

These same tissues were analyzed for PCB's, another group of long-life, widespread chemicals in the environment (BioScience 20, 958-964, 1970), and these results are found in Table 2. In terms of wet weight the highest values were found to occur in lobster egg masses. The lobster hepatopancreas and carcass also contained large amounts of PCB's. The eel had about 1 ppm of these materials whereas the hagfish had about one-half this quantity. As in the case of DDT, the fundulus and flounder had comparable levels. In terms of PCB's on a fat weight basis, there is extensive concentration observed in this compartment. The egg masses again contained the highest concentration followed by the flounder, eel, lobster carcass, and hagfish. The use of DDT/PCB ratios has been proposed as a means of determining the natural history of contamination of marine species (Bull. Env. Contam. and Tox. 7, 223-228, 1972). According to this concept, ratios of unity suggest that contamination of the marine environment by these two chlorinated hydrocarbons occurred at the same point in time. Higher ratios thus suggest initial contamination by DDT, low ratios recent contamination by PCB's. The low ratios indicated on the bottom of Table 2 imply that the intake of these materials for the lobster, a scavenger, is from materials contaminated more recently with PCB's than DDT's. The DDT/PCB ratios for all other species in question do not appear to be significantly far enough from unity to suggest anything other than simultaneous contamination by these pollutants.

1972 #3

DISTRIBUTION OF ^{14}C -2,4,5,2',5'-PENTACHLOROBIPHENYL IN THE LOBSTER, *Homarus americanus* AFTER INTRAVASCULAR ADMINISTRATION

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The polychlorinated biphenyls (PCB) possess high thermal and chemical stability in addition to a high dielectric constant. Due to these properties substantial quantities of PCB are used industrially. PCB residues are fairly widespread in the environment and have been detected in human adipose tissue (Price and Welch, Env. Hlth. Persp. 1, 73, 1972). Commercially available PCB are complex mixtures containing many isomers of varying chlorine content. For example, Aroclor 1254 has been separated into more than fifty components, of which the pentachlorobiphenyl isomers appear to be the chief constituents (Sissons and Welti, J. Chromatog. 60, 15, 1971). The distribution of radio-labeled 2,4,5,2',5'-pentachlorobiphenyl was investigated in the lobster since the data obtained should be representative of several, if not all, Aroclor 1254 components. In addition, the results could be compared with those reported earlier for DDT disposition in the lobster (Guarino, Pritchard, Call, Bering, Rall, Bull MDIBL 11, 29, 1971).

Adult lobsters of mixed sex weighing 360-560 g were purchased locally (Lunt's Lobster Pound, Trenton, Maine). 2,4,5,2',5'-Pentachlorobiphenyl - (2',5'-dichlorophenyl- ^{14}C) (^{14}C -PCB), 10.08 mCi/mM was obtained from Mallinckrodt. ^{14}C -PCB (0.20 mg/kg; 14.0×10^{12} dpm/kg) was injected into the pericardial sinus of the lobster as a solution in Emulphor-water (18 percent v/v).

All lobsters (n=14) were dosed on the same day and were returned to aquaria containing fresh circulating sea water (15°). Six animals were sacrificed 24 hours after treatment and the survivors (seven) were sacrificed one week after dosing. The "one week" lobsters were fed small pieces of mackerel starting 48 hours after treatment and terminating 48 hours before sacrifice. Plasma was removed from the pericardial sinus just prior to sacrifice, the dead animals were dissected, the tissues weighed and aliquots of plasma (100 μ l) or tissue (100-200 mg) were solubilized by digestion in 2 ml NCS (Amersham/Searle). Liquid scintillation solvent (18 ml; 5.0 g PPO and 250 mg POPOP/liter toluene) was added and the radioactivity determined in a liquid scintillation spectrometer. Counting efficiency varied from 62-80 percent for the biological samples. Recovery of ^{14}C -PCB administered to lobsters was determined by homogenizing the muscle tissue dissected from the lobster's carcass exclusive of fluids and organs sampled directly. Normally, 60-80 percent of the radioactivity injected was recovered.

TABLE 1
 SPECIFIC ACTIVITY (dpm/mg) OF VARIOUS TISSUES
 OF THE LOBSTER FOLLOWING INTRAVENOUS ADMINISTRATION
 OF ^{14}C -2,4,5,2',5'-PENTACHLOROBIPHENYL (0.20 mg/kg)

Tissue	dpm/mg tissue or/ μ l plasma Time after administration	
	24 hours	7 days
Hepatopancreas	183.4 \pm 24.7 (6)*	192.9 \pm 33.4 (7)
Green Gland	20.1 \pm 13.0 (6)	8.4 \pm 2.4 (7)**
Intestine	14.0 \pm 7.0 (6)	14.3 \pm 5.9 (7)
Heart	8.8 \pm 5.4 (6)	2.7 \pm 0.4 (7)**
Tail Muscle	5.1 \pm 1.8 (6)	1.6 \pm 0.1 (7)**
Claw Muscle	5.0 \pm 3.1 (6)	1.5 \pm 0.2 (7)**
Stomach	4.5 \pm 1.1 (6)	4.8 \pm 2.1 (7)
Gill	3.4 \pm 1.8 (6)	3.2 \pm 1.5 (7)
Male Gonad	8.3 (1)	1.6 (1)
Egg Masses	16.6 \pm 9.7 (5)	7.7 \pm 6.6 (6)
Plasma	1.33 \pm 0.41(6)	0.49 \pm 0.17(7)**

* Mean \pm S.D. (N)

** Significantly different from 24 hour ($P < 0.01$)

As shown in Table 1, the specific activity of the hepatopancreas was much higher than for any other individual organ at both assay times: 24 hours and seven days after PCB administration to the lobsters. This data also emphasizes the persistence of 2,4,5,2',5'-pentachlorobiphenyl in the hepato-

pancreas after one week since there is no significant decrease of the specific activity of the organ (Table 1) or in the percentage of the administered dose in this organ (Table 2). Similar results were noted for the intestine, stomach, gill, and egg masses. On the other hand, some organs and plasma showed a statistically significant decrease in radioactive content over the time period studied. Included in this category were the green gland, heart, tail muscle, and claw muscle. These observations suggest a redistribution or excretion of 2,4,5,2',5'-pentachlorobiphenyl or its metabolite(s) over a one-week period in the lobster. Fecal matter removed from the intestines of both 24-hour and seven day animals (during dissection) contained significant amounts of radioactivity, suggesting that some of the IV administered compound was being excreted by this route. Although the hepatopancreas has not yet been checked for oxidative metabolites of ^{14}C -PCB, it appears unlikely that significant levels of such metabolites are present since lobster gill and hepatopancreas subcellular fractions (10,000 x g supernatant and microsomes) possess negligible benzphetamine demethylase or aniline hydroxylase activity (Bend, Pohl, Fouts, Bull MDIBL, 1972). Moreover, it was previously reported that more than 90 percent of the radioactivity remaining in the hepatopancreas of the lobster following DDT administration was the parent insecticide rather than metabolites (Bull. MDIBL 11, 29, 1971).

TABLE 2
% ADMINISTERED RADIOACTIVITY REMAINING IN LOBSTER
AFTER PERICARDIAL INJECTION OF
 ^{14}C -2,4,5,2',5'-PENTACHLOROBIPHENYL (0.20 mg/kg)*

Tissue	Time after injection	
	24 hours	7 Days
Hepatopancreas	61.4 ± 8.3 (6)	60.5 ± 10.8 (7)
Green Gland	0.19 ± 0.12(6)	0.07 ± 0.01(7)**
Intestine	0.10 ± 0.09(6)	0.11 ± 0.05(7)
Heart	0.09 ± 0.04(6)	0.03 ± 0.01(7)**
Tail Muscle	4.36 ± 0.84(6)	1.57 ± 0.26(7)**
Stomach	0.36 ± 0.14(6)	0.53 ± 0.39(7)
Gill	0.61 ± 0.31(6)	0.59 ± 0.31(7)
Male Gonad	0.06	0.02
Egg Masses	0.48 ± 0.34(5)	0.37 ± 0.56(6)
Mean Total Recovery	67.59%	64.77%

* Mean ± S.D. (N)

** Significantly different from 24 hours ($P < 0.01$)

The slow clearance of 2,4,5,2',5'-pentachlorobiphenyl by the lobster after a single parenteral dose is not surprising, in view of the known persistence of PCB residues and their accumulation in

the food chain. In order to estimate the half-life of this compound in the lobster it will be necessary to study tissue levels for a prolonged time period after administration.

1972 #4

SOME PROPERTIES OF THE MICROSOMAL DRUG-METABOLIZING ENZYME SYSTEM IN THE LITTLE SKATE, *Raja erinacea*.

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The marine ecosystem is contaminated with pesticides, industrial chemicals, and other foreign organic chemicals. Consequently the ability of marine animals to metabolize xenobiotics is of interest. We studied xenobiotic metabolism in hepatic and gill tissues of some marine species indigenous to the Frenchman Bay area of Maine including the rock crab (*Cancer borealis*), lobster (*Homarus americanus*), little skate (*Raja erinacea*), dogfish shark (*Squalus acanthias*), and winter flounder (*Pseudopleuronectes americanus*) and found that aniline hydroxylase and benzphetamine demethylase specific activities were significantly higher in fortified liver microsomes (Table 1) from skate than for winter flounder or dogfish shark. Negligible activities for aniline and benzphetamine metabolisms were found in gill tissue of all species studied and in hepatopancreas from lobster and crab. Since the little skate possessed readily detectable levels of aniline hydroxylase and benzphetamine demethylase, some of the characteristics of these enzyme systems were investigated using the liver microsomal fraction.

TABLE 1
COMPARISON OF LIVER MICROSOMAL ANILINE HYDROXYLASE AND (d)-BENZPHETAMINE DEMETHYLASE ACTIVITIES IN THE LITTLE SKATE, WINTER FLOUNDER AND DOGFISH SHARK

Species	Specific Activity (nmoles product formed/min/mg microsomal protein)	
	aniline hydroxylase	(d)-benzphetamine demethylase
Little skate (<i>Raja erinacea</i>)	0.40*	1.41
Winter flounder (<i>Pseudopleuronectes americanus</i>)	0.03	0.11
Dogfish shark (<i>Squalus acanthias</i>)	0.07	0.27

*Data are from a single, representative experiment.