UNIVERSITI PUTRA MALAYSIA

HEAVY METALS IN SOIL FROM URBAN AREAS OF KLANG, MALAYSIA, AND THEIR HEALTH RISK ASSESSMENT USING IN VITRO DIGESTION METHOD

NURUL SYAZANI YUSWIR

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By

NURUL SYAZANI BINTI YUSWIR

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfilment of the Requirements for the Degree of Master of Science

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the Degree of Master of Science

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October 2015

Chair : Sarva Mangala Praveena, PhD
Faculty : Medicine and Health Sciences

Background: Klang District is considered as one of the area that undergoes a rapid urbanization and surrounded by land and sea based anthropogenic activities. These anthropogenic activities have given various impacts on environmental and human health. **Objective:** Thus, the aim of this study was to determine the total and bioavailable of heavy metal concentrations (Al, Fe, Zn, Cu, Co, Cd, Pb and Cr) in urban surface soil samples. In addition, this study also aimed to determine possible sources in Klang District urban surface soil using multivariate analysis. Besides, this study also aimed to determine health risks posed by bioavailable of heavy metal in urban soil on adult and children. **Methodology:** A total of 76 urban soils in Klang District have been sampled and analyzed for total and bioavailable of heavy metal concentrations (Al, Fe, Zn, Cu, Co, Cd, Pb and Cr) using Inductively Coupled Plasma-Optical Emission Spectrometry. Total heavy metal concentration was determined using *aqua regia* digestion method while bioavailable of heavy metal concentration was determined using Physiologically Based Extraction Test (PBET) *in vitro* digestion method. **Result:** Results showed that the bioavailable of heavy metal concentrations were in the order of Al (25 mg/kg), Fe (6.7 mg/kg), Zn (5.6 mg/kg), Cu (3.0 mg/kg), Co (0.22 mg/kg), Cd (0.14 mg/kg), Pb (0.11 mg/kg) and Cr (0.10 mg/kg), while concentration of total heavy metal were in the order of Fe (9090 mg/kg), Al (6171 mg/kg), Cu (294 mg/kg), Zn (276 mg/kg), Pb (53 mg/kg), Cr (16 mg/kg), Co (1.2 mg/kg) and Cd (0.71 mg/kg). From the Spearman correlation coefficient (r) value, significant correlation were observed for Al-Fe (r = 0.681), Cd-Co (r = 0.495), Cu-Zn (r = 0.232), Fe-Pb (r = 0.260), Fe-Zn (r = 0.239), Al-Cu (r = -0.503), Co-Pb (r = -0.241) and Cu-Fe (r = -0.492). Spearman correlation output showed that heavy metal such as Al and Fe may come from natural sources while heavy metal such as Cd, Co, Cr and Cu may come from anthropogenic sources. Cluster Analysis output showed that these heavy metals can be classified into four clusters namely Cluster 1 which consisted of Cd, Cr, Co and Pb, Cluster 2 consisted of Zn and Cu, Cluster 3 consisted of Fe and Cluster 4 consisted of Al. For Clusters 1 and 2, anthropogenic activities were believed as the sources while for Clusters 3 and 4, the heavy metals originated from natural sources. For the health risks, the results of Hazard Quotient and Total Risk showed that heavy metal contamination via soil ingestion pathway in Klang District may not pose carcinogenic and non-carcinogenic risk to human adult, but it may pose carcinogenic and non-carcinogenic risks to children. **Conclusion:** Output from this study can be used to fill the knowledge gap on effect of heavy metal and potential health risks due to heavy metal exposure in urban area especially in Malaysia.

Keywords: Urban soil, heavy metal, total, bioavailable, health risks

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia
sebagai memenuhi keperluan untuk Ijazah Master Sains

LOGAM BERAT DALAM TANAH DARI KAWASAN BANDAR KLANG, MALAYSIA, DAN PENILAIAN RISIKO KESIHATAN MENGGUNAKAN KAEDAH PENGHADAMAN *IN VITRO*

Oleh

NURUL SYAZANI BINTI YUSWIR

Oktober 2015

Pengerusi : Sarva Mangala Praveena, PhD
Fakulti : Perubatan dan Sains Kesihatan

Pendahuluan: Daerah Klang dianggap sebagai salah satu kawasan yang mengalami pembangunan pembandaran yang pesat dan dikelilingi oleh aktiviti antropogenik berdasarkan tanah dan laut. Aktiviti-aktiviti antropogenik ini telah memberikan pelbagai kesan terhadap alam sekitar dan juga kesihatan manusia. **Objektif:** Oleh itu, tujuan kajian ini adalah untuk menentukan kepekatan total dan kebolehdapatan logam berat (Al, Fe, Zn, Cu, Co, Cd, Pb dan Cr) di dalam permukaan sampel tanah di kawasan bandar. Di samping itu, kajian ini juga bertujuan untuk mengenal pasti hubungan antara kebolehdapatan logam berat, dan sumbernya di dalam permukaan tanah di bandar Klang dengan menggunakan analisis multivariat. Selain itu, kajian ini juga bertujuan untuk menentukan risiko kesihatan pada orang dewasa dan kanak-kanak di bandar Klang melalui kaedah kebolehdapatan logam berat di dalam tanah. **Metodologi:** Sebanyak 76 sampel tanah di kawasan bandar di Daerah Klang telah disampel dan dianalisis untuk menilai kandungan kepekatan total dan kebolehdapatan logam berat (Al, Fe, Zn, Cu, Co, Cd, Pb dan Cr) menggunakan Inductively Coupled Plasma-Optical Emission Spectrometry. Total kepekatan logam berat telah ditentukan dengan menggunakan kaedah "*aqua regia*" manakala kebolehdapatan kepekatan logam berat telah ditentukan dengan menggunakan kaedah Physiologically Based Extraction Test (PBET). **Hasil kajian:** Keputusan menunjukkan bahawa kebolehdapatan logam berat adalah dalam susunan Al (25 mg / kg), Fe (6.7 mg / kg), Zn (5.6 mg / kg), Cu (3 mg / kg), Co (0.22 mg / kg), Cd (0.14 mg / kg), Pb (0.11 mg / kg) dan Cr (0.10 mg / kg), manakala kepekatan total logam berat adalah dalam susunan Fe (9090 mg / kg), Al (6171 mg / kg), Cu (294 mg / kg), Zn (276 mg / kg), Pb (53 mg / kg), Cr (16 mg / kg), Co (1.2 mg / kg) dan Cd (0.71 mg / kg). Daripada nilai pekali korelasi Spearman (r), hubungan yang signifikan adalah Al-Fe ($r = 0.681$), Cd-Co ($r = 0.495$), Cu-Zn ($r = 0.232$), Fe-Pb ($r = 0.260$), Fe-Zn ($r = 0.239$), Al-Cu ($r = -0.503$), Co-Pb ($r = -0.241$) dan Cu-Fe ($r = -0.492$). Nilai pekali korelasi Spearman (r) di antara 0.681 - 0.503 menunjukkan bahawa logam berat seperti Al dan Fe mungkin datang daripada sumber semula jadi manakala logam berat seperti Cd, Co, Cr dan Cu mungkin datang daripada sumber antropogenik. Dapatan daripada Analisis Kluster menunjukkan bahawa logam berat ini boleh dikelaskan kepada empat kelompok di mana Kluster 1 terdiri daripada Cd, Cr, Co dan Pb, Kluster 2 terdiri daripada Zn dan Cu, Kluster 3 terdiri daripada Fe dan Kluster 4 terdiri daripada Al. Untuk Kluster 1 dan 2, aktiviti antropogenik dipercayai sebagai sumber kepada kandungan logam berat tersebut manakala bagi Kluster 3 dan 4, logam berat tersebut mungkin berasal daripada sumber semula jadi. Bagi Penilaian Risiko Kesihatan, keputusan Hazard Quotient dan Total Risk menunjukkan bahawa pencemaran logam berat melalui tanah di daerah Klang tidak mungkin menimbulkan risiko karsinogenik dan bukan karsinogenik kepada golongan dewasa, tetapi ia boleh menimbulkan risiko karsinogenik dan bukan karsinogenik kepada kanak-kanak. **Kesimpulan:** Hasil kajian ini boleh digunakan bagi mengisi jurang pengetahuan tentang kesan pencemaran tanah dan risiko kesihatan yang berpotensi di hadapi oleh populasi hasil daripada pendedahan logam berat di kawasan bandar terutamanya di Malaysia.

Kata Kunci: Tanah bandar, logam berat, total, kebolehdapatan, risiko kesihatan

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This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Masters of Science .The members of the Supervisory Committee were as follows:

Sarva Mangala Praveena, PhD
Senior Lecturer
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Chairman)

Zailina Hashim, PhD
Professor
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Member)

Ahmad Zaharin Aris, PhD
Associate Professor
Faculty of Environmental Sciences
Universiti Putra Malaysia
(Member)

BUJANG BIN KIM HUAT, PhD
Professor and Dean
School of Graduate Studies
Universiti Putra Malaysia

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Signature: ___________________
Name of
Chairman of
Supervisory
Committee : Dr Sarva Mangala Praveena

Signature: _____________________
Name of
Member of
Supervisory
Committee : Professor Zailina Hashim

Signature: _____________________
Name of
Member of
Supervisory
Committee : Associate Professor Ahmad Zaharin Aris

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LIST OF ABBREVIATIONS

µm	Micrometer
Ag	Silver
Al	Aluminium
As	Arsenic
AT	Averaging Time
At_{cancer}	Averaging Time For Cancer
$At_{non\ Cancer}$	Averaging Time For Non Cancer
ATSDR	Agency for Toxic Substances and Disease Registry
BW	Body Weight
CA	Cluster Analysis
Cd	Cadmium
CDI	Chronic Daily Intake
Co	Cobalt
Cr	Chromium
CSF	Cancer Slope Factors
Cu	Copper
ED	Exposure Duration
EF	Exposure Frequency
HCl	Hydrochloric Acid
Hg	Mercury
HNO_3	Nitric Acid
HQ	Hazard Quotient
HRA	Health Risk Assessment
IARC	International Agency For Research On Cancer
ICP-OES	Inductively Coupled Plasma-Optical Emission Spectrometry
Ingr	Ingestion Rate
IRIS	Integrated Risk Information System
LCR	Lifetime Cancer Risk
NaCl	Sodium Chloride
NaOH	Sodium Hydroxide
Ni	Nickel
Pb	Lead
PBET	Physiologically Based Extraction Test
RAIS	Risk Assessment Information System
Rfd	Reference Dose
SBET	Simplified Bioaccessibility Extraction Test
SBRC	Solubility/Bioavailable Research Consortium
Se	Selenium
SF	Slope Factor
SHIME	Simulator Of Human Intestinal Microbial Ecosystems Of Infants
SPSS	Statistical Package For Social Science
TR	Total Risk
USDA	United States Department Of Agriculture
USDOE	United Stated Department Of Environment
USEPA	United States Environmental Protection Administration
Zn	Zinc

CHAPTER 1

INTRODUCTION

1.1 Introduction

Malaysia is one of the countries in Southeast Asian that is currently undergoing a rapid urbanization. Urbanization in Malaysia rose dramatically from 26.8 % to 61.8 % between 1970 to 2000 (Jaafar, 2004). In addition, Malaysia is also projected to be an urban society with the majority (over 70 %) of the country's total population living in its cities (Hassan, 2009). Figure 1.1 shows the level of urbanization in Malaysia by Department of Statistics Malaysia from 1980 to 2010 while Figure 1.2 shows the distribution of urban centres in Peninsular Malaysia in 1957 and 2000. Over 60% of the Malaysian population are concentrated in the west part of Peninsular Malaysia. Thus, this area has most of the development activities (Bin, 2011). However, this rapid urbanization has produced environmental pollutions such as water and air pollution that have given impacts on vegetation, river and urban soil (Zarcinas et al., 2004; Henderson and Wang, 2007). According to Shazili et al., (2006), manufacturing along with port and shipping activities are the major contributors of pollution in the environment of west Peninsular Malaysia.

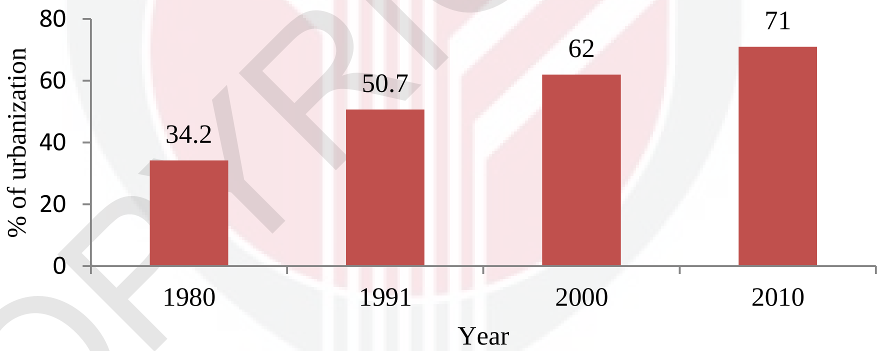


Figure 1.1: Percentage of urbanization in Malaysia
Source: Department of Statistics Malaysia, (2014)

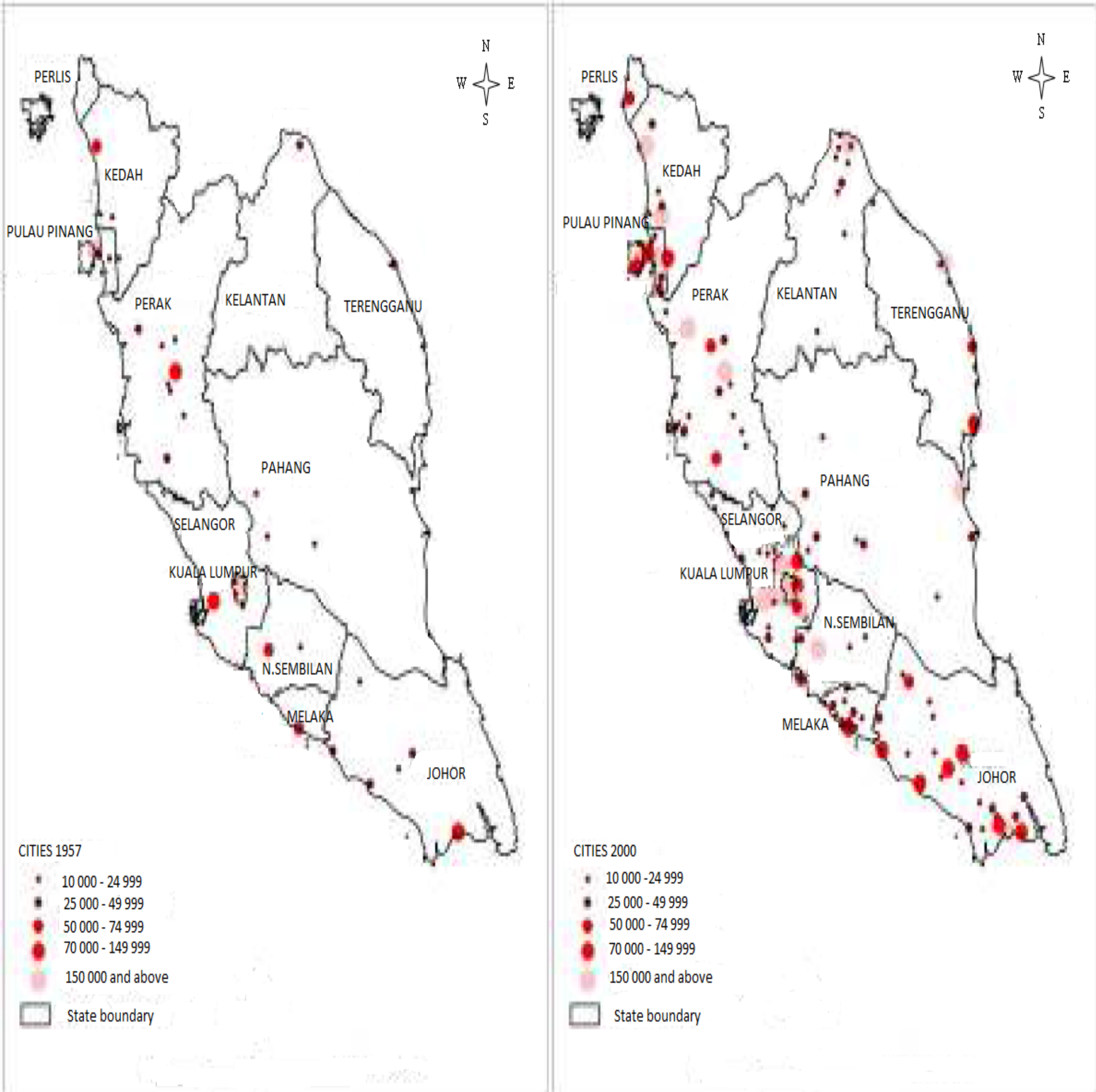


Figure 1.2: Distribution of urban centres in Peninsular Malaysia, 1957 and 2000
Source: Masron et al., (2012)

According to Wei and Yang (2010) and Foo et al., (2008), urban soils have become polluted due to the rapidly urbanization. Rapid urbanization has polluted the urban soil due to the land use activity by human. In urbanization process, raw land is converted and covered with pavement. The impervious surfaces created by buildings and pavement causes rainwater and snowmelt that carries environmental pollutants to flow quickly over the landscape, rather than soaking naturally into the urban soil or being absorbed by plants (Foster, 1990). There are many pollutants that can cause pollution in urban soil such as dioxin, persistent organic pollutants (POPs) and heavy metal (Habe et al., 2001; Foo et al., 2008; Wang et al., 2012). Among these pollutants in urban soil, heavy metal is one of the major environmental problems throughout the world because heavy metal has significant toxic effects and bioaccumulate in human, animals, microorganisms and plants (Mehdi et al., 2003; Majid et al., 2012). According to Coen et al., (2001) heavy metals can accumulate in human tissue as a result of rapid urbanization pollution and give toxic effect. In addition, bioaccumulation of heavy metal over prolonged time can become hazardous to human health after entering in their body system (Mehdi et al., 2003).

Studies done by researchers have stated that urban soils worldwide have high concentration of heavy metal (Wong et al., 2006; Thornton et al., 2008; Luo et al., 2012). In urban soil, heavy metal can be present in both natural and anthropogenic forms. Natural forms of heavy metal due to weathering of rock minerals are present at relative low concentrations. Mineral weathering of rocks is the primary source of background heavy metal in urban soil (Dakane, 2012). However, in recent years a number of anthropogenic sources have implied notable contributions to the increase of the heavy metal concentrations (Luo et al., 2011). This is supported by Wei and Yang (2010) in China, stating that the pollution sources of heavy metal in urban soil are mainly derived from anthropogenic activities. In urban soils, the anthropogenic sources of heavy metals include traffic emissions, industrial emissions, domestic emission, weathering of building and pavement surface and atmospheric deposition. Traffic emissions include vehicle exhaust particles, tire wear particles, weathered street surface particles and brake lining wear particles meanwhile industrial emissions includes the power plants, coal combustion, metallurgical industry, auto repair shop and chemical plants (Foo et al., 2008). A review done by Afroz et al., (2003) in Malaysia stated that the general major sources of pollution in Malaysia are from motor vehicles, open burning and industrial emissions. Additionally, Foo et al., (2008) in Kuala Terengganu (Terengganu) stated that vehicular emission is the most commonly known significant and increasing source of soil pollution in urban environment.

Elevation of heavy metal in urban soil due to anthropogenic sources can pose significant health risks to human. Heavy metal in urban soil can enter into human body from three different exposure pathways which are by accidental ingestion (Luo et al., 2011; Okorie et al., 2011), inhalation (Laidlaw and Filippelli, 2008; Schmidt, 2010) and dermal contact (Siciliano et al., 2009). Accidental ingestion of heavy metal in urban soil can occur due to mouthing behaviour, contacting with dirty hands or eating dropped food (USEPA, 2011). Moreover, according to Doyle et al., (2012) stated that accidental ingestion may occur through the inadvertent ingestion of soil or dust particles that adhere to food, objects and hands, or the deliberate ingestion of soil such as soil pica and geophagy, which is considered to be relatively uncommon in the general population. However, geophagy can be a relatively common practice in indigenous peoples on all continents (Doyle, 2012). Meanwhile, inhalation of heavy metal in urban soil requires the heavy metal to be present in air and then be inhaled whereas dermal absorption requires contact between heavy metal and skin during contact with urban soil (WHO, 2010). However, accidental ingestion is considered as an important pathway for heavy metal to enter the human body and is the largest area of concern in exposure route pathway in soil followed by dermal and inhalation (Paustenbach, 2000). Moreover, study by Intawongse et al., (2006) also suggest that accidental ingestion is an important exposure routes to humans from potentially harmful pollutants in soil including heavy metal.

1.2 Problem Statement

In Malaysia, Klang is located in west part of Peninsular Malaysia which is considered as one of the area that undergoes rapid urbanization (Bin, 2011). However, this rapid urbanization in Klang has given impacts on environment such as river and urban soil and produced environmental pollutions such as water and soil pollution (Zarcinas et al., 2004). Beside rapid urbanization, Klang is also an area that is surrounded by land and sea based anthropogenic activities. According to Shazili et al., (2006), anthropogenic activities in West Malaysia including Klang area has become one of the major contributor to environmental pollution in that area including heavy metal. This is supported by Yap et al. (2003), who stated that the major input sources of heavy metal in Klang District can directly contribute to high level of heavy metal concentrations in Klang waters, sediment, soil and biota. Other than that, studies by Ismail et al., (1993) and Yap et al., (2003) showed high concentrations of Cd, Pb, Cu and Zn detected in Port Klang sediments. In addition, studies done in Klang area by Lihan and Rahim, (2006) and Dayang et al., (2013) have also stated that the total heavy metal concentrations in urban soils sampled from Klang were highest for Cu, Zn, Ni, As, Cr and Cd concentrations.

So far, studies only employed total heavy metal concentration and explained about the pollution status, spatial behavior and speciation of heavy metal in Klang areas (Yap et al., 2003; Lihan and Rahim, 2006; Sany et al., 2012; Naji and Ismail., 2012; Dayang et al., 2013). However, these studies only measure the total heavy metal concentration in their soil sample and focusing on heavy metal pollution status of the soil. However, there are no any studies related to bioavailable of heavy metal in Klang urban soil which can be used to indicate human health.

1.3 Study Justification

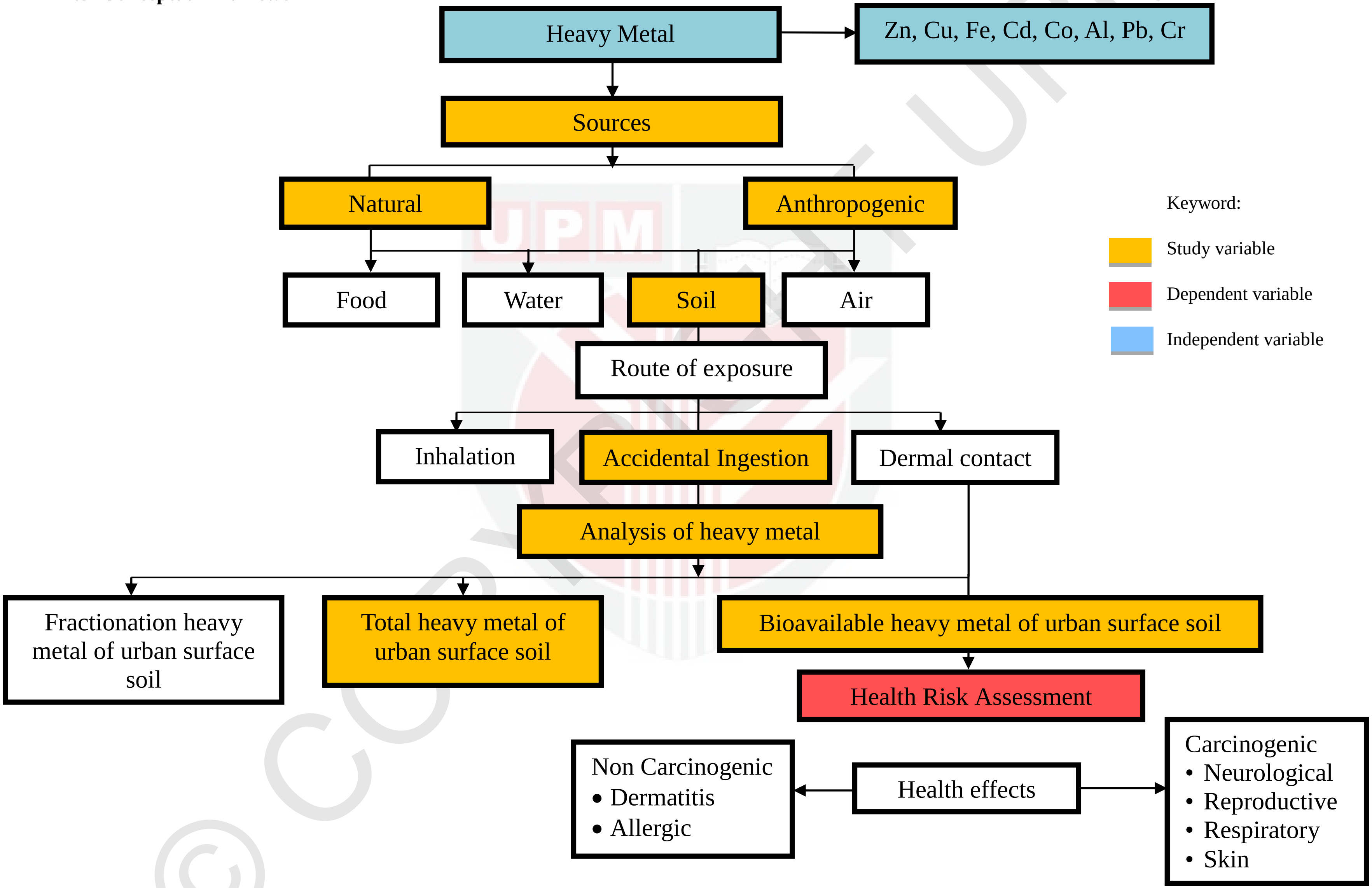
Rapid urbanization and anthropogenic activities in Klang area have polluted the urban soil. Studies done by Ismail et al., (1993), Yap et al., (2003), Lihan and Rahim, (2006) and Dayang et al., (2013) found a high concentration of heavy metal in Klang waters, sediment, soil and biota. From time to time, there are an increment in rapid urbanization and anthropogenic activities in Klang area. This situation will increase the concentration of heavy metal in Klang urban surface soil. Thus, there is a need for further research that evaluates the recent concentrations of heavy metal in Klang area and determines the possible sources of heavy metal pollution in Klang urban surface soil.

So far, heavy metal studies on urban soil in Klang have only focused on pollution status, spatial behaviour and speciation of heavy metal using total heavy metal concentration. Thus, there was a need for further research on Klang urban soil that evaluates impacts of heavy metal on human health by assessment of bioavailable form and incorporate health risk assessment. Bioavailable of heavy metal measures heavy metal concentration that is absorbed in gastrointestinal tract of human (Oomen et al., 2003). Bioavailable of heavy metal concentration would give accurate results on human health impacts via health risk assessment compared to total heavy metal.

1.4 Study Contribution

This study output would fill the gap about Klang area which includes information on the impact of heavy metal toward human health. Furthermore, findings from this study would also provide a baseline value for heavy metal contamination in urban soil and its impacts to human health that can be used in the recommendation for the management of heavy metals released to the environment. Findings from this study will also help in the policy making on development of housing area by providing a location for the type of land that are safe to be placed.

1.5 Conceptual Framework



1.6 Definition Of Term

1.6.1 Conceptual definition

Heavy metal

Any metallic element that has a relatively high density and is toxic or poisonous even at low concentration (Duruibe et al., 2007). Of the 92 naturally occurring elements, approximately 30 heavy metal are potentially toxic to humans such as Aluminium (Al), (Chromium) Cr, Cobalt (Co), Nickel (Ni), Copper (Cu), Arsenic (As), Selenium (Se), Argentum (Ag), Cadmium (Cd), Mercury (Hg) and Lead (Pb) (Morais et al., 2012).

Bioavailable

The fraction of an orally administered dose that reaches the systemic circulation (Versantvoort et al., 2004). There are 5 analytical methods used to determine the bioavailable of heavy metal which are Physiologically Bioavailable Extraction Test (PBET) method (British Geological Survey, UK), E DIN method (Ruhr Universitat Bochum, Germany), RIVM method (National Institute of Public Health and the Environment, The Netherlands), Simulator of Human Intestinal Microbial Ecosystems of Infants (SHIME) method (LabMET, Belgium) and TIM method (TNO Nutrition, The Netherlands) (Oomen et al., 2002).

Health Risk Assessment

Health risk assessment is a scientific process to estimate the probability of adverse health effects in humans who may be exposed to pollutants in contaminated environmental media. This process utilizes the tools of science and statistics to identify and measure a hazard, determine possible routes of exposure, and finally use that information to calculate a numerical value to represent the potential risk. A human health risk assessment consists of four steps. These steps include hazard identification, dose-response assessment, exposure assessment, and risk characterization (USEPA, 2002).

1.6.2 Operational definition

Heavy metal

Heavy metal (Zn, Cu, Fe, Cd, Co, Al, Pb, Cr) concentration in urban soil that are selected in this study was digested using *aqua regia* and Physiologically Based

Extraction Test method and then the concentration was determined and analyzed using Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES).

Bioavailable

Bioavailable of heavy metal in this study was measured using Physiologically Based Extraction Test (PBET) method. There are 5 analytical methods used in an *in vitro* digestion model to determine the bioavailable of heavy metal which are Physiologically Bioavailable Extraction Test (PBET) method (British Geological Survey, UK), E DIN method (Ruhr Universitat Bochum, Germany), RIVM method (National Institute of Public Health and the Environment, The Netherlands), Simulator of Human Intestinal Microbial Ecosystems of Infants (SHIME) method (LabMET, Belgium) and TIM method (TNO Nutrition, The Netherlands) (Oomen et al., 2002). Among 5 of the analytical methods used in an *in vitro* digestion model, PBET is one of the most widely used models.

Total

Total heavy metal in this study was measured using *aqua regia* method which is this method used the combination of hydrochloric acid and nitric acid.

Health Risk Assessment

In this study, health risk assessment was defined and done specifically on heavy metal exposure through the bioavailable of ingested urban soil. The assessment was evaluated through Hazard Quotient (HQ) for non-carcinogenic risks and Total Risk (TR) for carcinogenic risks to estimate the health risks of adult and children who are exposed to heavy metal through ingestion of urban soil.

Non carcinogenic risks were quantified by calculation of Hazard Quotient (HQ) as below: The oral route was the route of exposure observed in this study, therefore, oral RfD values from the IRIS database was used.

$$\text{Hazard Quotient (HQ)} = \frac{CDI}{RfD}$$

Where:
HQ = Non cancer Hazard Index of a health effect from intake of heavy metal
CDI = Chronic daily intake (mg/kg/day)
Oral RfD = Oral Reference dose of heavy metal (mg/kg/day)

Then, the hazard quotient value was compared with the following values of risk acceptability for non-carcinogenic health effects. In cases where the non-cancer HQ does not exceed unity (HI < 1), it is assumed that no chronic risks are likely to occur at the site.

Table 1.1: The risk acceptability for non-carcinogenic health effect (U.S. EPA, 2002)

Hazard Quotient (HQ)	
>1	Unacceptable
<1	Acceptable

As expressed earlier, carcinogens were assumed to not have an effective or safe threshold. Carcinogenic risk is expressed as slope factor (SF) and the following equation was used to quantify lifetime risk of cancer:

Total Risk (TR) = CDI x SF

Where:
CDI = Chronic daily intake (mg/kg/day)
SF = Slope factor (mg/kg/day)

Cancer slope factor values can be found on EPA's (2009) Integrated Risk Information System (IRIS). EPA guidelines specified that an acceptable risk is a lifetime cancer risk of no greater than 1 in 1,000,000. Thus, lifetime cancer risk (LCR) value would be referred to the following table to access the risk acceptability of carcinogenic health effect.

Table 1.2: The risk acceptability for carcinogenic health effects (USEPA, 2002)

Total Risk (TR)	
$<10^{-6}$	Clearly acceptable
10^{-6} to 10^{-4}	Acceptable
$> 10^{-4}$	Clearly unacceptable

1.7 Objectives

1.7.1 General Objective

To determine health risk assessment for bioavailable of heavy metal concentration (Al, Fe, Zn, Cu, Co, Cd, Pb and Cr) in urban soil of Klang District.

1.7.2 Specific Objectives

- To determine and compare the total and bioavailable of heavy metal concentration (Al, Fe, Zn, Cu, Co, Cd, Pb and Cr) in urban soil.
- To determine the correlations among bioavailable heavy metal concentrations.
- To identify possible sources of bioavailable heavy metal concentration in urban soil.
- To determine health risks through accidental ingestion exposure pathway of heavy metal exposure in urban soil based on element and population.

1.7.3 Hypotheses

- There was a significant difference between total and bioavailable of heavy metal concentration in urban soil.
- There were significant correlations between bioavailable heavy metal concentrations.

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