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Polychlorinated Biphenyl Congener Distributions in Winter Flounder As Related to Gender, Spawning Site, and Congener Metabolism†

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Concentrations of 17 polychlorinated biphenyl (PCB) congeners were measured in liver of gonadally mature winter flounder collected from Fox Island and Gaspee Point, RI, and from New Bedford Harbor, MA. These locations represent spawning sites with different degrees and sources of PCB contamination. The data demonstrate that the PCB concentrations and patterns in these fish reflect those of their spawning grounds, indicating that winter flounder caught in clean offshore waters could have substantial amounts of tissue PCB, that the content of PCB congeners in winter flounder liver is influenced little by sex or reproductive condition, and that flounder selectively metabolize PCB congeners with adjacent meta,para-unsubstituted carbon atoms. Based on congener concentrations and 2,3,7,8-tetrachlorodibenzo-*p*-dioxin equivalency factors, the non-ortho congeners, 77, 126, and 169, contributed more to the potential (mammalian) toxicity of PCB in flounder than more abundant, but less toxic, non-coplanar congeners.

Introduction

Polychlorinated biphenyls (PCBs) are widespread contaminants in the environment, where over 100 individual congeners have been identified (*1*). Relative concentrations of PCB congeners can provide information on their environmental source, metabolism, and toxic potential in tissues (*2–5*). A better understanding of the factors and processes influencing these distributions is critical to such interpretations.

Congener distributions in some PCB-exposed fishes suggest selective uptake and/or metabolism of specific PCB congeners (*2, 6*). In general, PCB congener bioaccumulation is proportional to lipid solubility (log K_{ow}) and, hence, chlorine content (*7, 8*). However, selective metabolism of congeners with adjacent meta,para-unsubstituted carbon atoms can facilitate excretion and lead to residue profiles which deviate from the apparent source (*9, 10*). Fish metabolize PCBs more slowly than do mammals (*11–13*), and thus the role of metabolism in determining congener distribution in fish is less certain. Since bivalves, which are winter flounder prey organisms, have little capacity to metabolize PCBs (*14, 15*), we compared PCB profiles in winter flounder with those of bivalves from the same site as one indication of whether selective congener metabolism may be occurring in flounder.

Sex differences in PCB disposition have been observed in gonadally mature fish. In some species, PCBs appear to partition into the eggs during gametogenesis (*16*) with whole body elimination rates enhanced during spawning (*17*); an equivalent process is reduced or does not occur in males of those species (*16*). Sex differences in PCB disposition might also be related to the activity of cytochromes P4501A (*18, 19*), enzymes which selectively metabolize some PCB congeners in mammals (*6, 20*) and which are also influenced by female sex steroids in fish (*19, 21*).

Few studies have addressed how strongly or consistently such factors influence congener distribution in migratory fish or in fish from highly contaminated sites. In this study, we measured 17 PCB congeners in the livers of spawning winter flounder from Fox Island and Gaspee Point, RI, and from New Bedford Harbor, MA, a site highly contaminated with PCBs (*22*), to evaluate whether gender, reproductive status, and metabolism affect PCB disposition in highly contaminated fish.

A key element of this study is that winter flounder from all three sites mix in one offshore population during summer, returning in winter to spawn as discrete populations (*23, 24*). Since the three spawning locations have different degrees and sources of PCB contamination (*25–27*), comparison of congener distributions in flounder and sediments could indicate whether these spawning sites are a major source of PCBs to these winter flounder. To this end, we compared PCB profiles in these flounder populations to previously published sediment PCB profiles from these sites.

We chose liver tissue rather than edible flesh for PCB analysis for two reasons. Liver tissue contains high concentrations of cytochrome P450 monooxygenases, enzymes which are involved in the metabolism of polynuclear aromatic hydrocarbons and polychlorinated biphenyls (*6, 20*). Further, liver lipid content is high compared to that of edible flesh (*10*), facilitating liver bioconcentration of highly lipophilic xenobiotics, such as PCBs. These two aspects (abundance of metabolic enzymes and high lipid content) make liver the most useful tissue for examining PCB congener distributions and for determining if selective congener metabolism occurs in these fish.

The distribution of individual PCB congeners in fish tissue has implications for both human and animal health. Congeners with chlorine substitution patterns conferring a coplanar configuration are generally thought to be more toxic than non-coplanar congeners (*28*). To facilitate the comparison of relative toxicity, individual PCB congener concentrations are commonly converted into 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) toxic equivalents using 'TCDD equivalency factors' (TEFs) derived from

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dose–response experiments in mammals (*29, 30*). We used TEFs to estimate the relative contribution of selected congeners to the potential toxicity (to mammals) of PCBs found in winter flounder tissue.

Materials and Methods

Chemicals. Congeners used as calibration standards were obtained from Ultra Scientific (North Kingstown, RI) and from the National Research Council of Canada. All solvents were high-purity, gas-chromatography grade from Burdick and Jackson Laboratories, Inc., Muskegon, MI.

Study Sites. Fox Island and Gaspee Point, RI, and New Bedford Harbor, MA [described in detail elsewhere (21)], are distinct winter flounder spawning grounds with different degrees of PCB contamination (Fox Island < Gaspee Point ≪ New Bedford Harbor).

Animals. Winter flounder (*Pleuronectes americanus*) were collected by fyke net as described previously (*31*) during the spring spawning season of 1988. Animals were gravid females (ovaries filled with eggs), spent females (ovary recently depleted of eggs but not yet regressed), and ripe males (actively milting) and ranged in size from 111 to 752 g wet wt, and from 19 to 43 cm total length. Fish from each site were of a similar size distribution. Bivalves (*Mya arenaria* and *Mercenaria mercenaria*) were collected from Tin Can Island inside the hurricane barrier in the New Bedford Harbor area via scuba diving. Bivalves were returned to the laboratory following field collection and held in flowing water overnight to purge gut contents. Bivalves were shucked and the tissue was frozen in solvent-rinsed, glass jars. Pooled samples were homogenized using a Virtis tissue homogenizer and were refrozen before analysis.

PCB Analysis. Individual flounder liver samples were extracted and analyzed, in triplicate, for total PCB (measured as Aroclor 1242 + 1254) and PCB congeners using methods previously described (*32*). All congeners were not completely resolved using these methods, and therefore contributions from unresolved compounds may occur for some congeners. Detection limits ranged from 10 to 50 ng/g of dry liver for A1242 + A1254 and from 0.35 to 3.5 ng/g for individual congeners. Prior to extraction, the samples were spiked with congener 100 (*33*) and octachloronaphthalene, compounds used as internal standards for the non-coplanar and coplanar congeners, respectively. Data were corrected for recovery. When the concentration (ng/g of dry liver) of a congener was below the limit of detection, one-half of the detection limit was used rather than zero for statistical analysis. Values were log-transformed prior to statistical analysis.

Bivalve samples were extracted using dichloromethane by one of two methods. Samples were combined with approximately 8–10 vol of anhydrous sodium sulfate. They were batch-extracted by using the Virtis homogenizer or more frequently by grinding the tissue sodium sulfate mixture with a mortar and pestle and using a column extraction technique, similar to that described by Ribick et al. (*34*). Prior to extraction, tissue was spiked (via injection) with a known quantity of PCB congeners 29 and 143 for use as procedural recovery standards. Extracts were reduced using rotary evaporation, applied to a fully activated 5-g Florisil column, and eluted with approximately 60 mL of hexane. After fractionation, extracts were reduced using rotary evaporation, transferred to a sample vial, blown just to dryness with nitrogen, and supplemented with an aliquot of hexane containing octachloronaphthalene as an internal standard. Samples were analyzed for chlorobiphenyl congeners by gas chromatography with electron capture detection using a Carlo Erba Model 4160 gas chromatograph equipped with a 30-m SE-52 capillary column (0.32 mm i.d.). Sediment PCB data were obtained from the literature (*25–27*).

Table 1. Characteristics of 17 PCB Congeners Measured in Winter Flounder

congener no.	ring 1	ring 2	inducer type[a]	comments
		Non-Coplanar		
52	25	25	2B	
47	24	24	2B	
101	25	245	2B	
105	34	234	mixed	*b, c*
118	34	245	mixed	*c*
151	25	2356		
153	245	245	2B	*d, e*
138	234	245	mixed	*c, e*
128	234	234	mixed	*c*
180	245	2 345	2B	
195	234	23 456		
194	2 345	2 345	2B	
206	2 345	23 456		
209	23 456	23 456		
		Coplanar		
77	34	34	1A	*b, f*
126	34	345	1A	
169	345	345	1A	*g*

[a] CYP1A (1A), CYP2B (2B), or mixed-type (1A and 2B) inducers (*20, 64*). [b] Readily metabolized in mammals (*29*). [c] Ineffective as an inducer of CYP1A in the fish scup (*36*). [d] Very low biodegradation rate in flounder (*16*). [e] Major component of technical PCB formulations (*62*). [f] 10^{-7} M strongly inhibits teleost CYP1A catalytic activity *in vitro* (*36*). [g] 10^{-4} M did not inhibit teleost CYP1A catalytic activity *in vitro* (*63*).

PCB Congener Selection. Fourteen non-coplanar congeners were measured in all samples, and three coplanar congeners were measured additionally in winter flounder (Table 1). The non-coplanar congeners were chosen based on relative ease of quantitation, availability of standards, and range of chlorination levels. These 17 congeners were of particular interest due to their environmental occurrence (congeners 52, 101, 105, 118, 138, 153, 180, 206) (*35*), potency as inducers of mammalian and teleost P4501A (CYP1A) (congeners 77, 126, 169) (*28, 36*) or mammalian P4502B (CYP2B) (congeners 52, 47, 101, 153, 180, 194) (*28*), evidence of metabolism by mammals (congeners 52, 77, 101, 105, 151) (*6*) or fish (congener 52) (*11*), or evidence of relatively high toxic potential in mammals (congener 128, 138) (*37*). Congeners 52, 77, 101, 105, 118, 126, 128, 138, 153, 169, and 180 have also been identified as relevant to human studies (*35*).

Potential Toxicity. Since there is a linear relationship between the toxicity of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) and its CYP1A induction potency in some systems (*29*), the toxicity of other compounds has been estimated relative to TCDD from their potency at CYP1A induction in mammals or mammalian cells. These derived 'TCDD equivalency factors' (TEFs) have been evaluated for PCB congeners in mammals (*38*) and for congeners 77 and 126 in fish (*39, 40*). TEFs were multiplied by congener concentrations in flounder tissue to derive congener-specific 'TCDD equivalent concentrations' (TECs).

Statistical Treatment of the Data. Data were analyzed using one- and two-way analysis of variance (ANOVA, Statview, Abacus Concepts Inc., 1988). For analysis of congener distributions (congener content

Table 2. Total PCB (A1242 + A1254) Concentrations in Winter Flounder Liver (n = 6) (Presented as μg/g of dry liver)[a]

sample type	Fox Island	Gaspee Point	New Bedford
gravid females	3.1 ± 3.6 (0.12 ± 0.01)	3.9 ± 1.4 (0.12 ± 0.02)	333 ± 387[b,e,f] (0.12 ± 0.02)
spent females	1.6 ± 0.8 (0.10 ± 0.01)	4.0 ± 1.1[e] (0.14 ± 0.02)	132 ± 72[e,f] (0.12 ± 0.01)
ripe males	4.0 ± 2.7 (0.11 ± 0.03)	13.3 ± 10.0[c,d] (0.11 ± 0.02)	124 ± 131[e,f] (0.13 ± 0.03)
sediment	0.1	0.5	8.0

[a] Values in parentheses represent μg of total PCB/g of liver lipid. Sediment values from refs 25–27. [b] One individual had 1050 μg of PCB/g of dry liver; without this fish, mean = 190 ± 181 μg/g. Significantly different by one-way ANOVA ($p < 0.05$) from the following: [c] Gravid females. [d] Spent females. [e] Fox Is. [f] Gaspee Pt.

Table 3. PCB Congener Concentrations in Winter Flounder Liver (Sexes Combined)

congener	Fox Island	Gaspee Point	New Bedford
		Non-Coplanars	
52	4.4 ± 4.0[a]	6.5 ± 5.7	1 100 ± 2 860
47	6.9 ± 5.9	14.3 ± 11.2	2 530 ± 4 220[b,c]
101	29.5 ± 25.3	49.5 ± 47.1	2 450 ± 3 490[b,c]
105	38.1 ± 29.8	92.1 ± 75.5	2 200 ± 1 770
151	9.0 ± 8.1	19.5 ± 19.0	422 ± 593[b,c]
118	100 ± 78.2	239 ± 173	12 800 ± 15 700[b,c]
153	181 ± 153	441 ± 337	11 000 ± 13 100[b,c]
138	122 ± 103	286 ± 231	7 070 ± 7 490[b,c]
128	18.3 ± 18.8	47.3 ± 48.5	1 370 ± 1 490[b,c]
180	53.7 ± 64.1	141 ± 159	1 010 ± 1 210
195	1.1 ± 0.6	1.4 ± 1.7	31.7 ± 40.7
194	10.2 ± 12.1	23.2 ± 26.2	78.9 ± 100.3
206	12.2 ± 12.3	32.6 ± 37.0	32.7 ± 44.3
209	8.3 ± 9.0	20.5 ± 23.3	3.1 ± 7.0
		Coplanars	
77	2.0 ± 2.7	4.6 ± 4.2	391 ± 356
126	1.2 ± 1.8	1.8 ± 2.1	48.6 ± 33.6
169	0.4 ± 0.1	0.4 ± 0.2	3.7 ± 4.6

[a] ng/g of dry liver ± SD; n = 18. Significantly different ($p < 0.05$) from the following: [b] Fox Is. [c] Gaspee Pt.

normalized to the sum of the congeners), distributions were first log-transformed to remove skewness, and then multivariate ANOVA (MANOVA) was used to generate eigenvectors (SuperANOVA, Abacus Concepts Inc., 1990) for use in discriminant analysis (DataDesk, Odesta Corp., Northbrook, IL).

Results and Discussion

Differences among Flounder Populations. Winter flounder showed strong site differences in liver concentrations of total PCB and the concentration and distribution of PCB congeners. New Bedford fish had up to 85- and 100-fold higher concentrations of total PCB (per g dry wt), respectively, than Gaspee Point and Fox Island fish (Table 2). These differences reflect the 16- and 80-fold difference in sediment PCB concentrations at these sites (Table 2). Because the lipid content of flounder liver was similar for fish from all three sites (0.10–0.18 g of lipid/g of dry liver), expressing PCB content on a per lipid weight basis did not alter our findings (Table 2). The large standard deviations of the PCB data (Tables 2 and 3) reflect the fact that these are migratory fish of different sizes and therefore ages, which have been exposed to the PCB concentrations at these sites for different lengths of time.

The PCB congener composition at these sites further illustrates between-site differences. The PCB patterns in the Narragansett Bay flounder (Gaspee Point and Fox Island) indicate a mixture of Aroclors 1254 and 1260 which is similar to that of the sediments at these sites (*25, 26*). In contrast, New Bedford flounder livers contain 20–30% A1242 and 70–80% A1254, a composition similar to that of lower New Bedford Harbor sediments (~31% A1242 and 69% A1254; ref 27, station D).

PCB congener profiles even more strongly indicate that the source of PCBs to the Narragansett Bay flounder differs from that of the New Bedford population. Discriminant analysis of the log-transformed concentrations of 17 PCB congeners in liver separated the groups according to site of capture (Figure 1a, ANOVA $p < 0.05$). Discriminant axis 1 (DA1), which accounts for most of the variance in the data (eigenvalues equal 45.1 and 0.9 for DA1 and DA2, respectively), strongly separated the New Bedford population from those of Fox Island and Gaspee Point. The small distinction between between Fox Island and Gaspee Point flounder along the DA2 axis (Figure 1) likely reflects a slight difference in PCB congener distribution with distance from the Providence River, the main source of PCBs to Narragansett Bay (*25,26*). Discriminant analysis of log-transformed, congener distributions (congener concentration/sum of congeners) gave similar results (Figure 1b, ANOVA $p < 0.05$). Two-way ANOVA (site x reproductive status) on these distributions showed that congeners 52, 47, 101, 118, and 77 were in higher relative abundance and that congeners 151, 153, 138, 180, 195, 194, 206, 209, and 169 were in lower relative abundance in New Bedford flounder as compared to Fox Island and/or Gaspee Point fish (Figure 2). Only congeners 128, 105, and 126 showed no site-related differences. Eigenvector plots ('loading' plots) produced similar results (data not shown).

The PCB congener distributions and the Aroclor concentrations both indicate that the winter flounder in this study are accumulating a significant amount of PCBs during residence at their respective spawning grounds. Our results, together with those of other studies (*41, 42*) provide evidence that PCB congeners in fish tissue can be used to identify environmental sources of PCBs.

Sex Differences in PCB Concentration and Distribution. Total PCB concentrations in liver were variable and not consistently related to flounder reproductive condition (Table 2). Male flounder from Gaspee Point had hepatic PCB concentrations significantly greater than those in gravid or spent females, which did not differ. No consistent sex differences were observed in Fox Island or New Bedford Harbor fish. Sex differences in the concentration and distribution of individual PCB congeners were examined by one- and two-way ANOVA. Although ripe males had greater concentrations of certain congeners than did females, the differences were not consistent among all sites. For example, at Fox Island, congeners 77, 126, and 169 were at higher concentrations in males, while at Gaspee Point congeners 52 and 128 were higher. Congeners 47 and 151 were elevated in males from both Fox Island and Gaspee Point, but no reproductive status-related differences in the concentration of these two congeners were observed in New Bedford Harbor fish. The distributions (congener concentration/sum of congeners) of only two congeners (118 and 128) were sexually differentiated at more than one site. Since there were no strong gender differences in PCB congener concentration or distribution, average congener values for flounder liver from each site (sexes combined) are presented in Table 3.

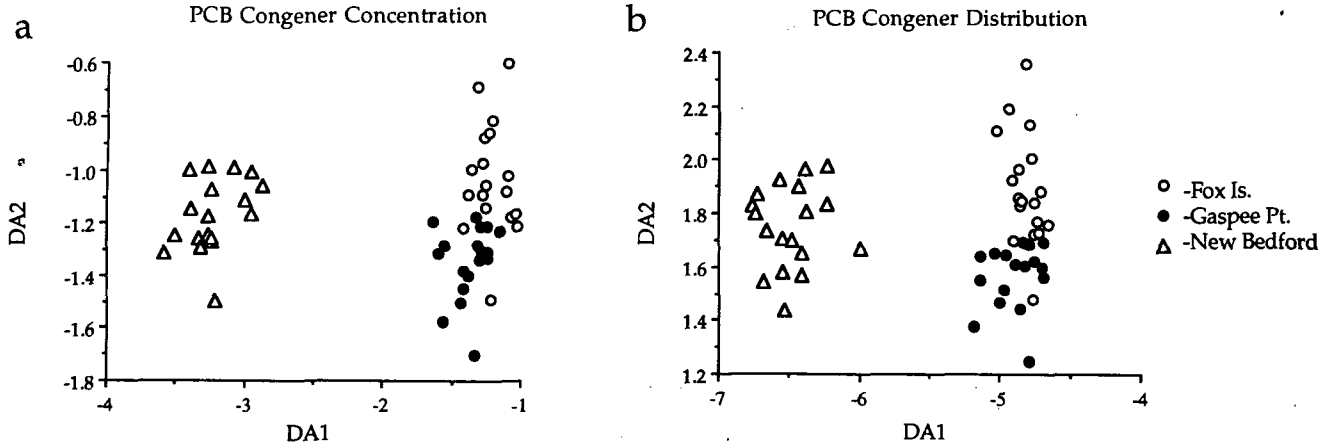

Figure 1. Winter flounder (n = 54) separated into discrete populations by collection site based on PCB congener (a) concentrations and (b) distributions (congener concentration/sum of congeners) in their liver. Eigenvectors calculated by MANOVA were used in discriminant analysis to derive discriminant axis components 1 and 2 (DA1, DA2). Data were log-transformed prior to analysis to remove skewness.

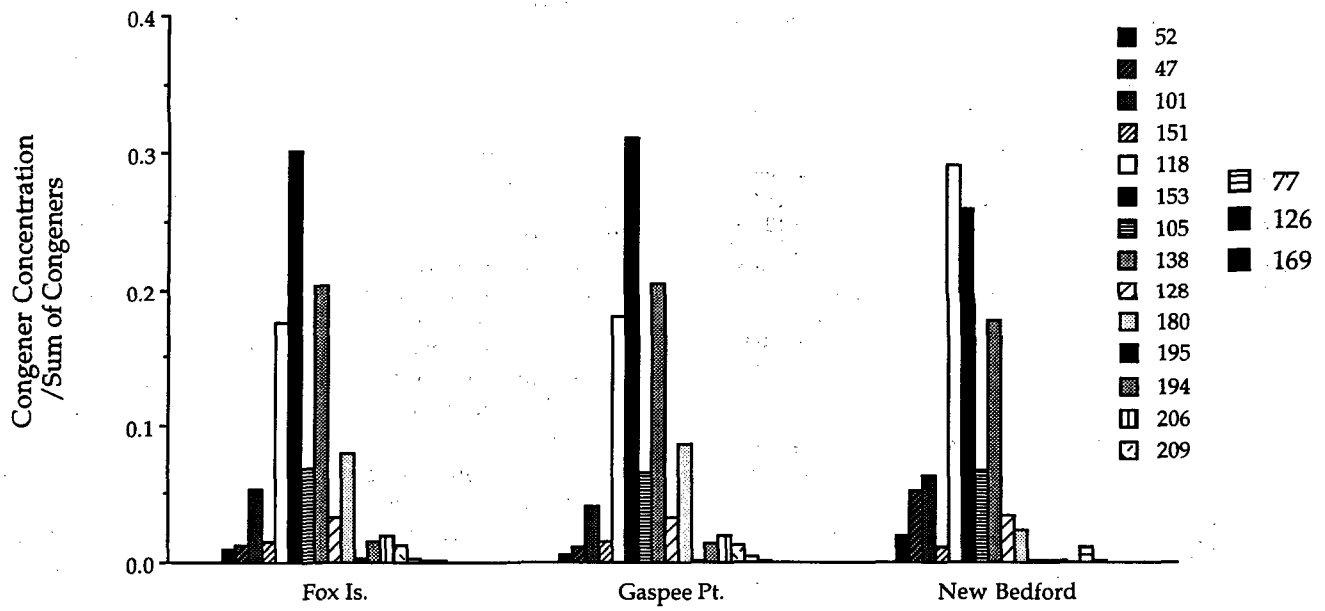

Figure 2. Relative concentrations of 17 PCB congeners (congener concentration/sum of congeners) in winter flounder liver.

Together these data indicate that reproductive status does not strongly or consistently influence PCB content in winter flounder liver. Laboratory studies have shown that female fish of some species can lose a significant percentage (50%) of their PCB body burden via spawning (*17*), but that males might be expected to lose less via this route (*16*, *43*). PCBs have been measured in winter flounder eggs (*23*), but the modest difference between spent and gravid females in the present study suggests that the redistribution of PCBs from liver to gonad might not substantially influence liver PCB content. Moreover, possible tissue saturation with PCBs could obscure sex differences in the New Bedford fish. Further work is necessary to determine how generally and strongly these factors affect PCB body burdens of highly contaminated fish as compared to slightly contaminated fish.

Mammalian CYP1A preferentially oxygenates coplanar congeners (*20*). The role of CYP1A in PCB congener metabolism is not yet established in winter flounder, but in other fish it has been linked to the metabolism of congener 77 (White, Gooch, and Stegeman, unpublished data). Since CYP1A content is suppressed in gravid female winter flounder (*21*), liver concentrations of the coplanar congeners 77, 126, and 169 might be expected to be lower in male than in female flounder. That the concentrations of these congeners are not reduced in male flounder suggests that their metabolism is not sexually differentiated. Therefore, CYP1A in winter flounder might not actively metabolize these congeners, or the rate of metabolism may be so slow in comparison to uptake that metabolism does not appreciably affect depuration and consequently bioaccumulation. In addition, congener 77 might inhibit its own metabolism as well as that of other compounds (*36*).

Evidence for PCB Congener Metabolism. Selective depletion, in flounder liver, of congeners 52, 101, and 151 suggests that these compounds are being selectively eliminated and presumably metabolized by these fish. Concentrations of congeners 52, 101, and 151, normalized to the concentration of congener 153, were 5–10-fold less in flounder liver than in sediment at all three sites (Table 4). Further, comparison of congener content normalized to congener 153 in New Bedford flounder with that in the bivalves *Mercenaria* and *Mya* (winter flounder prey organisms collected from New Bedford Harbor) showed a greater relative abundance of congeners 52, 101, and 151 in the bivalves (Table 5). The lack of congener depletion in bivalves (relative to sediment content) is in keeping with evidence that these organisms have little capacity to metabolize xenobiotics (*14*, *15*). It should be noted that

Table 4. Relative Concentrations of PCB Congeners, Normalized to Congener 153, in Sediment and Winter Flounder Liver[a]

	Fox Island		Gaspee Point		New Bedford	
congener no.	sediment	liver	sediment	liver	sediment	liver
52	–[b]	0.03	–	0.02	0.83	0.08
47	–	0.04	–	0.04	0.43	0.21
101	1.02	0.18	1.24	0.13	1.21	0.26
151	0.28	0.05	0.39	0.05	0.28	0.04
118	–	0.58	–	0.58	1.47	1.14
153	1	1	1	1	1	1
138	1.27	0.68	1.34	0.66	1.06	0.69
128	–	0.11	–	0.11	0.40	0.14
180	0.68	0.26	0.74	0.28	0.19	0.09
195	0.29	0.01	0.24	0.01	–	0.003
194	0.21	0.05	0.22	0.05	0.03	0.01
206	–	0.07	–	0.06	0.02	0.003
209	0.63	0.04	0.60	0.04	0.01	0

[a] Sediment values from refs 25–27. [b] Not analyzed.

Table 5. Congener Concentrations (Normalized to Congener 153) for Sediment (ref 27), Flounder Liver (This Study), and Bivalves (This Study) from New Bedford Harbor

	sample type			
congener	sediment	*P. americanus*	*M. areneria*	*M. mercenaria*
52	0.83	0.08	0.63	0.76
101	1.21	0.20	1.11	1.23
151	0.28	0.03	0.23	0.10
118	1.47	1.14	1.51	1.57
153	1.00	1.00	1.00	1.00
105	–[a]	0.22	0.18	0.16
138	1.06	0.61	0.83	0.73
128	0.40	0.10	0.10	0.06
180	0.19	0.10	0.04	0.10

[a] Not analyzed.

the sediment PCB congener values in Tables 4 and 5, taken from the literature, are nonetheless representative of sediment PCB profiles at our study sites. Congener profiles from Gaspee Point and Fox Island, which are widely separated in Narragansett Bay, have similar PCB congener distributions (*26*), indicating that the PCB profile does not change down the gradient. Likewise, profiles for the southern stations in New Bedford Harbor (A-I), of which our station (*D*) was one, are also very similar (*27*).

Congeners 52, 101, and 151 were the only congeners of the 17 we examined that had adjacent meta,para-unsubstituted carbon atoms (Table 1), suggesting that their low concentrations in winter flounder liver, relative to resident bivalves, may result from selective metabolism by flounder. In terrestrial mammals, PCB congeners with adjacent meta,para-unsubstituted carbon atoms are metabolized by CYP forms in the phenobarbital (PB)-inducible 2B subfamily (*20*). Fish are not responsive to PB or to PB-type inducers (*44*); however apparent homologues of 2B proteins have been described in fish (*45*), and we have detected proteins immunochemically related to CYP2B in winter flounder (unpublished). Therefore, it is possible that these enzymes may also be responsible for metabolizing PCB congeners with adjacent (meta,para) unsubstituted carbons in fish. Brown (*46*) has recently reached similar conclusions regarding PCB residue profiles in winter flounder.

Recent studies in marine vertebrates have shown evidence of selective metabolism of congeners with adjacent, unsubstituted meta,para carbons. Congener distribution patterns in bivalves relative to those of other

Table 6. TCDD Equivalent Concentrations (TEC, ng/g of dry liver) of Selected PCB Congeners in New Bedford Winter Flounder Liver (n = 18)

			TEC		
congener no.	congener concn[a] (ng/g of dry liver)	TEF[b,c]	% by wt[d]	ng/g	% concn[e]
77	391	0.002	2.5	0.8	4.5
126	48.6	0.3	0.3	15.0	85
169	3.7	0.001	0.02	0.004	0.02
105	2 200	0.000 8	14	1.8	10
118	12 800	0.000 006	83	0.08	0.5

[a] Average values for New Bedford Harbor flounder liver, taken from Table 3. [b] From refs 29 and 38. [c] TEFs from recent H4IIE cell studies: 1.8×10^{-5} (77); 2.2×10^{-2} (126); 5×10^{-4} (169); 7.6×10^{-6} (105); 3.6×10^{-7} (118) (*4*, Tillitt, personal communication). [d] Congener concentration as a percent of the sum of five congeners: (congener wt/congener sum wt) × 100. [e] Percent of toxic contribution.

marine organisms suggested specific congener metabolism in fish, seabirds, and marine mammals (*2*). Concentrations of congeners 52, 101, and 151 in seabird liver were depleted relative to concentrations in the prey of these birds (*47*). Juvenile sole fed a PCB-contaminated diet for 275 days had lesser relative amounts of congeners 52 and 101 in their livers than were present in the food, while the relative content of some other congeners, such as congener 180, was similar between food and fish liver (*10*). Together such findings lend support to our suggestion that the selective depletion of congeners with adjacent, unsubstituted meta,para carbon atoms in fish liver may be due to metabolism.

The concentrations of the highly chlorinated congeners, 194 and 209, were also lower by 3–16-fold in flounder liver as compared to that of sediment (Table 4). The large molecular volume of these congeners (*8*), the recalcitrance of highly chlorinated congeners to biotransformation (*6*), and the low concentrations of these congeners in bivalves from the same site (Table 5), suggest reduced bioavailability due to extreme hydrophobicity and sterically hindered biological uptake rather than metabolism.

Toxic Equivalents of PCB in Flounder Tissue. The potential (mammalian) toxicity of a biological organism can be estimated based on the toxic equivalents [TCDD equivalency factors (TEFs), see Materials and Methods] in that organism's tissues. Because TEF estimates vary depending on species and type of bioassay employed (*30*) and are continuously being reevaluated, we chose TEFs that were the most consistent between species and bioassay type. Using early estimates of TEF from Safe (*29, 38*), the highly toxic coplanar congeners, 77 and 126, contributed most (90%) of the TEC in flounder liver while the contribution of the mono-ortho congener, 118, was negligible (0.5%) (Table 6). Similar results were found for muscle tissue from New Bedford winter flounder collected in January 1987 (*32*) (data not shown). In contrast, using recent TEF estimates from Safe (*30*) the opposite conclusion can be drawn: congeners 77 and 126 together contributing less (36%) and congener 118 contributing most (53%) to the total flounder TEC. However, Safe's recent TEF estimate for the mono-ortho congener, 118, is highly conservative while the relative potency assigned congener 126 is low relative to most other studies (*4, 29, 30, 38*). In fact, recent work in mammalian H4IIE cells (*4*) produced TEF estimates for coplanar and mono-ortho congeners closer to Safe's earlier values (Table 6, footnote *b*). Thus, based on Safe's early TEF (*29, 38*) and Tillitt's recent TEF (ref 4; Tillitt, personal communication) data,

we conclude that trace concentrations of highly toxic coplanar congeners are much more important to the TEC of these winter flounder than less toxic congeners found in high concentrations.

There are a few other studies in which TEC values were calculated for individual PCBs in fish. In agreement with our results, Nimii and Oliver report that while congener 118 was one of the most abundant congeners in Lake Ontario trout, the high TEF of congener 126 resulted in the latter contributing the most to the TEC in those animals (*48*). In contrast, the TEC for the mono-ortho congener 105 equaled or exceeded that of congener 77 in fish from Waukegan Harbor, Lake Michigan (*49*).

TEC are often based on the analysis of PCBs in the whole body, and the relevance of TEC in unconsumed fish liver to humans may seem unclear. However, pharmacokinetic studies in vertebrates indicate that PCB congener distributions are very similar among tissues (*6*), and our results indicate that congeners in liver (Table 6) and muscle (data not shown) make similar relative contributions to TEC. It is important to note, however, that due to its greater mass the relative contribution of muscle PCB to the TEC will exceed that of liver.

The majority of TEF estimates available in the literature are based on mammalian models, and recent studies indicate that, in fish, the relative CYP1A induction potency and/or toxicity of different PCB congeners may differ. For example, ortho-substituted PCB congeners, including congener 118, are less potent inducers of CYP1A in some fish than in mammals (*36, 50*) and, thus, likely contribute little to the strong environmental induction of CYP1A in winter flounder (*31*). Walker and Peterson (*40*) recently showed that ortho-substituted PCBs were also less toxic to rainbow trout than would be expected based on mammalian studies. Due to the relative insensitivity of fish to ortho-substituted congeners found in that study (*40*), the calculation of winter flounder TECs using fish-dervied TEF values (data not shown) results in coplanar congeners contributing an even greater percentage of the potential toxicity of PCBs to flounder than to mammals, suggesting that the relative importance of different congeners to fish health likely differs from that to mammalian health.

Although we have used TEFs based on the toxicity of individual PCB congeners for calculating flounder TECs, it must be realized that interactive effects of PCB congeners with each other and/or with other xenobiotic contaminants may result in toxicities different from those predicted from individual TEFs. In rats, a combination of congeners produce toxicity, mutagenicity, and/or CYP1A1 induction greater than that produced by single congeners (*51, 52*). Conversely, PCBs can antagonize the effects of other xenobiotic compounds (*53, 54*). Since sediments from both Narragansett Bay and New Bedford contain a variety of potentially interactive xenobiotics (*55*), it is possible that the toxicity to mammals of flounder tissues from these sites differs from that based on PCB congener TEFs alone. It is clearly important that further research into such interactive effects be undertaken to strengthen the usefulness of TEFs in aquatic environmental work where exposure to contaminant mixtures is the rule.

PCB concentrations in New Bedford flounder flesh and liver exceed tolerance limits established in the United States and elsewhere. Total PCB concentrations measured in the edible flesh of New Bedford Harbor flounder exceed the FDA limit of 2 μg of PCB/g of wet edible tissue (*32*). In 1984, The Netherlands established maximum fish liver concentrations for PCB congeners 28, 52, 101, 118, 138, 153, and 180, congeners selected for their ease of quantitation, relative toxicity, and usefulness as indicators of the concentrations of tri- through hepta-chlorinated biphenyls (*56*). Average concentrations of congeners 118, 138, and 153 in New Bedford flounder liver exceed these limits.

Global Distribution of PCB Congeners in Aquatic Organisms. Comparison to PCB levels in other fishes puts the PCB concentrations in Fox Island, Gaspee Point, and New Bedford winter flounder into a global context. Total PCB concentrations in liver of New Bedford Harbor flounder (8.2–240 μg/g wet) are up to 5 times higher than those in fishes for which there are comparable data (i.e., 0.02–50 μg/g wet) from any other place in the world (*56–58*). The range of liver concentrations of congeners 77, 126, 105, and 118 in New Bedford fish are 8–270, 3–26, 74–1770, and 446–14 800 ng/g of wet liver, respectively. Concentrations of these congeners in fish liver have rarely been reported. Rattails from Carson Canyon and Hudson Canyon have mean levels of 9.9 and 129 ng of 105/g of wet liver, respectively (*57*); cod from the North Sea have 54–440 ng of 118/g of wet liver (*56*). Muscle PCB levels in New Bedford flounder also exceed those of most fish (*42, 47, 58–60*) with two exceptions. Walleye from Lake Kernaala, a heavily polluted lake in South Finland, have muscle PCB concentrations (14 μg/g wet, ref 61) similar to those of New Bedford flounder (0.1–7.1 μg/g wet, ref 32). The high whole body PCB content of Waukegan Harbor, IL, fishes (2.4–57 μg/g wet, ref 49) also suggests high muscle PCB content similar to that of New Bedford flounder. Muscle concentrations of congeners 77, 126, 105, and 118 are 0.1–14, nondetectable–1.1, 0.9–102, and 1.7–222, respectively, in New Bedford winter flounder (*32*), higher than those reported for these congeners in other fishes (*47, 56, 59, 60*) with one exception. Waukegan Harbor, IL, fish had whole body levels of 2–89 ng of 77/g of wet tissue and 80–483 ng of 105/g of wet tissue. In contrast to New Bedford, Fox Island and Gaspee Point flounder have moderate tissue PCB levels (0.1–1.9 and 0.3–5.8 μg/g of wet liver, respectively) similar to those of fishes from other sites (*56–58*). Pristine fish, by contrast, have much lower levels than the Narragansett Bay flounder. Cod collected off the Arctic coast of Norway have liver concentrations of congeners 77, 126, and total PCB of 0.01, 0.18–0.31, and 715–927 ng/g of liver lipid, respectively (*61*), values between 80- and 6500-fold lower than those in Fox Island flounder (nondetectable–65, nondetectable –48, and 3930–73 600 ng/g of liver lipid, respectively).

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