KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY, KUMASI

HEAVY METAL CONTENT OF SOME LOCAL SPICES ON THE GHANAIAN MARKET

BY

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DECLARATION

I hereby declare that this submission is my own work towards the MSc 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 other degree of the University except where due acknowledgment has been made in the text.

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DEDICATION

I dedicate this work to my parents Mr and Mrs John Raja Abban.

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ABSTRCT

A spice is a vegetable substance of indigenous or exotic origin which is or has a hot, pigment taste, used to enhance taste of foods or to add to them the stimulant ingredients contained in them. They are sparingly used but frequently used in Ghanaian meals. Seven heavy metals were determined in some natural spices on the Ghanaian market. Four of the metals namely zinc (Zn), manganese (Mn), copper (Cu) and iron (Fe) are essential micronutrients and lead (Pb), cadmium (Cd) and mercury (Hg) are toxic metals.

A total of eighty three samples comprising of fifteen spices were purchased from seven different markets at Asafo, Takoradi, Kaneshie, Ayigya, Makola and Tema. The spices were processed into the powdered form and subjected to a wet digestion procedure with a mixture of H_2O, HCl, HNO_3, $HClO_4$ and H_2SO_4. Sample solutions were analyzed for Hg using a Mercury Analyzer Model HG-5000 and for the other metals using SOLAAR (S Series 711239 v1.23) Flame Atomic Absorption Spectrometer. The range of total Hg concentration (µg/g) for all the spices from all the markets is ND-0.031 (ND- not detectable), that for Cd also ranged from ND- 1.41. The range for Fe is 65.35-221.15, that for Mn is from 14.93-24.96. Zn ranged from ND -17.70 and finally Cu is 4.22-22.91. Pb was not detected in any of the samples analyzed.

The amount of spice to be consumed in order to exceed the recommended daily intakes for the metals were calculated based on the recommended daily intake for the various metals. The amounts obtained were not unlikely to be consumed considering how sparingly they are used. Correlation analysis showed strong positive and negative correlation between the metals for the spices.

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CHAPTER ONE

1 INTRODUCTION

A spice is a vegetable substance of indigenous or exotic origin which is or has a hot, pigment taste, used to enhance taste of foods or to add to them the stimulant ingredients contained in them. It can also be defined as a dried seed, fruit, root, bark or vegetative substance derived from the non leafy parts of plants. It is used as a food additive for the purpose of flavouring or food flavourings, and sometimes as a preservative by killing or preventing the growth of harmful bacteria (Adamson, 2004).

The use of spices dates back to ancient times. Archaeological excavation has revealed that prehistoric man used the leaves of certain plants to enhance the flavour of the half-cooked foods which he ate (Baillon, 1877).

There are many reasons for which people use spices, though, taste probably tops the list. There are several spices that simply smell good and those smells can be alternately soothing or exciting. Some of the types of spices include Onion, Ginger, Garlic, Cloves, Dawadawa, Cinnamon, Rosemary, Thyme, Marjoram, Pepper, Black Pepper, Olive, Lime, Anise, Nutmeg, Curry, Almond, Turmeric, Oregano, Sage, Lemon balm, Peppermint, Dill, and Coriander. Over the centuries, certain spices have been said to heal every infection, disease and malady known to man. But there is no doubt that spices do have value far beyond enhancing the taste of food. However, besides adding to the taste, spices have multifarious functions that include combating foodborne microorganisms, reducing food poisoning, (Sherman and Billing, 1999) antioxidant function, (Pokorney, 1991) and antimicrobial activity (Shelef, 1983). Spices are also known to possess a wide range of medicinal values. Such values include fight against cancer causing cells, reduction of cholesterol level in the blood and prevention of several skin diseases (Shils, 1999).

Spices are plants and they grow in the soil. Minerals or elements needed for plants growth are also found in the soil and are used by these plants. Some of these elements or metals are of importance while others are not. Pollution of the general environment has increasingly gathered a global interest since the beginning of this century because it is through this process that some of these dangerous elements or metals get into the plant. In this respect, contamination of agricultural soils and of the environment with heavy metals in the last decades has raised public and scientific interest due to their dangerous effects on human health (Gilbert, 1984). Heavy metals may be present in agricultural soils at low levels but can be accumulated by plant over a period. This has led researchers all over the world to study the pollution with heavy metals of air, water, and foods to avoid their harmful effects (Oehme, 1989; Zakrzewski, 1991; Kennish, 1992) and to determine their suitability for human consumption.

People consume spices for several reasons. Some people prefer the processed form which is powdered to the fresh ones. This could be due to storage and convenience. Some surveys conducted have shown that these powdered forms are sometimes adulterated with other substances just to increase the quantity. The presence of some foreign substances such as sand from the environment or method of drying or milling has been reported (FAO/WHO, 2001). These added substances may affect the elemental composition or content by increasing the levels of trace metals if present. Element compositions in a particular plant vary greatly depending upon a number of factors, namely, soil parameters, fertilization management and climate. Also, element compositions vary from section to section within the same plant (Bhattacharjee *et al.*, 1998) but sometimes in the preparation two or more parts of the same plant are combined.

Heavy or trace metals play positive and negative roles in human life (Adriano, 1984; Divrikli *et al.*, 2003). Some of the heavy metals such as iron (Fe), zinc (Zn) and copper (Cu) are considered essential but they can become harmful above certain levels. Some metals like cadmium (Cd), lead (Pb) and mercury (Hg) have toxic roles in biochemical reactions in our body. Zinc is important during puberty, pregnancy, and menopause. If large doses of zinc (10-15 times higher than the recommended daily intake) which is 8-15 mg/day are consumed, stomach cramps, nausea, and vomiting may occur. Ingesting high levels of zinc for several months may cause anaemia, damage the pancreas, and decrease levels of high-density lipoprotein (HDL) cholesterol. Consuming low levels of zinc is at least as important a health problem as consuming too much zinc. Without enough zinc in the diet, people may experience loss of appetite, decreased sense of taste and smell, decreased immune function, slow wound healing, and skin sores. Too little zinc in the diet may also cause poorly developed sex organs and retarded growth in young men. If a pregnant woman does not get enough zinc, her babies may have birth defects (ATSDR, 2005).

Iron is an integral part of many proteins and enzymes that maintain good health. In humans, iron is an essential component of proteins involved in oxygen transport (Dallman 1986; Institute of Medicine, 2001). It is also essential for the regulation of cell growth and differentiation (Bothwell *et al.*, 1979; Andrews, 1999). A deficiency of iron limits oxygen delivery to cells, resulting in fatigue, poor work performance, and decreased immunity. Iron deficiency anaemia is associated with constipation, nausea, vomiting, and diarrhoea (Bhaskaram, 2001; Haas and Brownlie, 2001; Institute of Medicine, 2001). On the other hand, excess amounts of iron can result in increased risk of free radical damage and cancer and even death (Corbett, 1995).

Manganese is an extremely important mineral when stabilizing blood sugar. It has strong estrogenic properties, and as a result is the most important element for treating menopausal symptoms, osteoporosis, and postpartum depression. Inadequate manganese intake has been associated with parenteral nutrition, resulting in dermatitis, changes in hair pigmentation and slowed hair growth. Regular dislocation of joints (particularly knee joints) is associated also with insufficient manganese levels. On the other hand, excessive manganese levels increase the risk for tendon / ligament tears and pneumonia. Also loss of sex drive and sperm damage have also been observed in men. Excess manganese interferes with the absorption of dietary iron (Blaurock-Busch, 1997; ATSDR, 2008a).

Eating food or drinking water with very high cadmium content severely irritates the stomach, leading to vomiting and diarrhoea, hypertension, and sometimes death. Accumulation of lower levels of cadmium over a long period of time can lead to a build-up of cadmium in the kidneys and cause kidney damage (ATSDR, 2008b).

Copper helps the body to use iron. It is also important for nerve function, bone growth, enhanced body use of sugar and protection of cell membranes from destruction by free radicals. A wide range of cardiovascular and blood disorders may be attributed to copper deficiency. Water that contains higher than normal levels of copper may cause nausea, vomiting, stomach cramps, or diarrhoea (ATSDR, 2004).

Lead can cause neurological damage in foetus and young children. Lead exposure may also cause weakness in fingers, wrists, or ankles, small increases in blood pressure and anaemia. At high levels of exposure, lead can severely damage the brain and kidneys in adults or children and ultimately cause death; it also causes miscarriage (ATSDR, 2007).

Mercury causes severe birth defects, abortion and mental retardation. In addition to permanent damage to the brain and kidneys, mercury can damage the stomach and intestines, producing symptoms of nausea, diarrhoea, or severe ulcers. Symptoms included rapid heart rate and increased blood pressure (Hayter, 1980; ATSDR, 1999).

Thus, the benefits of micronutrients may be completely reversed if present at high levels as they may be detrimental to human health. Therefore, the World Health Organisation (WHO) has established levels of metals in foods above which, they should not be consumed. For this reason the levels of trace metals in our food should be of much importance and concern to us. The heavy metal or mineral content of spices is not a well researched area worldwide.

A research conducted in Saudi Arabia on some local spices revealed high levels of some trace metals (Al-Eed *et al*., 2002). The concentration of lead (Pb) ranged from trace to 14.30 mg/kg on dry weight basis, cadmium (Cd) ranged from 1.25 mg/kg to 3.05 mg/kg, cobalt (Co) from 0 to 0.64 mg/kg and that of selenium (Se) 0 to 13.3 mg/kg. Some of these concentrations are above the limit approved by WHO/FAO and this makes it dangerous due to the health implications associated with them (Gilbert, 1984; Satter *et al*., 1989). Other countries have also established daily intake for metals. For example the accepted daily intake in Canada for lead is 19.5 µg, for cadmium is 2.4 µg, for iron is 762 µg, for Cobalt is 0.2 µg and for Zinc is 452.1 µg (Meluzzi *et al*., 1996).

Countries such as Nigeria (Adeyeye *et al.*, 2000), Burkina Faso, Niger (Smith *et al*, 1996) are but a few in Africa who have done some research. Information available in Ghana dealt with some work that investigated the properties of spices as inhibitory agents of mycelial growth of *Aspegillus* species and of its toxin production.

Much information on heavy metal content of spices in Ghana is unavailable.

1.2 OBJECTIVES

The objectives of this study are

1. To determine the heavy metal content of some selected natural spices on the Ghanaian market.
2. To find out whether the elements occur in levels within the WHO limits
3. To determine if any correlation exist between the metals in a spice.

CHAPTER TWO

2 LITERATURE REVIEW

2.1 GENERAL BACKGROUND

Many people are not particularly interested in agriculture but spices arouse a fascination which goes well beyond their economic importance. This special status which spice plants in general enjoy is even more pronounced in the tropical origin. There is something subconsciously exciting about the flavours and aromas of these foodstuffs.

A spice is a dried seed, fruit, root, bark or vegetative substance used in nutritionally insignificant quantities as a food additive for the purpose of flavouring, and sometimes as a preservative by killing or preventing the growth of harmful bacteria (Adamson, 2004). Many of these substances are also used for other purposes, such as medicine, religious rituals, cosmetics, perfumery or eating as vegetables. For example, turmeric is also used as a preservative; licorice as a medicine, garlic as a vegetable (Dalby, 2001).

Though the term spice can be used to incorporate herbs, the distinction between herbs and spices can be described as follows, in the kitchen, spices are distinguished from herbs, which are leafy, green plant parts used for flavouring purposes (Bender and Bender, 2005). Herbs, such as basil or oregano, may be used fresh, and are commonly chopped into smaller pieces. Spices, however, are dried and often ground or grated into a powder. Small seeds, such as fennel and mustard seeds, are used both whole and in powder form.

2.2 HISTORY

The use of spices dates back to ancient times. Archaeological excavations have revealed that pre-historical men used the leaves of certain plants to enhance the flavour of the half-cooked food which were eaten. Spices are mention in the Bible with examples being Exodus 35:28, 1 Kings 10:15 etc, the Bible also mentions that in 1000 BC, Queen Sheba visited King Solomon in Jerusalem and offered him "120 measures of gold, many spices, and precious stones." (www.chaddsfordhistory.org).

Ancient Greeks and Romans were familiar with spices and paid high prices for them (Baillon, 1877). Abundant anecdotal information documents the historical use of herbs and spices for their health benefits (Rosengarten, 1969). Early documentation suggests that hunters and gatherers wrapped meat in the leaves of bushes, accidentally discovering that this process enhanced the taste of the meat, as did certain nuts, seeds, berries, and bark. Over the years, spices and herbs were used for medicinal purposes. Spices were also used as a way to mask unpleasant tastes and odours of food, and later, to keep food fresh (www.chaddsfordhistory.org). Ancient civilizations did not distinguish between those spices and herbs used for flavouring from those used for medicinal purposes. When leaves, seeds, roots, or gums had a pleasant taste or agreeable odour, it became in demand and gradually became a norm for that culture as a condiment. Spices were also valuable as items of exchange and trade.

Earlier on, spices were used as a source of trading. During the ancient Roman Empire, trading largely came from Arabia; traders supplied cassia, cinnamon, and other spices and deliberately kept the source of their products secret. The intent was to have a monopoly on the spice trade and the Arabians spun great tales about how they obtained the spices in order to keep their resource value high. They continued to keep the origins secret for several centuries from both Ancient Greek and Ancient Roman civilizations (Rosengarten, 1969;

www.chaddsfordhistory.org) until about the 1st century, AD, when the Roman scholar Pliny made the connection between the Arabian stories and the inflation of spices and herbs.

Spices and herbs played an important role in ancient Greek medical science. Hippocrates (460-377 BC) wrote about spices, including saffron, cinnamon, thyme, coriander, mint, and marjoram. Of the 400 herbal remedies utilized by Hippocrates, at least half are in use today (Tapsell *et al*., 2006). Roughly 500 years later, Theophrastus (372-287 BC), sometimes called the "Father of Botany," wrote two books that summarized the knowledge of over 600 spices and herbs.

Historically, culinary spices and herbs have been used as food preservatives and for their health-enhancing properties. Papyri from Ancient Egypt in1555 BC classified coriander, fennel, juniper, cumin, garlic and thyme as health promoting spices (Tapsell *et al*., 2006). Records from that time also note that labourers who constructed the Great Pyramid of Cheops consumed onion and garlic as a means to promote health.

As time went on, these foodstuffs became more abundant as crops were extended, first in the country of origin, then in other countries. Prices fell and less wealthy consumers were able to purchase spices, which were gradually becoming less thought of as luxury items. Although spices today are still important in trade, their per capita use for flavouring food has declined in Western civilizations, and certain spices must compete with synthetic flavourings. The demand for spices has remained large in Asia, where spices have a wider social and ceremonial significance than they ever attained in the West. Unlike earlier times when monopolies dominated the spice trade, commerce in spices is now relatively decentralized. Throughout the world, spices and herbs are frequently used in cuisine, largely to improve flavour and to provide new tastes (McCormick.com Spice Encyclopaedia).

Today, people are increasingly interested in enjoying spices and herbs for health benefits. As research is progressing, more evidence is supporting some of the anecdotal information supplied by our ancestors. Thousands of years ago our hunter/gatherer forefathers were forced to adapt to a wide range of climatic and other environmental conditions. Thus, before agrarian societies developed, humans consumed a wide variety of plant species. Some of these plants contained the basic macro and micronutrients needed for survival (carbohydrates, fats, proteins, vitamins and minerals) (Thudichum, 1895). Many of the pungent, stronger-tasting, and richly coloured plants also contained an array of "non-nutritive compounds" that had profound health benefits.

2.3 MINERALS/ HEAVY METALS AND THEIR EFFECTS

Minerals (inorganic nutrients) are materials found in foods that are essential for growth and health and do not contain the element carbon. These include water, sodium, potassium, chloride, calcium, phosphate, sulphate, magnesium, iron, copper, zinc, manganese, iodine, selenium, and molybdenum etc it is made up of both micro and macro nutrients or elements. These micro elements mostly fall under heavy metals. Cobalt is a required mineral for human health, but it is supplied by vitamin B_{12}. There is some evidence that chromium, boron, and other inorganic elements play some part in human nutrition, but the evidence is indirect and not yet convincing. Fluoride seems not to be required for human life, but its presence in the diet contributes to long term dental health. Some of the minerals do not occur as single atoms, but occur as molecules. These include water, phosphate, sulphate, and selenite (a form of selenium). Sulphate contains an atom of sulphur. We do not need to eat sulphate, since the body can acquire the entire sulphate it needs from protein (Odell and Sunde, 1997).

The statement that various minerals, or inorganic nutrients, are required for life means that their continued supply in the diet is needed for growth, maintenance of body weight in adulthood, and for reproduction. The amount of each mineral that is needed to support growth during infancy and childhood, to maintain body weight and health, and to facilitate pregnancy and lactation, are listed in a table called the Recommended Dietary Allowances (RDA). This table was compiled by the Food and Nutrition Board of the United State of America. All of the values listed in the RDA indicate the daily amounts that are expected to maintain health throughout most of the general population. The actual level of each inorganic nutrient required by any given individual is likely to be less than that stated by the RDA. The RDAs are all based on studies that provided the exact, minimal requirement of each mineral needed to maintain health (Institute of Medicine, 1999; 2001

Table 2.1 Recommended Daily Intake for Food Supplements (www.lenntech.com/recommended-daily-intake.htm)

Minerals	Recommended daily intake	Over dosage
Boron	< 20 mg	Toxic at anything more than low levels to man
Calcium	1000 mg	Doses larger than 1500 mg may cause stomach problems for sensitive individuals
Chlorine	3400mg (in chloride form)	Excessive intake as NaCl can lead to health problems in man
Chromium	120 µg	Doses larger than 200 µg are toxic and may cause concentration problems and fainting
Copper	2 mg	As little as 10 mg of copper can have a toxic effect
Fluorine	3.5 mg	No information found
Iodine	150 µg	No information found
Iron	15 mg	Doses larger than 20 mg may cause stomach upset, constipation and blackened stools

Magnesium	350 mg	Doses larger than 400 mg may cause stomach problems and diarrhoea
Manganese	5 mg	Excess manganese may hinder iron adsorption
Molybdenum	75 µg	Doses larger than 200 µg may cause kidney problems and copper deficiencies
Nickel	< 1 mg	Products containing nickel may cause skin rash in case of allergies
Phosphorus	1000 mg	Contradiction: the FDA states that doses larger than 250 mg may cause stomach problems for sensitive individuals
Potassium	3500 mg	Large doses may cause stomach upsets, intestinal problems or heart rhythm disorder
Selenium	35 µg	Doses larger than 200 µg can be toxic.
Sodium	2400 mg	Excess may cause congestive heart failure.
Vanadium	< 1,8 mg	Toxic to health of man.
Zinc	15 mg	Doses larger than 25 mg may cause anaemia and copper deficiency.

Much as we need these minerals for a healthy living, some of these minerals in higher or trace quantities may be damaging to our health. These minerals are called trace or heavy metals. Heavy metals are those with atomic weights from 63.546 to 200.590 (Kennish, 1992) and specific gravity higher than 4. Living organisms require trace amounts of some heavy metals, including cobalt, copper, iron, manganese, molybdenum, vanadium, strontium, and zinc, but excessive levels can be detrimental to the organism. These are termed as beneficial heavy metals. These elements, or some form of them, are commonly found naturally in foodstuffs, in fruits and vegetables, and in commercially available multivitamin products (Schrauzer, 1984). Others are found in the environment such as the soil and water where these plants are grown. Other heavy metals such as mercury, plutonium, and lead are toxic metals that have no known vital or beneficial effect on organisms, and their accumulation over time in the bodies of animals can cause serious illness.

There is a strong link between micronutrient nutrition of plants, animals and humans and the uptake and impact of contaminants in these organisms (De Leonardis *et al*., 2000; Yuzbasi *et al*., 2003). The content of essential elements in plants is conditional, the content being affected by the characteristics of a soil and the ability of plants to selectively accumulate the metals. Additional sources of heavy metals in plants are: rainfall, traffic density, use of oil or fossil fuels for heating, atmospheric dusts, plant protection agents, and fertilizers. Certain elements that are normally toxic are, for certain organisms or under certain conditions, beneficial. Examples include vanadium, tungsten, and even cadmium (Lane and Morel, 2000; Lane *et al*., 2005).

In medical usage, heavy metals are loosely defined and includes all toxic metals irrespective of their atomic weight: "heavy metal poisoning" can possibly include excessive amounts of iron, manganese, aluminium, or beryllium (the fourth lightest element) or such a semimetal as arsenic as well as heavy metals (Duffus, 2002).

Some of these metals are naturally found in the body and are essential to human health. Iron, for example, prevents anaemia, and zinc is a cofactor in over 100 enzyme reactions. High levels of zinc can result in a deficiency of copper, another metal required by the body.

Trace metals therefore, are metals in extremely small quantities, almost at the molecular level, that reside in or are present in animal and plant cells and tissue. They are a necessary part of good nutrition, although they can be toxic if ingested at excess quantities (Underwood, 1977).

Trace metals or micronutrients include iron, magnesium, zinc, copper, chromium, nickel, cobalt, vanadium, molybdenum, and selenium. Trace metals are depleted through the expenditure of energy by a living organism. They are replenished in animals by eating plants, and replenished in plants through the uptake of nutrients from the soil in which the plant grows. Human vitamin pills and plant fertilizers both contain trace metals as additional sources for trace metals (Schrauzer, 1984).

The elements under this study include, zinc, manganese, iron and copper being micronutrients. The rest are mercury, cadmium and lead being the toxic elements. It will therefore be necessary to know its use or what it does in the human body when spices taken contain them.

2.3.1 Copper

Copper acts as a catalyst in the formation of haemoglobin, the oxygen-carrying blood component (Lahey, 1975). The areas of highest concentrations in the body tissues are the liver and certain areas of the central nervous system, particularly the brain. Minerals including copper must be bound to a protein to be usable.

Functions of Copper in the body include, helping oxidize glucose and release energy, helping the body absorb iron, aiding the thyroid gland in balancing and secreting hormones, carrying oxygen in the blood stream thus supplying the body's tissues with oxygen. It is also needed for the functioning of the amino acid, tyrosine. It is essential for making red blood cells, necessary for the synthesis of the hormone adrenaline etc.

2.3.1.1 Low Copper/High Copper

Physical Symptoms of Low Copper include not enough oxygen in the cells, lowered levels of HDL cholesterol, skin problems, swollen ankles, anaemia. Low level causes the cells to suffocate and lack oxygen. It is linked to low enkephalins produced in the brain. Psychological Symptoms of Low Copper include auditory hallucinations, depression. Binge eaters have been found to have lower levels of copper.

Copper levels are more often too high than too low. High copper can be toxic. It causes headaches, hypoglycemia, increased heart rate, nausea, inhibition of urine production, anaemia, hair loss in women, Damage to the kidneys. Copper deposits in the brain and liver causing damage to them (Nolan, 1983). Excessive copper in children is associated with hyperactive behaviour, learning disorders such as dyslexia, and ear infections. High copper interferes with zinc, which is needed to manufacture digestive enzymes. Many high copper people dislike protein and are drawn to high-carbohydrate diets because they have difficulty digesting protein foods (Narang *et al*., 1991)

2.3.2 Manganese

Manganese is a trace metal that is present at very low amounts in our body. The human body most likely would contain around 20 milligrams of manganese and most of them will be found and concentrated in the bones, kidneys, liver and pancreas.

Manganese, which has antioxidant, free-radical-fighting properties, is important for proper food digestion and for normal bone structure. Manganese can help reduce fatigue levels, prevent the incidence and severity of osteoporosis, and even improve memory. Manganese is a trace mineral that helps the body convert protein and fat to energy. It also promotes normal bone growth, helps maintain healthy reproductive, nervous, and immune systems, and is involved in blood sugar regulation. In addition, manganese is involved in blood clotting and the formation of cartilage and lubricating fluid in the joints.

Manganese levels have also been shown to vary with the menstrual or estrus cycle in humans and animals, and low manganese intakes are associated with disruption of reproduction in animals (Penland and Johnson, 1993). Manganese deficiency had been linked to bone malformation, weakness, seizures, atherosclerosis, confusion, convulsions, eye problems, hearing problems, heart disorders, high cholesterol levels, hypertension, irritability, memory loss, muscle contractions, pancreatic damage, profuse perspiration, rapid pulse, tooth-grinding, tremors, and osteoporosis. Manganese is also linked to decreased superoxide dismutase (SOD) activity in white blood cells, which leaves the body more vulnerable to the damaging effects of free-radicals (ATSDR, 2008b).

2.3.3 Zinc

Zinc is an essential metal present in nearly every cell of the body. In addition, the body needs zinc to use nutrients for immunity, for wound healing and for maintaining the senses of taste and smell.

Zinc stimulates the work of about 100 enzymes that keep the body functioning normally and performs both structural and catalytic functions in many different enzymes and associated with many different metabolic processes, including the synthesis of the nucleic acids RNA and DNA. It is required for the transport of vitamin A from the liver, and as part of superoxide dismutase (SOD) helps protect cells from free radicals. Zinc is also required for normal growth and development, reproductive development and function, and to support the immune system, where it has been shown to increase T-lymphocytes and enhance other white blood cell functions. However, because of its effect on increasing white blood counts, higher intake of zinc (unless low) is contraindicated with leukaemia.

Low levels of zinc cause lung cancer, neural tube defects, slow growth, anorexia. A study involving cancer showed that children with malignancies in remission exhibited the same values for zinc, copper, and a zinc/copper ratio as did controls. In addition, maternal levels of zinc were decreased in women who bore children with neural tube defects (Hansen, 1983; ATSDR, 2005).

2.3.4 Iron

Iron is essential for the formation of haemoglobin, the chemical in the blood that carries oxygen to the cells. Low levels of iron cause anaemia (Lahey, 1975). In severe cases, the children become flabby, and they fail to grow normally. Milder cases of iron deficiency may not produce any physical symptoms, but children may learn at a slower pace than children with a proper amount of iron in their diet. The combination of rice, beans, and meat consumed with fresh citrus fruit provides an excellent source of absorbable iron (Gillooly *et al*., 1983).

Discussion of iron toxicity in this protocol is limited to ingested or environmental exposure. Iron overload disease (haemochromatosis), an inherited disorder, is discussed in a separate protocol. Iron is a heavy metal of concern, particularly because ingesting dietary iron supplements may acutely poison young children (e.g., as few as five to nine 30-mg iron tablets for a 30-lb child).
Ingestion accounts for most of the toxic effects of iron because iron is absorbed rapidly in the gastrointestinal tract. The corrosive nature of iron seems to further increase the absorption (Hallberg *et al*., 1997). Most overdoses appear to be the result of children mistaking red-coated ferrous sulphate tablets or adult multivitamin preparations for candy. Other sources of iron are drinking water, iron pipes, and cookware. Target organs are the liver, cardiovascular system, and kidneys.

2.3.5 Lead

Lead accounts for most of the cases of paediatric heavy metal poisoning (Chao and Kikano, 1993). It is a very soft metal and was used in pipes, drains, and soldering materials for many years. Millions of homes built before 1940 still contain lead (e.g., in painted surfaces), leading to chronic exposure from weathering, flaking, chalking, and dust (Myres and Easson, 1992). Every year, industry produces about 2.5 million tons of lead throughout the world. Most of this lead is used for batteries. The remainder is used for cable coverings, plumbing, ammunition, and fuel additives. Other uses are as paint pigments (Myres and Easson, 1992) and in PVC plastics, x-ray shielding, crystal glass production, pencils, and pesticides.

Lead can affect almost every organ and system in your body. The main target for lead toxicity is the nervous system, both in adults and children. Long-term exposure of adults can result in decreased performance in some tests that measure functions of the nervous system. It may also cause weakness in fingers, wrists or ankles. Lead exposure also causes small increases in blood pressure, particularly in middle-aged and older people and can cause anaemia. Exposure to high lead levels can severely damage the brain and kidneys (Loghman-Adham, 1997) in adults or children and ultimately cause death. High-level exposure in men can damage the organs responsible for sperm production (ATSDR, 2000). In pregnant women, high levels of exposure to lead may cause miscarriage (Bellinger *et al*., 1987).

2.3.6 Mercury

Mercury is generated naturally in the environment from the degassing of the earth's crust, from volcanic emissions. It exists in three forms: elemental mercury, organic and inorganic mercury. Mining operations, chloralkali plants, and paper industries are significant producers of mercury. Atmospheric mercury is dispersed across the globe by winds and returns to the earth in rainfall, accumulating in aquatic food chains and fish in lakes. Mercury compounds were added to paint as a fungicide until 1990. These compounds are now banned; however, old paint supplies and surfaces painted with these old supplies still exist.

Mercury continues to be used in thermometers, thermostats, and dental amalgam. (Many researchers suspect dental amalgam as being a possible source of mercury toxicity). Medicines, such as mercurochrome and merthiolate, are still available. Algaecides and childhood vaccines are also potential sources (Warkeny and Hubbard, 1953). Inhalation is the most frequent cause of exposure to mercury and has adverse health effects. The organic form is readily absorbed in the gastrointestinal tract (90-100%); lesser but still significant amounts of inorganic mercury are absorbed in the gastrointestinal tract (7-15%). Target organs are the brain and kidneys. High mercury exposure results in permanent damage of the nervous system, kidney damage, lung irritation, spontaneous abortion, eye irritation, skin rashes, vomiting and diarrhoea (Serafin and Verity, 1991; Soresen *et al*., 1999; Goyer and Clarkson, 2001).

2.3.7 Cadmium

Cadmium is a by-product of the mining and smelting of lead and zinc. It is used in nickel-cadmium batteries, PVC plastics, and paint pigments. It can be found in soils because insecticides, fungicides, sludge, and commercial fertilizers that use cadmium are used in agriculture. Cadmium may be found in reservoirs containing shellfish. Cigarettes also contain cadmium. Lesser-known sources of exposure are dental alloys, electroplating, motor oil, and exhaust. Inhalation accounts for 15-50% of absorption through the respiratory system; 2-7% of ingested cadmium is absorbed in the gastrointestinal system. Target organs are the liver, placenta, kidneys, lungs, brain, and bones (ATSDR, 2008a).

2.4 CONVENTIONAL CLASSIFICATION OF SPICES

A conventional classification of spices is based on degree of taste as: hot spices, mild spices, aromatic spices herbs and aromatic vegetables.

Table 2.2 Classification of Spices

Classes	Spices
Hot spices	Capsicum (chillies), Cayenne pepper, black and white peppers, ginger, mustard
Mild spices	Paprika, coriander
Aromatic spices	Allspice (pimento), cardamom, cassia, cinnamon, clove, cumin, dill, fennel, fenugreek, mace and nutmeg
Herbs	Basil, bay, dill leaves, marjoram, tarragon, thyme
Aromatic vegetables	Onion, garlic, shallot, celery

Table 2.3 Parts of Plant used as spices

Plant organs	Spice crops
Aril	Mace of nutmeg
Barks	Cassia, cinnamon
Berries	Allspice, black pepper, chilli
Buds	Clove
Bulbs	Onion, garlic, leek
Pistil	Saffron
Kernel	Nutmeg
Leaf	Basil, bay leaf, mint, marjoram, sage, curry leaf
Rhizome	Ginger, turmeric
Latex	Asafoetida
Roots	Angelica, horse-radish
Seeds	Ajowan, aniseed, caraway, celery, coriander, dill, fennel, fenugreek, mustard, poppy seed

2.5 TYPES OF SPICES

There are so many types of spices, some very common and some not. They are Onion, Ginger, Garlic, Cloves, Dawadawa, Cinnamon, Rosemary, Thyme, Marjoram, Pepper, Black Pepper, Olive, Lime, Nutmeg, Curry, Allspice, Almond, Turmeric, Oregano, Sage, Lemon balm, Peppermint, Dill and Coriander etc.

2.6 USES OF SPICES

Spices have so many uses. These uses includes: Addition of flavour to foods thus making food taste good, for Medicinal Purposes which includes fighting against cancer causing cells, reducing the cholesterol level in the blood and preventing several skin diseases. Other uses include, Religious rituals, Cosmetics, Perfumery and Aromatherapy.

2.7 IMPORTANCE OF SPICES

There are many reasons for which people use spices, though, taste probably tops the list. There are several spices that simply smell good and those smells can be alternately soothing or exciting. On the other hand, ginger is said to be a stimulant and the smell of ginger could be just what you need to pep up your mood. Over the centuries, certain spices have been said to heal every infection, disease and malady known to man. But there's no doubt that spices do have value far beyond enhancing the taste of food (Ziegler and Filer, 1996).

Spices play an important role in the nutrition of our daily diet. Scientists have done a lot of research on this and have found out that spices contain more antioxidants than fruits and vegetables. Spices contain more antioxidants when they are dried than when they are raw or fresh. Half teaspoon of spices will contribute more amounts of antioxidants than half a cup of fruits. Spices play an active role by acting as medicines. Cloves, oregano, allspice, cinnamon, sage, peppermint, thyme and lemon balm are some of the spices. These spices may be of a significant dietary source (Pokorney, 1991).

Spices, the predominant flavouring, colouring and aromatic agents in foods and beverages, are now gaining importance for their diversified uses. The nutritional, anti-oxidant, anti-microbial and medicinal properties of spices have far-reaching implications (Shelef, 1983). In the present scenario, the anti-diabetic, anti-hypercholesterolemic, anti-carcinogenic, anti-inflammatory effects of spices have paramount importance, as the key health issues of mankind nowadays are diabetes, cardio-vascular diseases, arthritis and cancer. Spices or their active principles could be used as possible ameliorative or preventive agents for these health disorders (Shils, 1999).

Extensive studies on animal models carried out indicate that spices could be consumed at higher dietary levels without any adverse effects on growth, organ weights, and food efficiency ratio and blood constituents. Curcumin, the colouring pigment present in turmeric, capsaicin, the pungent principle in red pepper, allicin, the active principle in garlic, gingerol, the pungent principle in ginger, saponin and fibre present in fenugreek are immensely valuable in health care with their multiple physiological effects (Stipanuk, 2000).

However, until recent times, the desiccation and freezing of food was not a viable option for those living in hot, humid climates; these societies discovered chemical preservation, in the form of salt and spices. As the former was only available in certain areas spices were often the only other option to protect food from insect infestation and microbial putrefaction (Sethi and Meena, 1997).

We now know that many of the strongly flavoured phytochemicals which give plants protection against insect and microbial attack are the same compounds that "preserve" our bodies, by protecting us against degenerative disease (Shils, 1999).

Today spices are consumed in much greater quantity and variety in warm, humid countries than in colder climates. India and Thailand have the highest consumption of spices; the warm Mediterranean countries follow somewhat behind these and other Eastern countries but are ahead of the United States. Chilly Scandinavian countries have the lowest spice consumption of all. Moreover, the importance of spices in helping to prevent chronic degenerative disease can be seen to correspond to the varying levels of spice utilization that occurs across different temperature zones. Cold countries, typically the most developed countries, tend to have much higher incidences of chronic degenerative diseases when compared to hotter regions.

There is the need to consume lots of spices on a daily basis as they can make us feel better, think better, age more slowly, and help to resist the onslaught of scourges like cardiovascular disease, cancer, diabetes, Alzheimer's disease and other chronic degenerative disorders (Shils, 1999).

2.8 SPICES AND THEIR HEALTH EFFECTS

Spices have numerous effects on the human body. Some of the health effects are discussed below.

2.8.1 Cloves (*Sypzygium aromaticum)*

Like other spices, cloves are available throughout the year. Cloves are the unopened pink flower buds of the evergreen clove tree. The buds are picked by hand when they are pink and dried until they turn brown in colour. Cloves resemble tiny nails. In fact, it English name is actually derived from the Latin word clavus, which means nail. Although cloves have a very hard exterior, their flesh features an oily compound that is essential to their nutritional and flavour profile.

Clove contains significant amounts of an active component called eugenol, which has made it the subject of numerous health studies, including studies on the prevention of toxicity from environmental pollutants like carbon tetrachloride, digestive tract cancers, and joint inflammation. In the United States, eugenol extracts from clove have often been used in dentistry in conjunction with root canal therapy, temporary fillings, and general gum pain, since eugenol and other components of clove (including beta-caryophyllene) combine to make clove a mild anaesthetic as well as an anti-bacterial agent (Amaechi *et al.*, 1999). Eugenol, the primary component of clove's volatile oils, functions as an anti-inflammatory substance. Clove also contains a variety of flavonoids, including kaempferol and rhamnetin, which also contribute to clove's anti-inflammatory and antioxidant properties (Friedman *et al*., 2002).

Like its fellow spices, clove's unique phytonutrient components are accompanied by an incredible variety of traditionally-recognized nutrients. Cloves are excellent source of manganese, a very good source of dietary fibre, vitamin C and omega-3 fatty acids and a good source of calcium and magnesium (Ensminger and Esminger, 1986).

2.8.2 Onion (*Allium cepa*)

Onions, like garlic, are a member of the Allium family, and are rich in powerful sulphur-containing compounds that are responsible for their pungent odours and for many of their health-promoting effect. Onions contain allyl propyl disulphide, (while garlic is rich in allicin, diallyl disulphide, diallyl trisulfide and others). In addition, onions are very rich in chromium, a trace mineral that helps cells respond to insulin, plus vitamin C, and numerous flavonoids.

The cysteine sulfoxides are primarily responsible for the onion flavour and produce the eye-irritating compounds that induce lacrimation (www.scientificamerican.com). Onion is not as potent as garlic since the sulphur compounds in onion are only about one-quarter the level found in garlic. Onions have a variety of medicinal effects. Early American settlers used wild onions to treat colds, coughs, and asthma, and to repel insects. In Chinese medicine, onions have been used to treat angina, coughs, bacterial infections, and breathing problems.

The regular consumption of onions has, like garlic, been shown to lower high cholesterol levels and high blood pressure, both of which help prevent atherosclerosis and diabetic heart disease, and reduce the risk of heart attack or stroke. These beneficial effects are likely due to onions' sulphur compounds, its chromium and its vitamin B6 content, which helps prevent heart disease by lowering high homocysteine levels, another significant risk factor for heart attack and stroke. Onions have been singled out as one of the small number of vegetables and fruits that contributed to the significant reduction in heart disease risk seen in a meta-analysis of seven prospective studies (Huxley and Neil, 2003).

Quercitin, an antioxidant in onions, and curcumin, a phytonutrient found in the curry spice turmeric, reduce both the size and number of precancerous lesions in the human intestinal tract (Yang *et al.*, 2004). Making onion and garlic a staple in your healthy way of eating may greatly lower your risk of several common cancers, (Fukushima *et al*., 1997) suggests a large data set of case-control studies from Southern European populations (Galeone *et al*., 2006). In addition, quercitin, the thiosulfinates exhibit antimicrobial properties and other flavonoids found in onions work with vitamin C to help kill harmful bacteria. Onion is effective against many bacteria including *Bacillus subtilis*, *Salmonella*, and *E. coli* (Augusti, 1996) making onions an especially good addition to soups and stews during cold and flu season.

The World Health Organisation (WHO) supports the use of onions for the treatment of poor appetite and to prevent atherosclerosis. In addition, onion extracts are recognised by WHO for providing relief in the treatment of coughs and colds, asthma and bronchitis. Onions are known to decrease bronchial spasms. An onion extract was found to decrease allergy-induced bronchial constriction in asthma patients.

Onions have a universal appeal. They are safely consumed by most people. However, consuming large quantities of onions can lead to stomach distress and gastrointestinal irritation that may result in nausea and diarrhoea. There are no known interactions with drugs except that they can potentiate the action of coagulants.

2.8.3 Pepper

With pepper, two separate parts of the plant, the fruit, which is an edible herb, and the ground seed which is used as a spice are of importance. The fruit of the sweet pepper, which is large and hollow, is popular for adding colour and flavour to salads and stir fry dishes.

2.8.3.1 Black pepper (*Piper nigrum*)

Black pepper (*Piper nigrum*) is a flowering vine in the family Piperaceae, cultivated for its fruit, which is usually dried and used as a spice and seasoning. The same fruit is also used to produce white pepper, red/pink pepper, and green pepper. The fruit, known as a peppercorn when dried, is a small drupe five millimetres in diameter, dark red when fully mature, containing a single seed.

Dried ground pepper is one of the most common spices in European cuisine and its descendants, having been known and prized since antiquity for both its flavour and its use as a medicine. The spiciness of black pepper is due to the chemical piperine (McGee, 2004).

Black pepper has long been recognized as a carminitive, (a substance that helps prevent the formation of intestinal gas), a property likely due to its beneficial effect of stimulating hydrochloric acid production. In addition, black pepper has diaphoretic (promotes sweating), and diuretic (promotes urination) properties.

Black pepper has demonstrated impressive antioxidant and antibacterial effects yet another way in which this wonderful seasoning promotes the health of the digestive tract Pepper also has antimicrobial properties (Dorman and Deans 2000). And not only does black pepper help you derive the most benefit from your food, the outer layer of the peppercorn stimulates the breakdown of fat cells, keeping you slim while giving you energy to burn. Black pepper is an excellent source of manganese, a very good source of iron and vitamin K, and a good source of dietary fibre.

2.8.3.2White pepper (*Piper nigrum*)

White pepper consists of the seed only, with the fruit removed. This is usually accomplished by allowing fully ripe berries to soak in water for about a week, during which the flesh of the fruit softens and decomposes. Rubbing then removes what remains of the fruit, and the naked seed is dried. Alternative processes are used for removing the outer fruit from the seed, including removal of the outer layer from black pepper produced from unripe berries (McGee, 2004).

White pepper which comes from the plant as black pepper has the same properties. It has long been recognized as a carminitive, (a substance that helps prevent the formation of intestinal gas), a property likely due to its beneficial effect of stimulating hydrochloric acid production. In addition, black pepper has diaphoretic (promotes sweating), and diuretic (promotes urination) properties (Dorman and Deans 2000). Too much will slow the rate at which certain drugs are cleared by the liver from the blood stream (www.mountainroseherbs.com).

2.8.3.3 Red pepper (chilli) (*Capsicum annuum*)

Red pepper, is a dried, ground fruit pod. It is also called cayenne pepper and provides the dominant flavour of chilli con carne. The chilli pepper, or more simply just "chilli", is the fruit of the plant Capsicum from the nightshade family, Solanaceae. These terms usually refer to the smaller, hotter types of capsicum; the mild larger types are called bell pepper (simply pepper in Britain and Ireland or capsicum in Australasia). The substances that give chilli peppers their heat is capsaicin (8-methyl-N-vanillyl-6-nonenamide) and several related chemicals, collectively called capsaicinoids.

Red chillies are very rich in vitamin C and provitamin A. Yellow and especially green chillies (which are essentially unripe fruit) contain a considerably lower amount of both substances. In addition, peppers are a good source of most B vitamins, and vitamin B6 in particular. They are very high in potassium and high in magnesium and iron. Pepper also has antimicrobial properties (Dorman and Deans 2000). Recent studies reveal that chilli peppers can have a wide range of benefits from helping alleviate pain in arthritis patients to acting effectively against cancer causing tumours (Daily Mail, 2007).

Experts say capsaicin, the chemical that gives spicy food its kick, could be used to kill tumours with few or no side effects for the patient Chilli peppers also have cardiovascular benefits, weight loss properties (by inducing thermogenesis) and helps to clear mucus from stuffed noses or congested lungs (Heidi, 2002). Chilli also helps to lower the risk of Diabetes; according to a study by the American Journal of Clinical Nutrition, the amount of insulin required to lower blood sugar after a meal is reduced if the meal contains chilli pepper. Canadian researchers believe that chillies could play a vital role in curing diabetes (Ahuja *et al.*, 2006). Red chili peppers, such as cayenne, have been shown to reduce blood cholesterol, triglyceride levels, and platelet aggregation, while increasing the body's ability to dissolve fibrin, a substance integral to the formation of blood clots. Cultures where hot pepper is used liberally have a much lower rate of heart attack, stroke, pulmonary embolism and also help fight against prostate cancer (Mori *et al*., 2006).

The 5th century Syriac Book of Medicines prescribes pepper (or perhaps long pepper) for such illnesses as constipation, diarrhoea, earache, gangrene, heart disease, hernia, hoarseness, indigestion, insect bites, insomnia, joint pain, liver problems, lung disease, oral abscesses, sunburn, tooth decay, and toothaches (Turner, 2004).

Chili peppers have a bad and mistaken reputation for contributing to stomach ulcers. Not only do they not cause ulcers, they can help prevent them by killing bacteria that may have been ingested, while stimulating the cells lining the stomach to secrete protective buffering juices. It is a good idea to keep cayenne pepper in the first aid kit. It is a marvellous aid for stopping internal bleeding that may result from an accident and it can help abort a heart attack in progress. One tablespoonful should be taken in a glass of water as soon as possible.

2.8.4 Garlic (*Allium sativum*)

Garlic is a member of the lily or Allium family, which also includes onions. Garlic is rich in a variety of powerful sulphur-containing compounds including thiosulfinates (of which the best known compound is allicin), sulfoxides (among which the best known compound is alliin), and dithiins (in which the most researched compound is ajoene). While these compounds are responsible for garlic's characteristically pungent odour, they are also the source of many of its health-promoting effects including cancer prevention (Fukushima *et al*., 1997; Andorfer *et al*., 2003). In addition, garlic is an excellent source of manganese, a very good source of vitamin B6 and vitamin C and a good source of selenium.

Numerous studies have demonstrated potential benefits of regular garlic consumption on blood pressure, platelet aggregation, serum triglyceride level, and cholesterol levels. Routine eating of garlic may also help stimulate the production of nitric oxide in the lining of blood vessel walls, which may help to relax them (Apitz-Castro *et al*., 1986; Spigelski and Jones, 2001). As a result of these beneficial actions, garlic can be described as a food that may help prevent cancer, atherosclerosis and diabetic heart disease, as well as reducing the risk of heart attack or stroke (Berthold and Sudhop, 1998; Fleischauer *et al*., 2000). The compounds in garlic responsible for its pungency also excite a neuron pathway providing cardiovascular benefits. Garlic's numerous beneficial cardiovascular effects are due to not only its sulphur compounds, but also to its vitamin C, vitamin B6, selenium, manganese calcium, potassium, iron and copper (Fugh-Berman, 2000; Bautista, *et al*., 2005).

One reason for garlic's beneficial effects may be its ability to lessen the amount of free radicals present in the bloodstream (Dillon *et al*., 2003). According to a study published in Life Sciences, a daily dose of 1 ml/kg body weight of garlic extract for six months resulted in a significant reduction in oxidant (free radical) stress in the blood of patients with atherosclerosis. Garlic is a very good source of vitamin C, the body's primary antioxidant defender in all aqueous (water-soluble) areas, such as the bloodstream, where it protects LDL cholesterol from oxidation. Since it is the oxidized form of LDL cholesterol that initiates damage to blood vessel walls, reducing levels of oxidizing free radicals in the bloodstream can have a profound effect on preventing cardiovascular disease (Superko and Krauss, 2000; Bhattacharya *et al*., 2004).

The selenium in garlic not only helps prevent heart disease, but also provides protection against cancer and heavy metal toxicity. A cofactor of glutathione peroxidase (one of the body's most important internally produced antioxidants), selenium also works with vitamin E in a number of vital antioxidant systems (Durak *et al.*, 2004) and another trace mineral, manganese, which also functions as a cofactor in a number of other important antioxidant defence enzymes.

Garlic, like onions, contains compounds that fight severe attacks in some cases of asthma and may also help reduce the pain and inflammation of osteoarthritis and rheumatoid arthritis. Allicin, is a powerful antibacterial and antiviral agent that joins forces with vitamin C to help kill harmful microbes and lower blood pressure, insulin and triglycerides (Lee *et al.*, 2003).

Side effects from garlic intake include upset stomach, bloating, bad breath, body odour, and a stinging sensation on the skin from handling too much fresh or dried garlic. Handling garlic may also cause skin lesions. Other, more rare side effects that have been reported by those taking garlic supplements include headache, fatigue, loss of appetite, muscle aches, dizziness described as vertigo (dizziness), and allergies such as an asthmatic reaction or contact dermatitis (skin rash).

2.8.5 Nutmeg (*Myristica frgrans*)

Unlike other herbs and spices, the health benefits of nutmeg have not been well known, although there are many benefits to this tasty spice. Nutmeg has a variety of health benefits and has been used as a healing herb for many centuries. It can help lower blood pressure and sooth a stomach ache as well as stop diarrhoea and help to detoxify the body. The essential oil, of the nutmeg is particularly effective for soothing many ailments, it is rich in phosphorus, calcium and iron. The most important species commercially known is the Common or Fragrant Nutmeg Myristica fragrans which belongs to the family Myristicaceae.

One of the interesting health benefits of nutmeg oil is its ability to stimulate the brain - it relieves stress and stimulates mental activity. It is even reputed to stimulate dreams. Its ability to improve concentration and increase efficiency was not lost on the ancient Greeks and Romans who used it as a brain tonic despite the fact that it was quite rare and costly.

Nutmeg has anti-inflammatory properties and can be used to treat joint and muscle pain. The oil works particularly well when it is massaged into the affected area (Devereux, 1996). It is an integral spice in Chinese medicine where it is used for stomach pain and inflammation as well as reducing joint swelling. In addition to being an excellent liver tonic which can help remove toxins from the liver, nutmeg oil is also a good herb for the kidney, helping it dissolve kidney stones as well as relieve infections of the kidney. Heart problems may also be somewhat alleviated by nutmeg, as it can help increase blood circulation and stimulate the cardio-vascular system. It is also good for digestion. It can get rid of both gas and stomach aches and can relieve vomiting, diarrhoea, and flatulence as well as encourage appetite. Nutmeg can also help with respiratory problems such as a cough from the common cold. It is an ingredient in cough syrups and is able to relieve asthmatic attack (www.indepthinfo.com).

While there are many health benefits of nutmeg, it is not advisable to be taken in high doses. It can be toxic and can cause serious problems. Not more than 30 grams (around 6 tablespoons) should be taken in a day, and even this amount may be considered excessive. In low doses, nutmeg produces no noticeable physiological or neurological response. Large doses of 60 g (~12 teaspoons) or more are dangerous, potentially inducing convulsions, palpitations, nausea, eventual dehydration, and generalized body pain (Gable, 2006).

2.8.6 Ginger (*Zingiber officinale*)

Ginger is a perennial spice which grows from underground rhizomes, which are often mistakenly called the "roots." Botanically it is the rhizome that provides the slightly hot, citrus-like taste, and wonderful aroma. Its family name is Zingiberaceae.

The rhizome has thick lobes coloured from tan to white. A highly valued variety, especially for medicinal uses, has a blue ring circling the fleshy inside of the rhizome. The nutrients present inside ginger, especially its volatile oils - gingerols and shogaols, accord a number of health benefits to its users (Chen *et al*., 2007). In fact, ginger has also been found to be effective in fighting some fatal ailments like cancer (Afshari and Taghizade, 2007). Ginger has been known to contain nutrients such as Calcium, Carbohydrate, Dietary Fibre, Iron, Magnesium, Manganese, Potassium, Protein, Selenium, Sodium, Vitamin C, E and B6.

Ginger has been found to be helpful in blocking the harmful effects of prostaglandin, a substance that can lead to inflammation of the blood vessels in the brain and even cause migraines. Ginger has been associated with alleviation of the feeling of nausea, even in case of pregnant women (Portnoi *et al.*, 2003). The anti-inflammatory properties of ginger make it effective in alleviation of pains associated with rheumatoid arthritis. Ginger promotes warmth in the upper respiratory tract and thus, is quite effective in treating cold and flu.

Ginger is quite effective for relief of cramps caused by stomach gas. Ginger makes the platelets less sticky and is thus, pretty helpful in case of circulatory disorders. Preliminary studies suggest that ginger may lower cholesterol and prevents the clotting of blood. Each of these effects may protect the blood vessels from blockage and the damaging effects of blockage such as atherosclerosis, which can lead to a heart attack or stroke. (Bordia *et al*., 1997).

There are a variety of uses suggested for ginger. Tea brewed from ginger is a folk remedy for colds. Ginger ale and ginger beer have been recommended as "stomach settlers" for generations in countries where the beverages are made, and ginger water was commonly used to avoid heat cramps in the US. Ginger has also been historically used to treat inflammation which several scientific studies support, though one arthritis trial showed ginger to be no better than a placebo or ibuprofen. Research on rats suggests that ginger may be useful for treating diabetes (Al-Amin and Zainab, 2006; Thomson *et al*., 2002). Side effects associated with ginger are rare, but if taken in excessive doses the herb may cause mild heartburn, diarrhoea and irritation of the mouth.

2.8.7 Rosemary (*Rosemaryinus officinalis*)

Rosemary grows on a small evergreen shrub belonging to the Labiatae family that is related to mint. Its leaves look like flat pine-tree needles, deep green in colour on top while silver-white on their underside. Its memorable flavour and unique health benefits makes it an indispensable spice for every kitchen.

Rosemary contains substances that are useful for stimulating the immune system, increasing circulation, and improving digestion. Rosemary also contains anti-inflammatory compounds that may reduce the severity of asthmatic attacks. It has antibacterial, antifungal, antiviral, spasmolytic, antioxidant (Aruoma *et al*., 1996; Kelm *et al*., 2000; Oluwatuyi *et al*., 2004). It is a smooth muscle modulating, analgesic, venotonic. It contains phenolic acids (rosmarinic acid), bitter diterpenes (carnosol, rosmanol) triterpenes (oleanic and ursolic acid), triterpene alcohols and flavonoids (Kelm *et al.*, 2000). The essential oil contained in the plant contains 1,8-cineole, α-pinene, camphor, β-pinene, borneol, iso-bornyl acetate, limonene, linalool, 3-octanone, terpineol and verbinol. In addition, rosemary has been shown to increase the blood flow to the head and brain, improving concentration and therapeutic properties (Hanrahan and Frey, 2005).

Rosemary is a good source of the minerals, iron and calcium, as well as dietary fibre. Fresh rosemary has 25% more manganese (which is somehow lost in the process of drying) and a 40% less calcium and iron, probably due to the higher water content. It also contains carbohydrates, sugar, soluble and insoluble fibre, sodium, vitamins, minerals, fatty acids and amino acids. (Ensminger and Esminger, 1986).

Rosemary is generally considered safe when taken in recommended doses. However, there have been occasional reports of allergic reactions. Large quantities of rosemary leaves, because of their volatile oil content, can cause serious side effects, including vomiting, spasms, coma and, in some cases, pulmonary edema (fluid in the lungs).

Because larger doses of rosemary may cause miscarriage, pregnant and nursing women should not use it in quantities other than those used for cooking (Lemonica *et al*, 1996). People with high blood pressure, ulcers, Crohn's disease, or ulcerative colitis should not take rosemary.

2.8.8 Thyme (*Thymus vulgaris*)

Thyme leaves are curled, elliptically shaped and very small, measuring about one-eighth of an inch long and one-sixteenth of an inch wide. The upper leaf is green-grey in colour on top, while the underside is a whitish colour.

Thyme has a long history of use in natural medicine in treatment of chest and respiratory problems including coughs, bronchitis, and chest congestion. Only recently, however, have researchers pinpointed some of the components in thyme that bring about its healing effects. The volatile oil components of thyme are now known to include carvacolo, borneol, geraniol, but most importantly, thymol (Bagamboula *et al*., 2004). Thymol - named after the spice itself - is the primary volatile oil constituent of thyme, and its health-supporting effects are well documented. In studies on aging in rats, thymol has been found to protect brain, kidney, and heart cell membranes when dietary supplementation with thyme was increased.

Thyme also contains a variety of flavonoids, including apigenin, naringenin, luteolin, and thymonin. These flavonoids increase thyme's antioxidant capacity, and combined with its status as a very good source of manganese, give thyme a high standing on the list of anti-oxidant foods (Kelm *et al.*, 2000). The volatile oil components of thyme have also been shown to have antimicrobial activity against a host of different bacteria and fungi. *Staphalococcus aureus*, *Bacillus subtilis*, *Escherichia coli* and *Shigella sonnei* are a few of the species for which thyme has been shown to have antibacterial activity (Kulevanova *et al.*, 2000).

For thousands of years, herbs and spices have been used to help preserve foods and protect them from microbial contamination. Research shows that both thyme and basil contain constituents that can both prevent contamination and decontaminate previously contaminated foods (Cosentino *et al.*, 1999). It is an excellent source of iron, a very good source of calcium, manganese and a food source of dietary fibre.

Thyme is reputed to affect the menstrual cycle, so its consumption is not recommended for pregnant or lactating mothers.

2.8.9 Anise (*Pimpinella anisum*)

Anise seed is a gray brown oval seed from <u>Pimpinella anisum</u> a plant in the parsley family. It is related to caraway, dill, cumin and fennel. Anise seeds smell and taste like licorice. Anise is native to Middle East and has been used as a medicine and as a flavour for medicine since prehistotoric times. Ancient Romans hung anise plant near their pillows to prevent bad dreams. They also used anise to aid digestion and ward off epileptic attacks (Chopra and Chandler, 1928).

Pythagoras claimed that the anise enhanced man's strength, cured insomnia, stimulated appetite, made men more fertile and facilitated digestion. It is rich in vitamins, the B complex (B1, B2), C, calcium, phosphorus, potassium, sulphur, iron, aromatic essences. It also has pharmacological properties which include, carminative, antispasmodic, expectorant, a pancreatic stimulant. The anise fruit has a nice taste and is used in phytotherapy. As a ripe fruit or juice, it is recommended for curing asthenia and for stimulating suckling mothers' lactation. It is also recommended for curing nervous asthenia, migraines, vertigos, rheumatism, bronchial asthma, gastric pains, and slow digestion. The oil is also antispasmodic, helping to relieve intestinal gas and spasmodic coughs (Blumenthal *et al*., 1998). Anise has been combined with cathartic laxatives to reduce the spasmodic cramping. It has modest antiparasitic actions and has been recommended by some practitioners to treat mild intestinal parasite infections (Weiss, 1985).

2.8.10 Aidan (Prekese) (*Tetrapleura tetraptera*)

Prekese (*Tetrapleura tetraptera*) which is an indigenous tree species belonging to the Mimosaceae family. In West Africa, the plant Tetrapleura tetraptera (locally known as Prekese) is popular among the Akans of Ghana for its use as a spice, as a dietary supplement rich in vitamins and a medicine for many ailments. Traditionally, the fruits, leaves, bark and roots are seen to have important medicinal properties.

Research has demonstrated how careful planting of Tetrapleura tetraptera in areas of high Billharzia transmission can reduce the rates of infection, offering countries with limited resource a more environmentally and financially friendly way of protecting their populations from this dreaded disease. It was found that it had anti-ulcer and anti-convulscant properties, confirming its ethno medicinal use to treat these symptoms. The active ingredients were found to be rapidly passed through the mammalian body, with little retention in tissues. They were also found to exhibit very few toxic effects, and were mutagenic only in the presence of other more dangerous chemicals which are not frequently found.

Prekese has many potential uses and should therefore not be allowed to be destroyed. For instance it is a potential source of raw material for the growing pole industry of Ghana, which is currently based on teak. Farmers are critical of negative environmental impact of teak therefore it is a suitable indigenous substitute which is agro-forestry and environmentally friendly (Adewunmi, 2008).

Biological study has shown that Prekese extract has some useful therapeutic action easing hypertension, and Asthma. Active constituents include Scopletin which appears to have a relaxing action on smooth muscle, helping to ease constriction in the Bronchioles of the lung, and on constricted blood vessels. Research has shown that *Tetrapleura tetraptera* also has anti-ulcerogenic, mollusicidal, and anti-microbial action (www.nkran.net/foodanddrink.jsp; www.ngrguardiannews.com).

2.8.11 Senegal pepper (Hwentea) (*Xylopia aethiopia*)

There are several widely used indigenous forest species which are harvested from the wild. *Xylopia aethiopia* or Ethiopia pepper or Senegal pepper or locally known as (hwentea), the seeds are used as spice and a substitute for pepper. As a spice Sénégal Pepper should always be used whole and ground, as the hull of the fruit lends the spice its aromatic notes whilst the seeds within lend pungency and bitterness to the flavour.

The chemical composition and mineral constituents of Senegal pepper, which is valued as a spice in Nigeria, were determined along with the physicochemical characteristics of the seed oil. The seeds had the following chemical compositions: moisture, ash, crude lipid, crude protein crude fibre and carbohydrate. Calcium and potassium were the major minerals in the seed. The extracted lipid was examined for the fatty acid composition. Linoleic and oleic acids were the predominant unsaturated fatty acids, while palmitic acid was the major saturated acid. The iodine value indicates that the seed oil is a non-drying type.

The essential oils of Senegal pepper and four Cameroonian plants used as spices in local food, showed antibacterial and antifungal activity (Tatsadjieu *et al*., 2003). Seeds are used for stomach-aches, dysentery, bronchitis, cancer, ulcers, fever and debility, rheumatism, post-partum management and fertility-enhancing, and vermifuge (Guardian newspaper, 2009).

2.8.12 Grain of paradise (famu wisa) (*Aframomum melegueta)*

Grains of paradise from the Zingiberaceae (ginger family) and known scientifically as *Aframomum melegueta* are native to some Africa's West coast countries, namely Ghana, Liberia, Ivory Coast, Togo and Nigeria. In the countries of origin, the seeds are used not only to flavour food, but they are also chewed on cold days to warm the body and also valued for their digestive properties.

In West Africa, grains of paradise are now hard to obtain, but still valuable for people following old recipes (e.g., for sausages or spiced wine). But this pungent grain is a worthy addition to many other everyday dishes. Their hotness is not as strong as in pepper, but more subtle and goes well with vegetables (potatoes, aubergines, pumpkin). The essential oil from grains of paradise is dominated by the sesquiterpene hydrocarbons humulene, α- and β-caryophyllene (together 83%) and their oxides (together 9%) (en.wikipedia.org). The powerful anti-inflammatory and antibiotic effects of grains of Paradise are proving to be potent and very successful in repeated laboratory tests conducted throughout the United States.

Historically, melegueta pepper and grains of paradise have been used to aid digestion, as a diuretic, reduce inflammation, relieve pain topically, slow the effects of aging, relieve toothaches and nausea, relieve migraines, as an antibiotic, increase breast milk production and relieve heartburn. As a stimulant, carminative and diuretic, the seeds are mainly used in some veterinary medicines. They also comfort and warm the weak, the cold and feeble stomach ache, help the aged. The seeds together with rhizomes are used in West African herbal medicines (www.theepicentre.com; Rosengarten, 1969).

2.8.13 Ashanti pepper (soro Wisa) (*Piper guineense*)

"False cubeb pepper" also known as "soro wisa" stems from Central Africa of the species *Piper guineense* (syn. P. clusii). Its fruits, also known as "ashanti pepper", indeed strongly resemble cubeb berries, but are prolate-elliptically shaped, smaller, smoother in surface and somewhat reddish coloured. Ashanti pepper tastes like the cubeb pepper, but fresher and less bitter.

The stalked berries are a little bit larger than pepper corns, having a furrowed surface. Most berries are hollow. They are sold whole and should be crushed or ground before usage. It belongs to the Piperaceae (pepper family). It has a pungent and bitter with a strong terpene aroma. It has anti-convulscant properties (Abila *et al*., 1996).

CHAPTER THREE

3 MATERIALS AND METHODS

3.1 STUDY AREA

The markets for this study were selected at random. In Kumasi the markets selected were Central, Asafo and Ayigya. In Accra, Kaneshie and Makola were selected. In Takoradi, the Market circle and the Tema market. A total seven markets were selected for the study.

3.2 APPARATUS

All glassware used were soaked in detergent solution, rinsed and soaked in 10% (v/v) HNO_3 overnight. They were rinsed with distilled water followed by 0.5% (w/v) $KMnO_4$, rinsed with distilled water and dried before use.

Thick walled 50mL digestion tubes, digestion blocks and a hot plate of the temperature range of 150-350^oC were used.

Automated Mercury Analyzer Model HG-5000 (Sanso Seisakusho Co., Ltd, Japan), equipped with mercury lamp operated at wavelength of 253.7 nm was used for the determinations of mercury. The signals were obtained on a Yokogawa Model strip chart recorder.

SOLAAR (S Series 711239 v1.23) Flame Atomic Absorption Spectrometer was used for the determination of zinc, cadmium, manganese, iron and copper.

3.3 REAGENTS

- Sulphuric Acid (H_2SO_4)
- Nitric Acid (HNO_3)
- Hydrochloric Acid (HCl)
- Perchloric Acid ($HClO_4$)

- Potassium Permanganate ($KMnO_4$)
- Tin (II) Chloride ($SnCl_2.H_2O$)

All reagents used were of analytical grade (BDH Chemicals Ltd, Poole. England). Double distilled water was used for the preparation of all solutions.

3.3.1 Preparation of Mercury Standard Solution

Mercury stock standard solution (1000mg/L) was prepared by dissolving 0.677g of $HgCl_2$ in 2ml HNO_3, 2ml $HClO_4$ and 5ml H_2SO_4 in a 50ml digestion flask with heating on a hot plate at a temperature of 200^oC for 30 minutes. The solution was then diluted to 50ml with distilled water.

Stannous Chloride solution (10%) was prepared by dissolving 10g of the salt in 100ml of 1M HCl. The solution was aerated with nitrogen gas at 50ml/min for 30 minutes to expel any elemental mercury.

3.3.2 Preparation of Standard solutions

The lead, Cadmium, Iron, Zinc, Manganese and Copper stock solutions of 1000ppm were prepared by dissolving 1g of pure metal in a minimum volume of 1% HCl and diluted to 1 litre with 1% HCl. Solutions (100ppm) of Lead, Cadmium, Iron, Zinc, Manganese and Copper were prepared by pipetting 10ml each of 1000ppm stock solution into in a 100ml volumetric flasks containing 10ml of 10% nitric acid and then made up to volume with distilled water. To obtain 2 and 4 ppm standard solutions for Fe, Mn and Zn, 2 and 4ml each of 100ppm solutions of Fe, Mn and Zn were pipetted into a 100ml volumetric flasks containing10ml 10% nitric acid and made up to the mark with distilled water. To obtain 3 and 9 ppm standard solutions for Cu, Pb and Cd, 3 and 9ml of 100ppm solutions of each of the three metals were pipetted into 100ml volumetric flasks containing 10ml of 10% nitric acid and filled to the mark with distilled water.

3.4 Sampling

A total of eighty three samples comprising fifteen different spices were purchased from different traders in the selected markets. Some of the spices were obtained in the powdered form and others in the raw form as their powdered form were not available. About 9- 11 samples were bought from each market. Table 3.1 below lists the spices, the source and the form in which it was obtained. Some spices were not available at all seven markets

Table 3.1 Spices, Their Sources And The Form In Which They Were Purchased.

	SPICE	SOURCE (MARKET)	FORM
1	THYME	TEMA CENTRAL TAKORADI KANESHIE	SEED POWDERED SEED SEED
2	AIDAN (PREKESE)	TEMA CENTRAL	FRIUT FRIUT
3	ANISE	TEMA TEMA TAKORADI MAKOLA ASAFO KANESHIE CENTRAL	POWDERED POWDERED/SACHET POWDERED POWDERED POWDERED POWDERED POWDERED

Table 3.1 Cont'd Spices, Their Sources And The Form In Which They Were Purchased.

	SPICE	SOURCE (MARKET)	FORM
4	SENEGAL PEPPER (HWENTEA)	ASAFO TEMA TAKORADI CENTRAL KANESHIE AYIGYA	FRIUT FRIUT FRIUT FRIUT FRIUT FRIUT
5	GARLIC	AYIGYA TEMA TAKORADI CENTRAL KANESHIE MAKOLA	CHOPPED BULB BULB BULB POWDERED/ BULB POWDERED
6	GINGER	TEMA TAKORADI MAKOLA ASAFO KANESHIE CENTRAL AYIGYA	POWDERED POWDERED POWDERED POWDERED POWDERED POWDERED POWDERED
7	RED PEPPER	TEMA TAKORADI MAKOLA ASAFO KANESHIE CENTRAL AYIGYA	POWDERED POWDERED POWDERED POWDERED POWDERED POWDERED POWDERED
8	ASHANTI PEPPER SORO WISA	TAKORADI MAKOLA ASAFO CENTRAL AYIGYA	SEED POWDERED SEED SEED SEED

Table 3.1 Cont'd Spices, Their Sources And The Form In Which They Were Purchased.

	SPICE	SOURCE (MARKET)	FORM
9	CLOVES	TEMA	POWDERED
		TEMA	POWDERED/SACHET
		TAKORADI	SEED
		MAKOLA	POWDERED
		ASAFO	POWDERED
		KANESHIE	POWDERED
		CENTRAL	POWDERED
		AYIGYA	POWDERED
10	ROSEMARY	TEMA	POWDERED
		TEMA	POWDERED/SACHET
		TAKORADI	POWDERED
		MAKOLA	POWDERED
		ASAFO	SEED
		KANESHIE	POWDERED
		CENTRAL	POWDERED
		AYIGYA	POWDERED
11	ANISE	TEMA	POWDERED
		TEMA	POWDERED/SACHET
		TAKORADI	POWDERED
		MAKOLA	POWDERED
		ASAFO	POWDERED
		KANESHIE	POWDERED
		CENTRAL	POWDERED
		AYIGYA	POWDERED
12	NUTMEG	TEMA	POWDERED
		TEMA	POWDERED/SACHET
		TAKORADI	POWDERED
		MAKOLA	POWDERED
		ASAFO	POWDERED
		KANESHIE	POWDERED
		CENTRAL	POWDERED
		AYIGYA	POWDERED

Table 3.1 Cont'd Spices, Their Sources And The Form In Which They Were Purchased.

	SPICE	SOURCE (MARKET)	FORM
13	ONION	AYIGYA CENTRAL	BULB BULB
14	BLACK PEPPER	TEMA MAKOLA	POWDERED/ SACHET POWDERED
15	WHITE PEPPER	TEMA AYIGYA MAKOLA	POWDERED/ SACHET POWDERED/ SACHET POWDERED

3.4.1 Sample Preparation

Samples that were in the raw form were washed thoroughly and dried in the oven at a temperature of $100^{o}C$ for between 30 to 60 minutes. Garlic and onion were dried at a temperature of $25^{o}C$ for 24 hours. They were then ground into the powdered form.

3.4.2 Digestion

About 1 g of each powdered sample was weighed into 50mL digestion tube, 1mL H_2O, 2mL HCl, 5mL of 1:1 HNO_3:$HClO_4$ and 2mL H_2SO_4 were added. Samples were allowed to stand for about 20mins at room temperature to enable the foam that formed to settle. They were then heated in a digestion block on a hot plate for about 2 hrs at a temperature of $150^{o}C$. The digested samples were allowed to cool and diluted to 50mL. Blanks were prepared alongside. The samples were then stored for analysis by the mercury analyzer and the atomic absorption spectrophotometer.

3.4.3 Recovery for Mercury

About 1g of each spice sample was weighed into three 50mL digestion tubes. Exactly 25ng of standard mercury chloride solution was added to one of the samples and 50ng was added to a second sample. No mercury was added to the third sample. The mixtures were digested using the digestion procedure used for the samples. The digests were analyzed for total mercury using the Mercury Analyzer. Each determination was carried out at least three times in order to ensure precision.

Percentage recovery for the spiked solutions was calculated using the relation

$$\% \text{ Recovery} = \frac{(\text{Hg in Spiked Solution} - \text{Hg in Non Spiked Solution}) \times 100}{(\text{Hg Added})}$$

3.4.4 Analysis of Samples

The samples were analyzed for mercury (Hg) using the Mercury Analyzer and SOLAAR (S Series 711239 v1.23) Flame Atomic Absorption Spectrometer was used for the determination of copper (Cu), lead (Pb), manganese (Mn), zinc (Zn), cadmium (Cd) and iron (Fe). The wavelengths of 253.7nm for Hg, 324.8nm for Cu, 217.0 for Pb, 251.6nm for Mn, 210.4nm for Zn, 228.8nm for Cd and 259.9nm for Fe were used. The concentrations of the metals were obtained on the spectrometer in mg/L after it was calibrated. Concentration in µg/g was calculated using the following relation.

$$\text{Conc } (\mu g/g) = \frac{\text{mg/L (conc of metal)} \times \text{Total volume (50ml)}}{\text{Weight of Sample (g)}}$$

For Hg: $\text{Conc} = \underline{(\text{Peak Height Sample} - \text{Peak Height Blank})} \times \underline{\text{Conc of Standard}}$

(Peak height of Standard – Peak height Blank)   Weight of Sample

Table 3.2 Operating Parameters for Solaar Flame Atomic Absorption Spectrometer

Metal	Wavelength (nm)	Burner height (mm)	Fuel flow (L/min)
Manganese	210.4	6.2	1.0
Lead	251.6	7.8	1.0
Iron	253.7	6.2	0.9
Copper	217.0	6.2	0.9
Zinc	228.8	6.2	0.9
Cadmium	259.9	8.6	0.9

Background Correction - D2 for all metals.

Flame type – Air-C_2H_2 for all metals.

Signal type – Continuous for all metals.

CHAPTER FOUR

4 RESULTS AND DISCUSSION

4.1 Total Metal Concentrations in the Different spices

The spices were analysed for seven heavy metals. Four of the metals namely zinc (Zn), manganese (Mn), copper (Cu) and iron (Fe) are essential micronutrients and lead (Pb), cadmium (Cd) and mercury (Hg) are toxic metals. A total of 83 spices were bought from seven different markets at Asafo, Takoradi, Kaneshie, Ayigya, Makola and Tema. Nine to twelve spices were purchased from each market.

Validity test for the method used in the analysis of the samples for Hg was established by recovery analysis. The recovery was from 86-90% with an average of 88%. The result is shown in table 4.1.

Table 4.1 Percentage Recovery of Mercury.

Rosemary Sample (g)	Hg added (ng)	Hg found (ng/g)	Recovered (ng/g)	% Recovery
1.025	0	21	0	
1.030	0	21	0	
1.035	25	43	22	88
1.062	25	43	22	88
1.072	50	66	45	90
1.034	50	64	43	86

Results for the metal concentrations in µg/g in all the spices are given in tables 4.2 to 4.8. The range of total Hg concentration for all the spices from all the markets is ND-0.031 (ND- not detectable), that for Cd also ranged from ND- 1.41. The range for Fe is 65.35-221.15, that for

Mn is from 14.93-24.96. Zn ranged from ND -17.70 and finally Cu is 4.22-22.91. Pb was not detected in any of the samples analyzed. Each table gives the values of the heavy metal contents analyzed for each spice purchased from each market.

Table 4.2 Mean Metal Concentrations (µg/g) In Spices From The Various Markets

Spice	Location	Hg µg/g	Cd µg/g	Cu µg/g	Zn µg/g	Fe µg/g	Mn µg/g
Clove	Asafo Market	0.010	N/D	11.14	10.60	151.03	17.41
	Central Market	0.008	1.41	11.47	10.36	149.76	19.76
	Ayigya Market	0.010	0.04	21.11	15.32	152.74	17.92
	Kaneshie Market	0.007	0.69	11.71	5.46	162.06	19.54
	Takoradi Market	0.009	0.80	11.30	12.96	150.61	18.71
	Tema Market	0.004	0.13	7.80	10.32	152.57	22.62
	Makola Market	0.004	N/D	7.81	14.00	152.90	18.49
Nutmeg	Asafo Market	0.006	0.26	15.82	N/D	147.60	18.52
	Central Market	0.002	0.79	20.04	10.41	150.86	17.27
	Ayigya Market	0.005	0.02	18.65	10.37	149.98	20.94
	Kaneshie Market	0.018	0.47	13.00	10.30	148.93	19.68
	Takoradi Market	0.007	0.66	15.93	10.64	153.65	18.03
	Tema Market	0.003	0.64	9.73	17.70	149.21	19.94
	Makola Market	0.016	1.02	6.64	10.62	153.68	16.99
Senegal pepper (Hwentea)	Asafo Market	0.005	0.92	15.58	10.38	160.18	19.39
	Central Market	0.005	N/D	14.07	5.31	152.66	20.49
	Ayigya Market	0.006	0.10	6.24	5.60	175.06	19.58
	Kaneshie Market	0.001	0.50	17.86	10.44	153.18	24.96
	Takoradi Market	0.006	1.29	7.88	10.58	150.67	17.69
	Tema Market	0.006	N/D	17.87	9.53	154.86	19.22

Table 4.2 (Cont'd) Mean Metal Concentrations (µg/g) In Spices From The Various Markets

Spice	Location	Hg µg/g	Cd µg/g	Cu µg/g	Zn µg/g	Fe µg/g	Mn µg/g
Anise	Asafo Market	0.014	0.42	9.66	10.34	149.58	18.62
	Central Market	0.020	0.19	9.11	10.09	150.34	18.77
	Ayigya Market	0.005	N/D	12.81	15.48	151.76	16.50
	Kaneshie Market	0.006	0.21	14.43	10.46	153.58	24.25
	Takoradi Market	0.008	0.95	10.53	16.08	119.91	15.28
	Tema Market	0.007	0.19	13.94	13.49	158.84	19.37
Red Pepper	Asafo Market	0.002	0.16	11.23	13.50	148.75	17.14
	Central Market	0.006	0.69	10.83	8.65	151.48	17.23
	Ayigya Market	0.003	N/D	10.63	17.17	150.56	19.43
	Kaneshie Market	0.007	0.33	10.78	13.18	152.31	18.65
	Takoradi Market	0.003	0.90	7.76	5.24	150.92	19.81
	Tema Market	0.002	0.15	13.94	6.03	148.83	18.08
	Makola Market	0.004	0.60	7.21	10.38	150.64	20.09
Rosemary	Asafo Market	0.026	0.63	8.29	13.01	154.32	18.27
	Central Market	0.018	0.30	5.91	7.73	65.35	21.31
	Ayigya Market	0.017	0.26	9.81	10.42	160.98	19.92
	Kaneshie Market	0.031	0.34	11.70	13.05	153.21	17.99
	Takoradi Market	0.019	0.46	8.79	10.67	150.74	20.41
	Tema Market	0.014	N/D	8.02	13.74	150.68	16.55

Table 4.2 (Cont'd) Mean Metal Concentrations (µg/g) In Spices From The Various Markets

Spice	Location	Hg µg/g	Cd µg/g	Cu µg/g	Zn µg/g	Fe µg/g	Mn µg/g
Ginger	Asafo Market	0.006	0.67	8.86	10.34	150.46	15.88
	Central Market	0.001	N/D	7.71	10.20	148.16	18.11
	Ayigya Market	0.007	0.25	13.82	13.43	152.12	16.03
	Kaneshie Market	0.002	0.96	12.37	10.56	155.82	19.14
	Takoradi Market	0.005	0.42	8.22	13.81	150.69	16.41
	Tema Market	0.006	N/D	8.26	10.68	159.47	19.55
	Makola Market	0.005	0.24	10.22	13.27	153.97	24.73
Grain of Paradise	Asafo Market	0.005	0.10	19.28	10.57	165.43	17.41
(fom wisa)	Central Market	0.005	0.50	19.38	10.43	153.89	19.42
	Ayigya Market	0.009	0.54	22.77	5.38	172.55	19.94
	Kaneshie Market	0.020	0.45	17.78	9.80	152.13	19.02
	Takoradi Market	0.011	0.27	14.76	13.96	152.92	19.75
	Tema Market	0.008	0.32	14.72	12.89	151.25	17.31
Ashanti Pepper	Asafo Market	0.004	N/D	8.17	10.23	154.08	20.94
(soro wisa)	Central Market	0.004	0.67	11.25	13.41	149.51	17.68
	Ayigya Market	0.007	0.55	9.41	5.80	151.09	19.52
	Takoradi Market	0.005	0.70	5.44	10.38	150.13	19.53
	Makola Market	0.004	N/D	9.13	13.54	154.20	17.39

Table 4.2 (Cont'd) Mean Metal Concentrations (µg/g) In Spices From The Various Markets

Spice	Location	Hg µg/g	Cd µg/g	Cu µg/g	Zn µg/g	Fe µg/g	Mn µg/g
White Pepper	Ayigya Market	0.002	0.49	4.22	10.57	155.33	16.13
	Makola Market	0.004	0.29	5.55	N/D	147.49	15.54
	Tema Market	0.001	0.06	6.64	13.16	152.34	19.79
Black Pepper	Tema Market	0.014	1.06	16.18	6.80	152.36	20.91
	Makola Market	0.012	0.58	18.64	9.74	150.26	17.59
Garlic	Central Market	0.006	1.35	11.41	13.80	148.86	18.71
	Ayigya Market	0.001	0.41	7.17	5.19	152.88	19.40
	Kaneshie Market	0.006	0.40	9.59	10.60	153.30	14.93
	Tema Market	0.002	0.60	8.96	10.18	149.52	17.54
	Makola Market	0.003	0.45	4.67	10.31	149.61	15.29
Onion	Central Market	0.003	0.90	7.85	12.83	145.16	18.12
	Ayigya Market	0.005	0.70	7.96	10.29	148.63	17.46
Thyme	Central Market	0.007	0.55	13.58	13.44	150.45	16.56
	Kaneshie Market	0.003	0.50	7.94	16.49	154.00	19.18
	Takoradi Market	0.018	0.60	10.51	10.25	146.27	18.55
	Tema Market	0.015	0.13	8.29	9.90	221.15	15.29
	Makola Market	0.013	0.51	8.82	10.79	154.98	19.57
Aidan (Prekese)	Central Market	0.003	1.4	7.36	15.77	149.39	17.75
	Tema Market	0.006	N/D	8.04	10.52	151.57	20.41

Table 4.3 Calculated Maximum Intake of Metal Levels Obtained By the Different Spices

	MAXIMUM INTAKE OBTAINED (70kg) BODY WEIGHT					
Spice/ RDI	Hg /g	Cd /g	Cu /g	Zn /g	Fe/g	Mn /g
RDI/ mg/day	0.016	0.071	2	15	15	5
Clove	1600	50	95	977	93	221
Nutmeg	889	70	100	847	98	250
Senegal pepper	2667	55	112	1418	86	200
Anise	800	75	140	933	94	206
Red pepper	2286	80	143	874	98	249
Rosemary	615	113	171	1092	93	235
Ginger	2286	74	145	1086	95	202
Grain of paradise	800	131	88	1074	88	251
Ashanti pepper	2286	101	178	1108	93	240
White pepper	4000	145	300	1139	97	253
Black pepper	1143	66	108	1540	98	240
Garlic	2667	53	275	1087	98	258
Onion	3200	80	250	1168	101	276
Thyme	889	118	150	910	68	250
Aidan	2667	51	250	951	99	245

*The calculations for the maximum intake were based on recommended daily intake values from Lenntech recommended daily intake on minerals and vitamins and Joint FAO/WHO expert committee on food additives.

4.1.1 Mercury

Clove was purchased from Asafo, Central, Ayigya, Kaneshie, Takoradi, Tema and Makola markets and the seven heavy metals were determined. Table 4.2 show the results obtained for the various heavy metals.

Mercury (Hg), one of the toxic elements with serious health implications was determined. The peak heights of the blanks were subtracted from the peak heights of all the samples. The precision of the procedure yielded results which agreed to within 5% with an average recovery of 88%. The range for mercury levels was from 0.004 to 0.010 µg/g with Asafo and Ayigya markets both recording 0.010 µg/g and Makola and Tema markets both recording the least level of 0.004 µg/g. It was observed that all the levels were low. Considering the value presented in table 4.3 for clove, the amount stated is not likely to be consumed. The recommended daily intake of Hg is 0.016 mg/day for a 70kg body weight according to Joint FAO/WHO expert committee on food additives. The market that recorded the next highest level of Hg in clove was in Takoradi followed by Central and Kaneshie.

Nutmeg was purchased from seven markets namely Asafo, Ayigya, Kaneshie, Tema, Makola, Takoradi and Central market. From table 4.2 the mercury levels in nutmeg ranged from 0.002 to 0.018 µg/g. The highest level was recorded in Kaneshie. Comparing the levels obtained, it was realized that some of levels were as low as 0.002 µg/g while others were as high as 0.018 µg/g. This indicates that the nutmeg spice came from different sources. Five markets recorded levels lower than 0.010 µg/g which indicate that soils in which the nutmeg were cultivated had low levels of Hg or Hg was not in the available form. This spice was purchased from the various markets in the powdered form. If this spice was adulterated, additives added were low in Hg.

Levels of Hg in nutmeg from the markets occurred, in the order: Kaneshie market > Makola > Takoradi > Asafo > Ayigya > Central market.

Senegal pepper (hwentea) was obtained from Asafo, Central, Ayigya, Kaneshie, Takoradi and Tema market. The Hg levels recorded for Senegal pepper were low as compared to those obtained in nutmeg. The total mercury concentration ranged from 0.001 to 0.006 µg/g. Sample from Tema, Takoradi and Ayigya recorded the highest level of 0.006 µg/g. Asafo and Central markets recorded the next highest level of 0.005 µg/g and Kaneshie market recorded the least mercury level in Senegal pepper.

Anise spices were obtained from Asafo, Central, Ayigya, Kaneshie, Takoradi and Tema. Mercury levels in anise ranged from 0.005 to 0.020 µg/g. Markets from Accra recorded values close to each other. This suggests that samples from Accra and Kumasi might have come from similar sources whiles that from Ayigya may come from a different source. The levels of Hg in anise ranged in the order: Central > Asafo > Takoradi > Tema > Kaneshie > Ayigya market in that order.

Red pepper was purchased from Asafo, Central, Ayigya, Kaneshie, Takoradi Makola and Tema. Hg level obtained ranged from 0.002 to 0.007 µg/g. The levels were low. Since it was purchased in the powdered form from the various markets if additives were added, it did not affect the levels of Hg. The levels of Hg in red pepper ranged in the order: Kaneshie > Central > Makola > Takoradi > Ayigya > Asafo > Tema markets.

The rosemary spice was purchased from Central, Asafo, Takoradi, Tema, Kaneshie and Ayigya markets. Table 4.2 shows the range of mercury in rosemary to be from 0.014 to 0.031 µg/g. The levels obtained for rosemary were high as compared to Hg levels in red pepper. These levels recorded could be as a result of the presence of some mercury in the soils where the rosemary plants were cultivated or if additives were added they also contained some amount of mercury. The levels of Hg in rosemary showed that Kaneshie > Asafo > Takoradi > Central > Ayigya > Tema.

Ginger was purchased from Asafo, Central, Ayigya, Kaneshie, Takoradi, Makola and Tema markets. Table 4.2 shows the levels of heavy metals recorded by ginger. Mercury levels ranged from 0.001 to 0.007 µg/g. The levels in ginger from the various markets were all low. Asafo and Tema both recorded 0.006 µg/g, Takoradi and Makola both 0.005 µg/g and the others varying levels. The highest level of Hg in ginger was recorded at Ayigya market followed by Tema, Asafo, Makola, Takoradi, Kaneshie and Central markets in that order.

Grain of paradise (famu wisa) was purchased from Asafo, Central, Ayigya, Kaneshie, Takoradi Makola and Tema. Table 4.2 shows the Hg concentrations analysed in grain of paradise. Mercury levels in grain of paradise ranged from 0.005 to 0.020 µg/g. Kaneshie market recorded the highest mercury level in grain of paradise followed by Takoradi, Ayigya, Tema, Central and Asafo markets.

Ashanti pepper (soro wisa) was purchased from Asafo, Central, Ayigya, Takoradi and Makola were markets. Table 4.2 shows the various metal concentrations in Ashanti pepper the various markets. Ashanti pepper recorded very low levels of mercury which ranged from 0.004 to 0.007

μg/g. Ayigya market recorded the highest level of Hg of all the markets from Kumasi. Asafo, Makola and Central all recorded a level of 0.004 μg/g while Takoradi recorded the lowest.

White pepper was obtained from Ayigya, Makola and Tema markets. From table 4.2, the mercury levels obtained by white pepper ranged from 0.001 to 0.004 μg/g. White pepper was one of the spices that recorded some of the lowest levels of mercury in this study. Makola recorded the highest level of Hg followed by Ayigya and Tema markets.

Black pepper was purchased from Tema and Makola markets. Table 4.2 shows the results of Hg that were determined in black pepper. The levels of mercury in black pepper ranged from 0.012 to 0.014 μg/g. These levels are high as compared to those in white pepper. These levels may be due to the addition of the fruit cover in the preparation of black pepper as; in the preparation of white pepper the fruit cover is excluded. Tema market recorded the highest and Makola market recorded the least.

Garlic was obtained from Central, Ayigya, Kaneshie, Tema and Makola markets. Table 4.2 shows the Hg concentrations determined in garlic from the various markets. The total Hg concentrations in garlic ranged from 0.001 to 0.006 μg/g. Garlic was also one of the spices which recorded some of the lowest levels of Hg in this study. Kaneshie and Central markets recorded the highest level of hg followed by Makola, Tema and Ayigya markets.

Onion was purchased from Central and Ayigya markets. From table 4.2, mercury levels ranged from 0.003 to 0.005 μg/g. These levels were pretty low as compared to levels those present in black pepper and rosemary. Ayigya market had the highest level and central market the least.

Thyme was purchased from Central, Makola, Kaneshie, Takoradi and Tema. Table 4.2 shows the results of the analysis of the heavy metals in thyme. Mercury levels in thyme ranged from 0.003 to 0.018 µg/g. These levels as compared to those in garlic or onion are a little high. Samples of thyme from the Takoradi market had the highest level followed by Tema, Makola, Central and Kaneshie markets in that order.

Aidan (Prekese) was purchased from Central in Kumasi and Tema markets. Table 4.2 shows the metal concentrations obtained from the analysis of aidan. Mercury levels in aidan ranged from 0.003 to 0.006 µg/g which were also low. Tema market recorded the highest followed by Central market.

The levels of mercury recorded by almost all the spices from the various markets were all low which suggests that the soils in which these spices were cultivated were low in Hg or the Hg was not in the available form for absorption. The range was from 0.001 to 0.031 µg/g. The recommended daily intake of Hg is 0.016 mg/day for a 70kg body weight according to Joint FAO/WHO expert committee on food additives. Considering the amounts presented in table 4.3, the recommended daily intake for Hg is unlikely to be exceeded. Considering the toxic nature of mercury it is satisfying to know that the level of this metal in the spices from the various markets were all safe for consumption.

Though the levels of Hg were low studies done elsewhere have shown that mercury can accumulate in plants and in soils where the levels of Hg are low (Matt, 1972). A study by Matt (1972) on mercury uptake from soil by some edible plants showed levels ranging from 0.387 to

2.447mg/kg. Another study done by Shaw and Panigrahi (1986) on the pattern of distribution of mercury in the tissues of some plant species collected around a chlor-alkali factory in India showed that different plants accumulated different levels of mercury in their tissues. A significant correlation was noted between the mercury concentration of the soil and the plant tissues and between different tissues.

4.1.2 Cadmium

Of all the toxic metals released in large quantities into the environment cadmium (Cd) generally is regarded as the mostly likely to accumulate in the human food chain (Johannesson, 2002). Cadmium in clove ranged from ND to 1.41 µg/g. The highest level of Cd recorded in this study was in clove from the Central market. The level of Cd found in clove was in the order from Central > Takoradi > Kaneshie > Tema > Ayigya markets. There was no Cd in cloves from Asafo and Makola markets.

The levels of Cd obtained for nutmeg were quite high with the range from 0.02 to 1.02 µg/g. Apart from the Ayigya market which recorded the least level of 0.02 µg/g all the others were all above 0.20 µg/g. These high levels may have been caused by additives since it was purchased in the powdered form from the various markets or from the soil in which it was cultivated. Makola market recorded the highest level of Cd in nutmeg followed by Kumasi Central, Takoradi, Tema, Kaneshie Asafo and Ayigya markets.

The Cd levels in Senegal pepper ranged from N/D to 1.29 µg/g. Cd was not detected in Senegal pepper from Central and Tema markets. Takoradi market recorded the highest followed by

Asafo, Kaneshie and then Ayigya. The levels recorded from markets from the same area varied as some recorded high levels whiles some were non detectable.

Cd levels in anise ranged from N/D to 0.95 µg/g. The levels obtained varied with some recording high levels and others recording low levels. Cadmium was not detected in anise from Ayigya market from Kumasi while Central and Asafo markets also from Kumasi had high values. Tema and Kaneshie showed a similar trend suggesting that this spice may have originated from similar environments. The anise samples were purchased from the various markets in the powdered form. The variation in the results show that, there is an indication of contamination from additives. Takoradi market recorded the highest level followed by Asafo, Kaneshie, Tema, Central and Ayigya markets.

Cadmium levels in red pepper from the various markets ranged from N/D to 0.90 µg/g. Cd was not detected in one of the samples but the other levels obtained suggests that red pepper plants were cultivated in soils a little high in Cd content. Since most of the powdered red pepper on the markets are adulterated, if additives were added they might have contributed to the levels. Levels of Cd in red pepper from Takoradi > Central > Makola > Asafo > Kaneshie > Tema > Ayigya.

The cadmium levels in rosemary from the various markets ranged from N/D to 0.63 µg/g. These levels were not so high compared to that of red pepper. The highest level of Cd in rosemary was found in Asafo followed by Takoradi, Kaneshie, Central and Ayigya market. Cadmium was not detected in rosemary from Tema market.

Cadmium levels in ginger ranged from N/D to 0.96 µg/g. The results obtained varied as some markets had non detectable levels while others had levels as high as 0.96 µg/g. Considering samples from the different markets in Kumasi and Accra it was noticed that the results obtained were varying which suggests that the samples did not come from similar sources. These levels indicate that some amount of Cd were in the soils where the ginger plants were cultivated. Cd in ginger from Kaneshie market > Asafo > Takoradi > Makola. Cd was not detected in Central and Tema markets.

The levels of Cd in the Grain of Paradise from the various markets ranged from 0.10 to 0.54 µg/g. These levels are rather low as compared to the levels obtained in ginger. The highest level was recorded in samples from Ayigya market and the least from Asafo all in Kumasi. The market that recorded the next highest level of Cd was Central market followed by Kaneshie, Tema and Takoradi markets.

Cadmium levels in Ashanti pepper ranged from N/D to 0.71 µg/g. Cd was not detected in Ashanti pepper from Makola and Asafo whiles Central, Ayigya and Takoradi recorded appreciable levels. Takoradi recorded the highest level of Cd in Ashanti pepper.

Cd levels in white pepper ranged from 0.06 to 0.49 µg/g. White pepper from Tema recorded the second lowest level of Cd in this study. The levels obtained in the spices from Makola and Ayigya markets were quite high as compared to that from Tema. The highest level of Cd in white pepper was recorded at Ayigya market.

The levels of cadmium in black pepper ranged from 0.58 to 1.06 µg/g. The levels obtained for samples from Tema and Makola markets were high enough and this could be due to the fact that the black pepper plants were cultivated in soils with some amount of Cd. The high levels could also be due to the addition of the fruit cover in the preparation of black pepper. Tema market recorded the highest level while Makola market recorded the least.

The levels of cadmium obtained in garlic ranged from 0.40 to 1.35 µg/g. Garlic was relatively rich in Cd concentrations as compared to some of the levels obtained by some of the spices. Garlic from Central market recorded the third highest Cd level in this study. These levels are as a result of the garlic plants being cultivated in soils with some levels of Cd or the garlic plants can accumulate Cd. Taking into consideration the maximum amount obtained in table 4.3, the recommended intake can be exceeded by people who consume large amounts of garlic, but for people who consume small amounts, the recommended intake will not be exceeded. The order of accumulation of Cd in garlic is in the order: Central market > Tema > Makola > Ayigya > Kaneshie market.

Cd levels recorded by onions from Central and Ayigya markets were high. The levels ranged from 0.70 to 0.90 µg/g. Central market obtained the highest level and Ayigya market the lowest level of Cd. From table 4.3, the maximum level of 0.90 µg/g gave an amount of 80g. Taking into consideration the quantity of onion some people can consume, these amounts can be exceeded.

Cadmium levels in thyme ranged from 0.13 to 0.60 µg/g. Takoradi obtained the highest level of Cd. Considering the quantity recorded in table 4.3 and the amount usually consumed, Cd levels in thyme may not exceed the recommended intake. The next highest level of Cd was obtained by Central followed by Makola, Kaneshie and Tema markets.

The Cd levels obtained in prekese ranged from N/D to 1.40 µg/g. The second highest level of Cd in this study was recorded in prekese from Central market. From table 4.3 about 51g of prekese would have to be consumed to exceed the maximum intake considering the maximum level. The whole fruit is usually used in cooking and the quantity used is dependent on the individual. For prekese that contains such levels, if large quantities are consumed then there is a probability that the daily intake can be exceeded.

The cadmium levels for all the spices ranged from ND to 1.41 µg/g. From table 4.3, the maximum intake obtained for Cd for the various spices were not likely to be consumed in a day to exceed recommended intake of 0.071mg/day for 70kg body weight with the exception of some spices like onions, garlic, prekese and red pepper. If these spices are consumed in large quantities there is a probability that the recommended intake will be exceeded. Cadmium is a toxic metal and with its accumulative nature poses serious health threat to humans. Thus the low levels obtained makes it satisfying to know that spices from these markets are healthy.

A study by Nnorom *et al.* (2007) on trace metals in bouillon cubes and some natural spices such as thyme reported levels for Cd ranging from ND to 5.05 mg/kg which is far above levels recorded for this study. Another research work done in Saudi Arabia on heavy metal levels of some common spices also showed high levels of 1.25 to 3.05 mg/kg for Cd (Al-Eed *et al*., 2002). The results in this study suggest that some of the spices analysed were cultivated in soil low in Cd while others were cultivated in soils with appreciable levels of cadmium.

Chizzola *et al.* (2003) reported that contents of heavy metals and metallic micronutrients were generally within the usual range in herbs, spices and medicinal plants from Austria. They concluded that the contamination levels with Cd and Pb could be described as low. Cadmium levels can be high in plants due to the fact that Cd is readily transported from soil to the upper part of plants especially, edible parts (Mengel and Kirkby, 1982). Its transfer from soil to the upper parts of crops is significantly greater than other heavy metals with the exception of zinc.

4.1.3 Copper

The normal levels of Copper (Cu) found in plants have a range 3-40 µg/g (Benton, 1998). Copper levels in clove ranged from 7.80 to 21.11 µg/g. Most of the levels from the various markets were in the range of 11.14-11.80 µg/g. Cu level in Clove from Ayigya market suggests that, of the three markets from Kumasi, it came from an area rich in copper than where the others came from. The highest level of Cu in clove was from Ayigya market with the least from Tema market. The next highest level was found in clove from Kaneshie followed by Central, Takoradi, Asafo and Makola in that order.

Copper levels in nutmeg from the various markets varied from 6.64 to 20.04 µg/g. Comparing the levels of Cu in nutmeg to that in clove it was realized that the differences between the levels were small. Ozkutlu *et al*., (2006) reported the range of copper to be between 3 to 11 µg/g with the highest level in nutmeg and black pepper. Comparing their values to the values obtained for nutmeg in this study, the values in this study were higher. It was also noticed that levels from markets in Kumasi were high as compared to those from Accra with the exception of Kaneshie market. Central market recorded the highest level followed by Ayigya, Takoradi, Asafo, Kaneshie, Tema and Makola.

The levels of Cu in Senegal pepper ranged from 6.24 to 17.87 µg/g. The least amount of Cu were observed in samples from Ayigya and the highest from Tema market. Kaneshie recorded the second highest followed by Asafo and Central markets in Kumasi. Considering the levels obtained it could be conjectured that samples from Takoradi and Ayigya were not cultivated in soils rich with Cu.

The Cu in anise ranged from 9.11 to 14.43 µg/g. Asafo and Central market in Kumasi recorded levels that were close to each other. Anise from Tema and Kaneshie also recorded close values. Copper from Ayigya market in Kumasi recorded far higher amount than the other two markets. Kaneshie obtained the highest level followed by Tema, Ayigya, Takoradi, Asafo and Central markets.

The copper in red pepper ranged from 7.21 to 13.94 µg/g. Three markets recorded values within the range of 10.63-10.83 µg/g while two markets obtained values below 10.00 µg/g. When these levels are compared to the other Cu levels in some other spices analysed in the study, the levels are low. Red pepper from Tema obtained the highest level followed by Asafo, Central, Kaneshie, Ayigya, Takoradi and Makola.

The copper levels obtained for rosemary ranged from 5.91 to 11.70 µg/g. Rosemary recorded some of the lowest levels of Cu in this study. The reason could be that the rosemary plants cannot accumulate Cu or they were cultivated in soils low in Cu content. Kaneshie market recorded the highest level of Cu in rosemary followed by Ayigya, Takoradi, Asafo, Tema and Central markets.

The levels of Cu studied in ginger varied from 7.71 to 13.82 µg/g. Ayigya market recorded the highest level of Cu while the Central market recorded the least. The Asafo and Central markets from Kumasi recorded low levels whilst Ayigya from Kumasi recorded a high level. Kaneshie and Makola from Accra recorded high levels while Tema obtained a low value. The markets that obtained the next highest levels were Makola and then Asafo followed by Tema, Takoradi and Central markets.

Copper levels recorded by grain of paradise ranged from 14.72 to 22.77 µg/g. The highest copper level obtained in this study was recorded by grain of paradise. Grain of paradise is naturally rich in Cu and the levels obtained in this study confirm it. Ayigya market obtained the highest level. Levels of Cu in grain of paradise from Ayigya market > central > Asafo > Kaneshie > Takoradi > Tema market.

The copper in Ashanti pepper from the various markets were low. Apart from the Central market which recorded a level above 11 µg/g, all the others recorded levels below 10 µg/g. The levels ranged from 5.44 to 11.25 µg/g. Central market obtained the highest level of Cu in Ashanti pepper followed by Ayigya, Makola, Asafo and Takoradi markets.

The Cu levels in white pepper ranged from 4.22 to 6.64 µg/g. Some of the lowest levels of Cu in this analysis were obtained by white pepper from the various markets. The lowest Cu level in this study was also recorded in white pepper from Ayigya. The highest level of Cu was obtained in white pepper from Tema, followed by Makola and Ayigya markets respectively.

Copper levels obtained for black pepper were all high. The levels ranged from 16.18 to 18.64 µg/g. This is in agreement with that found by Ozkutlu *et al.*, (2006) in nutmeg and black pepper. Comparing their values to the values of this study black pepper recorded higher values. Though white and black peppers come from or are obtained the same plant, the high levels obtained might have come from the fruit cover that is added in the preparation. Also, the white and black pepper on the market might have come from different plants. This means that the fruit cover might contain some amount of Cu. Makola market recorded the highest level and Tema market recorded the least level of Cu in white pepper.

The levels of Cu in garlic ranged from 4.67 to 11.41 µg/g. Apart from the samples from the Central market which gave 11.41 µg/g, all the other markets recorded levels below 10 µg/g. This may suggest that garlic from the other markets came from sources low in Cu or the garlic plant cannot accumulate much Cu. Central market > Kaneshie > Tema > Ayigya > Makola market in terms of Cu levels recorded.

Copper levels in onion ranged from 7.85 to 7.96 µg/g. The levels were on the lower side when compared to the normal range of 3-40 µg/g. This could be as a result of the onion plants coming from sources low in Cu. Ayigya market obtained the highest level while Central market recorded the lowest level.

With thyme, copper ranged from 7.94 to 13.58 µg/g. Apart from Central and Takoradi markets all the other markets levels below 10 µg/g. The levels of Cu in thyme from Central > Takoradi > Makola > Tema > Kaneshie market in that order.

The levels of copper in prekese ranged from 7.36 to 8.04 µg/g. Tema market recorded the highest level and Central market the lowest level. The levels obtained were on the lower side of the normal range which is 3 to 40 µg/g.

Ozkutlu *et al.* (2006) reported the range of copper to be between 3 to 11 µg/g with the highest level in nutmeg and black pepper. The values in this study ranged from 4.22 to 22.77 µg/g. Comparing the values of this study to those obtained by Ozkutlu *et al.* (2006) it is noticed that, the levels of this study are higher. Nutmeg and black pepper in their study recorded the highest levels. Nutmeg and black pepper in this study also recorded high levels. They concluded that the levels of copper obtained were far below the recommended levels of intake. A study by Divrikli *et al.* (2005) recorded values of copper in the range between 3.8 and 35.4 µg/g which were within the recommended daily intake of Cu of 2mg/day for 70kg body weight (www.lenntech.com). Taking into consideration the maximum intake calculated with the maximum levels presented for this study in table 4.3, exposure to Cu through consumption of these spices is not significant.

Most of the copper levels obtained in this study were low if compared to the normal range of 3-40 µg/g. Consuming these spices may not pose any health problems as copper is an essential micronutrient which when consumed has some important physiological functions in the body. Copper helps the body to use iron. It is also important for nerve function and bone growth. It also to helps the body to use sugar and protect cell membranes form being destroyed by free radicals (ATSDR, 2004).

4.1.4 Zinc

Zinc (Zn) is an essential element found virtually in every cell of the human body and plays vital roles in the healthy development and growth of the human body. Zinc plays an important role in growth and has recognized action on more than 300 enzymes by participating in their catalytic and regulatory action (Nnorom *et al*., 2007).

The normal levels of zinc range from 15 to 150 µg/g in most crops and pastures (Benton, 1998). Considering the levels of Zn recorded in clove which ranged from 5.46 to 15.32 µg/g, the levels can be considered to be low. The highest level of Zn was recorded in clove from Ayigya market followed by Makola, Takoradi, Asafo, Central Tema and Kaneshie

Levels of zinc in nutmeg ranged from N/D to 17.70 µg/g. The highest level of Zn in this study was recorded by nutmeg from Tema market. With the exception of nutmeg from Asafo in which Zn was not detected, most of the values were within 10.30-10.64 µg/g range. The levels of Zn in nutmeg follows in the order: Tema market > Takoradi > Makola > Central > Ayigya > Kaneshie. Zinc was not detected in nutmeg from Asafo.

Zn levels recorded in Senegal pepper were low as compared to those obtained in nutmeg and clove. The range is from 5.31 to 10.58 µg/g. All the markets recorded levels below the least normal level plants should have which is 15 µg/g (Benton, 1998). Divrikli *et al.* (2005) reported levels of zinc in spices and herbal plants in the range of 5.2-83.7 µg/g. These levels were high compared to the levels obtained in this study. Some of the levels obtained in this study are within the range of their study indicating that zinc levels vary widely depending on the type of plant and the area where it is cultivated. The levels of Zn in Senegal pepper from Takoradi > Kaneshie > Asafo > Tema > Ayigya > Central market.

Anise recorded a range of 10.09-16.08 µg/g for Zn. Three markets recorded a range of 10.09 to 10.48 µg/g and the other markets recorded levels above 12.81 µg/g. Some of these levels are low and are below the least level of Zn a plant should have. Comparing the results to the normal range it is realized that the levels recorded by anise are on the lower side of the normal range. Ayigya market recorded the second highest after Takoradi. Tema obtained the third highest followed by Kaneshie. Asafo and Central markets obtained the least levels of Zn in anise.

The Zn levels obtained in red pepper from the various markets varied from 5.24 to 17.17 µg/g. Red pepper from Ayigya recorded the second highest level of Zn in this study. All the markets had their zinc levels below 15 µg/g, the least within the normal range for Zn that a plant should have except for red pepper from Ayigya market. Zn levels in red pepper from the various markets was in the order: Ayigya > Asafo > Kaneshie > Makola > Central > Tema > Takoradi markets.

The levels of Zn in rosemary from the various markets ranged from 7.73 to 13.74 µg/g. The highest level of Zn was obtained in rosemary from Tema market and the least from Central market. These levels obtained were higher than some of the levels recorded by some of the spices discussed. The next highest level was found in Kaneshie followed by Asafo, Takoradi and Ayigya market in that order.

Zn levels in ginger ranged from 10.20 to 13.81 µg/g. Comparing the general levels of Zn obtained in this study, the levels recorded by ginger are quite high as most were above 10.20 µg/g though they were all below the normal range of 15 to 150 µg/g. The Takoradi market recorded the highest level of Zn in ginger. The second highest level was obtained by Ayigya followed by Makola, Tema, Kaneshie, Asafo and central markets.

The levels of zinc in Grain of paradise ranged from 5.38 to 13.96 µg/g. All the markets had their levels below the least level of the normal range which is 15-150 µg/g. Ayigya market recorded a level that was far lower than those obtained by the two other markets from Kumasi. Grain of paradise from Kaneshie also recorded a levels lower than that from Tema. Takoradi market had the highest level of Zn in Grain of paradise followed by Tema, Asafo, Central, Kaneshie and Ayigya market.

The range of zinc in Ashanti pepper was from 5.80 to 13.54 µg/g. Comparing the levels of Zn obtained by markets from Kumasi, Ayigya market recorded the lowest. Makola market in Accra recorded the highest level. Kumasi Central market recorded the second highest followed by Takoradi then Asafo and Ayigya markets.

White pepper had its Zn levels ranging from N/D to 13.61 µg/g. The levels recorded by Tema and Ayigya markets were high in this study. Tema market recorded the highest concentration of Zn and was followed by Ayigya market. Zn was not detected in white pepper from Makola.

Zn levels obtained in black pepper ranged from 6.8 to 9.74 µg/g. The levels were below the least level for a normal range in plant. White pepper recorded levels higher than that of black pepper for Zn contrary to what was observed for the other metals. Since they come from the same plant, this could be due to the fact that the fruit cover used in the preparation did not contain any appreciable level of Zn or both white and black pepper came from different plants. Makola market recorded the highest level and Tema market the least.

Levels of zinc in garlic from the various markets ranged between 5.19 and 13.80 µg/g. The Central market had the highest level Zn with Ayigya recording the least level. Comparing the levels of Zn in garlic from the two markets in Kumasi, it could be deduced that they originated from different sources. Kaneshie market obtained the next highest level of Zn followed by Makola and Tema markets.

The levels of zinc obtained in onion ranged from 10.29 to 12.83 µg/g. The highest level of Zn was found in Ayigya and the least in the Central market. The levels obtained were low when compared to the normal levels in plants.

The levels of zinc in thyme were all high except that from the Tema market ranging from 9.90 to 16.49 µg/g. Levels of zinc in thyme from Kaneshie > Central > Makola > Takoradi > Tema market in that order.

The levels of Zn in prekese ranged from10.52 to 15.77 µg/g. The highest level was obtained by Central market while Tema market recorded the least. The levels obtained were high compared to other levels of Zn obtained in other spices in this study.

A study by Nnorom *et al.* (2007) recorded a range of 1.00 to 48.10 µg/g for zinc. This indicates how low Zn levels can be in plants. They concluded that their results were high. Comparing their levels to that of this study, it can be said that the levels in this study were not high. Another study on micronutrients in spices showed levels of zinc ranging from 4 to 25 µg/g (Ozkutlu *et al.*, 2006). Their levels were almost in the same range as the levels in this study. They concluded that the levels recorded were within the low range of normal plant zinc. A study by Basgel and Erdemoglu (2005) found that micronutrient concentrations were generally higher in leaves than in the other parts above the ground.

The levels of zinc recorded for all the various spices ranged from ND to 17.70 µg/g. The low levels could be attributed to soils in which these spices were cultivated. The low levels could be explained by the fact that zinc is immobile in soil thus making it quite difficult for plants to absorb and transport. It is also not readily translocated within the plant especially in areas where the concentrations are low. Considering the maximum intakes presented in table 4.3, the amounts obtained for the various spices are not likely to be consumed. The recommended daily intake of 15mg/day for 70kg body weight (www.lenntech.com) is unlikely to be exceeded.

4.1.5 Iron

Plants require iron (Fe) for the production of chlorophyll which is needed for manufacture of plants food. It is also needed as a component of many enzymes associated with energy transfer, nitrogen reduction and fixation and liginin formation (www.echochemmicronutrient). The levels of iron in cloves from the different markets were in the range of 149.76 to 162.02 µg/g. The levels were all high in this study. The highest level of Fe was found in clove from Kaneshie market and the least in clove from the Central market. The next highest level of iron in clove was from Makola followed by Kaneshie, Tema, Asafo and Takoradi.

Levels of iron in nutmeg varied with a range of 147.60 to 153.68 µg/g. Most of the levels were within the range of 147 to 149 µg/g with the rest above 150 µg/g. Considering the levels of Fe obtained though lower than those recorded for clove, the levels are generally high. Nutmeg from Makola recorded the highest Fe level with the least from Asafo market. Takoradi market recorded the second highest level followed by Central market, Ayigya market, Tema market, Kaneshie and Asafo market.

The levels of iron in Senegal pepper ranged from 150.67 to 175.06 µg/g. The second highest level of Fe in this study was recorded by Senegal pepper from Ayigya market. The levels of Fe obtained for Senegal pepper were all high and consistent which suggest that plants have the ability to absorb and retain iron. The levels of iron in Senegal pepper from Ayigya > Asafo > Tema > Kaneshie > central > Takoradi market.

The levels of iron recorded by anise ranged from 119.91 to 158.84 µg/g. Takoradi recorded the second lowest level in this study that is 119.91 µg/g. This level as compared to other iron levels in anise from the different markets is low. The levels recorded by all the other markets were high and within the normal plant range of 20-300 µg/g. Tema market recorded the highest level followed by Kaneshie, Ayigya, Central, Asafo and Takoradi in that order.

The iron levels in red pepper ranged from 148.79 to 152.31 µg/g with Kaneshie market recording the highest level. The differences between the levels recorded by the markets are close. Additives added if any might have contributed to the levels since it was purchased in the powdered form. The market that obtained the next highest level of iron is Central market followed by Takoradi, Makola, Ayigya, Tema and Asafo markets respectively.

The levels of iron in rosemary ranged from 65.35 to 160.98 µg/g. All the levels obtained were high with the exception of rosemary from Central market. The lowest level of iron recorded in this study was in rosemary from Central market. Rosemary is a plant that is naturally rich in iron but that from central market was very low. This could be as result of this plant growing in a soil that was low in iron. The others however recorded high levels. Level of iron in rosemary was in the order Ayigya > Asafo > Kaneshie > Takoradi > Tema > Central market.

Iron recorded levels that ranged from 148.16 to 159.47 µg/g in ginger. Since ginger was purchased in the powdered form additives possibly added, may have contributed to the high values. The highest level of iron was from Tema followed by Kaneshie, Makola, Ayigya, Takoradi, Asafo and Central markets in that order.

Levels of iron in grain of paradise ranged from 151.25 to 172.55 µg/g. Markets from Kumasi recorded the highest levels with Ayigya market obtaining the highest. Kaneshie and Tema recorded the lowest levels.

Iron levels in Ashanti pepper ranged from 149.51 to 154.20 µg/g. Plants are rich in iron and all levels from the various markets obtained in this study confirms it. Makola market obtained the highest level of Fe in Ashanti pepper while Central market recorded the least level. The next highest level of Fe was obtained in Ashanti pepper from Asafo followed by Ayigya and Takoradi markets respectively.

Iron levels in white pepper ranged from 147.49 to 155.33 µg/g. The levels obtained were high as compared to the levels obtained for the other metals in white pepper. Ayigya market recorded the highest level and Makola market recorded the lowest. Tema market recorded the next highest level of Fe.

Iron levels recorded by black pepper ranged from 150.26 to 152.36 µg/g. Tema market obtained the highest level followed by Makola market. The levels recorded by both markets were all high. Comparing the levels in white pepper to levels in black pepper, their levels were not so different from each other. This suggests that the fruit cover added in the preparation of black pepper did not contain any appreciable levels of Fe. There is also the probability that the white pepper and black pepper on the markets might have come from different plants.

Iron levels in garlic ranged from 148.86 to 153.30 µg/g. Kaneshie market recorded the highest level of Fe followed by Ayigya, Makola, Tema and Central markets. The levels obtained in garlic from the various markets were all within the normal range of plant Fe.

Iron in onions from the markets ranged from 145.16 to 148.63 µg/g. The Central market recorded the highest level and Ayigya market the least. The levels are high and are of normal Fe range in plant.

The levels of iron in thyme ranged from 146.27 to 221.15 µg/g. From literature the thyme plant is an excellent source of Fe. The levels obtained were not that high except that obtained from Tema which recorded the highest level in this study. A study by Nnorom *et al.,* (2007) reported thyme having the highest level of Fe, 419.05 µg/g. This confirms that naturally the thyme plant can accumulate Fe. The levels of Fe in thyme was of the order: Tema market > Makola > Kaneshie > Central > Takoradi market.

Iron levels recorded in prekese ranged from 149.39 to 151.57 µg/g. Tema market had the highest level followed by the Central market.

The results obtained for iron in this study were all high except in two spices. The normal level of Fe in plants ranges from 20 to 300 µg/g (Benton, 1998) and obtained in this study were all within the normal range. A study by Nnorom *et a.,* (2007) reported results of some of spices recording iron levels as high as 419.05 µg/g. Plants normally contain high levels of iron and the relatively high levels in this study confirm the results of Onianwa *et al.,* (1999). Levels of iron are expected to be high in plant derived products which was shown by all the samples in this study. Thyme from Tema recorded the highest level of Fe, of 221.15 µg/g while rosemary from Central Market recorded the lowest level of 65.35 µg/g. The maximum Fe intakes obtained in this study (table 4.3) show that the amounts to be consumed are not likely to exceed the recommended daily intake of 15 mg/day for a 70kg body weight (www.lenntech.com). Thus the exposure to Fe through consumption of these spices is insignificant.

Iron is an integral part of many proteins and enzymes that maintain good health. In humans, iron is an essential component of proteins involved in oxygen transport (Dallman 1986; Institute of

Medicine, 2001). It is also essential for the regulation of cell growth and differentiation (Bothwell *et al*., 1979; Andrews, 1999).

4.1.6 Manganese

Manganese (Mn) is an important micronutrient needed by the body for certain processes such as helping to maintain sex hormone production, metabolism of Vitamins B1 and E, activation of various enzymes important for proper digestion to make the body healthy.

Manganese in clove from all the markets was found to have levels ranging from 17.41 to 22.62 µg/g. Considering the highest and the least Mn levels obtained, it was observed that the levels were not high and rather were lower than the results obtained by Ozkutlu *et al.*, (2006) which reported Mn levels within a range of 10 to 355 µg/g. The highest level was found in clove which is an excellent source of Mn. The clove spice did not record high levels in this study. This could be due to the fact that the soils in which the clove plants were cultivated were not that rich in Mn resulting in low accumulation. The normal range of Mn is 15- 150 µg/g (Benton, 1998). Tema market recorded the highest. The levels of Mn in clove was of the order Central market > Kaneshie > Takoradi > Makola > Ayigya > Asafo market.

The levels of manganese in nutmeg were all high comparing them to that of the other spices. The range is between 16.99 to 20.94 µg/g and above the lowest level of Mn a plant should contain (Benton, 1998). Ayigya market recorded the highest Mn level in nutmeg and Makola market recorded the least. The Tema market recorded the second highest Mn content followed by Kaneshie, Asafo, Takoradi, Central and Makola markets

The manganese in Senegal pepper ranged from 17.69 to 24.96 µg/g. The highest level of Mn in this study pertained to Senegal pepper from Kaneshie. Comparing Mn from the various markets, though the levels recorded were high they are lower when compared to the range of Mn 15- 150 µg/g obtained by Benton (1998). The Kaneshie market recorded the highest level followed by Central, Ayigya, Asafo, Tema and Takoradi markets in that order.

The Mn levels obtained for anise spice from the various markets ranged from 15.28 to 24.25 µg/g. The third highest level of Mn in this study was recorded by anise from Kaneshie. Comparing the levels of Mn obtained for the various markets, the levels were all high. Mn levels in anise was of the order: Kaneshie > Tema > Central > Asafo > Ayigya > Takoradi market.

The levels of Mn recorded by red pepper were all high ranging from 17.14 to 20.09 µg/g. However the Mn in red pepper obtained in this study were normal. Levels of Mn in red pepper was of the order: Makola > Takoradi > Ayigya > Kaneshie > Tema > Central > Asafo.

Mn levels in rosemary ranged from 16.55 to 21.31 µg/g. The least and the highest levels of Mn recorded by rosemary in this study were all high and above 15 µg/g. Central market recorded the highest followed by Takoradi, Ayigya, Asafo, Kaneshie and Tema markets in that order.

The levels of Mn in ginger ranged from 15.88 to 24.73 µg/g. The second highest level of Mn in this study was obtained in ginger from the Makola market which is 24.73 µg/g. The levels recorded for manganese were all above 15 µg/g. Levels of Mn in ginger from the various market had the order: Makola > Tema > Kaneshie > Central > Takoradi > Ayigya > Asafo market.

Levels recorded for the grain of paradise for manganese ranged from 17.31 to 19.94 µg/g. The highest was recorded by Ayigya market with Takoradi having the next highest level. Asafo and Tema markets recorded levels below 17.5 µg/g, all the other markets recording levels above 19 µg/g.

The levels of Mn in Ashanti pepper ranged from 17.39 to 20.94 µg/g. Comparing the three markets from Kumasi, Central market recorded the least, followed by Ayigya and Asafo. In general, Asafo market recorded the highest followed by Takoradi, Ayigya, Central and Makola markets.

Mn levels recorded by white pepper for the markets were quite high and ranged from 15.54 to 19.79 µg/g. The Tema market had the highest Mn level while the Makola market showed the least. Ayigya market recorded the second highest value. White pepper is naturally rich in Mn.
Black pepper is a plant naturally rich in manganese and the values realized confirm that. The levels ranged from 17.59 to 20.91 µg/g. The levels of Mn obtained for white pepper as compared to that of black pepper were not so different from each other. This suggests that Mn levels in the fruit cover added in the preparation of black pepper are low as compared to the other metals discussed earlier. Tema market obtained the highest level of Mn and Makola obtained the lowest level.

The manganese levels in garlic ranged from 14.93 to 19.40 µg/g. The levels recorded by all the markets were all high indicating the presence of normal Mn levels in the soil where they were cultivated. Ayigya market showed the highest level and Kaneshie market the least. The market that obtained the second highest level of Mn was Central followed by Tema and Makola markets respectively.

Manganese in onions ranged from 17.46 to 18.12 µg/g. The levels were high indicating that the onion plants were cultivated in soils with normal Mn levels. The Central market recorded the highest and Ayigya market the lowest.

The Mn in thyme ranged from 15.29 to 19.57 µg/g with the highest found in the Makola market followed by Kaneshie market, Takoradi, Central and Tema markets respectively.

Manganese levels in prekese ranged from 17.75 to 20.41 µg/g and these were all above the least normal level of Mn which is 15 µg/g. The Tema market recorded the highest while Central market recorded the lowest level.

A study by Gupta *et al.* (2003) on mineral composition of eight common spices reported high levels of Mn in the spices that were studied. The mean levels ranged from 33.6 µg/g in coriander to107.62 µg/g in carium. These levels are very high compared to that of this study.

Another study by Ozkutlu *et al.* (2006) on Cd and micronutrient of spices commonly consumed in Turkey showed Mn level with a range of 10 to 355 µg/g. The highest level was found in clove which seems to be an excellent source of Mn. These levels are very high compared to that obtained in this study. Though the levels of Mn obtained in this study are not very high, the results by Ozkutlu *et al.* (2006) indicate that plants could accumulate high levels of Mn when it is found in the soil where the plants are cultivated. It can also be low in plants which are naturally rich in Mn but cultivated in soils deficient in Mn. It was also reported that these levels obtained were noticeably lower than ones previously found in herbs and spice plants from different parts of the world (Ozkultu *et al*., 2006).

The general levels of Mn obtained by the spices were all above the normal range of 15-150 µg/g. When the results of this study are compared with the normal range of Mn, the levels were rather low. This could be attributed to the spices being cultivated in soils with some levels of Mn. The recommended daily intake of manganese is 5mg/day for 70kg body weight. Considering the amounts presented in table 4.3, exposure to Mn through consumption of these spices is not significant.

4.1.7 Lead

Lead was not detected in any of the samples. Gupta *et al*., (2003) on mineral composition of eight common spices showed that results were heavily scattered because Pb was detected in four out of the spices studied in the range of 1.57-8.69 µg/g. It was concluded that the contamination might have resulted from some extraneous factors. Al-Eed *et al.,* (2002) reported the range of lead to be from trace to 14.30 mg/kg. They concluded that these levels of lead were dangerous for human consumption. Chizzola *et al*., (2003) reported that the level of lead were generally low. These results indicate that lead can be detected in samples that come from environments that are contaminated with lead and that the levels could be high with serious detrimental effects. It can be low or non detectable in spices that come from soils that are low in lead concentration.

The spices from this study showed non detectable levels of Pb indicating that, the areas that these spices came from were not contaminated with lead or that the lead in the soil is not in the bioavailable form for plants to absorb. It is gratifying to know that spices from the markets in this study are free from lead contamination and therefore, are safe to consume.

4.2 Correlation Analysis

Analysis for correlation between the metals was done using excel. Tables 4.4 to 4.26 show the results obtained.

TABLE 4.4 GINGER

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	-0.1320	1				
Fe µg/g	0.2548	0.0200	1			
Mn µg/g	-0.1622	-0.1932	0.4266	1		
Zn µg/g	0.4843	-0.1295	-0.1051	0.1115	1	
Cu µg/g	0.1182	0.4775	0.2280	-0.0033	0.2423	1

TABLE 4.5 CLOVE

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.1758	1				
Fe µg/g	-0.2103	0.0269	1			
Mn µg/g	-0.3538	0.2945	0.1066	1		
Zn µg/g	-0.1197	-0.5241	-0.6571	-0.1053	1	
Cu µg/g	0.7073*	-0.0704*	0.0224	-0.2549	0.1875	1

The values with (*) showed significant correlation

TABLE 4.6 SENEGAL PEPPER

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.2251	1				
Fe µg/g	0.7389*	0.2954	1			
Mn µg/g	0.4981	0.2553	0.9440*	1		
Zn µg/g	0.4668	0.6534*	0.7734*	0.7983*	1	
Cu µg/g	0.2765	0.1003	0.6825*	0.8095*	0.7559*	1

TABLE 4.7 NUTMEG

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.2473	1				
Fe µg/g	0.2049	0.6497*	1			
Mn µg/g	-0.1306	-0.7891*	-0.5751*	1		
Zn µg/g	-0.0754*	0.3696	0.3099	0.2281	1	
Cu µg/g	-0.5702	-0.5298*	-0.2354	0.1502	-0.3394	1

TABLE 4.8 GARLIC

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.7415*	1				
Fe µg/g	0.6815*	0.6778*	1			
Mn µg/g	0.5755*	0.5702*	0.5511*	1		
Zn µg/g	0.8731*	0.8518*	0.8819*	0.4369	1	
Cu µg/g	0.8143*	0.8386*	0.8889*	0.6815*	0.9037*	1

The values with (*) showed significant correlations.

TABLE 4.9 ONION

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.9071	1				
Fe µg/g	0.9628*	0.9871*	1			
Mn µg/g	0.9525*	0.9922*	0.9994	1		
Zn µg/g	0.9144*	0.9998*	0.9898*	0.9942*	1	
Cu µg/g	0.9612*	0.9880*	1.0000*	0.9995*	0.9906*	1

TABLE 4.10 ROSEMARY

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.7379*	1				
Fe µg/g	0.7483*	0.4983	1			
Mn µg/g	0.7620*	0.5948*	0.7581*	1		
Zn µg/g	0.8128*	0.4639	0.9392*	0.7905*	1	
Cu µg/g	0.8759*	0.5109*	0.9319*	0.8228*	0.9051*	1

TABLE 4.11 BLACK PEPPER

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.9694*	1				
Fe µg/g	0.8401*	0.9476*	1			
Mn µg/g	0.8737*	0.9664*	0.9979*	1		
Zn µg/g	0.7047*	0.8574*	0.9769*	0.9608*	1	
Cu µg/g	0.7905*	0.9167*	0.9963*	0.9886*	0.9916*	1

The values with (*) showed significant correlations.

TABLE 4.12 RED PEPPER

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.3232	1				
Fe µg/g	0.8966*	0.4644	1			
Mn µg/g	-0.0448	0.2741	0.3118	1		
Zn µg/g	0.1180	-0.6587*	0.1011	-0.0155	1	
Cu µg/g	-0.1657	-0.6744*	-0.4386	-0.6981*	0.0466	1

TABLE 4.13 ANISE

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.2358	1				
Fe µg/g	0.5707*	0.1797	1			
Mn µg/g	0.5192*	0.2055	0.9547*	1		
Zn µg/g	0.2828	0.4739	0.8119*	0.7051*	1	
Cu µg/g	0.2419	0.1798	0.9234*	0.9225*	0.844*	1

TABLE 4.14 THYME

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	-0.0458	1				
Fe µg/g	0.6775*	0.6828*	1			
Mn µg/g	0.5990*	0.5260*	0.9015*	1		
Zn µg/g	0.7339*	0.3186*	0.6979*	0.6567*	1	
Cu µg/g	0.6923*	0.3404	0.6545*	0.6378*	0.9889*	1

The values with (*) showed significant correlations.

TABLE 4.15 SENEGAL PEPPER

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.6321*	1				
Fe µg/g	0.4304	0.6683*	1			
Mn µg/g	0.5281*	0.7381*	0.9826*	1		
Zn µg/g	0.3771	0.3092	0.7539*	0.7811*	1	
Cu µg/g	0.3713	0.7450*	0.9582*	0.9344*	0.5555*	1

TABLE 4.16 ASHANTI PEPPER

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.5811*	1				
Fe µg/g	0.9237*	0.5230*	1			
Mn µg/g	0.9445*	0.5255*	0.9920*	1		
Zn µg/g	0.6625*	0.3994	0.8976*	0.8530*	1	
Cu µg/g	0.8346*	0.4870	0.9241*	0.8925*	0.8667*	1

TABLE 4.17 AIDAN

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.3203	1				
Fe µg/g	0.9334*	0.6389*	1			
Mn µg/g	0.0316	0.5805*	0.2456	1		
Zn µg/g	0.8210*	0.8038*	0.9712*	0.3698	1	
Cu µg/g	0.9481*	0.6048*	0.9990*	0.2214	0.9599*	1

The values with (*) showed significant correlation.

TABLE 4.18 WHITE PEPPER

	Hg µg/g	Cd µg/g	Fe µg/g	Mn µg/g	Zn µg/g	Cu µg/g
Hg µg/g	1					
Cd µg/g	0.3960	1				
Fe µg/g	0.6218*	0.9654*	1			
Mn µg/g	0.6285*	0.9631*	1.0000*	1		
Zn µg/g	-0.1667	0.8394*	0.6686*	0.6622*	1	
Cu µg/g	0.7587*	0.8986*	0.9819*	0.9835*	0.5158*	1

The values with (*) significant correlations.

For the spices, there was a weak negative correlation between Cd and Hg in ginger. A weak negative correlation existed between Mn and Hg and Cd, another between Zn and Cd and Fe and another correlation between Cu and Mn all in ginger. All the rest gave a weak positive correlation.

A strong positive correlation was found between Cd and Hg for clove. All the rest showed weak positive and negative correlations. A strong positive correlation existed between Fe and Hg; Zn and Cd; Fe and Mn. A strong positive relation existed between Cu and Fe, Mn and Zn in Senegal pepper. The other metals gave weak positive correlations for Senegal pepper.

There was a weak negative and positive correlation between all the metals with the exception of Fe and Cd; Mn and Cd; Zn and Hg; Cu and Hg, Cd which showed a strong negative correlation. All these were observed for nutmeg.

A weak positive correlation between Zn and Mn was observed for garlic. All the metals showed strong positive correlation among each other. A strong positive correlation was observed in onion for all the metals. Weak positive correlations existed between Fe and Cd; Zn and Cd for rosemary. The rest exhibited strong positive correlations.

Strong positive correlations were shown by black pepper for all the metals involved. There was a strong positive and negative correlation between Fe and Hg, Cu and Mn and then weak positive and negative correlations for the others in red pepper. Strong positive correlations existed between Fe and Hg; Mn and Hg and Fe; Zn and Fe and Mn; Cu and Fe and the finally Mn and Zn. This was observed for Anise. All the others gave weak positive correlations.

Weak positive relations were observed between Fe and Hg, Zn and Cd and Hg for grain of paradise. All the rest gave a strong positive correlation. A strong positive correlation existed between all the metals for Ashanti pepper. There was a weak positive correlation between Cd and Hg; Mn and Hg and Fe; Zn and Mn and then Cu and Mn. The rest of the metals gave strong positive correlation between them for prekese. Lastly a weak negative and positive correlation existed between Zn and Hg; Cd and Hg. All the others gave strong positive correlations for white pepper.

The strong positive correlations that existed between the metals indicate that, as the concentration of a metal increases, the concentration of the other metal to which it is correlated also increases. A weak positive correlation means that the probability that an increase in the level of one metal causes another to increase is very small. The strong negative correlation meant that as the level of one metal increases the level of the other metal decreases. A weak negative correlation means that as the concentration of a metal increases the concentration of the other metal does not decrease or the decrease is very small. For strong positive and negative correlations the values should be 0.5 and above and -0.5 and above respectively.

CHAPTER FIVE

5 CONCLUSIONS AND RECOMMENDATIONS

5.1 CONCLUSION

From the analysis carried out, the following conclusions may be deduced from the results obtained.

- Some metal content in some natural spices found on the Ghanaian market were determined. The study reports the metal concentration levels in some spices from Makola, Asafo, Kaneshie, Takoradi, Ayigya, Kumasi Central and Tema markets. All the concentrations of the metals in the spices from the various markets were below the World Health Organization (WHO) limit.

- The concentration of the metals determined in the spices from all the markets considered in microgram/gram ranged from ND to 0.031 for Hg, ND to 1.41 for Cd, 65.35 to 221.15 for Fe, 14.93 to 24.96 for Mn, ND to 17.70 for Zn and 4.22 to 22.91 for Cu. Lead was not detected in any of the samples. The results obtained showed that the spices from the various markets in this study are not likely to pose any health risk to the public through consumption of the spices for both the toxic elements and the micronutrients determined.

- The correlation analysis done for the metals showed that, strong positive and negative correlation existed between the metals for both the markets and the spices.

5.2 RECOMMENDATIONS

The following recommendations are suggested as a result of the outcome of this study

- The concentrations of heavy metals in other natural spices on the Ghanaian market which are not covered by this research should be determined.
- Spices should be directly sampled from the areas of cultivation.
- Periodic monitoring of the levels of heavy metals in spices should be encouraged.

REFERENCES

Abila, B., Richens, A. and Davies, J. A. (1996). Anticonvulsant effects of extracts of the West African black pepper, *Piper guineense*. *J Ethnopharmacol,* 39: 113-117. PMID: 16400.

Adamson, M. W. (2004). *Food in Medieval Times*. ISBN 0-313-32147-7.

Adewunmi, C. O. (2008). Aidan –Success in fighting Bilharzia the natural way. Http://www.scienceinafrica.co.2a/3plant.htm.

Adeyeye, E. I., Arogundade, L. A., Akintayo, E. T., Aisida, O. A. and Alao, P. A. (2000). Calcium, zinc and phytate interrelationships in some foods of major consumption in Nigeria. *Food Chem*, 71: 435–441.

Adriano, D. C. (1984). Trace Metals in the Terrestrial Environment. New York: Verlag Springer.

Afshari, A. and Taghizadeh. (2007). The effect of ginger on diabetic nephropathy, plasma antioxidant capacity and lipid peroxidation in rats. *Food Chemistry,* 101: 148-153

Ahuja, K. D., Robertson, I. K., Geraghty, D. P. and Ball, M. J. (2006). Effects of chili consumption on postprandial glucose, insulin, and energy metabolism. *Am J Clin Nutr*, 84: 63-69. PMID: 16825682.

Al-Amin and Zainab, M. (2006). Anti-diabetic and hypolipidaemic properties of ginger (Zingiber officinale) in streptozotocin-induced diabetic rats. *British Journal of Nutrition,* 96: 660 - 666.

Al-Eed, M. A. Assubaie, F. N., El-Garawany, M. M., El-Hamshary, H. and El-Tayeb, Z. M. (2002). "Determination of heavy metal levels in common spices", *Egypt. J Appl Sci,* 17: 87-98.

Amaechi, B. T., Higham, S. M, and Edgar W. M. (1999). Techniques for the production of dental eroded lesions in vitro. *J Oral Rehabil*, 26: 97-102. PMID:12580.

Andorfer, J. H., Tchaikovskaya T. and Listowsky, I. (2004). Selective expression of glutathione S-transferase genes in the murine gastrointestinal tract in response to dietary organosulfur compounds. *Carcinogenesis*, 25: 359-367.

Andrews, N. C. (1999). Disorders of iron metabolism. *N Engl J Med*, 341: 1986-1995.

Apitz-Castro, R., Escalante, J. and Vargas, R. (1986). Ajoene, the antiplatelet principle of garlic, synergistically potentiates the antiaggregatory action of prostacyclin, forskolin, indomethacin, and dipyridamole on human platelets. *Thromb Res*, 42: 303-311.

Aruoma, O. I, Spencer, J. P. and Rossi, R. (1996). An evaluation of the antioxidant and antiviral action of extracts of rosemary and Provencal herbs. *Food Chem Toxicol,* 34: 449-456.

ATSDR (Agency for Toxic Substances and Disease Registry). (1999). Toxicological profile for Mercury. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

ATSDR (Agency for Toxic Substances and Disease Registry). (2000). Case studies in environmental medicine: lead toxicity. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

ATSDR (Agency for Toxic Substances and Disease Registry). (2004). Toxicological profile for Copper. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

ATSDR (Agency for Toxic Substances and Disease Registry). (2005). Toxicological profile for Zinc. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

ATSDR (Agency for Toxic Substances and Disease Registry). (2007). Toxicological profile for Lead. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

ATSDR (Agency for Toxic Substances and Disease Registry). (2008a). Draft toxicological profile for Cadmium. U.S. Department of health and human Services, Public Health Service.

ATSDR (Agency for Toxic Substances and Disease Registry). (2008b). Toxicological profile for Manganese. (Draft for Public Comment). Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

Augusti, K. (1996). Therapeutic values of onion (*Allium cepa L.*) and garlic (*Allium sativum L.*), *Indian J Exp Biol,* 34: 634–640.

Bagamboula, C. F., Uyttendaeleand, M. and Debevere, J. (2004). Inhibitory effect of thyme and basil essential oils, carvacrol, thymol, estragol, linalool and p-cymene towards *Shigella sonnei* and *S. flexneri*. *Food Microbio,* 21: 33-42.

Baillon, H. (1877). (Hachette, Paris) *Histoire des plantes,* Pp 1-2.

Basgel, S. and Erdemoglu, S. B. (2005). Determination of mineral and trace elements in some medicinal herbs and their infusions consumed in Turkey, *Sci Total Environ,* 359: 82-89.

Bautista, D. M., Movahed, P., Hinman, A., Axelsson, H. E., Sterner, O., Hogestatt, E. D., Julius, D., Jordt, S. E. and Zygmunt, P. M. (2005). Pungent products from garlic activate the sensory ion channel TRPA1. Proc *Natl Acad Sci,* U S A. PMID: 16103371.

Bellinger, D., Leviton, A., Rubinovitz, M., Needleman, H. and Waternaux, C. (1987). Longitudinal analysis of prenatal and postnatal lead exposure and early cognitive development. *N Engl J Med,* 316: 1037-1043

Bender, A. E. and Bender, D. A. (2005). Food and Nutrition. A Dictionary of Food and Nutrition.

Benton, J. J. (1998). Plant Nutrition Manual – Gardening. Pp 55-57.

Berthold, H. K. and Sudhop, T. (1998). Galic preparation for prevention of atherosclerosis. *Curr Opin Lipidol,* 9: 565-569.

Bhattacharjee, S., Dasgupta, P. and Paul, A. R., Ghoshal, S., Padhi, K. K. and Pandey, L. P. (1998). Mineral element composition of spinach. *J Sci Food Agric,* 77: 456–458.

Bhattacharya, K., Yadava, S., Papp, T., Schiffmann, D. and Rahman, Q. (2004). Reduction of chrysotile asbestos-induced genotoxicity in human peripheral blood lymphocytes by garlic extract. *Toxicol Lett*, 153: 327-332. PMID: 15454308.

Bhaskaram, P. (2001). Immunobiology of mild micronutrient deficiencies. *Br J Nut,* 85: 75-80.

Blaurock-Busch E. (1997). The Clinical Effects of Manganese (Mn). Mineral & Trace Element Analysis, Laboratory and Clinical Application. *A J Clinical Chemistry*, 33: 351-356

Blumenthal, M, Busse, W. R. and Goldberg, A. (1998). *The Complete German Commission E Monographs: Therapeutic Guide to Herbal Medicines*. Austin: American Botanical Council and Boston: Integrative Medicine Communications. Pp 82–83.

Bothwell, T. H., Charlton, R. W., Cook, J. D. and Finch, C. A. (1979). Iron Metabolism in Man. St. Louis: Oxford: Blackwell Scientific.

Bordia, A., Verma, S. K. and Srivastava, K. C. (1997). Effect of ginger (*Zingiber officinale Rosc*.) and fenugreek (*Trigonella foenumgraecum L*.) on blood lipids, blood sugar, and platelet aggregation ion patients with coronary heart disease. *Prostaglandins Leukot Essent Fatty Acids*, 56: 379-384.

Chao**,** J. and Kikano, G. E. (1993). Lead poisoning in children*. Am Fam Physician*, 47: 113-120.

Chen**,** J. C., Li-Jiau, H., Shih-Lu, W., Sheng-Chu, K., Tin-Yun, H. and Chien-Yun, H. (2007). Ginger and Its Bioactive Component Inhibit Enterotoxigenic Escherichia coli Heat-Labile Enterotoxin-Induced Diarrhoea in Mice. *Journal of Agricultural and Food Chemistry,* 55: 8390–8397.

Chizzola, R., Michitsch, H. and Franz, C. (2003). Monitoring of metallic micronutrients and heavy metals in herbs, spices and medicinal plants from Austria. *European Food Research and Technology*, 216: 407-411.

Chopra, R. N. and Chandler, A. C. (1928). *Anthelmintics and Their Uses in Medical and Veterinary Practice*. Baltimore: Williams & Wilkins Co: 159.

Cosentino, S., Tuberoso, C. I. and Pisano, B. (1999). In-vitro antimicrobial activity and chemical composition of Sardinian Thymus essential oils. *Lett Appl Microbiol*, 29: 130-135. PMID:12470.

Corbett, J. V. (1995). Accidental poisoning with iron supplements. *MCN Am J Matern Child Nurs*, 20: 234.

Daily Mail, (2007). Spicy foods 'could protect against cancer' 9th January.
Dalby, A. (2001). *Dangerous Tastes: The Story of Spice.* USA ISBN 0520236742.

Dallman, P. R. (1986). Biochemical basis for the manifestations of iron deficiency. *Annu Rev Nutr*, 6: 13-40.

De Leonardis, A., Macciola, V. and De Felice, M. (2000). Copper and iron determination in edible vegetable oils by graphite furnace atomic absorption spectrometry after extraction with diluted nitric acid. *International Journal of Food Science and Technology*, 35: 371–375.

Devereux, P. (1996). *Re-Visioning the Earth: A Guide to Opening the Healing Channels Between Mind and Nature*. New York: Fireside. pp. 261–262.

Dillon, S. A., Burmi, R. S. and Lowe, G. M. (2003). Antioxidant properties of aged garlic extract: an in vitro study incorporating human low density lipoprotein. *Life Sci*, 72: 1583-1594.

Divrikli, U., Saracoglu, S., Soylak, M. and Elci, L. (2003). Determination of trace heavy metal contents of green vegetable samples from Kayseri-Turkey by flame atomic absorption spectrometry. *Fresenius Environmental Bulletin*, 12: 1123–1125.

Divrikli, U., Horzum, N., Soylak, M. and Elci, L. (2006). Trace heavy metal content of some spices and herbal plants from Western Anatolia, Turkey. *International journal of food Science and Tech*, 41: 712-716.

Dorman, H. J. D. and Deans, S. G. (2000). Antimicrobial agents from plants: antibacterial activity of plant volatile oils. *Journal of Applied Microbiology*, 88: 308. doi:10.1046/j.1365-2672.2000.00969.x.

Duffus, J. H. (2002). Heavy metals" a meaningless term? (IUPAC Technical Report). Pure and Applied Chemistry, pp. 793-807. doi:10.1351/pac200274050793.

Durak, I., Aytac B., Atmaca, Y., Devrim, E., Avci, A., Erol, C, and Oral, D. (2004). Effects of garlic extract consumption on plasma and erythrocyte antioxidant parameters in atherosclerotic patients. *Life Sci*, 75: 1959-1966. PMID: 15306163.

Ensminger, A. H. and Esminger, M. K. J. (1986). Food for Health: A Nutrition Encyclopedia. Clovis, California: Pegus Pres. PMID: 15210.

FAO/WHO. (2001). Food Additives and Contaminations. Joint FAO/WHO Food Standards Programme. ALINORM01/12A.1 –289.

Fleischauer, A. T., Poole C. and Arab, L. (2000). Garlic consumption and cancer prevention: meta-analyses of colorectal and stomach cancers. *Am J Clin Nutr*, 2: 1047-1052.

Friedman**,** S**.** M., Henika, P. R. and Mandrell, R. E. (2002). Bactericidal activities of plant essential oils and some of their isolated constituents against Campylobacter jejuni, Escherichia coli, Listeria monocytogenes, and Salmonella enterica. *J Food Prot*, 65: 1545-1560.

Fugh-Berman, A. (2000). Herbs and dietary supplements in the prevention and treatment of cardiovascular disease. *Prev Cardiol*. 3: 24-32.

Fukushima, S., Takada, N., Hori, T. and Wanibuchi, H. (1997). Cancer prevention by organosulfur compounds from garlic and onion. *J Cell Biochem Suppl*, 27:100-105. PMID: 13650.

Gable, R. S. (2006). The toxicity of recreational drugs. *American Scientist,* 94: 206–208.

Galeone, C., Pelucchi, C., Levi, F., Negri, E., Franceschi, S., Talamini, R., Giacosa, A. and La Vecchia, C. (2006). Onion and garlic use and human cancer. *Am J Clin Nutr*, 84: 1027-1032. PMID: 17093154.

Gilbert, J. (1984). Analysis of food Contamination. Elsevier App. Sci Pups., London.ISBN:0-85334-255-5

Gillooly, M., Bothwell, T. H., Torrance, J. D., MacPhail, P., Derman, D. P., Bezwoda, W. R., Mills, W and Charlton, R. W. (1983). "The Effects of Organic Acids, Phytates and Polyphenols on the Absorption of Iron from Vegetables. *British Journal of Nutrition,* 49: 331–342.

Goyer, R. A. and Clarkson, T. W. (2001). Toxic Effects of Metals. In: Klassen, C.D. (Ed.), Casarett and Doull's toxicology: The basic science of poisons (6th ed., pp. 811-837). New York: McGraw-Hill Medical Publishing Division.

Gupta, K. K., Bhattacharjee, S., Kar, S., Chakrabarty, S., Thakar, P., Bhattacharyya, G. and Srivastava, S. C. (2003). Mineral composition of eight common spices. Comm. *Soil Plant Anal*, 34: 681-693.

Haas J. D. and Brownlie. T. (2001). Iron deficiency and reduced work capacity: a critical review of the research to determine a causal relationship. *J Nutr*, 131: 676-690.

Hallberg, L., Hulten, L. and Gramatkovski, E. (1997). Iron Absorption from the Whole Diet in Men: How Effective Is the Regulation of Iron Absorption? *American Journal of Clinical Nutrition,* 66: 347–356.

Hanrahan, C. and Frey, R. J. (2005). Rosemary. In: J. L. Longe, (ed). *The Gale Encyclopedia of Alternative Medicine.* Farmington Hills, MI: Thomson/Gale. ISBN 0787693960.

Hansen, C. R. Jr. (1983). Copper and zinc deficiencies in association with depression and neurological findings. *Biological Psychiatry,* 18: 395-401.

Hayter, J. (1980). Trace elements: implication for nursing. *J. Adv. Nurs*, 5: 91–101.

Heidi**,** A. (2002). The Chilli Pepper Diet: The Natural Way to Control Cravings, Boost Metabolism and Lose Weight, ISBN-10: 1558749268.

Huxley, R. R., and Neil, H. A. W. (2003). The relation between dietary flavonol intake and coronary heart disease mortality: a meta-analysis of prospective cohort studies, *European Journal of Clinical Nutrition,* 57: 904-908.

Institute of Medicine. Food and Nutrition Board. (1999). Dietary Reference Intakes: Calcium, Phosphorus, Magnesium, Vitamin D and Fluoride. National Academy Press. Washington, DC.

Institute of Medicine. (2001). Food and Nutrition Board. Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium and Zinc. Washington, DC: National Academy Press.

Johannesson, M. (2002). *A review of risk associated to cadmium, lead, mercury and zinc.* Pp 62

Kelm, M. A., Nair, M. G., Strasburg, G. M. and DeWitt, D. L. (2000). Antioxidant and cyclooxygenase inhibitory phenolic compounds from Ocimum sanctum Linn. *Phytomedicine,* 7: 7-13. PMID: 12240.

Kennish, M. J. (1992). Ecology of Estuaries. Anthropogenic effects. CRC. Press, Inc., Boca Raton, F1.

Kulevanova, S., Kaftandzieva, A. and Dimitrovska, A. (2000). Investigation of antimicrobial activity of essential oils of several Macedonian Thymus L. species (*Lamiaceae*). *Boll Chim Farm,* 139: 276-280. PMID: 12460

Lahey, F. (1975). A copper Deficiency Anaemia is Indistinguishable Haematologically from that of an Iron Deficiency Anaemia. *Clinical Significance of the Essential Biological Metals*, p. 57.

Lane T. W. and Morel, F. M. (2000). A biological function for cadmium in marine diatoms. *Proc Natl Acad Sci,* 97: 4627–4631. doi:10.1073/pnas.090091397. PMID 10781068.

Lane, T. W., Saito, M. A., George, G. N., Pickering, I. J., Prince, R. C. and Morel, F. M. (2005). Biochemistry: a cadmium enzyme from a marine diatom. *Nature,* 435: 42. doi:10.1038/435042a. PMID 15875011.

Lee, Y. L., Cesario, T., Wang, Y., Shanbrom, E. and Thrupp, L. (2003). Antibacterial activity of vegetables and juices. *Nutrition*, 19: 994-996.

Lemonica, I. P, Damasceno, D. C. and Di-Stasi, L. C. (1996). Study of the embryotoxic effects of an extract of rosemary (*Rosmarinus officinalis* L.) *Braz Med Biol Res*, 19: 223-227.

Loghman-Adham, M. (1997). Renal effects of environmental and occupational lead exposure. *Environ Health Perspect*, 105: 928-938.

Matt, K. J. (1972). Mercury Uptake from soil by various plant species, *Bulletin of environmental contamination and toxicology,* 8: 77-80.

McGee, H. (2004). *On Food and Cooking (Revised Edition)*. Scribner, 427–429. ISBN 0-684-80001-2. "Black Pepper and Relatives".

Meluzzi A., Simoncini F., and Sirri F. (1996). Feeding hens diets supplemented with heavy metals (chromium, nickel and lead). *Arch Geflugelkunde*, 3: 119-125

Mengel, K. and Kirkby, E.A. (1982). Principles of plant nutrition. International Potash institute Switzerland.

Mori, A., Lehmann, S., O'Kelly, J., Kumagai, T., Desmond, J., Pervan, M., McBride, W., Kizaki, M. and Koeffler, H. P. (2006). Capsaicin, a Component of Red Peppers, Inhibits the Growth of Androgen-Independent, p53 Mutant Prostate Cancer Cells. *Cancer Res*, 66: 3222-3229. PMID: 16540674.

Myres, A. W. and Easson, E. (1992). Lead in paint: an "old" problem revisited. *Environ Health Rev*, 36: 102-104.

Narang, R. L., Gupta, K. R., Narang, H. P. and Singh, R. (1991). Levels of copper and zinc in depression. *Indian Journal of Physiology and Pharmacology*, 10: 69-73.

Nnorom, I. C., Osibanjo, O. and Ogugua, K. (2007). Trace Heavy Metal levels of Some Bouillon Cubes, and Food Condiments Readily consumed in Nigeria. *Pakistan J of Nutrition*, 6: 122-127.

Nolan, K. R. (1983). Copper toxicity syndrome. *Journal of Orthomolecular Psychiatry*, 12: 270-282.

Odell, B. L. and Sunde, R. A. (1997). *Handbook of Nutritionally Essential Mineral Elements*. New York.

Oehme, F. W. (1989). Toxicity of heavy metals in the environment. Marcel Dekker, Inc., New York, Part 1, 1.

Oluwatuyi, M., Kaatz, G. W. and Gibbons, S. (2004). Antibacterial and resistance modifying activity of *Rosmarinus officinalis.Phytochemistry*, 65: 3249-3254.

Onianwa, P. C., Adetola, I. G., Iwegbue, C. M. A., Ojo, M. F. and Tella, O. O. (1999). Trace heavy metal composition of some Nigerian beverages and food drinks. *Food Chem*, 66: 275-279.

Ozkutlu, F., Sekeroklu, N. and Kara, M. (2006). Monitoring of Cadmium and Micronutrients in Spices Commonly Consumed in Turkey. *J of Agriculture and Biological Sciences*, 2: 223-226.

Penland, J. G. and Johnson, P. E. (1993). Dietary calcium and manganese effects on menstrual cycle symptoms. *Am J Obstet Gynecol*, 168: 1417–1423,

Pokorney, J. (1991). Natural antioxidants for food use. *Trends Food Sci. Technol*, 9: 223–227.

Portnoi, G., Chng, L. A. and Karimi-Tabesh, L. (2003). Prospective comparative study of the safety and effectiveness of ginger for the treatment of nausea and vomiting in pregnancy. *Am J Obstet Gynecol*, 189: 1374-1377.

Rosengarten, F. Jr. (1969). The Book of Spices, pp. 23-96, Jove Publ., Inc., New York.

Sattar, A., Wahid, M. and Durrani,S. K. (1989). Concentration of selected heavy metals in spices, dry fruits and plant nuts. *Plant Foods for Human Nutrition*, 39: 279-286**.**

Schrauzer, G. N. (1984). The Discovery of the Essential Trace Elements: An Outline of the History of Biological Trace Element Research. *In Biochemistry of the Essential Ultra-trace Elements*, pp 17–31. New York: Plenum.

Sethi, V. and Meena, M. R. (1997). Role of spices and their essential oils as preservatives antimicrobial agents: a review. *Indian Food Packer*, 51: 25–42.

Serafin, T. and Verity, A. (1991). Oxidative mechanisms underlying methyl mercury neurotoxicity. *International Journal of Developmental Neuroscience*, 9: 147-153

Shaw, B. P. and Panigrahi, A. K. (1986). Uptake and tissues distribution of mercury in some plant species collected from contaminated area in India: it ecological implications. *Journal Archives of Environmental Contamination*, 15: 439-446.

Shelef, L**.** A. (1983). Antimicrobial effects of spices. *J Food Saf*; 6: 29–44.

Sherman, P. W. and Billing, J. (1999). Darwinian gastronomy: why we use spices— spices taste good because they are good for us. *Bioscience*, 49: 453–463.

Shils, M. E. (1999). Magnesium. 9th Edition. In Shils, M. E, Olson, J. A, Shike, M., and Ross, A. C. (Eds) Modern Nutrition in Health and Disease. New York: Lippincott Williams and Wilkins, pp. 169-192.

Smith, G. C., Clegg, M. S., Keen, C. L. and Grivetti, L. E. (1996). Mineral value of selected plant foods common to Southern Burkina Faso and to Niamey, Niger, West Africa. *Int J Food Sci, Nutr*, 47: 41–53.

Sorensen**,** N., Murata, K., Butz-Jorgensen, E., Weihe, P. and Grandjean, P. (1999). Prenatal methymercury expposure as a cordiovscular risk factor at seven years of age. *Epidemiology*, 10: 370-375.

Spigelski, D. and Jones, P. J. (2001). Efficacy of garlic supplementation in lowering serum cholesterol levels. *Nutr Rev*, 59: 236-241.

Stipanuk, M. H. (2000). *Biochemical and Physiological Aspects of Human Nutrition*, 8: 305-350 Philadelphia.

Superko H. R. and Krauss R. M. (2000). Garlic powder, effect on plasma lipids, postprandial lipemia, low-density lipoprotein particle size, high-density lipoprotein subclass distribution and lipoprotein(a). *J Am Coll Cardiol*, 35 :321-326.

Tatsadjieu, L. N., Essia, Ngang, J. J. and Ngassoum, M. B. (2003). Etoa FX National High School of Agro-Industrial Science, Ngaoundere, Cameroon. *Fitoterapia*, 74: 469-472.

Tapsell, L. C., Hemphill, I, Cobiac, L., Patch, C. S., Sullivan, D. R., Fenech, M., 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. *Med J Aust*, 185: 4-24.

Thomson, M., Al Qattan, K. K. and Sawan, S. M. (2002). The use of ginger (Zingiber officinale Rosc.) as a potential anti-inflammatory and antithrombotic agent. *Prostaglandins Leukot Essent Fatty Acids*, 67: 475-478.

Thudichum, J. L. W. (1895). *The Spirit of Cookery: A Popular Treatise on the History, Science, Practice and Ethical and Medical Import of Culinary Art.* London and New York.

Turner, J. (2004). *Spice: The History of a Temptation*. Vintage Books. ISBN 0-375-70705-0.

Underwood, E. J. (1977). *Trace Elements in Human and Animal Nutrition*, 4th ed. New York: Academic Press.

Warkeny, J. and Hubbard, C. H. E. (1953). Acrodynia and mercury. *Journal of Pediatrics*, 42: 365-386.

Weiss, R. F. (1985). *Herbal Medicine*. Gothenberg, Sweden: Arcanum and Beaconsfield: Beaconsfield Publishers Ltd. Pp. 203–4.

WHO. (1972). Long term programme in environmental pollution control in Europe. The hazards to health of persistent substances in water. Technology documents on arsenic, lead, cadmium, manganese and mercury. Copenhagen.

Yang, J., Meyers, K. J., Van der, H. J and Liu, R. H. (2004). Varietal Differences in Phenolic Content and Antioxidant and Antiproliferative Activities of Onions. *J Agric Food Chem*, 52: 6787-6793. PMID:15506817.

Yuzbasi, N., Sezgin, E., Yildirim, M. and Yildirim, Z. (2003). Survey of lead, cadmium, iron, copper and zinc in Kasar cheese. *Food Additives and Contaminants*, 20: 464–469.

Zakrzewski, S. F. (1991). Principle of environmental toxicology. ACS Professional reference book, Washington, DC, 1.

Ziegler, E. E., and Filer, L. J. Jr. (1996) *Present Knowledge in Nutrition*, 7th. ed. Washington, D.C.: ILSI.

http://www.echochem.com/t_micronutrient.Micronutrient function: nutrient Deficiency systems.

http://www.indethinfo,com/nutmeg/health-effect.shtml

http://www.lenntech.com/recommended-daily-intake.htm

http://en.wikipedia.org/wiki/Aframomum_melegueta

http://www.acu-cell.com/pna:html.

http://www.mountainroseherb.com/learn/whitepepper

http//www.nkran.net/foodanddrink.jsp

http://www.the epicentre.com/spices/melegueta.html- Encyclopaedia of spices

http://www.ngrguardiannews.com/natural health/article 01-Guardian Newspaper. (2009). Researchers Explore Herbal Treatment for Tuberculosis.

http://www.chaddsfordhistory.org/history/herbs2.htm- McCormick.com Spice Encyclopedia. The History of Spices.

http://www.scientificamerican.com/article.cfm?id=what-is-the-chemical-proc