

COMPARATIVE PHYTOCHEMICAL, PROXIMATE, NEUTRACEUTICAL AND ANTIMICROBIAL PROPERTIES OF EXTRACTS OF SOME LOCAL PLANT SEEDS

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Abstract

*Plants from the same family and similar usage have been affected by natural phenomena which in turn affect their components. Certain illnesses could be resolved by increased use of foods rich in energy, proteins, iron and vitamins, especially those from the rural environment. However, ignorance of their nutritional properties and essential metabolites could hinder such benefits. Also as antimicrobial resistance is becoming increasingly alarming, there is a need to investigate and compare antimicrobial properties of these plants for man's needs. This study seeks to make a comparative analysis of the phytochemical, proximate and neutraceutical properties and antimicrobial activity of the pulverised plants seeds of *Brachystegia eurycoma*, *Mucuna sloanei*, *Detarium microcarpum* and *C. mannii*. The analyses were investigated by standard procedures and compared. For phytochemicals, *M. sloanei* contains more phytochemical components anthraquinone present in all the extracts, followed by *Brachystegia eurycoma* while *Detarium microcarpum* had the least content. For proximate analysis, carbohydrate and protein had the highest ($P < 0.05$) content in *M. sloanei* (66.29 ± 13.926 and 16.54 ± 0.002 compared to *B. eurycoma*; (59.19 ± 0.000 and 11.21 ± 0.000), and the lowest in *C. mannii*; 16.20 ± 0.0018) with fibre as the least ($2.89 - 2.3 \pm 0.000 - 0.003$) in all plants. For elemental values, *B. eurycoma* showed the highest ($P < 0.05$) composition in carbon (24.26 ± 0.02), oxygen (21.05 ± 0.01) and nitrogen (3.08 ± 0.00) content as against *Detarium microcarpum* with the least content. Antimicrobial activity was seen with a more significant inhibition zone ($P < 0.05$) in *B. eurycoma* ($21 - 3 \text{ mm} \pm 0.1 - 0.0$) compared to *M. sloanei*. ($18 - 2 \text{ mm} \pm 0.1 - 0.0$) with the least in *C. mannii* ($5 - 2 \text{ mm} \pm 0.0$). The results showed that the 4 pulverized plants seeds evaluated contained varying amounts of proximate, phytochemicals, minerals, and antimicrobial activities in the order of *M. sloanei*, *Brachystegia eurycoma*, *Detarium microcarpum* and *C. mannii*. The regular use and analytical study of these plants-seeds is highly recommended.*

Keywords: Phytochemical, Proximate, Neutraceutical, Antimicrobial, Local-plants-seeds.



Introduction

Plants are natural products and gift from God for the benefit of humanity (Natarajan *et al.*, 2012; Viebke, *et al.*, 2014). These plants are sufficiently provided with rich components domiciled at different parts at varying concentrations, necessary for eliciting its effects (Ojiako *et al.*, 2012; Ejere *et al.*, 2015; Ekwe *et al.*, 2016). In the past decades and even presently, plants have long served as useful ingredients for our nutritional purposes and in the treatment of diseases in both developing and developed countries (Muhammed *et al.*, 2011; Bauer and Bronstrup, 2014), numerous interesting chemical substances and compounds has also been synthesized from plants, which have been of great help in the past as well as recent practices. According to World Food and Agriculture Statistic Year book (2021) and World Bank (2016) surveys, 65 and 80 % of the World's populations rely mainly on plant and their products for their wellbeing (WHO, 2004; UNESCO, 2013; Pesic 2015; Tao *et al.*, 2018). Every food substance and plant consumed by humans has either a therapeutic, nutritional or toxic effect on the body, which play a basic role in healthy body development or pathologic inducement (Sofowora, 2008; Uhegbu *et al.*, 2011).

Unfortunately, millions of people in developing countries still suffers from malnutrition, especially infants and children of rural areas (WHF, 2005) which can be tremendously reduced with an increased use of foods rich in energy, proteins, iron and vitamins most especially those from the rural environment. Ignorance concerning the nutritional properties and presence of some phytochemicals, proximate and elemental components, which in turn influence the antimicrobial as well as other activities, are the major reasons for under- utilization of majority of these plants. The food use and consumption of these plants call for more research to provide information on their mineral, pharmacological, phytochemical compositions and properties or their constituents to ascertain their actual nutritional values, health and other medicinal importance. Therefore, this paper seeks to contribute by investigating and comparing the phytochemical, proximate and neutraceutical properties as well as the antimicrobial activity of the pulverized plants seeds of *Brachystegia eurycoma*, *Mucuna sloanei*, *Detarium microcarpum*, and *C. mannii* Naudin since these plants shared a common feature of being used as thickening agents in local delicacies in the South eastern region of Nigeria and are of the same family.

Review of Related Literature and Theoretical Framework

Brachystegia eurycoma (Harm) is a seasonal known legume and economic plant belonging to the family Fabaceae, mostly found in the rain forest zone with attractive description (APG, 2016; Ndukwu, *et al.*, 2017) with its ethno-medicinal and nutritional values, popular among the people of the South eastern part of Nigeria (Okoli, *et al.*, 2015). In some states of Nigeria, *B. eurycoma* is called achi in Igbo, akalado or eku in Yoruba, akpakpa or taura in Hausa, apauan by the Ijaws, dewan in Benin and odukpa in Efik, Ibibio and Annang (Obochi, *et al.*, 2007). *B. eurycoma* is known to exhibit variety of activities (Ajuru and Okoli, 2013; Onyeso, *et al.*, 2016; Ndukwu, *et al.*, 2017).

Mucuna is a genus of around 150 accepted species of annual and perennial legumes of the family fabaceae, and of pan-tropical distribution with India as one of the natural centers in the world (Eilitta, *et al.*, 2002; AGP, 2016). It is widespread in Africa from Sierra Leone east to DR Congo, and south to Angola, also in the Caribbean region, tropical America and islands of the Pacific Ocean (Jansen, 2005). In Nigeria, *Mucuna sloanei* are called different names at



different places; 'ukpo' by the Ibos; 'karasuu' by the Hausas; 'yerepe' by the Yorubas and 'ibaba' by the Annang, Efik and Ibibio but horse eye bean in English (Obochi *et al.*, 2007; Nwosu, 2012). The plants have been reported to possess useful constituents of high medicinal value of human and veterinary importance and also constitute an important raw material in Ayurvedic and folk medicines (Ijeh *et al.*, 2004; Sridhar and Rajeev, 2007; Nagatsua and Sawadab, 2009; Ojiako, *et al.*, 2012; Ejere *et al.*, 2015; Ekwe *et al.*, 2016). The seeds constitute as a good source of several alkaloids, antioxidants, antitumor and antibacterial compounds (Adebowale and Lawal, 2003).

Detarium microcapum (Guil and Perr) is an African tree known as sweet detar, sweet dattock or tallow tree belonging to the family fabaceae (AGP, 2016; Dayamba, *et al.*, 2016; Gaisberger *et al.*, 2017). It is an underutilized species of tree legume that grows naturally in the drier regions of West and Central African; from Senegal and Gambia east to Sudan and widely distributed in the semi-arid sub Saharan African which include Benin, Burkina Faso, Nigeria etc, (Mariod *et al.*, 2009; Padonou *et al.*, 2015). *D. microcapum* is one of the three species (*D. macrocapum* and *D. senegalense*) of *Detarium* plant. *D. microcapum* is called taura in Hausa, kanuri in Fulani, agashidam, etgsako in Tiv, iyede in Yoruba, ofor in Igbo, Annang, Ibibio and Efik, (Iwu, 1993). *D. microcapum* is known to contain bio-active and other substances in different parts for a variety of uses; the fruits are edible and rich in vitamin C, the leaves and seeds are used for cooking, and the root, stems bark are medicinal (Florence *et al.*, 2014).

Methodology

Sample Collection and Identification: The plants-seeds of *B. eurycoma*, *M. sloanei*, *C. manni* and *D. microcapum* were collected from Etim Ekpo and Itam markets/villages in Akwa Ibom State, Nigeria. The seeds were identified, authenticated and with UUPH A32 (ZVii), UUPH A32 (ZVi), UUPH 28 (e) and UUPH A32 (ZVii) as voucher numbers for *B. eurycome*, *M. sloanei*, *C. manni* and *D. microcapum*, respectively, from the Department of Pharmacognosy and Natural Product herbarium, Faculty of Pharmacy, University of Uyo. The samples were dried in an oven at 60 °C for 48 hours and then milled with an electric blender before being used for the analysis.

Seed Pulverization: The seeds of the different plants were pulverized into powder using a hammer mill machine. The pulverized seeds were sieved using a sieving apparatus of size 250 mm. The fine seed powders were separately stored in an air-tight container until needed for use in extraction and other studies.

Preparation of Extract: Extracts of the seeds of *B. eurycome*, *M. sloanei* and *D. microcapum* were carried out by cold maceration of the pulverized seeds using distilled water (aqueous) for 24 hours; and 70 % ethanol, 90 % methanol, as well as 99 % dichloromethane for 72 hours, with intermittent shaking. After 24 and 72 hours, the liquid extracts were filtered, and concentrated *in vacuo* at 40°C using a rotary evaporator. The respective dried crude extracts obtained were differently weighed and stored in a refrigerator at 4 °C for further studies (Sofoworo 1993, Trease and Evans, 2009).

Phytochemical Screening: This was carried out based on the procedure outlined by Sofowora (1993), Harbone (1998), and Trease and Evans (2009) to assess for phytoconstituents such as tannins, flavonoids, saponins, alkaloids, cardiac glycosides and anthraquinones.



Proximate Analysis: The seed samples were analyzed for their proximate compositions using the standard method as described by the Association of Official Analytical Chemists (AOAC), (1990).

Determination of Mineral Content: Calcium, sodium, potassium, magnesium and iron were determined according to the method of Shahidi *et al.* (1999). The phosphorus content of the digest was determined colourimetrically according to the method described by Nahapetian and Bassiri (1995).

Preparation and standardization of microbial culture: Microbial cultures (*Staphylococcus aureus*, *Escherichia coli*, *Aspergillus niger* and *Candida albicans*) were collected from the Department of Pharmaceutical Microbiology and Biotechnology, Faculty of Pharmacy, University of Uyo. The microorganisms were sub-cultured to obtain pure isolates by streaking on freshly prepared Nutrient agar (NA) and Sabouraud Dextrose Agar (SDA) media and incubated at 37 °C for 24 hours and at 28 °C for 48 hours for bacterial cultures and fungal cultures, respectively (Collins and Lyne. 1979). Bacterial cultures were standardised to the turbidity of 0.5 Marfarland nephelometer standard corresponding to cells of approximate density 1×10^8 cfu/mL; In contrast, the fungal cultures were standardised using a haemocytometer to clear turbidity (Ekong *et al.*, 2004).

Antimicrobial Activity of the Plant Seeds Extracts: The antimicrobial activity of the different pulverized plant seed extracts were carried out using the agar-well diffusion technique (AWD) on the standardized microbial inocula on NA and SDA plates for bacteria and fungi culture, respectively (Magaldi, *et al.*, 2004; Valgas, *et al.*, 2007).

Statistical Data Analysis: The results obtained were statistically analysed using Mean $\pm$ SEM, one-way ANOVA, followed by multiple comparison tests, with the aid of the GraphPad Instat-6 tool.

Findings /Discussion of Findings

Solvent Used	<i>M. sloanei</i>		<i>B. eurycoma</i>		<i>C. mannii</i>		<i>D. nicrocarpum</i>	
	Wt (g) of extract	% Yield	Wt (g) of extract	% Yield	Wt (g) of extract	% Yield	Wt (g) of extract	% Yield
Aqueous	39.50	31.60	17.10	13.68	8.05	6.44	33.05	26.44
Ethanol	7.70	6.16	10.00	8.00	35.25	44.20	51.12	40.90
Methanol	10.35	8.28	7.80	6.24	40.60	32.48	58.75	47.00
DCM	4.50	3.60	9.00	7.20	42.15	33.72	37.69	30.15

Table i: Percentage (%) Yield of the Plants Extracts (Weight of the sample for Extraction = 125 g)

Key: DCM= Dichloromethane, Wt= Weight

The extractable potentials of the solvents for the plants-seeds gave the following yield possibly due to their polarity and phytoconstituents of the seed, following the sequence: *M. sloanei*: Aqueous > methanol > ethanol > dichloromethane; *B. eurycoma*: Aqueous > ethanol > dichloromethane > methanol; *C. mannii*: Dichloromethane > methanol > ethanol >

aqueous whereas *D. microcapum*: Methanol > ethanol > dichloromethane > aqueous.

Phytochemical	<i>M. sloanei</i>				<i>B. eurycoma</i>				<i>C. manni</i>				<i>D. microcapum</i>			
Parameters	A	E	M	D	A	E	M	D	A	E	M	D	A	E	M	D
Saponins	+	+	+	-	+	+	+	-	-	-	-	-	+	+	+	-
Tannins	+	+	+	-	+	+	+	-	+	+	+	-	-	-	-	-
Flavonoids	+	+	+	-	+	+	+	-	+	+	+	-	+	+	+	-
Cardiac glycoside	-	+	+	+	-	-	-	-	-	-	-	-	-	-	+	+
Alkaloids	-	+	+	+	+	+	+	-	+	+	+	+	-	-	+	-
Anthraquinones	+	+	+	-	-	-	-	-	-	+	+	-	-	-	-	-

Table ii: Qualitative Phytochemical Analysis of the Pulverized Plants seeds

Key: - (Not present), + (Present), A (Aqueous), E (Ethanol), M (Methanol), D (Dichloromethane)

Phytochemical screening showed qualitative variations in secondary metabolites (saponins, tannins, flavonoids, cardiac glycosides, alkaloids and anthraquinones) in different extraction solvents (Table ii). The polar aqueous extracts showed the presence of tannins, saponin, flavonoids and anthraquinones only in *M. sloanei* with absence of alkaloids and cardiac glycosides, whereas in *B. eurycoma* and *C. manni*; tannins, flavonoids and alkaloids were present, in addition to saponins in *B. eurycoma*. But only flavonoids and saponins was present in *D. microcapum*. The ethanol extracts showed the presence of all phytochemical parameters in *M. sloanei*, 4 in *C. manni* (tannins, flavonoids, alkaloids and anthraquinones) and *B. eurycoma* (saponins, tannins, alkaloids and flavonoids), as well as 2 as the least in *D. microcapum* (saponins and flavonoids). Also, in methanol extracts; *M. sloanei* still showed the highest phytochemical parameters (saponins, tannins, flavonoids, cardiac glycosides, alkaloids and anthraquinones), followed by *D. microcapum* (saponins, flavonoids, cardiac glycosides and alkaloids), *B. eurycoma* (saponins, tannins, flavonoids and alkaloids) and *C. manni* (tannins, flavonoids, alkaloids and anthraquinones). The non-polar dichloromethane extracts showed the presence of, cardiac glycosides and alkaloids in *M. sloanei* alkaloids in *C. manni*, cardiac glycoside in *D. microcapum* and none in *B. eurycoma*, *C. manni* and *D. microcapum*. In all the four plants, *M. sloanei* possessed the highest number (6) of phytochemical parameters in both methanol and ethanol extracts, followed by *B. eurycoma*, and *C. manni*, while the least was *D. microcapum*. Hence this confirms the reason for their difference in antimicrobial and other activities (Joseph *et al.*, 2013; Johnny *et al.*, 2014). This possession of phytochemical parameters also aligned with already published articles (Badifu and Ogunsua, 1991; Obute and Adubor, 2007; Bamina, *et al.*, 2012; Igwe and Okwu, 2013). Tannins are astringent, bitter plant polyphenols that either bind and precipitate or shrink proteins. The astringency from the tannins is what causes the dry and puckery feeling in the mouth following the consumption of red wine or an unripened fruit. Their main function in nature seems to be one of protection; animals are deterred from eating plants high in tannins because of the bitter astringent taste. Tannins have traditionally been considered anti-

nutritional but it is now known that their beneficial or anti-nutritional properties depend upon their chemical structure and dosage (Muller-Harvey and McAllan, 1992). Recent studies have demonstrated that products containing chestnut tannins in the diet can improve wellbeing (Schiavone *et al.*, 2007). Flavonoids have been referred to as "nature's biological response modifiers" because of strong experimental evidence of their inherent ability to modify the body's reaction to allergens, viruses, and carcinogens. They show anti-allergic, anti-inflammatory (Yamamoto and Gaynor, 2006), anti-microbial (Cushine and Lamb, 2005) and anti-cancer activity. Flavonoids are antioxidants that play major roles in the protection of cells from the lethal effects of free radicals and their derivatives (Nyerhowo *et al.*, 2015). A diet rich in antioxidant compounds (like phenols and flavonoids) therefore helps to strengthen the antioxidant-based defence system in the human body (Nyerhowo *et al.*, 2015). Alkaloids are famous analgesics (Mothes, 1996) and have been utilized in a variety of ways in the treatment of diseases and during surgery due to their medicinal and pharmacological potency.

Content	<i>M. sloanei</i>	<i>B. eurycoma</i>	<i>C. manii</i>	<i>D. microcarpum</i>
Moisture	3.76±0.002	14.44±0.002	8.26±0.001	16.68±0.001
Protein	16.54±0.002	11.21±0.000	26.32±0.000	8.21±0.000
Fat	5.44±0.002	7.38±0.001	42.67±0.000	10.58±0.006
Ash	3.67±0.000	4.87±0.000	4.09±0.000	4.29±0.000
Fibre	2.30±0.000	2.89±0.000	2.46±0.003	2.86±0.000
CHO	66.29±13.926	59.19±0.000	16.20±0.018	57.37±0.002

Table iii: Proximate Composition (%) of the Pulverized Plants Seeds

Key: Values are presented as Mean±SEM, n=3.

The proximate compositions (%) of the pulverized plants seeds as seen in Table.iii showed significantly differences among the seeds. *M. sloanei* had the highest carbohydrate content of 66.29 %, followed by *B. euryoma* (59.19%) and *D. microcapum* (57.3 %) and *C. mannii* (16.20 %) as the least respectively. After carbohydrate, protein contents showed the next highest values in the sequence *C. mannii* (26.32%), *M. sloanei* (16.54 %), *B. euryoma* (11.21 %), and the least in *D. microcapum* (8.21 %). The results varied with that reported by Igwenyi and Akubugwo, (2010). These values give the seeds positive attributes as plant proteins are scarce and this protein contents can furnish the essential amino acids needed for healthy growth and repair of tissues (Igwenyi, 2008). *D. microcapum* showed the highest (16.68 %) moisture content and the least in *M. sloanei* (3.76%), whereas fat was highest in *C. mannii* (42.67 %), followed by *D. microcapum* (10.58 %), *B. euryoma* (7.38 %) with *M. sloanei* (5.44 %) as the least. The result is also in line with the work of Ejiofor, (1994). All the four plants seeds were observed to be generally low in crude fibre as the least value in *M. sloanei* (2.30 %), *C. mannii* (2.46 %), *D. microapum* (2.86 %) and *B. euryoma* (2.89 %). These values are close to the values reported by Akpata and Miachi (2001) and Barminas *et al.*, (2004) respectively. Fiber regulates bowel actions and may help to guard against colon and rectal cancer as well as in diabetes. Crude fiber is the inorganic residue left after the defatted food materials have been treated with boiling dilute hydrochloric acid, diluted sulphuric acid, boiling dilute sodium hydroxide, alcohol and ether. It is that portion of food

that is not used up by the body. Fiber shortens the transit time of food through the gastrointestinal tracts, reduces low density lipoprotein and hence keeps the gut healthy. Fiber supplements or fiber-rich foods may function as normal dietary agents by modulating the digestive and absorptive process (Okaka *et al.*, 2006). They are very important in promoting a range of physiological effects, including increased fecal bulk, water-holding capacity, absorption of organic molecules such as bile acids, cholesterol and toxic components (reduced bile acid and plasma-cholesterol levels), reduction of minerals and electrolytes (Igwenyi, 2008).

C. mannii recorded the highest (42.67 %) crude fat content, with *M. sloanei* as the least (5.44 %). Lipids are the principal form of stored energy (fat and oils) in most organisms and major constituents of cellular membranes. Specialized lipids serve as pigments (retinal, carotene), cofactors (vitamin K), detergents (bile salts), transporters (dolichoils in bacteria cell wall synthesis), hormones (vitamin D derivatives, sex hormones), extracellular and intracellular messengers (eicosanoids), and anchors for membrane proteins (covalently attached fatty acids, phosphatidyl inositol, etc) (Voet and Voet, 2004; Nelson and Cox, 2005). The ash content was slightly higher than crude fat but more in *B. eurycoma* (4.87 %) and less in *M. sloanei* (3.67 %). The result also showed comparative studies on the nutritional and anti – nutritional properties of indigenous seeds to that of other authors (Barminas *et al.*, 2004); Odenigbo and Obizoba, 2004).

There were significant differences ($P < 0.05$) in the carbohydrate, fat, protein and moisture contents of the pulverized plants seeds. This could be sequentially summarized thus; Carbohydrate content (*M. sloanei* > *B. eurycoma* > *D. microcapum* > *C. mannii*) >> Fat content (*C. mannii* > *D. microcapum* > *B. eurycoma* > *M. sloanei*) >> Protein content (*C. mannii* > *M. sloanei* > *B. eurycoma* > *D. microcapum*) >> Moisture content (*D. microcapum* > *B. eurycoma* > *C. mannii* > *M. sloanei*) >> Ash content (*B. eurycoma* > *D. microcapum* > *C. mannii* > *M. sloanei*) >> Fibre content (*B. eurycoma* > *D. microcapum* > *C. mannii* > *M. sloanei*). The proximate content of these 3 plants in their decreasing order, followed the sequence; Carbohydrate >> Fat >> Protein >> Moisture >> Ash >> Fibre. This is in line with already documented literatures (Akpata and Miachi 2001; Uhegbu *et al.*, 2009; Abiodun and Adeleke 2010; Johnny *et al.*, 2014).

Samples	Na	Ca	Mg	Fe	K	H	C	O	N	P
<i>B. eurycoma</i>	0.14± 0.00	0.77± 0.00	0.0± 0.00	0.34± 0.00	0.48± 0.00	3.16± 0.00	21.05± 0.01	24.26± 0.02	3.08± 0.00	0.17± 0.00
<i>M. sloanei</i>	0.02± 0.00	0.90± 0.00	0.06± 0.00	7.45± 0.00	0.51± 0.00	4.11± 0.00	9.18± 0.00	11.11± 0.01	5.05± 0.00	28.05± 0.01
<i>D. microcapum</i>	0.12± 0.00	0.35± 0.00	0.1± 0.00	0.02± 0.00	0.15± 0.00	2.85± 0.00	12.04± 0.01	18.01± 0.02	2.21± 0.00	0.15± 0.00
<i>C. manii</i>	2.48± 0.02	9.27± 0.01	28.38± 0.03	5.42± 0.00	19.35± 0.02	2.05± 0.02	8.55± 0.02	0.33± 0.00	0.21± 0.10	2.40± 0.02

Table iv: Nutraceutical/Elemental Content of the Pulverized Plants seeds

Key: Na (Sodium), Ca (calcium), Mg (magnesium), Fe (Iron), k (Potassium), H (Hydrogen), C (carbon), O (Oxygen), N (Nitrogen), P (Phosphorus)

Also, from Table iv, the result showed huge differences in elemental deposition. The 4 plants were found to show higher content in major (H, C, N, O, P) than minor (Mg, Na, Fe,

k, Ca) elements with significant variations. High deposition was seen in oxygen. The oxygen contents were found in the range of 24.26 to 24.30 mg/100g in *B. eurycoma*; 18.01 to 18.05 mg/100g in *D. microcapum*, 11.11 to 11.16 mg/100g in *M. sloanei* and the lowest in *C. mannii* 5.15 to 5.25 mg/100g. The same order (*B. eurycoma* > *D. microcapum* > *M. sloanei* > *C. mannii*) followed for calcium and sodium. Calcium as an important mineral is a structural component that contributes to physical strength of bones and teeth, in muscle concentration, blood coagulation, nerve impulse transmission etc. The iron contents were found to be highest in *M. sloanei* in 7.45 to 7.50 mg/100g, 5.42 to 5.45 mg/100g in *C. mannii*, 0.34 to 0.35 mg/100g in *B. eurycoma* and 0.02 to 0.03 mg/100g in *D. microcapum* as the least. Iron is a component of many proteins and enzymes, notably hemoglobin and cytochrome P450.

Deficiency of iron could induce iron deficiency anaemia which is more common in menstruating females and pregnant women (Dvlin, 200) the RDA of iron in adults in between 15.20-30 mg for children and 40mg for pregnant women (Vasudevan and Sreekumari, 2007). For Mg, *C. mannii* showed the highest content in the range of 28.38 to 29.01 mg/100g, followed 0.18 to 0.20 mg/100g in *D. microcapum*, 0.08 to 0.09 mg/100g in *B. eurycoma* while the least was seen in *M. sloanei* ranging from 0.06 to 0.08 mg/100g. For nitrogen and phosphorus, *M. sloanei* showed the highest content in the range of 5.05 to 5.07 and 28.05 to 28.15 mg/100g. *B. eurycoma* followed with 3.08 to 3.10 mg/100g and 0.17 to 0.19 mg/100g. Whereas 2.21 to 2.23 mg/100g and 0.13 to 0.17 mg/100g was for *D. microcapum* while the least was seen in *C. mannii* with 3.02 to 3.04 mg/100g and 2.35 to 2.55 mg/100g. The hydrogen contents were highest in *Mucuna sloanei* at the range of 4.10 to 4.13 mg/100g, 3.15 to 3.17 mg/100g with *B. eurycoma*, 2.75 to 2.95 mg/100g) in *D. microcapum*, with 2.00 to 2.15 mg/100g as the lowest in *C. mannii*. The highest carbon content was found in *B. eurycoma* at the range of 21.00 to 21.45 mg/100g, 12.02 to 12.06 mg/100g for *D. microcapum*, 9.15 to 9.21 mg/100g for *Mucuna sloanei* and the lowest was 8.50 to 8.60 mg/100g in *C. mannii*. Finally, the highest potassium content was found in *C. mannii*, at the range of 19.30 to 19.45 mg/100g. *Mucuna sloanei* followed at the range of 0.50 to 0.53 mg/100g, 0.46 to 0.52 mg/100g was seen in *B. eurycoma* and lastly 0.14 to 0.16 mg/100g for *D. microcapum* as the lowest.

The elemental content follows the sequence; Hydrogen: *M. sloanei* > *B. eurycoma* > *D. microcapum*; Carbon: *B. eurycoma* > *D. microcapum* > *M. sloanei*; Oxygen: *B. eurycoma* > *D. microcapum* > *M. sloanei*; Nitrogen: *M. sloanei* > *B. eurycoma* > *D. microcapum*; Phosphorus: *M. sloanei* > *B. eurycoma* > *D. microcapum*; Sodium: *B. eurycoma* > *D. microcapum* > *M. sloanei*; Calcium: *M. sloanei* > *B. eurycoma* > *D. microcapum*; Magnesium: *D. microcapum* > *B. eurycoma* > *M. sloanei*; Sodium is an electrolyte present in extracellular fluid and is essential for coregulating ATP with potassium (Linder, 1991) sodium is also important in the regulation of acid base balance (Vasudevan and Sreekumari 2007). Iron: *M. sloanei* > *B. eurycoma* > *D. microcapum* > *C. mannii*; Potassium: *M. sloanei* > *B. eurycoma* > *D. microcapum*. Potassium is a systemic electrolyte and is essential for coregulating ATP with sodium. Potassium is a major intracellular cation that maintains intracellular osmotic pressure (Vasudevan and Sreekumari 2007). This also aligned with their activity differences and already published work (Uhegbu *et al.*, 2009; Ameh *et al.*, 2010; Bolanle *et al.*, 2014).



Table v: Assessment of Antimicrobial Activity Test of the Pulverized Seeds of *M. sloanei*

Conc (g/mL)	Zone of Inhibition (mm)															
	<i>S. aureus</i>				<i>E. coli</i>				<i>A. niger</i>				<i>C. albicans</i>			
	M	E	A	D	M	E	A	D	M	E	A	D	M	E	A	D
0.1000	8±	12±	16±	18±	6±	11±	9±	14±	5±	11±	7±	12±	7±	15±	9±	7±
	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
0.0500	6±	8±	14±	15±	-	9±	6±	10±	-	6±	4±	8±	4±	12±	5±	3±
	0.0	0.0	0.0	0.0		0.0	0.0	0.0		0.0	0.0	0.0	0.0	0.0	0.0	0.0
0.0250	-	5±	11±	10±	-	6±	4±	7±		3±	-	5±	-	9±	-	-
		0.0	0.0	0.0		0.0	0.0	0.0	-	0.0		0.0		0.0		
0.0125	-	-	8±	6±	-	2±	-	5±	-	-	-	-	-	6±	-	-
			0.0	0.0		0.0		0.0						0.0		

Key: (-) NO Zone of inhibition/no activity, (values or numbers (Mean±SEM) Inhibition zone diameter (mm),

M-methanol, E-ethanol, A-aqueous, D-dichloromethane

Conc (g/mL)	Zone of Inhibition (mm)															
	<i>S. aureus</i>				<i>E. coli</i>				<i>A. niger</i>				<i>C. albicans</i>			
	M	E	A	D	M	E	A	D	M	E	A	D	M	E	A	D
0.1000	9±	11±	14±	16±	21±	22±	19±	18±	19±	15±	18±	21±	10±	18±	15±	14±
	0.0	0.0	0.1	0.0	0.1	0.2	0.1	0.0	0.1	0.0	0.0	0.1	0.0	0.0	0.0	±
	0															0.0
0.0500	4±	8±	11±	12±	15±	16±	16±	14±	12±	11±	14±	16±	5±	15±	9±	8±
	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	0															
0.0250	-	5±	7±	10±	4±	11	9±	8±	5±	8±	10	10±	-	11	5±	-
		0.0	0.0	0.0	0.0	±	0.0	0.0	0.0	0.0	±	0.0		±	0.0	
						0.0					0.0			0.0		
0.0125	-	3±	4±	7±	-	8±	6±	5±	-	-	7±	-	-	7±	-	-
		0.0	0.0	0.0		0.0	0.0	0.0			0.0			0		

Table vi: Assessment of Antimicrobial Activity Test of the Pulverized Seeds of *B. eurycoma*

Key: (-) No Zone of inhibition, (values or numbers (Mean±SEM) Inhibition zone diameter (mm),

M-methanol, E-ethanol, A-aqueous, D-dichloromethane

Conc(g/mL)	Zone of Inhibition (mm)															
	<i>S. aureus</i>				<i>E. coli</i>				<i>A. niger</i>				<i>C. albicans</i>			
	M	E	A	D	M	E	A	D	M	E	A	D	M	E	A	D
0.1000	4	12	15	17	10	16	16	14	12	16	18	19±	10	19	2±	21
	±	±	±	±	±	±	±	±	±	±	±	0.0	±	±	0.0	±
	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0		0.0	0.0		0.0
	0															
0.0500	-	9±	11	15	6±	12	11	10		10	15	17±	8±	17	10	16
		0.0	±	±	0.0	±	±	±	9±	±	±	0.0	0.0	±	±	±
			0.0	0.0		0.0	0.0	0.0	0.0	0.0	0.0			0.0	0.0	0.0
0.0250	-	7±	8±	11	-	-	-	-	-	9±	12	11±	-	14	8±	11
		0.0	0.0	±						0.0	±	0.0		±	0.0	±
				0.0							0.0			0.0		0.0
0.0125	-	4±	5±	-	-	-	-	-	-	-	10	-	-	10	7±	9±
		0.0	0.0								±			±	0.0	0.0
											0.0			0.0		

Table vii: Assessment of Antimicrobial Activity Test of the Pulverized Seeds of *D. microcarpum*

Key: (-) No Zone of inhibition, (values or numbers (Mean±SEM) Inhibition zone diameter

in mm, M-methanol, E-ethanol, A-aqueous, D-dichloromethane

Conc (g/mL)	Zone of Inhibition (mm)															
	<i>S. aureus</i>				<i>E. coli</i>				<i>A. niger</i>				<i>C. albicans</i>			
	M	A	D	E	M	A	D	E	M	A	D	E	M	A	D	E
0.1000	-	3±0	-	5±0	-	4±0	2±0	2±0	-	2±0	-	1±0	-	2±0	5±0	2±0
	.0		.0		.0	.0	.0	.0	.0		.0		.0	.0	0	
0.0500	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	-															
0.0250	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	-															
0.0125	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	-															

Table viii: Assessment of Antimicrobial Activity Test of the Pulverized Seeds on *C. manni*

Key: (-) No Zone of inhibition, (values or numbers (Mean±SEM) Inhibition zone diameter (mm),

M-methanol, E-ethanol, A-aqueous, D-dichloromethane

The assessment of antimicrobial activity of the different plant seed extracts was dose-concentration dependent with variations in microbial isolates inhibition as shown in Table v. Antimicrobial activity was seen in almost all the plant extracts from 22±0.0 mm to 2±0.0 mm for 0.100 g/ mL and 0.0125 g/ mL as the highest and least inhibition zone diameters (IZD) and concentrations. All the four microbial isolates showed varying IZD (mm) on the different pulverized plant seeds.

For *M. sloanei*, dichloromethane and aqueous extracts showed IZDs in all the concentrations (0.0125-0.1000 g/ mL) but not in all for methanol and ethanol extracts. The highest IZD was observed in dichloromethane (18±0.0), (14±0.0) and (12±0.0) while the least was seen in methanol extracts (8±0.0), (6±0.0) and (5±0.0) for *S. aureus*, *E. coli*, and *A. niger* respectively (Table 4.1.7.1). But for *C. albicans*, the highest and least IZD was observed in ethanol (15±0.0) and dichloromethane (3±0.0) extracts. Therefore, the assessment of antimicrobial activity of the different plants seeds extracts and microbial isolates for *M. Sloane* followed the sequence: *S. aureus*: Dichloromethane > Aqueous > Ethanol > Methanol; *E. coli*: Dichloromethane > Ethanol > Aqueous > Methanol; *A. niger*: Dichloromethane > Ethanol > Aqueous > Methanol; *C.albicans*: Ethanol > Aqueous > Methanol > Dichloromethane.

For *B. eurycoma*, as presented in Table 6, methanol extracts did not show activity (no IZD) in 0.025 and 0.0125 g/ mL concentrations in all the microbial isolates. Dichloromethane, aqueous and ethanol extracts showed varying IZD in all the microbial isolates. For *S. aureus*, the highest (16±0.1) and least (9±0.1) IZD was observed in dichloromethane and methanol extracts at 0.1000 g/ mL. For *E. coli*, ethanol and aqueous extracts showed the highest (22±0.2) and (19±0.1) IZD. For *A. niger*, the highest (21±0.1) and least (15±0.0) was observed in dichloromethane and ethanol extracts, while for *C. albicans*, ethanol and methanol extracts showcased the highest (18±0.0) and least (10±0.0) IZD respectively.

Therefore, assessment of antimicrobial activity test on the different isolates using different extracts could be shown by the sequence; *S. aureus*: Dichloromethane > Aqueous > Ethanol > Methanol; *E. coli*: Ethanol > Methanol > Aqueous > Dichloromethane; *A. niger*: Dichloromethane > Methanol > Aqueous > Ethanol; *C. albicans*: Ethanol > Aqueous > Dichloromethane > Methanol.

As presented in Table 7, the assessment of antimicrobial activity on the pulverized plants-seeds on *D. microcarpum* also showed varying IZD as observed in *S. aureus*, *A. niger* and *C. albicans* at 0.0250 and 0.0125 g/ mL except for *E. coli* with no activity (no IZD) for methanol, ethanol, aqueous and dichloromethane extracts. Methanol extracts had no activity (no IZD) at 0.0500 g/ mL, 0.0250 and 0.0125 g/mL in all microbial isolates. For *S. aureus*, the highest and least IZD was observed in dichloromethane (17 ± 0.0) and methanol (4 ± 0.0) extracts. For *E. coli*, ethanol and aqueous extracts showed same and highest IZD (16 ± 0.0) with the least IZD (10 ± 0.0) seen in methanol extracts. For *A. niger*, dichloromethane and methanol extracts showed highest (19 ± 0.0) and least (12 ± 0.0) IZD, while for *C. albicans*, the highest and least IZD was seen in dichloromethane (21 ± 0.0) and methanol (10 ± 0.0) extracts. This antimicrobial activity test assessment could be sequentially presented thus: *S. aureus*: Dichloromethane > Aqueous > Ethanol > Methanol; *E. coli*: Ethanol = Aqueous > Dichloromethane > Methanol; *A. niger*: Dichloromethane > Aqueous > Ethanol > Methanol; *C. albicans*: Dichloromethane > Ethanol > Aqueous > Methanol.

The assessment of antimicrobial activity test of the different microbial isolates and extracts attested to the fact that the pulverized plants-seeds of *B. eurycoma* has the highest and least antimicrobial activity (IZD) on *E. coli* (22 ± 0.0) and *S. aureus* (3 ± 0.0) and in *C. albicans* for dichloromethane extracts (21 ± 0.0) and the least for *S. aureus* (4 ± 0.0) at 0.1000 g / mL. With the least in the sequence been *M. sloanei* whose highest activity (IZD) was observed in *S. aureus* on dichloromethane extracts (18 ± 0.0) and least activity in *E. coli* with ethanol extracts (2 ± 0.0). Comparing activities in the different extraction solvents, the highest and least IZD was seen in dichloromethane and methanol extracts respectively.

For *C. mannii*, as presented in Table 8, antimicrobial activity was almost not feasibly seen (no IZD) especially in methanol extracts of the different pulverized plants seeds. Also, no activity (IZD) was seen in aqueous extracts on *S. aureus* and *A. niger*. But antimicrobial activity was only seen at a concentration of 1.000 g/ mL in with IZD range of 1 ± 0.0 to 5 ± 0.0 in dichloromethane and ethanol extracts in all the microbial isolates (*S. aureus*, *E. coli*, *A. niger* and *C. albicans* respectively). Therefore, *C. mannii* possessed the least antimicrobial activity compared to *B. eurycoma*, *D. microcarpum* and *M. sloanei*.

Critically looking at the results;

M. sloanei at a concentration of 1.000 g/mL, had the highest IZD of 18 ± 0.1 and 16 ± 0.0 on *S. aureus* with dichloromethane and aqueous extracts, while methanol extract showed the least IZD with 5 ± 0.0 on *A. niger*. Whereas at the least concentration of 0.0125 g/mL, highest and least IZD was observed at 8 ± 0.0 and 2 ± 0.0 for aqueous and ethanol extracts on *S. aureus* and *E. coli*. But for *B. eurycoma*, at a concentration of 1.000 g/mL, the highest IZD of 22 ± 0.2 on *E. coli* with ethanol extract and 21 ± 0.1 on *E. coli* and *A. niger* with methanol and dichloromethane extracts respectively, while methanol extract showed the least IZD with 9 ± 0.0 on *S. aureus*. Whereas at the least concentration of 0.0125 g/mL, highest and least IZD was observed only with ethanol extracts at 8 ± 0.0 and 3 ± 0.0 on *E. coli* and *S. aureus*. Also, for *D. microcarpum*, at a concentration of 1.000 g/mL, the highest IZD of 21 ± 0.0 on *C. albicans*

and 19 ± 0.0 on *C. albicans* and *A. niger* with ethanol and dichloromethane extracts was observed, while methanol extract showed the least IZD with 4 ± 0.0 on *S. aureus*. Whereas at the least concentration of 0.0125 g/mL , highest and least IZD was observed at 10 ± 0.0 on *A. niger* and *C. albicans* for aqueous and ethanol extracts and 4 ± 0.0 on *S. aureus* on ethanol extracts. But *C. mannii*, at a concentration of 1.000 g/mL , showed the least high IZD of 5 ± 0.0 on *S. aureus* and *C. albicans* with dichloromethane and aqueous extracts, whereas the least IZD was seen at 1 ± 0.0 on *A. niger* with dichloromethane extracts. This could be a function of the species and physicochemical components imbedded in the pulverized plants seed of these plants. Thus, the sequence of assessment of antimicrobial activity follows: *B. eurycoma* > *D. microcarpum* > *M. sloanei* > *C. mannii* while microbial isolates and extracts potential follow: Dichloromethane > Ethanol > Aqueous > Methanol. Though no detailed comparative work on these pulverized plants-seeds could be found, different publications on each plant agrees to these plants having varying antimicrobial activity but disagrees in respect to some species and IZD (Adekunle, 2000; Ebi and Afieroho, 2011; Igwe and Okwu, 2013; Igwe and Echeme, 2013; and Okoli, *et al.*, 015; Priya *et al.*, 2016).

Conclusion

These 4 plants are rich in carbohydrate and protein contents, some secondary metabolites (flavonoids, saponins, tannins, cardiac glycoside, alkaloids, etc) at varying degree; macro (H, C, O, N and P) and micro (Mg, Fe, Na, k and Ca) and elements necessary to solve man's needs. These could serve as fuel for the generation of the energy, metabolic activities, growth and development of the cell and all other activities recorded for these plants. Also, in an era where antimicrobial resistance is becoming increasingly alarming, there is a need to intensify research and usage of plants with antimicrobial properties as alternative to orthodox medications. However, this comparative study is necessary to ascertain previous documents on component, quantity and activity, effect of climatic, environmental and agricultural changes in plants and its activities, diversification in usage (for dietary deficiency, elemental supplement, toxicity studies), antimicrobial activity and further research work, enhance disease/infection diagnosis and effective therapeutic management and pharmaceutical products.

Therefore, routine research is recommended on plant-based nutrients for healthy consumption and the effect of climate and environmental change.

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