การสะสมของสารฆ่าแมลงกลุ่มออร์กาโนคลอรีนในห่วงโซ่อาหารของปลา ณ คลอง 7 พื้นที่เกษตรกรรมรังสิต จังหวัดปทุมธานี

นางสาว จุฑาสิริ โรหิตรัตนะ

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# ACCUMULATION OF ORGANOCHLORINE INSECTICIDE RESIDUES IN FOOD CHAIN OF FISH AT KHLONG 7, RANGSIT AGRICULTURAL AREA, PATHUM THANI PROVINCE

Miss Juthasiri Rohitrattana

# สถาบนวิทยบริการ

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JUTHASIRI ROHITRATTANA : ACCUMULATION OF ORGANOCHLORINE INSECTICIDE RESIDUES IN FOOD CHAIN OF FISH AT KHLONG 7, RANGSIT AGRICULTURAL AREA, PATHUM THANI PROVINCE. THESIS ADVISOR : ASST. PROF. KUMTHORN THIRAKHUPT, PH.D.,

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Fifty species of fish were collected during June 2004 to May 2005 at Khlong 7, Rangsit, Pathum Thani Province. Forty-two species were analyzed for accumulation and biomagnification of organochlorine (OC) insecticide residues. The compounds were  $\Sigma$ BHC ( $\alpha$ -,  $\beta$ -,  $\gamma$ - and  $\delta$ -BHC),  $\Sigma$ Heptachlor (heptachlor and heptachlor epoxide),  $\Sigma$ Endrin (endrin and endrin aldehyde), [DDT (p,p'-DDE, p,p'-DDD and p,p'-DDT), [Endosulfan (endosulfan I, endosulfan II and endosulfan sulfate), aldrin, dieldrin and methoxychlor. The major compounds detected in the fillets were DDT (<0.04-48.26 ppb) and ∑Endosulfan (0.44-49.18 ppb), followed by ∑Heptachlor (<0.02-26.51 ppb), ∑BHC (<0.05-20.76 ppb), [Endrin (<0.02-14.73 ppb), Dieldrin (<0.03-13.22 ppb), Methoxychlor (<0.02-13.18 ppb) and Aldrin (<0.02-9.88 ppb), respectively. DDT and Endosulfan concentrations in fillets increased with fish total body length. Biomagnification factor (BMF) values were 1.61-2.27 for ∑DDT and 4.19-8.80 for Endosulfan from primary consumers, herbivorous and detritivorous fish to the top predatory fish. Based on the Thai and FAO/WHO maximum residue limits (MRLs), all OC residue concentrations were below the levels that have been suggested to cause the adverse effects on human and wildlife.

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สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย

### CONTENT

ABSTRACT IN THAIiv
ABSTRACT IN ENGLISHv
ACKNOWLEDGEMENTSvi
CONTENTS
LIST OF FIGURESix
LIST OF TABLESxi
CHAPTER I INTRODUCTION
1.1 Objectives
1.2 Scope of the study
1.3 Benefits of this study
CHAPTER II LITERATURE REVIEW
2.1 Insecticides4
2.2 Bioconcentration, bioaccumulation and biomagnification7
2.3 Rangsit agricultural area8
2.4 Related studies
CHAPTER III METHODOLOGY11
3.1 Chemical reagents
3.2 Instruments and equipments
3.3 Sampling12
3.4 Species richness
3.5 Food chain investigation14
3.6 OCs analysis14
3.7 Accumulation of OC residues
3.8 Biomagnification of OC residues
3.9 Risk evaluation
CHAPTER IV RESULTS AND DISCUSSIONS
4.1 Species richness of fish
4.2 Food relationship investigation24

4.3 OCs analysis	
4.4 Calibration curve	
4.5 Limit of detection (LOD) and limit of quantitation (LOQ)	
4.6 Quality control	31
4.7 Accumulation of OC residues in fish	33
4.8 Biomagnification in fish food chain	49
4.9 Risk evaluation	60
CHAPTER V CONCLUSIONS AND RECOMMENDATIONS	63
REFERENCES	64
BIOGRAPHY	69
APPENDIX	70



# LIST OF FIGURES

# Figure

# Pages

3.1 Sampling sites at Khlong 7, Rangsit, Pathum Thani Province13
4.1 The hypothetical food relationship in fish community of Khlong 7, Rangsit,
Pathum Thani Province25
4.2 The hypothetical fish food web of Khlong 7, Rangsit, Pathum Thani Province26
4.3 The chromatogram of 17 mixed organochlorine insecticide standards in
hexane 100 ng/L27
4.4 Mean $\pm$ SE of $\sum$ BHC, $\sum$ Heptachlor, $\sum$ Endosulfan and $\sum$ DDT residue
concentrations in some herbivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 2005
4.5 Mean $\pm$ SE of $\sum$ Endrin, Aldrin, Dieldrin and Methoxychlor residue
concentrations in some herbivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 2005
4.6 Mean $\pm$ SE of $\sum$ BHC, $\sum$ Heptachlor, $\sum$ Endosulfan and $\sum$ DDT residue
concentrations in some omnivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 2005
4.7 Mean $\pm$ SE of $\sum$ Endrin, Aldrin, Dieldrin and Methoxychlor residue
concentrations in some omnivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 2005
4.8 Mean $\pm$ SE of $\sum$ BHC, $\sum$ Heptachlor, $\sum$ Endosulfan and $\sum$ DDT residue
concentrations in some carnivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 200540
4.9 Mean $\pm$ SE of $\sum$ Endrin, Aldrin, Dieldrin and Methoxychlor residue
concentrations in some carnivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 200541
4.10 Mean $\pm$ SE of $\sum$ BHC, $\sum$ Heptachlor, $\sum$ Endosulfan and $\sum$ DDT residue
concentrations in some detritivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 200542

4.11 Mean $\pm$ SE of $\sum$ Endrin, Aldrin, Dieldrin and Methoxychlor residue
concentrations in some detritivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 200542
4.12 Comparison of OC residue concentrations ( $\pm$ SE) in fish and water at
Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 200545
4.13 The relationship of $\sum$ DDT concentration and body weight of <i>P. gonionotus</i> ,
N. notopterus and P. protozysron at Khlong 7, Rangsit, Pathum Thani Province
during June 2004 to May 200547
4.14 The relationship of $\sum$ Endosulfan concentration and body weight of <i>P. gonionotus</i> ,
N. notopterus and P. protozysron Khlong 7, Rangsit, Pathum Thani Province
during June 2004 to May 200548
4.15 Illustration of the biomagnification of $\sum$ DDT through some fish food chains at
Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 200552
4.16 Illustration of the biomagnification of $\sum$ Endosulfan through some fish food
chains at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to
May 2005
4.17 The biomagnification of $\sum$ DDT in hypothetical fish food web of Khlong 7,
Rangsit, Pathum Thani Province during June 2004 to May 200557
4.18 The biomagnification of $\sum$ Endosulfan in hypothetical fish food web of Khlong 7,
Rangsit, Pathum Thani Province during June 2004 to May 200558
4.19 The overall biomagnification factor (BMF) of $\sum$ DDT and $\sum$ Endosulfan
in some fish food chain at Khlong 7, Rangsit, Pathum Thani Province during
June 2004 to May 2005

# 

## LIST OF TABLES

#### Table

# Pages

3.1 The gas chromatographic conditions using DB-35 MS column for the
study of mixed 17 standard organochlorine insecticide solution15
4.1 Species Richness of Fish in Khlong 7, Rangsit, Pathum Thani
Province during June 2004 to May 200521
4.2 The correlation coefficient $(R^2)$ and linear regression equation of 17
mixed organochlorine insecticide standards with the ranged of
1-100 ng/L (ppb)29
4.3 The limit of detection (LOD) and limit of quantitation (LOQ) of
organochlorine insecticides standard solution
4.4 The recovery percentage and %RSD of 17 mixed organochlorine
insecticides spiked in fish matrix
4.5 The individual OC residue concentrations ( $\pm$ SE) in fish during June 2004
to May 2005 at Khlong 7, Rangsit, Pathum Thani Province44
4.6 Biomagnification factors (BMF) of $\sum$ DDT in fish food chains during
June 2004 to May 2005 at Khlong 7, Rangsit, Pathum Thani Province50
4.7 Biomagnification factors (BMF) of $\sum$ Endosulfan in fish food chains
during June 2004 to May 2005 at Khlong 7, Rangsit, Pathum Thani
Province
4.8 OC residue concentrations in herbivorous and carnivorous fish at
Khlong 7, Rangsit, Pathum Thani Province during June 2004 to
May 2005 compared with MRLs61
4.9 OC residue concentrations in omnivorous and detritivorous fish at
Khlong 7, Rangsit, Pathum Thani Province during June 2004 to
May 2005 compared with MRLs62
A-1 Physical properties of organochlorine insecticides71
A-2 OC concentrations (±SE) found in herbivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 200572

A-3 OC concentrations (±SE) found in carnivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 200572
A-4 OC concentrations (±SE) found in omnivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 200573
A-5 OC concentrations (±SE) found in detritivorous fish at Khlong 7, Rangsit,
Pathum Thani Province during June 2004 to May 200573



# สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย

#### **CHAPTER I**

#### **INTRODUCTION**

Organochlorine insecticides (OCs) have been wildly used since the World War II because they are inexpensive and effective insecticide. Nevertheless, most OCs are the persistent organic pollutants (POPs) which generally have low water solubility, do not degrade easily in the environment, bioaccumulate in food chains, and have been linked to adverse health effects in both humans and wildlife (Abel, 1996; Gunn and Sterens, 1976; Hond, 2003; Moriarty, 1999; Newman, 2001).

Although most of organochlorine insecticides were banned, they have been found in the environment including organisms. Studies of pesticide residues in food conducted between 1982-1985 by the Food and Drug Administration and the Department of Medical Science found residues in 52% of 663 samples, including DDT (39%) and dieldrin (15%). A 1993 survey by the National Environment Board indicated no improvement. Pesticide residues were found in all soil samples, 86% of water samples, 32% of fruits, 25% of vegetables and 17% of field crops (Chumraskul, 1999; Sakulthientrong, 2001; Thapinta and Hudak, 2000).

Aquatic organisms are contaminated by OCs ingestion and dermal contact. These contaminants can accumulate in the adipose tissues due to the lipophilic characteristics and their resistance to breakdown. Organisms occupying high trophic levels are often exposed to high concentrations of such chemicals as a result of biomagnification (Klaassen, 2001).

Khlong 7 is located in Nong Sua district, Pathum Thani Province. It is one of 14 sub-canals that have been used to irrigate Rangsit agricultural areas for more than 100 years. Therefore, this area is undoubtedly one of the most contaminated spots of pesticides in the central plain of Thailand.

Fish is the major protein source for human. It can intake the contaminants by ingestion and dermal contact and these contaminants can accumulate in the adipose tissues. For example, a carnivorous fish can obtain the residue by feeding small invertebrates and small fish whereas an herbivorous fish can obtain the residue by feeding by feeding aquatic plants and algae (Braunbeck et al., 1998).

The species richness of fish was reported each month. The concentrations of 17 OCs in selected fish were determined which are  $\alpha$ -,  $\gamma$ -,  $\beta$ - and  $\delta$ -BHC, heptachlor, aldrin, heptachlor epoxide, endosulfan I, endosulfan II, 4,4'-DDE, -DDD and -DDT, dieldrin, endrin, endrin aldehyde, endosulfan sulfate and methoxychlor.

The study of feeding behavior and stomach content were used to draw the fish food chains and also fish food web. This information was used to estimate the biomagnification in fish community. Evaluating the patterns, levels, trends and effects of OCs in high trophic level consumers may therefore contribute to understand of both the contamination of aquatic ecosystems and the risks posed to the health of humans and wildlife.

#### 1.1 Objectives

- 1.1.1 To examine the species richness of fish and their food.
- 1.1.2 To determine the accumulation of organochlorine insecticide residues in each fish species.
- 1.1.3 To determine the biomagnification of organochlorine insecticides in the food chain.
- 1.1.4 To evaluate the risk of fish consuming from Khlong 7, Rangsit agricultural area, Pathum Thani Province.

#### 1.2 Scopes of the study

- 1.2.1 Fish sampling was performed monthly for one year from June 2004 to May 2005. All samples were collected along Khlong 7 canal in Nong Sua district, Pathum Thani Province.
- 1.2.2 Dominant species in each month are the species which are frequently found and have enough weight for extraction procedure.
- 1.2.3 The study focuses on 17 organochlorine insecticides, which are α-, γ-, βand δ-BHC, heptachlor, aldrin, heptachlor epoxide, endosulfan I, endosulfan II, 4,4'-DDE, -DDD and -DDT, dieldrin, endrin, endrin aldehyde, endosulfan sulfate and methoxychlor.

#### 1.3 Benefits of this study

The species richness and the dominant species of fish at Khlong 7, Rangsit agricultural area, Pathum Thani Province was known. Their feeding behavior were illustrated the fish food chains which related to the biomagnification of OCs. The concentrations of 17 OCs in fish at Khlong 7 were known. The level of OCs concentration in fish trophic levels can be compared with MRL standard for evaluating the risk in both human and wildlife.

สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย

#### **CHAPTER II**

#### LITERATURE REVIEW

#### **2.1 Insecticides**

The early insecticides were divided into 3 main groups. The first were formulated from salts of metals, including calcium arsenate, copper sulfate, lead arsenate, and sulfur (Klassen et al., 2001). These chemicals were almost completely displaced by synthetic organic insecticides after sulfur had been exempted. The second group was tar oils, which is effective in coating and killing the eggs and over wintering larvae of many insects. The third group was toxic extracts of plants. Pyrethrum is the best known of these substances. These extracts affect on the rapid knock down action and cause paralysis in insects (Pimsman, 1997). The major categories of insecticides are organochlorines, organophosphates, carbamates and pyrethroids.

#### 2.1.1 Organochlorine Insecticide:

Organochlorine insecticides are synthetic organic insecticides. They were discovered before World War II. These insecticide groups consist of carbon, hydrogen and chlorine. Organochlorine insecticides are non-polar molecule and lipid soluble; therefore, they are hardly degradable and highly stable in the environment (Pimsman, 1997). Organochlorine insecticides can be divided into 3 groups as follows:

#### 2.1.1.1 DDT and DDT Analogues

Othmar Zeidler, German scientist, discovered DDT in 1873. Its insecticide property was studied by Switchzerland scientist, Paul Muller, in 1942. Since then it was wildly used in agricultural area and household to increase the productivity yields and for the protection of malaria. From the properties of DDT, it easily accumulates in soil, sediment and organism. As a result of slowly degradation and persistence in the environment, DDT was banned for agricultural usage in 1983. The compounds in this group are TDE, DDD, methoxychlor, perthane, dicofol and chlorobenzilate. Only dicofol and chlorobenzilate are still used as acaricide.

#### 2.1.1.2 Hexachlorocyclohexane

Hexachlorocyclohexane was discovered by French and English scientists in 1940. BHC (benzenehexachloride) has 5 isomers by its toxicity. Gamma isomer, known as lindane, has the highest toxicity. Lindane can dissolve in water, about 100 folds better than DDT, and easily evaporate. Hence, it is a severity respiratory and ingests toxicant.

#### 2.1.1.3 Cyclodiene

The diene groups are synthesized by Diels-Alder reaction, the six carbon ring synthesis. These reaction developed DDT in controlling Malaria disease. The organochlorine insecticides were widely used because their mammalian toxicity was generally low and their usage was broad. Some insecticides in this group are chlordane, aldrin, dieldrin, endrin and endosulfan. Aldrin can be metabolized to dieldrin in plants and animals and this form is very stable in the soil environment. Dieldrin and endrin were banned in 1988 and 1981 because they were highly toxic in mammals. Most OCs are easily dissolved in lipid (lipid soluble). This property leads to the accumulation in adipose tissue except the endrin rich can be metabolized and is rapidly excreted.

DDT is the most concern of OCs because of its hardly degradation and stability in the environment. DDT can be transformed to DDD (DDD 1, 1'- (2,2-Dichloroethyl-idene) bis (4-chlorobenzene)) and DDA, respectively. These metabolized forms are excreted by urine, wherever, DDE is storage in the adipose tissue.

OCs were heavily used after their introduction, this insecticide group was used as malaria repellent, termite and other insect control. But the disadvantages of using compounds that do not readily degrade under environmental conditions soon became apparent. OCs affect neural transmission. The change from  $Na^+-K^+$  transport causes prolong depolarization.

Chlordane is the first commercial insecticide in this group. It is an effective insecticide but is negative for plants. Chlordane is possibly carcinogen and persistent in environment. Therefore, its was banned in 1995.

Aldrin, dietdrin and endrin are high toxicity and long persistent. Aldrin can be transformed to dieldrin (epoxide form) in plants and animals. This metabolite form is high persistent in soil for several years. Aldrin and dieldrin are possibly carcinogen and accumulate in organisms. Both of them were banned in 1988. Endrin is an isomer of dieldrin. It is high toxicity to warm-blooded animals. Endrin was banned in 1981.

Endosulfan is an OC that contains oxygen and sulfur molecules. There are 2 isomers of endosulfan; alpha and beta form. It is an effective insecticide as well as aldrin but is lower persistent. Endosulfan can be transformed to endosulfan sulfate and is very stable in the soil. Endosulfan can be transformed to endosulfan sulfate and is persistent in soil. Because endosulfan has no severity effect to mammals and not accumulate in body, it is the only one organochlorine insecticide group that is still permitted to use in USA.

Another OC group is chlorinated camphene. The common name of this group is toxaphene. The biological properties are similar as lindane and not accumulated in adipose tissue. However, toxaphene was banned in 1983 because it could be carcinogen in tested animals.

#### 2.1.2 Organophosphate insecticide:

OPs generally include organic compounds that contain a phosphorus atom. These compounds are widely used to control agricultural pest and disease vectors. The best known is parathion and malathion. The toxicity of OPs are the ability to inhibit cholinesterase, which involves phosphorylation at the biochemical target site. Although generally of the higher mammalian toxicity than OCs, OPs do not present the same problem of environmental persistence and biological uptake.

#### 2.1.3 Carbamate:

Carbamates are ester of carbamic acid. The best known is carbaryl. Like OPs, carbamates are potent inhibitors of cholinesterase. Their mammalian toxicity is generally lower than that of organophosphates.

#### 2.1.4 Pyrethroid:

Pyrethroid insecticides have their origin in the naturally occurring insecticides in pyrethrum extracts. Pyrethrum has rapid knockdown action and causes paralysis in insects, but has low toxicity to mammals. Its use in agriculture is limited because it readily degrades in sunlight.

#### 2.2 Bioconcentration, Bioaccumulation and Biomagnification

Bioaccumulation means the accumulation of a contaminant into an organism or biological community, resulting either from direct uptake from water (i.e. by bioconcentration) or from ingestion (i.e. by biomagnification).

Bioconcentration means the accumulation of water-borne contaminant by a non-dietary route. In fish, the major route is via the gills. Bioconcentration results from the physio-chemical partitioning between ambient water and organism and the morphological and physiological features of the gills.

Biomagnification means the accumulation of a food-borne contaminant by oral uptake and digestion. Biomagnification of fish is often measured or calculated as the increase in contaminant concentration in excess of bioconcentration. It results from the physio-chemical partitioning between intestinal fluids and organism and the special food uptake processes in the gut system (Braunbeck et al, 1998).

#### 2.3 Rangsit Agricultural Area

Rangsit Plain is local in the northern a part of the Bangkok Metropolitan. It is believed that this area had originated from the freshwater swamp forest. From the accumulation of freshwater sediment and a bar formed at the river mouth, an inland lagoon had been formed. With the seasonal precipitation, the water would be more diluted. Because it was pure wilderness and uninhabited condition, the Great Plain would be formed.

Since 1890, in the Reign of King Rama the V, the development of the Great Plain had initiated. The main objective of this revolution was to increase the ricegrowing areas for more export of rice production. Because of the lack of sufficient budget, this project needed the wealthy nobleman and high officials to join in excavating the desired canals.

Until a team of interested investors comprising the Prince Saisanidwongse, Phra Phratibathirachaprasong, Phra Nanapithabhasi, Luang Sathonrachayudd and Mr. Joachim Grassi had signed a contract to excavate the canals throughout the Kingdom for the duration of 25 years. On 9 March 1890, the digging up of the new main canal had launched. Starting from the bank of Chao Phraya River at Ban Mai subdistrict, south of Koh Yai, Pathum Thani town to Nakhon Nayok River at Plakod Hua Kwai subdistrict, Nakhon Nayok town, which was itself 12 m. wide and 56 km. long. This canal was named "Rangsitprayulasakdi" by King Rama the V. The canal was the largest canal that traversing the Great Plain (Thung Luang) and the Plain itself had been known as the Rangsit Plain. After the completion of the excavation of Rangsit canal including branch canals and side canal, the great number of people move in until large communities of great wealths in rice production and local trade were formed as a result of enough surface water all year and convenient trade routes.

The study area is in Pathum Thani Province which is located in the lower plain of Pasak and Chao Phraya river basins, north of Bangkok. The main product of the province comes from the paddy field which covers 70 percent of the province's total land area. Other products come from mango, coconut and tangerine groves (Office of Agricultural Economics, 2002). Rangsit irrigation system, situated east of Pathumthani province, composes of 14 sub-canals (Khlong). Each sub-canal is 20 km long and Khlong 7 is at the center of the irrigation system. Rangsit Prayun Sak canal, situated along the southern end of each sub-canal, is the main canal that receives water from sub-canals and transfers water to the Chao Phraya River which flows towards Bangkok.

Khlong 7, located in Nong Sua district, Prathum Thani Province is one of 14 sub-canals that have been used to irrigate Rangsit agricultural areas for more than 100 years. This area is undoubtedly one of the most contaminated spots of pesticides in the central plain of Thailand.

#### **2.4 Related Studies**

Yenchum and Sam-ang Si (2004) found the abnormality of hepatocyte in *Puntius gonionotus* and *Parambassis siamensis* from collected fish at Khlong 7, Rangsit, Pathum Thani Province. They inferred that the histopathological changes might come from the polluted water in this area.

Kumblad et al. (2001) studied on the distribution of DDT residue in fish from Songkhla Lake, Thailand. They reported that the mean of  $\sum$ DDT concentrations at different locations in the analyzed fish species (*Scatophagus argus, Protosus canius, Channa strita* and *Zonichthys nigrofasciata*) ranged from 33 to 170 ng/g lipid weight (0.086-7.7 ng/g fresh weight). They also found that the high temperature and solar radiation in the lake could result in a dilution effect, when DDT is distributed in a large amount of organic matter, followed by high biological degradation of the substance.

Chumraskul et al. (1995) reported the pesticide residue levels in fresh water fish in central agricultural areas. They found OC residues in 96.4% of the total sample. BHC, aldrin, heptachlor & epoxide, dicofol, dieldrin, endrin, endosulfan and DDT & derivatives were found between 0.001-0.08 ppm (mg/kg). Most are DDT &derivatives, followed by dieldrin whereas endrin, aldrin and heptachlor&epoxide were found less than 50% of the total sample.

Punwiriyapong and Tayapath (1998) studied the impact of some OC insecticides to mothers' and newborn babies's blood. Their result showed that the concentrate of 3 OCs;  $\alpha$ -BHC, p,p'-DDE and diedrin, which transferred from mothers to their babies were reduced 2.3-5.6 fold from mothers.

Punwiriyapong and Harutaitanasan (2001) investigated impact of OCs to farmer's blood. They found 8 OCs in their subjects; p,p'-DDE, -DDT, -TDE, o,p'-DDT, heptachlor-epoxide,  $\alpha$ -endosulfan,  $\beta$ -endosulfan and endosulfan sulfate, in 0.55-809.03 ppb. OC residues were found in 96% of subjects. P,p'-DDT, -TDE and o,p'-DDT were detected in 30%, 16% and 11% of subjects, respectively. They also reported that the amount of p,p'-DDE residue related to age, sex, income and number of OCs usage.

Stuetz et al., (2001) studied on OC residues in human milk of a Hmong hill tribe living in Northern Thailand. They found the estimated daily intakes of DDT, heptachlor and heptachlor epoxide by the infants exceeded up to 20 times the acceptable daily intakes as recommended by the FAO and WHO. This residue levels related with mother's experiences to OCs usage as a vector control agent and agricultural purposes.

Favari et al. (2002) studied on the biomagnification of pesticides at Ignacio Ramirez, Mexico. They found that OC and OP insecticides were bioconcentrated 2- to 10-fold from water to algae, 10- to 25-fold in zooplankton and 8- to 140-fold in fish.

Stefanelli et al. (2004) studied on the estimation of organochlorine pesticides through edible fish from the Italian Adriatic Sea during 1997. They found only p, p'-DDE and p, p'-DDD at level up to 25 ng/g (ppb) wet weight. The estimated daily intake was calculated for the general Italian population, and it was significantly lower than the pertinent acceptable daily intake (ADI).

#### **CHAPTER III**

#### METHODOLOGY

#### **3.1 Chemical reagents**

#### 3.1.1 Solvents

3.1.1.1 Hexane (PR)

3.1.1.2 Acetone (PR & AR)

3.1.1.3 Petroleum Ether (PR)

3.1.1.4 Diethyl Ether (PR)

#### **3.1.2 Chemicals**

3.1.2.1 Ottawa Sand

3.1.2.2 Granular Anhydrous Sodium Sulfate

3.1.2.3 Distilled Water

#### 3.1.3 Standard chemicals

EPA 8080 Pesticides Mix Catalog No. 47913, consisting of aldrin, alphabenzene hexachloride ( $\alpha$ -BHC), beta-benzene hexachloride ( $\beta$ -BHC), delta-bezene hexachloride ( $\delta$ -BHC), dieldrin, endosulfan I (alpha), endosulfan II (beta), endosulfan sulfate, endrin, endrin aldehyde, gamma-benzene hexachloride ( $\gamma$ -BHC), heptachlor ,heptachlor epoxide isomer B, methoxychlor, 4,4'-DDD, 4,4'-DDE and 4,4'-DDT from SUPLECO, were used.

#### **3.2 Instruments and equipment**

3.2.1 Aluminum foil and PE zip-lock for sample containers

3.2.2 Accelerated Solvent Extractor (ASE 100<sup>®</sup>) from Dionex

3.2.3 Gas Chromatography (GC) model 6890N with micro-Electron Capture Detectors ( $\mu$ -ECDs) equipped with Agilent 6783 A 8-sample tray autosampler from Agilent Technologies. The data system is an Agilent Chemstation G2070AA operated with Windows ME and an Agilent Kayak XA, 350 MHz Pentium II computer workstation.

3.2.4 Nitrogen evaporator (Turbo Vap II Concentration workstation, Zymark Corporation, Hopkinton, MA, USA)

3.2.5 Solid-Phase Extraction (SPE) with 500 mg Florisil<sup>®</sup>-PR Extract-Clean Columns (4.0 mL column size) from Alltech Associates, Inc.

3.2.6 Blender3.2.7 GC amber vial 2 mL3.2.8 Vial-glass- 2-mL capacity

3.2.9 Volumetric pipette

3.2.10 Glass beaker

All glassware used was washed with liquid soap and rinsed properly with distilled water, and then with pure acetone (AR). They were then baked in an oven at 200°C for 6-8 hrs.

#### **3.3 Sampling**

All samples were collected at Khlong 7 monthly from June 2004 to May 2005. Fish were caught by fish net. Samples were collected at 3 sampling sites; upstream, middle- and downstream. Number of fish found in each fish species including the weight and length were recorded. Fish fillets were wrapped with aluminum foil, placed in HDPE container for support. Samples were stored in the freezer at -34°C until analysis.



**Figure 3.1** Sampling sites at Khlong 7, Rangsit, Pathum Thani Province. U=Upper stream, M=Middle stream and L=Lower stream.

#### **3.4 Species richness**

All collected fish were identified to species. The nomenclature and systematic arrangement of fish followed that of Vidthayanon et al. The fish occurrences were compiled into species richness and the number of individuals.

#### 3.5 Food chains investigation

The fish's trophic niche; detritivorous, herbivorous, omnivorous or carnivorous, was determined from its stomach content along with the observation from its feeding behavior. Collected data were compared with other literature reviews. Finally, the aquatic fish food web in Khlong 7, Rangsit, Pathum Thani Province was drawn.

#### **3.6 OCs analysis**

#### 3.6.1 GC condition

OCs analysis was adapted from AOAC Method 983.21. The GC conditions were shown in table 3.1.

Under GC condition, the mixed OCs standard were injected and confirmed by single OCs standard. Then, the retention times of each OCs were presented and used for identification of OCs in the samples.

3.6.2 Calibration curve

The OC standard mixture was prepared for 5 concentration levels; 1, 5, 10, 50 and 100 ppb, then injected to GC under the condition shown in table 3.1. The calibration curve of 17 OCs was plotted from 5 concentration points that used for quantitation of OCs in the samples. The recalibrations were done every batch of sample (about 20 samples per batch).

GC Parameters	GC Conditions
Analytical column	DB-35MS, 35% phenyl methyl siloxane,
	$30\ m\ x\ 320\ mm,\ 0.25\ \mu m$ film thickness
Temperature program	100°C to 280°C at 12°C/min and held for
	10 min
Injection mode	Splitless mode with a 0.75 min vent delay
1 hand	
Injection temperature	260°C
Flow rate of carrier gas (He)	2 mL/min
232/2/21	
Flow rate of nitrogen gas (N <sub>2</sub> )	60 mL/min
35.1891×91×91	
Detector	micro-electron capture detector (µ-ECD)
Detector temperature	300°C

 Table 3.1 The gas chromatographic conditions using DB-35MS column for the study

 of mixed 17 standard organochlorine insecticide solutions

3.6.3 Limit of detection (LOD) and limit of quantitation (LOQ)

The limit of detection (LOD) and limit of quantitation (LOQ) are defined as the peak height of analyte in standard solution that signaled significantly different from the peak height of noise equal 3 and 10 times of LOD and LOQ, respectively.

#### 3.6.4 Quality control (SOP for AOAC method 983.21)

3.6.4.1 Blanks

To confirm the extraction and analysis were not contaminated with the interferences, the set of blanks must be done. The blank sets included solvent blank, system blank and fortified sample blank. These blanks were done every batchs of samples (about 20 samples per batch). The blanks must be free of contaminants or the concentration of a contaminated method analytes is at least.

3.6.4.2 Replications

The replicate samples were done for evaluate the repeatability. Triple replicate samples were checked to be sure the measurement is remaining stable. Then, %RSD was calculated from the below equation;

%RSD

SD Mean

3.6.4.3 Fortified sample recovery

Fortified sample were done every batch of samples (about 20 samples per batch) to ensure that the analyze is being properly extracted. The fillet (10 g) was spiked with mixed OC standard 1 ppm for 10  $\mu$ L. The recovery percentage of fortified samples was calculated from below equation and must be between 60-140%.

Recovery percentage =  $\frac{\text{amount of recovered}}{\text{amount of added}} \times 100$ 

3.6.5 Sample preparation (Zhuang, et al., 2004)

Fish fillets were grounded in room temperature and homogenized by the mixer. Mixed fillets were extracted with hexane : acetone (3 : 1) using accerrated solvent extractor (ASE) with. The operating conditions were as follows: pressure, 1500 psi; heating time, 5 min; flush volume, 70%; purge time, 90 s; and number of static cycles, 2. The extracts were cleaned up by SPE florisil column eluted with 6%, 15% and 50% diethyl ether in petroleum ether, respectively. After that the cleaned up samples were concentrated by evaporator for final volume 2 mL. The samples were ready for analysis or stored at -34 °C not longer than 6 months.

3.6.6 GC analysis

Extracted samples were analysed for the organochlorine residues by gas chromatography with micro-electron capture detector (GC/ $\mu$ ECD). The OC concentrations were calculated from the area under the peak at predefined retention time. The OC concentrations in samples were calculated from calibration curved using linear regression equation as followed equation;

y = mx + C

Concentration in sample =  $\underline{CAa}$ 

Where C = Concentration in standard solution, ppb (µg/g)

VB

A = Area under the peak of sample

B = Area under the peak of standard solution

a = Final volume of sample (ml)

V = Initiate weight of sample for extraction (g)

#### 3.7 Accumulation of OC residues

The dominant species in each month were used to determine the concentration of OCs residues. The accumulation of OCs in each species was compared by ANOVA at 95 confidential percent. The relationships between OCs concentrations and fish body length were tested by linear regression.

#### 3.8 Biomagnification of OC residues

OCs residues magnified through fish food web were observed from the increasing of OCs concentrations along the fish trophic levels. The biomagnification factor (BMF) was defined by predator versus prey ratio.

#### 3.8 Risk evaluation

The maximum residue limits (MRL) were used to evaluate the potential health risk compared with OC residue concentrations in fillets.



#### **CHAPTER IV**

#### **RESULTS AND DISCUSSIONS**

#### 4.1 Species Richness of fish

A total of 4,883 fish belonging to 50 species in 19 families was caught during the survey. They were classified by their foraging niches into 4 groups; herbivore, omnivore, carnivore and detritivore. The number of species of herbivorous, omnivorous, carnivorous and detritivorous fish were 10, 20, 14 and 6, respectively. The results of the fish caught were summarized in table 4.1.

The examples for herbivores are *Puntius gonionotus*, *Probarbus labeaminor*, *Cyclocheilichthys armatus*, *Dangila spilopleura*, etc. Detritivores are *Mystus mysticetus*, *Heterobargrus bocourti*, *Mystus singaringan*, etc. Omnivores are *Puntioplites proctozysron*, *Trichogaster trichopterus*, *Trichogaster microlepis*, *Oreochromis niloticus*, *Puntius brevis*, etc. Carnivores are *Anabas testudineus*, *Notopterus notopterus*, *Pristolepis fasciatus*, etc. Top carnivores are *Channa striatus* and *Oxyeleotris marmorata*.

Common species found throughout the year were Cyprinids and Gouramies. Some are economic species such as common silver barb (*Puntius gonionotus*), gouramies (*Trichogaster microlepis*), striped snake-head fish (*Channa striatus*), lesser spinyed fish (*Macrognathusn siamensis*), featherback fish (*Notopterus notopterus*) and climbbling perch (*Anabus testudineus*).

All fish species of Khlong 7 were found in the Chao Praya basin (Vidthayanon et al., 1997) and 22 fish species were also found in Bung Boraped (Thirakhupt and Phienlumplert, 2005). Hence, the observed diversity at Khlong 7 represented a significant relationship of the Chao Praya fish fauna. Some species; *Kryptopterus apogon, Pangasianodon hypophthalmus* and *Chitala ornata* were reported to occur in

the Chao Praya river but were not found in this study. This is probably due to the insecticide contamination that may change the water quality and affect to the susceptible species.



# สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย

No.	Scientific name		2004								2005				Foraging niche
		Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	1	
	Cyprinidae		1321					1.41	80			5			0.0000000
1	Puntius gonionotus (Bleeker)	61	37	21	-	33	17	10	19	6	5	-	51	260	Herbivore
2	Puntius altus (Gunther)	-	-	-	-	-	-	-	-	1	-	1	-	2	Herbivore
3	Puntius brevis (Bleeker)	7	-	2	-	1	-	1	-	-	1	1	-	13	Omnivore
4	Puntius schwanenfeldi (Bleeker)	7	-	3		7	3	8	17	-	2	6	4	57	Omnivore
5	Puntioplites proctozysron (Bleeker)	4	-	2	3	1	-	9	-	-	3	7	5	34	Omnivore
6	Dangila spilopleura Smith	3	-	-	-	-	1	6	-	- 11	-	1	2	13	Herbivore
7	Hypsibarbus wetmorei (Smith)	-	-	-	-	(-)	1	-	-	-	_	-	_	1	Herbivore
8	Osteochilus hasselti (Valenciennes)	-	-	-	-	-	-	-	1	-	-	-	-	1	Herbivore
9	Probarbus labeamajor (Robert)	166	12	9	-	2	-	5	-	-	3	-	-	197	Herbivore
10	Probarbus labeaminor (Robert)	-	-	-	-	15	1	-	-	-	-	-	12	28	Herbivore
11	Henicorhynchus siamensis (Beaufort)	2	-	-	-	-	-	2	-	-	-	1		3	Omnivore
12	Cyclocheilichthys armatus (Valenciennes)	6	-	10		2	-	4	3	-	-	-	1	26	Herbivore
13	Cyclocheilichthys apogon (Valenciennes)	-	-	-	1	2	-	-	-	-	-	-	-	3	Herbivore
14	Parachela siamensis	-0	1	-	-	-	-	-	-	-	2	-	-	1	Omnivore
15	Oxygaster anomalura Van Hasselt	-	-	-	-	-	-	-	-	22	12	-	3	15	Omnivore
16	Rasbora argyrotaenia Smith	-	12	-	-	6	1	. 1	-	-	3	-	27	50	Omnivore
17	Rasbora borapetensis Smith	10	-	2	-	18	84	118	37	76	12	-	47	404	Omnivore
18	Rasbora tornieri Ahl.	-	-	-	-	-	-	-	20	16	-	14	-	50	Omnivore
19	Rasbora sp.1	' -	1	1	-9	-	4	2	3	2	1	-	-	14	Omnivore
20	Esomus metallicus Ahl.	2	-	2	-	62	130	123	108	130	50	23	21	651	Omnivore
21	Oryzias minutillus Smith	0.70	-	-	-	14	76	6	1	-	37	4	36	170	Herbivore
22	Paralaubuca harmandi Sauvage	-	-	3	-	-		3			-	-	-	6	Omnivore
39	Belontiidae	6			16						188			3	Dentrore
23	Trichogaster trichopterus (Pallus)	17	20	4	1	32	13	25	12	10	2	3	9	148	Omnivore
24	Trichogaster microlepis (Gunther)	5	18	15	2	10	6	20	12	4	9	-	15	116	Omnivore
25	Trichopsis pumila (Anold)	3	81	1	-	22	19	22	61	88	40	4	7	348	Canivore

Table 4.1 Species Richness of Fish in Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

Table 4.1 Species Richness of Fish in Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005 (Continue).

No.	Scientific name	2004						2005			Total	Foraging niche			
		Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May		0 0
26	Trichopsis vittatus (Cuv. & Val.)	20	39	1	-	35	79	147	86	205	54	5	4	675	Canivore
27	Xenentodon cancila (Ham.)	-	-	-	-	-	2	3	11	9	3	5	2	35	Canivore
	Anabantidae														
28	Anabas testudineus Bloch	4	-	-	-	6	-	13	2	-	-	-	-	25	Canivore
	Eleotridae														
29	Oxyeleotris marmorata (Bleeker)	-	2	1	-	-	-	1	1	-	2	-	2	9	Canivore
	Toxotidae			//											
30	Toxotes chatareus (Ham Buch.)	-	-	-	-	(-	-	-	-	-	1	-	1	2	Canivore
	Pristolepidae					13 A.A.									- Dentity vete
31	Pristolepis fasciatus (Bleeker)	1	-	-	1		1	1	1	-	3	-	-	8	Canivore
	Channidae		- <u>/</u>			alala									
32	Channa striatus (Bloch)	-	-	-	-	-	1	2	2	-	-	1	1	7	Canivore
33	Channa micropeltes (Cuvier)	-	-	-	-	1	-	-	-	-	-	-	-	1	Canivore
48	Ambassidea			A	23	11.5/15	11.5.1							1	Omayore
34	Parambassis wolffii (Bleeker)	-	-	-	-	-	-	-	-	-	2	18	22	42	Canivore
35	Parambassis siamensis	-	-	4	-	21	23	1	19	27	125	164	94	478	Canivore
50	Syngnathidae		2			1								1	Debityone
36	Microphis boaja (Bleeker)	2	-	-	-	3	1	2	2	-	-	-	2	12	Canivore
	Clupeidae														
37	Clupeichthys aesarnensis Wongratana	' -	21	0	-0	6	10	6	9	17	149	361	76	649	Canivore
	Aplocheilidae	6	10	9.10		19/1	510	UT.	20	5					
38	Aplocheilus panchax (Ham.)	1	-		0.0	-	3	21	1	0_	1	6	-	12	Omnivore
	Mastacembelidae					5		0			9				
39	Mastacembelus armatus (Lacepede)	-0	-	-	-	-	-	2	1/	-	<b>N</b> -3	-	1	3	Detritivore
40	Macrognathus siamensis (Bloch)	1	-	-	2.01	1	-	1	-	-	-	-	2	5	Detritivore

Table 4.1 Species Richness	of Fish in Khlong 7, Ra	ngsit, Pathum Thani I	Province during June 26	004 to May 2005	(Continue).
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No.	Scientific name		2004								2005				Foraging niche
		Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May		
	Hermirhamphidae												S		
41	Zenarchopterus ectuntio (Ham.)	-	7	-	-	79	82	38	6	15	22	8	X	257	Omnivore
	Notopteridae							11 - 11							
42	Notopterus notopterus (Pallas)	2	6	-	3	-	-	5	-	-	-	-	6	22	Canivore
	Siluridae				8111										
43	Micronema bleekeri (Gunther)	-	-	-	-	-	-	1	-	-	8°- 7	-		1	Canivore
	Bagridae				6								1		
44	Mystus mysticetus (Cuv. & Val.)	- S	2	-	-	-	-	-	-	-	-	-	1	3	Detritivore
45	Heterobagrus bocourti Bleeker	-	3	-	-	-	-	-	-	2	-	-	-	3	Detritivore
46	Mystus singaringan	-	12	-	-	-	-	-	-	-		-	3	15	Detritivore
	Cichlidae					a lak			1						
47	Oreochromis niloticus (Linn.)	2	-	1	-	-	-	-	-	-	6 - 6	-		3	Omnivore
	Pangasiidae								100		3.		2.3		
48	Pangasius pleurotaenia (Sauvage)	-	-	-		0.57	-	-	-	1	-	-	-	1	Omnivore
49	Pangasius macronema Bleeker	- 0	-	-	-	1	-	3	-	-		-	- 3	4	Omnivore
	Synbranchidae										121-12				
50	Monopterus albus (zuiew)	-	-	-		1	-	1	-	-		-	- 2	1	Detritivore
	Total	8						1						4879	



#### 4.2 Food relationship investigation

The fish food chains in figure 4.1 were drawn based on the observation of feeding habits in the aquarium, the study on fish stomach contents and the information from literatures. Most food chains had four trophic levels, starting from herbivorous and detritivorous fish to the top carnivores.

Figure 4.1 shows only the food relationship of some common fish species. The head of the arrow demonstrates the predator. For example, *P. gonionotus* was consumed by *P.protozysron*. Then, *P.protozysron* was consumed by *P.fasciatus*. Lastly, *P.fasciatus* was consumed by *C.striatus*, the top predator.

Food chains in figure 4.1 were combined into the food web to demonstrate the hypothetical food relationship in fish community of Khlong 7. The fish food web was shown in figure 4.2.

The food relationship of the fish at Khlong 7 can be classified into 4 trophic levels. The lowest or the first trophic level, consisting of the producers (such as aquatic plants and algae) and detritus was not shown in the figure. Herbivorous fish were in the second trophic level because they consumed only aquatic plants and algae. Detritivorous fish were the carcass feeders; therefore, they were also at the second trophic level. Omnivorous and some carnivorous fish were stated in the third trophic level. The omnivorous fish fed on both plants and aquatic animals while carnivorous fish fed only on aquatic animals. The top predators were stated at the highest trophic level. In this study, it was found that *Channa striatus* and *Oxyeleotris marmorata* were important predators hunting on other fish at the lower trophic levels.


**Figure 4.1** The hypothetical food relationship in fish community of Khlong 7, Rangsit, Pathum Thani Province.



**Figure 4.2** The hypothetical fish food web of Khlong 7, Rangsit, Pathum Thani Province.

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26

Following GC condition by using micro-ECD as a detector in chapter 3, the mixture of 17 organochlorine insecticide standard solution, spiked sample and sample solution were injected and analyzed.

By using DB-35MS, The retention time of 17 mixed organochlorine insecticides standard were consequently separated by their retention time which were  $\alpha$ -BHC,  $\gamma$ -BHC,  $\beta$ -BHC, Heptachlor,  $\delta$ -BHC, Aldrin, Heptachlor epoxide, Endosulfan I, 4,4'-DDE, Dieldrin, Endrin, 4,4'-DDD, Endosulfan II, 4,4'-DDT, Endrin aldehyde, Endosulfan sulfate and Methoxychlor, respectively. The demonstration of 17 mixed standard organochlorine insecticides chromatogram (100 ng/L in hexane) was shown in figure 4.3.

In this study, 17 sorts of organochlorine insecticide were arranged to 8 groups based on their metabolite which were Aldrin,  $\sum$ BHC ( $\alpha$ -,  $\gamma$ -,  $\beta$ - and  $\delta$ -BHC),  $\sum$ DDT (4,4'-DDD, -DDE and –DDT), Dieldrin,  $\sum$ Endosulfan (endosulfan I, endosulfan II and endosulfan sulfate),  $\sum$ Endrin (endrin and endrin aldehyde),  $\sum$ Heptachlor (heptachlor and heptachlor epoxide), and Methoxychlor.



**Figure 4.3** The chromatogram of 17 mixed organochlorine insecticide standards in hexane 100 ng/L.

#### 4.4 Calibration curve

The calibration curves of five concentration levels of 17 mixed organochlorine insecticides standard solutions were constructed. Five concentrations between 1-100 ppb and were plotted by peak area versus concentration. All 17 mixed organochlorine insecticides exhibited linear relationship between peak area and concentration as shown in Table 4.2. The correlation coefficient ( $R^2$ ) were ranged between 0.994 - 0.998.  $R^2$  of all 17 mixed organochlorine insecticides were greater than 0.99 which were acceptable for GC quantitatative analysis (Jarupaiboon, 2003).

In addition, the slope value from linear regression equation is used for express the sensitivity of GC analysis. In this study, the sensitivity of 17 mixed organochlorine insecticides standard were prioritized that Endosulfan sulfate < 4,4'-DDT < Endrin aldehyde <  $\beta$ -BHC < Endosulfan II < 4,4'-DDD < Endrin < Methoxychlor < Endosulfan I < Dieldrin <  $\delta$ -BHC < 4,4'-DDE < Heptachlor epoxide < Heptachlor < Aldrin <  $\gamma$ -BHC <  $\alpha$ -BHC, repectively.

#### 4.5 Result of limit of detection (LOD) and limit of quantitation (LOQ)

Theoretically, the limit of detection (LOD) and limit of quantitation (LOQ) are defined as the peak height of analyte in standard solution that signaled significantly different from the peak height of noise equal three times of LOD and 10 times for LOQ of each compound. The result showed in table 4.3 that LOD of this study was 0.0007 - 0.0508 and LOQ was 0.0024 - 0.1695. Additionally, most analyzed fish samples were above LOD and LOQ, for this reason the analytes can be quantified. Nonetheless, when the concentrations of analytes were lower than LOD, this study described as below LOD.

Insecticides	Correlation	<b>Regression Formula:</b> y = mx+b x=amount (ng/L), y=area			
	Coefficient (R <sup>2</sup> )	Slope (m)	Intercept (b)		
α -BHC	0.998	5280	-72		
ү-BHC	0.996	4248	784		
β-BHC	0.998	1465	555		
Heptachlor	0.995	3840	5089		
δ-BHC	0.998	2501	-567		
Aldrin	0.997	3856	-780		
Heptachlor epoxide	0.995	2859	255		
Endosulfan I	0.994	2377	859		
4,4'-DDE	0.996	2594	-612		
Dieldrin	0.994	2439	299		
Endrin	0.995	1942	-265		
4,4'-DDD	0.997	1900	837		
Endosulfan II	0.997	1675	1026		
4,4'-DDT	0.997	1027	-518		
Endrin aldehyde	0.996	1248	2305		
Endosulfan sulfate	0.995	833	2203		
Methoxychlor	0.994 👝	2055	3335		

**Table 4.2** The correlation coefficient ( $R^2$ ) and linear regression equation of 17 mixed organochlorine insecticide standards with the ranged of 1-100 ng/L (ppb).

จุฬาลงกรณ์มหาวิทยาลัย

OC Incastisidas	LOD	LOQ		
OC Insecticides	(µg/L)	(µg/L)		
α -BHC	0.03	0.09		
۲ -BHC	0.05	0.2		
β-BHC	0.01	0.05		
Heptachlor	0.0007	0.002		
δ-BHC	0.05	0.1		
Aldrin	0.02	0.05		
Heptachlor epoxide	0.02	0.07		
Endosulfan I	0.05	0.2		
4,4'-DDE	0.04	0.1		
Dieldrin	0.03	0.1		
Endrin	0.02	0.07		
4,4'-DDD	0.003	0.009		
Endosulfan II	0.003	0.009		
4,4'-DDT	0.002	0.008		
Endrin aldehyde	0.002	0.007		
Endosulfan sulfate	0.002	0.007		
Methoxychlor	0.02	0.06		

**Table 4.3** The limit of detection (LOD) and limit of quantitation (LOQ) oforganochlorine insecticides standard solution.

# จุฬาลงกรณ์มหาวิทยาลัย

#### **4.6 Quality Control**

#### 4.6.1 Result of blank

System blank and solvent blank were performed each batch of sample, the result showed that system blank and solvent blank response in absence of interference.

Method blank was also carried through all the steps of sample preparation and analysis (in chapter 3) as if it were an actual sample. The result showed that there were no interferences occur during sample preparation and analysis procedure.

#### 4.6.2 Result of recovery and repeatability

Due to fish samples have a complex matrix, and matrix may play an importance role in the accuracy and precision of the measurement. Matrix (fish fillet) spiking were done by spike the triplicate of known concentration of 17 mixed organochlorine insecticides standard into the fish sample which had no present of 17 mixed organochlorine insecticides and then the samples were extracted by the method in chapter 3. The recovery percentage can be calculated by the equation below,

Recovery percentage = 
$$\frac{\text{amount of recovered}}{\text{amount of added}} \times 100$$

Usually the extraction efficiency, or matrix spiked recovery is required to be at least 70% (Kebbekus and Mitra, 1998) relate to Siriwong (2000) reported that the recovery percentage of organochlorine pesticide residues analyzed by using ECD as a detector should between 70 to 110. In this study, the result in table 4.4 showed that the means of recovery percentage of 17 mixed organochlorine insecticides were between 82.03 to 130.57. According to AOAC 983.21, all spiked matrix were in the acceptable range which were 60-140% because this study were done matrix spiked recoveries at the nanogram concentration level; spiked matrix concentration was very possible to loss the recovery yield at the low concentration level.

The repeatability showed by %RSD in table 4.4. The mean of %RSD was between 2.75 to 10.16. This %RSD was in the acceptable range determined by AOAC that referred to not exceed 21 at 10  $\mu$ g/L (AOAC, 1993). Therefore, the recovery percentages and repeatability of all analytes were accepted for this study.

Insecticides	Spiked Recovery	%RSD
	(%)	
α-BHC	130.57	2.75
γ-BHC	95.76	5.38
β-ВНС	127.53	2.96
Heptachlor	115.48	4.86
δ-BHC	123.80	4.16
Aldrin	119.36	5.83
Heptachlor epoxide	112.30	10.16
Endosulfan I	103.54	5.98
4,4'-DDE	95.24	7.59
Dieldrin	101.19	7.60
Endrin	107.20	6.73
4,4'-DDD	82.03	7.25
Endosulfan II	85.33	5.82
4,4'-DDT	109.38	4.99
Endrin aldehyde	85.97	4.18
Endosulfan sulfate	124.57	3.17
Methoxychlor	117.53	8.07

**Table 4.4** the recovery percentage of 17 mixed organochlorine insecticides spiked in fish matrix.

สถาบนวทยบรการ จุฬาลงกรณ์มหาวิทยาลัย

#### 4.7 Accumulation of OCs residues in fish

#### 4.7.1 OCs concentrations in fish

In this studied, the 17 OC compounds were described into 8 groups according to theirs metabolite forms.  $\sum$ DDT referred to 4,4'-DDD, 4,4'-DDE and 4,4'-DDT.  $\sum$ Endosulfan referred to endosulfan I, endosulfan II and endosulfan sulfate.  $\sum$ Heptachlor referred to heptachlor and heptachlor epoxide.  $\sum$ BHC were  $\alpha$ -BHC,  $\gamma$ -BHC,  $\beta$ -BHC and  $\delta$ -BHC.  $\sum$ Endrin were endrin and endrin aldehyde. Other groups included aldrin, dieldrin and methoxychlor.

The concentrations of organochlorine pesticide residues found in 42 fish species were  $\sum DDT > \sum Endosulfan > \sum Heptachlor > \sum BHC > \sum Endrin > Dieldrin > Methoxychlor > Aldrin, respectively. Additionally, there were statistically significant differences among detrivore, herbivore, omnivore and carnivore (Kruskal-Wallis One Way Analysis of Variance on Ranks, H = 10.232, df = 3, P-value =0.0017).$ 

In particular,  $\sum$ DDT and  $\sum$ Endosulfan residue concentrations were mostly found in every species.  $\sum$ DDT and  $\sum$ Endosulfan found among trophic levels were statistically different (Kruskal-Wallis One Way Analysis of Variance on Ranks,  $\sum$ DDT, H = 8.239, df = 3, P-value = 0.041, and  $\sum$ Endosulfan, H = 17.837, df = 3, Pvalue  $\leq$  0.001).

The OC residues in fish within the same foraging niche were different. This could be explained by the variation of lipid content and feeding behavior. Fish with higher lipid content could store the OC residues more than lower lipid content fish. Also, the feeding behavior involving with food consumed and preferred food could result the contaminant obtaining (Ruus et al., 1999).

Using One Way ANOVA (n=4, df=3, P<0.05), detritivorous species,  $\sum$ DDT residue concentrations were found in *Macrognathus siamensis* > *Mystus singaringan* > *Heterobagrus bocourti* > *Mystus mysticetus*, respectively whereas  $\sum$ Endosulfan

residue concentrations were found in *Macrognathus siamensis* > *Heterobagrus* bocourti > Mystus singaringan > Mystus mysticetus, respectively.

In herbivorous species,  $\sum$ DDT residue concentrations were found in Probarbus labeaminor > Puntius altus > Hypsibarbus wetmorei > Cyclocheilichthys armatus > Puntius gonionotus > Dangila spilopleura > Oryzias minutillus > Probarbus labeamajor, respectively while  $\sum$ Endosulfan residue concentrations were found in Puntius gonionotus > Puntius altus > Oryzias minutillus > Probarbus labeaminor > Dangila spilopleura > Cyclocheilichthys armatus > Probarbus labeamajor > Hypsibarbus wetmorei, respectively (One Way ANOVA; n=8, df=7, P<0.05).

In omnivorous species,  $\sum$ DDT residue concentrations were found in Trichogaster microlepis > Pangasius pleurotaenia > Oreochromis niloticus > Puntius brevis > Puntius schwanenfeldi > Trichogaster trichopterus > Puntioplites proctozysron > Rasbora sp.1 > Oxygaster anomalura > Paralaubuca harmandi >Esomus metallicus > Rasbora borapetensis> Henicorhynchus siamensis > Clupeichthys aesarnensis > Parachela siamensis > Rasbora tornieri > Rasbora argyrotaenia > Zenarchopterus ectuntio, respectively whereas  $\Sigma$ Endosulfan residue concentrations were found in Oreochromis niloticus > Trichogaster trichopterus > Rasbora sp.1 > Puntioplites proctozysron > Trichogaster microlepis > Oxygaster anomalura > Paralaubuca harmandi > Esomus metallicus > Puntius schwanenfeldi > Pangasius pleurotaenia > Puntius brevis > Rasbora tornieri > Zenarchopterus ectuntio > Rasbora borapetensis > Rasbora argyrotaenia > Parachela siamensis > Clupeichthys aesarnensis > Henicorhynchus siamensis, respectively (One Way ANOVA; n=18, df=17, P<0.05).

In carnivorous species,  $\sum$ DDT residue concentrations were found in *Channa* striatus > Oxyeleotris marmorata > Parambassis siamensis > Pristolepis fasciatus Parambassis wolffii > Micronema bleekeri > Toxotes chatareus > Anabas testudineus > Xenentodon cancila > Notopterus notopterus > Trichopsis pumila > Trichopsis vittatus, respectively whereas  $\sum$ Endosulfan residue concentrations were found in Oxyeleotris marmorata > Channa striatus > Parambassis siamensis > Pristolepis fasciatus > Anabas testudineus > Micronema bleekeri > Trichopsis vittatus > Parambassis wolffii > Notopterus notopterus > Xenentodon cancila > Toxotes chatareus > Trichopsis pumila, respectively (One Way ANOVA; n=12, df=11, P<0.05).

Figure 4.4-4.11 showed the concentration of organochlorine pesticide residues based on the consumption behavior which are detritivore, herbivore, omnivore, and carnivore.  $\sum$ DDT and  $\sum$ Endosulfan were dominantly represented in most of fish species. Carnivores, omnivores and herbivores exhibited more  $\sum$ DDT than others, whereas detritivores showed the high concentration of  $\sum$ Endosulfan. This could be explained by the metabolite compounds especially endosulfan sulfate that was very stable in the sediment and transferred to detritivores. Some detritivorous fish presented higher concentration of OCs than other groups. This could be explained by their bottom feeding behavior that might uptake the contaminants by both carcass and sediment feeding.

สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย



**Figure 4.4** Means  $\pm$  SE of  $\sum$ BHC,  $\sum$ Heptachlor,  $\sum$ Endosulfan and  $\sum$ DDT residue concentrations in some herbivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.





**Figure 4.5** Means  $\pm$  SE of  $\sum$ Endrin, Aldrin, Dieldrin and Methoxylchlor residue concentrations in some herbivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.



**Figure 4.6** Means  $\pm$  SE of  $\sum$ BHC,  $\sum$ Heptachlor,  $\sum$ Endosulfan and  $\sum$ DDT residue concentrations in some omnivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.



**Figure 4.7** Means  $\pm$  SE of  $\sum$ Endrin, Aldrin, Dieldrin and Methoxylchlor residue concentrations in some omnivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.



**Figure 4.8** Means  $\pm$  SE of  $\sum$ BHC,  $\sum$ Heptachlor,  $\sum$ Endosulfan and  $\sum$ DDT residue concentrations in some carnivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.



**Figure 4.9** Means  $\pm$  SE of  $\sum$ Endrin, Aldrin, Dieldrin and Methoxylchlor residue concentrations in some carnivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.



**Figure 4.10** Means  $\pm$  SE of  $\sum$ BHC,  $\sum$ Heptachlor,  $\sum$ Endosulfan and  $\sum$ DDT residue concentrations in some detritivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.



**Figure 4.11** Means  $\pm$  SE of  $\sum$ Endrin, Aldrin, Dieldrin and Methoxylchlor residue concentrations in some detritivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

The means of the highest concentration of  $\sum$ DDT,  $\sum$ Endosulfan,  $\sum$ Heptachlor,  $\sum$ BHC,  $\sum$ Endrin, Dieldrin, Aldrin, Methoxychlor were 48.26, 49.18, 26.51, 20.76, 14.73, 13.22, 9.88 and 13.18 ppb, respectively. These data related to Chumraskul (1995) study that they found OC residues in fresh water fish in central agricultural areas between 1-80 ppb and most were  $\sum$ DDT.

Table 4.5 showed the individual OC residue concentrations found in carnivorous fish during June 2004 to May 2005. The highest concentration level was 4,4'-DDT, followed by endosulfan sulfate, heptachlor, endosulfan I, endosulfan II, 4,4'-DDE,  $\delta$ -BHC, 4,4'-DDD, dieldrin,  $\beta$ -BHC, aldrin, heptachlor epoxide, endrin aldehyde, endrin,  $\gamma$ -BHC, methoxychlor and  $\alpha$ -BHC, respectively. The large amount of 4,4'-DDT could be explained by the greatly potential persistent due to its highly octanol-water coefficient value (log P<sub>ow</sub>=6.91). Even through DDT was banned for several years (since 1983) but it still circulates in the environment because of the heavily used for agricultural purposes in the past decade. Monthly means of each OC compound were statistically different (n=12, df=11, P<0.05), indicating that the concentration of each OC in fish at Khlong 7 was not steady. This could result from the monthly different in fish diversity and fish caught. Thus, the levels of OC residues found in fish throughout the year were dissimilar.

OC concentrations in common species frequently found during the year such as the common silver barb (*Puntius gonionotus*) were significantly different throughout the year, although there were some months that the OC residues were not different. For example, DDT, Endosulfan and Heptachlor were similar in June, July and August, 2004. For the other species, the snaked-head fish (*Channa striatus*) OC residues were significantly different throughout the year. This is probably because of the size difference of the fish caught.

Name	OC residue concentrations (ppb)												
	2004 2005						Mean± SE						
	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	
a BHC	0.0530	0.1430	0.2150	0.0000	0.0567	0.4160	0.4370	0.6730	0.3060	0.2790	0.3100	0.4000	$0.274 \pm \! 0.056$
g BHC	0.2740	0.1840	0.6380	0.9490	0.4170	0.9240	0.3560	0.2390	0.6260	0.0618	0.0972	0.4170	$0.432 \pm 0.085$
b BHC	0.8420	0.7100	2.1190	2.8400	0.6140	2.9200	2.0930	2.6540	0.6360	1.8020	1.1410	0.0582	$1.536 \pm 0.285$
Heptachlor	2.6700	2.6330	3.1510	2.1470	2.1320	6.2540	0.6390	5.8640	2.5700	4.9650	3.9090	1.2250	$3.180 \pm 0.506$
d BHC	0.4640	1.0120	2.0600	2.5640	1.1170	1.8470	1.3960	1.9790	1.1890	1.5220	1.3150	3.6680	$1.678 \pm 0.242$
Aldrin	0.5870	1.0490	1.3770	2.5340	1.1770	2.0420	1.1580	2.3020	0.9730	1.8470	0.7810	1.2730	$1.425 \pm 0.178$
Heptachlor Epoxide	0.3630	1.4820	1.1850	0.0000	0.1890	1.7920	0.8310	2.1350	1.4170	1.6640	1.5360	0.8740	1.12±0.195
Endosulfan I	1.1090	6.7600	2.1990	0.5120	2.3720	4.9340	3.1710	3.1470	2.0410	4.7740	4.2670	1.5920	3.073±0.525
4,4 DDE	1.4570	1.7390	1.7360	2.6350	3.2000	1.7980	1.7290	3.3690	1.6850	2.5620	1.6710	5.5740	$2.430 \pm 0.341$
Dieldrin	0.3620	1.3740	1.2500	0.5020	0.1410	2.7200	1.3630	2.2130	1.4800	2.5460	2.7050	2.0990	$1.563 \pm 0.262$
Endrin	0.6010	0.2010	0.5420	0.2670	0.1050	0.7300	0.5320	1.0850	0.7220	0.8720	0.5200	2.6340	0.734±0.191
4,4 DDD	0.8860	2.1880	1.5080	0.0000	1.7780	2.6210	1.4160	2.4840	1.4400	2.1090	2.0100	0.5260	$1.580 \pm 0.229$
Endosulfan II	2.1870	3.2430	1.5180	4.0880	2.6340	3.0600	3.3250	2.5660	1.5320	2.8380	2.3480	1.9320	$2.606 \pm 0.220$
4,4 DDT	6.5600	7.0180	8.5560	4.9730	3.1380	4.4210	6.2080	10.2850	2.9760	4.2710	2.8300	3.8410	5.423±0.067
Endrin Aldehyde	0.1770	0.4370	0.0516	0.2750	0.9870	0.9480	0.4010	0.2960	0.3530	0.0000	0.2450	6.6060	$0.898 \pm 0.026$
Endosulfan Sulfate	2.5040	2.3460	3.4200	2.8280	3.3170	5.4380	5.1050	4.5650	1.7750	4.8240	3.5100	1.1260	3.396±1.366
Methoxychlor	0.0000	0.0000	0.0000	0.0000	0.5050	0.0000	0.0560	0.0000	0.0000	0.0000	0.0000	4.1500	0.393±0.034
Total	21.0960	32.5190	31.5256	27.1140	23.8797	42.8650	30.2160	45.8560	21.7210	36.9368	29.1952	37.9952	

Table 4.5 The individual OC residue concentrations (±SE) in fish during June 2004 to May 2005 at Khlong 7, Rangsit, Pathum Thani Province.

สถาบันวิทยบริการ หาลงกรณ์มหาวิทยาลัย

44

#### 4.7.2 OCs concentrations in fish compared with water



**Figure 4.12** Comparison of OC residue concentrations (±SE) in fish compared with water at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

The means of OC residue concentrations during the sampling time at Khlong 7 were compared between concentrations found in water and concentrations found in fish. From figure 4.12, the OC concentrations found in fish were higher than OC concentrations found in water. The concentrations were increased more than 100 times from background water to fish. This could be inferred that OC concentrations in fish might accumulate from water in addition to ingestion. Furthermore, fish might intake OCs from their living habit from sediment as well as from water (Caldas et al.,1999).



#### 4.7.3 Relationship of body weight and OCs concentration

Three fish species were studied for the relationship between OCs concentrations and the total body length. *P. gonionotus*, *N. notopterus* and *P. protozysron* represented for herbivorous, carnivorous and omnivorous fish, respectively.  $\Sigma$ DDT and  $\Sigma$ Endosulfan were selected to demonstrate the relationship shown in figure 4.13-4.14. Analysis using linear regression showed that R<sup>2</sup> values for  $\Sigma$ DDT of *P. gonionotus*, *N. notopterus* and *P. protozysron* were 0.775 (n=12, P<0.001), 0.818 (n=18, P<0.001) and 0.387 (n=12, P=0.031), respectively and R<sup>2</sup> values for  $\Sigma$ Endosulfan were 0.559 (n=12, P=0.005), 0.703 (n=18, P<0.001) and 0.578 (n=12, P=0.004), respectively. The results were shown in Figure 4.4 and Figure 4.5, repectively.

Consistent with  $R^2$  and P value, the OCs concentrations were significantly related with fish body weight. This result agreed with Dissayawong (1979) and Tanabe (1994) who reported that OCs concentrations increased in relation to body weight and length. According to organochlorine compounds likely to accumulate in organic tissues due to their lipophilicity and persistence to degradation, OCs expected in view of random distribution of OCs storage in fish muscle in all sorts of organic tissues, not only in fat (Vives et al,2005). Furthermore, the increase of OC levels assumed to result from a higher contribution of fish in the diet of larger fish (Weber and Goerke, 2003).

สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย



Weight (g)

**Figure 4.13** The relationship of  $\sum$ DDT concentration and body weight of *P*. *gonionotus N. notopterus* and *P. protozysron* at Khlong 7, Rangsit, Pathum Thani Province.



#### Weight (g)

**Figure 4.14** The relationship of  $\sum$ Endosulfan concentration and body weight of *P*. *gonionotus N. notopterus* and *P. protozysron* at Khlong 7, Rangsit, Pathumthani Province.

#### **4.8** The biomagnification in fish food chain

The OCs concentrations in each fish species were used to determine the biomagnification in fish food chain. The biomagnification factors (BMF) were used to explain the increase of OCs concentrations along the trophic levels. The BMF was calculated from the following equation:

### $BMF = \underline{Concentration in Predator (ng/g)}$ Concentration in Prey (ng/g)

In this study,  $\sum$ DDT and  $\sum$ Endosulfan showed the highest concentrations. It is probably because they were heavily used in the past and they have the characteristics of slowly degraded and long-term persistent in the environment. Both of them have been most concerned as the insecticides that can transfer through the food chain and accumulate in the organisms. The BMF of  $\sum$ DDT was shown in table 4.6 and illustrated in figure 4.15 whereas the BMF of  $\sum$ Endosulfan was shown in table 4.7 and illustrated in figure 4.16.

The BMF values showed the increasing of OC concentrations along the food chains, indicating that the concentrations found in predators were higher than preys. The higher BMF values were resulted from the foraging behaviors. For example, the BMF values of *P. wolffii* versus *P. labeamajor* equal to 10.46, this demonstrated that carnivorous fish (*P. wolffii*) had more contaminant than herbivorous fish (*P. labeamajor*) via ingested fish and plankton whereas herbivorous fish ate mainly on aquatic plants that contained lower OC concentrations.

From figure 4.6 and figure 4.7, the number under the fish names referred to OC concentrations and the number under the arrows referred to BMF values. These figures showed the magnifying of OC concentrations through fish food chains including their BMF values of predators versus preys.

Predators / Preys	Concentration in [Predators] / [Preys] (ng/g) (ng/g)	BMF
Herbivorous feeder		
N. notopterus / P. gonionotus	4.75 / 3.28	1.45
A. testudieneus / P. gonionotus	5.71 / 3.28	1.74
H. siamensis / P. gonionotus	3.74 / 3.28	1.14
P. protozysron / P. gonionotus	9.66 / 3.28	2.95
R. sp.1 / P. gonionotus	7.05 / 3.28	2.15
N. notopterus / P. labeamajor	4.75 / 0.85	5.59
A. testudieneus / P. labeamajor	5.71 / 0.85	6.72
P. wolffii / P. labeamajor	8.89 / 0.85	10.46
O. anomalura / P. labeamajor	5.87 / 0.85	6.91
T. chatareus / P. labeamajor	6.68 / 0.85	7.86
P. protozysron / C. amatus	9.66 / 3.92	2.46
R. sp.1 / C. amatus	7.05 / 3.92	1.80
Detritivorous feeder		
C. striatus / M. mysticetus	48.26 / 10.09	4.78
O. marmorata / M. mysticetus	25.22 / 10.09	2.50
C. striatus / H. bocourti	48.26 / 14.30	3.37
Omnivorous feeder		
P. brevis / H. siamensis	14.18 / 3.74	3.79
O. niloticus / H. siamensis	15.41 / 3.74	4.12
P. fasciatus / P. protozysron	14.39 / 9.66	16 21.49
O. niloticus / R. sp.1	15.41 / 7.05	2.19
P. brevis / O. anomalura	14.18 / 5.87	2.42

**Table 4.6** Biomagnification factors (BMF) of ∑DDT in some fish food chains at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

Predators / Preys	Concentration in	BMF
	[Predators] / [Preys] (ng/g) (ng/g)	
Carnivorous feeder	(***********	
P. siamensis / N. notopterus	24.54 / 4.75	5.17
P. siamensis / A. testudieneus	24.54 / 5.71	4.30
P. fasciatus / P. wolffii	14.39 / 8.89	1.62
P. fasciatus / T. chatar <mark>eus</mark>	14.39 / 6.68	2.15
O. niloticus / A. testudieneus	15.41 / 5.71	2.70
O. marmorata / P. brevis	25.22 / 14.18	1.78
O. marmorata / O. niloticus	25.22 / 15.41	1.64
O. marmorata / P. fasciatus	25.22 / 14.39	1.75
O. marmorata / P. siamensis	25.22 / 24.54	1.03
C. striatus / O. niloticus	48.26 / 15.41	3.13
C. striatus / P. fasciatus	48.26 / 14.39	3.35
C. striatus / P. siamensis	48.26 / 24.54	1.97

**Table 4.6** Biomagnification factors (BMF) of ∑DDT in some fish food chains at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005 (continue).

สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย

P. gonionotus	2.95	P. protozysron	1.49	P.fasciatus	3.35	C.striatus
3.28		9.66		14.39		48.26
P. gonionotus	1.14	H. siamensis	3.79	P. brevis	1.78	O. marmorata
3.28		3.74		14.18		25.22
P. gonionotus	2.15	R. sp.1	2.19	O. niloticus	3.13	C.striatus
3.28		7.05		15.41		48.26
P. labeamajor	7.86	T.chatareus	2.15	P.fasciatus	3.35	C.striatus
0.85		6.68	2.12	14.39		48.26
P. labeamajor	6.91	O. anomalura	2.42	P. brevis	1.78	O. marmorata
0.85	5 50	5.87	5 17	14.18	1.07	25.22
P. labeamajor	J.J9	N. notopterus	J.17	P.siamensis	1.97	C.striatus
0.85	0.15	4.75	1.40	24.54	2.25	48.26
C. amatus	2.46	P. protozysron	1.49	P.fasciatus	3.35	C.striatus
3.92		9.66		14.39		48.26
C. amatus	1.80	R. sp.1	2.19	O. niloticus	3.13	C.striatus
3.92	1 45	7.05	5 17	15.41	1 07	48.26
P. gonionotus		N. notopterus	<u> </u>	P.siamensis	1.97	C.striatus
3.28		4.75		24.54		48.26
P. gonionotus	1.74	A. testudineus	4.30	P.siamensis	1.03	O. marmorata
3.28	10.46	5.71	1.62	24.54	1.75	25.22
P. labeamajor		P. wolffii		P.fasciatus		O. marmorata
0.85	(7)	8.89	2 70	14.39	3 13	25.22
P. labeamajor	<u>0.72</u> →	A. testudineus	2.70	O. niloticus	<b>→</b>	C.striatus
0.85	4 70	5.71		15.41		48.26
M. mysticetus	4.78	C.striatus				
10.09	2.50	48.26				
M. mysticetus	2.50	O. marmorata				
10.09	2 27	25.22				
H. bocourti	→ 	C.striatus				
14.30		48.26				

**Figure 4.15** Illustration of the biomagnification of  $\sum$ DDT through some fish food chains at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

Predators/Preys	Concentration in [Predators] / [Preys]	BMF
Harbiyaraya faadar	(ng/g) (ng/g)	
ner bivorous reeder	7 00 / 2 25	2.54
P. protozysron / P. gonionotus	1.99 / 3.25	2.56
R. sp.1 / P. gonionotus	8.49 / 3.25	2.61
T.chatareus / P. labeamajor	2.64 / 0.46	5.74
O. anomalura / P. labeamajor	7.06 / 0.46	15.35
N. notopterus / P. labeamajor	5.59 / 0.46	12.15
P. wolffii / C. amatus	6.17 / 2.23	2.77
P. protozysron / C. amatus	7.99 / 2.23	3.58
R. sp.1 / C. amatus	8.49 / 2.23	3.81
N. notopterus / P. labeaminor	5.59 / 1.72	3.25
A. testudineus / P. gonionotus	10.64 / 3.25	3.27
P. wolffii / P. labeaminor	6.17 / 1.72	3.59
	2/18/18/202	
Detritivorous feeder	6	
C.striatus / M. mysticetus	39.48 / 1.39	28.40
O. marmorata / M. mysticetus	40.12 / 1.39	28.86
Omniverous feeder	A	
	22.10.77.00	4.15
P.fasciatus / P. protozysron	33.1977.99	4.15
O. niloticus / R. sp.1	17.71 / 8.49	2.09
T. trichopterus / O. anomalura	10.04 / 7.06	<b>b C</b> 1.42
Carnivorous feeder		
P.fasciatus / T.chatareus	33.19 / 2.64	12.57
T. trichopterus / N. notopterus	10.04 / 5.59	1.80
O. niloticus / P. wolffii	17.71 / 6.17	2.87
O. niloticus / A. testudineus	17.71 / 10.64	1.66
P.fasciatus / P. wolffii	33.19 / 6.17	5.38

**Table 4.7** Biomagnification factors (BMF) of ∑Endosulfan in some fish food chains at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

**Table 4.7** Biomagnification factors (BMF) of ∑Endosulfan in some fish food chains at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005 (continue).

Predators/Preys	Concentration in [Predators] / [Preys] (ng/g) (ng/g)	BMF
Carnivorous feeder (continue)		
O. marmorata / O. niloticus	40.12 / 17.71	2.27
O. marmorata / P. fasciatus	40.12 / 33.19	1.21
O. marmorata / T. trichopterus	40.12 / 10.04	4.00
C. striatus / O. niloticus	39.48 / 17.71	2.23
C. striatus / P. fasciatus	39.48 / 33.19	1.19
C. striatus / T. trichopterus	39.48 / 10.04	3.93

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**Figure 4.16** The illustration of the biomagnification of ∑Endosulfan through some fish food chains at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

Figure 4.17-4.18 shown the biomagnification of  $\sum$ DDT and  $\sum$ Endosulfan. The number under the fish names referred to total concentration (ppb). The overall biomagnification of  $\sum$ DDT and  $\sum$ Endosulfan were summarized in figure 4.19.  $\sum$ DDT was magnified from primary consumers through higher consumers on average 1.61, 2.11 and 2.27 times, respectively whereas  $\sum$ Endosulfan was magnified 4.19, 6.73 and 8.80 times, respectively. OC concentrations were highest in the top predators such as the snake head fish (*Channa striatus*) and the sleepy goby (*Oxyeleotris marmorata*) due to their high trophic positions.

Most BMFs in this study were consistent with the food chain transfer of contaminants, with concentrations increasing from the lower trophic level to the higher trophic level. However, a few exceptions showed that organisms at the higher trophic level had lower OC concentrations than their preys. For examples, detritivorous fish; spotted spiny eel and *M. siamensis*. One explanation is that the predator may consume a small number of preys which contain high OC residues. The specific capacity of different species to degrade OC compounds is probably another main factor explaining the observed interspecific patterns in OC levels. Bernhoft et al. (1997) reported that the polar bear's (consume fish) high ability to metabolize many OCs could explain the lower concentration of these compounds in this animal compared with the white whale (consume plankton) from the same area. This could suggest that detritivorous fish may have slower ability to metabolize OCs than other fish groups. Moreover, the detritivores could get the contaminants from the sediment and from buried substances such as the carcass of the higher trophic level organisms.

## จุฬาลงกรณมหาวทยาลย



**Figure 4.17** The biomagnification of  $\sum$ DDT in hypothetical fish food web of Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.







**Figure 4.19** The overall biomagnification factor (BMF) of  $\sum$ DDT and  $\sum$ Endosulfan in some fish food chain at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

#### 4.9 Risk evaluation

The OC residue concentrations were compared with the maximum residue limits (MRLs) in both Thailand and international standard. Table 4.8-4.9 showed OC residue concentrations in herbivorous, carnivorous, omnivorous and detritivorous fish.

From table 4.8-4.9, the OC residue concentrations of all samples did not exceed the maximum residue limits notified by the Ministry of Public Health, Thailand and FAO/WHO Guidelines. Thus, the OC concentrations in fish at Khlong 7, Rangsit, Pathum Thani Province in particular  $\Sigma$ DDT,  $\Sigma$ Endosulfan,  $\Sigma$ Heptachlor,  $\Sigma$ BHC,  $\Sigma$ Endrin, Dieldrin, Aldrin and Methoxychlor were below levels that were suggested to cause adverse effects in human and wildlife.

However, there are some awareness of the insecticide residues in fish fillets of some species. It is recommended avoid to eating the bottom feeder fish because they contain high level of OCs than other species. Boiling, baking, grilling or streaming are better cooking methods to remove fat and residues away from foods.


Scientific name	OCs concentrations (ppm)									
	∑внс	∑Heptachlor	Endosulfan	<b>∑DDT</b>	DEndrin	Aldrin	Dieldrin	Methoxychlor		
Herbivores	1.	- Andreastan and an and	a la constante de la constante			1111710	Sector Sector			
Puntius gonionotus	0.003	0.003	0.003	0.003	0.001	0.000	0.000	0.000		
Puntius altus	0.002	0.001	0.003	0.007	0.007	0.000	0.000	0.000		
Dangila spilopleura	0.002	0.002	0.001	0.003	0.006	0.001	0.000	0.000		
Hypsibarbus wetmorei	0.001	0.004	0.000	0.005	0.000	0.000	0.000	0.000		
Probarbus labeamajor	0.002	0.000	0.000	0.000	0.000	0.000	0.000	0.000		
Probarbus labeaminor	0.001	0.009	0.001	0.009	0.004	0.000	0.000	0.001		
Cyclocheilichthys armatus	0.002	0.002	0.000	0.003	0.001	0.000	0.000	0.000		
Oryzias minutillus	0.005	0.006	0.002	0.002	0.003	0.000	0.001	0.000		
Carnivores		11			0.022					
Trichopsis pumila	0.001	0.002	0.002	0.005	0.000	0.001	0.000	0.000		
Trichopsis vittatus	0.003	0.003	0.007	0.003	0.001	0.000	0.001	0.000		
Xenentodon cancila	0.004	0.004	0.005	0.006	0.001	0.000	0.001	0.000		
Anabas testudineus	0.002	0.004	0.010	0.006	0.000	0.000	0.000	0.000		
Oxyeleotris marmorata	0.012	0.026	0.040	0.025	0.000	0.006	0.013	0.000		
Toxotes chatareus	0.000	0.002	0.003	0.007	0.000	0.000	0.000	0.000		
Pristolepis fasciatus	0.006	0.009	0.033	0.014	0.002	0.004	0.003	0.000		
Channa striatus	0.020	0.024	0.039	0.048	0.014	0.009	0.012	0.006		
Parambassis wolffii	0.005	0.005	0.006	0.009	0.006	0.007	0.003	0.002		
Parambassis siamensis	0.012	0.023	0.037	0.024	0.004	0.007	0.012	0.002		
Notopterus notopterus	0.001	0.001	0.006	0.005	0.000	0.000	0.000	0.000		
Micronema bleekeri	0.000	0.000	0.009	0.008	0.002	0.000	0.006	0.000		
MRL	0.5	0.3	0.3	5	0.3	0.1	0.3	5		
MRL <sup>#</sup>	N.D.	0.3	0.3	5	N.D.	0.3	0.3	5		

Table 4.8 OC residue concentrations in herbivorous and carnivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005 compared with MR

MRL<sup>\*</sup> referred to the Ministry of Public Health, Thailand MRL<sup>#</sup> referred to FAO/WHO, 1992

Scientific name	OCs concentrations (ppm)									
	∑внс	∑Heptachlor	∑Endosulfan	∑DDT	<b>D</b> Endrin	Aldrin	Dieldrin	Methoxychlor		
Omnivores					· · · · ·					
Puntius brevis	0.002	0.002	0.002	0.014	0.000	0.000	0.000	0.000		
Puntius schwanenfeldi	0.007	0.005	0.004	0.013	0.000	0.002	0.000	0.000		
Puntioplites proctozysron	0.003	0.001	0.008	0.010	0.002	0.000	0.000	0.000		
Henicorhynchus siamensis	0.000	0.002	0.000	0.004	0.000	0.000	0.000	0.000		
Parachela siamensis	0.003	0.000	0.000	0.003	0.000	0.000	0.000	0.000		
Oxygaster anomalura	0.003	0.003	0.007	0.006	0.000	0.000	0.000	0.000		
Rasbora argyrotaenia	0.000	0.000	0.001	0.000	0.002	0.001	0.000	0.000		
Rasbora borapetensis	0.003	0.003	0.001	0.004	0.002	0.001	0.000	0.000		
Rasbora tornieri	0.000	0.000	0.002	0.001	0.000	0.000	0.000	0.000		
Rasbora sp.1	0.009	0.002	0.009	0.007	0.000	0.000	0.002	0.000		
Esomus metallicus	0.002	0.002	0.005	0.005	0.000	0.000	0.000	0.000		
Paralaubuca harmandi	0.004	0.003	0.007	0.005	0.000	0.000	0.000	0.000		
Trichogaster trichopterus	0.003	0.004	0.010	0.011	0.006	0.001	0.000	0.000		
Trichogaster microlepis	0.004	0.004	0.007	0.024	0.005	0.002	0.000	0.000		
Clupeichthys aesarnensis	0.001	0.002	0.000	0.004	0.000	0.000	0.000	0.000		
Zenarchopterus ectuntio	0.002	0.002	0.002	0.000	0.000	0.000	0.000	0.000		
Oreochromis niloticus	0.008	0.006	0.018	0.015	0.004	0.002	0.000	0.000		
Pangasius pleurotaenia	0.000	0.003	0.003	0.016	0.003	0.000	0.000	0.000		
Detritivores		and the		6. · · · · · · · · · · · · · · · · · · ·	and the	1 E .	14 12			
Macrognathus siamensis	0.005	0.006	0.049	0.035	0.010	0.006	0.009	0.013		
Mystus mysticetus	0.001	0.001	0.010	0.010	0.001	0.000	0.000	0.000		
Heterobagrus bocourti	0.000	0.000	0.026	0.014	0.002	0.000	0.000	0.000		
Mystus singaringan	0.000	' 0.001	0.014	0.018	0.006	0.000	0.002	0.001		
MRL	0.5	0.3	0.3	0 0 0 5	0.3	0.1	0.3	5		
MRL <sup>#</sup>	N.D.	0.3	0.3	5	N.D.	0.3	0.3	5		

Table 4.9 OCs concentrations in omnivorous and detritivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005 compared with MRLs.

MRL<sup>\*</sup> referred to the Ministry of Public Health, Thailand MRL<sup>#</sup> referred to FAO/WHO, 1992

### **CHAPTER V**

#### CONCLUSIONS AND RECOMMENDATIONS

- 1. Fifty fish species in 19 families were found at Khlong 7, Rangsit, Pathum Thani Province during the sampling from June 2004 to May 2005. The dominant groups were Cyprinids and Guramies of which some were economic species.
- 2. Fish food chains of Khlong 7, Rangsit, Pathum Thani Province were drawn based on the fish foraging niches and then were combined into the fish food web.
- The predominant concentration of OC residues were ∑DDT and ∑Endosulfan, followed by ∑Heptachlor, ∑BHC, ∑Endrin, Dieldrin, Aldrin, Methoxychlor, respectively.
- 4. Due to the highest trophic positions, the top predators such as the snake-head fish (*Channa striatus*) and the sleepy goby (*Oxyeleotris marmorata*) were found with highest OC concentrations.
- The concentration of OC residues, ∑DDT and ∑Endosulfan, were magnified through the food chain. The increases were observed by the biomagnification factors (BMFs) between predators and preys.
- 6. According to the maximum residue limits (MRLs) in both Thailand and international standard, the OC concentrations in fish at Khlong 7, Rangsit, Pathumthani Province in particular ∑DDT, ∑Endosulfan, ∑Heptachlor, ∑BHC, ∑Endrin, Dieldrin, Aldrin and Methoxychlor were below levels that have been suggested to cause adverse effects in human and wildlife.
- 7. To complete the food chain and food web of Khlong 7 aquatic community, the concentration of OC residues in aquatic plants, aquatic invertebrates, sediment and water should be investigated. Then, the biomagnification through the complex food web would be demonstrated and the consumer risk could be predicted.
- 8. As a result of OC residues in fish, it can be used as the fundamental information for insecticide control regulation in permission of the insecticide usage in agricultural purposes.

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สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย

## **BIOGRAPHY**

Miss Juthasiri Rohitrattana was born on May 28, 1981 in Bangkok, Thailand. She attended Saipanya School and graduated in 1996. She received her Bachelor's Degree in General Science from Faculty of Science, Chulalongkorn University, Bangkok, Thailand in 2001. She pursued her Master Degree studies in International Postgraduate Programs in Environmental Management, Inter-Department of Environmental Management, Chulalongkorn University, Bangkok, Thailand in May 2003. She finished her Master Degree of Science in Environmental Management in October 2005.



สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย

# สถาบันวิทยบริการ จุฬาลงกรณ์มหาวิทยาลัย

APPENDIX

Compounds	Physical Properties									
	Log Pow	$\mathbf{S}_{\mathbf{w}}$	Vp	K <sub>H</sub>						
		( <b>mg/L</b> )	(mmHg)	(atm-m <sup>3</sup> /mole)						
a-BHC	3.80	2.00	-	1.22 x 10 <sup>-5</sup>						
b-BHC	3.78	0.24	-	4.40 x 10 <sup>-7</sup>						
d-BHC	4.14	10.00	3.52 x 10 <sup>-5</sup>	4.29 x 10 <sup>-7</sup>						
g-BHC	3.72	7.20	4.20 x 10 <sup>-5</sup>	5.14 x 10 <sup>-6</sup>						
BHC	4.26	8.00	7.83 x 10 <sup>-4</sup>	2.56 x 10 <sup>-4</sup>						
DDD	6.02	0.09	1.35 x 10 <sup>-6</sup>	6.60 x 10 <sup>-6</sup>						
DDE	6.51	0.04	-	4.16 x 10 <sup>-5</sup>						
DDT	6.91	0.01	$1.60 \ge 10^{-7}$	8.32 x 10 <sup>-6</sup>						
Endosulfan I	3.83	0.51	3.00 x 10 <sup>-6</sup>	7.09 x 10 <sup>-6</sup>						
Endosulfan II	3.83	0.45	6.00 x 10 <sup>-7</sup>	3.91 x 10 <sup>-7</sup>						
Endosulfan	3.66	0.48	-	3.25 x 10 <sup>-7</sup>						
Sulfate										
Endosulfan	3.83	0.33	1.73 x 10 <sup>-7</sup>	6.50 x 10 <sup>-5</sup>						
Endrin	5.20	0.25	3.00 x 10 <sup>-6</sup>	6.36 x 10 <sup>-6</sup>						
Endrin	4.80	0.02	2.00 x 10 <sup>-7</sup>	4.18 x 10 <sup>-6</sup>						
Aldehyde										
Aldrin	6.50	0.02	1.20 x 10 <sup>-4</sup>	4.40 x 10 <sup>-5</sup>						
Dieldrin	5.40	0.20	5.89 x 10 <sup>-6</sup>	1.00 x 10 <sup>-5</sup>						
Heptachlor	6.10	0.18	4.00 x 10 <sup>-4</sup>	2.94 x 10 <sup>-4</sup>						
Heptachlor	4.98	0.20	1.95 x 10 <sup>-5</sup>	2.10 x 10 <sup>-5</sup>						
Epoxide	0 1 1 1 10									
Methoxychlor	5.08	0.10	2.58 x 10 <sup>-6</sup>	2.03 x 10 <sup>-7</sup>						

 Table A-1 Physical properties of organochlorine insecticides.

Available from: http://chem.sis.nlm.nih.gov/chemidplus/

Scientific name	OCs concentrations (ppb)								
	∑BHC	∑Heptachlor	∑Endosulfan	∑DDT	∑Endrin	Aldrin	Dieldrin	Methoxychlor	
Puntius gonionotus	2.54±0.24	2.78±1.00	3.25±0.14	3.28±0.54	1.29±0.09	0.57±0.25	0.27±0.06	0.26±0.01	
Puntius altus	1.19±0.30	$1.18\pm0.88$	3.21±0.61	7.30±1.25	7.10±0.24	$0.83 \pm 0.04$	< 0.03	< 0.02	
Dangila spilopleura	2.12±0.15	2.21±0.81	1.37±0.08	2.95±0.48	5.72±2.86	1.36±0.29	0.32±0.04	< 0.02	
Hypsibarbus wetmorei	1.12±0.28	0.35±0.18	0.44±0.13	5.36±1.35	< 0.02	< 0.02	< 0.03	< 0.02	
Probarbus labeamajor	2.02±0.21	< 0.02	0.45±0.13	0.85±0.20	< 0.02	< 0.02	< 0.03	< 0.02	
Probarbus labeaminor	1.19±0.15	0.93±0.46	1.72±0.30	8.95±1.30	4.56±2.28	< 0.02	$0.56 \pm 0.04$	1.57±0.00	
Cyclocheilichthys armatus	2.26±0.22	1.58±0.78	0.67±0.11	3.91±1.05	0.13±0.09	$0.09\pm0.00$	< 0.03	0.09±0.00	
Oryzias minutillus	4.92±0.92	0.57±0.29	2.23±0.74	1.89±0.63	3.41±1.71	< 0.02	1.10±0.07	< 0.02	

Table A-2 OCs concentrations (±SE) found in herbivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

Table A-3 OCs concentrations (±SE) found in carnivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

Scientific name	OCs concentrations (ppb)								
	∑BHC	∑Heptachlor	∑Endosulfan	∑DDT	∑Endrin	Aldrin	Dieldrin	Methoxychlor	
Trichopsis pumila	$1.68\pm0.18$	2.42±1.21	1.79±0.30	4.54±0.43	< 0.02	1.11±0.31	< 0.03	< 0.02	
Trichopsis vittatus	2.78±0.29	3.02±1.19	6.61±0.14	3.03±0.29	1.23±0.14	0.78±0.41	1.16±0.56	0.12±0.08	
Xenentodon cancila	4.01±0.17	3.57±1.77	5.32±0.60	5.59±0.76	1.65±0.03	$0.22 \pm 0.00$	1.48±0.56	< 0.02	
Anabas testudineus	2.13±0.23	3.52±0.98	$10.64 \pm 1.04$	5.71±0.92	0.84±0.27	0.29±0.00	$0.80 \pm 0.00$	< 0.02	
Oxyeleotris marmorata	12.19±1.89	26.51±2.31	40.12±2.52	25.22±1.49	< 0.02	6.27±0.26	13.22±0.09	< 0.02	
Toxotes chatareus	< 0.05	2.44±1.22	2.64±0.32	6.68±1.03	0.96±0.48	$0.68 \pm 0.14$	0.74±0.39	< 0.02	
Pristolepis fasciatus	5.86±1.46	8.96±4.48	33.19±1.57	14.39±1.68	2.31±1.16	4.15±0.07	3.37±0.18	< 0.02	
Channa striatus	20.76±2.46	24.13±1.24	39.48±1.81	48.26±5.34	14.73±3.00	9.88±1.14	12.76±1.12	6.03±0.00	
Parambassis wolffii	5.35±0.72	5.08±2.54	6.17±0.58	$8.89 \pm 0.81$	6.32±0.29	< 0.02	3.31±0.04	2.09±0.00	
Parambassis siamensis	12.89±2.31	23.32±0.73	37.35±2.16	24.54±1.06	3.81±0.76	$6.57 \pm 0.68$	12.11±1.59	2.43±0.00	
Notopterus notopterus	1.35±0.19	1.29±0.4	5.59±1.12	4.75±1.02	0.69±0.19	0.24±0.11	0.46±0.19	$0.08\pm0.00$	
Micronema bleekeri	0.69±0.15	0.14±0.07	9.62±2.44	8.37±1.80	2.42±0.67	0.33±0.17	6.29±0.31	< 0.02	



Scientific name	OCs concentrations (ppb)							
	<b><i><u>SBHC</u></i></b>	<b>∑</b> Heptachlor	∑Endosulfan	∑DDT	∑Endrin	Aldrin	Dieldrin	Methoxychlor
Puntius brevis	$2.50\pm0.58$	2.61±1.30	2.00±0.03	14.18±4.27	< 0.02	$0.20 \pm 0.00$	0.73±0.03	< 0.02
Puntius schwanenfeldi	7.33±0.73	4.65±2.06	4.21±0.78	13.42±2.94	< 0.02	2.27±1.15	0.35±0.00	< 0.02
Puntioplites proctozysron	2.99±0.47	1.27±0.38	7.99±0.83	9.66±1.55	1.76±0.06	0.83±0.42	0.95±0.43	0.67±0.00
Henicorhynchus siamensis	< 0.05	2.45±1.23	0.63±0.21	3.74±0.41	< 0.02	< 0.02	< 0.03	< 0.02
Parachela siamensis	3.06±0.34	0.42±0.21	0.87±0.29	3.30±0.59	< 0.02	< 0.02	< 0.03	< 0.02
Oxygaster anomalura	3.12±0.78	3.49±1.13	7.06±0.50	5.87±0.77	0.58±0.29	0.82±0.11	< 0.03	< 0.02
Rasbora argyrotaenia	0.78±0.11	0.90±0.45	1.09±0.36	0.63±0.21	2.27±0.03	1.45±0.09	< 0.03	0.72±0.56
Rasbora borapetensis	3.29±0.31	2.95±1.37	1.43±0.25	3.80±0.74	1.66±0.67	1.03±0.73	0.93±0.79	0.20±0.14
Rasbora tornieri	0.94±0.14	0.35±0.14	1.80±0.13	1.29±0.22	0.61±0.04	0.39±0.00	0.52±0.48	< 0.02
Rasbora sp.1	9.14±1.12	2.09±1.05	8.49±1.17	7.05±0.88	0.41±0.20	0.81±0.00	1.58±0.00	< 0.02
Esomus metallicus	2.49±0.38	2.32±0.86	4.82±0.27	4.59±0.52	0.27±0.00	0.41±0.24	0.25±0.00	< 0.02
Paralaubuca harmandi	$3.62 \pm 0.56$	3.01±0.97	6.64±0.37	4.73±0.50	0.56±0.28	0.69±0.10	< 0.03	< 0.02
Trichogaster trichopterus	3.47±0.36	4.52±1.58	10.04±2.18	11.35±1.96	5.78±0.67	1.45±0.32	0.22±0.14	0.89±0.09
Trichogaster microlepis	3.63±0.46	4.28±1.35	7.61±1.01	24.28±5.79	4.57±0.09	1.85±0.67	0.54±0.44	< 0.02
Clupeichthys aesarnensis	$1.05 \pm 0.07$	2.41±1.20	0.84±0.28	3.55±0.34	0.11±0.06	0.33±0.17	< 0.03	< 0.02
Zenarchopterus ectuntio	2.01±0.29	1.99±0.90	1.51±0.50	< 0.04	0.79±0.39	< 0.02	0.19±0.08	< 0.02
Oreochromis niloticus	8.59±0.90	6.16±3.08	17.71±4.40	15.41±2.65	3.83±1.91	2.03±0.13	< 0.03	< 0.02
Pangasius pleurotaenia	0.69±0.17	3.12±1.47	2.87±0.36	16.44±2.99	3.03±0.94	0.55±0.07	0.46±0.13	< 0.02

Table A-4 OCs concentrations (±SE) found in omnivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

Table A-5 OCs concentrations (±SE) found in detritivorous fish at Khlong 7, Rangsit, Pathum Thani Province during June 2004 to May 2005.

Scientific name	OCs concentrations (ppb)								
	∑внс	∑Heptachlor	∑Endosulfan	∑DDT	∑Endrin	Aldrin	Dieldrin	Methoxychlor	
Macrognathus siamensis	5.16±0.94	5.50±0.37	49.18±4.49	34.6±2.40	10.42±5.21	5.69±1.38	9.55±0.09	13.18±0.95	
Mystus mysticetus	$1.40\pm0.18$	1.03±0.19	10.48±1.84	$10.09 \pm 1.69$	1.45±0.73	0.33±0.15	0.92±0.20	0.63±0.00	
Heterobagrus bocourti	$0.06 \pm 0.02$	0.96±0.48	25.72±4.48	14.29±2.38	1.69±0.85	0.44±0.12	0.07±0.02	< 0.02	
Mystus singaringan	0.51±0.76	1.04±0.15	14.63±1.97	17.68±2.42	5.60±2.52	0.33±0.10	1.69±0.09	1.02±0.00	
					01110				