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Comparative Phytochemical, Proximate and Some Mineral Composition of the Leaves and Stem Bark of *Spondia mombin* (L. anacardiaceae)

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Authors' contributions

This work was carried out in collaboration between both authors. Author EDK designed the study, performed the statistical analysis, wrote the protocol and also wrote the first draft of the manuscript.

Author EOO managed the analyses of the study and literature searches. Both authors read and approved the final manuscript.

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ABSTRACT

Aims: Quantitative phytochemical, proximate and some mineral content of the leaves and stem bark of *Spondia mombin*, a plant commonly found in Southern Nigeria was studied with a view of ascertaining their nutritional potential and justification for their ethnopharmacological uses.

Place and Duration of Study: The study was conducted in the research Laboratory of the Department of Biochemistry, Federal University Otuoke, Bayelsa State, Nigeria, between February – March 2019.

Methodology: Proximate composition, quantitative phytochemical, and mineral contents of the samples were studied using standard analytical methods of the Association of Analytical Chemist (AOAC).

Results: Proximate analysis of the leaves and stem bark showed that there was no significant difference ($P > 0.05$) between the carbohydrate and protein content of both leaves and stem bark while the stem bark had significantly higher ($P < 0.05$) values for ash, fat and fibre when compared to the leaves which incidentally had significantly higher ($P < 0.05$) moisture content. Quantitative phytochemical analysis revealed that there was no significant difference ($P > 0.05$) between the

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leaves and stem bark with reference to tannin, oxalate and phytate, the stem bark, however, showed significantly higher ($P < 0.05$) values for alkaloids, saponin and cyanogenic glycosides when compared to the leaves, although flavonoids and phenols were significantly ($P < 0.05$) higher in the leaves. Mineral (elemental) analysis of both leaves and stem bark indicated the presence of macro and micro minerals in varying amounts.

Conclusion: These findings indicated that *S. mombin* contained an appreciable amount of nutrients and minerals which could be included in the diet to supplement our daily nutrients and animal feeds, while the presence of the phytochemical may justify their use in ethnomedicine for the prevention, treatment and management of diverse health challenges, the phytochemicals may also act as lead compounds in the development of new drugs to counteract the present drug-resistant phenomenon.

Keywords: Minerals; phytochemicals; proximate composition; *Spondia mombin*.

1. INTRODUCTION

Green medicine is an important alternative therapeutic aid, owing to their broad spectrum of phytochemicals. These organic substances are safe, reliable, accessible, economical and products of nature with vast potentials for sustainability in the global combat of multidrug-resistant strains of microbes. There is a need to channel research to exploit and establish the wealth of untapped phytoconstituents with profound physiological importance.

Spondia mombin is a green-based fructiferous medicinal tree belonging to the family *Anacardiaceae*. It grows in the coastal areas covering the rain forest and other tropical regions [1]. The plant is widely reported for its medicinal values cutting across various aspects of human health and well-being. It is popular for eating, making ice lolly and also employed in managing several conditions locally, such as induced labour, prevent abortion and control hemorrhagic associated with child delivery [2], candidiasis, fever, washing cuts, sores, burns, worm expellant [3,4], diuretic, useful in preventing vomiting, dysentery, diarrhea, haemorrhoids, gonorrhoea and leucorrhoea [5]. Isolated compounds from *S. mombin* were also reported to be antiinfective [3, 6,7]. The vast ethnomedicinal claims of *S. mombin* have been a subject of concern with limited data to validate the bioactive phytoconstitution of the stem bark and leaves. Thus, the present study is designed to ascertaining and validating the acclaimed use of this plant in green medicine and possible incorporation in food.

2. MATERIALS AND METHODS

2.1 Equipment

Some of the equipment used in the course of the study includes: glasswares, micropipette, heating

mantle (CAP.500MI), refrigerator (HTF219H), UV-visible spectrophotometer (792N), water bath (KW-1000C), weighing balance (905N), spatula, drying oven (MINO/30), water distiller (SZ-1), crucible, evaporating dishes, Kjeldahl flasks, digestion rack, condenser, Soxhlet extractor, rotary evaporator (RE52-1), desiccator, Buchner funnel, suction pump, muffle furnace (SX-5-12).

2.2 Chemicals and Reagents

The chemicals and reagents used were of analytical grade. They include petroleum spirit (ether), sulphuric acid, sodium hydroxide, anhydrous sodium sulphate, anhydrous copper sulphate, mayer's reagent, dragendoff reagent, ethyl acetate, aluminum chloride, ammonia solution, olive oil, ethanol (80%), ferrous sulphate, ferrous chloride, lead acetate, bromine water, Fehling solution A and B, chloroform, 40% sodium hydroxide, boric acid, methyl red, methylene blue, 0.1M hydrochloric acid.

2.3 Collection of Plant Material/Sample Pretreatment

The plant was collected in Otuoke, Ogbia Local Government Area of Bayelsa State, Nigeria. Identified and authenticated with voucher No. NDU/P/008 at the Niger Delta University, Bayelsa State, Nigeria. The leaves and stem bark were air-dried, blended, and subjected to cold extraction at room temperature, using ethyl acetate solvent and concentrated under vacuum with the aid of rotary evaporator at 40 °C separately.

2.4 Preparation of Plant Material

The leaves and stem bark were shade dried at room temperature and then blended. 30 g each of the samples was extracted with 300 ml of 80% aqueous ethanol using the Soxhlet setup for 72 hours before being concentrated under vacuum.

2.5 Quantitative Phytochemical Analysis

Determination of alkaloid content was done by the Harbone gravimetric method [8], flavonoids were quantified using the method reported by Bohm and Kocipal- Abyazan [9], tannins were evaluated using the Burns method [10], cyanogenic glycosides using the method of Wang and Filled [11], saponins were determined using the froth method [12], phenols using the technique adopted by AOAC [13] and phytates by method of Garcia-Villanova et al. [14].

2.6 Quantitative Proximate Analysis

The following nutritive analyses: Moisture content, ash content, crude protein content, crude lipid content, crude fibre content, and total carbohydrates were carried out using the AOAC methods [13]. The energy (cal.) values were obtained by the method of Vadivel and Janardhanam [15].

2.7 Quantitative Elemental Analysis

The macro and micro-mineral of biomedical significance: Chromium, iron, magnesium, manganese, zinc, copper, calcium, sodium, potassium, phosphorus, sulphur and chloride present in the plant samples were assayed using the method of AOAC [13].

2.8 Statistical Analysis

Data obtained from the study were expressed as a mean of three (3) replicates \pm standard deviation (SD). Statistical Analysis of data was performed using the SPSS statistical software (Statistical package for social science version 16). The obtained data were analyzed using a one-way analysis of variance (ANOVA) followed by post hoc Tukey. $P < 0.05$ was considered as statistically significant.

3. RESULTS AND DISCUSSION

3.1 Quantitative Phytochemical Analysis

The quantitative phytochemical analysis of *S. mombin* leaves and stem bark indicated the presence of alkaloids, flavonoids, tannins, cyanogenic glycosides, saponins, phenols, oxalates and phytates in varying amount. The alkaloids are amines in nature and have been associated with medicinal use for centuries. Therapeutic properties of alkaloids include cytotoxicity, analgesic, antispasmodic and

antibacterial [16,17,18]. Studies have also shown that the alkaloids have anti-HIV and anti-parasitic, anti-tumour, antioxidant, anti-genotoxic, and hallucinogenic properties [19]. Flavonoids consist of a large group of polyhydroxylaromatic compounds having a generic benzo- γ -pyrone skeleton. The flavonoids are known to exhibit antiinfective activities against wider lines of microorganisms [20]. This is due to their ability to annex extracellular and soluble proteins of the bacteria cell wall [21]. The flavonoids have strong free radical scavenging mode [22]. They often act by terminating ROS formation either by inhibition of enzymes or by chelating trace elements involved in free radical generation and by in a way promoting the system's biochemical antioxidant defense [23,24,25]. Several flavonoids have been reported to have hepatoprotective, anticancer, and anti-inflammatory activities [26,27,28]. Saponins are known plant glycosides with characteristic foaming and bitter taste. They have cytotoxic effect, anti-inflammatory, anticancer and have tumour inhibiting potentials [29]. They are known antifungal and also show beneficial cholesterol-lowering activity [30], precipitate and coagulate red blood cells. The saponins are also known to boost fertility and as a possible bioactive agent responsible for the aphrodisiac effects [31, 32,33]. Many tannins have been implicated to reduce the mutagenic and carcinogenic activities of most mutagens and carcinogens and this is largely due to their antioxidant properties [34,35]. Tannins also can facilitate blood clotting, decrease blood pressure, lower serum lipid parameters, and with strong antimicrobial activities [36]. Oxalates are phytochemicals found in large amounts in many plants and have little or no effect on the human [37]. Studies examining the effect of phytates, have it that they are important in regulating vital cellular function. Both *in vivo* and *in vitro* experiments have shown anticancer therapeutic effects of phytates [38].

3.2 Quantitative Proximate Analysis

The bedrock of nutritional fact is the proximate analysis. It provides vital data that are empirical. The nutritional values of some underutilised multi-purpose plants like *S. mombin* is necessary, because it will in no small measure create awareness, remove the cultural barrier and improve the consumption rate of this plant. The major indices of nutritional values include carbohydrate, protein, fat, ash, and moisture. Proximate analysis of the leaves and stem bark of *S. mombin* indicated the presence of the six

basic nutrients. Carbohydrates are primary metabolites essential for survival, biochemical sources of energy and building blocks for most secondary metabolites. The biomolecules that can hydrolysed to yield a six-carbon monomeric unit (glucose) which can be utilised immediately or stored as glycogen in the muscles and liver for future use [39,40]. Proteins are important primary metabolites responsible for the biosynthesis of hormones, enzymes, and blood plasma. They are immune boosters and can help in cell division as well as growth [41]. Lipids are other building blocks for secondary metabolites. They yield more energy per gram than carbohydrates. Dietary fats are needed not only for their high energy value but also for fat-soluble vitamins and essential fatty acids contained in the fats of natural foods. Lipids also help to regulate blood pressure and play a useful role in the synthesis and repair of vital cell parts [42]. Moisture content often regarded as water is a universal solvent; it dissolves other substances, carries nutrients and

other materials throughout the body thus making the entire body system to function optimally [43]. Fibres are part of plant food which can neither be digested nor absorbed by the digestive system. Generally dietary fibre slows down the rate of glucose absorption into the bloodstream, thereby reducing the risk of hyperglycemia. They also reduce the level of plasma cholesterol and prevent colon cancer and cardiovascular diseases [44,45]. Ash content of plant-based food is the function of the mineral elements present, dietary ash has proved helpful in establishing and maintaining the acid-alkaline balance of the blood system as well as controlling hyperglycemia conditions [46]. Adequate and balanced nutrition as depicted by the presence of all major food components in *S. mombin* leaves and stem bark can promote good health and boost immunity against a wide range of diseases [47]. The low calorific value of the leaves and stem bark may also help obese patients to come down on their body weight.

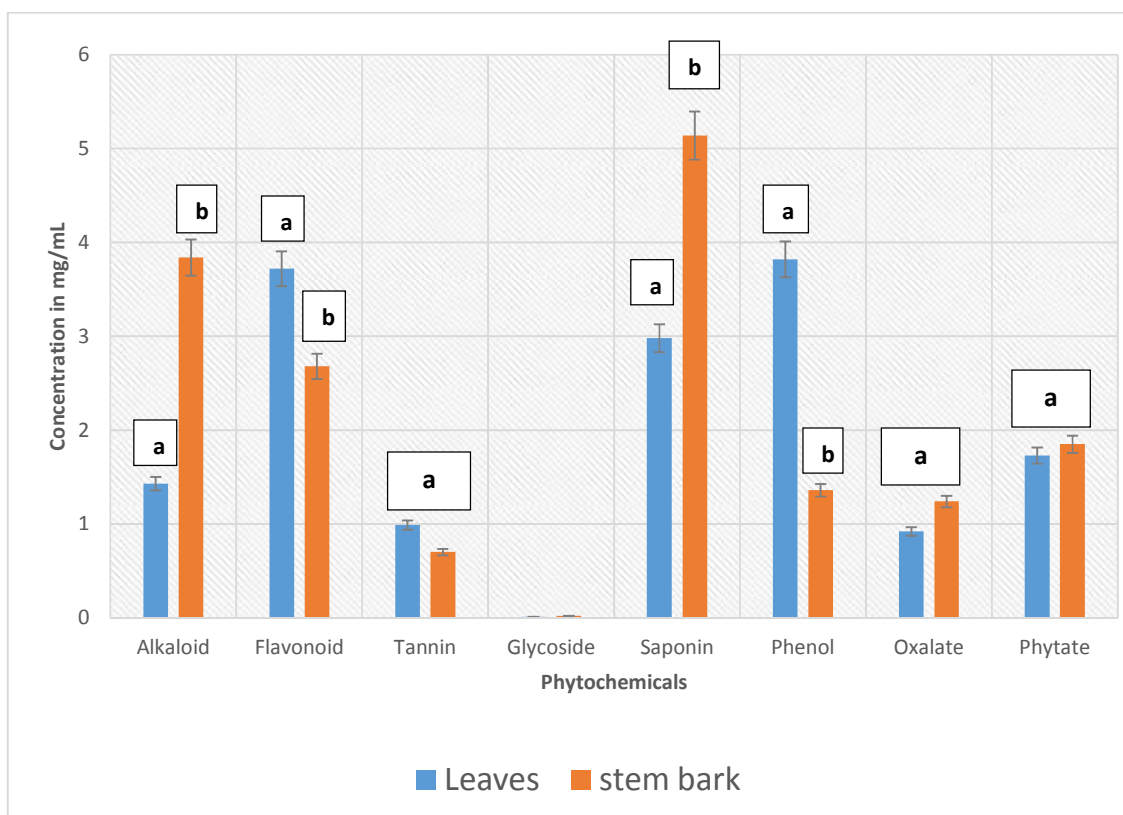


Fig. 1. Phytochemical composition of *S. mombin* leaves and stem bark

The values of phytochemical parameters are expressed as a mean of three replicate determinations ($n=3$) \pm standard deviation (SD). Values with different superscript letters are significantly different ($P < 0.05$), one-way analysis of variance (ANOVA) followed by post hoc Tukey

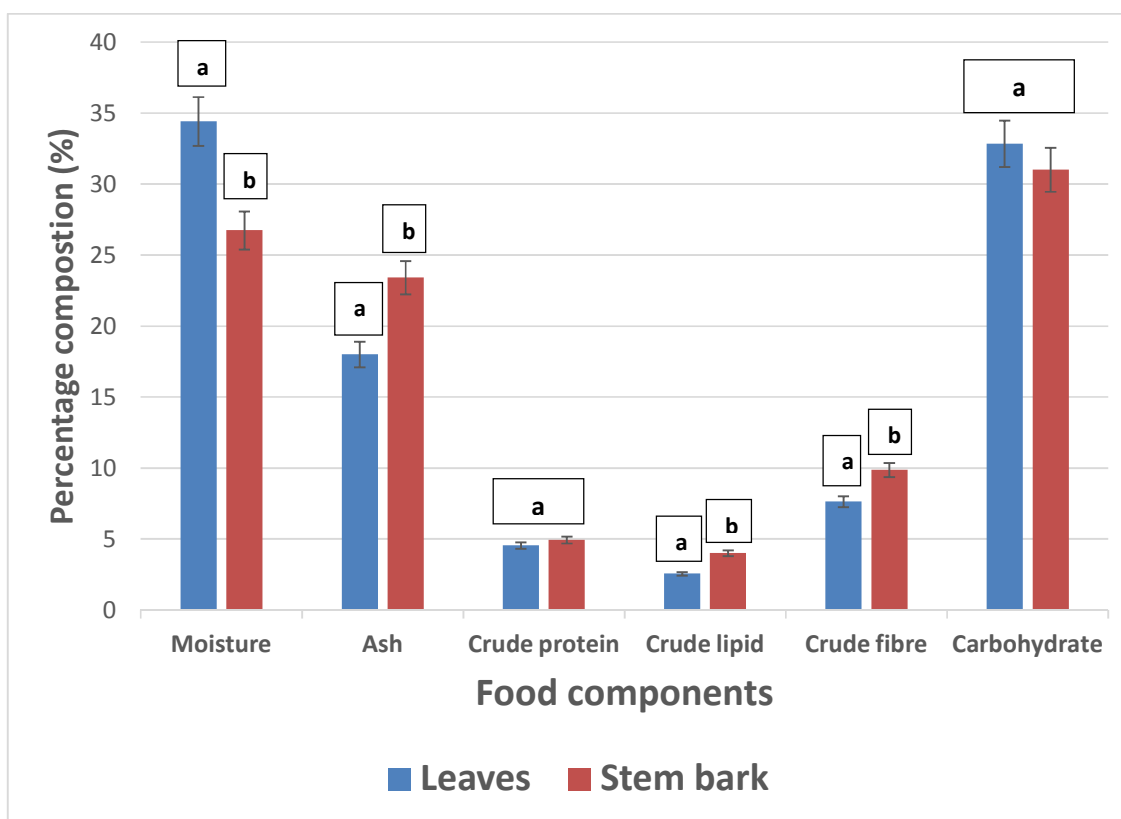


Fig. 2. Proximate composition of *S. mombin* leaves and stem bark

The values of proximate composition are expressed as a mean of three replicate determinations ($n=3$) \pm standard deviation (SD). Values with different superscript letters are significantly different ($P < 0.05$), one-way analysis of variance (ANOVA) followed by post hoc Tukey

3.3 Quantitative Elemental Analysis

Analysis of *S. mombin* leaves and stem bark reveals that they are rich in minerals like chromium, iron, magnesium, manganese, zinc, copper, potassium, phosphorus, sulphur and chlorine. Potassium, which is the principal base ion of intracellular fluid, sodium, which is the main cation of extracellular fluid, and collectively functions to maintain the electrical potential of the nervous system and hence proper functioning of the nerves. Both cations are also very important in the regulation of water and electrolyte balance and acid/base balance in the body. The results also indicated the presence of calcium, which is an essential constituent of bones and teeth, it is also an important factor for many metabolic processes like nerve function, muscle contraction, maintenance of regular heartbeats and blood clotting. Magnesium which is also a constituent of bones and teeth was also detected in varying quantities in *S. mombin* leaves and stem bark. Magnesium is needed for

the proper functioning of muscles and nervous tissues, it is also a co-factor of many enzymes (kinases) particularly those of glycolysis and many ATP-dependent reactions (oxidative phosphorylation, cell replication, nucleotide metabolism and protein biosynthesis). The biological functions of zinc can be broadly classified into three namely catalytic activities, as it is a co-factor of over 300 enzymes catalyzed reactions, regulatory functions, and structural function [48,49]. Over 90% of zinc can be found in bones and teeth [48]. Zinc supplementation at a dose of 5 mg/day for two weeks has been reported to exert strong aphrodisiac property [50], its supplementation has also been recommended as an adjunct therapy during the treatment of diarrhoea in children [51]. The importance of iron in health and disease was recognized by man from ancient times. It is an essential component of all living organism, where it plays a key role in haemoglobin formation and oxygen transport [52]. Thus Proper and appropriate balance of these minerals in the

human body can promote disease resistance which may in turn translate into optimal functioning of all cells, tissues, organs, and systems of the body. Chloride exist as salts, helps to maintain the electrical balance in the nervous system, and is also involved in intracellular and extracellular transport [53]. Sulphur is an important element for the entire biological kingdom due to its incorporation into proteins and other biomolecules. Sulphur is an essential part of many enzymes and antioxidant molecules like glutathione and thioredoxin. Some sulphur-containing compounds can efficiently form a line of defense against reactive oxygen and nitrogen species [54]. They are well known in the treatment of oxidative stress-induced pathological disorders [55]. Disulphide bonds are usually found in proteins that are localized in the extracellular

environment or within proteins that are on the cell surface. These disulphide bonds occur because of the oxidizing conditions in the extracellular environment. In the intracellular environment however, reducing conditions are maintained and disulphide bonds are absent. Several proteins are responsible for maintaining this reducing environment and these include glutathione, thioredoxin, and glutaredoxin [56].

The quantitative values of phytochemicals, proximate and mineral compositions of the leaves and stem bark of *S. mombin* found in this study are however of proximity with literatures [2,3,5], although with some little variations which may be adduced to differences in climatic conditions of regions in which samples were obtained [57].

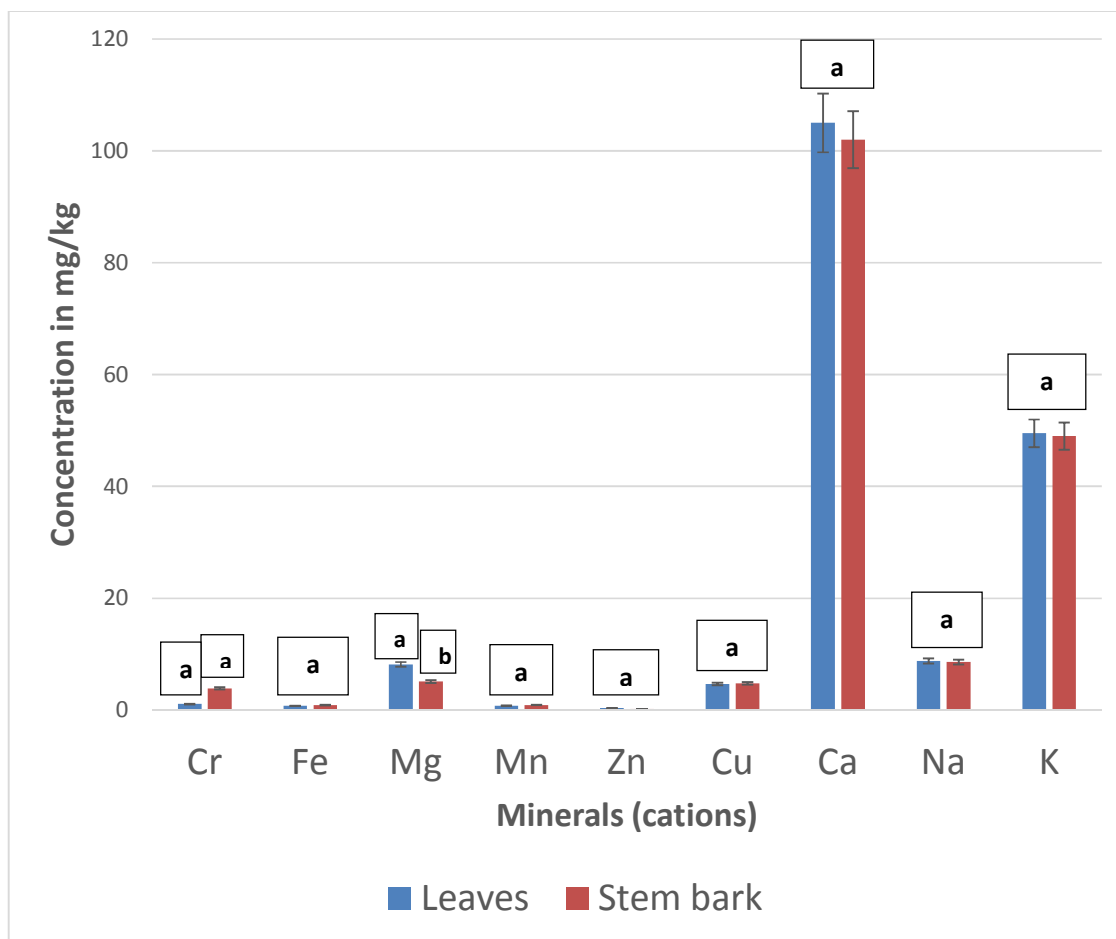


Fig. 3a. Mineral composition (cations) of *S. mombin* leaves and stem bark

The values of mineral composition (cations) are expressed as a mean of three replicate determinations ($n=3$) \pm standard deviation (SD). Values with different superscript letters are significantly different ($P < 0.05$), one-way analysis of variance (ANOVA) followed by post hoc Tukey

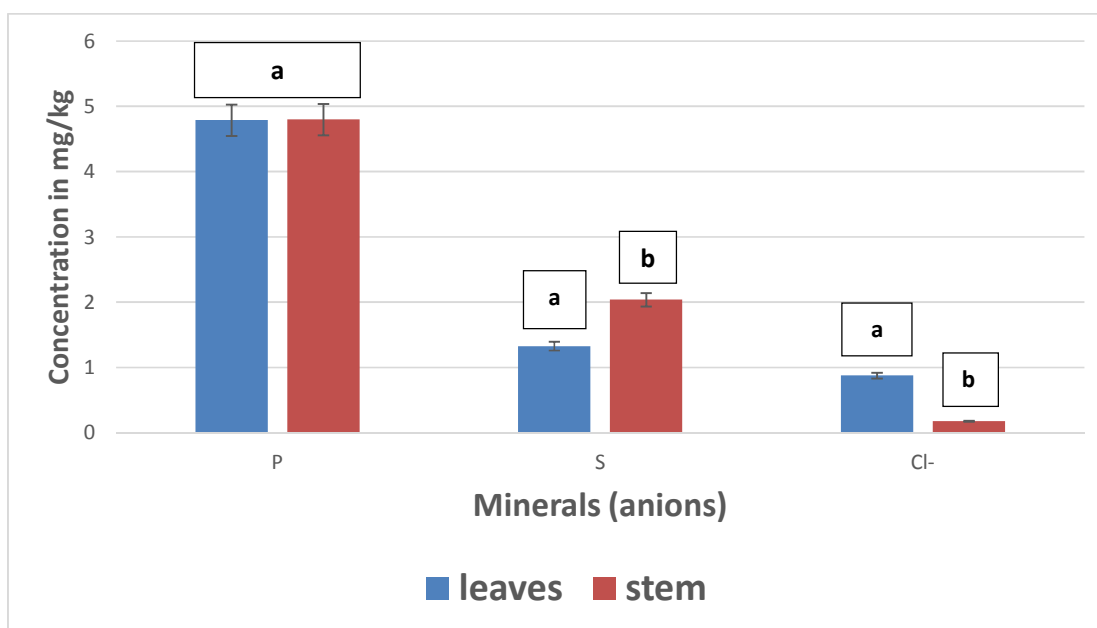


Fig. 3b. Mineral composition (anions) of *S. mombin* leaves and stem bark

The values of mineral composition (anions) are expressed as a mean of three replicate determinations ($n=3$) \pm standard deviation (SD). Values with different superscript letters are significantly different ($P < 0.05$), one-way analysis of variance (ANOVA) followed by post hoc Tukey

4. CONCLUSION

This study has shown that the leaves and stem bark of *S. mombin* is significantly rich in phytochemicals with great pharmacological potentials, hence justifying their ethnomedicinal claims in green medicine. The nutritional fact and the elemental composition of the leaves and stem bark are comparatively significant, thus justifying the nourishment ascribed to the plant.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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