CONCENTRATION OF Cd, Cr, Cu, Pb, AND Zn IN FISHES ALONG SEMBULAN RIVER

KWONG YE LENG

THIS DISSERTATION IS SUBMITTED TO FULFILL THE REQUIREMENTS TO OBTAIN A DEGREE IN BACHELOR OF SCIENCE WITH HONOUR

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KAMSIA BUDIN
Nama Penyelia

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I declare that this dissertation entitled "CONCENTRATION OF Cd, Cr, Cu, Pb, AND Zn IN FISHES ALONG SEMBULAN RIVER" is the results of my own work except as cited in the references.

KWONG YE LENG
(BS 07110015)

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1. **SUPERVISOR**
   (MISS KAMSIA BUDIN)

2. **EXAMINER 1**
   (MISS SITI AISHAH MOHD ALI)

3. **EXAMINER 2**
   (MISS FARRAH ANIS FAZLIATUL ADNAN)

4. **DEAN**
   (PROF. DR. MOHD. HARUN ABDULLAH)

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ABSTRACT

This study was conducted to determine the concentrations of Cd, Cr, Cu, Pb, and Zn in fishes collected along Sembulan River. A total of four sampling points were set along the river according to their point source of pollution. Water and fish samples were collected along the river and dissected to different parts which were the gills, liver, gastro intestine, head, and muscle. All the organs were digested using nitric acid digestion method followed by analysis using ICP-OES. The trends of the heavy metals concentrations in gills, liver, gastro intestine, head, muscle, and water were analyzed. Overall, the heavy metals concentrations in all the fish organs showing the similar trends. The trends of heavy metals concentrations in gills and liver are the same which is Zn>Cr>Pb>Cu>Cd, while the trend of gastro intestine and muscle are the same which is Zn>Cr>Cu>Pb>Cd. The overall trend of head is Zn>Cr>Cu>Cd>Pb. In this study, concentrations of Zn were found to be the highest in all organs and water samples. The metals concentration in head and muscle of fishes along Sembulan River had exceeded the permissible level of USEPA. The good news is, the heavy metals concentrations in head and muscle of the fishes are still away low compare to *Akta Makanan 1985* in Malaysia. Based on this, we can conclude that the fishes in Sembulan River are safe for human consumption, but, precautions still needed for other pollutants and metals.

KEPEKATAN Cd, Cr, Cu, Pb DAN Zn DI DALAM IKAN SEPANJANG SUNGAI SEMBULAN

ABSTRAK

Kajian ini dijalankan untuk menentukan kepekatan logam Cd, Cr, Cu, Pb, dan Zn dalam ikan sepanjang Sungai Sembulan. Sebanyak empat lokasi persampelan telah ditentukan berdasarkan kepada sumber pencemaran lokasi tersebut. Sampel ikan dan air dikumpulkan di sepanjang sungai tersebut dan organ-organ seperti insang, hati, sistem pencernaan, kepala, dan otot telah dibedahkan. Organ-organ yang telah dibedah akan dicerna dengan menggunakan kaedah pencernaan asid nitrik dan diikuti dengan analisis kandugan logam menggunakan ICP-OES. Trend kepekatan logam berat di insang, hati, sistem pencernaan, kepala, otot, dan air telah dianalisis. Secara keseluruhan, kepekatan logam berat dalam semua organ ikan menunjukkan trend yang sama. Trend dalam insang dan hati adalah sama di mana Zn> Cr> Pb> Cu> Cd manakala trend dalam sistem pencernaan dan otot adalah sama iaitu Zn> Cr> Cu> Pb> Cd. Trend di bahagian kepala adalah Zn> Cr> Cu > Cd> Pb secara keseluruhan. Dalam kajian ini, kepekatan Zn adalah yang tertinggi dalam semua organ dan sampel air. Kepekatan logam di kepala dan otot ikan di sepanjang Sungai Sembulan telah melebihi tahap yang dibenarkan dari USEPA. Khabar baik adalah kepekatan logam berat di kepala dan otot ikan-ikan masih jauh lebih rendah berbanding dengan Akta Makanan 1985 di Malaysia. Berdasarkan ini, kita boleh menyimpulkan bahawa ikan-ikan di Sungai Sembulan adalah selamat untuk dimakan oleh manusia, namun, tindakan pencegahan masih perlu untuk bahan pencemar and logam lain.

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Cd	cadmium
Cr	chromium
Cu	copper
Pb	lead
Zn	zinc
CO_3^{2-}	carbonate ion
OH^-	hydroxide ion
Cl^-	chloride ion
SO_4^{2-}	sulphate ion
$Cd(H_2O)_6^{2+}$	cadmium dodecahydro ion
$CdCl_2$	cadmium chloride
Cr^{3+}	chromium ion
$Cr(OH)^{2+}$	chromium hydroxide ion
$Cr(OH)_2^+$	chromium hydroxide II ion
$Cr(OH)_4^-$	chromium hydroxide IV ion
CrO_4^{2-}	chromate
$Cr_2O_7^{2-}$	dichromate
$PbCO_3$	lead carbonate
CO_2	carbon dioxide
NO_3^-	nitrate ion
NH_4^+	ammonium ion

Ca^{2+}	calcium ion
Mg^{2+}	magnesium ion
HNO_3	nitric acid
H_2SO_4	sulphuric acid
HCl	hydrochloric acid
H_2O	water
EtOH	ethanol
Et_2O	diethyl ether
MeOH	methanol
AcOH	acetyl acid
EtOAc	ethyl acetate
NH_4OH	ammonium hydroxide
NH_3	ammonia
ATSDR	Agency for Toxic Substances and Disease Registry
DOE	Department of Environment
FAO	Food and Agriculture Organisation
USEPA	United State Environmental Protection Agency
WHO	World Health Organisation
PTWI	permissible tolerable weekly intake
CAC	Codex Alimentarius Comission
NAS-NRC	National Academy of Sciences-National Research Council
ICP-OES	Inductively Coupled Plasma-Optical Emission Spectrometry
alk.	alkaline

liq.	Liquid
sl.	slightly
sol.	soluble
conc.	concentrated
insol.	insoluble
cryst.	crystal
subl.	sublime
NR	not recorded
ND	no data
ppb	part per billion
ppm	part per million
kPa	kilopascal
mg	milligram
mL	milliliter
µm	micrometer
µL	microliter
µg/kg	microgram per kilogram
mg/kg	milligram per kilogram
mg/L	milligram per liter
µg/L	microgram per litre
g/cm^3	gram per sentimeter cube
&	and
%	percent

$\geq$	greater than or equal
$>$	greater than
°C	degree celcius
M	Molarity
ft	feet
km	kilometre
DO	dissolved oxygen
EC	Electrical conductivity

CHAPTER 1

INTRODUCTION

1.1 Metal pollution

Metals can originate both from natural and anthropogenic sources in receiving water bodies. Heavy metals can enter a water supply by industrial and consumer waste, or even from acidic rain breaking down soils and releasing heavy metals into streams, lakes, rivers, and groundwater (Kiliç, 2009). Some of the metals are also essential for animals and human health. These metals such as Zn, Fe, Sc, and Bo are known as micronutrient. However, they could become harmful to the aquatic organism when their concentrations are exceed the toxicity threshold and these are typically caused by excessive discharged and emissions of metals from man-made sources (Novotny, 1995).

Heavy metals that entered into the aquatic environment will undergoes bioaccumulation which the concentration of the heavy metal will be increase in a living tissue as it moves through the food chain. As a result, top predators in a food chain may have concentrations of heavy metals in their tissues that are significantly higher than the concentrations found in water (EPA, 2008).

Aquatic organisms such as fish have been used in comparative monitoring of pollution effects in river systems. Furthermore, biomonitoring approach is also has proved to be promising as a reliable means of quantifying biological effects of complex effluents (He *et al.*, 1998). The heavy metal concentration in fish tissues may reflects the time of exposure of the metals via water or food. Besides, it also can demonstrate the current situation of the animals before toxicity affects the ecological balance of populations in the aquatic environment.

Fishes are a major component in most aquatic environment, and thousand of species occur in streams, river, lakes, and estuaries (APHA, 1995). They are an important part of healthy diet and also a lean, low-calorie source of protein. Fishes may exposed to the pollutants in the water or through the food that they eat. The pollutants that entered into their body may stored in skin, fat, internal organs, or muscles. Therefore, eat fish that taken from polluted location such as Sembulan River might hazardous to human health.

Sembulan River is located in Sembulan, Kota Kinabalu. This river passed through various housing areas and commercial areas in Kota Kinabalu and finally discharging to South China Sea. Areas nearby Sembulan River contribute to the pollution of the river or the metal concentration as there is more than 15 main tributaries and major drains flow into it (Sundari *et al.*, 2006). The river has been polluted by uncontrolled discharge of mainly domestic wastewater originated from the surrounding towns.

1.2 Importance of study

Sembulan River is located in Sembulan, Kota Kinabalu, Sabah. This river receives most of the effluent from the commercial area in Kota Kinabalu. Besides that, the village nearby which is *Kampung Sembulan* is still discharge their waste water into the river. Furthermore, the villagers from Sembulan River will also catch the fishes in the river for consumption or to be sold in the market. Therefore, it is important to determine the heavy metals concentrations in fishes in the river which plays an important role in food supply and economic sources for the villagers nearby.

1.3 Scope of study

The main focus of this study is to determine the concentration of heavy metals in fish gills, muscles, liver, and gastro intestine and compare the concentration of heavy metals especially contained in the fish muscle with the permissible level that has been set by the Malaysian government and United Stated Environmental Protection Agency (USEPA). However, water samples from the sampling site were also obtained in order to get the correlation between the concentrations of heavy metal in fishes with the concentrations of heavy metal in water.

1.4 Objective of study

i. To determine the Cd, Cr, Cu, Pb, and Zn concentrations in fishes of Sembulan River.
ii. To compare the concentrations of Cd, Cr, Cu, Pb, and Zn in fish head and muscle with the permissible level from *Akta Makanan 1985* and United State Environmental Protection Agency (USEPA).
iii. To determine the correlation between heavy metal in fishes and the concentration of heavy metal in river water.

CHAPTER 2

LITERATURE REVIEW

2.1 Heavy metals and its toxicity effects

Heavy metal poses a serious problem to environment because they do not decompose or become eliminated from the environment. In fish, the toxic effects of heavy metals may influence physiological functions, individual growth rates, reproduction and mortality (Amundsen *et al.*, 1997). The characteristic, properties, and toxicity of the studied metals thoroughly were discussed in this chapter.

2.1.1 Cadmium

Cadmium (Cd) is a metallic element in Group IIB of the periodic table which has the atomic number 48, an atomic weight of 112.41, and a valence of 2. Its natural isotopes are ^{106}Cd, ^{108}Cd, ^{110}Cd, ^{111}Cd, ^{112}Cd, ^{113}Cd (radioactive), ^{114}Cd, and ^{116}Cd (Filov *et al.*, 1993). Cadmium can be found in the earth crust associated with zinc, lead, and copper ores (ATSDR, 2008). Pure cadmium is a soft metal in other word it is malleable and ductile with silver-white in colour.

Cadmium has no allotropic modifications. In air, it is covered with and oxide (CdO) film that will protects it from further oxidation. It exhibits the oxidation state +2 in most compounds (Filov *et al.*, 1993). Cadmium will burns to the oxide CdO when it is strongly heated. When alkaline is added to the solution of cadmium salt, the hydroxide $Cd(OH)_2$ precipitates as the solution of cadmium salts have an acid reaction. Cadmium halides are readily soluble in water (Filov *et al.*, 1993). Cadmium forms a large number of complex compounds. The physicals and chemical properties of cadmium and its compounds are listed in Appendix A.

Cadmium is emitted to soil, water, and air by non-ferrous metal mining and refining manufacture and application of phosphate fertilizers, fossil fuel combustion, and waste incineration and disposal (ATSDR, 2008). In water, it is exists as the hydrated ion or as ionic complexes with other inorganic or organic substances. The soluble forms of cadmium migrate in water whereas the insoluble forms are immobile and will deposit and accumulate in sediments. Cadmium can be accumulated in aquatic organisms. Some cadmium that entered into the water is taken up and concentrated in the liver and kidney of the aquatic animals that eat the aquatic plants. Cadmium concentrates in freshwater and marine animals to concentrations hundreds to thousands of times higher than in the water (EPA, 1979). The average abundance of Cadmium in streams is 1 µg/L whereas in groundwater it is from 1 to 10 µg/L (APHA, 1995).

Cadmium may be released to water by natural weathering processes, discharge from industrial facilities or sewage treatment plants, atmospheric deposition, leaching from landfills or soil, or phosphate fertilizers. Smelting of nonferrous metal ores has been estimated to be the largest anthropogenic source of cadmium released into the aquatic environment (ATSDR, 2008). A large proportion of the cadmium load in the aquatic environment is due to diffuse pollution originating from many different sources rather than from point sources (ATSDR, 2008).

Cadmium is more mobile in aquatic environments than most other heavy metals, for an example, lead (Pb). In most natural surface waters, the affinities of complexing ligands for cadmium generally follow the order of humic acids > CO_3^{2-} > OH^- ≥ Cl^- ≥ SO_4^{2-} (EPA, 1979). In unpolluted natural waters, most cadmium transported in the water column will exist in the dissolved state as the hydrated ion $Cd(H_2O)_6{}^{2+}$. Minor amounts of cadmium are transported with the coarse particulates, and only a small fraction is transported with the colloids. Cadmium can be removed from solution by exchange of cadmium for calcium in the lattice structure of carbonate minerals in unpolluted water.

In polluted or organic-rich waters, adsorption of cadmium by humic substances and other organic complexing agents plays a dominant role in transport, partitioning, and remobilization of cadmium (EPA, 1979). Cadmium concentration in water is inversely related to the pH and the concentration of organic material in the water (EPA, 1979). The solubility of Cadmium in natural water is controlled by carbonate equilibria (APHA, 1995). Aqueous cadmium is not strongly influenced by the oxidizing or reducing potential of the water as it is exists only in the +2 oxidation state in water. However, cadmium may form cadmium sulphide under reducing conditions which is poorly soluble and tends to precipitate. The hardness or alkalinity of the natural water will affect the permittivity of Cadmium, in which, the softer the water, the lower the permitted level of Cadmium (APHA, 1995).

Cadmium exposure occurs mainly through oral route and inhalation. It is extremely toxic and accumulates in kidneys and liver, with prolonged intake at low levels sometimes leading to dysfunction of the kidneys (APHA, 1995). Most of the cadmium content in fish or other seafood is highly absorbable in $CdCl_2$ form; in humans, the efficiency of gastrointestinal absorption of cadmium has been reported to be approximately 3–8% of the ingested load (Castro-Gonzalez and Mendez-Armenta, 2008).

Cadmium induces renal tubular dysfunction, proteinuria and chronic renal insufficiency in kidney as well as aortic and coronary artherosclerosis, increases cholesterol and free fatty acids in heart. Besides that, it also affects lung (fibrosis), skeletal system, testes, placenta, brain and the central nervous system (CNS) of children which causing neurological disorder such as learning disabilities and hyperactivity may occur (Gonzalez and Armenta, 2008).

2.1.2 Chromium

Chromium is the first element which is a metallic element in Group VIB in the periodic table with atomic number of 24, an atomic weight of 51.99, and valence of 0 and 2 through 6 (APHA, 1995). Chromium is a naturally-occurring element found in rocks, animals, plants, and soil. But, it is found chiefly in chrome-iron ore ($FeO \cdot Cr_2O_3$). The three main forms of chromium are chromium(0), chromium(III), and chromium(VI). However, chromium(0) does not occur naturally and the divalent state is relatively unstable and is readily oxidized to the trivalent state in chromium compounds are stable in this state. Small amounts of chromium(III) are considered to be a necessity for human health (ATSDR, 2008).

Chromate(IV) which is also hexavalent is the second most stable state. However, hexavalent chromium rarely occurs naturally, but is produced from anthropogenic sources (ATSDR, 2008). Chromium in the hexavalent state only will occur naturally in the rare mineral crocoite (PbCrO4) (ATSDR, 2008). Chromium can change from one form to another in water and soil, depending on the conditions present.

In natural waters trivalent chromium exists as Cr^{3+}, $Cr(OH)^{2+}$, $Cr(OH)_2^{+}$, and $Cr(OH)_4^{-}$ whereas in hexavalent form chromium exists as CrO_4^{2-} and as $Cr_2O_7^{2-}$ (APHA, 1995). Cr^{3+} would be expected to form strong complexes with amines, and would be

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