

FINAL REPORT

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PROJECT ACTIVITIES

Escambia and Santa Rosa Counties in Northwest Florida have experienced continuing deterioration of environmental health due to pollution from point sources (industrial, military, and Superfund sites) and non-point sources (stormwater runoff, septic tanks, lead-contaminated homes, contaminated aquifers, and other diffused sources of pollution). In response to the emerging evidence of groundwater contamination from area Superfund sites and the community's general concerns about regional environmental pollution and potential impacts on human health, the University of West Florida (UWF) initiated environmental health studies in Escambia and Santa Rosa counties in 2002. The objectives of these studies are to identify potential pollutant exposure risks and assist the Northwest Florida community in its efforts to set priorities for improving the health status and quality of life of area residents.

The current project explored environmental health concerns that have not been previously addressed in the two counties. Specifically, this project investigated whether game fish caught in local ponds and lakes serve as an additional source of contaminants to human consumers. In this task, we collected largemouth bass and analyzed tissue samples for dioxins, PCBs, and mercury levels, and compared the data to U.S. EPA screening values. The second task of this study investigated body burdens of mercury in women of child-bearing age to assess whether the high rates of atmospheric mercury deposition in Northwest Florida are reflected in the local population. Hair samples were collected from area residents and data are compared to results from previous national studies, such as NHANES. The third task monitored the health and pollutant levels in a previously untested residential community near the Escambia Wood Treating Company (ETC) Superfund site. Current residents of the Clarinda Triangle area were offered a complete health screening, and biomonitoring for serum dioxins and dioxin-like PCBs and urine arsenic levels to assess their exposure to the ETC contaminants. This research program is pertinent to the "Healthy People 2010" focus area of Environmental Health, and is in alignment with the NCEH's performance Goal 1: Determine human health effects associated with environmental exposures. This study enhanced the overall assessment of environmental impacts on public health in Northwest Florida. The information gathered will assist local residents in understanding their potential exposure to persistent contaminants and provide information on whether dietary or behavioral modifications may be necessary to reduce body burdens.

Task 1: Survey of contamination in freshwater fish from lakes and ponds in Escambia and Santa Rosa Counties

Introduction

Aquatic organisms bioaccumulate and bioconcentrate some environmental pollutants, which over time may be concentrated to levels that cause physiological impairment in humans. Segments of the human population with increased toxic exposure risk include consumers of commercially harvested seafood, recreational fishers, and segments of society that may rely on harvestable species for subsistence. Monitoring of fish and shellfish tissues is common throughout the country as an indicator of potential human exposure to contaminants in sediments and water. These analyses are used for the establishment of consumption advisories for specific species and size classes in specific regions or water bodies.

The State Florida currently has consumption advisories for largemouth bass in most freshwaters of the State, with some areas having high enough values to issue “no consumption” advisories for pregnant or nursing women. Deposition of mercury from combustion sources in rainfall is considered to be the major pathway of environmental contamination (U.S. EPA 1997). Although local variation in such loading is suspected (U.S. EPA 1997), the resolution of national scale data assumes diffuse loading from known point sources, such as coal fired power plants. In addition to defining regional contamination of mercury, PCBs, and Dioxins in largemouth bass as an evaluation of human health risk, this investigation also documented small-scale variation in mercury contamination of impounded surface waters as tissue concentrations in largemouth bass.

Methods

The US EPA’s Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories, Volume 1: Fish Sampling and Analysis, Third Edition (U.S. EPA 2000) was followed in the collection and processing of fish specimens. As soon as possible following collection, specimens were wrapped in aluminum foil, labeled, and stored below 4 °C until tissue processing. Each sample was assigned and tracked by a unique number on all documentation and records. Fish were measured, weighed, filleted, and muscle tissue homogenized prior to analysis of pollutants. Total Mercury was determined by the State of Florida. PCBs and Dioxins were determined by Pace Laboratories, Inc. Toxic Equivalency factors (TEQ) were used from Van den Berg et al. (1998) for mammalian toxicity. Data were reduced and analyzed in Microsoft Excel spreadsheets. Graphic presentation of the data was accomplished with Kaleidagraph software.

Results

Twenty-two locations were sampled for largemouth bass (Table 1.). Of those, eleven yielded enough fish of appropriate sizes to be processed completely for this study, with a total count of 56 specimens. Characteristics of these samples are listed in Table 2. Contaminant concentrations for these samples are listed in Table 3, and averages for each site are shown in Figure 1. Seven out of Eleven locations had samples exceeding the USPA screening value of 0.40 mg/Kg Hg, and five out of eleven sample locations had samples exceeding the higher State of Florida screening value of 0.50 mg/Kg Hg.

Mercury (Hg) concentrations in fish tissues were lowest in the southernmost samples: Bayou Chico and the Tiger Point golf course (Tiger Point GC). Interestingly, a sample from the Tiger Point sewage treatment plant effluent holding pond (Tiger Point WWT) was also among the lowest for Hg, despite this pond being essentially a flow through system where concentration of contaminants from the constant flow of wastewater might be expected. A larger specimen recovered from this pond did rank higher in the list, as might be expected from bioconcentration with age. Highest concentrations recovered were in samples from Woodbine Springs, and samples from western Escambia County. Samples from Woodbine Springs in this study were consistent with the trend of age related Hg contamination found by the Florida Fish and Wildlife Commission (FWC; Figure 2). This graph also indicates lower contamination for larger fish elsewhere in the region than for those found in Woodbine Springs, suggesting that the findings for this pond are not indicative of widespread contamination, but represent a localized source of either enhanced atmospheric deposition or historical land use.

For combined PCB, Dioxin and Furan toxicity equivalency calculations, only two of the eleven exceeded the US EPA screening value of 0.26 ng/Kg: Bayou Chico and the Tiger Point WWT. Both of these samples contained elevated PCB concentrations, and the latter was influenced by the single larger specimen that would have accumulated more of these materials with age. Despite similar PCB tissue concentrations between the single large specimen from the Tiger Point WWT sample and the three fish composite from Bayou Chico (Table 2), the TEQ value for Bayou Chico was nearly 4 times the TEQ for the WWT sample, indicating a prevalence and bioavailability of more highly chlorinated and more toxic congeners in the Bayou Chico site. All other samples had relatively low levels of organic contaminants.

Mercury, Dioxin-Furans, and PCBs should bioaccumulate with both age and trophic status in these fishes, and patterns of increasing body burdens with size or age should be apparent if sources of the contaminants are uniform across all sites. The data from Woodbine Springs (Figure 2) suggest this uniformity does not exist for Mercury, a compound for which the largest potential source is atmospheric deposition. Analysis of all the body burden data from the present study as a function of fish size for mercury, Dioxin-Furans, and PCBs also does not show clear patterns of increasing tissue burdens with size, suggesting that for all these compounds, spatially explicit contamination is affecting bioaccumulation rates. Examination of the data spatially over the three-county geography does show some patterns in the distribution of contaminant accumulation (Figures 4-6). For mercury, the highest concentrations were recovered just inland from the coast. Low concentrations were recovered at the stations closest to the coast at Tiger Point Golf Course and the adjacent Tiger Point sewage treatment plant, and the headwaters of Bayou Chico. It is instructive to note that the flow through of sewage effluent at the Tiger Point waste water plant did not result in elevated mercury in bass sampled there. Further inland, samples had elevated mercury, but the concentrations were not as high as those recovered further south.

Organic contaminants also displayed geographic specificity, but in patterns different than mercury. Dioxin-Furan concentrations were all relatively low (EPA Screening TEQ at 0.26 ng/Kg), with a high concentration found at an RV park pond (Cedar Lakes). PCB concentrations were higher for bass in Bayou Chico headwaters, a known industrial area, and at the Tiger Point treatment plant pond, where constant flow would increase exposure over time. None of these samples, however, exceeded EPA screening values for these contaminants.

Table 1. Sampling locations attempted in this study

Site	County	Pond/Lake
E-1	Escambia	Lake Stone
E-2	Escambia	Wiggins Lake
E-3	Escambia	Lakeside at Barth
E-4	Escambia	Lake Frederic
E-5	Escambia	Bronson Field
E-6	Escambia	NAS Golf Course Pond
E-7	Escambia	Bayou Chico Headwater Ponds
E-8	Escambia	Cantonment Indus. Pk. Pond
E-9	Escambia	Langley-Bell 4-H
E-10	Escambia	Crescent Lake
E-11	Escambia	Hwy98/Fairfield Sand Pit
SR-1	Santa Rosa	Bear Lake
SR-2	Santa Rosa	Woodbine Springs Lake
SR-4	Santa Rosa	Lake Kristina Campground
SR-5	Santa Rosa	Blackjack Creek Campground
SR-6	Santa Rosa	Cedar Lakes RV Park
SR-7	Santa Rosa	Tiger Point WWTP
SR-8	Santa Rosa	Tiger Point Golf Course
SR-9	Santa Rosa	Holley-Navarre Golf Course
OK-1	Okaloosa	Hurricane Lake
SR-A1	Santa Rosa	McMillion Family Pond-E. Milton
E-A1	Escambia	Bluff Springs Campground

Table 2. Sample characteristics for largemouth bass sampling in lakes and ponds in Northwest Florida.

Santa Rosa County

Site	Composite	n	Mean Length	Mean Weight	Mean Age	Length Range	Length SD	Length SE	Weight Range	Weight SD	Weight SE	% Lipids
Bear Lake 1	051017A	3	361.67	703.06	3.67	327-412	44.61	25.76	470-1080	329.48	190.22	0.4
Bear Lake 2	051017B	3	330.00	481.73	2.00	307-358	25.87	14.93	370-630	133.98	77.35	0.5
Bear Lake 3	051017C	3	337.67	555.08	2.67	317-371	29.14	16.83	456-740	160.27	92.53	0.6
Lake Kristina	060322A	3	307.00	356.71	2.33	283-322	21.00	12.12	250-470	110.08	63.56	0.2
Cedar Lakes	060323A	3	348.33	553.33	3.67	297-403	53.08	30.64	340-860	272.27	157.20	0.2
Woodbine Springs 1	060511A	4	301.25	323.91	1.00	297-310	5.97	2.98	310-340	12.34	6.17	0.2
Woodbine Springs 2	060511B	3	321.00	435.73	2.00	306-331	13.23	7.64	377-480	52.86	30.52	0.2
Woodbine Springs 3	060511C	3	345.33	463.33	2.33	340-350	5.03	2.91	420-530	58.59	33.83	0.3
Tiger Pt GC	060603A	3	332.33	408.20	3.00	309-358	24.58	14.19	330-520	99.28	57.32	0.2
Tiger Pt WWT	060405A	3	305.00	361.26	1.00	275-330	27.84	16.07	268-454	93.38	53.91	0.2
Tiger Pt WWT	060612A	1	398.00	750.00	3.00	1398			750			0.2
Tiger Pt WWT	070315A	1	510	1068								.2

Escambia County

Site	Composite	n	Mean Length	Mean Weight	Mean Age	Length Range	Length SD	Length SE	Weight Range	Weight SD	Weight SE	% Lipids
Lake Stone 1	051012A	4	345.00	554.86	3.25	325-387	28.51	14.25	463-810	316.56	158.28	0.0
Lake Stone 2	051012B	4	330.50	488.31	3.25	304-370	30.87	15.44	362-700	87.01	43.50	0.0
Lake Stone 3	051012C	4	335.75	468.45	4.00	318-367	22.97	11.48	383-630	211.67	105.84	0.0
Bayou Chico	060315A	3	365.67	573.82	3.33	346-414	42.10	24.31	361-860	272.27	157.20	0.2
Langley-Bell 4H	060712A	3	296.00	272.30	3.67	281-312	15.52	8.96	243-307	32.13	18.55	1.9
Langley-Bell 4H	060712B	1	430.00	780.00	5.00	430						0.1
Fairfield Pond	060718C	3	361.67	601.45	3.00	315-415	50.33	29.06	374-890	263.26	151.99	0.1
Fairfield Pond	060720A	1	532.00	1970.0	6.00	532						0.2
Fairfield Pond	070225A	3	332.33	450		306-366	30.66	17.70	360-590	122.88	70.95	0.3

Okaloosa County

Site	Composite	n	Mean Length	Mean Weight	Mean Age	Length Range	Length SD	Length SE	Weight Range	Weight SD	Weight SE	% Lipids
Hurricane Lake	060629A	1	340	376.89	4.00	340			377			0.2

Table 3. Contaminant data for largemouth bass samples from lakes and ponds in Northwest Florida.

Santa Rosa County

Site	Composite	Hg	TEQ _{DF} ng/Kg	TEQ _P ng/Kg	TEQ _{DFP} ng/Kg	ΣPCBs ug/Kg	OCDF ng/Kg	OCDD ng/Kg	Total TCDF ng/Kg	Total TCDD ng/Kg
Bear Lake 1	051017A	0.41	0.0794	0.0224	0.1019	0.2405	0.3000	1.3000	0.3600	1.5490
Bear Lake 2	051017B	0.34	0.0787	0.0222	0.1009	0.1477	0.1300	0.2800	0.2740	0.4850
Bear Lake 3	051017C	0.34	0.0404	0.0217	0.0621	0.3045	0.1200	0.3300	0.2160	0.4080
Lake Kristina	060322A	0.43	0.0994	0.0549	0.1543	0.2375	<DL	0.6600	<DL	<DL
Cedar Lakes	060323A	0.84	0.1338	0.0579	0.1917	2.1385	<DL	0.8800	0.1300	<DL
Woodbine Springs 1	060511A	1.6	0.0541	0.0412	0.0953	0.9903	0.5200	4.3000	0.0430	<DL
Woodbine Springs 2	060511B	2.5	0.0453	0.0348	0.0801	0.5171	0.1800	0.4100	0.0640	<DL
Woodbine Springs 3	060511C	2.3	0.0509	0.0594	0.1103	1.3437	1.2000	2.9000	<DL	<DL
Tiger Pt GC	060603A	0.2	0.0533	0.0692	0.1225	4.1940	0.1800	0.4300	<DL	<DL
Tiger Pt WWT	060405A	0.58	0.1035	0.1216	0.2251	3.6187	<DL	0.5600	0.4300	<DL
Tiger Pt WWT	060612A	0.091	0.0270	0.2523	0.2793	10.9670	0.0185	0.0120	0.1900	0.0145
Tiger Pt WWT	070315A	0.13	0.1000			6.080				

Escambia County

Site	Composite	Hg	TEQ _{DF} ng/Kg	TEQ _P ng/Kg	TEQ _{DFP} ng/Kg	ΣPCBs ug/Kg	OCDF ng/Kg	OCDD ng/Kg	Total TCDF ng/Kg	Total TCDD ng/Kg
Lake Stone 1	051012A	0.51	0.0297	0.0224	0.0521	0.2133	0.1300	0.0000	0.2600	0.1000
Lake Stone 2	051012B	0.51	0.0312	0.0223	0.0534	0.1407	0.1100	0.2400	0.2300	0.3020
Lake Stone 3	051012C	0.58	0.0288	0.0224	0.0511	0.1101	0.1100	0.2300	0.2110	0.2300
Bayou Chico	060315A	0.035	0.0781	1.0320	1.1100	10.1025	0.0550	0.1000	0.0700	<DL
Langley-Bell 4H	060712A	0.96	0.0201	0.0286	0.0487	0.4763	0.0275	0.0135	0.0135	0.0090
Langley-Bell 4H	060712B	1.1	0.0286	0.0287	0.0573	0.4379	0.0160	0.0080	0.0080	0.0290
Fairfield Pond	060718C	0.57	0.0496	0.0396	0.0892	3.6039	0.0130	0.0320	0.0145	0.0335
Fairfield Pond	060720A	1.3	0.0807	0.1538	0.2344	8.2814	0.0115	0.0180	0.0185	0.3800
Fairfield Pond	070225A	0.66	0.0690			4.320				

Okaloosa County

Site	Composite	Hg	TEQ _{DF} ng/Kg	TEQ _P ng/Kg	TEQ _{DFP} ng/Kg	ΣPCBs ug/Kg	OCDF ng/Kg	OCDD ng/Kg	Total TCDF ng/Kg	Total TCDD ng/Kg
Hurricane Lake	060629A	0.42	0.0212	0.0262	0.0474	0.2528	0.025	0.0085	0.0085	0.098

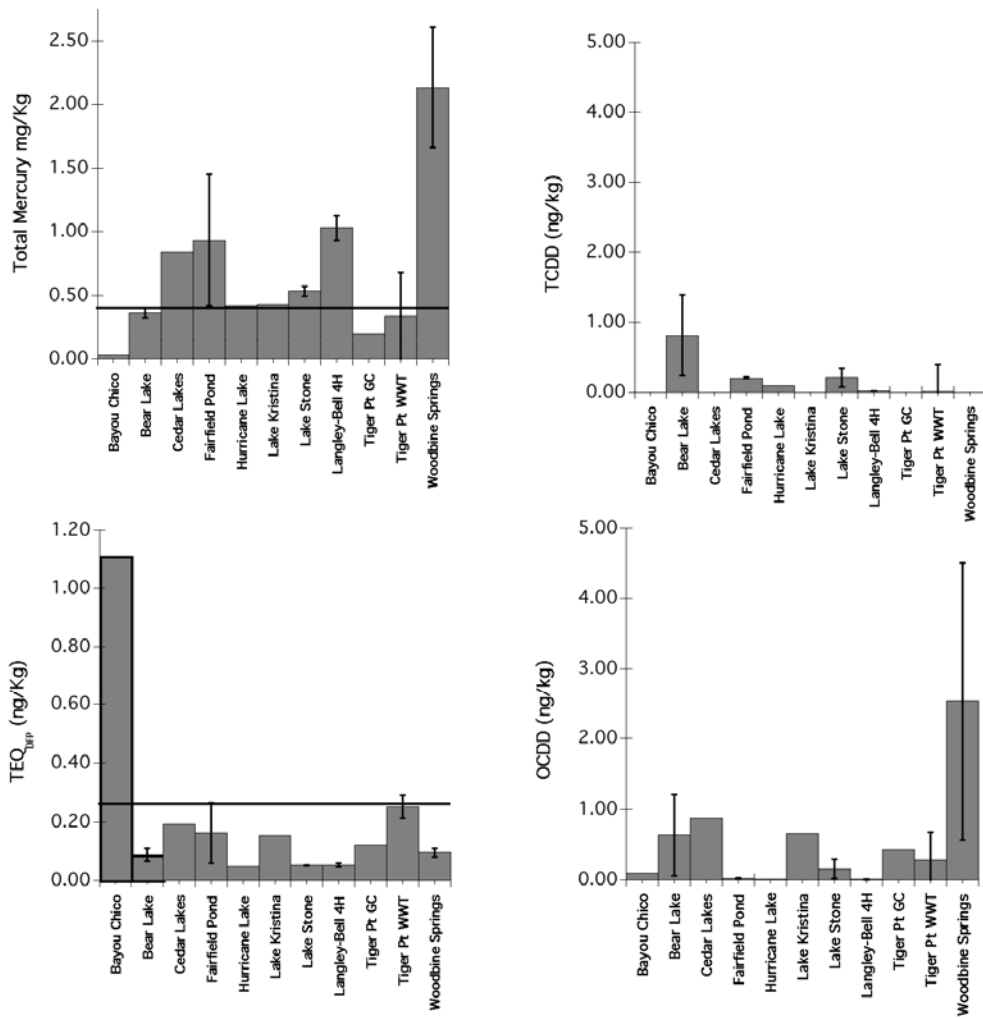


Figure 1. Summary data on contaminant levels in lake and pond bass in Northwest Florida.

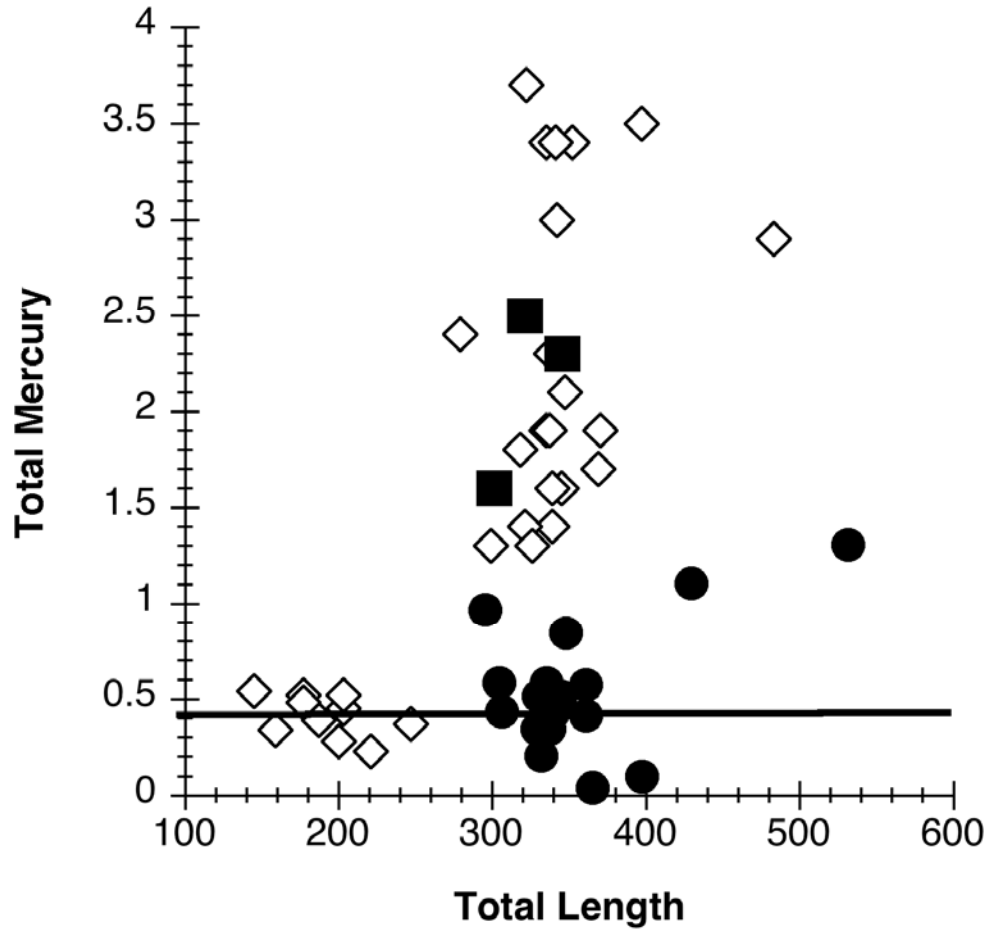


Figure 2. Mercury in bass data collected over Escambia, Santa Rosa, and Okaloosa Counties (solid circles and squares), compared to multiyear data from the Florida Fish and Wildlife Commission sampling of bass in Woodbine Springs (diamonds). The results from the UWF work for Woodbine Springs are indicated by the solid squares. Data from all other locations sampled by UWF are indicated by solid circles.

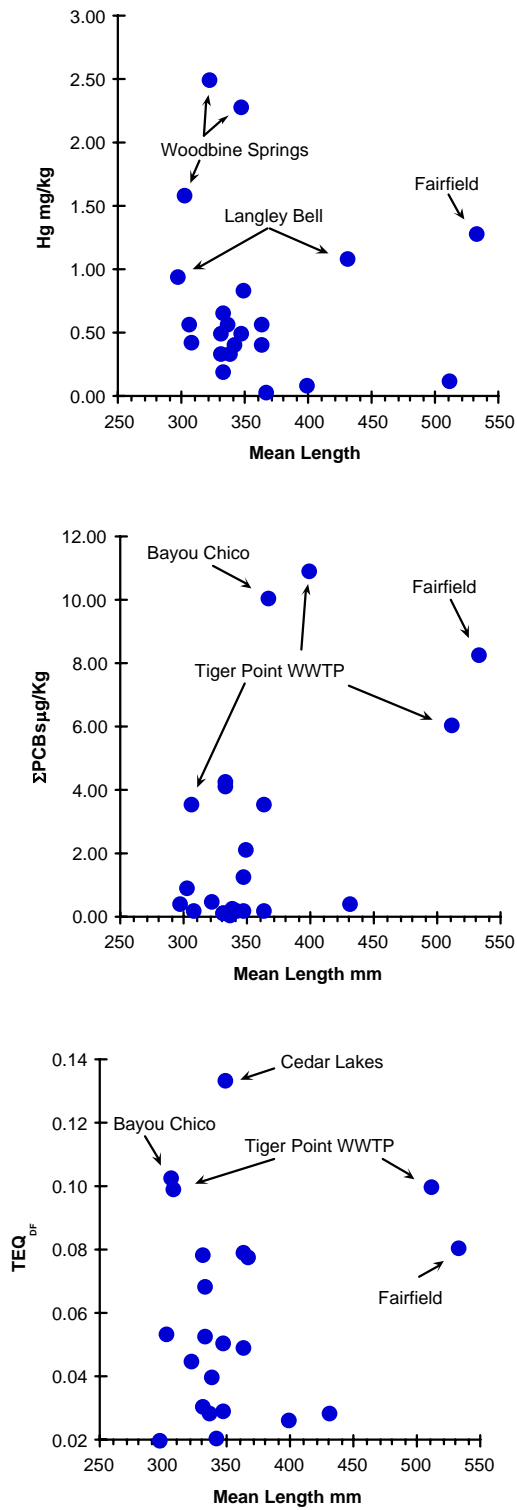


Figure 3. Contaminant concentrations in largemouth bass as a function of fish length as the mean for composites or single fish for larger specimens (Table 1).

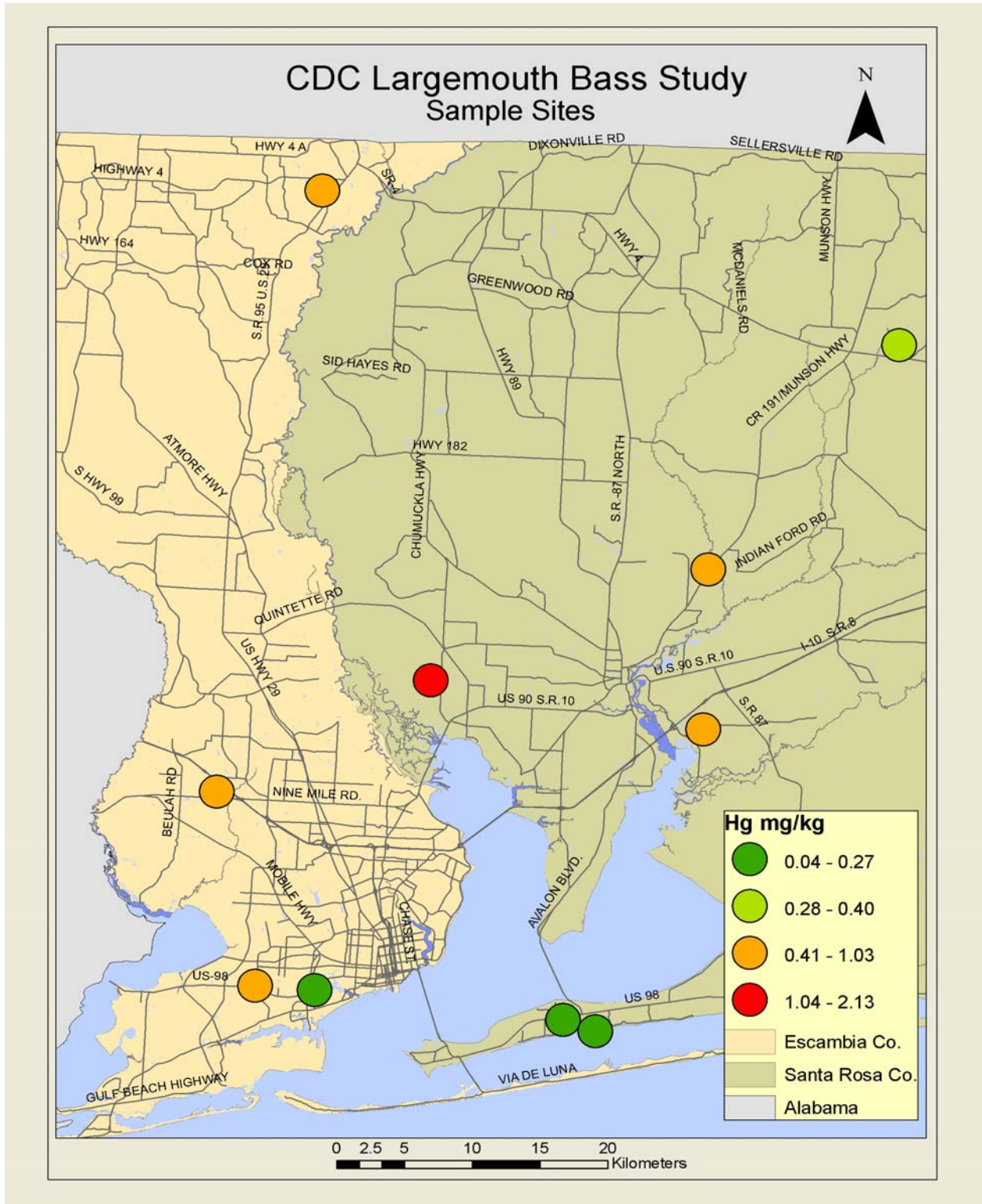


Figure 4. Geographic distribution of average mercury concentrations in largemouth bass from ponds over the northwest Florida region. Highest concentrations were recovered from Woodbine Springs (red symbol).

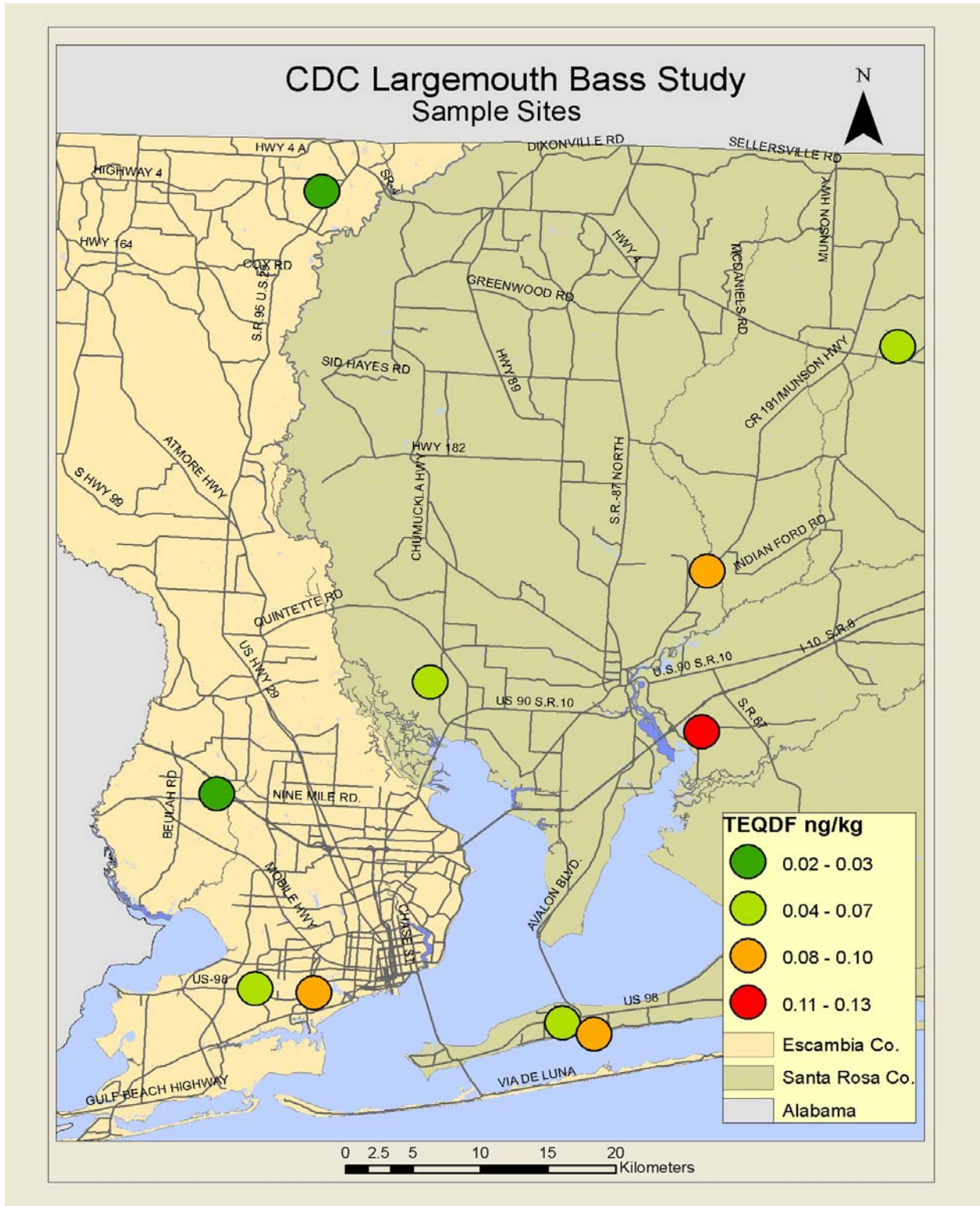


Figure 5. Geographic distribution of Dioxin-Furan TEQ values from largemouth bass tissue recovered from ponds over the northwest Florida region. Highest value was obtained from a sample at Cedar Lakes (red symbol).

Evaluation

The project goals were met by sampling and analyzing the contaminant loads of largemouth bass from 11 different lakes/ponds, including 10 locations that were not studied earlier. Several owners of private ponds/lakes allowed us to sample for this study, aiding in meeting our objectives.

The results are being prepared for publication in a professional journal. The data will be shared with the Florida Department of Health, county health departments, and the Florida Department of Environmental Protection. The final report, including site-specific data presented in a GIS map format, will be posted on the CEDB's website for public access. Knowledge of the contaminant loads would be helpful to recreational fishers to be selective in consuming largemouth bass collected from regional lakes and ponds.

The above outcomes are in accord with the goals of "Healthy People 2010" in the area of environmental health: 8-8: "increase the proportion of assessed rivers, lakes, and estuaries that are safe for fishing and recreational purposes;" and 8-10: "reduce the potential exposure to persistent chemicals by decreasing fish consumption levels."

Task 2. Evaluation of mercury levels in hair samples from women of child-bearing age in Escambia and Santa Rosa Counties, Florida.

Introduction

According to the U.S. EPA, nearly 75% of fish consumption advisories in the United States have been issued due to mercury contamination (U.S. EPA, 2003). Despite a significant reduction in the use and release of mercury from industrial processes since the 1970s, mercury contamination associated with increased fossil fuel combustion poses a growing contamination problem in many areas (U.S. EPA, 2000). The State of Florida has issued fish consumption advisories due to mercury levels for all four of the major rivers in Northwest Florida (Escambia, Perdido, Blackwater, and Yellow Rivers) for multiple fish species, including bluegill, redear, longear and spotted sunfish, warmouth, chain pickerel, largemouth bass, bowfin, and gar (FDOH, 2006). Likewise, 62 salt-water fish species carry consumption advisories for all coastal waters of Florida. Other fish species, such as speckled trout, cobia, and gafftopsail catfish, have also exhibited elevated levels of mercury in various locations in Florida (Adams et al., 2003a).

Previous studies have found that coastal populations commonly exhibit higher levels of methylmercury than inland populations, most likely as a result of higher fish consumption (Knobeloch et al., 2005; Mahaffey, 2005). The Mercury Deposition Network (MDN) of the National Atmospheric Deposition Program (NADP) has reported that over the past five years, the Gulf Coast and the Florida Peninsula received the highest levels of wet deposition of total mercury in the United States (NADP, 2007). The station that consistently records among the highest levels of the more than 85 MDN stations is located in Baldwin County, AL, which borders the far western Florida Panhandle. Although mercury is deposited primarily in its inorganic form, it is converted by microbial activity into methylmercury at particularly high rates in coastal wetlands, such as those found along the Gulf of Mexico (U.S.G.S, 2000). Thus, the high levels of atmospheric deposition and subsequent methylation, and high fish consumption

rates could potentially expose the human population in the Florida Panhandle to high levels of mercury, primarily through the consumption of regionally caught fish species.

The human nervous system is extremely vulnerable to methyl mercury (MeHg), and exposure to high levels of MeHg during the last two trimesters of pregnancy produces documented neurodevelopmental problems in children (McDowell et al., 2004). For example, higher maternal mercury levels are associated with lower infant cognition (Oken et al., 2005). Methyl mercury has a half-life of approximately 60 days in the human body, and therefore consumption of mercury-contaminated fish on a regular basis increases the risk of health problems, particularly in children (Knobeloch et al., 2005). The National Health and Nutrition Examination Survey (NHANES) reported that hair mercury levels in frequent fish consumers were three-fold higher for women and two-fold higher for children compared with nonconsumers (McDowell et al., 2004). Analysis of human hair for mercury is useful as an indicator of methylmercury exposures (Mahaffey, 2005). Methylmercury is incorporated into the hair follicle during growth (approximately 1 cm per month), and therefore hair-mercury concentrations reflect longer-term averages than levels in the blood (Budtz-Jorgensen et al., 2004; Mahaffey, 2005).

In the present study, we investigated mercury levels in human hair to assess the exposure of a highly susceptible segment of the local population in Northwest Florida- women of child-bearing age. Many of the fish consumption surveys that have been issued in the U.S specifically target this subset of the population because of the concerns that maternal intake of contaminants poses a major health risk to a developing fetus. In conjunction with the hair analysis, this study surveyed the fish consumption practices of the sampled volunteers as part of an overall dietary assessment and in an attempt to evaluate whether levels in hair reflect consumption rates. To our knowledge, no previous studies have investigated mercury levels in the local human population in the western Florida Panhandle.

Methods

Pregnant and non-pregnant women 16–49 years of age, who had resided in Santa Rosa and Escambia Counties, FL for at least one year, were included in the present study. This study was approved by the Institutional Review Board for Human Research Participants Protection of the University of West Florida. All participants provided their written informed consent. Subjects were recruited through written announcements that were distributed through local health clinics, newspapers, the University of West Florida website, community groups, and social clubs. Hair samples were collected at various locations throughout the sampling area by staff from the University of West Florida, the Escambia County Health Department, and the Santa Rosa County Health Department, following the methods of McDowell et al. (2004). Each participant was assigned an identification number. Approximately 100 strands of hair (~ 50 mg) were gathered and cut from the occipital region of the scalp using stainless steel surgical scissors. The ends of the hair strands closest to the scalp were placed in a 1.5 × 2 in. Post-it Note® (3M Corporation). The paper was folded in half over the hair and the end of hair closest to the scalp was marked on the paper with an arrow. A plastic paper clip was placed over the paper to secure the hair sample and the sample was then placed in a resealable plastic bag labeled with the participant identification number. Hair samples were stored at room temperature until shipping. Two inches of hair, representing two to six months of growth, were analyzed for total mercury.

The concentration of mercury in the hair study samples was determined at RTI International (Research Triangle Park, N.C.) by cold vapor atomic fluorescence spectrometry (CVAFS). Prior to digestion, the hair samples were treated with acetone to minimize the potential for external contamination. Small hair sample aliquots (approximately 5 mg) were then digested using high-purity acids and oxidants in an oven heated to 90°C. In addition to matrix decomposition, this procedure served to convert mercury present in the samples to the Hg (II) form. When acidified samples containing Hg (II) were introduced into the CVAFS instrument and subjected to a reducing agent, Hg (0) vapor was formed. This vapor was passed directly to Hg-specific atomic fluorescence detector with a stream of argon gas. A thorough description of this analytical method is provided by Pellizzari et al (1999).

In addition to the study samples, numerous quality controls were processed with each sample batch to continuously monitor analytical performance. These controls included method blanks to monitor the analytical background from the reagents and the procedure, mercury-fortified method controls to assess analyte recovery in the absence of sample matrix, certified reference materials to monitor accuracy, and duplicate preparations and analyses to assess sample preparation and analysis precision. Quality control data using the employed method for this investigation were consistent with the data collected by this laboratory in support of the 1999-2000 National Health and Nutrition Examination Survey (NHANES) (McDowell et al., 2004). The method quantitation limit (MQL) = 0.0244 µg Hg/g hair. The average recovery for low level method control samples (n=26) containing 20 pg Hg/mL was 91.8% with a 14% RSD. The average recovery for high level method control samples (n=25) containing 60 pg Hg/mL was 95.6% with a 13% RSD. The average recovery for hair certified reference material (NIES-13) (n=71) was 114% with a 13% RSD. Fifty-nine samples were prepared in duplicate of which six were less than the MQL. Fifty-five (93%) of the duplicate samples had % RSDs less than 20% and 36 (61%) had % RSDs less than 10%.

Each participant was given a questionnaire, which was filled out at the time of sampling. The questionnaire inquired about the participant's use of hair dye or hair relaxer and seafood consumption practices. Seafood consumption practices were evaluated as follows: 1) number of meals (0, 1, 2, 3, >3) consumed during the 30 days immediately prior to sampling, 2) number of meals (0, 1, 2, 3, >3) consumed over a 30 day period, two months prior to sampling, and 3) rating of seafood consumption over the 60 day period prior to sampling (normal, lower than normal, higher than normal). Participant awareness of Florida's fish consumption advisories was also noted. In addition, each volunteer was interviewed regarding demographic information. Demographic data included zip code of residence, race/ethnicity, length of residency, age, and pregnancy status.

Statistical Analysis

Relationships between mercury levels in hair, participant demographic data, and seafood consumption practices were assessed by generalized linear models (GLM) using SPSS version 13 (SPSS Inc., Chicago, IL). Mercury hair data were log transformed to satisfy the assumptions of the GLM. Data in the manuscript are reported as back-transformed variables (geometric mean=GM). Arithmetic means (AM) and related descriptive statistics are also reported for comparison purposes.

Results

Hair samples were obtained from a total of 601 women, of whom 449 (74.5%) classified themselves as White, 65 (10.8%) as Black, 28 (4.6%) as Asian, 37 (6.1%) as Hispanic, 12 (2.0%) as Other, and 10 (1.9%) did not provide race data. The majority of the participants lived in Escambia County, FL (454 or 75.6%) and the remainder lived in Santa Rosa County, FL (147 or 24.4%). The age distribution of the participants was as follows: 25 (4.2%) were 15-19 years old, 187 (31.1%) were 20-29 years old, 185 (30.8%) were 30-39 years old, and 204 (33.9%) were 40-49 years old. A total of 83 (13.8%) women reported being pregnant, of whom 29 (34.9%) were in the first trimester, 28 (33.7%) were in the second trimester, and 22 (26.5%) were in the third trimester. Four women did not report pregnancy stage. One woman did not answer the participant questionnaire completely. Table 1 summarizes the responses to the participant questionnaire.

Table 2 summarizes mercury concentrations in the entire sampling group and by participant characteristics. Of the 601 women sampled, 95 (15.8%) were found to have mercury levels that exceeded the US EPA advisory level of 1.0 $\mu\text{g/g}$. The percentage of women in the Pensacola cohort exceeding the EPA advisory level increased with seafood consumption (Figure 1). Of the 95 women who exceeded the advisory level, 60 (62.5%) ate more than three fish/seafood meals in the 30 days prior to sampling and 75 (78%) ate three or more meals in that period. Mercury levels in 87 of the 601 participants (14.5%) were between 0.5 and 0.99 $\mu\text{g/g}$ and 420 of the participants (70%) had mercury levels below 0.5 $\mu\text{g/g}$. In the 0.5-0.9 $\mu\text{g/g}$ group, 43 women (49.4%) ate more than three seafood meals in the 30 days prior to sampling and 57 (65.5%) ate three or more meals in that period. Of the participants with <0.5 $\mu\text{g/g}$ mercury, 106 (25%) ate more than three seafood meals and 170 (40.4%) ate three or more seafood meals in the 30 days prior to sampling. Approximately 31% of the women reported being aware of the Florida Fish Consumption Advisory. Black women were the least likely to know about the advisory and Hispanic women had the highest awareness (Table 3). Fewer pregnant women knew about the fish advisory than non-pregnant women (Table 3).

We found no significant differences in mercury levels between age groups, between women who were pregnant and those who were not, or between women who used hair dye and those who did not. Mercury levels were found to differ significantly between racial/ethnic groups ($p<0.0001$). Age and seafood consumption practices were included in the final model. Post-hoc analysis using a Bonferroni correction found that Black women had significantly lower mercury levels than any other group ($p<0.02$). Women who were aware (GM=0.23 $\mu\text{g/g}$) of the Florida Fish Consumption Advisory had significantly lower mercury levels ($p=0.043$) than women who were unaware (GM=0.31 $\mu\text{g/g}$) of the advisory (seafood consumption practices and race were included in the model).

Seafood consumption rating (usual, lower than normal, higher than normal) was found to significantly affect mercury levels ($p=0.04$), when age, race, and seafood consumption practices (30 days prior and 30 day period, two months prior to sampling) were included as covariates in the final model. Women who reported eating higher than normal amounts of fish or shellfish over the 60 days prior to sampling had significantly higher mercury concentrations than women who reported eating a “less than normal” amount ($p=0.034$, Bonferroni correction). The number

of seafood meals consumed in the 30 days immediately prior to sampling significantly affected mercury levels in the participants ($p < 0.0001$). Age, race, and number of meals consumed over a 30 day period, two months prior to sampling were included in the model. Women who consumed one, two, three and greater than three meals had significantly higher mercury levels than women who did not consume any fish/shellfish in the 30 day period prior to sampling ($p < 0.001$, Bonferroni correction). The number of seafood meals consumed over a 30 day period, two months prior to sampling was also found to have a significant effect on mercury levels ($p = 0.033$). Age, race, and number of seafood meals consumed 30 days prior to sampling were included in the final model. Women who ate three meals were found to have significantly higher mercury levels than women who ate no seafood in that period ($p = 0.014$).

Discussion

The nutritional benefits of consuming fish have been well publicized in recent years and they include the presence of high-quality protein, vitamins and other essential nutrients, low levels of saturated fat, and high levels of omega-3 polyunsaturated fatty acids (Domingo et al., 2007). Likewise, longer gestation, increased birth weight, reduced risk of intrauterine growth retardation, and lower prevalence of pregnancy-induced hypertension have been reported in women who consumed high levels of fish during pregnancy (Xue et al., 2007). In fact, maternal seafood consumption during pregnancy of less than 340 g per week has been associated with increased risk of low verbal intelligence quotient (IQ) and increased risk of suboptimum outcomes for prosocial behavior, fine motor, communication, and social development scores (Hibbeln et al., 2007). However, the U.S. EPA has reported that fish and shellfish are the main sources of methylmercury exposure to humans (U.S. EPA, 2007) and the agency has therefore issued a joint advisory with the U.S. Food and Drug Administration (FDA) that warns pregnant women, women who may become pregnant, nursing mothers, and young children to avoid some types of fish and eat fish and shellfish that are lower in mercury (U.S. EPA, 2004). Sensitivity of the fetal nervous system to methyl mercury is well-documented and other effects such as a relationship between elevated mercury levels in pregnant women and delivery before 35 weeks' gestation have also been reported (Mahaffey, 2005; Xue et al., 2007).

Mean mercury levels (GM=0.25 $\mu\text{g/g}$ and AM=0.56 $\mu\text{g/g}$) for the entire cohort in the current study were slightly higher than those reported by McDowell et al. (2004) from 1726 randomly selected women of child-bearing age (GM=0.20 $\mu\text{g/g}$, AM= 0.47 $\mu\text{g/g}$), who participated in the 1999-2000 National Health and Nutrition Examination Survey (NHANES). Two previous studies have been identified that analyzed hair samples from women in Florida (Knobeloch et al., 2005; Patch et al., 2005). Knobeloch et al. analyzed a small sample of child-bearing age women ($n=8$) and reported an AM of 0.40 $\mu\text{g/g}$. Patch et al. (2005) reported an AM of 1.03 $\mu\text{g/g}$ in 156 women of childbearing age from across Florida. Knobeloch et al (2005) did not report the specific regions of Florida from which samples originated, whereas the majority of samples in the Patch et al. (2005) study originated from the lower Florida Peninsula and not the Panhandle. A recent study by Warner (2007) that examined mercury levels in coastal Alabama anglers and residents, reported an AM 0.55 $\mu\text{g/g}$ and a median of 0.37 $\mu\text{g/g}$ in women.

Although the levels of mercury we observed for the Pensacola cohort exceeded those reported nationally for the NHANES and the Knobeloch et al. (2005) studies, the levels were lower than

those reported in the Patch et al. study. That national levels are lower than those we found is not unexpected, since women who live in coastal areas generally consume greater amounts of fish than those who live in interior regions and thus are more likely to exhibit higher mercury levels. Warner (2007) reported a nearly identical AM to that reported in the present study. Although the sample size in her study was significantly smaller (n=19) than the Pensacola cohort, the women sampled in her study are more likely to be exposed to the same sources of seafood, due to the proximity of Pensacola to Dauphin Island, AL, the site of sample collection.

However, reasons for the differences between our results and the Patch et al. study are more obscure. Of the 156 women sampled in the Patch et al. study, 94% were from the Florida Peninsula. Theoretically, regional differences in the mercury levels of locally caught fish and differences in consumption rates could account for the observed levels. Adams et al. (2003b) found that mercury levels in several marine fish species from the southern Florida Gulf Coast and the Florida Keys/Florida Bay regions were often higher than mercury levels in fish of the same species and size sampled from the Atlantic coast of Florida. However, the actual sources of the fish consumed by the participants in the respective studies are unknown, since most reported consuming seafood bought from a store or restaurant, rather than catching the fish themselves. The differences may also result from variations in consumption rates and portion sizes in study participants. However, we were unable to directly compare consumption rates between the studies because Patch et al. (2005) did not report regional consumption rates for women of childbearing age and we did not specifically define portion size in our questionnaire.

Alternatively, participation in the Patch et al. (2005) study required the enrollees to pay a fee, whereas participation in our study and the Warner study was free. Several studies have reported that higher mercury levels are associated with higher socioeconomic status (college education and annual household income) due a participant's ability to buy more seafood, especially the more expensive predatory fish that contain higher levels of mercury (Knobeloch et al., 2005; Mahaffey, 2005; Xue et al., 2007). Therefore, participation in the Patch et al. (2005) study was potentially skewed towards persons of higher socioeconomic status, which could in part account for the higher levels of mercury.

The United States Environmental Protection Agency (EPA) has set a Reference Dose of 0.1 mg/kg/day of mercury for children and women of child-bearing age, which corresponds to a hair mercury concentration at 1.0 µg/g (U.S. EPA, 1997). Consumers with hair mercury levels above this dose are advised to stop consuming fish known to have high mercury levels. According to the NHANES study, approximately 12% of women exhibit hair mercury levels above the EPA advisory level, nationally (McDowell et al., 2004). Nearly 16% of the women monitored in the present study exhibited hair mercury levels above the advisory level. Consistent with the higher mean levels found in their analyses, Patch et al. (2005) found that mercury levels in 33.9% of the women sampled in Florida exceeded the advisory level. In contrast, Warner (2007) reported that only 10.5% of the women in her study exceeded the advisory level. These data suggest that regional differences in exposure may be present even within a single state due to the type of seafood consumed, origin of the consumed seafood and consumption rates.

In contrast to the NHANES study and others (Patch et al., 2005; Xue et al., 2007), we did not find a significant relationship between age and mercury levels in the Pensacola cohort, despite an

increasing trend in GMs with age. However, race was a significant factor in predicting mercury levels. As has been reported previously (McDowell et al., 2004; Patch et al., 2005; Xue et al., 2007), Black women were found to have significantly lower levels of mercury in the present study than all other race categories. In contrast to previous studies, women who classified themselves as Asian did not have the highest mercury levels in our cohort (based on GM).

We found a significant relationship between the level of seafood consumption in women of childbearing age and mercury levels in hair samples. Mercury levels increased with the number of seafood meals. Interestingly, even eating one meal in the 30 day period prior to sampling caused a significant elevation of hair mercury levels over those in women who did not consume any seafood prior to sampling. Because we analyzed the samples for total mercury and because seafood is the primary source of organic mercury for the general population, the mercury levels in those women who did not consume any seafood are likely to represent ingested inorganic mercury or organic mercury that has been deposited on the sampled hair (Mahaffey, 2005). The levels that we observed in the five consumption categories (0, 1, 2, 3, >3 meals in the 30 days prior to sampling) were nearly identical to those observed by McDowell et al. (2004) and Patch et al. (2005) for the same categories. This suggests that despite the differences in overall means/medians between the three studies, in general, one can expect similar mercury levels at a given rate of consumption. Extremes at each level of consumption are driven by the types of fish consumed, that is, elevated mercury can result from moderate or low level consumption of fish with high mercury concentrations or from high rates of consumption of fish with moderate mercury concentrations.

Knowledge of the Florida fish consumption advisory was fairly low (31%) in the Pensacola cohort. However, those women who knew about the advisory exhibited statistically lower levels of mercury than those who did not know about the advisory. Despite having the lowest awareness of the advisory, Black women had the lowest mercury levels, potentially because the fish species most commonly consumed by the women sampled carry a lower body-burden of mercury. Our data suggest that although a lack of advisory awareness amongst the Black women in our cohort did not result in exceptional mercury levels, in general, knowledge of the advisory does appear to impact concentrations. Overall, advisory awareness was higher in the Pensacola cohort than that reported by Knobloch et al. (2005) for a national cohort of women (20%). In that study, Asian women had the lowest awareness (7%), followed closely by Black (11%) and Hispanic (12%) women. The data from the present study also demonstrated that pregnant women in the Pensacola area were much less likely to know about the consumption advisory than non-pregnant women.

Recent studies have shown that nationally, women of childbearing age, especially women who are or plan to become pregnant, have been eating less seafood in an attempt to reduce mercury intake. A study by Oken et al. (2003) found that in response to the national advisory issued jointly by the FDA and EPA, pregnant women reduced fish consumption to levels below beneficial amounts. That is, the women didn't just reduce intake of species that have higher mercury levels. Instead they reduced consumption of all fish. The risks to children of mothers whose fish consumption was reduced or eliminated during pregnancy from the loss of nutrients have been found to be greater than the risks of harm from exposure to trace contaminants such as mercury (Hibbeln et al., 2007). Therefore, the benefits of fish consumption must also be taken

into account when offering information on fish consumption advisories to women of child-bearing age, and although as we have found in the Pensacola cohort, mercury levels can be reduced in women who are aware of fish advisories, they should also be made aware of the potential negative impacts of improperly reducing fish consumption. The first step appears to be

Participant Characteristics	Geometric mean Hg	95% CI	No.	% of Total
1. Hair Treatment 30 days prior				
yes	0.26	0.23 - 0.29	195	33%
no	0.24	0.20 - 0.28	405	68%
2. Fish consumption (meals) in 30 days prior to sampling				
0	0.08	0.07 - 0.10	82	14%
1	0.19	0.15 - 0.24	96	16%
2	0.20	0.17 - 0.24	119	20%
3	0.29	0.23 - 0.37	93	16%
>3	0.47	0.40 - 0.54	210	35%
3. Fish consumption (meals) over a 30 day period, two months prior to sampling .				
0	0.10	0.08 - 0.13	93	16%
1	0.15	0.12 - 0.19	89	15%
2	0.22	0.18 - 0.27	131	22%
3	0.36	0.28 - 0.45	92	15%
>3	0.45	0.38 - 0.52	196	33%
4. Fish Consumption over 60 days				
Usual	0.26	0.23 - 0.29	473	79%
Higher	0.27	0.19 - 0.36	58	10%
Lower	0.21	0.15 - 0.28	70	12%
5. Type of fish or shellfish consumed.				
Saltwater	*	*	281	47%
Freshwater	*	*	105	18%
Shellfish	*	*	332	55%
6. Source of Fish				
Caught	*	*	94	16%
Store	*	*	365	61%
Restaurant	*	*	389	65%
7. Source of Shellfish				
Caught	*	*	36	6%
Store	*	*	295	49%
Restaurant	*	*	354	59%
8. Awareness of Florida Fish Consumption Advisory				
Yes	0.23	0.20 - 0.26	187	31%
No	0.31	0.26 - 0.36	413	69%

* Means are not provided because participants marked as many of the options as they felt warranted. actually getting appropriate information to women. Our cohort has demonstrated that previous efforts have been only minimally successful.

Table 1. Summary of responses to participant questionnaire and geometric means and 95% CI for mercury levels ($\mu\text{g/g}$) in hair of the respondents.

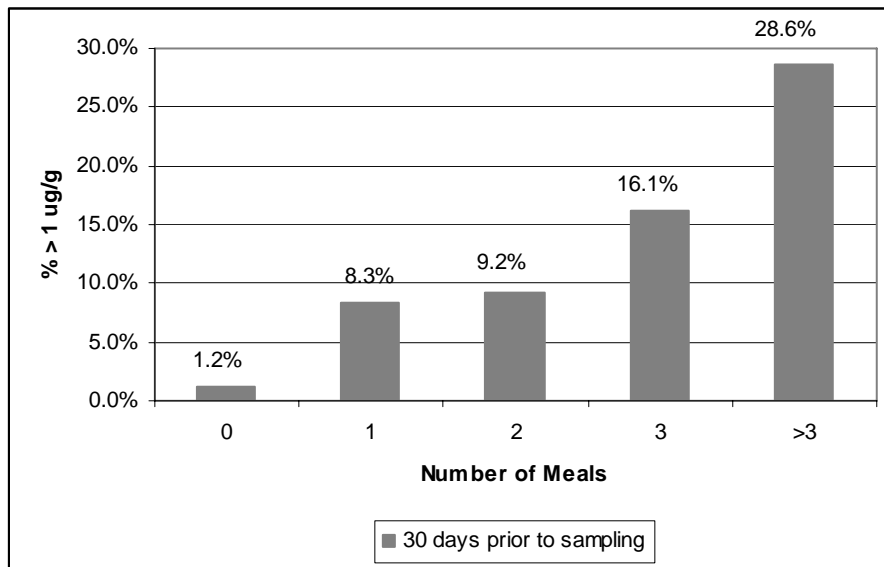
Table 2. Mercury concentrations in the entire sampling group and by participant characteristics. Back-transformed and untransformed statistics are provided for comparison.

	N	Back Transformed (Geometric)				Untransformed (Arithmetic)			
		Mean	95% CI	Minimum	Maximum	Mean	Median	Std. Dev.	95% CI
Total	601	0.25	0.22 - 0.28	0.02	22.14	0.56	0.27	1.17	0.46 - 0.64
Asian	28	0.28	0.15 - 0.51	0.02	22.14	1.36	0.29	4.22	-0.2 - 2.99
Black	65	0.11	0.08 - 0.14	0.02	2.67	0.22	0.10	0.39	0.12 - 0.32
Hispanic	37	0.26	0.18 - 0.37	0.02	2.30	0.46	0.23	0.53	0.28 - 0.63
Not Reported	10	0.40	0.21 - 0.73	0.10	1.26	0.56	0.41	0.44	0.26 - 0.85
Other	12	0.61	0.26 - 1.42	0.05	10.65	1.52	0.52	2.92	-0.3 - 3.37
White	449	0.28	0.24 - 0.30	0.02	4.12	0.54	0.30	0.66	0.47 - 0.59
Not Pregnant	515	0.27	0.23 - 0.29	0.02	22.14	0.58	0.28	1.17	0.47 - 0.67
Pregnant	83	0.20	0.15 - 0.25	0.02	10.65	0.43	0.19	1.19	0.17 - 0.69
16-19 years old	25	0.19	0.10 - 0.32	0.02	10.65	0.69	0.14	2.10	-0.1 - 1.55
20-29 years old	187	0.21	0.17 - 0.24	0.02	3.56	0.43	0.24	0.57	0.34 - 0.51
30-39 years old	185	0.26	0.21 - 0.31	0.02	22.14	0.64	0.28	1.75	0.38 - 0.89
40-49 years old	204	0.32	0.26 - 0.36	0.02	4.03	0.57	0.32	0.66	0.48 - 0.66

Table 3. Fish advisory awareness according to race and pregnancy status.

Group	FishAdvisory		% Aware
	Unaware	Aware	
Asian	21	7	25.0%
Black	52	13	20.0%
Hispanic	24	13	35.1%
Not Reported	8	3	27.3%
Other	8	4	33.3%
White	300	147	32.9%
Pregnant	347	168	32.6%
Not Pregnant	64	19	22.9%
Total	413	187	31.2%

Figure 1. Percentage of women in the Pensacola cohort exceeding the EPA advisory level based on seafood consumption



Evaluation

Our goal was to recruit 400 women for the hair mercury determinations. Publicity of information on this project through diverse media (newspapers, radio and television, and websites) elicited substantial community interest, and we exceeded our goal by recruiting 601 women for this study, representing the following ethnicity: White (74.6%), Black (10.8%), Asian (4.6%), and Hispanic (6.1%) – reflecting the regional population profile.

We sent to each participant a letter containing the results of mercury analysis, how the determined Hg level compared with the EPA reference value, and a copy of the latest Florida DOH Fish Consumption Advisory, to enable them to make informed choices of fish consumption. Individuals whose hair Hg levels were $\geq 5\mu\text{g/g}$ were re-examined and referred to their physician for further follow-up. The results from client satisfaction survey indicate that the participants were very satisfied (see attached summary).

We presented a progress report (poster presentation) of hair Hg results at the Florida Statewide Epidemiology Seminar on 10/31/2006. Our findings were highlighted in epidemiology updates posted by the Florida DOH at the URL below.

http://www.doh.state.fl.us/disease_ctrl/epi/EPI_Updates/2007/01_25_2007.pdf

We plan to publish our results in a professional journal. The final report will be posted on the CEDB's website for public access.

The above outcomes are pertinent to Goal 8-25, "(Developmental) Reduce exposure to the population to pesticides, heavy metals, and other toxic chemicals, for "Healthy People 2010."

Hair Hg monitoring project: results of client satisfaction survey

Question		Was the location of the clinic convenient for you?	How did you feel about the waiting time in the clinic?	Were you satisfied with the treatment you received by the staff during your visit?	Satisfied with Clerical Staff?	Satisfied with Nursing Staff?	Satisfied with Physician?	How did you feel about the services you received during your visit?	Were you satisfied with the privacy provided for the interview?	Did you feel satisfied that the information shared would remain confidential?
N	Valid	562	555	558	454	492	271	547	553	551
	Missing	52	59	56	160	122	343	67	61	63
Mean		4.77	4.79	4.84	4.81	4.86	4.81	4.83	4.82	4.83
Minimum		2	2	1	1	3	3	1	2	2
Maximum		5	5	5	5	5	5	5	5	5

Legend 1= Unsatisfied 2= Less than satisfied 3= Satisfied 4= Somewhat Satisfied 5= Very Satisfied

Task 3. Health screening profile of a community near the Escambia Wood Treatment Co. Superfund site in Pensacola, FL.

Introduction

The Escambia Treating Company (ETC) operated a wood treating facility in eastern Pensacola, FL from 1942 to 1982. This abandoned facility was added to the U.S. Environmental Protection Agency's (EPA) National Priorities List in 1994, since the soil and groundwater were found to be contaminated by pollutants of concern- pentachlorophenol (PCP), polychlorinated dibenzo-p-dioxins and polychlorinated dibenzofurans (PCDD/F), polycyclic aromatic hydrocarbons (PAHs), arsenic, and lead. Polychlorinated Biphenyls (PCBs) are also known to be present at the ETC site. In view of potential health risks from exposure to contaminants, the EPA relocated approximately 361 families from the neighborhoods north and south of ETC during 1997-2003. Recently, the EPA collected additional soil samples near the ETC site in a previously untested area called Clarinda Triangle, and found elevated levels of several contaminants of concern (ATSDR, 2005). Clarinda Triangle is an area of mixed residential and commercial properties near the ETC site, which is bordered to the west by Pace Boulevard, to the east by Palafox Street, to the south by Clarinda Lane, and to the north by West Loretta Street (Figure 1). The Clarinda Triangle area soil samples had arsenic, dioxins, lead, and polycyclic aromatic hydrocarbons (PAHs) above the Florida Department of Environmental Protection's (DEP) Soil Cleanup Target Levels (SCTLs) for residential areas (ATSDR, 2005). However, the Florida DOH categorized the Clarinda Triangle area surface soil as "No Apparent Public Health Threat" and stated that the highest measured levels of arsenic, dioxins, lead, and PAHs were unlikely to have non-cancer health effects on either children or adults (ATSDR, 2005).

The DOH assessment was based solely on soil levels and despite their conclusions on the health threat, the DOH issued the following warning to residents: "residents and workers should avoid hand-to-mouth contact with surface soil, and should avoid consuming vegetables and fruits grown in contaminated soil, especially root crops. Residents and workers should avoid working in dusty conditions. Persons who are not able to avoid dusty conditions should use a dust mask." Because the ETC plant ceased operation in 1982 and the Clarinda soil assessment was conducted 22 years later, soil levels of contaminants are likely to have been higher at the time of plant closure. Thus, the potential exists for long-term residents of Clarinda Triangle to have accumulated significant levels of the ETC contaminants through the types of exposures described in the DOH warning. We performed clinical evaluations and biomonitoring for contaminants of concern in residents of the Clarinda Triangle area. Biomonitoring included analyses for PCDD/F, and dioxin-like PCBs (dl-PCBs) in serum and arsenic in urine.

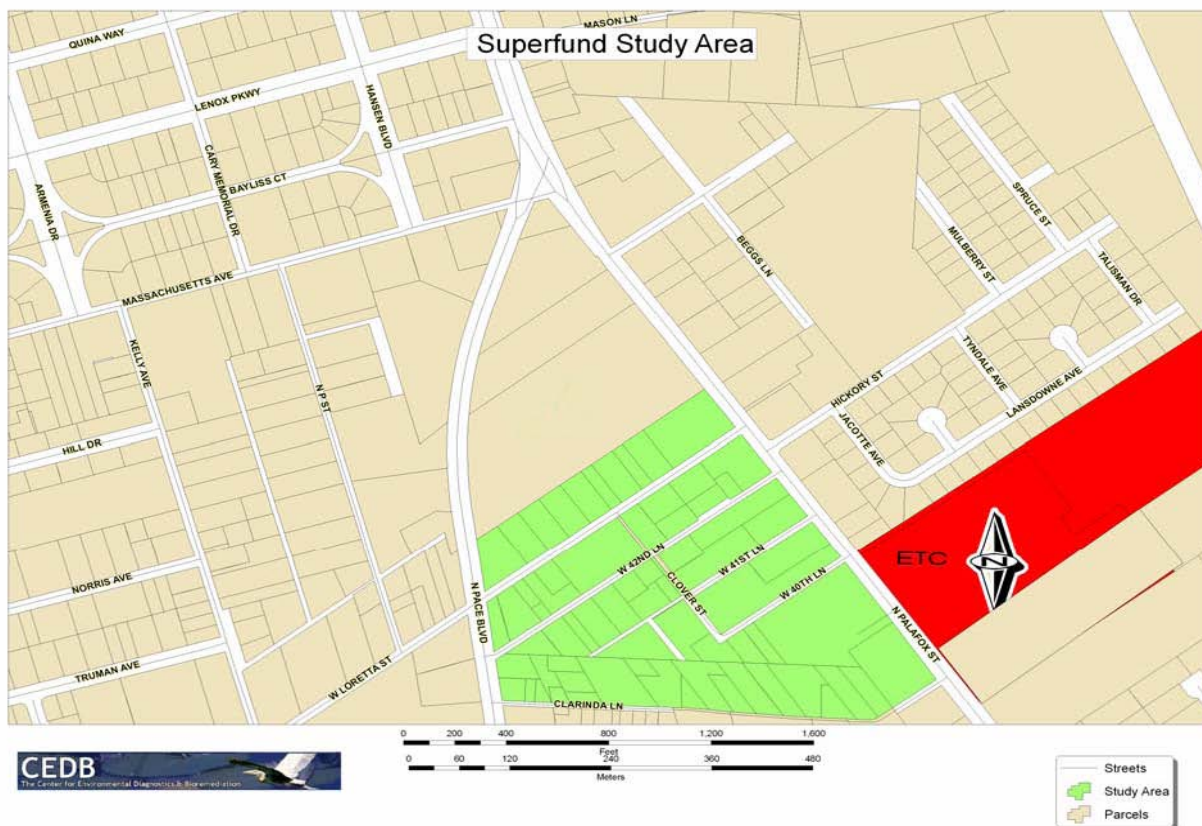


Figure 1. Location of Clarinda Triangle study area relative to the ETC Superfund site.

METHODS

We recruited current and former residents of the area known as Clarinda Triangle, which neighbors the ETC Superfund site. The study plan was reviewed and approved by the University of West Florida Institutional Review Board. All participants were provided detailed information on the study and completed appropriate consent forms. Participants underwent a physical exam conducted by a nurse practitioner and were queried about their medical history using a fixed panel of questions. These questions investigated demographics, exposure history, and health conditions such as cancer, hypertension, diabetes, alcohol usage, and smoking history, among others. Blood and urine specimens were collected for routine tests (Complete Metabolic Profile, lipid panel, thyroid panel, Complete Blood Count with differential, urinalysis, and hepatitis panel). Urine was also analyzed for total and inorganic arsenic using inductively-coupled plasma-mass spectrometry (ICP-MS) and flow injection atomic absorption spectrometry (FIAS), respectively. These samples were analyzed by a commercial laboratory, Laboratory Corporation of America (Labcorp, Pensacola, FL).

Serum samples were also collected for analysis of PCDD/F and dl-PCBs. Participants were requested to abstain from eating a fatty meal prior to sampling. A phlebotomist collected blood in eight 10 ml Monoject collection tubes (Sherwood Medical Co., St. Louis, MO) using a

standardized protocol. Collection tubes were labeled and placed upright at room temperature for clotting. After 30 minutes, samples were centrifuged for 15 minutes at 1000 x g. Using a disposable glass pipette, 20-30 ml of serum was transferred from each tube into one pre-cleaned 30 ml glass vial with a Teflon coated top, frozen at -20°C and shipped overnight on dry ice to Analytical Perspectives (Wilmington, NC) for PCDD/F and dl-PCB analysis.

Fasting blood samples were also drawn for total lipid analysis. The sample was drawn into a 10 ml Monoject collection tube and subsequently separated into two Corvac blood collection tubes (6 ml; Sherwood-Davis & Geck, St. Louis, MO) for duplicate analysis of total lipids. The samples were allowed to clot for ~ 30 minutes, centrifuged for 15 minutes, and analyzed for total lipids by LabCorp (Pensacola, FL) using a colorimetric assay (Bragdon, 1951).

Analysis of PCDD/F and DL-PCBs

The serum samples were analyzed for seventeen 2,3,7,8-substituted PCDD/F congeners and 12 dl-PCBs (PCB-77, PCB-81, PCB-105, PCB-114, PCB-118, PCB-123, PCB-126, PCB-156, PCB-157, PCB-167, PCB-169, PCB-189) using an in-house HR-MS method known as WHO-2S, which is a modification of USEPA Methods 8290B and 1668A. In this method, two separate extracts were produced for each sample and analyzed concomitantly on two separate columns using the same GC temperature and MS monitoring programs. Extraction standards (1 ng for each congener and 2 ng for $^{13}\text{C}_{12}$ -OCDD/F dissolved in 10 μL toluene or 1 mL acetone plus acetone rinses) were added to room temperature serum samples. After homogenization, formic acid was added to each sample and mixed on a shaker. Hexane liquid-liquid partitioning extraction was then carried out. The hexane extract was fortified with cleanup standards (CS) and fractionated on a multi-layered acid/base-coated silica gel column in tandem with a Florisil (PCDD/F) or carbon column (dl-PCBs). The fractions were collected and prepared for GC/MS by the addition of injection standards (JS). The final extract (1 μl) was injected in a splitless mode onto a 60 m DB-5MS (PCDD/F) or a 30 m DB-1 capillary column coupled to a magnetic sector instrument and analyzed under high-resolution GC (Agilent 6890 Series; Palo Alto, CA) and MS conditions (Waters AutoSpec Ultima; <100 ppm mass resolution at 5% peak width). Results were lipids adjusted and expressed as picograms per gram serum lipid (pg.g lipid $^{-1}$).

Results

Demographics of the 31 participants are summarized in Table 1. The majority of the participants were African-American, closely reflecting national census data for the neighborhood surrounding the ETC site (ATSDR, 1995). Participant age ranged from 13 to 80 years (mean = 47 years) and length of residency ranged from 1 to 52 years (mean= 21.0 years).

Table 1. Demographic profile of the 31 participants in the present study.

Characteristic		# Participants	% of Participants
Race	African-American	30	97%
	Caucasian	1	3%
Gender	M	12	39%
	F	19	61%
Age	Range = 13-80 years		
	<20	1	3%
	20-40	8	26%
	41-60	17	55%
	>60	5	16%
Length of Residency	minimum=1 yr maximum=52 yr mean=17.1 yr		

Health Screening

During the physical exams of the participants, the nurse practitioner performed a thorough inspection of the skin. No findings of chloracne were reported. One participant reported a previous diagnosis of cancer (skin).

Eight of the participants reported taking medication to control their diabetes (25.8%). Commonly, members of the general population may be unaware of diseases they have and, thus, do not report their occurrence. Therefore, to better estimate the actual number of individuals that were diabetic in the Clarinda cohort, fasting blood glucose levels were measured. The American Diabetes Association recommends the following screening levels for a fasting blood glucose test: 70 to 99 mg/dL (3.9 to 5.4 mmol/L) -normal glucose tolerance, 100 to 125 mg/dL (5.5 to 6.9 mmol/L) - impaired fasting glucose (pre-diabetes), 126 mg/dL (7.0 mmol/L) and above - probable diabetes. Of the 31 participants, 21 clients exhibited normal blood glucose, seven exhibited impaired fasting blood glucose (100 to 125 mg/dL), and 3 clients had a blood glucose level \geq 126 mg/dL. All three of the participants in the probable diabetes category reported taking medication to control their condition.

Hypertension (HTN) is defined as a condition in which the blood pressure (BP) is higher than 140 mm Hg systolic or 90 mm Hg diastolic on three separate readings recorded several weeks apart (Venues and Thomas, 1997). Thirteen participants reported taking medication for

hypertension (41.9%). In addition to the self-reported hypertension data that were collected, each participant had their blood pressure checked during the study. Over the course of the study, 9 of the 31 participants (29%) exhibited a high blood pressure reading (>140 mm Hg systolic and/or at >90 mm Hg diastolic). Nationwide prevalence for a person being told that they have high blood pressure was 24.8% in 2003, with 25.8% of whites and 31.4% of African Americans reporting a diagnosis (CDC, 2003a). In Florida, the prevalence of hypertension was 26.5% in 2002, with 27.9% of whites and 33.3% of African Americans reporting a diagnosis of hypertension (CDC, 2003a).

The Clarinda cohort was screened with a hepatitis panel, including hepatitis A antibody, hepatitis B core antibody, and hepatitis C antibody. Nineteen participants (61.3%) were positive for hepatitis A, none were positive for hepatitis B, and only one participant was positive for hepatitis C. According to data collected by the Florida Hepatitis and Liver Failure Prevention and Control Program and the Florida Department of Health, hepatitis A is the most common form of acute viral hepatitis in the United States, and it is one of the 10 most commonly reported infectious diseases in the U.S (Katz, 1999). The CDC reports that 33% of the US population has been infected with hepatitis A (CDC, 2004). The results of the screen indicate that the prevalence of infection for hepatitis A was higher in the Clarinda cohort than observed nationally.

Participants also received a panel of tests to determine liver and kidney status. Clients received a laboratory test for AST (aspartate aminotransferase), ALT (alanine aminotransferase), and GGT (gamma glutamyl transferase). The analytical facility, Labcorp, considers AST and ALT to be elevated at >40 IU/L and GGT to be elevated at >65 IU/L. Elevated AST was observed in 3 participants (9.7%) and elevated ALT was observed in 7 clients (22.6%). Likewise, one participant (3.2%) exhibited elevated GGT. All of the participants exhibited creatinine levels, BUN levels, and BUN/creatinine ratios within normal limits, suggesting that kidney function was not impaired.

The participants were tested for RBCs, HGB, and HCT. The thresholds for low RBC were $<3.8 \times 10^6/\mu\text{l}$ in females and $<4.1 \times 10^6/\mu\text{l}$ in males, for low HGB $<11.5 \text{ g/dL}$ in females and $<12.5 \text{ g/dl}$ in males, and for low HCT $<34\%$ in females and $<36\%$ in males. One female exhibited slightly lowered HGB (11.3 g/dL) and HCT (33.5). The remaining participants were within normal ranges for the three indices.

Participants were weighed and measured to determine body mass index (BMI). According to the CDC, increases in BMI are often accompanied by increased risks for several diseases related to obesity. These include premature death, cardiovascular disease, high blood pressure, osteoarthritis, some cancers, and diabetes. BMI does not indicate that a disease is present, rather it is a predictive factor for disease risk. Normal BMI ranges from 18.5 to 24.9, BMI between 25.0 and 29.9 signifies that a person is overweight, and a BMI ≥ 30.0 indicates obesity. Three participants in the study (11.1%) had a BMI within the normal range, seven (25.9%) were classified as overweight, and 17 (63%) were classified as obese. BMI could not be calculated for four of the participants. Nationally, 34% of adults are classified as overweight and 30% are classified as obese (CDC, 2003b). In Escambia County in 2002, 32.6% of the population was considered overweight and 25% was classified as obese (FDOH, 2003).

The CDC reported that in 2002, 23.0% of adults nationwide smoked cigarettes. In the same year, 22.0% of adult Floridians reported cigarette use: 23.4% of Caucasian adults, 17.5% of African American adults, 23.5% of men, and 20.7% of women were smokers (CDC, 2003a). Active tobacco use (cigars, cigarettes, or smokeless tobacco) in the Clarinda cohort was reported by four participants (12.9%). The remaining 27 clients (87.1%) were not using tobacco at the

time of the screening: 17 clients (54.8%) had never used tobacco and 10 clients (32.3%) used tobacco at one time, but had quit.

Toxicological Profile

Total arsenic levels fell below the method detection limit in 16 of the participants. Mean total arsenic in those participants with detectable levels was 19.8 µg/l (range 12-36 µg/l). The reference interval for total arsenic is 0-50 µg/l. No inorganic arsenic was detected in any of the participants.

Summary data for TEQs for the Clarinda cohort are presented in Table 2. Mean TCDD/F TEQs for the Clarinda cohort was 22.7 pg/g using the 1998 WHO TEF values and 21.8 using the revised 2005 WHO TEF values. Table 3 presents comparison TCDD/F TEQ data from three previous studies of persons exposed to contaminants from wood-treatment facilities (Dahlgren et al., 2003; Dahlgren et al., 2004; Karouna-Renier et al., 2007) and comparison data from four control populations from across the United States, which were not exposed to any known point sources of PCDD/F (Orloff et al., 2001; Dahlgren et al., 2003a; Dahlgren et al., 2004; Schechter and Tung, unpublished data). Table 4 presents comparison data for PCB TEQs calculated using the 1998 WHO TEF values.

Table 2. Preliminary summary of TEQs calculated for the Clarinda cohort. Means, ranges, and percentiles are presented for TEQs calculated using WHO TEFs from 1998 and from 2005.

	Mean	Range	50th	75th	90th	95th
TEQD 1998	22.7	5.2 - 68.2	17.3	30.6	40.78	53.8
TEQD 2005	21.8	5 - 63.8	17	30	38.98	50.66
TEQP 1998	16.2	1.9 - 40.3	11.9	28.9	36.42	39.22
TEQP 2005	7.4	0.9 - 23	5.9	10.3	15.9	20.06
TEQT 1998	38.9	7.3 - 100.8	35	53.9	70.9	88.74
TEQT 2005	29.2	6.9 - 79.7	23.3	39	52.18	67.94

	Karouna-Renier (2007)		Orloff et al. (2001) Comparison		Schechter & Tung Gen. Population pooled (n=100)		Dahlgren (2003)			Dahlgren (2004)			Controls (n=200)
	mean	range	mean	95 th	mean	95 th	Gen. Popul.	Wood Trtmt. Residents	Wood Trtmt. Residents	Wood Trtmt. Residents	95 th		
TEQD	45.2	8.4 - 711.1	121.3	21	37.5	15.8	15.1	25.6	12.7 - 33.3	21.7	2.8 - 78.1	74.7	19.6

Table 3. Comparison data for TCDD/F TEQs calculated using 1998 WHO TEFs.

Table 4. Comparison data for PCB TEQs.

Congener	Schechter & Tung Gen. Population pooled (n=100)	Dahlgren (2004) Control mean	Marchand (2004) hospital patients mean	Dahlgren et al. (2004) Wood Treatment Residents		
	mean	mean	mean	range	95 th perc.	
TEQP 1998	9.3	12	20.8	10.0	1.2 - 31.4	26.4

Evaluation

Our goal was to perform biomonitoring for 40 individuals in the Clarinda Triangle area. Recruitment of participants for this study was somewhat difficult largely because of concerns on whether the results would in any way affect the EPA's proposals/plans to relocate individuals residing in this area. In view of this, our ability to thus far recruit 31 participants (77.5% of our goal) can be considered a very good accomplishment. We will continue our efforts to recruit additional individuals during the remainder of the project period.

The results were shared individually to the participants. The finding of what appeared to be relatively low levels of the analyzed contaminants in most cases alleviated concerns of residual contaminants resulting from past exposures to ETC releases.

The results are being statistically analyzed and will be prepared for publication in a professional journal. The final report will be posted on the CEDB's website for public access. The above outcomes are pertinent to Goal 8-12, "minimize the risks to human health and the environment posed by hazardous sites," for "Healthy People 2010."

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