HEAVY METAL EXPOSURE ASSESSMENT USING NAIL AS BIO-INDICATOR AMONG THE FARMERS IN MALAYSIA.

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ABSTARCT

Heavy metals, particularly cadmium, lead, and arsenic, constitute a significant potential threat to human health. Chronic exposure to toxic heavy metal can lead to deleterious human health effect. The aim of this study was to determine the level of heavy metal concentration among the farmers in Malaysia as well as to understand the correlation between the heavy metal levels with the heavy metal sensitivity symptoms. The study comprised of 33 farmers. Nails sample was collected among the farmers and the adopted questionnaire were used to obtained demographic status and related symptoms. Inductively coupled plasma mass spectrometry (ICP-MS) was used to analyze the nail sample and the Statistical Package for Social Sciences (SPSS) were used to analyze the data obtained In this study, there were significant correlations between the working experiences with level of arsenic, cadmium, nickel and lead, ($r=0.499$, $r=0.398$, $r=0.454$, $r=0.498$., $p<0.05$). Besides, the analysis of the relationship between the age and levels of arsenic ($r=0.499$), cadmium ($r=0.398$), nickel ($r=0.454$), and lead ($r=0.498$), ($p<0.05$) showed significant relationship. This finding suggests that agriculture activities could contribute to the accumulations of heavy metals in farmers. Hence, the control of environmental levels of and human exposure to these metals to prevent adverse health effects is still an important public health issue.

ABSTRAK

Logam berat, terutamanya kadmium, plumbum, dan arsenik, menpunyai potensi yang besar mengancami kesihatan manusia. Pendedahan kronik terhadap logam berat toksik boleh menyebabkan kesan kesihatan manusia. Tujuan kajian ini adalah untuk menentukan tahap logam berat di kalangan petani, Malaysia dan juga untuk memahami hubugan antara tahap logam berat dengan gejala sensitiviti logam berat. Kajian ini terdiri daripada 33 petani. Sampel kuku telah dikumpulkan di kalangan petani dan soal selidik telah digunakan untuk mengumpulkan status demografi diperolehi dan gejala-gejala kesihatan yang berkaitan. *Inductively Coupled Plasma – Mass Spectrometry* (ICP-MS) telah digunakan untuk menganalisis sampel kuku dan *Statistical Package for Social Sciences* (SPSS) telah digunakan untuk menganalisis data yang diperolehi dalam kajian ini, terdapat hubungan yang signifikan antara pengalaman kerja dengan tahap arsenik , kadmium, nikel dan plumbum, ($r = 0,499$, $r = 0,398$, $r = 0,454$, $r = 0,498$., $p < 0.05$). Selain itu, analisis hubungan antara umur dan tahap kandungan arsenik ($r = 0,499$), kadmium ($r = 0,398$), nikel ($r = 0,454$), dan plumbum ($r = 0,498$), ($p < 0.05$) menunjukkan hubungan yang signifikan . Kajian ini telah menunjukkan bahawa aktiviti pertanian boleh menyumbang kepada pengumpulan logam berat dalam badan petani. Oleh itu, kawalan tahap alam sekitar dan pendedahan manusia kepada logam diperlukan untuk mengelakkan kesan kesihatan yang buruk dan keseimbangan alam sekitar.

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LIST OF ABBREVIATIONS

ACGIH	American Conference for Governmental Industrial Hygienists
Al	Aluminum
ATSDR	Agency for Toxic Substances and Disease Registry
Be	Beryllium
Cd	Cadmium
Cr	Chromium
Cu	Copper
DOSH	Department of Occupational Safety and Health
Fe	Iron
ICP-MS	Inductively Coupled Plasma-Mass Spectrometry
Mn	Manganese
Ni	Nickel
OSHA	Occupational Safety and Health Act
Pb	Lead
SPSS	Statistical Package for Social Sciences
USECHH	Use and Standards of Exposure of Chemical Hazardous to Health
Zn	Zinc

CHAPTER 1

INTRODUCTION

1.1 INTRODUCTION

Metals are elements can be obtained naturally in the earth's crust (Morais, 2012). Some of the metals found in environment are essential nutritionally while others are xenobiotic (Hu, 2002). Zinc and iron are important metals that needed in our body for human growth especially in puberty stage. However, among the 92 naturally occurring metals elements, mostly 30 metals and metalloids poses toxic effect to human, Beryllium (Be), Boron (B), Lithium (Li), Aluminum (Al), Titanium (Ti), Vanadium (V), Chromium (Cr), Manganese (Mn), Copper (Cu), Arsenic (As), Selenium (Se), Strontium (Sr), Molybdenum (Mo), Palladium (Pd), Silver (Ag), Cadmium (Cd), Tin (Sn), Antimony (Sb), Tellurium (Te), Nickel (Ni), Barium (Ba), Tungsten (W), Platinum (Pt), Gold (Au), Mercury (Hg), Lead (Pb) and Bismuth (Bi) (Morais, 2012). All these metals are widely used in industry where Pb, Cd, and Hg are generally have the most toxic to human health (Ming-Ho, 2005). Heavy metals can be defined as a group of metallic elements that exhibit certain chemicals and electrical properties where their density are generally greater than 5 g/cm^3 or five times higher than water density (Ming-Ho, 2005).

Nowadays, agriculture industries in Cameron Highland especially vegetation and flower cultivation has an over-capacity and is experiencing a highly competitive situation in the domestic market. The massive growth of the agriculture sector has been accompanied by the large-scale application of fertilizer and pesticide in order to stimulate the growth of plants. Inorganic fertilizer such as ammonium bicarbonate,

herbicides and bactericides are commonly been used to keep plant healthy. The use of pesticides and fertilizer in agriculture has increased the exposure of heavy metals among the farmer. Workers can exposed to the heavy metal fumes or gases even a low level chronic exposure by spraying, dusting and fumigation of the pesticide. Most of the heavy metals are highly toxic because it acts as ions or in certain compounds they are soluble in water and can easily absorbed into plant and animal tissues (Ming-Ho, 2005). In addition, most heavy metals has long lifetime that they remove very slowly from the body once it has been absorbed. Besides, they will accumulate in the environment and cause a long lasting environmental impact.

The presence of heavy metals in air, soil and water has become a serious global issue that causes the adverse effect to the environment, but mainly of the heavy metals are released to the environment by anthropogenic means such as the use of pesticides in agricultural sectors (Khlifi & Hamza-Chaffai, 2010). Heavy metals are toxic that can cause serious health effect when expose to it. The exposure can be either high acute exposure to toxicant or low level chronic exposure (Khudzari et al., 2012). According to Saat et al. (2013), long term exposure to heavy metals can cause Parkinson, Alzheimer, heart disease and lung cancer whereas short term exposure are usually reversible, typically has general symptoms, such as weakness or headache. This make diagnose for short term exposure to heavy metals very difficult unless a clinician has the knowledge and training and able to order the correct diagnostic test (Hu, 2002).

There are increases in numbers of research concerning the exposure of heavy metals among workers. Significant control measure has also been carried out in order to reduce the number of toxic metals in environment as well as in workplace. This has induced the study of human nails as a bio-indicator for detection of heavy metals among the Malaysia farmers. During the research, the levels of heavy metals in nail samples from farmers will be determined and correlated with their demographic factors. The result from this assessment will eventually help the agriculture sector to establish a better prevention method in order to decrease the heavy metals exposure among the farmers.

1.2 STUDY BACKGROUND

The oldest axiom of toxicology suggests 'the dose makes the poison' (Rozman & Doull, 2001). Everything can be harmful to human health with enough doses given. According to U.S. Agency for Toxic substances and Disease Registry ATSDR (2013), the first, second, third and seventh hazardous heavy metals on the list are arsenic, lead, mercury and cadmium respectively. Therefore, this study examines the rate of heavy metals among farmers. This research assessment will includes the lead (Pb), cadmium (Cd), mercury (Hg), and arsenic (As) which were generally considered as the most toxic heavy metal that affect human health.

Cameron Highland is located (4°28'N, 101°23'E) at the state of Pahang, Malaysia amid the lofty peaks of Banjaran Titiwangsa with a total area 71,218 hectares (Munisamy et al., 2013). The area experience mild temperature, ranging from 14 to 24°C throughout the year with averages rainfall 2660 mm yr^{-1}, high humidity and there is no marked dry season (Barrow et al., 2009). Therefore, it is popular tourist and recreational area. In addition, it has been recognized as key agricultural area, for vegetable, flower and tea (Gasim et al., 2009). Besides that, there are 66 % of the land are steep that having gradient greater than 20° (Munisamy et al., 2013). Generally, steep lands are not suitable for agriculture activities but favorable cool climate has encouraged the development of several agriculture activities. As time move on, most of the forest area was intensively used for vegetable cultivation. According to Abdullah et al. (2005), vegetable cultivation has total 47 %, followed by tea 44 %, flowers 7 % and fruits 1 %.

Other than that, agriculture activities performed in Cameron Highland are aided by chemical control in order to maintain and stimulate the growth of plant. Therefore, large input of chicken manure, chemical fertilizer and other agrobiocides such as lime, burnt rice husk, soymeal and chicken dung are added to sustain vegetable cultivation as part of intensive cropping cycle (Munisamy et al., 2013). Most of the chemicals contain heavy metals and tend to accumulate over time in soil and disperse in the ambient environment. Farmers who used pesticides and fertilizer are at risk to expose to the heavy metal. Spraying, dusting and fumigation allow farmers to inhaled the heavy

metals fumes and gases into the body, it become worst if the farmers did not wear PPE which might increase the chances to ingest the contaminant soil that accumulate in their fingernails. Without the PPE, the heavy metal fumes that disperse in the air might easily absorbed by the human skin. Besides, most of the farmers preferred to work with naked foot increase the possibility in exposure.

According to Mielke et al. (1999), the issue of heavy metal poisoning has been described as a 'silent epidemic'. This is because unless the toxic level is high enough if not the symptoms of poisoning are not apparent and maybe show only in adulthood, making the number of undiagnosed cases very high. Chronic exposure to high level of heavy metals elements can cause chronic toxicity effect such as hypertension and renal toxicity (Hu, 2002). The chronic exposures are usually irreversible and often use to describe carcinogenicity, teratogenicity and mutagenicity. According to Morais (2012), lead has no essential function in human body. Lead in human will affect various vital organs such as kidney, lung and muscles. Accumulation of Pb will cause anemia, renal tubular dysfunction and lead to secondary effect on the gastrointestinal tract include nausea, anorexia and abdominal cramps (Ming-Ho, 2005).

As a result, those farmers have their responsibility to take their own safety and health into account while conduct dangerous work practice. According to McDonald (2009), 'safety is everyone job'. Therefore, farmers should aware about the Personal Protective Equipment (PPE) and possess good awareness and work practice in order to minimize the exposure to heavy metals.

1.3 PROBLEM STATEMENT

The challenge of producing enough food is in increasing trend and affecting most of the developing countries as they priorities support for industry ahead of agriculture. However, many country including Malaysia have realized the need for food security and self-sufficiency and have embarked on an intensive agricultural activities (Barrow et al., 2009). Most of the tropical highlands are used for crop and suffering from environmental damage due to intensive farming with little or no enforcement control. The Cameron Highland of Malaysia is one of the examples whereby large scale

of forest is clear and intensively used for vegetation and flower cultivation. According to Barrow et al. (2009), vegetables from Cameron Highland used to supply for half of Malaysian needed and generate foreign earnings between RM 56 million-RM 100 million from vegetables and RM 20 million from flower production. Furthermore, Cameron supplied over 60 % of Malaysia's vegetables and fruit in 2006 and generated considerable export earnings from vegetables and flowers (Personal Communication 2006 Cameron Highland Growers Association).

Pesticide pollution in Cameron Highland has attracted research attention for over 20 years and there have been many efforts putting in to control it (Barrow et al., 2009). Recent research has reported the widespread of illegal pesticides where they are cheaper and more effective compared to approve pesticides. Besides that, the main organic fertilizer such as chicken manures and sewage sludge (bio solids) are widely used by farmers where they contain heavy metals contaminants. The worse scenario is there is no Federal Limit on heavy metals contaminants that are generally applied to all fertilizer. These chemicals are utilized intensively by farmers to control the pest infection and sustain the growth of plant thus increase the rate of plantation production. As a matter of fact, although these chemicals are known to produce beneficial impacts as desired by the farmers, the heavy metals content of those chemical are still unknown (Barrow et al., 2009). In this case, farmers may indirectly expose to heavy metals element through inhalation, ingestion and absorption. The accumulation of heavy metal in our body may pose detrimental effect to human health and thus reduces the quality of life.

A recent study on heavy metal in hair and nails among farmers in Kelantan, Malaysia is carry out with the result showed that there was mean different of Pb levels between age group in hair samples ($p<0.05$). Futhermore, using te pearson correlation, the result indicated that were significant relationship between working experience and the level of Pb ($r = -0.020$, $p<0.05$) (Saat et al., 2013). Another study was conducted to assess the heavy metal among farmers of Muda Agricultural Development Authority in Malaysia. This study found that there were significant correlations between working period with level of lead and arsenic ($r =0.315$ and $r = 0.242$ respectively), and age with lead level ($r = 0.175$, $P<0.05$) (Ghazali et al., 2012).

This research will be conducted to assess the rate of heavy metals exposure among the farmers at Cameron Highland. The nail tissue will be collected due to their simple sample collection, storage and preparation of analysis. The nail is better biomarker in occupational exposure to pollutants compare to blood and urine due to their slow growth rate 3.47 mm per month and able to trace elements for the duration of 2 to 18 months (Ghazali et al., 2006). In addition, the highest level of heavy metals element among the farmers will be determined so that the workers will understand the importance of PPE to reduce the heavy metals from entering their body.

1.4 RESEARCH QUESTION

1. What are the contributing factors to the heavy metal accumulates among the farmers?
2. What are the levels of concentration of each farmer?
3. What is the highest level of heavy metal element that the farmers expose to?
4. What are the relationships between heavy metal and symptoms of heavy metal sensitivity that exist from the exposure of heavy metals among the farmers?

1.5 STUDY OBJECTIVE

1.5.1 General Objective

1. To assess the level of heavy metal concentration using nail as bio-indicator among the farmers in agriculture sector.

1.5.2 Specific Objective

1. To determine the risk factors associated with the heavy metal accumulates among the farmers.
2. To assess the rate of heavy metal concentration among the farmers.
3. To identify the most dominant element of heavy metals concentrations among the farmers.

4. To determine the relationship between heavy metal and symptoms of heavy metal sensitivity that associated from the exposure of heavy metal among the farmers.

1.6 RESEARCH HYPOTHESIS

H_1: The age of workers and working experience show significant influenced to the heavy metal accumulate among the farmers.

H_2: The heavy metals level showed significant high among the farmers.

H_3: The level of arsenic show significant higher among the farmers.

H_4: There are is an association between heavy metal and symptoms of heavy metal sensitivity that exist from the exposure of heavy metals among the farmers.

1.7 SIGNIFICANCE OF STUDY

Heavy metals exposure is known to be dangerous to the health status of workers and may affect the work performance. Therefore, the finding in this study about the rate of heavy metals exposure is essential to create awareness for the individual farmers. This research provides information on current level of heavy metals among the farmers so that they will be aware to heavy metals exposure during conduct their work. Hence, they can understand the dangerous scenario and increase their safety awareness by wearing appropriate PPE and practice safety manner in their task.

Similarly, this study can serve as an additional source of reference for future studies regarding occupational heavy metals exposure. In a previous study, they have been proven working as a farmer is at risk expose to heavy metals. The duration of work have significant influence to the level of heavy metals exposure. In addition, this study will help to strengthen the findings of future research and also able to allow more study be conducted to increase the level of knowledge, understanding and good practice towards heavy metals in industries and environment.

Moreover, the study of the factors that contributes heavy metals accumulation among the farmers will help them to prevent the reoccurrence of the problem. Besides, the farmers can minimize the exposure of heavy metals by job rotation or use of quality PPE to prevent the exposure to pesticides.

1.8 SCOPE OF STUDY

A cross-sectional study will be conducted to assess the rate of heavy metals exposure among the farmers. This study will focus on farmers who are working with fertilizer and pesticide for a prolonged time. A total of 32 workers took part in this study. The questionnaire was used consist of background of farmers, number of years working as farmers, and the related heavy metal sensitivity symptoms. The nail samples were collected by participants from all ten fingers and stored at room temperature prior to sample preparation.

The biological tissues (nails) of farmers from Cameron Highland were analyzed to trace different types of heavy metal elements. The analysis will be carried out by inductively coupled plasma-mass spectrometry (ICP-MS). The highest level of heavy metals will be determined and correlated with demographic factors.

1.9 OPERATIONAL DEFINITION OF VARIABLES

1.9.1 *Heavy metals:* Heavy metals are able to form positive ions in solution and their density is five times greater than water (Naja & Volesky, 2009). According to Duffus (2002) said that in relation to the position of the element in the periodic table, due to the relation to chemical properties of compounds that include the element. Most heavy metals are commonly associated with toxicity problems and environment pollution when they in soluble form (Chen, 2013).

1.9.2 *Anthropogenic*: An activity that is not naturally occurs in environment and is due to human activities. Examples of anthropogenic activities are mining, agriculture, manufacturing industry, and construction. These situations are often referring to certain events, actions or effect whose origin can be traced to the activities of individual.

1.9.3 *Xenobiotic:* Chemical or other stressor that does not occur or appear naturally in the environment and is generate via anthropogenic activities (Tatiya, 2011). According to Yates (2011) xenobiotic is a substance which is not normally produced or expected to be present found in an organism.

1.9.4 *Acute toxicity:* Short-term exposure (minutes, hours, and days) to toxicant with symptoms appear or worsen rapidly.

1.9.5 *Chronic toxicity:* Long-term exposure (weeks, months, and years) to hazardous source which the diseases appear over an extended period of time.

1.9.6 *Pesticides*: Pesticide is any natural or synthetic compounds which are used by man to control pest. The pest may be insects, plant disease, fungi, weeds, nematodes, and snails. The term pesticide covers a wide range of compounds including insecticides, fungicides, herbicides, rodenticides, molluscicides, and plant growth regulators (Aktar et al., 2009).

1.9.7 *Fertilizers:* Fertilizers are substances that are either added to soil or applied directly to plants to enhance the growth and strength of plants (Daniel et al., 2004). There are two types of fertilizers organic fertilizers and inorganic fertilizers. Organic fertilizers made from natural compound such as

animal manure, sea weed, peat moss and guano while inorganic fertilizer are usually mix of various chemicals such as ammonia, sodium nitrates, rock phosphate, and muriate of potash (Aktar et al., 2007).

1.10 CONCEPTUAL FRAMEWORK

Figure 1.1 shows the conceptual framework of this study which intended to study the relationship between the heavy metal exposure and health effect that poses among the farmers. The level of heavy metal will be assessed using biological material. The nail of the workers will be collected and analyzed using (ICP-MS).

Occupational factors such as working duration will be considered in this study. The study of occupational factors is to understand whether those factors will affect the level of heavy metal accumulation in the workers body. Personal factors such as age also will be considered in this study.

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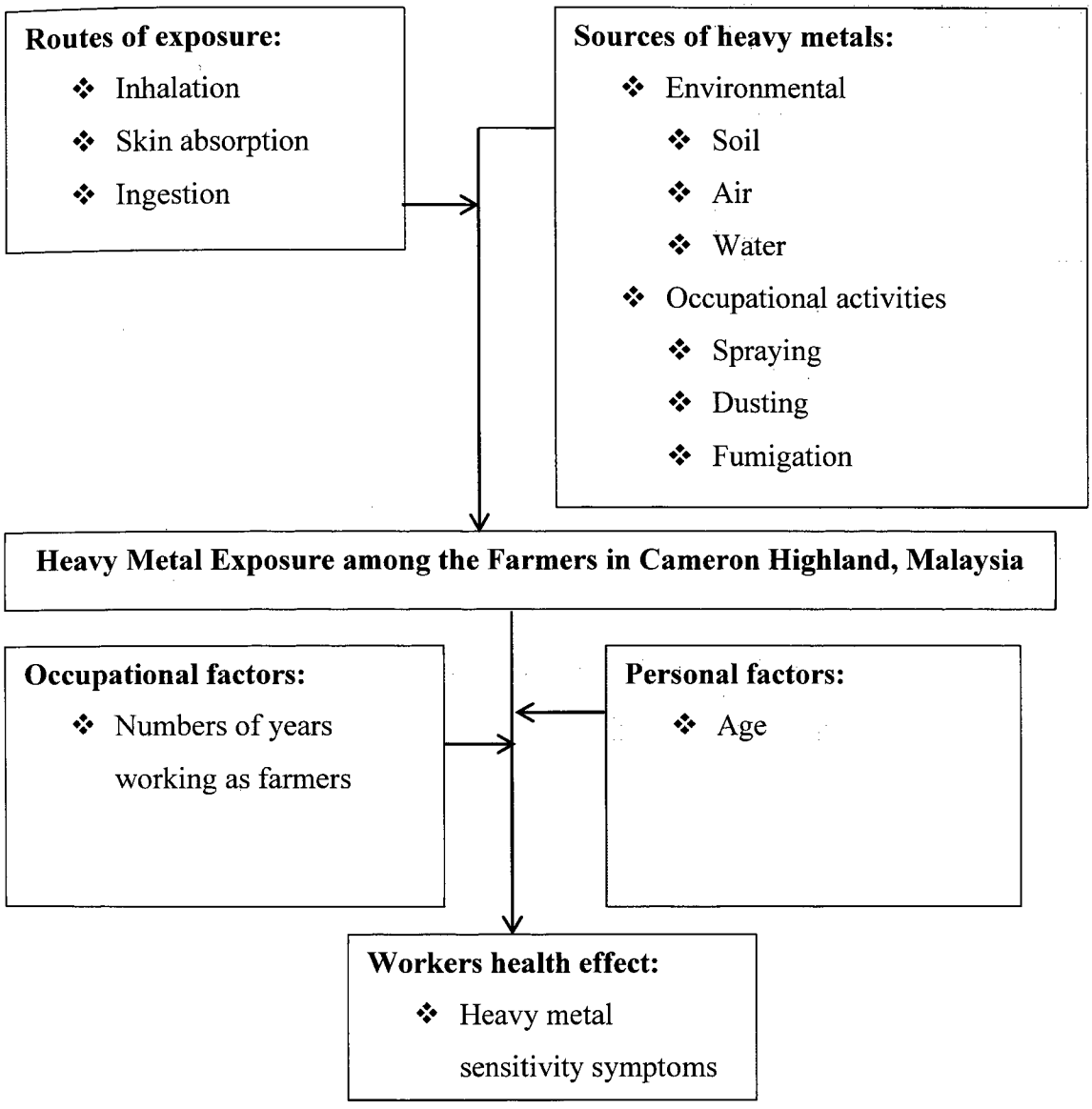

Figure 1.1: Factors associated with heavy metal exposure and farmers health impairment

Notes:

❖	Variable that will be studied

CHAPTER 2

LITERATURE REVIEW

2.1 INTRODUCTION

In this chapter, the past researches related to this study will be discussed in order to get more knowledge and related information. This chapter mainly focuses on the definition of heavy metal, sources of heavy metal, routes of exposure, and health effect of heavy metal. Hence, this chapter is essential in order to understand the heavy metal in detail and they way to minimize the exposure towards heavy metal.

2.2 DEFINITIONS OF HEAVY METALS

Heavy metal is a general collective term applied to the group of metals and metalloids with an atomic density five times greater than water (Chen, 2013). They are particularly toxicological important. Most of the metallic elements play an essential role in the function of living organisms; they supply a nutritional requirement and fulfill a physiological role (Naja & Volesky, 2009). Heavy metals can be defined as having the molecular weight above 40 whereas any metal that molecular weight less than 40 is called light metal (Chen, 2013). Heavy metal is a very imprecise term that used loosely to refer the element and its compounds. (Duffus, 2002) It is based on categorization of different criteria:

a) Good conductor of electricity.
b) High thermal conductivity.
c) High density, which is above $5g/cm^3$.

d) Malleability and ductility, able to draw into sheet and wires.
e) Normally has oxidation numbers 0, +1, +2, +3, and +6.
f) A zero-valent metal is present in solid form. Mercury is the only exception.
g) Heavy metal may exist in nature as metal oxides, metal carbonate, or metals sulfate.
h) Heavy metal may present in nature as soluble species that are complexes with various anionic substances called ligands.

Heavy metals can be divided into three groups according to their importance to living things such as essential light metals, essential heavy metals, and toxic heavy metals.

2.2.1 Essential Light Metals

Light metals are various metals that have low atomic mass such as sodium (Na), potassium (K), magnesium (Mg), and calcium (Ca) (Chen, 2013). There present of those light metals are essential in sustaining biological growth. Both sodium and potassium have high concentration in our body and participate in various metabolic processes (Chen, 2013). However, high sodium intake may increase the risk of mortality from cardiovascular disease (Umesawa et al., 2008). High concentration of potassium in human body can cause adverse effect such as nausea, diarrhea, irregular heartbeat, and vomiting. The recommended daily allowance (RDA) for sodium is 2,400 mg/day while potassium is 3,500 mg/day (Chen, 2013).

Magnesium is needed for building the skeleton and help muscle to carry out their function; it is widely distributed throughout the human body (Landner & Reuther, 2004). Magnesium deficiency will lead to cardiovascular disease, insomnia, anxiety, muscle and joint pain whereas if overdose or overconsumption will cause low stomach acid, depression, low blood pressure and higher risk for several cancers (Umesawa et al., 2008). The RDA is 400 mg/day (Chen, 2013).