The Phytochemical and Proximate Composition of White Mangrove Leaves (*Laguncularia Racemosa*)

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ABSTRACT

The mangrove leave (Laguncularia racemosa) also known as white mangrove obtained at the deep forest areas of south- south region of Nigeria is one of the most productive and biologically important plant resources that contribute to the community structure of West African mangrove forests and breeding place for marine species, and as medicine and food for coastal dwellers. However, its phytonutients are yet to be evaluated. This study is to investigate phytochemical profile and proximate nutrient compositions of Laguncularia racemosa leaves. From the analysis, the phytochemical profile revealed the followings; Alkaloid 6.01 %, Saponin20.23%, Tannin 65.21%. Flavonoid 7.45% and phytate recorded 1.10%. These values are moderate when compared with other plants analyzed in Nigeria indicating that the plant can serves as alternative to antibiotics (Drugs) in livestock industries due to its pharmacological properties particularly now that pathogens are building more resistance to drugs. Data obtained for proximate composition shows crude fibre recording 30.87%, carbohydrates 23.001% and protein 9.725%. This finding indicates that the analyzed leaves can be implored as alternative feed ingredient in animals especially ruminants and non-ruminants.

KEYWORDS:

Laguncularia Racemosa Leaves, Phyto-chemicals, proximate composition.

I. INTRODUCTION

Approximately one-fourth of the world's tropical coastline is dominated by mangroves plants and they extend over 15.5 million hectares worldwide (Macinntosh and Zisman,1997) The most extensive and luxurious mangroves extend across the indo-pacific regions, where they are best developed in the delta system of major rivers.

The largest single area of mangroves plants in the world lays in the Bangladesh part of sunder bands covering an area almost 600,000 hectares including waterways. There are about 6.9 million hectares in the indo – pacific region (Cloug.1993; Macintosh and Zisman.*1997*), 3.5 million hectares in Africa with good proportion in the southern Nigeria and 4.1 hectares in the American including the Caribbean. (Untawale, *et al.*1992; Zahran and Al- kaf.1996; Macintosh and Zisman.1997). Mangroves plants also penetrate some temperate zones, but there is a rapid decrease in the number of species with increasing latitude (Macintosh and Zisman.1997)

A chemical and pharmacological survey of mangrove plants in the Australia region revealed that several species of mangroves plants leaves possess antiviral activity and healing properties in popular/folk medicine that are attributed to *Rhizophora* trees (Red mangrove) (Bandaranayake,2002). Similarly the root, leaf and stem extracts of *Rhizophora* trees have inhibitory properties affecting the growth of various human pathogenic organisms and among these are bacteria, fungi and viruses, (Bandaranayake,2002).It has been reported that mangrove plant cured throat cancer with gargles of mangrove bark, (Lesile Tailor,2003). Bark of red mangrove trees have been used in folk remedy for a wide array of diseases, (Marius.1985).More recently, (Alarcon-Aguilara et al.,1998) reported that extracts of *Rhizophora mangle* (Black mangrove) had anti-diabetic and anti-hyperglycemic properties. (Itigowa et al., 2001) asserted that *Avicennia plants,* especially *Laguncularia racemosa leaves* are used in traditional medicine that might serve as lead for the development of novel drugs.

The influence of mangroves trees on reproductive health and their performance enhancement attributes in human and animal has been reported by (Lesile Tailor, 2003) to be due to the following phytochemicals: Alkaloids, Lignins, Flavonoids, Lipids, Benzernoids, Steroids, Alkanes, Tanin and Saponins. The use of such phytochemical extracts by herbalists to improve the reproductive hormones and the overall performance of animals and man was associated to its phytochemical (aphrodisiac) properties as reported by (Sofowora.1993; Amin.et al. 1996; Yakubu et al., 2003; Ratnasooriya and Dharmasiri. 2000)

The results of proximate composition and phytochemical analysis of *Laguncularia racemosa* leaves may likely play the role of alternative antibiotic growth promoters that are indigenous and medicinal plants known to possess a wide range of antibacterial and antifungal properties. They could be employed as feed additive in animal dietary manipulation.

Amongst the numerous documentations of the trado-medical application of *Laguncularia racemosa,* little has been reported on its medicinal (Ethno-veterinary) influence on the reproductive physiology and growth performance of animals and man. Biologically, the relevance of its phytochemicals on general health improvements still remains obscured. Therefore, the objective of this study is to un-earth phytochemicals and nutritional profile of *Laguncularia racemosa* to tap from its potential medicinal (ethno-veterinary) and the nutritional value for the benefits of animals and mankind.

II. MATERIALS AND METHODS

Whole fresh leaves of *Laguncularia racemosa* leaves were harvested fresh from the Eagle Island, Port Harcourt in Rivers state of Nigeria. They were oven dried at 78^0C for two hours in accordance with the methods of (Wekhe and Oboh, 2007). Thereafter, proximate composition and phytochemicals analysis were carried out in the Department of Food Science and Technology, Rivers State University of Science and Technology Port Harcourt, Rivers state. The leaves were milled into powdery form using local grinding machine in the nearby market closed to the University and was finally stored in a clean covered container to prevent microbial contamination and spoilage of the products.

Moisture Content

The method of *(AOAC.1990*) was used to determine the moisture and Ash contents of *Laguncularia racemosa* leaves. 5kg of the ground sample was weighed into previously heated, clean and dry aluminum dish. The dish and the content were then placed in an air oven for one hour at a temperature of 110^0C. The dish was removed from the oven, cooled in a desiccators and re-weighed. The percentage moisture content of the sample was calculated from the weight loss.

$$\text{Moisture (\%)} = \frac{Loss\ of\ weight}{wt\ of\ sample} x\ 100$$

Ash content

The method of the (*AOAC.1990*) was also used. 5g sample was weighed into a previously ignited and clean porcelain crucible (Dish). The crucible and the content were then transferred to a muffle furnace and allowed to ash for one hour at 500^0C. At the end of the exercise, the crucible with its content was removed from the furnace and cooled in a desiccators and weighed again. The percentage ash content of the sample was then calculated as follows;

$$\text{Ash (\%)} = \frac{weight\ of\ ash}{weight\ of\ sample} x\ 100$$

Crude Fat

The crude fat was determined using the Sechelt extraction method (micro extraction unit) 5g sample was weighed after moisture determination onto a Whitman number 1 filter paper. This was then placed on the extraction unit and extracted for three hours using petroleum ether as solvent. At the end of the extraction process, the ether was evaporated and the weight of the extraction flask taken. The difference in weight before and after extraction was recorded as the amount of the fat or ether extract.

$$\text{Crude Fat (\%)} = \frac{wt\ of\ ether\ extract}{wt\ of\ sample} x\ 100$$

Crude Protein

The method of the Association of Official Analytical Chemist Washington D.C. (1990*)* was used. 2g of the sample was weighed into a 100ml conical flask and were added one and a half tablet of kjedahl catalyst and 10ml of Nitrogen-free concentrated sulphuric acid. The mixture was heated slowly for digestion in a fume cupboard with the flask placed at an angle of 40^0 for 30 minutes. Heating was then increased and continued until frothing ceased. The sample was allowed to cool and then transferred into 100ml volumetric flask and made to volume with

distilled water. 10ml of the digest was introduced into 100ml Kjedahl distillation flask and 10ml of 45% NAOH was added. The ammonia liberated was steam distilled into a 5ml boric acid in a conical flask until 50ml of the distillate was obtained. This was back titrated against 0.05N H_2SO_4 to give the nitrogen content of the sample. A blank determination was also carried out and subtracted from the sample reading and the %N was calculated thus:

$$N(\%) = \frac{(Titre - Blank)\ x\ Normally\ of\ \ acid\ x 1.4}{Weight\ of\ \ sample}\ x\ 100$$

The percentage crude protein content of the sample was then calculated thus: % crude protein = % N x 6.25

Total Available Carbohydrate

Manual Anthrone method of (Osborne and Voogt 1978) was used in the determination of total available carbohydrate of the milled samples, 2.5g of the milled sample was digested using 13ml of 52% per hydrochloric acid (diluted with water in the ratio of 270ml: 100ml). 1ml of the digest was pipetted into a test tube and 5ml of freshly prepared Anthrone Reagent was added, mixed and allowed to stand in a boiling water bath for exactly 12 minutes. The test tube and its content were then removed and cooled quickly to room temperature. The absorbance of the samples mixture and standard were then read at 630nm against the reagent blanks, and the total available carbohydrate content was then calculated thus:

$$\text{Total available carbohydrate (as \% glucose)} = \frac{25\ x\ b}{a\ x\ w}$$

Determination of Crude Fibre

5g of the moisture free sample was extracted for three hours with petroleum ether using a sox let apparatus. The fat free material was placed in a 200ml beaker and 50ml of 1.25%w/v sulphuric acid was added and covered with a wash glass. The content of the beaker was heated gently on a hot plate for 30 minutes (acid hydrolysis). After acid hydrolysis, the content of the beaker was filtered under vacuum through a Buchner funnel fitted with filter paper and washed with boiling water until the washing was no longer acid to litmus.

The residue was washed back into the original flask using a wash bottle containing 1.25% NAOH. This was boiled for 30 minutes covered with a wash glass. The resulting insoluble material was transferred to a dried weighed Ash less filter paper and washed thoroughly first with hot water and then with 15ml of Ethanol (95%) by volume. The filter paper was dried at 100^0C to a constant weight for one hour. The filter paper and content was incinerated to an ash at 500^0C for one hour. The ash was allowed to cool and then weighed.

The weight of the ash was subtracted from the increase of weight on the paper due to the insoluble material and the difference reported as fibre.

$$\text{Crude Fibre (\%)} = \frac{wt\ of\ \ fibre}{wt\ of\ sample}\ x100$$

Analysis of the following Phytochemicals;

(a) Taninn
(b) Alkaloid
(c) Saponin
(d) Flavonoid
(e) Phytate

Tannin – The method for the determination of Tannin was by method of Mega as described by (Akinmutimi. 2006)

Alkaloid – Alkaloid was determined using (Harborne method. 1998). **Saponin** – The method used was that of (Obadoni and Ochuko. 2001). **Flavonoid** – Flavonoid determination was by the method of (Bohm and Koclpal _ Abayazan.1994)

Phytate- lucus and Markakas method as described by (Akinmutimi.2006)

III. RESULTS AND DISCUSSION

The results of the phytochemicals analysis of the forest plant (Table1) revealed that it contains favorable phytochemicals of Tannin, Saponin, Alkaloid, Flavonoid and Phytate. The findings in this study agreed with the work of (Bandaranyake, 1998a and 1998b*)* who reported that, mangrove plants are rich sources of steroids, triterpens, saponins, flavonoids, alkaloids and tannins.

(Wekhe, 2002) reported that alkaloids are used as antiparasites, antispasmodic and bacterial antigens. (Ahamefule et al., 2006) submitted that flavonoid and alkaloid present in some mangrove plants such as *Laguncularia racemosa* function in protection against inflammation, allergies, and microbial infestations. (Kawo, 2009) reported the pharmacological activities of phytochemicals in plants to include antimicrobial, inflammation inhibiting and cytotoxic activities. Thus livestock farmers are encouraged to use this mangrove plant due to its pharmacological benefits thereby saving funds meant for drugs (antibiotics), since most of this phytochemicals are present in this analyzed *Laguncularia racemosa leaves*.

(Bandaranayake, 2002 *and* Lesile*, 2003)* reviewed the role of flavonoids, alkaloid and saponin as therapeutic agents and have implicated the flavonoids components in forest plants such as mangrove leaves in enhancing aphrodisiac properties and indirectly influencing the production of estrogen/testosterone in animals and Man. Animals that consumes this forest leaves are likely to experience high libido and significant increase in reproductive performance. Flavonoid is the most common widely distributed groups of phytochemicals in forest plants (phenolics) as reported by (Ahamefule *et al*., 2006). Its biological function includes protection against allegies, platelet aggregation, microbes, ulcer and tumors, he further stated that several biological activities such as cytotoxic, anti-neoplastic, antibacterial, ant herpetic, steroidal, and anthelminthic are reported to influence defense against invading parasites. Therefore, it's in line to state that the use of this test mangrove leaves in animal feeds manipulation will assist in improving reproductive efficiency and also help in enhancing body immunity against the possible various diseases infestations.

Table 1; Phytochemicals present in Laguncularia Racemosa

Tanin %	**65.21**
Alkaloid %	**6.01**
Saponin %	**20.23**
Flavonoid %	**7.45**
Phytate %	**1.10**

Table 2; proximate compositions of Laguncularia Racemosa

Ash%	**10.70**
Moisture %	**20.00**
Protein %	**9.725**
CHO %	**23.001**
Lipid %	**5.70**
Fibre %	**30.874**

The obtained results of this phytochemical analysis also lend credence to the finding of (Akindahunsi and Salawu, 2005) on Bidens Pilosa leaves (Abere oloko). He reported that the phytochemicals in leaves of Bidens Pilosa exhibit cytotoxic effect and growth inhibition against abnormal cells and have inflammatory and anticancer properties. They also show tumor inhibiting activity in animals. Therefore, the leaves of *Laguncularia racemosa* could be used as an alternative to antibiotics in livestock industries.

The results of the proximate composition of *Laguncularia racemosa* leaves in Table2 revealed low protein content of 9.725% when compared with *Amaranthus caudotus* 20.59%, (Etuk *et al*.,1998). *Piper Guinese* 29.78%, *Talinum triangular* 31.00%. (Akindahunsi and Salawu 2005) The values of Ash content of 10.70% recorded in this study requires further investigation to ascertain the types of minerals available as they are necessary in body metabolism, functions and maintenance. The Ash value is higher than *Occimum graticimum* 8.00% and *Hibiscus esculentus* 8.00%. (Akindahunsi and Salawu 2005) The high Ash content in this test leaves is a reflection of the minerals untapped deposit in the forest plant. The values of crude Fat 5.70% content was moderate when compares with those of *Talinum triangulare 5.90%, Baseila alba8.71%, as reported by* (Akindahunsi and Salawu 2005) and *Bidens Pilosa leaves contain 7.49% crude fat* (Alikwe *et al*., 2013).Moderate fat content assist in absorption of fat soluble vitamins in diet and serves as additional source of energy. However, excess fat intake is associated with cardiovascular disorder such as atherosclerosis and cancer, (Antia *et al*.2006) The results on crude fibre content of 30.00% was higher when compares with the work of (Akindahunsi and Salawu 2005) on *Talinum triangulare* 6.20%, *Piper guineeses* 6.40%, and bitter leaves *vernonia amygdalina,* 6.5%and *Bidens pilosa leaves*18.13% (Alikwe et al.,2013). Forest plants and non-starchy vegetables are the richest sources of dietary fibre, (Agostoni *et al.*1995) They are essentials in the cure of obesity, diabetes and gastrointestinal disorders (Saldanha,1995) The nutrient contents in this leaves indicates that its can comfortably be used in feed manipulation in animals especially the Ruminants and Non-Ruminants

IV. CONCLUSION AND RECOMMENDATION

More knowledge of the chemical constituents of these forest plants(*Laguncularia racemosa)* is desirable, not only for the discovery of new nutrient relevance that could possibly replace some expensive feed ingredients which are in high competition with Man, but also because such information may be of further values to those interested in deciphering the actual phytochemical substances that are ethno-veterinary important or responsible for the various medicinal and other therapeutic agents that could probably influenced reproductive hormones and growth performance of animals and man in general. The phytochemical analysis obtained in this study revealed no toxic properties, however further study is necessary to thoroughly investigate presence of any possible toxic or ant-nutritional substances that may challenge the efficient use this forest plants.

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