Determination of Proximate and Mineral Elements Compositions in the Bark of Dacryodes edulis (G.Don) H.J. Lam

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Abstract

Determination of proximate, mineral elements compositions, total antioxidant capacity and total phenolic contents of the bark of Dacryodes edulis (G. Don) H.J. Lam was carried out. Proximate study revealed that the bark is adequate in nutrients and minerals. Macro nutrient like protein Carbohydrates was determined with a content level of 58±0.16mg/kg. Other macro nutrients were also relatively high in content level. While crude Protein had a content level of 61.73±0.11mg/kg, crude Lipid had a content level of 48.56±0.20mg/kg. Proximate analysis for crude Fibre, Vitamin C, Ash, and Moisture also revealed their contents levels to be 61.66±0.92mg/kg, 24.01±0.16mg/kg, 52.37±0.38mg/kg and 14.56±0.18 mg/kg, respectively. The existence of essential minerals and their contents levels analysed showed that Dacryodes edulis contains sodium (Na; 7.20±0.04) mg/kg, potassium (K; 2.57±0.25) mg/kg and Iron (Fe; 17.76±0.02) mg/kg, calcium (Ca; 83.24±0.89)mg/kg, Zinc (Zn; 3.65±0.08)mg/kg, and manganese (Mn; 241.28±) mg/kg, respectively, values range that are non-lethal, therapeutic and idea for the maintenance of good health except for Manganese that was on the very high side. Dacryodes edulis bark was found to be also very rich in anti-oxidants and phenols, with total anti-oxidant capacity of 103.02±0.62mgVce and phenol (total) content of 112.95±0.56GAE100-1 respectively.

Keywords: Promixate, Mineral elements, Total antioxidant capacity, Total Phenolic content, Dacryodes edulis.

Introduction

The contribution of bark of tree and its constituents to medicinal ethno pharmaceutical are laudable. This is because it serves many medicinal purposes in major parts of Africa. *Dacryodes edulis* (G. Don) H.J Lam, the African pear, is fir fruit common to the gulf of guinea region, Westand Central Africa¹. The generic name *Dacryodes* was obtained from the Greek word '*Dakruon*', meaning '*tear*', which refers to resin droplets on the bark surface of its member, while '*Eduli*' meaning '*edible*', emphasizes the tremendous interest in the cultivation of this nutrients fruit plant^{2,3}. It is a tree species with a very notable short trunk and a cavernous dense crown. It reaches a height of about 18-40m in the forest but not go beyond 12m in plantations⁴.

Dacryodes edulis ideally does well in a dim, moist, tropical forest in some major parts of Africa. However, it burgeons adaptively to digression in temperature, soil type, and humidity. The universal existence extends from Sierra Leone in the West, Uganda in the east Angola in the south, Nigeria in the North. Malaysia also cultivates this fruit tree species⁵. Apart from this plant bearing edible fruits, the leaves, stems, roots and bark are been used as medicine for the treatment of some diseases^{4,6-8}. The plant's bark is ashy grey in colour and coarse with droplets of resin^{9,10}. While extract from the tree bark are used to remedy

problems of dysentery, mouthwash, for tonsillitis, and general oral hygiene¹¹⁻¹³ extracts from the root and bark are used in the treatment of leprosy and malaria. The powdered form obtained from the bark of *Dacryodes is* added with malequeta pepper as an anti-dysenteric for anaemia¹⁴. Its wood finds usefulness incarpentry, shelter, road and many traditional medicines for when the bark is grounded and mixed with palm kernel oil, it becomes a potent ointment for healing injuries⁴. The leaves are compound with 5-8 pairs of leaflets in a stalk. The fleshy mesocap of the fruit is dark blue or violet with a very big seed which is used directly for cultivation of the African pear tree. Phytochemical screening of the fruit, seed and leaves have shown that alkaloids, tannins, flavonoids, saponins, glycosides (cyanogenic), and phytatesare present^{8,16,23}.

Phytochemical compounds play very significant defensive roles in plants. They protect plant cells from environmental drought, stress, pathogenic attack, pollution, and UV exposure. These compounds are also referred to as secondary metabolites and have various biological properties which make many people in Nigeria use the various plants for medicinal purposes⁸. Generally, the minerals contained in the plant play some specific functions in plants and animals which enable them to function properly and maintain wellbeing. This study, therefore, aims to determine the bioactive components responsible for the

medicinal properties of *D.edulis* bark. Thus the major objective of this study toevaluate the proximate, mineral composition, total antioxidant capacity and content of phenol (total) of the bark of *D. edulis* (G.Don) H.J. Lam.

Materials and Methods

Reagents: All reagents used in this research were of analytical grades with high purity.

Collection of Samples: Triplicate samples of *Dacryodes edulis* bark were obtainedfrom an urban area in Benin-City (6⁰ 18.1510^IN and 5⁰37.2508^IE), Edo state, Nigeria. Benin-city lies within the tropical rainforest of Nigeria in the Western of Africa. Samples obtained from the tree barkwere dried in air under room temperature for a period of one month at Moist Forest Research Station, Benin-City.

Processing of plant samples: The bark samples were granulated using a ceramic mortar and pestle into a powdered form and stored in covered containers for further analysis.

Proximate Composition Determinations: Moisture content determination: 2g of powdered *Dacryodes edulis* bark were put in a ceramic crucible and heated at 105°C to achieve a constant weight. Following this, the moisture content was obtained, being loss in weight of the original sample, which waslatter expressed as percentage moisture content¹⁷.

Crude protein determination: This was obtained following the Kjeldahl method though with slight variation. This involves digesting 0.5g of the samples with 5ml of concentrated H₂SO₄ in using Kjeldahl catalyst. Samplesnitrogen inherent in protein was converted to ammonium sulphate. This was introduced to 2.5 ml Brucine reagent (2.5%), followed by H₂SO₄ (98%, 5ml). And a clouded subtratewas formed whose absorbance at wavelength 470 nm was determined. This was followed by calculating the percentage nitrogen, times 6.25 to getthe crude protein value¹⁸.

Crude lipid determination: This was achieved through the method of Soxhlet extraction. 10g of the powdered bark sample and wrapped in a watzmanfilter paper and carefully positioned in a thimble. The thimble was cotton-wool laced and thereafter placed in the extraction column linked to a condenser. This was followed by extracting the lipid using $n - \text{Hexane} (200 \text{ ml})^3$.

Ash content determination: This involves transferring 2 g of the pulverized samples to a crucible and ignited at 500° C for 6 hours. This was followed by cooling in a desiccator and weighed under room temperature to get its ash content.

Carbohydrate determination: This was carried out by subtracting the total of moisture, protein, lipid, fiber and the contents of ash from 100^{19} .

Crude fiber determination: This involves heating 5g of the

powdered bark samplewith 200 ml of H_2SO_4 (1.25%) for a period of thirty minutes and filtered (Buchner funnel). The residue obtained was distilled water washed to be free of acid. This was followed by boiling the residue in 200 ml NaOH (1.25%) for about 30 minutes. It was then filtered again and washed free of alkaline with distilled water. The substrate was rinsed with HCl (10%) first and then and with ethanol (twice). Finally it was rinsed with petroleum ether three times. The residue was dried in the ovenat 105° C overnight. After cooling in a desiccator, it was ignited in a muffle furnace at 550° C for ninety minutes which enabled the weight of the crude fibre to be obtained 18.

Minerals Determination: Some Minerals contents of *D. edulis* were determine using atomic absorption spectrometry, flame photometry and spectrophotometry methods¹⁸.

Wet digestion of sample: For wet digestion of the sample, exactly (1g) was introduced into a digesting glass tube. This was followed by the addition of 12ml of HNO₃ to the samples and mixture and kept overnight under room temperature. 4 ml HClO was then added to this mixture and was kept in the fumes chamber for digestion. The temperature was gradually increased, from 50°C and increasing up to 250-300°C. The digestion took about 70- 85 minutes with the appearance of white fumes as an indicator. The mixture was allowed to cool down and the contents of the tubes were introduced into volumetric flasks (100 ml) where the volumes of the contents were made to 100 ml with distilled water.

The digested solution (wet) was thereafter transferred to plastic bottles and labeled appropriately. The stored digest were used for mineral determination.

Sodium (Na) and Potassium (K) determination via flame photometer Principle: The flame photometer measures the emission of radiant energy when atoms of an element return to their ground state after their excitation by the high temperature of the flame. The degree of emission is associated to the concentration of the element in the solution.

Procedure: Analysis of Na and K were carried out via flame photometry method.

Determination of Iron (Fe), Calcium (C), Zinc (Zn) and Manganese (Mn): The same wet digested dry *Dacryodes edulis* bark sample solutions was analyzed for the assay of Iron content, Calcium, Zinc and Manganese using the Atomic absorption spectrophotometer with Model Agilent55B. Total antioxidant capacity determination: Phospho-molybdenium methodin line with the procedure described by Prieto P., Pineda M. and Angulilar M.²⁰.

Total Phenolic content determination: The Phenol (Total) content were determined via Folin-ciocalteu spectrophotometric method³.

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Results and Discussion

Table-1 shows the presence and contents levels of carbohydrates, crude protein, crude lipid, moisture, Vitamin c, Ash, and crude Fibre that were determined from the bark of D.edulis and their values were reported in triplicate together with their % mean ± SD inclusive, as each experiment was repeated 3 times. Table1 also contained the triplicate results of the minerals, such as, Sodium (Na), Iron (Fe), Manganese (Mn), Calcium (Ca), Potassium (K), Zinc (Zn), Total Anti-oxidant capacity (TA-OC), and Total Phenolic Content (TPC) including their % mean \pm SD.

The proximate analysis carried out in this study showed that adequate nutrients composition is inherent in the bark of D. edulis (G.Don). The levels of the nutrients determined were generally high, as some (e.g. Carbohydrates, crude Lipid and crude Protein) were within reference standards, others (e.g.Ash, crude Fibre and crude Protein) were above. Results from Table

1 showed crude protein having the highest value of 61.73±0.11 mg/kg, which was closely followed by crude Fibre (61.66±0.92 mg/kg), Carbohydrates (58.51±0.16 mg/kg), Ash (52.37±0.16 mg/kg), crude Lipid (48.56±0.20 mg/kg), Vitamin C (24.01±0.16 mg/kg) and a Moisture content of 14.56±0.18 mg/kg. The presence of Vitamin C as part of the proximate composition of the bark of Dacryodes edulis underpins and enhances its interest in medicinal or health purpose. Vitamin C helps the body to fight against illnesses by stabilising the immune system cells. This is an attribute that could have made Dacryodes edulis to standout amongst others in terms of medicinal value. Vitamin C also known as ascorbic acid, whose determined content level was 24.01±0.16 mg/kg, though appearing relatively low, but it is of a therapeutic level needed by the body, as overdose of ascorbic acid (arising from high content level) endangers the kidneys. This is because, a very high level of Vitamin C leads to the acidification of the urine, which causes kidney stones.

Table-1 Results for proximate analysis on powdered dry weight basis of the bark of Dacryodes edulis, some mineral concentrations, Total antioxidant capacity and Total phenolic content

Parameters	1st	2nd	3rd	Mean/Standard Deviation
Carbohydrates	58.42	58.70	58.41	58.51 <u>+</u> 0.16 mg/kg
Crude Protein	61.84	61.73	61.63	61.73 <u>+</u> 0.11 mg/kg
Crude Lipid	48.42	48.70	48.47	48.56 <u>+</u> 0.20 mg/kg
Moisture	14.36	14.63	14.60	14.56 <u>+</u> 0.18 mg/kg
Vitamin C	23.94	23.90	24.20	24.01 <u>+</u> 0.16 mg/kg
Ash	52.11	52.21	52.81	52.37 <u>+</u> 0.38 mg/kg
Crude Fibre	61.10	61.17	62.73	61.66 <u>+</u> 0.92 mg/kg
Sodium	7.16	7.21	7.24	7.20±0.04 mg/kg
Potassium	2.57	2.58	2.59	2.57±0.25 mg/kg
Iron	17.75	17.78	17.76	17.76±0.02 mg/kg
Calcium	82.43	84.20	83.10	83.24 <u>+</u> 0.89 mg/kg
Zinc	3.74	3.61	3.59	3.65 <u>+</u> 0.08 mg/kg
Manganese	241.35	242.40	240.09	241.28 <u>+</u> 1.16 mg/kg
Total Antioxidant Capacity	103.41	102.30	103.34	103.02±0.62 mg/Vce
Total Phenolic Content	113.56	112.47	112.84	112.95±0.56 GAE100 ⁻¹

Dacryodes edulis was also found to contain essential minerals, such as Na (7.20±0.04 mg/kg), K (2.57±0.25 mg/kg), Fe (17.76±0.25 mg/kg), Ca (83.24±0.89), Zn (3.65±0.08), and Mn (241.28±1.16) which are needed by living cells. Distorted enzymatic activities and poor electrolyte balance of blood fluid are recognized to be related to inadequate Na, K, Zn etc. The relatively low level of sodium determined is nutritionally ideal for both hypertensive and non-hypertensive patients, as high blood pressure are associated with high level of sodium intake²¹. Potassium being one of the essential minerals is needed to keep water at cell level balanced, ensureappropriate body pH, and help in protein and carbohydrate metabolism²⁵.

It is also recognized that increase intake of potassium reduces blood pressure to about 3.2 mmHg, thus reducing mortality by over 7%. However, high consumption of food very high in potassium level could trigger irregular blood pressure, nausea, or slow pulse²⁶.

It is interesting to note that the potassium level as determined in this study was not on the high rather, low $(2.57\pm0.25~\text{mg/kg})$ and of the rapeutic level. Iron is a vital mineral in the formation of hemoglobin and myoglobin, which function in oxygentransport²³.

The relatively low level of Iron determined (17.76±0.02 mg/kg) alleys the fears of developing possible liver failure due to overload of Iron in the bodyon consumption/usage of D.edulisbark. These essential minerals are good reducing agents with anti-oxidant characteristics that fight against terminal diseases like cancer. Calcium is endeared as a key player in bone formation and other physiologic systems in the body. It's a mineral that is strongly linked to many of the functions that Vitamin D plays in the body. Besides, it's essential for blood clothing, stabilisation of blood pressure and a vital mineral for communicating essential information among cells, as well as, contributing to normal brain function. It is the mineral that is found to be most copious in the human body. Hence it is recommended to be eaten constantly to build bone and maintain the blood level of calcium, which the bark of *D.edulis* is richly endowed (83.24±0.89 mg/kg).

The very low level of Zinc determined, relatively [almost] in trace concentration (3.65±0.08 mg/kg) is vital for the physiologic functions of living tissues and, regulation of many biochemical process in the body⁴. Manganese, being a trace element present in tiny amounts in our body helps in sex hormones formation, and that of connective tissue, bones and blood clothing factors in the body. Besides, it also plays role in carbohydrates and fat metabolisms and other physiologic functions. However, excess manganese can interfere with the absorption of dietary iron, thereby resulting in iron-deficiency anemia on a long term usage. Manganese determined in this study was on the very high (241.28±1.16 mg/kg). Total Antioxidant Capacity in *Dacryodes edulis* as determined was high

(103.02±0.62mgVCE). The most important role of anti-oxidants is to neutralise the effect of free radicals which damage health cells. It also strengthensthe immune system and prevent diseases (e.g. cardiovascular disease and cancer), thus imparting good health and longevity. Phenols help in providing the body defence against oxidative stress or damage arising from oxidising agents and free radicals, particularly when the internal enzymatic mechanisms fail or become inadequate^{24,25}. Besides, they are known for their excellent ability to prevent fatty acids from oxidative decay²⁶. The Total Phenolic Content in Dacryodes edulis as determined was quite $(112.95\pm0.56\text{GAE }100^{-1}).$

Conclusion

Determination of the proximate, mineral elements compositions, total antioxidant capacity and content of phenol (total)of the bark of *D.edulis* (G.Don) H.J. Lam showed that it is nutritionally adequate in proximate compounds of Carbohydrates, Protein, Lipids and Vitamin C, and not lacking in essential food fibre and moisture as well as, essential minerals of sodium, potassium, iron, calcium, and zinc in contents levels needed for a healthy life, except manganese, with a very high content level, which calls for a control use of any decoction from the bark of *Dacryodes edulis* (G.Don) H.J. Lam.

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