Proc. 14th Conf. Great Lakes Res. 1971
157-169. Internat. Assoc. Great Lakes Res.

PCBs IN FISH FROM THE MILWAUKEE REGION[1]

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Abstract. The results of this investigation provided spectrochemical confirmations of the presence of chlorobiphenyls (PCBs) and estimates for the composition and concentration of the PCB mixtures in fish from the Milwaukee River and Lake Michigan. The composition of the PCBs in fish varied with location of capture and its relation to PCB sources. Fish from the upper Milwaukee River contained from 2.3 to 15.4 μg/gm of Aroclor 1260 equivalent. Major sources of Aroclors 1242 and 1248 were found in the city of Milwaukee, and the fish in this region contained 405 μg/gm Aroclor 1248 equivalent. Lake Michigan fish contain residues resembling Aroclor 1254 at levels of approximately 18.6 to 22.4 μg/gm. (Key words: Aroclors; chlorobiphenyls; chlorinated hydrocarbons; PCBs; Lake Michigan).

INTRODUCTION

The chlorobiphenyls have stimulated considerable research interest since Jensen and Widmark first confirmed the presence of these chlorocarbons five years ago. Although the chlorobiphenyl mixtures (PCBs) were utilized in electrical and heat transfer systems (Hubbard 1964) before the advent of chlorinated pesticides such as DDT, it was not until pesticide research and environmental protection had matured through the use of gas chromatography and mass spectrometry that the PCBs were shown to coexist with pesticides in many organisms. Within the past few years, the PCBs have been found throughout the environment, particularly in organisms associated with natural waters which receive wastes from urban centers. Reviews of the chemical and physiological properties and the occurrences of the PCBs have been presented (Hubbard 1964; Veith and Lee 1970a; Peakall and Lincer 1970; National Swedish Environment Protection Board 1970).

While estimates of the levels of PCBs in environmental systems can rapidly be made with present analytical instrumentation, assessments of the long-term physiological consequences of the exposure of food chain systems to sublethal concentrations of PCBs are more difficult. Table 1 presents a brief summary of the toxicological effects of the PCBs, many of which must be regarded as preliminary identification.

Generally, the acute toxicity of the PCB mixtures to birds are of the same order of magnitude as DDT, although some variations are observed which result from differences in the slope of the feed dose-response curves for PCBs and DDT (Prestt, Jefferies and Moore 1970; Heath et al. 1970). The PCBs produce greater enzyme induction in avian, rat, and dog livers than does DDT (Risebrough et al. 1968; Street et al. 1969). In regard to enzyme induction and acute toxicity to some invertebrates, the PCBs are more active than DDT but less active than some of the oxygenated cyclodiene pesticides such as dieldrin and heptachlor epoxide (Lichtenstein et al. 1969; Street et al. 1969). The comparatively high

[1]Supported by the Wisconsin Department of Natural Resources, a NDEA Title IV Fellowship, and by the University of Wisconsin Research Committee. Additional support was given by the University of Wisconsin Engineering Experiment Station, Department of Civil Engineering, and Water Resources Center.

Table 1. Summary of PCB toxicities.

Organism	PCB Mixture	Dose or concentration	Remarks	Reference
Rats and dogs	A-1254	1,10,100 μg/gm	no effects after 2 yrs.	Monsanto (1970)
Chickens	A-1242	200-400 μg/gm	produced hydropericardial oedema, growth depression	Flick et al. (1965)
Bengalese finches	varied	low dose high dose	more toxic than DDT less toxic than DDT	Prestt et al. (1970)
Penned birds	A-1254 varied	25,50 mg/kg 5 day feedings	no effect after 5 days less toxic than DDT	Heath et al. (1970)
Avian livers	A-1254		increased steroid metabolism	Risebrough et al. (1968)
Hepatic enzymes	varied	varied	greater induction than DDT, less than heptachlor epoxide	Street et al. (1969)
Daphnia magna	varied	continuous bioassay	25 μg/l safe level A-1254 most toxic	Nebeker (1970)
Gammarus Oceanicus	A-1254	0.001-0.01 mg/l colloidal	lethal threshold in sea sublethal necrosis	Wildish (1970)
Juvenile pinfish Oysters Juvenile shrimp	A-1254	100 μg/l 100-1.0 μg/l 100 μg/l	no effect after 48 hr 100-19% shell reduction 80% mortality after 24 hr	Duke et al. (1970)
Juvenile shrimp	A-1254	1 μg/l	some mortality	Duke (personal communication)[1]
Crayfish	A-1254	continuous —7 day	TL_{50}; 80 μg/l	Stalling (1970)
Glass-shrimp			TL_{50}; 3.0 μg/l	
Damselfly		—4 day	TL_{50}; 200 μg/l	
Rainbow Trout		—10 day	TL_{50}; 8 μg/l	
Bluegills			TL_{50}; 443 μg/l	
Channel catfish		—15 day	TL_{50}; 741 μg/l	
Bluegills, trout Channel catfish	A-1254	1 μg/l	adverse physiological effects in continuous flow	Stalling (personal communication)[2]
Salmon	54% Cl	1 μg/l	egg mortality—16% for 9.2 μg/gm fat; 100% for 34.0 μg/gm fat	Johansson et al. (1970)

[1]T. W. Duke, 1970, Bureau of Commercial Fisheries, Pesticide Field Station, Gulf Breeze, Florida, personal communication to G. Fred Lee.
[2]D. L. Stalling, 1970, Fish-Pesticide Research Laboratory, Route 1, Columbia, Missouri.

mortality and/or adverse physiological effects in aquatic organisms caused by the PCBs at the lower μg/l levels (Duke, Lowe and Wilson 1970; Johansson, Jensen and Olsson 1970; Stalling 1970) is of interest because μg/l quantities of PCBs are found in municipal and industrial waste waters (Duke et al. 1970; Veith and Lee 1970b).

The purpose of this study was to examine fish from the Milwaukee River and nearshore (Wisconsin) region of Lake Michigan for the presence of PCBs. The objectives of the study were to relate the composition and quantities of the PCB mixtures to known PCB sources or areas of the river which have been contaminated with PCB-containing wastes.

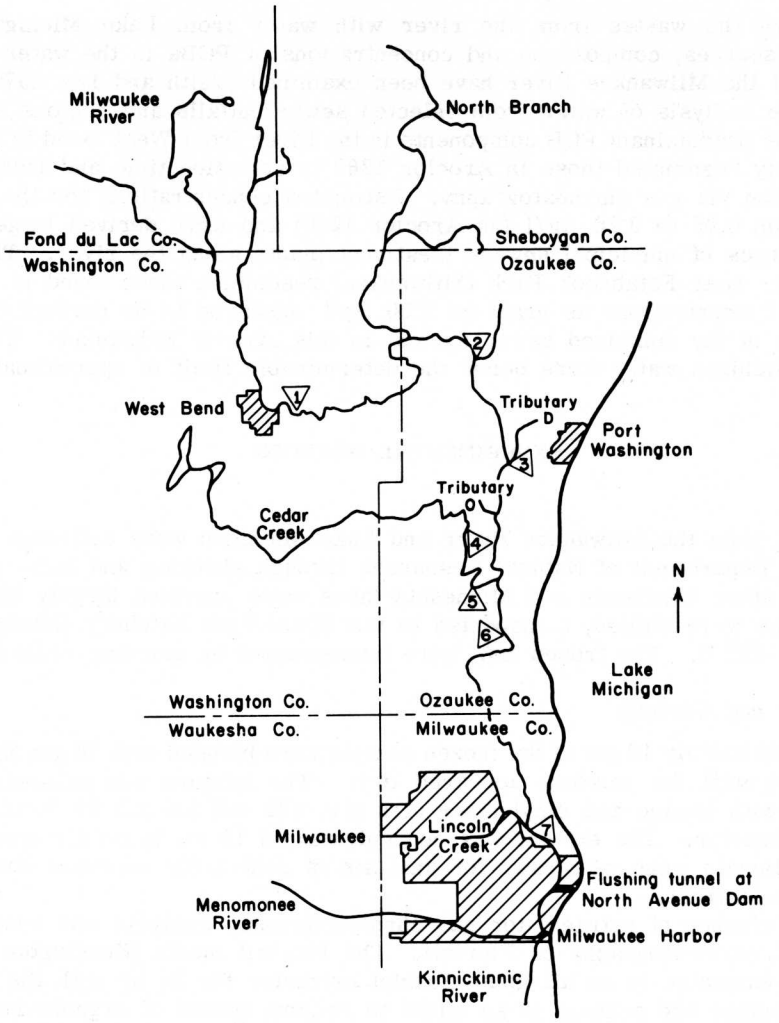

FIG. 1. Milwaukee River Watershed (Milwaukee River Study Committee 1968) ▽ -Muncipal STP* Outfall; 1-West Bend; 2-Fredonia; 3-Saukville; 4-Grafton; 5-Cedarburg; 6-Thiensville; 7-Milwaukee combined sewer outfalls.

*STP-Sewage Treatment Plant

Figure 1 presents the Milwaukee River watershed and major industrial and sewage treatment plant (STP) outfalls into the river. The Milwaukee River originates in the Kettle Moraine area of southern Fond du Lac and Sheboygan counties, Wisconsin, and flows in a southerly direction for approximately 95 stream miles into Lake Michigan at Milwaukee. The major municipalities along the river include West Bend, Saukville, Grafton, Cedarburg, Thiensville and Milwaukee, all of which discharge municipal and industrial wastes to the river. The major physical alterations of the natural river have been the construction of a total of 22 dams and spillways for power (Martin 1965), the dredging of the river from the mouth to the Milwaukee North Avenue dam, and the construction of a flushing tunnel just below the North Avenue dam in the city of Milwaukee

for flushing the wastes from the river with water from Lake Michigan.

The sources, composition and concentrations of PCBs in the water and plankton of the Milwaukee River have been examined (Veith and Lee 1970b) through the analysis of water from selected sewer outfalls and regions of the river. The predominant PCB components in the River from West Bend to Thiensville closely resembled those in Aroclor 1260 in retention time and relative concentration via gas chromatography. Estimated concentrations for the PCBs ranged from 0.05 to 0.26 μg/l (as Aroclor 1260) and were derived largely from the discharges of municipal sewage treatment plants along the river. The PCBs in the river near Estabrook Park (Milwaukee) resembled those found in Aroclor 1242, and concentrations as great as 2.80 μg/l appeared to be derived from the discharges of the combined sewer system in this area of Milwaukee. The PCBs in Lake Michigan water were below the determinable limit of approximately 0.01 μg/l.

EXPERIMENTAL METHODS

Sampling

Fish from the Milwaukee River and Lake Michigan were collected by the Wisconsin Department of Natural Resources through shocking and seining. The fish from other Wisconsin and Minnesota lakes were provided largely by anglers. All samples were chilled, transported to the Nevin Fish Hatchery (Madison), and frozen at -20^{o} C. The frozen fish were homogenized by grinding while frozen.

Extraction and Cleanup

Approximately 10 gm of the frozen sample were blended with 70 gm Na_2SO_4 (anhydrous) until the mixture appeared dry. The mixture was exhaustively extracted with hexane and ethyl ether (1:1 v/v, 170 ml) for 3.5 hr in an all-glass Soxhlet extractor. The extract was concentrated to 15 ml in an air stream, and suitable aliquots were withdrawn for analysis of nonvolatile material (fats and oils) in the extract.

The cleanup of extracts for gas chromatographic analysis was conducted with liquid chromatography on Florisil. The Florisil media (Kensington Scientific) was extracted in an all-glass Soxhlet extractor for 24 hr with the azeotrope of hexane and acetone in an effort to remove traces of organic impurities. The solvent was evaporated from the Florisil at 100^{o} C and the solid was heated at 650^{o} C for 2.5 hr for activation. If not used immediately after heating, the Florisil was warmed to 105^{o} C just prior to its use.

The Florisil column consisted of a 25 mm O.D. glass column which was filled with 19 gm of Florisil (Reynolds 1969). The column was fitted with a glass frit and Teflon stopcock at the bottom and was topped with 10 gm anhydrous Na_2SO_4 to prevent deactivation from water in the extract. The extracts (15 ml) were placed on the dry column and eluted with hexane (200 ml) at 3 to 5 ml/min to obtain DDE, some DDT, heptachlor, aldrin, toxaphene and PCBs, if present. The receiving flask was changed, and the column was eluted with 20% ethyl ether in hexane (200 ml) to obtain DDT, DDD, dieldrin, heptachlor epoxide and lindane, if present (Reynolds 1969). The samples were concentrated to appropriate volumes for GLC analysis.

In general, the 19 gm Florisil column was suitable only for removing the bulk of the co-extracted nonvolatile materials and dieldrin from the chlorobiphenyl and DDT family of pesticides.

Situations arose with samples from some regions which indicated that the 25 mm O.D. Florisil column did not adequately remove chlorinated pesticides

from the PCBs. Therefore, the column design was modified in an effort to optimize parameters in accordance with liquid chromatography theory. The column diameter was reduced to 9 mm I.D. Optimal conditions appeared to be an 8 to 10 gm x 9 mm I.D. column of approximately 100 mesh Florisil which was eluted with 50 to 60 ml hexane followed by ether/hexane or methylene chloride/hexane mixtures. In addition, samples for which the mutual interferences between the PCBs and pesticides were significant were chromatographed on the silicic acid column described by Armour and Burke (1970). However, since the efficiencies of all of the liquid chromatographic separations were not entirely reproducible, the more difficult to interpret analyses of Lake Michigan fish were routinely replicated to increase the reliability of the analyses.

Confirmatory Evidence

Confirmations of PCBs in representative samples from the study area were obtained with IR and mass spectral data. Initial confirmations were made by extracting and isolating sufficient quantities of the PCB components from fish of the Milwaukee River via semi-preparative gas chromatography, comparing IR spectra of a component with the corresponding component (GLC retention time) of the Aroclor mixtures, and determining molecular weights and chlorine numbers by direct insertion into a double-focusing mass spectrometer. IR data were obtained from micropellets with a Beckman IR-10 spectrometer equipped with a beam condenser. Mass data were obtained on an AEI MS-902 mass spectrometer located in the University of Wisconsin, Department of Chemistry (Professor P. E. Bender). In addition, a Varian CH-7 mass spectrometer interfaced with an Aerograph 1740 gas chromatograph has been employed in confirming the presence of some PCBs in Lake Michigan fish. The PCB components isolated from Milwaukee River fish were those with m/e of 324 to 426. The PCB components confirmed in this preliminary study of Lake Michigan fish were those with m/e of 256 to 324.

Analytical Gas Chromatography

Quantitative gas chromatographic analyses were conducted on an Aerograph 1745-20 gas chromatograph equipped with concentric tube electron capture detectors (^3H, 250 mc) and a 50:50 effluent splitter for simultaneous analysis with electron capture and flame ionization detectors. The most suitable columns included 2.0 m x 1.8 mm (I.D.) glass coils which were packed with OV-101 (3%), OV-101/XE-60 (3/3%), and OV-101/QF-1 (3/4.5%) on Gas Chrom Q (120/140 mesh). The carrier gas (purified N_2) was maintained at 21 ml/min; and the injector, column and detector temperatures were 240° C, 180° C and 220° C, respectively.

There are three major approaches to estimating the quantity of PCBs in the cleaned up extracts. Perhaps the most exact method for PCB analyses is that of determining the concentration of each PCB component in a manner similar to other pesticides. This method required analytical standards for each component (or major components). Pure isomers of PCB have not been readily available, and it has been necessary to isolate components of PCB via GLC trapping of the Aroclor mixtures. The major components of the PCB (Aroclors) which have been found in Wisconsin waters have been isolated and recombined to form four standard mixtures. The components are well resolved and have been adjusted in concentration to similar peak heights. Only preliminary standardization of these standards has been accomplished, and a more thorough evaluation will be necessary before they can be applied in routine analyses.

The second method is assigning weight-equivalents by measuring one or more peak heights of the mixture. This method has been employed in many of the studies reported to date, and in most instances, the chosen standard PCB mixture is Aroclor 1254 (or comparable mixture). While this method may be more rapid than the others, the composition of the mixture of PCB may be altered in the environment, and the peak-height method may not produce consistent results. Also, several of the PCB components in Aroclor 1254 elute simultaneously with the DDT family of pesticides and would cause high results if adequate separation were not attained in the column cleanup step.

The third method involves a visual comparison of each chromatogram to the most similar Aroclor mixture(s) and quantitation by comparing the area of the chromatogram to an area-standard curve. This method has been used in this study and provides qualitative and quantitative information regarding the PCB composition. The major limitation of this method is that it may include areas of components which are not PCBs. It requires that each sample or group of samples be examined by more specific methods and thereby increases the time needed for each sample. However, the time limitation is not viewed as a major problem in exploratory studies such as this work.

Precision and Accuracy

The reproducibility of the analysis of fish for the PCBs is summarized in Table 2. The data are derived from six replicate samples (10 gm) of an uncontaminated fish from Trout Lake in Vilas County, Wisconsin, which were "spiked" with 100 μg of Aroclor 1254 in 200 μg of hexane, six replicates of a composite goldfish sample and six replicates of a rainbow trout homogenate. Each sample was injected twice to estimate the relative error due to injection and planimetering (three integrations), which was less than ± 1.5%.

The "spiking" procedures may not produce a representative sample since the organochlorine mixture is not likely to be assimilated into the tissues in the same manner as in environmental samples. Nonetheless, the average recovery was estimated to be 87.2 ± 3.2%. The relative error was ± 3.7% and was less than in the analysis of environmental samples, reflecting the fact that

Table 2. Quantitative analysis of PCBs in fish.

Sample	Number of replicates	Aroclor 1254 added (μg/gm)	Recovery (percent)*
Cisco			Range: 83.1 - 91.4
Trout Lake	6	10.0	Mean: 87.2 ± 3.2
("spiked")			Relative error: ± 3.7%
	Number of replicates	**Most similar aroclor**	**Aroclor concentration (μg/gm)***
Goldfish			Range: 370-446
Milwaukee River	6	1248	Mean: 405 ± 20
			Relative error: ± 5%
Rainbow Trout			Range: 17.1 - 21.2
Lake Michigan	6	1254	Mean: 19.0 ± 1.0
			Relative error: ± 5.2%

*Each value represents the average of two injections and total area determinations.

TABLE 3. Results of analysis of Milwaukee River fish for PCBs.

Location	1969 Date	Species and body weight (Kg) or length*	Fat percent	Most similar Aroclor	Concentration** (μg/gm)
Above dam, Grafton	5-27	Redhorse (*Moxostoma macrolepidotum*), 1.2	2.4	1260	14
" " "	"	Carp (*Cyprinus carpio*), 1.1	1.4	1260	15
" " "	"	3 White Suckers (*Catostomus commersonnii*) 0.6, 0.6, 0.7	2.1	1254	2
" " "	"	3 Small mouth bass, 8.8-11.1 inches	1.1	1260	***
" " "	"	Northern Pike, 13.3 inches	2.1	1260	3
" " "	"	7 Bullheads, 5.2-6.6 inches	1.7	1260	***
Below dam, Grafton	"	2 White Suckers, 0.4, 0.4	4.8	1260	10
" " "	5-28	Northern Pike, 20.4 inches	3.9	1260	24
" " "	"	3 White Suckers, 11.1-11.4 inches	3.7	1260	4
" " "	"	2 Carp, 14.6, 13.2 inches	5.5	1260	38
" " "	"	2 Carp, 14.2, 14.6 inches	6.1	1260	25
Above dam, Thiensville	5-27	Carp, 1.6	2.8	1260	13
" " "	"	4 Redhorse, 8-13 inches	2.8	1260	19
" " "	"	Carp, 12 inches	8.1	1260	16
" " "	"	3 Pumpkinseed (*Lepomis gibbosus*) 5 inches	2.4	1260	14
" " "	"	White Sucker, 12.8 inches	4.7	1260	11
" " "	"	Northern Pike, 24.6 inches	2.0	1260	22
" " "	"	9 Bullheads, 5.1-7.1 inches	1.7	1260	21
Estabrook Park	5-28	7 Goldfish (*Caraesus auratus*) 1-7 inches	18.7	1248	405
" "	"	4 Goldfish, 3-8 inches	7.3	1248	139
" "	"	2 White Suckers, 11.5-13.7 inches	3.9	1248	64
Milwaukee Harbor	6-3	Chub, 12 inches	23.2	1254	20
" "	"	Chub, 11 inches	23.1	1254	23
" "	6-5	Lake Trout (*Salvelinus namaycush*), 2.3 inches	22.9	1254	26
" "	6-3	Yellow Perch (*Perca flavescens*), 10 inches	3.6	1254	4
" "	6-5	Coho Salmon (*Oncorhynchus kisutch*), 4.0	24.1	1254	18

*Furnished by Wisconsin Conservation Division
**Concentration based on total area
***Below the determinable limit of 2.0 μg/gm for 10 gm sample

TABLE 3. Results of analysis of Lake Michigan fish for PCBs, cont'd.

Location	1969 Date	Species and body weight (Kg) or length*	Fat percent	Most similar Aroclor	Concentration** (μg/gm)
Milwaukee Shorewood	6-5	Chinook Salmon (*Oncorhynchus tschawytscha*) 27 inches	25.3	1254	19
" "	6-3	Coho, 5.28 lbs., 23.5 inches	8.4	1254	14
" "	"	Coho, 2.8 lbs., 20.7 inches	8.3	1254	25
" "	"	Coho, 24 inches	11.2	1254	24
" "	"	Yellow Perch, 13.1 inches (spawned)	2.5	1254	4
" "	"	4 Yellow Perch, 7.2-8.5 inches	4.0	1254	4
" "	"	Rainbow Trout, 10.5 inches	7.1	1254	10
Kewaunee County	4-16	Rainbow Trout (*Salmo gairdnerii*, Spring 1967, 24 inches	8.5	1254	19
" "	"	Rainbow Trout, 27.3 inches	11.8	1254	17
" "	"	Rainbow Trout, 21.1 inches, (Spring, 1968)	8.6	1254	16
" "	"	Rainbow Trout, 16.8 inches, (Spring, 1968)	8.0	1254	6
" "	"	Rainbow Trout, 25.0 inches, (Spring, 1966)	7.2	1254	25
" "	"	Rainbow Trout, 25.9 inches	7.0	1254	16
" "	"	Rainbow Trout, Spring 1967, 28 inches	10.1	1254	22
Sucker Creek, Ozaukee	5-1	2 White Suckers, 15 inches	5.7	1254	12
" " "	"	8 Red-tailed Chubs	3.4	1248	38
Sauk Creek, Ozaukee	"	2 White Suckers, 12 inches	5.0	1254	16
" " "	"	White Sucker, 2 lbs.	1.4	1254	2
" " "	"	Rainbow Trout	1.4	1254	2
" " "	"	Rainbow Trout, 23 inches	2.6	1254	79
" " "	"	10 Creek Chubs	3.1	1254	20
Point Creek, Manitowoc	"	3 White Suckers, 14 inches	4.2	1248	50
Oconto River, Green Bay	5-7	Walleye, 2 lbs.	4.1	1248	43

*Furnish by Wisconsin Conservation Division
**Concentration based on total area

the data are derived from "spiked" samples. The overall relative error from extraction, cleanup and analysis was approximately ± 5%. The concentration of Aroclor 1248 in the goldfish was 405 ± 20 μg/gm, and that of Aroclor 1254 in the trout was 19.0 ± 1.0 μg/gm on a whole fish basis.

RESULTS AND DISCUSSION

The fish from five areas of the Milwaukee River and five areas of western Lake Michigan were sampled in the spring of 1969. The results of the analyses of the fish on a whole fish basis are presented in Table 3. The qualitative data show that the more highly chlorinated biphenyls were predominant in the fish from the Grafton and Thiensville areas and that the residues could best be determined as Aroclor 1260. These results are in agreement with the results of the water analyses in these areas (Veith and Lee 1970b). The predominance of isomers in the fish changed from Aroclor 1260 to Aroclor 1248 in the Estabrook-East Riverside Park area (city of Milwaukee), although the late-eluting Aroclor 1260 components were evident in concentrated extracts. Figure 2 compares the chromatograms of an extract of a composite goldfish sample from Estabrook Park (Fig. 2a) with those of Aroclors 1242 and 1248 (Fig. 2b). The peak heights of Aroclors 1242 and 1248 have been normalized to the major component eluting at 7.3 min. The earlier-eluting components in the fish closely resemble the Aroclor 1242 mixture while the later-eluting components are too large on a relative basis to be assigned to Aroclor 1242. Therefore, the PCBs have been conservatively estimated as Aroclor 1248 which underestimates the early components.

The mixtures of PCBs in the fish from the Milwaukee harbor area were not the same as those several miles from the mouth at Estabrook Park. The fish in the harbor, together with all of the fish from Lake Michigan, contained residues which produced chromatograms resembling that of Aroclor 1254. Figure 3 is a chromatogram of a Lake Michigan rainbow trout extract (hexane fraction) after preliminary cleanup on a 19 gm - 25 mm O.D. Florisil column (Reynolds 1969). Nine of the major components have retention times identical with major components of Aroclor 1254. DDE is evidenced by the large peak at 7.0 min. pp' DDT is indicated by the increased height of the peak at 13.0 min with respect to the peak at 10.5 min which is not found in the corresponding peaks in Aroclor 1254. Further separation of the components by liquid chromatography (Armour and Burke 1970) removed the DDT and DDT metabolites and produced subsequent GLC chromatograms similar to Aroclor 1254. The components at 21.5 and 26 min correspond with components in the Aroclor 1260 mixture; however, the nature of these compounds have not been confirmed.

The quantitative data show that the concentration of Aroclor 1260 in the fish from above the Grafton dam ranged from below or near the determinable limit in bullheads, bass and white suckers to 15.4 μg/gm in a carp. Above the dam at Thiensville, the concentrations of Aroclor 1260 were similar in that they ranged from 10 μg/gm in white suckers to 22.3 μg/gm in a northern pike. The data from fish in the Estabrook Park region indicated that, not only were the lesser chlorinated biphenyls predominant, but also the concentrations of the total mixture increased by a factor of about 20 to 405 μg/gm in the composite of small goldfish. Thus, the concentrations of PCBs in the fish reflected the increased levels of PCBs in the water near Estabrook Park.

In the harbor area, the Aroclor 1254 isomers were present at concentrations which varied from 4 μg/gm in the immature yellow perch to 26 μg/gm in the lake trout. The lake trout and coho salmon captured in the harbor contained residues similar to the fish from Lake Michigan (off Milwaukee Shorewood

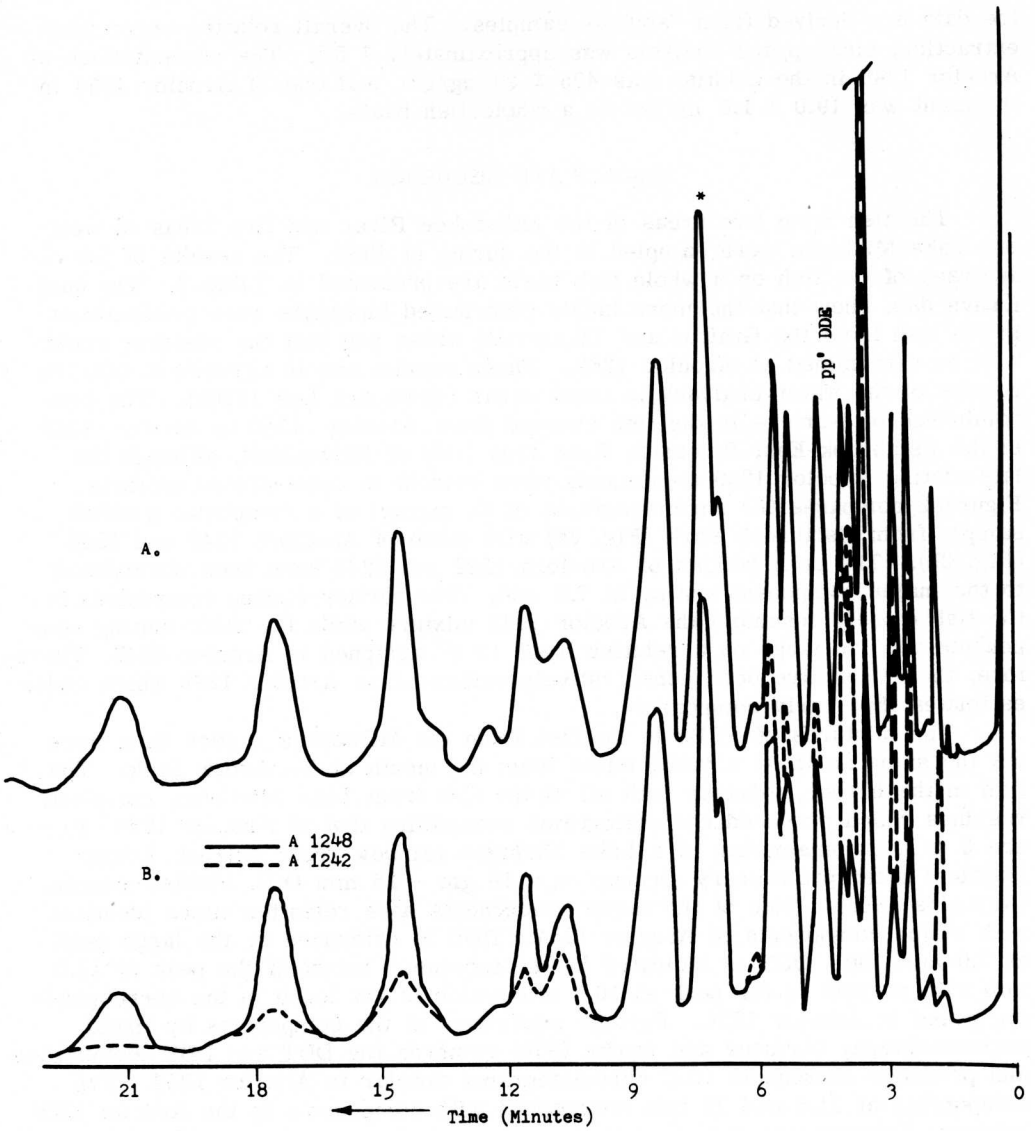

FIG. 2. Comparison of chromatograms of Aroclor 1248 with the extract of goldfish from the Milwaukee River. A. Goldfish from Estabrook Park; 1.3% fat; injection equivalent to 0.1 μg whole fish. B. 40 μg Aroclor 1248 (——) and Aroclor 1242 (---) normalized to component at 7.3 min.

and Kewaunee County) in that the latter also contained Aroclor 1254 ranging from 19 to 22 μg/gm.

The white sucker samples from Sucker Creek and Sauk Creek (Washington County), both of which discharge to Lake Michigan, contained 12 and 16 μg/gm of Aroclor 1254, respectively. These data indicated that the fish may represent fish from Lake Michigan. The 23 inch rainbow trout captured off Ozaukee County contained 79 μg/gm PCB (as Aroclor 1254) which was the highest level determined in Lake Michigan fish. The composite of three white suckers from

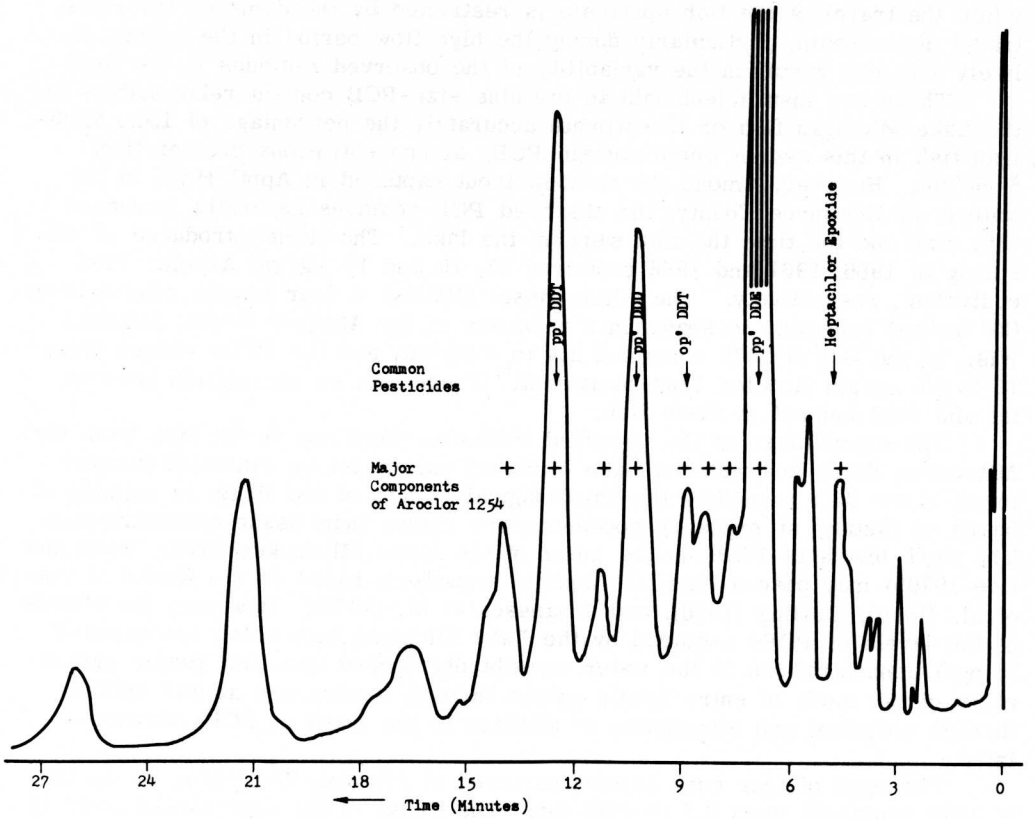

FIG. 3. Chromatogram of concentrated extract of rainbow trout 10.3% fat; injection equivalent to 0.2 mg whole fish; (+) retention times of major components of Aroclor 1254.

Point Creek (Manitowoc County) contained 50 μg/gm of Aroclor 1248 which indicated that these fish may have been exposed to relatively high concentrations of Aroclor 1248 in addition to the apparent Aroclor 1254 in Lake Michigan water and/or the fish food chain.

These results point out several interesting aspects of this study of the Milwaukee River. The first is the variability of the "apparent PCB mixtures" which can be found in fish from relatively small study areas such as sections of the Milwaukee River and the consistency of the PCB composition in fish from the Lake Michigan system. The residues in the fish from the river appear to reflect the PCB discharges in the vicinity of the sampling site (Veith and Lee, unpublished) and the history of the fish movement. On the other hand, the composition of PCB in Lake Michigan fish best resembled Aroclor 1254, and little variation was noted in the fish examined by this study. Since Aroclor 1248 components were not observed in Lake Michigan fish, this evidence may indicate that the "apparent Aroclor 1254" in Lake Michigan fish is a stable mixture of PCB components which results from the degradation of lesser chlorinated PCB components.

The second point of interest is the lack of any correlation between the PCB concentration in the fish and the size or lipid content of the fish. The large variations in the PCB in fish in the Milwaukee River likely result from the difference in feeding habits and residence time near the PCB sources.

While the travel of the fish upstream is restricted by the dams on the river, travel downstream, particularly during the high flow period in the spring, is likely and may result in the variability of the observed residues in the fish.

There are insufficient data to examine size-PCB content relationships in the Lake Michigan fish or to estimate accurately the percentage of Lake Michigan fish in this region which contain PCBs at concentrations greater than 5 μg/gm. However, among the rainbow trout captured in April 1969, in the waters off Kewaunee County, the observed PCB residues generally increased with size and the time the fish were in the lake. The trout introduced in the spring of 1966, 1967 and 1968 contained 25, 19 and 17 μg/gm Aroclor 1254 equivalent, respectively. The edible tissue (fillets) of four female coho salmon (28 inches) returning to spawn in a tributary of the Ahnapee River, Algoma, Wis., in the fall of 1970 contained 2.4 to 7.5% fat, and the PCBs ranged from 10 to 17 μg/gm Aroclor 1254 equivalent. There was no correlation between fat and PCB content in these fish.

The significance of the observed PCB concentrations in the fish from the Milwaukee River and western Lake Michigan cannot yet be evaluated because much of the data regarding the physiological effects of the PCBs in animals is based on dietary or exposure concentrations rather than tissue concentrations. The μg/l levels of PCBs in the water of the lower Milwaukee River (Veith and Lee 1970b) may pose a threat to aquatic organisms based on the works of Duke et al. (1970), Stalling (1970), and Johansson et al. (1970). However, the effects of the levels of PCBs acquired by the Lake Michigan fish which are exposed to ng/l concentrations in the water undoubtedly depend upon the kinetic parameters of the mode of entry (biotic uptake through feeding and abiotic uptake through sorption) and elimination in addition to the mass of PCBs accumulated.

The eggs of four coho salmon captured at Algoma, Wisconsin, in the fall of 1970 contained from 6.7 to 9.5% fat. The PCBs in the eggs ranged from 12 to 17 μg/gm as Aroclor 1254 on a fresh weight basis. Johansson et al. (1970) have demonstrated that the mortality of salmon eggs increased from 16% in eggs containing 9 μg/gm PCB (fat weight basis) to 100% in eggs containing 34 μg/gm PCB. Although similar studies with Lake Michigan fish have not yet been reported, the data suggest that the PCBs in Lake Michigan could influence the reproductive success of fish populations. Studies are needed to determine if relationships similar to those reported by Johansson exist in the fisheries of Lake Michigan.

The analyses of concentrated extracts from fish collected from Trout Lake (Vilas County, Wis.), Lake Hanska (Brown County, Minn.) and Lake Mendota (Dane County, Wis.) failed to produce any indication of PCB in the fish. None of these lakes receive industrial wastes, although Lake Mendota receives urban runoff from the city of Madison. Lake Hanska receives considerable agricultural drainage via several networks of drainage ditches. The absence of PCB in these lakes indicates that the PCB contamination through application with pesticide formulations and/or aerial transport has not been significant in these regions.

SUMMARY

The analysis of fish from the Milwaukee River and nearshore areas of western Lake Michigan have shown that the PCBs are present throughout the river and the study area in Lake Michigan. The composition of PCB in fish in the river varied with sampling location and were comparable to the composition of PCBs in waste discharges. In contrast, the composition of PCBs in

Lake Michigan was remarkably constant and closely resembled that of Aroclor 1254. The concentrations of PCBs in fish were highly variable and ranged from below the determinable limit to 405 μg/gm on a whole fish basis. Approximately 50% of the Lake Michigan fish contained PCB residues in the range of 15 to 25 μg/gm as Aroclor 1254.

ACKNOWLEDGMENTS

The assistance of Paul Bender of The University of Wisconsin Department of Chemistry and Alfred Thurston of the EPA Southeast Water Laboratory is appreciated.

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