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The determination of elemental composition of five indigenous herbal plants used in the treatment of snake bite in the vicinity of Billiri and Kaltungo Local Government areas of Gombe State, Nigeria.

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ABSTRACT

The determination of elemental compositions of *Gardenia terniforlia, Annona senegalens, Combretum*, *Balanite aegyptiaca and Borreria verticillata* herbal plants used in treatment of snake bite in the vicinity of Billiri and Kaltungo Local Government areas of Gombe State were analyzed using Atomic Absorption spectrophotometer (AAS) spectrophotometer method. The element content appeared to be dependent on the type and the part of the plant samples. The leaves and stem/bark of *combretum* epiphyte have the highest concentration in Mn of 90.9±0.0024 and 43.35±0.0009, while the Cd has the least amount of 0.4±0.0006 and 0.2±0.0003 respectively. The highest amount of Zinc in leaves and stem/bark of *Gardenia ternifolai* epiphyte were 35.82±0.0019 and 34.63±0.0033 respectively, while the least amount were found in Ni and Cr for leaves of *Gardenia ternifolia epiphyte* and least in Cd for the stem. The root bark of *Annona senegalensis* and *Balanite aegyptiaca* has the highest concentration of Zn, and least of Ni and Cd respectively. Whereas the graphs shows the highest concentration of Mn in *Borerria verticillata* root and Cd has the least amount. Considerable amount of sodium was recorded in the samples investigated with highest amount in root bark of *balanite aegyptiaca* 750mg/kg and the least amount was found in leaves of *combretum epiphyte* 110mg/kg. Pb was not detected in the entire samples.

Keywords: Elemental composition, herbal plants, Atomic Absorption spectrophotometer.

INTRODUCTION

The Nigerian Ministry of Health reports that snake bites kill every year. Research in recent years has provided critical data on the conditions that promote snake-human contact, snake bite patient profile, and the necessity of in-state production of large quantities of affordable antivenin. At this time, development of anti-venom facilities in Nigeria is in place due to the EchiTab Study Group of Liverpool, England, and novel research with herbal medicine is being conducted at the University of Nigeria Nsuka [1, 2]. Venomous snake species which inhabit Nigeria include *Bitis gabonica* (gaboon viper), *Bitis arietans* (puff adder) and *Naja nigricollis* (black necked spitting cobra). However, *Echis ocellatus*, the West African carpet viper, is the most common. It accounts for 90% of bites and 60% of the fatalities in the country, which add up to 20% of all African cases [3,4].

Generally, the use of herbs to treat disease is almost universal among non-industrialized societies, and is often more affordable than purchasing expensive modern pharmaceuticals.

Medicinal plants are plants that have at least one of their parts (leaves, stem, bark or roots) used for therapeutic purposes [5,6]. [7], defined medicinal plants as any plant which contains substances that can be used for therapeutic purposes in one or more of its organ or substances which are precursors for the synthesis of useful drugs. The herbs which appear most effective as per the symptoms of snake bites are relatively non-toxic and have substantial documented efficacy, among them some are *Aristolochia* species, *Cissus assamica, Echinacea* species, *Guiera senegalensis, Hemidesmus indicus, Parkia biglobosa, Securidaca longipedunculata, Tamarindus indica, Trianosperma tayuya, Thea sinensis, Withania somnifera* [8,9]. *Gardenia ternifolia* is a genus of flowering plants in the coffee family, Rubiaceae, native to the tropical and subtropical regions of Africa, southern Asia, Australasia and Oceania [10, 11]. *Annona senegalensis* is a species of flowering plant in the custard apple family, Annonaceae [12]. A traditional food plant in Africa, the fruits of *A. senegalensis* have the potential to improve nutrition, boost food security, foster rural development and support sustainable land care. Well known where it grows naturally. *Combretum* make up the type genus of the family Combretaceae. *Balanites aegyptiaca* a member of the family Zygophyllaceae, is one of the most common but neglected wild plant species of the dry land areas of Africa and South Asia [13]. *Borreria verticillata* is a woody, bushy shrub. It is a perennial reaching 1 m in height, glabrous and erect with many ramifications. It is very common in humid areas [14].

Since pre-historical time, snake bite has been an issue in the areas of Billiri and Kaltungo local government areas of Gombe State. Many researchers have worked on different snake bite medicinal plants. But certain essential elemental pollutants in these medicinal plants were normally not part of the results presented in literature. Hence there is need to carry out elemental analyses of some medicinal plants so as to establish scientific data, with respect to safety and mineral benefits derived from them.

Elemental analysis is a process where a sample of a compound is analyzed to determine its composition and some isotopic composition. Micro analysis has provided elemental analysis services for customers in a great number of industries, research and academic research. These can be qualitative and quantitative. A great number of research conducted on these medicinal plants often focus on the organic actions of these medicines with little attention going to the elemental contents [15]. Due to their potential impact on human health, the pharmacological properties of these medicines must be studied. The pharmacological properties have been attributed to active chemical contents. Though trace elements have been reported to play a role in the formation of these active chemical constituents, a direct correlation between elemental composition of the medicinal plants and their remedial properties has not yet been established [15,16]. However, lack of knowledge of the elemental constituents of these medicinal plants often pose danger to consumers as some may contain toxic elements. Also the

dose rates of many of these medicinal plants are not well defined and left to the judgment of the users [17].Thus, screening of the elemental composition of these medicinal plants is highly essential [5]. The importance of trace elements in various metabolic activities in the human body and also the increasing industrialization and environmental pollution, this present study, elemental analysis of some medicinal plants used in Ghana was carried out using Instrumental Neutron Activation Analysis to study trace elements in some selected medicinal plants and relate the levels of the trace elements to its traditional uses for some time now, several techniques such as AAS, XRF, ICP – MS, PIXE etc have been used for the analysis of trace elements.

MATERIALS AND METHODS

Heavy metals determination: This was achieved by means of absorbance measurements of dilute solution using the advantage of the sensitivity of AAS spectrophotometer. The instrument was calibrated by allowing some warm up time for the AAS machine and the control knob was adjusted until the meter reads 0% transmittance. Two glass cuvettes was obtained and blotted with tissue paper. One of them was used for solvent blank which was distilled water while the other was used for the analyte samples. The absorbance reading was measured for the each metal serial dilution of the various samples, calibration curves for each metal was prepared for which unknown concentrations in the sample was extrapolated. A plot of the absorbance (A) versus concentration gives a straight line graph.

Determination of K and Na using FES: The analyte sample was introduced into flame as a gas.The thermal energy dried the solvent, break the chemical bonds and excited atoms which emit characteristic light of specific wave length as it returns to the ground state.This is disperse by a grating or prism detected by the spectrometer.

The blank compensate for any absorption by the solvent containing the compound and serves to eliminate unwanted absorption due to the solvent.

Statistical Analysis: Each experiment was carried out in triplicate and result were expressed as mean ± SD (n=3).

RESULTS

Table 1: Concentration of some heavy metals

Plants	Cr (mg/kg)	Pb (mg/kg)	Zn (mg/kg)	Mn (mg/kg)	Cd (mg/kg)	Ni (mg/kg)
Leaves of Combretum epiphyte	0.65 ±0.0007	<0.0001	35.93± 0.0022	90.9 ±0.0024	0.4 ±0.0006	<0.0001
Stem/bark of Combretum epiphyte	0.80 ±0.0009	<0.0001	37.02 ±0.0096	43.35 ±0.0009	0.2 ±0.0003	0.15 ±0.0005
Leave of G. ternifolia epiphyte	0.15 ±0.0009	<0.0001	35.82 ±0.0019	24.70 ±0.0000	0.25 ±0.0002	0.15 ±0.0005
Stem/bark of G. ternifolia epiphyte	<0.0001	<0.0001	34.63 ±0.0033	16.15 ±0.0007	0.2 ±0.0002	
Root bark of Annona Senegalenses	0.06 ±0.0008	<0.0001	26.49 ±0.0018	19.2 ±0.0006	0.86 ±0.0001	0.35 ±0.0004
Root bark of Balanite aegyptiaca	<0.0001	<0.0001	22.84 ±0.0015	15.85 ±0.0002	0.25 ±0.0002	2.1 ±0.0004
Root of Boreria verticillata	<0.0001	<0.0001	29.49 ±0.0032	69.1 ±0.0015	0.3 ±0.0002	

Key: Cr-Cromium,Pb- Lead, Zn- Zinc, Mn-Manganese, Cd- Cadmium, Ni- Nickel

Table 2: Levels of Na and K

Plants	Na(mg/kg)	K(mg/kg)
Leaves of Combretum epiphyte	110	6600
Stem/bark of Combretum epiphyte	180	4600
Leave of G. ternifolia epiphyte	335	13600
Stem/bark of G. ternifolia epiphyte	345	7800
Root bark of Annona Senegalenses	270	3400
Root bark of Balanite aegyptiaca	750	8200
Root of Boreria verticillata	225	5000

Key: Na-Sodium, K-Potassium

DISCUSSION

The result for the leaves of Combretum epiphyte, stem bark of Combretum epiphyte, leaves of Gardenia ternifolia epiphyte, stem bark of Gardenia ternifolia's epiphyte, root bark of Annona senegalensis, root bark of Balanite aegyptiaca and root of Boreria verticillata studied are presented in the table. The heavy metals distribution in mg/kg of the selected plants is presented in Table 1. The element content appeared to be

dependent on the type and the part of the plant samples. The leaves of combretum epiphyte have the highest concentration in Mn while the Cd has the least amount of Combretum. Highest concentration of Mn was found in stem/bark of Combretum epiphyte and the least amount was found in Cd. The highest amount of Zinc in leaves of Gardenia ternifolai epiphyte and the least amount were found in Ni and Cr. Highest amount of Zn was found in stem/bark of Gardenia ternifolia epiphyte and the least amount was found in Cd. The root bark of *Annona senegalensis* has the highest concentration of Zn and the least of Ni. The root bark Balanite aegyptiaca has the highest amount in Zn and the least amount was found in Cd. Whereas the graphs shows the highest concentration of Mn in Borerria verticillata root and Cd has the least amount.

Zinc results indicates that the leaves of Combretum epiphyte, stem bark of Combretum epiphyte, leaves of Gardenia ternifolia epiphyte, stem bark of Gardenia ternifolia's epiphyte, root bark of Annona senegalensis, root bark of Balanite aegyptiaca and root of Boreria verticillata has the following concentrations: 35.93 mg/kg ,37.02 mg/kg, 35.82 mg/kg, 34.63 mg/kg, 26.49 mg/kg, 22.84 mg/kg and 29.49 mg/kg respectively. Zinc is essential constituents of enzymes involve in carbohydrate and protein metabolism and nucleic acid synthesis. Its deficiency results in impaired growth and development, skin lesion and loss of appetite [18,19].WHO recommended limit of zinc in medicinal plants is 50mg/kg, while its intake in food is 11mg/day [10].

Cadmium was detected in the entire samples. The amount recorded for cadmium were:0.4mg/kg, 0.2mg/kg, 0.25mg/kg, 0.2mg/kg, 0.85mg/kg, 0.25mg/kg and 0.3mg/kg for leaves of *Combretum epiphyte*, stem/bark of *Combretum epiphyte,* leaves of *Gardenia ternifolia epiphyte,* stem/bark of *Gardenia ternifolia's epiphyte,* root bark of *Annona senegalensis,* root bark of *Balanite aegyptiaca* and root of *Boreria verticillata*, respectively.Cd is extremely toxic even at low concentrations .It causes learning disabilities and hyperactivity in children.WHO recommends level of Cd in medicinal plants is 0.3mg/kg [12]. Leaves of *combretum epiphyte* and root bark of *Annona senegalensis* were slightly above the permissible.

Lead was not detected in all the samples has low geochemical mobility and bioavailability. Its transportation to above ground tissues in plants is minimal due to its retention in roots and precipitation [10]. Lead is toxic metal and non essential element for human body as it causes a rise in blood pressure, kidney damage and miscarriage.

Chromium was present in leaves of *combretum epiphyte*, stem/bark of *combretum epiphyte,* leaves of *gardenia ternifolia epiphyte* and root bark of *Annona senegalens* with the concentrations, 0.65mg/kg, 0.80mg/kg, 0.15mg/kg and 0.60mg/kg respectively.At high concentration chromium is toxic andcarcinogenic.WHO permissible of chromium in medicinal is 1.5mg/kg while its intake dietery is 0.2mg [9]. The levels of chromium in the plant samples were below the permissible level.

Manganese was recorded in all the plants. The highest amount 90.9mg/kg was observed in leaves of combretum epiphyte and the least amount 15.85mg/kg in root bark of Balanite aegyptiaca. Manganese was found below the permissible limit, because its require for growth of animals and plants. Its deficiency produces severe skeletal and

reproductive abnormalities in mammals. High concentration Mn causes hazardous effects on lungs and brains of humans. The maximum permissible of Mn in medicinal plants is 200mg/kg, while its daily intake is 11mg [9].

Nickel was detected in leaves of *Gardenia ternifolia* epiphyte, root bark of *Annonas enegalenses* and root bark of *Balanite aegyptiaca* with their concentrations 0.15mg/kg, 0.35mg/kg and 2.1mg/kg respectively.The concentration of Ni in root bark of *Balanite aegyptiaca* slightly above the tolerable limit which is 0.9mg/kg [9].

Concentrations of Essential Elements: The leaves of Combretum epiphyte, stem/bark of Combretum epiphyte, leaves of Gardenia ternifolia epiphyte, stem/bark of Gardenia ternifolia's epiphyte, root bark of Annona senegalensis, root bark of Balanite aegyptiaca and root of Boreria verticillata were analyzed for the level of Na and K. The result is presented in Table 2. Considerable amount of sodium was recorded in the samples investigated with highest amount in root bark 0f balanite aegyptiaca 750mg/kg and the least amount was found in leaves of combretum epiphyte 110mg/kg. Sodium is one of the chief extracellular ions in the body. It involves in the production of energy, transport of amino acids and glucose into the body cells and its deficiency results in hyponatremia [11].The tolerable limit for Na 123.7 to 264.7mg/kg [7]. The amounts of potassium in the samples investigated as shown in Table 2 recorded the highest amount in leaves of gardenia ternifolia epiphytes and least amount was recorded in root bark of *Balanite aegyptiaca* and all the samples are within the safety limits of 120.0-473.7 [7]. Potassium is the principal intracellular cation. It helps to regulate osmotic pressure and pH equilibrium. The recommended daily intake is 4700 mg [13]. Its deficiency causes muscles weakness, decrease reflex responses and respiratory paralysis.

The concentration of heavy metals such as Cr, Pb, Zn, Mn, Cr, Ni and essential elements such as Na and K are within safe limits as described by [11], except for Cd whose concentration was slightly above the safe limits in root bark of annona senegalensis and leaves of combretum epiphyte. Pb was not detected in the entire samples.

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