



Faculty of Resource Science and Technology

Assessment of Heavy Metals in Water, Sediment and Fishes of the Baleh River, Sarawak, Malaysia

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Assessment of Heavy metals in Water, Sediment and Fishes of the Baleh River,
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DECLARATION

I declare that the work in this thesis was carried out in accordance with the regulations of Universiti Malaysia Sarawak. It is original and is the result of my work, unless otherwise indicated or acknowledged as referenced work. This thesis has not been accepted for any degree and is not concurrently submitted in candidature of any other degree.

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ABSTRACT

Pollution of heavy metals in aquatic environment is a subject of concern as rivers play an important role to the local inhabitants for various daily activities. The objective of the study is to determine the distribution and contamination status of heavy metals (Al, Cr, Cu, Mn, Ni, Zn, Fe, As, Se, Hg) in water, sediment and fishes of the Baleh River. Water and sediment samples were collected from 50 stations and 22 fish species were caught for metal analysis. For water, acid extractable metals were determined and only Al was found higher than the drinking water guideline due to enhanced surface runoff. Dissolved and total metal contents in 10 stations were compared with the acid extractable metals. The concentration of total metals were significantly higher than the dissolved and acid-extractable metals. The contamination status of sediment was evaluated using the geochemical indices with the baseline concentrations determined based on the cumulative distribution curves. The assessment suggested low contamination risk in sediment. For fishes, metals tend to be accumulated in gills, except Hg which was found higher in muscle. The metal concentration in muscle was compared against Malaysia Food Act; Al was detected at a concentration exceeding the permissible level for most species possibly due to the elevated Al concentration in water. Hg was found in higher concentrations in carnivorous and bottom feeders due to their feeding habits. The hazard index calculated was greater than 1 for some species suggesting possible adverse health effects where the consumption limits was between 143.54 g and 716.80 g per week. Overall, the study concluded low contamination risk in water and sediment of the Baleh River, with fish species demonstrating potential Al and Hg accumulation where continuous monitoring is recommended.

Keywords: heavy metals; contamination status; baseline values; risk assessment

Penilaian Logam Berat dalam Air, Sedimen dan Ikan di Batang Baleh, Sarawak, Malaysia

ABSTRAK

Pencemaran logam berat dalam persekitaran akuatik adalah satu subjek yang membimbangkan kerana ia memainkan peranan yang penting kepada penduduk tempatan untuk pelbagai aktiviti harian. Kajian ini bertujuan untuk mengkaji status dan pencemaran logam berat (Al, Cr, Cu, Mn, Ni, Zn, Fe, As, Se, Hg) di Sungai Baleh. Air dan sedimen sampel telah dikumpul dari 50 stesen dan 22 spesies ikan telah ditangkap untuk analisis logam. Untuk air, logam yang diekstrak dengan asid telah dinilai dan Al didapati tinggi daripada garis panduan air minuman akibat larian sedimen diserap masuk ke dalam air. Logam terlarut dan keseluruhan logam di 10 stesen telah dibandingkan dengan logam yang diekstrak dengan asid. Kepekatan keseluruhan logam adalah signifikan tinggi daripada logam terlarut dan logam yang diekstrak dengan asid. Status pencemaran sedimen telah dinilai menggunakan indeks geokimia berdasarkan regresi linier yang diperolehi dari keluk kumulatif. Keputusan menunjukkan bahawa tiada pencemaran logam berat di dalam sedimen. Untuk ikan, logam didapati tinggi dalam insang, kecuali Hg lebih tinggi dalam otot. Kepekatan logam dalam otot dibandingkan dengan Akta Makanan Malaysia; Al di kebanyakan spesies didapati lebih tinggi daripada nilai yang dicadangkan. Tabiat permakanan karnivor spesies juga mengakibatkan kepekatan Hg yang tinggi. Indeks bahaya yang tinggi daripada 1 untuk beberapa spesies mencadangkan bahawa kemungkinan kesan kesihatan yang berbahaya manakala had pemakanan adalah di antara 143.54 g hingga 716.80 g seminggu. Secara keseluruhannya, kajian ini menunjukkan bahawa pencemaran yang rendah di dalam air dan sedimen di Sungai Baleh, dengan potensi pengumpulan Al dan Hg dalam ikan manakala pemantauan berterusan adalah dicadangkan.

Kata Kunci: *logam berat; status pencemaran; nilai-nilai asas; penilaian risiko*

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LIST OF ABBREVIATIONS

CF	Contamination factor
DO	dissolved oxygen
CRM	Certified Reference Materials
FAO	Food and Agriculture Organization
GPS	Global positioning system
HI	Hazard index
I_{geo}	Geo-accumulation index
LoD	Limit of detection
LoQ	Limit of quantification
MFA	Malaysia Food Act
MP-AES	Microwave Plasma Atomic Emission Spectrometer
NSDWQ	National Standard Drinking Water Quality
PCA	Principal Component Analysis
PLI	Pollution load index
WHO	World Health Organization

CHAPTER 1

INTRODUCTION

The presence of heavy metals in the aquatic ecosystem is of great concern due to their potential to be accumulated in the food chains. Heavy metals can be introduced into the aquatic environment *via* anthropogenic input such as industrial discharge, mining, logging and agricultural activities (Ogoyi *et al.*, 2011; Ling *et al.*, 2012). Metals such as Fe, Cu, Zn and Mn are essential elements as they play an important role in various biochemical and physiological functions. However, excess intake of essential metals could cause detrimental effects. Other elements such as Hg, Pb and Se are non-essential as they are toxic, even in trace amount (Somer, 1974; Kalay & Canli, 2000).

Heavy metals that are discharged into the aquatic environment will undergo several pathways including adsorption, complexation and precipitation of which adsorption is the primary process due to the strong affinities of metals for Fe and Mn hydroxides, as well as organic metals (Nobi *et al.*, 2010). Ahmad *et al.* (2008) evidenced that metals are highly accumulative in sediments due to the presence of organic matter in sediment. Hence sediment is a sink for heavy metals, capable of accumulating tremendous metals concentration (Hasrizal *et al.*, 2015). The accumulation, bioavailability and toxicity of metals in aquatic environments are governed by their speciation which is affected by physical and chemical factors such as pH, salinity, temperature, redox potential and types of organic ligands (Tokalioglu *et al.*, 2000; Ebrahimpour & Mushrifah, 2008; Sim *et al.*, 2015). Under favorable conditions, metals are convertible from non-solubilize form into solubilize form, becoming more available hence causing pollution of water bodies (Sahuquillo *et al.*, 2003; Nobi *et al.*, 2010). Typically under low pH condition, increased competition between hydron, H^+ and dissolved metals for ligands

could increase the dissolution of metals in water bodies (Idriss & Ahmad, 2013; Hasrizal *et al.*, 2015). Likewise, under low dissolved oxygen environment, the mobility of some elements Cd, Cu, Pb and Zn can be improved (Atkinson *et al.*, 2007). The presence of heavy metals in water may cause toxicity in aquatic organisms even at trace level. The metals can be transferred to human *via* food chain and become toxic when they are taken in excess (Jaishankar *et al.*, 2014). For example, excess intake of Al has a risk of development of Alzheimer disease while overdose of Pb can cause disorder to central nervous system (WHO, 2006).

Several studies on heavy metals have been conducted to assess the degree of heavy metal contamination in the river systems. Most of the heavy metals studies focused on gathering the data related to the levels of toxic metals in the environment (Saeed & Shaker, 2008; Ebrahimpour & Mushrifah, 2008; Bouraie *et al.*, 2010; Sany *et al.*, 2013). Various quantitative indexes such as geo-accumulation index and contamination factor are employed to evaluate the contamination status of heavy metals in order to identify, monitor and control the sources of the metals (Wan *et al.*, 2012; Seyedeh *et al.*, 2013).

The Baleh River, locally known as Batang Baleh, is located at about 95 km from its confluence with the Rajang River in Kapit Division, Sarawak, Malaysia. The river is identified potentially viable for hydroelectric development (Wong, 2014). Currently, the area is largely covered with natural vegetation and sparsely populated with limited accessibility. Logging is the most prominent activity in the vicinity that leads to massive debris carried down from Melatai River, upstream of the Baleh River in 2010 (Sibon, 2010). Other development in Baleh includes construction of a bridge which was undertaken in 2013

(NewsDesk, 2013). The distribution of heavy metals in the environment is a concern as the river serves an important role to the local inhabitants for various daily activities as means of transportation and source of protein. Despite the increasing development, the status of heavy metals in the environment of the Baleh River is scarcely reported. It is essential to investigate the distribution of heavy metals in the Baleh River and its tributaries, whether the land use activities have contaminated the river and whether fishes caught from the river are safe for consumption with regards to the metal contents. According to studies, logging may contribute to an increase of metal loadings of the river (Ahmad *et al.*, 2008; Rohasliney *et al.*, 2014; Soo *et al.*, 2016).

In view of the use of the Baleh River for various daily activities as means of transportation and source of protein, it is important that a comprehensive study is conducted to evaluate the status of heavy metals in water, sediment and fishes of the Baleh River. Therefore, the aim of this study is to determine and evaluate the heavy metals present in water, sediment and fish of this river. It is essential to capture the baseline data before further development is introduced. The study would provide baseline data of heavy metals of the Baleh River serving as the benchmark for future monitoring. The specific objectives are

1. To determine the distribution of heavy metals in water, sediment and fishes along the Baleh river;
2. To assess the contamination status of the Baleh River;
3. To compare the acid-extractable, dissolved, and total dissolved metal concentration;
4. To assess the human health risk of metals in fishes.

CHAPTER 2

LITERATURE REVIEW

2.1 Heavy metals

Heavy metals are metallic elements of the periodic table with relatively higher density than water. Pollution of heavy metals in the aquatic ecosystem is a current issue and it may reach an alarming state due to their non-biodegradable and accumulative nature. Metals are naturally occurring in the Earth's crust. They are mainly introduced into the aquatic environments *via* natural weathering and volcanic eruption (Bradl, 2002). However with the escalating anthropogenic activities, the metals loadings in the environment have consistently increased (Ogoyi *et al.*, 2011; Stihl *et al.*, 2006). As heavy metals are not degradable, they are continuously deposited and integrated in water, sediment and aquatic organisms leading to heavy metal pollution in water bodies (Ogoyi *et al.*, 2011). Despite the toxicological effect of heavy metals, elements such as Cu, Fe, Mn, Ni and Zn are essential nutrients to support various biochemical and physiological functions of living cells when in trace amount (WHO, 1996). Table 2.1 show the permissible guideline and effect of heavy metals present in drinking water according to WHO (2006).

Studies revealed that more than 90 % of heavy metals in the marine environment have settled down and integrated into sediment (Amin *et al.*, 2009; Zahra *et al.*, 2013; Zheng *et al.*, 2008). These elements are deposited and immobilized through adsorption, complexation and are integrated into the lattice structure (Du *et al.*, 2009; Lin *et al.*, 2013). Adsorption takes place as a result of strong affinities for Fe and Mn oxyhydroxides and particulate organic matter. Some free metals ions remain dissolved in water due to their specific chemical states which can be altered depending on the physical and chemical characteristics of water. For example,

low pH could enhance the metal solubility and decrease the adsorption capacity of solid surface.

Table 2.1: The permissible guideline and effect of heavy metals in drinking water (WHO)

Elements	Permissible level (mg/ L)	Effect
Aluminum	0.2	Risk factor for the development of Alzheimer disease
Arsenic	0.01	Possibly carcinogen
Chromium	0.05	Possibly carcinogen
Copper	2	Essential trace element; toxic to plant and algae in high level
Iron	1	Essential trace element; toxic at high level
Lead	0.01	Central nervous system failure
Manganese	0.4	Essential trace element; cause neurological toxic effect
Mercury	0.006	Kidney failure
Nickel	0.07	Possibly carcinogen
Selenium	0.01	Essential trace element; toxic at high level
Zinc	3	Essential trace element; toxic to plant at high levels

2.1.1 Aluminum (Al)

Al is the third most abundant element in the Earth's crust (David, 2005). It comprises of about 0.9 to 6.5 % in soils. The permissible level of Al in drinking water is 54 µg/L which is much lower than the typical concentration in freshwater streams, 400 µg/L, according to APHA (2005). The occurrence of Al in natural waters is mainly affected by pH and suspended mineral particles. It is a non-essential element for living organisms. At pH less than 4, Al exists in cation form, Al^{3+} while $\text{Al}(\text{OH})_4^-$ dominates at higher pH. A concentration higher than 1.5 mg/L would lead to toxic effect whilst concentration lowers than 200 µg/L poses a minimal risk. Excessive intake of Al by humans could cause Alzheimer's disease.

2.1.2 Arsenic (As)

As is a metalloid element which has chemical and physical properties between a metal and a non-metal (WHO, 2001). Arsenic mainly exists in four oxidation states, -3 , 0 , $+3$, and $+5$. The oxidation state of arsenite, AsO_3^{3-} is common under reducing conditions while arsenate, AsO_4^{3-} predominates under oxygenated conditions (WHO, 2001; Atamirano-Lozano *et al.*, 2006). Arsenic can be classified into organic and inorganic forms. Inorganic arsenic is mainly introduced into the environment through anthropogenic activities and may pose harmful threats to human health at high level (WHO, 2001). According to APHA (2005), the natural level of As is typically less than $10\text{ }\mu\text{g/L}$. It is harmful to aquatic organisms at concentration exceeding $72\text{ }\mu\text{g/L}$. The maximum concentration should not exceed $140\text{ }\mu\text{g/L}$. The chemical state of As is affected by several factors including the redox condition, the degree of biological activity, type of water source and anthropogenic sources (WHO, 2001). The background concentrations of As in soil is about 5 mg/kg however it may range from as low as 1 mg/kg to as high as 40 mg/kg . The variation of As in soils depends profoundly on the geological formations (WHO, 2001).

2.1.3 Copper (Cu)

Cu is an essential trace element but intake of more than 15 mg/day can cause health problem such as skin allergies, nausea and diarrhea (ATSDR, 2007). Cu is commonly found in the environment and contamination of Cu is often associated with mining, corrosion of Cu pipes and burning of coal. Cu is a transitional metal and there are four oxidation states: elemental Cu, cuprous ion Cu (I), cupric ion Cu (II) and Cu (III) (Georgopoulos *et al.*, 2001). Cu is found in suspended, colloidal and dissolving states in water. The dissolving form of Cu, mainly Cu^{+2} , will bind to organic and inorganic ligands through complexation at higher pH

(Odobasic, 2012). On the contrary, lower pH will increase the solubility of Cu. The bioavailability of Cu is influenced by the water hardness, redox potential, ionic strength, suspended matter, presence of complex ligands and interaction between water and sediments. Dissolved Cu can be adsorbed into aquatic organisms through the skin resulting in accumulation (Odobasic, 2012).

2.1.4 Iron (Fe)

Fe is naturally abundant in the environment. It plays an important role for oxygen and electron transfer acting as a metalloenzymes such as oxidase, reductases and dehydrases (Cotton & Wilkinson, 1998). Fe is also a starter for some chemical processes in water due to its high reactivity. The speciation of trace elements in the aqueous environment can be influenced by the concentrations of Fe (III) and Fe (II). For example, Fe (III) precipitates completely into hydrous ferric oxide and controlling the mobility and toxicity of other metals. The solubility of ferrous ion (Fe^{2+}) is affected by the carbonate concentration. Additionally, ferrous ion (Fe^{2+}) can be oxidized into ferric state (Fe^{3+}) and may be hydrolyzed to form red, insoluble hydrated ferric oxide when it is in contact with air or additional oxidants. Ferric ions mostly in complex form and only dissolved into water during acidic condition (APHA, 2005). Fe overload can cause lethal effect and may lead to several health diseases, such as fatigue, weakness, joint pain, abdominal pain, or organ damage (ATSDR, 2007).

2.1.5 Lead (Pb)

Pb is a toxic metal which could cause illnesses to the body system such as neurological, hematological, cardiovascular and renal systems. Essentially, Pb is rare in the Earth's crust and exists primarily as Pb sulfide (Atamirano-Lozano, 2006). Human activities such as

refining and burning fossil fuels, mining and manufacturing are associated with release of Pb into the environment (WHO, 2010b). The common aqueous species of Pb are Pb^{2+} hydroxide and carbonate complexes (APHA, 2005). According to APHA (2005), the average concentration of Pb in the Earth's crust is 13 ppm. In soils, it ranges from 2.6 to 25 ppm. In streams, it is 3 $\mu\text{g/L}$, and in ground waters, it is normally less than 0.1 mg/L. In air, its concentration may be lower than 1.5 $\mu\text{g/m}^3$. Once Pb exceeds the permissible level of 0.015 mg/L, additional removal steps are necessary for the water treatment plant. Similar to other elements, Pb tends to dissolve into water at lower pH. The element is insoluble at higher pH (ATSDR, 2007).

2.1.6 Manganese (Mn)

Mn is the 12th most abundant element in the Earth's crust. It is generally found in rocks, soil, groundwater, and surface water (Nadaska *et al.*, 2012). Anthropogenic activities could introduce Mn into the environment. There are three general oxidation states: Mn (II), Mn (III) and Mn (IV). Mn (II) is the most stable form in aqueous solution, while Mn (III) and Mn (IV) are only stable in the solid phase of soil (Mcbride, 1994; APHA, 2005). The mobility of Mn in soil is greatly influenced by pH, wetness, organic matter content and biological activities. An acidic condition with low redox potential will render Mn more readily dissolved into the water increasing the mobility of Mn (Walna *et al.*, 2010). However Mn will form complexation with organic matter, oxides and silicates at pH above 6 reducing its solubility. The concentration of Mn in water should not exceed 0.5 mg/L as it will cause disorder of the center nervous system (ATSDR, 2012).

2.1.7 Mercury (Hg)

Hg is a toxic element that can be found in the environment. The most common form of mercury, which is inorganic mercury could enter the environment naturally or via anthropogenic activities (Bella & Hilliker, 2003; Bose-O'Reilly *et al.*, 2010). The concentration of Hg in the Earth's crust is about 0.09 ppm. In soil, it ranges from 30 to 160 ppb. In stream and groundwater, it is 0.07 µg/L and 0.5- 1 µg/L, respectively (APHA, 2005). Under low pH and anoxic condition, Hg can undergo methylation process and transform into most toxic form, which is methylmercury. Formation of methylmercury is controlled by changes in the nutrients and oxygen level, temperature and organic matter content (Bella & Hilliker, 2003). Methylmercury is hard to eliminate from the body and it can be transferred to higher trophic level of food chain due to its bioaccumulation properties. Excess methylmercury in human body can cause neurotoxin and has the ability to cross the blood-brain and placental barriers affecting the neuron system (Bose-O'Reilly *et al.*, 2010).

2.1.8 Nickel (Ni)

Nickel is a transition metal that is commonly found in the natural environment and plays an important role in many biochemical processes. The concentration of Ni in the Earth's crust is 1.2 ppm. In soil, the concentration is typically 2.5 ppm and in streams, it is often less than 1 µg/L (APHA, 2005). The aqueous form of Ni is controlled by pH, redox conditions, ionic strength, temperature and the form of inorganic and organic ligands (Mandal *et al.*, 2002; Cornellis *et al.*, 2005). The most common aqueous species is Ni (II). Ni formed complexation with hydroxide, carbonates and organic ligand in oxidation conditions (APHA, 2005). High Ni concentration in human body can cause various health effect including skin allergies, lung

fibrosis, cancer of the respiratory tract and iatrogenic nickel poisoning (Cempel & Nickel, 2006).

2.1.9 Selenium (Se)

Se is a semi-metal and it possesses intermediate properties between a metal and a non-metal. The natural forms of Se are selenite (SeO_3^{2-}), selenide (Se^{2-}), selenate (SeO_4^{2-}) and the selenium element (Mehdi *et al.*, 2013). These anionic forms are highly soluble, mobile, bio-available and potentially toxic. They are greatly influenced by pH (Mehdi *et al.*, 2013). In water, Se originates from atmospheric deposits or contaminated soil drainage. Selenite and selenide may be released through erosion. Selenate and selenide mostly present in aqueous state where selenite is easily bound to suspended sediments. The concentration of Se in the Earth's crust is about 0.2 ppm. In soil, it ranges between 0.27 and 0.74 ppm while in stream, it is typically 0.2 $\mu\text{g/L}$ (APHA, 2005).

2.1.10 Zinc (Zn)

Zn is generally found in the environment due to natural processes and human activities including mining and smelting operations, domestic and industrial sewage, road surface runoff and erosion of agricultural soils. The average abundance of Zn in the Earth's crust is 76 ppm; in soils it ranges from 25 to 68 ppm; in streams is 20 $\mu\text{g/L}$, and in ground water it is less than 0.1 mg/L (APHA, 2005). Zn has negative redox potential and is readily oxidized in water. Under acidic condition, the solubility of Zn is increased and exists as Zn (II). Meanwhile at pH 6, it is in divalent form that may bound to organic substances or present as ZnOH^+ . The toxicity of Zn is governed by the water hardness, alkalinity, and organic matter content (Odobasic, 2012).

2.2 Heavy metals in water and sediment

As reported, about 90 % of heavy metals are accumulated in sediment and only a small amount is dissolving in water bodies (Amin *et al.*, 2009; Zahra *et al.*, 2013; Zheng *et al.*, 2008; Ikem *et al.*, 2003), hence, sediment acts as a sink for heavy metals. Hasrizal *et al.* (2015) examined the distribution of Al, Fe, Mn, Zn, Cu and Co in surface sediments of the East Coast of Peninsular Malaysia. The results revealed that Zn-Fe ($r = 0.71$, $P < 0.05$) and Al-Cu ($r = 0.63$, $P < 0.05$) were correlated suggesting that these elements were probably originated from similar sources (Hossain *et al.*, 2015). Authors also reported that the distribution pattern of Zn is closely related with iron-rich deposits as iron hydroxide are able to adsorb large quantities of metals through cation exchange process; in addition, iron oxides also play an important role in trapping metals in aquatic sediment (Horowitz & Elrick, 1997). Besides that, the concentration of Al was higher than other elements indicating its origin from both natural and anthropogenic sources. The aluminium-rich minerals are easily transported to the water via surface run-off (Chester, 2000).

Similary, Seyedeh *et al.* (2013) examined the distribution of As, Cu, Cd, Cr, Ni, Pb, Zn, Pb, Mn, Al and Fe in the coastal sediments of Pork Klang, Selangor, Malaysia. High concentrations of Cu (17.43 mg/kg), Pb (59.45 mg/kg) and Cr (46.40 mg/kg) were found in the areas adjacent to surface runoff and industrial outlets, which can directly release organic and inorganic pollutants. Generally, the concentration of Cu (10.34 mg/kg), Cd (0.22 mg/kg), Cr (27.00 mg/kg), Ni (4.16 mg/kg), Pb (31.30 mg/kg), and Zn (26.00 mg/kg) were found lower at the stations with strong water current which reduces the chemical reactions (adsorption and complexation) between metals and sediments (Saeed & Shaker, 2008).

Wan *et al.* (2012) examined the geochemical factors that influence As, Cd, Cr, Ni and Pb in water and sediment of the Langat River, Malaysia. The results revealed that the concentration of As and Cr in sediment were negatively correlated with sediment pH as both elements are immobile anion under acidic condition and tend to be sorbed by soil colloids (Hooda, 2010). On the contrary, the concentration of Ni is positively correlated with sediment pH and this indicated that as pH increased so did the binding of Ni to the sediment solid phase. According to Campbell and Stokes (1989), increasing pH would reduce Ni mobility as insoluble hydroxides or carbonates will be formed. Positive correlation was similarly established between salinity and As, Cr as well as Ni in water according to the study of Wan *et al.* (2012). This is because the increased salinity would increase the competition between cations and metals for binding sites on clay-organic particles surface. Due to this antagonistic relationship, more metals will be desorbed from the sediment into the water column (Elzahabi & Yong, 2001).

Ling *et al.* (2012) determined the concentration of Fe, Zn, Pb, Cu and Cd over rainy and dry months in a tropical river. The findings reported that the concentration of heavy metals increased during rainy season due to runoff containing fertilizers and overflowing from the adjacent oxidation ponds. The findings of Ling *et al.* (2012) revealed that Cr exhibited higher concentration under aerobic condition with pH above 7 due to the formation of hexavalent chromium, which is soluble in water (Gauglhofer & Bianchi, 1991). Besides that, higher concentration of Zn was found in water near the stations with batteries, old tyres, glasses and plastics that serve as potential source of Zn (Cretaz & Barten, 2007).

Idriss and Ahmad (2013) determined the concentrations of Ni and Cr in sediment of the Juru River, Penang, Malaysia and investigated the effects of pH and organic matter. The results revealed that the presence of organic substances under reducing condition facilitated the binding of Ni and Cr onto sediment (Ritchie & Posner, 1982). Besides that, acidic pH ranges from 3.96 to 6.59 also showed higher capability for the metals to be bound onto sediment. Ni appeared to associate with sediment matrix becoming less mobility in water bodies. Cr is a stable element that can only associate with carbonate and exchangeable state under acidic condition.

Assessment of metals including As, Cu, Cr, Cd, Ni, Pb, Hg and Zn in surface waters and sediments was carried out by Sany *et al.* (2013). The results revealed that the concentrations of Cd, Ni, Zn, Hg, As and Cr in sediment decreased with increasing rainfall. This suggests that the exchangeable metals are easily disturbed by the flowing water (Lim *et al.*, 2006; Li *et al.*, 2009). Additionally, less anthropogenic activities such as shipping and logging were carried out during wet season and hence, metals concentration in water column becomes lower compared to dry season. However, the concentration of Zn and Cu appear to be higher during wet season suggesting that these elements are mainly sheared off from soil erosion.

Similarly, Bouraie *et al.* (2010) determined the heavy metal concentrations in surface river water and sediments at the Nile Delta in Egypt. The results showed that concentrations of heavy metals were the highest at the discharge point of industries. Noticeably, most the concentrations of heavy metals in water were higher during winter closure period due to lower pH and microbial activities (Bouraie, 2010). At higher pH, heavy metals may precipitate as insoluble hydroxides, oxides and carbonates. Besides, there is a significant positive

correlation between the concentration of Fe and Al. This suggests that Fe and Al originate from same source during discharge process (Degroot & Allersma, 1975). However, concentration of Mn and Zn was found lower during winter closure period. This may due to the consumption of these two elements by phytoplankton (Emerson & Lewis, 1939).

Ebrahimpour and Mushrifah (2008) determined Cd, Cu and Pb in water and sediment. The results showed that heavy metals were released under acidic condition corroborating the findings of Samecka-Cymerman and Kempers (2001). Tokalioglu *et al.* (2000) likewise reported increase of Cd, Cu and Pb in water in acidic condition. This is because the hydrogen ions in water column are competing with the metals for binding sites consequently releasing the metal ions (Ersoz & Barrott, 2012).

A study by Başıyigit and Tekin-Özan (2012) reported that dissolved oxygen level in water is negatively correlated with the concentration of Cd, Cr, Cu, Fe, Mn, Mo, Ni, Pb and Zn. On the other hand, temperature is negatively correlated with Cu and Se but positively correlated with other metals. This is because the dissolved oxygen was reduced during higher temperature. As a consequence, the dissolution metals in sediment were encouraged. The finding was in good agreement with the study of Atkinson *et al.* (2007) who reported that metals bound onto sediment were easily disturbed and released into water bodies under the condition of low dissolved oxygen, especially Fe and Mn, due to the slower oxidative precipitation.

Ogunfowokan *et al.* (2013) studied the speciation of heavy metals in water and sediments from the Asunle River, Nigeria. The concentration of Zn, Mn, Cu, Ni and Cd appeared to be

higher in water due to low association with minerals to form crystal structure and low capability to bind with organic matters in sediment (Sposito *et al.*, 1982). The mobility factor was studied where Ni exhibited the highest mobility of 92.48 %. Elements high in mobility appeared to be more bioavailable and susceptible to transformation (Xian, 1987). The study concluded that the level of dissolved heavy metals is much lower than their background levels in sediment.

2.3 Assessment of contamination

The geo-accumulation index (Igeo) introduced by Muller (1969) is used to better understand the degree of contamination in surface sediments, and these results reflect the effect of lithogenic sources (Wan *et al.*, 2012; Hasrizal *et al.*, 2015; Mohamad *et al.*, 2015). The geoaccumulation index is calculated by comparing the baseline levels with the concentration of metals detected in sediment. Contamination factor (CF) was applied to assess the state of conservation of an environment and to monitor the contamination status (Fishbein, 1981; Davis *et al.*, 2009). In the study of Seyedeh *et al.* (2012), the contamination factor was applied to evaluate sediment of the Port Klang coastal area, Selangor, Malaysia. The findings indicated that all metals studied were influenced by anthropogenic inputs, especially toxic elements such as As, Cd, Hg and Pb that were enriched at stations close to the berth line and the Klang River mouth (Seyedeh *et al.*, 2012). Pollution load index (PLI) was applied to evaluate the overall level of soil pollution across the sampling sites (Bhuiyan *et al.*, 2010). The PLI is a simple and useful means for assessing the level of heavy metal pollution. Four pollution levels were defined: no pollution ($PLI < 1$), moderate pollution ($1 < PLI < 2$), heavy pollution ($2 < PLI < 3$), and extremely heavy pollution ($PLI > 3$) (Liu *et al.*, 2014).

2.4 Determination of heavy metals in fish

Fishes are on the higher trophic level of the aquatic food chain. They can accumulate heavy metals from food, water and sediment (Yilmax *et al.*, 2007; Zhao *et al.*, 2012). Overload of heavy metals in fishes will cause lethal effects such as renal failure, liver damage, cardiovascular diseases and even death (Al-Busaidi *et al.*, 2011). Human are exposed to the risk of heavy metals via consumption of fishes (Luoma & Rainbow, 2008).

Ley *et al.* (2016) determined the level of Hg, MeHg and Se in grey-eel catfish (*Plotosus canius*) and giant mudskipper (*Periophthalmodon schlosseri*) from the Strait of Malacca. The results revealed that *Periophthalmodon schlosseri* accumulated higher concentration of Hg in the gill membrane due to the direct uptake of Hg from the surrounding water during the respiration process. On the other hand, the liver of *Periophthalmodon schlosseri* was found to contain the highest levels of total Hg, MeHg and Se, followed by the gastrointestinal tract. According to Regine *et al.* (2006), gill was a good indicator of direct exposure of Hg because it controlled the adsorption of Hg prior to transportation to the targeted organs *via* the circulating system. The study of Ley *et al.* (2016) revealed that Se concentration in muscle was negatively correlated ($P < 0.05$) with Hg in both species. According to Burger *et al.* (2013), the concentration of Hg was influenced by the Se content in the environment but further mechanistic studies are needed to examine the correlation.

Alina *et al.* (2012) determined the Hg, As and Pb in marina fish and shellfish along the Straits of Malacca. The results revealed that bottom feeding species (demersal) accumulated higher concentration of Hg, As and Pb than pelagic species, which inhabit in the upper layer of river (Fisheries Research Institute, 2004). This finding was corroborated by Ahmad *et al.* (2008) on

the study of temporal variability of heavy metals in fish of Tasik Chini, Penisular Malaysia. According to Ahmad *et al.* (2008), bottom feeder tends to accumulate heavy metals as metals deposition in sediment is easily taken up for accumulation. Despite the profound concentrations of metals in demercial species caught in the Straits of Malacca and Tasik Chini, the levels reported were relatively lower than those samples from Langkawi Island (Irwandi & Farida, 2009).

Rohasliney *et al.* (2014) determined Cd, Ni and Pb levels in fish from the Kelantan River, Malaysia. The findings revealed that the concentration of Cd, Ni and Pb in fish collected during wet season was significantly higher ($P < 0.05$) compared to the dry season. According to Igwegbe *et al.* (2015), the increase of surface runoff during wet season has increased the dissolution of metals, which in turn, increases the metal uptakes by fishes. However, the finding of Obasohan and Eguavoen (2008) was in contrast with the results reported by Rohasliney *et al.* (2014). Obasohan & Eguavoen (2008) reported that the heavy metals in fish were higher during dry season due to high temperature, which increased the activity, ventilation, metabolic rate and feeding habits (Nussey *et al.*, 2000). The low heavy metal concentrations in the wet season on the other hand were associated with the dilution of metals (Obasohan & Eguavoen, 2008). Rohasliney *et al.* (2014) also reported that the herbivorous species (*B. schwanenfeldii* and *O. hasseltii*) tend to accumulate lower concentration of metals due to their lower trophic level where they feed on the aquatic macrophytes and submerged land plants (Ambak & Zaidi, 2010).

Renieri *et al.* (2014) examined the occurrence of Cd, Pb and Hg in fish of the Mediterranean region. According to Castro-González *et al.* (2008), the fish can accumulate metals by

ingestion of food and direct penetrate through the gills. Several studies indicates that the accumulation of metals in fish tissues are difficult to predict because it is governed by numerous abiotic and biotic factors such as temperature, salinity, pH, species, gender, age, size and physiologic conditions (Storelli *et al.*, 2005; Castro-González *et al.*, 2008; Conti *et al.*, 2012). However, accumulation of metals in fish tissues generally depends on the ability of metal binding proteins such as metallothioneins. The metal accumulation is often evaluated based on the concentrations in gills as they are the uptake site of waterborne ions and directly reflect the concentration in water bodies. Cd is easily affected by the salinity and temperature in the aquatic environment and this element is found usually in trace in the water bodies (Creti *et al.*, 2010; Castro-González *et al.*, 2008). Cd can bind with Ca^{2+} leading to serious hypocalcaemia and eventually death in aquatic organisms. Meanwhile, Pb does not originate from diet but most likely from contaminated water (Creti *et al.*, 2010). The metal accumulation ability of fishes is largely affected by its natural habitat as well. Benthic behavior fish which lives at the bottom of the river will accumulate higher degree of heavy metals due to the closer relationship with sediment. Predator at higher trophic levels appeared to accumulate higher Hg concentration due to higher mobility and metabolism (Marcotrigiano & Storelli, 2003). Meanwhile, carnivores or large predator accumulate higher metals due to highly active metabolism and high food consumption (Damiano *et al.*, 2011).

Bashir *et al.* (2013) examined the concentration of heavy metals in fishes from the coastal waters of Kapar (industrialized zone) and Mersing (less industrialized zone) in Malaysia (Bashir *et al.*, 2013). Microwave digestion method was used because it is more efficient due to shorter time, less acid consumption and volatile compounds are able to be retained in the solution (Gulmini *et al.*, 1994; Krushevska *et al.*, 1993). The results showed that muscle

demonstrated lower metal concentrations compare to liver and gills. Liver acts as the metal storage, hence accumulating higher concentration of metals (Agusa *et al.*, 2005). Meanwhile, gills as a part directly exposed to the water bodies is highly vulnerable to toxic chemicals due to its large surface area that facilitates toxicant interaction and absorption (Evans, 1987).

Similarly, the concentrations of heavy metals (Cu, Zn, Pb, Cd, Fe and Mn) were analyzed in the liver, gills and muscles of benthic and pelagic fish species (El-Moselhy *et al.*, 2014). The results revealed that muscles possessed the lowest concentrations of all metals while liver accumulated the highest concentration Cu, Zn and Fe. The highest concentration of Pb and Mn are found in gills. Gills is the part involves in of metal ion exchange from water and has large surface area that facilitate the adsorption of toxic metals (Dhaneesh *et al.*, 2012). A few studies revealed that Pb tend to accumulate in gills (Kargin, 1998; Avenant-Oldewage & Marx, 2000; Zhao *et al.*, 2012) while Eisler (2010) reported that gills accumulate higher concentration of Mn compare to soft tissues, muscle. The findings of El-Moselhy *et al.* (2014) reported that pelagic fish species accumulated higher concentration of Cd in muscle which is in contrary to the findings of other researchers (Yi *et al.*, 2011; Renieri *et al.*, 2014). Zhao *et al.* (2012) reported that low mobility of fish which has limited range of activity will accumulate less heavy metal. Results of El-Moselhy *et al.* (2014) also revealed that large fish showed very high concentrations of Cd in liver where the finding is associated to the age of the fish since Cd is difficult to be excreted from liver (Khaled, 2004).

2.5 Health risk assessment

Hazard Quotient (HQ) or Hazard Index (HI), set by the US Environmental Protection Agency, are commonly used to assess the potential health risks associated with exposure to a variety of metals *via* fish ingestion (USEPA, 2000). Both measures are often estimated according to the experimental data and modelling assumptions (Zhao *et al.*, 2012). Several studies have evaluated the human health risk of Hg in fish based on the hazard index (HI) calculated according to the estimated daily dose and reference dose (Mohamad *et al.*, 2015; Sim *et al.*, 2016; Ahmad *et al.*, 2016). Hazard index < 1 suggests unlikely adverse health effect whereas HI > 1 indicates probability of adverse health effects (Sim *et al.*, 2016). In the study of Sim *et al.* (2016), the fish consumption rate and body weight for different populations are assumed based on the guideline of US EPA (Health Canadian, 2007). On the other hand, Ahmad *et al.* (2016) adopted the body weight and fish consumption rate from the study of Azmi *et al.* (2009) for Malaysia population. Similarly, Alam *et al.* (2015) evaluated the contamination of heavy metal in tilapia fish collected from Langat Basin, Malaysia based on hazard quotient and hazard index. According to Alam *et al.* (2015), US EPA fish advisories have made available to the public a compendium of information on locally issued fish advisories and safe eating guidelines to inform people about the recommended level of consumption for fish caught in local waters. Unfortunately, this information is not well developed in Malaysia.

2.6 Determination of heavy metals in reservoir

Avila-Pérez *et al.* (1999) studied the heavy metal concentrations in water and bottom sediments of Maxican reservoir. The findings revealed that Zn, Fe, Cr and Pb were higher before flowing into the reservoir while concentration decreases when it is nearer to the dam due to the self-sedimentary ability. Sediment is primarily composed of silicates and aluminium silicates in the form of clays that facilitates the adsorption of metals onto sediment and encourages the self-cleaning process (Dekov *et al.*, 1997). The Pearson correlations analysis demonstrated intermetallic correlation between Fe-Zn, Fe-Pb, Zn-Pb in surface water; whilst Cu, Zn and Pb in sediment were positively correlated. The result indicated that these elements shared similar contamination sources.

Kasper *et al.* (2014) studied how reservoir stratification affects methylmercury levels in river water, plankton and fish downstream. The results revealed that the concentration of MeHg in anoxic hypolimnetic water was higher than surface water and decreased gradually until 200 km downstream from the dam during dry season. The reservoir stratification provided anoxic hypolimnetic condition which facilitates the accumulation of MeHg (Canavan *et al.*, 2000). The MeHg accumulation increased after 200 km downstream and is mostly accumulated by planktons and fishes and this suggested that MeHg exported to downstream were accumulated by biota. This may due to the presence of large deep ponds that may trap MeHg-rich particles (Schetagne & Verdon, 1999). Hence, Hg in water and sediment may be influenced by the distance downstream from a dam. On the other hand, tributaries below the reservoir may dilute the concentration of Hg below the dam (Schetagne & Verdon, 1999). However, Li *et al.* (2013) indicated that there is no significant difference in Hg levels in water and plankton from

100 km downstream from the dam for a reservoir less than 1 year old. MeHg concentration is lower during wet season and this may be due to the dilution factor (Canavan *et al.*, 2000).

Tuomola (2005) examined the mercury levels in fish five years after construction of Lago Manso Reservoir, Brazil. There is a significant correlation between weight and length with the Hg levels. The results suggest that larger fishes exhibit higher Hg levels corroborating the findings of Driscoll *et al.* (1995) and Lindqvist *et al.* (1991). Besides, carnivores fish appear to accumulate higher Hg compare with herbivores and omnivores fishes. *Pintado* caught from downstream recorded 108 ng/g mercury in year 1999 before impoundment; this value increased to 440 ng/g in the year 2002 after construction of the dam. Meanwhile, *Dourado* recorded 817 ng/g of mercury in year 2002 but the concentration increased to 1323 ng/g in 2005. In this study, fishes of comparable length and weight were compared. After 1 year, Tuomola *et al.* (2008) examined the mercury development in relation to abiotic characteristics and carbon sources of this reservoir again. The dissolved organic carbon has increased while dissolved oxygen was decreased, even lower at downstream after six year impoundment of dam. This condition would favor the bioavailability of Hg (Tuomola, 2005). Low dissolved oxygen may be a result of decomposition of vegetation after flooding. Hg concentration in all fishes, both in the reservoir and downstream, were found to increase significantly, five times or more than the background values. The results indicate that impoundment of hydroelectric dam is associated with the bioaccumulation of Hg in fishes.

2.7 Metal quantifying methods

According to Habibah *et al.* (2014), heavy metals in sediment can be fractionated into four geochemical phase; they are the easily leacheable and ion exchangeable (ELFE) fraction, acid reducible (AR) fraction, organic oxidizable (OO) fraction and resistant fractions (RR) (Badri, 1984; Habibah *et al.*, 2014). In ELFE fraction, the adsorbed metals on exchangeable surfaces of clay minerals and organic matter were extracted by using centrifugation with addition of ammonium acetate ($\text{NH}_4\text{CH}_3\text{COO}$) (Khairiah *et al.*, 2012). Metals combined with hydrous and amorphous Fe and Mn oxides, carbonates and sulphides were extracted as AR fraction using centrifugation with addition of hydroxylamine hydrochloride ($\text{NH}_2\text{OH.HCl}$). For OO fraction, metals were leached out using ammonium acetate acidific with hydrochloric acid (HCl) (Khairiah *et al.*, 2012). Lastly, heavy metals trapped in the mineral silicates or in the basic rock at each particular area, referred to as RR fraction, were extracted using acid digestion method with nitric acid and perchloric acid (Habibah *et al.*, 2014).

Acid digestion is the method commonly used to extract heavy metals from water and biological samples (Ashraf *et al.*, 2012; Ling *et al.*, 2012; Wan *et al.*, 2012; Idriss & Ahmad, 2013; Seyedeh *et al.*, 2013; Sim *et al.*, 2015). In the study of Ashraf *et al.* (2012), two types of digestion procedures were applied which are dry-ashing and wet-ashing in the fish samples. Generally, the samples were ashed under 400 °C for dry-ashing while the samples were heated using high pressure decomposition vessels for wet-ashing method with HNO_3 . The study of Yang *et al.* (2013) revealed that microwave digestion procedure was easier and more efficient with less contamination than wet-ashing and dry-ashing.

It is important to evaluate different digestion methods to precisely determine the elemental concentrations in water. Chen and Ma (2001) compared three aqua regia digestion methods namely, hotplate aqua regia, microwave aqua regia and microwave aqua regia with Hydrogen fluorid, HF using certified reference materials. The study revealed that both hotplate and microwave aqua regia methods demonstrated satisfactory performance on recovery of Al, Ca, Fe, K, Mg, P, Ba, Cd, Cr, Cu, Mn, Ni, Pb and Zn. However, analyses of As using microwave digestion method and analyses of Se using both hotplate and microwave method attained poor recovery. Similarly, Hseu (2004) evaluated the efficiency of four digestion methods: nitric acid, dry ashing, nitric–perchloric acid and sulfuric acid. The results indicated that nitric acid procedure was the most efficient for recovery of Cd, Mn and Ni from most samples while sulfuric acid appeared to have the lowest recovery for Pb. Nitric–perchloric acid procedure was not recommended because perchloric acid is potentially hazardous during digestion and its recovery performance is relatively poor.

Several studies revealed that the analytical technique to quantify metal ions in samples has been atomic spectroscopy, or more specifically, atomic absorption spectrometry (AAS), atomic emission spectrometry (ICP-AES), inductively coupled plasma mass spectrometry (ICP-MS) (Perez, *et al.*, 2001; Ling *et al.*, 2012; Bashir *et al.*, 2013; Renieri *et al.*, 2014; Sim *et al.*, 2015).

CHAPTER 3

MATERIALS AND METHODS

3.1 Study area

The Baleh River is a tributary of Rajang River. A total of five samplings were carried out at the Baleh River in February, April, June, August and November of 2015. Water, sediment and fish samples were collected from 50 stations along the main river and tributaries of the Baleh River from upstream, downstream and mid-stream at the Mengiong River. The sampling stations are indicated in Figure 3.1 and the corresponding GPS coordinates are summarized in Table 3.1. For fish samples, a total of 22 species were caught.

Table 3.1: The sampling dates and GPS coordinates of the sampling stations

Stations	GPS coordinates	Locations	Dates
1	N02°01'42.9" E113°08'47.2"	Selau River	13/08/2015
2	N02°01'36.2"E113°09'31.2"	Baleh River	13/08/2015
3	N02°01'30.7" E113°09'51.0"	Mouth of Mujong River	13/08/2015
4	N02°01'37.5" E113°10'39.2"	Mujong River	13/08/2015
5	N02°01'39.5"E113°10'54.9"	Mujong River	13/08/2015
6	N02°00'43.6" E113°11'37.2"	Baleh River	13/08/2015
7	N01°59'47.1" E113°12'59.4"	Baleh River	13/08/2015
8	N01°59'28.9" E113°14'24.0"	Mouth of Sebatah River	13/08/2015
9	N01°58'39.1"E113°14'55.2"	Mouth of Won River	13/08/2015
10	N01°57'59.3" E113°17'06.7"	Mouth of Selaut River	12/08/2015
11	N01°56'56.2" E113°19'44.2"	Baleh River	12/08/2015
12	N01°55'39.5" E113°22'45.8"	Baleh River	12/08/2015
13	N01°53'28.2"E113°25'21.6"	1 km below Gaat River	12/08/2015
14	N01°52'30.5"E113°26'08.6"	Gaat River	12/02/2015
15	N01°51'19.7"E113°33'23.1"	1 km below Merirai River	12/02/2015
16	N01°51'36.1"E113°34'39.1"	Merirai River	12/02/2015
17	N01°49'34.2"E113°39'48.6"	Mouth of Entawau River	12/02/2015
18	N01°48'45.4"E113°46'14.2"	Putai River	12/02/2015
19	N01°48'22.9"E113°46'20.6"	Proposed dam site	12/02/2015
20	N01°37'55.4"E113°38'05.0"	Tamawai Atan	12/02/2015
21	N01°37'51.8"E113°40'07.4"	Jugau River	12/02/2015
22	N01°36'12.2"E113°44'40.6"	Mouth of Entuloh River	12/02/2015
23	N01°35'28.8"E113°47'23.1"	1 km below Melatai River	04/04/2015

Table 3.1: Continued

24	N01°35'21.9"E113°47'44.7"	Melatai River	04/04/2015
25	N01°34'01.4"E113°55'38.3"	1 km below Laie River	04/04/2015
26	N01°34'06.3"E113°55'38.3"	Laie River	04/04/2015
27	N01°34'16.5"E114°08'40.7"	Seranai River	05/04/2015
28	N01°33'53.4"E114°12'37.8"	1 km below Kupet River	05/04/2015
29	N01°33'54.2"E114°12'53.0"	Kupet River	05/04/2015
30	N01°34'05"N E114°15'16.7"	Ukit River	22/11/2015
31	N01°34'07.7" E114°15'26.7"	Baleh River	22/11/2015
32	N01°34'01.1" E114°15'47.2"	Kian River	22/11/2015
33	N01°34'04.3" E114°16'45.8"	Tor River	22/11/2015
34	N01°33'55.2" E114°18'38.9"	Irak River	22/11/2015
35	N01°34'02.0" E114°20'47.2"	Penganen River	22/11/2015
36	N01°34'05.6" E114°21'14.0"	Selentang River	06/04/2015
37	N01°34'27.3"E114°24'01.3"	5 km below Long Singut long house	06/04/2015
38	N01°34'41.2"E114°28'03.0"	Baleh River	07/04/2015
39	N01°34'54.0" E113°22'31.4"	Sekawi River	04/04/2015
40	N01°34'23.0" E113°20'40.2"	Entemu River	04/04/2015
41	N01°33'29.9" E113°18'54.4"	Mengiong River below Lengkuas River	13/06/2015
42	N01°32'07.1" E113°18'00.2"	Mengiong River	13/06/2015
43	N01°31'59.3" E113°17'57.8"	Mengiong River below Seringin River	13/06/2015
44	N01°31'48.6" E113°17'17.2"	Mengiong River	13/06/2015
45	N01°31'40.4" E113°17'24.0"	Uga River	12/06/2016
46	N01°30'52.2" E113°14'57.7"	Meranu River	12/06/2016
47	N01°30'41.0" E113°14'37.3"	Mengiong River below Entalawang River	12/06/2016
48	N01°30'15.9" E113°13'53.9"	Entalawang River	12/06/2016
49	N01°30'29.6" E113°13'35.0"	Mengiong River	12/06/2016
50	N01°30'49.7" E113°12'38.9"	Mengiong River upstream	12/06/2016

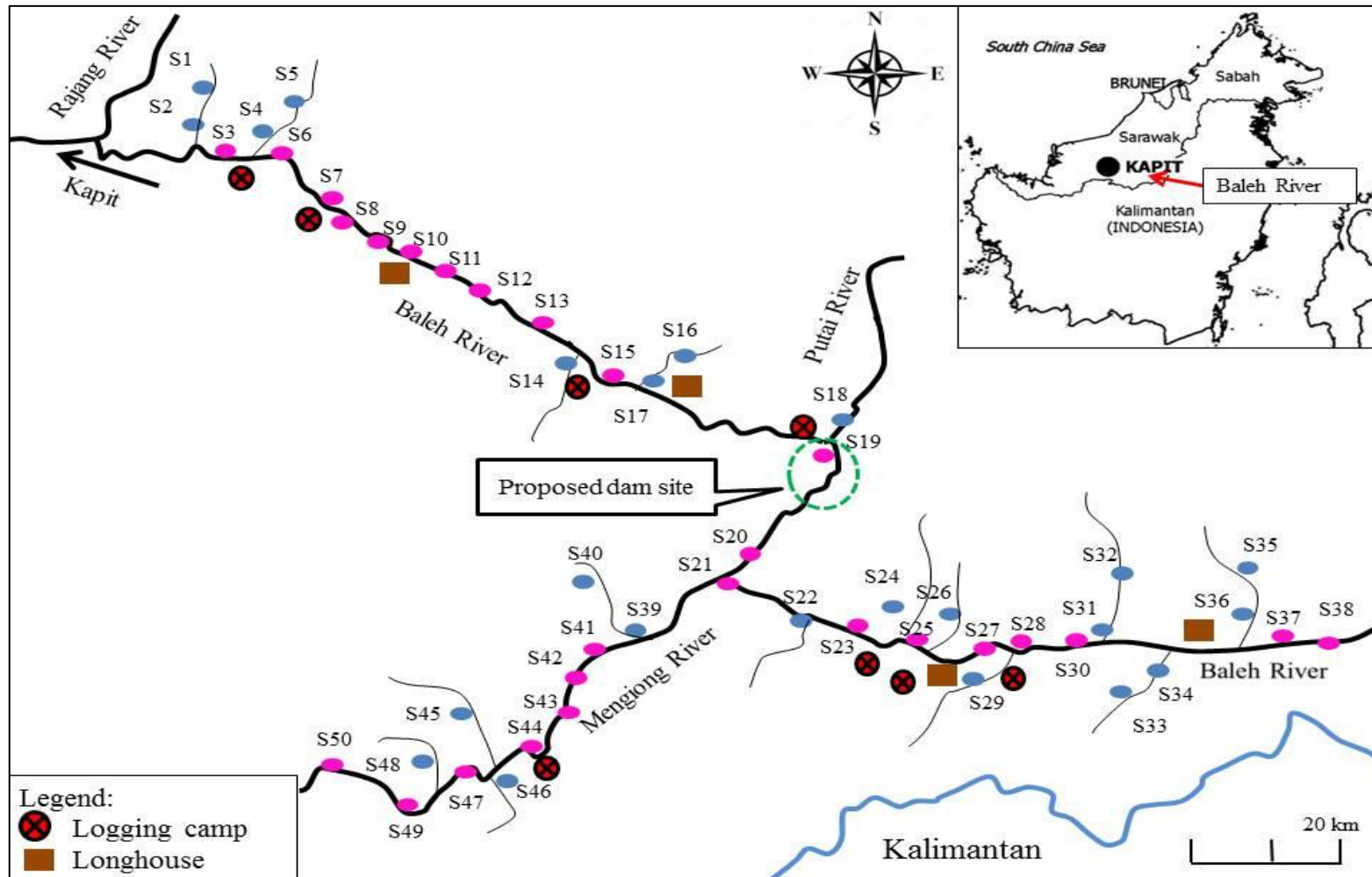


Figure 3.1: Pink markers indicate sampling stations at the main river whilst blue markers indicate tributaries of Baleh River

3.2 Sample collection

1 L of water samples were collected from sub-surface of 5- 30 cm and acidified with 5 mL of 2 M HNO₃. Submerged sediments, at least 30 cm below the water, were obtained by a grab sampler and stored in plastic bags. Fish samples were caught using the netting method, which is three layers net and gill net. The corresponding total length of fishes is recorded in Table 3.2. The samples were kept in a cooler box with ice. Upon arrival at the laboratory, the samples were transferred to the cold room at –20 °C until further analysis.

Table 3.2: Fish species identified and the range of total length

No	Species	Local/ common name	Range of total length (cm)
1.	<i>Oxygaster anomalura</i>	Lalang/ Seluang	17.0-22.5
2.	<i>Luciosoma setigerum</i>	Wader	25.3-26.4
3.	<i>Leiocassis micropogon</i>	Tugok	13.5-21.7
4.	<i>Barbonymus collingwoodii</i>	Kepiat	11.7-14.7
5.	<i>Hemibagrus wyckii</i>	Baung/ Kurai	35.0-36.6
6.	<i>Hampala bimaculata</i>	Adong	18.2-23.0
7.	<i>Hemibagrus planiceps</i>	Baung	24.0-26.0
8.	<i>Macrognathus maculatus</i>	Tilan kapar/ Tilan	30.2-31.3
9.	<i>Barbonymus schwanenfeldii</i>	Tengadak	19.0-28.8
10.	<i>Anematichtys armatus</i>	Buin/ lawak	18.0-23.0
11.	<i>Tor tambroides</i>	Empurau	20.5-25.7
12.	<i>Lobocheilos bo</i>	Kulong	18.0-28.0
13.	<i>Tor dourenensis</i>	Semah	14.0-26.0
14.	<i>Cyclocheilichthys apogon</i>	Emperas/Engkaras	16.0-18.5
15.	<i>Osteochilus schlegelii</i>	Kebali	29.4-30.5
16.	<i>Osteochilus vittatus</i>	Bantak	18.0-24.5
17.	<i>Pangasius macronema</i>	Langu/ Riu	33.0-37.2
18.	<i>Luciosoma spilopleura</i>	Dalang/ Penyuar	18.0-24.5
19.	<i>Labiobarbus fasciatus</i>	Masik	23.7-27.0
20.	<i>Pseudolais micronemus</i>	Shortbarbel pangasius	23.5-36.2
21.	<i>Rasbora borneensis</i>	Seluang buluh	9.0-14.5
22.	<i>Channa lucius</i>	Runtuk/ Gabus cina	13.5-19.5

3.3 Sample preparation and digestion

3.3.1 Water samples

Water samples were digested based on Standard Method of American Public Health Association (APHA) (1998). The acidified water samples were filtered through 0.45 μm membrane filters. Approximately 5 mL of concentrated HNO_3 was added to 100 mL of sample and it was then heated to slow boil on a hotplate until the final volume was approximately 10-20 mL. The samples were left to cool to room temperature and then filtered through 0.45 μm membrane filters. The filtrate was diluted to 100 mL and stored in refrigerator with 4°C prior to analysis. The metal concentration determined with this method is referred to as the acid extractable metal contents consisting of dissolved and weakly sorbed particles.

3.3.1.1 Dissolved metals and total metals method

Water samples from 10 stations downstream, S2, S5, S13, S14, S15, S16, S17, S18, S19 and S20, were also analyzed for their dissolved and total metal contents. Water samples for dissolved metals analysis were filtered through 0.45 μm of filter paper prior to addition of acid for preservation. On the other hand, samples of total metals were preserved immediately by adding acid and proceed without filtration (Zarazua *et al.*, 2006; Kim & Steele, 2004). Approximately 5 mL of concentrated HNO_3 was added to 100 mL of both dissolved metals and total metals samples. The samples were then heated to slow boil on a hotplate until the final volume was approximately 10-20 mL. The samples were filtered through 0.45 μm membrane filters after cool to room temperature. The filtrate was diluted to 100 mL and stored in refrigerator with 4°C prior to analysis.

3.3.2 Sediment samples

Sediment samples were air-dried, sieved through a 0.5 mm sieve, homogenized and ground to fine powder using ceramic mortar and pestle. The 0.5 mm sieve was chosen because it comprises all primary soil particles (USDA, 2014). A total 0.50 g of sediment samples were digested with 6 mL concentrated HNO₃ and 2 mL HCl. The samples were digested using a microwave digester (CEM-MARS 6; USA) at 180 °C for 40 min, and cool cold down for 20 min. The digested samples were filtered through 0.45 µm membrane filters and diluted to 100 mL. The samples were kept in a refrigerator with 4 °C prior to analysis.

3.3.3 Fish samples

Fish samples were washed under tap water to remove dirt and dissected according to muscle, belly and gills using a ceramic knife after took out from the cold room. The samples were oven-dried at 105 °C for approximately 2 hours and ground homogenize by using ceramic mortar and pestle. A total 0.25 g of fish samples were digested in 6 mL HNO₃ and 1 mL HCl using the microwave-digester at 180 °C for 40 min, and cool cold down for 20 minutes. The digested samples were filtered through 0.45 µm membrane filters and diluted to 100 mL. The filtrate samples were also kept in a refrigerator with 4 °C prior to analysis.

3.4 Samples analysis

All samples were digested in triplicates and subjected to metal analysis including Al, Cr, Cu, Mn, Ni, Fe, Zn, As and Se using a Microwave Plasma Atomic Emission Spectrometer (Agilent MP-AES 4200; USA) whilst Hg was analyzed using a mercury analyzer (Perkin Elmer FIMS 400; USA). The limit of detection (LoD) and limit of quantification (LoQ) were calculated as the concentrations three times and ten times the standard deviation of the

background noise recorded on 20 measurements of the procedural blank, respectively (Djedjibegovic *et al.*, 2012). Detection limits of MP-AES were Al (0.03 ppm); Cr (0.01 ppm); Cu (0.01 ppm); Mn (0.002 ppm); Ni (0.004 ppm); Pb (0.02 ppm); Zn (0.04ppm); Fe (0.01 ppm); As (0.5 ppm); Se (0.2 ppm). Meanwhile, the detection limit of mercury analyzer was 0.5 ppb. Any metals concentration below the detection limit, are indicated < LOD.

3.5 Assessment of the contamination

The contamination status of sediment was evaluated based on the geo-accumulation index (I_{geo}), contamination factor (CF) and pollution load index (PLI). Geo-accumulation index, I_{geo} (Muller, 1969) is used to illustrate the enrichment of metal concentration above the baseline concentrations as shown in equation 1. The I_{geo} scale consists of seven grades (0-6) ranging from unpolluted to highly pollute as shown in Table 3.3.

$$I_{geo} = \log_2 \frac{C_{sample}}{1.5 \times C_{background}} \quad [1]$$

where C_{sample} = concentration of the element in a sample
 $C_{background}$ = baseline value

Table 3.3: The classification of sediment quality based on I_{geo} value

I_{geo} value	Class	Sediment quality
≤ 0	0	Unpolluted
0-1	1	Unpolluted to moderately polluted
1-2	2	Moderately polluted
2-3	3	Moderately to strong polluted
3-4	4	Strong polluted
4-5	5	Strongly to extremely polluted
> 6	6	Extremely polluted

The formula of contamination factor (CF) is expressed in equation 2, where sediment with $CF < 1$ is classified as low in contamination. Sediments with $1 \leq CF < 3$ are moderately

contaminated and $3 \leq CF < 6$ is considerably contaminated (Thomlinson *et al.*, 1980). The pollution load index (PLI) is calculated based on equation 3. The PLI value varies from 0 (unpolluted) to 1 (highly polluted).

$$CF = \frac{C_{sample}}{C_{background}} \quad [2]$$

where C_{sample} = concentration of the element in a sample
 $C_{background}$ = baseline value

$$PLI = (CF_1 \times CF_2 \times CF_3 \times \dots CF_n)^{1/n} \quad [3]$$

where CF= contamination factor
n= number of metals

The geochemical baseline, $C_{background}$ values were determined based on the linear regression of the cumulative distribution frequency (Wei & Wen, 2012; Teng *et al.*, 2009). The data (element concentrations) was arranged in the ascending order and each frequency was added to the sum of its predecessors. Then, in order to calculate the cumulative frequency, each added frequency was divided with the total value of the data sets where the ultimate cumulative frequency is equal to 1. Cumulative frequency curve was plotted with x-axis representing the element concentrations (arranged in ascending order) whilst y-axis representing the cumulative frequency. The regression line with $P < 0.05$ and $R^2 > 0.9$ was modeled using Microsoft Excel 2010. If the criteria ($P < 0.05$ and $R^2 > 0.9$) are not met, the maximum value was removed and the linearity as well as R^2 was recalculated. This procedure was repeated until the above two criteria were met (Wei & Wen, 2012). The outlier was attributed to input from anthropogenic activities whilst the value which can fit into the regression line suggests natural existence where the highest value of the regression fit is considered the baseline level. Curves with one inflexion point indicate minimal anthropogenic input, representing the natural concentrations (Raimann *et al.*, 2005). A second inflexion point

would suggest possible anthropogenic contribution where samples found between the first and second may or may not be affected by the disturbance (Matschullat *et al.*, 2000).

3.6 Determination of moisture content

In the present study, the metals concentrations in fishes were reported in dry weight basis for consistency. The guideline concentrations reported by Food and Agriculture Organization (FAO) (1983) and Malaysia Food Act (MFA) (1983) are in wet weight. Therefore, the metals concentrations identified in fishes which include Cr, Cu, Fe, Mn, Ni, Al, Zn, Pb, As, Se and Hg were converted to wet weight based on the moisture content determined according to loss-on-drying. The weights of fish before and after oven-dried, in 105 °C were recorded and the moisture content was calculated based on the equation 4.

$$\text{Moisture content (\%)} = \frac{W_{\text{before}} - W_{\text{after}}}{W_{\text{before}}} \times 100\% \quad [4]$$

Where W_{before} = weight before dry
 W_{after} = weight after dry

3.7 Health risk assessment

The hazard risk of Hg is further evaluated based on the Hazard Index (HI), developed by the United States Environmental Protection Agency (US EPA) based on the following equation 5 and 6 (Singh *et al.*, 2014; Sim *et al.*, 2016). The reference dose (RfD) for total Hg is 0.0003 mg/kg body weight per day, whilst the bioavailability factor was assumed to be 1 in order to avoid the underestimating of the health hazard indices, defined by USEPA (1997).

$$\text{HI} = \frac{ED}{RfD} \quad [5]$$

where ED = estimated daily dose (mg/kg/BW/day)
 RfD = reference dose (mg/kg/BW/day)

$$ED = \frac{C_{sample} \times IR \times BF}{BW} \quad [6]$$

Where C_{sample} = concentration of elements in samples (mg/kg)

IR = fish consumption rate (g/ day)

BF = bioavailability factor

BW = body weight (kg)

The fish consumption rate and body weight are assumed based on the guideline of US EPA (Health Canadian, 2007) and the guideline recommended by Azmi *et al.* (2009) (Table 3.4) was used to provide more representative calculation for the population in Malaysia. Hazard index (HI) below one indicates no health risk and HI above 1 and 10 suggests moderate and high hazard respectively. Exposure ratio (ER) and consumption limits (CR, g of fish/week) were calculated to suggest the consumption rate/week based on the equation 7 and 8. The provisional tolerable daily intake (pTDI) of Hg proposed by Health Canada (2007) for children is 0.2 µg/kg/BW/day, while 0.47 µg/kg for adult women and men.

$$ER = \frac{ED}{pTDI} \quad [7]$$

where ER = exposure ratio

ED = estimate daily dose (mg/kg/BW/day)

pTDI= provisional tolerable daily intake (µg/kg/BW/day)

$$CR = \frac{pTDI \times BW \times 7(day/week)}{C_{Hg} (\mu g/g)} \quad [8]$$

where CR = consumption limits (g/week)

pTDI= provisional tolerable daily intake (µg/kg/BW/day)

BW = body weight (kg)

C_{Hg} = Total Hg concentration in fish (µg/g)

Table 3.4: Fish consumption rate and body weight of different population groups

Country	Population	Body weight (kg)	Fish consumption rate (kg/day)
Canada	1-4 years old (children)	14.4 ¹	0.02
	5-11 years old(children)	26.4 ¹	0.02
	Women (adult)	65.0 ¹	0.08
	Men (adult)	73.0 ¹	0.08
Malaysia	18-59 years old women (adult)	58.44 ²	0.16
	18-59 years old men (adult)	66.56 ²	0.16

Health Canada (2007)¹

Azmi *et al.* (2009)²

3.8 Statistical analyses

The statistical analyses were conducted using the IBM SPSS statistic 20, Matlab R2012b, and Microsoft Excel 2010. Descriptive statistics including the mean, maximum, minimum, standard deviation, and coefficient of variance were calculated. The metal contents were subjected to Principal Component Analysis (PCA) to reveal the clustering pattern of the data according to sampling locations and fish species. Prior to PCA, the data was transformed using square-root function and standardized by subtracting the samples mean from each data point, and dividing by the standard deviation. This was done to ensure that all variables are comparable. Pearson's correlation analysis was performed to identify the correlation between two elements where performed based on 95 % significance level. One way ANOVA was used to examine the significant difference in means of different sample populations and for the comparison between dissolved, acid-extractable and total dissolved metal contents. Meanwhile Tukey's Test was applied for multiple comparisons. The baseline concentrations were derived from linear regression fit of the cumulative distribution frequency. As suggested by Wei and Wen (2012), the linear regression of the cumulative frequency curve, R^2 , must be greater than 0.90 with linearity of $P < 0.05$.

3.9 Quality assurance and quality control

All samples were digested in triplicates and the analytical measurements were taken in triplicates to ensure reliable and representative results. Blank samples were analyzed for potential contamination. All glassware was soaked in 10 % HNO₃ overnight before use. The method was validated using the Certified Reference Materials (CRM) of stream sediment, STSD-1 for sediment whilst ERM BB-422 for fish.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Heavy metals in water and sediment

4.1.1 Method validation

The recovery percentages of Cu, Fe, Mn, Ni and Zn in sediment of CRM (stream sediment, STSD-1) are summarized in Table 4.1. For Al, spiked method was used attaining a percentage of 108.69 %. Other elements demonstrated recovery percentages between 96 and 128 %, falling within the acceptable range of 75–125 % (Cussen & Hensman, 2011) with Ni showing slightly higher degree of uncertainty.

Table 4.1: Mean metals concentration recovered and the recovery percentages in sediment

Elements	Expected value (mg/kg)	Observed value (mg/kg)	Recovery (%)
Al*	2000	2173.93±119.94	108.69±6.00
As	17	15.25±1.64	89.73±9.65
Cu	36	35.13±0.73	96.32±2.04
Fe	35000	38674.37±604.39	110.47±1.73
Mn	3740	3714.87±119.94	99.32±0.68
Ni	18	23.17±1.13	125.10±5.92
Zn	165	173.83±1.54	104.70±1.37
Hg	0.11	0.13±0.04	118.55±35.39

* spiked method

4.1.2 Distribution of metals in water and sediment

The mean concentrations of heavy metals in water and sediment are summarized in Table 4.2 while the significant difference across stations are shown in Appendix A and B. Heavy metals in water and sediment were examined independently using PCA to demonstrate the clustering pattern. The scores plots of PC2 vs PC1 are shown in Figure 4.1. For water, samples from the Mengiong River S43, S44, S45, S47, S48, S49 and S50 were seen to occupy the right quadrant of the scores plot whilst S40, S41, S42 and S46 formed another cluster, evidently distinguishable from other stations. Meanwhile, S31 stands alone at the upper right quadrant

whilst S23, S24, S25 and S27 clustered. The downstream of Baleh River from S1 to S12 and tributaries from S30 to S36 on the other hand occupy the left quadrant. For sediment, S3, S4, S6, S7, S8, S9, S10, S11, S12 and S13 appear to occupy the right quadrant forming a cluster distinguishable from other stations. Statistically, significant differences between stations are confirmed for water and sediments ($P < 0.05$).

Table 4.2: The mean concentrations of metals in (a) water and (b) sediment across 50 stations

(a)

Stations	Elements (mg/L)										
	Al	Cr	Cu	Mn	Ni	Pb	Zn	Fe	As	Se	Hg
1	0.33±0.03	<LOD	<LOD	0.13±0.00	<LOD	0.01±0.00	<LOD	0.02±0.03	<LOD	<LOD	<LOD
2	0.07±0.02	<LOD	0.01±0.00	0.03±0.00	<LOD	0.03±0.01	0.03±0.01	0.27±0.03	<LOD	<LOD	<LOD
3	0.39±0.04	<LOD	<LOD	0.12±0.01	<LOD	0.01±0.01	<LOD	0.38±0.32	<LOD	<LOD	<LOD
4	0.48±0.05	<LOD	<LOD	0.15±0.00	<LOD	0.01±0.00	<LOD	0.24±0.19	<LOD	<LOD	<LOD
5	0±0.00	<LOD	0.01±0.00	<LOD	<LOD	0.03±0.01	0.02±0.00	0.06±0.00	<LOD	<LOD	<LOD
6	0.16±0.02	<LOD	<LOD	0.06±0.00	<LOD	<LOD	<LOD	0.05±0.04	<LOD	<LOD	<LOD
7	0.10±0	<LOD	<LOD	0.05±0.00	<LOD	0.01±0.00	<LOD	0.00±0.00	<LOD	<LOD	<LOD
8	0.12±0	<LOD	<LOD	0.06±0.00	<LOD	<LOD	<LOD	0.03±0.02	<LOD	<LOD	<LOD
9	0.10±0.01	<LOD	<LOD	0.05±0.00	<LOD	<LOD	<LOD	0.03±0.04	<LOD	<LOD	<LOD
10	0.12±0	<LOD	<LOD	0.05±0.01	<LOD	<LOD	<LOD	0.08±0.10	<LOD	<LOD	<LOD
11	0.08±0.06	<LOD	<LOD	0.05±0.00	<LOD	<LOD	<LOD	0.15±0.18	<LOD	<LOD	<LOD
12	0.11±0.01	<LOD	<LOD	0.06±0.00	<LOD	<LOD	<LOD	0.13±0.17	<LOD	<LOD	<LOD
13	0.03±0.00	<LOD	0.01±0.00	0.03±0.01	<LOD	0.02±0.01	0.02±0.01	0.19±0.03	<LOD	<LOD	<LOD
14	0.26±0.00	<LOD	0.01±0.00	<LOD	<LOD	0.03±0.01	0.01±0.01	0.06±0.00	<LOD	<LOD	<LOD
15	0.10±0.00	<LOD	0.01±0.00	0.04±0.00	<LOD	0.02±0.01	0.01±0.01	0.25±0.01	<LOD	<LOD	<LOD
16	0.2±0.00	<LOD	0.01±0.00	0.14±0.01	<LOD	0.02±0.00	0.01±0.01	2.00±2.23	<LOD	<LOD	<LOD
17	0.39±0.04	<LOD	<LOD	0.1±0.00	<LOD	0.03±0.00	0.03±0.01	0.53±0.01	<LOD	<LOD	<LOD
18	0.10±0.00	<LOD	<LOD	0.02±0.00	0.01±0.01	0.03±0.00	0.04±0.01	0.17±0.00	<LOD	<LOD	<LOD
19	0±0.01	<LOD	<LOD	0.08±0.00	<LOD	0.03±0.00	0.04±0.01	0.45±0.01	<LOD	<LOD	<LOD
20	0.11±0.07	<LOD	<LOD	0.02±0.00	<LOD	0.03±0.00	0.02±0.00	0.13±0.01	<LOD	<LOD	<LOD
21	0.04±0.02	<LOD	<LOD	0.07±0.00	<LOD	0.03±0.00	0.03±0.00	0.35±0.00	<LOD	<LOD	<LOD
22	0.06±0.00	<LOD	0.01±0.00	0.02±0.00	0.01±0.00	0.03±0.00	0.05±0.02	0.19±0.03	<LOD	<LOD	<LOD
23	1.85±0.21	<LOD	0.01±0.00	0.48±0.05	0.01±0.00	0.02±0.00	0.03±0.01	1.51±0.18	<LOD	<LOD	<LOD
24	1.06±0.06	<LOD	0.01±0.00	0.45±0.03	<LOD	0.01±0.00	0.02±0.00	1.48±0.06	<LOD	<LOD	<LOD

Table 4.2(a): Continued

25	0.07±0.00	<LOD	<LOD	0.03±0.00	<LOD	<LOD	0.02±0.01	0.19±0.00	<LOD	<LOD	<LOD
26	0.76±0.01	<LOD	0.01±0.00	0.21±0.01	0.02	0.01±0.00	0.02±0.00	1.02±0.02	<LOD	<LOD	<LOD
27	0.18±0.01	<LOD	<LOD	0.05±0.00	0.01±0.00	<LOD	0.01±0.00	0.34±0.03	<LOD	<LOD	<LOD
28	0.11±0.00	<LOD	<LOD	0.03±0.00	<LOD	0.01±0.00	0.01±0.00	0.24±0.00	<LOD	<LOD	<LOD
29	0.26±0.00	<LOD	<LOD	0.15±0.00	<LOD	<LOD	<LOD	0.60±0.03	<LOD	<LOD	<LOD
30	6.11±0.19	<LOD	<LOD	1.74±0.04	<LOD	<LOD	<LOD	6.00±0.17	<LOD	<LOD	<LOD
31	0±0	<LOD	<LOD	0.02±0.00	<LOD	<LOD	<LOD	0.08±0.00	<LOD	<LOD	<LOD
32	0.03±0	<LOD	<LOD	0.02±0.00	<LOD	<LOD	<LOD	0.23±0.02	<LOD	<LOD	<LOD
33	0.03±0.02	<LOD	<LOD	0.02±0.01	<LOD	<LOD	<LOD	0.12±0.04	<LOD	<LOD	<LOD
34	0.06±0.01	<LOD	<LOD	0.02±0.00	<LOD	<LOD	<LOD	0.13±0.00	<LOD	<LOD	<LOD
35	0.06±0.00	<LOD	<LOD	0.03±0.00	<LOD	<LOD	<LOD	0.24±0.00	<LOD	<LOD	<LOD
36	0.15±0.01	<LOD	<LOD	0.05±0.00	<LOD	0.01±0.00	<LOD	0.28±0.00	<LOD	<LOD	<LOD
37	0.11±0.01	<LOD	<LOD	0.03±0.00	<LOD	0.01±0.00	<LOD	0.18±0.00	<LOD	<LOD	<LOD
38	0±0.00	<LOD	0.01±0.00	0.02±0.00	<LOD	<LOD	<LOD	0.15±0.01	<LOD	<LOD	<LOD
39	0.18±0.01	<LOD	0.01±0.00	0.10±0.00	<LOD	<LOD	<LOD	0.44±0.02	<LOD	<LOD	<LOD
40	0.23±0.01	<LOD	0.01±0.00	0.06±0.00	<LOD	0.01±0.00	<LOD	0.44±0.01	<LOD	<LOD	<LOD
41	0.09±0.01	<LOD	0.01±0.00	0.05±0.00	<LOD	<LOD	<LOD	0.31±0.02	<LOD	<LOD	<LOD
42	0.77±0.09	<LOD	0.01±0.00	0.23±0.00	<LOD	0.01±0.00	<LOD	1.18±0.13	<LOD	<LOD	<LOD
43	0.71±0.06	<LOD	0.01±0.00	0.23±0.00	<LOD	0.01±0.00	<LOD	1.02±0.07	<LOD	<LOD	<LOD
44	0.53±0.03	<LOD	0.01±0.00	0.2±0.00	<LOD	0.01±0.00	<LOD	0.66±0.03	<LOD	<LOD	<LOD
45	0.16±0.01	<LOD	0.01±0.00	0.08±0.00	<LOD	<LOD	<LOD	0.36±0.05	<LOD	<LOD	<LOD
46	0.68±0.02	<LOD	0.01±0.00	0.24±0.01	<LOD	0.01±0.00	<LOD	0.95±0.05	<LOD	<LOD	<LOD
47	0.84±0	<LOD	0.01±0.00	0.22±0.00	<LOD	0.01±0.00	<LOD	0.91±0.01	<LOD	<LOD	<LOD
48	0.72±0.04	<LOD	0.01±0.00	0.29±0.01	0.01±0.00	0.01±0.00	<LOD	1.16±0.05	<LOD	<LOD	<LOD
49	0.72±0.02	<LOD	0.01±0.00	0.245±0.00	0.01±0.00	<LOD	<LOD	1.20±0.06	<LOD	<LOD	<LOD
50	0.33±0.03	<LOD	<LOD	0.13±0.00	<LOD	0.01±0.00	<LOD	0.02±0.03	<LOD	<LOD	<LOD

<LOD: below detection limit

Table 4.2(b): Continued

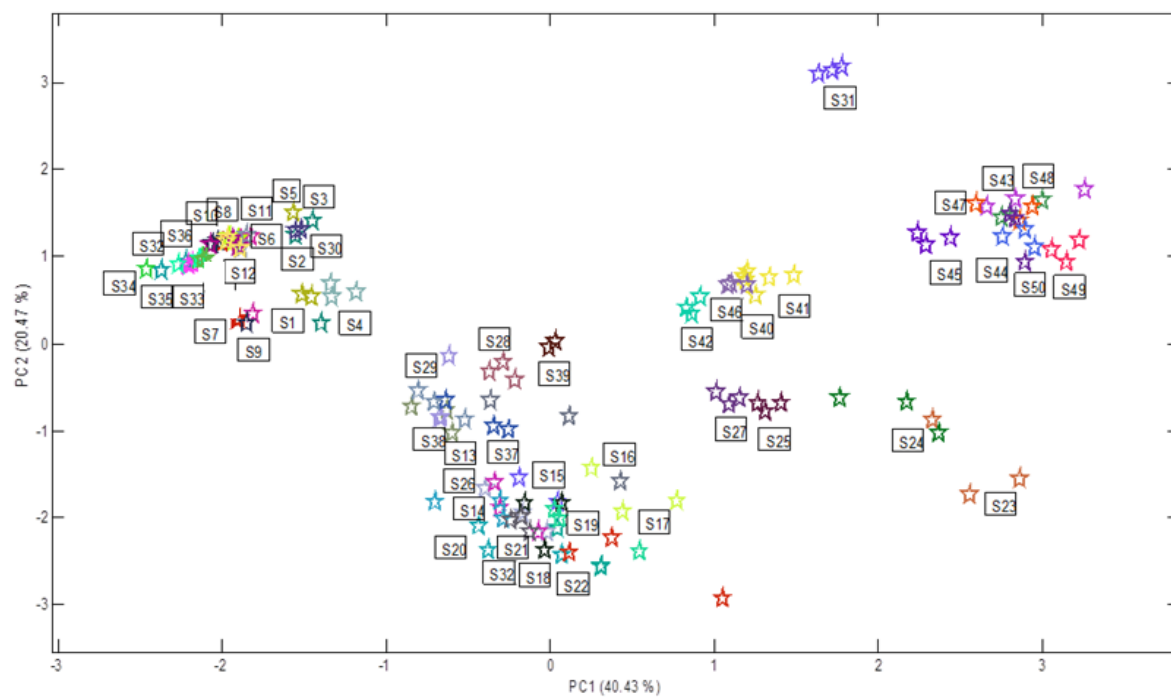
Stations	Elements (mg/kg)								
	Al	Cr	Cu	Mn	Ni	Pb	Zn	Fe	Hg
1	72670.20±1187.44	42.38±1.41	23.61±0.93	694.12±16.31	<LOD	16.87±0.90	52.28±1.58	43324.17±814.81	0.11±0.02
2	38854.88±5170.26	25.32±3.45	15.01±0.09	354.33±4.23	<LOD	15.84±0.46	43.05±1.89	21983.52±811.27	0.07±0.04
3	91135.25±3140.82	55.61±1.81	29.22±1.02	99.00±2.95	<LOD	29.64±0.94	42.97±1.95	50207.60±1082.04	0.07±0.00
4	70219.25±1968.56	39.26±2.64	22.63±0.16	638.20±8.81	<LOD	14.44±0.25	57.20±1.15	41448.62±883.25	0.10±0.01
5	38924.46±6547.16	31.67±2.05	14.45±0.74	253.07±18.54	<LOD	11.51±0.85	43.06±1.01	23018.48±395.51	0.10±0.04
6	50850.64±3903.13	34.54±2.05	18.56±0.46	466.73±11.49	<LOD	12.60±0.45	43.78±1.07	35707.88±1491.55	0.07±0.00
7	56764.01±1777.58	36.89±1.01	18.92±0.87	461.32±23.90	<LOD	14.30±0.24	46.33±1.85	37133.87±1539.27	0.08±0.01
8	62765.21±2266.09	37.74±2.72	21.72±0.74	632.94±24.30	<LOD	16.07±0.15	51.24±1.81	42186.81±820.43	0.09±0.00
9	57898.58±5001.52	36.29±1.63	19.32±0.88	486.95±21.24	<LOD	14.22±0.32	45.12±2.53	38079.50±1476.33	0.07±0.01
10	33081.52±3097.98	28.23±1.65	12.22±0.08	280.58±18.14	<LOD	8.46±0.31	35.02±2.05	27645.42±1178.18	0.05±0.01
11	69125.62±9884.65	47.47±5.80	25.89±0.96	772.57±35.53	<LOD	13.65±0.77	55.53±3.42	44707.41±2136.93	0.11±0.02
12	67839.52±1882.60	40.40±0.35	22.64±0.28	638.27±22.54	<LOD	16.53±0.31	53.03±2.00	41938.12±1603.09	0.11±0.03
13	28064.90±2088.00	19.01±1.52	12.13±0.60	273.51±6.05	<LOD	12.18±0.50	40.58±1.33	20349.81±385.96	0.07±0.05
14	29033.21±3007.60	40.06±1.77	17.39±0.21	331.12±12.45	<LOD	13.43±0.15	53.88±3.20	27488.94±372.83	0.13±0.03
15	21773.43±1391.28	25.18±3.13	14.26±1.32	296.90±28.08	<LOD	14.47±1.21	49.11±4.35	24903.36±2782.95	0.14±0.03
16	38138.18±1328.91	39.62±4.52	16.81±1.00	323.66±37.25	<LOD	12.30±1.31	55.32±7.89	25073.76±1841.28	0.11±0.02
17	42128.85±1209.87	43.29±0.60	20.71±0.68	463.90±14.91	6.74±2.21	18.71±0.98	48.84±3.98	28107.94±1320.35	0.15±0.03
18	39650.48±6322.06	24.02±5.08	11.73±1.02	230.00±14.05	1.39±1.96	13.00±0.47	35.27±1.09	19845.29±1264.72	0.08±0.06
19	45824.90±3683.60	36.2±0.46	17.46±1.47	432.94±80.63	5.12±0.50	19.57±2.65	46.55±4.42	28608.18±2864.37	0.10±0.04
20	34001.13±3683.60	30.74±1.35	15.65±0.92	444.23±37.27	4.87±0.44	22.81±6.24	41.79±3.02	25195.06±2328.93	0.07±0.05
21	37641.96±4398.27	34.11±3.60	15.99±1.09	392.65±8.67	<LOD	17.49±0.68	43.12±1.01	24872.10±1710.15	0.13±0.03
22	45053.51±3223.54	41.01±2.17	15.47±0.43	304.00±13.06	<LOD	15.28±0.38	38.00±2.36	22058.29±791.81	0.15±0.03
23	74628.00±6668.50	61.03±5.58	24.07±2.08	340.19±32.42	11.57±8.18	19.09±1.30	58.04±6.34	29749.39±2933.04	0.17±0.08
24	68684.55±8422.23	53.16±3.72	21.38±0.14	343.46±2.70	<LOD	19.08±0.52	49.37±0.81	27615.77±594.23	0.25±0.05
25	74957.60±10273.21	56.05±1.95	27.16±1.46	668.28±16.11	17.27±0.94	22.10±0.21	56.73±3.17	32525.14±120.00	0.17±0.00

Table 4.2 (b): Continued

26	86421.30±9797.65	47.41±2.73	24.96±0.36	742.60±15.88	12.88±0.23	24.68±0.38	58.96±0.71	33732.10±521.26	0.19±0.02
27	73629.72±2276.57	52.46±14.00	23.18±0.87	511.58±24.30	9.82±6.97	19.86±0.67	53.92±2.71	28074.49±1194.03	0.30±0.06
28	82902.10±4770.65	64.06±5.62	32.14±0.92	716.70±25.26	26.42±1.84	21.03±0.74	68.55±3.64	34566.95±1204.83	0.23±0.01
29	85605.37±7637.54	62.12±9.03	34.1±3.10	737.00±77.10	20.41±14.54	21.92±1.04	67.39±4.33	35368.67±2947.40	0.26±0.06
30	51206.77±1728.25	43.72±1.64	13.62±0.36	290.86±13.00	<LOD	6.79±0.44	37.28±1.48	27483.92±422.07	0.02±0.01
31	54348.80±3808.49	54.20±3.04	28.12±1.43	546.52±50.40	<LOD	8.64±0.66	52.96±0.39	42429.04±2871.82	0.02±0.00
32	56733.22±2384.64	47.59±0.88	23.00±0.60	522.60±50.20	<LOD	8.97±0.74	52.63±1.06	38566.32±1867.92	0.03±0.00
33	50631.42±2623.37	43.38±0.55	14.51±0.31	395.28±83.37	<LOD	6.03±0.87	37.59±0.40	29782.27±693.88	0.01±0.00
34	47404.15±3041.17	40.28±2.10	15.3±0.54	398.14±41.61	<LOD	6.92±0.17	41.51±0.68	31432.73±579.14	0.01±0.00
35	49920.28±1048.68	49.79±2.66	22.43±0.50	448.31±8.15	<LOD	7.39±0.47	48.34±0.86	39948.61±819.31	0.01±0.00
36	52854.22±2291.71	53.40±1.03	22.21±0.46	527.10±44.59	<LOD	8.09±1.07	44.51±0.81	38824.51±2004.85	0.03±0.00
37	81530.14±8349.58	65.27±4.40	28.81±0.34	453.59±19.46	19.94±1.06	20.97±0.40	56.11±1.85	32184.64±1388.45	0.39±0.27
38	85413.15±11675.91	65.86±6.12	36.44±1.76	697.17±23.77	23.48±2.43	19.55±0.84	63.39±4.10	35094.83±2257.68	0.20±0.02
39	61334.41±1534.90	40.66±0.56	23.06±0.26	533.34±28.24	15.99±0.70	19.86±0.28	54.78±1.24	35216.12±1667.21	<LOD
40	62320.59±1808.97	46.48±1.96	21.57±0.32	489.11±6.44	15.64±0.56	18.52±0.35	40.37±0.44	32742.21±206.03	<LOD
41	49071.05±1503.60	34.30±1.27	16.79±0.55	258.63±10.59	11.65±0.90	15.77±0.47	45.40±1.19	28283.65±1091.92	<LOD
42	51024.34±2173.53	35.89±1.78	11.89±0.32	94.13±1.42	7.54±0.57	15.76±0.22	36.50±1.15	26153.83±106.53	<LOD
43	48801.52±3630.62	33.64±12.14	16.31±0.18	335.91±8.13	10.80±0.56	15.99±0.49	36.72±0.25	27613.73±296.60	<LOD
44	56516.98±2023.72	39.74±0.30	19.38±0.14	374.08±8.62	14.49±1.02	18.90±0.24	46.01±0.74	31626.10±520.06	<LOD
45	49685.93±7040.12	35.25±4.08	18.29±0.51	251.88±26.14	14.14±1.03	18.06±0.21	44.14±1.79	22823.12±2032.29	<LOD
46	58362.87±2082.28	43.21±0.68	21.05±0.57	385.95±25.41	15.01±1.24	19.54±1.01	43.65±1.34	30423.63±656.38	<LOD
47	52677.56±840.18	36.28±0.96	18.53±0.16	379.67±13.50	12.14±0.41	17.78±0.14	41.67±0.31	29619.24±433.44	<LOD
48	50722.53±1274.16	35.42±1.37	16.90±0.40	344.82±5.10	11.84±0.24	16.87±0.53	40.59±1.35	27800.53±51.17	<LOD
49	40121.31±1977.25	30.37±1.49	15.67±0.43	365.65±48.64	10.78±0.69	14.09±0.63	39.73±1.05	27789.48±537.27	<LOD
50	43671.07±10553	31.08±1.29	16.44±0.55	300.39±18.72	11.05±1.19	15.16±0.20	44.55±2.10	27338.29±613.10	<LOD

<LOD: below detection limit

(a)



(b)

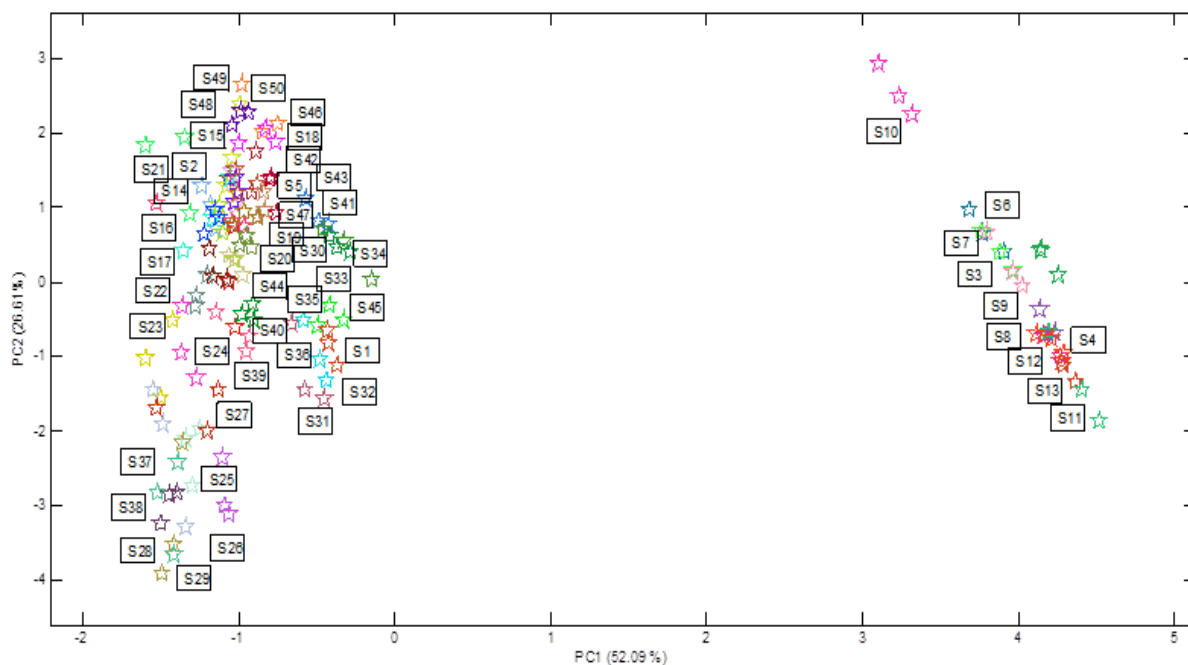


Figure 4.1: The scores plot of PC2 vs PC1 for heavy metals in (a) water (b) sediment

Table 4.3 summarizes the mean metal concentrations found in water and sediment in comparison to various guidelines. Typically, in the present study higher metal concentrations are found in sediment as it is the major depository that holds more than 99 % of the total metals present in the aquatic environment (Davies & Allison, 2006; Abdullah *et al.*, 2007; Ebrahimpour & Mushrifah, 2008; Idriss & Ahmad, 2013). The most abundant element detected in water was Fe with an average of 0.58 mg/L. Fe is commonly found in two states, Fe^{2+} and Fe^{3+} . In rivers, the median concentration of Fe, typically referred to Fe^{2+} , is 0.7 mg/L (WHO, 1996). Under reducing environment, the concentration could increase to 1.0 mg/L as a result of enhanced dissolution of insoluble Fe^{3+} to soluble Fe^{2+} (Colter & Mahler, 2003). Al is the second most abundant element in water, detected at an average of 0.44 mg/L. The average concentration of Al (0.44 mg/L) is higher than the permissible level for drinking water at 0.2 mg/L. The metal concentrations reported in this study refer to acid extractable content that constitutes of dissolved and weakly-sorbed metals on particulates hence is expectedly higher than the drinking water guideline that is often referred to the dissolved metals content. Meanwhile, Pb is detected at 0.01 mg/L, complying with the permissible level for drinking water. According to the National Standard of Drinking Water in Malaysia (MOH, 2000), the level of Pb detected falls within the recommended raw water quality for drinking water (NSDWQ: 0.05 mg/L). Other elements including Cr, Hg, As and Se are undetected whilst Cu, Zn, Ni, Mn and Fe are below the guideline with Fe exhibiting moderately high concentration above 0.5 mg/L. For sediment, the metal concentration is compared to the Canadian Sediment Quality Guidelines (CEQG, 2001). Cu, Ni, Zn and Pb were below the rare effect level, indicative of low risk of adverse effects. The concentration of Mn is slightly higher, categorized below threshold effect level, whilst Fe falls between occasional and probable effect level.

Table 4.3: The mean of metal contents in water and sediment in comparison to various guidelines

Elements	Water (mg/L)	Sediments (mg/kg)	Permissible range of water (mg/L)		Guideline of sediment quality(mg/kg)				
			WHO	NSDWQ	REL	TEL	OEL	PEL	FEL
Al	0.44±0.92	55452.41±16765.53	0.20	0.20	-	-	-	-	-
Cu	0.01±0.00	20.18±5.78	2	1.0	22	36	63	200	700
Fe	0.58±0.93	31573.00±6985.79	1	1	-	20000	30000	40000	-
Mn	0.14±0.26	434.48±164.24	0.4	0.2	-	460	780	1100	-
Ni	0.02±0.00	6.22±7.68	0.07	0.02	-	-	47	-	-
Zn	0.01±0.01	47.65±8.16	3	3	80	120	170	310	770
Pb	0.01±0.01	15.82±4.97	0.01	0.05	-	36	83	130	-
Cr	<LOD	41.82±11.13	0.05	0.05	-	43	76.5	110	-
As	<LOD	<LOD	0.01	0.01	-	9.8	21.4	33	-
Se	<LOD	<LOD	0.01	0.01	-	-	-	-	-
Hg	<LOD	<LOD	0.006	0.001	-	0.18	0.64	1.1	-

WHO: World Health Organisation

NSDWQ: National Standards Drinking Water Quality

REL: rare effect level; TEL: threshold effect level; OEL: occasional effect level; PEL: probable effect level; FEL: frequent effect level

<LOD: below detection limit

The mean metal distributions in water and sediment are shown in Figure 4.2. Apparently, Al is found in the highest concentration in water at S31 (6.11 mg/L), followed by S23 (2.02 mg/L), S24 (1.85 mg/L) and S25 (1.06 mg/L). The concentration of Fe at S31, S25 and S23 are observed in a similar order, corresponding to 1.94 mg/L, 1.51 mg/L and 1.48 mg/L, respectively. Interestingly, the concentration of Mn also demonstrates similar trend with the highest concentration detected at S31 (1.74 mg/L), followed by S23 (0.57 mg/L), S24 (0.48 mg/L) and S25 (0.45 mg/L). The concentrations of Al and Fe are positively correlated in pH 7.2. Al and Fe are the most abundant metals in the Earth's crust demonstrating considerably good correlation above pH 5.5. In addition, Mn also shares similar redox behavior with Fe. Under oxidizing environment, dissolving Mn^{2+} and Fe^{2+} are precipitated yielding insoluble Mn^{4+} and Fe^{3+} (Dvorak & Skipton, 2014). The variation of Mn corresponds well with the trend observed for Fe, indicating that both elements are subjected to similar geochemical process.

The elevated Al, Fe and Mn in water correspond well with turbidity ($R = 0.81$, $P < 0.05$ for Al; $R = 0.83$, $P < 0.05$ for Mn; $R = 0.80$, $P < 0.05$ for Fe). The turbidity at S31 is found at an average of 1800 NTU suggesting accelerated surface runoff (Heng, 2016). Meanwhile, S23 (1176.6 NTU), S24 (1159.5 NTU) and S25 (750.8 NTU) from upstream are also characterized by high turbidity as a result of intensive logging activities with observable turbid water. Hence, elevated Al, Fe and Mn are likely associated with the transformation and desorption of the elements from eroded sediment. This mixing and desorption process can be further enhanced by rainfall land shear stress (Singh *et al.*, 2000).

The exposure to Al is a risk factor for the development of Alzheimer disease in human. Al residue in water could be reduced *via* coagulation process at an ideal pH upon treatment. On the other hand, over exposure to Mn could cause neurological effect. The manganese greensand has been used to efficiently remove Fe and Mn (Chen *et al.*, 2009). This process uses a specially formulated filter media made from a naturally mined form of glauconite greensand, layered with manganese oxide, to oxidize iron and manganese in water. Numerous of methods were applied to generate the greensand process which are continuous regeneration (CR) or intermittent regeneration (IR) or, in some cases, both. For continuous regeneration (CR), chlorine and KMnO_4 are added to oxidize Fe and Mn. Intermittent regeneration (IR) is used when the water contains relatively more Mn (Hungerford & Terry Inc, 2003).

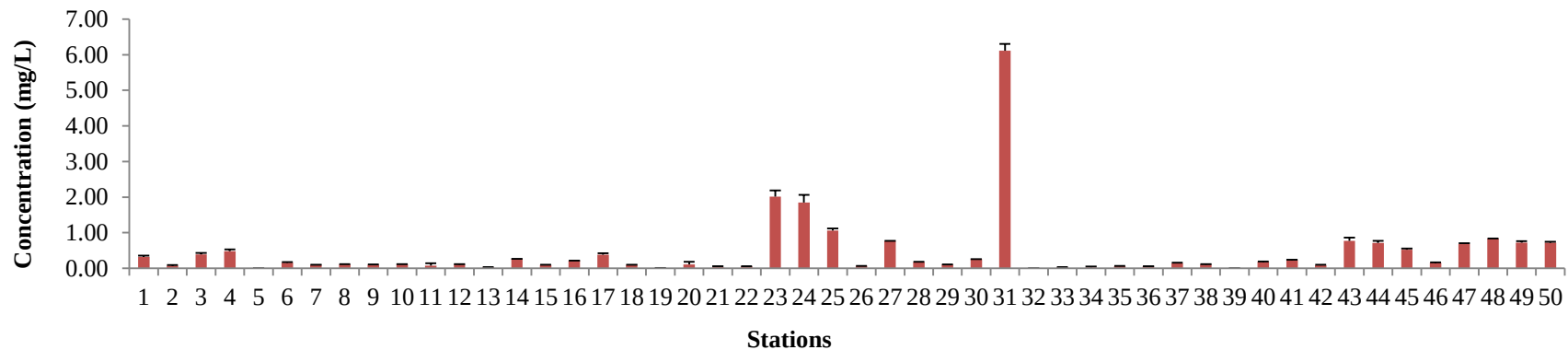
Elevated Pb was found at the middle stream, from S13 to S24, with concentration ranging between 0.02 mg/L to 0.03 mg/L, exceeding the permissible level of drinking water (0.01 mg/L). Pb is a non-essential element for plants and animals and ingestion of Pb can lead to cumulative poison (WHO, 2006). The most common aqueous species of Pb is Pb^{2+} . The element is commonly used in gasoline as an anti-knock agent in the form of tetraethyl lead (WHO, 2006). The elevated Pb may be associated to the boating activities where the concentration is profoundly governed by the pH of water (WHO, 2006). Pure Pb will dissolve into water at lower pH (Kim *et al.*, 2011). Hence, in the present study the acidification of samples prior to filtration could result in higher concentration of Pb.

Ni (< 0.02 mg/L), Cu (< 0.01 mg/L) and Zn (< 0.04 mg/L) were consistently found in trace amounts in both main rivers and tributaries of the Baleh River. Cu is an element naturally present in soils and runoff into river systems *via* natural weathering or anthropogenic sources.

In natural waters, the concentration of Cu ranges between 0.001 and 0.1 mg/L when Cu^{2+} predominates (WHO, 2006). For Zn and Ni, the natural abundance is typically below 0.01 mg/L and 0.003 mg/L, respectively (WHO, 2006). The concentrations of Zn (0.02 mg/L) and Ni (0.004 mg/L) are slightly higher and they are distributed variably across all the stations. On the other hand, Cr, Hg, As and Se were not detected in water. These elements are non-essential for plants.

(a) Al

(i) water



(ii) sediment

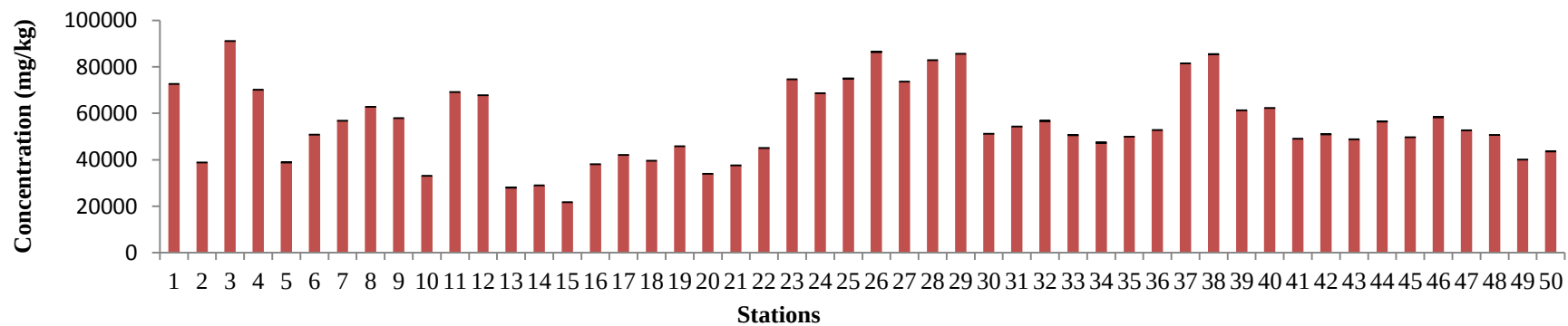


Figure 4.2 (a) to (i): The mean metal concentrations in water and sediment according to sampling stations

(b)Cr

(i) water

<LOD

(ii) sediment

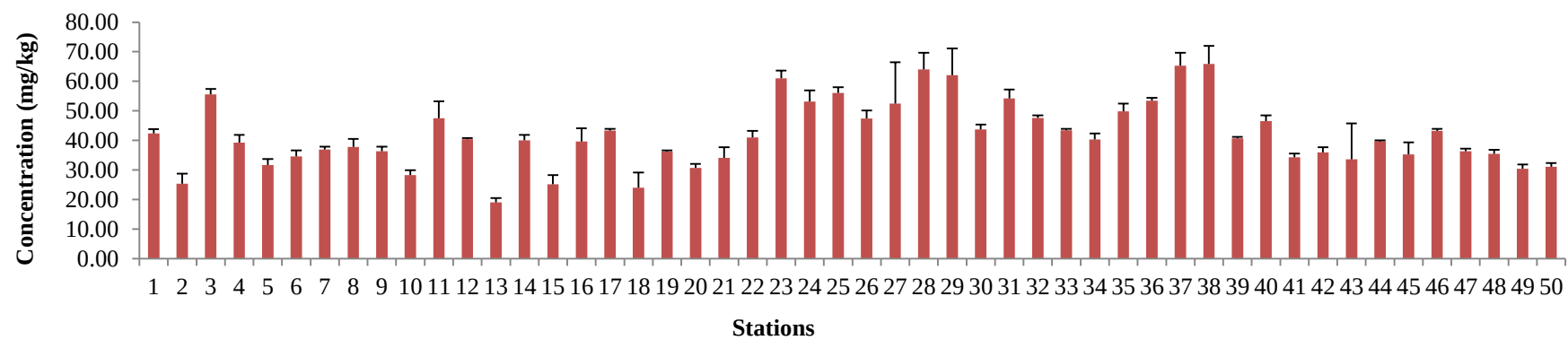
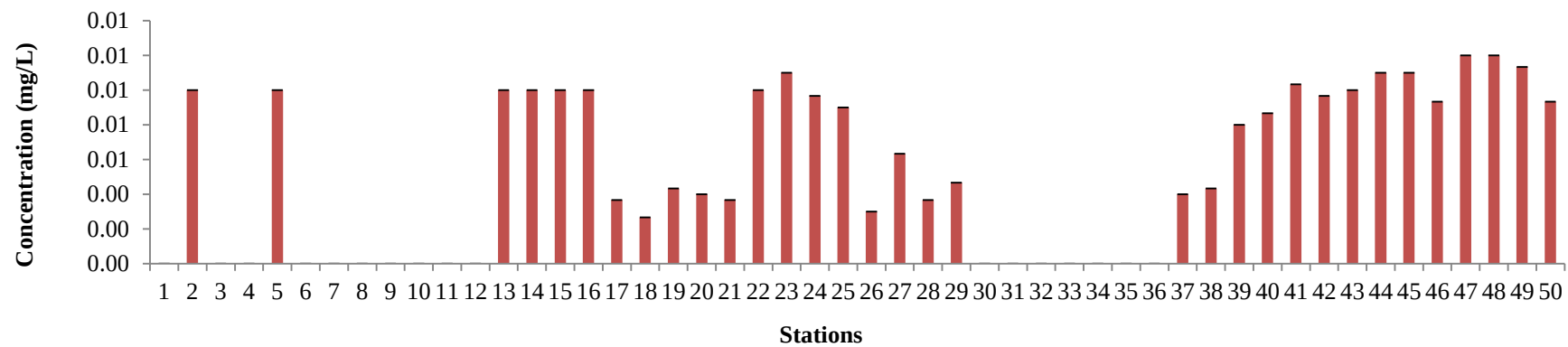


Figure 4.2: Continued

(c) Cu

(i) water



(ii) sediment

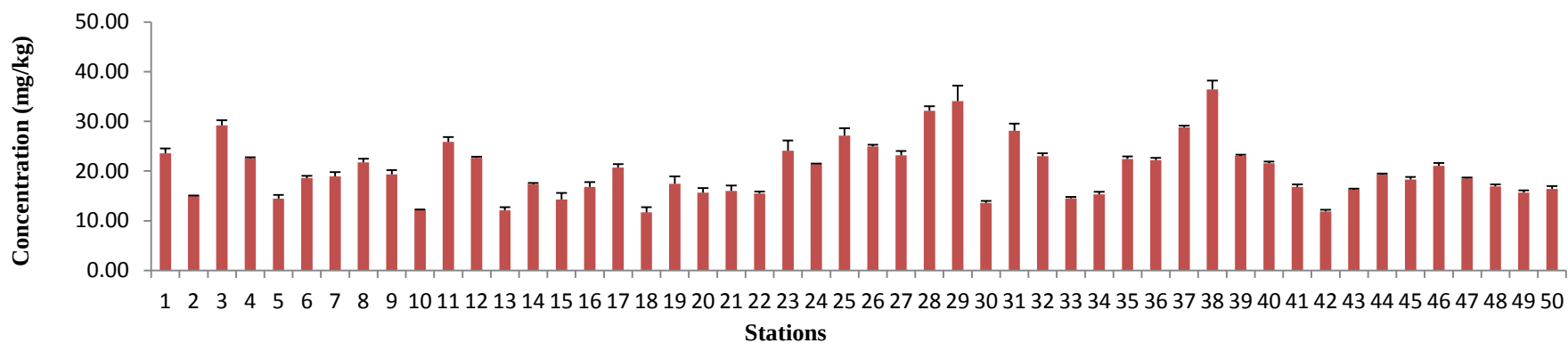
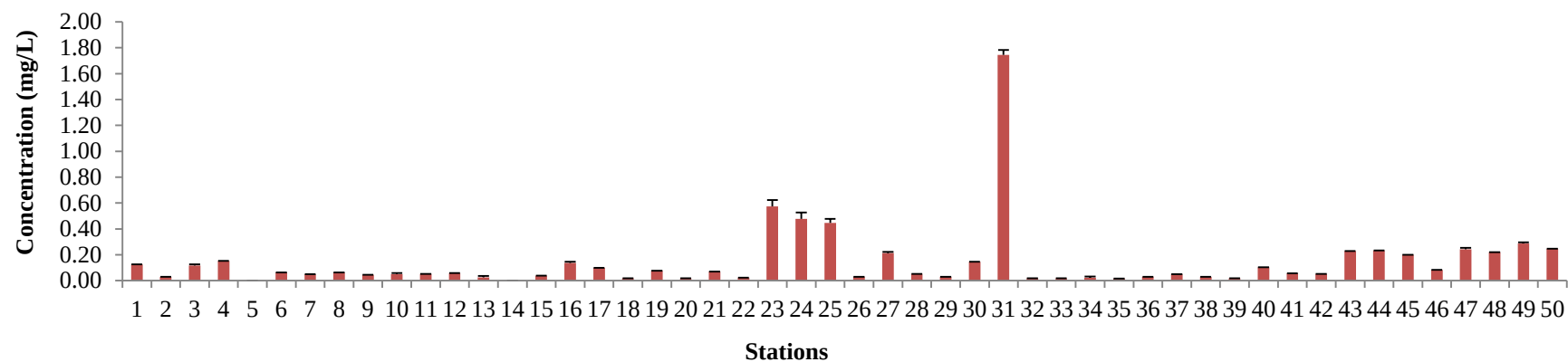


Figure 4.2: Continued

(d) Mn

(i) water



(ii) sediment

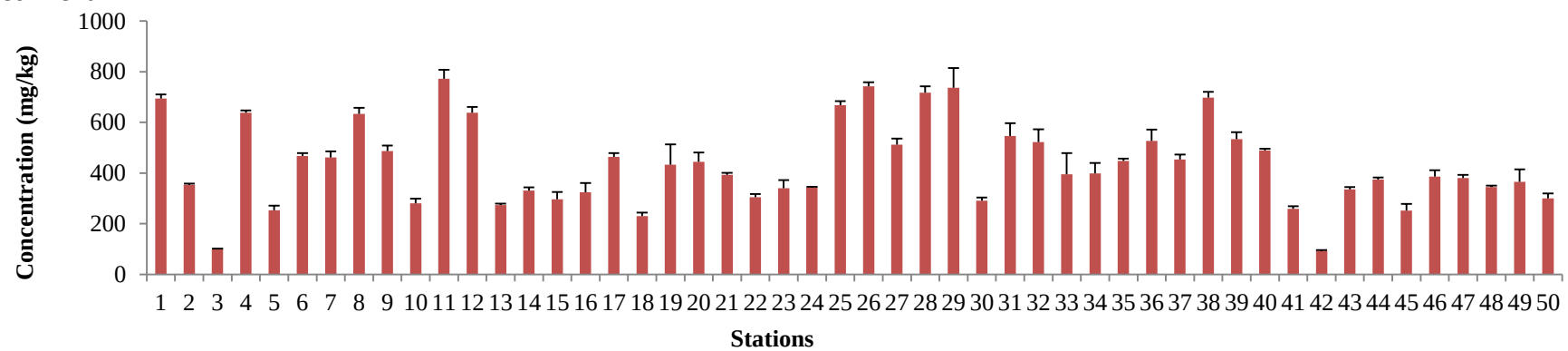
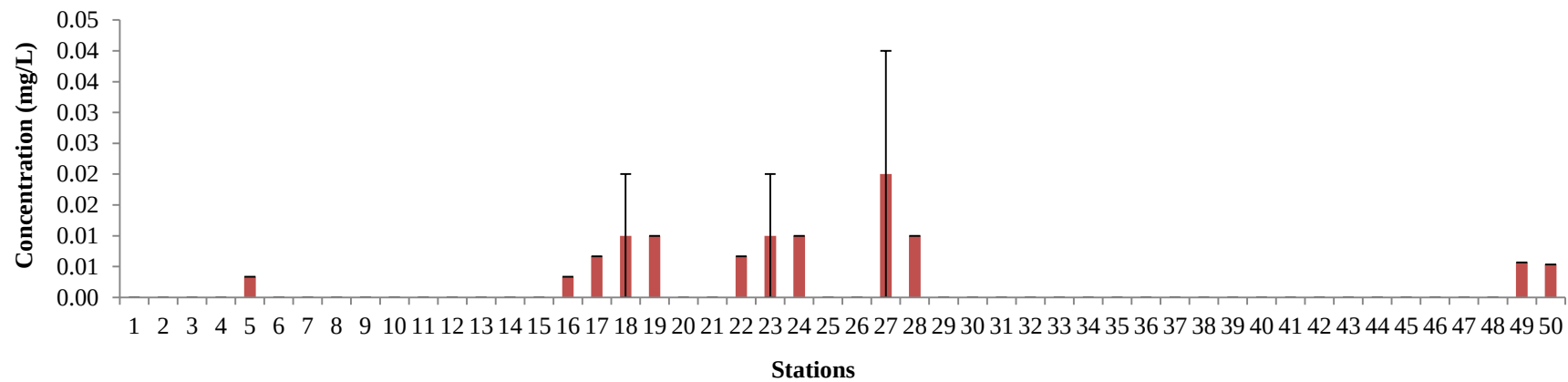


Figure 4.2: Continued

(e) Ni

(i) water



(ii) sediment

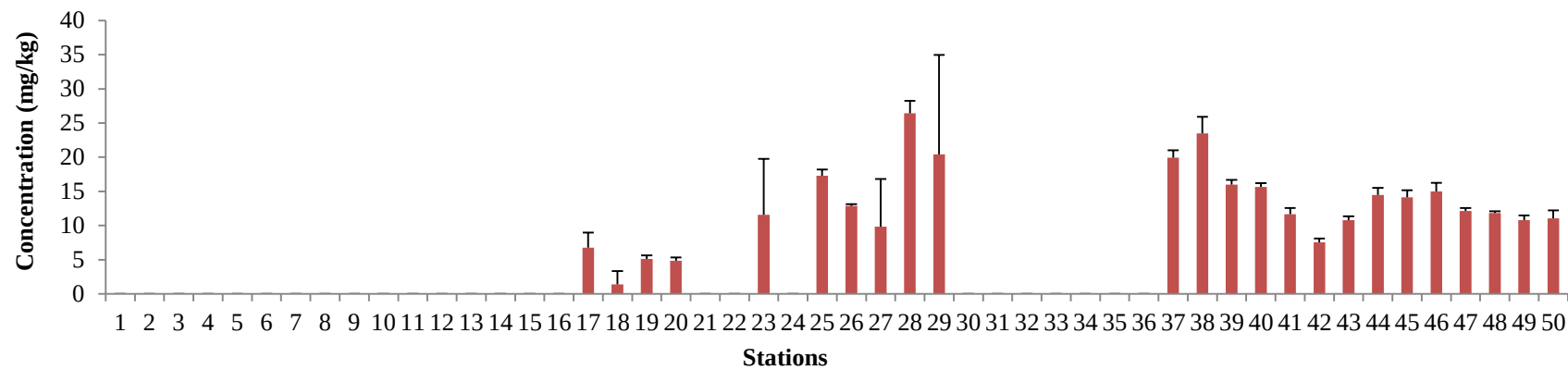
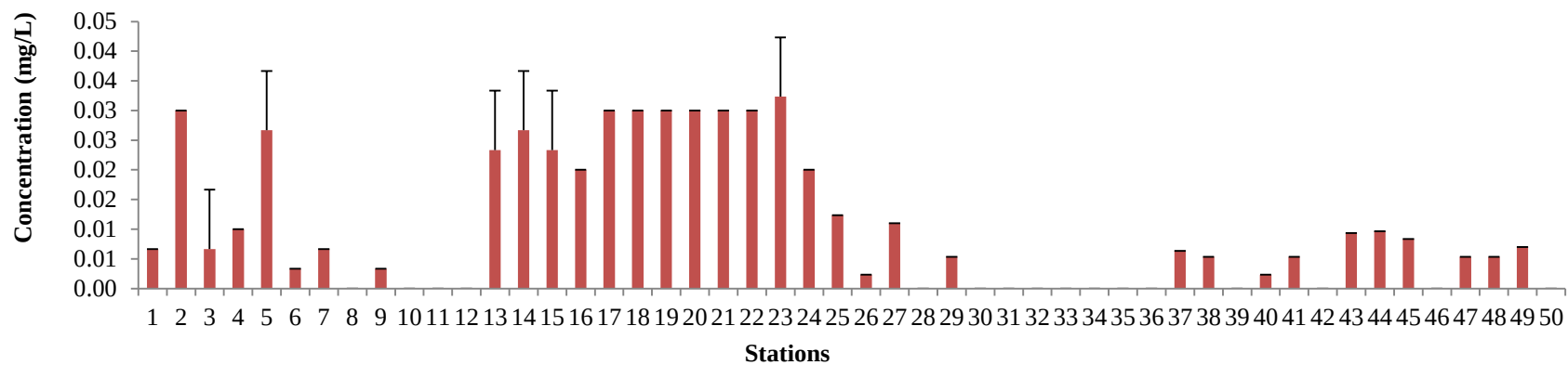


Figure 4.2: Continued

(f) Pb

(i) water



(ii) sediment

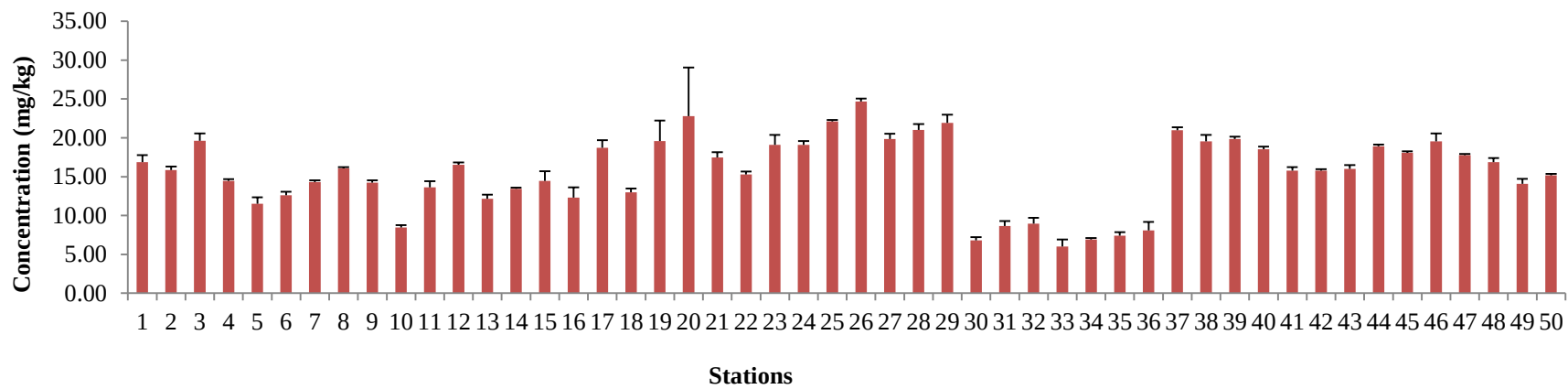
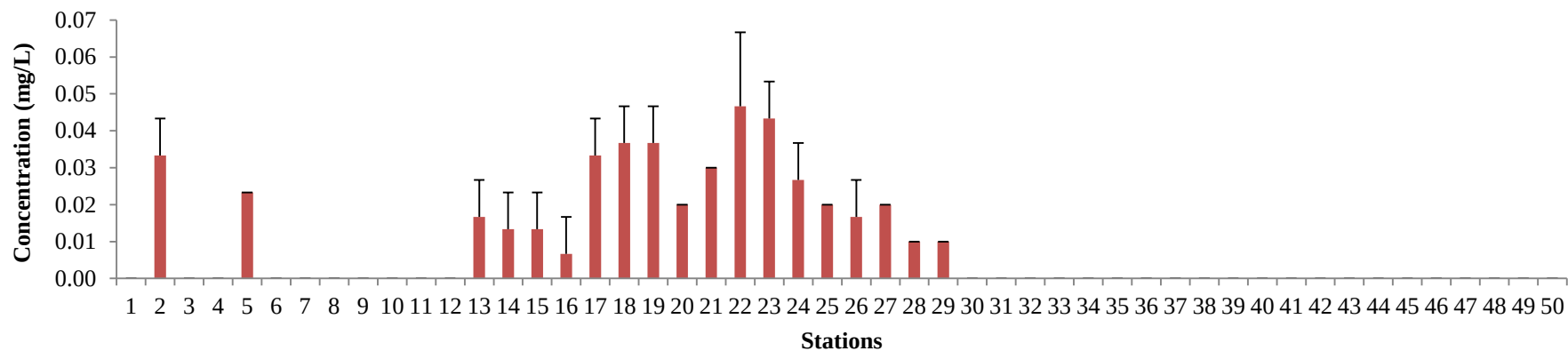


Figure 4.2: Continued

(g) Zn

(i) water



(ii) sediment

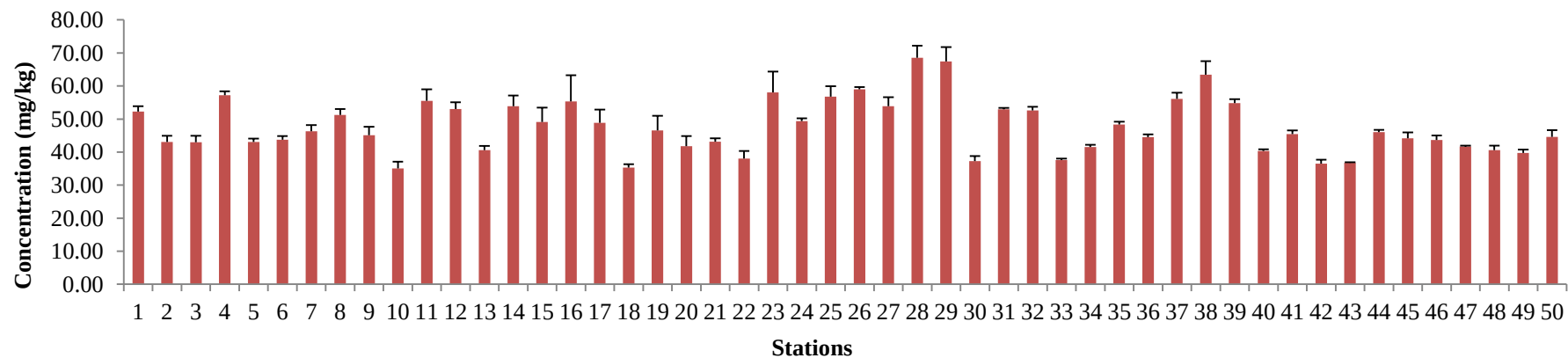
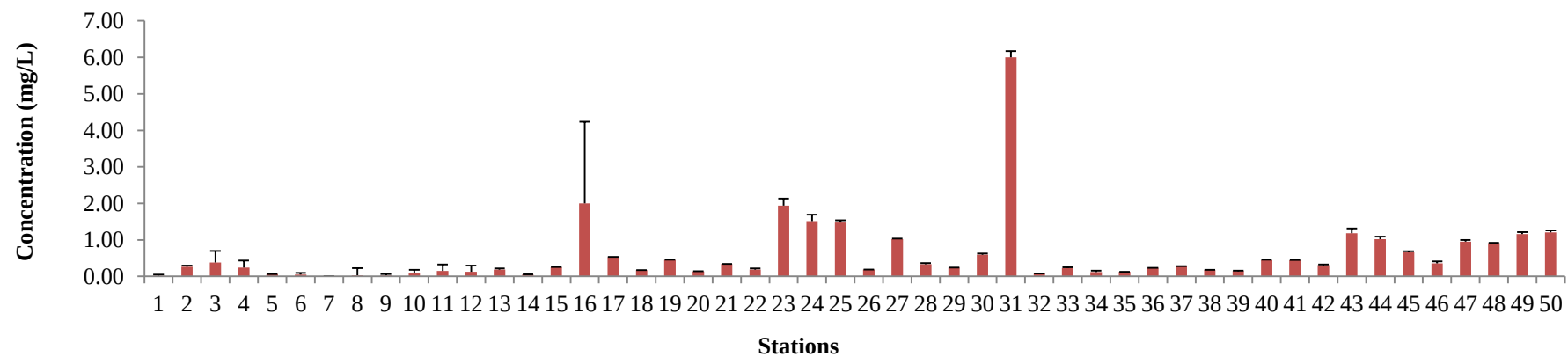


Figure 4.2: Continued

(h) Fe

(i) Water



(ii) sediment

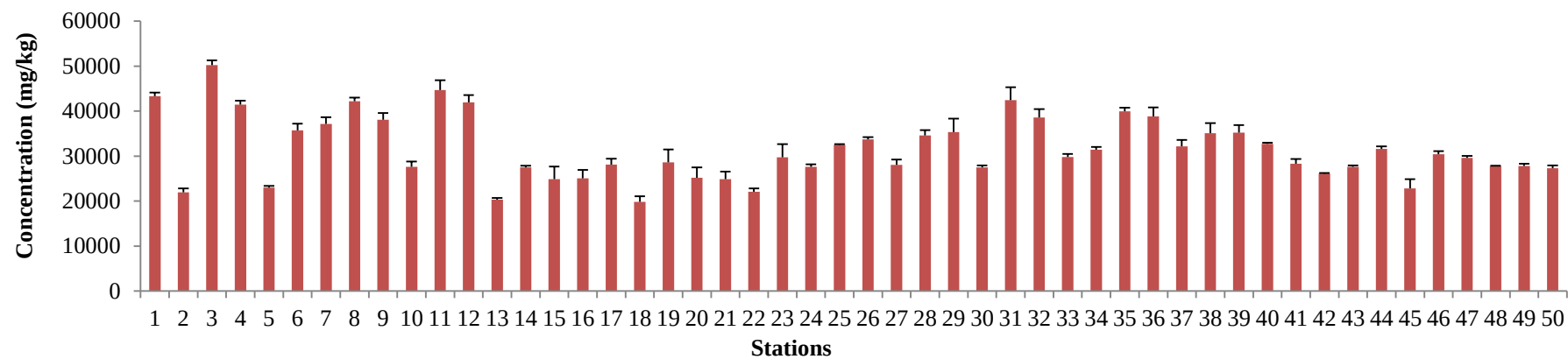
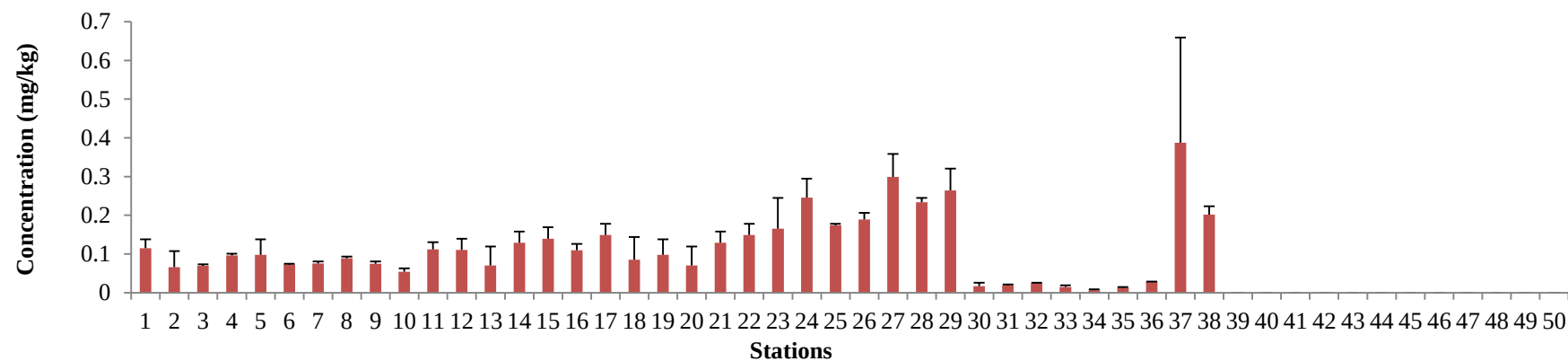


Figure 4.2: Continued

(i) Hg

(i) water

(ii) sediment



<LOD: below detection limit

Figure 4.2: Continued

4.1.3 Comparisons between dissolved, acid-extractable and total metal contents

Essentially, the most common types of metal analysis are dissolved and total dissolved metals, whilst acid extractable method has become more widespread as an alternative to either of the total or dissolved analysis (ENCO, n.d.). According to literatures, the acid-extractable analysis removes the biases introduced to samples analysed for total dissolved metals and provides more accurate prediction on the environment condition (ENCO, n.d.; Kim & Steele, 2004). The results of different fractions of metal contents are tabulated in Table 4.4. Statistically, significant differences are confirmed between dissolved, acid extractable and total metals contents ($P < 0.05$) for all the elements except Ni whilst Hg was not detected. The results revealed that total dissolved metals are much higher than dissolved metals, agreeing well with the findings of Ogunfowokan *et al.* (2013). Meanwhile, acid-extractable Mn and Pb show significant difference ($P < 0.05$) from dissolved metals; for other elements, they are comparable with no significant difference. This observation agrees well with the findings of Kim and Steele (2004) who stated that acid-extractable Fe, Mn and Pb are significantly higher than the dissolved concentrations. According to Kim and Steele (2004), acid-extractable metals are more representative for environmental considerations as the method of dissolved metals could filter out the precipitated iron oxides which consequently underestimate the iron content. Apart from that, toxic effects of metals may be underestimated because of the bioavailability of the metal from suspended solids are being filtered out. The nitric acid added to preserve dissolved metals would cause desorption of metals from clay sediments indicating the available metal concentrations.

Table 4.4: The dissolved, acid extractable and total metals in water samples

Elements (mg/L)		Stations									
		S2	S5	S13	S14	S15	S16	S17	S18	S19	S20
Al	DM	<LOD ^a	<LOD ^a	<LOD ^a	0.05±0.01 ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a
	AE	0.78±0.04 ^a	0.09±0.01 ^a	0.81±0.04 ^a	0.34±0.04 ^a	0.70±0.01 ^a	0.60±0.04 ^a	0.03±0.01 ^a	0.65±0.10 ^a	0.05±0.01 ^a	0.65±0.04 ^a
	TM	30.91±2.88 ^b	1.98±0.03 ^b	27.76±2.32 ^b	12.28±1.60 ^b	26.38±2.00 ^b	18.18±2.23 ^b	0.73±0.03 ^b	22.17±2.59 ^b	1.07±0.03 ^b	18.54±1.71 ^b
Mn	DM	<LOD ^a	<LOD ^a	<LOD ^a	0.01±0.00 ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a
	AE	0.4±0.03 ^b	0.03±0.00 ^b	0.39±0.02 ^b	0.18±0.02 ^b	0.70±0.031 ^b	0.37±0.01 ^b	0.03±0.00 ^b	0.38±0.07 ^b	0.02±0.00 ^b	0.39±0.02 ^b
	TM	0.61±0.01 ^c	0.04±0.00 ^c	0.55±0.02 ^{cc}	0.23±0.03 ^c	0.81±0.0 ^c	0.41±0.03 ^c	0.03±0.00 ^c	0.51±0.02 ^c	<LOD ^a	0.46±0.0 ^c
Fe	DM	<LOD ^a	<LOD ^a	<LOD ^a	0.04±0.01 ^a	0.01±0.00 ^a	0.05±0.01 ^a	0.04±0.02 ^a	0.06±0.00 ^a	0.05±0.00 ^a	0.07±0.02 ^a
	AE	0.77±0.05 ^a	0.12±0.00 ^a	0.83±0.04 ^a	0.33±0.05 ^a	0.95±0.01 ^a	0.78±0.04 ^a	0.17±0.02 ^a	0.85±0.20 ^a	0.23±0.02 ^a	1.10±0.05 ^a
	TM	45.65±7.51 ^b	2.27±0.06 ^b	39.86±3.63 ^b	12.74±3.34 ^b	38.33±3.28 ^b	22.71±2.87 ^b	1.06±0.05 ^b	30.84±3.83 ^b	1.18±0.04 ^b	25.02±1.37 ^b
Cr	DM	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a
	AE	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LO ^a	<LOD ^a
	TM	0.03±0.01 ^b	<LOD ^a	0.03±0.00 ^b	0.01±0.00 ^b	0.03±0.00 ^b	0.02±0.00 ^b	<LOD ^a	0.02±0.00 ^b	<LOD ^a	0.02±0.00
Cu	DM	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a
	AE	0.01±0.00 ^b	<LOD ^a	0.01±0.00 ^b	<LOD ^a	0.01±0.00 ^b	<LOD ^a	<LOD ^a	0.01±0.01 ^b	<LOD ^a	0.01±0.00 ^b
	TM	0.03±0.01 ^c	<LOD ^a	0.03±0.00 ^c	0.01±0.00 ^b	0.03±0.00 ^c	0.02±0.00 ^b	<LOD ^a	0.02±0.01 ^c	<LOD ^a	0.02±0.00 ^c
Ni	DM	<LOD ^a	<LOD ^a	<LOD ^a	<LOD	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a
	AE	<LOD ^a	<LOD ^a	<LOD ^a	0.01±0.00	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a
	TM	<LOD ^a	<LOD ^a	<LOD ^a	<LOD	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a
Pb	DM	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	0.01±0.00 ^a	<LOD ^a	<LOD ^a	<LOD ^a
	AE	0.01±0.00 ^b	0.01±0.00 ^b	0.01±0.00 ^b	0.01±0.00 ^b	0.01±0.00 ^b	0.01±0.00 ^b	0.01±0.00 ^a	0.02±0.00 ^b	0.01±0.00 ^b	0.02±0.00 ^b
	TM	0.02±0.00 ^b	<LOD ^a	0.03±0.00 ^c	0.01±0.00 ^b	0.02±0.00 ^b	0.01±0.00 ^b	0.01±0.00 ^a	0.02±0.00 ^b	<LOD ^a	0.02±0.01 ^b
Zn	DM	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a
	AE	0.01±0.00 ^b	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a	<LOD ^a
	TM	0.04±0.01 ^c	<LOD ^a	0.04±0.02 ^b	0.02±0.01 ^b	0.04±0.01 ^b	0.04±0.01 ^b	0.02±0.01 ^b	0.02±0.01 ^b	0.01±0.00 ^b	<LOD ^a

DM: Dissolved metals

AE: Acid-extractable metals

TM: Total metals

<LOD: below detection limit

4.1.4 Assessment of contamination

In the present study, the baseline values which determined based on the linear regression of cumulative distribution curves was shown in Figure 4.3. All the metals concentration were fit into the regression line under the condition of $P < 0.05$ and $R^2 > 0.9$ with no inflection point which indicated that the concentration of metals in Baleh River representing natural concentrations. Table 4.5 summarizes the baseline metals of the Baleh River in comparison to the world average proposed by Martin and Meybeck (1979). The baseline levels determined were relatively lower for all elements, except Fe where the value derived was slightly higher than the world average but within the range reported in other literatures, ranging from 73000 mg/kg to 91000 mg/kg (De Lima Rodrigues *et al.*, 2013). A study by Aprile and Bouvy (2010) reported that the concentrations of Fe and Mn are higher in the tropical river. For assessment of contamination, it will be more representative if the baseline values are derived from the area of study (Galuszka, 2007; Matschullat *et al.*, 2000; Reimann *et al.*, 2005; Wei & Wen, 2012) and these values may be used as the benchmark for future monitoring.

Table 4.5: Baseline concentrations of metals based on cumulative distribution frequency and comparison with the World Average (Martin & Meybeck, 1979)

Elements	World average (Martin & Meybeck, 1979)	Baselines concentration in the present study (mg/kg)
Al	-	91135.25
Cr	100	65.86
Cu	100	36.44
Mn	1050	772.57
Ni	90	26.42
Pb	-	24.68
Zn	350	68.55
Fe	48000	50207.60
Hg	-	0.39

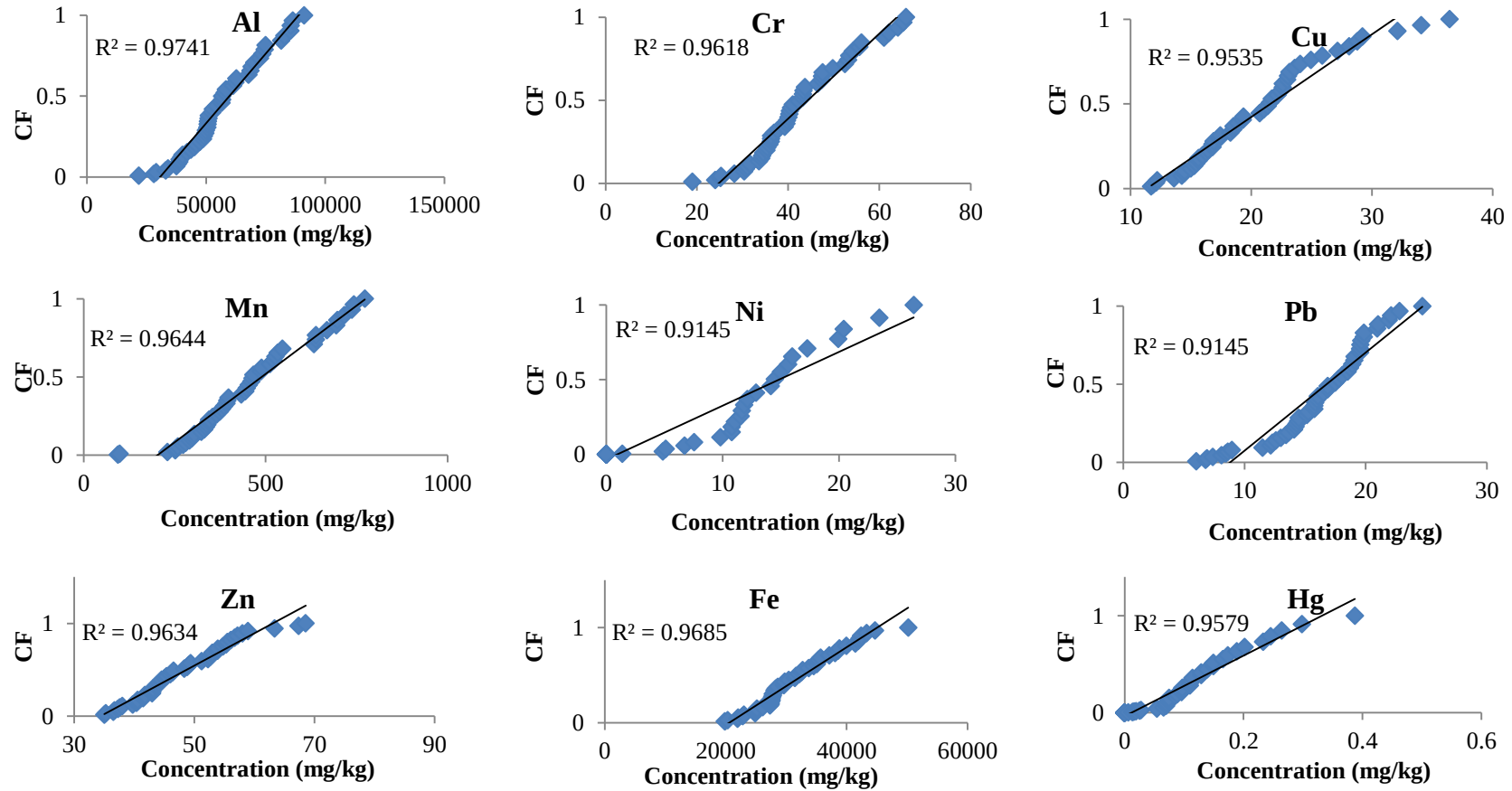


Figure 4.3: The cumulative frequency curves of metal concentrations in sediment of Baleh River

The mean contamination factors (CF) across all stations are consistently < 1 suggesting no risk of contamination (Table 4.6). The geo-accumulation indices, I_{geo} , correspondingly demonstrate minimal pollution risk for all elements ($I_{geo} < 0$) as shown in Table 4.7. The pollution load index (PLI) likewise infers minimal risk of contamination for all elements, $PLI < 1$. Baleh region is referred to as Heart of Borneo. It is largely covered with natural grassland and sparsely populated with limited accessibility. The only prominent activity is logging where no other obvious anthropogenic activities are identified at the moment. The baseline values reported is highly recommended to serve as the benchmark for future monitoring.

Table 4.6: The contamination factor (CF) and pollution load index (PLI) of metals from different stations

Stations	Elements (CF)									PLI
	Al	Cr	Cu	Mn	Ni	Pb	Zn	Fe	Hg	
1	0.797	0.643	0.648	0.898	0.000	0.684	0.763	0.863	0.294	0.668
2	0.426	0.384	0.412	0.459	0.000	0.642	0.628	0.438	0.168	0.417
3	1.000	0.844	0.802	0.128	0.000	0.796	0.627	1.000	0.179	0.545
4	0.770	0.596	0.621	0.826	0.000	0.585	0.834	0.826	0.246	0.625
5	0.427	0.481	0.397	0.328	0.000	0.466	0.628	0.458	0.250	0.416
6	0.558	0.524	0.509	0.604	0.000	0.510	0.639	0.711	0.189	0.500
7	0.623	0.560	0.519	0.597	0.000	0.580	0.676	0.740	0.193	0.528
8	0.689	0.573	0.596	0.819	0.000	0.651	0.747	0.840	0.227	0.605
9	0.635	0.551	0.530	0.630	0.000	0.576	0.658	0.758	0.192	0.532
10	0.363	0.429	0.335	0.363	0.000	0.343	0.511	0.551	0.139	0.355
11	0.758	0.721	0.710	1.000	0.000	0.553	0.810	0.890	0.287	0.678
12	0.744	0.613	0.621	0.826	0.000	0.670	0.774	0.835	0.284	0.642
13	0.308	0.289	0.333	0.354	0.000	0.493	0.592	0.405	0.180	0.350
14	0.319	0.608	0.477	0.429	0.000	0.544	0.786	0.548	0.330	0.485
15	0.239	0.382	0.391	0.384	0.000	0.586	0.716	0.496	0.357	0.423
16	0.418	0.602	0.461	0.419	0.000	0.499	0.807	0.499	0.282	0.479
17	0.462	0.657	0.568	0.600	0.255	0.758	0.712	0.560	0.383	0.526
18	0.435	0.365	0.322	0.298	0.052	0.527	0.514	0.395	0.218	0.298
19	0.503	0.550	0.479	0.560	0.194	0.793	0.679	0.570	0.250	0.469
20	0.373	0.467	0.429	0.575	0.184	0.924	0.610	0.502	0.180	0.420
21	0.413	0.518	0.439	0.508	0.000	0.708	0.629	0.495	0.330	0.493
22	0.494	0.623	0.424	0.393	0.000	0.619	0.554	0.439	0.383	0.483
23	0.819	0.927	0.661	0.440	0.438	0.773	0.847	0.593	0.424	0.631

Table 4.6: Continued

24	0.754	0.807	0.587	0.445	0.000	0.773	0.720	0.550	0.630	0.647
25	0.822	0.851	0.745	0.865	0.654	0.895	0.828	0.648	0.446	0.736
26	0.948	0.720	0.685	0.961	0.488	1.000	0.860	0.672	0.485	0.733
27	0.808	0.797	0.636	0.662	0.372	0.805	0.787	0.559	0.766	0.670
28	0.910	0.973	0.882	0.928	1.000	0.852	1.000	0.688	0.598	0.859
29	0.939	0.943	0.936	0.954	0.773	0.888	0.983	0.704	0.677	0.859
30	0.562	0.664	0.374	0.376	0.000	0.275	0.544	0.547	0.043	0.341
31	0.596	0.823	0.772	0.707	0.000	0.350	0.773	0.845	0.049	0.484
32	0.623	0.723	0.631	0.676	0.000	0.363	0.768	0.768	0.064	0.476
33	0.556	0.659	0.398	0.512	0.000	0.244	0.548	0.593	0.037	0.349
34	0.520	0.612	0.420	0.515	0.000	0.280	0.606	0.626	0.016	0.321
35	0.548	0.756	0.616	0.580	0.000	0.299	0.705	0.796	0.033	0.412
36	0.580	0.811	0.609	0.682	0.000	0.328	0.649	0.773	0.071	0.468
37	0.895	0.991	0.791	0.587	0.755	0.850	0.819	0.641	0.993	0.802
38	0.937	1.000	1.000	0.902	0.889	0.792	0.925	0.699	0.518	0.836
39	0.673	0.617	0.633	0.690	0.605	0.805	0.799	0.701	0.000	0.687
40	0.684	0.706	0.592	0.633	0.592	0.750	0.589	0.652	0.000	0.004
41	0.538	0.521	0.461	0.335	0.441	0.639	0.662	0.563	0.000	0.510
42	0.560	0.545	0.326	0.122	0.286	0.639	0.532	0.521	0.000	0.397
43	0.535	0.511	0.448	0.435	0.409	0.648	0.536	0.550	0.000	0.504
44	0.620	0.603	0.532	0.484	0.548	0.766	0.671	0.630	0.000	0.601
45	0.545	0.535	0.502	0.326	0.535	0.732	0.644	0.455	0.000	0.522
46	0.640	0.656	0.578	0.500	0.568	0.792	0.637	0.606	0.000	0.617
47	0.578	0.551	0.508	0.491	0.459	0.720	0.608	0.590	0.000	0.558
48	0.557	0.538	0.464	0.446	0.448	0.684	0.592	0.554	0.000	0.530
49	0.440	0.461	0.430	0.473	0.408	0.571	0.580	0.553	0.000	0.485
50	0.479	0.472	0.451	0.389	0.418	0.614	0.650	0.545	0.000	0.495

Table 4.7: The geo-accumulation index, I_{geo} of metals from different stations

Stations	Elements (I_{geo})										
	Al	Cr	Cu	Mn	Ni	Pb	Zn	Fe	As	Se	Hg
1	-0.912	-1.221	-1.211	-0.739	0.000	-1.134	-0.976	-0.798	0.000	0.000	-2.353
2	-1.815	-1.964	-1.864	-1.710	0.000	-1.225	-1.256	-1.776	0.000	0.000	-3.160
3	-0.585	-0.829	-0.904	-3.549	0.000	-0.915	-1.259	-0.585	0.000	0.000	-3.066
4	-0.961	-1.331	-1.272	-0.861	0.000	-1.358	-0.846	-0.862	0.000	0.000	-2.608
5	-1.812	-1.641	-1.919	-2.195	0.000	-1.686	-1.256	-1.710	0.000	0.000	-2.585
6	-1.427	-1.516	-1.558	-1.312	0.000	-1.555	-1.232	-1.077	0.000	0.000	-2.992
7	-1.268	-1.421	-1.530	-1.329	0.000	-1.372	-1.150	-1.020	0.000	0.000	-2.961
8	-1.123	-1.388	-1.331	-0.873	0.000	-1.204	-1.005	-0.836	0.000	0.000	-2.724
9	-1.239	-1.445	-1.500	-1.251	0.000	-1.381	-1.188	-0.984	0.000	0.000	-2.966
10	-2.047	-1.807	-2.162	-2.046	0.000	-2.129	-1.554	-1.446	0.000	0.000	-3.430
11	-0.984	-1.057	-1.078	-0.585	0.000	-1.440	-0.889	-0.752	0.000	0.000	-2.384
12	-1.011	-1.290	-1.272	-0.860	0.000	-1.163	-0.955	-0.845	0.000	0.000	-2.403
13	-2.284	-2.378	-2.172	-2.083	0.000	-1.604	-1.341	-1.888	0.000	0.000	-3.062
14	-2.235	-1.302	-1.652	-1.807	0.000	-1.463	-0.932	-1.454	0.000	0.000	-2.183
15	-2.650	-1.972	-1.939	-1.965	0.000	-1.355	-1.066	-1.597	0.000	0.000	-2.072
16	-1.842	-1.318	-1.701	-1.840	0.000	-1.589	-0.894	-1.587	0.000	0.000	-2.414
17	-1.698	-1.190	-1.400	-1.321	-2.556	-0.984	-1.074	-1.422	0.000	0.000	-1.971
18	-1.786	-2.040	-2.221	-2.333	-4.839	-1.510	-1.544	-1.924	0.000	0.000	-2.784
19	-1.577	-1.448	-1.647	-1.420	-2.952	-0.920	-1.143	-1.396	0.000	0.000	-2.585
20	-2.007	-1.684	-1.804	-1.383	-3.024	-0.699	-1.299	-1.580	0.000	0.000	-3.062
21	-1.861	-1.534	-1.773	-1.561	0.000	-1.082	-1.254	-1.598	0.000	0.000	-2.183
22	-1.601	-1.269	-1.821	-1.931	0.000	-1.277	-1.436	-1.772	0.000	0.000	-1.971
23	-0.873	-0.695	-1.183	-1.768	-1.777	-0.956	-0.825	-1.340	0.000	0.000	-1.823
24	-0.993	-0.894	-1.354	-1.755	0.000	-0.956	-1.058	-1.447	0.000	0.000	-1.251
25	-0.867	-0.818	-1.009	-0.794	-1.199	-0.744	-0.858	-1.211	0.000	0.000	-1.750
26	-0.662	-1.059	-1.131	-0.642	-1.621	-0.585	-0.802	-1.159	0.000	0.000	-1.630
27	-0.893	-0.913	-1.238	-1.180	-2.012	-0.899	-0.931	-1.424	0.000	0.000	-0.970
28	-0.722	-0.625	-0.766	-0.693	-0.585	-0.816	-0.585	-1.123	0.000	0.000	-1.326
29	-0.675	-0.669	-0.681	-0.653	-0.957	-0.756	-0.610	-1.090	0.000	0.000	-1.147
30	-1.417	-1.176	-2.005	-1.994	0.000	-2.448	-1.464	-1.454	0.000	0.000	-5.128
31	-1.331	-0.866	-0.959	-1.084	0.000	-2.099	-0.957	-0.828	0.000	0.000	-4.924
32	-1.269	-1.054	-1.249	-1.149	0.000	-2.045	-0.966	-0.966	0.000	0.000	-4.545
33	-1.433	-1.187	-1.913	-1.552	0.000	-2.618	-1.452	-1.338	0.000	0.000	-5.344
34	-1.528	-1.294	-1.837	-1.541	0.000	-2.419	-1.309	-1.261	0.000	0.000	-6.591
35	-1.453	-0.989	-1.285	-1.370	0.000	-2.325	-1.089	-0.915	0.000	0.000	-5.499
36	-1.371	-0.888	-1.299	-1.137	0.000	-2.193	-1.208	-0.956	0.000	0.000	-4.402
37	-0.746	-0.598	-0.924	-1.353	-0.991	-0.820	-0.874	-1.226	0.000	0.000	-0.595
38	-0.679	-0.585	-0.585	-0.733	-0.755	-0.921	-0.698	-1.102	0.000	0.000	-1.534
39	-1.156	-1.281	-1.245	-1.120	-1.309	-0.898	-0.909	-1.097	0.000	0.000	0.000
40	-1.133	-1.088	-1.341	-1.244	-1.342	-0.999	-1.349	-1.202	0.000	0.000	0.000
41	-1.478	-1.526	-1.703	-2.164	-1.767	-1.231	-1.179	-1.413	0.000	0.000	0.000

Table 4.7: Continued

42	-1.422	-1.461	-2.201	-3.622	-2.393	-1.232	-1.494	-1.526	0.000	0.000	0.000
43	-1.486	-1.554	-1.745	-1.787	-1.876	-1.211	-1.486	-1.447	0.000	0.000	0.000
44	-1.274	-1.314	-1.496	-1.631	-1.452	-0.970	-1.160	-1.252	0.000	0.000	0.000
45	-1.460	-1.487	-1.580	-2.202	-1.487	-1.036	-1.220	-1.722	0.000	0.000	0.000
46	-1.228	-1.193	-1.377	-1.586	-1.401	-0.922	-1.236	-1.308	0.000	0.000	0.000
47	-1.376	-1.445	-1.561	-1.610	-1.707	-1.058	-1.303	-1.346	0.000	0.000	0.000
48	-1.430	-1.480	-1.693	-1.749	-1.742	-1.134	-1.341	-1.438	0.000	0.000	0.000
49	-1.769	-1.702	-1.803	-1.664	-1.879	-1.394	-1.372	-1.438	0.000	0.000	0.000
50	-1.646	-1.668	-1.734	-1.948	-1.843	-1.288	-1.207	-1.462	0.000	0.000	0.000

4.1.5 Correlations of metals

Pearson's correlation analysis was employed to examine the intermetallic relationships in water and sediment (Table 4.8). As expected, significant positive correlations are deduced between Al- Fe- Mn in water. For sediment, notable correlation is established between Zn- Cu, Cr- Zn, and Cu- Cr. According to Ashraf (2012), a strong correlation between two elements may be an occurrence of strong dependence of both metals on the same causal factor. The positive correlation between Al and Fe suggest that these two elements originate from same source during discharge process (Degroot & Allersma, 1975; Xue *et al.*, 2006).

Table 4.8: The Pearson's correlation of metals in (a) water and (b) sediment

(a)

	<i>Al</i>	<i>Cu</i>	<i>Mn</i>	<i>Ni</i>	<i>Pb</i>	<i>Zn</i>	<i>Fe</i>
Al	1.000						
Cu	0.072	1.000					
Mn	0.991**	0.076	1.000				
Ni	0.115	0.105	0.095	1.000			
Pb	-0.031	0.294	-0.055	0.360	1.000		
Zn	0.016	0.177	-0.003	0.504	0.806	1.000	
Fe	0.893**	0.167	0.909**	0.174	-0.012	0.000	1.000

** $r \geq 0.893$ indicates significance at $P=0.01$; $n=150$

(b)

	<i>Al</i>	<i>Cr</i>	<i>Cu</i>	<i>Mn</i>	<i>Ni</i>	<i>Pb</i>	<i>Zn</i>	<i>Fe</i>	<i>Hg</i>
Al	1.000								
Cr	0.217	1.000							
Cu	0.222	0.954**	1.000						
Mn	0.559	0.089	0.180	1.000					
Ni	0.468	0.575	0.654*	0.273	1.000				
Pb	0.056	0.755*	0.810*	-0.028	0.666*	1.000			
Zn	0.010	0.918**	0.945**	-0.002	0.538	0.848*	1.000		
Fe	0.628*	-0.249	-0.223	0.577	-0.034	-0.516	-0.421	1.000	
Hg	0.429	0.272	0.274	0.343	0.219	0.252	0.221	0.025	1.000

** $r \geq 0.918$ indicates significance at $P= 0.01$; * $r \geq 0.628$ indicates significance at $P=0.05$; $n=150$

4.2 Heavy metals in fishes

4.2.1 Method validation

The recovery percentages of As, Cu, Zn, Mn and Hg in fish based on the CRM are summarized in Table 4.9. For Al, spiked method was used attaining a percentage of 103.00 %. Other elements demonstrated recovery percentages between 91.75 and 118.18 %, falling within the acceptable range of 75– 125 % (Cussen & Hensman, 2011).

Table 4.9: Mean metals concentration recovered and the recovery percentages in fish

Elements	Expected value (ppm)	Observed value (ppm)	Recovery (%)
As	12.70	12.59±1.98	99.13±15.61
Cu	1.67	1.77±0.23	105.99±13.66
Mn	0.37	0.42±0.04	113.51±9.49
Zn	16.00	14.68±0.67	91.75±4.17
Hg	0.11	0.13±0.02	118.18±13.89
Al*	0.10	0.1±0.01	103.00±5.77

* spiked method

4.2.2 Distribution of metals in fish by parts (muscle, belly, gills)

Heavy metals in fishes were examined using PCA to demonstrate the clustering pattern of samples according to parts. Figure 4.4 illustrates the scores plot of PC2 versus PC1 with a total variance explained of 58.13 %. The metal distributions in gills are noticeably distinguishable from muscle and belly suggesting dissimilar metals accumulation pattern.

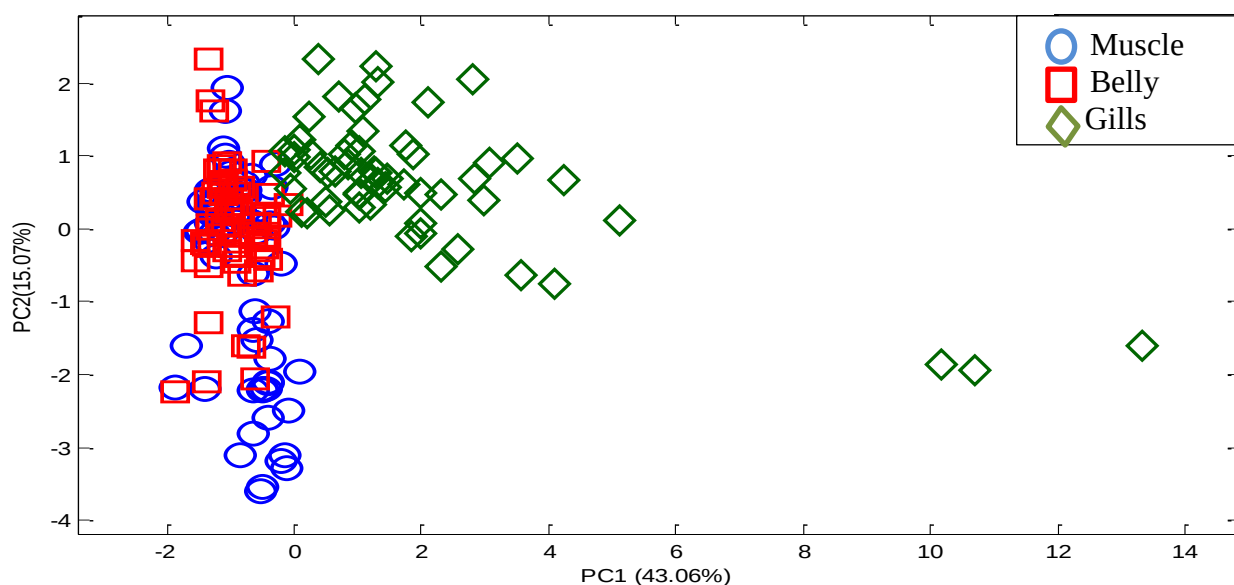


Figure 4.4: The scores plot of PC2 vs PC1 for heavy metals in fishes according to parts

Table 4.10 summarizes the metal contents in various species of fish according to parts (muscle, belly and gills). Statistically, no significant difference is deduced between muscle and belly whilst gills demonstrated significant difference with other parts ($P < 0.05$) for all elements. The metals content in muscle and belly are measured due to the significance on human consumption, while the level of metal in gills reflects the concentration of metals in water (Taweel, 2011). Most elements are found in higher concentrations in gills. This includes Cr, 2.58 mg/kg dry weight (dw), Fe (725.26 mg/kg dw), Mn (91.91 mg/kg dw), Pb (5.40 mg/kg dw), Zn (99.01 mg/kg dw) and Al (918.43 mg/kg dw). This result corroborates the findings of Dhaneesh *et al.* (2012) and El-Moselhy *et al.* (2014) who stated that Pb and Mn are prone to accumulation in gills. Other studies likewise revealed that Pb tends to accumulate in gills due to its primarily larger surface area that facilitates greater interaction and absorption. (Kargin, 1998; Avenant-Oldewage & Marx, 2000; Zhao *et al.*, 2012). The studies of Ahmed and Bibi (2010) further indicates that Pb entering the body of fish through gills is bound to the mucus layer and is subsequently absorbed by other tissues. The gills exhibit

relatively weaker detoxification therefore the metal burden in gills is high (Taweel, 2011). The concentration of Fe, Mn and Al are particularly high in gills as these elements are the most abundant in water. The metals can penetrate directly through gills when their concentrations in water and sediment increase (Bashir *et al.*, 2013). A study by Rajkwaska and Protasowicki (2012) demonstrates that Fe penetrates mainly through gills, while Mn is absorbed through gills and skins. Gills also demonstrate higher affinity for Zn and Cu. However this depends primarily on the species nature (Rajkwaska & Protasowicki, 2011). Evidently, *Oxygaster anomalura*, *Lobocheilos setigerum*, *Leocassis macropogon*, *Hemibagrus wyckii*, *Hemibagrus planiceps*, *Tor tambroides*, *Luciosoma spilopteura*, *Pengasius micronemus* and *Rasbora borneensis* indicate higher Cu in muscle than gills. *Oxygaster anomalura*, *Leocassis macropogon* and *Rasbora borneensis* are carnivores whilst *Lobocheilos setigerum*, *Hemibagrus wyckii*, *Hemibagrus planiceps*, *Tor tambroides*, *Luciosoma spilopteura* and *Pengasius micronemus* are omnivores. This may conclude that carnivores and omnivores tend to accumulate higher Cu in muscle. Inevitably, abiotic and biotic factors also play an important role in metal accumulation. The abiotic factors include biological habitat, chemical forms, temperature, salinity, pH, dissolved oxygen concentration and water transparency (Storelli & Marcotrigiano, 2005) whilst biotic factors encompass species, sex, body mass, age, physiologic conditions and nourishment sources (Castro-González & Méndez-Armenta, 2008). The accumulation of Hg is widely reported in fish. The element is found to be consistently higher in muscle and belly (0.01– 3.59 mg/kg dw) compared to that in gills (lower than detection limit to 0.44 mg/kg dw). Sackett *et al.* (2013) revealed that Hg elimination in muscle is relatively slow therefore tends to be accumulated over time. Besides, a study by Hosseini *et al.* (2013) reported that the metallothionein protein in muscle has a high affinity for Hg rendering the element hard to be eliminated.

Table 4.10: Metal contents in various species of fish according to (a) muscle, (b) belly and (c) gills

(a)

Part	Species	Elements (dry weight, mg/kg)								
		Cr	Cu	Fe	Mn	Ni	Pb	Zn	Al	Hg
Muscle	<i>O. anomalura</i>	<LOD ^a	10.67±1.89 ^b	76.00±13.06 ^a	2.67±1.89 ^{a,b}	<LOD ^a	5.33±1.89 ^{b,c,d}	69.33±7.54 ^g	20.22±2.26 ^{a,b}	0.48±0.32 ^{a,b,c,d,e}
	<i>L. setigerum</i>	<LOD ^a	12±0.00 ^b	102.67±15.43 ^{a,b}	4.00±0.00 ^{a,b,c}	<LOD ^a	5.33±1.89 ^{b,c,d}	52.00±3.27 ^{e,f,g}	43.15±13.16 ^{a,b}	0.66±0.03 ^{c,d,e,f}
	<i>L. micropogon</i>	<LOD ^a	12±0.01 ^b	245.33±43.49 ^{a,b,c}	4.00±0.00 ^{a,b,c}	<LOD ^a	8.00±0.01 ^d	26.67±1.89 ^{a,b,c,d,e}	216.55±43.42 ^d	1.38±0.10 ^g
	<i>B. collingwoodii</i>	1.45±2.06 ^a	2.71±2.23 ^a	265.56±325.24 ^{a,b,c}	3.20±2.53 ^{a,b,c}	<LOD ^a	1.15±1.62 ^{a,b}	20.17±2.35 ^{a,b,c,d}	<LOD ^a	0.21±0.02 ^{a,b,c}
	<i>H. bimaculata</i>	<LOD ^a	<LOD ^a	32.00±8.64 ^a	1.33±1.89 ^{a,b}	<LOD ^a	1.33±1.89 ^{a,b}	21.33±1.89 ^{a,b,c,d}	14.04±1.09 ^{a,b}	3.59±0.32 ⁱ
	<i>H. wyckii</i>	<LOD ^a	12±0.00 ^b	56.00±5.66 ^a	4.00±0.00 ^{a,b,c}	<LOD ^a	6.67±1.89 ^{c,d}	28.00±3.27 ^{a,b,c,d,e}	14.42±2.58 ^{a,b}	1.94±0.08 ^h
	<i>H. planiceps</i>	<LOD ^a	8.00 ^b	392.57±76.24 ^{b,c}	6.67±1.89 ^{b,c}	1.33±1.89 ^a	4.00±0.00 ^{a,b,c,d}	33.33±3.77 ^{b,c,d,e,f}	281.50±11.40 ^e	2.26±0.13 ^h
	<i>B. schwanenfeldii</i>	1.42±2.01 ^a	0.84±1.19 ^a	36.19±9.01 ^a	2.43±0.46 ^{a,b}	<LOD ^a	2.57±1.85 ^{a,b,c}	<LOD ^a	13.59±19.21 ^{a,b}	0.13±0.02 ^{a,b}
	<i>T. tambroides</i>	<LOD ^a	9.33±1.89 ^b	88.00±18.18 ^a	1.33±0.00 ^{a,b}	<LOD ^a	4.00±0.00 ^{a,b,c,d}	29.33±4.99 ^{b,c,d,e}	62.69±32.64 ^b	0.14±0.18 ^{a,b}
	<i>L. bo</i>	<LOD ^a	1.33±1.89 ^a	144.00±25.92 ^{a,b}	13.33±1.89 ^d	<LOD ^a	<LOD ^a	114.67±22.23 ^h	134.18±18.46 ^c	0.60±0.10 ^{b,c,d,e,f}
	<i>T. dourenensis</i>	<LOD ^a	1.33±1.89 ^a	146.58±35.27 ^a	1.33±1.89 ^{a,b}	1.33±1.89 ^a	2.67±1.89 ^{a,b,c}	36.67±5.73 ^{b,c,d,e,f}	17.66±1.79 ^{a,b}	0.40±0.01 ^{a,b,c,d}
	<i>C. apogon</i>	1.41±1.99 ^a	<LOD ^a	50.51±24.19 ^a	8.26±1.73 ^{c,d}	<LOD ^a	2.14±1.51 ^{a,b}	14.00±10.00 ^{a,b,c,d}	<LOD ^a	0.01±0.00 ^a
	<i>O. schelegelii</i>	<LOD ^a	<LOD ^a	102.14±121.82 ^{a,b}	1.33±1.89 ^{a,b}	1.33±1.89 ^a	2.67±1.89 ^{a,b,c}	13.33±1.89 ^{a,b,c}	14.36±0.91 ^{a,b}	0.32±0.05 ^{a,b,c,d}
	<i>P. macronema</i>	<LOD ^a	<LOD ^a	463.50±36.12 ^c	1.33±1.89 ^{a,b}	<LOD ^a	<LOD ^a	28.00±0.00 ^{a,b,c,d,e}	13.19±4.11 ^{a,b}	0.12±0.07 ^{a,b}
	<i>O. vittatus</i>	<LOD ^a	<LOD ^a	10.56±5.14 ^a	<LOD ^a	1.33±1.89 ^a	<LOD ^a	28.00±3.27 ^{a,b,c,d,e}	20.22±2.82 ^{a,b}	1.09±0.28 ^{f,g}
	<i>L. Spilopteura</i>	<LOD ^a	10.67±1.89 ^b	90.67±4.99 ^a	4.00±0.00 ^{a,b,c}	<LOD ^a	4.00±0.01 ^{a,b,c,d}	42.67±1.89 ^{d,e,f,g}	43.87±1.90 ^{a,b}	0.70±0.03 ^{c,d,e,f}
	<i>L. fasciatus</i>	<LOD ^a	3.21±0.22 ^a	68.90±63.39 ^a	4.38±0.65 ^{a,b,c}	8.71±1.65 ^b	<LOD ^a	51.58±17.53 ^{e,f,g}	4.42±6.25 ^a	0.01±0.00 ^a
	<i>P.s micronemus</i>	<LOD ^a	2.12±0.05 ^a	27.87±2.95 ^a	1.80±0.06 ^{a,b}	1.74±1.04 ^a	<LOD ^a	<LOD ^a	10.59±8.11 ^{a,b}	0.13±0.1 ^{a,b}
	<i>M. maculatus</i>	<LOD ^a	2.20±0.28 ^a	31.09±3.35 ^a	4.15±0.41 ^{a,b,c}	1.82±1.67 ^a	<LOD ^a	37.51±8.62 ^{c,d,e,f}	15.16±21.42 ^{a,b}	0.94±0.06
	<i>R. borneensis</i>	<LOD ^a	0.67±0.95 ^a	17.68±0.90 ^a	3.13±0.31 ^{a,b,c}	1.23±0.38 ^a	<LOD ^a	58.55±2.51 ^{f,g}	<LOD ^a	0.37±0.01 ^{a,b,c,d}
	<i>A. armatus</i>	<LOD ^a	0.66±0.94 ^a	24.48±2.21 ^a	1.53±0.33 ^{a,b}	1.46±0.42 ^a	<LOD ^a	24.93±8.22 ^{a,b,c,d,e}	<LOD ^a	0.76±0.03 ^{d,e,f}
	<i>C. lucius</i>	<LOD ^a	<LOD ^a	21.35±0.35 ^a	6.69±2.58 ^{b,c}	1.02±0.05 ^a	<LOD ^a	7.55±0.01 ^{a,b}	<LOD ^a	1.07±0.02 ^{f,g}

Table 4.10: Continued
(b)

Part	Species	Elements (dry weight, mg/kg)								
		Cr	Cu	Fe	Mn	Ni	Pb	Zn	Al	Hg
Belly	<i>O. anomalura</i>	<LOD ^a	<LOD ^a	75.51±19.93 ^{a,b,c}	3.13±1.23 ^{a,b}	1.35±1.01 ^{a,b,c}	7.09±0.59 ^{e,f}	58.54±8.91 ^{g,h}	13.50±4.46 ^a	0.52±0.04 ^{b,c,d,e,f}
	<i>L. setigerum</i>	0.24±0.06 ^a	0.50±0.42 ^{a,b,c}	59.42±18.76 ^{a,b,c}	2.79±2.69 ^{a,b}	0.89±0.87 ^{a,b}	6.42±0.73 ^{d,e,f}	37.27±2.34 ^{e,f,g}	6.18±0.30 ^a	0.66±0.06 ^{d,e,f}
	<i>L. micropogon</i>	0.62±0.23 ^a	3.14±0.29 ^{c,d}	171.94±9.59 ^{d,e}	5.82±0.52 ^{a,b,c}	4.75±1.76 ^{b,c,d,e}	5.87±0.82 ^{c,d,e,f}	13.01±13.81 ^{a,b,c,d}	368.14±110.00 ^c	1.36±0.02 ^g
	<i>B. collingwoodii</i>	<LOD ^a	0.72±1.02 ^{a,b,c}	54.46±37.21 ^{a,b,c}	2.31±0.69 ^{a,b}	<LOD ^a	<LOD ^a	5.38±7.61 ^{a,b,c}	<LOD ^a	0.22±0.01 ^{a,b,c}
	<i>H. bimaculata</i>	<LOD ^a	<LOD ^a	31.36±2.52 ^{a,b,c}	1.33±1.84 ^a	1.33±1.89 ^{a,b,c}	2.67±1.89 ^{a,b,c}	32.00±6.53 ^{d,e,f}	19.35±3.63 ^a	3.63±0.36 ⁱ
	<i>H. wyckii</i>	0.25±0.07 ^a	0.19±0.26 ^{a,b}	<LOD ^a	1.99±0.19 ^{a,b}	4.3±1.10 ^{b,c,d,e}	3.03±0.07 ^{a,b,c}	1.72±0.1 ^{a,b}	6.10±0.32 ^a	1.62±0.11 ^{g,h}
	<i>H. planiceps</i>	0.60±0.07 ^a	2.80±0.62 ^{a,b,c,d}	104.59±55.69 ^{c,d,e}	1.96±0.22 ^a	1.45±0.04 ^{a,b,c}	6.90±1.25 ^{e,f}	1.29±0.85 ^{a,b}	252.38±18.66 ^b	1.99±0.18 ^h
	<i>B. schwanenfeldii</i>	1.40±1.98 ^a	1.64±1.17 ^{a,b,c,d}	31.47±3.46 ^{a,b,c}	1.96±0.24 ^a	<LOD ^a	2.54±1.85 ^{a,b}	<LOD ^a	<LOD ^a	0.14±0.01 ^{a,b,c}
	<i>T. tambroides</i>	<LOD ^a	1.33±1.89 ^{a,b,c}	180.00±26.13 ^e	4.00±0.00 ^{a,b,c}	<LOD ^a	<LOD ^a	22.67±1.89 ^{b,c,d,e}	202.09±41.17 ^b	0.56±0.01 ^{c,d,e,f}
	<i>L. bo</i>	0.27±0.14 ^a	4.40±0.15 ^d	68.39±5.59 ^{a,b,c}	6.48±1.80 ^{a,b,c}	5.98±1.01 ^e	6.39±0.38 ^{d,e,f}	37.58±6.13 ^{e,f,g}	25.97±0.82 ^a	0.38±0.06 ^{a,b,c,d,e}
	<i>T. dourenensis</i>	0.32±0.09 ^a	0.52±0.32 ^{a,b,c}	11.65±16.48 ^{a,b}	1.61±0.18 ^a	4.11±0.56 ^{b,c,d,e}	2.33±0.30 ^{a,b}	2.02±0.31 ^{a,b}	6.65±0.73 ^a	0.38±0.01 ^{a,b,c,d,e}
	<i>C. apogon</i>	1.42±2.00 ^a	1.38±0.97 ^{a,b,c}	97.80±36.61 ^{b,c,d,e}	12.23±4.28 ^d	<LOD ^a	1.47±2.08 ^{a,b}	18.02±0.48 ^{a,b,c,d,e}	38.06±40.78 ^a	0.01±0.00 ^a
	<i>O. schelegelii</i>	0.31±0.07 ^a	0.78±0.55 ^{a,b,c}	73.28±14.03 ^{a,b,c}	1.88±0.51 ^a	11.76±2.92 ^f	1.31±0.55 ^{a,b}	6.38±1.18 ^{a,b,c}	19.53±0.53 ^a	0.18±0.04 ^{a,b,c}
	<i>P. macronema</i>	0.26±0.06 ^a	1.79±0.35 ^{a,b,c,d}	72.29±8.89 ^{a,b,c}	1.93±0.11 ^a	1.37±0.85 ^{a,b,c}	7.64±0.75 ^f	7.30±1.18 ^{a,b,c}	14.09±5.92 ^a	0.28±0.24 ^{a,b,c,d}
	<i>O. vittatus</i>	0.31±0.09 ^a	2.46±1.27 ^{a,b,c,d}	70.04±6.89 ^{a,b,c}	3.78±0.82 ^{a,b}	5.59±1.00 ^{d,e}	4.18±0.41 ^{b,c,d,e}	0.72±0.20 ^{a,b}	33.15±3.33 ^a	0.53±0.07 ^{b,c,d,e,f}
	<i>L. Spilopteura</i>	0.48±0.23 ^a	0.86±0.67 ^{a,b,c}	41.30±17.33 ^{a,b,c}	2.53±0.52 ^{a,b}	5.05±0.37 ^{c,d,e}	3.43±0.72 ^{b,c,d}	1.31±0.50 ^{a,b}	44.49±16.44 ^a	0.68±0.07 ^{d,e,f}
	<i>L. fasciatus</i>	1.35±1.91 ^a	3.23±0.09 ^{c,d}	87.27±61.75 ^{a,b,c,d}	9.67±1.07 ^{c,d}	5.85±0.58 ^e	<LOD ^a	14.74±10.60 ^{a,b,c,d}	<LOD ^a	0.01±0.00 ^a
	<i>P. micronemus</i>	<LOD ^a	0.78±1.11 ^{a,b,c}	52.34±5.59 ^{a,b,c}	4.51±0.89 ^{a,b,c}	1.47±1.30 ^{a,b,c}	<LOD ^a	24.84±1.43 ^{c,d,e}	38.32±3.81 ^a	0.12±0.01 ^{a,b}
	<i>M. maculatus</i>	<LOD ^a	2.83±0.16 ^{b,c,d}	46.22±2.49 ^{a,b,c}	4.18±0.99 ^{a,b,c}	1.04±0.74 ^{a,b,c}	<LOD ^a	47.20±1.69 ^{f,g,h}	<LOD ^a	0.82±0.04 ^f
	<i>R. borneensis</i>	<LOD ^a	2.22±0.39 ^{a,b,c,d}	28.39±1.12 ^{a,b,c}	7.69±1.55 ^{b,c,d}	1.95±0.87 ^{a,b,c,d,e}	<LOD ^a	66.88±4.81 ^h	<LOD ^a	0.31±0.04 ^{a,b,c,d}
	<i>A. armatus</i>	<LOD ^a	2.53±0.44 ^{a,b,c,d}	30.11±4.74 ^{a,b,c}	2.32±0.33 ^{a,b}	1.74±0.63 ^{a,b,c,d}	<LOD ^a	11.54±8.22 ^{a,b,c,d}	<LOD ^a	0.77±0.03 ^{e,f}
	<i>C. lucius</i>	<LOD ^a	<LOD ^a	33.29±1.78 ^{a,b,c}	3.74±0.21 ^{a,b,c}	1.20±0.58 ^{a,b,c}	<LOD ^a	<LOD ^a	<LOD ^a	1.46±0.07 ^g

Table 4.10: Continued
(c)

Part	Species	Elements (dry weight, mg/kg)								
		Cr	Cu	Fe	Mn	Ni	Pb	Zn	Al	Hg
Gills	<i>O. anomalura</i>	<LOD ^a	<LOD ^a	164.29±23.12 ^{a,b}	50.84±30.81 ^a	1.76±0.78 ^{a,b}	6.24±0.24 ^{b,c,d,e}	170.25±60.00 ^d	54.24±15.47 ^a	0.14±0.04 ^{a,b,c}
	<i>L. setigerum</i>	3.48±1.75 ^a	1.52±1.23 ^{a,b}	1427.54±210.32 ^e	55.20±7.89 ^a	1.57±1.36 ^{a,b}	7.78±1.11 ^{d,e}	153.75±5.02 ^{c,d}	2202.29±808.11 ^c	0.07±0.01 ^{a,b}
	<i>L. micropogon</i>	1.54±0.34 ^a	1.737±1.22 ^{a,b}	601.55±65.90 ^{a,b,c,d}	121.88±22.34 ^{a,b}	0.88±0.64 ^a	6.96±0.73 ^{c,d,e}	58.84±9.51 ^{a,b}	707.00±25.83 ^{a,b}	0.38±0.13 ^{b,c}
	<i>B. collingwoodii</i>	1.57±2.23 ^a	<LOD ^a	271.21±54.73 ^{a,b,c}	68.23±3.10 ^a	<LOD ^a	2.85±2.02 ^{a,b,c}	88.34±3.95 ^{a,b,c}	343.09±146.38 ^a	0.07±0.02 ^{a,b}
	<i>H. bimaculata</i>	2.18±1.26 ^a	0.50±0.00 ^{a,b}	411.87±32.65 ^{a,b,c}	52.20±1.94 ^a	2.47±1.65 ^{a,b}	6.86±0.07 ^{c,d,e}	137.74±9.07 ^{a,b,c}	673.98±64.68 ^{a,b}	0.38±0.04 ^{b,c}
	<i>H. wyckii</i>	0.78±0.17 ^a	<LOD ^a	190.23±71.76	27.06±2.17 ^a	2.16±1.64 ^{a,b}	6.50±0.32 ^{b,c,d,e}	58.01±7.60 ^{a,b}	160.15±88.40 ^a	0.44±0.08 ^c
	<i>H. planiceps</i>	4.56±2.55 ^a	2.78±0.31 ^{a,b}	1111.88±19.29 ^{d,e}	78.74±6.31 ^a	0.76±0.65 ^a	10.08±0.74	76.17±4.55 ^{a,b,c}	1782.43±374.67 ^{b,c}	0.39±0.01 ^{b,c}
	<i>B. schwanenfeldii</i>	3.97±2.84 ^a	0.73±1.03 ^{a,b}	372.97±68.24 ^{a,b,c}	46.82±4.27 ^a	<LOD ^a	3.42±2.49 ^{a,b,c,d}	71.50±4.99 ^{a,b,c}	458.03±111.71 ^a	0.07±0.04 ^{a,b}
	<i>T. tambroides</i>	11.57±1.42 ^b	6.21±0.88 ^c	4697.84±252.41 ^f	278.35±98.38 ^c	<LOD ^a	9.26±0.92	79.25±5.74 ^{a,b,c}	6408.36±867.51 ^d	0.43±0.25 ^c
	<i>L. bo</i>	4.99±1.04 ^a	2.28±1.20 ^{a,b}	815.59±93.13 ^{c,d}	194.34±21.71 ^{b,c}	1.42±1.42 ^{a,b}	6.18±0.91 ^{b,c,d,e}	121.61±11.99 ^{a,b,c}	1144.33±294.02 ^{a,b,c}	0.07±0.05 ^{a,b}
	<i>T. dourenensis</i>	1.58±0.52 ^a	1.96±0.22 ^{a,b}	590.10±154.09 ^{a,b,c,d}	75.76±7.74 ^a	2.33±1.20 ^{a,b}	6.33±0.72 ^{b,c,d,e}	138.31±19.44 ^{a,b,c}	591.73±39.77 ^{a,b}	0.18±0.14 ^{a,b,c}
	<i>C. apogon</i>	2.53±2.53 ^a	<LOD ^a	373.38±1.21 ^{a,b,c}	239.86±0.56 ^c	<LOD ^a	1.97±1.97 ^{a,b}	143.17±0.35 ^{b,c,d}	442.75±19.48 ^a	<LOD ^a
	<i>O. schelegelii</i>	1.94±0.56 ^a	0.60±0.44 ^{a,b}	423.73±33.09 ^{a,b,c}	29.56±9.71 ^a	2.41±0.38 ^{a,b}	7.80±0.43 ^{d,e}	58.45±13.06 ^{a,b}	560.02±55.12 ^{a,b}	0.09±0.07 ^{a,b,c}
	<i>P. macronema</i>	1.01±0.08 ^a	0.93±0.71 ^{a,b}	251.13±39.20 ^{a,b,c}	85.74±3.75 ^a	2.59±0.37 ^{a,b}	7.83±0.64 ^{d,e}	80.52±4.72 ^{a,b,c}	266.54±112.18 ^a	0.05±0.04 ^{a,b}
	<i>O. vittatus</i>	3.17±1.13 ^a	1.96±1.41 ^{a,b}	693.17±114.20 ^{b,c,d}	56.56±21.39 ^a	1.65±0.89 ^{a,b}	7.69±1.23 ^{d,e}	77.18±0.59 ^{a,b,c}	1006.61±257.03 ^{a,b,c}	0.13±0.06 ^{a,b,c}
	<i>L. Spilopteura</i>	1.45±0.30 ^a	1.39±0.56 ^{a,b}	565.90±157.46 ^{a,b,c,d}	43.86±2.71 ^a	1.53±0.40 ^{a,b}	7.58±0.86 ^{d,e}	155.63±13.37 ^{c,d}	756.11±267.81 ^{a,b}	0.17±0.14 ^{a,b,c}
	<i>L. fasciatus</i>	1.62±2.29 ^a	3.36±0.58 ^b	839.38±506.24 ^{c,d,e}	97.42±6.08 ^{a,b}	7.05±0.17 ^c	<LOD ^a	41.10±1.67 ^a	551.85±22.41 ^{a,b}	<LOD ^a
	<i>P. micronemus</i>	<LOD ^a	<LOD ^a	274.09±42.24 ^{a,b,c}	61.60±3.02 ^a	<LOD ^a	<LOD ^a	74.32±5.13 ^{a,b,c}	375.12±106.01 ^a	0.09±0.01 ^{a,b,c}
	<i>M. maculatus</i>	<LOD ^a	<LOD ^a	22.26±0.23 ^a	91.83±4.21 ^a	2.05±0.00 ^{a,b}	3.25±0.43 ^{a,b,c}	82.21±0.00 ^{a,b,c}	22.38±0.01 ^a	0.17±0.00 ^{a,b,c}
	<i>R. borneensis</i>	<LOD ^a	<LOD ^a	46.73±3.90 ^a	52.31±3.28 ^a	4.65±1.35 ^{a,b,c}	4.22±0.92 ^{a,b,c,d}	207.41±8.25	9.18±9.81 ^a	0.24±0.07 ^{a,b,c}
	<i>A. armatus</i>	<LOD ^a	1.00±0.01 ^{a,b}	545.47±118.61 ^{a,b,c,d}	69.68±5.41 ^a	0.69±0.69 ^a	<LOD ^a	76.94±3.58 ^{a,b,c}	700.59±184.86 ^{a,b}	0.11±0.05 ^{a,b,c}
	<i>C. lucius</i>	<LOD ^a	<LOD ^a	124.77±2.71 ^{a,b}	262.01±8.90 ^c	5.03±0.94 ^{b,c}	3.55±0.38 ^{a,b,c,d}	85.16±3.52 ^{a,b,c}	46.86±8.87 ^a	0.26±0.01 ^{a,b,c}

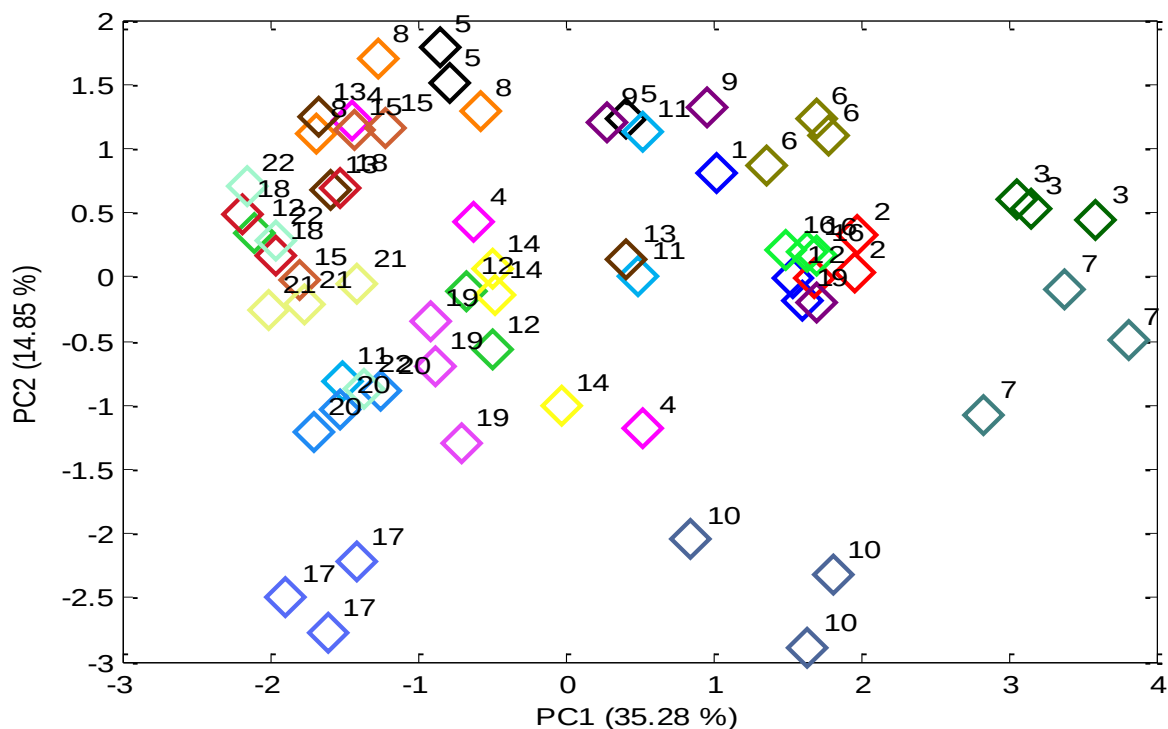
Different letters indicate significant differences ($P<0.05$)

The metal concentrations in muscle, belly and gills were examined separately using PCA to demonstrate the clustering pattern according to species. Figure 4.5(a), (b) and (c) illustrate the scores plot according to parts and different colour indicates different species. Noticeably, the distribution of metals in muscle of *Hemibagrus planiceps* (F7), *Leiocassis micropogon* (F3), *Lobocheilos bo* (F10) and *Labiobarbus fasciatus* (F17) are distinguishable from other species. For belly, *Hemibagrus planiceps* (F7) and *Leiocassis micropogon* (F3) occupy the bottom right quadrant of the scores plot, evidently discernible from other species. For metal contents in gills, *Hemibagrus planiceps* (F7) and *Tor tambroides* (F9) are separable from other species. The metals content in muscle, belly and gills of varying species were compared across all elements, respectively; statistical difference was consistently established ($P < 0.05$), except Cr in muscle and belly and Ni in gills.

Hemibagrus planiceps is a typical omnivorous bottom feeder found in the river basins with sandy or rocky bottom (Ng & Kottelet, 2013). It accumulates the highest concentrations of Al in its muscle with statistical difference from other species, at an average concentration of 181.50 mg/kg dw (Figure 4.6a (i)). *Lobocheilos bo* on the other hand is a herbivorous bottom feeder that mainly feed on algae and aquatic plants (Kottelat & Tan, 2008). It demonstrates the highest concentration of Zn (114.67 mg/kg dw) in the muscle (Figure 4.6b (i)). *Leiocassis micropogon*, a carnivorous bottom feeder is found to accumulate the highest Al in its belly at 364.81 mg/kg dw (Figure 4.6c (i)). According to Ahmad *et al.* (2015), the bioaccumulation rate of bottom feeders (demersal) is usually higher than fishes live in the upper layer of river (pelagic species). These elements from sediment are easily taken by demersal species due to close relationship with sediment. However, Topping (1973) suggested that plankton feeding fish contained higher concentrations of Zn and Pb than bottom feeding fish. Therefore, metal

accumulation of fish is greatly influenced by its feeding habit. *Labiobarbus fasciatus*, a bentopelagic carnivorous species that mainly feeds on aquatic insects and invertebrates demonstrated the highest concentration of Ni in the muscle, 8.71 mg/kg (Figure 4.6d (i)). Al-Busaidi *et al.* (2011) suggested that high heavy metal accumulation in carnivorous species is due to their feeding at higher trophic levels. In this study, *Hampala bimaculata*, an omnivorous pelagic species, demonstrates the highest Hg in muscle and belly at an average of 3.59 mg/kg and 3.63 mg/kg dw, respectively (Figure 4.6e (i) & (ii)). It is widely reported that Hg is often higher in the predatory fish, as larger organisms exhibit higher contaminant level compared to organisms at the bottom of the food chains (Hosseini *et al.*, 2013). Besides the feeding habit, the metal accumulation in fish is further complicated by other factors including the exposure and availability of food (Florence 1982; Henry *et al.* 2004). *Tor tambroides*, a typical omnivore tends to accumulate metals in its gills which include Cr (11.57 mg/kg), Fe (4697.84 mg/kg) and Al (6408.36 mg/kg) in dw (Figure 4.6F (i)). According to the study by Ahmad *et al.* (2015), *Tor tambroides* always show higher ventilation of gills compare to other fish species especially under stress condition, for example high total solid suspended (TSS) environment. As sediment begins to accumulate in the gill filaments, *Tor tambroides* could excessively open and close their gills and this may consequently accumulate higher metals concentration. High Al concentration in the aquatic environment may cause the clogging of gills, especially the Al in the form of low molecular weight (Oughton *et al.*, 1992). However, low risk of toxicity is found despite high level of Al at pH 6.5 to 7.5 as Al tend to form complexes with organic ligands hence less bioavailable (Baker & Schofield, 1980; Bertsch, 1990).

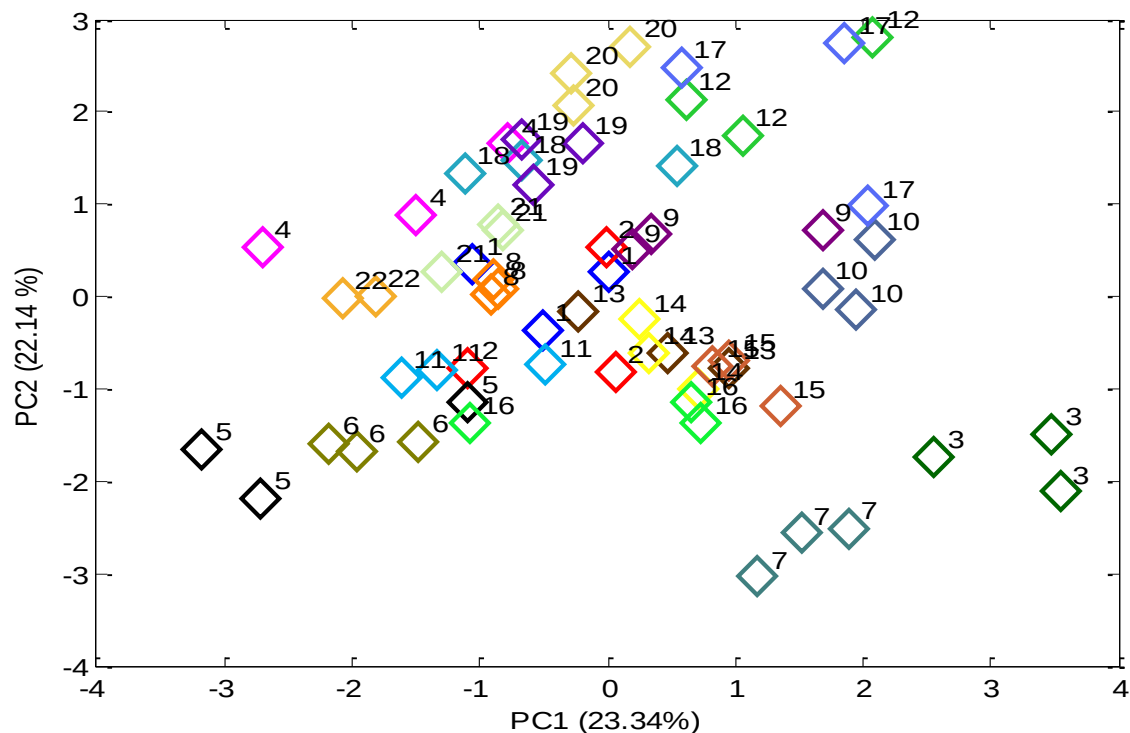
(a)



No	Species	No	Species
1	<i>Oxygaster anomalura</i>	12	<i>Cyclocheilichthys apogon</i>
2	<i>Luciosoma setigerum</i>	13	<i>Osteochilus schlegelii</i>
3	<i>Leiocassis micropogon</i>	14	<i>Pengasius macronema</i>
4	<i>Barbonymus collingwoodii</i>	15	<i>Osteochilus vittatus</i>
5	<i>Hampala bimaculata</i>	16	<i>Luciosoma spiloptera</i>
6	<i>Hemibagrus wyckii</i>	17	<i>Labiobarbus fasciatus</i>
7	<i>Hemibagrus planiceps</i>	18	<i>Pseudolais micronemus</i>
8	<i>Barbonymus schwanenfeldii</i>	19	<i>Macrognaathus maculatus</i>
9	<i>Tor tambroides</i>	20	<i>Rasbora borneensis</i>
10	<i>Lobocheilos bo</i>	21	<i>Anematichthys armatus</i>
11	<i>Tor dourenensis</i>	22	<i>Channa lucius</i>

Figure 4.5: The scores plot of PC2 vs PC1 for heavy metals in (a) muscle, (b) belly and (c) gills for different species

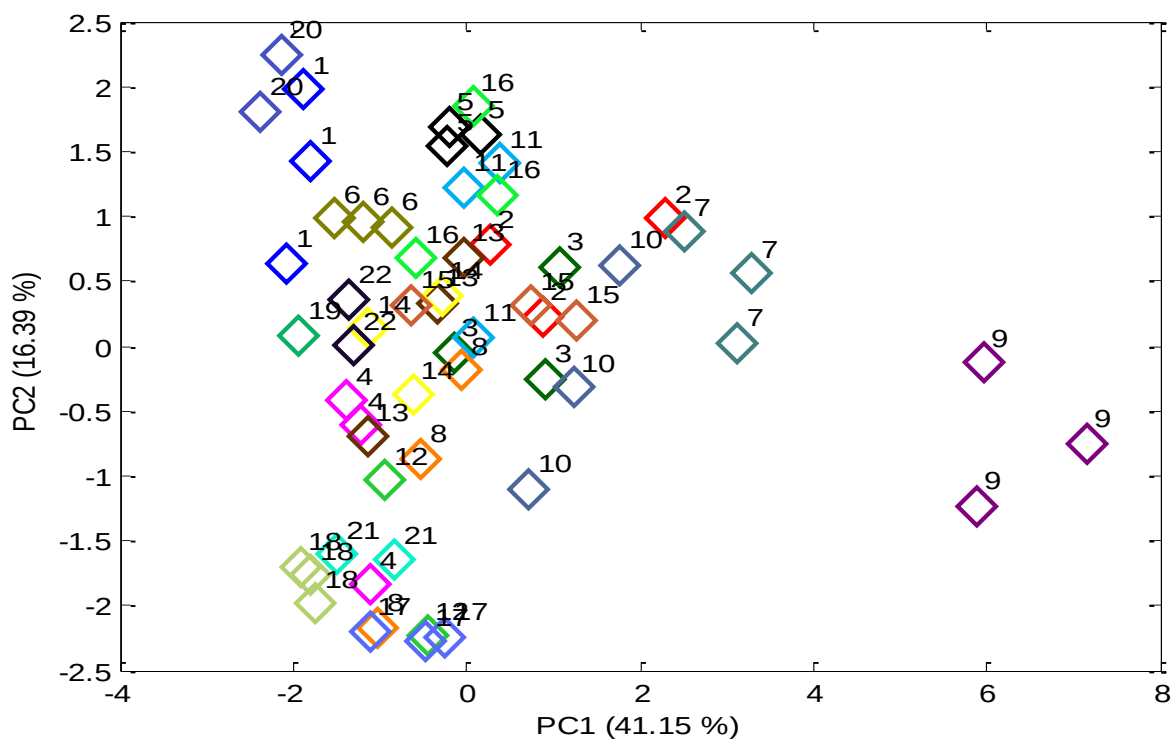
(b)



No	Species	No	Species
1	<i>Oxygaster anomalura</i>	12	<i>Cyclocheilichthys apogon</i>
2	<i>Luciosoma setigerum</i>	13	<i>Osteochilus schlegelii</i>
3	<i>Leiocassis micropogon</i>	14	<i>Pengasius macronema</i>
4	<i>Barbonymus collingwoodii</i>	15	<i>Osteochilus vittatus</i>
5	<i>Hampala bimaculata</i>	16	<i>Luciosoma spiloptera</i>
6	<i>Hemibagrus wyckii</i>	17	<i>Labiobarbus fasciatus</i>
7	<i>Hemibagrus planiceps</i>	18	<i>Pseudolais micronemus</i>
8	<i>Barbonymus schwanenfeldii</i>	19	<i>Macrogathus maculatus</i>
9	<i>Tor tambroides</i>	20	<i>Rasbora borneensis</i>
10	<i>Lobocheilos bo</i>	21	<i>Anematichthys armatus</i>
11	<i>Tor dourenensis</i>	22	<i>Channa lucius</i>

Figure 4.5: Continued

(c)

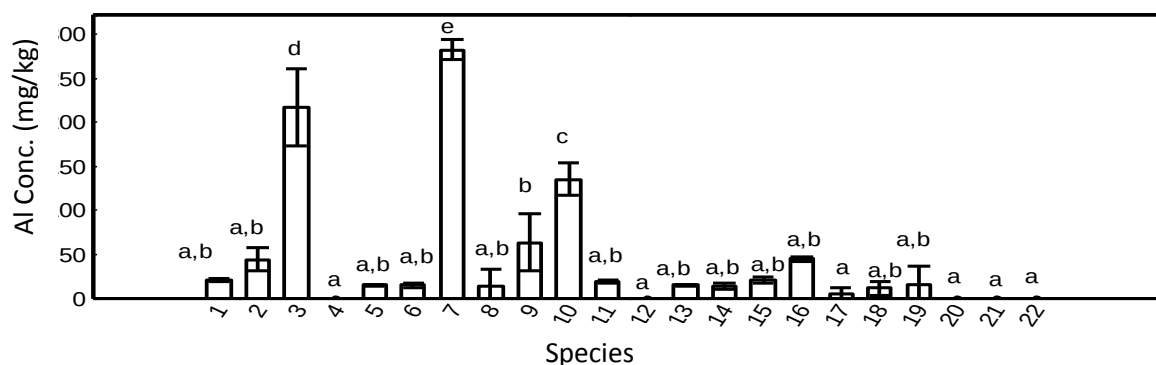


No	Species	No	Species
1	<i>Oxygaster anomalura</i>	12	<i>Cyclocheilichthys apogon</i>
2	<i>Luciosoma setigerum</i>	13	<i>Osteochilus schlegelii</i>
3	<i>Leiocassis micropogon</i>	14	<i>Pengasius macronema</i>
4	<i>Barbonymus collingwoodii</i>	15	<i>Osteochilus vittatus</i>
5	<i>Hampala bimaculata</i>	16	<i>Luciosoma spiloptera</i>
6	<i>Hemibagrus wyckii</i>	17	<i>Labiobarbus fasciatus</i>
7	<i>Hemibagrus planiceps</i>	18	<i>Pseudolais micronemus</i>
8	<i>Barbonymus schwanenfeldii</i>	19	<i>Macrognaathus maculatus</i>
9	<i>Tor tambroides</i>	20	<i>Rasbora borneensis</i>
10	<i>Lobocheilos bo</i>	21	<i>Anematichthys armatus</i>
11	<i>Tor dourenensis</i>	22	<i>Channa lucius</i>

Figure 4.5: Continued

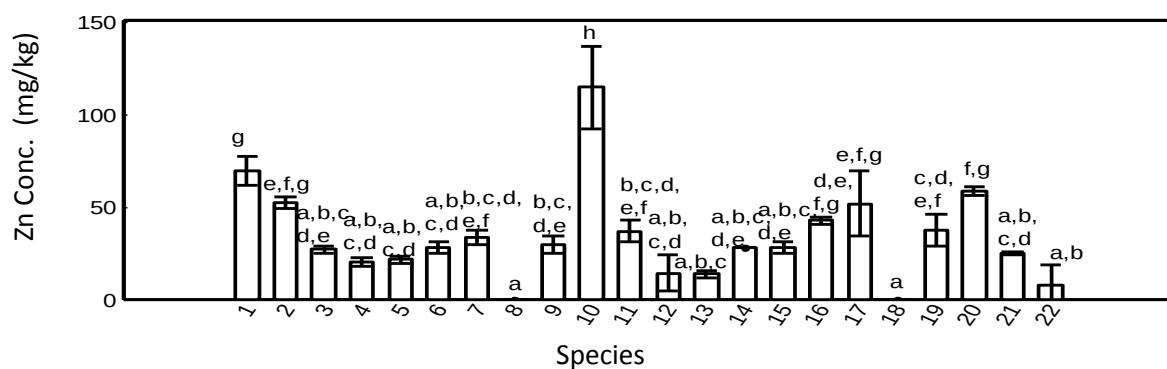
(a) *Hemibagrus planiceps*

(i) Muscle



(b) *Lobocheilos bo*

(i) Muscle



(c) *Leiocassis micropogon*

(i) Belly

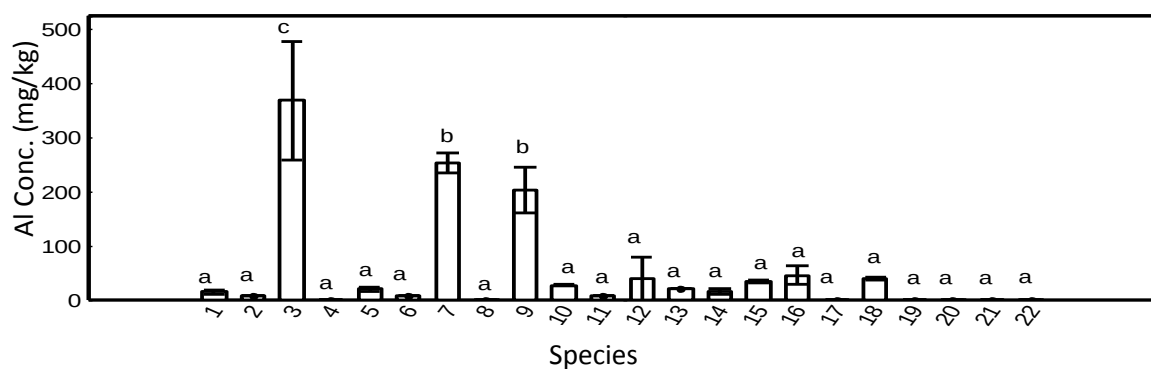
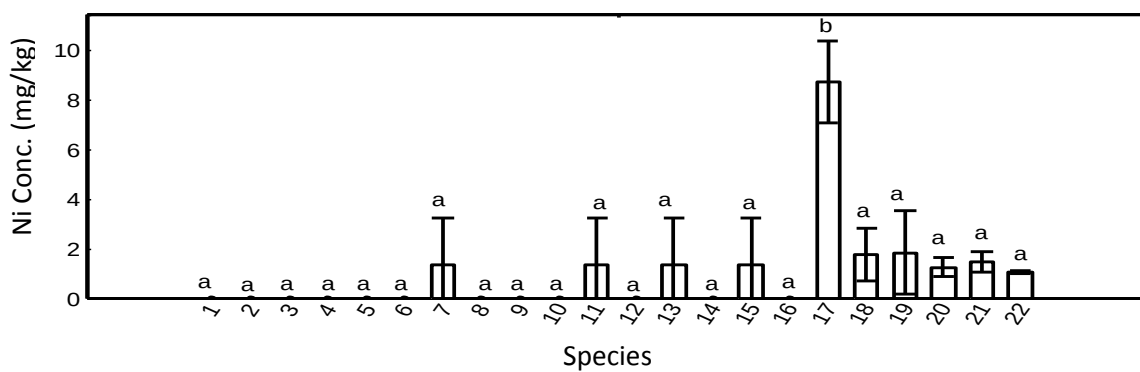


Figure 4.6: The concentrations of heavy metals in different part of fishes

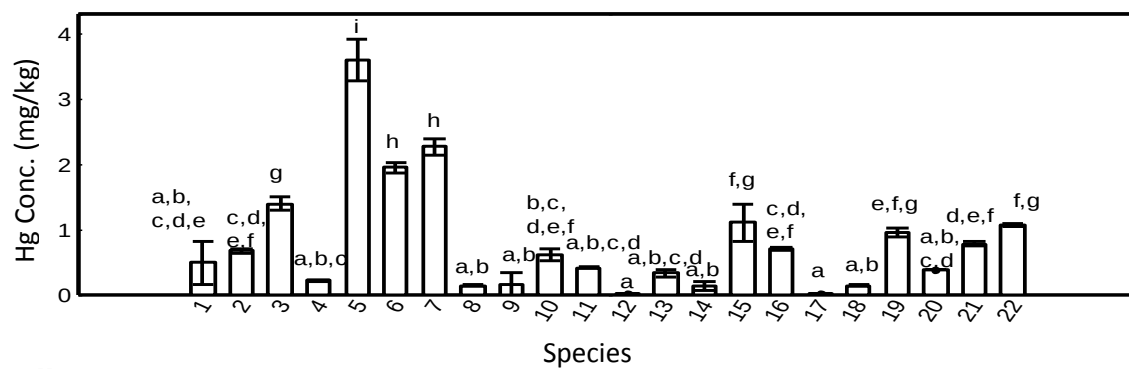
(d) *Labiobarbus fasciatus*

(i) Muscle



(e) *Hampala bimaculata*

(i) Muscle



(ii) Belly

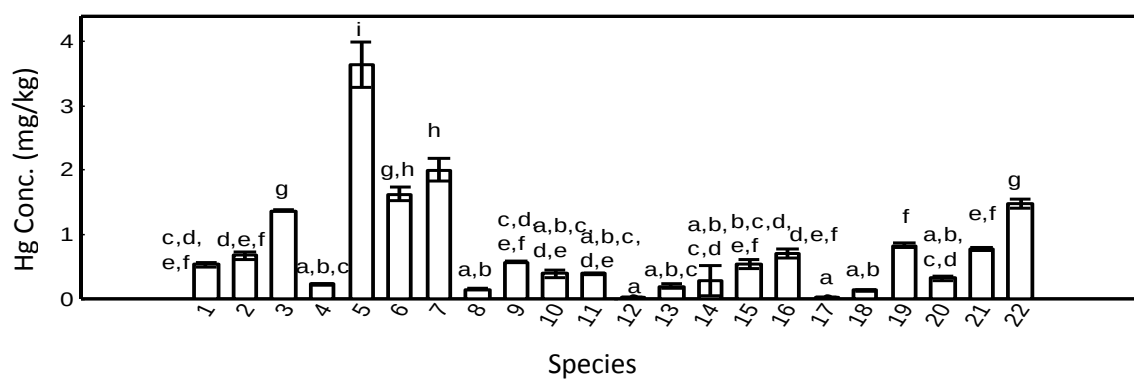
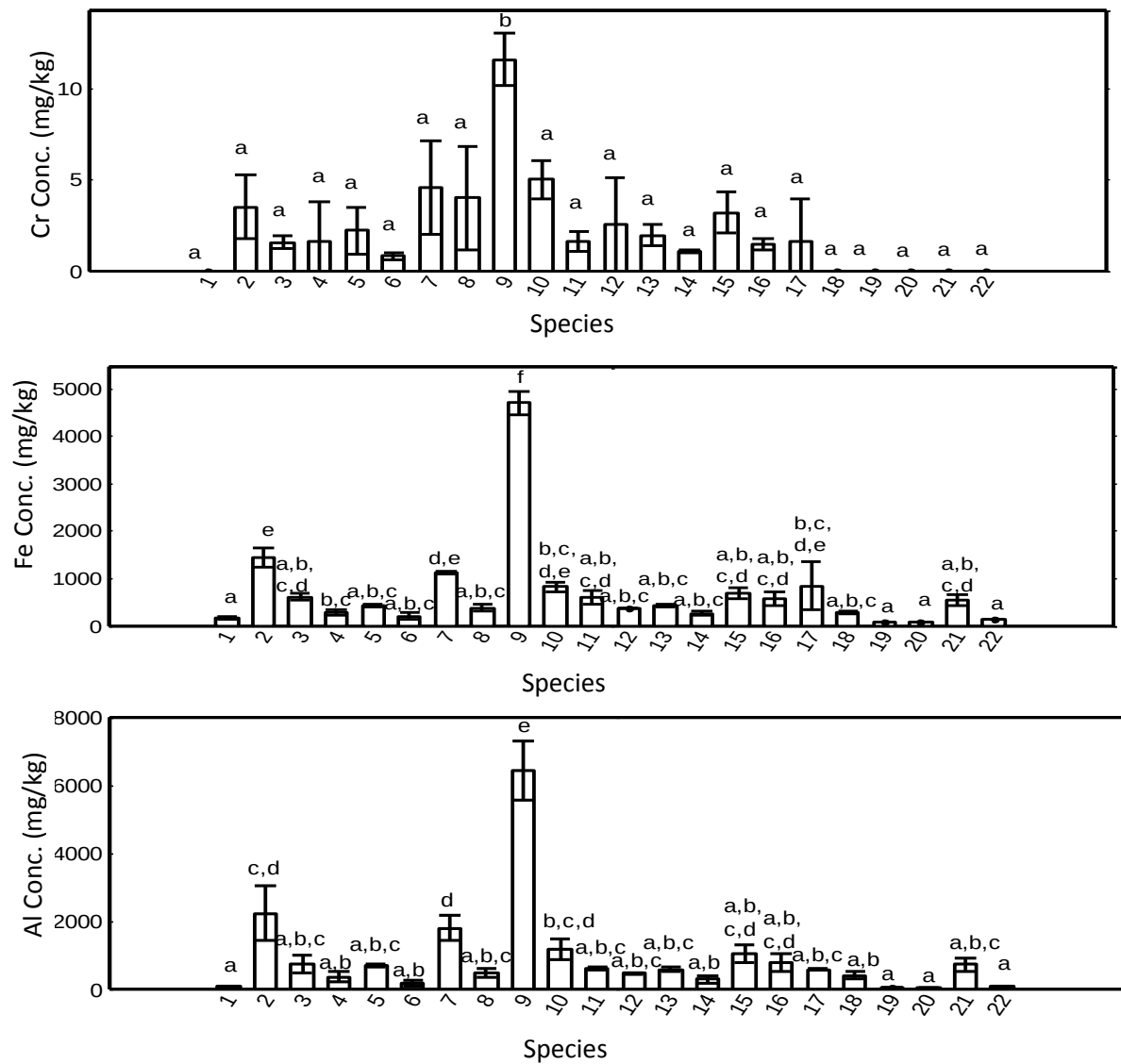


Figure 4.6: Continued

- (f) *Tor tambroides*
(i) Gills



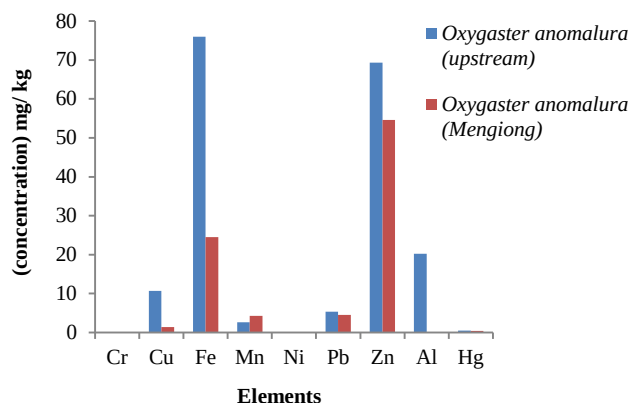
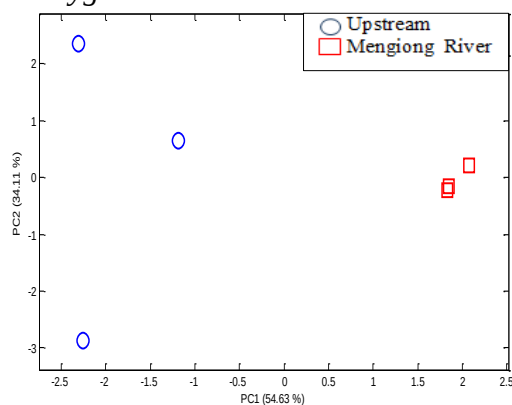
Different letters between species indicate significant differences ($p < 0.05$)

Figure 4.6: Continued

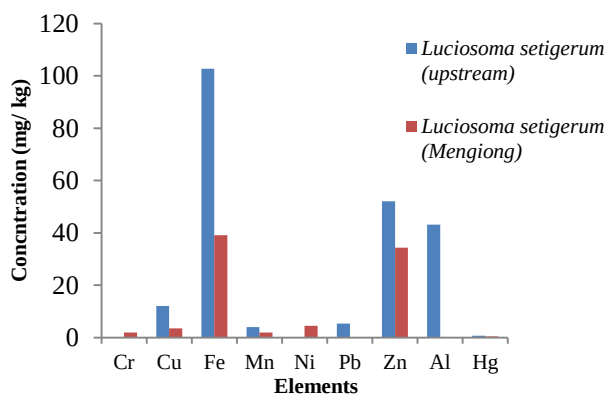
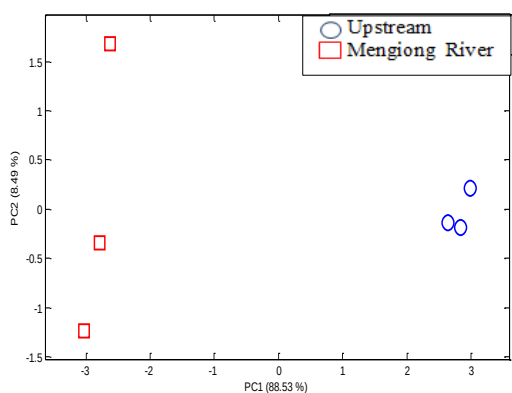
4.2.3 Distribution of metals in fish from upstream and downstream of Baleh River and Mengiong River

The sampling stations are mainly divided into upstream, downstream and mid stream at the Mengiong River. Five similar species were found in upstream and mid-stream of the Baleh River (*Oxygaster anomalura*, *Luciosoma setigerum*, *Hampala bimaculata*, *Tor dourenensis* and *Cyclocheilichthys apogon*); *Hemibagrus planiceps* was the only species found in upstream, mid stream and downstream. The metal concentrations found in fishes caught from different stretches of the river were analysed and compared. PCA was employed to demonstrate the clustering pattern of metal concentrations in fishes according to locations. Noticeably, all species of fish caught from different area of the river are distinguishable. *Oxygaster anomalura*, *Luciosoma setigerum*, *Hampala bimaculata*, *Tor dourenensis*, *Cyclocheilichthys apogon* and *Hemibagrus planiceps* from upstream demonstrated higher Fe, Zn and Al, $P < 0.05$, except Fe in *Tor dourenensis* and *Cyclocheilichthys apogon* from Mengiong River significant higher ($P < 0.05$); no significant difference is found for other elements (Figure 4.7). The higher Al and Fe accumulation in fishes from upstream correspond to the higher concentrations in water. Observably, the upstream of the Baleh River is exposed to intensive logging activity with turbid water. Elevated Al and Fe were detected in the river water and consequently metals content in fish were higher compared to other areas. On the other hand, Fe and Zn were found higher in *Oxygaster anomalura*, *Tor dourenensis* and *Cyclocheilichthys apogon* caught at the Mengiong River possibly due to the wet season during sampling. A few studies reported that fishes tend to accumulate higher heavy metals during wet season (Olowoyo, 2010; Gbaruko & Friday, 2007). According to Igwegbe *et al.* (2015), the increases of surface runoff during wet season have increased the dissolution of metals into the water, which in turn, increases the metal uptakes by fishes.

i. *Oxygaster anomalura*



ii. *Luciosoma setigerum*



iii. *Hampala bimaculata*

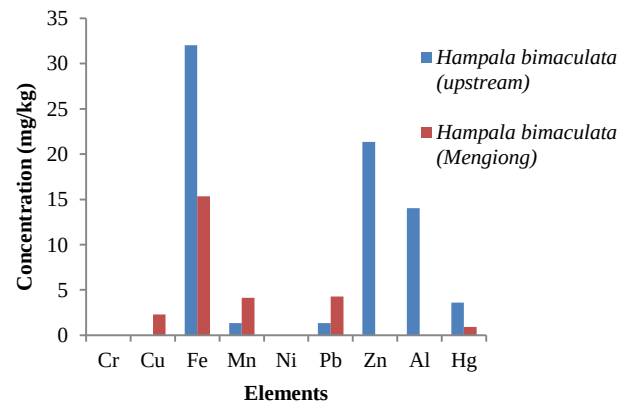
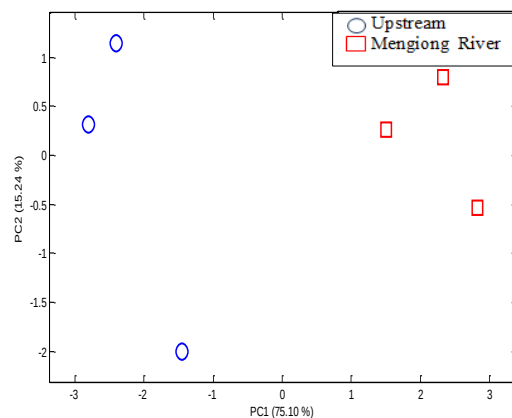
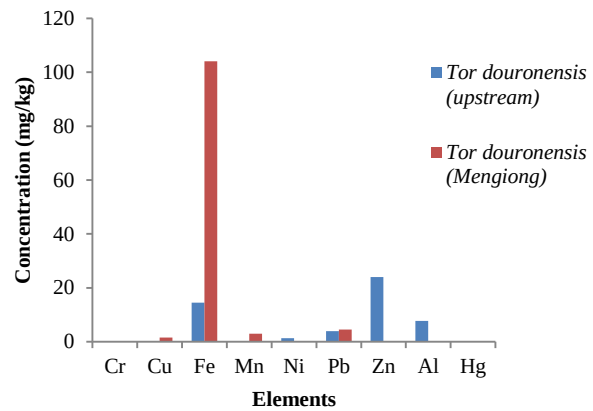
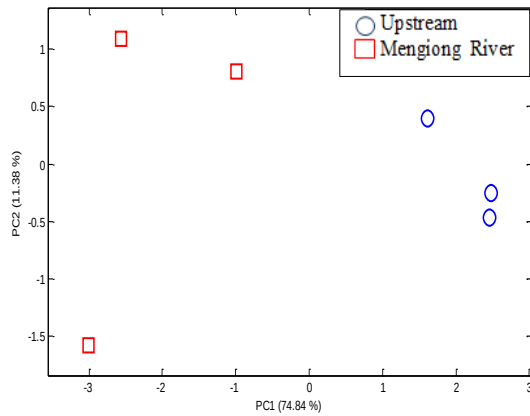
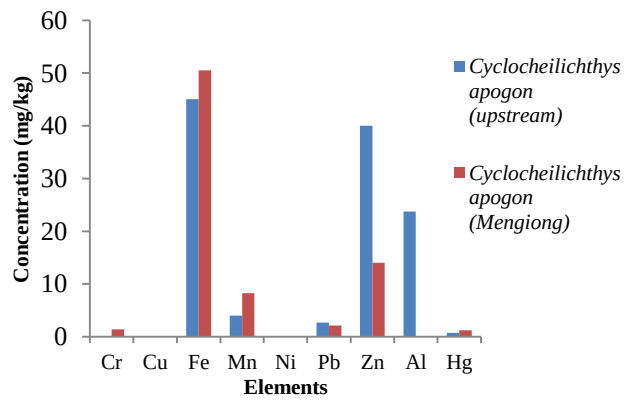
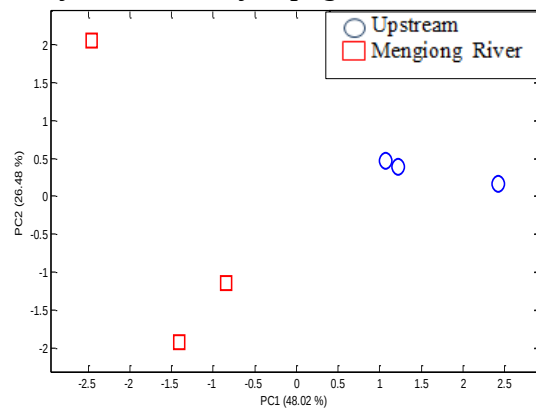


Figure 4.7: The scores plot of PC2 vs PC1 for heavy metals in fishes caught from upstream, mid-stream and downstream with the bar chart illustrating the metal distributions

iv. *Tor douronensis*



v. *Cyclocheilichthys apogon*



vi. *Hemibagrus planiceps*

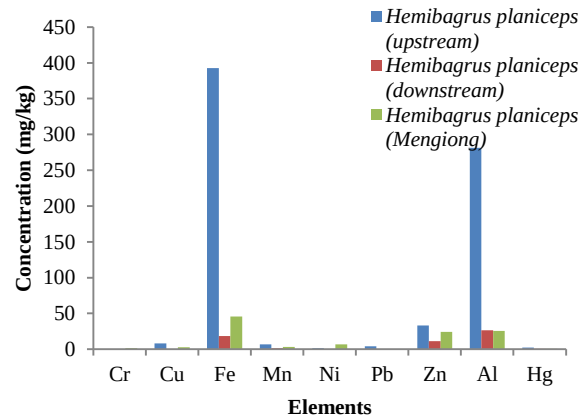
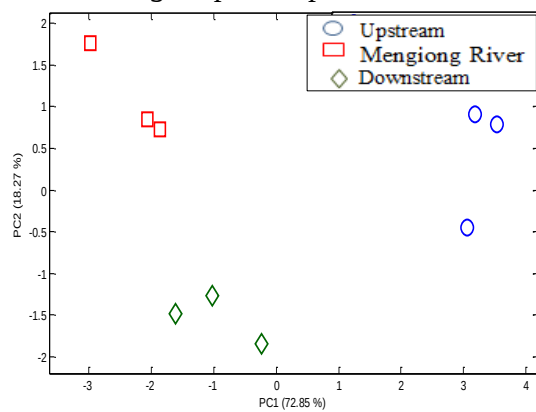


Figure 4.7: Continued

4.2.4 Comparison of the metals content against the guidelines

In this study, the metals are reported in dry weight for consistencies. For comparison with the stipulated guideline, the metals in wet weight are estimated based on the moisture content.

The moisture content in fish ranges from 77.38 % to 84.54 % (Table 4.11).

Table 4.11: Moisture content (%) of 22 species of fish

Fish species	Moisture content (%)	Fish species	Moisture content (%)
<i>Oxygaster anomalura</i>	82.00	<i>Cyclocheilichthys apogon</i>	80.00
<i>Luciosoma setigerum</i>	80.35	<i>Osteochilus schlegelii</i>	79.59
<i>Leiocassis micropogon</i>	80.34	<i>Pengasius macronema</i>	82.17
<i>Barbonymus collingwoodii</i>	79.83	<i>Osteochilus vittatus</i>	78.15
<i>Hampala bimaculata</i>	83.56	<i>Luciosoma spilopteura</i>	80.57
<i>Hemibagrus wyckii</i>	78.10	<i>Labiobarbus fasciatus</i>	77.00
<i>Hemibagrus planiceps</i>	80.90	<i>Pseudolais micronemus</i>	84.54
<i>Barbonymus schwanenfeldii</i>	81.00	<i>Macragnathus maculatus</i>	80.41
<i>Tor tambroides</i>	77.38	<i>Rasbora borneensis</i>	83.72
<i>Lobocheilos bo</i>	78.90	<i>Anematichthys armatus</i>	81.81
<i>Tor dourenensis</i>	79.92	<i>Channa lucius</i>	81.44

This strategy of comparison leads to uncertainties whether concentrations in dry weight and estimated wet weight are comparable, especially for volatile elements such as Hg. In order to demonstrate that the estimated wet and dry weight concentrations are comparable, three selected species (*Tor douronensis*, *Tor tambroides* and *Lobocheilos bo*) were digested in wet and dry weight for analysis. The estimated wet weight were then calculated and compared with the actual wet weight concentration. The metals contents in estimated wet weight and actual wet weight are tabulated in Table 4.12; statistically, there are no significant different for all the elements, except Zn in *Tor douronensis*. As suggested by Adrian and Steven (1979), dry weight is more applicable as it is difficult to attain consistent weight of tissues due to the presence of 60 to 80 % of moisture (Adrian & Stevens, 1979).

Table 4.12: Heavy metals content in estimated wet weight and actual wet weight based on moisture content

Element		Concentration (mg/kg)		
		<i>Tor douronensis</i>	<i>Tor tambroides</i>	<i>Lobocheilos bo</i>
Al	Estimated	<LOD	<LOD	8.36±7.24
	Actual	<LOD	<LOD	6.72±11.64
Cr	Estimated	<LOD	<LOD	<LOD
	Actual	<LOD	<LOD	<LOD
Cu	Estimated	<LOD	<LOD	<LOD
	Actual	<LOD	<LOD	<LOD
Fe	Estimated	<LOD	1.74±3.01	<LOD
	Actual	<LOD	3.14±5.44	3.82±6.62
Mn	Estimated	0.03±0.05	0.03±0.05	<LOD
	Actual	<LOD	<LOD	0.74±0.64
Ni	Estimated	<LOD	<LOD	<LOD
	Actual	1.08±1.88	<LOD	<LOD
Pb	Estimated	<LOD	<LOD	<LOD
	Actual	<LOD	<LOD	<LOD
Zn	Estimated	<LOD	11.32±9.80	<LOD
	Actual	17.90±1.66	13.98±12.66	<LOD
Hg	Estimated	0.15±0.02	0.28±0.02	0.14±0.01
	Actual	0.12±0.03	0.25±0.01	0.15±0.01

<LOD: below detection limit

The concentrations in estimated wet weight (ww) are compared against the guideline of Food and Agriculture Organization (FAO) (1983) and Malaysia Food Act (MFA) (1983) (Table 4.13). *Oxygaster anomalura*, *Luciosoma setigerum*, *Leiocasis micropogon*, *Hemibagrus wyckii*, *Hemibagrus planiceps*, *Barbonymus schwanenfeldii*, *Tor tambroides*, *Tor douronensis*, *Osteochilus schlegelii* and *Luciosoma spilopteura* were found with concentrations of Pb higher than the recommended level of FAO, 0.5 mg/kg (ww) (LOM, 2006). However, the value reported is lower than permissible level of MFA, 2.0 mg/kg and some literature findings that range from not detected to 18.5 mg/kg (ww) (Mwashote, 2003; Hasyimah *et al.*, 2011). Overdose of Pb has been associated to disorder of central nervous system and kidney failure affecting the central information processing system, causing

learning disabilities, poor muscle coordination and hearing damage (ATSDR, 2008). Al in *Oxygaster anomalura*, *Luciosoma setigerum*, *Leiocassis micropogon*, *Hampala bimaculata*, *Hemibagrus wyckii*, *Hemibagrus planiceps*, *Barbonymus schwanenfeldii*, *Tor tambroides*, *Lobocheilos bo*, *Tor dourenensis*, *Osteochilus schlegelii*, *Pengasius macronema*, *Osteochilus vittatus*, *Luciosoma spilopteura* and *Macrognathus maculatus* were higher than 2.0 mg/kg however, the value reported is lower than some literature findings that ranges from 132.9 mg/kg to 320.6 mg/kg in wet weight (Merciai *et al.*, 2014; Ismail & Saleh, 2012). Higher concentration of Al in body has been associated with brain and bone disease, for example, Alzheimer disease (ATSDR, 2008). Hg is detected in *Hampala bimaculata* at an average of 0.57 mg/kg ww, slightly exceeding the permissible level of 0.5 mg/kg. Nevertheless, this value is lower compared to the findings of Ahmad *et al.* (2015) that ranges from not detected to 2.7 mg/kg in wet weight. The toxicity of Hg is associated to the central nervous system including loss of coordination, blurred vision or blindness, and hearing and speech impairment. Other elements are comfortably below the permissible level of FAO and MFA.

Table 4.13: Comparison of the metals content in fish muscle (in wet weight) with the guideline

Elements (mg/kg)		Cr		Cu		Pb		Zn		Al		Hg	
Fish species	Guidelines	FAO 1	MFA -	FAO 30	MFA 30	FAO 0.5	MFA 2	FAO 30	MFA 100	FAO 2	MFA 2	FAO 0.5	MFA 0.5
<i>Oxygaster anomalura</i>		<LOD		2.24		1.12		14.56		4.25		0.10	
<i>Luciosoma setigerum</i>		<LOD		2.40		1.07		10.40		8.63		0.13	
<i>Leiocasis micropogon</i>		<LOD		2.40		1.6		5.33		43.31		0.28	
<i>Barbonymus collingwoodii</i>		0.29		0.54		0.23		4.03		<LOD		0.04	
<i>Hampala bimaculata</i>		<LOD		<LOD		0.21		3.41		2.25		0.57	
<i>Hemibagrus wyckii</i>		<LOD		2.64		1.47		6.16		3.17		0.43	
<i>Hemibagrus planiceps</i>		<LOD		1.52		0.76		6.33		53.49		0.43	
<i>Barbonymus schwanenfeldii</i>		0.28		0.17		0.51		<LOD		2.72		0.03	
<i>Tor tambroides</i>		<LOD		2.15		0.92		6.75		14.42		0.03	
<i>Lobocheilos bo</i>		<LOD		0.28		<LOD		24.08		28.18		0.13	
<i>Tor dourenensis</i>		<LOD		0.27		0.53		7.33		3.53		0.08	
<i>Cyclocheilichthys apogon</i>		<LOD		<LOD		0.43		2.8		<LOD		<LOD	
<i>Osteochilus schlegelii</i>		<LOD		<LOD		0.53		2.67		2.87		0.06	
<i>Pengasius macronema</i>		<LOD		<LOD		<LOD		5.04		2.37		0.02	
<i>Osteochilus vittatus</i>		<LOD		<LOD		<LOD		6.16		4.45		0.24	
<i>Luciosoma spiloptera</i>		<LOD		2.45		0.92		9.81		10.09		0.16	
<i>Labiobarbus fasciatus</i>		<LOD		0.74		<LOD		11.86		1.02		<LOD	
<i>Pseudolais micronemus</i>		<LOD		0.32		<LOD		<LOD		1.59		0.02	
<i>Macrognathus maculatus</i>		<LOD		0.44		<LOD		7.50		3.03		0.19	
<i>Rasbora borneensis</i>		<LOD		0.11		<LOD		9.37		<LOD		0.06	
<i>Anematichthys armatus</i>		<LOD		0.12		<LOD		4.49		<LOD		0.14	
<i>Channa lucius</i>		<LOD		<LOD		<LOD		1.43		<LOD		0.20	

FAO: Food and Agriculture Organization (1983)

MFA: Malaysia Food Act (1983)

<LOD: below detection limit

4.2.5 Health risk assessment of Hg

The health risk assessment of Hg is evaluated based on the Hazard Index (HI), developed by United States Environmental Protection Agency (US EPA). Overall, hazard risks estimates based on the Canada population were the lowest for adults and the highest for infant below 1 year old (Table 4.14). The hazard index of *Leiocassis micropogon*, *Hampala bimaculata*, *Hemibagrus wyckii* and *Hemibagrus planiceps* are greater than 1 for all population groups considered whilst *Osteochilus vittatus* shows moderate hazard to children 1-4 years old. The suggested consumption limits of these species range from 35.37 g to 84.00 g/week for children 1-4 years; 64.84 g to 154.00 g/week for children 5-11 years; 159.65 g to 325.00 g/week for adult women; 179.30 g to 365.00 g/week for adult men. For species with hazard indices below one, the consumption rate was similarly calculated. The recommended consumption limits ranges from 1848.00 to 5110.00 g/week.

The result of the health risk of Hg based on the population of Malaysia was tabulated in Table 4.15. The hazard indices of *Luciosoma setigerum*, *Leiocassis micropogon*, *Hampala bimaculata*, *Hemibagrus wyckii*, *Hemibagrus planiceps*, *Macrogathus maculatus*, *Anematichthys armatus*, *Lobocheilos bo*, *Osteochilus vittatus*, *Luciosoma spilopleura* and *Channa lucius* are greater than 1 for all adult women and men. The suggested consumption limits ranging from 143.54 - 629.35 g/week for adult women and 163.48 - 716.80 g/week for adult men. For species with hazard indices lower than 1, the suggested consumption limits range from 818.16 - 4090.80 g/week for adult women and 931.84 - 4659.20 g/week for adult men.

Generally, the hazard indices calculated based on the recommended body weight and fish intake of Malaysia population was higher compared to that derived with the assumption of US EPA. This is due to the lower body weight and higher fish intake assumptions for Malaysian. In conclusion, fishes with hazard index greater than 1 would require continuous monitoring to ensure safe consumption.

Table 4.14: The health risk assessment of Hg based on the guideline of Canada population

Fish species	Hg Conc. (mg/kg)	Estimate dose (ED) (mg/kg/day)				Hazard index (HI)				Exposure ratio (ER)				Consumption limit (CR) (g), of fish/week			
		1-4 yr	5-11yrs	women	man	1-4yrs	5-11yrs	women	man	1-4yrs	5-11yrs	women	man	1-4yrs	5-11yrs	women	man
<i>O. anomalura</i>	0.10	0.0001	0.0001	0.0001	0.0001	0.46	0.25	0.41	0.37	0.69	0.38	0.26	0.23	201.60	369.60	910.00	1022.00
<i>L. setigerum</i>	0.13	0.0002	0.0001	0.0002	0.0001	0.60	0.33	0.53	0.47	0.90	0.49	0.34	0.30	155.08	284.31	700.00	786.15
<i>L. micropogon</i>	0.28	0.0004	0.0002	0.0003	0.0003	1.30	0.71	1.15	1.02	1.94	1.06	0.73	0.65	72.00	132.00	325.00	365.00
<i>B. collingwoodii</i>	0.04	0.0001	0.0000	0.0000	0.0000	0.19	0.10	0.16	0.15	0.28	0.15	0.10	0.09	504.00	924.00	2275.00	2555.00
<i>H. bimaculata</i>	0.57	0.0008	0.0004	0.0007	0.0006	2.64	1.44	2.34	2.08	3.96	2.16	1.49	1.33	35.37	64.84	159.65	179.30
<i>H. wyckii</i>	0.43	0.0006	0.0003	0.0005	0.0005	1.99	1.09	1.76	1.57	2.99	1.63	1.13	1.00	46.88	85.95	211.63	237.67
<i>H. planiceps</i>	0.43	0.0006	0.0003	0.0005	0.0005	1.99	1.09	1.76	1.57	2.99	1.63	1.13	1.00	46.88	85.95	211.63	237.67
<i>B. schwanefeldii</i>	0.03	0.0000	0.0000	0.0000	0.0000	0.14	0.08	0.12	0.11	0.21	0.11	0.08	0.07	672.00	1232.00	3033.33	3406.67
<i>T. tambroides</i>	0.03	0.0000	0.0000	0.0000	0.0000	0.14	0.08	0.12	0.11	0.21	0.11	0.08	0.07	672.00	1232.00	3033.33	3406.67
<i>L. bo</i>	0.13	0.0002	0.0001	0.0002	0.0001	0.60	0.33	0.53	0.47	0.90	0.49	0.34	0.30	155.08	284.31	700.00	786.15
<i>T. dourenensis</i>	0.08	0.0001	0.0001	0.0001	0.0001	0.37	0.20	0.33	0.29	0.56	0.30	0.21	0.19	252.00	462.00	1137.50	1277.50
<i>C. apogon</i>	<LOD	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>O. schlegelii</i>	0.06	0.0001	0.0000	0.0001	0.0001	0.28	0.15	0.25	0.22	0.42	0.23	0.16	0.14	336.00	616.00	1516.67	1703.33
<i>P. macronema</i>	0.02	0.0000	0.0000	0.0000	0.0000	0.09	0.05	0.08	0.07	0.14	0.08	0.05	0.05	1008.00	1848.00	4550.00	5110.00
<i>O. vittatus</i>	0.24	0.0003	0.0002	0.0003	0.0003	1.11	0.61	0.98	0.88	1.67	0.91	0.63	0.56	84.00	154.00	379.17	425.83
<i>L. spilopteura</i>	0.16	0.0002	0.0001	0.0002	0.0002	0.74	0.40	0.66	0.58	1.11	0.61	0.42	0.37	126.00	231.00	568.75	638.75
<i>L. fasciatus</i>	<LOD	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>P. micronemus</i>	0.02	0.0000	0.0000	0.0000	0.0000	0.09	0.05	0.08	0.07	0.14	0.08	0.05	0.05	1008.00	1848.00	4550.00	5110.00
<i>M. maculatus</i>	0.19	0.0003	0.0001	0.0002	0.0002	0.88	0.48	0.78	0.69	1.32	0.72	0.50	0.44	106.11	194.53	478.95	537.89
<i>R. borneensis</i>	0.06	0.0001	0.0000	0.0001	0.0001	0.28	0.15	0.25	0.22	0.42	0.23	0.16	0.14	336.00	616.00	1516.67	1703.33
<i>A. armatus</i>	0.14	0.0002	0.0001	0.0002	0.0002	0.65	0.35	0.57	0.51	0.97	0.53	0.37	0.33	144.00	264.00	650.00	730.00
<i>C. lucius</i>	0.20	0.0003	0.0002	0.0002	0.0002	0.93	0.51	0.82	0.73	1.39	0.76	0.52	0.47	100.80	184.80	455.00	511.00

<LOD: below detection limit

Table 4.15: The health risk assessment of Hg based on the population in Malaysia

Fish species	Hg conc. (mg/kg)	Estimate dose (mg/kg)		Hazard index (HI)		Exposure ratio (ER)		Consumption limit (CR) (g), of fish per week	
		women	men	women	men	women	men	women	men
<i>O. anomalura</i>	0.1	0.0003	0.0002	0.91	0.80	0.58	0.51	818.16	931.84
<i>L. setigerum</i>	0.13	0.0004	0.0003	1.19	1.04	0.76	0.66	629.35	716.80
<i>L. micropogon</i>	0.28	0.0008	0.0007	2.56	2.24	1.63	1.43	292.20	332.80
<i>B. collingwoodii</i>	0.04	0.0001	0.0001	0.37	0.32	0.23	0.20	2045.40	2329.60
<i>H. bimaculata</i>	0.57	0.0016	0.0014	5.20	4.57	3.32	2.92	143.54	163.48
<i>H. wyckii</i>	0.43	0.0012	0.0010	3.92	3.45	2.50	2.20	190.27	216.71
<i>H. planiceps</i>	0.43	0.0012	0.0010	3.92	3.45	2.50	2.20	190.27	216.71
<i>B. schwanenfeldii</i>	0.03	0.0001	0.0001	0.27	0.24	0.17	0.15	2727.20	3106.13
<i>T. tambroides</i>	0.03	0.0001	0.0001	0.27	0.24	0.17	0.15	2727.20	3106.13
<i>L. bo</i>	0.13	0.0004	0.0003	1.19	1.04	0.76	0.66	629.35	716.80
<i>T. dourenensis</i>	0.08	0.0002	0.0002	0.73	0.64	0.47	0.41	1022.70	1164.80
<i>C. apogon</i>	<LOD	-	-	-	-	-	-	-	-
<i>O. schlegelii</i>	0.06	0.0002	0.0001	0.55	0.48	0.35	0.31	1363.60	1553.07
<i>P. macronema</i>	0.02	0.0001	0.0000	0.18	0.16	0.12	0.10	4090.80	4659.20
<i>O. vittatus</i>	0.24	0.0007	0.0006	2.19	1.92	1.40	1.23	340.90	388.27
<i>L. spilopteura</i>	0.16	0.0004	0.0004	1.46	1.28	0.93	0.82	511.35	582.40
<i>L. fasciatus</i>	<LOD	-	-	-	-	-	-	-	-
<i>P. micronemus</i>	0.02	0.0001	0.0000	0.18	0.16	0.12	0.10	4090.80	4659.20
<i>M. maculatus</i>	0.19	0.0005	0.0005	1.73	1.52	1.11	0.97	430.61	490.44
<i>R. borneensis</i>	0.06	0.0002	0.0001	0.55	0.48	0.35	0.31	1363.60	1553.07
<i>A. armatus</i>	0.14	0.0004	0.0003	1.28	1.12	0.82	0.72	584.40	665.60
<i>C. lucius</i>	0.2	0.0005	0.0005	1.83	1.60	1.17	1.02	409.08	465.92

<LOD: below detection limit

CHAPTER 5

CONCLUSIONS AND RECOMMENDATIONS

The concentration of heavy metals from upstream and downstream of the Baleh River as well as Mengiong River was determined. The findings of the study indicated low risk of metals contamination in water of the Baleh River although some stations were characterized with higher Fe, Al, Mn and Pb, likely due to accelerated sediment runoff as a result of land clearing and boating activities. Significant positive correlations were deduced between Al-Fe-Mn in water suggesting that these elements may originate from the same source. For sediment, no risk of contaminations was identified at the moment. The baseline metals in sediment of the Baleh River were derived and these values may be used as the benchmark for future monitoring. For fishes, metals in gills were found higher than muscle and belly. The concentration of Al in *Oxygaster anomalura*, *Luciosoma setigerum*, *Leiocassis micropogon*, *Hampala bimaculata*, *Hemibagrus wyckii*, *Hemibagrus planiceps*, *Barbonymus schwanenfeldii*, *Tor tambroides*, *Lobocheilos bo*, *Tor dourenensis*, *Osteochilus schlegelii*, *Pengasius macronema*, *Osteochilus vittatus*, *Luciosoma spilopteura* and *Macrognathus maculatus* were higher than the guideline level set by FAO and MFA; whilst Hg in *Hampala bimaculata* was found exceeding the permissible guideline of 0.5 mg/kg ww. The hazard index of Hg in *Luciosoma setigerum*, *Leiocassis micropogon*, *Hampala bimaculata*, *Hemibagrus wyckii*, *Hemibagrus planiceps*, *Macrognathus maculatus*, *Anematichythys armatus*, *Lobocheilos bo*, *Osteochilus vittatus*, *Luciosoma spilopleura* and *Channa lucius* are greater than 1 for adult women and men (calculated based on the body weight and fish intake for population in Malaysia). Therefore, continuous monitoring is necessary to ensure the fish are safe for consumption.

REFERENCES

- Abdel-Moati, M. A., & El-Sammak, A. A. (1997). Man-made impact on the geochemistry of the Nile Delta Lakes: A study of metals concentrations in sediments. *Water, Air and Soil Pollution*, 97, 413-429.
- Abdullah, M. H., Sidi, J., & Aris, A. Z. (2007). Heavy Metals (Cd, Cu, Cr, Pb and Zn) in Meretrix *Meretrix röding*, water and sediments from estuaries in Sabah, north Borneo. *International Journal of Environmental and Science Education*, 2(3), 69 – 74.
- Adrian, W. J., & Stevens, M. L. (1977). Wet versus dry weights for heavy metal toxicity determinations in duck liver. *Journal of Wildlife Diseases*, 15(1), 125-126.
- Agency for Toxic Substances and Disease Registry (ATSDR). (2007). *Minimal risk levels for hazardous substances (MRLs)*. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.
- Agusa, T., Kunito, T., Yasunaga, G., Iwata, H., Subramanian, A., & Ismail, A. (2005). Concentrations of trace elements in marine fish and its risk assessment in Malaysia. *Marine Pollution Bulletin*, 51, 896–911.
- Ahmad, A. K., Shuhaimi-Othaman, M., & Mushrifah, I. (2008). The heavy metal variability in fish of Tasik Chini, Peninsular Malaysia. *The World Lake Conference*, 12, 2131-2135.
- Ahmad, A. K., Siti Munirah, A. H., & Shuhaimi-Othman, M. (2015). Preliminary test of fish respiratory and locomotive signal using multispecies freshwater bio indicator (MFB). *Jurnal Teknologi (Science and Engineering)*, 72(5), 155-158.
- Ahmad, N. I., Noh, M. F., Mahiyuddin, W. R., Jaafar, H., Ishak, I., & Azmi, W. N. (2015). Mercury levels of marine fish commonly consumed in Peninsular Malaysia. *Environmental Science and Pollution Research*, 22(5), 3672–3686.

- Ahmed, M. S., & Bibi, S. (2010). Uptake and bioaccumulation of water borne lead (Pb) in the fingerlings of a freshwater Cyprinid, *Catla catla*. *Journal of Animal and Plant Science*, 20(3), 201-207.
- Akoto, O., Bruce, T. N., & Darko, G. (2008). Heavy metals pollution profiles in streams serving the Owabi reservoir. *African Journal of Environmental Science and Technology*, 2(11), 354–359.
- Alam, L., Mokhtar, M., Alam, M., Bazri, A., Kathijotes, N., Goh, C. T., & Lee, K. E. (2015). Assessment of environmental and human health risk for contamination of heavy metal in tilapia fish collected from Langat Basin, Malaysia. *Asian Journal of Water, Environment and Pollution*, 12(2), 21–30.
- Alina, M., Azrina, A., Yunas, A. S., Zakiuddin, S., Izuan, E. H., & Rizal, R (2012). Heavy metals (mercury, arsenic, cadmium, plumbum) in selected marine fish and shellfish along the straits of Melacca. *International Food Research Journal*, 19(1), 135-140.
- Altamirano-Lozano, M., Beyersmann, D., Carter, D. E., Fowler, B. A., Fubini, B., & Williams, R. (2006). Cobalt in hard metals and cobalt sulfate, gallium arsenide, indium phosphide and vanadium pentoxide. *IARC Monographs on the Evaluation of Carcinogenic Risks to Humans*, 86.
- Al-Busaidi, M., Yesudhasan, P., Al-Mughairi, S., Al-Rahbi, W. K., Al-Harthy, K. S., & Al-Mazrooei, N. A. (2011). Toxic metals in commercial marine fish in Oman with reference to national and international standards. *Chemosphere*, 85(1), 67–73.
- Ambak, M. A., & Zaidi, Z. (2010). Freshwater fish diversity in Sungai Kelantan. *Journal of Sustainability Science and Management*, 5(1), 13–20.

- American Public Health Association (APHA). (2005). *Standard methods for the examination of water and wastewater*. Washington, D.C.: American Public Health Association-American Water Work Association-Water Environment Federation.
- Amiard, J. C., Amiard-Triquet, C., Barka, S., Pellerin, J., & Rainbow, P. S. (2006). Metallothioneins in aquatic invertebrates: Their role in metal detoxification and their use as biomarkers. *Aquatic Toxicology*, 76(2), 160–202.
- Amin, B., Ismail, A., Arshad, A., Yap, C. K., & Kamarudin, M. S. (2009). Anthropogenic impacts on heavy metal concentrations in the coastal sediments of Dumai, Indonesia. *Environmental Monitoring and Assessment*, 148(1), 291–305.
- Aprile, F. M., & Bouvy, M. (2010). Heavy metals in surface waters from a tropical river basin, Pernambuco State, northeastern Brazil. *Acta Scientiarum Biological Sciences*, 32(4), 357-364.
- Ashraf, M. A., Maah, M. J., & Yusoff, I. (2012). Speciation of heavy metals in the sediments of former tin mining catchment, Iranian. *Journal of Science and Technology*, A2, 163–180.
- Atkinson, C. A., Jolley, D. F., & Simpson, S. L. (2007). Effect of overlying water pH, dissolved oxygen, salinity and sediment disturbances on metal release and sequestration from metal contaminated marine sediments. *Chemosphere*, 69(9), 1428–1437.
- Avenant-Oldewage, A., & Marx, H. M. (2000). Bioaccumulation of chromium, copper and iron in the organs and tissues of *Clarias gariepinus* in the Olifants River, Kruger National Park. *Water Sanitation*, 26, 569-582.
- Avila-Pérez, P., Balcazar, M., Zarazua-Ortega, G., Barcelo-Quintal, I., & Díaz-Delgado, C. (1999). Heavy metal concentrations in water and bottom sediments of a Mexican reservoir. *Science of the Total Environment*, 234, 185–196.

- Azmi, M. Y., Junidah, R., Mariam, S. A., Safiah, M. Y., Fatimah, S., Norimah, A. K., Poh, B. K., Kandiah, M., & Tahir, A. (2009). *Malaysian Journal of Nutrition*, 15(2), 97-119.
- Badri, M. A., (1984). Identification of heavy metal toxicology levels in soil waste by chemical speciation. *Conservation and Recycling*, 7(2), 25-270.
- Baki, A., Dkhil, M. A., & Quraishy, A. (2011). Bioaccumulation of some heavy metals in tilapia fish relevant to their concentration in water and sediment of Wadi Hanifah, Saudi Arabia. *African Journal of Biotechnology*, 10(13), 2541–2547.
- Basaham, A. S., & Al-Lihaibi, S. S. (1993). Trace elements in sediments of the western Gulf. *Marine Pollution Bulletin*, 27, 103- 107.
- Bashir, F. H., Othman, M. S., Mazlan, A. G., Rahim, S. M., & Simon, K. D. (2013). Heavy metal concentration in fishes from the coastal waters of Kapar and Mersing, Malaysia. *Turkish Journal of Fisheries and Aquatic Sciences*, 13, 87–94.
- Başığit, B., & Tekin-özan, S. (2013). Concentrations of some heavy metals in water, sediment, and tissues of Pikeperch (*Sander lucioperca*) from Karataş Lake related to physico-chemical parameters, fish size, and seasons. *Journal of Environmental Studies and Sciences*, 22(3), 633–644.
- Bella, N. L., & Hilliker, A. (2003). Mercury in the Environment. *The Traprock*, 2(11), 33–36.
- Bertsch, R. M. (1990). Aluminum speciation: Methodology and applications, in S. A. Norton, S. E. Lindberg and A. L. Page (eds.). *Acid Precipitation, Advances in Environmental Sciences*, 4, 1-11.
- Bhuiyan, M. A. H., Parvez, L., Islam, M., Dampare, S. B., & Suzuki, S. (2010). Heavy metal pollution of coal mine-affected agricultural soils in the northern part of Bangladesh. *Journal of Hazardous Materials*, 173, 384–392.

- Bodek, I., Lyman, W. J., Reehl, W. F., & Rosenblatt, D. H. (1988). *Environmental inorganic chemistry*. New York: Pergamon Press.
- Bose-O'Reilly, S., McCarty, K., Steckling, N., & Lettmeier, B. (2010). Mercury exposure and children's health. *Current Problems in Pediatric and Adolescent Health Care*, 40(8), 186–215.
- Bradl, H. (2002). *Heavy metals in the environment: Origin, interaction and remediation volume 6*. London: Academic Press.
- Burger, J., Jeitner, C., Donio, M., Pittfield, T., Gochfeld, M., (2013). Mercury and selenium levels: Mercury molar ratios of brain, muscle, and other tissues in bluefish (*Pomatomus saltatrix*) from New Jersey, USA. *Science of the Total Environment*, 443, 278-286.
- Burton, A. G. (1992). *Assessing sediment quality: Sediment toxicity assessment*. Boca Raton: CRC Press.
- Campbell, P. G. C., & Tessier, A. (1989). *Geochemistry and bio availability of trace metals in sediments*, In: A. Boudou and F. Ribeyre, Eds., *aquatic ecotoxicology: Fundamental concepts and methodologies, Volume 1*. Boca Raton: CRC Press.
- Canadian Environmental Quality Guidelines (CEQG). (2001). *Canadian sediment quality guidelines for the protection of aquatic life*. Canadian: Canadian Council of Ministers of the Environment. Retrieved 24^h June, 2015 from <http://ceqg-rcqe.ccme.ca/en/index.html#void>
- Canavan, C. M., Caldwell, C. A., & Bloom, N. S. (2000). Discharge of methylmercury enriched hypolimnetic water from a stratified reservoir. *Science of the Total Environment*, 260, 159–170.
- Castro-González, M. I., & Méndez-Armenta, M. (2008). Heavy metals: Implications associated to fish consumption. *Environmental Toxicology and Pharmacology*, 26, 263–271.
- Cempel, M., & Nikel, G. (2006). Nickel: A review of its sources and environmental toxicology. *Polish Journal of Environmental Studies*, 15(3), 375–382.

- Chen, M., & Ma, L. Q. (2001). Comparison of three aqua regia digestion methods for twenty Florida Soils. *Soil Science Society of America Journal*, 65(2), 491-495.
- Chen, W. H., Tseng, S. I., Lee, T. L., Kan, C. C., & Wan, M. W. (2009). The optimization study of iron and manganese removal from groundwater by hollow-fiber. Retrieved 7th October, 2016 from http://ir.cnu.edu.tw/retrieve/43782/98_154_pro.pdf
- Chester, R. (2000). *Marine geochemistry*. London, UK: Academic Press.
- Colter, A., & Mahler, R. L. (2003). Iron in drinking water. *Environmental Protection*, 2, 3–6.
- Conti, G. O., Copat, C., Ledda, C., Fiore, M., Fallico, R., Sciacca, S., & Ferrante, M. (2012). Evaluation of heavy metals and polycyclic aromatic hydrocarbons (pahs) in *Mullus barbatus* from Sicily Channel and risk-based consumption limits. *Bulletin of Environmental Contamination and Toxicology*, 88, 946–950.
- Conti, M. E., & Cecchetti, G. (2001). Biological monitoring: Lichens as bio-indicators of air pollution assessment. *Environmental Pollution*, 114, 471–492.
- Cornellis, R., Caruso, J., Crews, H., & Heumann, K. (2005). *Handbook of elemental speciation II: Species in the environment, food, medicine and occupational health*. Chichester: John Wiley and Sons Limited Company.
- Cotton, F., A., & Wilkinson, G. (1998). *Advanced inorganic chemistry, 3rd edition*. New York: Wiley.
- Cretaz, A. L. D. L., & Barten, P. K. (2007). *Land use effects on streamflow and water quality in the Northeastern United States*. United States: CRC Press.
- Creti, P., Trinchella, F., & Scudiero, R. (2010). Heavy metal bioaccumulation and metallothionein content in tissues of the sea bream *Sparus aurata* from three different fish farming systems. *Environmental Monitoring and Assessment*, 165, 321–329.

- Cussen, D. I., & Hensman, C. E. (2011). *Determination of mercury and other trace metals in hydrocarbon using the Anton-Paar High Pressure Asher (HPA)*. Seattle: Frontier Geosciences Inc.
- Damiano, S., Papetti, P., & Menesatti, P. (2011). Accumulation of heavy metals to assess the health status of swordfish in a comparative analysis of Mediterranean and Atlantic areas. *Marine Pollution Bulletin*, 62, 1920–1925.
- David, R. (2005). *CRC handbook of chemistry and physics*. Boca Raton: CRC Press.
- Davies, O. A., & Allison, M. E. (2006). Bioaccumulation of heavy metals in water, sediment and periwinkle (*Tympanotonus fuscatus var radula*) from the Elechi Creek, Niger Delta. *African Journal of Biotechnology*, 5(10), 968-973.
- Davis, H. T., Marjorie, C., McDermott, S., & Lawson, A. B. (2009). Identifying natural and anthropogenic sources of metals in urban and rural soils using GIS-based data, PCA, and spatial interpolation. *Environmental Pollution*, 157(8–9), 2378–2385.
- Dean, J. R. (2002). *Methods for environmental trace analysis*. New York: Wiley.
- Department of Environment (DOE). (2012). *Malaysia: Environmental Quality Report 2012, Ministry of Science, Technology and the Environment, Putrajaya, Malaysia*. Retrieved 10th October 2015 from <https://enviro.doe.gov.my/view.php?id=15791>.
- Degroot, A. J., & Allersma, E. (1975). *Field observation on the transport of heavy metals in sediments, heavy metals in the aquatic environment*. Pennsylvania: Krenke.
- Dekov, V. M., Komy, Z., Araújo, F., Van Put, A., & Van Grieken, R. (1997). Chemical composition of sediments, suspended matter, river water and ground water of the Nile (Aswan–Sohag traverse). *Science of the Total Environment*, 20, 195–210.

- De Lima Rodrigues, A. S., Malafaia, G., Costa, A. T., & Junior, H. A. N. (2013). Background values for chemical elements in sediments of the Gualazo Do Norte river basin, MG, Brazil. *Revista de Ciencias Ambientais*, 7(2), 15-32.
- Dhaneesh, K. V., Gopi, M., Ganeshamurthy, R., Kumar, T. T. A., & Balasubramanian, T. (2012). Bio-accumulation of metals on reef associated organisms of Lakshadweep Archipelago. *Food Chemistry*, 131(3), 985–991.
- Djedjibegovic, J., Skrbo, T. L. A., Aleksandra, M., & Sober, M. (2012). Contents of cadmium, copper, mercury and lead in fish from the Neretva river (Bosnia and Herzegovina) determined by inductively coupled plasma mass spectrometry (ICP-MS). *Food Chemistry*, 131, 469–476. doi: 10.1016/j.foodchem.2011.09.009
- Driscoll, C. T., Blette, V., Yan, C., Schofield, C. L., Munson, R., & Holsapple, J. (1995). The roll of dissolved organic carbon in the chemistry and bioavailability of mercury in remote Adirondack lakes. *Water, Air Soil Pollution*, 80, 499-508.
- Du, L. G., Rinklebe, J., Vandecasteele, B., Meers, E., & Tack, F. M. (2009). Trace metal behaviour in estuarine and riverine floodplain soils and sediments: A review. *Science of the Total Environment*, 407, 3972–3985.
- Dvorak, B. I., & Skipton, S. O. (2014). *Drinking water: Iron and manganese*. Lincoln: NebGuide.
- Ebrahimpour, M., & Mushrifah, I. (2008). Heavy metal concentrations in water and sediments in Tasik Chini, a freshwater lake, Malaysia. *Environmental Monitoring and Assessment*, 141(1-3), 297–307.
- Eisler, R. (2010). *Compendium of trace metals and marine biota II*. Amsterdam: Vertebrates Elsevier.

- El Bouraie, M. M., El Barbary, A. A., Yehia, M. M., & Motawea, E. A. (2010). Heavy metal concentrations in surface river water and bed sediments at Nile Delta in Egypt. *Finnish Peatland Society*, 61(1), 1–12.
- El-Moselhy, K. M., Othman, A. I., Abd El-Azem, H., & El-Metwally, M. E. (2014). Bioaccumulation of heavy metals in some tissues of fish in the Red Sea, Egypt. *Egyptian Journal of Basic and Applied Sciences*, 1(2), 97–105.
- Elzahabi, M., & Yong, R. N. (2001). PH influence on sorption characteristics of heavy metal in the vadose zone. *Engineering Geology*, 60(1-4), 61-68.
- Emerson, R., & Lewis, C. M. (1939). Factors influencing the efficiency of photosynthesis, American. *Journal of Botany*, 26, 808–822.
- ENCO (n.d.). *Inorganic (metals) analyses*. Florida: ENCO Chemistry Book III. Retrieved 20 June, 2016 from http://www.encolabs.com/forms/ChemBook/ENCOChembook_III.pdf.
- Ersoz, M., & Barrott, L. (2012). *Best practice guide on metal removal from drinking water by treatment*. London: International Water Association.
- Evans, D. H. (1987). The fish gill: Site of action and model for toxic effects of environmental pollutants. *Environmental Health Perspectives*, 71, 47–58.
- Ezaimah, N. (2016). *Fish fauna in Batang Baleh, Kapit, Sarawak*. (Unpublished Master's thesis). Universiti Malaysia Sarawak, Sarawak, Malaysia.
- Ezeonyejiaku, C. D., & Obiakor, M. O. (2013). Physicochemical and heavy metal profile of surface water, anthropogenic activities, and community health implications. *Journal of Environmental Conservation Research*, 1(2), 40-48.

- Fabbri, P., Gabbianelli, G., Locatelli, C., Lubrano, P., Tormbini, C., & Vassura, I. (2001). Distribution of mercury and other heavy metals in core sediments of northern Adriatic Sea. *Water, Air and Soil Pollution*, 129, 143–153.
- Facetti, J., Dekov, V. M., & Van-Grieken, R. (1998). Heavy metals in sediments from the Paraguay River: A preliminary study. *Science of the Total Environment*, 209, 79-86.
- Fergusson, J. E. (1990). *The heavy elements: Chemistry, environmental impact and health effects*. Oxford: Pergamon Press.
- Fishbein, L. (1981). Sources, transport and alterations of metal compounds: An overview of arsenic, beryllium, cadmium, chromium, and nickel. *Environmental Health Perspectives*, 40-43.
- Fisheries Research Institute (2004). *Field guide to selected commercial marine fishes of Malaysian waters*. Malaysia: Fisheries Research Institute.
- Florence, T. M. (1982). Development of physico-chemical speciation procedures to investigate the toxicity of copper, lead, cadmium and zinc towards aquatic biota. *Analytica Chimica Acta*, 141, 73–94.
- Franc, S., Vinagre, C., Cacador, I., & Henrique, C. N. (2005). Heavy metal concentrations in sediment, benthic invertebrates and fish in three salt marsh areas subjected to different pollution loads in the Tagus Estuary (Portugal). *Marine Pollution Bulletin*, 50, 993–1018.
- Gaughhofer, J., & Bianchi, V. (1991). *Chromium in Merian, E. Metals and their compounds in the environment: occurrence, analysis and biological relevance*. Germany: Wiley-VCH.
- Gbaruko, B. C., & Friday, O. U. (2007). Bioaccumulation of heavy metals in some fauna and flora. *Journal of Environmental Science*, 4(2), 197 – 202.

- Georgopoulos, P. G., Roy, A., Yonone-Lioy, M. J., Opiekun, R. E., & Lioy, P. J. (2001). Copper: Environmental dynamics and human exposure issues. *Journal Toxicology Environmental Health*, 4(4), 341–394.
- Gorur, F. K., Keser, R., Akcay, N., & Dizman, S. (2012). Radioactivity and heavy metal concentrations of some commercial fish species consumed in the Black Sea Region of Turkey. *Chemosphere*, 87, 356-362.
- Gulmini, M., Ostacoli, G., & Zelano, V. (1994). Comparison between microwave and conventional heating procedures in tessier's extractions of calcium, copper, iron and manganese in a lagoon sediment. *Analyst*, 119, 2075–2080.
- Guimarães, J. R. D., Malm, O., & Pfeiffer, W. C. (1995). A simplified radiochemical technique for measurements of net mercury methylation rates in aquatic systems near gold mining areas, Amazon, Brazil. *Science of the Total Environment*, 175, 151-162.
- Guimarães, J. R. D., Meili, M., Malm, O., & Brito, E.M. (1998). Hg methylation in sediments and floating meadows of a tropical lake in the Pantanal floodplain, Brazil. *Science of the Total Environment*, 213, 165-175.
- Habibah, J., Khairiah, J., Ismail, B. S., & Kadderi, M. D. (2014). Manganese speciation in selected agricultural soils of Peninsular Malaysia. *American Journal of Environmental Science*, 10(2), 148-156.
- Hasrizal, S., Siti, N. H., Khawar, S., Joseph, B., & Yuzwan, M. (2015). Spatial distribution of selected heavy metals in surface sediments of the EEZ of the East Coast of Peninsular Malaysia. *International Journal of Oceanography*, 1, 1-10.
- Hasyimah, A. K. N., Noik, J., Teh, V., & Lee, C. (2011). Assessment of cadmium (Cd) and lead (Pb) levels in commercial marine fish organs between wet markets and supermarkets in Klang Valley, Malaysia. *International Food Research Journal*, 18, 795–802.

- Health Canada. (2007). *Human health risk assessment of mercury in fish and health benefits of fish consumption*. Ottawa, Canada: Health Canada. Retrieved 6th December 2016, from http://www.hc-sc.gc.ca/fn-an/pubs/mercur/merc_fish_poisson-eng.php
- He, Z., Song, J., Zhang, N., Zhang, P., & Xu, Y. (2009). Variation characteristics and ecological risk of heavy metals in the south Yellow Sea surface sediments. *Environmental Monitoring and Assessment*, 157, 515– 528.
- Heng, L. E. T. (2016). *Water quality and sediment characteristics of the Batang Baleh* (Unpublished Master's thesis). Universiti Malaysia Sarawak, Sarawak, Malaysia.
- Henry, F., Amara, R., Courcot, L., Lacouture, D., & Bertho, M. L. (2004). Heavy metals in four fish species from the French coast of the Eastern English Channel and Southern Bight of the North Sea. *Environmental International*, 30, 675–683.
- Hilal, A., & Ismail, N. S. (2008). Heavy metals in eleven common species of fish from the Gulf of Aqaba, Red Sea. *Jordan Journal of Biological Sciences*, 1(1), 13-18.
- Hooda, P. S. (2010). *Trace elements in soil*. Chichester, United Kingdom: John Wiley & Sons.
- Horowitz, A. J., & Elrick, K. A. (1997). The relation of stream sediment surface area, grain size and composition to trace element chemistry. *Applied Geochemistry*, 2(4), 437-451.
- Hosseini, M., Nabavi, S. M. B., & Parsa, Y. (2013). Bioaccumulation of mercury in trophic level of benthic, benthopelagic, pelagic fish species and sea bird from Arvand River, Iran. *Biological Trace Element Research*, 156(1-3), 175-180.
- Hossain, M. A., Ali, M. N., Islam, M. S., & Hossain, H. M. Z. (2015). Spatial distribution and source apportionment of heavy metals in soils of Gebeng industrial city, Malaysia. *Environmental Earth Sciences*, 73(1), 115-126.

- Hou, D., He, J., Lu, C., Ren, L., Fan, Q., & Wang, J. (2013). Distribution characteristics and potential ecological risk assessment of heavy metals (Cu, Pb, Zn, Cd) in water and sediments from Lake Dalinouer, China. *Ecotoxicology and Environmental Safety*, 93, 135–144.
- Hseu, Z. Y. (2004). Evaluating heavy metal contents in nine composts using four digestion methods. *Bioresource Technology*, 95(1), 53–59.
- Hungerford & Terry Inc. (2003). *Greensand process removes iron, manganese, arsenic from groundwater*. Retrieved 30th November 2016 from <http://www.wwdmag.com/arsenic/greensand-process-removes-iron-manganese-arsenic-groundwater>
- Idriss, A. A., & Ahmad, A. K. (2013). Heavy metals nickel and chromium in sediments in the Juru River, Penang, Malaysia. *Journal of Environmental Protection*, 11, 1245–1250.
- Igwegbe, A. O., Negbenebor, C. A., Chibuzo, E. C., Badau, M. H., & Agbara, G. I. (2015). Effect of season and fish smoking on heavy metal contents of selected fish species from three locations in Borno state of Nigeria. *Asian Journal of Science and Technology*, 6(2), 1010-1019.
- Irwandi, J. & Farida, O. (2009). Mineral and heavy metal contents of marine fin fish in Langkawi island, Malaysia. *International Food Research Journal*, 16(2), 105-112.
- Ikem, A., Egiebor, N. O., & Nyavor, K. (2003). Trace elements in water, fish and sediment from Tuskegee Lake, Southeastern USA. *Water, Air and Soil Pollution*, 149(1-4), 51–75.
- Ismail, I., & Saleh, I. M. (2012). Analysis of heavy metals in water and fish (*Tilapia* sp.) samples from Tasik Mutiara, Puchong. *The Malaysian Journal of Analytical Sciences*, 16(3), 346 – 352.
- Jaishankar, M., Tseten, T., Anbalagan, N., Mathew, B. B., & Beeregowda, K. N. (2014). Toxicity, mechanism and health effects of some heavy metals. *Interdisciplinary Toxicology*, 7(2), 60-72.
- Järup, L. (2003). Hazards of heavy metal contamination. *British Medical Bulletin*, 68, 167–182.

- Kabata-Pendias, A. (2011). *Trace elements in soils and plants*. Boca Raton: CRC Press.
- Kalay, M., & Canli, M. (2000). Elimination of essential (Cu, Zn) and non-essential (Cd, Pb) metals from tissues of a freshwater fish *Tilapia zilli*. *Turkish Journal of Zoology*, 24, 429- 436.
- Kargin, F. (1998). Metal concentrations in tissues of the freshwater fish *Capoeta barroisi* from the Seyhan River (Turkey). *Bulletin of Environmental Contamination and Toxicology*, 60, 822-828.
- Kasper, D., Forsberg, B. R., Amaral, J. H. F., Leito, R. P., Py-Daniel, S. S., Bastos, W. R., & Malm, O. (2014). Reservoir stratification affects methylmercury levels in river water, plankton, and fish downstream from Balbina hydroelectric dam, Amazonas, Brazil. *Environmental Science and Technology*, 48(2), 1032–1040.
- Kasper, D., Palermo, E. F. A., Branco, C. W. C., & Malm, O. (2012). Evidence of elevated mercury levels in carnivorous and omnivorous fishes downstream from an Amazon reservoir. *Hydrobiologia*, 694, 87–98.
- Kennish, M. J. (1992). *Practical handbook of estuarine and marine solution*. New York: CRC Press.
- Khairiah, J., Tharmendren, M. S. M., Habibah, H. Z., Wan Kamal, W. I., & Ismail, B. S. (2012). Heavy metal content in paddy soils of Ketara, Besut, Terengganu, Malaysia. *World Applied Sciences Journal*, 19 (2), 183-191.
- Khaled, A. (2004). Seasonal determination of some heavy metals in muscle tissues of *Siganus rivulatus* and *Sargus sargus* fish from El-Mex Bay and Eastern Harbor, Alexandria, Egypt. *Egyptian Journal of Aquatic Biology and Fisheries*, 8(1), 65-81.
- Kim, B., & Steele, K. F. (2004). Comparison of dissolved and acid-extractable metal concentrations in groundwater, Eastern Arkansas. *Journal of the Arkansas Academy of Science*, 58, 117-120.

- Kim, E. J., Herrera, J. E., Huggins, D., Braam, H., & Koshowski, S. (2011). Effect of pH on the concentrations of lead and trace contaminants in drinking water: A combined batch, pipe loop and sentinel home study. *Water Research*, 45, 2763-2774.
- Kottelat, M., & Tan, H. H. (2008). A synopsis of the genus *Lobocheilos* in Java, Sumatra and Borneo, with descriptions of six new species (Teleostei: Cyprinidae). *Ichthyological Exploration of Freshwaters*, 19(1), 27–58.
- Kotze, P. H., Preez, H. H., & Van-Vuren, J. H. J. (1999). Bioaccumulation of copper and zinc in *Oreochromis mossambicus* and *Clarias garicpinus*, from the Olifants River, Mpumalanga, South Africa. *Water South Africa*, 25, 99–110.
- Krushevskaya, A., Barnes, R. M., & Amarasinghe, C. (1993). Decomposition of biological samples for inductively coupled plasma atomic emission spectrometry using an open focused microwave digestion system. *Analyst*, 118, 1175–1181.
- Laws of Malaysia (LOM). (2006). *Food act 1983 (Act 281)*. Kuala Lumpur, Malaysia: The Commissioner of Law Revision.
- Ley, J. L., Ahmad, Z. A., Hazzeman, H., Fatimah, M.Y., & Zailina, H. (2016). The levels of mercury, methylmercury and selenium and the selenium health benefit value in grey-eel catfish (*Plotosus canius*) and giant mudskipper (*Periophthalmodon schlosseri*) from the Strait of Malacca. *Chemosphere*, 152, 265-273.
- Lim, D., Jung, H., Choi, J., Yang, S., & Ahn, K. (2006). Geochemical compositions of river and shelf sediments in the Yellow Sea: Grain-size normalization and sediment provenance. *Continental Shelf Research*, 26(1), 15–24.
- Ling, T. Y., Kho, C. P., & Nyanti, L. (2012). Spatial and temporal variations of heavy metals in a tropical river. *World Applied Sciences Journal*, 16(4), 550-559.

- Liu, G., Yu, Y., Hou, J., Xue, W., Liu, X. H., Ahmed, A., & Hayat, T. (2014). An ecological risk assessment of heavy metals pollution of the agricultural ecosystem near a lead-acid battery factory. *Ecological Indicators*, 47, 210-218.
- Lindqvist, O., Johansson, K., Aastrup, M., Bringmark, L., Hovsenius, G., Håkansson, L., & Timm, B. (1991). Mercury in the Swedish environment - recent research on causes, consequences and corrective methods. *Water, Air and Soil Pollution*, 55, 1-251.
- Lin, Y. C., Chang-Chien, G. P., Chiang, P. C., Chen, W. H., & Lin, Y. C. (2013). Multivariate analysis of heavy metal contaminations in seawater and sediments from a heavily industrialized harbor in Southern Taiwan. *Marine Pollution Bulletin*, 76, 266–275.
- Li, J., He, M., Sun, S., Han, W., Zhang, Y., & Gu, Y. (2009). Effect of the behavior and availability of heavy metals on the characteristics of the coastal soils developed from alluvial deposits. *Environmental Monitoring and Assessment*, 156(1), 91–98.
- Li, S., Zhou, L., Wang, H., Xiong, M., Yang, Z., & Chang, J. (2013). Short-term impact of reservoir impoundment on the patterns of mercury distribution in a subtropical aquatic ecosystem, Wujiang. *Environmental Science and Pollution Research*, 20(7), 4396-4404.
- Łuczyńska, J., Tońska, E., & Łuczyński, I. M. J. (2009). Essentials mineral components in the muscles of six freshwater fish from the Mazurian Great Lakes (northeastern Poland). *Archives of Polish Fisheries*, 17, 171-178.
- Luoma, S. N., & Rainbow, P. S. (2008). *Metal contamination in aquatic environment: Science and lateral management*. Cambridge: Cambridge University Press.
- Mandal, R., Hassan, N. M., Murimboh, J., Chakrabarti, C. L., & Back, M. H. (2002). Chemical speciation and toxicity of nickel species in natural waters from the Sudbury Area (Canada). *Environmental Science and Technology*, 36(7), 1477-1484.

- Marcotrigiano, G. O., & Storelli, M. M. (2003). Heavy metal, polychlorinated biphenyl and organochlorine pesticide residues in marine organisms: Risk evaluation for consumers. *Veterinary Research Communications*, 27, 183–195.
- Martin, J., & Meybeck, M. (1979). Elemental mass-balance of material carried by major world rivers. *Marine Chemistry*, 7, 178-206.
- Matilainen, T. (1995). Involvement of bacteria in methyl-Hg formation in anaerobic lake waters. *Water, Air and Soil Pollution*, 80, 757-764.
- Matschullat, J., Ottenstein, R., & Reimann, C. (2000). Geochemical background—can we calculate it? *Environmental Geology*, 39(9), 990–1000.
- Mcbride, M. B. (1994). *Environmental chemistry of soils 1st Edition*. Oxford: University Press.
- Merciai, R., Guasch, H., Kumar, A., Sabater, S., & García-Berthou, E. (2014). Trace metal concentration and fish size: Variation among fish species in a Mediterranean river. *Ecotoxicology and Environmental Safety*, 107, 154–161.
- Mehdi, Y., Hornick, J., Istasse, L., & Dufrasne, I. (2013). Selenium in the environment, metabolism and involvement in body functions. *Molecules*, 18(3), 3292–3311.
- Ministry of Health (MOH). (2000). *National drinking water quality standard (NDWQS)*. Putrajaya, Malaysia: Ministry of Health, Engineering Services Division.
- Mohamad, Z. A., Nur Rashidah, A. M., Ahmad, S., Zaini, H., & Mohamad, T. A. (2015). Evaluation of Heavy metal contamination levels of Balok River sediment in Pahang, Malaysia based on Geoaccumulation Index and supported with enrichment factor. *Malaysian Journal of Analytical Sciences*, 9(4), 707 – 714.
- Muller, G. (1969). Index of geoaccumulation in sediments of the Rhine River. *GeoJournal*, 2(3), 108–118.

- Mwashote, B. M. (2003). Levels of cadmium and lead in water, sediments and selected fish species in Mombasa, Kenya. *Western Indian Ocean Journal Marine Science*, 2(1), 25–34.
- Nadaska, G., Lesny, J., & Michalik, I. (2012). Environmental aspect of manganese chemistry. *Hungarian Electronic Journal of Science*, 10(2), 1–16.
- Nagajyoti, P. C., Lee, K. D., & Sreekanth, T. W. N. (2010). Heavy metals, occurrence and toxicity for plants: A review. *Environmental Chemistry Letters*, 8(3), 199–216.
- Naji, A., Ismail, A., & Ismail, A. R. (2010). Chemical speciation and contamination assessment of Zn and Cd by sequential extraction in surface sediment of Klang River, Malaysia. *Microchemical Journal*, 95, 285–292.
- Nduka, J. K., & Orisakw, O. E. (2011). Water-quality issues in the Niger Delta of Nigeria: A look at heavy metal levels and some physicochemical properties. *Environmental Science and Pollution Research*, 18(2), 237–246.
- NewsDesk. (2013, May 1). Batang Baleh bridge to benefit 160 long house. New Sarawak Tribune. Retrieved 12th April, 2014 from <http://www.newsarawaktribune.com/news/4238/Batang-Baleh-Bridge-to-benefit-160-longhouses/>
- Ng, H. H., & Kottelet, M. (2013). Revision of the Asian catfish genus *Hemibagrus bleeker*, 1862 (Teleostei: Siluriformes: Bagridae). *The Raffles Bulletin of Zoology*, 61(1), 205–291.
- Nobi, E. P., Dilipan, E., Thangaradjou, T., Sivakumar, K., & Kannan, L. (2010). Geochemical and geo-statistical assessment of heavy metal concentration in the sediments of different coastal ecosystems of Andaman Islands, India. *Estuarine Coastal and Shelf Science*, 87, 253–264.
- Nussey, G., Van Vuren, J. H. J., & Du Preez, H. H. (2000). Bioaccumulation of chromium, manganese, nickel and lead in the tissues of the moggel, *Labeo umbratus* (Cyprinidae), from Witbank Dam, Mpumalanga S. Africa. *Water and Sanitation Africa*, 26(2), 269–284.

- Obasohan, E. E., & Eguavoen, O. I. (2008). Seasonal variations of bioaccumulation of heavy metals in a freshwater fish (*Erpetoichthys calabaricus*) from Ogba River, Benim City, Nigeria. *African Journal of General Agriculture*, 4, 153–163.
- Odobasic, A. (2012). Determination and speciation of trace heavy metals in natural water by DPASV. *Environmental Sciences*, 5, 429–456.
- Ogoyi, D. O., Mwita, C. J., Nguu, E. K., & Shiundu, P. M. (2011). Determination of heavy metal content in water, sediment and microalgae from Lake Victoria, East Africa. *The Open Environmental Engineering Journal*, 4, 156–161.
- Ogunfowokan, A. O., Oyekunle, J. A. O., Atoyebi, A. O., & Lawal, A. (2013). Speciation study of heavy metals in water and sediments from Asunle River of the Obafemi Awolowo University, Ile-Ife, Nigeria. *International Journal of Environmental Protection*, 3(6), 6–16.
- Olowoyo, D. N., Ajayi, O. O., Amoo, I. A., & Ayeisanmi, A. F. (2010). Seasonal variation of metal concentrations in catfish, blue crab and crayfish from Warri Coastal Water of Delta State, Nigeria. Pakistan. *Journal of Nutrition*, 9(11), 1118-1121.
- Oughton, D. H, Salbu, B., Bjornstad, H. E., & Day, J. P. (1992). Use of an aluminium-26 tracer to study the deposition of aluminium species on fish gills following mixing of limed and acidic waters. *Analyst*, 117(3), 619-621.
- Perez, C. B., Boia, C., Pombo, L., & Rebelo, L. (2001). Determination of trace metals in fish species the Ria de Aveiro (Portugal) by electrothermal atomic adsorption spectrometry. *Food Chemistry*, 75, 93-100.
- Rajkowska, M., & Protasowicki, M. (2011). Distribution of selected metals in bottom sediments of lakes Insko and Wiola (Poland). *Ecological Chemistry and Engineering*, 18(5–6), 805–812.

- Regine, M. B., Gilles, D., Yannick, D., Alain, B., (2006). Mercury distribution in fish organs and food regimes: significant relationships from twelve species collected in French Guiana (Amazonian basin). *Science of the Total Environment*, 368, 262-270.
- Reimann, C., & Garrett, R. G. (2005) Geochemical background: Concept and reality. *Science of the Total Environment*, 350, 12-27.
- Renieri, E., Alegakis, A., Kiriakakis, M., Vinceti, M., Ozcagli, E., & Tsatsakis, A. (2014). Cd, Pb and Hg biomonitoring in fish of the Mediterranean Region and risk estimations on fish consumption. *Toxics*, 2(3), 417–442.
- Renoldi, M., Camusso, M., & Tartari, G. (1997). The highly polluted Lambro River Italy: Dissolved and solid transport of Cu, Cr and Fe. *Water, Air and Soil Pollution*, 95, 99-118.
- Rezayi, M., Ahmadzadeh, S., Kassim, A., & Heng, L. Y. (2011). Thermodynamic studies of complex formation between Co (SALEN) ionophore with chromate (II) Ions in AN-H₂O binary solutions by the conductometric method. *International Journal of Electrochemical Science*, 6, 6350–6359.
- Ritchie, G. P. S., & Posner, A. M. (1982). The effect of pH and metals binding on the transport properties. *Journal of Soil Science*, 33, 233-247.
- Rohasliney, H., Tan, H. S., Noor Zuhartini, M. M., & Tan, P. Y. (2014). Determination of heavy metal levels in fishes from the lower reach of the Kelantan River, Kelantan, Malaysia. *Tropical Life Sciences Research*, 25(2), 21–39.
- Sackett, D. K., Cope, W. G., Rice, J. A., & Aday, D. D. (2013). The influence of fish length on tissue mercury dynamics: Implications for natural resource management and human health risk. *International Journal of Environmental Research and Public Health*, 10(2), 638-659.

- Saeed, S. M., & Shaker, I. M. (2008). *Assessment of heavy metals pollution in water and sediments and their effect on Oreochromis Niloticus in the Northern Delta Lakes, Egypt*. Egypt: 8th International Symposium on Tilapia in Aquaculture 2008, World Fish Center.
- Sahuquillo, A., Rigol, A., & Rauret, G. (2003). Overview of the use of leaching/extraction tests for risk assessment of trace metals in contaminated soils and sediments. *Trends in Analytical Chemistry*, 22, 152–159.
- Samecka-Cymerman, A., & Kempers, A. J. (2001). Concentrations of heavy metals and plant nutrients in water, sediments and aquatic macrophytes of anthropogenic lakes former open cut brown coal/mines differing in stage of acidification. *Science of the Total Environment*, 281, 87-98.
- Sany, S. B. T., Salleh, A., Sulaiman, A. H., Sasekumar, A., Rezayi, M., & Tehrani, G. M. (2013). Heavy metal contamination in water and sediment of the Port Klang coastal area, Selangor, Malaysia. *Environmental Earth Sciences*, 69(6), 2013–2025.
- Schetagne, R., Doyon, J. F., & Fournier, J. J. (2000). Export of mercury downstream from reservoirs. *Science of the Total Environment*, 260, 135-145.
- Schetagne, R., & Verdon, R. (1999). *Post-impoundment evolution of fish mercury levels at the La Grande Complex, Quebec, Canada (from 1978 to 1996)*. Berlin: Springer.
- Seyedeh, B., Tavakoly, S., Aishah, S., Majid, R., Naghmeh, S., & Ghazaleh, M. T. (2013). Distribution and contamination of heavy metal in the coastal sediment of Pork Klang, Selangor, Malaysia. *Water, Air and Soil Pollution*, 1(224), 1476-1481.
- Sharpe, S., & Mackay, D. (2000). A framework for evaluating bioaccumulation in food webs. *Environmental Science and Technology*, 34(12), 2373-2379.

- Sibon, P. (2010, October 14). Log-jam a man-made disaster. *Borneo Post Online*. Retrieved 12th February, 2015 from <http://www.theborneopost.com/2010/10/14/%E2%80%98logjam-a-man-made-disaster%E2%80%99/>
- Sim, S. F., Ling, T. Y., Nyanti, L., Lee, T. Z. E., Mohd Irwan Lu, N. A. L., & Bakeh, T. (2014). Distribution of major and trace elements in a tropical hydroelectric reservoir in Sarawak, Malaysia. *International Scholarly Research Notices*, 1, 1-12.
- Sim, S. F., Ling, T. Y., Nyanti, L., Gerunsin, N., Wong, Y. E., & Kho, L. P. (2016). Assessment of heavy metals in water, sediment, and fishes of a large tropical hydroelectric dam in Sarawak, Malaysia. *Journal of Chemistry*, 1, 1-10.
- Singh, A. K., Srivastava, S. C., Verma, P., Ansari, A., & Verma, A. (2014). Hazard assessment of metals in invasive fish species of the Yamuna River, India in relation to bioaccumulation factor and exposure concentration for human health implications. *Environmental Monitoring and Assessment*, 186(6), 3823-3836.
- Singh, S. P., Tack, F. M. G., Gabriels, D., & Verloo, M. G. (2000). Heavy metal transport from dredged sediment derived surface soils in a laboratory rainfall simulation experiment. *Water, Air and Soil Pollution*, 118(1), 73-86.
- Somer, E. (1974). Toxic potential of trace metals in foods. *Journal of Food Science*, 39, 215–217.
- Soo, C. L., Ling, T. Y., Nyanti, L., & Apun, K. (2016). Assessment of the characteristic of nutrients, total metals, and fecal coliform in Sibu Laut River, Sarawak, Malaysia. *Applied Water Science*, 6(1), 77-96.
- Sposito, G., Lund, L. J., & Chang, A. C. (1982). Selective and partial extraction analyses of transported overburden for gold exploration in the Yilgarn Craton, Western Australia. *Soil Science Society of America*, 46, 260-264.

- Stihi, C., Bancuta, A., Popescu, I. V., Virgolici, M., Cimpoca, V., Gugiu, M., & Vlaicu, G. (2006). Air pollution studies using PIXE and ICP methods. *Journal of Physics: Conference Series*, 41, 565–568.
- Storelli, M. M., & Marcotrigiano, G. O. (2005). Bioindicator organisms: Heavy metal pollution evaluation in the Ionian sea (Mediterranean sea–Italy). *Environmental Monitoring and Assessment*, 1(102), 159–166.
- Taweel, A., Shuhaimi-Othman, M., & Ahmad, A. K. (2011). Heavy metals concentration in different organs of tilapia fish (*Oreochromis niloticus*) from selected areas of Bangi, Selangor, Malaysia. *African Journal of Biotechnology*, 10(55), 11562–11566.
- Teng, Y., Ni, S., Wang, J., & Niu, L. (2007). Geochemical baseline of trace elements in the sediment in Dexing area, South China. *Environmental Geology*, 57(7), 1649–1660.
- Tokalioglu, Ş., Karta, Ş., & Elçi, L. (2000). Speciation and determination of the heavy metals in lake waters by atomic absorption spectrometry after sorption on Amberlite XAD-16 Resin. *Analytical Sciences*, 16, 1169–1174.
- Topping, G. (1973). Heavy metals in fish from Scottish waters. *Aquaculture*, 11, 373–377.
- Tuomola, L. (2005). Mercury levels in fish five years after construction of Lago Manso Reservoir, Brazil. *Tropical Ecology*, 11, 1–28.
- United States Department of Agriculture (USDA) (2014). *Kellogg Soil Survey Laboratory Methods Manual*. Retrieved 13th October, 2016 from http://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/stelprdb1253872.pdf
- USEPA (United States Environmental Protection Agency) (1997). *Exposure Factors Handbook (1997 Final Report)*. Washington, DC: U.S. Environmental Protection Agency.

- USEPA (United States Environmental Protection Agency) (2000). *Guidance for assessing chemical contaminant data for use in fish advisories*. Washington, DC: Office of Science and Technology and Office of Water. Retrieved 6th December, 2016 from <https://www.epa.gov/sites/production/files/2015-06/documents/volume2.pdf>
- Tuomola, L., Niklasson, T., & Hylander, L. D. (2008). Fish mercury development in relation to abiotic characteristics and carbon sources in a six-year-old, Brazilian reservoir. *Science of the Total Environment*, 390(1), 177–187.
- Walna, B., Spychalski, W., & Ibragimow, A. (2010). Fractionation of iron and manganese in the horizons of a nutrient-poor forest soil profile using the sequential extraction method. *Journal of Environment Studies and Science*, 19(5), 1029–1037.
- Wang, S. X. (2001). Molecular mechanisms of metal toxicity and carcinogenesis. *Molecular Cell Biochemistry*, 222, 3–9.
- Warren, L. A., & Haack, E. A. (2001). Biogeochemical controls on metal behaviour in freshwater environments. *Earth-Science Reviews*, 54, 261–320.
- Wei, C., & Wen, H. (2012). Geochemical baselines of heavy metals in the sediments of two large freshwater lakes in China: Implications for contamination character and history. *Environment Geochemistry and Health*, 34, 737-748.
- Wintz, H., Fox, T., & Vulpe, C. (2002). Responses of plants to iron, zinc and copper deficiencies. *Biochemical Society Transactions*, 30, 766–768.
- Wong, J. (2014). *SEB to invest RM10bil to build two more hydroelectric dams*. Retrieved 14th February, 2014 from <http://www.thestar.com.my/Business/Business-News/2014/11/14/SEB-to-invest-RM10b-more-Its-planning-to-build-another-two-hydroelectric-dams-over-the-next-5-years/?style=biz>

- World Health Organization (WHO). (1996). *Trace elements in human Nutrition and health*. Geneva: WHO Press, World Health Organization.
- World Health Organization (WHO). (2001). *Arsenic and arsenic compounds (Environmental health criteria 224), 2nd edition*. Geneva: WHO Press, International Programme on Chemical Safety.
- World Health Organization (WHO). (2010). *Exposure to lead: A major public health concern*. Geneva: WHO Press, World Health Organization 6.
- Xian, X. (1987). Chemical partitioning of cadmium, zinc, lead and copper in soils near smelters. *Journal Environmental Science and Health*, 6, 527 – 541.
- Xue, N., Seip, H. M., Guo, J., Liao, B., & Zeng, Q. (2006). Distribution of Al-, Fe-, and Mn-pools and their correlation in soils from two acid deposition small catchments in Hunan, China. *Chemosphere*, 65(11), 2468-2476.
- Yang, L., Li, Y., Ma, X., & Yan, Q. (2013). Comparison on dry-ashing, wet-ashing and microwave digestion for determination of trace elements in *Periostracum serpentis* and *Periostracum cicadae* by ICP-AES. *Journal of the Chilean Chemical Society*, 58(3), 1876-1879.
- Yi, Y., Yang, Z., & Zhang, S. (2011). Ecological risk assessment of heavy metals in sediment and human health risk assessment of heavy metals in fishes in the middle and lower reaches of the Yangtze River basin. *Environmental Pollution*, 159(10), 2575–2585.
- Yilmaz, F., Zdemir, N., Demirak, A., & Tuna, A. L. (2007). Heavy metal levels in two fish species *Leuciscus cephalus* and *Lepomis gibbosus*. *Food Chemistry*, 100(2), 830–835.
- Zahra, A., Hashmi, M. Z., Malik, R. N., & Ahmed, Z. (2013). Enrichment and geo-accumulation of heavy metals and risk assessment of sediments of the Kurang Nallah—feeding tributary of the Rawal Lake Reservoir, Pakistan. *Science of the Total Environment*, 1, 925-933.

- Zarazua, G., Avila-Perez, P., Tejeda, S., Barcelo-Quintal, I., & Martinez, T. (2006). Analysis of total and dissolved heavy metals in surface water of a Mexican polluted river by total reflection X-ray fluorescence spectrometry. *Spectrochimica Acta - Part B Atomic Spectroscopy*, 61(10-11), 1180–1184.
- Zhao, S., Feng, C., Quan, W., Chen, X., Niu, J., & Shen, Z. (2012). Role of living environments in the accumulation characteristics of heavy metals in fishes and crabs in the Yangtze River Estuary, China. *Marine Pollution Bulletin*, 64(6), 1163–1171.
- Zheng, N., Wang, Q., Liang, Z., & Zheng, D. (2008). Characterization of heavy metal concentrations in the sediments of three freshwater rivers in Huludao City, northeast China. *Environmental Pollution*, 154, 135-142.

APPENDICES

Appendix A

The mean concentrations of metals in water across 50 stations (<LOD indicates below detection limit)

Stations	Elements (mg/L)										
	Al	Cr	Cu	Mn	Ni	Pb	Zn	Fe	As	Se	Hg
1	0.33 ^{e,f,g,h,i}	<LOD	<LOD ^a	0.13 ^{g,h,i}	<LOD ^a	0.01 ^a	<LOD ^a	0.02 ^a	<LOD	<LOD	<LOD
2	0.07 ^{a,b,c,d}	<LOD	0.01 ^{f,g}	0.03 ^{a,b,c}	<LOD ^a	0.03 ^d	0.00 ^{d,e,f,g}	0.27 ^{a,b,c}	<LOD	<LOD	<LOD
3	0.39 ^{h,i}	<LOD	<LOD ^a	0.12 ^{f,g,h,i}	<LOD ^a	0.01 ^a	<LOD ^a	0.38 ^{a,b,c}	<LOD	<LOD	<LOD
4	0.48 ^{h,i,j}	<LOD	<LOD ^a	0.15 ^{i,j,k}	<LOD ^a	0.01 ^{a,b}	<LOD ^a	0.24 ^{a,b,c}	<LOD	<LOD	<LOD
5	0.00 ^{a,b}	<LOD	0.01 ^{f,g}	<LOD ^a	<LOD ^{a,b}	0.03 ^d	0.02 ^{b,c,d,e,f}	0.06 ^a	<LOD	<LOD	<LOD
6	0.16 ^{a,b,c,d,e}	<LOD	<LOD ^a	0.06 ^{b,c,d,e,f}	<LOD ^a	<LOD ^a	<LOD ^a	0.05 ^a	<LOD	<LOD	<LOD
7	0.10 ^{a,b,c,d}	<LOD	<LOD ^a	0.05 ^{a,b,c,d,e}	<LOD ^a	0.01 ^a	<LOD ^a	<LOD ^a	<LOD	<LOD	<LOD
8	0.12 ^{a,b,c,d,e}	<LOD	<LOD ^a	0.06 ^{b,c,d,e,f}	<LOD ^a	<LOD ^a	<LOD ^a	0.03 ^a	<LOD	<LOD	<LOD
9	0.10 ^{a,b,c,d}	<LOD	<LOD ^a	0.05 ^{a,b,c,d,e}	<LOD ^a	<LOD ^a	<LOD ^a	0.03 ^a	<LOD	<LOD	<LOD
10	0.12 ^{a,b,c,d,e}	<LOD	<LOD ^a	0.05 ^{a,b,c,d,e}	<LOD ^a	<LOD ^a	<LOD ^a	0.08 ^a	<LOD	<LOD	<LOD
11	0.08 ^{a,b,c,d}	<LOD	<LOD ^a	0.05 ^{a,b,c,d,e}	<LOD ^a	<LOD ^a	<LOD ^a	0.15 ^{a,b}	<LOD	<LOD	<LOD
12	0.11 ^{a,b,c,d,e}	<LOD	<LOD ^a	0.06 ^{a,b,c,d,e,f}	<LOD ^a	<LOD ^a	<LOD ^a	0.13 ^a	<LOD	<LOD	<LOD
13	0.03 ^{a,b,c}	<LOD	0.01 ^{f,g}	0.03 ^{a,b,c}	<LOD ^a	0.02 ^{c,d}	<LOD ^{a,b,c,d,e}	0.19 ^{a,b,c}	<LOD	<LOD	<LOD
14	0.26 ^{d,e,f,g,h}	<LOD	0.01 ^{f,g}	<LOD ^a	<LOD ^a	0.03 ^d	<LOD ^{a,b,c,d}	0.06 ^a	<LOD	<LOD	<LOD
15	0.10 ^{a,b,c,d}	<LOD	0.01 ^{f,g}	0.04 ^{a,b,c,d}	<LOD ^a	0.02 ^{c,d}	<LOD ^{a,b,c,d}	0.25 ^{a,b,c}	<LOD	<LOD	<LOD
16	0.22 ^{a,b,c,d,e,f,g}	<LOD	0.01 ^{f,g}	0.14 ^{h,i}	<LOD ^{a,b}	0.02 ^{b,c,d}	<LOD ^{a,b}	2.00 ^e	<LOD	<LOD	<LOD
17	0.39 ^{f,g,h,i}	<LOD	<LOD ^{a,b,c}	0.10 ^{d,e,f,g,h,i}	0.01 ^{a,b}	0.03 ^d	<LOD ^{d,e,f,g}	0.53 ^{a,b,c}	<LOD	<LOD	<LOD
18	0.10 ^{a,b,c,d,e}	<LOD	<LOD ^a	0.02 ^{a,b}	0.01 ^b	0.03 ^d	0.04 ^{e,f,g}	0.17 ^{a,b,c}	<LOD	<LOD	<LOD
19	0.00 ^{a,b}	<LOD	<LOD ^{a,b}	0.08 ^{b,c,d,e,f,g,h}	0.01 ^b	0.03 ^d	0.04 ^{e,f,g}	0.45 ^{a,b,c}	<LOD	<LOD	<LOD
20	0.11 ^{a,b,c,d,e}	<LOD	<LOD ^{a,b}	0.02 ^{a,b}	<LOD ^a	0.03 ^d	0.02 ^{a,b,c,d,e}	0.13 ^{a,b}	<LOD	<LOD	<LOD
21	0.04 ^{a,b,c,d}	<LOD	<LOD ^{a,b}	0.07 ^{b,c,d,e,f,g}	<LOD ^a	0.03 ^d	<LOD ^{c,d,e,f,g}	0.35 ^{a,b,c}	<LOD	<LOD	<LOD
22	0.06 ^{a,b,c,d}	<LOD	0.01 ^{f,g}	0.02 ^{a,b,c}	0.01 ^{a,b}	0.03 ^d	0.05 ^g	0.19 ^{a,b,c}	<LOD	<LOD	<LOD
23	2.02 ⁿ	<LOD	0.01 ^g	0.57 ^o	0.01 ^b	0.03	0.04 ^{f,g}	1.94 ^{d,e}	<LOD	<LOD	<LOD

Appendix A: Continued

24	1.85 ⁿ	<LOD	0.01 ^{f,g}	0.48 ⁿ	0.01 ^b	0.02 ^{b,c,d}	<LOD ^{b,c,d,e,f,g}	1.51 ^{c,d,e}	<LOD	<LOD	<LOD
25	1.06 ^m	<LOD	0.01 ^{f,g}	0.45 ⁿ	<LOD ^a	0.01 ^{a,b,c}	<LOD ^{a,b,c,d,e}	1.48 ^{b,c,d,e}	<LOD	<LOD	<LOD
26	0.07 ^{a,b,c,d}	<LOD	<LOD ^a	0.03 ^{a,b,c}	<LOD ^a	<LOD ^a	<LOD ^{a,b,c,d,e}	0.19 ^{a,b,c}	<LOD	<LOD	<LOD
27	0.76 ^l	<LOD	0.01 ^{c,d,e,f}	0.21 ^{k,l}	0.02 ^c	0.01 ^{a,b,c}	<LOD ^{a,b,c,d,e}	1.02 ^{a,b,c,d,e}	<LOD	<LOD	<LOD
28	0.18 ^{a,b,c,d,e,f,g}	<LOD	<LOD ^{a,b,c}	0.05 ^{a,b,c,d,e}	0.01 ^b	0.00 ^a	<LOD ^{a,b,c}	0.34 ^{a,b,c}	<LOD	<LOD	<LOD
29	0.11 ^{a,b,c,d,e}	<LOD	<LOD ^{b,c,d,e}	0.03 ^{a,b,c}	<LOD ^a	0.01 ^a	<LOD ^{a,b,c}	0.24 ^{a,b,c}	<LOD	<LOD	<LOD
30	0.26 ^{c,d,e,f,g,h}	<LOD	<LOD ^a	0.15 ^{i,j}	<LOD ^a	<LOD ^a	<LOD ^a	0.60 ^{a,b,c,d}	<LOD	<LOD	<LOD
31	6.11 ^o	<LOD	<LOD ^a	1.74 ^p	<LOD ^a	<LOD ^a	<LOD ^a	6.00 ^f	<LOD	<LOD	<LOD
32	0.00 ^{a,b}	<LOD	<LOD ^a	0.02 ^{a,b}	<LOD ^a	<LOD ^a	<LOD ^a	0.08 ^a	<LOD	<LOD	<LOD
33	0.03 ^{a,b,c}	<LOD	<LOD ^a	0.02 ^{a,b}	<LOD ^a	<LOD ^a	<LOD ^a	0.23 ^{a,b,c}	<LOD	<LOD	<LOD
34	0.03 ^{a,b,c}	<LOD	<LOD ^a	0.02 ^{a,b,c}	<LOD ^a	<LOD ^a	<LOD ^a	0.12 ^a	<LOD	<LOD	<LOD
35	0.06 ^{a,b,c,d}	<LOD	<LOD ^a	0.02 ^{a,b}	<LOD ^a	<LOD ^a	<LOD ^a	0.13 ^a	<LOD	<LOD	<LOD
36	0.06 ^{a,b,c,d}	<LOD	<LOD ^a	0.03 ^{a,b,c}	<LOD ^a	<LOD ^a	<LOD ^a	0.24 ^{a,b,c}	<LOD	<LOD	<LOD
37	0.15 ^{a,b,c,d,e}	<LOD	<LOD ^{a,b,c,d}	0.05 ^{a,b,c,d,e}	<LOD ^a	0.01 ^a	<LOD ^a	0.28 ^{a,b,c}	<LOD	<LOD	<LOD
38	0.11 ^{a,b,c,d,e}	<LOD	<LOD ^{b,c,d}	0.03 ^{a,b,c}	<LOD ^a	0.01 ^a	<LOD ^a	0.18 ^{a,b,c}	<LOD	<LOD	<LOD
39	0.00 ^a	<LOD	0.01 ^{d,e,f,g}	0.02 ^{a,b}	<LOD ^a	<LOD ^a	<LOD ^a	0.15 ^{a,b}	<LOD	<LOD	<LOD
40	0.18 ^{a,b,c,d,e,f,g}	<LOD	0.01 ^{e,f,g}	0.10 ^{e,f,g,h,i}	<LOD ^a	<LOD ^a	<LOD ^a	0.44 ^{a,b,c}	<LOD	<LOD	<LOD
41	0.23 ^{b,c,d,e,f,g}	<LOD	0.01 ^{f,g}	0.06 ^{a,b,c,d,e,f}	<LOD ^a	0.01 ^a	<LOD ^a	0.44 ^{a,b,c}	<LOD	<LOD	<LOD
42	0.09 ^{a,b,c,d}	<LOD	0.01 ^{f,g}	0.05 ^{a,b,c,d,e}	<LOD ^a	<LOD ^a	<LOD ^a	0.31 ^{a,b,c}	<LOD	<LOD	<LOD
43	0.77 ^l	<LOD	0.01 ^{f,g}	0.23 ^{l,m}	<LOD ^a	0.01 ^{a,b}	<LOD ^a	1.18 ^{a,b,c,d,e}	<LOD	<LOD	<LOD
44	0.71 ^{k,l}	<LOD	0.01 ^g	0.23 ^{l,m}	<LOD ^a	0.01 ^{a,b}	<LOD ^a	1.02 ^{a,b,c,d,e}	<LOD	<LOD	<LOD
45	0.53 ^{i,j,k}	<LOD	0.01 ^g	0.20 ^{j,k,l}	<LOD ^a	0.01 ^{a,b}	<LOD ^a	0.66 ^{a,b,c,d,e}	<LOD	<LOD	<LOD
46	0.16 ^{a,b,c,d,e,f}	<LOD	0.01 ^{f,g}	0.08 ^{c,d,e,f,g,h}	<LOD ^a	<LOD ^a	<LOD ^a	0.36 ^{a,b,c}	<LOD	<LOD	<LOD
47	0.68 ^{j,k,l}	<LOD	0.01 ^g	0.24 ^{l,m}	<LOD ^a	0.01 ^a	<LOD ^a	0.95 ^{a,b,c,d,e}	<LOD	<LOD	<LOD
48	0.84 ^{l,m}	<LOD	0.01 ^g	0.22 ^l	<LOD ^a	0.01 ^a	<LOD ^a	0.91 ^{a,b,c,d,e}	<LOD	<LOD	<LOD
49	0.72 ^{k,l}	<LOD	0.01 ^g	0.29 ^m	0.01 ^{a,b}	0.01 ^a	<LOD ^a	1.16 ^{a,b,c,d,e}	<LOD	<LOD	<LOD
50	0.72 ^k	<LOD	0.01 ^{f,g}	0.25 ^{l,m}	0.01 ^{a,b}	<LOD ^a	<LOD ^a	1.20 ^{a,b,c,d,e}	<LOD	<LOD	<LOD

Appendix B

The mean of concentration of metals in sediment across 50 stations (<LOD indicates below detection limit)

Stations	Elements (mg/kg)								
	Al	Cr	Cu	Mn	Ni	Pb	Zn	Fe	Hg
1	72670.20 ^{n,o,p,q,r}	0.21 ^a	0.12 ^a	694.12 ^{r,s}	<LOD ^a	0.08 ^a	0.26 ^a	43324.17 ^{s,t}	0.11 ^{a,b,c,d,e}
2	38854.88 ^{a,b,c,d,e,f}	25.32 ^{b,c}	15.01 ^{b,c,d,e,f,g}	354.33 ^{b,c,d,e,f,g,h,i,j}	<LOD ^a	15.84 ^{g,h,i,j,k,l,m,n}	43.05 ^{b,c,d,e,f}	21983.52 ^{a,b,c}	0.07 ^{a,b,c}
3	91135.25 ^r	0.28 ^a	0.14 ^a	99.00 ^a	<LOD ^a	0.15 ^a	0.21 ^a	50207.60 ^u	0.07 ^{a,b,c}
4	70219.25 ^{m,n,o,p,q}	0.19 ^a	0.11 ^a	638.20 ^{p,q,r}	<LOD ^a	0.07 ^a	0.28 ^a	41448.62 ^{q,r,s,t}	0.10 ^{a,b,c,d}
5	38924.46 ^{a,b,c,d,e,f}	31.67 ^{b,c,d}	14.45 ^{b,c,d,e,f}	253.07 ^{b,c}	<LOD ^a	11.51 ^{c,d,e,f,g}	43.06 ^{b,c,d,e,f}	23018.48 ^{a,b,c,d,e}	0.10 ^{a,b,c,d}
6	50850.64 ^{c,d,e,f,g,h,i,j,k,l,m}	0.17 ^a	0.09 ^a	466.73 ^{j,k,l,m,n}	<LOD ^a	0.06 ^a	0.22 ^a	35707.88 ^{k,l,m,n,o,p,q}	0.07 ^{a,b,c,d}
7	56764.01 ^{e,f,g,h,i,j,k,l,m,n,o}	0.18 ^a	0.09 ^a	461.32 ^{h,i,j,k,l,m,n}	<LOD ^a	0.07 ^a	0.23 ^a	37133.87 ^{l,m,n,o,p,q,r}	0.08 ^{a,b,c,d}
8	62765.21 ^{i,j,k,l,m,n,o,p}	0.19 ^a	0.11 ^a	632.94 ^{o,p,q,r}	<LOD ^a	0.08 ^a	0.25 ^a	42186.81 ^{r,s,t}	0.09 ^{a,b,c,d}
9	57898.58 ^{f,g,h,i,j,k,l,m,n,o}	0.18 ^a	0.09 ^a	486.95 ^{k,l,m,n}	<LOD ^a	0.07 ^a	0.22 ^a	38079.50 ^{m,n,o,p,q,r,s}	0.07 ^{a,b,c,d}
10	33081.52 ^{a,b,c}	0.14 ^a	0.06 ^a	280.58 ^{b,c,d,e}	<LOD ^a	0.04 ^a	0.17 ^a	27645.42 ^{c,d,e,f,g,h}	0.05 ^{a,b,c}
11	69125.62 ^{l,m,n,o,p,q}	0.23 ^a	0.13 ^a	772.57 ^s	<LOD ^a	0.07 ^a	0.28 ^a	44707.41 ^{t,u}	0.11 ^{a,b,c,d,e}
12	67839.52 ^{j,k,l,m,n,o,p,q}	0.20 ^a	0.11 ^a	638.27 ^{p,q,r}	<LOD ^a	0.08 ^a	0.26 ^a	41938.12 ^{r,s,t}	0.11 ^{a,b,c,d,e}
13	28064.90 ^{a,b}	19.01 ^b	12.13 ^{b,c,d}	273.51 ^{b,c,d,e}	<LOD ^a	12.18 ^{d,e,f,g}	40.58 ^{b,c,d,e}	20349.81 ^{a,b}	0.07 ^{a,b,c}
14	29033.21 ^{a,b}	40.06 ^{d,e,f,g}	17.39 ^{f,g,h,i,j}	331.12 ^{b,c,d,e,f,g}	<LOD ^a	13.43 ^{f,g,h,i,j}	53.88 ^{h,i,j,k,l,m,n}	27488.94 ^{c,d,e,f,g,h}	0.13 ^{a,b,c,d,e}
15	21773.43 ^a	25.18 ^{b,c}	14.26 ^{b,c,d,e,f}	296.90 ^{b,c,d,e}	<LOD ^a	14.47 ^{g,h,i,j,k,l}	49.11 ^{e,f,g,h,i,j,k,l}	24903.36 ^{a,b,c,d,e,f}	0.14 ^{a,b,c,d,e}
16	38138.18 ^{a,b,c,d,e}	39.62 ^{d,e,f,g}	16.81 ^{e,f,g,h,i}	323.66 ^{b,c,d,e,f}	<LOD ^a	12.30 ^{d,e,f,g,h}	55.32 ^{j,k,l,m,n}	25073.76 ^{a,b,c,d,e,f}	0.11 ^{a,b,c,d,e}
17	42128.85 ^{b,c,d,e,f,g,h}	43.29 ^{d,e,f,g,h,i}	20.71 ^{i,j,k,l}	463.90 ^{i,j,k,l,m,n}	6.74 ^{a,b,c,d,e}	18.71 ^{k,l,m,n,o,p,q}	48.84 ^{e,f,g,h,i,j,k,l}	28107.94 ^{e,f,g,h,i}	0.15 ^{a,b,c,d,e}
18	39650.48 ^{a,b,c,d,e,f,g}	24.02 ^{b,c}	11.73 ^b	230.00 ^b	1.39 ^{a,b}	13.00 ^{e,f,g,h,i}	35.27 ^b	19845.29 ^a	0.08 ^{a,b,c,d}
19	45824.90 ^{b,c,d,e,f,g,h,i}	36.20 ^{c,d,e,f}	17.46 ^{f,g,h,i,j}	432.94 ^{f,g,h,i,j,k,l,m,n}	5.12 ^{a,b,c,d}	19.57 ^{m,n,o,p,q}	46.55 ^{d,e,f,g,h,i,j,k}	28608.18 ^{f,g,h,i,j}	0.10 ^{a,b,c,d}
20	34001.13 ^{a,b,c,d}	30.74 ^{b,c,d}	15.65 ^{e,f,g,h}	444.23 ^{f,g,h,i,j,k,l,m,n}	4.87 ^{a,b,c}	22.81 ^{q,r}	41.79 ^{b,c,d,e}	25195.06 ^{a,b,c,d,e,f}	0.07 ^{a,b,c}
21	37641.96 ^{a,b,c,d,e}	34.11 ^{c,d,e}	15.99 ^{e,f,g,h,i}	392.65 ^{e,f,g,h,i,j,k,l}	0.00 ^a	17.49 ^{i,j,k,l,m,n,o,p}	43.12 ^{b,c,d,e,f}	24872.10 ^{a,b,c,d,e,f}	0.13 ^{a,b,c,d,e}
22	45053.51 ^{b,c,d,e,f,g,h,i}	41.01 ^{d,e,f,g,h}	15.47 ^{d,e,f,g,h}	304.00 ^{b,c,d,e}	0.00 ^a	15.28 ^{g,h,i,j,k,l,m,n}	38.00 ^{b,c,d}	22058.29 ^{a,b,c,d}	0.15 ^{a,b,c,d,e}
23	74628.00 ^{o,p,q,r}	61.03 ^{j,k,l}	24.07 ^{m,n,o}	340.19 ^{b,c,d,e,f,g,h,i}	11.57 ^{b,c,d,e,f}	19.09 ^{l,m,n,o,p,q}	58.04 ^{l,m,n,o}	29749.39 ^{f,g,h,i,j,k,l}	0.17 ^{a,b,c,d,e}

Appendix B: Continued

24	68684.55 ^{k,l,m,n,o,p,q}	53.16 ^{g,h,i,j,k,l}	21.38 ^{k,l,m}	343.46 ^{b,c,d,e,f,g,h,i,j}	<LOD ^a	19.08 ^{l,m,n,o,p,q}	49.37 ^{e,f,g,h,i,j,k,l,m}	27615.77 ^{c,d,e,f,g,h}	0.25 ^{c,d,e,f}
25	74957.60 ^{o,p,q,r}	56.05 ^{i,j,k,l}	27.16 ^{o,p,q}	668.28 ^{q,r,s}	17.27 ^{e,f,g,h}	22.10 ^{p,q,r}	56.73 ^{l,m,n}	32525.14 ^{h,i,j,k,l,m,n}	0.17 ^{a,b,c,d,e}
26	86421.30 ^{q,r}	47.41 ^{f,g,h,i,j}	24.96 ^{n,o,p}	742.60 ^{r,s}	12.88 ^{c,d,e,f,g}	24.68 ^f	58.96 ^{m,n,o,p}	33732.10 ^{i,j,k,l,m,n,o,p}	0.19 ^{a,b,c,d,e}
27	73629.72 ^{n,o,p,q,r}	52.46 ^{g,h,i,j,k,l}	23.18 ^{m,n}	511.58 ^{l,m,n,o}	9.82 ^{a,b,c,d,e,f}	19.86 ^{n,o,p,q,r}	53.92 ^{h,i,j,k,l,m,n}	28074.49 ^{d,e,f,g,h,i}	0.30 ^{e,f}
28	82902.10 ^{q,r}	64.06 ^l	32.14 ^{r,s}	716.70 ^{r,s}	26.42 ^h	21.03 ^{o,p,q,r}	68.55 ^p	34566.95 ^{j,k,l,m,n,o,p,q}	0.23 ^{c,d,e,f}
29	85605.37 ^{q,r}	62.12 ^{k,l}	34.10 ^{s,t}	737.00 ^{r,s}	20.41 ^{f,g,h}	21.92 ^{p,q,r}	67.39 ^{o,p}	35368.67 ^{k,l,m,n,o,p,q}	0.26 ^{d,e,f}
30	51206.77 ^{c,d,e,f,g,h,i,j,k,l,m}	43.72 ^{d,e,f,g,h,i}	13.62 ^{b,c,d,e}	290.86 ^{b,c,d,e}	<LOD ^a	6.79 ^a	37.28 ^{b,c,d}	27483.92 ^{c,d,e,f,g,h}	0.02 ^{a,b}
31	54348.80 ^{e,f,g,h,i,j,k,l,m,n}	54.20 ^{h,i,j,k,l}	28.12 ^{p,q}	546.52 ^{n,o,p,q}	<LOD ^a	8.64 ^{a,b,c,d}	52.96 ^{g,h,i,j,k,l,m}	42429.04 ^{r,s,t}	0.02 ^{a,b}
32	56733.22 ^{e,f,g,h,i,j,k,l,m,n,o}	47.59 ^{e,f,g,h,i,j}	23.00 ^{m,n}	522.60 ^{m,n,o,p}	<LOD ^a	8.97 ^{a,b,c,d,e}	52.63 ^{f,g,h,i,j,k,l,m}	38566.32 ^{n,o,p,q,r,s}	0.03 ^{a,b}
33	50631.42 ^{c,d,e,f,g,h,i,j,k,l}	43.38 ^{d,e,f,g,h,i}	14.51 ^{b,c,d,e,f}	395.28 ^{e,f,g,h,i,j,k,l}	<LOD ^a	6.03 ^a	37.59 ^{b,c,d}	29782.27 ^{f,g,h,i,j,k,l}	0.01 ^{a,b}
34	47404.15 ^{b,c,d,e,f,g,h,i}	40.28 ^{d,e,f,g,h}	15.30 ^{c,d,e,f,g,h}	398.14 ^{e,f,g,h,i,j,k,l,m}	<LOD ^a	6.92 ^{a,b}	41.51 ^{b,c,d,e}	31432.73 ^{g,h,i,j,k,l,m}	0.01 ^a
35	49920.28 ^{c,d,e,f,g,h,i,j,k,l}	49.79 ^{f,g,h,i,j,k}	22.43 ^{l,m,n}	448.31 ^{f,g,h,i,j,k,l,m,n}	<LOD ^a	7.39 ^{a,b}	48.34 ^{e,f,g,h,i,j,k,l}	39948.61 ^{p,q,r,s,t}	0.01 ^{a,b}
36	52854.22 ^{d,e,f,g,h,i,j,k,l,m}	53.40 ^{g,h,i,j,k,l}	22.21 ^{l,m,n}	527.10 ^{n,o,p}	<LOD ^a	8.09 ^{a,b,c}	44.51 ^{b,c,d,e,f,g,h}	38824.51 ^{o,p,q,r,s,t}	0.03 ^{a,b}
37	81530.14 ^{p,q,r}	65.27 ^l	28.81 ^{q,r}	453.59 ^{g,h,i,j,k,l,m,n}	19.94 ^{f,g,h}	20.97 ^{o,p,q,r}	56.11 ^{k,l,n,n}	32184.64 ^{h,i,j,k,l,m,n}	0.39 ^f
38	85413.15 ^{p,q,r}	65.86 ^l	36.4 ^t	697.17 ^{r,s}	23.48 ^{g,h}	19.55 ^{m,n,p,q}	63.39 ^{n,o,p}	35094.83 ^{k,l,m,n,o,p,q}	0.20 ^{b,c,d,e,f}
39	61334.41 ^{h,i,j,k,l,m,n,o}	40.66 ^{d,e,f,g,h}	23.06 ^{m,n}	533.34 ^{n,o,p}	15.99 ^{d,e,f,g,h}	19.86 ^{n,o,p,q}	54.78 ^{i,j,k,l,m,n}	35216.12 ^{k,l,m,n,o,p,q}	<LOD ^a
40	62320.59 ^{i,j,k,l,m,n,o,p}	46.48 ^{e,f,g,h,i}	21.57 ^{k,l,m,n}	489.11 ^{k,l,m,n}	15.64 ^{c,d,e,f,g,h}	18.52 ^{k,l,m,n,o,p,q}	40.37 ^{b,c,d,e}	32742.21 ^{h,i,j,k,l,m,n,o}	<LOD ^a
41	49071.05 ^{c,d,e,f,g,h,i,j}	34.30 ^{c,d,e}	16.79 ^{e,f,g,h,i}	258.63 ^{b,c,d}	11.64 ^{b,c,d,e,f}	15.77 ^{g,h,i,j,k,l,m,n}	45.40 ^{c,d,e,f,g,h,i}	28283.65 ^{e,f,g,h,i}	<LOD ^a
42	51024.34 ^{c,d,e,f,g,h,i,j,k,l,m}	35.89 ^{c,d,e,f}	11.89 ^{b,c}	94.13 ^a	7.54 ^{a,b,c,d,e}	15.76 ^{g,h,i,j,k,l,m,n}	36.50 ^{b,c}	26153.83 ^{b,c,d,e,f,g}	<LOD ^a
43	48801.52 ^{c,d,e,f,g,h,i,j}	33.64 ^{c,d,e}	16.31 ^{e,f,g,h,i}	335.90 ^{b,c,d,e,f,g,h}	10.80 ^{a,b,c,d,e,f}	15.99 ^{g,h,i,j,k,l,m,n}	36.72 ^{b,c,d}	27613.73 ^{c,d,e,f,g,h}	<LOD ^a
44	56516.98 ^{e,f,g,h,i,j,k,l,m,n,o}	39.74 ^{d,e,f,g}	19.38 ^{i,j,k,l}	374.08 ^{c,d,e,f,g,h,i,j,k}	14.49 ^{c,d,e,f,g}	18.90 ^{l,m,n,o,p,q}	46.01 ^{c,d,e,f,g,h,i,j}	31626.10 ^{g,h,i,j,k,l,m}	<LOD ^a
45	49685.93 ^{c,d,e,f,g,h,i,j,k}	35.25 ^{c,d,e}	18.29 ^{g,h,i,j,k}	251.88 ^{b,c}	14.14 ^{c,d,e,f,g}	18.06 ^{j,k,l,m,n,o,p}	44.14 ^{b,c,d,e,f,g,h}	22823.12 ^{a,b,c,d,e}	<LOD ^a
46	58362.87 ^{g,h,i,j,k,l,m,n,o}	43.21 ^{d,e,f,g,h,i}	21.05 ^{k,l,m}	385.95 ^{e,f,g,h,i,j,k,l}	15.01 ^{c,d,e,f,g}	19.54 ^{m,n,o,p,q}	43.65 ^{b,c,d,e,f,g}	30423.63 ^{f,g,h,i,j,k,l}	<LOD ^a
47	52677.56 ^{d,e,f,g,h,i,j,k,l,m}	36.28 ^{c,d,e,f}	18.53 ^{h,i,j,k}	379.67 ^{d,e,f,g,h,i,j,k}	12.14 ^{b,c,d,e,f}	17.78 ^{j,k,l,m,n,o,p}	41.67 ^{b,c,d,e}	29619.24 ^{d,g,h,i,j,k}	<LOD ^a
48	50722.53 ^{c,d,e,f,g,h,i,j,k,l}	35.42 ^{c,d,e}	16.90 ^{e,f,g,h,i}	344.82 ^{b,c,d,e,f,g,h,i,j}	11.84 ^{b,c,d,e,f}	16.87 ^{h,i,j,k,l,m,n,o}	40.59 ^{b,c,d,e}	27800.53 ^{c,d,e,f,g,h,i}	<LOD ^a
49	40121.31 ^{a,b,c,d,e,f,g}	30.37 ^{b,c,d}	15.67 ^{e,f,g,h}	365.65 ^{c,d,e,f,g,h,i,j,k}	10.78 ^{a,b,c,d,e,f}	14.09 ^{g,h,i,j,k}	39.73 ^{b,c,d,e}	27789.48 ^{c,d,e,f,g,h,i}	<LOD ^a
50	43671.07 ^{b,c,d,e,f,g,h,i}	31.08 ^{b,c,d}	16.44 ^{e,f,g,h,i}	300.39 ^{b,c,d,e}	11.05 ^{b,c,d,e,f}	15.16 ^{g,h,i,j,k,l,m}	44.55 ^{b,c,d,e,f,g,h}	27338.29 ^{c,d,e,f,g,h}	<LOD ^a

Appendix C

Habitat, ecology and feeding habit of fish (Ezaimah, 2016)

No.	Fish name	Habitat and ecology	Feeding habit
1	<i>Oxygaster anomalura</i>	-pelagic species, found at the surface water	-Carnivorous (fish, insect)
2	<i>Luciosoma setigerum</i>	-found in slowly-flowing rivers with substrates of leaf litter and submerged woody structures.	-Omnivorous (insect, plant)
3	<i>Leiocassis micropogon</i>	-dermesal species, inhabit sandy or rocky bottom.	-Carnivorous (fish, insect)
4	<i>Barbonymus collingwoodii</i>	-inhabits streams with fast flowing and relatively cold waters, over stones and gravel, in upland areas.	-Herbivorous (plant, algae)
5	<i>Hampala bimaculata</i>	-found in clear, well-oxygenated, running waters with substrates of sand, gravel, rock or mud, typically flowing through tropical forest.	-Omnivorous (fish, insect, plant)
6	<i>Hemibagrus wyckii</i>	-demersal species, found in larger rivers with fast flowing water over muddy bottom.	-Omnivorous (fish, plant)
7	<i>Hemibagrus planiceps</i>	-demersal species, found in the river basins with sandy or rocky bottom.	-Omnivorous (fish, aquatic invertebrate, nematode, plants)
8	<i>Barbonymus schwanenfeldii</i>	-found in medium size river, especially tributaries.	-Herbivorous (plant, algae)
9	<i>Tor tambroides</i>	-upper stream with clean, fast flowing water, rocky, high oxygen.	-Omnivorous (fruit, plant, fish)
10	<i>Lobocheilos bo</i>	-found in shallow, fast flowing areas with gravel substrate.	-Herbivorous (algae, plant)
11	<i>Tor dourenensis</i>	-upper stream with clean, fast flowing water, rocky, high oxygen.	-Omnivorous (fish, insect, plant)
12	<i>Cyclocheilichthys apogon</i>	-bentopelagic species, typically found around surfaces, such as plant, leaves, branches and tree roots.	-Omnivorous (fish, insect, plant)
13	<i>Osteochilus schelegelii</i>	-bentopelagic species, occur from midwater to bottom depth in large and medium sized rivers.	-Herbivorous (detritus, plant)
14	<i>Pengasius macronema</i>	-bentopelagic species	-Omnivorous (insect, plant)
15	<i>Osteochilus vittatus</i>	-bentopelagic species, found in large streams with slow current and muddy to sandy substrate	-Omnivorous (fish, insect, detritus)
16	<i>Luciosoma spiloptera</i>	-bentopelagic species, found in high gradient rivers with rocky, stony or gravel bottoms.	-Omnivorous (fish, insect, plant)
17	<i>Lobiobarbus fasciatus</i>	-bentopelagic species.	-Carnivorous (insect, aquatic invertebrates)
18	<i>Pseudolais micronemus</i>	-bentopelagic species.	-Omnivorous (fish, insect, plant, fruit seed)
19	<i>Macrognaathus maculatus</i>	-bentopelagic species, often occurs in clear water over rocky bottom in flowing streams.	-Carnivorous (insect)
20	<i>Rasbora borneensis</i>	-bentopelagic species.	-Carnivorous (insect)
21	<i>Anematichthys armatus</i>	-bentopelagic species, occurs at midwater to bottom levels in rivers and streams.	-Carnivorous (zooplankton, small crustaceans, chironomids, and insect larvae)
22	<i>Channa lucius</i>	-bentopelagic species, often found in areas with plenty of aquatic vegetation and submerged woody plants.	-Carnivorous (fish, prawn)

Appendix D

Photos of the fish species caught



Hampala bimaculata



Luciosoma setigerum



Barbonymus schwanenfeldii



Osteochilus vittatus



Tor dourenensis



Pseudolais micronemus



Hemibagrus planiceps



Hemibagrus wyckii



Cyclocheilichthys apogon

Appendix E
Photos of the Baleh River



