DEPURATION OF HIGHLY TOXIC CHLORINATED COMPOUNDS BY THE AMERICAN OYSTER *CRASSOSTREA VIRGINICA*: A FIELD STUDY

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ABSTRACT

Depuration of toxic chlorinated compounds such as the planar 3,3',4,4'-tetrachlorobiphenyl (77); 3,3',4,4',5-pentachlorobiphenyl (126) and 3,3',4,4',5,5'-hexachlorobiphenyl (169), 2,3,7,8-tetrachlorodibenzo -p-dioxin (TCDD) and 2,3,7,8-tetrachlorodibenzofuran (TCDF) was monitored during a 50 day field study, during which american oysters (Crassostrea virginica) were transplanted from a heavily polluted area (Hosuton Ship Channel) to a relatively non impacted area (Hanna's Reef). Estimated half-lives for planar PCB congeners 77 and 126 were 88 and 107 days respectively. Estimated half-lives for TCDD and TCDF were 33 and 37 days, respectively. Compared with ortho-substituted PCB congeners of the same chlorination level, these toxic halogenated compounds take longer to depurate from the oysters.

INTRODUCTION

The general concern about the occurrence of highly toxic PCB congeners, i.e. the planar 3,3',4,4'-tetrachlorobiphenyl (77); 3,3',4,4',5-pentachlorobiphenyl (126) and 3,3',4,4',5,5'-hexachloro-biphenyl (169), 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) and 2,3,7,8-tetrachlorodibenzo-*p*-furan (TCDF) in different environmental compartments is associated with their potential adverse environmental and human health effects. These compounds elicit a diverse spectrum of toxic and biochemical responses including body weight loss, dermal disorders, liver damage, thymic atrophy, reproductive toxicity and immunotoxicity and the induction of CYP1A1 and CYP1A2 gene expression [1,2,3,4]

The objective of this paper is to discuss the depuration of highly toxic planar compounds by the american oyster, *Crassostrea virginica*, and to evaluate the possible health risks that the consumption of contaminated bivalves allowed to depurate in clean waters might pose to human beings.

MATERIALS AND METHODS

Approximately 150 oysters were collected from a known contaminated area near the Houston Ship Channel in Galveston Bay and transplanted to a relatively pristine area, Hanna Reef (Figure 1). Oysters were sampled in groups of 20 individuals during the 3rd, 6th, 18th, 30th, and 50th days after transplantation.

Planar PCB Congeners

The analytical procedure used for the extraction, initial fractionation and cleanup of oyster tissue samples for aliphatic and aromatic (PAHs) hydrocarbons, polychlorinated biphenyls (PCBs), including planar congeners, and chlorinated pesticides analyses have been fully described elsewhere. [5] Before proceeding with the isolation of planar PCB congeners, PCB #81 was added to the extracts as internal standard.

Planar PCBs were isolated from *non-ortho* PCB congeners and other chlorinated compounds using glass chromatographic columns packed with 2 g of the adsorbent, a 1:20 mixture of AX-21 activated charcoal and LPS-2 silica gel. The column was eluted with 50 ml of 1:4 methylene chloride:cyclohexane, 30 ml of 9:1 methylene chloride:toluene, and 40 ml of toluene. [6]

Planar PCB congeners were analyzed by fused-silica capillary column GC-ECD (Ni^{63}) using a Hewlett Packard 5880A GC in splitless mode as previously described. [6] Detection limits for individual planar PCB congeners, calculated on the basis of 2 grams (dry weight) sample size with 2% by volume of the extract injected into the GC-ECD, was 50 pg g^{-1} dry weight.

Dibenzo-*p*-dioxins and Dibenzo-*p*-furans

The analytical procedure used for the extraction, clean-up and isolation of polychlorinated dibenzo-*p*-dioxins (PCDD) and polychlorinated dibenzofurans (PCDF) have been published elsewhere.[7] Briefly, oyster tissues were extracted with methylene chloride and concentrated to 100 ml in hexane. To eliminate interferences caused by lipidi material, the hexane extract was treated for 2 h. with a mixture of concentrated sulfuric acid and silica gel. After filtration and concentration, the sample extract was further purified by mixed-bed silica column chromatography (8 g of 44% sulfuric acid/silica gel and 4 g of 33% 1 N sodium hydroxide/silica gel) and basic alumina column chromatography.

Sample extracts were fractionated using a low pressure Michael-Miller type chromatographic columns fitted with removable Teflon threaded couplings, stopper, an adjustable flow valve and a 250 ml glass solvent reservoir. The column was packed with 2 g of the 20:1 charcoal/silica gel adsorbent mixture and sequentially eluted with 60 ml of 1:4 methylene chloride:cyclohexane, 40 ml of 1:9 methylene chloride:toluene and 25 ml of toluene to isolate the mono- and non-*ortho* PCB congeners respectively.

PCDD and PCDF are collected by eluting the flipped column "backwards" with 200 ml of toluene. TCDD and TCDF were analyzed by a high resolution gas chromatograph coupled to a high resolution mass spectrometer (HRGC/HRMS).

RESULTS AND DISCUSSION

The concentrations of three highly toxic planar PCB congeners, i.e., PCB 77, 126 and 169, and TCDD and TCDF in transplanted oysters are summarized in Table 1. Except for PCB 169, the concentrations of these compounds measured in oysters from the Houston Ship Channel area were in the low part per trillion (pg g^{-1}) to part per billion (ng g^{-1}) range. PCB 169 was present at concentrations near or below the detection limit. When transplanted to the Hanna Reef area, the oysters slowly depurated the planar PCB congeners, TCDD and TCDF. These compounds were still present at relatively high concentrations by the end of the 50-days depuration period (Figure 2).

Kinetics parameters describing the depuration of planar PCB congeners, TCDD and TCDF can be calculated according to the first-order equation:

$$dC_t/d_t = k_u C_w - k_d C_t \quad (1)$$

where C_t is the concentration of the analyte in the tissue at time = t, C_w is the concentration in water and k_u and k_d are the uptake and depuration constants, respectively. If the concentration in the water at the depuration site is regarded as zero, i.e., $C_w = 0$, the equation can be reduced to:

$$dC_t/dt = - k_d C_t \quad (2)$$

or

$$\text{Log } C_t = \log C_o - k_d t / 2.303 \quad (3)$$

From this equation, the relationship to calculate the biological half-life ($t_{1/2}$) can be deduced:

$$t_{1/2} = 0.693 / k_d \quad (4)$$

Depuration rate of congener 77 was higher than the rate observed for congener 126. The estimated depuration constants (k_d) for congeners 77 and 126 were 0.0079 and 0.0064 $days^{-1}$, respectively. These values are lower than the range of values observed for other PCB congeners within the same homolog group. Compared to *ortho*-substituted congeners in the corresponding homolog groups, planar PCBs take longer to equilibrate out of the lipid pools of these organisms. Estimated half-lives for congeners 77 and 126 are 88 and 107 days, respectively. The depuration constants for TCDD and TCDF were similar (0.0210 and 0.0189 $days^{-1}$, respectively). Estimated half-lives are 33 and 37 days for TCDD and TCDF, respectively.

The long time needed for these highly toxic compounds to be depurated might be of significant importance in projects such as the Mississippi oyster relaying effort designed to transplant oysters from polluted to clean waters before harvesting them for human consumption [8]. Bohem and Quinn[9] reported that transplanting of shellfish from polluted to cleaner environments has been used as a means of increasing harvestable yields of the hard shell clam (*Mercenaria mercenaria*) in Narragansett Bay, Rhode Island. However, oysters that are allowed to depurate in a clean environment might show lower concentrations of organic contaminants but still maybe high enough to pose a threat for human consumption.

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	Concentration (pg/g dry weight)					
Compound	Day 0	Day 3	Day 6	Day 18	Day 30	Day 50
3,3',4,4'-Tetrachloro-biphenyl PCB 77	980	900	800	750	740	630
3,3',4,4',5-Pentachloro-biphenyl PCB 126	220	170	250	210	190	150
3,3',4,4',5,5'-Hexachloro-biphenyl PCB 169	96	106	ND	76	ND	ND
2,3,7,8-Tetrachlorodibenzo-p-dioxin TCDD	50	47	38	29	24	17
2,3,7,8-Tetrachorodibenzofuran TCDF	169	132	124	98	82	60
TOTAL TEQ[a]	104	91	83	71	59	44

Table 1.- Concentration of toxic chlorinated compounds in the american oyster (*Crassostrea virginica*) during the depuration experiment. (ND= non detected); a: based on Safe, 1990 [3)]

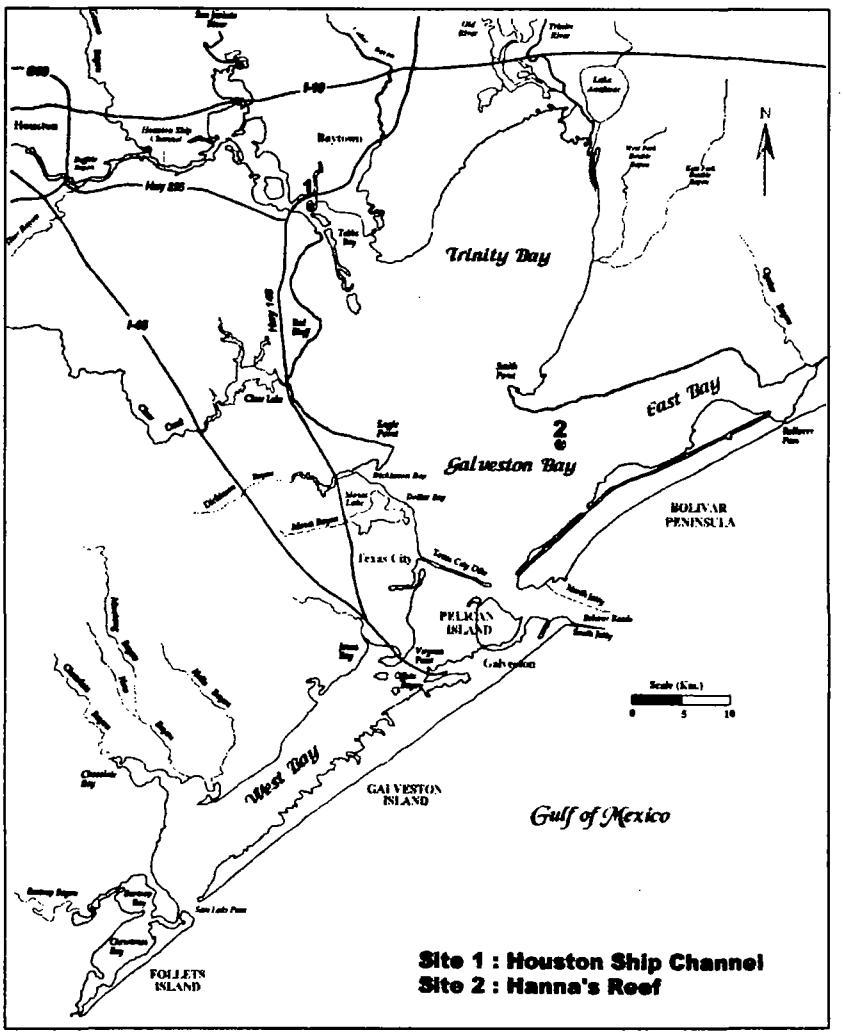

Figure 1.- Map of Galveston Bay, Texas showing the locations of the oyster transplant.

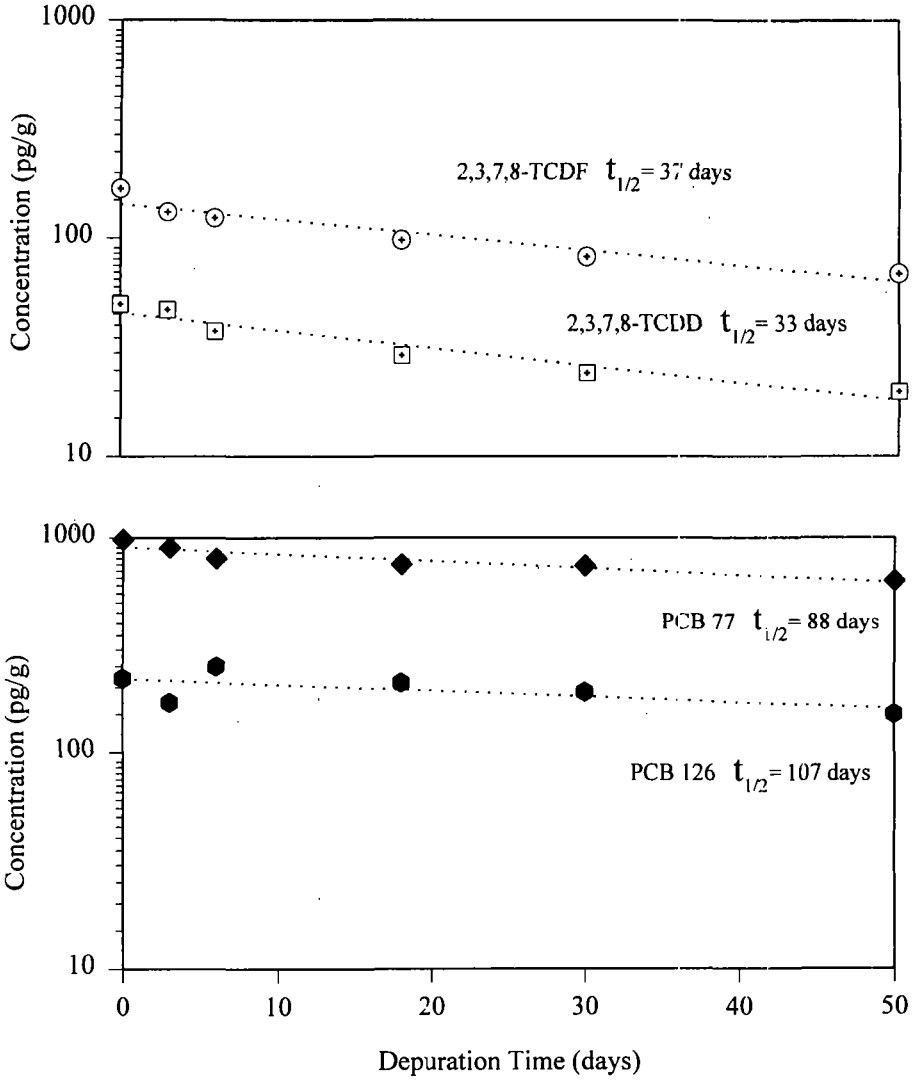

Figure 2.- Concentrations of toxic chlorinated compounds in the american oyster during the 50 day depuration experiment.