

Residues of Toxaphene in Finfish and Shellfish from Terry and Dupree Creeks, Georgia, U.S.A.

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ABSTRACT: To better characterize human health risks associated with potentially contaminated seafood, 56 composite samples of edible tissue of several finfish and shellfish species were analyzed for residues of toxaphene using gas chromatography with electron capture and negative ion mass spectrometric detection (GC-ECD and GC-ECNI-MS). Toxaphene in these samples, collected in 1997 near a former toxaphene plant in Brunswick, Georgia, were previously reported as non-detectable using non-selective techniques. Estimated total toxaphene concentrations (Σ TOX) ranged from less than 0.01 to 26 $\mu\text{g g}^{-1}$ on a wet tissue basis. Smaller, bottom dwelling finfish such as croaker, mullet, and spot exhibited the highest Σ TOX (0.76–26 $\mu\text{g g}^{-1}$), larger predatory fish including seatrout contained intermediate levels (0.08–4.4 $\mu\text{g g}^{-1}$), and shellfish (blue crab and shrimp) contained the lowest levels (< 0.01 to 0.27 $\mu\text{g g}^{-1}$). For a given species, samples from the site furthest from the toxaphene plant had lower Σ TOX than samples from the other 3 sites. On a congener specific basis, levels ranged from < 0.0025 to 3.5 $\mu\text{g g}^{-1}$. Congener distributions were, in general, dominated by 2-exo,3-endo,6-exo,8,9,10-hexachlorobornane (Hx-Sed) and 2-endo,3-exo,5-endo,6-exo,8,9,10-heptachlorobornane (Hp-Sed), breakdown products of Cl_8 - Cl_{10} toxaphene homologs. Other prominent congeners confirmed by GC-ECNI-MS included Parlar numbers 26, 40/41, 42, 44, 50, 62, and 63, as well as several unidentified Cl_6 - Cl_9 homologs. Minor differences in congener distribution among species and sampling locations suggested that exposure regimes and/or intrinsic biotransformation capabilities were not uniform. These results indicate that toxaphene residues were detectable in all species surveyed and at concentrations higher than estimated previously.

Introduction

Toxaphene is the trade name of a complex organochlorine pesticide formulation manufactured in the United States by Hercules, Inc. at their Brunswick, Georgia facility. Technical grade toxaphene was produced by exhaustive chlorination of camphene, resulting in a complex mixture of several hundred Cl_6 - Cl_{10} monoterpene compounds with an average chlorine content of 70% by weight (Saleh 1991). Due to its physicochemical and toxicological properties, toxaphene has the potential to accumulate locally in sediment (Veith and Lee 1971) and biota (Pollock and Kilgore 1978) and exert toxic effects in the aquatic environment (U.S. Environmental Protection Agency 1980). Residues of toxaphene are also transported over long distances via the atmosphere (Stanley et al. 1971) and as a result are detected in biota in remote regions such as the Arctic (Bidleman et al. 1993). Although it was banned in the U.S. and other countries during the 1980s, toxaphene continues to be an environmental quality issue at local, regional, national, and international levels (Agency for Toxic Substances and Disease Registry 1998;

U.S. Environmental Protection Agency 1998; de Geus et al. 1999).

In the aquatic environment, in situ processes act to modify the distribution of individual congeners in technical toxaphene. A pronounced shift toward lower chlorinated homologs was first reported in reducing salt marsh sediments spiked with toxaphene (Williams and Bidleman 1978). In sediments from lakes collected decades after treatment with toxaphene, the two most abundant toxaphene residue congeners were identified as 2-exo,3-endo,6-exo,8,9,10-hexachlorobornane and 2-endo,3-exo,5-endo,6-exo,8,9,10-heptachlorobornane (Stern et al. 1996). These compounds, also known as Hx-Sed and Hp-Sed, respectively, are only very minor components of virgin toxaphene. Experimental evidence indicated that Hx-Sed and Hp-Sed result from dechlorination of Cl_8 and Cl_9 toxaphene components under reducing conditions (Fingerling et al. 1996). These weathered, sediment-associated residues of toxaphene are also available to biota, including forage and predator fish (Miskimmin et al. 1995; Maruya and Lee 1998). In heavily contaminated systems (Calero et al. 1993; Maruya et al. 2000), the complex toxaphene residue patterns in sediment and tissue are in contrast to samples from remote areas, where the pattern is characterized by fewer, recalcitrant compounds (Alder and Vieth 1996).

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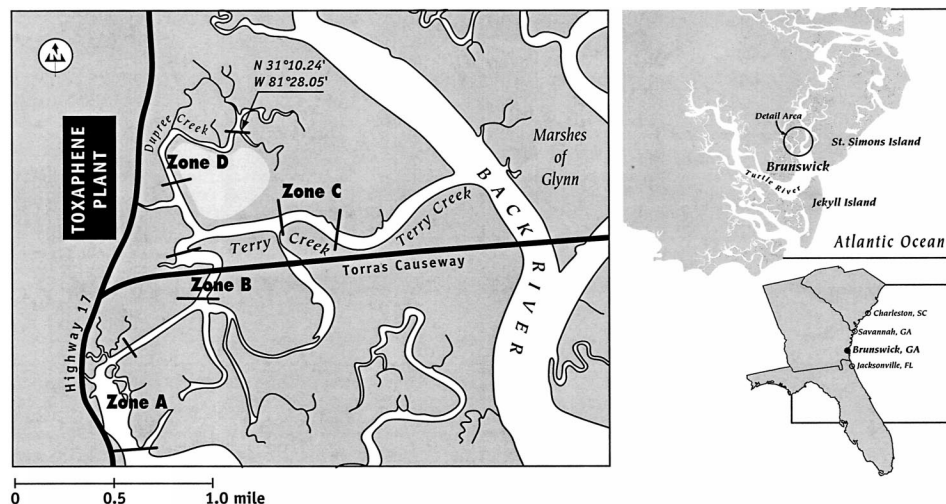


Fig. 1. Map of study area, Terry and Dupree Creeks marsh, near the Hercules Inc. facility in Brunswick, Georgia. Samples were collected in each of four zones (A–D).

Gas chromatography with electron capture detection (GC-ECD), the basis for standard protocols such as U.S. Environmental Protection Agency (1996) Method 8081, offers good sensitivity and repeatability for organochlorine residues, including toxaphene. At the same time, however, this method suffers from poor selectivity, and hence, specificity for toxaphene. The utility of GC-ECD as a sole technique for the characterization of toxaphene residues based on unmodified technical mixtures as standards of reference is further compromised by profound changes in congener profiles. In recent years, the application of electron capture negative ionization mass spectrometry (GC-ECNI-MS) and the commercial availability of pure standards have allowed analysts to better characterize toxaphene contamination (Muir and de Boer 1995). Thus, GC-ECNI-MS used in conjunction with GC-ECD can help distinguish toxaphene residues from other common contaminants (e.g., PCBs, chlordanes, and DDTs).

In 1997, the Georgia Department of Natural Resources conducted a study in the Brunswick area to determine the levels of toxaphene in several species of finfish and shellfish consumed by humans (Georgia Department of Natural Resources 1997). Such information was needed to better assess human health risks due to toxaphene contamination. Specimens of each target species were collected from several tidal creek sites near the former toxaphene facility, composited, and analyzed using protocols based on U.S. Environmental Protection Agency (1996) Method 8081. As defined by a consortium of regulatory and industry representatives (Carlin et al. 2000), toxaphene in each and every one of these samples was reported as not de-

tectable. In this study, we combined GC-ECD with GC-ECNI-MS and re-analyzed these samples to provide more definitive estimates of toxaphene residue levels and also to determine the concentrations of a suite of individual toxaphene congeners.

Materials and Methods

SAMPLE COLLECTION AND COMPOSITING

In the spring of 1997, blue crab (*Callinectes sapidus*), shrimp (*Penaeus* sp.), and nine different species of finfish were collected in four zones (A–D) of the Terry-Dupree Creeks estuarine marsh near the Hercules, Inc. facility in Brunswick, Georgia (Fig. 1). Using gill nets and/or baited wire traps, up to 15 individual fish/blue crab per species were collected in each zone, measured, and tagged. To maintain size similarity, the total length of smallest individual was > 75% of the total length of the largest individual. Shrimp (15–20 individuals; 300–400 g total) were collected in each zone using cast nets and placed in re-sealable plastic bags. All specimens were kept on ice. After return to shore, edible tissue consisting of scaled, skin-on filets or blue crab lump and claw meat from 2–5 individuals were processed and pooled into 1–3 composite samples per zone for each species (Table 1). For shrimp, heads and exoskeletons were removed. Pooled tissues were then ground using a food processor, transferred to clean glass jars, and frozen. A total of 56 composite samples were delivered in ice-filled coolers to the Skidaway Institute of Oceanography for analysis. Upon receipt, samples were immediately unpacked and placed in a freezer.

TABLE 1. Number of composite edible tissue samples by species and by location. See Fig. 1 for location of zones.

Species	Zone	No. Composite Samples
Blue crab (<i>Callinectes sapidus</i>)	A, B, D	3 per zone
	C	2
Shrimp (<i>Penaeus</i> sp.)	A, B	1 per zone
	C	3
	D	2
Atlantic croaker (<i>Micropogonias undulatus</i>)	B, D	2 per zone
Striped mullet (<i>Mugil cephalus</i>)	A, B, D	3 per zone
Red drum (<i>Sciaenops ocellatus</i>)	A, B, D	1 per zone
Spotted seatrout (<i>Cynoscion nebulosus</i>)	A	3
	B	2
	C, D	1 per zone
Spot (<i>Leiostomus xanthurus</i>)	A, D	3 per zone
	B	1
	C	2
Yellowtail (<i>Bairdiella chrysoura</i>)	A	3
Black drum (<i>Pogonias cromis</i>)	A	1
Flounder (<i>Paralichthys</i> sp.)	D	1
Whiting/kingfish (<i>Menticirrhus</i> sp.)	D	1

SAMPLE PROCESSING

For each sample, a 5 to 20 g aliquot of thawed tissue was homogenized with ~50 g of kiln fired, anhydrous Na_2SO_4 using a solvent-rinsed, ceramic mortar and pestle. This mixture was placed in a glass thimble and extracted with 400 ml CH_2Cl_2 in a glass Soxhlet apparatus for ≥ 16 h. The CH_2Cl_2 extract was transferred to a glass ampoule/flask/Snyder column apparatus and reduced to < 10 ml using a heated water bath. The extract was then evaporated to dryness in a fume hood and extractable lipid determined gravimetrically to the nearest mg using a microbalance. The sample was redissolved in hexane and applied to a 500 (L) $\times$ 11 mm (OD) glass column packed with 18.0 g 1.0% water deactivated Florisil (60–100 mesh chromatographic grade, Fisher Scientific, Fair Lawn, New Jersey), prepared as described in detail in Maruya and Lee (1998). To minimize breakthrough of lipid, a maximum of 200 mg extractable lipid was applied to each Florisil column. Two fractions were collected from Florisil, the first fraction (F1) was eluted with 50–80 ml hexane to capture PCBs and other nonpolar interferences, and the second (F2) was eluted with 150 ml of a 20% (v/v) mixture of CH_2Cl_2 and hexane to capture toxaphene residues. Both fractions were reduced and F2 exchanged to hexane using a TurboVap II (Zymark Inc., Hopkington, Massachusetts). Water bath temperature and N_2 ($> 99.99\%$) pressure were maintained at 55°C and 8 psi, respectively. Extracts in hexane were reduced to 1.0 ml and transferred to amber 2-ml glass vials sealed with Teflon-lined silicone rubber septa for subsequent GC analysis.

GAS CHROMATOGRAPHY

F1 and F2 extracts (1 μl) were injected into a Varian 3400CX GC-ECD with an 8200 autosampler.

Varian Star chromatography software (version 4.01) was used to acquire and analyze the chromatographic runs. A 30 (L) $\times$ 0.25 mm (OD) fused silica capillary column coated with 0.25 μm DB-5 was used to separate toxaphene components (J&W Scientific, Folsom, California). The GC oven was programmed as follows: 120°C (hold 1 min); ramp to 260°C at 2°C min^{-1} (hold 15 min). The injector and detector were maintained isothermal at 270°C and 330°C , respectively. The total run time was 86 min. To confirm the presence of individual toxaphene analytes, extracts of at least one sample per species were analyzed on a Hewlett Packard 6890 Series II GC coupled to a 5973 mass selective detector operating in the ECNI mode. Fused silica columns, 30 (L) $\times$ 0.25 mm (OD), coated with either 0.25 μm of DB-5 or DB-XLB were used for this analysis. Methane at a pressure of ~1 torr was used as the moderating gas. The quadrupole mass spectrometer (MS) and ion source were maintained at 106°C and 150°C , respectively. The MS was scanned between 200–500 daltons at $1.3 \text{ cycles s}^{-1}$.

Concentrations of individual toxaphene congeners were determined by GC-ECD based on a 22-component mixture of chlorinated monoterpenes (TM2, Dr. Ehrenstorfer, Augsburg, Germany). The 17 chlorobornane and 5 chlorocamphene congeners in TM2 represent $\text{Cl}_6\text{--Cl}_{10}$ homologs (Table 2). Serial dilutions of TM2 ranging from 2–100 pg in hexane were used to generate a 6-point calibration curve and to compute mean response factors. Concentrations of several prominent congeners not in TM2, including Hx-Sed and Hp-Sed (Table 2) and Cl_7 and Cl_8 compounds whose structures are presently unknown (Hp-1, Oc-1, Oc-2, and Oc-3; Fig. 2) were estimated using the mean response factor

TABLE 2. Structure, homolog, and chromatographic properties of individual chlorinated monoterpene (toxaphene) target analytes. Structural formulas are for chlorobornanes except for P11, 12, 15, 25, and 31 which are chlorocamphenes (CC). Relative retention time (RRT) data from Smalling and Maruya (2001). na = not available.

Parlar No.	Homolog	Structure	RRT (DB-5)	RRT (DB-XLB)
11	6	5,5,6-exo,8,9,10-hexaCC	0.585	0.599
12	6	5-exo,6-endo,8,9,9,10-hexaCC	0.591	0.603
15	6	5-exo,6-endo,7-anti,8,9,10-hexaCC	0.619	0.636
Hx-Sed*	6	2-exo,3-endo,6-exo,8,9,10	0.621	0.653
21	7	2,2,5,5,9,10,10	0.661	0.675
Hp-Sed*	7	2-endo,3-exo,5-endo,6-exo,8,9,10	0.662	0.669
25	7	5,5,6-exo,8,9,9,10-heptaCC	0.695	0.707
26	8	2-endo,3-exo,5-endo,6-exo,8,8,10,10	0.708	0.687
31	8	5,5,6-exo,8,8,9,9,10-octaCC	0.748	0.738
32	7	2,2,5-endo,6-exo,8,9,10	0.748	0.777
38	8	2,2,5,5,9,9,10,10	0.792	0.801
39	8	2,2,3-exo,5-endo,6-exo,8,9,10	0.813	0.812
40	8	2-endo,3-exo,5-endo,6-exo,8,9,10,10	0.824	0.828
41	8	2-exo,3-endo,5-exo,8,9,9,10,10	0.824	0.820
42a/b	8	2,2,5-endo,6-exo,8,8,9,10 (or 8,9,9,10)	0.832	0.839
44	8	2-exo,5,5,8,9,9,10,10	0.844	0.844
50	9	2-endo,3-exo,5-endo,6-exo,8,8,9,10,10	0.888	0.860
51	8	2,2,5,5,8,9,10,10	0.900	na
56	9	2,2,5-endo,6-exo,8,8,9,10,10	0.945	0.928
58	9	2,2,3-exo,5,5,8,9,10,10	0.959	0.947
59	9	2,2,5-endo,6-exo,8,9,9,10,10	0.968	0.955
62	9	2,2,5,5,8,9,9,10,10	1.000	1.000
63	9	2-exo,3-endo,5-exo,6-exo,8,8,9,10,10	1.016	1.008
69	10	2,2,5,5,6-exo,8,9,9,10,10	1.151	1.127

* Not present in the 22-component TM2 mixture (Ehrenstorfer unpublished data).

for all 22 TM2 components. The retention times for Hx-Sed and Hp-Sed were recorded from a solution provided by Dr. G. Fingerling (Technical University Munich, Germany). For congeners that were split between F1 and F2, the concentration was reported as the sum of the 2 fractions, providing that its presence was confirmed in each fraction by GC-ECNI-MS (see also Quality Assurance/Quality Control).

Total toxaphene concentration (Σ TOX) was estimated by calibrating the GC-ECD with a technical toxaphene product standard (TTX) provided by J. Hoffman (Hercules Inc., Wilmington, Delaware). A known mass of TTX was diluted in CH_2Cl_2 to create a concentrated stock ($\sim 2,800 \mu\text{g ml}^{-1}$). Serial dilutions were then created in hexane at concentrations between 0.28 – $55 \mu\text{g ml}^{-1}$. An average response factor for TTX was computed by summing the areas of all peaks in the unresolved hump of unmodified toxaphene (Fig. 2), and dividing by the known standard mass. The TTX response was then applied to the summed area of peaks eluting within a specified retention time window (25–60 min; Fig. 2). For samples with detectable toxaphene residues in both F1 and F2 extracts using GC-ECNI-MS (see also Quality Assurance/Quality Control), Σ TOX was reported as the sum of both fractions. Areas for peaks corresponding to non-toxaphene compounds eluting within this time

window (e.g., Cl_4 - Cl_7 PCBs in F1 extracts, and organochlorine pesticides such as chlordanes and DDTs in F2 extracts) were subtracted from estimates of Σ TOX. Retention times for PCB and pesticide analytes subtracted in this fashion were recorded from standard reference mixtures SRM2262 and 2261, respectively (National Institute of Standards and Technology, Gaithersburg, Maryland).

QUALITY ASSURANCE/QUALITY CONTROL

Individual congener and total toxaphene concentrations were validated against a comprehensive, performance based set of quality assurance/quality control (QA/QC) criteria. Procedural blanks and toxaphene spiked reference tissue were analyzed in conjunction with the samples. Surrogate compounds (dibromooctafluorobiphenyl or DBOFB and α -HCH) were added to each sample prior to extraction to track target analyte recovery. Initial GC-ECD calibration with the TM2 standard resulted in r^2 values exceeding 0.99 for all 22 components. Continuing instrument calibration was monitored using mid-level TM2 and TTX solutions injected every 10–12 samples; the mean relative percent deviation from the initial calibration response for TM2 analytes was maintained at or below 15% for the duration of the study. Mass cali-

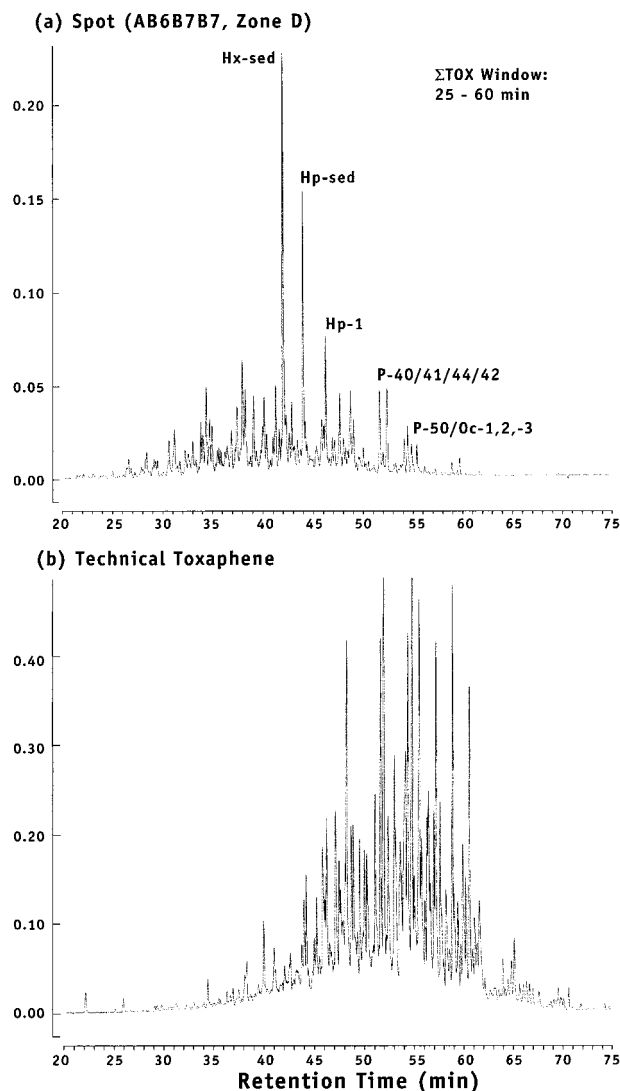


Fig. 2. GC-ECD F2 chromatogram of (a) edible tissue of spot (*Leiostomus xanthurus*) from Dupree Creek (zone D) and (b) unmodified technical toxaphene. Note the predominance of Hx-Sed and Hp-Sed and a shift toward earlier eluting peaks in (a) relative to (b).

bration of the GC-MS system used for analyte confirmation was performed weekly.

Five procedural blanks consisting of ~50 g kiln fired Na_2SO_4 were processed and analyzed using the procedures followed for tissue samples. No target toxaphene congeners were detectable by GC-ECD or GC-ECNI-MS in any blank sample; ECD-based ΣTOX in these blanks ranged between 2.2 to 9.6 ng g^{-1} ($\bar{x} \pm \sigma$: $6.76 \pm 2.89 \text{ ng g}^{-1}$). Since toxaphene levels in most of our samples were in the $\mu\text{g g}^{-1}$ range, ΣTOX were not corrected for blank levels. Mean surrogate recoveries ($\bar{x} \pm \sigma$) for DBOFB and α -HCH were $92 \pm 18\%$ and $79 \pm 19\%$,

respectively. The mean recoveries of TM2 components and TTX spiked into reference seatrout muscle were $107 \pm 11\%$ and 94% , respectively. Twelve of the 22 TM2 components eluted in F2, 5 eluted in F1, and the remaining 5 were split between F1 and F2.

The presence of a toxaphene congener was confirmed using GC-ECNI-MS if the retention time ($\pm 0.1 \text{ s}$) and mass spectrum ($\geq 70\%$) matched that of a target analyte in TM2 (Table 2). Nine individual congeners, Hx-Sed and Hp-Sed, P-26, P-40, P-41, P-42, P-50, P-62, and P-63, were confirmed in the majority of samples (Fig. 3). Since $[\text{M-Cl}]^-$ fragment ions are typically predominant for toxaphene congeners using ECNI-MS (Jansson and Wideqvist 1983), the degree of chlorination for several unidentified toxaphene congeners (e.g., Hx-1, Hp-1, Hp-2, and Hp-3; Oc-1, Oc-2, and Oc-3) was also assigned. Several Cl_7 - Cl_{10} PCB congeners, the majority of which eluted outside the 25–60 min ΣTOX retention time window, were confirmed in the majority of F1 extracts. These PCBs were attributed to Aroclor 1268 contamination originating from a different tidal creek system in the Turtle-Brunswick river estuary, approximately 20 miles away by boat from our study site (Maruya and Lee 1998). Peaks corresponding to PCB congeners in Aroclor 1268 that eluted within the ΣTOX retention time window (e.g., IUPAC numbers 153, 187, 202, and 201) were not included in estimated ΣTOX calculations. Pesticide analytes that were potential interferences in our toxaphene congener specific analyses (e.g., oxychlordane, 4,4'-DDE, 4,4'-DDT) were not detected in our samples using GC-ECNI or electron impact MS.

DATA AND STATISTICAL ANALYSES

All instrument calibration and sample concentration data were compiled and analyzed using Microsoft Excel 97 SR-2 spreadsheet software. Statistical evaluation of the data using analysis of variance (ANOVA) and *t*-tests were performed using the data analysis tool in Excel.

Results and Discussion

TOTAL TOXAPHENE CONCENTRATIONS

Total toxaphene concentrations on a wet weight basis (ΣTOX) ranged from 1.7 ng g^{-1} for blue crab (zone A) to 26,000 ng g^{-1} for mullet (zone B) (Table 3), a range spanning over 4 orders of magnitude. Normalized to extractable lipid content, total toxaphene concentrations ($\Sigma\text{TOX}_{\text{lip}}$) ranged from 0.59 $\mu\text{g g}^{-1}$ for blue crab (zone A) to 270 $\mu\text{g g}^{-1}$ for mullet (zone B); lipid normalization reduced the variability in ΣTOX to a factor of ~500. Using linear regression analysis, ΣTOX and extractable lipid content were positively correlated (ΣTOX

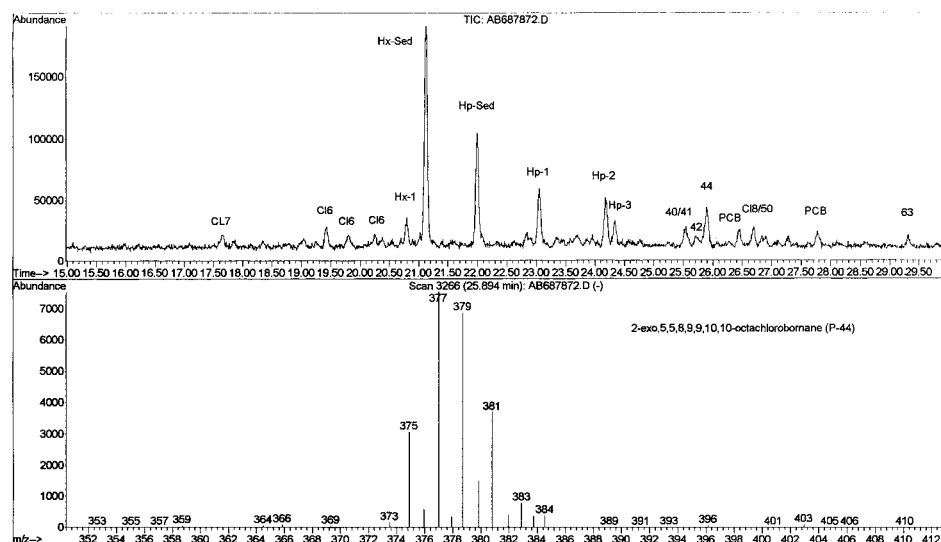


Fig. 3. GC-ECNI-MS (top panel) total ion chromatogram of spot (*Leiostomus xanthurus*) (zone D) and (bottom panel) mass spectral confirmation of Parlar 44, a Cl_8 component of technical toxaphene (CTT). Also prominent are other known CTTs (e.g., P40/41, 50, and 63), breakdown products (Hx-Sed and Hp-Sed), and several unidentified Cl_6 - Cl_8 bornane residues (e.g., Hx-1 and Hp-1).

(ng g^{-1} wet weight) = $593 \times \% \text{ lipid} + 453$; $n = 56$; $r^2 = 0.32$; $p < 0.001$) indicating that lipid content is an important co-varying factor. Because ΣTOX and $\Sigma\text{TOX}_{\text{lip}}$ were also strongly correlated ($\Sigma\text{TOX}_{\text{lip}}$ (in $\mu\text{g g}^{-1}$) = $0.0082 \times \Sigma\text{TOX} + 20.9$; $n = 56$; $r^2 = 0.72$; $p < 0.001$), it can be inferred that

any trends noted for ΣTOX may be generalized to a lipid basis as well.

Species Differences

The 13 samples with the highest ΣTOX were composites of mullet, spot, and croaker (Table 3).

TABLE 3. Ranking of samples according to ΣTOX (ng g^{-1} wet weight) in descending order.

Species	Zone	% Lipid	ΣTOX	Species	Zone	% Lipid	ΣTOX
Mullet	B	11	26,000	seatrout	B	2.8	940
Spot	D	13	23,000	yellowtail	A	4.3	840
Mullet	B	6.3	17,000	spot	A	19	760
Spot	B	15	16,000	red drum	D	0.55	490
Spot	D	9.6	13,000	yellowtail	A	2.1	370
Spot	C	14	12,000	blue crab	D	0.60	270
Croaker	B	14	11,000	yellowtail	A	1.5	230
Croaker	D	12	11,000	shrimp	D	0.69	210
Mullet	B	8.2	8,300	shrimp	C	0.99	150
Spot	D	12	7,600	shrimp	B	1.1	120
Croaker	B	10	6,300	seatrout	A	1.9	110
Spot	C	13	5,700	blue crab	B	0.40	95
Croaker	D	7.2	5,300	shrimp	C	0.93	93
Seatrout	C	6.7	4,400	blue crab	C	0.88	93
Mullet	A	11	4,100	shrimp	D	0.53	91
Seatrout	B	6.9	3,600	flounder	D	0.24	90
Sseatrout	D	2.3	3,300	red drum	A	0.93	79
Spot	A	16	2,400	blue crab	D	0.56	75
Mullet	D	2.1	2,100	shrimp	C	0.72	72
Whiting	D	3.4	2,100	blue crab	C	1.0	66
Mullet	D	2.2	2,000	blue crab	B	0.31	61
Mullet	A	4.2	1,800	black drum	A	1.5	51
Red drum	B	1.7	1,400	blue crab	D	0.43	49
Mullet	D	2.6	1,300	blue crab	A	1.1	44
Spot	A	21	1,300	blue crab	B	0.50	36
Seatrout	A	12	1,300	shrimp	A	1.5	≤ 12
Seatrout	A	6.2	1,300	blue crab	A	1.0	6.3
Mullet	A	3.0	970	blue crab	A	0.29	1.7

TABLE 4. Species differences in Σ TOX were significant using one-way ANOVA.

Groups	Count	Sum	Average	Variance		
Blue crab	11	798	73	5,221		
Mullet	9	63,570	7,063	76,995,600		
Seatrout	7	14,950	2,136	2,592,795		
Shrimp	7	748	107	3,892		
Spot	9	81,760	9,084	56,691,678		
ANOVA						
Source	SS	df	MS	F	p-value	F crit
Between groups	6.15×10^8	4	1.54×10^8	5.38250	0.002	2.61899
Within groups	1.09×10^9	38	28,556,067			
Total	1.7×10^9	42				

Conversely, 8 of the 11 samples with the lowest Σ TOX were blue crab. Roughly half of all samples (27 of 56) had Σ TOX $> 1,000$ ng g⁻¹. Overall species differences in mean Σ TOX were observed using one-way ANOVA (Table 4). Of the 5 species with 7 or more composite samples (blue crab, mullet, seatrout, shrimp, and spot), spot had the highest mean levels of toxaphene (9,100 ng g⁻¹) and blue crab the lowest (72.6 ng g⁻¹). Using 2-tailed *t*-tests assuming unequal variances, significant ($p = 0.05$) differences in mean Σ TOX were noted for blue crab and each of the 3 finfish species (mullet, seatrout, and spot), shrimp and each of the 3 finfish, and seatrout and spot. No differences in mean Σ TOX were noted for blue crab and shrimp, mullet and spot, or mullet and seatrout.

Zone (Sampling Location) Differences

Since toxaphene levels were previously determined to be species dependent, Σ TOX grouped by species were analyzed by one-way ANOVA to assess for differences among the 4 sampling zones. This was possible for 3 species (blue crab, mullet, and spot) since Σ TOX values for a minimum of 3 replicate samples were needed for at least 2 different zones (Table 5). For blue crab, no difference in mean Σ TOX among zones A, B, and D was observed ($p = 0.24$). For mullet, however, the mean Σ TOX for zone B (17,000 ng g⁻¹) was significantly

higher than mean Σ TOX for zones A or D (2,300 and 1,800 ng g⁻¹, respectively). For spot, the mean Σ TOX for zone D was higher than for zone A (15,000 versus 1,500 ng g⁻¹). For a given species, Σ TOX for zones B (Terry Creek at bridge) and D (Dupree Creek) were, in general, elevated relative to zone A (Terry Creek at Lanier Basin).

An insufficient number of samples precluded statistical comparisons for all species in zone C, and for blue crab and shrimp (all zones). The mean Σ TOX for zone C spot (8,900 $\pm$ 3,150 ng g⁻¹, $n = 2$) was however, intermediate between mean levels for zones D and A. For blue crab, mean Σ TOX decreased from zone D (130 $\pm$ 99 ng g⁻¹) to zone A (17 $\pm$ 19 ng g⁻¹) with intermediate values for zones B (64 $\pm$ 24 ng g⁻¹) and C (80 $\pm$ 14 ng g⁻¹). Similarly, mean Σ TOX decreased for shrimp from zone D (150 $\pm$ 60 ng g⁻¹) through zones B and C (120 ng g⁻¹, $n = 1$; 105 $\pm$ 33 ng g⁻¹) to zone A (≤ 12 ng g⁻¹, $n = 1$). Clearly, biota from zones D (i.e., Dupree Creek), B, and C (lower Terry Creek near the Torres Causeway Bridge) contained higher toxaphene levels than those collected from zone A (i.e., the Lanier Basin area). This decreasing trend in Σ TOX for all species strongly suggests a spatial toxaphene contamination gradient away from the Hercules, Inc. discharge canal in Dupree Creek (Fig. 1).

TABLE 5. *t*-test comparisons indicate significant differences in Σ TOX among sampling zones for mullet and spot.

	Mullet			Spot	
	Zone D	Zone B	Zone A	Zone D	Zone A
Mean	1,800	17,100	2,290	14,533	1,487
Variance	190,000	78,330,000	2,629,300	61,053,333	698,533
Observations	3	3	3	3	3
Hypothesis	0		0	0	
df	2		2	2	
<i>t</i> stat	-2.9906		-2.8509	2.8756	
P($T \leq t$) 1-tail	0.05		0.05	0.05	
<i>t</i> crit 1-tail	2.9200		2.9200	2.9200	
P($T \leq t$) 2-tail	0.0960		0.1042	0.1026	
<i>t</i> crit 2-tail	4.3027		4.3027	4.3027	

TABLE 6. Individual congener concentrations ($\bar{x} + \sigma$ in units of ng g^{-1}) for zone D finfish and shellfish. Value in parentheses are 1s.

	Mullet (n = 3)	Spot (n = 3)	Seatrout (n = 1)	Blue crab (n = 3)	Shrimp (n = 2)
Hx-Sed	122 (16.7)	1,420 (833)	331	4.12 (0.48)	10.2 (3.15)
Hp-Sed	300 (64.9)	1,130 (735)	502	2.52 (1.28)	7.42 (2.14)
P-26/Hp-1	61.6 (26.0)	549 (226)	129	7.54 (6.30)	6.86 (2.84)
P-40/41	16.0 (3.91)	136 (59.5)	40.2	0.78 (0.43)	0.91 (0.28)
P-42	16.3 (8.96)	94.4 (36.1)	26.3	≤ 1.29	0.50 (0.22)
P-44	27.8 (25.2)	355 (147)	100	1.50 (1.00)	≤ 2.58
P-50/Oc-1	27.7 (2.42)	178 (72.9)	46.2	2.41 (1.17)	4.21 (1.75)
P-62	11.4 (2.24)	51.3 (26.9)	17.8	0.86 (0.04)	1.00 (0.39)
ΣTOX	1,800 (360)	15,000 (6,400)	3,300	130 (98)	150 (60)

TOXAPHENE CONGENER CONCENTRATIONS AND PROFILES

The concentrations of 23 toxaphene congeners (or congener pairs) on a wet weight basis ranged from nondetectable ($< 0.25 \text{ ng g}^{-1}$) to a maximum of $3,450 \text{ ng g}^{-1}$ for Hp-Sed (mullet, zone B). Of the 9 congeners confirmed by GC-ECNI-MS, Hx-Sed and Hp-Sed were, in general, the most abundant followed by (in order of decreasing abundance) P-26/Hp-1, 44, 50/Oc-1, 40/41, 42, 62, and 63. In zone D samples for example, mean concentrations ranged from 0.78 ng g^{-1} for P-40/41 in blue crab to $1,420 \text{ ng g}^{-1}$ for Hx-Sed in spot (Table 6). One exception was noted for blue crab, where P-26/Hp-1 was more abundant than either Hx-Sed or Hp-Sed.

The distribution of confirmed toxaphene congeners was, in general, similar for all samples (Fig. 4). In previous studies at this site, Hx-Sed and Hp-Sed were also the most abundant toxaphene congeners in sediment and biota (Vetter and Maruya 2000). Because these congeners are only minor components of unmodified technical toxaphene, they are widely believed to be dead-end metabolites of reductive dechlorination in contaminated,

anoxic sediments (Stern et al. 1996). Their predominance has also been observed in native rainbow trout from toxaphene-treated lakes (Miskimmin et al. 1995). Several Cl_8 congeners (e.g., P-40, P-41, P-42, and P-44) were also abundant in most of our samples at levels as high as 733 ng g^{-1} (P-44 in mullet, zone B). Relatively few Cl_9 homologs were detected; the 2 congeners that were prominent (P-62 and P-63) were present at lower levels (maximum of 175 ng g^{-1} in mullet, AB68863). No Cl_{10} compounds were detected.

Several unidentified Cl_5 - Cl_9 toxaphene residue compounds were also detected in many of these samples (Figs. 2 and 3). Pentachlorobornanes and hexachlorobornanes have been previously identified in Terry and Dupree Creeks sediments (Maruya et al. 2000) and biota (Maruya unpublished data) as have several heptachlorobornanes, including Hp-1 (Vetter and Maruya 2000). A series of Cl_8 compounds eluting after P-44, designated Oc-1, Oc-2, and Oc-3, were also prominent in the majority of our samples. Although their structures remain unknown, they likely occur in technical toxaphene in relatively high abundances. A comparison of individual congener concentrations and ΣTOX estimates indicate that, in many cases, only a small fraction ($< 35\%$) of the toxaphene congeners in our samples have been positively identified (Table 6).

Species and Zone Effects

A difference in congener distribution was noted between fish and crustacean species (Fig. 4). Specifically, shrimp and blue crab had relatively less Hp-Sed and more P-26/Hp-1 and P-50/Oc-1 than fish. Congener profiles for shrimp and blue crab were similar, suggesting a similar exposure regime and/or elimination/metabolic capability. These crustacean species are highly mobile and feed on planktonic organisms as well as detritus. Profile differences among the various species of fish appeared to be limited to Hx-Sed and Hp-Sed (Fig. 4).

Little is known concerning the metabolism of

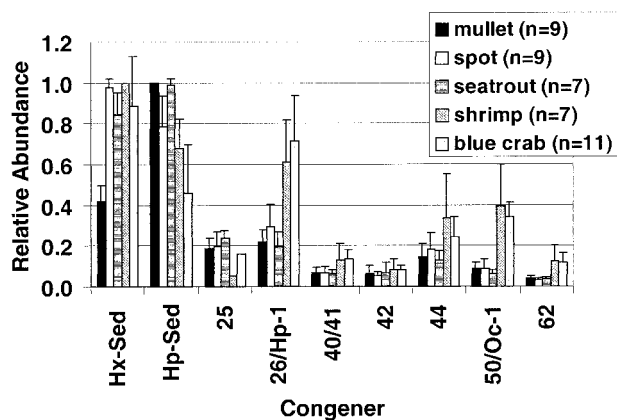


Fig. 4. Mean congener distributions illustrate differences in exposure regime and/or biotransformation capability among finfish and shellfish species. Error bars represent $+1\sigma$.

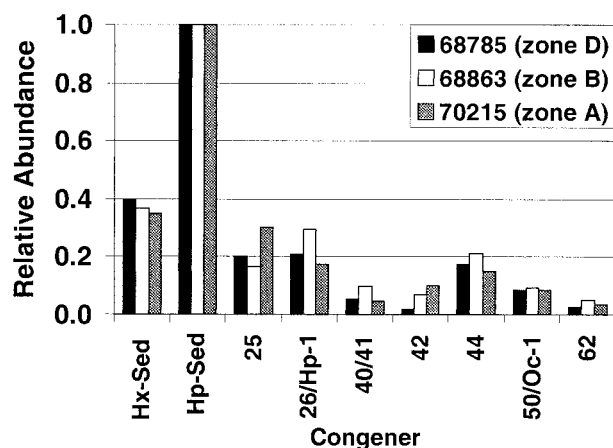


Fig. 5. The similarity of congener distributions in mullet from different sampling locations suggest a uniform exposure regime and/or a high, local mobility for this species.

toxaphene in finfish and shellfish, with the available data suggesting highly variable biotransformation capabilities among species (Saleh 1991). Recent studies indicate that the elimination of Hx-Sed and Hp-Sed by fish is relatively rapid with respect to P-26 and P-50, two congeners considered to be among the most recalcitrant (Fisk et al. 1998; Smalling et al. 2000). The enantioselective nature of the elimination process strongly suggests that these fish actively metabolize Hx-Sed and Hp-Sed (Smalling et al. 2000). If one assumes that all species in this study similarly eliminate toxaphene residues and that Hx-Sed is eliminated faster than Hp-Sed (Smalling et al. 2000), the predominance of Hx-Sed in spot, croaker, and the 2 crustacean species may indicate uninterrupted exposure to sources of toxaphene contamination. This may be due to exposure to sources enriched with Hx-Sed, e.g., tidal creek water at the site (Maruya unpublished data). The predominance of Hp-Sed in species such as mullet and seatrout suggests intermittent exposure and/or exposure to Hp-Sed enriched media. Mullet are known to ingest sediment in search of food, they in turn are preyed upon by seatrout.

Spatial differences in toxaphene profiles by species were not as obvious. Congener distributions for mullet (Fig. 5) and seatrout (not shown) from zones A, B, and D were similar, suggesting that exposure regimes were uniform throughout the study area and/or that fish moved freely among study zones. Location-specific profiles for shrimp (Fig. 6) and blue crab (not shown) were more variable, with congener complexity decreasing from zone D (Dupree Creek) to zone A (Terry Creek at Lanier Basin). Because of their lower hydrophobicity, Hx-Sed and, to a lesser extent, Hp-Sed are

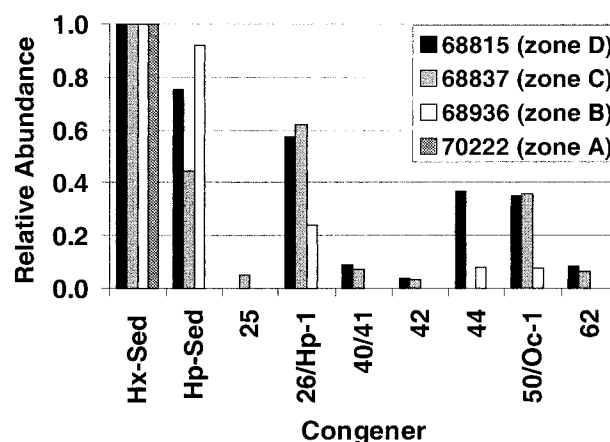


Fig. 6. The difference in congener distributions for shrimp from different sampling locations suggests a spatially varying contaminant exposure regime for this species. The predominance of Hx-Sed may indicate preferential uptake of toxaphene residues via the water column.

more efficiently transported from the zones of heaviest toxaphene contamination (i.e., zones B–D). It follows that exposure via water absorption may be more important for the crustacean species. Caution should be exercised when interpreting blue crab and shrimp profiles as the concentrations of individual congeners in most of these samples were relatively low.

COMPARISONS WITH OTHER STUDIES

Total and individual toxaphene congener concentrations in our samples were compared with levels for various aquatic species from other regions compiled by de Geus et al. (1999) and Saleh (1991). Σ TOX in our samples were as high as maximum levels reported for contaminated fish eating birds, and 20 times higher than those reported for other marine fish species. Only blubber of white beaked dolphins from Newfoundland had higher toxaphene levels (38–46 $\mu\text{g g}^{-1}$). Σ TOX for bottom dwelling, forage species in our study were four fold higher than Lake Superior lake trout and more than 30 times higher than rainbow smelt from Lakes Michigan and Ontario. Average toxaphene in carp, a bottom dwelling fish, from the Great Lakes was reported to be 3.3 $\mu\text{g g}^{-1}$, roughly a third of the mean Σ TOX for spot from Dupree Creek (zone D, 9.1 $\mu\text{g g}^{-1}$). Toxaphene levels for spotted seatrout in our study were comparable to those for Great Lakes salmonids. Very little toxaphene data is available for invertebrates; concentrations reported previously were typically low (< 0.002 $\mu\text{g g}^{-1}$).

Maximum concentrations for Hx-Sed and Hp-Sed (up to 3.4 $\mu\text{g g}^{-1}$ wet weight) in our samples were two or more orders of magnitude higher than

previously reported for any toxaphene congener. The majority of congener specific data available are for P-26 and P-50, as they comprise a large percentage of the total toxaphene detected in biota from intensely studied regions, e.g., the north Atlantic and Arctic Oceans. Miskimmin et al. (1995) reported maximum total toxaphene levels of $\sim 0.1 \mu\text{g g}^{-1}$ in Chatwin and Peanut Lake rainbow trout where Hx-Sed and Hp-Sed were also predominant, but did not give estimates of individual congener concentrations.

Toxaphene concentrations and distributions similar to those reported herein were noted for other species collected independently during 1997–1998 from Terry and Dupree Creeks, including grass shrimp (*Palaemonetes* sp.) and mummichogs (*Fundulus* sp.) (Vetter and Maruya 2000). In contrast, ΣTOX in this study were, in general, two to three orders of magnitude higher than in similar species caught in other areas of the Turtle-Brunswick River estuary and St. Simons Sound (Maruya and Lee 1998). Estimated toxaphene concentrations based on packed column GC-ECD in shrimp (*Penaeus* sp.) tails, anchovy (*Anchoa* sp.), and star drum (*Stellifer* sp.) collected at our site in the early 1970s ranged from 0.6–2.5, 8.6–20, and 1.1–3.2 $\mu\text{g g}^{-1}$ wet weight, respectively (Reimhold and Durant 1974).

IMPLICATIONS FOR ECOLOGICAL AND HUMAN HEALTH EFFECTS

ΣTOX in our samples ranged from < 0.05 to $\sim 26 \mu\text{g g}^{-1}$, estimates that are in many cases considerably higher than originally reported using analytical methods based on U.S. Environmental Protection Agency (1996) Method 8081 (Georgia Department of Natural Resources 1997) and specific criteria defining the presence of toxaphene (Carlin et al. 2000). This discrepancy is not surprising as toxaphene was detected using the criteria of Carlin et al. (2000) if a minimum of four GC peaks in the back half of the ECD chromatogram of unmodified technical toxaphene (e.g., TTX; Fig. 2) were determined to be present in a sample. These peaks were quantified as toxaphene if and only if their relative proportions in the sample matched that of TTX. The demarcation of the back half of the ECD chromatogram is the highest responding peak in technical toxaphene, seen in Fig. 2 as eluting between 55 and 60 min. It is clear that selective weathering and/or biotransformation of technical toxaphene in reducing sedimentary environments results in a highly modified, left-shifted residue pattern in sediment (Williams and Bidleman 1978; Stern et al. 1996) and in local food web organisms including fish (Miskimmin et al. 1995; Maruya et al. 2000). It is evident from the similar, left-shifted

patterns in our composite samples (Fig. 2) that the criteria defined by Carlin et al. (2000) were not optimized for detecting residues resulting from technical toxaphene (weathered toxaphene).

Another important difference was the pre-separation of nonpolar compounds, including PCBs, from toxaphene residues using solid-liquid chromatography in our study. Even though we did not achieve complete separation using Florisil, the bulk of the total PCB mass in our samples eluted in F1 whereas toxaphene residues eluted primarily in F2. This was confirmed using GC-ECNI-MS. Neither pre-separation of PCBs nor GC-MS confirmation of toxaphene was performed using the 8081-based method (Carlin et al. 2000). Fortuitously, the PCB congeners prominent in our samples are late eluting $\text{Cl}_7\text{-Cl}_{10}$ homologs, whose origins have been previously linked to Aroclor 1268 (Kannan et al. 1997; Maruya and Lee 1998). The potential for PCB interference using our ΣTOX method (25–60 min window) is lessened whereas the potential for interference by these same PCBs in selecting and identifying back half toxaphene components using the criteria of Carlin et al. (2000) is increased. Improved cleanup (e.g., Krock et al. 1997) and additional confirmatory techniques are needed to better characterize weathered toxaphene residues. Our work represents the first application of chromatographic cleanup and GC-MS in characterizing the complex weathered toxaphene profile observed in these samples.

Both species- and location-specific trends in toxaphene levels were also observed. Bottom feeding finfish such as spot, croaker, and mullet exhibited the highest levels, whereas the lowest concentrations were consistently found in the two shellfish species (blue crab and shrimp). Larger, predatory finfish such as seatrout and red drum had intermediate contamination levels ranging from < 0.1 to $\sim 5 \mu\text{g g}^{-1}$. Spatial differences in contamination were also apparent with tissue levels elevated at sites closest to the original source of toxaphene contamination. Subtle differences in congener distribution among species may be the result of varying exposure regimes (e.g., uptake mechanism, time spent in the contaminated creek habitats) and/or intrinsic elimination/metabolic capabilities.

Science-based regulatory action levels and risk information in general for weathered toxaphene are extremely limited (Agency for Toxic Substances and Disease Registry 1998). Translation of our results into ecological and human health risks is typically based on extrapolations to unmodified technical toxaphene. Recent investigations indicate that weathered toxaphene residues at sub ppm ($= \mu\text{g g}^{-1}$) tissue levels may affect reproduc-

tion and possibly induce DNA damage in grass shrimp (*Palaemonetes* sp.), an important species in salt marsh ecosystems (Lee and Maruya 1999). Other experiments suggest that *Fundulus* are less susceptible to toxaphene contamination based on a battery of both acute and chronic toxicological endpoints (Maruya and Lee unpublished data).

It was recently documented that *Fundulus* can rapidly eliminate some residues of toxaphene, including Hx-Sed and Hp-Sed, the two predominant congeners found in the Terry-Dupree Creeks ecosystem (Smalling et al. 2000; Vetter and Maruya 2000). Other congeners such as P-26, P-50, and B7-1000, a recently identified Cl₇ bornane isolated from Dupree Creek sediments (Vetter et al. 2001), were much more recalcitrant. These congeners appear to be resistant to metabolism in other species (e.g., marine and terrestrial mammals; Boon et al. 1998) as well as in fish (Delorme et al. 1988; Fisk et al. 1998). It follows that the more labile toxaphene residues in biota from our study area will be removed via elimination/metabolic processes (natural attenuation) if contamination sources are reduced and/or eliminated. The long-term ecological and human health risks from toxaphene in this system can thus be expected to shift to recalcitrant congeners.

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LITERATURE CITED

- AGENCY FOR TOXIC SUBSTANCES AND DISEASE REGISTRY. 1998. Draft public health assessment for Terry Creek dredge spoil area, Brunswick, Glynn County, Georgia. CERCLIS no. GAD982112658. Agency for Toxic Substances and Disease Registry, Atlanta, Georgia.
- ALDER, L. AND B. VIETH. 1996. A congener specific method for the quantification of camphechlor (toxaphene) residues in fish and other foodstuffs. *Fresenius Journal of Analytical Chemistry* 354:81-92.
- BIDLEMAN, T. F., M. D. WALLA, D. C. G. MUIR, AND G. A. STERN. 1993. Selective accumulation of polychlorocamphenes in aquatic biota from the Canadian Arctic. *Environmental Toxicology and Chemistry* 12:701-709.
- BOON, J. P., H. M. SLEIDERINK, M. S. HELLE, M. DEKKER, A. VAN SCHANKE, E. ROEX, M. T. J. HILLEBRAND, H. J. C. KLAMER, B. GOVERS, D. PASTOR, D. MORSE, P. G. WESTER, AND J. DE BOER. 1998. The use of microsomal in vitro assay to study phase I biotransformation of chlorobornanes (toxaphene) in marine mammals and birds. Possible consequences of biotransformation for bioaccumulation and genotoxicity. *Comparative Biochemistry and Physiology Part C* 121:385-403.
- CALERO, S., I. FOMSGAARD, M. L. LACAYO, V. MARTINEZ, AND R. RUGAMA. 1993. Toxaphene and other organochlorine pesticides in fish and sediment from Lake Xolotlan, Nicaragua. *International Journal of Environmental Analytical Chemistry* 53: 297-305.
- CARLIN, F. J., H. L. REVELLS, AND D. L. REED. 2000. The application of standard methods for the determination of toxaphene in environmental media. *Chemosphere* 41:481-486.
- DE GEUS, H. J., H. BESSELINK, A. BROUWER, J. KLUNGSOYR, B. MCHUGH, E. NIXON, G. G. RIMKUS, P. G. WESTER, AND J. DE BOER. 1999. Environmental occurrence, analysis and toxicology of toxaphene compounds. *Environmental Health Perspectives* 107:115-144.
- DELORME, P. D., D. C. G. MUIR, W. L. LOCKHART, K. H. MILLS, AND F. J. WARD. 1988. Depuration of toxaphene in lake trout and white suckers in a natural ecosystem following a single I. P. dose. *Chemosphere* 10:1965-1973.
- FINGERLING, G. M., N. HERTKORN, AND H. PARLAR. 1996. Formation and spectroscopic investigation of two hexachlorobornanes from six environmentally relevant toxaphene components under anaerobic conditions. *Environmental Science and Technology* 30:2984-2992.
- FISK, A. T., R. J. NORSTROM, C. D. CYMBALISTY, AND D. C. G. MUIR. 1998. Dietary accumulation and depuration of hydrophobic organochlorines: Bioaccumulation parameters and their relationship with the octanol/water partition coefficient. *Environmental Toxicology and Chemistry* 17:951-961.
- GEORGIA DEPARTMENT OF NATURAL RESOURCES. 1997. Study plan for the analyses of seafood from Terry and Dupree Creeks, Brunswick, Georgia. Georgia Department of Natural Resources, Environmental Protection Division, Atlanta, Georgia.
- JANSSON, B. AND U. WIDEQVIST. 1983. Analysis of toxaphene (PCC) and chlordane in biological samples by NCI mass spectrometry. *International Journal of Environmental Analytical Chemistry* 13:309-321.
- KANNAN, K., K. A. MARUYA, AND S. TANABE. 1997. Distribution and characterization of polychlorinated biphenyl congeners in soils and sediments from a Superfund site contaminated with Aroclor 1268. *Environmental Science and Technology* 31: 1483-1488.
- KROCK, B., W. VETTER, AND B. LUCKAS. 1997. PCB/toxaphene group separation on silica prior to congener specific determination of toxaphene in fish and other samples by GC/ECD. *Chemosphere* 35:1519-1530.
- LEE, R. F. AND K. A. MARUYA. 1999. Embryo development effects and DNA damage in grass shrimp (*Palaemonetes pugio*) from Terry/Dupree Creek. U.S. Environmental Protection Agency Region 4, Atlanta, Georgia.
- MARUYA, K. A. AND R. F. LEE. 1998. Aroclor 1268 and toxaphene in fish from a southeastern U.S. estuary. *Environmental Science and Technology* 32:1069-1075.
- MARUYA, K. A., S. G. WAKEHAM, W. VETTER, AND L. FRANCENDESE. 2000. Prominent chlorobornane residues in estuarine sediments contaminated with toxaphene. *Environmental Toxicology and Chemistry* 19:2198-2203.
- MISKIMMIN, B. M., D. C. G. MUIR, D. W. SCHINDLER, G. A. STERN, AND N. P. GRIFT. 1995. Chlorobornanes in sediments and fish 30 years after toxaphene treatment of lakes. *Environmental Science and Technology* 29:2490-2495.
- MUIR, D. C. G. AND J. DE BOER. 1995. Recent developments in the analysis and environmental chemistry of toxaphene with an emphasis on the marine environment. *Trends in Analytical Chemistry* 14:56-66.
- POLLOCK, G. A. AND W. W. KILGORE. 1978. Toxaphene. *Residue Reviews* 50:87-140.
- REIMHOLD, R. J. AND C. J. DURANT. 1974. Toxaphene content of estuarine fauna and flora before, during, and after dredging toxaphene-contaminated sediments. *Pesticides Monitoring Journal* 8:44-49.
- SALEH, M. A. 1991. Toxaphene: Chemistry, biochemistry, toxicity and environmental fate. *Reviews of Environmental Contamination and Toxicology* 118:1-85.
- SMALLING, K. L. AND K. A. MARUYA. 2001. Gas chromatographic separation of toxaphene residues by DB-XLB. *Journal of Separation Science* 24:104-108.

- SMALLING, K. L., K. A. MARUYA, AND W. VETTER. 2000. Elimination of toxaphene residues by the mummichog (*Fundulus* sp.). Abstracts of the 219th American Chemical Society National Meeting, San Francisco, California, March 26–30, 2000, American Chemical Society, Washington, D.C.
- STANLEY, C. W., J. E. BARNEY, M. R. HELTON, AND A. R. YOHS. 1971. Measurement of atmospheric levels of pesticides. *Environmental Science and Technology* 5:430–435.
- STERN, G. A., M. D. LOEWEN, B. M. MISKIMMIN, D. C. G. MUIR, AND J. B. WESTMORE. 1996. Characterization of two major toxaphene components in treated lake sediment. *Environmental Science and Technology* 30:2251–2258.
- U.S. ENVIRONMENTAL PROTECTION AGENCY. 1980. Ambient water quality criteria for toxaphene. U.S. Environmental Protection Agency Report no. 440/5–80–076, Washington, D.C.
- U.S. ENVIRONMENTAL PROTECTION AGENCY. 1996. Organochlorine pesticides by gas chromatography, Method 8081A, Rev. 1, p. 8081–1–75. In *Test Methods for Evaluating Solid Waste*, SW-846, U.S. Environmental Protection Agency, Office of Solid Waste and Emergency Response, Washington, D.C.
- U.S. ENVIRONMENTAL PROTECTION AGENCY. 1998. A multimedia strategy for priority persistent, bioaccumulative, and toxic (PBT) pollutants. U.S. Environmental Protection Agency Persistent, Bioaccumulative and Toxic Pollutants Plenary Group, Washington, D.C.
- VEITH, G. D. AND G. F. LEE. 1971. Water chemistry of toxaphene: Role of lake sediments. *Environmental Science and Technology* 5: 230–234.
- VETTER, W. AND K. A. MARUYA. 2000. Congener and enantioselective analysis of toxaphene residues in sediment and biota from a contaminated estuarine wetland. *Environmental Science and Technology* 34:1627–1635.
- VETTER, W., E. SCHOLZ, B. LUCKAS, AND K. A. MARUYA. 2001. Structure of a persistent heptachlorobornane in toxaphene agrees with molecular model predictions. *Journal of Agricultural and Food Chemistry* 49:759–765.
- WILLIAMS, R. R. AND T. F. BIDDLEMAN. 1978. Toxaphene degradation in estuarine sediments. *Journal of Agricultural and Food Chemistry* 26:280–282.

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