PCDD/F, PCB, DIOXIN-LIKE PCB, AND PBDE IN FISH OIL USED AS DIETARY SUPPLEMENT IN SWITZERLAND

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Introduction

Fish oil is a valuable by-product of fish meal production and is widely used as food and feed supplement. It is rich in polyunsaturated fatty acids (PUFA) which are essential lipids that cannot be synthesized by mammalians. Therefore, PUFA must be ingested via the diet. Levels of one important category of polyunsaturated fatty acids, the so called ω-3 fatty acids like eicosapentaenoic acid (EPA, 20 carbons) and docosahexaneoic acid (DHA, 22 carbons), are especially high in fish oil. Up to 25% or more of the total fat content of wild and farmed salmon is in the form of ω-3 fatty acids.[1] ω-3 fatty acids have therapeutic benefits in the prevention and treatment of cardiovascular, immune, and arthritic diseases. They lower blood pressure and pulse, reduce inflammation, and improve vascular and platelet function.[2] Further possible effects of ω-3 fatty acids on inflammatory diseases, insulin resistance, diabetes and obesity, cancers, mental health and neurodegenerative diseases were reported.[3,4] However, fish oil can be a major source of lipophilic persistent organic pollutants (POPs) such as polychlorinated dibenzo-*p*-dioxins and dibenzofurans (PCDD/F), polychlorinated biphenyls (PCB), and polybrominated diphenyl ethers (PBDE). Via the food chain, these and other highly lipophilic compounds accumulate in the lipid compartment of fish. Fish oil collected from species caught in contaminated waters or from farmed fish fed with contaminated feed may contain markedly higher amounts of POPs than fish originating from less polluted sites. The daily intake of fish oil capsules as dietary supplement and source of PUFA might result in an undesirable uptake of POPs. Therefore, in a first survey six fish oil containing products available on the Swiss market were analyzed for their content of indicator PCB (PCB-28, 52, 101, 138, 153, and 180), dioxin-like PCB (DL-PCB, PCB-77, 81, 105, 114, 118, 123, 126, 156, 157, 167, 169, and 189), the 17 2,3,7,8-substituted PCDD and PCDF, as well as PBDE (BDE-28, 47, 99, 100, 153, 154, 183, and 209).

Materials and Methods

Sample Description. Fish oil capsules from six different suppliers were bought in a pharmacy (n = 5) and in a supermarket (n = 1). Detailed descriptions of the different brands are given in Table 1. At least 19 different brands of fish oil capsules are available on the Swiss market; therefore this first survey does only represent about 30% of all brands available.[5]

Sample Preparation and Analysis. For each sample, between 30 and 40 fish oil capsules were pierced with a syringe needle and the content was squeezed out into a pre-washed glass vial. About 15 g fish oil was used for the clean-up. The samples were spiked with $^{13}C_{12}$-labeled PCDD/F, PCB, and PBDE surrogates of the above mentioned congeners. After addition of 25 mL *n*-hexane the mixture was cleaned-up as described previously.[6] In brief, clean-up of the samples included chromatography with *n*-hexane on a large scale multilayer silica gel column followed by chromatography on carbon AX-21. The PBDE and di-*ortho*-substituted PCB were collected as fraction passing the carbon AX-21 column whereas the PCDD/F and the mono- and non-*ortho*-substituted PCB were collected after backflush of the carbon AX-21 column with toluene. Analyses of PCDD/F, PCB, and PBDE were carried out using GC/HRMS on a 30 m × 0.25 mm RTX-5 Sil-MS capillary column (film thickness 0.10 µm) at a mass resolution of 8000.

Table 1: Sample description and recommended daily doses.

Sample	Origin of fish oil	Fish oil/capsule [mg]	EPA/capsule [mg]	DHA/capsule [mg]	Recommended daily dose [mg]
S1	Salmon	500	90 (18%)	60 (12%)	1500
S2	Pacific fish with lipid extract from green-lipped mussel (*perna canaliculus*, New-Zealand)	500	90 (18%)	60 (12%)	1500
S3	Pacific fish, mainly offshore fish from the coast of Peru	740	240 (32%)	160 (22%)	740
S4	Pacific and south Atlantic fish	750	135 (18%)	90 (12%)	2250
S5	not specified	500	90 (18%)	60 (12%)	1500
S6	Pacific fish	500	90 (18%)	60 (12%)	1000

Results and Discussion

Detailed results are summarized in Table 2. Whereas PCB and PBDE could be detected in all fish oil samples only few of the 17 2,3,7,8-substituted PCDD/F congeners could be measured above their limits of detection (LODs). From tetra- to octaCDD/F the LODs varied between 0.1 and 0.4 pg/g lipid weight (lw). The WHO-TEQ_{max} in Table 2 was calculated including the entire LODs in the case of non-detected congeners. For calculation of WHO-TEQ_{min} only congeners detected above their LODs were considered. In most of the samples, only 1,2,3,4,6,7,8-HpCDD, OCDD, 2,3,7,8-TCDF, 1,2,3,4,6,7,8-HpCDF, and OCDF were detectable, and the concentrations were close to the corresponding LODs. Even when considering the WHO-TEQ_{max} all the samples were clearly below the Swiss tolerance value of 2 pg/g, which is based on the European Council regulation (EC) No 2375/2001 from 2001.[7] The WHO-TEQ values of the DL-PCB were between 0.038 and 2.0 pg/g lw. PCB-126 contributed up to 83% of the WHO-TEQ (average 74% ± 7.5) followed by PCB-118 (9.4% ± 3.4) and PCB-156 (6.9 ± 1.4). Similar compositions were reported for fish from Swiss plateau lakes.[6] In all samples, the WHO-TEQ of DL-PCB clearly surpassed the PCDD/F but were still below the maximum level of 10 pg/g established in the new regulation (EC) No 199/2006.[8] The sum of the six indicator PCB (PCB-28, 52, 101, 138, 153 and 180) was between 0.23 and 17 ng/g with patterns dominated by PCB-153 followed by PCB-138 and PCB-180.

In a study on persistent halogenated contaminants in fish oil Jacobs *et al*. reported total PCB concentrations (sum of seven indicator PCB, including PCB-118) of 92 to 202 ng/g in cod liver and distinctly lower levels of 19 to 49 ng/g in whole body fish oils.[9] The PCB levels in the present study are in good accordance with the results found by Jacobs and coworkers: Four of the samples (S1, S2, S4, and S6) exhibited total PCB levels between 13 and 17 ng/g, sample S3 was clearly less contaminated (2.1 ng/g), and the PCB concentration in sample S5 (0.23 ng/g) was close to the method blank level. On the other hand, PCB concentrations in fish from Swiss plateau lakes were in the range of 210 to 1300 ng/g lipid, and even in fish from remote Swiss alpine lakes PCB concentrations were in a similar range (180 to 1300 ng/g lw).[6,10] Compared to these fairly high PCB levels, the concentrations in fish oil which mainly originate from Pacific offshore fish were by one to two orders of magnitude lower.

The PBDE levels (sum of BDE-28, 47, 99, 100, 153, 154 and 183) in fish oil were between 0.069 and 3.8 ng/g. At the upper end of the range, sample S1 exhibited by far the highest PBDE concentration. The levels in the remaining samples were between 0.069 and 0.78 ng/g. In the above mentioned study, Jacobs *et al*. reported PBDE levels in cod liver oil of 15 to 22 ng/g and for whole body fish oil levels up to 2.7 ng/g.[9] Obviously, the results of the present work are in good agreement with this study. Elevated PBDE concentrations of 9 to 22 ng/g lw in farmed salmon from different regions were reported by Hites and Carpenter, meanwhile wild pacific salmon had clearly lower levels.[1,11] In wild and farmed fish from Switzerland PBDE levels of 36 to 165 ng/g lw and 12 to 24 ng/g lw, respectively, were observed.[12] Compared to these levels, PBDE concentrations in the investigated fish oil samples are by one to two orders of magnitude lower.

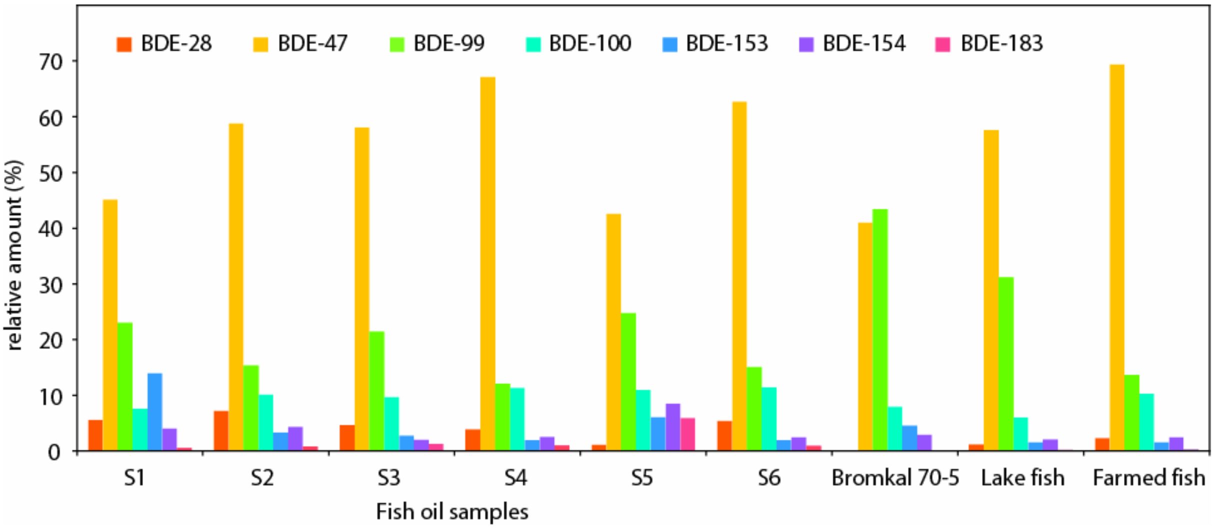
In Figure 1, normalized PBDE congener patterns of the six samples are displayed together with the patterns of the technical pentaBDE product Bromkal 70-5, wild fish from Swiss plateau lakes, and farmed fish from

Switzerland. With exception of sample S1, the fish oil patterns dominated by BDE-47, BDE-99, and BDE-100 are very similar. Sample S1 exhibits the highest PBDE level (3.8 ng/g) and its pattern is characterized by a relatively high contribution of congener BDE-153. The PBDE congener patterns of the fish oil samples are similar to the average pattern of whitefish from Swiss plateau lakes (n = 8) and farmed fish from Switzerland (n = 4) as well as to congener patterns of salmon reported by Hites and coworkers.[11,12] Compared to the technical pentaBDE product Bromkal 70-5 the fish oil samples show significantly higher ratios BDE-47/BDE-99 which might be due to metabolic degradation of BDE-99 or reduced absorption of this congener. Decabromodiphenyl ether (BDE-209) was determined in all samples but all the levels were close to or even below the method blank level. Therefore, no clear statement concerning this congener can be given at the moment and the results in Table 2 are given in parentheses.

Table 2: Total PCB and PBDE concentrations in fish oil (ng/g and pg WHO-TEQ/g, respectively).

Sample	Total PCB	Total PBDE	BDE-209	DL-PCB WHO-TEQ	PCDD/F WHO-TEQ_{max}	PCDD/F WHO-TEQ_{min}
S1	16	3.8	(0.48)	1.3	0.54	0.058
S2	13	0.72	(0.67)	1.1	0.83	0.45
S3	2.1	0.23	(0.27)	0.50	0.59	0.020
S4	17	0.78	(1.5)	2.0	0.72	0.51
S5	0.23	0.069	(0.12)	0.038	0.32	0.067
S6	13	0.68	(0.88)	1.2	0.81	0.47
Method blank	0.084	0.048	(0.57)	0.18	n.d.	n.d.

Figure 1: Normalized PBDE patterns of six fish oil samples (S1 to S6), Bromkal 70-5 DE, lake fish, and farmed fish from Switzerland.



Conclusions

In all investigated fish oil samples concentrations of PCDD/F, PCB, DL-PCB, and PBDE were fairly low. The concentrations were similar to concentrations reported for fish from the relatively uncontaminated Pacific. This is consistent with the fact that most of the analyzed products were indeed extracts from offshore fish from the Pacific.

The supply of PUFA via intake of fish oil capsules is an alternative to the consumption of fish. Therefore, the health benefits of dietary supplements such as fish oil have to be counterbalanced against possible negative longterm effects caused by possible accumulation of POPs. In Switzerland, the average daily fish consumption by adults is 22 g/day.[13] Based on lipid contents in fresh fish of 1 to 5% this corresponds to the intake of 0.2 to 1 g of fish lipids. This amount is similar to suppliers' recommendations of daily doses of 0.75 to 2.25 g of fish oil. As lipid weight based levels of PCDD/F, PCB, DL-PCB and PBDE in fish can exceed the respective concentrations in fish oil by up to two orders of magnitude, intake of fish oil capsules does not significantly contribute to the average dietary intake of these persistent contaminants.

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