P C B N E W S L E T T E R

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ANALYTICAL METHODS

AND

CHEMICAL CHARACTERISTICS

MONS 087252

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MS Identification of PCB's

A manuscript entitled "Use of Isotopic Abundance Ratios in the Identification of Polychlorinated Biphenyls by Mass Spectrometry" by J. W. Rote and W. J. Morris has been submitted to the Journal of the Association of Official Analytical Chemists for publication.

We describe a method which provides unambiguous identification of PCB's in the presence of interfering substances. The mass spectra of PCB's, polychlorinated terphenyls (PCT's) and chlorinated naphthalenes (CN's) are characterized by a strong parent ion cluster with an isotopic abundance ratio determined by the number of chlorine atoms substituted on the rings. Although most workers using MS are probably aware of these isotopic clusters, no one to date has compared theoretical ion intensities to observed spectra.

From the natural abundance ratios of chlorine and carbon isotopes, we have calculated the theoretical probability of the occurrence of ions of different masses in the molecular cluster for all degrees of chlorination in CN's, PCB's and PCT's. Using a computer controlled Finnigan GC-MS system, we have taken the mass spectra of mono- through decachlorobiphenyl and have compared the observed isotopic abundance ratios to the theoretical values. A chi-square test shows the matching of observed to expected intensities significant at the 99% level for all PCB's.

As the probability of the occurrence of ^{37}Cl increases with increasing degree of chlorination, there is a shift of the most intense ion in the cluster from the parent ion (M^+) to M^++2, then to M^++4. These patterns of intensity are unique for each chemical species, thus providing a ready means of identification.

An additional tool in identifying PCB's is found in their fragmentation pattern. The spectra show the molecular cluster always to be the most intense with a loss of even numbers of chlorine atoms from the molecule favored upon fragmentation. PCB spectra that show the M^+-Cl or M^+-Cl_2 ions more intense than the parent ion usually indicate that there is more than one isomer present under the GC peak being scanned. Characterization of Aroclor mixtures by GC-MS shows that this is often the case.

As more laboratories turn to GC-MS analysis of environmental pollutants, the use of isotopic abundance ratios and fragmentation patterns will become increasingly more valuable as an aid to identification.

Copies of the plotted spectra of mono- through decachlorobiphenyl and tables of theoretical ion intensities for CN's, PCB's, and PCT's along with observed ion intensities for the PCB's are available upon request.

Leon D. Sawyer, Pesticide Specialist; Food and Drug Administration, 240 Hennepin Ave., Minneapolis, MN 55401

Collaborative Study on the Recovery and Gas Chromatographic Quantitation of Polychlorinated Biphenyls in Chicken Fat and Polychlorinated Biphenyl-DDT Combinations in Fish

This is an Associate Referee Report on the project "Extension of Multiple Chlorinated Pesticide Residue Method to Polychlorinated Biphenyls." This paper presented at the 86th Annual Meeting of the Association of Official Analytical Chemists, Oct. 9-12, 1972, at Washington, D.C.

Nine laboratories collaborated on the analyses of PCB in chicken fat and DDT/PCB combinations in fish. Existing AOAC multipesticide methodology with GLC quantitation was employed. One solution containing a mixture of Aroclors 1254 and 1260 was analyzed by GLC only. The fish samples were subjected to a published silicic acid procedure for separating the DDT/PCB mixtures. The DDT analogs were quantitated before and after the separation. The PCB content was quantitated by total peak height and total area comparisons against appropriate Aroclor(s) using EC-GLC, and additionally in 6 laboratories by total area comparisons using halogen specific detection. The EC-GLC data demonstrated better accuracy and precision. The following PCB recoveries were obtained using total peak height comparisons: 5 ppm mixed Aroclor solution, 100 $\pm$ 4%; 8 ppm 1242 fortified chicken fat, 101 $\pm$ 13%; 7.5 ppm 1248 fortified chicken fat, 96 $\pm$ 9%; incurred 1242 chicken fat, 9.2 ppm $\pm$ 8%; 6 ppm 1254 fortified fish, 75 $\pm$ 14%; 6 ppm 1260 fortifiad fish, 75 $\pm$ 15%; and an environmentally incurred residue in fish, 4.5 ppm $\pm$ 20%. The 2 Aroclor fortified fish samples were concurrently spiked with the p,p' isomers of DDE, TDE, and DDT at levels of 4, 1, and 3 ppm, respectively. After silicic acid separation the combined recoveries for these 2 samples were: DDE, 86 $\pm$ 13%; TDE, 89 $\pm$ 20%; and DDT, 84 $\pm$ 17%. Environmentally incurred DDTs were recovered at 4.1 ppm $\pm$ 14% for p,p'-DDE, 0.7 ppm $\pm$ 24% for o,p'-DDT, and 2.7 ppm $\pm$ 17% for p,p'-DDT. The DDT values calculated before the silicic acid separation compared favorably with those summarized.

J. A. Armour, Division of Chemical Technology, U. S. Food and Drug Administration, Washington, D.C. 20204

Quantitative Perchlorination of Polychlorinated Biphenyls as a Method for Confirmatory Residue Measurement and Identification

The perchlorination procedure for derivatization of PCB described by Berg, Diosady, and Raee has been modified to achieve

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a micro scale quantitative conversion (greater than 90%) of commercial PCB preparations, (Aroclors®), to decachlorobiphenyl. Cleaned up sample extracts containing PCB residues (1 µg-20 µg) are reacted with antimony pentachloride in the presence of chloroform to form decachlorobiphenyl. This procedure converts a multicomponent mixture to a single derivative detectable by EC-GLC thus providing an easy method for quantitating and identifying PCB residues and at the same time increasing the sensitivity of detection. The usefulness of the perchlorination procedure is demonstrated by comparing results for environmentally contaminted samples quantitated by measuring the total area of the EC-GLC response for the residue against the Aroclor it most closely resembles to results obtained by measuring the single peak of the decachlorobiphenyl derivative and expressing the results in terms of the particular Aroclor.

James J. Lichtenberg, Group Leader, Pesticides Identification, United States Environmental Protection Agency, National Environmental Research Center, Cincinnati, Ohio 45268

Formaldehyde solution has been found effective for preserving water samples to be analyzed for PCB's. Quantitative recoveries are achieved over a 14-day holding period when PCB's are dosed into natural river water and preserved with approximately 15 ml of formaldehyde solution per liter of sample. The PCB's are recovered intact with no interfering artifacts or no apparent chemical effect on the PCB's at the 50-1000 ng per liter level. Samples not preserved show significant "loss" of PCB's if held longer than 8 hours. As much as 10% loss has been observed in a 24 hour period. The rate of loss slows significantly after 2 to 3 days so that after 7 days the recoveries are 60 to 70%. These effects were observed on samples stored under normal room temperature and lighting conditions. Storage of samples in the dark, in a refrigerator, retarded the loss somewhat but the same trend was noted.

Ronald C. Webb, Research Chemist, United States Environmental Protection Agency, Southeast Environmental Research Laboratory, Athens, Georgia 30601

Copies are available of a paper on "Quantitative PCB Standards for Electron Capture Gas Chromatography." The abstract may be of interest:

The weight of PCB represented by each electron capture gas chromatographic (EC-GC) peak in solutions of Aroclors 1221-1260 has been determined. The Aroclor samples from which these

solutions were prepared are proposed as quantitative PCB standards. Their compositions were determined by elemental analysis, gas chromatography with a Coulson conductivity detector, and combined gas chromatography-mass spectrometry. Retention times relative to p,p'-DDE are recommended to designate individual GC peaks of PCB's. A table is given for each Aroclor showing the weight percent of each EC-GC peak in the mixture. A procedure using Aroclors 1242, 1254, and 1260 is recommended for analyzing environmental samples containing more than one Aroclor mixture. Stock solutions of the Aroclors in isooctane are stable except when directly exposed to sunlight. Ampoules of the Aroclor solutions are offered.

Laverre D. Uhlken, Richard W. Carlson, and Robert M. Tyo, United States Environmental Protection Agency, National Water Quality Laboratory, Duluth, Minnesota 55804

Apparent Volatility of PCB's as used in Continuous-Flow Bioassays

Recent studies regarding the toxicity of PCB's to Daphnia under static conditions have indicated that the concentration of PCB's in the water decreases markedly with time after the PCB's are added as an acetone solution. Moreover, the measured concentration of PCB's in continuous-flow bioassay chambers is often much less than that expected from the amount of PCB's added. Although it is likely some of the PCB's are removed from solution through sorption processes, there are indications that PCB's may be lost through vaporization.

Acetone solutions of Aroclors® 1248, 1260 and 2,2'-dichlorobiphenyl were added to Lake Superior water and the concentrations of each were determined initially and after 1, 2, 4, and 7 days. Glassware samples were taken after 7 days, extracted and analyzed by GLC. The concentration of Aroclors® 1248, 1260 and 2,2'-dichlorobiphenyl decreased nearly 50 percent in 24 hours and nearly 90 percent in one week (Figure 1). One to seven percent of the PCB's could be accounted to being adsorbed onto the glassware. In essence, 83-89 percent of the total Aroclor® could not be accounted for in the water and it was suggested that a considerable amount of Aroclor® entered the atmosphere. Similar results were obtained using Triton X-100 instead of acetone as is illustrated in Figure 2.

Another test was made to check possible air loss of Aroclor® 1016, 0.5 and 1.0 µg of Aroclor® 1016 (in acetone) was added to 100 ml of lake water in a separatory funnel (10 ng/ml). The top of the funnel was fitted with a polyurethane foam plug. Nitrogen was blown through the bottom outlet, the rate of flow controlled with a Teflon® stopcock. Care was taken not to produce droplets of water in the air stream. The polyurethane foam plugs, prior

to use, were consecutively washed with a mixed-polar solvent (85 ml hexane: 15 ml ethyl ether), acetone and hexane. The water/PCB mixture was aerated with nitrogen for two hours, the polyurethane foam plugs removed and extracted. The sample extracts were analyzed by GLC. A clean plug exposed over lake water alone served as the control. Figure 3 illustrates the similarity between the Aroclor® 1016 collected by the foam plugs and the analytical standard of 1016. Of the nine major peaks representing 1016, eight could be confirmed (by retention time) to be collected by the polyurethane foam plugs.

Additional studies to examine the air over large continuous-flow bioassay systems for PCB's were conducted. Two 40 mm polyurethane foam plugs were placed in a glass tube 15 mm ID and 300 mm in length. Sampling tubes attached to vacuum outlets were placed over two continuous-flow bioassay systems using Aroclors® 1248 and 1260 as toxicants. The two systems were in the same room and in close proximity. Air above the diluter and tanks was sampled for a period of three days. The plugs were extracted and analyzed by GLC. The analysis demonstrated the presence of Aroclors® 1248 and 1260 in the plugs. Air sampled above the 1260 diluter contained mostly Aroclor® 1260, although, some Aroclor® 1248 was found and air sampled above the 1248 diluter contained mostly Aroclor® 1248, although, a smaller amount of Aroclor® 1260 was found. Although, variables such as particle sizing, blanks, time or volume of air were not controlled, the preliminary data demonstrates that PCB's being used in continuous-flow bioassays are present in the air as a mixture of individual PCB's closely resembling the PCB in the tank, Figure 4. More detailed studies are underway to confirm preliminary findings.

Figure 1 - See page 6.

Figure 2 - See page 6.

Figure 3 - See page 7.

Figure 4 - See page 8.

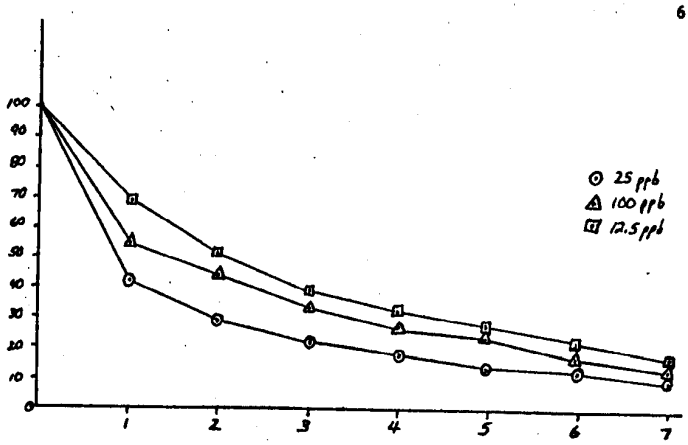


FIGURE 1. Decrease in the water concentration of 2,2'-Dichlorobiphenyl WITH TIME
Ordinate - Ascent of initial PCB
Abcissa - Time (DAYS)

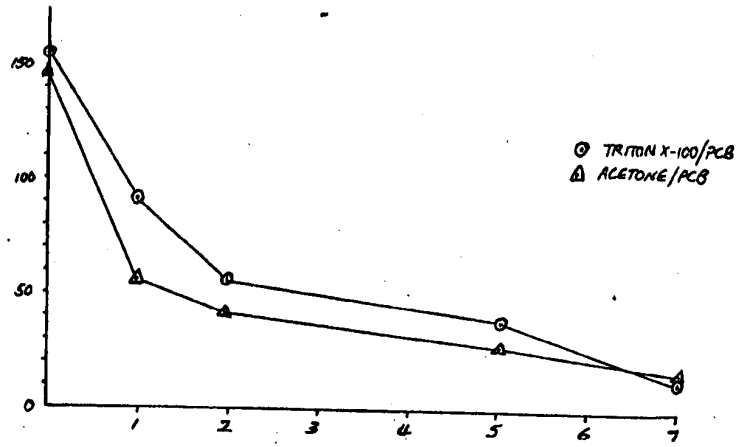
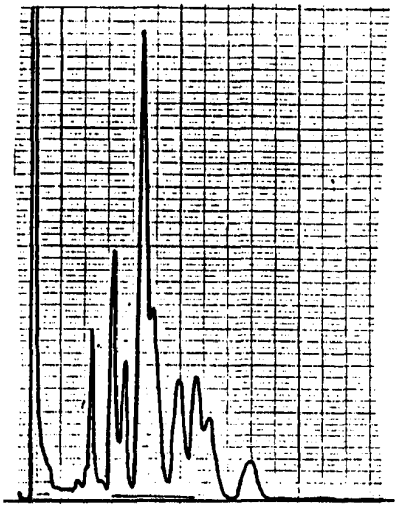
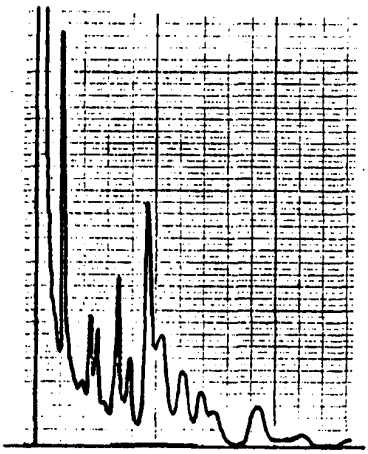


FIGURE 2. Decrease in the concentration of 2,2'-Dichlorobiphenyl when added in a solution of Triton X-100 and acetone
Ordinate - Concentration (µg/L)
Abcissa - Time (Days).



a)



b)

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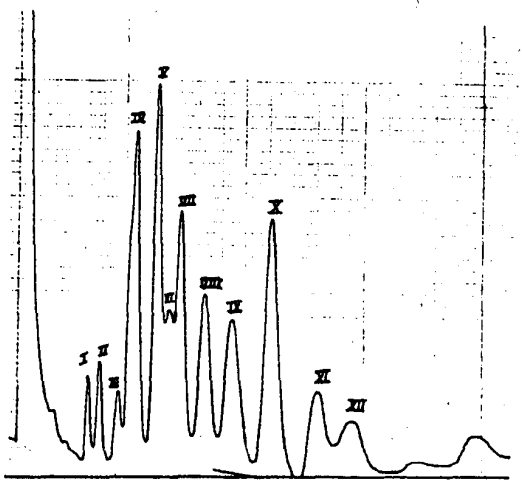
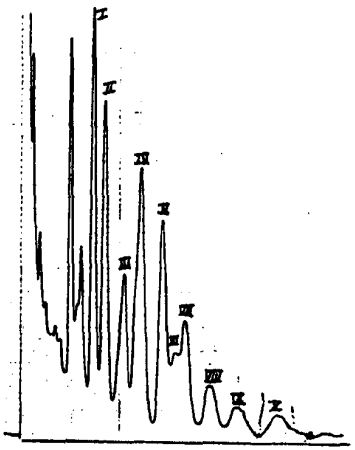
FIGURE 3. a) Chromatogram of a 0.69 ng injection of Aroclor®1016
b) Chromatogram of Polyurethane Plug Extract

GLC conditions: 3% OV-1 6'x¼" glass column on Chromosorb W 60/80 mesh
oven: 180°C Ni63 Detector 50ml/min N_2 flow



a)



b)

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FIGURE #. COMPARATIVE CHROMATOGRAMS OF AROCLOR 1260
a) 0.35 ng 1260 WITH 12 MAJOR PEAKS I-XII
b) EXTRACT FROM POLYURETHANE PLUGS (I-X)
GLC CONDITIONS: 3% OV-1 6'x1/8" GLASS COLUMN CHROMOSORB W 60/80 MESH OVEN 210°C Ni⁶³ DETECTOR 70 ML/MN N_2 FLOW

ENVIRONMENTAL OCCURRENCE AND FATE

Kenneth J. Collyard, Midwest Research Institute, 425 Volker Boulevard, Kansas City, Missouri 64110

PCB Extraction of Sewage Sludge Obtained from Various Areas of Colorado and Kansas City

Wet sewage sludge (mostly anaerobic) was collected from secondary sewage treatment plants in Colorado: Ft. Collins, Greeley, Denver and the Air Force Academy. Along with the Ft. Collins sludge, outfall from the sewage treatment plant (STP) and a river sediment sample 400 yd downstream from the STP was extracted for PCB's.

Sewage Sludge Extraction

Wet sewage sludge was air dried, pulverized and passed through a No. 9 Tyler sieve.

Twenty grams of dry sludge was weighed into a 16-oz French bottle, enough water was added just to saturate the sludge and 200 ml of pesticide quality acetone was added. The matrix was extracted using a high-speed ultrasonic homogenizer, Polytron (Model PT-205T), Brinkman Instrument, Westbury, New York.[1] The Polytron extraction at maximum power for 30 sec eliminates an 8-hr soxhlet extraction. The sludge particles are reduced to a very fine "powder."

The extract is then filtered on No. 42 Whatman paper using a Buchner funnel. Concentrate the filtrate to ca. 50 ml washing the concentrate into a 1L separatory funnel containing 300 ml 1% NaCl solution and extracted with 100 ml hexane twice and twice more with 100 ml benzene. Combine the four extracts in a 1L separatory funnel and wash with 300 ml 1% NaCl solution. Collect any interface that forms and use Na_2SO_4 to dry the extract. Concentrate the extract to ca. 10 ml and elute (200 ml hexane) through a 40 ml Na_2SO_4 bracketed Florisil column prerinsed e hexane.

The eluate was concentrated to 10 ml and placed on a 40 ml Al_2O_3 column (Merck No. 71707) prerinsed ē 75 ml hexane and bracketed by 1 in. of grenular Na_2SO_4. Two-hundred ml hexane was used for elution of any PCB's. Concentrating this extract yielded a 0.25 ml clear viscous solution.

IR Analysis

One milliliter of the 0.25 ml viscous extract was sandwiched between two, 13-mm diameter KBr pellets and placed in both a Beckman IR-33 and IR-20A infrared spectrophotometer. A reference spectrum of Aroclor 1254 was used for comparison.

[1] Johnsen, R. E. and R. I. Starr, Ag. and Food Chem., 20 (1) 48, 1972.

The extract and refarence spectrum were identical except for an aliphatic contaminant in the extract showing intense C-H bands at 3000-2800 cm^{-1}, 1460 cm^{-1} and 1380 cm^{-1}.

Mass Spectrographic Analysis

A Beckman gas chromatograph (Model GC-4) interfaced with a low resolution AEI (Model MS-12) mass spectrometer was used for PCB confirmatory analysis.

Parameters

Column: 7-ft x 0.0625-in. stainless steel 3% OV-1 on Chromsorb W 80/100 mesh
Helium flow rate: 20 ml/min
Inlet: 240° C
Detector FID: 300° C
Separator: 250° C

Column: 50-250° C, 30-min program
Ion source: 250° C at 70 ev potential
Accelerating voltage: 1 Kv
Scan time: 4 sec
m/e: 12-1000

A 0.04 ml sample of the viscous extract was injected on the GLC and temperature programmed to 250° C. Standard 1254 came off between 20 and 36 min with a characteristic isomer peak (ca. 20 peaks) pattern. Mass spectra of the extract was taken on major peaks throughout the run.

The latest and earliest PCB eluting peaks were the heptachloro and tetra chlorobiphenyl isomers, respectively. The intermediate peaks consisted of isomer mixtures.

A direct probe mass spec of this same extract indicated a "clean" spectra (lack of other ionizing species between 200-400 m/e).

Intense fragment ions produced by the consecutive loss of chlorine atoms were quite apparent and the molecular ion was 392.

The clarity of the isomer fragmentation pattern on the GC/MS was not apparent with florisil clean-up alone. Only with the additional Al_2O_3 column clean-up was it possible to get such clean data by IR, GLC/MS.

Gas Chromatography

A 1/100 hexane dilution of the extract was injected onto a Micro-Tek (Model MT-220) gas chromatograph using a ^{63}Ni electron capture detector under the following parameters:

Detector temperature: 300° C
Inlet: 210° C
Transfer: 250° C
Oven temperature: 200° C isothermal
Columns: 6-ft x 1/4-in. glass, 3% Degs, 2% OV-1/3% QF-1

The peak pattern for the extract was almost a perfect match for Aroclor 1254, except for some early "contaminant" peaks. It was surprising to see the almost matched peak height ratios of the PCB isomers to the standard Aroclor. If a sewage plant were at all efficient, some of the isomers should have been partially digested. This was not the case in this sludge sample. The PCB concentration was calculated by area matching with standard Aroclor 1254. The PCB concentration found in the Ft. Collins sludge was 125 ppm.

Coulson Electrolytic and Microcoulometric Analysis

Coulson electrolytic and microcoulometric analysis was also performed on this extract as a comparison of a stoichometric detector to that of electron capture. The early contaminants also showed up using these two detectors but did not interfere with the pattern matching to standard Aroclor 1254.

The river bed soil sample was extracted in the same manner as described above. GLC analysis using an electron capture detector indicated low ppm levels of chlorinated hydrocarbons but not having exact Aroclor patterns. Without confirmatory analysis one can only speculate that these peaks represent partially digested chlorinated-type compounds.

The sewage sludge samples taken from the city of Greeley came from a dry bed approximately 1 year old.

The sewage sludge from Denver and the Air Force Academy was also extracted in the same manner and analysed by EC/GLC only. The Greeley sludge indicated PCB's to be present; however, the lower chlorine-containing isomers were partially digested; however, relative retention times to Aldrin were identically matched to Aroclor 1254. Without mass spec data we do not confirm the results.

The Air Force Academy indicated fewer GLC peaks (less complicated pattern), however, strong electron capturing peaks were obtained and are in the process of being identified by confirmatory methods.

Associated with this work a complete set of chlorinated naphthalene standards were identified by GLC. Even though a complete matching of peak patterns were never observed this does not rule out their presence.

Note:

The cities of Ft. Collins and Greeley each have a population of 40,000 and is nonindustrialized. The question of where 125 ppm of

Aroclor in sludge comes from in a nonindustrialized city along the eastern slope of the Rockies, suggests that PCB contamination is not solely industrial in origin.

Kansas City Sewage Sludge Analysis for PCB's

Five major sewage treatment plants (STP) in the metropolitan Kansas City area were sampled for anaerobic/aerobic sewage sludge, effluent and river sediment.

The same extraction and clean-up techniques used for Colorado sludge was also used on the Kansas City sludge.

Electron capture gas chromatography using ^{63}Ni and basically the same parameters indicate the presences of PCB's in almost all sludge samples.

Gas chromatographic identification of sewage sludge extracts indicated the presence of Aroclor 1254, 1260 and 1262. Confirmatory analysis was obtained on a medium resolution Varian Mat CH-4 mass spectrometer.

Direct probe MS analysis indicated additional clean up was not necessary for PCB identification. This type of information indicates sewage sludge can vary in the degree of contamination. The extract was, therefore, concentrated to a 10 ml volume and passed through a second Al_2O_3 column whereby the first 50 ml of the 200 ml hexane eluting solvent was discarded (containing most of the contaminants) and a mass spec (MS) run on the last 150 ml concentrate. The MS data indicated an octochloro biphenyl molecular ion along with the hepta and hexachlorobiphenyl fragmentation pattern. This additional cleanup was necessary for confirmatory analysis.

Results

Johnson County STP sludge indicates the presence of Aroclor 1262 at a concentration of 80 ppm. The effluent to the STP was calculated at 4 ppb with a 7×10^6 gal/day output. Total PCB contribution from this STP is ca. 1/2 lb/day.

The Platte County STP indicated a PCB concentration of 56 ppm.

A printing company aerobic digester indicated 15.0 ppm PCB's.

The Blue River sewage treatment plant "primary cake," before incineration, contained 21.0 ppm of Aroclor 1254 with indications of isomer digestion.

C. S. Giam, R. L. Richardson, and M. K. Wong, Department of Chemistry, Texas A&M University, College Station, Texas 77843

Chlorinated Hydrocarbon Residues in Gulf of Mexico and Northern Caribbean Biota

In cooperation with the Department of Oceanography, we have just completed a baseline study of chlorinated hydrocarbon pollutants in the Gulf of Mexico and Northern Caribbean marine biota. The project was sponsored by the International Decade of Ocean Exploration of the National Science Foundation.

DDT, its metabolites and polychlorinated biphenyls (PCBs) were found widely distributed throughout the Gulf of Mexico and the Caribbean. However, the level is generally low. Most of the samples that gave higher chlorinated hydrocarbon values were from the coastal areas, indicating the increased pollution level. With the exception of the livers and muscles of some larger fish, e.g. shark and tuna, which have higher concentration of pollutants, the level of chlorinated hydrocarbons in plankton was found to be comparable to that of small whole fish and muscle tissue of larger fish (Table I).

Preliminary analyses of some samples (zooplankton, fish) from the Antarctic also indicated the presence of chlorinated hydrocarbons in most of the samples. The level is very low, about two orders of magnitude lower than that in the biota of the Gulf of Mexico and Northern Caribbean. It is obvious that more samples of similar species need to be taken and analyzed for a better comparison of relative degrees of pollution.

TABLE 1

Concentration of chlorinated hydrocarbons in Gulf of Mexico and Northern Caribbean plankton and fish (parts per billion wet weight)

	Plankton	Small whole fish (6" or less) and fish muscle	Fish liver
Total DDT range	< 0.2-34	1-139	13-1505
Mean total DDT	11	39	282
PCB range	< 3-1055	1-527	18-1300
Mean PCB	94	49	290
Number samples analysed	29	43	13

Richard E. Johnsen, Associate Professor, Pesticide Research Laboratory, Colorado State University, Fort Collins, Colorado 80521

The degradation of Aroclor 1254 and eight PCB isomers in soil alone and soil amended with cattle manure was studied after a one month incubation under flooded conditions in the laboratory. The isomers used were 4,4'-dichloro, 2,5,2',5',-2,6,2'6'-,3,4,3',4'-, 3,5,3',5'-tetrachloro, 2,4,5,2',5'-pentachloro and 2,4,5,2',4',5',- and 2,4,6,2',4',6'-hexachlorobiphenyl. Recoveries were quite variable, ranging from 31-100%, indicating possible selective retention by the soil as found for DDT. There were no indications of any metabolism in this system which when treated with p,p'-DDT degrades almost 100% of the DDT to TDE and lesser amounts of other metabolites.

Additional work in this area is continuing.

R. B. Maybury, Pesticide Residue Unit, Analytical Services Section, Canada Department of Agriculture, Ottawa, Ontario, K1A 0C5, Canada

PCB's in Sewage Sludge, Soils and Crops in Ontario

A survey is being made of the PCB content of sewage sludges being applied as fertilizer to agricultural land in Ontario. The resulting levels of PCB's in the soil are being examined, as well as the possibility of translocation into crops grown on treated land. Results so far have indicated the presence of PCB's in the range 0.1 to 9 ppm in all sludges from sewage treatment plants.

Gilman D. Veith, United States Environmental Protection Agency, National Water Quality Laboratory, Duluth, Minnesota 55804

Current Levels of Chlorobiphenyls in Lake Michigan Fish[1/]

Approximately 1400 fish from 11 regions of Lake Michigan were captured in the fall of 1971 to provide the basis of baseline data for the concentration of major chlorinated hydrocarbons in the 1971 fish, detailed characterization of other trace chemicals in the fish, and preservation for future reference and study. The PCB mixture most commonly observed was similar to Aroclor 1254 although smaller fish from some nearshore waters contained PCB's found in Aroclor 1221 and Aroclor 1242.

Table 1 presents a preliminary summary of the ranges and means of PCB levels for 13 species. The data were averaged over the entire

1/ Much of this study was completed at the University of Wisconsin, Water Chemistry Program, Madison, Wisconsin.

lake and the ranges therefore reflect variations due to region, lipid content, and size of fish. As would be expected, the larger fish with high lipid content, such as the trout and salmon, contain higher concentrations of PCB's ranging from a mean of 8.8 μg/gm (as Aroclor 1254) in brown trout to a mean of 15 μg/gm in chinook salmon. The PCB's in suckers, smelt, and whitefish averaged less than 4.1 μg/gm while the mean concentrations of PCB's in alewives, chubs, carp, and perch ranged between 4.7 and 5.9 μg/gm.

The results of more detailed analysis of data and of the qualitative analyses will be presented in the near future.

TABLE 1

Summary of PCB concentrations in the 1971 Lake Michigan fish

Species	Number of fish analyzed	Concentration of PCB's (μg/gm) as Aroclor® 1254) Range	Mean[a]
Alewife	76	2.5 - 8.9	4.7
Brown trout	16	6.7 - 12	8.8
Carp	42	1.7 - 11	5.5
Chub	229	3.4 - 8.1	5.3
Chinook salmon	21	9.9 - 24	15
Coho salmon	56	3.6 - 17	12
Lake trout	157	8.1 - 21	14
Perch	48	4.2 - 11	5.9
Rainbow trout	10	8.8 - 12	10
Red sucker	16	2.7 - 3.2	2.9
Smelt	35	0.7 - 3.2	2.1
White sucker	50	2.1 - 11	4.1
Whitefish	43	1.5 - 6.1	3.5

[a] Average of all locations sampled.

TOXICITY AND OTHER BIOLOGICAL EFFECTS

Albert M. Gardner, Chemist, Division of Chemical Technology, FDA, Washington, D.C. 20204

Water Soluble Metabolites of PCB's

A brief study was conducted to determine whether PCB's were degraded to water-soluble metabolites in animals. A 2.5 Kg rabbit was given a 10 mg dose of synthetically prepared 2,5,2',5' tetrachloro biphenyl (TCB), a component of Aroclor® 1254. Urine and feces of this rabbit were collected daily for two weeks.

A dihydroxy, dihydro-TCB seemed to be the main water-soluble metabolite of 2,5,2',5' TCB in rabbit urine. Two monohydroxy TCB's were also found in free and conjugated forms in the urine.

The monohydroxy 2,5,2',5' TCB's have been synthetically prepared. Two of these synthetic compounds were identical to the urinary metabolites in gas chromatographic and TLC analyses. Work is now in progress to establish the isomeric identity of the three urinary metabolites.

Robert E. Reinhert, and Wayne A. Willford, Bureau of Sport Fisheries and Wildlife, Great Lakes Fisheries Laboratory, Ann Arbor, Michigan

In November 1972, 18 lake trout from Lake Michigan and the Marquette State Fish Hatchery were stripped of eggs, fertilized, and the eggs transferred to the Great Lakes Fishery Laboratory and the Hammond Bay Biological Station. The 25,000+ eggs were divided into lots containing approximately 300 eggs from each fish for incubation at either a constant temperature (6° C), at a temperature cycle similar to that of Lake Michigan, or in water taken directly from Lake Huron.

Eggs are being analysed for PCB's and DDT residues and observations on the hatching success will be continued until complete absorption of the yolk sac has occurred. The embryological development of the eggs is also being studied.

David L. DeFoe, United States Environmental Protection Agency, National Water Quality Laboratory, Duluth, Minnesota

Two fathead minnow life cycle exposures with PCB Aroclors® 1248 and 1260 are in the ninth and final month. Spawning is expected to terminate by mid-January. Toxicant concentrations are approximately 3, 1, 0.33, and 0.11 ppb.

The three objectives of this experiment are:

1. To determine the no effect level of these Aroclors®, on a full life cycle of the fathead minnow. The parameters used are survival, growth, and reproduction.

2. To determine biological magnification of PCB's. Samples were taken at periodic intervals throughout the life cycle to determine PCB accumulation.

For a detoxification study, the excess fish from the 60 day thinning were placed in a clean water system for approximately 60 days with 14 day sampling intervals.

3. To investigate the effect of the solvent (acetone) on the exposure system.

Acetone was the solvent used as the delivery media for the PCB's. Because of this, two types of controls were used. One set of duplicate control tanks received the same amount of acetone as did the toxicant tanks (approximately 2 ppm). The second set of controls received only clean water.

There appears to be an effect on the algal and bacteria growth. This could have an effect on some test organisms, especially those which show grazing characteristics.

The analysis of the data on survival, growth and reproduction will be complete in several months.

Tissue residue analysis will begin in February.