



PRINT ISSN 1119-8362
ELECTRONIC ISSN 1119-8362

Full-text Available Online at

<https://www.ajol.info/index.php/jasem>
<http://www.bioline.org.br/ja>

J. Appl. Sci. Environ. Manage.
Vol. 23 (1) 65–74 January 2019

Proximate Composition, Vitamin, Mineral and biologically Active Compounds Levels in Leaves of *Mangifera indica* (Mango), *Persea americana* (Avocado pea), and *Annona muricata* (Sour sop)

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ABSTRACT: The high rate of food insecurity in Nigeria has resulted to malnutrition and sicknesses especially in the rural areas. To address this challenges, this study assessed the levels of biologically active compounds, some essential proximate, vitamin and mineral in leaves of Mango, Avocado pea and Sour sop to determine their nutritional and health benefits. The results indicate that the highest significant ($P < 0.05$) values of crude protein (17.94 ± 0.99 %), calorific value (370.47 ± 1.01 KJ), carbohydrate (66.04 ± 1.00 %), thiamine (0.52 ± 0.01 mg), beta-carotene (115.50 ± 0.01 mg) and K (0.60 ± 0.01 %), alkaloid (2.26 ± 0.01 %) and saponin (1.33 ± 0.01 %) were observed in Avocado pea while ether extract (4.30 ± 0.95 %), ash (8.24 ± 0.99 %), crude fiber (10.60 ± 0.95 %), ascorbic acid (13.20 ± 0.90 mg), riboflavin (0.21 ± 0.01 mg), Ca (4.41 ± 0.01 %), P (0.40 ± 0.01 %), tannin (1.38 ± 0.01 %), flavonoid (0.85 ± 0.01 %) and phenol (0.37 ± 0.01 %) were obtained in Mango. The highest content of niacin (0.41 ± 0.01 mg), Mg (1.70 ± 0.01 %), N (2.98 ± 0.99 %) and phytate (0.30 ± 0.01 %) were recorded in sour sop. The highest values of Na was statistically the same ($P > 0.05$) in Mango (0.23 ± 0.01 %) and Sour sop (0.23 ± 0.01 %) but significantly ($P < 0.05$) higher than 0.19 ± 0.01 % in Avocado pea. Similarly the highest moisture content was observed in Avocado pea (21.74 ± 0.99 %) which is statistically equal with 20.10 ± 0.90 % in Mango but significantly ($P < 0.05$) higher than 16.58 ± 1.00 % in Sour sop. The leaves of Mango and Avocado pea are potential source of vitamins, mineral, phytochemical and proximate composition and is strongly recommended for nutritional and therapeutic uses to enhance good health of people.

DOI: <https://dx.doi.org/10.4314/jasem.v23i1.11>

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Dates: Received: 30 November 2018; Revised: 06 January 2019; Accepted 18 January 2019

Key words: proximate, mineral, vitamins, phytochemicals, mango, avocado, sour sop.

The high rate of food insecurity in Nigeria has resulted to malnutrition. Indeed, various forms of sicknesses are being experienced by people especially in the rural areas due to poor nutrition vis-a-vis low level of immunity against diseases. Most underutilized fruits trees have appreciable health benefits and contribute significantly to poverty alleviation in low and middle income nations (Hall *et al.*, 2009; Padulosi *et al.*, 2006; Williams and Haq, 2000). The appreciation of this concept in the recent past, has led to a paradigm shift of researchers into exploring the optimal use of underutilized fruit trees with emphasis on their nutritive, health-beneficial constituents, dietary fibre and phytochemicals. Natural products, either as pure compounds or as standardized plant extracts, provide unlimited opportunities for new drug because of the unmatched availability of chemical diversity (Pathak *et al.*, 2010). Phytochemical screening is an important method of identifying bioactive compound that will be useful in creating new drugs. These simple, cheap,

sensitive, selective and rapid chemical tests to determine the presence of certain groups of compounds is an initial step to select plants for further phytochemical studies (Ibrahim, 2004).

Persea americana (common name: Avocado, family: Lauraceae) is commercially important fruit tree that have cardiovascular benefits. Studies have been carried out on *P. americana* including phytochemical studies on avocado seeds (Leite *et al.*, 2009; Ding *et al.*, 2007), avocado pulp for wound healing and hair growth (Ding *et al.*, 2007), as an aphrodisiac (Duke and Vasquez 1994), *in vitro* growth inhibitory effects against cancer (Lu *et al.*, 2009), oil extracted from the pulp used in cosmetics (Bleinroth and Castro, 1992), and acute and sub-chronic safety of orally-administered seed extract was evaluated in the rat and it was found to be safe even at high doses (Dabas *et al.*, 2011). *Mangifera indica* Linn (common name: Mango, Family: Anacardiaceae) is evergreen fruit tree spreading with dense rounded crown and the fruit is

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greatly relished for its succulence, exotic flavour and delicious taste in most countries of the world (Bhatnagar and Subramanyam, 1973). It is used in control of heart diseases, urinary disorders, dysentery, eye diseases, diarrhea, syphilis, ulcer, diabetes, kidney stone, sunstroke, tuberculosis, intestinal disorder, and blood purification, nasal bleeding, in piles and in heart diseases (Cheikhoussef *et al.*, 2011; Achuta *et al.*, 2010; Agrawal, 1986). *Annona muricata* Linn (common name: Sour sop, Family: Annonaceae) is a deciduous fruit tree that produces a heart-shaped, highly aromatic fruit. The fruit's nectar is commonly used in smoothies and yoghurts, giving this plant yet another cultural use (Lutchmedial *et al.*, 2004). Studies have reported an array of phytochemicals with a corresponding high antioxidant activity for inedible morphological parts of Brazilian varieties of *Annona muricata* (Gomes *et al.*, 2010), the efficacy of leaf extracts of some Ghanaian varieties against jaundice (Arthur *et al.*, 2012), and the use of the fruit pulp as lactagogue and as cure for diarrhea and dysentery in Nigeria (Adewole and Caxton-Martins, 2006). Research on *A. muricata* has focused on the bark of the tree and roots for pharmaceutical purposes (Kimbonguila *et al.*, 2010) and little attention has been paid to the leaves, which is usually used in traditional medicine remedies. Literature search has shown that there are paucity of scientific research publication on the phytochemical, mineral and vitamin content in leaves of *A. muricata*, *P. americana*, and *M. indica* in South east Nigeria. This study, therefore, investigate key parameters for assessing the health potential of the fruit trees (*Annona muricata*, *Persea americana*, and *Mangifera indica*) to complement literature in making known the potential of these species as well as provide a stepping-stone for further research works in other uses of the fruit trees.

MATERIALS AND METHODS

Study area: The study was carried out at Umuanunu in Umunumo, Ehime Mbano Local Governemnt Area of Imo State, Nigeria. Umunumo is situated within latitude 5°40' and longitude 7°17' E at an altitude of 252 m and a population of 74,009 (Fallingrain, 2016).

Sample collection and analysis: A reconnaissance survey was carried out prior to sample collection to identify plant species that are common in Umuanunu and are utilized by the people for therapeutic purposes. The survey showed that *M. indica*, *P. americana*, and *A. muricata* are the common plant species used for herbal remedies by the inhabitants of Umuanunu. Five (5) stands each of *M. indica*, *P. americana*, and *A. muricata* were selected randomly from each of the three (3) kindred's in Umuanunu. Fresh leaves were

collected randomly from fifteen (15) stands each of *Annona muricata*, *Mangifera indica*, and *Persea americana* using well cleaned secateurs. The leaves were collected separately from various branches of each of the tree species, placed separately in large envelopes, labeled well, covered in a wooden box to avoid contamination, and taken to the Laboratory for pre-treatment and analysis. The leaves from each plant species were removed from the stalk separately, thoroughly washed with running tap water to remove dust, pollen grains and other debris. Thereafter, the leaves from each plant species were air-dried separately for 14 days at room temperature (24°C) to constant weight. The dried leaves from each plant species were milled with Wiley milling Machine (Model ED-5) to fine powder and stored separately in well cleaned and dried plastic bottles, labeled well. Aliquot portions of the powdered leaves was weighed and used for analysis of phytochemical, mineral, proximate, and vitamin content of the leaves.

Determination of proximate composition of *Mangifera indica*, *Persea americana* and *Annona muricata* leave: The dry matter, moisture, ash, crude fat, crude protein (nitrogen x 6.25) and crude fiber contents were determined using the standard methods of the Association of Official Analytical Chemists (AOAC, 2000) while the carbohydrate content and calorific content were determined according to the method of James (1995).

Determination of mineral content: The AOAC (2000) method was used to determine the mineral content (Ca, Mg, K, Na, P, and N) using the atomic absorption spectrophotometric method (Uzoekwe and Mohammed, 2015).

Determination of vitamin content: Beta carotene (precursor of vitamin A), thiamin (vitamin B1), riboflavin (vitamin B2), ascorbic acid (vitamin C) and niacin (vitamin B3) content of the samples were determined using the method of Pearson (1976).

Determination of phytochemical content: The procedure described by Usunobun *et al.* (2015) was used to determine the phytochemical content with no modification.

Phytochemical Screening: The phytochemicals determined in the ethanolic leaf extracts include: flavonoids, alkaloids, tannins, saponins, phenol, and phytaes. This test was performed using standard procedures (Sofowora, 1993, Trease and Evans, 1989, Ayoola *et al.*, 2008).

Data analysis: The data generated from the study is subjected to one way analysis of variance (ANOVA) using Statistical Package for Social Sciences (SPSS) and means separated by Duncan Multiple Range Test (DMRT) to compare the differences in means at $P < 0.05$.

RESULTS AND DISCUSSION

The data generated from the analysis of samples in the Laboratory were subjected to one way analysis of variance and the results of the separation of means using Duncan Multiple Range Test is presented in Tables 4.1, 4.2, 4.3, and 4.4, respectively.

Proximate composition of leaves of *Mangifera indica*, *Persea americana* and *Annona muricata*: The result of the proximate composition of leaves of *Mangifera indica*, *Persea americana* and *Annona muricata* is summarized in Table 1. The values of the proximate composition was significantly raised and the significant differences was evidenced amongst the

three plant species studied. The highest moisture content (MC) was obtained in *P. americana* (21.74 ± 0.99 %) which is statistically equal ($P > 0.05$) with the value obtained in *M. indica* (20.10 ± 0.90 %) but significantly ($P < 0.05$) higher than the value recorded for *A. muricata* (16.58 ± 1.00 %). In this study, the value of moisture content in *P. americana* is well above the value of moisture content (5.33 ± 0.62 g/100g) reported in *P. americana* leaves by Arukwe *et al.* (2012). Low moisture content would therefore hinder the growth of spoilage microorganisms and enhance shelf life (Ruberto and Baratta, 2000). The highest content of ether extract (fat) was obtained in leaves of *M. indica* (4.30 ± 0.95 %) but the value is not significantly different ($P > 0.05$) from the values obtained in *P. americana* leaves (3.84 ± 0.99 %) and *A. muricata* (2.98 ± 1.00 %). The highest value of ether extract (fat) obtained in leaves of *M. indica* (4.30 ± 0.95 %) is relatively higher than 4.01 ± 0.16 g/100g reported in leaves of *P. americana* (Arukwe *et al.*, 2012). Dietary fat increases the palatability of food by absorbing and retaining flavours (Antia *et al.*, 2006).

Table 1: Proximate composition of the plants

Sample	MC (%)	Ash (%)	CF (%)	CP (%)	Ether (%)	N (%)	CHO (%)	CAL (Kcal)
<i>M. indica</i>	20.10 ± 0.90^a	8.24 ± 0.99^a	10.60 ± 0.95^a	16.25 ± 1.00^b	4.30 ± 0.95^a	2.60 ± 0.95^a	60.61 ± 0.95^b	346.14 ± 1.01^c
<i>P. americana</i>	21.74 ± 0.99^a	5.60 ± 1.10^b	6.58 ± 1.01^b	17.94 ± 0.99^a	3.84 ± 0.99^a	2.87 ± 1.01^a	66.04 ± 1.00^a	370.47 ± 1.01^a
<i>A. muricata</i>	16.58 ± 1.00^b	5.96 ± 1.01^b	7.24 ± 1.01^b	15.74 ± 1.01^b	2.98 ± 1.00^a	2.52 ± 0.99^a	65.56 ± 1.00^a	352.02 ± 1.01^b

Mean \pm standard deviation of triplicate determinations; Values on the same column with different superscripts are significantly different ($p < 0.05$).

The highest content of ash and crude fiber were obtained in the leaves of *M. indica* (8.24 ± 0.99 and 10.60 ± 0.95 %) and these values are significantly ($P < 0.05$) higher than their corresponding values in *A. muricata* (5.96 ± 1.01 and 7.24 ± 1.01 %) and *P. americana* (5.60 ± 1.10 and 6.58 ± 1.01 %), respectively. The highest content of ash and crude fiber in leaves of *M. indica* may be attributed to the ability of the plant to synthesize these proximate substances more than *A. muricata* and *P. americana*. The highest value of ash (8.24 ± 0.99 %) and crude fiber (10.60 ± 0.95 %) obtained in this study is lower than values reported in *Vernonia colorate* (15.86 %) and *Moringa oleifera* (15.09 %) (Antia *et al.*, 2006; Lockeet *et al.*, 2000) as well as 19.38 ± 4.34 and 38.40 ± 5.12 g/100g obtained in *P. americana* leaves (Arukwe *et al.*, 2012) and 20.05 % obtained in leafy vegetable commonly consumed in Nigeria such as *Talinum triangulare* (Akindahunsi and Salawu, 2005). The result therefore suggests a high deposit of proximate composition in *M. indica* leaves. Fiber cleanses the digestive tract by removing potential carcinogens from the body, prevents the absorption of excess cholesterol and adds bulk to the diet and prevents the intake of excess starchy food (Mensah *et al.*, 2008) and may therefore guard against metabolic conditions such as hypercholesterolemia

and diabetes mellitus (Usunobun *et al.*, 2015; Henry, 2004). Dietary fiber has a positive effect in the management of diabetes by controlling post-prandial hyperglycemia, delays gastric emptying or increase the viscosity of gastro-intestinal tract content thereby suppressing digestion of carbohydrate and delays its absorption (Usunobun *et al.*, 2015). The substantial amount of fiber in *Annona muricata* leaves shows that they can help in keeping the digestive system healthy and functioning properly. Fiber aids and speeds up the excretion of waste and toxins from the body, preventing them from sitting in the intestine or bowel for too long, which could cause a build-up and lead to several diseases (Hunt *et al.*, 1980). Adequate intake of dietary fiber can lower the serum cholesterol level, risk of coronary heart disease, hypertension, constipation, diabetes, colon and breast cancer (Ishida *et al.*, 2000; Rao and Newmark, 1998). The Recommended Daily Allowance (RDA) of fiber for children, adults, pregnant and lactating mothers are 19-25, 21-38, 28 and 29 g, respectively (Jimoh *et al.*, 2011).

The highest content of crude protein and calorific value were obtained in the leaves of *P. americana* (17.94 ± 0.99 % and 370.47 ± 1.01 KJ) and these values

are significantly ($P < 0.05$) higher than the values obtained in *M. indica* leaves (16.25 ± 1.01 % and 346.14 ± 1.01 KJ) and *A. muricata* (15.74 ± 1.01 % and 352.02 ± 1.01 KJ). The highest value of crude protein (17.94 ± 0.99 %) in leaves of *P. americana* is higher than protein value of *Momordica foecide* (4.6 %) leaves consumed in Nigeria and Swaziland, *Lesianthera africanas* (13.1 %) (Hassan and Umar, 2006; Ogle and Grivetti, 1985; Isong and Idiong, 1997) but lower than *Amaranthus candatus* (20.5% DW), *Piper guineeses* (29.78 % DW) and *T. triangulare* (31.00 % DW) (Antia *et al.*, 2006; Akindahunsi and Salawu, 2005; Etuk *et al.*, 1998), and 25.54 ± 2.52 g/100g reported in *P. americana* leaves (Arukwe *et al.*, 2012). High amount of protein is essential for animal growth and increased milk production (Tangka, 2003). Plant proteins are a source of food nutrient especially for the less privileged population in developing countries including Nigeria and proteins are one of the macromolecule and it is an alternate energy source when other energy sources are in short supply (Usunobun *et al.*, 2015). Food protein are building block units and it is needed to produce vital hormones, important brain chemicals, antibodies, digestive enzymes, and necessary elements for the manufacture of DNA (Usunobun *et al.*, 2015; Bailey, 2008).

The highest value of CHO was obtained in *P. americana* (66.04 ± 1.00 %) and the value is statistically equal ($P > 0.05$) with the value obtained in *A. muricata* (65.56 ± 1.00 %) but significantly ($P < 0.05$) higher than 60.61 ± 0.95 % observed in *M. indica*. The value of CHO obtained in *P. americana* (66.04 ± 1.00 %) in this study is lower than 75.0% DW reported for *Corchorus tridens* and 82.8 % for sweet

potatoes leaves (Asibey-Berko and Taiye, 1999) but substantially higher than 7.34 ± 0.41 g/100g reported in *P. americana* by Arukwe *et al.* (2012). Thus, the carbohydrate content contributes to the energy value in *Annona muricata*. Carbohydrates are essential for the maintenance of life in both plants and animals and also provide raw materials for many industries (Ebun-Oluwa and Alade, 2007). Carbohydrates produced by plants are one of the three main energy sources in food, along with protein and fat. When animals eat plants, energy stored as carbohydrates is released by the process of respiration, a chemical reaction between glucose and oxygen to produce energy, carbon dioxide, and water (Usunobun *et al.*, 2015). Glucose is also used by animal cells in the production of other substances needed for growth (Westman, 2002).

Vitamin content in leaves of Mangifera indica, Persea americana and Annona muricata: The result of the vitamin content in leaves of the three plants species (*M. indica*, *P. americana*, and *A. muricata*) investigated in this study is summarized in Table 2. The result revealed the presence of ascorbic acid, niacin, riboflavin, thiamine and beta-carotene in leaves of the plant species. The highest content of ascorbic acid and riboflavin was obtained in the leaves of *M. indica* (13.20 ± 0.90 and 0.21 ± 0.01 Mg) and these values are significantly ($P < 0.05$) higher than their corresponding values in *P. americana* (8.80 ± 0.10 and 0.15 ± 0.01 Mg) and in *A. muricata* (8.36 ± 1.01 and 0.17 ± 0.01 Mg), respectively. In this study, the highest value of ascorbic acid obtained in *M. indica* is lower than 29.92 ± 0.11 Mg/100g reported in *M. indica* by Okwu and Ezenagu (2008) and 38.16 ± 1.94 in *A. muricata* (Usunobun *et al.*, 2015).

Table 2: Vitamin content of the plants

Sample	Ascorbic (mg)	Niacin (mg)	Riboflavin (mg)	Thiamine (mg)	β -carotin (mg)
<i>M. indica</i>	13.20 ± 0.90^a	0.38 ± 0.00^b	0.21 ± 0.01^a	0.48 ± 0.01^b	22.60 ± 0.01^c
<i>P. americana</i>	8.80 ± 0.10^b	0.29 ± 0.01^c	0.15 ± 0.01^c	0.52 ± 0.01^a	115.50 ± 0.01^a
<i>A. muricata</i>	8.36 ± 1.01^b	0.41 ± 0.01^a	0.17 ± 0.01^b	0.39 ± 0.01^c	43.25 ± 0.99^b

Mean \pm standard deviation of triplicate determinations; Values on the same column with different superscripts are significantly different ($p < 0.05$).

Ascorbic acid (vitamin C) as an antioxidant helps to prevent cell damage caused by free radicals in the body, thus, reducing the rate of aging and development of heart disease, cancer and arthritis as well as optimizes the body's immune function and enhances wound healing (Iqbal *et al.*, 2004). Riboflavin (vitamin B2) is needed to break down proteins into amino acids, fats, and carbohydrates in the form of glucose, hence, convert nutrients from food into usable bodily energy that helps to maintain a healthy metabolism but its (riboflavin) poor status interferes with iron handling and contributes to the etiology of

anemia when iron intake is low while its deficiency has been implicated as a risk factor for cancer (Powers, 2003). The National Diet and Nutrition Survey of young people aged 4-18 years (Gregory and Lowe, 2000) reported a high prevalence of poor riboflavin status, determined biochemically, among adolescent girls in the United Kingdom. The highest content of thiamine and Beta-carotene were obtained in leaves of *P. americana* (0.52 ± 0.01 and 115.50 ± 0.01 Mg) and these values are significantly ($P < 0.05$) higher than their corresponding values obtained in *M. indica* (0.48 ± 0.01 and 22.60 ± 0.01 Mg) and *A. muricata*

(0.39±0.01 and 43.25±0.99 Mg), respectively. The highest value of thiamine in *P. americana* (0.52±0.01 Mg) in this study is relatively higher than 0.45 ± 0.11 Mg/100g observed in *M. indica* (Okwu and Ezenagu, 2008). Thiamine (vitamin B1) helps the body's cells change carbohydrates into energy, thus, making carbohydrate available for the brain and nervous system, plays a role in muscle contraction and conduction of nerve signals and its deficiency is associated with neurological problems, including cognitive deficits and encephalopathy (Gibson *et al.*, 2016) The highest content of niacin was recorded for *A. muricata* (0.41±0.01 Mg) and the value is significantly ($P<0.05$) higher than their corresponding values obtained in *M. indica* (0.38 ± 0.00 Mg) and *P. americana* (0.29 ± 0.01 Mg). The highest value of niacin (0.41 ± 0.01 Mg) is lower than 0.75 ± 0.20 Mg/100g reported in *M. indica* (Okwu and Ezenagu, 2008). Niacin (vitamin B3) helps the body break down carbohydrates, fats and proteins into energy, plays a role in gland and liver function, removal of harmful chemicals from the liver, relieve of migraine headache, circulation problems and dizziness, and reducing diarrhea associated with cholera but its (niacin) deficiency result to depression, pellagra, canker sores, poor circulation and fatigue (Prousky and MSc, 2010).

Mineral content (%) in leaves of Mangifera indica, Persea americana, and Annona muricata: The result of the mineral content in leaves of *M. indica*, *P. americana*, and *A. muricata* is summarized in Table 3. The result show that the highest content of Ca (4.41±0.01 %) and P (0.40±0.01 %) were obtained in the leaves of *M. indica* and these values were significantly ($P<0.05$) higher than values obtained in leaves of *A. muricata* (4.20±0.01 and 0.28±0.99 %) and *P. americana* (3.61±0.01 and 0.38±0.01 %). The highest value of Ca and P in leaves of *M. indica* may be attributed to the inherent ability of *M. indica* to take

up Ca and P from the soil more than *P. americana* and *A. muricata* and translocate them (Ca and P) to their leaves. The highest value of Ca (4.41±0.01 %) in *M. indica* is higher than 3.82±0.10 reported by Okwu and Ezenagu (2008) but the value of P (0.40±0.01 %) in *M. indica* is lower than 0.78±0.10 reported by Okwu and Ezenagu (2008) in South east Nigeria. Calcium is necessary for the coagulation of blood, proper functioning of the heart and nervous system and the normal contraction of muscles as well as aid in the formation of bones and teeth (Usunobun and Okolie, 2017). Phosphorus functions as a structural component of bones and teeth and DNA/RNA and enables the bipolarity of lipid membranes and circulating lipoproteins (Calvo and Lamberg-Allardt, 2015). The highest content of Mg (1.70±0.01 %) was obtained in leaves of *A. muricata* while the highest content of K (0.60±0.01 %) was obtained in leaves of *P. americana*. The highest value of Mg (1.70±0.01 %) in this study is well below 96.19±8.01% reported in leaves of *A. muricata* (Usunobun and Okolie, 2017). Similarly, the highest value of K (0.60±0.01 %) in *P. americana* is well below 148.92±0.12 obtained in leaves of *P. americana* by Arukwe *et al.* (2012). This may be attributed to differences in location vis-a-vis the type of species investigated as well as soil quality. Magnesium is a component of chlorophyll and it is an important mineral element in connection with ischemic heart disease and calcium metabolism in bones (Ishida *et al.*, 2000). Potassium is the principal cation in intracellular fluid and functions in acid-base balance, regulation of osmotic pressure, conduction of nerve impulse, muscle contraction particularly the cardiac muscle, cell membrane function and Na⁺/K⁺-ATPase. Potassium deficiency affects the collecting tubules of the kidney, resulting in the inability to concentrate urine, and also causes alterations of gastric secretions and intestinal motility (Streeten and Vaughan-Williams, 1952).

Table 3: Mineral content of the plants

Sample	Ca (%)	Mg (%)	K (%)	Na (%)	P (%)	N (%)
<i>M. indica</i>	4.41±0.01 ^a	1.58±0.01 ^b	0.55±0.01 ^b	0.23±0.01 ^a	0.40±0.01 ^a	2.60±0.01 ^a
<i>P. americana</i>	3.61±0.01 ^c	1.28±0.01 ^c	0.60±0.01 ^a	0.19±0.01 ^b	0.38±0.01 ^b	2.87±1.01 ^a
<i>A. muricata</i>	4.20±0.01 ^b	1.70±0.01 ^a	0.49±0.01 ^c	0.23±0.01 ^a	0.28±0.99 ^c	2.98±0.99 ^a

Mean ± standard deviation of triplicate determinations; Values on the same column with different superscripts are significantly different ($p<0.05$).

The content of Na is statistically the same ($P>0.05$) in leaves of *M. indica* (0.23±0.01 %) and *A. muricata* (0.23±0.01 %) but the values are significantly ($P<0.05$) higher than 0.19±0.01 % obtained in leaves of *P. americana*. The highest value of Na (0.23±0.01 %) obtained in this study is relatively lower than 0.38±0.11 mg/100 g reported in leaves of *M. indica* (Okwu and Ezenagu, 2008) but substantially lower

than 694.86±10.65 % in leaves of *A. muricata* (Usunobun and Okolie, 2017). Sodium is involved in the regulation of plasma volume, acid-base balance, nerve and muscle contraction (Akpanyung, 2005). Sodium remains one of the major electrolytes in the blood and without sodium, the body may not be hydrated, as it would dry up. The highest content of N (2.98±0.99 %) is recorded for *A. muricata* but the

value is not significantly ($P>0.05$) different from the values obtained in leaves of *P. americana* ($2.87\pm 1.01\%$) and *M. indica* ($2.60\pm 0.01\%$). Since N plays an important role in photosynthesis, its (N) deficiency would result in lower dry matter yield and foliage and this can affect the quantity of phytochemical and proximate composition of the leaves. Adequate level of macronutrients is required for continuous generation of immune cells in bone marrow and the clonal expansion of lymphocytes in response to antigenic stimulation (Chandra, 1990). Majority of antioxidant enzymes or defense systems of the body and processes involved in lipid metabolism in general make use of mineral elements, and an imbalance in these elements usually leads to nutritional disorders and complications of nutritionally related diseases for example diabetes (Chandra, 1990). The presence of most of the mineral elements in *M. indica* and *P. americana* leaves could, therefore, be relevant in exerting anti-hyperglycemic activity and the amelioration of the attendant macro-vascular complications. The broad distribution of phytochemicals, minerals and antioxidants in *Annona muricata* leaves sampled from Umuanunu support, as well as provide a basic rationale for their use in folk medicine. From the foregoing, this work therefore indicates that *Annona muricata* leaves, besides serving as good source of pharmacologically active phytochemicals, antioxidants and thus free radical scavengers may also be useful as supplements in human and animal nutrition as they are biodegradable, environmentally friendly, and cost-effective and would meet the demand of Nigeria as a country to go green.

Phytochemical content (%) in leaves of Mangifera indica, Persea americana, and Annona muricata: The result of the phytochemical content in leaves of *M. indica*, *P. americana*, and *A. muricata* is summarized in Table 4. The result indicate that the highest content of tannins ($1.38\pm 0.01\%$), flavonoid ($0.85\pm 0.01\%$), and phenol ($0.37\pm 0.01\%$) were obtained in leaves of *M. indica* and these values were higher than their corresponding values in leaves of *P. americana* (0.51 ± 0.01 , 0.44 ± 0.01 , and $0.04\pm 0.01\%$) and *A. muricata* (0.34 ± 0.01 , 0.26 ± 0.01 , and $0.18\pm 0.01\%$).

The high content of tannins, flavonoid, and phenol in leaves of *M. indica* may be attributed to the inherent ability of the plant to generate these phytochemicals more than *P. americana* and *A. muricata*. The secretion of phytochemicals vary from one to another (Tariq and Reyaz, 2013). Hence, some plants produce more of these compounds while others synthesizes minimal quantity. Herbs that have tannins are used for the treatment of intestinal disorders like dysentery and diarrhea (Bajai, 2001), hence supporting the reasons why *M. indica*, *P. americana*, and *Annona muricata* is used for therapeutic purposes by the people of Umuanunu. Tannins are known to be useful for the prevention of cancer, treatment of inflamed or ulcerated tissues. (Adegboye *et al.*, 2008; Okwu and Emineke, 2006; Li *et al.*, 2003) and exhibit cytotoxic effects and growth inhibition, thus, making it suitable as tumor inhibiting agent (Asl and Hossein, 2008; Akindahunsi and Salawu, 2005). Flavonoids and phenolics are free radical scavengers that prevent oxidative cell damage, and have strong anticancer activities (Ugwu *et al.*, 2013; Pourmorad *et al.*, 2006) and they might induce mechanism that affect cancer cells and inhibit tumor invasion (Rafat *et al.*, 2008). These activities is attributed to their ability to neutralize free radicals (Ugwu *et al.*, 2013; Omale and Okafor, 2008; Pourmorad *et al.*, 2006), the presence of conjugated ring structures and carboxylic group that inhibit lipid peroxidation (Rice-Evans *et al.*, 1997). The highest content of phytate ($0.30\pm 0.01\%$) was obtained in leaves of *A. muricata* and the value was significantly ($P<0.05$) higher than values obtained in *M. indica* ($0.27\pm 0.01\%$) and *P. americana* ($0.26\pm 0.01\%$). The low phytate in the species studied suggest that the plants will not impact negatively on protein and lipid utilization in man (Kumar *et al.*, 2010). Similarly, the highest content of alkaloid ($2.26\pm 0.01\%$) and saponin ($1.33\pm 0.01\%$) were recorded in leaves of *P. americana* and these values are significantly ($P<0.05$) higher than their corresponding values in leaves of *M. indica* (1.96 ± 0.01 and $1.24\pm 0.01\%$) and *A. muricata* (1.48 ± 0.01 and $1.28\pm 0.01\%$). The high content of alkaloid and saponin in leaves of *P. americana* may be attributed to the inherent ability of the plant to produce these phytochemicals more than *A. muricata* and *M. indica*.

Table 4: Phytochemical (%) content of the plants

Sample	Tannin	Alkaloid	Flavonoid	Phenol	Saponin	Phytate
<i>M. indica</i>	1.38 ± 0.01^a	1.96 ± 0.01^b	0.85 ± 0.01^a	0.37 ± 0.0^a	1.24 ± 0.01^c	0.27 ± 0.01^b
<i>P. americana</i>	0.51 ± 0.01^b	2.26 ± 0.01^a	0.44 ± 0.01^b	0.04 ± 0.00^c	1.33 ± 0.01^a	0.26 ± 0.01^b
<i>A. muricata</i>	0.34 ± 0.01^c	1.48 ± 0.01^c	0.26 ± 0.01^c	0.18 ± 0.01^b	1.28 ± 0.01^b	0.30 ± 0.01^a

Mean \pm standard deviation of triplicate determinations; Values on the same column with different superscripts are significantly different ($p<0.05$).

The ability to generate bioactive compounds or active ingredients differ from one plant species to another.

Alkaloids are beneficial chemicals to plants serving as repellent to predators and parasites (Usunobun *et al.*,

2015). Several alkaloid containing medicinal plants were used by the early man as pain relievers, as recreational stimulants or in religious ceremonies to enter a psychological state to achieve communication with ancestors (Heinrich *et al.*, 2004; Gurib-Fakin, 2006). Saponin in medicinal plants are responsible for most biological effects related to cell growth and division in humans and have inhibitory effect on inflammation (Okwu and Emineke, 2006, Liu and Henkel, 2002; Just *et al.*, 1998). Saponins have the property of precipitating and coagulating red blood cells (Yadav and Agarwala, 2011). Some of the characteristics of saponins include formation of foams in aqueous solutions, hemolytic activity, cholesterol binding properties and bitterness (Okwu, 2004; Sodipo *et al.*, 2000). Saponin is very useful in formulation of drugs in food, drinks and beverage industries as foaming agents (Fenwick *et al.*, 1983), as antioxidants, preservatives and flavouring agents (You *et al.*, 1993; Fenwick *et al.*, 1983). Indeed, *M. indica* (ascorbic acid, riboflavin), *P. americana* (thiamine, beta-carotene), and *A. muricata* (niacin) leaves are reservoirs for free radical scavenging molecules such as vitamins and their utilization should be strongly recommended for good health. The plants are also rich in phytochemicals (tannins, flavonoids, phenol, alkaloid, and saponin), proximate (ash, crude fibre, crude protein, calorific value, carbohydrate) which are basically rich in antioxidant activities, and minerals (Ca, P, Na, K, Mg, and Na). Generally, the content of tannins, flavonoids, and phenols in *M. indica* were > *P. americana* > *A. muricata* while alkaloid and saponin in *P. americana* were > *M. indica* > *A. muricata*. Consequently, the utilization of *M. indica* and *P. americana* by the local people will boost their immunity against some diseases.

Conclusion: The study show that the three plant species (*M. indica*, *P. americana*, and *A. muricata*) contain varying quantity of phytochemicals, vitamins, minerals and proximate composition in their leaves. The content of tannins, flavonoids, phenols, ascorbic acid, riboflavin, Ca, and P mostly occurred in *M. indica* while alkaloid, saponin, thiamine, beta-carotene, and K were highest in *P. americana*. The use of *M. indica* and *P. americana* leaves should be strongly recommended for the good health of inhabitants of the study area as well as other Nigerians.

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