



UNIVERSITI PUTRA MALAYSIA

***OCCURRENCE OF HEAVY METAL IN PADDY SOIL AND RICE
GRAIN NOODLE ASSOCIATED WITH POTENTIAL HEALTH RISKS***

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ASSOCIATED WITH POTENTIAL HEALTH RISKS**



BY

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**Thesis submitted in fulfilment of the requirement for the
degree of Bachelor Science (Environmental and Occupational Health) from
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ABSTRACT

OCCURRENCE OF HEAVY METAL IN PADDY SOIL AND RICE GRAIN ASSOCIATED WITH POTENTIAL HEALTH RISKS

ZANJABILA BINTI HANAFI

Introduction: Rice is a staple food among Malaysian. However, little do we know about the heavy metal pollution in the paddy soil and rice grain, followed by its effect that can give potential health risks to human. **Objectives:** The aim of this study was to determine both total and bioavailable heavy metal (As, Cd, Pb, Cr, Cu) concentrations in paddy soil and rice grain associated with potential risks to environment and human health, respectively. **Method:** A total of 13 paddy soil and rice grain were extracted for total and bioavailable concentration using nitric acid digestion and hydrogen peroxide method and static *in-vitro* digestion model, respectively, for heavy metals determination using Inductively Coupled Plasma-Mass Spectrometry (ICP-MS). Descriptive analysis were calculated using Excel 2013. **Result:** Overall total concentration of paddy soil showed this decreasing trend Pb (0.0082 mg/kg) > Cr (0.0054 mg/kg) > Cu (0.0038 mg/kg) > As (0.0026 mg/kg) > Cd (0.0001 mg/kg), whereas the bioavailable concentration in the rice grain was Cr (0.2701 mg/kg) > Cu (0.0896 mg/kg) > Pb (0.0458 mg/kg) > As (0.0139mg/kg) > Cd (0.0053 mg/kg). Geoaccumulation index (Igeo) with background concentration (Igeo<0) for paddy soil samples. The bio-concentration factor (BCF) were found to be potential to accumulate heavy metal (>1) for all heavy metals. For Health Risk Assessment (HRA) involving rice grain samples, in adult and children, there were non-carcinogenic risks of Cu and Cr, where hazard quotient (HQ>1) and carcinogenic risk of As in adult where lifetime cancer risk (LCR >10⁻⁴). Whereas, in children, no risk were present. **Conclusion:** The study findings showed that although paddy soils were uncontaminated but potential health risks were present in most of the rice grain samples. This indicated that further research work to be focused on rice grains characteristics in order to understand its bioaccumulation capacity.

Keywords : *Paddy soil, Rice grain, Total, Bioavailable, Heavy metal, Risk, Environment, Human health*

ABSTRAK

KEJADIAN LOGAM BERAT DALAM TANAH PADI DAN BERAS DENGAN PERKAITAN RISIKO KESIHATAN YANG BERPOTENSI

ZANJABILA BINTI HANAFI

Pengenalan: Nasi merupakan makanan ruji masyarakat Malaysia. Namun, sedikit sekali fakta tentang pencemaran logam berat dalam tanah padi dan beras, yang dapat menyebabkan potensi risiko kesihatan kepada manusia. **Objektif:** Tujuan utama kajian ini adalah untuk menentukan kedua-dua konsentration logam berat (As, Cd, Pb, Cr, Cu) penuh dan bioavailabiliti di dalam tanah padi dan beras, serta mengaikkannya dengan risiko kesihatan yang berpotensi. **Metodologi:** Sebanyak 13 tanah padi dan beras digunakan untuk menentukan konsentration penuh dan bioavailabiliti dengan menggunakan kaedah pencernaan asid nitrik dan hidrogen peroxide untuk sampel tanah padi, serta kaedah statik pencernaan *in-vitro*. Untuk menentukan logam berat, alat 'Inductively Coupled Plasma-Mass Spectrometry (ICP-MS)'. Semua analisis penjelasan dijalankan menggunakan Excel 2013. **Keputusan:** Secara keseluruhan, konsentration penuh untuk tanah padi menunjukkan tren yang menurun Pb (0.0082 mg/kg) > Cr (0.0054 mg/kg) > Cu (0.0038 mg/kg) > As (0.0026 mg/kg) > Cd (0.0001 mg/kg), manakala konsentration bioavailabiliti untuk beras pula, Cr (0.2701 mg/kg) > Cu (0.0896 mg/kg) > Pb (0.0458 mg/kg) > As (0.0139mg/kg) > Cd (0.0053 mg/kg). Index Geoakumulasi (Igeo) adalah tidak tercemar Igeo<0) untuk semua sampel tanah padi. Faktor biokonsentration (BCF) pula menunjukkan semua logam berat mempunyai potensi untuk mengakumulasi logam berat (BCF>1). Untuk Penilaian Risiko Kesihatan (HRA) melibatkan sampel beras, pada orang dewasa dan kanak-kanak, ada risiko 'non-carcinogenic' pada Cu dan Cr, apabila 'hazard quotient' (HQ>1), serta risiko 'carcinogenic' As pada orang dewasa apabila 'lifetime cancer risk' (LCR >10⁻⁴). Manakala, pada kanak-kanak tiada risiko yang dapat dikenalpasti. **Konklusi:** Dapatan kajian menunjukkan, walaupun tanah padi tidak tercemar, namun potensi risiko kesihatan dapat dikenalpasti dalam kebanyakan sampel beras. Ini memberi tanda bahawa kajian lanjut perlu memberi perhatian kepada ciri-ciri beras, supaya dapat memahami tentang kapasiti akumulasi logam beratnya.

Kata kunci : Tanah padi, Beras, Konsentration Penuh, Bioavailabiliti, Logam Berat, Kesihatan Manusia

TABLE OF CONTENTS

DECLARATION	iii
ACKNOWLEDGEMENTS	v
ABSTRACT	vi
ABSTRAK	vii
LIST OF TABLES	x
LIST OF FIGURES	xi
LIST OF ABBREVIATIONS	xiii
CHAPTER 1 INTRODUCTION	1
1.1 Research Background	1
1.2 Problem Statement	4
1.3 Study Justification	5
1.4 Research Objective	6
1.4.1 General Objective	6
1.4.2 Specific Objectives	6
1.5 Study Hypothesis	6
1.6 Conceptual Framework	7
CHAPTER 2 LITERATURE REVIEW	8
2.1 Paddy	8
2.1.1 Paddy Soils And Rice Grains	9
2.2 Heavy Metal Bioavailability	10
2.3 Heavy Metal and its Health Effects	11
2.4 Heavy Metal Uptake Mechanism	14
2.5 Heavy Metal Accumulation	15
2.6 Method to analyze Heavy Metal Concentration in Paddy Soils and Rice Grains	16
2.7 Health Risk Calculation	17
2.8 Previous Studies of Paddy Soil and Rice Grain in Malaysia	18
CHAPTER 3 METHODOLOGY	21

3.1 Research Design	21
3.2 Study Location	21
3.3 Sample Collection	23
3.4 Study Methodology	23
3.4.1 Preparation Of Samples	24
3.4.2 Soil Samples Digestion	26
3.4.3 Rice Grain Samples Digestion	28
3.4.4 Geoaccumulation Index (Igeo)	33
3.4.5 Bioconcentration Factor (BCF)	34
3.4.6 Health Risk Assessment (HRA)	35
3.5 Quality Control and Quality Assurance	38
3.6 Data Analysis	38
CHAPTER 4 RESULT	39
4.1 The Total and Bioavailable Heavy Metal Concentration	39
4.2 Geoaccumulation Index (Igeo)	42
4.3 Bioconcentration Factor (BCF)	43
4.4 Health Risk Assessment (HRA)	44
CHAPTER 5 DISCUSSIONS	47
5.1 The Total and Bioavailable Heavy Metal Concentration	47
5.2 Geoaccumulation Index (Igeo)	48
5.3 Bioconcentration Factor (BCF)	48
5.4 Health Risk Assessment (HRA)	49
CHAPTER 6 CONCLUSION AND RECOMMENDATION	50
REFERENCES	51

LIST OF TABLES

	Page
Table 2.3.1 Agents Classified By The IARC Monographs, Volumes 1–120	11
Table 2.3.2 List of Classifications, Volumes 1–120	11
Table 2.3.3 List of Classifications by cancer sites with sufficient or limited evidence in humans, Volumes 1 to 120	12
Table 2.2.1 Agents Classified By The IARC Monographs, Volumes 1–120	11
Table 2.8.1 Summary of previous studies compared to present study	15-16
Table 3.4.4.1 Background value for each heavy metal	30
Table 3.4.4.2 The seven classes of Igeo index	30
Table 3.4.5.1 Bioconcentration Factor representation	31
Table 3.4.6.1 Variable used in health risk assessment	32
Table 3.4.6.2 RfD and CSF value for each heavy metal	33
Table 4.1.1 Total Heavy Metal Concentration in Paddy Soil samples	36
Table 4.1.2 Bioavailable Heavy Metal Concentration in Rice Grain	37
Table 4.2.1 Geoaccumulation Index (Igeo) of heavy metals in paddy soil	38
Table 4.3.1 Bioconcentration Factor of heavy metals in rice grain	39
Table 4.4.1 HQ and LCR values for adult (n=13)	41
Table 4.4.2 HQ and LCR values for children (n=13)	42

LIST OF FIGURES

		Page
Figure 1.6	Conceptual Framework of the research study	7
Figure 2.1	Picture of paddy plant	8
Figure 2.2	Three-step concept of heavy metal bioavailability in soils to plant	10
Figure 2.4	Example of a plant uptake mechanism via evapotranspiration	13
Figure 3.2.1	Study locations of Sabak Bernam, Tanjung Karang, Sekinchan, Yan and Besut paddy fields in Malaysia	19
Figure 3.4.1	Process flow of this research methodology	20
Figure 3.4.1.1	Paddy soil samples before air-dried and homogenized	21
Figure 3.4.1.2	Rice grain samples before air-dried and homogenized	21
Figure 3.4.1.3	Samples were air-dried for 3-4 days	21
Figure 3.4.1.4	Samples were homogenized by homogenized by mortar and pestle, and sieved through a 2mm mesh screen	22
Figure 3.4.1.5	Paddy soil samples after air-dried and homogenized	22
Figure 3.4.1.6	Rice grain samples after air-dried and homogenized	22
Figure 3.4.2.1	Process Flow of Nitric Acid and Hydrogen Peroxide Digestion Method for Soil Samples	23
Figure 3.4.2.2	Preparing the nitric acid before digestion experiment	24
Figure 3.4.2.3	Soil samples were weighed carefully to 1g	24
Figure 3.4.2.4	Soil samples after adding the nitric acid	24
Figure 3.4.2.5	Soil samples heated in the oven for 2 hours at 50°C	24
Figure 3.4.2.6	Soil samples were added hydrogen peroxide and left to digest overnight	24

Figure 3.4.2.7	Soil samples were filtered using 0.45µm Whattman filter paper	26
Figure 3.4.3.1	The human digestion process when ingesting the rice samples	27
Figure 3.4.3.2	Process Flow of Standardized Static <i>in-vitro</i> digestion model for Rice Grain Samples	27
Figure 3.4.3.3	Transferring the previous digested solution	28
Figure 3.4.3.4	Rice grain samples in the water-bath shaker	28
Figure 3.4.3.5	pH meter used to adjust the pH and the labelled tubes to transfer in every phase	28
Figure 3.4.3.6	Rice samples were finally filtered using 0.45µm Whattman filter paper	28
Figure 3.4.3.7	Heavy metal calibration solution and standard solution for ICP-MS	29
Figure 3.4.2.8	Both paddy soil and rice grain samples were brought to analyze by ICP-MS	29
Figure 4.2.1	Geoaccumulation index (I _{geo}) values in paddy soil.	38
Figure 4.3.1	Bioconcentration factor (BCF) values which involved paddy soil and rice grain samples concentration.	39

LIST OF ABBREVIATIONS

GDP	Gross Domestic Product
UNDESA	United Nation Department of Economic and Social Affairs
MOA	Ministry of Agriculture
ICP-MS	Inductive Coupled Plasma – Mass Spectrometry
IRIS	Integrated Risk Information System
IARC	International Agency of Research on Cancer
GERM	Geochemical Earth Reference Model
USEPA	U.S. Environmental Protection Agency
HQ	Hazard Quotient
LCR	Lifetime Cancer Risk
Igeo	Geochemical Index
BCF	Bioconcentration Factor
HRA	Health Risk Assessment
SSF	Simulated Salivary Fluid
SGF	Simulated Gastric Fluid
SIF	Simulated Intestinal Fluid
SOM	Soil Organic Matter

CHAPTER 1

INTRODUCTION

1.1 Research Background

Agriculture sector has continued to expand in 2015 with a contribution of 8.9 per cent to the Gross Domestic Product (GDP). Oil palm was a major contributor to the GDP of agriculture sector at 46.9 per cent followed by other agriculture (17.7%), livestock (10.7%), fishing (10.7%), rubber trees (7.2%) as well as forestry & logging (6.9%) in 2015, stated by the Department of Statistics Malaysia in Selected Agricultural Indicators, Malaysia, 2016.

Paddy is known to be the staple food among Malaysian community. Today, Malaysia is able to produce rice approximately 71.6% (Statistics Booklet of Plant (Food Crops of Sub-Sector) in Malaysia, 2015). Now, with the gradual increase of population growth, there is rising demand of food supply, especially rice. The population in Malaysia is 30,331,000 in 2015 and projected to continue growing linearly and is expected to reach approximately 43 million by the end of 2050 (UNDESA, 2015). The population in Malaysia is now growing with 2.7% per annum. Therefore, the estimated additional 45,000 tons of rice is required for each year to maintain the current consumption; which

is about 180,000 tonnes of rice per month. The increasing demand for food supply are due to two factors; world population growth and rising domestic incomes, especially those who live in developing countries like Malaysia (Azizan, 2015; Najim, et al., 2007). Nonetheless, in 2016 only, the production volume of rice in Malaysia have totaled to 1.77 million tons. The Ministry of Agriculture and Agro-Based Industry Malaysia (MOA), has set objectives are to ensure the rice production is at least 90% self-sufficiency level (SSL) and to ensure rice supply to consumers is sufficient, of quality and easily obtainable. (Ministry of Agriculture, 2016).

Thus, to meet the goal and demand, Department of Agriculture (DOA) of Malaysia, has provided various incentives to encourage rice production, such as subsidized seeds, fertilizer, pesticides, and irrigation (Grain and Feed Annual, 2016). The encouragement has made huge amount of fertilizers and pesticides has been used to meet the production number aim by the country. However, poor management practices and excessive use of chemicals overtime has led to environmental pollution such as soil, water and air. Pollutants like heavy metal is the most prominent because it affect agricultural production, food quality and eventually threaten human health (Jaishankar et al., 2014).

Heavy metals from natural and anthropogenic activities gathered in soil making both soil and paddy plant prone to contamination. This accumulation of heavy metals brings detrimental effects to human health through respiration, consumption and dermal contact absorption of rice (Jiao et al., 2015). Heavy metal in paddy soil can accumulate and concentrated in the soil for years due to its persistency. Soil act as sinks for heavy

metals and other pollutants (Omar et al., 2015). This accumulation can also lead to the uptake of heavy metals into the rice grain. Paddy plant has the ability to absorb any pollutant effectively from the water to the soil to the plant and even to the rice grain, due to its water-flooded condition and anaerobic state that results in higher mobilization or uptake. Normal production would take the raw harvested rice grains to the factory and packed for selling. There is no tendency that the heavy metal would disappear from the rice grain. When, it reaches the buyer, the seller, the retailer and the consumer, the rice grain would still be contaminated with certain heavy metals that would have an impact on human health. The health risk exist and it is still there in our food that we consumed, since heavy metal is a non-biodegradable and persistent in properties (Praveena & Omar, 2017).

Heavy metals like Arsenic (As), Cadmium (Cd), Lead (Pb), Chromium (Cr) and Copper (Cu) are heavy metals that are essential to be studied. International Agency of Research on Cancer (2012), has classified As and Cd in carcinogenic group, Pb in the possibly carcinogenic, Cr and Cu as non-carcinogenic. While, the main source of heavy metal accumulation in human body is through diet or ingestion of food contaminated by heavy metal. Different heavy metals have different toxicity and different organs that they accumulates based on their toxicity (Mahurpawar, 2015). According to Emese (2008), oral exposure to As could develop skin lesions like hyperkeratinization corns and warts, also known as arsenicosis. While, Cd could lead to gastro-intestinal toxicity and Pb affects the human's heme bio-synthesis pathway. Long term exposure to Cu and Cr might lead to severe health effects like abdominal pain, vomiting and dermatitis.

1.2 Problem Statement

As the country set objective to meet 90% self-sufficiency level (SSL), the encouragement to produce high amount of rice production has also increase the amount of fertilizers and pesticides used in the paddy fields. Habibah et al. (2011); Khairiah et al. (2012) & Roslaili et. al. (2015) had stated that heavy metal contained in fertilizers and pesticides has a higher chance to be present in the paddy field due to the high usage that are being used. With the high amount of heavy metal, it will lead to deterioration of paddy soil quality and high accumulation in the rice grain as discussed by Haris et al. (2013); Juen et al. (2014) & Khan et al. (2017).

Although, there is quite an amount of previous studies that has done research on heavy metal concentration in paddy soil and rice grain in Malaysia (Habibah et al., 2011; Khairiah et al, 2012; Yin et al., 2012; Haris et al., 2013; Norbaya et al., 2014; Juen et al., 2014; Hossain et al., 2015 ; Roslaili et. al., 2015; Khan et al., 2017). However, most of these studies only focuses on the quality of paddy soil and rice grain separately with limited consideration on the potential health risks to humans via rice consumption. Investigating heavy metal in paddy soil and rice grain is also crucial to understand the whole heavy metal accumulation from paddy soil to human health. Moreover, previous studies have focused on total form of heavy metal concentration only, which can overestimate the risk of heavy metal effect on human.

1.3 Study Justification

Thus, potential health risks to humans through rice consumption were assessed in this study along with the soil quality. Total and bioavailable heavy metal concentration in soil and grain samples were determined, followed by calculating the Bioconcentration factor (BCF) to detect the heavy metal accumulation.

Using the suitable heavy metal form will help us to assess the health risk and effects accurately. Thus, bioavailable concentration is found to be the most appropriate form to be used based on study done by Omar et. al (2015). According to the general interpretation in pharmacology, oral bioavailability is presently defined as the fraction of an orally administered dose that reaches the systemic circulation. (Oomen et.al, 2003). More realistic estimation of human heavy metal exposure was produced. Thus, this present study would investigated the bioavailable concentration of heavy metal in rice grain samples, along with the total heavy metal concentration in paddy soil samples.

1.4 Research Objective

1.4.1 General Objective

To determine the heavy metal concentration (As, Pb Cd, Cr, Cu) of paddy soil and rice grain associated with potential health risk to environment and human respectively in Sabak Bernam, Tanjung Karang, Sekinchan, Besut and Yan.

1.4.2 Specific Objectives

- a) To determine the total heavy metal concentration in paddy soil samples and bioavailable heavy metal concentration in rice grain samples
- b) To evaluate the geoaccumulation index (Igeo) for paddy soil samples and the bioconcentration factor (BCF) for paddy soil and rice grain samples, respectively
- c) To assess the potential human health risk through rice consumption

1.5 Study Hypothesis

- a) There is a potential health risk through rice consumptions to humans

1.6 Conceptual Framework

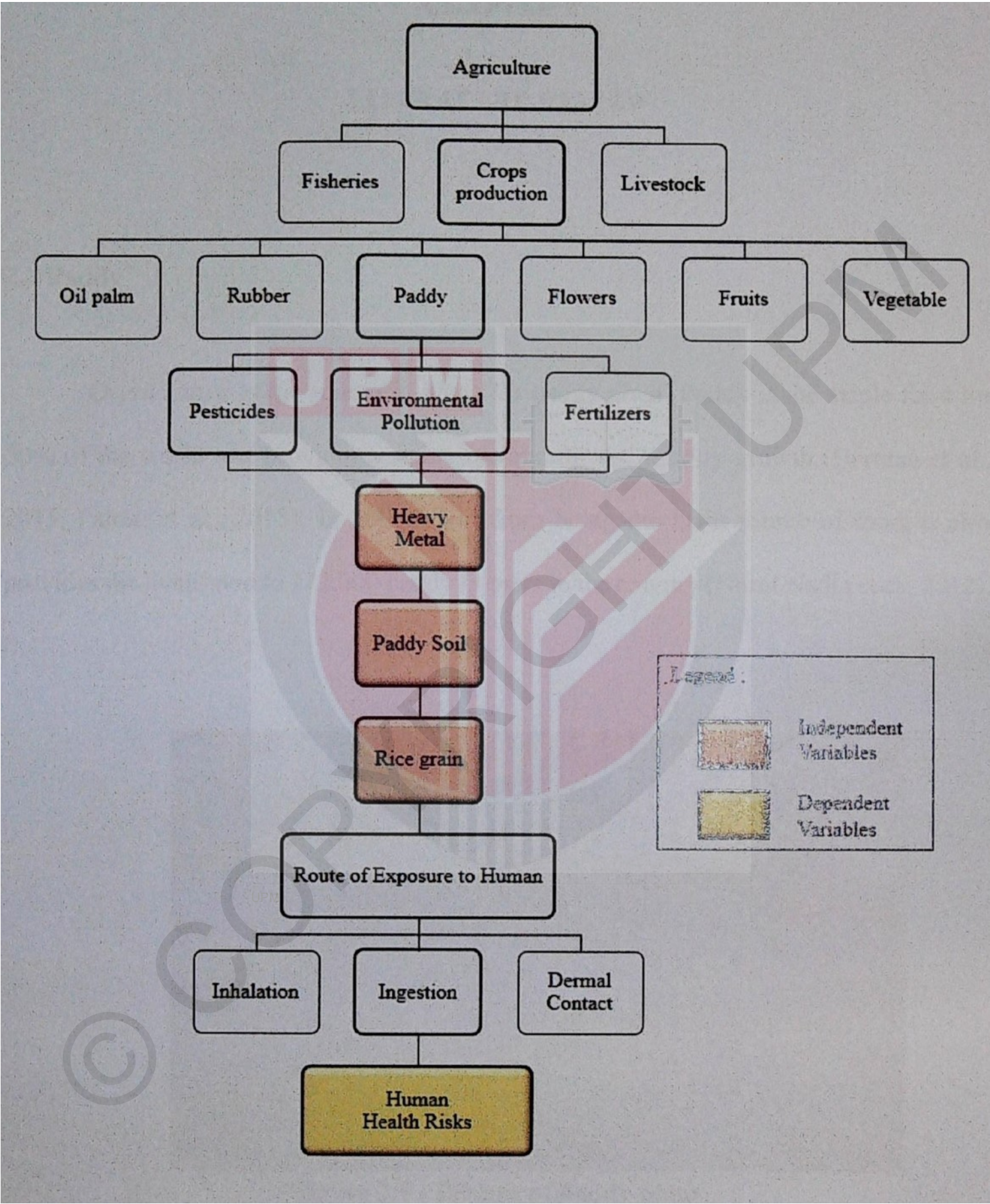


Figure 1.6 : Conceptual Framework of the research study

CHAPTER 2

LITERATURE REVIEW

2.1 Paddy

Oryza sativa is the scientific name for paddy plant. Paddy is the staple food for 50% of the world and provides a stable income for the country growth (Herman et al., 2015; Fahmi et al., 2013). In 2009, apart from being the main source of food, it also provides the livelihood to 172,000 paddy farmers in the country (Nurul Nadia et al., 2012).



Figure 2.1 : Picture of paddy plant

2.1.1 Paddy Soils And Rice Grains

Paddy soils consist of heterogeneous media comprising decomposed rock fragments, clay minerals, oxides of Fe, Al and Mn, organic materials, organo-metallic complexes and soil solutions (Alloway, 1995). In cultivated paddy areas, the soils cannot be considered natural, as they have been highly modified by management practices including levelling, cultivation, puddling, submerging and fertilizing (Prakongkep et al., 2008). Apart from supplying micronutrients for the plant growth, some of the fertilizers and pesticides might introduce toxic heavy metals into the soils.

Pollutants in the soil can be absorbed by the plants through their roots via vascular system. The uptake of metals in excessive amount may either cause harm to the plants itself or enter the food chain and accumulate when these plants are taken up. Metals accumulated in human body through the food chain can affect the central nervous system and cause many diseases (Coen et al. 2001; Zhao et al. 2009). Thus, rice is a crucial to be investigated as rice has been dominating staple food for more than two billion people especially from Asia (World Health Organization., 2003). World Health Organization (2003) has indicated that average daily intake of rice per person ranges from 9 g in west countries and 600 g in Asia which clearly showed that rice contribution to toxic elements exposure in human is crucial to be investigated.

2.2 Heavy Metal Bioavailability

There are various definitions on the term bioavailability. The first definition for heavy metal bioavailability is the available amount of the total heavy metal content in the soil. Secondly, the fraction of dissolved heavy metal that is controlled by physiological process and transfer from soil to plant. Thirdly, a physiologically induced bioaccumulation or biological effect of heavy metal within plants or other organisms which depends on a number of complex processes including translocation, metabolism, and detoxification. Simply said, the heavy metal can exist, accumulate, pollute and mobile from the soil to grains (Kim et al., 2015).

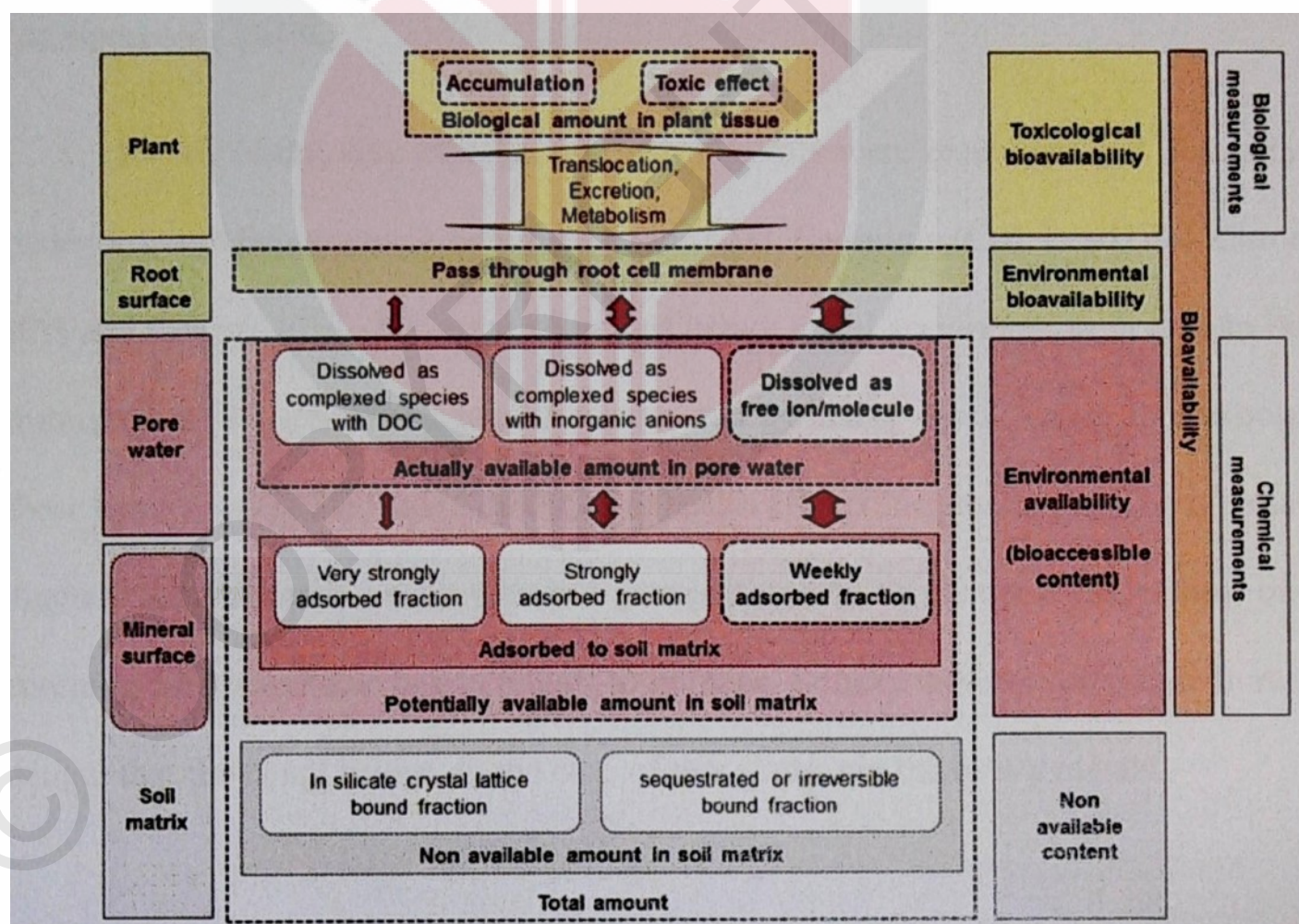


Figure 2.2 Three-step concept of heavy metal bioavailability in soils for plants

Source : Kim et al. (2015)

2.3 Heavy Metal and its Health Effects

There are abundance of heavy metals that are naturally occurring and essential in the nature. Heavy metal is a universal terminology that is used to express metallic elements that encompass average density greater than 5 g/cm³ and have a high atomic weight (Yusof, 2014). The heavy metal works in a way where the toxicity of it will affect the living organism body system due to chemical reactivity of ions interact with cellular structural proteins, membrane system and enzymes. Some organs accumulate the heavy metal in it, as the example kidney, and it is called as target organ. Different heavy metals have different toxicity and different organs that they accumulates based on their toxicity (Mahurpawar, 2015).

In this study, five elements of heavy metals were examined and determined in paddy soil and rice grains, which are Arsenic (As), Cadmium (Cd), Lead (Pb), Chromium (Cr) and Copper (Cu). The main source of heavy metal accumulation in human body is through diet or ingestion of food contaminated by heavy metal. Long time exposure to these heavy metals can lead to detrimental health effects such as cancer. The International Agency for Research on Cancer has grouped agents or elements depending on their carcinogenicity and also health effects to humans. It make it easier for people to refer the danger that the agent brings, in the case of this study, are the heavy metals.

Table 2.3.1 : Agents Classified By The IARC Monographs, Volumes 1–120

Group	Carcinogenicity	Agents
Group 1	Carcinogenic to humans	120
Group 2A	Probably carcinogenic to humans	81
Group 2B	Possibly carcinogenic to humans	299
Group 3	Not classifiable as carcinogenic to humans	502
Group 4	Probably not carcinogenic to humans	1

Table 2.3.2 : List of Classifications, Volumes 1–120

Heavy Metal	CAS No.	Group	Carcinogenicity
Arsenic, As	7440-38-2	1	Carcinogenic to humans
Cadmium, Cd	7440-43-9	1	Carcinogenic to humans
Lead, Pb	7439-92-1	2B	Possibly carcinogenic to humans
Chromium, Cr	7440-47-3	3	Not classifiable as carcinogenic to humans
Copper, Cu	1038-28-6	3	Not classifiable as carcinogenic to humans

Table 2.3.3 : List of Classifications by cancer sites with sufficient or limited evidence in humans, Volumes 1 to 120

Cancer Site	As	Cd	Pb	Cr	Cu
Lip, oral cavity, and pharynx	X				X
Digestive organs			X		
Respiratory organs		X		X	
Bone, skin, soft tissue	X				
Breast and female genital organs					
Male genital organs	X	X			
Urinary tract	X	X			
Endocrine glands					

2.4 Heavy Metal Uptake Mechanism

Heavy metals are a compound that can cycle and moves through water, biosphere and soils, it is the same as the phosphorus cycle in the ecosystem. Majority of the heavy metals does not exist in the state of gas so it exist in the soil as ionic species that can be taken up and accumulates by plants and passed to consumers that eat the plant in the food chain (Irfan et al., 2013). According to Tangahu et al. (2011), water and evaporations from plant leaves act as pump to absorb the nutrients and other substance in the soil through the root to other parts of the plants, it is called as evapotranspiration, and is responsible for moving the heavy metal into a crop.

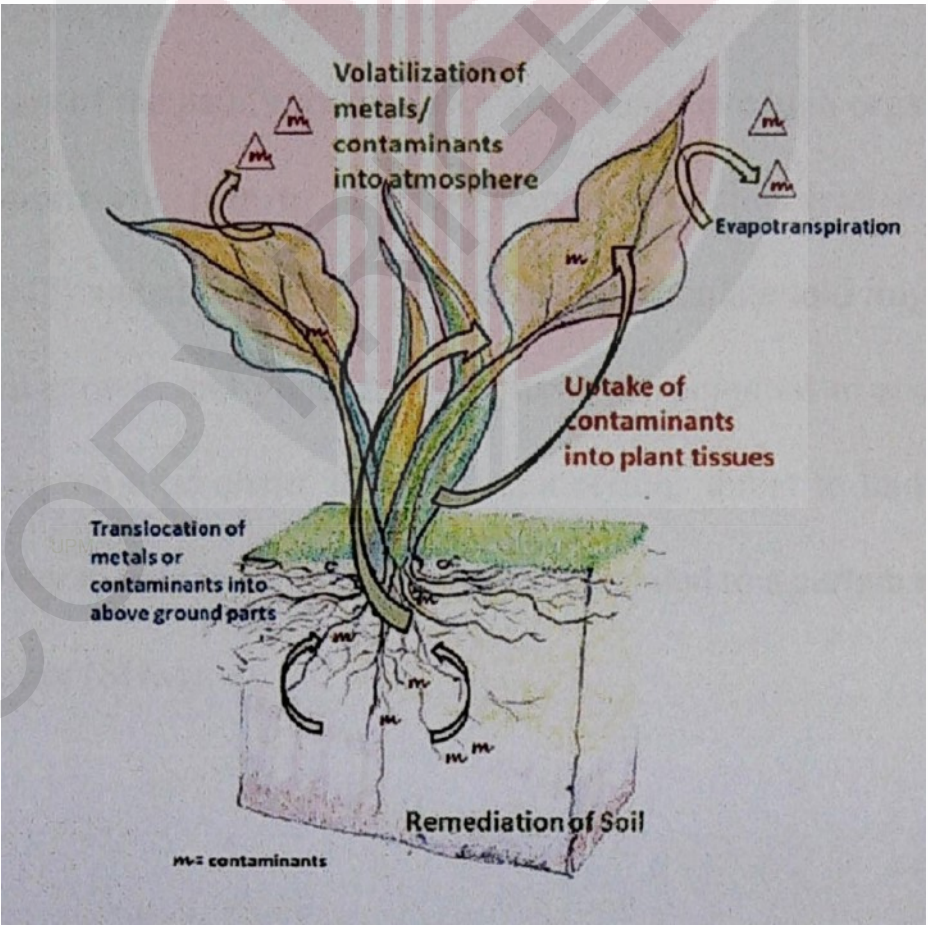


Figure 2.4 : Example of plant uptake mechanism via evapotranspiration

Source : Datta et al. (2013)

2.5 Heavy Metal Accumulation

Sources for heavy metal accumulation in paddy soils and rice grains are through the emissions from the industrial areas, mine tailings, disposal of high metal wastes, leaded gasoline and paints, land application of fertilizers, animal manures, sewage sludge, pesticides, wastewater irrigation, coal combustion residues, spillage of petrochemicals, and atmospheric deposition (Wuana and Okieimen, 2011).

The nature of the paddy soil of which acts as sinks for heavy metals and many other pollutants that persist for long time periods. Paddy soils were kept submerged for long period of times and it acts as which may cause reduction and subsequent mobilization of various metals (Tariq and Rashid, 2013).

The properties of the paddy soil and rice grain that have high organic content and high cellulose, respectively, help to hold the heavy metal thus lead to accumulation. Zhouping et al. (2015) stated that heavy metals that accumulate and migrate in soil not only influence plant growth and yield, but also have the potential to accumulate in the human body through the food chain, thus posing a serious threat to human health. The accumulation of heavy metals in soil and rice grains is related to a certain extent to the pH and soil organic matter (SOM).

2.6 Method to analyze Heavy Metal Concentration in Paddy Soils and Rice Grains

There are various methods to analyze heavy metal concentration. Different methods for specific forms of heavy metal, such as total and bioavailable heavy metal concentration that were determined in this study for paddy soil and rice grain samples, respectively.

For total heavy metal concentration for the paddy soil samples, some methods available are the *aqua regia* digestion, nitric acid-hydrogen peroxide digestion, nitric acid-hydrochloric acid digestion. For this present study, nitric acid-hydrogen peroxide digestion method were used. It is a wet digestion method that used heat that provide complete digestion of the heavy metals within 24 hours.

Whereas, to get the bioavailable heavy metal concentration for the rice grain samples via human rice consumption through oral ingestion route, *in-vitro* method was chosen to imitate the human digestion process. Wragg and Cave (2003), had even critically review all the *in-vitro* methods available, such as Potentially Bioavailable Sequential Extraction (PBASE), Gastrointestinal Analogues, The Physiologically Based Extraction Test (PBET), Simplified Bioaccessibility Extraction Test (SBET), *In-Vitro* Gastrointestinal Method (IVG) and more. Though, a standardised static *in vitro* digestion method suitable for food from Minekus et al. (2014) were used for this present study.

2.7 Health Risk Calculation

The heavy metal parameters for health risk calculation in this study were Copper (Cu), Chromium (Cr), Cadmium (Cd), Lead (Pb) and Arsenic (As). Based on the classification of carcinogenicity by the International Agency for Research on Cancer (IARC), Cu and Cr are classified as non-carcinogenic risk. While, Cd, Pb and As were carcinogenic. Though, in the health risk calculation, Cd was considered as non-carcinogenic risk along with Cu and Cr, due to the absence of Cancer Slope Factor (CSF) value through the ingestion route.

There are three routes of exposure in general, which are, inhalation, ingestion and dermal contact. This study only focused on the ingestion route, since, the rice grain were eaten by the humans. The Ingestion Rate, Exposure Frequency, Exposure Duration, Body Weight and Average Time of exposure, were all included in the calculation, just as the formula of health risk assessment via ingestion. The health risk were determined potential to exist when Hazard Quotient (HQ) for non-carcinogenic risks were (>1) and when Lifetime Cancer Risk (LCR) for carcinogenic risks were ($>1 \times 10^{-4}$).

2.8 Previous Studies of Paddy Soil and Rice Grain in Malaysia

Previous studies on paddy soil done by Habibah et al (2011); Khairiah et al. (2012); Haris et al. (2012); Norbaya et al. (2014); Amjed et al. (2015), had all assessed on the soil quality through various methods. Though, assessing the soil quality only, could not determine the rice grain quality, and vice versa.

Whereas, previous studies done on both paddy soil and paddy plant done by Roslaili et al. (2015); Khan et al. (2016), had focused on the uptake and the bioavailability of heavy metal. However, the research had only assessed the quality of the paddy soil and paddy plant. Nonetheless, health risk to human should be taken into consideration too, since rice grain is our staple food in Malaysia.

A study done by Juen et al. (2014) that had collected from soil, plant and grain samples to assess the heavy metal translocation efficiency, but it has not further assess on the potential health risks to human.

Although, recent study done by Praveena & Omar (2015; 2017) had assessed on the heavy metal concentration of rice grain, in both total and available form. Thus, associated it with potential health risks. What differ in this present study is that, the total and bioavailable heavy metal concentration, were determined for paddy soil and rice grain, respectively. Further evaluation on paddy soil quality and human health risks were also conducted accordingly.

Table 2.8.1 : Summary of previous studies compared to present study

Authors & Date of Publication	Paddy			Rice		Research Focus / Keywords		Human	
	soil	plant			grain			Total	Bioavailable health risks
Habibah et al, 2011	X					Speciation of heavy metals		X	
Khairiah et al.,2012	X					Heavy metal content		X	
Haris et el., 2012	X					Geoaccumulation index and enrichment factor		X	
Juen et al., 2014	X	X			X	Bioconcentration and Translocation Efficiency of Metals in Paddy		X	
Norbaya et al., 2014	X					Analysis And Pollution Assessment		X	

Authors & Date of Publication	Research Focus			Human		
	Paddy soil	Paddy plant	Rice grain	Total	Bioavailable	Health risks
Amjed et al., 2015	X			X		
Roslaili et al.,2015	X	X		X		
Khan et al, 2016	X	X			X	
Praveena & Omar, 2017			X	X	X	X
Present study	X		X	X	X	X

CHAPTER 3

METHODOLOGY

3.1 Research Design

This study is a cross-sectional design study. It is purely descriptive to determine the heavy metal concentration in the paddy soil and rice grain. Data were collected at a single point in time for the analysis and result were calculated to associate with the potential health risks to environment and human, respectively.

3.2 Study Location

The chosen paddy fields are responsible for the rice production in Malaysia. Paddy field in Sabak Bernam, Tanjung Karang and Sekinchan (Selangor), Besut (Terengganu) and Yan (Kedah).

In Selangor, these three paddy fields of Sabak Bernam, Tanjung Karang and Sekinchan are located close together, they are about 102km north of Kuala Lumpur. It was stated by the Ministry of Agriculture & Agro-Based Industry Malaysia (2016), that the overall area of planted paddy field in Selangor was 19,856 hectares with 9057 paddy farmers. Even though, paddy fields in Selangor were not big of an area rather than of in other states of Malaysia, but, it was the highest paddy producers with 6.15 tonnes metric per season.

While in Terengganu, a project called IADA KETARA under the MOA had covered an area of 208,212 hectares which consist of 122,831 hectares paddy fields in Besut and 85,381.70 hectares in Setiu. It was set in the 3rd National Agriculture Policy that targeted for the project to reach 6.5 million tonners per hectares, which is needed to reach 90% self-sufficiency level (MOA, 2017).

Whereas, in Kedah, an agency under MOA, which is , Muda Agriculture Development Authority (MADA) has managed and operated the paddy fields in Yan. The place had 19 unit farmers' shares and had an area of 3,590 hectares. The paddy estate project contributed revenues of RM 1,898,855 per annum in the economic sense (MADA, 2012).

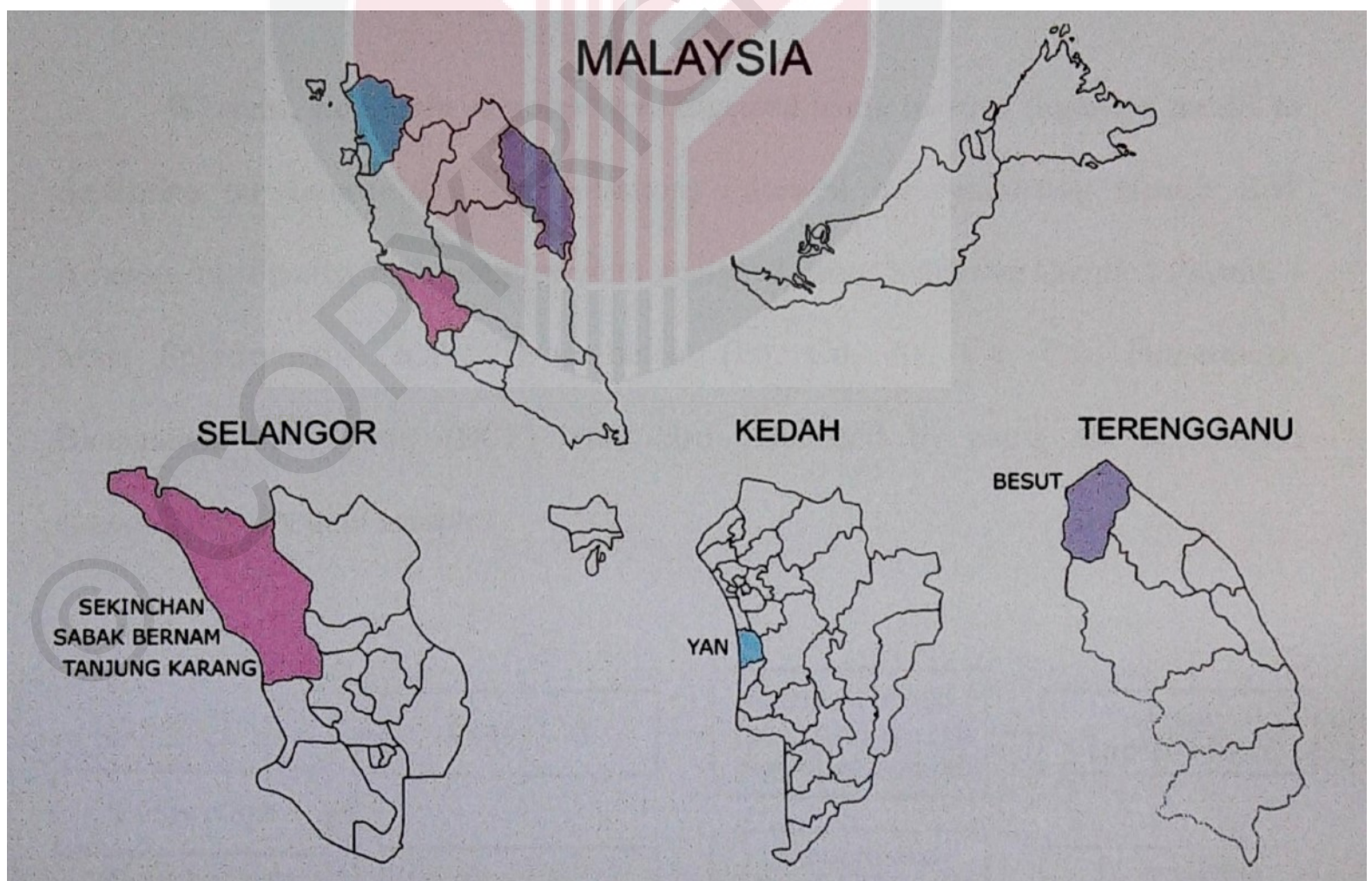


Figure 3.2.1: Study locations of Sabak Bernam, Tanjung Karang, Sekinchan, Yan and Besut paddy fields in Malaysia

3.3 Sample Collection

Paddy soil and rice grain samples were collected from the paddy fields in Sabak Bernam, Tanjung Karang and Sekinchan (Selangor), Besut (Terengganu) and Yan (Kedah), respectively. Three plots of paddy soil samples and three plots of rice grain samples were taken for paddy fields in

3.4 Study Methodology

For the paddy soil samples, nitric acid and hydrogen peroxide digestion method were used to determine the total concentration, followed by calculating the Geoaccumulation Index (Igeo).

Whereas, rice grain samples were digested using *in-vitro* digestion model to determine the bioavailable concentration, followed by conducting Health Risk Assessment. Finally, both samples were analyzed using Inductive Coupled Plasma – Mass Spectrometry (ICP-MS) to detect (Pb, Cd, As, Cu, Cr). Futhermore, Bioconcentration Factor (BCF) were also calculated by using the determined concentration by both samples.

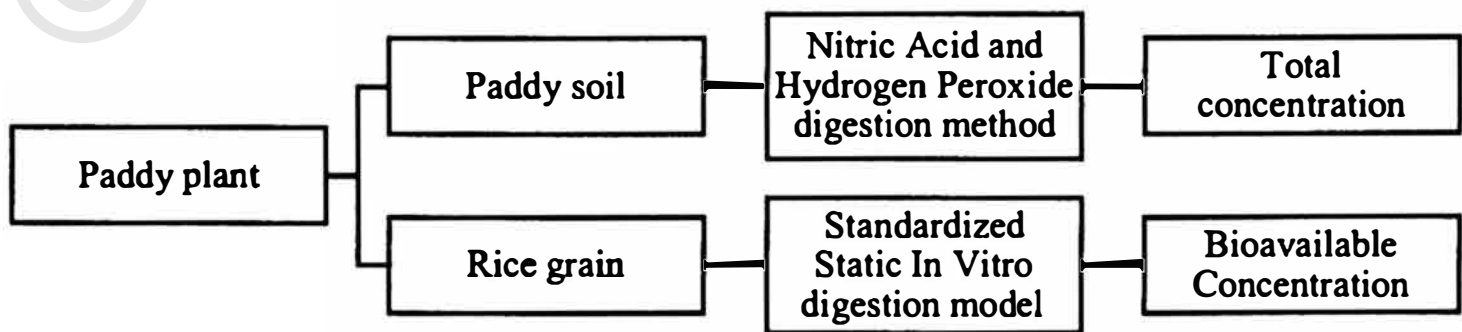


Figure 3.4.1 : Process flow of this research methodology

3.4.1 Preparation Of Samples

The samples of paddy soil and rice grains were air-dried, homogenized by mortar and pestle, and sieved through a 2mm mesh screen before storage in polyethylene bags.



Figure 3.4.1.1 : Paddy soil samples before air-dried and homogenized



Figure 3.4.1.2 : Rice grain samples before air-dried and homogenized

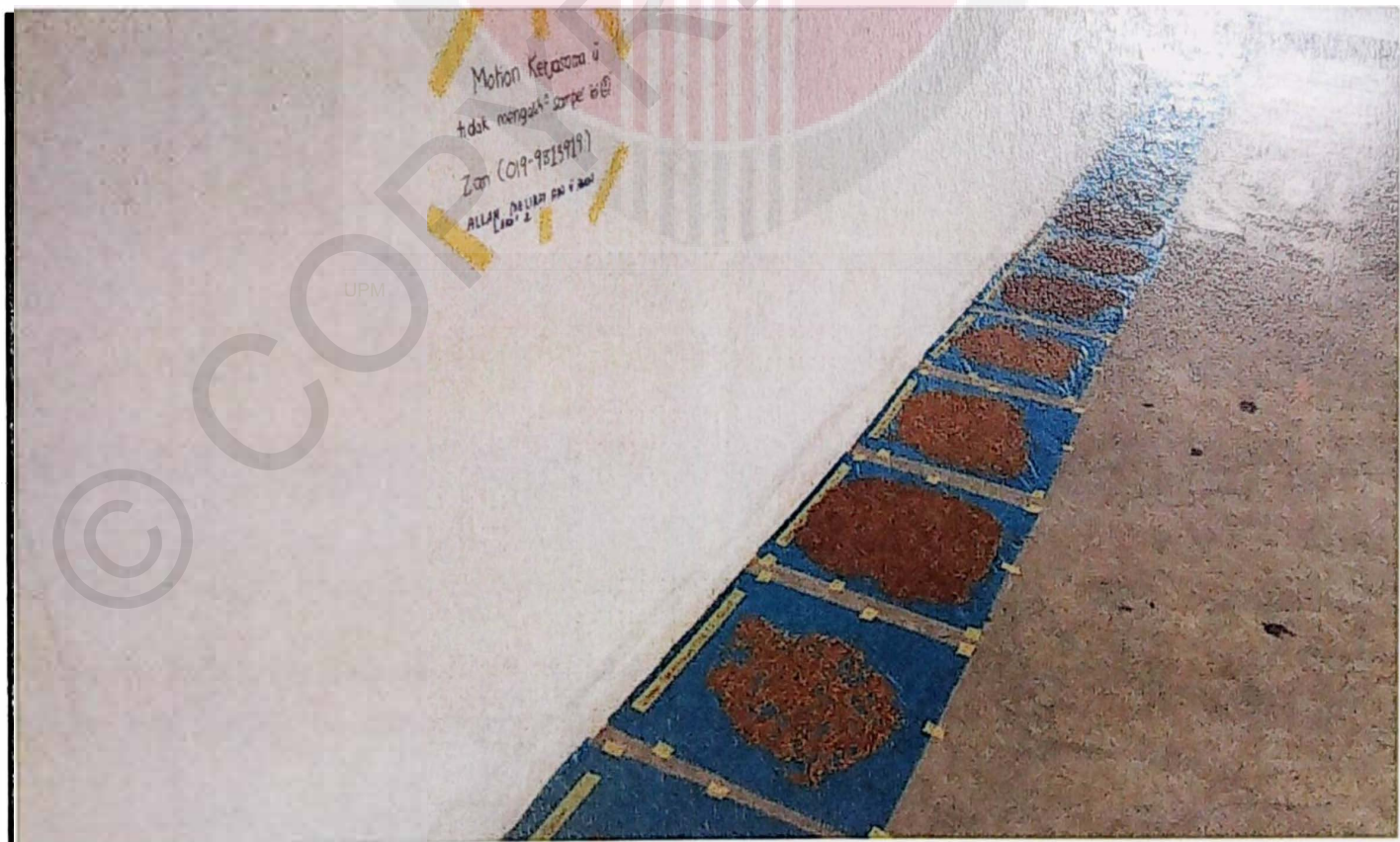


Figure 3.4.1.3 : Samples were air-dried for 3-4 days



Figure 3.4.1.4 : Samples were homogenized by homogenized by mortar and pestle, and sieved through a 2mm mesh screen



Figure 3.4.1.5 : Paddy soil samples after air-dried and homogenized

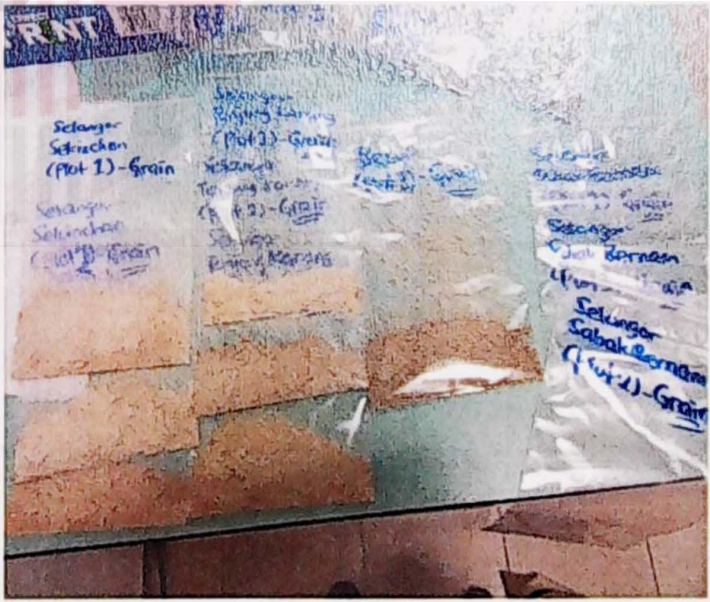


Figure 3.4.1.6 : Rice grain samples after air-dried and homogenized

3.4.2 Soil Samples Digestion

Paddy soil samples (n=13) were digested using nitric acid and hydrogen peroxide digestion method. A total of 1g paddy soil sample was added with 10 mL nitric acid (HNO₃, 0.05M) and digested for 2 hours at a constant temperature of 50 °C. After heating, samples were brought out from the oven and left to cool to room temperature. Next, 5 mL of 30% hydrogen peroxide (H₂O₂) was added in, which acted to digest the inorganic compounds, and were left at room temperature overnight. Lastly, samples were filtered through 0.45µm Whattman filter paper and kept in chiller at 4°C before further analysis.

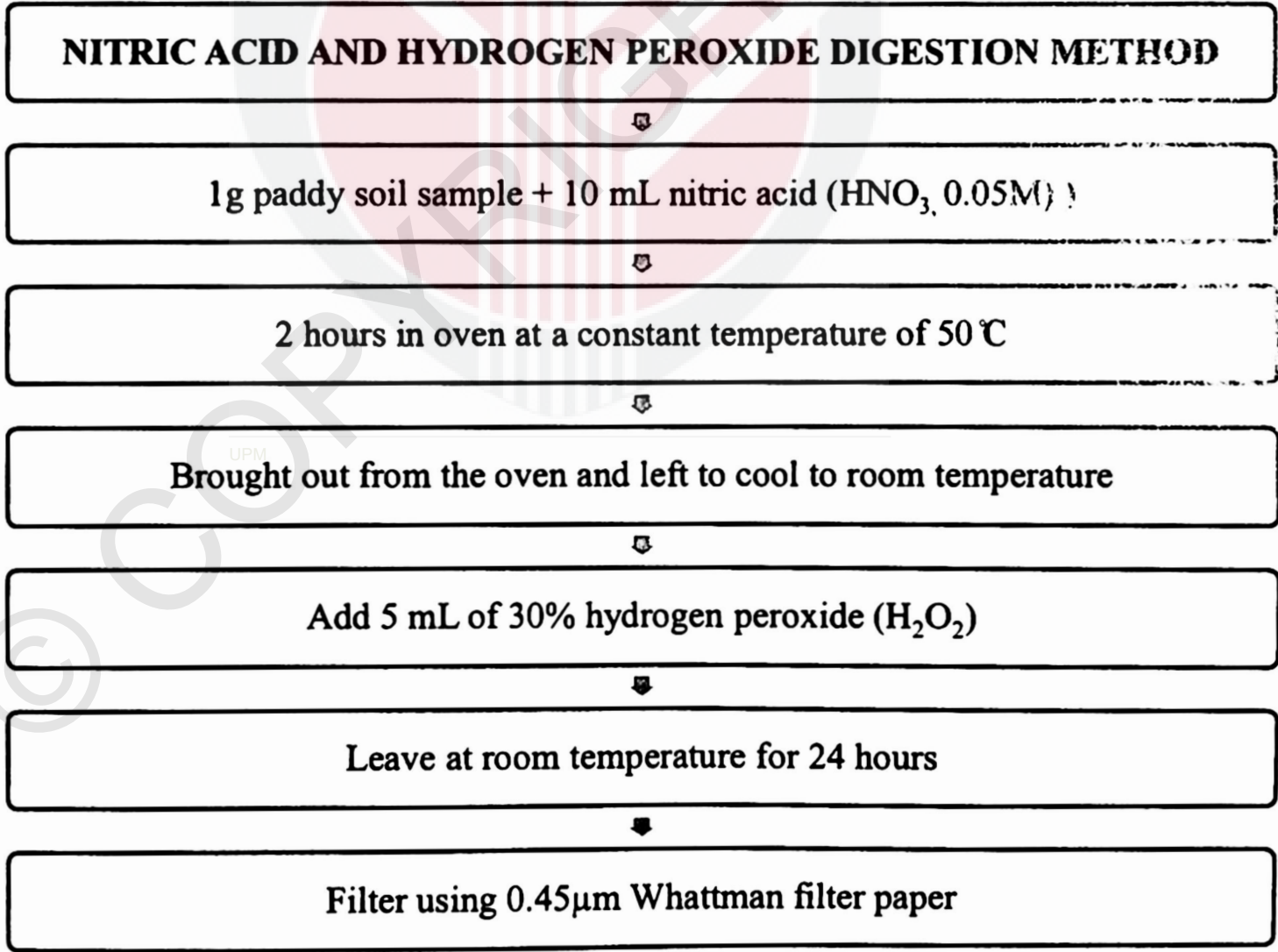


Figure 3.4.2.1 : Process Flow of Nitric Acid and Hydrogen Peroxide Digestion Method for Soil Samples

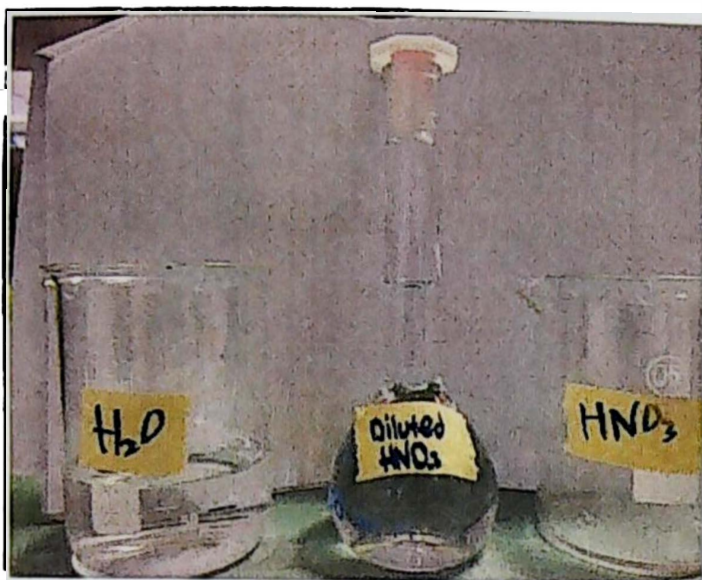


Figure 3.4.2.2 : Preparing the nitric acid before digestion experiment



Figure 3.4.2.3 : Soil samples were weighed carefully to 1g

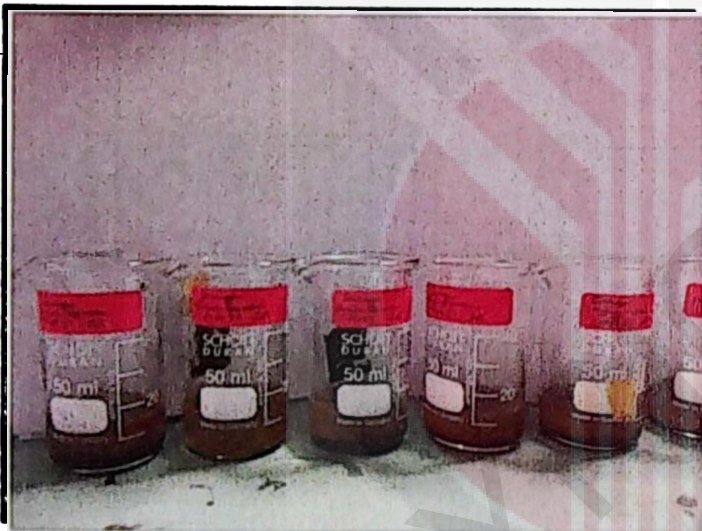


Figure 3.4.2.4 : Soil samples after adding the nitric acid



Figure 3.4.2.5 : Soil samples heated in the oven for 2 hours at 50 °C

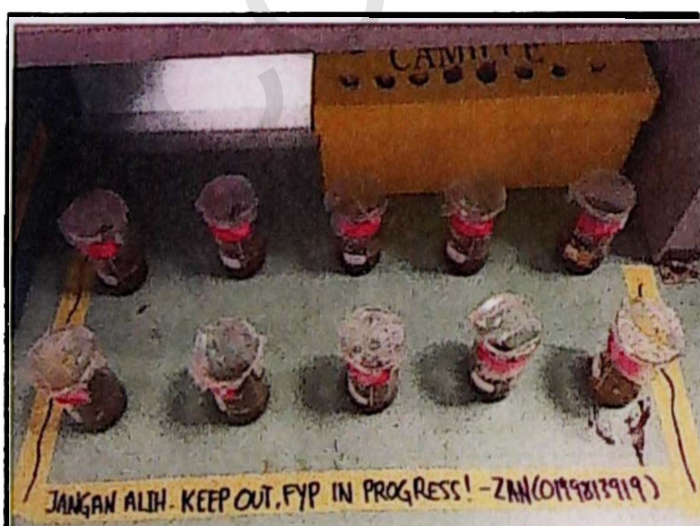


Figure 3.4.2.6 : Soil samples were added hydrogen peroxide and left to digest overnight

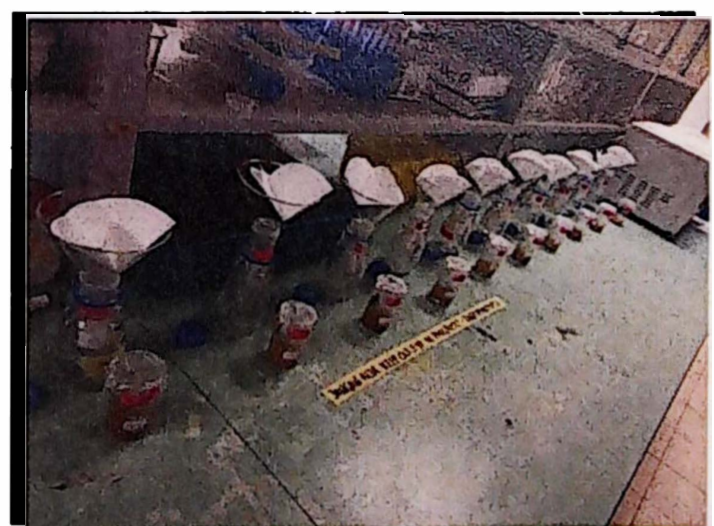


Figure 3.4.2.7 : Soil samples were filtered using 0.45µm Whattman filter paper

3.4.3 Rice Grain Samples Digestion

Rice grain samples (n=13) were digested using a standardized static *in vitro* digestion method by Minekus et al. (2014). The model were made to attain the bioavailable concentration of the heavy metal in the rice grain samples, which are the amount of a compound that is released from the matrix and is considered to be available for absorption through the gut wall. (Minekus et al.,2014).

Before the experiment, all the simulated digestion fluids were prepared, which are the Simulated Salivary Fluid (SSF), Simulated Gastric Fluid (SGF) and Simulated Intestinal Fluid (SIF), which are made up of electrolyte stock solutions, enzymes, calcium chloride (CaCl_2), and water. The experiment started off with the oral phase, 1g of rice grain samples were added with 3.5mL SSF, 2.5 μL of CaCl_2 and 975 μL of deionized water. The solution were stirred for 1 minute and stand for 1 minute. Followed by the gastric phase, 3mL of previous digested solution from the oral phase was added with 7.5mL SGF, 5 μL of CaCl_2 and 0.2mL hydrochloric acid (HCl). Then, pH were adjusted to reach pH 3.0. Next, 0.695 μL of deionized water were added. The digested samples were then placed into a water-bath shaker such that in the human temperature of 37°C for 2 hours at 100 rpm. After that, digested samples were let to stand for 20 to 30 minutes.

Finally, in intestinal phase, 10mL of previous digested solution was added with 11mL SIF, 2.5mL of fresh bile, 40 μL of CaCl_2 , 0.15mL of sodium hydroxide (NaOH). Then, pH were adjusted to reach pH 7.0. Next, 1.31mL of deionized water were added. The digested sample was again placed into a water-bath shaker such that the human

temperature of 37 °C for 2 hours at 100 rpm. After that, digested samples were left to stand till precipitated. Lastly, digested rice samples were filtered through 0.45µm Whattman filter paper and kept in chiller at 4°C before further analysis.

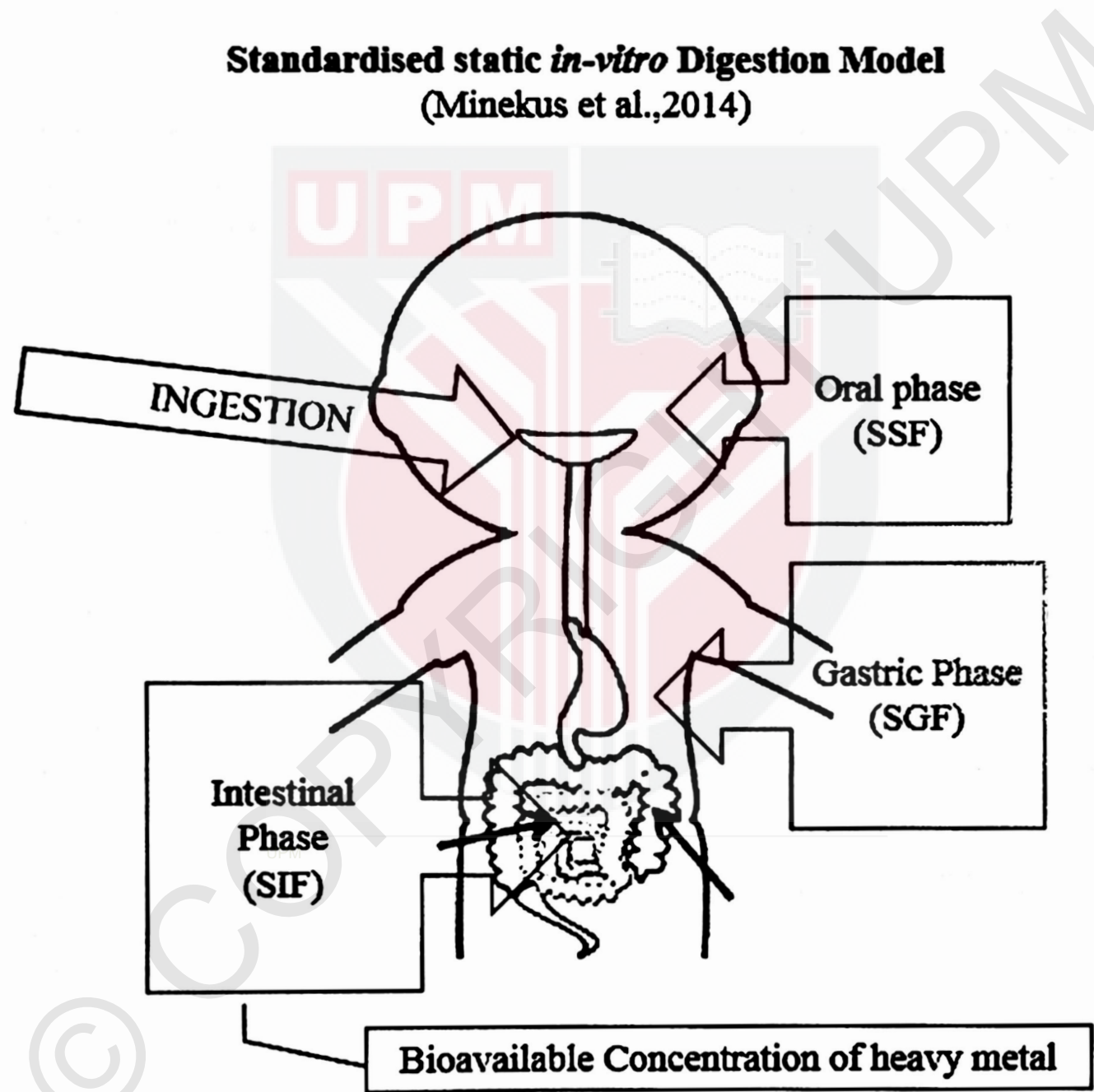


Figure 3.4.3.1 : The human digestion process when ingesting the rice samples

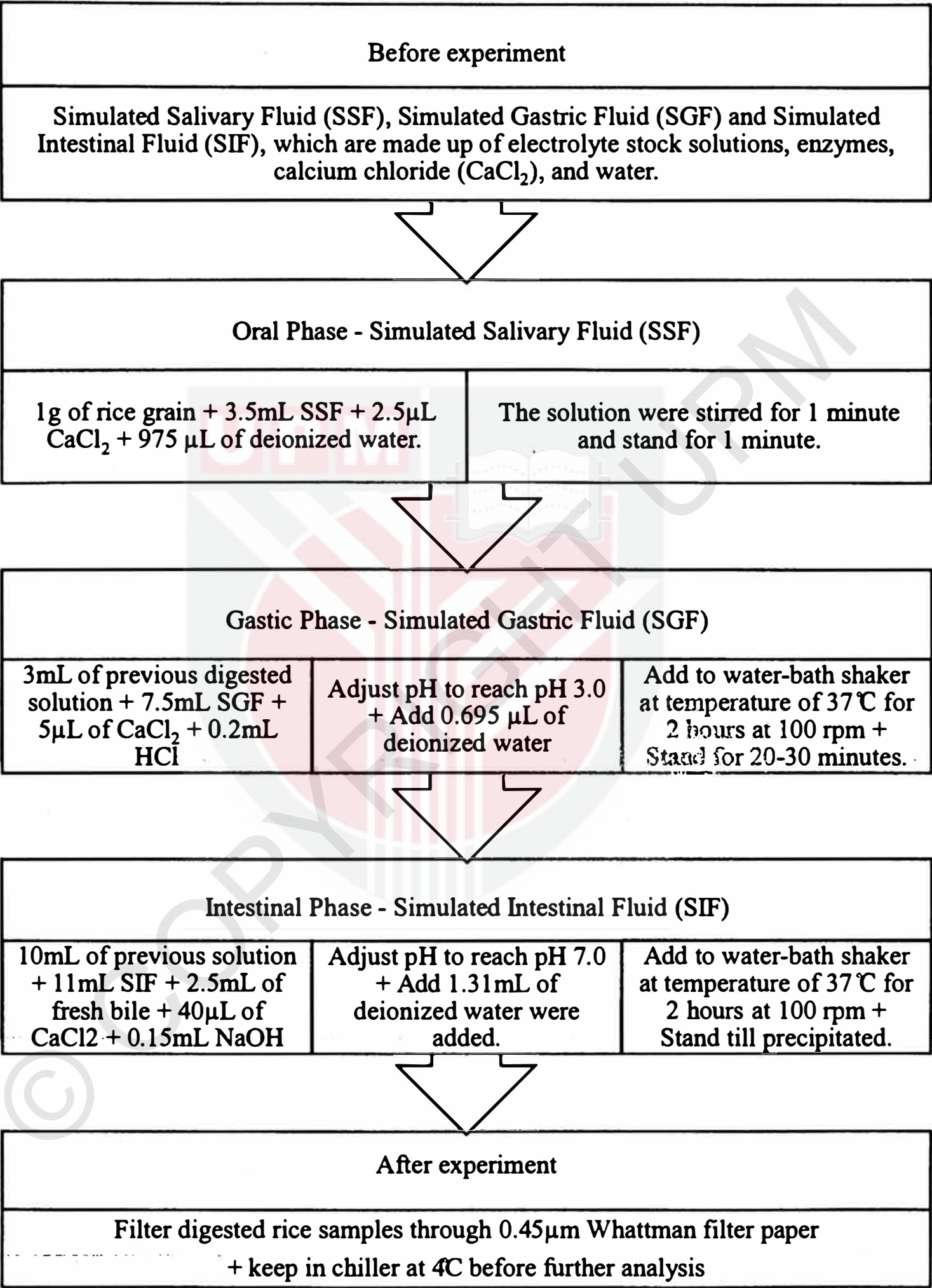


Figure 3.4.3.2 : Process Flow of Standardized Static *in-vitro* digestion model for Rice Grain Samples



Figure 3.4.3.3 : Transferring the previous digested solution



Figure 3.4.3.4 : Rice grain samples in the water-bath shaker



Figure 3.4.3.5 : pH meter used to adjust the pH and the labelled tubes to transfer in every phase

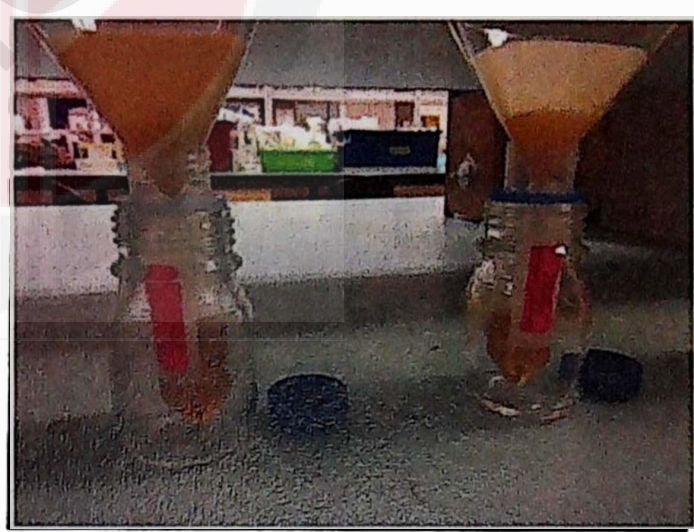


Figure 3.4.3.6 : Rice samples were finally filtered using 0.45µm Whatman filter paper



Figure 3.4.3.7 : Heavy metal calibration solution and standard solution for ICP-MS



Figure 3.4.2.8 : Both paddy soil and rice grain samples were brought to analyze by ICP-MS

3.4.4 Geoaccumulation Index (Igeo)

The geoaccumulation index (Igeo) was determined by using the following equation;

$$I_{geo} = [\log_2 C_n / 1.5 \times B_n]$$

Where,,

Cn = Concentration (mean) of heavy metals as calculated

Bn = Geochemical background value of shale or soil in the area (reference)

1.5 = Correction factor due to lithological variations.

In this study, the background value (Bn) were obtained from Rudnick & Gao (2004) in Geochemical Earth Reference Model (GERM) Reservoir Database (2017).

Table 3.4.4.1 : Background value for each heavy metal

Heavy metal	Cu	Cd	Cr	Pb	As
Background Value (µg/g)	28	0.102	92	17	6.6

Table 3.4.4.2 : The seven classes of Igeo index

Geoaccumulation index (Igeo)	Class	Pollution Intensity
0	0	Background concentration
0-1	1	Unpolluted
1-2	2	Moderately to unpolluted
2-3	3	Moderately polluted
3-4	4	Moderately to highly polluted
4-5	5	Highly polluted

*Source: Singh et all, (2003)

3.4.5 Bioconcentration Factor (BCF)

Biocencentration factor (BCF) is the ratio of the content of heavy metals in parts of paddy like grain to that in the soil. BCF was determined by using the following equation;

$$BCF = C_{rice} / C_{soil}$$

Where,

C_{rice} = Bioavailable heavy metal concentration in rice grain samples

C_{soil} = Total heavy metal concentration in paddy soil.

When $BCF \leq 1.00$, it indicates that the plant can only absorb but not accumulate metal, and if a $BCF > 1.00$, it means that the plant have the potential to accumulate metal (Liu et al. 2009).

Table 3.4.5.1 : Bioconcentration Factor representation

Bioconcentration factor (BCF)	Represent
≤ 1.00	Only absorb but not accumulate metal
> 1.00	Potential to accumulate metal

*Source : Liu et al. (2009)

3.4.6 Health Risk Assessment (HRA)

A quantitative health risk assessment (HRA) functioned to generate a numerical measure of the risk or safety of the heavy metal exposure to human via ingestion. A chronic health risk in a non-cancer endpoint is known as non-carcinogenic risk and expressed using hazard quotient (HQ). Whereas, chronic health risks that leads to cancer endpoint is known as carcinogenic risk and expressed using Lifetime Cancer Risk (LCR). The formulas were based on United States Environmental Protection Agency (US EPA), (2014);

Non-carcinogenic risks,

$$ADD = C_{\text{rice}} \times IR \times ED \times EF / BW \times AT$$

$$HQ = ADD / RfD$$

Carcinogenic risks,

$$LADD = C_{\text{rice}} \times IR \times ED \times EF / BW \times AT$$

$$LCR = LADD \times CSF$$

Where,

Table 3.4.6.1 : Variable used in health risk assessment

	Name	Unit	Adult	Children
C _{rice}	Bioavailable concentration of rice	mg/kg	Value as obtained	
IR	Ingestion Rate	g/day	600	198.4
EF	Exposure Frequency	days/year	365	365
ED	Exposure Duration	years	35	6
BW	Body Weight	kg	62.65	19.5
AT	Average Time	days	LADD = 25550 days ADD = ED X 365	

In determining the health risks in both adult and children, the values for IR, EF, and BW, for adults were 600g/day, 365 days/year and 62.65kg, for children 198.4g/day, 365 days/year, 19.5kg, respectively, LADD = 25550days and ADD = ED X 365, were all obtained from the studies done by Department of Statistics Malaysia (2014), Norimah, Safiah, and Jamal (2008) and Zhuang, McBride, and Xia (2009). ED values were substituted as suggested by the U.S. Environmental Protection Agency (US EPA), 35 years average for adult and 6 years for children.

While, Reference Dose (RfD) and Cancer Slope Factor (CSF) values were obtained from the Integrated Risk Information System (IRIS) Chemical Assessment Summary by U.S. Environmental Protection Agency (US EPA), (2018).

Table 3.4.6.2 : RfD and CSF value for each heavy metal

Heavy metal	Reference Dose for Oral Exposure, RfD (mg/kg-day)	Cancer Slope Factor for Oral Exposure, CSF (mg/kg)/day	References
Cu	0.001	-	
Cd	0.001	-	Integrated Risk
Cr	0.003	-	Information System
Pb	-	0.0085	(IRIS), US EPA
As	-	1.5	

Since there are no available CSF data for Cd through ingestion pathway yet, it was considered as non-carcinogenic health risks along with Cu and Cr for the calculation. Only Pb and As acted as the carcinogenic health risks.

3.5 Quality Control and Quality Assurance

The laboratory apparatus were carefully soaked overnight with nitric acid, rinsed with deionized water and dried completely in the oven before usage. All beakers and tubes were properly labelled upon transferring. The simulated digestion fluids, like SSF, SGF and SIF, were prepared right before the *in vitro* digestion experiment was conducted to avoid the reduction of enzymatic activities. Inductively coupled plasma mass spectrometry (ICP-MS) was calibrated with Multi-Element Calibration Standard of 10 mg/L before the machine was run for detection. Heavy metal standards solution were put in between every three samples during analysis. The use of internal standard in ICP-MS is to correct for instrument instability and/or signal drift and non-spectral interferences (signal suppression or signal enhancement caused by the matrix). Moreover, internal standardization can bring an improvement in both precision and accuracy (Vanhaecke, Vanhoe & Damst, 1992).

3.6 Data Analysis

The mean values for the total and bioavailable heavy metal concentration were obtained directly from the ICP-MS. All descriptive analysis (Geoaccumulation index, Bioconcentration factor, and Health Risk Assessment) were performed using Microsoft Office Professional Plus Excel 2013.

CHAPTER 4

RESULT

4.1 The Total and Bioavailable Heavy Metal Concentration

Table 4.1.1 and Table 4.1.2 showed the mean of total and bioavailable heavy metal (As, Cd, Pb, Cr, Cu) in paddy soil and rice grain, respectively. Paddy soil showed decreasing trend from ; Pb (0.0082 mg/kg) > Cr (0.0054 mg/kg) > Cu (0.0038 mg/kg) > As (0.0026 mg/kg) > Cd (0.0001 mg/kg)

Whereas the bioavailable heavy metal concentration in the rice grain was much higher than the paddy soil, even in decreasing trend; Cr (0.2701 mg/kg) > Cu (0.0896 mg/kg) > Pb (0.0458 mg/kg) > As (0.0139mg/kg) > Cd (0.0053 mg/kg). Pb and Cr were both found to be the highest concentration in paddy soil and rice grain, respectively. While, Cd was found to be the lowest toxic element in both.

Table 4.1.1 : Total Heavy Metal Concentration in Paddy Soil samples

Sample ID	Sample Names (n=13)	Mean concentration (mg/kg)				
		Cu	Cd	Cr	Pb	As
1	Sabak Bernam (Plot 1) Soil	0.0029	0.0001	0.0074	0.0071	0.0023
2	Sabak Bernam (Plot 2) Soil	0.0037	0.0001	0.0063	0.0081	0.0027
3	Sabak Bernam (Plot 3) Soil	0.0033	0.0001	0.0060	0.0062	0.0023
4	Tanjung Karang (Plot 1) Soil	0.0037	0.0001	0.0052	0.0065	0.0025
5	Tanjung Karang (Plot 2) Soil	0.0036	0.0001	0.0070	0.0096	0.0039
6	Tanjung Karang (Plot 3) Soil	0.0036	0.0001	0.0064	0.0066	0.0025
7	Sekinchan (Plot 1) Soil	0.0044	0.0002	0.0078	0.0111	0.0028
8	Sekinchan (Plot 2) Soil	0.0033	0.0001	0.0062	0.0091	0.0028
9	Sekinchan (Plot 3) Soil	0.0031	0.0001	0.0055	0.0072	0.0024
10	Yan (Plot 1) Soil	0.0047	0.0001	0.0073	0.0057	0.0023
11	Yan (Plot 2) Soil	0.0025	0.0001	0.0050	0.0052	0.0024
12	Yan (Plot 3) Soil	0.0052	0.0001	0.0061	0.0068	0.0029
13	Besut (Plot 1) Soil	0.0053	0.0002	0.0045	0.0167	0.0014

Table 4.1.2 : Bioavailable Heavy Metal Concentration in Rice Grain samples

Sample ID	Sample Names (n=13)	Mean concentration (mg/kg)				
		Cu	Cd	Cr	Pb	As
14	Sabak Bernam (Plot 1) Grain	0.1715	0.0004	0.1072	0.0297	0.0169
15	Sabak Bernam (Plot 2) Grain	0.0739	0.0004	0.2216	0.0331	0.0181
16	Sabak Bernam (Plot 3) Grain	0.0943	0.0028	0.2759	0.2645	0.0132
17	Tanjung Karang (Plot 1) Grain	0.1191	0.0011	0.3857	0.0389	0.0183
18	Tanjung Karang (Plot 2) Grain	0.0741	0.0503	0.2734	0.0185	0.0121
19	Tanjung Karang (Plot 3) Grain	0.1048	0.0002	0.3961	0.0257	0.0172
20	Sekinchan (Plot 1) Grain	0.0774	0.0004	0.2763	0.0154	0.0200
21	Sekinchan (Plot 2) Grain	0.0586	-0.0003	0.2249	0.0085	0.0092
22	Sekinchan (Plot 3) Grain	0.0649	-0.0003	0.2852	0.0975	0.0100
23	Yan (Plot 1) Grain	0.0691	0.0009	0.2683	0.0151	0.0120
24	Yan (Plot 2) Grain	0.0997	0.0045	0.2748	0.0116	0.0100
25	Yan (Plot 3) Grain	0.0501	-0.0002	0.2070	0.0120	0.0090
26	Besut (Plot 1) Grain	0.1076	0.0081	0.3150	0.0243	0.0147

4.2 Geoaccumulation Index (Igeo)

Geoaccumulation index (Igeo) were in background concentration where (Igeo<0) for each heavy metal elements. Which falls in the Class 0. The index obtained as in Table 4.2.1 and Figure 4.2.1.

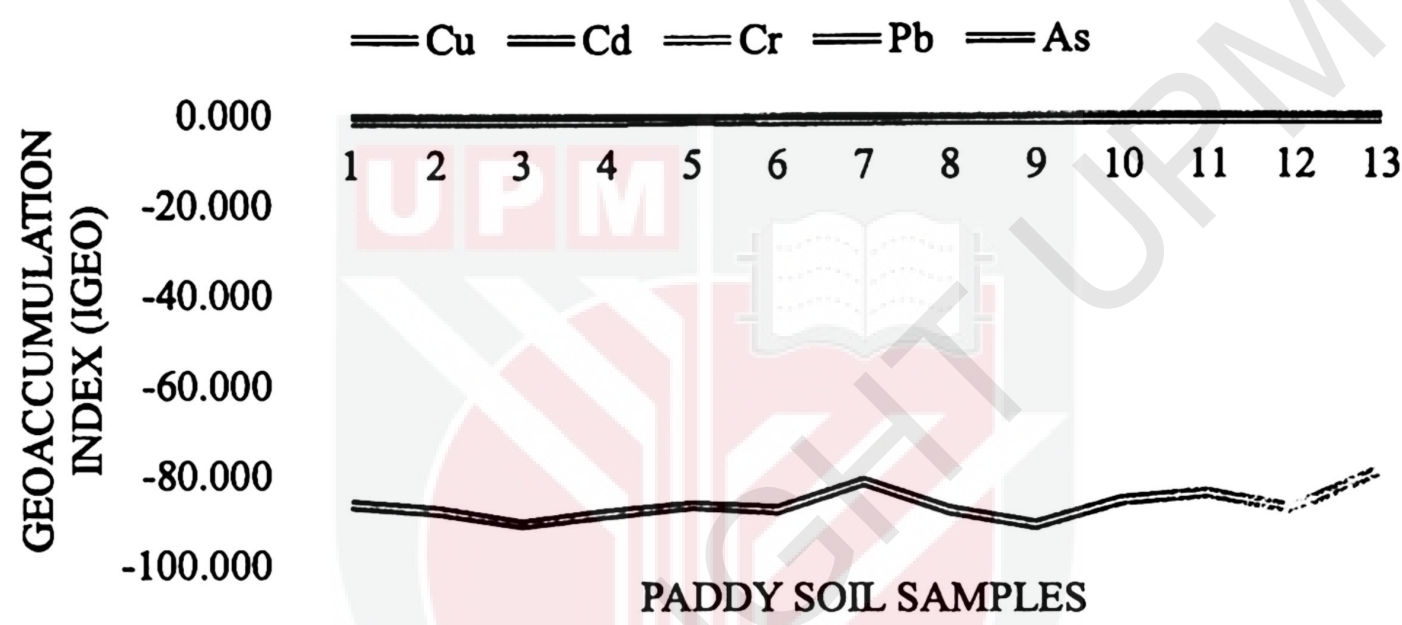


Figure 4.2.1 : Geoaccumulation index (Igeo) values in paddy soil.

Table 4.2.1 : Geoaccumulation Index (Igeo) of heavy metals in paddy soil

Geoaccumulation Index	Class	Pollution Intensity	Heavy Metals
0	0	Background concentration	Cu, Cd, Cr, Pb, As
0-1	1	Unpolluted	
1-2	2	Moderately to unpolluted	
2-3	3	Moderately polluted	
3-4	4	Moderately to highly polluted	
4-5	5	Highly polluted	

4.3 Bioconcentration Factor (BCF)

The bio-concentration factor (BCF) were found to be potential to accumulate ($BCF > 1$) for each heavy metal elements, as shown in Figure 4.3.1.

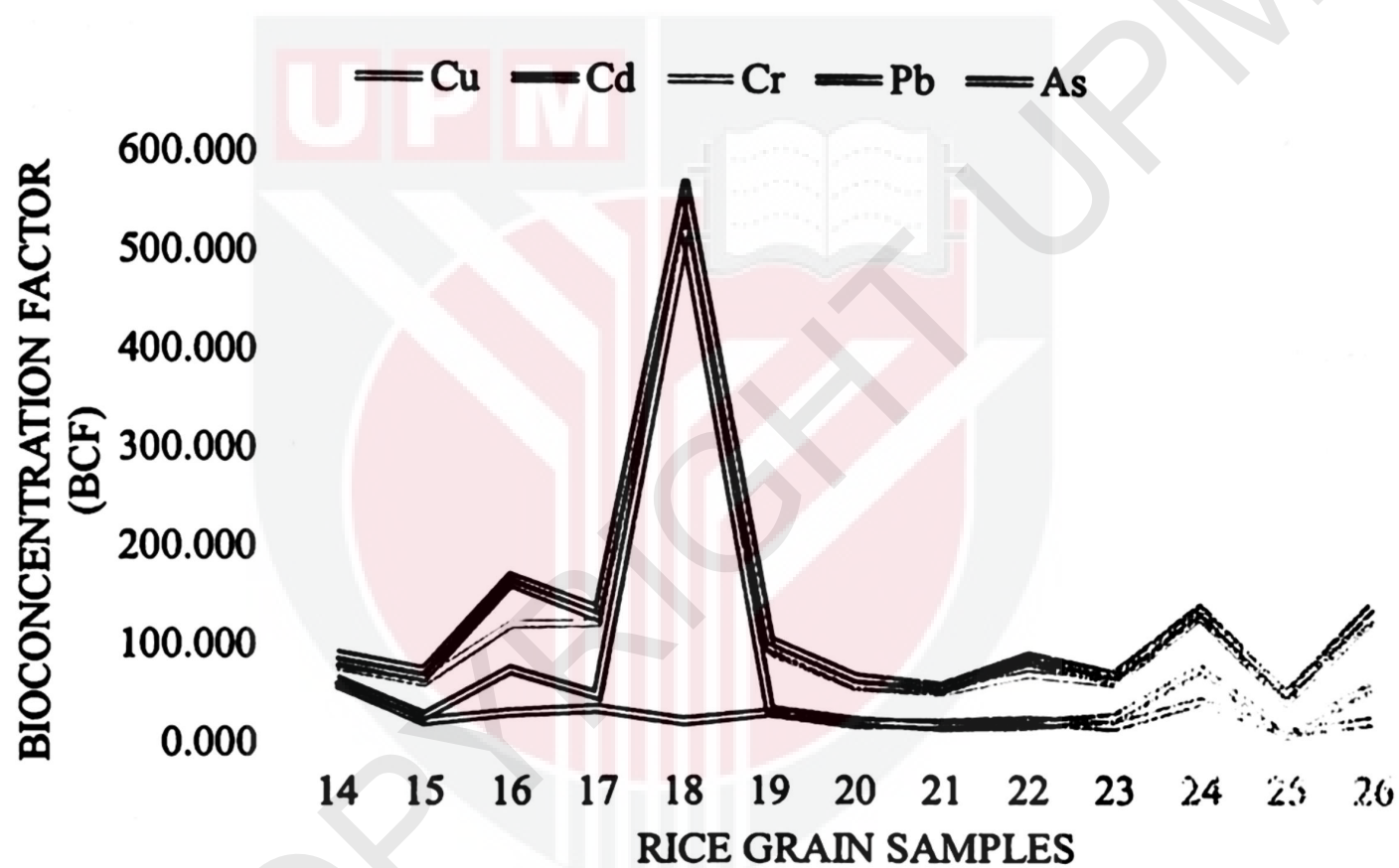


Figure 4.3.1 Bioconcentration factor (BCF) values which involved paddy soil and rice grain samples concentration.

Table 4.3.1 : Bioconcentration Factor of heavy metals in rice grain

Bioconcentration factor (BCF)	Represent	Heavy Metals
≤ 1.00	Only absorb but not accumulate metal	
> 1.00	Potential to accumulate metal	Cu, Cd, Cr, Pb, As

4.4 Health Risk Assessment (HRA)

Health Risk Assessment (HRA) was calculated for the average adults and children, involving rice grain samples.

Results of HRA for adult shows that there were potential health risks of non-carcinogenic risk, Copper (Cu) and Chromium (Cr), where hazard quotient ($HQ > 1$). While, for non-carcinogenic risk, heavy metal Arsenic (As) has potential risks, where lifetime cancer risk ($LCR > 1 \times 10^{-4}$).

Whereas, in children, there were potential non-carcinogenic risk for Copper (Cu) and Chromium (Cr), where hazard quotient ($HQ > 1$). While, for non-carcinogenic risk, the heavy metal elements were at the acceptable level, where, ($LCR > 1 \times 10^{-6}$).

The values calculated were shown as in the given tables.

Table 4.4.1 HQ and LCR values for adult

Sample Names	HQ	HQ	HQ	LCR	LCR
	Cu	Cd	Cr	Pb	As
14 Sabak Bernam (Plot 1) Grain	1.6E+00	3.8E-03	3.4E-01	1.2E-06	1.2E-04
15 Sabak Bernam (Plot 2) Grain	7.1E-01	4.0E-03	7.1E-01	1.3E-06	1.3E-04
16 Sabak Bernam (Plot 3) Grain	9.0E-01	2.7E-02	8.8E-01	1.1E-05	9.4E-05
17 Tanjung Karang (Plot 1) Grain	1.1E+00	1.1E-02	1.2E+00	1.6E-06	1.3E-04
18 Tanjung Karang (Plot 2) Grain	7.1E-01	4.8E-01	8.7E-01	7.5E-07	8.7E-05
19 Tanjung Karang (Plot 3) Grain	1.0E+00	1.5E-03	1.3E+00	1.0E-06	1.2E-04
20 Sekinchan (Plot 1) Grain	7.4E-01	4.0E-03	8.8E-01	6.3E-07	1.4E-04
21 Sekinchan (Plot 2) Grain	5.6E-01	-3.0E-03	7.2E-01	3.5E-07	6.6E-05
22 Sekinchan (Plot 3) Grain	6.2E-01	-2.8E-03	9.1E-01	4.0E-06	7.2E-05
23 Yan (Plot 1) Grain	6.6E-01	8.9E-03	8.6E-01	6.1E-07	8.6E-05
24 Yan (Plot 2) Grain	9.5E-01	4.3E-02	8.8E-01	4.7E-07	7.2E-05
25 Yan (Plot 3) Grain	4.8E-01	-2.0E-03	6.6E-01	4.9E-07	6.4E-05
26 Besut (Plot 1) Grain	1.0E+00	7.7E-02	1.0E+00	9.9E-07	1.1E-04

Legend :

Noncarcinogenic risk; Hazard quotient (HQ) = Cu, Cd and Cr

Carcinogenic risk; Lifetime Cancer Risk (LCR) = Pb and As

Potential Health Risk; $HQ > 1$, $LCR > 1 \times 10^{-4}$

Table 4.4.2 HQ and LCR values for children

Sample Names	HQ	HQ	HQ	LCR	LCR
	Cu	Cd	Cr	Pb	As
14 Sabak Bernam (Plot 1) Grain	1.7E+00	4.0E-03	3.6E-01	2.2E-07	2.2E-05
15 Sabak Bernam (Plot 2) Grain	7.5E-01	4.2E-03	7.5E-01	2.5E-07	2.4E-05
16 Sabak Bernam (Plot 3) Grain	9.6E-01	2.9E-02	9.4E-01	2.0E-06	1.7E-05
17 Tanjung Karang (Plot 1) Grain	1.2E+00	1.1E-02	1.3E+00	2.9E-07	2.4E-05
18 Tanjung Karang (Plot 2) Grain	7.5E-01	5.1E-01	9.3E-01	1.4E-07	1.6E-05
19 Tanjung Karang (Plot 3) Grain	1.1E+00	1.5E-03	1.3E+00	1.9E-07	2.2E-05
20 Sekinchan (Plot 1) Grain	7.9E-01	4.2E-03	9.4E-01	1.1E-07	2.6E-05
21 Sekinchan (Plot 2) Grain	6.0E-01	-3.2E-03	7.6E-01	6.3E-08	1.2E-05
22 Sekinchan (Plot 3) Grain	6.6E-01	-3.0E-03	9.7E-01	7.2E-07	1.3E-05
23 Yan (Plot 1) Grain	7.0E-01	9.4E-03	9.1E-01	1.1E-07	1.6E-05
24 Yan (Plot 2) Grain	1.0E+00	4.6E-02	9.3E-01	8.6E-08	1.3E-05
25 Yan (Plot 3) Grain	5.1E-01	-2.1E-03	7.0E-01	8.9E-08	1.2E-05
26 Besut (Plot 1) Grain	1.1E+00	8.2E-02	1.1E+00	1.8E-07	1.9E-05

Legend :

Noncarcinogenic risk; Hazard quotient (HQ) = Cu, Cd and Cr

Carcinogenic risk; Lifetime Cancer Risk (LCR) = Pb and As

Potential Health Risk; $HQ > 1$, $LCR > 1 \times 10^{-4}$

CHAPTER 5

DISCUSSIONS

5.1 The Total and Bioavailable Heavy Metal Concentration

Overall, the results have indicated the total heavy metal concentration of paddy soil that showed an decreasing trend from Pb (0.0082 mg/kg) > Cr (0.0054 mg/kg) > Cu (0.0038 mg/kg) > As (0.0026 mg/kg) > Cd (0.0001 mg/kg), whereas the bioavailable heavy metal concentration in the rice grain was Cr (0.2701 mg/kg) > Cu (0.0896 mg/kg) > Pb (0.0458 mg/kg) > As (0.0139mg/kg) > Cd (0.0053 mg/kg). The bioavailable concentration of rice grain were seen much likely higher than the total concentration in the paddy soil.

The heavy metal concentration in the rice grain was found to be higher than paddy soil heavy metal concentration. This might be due to its direct exposure to sprayed pesticides used in the paddy fields like herbicides and fungicides used, namedly Isoprothiolane, Propiconazole and Pretilachlor, that contains heavy metal Cd, Pb and Cu (Gimeno-Garcia et al., 1995; Musa et al., 2007) and many other factors such as inorganic air pollutant through particulate matter that can seep into the leaf of plants (Bosco et al.,2015).

5.2 Geoaccumulation Index (I_{geo})

Geoaccumulation index (I_{geo}) values were all in background concentration (I_{geo}<0) level, indicating that the paddy soils were not polluted with heavy metal. Since, the index was used to assess the presence and intensity of anthropogenic contaminant deposition on soil and sediments. Thus, heavy metal were not accumulated in the soils. In case of underestimation, these might be due to the characteristics of heavy metals in the soil from anthropogenic sources, which tend to be more mobile, hence bio-available than pedogenic or lithogenic ones (Maurizio, 2016). Though, further investigation should be conducted.

5.3 Bioconcentration Factor (BCF)

While, the bioconcentration factor (BCF) were found to be potential to accumulate metal (BCF>1) for each heavy metal which correspond to be detected. This might be due to the process of evapotranspiration that could pump the heavy metal from the soil via uptake mechanism through translocation. Just as shown by Datta et al. (2013) and supported by (Irfan et al., 2013) and Tangahu et al. (2011).

5.4 Health Risk Assessment (HRA)

Potential non-carcinogenic risks of Cu and Cr were identified for both adult and children where hazard quotient, ($HQ > 1$). While, potential carcinogenic risks of As were not acceptable for adult ($LCR > 1 \times 10^{-4}$),, but both Pb and As were acceptable for children where ($LCR > 1 \times 10^{-6}$).

Previous study by Praveena & Omar (2017), also found out that heavy metals such as Pb and As, that were potential to have carcinogenic health risk to adult and children from the marketed rice grain. Thus, this present study has opened an estimation of rice grain that were obtained directly from the paddy field.

Both potential non-carcinogenic and carcinogenic health risks in this study that were resulted from the health risk assessment (HRA) should be reconfirmed. This is due to the values that were adopted from previous reports were prone to certain limitations.

In addition, the Malaysian ingestion rate (IR) and body weight (BW) values in this study were from the studies done by Department of Statistics Malaysia (2014), Norimah, Safiah, and Jamal (2008) and Zhuang, McBride, and Xia (2009). Thus, a complete questionnaire is needed to obtain for the actual Malaysian IR and BW values to be used in HRA.

CHAPTER 6

CONCLUSION AND RECOMMENDATION

In conclusion, Total concentration of paddy soil showed decreasing trend Pb (0.0082 mg/kg) > Cr (0.0054 mg/kg) > Cu (0.0038 mg/kg) > As (0.0026 mg/kg) > Cd (0.0001 mg/kg). Whereas the bioavailable concentration in the rice grain was Cr (0.2701 mg/kg) > Cu (0.0896 mg/kg) > Pb (0.0458 mg/kg) > As (0.0139mg/kg) > Cd (0.0053 mg/kg). Heavy metal concentration of rice grain is higher than paddy soil.

Geoaccumulation index (I_{geo}) with background concentration ($I_{geo} < 0$) for paddy soil samples. The bioconcentration factor (BCF) were found potential to accumulate heavy metal ($BCF > 1$) for all heavy metals.

For Health Risk Assessment (HRA) involving rice grain samples, potential non-carcinogenic risks of Cu and Cr were identified for both adult and children where hazard quotient, ($HQ > 1$). While, potential carcinogenic risks of As were not acceptable for adult ($LCR > 1 \times 10^{-4}$), but both Pb and As were acceptable for children where ($LCR > 1 \times 10^{-6}$),

As for the recommendation, the results has indicated that further research need to be focused on rice grains characteristics in order to understand its bioaccumulation capacity. The HRA also needs the real values that sync with Malaysian population to get the accurate estimation of risk.

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