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DEPARTMENT OF FOOD SCIENCE AND TECHNOLOGY

HEAVY METAL CONTAMINANTS OF SELECTED CULINARY HERBS AND SPICES AVAILABLE IN SOME GHANAIAN MARKETS

BY

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JUNE, 2018

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A THESIS SUBMITTED TO THE DEPARTMENT OF FOOD SCIENCE AND TECHNOLOGY IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF THE DEGREE OF MASTER OF SCIENCE IN FOOD QUALITY MANAGEMENT

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DECLARATION

I, hereby declare this thesis is the outcome of my own original research toward the award of the MSc. Degree and that, to the best of my knowledge, it contains no material previously published by another person or material which has been accepted for the award of any degree or diploma at Kwame Nkrumah University of Science and Technology, Kumasi or any other educational institution, except where due acknowledgment has been made in the text.

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ABSTRACT

There is increasing global concerns about the safety of culinary herbs and spices available on market shelves. The Food and Drugs Authority (FDA) of Ghana rarely samples culinary herbs and spices on the market for its safety. To ascertain the safety of culinary herbs and spices on the Ghanaian market, the concentrations of Cd, Cr and Pb in ten commonly and frequently used culinary herbs and spices sampled from four large markets in the Accra and Kumasi Metropolis were determined. The dried samples were taken through wet digestion, in triplicate, with a 12 ml HCl and HN03 acid mix to obtain clear solutions which were subsequently analyzed with AAS. All the samples studied contained no detectable traces of Cd. Five of the ten sampled culinary herbs and spices contained detectable traces of Pb, whilst two of the ten contained detectable traces of Cr. Turmeric recorded the highest mean Cr concentration (0.42± 0.03 mg/kg). The mean concentrations of Pb present in five of the sampled herbs and spices exceeded the maximum permissible limit set by the WHO at 0.1 mg/kg (Anise; 8.95± 0.05 mg/kg, Cloves; 5.50±0.05 mg/kg, Fennel; 6.52±0.06 mg/kg, Rosemary; 4.55±0.06 mg/kg and Turmeric 0.85 ±0.08 mg/kg). The results indicate that the culinary herbs and spices on the market may contain high levels of heavy metals which are a potential risk to human health.

TABLE OF CONTENT

LIST OF TABLES

LIST OF EQUATIONS

ABBREVIATIONS

CAC	:	Codex Alimentarius Commission of the FAO/WHO

FOA	:	Food and Agriculture Organization of the United Nations
WHO	:	World Health Organization
JECFA	:	Joint Expert Committee on Food Additives of FAO/WHO
IARC	:	International Agency for Research on Cancer
CRN	:	Council for Responsible Nutrition
AAS	:	Atomic Absorption Spectroscopy
µg/kg bw	:	microgram per kilogram per body weight
mg/kg bw	:	milligrams per kilogram per body weight
ppb	:	parts per billion
ppm	:	parts per million
FDA	:	Food and Drugs Authority
GSA	:	Ghana Standards Authority
ESA	:	European Spice Association
ECHA	:	European Chemical Agency
MPL	:	Maximum permissible limit

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DEDICATION

This work is dedicated to the memory of Edna Nortey Lantei, caterer par excellence! You will be sorely missed but never forgotten.

CHAPTER ONE

INTRODUCTION

1.1 Background

Since hunter gatherer times when human ancestors accidentally realized the health benefits and taste enhancing properties of certain leaves, roots, and berries, the use of herbs and spices has been gradually refined, with culinary herbs spices spanning a wide range of plants (Halvorsen *et al.*, 2006).

The beginning of the global herbs and spices trade circa 3000 BC made herbs and spices a worldwide sensation and they have since taken the culinary world by storm –from the blandest and mildest to the spiciest and most colorful (Zheng and Wang, 2001). The earliest recording of herbs and spices for medicinal purposes was in 1555 BC in Egypt by Papyri in ancient Egypt (Tapsell *et al.*, 2006).

Culinary herbs and spices are those that are derived from the flower, stigma, root, bark, leaf, etc of plants for flavoring and enhancing the taste of food (Opara *et al.*, 2014) and are used in cooking to flavor, spice or add color to dishes. With the surge in health consciousness and individual desire to eat healthy, culinary herbs and spices are more in demand now and are the favored choice over artificial flavor enhancers that dominated the spice rack for decades (Lampe, 2003). However, proliferation of anthropogenic activities such as mining, agriculture with the application of sewerage sludge and fertilizers (Roy and Gupta, 2016) and use of leaded fuels compounded with the growth of human populations have led to continuous pollution of the environment with heavy metal deposits (Slavík *et al.*, 2012). Heavy metals like cadmium (Cd), Lead (Pb) and Chromium (Cr) deposits in the environment are a direct result of such anthropogenic activities and these deposits are eventually transferred from the soil to crops and water sources (Krejpcio *et al.*, 2007).

Heavy metal contamination of foods, culinary herbs and spices and other food sources can occur directly through contaminated soil, agricultural practices (Dudka *et al.*, 1996; Slavík *et al.*, 2012) as well as from storage and processing practices according to the European spice association (ESA, 2013). This poses a food safety concern because food can only be deemed safe for consumption if its intake will pose no harm when prepared or eaten according to its purposed use (Haas *et al.*, 1999).

The periodic table of elements does not have a classification for heavy metals. The metals that are clinically classified as heavy metals fall within the transition metals and poor metals sections of the periodic table (Duffus, 2002). Due to the term heavy, it will be precise that it refers to elements with atomic weights of 200.5 daltons and above but most of them do not have this high atomic weight (Duffus, 2002). In clinical terms therefore, heavy metals refer to any metal to which exposure is undesirable and which posses a potential hazard to those it comes in contact with (Baldwin *et al.*, 1999).

Since the first incidence of food poisoning by ingesting contaminated rice (Leblanc *et al.*, 2000), heavy metals of clinical importance include arsenic (As), cadmium (Cd), chromium (Cr), zinc (Zn), lead (Pb), cobalt (Co), copper (Cu), and mercury (Hg). Heavy metals, like zinc, copper, cobalt and chromium in the right doses have beneficial purposes in human systems by being integral parts of coenzymes (Nkansah *et al.*, 2010) which drive the metabolic pathway or metabolic processes. Other heavy metals like Hg, As, Pb are highly injurious to human health, even in small doses and can cause acute illnesses or accumulate in the body to pose chronic health problems at a later time (Baldwin *et al.*, 1999).

The rate of human contact with toxic heavy metals has increased due to a population growth which places a huge demand on industry and the land. Industrial spills and improper disposal of chemical waste contaminate soils which knowingly or unknowingly may be used to cultivate crops (Dudka *et al.*, 1996). Anthropogenic activities in nature such as mining, fertilizer based agriculture (including aquaculture) and fishing with chemicals (Eneji and Annune, 2011) also contribute to conveying and depositing heavy metals in soils and water that could be used for cultivating crops. Cadmium, lead and zinc have contaminated large areas of farmland reclaimed from historic or current mining sites (Dudka *et al.*, 1996). In Ghana for instance, the current proliferation of illegal small scale mining activities locally known as *galamsey* in the Pra River Basin (PRB) which is accompanied with the unregulated use of mercury for detecting the gold ore has led to a contamination of the soil and water bodies in the area (Yidana *et al.*, 2011).

Recent studies have also uncovered the unsanitary practice of using waste water to irrigate vegetables meant for consumption (Roy *et al.*, 2016) as well the effects of unsupervised use or overuse of chemicals for the cultivation of many vegetables and herbs (Nkansah *et al.*, 2010). The recent cases of the detection of elevated levels of Lead in some brands of turmeric in the United States of America has brought into sharp focus the health risks which may be inherent in consuming certain culinary herbs and spices (Addady, 2016). The results of investigations carried out by the FDA of the United States of America caused these brands of turmeric to be withdrawn from the shelves (Cowell *et al.*, 2017). The joint FAO and WHO standards program through the Codex Alimentarius Commission (CAC) collaborate regularly with the JECFA to examine the results of tests carried out to analyze the dose-response curves of heavy metal contamination. Based on these results, the provisional tolerable monthly and weekly intake (PTMI and PTWI) quantities of heavy metals allowed in foods are revised periodically to ensure the safety of food and water (FAO/WHO, 2011).

1.2 Problem Statement and justification

There is increasing global concern about massive contamination of culinary herbs and spices like curry powder, oregano, black pepper and turmeric (Addady, 2016; Cowell *et al.*, 2017) with heavy metal contaminants like lead. Strictly regulated markets in the US, Europe and other countries which have well resourced regulatory bodies still record incidences of contamination of culinary herbs and spices. Regulatory bodies in Ghana are not regularly sampling culinary herbs and spices on the market to monitor their safety; perhaps because these food additives are not considered a threat. Some studies have been carried to determine the heavy metal concentrations in local herbs and spices in Ghana and Nigeria (Gaya *et al.*, 2016; Nkansah *et al.*, 2010) but the scope of those studies did not include imported spices like turmeric and curry powder. Investigating culinary herbs and spices, including imported spices like turmeric, available on the Ghanaian market for concentrations of heavy metal contaminants and comparing the detected concentrations to existing standards is imperative. It will aid regulatory bodies increase and improve monitoring methods as well as assure the public of the safety of these food ingredients.

1.3 Objectives

1.3.1 Main objective

- To determine the safety of ten (10) selected culinary herbs and spices available in some Ghanaian markets in relation to the heavy metal contaminants; cadmium (Cd), chromium (Cr) and lead (Pb).

1.3.2 Specific objectives

- To determine the concentrations of the heavy metals Cd, Cr and Pb in the sampled culinary herbs and spices.
- To estimate the daily intake (EDI) of the concentrations of Cd, Cr, and Pb found in the sampled culinary herbs and spices.
- Compare the EDI to reference doses (R_fD) obtained from international regulatory bodies to determine the safety of the sampled culinary herbs and spices.

CHAPTER TWO

LITERATURE REVIEW

2.1 Heavy metal contamination of culinary herbs and spices

Heavy metals should in practice refer to metals in the periodic table with atomic mass 200 and above, e.gs mercury and bismuth (Duffus, 2002). Copper (Cu), zinc (Zn), nickel (Ni) are examples of heavy metals required by the body for essential metabolic processes (Nkansah and Amoako, 2010).

Occasional excesses of copper, zinc and nickel in the metabolic pathway yields a range of malfunction in human metabolism and possibly disease (Gaya and Ikechukwu, 2016) but these are rarely classified under heavy metal contamination related clinical cases. In food safety practice, heavy metals have come to encompass any metal which when exposed to human populations may constitute a potential hazard or yield or elicit clinically undesirable physiological responses (Baldwin *et al.*, 1999). Arsenic, cadmium, chromium, mercury, lead and tin are few of the metals of interest in food risk assessment studies.

Pesticide residues are a major pathway for heavy metal contamination of soils and eventually food cultivated on the soil (Dudka *et al.*, 1996; Leblanc *et al.*, 2000). Industrial effluents (Slavík *et al.*, 2012) and workplace accidents or exposure (Baldwin *et al.*, 1999) are other modes of contact with heavy metals which have acute to chronic clinical implications. Increased mining activity (both legal and illegal) which lays arable lands waste, industrialization, improper disposal of chemical wastes and steep competition on the global food market has inadvertently opened a window for the possible contamination of almost very consumable product. The indiscriminate use of chemicals which may contain raw forms or compounds of Cd, Cr and Pb make them available in the ecosystem. Thus the contact rate of planted food or water with these toxic chemicals have increased, increasing their propensity to enter the food chain (Yidana *et* *al.*, 2011). These toxic chemicals are often bioaccumulated or concentrated in plant and animal tissues and hence released into humans who use these plant and animal tissues as food (Eneji and Annune, 2011; Haas *et al.*, 1999).

Evidence of possible heavy metal contamination of foods over the decade have included contamination of rice, potatoes and barley planted in cadmium contaminated fields (Dudka *et al.*, 1996), contamination of red chili with Sudan 1 dye to make it appear fresher, adulteration of ground ginger or paprika with fillers like corn starch (Dhanya *et al.*, 2010) and economically motivated contamination of turmeric with lead chromate (Cowell *et al.*, 2017).

Cadmium sulphide is mainly used as a pigment in paints, plastics, ceramics, glass, textiles, and paper. The major pathways of exposure of cadmium to the general population are through food and water contaminated with cadmium, and also through inhalation of cadmium compounds (IARC, 2011). Cadmium is also a major component in some phosphate fertilizers employed for agriculture (IARC, 2011).

Clinical investigation into the effects of residues of toxic heavy metals in consumables combined with a food safety interest has been a strong focus of the FAO of the WHO. The organization is constantly working to narrow the levels and totally eliminate heavy metal contaminants from food and water, including culinary herbs, spices and condiments Various countries, also concerned with food safety and meeting world trade standards, have set up regulatory bodies to ensure consumers are not put at risk of disease or ill health. It is the duty of such regulatory bodies to check that any consumable placed within the reach of consumers does not contain any potential hazards (Dhanya and Sasikumar, 2010).

In Ghana, the Ghana Standards Authority (GSA) and Food and Drugs Authority (FDA) are responsible for assuring the public of the safety or otherwise of all consumables. It is necessary to assess culinary herbs and spices that are liable to adulteration to check unfair competition among producers and above all assure consumers protection against the fraudulent practices that place them at risk (Dhanya *et al.*, 2010).

2.1.1 Detection of heavy metal contaminants with Atomic Absorption Spectroscopy (AAS)

The premise for atomic absorption spectroscopy is that atoms absorb energy to reach an excited state at a particular wavelength and then emit the absorbed energy at a different wavelength when returning to the ground state (Bader, 2011). Using special light sources, like hollow cathode lamps, and carefully selecting specific wavelengths, individual elements can be specifically determined (Perkin-Elmer, 1996). Each AAS equipment has a limit of detection (LOD) beyond which certain concentrations of elements cannot be detected (Perkin-Elmer, 1996). AAS integration or measuring time of 1-3 seconds is mostly used for routine analyses though precision can be improved by increasing the integration time to a an estimated maximum of 10 seconds, beyond which there is no further improvement on precision (Perkin-Elmer, 1996) AAS has been successfully used to determine the concentrations of elements of interest in food products, (Kurian, 2012; Leblanc, *et al.* 2000). AAS has also been successfully employed to determine the presence and concentrations of heavy metal content in various foods including culinary herbs and spices (Al-Eed, *et al.*,

2002; Gaya and Ikechukwu, 2016; Krejpcio, *et al.*, 2007, Leblanc, *et al.*, 2000; Nkansah and Amoako, 2010). The atomic absorption spectrometer is calibrated with standard a solution of the element of interest before the clear liquid obtained from digesting the food suspected to contain the element of interest is loaded into the AAS equipment for a determination of the concentrations of the element of interest (Bader, 2011, Perkin Elmer 1996).

2.2 Risk assessment of culinary herbs and spices contaminated with heavy metals

Risk refers to the probability or chance of an incident occurring (Haas *et al.*, 1999). Risk assessment is defined as "the process of estimating both the probability that an event will occur, and the probable magnitude of its adverse effects in economic, health or safety related or ecological terms, over a specified period of time" (Haas *et al.*, 1999).

Assessing any food or drink for risk involves doing four specific tasks (Haas *et al.*, 1999); Hazard identification, where the type of hazard and the extent of harm it can cause are defined; Exposure assessment, which involves knowing the concentrations of the identified hazard and estimating how often it may be ingested; Dose response assessment, where the degree of exposure is used to quantify the adverse effects of the hazard; and Risk characterization which refers to employing probabilistic methods to state the potential impact of the hazard. This is done taking into consideration the severity of the hazard's effects as well as the amount or length of exposure to said hazard (Haas *et al.*, 1999).

The Codex Alimentarius Commission (CAC) classifies heavy metal contaminants of culinary herbs and spices as chemical hazards which are toxic (CAC, 2013). The toxicities of heavy metals are determined through observation of their adverse reactions elicited after exposure to certain quantities or concentrations of the heavy metals (Needleman, 2013).

Exposure assessment of toxic heavy metal contaminants indicate that contact is mostly through consuming contaminated food and water; with inhalation and skin contact during accidental spillage making up a smaller proportion (Baldwin *et al.*, 1999; Needleman, 2013; Slavík *et al.*, 2012). It is a fact that the concentrations of contaminants in water can be easily measured or estimated (Eneji and Annune, 2011; Singh and Agrawal, 2010). A bioconcentration factor (BCF) can be used to determine the tendency of a substance in water to accumulate in plant tissue and subsequently be a risk to humans who consume them (Haas *et al.*, 1999). The length of time and the frequency with which these substances are exposed to humans contribute to how much of it is bio-accumulated or biomagnified in human tissue during their lifetime. To narrow in on how much of a contaminant can be safely consumed; dose response assessments are carried out (Haas *et al.*, 1999). This is done by exposing populations of laboratory organisms to specific concentrations/doses of the identified hazard. The responses elicited at the specific administered doses are recorded and used to determine the safety of the potential hazard (Eneji and Annune, 2011; Haas *et al.*, 1999; IARC, 2004; Needleman, 2004).

A dose is defined as the amount of a substance or chemical which when ingested will pose no harm or have adverse effects on health (Haas *et al.*, 1999). Dose-response assessments are carried out is to acquire a mathematical relationship between the quantity or concentration of a toxicant or microorganism a human can be exposed to, and the risk of the negative reactions from that quantity (Haas *et al.*, 1999). If a smaller dose of a toxicant can elicit adverse effects in a considerable population of test subjects, that toxicant is deemed very potent (Haas *et al.*, 1999). The slope of the dose-response curve is called the potency factor (PF) (Haas *et al.*, 1999).

In the course of studying the safety of chemicals, predetermined quantities can be administered to the test population to yield or elicit a specific response. The predetermined quantity or dose is referred to as the benchmark dose (BMD) and the response elicited is the benchmark response (Haas *et al.*, 1999).

Using these principles, the CAC and JECFA are able to set the permissible tolerable weekly intake (PTWI) and permissible tolerable monthly intake (PTMI) levels for cadmium, chromium and lead in food and water. The PTWI and PTMI are aggregations of the R_fD – reference dose; which is the concentrations of these chemicals which on assumption can be consumed without causing any adverse effect to humans. The R_fD is also called the Allowable Daily Intake (ADI) (Haas *et al.*, 1999). The ADI is expressed in mg/kg bw.

To estimate the risk involved in ingesting particular concentrations of a substance three factors being; how much of the substance is consumed daily over a period of time also known as the chronic daily intake, the R_fD, and the potency factor (PF) of the substance in question are considered (Haas *et al.*, 1999). The chronic daily intake (CDI) of any substance under investigation is computed from data which is collected from the sampled population. The mathematical equation used for the computation is depicted in Equation 2.1;

$$CDI = \frac{C \times CR \times EFD}{BW} \times \frac{1}{AT} \qquad \text{(Eq. 2.1)}$$

Where C is the concentration of the toxicant; CR is the contact rate (how much of the toxicant is consumed at a time); EFD is the exposure frequency and duration; how often the toxicant is consumed (days/years) and for how long in the consumer's lifetime (years); BW is the average body weight per kg during the exposure and AT is averaging time; the period over which the exposure is averaged.

The risk of consuming a suspected toxicant which is non carcinogenic is determined using Equation 2.2

$$Risk = PF\,(CDI - R_fD) \qquad \text{(Eq. 2.2)}$$

Where PF is potency factor, CDI is chronic daily intake and R_fD is the reference dose for a non-carcinogen (Haas *et al.*, 1999).

If the calculated risk is equal to or greater than 1 (i.e. Risk $\geq$ 1), then immediate steps must be taken to mitigate the risk by regulating the substance (Haas *et al.*, 1999). There are two methods for characterizing the risk of being exposed to toxicants; one for carcinogenic toxicants and one for non- carcinogenic toxicants, because carcinogenic toxicants do not show the same dose response relationship as non-carcinogenic toxicants (Haas *et al.*, 1999). Whereas the dose response curves for non-carcinogenic toxicants are usually sigmoidal, the dose response curve for carcinogenic toxicants is a straight line *(*Haas *et al.*, 1999; IARC, 2017). The linear relationship of the dose of a carcinogen to the response elicited indicates a high slope and hence a high potency factor PF (Haas *et al.*, 1999).

The implication of this relationship is that there is no tolerable threshold concentration of carcinogenic toxicants which can be safely consumed by humans. Consumption of any dose or amount puts one at risk of the adverse effects associated with consuming such toxicants (Haas *et al.*, 1999). The incremental lifetime risk of consuming a carcinogen is determined using Equation 2.3 below:

$$\text{Risk} = \text{CDI} \times \text{PF} \qquad \text{(Eq. 2.3)}$$

Regulatory agencies like the European Food Safety Association (EFSA) and the Agency for Toxic Substances and Disease Registry (ATSDR) of the USA list the dietary slope factors for hazardous food contaminants.

2.2.1 Human health risk assessment

The estimated health risk associated with consumption of heavy metal contaminants of food under investigation is usually calculated based on an estimated daily intake (EDI) which differs from the CDI only by being an estimate because EDI and CDI connote the same thing *(*Haas *et al.*, 1999). This depicted in Equation 2.4

$$\text{EDI} = \frac{(C \times AC)}{bw} \qquad \text{(Eq. 2.4)}$$

Where C (mg/kg) is the concentration of heavy metal in the raw food; AC is the average dry weight of the raw food consumed by local inhabitants measured in g/day/person and bw is the average body weight.

Studies conducted on different continents have suggested daily culinary herbs and spice intake as low as 4 g/serving and as high as 20.4 g/serving (Lampe, 2003). Meals prepared and consumed daily are observed to contain a mean of 10.4 g/serving of culinary herbs and spices (Siruguri and Bhat, 2015).

2.3 Economic importance of herbs and spices

Culinary herbs and spices are a subset of the wider range of herbs and spices which are used for a variety of purpose spanning aesthetic uses (aromatherapy) to specific medicinal indicative uses (Zheng and Wang, 2001). They are used to enhance the taste, aroma and even appearance of foods (Lampe, 2003). Culinary herbs and spices have been proven to contain microelements which make them useful antioxidants in human metabolism (Kurian, 2012). Numerous research has been carried out to determine the health benefits of some culinary herbs and spices (Halvorsen *et al.*, 2006; Kurian, 2012; Lampe, 2003; Opara and Chohan, 2014; Zheng and Wang, 2001), bacterial load in some culinary herbs and spices (Ahene *et al.*, 2011) as well as the heavy metal content of selected culinary herbs and spices on the local markets of Saudi Arabia (Al-Eed *et al.*, 2002) and Nigeria (Gaya *et al.*, 2016).

2.4 Use of culinary herbs and spices

Ghanaian dishes are mostly spicy and flavorful and no dish is complete without the addition of culinary herbs and or spices during the meal preparation (Nkansah *et al.*, 2010).

Traditional herbs and spices like cloves, negro pepper, ginger, shallots, African locust bean and garlic are used regularly to augment the flavor and taste of many soups, stews, stir fries and are used in marinating sauces for meats or certain foods before they are fried (Ahene *et al.*, 2011; Nkansah *et al.*, 2010).

Until recently imported flavor packets were widely used in homes and restaurants due to the pertaining view that traditional spices were only suited for preparing wholly local cuisine. Recent news articles about the health benefits of using natural, unprocessed herbs and spices have led to a marginal increase in the purchase and use of local culinary herbs and spices such as fennel, cloves, African locust bean, and turmeric (Ahene *et al.*, 2011).

One study found that in comparison to other culinary herbs such as, oregano, sage and rosemary, cloves had a high mean antioxidant value (Kurian, 2012). This confirmed another research work that had been carried out to determine the content of antioxidants present in foods consumed in the United States (Halvorsen *et al.*, 2006). Another extensive research determined the antioxidant activity and phenolic compounds in 27 culinary herbs and 12 medicinal herbs concluding that the culinary herbs contained higher antioxidant capacity (Zheng and Wang, 2001).

The results of most of these studies have been published in the general media and are accessible to the populace. This has led to an increase in the use of a variety culinary herbs and spices, some of which may have inherent risk factors. Without appropriate food risk communication (Haas *et al.*, 1999), these inherent risk factors may be ignored due to the one sided emphasis on their health benefits.

Daily consumption of collective culinary herbs and spices range from 0.5g per person per day to 4g per person per day across four continents (Lampe, 2003). The study also discovered that in India, turmeric consumption alone by individuals was estimated at 1.5g per day. Culinary herbs and spices are also a huge part of Ethiopian dishes, where their medicinal purposes are part of the reason for their inclusion in the local diet (Opara *et al.*, 2014). Other studies iterated that the fiber and protein content of some culinary herbs and spices are comparable to some whole grains (Lampe, 2003; Kurian, 2012).

2.5 Food safety and risk factors of culinary herbs and spices

The European Spice Association (ESA) reports that another area which may give cause to pose food safety and risk questions is the harvesting, storage and processing practices which may be employed after cultivation (ESA, 2013). This is because apart from the possibility of inadvertent contamination from soil which may have metallic or magnetic components easily absorbed by the plants (ESA, 2013) and water or waste water use compounded by overuse of pesticides which definitely leave residues (Singh *et al.*, 2010), adulteration to enhance the aesthetic form of harvested herbs and spices (Cowell *et al.*, 2017) as well as economically motivated adulteration along the value chain can also introduce contaminants into culinary herbs and spices. The metal surfaces that the harvested roots, leaves and bark come in contact with during storage and processing is also another avenue through which packaged culinary herbs and spices on the supermarket shelf may become contaminated with heavy metals (ESA, 2013).

Culinary herbs and spices, like most foods cultivated for consumption, can pose safety and risk questions to demographic groups of interest like children and the aged. This is because they may be cultivated in environments where anthropogenic activities may have led to pollution of the soil or water being used to irrigate farm beds (Leblanc *et al.*, 2000; Roy *et al.*, 2016; Slavík *et al.*, 2012).

A recent US public health paper reported that lead chromate adulteration of turmeric imported from India and Bangladesh is of increased health concern to consumers in the USA

(Cowell *et al.*, 2017). Lead chromate has a vibrant yellow color and because of this, many farmers and retailers apply it to impart a deeper color to the turmeric roots to feign a look of freshness and wholeness even when it has been in stock over a lengthy period (Addady, 2016; Cowell *et al.*, 2017). It is evident that many of these retailers and farmers are ignorant of the damaging side effects of their actions.

Other forms of adulteration of culinary herbs and spices that has been reported include ground pawpaw seeds added to black pepper and corn flour added to ground red pepper to either increase the volume/weight or enhance their appearance (Dhanya *et al.*, 2010). The adulterants mixed into culinary herbs and spices have a varied range of health and safety risk they pose to consumers. Some are a mere nuisance, causing a change in the taste and or consistency of prepared meals. Other adulterants pose more hazardous risks to the health of the consumer ranging from mild stomach upset to severe physiological reactions.

The prevailing storage options for whole culinary herbs and spices available in local markets in sub-Saharan Africa and specifically Ghana, where the spices are kept and displayed in open top pans or bowls creates an avenue for environment pollution with aerial pollutants like lead (Pb) as well as microorganism contamination (Al-Eed *et al.*, 2002; Gaya *et al.*, 2016). A study conducted on culinary herbs and spices on sale in a local market in Ghana isolated 8 genera of fungal species, some of which were potential hazards (Ahene *et al.*, 2011).

Culinary herbs and spices are often times not used in quantities that will elicit adverse reactions immediately on ingestion (Lampe, 2003) even if they are contaminated –this does not hold true in cases where the contaminant is present in large quantities or is a very potent substance (Baldwin *et al.*, 1999). Heavy metal contaminants that may be found in trace or minute quantities in food or water meant for ingestion, will rather bio-accumulate in the body upon intake and wreak havoc later (Eneji *et al.*, 2011; Needleman, 2004).

In the same vein, culinary herbs and spices are ingested daily and even if it is in small quantities, the chronic daily intake of any toxic contaminants found in these otherwise healthy culinary herbs and spices makes determining their safety or risk a worthwhile study.

The mechanisms of how bioaccumulation of toxic heavy metals occurs and affect plants (Smical *et al.*, 2008), invertebrates, fish (Eneji and Annune, 2011; Smical *et al.*, 2008), and mammals (Midzi, 2012) have been studied alongside the clinical cases detected in humans. In humans, heavy metal bioaccumulation and biomagnifications generally occurs in the liver, kidneys, and lungs due to the excretory functions of these organs and the affinity of the heavy metals to localized enzymes (Baldwin *et al.*, 1999; ECHA and IARC, 2012; Gaya *et al.*, 2016; Needleman, 2004).

2.5.1 Physiological effects of ingesting heavy metal contaminants

The clinical effects of consuming cadmium (Cd) and lead (Pb) have been documented in detail. They include cardiovascular failures, kidney malfunctions, teratogenesis, mutations, breaking of DNA strands, physical and mental retardation of child development (Baldwin *et al.*, 1999) and a myriad other diseases in adults including atrophy of bones. Atrophy of bones is attributed to lead, when it actively competes with calcium (Ca), in the presence of protein deficient diets, for binding to enzymes during bone synthesis (Needleman, 2004).

Cadmium and its compounds have been proven through laboratory experiments, using rats, to cause tumors at different tissue sites when administered through several exposure routes (IARC, 2011). Tests carried out by the European Chemical Association (ECHA) observed increased incidence of chromosomal aberrations in the lymphocytes of workers occupationally exposed to cadmium (ECHA and IARC, 2012).

Lead (Pb) is considered highly toxic and its level in food and water is strictly regulated by many governments as well as by the WHO through its FAO arm together with the Codex Alimentarius Commission (CAC). Lead commands a lot of scrutiny due its proven detrimental effects on child development, as well as its disease causing propensity in adults (Baldwin *et al.*, 1999; Gaya *et al.*, 2016; Needleman, 2004; Nkansah *et al.*, 2010; Ogunseitan *et al.*, 2007; WHO, 2011). It is highly dangerous to children since it impairs their physiological and psychological development (Needleman, 2004). Lead levels in blood is also implicated in several disease categories such as its effect on hypertension, anemia, neuropathy, congenital anomalies and dental caries (Ogunseitan and Smith, 2007).

Prior to 1980, the risk lead posed to the intellectual growth of children was underestimated and was therefore set at 40 µg/dL. It was later adjusted downward to 10 µg/dL as more research was carried out to expose the chronic effects of lead (Dudka *et al.*, 1996) and much later adjusted further down to 8 µg/dL (CAC, 2013). Lead uptake from the gut is especially easy when calcium deficient diets are consumed (Baldwin *et al.*, 1999).

Chromium (Cr) in its trivalent form Cr (III) is currently considered essential to some human metabolic processes (CRN, 2013). The Council for Responsible Nutrition, USA, records a study on Cr that found that 1000 µg per day of Cr in the form of chromium picolinate decreased symptoms of type II diabetes in patients (CRN, 2013). Chromium in Cr (VI) form has more oxidizing power and is carcinogenic (IARC & WHO, 2017). Human contact with Cr (VI) is more prone through inhalation in the stainless steel industry (CRN, 2013). The CRN notes that study on the safety of Cr is still inconclusive but the available data supports the idea that Cr is safe for consumption, is not a potent toxicant, and has a wide margin of safety (CRN, 2013).

2.5.2 Regulation of heavy metal contamination

Extensive tests are routinely carried out to conclusively determine the toxicities of many heavy metals by the FAO and the WHO through their joint food safety programs under the auspices of the Codex Alimentarius Commission (CAC). Subsequent to the dose response analyses of these substances, permissible tolerable daily, and weekly and monthly ingestion (PTDI, PTWI and PTMI) quantities have been set by the CAC to advice, guide and standardize their presence in food and water (CAC, 2013).

The toxicity and carcinogenicity of cadmium was determined rather early in the clinical investigation into heavy metal ingestion and their classification done accordingly. The roles chromium and lead played however in disrupting metabolic processes and its cancer causing ability have been quite elusive, making further research on them through the years a priority for the IARC as evidenced by their periodic review of its classification (IARC, 2012; IARC/WHO, 2017).

Cadmium (Cd) a soft bluish white metal occurs as a minor component in zinc ores and exists in nature at an average concentration of 0.1 to 05ppm. Cadmium compounds were industrially used as color pigments to color glass and stabilize plastics until being listed as toxic (ECHA and IARC, 2012). Based on estimated daily intake through food and water, the CAC in 2013 established a PTMI of 25 µg/kg bw translating to approximately 0.8 µg/kg bw per day. The IARC in 2011 estimated that average cadmium levels in the U.S. food supply are between 2 –40 ppb with the average adult consuming approximately 30 µg of cadmium daily (CRN, 2013).

Chromium (Cr) occurs in nature as a metal. It is used commercially in combination with steel to produce stainless steel because it is highly resistant to corrosion. Cr can exist in trivalent (III) and hexavalent (VI) ionic states. The Council for Responsible Nutrition of the U.S.A. reports that Cr metal and Cr(III) are not considered toxic but Cr (VI) is toxic and carcinogenic

(CRN, 2013). Cr (III) or trivalent Cr is considered an essential nutrient in human, used in trace amounts in insulin and lipid metabolism (CRN, 2013). The EFSA in 2010 evaluated the safety of Cr III as an added nutrient to food intended for use by the general population with focus on the potential genotoxicity of Cr III (CRN, 2013). This study was conducted on the back of previous studies carried out by WHO in 1996 and the Institute of Medicine, USA in 2001 (CRN, 2013). In 2003, the Expert Group on Vitamins and Minerals in also run a similar study (CRN, 2013). A study indicated that accumulation of chromium in edible plants may prove hazardous to animals (Gaya and Ikechukwu, 2016). However, the results of an EFSA study in 2010 were conflicting; some evidence indicated that certain Cr III compounds, at high concentrations, may cause chromosomal damage in vitro. In the same report however, the EFSA states that Cr III compounds do not show genotoxicity in vivo, neither did long term studies where mice and rats were fed various Cr III compounds yield evidence of carcinogenicity (CRN, 2013). The updated IRAC classification of carcinogenic substances, last update 27th October 2017, lists Cr III compounds as a group 3 carcinogen (IARC, 2012) and Cr VI as a group 1 carcinogen. However, because Cr VI is not produced in dietary form by any biological system, it does not pose any risk through the dietary system(CRN, 2013). Due to this accepted view and the inconclusive studies on Cr III as a possible carcinogen, the acceptable daily intake (ADI) of Cr III is within a wide safety margin of between 25 µg/kg day^{-1} (for young women, 35 µg/kg/day for young men) to 250 µg/kg day^{-1} based on work carried out by the various health organizations and the EFSA (2010) (CRN, 2013).

Lead (Pb) occurs in nature at between 13 to 20 ppm, mostly as oxides and sulphides (Baldwin *et al.*, 1999). Lead poisoning is a focal point in food safety and risk management because of the ease with which it can be brought into contact with edible food and drink. This is due to its wide distribution in ecosystems stemming from human activities. Lead is a by-product of industrialization. Also for a good number of years in the recent past, it was used as the main component of several widely used chemicals such as leaded gasoline, paints for buildings as well as paint for coating children's toys (Ogunseitan *et al.*, 2007), which increased its contact rate alarmingly.

In 2011, the joint FAO and WHO standards program proposed a withdrawal of the provisional tolerable weekly intake (PTWI) of 25 µg/kg bw of lead which was in use as a standard. This translated into an allowable daily intake (ADI) of approximately 3.6 µg/kg bw. The withdrawal was informed by further study which indicated that even at that level, children could lose 3IQ points (FAO/WHO, 2011). There is now a zero tolerance for lead in especially foods meant to be consumed by children because they are more susceptible to its immediate effects when ingested in either acute or chronic quantities (Needleman, 2004).

Whereas the WHO easily classified the carcinogenicity of cadmium as a Group 1 carcinogen (2012) based on sufficient evidence from studies in laboratory animals and humans (ECHA and IARC, 2012), the classifications for Cr and Pb have been reviewed over time as more studies are conducted to gauge their effects through dose-response analysis (IARC, 2017).

In 1990, Cr III was classified as a Group 3 carcinogen whilst Cr IV was classified a Group 1 carcinogen (IARC, 2012). Lead was first classified under Group 2B in 1987. Then in 2006 it was elevated to Group 2A and briefly downgraded to Group 3 in the same year (IARC, 2012) until 2017 when it was classified as a Group 1 carcinogen (IARC, 2017).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Materials

3.1.1 Sampling of materials

Using a single step simple probability sampling, questionnaires were distributed to some thirty (30) households and ten (10) restaurants in Madina, Accra to collect data on their use of culinary herbs and spices. Respondents, assisted by the researcher, were asked to choose their preferred and most frequently used culinary herbs and spices in no specific order from an extensive list. The list included many of the culinary herbs and spices like; dawadawa, nutmeg, anise, curry powder, oregano, coriander, cinnamon, five star and calabash nutmeg available on the Ghanaian market.

The data obtained was used to determine the frequency of usage of named culinary herbs and spices. Subsequently, ten (10) common and most used culinary herbs and spices were selected for investigation. Culinary herb and spice samples were collected from four (4) local markets; Madina, Malata, Makola and Suame. They were classified according to their English, local and scientific nomenclature and whether they are imported or grown locally (Table 3.1). Reagents for digesting the samples, HNO_3 (nitric acid) and HCl (hydrochloric acid) and standards were obtained from Sigma-Aldrich.

3.1.2 Sample preparation

Dried whole culinary herbs and spices were obtained from the local markets, cleaned by washing with distilled water and sundried for 12 hours to remove all moisture. The whole samples were pounded in a mortar to obtain slightly coarse powders except the turmeric and nutmeg which were grated with a stainless steel grater to obtain fine powders. Different brands of curry powder, which is sold already processed, were also sampled. The powders were kept dry and sealed for analysis.

Table 3.1: Classification of culinary herb and spice samples under investigation

English name	Local name	Scientific name	Origin unto market
African locust beans	Dawadawa	*Parkia biglobosa*	Grown locally
Anise	Nketenkete	*Pimpinella anisum*	Imported
Cloves	Pɛprɛ	*Syzygium aromaticum*	Grown locally
Calabash nutmeg	Wedeaba	*Monodora myristica*	Grown locally
Curry powder	-	None (A mixture of spices; curry leaf, coriander, cumin, turmeric, etc)	Imported
Fennel	Ɛmoo aba	*Foeniculum vulgare*	Imported
Negro pepper	Hwentia	*Xylopia aethiopica*	Grown locally
Nutmeg	-	*Myristica fragrans*	Imported
Rosemary	-	*Rosmarinus officinalis*	Imported
Turmeric	-	*Curcuma longa*	Imported

3.1.3 Wet Digestion of Samples

To carry out a determination of the heavy metal concentrations, a wet digestion of the dried samples was undertaken (Bader, 2011). The wet digestion was carried out with a concentrated HNO_3 and HCl mixture in a 3:1 ratio. 0.5 g each of the dried samples was weighed into test tubes and to each was added a 12 ml of the acid mixture.

Using a thermostat controlled heating block, the content of the test tube was heated to 100 ºC. The temperature was gradually increased to 200 ºC and this temperature maintained for an hour. The test tubes were allowed to gradually cool. A 1 ml of HCl was added to each digestion mixture and the contents were retuned into the oven for heating. The digestion process of heating, cooling and addition of 1 ml of HCl, was repeated until a clear solution was obtained.

The clear solution was transferred into a 50 ml volumetric flask and double distilled deionized water was added on to top the solution up to the mark.

3.2 Heavy metal determination with AAS

The heavy metal determination was performed with a Perkin-Elmer model novAA 400p Atomic Absorption Spectrometer (AAS) with a single beam and deuterium background correction. A standard solution containing known concentrations of the heavy metals under investigation together with a blank solution was used to calibrate the equipment using a linear calibration through zero. A single beam hollow cathode lamp of cadmium, chromium, and lead were each used at specific wavelengths to detect each metal.

The AAS was run on triplicate digested clear solutions for each of the sampled culinary herbs and spices. The linearity (R^2) of the standards run ranged from 0.9897 to 0.9983. Recovery for the metals detected ranged from 98% to 101%. The limit of detected (LOD) was pegged at 0.01 ppm/mL, and the measuring time was 3 seconds, standard for accurate determination.

3.3 Data Analysis

The concentrations of the heavy metals detected in the samples analyzed were extrapolated from calibration curves presented in mg/L and converted to mg/kg of dry weight using Equation 3.1.

$$\frac{\text{Volume of final solution from digestion}}{\text{Weight of sample taken for digestion}} \qquad \text{Eq. 3.1}$$

The descriptive statistical parameters; mean and standard deviation values, were calculated using MS Excel computer program.

A risk assessment for safety was done based on estimating the daily dietary intake of the detected concentrations of heavy metals in the contaminated culinary herbs and spice and comparing the results to the acceptable daily intake (ADI) levels or reference dose R_fD set by the WHO. The estimated dietary intake (EDI) was calculated using equation 2.4 (page 11). The average dry weight of culinary herbs and spices consumed daily (AC) was taken as 10 g per day per average body weight of 60 kg (Lampe, 2003; Nkansah and Amoako, 2010; Siruguri and Bhat, 2015).

A point risk assessment for ingesting the culinary herbs and spices contaminated with lead was calculated using Eq. 2.3 (page 11).

The chronic daily intake (CDI) is the same as the EDI and the potency factor (PF) also called the slope factor was obtained from the Agency for toxic substances and disease registry (ATSDR) of the USA (ATSDR, 2007).

Slope factor for dietary intake of lead (ATSDR, 2007);

Infant and toddlers (breast-fed and formula-fed) – 0.24 µg/dL (2.4×10^{-4} mg/kg)

Adult females (duplicate diet study) – 0.034 µg/dL (3.4×10^{-5} mg/kg)

Adult males (experimental study) – 0.027 µg/dL (2.7×10^{-5} mg/kg)

CHAPTER FOUR

RESULTS AND DISCUSSION

The mean concentrations of cadmium (Cd), lead (Pb) and Chromium (Cr) and the range of their concentrations present in the sampled culinary herbs and spices are recorded in Table 4.1.

Table 4.1 Concentrations of heavy metals in sampled culinary herbs and spices

	Conc. mg/kg				
	[Cd]	**[Pb]**		**[Cr]**	
Spice Group		**Mean ±SD**	**Range**	**Mean ±SD**	**Range**
Nutmeg	<DL	<DL		0.13 ±0.04	0.11 - 0.15
Turmeric	<DL	0.850 ±0.08	0.76 -0.94	0.42 ±0.03	0.39 - 0.45
Calabash nutmeg	<DL	<DL		<DL	
Fennel	<DL	6.525 ± 0.06	6.05 - 7.02	<DL	
Cloves	<DL	5.503 ±0.05	0.11 - 10.90	<DL	
Rosemary	<DL	4.557 ±0.03	0.11 - 6.08	<DL	
Negro pepper	<DL	<DL		<DL	
African locust beans	<DL	<DL		<DL	
Curry powder	<DL	<DL		<DL	
Anise	<DL	8.950±0.05	8.90 - 9.00	<DL	

Key: <DL: Below the AAS detection limit of 0.01 mg/L

Culinary herbs and spices are classified as condiments and thus, the levels of heavy metals were compared with the acceptable safety standards also known as the maximum permissible limits (MPL) for food condiments with dry weight content above 50% established by FAO/WHO. These standards are: 0.1 mg/kg for Cd, 0.1 mg/kg for Pb, and 10 mg/kg for Cr (CAC, 2013; CRN, 2013).

The highest mean value was recorded for detected concentrations of lead; 8.95 ±0.05 mg/kg. The concentration for chromium detected in the samples ranged from 0.11 to 0.45 mg/kg. No cadmium concentrations were detected in the sampled herbs and spices.

Some of the samples did not have detectable levels of heavy metal and hence can be presumed to have met the FAO/WHO acceptable standards. Five of the sampled culinary herbs and spices; anise, cloves, fennel, rosemary and turmeric, had elevated levels of lead. Anise recorded the highest lead concentration of 8.950±0.05 mg/kg, followed by fennel with 6.525±0.06 mg/kg whilst turmeric recorded the lowest lead concentration (0.85±0.08 mg/kg). None of the samples analyzed contained detectable concentrations of chromium except nutmeg and turmeric which recorded mean concentration levels of 0.13±0.04 mg/kg and 0.42±0.03 mg/kg respectively. These mean values fall safely below the MPL for Cr in foods.

Lead is a divalent cat-ion which binds strongly to sulfhydryl groups on proteins, distorting enzymes and structural proteins (Needleman, 2004). It is the most recognized toxic environmental metal pollutant because of its contact rate (Krejpcio *et al.*, 2007). Lead (Pb) primarily affects the central nervous system (CNS) amongst other organs in the body and is implicated in reproductive, renal, nervous, cardiovascular and developmental malfunctions (Needleman, 2004). The FOA/WHO permissible limit for lead in food additives was 0.2 mg/kg (CAC, 2013). Since lead was elevated to a Group 1 carcinogen (IARC, 2017), the FOA/WHO have met and are reviewing the permissible limits of lead in foods (FAO *et al.*, 2017). The current consensus is to withdraw the existing permissible tolerable weekly intake (PTWI) of 25 µg/kg bw for adults (FAO *et al*., 2017). There is a zero tolerance for carcinogens in foods. The mean lead concentrations detected in anise (8.95±0.05 mg/kg), cloves (5.50±0.05 mg/kg), fennel (6.52±0.06 mg/kg), rosemary (4.55±0.06 mg/kg) and turmeric (0.85 ±0.08) far exceed the current WHO standards.

Cadmium and its compounds are classified as human carcinogens (IARC, 2011). Increasing its content in food will be harmful to human health. WHO set a permissible monthly intake of cadmium (Cd) at 25 µg/kg (CAC, 2013). No detectable concentrations of Cd were found in all the sampled herbs and spices.

Humans are primarily exposed to chromium through food (CRN, 2013). Studies have shown that Cr (III) is essential for some human metabolic processes but Cr (VI) which has more oxidative power can be harmful (Gaya and Ikechukwu, 2016). Cr III has a wide safety margin and thus the reference dose (R_fD) and permissible limits in foods is high. The EFSA R_fD for chromium is 250 µg/kg day$^-$ and the WHO MPL is 0.10 mg/kg. The mean chromium levels detected in the sampled herbs and spices varied from 0.13 mg/kg in nutmeg to 0.42 mg/kg in turmeric.

A study in Poland investigated concentrations of lead, cadmium, copper and zinc in selected herbs and spices and found varied elevated lead (0.25 mg/kg in cloves and 1.4 mg/kg in cinnamon) and cadmium (0.14 mg/kg in cinnamon) levels, (Krejpcio *et al*., 2007). A study conducted on selected spices in Nigeria also recorded high mean values of lead in spices; 3.80 mg/kg in cloves, 4.71 mg/kg in calabash nutmeg and 3.80 mg/kg in nutmeg (Gaya and Ikechukwu, 2016). These levels are rather alarming especially in light of the current review of acceptable lead levels in consumables (FAO *et al.*, 2017), while a study conducted in Ghana found levels of lead (0.17 -0.19 mg/kg) higher than the WHO accepted limit (0.10 mg/kg), present in some culinary herbs and spices (Nkansah and Amoako, 2010). Very few studies have been carried out to determine the levels of chromium in herbs and spices due to the assurance of its wide margin of safety. Sampled culinary herbs and spices in Nigeria recorded mean chromium values as high as 2.83 mg/kg in cloves and 3.76 mg/kg in nutmeg (Gaya and Ikechukwu, 2016).

Turmeric recorded chromium levels ranging from 0.39 mg/kg to 0.45 mg/kg. The mean lead level in turmeric was 0.85 mg/kg. This indicates that the phenomenon noticed in the USA about contamination of turmeric with lead through the application of lead chromate to turmeric rhizomes, may not be localized only to that market (Cowell *et al.*, 2017).

The increased levels of Pb in anise, cloves, fennel and rosemary; and chromium in nutmeg and turmeric indicate that those plants are perhaps more prone to accumulate these elements from the environment (Krejpcio *et al.*, 2007).

Researchers in various jurisdictions investigating heavy metal concentrations in herbs and spices affirm that the heavy metal content of sampled culinary herbs and spices vary depending on environmental pollution levels, storage, processing, technology used in processing country of origin and plant part which is used (Al-Eed *et al.*, 2002; ESA, 2013;
Gaya and Ikechukwu, 2016; Krejpcio *et al.*, 2007; Nkansah and Amoako, 2010; Opara and Chohan, 2014).

The estimated daily dietary intake of the sampled culinary herbs and spices containing the mean quantified concentrations of heavy metals are presented below in Table 4.2.

Table 4.2 Estimated dietary intake (EDI) of the detected heavy metals (mg/kg day^{-1})

Spice Group	Pb	Cr
Nutmeg		0.022
Turmeric	0.142	0.070
Fennel	1.088	
Cloves	0.917	
Rosemary	0.760	
Anise	1.492	

When referenced to the WHO permissible tolerable daily intake (PTDI) of lead, 8 µg/kg (0.008 mg/kg) (CAC, 2013), a figure currently under review, the EDI of the herbs and spices which showed traces of lead (Pb) are intolerable and above the standard. In contrast, the EDI for Cr falls below the daily MPL of 250 µg/kg or 0.25 mg/kg (CRN, 2013).

Currently, lead (Pb) is classified as a Group 1 carcinogen. A point risk assessment for ingesting the culinary herbs and spices contaminated with lead can be calculated using Eq. 2.3 (page 11)

For example, the risk of consuming cloves contaminated with 0.917 mg/kg (EDI/CDI) of lead (Pb) can be calculated for adult males and females, using the dietary slope factor for these groups obtained from the Agency for Toxic Substances and Disease Registry (ATSDR).

The point risk involved in ingesting 10 mg/kg of cloves contaminated with 0.917 mg/kg of lead for an adult female (PF 3.4×10^{-5} mg/kg) will be 3.118×10^{-5}, while that for an adult male (PF 2.7×10^{-5} mg/kg) will be 2.475×10^{-5}. The calculated risk is greater than one (1) hence the risk of the individual reacting adversely to the health hazard is implied.

CHAPTER FIVE

CONCLUSION AND RECOMMENDATION

5.1 Conclusion

No concentration of cadmium was detected in the sampled culinary herbs and spices. The limit of detection of the atomic absorption spectroscopy may have hampered the detection of concentrations of cadmium.

Chromium concentrations detected in the sampled culinary herbs and spices were low; only two of the analyzed samples, nutmeg and turmeric, recorded mean levels of chromium. The recorded mean concentrations fall below the maximum permissible limit (MPL) of 10 mg/kg. The estimated dietary intake of chromium concentrations in the sampled nutmeg and turmeric pose no significant health risk because it falls below allowable daily intake or reference dose (R_fD) of 0.25 mg/kg.

Mean lead concentrations detected in five (5) of the sampled culinary herbs and spices were relatively high and exceeded the current maximum permissible limit set by the FAO/WHO Joint committee; 0.10 mg/kg. The five sampled culinary herbs and spices with elevated lead levels pose significant risks as food hazards. The estimated dietary intake of lead concentrations in the sampled anise, cloves, fennel, rosemary and turmeric pose a significant health risk because they exceed the current allowable daily intake of 0.008 mg/kg. The health risk is of high concern since lead is classified as a carcinogen.

5.2 Recommendation

Food safety regulatory bodies must recognize culinary herbs and spices as potential vehicles for introducing heavy metal contamination into a diet and place them under constant and continuous monitoring like any other food products.

REFERENCES

Addady, M. (2016, August). Turmeric recall expanded after confirming presence of Lead. Fortune Magazine, Chicago, New York, p. 16.

ATSDR, Agency for Toxic Substances and Diseases Registry (2007). ATSDR minimal risk levels and worksheets, Appendices, Vol. 13 a. https://doi.org/https://www.atsdr.cdc.gov/toxprofiles/tp13-a.pd

Ahene, R.E., Odamtten, G.T. and Owusu, E. (2011). Fungal and bacterial contaminants of six spices and spice products in Ghana. African *Journal of Environmental Science and Technology*, Vol. 5, Issue 9, pp. 633–640.

Al-Eed, M., Assubaie, F., El-Garawany, M., El-Hamshary, H. and ElTayeb, Z. (2002). Determination of heavy metal levels in common spices. *Journal of Applied Science*, Vol. 17, p. 87. Retrieved from http://www.kfu.edu.sa/en/Deans/Research/Documents/1026.pdf

Bader, N. R. (2011). Sample preparation for flame atomic absorption spectroscopy: An overview. *Rasayan Journal of Chemistry*, Vol. 4, Issue 1, pp. 49–55. https://doi.org/10.1016/0584-8547(96)01507-8

Baldwin, D. R. and Marshall, W. J. (1999). Heavy metal poisoning and its laboratory investigation. *Annals of Clinical Biochemistry*, Vol. 36, Part 3, pp. 267–300. https://doi.org/10.1177/000456329903600301

CAC, Codex Alimentarius Commission. (2013). Joint FAO/WHO Food Standards Programme and Codex Alimentarius Commission. Report on the 17th session of the codex committee on food additives. (No. ALINORM 85/12). Roma.

CRN, Council for Responsible Nutrition. (2013). Chromium. Vitamin and Mineral Safety, Vol. 1, 3rd Edition.

Cowell, W., Ireland, T., Vorhees, D. and Heiger-Bernays, W. (2017). Ground turmeric as a source of lead exposure in the United States. Public Health Reports (Washington, D.C. : 1974), Vol. 132, Issue 3, pp. 1–5. https://doi.org/10.1177/0033354917700109

Dhanya, K. and Sasikumar, B. (2010). Molecular marker based adulteration detection in traded food and agricultural commodities of plant origin with special reference to spices. *Current Trends in Biotechnology and Pharmacy*, Vol. 4, Issue 1, pp. 454–489.

Dudka, S., Piotrowska, M. and Terelak, H. (1996). Transfer of cadmium, lead, and zinc from industrially contaminated soil to crop plants: a field study. *Environmental Pollution* (Barking, Essex : 1987), Vol. 94, Issue 2, pp. 181–188. https://doi.org/10.1016/S0269-

7491(96)00069-3

Duffus, J. H. (2002). “Heavy metals ” – A meaningless term? *Pure and Applied Chemistry*, Vol. 74, Issue 5, pp. 793–807. https://doi.org/10.1007/s001340101055

Eneji, I. S. and Annune, P. A. (2011). Bioaccumulation of heavy metals in fish (Tilapia Zilli and Clarias Gariepinus) organs from River Benue , North – Central Nigeria. Pakistan *Journal of Analytical Environmental Chemistry*, Vol. 12, Issue 1 & 2, pp. 1–8.

ECHA, European Chemical Association, and International Agency for Research on Cancer (IARC). (2012). Cadmium and Cadmium compounds. European Chemicals Agency (Vol. 1993). https://doi.org/10.1002/14356007.a04

ESA, European Spice Association. (2013). Metallic and magnetic contaminations in herbs and spices – Risk evaluation. European Spice Association, October edition, pp. 1–9.

FAO/WHO, Food and agricultural organization of the World Health Organization. (2011). JOINT FAO/WHO Food standards programme and Codex Committee on contaminants in foods. 5th Session. The Hague , The Netherlands. March 2011. Codex Alimentarius Commision (Vol. CF/5 INF/1).

FAO, WHO, and Codex Alimentarius Commission C.A.C. (2017). Joint FAO/WHO Food standards programme and Codex Alimentarius Commission, 40th session. Report of the 11th session of the codex committee on contaminats in food.

Gaya, U. I. and Ikechukwu, S. A. (2016). Heavy metal contamination of selected spices obtained from Nigeria. *Journal of Applied Sciences and Environmental Management*, Vol. 20, Issue 3, pp. 681–688.

Haas, C.N, Rose, J.B. and Gerba, C. P. (1999). Quantitative Microbial Risk Assessment. New York: John Wiley & Sons.

Halvorsen, B. L., Carlsen, M. H., Phillips, K. M., Bøhn, S. K., Holte, K., Jacobs, D. R. and Blomhoff, R. (2006). Content of redox-active compounds (i.e. antioxidants) in foods consumed in the United States. *American Journal of Clinical Nutrition*, Vol. 84, Issue 1, pp. 95–135.

IARC, International Agency for Research on Cancer. (2004). Arsenic and Arsenic Compounds (Vol. 2002).

IARC, International Agency for Research on Cancer. (2011). Cadmium and Cadmium compounds. IARC Monographs on the Evaluation of Carcinogenic Risks to Humans (Vol. 100 C). https://doi.org/10.1002/14356007.a04

IARC, International Agency for Research on Cancer. (2012). Agents Classified by the IARC Monographs , Volumes 1 – 104. IARC Monographs (Vol. 7). https://doi.org/10.1007/s13398-014-0173-7.2

IARC, International Agency for Research on Cancer. (2017). Monographs on the evaluation of carcinogenic risks to humans.

IARC, International Agency for Research on Cancer, and World Health Organization. (2017). IARC Monographs on the Evaluation of Carcinogenic Risks to Humans. Volume 1-120. Retrieved from http://monographs.iarc.fr/ENG/Classification/

Krejpcio, Z., Król, E. and Sionkowski, S. (2007). Evaluation of heavy metals contents in spices and herbs available on the Polish market. *Polish Journal of Environmental Studies*, Vol. 16, Issue 1, pp. 97–100.

Kurian, A. (2012). Health benefits of herbs and spices. Cambridge, UK; Woodhead Publishing Limited. pp. 72-88 https://doi.org/10.1533/9780857095688.72

Lampe, J. W. (2003). Spicing up a vegetarian diet : chemopreventive effects of phytochemicals. *American Journal of Clinical Nutrition*, Vol. 78, pp. 579–583.

Leblanc, J., Malmauret, L., Guérin, T. and Bordet, F. (2000). Food additives and contaminants estimation of the dietary intake of pesticide residues , lead , cadmium , arsenic and radionuclides in France. *Food Additives and Contaminants*, Vol. 17, Issue 11, pp. 925–932. https://doi.org/10.1080/026520300750038108

Midzi, E. M. (2012). Cadmium and lead concentrations in livers and kidneys of cattle slaughtered at Grootfontein Abattoir in Namibia.

Needleman, H. (2004). Lead Poisoning. *Annual Review of Medicine*, Vol. 55, Issue 1, pp. 209–222. https://doi.org/10.1146/annurev.med.55.091902.103653

Needleman, H. L. (2013). History of Lead poisoning in the world. Climate Change 2013 - *The Physical Science Basis*, Vol. 53, Issue 9, pp. 1–30. https://doi.org/10.1017/CBO9781107415324.004

Nkansah, M. A. and Amoako, C. O. (2010). Heavy metal content of some common spices available in markets in the Kumasi metropolis of Ghana. *American Journal of Scientific and Industrial Research*, Vol. 1, Issue 2, pp. 158–163. https://doi.org/10.5251/ajsir.2010.1.2.158.163

Ogunseitan, O. A. and Smith, T. R. (2007). The cost of environmental lead (Pb) poisoning in Nigeria. *African Journal of Environmental Science and Technology*, Vol. 1, Issue 2, pp. 27–36.

Opara, E. I. and Chohan, M. (2014). Culinary herbs and spices : Their bioactive properties , the contribution of polyphenols and the challenges in deducing their true health benefits. *International Journal of Molecular Sciences*, Vol. 15, pp. 19183–19202. https://doi.org/10.3390/ijms151019183

Roy, S. and Gupta, S. (2016). Effect of wastewater irrigation on soil and some selected vegetables grown in Asansol , West Bengal. *International Journal of Environmental Sciences*, Vol. 6, Issue 5, pp. 894–904. https://doi.org/10.6088/ijes.6084

Singh, A. and Agrawal, M. (2010). Effects of municipal waste water irrigation on availability of heavy metals and morpho-physiological characteristics of Beta vulgaris L . *Journal of Environmental Biology*, Vol. 31, September edition, pp. 727–736.

Siruguri, V. and Bhat, R. V. (2015). Assessing intake of spices by pattern of spice use, frequency of consumption and portion size of spices consumed from routinely prepared dishes in southern India. *Nutrition Journal*, Vol. 14, Issue 1, pp. 1–9. https://doi.org/10.1186/1475-2891-14-7

Slavík, R., Julinová, M. and Labudíková, M. (2012). Screening of the spatial distribution of risk metals in topsoil from an industrial complex. *Ecological Chemical Engineering Science*, Vol. 19, Issue 2, pp. 259–272. https://doi.org/10.2478/v10216-011-0020-0

Smical, A.-I., Hotea, V., Oros, V., Juhasz, J. and Pop, E. (2008). Studies on transfer and bioaccumulation of heavy metals from soil into lettuce. *Environmental Engineering and Management Journal*, Vol. 7, Issue 6, pp. 609–615.

Tapsell, L. C., Hemphill, I., Cobiac, L., Patch, C. S., Sullivan, D. R., Fenech, M., Patch C.S., Roodenrys, S., Keogh, J.B., Clifton, P.M., Williams, P.G., Fazio, V.A. and Inge, K. E. (2006). Health benefits of herbs and spices: the past, the present, the future. *Medical Journal of Australia*, Vol. 185, Suppl 4, S4-24. Retrieved from http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=17022438

The Perkin-Elmer Corporation, (1996). Analytical methods for atomic absorption spectroscopy: a manual. Perkin-Elmer Corporation, USA. pp 3-9, 38.

WHO, World health organization. (2011). Lead in drinking water.Vol. WHO/SDE/WS.

Yidana, S. M., Sakyi, P. A. and Stamp, G. (2011). Analysis of the suitability of surface water for irrigation purposes : The southwestern and coastal river systems in Ghana. *Journal of Water Resource and Protection*, 2011, October edition, pp. 695–710. https://doi.org/10.4236/jwarp.2011.310080

Zheng, W. and Wang, S. Y. (2001). Antioxidant activity and phenolic compounds in selected herbs. *Journal of Agricultural and Food Chemistry*, Vol. 49, Issues 11, pp. 5165–5170. https://doi.org/10.1021/jf010697n

APPENDIX

EXPOSURE ASSESSMENT QUESTIONNAIRE

QUESTIONS ABOUT CULINARY HERBS AND SPICE USAGE IN HOMES AND EATERIES

INSTRUCTIONS: Kindly supply the appropriate answers in space provide.
PLEASE MARK LIKE THIS: ⓧ

BIODATA							
GENDER	**RELATIONS**	**EDUCATION**	**AGE (Y)**		**WGT (kg)**		
M○	Single ○	Informal ○	0	0	0	0	0
F○	Married ○	Basic ○	1	1	1	1	1
	Divorced ○	JHS ○	2	2	2	2	2
		SHS ○	3	3	3	3	3
		Tertiary ○	4	4	4	4	4
			5	5	5	5	5
			6	6	6	6	6
			7	7	7	7	7
			8	8	8	8	8
			9	9	9	9	9
SCOPE OF COOKING: HOME ○ **CATERING FACILITY** ○							

1. Do you use culinary herbs?
○Yes
○No

2. Select your most used (favorite) herbs or spices from the list below
○Anise (nketenketen)
○Cloves (pepre)
○Curry Powder

○Fennel (emoo aba)
○Rosemary
○Turmeric
○Pre-mixed local spices containing Fennel/Rosemary/Anise/Cloves

3. What number of people do you normally cook for?
○5 and below
○10 and below
○Above 10 but below 20
○Above 20
○Above 100

4. What quantities of your preferred culinary herbs or spices do you use per meal preparation?
○Pinch
○Quarter teaspoon
○Half teaspoon
○Teaspoonful
○Handful
○Tablespoonful
○Other weight or measure: ………………….

5. How often do you use your preferred culinary herbs or spices?
○Rarely
○Once a week
○Twice a week
○Thrice a week
○Daily

6. How long (in years) have you been using this/these culinary herbs and spice?
○0 ○ 0
○1 ○ 1
○2 ○ 2
○3 ○ 3
○4 ○ 4
○5 ○ 5
○6 ○ 6
○7 ○ 7
○8 ○ 8
○9 ○ 9

7. Do you have any safety or health concerns about your preferred culinary herbs or spices?

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