UNIVERSITI PUTRA MALAYSIA

DISTRIBUTION OF SELECTED TRACE ELEMENTS AND STABLE ISOTOPES AT VARIOUS TROPHIC LEVELS OF SUNGAI PULAI ESTUARY, JOHOR, MALAYSIA

AQILAH MUKHTAR

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By

`AQILAH BINTI MUKHTAR

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

December 2015

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DEDICATION

The thesis is especially dedicated to my family

late father, Mukhtar bin Takei, beloved mother, Saodah binti Abdul Talib, lovely husband Che Mohd Nur Yusran bin Che Mohd Norodi

and

strong supporters from my siblings, Nor Faizah binti Mukhtar, Nabihah binti Mukhtar and Dr. Ahmad Faiq bin Mukhtar.

May Allah bless and protect them all.

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

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December 2015

Chairman : Syaizwan Zahmir Zulkifli, PhD
Faculty : Science

Trace metals in food web of seagrass ecosystem of Sungai Pulai in South of Johor was determined using combination of stable isotopes and heavy metals analyses. Stable isotope analysis was applied in order to construct the food web of seagrass area referred to the scientific ratios of $\delta^{13}C$ and $\delta^{15}N$. In general, complex structure of seagrass ecosystem consisted of five trophic levels. Primary producers were located in the first trophic level, while second and third trophic levels consist of echinoderms and mollusks. Fish and mollusk made up in trophic level four and five. Selected trace metals (Co, Ni, As, Cd and Pb) concentration were evaluated in sediments and marine organisms to determine the metal pollution status effected from the anthropogenic activities around the ecosystem. In this study, total heavy metal and sequential extraction technique were used to determine the concentrations of metals in sediments. Result showed concentration pattern of trace metals for total extraction technique of sediment can be arranged as Ni > Pb > As > Co > Cd. While, arrangement of trace metals for sequential extraction technique such follows: Pb > Ni > As > Co > Cd. Distribution of trace metals in sediment come from both naturally occurrence and anthropogenic activities. However, most of metal sources present are naturally on the earth's crust. After having assessment of Enrichment Factor (EF), Geo-accumulation index (I_{geo}), Contaminant Factor (CF), Pollution Load Index (PLI) and Sediment Quality Guideline (SQGs), low concentrations of metal were detected in sediment collected from seagrass bed and categorized as unpolluted. However, some of benthic organisms have been affected with metal concentrations in the coastal environment. Eggs of *Tachypleus gigas* consisted of As concentration over the threshold, while Family Pinnidae and *Placuna* sp. have high concentration of Cd and As. Stable isotope and metal analysis are perfectly combination to evaluate the biomagnification and biodilution of trace metal. Different in accumulations of metals in organisms depend on assimilation efficiencies and efflux rates of metals in organism. Movement of metals in seagrass ecosystem involved biota that correlated with stable isotope ratio of $\delta^{13}C$ and $\delta^{15}N$. All selected trace metals have biodiluted throughout the seagrass food web. The concept of mobility of trace metals in seagrass ecosystem was clearly designed across the trophic level.

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

TABURAN LAGAM SURIH TERPILIH DAN STABIL ISOTOP PADA PARAS TROFIK PELBAGAI DI MUARA SUNGAI PULAI, JOHOR, MALAYSIA

Oleh

`AQILAH BINTI MUKHTAR

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Pengerusi : Syaizwan Zahmir Zulkifli, PhD
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Logam berat dalam rangkaian makanan di ekosistem rumput laut Sungai Pulai di Selatan Johor ditentukan menggunakan kombinasi analisis-analissis isotop stabil dan logam berat. Isotop stabil telah digunakan untuk membina rangkaian makanan rumput laut berdasarkan nisbah saintifik $\delta^{13}C$ and $\delta^{15}N$. Umumnya, struktur ekosistem rumput laut yang kompleks mempunyai lima tahap trofik. Pengeluar utama terletak di tahap trofik pertama, manakala tahap trofik kedua dan ketiga terdiri daripada ekinoderma dan moluska. Tahap tropik keempat dan kelima terdiri daripada moluska dan ikan. Kepekatan logam berat terpilih (Co, Ni, As, Cd dan Pb) yang terkandung dalam tanah dan organisma laut dinilai untuk menentukan status pencemaran logam berat kesan daripada aktiviti antropogenik di sekeliling ekosistem yang terhad. Kajian ini menggunakan dua teknik yang berbeza untuk menentukan kepekatan logam berat dalam tanah; teknik pengekstrakan keseluruhan logam berat dan teknik pengekstrakan berturutan. Secara umum, corak logam berat untuk teknik pengekstrakan keseluruhan mempunyai turutan seperti berikut: Ni > Pb > As > Co > Cd. Susunan logam berat untuk teknik pengekstrakan berturutan seperti berikut Pb > Ni > As > Co > Cd. Taburan logam berat dalam tanah terhasil secra semula jadi dan aktiviti antropogenik. Namun begitu, kebanyakan logam adalah terjadi secra semulajadi di kerak bumi. Selepas penilaian tanah menggunakan faktor pengayaan (EF), geo-pengumpulan indeks (I_{geo}), faktor pencemaran (CF), beban pencemaran indeks (PLI) dan garis panduan kualiti tanah (SQGs), kepekatan logam yang rendah telah dikenalpasti dalam tanah dan boleh dikategori sebagai tidak tercemar. Walaubagaimanapun, sesetengah organisma bentik telah dicemari dengan kepekatan logam berat in kawasan perairan. Telur *Tachypleus gigas* mengandungi kepekatan logam As yang melebihi ambang dan Keluarga Pinnidae dan *Placuna* sp. mempunyai kepekatan Cd yang tinggi. Biopengumpulan logam boleh berlaku melalui tahap trofik. Dalam kajian ini, kesemua logam mempunyai biopencairan melalui rantain makanan di kawasan rumput laut. Isotope stabil dan analisis logam adalah kombinasi yang sempurna untuk menilai biopembesaran dan biopencairan logam berat. Perbezaan pengumpulan logam dalam organisma bergantung kepada keberkesanan assimilasi dan kadar pengurangan logam dalam organism. Pergerakan logam dalam ekosistem rumput laut melibatkan hidupan yang mempunyai hubungkait dengan nisbah isotop stabil, $\delta^{13}C$ dan $\delta^{15}N$. Konsep peralihan logam berat dalam ekosistem rumput laut jelas dipamerkan bersama tahap trofik.

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

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LIST OF ABBREVIATIONS

ANOVA	Analysis of variance
ATSDR	Agency for Toxic Substances and Disease Registry
BMF	Biomagnification factors
CF	Contamination factor
CF-IRMS-EA	Continuous Flow Isotopic Ratio Mass Spectrometer with Elemental Analyzer
CO_2	Carbon dioxide
CRM	Certified Reference Material
d. w.	Dry weight
DIC	Dissolved inorganic carbon-pool
DO	Dissolve oxygen
EF	Enrichment factor
EFLE	Easily, freely or leachable and exchangeable
EPA	Environmental Protection Agency
ERL	Effects range-low
ERM	Effects range-median
FAO	Food and Agriculture Organization
FWMF	Food web magnification factors
HCl	Hydrochloric acid
$HClO_4$	Perchloric acid
HNO_3	Nitrite acid
IARC	International Agency for Research on Cancer
ICP-MS	Inductively coupled plasma- mass spectrometry
I_{geo}	Index geoaccumulation
IPCS	International Programme on Chemical Safety
IRMS	Isotopes Ratio Mass Spectrometry
JECFA	Joint FAO/WHO Expert Committee on Food Additives
MMEA	Malaysian Maritime Enforcement Agency
N_2	Nitrogen gas
$NaSO_4$	Sodium hydroxide
$NH_2OH.HCl$	Hydroxyl-ammonium chloride
NH_4CH_3COO	Ammonium acetate
NOAA	National Oceanic and Atmospheric Administration's
NPL	National Priorities List
NTS	Non-Traditional Security
PLI	Pollution load index
POM	Particulate organic matter
PTDI	Provisional tolerable daily intake
RfD	Reference dose
RSIS	Rajaratnam School of International Studies
SD	Standard deviation
SET	Sequential extraction technique
SQGs	Sediment quality guideline
TDS	Total dissolve solid
TL	Trophic level
UNEP	United Nations Environment Programme
USFDA	United States Food and Drug Administration
VPDB	Vienna Pee Dee Belemnite

WHO	World Health Organization

CHAPTER 1

INTRODUCTION

1.1 General Introduction

Environmental quality is usually linked with human population and anthropogenic activities. Figure 1.1 shows human population growth occurring in the states of Peninsular Malaysia. Increasing human population number enhance waste load discharges to land and subsequently release to the rivers and marine water system (Zelina et al., 2011). Development of natural areas is essential to support human population and people requirements. However, improper planning and mitigation program have resulted many unexpected environmental impacts such as flash flood, species extinction from logging activities, haze and others environment issues. Rapid urban development, domestic and agriculture activities in Johor release significant amount of pollutants into the surrounding create environmental stress on ecosystem and human health (Table 1.1).

Most of the land-based and sea-based pollutants released into the environment will be ended and accumulated in the marine environment. For example, the factories releasing polluted air into the atmosphere then the rain wash the waste accumulated down into the sea water. Otherwise, the industries discharge effluent into the rivers will have end flow in the marine water. Such the rapid development of the coastal area, the maritime activities example the shipping activities for the fisheries and international trade enhance the maritime traffic potential to dump oil and accidental spills other wastes into marine water. For example, in 2010 Bunga Kelana 3 ship was on its way from Bintulu to a Petronas refinery in Malacca was spillage of about 2,000 tonnes of crude oil (New Straits Times, 2010). Nowadays, the oil spills on the Strait of Johor was common and not the new issue to be countered.

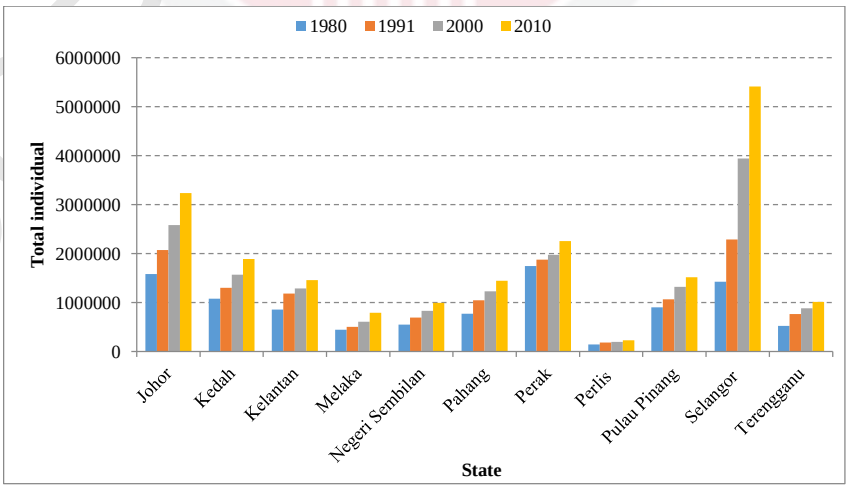

Figure 1.1. Population in the West Peninsular of Malaysia from 1980 to 2010
(Source: Department of Statistic, 2010)

Table 1.1. Sources of land-based pollution in Johor

Source	Source
▪ Earthworks ▪ Wet markets ▪ Abattoirs / slaughterhouses ▪ Chicken processing stalls ▪ Landfills near rivers ▪ Squatters on river reserves ▪ Shared accommodation for workers at construction sites ▪ Plastic bags	▪ Aging individual septic tanks ▪ Old sewage treatment plants ▪ Restaurants and food stalls ▪ Sand mines in and upstream of rivers ▪ Pig farming areas ▪ Aquaculture in tidal flats ▪ Logging of permanent forest reserves

(Source: Zelina et al., 2011)

According to the report published by Centre for Non-Traditional Security (NTS) and Rajaratnam School of International Studies (RSIS), rapid development for industrialisation and urbanisation in Iskandar Malaysia, South of Johor increase pollution transported through runoff (Hangzo and Cook, 2014). Plus, changes in land used give the result in the changes of water quality endanger the seagrass meadows that support revering of mangroves tract and endangered species such as the seahorse, dugong and sea turtle. Commercially important fishes, crabs and prawns, and invertebrates' species such as sea stars, sea cucumbers and sea anemone that thrive in the seagrass patches could be affected as well. The function of food web in coastal area could to be disturbed concomitant with the anthropogenic activities. The illustration on how the pollutants release in the aquatic ecosystem shown in the Figure 1.2.

Monitoring activities in contaminated coastal area need to be carried out as fast as the developing activities of industrialisation and urbanisation in order to have proper touch in counter the environmental problems in the short-term. Thus, scientific study background focus on the current situation must be referred. Present study has focused to provide the current status of the trace elements widely distributed in South of Johor. This baseline can be referred for further study on controlling and monitoring the study area affects from the anthropogenic activities.

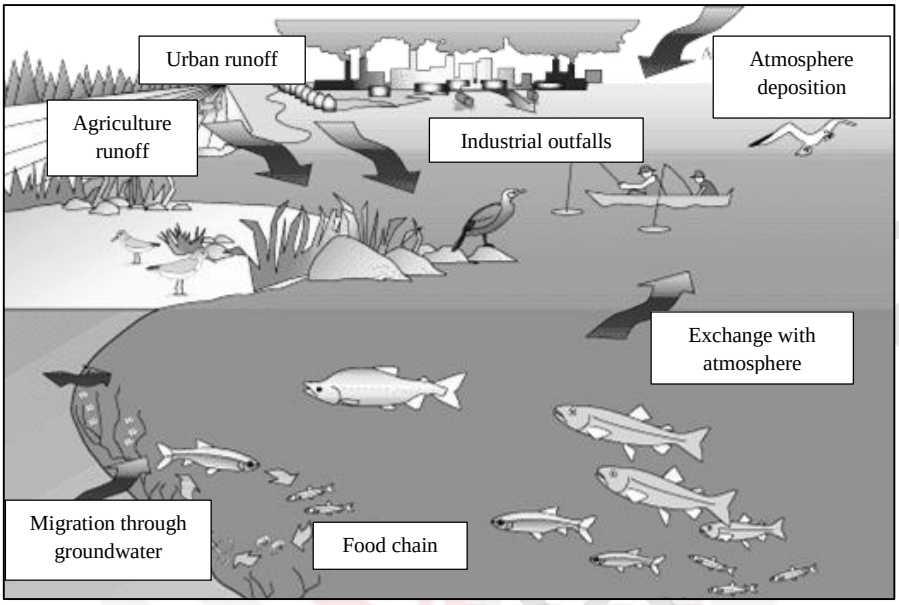

Figure 1.2. Sources of pollution in the coastal area
(Source: The Lake Huron Centre for Coastal Conservation, 1998-2015)

1.2 Problem Statement and Justification

Many studies have been conducted on the heavy metal pollution in the coastal area of Malaysia (Ismail et al., 1993; Yap et al., 2002, 2003; Shazali et al., 2006; Zulkifli et al., 2010). Although there have many documented of heavy metals status Malaysia waterways, but there had less study on trace elements record in seagrass area.

Merambong shoal seagrass area located near to the Port of Tanjung Pelepas and the developing area of Iskandar Malaysia. The increasing population number demands highly develop of Iskandar Malaysia through the urbanisation, industrial, domestic's activities and international trades. The insistence for much better facilities were on going construct to facilitate domestic life in the future. However, these activities lead to the trace elements discharge and sedimentation of seagrass bed contribute to the destroying of seagrass meadows which is important habitat and nursery for various marine organisms (Japar et al., 2006). Therefore, in ths study, the distribution of trace elements in the biota that exist in seagrass food web were determined using combination of stable isotope and trace elements analysis effective approach for evaluating the coastal ecosystem and human health.

1.3 Research Objectives

The mobility of heavy metals throughout the food web in the seagrass area can be completed by having following objectives of study:

I. Geochemical speciation and risk assessment of selected trace elements (Co, Ni, As, Cd, Pb) in surface sediments collected from seagrass area of Pulai River estuary;
II. To determine the concentration of trace elements in the marine organism effected from metal pollutants; and
III. Stable isotope ($\delta^{13}C$ and $\delta^{15}N$) analysis as a tool to construct the food web structure and trace elements pattern in seagrass area of Pulai River estuary.

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Article for Publication

Mukhtar, A., Zulkifli, S. Z., Mohamat-Yusuff, F., Ismail, A., & Miyazaki, N. (2015). Stable Isotope ($\delta^{13}C$ and $\delta^{15}N$) Analysis as a Tool to Quantify the Food Web Structure in Seagrass Area of Pulai River estuary. *Malayan Nature Journal*. (Submitted)

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1. **Mukhtar, A.**, Zulkifli, S. Z., Mohamat-Yusuff, F., Ismail, A., & N. Miyazaki (2013). A Preliminary Determination of Food Web in a Small Mangrove of Pantai Remis (Selangor, Malaysia) Using Stable Isotope Analysis. The Regional Fundamental Scienc Congress 2013, Faculty of Veterinar, Universiti Putra Malaysia.
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