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MINERALS AND PHYTOCHEMICAL CONSTITUENTS OF *MANGIFERA INDICA* SEED KERNEL OBTAINED FROM EASTERN NIGERIA

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ABSTRACT: *The analysis on the seed kernel of Mangifera indica was carried out in the laboratory with the view of determining its phytochemical constituents as well as its mineral constituent. The sample was screened for its phytochemicals and the minerals determination was performed by the ashing method with the aid of a muffle furnace and bulk scientific atomic absoption spectrometer (AAS). Results obtained revealed that the seed kernel is rich in phytonutrients as it contains alkaloids, tannins, phenols, saponins, glycosides, flavonoids and steroids. Similarly, the mineral composition revealed that the seed kernel contained essential minerals such as; Potassium, 0.013mg/g, Sodium 0.788mg/g, Magnesium 0.005mg/g, Calcium 0.009mg/g, Manganese 0.039mg/g, Iron 0.101mg/g, Zinc 0.058mg/g, Copper 0.0016mg/g, Cobalt 0.010mg/g and Chromium 0.008mg/g. These results not only highlighted the usefulness of this sample but gives credence to fact that the Mangifera indica seed kernel could act as a veritable food supplements for both domesticated animals and man.*

KEYWORDS: Mangifera Indica, Phytochemicals, Minerals, Seed Kernel

INTRODUCTION

Mangifera indica commonly known as mango plant is a tree found in every part of Nigeria. It is basically cultivated for it fruits which are usually used as food. In Nigeria the juicy part of the fruit is consumed and the hard shell containing the kernel is discarded. Usually the hard shell containing the kernel is of no value as it is found littered everywhere especially during the mango harvest season. The mango belongs to genus *Mangifera*, which consists of numerous species of tropical fruits in the family of *Anacardiaceae. Mangifera indica* is native to India and Southeast Asia where it has been cultivated over the years for the good qualities of the fruits. Currently, mango is also grown in Central America, Africa and Australia. It is found in the wild in Nigeria and cultivated varieties have been introduced to other warm regions of the world. It is the largest fruit-tree in the world, capable of a height of one-hundred feet and an average circumference of twelve to fourteen feet, sometimes reaching twenty. This plant contain high level of phytonutrients as its fruit, root, bark and leaves are used by traditional medicine practioners to cure various diseases and disorders. Its woody trunk is also used as fuel and for furniture making. Extracts from the seed, leaves and roots are specially use in control of diarrhoea, syphilis, ulcer, diabetes, kidney stone, sunstroke, tuberculosis, intestinal disorder, blood purification, nasal bleeding, ameobiosis, in piles and in heart diseases. Mango contain chemical like Gallic acid, Mangeferin, ellagic acid, soluble sugar, protein and oil.[1].In remote area, villagers depend upon the folk medicine

and house hold remedies. Mango is one of wild plants growing naturally on west land which is very important to cure several diseases of rural peoples and domesticated animals; It is used in the worship by Hindu as a holy plant. The fruit, root, bark, flower and leaves are used by tribals and Indians to cure various diseases and disorders, the Fruit contain Gallic acid, gallotanin, mangiferin, elagic-acid sugars, protein, carotene xanthophylls, vitamin A and C. its Seed contain gallotanin, stearic acid, methyl alcohol. Its Leaves contain mangiferin, glucose, galactose, xylose, gallic acid, glycoside and several other secondary metabolites [2]

Various parts of plant are used as a dentifrice, antiseptic, astringent, diaphoretic, stomachic, vermifuge, tonic, laxative and diuretic and to treat diarrhea, dysentery, anaemia, asthma, bronchitis, cough, hypertension, insomnia, rheumatism, toothache, leucorrhoea, haemorrhage and piles. All parts are used to treat abscesses, rabid dog or jackal bite, tumour, snakebite, stings, heat stroke, miscarriage, anthrax, blisters, wounds in the mouth, tympanitis, colic, diarrhea, indigestion, bacillosis, bloody dysentery, liver disorders, excessive urination, tetanus and asthma. Various Chemical constituents are found in different species of mango. These constituents are always of an interest. Especially the polyphenolics, flavonoids, triterpenoids are the different chemical constituents of the plant[3].Mangiferin a (xanthone glycoside) is major bioactive constituent, isomangiferin, tannins and gallic acid derivatives also are bioactive.. The bark is reported to contain protocatechic acid, catechin, mangiferin, alanine, glycine, γ-aminobutyric acid, shikimic acid and the tetracyclic triterpenoids. Mango seed kernel extract is one of the most important phytomedicine, traditionally claimed for its anti-diarrhoeal activity. It is employed to kill abdominal worms, cure for vomiting, diarrhoea and hyper-acidity.it has been reported recently that this extract can be used as an anti-diarrhoeal agent. The extract inhibited the growth of virulent strains of *Salmonella*. It may due to prevention of biofilm and action on pathogens.[4] Mango seed kernels are rich in carbohydrates, proteins, fat, minerals and vitamins, hence can be used as potential source of nutrients for human and animal feed. Mango seed kernel also contains considerable amount of phenolic compound as well as high levels of antioxidant. The plant extracts have been shown to exhibit antimicrobial activity against *Staphylococcus aureus, Bacillus subtilis, Escherichia coli, and Pseudomonas aeruginosa* [5]. Mango is a fruit that should be included in everyone's diet for its multifaceted biochemical actions and health-enhancing properties. Mangifera indica L. (mango) is known as "the king of fruits" because it is the most popular fruit in tropical region. There are different mango cultivars (Kensington Pride, Tommy Atkins, Osteen, Maya, Kent, Irwin, Glenn,and Keitt varieties) [6]. Chemical analysis of mango pulp provides evidence that it has a relatively high content in calories and is an important source of potassium, fiber, and vitamins. Like mango mesocarp and exocarp, mango seed kernels are also equally rich in polyphenols with potent antioxidant activity. Mango seed kernels contain tannin, gallic acid, coumarin, caffeic acid, vanillin, mangiferin, ferulic acid, and cinnamic acid. [7] In traditional medicine the different parts of the mango tree (fruit pulp, extracts of fruit kernel, leaves, and stem bark) are used for their health properties [8].

MATERIALS AND METHOD

SAMPLE COLLECTION AND PREPARATION

Mangifera indica fruits were bought from Ihiagwa market in Owerri north L.G.A Imo state Nigeria. The seed kernels were obtained from the fruit of the plant after removal of both the

mesocap and the epicap. They were wash and sliced into bits and room dried for a period of 8 weeks, after which the seeds were ground to powder and stored in air tight container. [9]

Phytochemical screening of the sample

Frothing test for Saponins

This test is based on the ability of the saponins to produce froth in aqueous solution. 5g of the Leaf sample was weighed into a test tube and 50cm^3 of water was added and extracted after 4hours. The water extract was shaken vigorously in a conical flask. The production of a stable froth indicates the presence of saponins in the sample

Test for Flavonoids

5g of the sample was soaked with 50cm^3 of water and then filtered. To the filtrate was added drops of ammonia and 3cm^3 of concentrated H_2SO_4. A yellow precipitate which disappears on storage indicates the presence of flavonoids.

Test for Alkaloids

5g of the sample was extracted using 20% acetic acid in ethanol .5cm^3 of the extract was treated with Wagner's reagent (iodine crystals and KI). A yellowish-brown precipitate indicates the presence of alkaloids.

Test for Tannins

5g of the sample was weighed into a beaker and 50cm^3 of water was added and allowed to soak properly for 4 hours and extracted. The extract was treated with drops of ferric chloride. A blue-black precipitate indicates the presence of tannins.

Test for Steroids

5cm^3 of the water extract was treated with concentrated H_2SO_4 in acetic anhydride. The formation of a blue-green color indicates the presence of steroids.

Test for Phenols

20cm^3 of the water extract was treated with 5cm^3 of concentrated sulphuric acid and drops of sodium nitrate ($NaNO_3$). 2cm^3 of sodium hydroxide was added to the mixture. A blue precipitate indicated the presence of phenols.

Test for Glycosides

20cm^3 of the water extract was treated with Fehling solutions of A and B in equal amount and boiled. A brownish red precipitate indicates the presence of glycoside.

Determination of Minerals by Ashing Method

5.0g of dried plant sample was weighed into a petridish. The sample was ashed for six hours at about 500^{o}C in a muffle furnace. The ash was cooled and extracted with 2-3cm^3 of hydrochloric acid (SG 1-4) and evaporated to dryness. The residue was re-extracted with 20ml of 25% V/V HCl and transferred into a 100cm^3 volumetric flask and diluted to the mark

with water. The elementary determinations were achieved by direct aspiration of the ash solution into the flame of the AAS (Atomic Absorption Spectrophotometer) into the spray chamber through the capillary tube and nebulizer. Each metal was determined by using the metal's hollow cathode lamp. The readings were taken in triplicate and the mean value was obtained.

RESULTS AND DISCUSSION

The results obtained from the phytochemical screening of the mango seed kernel are enumerated in table 1 below. The seed kernel is rich in phytonutrients as it contains alkaloids, tannins, phenols, saponins, glycosides, flavonoids and steriods

Table 1: Results of Preliminary Phytochemical Analysis for Mangifera Indica Seed Kernel.

Sample	Phytochemical	Result
A	Alkaloids	+
B	Tannins	+
C	Phenols	+
D	Saponins	+
E	Glycosides	+
F	Flavonoids	+
G	Steroids	+

Alkaloids are vast and vary alot in their activity when ingested by man and livestock. Some alkaloids are useful and important in medicine and constitute most of the valuable drugs currently used by humans. They are reported to have marked physiological effect on animals [10].

Phenolics are broadly distributed in plants and are the most abundant secondary metabolites of plants. Plant phenolics have drawn increasing attention due to their potent anti-oxidant properties and their marked effects in the prevention of various oxidative stress associated diseases such as cancer. In the last few years, the identification and development of phenolic compounds or extracts from different plants has become a major area of health and medical-related research

Flavonoids have been shown to be highly effective scavengers of most oxidizing molecules [11]. In addition, tannin was found in the plant at a concentration range. Plant leaves with high tannin content have been used successfully as hops alternative in beer [12]

Flavonoids are polyphenolic compounds based on a C_{15} ($C_6C_3C_6$) framework. The heterocyclic six-membered C-ring is sometimes replaced by a 5-membered ring. The oxidation state of the C-rings is used to classify flavonoids into different categories such as Flavan-3-ols, flavonones, flavones, isoflavones and flavonols. Flavonoids are the major nutraceutical ingredients that are in plants. The best described property of almost every group of flavonoids is their capacity to act as anti-oxidants. The flavones seem to be the most powerful flavonoid for protecting the body against reactive oxygen species (ROS).

Antibacterial activity has been displayed by a number of flavonoids, Quercetin has been reported to completely inhibit the growth of *Staphylococcus aureus*[13]. Flavonoid also possesses anti-inflammatory and analgesic effect as well as anti-ulcerogenic activity [14]. The seed kernel of *Mangifera indica* is a remedy for Saponins are foam forming in nature and have been implicated as a bioactive antibacterial remedy for gonorrhea and jaundice [15, 16]. This is probably due to antibacterial action of saponins component of plant [17]. Saponins are also utilized in foods that need sustained foam volume such as ice-creams. In plants, saponins may serve as anti-feedants to protect the plant against microbes and fungi. Some plant saponins may enhance nutrient absorption and aid in animal digestion. Saponins have been used as a pharmacological and immunological agent that modifies the effect of other agents in vaccines. Saponins from plants have been shown to significantly augment the cytotoxicity of immunotoxins and other target toxins directed against human cancer cells. [18]

Tannins are astringent, bitter plant polyphenol compounds that bind to and precipitate proteins and various other organic compounds including amino acids and alkaloids. The tannin compounds are widely distributed in many species of plants where they play a role in protection from predation and perhaps also as pesticides and in plant growth regulation [19]. The astringency from tannin is what causes the dry puckery feeling in the mouth following the consumption of unripe fruits or red wine. Tannins are important ingredients used in process of making tannin leather. Medicinally, tannins are used as anti-diarrhea, hemostatic and anti-hemorrhoid compounds. [20]

The presence of Phenolic compounds in mangifera indica seed kernel indicates that this plant might be an anti-microbial agent. This is because phenols and phenolic compounds have been extensively used in disinfections and remains the standard with which other bactericides are compared. Phenolic compounds act as electron donors and are readily oxidized to form phenolate ions, this gives rise to protonated phenol which is used as a cleaning agent. Phenols have antioxidant properties. The presence of phenol further indicates that the extract could act as anti-inflammatory, anti-clotting, immune enhancers and hormone modulators. [21]

Glycosides are molecules in which a sugar is bound to another functional group via a glycosidic bond. Glycosides play numerous important roles in living organisms. Many plant store chemicals in form of inactive glycosides. Many such plant glycosides are used as medications. Some glycosides have shown some evidence of pharmacological effects in patients with hypertension or with type-2 diabetes. [22]

The mineral composition showed that the seed kernel of *Mangifera indica* contained essential minerals such as; Potassium, 0.013mg/g, Sodium 0.788mg/g, Magnesium 0.005mg/g, Calcium 0.009mg/g, Manganese 0.039mg/g, Iron 0.101mg/g, Zinc 0.058mg/g, Copper 0.0016mg/g, Cobalt 0.010mg/g and Chromium 0.008mg, table 2 below;

TABLE 2: Mineral Composition of *Mangifera Indica* Fruit Nut

Mineral Elements	**Composition (Mg/g)**
Potassium	0.013,
Sodium	0.788
Magnesium	0.005
Calcium	0.009
Manganese	0.038

Iron	0.101
Zinc	0.058
Copper	0.016
Cobalt	0.010
Chromium	0.008

Sodium and potassium are essential for human health. They are important ions in the body and are associated with many physiologic and pathophysiologic processes. Concentrations of sodium in the blood that exceed or do not reach the normal value range are called hypernatremia or hyponatremia, respectively. Sodium is the most common electrolyte in extracellular fluid, its main role is in controlling water distribution and fluid balance in the body. Water follows sodium, so high levels of sodium in a fluid compartment take water with it. Other functions of sodium include: promote transmission of nerve impulses, activate several enzymatic reactions, assist with regulation of acid-base balance, promote myocardial, skeletal, and smooth muscle contractility. Sodium levels are influenced by the antidiuretic hormone (ADH). Increased secretions of ADH causes more water to be reabsorbed in the kidneys and decreased ADH secretion allows more water to be excreted. Sodium levels are also influenced by aldosterone. High aldosterone levels promote the re-absorption of sodium in the kidneys' distal tubules. Acid –base balance is maintained by the sodium combining with chloride and bicarbonate ions. Sodium, which is mainly extracellular, works with potassium, mainly intracellular, to maintain the balance in intracellular and extracellular fluids through the sodium-potassium pump. This pump system has an important role in conducting impulses in muscle and nerve fibers. [23, 24]

Magnesium is an essential element is necessary for the biochemical functioning of numerous metabolic pathways. Inadequate magnesium in the body may impair biochemical processes dependent on sufficiency of this element. Magnesium is useful in the prevention and treatment of many common health conditions including; migraine headache, metabolic syndrome, diabetes, hyperlipidemia, asthma, premenstrual syndrome, pre-eclampsia, and various cardiac arrhythmias. Magnesium may also be considered for prevention of renal calculi and cataract formation, as an adjunct or treatment for depression, and as a therapeutic intervention for many other health-related disorders. Magnesium is effective for mild-to-moderate depression in adults. It works quickly and is well tolerated without the need for close monitoring for toxicity. Magnesium is the fourth most abundant mineral in the body. It has been recognized as a cofactor for more than 300 enzymatic reactions, where it is crucial for adenosine triphosphate (ATP) metabolism. Magnesium is required for DNA and RNA synthesis, reproduction, and protein synthesis. Moreover, magnesium is essential for the regulation of muscular contraction, blood pressure, insulin metabolism, cardiac excitability, vasomotor tone, nerve transmission and neuromuscular conduction. Imbalances in magnesium status might result in unwanted neuromuscular, cardiac or nervous disorders. Based on magnesium's many functions within the human body, it plays an important role in prevention and treatment of many diseases. Low levels of magnesium have been associated with a number of chronic diseases, such as Alzheimer's disease, insulin resistance and type-2 diabetes mellitus, hypertension, cardiovascular disease (e.g., stroke), migraine headaches,and attention deficit hyperactivity disorder (ADHD) [25, 26]

Calcium is an essential nutrient that has many functions in human health. Calcium is the most abundant mineral in the body with 99% found in teeth and bone. Only 1% is found in serum. Calcium metabolism involves other nutrients including protein, vitamin D, and phosphorus. Bone formation and maintenance is a lifelong process. Early attention to strong bones in childhood and adulthood will provide more stable bone mass during the aging years. Adequate calcium intake can reduce the risk of fractures, osteoporosis, and diabetes in some populations. The dietary requirements of calcium and other collaborative nutrients vary slightly around the world. Lactose intolerance due to lactase deficiency is a common cause of low calcium intake. [27,28]

Manganese is an important metal for human health, being absolutely necessary for development, metabolism, and the antioxidant system. Nevertheless, excessive exposure or intake may lead to a condition known as manganism, a neurodegenerative disorder that causes dopaminergic neuronal death and parkinsonian- like symptoms. it is required for growth, development, and maintenance of health. Routes of Manganese exposure are mainly through dietary intake, dermal absorption, and inhalation. Accordingly, the primary source of Manganese intoxication in humans is due to occupational exposure as in miners, smelters, welders, and workers in dry-cell battery factories. Significant neurological dysfunction has been associated with Manganese exposure, but the exact mechanisms underlying the neurotoxic effects of Manganese remain unclear {29,30}

Iron is an essential element for most life on earth, including human beings by participating in a wide variety of metabolic processes, including oxygen transport, DNA synthesis, and electron transport. Iron is needed for a number of highly complex processes that continuously take place on a molecular level and that are indispensable to human life; the transportation of oxygen around the body. Iron is required for the production of red blood cells (a process known as haematopoiesis), but it is also part of haemoglobin binding to the oxygen and thus facilitating its transport from the lungs via the arteries to all cells throughout the body. About 70% of the body's iron is bound to hemoglobin in red blood cells. The rest is bound to other proteins (transferrin in blood or ferritin in bone marrow) or stored in other body tissues. When red blood cells die, their iron is released and carried by transferring to the bone marrow and to other organs such as the liver and spleen. In the bone marrow, iron is stored and used as needed to make new red blood cells. Once the oxygen is delivered the iron (as part of haemoglobin) binds the carbon dioxide which is then transported back to the lung from where it gets exhaled. Iron is also involved in the conversion of blood sugar to energy. Metabolic energy is crucial for athletes since it allows muscles to work at their optimum during exercise or when competing. The production of enzymes (which play a vital role in the production of new cells, amino acids, hormones and neurotransmitters) also depends on iron, this aspect becomes crucial during the recovery process from illnesses or following strenuous exercise or competing. Iron is one of the important elements necessary for the metabolism of the human body. It is found in two forms, essential iron for normal function of the body and the reserve for times of needs. The essential iron is mostly haemoproteins and is present in haemoglobin or erythron and is the major part of the body iron. The other important position of essential iron is in enzymes required for mitochondrial function and DNA Synthesis The stored iron compounds, in the form of ferritin and haemosiderin are present mainly in the reticulo-endothelial system. Ferritin, a protein has an outershell and an inner core which contains the iron deposited as ferric hydroxy phosphate complex. The iron requirements are for growth, physiological losses and uterine losses due to menstruation and pregnancy.

Increased catecholamine levels in children leading to abnormal behaviour has been found associated with iron deficiency such. A number of metals as lead and cadmium enter the body via the iron absorptive mechanism. Occasionally iron overload develops in patients with liver disease especially cirrhosis [31,32,33]

Over 200 zinc metallo-enzymes exist in the human body. Of these, many e.g., carbonic anhydrase, alkaline phosphatase, carboxypeptidase A,B perform a variety of functions. However, zinc deficiency does not exert its effects through deficient function of these enzymes alone. Other important roles attributed to zinc include maintenance of adequate immune function and brain development. Zinc is crucial to the maintenance of satisfactory growth in childhood. Zinc deficiency has been shown to affect the function of human growth hormone by modulating with the function of the polypeptide hormone-receptor. A growth limiting mild zinc deficiency state has been described in young boys with short stature. The essential role of zinc in the maintenance of structure of bio-membranes, DNA and RNA synthesis and metabolism of essential fatty acids, makes it an extremely important micronutrient in pregnancy. A close association exists between zinc status and normal fetal growth. It is thus possible that postnatal low zinc intake, especially associated with poorly fortified formulae, may lead to hypozincemia and poor growth. Serum albumin and pre-albumin metabolism are also closely dependent on zinc status. [34, 35, 36, 37, 38, 39]

Copper is an important trace element that plays a very important role in the biochemistry of all living organisms and affects enzymes activity as a cofactor or as a fundamental structure of many metalloenzymes such as superoxide dismutase, ceruloplasmin, lysyl oxidase, cytochrome oxidase and tyrosinase. Copper is necessary for structural and catalytic properties of cuproenzymes. Copper deficiency is usually the consequence of decreased copper stores at birth, inadequate dietary copper intake, poor absorption, and elevated requirements induced by rapid growth, or increased copper losses. The manifestations of copper deficiency are anemia, neutropenia, and bone abnormalities. *Copper is essential for cellular respiration, free radical defence, neurotransmitter function and tissue biosynthesis. Excessive copper accumulation is toxic in all species as it leads to hepatic cirrhosis, hemolytic anemia and degeneration of basal ganglia.* *[40,41]*

Cobalt is a component of Vitamin B12, it induces erythropoietin and blocks iodine uptake by the thyroid. It has a role to play in methionine metabolism where it controls the transfer of enzymes like homocysteine methyltransferase. Deficiency produces cardiomyopathy, congestive cardiac failure, pericardial effusion, polycythemia,and thyroid enlargement. Cobalt is not easily absorbed from the digestive tract. It is stored in the red blood cells and the plasma, as well as in the liver, kidney,spleen, and pancreas. Cobalt has both beneficial and harmful effects on human health. Cobalt is beneficial for humans as it increases red blood cell production in healthy people, but only at very high exposure levels. Deficiency of cobalt also leads to fatigue, digestive disorders, and neuromuscular problems and decreased availability of B12. Vitamin B12 is a collective term for a group of cobalt-containing compounds known as corrinoids which are also known as cobalamins. The principal cobalamins are: cyanocobalamin, hydroxocobalamin and the two coenzyme forms: methylcobalamin and deoxyadenosyl cobalamin (adenosylcobalamin). Vitamin B12 is involved in the metabolism of every cell of the body, especially affecting DNA synthesis and regulation but also fatty acid synthesis and energy production. Vitamin B12 controls production of blood platelets and red and white blood cells, normal nerve cell activity –

formation of myelin basic protein, DNA and RNA replication, and production of the mood-affecting substance SAM (S-adenosyl-L-methionine). [39,42]

The importance of Chromium is for biosynthesis of glucose tolerance factor. The deficiency causes impairment of glucose tolerance while toxicity results in renal failure, dermatitis, and pulmonary cancer. Certain spices such as black pepper contain high concentrations of chromium. Chromium is excreted principally in the urine and in small quantities in the hair, sweat, and bile. The major route of elimination after absorption is fecal. Chromium is a human carcinogen primarily when inhaled or upon exposure in occupational settings. Lung cancer has been established as a consequence of hexavalent chromium exposure in smokers and nonsmokers. Hyperglycemia may be associated with raised plasma chromium and increased urinary excretion. Chromium is biologically active as part of an oligopeptide – chromodulin potentiating the effect of insulin by facilitating insulin binding to receptors at the cell surface, with chromium acting as a cofactor of insulin, Chromium activity in the organism is parallel to insulin functions. Absorbed Chromiun circulates in blood bound to the β-globulin plasma fraction and is transported to tissues bound to transferrin. [39,42.43]

CONCLUSION

Mangifera indica seed kernel is a very useful nutraceutical and should be included in our daily diet because of its phytochemical and essential mineral contents. Eating a diet made with this seed kernel will not only nourish the body but also help the body fight certain common human diseases

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