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Distributions and Congener Patterns of PCBs in Fish from Major Aquaculture Areas in the Pearl River Delta, South China

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ABSTRACT

The distribution and concentrations of polychlorinated biphenyls (PCBs) were determined in surface sediments and fish collected from freshwater fishponds in six major aquaculture areas of the Pearl River Delta. The concentrations of total PCBs ranged from 7.32 to 36.2 ng/g (dry weight) in sediments and 5.15 to 226 ng/g (lipid weight) in five species of fish, with higher concentrations in fishponds from two industrialized areas. Feeding habits of fishes played a significant role on the accumulation of PCBs and their homologue patterns in fish tissues, with higher concentrations in muscle and viscera of mandarin fish (*Siniperca kneri*), and tilapia (*Tilapia mossambica*) and lower in grass carp (*Ctenopharyngodon idellus*). In muscle, IUPAC No. 118, 138, 81/87, 153, 180, 52, 49, 99, and 44 congeners were the most dominant out of the 36 congeners measured in the present study. The contents of PCBs in fish cultivated in the Pearl River Delta were rather low when compared with the maximum concentration of total PCBs of 2.0 μg/g (wet weight), imposed by the U.S. Food and Drug Administration edible seafood. However, due to the bioaccumulation and biomagnification nature of PCBs through the food chain, continuous monitoring of PCBs as well as other Persistent Organic Pollutants in this rapidly developed region is encouraged.

Key Words: polychlorinated biphenyls, freshwater fishponds, pond sediment, fish muscle, fish viscera.

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INTRODUCTION

Environmental impacts caused by trace organic pollutants such as polychlorinated biphenyls (PCBs), polycyclic aromatic hydrocarbons (PAHs), polychlorinated dibenzo-p-dioxins (PCDDs), polychlorinated and dibenzofurans (PCDFs) have received increasing attention due to their persistency and tendency to bioaccumulate through the food chains (Lee *et al.* 2001). Once released into the aquatic environment, they can be easily adsorbed onto suspended particles or taken up and concentrated by aquatic organisms, and could even be biomagnified along the food chain, and pose potential hazards to other living organisms, including human beings (Ashley *et al.* 2000; Fontenot *et al.* 2000; Pruell *et al.* 2000; Kassa and Bisesi 2001).

PCBs have been used in the past decades as anti-conductivity lubricating oil and an additive in the plastic and painting industries due to their special physicochemical properties such as low reactivity, high dielectric constant, and degradability throughout the world. As a consequence, PCBs have been dispersed on a global scale, even found in pristine places such as the Arctic and Antarctic (Bard 1999). In China, about 9×10^6 kg of PCBs were used as insulators in electrical transformers and lubricating oils since the 1960s. Although banned in the 1980s, some old transformers and capacitors containing large amounts of PCBs have not been disposed of properly and some of them are still in use (Hong *et al.* 1997). Used electrical equipment and the leakage from broken transformers continue to be a major anthropogenic source for PCBs transferred to the environment (Fu *et al.* 2003). In most regions, particularly those areas far away from industrial activities, PCBs levels remain constant or even increase as a consequence of environmental redistribution (Borrell *et al.* 2001).

Being a fluvial deltaic plain with an area of 10,000 km^2, the Pearl River Delta has been known as the "homeland for rice and fish" due to its fertile soil and abundant water resources. The dike-pond systems for integrated agriculture and aquaculture are centuries-old traditions for maximizing energy sources, with the mulberry-dike-fish-system a well-known representative. Leaves of the mulberry trees that grow on dikes of the fishponds are fed to silkworms and the cocoons after extraction of silk are used as fish food. The pond mud is excavated and used as fertilizer for growing the mulberry trees. In addition, digested pig manure is discharged into fishponds and serves as pond fertilizer for enriching the pond water, where several fish species are cultivated in the same pond. All the resources are therefore utilized efficiently under polyculture of fish having different feeding modes, with animal manure as a major nutrient source (Wong *et al.* 2004). However, the rapid industrialization and urbanization in the region during the past two decades have drastically changed the local environment, in particular the dike-pond systems. The traditional fishpond culture models (such as mulberry-dike-fishponds) have given way to monoculture of fish, usually under high density, in which a large amount of forage and high protein feed pellets are used. However, the pond mud is no longer used as fertilizer for crops growing on the dikes. A large amount of chemical pollutant from the discharge of industrial and urban effluent has caused the deterioration of water quality. The pond mud may therefore act as a sink for various chemical compounds, and the fish will become more susceptible to various diseases.

The presence of persistent organic pollutants such as hexachlorocyclohexanes (HCHs), polycyclic aromatic hydrocarbons (PAHs), dichlorodiphenyltri chloroethanes (DDTs), and PCBs in water and sediments from the Pearl River Delta have been reported recently (Liang *et al.* 1999; Kang *et al.* 2000; Mai *et al.* 2002), but there is a lack of information on PCBs in fish cultivated in freshwater fishponds (Zhou and Wong 2000; Zhou *et al.* 1999). Furthermore, most of the investigations on PCBs are limited to total PCBs instead of congeners (Zhou *et al.* 1999; Kang *et al.* 2000; Fu *et al.* 2003). In view of the this background, the objectives of the present study were to: (1) investigate the concentrations and conger patterns of PCBs in sediments and fish from fishponds of major aquaculture sites around the Pearl River Delta; (2) compare the levels of PCBs contamination in fish collected from these fishponds; and (3) assess the uptake of PCBs by different fish species that have different feeding habits.

MATERIALS AND METHODS

The sampling sites were located at six different regions representing the major aquaculture areas in the Pearl River Delta (Figure 1) (Fangcun Country of

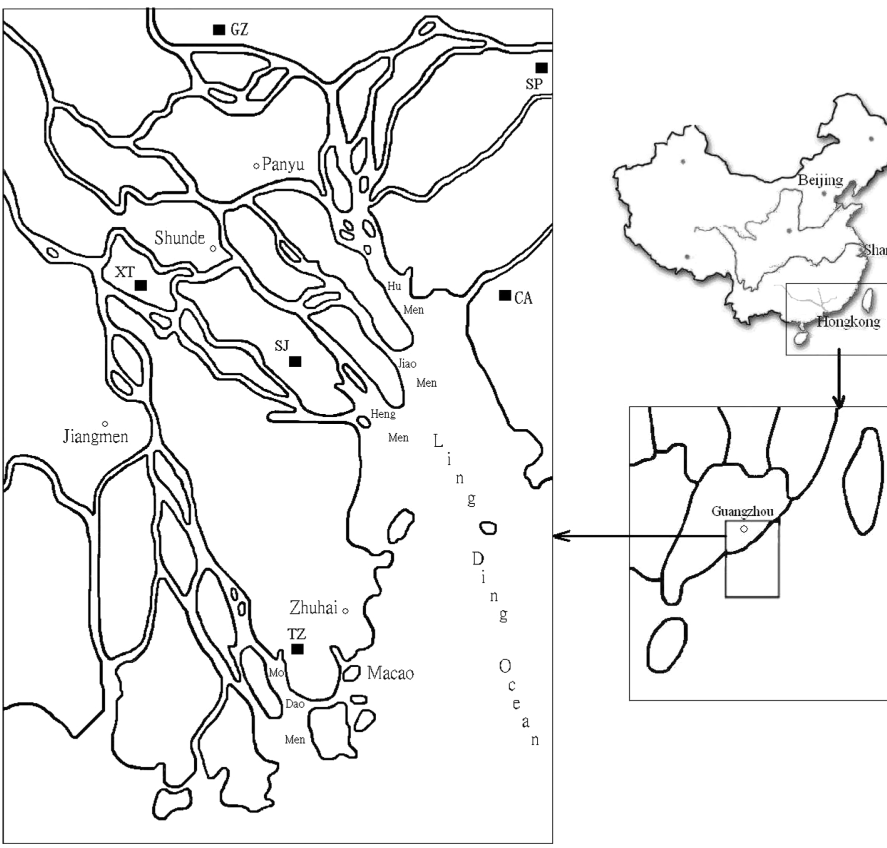

Figure 1. Locations of fishponds of six major aquaculture areas within the Pearl River Delta (refer to Table 1 for site details).

Guangzhou City [GZ], Xingtan Town [XT] of Shunde City, Shipai Town [SP] of Dongguan City, Changan Town [CA] of Shengzhen City, Tanzhou Town [TZ] of Zhuhai City, Sanjiao Town [SJ] of Zhongshan City). The fishponds represented two types of aquaculture models: (1) the high-density monoculture model in which high-quality fish such as mandarin fish (*Siniperca kneri*) are fed with smaller live fish such as cuvier fish (*Cirrhina molitorella*), tilapia (*Oreochromis mossambicus*) is also cultivated under high density and sometime serves as feed for mandarin fish; and (2) the polyculture model in which different fish species including bighead (*Aristichthys nobilis*), grass carp (*Ctenopharyngodon idella*), and crucian carp (*Carassius auratus*) are raised simultaneously. CA and GZ are the two sites with rapid industrialization and urbanization (Table 1).

Sample collection was conducted during May–June 2000. Fish samples were collected manually using nets from 2–4 fishponds at each site (refer to Table 1 for the number, length, and weight of five different species collected from six study sites). Sediments were collected using a grab sampler (Ekman-Brige), with six samples collected from each pond, and each sample consisted of 3–5 sub-samples, which were mixed thoroughly on site. All samples were transported at a temperature under 4°C to the laboratory at the Croucher Institute of Environmental Sciences within the same day. All the fish were divided into muscle and viscera, before they were homogenized and freeze-dried. The sediment samples were also freeze-dried. All fish and sediment samples were stored in desiccators (at about 20°C) until analysis.

The extraction of PCBs was conducted according to the method described in AOAC (1990). Freeze-dried fish samples (10 g) were extracted by 80 mL *n*-hexane in a water bath for 8 h using a soxhlet apparatus. Lipid content was measured by evaporating the solvent until a constant weight was obtained. Twenty mL of n-hexane was added to dissolve the lipid and transferred into the separatory funnel. They were then washed by concentrated sulfuric acid and passed through a microflorisil column to remove most of the lipid and interference compounds.

Freeze-dried sediment samples were passed through a 1 mm mesh sieve to separate the stones, leaves, and dead invertebrates before grinding into powder using a mortar and a pestle. About 10–15 g sediment was transferred into a soxhlet apparatus and extracted by 80 ml acetone: *n*-hexane (1:1, v/v) in a water bath for 12 h. Before extraction, sediment was soaked with about 10 mL water for 30 min. The sediment extracts were then cleaned by concentrated sulfuric acid, copper powder, and a microflorisil column (Erickson 1997).

The determination of PCBs (Method 8082, USEPA 1996) for fish and sediment samples was conducted using a Hewlett-Packard 6890 GC-ECD equipped with a ^{63}Ni electron-capture detector and an automatic sampler. The capillary column was a 30 m DB-1 (100% dimethysiloxane) (CJ & W Scientific, USA) with an internal diameter of 0.25 mm and a stationary phase thickness of 0.25 μm. The oven temperature was programmed from the initial temperature of 80°C (holding for 1 min), then to 180°C (holding for 5 min) at the rate of 20°C/min; and to 280°C at the rate of 5°C/min. The injection was operated in splitless mode at 250°C and detector temperature was maintained at 300°C. Nitrogen gas was used as carrier gas and make-up gas.

A mixture of 36 PCB congeners in isooctane (Accustandard, USA) was used as the PCB standard. These 36 congeners are considered environmentally threatening due to their frequency of occurrence in environmental samples, abundance in

Table 1. Site descriptions and details of fish samples collected from the freshwater fishponds in the Pearl River Delta.

Sampling sites			Polyculture			Monoculture	
Abbreviations	Site details	Parameters	Bighead	Grass carp	Crucian carp	Tilapia	Mandarin fish
TZ	Aquaculture base	Number	3	3	—	—	—
Zhuhai	of the Dajating	Length (cm)	39–41	51–53	—	—	—
	Company	Wet weight (g)	1250–1750	1800–2200	—	—	—
SJ	Banana and grass on	Number	3	3	5	4	4
Zhongshan	the pond dikes	Length (cm)	31–38	35–48	19–24	21–22	19–25
		Wet weight (g)	510–830	1200–2060	190–210	200–260	180–230
XT	Near a small road and	Number	—	4	5	—	12
Shunde	a stream	Length (cm)	—	45–50	21–25	—	11–17
		Wet weight (g)	—	1500–2150	220–280	—	50–120
GZ	Near a factory	Number	3	3	4	5	5
Guangzhou	producing fish feeds	Length (cm)	45–51	46–55	31–33	31–36	25–31
		Wet weight (g)	2000–2500	1750–2750	400–420	350–500	260–320
SP	Pig styles and duck	Number	3	4	4	5	20
Dongguan	farms nearby	Length (cm)	32–39	41–56	24–29	19–22	16–19
		Wet weight (g)	550–850	1450–2260	280–350	200–260	65–100
	Near industry	Number	3	3	4	4	—
CA	districts and a	Length (cm)	41–45	51–60	26–31	31–37	—
Shengzhen	transform capacity station	Wet weight (g)	850–1500	2100–3000	310–380	390–520	—

Aroclors, and their potential toxicity (McFarland and Clarke 1989). They included Group A: three congeners are the most toxic and characterized as pure 3-Methyl cholanthrene-type (3-MC) inducer, and six congeners are mixed-type inducers and are very abundant in Aroclors as well as in the environment (IUPAC No: 77, 126, 169, 105, 118, 128, 138, 156, 170); Group B: seven congeners are Phenobarbital-type (PB) inducers for Mixed-Function Oxidase enzymes and are less toxic but most abundant in the environment (87, 99, 101, 153, 180, 183, 194); and Group C: ten congeners are weak or not inducers but more common in animal tissues (18, 44, 49, 52, 70, 74, 151, 177, 201); Group D: ten congeners have potential toxicity, but with very low presence in animal tissues (37, 81, 114, 119, 123, 157, 158, 167, 168, 189).

Octachloronaphthalene (OCN) was used as an internal standard. Standard reference materials (SRM) 2977 for mussel tissue (organic contaminants and trace elements) and 1939a for river sediments (PCBs congeners) were obtained from the U.S. Department of Commerce, National Institute of Standards and Technology Certificate of Analysis for QA/QC purpose. A one-way ANOVA test, followed by the least significant difference test (LSD) in the statistical software program SPSS were used to evaluate any significant differences (at $p < .05$) in terms of PCB concentrations among fish and sediment samples.

RESULTS AND DISCUSSION

Sediment Samples

For most of the persistent organic pollutions (POPs), including PCBs in the aquatic environment, sediments serve as the final sinks due to their hydrophobic and persistent nature (Menone *et al.* 2001). Therefore, the study of PCB concentrations in sediments could reflect not only the present but also the past environmental status of PCBs.

The patterns of PCB congers and total PCB concentrations in the sediments collected from the six study sites are shown in Figure 2. The total concentrations of PCBs in sediments ranged from 7.32 to 36.2 ng/g (dry weight), with an average of 14.5 ng/g. The highest value (36.2 ng/g) was obtained at the CA site, followed by the GZ, SP, XT, SJ, and TZ sites. There were significant differences between CA and the other five sites ($p < .01$), and also between GZ and four other sites ($p < .01$), whereas no significant differences were observed among the SP, XT, SJ, and TZ sites. This was probably due to the fact that the aquaculture and agriculture activities of both CA and GZ sites had been diminished and the sites had been gradually transformed to industrial zones during the past 20 years. The total area of the CA fishpond is now only measured at 0.35 ha. The site is surrounded by an electrical power plant, a transformer station, and a leather shoe making factory, all about 50 m distant. The water of the fishponds is rather stagnant, which has further aggravated the situation, as it only relies on rainfall for replenishing the pond water. This resulted in the ΣPCBs being 2.5–5-fold higher than other sites. GZ is situated next to a factory producing fish feeds, whereas other sites are located at more remote areas.

In general, the representative congeners were IUPAC No. 87/81, 153, 118, 138, 52, 170, 49, 70, 101, and 119 among all the sites. The homologue patterns of PCB congeners of CA were characterized by: 118 > 87/81 > 138 > 153 > 52 > 70 > 49 > 180 > 170 > 101 > 18, especially the lower chlorine substituted congeners including

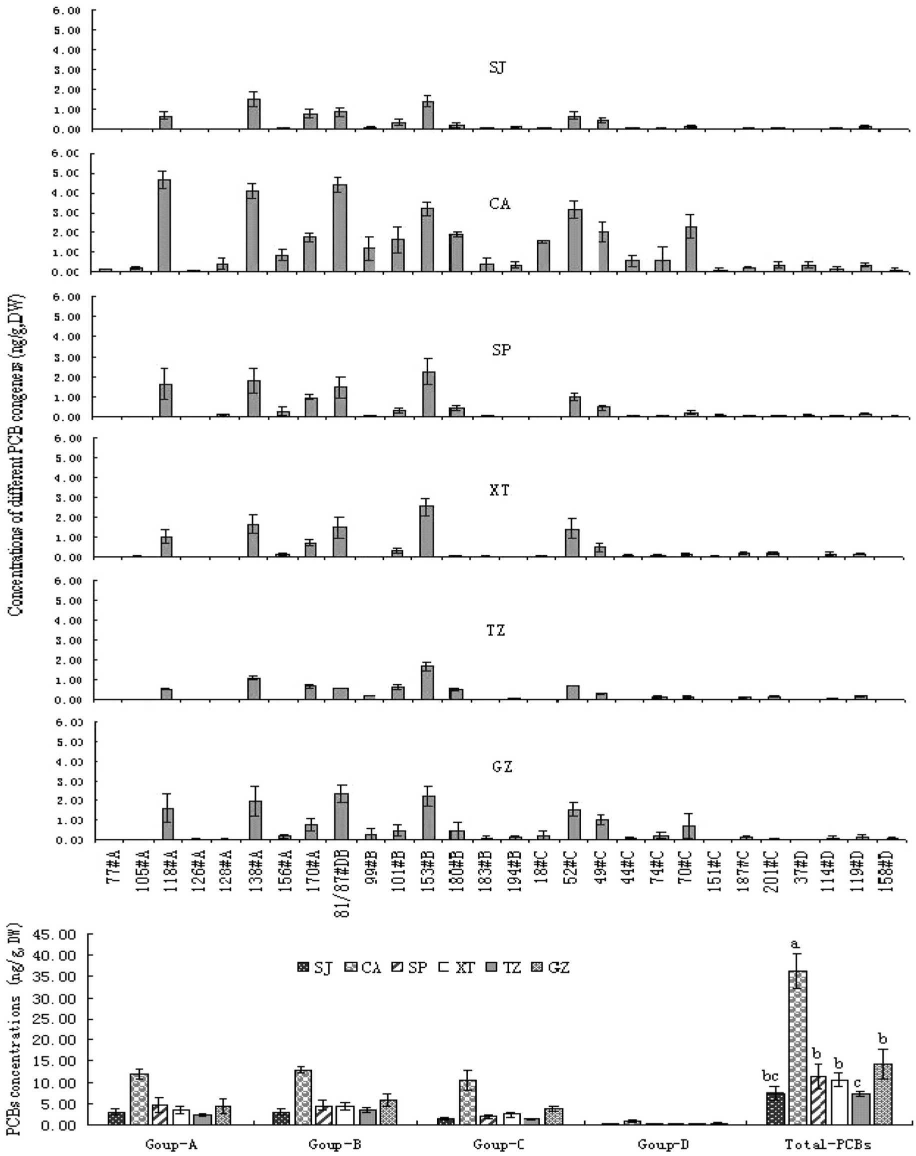

Figure 2. The patterns of PCBs congeners and total concentrations of PCBs in the pond sediments from six major aquaculture areas in the Pearl River Delta (refer to Table 1 for site details). Error bars indicate standard deviations. For total PCBs, same letters on top of bars indicate no significant difference at ($p < .05$).

18 and 70, which were either absent or existed at very lower concentrations at other sites. This further confirmed that point source contamination was evident at the CA site, because less chlorinated PCB congeners are more prone to be decomposed (Miao *et al.* 2000). On the contrary, the SJ site is a rural area where traditional

agriculture and aquaculture have been maintained, which accounted for the lowest concentration of PCBs in the sediments (average value 7.32 ng/g, dw), with the ranking of congeners: 138 > 153 > 81/87 > 170 > 118 > 52 > 49.

In general, the present study indicated that the total PCBs of pond sediments (7.32–36.2 ng/g, dw) are higher than the pond sediments (0.646–9.95 ng/g, dw) located within Mai Po Marshes (Hong Kong), a remote nature reserve (Liang *et al.* 1999). It has been observed that river sediments of the West River (a tributary of the Pearl River) were 11.1–14.9 ng/g, dw (Mai *et al.* 2002), and the more polluted river sediments of the Shing Mun River (an industrially polluted stream, with a variety of factories such as plasticizers, electricity, and painting, situated in the New Territories of Hong Kong) were 43.0–461 ng/g, dw (Zhou *et al.* 1999).

Fish Samples

The average concentrations of total PCBs in muscle of the 5 fish species in descending order (ng/g, lipid weight) were: 166 for tilapia, 156 for mandarin fish, 131 for bighead, 97.4 for crucian, and 91.6 for grass carp. The average concentrations of total PCBs in viscera were slightly different, with mandarin fish having the highest concentration of 195, followed by tilapia 190, bighead 179, crucian 148, and grass carp 112. The analysis of homologue patterns of different PCB congeners showed that IUPAC No. 118, 153, 81/87, 138, 170, 119, 114, 180, 101, 99, 52, 49, 74, and 70 were commonly detected in most of the fish samples (as well as the sediment samples); 126, 128, 157, 167, 168, 169, and 37 were not detected (Figure 3).

The concentration and distribution of PCBs in fish are controlled by many factors including the species, the lipid content in body, age, size, gender, growth rate, and food choice (Bremle an Larsson 1998; Ashley *et al.* 2000). In general, the position in the food chain or feeding habits of the organism play an important role in the accumulation of the POPs, with species situated at higher trophic levels tending to accumulate the most POPs, including PCBs (Johnson *et al.* 1996; Zhou *et al.* 1999; Gunnarsson and Skold 1999). Mandarin fish is a carnivorous species, whereas tilapia is an omnivorous species, which accounted for their higher uptake of PCBs in both muscle and viscera. Bighead is a zooplanktivorous species that filters zooplankton, which may concentrate pollutants from water due to its great body surface (Wang *et al.* 1998). Significant correlation coefficients were observed between total PCBs in sediments with fish muscle ($r = .92$, with $p < .01$) and viscera ($r = .88$, $p < .01$). The positive correlation of PCBs between the sediments and biota samples often occurs in environments near contaminated sites (Bazzanti 1997). This explained why fish collected from the CA site had the higher total PCBs in both muscle and viscera.

The representative congeners in all fish samples, in descending order were: 153 > 138 > 118 > 87/81 > 180 > 52 > 170. This may indicate that the highly chlorinated congeners (138, 153, and 180) had perhaps greater resistance to metabolism and elimination than the lower congeners and so more prone to accumulate in fish, which occupied the higher trophic position (Jacob and Boer 1994). However, highly chlorinated congeners such as 194, 201, and 189 generally possessed lower bioavailablity due to their large molecular structure (Bremle *et al.* 1995).

It was also observed that fish viscera usually contained higher chlorinated congeners of PCBs such as hexa- and penta-chlorobiphenyl when compared with muscle,

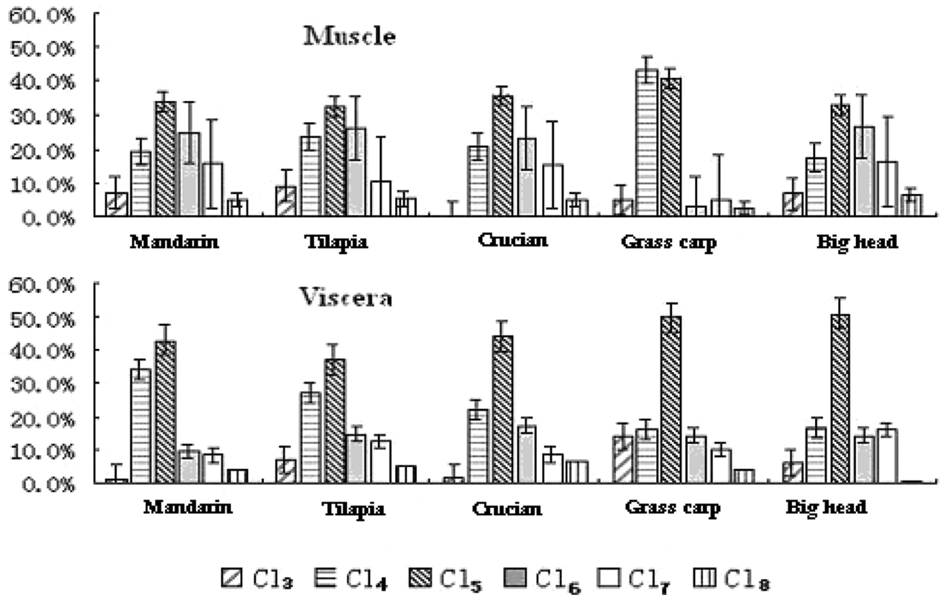

Figure 3. Percentages of different chlorine substituted congeners in muscle and viscera of fish collected from different fish species. Error bars indicate standard deviations.

indicating the lipophilic compounds like PCBs are prone to accumulate in lipids, especially for the moderately chlorinated PCB congeners.

The biological activity of individual PCBs is a function of extent and pattern of chlorine substitution (Storelli and Marcotrigiano 2003). "Congener-specific" PCB analysis of biotic tissues has gained increasing importance in assessing possible links between PCB exposure and toxic effects. In the present study, 36 congeners from di- to octa-chlorobiphenyls were analyzed in all fish samples. Results showed that the flesh-eater species (such as mandarin fish, and to a certain extent tilapia and bighead) had higher concentrations of higher chlorinated PCB congeners than other fish species such as grass carp. For example, penta-, hexa-, hepata-, and octa-chlorobiphenyls accounted for up to 70–80% in muscle of mandarin fish, 50–60% for tilapia and only 40–45% for grass carp (Figure 3). Our early investigation studying PCB congeners in muscle of tilapia collected from streams in Hong Kong also indicated that these congeners accounted for almost 60% of the total PCBs (Zhou *et al.* 1999).

Based on the present results, the concentrations of PCBs in fish cultivated in fishponds within the Pearl River Delta were low, when compared with the maximum concentration of total PCBs of 2.0 μg/g (wet weight) in seafood imposed by the U.S. Food and Drug Administration (FDA 2001).

On the other hand, when based on the classification of McFarland and Clarke (1989), PCBs Group B (81/87, 99, 101, 153, 180, 183, 194) accounted for the highest proportion of 32–47% of total PCBs in the muscle of different species (except grass carp, in which PCBs congeners were dominated by Group C), followed by Group C (18, 44, 49, 52, 70, 74, 151, 177, 187, 201) of 20–24%, Group A (105, 118, 126, 128, 138, 156, 169, 170) of 19–34%, whereas Group D (37, 114, 119, 123, 157, 158, 167, 168, 189) accounted for the lowest proportion of 7–17%. The potential health effects of PCB Group A should not be overlooked as they are the most toxic and characterized as pure 3-Methyl cholanthrene-type (3-MC) inducers and mixed-type inducers (McFarland and Clarke 1989).

CONCLUSION

The rapid socioeconomic development in the Pearl River Delta has imposed adverse environmental effects. Based on the present results on the concentrations of PCBs in sediments and fish collected from six major aquaculture sites in the area, there are potential point sources of PCB contamination. The feeding habit or the trophic level of fish species in the food chain plays a major role in the abundance and relative ratio of the homologue patterns of PCBs in fish. The fact that carnivorous and omnivorous fish usually have higher concentrations of PCBs and a higher proportion of highly chlorinated congeners in their body seemed to provide an obvious evidence of biomagnification. There is an urgent need to monitor PCBs as well as other persistent organic pollutants in our rapidly changing environment.

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