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Tissue-specific and whole-fish accumulation of polychlorinated biphenyls by juvenile Baltic salmon (*Salmo salar* L.) after oral gavage exposure

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The bioaccumulation and accumulation efficiencies of four different polychlorinated biphenyl congeners (PCB-18, PCB-44, PCB-137, PCB-169) were measured in short-term (14-42-d) bioaccumulation tests performed on juvenile Baltic salmon (*Salmo salar* L.). The PCBs were administered to the fish through oral gavage implantation of gelatine capsules once a week. The given dosages were similar to the concentrations found in the main prey of Baltic salmon in the Gulf of Finland. The accumulation efficiencies of the four PCB congeners occurred in the order: PCB-44 > PCB-18 ≈ PCB-137 > PCB-169. All the four PCB congeners accumulated to higher concentrations in lipid-rich intestines as compared with PCB bioaccumulation in liver, muscle sample and carcass remaining after those tissue removals. While the relative accumulation efficiency of the salmon intestine was 21.3% (SD = 10.1%), as much as 74.0% (SD = 12.7%) of the PCBs was assimilated in a salmon carcass. Average total accumulation efficiency of total PCB was 71%.

Introduction

The ecotoxicological risk of polychlorinated biphenyls (PCBs) in aquatic systems including catchments and remote areas continues to be a topic of concern due to their high chemical, thermal and biological stability as well as their toxicity (Hutzinger *et al*. 1974, Lech and Peterson 1983, Tanabe *et al*. 1987, Manahan 1994, Willman *et al*. 1997, Van den Berg *et al*. 1998, Isosaari *et al*. 2004). Although their production and use were prohibited or at least strictly regulated in many countries during the early 1970s, the levels found in biological samples remain high due to the persistence of PCBs and to leakage from PCB-containing products and wastes (Andersson *et al*. 1997).

Salmonid fish acquire their PCB load mainly by consuming food contaminated by PCBs (Shaw and Connell 1986, Stow and Carpenter 1994, Madenjian *et al*. 2000, Vuorinen *et al*. 2002). Thus, the major route of bioaccumulation of PCBs by fish is consumption of contaminated food followed by passage through the stomach walls. Since the early 1980s, high concentrations of PCBs have been detected in Baltic salmon (*Salmo salar*) and Bothnian Bay trout (*Salmo trutta*) in Finland, Europe, as well as in other fish throughout the world (Swain 1983, Vuorinen *et al*. 1985, Tanabe *et al*. 1987). PCB and dioxin

concentrations in fish have decreased over time, but they seemed to be levelled out in Baltic fishes during last decade (HELCOM 2004). The concentrations observed recently may still have remarkable effects on the health of organisms. A recent study of Vuorinen *et al*. (2002) pointed out that high coplanar PCB and PCDF (polychlorinated dibenzo-*p*-furans) concentrations in Baltic salmon coincided with an outbreak of yolk-sac mortalities (M74 syndrome — a reproductive disturbance) at the beginning of the 1990s.

The solubility of PCBs in water is very low, and generally decreases with increasing chlorination (Hutzinger *et al*. 1974). The very same features that made the extraordinarily stable PCBs so industrially desirable also affect their tendency to partition into biologic tissues (Swain 1983, Manahan 1994) or into sediments, which act as xenobiotic sinks where the compounds can reenter the foodweb. Additionally, specific substitution patterns of PCBs, such as the number of chlorine atoms in the *ortho*-position and the number of adjacent chlorines, play an important role in their biomagnification potential (Shaw and Connell 1986). The physicochemical characteristics of PCBs have been described in detail (Hutzinger *et al*. 1974) and are shown in Table 1 for the four PCBs used in this study: PCB-18 is *tri*-chlorinated, PCB-44 *tetra*-chlorinated and PCBs 137 and 169 *hexa*-chlorinated. PCB-169 deviates from its *ortho*-substituted congeners in being coplanar with no chlorine in the *ortho*, i.e. 2,2´,6,6´ position. This non-*ortho* IUPAC congener with no unsubstituted carbon atoms adjacent on the biphenyl ring therefore exhibits maximum TCDD-like (2,3,7,8-tetrachlorodibenzo-*p*-dioxin) activities such as high stability, and is among the most toxic PCB congeners in mammals and fish (Willman *et al*. 1997, Van den Berg *et al*. 1998).

A great deal of information is available about whole-fish accumulation efficiencies of different organochlorines (OCs) and PCB congeners (Niimi and Oliver 1983, Burreau *et al*. 1997, Dabrowska *et al*. 1999, Madenjian *et al*. 2000, Andersson *et al*. 2001, Wong *et al*. 2002, Buckman *et al*. 2004, Isosaari *et al*. 2004) but information on the tissue specific (muscle, liver, intestine, gonads, carcass) accumulation and allocation in individual fish is limited (Tietge *et al*. 1998, Nichols *et al*. 2001, Jørgensen *et al*. 2002).

Often the comparability between the different field sampling studies of OCs is also rather poor due to differences in sampling of the fish (fillet with skin or not etc.). Amrhein *et al*. (1999) showed that individual variation in tissue-specific concentrations are high and estimation of whole fish concentrations from fillet data is difficult. Most of the PCB analyses in fishes have been made from muscle or fillet samples, because advisory criteria for human consumption are often based on fillet concentrations. Whole fish concentrations are, however, needed for bioaccumulation, accumulation efficiency and trophic transfer estimates (Amrhein *et al*. 1999, Madeijan *et al*. 2000).

This study reports short-term PCB bioaccumulation in liver, lipid-rich intestine, muscle, gonads and the remnant fish carcass (with skin) after oral gavage exposure. Secondly, we also calculated the retention efficiencies of four PCB congeners by analysing whole-body chemical residues of Baltic Sea salmon. Thirdly, whole-fish/muscle-PCB ratios were estimated in order to demonstrate the variation in PCB allocation

Table 1. Properties of the polychlorinated biphenyl congeners used in this study. IUPAC = International Union of Pure and Applied Chemistry.

IUPAC No.	Number of chlorines	Percentage chlorine	Substitution pattern	*o-m-p*[a]	Log K_{ow}[b]
18	3	41.3	2,2´,5	2-1-0	5.24
44	4	48.6	2,2´,3,5´	2-2-0	5.75
137	6	58.9	2,2´,3,4,4´,5	2-2-2	6.83
169	6	58.9	3,3´,4,4´,5,5´	0-4-2	7.42

[a] number of chlorine atoms in the *ortho*, *meta* and *para* positions.
[b] octanol-water partition coefficient values from Hawker and Connell (1988).

between juvenile salmon individuals after short-term PCB exposure.

Material and methods

Chemicals

The studied polychlorinated biphenyl congeners (PCBs) 2,2´,5-trichlorobiphenyl (PCB-18), 2,2´,3,5´-tetrachlorobiphenyl (PCB-44), 2,2´,3,4,4´,5-hexachlorobiphenyl (PCB-137) and 3,3´,4,4´,5,5´hexachlorobiphenyl (PCB-169) as well as the 2,4,6-trichlorobiphenyl (PCB-30) used as an internal standard in the compound concentration analyses were purchased from AccuStandard Inc. (New Haven, CT), and all the PCBs had a purity of ≥ 99.0% (SD = 0.5%). The four PCB congeners were selected to encompass a range of lipophilicity and some structural variation as shown in Table 1. The log-transformed octanol-water partition coefficients indicating the lipophilicities of the compounds and thus their bioaccumulation potentials according to Hawker and Connell (1988) varied from 5.24 for PCB-18 to 7.42 for PCB-169.

The PCB congeners were weighed with a microbalance and dissolved with slow mixing into pure commercial fish liver oil to create a stock solution of 151.3 mg kg^{-1} of the total PCBs, and of 32.7 (18) 38.3 (44) 40.7 (137) 39.7 (169) mg l^{-1} of the individual PCBs. All solvents, anhydrous sodium sulphate and concentrated sulphuric acid used in sample preparation for PCB concentration analyses were of analytical grade, except acetone and hexane, which were HPLC grade.

Experimental fish

Juvenile Baltic salmon (*Salmo salar*) weighing on average 124.0 g (SD = 35.0) were obtained from Hanka-Taimen Oy aquaculture station (Finland). Fish were kept in three groups of 20 (PCB-exposed), 10 (carrier control fish) and 14 (pure control fish) individuals in flow-through 220 l pools. The dry feed of fish (Biomar Ecostart, pellets > 2 mm; 48% protein, 23% fat, 12% carbohydrate and 6.5% ash according to the manufacturer) was analysed as uncontaminated and for fat content (mean = 27.8%, SD = 3.0, n = 4) as described later and the fish were fed once a day in excess with this food. Prior to fish marking, they were anaesthetized with commercial clove oil, which was diluted with 95% ethanol (1:9 v/v) and dosed at approximately 40 mg l^{-1} per fish (Hoskonen and Pirhonen 2004). The fish were PIT-marked (Biomark Inc., Boise, ID, USA) into the back muscle between the dorsal and adipose fins using 12 G needles. Thereafter the fish were acclimatized for eight weeks in continuously exchanging water (0.5 l min^{-1}) at 12 ± 0.5 °C with a 16:8 light:dark photoperiod.

Experimental design and fish sampling

Prior to exposures the fish were again equally anaesthetized with clove oil. Commercial 0.5 ml gelatine capsules were filled with 12.5 μl of the PCB stock solution, and the capsules cautiously inserted into the stomach of the anaesthetized fish by placing a plastic inserter tube into the mouth of the fish and pushing the filled capsule through the tube with a glass rod. Fish were given spiked capsules weekly and were sampled after 14 (n = 7) and 25 (n = 9) days of exposure. Additionally, four fishes (which received 4 spiked capsules) exposed for 25 days were allowed to assimilate the given PCBs for another two weeks. The absorption times after administering the last capsules were 7, 4 and 21 days before the fish sampling, for 14, 25 and 42 days exposures, respectively. Thus the individual congener dosages were 0.90 (SD = 0.09) and 1.79 (SD = 0.17) μg per fish, and the PCB total dosages 3.58 and 7.17 μg per fish for 14-day exposure and 25- as well as 42-day exposure, respectively. The PCB single dosages per fish fresh weight were 30.2 (SD = 5.9), 48.9 (SD = 14.6) and 49.4 ng g^{-1} fw (fresh weight) (SD = 12.3) for 14, 25 and 42 days exposures, respectively. These dosages were chosen to be of the same order of magnitude as the total PCB concentrations (hereafter 'total PCBs') measured recently by Vuorinen *et al.* (2002) for the main prey of Baltic salmon, 1 to 3-year-old Baltic herring (31 ng g^{-1} fw) and 2 to 10-year-old sprat (77 ng g^{-1} fw).

At each sampling time, fish weight and length were measured and fish were opened. Tissue samples for PCB analyses were taken from the back and side muscles (mean = 7.5 g fw, SD = 1.5), liver (mean = 1.2 g fw, SD = 0.5), intestine (mean = 6.4 g fw, SD = 2.3) and gonads (mean = 0.3 g fw, SD = 0.2). A muscle cross-section from spinal column to abdomen, including the subcutaneous fatty layer towards the abdomen, served as a subsample of muscle. Total weight of the organs examined and the mean weight of 118.4 g fw (SD = 29.8) for whole-fish (fish carcass remaining) was measured for the calculation of the total PCBs in fish. Possible contents of the intestine were flushed out with distilled water before the weighing and sampling. All the samples were enfolded in aluminium foil prior to freezing at –18 °C (fish carcass) and at –85 °C (tissue samples).

The exposures were carried out in continuously exchanging water (0.5 l m^{-1}) at 12 ± 0.5 °C with a 16:8 light:dark photoperiod. Water temperature was measured every 15 min with temperature minilogs (Vemco Ltd, Shad Bay, NS, Canada) from the positive control and exposure pools. Fish faeces in the pools were cleaned daily with a syphon.

The temporal treatments were not replicated to prevent the fasting observed when fish were held alone. Control fish, carrier and pure, for each exposure treatment, were kept under similar conditions and sampled simultaneously with the exposed fish. Fish serving as positive controls received the same amounts of pure fish liver oil (PCB vehicle alone) in gelatine capsules as the exposed fish (25 μl: 14 days treatments, and 50 μl: 25 and 42 days treatments) whereas pure control fish received none. Details of the experimental design, exposure doses and sampling are shown in Table 2.

PCB analyses

Gonads, muscle, liver and intestine samples of four fish per PCB exposures and pure control fish were ground until homogenous in a mortar with approximately four times more anhydrous sodium sulphate than the sample mass. Whole-fish (carcass remaining with the skin) were homogenized with a steel blender (GWB, New Hartford, CONN.). Subsamples of 3–8 g fw of homogenized carcass, 0.05–0.5 g fw of gonads, 2–4 g fw of muscle, 0.5–2 g fw of liver and 2–5 g fw of intestine, respectively, were used for PCB analyses.

Fish food (2–3 g fw, n = 4) as well as the commercial fish liver oil (n = 4) used as a PCB vehicle were analysed simultaneously. The congener concentrations in the PCB stock solution (n = 3) were also measured. The PCB-18, -44, -137 and -169 concentrations in food and liver oil were under analytical detection limits (< 0.1–0.5 ng g^{-1} fresh weight). PCBs in the tissue samples of control fish (carrier control, n = 12) were analysed and the concentrations of the four studied congeners were under analytical detection limits.

All the tissue samples including the fish carcass remaining, fish food and fish liver oil were extracted for six hours with petroleum ether:

Table 2. Experimental design for PCB exposures of juvenile Baltic salmon. Number of individuals per treatment and analyses and PCB dosages. PCB column indicates fish exposed to PCBs 18, 44, 137 and 169; and C+ and C– positive (carrier only) and negative control treatments, respectively.

	Exposure treatment								
	14 days			25 days			42 days		
	PCB	C+	C–	PCB	C+	C–	PCB	C+	C–
No. of fish per treatment	7	4	5	9	4	5	4	2	4
No. of fish per PCB analyses	3	–	4	4	–	4	4	–	4
Dosage of total PCBs (µg per fish)	3.6	–	–	7.2	–	–	7.2	–	–
Dosage of total PCBs (ng g^{-1} fish fw)	30.2	–	–	48.9	–	–	49.4	–	–
Fish liver oil dose (µl)	25	25	–	50	50	–	50	50	–

acetone:hexane:diethyl ether (9:5.5:2.5:1, v/v) using a warm Soxhlet extraction method. After extraction the samples were reduced to a few ml using a rotary evaporator and further made up to about six ml with hexane into tared glass tubes. Hexane extracts were evaporated with a gentle stream of nitrogen gas until dry and the remaining total lipids were determined gravimetrically. The extracts were immediately redissolved into approximately 3 ml of hexane, the same amount of concentrated sulphuric acid was added to decompose and remove the lipids in the extract, and the samples were shaken manually for one minute. Two phases were allowed to separate overnight in warm and the solvent extract was transferred to glass tubes. This procedure was repeated twice (muscle, gonad and carcass samples) or three times (liver and intestine samples) for the remainding lipid deposit. The hexane extracts were then reduced to a few ml under a gentle stream of nitrogen gas. For a necessary additional clean-up, the residue was then introduced to a standardized Florisil-column (Florisil PR 60-100 mesh, Fluka Chemie AG, Buchs, Switzerland), which was activated at 160 °C and deactivated with 1.25% of water. The column was flushed with hexane and, prior to analyses, the residue in the second fraction was again reduced to 0.5 ml under a gentle stream of nitrogen gas. The PCB stock solution was dissolved into a few ml of hexane, PCB-30 was added as an internal standard, and the samples were purified with concentrated sulphuric acid.

The analysis of the PCBs was completed on a Nordion Micromat HRGC 412 gas chromatograph equipped with two 25 m quartz capillary columns (NB-1701 and NB-54, 0.25 mm, 0.25 μn) and with two Ni-63 EC detectors using helium as the carrier gas. The injection was splitless at 250 °C, the analysis programmed from 100 °C to 250 °C at 4 °C per min where it was held for 15 min. The compounds were identified and quantified using electron capture detection, and the concentrations calculated using response factors determined from individual standard samples included at least twice in each sample lot. Quality assurance included duplicate samples when necessary and use of method blanks of pure solvents at least twice per sample lot. The results presented in this paper are not corrected with the recovery percentage, including the internal standard (recovery percentage of internal standard was > 90%).

Calculations and statistics

Accumulation efficiency (AE,%), the amount of compound measured relative to that given, was calculated in total as the sum of accumulation efficiencies for each organ or tissue in relation to the total amount given to the fish. Relative accumulation efficiencies were calculated as proportions of each PCB in each organ or tissue sample from the total in all the samples. The total PCBs in whole fish were calculated as the sum of individual organ or tissue sample total concentrations multiplied by the sum of individual organ or tissue sample weights. Muscle sample and carcass PCB concentrations were multiplied by fish total weights to obtain the predicted whole fish values. Comparisons between predicted and observed PCB totals were made by regression analysis. Whole fish to muscle tissue and whole fish to carcass ratios were also calculated.

Paired-samples *t*-tests were performed in SPSS® 11.0 for Windows (Statistical Product Service Solutions Inc., Chicago, IL) to compare differences between tissue lipid contents in PCB-exposed and control-treated fish. Tissue lipid content comparisons between tissues were done with one-way analysis of variance (ANOVA) in SPSS. PCB concentration comparisons between the tissues, PCBs and time were performed with three-way ANOVA in SPSS. Two-way ANOVA for arcsine of the square-root transformed relative and total accumulation efficiency relations was used to compare the treatments. In the case of multiple comparisons, Tamhane's T2 post hoc test in SPSS was used with the analysis of variances. Statistical significance was set to the level of $p < 0.05$.

Results

Tissue lipid contents

During the exposure experiment, the lipid contents in all the tissues remained unchanged over

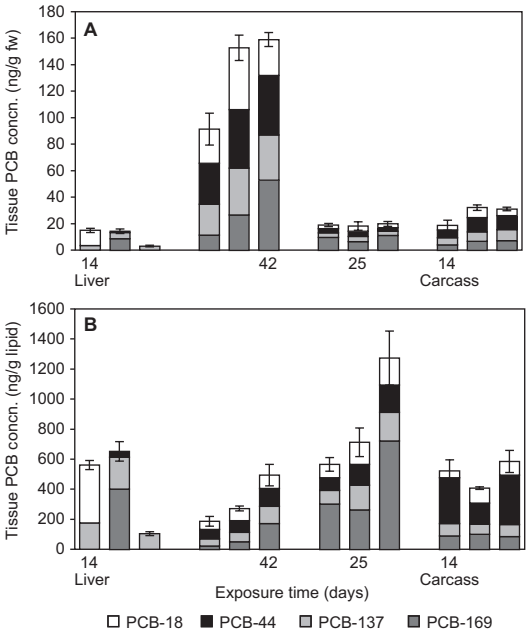

Fig. 1. PCB tissue concentrations (**A**) on a fresh weight (ng g^{-1} fw), and (**B**) on a lipid weight (ng g^{-1} lipid) basis after 14, 25 and 42 days of exposures. Vertical lines represent SE of sum of all PCB tissue concentrations.

time (ANOVA: $0.079 < p < 0.648$). The tissue lipid contents of the PCB-exposed and the control Baltic salmon were also similar (*t*-test: $0.597 < p < 0.970$). Half of the intestine fresh weight (mean = 49.7%, SD = 16.2%) of both the PCB-exposed and control-treated salmon consisted of tissue lipids, considerably more than in any other tissue (ANOVA: $p < 0.001$). Significant differences were also observed in carcass lipid content when compared with that in all the other tissues (ANOVA: $p \leq 0.001$). Liver and muscle sample contained less than 3% tissue lipids in their fresh weight (mean = 2.8%, SD = 0.6% and mean = 2.5%, SD = 1.0%, respectively), and carcass contained more than twice those amounts (mean = 6.4%, SD = 2.2%) with significant differences between them (ANOVA: $p = 0.001$ and $p < 0.001$, respectively). The lipid content of fish gonads was low with high individual variability (mean = 1.9%, SD = 1.3%) and equalled the liver and muscle lipid contents (ANOVA: $p = 0.062$ and $p = 0.450$, respectively). The low lipid content of fish gonads and the relatively high variation in the measurements indicated that while some individuals probably were at the early onset of their maturation process, most of the fish were still sexually immature.

PCB distribution in the body tissues

On a fresh weight basis, all the four PCB congeners had the highest concentrations in lipid-rich intestine (Fig. 1, ANOVA: $p < 0.001$). Normalization to tissue lipid contents did not eliminate all the variation in PCB uptake within the various tissue targets over time (ANOVA: $p < 0.001$). Within-individual variation between different tissues, however, was diminished. Higher lipid-based concentrations in intestine and muscle on day 42 as compared with those on either day 14 or day 25 were observed for all the four PCB congeners (ANOVA: $0.007 < p < 0.045$).

On a fresh weight basis, PCB-18 was the sole congener whose concentrations increased from day 14 to day 25 and then increased to day 42. This pattern of PCB-18 was similar in all tissues except liver where the concentration was the highest at day 14. It was rather quickly removed from the liver with no detection at day 25 and day 42. Fresh-weight-based concentrations of all the PCBs in carcass increased from day 14 to day 42 (ANOVA: $p = 0.010$). In intestine and muscle, the concentrations of coplanar PCB-169 continued to increase from day 25 to day 42 when the compound was no longer being given to the fish. None of the PCBs were detected in the gonads of the juvenile Baltic salmon.

Relative accumulation (%, total amount of the specific congener in the specific tissue) into the different tissues reveal the significance of carcass in total PCB accumulation in Baltic salmon (Fig. 2). While the relative accumulation efficiency of the salmon intestine was 21.3% (SD = 10.1%), as much as 78.0% (SD = 12.7%) of the PCBs was assimilated in salmon carcass. Total PCB accumulation efficiency of PCB given to the fish calculated as an average of all sampling times was 71.1% (SD = 31.1%).

Accumulation efficiencies

The accumulation efficiency of PCBs 18 and 44 was highest after the four consecutive dosages

given on day 25 (mean = 89.2%, SD = 26.6% and mean = 97.6%, SD = 35.2%, respectively), whereas with PCB-169 the highest accumulation efficiencies were observed on day 42 (mean = 67.2%, SD = 11.2%). The accumulation efficiencies of PCB-169 increased over time (mean = 52.2%, SD = 31.2%; mean = 60.5%, SD = 25.9%; mean = 67.2%, SD = 11.2%, on sampling days, respectively). PCB-137 seemed to attain a steady state in fish as accumulation efficiencies of 66.8% (SD = 63.0%), 64.6% (SD = 28.0%) and 64.5% (SD = 8.5%) were measured on days 14, 25 and 42, respectively. As an average of all sampling days, we found that the accumulation efficiencies of the four PCB congeners occurred in the following order: PCB-44 > PCB-18 ≈ PCB-137 > PCB-169.

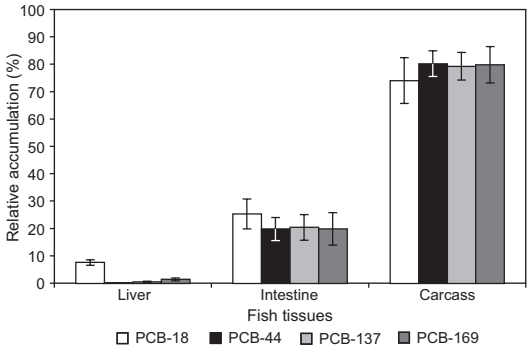

Fig. 2. Relative accumulation (total amount of the specific congener in the specific tissue ± SD) in fish tissues (liver, intestine and carcass) for the four PCB congeners studied as an average of 14, 25 and 42 days of exposures.

Estimation of whole fish to muscle concentration ratio

Whole-fish/muscle-PCB ratio was high (mean = 2.05, SD = 1.06 for total sum of all 4 congeners) and the totals predicted from the muscle samples had very high variation between individuals. Whole-fish/carcass-PCB ratio was 1.20 (SD = 0.17) for the sum of all 4 congeners.

Discussion

Quality control

Fish were fed once a day in excess but they did not grow during the treatments. During the experimental period, the mass change in the PCB-exposed and control fish was < 1% and 1%–3%, respectively. The length of both the exposed (mean = 0.81%) and the control (mean = 2.27%) fish increased. Significant growth retardation was shown earlier for juvenile coho salmon (*Oncorhynchus kisutch*) in a 165 day PCB-exposure for congeners 77, 153 and 155 (Gruger *et al*. 1975). This indicates some stress undergone by the PCB-exposed fish. Nonetheless, no measurable lipid mobilisation or loss took place in the fish during the treatments.

Fish faeces as well as the food pellets not eaten by the fish were cleaned daily from the pools with a continuous water flow-through system. Thus, taking into account also the highly hydrophopic nature of the PCBs administered to salmon in this study, any reuptake of PCBs either from the waterphase or from the fish faeces should have been negligible. This was verified by an equilibrium dialysis technique with semi-permeable dialysis membranes and by faeces PCB measurements in our earlier studies with the same PCBs administered orally to burbot (*Lota lota*) Pääkkönen *et al*. (2005).

The PCB congeners were administered to salmon by oral gavage exposure. It is widely regonized that type of food, dosing method and carrier influence the accumulation of OCs to organisms (Burreau *et al*. 1997, Andersson *et al*. 2001). Oral gavage exposure is a less "natural" dosing method than many others but it makes it possible to give an accurate dose to each individual in the school of fish. The whole fish accumulation efficiencies in this short-term exposure study were similar to those observed earlier which indicated that the dosing method was workable.

PCB accumulation and distribution in the body tissues

Because PCBs are highly soluble in lipids, they should accumulate in tissues and organs rich in lipids following their K_{ow}-dependent release rates. Bioaccumulation of PCBs is a physico-

chemical process and at first, the accumulation is predominantly governed by equilibrium partitioning of the compounds between the organism and the fish liver oil used as a carrier. The accumulation of PCBs from fish liver oil containing PCB takes place as partition across the membrane lining the gut into the bloodstream. Prior to this, the surface adsorption properties, i.e. the strength of adsorption of the PCBs on the membrane, will affect accumulation (Sawhney 1986). After passage through the walls of gastrointestinal tract and absorbtion, the PCBs are then partitioned into the blood and then from blood to tissues (Shaw & Conell 1986) followed by an equilibrium partitioning between different tissues and organs inside a fish. The blood flow in different tissues drives the initial distribution of an administered dose but tissue lipid content determines where the dose will end up.

In our experiment, all the four PCB congeners accumulated more into the lipid-rich intestine of Baltic salmon than into other tissues. In addition to differences in the total PCB concentrations, there were differences in concentrations of individual PCB congeners among different tissues. The congener-specific accumulation was mostly related to the K_{ow}-dependent uptake processes (differences in absorption across the gastrointestinal epithelium, accumulation, retention, biotransformation). The coplanar PCB-169 with the highest K_{ow} value was taken up least within the 14 and 25 days exposures. PCB-169 seemed to distribute into tissues and organs of fish more slowly than its *ortho*-substituted congeners. In intestine, the concentration of PCB-169 continued to increase from day 25 to day 42 when the compound was no longer being given to the fish. At the same time, the concentrations of PCB-18 decreased while those of PCBs 44 and 137 remained unchanged.

The tissue-specific accumulation differs remarkably between different fish species. For example, comparison between our earlier studies with burbot (*Lota lota*, Pääkkönen *et al*. 2005) and salmon indicated that while the main portion of the total PCBs in Baltic salmon was accumulated in the fish carcass (which is mostly muscle), liver with practically an equal contribution (mean = 71.6%, SD = 6.8%) was the main target of the PCBs in burbot. Secondly, whereas intestine accounted for as much as 22% and liver less than 1% of the total accumulation in salmon, burbot carcass accounted for 12.5% (SD = 6.2%) and intestine only 5.0% (SD = 2.7%) of the total accumulated PCB in burbot during the short-term experiment.

Accumulation efficiencies

Total accumulation efficiency of the total PCBs in Baltic salmon averaged 71% and varied from 38% to 88%, whereas for the *ortho*-substituted congeners 18, 44 and 137 it averaged 75% and for PCB-169 61%. These efficiencies are similar to those observed earlier. Adult rainbow trout, in which gelatin capsules spiked with a mixture of 31 *di*-chloro- to *deca*-chlorobiphenyl congeners were inserted into the fish stomach, were shown to accumulate 75% of the PCBs with a variation of 62% to 85% (Niimi and Oliver 1983). Wong *et al*. (2002) measured accumulation efficiencies of 50%–80% for *ortho*-substituted PCBs 95 and 136 in 40 days dietary exposure (feed) and Isosaari *et al*. (2004) observed a mean efficiency of 78% for 25 *ortho*-substituted congeners in adult Atlantic salmon in dietary exposure (feed) of 210 days. The total retention efficiency of the total PCBs for burbot (*Lota lota*, Pääkkönen *et al*. 2005) was 81.7% (SD = 28.1%) in 28 days with the same congeners as in this study after dietary exposure by natural fish prey. Further, the short-term uptake efficiency of five PCB congeners administered to pike (*Esox lucius*) in natural food varied from 42% to 82% (Burreau *et al*. 1997).

Our results revealed some differences between different congeners in the total accumulation efficiencies in Baltic Sea salmon. These were highest for the *tetra*-chlorinated PCB-44, although quite variable, and lowest for *hexa*-chlorinated PCBs 137 and 169. Adult Atlantic salmon also show the highest accumulation efficiencies for *tetra*-chlorinated PCBs (Isosaari *et al*. 2004). The total accumulation efficiencies of PCB-169 increased from 14 days to 42 days from 52% up to 67% indicating that this coplanar congener most likely takes longer than its *ortho*-substituted congeners to reach an equilibrium distribution between organs and tissues in fish. This is also supported by the fact that

an 84% accumulation efficiency for PCB-169 was attained in 30 weeks of exposure in Atlantic salmon (Isosaari *et al*. 2004).

It seems also that PCB-169, with no unsubstituted carbon atoms that are adjacent on the biphenyl ring, is resistant to metabolism as these positions are also preferentially oxidized by the cytochrome P450 system (Van den Berg *et al*. 1998). PCB-137 seemed to attain an apparent steady state in fish as similar accumulation was measured at the consecutive sampling times. The PCBs 18 and 44 with no chlorine substitution in the *para* positions were eliminated by Baltic salmon juveniles to some extent, because the continuing follow-up with no additional PCB exposure from day 25 to day 42 resulted in lower measurable compound total accumulations than those on day 25. Dimished accumulation efficiencies of these two PCBs was not caused by a growth dilution effect, because the fish growth rate during the short-term experiments was negligible.

In summary, fish are known to metabolize most PCBs with a retention propensity for the higher and selective elimination of the lower chlorinated PCBs (Niimi and Oliver 1983). On the basis of earlier investigations introduced above and our study it appears that the average dietary accumulation efficiencies of PCBs in fish rarely exceed 78%–84% and are not influenced by the degree of chlorination but rather by their substitution pattern.

Estimation of the whole-fish to muscle concentration ratio

The average whole-fish to muscle (no skin) PCB-concentration ratio (2.05) obtained in this study was slightly higher than the average whole-fish to skin-on fillet PCB concentration ratios observed earlier for coho salmon (1.70) and rainbow trout (1.47) (Amrhein *et al*. 1999). Based on the high individual variation in the ratios observed in these two studies, it seems that the estimation of the whole fish PCB concentrations simply from muscle or fillet data is unreliable. We can speculate that in our study, the high individual variation is due to the short exposure duration: fish never came to steady-state internally and the rate to reach the equilibrium changes between individuals. However, this is always true in the sampling of the wild fish because the exposure to PCBs fluctuates according to the changes in feeding intensity, prey selection, starving periods etc.

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