ISSN (Print): 2476-8316
ISSN (Online): 2635-3490
Dutse Journal of Pure and Applied Sciences (DUJOPAS), *Vol. 5 No. (2a) December 2019*

Metal Toxicity in *Terminelia avicennioides* and *Sida acuta* Medicinal Plants from Gbongbofu and Swatamukun in Bida, Niger State

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Abstract

Traditional herbs are the oldest remedies of infirmities and man's dependence on plants for health care is as old as civilization. The leaves, stems, barks and fruits of Terminalia avicennioides and the leaves and stems only of Sida acuta were randomly collected from Swatamukun and Gbongbofu based on the diverse cultural traditions associated with the use of these plants. The medicinal plant samples were digested using Method 975.03 of the AOAC and thereafter analyzed using AA500 Atomic Absorption Spectrophotometer for Cd, Fe, Pb, Mg, Mn and Zn contents. Also, proximate analyses of the samples were carried out using standard methods. Results showed that the metal contents varied in both plant parts from the two locations except Pb with a concentration of 13.15 µg/g in all the samples, which exceeded the 10.0 µg/g of WHO permissible limit. The range of concentrations in both Swatamukun and Gbongbofu for Fe, Mg, Mn and Zn were 0.00-77.00, 0.00-19.00, 0.00-47.10 and 4.00-35.60 µg/g respectively. Thus, Fe content was highest in most of the plant samples. The same concentration of Mn and Zn was obtained in the stems of Sida acuta from Gbongbofu (35.6 µg/g) while the fruit of Terminalia avicennioides had 31.5 µg/g of Zn, which are below the 100 µg/g of WHO permissible limit for Zn in medicinal plants. Cadmium was not detected in any of the plant samples. Terminelia avicennioides contained higher ash, crude fat, crude lipid and crude fiber contents as well as moisture content of 64.67, 76.00 and 78.00 % in the leaves, stems, and fruits than Sida acuta, which implies that it cannot be kept for long time. However, carbohydrate was higher in Sida acuta than Terminelia avicennioides,

Keywords: Traditional medicinal herbs; health risk assessment; brass industries; toxicity.

INTRODUCTION

Brass is one of the most famous objects in African art, produced firstly by the Kingdom of Ife and then the Benin Empire. Brass work continued to be important art in some other parts of

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Nigeria. Brass industry in Bida, Niger State, has its history traced back to the Egyptian immigrants that previously settled there, who specialized mainly in production of bangles, cups, rings and swords (Gbate&Usman, 1988). The brass industrial sites are located within the residential areas and the public concern over the possible and potential health effects has been undermined (Iyaka&Kakulu, 2012). Brass is an alloy made of Cu and Zn, with bright gold-like appearance, malleability than bronze, with relatively low melting point of 900 °C to 940 °C. In order to increase the properties of brass, certain metals like Al, Fe, Mn, Pb, Sn and Si are sometimes added to improve the quality of the brass products. Addition of Pb will enhance the machinability of brass, which is often added in concentrations of about 2 % because it has a lower melting point than the other constituents of brass(Khan et al., 2015).

Various cases of human disease, disorders, malfunction and deformity of organs due to this metal toxicity have been reported in the past few decades. Along with human beings, animals and plants are affected by a reasonable amount of these metals (Kirmaniet al., 2011; Abdullahi et al., 2001; Moseset al., 2012).They are metallic elements that are toxic and have high densityand atomic weight. Such metals with a potential negative health effect or environmental impact may be termed heavy metals (Kabata-Pendias, 2011).

Kabata-Pendias(2011) observed that plants may act as a geochemical storage for contaminants, and as a natural buffer controlling the environmental cycling of the chemical elements to the atmosphere, hydrosphere and biota. The concern for heavy metals in plants is directly related to their interactions within all the systems through the food chain (Sridhara, Kamalaa& Raj, 2008).

Medicinal plants contain different chemical compositions due to so many factors, including climatic conditions, fertilizer, pesticide, geographical distribution, age of plant, source of collection, altitude, period of harvesting, manufacturing practices (Liu, 2011). Medicinal plants may be easily contaminated by absorbing heavy metals from soil, water and air. Usually plant is subjected to contamination through atmospheric deposition of heavy metals from point sources including metalliferous mining, smelting and different industrial activities. Some other sources of plants contamination involve use of fertilizers, pesticides, sewage sludge and organic manures (Singhet al., 1997).

Sida acuta popularly known as a weed is of utmost importance and its values and potentials have earned it inclusion among the medicinal plants of the world. Many researchers have reported its strong pharmacological activities (Karou et al.,2006). *Sida acuta* is traditionally used in the treatment of malaria, diarrhoea and many other diseases. Its application in the therapeutic management of disturbing conditions such as asthma, renal inflammation, colds, fever, headache, ulcers and worm infections in regions around Central America has been reported (Caceres, 1987). The plant has also been found applicable for the treatment of snake bite (Oteroet al., 2000a; 2000b) with an indication that the ethanol extract of the plant had an effect against the venom of Bothroxathrox.

*Terminaliaavicennioides*have been found to be useful against *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa* in patients suffering from complicated respiratory tract infections for their antimicrobial activity. Research showed that the ethanolic extracts of the plant exhibit a significant antimicrobial activity against *Staphylococcus aureus, Escherichia coli* and *Pseudomonas aeruginosa*. The bioactive constituents like phenols, steroids, glycosides, flavonoids, tannins, ellagic acids, detected in this plant are responsible for the observed antimicrobial activity and hence, its potential use as antimicrobial agent (Mann et al., 2008).
This work is aimed at determining the metal contents of two medicinal plant samples from two brass-making sites. i.e. Gbongbofu and Swatamukun in Bida, Niger State, Nigeria. The

study will further determine the human health risk associated with these metals and create awareness on the effect of the brass industries and some of the additives, which are added to improve the brass products.

MATERIALS AND METHODS

The medicinal plants samples (leaves, stems, barks and fruits) were randomly collected from Gbongbofu and Swatamukun in Bida, Niger State, Nigeria. Shovel, trowel, stainless steel knife, polyethylene bags, paper envelopes and gloves were used for the collection of the plant samples. The simple random sampling approach was used. They were placed in labeled paper envelopes in order to avoid loss of moisture content and taken for analysis in the laboratory of the Department of Chemistry, Ibrahim Badamasi Babangida University, Lapai, Niger State, Nigeria. The fresh plant samples were spread on labeled plastic trays and air-dried for 7 days at a temperature of about 35 ^{o}C. The air-dried samples were pounded using a wooden mortar and pestle. Then, the samples were blended using Kenwood blender BL480 and sieved through a 1 mm sieve. The fine samples were then stored in labeled air-tight polythene bags ready for analysis. All glassware and digestion vessels were soaked in 10 % nitric acid overnight and rinsed with distilled water.

For wet digestion of the plant samples, Method 975.03 of AOAC (2006) was used in the wet digestion of the plant samples.1.00 g of the sieved plant sample was weighed into a 150 ml beaker. 10 ml of concentrated HNO_3 and 3 ml of 60 % $HClO_4$ were added and allowed to soak thoroughly, while a deep brown fume and frothing was observed. The contents of the beaker were heated gently at first on a Jenway 1000 hot plate with stirrer until the frothing ceased; the heating was continued until the HNO_3 was almost evaporated and cooled. 10 ml of concentrated HNO_3 acid was added to the light brown solution. The heating was continued until white fumes of $HClO_4$ were observed indicating complete digestion of the sample. The digest was cooled and 10 ml of 1:1 HCl and 10 ml of 5 % La_2O_3 solution were added. This was followed by filtration using Whatman No.42 Ashless filter paper into a 50 ml volumetric flask and the solution was made up to mark with distilled water. Each sample solution was stored in a labeled plastic bottle ready for analysis using AA500 Atomic absorption spectrophotometer at National Cereals Research Institute (NCRI), Badeggi, Niger State. Each determination was in triplicate and a blank determination was also carried out.

For moisture and ash contents determination, 5.00 g of each of the fresh plant samples was weighed using Ohaus Scout Pro weighing balance and kept inside DHG-9202 series thermal electric thermostatic drying oven at 105 ^{o}C in order to determine the fresh weight moisture content. The samples were dried to constant weight, cooled in desiccators, weighed and their masses were recorded. For the ash content, 5.00 g of each sieved sample was weighed into a porcelain crucible of known mass. Then, the samples were charred with a Bunsen burner before transferring to Model SXL-1008 Muffle furnace at 550 °C and allowed to ash for 8 hours. The samples were then cooled in a desiccator, before weighing and the masses recorded.

The crude protein determination was by the Kjeldahl method**.** 1.00 g of the sieved sample and Kel-Pac catalyst were transferred into a labelled Kjeldahl flask. 35 ml concentrated H_2SO_4 was added and the blower was turned on, and set burners on '5' for digestion until the digest was clear, before setting the burner to 'Hi' for 30 more minutes. The flask was rotated continuously throughout the digestion. The digest was then cooled before the addition of 400 ml deionized water and the digest in the flask was swirled to dissolve salts; before distillation process, which involve addition of 75 ml of boric acid solution to a 500 ml Erlenmeyer flask. The delivery tube from the condenser was placed into the boric acid solution before turning water on to the distillation system and set burners on '4'. 0.5 g of

zinc and a scoop of boiling stones were added to flask which was done under the hood. 100 ml of 50 % NaOH was added slowly to the Kjeldahl flask which was placed on a burner and mixed thoroughly. The heating continued until 250 ml of distillate was collected and the receiving flask was removed and replaced with beaker containing 400 ml of deionized water. The distillate was then titrated with 0.1 M HCl, and the amount of acid used was recorded. The crude protein was calculated as % N_2 (Nitrogen) × 6.25

For the crude fiber determination, 2.00 g of the sieved plant sample was extracted with ether and transferred into 600 ml beaker, avoiding fibre contamination from paper or brush and 1.00 g of the prepared asbestos, 200 ml of boiling 1.25 % H_2SO_4, and 1 drop of diluted antifoam were added. The beaker was placed on the digestion apparatus with pre-adjusted hot plate and boiled exactly for 30 minutes, rotating beaker periodically to keep solids from adhering to the sides of the beaker and was removed. The solution was filtered into a beaker using a Buchner funnel. The beaker was rinsed with 50 ml of boiled water, and washed through the funnel. The process was repeated with three of the 50 ml portions of deionized water, and sucked dry. The mat and the residue were removed by snapping the bottom of the Buchner funnel against the top while covering the stem with thumb or fore-finger, and then replaced back to the beaker. 200 ml of boiling 1.25 % NaOH was added and boiled exactly for 30 minutes, removed and filtered as described above. The process was repeated but this time; it was washed with 25 ml of boiling 1.25 % H_2SO_4, and three 50 ml portions of H_2O, 25 ml of alcohol.

For crude fat determination, a copper wire of approximately 0.5 cm was placed on each of the labeled beaker and dried at 100 ^oC for 15 minutes. It was cooled in a desiccator and weighed before turning on water to condensers. The sample was then placed on the extraction thimble and the thimble was clamped to an extractor. 50 ml of anhydrous ether was measured into the beaker and attached snuggly to the extractor and all the burners were turned to "high" setting and the switch was turned on. When dripping occurred through the extraction thimble, the extractor was turned off and all settings were turned to low before turning the extractor on again. Extraction was at the rate of 2 to 3 drops/second for 16 hours, and burners were shut-off through the main switch and the beaker was removed. The thimble was replaced with a glass tube and the contents boiled, collected in a glass tube and the beaker was boiled until almost dry. Blank determinations were carried out and all determinations were in triplicate. The total carbohydrate was obtained by difference.

RESULTS AND DISCUSSION

Results of the heavy metals in the two medicinal plants from the two locations (Swatamukun and Gbongbofu) are given in Figures 1 & 2. The results of the various nutrients are summarized in Figure 3 and 4.

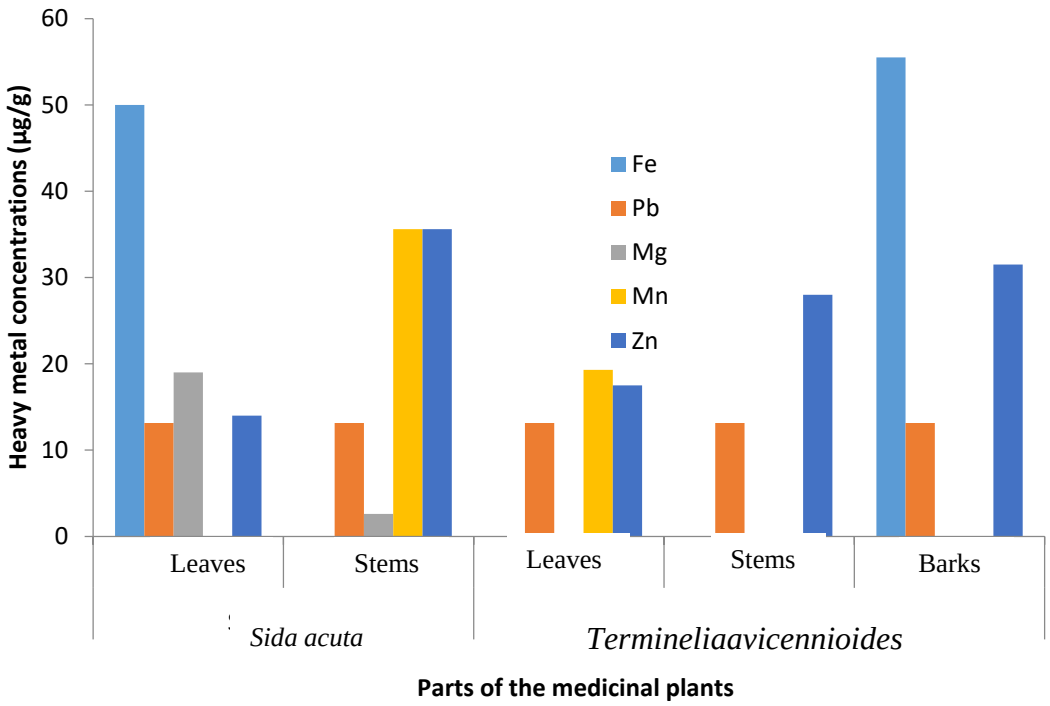

Figure 1: Total metal contents in the medicinal plants from Gbongbofu

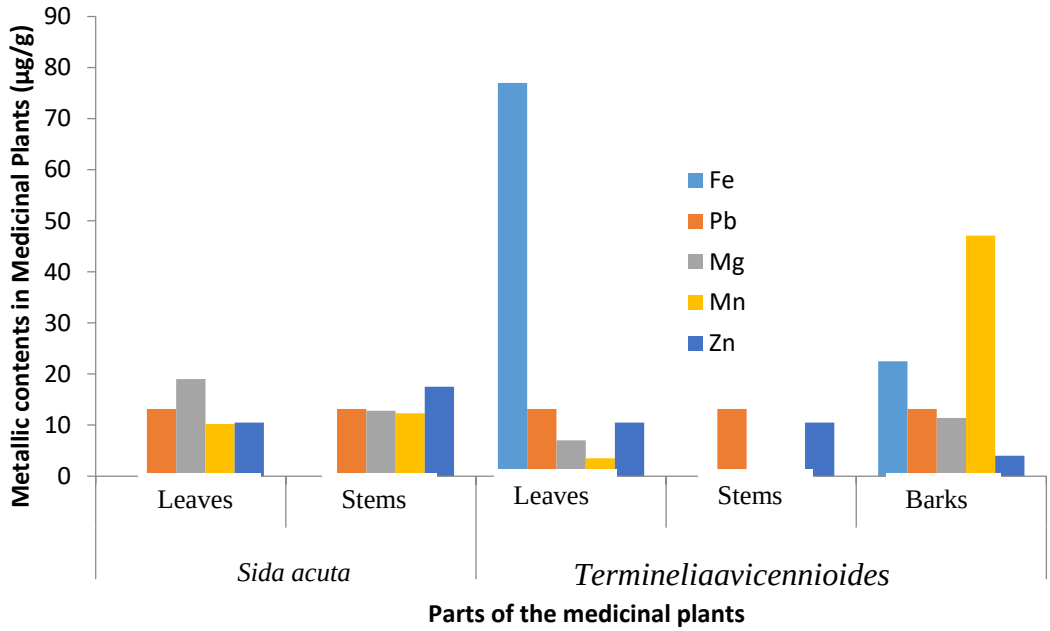

Figure 2: Total metal contents in the medicinal plants from Swatamuukun

It is necessary to recall two well-known facts about heavy metals. First, a heavy metal is not toxic; it is only toxic when its concentration in a medicinal plant exceeds a certain threshold concentration ("It is the dose that makes the effect"). The second fact is that some heavy metals are called micronutrients and they have essential functions in plant and animal cells. This has been shown for Co, Cu, Fe, Mn, Mo, Ni and Zn. As revealed in this study from Figures 1 and 2, Fe, Zn and Mn are higher in both the *Terminelia avicennioides and Sida acuta*. These medicinal plants (*Sida acuta* and *Terminalia avicennioides*) obtained from Gbongbofu and Swatamukun as shown in Figures 1 and 2, had the same concentration of Pb as 13.15 µg/g in all the parts of the two medicinal plants studied. This value when compared with the WHO (2007) and WHO (2011) maximum permissible limit of 10.0µg/g is higher. As

revealed in Figures 1 and 2, the concentrations of Mn and Znin the stem of *Sida acuta* from Gbongbofu had the same value of 35.6 µg/g. The fruit of *Terminalia avicennioides* Zn concentration of 31.5 µg/g, which was below the 100 µg/g of WHO (2011) permissible limit for Zn in herbal material.

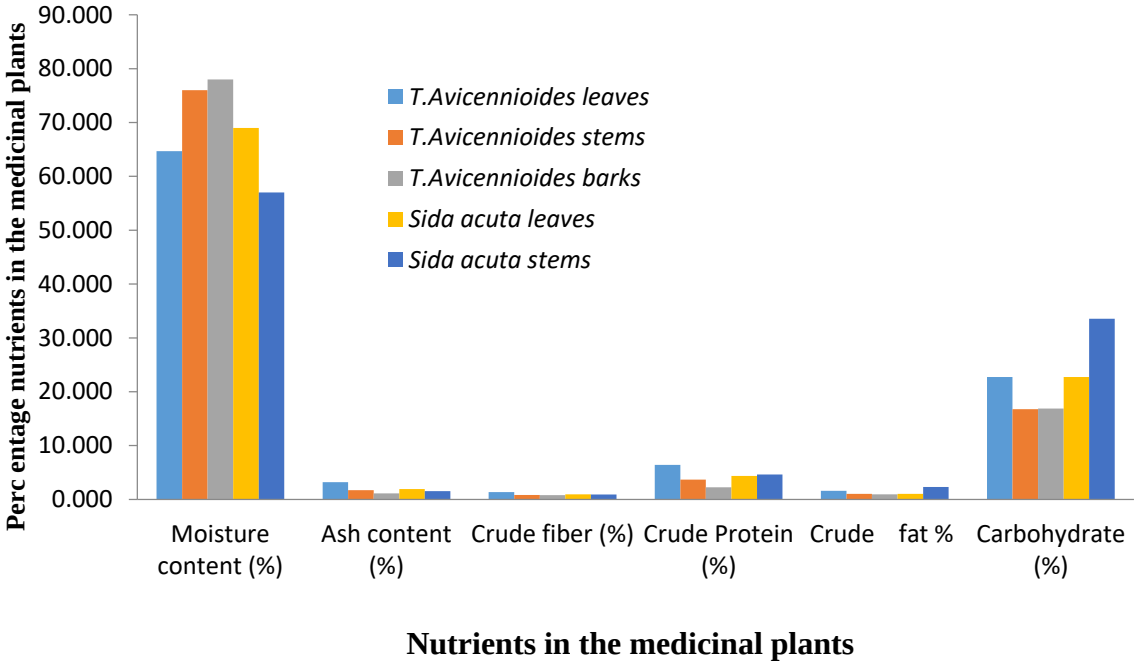

Figure 3: Total nutrients in the medicinal plants from Gbongbofu

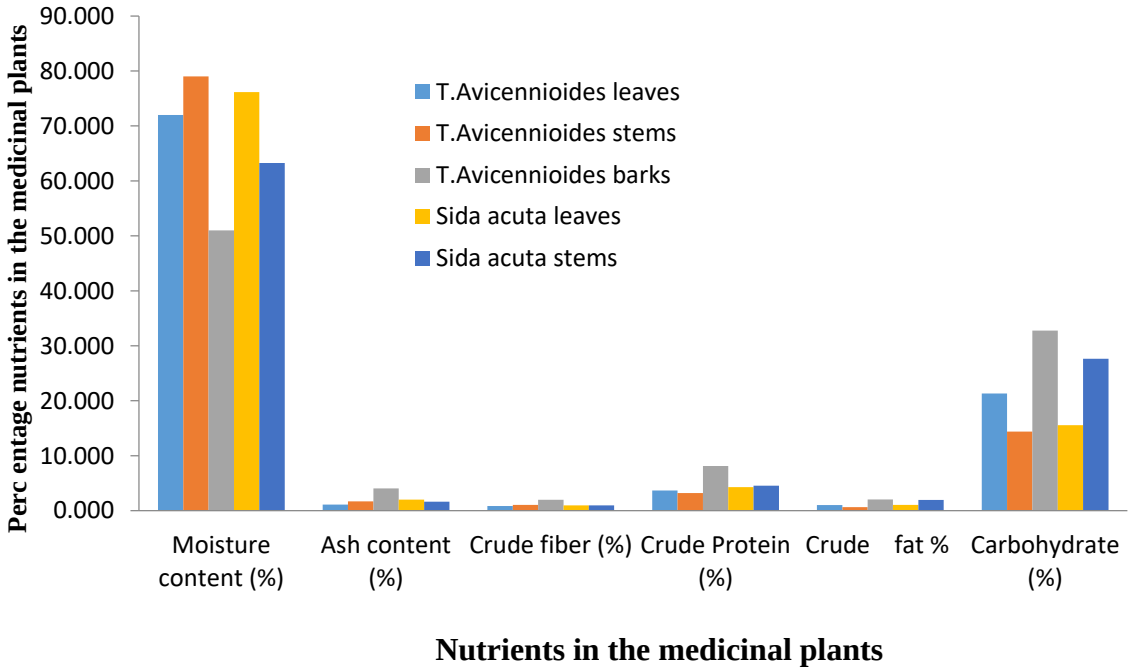

Figure 4: Total nutrients in the medicinal plants from Swatamukun

As shown in Figures 3 and 4, *Terminelia avicennioides* had high moisture content of 64.67, 76.00 and 78.00 % for the leaves, stems, and fruits respectively and 57.00 and 69.00 % in the stems and leaves respectively of *Sida acuta* from Gbongbofu. Similarly, the moisture contents

in *Terminelia avicennioides* from Swatamukun were 51.00, 72.00 and 79.00 % in the barks, leaves and stems respectively while *Sida acuta* leaves and stems showed moisture contents of 716.15 and 63.24 % respectively. The higher percentage of moisture in *Terminelia avicennioides*will not allow the plant to be kept for longer time, because moisture content of any food material can be used to measure its keeping quality.

The percentage ash in the *Terminelia avicennioides* ranged from 1.13 to 4.07 %, which is higher than the ash content obtained from *Sida acuta* that ranged from 1.19 to 2.43 %. This implies that the *Terminelia avicennioides* contain more mineral content than *Sida acuta*.
As revealed in Figures 3 and 4, the percentage of crudefat obtained from *Sida acuta* in the stems and leaves from Gbongbofu is higher than the percentage of crude fat obtained in *Terminelia avicennioides* with value ranging from 1.04to 2.32 % and 0.66 % to 2.03 % respectively. From Swatamukun, the crude fat ranged from 0.65 % to 2.03 % in the stems, leaves and fruits.

Terminelia avicennioides recorded the highest crude fiber content of 2.00 % compared to *Sida acuta* with a percentage value of 1.26 %. The high fiber content recorded in *Terminelia avicennioides* was from its barks and stems. This shows that the stems and the barks contain more fiber than the other parts of the two medicinal plants.

The percentage crude protein in the *Terminelia avicennioides* ranged from 2.27 to 8.13 % and is higher than the crude protein obtained in *Sida acuta* that range from 3.45 to 5.06 %. This implies that the *Terminelia avicennioides* contain more protein than *Sida acuta*.
The highest value of carbohydrate recorded was in the stem of *Sida acuta* from Gbongbofu with a value of 33.58 %, which is slightly higher than the 32.77 % obtained from *Terminelia. avicennioides* bark and shows that the percentage of carbohydrate obtained in *Terminelia avicennioides* is less than the percentage of carbohydrate obtained in *Sida acuta*. Carbohydrates play important roles in many important roles in living organisms (WHO/FAO, 1998).

CONCLUSION

From this study, various concentrations of six metals of interest namely, Cd, Fe, Pb, Mg, Mn and Zn were determined. Cadmium (Cd) was not detected in any of the two medicinal plants' samples from the two brass-making areas. Thus, the WHO permissible limit of 0.10 µg/g maximum for Cd was not exceeded in any of the samples, Amongst the other metals, only Pb exceeded the WHO permissible limit of 10.0 µg/g. Lead (Pb) has no beneficial effect to plants and animals and is a carcinogen. It is therefore concluded that the medicinal plants cultivated in the Gbongbofu and Swatamukun are polluted with Pb, which is toxic to plants, animals and human beings and soil to plant transfer of Pb is a key route of human exposure through food chain.

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