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# Determination of the Levels of Some Heavy Metals in Cocoa Beans from Selected Cocoa- Growing Areas in Western and Ashanti Regions of Ghana

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## Abstract

In this study, six heavy metals in cocoa beans from some cocoa-growing towns in the Western and Ashanti regions were determined after acid digestion using analytical grades of concentrated nitric acid and concentrated hydrochloric acid. Cadmium, lead, copper, Manganese, Iron and zinc in the samples were determined by atomic absorption spectroscopy. The metal levels in cocoa beans from the Western region, expressed in mg/ kg varied from 0.045 to 0.066 with mean value of 0.054 for cadmium, from 0.013 to 0.03 with mean value of 0.02 for lead, from 46.47 to 55.17 with mean value of 51.98 for copper, from 48.36 to 64.65 with mean value of 55.18 for manganese, from 43.80 to 53.11 with the mean value of 47.51 iron, from 43.04 to 52.06 with the mean value of 48.29 for zinc. That of Ashanti region ranged from 0.05 to 0.065 with the mean value of 0.056 for cadmium, from 0.014 to 0.02 with the mean value of 0.017 for lead, from 47.43 to 54.17nwith the mean values of 49.10 for copper, from 47.15 to 57.34 with the mean value of 54.62 for manganese, from 50.23 to 63.87 with the mean value of 54.63 for iron and from 53.02 to 58.71 with the mean value of 56.49 for zinc. For overall conclusion, heavy metals were present in all samples but the levels of zinc, copper, iron and manganese which are considered as essential elements were high as compared to the toxic cadmium and lead. Levels according to codex set up by FAO/WHO therefore makes cocoa beans from the Western and Ashanti regions of Ghana analyzed in this study safe for consumption.

## Background to the Study

With the development of industries and modernization of agriculture, soil pollution has become more increasingly serious. The heavy metal concentrations are so high in soils of many areas that they can poison the soil-plant system, degenerate the soil, and reduce the quality of products of crops. Moreover, they can threaten the health of animals and human beings upon bioaccumulation in food chain(Alloway, 1992. Beevers, Schrader, Flesher &Hgernan, 1965). Based on this, the studies of the elemental composition of food crops is increasingly becoming much relevant not only to nutritionists and toxicologists but to the general public as well. Though some micro elements are very essential for the proper functioning of the body, the toxicity of others makes their presence in food a cause for concern. The sources of these metals in food may vary widely ranging from the soil on which the plants are grown to the conditions they are subjected to during and after crop production. Conditions such as mining and smelting of metal ores, Industrial emissions and application of insecticides and fertilizers (Anthropogenic activities) have all contributed to the elevated levels of heavy metals in the environment (Alloway, 1994).

The threat that heavy metals pose to human and animal health is exacerbated by their long-term persistence in the environment. To substantiate this, toxicological effects of heavy metals like lead on human include inhibition of hemoglobin formation, sterility, hypertension and mental retardation in children (Hermandez, Probst&Wrich, **1995**), while the major hazard to human health of cadmium is its chronicaccumulation in the kidney where it causes dysfunction if the concentration in the kidney cortex exceeds 200mg/kg fresh weight (GU, Lin, HUR,Zhuge&Zhou, 2005). In addition, though copper is an essential element, it may be toxic to both human and animals when its concentration exceeds the safe limits and its concentration in some human tissues such as thyroid can be changed dependent on the tissue state providing even cancerous or non-cancerous effects(Bakirdere&Yaman, 2008; Yaman, Akdeniz, Bkirdere&Atici, 2005). The interaction between these metals and solid phases of soil, soil water and air within and above soil depends on a variety of chemical factors and determination of heavy metal transport and fate. Absorption of metals from soil water to soil particles is most important chemical determinant that limits mobility in soil (Curtis &Smith 2002). Heavy metals from soil enter plants primarily through the root system. In general, plant roots are the most important site for uptake of chemicals from soil (Bell,1992).

It is worth- mentioning that though cocoa beans from Ghana has been reported to be of relatively low levels of heavy metals within the acceptable limits, most of these research works dates back to the 1990s (Dahiya, Karpe, Hedge, Sharma, 2005). Owing to this, the safe levels of heavy metals in the Ghanaian cocoa beans is questionable as there has been an alarming increase in mineral mining activities as well as anthropogenic activities.The patronage in cocoa products is likely to be on the ascendency following the recent discovery of high levels of polyphenols in cocoa and their great benefits to health (Engler, Engler, Chen, 2004; Cooper, Donovan, Waterhouse, Williamson, 2008). The polyphenols (catechins and epicatechins) have been detected to have good antioxidant and free radical scavenging properties leading to lower risk of cardiovascular diseases. Recent research has confirmed that cocoa has higher levels of essential polyphenols than in red wine and tea (Buijsse, Feskens, Kok, &Kromhout, 2006).The consumption of cocoa products especially in Ghana is not only because of the above-mentioned benefit but both invitro and epidemiological studies on health benefits of cocoa consumption produced significant positive results on cardiovascular health (Buijsse, et. al; 2006). This high consumption level of cocoa products in recent times calls for an analytical interventions to determine the heavy metal levels.combustion and other industrial activities contribute the most importance source of heavy metal exposure.

## Cocoa Production In Ghana

In 1879, cocoa was introduced in Ghana from Fernando Po and it is attributed to a Ghanaian farmer called Tetteh Quashie (Coe & Coe,1996). Cocoa has since then become a major bane to the Ghanaian economy. Ghana in the 1990s was exporting about 40,000 tons of cocoa per annum which made Ghana the largest producer of cocoa in the world. Ghana in the mid-1960s became the leading producer and exporter of cocoa with 570,000 tons per annum (Gordon, 1976). This is because cocoa production is characterized by it relative advantage of quick maturity and less labour intensive over other Cush crops like oil palm, coconut, cola and sheer nut at that time. However, due to factors like poor administration and effect of pest, diseases and contamination, cocoa production in 1964/65 suffered a decline from over 550,000 tons to less than 160,000 tons in 1983/84 in a period of a decade (Raffaclli, 1995). In 1990s cocoa production in Ghana rejuvenated both in quantity and quality wise as a result of effective fermentation and drying through the initiative by the Ghana cocoa Board. As a result of this initiative, there was in improvement in the taste of cocoa from Ghana and to add to this, it was associated with low levels of contamination (from heavy metals, pesticides etc.) as compared to beans from other cocoa producing countries (Leiter& Harding, 2004).

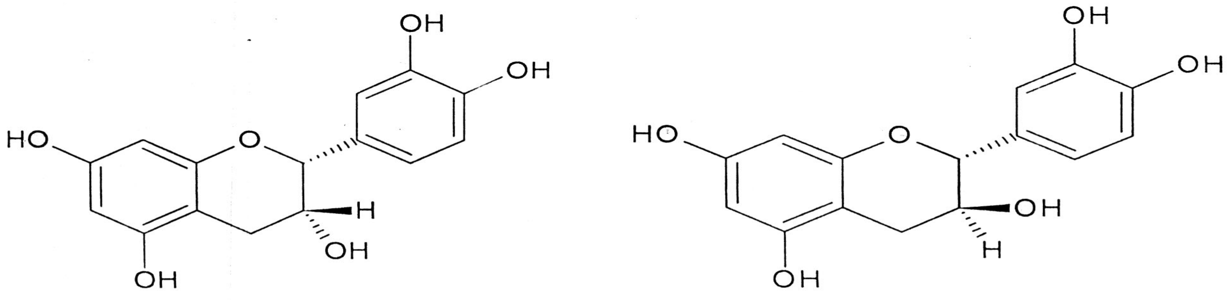
The intermittent reduction in cocoa production over the years was as a result of the fact that Ghanaian cocoa farmers reverted to traditional methods of controlling pest because they could not afford pesticides. Recently, the greatest challenge confronting cocoa production in Ghana is the ability to increase cocoa yield through the use of these modern and sophisticated methods of averting pest without compromising the quality of cocoa beans for human consumption. From the time of ripening of the cocoa pods to the marketing and processing of the beans, many processes are adopted by the Ghanaian cocoa farmer in order to ensure the quality of beans produced. This has culminated into the current production of 1,000,000 tons annually.

## Some of the benefits of drying of the cocoa beans are:

It removes both water and excess acidity from the beans; it also ensures longer storage of the cocoa beans. The commonest method of drying in Ghana is the use of the sun (sun drying) (Newman, 2000m). During drying, turning is done from time to time to ensure a uniform mixing of the beans. There is a possibility of aerial deposition of some heavy metals onto the shells of the beans. Through the adoption and implementation of quality control practices which are in concord with the international standard by Cocoa Board, Ghana consistently produces cocoa beans which are of high quality for several years. One of the cardinal interventions is the “mass spraying” exercise which is under taken four times each year in cocoa regions of the country. This ensures the prevention of pest and diseases but it also helps to curtail the indiscriminate use of unauthorized pesticides. All these have culminated into the achievement of one million metric tons of cocoa currently.

## Health Effects Of Cocoa Polyhenols

The discovery of polyphenols in cocoa and its products in the early 1909 has given the grounds for a lot of interest in the scientific world (Freudenbergg, Cox & Braun, 1932; Adam, Hardy, Nierenstein, 1931). These compounds are not only found in cocoa but also in fruits, red wine and tea(Kim & Keeney, 1984). However, apart from the discovery of these compounds in foods, the health effects of these compounds are of great importance to the chemists, food scientists and nutritionists. Polyphenols have in recent times become the centre of research for many scientists due to their health benefits to man. Some of these benefits include; Anti-carcinogenic, anti-microbial and anti-inflammatory effects. The type of polyphenol present in cocoa products has been found to be flavanols (flavan -3-ols). They form part of flavonoid which is also a sub- class of polyphenols. The flavanols in cocoa may be monomeric, dimeric or polymeric. The two main monomeric flavanols present in cocoa are: (-) – epicatechins and (+)-catechins. Their structures are presented below;



(-) - epicatechin

(+) - catechin

**Figure 2.1 Chemical structure of monomeric flavanols in Cocoa**

The two main types of dimericflavamolwhich occur in cocoa are: Proanthocyanidin $B_2$ and B5 Since the dimericflavanols are also called proanthocyanidins. The polymeric combinations of these monomers called prcoyanidins of chains up to 10 units have been found in cocoa(Cook &Samman, 1996). The major components of the polyphenols in cocoa beans are the epicatechins. These polyphenols content in the products of cocoa depend on several factors and conditions. These determinant factors range from biological to processing conditions. To develop a desired flavor in cocoa beans, optimum fermentation and drying of the cocoa beans are crucial. Depending on the atmospheric conditions, fermentation takes between 5 to 6 days. Conversely, fermentation in spite of its importance in the production of quality cocoa beans, it has been found to decrease the flavonoid content of cocoa beans [50]. Other processes like drying and roasting also contribute to the decrease in the flavonoid content in cocoa beans. However, as shown by several studies, the intake of cocoa may have cardiovascular benefits through anti-platelet function, higher density of lipoprotein, cholesterol oxidation and lowering of blood pressure(Keen, 2001; Ren, Wang, Zhu, Zhang, 2003). It has recently been published in a publication on polyphenol content in cocoa bean from different countries that cocoa beans from Ghana are of very high antioxidant properties and so rated to be of high significance to health (Othman, Ismail, Abdul Ghani,&Adenan, 2007).

## Mechanisms Of Heavy Metal Uptake By Plant

Contaminant uptake by plants and its mechanisms have been being explored by several researchers. It could be used to optimize the factors to improve the performance of plant uptake. According to Sinha et al.(Sinha, Herat, Tandon, 2004), the plants act both as “accumulators” and “excluders”. Accumulators survive despite concentrating contaminants in their aerial tissues. The contaminants biodegrade or bio transform into inert forms in their tissues. The excluders restrict contaminant uptake into their biomass. Plants have evolved highly specific and very efficient mechanisms to obtain essential micronutrients from the environment, even when present at low ppm levels. Plant roots, aided by plant-produced chelating agents and plant-induced pH changes and redox reactions, are able to solubilize and take up micronutrients from very low levels in the soil, even from nearly insoluble precipitates.

Plants have also evolved highly specific mechanisms to translocate and store micronutrients. These same mechanisms are also involved in the uptake, translocation, and storage of toxic elements, whose chemical properties simulate those of essential elements. Thus, micronutrient uptake mechanisms are of great interest to phytoremediation (US. Department,2004). The range of known transport mechanisms or specialized proteins embedded in the plant cell plasma membrane involved in ion uptake and translocation include (Alloway,1992) proton pumps (″-ATPase’s that consume energy and generate electrochemical gradients), (Beevers, et; al. ,1965) co- and anti-transporters (proteins that use the electrochemical gradients generated by ″-ATPase to drive the active uptake of ions), and (Alloway,1994) channels (proteins that facilitate the transport of ions into the cell). Each transport mechanism is likely to take up a range of ions. A basic problem is the interaction of ionic species during uptake of various heavy metal contaminants. After uptake by roots, translocation into shoots is desirable because the harvest of root biomass is generally not feasible. Little is known regarding the forms in which metal ions are transported from the roots to the shoots (U.S. Department of energy, 2004).

Plant uptake-translocation mechanisms are likely to be closely regulated. Plants generally do not accumulate trace elements beyond near-term metabolic needs. And these requirements are small ranging from 10 to 15 ppm of most trace elements suffice for most needs(U.S. Department of energy, 2004). The exceptions are “hyper accumulator” plants, which can take up toxic metal ions at levels in the thousands of ppm. Another issue is the form in which toxic metal ions are stored in plants, particularly in hyper accumulating plants, and how these plants avoid metal toxicity. Multiple mechanisms are involved. Storage in the vacuole appears to be a major one .Water, evaporating from plant leaves, serves as a pump to absorb nutrients and other soil substances into plant roots. This process, termed evapotranspiration, is responsible for moving contamination into the plant shoots as well. Since contamination is from roots to the shoots, which are harvested, contamination is removed while leaving the original soil undisturbed. Some plants that are used in phytoextraction strategies are termed "hyper accumulators." They are plants that achieve a shoot-to-root metal-concentration ratio greater than one. Non-accumulating plants typically have a shoot-to-root ratio considerably less than one. Ideally, hyper accumulators should thrive in toxic environments, require little maintenance and produce high biomass, although few plants perfectly fulfill these requirements (Salido, Hasty, Lim, Butcher, 2003 ).

Metal accumulating plant species can concentrate heavy metals like Cd, Zn, Co, Mn, Ni, and Pb up to 100 or 1000 times those taken up by non-accumulator (excluder) plants. In most cases, microorganisms, bacteria and fungi, living in the rhizosphere closely associated with plants, may contribute to mobilize metal ions, increasing the bioavailable fraction. Their role in eliminating organic contaminants is even more significant than that in case of inorganic compounds (Eradei, Mezosi, Vass, Foglein, &Bulik, 2005). Heavy metal uptake by plant through phytoremediation technologies is using these mechanisms of phytoextraction, phytostabilisation, rhizofiltration, and phytovolatilization as shown in figure 2.2

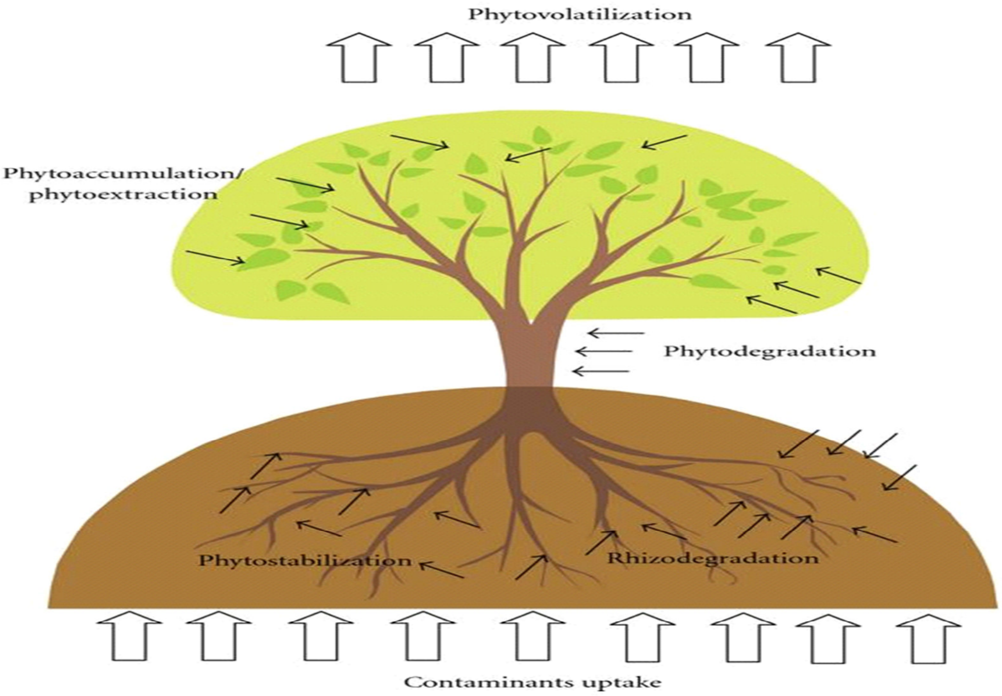


**Figure 2:2 Mechanisms of heavy metals uptake by plant through phytoremediation technology.**

Phytoextraction is the uptake/absorption and translocation of contaminants by plant roots into the above ground portions of the plants (shoots) that can be harvested and burned gaining energy and recycling the metal from the ash (Hinchman, Negri, Gatliff, 1995). Phytostabilisation is the use of certain plant species to immobilize the contaminants in the soil and groundwater through absorption and accumulation in plant tissues, adsorption onto roots, or precipitation within the root zone preventing their migration in soil, as well as their movement by erosion and deflation (Hinchman, et; al. 1995). Rhizofiltration is the adsorption or precipitation onto plant roots or absorption into and sequesterization in the roots of contaminants that are in solution surrounding the root zone by constructed wetland for cleaning up communal wastewater. Phytovolatilization is the uptake and transpiration of a contaminant by a plant, with release of the contaminant or a modified form of the contaminant to the atmosphere from the plant. Phytovolatilization occurs as growing trees and other plants take up water along with the contaminants. Some of these contaminants can pass through the plants to the leaves and volatilize into the atmosphere at comparatively low concentrations (Hinchman, et; al.). Plants also perform an important secondary role in physically stabilizing the soil with their root system, preventing erosion, protecting the soil surface, and reducing the impact of rain. At the same time, plant roots release nutrients that sustain a rich microbial community in the rhizosphere. Bacterial community composition in the rhizosphere is affected by complex interactions between soil type, plant species, and root zone location. Microbial populations are generally higher in the rhizosphere than in the root-free soil. This is due to a symbiotic relationship between soil microorganisms and plants. This symbiotic relationship can enhance some bioremediation processes. Plant roots also may provide surfaces for sorption or precipitation of metal contaminants (Sas-Nowosietska, Galimska-Stypa, Kucharski, Zielonka, Malkowski& Gray, 2008).

In phytoremediation, the root zone is of special interest. The contaminants can be absorbed by the root to be subsequently stored or metabolized by the plant. Degradation of contaminants in the soil by plant enzymes exuded from the roots is another phytoremediation mechanism(Hinchman, Negri, Gatliff, 1995). For many contaminants, passive uptake via micropores in the root cell walls may be a major route into the root, where degradation can take place (Hinchman, Negri, Gatliff).

## Sources Of Heavy Metals In Contaminated Soils

Heavy metals occur naturally in the soil environment from the pedogenetic processes of weathering of parent materials at levels that are regarded as trace (<1000 mg kg−1) and rarely toxic(National Ground Water Association Copyright 2001; WHO, 2000). Due to the disturbance and acceleration of nature’s slowly occurring geochemical cycle of metals by man, most soils of rural and urban environments may accumulate one or more of the heavy metals above defined background values high enough to cause risks to human health, plants, animals, ecosystems, or other media (Chatia, Kojima, Satokawa, 2009). The heavy metals essentially become contaminants in the soil environments because (i) their rates of generation via man-made cycles are more rapid relative to natural ones, (ii) they become transferred from mines to random environmental locations where higher potentials of direct exposure occur, (iii) the concentrations of the metals in discarded products are relatively high compared to those in the receiving environment, and (iv) the chemical form (species) in which a metal is found in the receiving environmental system may render it more bioavailable (Chatia, Kojima &Satokawa, 2009). A simple mass balance of the heavy metals in the soil can be expressed as follows (Andrianisa, Ito, Sasaki Aizawa&Unita, 2008)

$$M\mathrm{total} = (Mp + Ma + Mf + M\mathrm{ag} + M\mathrm{ow} + M\mathrm{ip}) - (M\mathrm{cr} + Ml)$$

where “ $M$ ” is the heavy metal, “ $p$ ” is the parent material, “ $a$ ” is the atmospheric deposition, “ $f$ ” is the fertilizer sources, “ a g ” are the agrochemical sources, “ o w ” are the organic waste sources, “ i p ” are other inorganic pollutants, “ c r ” is crop removal, and “ $l$ ” is the losses by leaching, volatilization, and so forth. It is projected that the anthropogenic emission into the atmosphere, for several heavy metals, is one-to-three orders of magnitude higher than natural fluxes (Ampiah-Bonney, Tyson &Lauza, 2007). Heavy metals in the soil from anthropogenic sources tend to be more mobile, hence bioavailable than pedogenic, or lithogenic ones (Yuof&Malek, 2009; WHO, 2001). Metal-bearing solids at contaminated sites can originate from a wide variety of anthropogenic sources in the form of metal mine tailings, disposal of high metal wastes in improperly protected landfills, leaded gasoline and lead-based paints, land application of fertilizer, animal manures, biosolids (sewage sludge), compost, pesticides, coal combustion residues, petrochemicals, and atmospheric deposition (Gaur&Adholeya, 2004; Rakhshaee, Giahi&Pourahmad, 2009) are discussed hereunder.

### Fertilizers

Historically, agriculture was the first major human influence on the soil (European Commission DG ENV. E3. ,2002). To grow and complete the lifecycle, plants must acquire not only macronutrients (N, P, K, S, Ca, and Mg), but also essential micronutrients. Some soils are deficient in the heavy metals (such as Co, Cu, Fe, Mn, Mo, Ni, and Zn) that are essential for healthy plant growth (Musselman QEP,http://www.cet **),** and crops may be supplied with these as an addition to the soil or as a foliar spray. Cereal crops grown on Cu-deficient soils are occasionally treated with Cu as an addition to the soil, and Mn may similarly be supplied to cereal and root crops. Large quantities of fertilizers are regularly added to soils in intensive farming systems to provide adequate N, P, and K for crop growth. The compounds used to supply these elements contain trace amounts of heavy metals (e.g., Cd and Pb) as impurities, which, after continued fertilizer, application may significantly increase their content in the soil (Cheny, You, Yu &Chen, 2009). Metals, such as Cd and Pb, have no known physiological activity. Application of certain phosphatic fertilizers inadvertently adds Cd and other potentially toxic elements to the soil, including F, Hg, and Pb (Rodrignez, Lopez-Bellido, Carnicer, Recreo, Tallos, Monteagudo, 2005).

### Pesticides

Several common pesticides used fairly extensively in agriculture and horticulture in the past contained substantial concentrations of metals. For instance in the recent past, about 10% of the chemicals have approved for use as insecticides and fungicides in UK were based on compounds which contain Cu, Hg, Mn, Pb, or Zn. Examples of such pesticides are copper-containing fungicidal sprays such as Bordeaux mixture (copper sulphate) and copper oxychloride (Roy, Labelle & Mehta, 2005). Lead arsenate was used in fruit orchards for many years to control some parasitic insects. Arsenic-containing compounds were also used extensively to control cattle ticks and to control pests in banana in New Zealand and Australia, timbers have been preserved with formulations of Cu, Cr, and As (CCA), and there are now many derelict sites where soil concentrations of these elements greatly exceed background concentrations. Such contamination has the potential to cause problems, particularly if sites are redeveloped for other agricultural or nonagricultural purposes. Compared with fertilizers, the use of such materials has been more localized, being restricted to particular sites or crops (Wagner-Dobler**,**http://www.gbf.de/mercury_remediation1/pdf- **).**

## Bio solids and Manures

The application of numerous biosolids (e.g., livestock manures, composts, and municipal sewage sludge) to land inadvertently leads to the accumulation of heavy metals such as As, Cd, Cr, Cu, Pb, Hg, Ni, Se, Mo, Zn, Tl, Sb, and so forth, in the soil (Traunfeld& Clement, 2001). Certain animal wastes such as poultry, cattle, and pig manures produced in agriculture are commonly applied to crops and pastures either as solids or slurries (Wagner-Dobler, http://www.gbf.de/mercury_remediation1/pdf- **).** Although most manure are seen as valuable fertilizers, in the pig and poultry industry, the Cu and Zn added to diets as growth promoters and As contained in poultry health products may also have the potential to cause metal contamination of the soil (Wagner-Dobler, Resaee, Derayat, Mortazari, Yamini,http://www.gbf.de/mercury_remediation1/pdf- **).** The manures produced from animals on such diets contain high concentrations of As, Cu, and Zn and, if repeatedly applied to restricted areas of land, can cause considerable buildup of these metals in the soil in the long run.

Biosolids (sewage sludge) are primarily organic solid products, produced by wastewater treatment processes that can be beneficially recycled (Sas- Nowosietska, Galimska- StypaKucharski, Zielonkaa&Malkowskia, 2008).Land application of biosolids materials is a common practice in many countries that allow the reuse of biosolids produced by urban populations (Erakhrumen&Agbontalor, 2007). The term sewage sludge is used in many references because of its wide recognition and its regulatory definition. However, the term biosolids is becoming more common as a replacement for sewage sludge because it is thought to reflect more accurately the beneficial characteristics inherent to sewage sludge (US. Environmental Protection Agency, 2000). It is estimated that in the United States, more than half of approximately 5.6 million dry tonnes of sewage sludge used or disposed of annually is land applied, and agricultural utilization of biosolids occurs in every region of the country. In the European community, over 30% of the sewage sludge is used as fertilizer in agriculture (US. Environmental Protection Agency, 2000) . In Australia over 175 000 tonnes of dry biosolids are produced each year by the major metropolitan authorities, and currently most biosolids applied to agricultural land are used in arable cropping situations where they can be incorporated into the soil (Roy, Labelle, Mehta, 2005).

There is also considerable interest in the potential for composting biosolids with other organic materials such as sawdust, straw, or garden waste. If this trend continues, there will be implications for metal contamination of soils. The potential of biosolids for contaminating soils with heavy metals has caused great concern about their application in agricultural practices (Moreno, Anderson, Stewart & Robinson, 2008). Heavy metals most commonly found in biosolids are Pb, Ni, Cd, Cr, Cu, and Zn, and the metal concentrations are governed by the nature and the intensity of the industrial activity, as well as the type of process employed during the biosolids treatment (Prasad, De Oliverira, Freitas, 2003). Under certain conditions, metals added to soils in applications of biosolids can be leached downwards through the soil profile and can have the potential to contaminate groundwater (Liu, Jiang, Liu, Xin, Hou, 2000). Recent studies on some New Zealand soils treated with biosolids have shown increased concentrations of Cd, Ni, and Zn in drainage leachates (Bhattacharya, Banerjee, &Gopal, 2006).

## Wastewater

The application of municipal and industrial wastewater and related effluents to land dates back 400 years and now is a common practice in many parts of the world (Van Ginneken&Guisson et.al; 2007). Worldwide, it is estimated that 20 million hectares of arable land are irrigated with waste water. In several Asian and African cities, studies suggest that agriculture based on wastewater irrigation accounts for 50 percent of the vegetable supply to urban areas (Sinha, Hearat,&Taudon, 2004). Farmers generally are not bothered about environmental benefits or hazards and are primarily interested in maximizing their yields and profits. Although the metal concentrations in wastewater effluents are usually relatively low, long-term irrigation of land with such can eventually result in heavy metal accumulation in the soil.

## Metal Mining and Milling Processes and Industrial Wastes

Mining and milling of metal ores coupled with industries have bequeathed many countries, the legacy of wide distribution of metal contaminants in soil. During mining, tailings (heavier and larger particles settled at the bottom of the flotation cell during mining) are directly discharged into natural depressions, including onsite wetlands resulting in elevated concentrations (US Department of Ernergy, 2004). Extensive Pb and zinc Zn ore mining and smelting have resulted in contamination of soil that poses risk to human and ecological health. Many reclamation methods used for these sites are lengthy and expensive and may not restore soil productivity. Soil heavy metal environmental risk to humans is related to bioavailability. Assimilation pathways include the ingestion of plant material grown in (food chain), or the direct ingestion (oral bioavailability) of, contaminated soil (Salido, Hasty, Lim, Butcher, 2003 ).

Other materials are generated by a variety of industries such as textile, tanning, petrochemicals from accidental oil spills or utilization of petroleum-based products, pesticides, and pharmaceutical facilities and are highly variable in composition. Although some are disposed of on land, few have benefits to agriculture or forestry. In addition, many are potentially hazardous because of their contents of heavy metals (Cr, Pb, and Zn) or toxic organic compounds and are seldom, if ever, applied to land. Others are very low in plant nutrients or have no soil conditioning properties

### Air-Borne Sources

Airborne sources of metals include stack or duct emissions of air, gas, or vapor streams, and fugitive emissions such as dust from storage areas or waste piles. Metals from airborne sources are generally released as particulates contained in the gas stream. Some metals such as As, Cd, and Pb can also volatilize during high-temperature processing. These metals will convert to oxides and condense as fine particulates unless a reducing atmosphere is maintained (Erdei, Mezosi, Vass, Foglein&Bulik, 2005). Stack emissions can be distributed over a wide area by natural air currents until dry and/or wet precipitation mechanisms remove them from the gas stream. Fugitive emissions are often distributed over a much smaller area because emissions are made near the ground. In general, contaminant concentrations are lower in fugitive emissions compared to stack emissions. The type and concentration of metals emitted from both types of sources will depend on site-specific conditions. All solid particles in smoke from fires and in other emissions from factory chimneys are eventually deposited on land or sea; most forms of fossil fuels contain some heavy metals and this is, therefore, a form of contamination which has been continuing on a large scale since the industrial revolution began. For example, very high concentration of Cd, Pb, and Zn has been found in plants and soils adjacent to smelting works. Another major source of soil contamination is the aerial emission of Pb from the combustion of petrol containing tetraethyl lead; this contributes substantially to the content of Pb in soils in urban areas and in those adjacent to major roads. Zn and Cd may also be added to soils adjacent to roads, the sources being tyres, and lubricant oils (Erdei, Mezosi, Vass, Foglein&Bulik, 2005).

### Some Factors Influencing Plant Uptake Of Metals.

Soil represents the major repository of trace elements over geologic time. On a worldwide basis, soil shows an average composition close to the earth crust but the near –surface parent material from which soils are derived is not uniform and soil-forming processes differ markedly from one climate region to another, accounting for considerable overall variations in trace metal concentrations (Krauskopf, 1972).The major factor governing the availability of nutrients, metals and ions to plants in the soil is chiefly the uncomplexed ion (Janne&Luoma, 1977). This is due to the fact that in order for root uptake to occur, a soluble species must exist adjacent to the root membrane for some finite period. The form of this solution species will have an enormous influence on its longevity in soil solution, mobility in soils and on the rate and extent of uptake, and perhaps mobility and toxicity in the plant (Tiffin, 1977). Once deposited, metal- containing materials are subject to chemical and microbial modification with metal solubility ultimately approaching thermodynamic equilibrium with native soil minerals and organic matter. The rate and extent of solubility are governed by the physicochemical properties of the deposited material, soil processes and soil properties. Evidence indicates that soil microorganisms may play an important role by the producing soluble ligands with high affinity for metals (Willdung et. al; 1976: Wildung,& Garland Drucker, 1977). Metals entering the soil as stable organo-complexes, such as those used in fertilization to correct micronutrient deficiencies or those possibly present in discharge from a nuclear fuel separation facility, may be initially highly stable (Norvell, & Lindsay,1969; Wildung, Garland, 1974). The duration of solubility and mobility in the soil will be a function of the stability of the complex to be substituted by major competing ions such as Ca and H (Lahan& Hochberg, 1976; Norvel, 1972) and the stability of the organic ligand to microbial decomposition (Erdei, Mezosi, Vass, Foglein&Bulik, 2005). It may be concluded that the soil's physicochemical parameters which are most important in influencing the solubility of metals include: solution composition (organic and inorganic soluble), Eh and PH: type and density of charge on soil colloids; and reactive surface area (Keeney & Wilding, 1972). These phenomena will be dependent upon soil properties including metal concentration and form, particle size distribution, quantity and reactivity of hydrous oxides, mineralogy microbial activities and aeration (Janne&Luoma, 1977). These soil properties vary geographically and will be a function of the combined effects of parent material, topography, climate, time, man's activities and biological processes (Janney, 1994).

**Figure2.3 Factors which are affecting the uptake mechanisms of heavy metals.**

### Properties of Medium

Agronomical practices are developed to enhance remediation (pH adjustment, addition of chelators, fertilizers) (Prasad, De Oliveira, Freitas, 2003). For example, the amount of lead absorbed by plants is affected by the pH, organic matter, and the phosphorus content of the soil. To reduce lead uptake by plants, the pH of the soil is adjusted with lime to a level of 6.5 to 7.0 (Trunfeld& Clement, 2001).

### The Root Zone

The Root Zone is of special interest in phytoremediation. It can absorb contaminants and store or metabolize it inside the plant tissue. Degradation of contaminants in the soil by plant enzymes exuded from the roots is another phytoremediation mechanism. A morphological adaptation to drought stress is an increase in root diameter and reduced root elongation as a response to less permeability of the dried soil (Merki, Schultze-Kraft &Infante, 2005).

### Vegetative Uptake

Vegetative Uptake is affected by the environmental conditions (Burken&Sdinoor, 1996). The temperature affects growth substances and consequently, root length. Root structure under field conditions differs from that under greenhouse condition(Merki, Schultze-Kraft &Infante, 2005) . The success of phytoremediation, more specifically phytoextraction, depends on a contaminant-specific hyperaccumulator (Tu, Ma, Fayiga&Zillioux (2004). Understanding mass balance analyses and the metabolic fate of pollutants in plants are the keys to proving the applicability of phytoremediation (Mwegoha, 2008). Metal uptake by plants depends on the bioavailability of the metal in the water phase, which in turn depends on the retention time of the metal, as well as the interaction with other elements and substances in the water. Furthermore, when metals have been bound to the soil, the pH, redox potential, and organic matter content will all affect the tendency of the metal to exist in ionic and plant-available form. Plants will affect the soil through their ability to lower the pH and oxygenate the sediment, which affects the availability of the metals (Fritioff&Greger, 2003), increasing the bioavailability of heavy metals by the addition of biodegradable physicochemical factors, such as chelating agents and micronutrients (Van Ginneken&Guisson et.al; 2007)

### Addition of Chelating Agent

The increase of the uptake of heavy metals by the energy crops can be influenced by increasing the bioavailability of heavy metals through addition of biodegradable physicochemical factors such as chelating agents, and micronutrients, and also by stimulating the heavy-metal-uptake capacity of the microbial community in and around the plant. This faster uptake of heavy metals will result in shorter and, therefore, less expensive remediation periods. However, with the use of synthetic chelating agents, the risk of increased leaching must be taken into account (Van Ginneken&Guisson et.al; 2007) The use of chelating agents in heavy-metal-contaminated soils could promote leaching of the contaminants into the soil. Since the bioavailability of heavy metals in soils decreases above pH 5.5–6, the use of a chelating agent is warranted, and may be required, in alkaline soils. It was found that exposing plants to EDTA for a longer period (2 weeks) could improve metal translocation in plant tissue as well as the overall phytoextraction performance. The application of a synthetic chelating agent (EDTA) at 5 mmol/kg yielded positive results (Roy, Labelle, Mehta, 2005). Plant roots exude organic acids such as citrate and oxalate, which affect the bioavailability of metals. In chelate-assisted phytoremediation, synthetic chelating agents such as NTA and EDTA are added to enhance the phytoextraction of soil-polluting heavy metals. The presence of a ligand affects the biouptake of heavy metals through the formation of metal-ligand complexes and changes the potential to leach metals below the root zone (Seuntjens, Nowack&Schulin, 2004).

### Methods ForAnalysing Heavy Metals.

The determination of heavy in organic matrices requires analytical techniques with high sensitivity and low detection limits. These methods with the above-mentioned specifications include inductively coupled plasma mass spectrometry (ICP –MS), inductively coupled plasma-optical emission spectrometry (ICP-OES), Electro-thermal atomic absorption spectrometry (ETAAS) which is the most widely used, flame atomic absorption spectrometry (F-AAS), graphite furnace spectrometry (GF-AAS) and A-ray fluorescence, neutron activation analysis (NAA).

### Principles Of Atomic Absorption Spectroscopy

The technique makes use of absorption spectrometry to assess the concentration of an analyte in a sample. It requires standards with known analyte content to establish the relation between the measured absorbance and the analyte concentration and relies therefore on the Beer-Lambert Law. In short, the electrons of the atoms in the atomizer can be promoted to higher orbitals (excited state) for a short period of time (nanoseconds) by absorbing a defined quantity of energy (radiation of a given wavelength). This amount of energy, that is wavelength, is specific to a particular electron transition in a particular element. In general, each wave length corresponds to only one element, and the width of an absorption line is only of the order of a few Pico meters (pm), which gives the technique its elemental selectivity.

The radiation flux without a sample and with a sample in the atomizer is measured using a detector, and the ratio between the two values (the absorbance) is converted to analyte concentration or mass using the Bee – Lambert Law.

The instrumentation is designed in such a way that, in order to analyze a sample for its atomic content, it is to be atomized. The atomizers most commonly used nowadays are flames and electro thermal (graphite tube) atomizers. The atoms should then be irradiated by optical radiation and the radiation source could be an element-specific line radiation source or a continuum radiation source. The radiation then passes through a monochromator in order to separate the element-specific radiation from any other radiation emitted by the radiation source, which is finally measured by a detector. Figure 2.2 shows a block diagram of atomic absorption spectrometer.

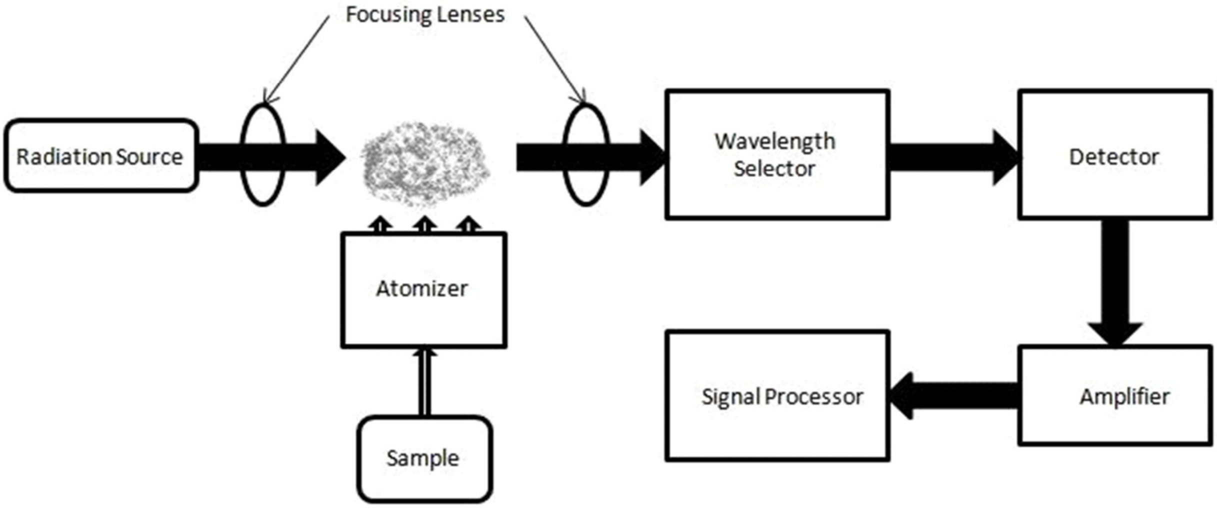


**Figure 2.4 Schematic diagram of the operation of atomic absorption spectrometer**

### Toxicity Of The Analysed Heavy Metal

A heavy metal is a member of a loosely- defined subset of elements that exhibit metallic properties. It mainly includes the transition metals, some metalloids, lanthanides, and actinides. Many different definitions have been proposed –some based on density, some on atomic number or atomic weight and some on chemical properties or toxicity (McCarthy, 2012). Heavy metals constitute a very heterogeneous group of elements widely varied in their chemical properties and biological functions. The term" heavy metals "defined as commonly held for those metals, which have specific weights more than 5gcm$^{-3}$ (John, 2002). Heavy metals are kept under environmental pollutant category due to their toxic effects in plants, human and food. The toxicity of these metals has two main aspects: (a) the fact that they have no known metabolic functions, but when present in the body they disrupt normal cellular processes, leading to toxicity in a number of organs; (b) the potential , particularly, of cadmium and lead to accumulate in the biological tissues, a process known as bioaccumulation. This occurs because the metals, once taken up into the body, are stored in particular organs like kidney and liver which are excreted at a slow rate compared with its intake. The toxicities of the individual heavy metals are discussed below:

### Lead (Pb)

The toxic effects of lead, like those of mercury, have been principally established in studies on people exposed to lead in the course of their work. Short-term exposure of high levels of lead may affect brain function by interfering with neurotransmitter release and synapse formation (Dahiya, Karpe, Hedge & Sharma, 2005**).** Exposure to lead has been associated with reduced IQ, Learning disabilities, slow growth, hyperactive, antisocial behavior and impaired hearing. Lead is known to damage the kidney, liver and reproductive system, basic cellular processes and brain function (Mo, Choi & Robinson, 1988). In addition to this, the most critical effect of low-level lead exposure is an intellectual development of young children and, like mercury, lead crosses the placental barrier and accumulates in the foetus.

### Cadmium (Cd)

Cadmium can mainly be found in the earth's crust. It always occurs in combination with Zinc (Zn). Cadmium also consists in the industries as an inevitable by –product of Zn, lead and copper extraction. After being applied, it enters the environment mainly through the ground, because it is found in manures and pesticides. The principal toxic effect of cadmium is its toxicity to the kidney, although it has also been associated with lung damage (including induction of lung tumor) and skeletal changes in occupationally exposed populations. Cadmium is relatively poorly absorbed into the body, but once absorbed, it is slowly excreted. Like other metals, it accumulates in kidney causing renal damage. An exposure to significantly higher cadmium levels occurs when people smoke. Tobacco smoke is first transported to the liver through the blood. There, it is bond to protein to form complexes that are transported to the kidney. This accumulation damages filtering mechanism causing difficulty in the excretion of essential proteins and sugars form the body.

### Copper (Cu)

Research shows that short period of exposure to high levels of copper can cause gastrointestinal disturbance, including diarrhoea, stomach cramps, nausea and vomiting. Using water with elevated levels of copper over many years may cause liver and kidney damage. The seriousness of these effects can be expected to increase with increased copper levels or length of exposure.Children under one year of age are more sensitive to copper than adults. Other persons who are highly susceptible to copper toxicity include people with liver damage or Wilson's disease (US EPA, 1984). Long-term exposure to copper can cause irritation of the nose, mouth and eyes and it causes headaches, stomach- aches, dizziness etc. High uptake of copper may lead to liver and Kidney damage and even death.

### Zinc (Zn)

Although Zinc (Zn) is an essential requirement for good health, excess zinc can be harmful. Excessive absorption of zinc suppresses copper and iron absorption (. http://www.epa.gov/nrmrl/wswrd/cr/corr-res-copper.html). The U.S. Food and Drugs Administration (FDA) have stated that zinc damages nerve receptors in the nose, which can cause anosmia. Reports of anosmia were also observed in the 1930s when Zn preparations were in a failed attempt to prevent polio infections (Fosmire, 1990). The problem with consuming too much zinc is that it actually prohibits ones metabolism from absorbing the other vitamins and minerals needed by the body. Again, Zn toxicity lowers the body's immunity and good cholesterol levels. In August 2008, the journal "Neurology" reported on four patients suffering from neuropathy and other neurological symptoms typical of zinc poisoning and copper deficiencies.

### Iron (Fe)

The University of Maryland Medical Centre reports that there may be evidence that high iron levels may increase your risk of certain cancers, including breast cancer. The " Journal of clinical Biochemistry and Nutrition" notes that breast cancer is the most common type of cancer among women around the globe, and excessive iron intake may be one reason why so many women- develop this type of cancer (Oxford &Obeng, 1985) storing excess amount of iron in the body may also increase the risk of heart disease. The office of Dietary Supplements of the National Institute of Health notes that too much iron accumulation in the body may encourage which can then lead to a heart disease. (Yun-Jung et, al; 2009)

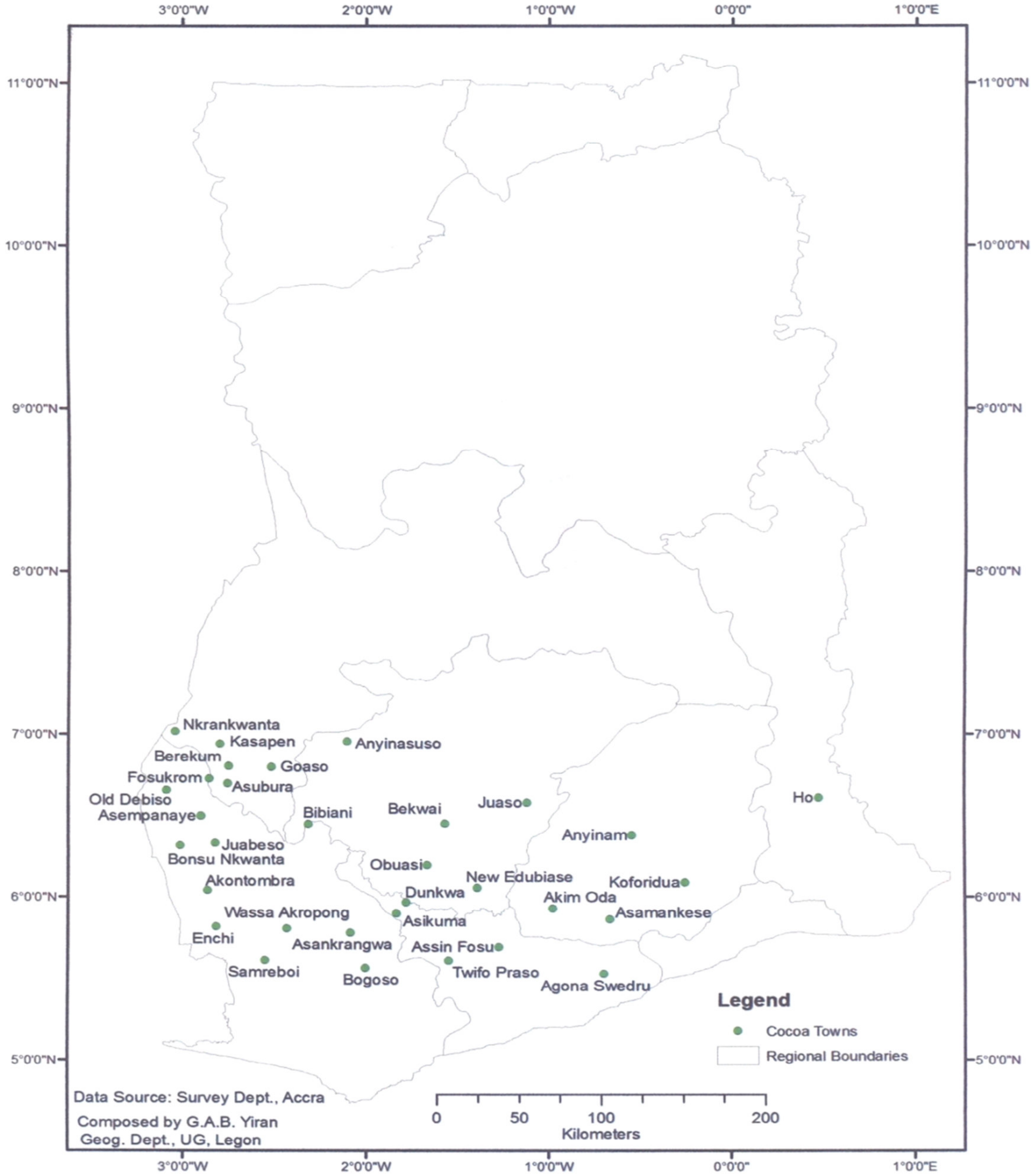
### Manganese [Mn]

Manganese is a common element in the earth's crust, water and particulate matter in the atmosphere. This element is used in manufacturing steel, dry cell batteries, electrical coils, ceramics, fertilizers and fungicides. However, toxic effects from oral ingestion of manganese are rare and include lethargy, changes in muscle tone and posture, coma and involuntary movements.

## Geographical Description Of The Samples Area

Ghana is situated on the west coast of Africa about 750 north of the equator between latitude 4 and 11.5 N and 3.111 west. It shares boundaries with Burkina Faso to the North, Togo to the east, La cote d'viore to the west and

Gulf of Guinea (part of Atlantic Ocean) to the south. Generally, the Climate of Ghana is tropical and two main types of vegetation exist. These are the rain forest and savanna grassland. The forest vegetation is characterized by high temperatures and heavy rainfall almost throughout the year and is usually divided into rain forest and semi-deciduous forest. The forest vegetation promotes very rapid plant growth (Dickson &Benneh, 1998). Cocoa thrives well in the forest regions of Ghana which covers the south western part and compresses 6 out of the 10 political regions of the country. Samples of cocoa beans from Western region and Ashanti region were taken during this work. Figure 3.1 shows a map of major cocoa-growing towns from some of which cocoa beans used for the analysis in this work were taken.



Data Source: Survey Dept., Accra
Composed by G.A.B. Yiran
Geog. Dept., UG, Legon

**Figure 3.2 Map Showing major Cocoa growing areas in Ghana**

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**Figure 3.1 a map showing the major cocoa-growing towns from some of which samples were taken.**

## Collection Of Samples

Samples of fermented and dried cocoa beans were obtained from cocoa farmers in some selected cocoa growing towns in Ashanti region and Western region. However, few cocoa fruits were obtained from the farms of some farmers before the beans were dried.

Sampling was done such that the towns from which cocoa beans were obtained were well spread throughout the region with each of the towns having equal opportunity of being chosen. Western region was divided into four areas (Northern part, Eastern part, Southern part and western part). The names of the cocoa farming towns in each of the demarcated areas were written on separate sheets of papers in a bowl and one town each was picked at random after thoroughly mixing them until all the demarcated areas were duly represented (one town from each divided part). In each town visited, dried cocoa beans samples were obtained from five different cocoa farmers and kept in clean and dry polyethylene bags. The same procedure was employed during sampling in Ashanti region. Below shows some photographs during the sampling expedition.

**Figure 3.2 Cocoa fruit being collected from farmers at Mampong in the Ashanti region of Ghana (AM005)**

**Figure 3.3 Some of the cocoa samples from Sefwi-Asempaneye in the Western region of Ghana (SA005)**

**Figure 3.4 Some of the cocoa samples fromBogoso in the Western Region of Ghana (SB002)**

**Figure 3.5 Some of the cocoa samples from Bekwai in the Ashanti Region of Ghana (AB003)**

**Figure 3.6 some of the samples from Tepa-Anyinasosu in the Ashanti Region of Ghana (AT004)**

**Figure 3.7 Cocoa beans from Juaso in the Asanti region of Ghana(AJ001)**

**Table 3.1 shows the sampling location (towns) of the cocoa beans.**

| Region | Town | Numbers of samples |
|---|---|---|
| | 1. Kasapen (SK) | 5 |
| **Western** | 2. Enchi (SE) | 5 |
| | 3. Sefwi–Juabeso(SJ) | 5 |
| | 4. Asempaneye(SA) | 5 |
| | 5. Bogoso(SB) | 5 |
| | 1.Juaso(AJ) | 5 |
| **Ashanti** | 2.TepaAnyinasuso(AT) | 5 |
| | 3.Bekwai(AB) | 5 |
| | 4. Obuasi (AO) | 5 |
| | 5 Mampong(AM) | 5 |
| **Total** | | **50** |

**Sample Preparation And Pre-Treatment**

The dry cocoa samples obtained were kept in clean, dry glass bottles for further treatment. In handling the cocoa beans, gloves were worn to avert external contamination which will potentially discredit the work. Care was taken to also ensure that water and other external reagents did not come into contact with the beans before subjected them to milling, digestion and subsequently, analysis by the use of Flame Atomic Absorption Spectrometer. (F-AAS)

## CHEMICALS AND REAGENTS

All reagent used for this work were of analytical grade. Digestion of samples was performed using nitric of acid (65%) and HCL (30%), both obtained from Merck, Germany. Petroleum ether, HCl, Selenium catalysts were used for the proximate analysis for the determination of the fat protein and carbohydrate content in the cocoa beans.De- ionized water (distilled water) obtained from the Chemistry department, KNUST was used for all the analytical work.

### Equipment And General Apparatus

All samples were collected, sealed and stored in pre-cleaned and dry low-density polyethylene bags. Samples were homogenized using a milling machine at the pharmaceutical department at KNUST. Digested samples were stored in polyethylene vials. The vials were pre-cleaned by soaking in ultrapure nitric acid for 24 hours, rising thoroughly with de-ionized water and dried at 30 c in an oven. Every glassware used in the analysis was initially washed with detergent and water after which they were soaked in ultrapure nitric acid for 24 hours. The glassware were then rinsed several times with de-ionized water and dried at 100. Analysis of heavy metal concentration was performed using Buck Scientific model VEP 210.

### Preparation Of Standards

Standards of heavy metals were prepared from multi- element standard stock solution (obtained from Inorganic Ventures Inc., USA) in 10% nitric acid and 2% HCl. Single- element standards of most of the elements having very low concentrations were also prepared for calibration. The working standard solutions were all prepared by serial dilution of the stock solution with de-ionized water in 100ml volumetric flask.

### Digestion Of Samples

This is a critical sample preparation step in quantitative analysis and steps have to be taken to ensure the reliability and reproducibility of the results by ensuring that samples are free from contamination and to avoid or minimize loss of analyte. The aqua regia for the digestion was prepared by mixing 3:1 volumes of HCL and $HNO_3$ respectively in a hood. After storing the prepared aquaregia for 2 days to ensure a complete reaction and a uniform homogenous mixture between the acids, digestion of the samples commenced. 1g of the sample was weighed using a balance into 30ml of the aqua regia in a 50ml conical flask.

The content of the flask was heated on a hot plate at $40^{0c}$ in a hood with the flask partially covered. Digestion continued until almost all the 30ml of the aqua regia and its content was evaporated to dryness before taking it out of the hot plate. On cooling, de-ionized water was added to make it up to the 50ml mark (Lokeshwari, Chandrappa, 2006)] before being filtered into pre-cleaned polypropylene tubes for analysis. All the samples were subjected to this procedure. Blank samples were digested and analyzed in the same way as described for the cocoa samples. Triplicate digestions were conducted for each sample.

**Table 3.2 Wavelengths and detection limits analysis.**

| Element | Wavelength (nm) | Detection limit |
|---|---|---|
| Cadmium | 228.9 | 0.005 |
| Lead (Pb) | 283.3 | 0.100 |
| Iron (Fe) | 248.3 | 0.03 |
| Copper (Cu) | 324.8 | 0.02 |
| Magnesium (Mn) | 279.5 | 0.01 |
| Zinc (Zn) | 213.9 | 0.005 |

### Quality Assurance

Sample containers and glassware used in the analysis were first cleaned with metal free nonionic detergent solution, rinsed thoroughly with de-ionized water and soaked in nitric acid for 24 hours. They were then washed several times with de-ionized water prior to the use. Blanks, consisting of de-ionized water, chemicals and reagents used for the digestion were subjected to similar sample preparation and analytical procedure in an effort to reduce the effect of contamination arising from chemical reagents, distilled water and glassware used in the analysis. Accuracy of the method was evaluated through the analysis of two reference materials: NIST 1547 SRM certified Peach Leaves and IAEAV-10 SRM certified Hay Powder.

### Quality Assurance Of Analysis

In an effort to obtain results that are accurate and reproducible in this analysis , a number of quality control measures were ensured; from the initial sampling process to the final analysis of the metals using flame atomic absorption spectrometer (F-AAS**)** instrument. Sampling of Cocoa beans from Western and Ashanti region ensured that the representative samples from the major cocoa –growing towns from the major cocoa –growing towns were taken. Sampling was done according to the codex general standard for contaminants and toxins in food (Roy, Labelle, Mehta, 2005) strict precautions were taken to reduce contamination during the handling and preparation of samples. As mention earlier in this work all reagents were of analytical grade and sample containers and apparatus were washed and rinsed thoroughly prior to their use. Since reagents could be reliable sources of contamination in analytical work, high purity reagents and water were used in this work. Again, reagent blanks were analyzed. The concentrations reported in this work are thus actual concentrations of the samples relative to the reagent blanks. Moreover, the accuracy and reliability of the measurements were ascertained with the analysis of two reference materials; NIST 1574 SRM certified Peach Leaves and IAEA-V-10 SRM certified Hay Powder. These measured values for this work along with their corresponding certified or reported values for the reference materials are presented in table 4.1 and 4.2.

## 4. TABLE OF RESULTS

**Table 4.1 Levels of heavy metals in IAEA – V-10 Standard reference materials (Hay Powder)**

| Metal | This work | Reported values |
|---|---|---|
| Pb | 1.55(1.42-1.51) | 1.6(08-1.9) |
| Cd | 0.04(0.03-0.068) | 0.03(0.02-0.05) |
| Fe | 185.5(175-189) | 186(177-190) |
| Zn | 23.8(22.60-25.10) | 24(23-25) |
| Mn | 48.2(43.30-50.4) | 47(44-51) |
| Cu | 9.90 (9.2-10.3) | 9.4(8.8-9.7) |

**Table 4.2 Leaves of heavy metals in NIST standard reference material 1547 (Peach Leaves)**

| Metal | This work | Reported Value |
|---|---|---|
| **Pb** | 43.48±2.62 | 45±3 |
| **Cd** | 0.12±0.03 | 0.11±0.02 |
| **Fe** | 289±8.8 | 300±20 |
| **Zn** | 27.56±2.1 | 25±3 |
| **Mn** | 89. 76±3.1 | 91±4 |
| **Cu** | 11.74±2.0 | 12±1 |

**Table 4.3 Levels of some heavy metals (mg/kg) in cocoa beans from some cocoa- growing towns in the Western region of Ghana.**

| TOWN | SAMPLE | Cd | Pd | Cu | Mn | Fe | Zn |
|---|---|---|---|---|---|---|---|
| | **SK001** | 0.039±0.00 | 0.015±0.00 | 48.18±0.04 | 67.73±0.02 | 39.82±0.04 | 67.15±0.00 |
| **KASAPEN** | **SK002** | 0.025±0.001 | 0.011±0.001 | 56.00±0.11 | 49.15±0.07 | 48.35±0.04 | 46.65±0.04 |
| | **SK003** | 0.063±0.002 | 0.013±0.001 | 52.75±0.03 | 56.67±0.03 | 56.35±0.04 | 47.33±0.04 |
| | **SK004** | 0.055±0.000 | 0.014±0.00 | 54.17±0.02 | 41.83±0.02 | 41.78±0.04 | 52.84±0.02 |
| | **SK005** | 0.045±0.002 | 0.013±0.00 | 48.46±0.05 | 57.85±0.03 | 44.58±0.04 | 39.38±0.03 |
| | **MEAN** | **0.045±0.001** | **0.013±0.0004** | **51.19±0.05** | **64.65±0.03** | **53.11±0.04** | **50.67±0.034** |
| **ENCHI** | **SE001** | 0.045±0.003 | 0.013±0.001 | 45.91±0.06 | 45.66±0.02 | 41.81±0.06 | 41.65±0.02 |
| | **SE002** | 0.085±0.003 | 0.015±0.001 | 43.50±0.04 | 68.57±0.03 | 8.83±0.02 | 48.79±0.06 |
| | **SE003** | 0.061±0.003 | 0.014±0.002 | 43.65±0.04 | 62.84±0.03 | 55.32±0.03 | 44.83±0.01 |
| | **SE004** | 0.071±0.00 | 0.014±0.001 | 50.38±0.04 | 42.16±0.01 | 42.65±0.00 | 36.18±0.06 |
| | **SE005** | 0.062±0.002 | 0.019±0.002 | 48.92±0.05 | 61.74±0.02 | 42.80±0.05 | 48.25±0.02 |
| | **MEAN** | 0.065±0.002 | 0.015±0.003 | 46.47±0.05 | 56.19±0.02 | 44.28±0.03 | 43.94±0.03 |
| **ASEMPANEYE** | **SA001** | 0.039±0.00 | 0.017±0.002 | 45.29±0.02 | 48.94±0.02 | 43.14±0.03 | 46.80±0.05 |
| | **SA002** | 0.025±0.001 | 0.021±0.002 | 65.25±0.04 | 57.92±0.05 | 46.72±0.03 | 43.26±0.02 |
| | **SA003** | 0.063±0.002 | 0.019±0.002 | 62.28±0.00 | 42.25±0.03 | 44.16±0.02 | 39.16±0.02 |
| | **SA004** | 0.055±0.00 | 0.018±0.001 | 47.41±0.06 | 58.26±0.02 | 49.42±0.00 | 43.77±0.05 |
| | **SA005** | 0.045±0.002 | 0.016±0.001 | 41.34±0.02 | 64.26±0.02 | 35.55±0.00 | 42.19±0.04 |
| | **MEAN** | **0.045±0.001** | **0.018±0.002** | **52.31±0.03** | **54.33±0.03** | **43.80±0.02** | **43.04±0.04** |
| **SEFWI-JUABESO** | **SJ001** | 0.069±0.003 | 0.017±0.00 | 65.85±0.00 | 47.64±0.03 | 45.54±0.04 | 59.03±0.17 |
| | **SJ002** | 0.055±0.002 | 0.014±0.001 | 49.25±0.03 | 64.17±0.02 | 40.65±0.04 | 46.71±0.02 |
| | **SJ003** | 0.078±0.003 | 0.013±0.002 | 67.85±0.04 | 40.64±0.02 | 48.68±0.04 | 63.86±0.03 |
| | **SJ004** | 0.050±0.004 | 0.061±0.063 | 48.65±0.04 | 48.28±0.06 | 63.28±0.03 | 47.78±0.02 |
| | **SJ005** | 0.078±0.005 | 0.022±0.001 | 44.24±0.02 | 61.17±0.04 | 47.60±0.07 | 41.26±0.01 |
| | **MEAN** | 0.066±0.003 | 0.025±0.013 | 55.17±0.03 | 52.38±0.03 | 49.15±0.04 | 51.73±0.05 |
| **BOGOSO** | **SB001** | 0.029±0.002 | 0.020±0.002 | 68.55±0.00 | 69.57±0.02 | 42.27±0.02 | 47.86±0.03 |
| | **SB002** | 0.080±0.002 | 0.017±0.002 | 69.85±0.04 | 46.18±0.06 | 49.91±0.05 | 53.18±0.05 |
| | **SB003** | 0.042±0.003 | 0.033±0.002 | 46.79±0.04 | 42.71±0.05 | 43.25±0.04 | 41.68±0.05 |
| | **SB004** | 0.038±0.003 | 0.044±0.002 | 41.28±0.03 | 36.16±0.02 | 45.25±0.04 | 49.35±0.03 |
| | **SB005** | 0.068±0.003 | 0.057±0.003 | 57.28±0.03 | 47.18±0.03 | 55.45±0.00 | 68.25±0.02 |
| | **MEAN** | **0.05±0.003** | **0.03±0.002** | **54.75±0.03** | **48.36±0.04** | **47.23±0.03** | **52.06±0.04** |

**Table 4.4 Levels of some heavy metals (mg/kg) in samples of cocoa beans from some cocoa- growing towns in the Ashanti region of Ghana.**

| TOWN | SAMPLE | Cd | Pd | Cu | Mn | Fe | Zn |
|---|---|---|---|---|---|---|---|
| **JUASO** | **AJ001** | 0.066±0.005 | 0.014±0.001 | 61.29±0.15 | 61.17±0.03 | 66.45±0.14 | 48.86±0.02 |
| | **AJ002** | 0.052±0.00 | 0.021±0.001 | 49.87±0.10 | 46.26±0.02 | 48.43±0.06 | 49.78±0.04 |
| | **AJ003** | 0.078±0.005 | 0.021±0.001 | 43.49±0.15 | 41.72±0.04 | 68.16±0.01 | 60.64±0.09 |
| | **AJ004** | 0.051±0.002 | 0.032±0.001 | 55.65±0.09 | 49.39±0.04 | 43.81±0.03 | 57.27±0.02 |
| | **AJ005** | 0.042±0.003 | 0.012±0.001 | 60.56±0.12 | 37.20±0.05 | 44.34±0.14 | 62.15±0.02 |
| **MEAN** | | **0.058±0.003** | **0.02±0.001** | **54.17±0.1** | **47.15±0.04** | **54.24±0.08** | **55.74±0.04** |
| **TEPA** | **AT001** | 0.085±0.00 | 0.012±0.001 | 43.75±0.10 | 67.71±0.02 | 63.67±0.12 | 48.94±0.03 |
| | **AT002** | 0.070±0.002 | 0.034±0.002 | 48.38±0.04 | 41.25±0.03 | 39.27±0.72 | 44.91±0.04 |
| | **AT003** | 0.076±0.00 | 0.015±0.001 | 44.56±0.09 | 71.15±0.03 | 48.45±0.05 | 42.93±0.02 |
| | **AT004** | 0.040±0.002 | 0.014±0.001 | 41.48±0.13 | 56.25±0.02 | 71.26±0.11 | 67.16±0.03 |
| | **AT005** | 0.024±0.00 | 0.013±0.001 | 63.36±0.03 | 49.68±0.03 | 44.39±0.04 | 61.15±0.02 |
| | **MEAN** | **0.059±0.0008** | **0.018±0.001** | **48.31±0.08** | **57.21±0.03** | **53.41±0.21** | **53.02±0.03** |
| **BEKWAI** | **AB001** | 0.049±0.002 | 0.011±0.000 | 38.55±0.10 | 61.14±0.02 | 51.58±0.03 | 66.24±0.03 |
| | **AB002** | 0.041±0.003 | 0.012±0.001 | 63.65±0.10 | 48.35±0.03 | 47.81±0.06 | 71.14±0.02 |
| | **AB003** | 0.064±0.001 | 0.013±0.002 | 39.86±0.04 | 57.21±0.04 | 69.80±0.04 | 62.38±0.04 |
| | **AB004** | 0.045±0.005 | 0.023±0.001 | 47.81±0.05 | 63.82±0.02 | 42.17±0.07 | 49.35±0.03 |
| | **AB005** | 0.052±0.002 | 0.019±0.001 | 47.27±0.04 | 43.27±0.02 | 39.78±0.04 | 44.44±0.03 |
| **MEAN** | | **0.050±0.003** | **0.016±0.001** | **47.43±0.07** | **54.76±0.03** | **50.23±0.05** | **58.71±0.03** |
| **OBUASI** | **AO001** | 0.062±0.001 | 0.016±0.001 | 41.71±0.04 | 48.87±0.05 | 66.59±0.06 | 49.18±0.03 |
| | **AO002** | 0.075±0.00 | 0.022±0.002 | 36.26±0.06 | 66.14±0.04 | 72.36±0.09 | 66.24±0.02 |
| | **AO003** | 0.057±0.003 | 0.012±0.001 | 56.75±0.10 | 66.73±0.02 | 48.86±0.07 | 60.78±0.03 |
| | **AO004** | 0.068±0.002 | 0.019±0.001 | 62.78±0.05 | 62.18±0.02 | 64.90±0.05 | 66.24±0.04 |
| | **AO005** | 0.061±0.004 | 0.014±0.001 | 41.81±0.05 | 39.22±0.03 | 66.63±0.09 | 44.93±0.02 |
| **MEAN** | | **0.065±0.002** | **0.017±0.001** | **47.86±0.06** | **56.63±0.03** | **63.87±0.07** | **57.47±0.03** |
| **MAMPONG** | **AM001** | 0.060±0.002 | 0.013±0.001 | 38.94±0.02 | 44.36±0.02 | 63.48±0.14 | 49.28±0.03 |
| | **AM002** | 0.042±0.003 | 0.016±0.001 | 47.75±0.10 | 72.64±0.02 | 43.40±0.05 | 66.24±0.02 |
| | **AM003** | 0.056±0.004 | 0.012±0.001 | 61.35±0.06 | 58.75±0.03 | 53.25±0.00 | 60.78±0.03 |
| | **AM004** | 0.045±0.002 | 0.017±0.002 | 42.79±0.04 | 66.16±0.03 | 49.62±0.07 | 66.24±0.04 |
| | **AM005** | 0.050±0.002 | 0.013±0.001 | 47.70±0.05 | 44.79±0.04 | 47.36±0.09 | 44.93±0.02 |
| **MEAN** | | **0.051±0.003** | **0.014±0.001** | **47.71±0.05** | **57.34±0.03** | **51.42±0.07** | **57.49±0.03** |

**Table 4.5 The Mean heavy metal concentrations (mg/kg) in cocoa beans from some of the various cocoa-growing towns in the Western region of Ghana**

| TOWN | Cd | Pd | Cu | Mn | Fe | Zn |
|---|---|---|---|---|---|---|
| **KASAPEN** | 0.045±0.001 | 0.013±0.0004 | 51.19±0.05 | 64.65±0.03 | 53.11±0.04 | 50.67±0.03 |
| **ENCHI** | 0.065±0.002 | 0.015±0.003 | 46.47±0.05 | 56.19±0.02 | 44.28±0.03 | 43.94±0.03 |
| **ASEMPANEYE** | 0.045±0.001 | 0.018±0.002 | 52.31±0.03 | 54.33±0.03 | 43.80±0.02 | 43.04±0.04 |
| **JUABESO** | 0.066±0.003 | 0.025±0.013 | 55.17±0.03 | 52.38±0.03 | 49.15±0.04 | 51.73±0.05 |
| **BOGOSO** | 0.05±0.003 | 0.03±0.002 | 54.75±0.03 | 48.36±0.04 | 47.23±0.03 | 52.06±0.04 |
| **MEAN** | **0.054±0.002** | **0.02±0.0004** | **51.98±0.04** | **55.18±0.03** | **47.51±0.03** | **48.29±0.04** |

**Table 4.6 The Mean of the concentrations of heavy metals (mg/kg) in cocoa beans from some of the cocoa-growing towns in the Ashanti region of Ghana.**

| TOWN | Cd | Pb | Cu | Mn | Fe | Zn |
|---|---|---|---|---|---|---|
| **JUASO** | 0.058±0.003 | 0.02±0.001 | 54.17±0.1 | 47.15±0.04 | 54.24±0.08 | 55.74±0.04 |
| **TEPA** | 0.059±0.0008 | 0.018±0.001 | 48.31±0.08 | 57.21±0.03 | 53.41±0.21 | 53.02±0.03 |
| **BEKWAI** | 0.05±0.003 | 0.016±0.001 | 47.43±0.07 | 54.76±0.03 | 50.23±0.05 | 58.71±0.03 |
| **OBUASI** | 0.065±0.002 | 0.017±0.001 | 47.86±0.06 | 56.63±0.03 | 63.87±0.07 | 57.47±0.03 |
| **MAMPONG** | 0.051±0.003 | 0.014±0.001 | 47.71±0.05 | 57.34±0.03 | 51.42±0.07 | 57.49±0.03 |
| **MEAN** | **0.056±0.0002** | **0.017±0.001** | **49.10±0.07** | **54.62±0.03** | **54.63±0.10** | **56.49±0.03** |

**Table 4.7 The Mean of the concentrations of heavy metals (mg/kg) in cocoa beans from the Western and Ashanti regions of Ghana West –Africa.**

| REGION | Mean Cd | Mean Pd | Mean Cu | Mean Mn | Mean Fe | Mean Zn |
|---|---|---|---|---|---|---|
| **WESTERN** | 0.054±0.002 | 0.02±0.0004 | 51.98±0.04 | 55.18±0.03 | 47.51±0.03 | 48.29±0.04 |
| **ASHANTI** | 0.056±0.0002 | 0.017±0.001 | 49.10±0.07 | 54.62±0.03 | 54.63±0.10 | 56.49±0.03 |

**Percentages of ash, fibre, protein, moisture, carbohydrate and fat in cocoa beans from Western and Ashanti regions.**

**Table 4.8 Moisture**

| Sample Town | Wt of crucible | W6 of crucible + sample | Wt of crucible + Dry sample | Wt, of sample taken | Wt, of moisture | % moisture | Mean |
|---|---|---|---|---|---|---|---|
| **Kasapen 1** | 38.7167 | 40.8382 | 40.7714 | 2.1216 | 0.0668 | 3.15 | 3.15 |
| **2** | 43.6403 | 45.8273 | 45.7584 | 2.1870 | 0.0689 | 3.15 | |
| **Asempaneye 1** | 21.7893 | 23.8484 | 23.7593 | 2.0591 | 0.0891 | 4.44 | 4.39 |
| **2** | 23.7281 | 25.7360 | 25.6469 | 2.0079 | 0.0891 | 4.44 | |
| **Bekwai 1** | 42.1867 | 44.2172 | 44.1255 | 2.030 | 0.0917 | 4.52 | 4.46 |
| **2** | 48.9408 | 51.0137 | 50.9226 | 2.0729 | 0.0911 | 4.39 | |
| **Juaso 1** | 42.1129 | 44.1415 | 44.0957 | 2.0286 | 0.0458 | 2.26 | 2.23 |
| **2** | 43.2416 | 45.2521 | 45.2078 | 2.0105 | 0.0443 | 2.20 | |

**Table 4.9 Ash**

| | | | ASH | | | | |
|---|---|---|---|---|---|---|---|
| **Sample** | **Wt of crucible** | **W6 of crucible + sample** | **Wt of crucible + Dry sample** | **Wt, if sample taken** | **Wt, of Ash** | **% ash** | **Mean** |
| **Kasapen 1** | 21.1226 | 23.1752 | 21.1958 | 2.0526 | 0.0732 | 3.57 | 3.40 |
| **2** | 19.5487 | 21.5507 | 19.6132 | 2.0020 | 0.0645 | 3.22 | |
| **Asempaneye1** | 19.7290 | 21.7595 | 19.7982 | 2.0305 | 0.0692 | 3.41 | 3.69 |
| **2** | 18.7902 | 20.8305 | 18.8709 | 2.0403 | 0.0807 | 3.96 | |
| **Bekwai 1** | 18.1538 | 20.2742 | 18.2366 | 2.1204 | 0.0828 | 3.90 | 3.52 |
| **2** | 18.8804 | 20.9438 | 18.9451 | 2.0634 | 0.0647 | 3.14 | |
| **Juaso 1** | 19.6858 | 21.7326 | 19.7494 | 2.0468 | 0.0636 | 3.11 | 3.19 |
| **2** | 18.7208 | 20.7861 | 18.7883 | 2.0653 | 0.0675 | 3.27 | |

**Table 4.10 Fat**

| Sample | Wt of flask | W6 of flask + fat | Wt of sample taken | Wt, of Fat | % Fat | Mean |
|---|---|---|---|---|---|---|
| **Kasapen 1** | 121.6800 | 122.6110 | 2.0373 | 0.9310 | 45.70 | 45.52 |
| **2** | 122.9810 | 123.9050 | 2.0386 | 0.9240 | 45.33 | |
| **SefwiAsempaneye 1** | 123.4610 | 124.3470 | 2.0256 | 0.8860 | 43.74 | 43.85 |
| **2** | 122.8610 | 123.7530 | 2.0297 | 0.8920 | 43.94 | |
| **Bekwai 1** | 118.3380 | 119.2740 | 2.0417 | 0.9360 | 45.84 | 45.57 |
| **2** | 119.4240 | 120.3450 | 2.0331 | 0.9210 | 45.30 | |
| **Juaso 1** | 183.9980 | 184.7420 | 2.0317 | 0.7440 | 36.62 | 36.72 |
| **2** | 179.4240 | 180.2380 | 2.0421 | 0.7520 | 36.82 | |

**Table 4.11 Fibre**

| Sample | Wt of crucible +Asbestos +fibre | W6 of crucible + Asbestos | Wt of Sample taken | Wtt of fibre | % fibre | Mean |
|---|---|---|---|---|---|---|
| **Kasapen 1** | 25.7019 | 25.6808 | 2.0373 | 0.0211 | 1.04 | 1.03 |
| **2** | 24.1040 | 24.0832 | 2.0386 | 0.0208 | 1.02 | |
| **SefwiAsempaneye 1** | 29.6899 | 29.6695 | 2.0256 | 0.0204 | 0.01 | 1.02 |
| **2** | 11.0339 | 11.0132 | 2.0297 | 0.0207 | 0.02 | |
| **Bekwai 1** | 11.5763 | 11.5543 | 2.0417 | 0.0220 | 1.08 | 1.09 |
| **2** | 25.7076 | 25.6852 | 2.0331 | 0.0224 | 1.10 | |
| **Juaso 1** | 24.0715 | 24.0495 | 2.0317 | 0.0220 | 1.08 | 1.07 |
| **2** | 11.6438 | 11.6222 | 2.0421 | 0.0216 | 1.06 | |

**Table 4.12 PROTEINS**

| Sample | Initial Reading | Final Reading | Titric | Average Titre | Wt, of sample taken | % Protien |
|---|---|---|---|---|---|---|
| **Kasapen 1** | 0.00 | 3.55 | 3.55 | | 2.0456 | 14.34 |
| **2** | 3.55 | 7.00 | 3.45 | 3.50 | | |
| **Asempaneye 1** | 7.00 | 10.45 | 3.45 | | 2.0561 | 13.97 |
| **2** | 10.45 | 13.85 | 3.40 | 3.43 | | |
| **Bekwai 1** | 13.85 | 17.20 | 3.35 | | 2.0398 | 13.74 |
| **2** | 17.20 | 20.55 | 3.35 | 3.35 | | |
| **Juaso 1** | 20.55 | 24.00 | 3.45 | | 2.0465 | 13.91 |
| **2** | 24.00 | 27.35 | 3.35 | 3.40 | | |

**Table 4.13 CARBOHYDRATES**

| | |
|---|---|
| **Kasapen** | 32.56 |
| **Sefwi- Asempaneye** | 32.08 |
| **Bekwai** | 31.62 |
| **Juaso** | 42.88 |

**% of carbohydrate = 100% -%Moisture+%Fat +%Protein +%Fibre + %Ash**

### Levels of heavy metals in cocoa beans

### Lead

With reference to the analysis performed in this study, a mean lead concentration ranging from 0.013mg/kg t in samples from Kasapen to 0.03mg/kg in samples from Bogoso was obtained in cocoa beans farm the individual to town of the western region of Ghana. This was obtained by computing the mean lead concentrations of the individual samples from the various towns in the Western region of Ghana (Kasapen, Asenpanaye, Juabeso, Enchi and Bogoso) However, the mean lead concentrations in cocoa beans from the aforementioned towns in the Western region were found to be quite uniform. The level of lead obtained in this work compares favourably with lead level in most fruits and vegetables. Codex alimentarius has set a maximum of 0.1mg/kg for most of these fruits and vegetables. However, the level of lead in cocoa beans from the western region in comparison with the codex alimetarius maximum level is within the permissible limits. It is worth-mentioning that the level of lead in the individual five samples from each town before the mean for the towns were calculated were also found within the permissible limits from table 4.3. This means that the unprocessed cocoa beans from the western regions of Ghana are of quality in terms of the levels of lead in them. Below compares the mean concentrations of lead in cocoa beans from the sample towns in the Western region.

**Figure 4.28 Comparison of the levels of lead in cocoa beans from the Western region with the codex maximum limits for lead (Pb)**

Similar observations were made concerning the mean lead concentrations of individual samples towns in the Ashanti region of Ghana. The mean lead concentrations, with reference to table 4.4 ranged from 0.014mg/kg in samples from Mampong to 0.02mg/kg in samples from Juaso in the Ashanti region of Ghana. Comparatively, the levels of lead in cocoa beans from the Ashanti region of Ghana also fall well below the maximum permissible limits. Below compares the levels of mean lead in cocoa beans from the various sample towns in the Ashanti region with the condex maximum limits.

**Figure 4.29 Comparison of the levels of lead (Pb) in cocoa beans from the sample towns in Ashanti region with the codex maximum limit for Pb.**

Finally the mean lead concentrations in cocoa beans from the sample town in the western and Ashanti region were also compared with the codex maximum limits. Again, the levels of lead in all cocoa samples from western and Ashanti regions were found to falls below the codex maximum limits as:

**Figure 4.30 Comparison of the concentration of lead (Pb) in cocoa samples from Western region and Ashanti region with codex maximum limit**

Comparatively, while the levels of lead obtained in this study fall well below the maximum permissible limits, Rankin etal (2005) has reported even lower levels of lead (0.005kmg/kg) in cocoa beans from Nigeria using ICP-MS.

Lead is very detrimental to human health and it is regarded to be one of the most toxic metals with higher probability of incorporating into most food. In most diseases, lead poisoning has been implicated and it is considered by the united states of Agency for Toxic substances and Disease Registry (ATSDR) as one of the most toxic metals. What exacerbates the situation is that the targets of lead toxicity are the cardiovascular system (Heart), Kidneys and the developing nervous systems. Owing to this children have been found to be more susceptible to the health effects of lead toxicity than adults.

Cocoa products have been reported to contain relatively high levels of lead as compared to other similar food products. (Dahiya, Karpe, Hedge & Sharma, 2005). Though cocoa beans as a raw material for the manufacture of cocoa products contribute to the elevated levels of lead in cocoa products the greatest percentage of lead contamination of cocoa products occur during processing of beans due to various industrial process that may require the use of metal parts some of which may contain trace amounts of the metal (Rakin, Nriagu, Agyarwal, Arowolo, Adebayo &Flegal, 2005). Anthropogenic activities are the main sources of increase in the levels of lead in both soil and food. This is because lead has been used in a number of applications such as paints, car batteries and as components in anti-knocking agents in fuel.

### Cadmium

Table 4.3 and Table 4.4 show the mean (±standard deviation) of the concentrations of cadmium in cocoa beans from the various towns in western and Ashanti regions of Ghana respectively. The concentration of cadmium (Cd) in cocoa beans from the western region ranged from 0.045mg/kg in samples from Kasapen and

Asempaneye to 0.66mg/kg in samples from Juabeso. That of Ashanti region ranged from 0.05mg/kg in samples from Bekwai to 0.065mg/kg in samples from Obuasi. The levels of cadmium in samples from the towns in the Western region as well as Ashanti region were found to be fairly uniform. Though the specific allowable limits for cocoa beans are not readily available, the levels of Cd obtained in this work compares favourably with the maximum limit of 0.1mg/kg fresh weight set for most plant parts (excluding the leaves). The mean levels of cadmium in cocoa beans from western and Ashanti region were found to be 0.054mg/kg and 0.056mg/kg respectively. Though the levels were found to be closer to the permissible limits, they still fall below the allowable limits set by codex as presented graphically below:

**Figure 4.31 Comparison of the mean levels of cadmium in cocoa beans from the Western and Ashanti regions with codex maximum limits**

The availability of cadmium in cocoa beans may be attributed to its natural occurrence in the soil and anthropogenic activities. Cadmium levels in the soil ranges from 0.01 – 7mg/kg (Bohn, McNeal, O' Connor, 1985 )Because of the high cation exchange capacity of most tropic soil the levels of heavy metals such as cadmium in the tropic soil are said to be very low (Nartey, 2009). Cadmium which is found most fertilizers may get incorporated in plants grown with the use of these fertilizers. Once cadmium gets into the soil it is found to accumulate in the top soil where it binds strongly to organic matter and prevented from entering soil solution. The high sorption of capacity of tropic soil may prevent its access to the plant and so reduce the risk of contamination in food. The involvement and availability of cadmium in soil is low and best under low pH conditions (Nartey,2009). Thus the use of pesticides containing trace amounts on cocoa plant to prevent and cure numerous diseases is a major contributory factor of the presence of heavy metals in soil and may consequently accumulate in plant and soil over time which may lead to toxicity. It is reported that cadmium tends to accumulate more in leaves of plants than in the seeds **(**Herrero& Martin, 1993; Allotey, Jackson & Morgan, 1990**).** The seriousness of cadmium toxicity is that it accumulates in all levels of food chain and concentrate in the liver and kidneys causing disorders. The pathway of cadmium toxicity is through consumption of contaminated food. However, the levels of cadmium in cocoa beans analyzed in the work may not pose any significant health hazard due to relatively low concentrations. The levels are in agreement in agreement with cadmium levels in cocoa reported by Mounicou et al (2003).

**Other Elements (Zinc, Copper, Manganese and Iron)**

The presences of these metals in food mostly are regarded relatively non- toxic or essential in living organisms. However, the levels of these metals analyzed in this work were much higher than the levels of cadmium and lead which are considered irrelevant in living organisms and are termed as toxic elements or metals. The mean levels of these metals in cocoa beans from the towns in the Western and Ashanti region are shown in table 4.5 and 4.6 respectively. Iron and Zinc were the highest in all the samples in this work corroborating their normal composition in plants relative to other metals. The levels of these metals are mostly not of significant health interest especially at low concentration because they are considered to be essential in plants, animals and man. Conversely, their presence at high concentration beyond requirements may pose health problem to living organisms by interfering with the functions of other essential elements.

The mean concentration of Zinc ranged from a low of 43.04mg/kg in samples fromAsempaneye in the Western region to 58.71mg/kg in samples from Bekwai in the Ashanti region. That of iron ranged from 43.80mg/kg in samples from Asempaneye in the Western region to 63.874mg/kg in samples from Obuasi in the Ashanti region. However, the concentrations of Zn and iron in cocoa beans from the Western region , upon the analysis, were found to be of lower levels as compared to those from Ashanti region of Ghana. These relative lower levels of these metals recorded from the Western region may be attributed to factors such as heavy rainfall pattern in the region, soil characteristics and low levels in the soil. The Western region is known to have the highest rainfall pattern in Ghana, a condition which may bring about leaching of most of these ions.

The mean concentrations of Manganese and copper were also high with that of Manganese ranging from 43.5mg/kg in samples from Asemaneye in the Western region to 68.74mg/kg in samples from Juaso in the Ashanti region. The mean concentration of Manganese of and Copper were also high with that of Manganese ranging from 47.15mg/kg in samples from Juaso in the Ashanti region to 64.65mg/kg in sample from Kasapen in the Western region. That of copper also ranged from low of 46.47mg/kg in samples from Enchi to a high of 55.17mg/kg in samples from Juabeso all in the Western region (Table4.5 and4.6). Cocoa is known to be a rich source of copper. Due to the continual use of copper –containing pesticides on cocoa farm to avert pest and diseases, these high levels of the metal in the beans are likely to increase over time. Copper intake in the body has been found to be primarily through diet, though this amount usually does not exceed the average dietary requirements of 10-12mg/day for adults **(**WHO, 1998**).**

The probable increase in the levels of copper in Ghana over the years ahead is as a result of the fact that copper is probably the most widely used heavy metal on cocoa farms as fungicide. Oxides and hydroxides of copper are used as fungicides and sprayed on the cocoa plants about four times each to prevent and control the occurrence of diseases and pests on the cocoa farm. Fungicides such kocide 101 (Cu $(OH)_2$, 22%) and nordox 50 ($Cu_2O$, 50%) contain high levels of copper which upon application, get incorporated into the cocoa plant and the soil and may accumulate Irrespective of their concentration in the soil however, the uptake of most heavy such as copper by plants is dependent on a number on a factor such as pH, organic matter content, state of the metal etc. the uptake of copper for instance is improved under acidic conditions and the presence of high organic matter content of the soil (Tang, Wilke& Huang, 1999).

In spite of the fact copper occurs naturally in all plants (and animals) and essential in a number of biological processes in the body such as hemoglobin formation, drug and carbohydrate metabolism and antioxidant defense mechanisms, at excessive levels however, toxicity may result. Mining activities and the application of phosphate fertilizers account for two of the main sources of release of copper into the soil and in the plants (Georgopoulos, Yonone-Loy &Opiekum, 2001). In cocoa farm however the single largest sources of copper is pesticide usage. Manganese is one of the commonest as well as most abundant metals in the environment. Manganese and its compounds exist naturally in the environment though its levels may be increased through human activities such as mining operations, burning of fossil fuels and the addition of pesticides and fertilizers on the farm.

One major source of Manganese is that an organic compound of manganese; methylcyclopentadienyl manganese tricarbonyl has been used as a fuel additive to control knocking in fuel combustion engines over the decades in Ghana. This ends up with the release of manganese from fuel combustion into the environment which eventually gets into the soil by precipitation through rainfall. Once manganese settles on the soil, the soil type and its chemical state are some of the factors that determine its mobility. Manganese is an essential trace element necessary for good health. In humans and animals, some of the remarkable roles played by are: bone mineralization, protein and energy metabolism, metabolic regulation, cellular protection from damaging free radical species etc. (Wedler, 1994).

## Conclusion

The analysis of the metals Cd. Pb, Cu, Mn, Fe, and Zn in cocoa beans were performed by use of AAS instrument. The reliability of the procedures employed in the analysis was ascertained by the use of two standard reference materials which were in good agreement with their reported valuesHowever, it should be emphasized that the concentrations of the metals obtained in this work were below the recommended maximum limits set by WHO/FAO and hence possess less or no risk upon consumption. From this work done, it can be concluded that cocoa beans produced in the country, mainly from the Western and Ashanti regions provide generally safe levels of heavy metals and of good and quality nutritional values when consumed.

From the results of the analysis, metals concentrations did not differ significantly among the regions samples except in the case of Cd and Cd which had relatively low levels in the western region as well as the Ashanti region of the country. The generally uniform levels of the metals in the various regions may be due to similar soil types and characteristics as well as general agricultural practices occurring within the various cocoa growing regions of the country. The concentration of Cu, Mn, Zn, and Fe were observed to be comparatively quite high. Though, these metals are considered as essential elements in plants, higher concentrations than needed may pose some level of risk upon consumption. The relatively high levels of Cu are most likely due to its significant application on the cocoa farms as fungicides. Alternative sources of fungicides may go a long way curbing the relatively high levels of this metal.

## Recommendations

Since the processing of cocoa beans usually involves the pressing of the pulverized cocoa nib (cocoa mass/liquor) to separate the solids (powder/cake) form the cocoa butter (fats), it is recommended that a research is conducted to find which of the separated parts contains the highest levels of heavy metals and vice versa for scientific advancements.

Also, the processing of cocoa beans involves the separation of the shells from the nib and so it is encouraged that the determination of the levels of heavy metals in both the shells and the nibs be made separately for comparison.

Analysis of soils from the various cocoa -growing regions is highly encouraged to determine the levels of these metals in the soil and the extent of uptake into the cocoa plant.

Research to determine the levels of heavy metals especially, Hg in cocoa beans from the mining towns is recommended to ascertain the real effect of mining activities on the levels of heavy metals in cocoa beans.

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