DETERMINATION OF LEAD AND CADMIUM IN SOME HERBAL PREPARATIONS SOLD BY HAWKERS IN ZARIA AND ENVIRONS

BY

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A THESIS SUBMITTED TO THE POST GRADUATE SCHOOL, AHMADU BELLO UNIVERSITY, ZARIA IN PARTIAL FULFILMENT OF A REQUIREMENT FOR THE AWARD OF MASTERS OF SCIENCE DEGREE (MSc) IN PHARMACEUTICAL CHEMISTRY.

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DECLARATION

I hereby declared that this research entitled "Determination of lead and cadmium in some herbal preparations sold by hawkers in Zaria and environs" has been carried out by me in the department of pharmaceutical and medicinal chemistry under the joint supervision of Dr. M.T. Odunola and Prof. M. Garba. The information derived from the literature has been duly acknowledged in the text and a list of references provided. No part of this thesis was previously presented for another degree at any university.

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SIGNATURE DATE

CERTIFICATION

This thesis entitled “Determination of lead and cadmium in some herbal preparations sold by hawkers in Zaria and environs" by Muhammad Kabir Shehu, meets the regulations governing the award of the Degree of MASTER OF SCIENCE of Ahmadu Bello University, Zaria and is approved for its contribution to knowledge and literary presentation.

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DEDICATION

This work is dedicated to my parents, wife and children

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All praise be unto Allah the creator and sustainer of the universe. I sincerely wish to express my profound and unalloyed gratitude to Allah who in His infinite mercy gave me the ample opportunity to accomplish this valuable task successfully. 1 also thank him for all His favour on me and my family of whatever magnitude. May His peace and blessings continue to be on His beloved prophet and messenger, Muhammad (S.A.W.), his household, his outstanding righteous companion and all those who trail their path until the end of time.

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ABSTRACT

Most people administer herbal preparations of different types without considering their side effects. Six sites were selected for the study based on the frequency of consumption. The areas were Kofar Doka, Danmagaji, Tudunwada, Sabongari, Kwangila and Samaru. Five different samples were randomly collected from each area of the study. Wet digestion method was employed to obtain clear solution of the sample. Atomic Absorption Spectrophotometric technique was used to quantify the levels of Lead and Cadmium in the digested samples. The results obtained showed that the mean level of Lead in the analysed samples ranges from 0.086 to 0.258mg/l, while that of Cadmium ranges from 0.005 to 0.022mg/l. The levels of Lead and Cadmium were found to exceed the WHO and USEPA permissible maximum guideline limits.

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ABBREVIATIONS

AAS	-	Atomic Absorption Spectrophotometer
ATSDR	-	Agency for Toxic Substances and Disease Registry
ANOVA	-	Analysis of Variance
Cd	-	Cadmium
CNS	-	Central Nervous System
CEC	-	Commission of European Communities
^0C	-	Degree Centigrade (celcius)
df	-	Degree of freedom
EPA	-	Environmental Protection Agency
F	-	Distribution value (Table Value)
HNO_3	-	Trioxonitrate (V) acid
H_2O_2	-	Hydrogen Peroxide
IATC	-	International Agency for Resources of Cancer
ND	-	Not Detected
Ml	-	Milliliter
M	-	Molarity (Molar)
μg	-	Microgram
mA	-	Milliampheres
nm	-	Nanometers
PVC	-	Polyvinyl Chloride

pH	-	Hydrogen ion Concentration
Pb	-	Lead
S.E.M	-	Standard Error of Mean
USEPA	-	United State Environmental Protection Agency

CHAPTER ONE

1.0 INTRODUCTION

1.1 Metals and Non Metals

A metal is a substance, which conducts electricity, has a shiny luster, malleable and ductile. A non metal is a substance which doesn't conduct electricity and is neither malleable nor ductile but brittle (Bentor,2006). Also according to Encarta Encyclopedia (2006), metals are group of chemical elements that exhibit all or most of the following physical properties: they are solid at ordinary temperatures, Opaque, except in extremely thin films; good electrical and thermal conductors; lustrous when polished; and have a crystalline structure when in solid state. Generally, elements are classified as metals and non metals (Atkins and Jones, 1998). At present more than 100 elements have been discovered out of which about 80 of them are classified as metals while 20 have properties of non metals (Miller, 1984). Metals and non metals are separated in the periodic table by a diagonal line of elements. Elements to the left of this diagonal are metals and elements to the right are non metals (Encarta, 2006).

Physically most metals are grayish in colour, but bismuth is pinkish, copper is red and gold is yellow. But some metals display more than one colour such phenomenon is called pleochroism.

The melting points of metals range from about-39^{0}C for mercury to 3410^{0}C for tungsten. Osmium and Iridium (SPG 22.6) are the most dense metals while lithium (SPG 0.53) is the least dense. The majority of metals crystallize in the cubic system, but some crystallize in the hexagonal and tetragonal systems. Bismuth has the lowest electrical conductivity of the metallic elements and silver the highest at ordinary temperature (Encarta, 2006).

Chemically, metal atoms tend to loose electrons and form positive ions, while non metal generally gain electrons to form negative ions. On the other hand, metal have relatively low first ionization energies (Miller, 1984). Thus, metallic elements can combine with one another and with certain other elements, either as compounds, solutions, or as intimate mistures (Encarta, 2006).

According to the periodic table, metals are divided into three categories: alkali metals, alkali earth metals and the transition or heavy metals. The alkali metals are very reactive metals that do not occur freely in nature. These metals have only one electron in their outer most shells and they are ready to lose such electron in ionic bonding with other elements. As with all metals, the alkali metals are malleable, ductile and good conductors of heat and electricity but softer than most other metals (Bentor, 2006).

The alkali earth metals are those found in group II of the period table and thus have two electrons in their outer most shells. These metals are harder and less reactive than the alkali metals. The transition elements or metals are located in between group II and III of the periodic table. In transition metals electrons in both penultimate and the outer most shells participate in chemical bonding with other atoms. Thus, they exhibit several common oxidation states and variable valences (Bentor, 2006). The cadmium (Cd) which is one of the metals to be analysed in this research work is a typical example of a transition metal. The transition metals are paramagnetic and can form large number of tightly bound complex ions (Seinko and Plane, 1981).

Apart from the transition metals, metallic elements could be described as heavy metal or trace elements. The term heavy metal refers to any metallic chemical element that has a relatively high density and is toxic or poisonous at low concentrations. Heavy metals are natural components of the earth crust, they cannot be degraded or destroyed.

To a small extent they enter into our bodies via food, drinking water and air (Lentech, 2006). The toxic heavy metals usually pollute our environment. Thus, the three most pollutants and

dangerous heavy metals are lead, cadmium and mercury (Lentech, 2006).

1.2 Sources Of Metals

Metals usually or naturally exist in form of ores in the earth's crust. These ores are exposed into the surface of the earth through the processes of weathering, erosion and mining (ATSDR, 1993). Traces of these metals can thus be deposited and distributed into rivers, soil and air. It is from these sources that metals accumulate in aquatic inhabitants, plants and animals. At present, there are many sources of exposure to heavy metals. This could come from toxic waste dump, burnsites of agricultural chemical products, mercury amalgam, dental fillings, lead based paints, tap water and chemical residues in processed foods (WHO, 1984). Also personal care products such as cosmetics, mouth wash, tooth paste, soap shampoo and other hair care goods can serve as sources of contamination (Mercola and Droege, 2003).

In todays industrial society, there is no escaping exposure to toxic metals and chemical. This is because it has been found out that a lot of industrial processes released significant amount of toxic metals into the atmosphere. Such industries include; cement, electroplating, fuel combustion, motor vehicles, tobacco, petroleum

and coal product manufacturing, pulp and paper, etc. Thus, as a result of environmental pollution, metals especially the toxic ones are present in the air we breath, the water we drink, the food we eat and to some extent even in the pharmaceuticals we take (Elson and Haas, 1984).

Also according to the environmental technical report by WHO (2002), metals occur in air in different phases, as solids, gases or absorbed to particle having aerodynamic sizes ranging from below 0.01 to 100 micrometers and above.

These particles are emitted from fossil fuel combination, motor vehicle exhaust and wood burning. Several toxic metals including cadmium, lead and arsenic are associated with fine particulate matter in ambient air (U.S. EPA, 1996).

1.3 Toxicity Of Lead And Cadmium

At present, toxic heavy metals such as cadmium and lead have polluted our atmosphere, our waters, our soil and our food chain (Elson and Haas 1984). These heavy metals are taken into the body by inhalation, ingestion and absorption through the skin.

Lead serves no useful purpose in the human body and its presence in the body can lead to toxic effects, regardless of the exposure pathway or route (ATSDR, 1999). Lead toxicity can affect almost every organ and system in our bodies, but the most

sensitive is the central nervous system, particularly in children. Lead also damages kidneys and the immune system (ATSDR, 1999).

Toxicity usually occurs when there is exposure to the toxic substance. Thus, people can be exposed to lead either occupationally or environmentally. The most common occupational exposure to lead is encountered in the manufacturing of lead batteries, paints and colours, lead compound, rubber products and glass, grinding, dicing and cutting by power tools, demolition of old industrial buildings and cutting of lead pipes (Mercola and Droege 2003). While the most common environmental non industrial exposure to lead is via the drinking water, and in communities residing around incinerators, toxic dumps and manufacturing industries utilizing lead and releasing it into the environment. Also environmental exposure to lead could be encountered through leafy vegetables grown in lead contaminated soil, improperly glazed, ceramics, lead crystals and certain herbal folk remedies.

Case studies in the environmental medicine have shown that acute high lead exposure can cause serious physiological effects, including death or long term damage to brain function and organ systems. The effects of lead exposure vary according to exposure time, levels and other factors.

Lead primarily affects the peripheral and central nervous system, renal function, blood cells and metabolism of vitamin D and calcium. Lead can also cause hypertension, reproductive toxicity and developmental effect. Lead also inhibits several enzymes that are critical to the synthesis of heme (ATSDR, 1997).

Also studies by the Environmental Protection Agency (EPA) and other International Regulatory Agencies, have clearly shown that chronic low level exposure to lead is associated with many societal problems, such as brain dysfunction in children exposed to lead in drinking water, neurobehavioral changes in adults, hypertension and chronic kidney disease. Exposure to lead is more dangerous for young and unborn children. Unborn children can be exposed to lead through their mothers. Harmful effects include premature births, smaller babies, decreased mental ability in the infant, learning difficulties, and reduced growth in young children. These effects are more common after exposure to high levels of lead (ATSDR, 1993).

Similarly in adults, lead may decrease reaction time, cause weakness in fingers wrists, or ankles, and possibly affect the memory. Lead may cause anemia, abortion and can damage the male reproductive system (ATSDR, 1993).

Cadmium is a naturally occurring metal which can be found in food, water and cigarette smoke. It is a very powerful and toxic metal that affects a lot of organs and systems in our bodies (Mercola and Droege, 2003).

People are exposed to cadmium not only through foods but also through drinking contaminated water and breathing cadmium-contaminated air such as near burning waste, battery manufacturing, metal soldering or welding. Cadmium is also present in cigarette smoke, thus smoking doubles the average daily in take (Mercola and Droege, 2003). Also according to ATSDR (1989) environmental exposure to cadmium can occur via the diet and drinking water.

Studies indicated that acute oral exposure to cadmium have caused fatalities in humans (ATSDR, 1989) . Exposure to lower amounts may cause gastrointestinal irritation, vomiting, abdominal pain and diarrhea (ATSDR, 1989). Longer term exposure to cadmium primarily affects the kidney, resulting in tubular proteinosis. Also another disease due to cadmium known as “itai-itai” affects the skeletal system (Goyer,1991).

In halation exposure to cadmium and cadmium compounds may cause adverse effects which include headache, chest paint,

muscular weakness, pulmonary edema and to some extent even death could result (USAF, 1990).

One of the greatest effect of cadmium is that it depletes selenium in the body because selenium is essential for cadmium removal (Ghosh and Bhattachaya, 1992). Selenium atoms usually combine with cadmium atoms and are eliminated out of the body via the bile systems. Thus, when selenium is depleted by cadmium, there will be less selenium to form the deiodinase enzymes which convert T_4 to T_3, resulting in Low T_3 and hypothyroidism. Also there is less selenium to form glutathione peroxidase, one of the body's prime antioxidants. This results in greater levels of reactive oxygen species, and hydrogen peroxide, which lead to an increased production of thyroid hormone and damage to the thyroid gland (Ghosh and Bhattachaya, 1992). Thus, cadmium appears to be largest single contributor to autoimmune thyroid diseases.

1.4 Analysis of metals

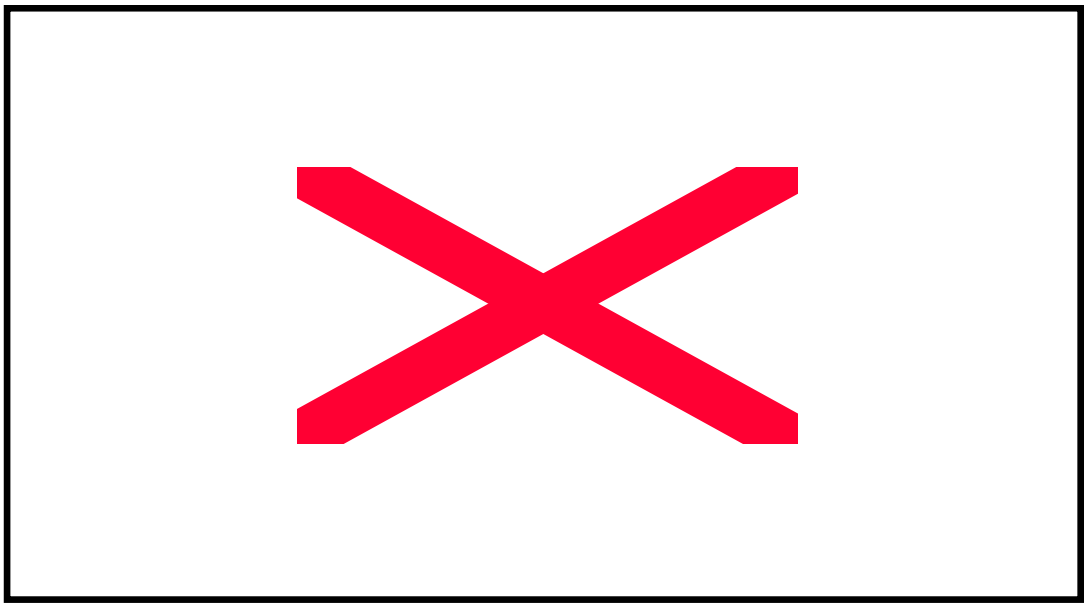
Although there are different methods of metal analyses, but according to Ranganna (2002) Atomic Absorption Spectroscopy (AAS) is an important analytical technique which is widely used in the analyses of metals in foods and other allied products. Crompton (1991) also stated that since its inception in 1955, AAS has been the standard tool employed in the environmental analysis

of trace levels of metals in different samples. In another related statement, Angerer (1992) reported that AAS is at present the most powerful and versatile technique available for determining the levels of naturally occurring heavy metals in different materials. Sahito *et al* (2001) also studied the levels of trace elements in two varieties of indigenous medicinal plants using AAS. The results obtained clearly showed that the method is reliable and can be applied to any type of material.

The principle of operation of atomic absorption instrumentation is relatively simple (Jon, 1996). Radiation from an external light source, hollow cathode lamp which is characteristics of the cathode material, usually a single element (analyte) is emitted. This beam which correspond to the energy for an electronic transition from the ground state to an excited state, is passed through the flame. The flame gases are treated as a medium containing free, un excited atoms capable of absorbing radiation from the external source. This occurs when the radiation corresponds exactly to the energy required for a transition of the test element from the ground electronic state to an upper excited level. The absorbed radiation then passes through a monochromator that isolates the exciting spectral line of the light source and into a detector. The absorption of radiation from the

light source depends on the population of the ground state, which is proportional to the solution concentration sprayed into the flame. Absorption is measured by the difference in transmitted signal in the presence and absence of the test element (Jon, 1996). But according to David *et al* (1983) the proportion of incident radiation which is absorbed by the atoms in the flame is measured and related to the number (concentration) of atoms in the flame in a manner directly comparable to molecular absorption spectroscopy.

David and Hazel (1983) represented a schematic instrumental design of AAS as below:



CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Herbal Preparations

Herbal preparations are those formulations prepared from different parts of plants which are generally used for the relief of symptoms or to cure certain diseases. In herbal medicine, the word herb applies technically to any plant or plant part used for its medicinal, flavouring or fragrant properties. Thus, leaves, flowers, steams, roots, seeds, fruits and bark can all be considered as the constituents of herbal medicine (Kenneth, 2006). Herbs are available in a variety of forms, including fresh, dried, in tablet, capsules or bottled in liquid forms (Rita, 1996).

Herbal remedies or medications have been practiced and used for thousands of years. Today, about 64% of the world population relies solely on herbal medicines. This is because modern pharmaceuticals cannot treat every condition effectively and some drugs have unwanted side effects (Kenneth, 2006). Hence, in the late 20th century herbal medicine made a tremendous come back as people began to seek alternatives to these drugs (Kenneth, 2006 & Sahito *et al.*, 2001).

According to Leslie (2004), traditional herbal medicines are prepared in several ways which usually vary based upon the plant

used and sometimes on the condition that is being treated. Generally, herbal preparations could be in form of: in fusions (hot teas); decoctions (boiled teas) tinctures (alcohol and water extracts); macerations (cold soaking); compress; poultice; creams; essential oils.

The biological or therapeutic activity of a herbal medicine is dependent upon the presence of chemically active ingredients. These ingredients can be classified into major group of chemicals such as essential oils, alkaloids, acids, steroids, tannins and so forth. Each one of these classes of chemicals or natural product may have a preferred effective method of extraction (Leslie, 2004).

As already stated by Leslie (2004) and Rita (1996), some of the active ingredient of medicinal herbs can be extracted and prepared by infusion. This method is used to extract the active principles of herbs through the action of hot water. The preparation of infusions is similar to the way of preparing tea. Infusions are typically used for delicate herbs such as leaves, flowers and fresh tender parts. This technique is the most common and cheapest method of extracting the medicinal compounds of herbs (Rita, 1996).

Decoction is another method of preparing herbal remedies. This method is applied when working with tougher and more fibrous plants such as bark and roots parts (Leslie, 2004). This method is

adopted because, the roots and bark of a medicinal plant are less permeable than the aerial parts and do not liberate their active principles by simple infusion.

Thus, instead of steeping in hot water, the plant material is boiled for a longer period of time to soften the harder woody material and release its active constituent (Leslie, 2004 and Rita, 1996).

In another development, the active ingredient of a medicinal plant is prepared and extracted after using alcohol and water mixture. This type of preparation is refer to as tinctures. This applied when plant have active principles or constituents that are not soluble in water or when a larger quantity is prepared and wanted for longer time storage. Thus, many tincture when properly prepared can last for several years or more without losing potency. The percentage of alcohol usually helps to determine the shelf-life of a particular tincture. The higher the proportion of the alcohol the longer the shelf life. Some times the percentage of alcohol and water is unique to the herbs that are used as some active ingredients are more soluble in alcohol and others more soluble in water (Leslie, 2004).

Maceration is another important method of preparing and extracting the active ingredients of a medicinal plant. This method

of preparation is the easiest, because the extraction is done using cold water.

The fresh or dried plant material is simply covered in cold water and soaked overnight. The herb is strained out and the liquid is taken. This is normally used for very tender plants and/or fresh plants, or those with delicate chemical constituents that might be damaged by heating or degraded in alcohol (Leslie, 2004).

2.2 Lead

Lead is a naturally occurring bluish gray metal found in small amounts in the earth's crust. It has no special taste or smell and can be found in all parts of our environment (ATSDR, 1993). It is present in a number of minerals, the principal one being galena which is chemically known as lead sulphide (WHO, 1984). Human activities such as mining manufacturing and the burning of fossil fuel are the major sources of environmental lead (ATSDR, 1993). Thus, lead is consequently present in air, food, water, soil, dust and snow. In the environment, lead exists almost entirely in its inorganic form, but small amount of organic lead results from the use of leaded gasoline and from natural alkylation's processes that produce methyl lead compounds (Harrison and Laxen, 1978).

Lead has many different uses, but the most important is in the production of batteries. It is also used in ammunition, metal

products (solder and pipes), roofing and devices for shielding x-rays (ATSDR, 1993). For the purpose of good health, lead from gasoline, paints, ceramic, and pipe solder has been reduced significantly in recent years (ATSDR, 1993). The major sources of lead in drinking water are lead plumbing, soil carried into water by rain and wind, and waste water from industries that use lead (Diane, 1994).

With respect to lake and river water, the natural lead content has been estimated to be 1-10 microgram/L, but higher concentrations have been recorded in some areas where contamination occurred as a result of industrial processes (WHO, 1973).

The concentration in finished or treated water prior to its distribution are generally lower than in source waters since lead is partially removed by most conventional water-treatment processes (WHO, 1984). The levels in drinking water can be much higher due to the use of lead service pipes and lead-lined storage tanks (USEPA, 1977). Also high levels of lead can result when the water is soft or has a low pH. These conditions tend to produce the highest level of lead (WHO, 1979 and US EPA, 1980). The current use of lead pipes in some towns and cities in certain countries results in undesirable high levels of lead in the tap-water (NRC, 1977 and WHO, 1979).

2.2.1 Routes Of Exposure To Lead

There are many routes or ways through which humans can be exposed to lead. Among the major sources are drinking lead contaminated water, contaminated food stuff, lead-based paint, leaded gasoline, breathing contaminated air and dust (Mercola and Droege, 2003: WHO, 1984).

According to WHO (1984), it is very difficult to define precisely the average exposure in terms of the concentration of lead in drinking water. This is because of the very wide variation in the levels of lead produced at the tap. These levels depend critically on factors such as stagnation time of the water in lead service pipe or in household plumbing. Even in the same water supply area, there may be considerable home-to-home variations in the level of lead as a result of differences in the length of pipe water, use patterns, and types of deposit that have built up. Based on a water consumption of 2 litres per day, calculations show that the daily in take of lead from water varies from 0.01 - 1mg or more. Such estimates are based on the assumption that all lead is consumed. Since tap water is used for cooking, food preparations and other related processes, it therefore provides additional opportunity for ingesting lead into the body system (WHO, 1984).

Food is another medium or route through which people can be exposed to lead. This is because lead is found to be present in a wide variety of food stuffs. The amounts of lead depends on the type of food. Canned foods were found to contain the highest levels of lead as a result of using lead solders in the manufacturing process. But many fresh vegetables, cereals and fruits contain small quantities of lead as a result of limited absorption of the metal from the soil in which they are grown (Drill *et al*., 1979). It has been estimated that the daily intake of lead ranges from less than 100 to over 500μg/day (USEPA, 1977; WHO, 1977 and 1972).

But the world wide average of daily intake for adults is estimated at about 200μg/day. The intake of lead by women is generally less than that of men. This is because women usually eat less than their male counterparts (WHO, 1984). It has been estimated that children aged 1-5 years ingest about 90μg of lead per day (Drill, 1979). Additional lead in food can arise from contamination by cooking vessels, such as pots that have soldered joints and some lead comes from tap water used for preparing food. Generally, the major source of ingested lead is food (WHO, 1984).

Air is also another route through which lead can be inhaled into our body. It has been found out that in rural areas, the average level of lead is 0.1$\mu g/m^3$, while in urban areas the level ranges from 0.5 - 2$\mu g/m^3$ (WHO, 1977 and 1973; Drill, 1979).

The levels of lead in a particular area depend on the type and extent of the emission source. At present, most of the airborne lead in non-industrialized cities comes from motor vehicles traffic. Generally, people living near busy highways will be most exposed. In some industrialized areas, average ambient air levels as high as 6 microgram/m^3 have been recorded (WHO, 1973 and 1977). Most of the lead in the air is in form of fine particles. When these particles are inhaled, only 20-60% will be deposited in the respiratory system (WHO, 1977). The deposited lead from air usually contaminates the soil and any other substance that is exposed.

Studies also indicated that soil, dust and house hold paints contain elevated levels of lead. Thus as a result of the “hand to mouth” activity of some people particularly the young children, they may be significantly exposed to such sources (WHO, 1973 and 1977).

2.2.2 Metabolism Of Lead

Elemental lead and inorganic lead compounds are usually absorbed through ingestion or inhalation. Organic lead such as

tetraethyl lead, is absorbed to a significant degree through the skin. Pulmonary absorption of lead is efficient and depends on the size of lead particles and on the depth as well as the rate of breathing (EPA, 1977 and WHO, 1977). Children absorb up to 50 percent of the amount of lead ingested, whereas adults absorb only about 10 - 20 percent. The values depend on whether the contaminated food or water is consumed on a full or an empty stomach. This is because gastrointestinal absorption of lead is enhanced by fasting (Rabinowitz, 1974).

This finding has also been confirmed in mice (Garber, 1974). Absorption of lead from the gastrointestinal tract is also influenced by some other factors such as presence of calcium, phosphorous, iron, copper and Zinc in the diet (EPA, 1980, WHO, 1977 and Underwood, 1977).

The absorbed lead enters into the blood stream or plasma and becomes distributed into soft tissues and bone. In the blood, around 95 - 99% of the lead is sequested in red cells, where it is bound to hemoglobin and other components. After a prolonged exposure an equilibrium is reached between the blood and soft tissues (WHO 1984). The largest proportion of the absorbed lead is incorporated into the skeleton, which contains more than 90 percent of the body's total lead burden, (Underwood, 1977).

Lead is excreted mainly in the urine and in the feces. The urine excretion of lead depends on the glomerular filtration and tubular secretion. Lead also appears in hair, nails, sweat, saliva and breast milk, (WHO 1980). The respective half-lives of lead in blood, soft tissue and bones have been estimated to be 25 days. (USEPA 1977, WHO 1977 and Rabinowtz, 1974), 40 days (Rabinowtz, 1974) and 27years (Drill, 1979).

2.2.3 Health Effects Of Lead

In humans, exposures to lead can result in a wide range of biological effects depending on the level and duration of exposure. Lead in high doses has been recognized for centuries as a cumulative general metabolic poison. Thus, various effects occurs over abroad range of doses, with the developing foetus and infant being more sensitive than the adult (Lentech, 2006 and WHO, 1984). High levels of exposure may result in toxic biochemical effects in humans which in turn cause problems in the synthesis of haemoglobin, effects on the kidneys, gastrointestinal tract, joints and reproductive system, and acute or chronic damage to the nervous system. The health effects of lead vary according to exposure timing and levels, and other factors, although some effects may be latent (ATSDR, 1999).

According to a case study conducted by ATSDR (1999), the nervous system is the most sensitive target of lead exposure. The neurologic effects of lead is more serious in feutuses and young children. This is because their brains and nervous systems are still developing and the blood-brain barrier is incomplete. Thus, neurologic effects in children generally occur at lower blood levels than in adults.

Also according to another report by ATSDR (1993), the central nervous system (CNS) becomes severely damaged at blood lead concentration of 40microgram/dL, causing a reduction in nerve conduction velocities and neuritis. It has also been suggested that lead may cause some neurological and behavioural effects in adults. These effects include malaise; forgetfulness; irritability; lethargy; impaired concentration; depression and mood changes; impotence; increase nervousness; parasthesia; dizziness; and cognitive performance (ATSDR, 1999).

In another related studies, it has been noted that lead exposure can result into renal effects such as Fanconi-like syndromes, chronic nephropathy and gout. These effects can be seen in morphological changes in the kidney epithelium, increase in excretion rates of many different compounds, reduction in

glomerular filtration rate and an altered plasma albumin ratio (Goyer, 1985).

It has been found out that exposure to lead contributes significantly to the onset of anemia. This is because lead inhibits several enzymes that are critical to the synthesis of heme, causing a decrease in blood hemoglobin (ATSDR, 1999). Lead at low levels can reduce the activity of an enzyme, porphobilinogen synthase. This enzyme is involved in normal heme synthesis at the stage of conversion of aminolevulinic acid to porphobilinogen. Thus, a decrease in the activity of this enzyme may be used as an index of exposure to lead.

Lead can induce two types of anemia which are often accompanied by basophilic stippling of the erythrocytes (ATSDR, 1999). The Hemolytic anemia which manifests Immediately after the exposure is associated with acute high – level of lead . While frank anemia is evident only when the blood level of lead is significantly elevated for along period of time (ATSDR, 1999). Also the impairment of heme synthesis by lead can affect other processes in the body such as neural, renal, endocrine and hepatic pathways.

Koo *et al* (1991) indicated that a strong inverse correlation exists between blood levels of lead and vitamin D. They further stated

that lead interferes with a hormonal form of vitamin D which affects multiple processes in the body including cell maturation and skeletal growth. Lead impedes vitamin D conversion into its hormonal form, 1, 25 – dihydroxy vitamin D, which is largely responsible for the maintenance of extra cellular and intral cellular calcium homeostasis. But these adverse effects seems to be restricted to children with chronically high blood level of lead. Thus, Rosen, *et al* (1980) noted that in lead exposed children with blood levels of 33 - 55 microgram/dl, 1, 25 - dihydroxy vitamin D levels were reduced to levels comparable to those observed in children with severe renal in sufficiency.

A relationship between lead exposure and hypertension has also been established. Lead exposure has been recognized as one of the factors that may contribute to the on set and development of hypertension (Victery, *et al*., 1988, Schwartz, 1995 and Korrick *et al.,* 1999). Hu (1991) also noted that adults who experienced lead poisoning as children had a significantly higher risk of hypertension 50 years later. Also studies of veterans and nurses revealed that increase odds of hypertension have been associated with the higher levels of lead.

Recent reproductive function studies in humans suggested relationship between lead exposure and certain reproductive and

developmental outcomes (ATSDR, 1997). Thus, Alexander, *et al.,* (1996) indicated that current occupational exposure to lead may decrease sperm count and increase abnormal sperm frequencies. While long term lead exposure may also diminish sperm concentrations, total sperm counts and total sperm motility. These effects may begin when the blood level of lead reaches 40μg /dl (ATSDR, 1999). It has also been observed and noted that exposure to lead increases the frequency of miscarriages and stillbirths among women. Studies of women living in the vicinity of lead smelter versus those living some distance away have shown an increase frequency of spontaneous abortions (Nordstrom *et al.,* 1979) and miscarriages with still births (Baghurst *et al*., 1987 and Mc Michael *et al*., 1986). Results of another resent retrospective study indicated that women who experienced overt child – hood lead poisoning 50 years earlier may have also experienced a higher rate of spontaneous abortions and miscarriages (HU, 1991).

2.3 Cadmium

Cadmium is a naturally occurring metal which is uniformly distributed in trace amount in the earth's crust. It is not usually found in pure state, but inform of a mineral, greenokite (cadmium

blend) which is chemically known as cadmium sulfide (ATSDR, 1989 and WHO, 1984).

Practically, all Zinc ores contain small amount of cadmium. Thus, it is obtained usually as a by product of Zinc extraction. As a result of pollution, cadmium contaminates our environment, thus it is found in air, food, soil, plants and water. Cadmium is also present as an impurity in several products such as phosphate fertilizers, detergents, refined petroleum products, insecticides and fungicides (ATSDR, 1989). Strong evidence also indicated that cadmium is present in tobacco or cigarette smoke. Hence, it has been estimated that one cigarette normally contains 1 - 2μg (WHO, 1984).

Cadmium has a limited number of applications, but is used for a variety of consumer and industrial materials. It's principal uses includes: protective plating for steel, stabilizers for polyvinyl chloride (PVC), pigments in plastics and glasses, electrode material in nickel-cadmium batteries and as component of various alloys (WHO, 1992).

Cadmium compounds have varying degrees of solubility ranging from very soluble to nearly insoluble. The solubility is influenced by the nature of the source of the cadmium and the acidity of the

medium. The solubility affects both the absorption and toxicity of the metal (ATSDR, 1989).

It has been reported that the level of cadmium in public water supply is normally very low. This is because only tiny amount of this metal exist in raw water and even where the levels are elevated, many conventional water-treatment processes will remove much of the cadmium (NRC, 1977). But where the level is high despite the treatment processes it could be due to the use of plated plumbing fittings, silver-base solders and galvanized iron piping materials (WHO, 1984). Generally, the levels of cadmium in unpolluted waters have been found to be less than 1μg /L (Hiatt and Juff, 1975).

2.3.1 Routes Of Exposure To Cadmium

Humans are exposed to cadmium through a different ways or media. The fact that cadmium is found in the water we drink, the food we eat, the air we breath and in some medicinal plants we use (NRC, 1977, ATSDR, 1989 and Sahito *et al.,* 2001), it can therefore easily get into our body systems.

Drinking water normally contains very low concentrations of cadmium, of the order of 1μg/l or less (NRC, 1977 and WHO, 1972). But occasionally, levels up to 5μg/l have been reported

(NRC, 1977) and on rare occasions levels up to 10μg/L have been detected (WHO, 1972). In some areas well water may contain elevated concentrations of cadmium (NRC, 1977). Thus, estimated daily exposure to cadmium via water, based on a water consumption of 2 litres per day, ranges from substantially less than 1 microgram to over 10 microgram per day. These estimates are based on the presumption that all the cadmium from the water is ingested (WHO, 1984).

Report has already shown that food products account for most of the human exposure to cadmium, except in the vicinity of cadmium-emitting industries (Kaneta *et al.,* 1986).

Most foods stuffs contain traces of cadmium, also crops grown in polluted soil or irrigated with polluted water may contain increase concentrations, as may meat from animals grazing on contaminated pastures (WHO, 1984). Thus, people who eat such foods will tend to ingest more cadmium in to their systems.

It has also been noted that air is another medium through which humans can be exposed to cadmium (USAF, 1990). But the levels of cadmium in ambient air are generally found to be low (WHO, 1984). Long term average concentrations may vary from less than 0.001 to 0.5μg/m^3 depending on the degree of industrialization and presence of cadmium emitting industries. It has been estimated

that members of the general population will generally inhale less than 0.05 microgram/day (WHO, 1984). But for unusually polluted areas, maximum values as high as 3.5 microgram/day have been estimated.

Another source of exposure to cadmium is cigarette smoke. This is because it has been reported that cadmium is present in tobacco where by one cigarette normally contains 1 - 2 microgram. At elevated temperature, cadmium is volatile, thus some of the metal will be inhaled during smoking (WHO, 1984), and probably 50% of this will be deposited in the lungs.

2.3.2 Metabolism Of Cadmium

Cadmium is absorbed into the body system through ingestion or through the lungs. But research has shown that cadmium is more efficiently absorbed from the lungs than from the gastrointestinal tract (ATSDR, 1989).

The absorption efficiency is a function of solubility of the specific cadmium compound as well as its exposure concentrations and route. Alimentary absorption is affected by a number of factors, such as age, calcium, iron, zinc and protein deficiency (WHO, 1984).

The state of the stomach is also likely to influence the amount absorbed, with a fasting stomach probably providing the maximum

up take in contrast with a full stomach. Dietary factors, such as iron, calcium and protein deficiency, may increase the gastrointestinal absorption rate (Flanagan, 1978).

The absorbed cadmium is transported in the blood by red blood cells and high-molecular weight proteins such as albumin (Goyer, 1991) and be come concentrated in certain parts of the human body (Underwood, 1977).

According to a report by Goyer (1991) both the liver and the kidneys act as stores of cadmium (about 50% of any accumulated cadmium is found in these organs). The cadmium is to a large extent bound to a protein of low relative molecular mass, known as metallothioneine which is believed to be playing a vital role in cadmium transport and absorption (WHO, 1980).

Cadmium has a long biological half life in the body (13 - 38 years) and accumulates with age. It has been found that, the new born are virtually free of cadmium (NRC, 1977) because the placenta acts as a fairly efficient barrier to cadmium (ATSDR, 1989).

As with most metallic elements, there is little or no direct metabolic conversions of cadmium in the body, but rather binding to various biological components, such as protein and non protein sulfhydryl groups and anionic groups of various macromolecules (ATSDR, 1989). The binding of cadmium to the protein, metallothionein is of

special importance and instrumental in determining the disposition of cadmium in the body.

Studies have shown that excretion of cadmium from the body system is usually rather slow. The principal route of excretion is via the urine, with average daily excretion of about 2 - 3 microgram in humans (ATSDR, 1989). On a group basis, urinary cadmium is generally regarded as a good indication of the body burden. The unabsorbed cadmium is removed from the gastrointestinal tract by fecal excretion.

Typical daily cadmium excretion has been reported to be about 0.01% of the total body burden. Thus, the daily excretion represent only small percentage of the total body burden (Tsuchiya *et al.*, 1972; Friberg *et al*., 1974).

2.3.3 Health Effects Of Cadmium

The health effects of cadmium exposure in both animals and humans have been studied extensively (NRC, 1977). In humans, it has been reported that most of the severe cases of oral cadmium toxicity have been associated with ingestion of foods or fluids which are contaminated with cadmium.

Thus, acute effects have been identified where foods or fluids have been contaminated by cadmium from plated vessels or containers. High doses of cadmium are known to cause gastrointestinal

irritation resulting in vomiting, abdominal pain and diarrhea (ATSDR, 1989). Lauwerys (1979) reported that the emetic thresh hold for cadmium in drinking water was about 16 microgram/L and CEC (1978) reported that 3 microgram/L was an emetic thresh hold. Health effects have also been demonstrated in industrial workers heavily exposed to cadmium oxide fumes and dust (CEC, 1978).

In line with this, bronchitis, emphysema, anemia and renal stones have been reported (NRC, 1977). The renal cotex has been generally accepted as the most critical organ for cadmium accumulation in man. Friberg *et al*., (1974) estimated that this critical effect which is characterized by tubular proteinuria will not occur in humans until the cadmium concentration in the renal cotex exceeds 200μg. In another report, it has been stated that the classical renal effects of cadmium poisoning are associated with proteinuria, glucosuria and amino aciduria. But where the exposure is high, irreversible renal injury could occur (CEC, 1978). Dietary intake of cadmium has also been implicated in osteomalacia, osteoporosis and spontaneous fractures. These conditions are collectively known as “itai-itai” (Ouch-Ouch) disease and originally documented in post menopausal women in cadmium contaminated areas of Japan (Friberg, *et al*, 1974). Cadmium

exposure has also been implicated in hypertensive disorders, but this conditions is currently not thoroughly understood or verified (ATSDR, 1989).

There have been many studies in which experimental animals have been dosed with cadmium. There is evidence of hypertension after long term low level oral exposure and teratogenic, mutagenic and carcinogenic effects after injection of high doses (CEC, 1978). Also it has been noticed that short term administration of cadmium in drinking water at a level of 10 microgram/L has been associated with partial inhibition of the gastrointestinal absorption of iron (CEC, 1978).Stowe *et al*., (1972) noted that exposure of rabbits to 1.5mmol cadmium chloride in drinking water produced histological alternations in the liver but no clinical signs of toxicity. In another study by Kotsonis and Klaassen (1978), rats exhibited proteinuria after receiving cadmium chloride in drinking water for six weeks at 30 or 100μg/L. Evidence for cadmium-induced immuno toxicity in animals is also available. This is because Koller *et al*. (1975) noted a decrease in the number of spleen placque-forming cells in mice receiving cadmium at 0.6μg/kg/day for 10 weeks and Blakley (1985) reported a dose-dependent suppression of the humoral

immune system in mice receiving cadmium in drinking water at concentrations of 5 - 50μg/L for three weeks.

According to USAF (1990), inhalation of cadmium fumes or dust may result in a wide range of effects, including a metallic taste, headache, dyspnea, chest pains, cough with foamy or bloody sputum and muscular weakness. Severe exposure may result in pulmonary edema and death. If the pulmonary edema is resolved, late occurring kidney or liver damage may develop. Friberg *et al.,* (1975) indicated that exposure to 1μg/m^3 of cadmium for 8 hours is instantly dangerous to humans and the world health organization (WHO, 1980) identified 0.5μg/m^3 of cadmium as the thresholds for respiratory effects resulting from 8-hours exposure.

It has been reported that cadmium is also a probable human carcinogen. This report is based on a limited evidence from multiple occupational exposure studies and adequate animal data (ATSDR, 1989). But in another development, the evidence that cadmium may be carcinogenic to man is rather weak (IARC, 1976), although similar report stated that prolonged and heavy industrial exposures may constitute an increased risk of prostate cancer (Kipling and Waterhouse, 1969; Leman 1976).

2.4 Scope And Objective

Plants play an important role in our life. Plants not only provided us with nutrition but also have some medicinal values. Thus, a large number of herbs and plants are widely used for the treatment of various disease (Karamat *et al.,* 2002).

Presently, many patients have been turning to traditional medicine as an alternative way in search of the relief which the orthodox medical care has not been able to provide (Sahito *et al.,* 2001). As a result, medicinal plants are now increasingly being used in raw, semi processed and polypharmaceutical forms as medicine which is playing a significant role in providing relief to a great number of people. Studies have shown that most of the medicinal plants used in herbal formulations contain certain amount of trace elements or heavy metals (Karamat *et al.,* 2002). This was evidently observed in some medicinal plants such as catharantus roseus (Sahito *et al.,* 2001), cymbopogan Jwarancusa (Karamat *et al.,* 2002) and Ayurvedic medicinal plants (Rajukar *et al.,* 1998).

This study is only limited to the traditional, herbal formulations that are now being frequently prepared and sold by hawkers in Zaria and its environs. Thus, the research is being focused towards the

quantitative estimation of the levels of cadmium and lead in the ready made samples of such formulations.

These formulations are according to the hawkers being locally used for the treatment of different types of ailments such as typhoid, malaria, haemorrhoids (pile), stomach problem, backache, etc.

Thus, the aims and objectives of this research are as follows:-

i. to establish the presence or otherwise of cadmium and lead in the herbal formulations.
ii. to quantify the levels of cadmium and lead in the samples collected.
iii. to correlate the levels of such metals with that of WHO standard.
iv. to inform the general public about the health hazard associated with the local administration of any such herbal formulations.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Materials

3.1.1 Reagents

Trioxonitrate (v) acid (conc.). Analar grade, VWR international Ltd. Poole BH151 TD, England.

Hydrogen peroxide (30%). BDH chemical England.

Lead trioxonitrate (v). Analar grade, Hopkins and Williams England.

Cadmium trioxonitrate (v). Analar grade, May and Baker, England

Deionized water. Obtained from Kaduna Polytechnic.

3.1.2 Equipment And Glassware

Atomic Absorption Spectrophotometer (AAS), solar 32 unicam 969 model with it's accessories.

Analytical weighing balance type Acculab AL-64.

pH – meter, fisher brand hydrus 300 model.

Hot plate, stuart scientific model.

Beakers, 50ml, 100ml and 250ml

Volumetric flask, 100ml

Measuring cylinder, 200ml

Pipette, 10ml, 20ml

Test – tubes,10ml, 20ml

Filter paper, Wattman No.1

Universal indicator paper

Polyethylene bottles, 100ml

3.2 METHODS

3.2.1 Sample Collection

The sampling areas were: Kofar Doka, Danmagaji, Tudun Wada, Sabongari, Kwangila and Samaru. The samples collected comprised of those used in the treatment of typhoid, stomach problem, Yellow fever, haemorhoids (pile) and waist pain.

The samples, were collected in a clean polyethylene bottles and labeled accordingly so as to avoid any form of misrepresentation.

The samples were then kept in a refrigerator at 10^0C while awaiting analysis.

3.2.2 Preparation Of The Calibration Curve

3.2.2.1 Lead

A stock solution of 1000mg/L was prepared using lead trioxonitrate (v) salt (1.599g/L). A 10mg per litre solution was then prepared from the stock. Standard working solutions ranging from 0.1 to 0.5 mg/L were

prepared by serial dilution of the resulting solution using deionized water (to preserve the solution, 100ml of each standard solution was taken into a beaker and the pH was adjusted to 2.5 by adding few drops of 1M $HN0_3$).

Each of the standard solutions was then aspirated into the atomic absorption spectrophotometer and the corresponding absorbance was read directly. The nebulizer, atomizer and burner were flushed each time with deionized water after each standard solution was aspirated before the next one. The instrument's stability was also checked at interval by introducing the highest working standard solution and the blank.

3.2.2.2 Cadmium

For the preparation of cadmium standard curve, a stock

solution of 100mg/L was prepared using an analar grade of cadmium trioxonitrate (v) salt (0.2107g/L). Then standard working solutions were prepared by serial dilution using deionized water. Also to preserve the solution, 100ml of each standard was taken into a clean beaker and the pH was adjusted to 2.5 by adding few drops of 1M $HN0_3$.

Then each of the standard solutions were aspirated directly into the atomic absorption spectrophotometer and the absorbances were read.

3.2.3 Sample Preparation And Treatment

The herbal samples collected were prepared using a wet ash digestion procedure (Sahito *et al.*, 2001; Yaman and Bakirdere, 2002). All

glasswares used were thoroughly washed and allowed to dry in an oven. A 100ml aliquot of each herbal sample was measured and poured into a separate 250ml beaker. The beakers were then placed on a hot plate and heated until the water content evaporated to dryness. The residue was allowed to cool after which 5ml of deionized water was added to dampen the residue. A 20ml of concentrated trioxonitrtae (v) acid was added followed by heating under reflux at 100^0C for about 2hrs.

The sample was allowed to cool, then 10ml of hydrogen peroxide (H_20_2) was added and the mixture was heated again on a hot plate until a clear digest was obtained. The residual mixture was further diluted with 20ml of deionized water and filtered. The filtrate was then transferred into a 100ml volumetric flask and made up to the mark with deinozed water. Each of the digest solution was poured into a polyethylene bottle, labeled accordingly and stored for atomic absorption analysis.

3.2.4 Determination Of Lead

The lead contents of the digested samples were determined using atomic absorption spectrophotometric technique (Sahito *et al.,* 2001; Ranganna *et al.,* 2002; Yaman and Bakirdere, 2002). The AAS was equipped with hollow cathode lamps of lead and the condition of operation were as follows:

1. Spectral line: 217nm
2. Lamp current: 10mA
3. Fuel type: Air/acetylene
4. Band pass: 0.5nm

The instrument was switched on and allowed to warm up for 15 minutes.Then the flame was switched on and allowed to stabilize for 10minutes. All the necessary adjustments were made to achieve the most sensitive response for the element to be analysed. The system was aspirated or flushed with deionized water. Then the sample solutions were aspirated one after the other. But the system was being flushed each time with deionized water after each sample was aspirated before the next one.

3.2.5 Determination of Cadmium

The Cadmium contents of the digested samples were also determined using AAS method (Sahito *et al.,* 2001; Ranganna *et al.,* 2002; Yaman and Bakirdere, 2002). The AAS was equipped with cadmium hollow cathode lamp and the conditions of the operation were as follow:

1. Spectral line: 228.8nm
2. Lamp current: 10mA
3. Fuel type: Air/acetylene
4. Band pass: 0.5nm

After setting all the conditions, the instrument was operated as described in **3.2.4** above.

3.2.6 Data Analysis

Analysis of variance (ANOVA) was used to determine whether there is any significant difference between the mean concentrations of lead collected from the various locations of Zaria and environs. Also the same method was applied to see whether there is any variation in the mean concentrations of cadmium across the various locations of the sampling points.

CHAPTER FOUR

4.0 RESULTS

4.1 Lead contents of the different herbal formulations

Formulations for the treatment of yellow fever which was collected from Danmagaji area was found to contain the highest concentration of lead (0.424mg/l),while formulation for the treatment of pile collected from Kwangila area contains the least (0.029mg/l).

But on average basis, formulation for the treatment of waist pain contains the highest mean level of lead, (0.258mg/l) where as the lowest mean level (0.086mg/l) was detected in the formulation for the treatment of Typhoid.

Table 1 shows the individual as well as the mean concentrations of lead in the analyzed samples.

Table 1: Concentrations of Lead (mg/l) in the different herbal formulations sold by hawkers in Zaria and environs.

LOCATIONS	HERBAL FORMULATIONS					
	A typhoid	B Stomach Ache	C Yellow Fever	D Pile	E Waist pain	Mean± S.E.M
Kofar Doka	ND	ND	0.138	0.128	0.311	0.115±.0.057
Tudun wada	0.191	0.290	0.107	0.230	0.211	0.206±0.029
Sabon Gari	0.126	0.135	0.153	0.094	0.257	0.153±0.027
Danmagaji	ND	0.289	0.424	0.244	0.322	0.256±0.070
Kwangila	0.085	0.244	0.211	0.029	0.136	0.141±0.039
Samaru	0.115	0.054	0.145	0.071	0.313	0.140±0.046
Mean ±	0.086	0.1689	0.196	0.133	0.258	
S.E.M	0.031	0.051	0.048	0.036	0.030	

4.2 Cadmium contents of the various formulations

Based on the findings of this analysis, significant amount of cadmium was detected in seventeen (56.7%) out of the thirty (30)

samples collected for the research. Thirteen (13), i.e (43.3%) of the sample formulations were found to be free of cadmium. On a more specific basis, the highest concentration (0.099mg/l) was detected in the concoction for the treatment of stomach ache, while the lowest concentration (0.004mg/l) was detected in formulation for the treatment of pile at Sabon Gari area. But generally, yellow fever formulation has the highest mean level of cadmium (0.028mg/l), while formulation for the treatment of waist pain has the lowest mean level (0.005mg/l). All of the herbal formulations collected at Danmagaji area where found to be free of this toxic metal. Thus, Table 2 clearly shows the individual as well as the mean concentrations of cadmium detected in the five different types of formulations sold by hawkers at various locations in Zaria and environs.

Table 2: Concentrations of cadmium (mg/l) in the different herbal formulations sold by hawkers in Zaria and environs.

LOCATIONS	HERBAL FORMULATIONS					
	A typhoid	**B stomac Ache**	**C Yellow Fever**	**D Pile**	**E Waist Pain**	**Mean ± S.M.E**
Kofar Doka	ND	0.010	0.023	0.010	ND	0.008±0.004

Tudun wada	0.042	ND	0.060	0.023	ND	0.025±0.011
Sabon Gari	ND	0.030	ND	0.004	0.031	0.013±0.007
Danmagaji	ND	ND	ND	ND	ND	0.000±0.000
Kwangila	0.019	0.099	0.018	0.036	ND	0.035±0.017
Samaru	0.069	0.069	0.066	0.029	ND	0.047±0.013
Mean ± S.E.M	0.022 0.012	0.020 0.011	0.028 0.012	0.017 0.006	0.005 0.005	

4.3 Comparison of the levels of lead and cadmium (mg/l) at sample point locations

On the comparative basis, the mean lead contents detected in the herbal formulations were significantly different from those of cadmium. Generally the mean lead contents were higher than the mean levels of cadmium in almost all of the samples analyzed. The only exception was observed specifically in one of the samples

Fig. 2: Mean Concentrations of cadmium (mg/l) in the different formulations

collected at Kofar Doka where the level of cadmium was higher than that of lead.

Table 3 gives a summary for the comparison between the level of lead and cadmium at different sample point locations.

Table 3. Levels of Cadmium and Lead at sample point locations (mg/l)

	Kofan doka		**Tudunwada**		**Sabongari**		**Kwangila**		**Samaru**		Danmagaji	
Samples	Pb	Cd	Pb	Cd	Pb	Cd	Pb	Cd	Pb	Cd	Pb	Cd
Typhoid	ND	ND	0.191	0.042	0.126	ND	0.085	0.019	0.115	0.070	ND	ND
Stomach Ache	ND	0.010	0.290	ND	0.135	0.030	0.244	0.009	0.054	0.070	0.290	ND
Yellow fever	0.138	0.023	0.107	0.060	0.153	ND	0.211	0.018	0.145	0.067	0.424	ND
Pile	0.128	0.010	0.230	0.023	0.094	0.004	0.029	0.036	0.071	0.029	0.244	ND
Waist Pain	0.311	ND	0.210	ND	0.257	0.031	0.136	ND	0.313	ND	0.322	ND

4.4 Comparison of the mean concentrations of lead at different locations

One way ANOVA was used to test whether the mean concentrations of lead were significantly different in the various formulations collected from different areas in order to study the effect of the environmental factors. Table 4 shows the results obtained for this correlation.

Table 4: Analysis of variance (ANOVA) summary table for the lead content (mg/l)

Sources Of variation	Sum of square	Df	Mean square	F	Sig.
Between Groups	0.101	4	0.025	2.652	0.057
Within groups	0.239	25			
Total	0.340	29			

4.5 comparison of the mean levels of cadmium in the different formulations for the various sampling areas

Similarly, one way ANOVA was applied to test whether the mean levels of cadmium were significantly different in the various formulations sampled from the six areas of Zaria and environs. Table 5 clearly illustrated the summary of such findings.

Sources Of variation	**Sum of square**	**Df**	**Mean square**	**F**	**Sig.**
Between Groups	0.002	4	0.000	0.765	0.558
Within groups	0.014	25	0.001		
Total	0.015	29			

CHAPTER FIVE

5.0 DISCUSSION

Considering the results obtained from the quantitative estimation of Lead (Table 1), it shows that appreciable amounts of this metal have been detected in almost 90% of the analyzed samples. But the remaining 10% of the samples being analysed were found to be free of this toxic metal. This shows that majority of herbal preparations have been contaminated with this toxic metal which could be as a result of using contaminated water during preparation or due deposition of contaminated dust on the exposed herbal materials. The sample of the herbal preparation used for the treatment of waist pain contains the highest mean concentration of lead (0.258mg/l). While the least mean level (0.086mg) was recorded in the sample for the treatment of typhoid fever. But on the individual point of view, the sample for treatment of yellow fever which was collected from Danmagaji area was found to contain the highest level of lead (0.424mg/l). While the least level (0.029mg/l) was detected in the sample for the treatment of pile which was collected from Kwangila area. The highest level of lead in this instance could be attributed to the influence of environmental pollution, which contributed to the presence of this metal in the study area. However, levels of lead in

most samples exceeded that of the WHO guideline limit of 0.01mg/l. This is not unexpected because previous research conducted on the level of lead in some food and water samples also recorded an elevated levels of this toxic metal (Musa *et al.,* 2004, Lawal *et al.,* 2006 and Odunlola, 2006).

The presence of lead in these sample preparations could be attributed to factors like discharge of lead through industrial effluents, it's use in batteries, paints, roofing and piping materials. Also the use of lead as an additives in petrol is another factor of great concern, because the metal is emitted through the exhaust fumes of machineries into the environment, (ATSDR, 1993 and WHO, 1994).

The presence of lead in the analysed samples could also be attributed to the way and manner such formulations are being prepared and handled. Almost all of these formulations are being prepared using water as a solvent or medium. Such water might have already been contaminated with lead and

when mixed with the herbal material it significantly increased the amount of lead in the formulations. The part of plant materials that is being used for the preparation may itself contain significant amount of lead. This could be true if the plant was grown in the contaminated soil (Diane, 1994). Lead in air may also become

deposited on the part of plant materials to be used when it is not properly stored or kept (ATSDR, 1993). Similarly, the presence of lead in the analyzed samples might be connected to the type of containers used during the course of preparation. This is because according to Diane (1994), some dishes or pot contain significant amount of lead on their surfaces, thus when acidic food or drinks are used they can easily leach out the lead content into the solution. This is not surprising as the pH of most of these preparations fall within the acidic range.

On comparing the mean levels of lead content of the analysed samples using ANOVA (Table 4), the results revealed an insignificant statistical difference ($P>0.05$) in the mean values across the various samples.

With respect to the cadmium content, Table 2 indicated that 56.7% of the analysed herbal formulations contain significant amount of the metal. While 43.7% of the remaining samples analysed have been found to contain no any cadmium. On the individual point of view, the sample for the treatment or remedy of stomach ache which was collected from Kwangila area contains the highest level (0.099mg/l) of cadmium, but the samples formulation for the

treatment of pile (haemorrhoids) collected from Sabongari area has the least value (0.004mg/l).

In another case, all the herbal samples collected from Dan Magaji study site were found to be free of cadmium. This is quite interesting and it could be as a result of insufficient sources of cadmium in this location of study.

Generally, the samples formulations for the treatment of yellow fever was found to contain the highest mean level (0.028mg/l) of cadmium, while the least mean level (0.005mg/l) was detected in the samples preparation for the treatment of waist pain.

The detection of significant amount of cadmium in this research is not surprising, because the results obtained are in quite agreement with the findings of the previous research (Sahito *et al.,* 2001 and Lawal *et al.,* 2006). The results also further confirm the findings of Elson and Ha'as (1984) who reported that metals especially the toxic ones are present in the air we breath, the water we drink, the food we consume and to some extents even in the pharmaceuticals we use.

The presence of cadmium in the analyzed samples could be attributed to many factors among which include the use of cadmium in phosphate fertilizer which can eventually contaminate the soil where such herbs are being grown (ATSDR, 1989). Also

the use of cadmium in batteries serves as another factor that can introduce significant amount of such metal into our environment. The release of cadmium from this source can easily contaminate the source of water being used in the preparation of the herbal formulations and at the same time it can also contaminate the soil, where the herbs are growing. Application of cadmium in some alloys (Lenntech, 2006) could on another way round serves as a contributory factor through which the metal is introduced into our environment. Thus, these alloys can serve as agents through which our household products could become contaminated. Another important factor through which cadmium is distributed into our environment is the use of the metal in production of pigments (ASTDR, 1989) which can easily be in direct or indirect contact with the water we use and the food we consume.

Another factor that contribute significant amount of cadmium into our environment is through the discharge of industrial waste (ASTDR, 1989). This can eventually get in contact with the source of our drinking water which is one of the ways through which exposure to such metal could result. In another way, the frequent use of detergent which contain cadmium as impurities could also serve as another factor that contribute to the presence of the metal in the analyzed herbal formulations.On the comparative basis, it

has also been noticed that there is no significant variation in the mean levels of cadmium across the different traditional herbal formulations (Table 5).

Table 3 illustrated a comparison between the level of Lead and Cadmium determined in the different formulations collected from the various sampling points. As it can be seen in figure 3, the typhoid formulation obtained from Kofar Doka area contain neither lead nor cadmium. But the formulation for the treatment of stomach ache was found to contain only cadmium. In the case of the sample for the treatment of yellow fever collected from Kofar Doka the level of lead was found to be quite higher than that of cadmium. Similarly, the level of lead was also found to be higher when compared to that of cadmium in the concoction for the treatment of pile from the same area. While the sample for the treatment of waist pain was found to contain only lead.

Figure 4 gives a summary bar graph comparing the levels of lead and cadmium detected at Danmagaji area. It indicated that lead was present in all the samples except that of typhoid formulations, whereas cadmium was found to be completely absent. Also in the samples collected from Tudun Wada area the levels of lead was found to be higher in all respect than that of cadmium. Figure 6 also indicated that lead was detected in all the samples collected

from Sabon Gari area and found to be higher than cadmium which was only detected in three of the samples at lower concentration.

In the case of samples collected from Kwangila area the level of lead detected was found to be higher than that of cadmium except in the formulation for the treatment of pile where that of cadmium was a little bit higher. But in Samaru area of study, the level of lead in the herbal formulations was found to be higher as compared to that of cadmium except in the sample for the treatment of stomach ache where the reverse is the case.

From this point of view, the results showed that the herbal samples contain more lead than cadmium. Thus, the most likely sources of lead that must have made its way through the herbal formulations could be from traffic emissions and via the use of contaminated water (ATSDR, 1989).

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

Considering the results obtained from this research, it is apparent that the herbal preparations sold by hawkers in Zaria and environs contain significant amount of lead and cadmium. However, the concentration of lead exceeds that of cadmium in almost all of the analyzed samples. The lead content exceeds the WHO and USEPA permissible maximum guideline limits of 0.01 and 0.015mg/l in water respectively. Similarly, the mean levels of cadmium detected exceeded the WHO guideline limit set at 0.003mg/l and that of USEPA 0.005mg/l. The high levels of these metals might be due to their high concentration in the soil as well as the stream that is flowing close to the area which may carry industrial, petrochemical and agroallied chemical wastes. Also the prevalence of mechanic garages and motor parks together with heavy traffic congestion around the town may serve as the other contributory factors that pollute the environment with this toxic metal.

It is well known fact that both lead and cadmium are highly toxic metals that can affect almost all organ systems of the human body.

The most affected organs include the liver, kidney, placenta, lungs and the central nervous system (ATSDR, 1989). The toxic effects include hypertension, fatigue, hemolytic anaemia, cancer, abdominal pain, impotence, peripheral neuropathy, etc (ATSDR, 1999). Thus, excessive or continuous consumption of these herbal formulations may cause the accumulation of these metals in the body systems which may lead to one or more of the above mentioned disorders.

6.2 Recommendations

Based on the findings of this research, the following recommendations were made:

1. Industries should be taught and be well informed about the importance of proper waste disposal and should be monitored and prosecuted in the event of non-compliance
2. The consumers of these herbal preparations in the study area should be screened periodically for heavy metals which may be accomplished by analyzing the urine, blood, hair and fingernails.
3. An agency which will serve with the responsibility of both monitoring the quality and usage of the traditional medicine should be inaugurated.

4. There is need for the government to equip our research and academic institutions with the latest equipment and competent personnel for effective environmental monitoring and analysis.
5. The frequent use of lead and cadmium in batteries, paints, water pipes, pigments, fertilizer and in other household products should be discouraged and minimized as much as possible.

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