidative stress; a substrate for ascorbate peroxidase in plants (Higdon, 2006). Physiologically, Ascorbic acid acts as an enzymatic cofactor. It helps in synthesis of collagen, carnitine and neurotransmitters; the synthesis and metabolism of tyrosine; and the metabolism of microsome (Odoemelam, 2005). Ascorbic acid acts as a proton donor to eight different enzymes (Gropper et al., 2005).

Elemental analyses were carried out on the ash content after digestion and dilution using flame photometer and atomic absorption spectrophotometer. The result of the mineral content is presented in table 1. From the result obtained, the value of Iron was found to be 33.20±0.12mg/g. This value is closer to 50.67mg/kg for Locust bean seed (Olujibo, 2012) and higher than 2.73mg/g for watermelon seed (Tabiri et al., 2016). Iron is needed for red blood cell formation. An insufficiency of iron compounds in the body shows itself as iron-deficiency anaemia. Calcium was found to be 18.04±0.05mg/g. This value is lower than 1.47033g/kg for locust bean seed (Olujibo, 2012).

Magnesium was found to be 0.16±0.01mg/g. This value is lower than 30.60mg/kg reported for locust bean seed (Olujibo, 2012). Potassium was found to be 2.50±0.02mg/g. This is higher than 0.22mg/g reported for watermelon [Tabiri et al., 2016]. Increasing potassium intake through fruits and vegetables has been associated with an improvement in healthy bone formation in children, adults and the elderly. Evidence from variety of studies has demonstrated that increased potassium intake reduces systolic (maximum) and diastolic (minimum) blood pressure in adults in both hypertensive and non-hypertensive patients (Sarit, 2013). The value of Zinc was found to be 34.00±0.14mg/g. This value is higher than 0.81mg/g for watermelon (Tabiri et al., 2012). Zinc is an essential trace mineral. According to (Deshphande et al., 2013), "Zinc is required for the metabolic activity of 300 of the body enzymes, and is considered essential for cell mitosis and meiosis and synthesis of DNA and protein. These enzymes are involved in the metabolism of protein, carbohydrate and fat. Zinc is also critical to tissue growth and maintenance, immune system function, prostaglandin production, bone mineralization, proper thyroid function, blood clothing, cognitive functions, foetal growth and sperm production". The value of Phosphorus was found to be 9.80±0.10mg/g. This value is higher than 0.22mg/g reported for watermelon seed (Tabiri et al., 2016). Phosphorus is one mineral that perfumes a number of important functions. It combines with Calcium to form a relatively soluble compound calcium phosphate, which gives strength and rigidity to bones and teeth. Phosphorus like calcium is needed not only for growth of skeleton but also for its maintenance. Sodium was found to be 0.05±0.02mg/g. This is lower than 0.08mg/g for watermelon (Tabiri et al., 2016).

6. Conclusion

Chrysophyllum albidum seed contains a high amount of lipid, fibre, protein, iron, zinc, phosphorus and calcium as shown on the table for comparison above. These are essential body materials. This makes the seeds a good source of minerals, protein, fat and oil. The ascorbic acid content of the seed which is present in low concentration is however significant and can be used for vitamin c supplement. The result of the proximate and elemental analysis is supportive of the high nutritional value of the seeds. Thus, it is suggested that the seed can be processed as a component of animal feed stock for poultry, which is undergoing experimental examination and the result shall be published in the next phase of this work.

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Annexure

Calculations

Determination of Moisture Content: The moisture content is calculated thus:

% moisture = $\frac{loseinweightondrying}{initialsampleweight} \times \frac{100}{1}$

Weight of empty beaker (a_1) = 24.4g

Weight of empty beaker + sample (a_2) = 29.4g

Weight of empty beaker + dry sample (a_3) = 25.1g

% moisture content = $\frac{a_3 - a_1}{a_2 - a_1} \times \frac{100}{1}$

$= \frac{25.1 - 24.4}{29.4 - 24.4} \times \frac{100}{1} = \frac{0.7}{5} \times \frac{100}{1} = 0.14 \times 100 = 14\%$

Determination of Ash Content: The ash content is calculated as follows:

Weight of empty beaker (b_1) = 30.1g

Weight of empty beaker + sample (b_2) = 35.1g

Weight of empty beaker + dry ignited sample (b_3) = 30.3g

% Ash = $\frac{b_3 - b_1}{b_2 - b_1} \times \frac{100}{1}$

$= \frac{30.3 - 30.1}{35.1 - 30.1} \times \frac{100}{1} = \frac{0.2}{5} \times \frac{100}{1} = 0.04 \times 100 = 4\%$

Determination of Crude Fat Content: The percentage crude fat was calculated as follows;

Weight of empty round bottom flask (c_1) = 24.1g

Weight of round bottom flask + sample (c_2) = 29.1g

Weight of round bottom flask + (dry fat), lipid (c_3) = 25.2g

% crude fat = $\frac{c_3 - c_1}{c_2 - c_1} \times \frac{100}{1}$

$= \frac{25.2 - 24.1}{29.1 - 24.1} \times \frac{100}{1} = \frac{1.1}{4.3} \times \frac{100}{1} = 0.2558 \times 100 = 25.58\%$

Determination of Crude Fibre Content: The percentage crude fibre content is calculated thus;

Weight of empty beaker (d_1) = 97.7g

Weight of empty beaker (d_2) = 102.7g

Weight of empty beaker + dry fibre (d_3) = 98.5g

% crude fibre = $\frac{d_3 - d_1}{d_2 - d_1} \times \frac{100}{1}$

$= \frac{98.5 - 97.7}{102.7 - 97.7} \times \frac{100}{1} = \frac{0.8}{5} \times \frac{100}{1} = 0.16 \times 100 = 16\%$

Determination of Carbohydrate Content: The total carbohydrate content is calculated as shown below:

Protein = 25.76

Fat = 25.58

Fibre = 16.00

Ash = 4.00

71.38

% carbohydrate content= 100 – 71.38= 28.66%

Determination of Crude Protein Content: The crude protein content obtained from the analysis is calculated thus;

% Nitrogen = $\frac{mlofHCl(sample) - mlofHCl(blank) \times molarityofHCl \times 100 \times 100 \times 14}{weightofsample \times mlofdigest \times 1000}$

% Protein = % Nitrogen × Protein factor = 4.121 × 6.25 = 25.76%

Determination of Ascorbic Acid Content: The ascorbic acid (Vitamin C) content is given below;

Burette Readings	1st (cm^3)	2nd (cm^3)	3rd (cm^3)
Final	5.80	25.70	43.60
Initial	0.00	20.40	37.90
Volume of iodine used	5.80	5.30	5.70

Average Volume of iodine used = $\frac{(5.80 + 5.30 + 5.70)}{3} cm^3 = \frac{16.8}{3} cm^3 = 5.60 cm^3$

Volume of blank titre = 18.5cm^3

Volume of iodine used = 5.60cm^3

Standard Ascorbic Acid in 10ml of H_2O = 0.20g/l

Weight of Sample = 5g

% Ascorbic acid = $\frac{18.5 - 5.60 \times 0.20 \times 100}{10 \times 5} = \frac{258}{50} = 5.16\%$