

MASTER'S THESIS

Toxicological assessments of PAHs, OCPs and heavy metals in sediments at Mai Po and Deep Bay, Hong Kong

Kwok, Chun Kit

Date of Award:
2008

[Link to publication](#)

General rights

Copyright and intellectual property rights for the publications made accessible in HKBU Scholars are retained by the authors and/or other copyright owners. In addition to the restrictions prescribed by the Copyright Ordinance of Hong Kong, all users and readers must also observe the following terms of use:

- Users may download and print one copy of any publication from HKBU Scholars for the purpose of private study or research
- Users cannot further distribute the material or use it for any profit-making activity or commercial gain
- To share publications in HKBU Scholars with others, users are welcome to freely distribute the permanent URL assigned to the publication

**Toxicological Assessments of PAHs, OCPs and Heavy
Metals in Sediments at Mai Po and Deep Bay,
Hong Kong**

KWOK Chun Kit

A thesis submitted in partial fulfillment of the requirements
for the degree of
Master of Philosophy

Principal Supervisor: Prof. WONG Ming Hung

Hong Kong Baptist University

May 2008

Abstract

The main objective of this study was to investigate heavy metal (As, Cd, Cr, Cu, Pb and Zn), polyaromatic hydrocarbon (PAH) and organochlorine pesticide (OCP) contaminations in sediments, fish (crucian carp [*Carassius carassius*], grey mullet [*Mugil cephalus*], tenpounder [*Elops saurus*], Indo-pacific tarpon [*Megalops cyprinoids*], tilapia [*Oreochromis mossambicus*], snakehead [*Channa maculata*] and mud carp [*Cirrhinus molitorella*]), shrimps (*Metapenaeus ensis*) and eggs of ardeids (such as little egrets [*Egretta garzetta*]) of Mai Po Ramsar site as well as in sediments of the nearby Shenzhen River (SZR) and Kam Tin River (KTR). Sediments of 66 locations at Mai Po Ramsar site and the rivers were collected between 2003 and 2006. Concentrations of heavy metals, PAHs and OCPs in sediments were found to decrease in the order of SZR > KTR > mangrove and mudflat > seaward side of gei wais > landward side of gei wais, suggesting discharges from SZR and KTR were major sources of the pollutants in Mai Po. Sediments of Mai Po were also subjected to a more severe heavy metal contamination over the past 10 years when compared to a previous study conducted in 1997. Bioaccumulations of As, Cd, PAHs and OCPs were detected in fish and shrimps of Mai Po, as reflected by the high bioaccumulation factors (BAFs) (about 130 and 700% for As and Cd respectively) and biota-sediment

accumulation factors (BSAFs) (3.83-45.8 for OCPs; 1.7-6.3 for PAHs). Among different fish species, tilapia was subjected to a higher degree of bioaccumulation of the pollutants and this is probably related to the sediment-feeding nature of the species. Significant ($p < 0.01$) linear regressions were found between pollutants in different biota samples, including OCPs in eggs of ardeids and prey fish as well as Cr, Ni, Pb and Zn in bird eggs and eggshells. Estimated concentrations of OCPs, Cr, Ni, Pb and Zn in ardeids eggs of Mai Po were within ranges reported in Hong Kong, China and Pakistan. Results of toxicity tests (Microtox® solid-phase test, *Daphnia* mortality test, algal growth inhibition test and ryegrass seed germination/root elongation test) revealed differential toxicities among sediments collected in different locations of Mai Po, with the highest toxicity found in mudflat sediments while sediments of gei wai 24g the lowest. Different biomarker studies were also used to evaluate toxicity of heavy metal and organic pollutions in Mai Po. Hepatic metallothionein (MT) was induced in tilapia fed with artificial fish feed that included 30% (w/w) of dried sediment from the mudflat, suggesting induction of MT could be responsive to heavy metals in the diets. Glutathione (GSH), 7-ethoxyresorufin-*O*-deethylase (EROD), and MTT assays were also used to evaluate organic contaminations in Mai Po sediments. GSH, in particular, was found to be closely related to concentrations of total PAHs and OCPs in sediments and the

corresponding regressions can hopefully be used to predict PAH and OCP contaminations in sediments of Mai Po.

Table of contents

Declaration	i
Abstract	ii
Acknowledgements	v
Table of Contents	vi
List of Tables	xiii
List of Figures	xv
List of Appendices	xvii
CHAPTER 1 INTRODUCTION	1
1.1 Background information and ecological values of Mai Po Ramsar Site.....	1
1.1.1 Physical characteristics of Mai Po Ramsar site.....	1
1.1.2 Ecological significances of Mai Po Ramsar site.....	2
1.2 Review of the pollution status of Mai Po Ramsar Site over the past years.....	7
1.2.1 Background information about heavy metals.....	7
1.2.1.1 Heavy metal contamination in sediments of Mai Po.....	8
1.2.1.2 Heavy metal contamination in biota of Mai Po.....	9
1.2.2 Background information about Persistent Organic Pollutants (POPs).....	14
1.2.2.1 POPs in sediment of Mai Po.....	22

1.2.2.2	POPs in biota of Mai Po.....	23
1.3	Previous toxicological and biomarker studies concerning Mai Po Ramsar site....	24
1.4	Objectives of the present study.....	26

CHAPTER 2 CONTAMINATION OF POLYCYCLIC AROMATIC HYDROCARBONS (PAHs) AND ORGANOCHLORINE PESTICIDES (OCPs) IN SEDIMENTS AND BIOTA AROUND MAI PO RAMSAR SITE.....27

2.1	Introduction.....	27
2.1.1	PAHs and OCPs contamination in Mai Po and Deep Bay.....	27
2.1.2	Biomagnifications of PAHs and OCPs in fish and waterbirds.....	31
2.1.3	Objectives.....	33
2.2	Materials and methods.....	34
2.2.1	Sample collection.....	34
2.2.1.1	Mai Po.....	34
2.2.1.2	Jiangsu Province.....	35
2.2.2	Sample pretreatment.....	35
2.2.3	Soxhlet extraction and cleanup.....	37
2.2.4	Gas chromatography and mass spectrometry (GS/MS).....	39
2.2.5	Statistical analysis and construction of contour maps.....	40

2.3	Results.....	41
2.3.1	Spatial distribution of PAHs and OCPs in Mai Po sediments.....	41
2.3.2	PAHs source analysis.....	44
2.3.3	PAHs and OCPs in fish and shrimps of Mai Po.....	46
2.3.4	BSAFs for PAHs and OCPs in fish and shrimps of Mai Po.....	47
2.3.5	BSAFs for PAHs and OCPs in ardeids eggs of Jiangsu Province.....	48
2.4	Discussion.....	49
2.4.1	PAH and OCP contaminations in sediments and biota of Mai Po.....	49
2.4.1.1	Spatial distribution of PAHs and OCPs in sediments.....	49
2.4.1.2	Sources of PAHs in sediments.....	59
2.4.1.3	PAHs and OCPs in biota samples.....	60
2.4.2	BSAFs for PAHs and OCPs in biota of Mai Po.....	64
2.4.3	Extrapolation of OCP contamination in eggs of ardeids from prey fish.....	66
2.5	Conclusion.....	69
CHAPTER 3 CONTAMINATION OF HEAVY METALS IN SEDIMENTS AND BIOTA AROUND MAI PO RAMSAR SITE.....		71
3.1	Introduction.....	71
3.1.1	Heavy metal contamination around Mai Po Ramsar site.....	71

3.1.2	Bioaccumulations of metals in fish and birds.....	74
3.1.3	Objectives.....	77
3.2	Materials and methods.....	78
3.2.1	Sample collection.....	78
3.2.1.1	Mai Po.....	78
3.2.1.2	Jiangsu Province.....	79
3.2.2	Sample pretreatment.....	79
3.2.3	Acid digestion and metal quantifications.....	81
3.2.4	Statistical analysis and construction of contour maps.....	82
3.3	Results.....	83
3.3.1	Spatial distribution of heavy metals in sediments around Mai Po.....	83
3.3.2	Heavy metals in muscle and viscera of fish from Mai Po.....	84
3.3.3	Heavy metals in eggs, eggshells and feathers of ardeids from Mai Po and Jiangsu Province, China.....	86
3.4	Discussion.....	92
3.4.1	Contamination status of heavy metals in sediments of Mai Po.....	92
3.4.1.1	Sources of heavy metals.....	92
3.4.1.2	Heavy metal contamination in sediments of Mai Po in the past 10 years.....	94

3.4.1.3 Comparison with other studies and international guidelines.....	96
3.4.2 Comparison of BAFs for heavy metals in fish.....	98
3.4.3 Relationships between heavy metals in eggs and eggshells of bird.....	102
3.5 Conclusion.....	107

CHAPTER 4 TOXICOLOGICAL ASSESSMENTS OF SEDIMENTS COLLECTED AROUND MAI PO RAMSAR SITE USING BIOMARKERS AND DIFFERENT TROPHIC ORGANISMS.....109

4.1 Introduction.....	109
4.2 Materials and methods.....	113
4.2.1 Toxicity tests.....	113
4.2.1.1 Elutriate preparation.....	113
4.2.1.2 Microtox ® test.....	113
4.2.1.3 Algal growth inhibition test.....	114
4.2.1.4 <i>Daphnia</i> mortality test.....	115
4.2.1.5 Ryegrass seed germination/root elongation test.....	116
4.2.2 Metallothionein bioassay.....	117
4.2.2.1 Collection of tilapia and preparation of fish feed.....	117
4.2.2.2 Experimental setup and MT determination.....	118

4.2.3	GSH, EROD and MTT bioassay.....	120
4.2.3.1	Extraction of PAHs, OCPs and changing of solvent (n-hexane to DMSO).....	120
4.2.3.2	Cell culture.....	120
4.2.3.3	GSH extraction and determination.....	121
4.2.3.4	EROD determination.....	122
4.2.3.5	MTT assay.....	122
4.2.4	Calculation of effective concentrations and statistical analysis.....	123
4.3	Results.....	123
4.3.1	EC ₅₀ and EC ₁₀ of sediments and elutriates of Mai Po.....	123
4.3.2	Metallothionein concentrations in livers of tilapia.....	124
4.3.3	GSH, EROD and MTT responses in rat hepatoma cells and duck embryo fibroblast.....	125
4.3.4	Relationships between GSH responses and concentrations of PAH and OCP in sediments.....	126
4.4	Discussion.....	126
4.4.1	Toxicity of sediments of Mai Po Ramsar site.....	126
4.4.2	Use of metallothionein as a potential biomarker for heavy metal pollution in sediments.....	137

4.4.3 Use of GSH, EROD and MTT assays to evaluate PAH and OCP contaminations in sediments.....	141
4.5 Conclusion.....	146
CHAPTER 5 GENERAL CONCLUSION AND RECOMMENDATIONS.....	148
5.1 Heavy metal, PAH and OCP contaminations in sediments and fish of Mai Po Ramsar site.....	148
5.2 Toxicity of Mai Po sediments.....	151
5.3 Use of metallothionein, glutathione, EROD and MTT assays for evaluating heavy metal and organic pollution in sediments of Mai Po.....	154
5.4 Evaluation of heavy metals and OCPs in bird eggs of Mi Po.....	159
5.5 Prospects of the present study and recommendations.....	162
List of References.....	164
Appendices.....	188
Curriculum Vitae.....	193