Fish Reproductive Health Assessment in PCB Contaminated Regions of the Housatonic River, Massachusetts, USA: Investigations of Causal Linkages Between PCBs and Fish Health

Final Report of Phase II Studies
Interagency Agreement 1448-50181-99-H-008
8 July 2003

Prepared for:

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Suggested Citation:

Tillitt, D.E., D.M. Papoulias, and D.R. Buckler. 2003. Fish reproductive health assessment in PCB contaminated regions of the Housatonic River, Massachusetts, USA: Investigations of causal linkages between PCBs and fish health. Final Report of Phase II Studies, Interagency Agreement 1448-50181-99-H-008 U.S. Geological Survey, Columbia Environmental Research Center, 4200 New Haven Rd., Columbia, MO. Biochemistry & Physiology Branch Report, FY03-30-10, 219 p.

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1.0 EXECUTIVE SUMMARY

The Housatonic River is an important natural resource in western Massachusetts and western Connecticut. There is concern that contamination by polychlorinated biphenyls (PCBs) in the Housatonic River has resulted in adverse health effects on the area's fish and wildlife. This document reports the second portion (Phase II) of a set of studies designed to evaluate PCBs in the Housatonic River. Studies conducted in Phase I (Tillitt et al. 2003) were designed to: 1) evaluate the survival, growth and development of offspring of fish collected in the Housatonic River; and 2) quantify the amount of PCBs in Housatonic River fish. Studies conducted in Phase II were designed to provide a more complete test of the hypothesis that PCBs are causally linked to the adverse effects observed in largemouth bass (*Micropterus salmoides*) from the Housatonic River. In Phase II studies, contaminants present in largemouth bass from the Housatonic were extracted and introduced into clean embryos of largemouth bass, medaka (*Oryzias latipes*), and rainbow trout (*Oncorhynchus mykiss*). Concentrations for adverse effects were established, based upon the chemicals present in the extracts.

Adult largemouth bass were collected from three locations along the Housatonic River (Deep Reach, GE River Mile 126-127; Woods Pond, GE River Mile 124-125; and Rising Pond, GE River Mile 105-106) and a reference site (Three-Mile Pond, east of the Housatonic River at GE River Mile 94). The fish were transported to the Columbia Environmental Research Center, U.S. Geological Survey, Columbia, MO. A sub-sample of the fish was used for spawning and production of offspring for the Phase I studies. At the conclusion of the reproductive portion of the Phase I study, the fish were sacrificed to obtain chemical extracts for subsequent injection into clean eggs in Phase II studies.

The adult largemouth bass from the Housatonic River sites contained elevated tissue concentrations of PCBs. Across all three sites, the mean concentrations of total PCBs were 43-149 µg/g (wet wt.) in carcass homogenate samples and 160-448 µg/g in ovary samples. The mean concentrations of total PCBs in whole fish homogenates and ovaries from the reference site were 0.1 µg/g and 0.6 µg/g, respectively. Polychlorinated dibenzofurans (PCDFs), contaminants of commercial PCB mixtures, were also elevated (approximately 90-fold in the tissues and 150-fold in the ovaries based on dioxin-like toxic potency, TEQs) relative to the reference fish. Polychlorinated dibenzo-p-dioxins (PCDDs) were elevated (approximately 5 to 10 fold in the tissues and 15 to 20 fold in the ovaries based on TEQs) relative to the concentrations in fish from

the reference site. The mean total dioxin-like potency in largemouth bass from the Housatonic River was 37-65 pg TEQs/g in the tissues and 109-135 pg TEQs/g in ovaries, while the fish from the reference site contained 1.0 pg TEQs/g in either whole carcass or ovary samples.

Findings from Phase I Studies -- The adult largemouth bass collected from the Housatonic River had several symptoms that were consistent with PCB-related toxicosis, including elevated liver enzyme (ethoxyresorufin-O-deethylase, EROD) activities, abnormal gonadal histology, and elevated occurrences and rates of macrophage aggregates. In general, organ weights and steroid hormone concentrations were not different in adult largemouth bass from the Housatonic River than in fish from the reference site, Three-Mile Pond. The offspring of largemouth bass collected from the Housatonic River showed a suite of symptoms that was consistent with PCB-related toxicity, including delayed development, reduced survival at swim-up, cytochrome P450 1A induction, developmental deformities, and reduced weight gain. Some deformities, such as lack of inflation of the swim bladder and craniofacial deformities that were observed exclusively in the fry from the Housatonic River, suggest an Ah-receptor agonist etiology. These same symptoms were not observed in the offspring of largemouth bass from the reference location, Three-Mile Pond. This suite of indicators, the specific patterns of their occurrence, and the magnitudes of their responses at this critical life-stage in fishes were indicative of an adverse effect related to PCBs or other dioxin-like compounds. Exposure indicators and adverse effects observed in largemouth bass and their offspring from all study locations on the Housatonic River are consistent with PCB-related toxicity in these populations. Additionally, the data from these experiments indicate that all of the study sites on the Housatonic River (Deep Reach, Woods Pond, and Rising Pond) were above threshold concentrations for adverse effects in largemouth bass.

Findings from Phase II Studies – Parallel and similarly-shaped dose-response curves for mortality and effects were observed for extracts and reference toxicants independent of species exposed, however, LD50s and ED50s for extracts were lower than those for PCB 126 and 2,3,7,8-TCDD. The swim-up stages of medaka and rainbow trout were generally more sensitive than largemouth bass to the toxic effects of the extracts and the reference standards. Survival in swim-up largemouth bass in Phase II studies was generally higher than survival in Phase I studies. Differences in site-specific mortality were not notable in swim-up largemouth bass from

Phase II studies, while in Phase I studies Deep Reach offspring showed poorer survival. However, mean mortality for Woods Pond offspring of females which averaged 128 pg TEQ/g ovarian tissue (Phase I results) falls on the predicted dose-response curve from the in ovo injection studies in Phase II and estimated mean effects exhibited in the Phase I Woods Pond offspring are close to the corresponding predicted dose-response curve from Phase II.

Effects from exposure to the reference toxicants, PCB126 and 2,3,7,8-TCDD that were common to all species we tested included craniofacial deformities, edema, and hemorrhaging. These compounds also caused largemouth bass to have problems inflating the swim bladder, and to have deformed fins. Largemouth bass exposed to PCB 126 were also observed to have the swim bladder external to the body. Fish injected with Housatonic River extracts generally showed adverse effects that were consistent with those exposed to reference toxicants and the offspring from the Phase I studies. These effects included: craniofacial, eye, fin, and opercular deformities, edema and problems associated with swim bladder inflation. External swim bladder was observed in largemouth bass exposed to Deep Reach and Woods Pond extracts. Three-Mile Pond-exposed fish had fewer types and incidence of pathologies than those exposed to the Housatonic River extracts.

Threshold concentrations for adverse effects (mortality plus pathology) in largemouth bass were established for Woods Pond extract and PCB 126. The geometric mean of the NOEL and LOEL for swim-up fry after in ovo exposure to Woods Pond extract was estimated to be 83 pg TEQ/g egg. No NOEL was observed for PCB 126, however the LOEL was 75 pg TEQ/g egg.

INTRODUCTION

The Housatonic River is a valuable aquatic resource, both aesthetically and economically (Orciari and Leonard 1990). Draining over 2000 square miles, the Housatonic flows south through a series of impoundments in western Massachusetts and western Connecticut, terminating in Long Island Sound. During the past two decades there has been increasing concern regarding the threat posed by the presence of highly toxic environmental contaminants to the fish and wildlife inhabiting the river. The principal cause of this contamination is a polychlorinated biphenyl (PCB) point source located on the East Branch of the Housatonic River at Pittsfield, Massachusetts. Total PCB concentrations in fish downstream of this source have been reported at levels as high as 418 mg/Kg (Rick Beach, personal communication). Although PCBs are considered to be the major toxic input, polychlorinated dibenzo-*p*-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs) also contribute to contamination of the river (Eitzer 1993).

PCBs, PCDDs, and PCDFs are collectively referred to as planar halogenated hydrocarbons (PHHs). The toxic effects of PHHs and structurally similar compounds are thought to be mediated through contaminant binding to a cytosolic aryl hydrocarbon receptor (Ah-R). This ligand-activated transcription factor binds with selected sites on DNA called dioxin responsive elements which control the expression of genes that encode both Phase I and Phase II enzymes. The induction of these proteins is part of a response to dioxin-like chemicals, which leads to alterations in cellular homeostasis (DeVito and Birnbaum 1995). Binding of PHHs to the Ah-R has been linked to several molecular events including the production of electrophilic metabolites and oxygen radicals, reduced capacity for xenobiotic metabolism, and alteration in the rates of endogenous substrate metabolism (Stegeman and Hahn 1994). In fish, early life stages appear to be particularly sensitive to the effects of Ah-R ligands (Mehrle et al. 1988; Walker and Peterson 1991), and evidence indicates the specific involvement of cytochrome P450 enzymes in this embryo-toxic response (Cantrell et al. 1996; 1998).

A detailed assessment of the toxicological impact of PHHs on fish from the Housatonic River has not previously been conducted. Using a combination of validated field and laboratory-based

tools, the current project evaluates the potential for contaminants in the Housatonic River to elicit embryo-toxic effects in fish that could ultimately result in altered population size and structure.

3.0 OBJECTIVES AND EXPERIMENTAL DESIGN

The studies conducted within this project were designed to test two fundamental hypotheses. The first set of studies (Phase I) addressed the sensitivity of fish to Housatonic River contaminants. The second set of studies (Phase II) was directed toward determination of the mechanism of toxicity of Housatonic River contaminants. Specifically, the hypotheses tested in the two Phases were as follows: Phase I) early life stages of fish species in the Housatonic River are sensitive to the amount and composition of the PHHs present, and Phase II) the PHHs present in fish from the Housatonic River elicit detrimental effects through an Ah-R mediated mechanism of toxicity.

3.1 Study Objectives

To test the first hypothesis, studies in Phase I were designed to evaluate the survival and development of offspring of fish collected from three PCB-contaminated locations in the Housatonic River and from an off-river reference site in the basin. Phase I studies had the following primary objectives:

1) Determine appropriate rearing conditions for embryos of representative species from the Housatonic River.
2) Determine the dioxin-like effects present in embryos and early life stages of offspring from fish collected from four areas of interest in the Housatonic River Basin.
3) Determine the concentration of PCBs and other organic contaminants present in the carcass and ovaries of fish collected from the four areas of interest.

To test the second hypothesis, studies in Phase II were designed to determine the mechanisms of embryo-toxic effects of contaminants found in fish from the Housatonic River Basin sites. Phase II studies had the following primary objectives:

1) Develop extracts of organic contaminants from largemouth bass (*Micropterus salmoides*) collected from each of the four areas of interest for use in egg injection studies, and characterize the PCBs and other hydrophobic organic chemicals present in those extracts.
2) Determine the embryo-toxic effects of the extracts in egg injection studies with largemouth bass, medaka (*Oryzias latipes*), and rainbow trout (*Oncorhynchus mykiss*).
3) Determine the extent to which an additive model of dioxin-like toxicity explains the effects observed in embryos and fry in the Phase I rearing studies and in the Phase II egg injection studies.

3.2 Experimental Design

Phase I of the project involved the collection of brood fish from four study sites in the Housatonic River Basin, spawning of the fish in experimental ponds, and subsequent rearing of offspring in the laboratory. Stage-specific mortality determinations, gross pathology assessments, histological examinations, and biochemical measurements were made on the developing embryos and resultant fry. Information on survival, development, and growth was augmented with exposure assessment. The carcasses of brood fish and portions of ovaries collected during this phase of the project were analyzed for organochlorine chemicals, including pesticides, PCBs, dioxins, and furans. Additionally, samples were obtained from adult fish for assessment of biological indicators of contaminant exposure. Biochemical and physiological measurements in the adult largemouth bass included ethoxyresorufin-O-deethylase induction (EROD, a measure of cytochrome P450 induction); gross histology of liver and gonads; measurement of plasma concentrations of estrogen and testosterone and their ratios (E/T ratios); and measurement of plasma concentrations of vitellogenin. This exposure and effects information has been previously reported in our Interim Report of Phase I Studies (Tillitt et al. 2003).

During Phase II studies, chemical mixtures extracted from Housatonic River largemouth bass were used to develop dose-response relationships for embryo toxicity in the laboratory, exclusive of other environmental stressors. The studies were designed to allow controlled laboratory exposures of fish eggs and developing embryos to extracts obtained from bass from the four locations of interest in the Housatonic River Basin. Egg injection techniques and subsequent monitoring of the developing embryos and fry were used to assess toxic effects. The egg injection procedures effectively mimic the maternal transfer of hydrophobic contaminants, such as PCBs, to developing oocytes. Moreover, the toxic effects observed in previous egg injection studies have been shown to be similar to those observed in studies where eggs obtained contaminants through maternal deposition (Walker et al. 1994; 1996). The results of the Phase I field-laboratory studies, when evaluated in the context of the predictive dose-response models developed in the Phase II laboratory studies, help determine the extent to which the early life-stage toxicity observed in Housatonic River largemouth bass may be attributed to PCBs.

4.0 METHODS

4.1 Collection of Largemouth Bass from Housatonic River

Adult largemouth bass were collected from three sites on the Housatonic River and from one off-river reference site in the basin during the first week of May 1999. The timing of sampling was intended to correspond with the onset of spawning condition. U.S. Fish and Wildlife Service personnel collected the fish, using their electro-shocking collection protocols. The three Housatonic River sampling sites included Rising Pond (GE River Mile 105-106), Woods Pond (GE River Mile 124-125), and Deep Reach (GE River Mile 126-127). The off-river reference site was Three-Mile Pond, which is located east of the Housatonic River near GE River Mile 94. More specific locations of these sites are shown in Figure 1. The fish were held in cages at the collection sites until May 12, 1999, when they were transported to CERC in Columbia, MO. The fish from the four collection sites were transported by truck in separate aerated compartments, each containing approximately 1135 L of water. The fish were treated with 3-5 parts per thousand salt to reduce handling stress during the 20-h trip to CERC (Piper et al. 1982). Upon arrival at CERC on May 13, 1999, the largemouth bass were immediately stocked into

experimental ponds for the Phase I studies. After completion of spawning, the brood fish were sacrificed on June 3, 1999 for subsequent extract preparation.

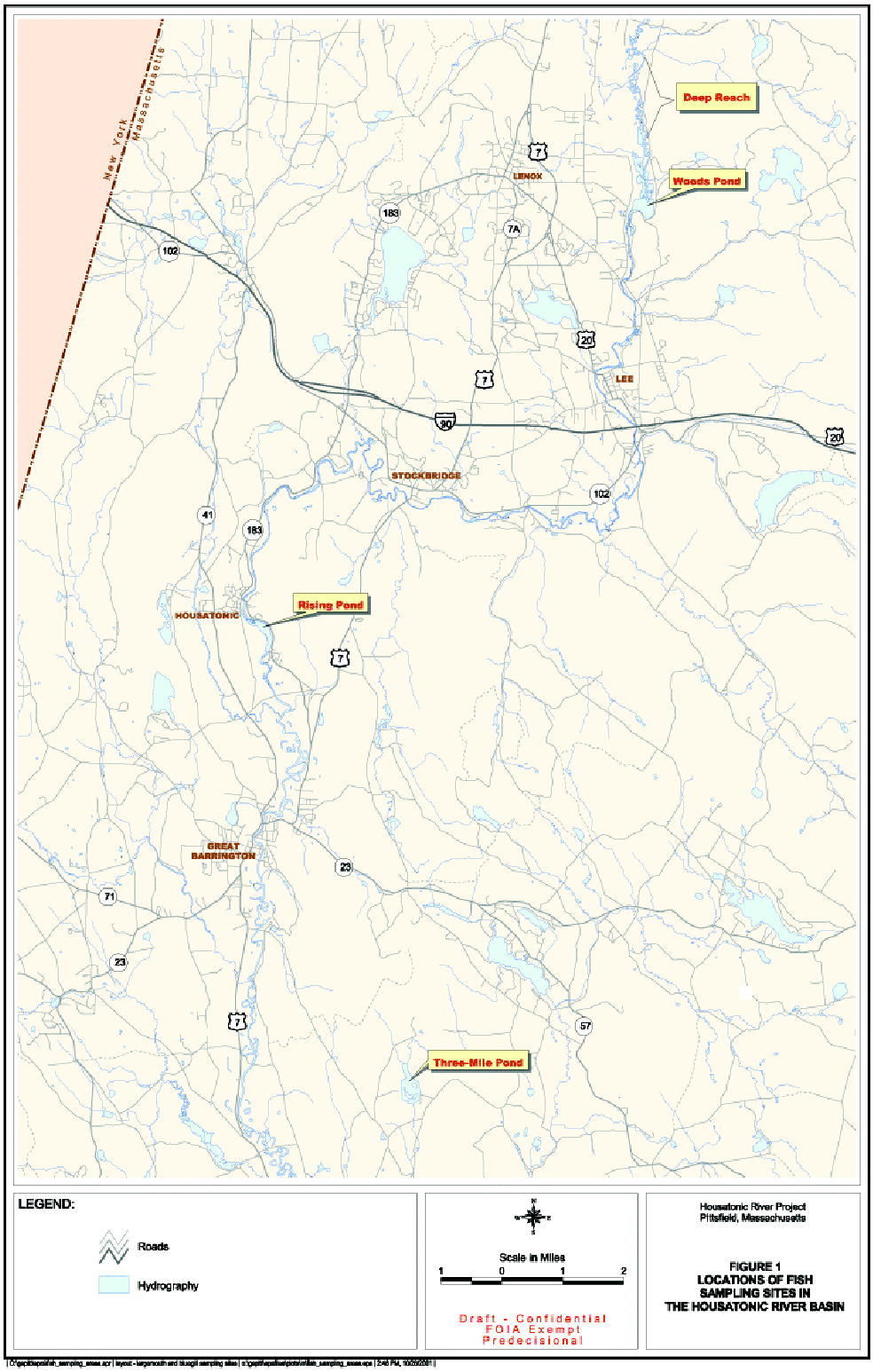

Figure 1. Locations of fish sampling sites in the Housatonic River Basin.

4.2 Processing of Largemouth Bass to Obtain Contaminant Extracts

The largemouth bass from the Housatonic River were sacrificed at the end of the reproduction portion of the Phase I studies to obtain tissues from which the dosing solutions were prepared for use in the Phase II egg injection studies. For each of the four study sites, a 15-18 kg mass of largemouth bass tissue (multiple individuals of both sexes) was collected. A detailed description of the methods used for extract preparation can be found in Appendix 1, but the procedures were generally as follows. Composites of the carcasses of fish were created for each site, after removal of the ovaries from females (testes were not removed from males). The tissue samples were dehydrated by addition of anhydrous sodium sulfate and no method recovery compounds were added. The four large composite samples (one for each sampling site) were column extracted with methylene chloride and the solvent was removed from the lipid by rotoevaporation. Contaminants were isolated from the lipids using large-scale dialysis through a polyethylene membrane. Dialytic recoveries, as checked by ^{14}C-labeled 2,5,2',5'-tetrachlorobiphenyl and mirex, averaged 77%. Dialysates were composited and a small portion of the extract (~0.1%) was used to determine percent lipid (CERC SOP P.461). Any remaining co-dialyzed lipids were removed from the extracts by reactive column cleanup on acid- and base-treated silica gels and adsorbent chromatography on activated silica gel (CERC SOP P.186). These extracts were further purified by high performance gel permeation chromatography (HPGPC) (Meadows et al. 1993; Meadows et al. 1996; CERC SOP P.464). Dosing solutions were prepared containing graded doses of the isolated contaminants using triolein as the diluent and injection vehicle (CERC SOP P.461). This procedure resulted in purified dosing solutions of PCBs, PCDDs, and PCDFs from the original tissue samples.

4.3 Chemical Analysis of Largemouth Bass Carcasses and Extracts

There were two types of samples analyzed in the Phase II portion of this study. Portions of each fish carcass homogenate were taken for routine chemical analysis and portions of the final extract used for the dosing solutions were also analyzed for confirmation of the doses used for injection (Figure 2). Recovery standards were added to the analytical samples either in the sub-

sampled fish homogenates or into the sub-samples of the purified extract dialysate. Triplicate portions of the final purified dialysates from HPGPC (prepared as described above) were removed for analysis using the procedures outlined in Figure 2. Samples were diluted using estimates of total PCBs (UV detection) in order to prevent overloading of the porous graphitic carbon (PGC) column. These diluted samples were fractionated on PGC (CERC SOP P.480) into the following fractions: PGC 1 (*ortho*-chlorinated or congener-specific PCB congeners and organochlorine pesticides), PGC 2 (non-*ortho*-chlorinated PCBs), and PGC 3 (polychlorinated dibenzo-*p*-dioxins and furans). The PGC 1 fraction was subsequently analyzed by GC/electron capture detection (GC/ECD). The PGC 2 and PGC 3 fractions were analyzed by GC/high resolution mass spectrometry (GC/HRMS). The PGC 3 fraction was further purified on basic alumina before the GC/HRMS analysis (CERC SOP P.193). The analyses also incorporated a procedural blank and a procedural spike as quality control samples. In order to monitor method recoveries through the HPLC carbon and alumina cleanup steps, the final purified dialysates and the quality control samples had the following method recovery compounds added to them: PCB 029 (2,4,6-trichlorobiphenyl), PCB 155 (2,2',4,4',6,6'-hexachlorobiphenyl), PCB 204 (2,2',3,4,4',5,6,6'-octachlorobiphenyl), four ^{13}C-labeled non-*ortho* PCB congeners, and seventeen ^{13}C-labeled 2,3,7,8-substituted dioxins and furans. Procedural spikes in the PHH portion of the analysis also received PCBs (mixed Aroclors 1242, 1248, 1254, and 1260) and native (^{12}C) dioxin and furan congeners.

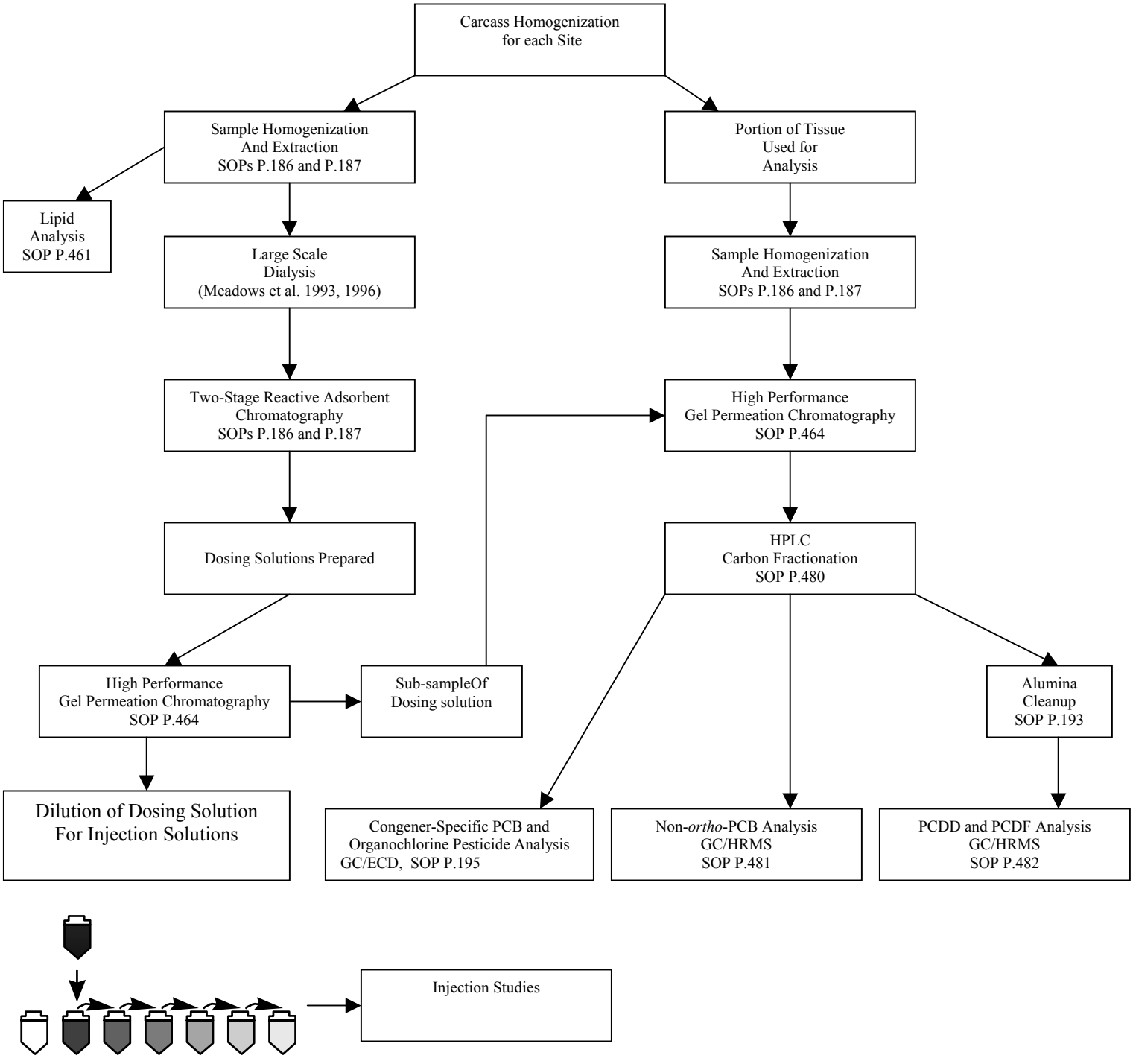

Figure 2. Preparation of dosing solutions and analytical scheme for confirmation of PCB congeners, organochlorine pesticides, polychlorinated dibenzo-*p*-dioxins, and polychlorinated dibenzofurans concentrations in dosing solutions.

Gas chromatographic method for congener-specific or ortho-chlorinated PCB congeners – The PGC 1 fractions containing PCB *ortho*-chlorinated congeners (ie. congener-specific PCB analysis) were adjusted to final volumes of 10 mL. Two instrumental internal standards (400 ng each of PCB congener 030 and 207) were added at this point to these fractions. Individual PCB congeners were subsequently measured by GC/EDC. Analyses were performed using Hewlett-Packard 5890 Series II GCs with cool on-column capillary injection systems and Hewlett-Packard 7673 autosamplers (CERC SOP P.195). For all analyses, a 3-m section of 0.53 mm i.d. uncoated and deactivated capillary retention gap guard column (J&W Scientific/Agilent, Wilmington, DE) was attached to the front of each analytical column by a Press-Tight® connector (Resteck Corporation, Bellefonte, PA). The analytical columns included a 60-m x 0.25-mm DB-5 column (0.25 μm 5% phenyl-, 95% methylsilicone) and a 60-m x 0.25-mm DB-17 column (0.25 μm 50% phenyl-, 50% methylsilicone); both from J&W Scientific/Agilent, Folsom, CA. The H_2 carrier gas was pressure regulated at 25 psi. The temperature program for the PCB analysis was as follows: initial temperature 60° C, immediately ramped to 150° C at 15° C/min, then ramped to 250° C at 1° C/min, and finally ramped to 320° C at 10° C/min and held for 1 min. The electron capture detector temperature was 330° C.

Capillary GC/ECD data were collected, archived in digital form, and processed using a Perkin-Elmer chromatography data system, which included the Model 970 interface and Version 6.1 of Turbochrom Workstation™ chromatography software (Perkin-Elmer Corporation, Norwalk, CT), on a Pentium III microcomputer (CERC SOP P.483). Six levels of a PCB standard (designated A1111, which is a combination of Aroclors 1242, 1248, 1254, and 1260 in a 1:1:1:1 (w:w:w:w) ratio) were used for PCB congener calibration, with total PCB concentrations ranging from 50 to 8000 ng/mL. The elution (on two capillary columns) and concentrations of individual PCB congeners in the A1111 calibration standard have been verified against certified individual PCB standards (Accustandard, New Haven, CT) using previously reported methods (Frame 1997; Krupcík et al. 1992; Krupcík et al. 1993).

The method detection limits (MDLs) and method quantitation limits (MQLs) for individual PCB *ortho*-chlorinated congeners and for total PCBs were based on procedural blank results according to the method outlined by Keith et al. (1983) and Keith (1991).

Gas chromatographic method for organochlorine pesticides – Organochlorine pesticides were analyzed with the *ortho*-chlorinated PCBs in the PGC1 fraction by GC/ECD (CERC SOP P.459). The instrumental components were the same as for the PCB congener analysis discussed above. The analytical column used was the DB-17 column, described in detail above. Capillary GC/ECD data were collected, archived in digital form, and processed using a Perkin-Elmer chromatography data system, which included the Model 970- interface and Version 6.1 of Turbochrom Workstation™ chromatography software, on a Pentium III microcomputer (CERC SOP P.483). Six levels of OC pesticide standards were used for calibration, with each pesticide at concentrations ranging from 0.5 to 80 ng/mL. Detection limits were calculated as discussed above for PCB congeners.

GC/HRMS method for non-ortho-chorinated PCB congeners – The non-*ortho*-chlorinated PCB fractions (PGC 2) were spiked with 5 ng of instrumental internal standard (^{13}C-labeled 2,2',4,5,5'-pentachlorobiphenyl (PCB #101). At a final volume of 50 µL, the non-*ortho*-PCBs were determined by GC/HRMS, monitoring two sequential mass windows during the chromatographic separation (CERC SOP P.481; Peterman et al. 1996). GC/HRMS analysis was performed with a Hewlett-Packard 5890A capillary gas chromatograph interfaced to a VG 70-250AS high-resolution mass spectrometer. A Hewlett-Packard 7673 autosampler was used to introduce 2 µL of the extract onto a 2.5 m x 320 µm deactivated fused silica retention gap via heated (285 °C) direct on-column injection with a Restek spiral Uniliner. The analytical column was a 50 m x 200 µm x 0.11 µm Ultra-1 capillary column. The GC oven was held at 120° C for 1 min, programmed to 240° C at 2.2° C/min, then ramped to 310° C at 5° C/min, and a final hold of 5 min. Helium carrier gas was maintained at 45 psi with an initial linear velocity of 27 cm/sec.

The VG GC/HRMS system was tuned to a resolution of 10,000 and calibrated using perfluorodecalin. Mass windows were established for two groups of non-*ortho*-PCBs. Within

each mass window, the two most abundant ions were measured for positive identification and quantitation of each analyte. The ion responses were quantified and averaged. Also, additional ions monitored the responses of higher chlorinated, potentially interfering PCB congeners, Cl_{4-8} naphthalenes (PCNs), Cl_{3-5} terphenyls (PCTs), Br_5- and Cl_6-diphenyl ethers (residual carryover from PGC 1), and Cl_4-PCDF (to ensure no breakthrough of PCDFs).

A calibration curve describing the response of each native congener (0.25 to 2,500 pg/µL) to that of its ^{13}C-labeled surrogate was used. Quantification is inherently corrected by the ^{13}C-isotopically labeled surrogates, which account for analytical losses during isolation procedures and variations in the instrumental analysis.

Molecular ion responses of certain PCB congeners were measured to ensure that their fragment ion responses did not contribute an interference >10% to the responses of the respective non-*ortho*-PCBs. Column performance was verified by analyzing standards of individual congeners, labeled congeners, and congeners from Aroclor mixtures. Because non-*ortho*-Cl_5-PCB 126 is only minimally resolved from Cl_6-PCB 129, PCB 129's molecular ion response was monitored to assure that its fragment ion response (3.5% abundance) did not contribute an interference of >10% to the response of PCB 126. Adequate mass resolution was verified by monitoring Cl_{4-8} ions of PCNs.

Criteria for Confirmation: For the positive identification and quantitation of each congener, the following criteria were established and met in this study:

1. Peak areas for the selected ion responses must be greater than three times background noise.
2. Native ion peaks must occur at retention times from -1 to +3 sec of that for the corresponding ^{13}C-labeled ion peaks, which generally elute about 1 sec earlier.
3. The ion ratio for the two principal ion responses must be within the acceptable range (±15%).

GC/HRMS method for polychlorinated dibenzo-p-dioxins and furans -- The PCDD/PCDF fractions (PGC3) were eluted through basic alumina to remove potential co-contaminants such as chlorinated diphenyl ethers (PCDEs) and residual PCNs and PCBs (CERC SOP P.480) prior to

chromatographic analysis. A total of 1 ng of the internal standard, ^{13}C-labeled 1,2,3,4-TCDD (tetrachlorodibenzo-p-dioxin), was added and the volume reduced to ~25 µL. The GC/HRMS analysis was performed using a Hewlett-Packard 5890A capillary gas chromatograph interfaced to a VG 70-AS high-resolution mass spectrometer. Five sequential selected ion windows were monitored. (CERC SOP P.482). A Hewlett-Packard 7673 autosampler injected 2µL of the extract into the GC/HRMS. A spiral uniliner, 2.5m x 320µm deactivated fused silica retention gap, and heated (285° C) direct inlet were employed. The analysis was conducted with a 50m x 200µm x 0.11µm Ultra-2 (Agilent Technologies, Wilmington, DE) capillary column, with an initial hold of 1 min at 120° C followed by a ramp to 200° C at 20° C/min, another ramp to 300° C at 2.3° C/min, and a final hold of 5 min. The helium carrier gas was maintained at 44 psi with an initial linear velocity of 25 cm/sec.

The VG GC/HRMS system was tuned to a resolution of 10,000 and calibrated using perfluorokerosene. Mass windows were established for five ion groups to measure Cl_{4-8} PCDFs and PCDDs. Within each mass window, the two most abundant ions were measured for positive identification and quantitation of each analyte. The ion responses were quantified and averaged. Additional ions monitored any responses from potentially interfering Cl_{5-9}-PCDEs and Cl_{5-7}-polychlorinated terphenyls (PCTs), and dioxin-like Cl_{6-7}-PCNs, Cl_{3-8} dibenzothiophenes (PCDTs), and Cl_{3-8} phenanthrene and anthracenes. A calibration curve describing the response of each native congener to that of a ^{13}C-labeled surrogate congener was used for quantification.

Column performance was verified by analyzing standards of individual components and observing the chromatographic resolution of the TCDDs, HxCDDs (hexachlorodibenzodioxins), and HxCDFs (hexachlorodibenzofurans). Similarly, relative retention times for all other congeners of interest were evaluated with respect to labeled analogs. Adequate mass resolution was verified by monitoring Cl_{6-7} PCNs ions versus ion responses of ^{13}C-TCDDs and of native TCDD versus ^{13}C-TCDF. Lock-mass and lock-mass-check ions were used to maintain and verify the accuracy of mass measurement.

Criteria for Confirmation: For the positive identification and quantitation of a particular congener, the following criteria were established and met in this study:

1. Peak areas for the selected ion responses must be greater than three times background noise.
2. For congeners with isotopically-labeled analogs, the ion peaks for the native must occur at retention times from -1 to +3 sec of that for the corresponding ^{13}C-labeled ion peaks, which generally elute about 1 sec earlier.
3. For OCDF (without an isotopically-labeled analog), ion responses in sample analyses must occur at RRTs from -0.2 to 0.5% of ^{13}C-labeled OCDD, analogous to the window above.
4. The ion ratio for the two principal ion responses must be within the acceptable range (±15%).

4.4 Procurement of Eggs of Largemouth Bass, Medaka, and Rainbow Trout

Largemouth Bass – Brood stock were obtained on two separate occasions and placed in experimental ponds for spawning at CERC. Personnel of the Missouri Department of Conservation made the first collection of fish. On April 27, 2000, 16 male and 18 female largemouth bass in spawning condition were captured from Little Dixie Lake Conservation Area, 1821 State Road RA, Fulton, MO, by electro-shocking. The fish were hauled directly to CERC (approximately 10 miles) and stocked into experimental ponds. Spawning began on April 28, 2000 and ended on May 5, 2000. On May 5, 2000 the fish were removed from the ponds. For additional availability of eggs, a second set of brood stock was obtained from the Genoa National Fish Hatchery, U.S. Fish and Wildlife Service, S5689 State Road 35, Genoa, WI, on May 8, 2000. Upon arrival at CERC (after approximately 12 hours in transit), the fish were held in raceways at CERC until May 15, 2000 when they were stocked in experimental ponds for spawning. Spawning commenced on May 17, 2000 and was completed by May 23, 2000, when they were removed from the ponds. Carcasses of a subset of all brood fish from Genoa National Fish Hatchery and those collected from Little Dixie Lake were frozen and portions of fish from each source were sent to the Patuxent Analytical Control Facility, U.S. Fish and Wildlife Service, Laurel, Maryland, for chemical residue analysis.

Spawning procedures for the fish were essentially as described in the report of Phase I studies (Tillitt et al. 2003). Briefly, the fish were stocked into two 0.1-hectare ponds for spawning. Each pond was provided with six spawning substrates (0.5 x 0.6 m mats constructed from latex-

coated coconut fiber, placed at depths from 0.3 to 1.0 m (Figure 3). Each nesting site was marked with a float and spawning behavior was monitored twice daily. Once spawns were confirmed on the mats, they were retrieved and brought into the laboratory. In the laboratory, mats were placed in 1.5% sodium sulfite solution for 5-7 min to minimize egg adhesiveness, allowing for egg removal, separation, and subsequent injection.

Medaka – Broodstock of the dr-R strain were maintained in well water under an 18:6 light-dark photoperiod at 25-27^{o}C. The fish were fed three times per day with brine shrimp (*Artemia salina*; 48 h old) and finely ground flake food that was high in xanthathin (Wardley Spectra IV, The Wardley Corp., Secaucus, NJ). Eggs were collected during morning hours on the day before exposures began. The eggs were carefully selected so that only healthy, fertilized eggs were used.

Rainbow trout - Eggs of the Fish Lake strain of rainbow trout were obtained from the Ennis National Fish Hatchery, U.S. Fish and Wildlife Service, Ennis, Montana, between January 31 and March 21, 2001. During this period, weekly shipments of green unfertilized eggs (3 to 6 females kept separate) and milt (3 sets of 2 males pooled) were received. The shipments included approximately 3000 eggs from each female. The eggs were sent Fed-Ex overnight packed in an egg-shipping crate with ice in trays above and below those trays containing eggs and milt. Eggs arrived between 9:30 and 10:00 AM, the crate was immediately opened and the temperature in the egg bags was checked. An incubator in the laboratory was set to the measured temperature and allowed to acclimate during which time the egg crate was repacked to prevent rapid temperature change in the eggs and milt. All eggs and milt were quickly transferred to the incubator and then brought up to 12^{o} C at a rate of 1^{o} C per hour. Eggs were fertilized using the dry method in the late afternoon and placed in trout Heath trays maintained at 12 to 12.5^{o} C and allowed to water-harden overnight.

Figure 3. Spawning mats used in experimental ponds during Housatonic River Basin largemouth bass Phase II studies.

4.5 Embryo Injection

Embryos were exposed following a slightly modified nano-injection procedure described by Walker et al. (1996). Immediately prior to injection, embryos were placed in an agarose matrix to provide the necessary stability for the injection process. A sterile saline solution was added to cover the eggs. Aluminosilicate capillary tubes (1.0 mm OD and 0.53 mm ID; Sutter Instrument Co., Novato, CA) were pulled on a pipette puller (Narishige, Boyce Scientific, MO) to make needles with a 5-10 μ OD tip when beveled at a 15^{o} angle using a diamond wheel pipette grinder (Narishige, Boyce Scientific, MO). Cleaned and siliconized (Sigmacote; Sigma Chemical Co., St. Louis, MO) needles were filled using a pipettor with femtotips (Eppendorf, Madison, WI). Triolein (95% purity, Sigma Chemical Co., St. Louis, MO) was used as the carrier solvent and was filter-sterilized using a 5-cc syringe with disposable 0.22-μ filter (Corning Glass Inc., Corning, NY) before use. Injections of material at approximately 0.1% of egg volume (0.5nL per egg for largemouth bass and medaka, 40 nL per egg for rainbow trout) were made directly into the egg yolk using a Narashigi picoinjector (PLI-188, Nikon, Inc., Melville, NY) and micro-manipulator (MM-3, Stoelting Co., Wood Dale, IL).

The experimental design for these experiments included the development of dose-response curves for the organic extracts of Housatonic River fish with largemouth bass, medaka, and rainbow trout. The chemicals used as standards for comparison to the Housatonic extracts included 2,3,7,8-TCDD and PCB 126 (3,3',4,4',5-pentachlorobiphenyl). TCDD was chosen as a standard because it is the hallmark chemical for Ah-receptor (Ah-R) related responses. PCB 126 was chosen as the other standard because it often accounts for the majority of the dioxin-like potency of PCB mixtures in the environment and has the greatest toxic equivalency factor (TEF) of all of the 209 PCB congeners in fish (van den Berg et al. 1998).

To develop dose-response relationships, five doses of each Housatonic River fish extract (and five to six doses of the chemical standards) were injected (see Figure 4 and Tables 1-3). A set of uninjected eggs and triolein-injected eggs was also included with each dosing experiment. Multiple trials were conducted for each exposure series. The actual amounts injected were

originally calculated to bracket the concentrations observed in fish from the study sites. The doses are expressed in units of gram-equivalents (g-EQ). One g-EQ equals the amount of extract that corresponds to one gram of Housatonic River fish, normalized to lipid content (Tillitt and Wright 1997). The doses were planned to span the range of chemicals observed in the field-collected samples (Tillitt et al. 2003).

The exposure concentrations of the extracts injected into the eggs were evaluated using the toxic equivalency approach. This approach assumes an additive model of toxicity for the dioxin-like congeners (see van den Berg et al. 1998 for further details on this approach). From this approach, the contribution of the planar PCBs (non-*ortho*-chlorinated congeners) to the overall dioxin-like toxicity may be estimated. The toxic equivalency factors (TEFs) used were those developed from fish embryo mortality (van den Berg et al. 1998). The dioxin toxic equivalents (TEQs) estimated for each dose of the extracts allows calibration of the dose-response relationship for the extracts against the chemical standards, TCDD and PCB 126. If the prevailing mode of toxic action of the extract is through an Ah-R mediated pathway, then it would be expected that the slope of the dose-response curves for the extracts and standards would be parallel. Additionally, comparison of the median values for mortality (LD50) between the extracts and standards also allows evaluation of the appropriateness of an additive model of toxicity for the Housatonic River fish extracts. If the slopes of the extract dose-response curves are parallel to those of the standards and the toxicity is additive, then the contribution of PCBs to the overall dioxin-like toxicity can be estimated from these studies.

Figure 4. Injection of largemouth bass egg.

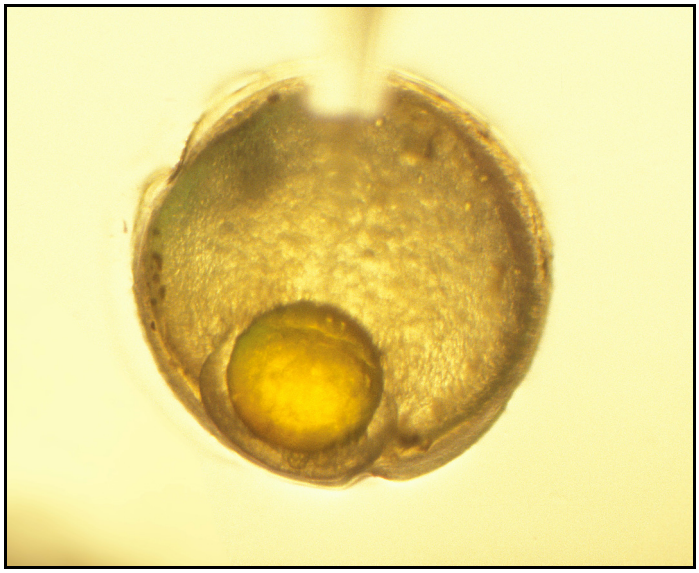

Table 1. Concentrations (by component) in doses delivered to eggs of largemouth bass, expressed on a per gram of egg basis. Injection volumes for largemouth bass were 0.5 nL of solution per egg.

		Dose					
	units/g egg	**1**	**2**	**3**	**4**	**5**	**6**
Three-Mile Pond Extract	**g-eq tissue**	0.03	0.1	0.5	2.5	5.0	---
	ug tPCBs	0.001	0.002	0.02	0.08	0.15	---
	pg TEQs	0.030	0.06	0.6	3.0	6.0	---
Rising Pond Extract	**g-eq tissue**	0.03	0.1	0.5	2.5	5.0	---
	ug tPCBs	0.5	1.0	9.0	45	90	---
	pg TEQs	1.0	2.0	16	81	161	---
Woods Pond Extract	**g-eq tissue**	0.03	0.1	0.5	2.5	5.0	---
	ug tPCBs	1.0	2.0	19	93	185	---
	pg TEQs	0.6	1.0	12	59	118	---
Deep Reach Extract	**g-eq tissue**	0.03	0.1	0.5	2.5	5.0	---
	ug tPCBs	2.0	3.0	31	155	310	---
	pg TEQs	0.6	1.0	13	64	128	---
PCB 126	**ng PCB 126**	15	44	133	400	1200	---
	pg TEQs	75	220	665	2000	6000	---
2,3,7,8-TCDD	**pg TCDD**	70	220	670	2000	6000	---
	pg TEQs	70	220	670	2000	6000	---

--- Only five doses of extracts and positive controls were used with largemouth bass.

Table 2. Concentrations (by component) in doses delivered to eggs of medaka, expressed on a per gram of egg basis. Injection volumes for medaka were 0.5 nL of solution per egg.

		Dose					
	units/g egg	**1**	**2**	**3**	**4**	**5**	**6**
Three-Mile Pond Extract	**g-eq tissue**	0.03	0.1	0.5	2.5	5.0	---
	ug tPCBs	0.001	0.002	0.02	0.08	0.15	---
	pg TEQs	0.030	0.06	0.6	3.0	6.0	---
Rising Pond Extract	**g-eq tissue**	0.03	0.1	0.5	2.5	5.0	---
	ug tPCBs	0.5	1.0	9.0	45	90	---
	pg TEQs	1.0	2.0	16	81	161	---
Woods Pond Extract	**g-eq tissue**	0.03	0.1	0.5	2.5	5.0	---
	ug tPCBs	1.0	2.0	19	93	185	---
	pg TEQs	0.6	1.0	12	59	118	---
Deep Reach Extract	**g-eq tissue**	0.03	0.1	0.5	2.5	5.0	---
	ug tPCBs	2.0	3.0	31	155	310	---
	pg TEQs	0.6	1.0	13	64	128	---
PCB 126	**ng PCB 126**	5.0	15	44	133	400	1200
	pg TEQs	25	75	220	665	2000	6000
2,3,7,8-TCDD	**pg TCDD**	20	70	220	670	2000	6000
	pg TEQs	20	70	220	670	2000	6000

--- Only five doses of extracts were used with medaka.

Table 3. Concentrations (by component) in doses delivered to eggs of rainbow trout, expressed on a per gram of egg basis. Injection volumes for rainbow trout were 40 nL of solution per egg.

		Dose					
	units/ g egg	**1**	**2**	**3**	**4**	**5**	**6**
Three-Mile Pond Extract	**g-eq tissue**	0.02	0.04	0.45	2.2	4.5	---
	ug tPCBs	0.0007	0.001	0.01	0.07	0.13	---
	pg TEQs	0.03	0.05	0.5	3.0	5.0	---
Rising Pond Extract	**g-eq tissue**	0.02	0.04	0.45	2.2	4.5	---
	ug tPCBs	0.4	0.8	8.0	40	81	---
	pg TEQs	0.7	1.0	14	72	145	---
Woods Pond Extract	**g-eq tissue**	0.02	0.04	0.45	1.1	2.2	---
	ug tPCBs	0.8	1.7	17	42	83	---
	pg TEQs	0.5	1.0	11	27	53	---
Deep Reach Extract	**g-eq tissue**	0.02	0.04	0.45	1.1	2.2	---
	ug tPCBs	1.4	2.8	28	70	139	---
	pg TEQs	0.6	1.0	11	29	57	---
PCB 126	**ng PCB 126**	1.0	4.0	13	40	120	360
	pg TEQs	5.0	20	65	200	600	1800
2,3,7,8-TCDD	**pg TCDD**	11	22	67	200	600	1800
	pg TEQs	11	22	67	200	600	1800

--- Only five doses of extracts were used with rainbow trout.

4.6 Embryo Rearing, Fry Culture, and Sampling Procedures

Embryo Incubation and Larval Grow Out:

Samples of eggs from each spawning event were distributed into three replicates of 10 eggs each for each treatment in studies with largemouth bass and medaka, and three replicates of 24 eggs each for each treatment in studies with rainbow trout. Growth, survival, and pathological evaluations were made for each replicate. There was also a separate set of injections (at or near the expected LD50 of each of the mixtures) along with a set of triolein-injected eggs that were incubated and sampled temporally for histopathological and biochemical evaluations. The developing bass embryos were incubated generally as described by Piper et al. (1982).

Largemouth bass **-** The hatching and fry rearing system for largemouth bass consisted of rearing baskets suspended in a constant-temperature (27^{o} C), aerated, flow-through water bath. Eggs were hatched and fry were raised in the rearing baskets placed in stainless steel raceways (137.2 X 35.6 X 30.5 cm) that were supplied with a constant flow of well-water (300 ml/min). The rearing baskets were constructed from 100 mL polyethylene specimen cups with three 2 x 3 cm openings symmetrically cut around the sidewalls of the container. The openings were covered with stainless steel screen (30 X 30 mesh, 0.010" diameter wire) to retain embryos, yet allow flow of water into and out of the containers. Air stones were placed in the surrounding water bath to enhance the movement of water through the rearing baskets. Baskets were placed in the water column to attain an approximate volume of 60 mL for the 10 eggs contained in each basket. Eggs and hatched embryos were maintained in the rearing baskets until the fry reached the 15d post swim-up life stage. The rearing baskets were cleaned daily and dead eggs were removed with a pipette. After swim-up and the onset of exogenous feeding, the fish were fed 24-hr-old brine shrimp (*Artemia salina)* by hand pipetting a known concentration of nauplii into each chamber. The culture of the artemia followed standard practices (CERC SOP P.525). The nauplii were fed to the largemouth bass fry at 3-hour intervals from 6:00 am to 9:00 pm.

Medaka **–** Medaka embryos were reared in petri dishes in a 27^{o} C incubator until approximately 24 h before hatch (~8 days). They were then moved to 150-ml glass side-arm test tubes (Corning

Glass Inc., Corning, NY) suspended in a 25-27 ^{o}C water bath. A gentle stream of air was used to roll the embryos in the water column. Upon hatch, medaka larvae were moved to rearing baskets as described above for largemouth bass, where they were maintained for 15d. Fry were fed three times per day with brine shrimp (*Artemia salina*; 24-h old) and finely ground flake food high in xanthathin (Wardley Spectra IV, The Wardley Corp., Secaucus, NJ). Photoperiod was 16 hours light: 8 dark and lighting intensity was approximately 100-200 lux. Dead fry, feces, and excess food were removed by pipette prior to each feeding.

Rainbow trout **-** Rainbow trout eggs were injected according to SOP P.535. After injection, the eggs for each replicate were placed in labeled baskets and returned to Heath trays. The eggs were checked daily, Monday through Friday, and any white eggs (dead) removed. All removed eggs were cleared with a 15% acetic acid solution and identified as fertile or unfertile. The number of eggs removed and their status was recorded. This information was used to adjust mortality results so that only fertilized eggs were included. Sampling times were expressed as Daily Temperature Units (DTU). Study completion was at 600 DTU, approximately the time of swim-up.

4.7 Pathological, Histological, and Biochemical Evaluations

Mortality was monitored daily for all species. Deformities in the developing embryos and fry were monitored throughout development. Standard symptoms of an Ah-R mediated mode of action (Peterson et al. 1993) including measures of gross physical abnormalities and pathology (e.g. edema, hemorrhage, and craniofacial anomalies) were noted and recorded. Embryos and fry were observed under a dissecting microscope and photographs were taken to document examples of the deformities observed. Gross pathological observations were made on largemouth bass at hatch, swim-up, and 15d post swim-up. Observations were made on medaka at hatch, approximately 100h post hatch (about the time of yolk absorption), and at 15d post hatch. Rainbow trout were observed at hatch and at study completion (600 DTU). At the end of the studies, largemouth bass and medaka were weighed and measured for length. All survivors from all studies were preserved in 10% neutral-buffered formalin.

Preparation of fry for histological analysis was performed in general accordance with CERC SOP P.549. Formalin-preserved fry were washed in 10 mM HEPES (pH 7.4) and dehydrated by immersion in graded aqueous solutions of ethanol ranging from 50% to 100% ethanol. This was followed by immersion in xylene and subsequent infiltration with paraffin. The paraffin-embedded samples were sectioned at 5 μm and placed onto silanized slides (CERC SOPs P.548 and P.550), then stored at room temperature until staining.

In preparation for histochemical staining, tissue sections were dewaxed with xylene and then rehydrated to water by immersion in graded solutions containing decreasing concentrations of ethanol ranging from 100% to 0% (CERC SOP P.540). Tissue samples were then stained following the general nuclear stain procedure for hematoxylin and eosin (CERC SOP P.539) or Masson Trichrome (Moore 1996).

The level of cytochrome P450 induction observed following contaminant exposure is often used to infer the presence of compounds such as PCBs that can exert effects through the aryl hydrocarbon receptor (Ah-R). Immuno-detection of cytochrome P450 in paraffin-embedded tissue sections was accomplished using an indirect peroxidase-labeling method (CERC SOPs P.551 and P.547). The tissue sections were dewaxed and rehydrated in a PBS solution containing 1% bovine serum albumin. The rehydrated slides were incubated in 0.5% H_2O_2 in methanol for 5 min to block endogenous peroxidase activity. The tissue sections were then immunochemically stained using monoclonal antibody (mAb) 1-12-3 made against scup P450E as the primary antibody (Park et al. 1986), and observed for peroxidase staining (red-brown deposit) using light microscopy. Companion sections were incubated with a non-specific monoclonal immunoglobulin G2 (Smolowitz et al. 1991) to confirm the specificity of the observed responses. Selected sections were counter stained with Mayer's hematoxylin to enable differentiation of cellular structures. Qualitative assessments were then made as to the extent and intensity of the responses in various tissue types.

4.8 Statistical Analysis

Prior to analyses, data were tested using the Kolmogorov test for normality and Levene's test for homogeneity of variances. Only those dose-response trials for which control (triolein and uninjected) survival was $\geq$ 50% (largemouth bass) or $\geq$ 70% (medaka and rainbow trout) were analyzed statistically for effects on survival, weight, or length. The selection of minimum survival criteria was derived from the American Society for Testing and Materials (ASTM) Standard Guide E 1241-98 for conducting early life stage toxicity tests with fishes (ASTM 2002). The ASTM Standard Guide recommends the use of $\geq$ 70% control survival as a test acceptability criterion for studies conducted with rainbow trout. Different acceptability criteria are provided for other commonly tested species. However, due to their comparatively infrequent use as laboratory test organisms, ASTM recommendations are not provided for medaka and largemouth bass. The rainbow trout acceptance criterion was adopted for medaka, since this species is relatively easily cultured in the laboratory. An acceptance criterion of $\geq$ 50% survival was selected for largemouth bass due to the species' lack of standard culturing techniques and its r-selected reproductive strategy (MacArthur and Wilson 1967). ANOVA or alternatively, Kruskal-Wallis as the non-parametric test, were used for all evaluations. To evaluate treatment effects, mean comparisons were made with triolein-injected controls. Mulitiple mean comparison procedures used were either Least Square Means or Bonfferroni. Differences were evaluated at the 0.05 alpha level. All analyses were performed using SAS software v.8 (SAS Institute 2000).

The incidence of gross pathologies observed for a given chemical/extract during development was first summarized on an occurrence per 1000 individuals basis by summing all observations of a pathology at a given developmental stage across all trials, dividing the sum by the number of fish examined, and multiplying the quotient by 1000. These data are presented for both specific doses of a chemical or extract, and also for all doses of a chemical or extract combined. Because many of these trials did not meet the test acceptability criteria, only a subset of these data were used in subsequent statistical evaluations.

For each species, a matrix is presented that identifies those pathologies that were common to both PCB 126-exposed fish and to fish exposed to the Housatonic River fish extracts and/or 2,3,7,8-TCDD. Comparisons were made at equivalent TEQ levels across chemicals and extracts. To be included in the matrix, a given pathology had to be present in exposed fish at a level that was greater than twice that observed in the negative controls (uninjected and triolein).

A subset of the swim up pathology data was evaluated for the effect of chemical or extract exposure on the percentage of fish developing one or more pathologies. Significant differences between controls and dosed individuals were tested using Chi-square analysis. Only those trials with $\geq$ 50% survival (largemouth bass) or $\geq$ 70% survival (medaka and rainbow trout) in negative controls (uninjected and triolein) were used in these analyses, using the same rationale for acceptance criteria outlined above. Multiple trials meeting these criteria were pooled for a chemical or extract.

Dose-response curves were developed and LD50 and ED50 concentrations with 95% CI and slopes were estimated for largemouth bass and medaka at swim up (yolk nearly completely absorbed), and for rainbow trout at 600 DTU (yolk absorption and the end of the trout study). An additional LD50 was also calculated for medaka at the conclusion of the study (15 d post swim up) an ED50 was not calculated because there were almost no pathologies observed at this stage. No ED50s were calculated for bass at the end of the study because so few survived the transition from endogenous to exogenous feeding. Only trials for which negative control (uninjected and triolein) survival was $\geq$ 50% (largemouth bass) or $\geq$ 70% (medaka and rainbow trout) and effects (mortality or mortality plus pathology) were < 50% in negative controls and $\geq$ 50% at the highest dose were used. For ED50 estimates, individuals were considered affected when they were either dead, or showed one or more pathologies. The rationale for the control survival acceptance criteria has been previously explained. A similar rationale was used for the < 50% acceptance criterion for combined effects in the negative controls. The criterion of $\geq$ 50% occurrence of combined effects at the highest dose, was established to avoid extrapolation beyond the range of the observed data, thereby ensuring acceptable confidence intervals for the ED50. Only trials meeting the acceptance criteria were evaluated for fit to a dose-response curve. In a few cases where acceptance criteria were met, one or more treatments exhibited

extensive mortality due to fungal infection. These trials are identified in the survival tables and were not further evaluated.

Graphs depict predicted dose-response curves while data points are the original data from experiments. All data were tested for normality and homogeneity of variances prior to analysis. Comparisons were made to pooled controls if the were not significantly different, or to triolein controls if they were. Probit analysis with Abbott's correction was used for data meeting the assumptions for parametric analysis, while Spearman-Karber was used for data that failed to meet those assumptions. TOXSTAT v3.5 was used for these analyses (West 1996). Slopes were calculated by conducting a regression analysis on the linear portion of the dose-response curves.

5.0 RESULTS

5.1 Chemical Analysis of Largemouth Bass Carcass Extracts

Congener-specific PCBs – Congener-specific PCB analysis was conducted on the final extract dosing solutions made from fish carcasses collected from the Housatonic River and reference site (Tables 4 and 5). The results were normalized to the initial tissue mass of the carcasses (Table 4) and concentrations were normalized to the volume of the final extract and top dosing solution (Table 5). A total of 139 individual congeners were reported. The sum of the concentrations of the congeners was used to quantify total concentrations of PCBs in these same samples. Details of the analytical results are presented in an analytical report (CERC Organic Chemistry Section, Project Report # 3307-70L1E) that are attached in Appendix 1 and summarized below.

The concentrations of congener-specific PCBs (ng/g wet weight) in the dosing solutions made from the carcasses of largemouth bass from the Housatonic River were elevated (600-2100 fold greater) at all locations relative to the dosing solutions made from largemouth bass carcasses from the reference site, Three-Mile Pond (Tables 4-5). Concentrations (mean and standard deviation) of total PCBs in the dosing solutions made from carcasses of the largemouth bass were 62,000 1,200 ng/g at Deep Reach; 37,000 0 ng/g at Woods Pond; 18,000 580 ng/g at Rising Pond; and 30 0.02 ng/g at Three-Mile Pond (Table 4). These are significantly lower

concentrations of total PCBs than were present in the original largemouth bass carcasses in the Phase I report of this study (Tillitt et al. 2003). In the Phase I report the concentrations of total PCBs were found to be 149,000 7,900 ng/g at Deep Reach; 108,000 17,600 ng/g at Woods Pond; 43,000 2,500 ng/g at Rising Pond; and 106 0.3 ng/g at Three-Mile Pond (Tillitt et al. 2003, Table 24). The lower amounts of total PCBs in the dosing solutions relative to the amounts that were in the original fish carcass's was due to reduced process recoveries in the large-scale extraction and clean-up of the extracts. The overall recoveries of total PCBs for the fish processing, based on the concentrations in the dosing solutions relative to the amounts in the fish carcasses, were 42% for the Deep Reach and Rising Pond dosing solution, 34% for the Woods Pond-derived dosing solution, and 28% for the dosing solution made from the 3-Mile Pond fish carcasses. These are lower than previously obtained from similar large-scale extraction procedures in which recoveries were on the order of 70-80% of the original material (Meadows et al 1996; Wright and Tillitt 1999). However, the losses of the PCBs during the extraction and dialytic procedures were not biased, and the resultant composition of the PCBs in the dosing solutions had the same relative composition of individual PCB congeners as the original largemouth bass carcasses. This was confirmed by a comparison of the PCB congener patterns measured in the dosing solution and in the fish carcasses using principle components analysis (PCA, data not shown) in accordance with the descriptions provided in the PhaseI report (Tillitt et al. 2003). So, even though the concentrations of the PCB congeners was significantly lower in the dosing solutions than would have been expected, the composition of those congeners did not change relative to the PCB congener patterns found in the largemouth bass taken from the Housatonic River or reference area (Three-Mile Pond). Thus, the relative potency estimates derived from these injection studies are good predictions of the toxicity of the mixtures, when the concentrations in the carcasses are corrected for losses or the concentrations in the dosing solutions are used for the exposure estimates.

The QC check samples for this set of samples and analyses all had results that were within the data quality objectives for these measurements (See Appendix 1, CERC Organic Chemistry Section Project Report # 3307-70L1E for details).

Organochlorine Pesticides **-** The concentrations of organochlorine (OC) pesticides were measured in the final extract dosing solutions made from fish carcasses collected from the Housatonic River and reference site (Tables 6 and 7). The pesticides measured were hexachlorobenzene (HCB), pentachloroanisole (PCA), alpha-hexachlorocyclohexane (alpha-BHC), beta-hexachlorocyclohexane (beta-BHC), delta-hexachlorocyclohexane (delta-BHC), heptachlor, heptachlor epoxide, dacthal, dieldrin, endrin, oxychlordane, cis-chlordane, trans-chlordane, cis-nonachlor, trans-nonachlor, o,p=-DDE, o,p=-DDD, o,p=-DDT, p.p=-DDE, p,p=-DDD, p,p=-DDT, endosulfan I, endosulfan II, endosulfan sulfate, methoxychlor, and mirex (Phase I Report, Table 21). Details of the analytical results are presented in an analytical report (CERC Organic Chemistry Section, Project Report # 3307-70L1E) that are attached in Appendix 1 and summarized below.

The concentrations of organochlorine pesticides in the dosing solutions made from largemouth bass taken from the Housatonic River were at or near detection limits for all chemicals except p,p'-DDE (31-42 ng/g, Table 6; or 45-55 ng/uL, Table 7). The greatest doses of p,p'-DDE delivered to the fish eggs resulted from injections of the Deep Reach extract at dose 5 (92 ng/g in rainbow trout or 210 ng/g in medaka and largemouth bass). These values of p,p'-DDE are below the threshold for embryo toxicity of 1270 ng/g for rainbow trout (Hopkins et al. 1969). Thus, OC pesticides present in the extracts of the largemouth bass from the Housatonic River or the reference site (Three-Mile Pond) are not thought to have a direct toxic effect on the survival or development on the embryos tested in this series of experiments.

Non-ortho-PCB congeners - Concentrations of non-*ortho*-substituted PCB congeners (PCB 81, PCB 77, PCB 126, and PCB 169) were measured in dosing solutions made from largemouth bass carcasses taken from the study area and reference site (Tables 8-9). The concentrations were reported as amounts normalized to the mass of the fish tissue used to make each extract (Table 8, pg/g of carcass) or the mass of non-*ortho*-PCB congeners were normalized to the volume of the final extract (Table 9, pg/uL of extract). These PCB congeners act on developmental and reproductive processes through the Ah-receptor in fish. The dioxin-like toxicity imposed by these PCB congeners was summarized with an additive model of toxicity with the TEF/TEQ approach (van den Berg et al. 1998) in a subsequent section of this report. Details of the analytical results are presented in an analytical report (CERC Organic Chemistry Section, Project Report # 3307-70L1E) which is attached in Appendix 1 and summarized below.

The concentrations of non-*ortho*-PCBs in the dosing solutions made from carcass homogenates of largemouth bass from all locations along the Housatonic River were elevated (50-2000 fold greater) relative to concentrations observed in dosing solutions prepared from carcass homogenates of largemouth bass collected at the reference site, Three-Mile Pond (Table 8-9). Concentrations of all of the non-*ortho*-PCB congeners were small in the dosing solutions made from largemouth bass from Three-Mile Pond (Table 8-9). The dosing solutions made from fish collected at Rising Pond, the furthest downstream site on the Housatonic River, contained 37 5.8 pg/g; 810 72 pg/g; 1,500 58 pg/g; and 210 5.9 pg/g of PCB 81, PCB 77, PCB 126, and PCB 169, respectively. The concentrations of non-ortho PCBs in largemouth bass carcass homogenates collected at the two upstream sites on the Housatonic River, Woods Pond and Deep Reach, were similar to one another. The concentrations in the dosing solutions made from carcasses of largemouth bass collected from Woods Pond were 83 24 pg/g; 1,700 120 pg/g; 2,400 58 pg/g; and 300 26 pg/g of PCB 81, PCB 77, PCB 126, and PCB 169, respectively. Concentrations of these same PCB congeners in the dosing solutions made from carcasses of largemouth bass collected from the Deep Reach section of the Housatonic River were 194 33 pg/g; 2,000 450 pg/g; 2,500 450 pg/g; and 360 44 pg/g of PCB 81, PCB 77, PCB 126, and PCB 169, respectively. These concentrations in the dosing solutions are given in units of pg per gram equivalent of fish tissue in the original extract (Table 8), but have also been given in the form of a concentration in the dosing solutions (Table 9).

The QC check samples associated with this batch were within acceptable limits of the data quality objectives for most of the samples. Details of the quality control for this set of measurements are presented in an analytical report (CERC Organic Chemistry Section, Project Report # 3307-70L1E) which is attached in Appendix 1.

Polychlorinated dibenzo-p-dioxins and polychlorinated dibenzofurans - Concentrations of 2,3,7,8-substituted polychlorinated dibenzo-p-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs) were measured in the dosing solutions made from the carcasses of largemouth bass from the study area and reference site (Tabes 10-11). These PCDD/F congeners act on developmental and reproductive processes through the Ah-receptor in fish. The dioxin-like toxicity imposed by these PCDD/F congeners was summarized with an additive model of toxicity using the TEF/TEQ approach (van den Berg et al. 1998) and is discussed in a later section of this report. Details of the analytical results are presented in an analytical report (CERC Organic Chemistry Section, Project Report # 3307-70L1E) that attached in Appendix 1 and summarized below.

The concentrations of PCDD/F congeners in the dosing solutions made from carcasses of largemouth bass collected from the Housatonic River sites were elevated relative to the reference site, Three-Mile Pond (Table 10-11). The PCDD/F congeners were at or near the limits of detection (or quantitation) in the dosing solutions made from extracts of carcasses of fish collected from the reference site, Three-Mile Pond. Only 2,3,7,8-TCDF in the Three-Mile Pond dosing solution extracts was consistently measured at concentrations above the procedural blanks. The mean total TEQ concentrations that were contributed by PCDD/F congeners in the dosing solutions made from fish collected at the reference site, Three-Mile Pond, was 1.2 pg TEQ/g of tissue extracted (Table 10). PCDD/F congeners in the dosing solutions made from largemouth bass collected from the Housatonic River were found at similar concentrations among all of the collection sites (Table 10). The concentrations of PCDFs in the dosing solution made from Housatonic River fish carcasses were greater than the concentrations of PCDDs, as would be expected with the major source of chemicals being PCBs. 2,3,4,7,8-pentachlorodibenzofuran (2,3,4,7,8-PCDF) was the PCDD/F found to be in the greatest amount (over and above concentrations in the procedural blanks) in the dosing solutions made from

largemouth bass carcasses from the Housatonic River (Table 10). The 2,3,7,8-tetrachlorodibenzofuran (2,3,7,8-TCDF) congener was also elevated in the all of the Housatonic River-derived dosing solutions. 2,3,7,8-tetrachlorodibenzo-p-dioxin (2,3,7,8-TCDD) and 1,2,3,7,8-pentachlorodibenzo-p-dioxin (1,2,3,7,8-PCDD) were the only PCDD congeners that were elevated above background or procedural blank samples in the dosing solutions made from carcasses of largemouth bass collected in the Housatonic River.

The QC check samples were all within expected range, with a few exceptions (see CERC Organic Chemistry Section, Project Report # 3307-70L1E, Appendix 1 fro details).

Dioxin Toxic Equivalents (TEQs) - The toxicity of chlorinated dioxins, furans, and planar PCBs is expressed through the aryl hydrocarbon receptor (Ah-R). The untoward effects of PCDDs, PCDFs, and planar PCBs in fish and wildlife largely occur through an additive model of toxicity (van den Berg et al. 1998; Tillitt 1999). Therefore, we have taken the exposure data (concentrations of doxins, furans, and planar PCBs) measured in the dosing solutions and converted the chemical concentrations into dioxin-like potency, using dioxin toxic equivalence factors (TEFs) and an additive model of toxicity. The TEFs used for this section and report were those TEFs designated for ecological risk assessment of PCBs, PCDDs, and PCDFs in fish (van den Berg et al. 1998). We used the fish-specific TEF values from the World Health Organization report of the international workshop on the TEF/TEQ approach. The analytes that were found to have concentrations at or below the limits of detection or quantitation were assigned the limit of detection or quantitation, respectively, for purposes of TEQ calculation in the samples.

Dioxin toxic equivalents (TEQs) in the dosing solutions made from largemouth bass carcasses used in the fish reproduction studies were derived from measured concentrations of planar (non-ortho) PCBs, 2,3,7,8-substituted PCDDs, and 2,3,7,8-substituted PCDFs in the samples (Table 12). The fish-based TEFs from van den Berg et al. (1998) were used to develop potency values (TEQs) for the extracted material in the dosing solutions. The concentrations were reported as amounts normalized to the mass of the fish tissue used to make each extract (pg/g of carcass) or the mass of PCDD/PCDF congeners were normalized to the volume of the final extract (pg/uL of extract). The concentrations of TEQs in the dosing solution extracts developed from largemouth

bass from Three Mile Pond were small (Table 12). Mean TEQs were 1.2 0.1 pg/g (1.8 0.2 pg/uL) in the dosing solution derived from carcass homogenates of largemouth bass from the reference site, Three Mile Pond (Table 12). Contributions to the total TEQs in the dosing solution from the reference site, Three Mile Pond, were mainly from chlorinated dioxins (~70%), with most of the remainder of the TEQs derived from PCDFs (29%). The contribution of PCBs was only 1% of the total TEQs in this dosing solution from Three Mile Pond. The exact proportions of the relative contributions of the different classes of compounds to the total TEQs are not considered to be very accurate for these samples due to the small amounts present in the Three Mile Pond extracts and samples. Most of the PCDDs and PCDFs were below detection in these samples. Thus, the estimated total TEQs in the dosing solution made from carcasses of largemouth bass from Three Mile Pond reported here, were likely to be an over-estimate, biased by the use of detection limits in the calculations. The dioxin-like potencies in the dosing solution extract of largemouth bass from Three Mile Pond are considered small and at or near background.

The concentrations of TEQs in the dosing solutions made from largemouth bass carcasses from the Housatonic River were elevated at all three of the collection sites. The mean concentrations of TEQs in dosing solutions derived from Housatonic River largemouth bass carcasses were 32 0.3, 23.6 0.6, and 25.5 3.6 pg TEQs/g of tissue extracted from Rising Pond, Woods Pond, and Deep Reach locations on the river, respectively (Table 12). The TEQs in these same dosing solutions normalized to the final volume of the extracts were 41.2 0.4, 33.5 0.8, and 33.2 4.6 pg/uL of extract (Table 12). The contributions of PCB-related TEQs and PCDF-related TEQs were the greatest at all three sites and combined to account for approximately 85-92% of the measured TEQs. The large majority of the PCB-related TEQs from all three studies areas on the Housatonic River were due to the contribution of PCB 126. The pentachlorinated dibenzofuran, 2,3,4,7,8-PCDF, contributed the greatest (65-73%) to the dioxin and furan-related TEQs in the dosing solutions made from largemouth bass carcasses from the Housatonic River study sites. These proportions were similar to the relative contributions of these classes of compounds (PCBs, PCDDs, and PCDFs) compared to the relative contributions measured in the carcasses in the Phase I studies (Tillitt et al. 2003).

5.2 Chemical Analysis of Ovaries of Largemouth Bass Broodstock

The quality of the eggs from both the Little Dixie reservoir and the Genoa National Fish Hatchery was good. In general, organochlorine chemical and metals contaminant levels were below thresholds for early life-stage effects (see tabulated results in Appendix).

Table 4. Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**001**	**003**	**004**	**005**	**006**	**007**	**008**	**009**	**010**	**015**	**016**	**017**	**018**	**019**
Three-Mile Pond, 3MP-A	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.05	< 0.01	< 0.02	< 0.07	< 0.14
Three-Mile Pond, 3MP-B	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.05	< 0.01	< 0.02	< 0.07	< 0.14
Three-Mile Pond, 3MP-C	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.05	< 0.01	< 0.02	< 0.07	< 0.14
Mean										**0.05**				
SD										**0.00**				
Rising Pond, RP-A	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	0.92	1.4	1.3	1.3
Rising Pond, RP-B	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	1.5	1.2	1.5
Rising Pond, RP-C	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	1.4	1.2	1.2
Mean												**1.4**	**1.2**	**1.3**
SD												**0.05**	**0.05**	**0.16**
Woods Pond, WP-A	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	8.2	7.2	7.0
Woods Pond, WP-B	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	7.8	6.4	6.6
Woods Pond, WP-C	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	8.2	6.6	9.9
Mean												**8.1**	**6.7**	**7.8**
SD												**0.20**	**0.37**	**1.8**
Deep Reach DRP-A	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	14	11	11
Deep Reach DRP-B	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.4	< 0.01	14	11	9.3
Deep Reach DRP-C	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	12	8.8	24
Mean												**14**	**10**	**15**
SD												**1.3**	**1.1**	**8.3**

Table 4 (Cont.). Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**020**	**022**	**024**	**025**	**026**	**027**	**028**	**031**	**032**	**033**	**034**	**035**	**037,059**	**040**
Three-Mile Pond, 3MP-A	< 0.01	< 0.04	< 0.01	< 0.01	< 0.01	< 0.04	< 0.04	< 0.02	< 0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02
Three-Mile Pond, 3MP-B	< 0.01	< 0.04	< 0.01	< 0.01	< 0.01	< 0.04	< 0.04	< 0.02	0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02
Three-Mile Pond, 3MP-C	< 0.01	< 0.04	< 0.01	< 0.01	< 0.01	< 0.04	< 0.04	< 0.02	0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02
Mean														
SD														
Rising Pond, RP-A	< 0.01	0.31	< 0.01	1.4	2.3	0.51	4.9	0.66	1.3	< 0.04	< 0.17	< 0.10	0.47	0.42
Rising Pond, RP-B	< 0.01	0.29	< 0.01	1.3	2.4	0.55	4.8	0.77	3.1	< 0.04	< 0.17	< 0.10	0.51	0.49
Rising Pond, RP-C	< 0.01	0.26	< 0.01	1.5	2.5	0.59	4.8	0.82	3.3	< 0.04	< 0.17	< 0.10	0.48	0.43
Mean		**0.29**		**1.4**	**2.4**	**0.55**	**4.9**	**0.75**	**2.6**				**0.49**	**0.45**
SD		**0.03**		**0.08**	**0.06**	**0.04**	**0.07**	**0.08**	**1.1**				**0.02**	**0.03**
Woods Pond, WP-A	< 0.01	1.2	< 0.01	8.9	16	3.1	21	4.4	17	1.7	< 0.17	< 0.10	1.9	1.3
Woods Pond, WP-B	< 0.01	1.4	< 0.01	8.2	16	3.1	20	4.4	17	1.4	< 0.17	0.17	1.2	1.3
Woods Pond, WP-C	< 0.01	1.7	< 0.01	8.7	15	2.9	21	4.6	16	1.7	< 0.17	< 0.10	1.9	1.1
Mean		**1.4**		**8.6**	**16**	**3.1**	**21**	**4.4**	**17**	**1.6**			**1.7**	**1.2**
SD		**0.24**		**0.37**	**0.19**	**0.14**	**0.19**	**0.13**	**0.74**	**0.19**			**0.39**	**0.12**
Deep Reach DRP-A	< 0.01	1.8	< 0.01	6.8	18	5.0	18	7.7	39	0.90	< 0.17	0.90	< 0.02	1.7
Deep Reach DRP-B	< 0.01	2.1	< 0.01	5.9	20	5.5	19	5.9	38	1.7	3.0	< 0.10	2.1	1.6
Deep Reach DRP-C	< 0.01	2.4	< 0.01	7.3	18	5.4	21	7.8	30	0.49	0.49	< 0.10	2.9	2.3
Mean		**2.1**		**6.7**	**18**	**5.3**	**20**	**7.1**	**36**	**1**				**1.8**
SD		**0.32**		**0.7**	**1.2**	**0.3**	**1.1**	**1.0**	**5.2**	**0.6**				**0.38**

Table 4 (Cont.). Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**041**	**042**	**043**	**044**	**045**	**046**	**047**	**048**	**049**	**051**	**052**	**053**	**054**	**055**
Three-Mile Pond, 3MP-A	< 0.02	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.08	< 12	< 0.08	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02
Three-Mile Pond, 3MP-B	< 0.02	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.09	< 12	< 0.08	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02
Three-Mile Pond, 3MP-C	< 0.02	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.08	< 12	< 0.08	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02
Mean							**0.08**							
SD							**0.00**							
Rising Pond, RP-A	< 0.02	11	1.1	12	0.39	0.35	170	51	96	8.4	72	7.9	< 0.02	< 0.02
Rising Pond, RP-B	< 0.02	13	1.4	12	0.49	0.36	170	< 12	93	8.5	70	7.8	< 0.02	< 0.02
Rising Pond, RP-C	0.03	11	2.9	12	0.47	0.33	170	19	95	8.5	72	7.8	< 0.02	< 0.02
Mean		**11**	**1.8**	**12**	**0.45**	**0.35**	**170**		**95**	**8.5**	**71**	**7.9**		
SD		**1.1**	**0.96**	**0.20**	**0.05**	**0.01**	**0.0**		**1.6**	**0.04**	**1.1**	**0.08**		
Woods Pond, WP-A	< 0.02	80	4.0	39	0.32	1.3	420	220	260	33	210	30	0.64	< 0.02
Woods Pond, WP-B	0.47	82	2.8	38	0.94	1.3	430	230	270	32	220	30	0.63	< 0.02
Woods Pond, WP-C	< 0.02	81	3.2	39	0.76	1.1	410	210	260	32	210	30	0.76	< 0.02
Mean		**81**	**3.3**	**38**	**0.67**	**1.2**	**420**	**220**	**260**	**32**	**210**	**30**	**0.68**	
SD		**0.64**	**0.61**	**0.70**	**0.32**	**0.12**	**10**	**10**	**5.8**	**0.59**	**5.8**	**0.26**	**0.07**	
Deep Reach DRP-A	< 0.02	93	5.4	47	1.7	1.7	570	610	310	41	280	37	0.42	< 0.02
Deep Reach DRP-B	< 0.02	92	6.6	51	< 0.12	1.2	560	590	310	43	280	39	1.6	< 0.02
Deep Reach DRP-C	< 0.02	92	6.8	50	< 0.12	1.8	580	690	320	43	280	39	0.45	< 0.02
Mean		**92**	**6.3**	**49**		**1.5**	**570**	**630**	**310**	**42**	**280**	**38**	**0.81**	
SD		**0.3**	**0.75**	**1.8**		**0.33**	**10**	**53**	**6**	**1**	**0**	**1**	**0.65**	

Table 4 (Cont.). Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**056,060**	**057**	**058**	**063**	**064**	**066**	**067**	**069**	**070**	**071**	**072**	**074**	**075**	**082**
Three-Mile Pond, 3MP-A	< 1.7	0.03	< 0.01	0.01	< 0.02	0.08	< 0.06	< 0.01	< 0.20	< 0.23	< 0.01	< 0.13	< 0.01	0.02
Three-Mile Pond, 3MP-B	< 1.7	0.03	< 0.01	0.01	< 0.02	0.08	< 0.06	< 0.01	< 0.20	< 0.23	< 0.01	< 0.13	< 0.01	0.02
Three-Mile Pond, 3MP-C	< 1.7	0.03	< 0.01	0.01	< 0.02	0.08	< 0.06	< 0.01	< 0.20	< 0.23	< 0.01	< 0.13	< 0.01	0.02
Mean		**0.03**		**0.01**		**0.08**								**0.02**
SD		**0.00**		**0.00**		**0.00**								**0.00**
Rising Pond, RP-A	3.0	0.39	0.85	1.7	3.9	30	0.49	0.39	10	56	4.1	9.7	1.9	10
Rising Pond, RP-B	3.3	0.07	0.78	1.7	3.9	29	0.55	0.46	10	55	4.0	9.6	2.3	10
Rising Pond, RP-C	2.9	0.13	0.76	1.7	4.0	29	0.50	0.47	11	56	4.0	9.6	2.2	10
Mean	**3.1**	**0.20**	**0.80**	**1.7**	**4.0**	**29**	**0.52**	**0.44**	**10**	**56**	**4.0**	**9.6**	**2.1**	**10**
SD	**0.22**	**0.17**	**0.05**	**0.01**	**0.06**	**0.27**	**0.03**	**0.04**	**0.10**	**0.35**	**0.08**	**0.07**	**0.22**	**0.07**
Woods Pond, WP-A	21	1.8	2.3	3.4	12	75	1.0	1.3	43	120	8.5	29	4.5	15
Woods Pond, WP-B	19	1.6	2.2	4.4	13	73	1.7	1.1	36	120	10	29	5.6	15
Woods Pond, WP-C	18	0.91	1.5	3.9	12	74	1.2	1.5	43	120	9.9	29	3.6	15
Mean	**20**	**1.4**	**2.0**	**3.9**	**12**	**74**	**1.3**	**1.3**	**41**	**120**	**9.6**	**29**	**4.6**	**15**
SD	**1.3**	**0.45**	**0.43**	**0.51**	**0.42**	**1.1**	**0.39**	**0.21**	**3.7**	**0.0**	**0.94**	**0.29**	**1.0**	**0.19**
Deep Reach DRP-A	52	< 0.02	< 0.01	2.5	17	81	1.7	0.42	57	150	6.6	44	2.9	23
Deep Reach DRP-B	52	0.39	< 0.01	2.7	16	80	2.0	2.7	59	150	6.6	42	3.1	24
Deep Reach DRP-C	53	2.3	< 0.01	2.3	17	82	< 0.06	0.90	60	150	7.2	44	3.2	25
Mean	**52**			**2.5**	**17**	**81**	**1.8**	**1.3**	**59**	**150**	**6.8**	**43**	**3.1**	**24**
SD	**0.50**			**0.24**	**0.32**	**0.83**	**0.20**	**1.2**	**1.3**	**0.0**	**0.33**	**1.0**	**0.13**	**1.0**

Table 4 (Cont.). Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**083**	**084**	**085**	**086**	**087**	**090**	**091**	**092**	**095**	**096**	**097**	**099**	**101**	**102**
Three-Mile Pond, 3MP-A	< 0.01	< 0.17	18	< 0.01	< 0.29	0.04	< 0.08	< 0.17	< 0.19	< 0.01	< 0.29	0.26	0.35	< 0.02
Three-Mile Pond, 3MP-B	< 0.01	< 0.17	18	< 0.01	< 0.29	0.04	< 0.08	< 0.17	< 0.19	< 0.01	< 0.29	0.27	0.36	< 0.02
Three-Mile Pond, 3MP-C	< 0.01	< 0.17	18	< 0.01	< 0.29	0.04	< 0.08	< 0.17	< 0.19	< 0.01	< 0.29	0.26	0.36	< 0.02
Mean			**18**			**0.04**						**0.26**	**0.36**	
SD			**0.07**			**0.00**						**0.00**	**0.00**	
Rising Pond, RP-A	3.1	17	99	1.6	95	88	65	130	140	0.85	57	280	450	3.4
Rising Pond, RP-B	3.0	16	97	1.5	94	87	64	130	140	0.78	56	280	450	3.1
Rising Pond, RP-C	3.0	17	98	1.7	95	87	66	130	140	0.83	56	280	450	2.8
Mean	**3.0**	**16**	**98**	**1.6**	**95**	**87**	**65**	**130**	**140**	**0.82**	**56**	**280**	**450**	**3.1**
SD	**0.02**	**0.21**	**0.90**	**0.06**	**0.80**	**0.46**	**1.0**	**0.00**	**0.00**	**0.04**	**0.72**	**0.00**	**0.0**	**0.27**
Woods Pond, WP-A	9.2	49	150	3.1	260	130	130	320	430	2.9	130	450	1,000	2.4
Woods Pond, WP-B	8.9	49	150	3.6	250	120	130	320	430	2.8	130	450	1,000	0.5
Woods Pond, WP-C	9.0	48	150	2.8	250	120	130	320	430	3.2	120	450	1,000	2.1
Mean	**9.0**	**49**	**150**	**3.2**	**250**	**120**	**130**	**320**	**430**	**3.0**	**130**	**450**	**1,000**	**1.7**
SD	**0.13**	**0.53**	**0.0**	**0.42**	**5.8**	**5.8**	**0.0**	**0.0**	**0.0**	**0.20**	**5.8**	**0.0**	**0.0**	**1.0**
Deep Reach DRP-A	12	71	200	2.5	410	140	170	540	640	2.9	180	530	1,500	2.9
Deep Reach DRP-B	12	72	190	4.8	410	140	180	530	640	3.5	180	520	1,500	3.1
Deep Reach DRP-C	13	73	200	5.0	420	140	180	570	660	2.3	190	530	1,500	4.1
Mean	**12**	**72**	**200**	**4.1**	**410**	**140**	**180**	**550**	**650**	**2.9**	**180**	**530**	**1,500**	**3.4**
SD	**0.50**	**1**	**5.8**	**1.3**	**6**	**0**	**6**	**21**	**10**	**0.63**	**6**	**6**	**0**	**0.6**

Table 4 (Cont.). Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**105**	**109**	**110**	**112**	**113**	**114**	**115**	**117**	**118**	**119**	**122**	**123**	**128**	**129**
Three-Mile Pond, 3MP-A	< 0.24	0.09	< 0.40	0.01	0.08	0.01	< 0.02	< 0.02	0.56	0.04	< 0.01	< 0.01	0.14	0.02
Three-Mile Pond, 3MP-B	< 0.24	0.09	< 0.40	0.01	0.08	0.01	< 0.02	< 0.02	0.55	< 0.04	< 0.01	< 0.01	0.14	0.02
Three-Mile Pond, 3MP-C	< 0.24	0.09	< 0.40	0.01	0.08	0.01	< 0.02	< 0.02	0.57	< 0.04	< 0.01	< 0.01	0.14	0.02
Mean		**0.09**		**0.01**	**0.08**	**0.01**			**0.56**				**0.14**	**0.02**
SD		**0.00**		**0.00**	**0.00**	**0.00**			**0.01**				**0.00**	**0.00**
Rising Pond, RP-A	87	110	270	0.90	5.5	2.6	2.8	29	250	50	0.67	6.0	140	19
Rising Pond, RP-B	85	110	270	0.83	5.2	3.3	2.8	29	260	49	0.75	5.9	140	21
Rising Pond, RP-C	86	110	270	0.77	5.3	3.1	2.8	29	250	50	0.77	6.0	140	20
Mean	**86**	**110**	**270**	**0.83**	**5.3**	**3.0**	**2.8**	**29**	**250**	**50**	**0.73**	**6.0**	**140**	**20**
SD	**1.0**	**0.0**	**0.0**	**0.06**	**0.16**	**0.36**	**0.02**	**0.25**	**5.8**	**0.34**	**0.05**	**0.07**	**0.00**	**0.84**
Woods Pond, WP-A	150	220	600	0.39	14	11	6.9	28	520	96	1.3	6.3	260	41
Woods Pond, WP-B	140	220	600	1.7	14	8.4	6.7	29	530	96	1.1	5.9	250	41
Woods Pond, WP-C	140	220	580	0.37	15	8.2	6.8	27	510	95	1.1	6.2	250	39
Mean	**140**	**220**	**590**	**0.82**	**14**	**9.3**	**6.8**	**28**	**520**	**96**	**1.1**	**6.1**	**250**	**40**
SD	**5.8**	**0.0**	**12**	**0.77**	**0.62**	**1.7**	**0.09**	**0.58**	**10**	**0.56**	**0.12**	**0.21**	**5.8**	**0.82**
Deep Reach DRP-A	250	240	880	2.0	17	20	10	53	870	110	1.7	8.5	390	87
Deep Reach DRP-B	240	240	890	1.9	17	20	10	52	830	110	2.0	8.6	380	83
Deep Reach DRP-C	260	240	1,000	0.5	18	19	9.9	57	860	110	1.8	9.9	420	95
Mean	**250**	**240**	**920**	**1.5**	**17**	**20**	**10**	**54**	**850**	**110**	**1.8**	**9.0**	**400**	**88**
SD	**10**	**0**	**70**	**0.8**	**0.5**	**0.3**	**0.2**	**2.9**	**21**	**0**	**0.14**	**0.79**	**21**	**5.8**

Table 4 (Cont.). Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**130**	**131**	**132**	**133**	**134**	**136**	**137**	**138**	**139**	**141**	**144**	**146**	**147**	**149**
Three-Mile Pond, 3MP-A	0.06	< 0.02	< 0.33	0.04	0.06	0.05	0.05	1.0	< 0.02	0.14	0.03	0.31	< 0.01	0.31
Three-Mile Pond, 3MP-B	0.06	< 0.02	< 0.33	0.04	0.05	0.05	0.05	1.1	< 0.02	0.14	0.03	0.31	< 0.01	0.31
Three-Mile Pond, 3MP-C	0.06	< 0.02	< 0.33	0.04	0.05	0.05	0.05	1.0	< 0.02	0.13	0.03	0.31	< 0.01	0.31
Mean	**0.06**			**0.04**	**0.05**	**0.05**	**0.05**	**1.0**		**0.13**	**0.03**	**0.31**		**0.31**
SD	**0.00**			**0.00**	**0.00**	**0.00**	**0.0**	**0.0**		**0.00**	**0.00**	**0.00**		**0.00**
Rising Pond, RP-A	77	4.4	190	63	34	44	36	1,300	3.7	370	82	510	15	1,000
Rising Pond, RP-B	77	4.6	190	62	33	44	36	1,300	5.2	360	81	500	13	1,100
Rising Pond, RP-C	77	4.4	190	62	33	44	36	1,300	4.4	360	82	510	15	1,100
Mean	**77**	**4.5**	**190**	**62**	**33**	**44**	**36**	**1,300**	**4.4**	**360**	**82**	**510**	**14**	**1,100**
SD	**0.18**	**0.09**	**0.0**	**0.52**	**0.31**	**0.06**	**0.20**	**0.0**	**0.77**	**5.8**	**0.43**	**5.8**	**0.73**	**58**
Woods Pond, WP-A	130	12	700	100	86	160	62	2,800	11	860	230	870	19	2,500
Woods Pond, WP-B	130	11	690	100	85	160	62	2,800	11	850	230	870	18	2,400
Woods Pond, WP-C	130	11	670	100	84	160	62	2,800	11	840	230	860	18	2,400
Mean	**130**	**11**	**690**	**100**	**85**	**160**	**62**	**2,800**	**11**	**850**	**230**	**870**	**18**	**2,400**
SD	**0.0**	**0.24**	**15**	**0.0**	**0.74**	**0.0**	**0.33**	**0**	**0.14**	**10**	**0.0**	**5.8**	**0.96**	**58**
Deep Reach DRP-A	180	21	1,300	120	140	270	91	4,700	23	1,400	330	1,200	28	3,900
Deep Reach DRP-B	180	21	1,200	130	130	270	89	4,700	22	1,400	340	1,200	30	4,000
Deep Reach DRP-C	190	22	1,300	140	140	280	97	5,100	24	1,500	340	1,300	33	4,100
Mean	**180**	**21**	**1,300**	**130**	**140**	**270**	**92**	**4,800**	**23**	**1,400**	**340**	**1,200**	**30**	**4,000**
SD	**5.8**	**0.45**	**60**	**10**	**5.8**	**5.8**	**4.0**	**230**	**1.3**	**60**	**6**	**60**	**2.4**	**100**

Table 4 (Cont.). Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**151**	**153**	**156**	**157**	**158**	**163**	**164**	**166**	**167**	**170**	**171**	**172**	**173**	**174**
Three-Mile Pond, 3MP-A	0.12	1.6	0.04	< 0.04	0.09	0.23	0.04	< 0.01	0.04	0.42	0.06	0.08	< 0.02	0.09
Three-Mile Pond, 3MP-B	0.12	1.6	0.04	< 0.04	0.09	0.24	0.04	< 0.01	0.04	0.41	0.06	0.08	< 0.02	0.09
Three-Mile Pond, 3MP-C	0.11	1.6	0.06	< 0.04	0.09	0.23	0.04	< 0.01	0.04	0.43	0.06	0.08	< 0.02	0.10
Mean	**0.12**	**1.6**	**0.05**		**0.09**	**0.23**	**0.04**		**0.04**	**0.42**	**0.06**	**0.08**		**0.09**
SD	**0.00**	**0.01**	**0.01**		**0.00**	**0.00**	**0.0**		**0.0**	**0.01**	**0.00**	**0.0**		**0.00**
Rising Pond, RP-A	370	2,800	73	17	150	450	130	6.0	57	920	150	140	6.5	360
Rising Pond, RP-B	360	2,700	74	17	150	460	110	6.0	56	930	150	140	6.2	360
Rising Pond, RP-C	370	2,800	76	17	150	450	130	5.9	56	940	150	140	6.6	370
Mean	**370**	**2,800**	**74**	**17**	**150**	**450**	**120**	**5.9**	**56**	**930**	**150**	**140**	**6.4**	**360**
SD	**5.8**	**58**	**1.8**	**0.04**	**0.0**	**5.8**	**12**	**0.06**	**0.25**	**10**	**0.0**	**0.00**	**0.20**	**5.8**
Woods Pond, WP-A	960	4,900	120	21	320	840	280	7.7	92	1,600	350	270	24	1,100
Woods Pond, WP-B	960	4,900	130	22	320	820	290	7.8	92	1,600	350	270	22	1,100
Woods Pond, WP-C	950	4,800	130	21	310	820	290	7.6	90	1,600	350	260	26	1,100
Mean	**960**	**4,900**	**130**	**21**	**320**	**830**	**290**	**7.7**	**91**	**1,600**	**350**	**270**	**24**	**1,100**
SD	**5.8**	**58**	**5.8**	**0.74**	**5.8**	**12**	**5.8**	**0.11**	**1.1**	**0.0**	**0.0**	**5.8**	**1.9**	**0.0**
Deep Reach DRP-A	1,300	7,400	160	25	520	1,300	270	10	140	2,500	590	480	46	1,900
Deep Reach DRP-B	1,300	7,400	160	27	520	1,300	410	10	140	2,600	590	470	48	2,000
Deep Reach DRP-C	1,400	7,800	160	26	560	1,400	320	11	150	2,600	590	480	50	1,900
Mean	**1,300**	**7,500**	**160**	**26**	**530**	**1,300**	**330**	**10**	**140**	**2,600**	**590**	**480**	**48**	**1,900**
SD	**60**	**230**	**0**	**1**	**23**	**60**	**70**	**0.3**	**6**	**60**	**0**	**6**	**2**	**60**

Table 4 (Cont.). Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**175**	**176**	**177**	**178**	**179**	**180**	**183**	**185**	**187**	**189**	**190**	**191**	**193**	**194**
Three-Mile Pond, 3MP-A	< 0.10	0.02	0.08	0.04	< 0.06	0.91	0.28	0.02	0.59	0.02	0.14	< 0.01	0.06	0.13
Three-Mile Pond, 3MP-B	< 0.10	0.02	0.08	0.04	< 0.06	0.91	0.28	0.02	0.60	0.02	0.14	< 0.01	0.06	0.14
Three-Mile Pond, 3MP-C	< 0.10	0.02	0.08	0.04	< 0.06	0.90	0.30	0.02	0.60	0.02	0.16	< 0.01	0.06	0.14
Mean		**0.02**	**0.08**	**0.04**		**0.91**	**0.29**	**0.02**	**0.60**	**0.02**	**0.15**		**0.06**	**0.13**
SD		**0.00**	**0.00**	**0.00**		**0.00**	**0.01**	**0.00**	**0.01**	**0.00**	**0.01**		**0.00**	**0.00**
Rising Pond, RP-A	30	11	260	140	54	1,700	510	60	1,100	29	210	28	110	250
Rising Pond, RP-B	31	13	260	140	55	1,700	510	61	1,100	29	210	28	110	250
Rising Pond, RP-C	31	8.6	260	140	54	1,700	510	61	1,100	29	210	28	110	250
Mean	**31**	**11**	**260**	**140**	**54**	**1,700**	**510**	**61**	**1,100**	**29**	**210**	**28**	**110**	**250**
SD	**0.42**	**2.0**	**0.0**	**0.00**	**0.57**	**0.0**	**0.0**	**0.47**	**0.0**	**0.19**	**0.0**	**0.17**	**0.00**	**0.00**
Woods Pond, WP-A	66	93	590	300	230	3,200	1,100	150	2,100	53	480	61	210	520
Woods Pond, WP-B	66	88	600	300	230	3,200	1,100	150	2,100	53	480	60	210	520
Woods Pond, WP-C	66	89	590	300	220	3,200	1,100	150	2,100	53	480	62	210	520
Mean	**66**	**90**	**590**	**300**	**230**	**3,200**	**1,100**	**150**	**2,100**	**53**	**480**	**61**	**210**	**520**
SD	**0.08**	**2.8**	**5.8**	**0.0**	**5.8**	**0.0**	**0.0**	**0.0**	**0.0**	**0.08**	**0.0**	**0.81**	**0.0**	**0.0**
Deep Reach DRP-A	50	200	1,000	490	350	5,700	2,100	250	4,100	100	1,000	120	390	990
Deep Reach DRP-B	67	170	1,000	490	340	5,700	2,100	250	4,100	100	990	110	390	970
Deep Reach DRP-C	49	180	1,000	480	350	5,800	2,100	250	4,000	100	990	120	400	990
Mean	**55**	**180**	**1,000**	**490**	**350**	**5,700**	**2,100**	**250**	**4,100**	**100**	**990**	**120**	**390**	**980**
SD	**10**	**20**	**0**	**10**	**10**	**60**	**0**	**0**	**60**	**0.0**	**10**	**5.8**	**5.8**	**10**

Table 4 (Cont.). Concentrations (ng/g) of PCB congeners in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**195**	**196**	**197**	**198**	**199**	**200**	**201**	**202**	**203**	**205**	**206**	**208**	**209**	**Total PCBs**
Three-Mile Pond, 3MP-A	0.06	< 0.19	< 0.15	< 0.01	0.17	< 0.02	< 0.02	0.06	0.15	0.01	0.10	0.02	0.05	30
Three-Mile Pond, 3MP-B	0.06	< 0.19	< 0.15	< 0.01	0.17	< 0.02	< 0.02	0.06	0.16	0.01	0.10	0.03	0.05	30
Three-Mile Pond, 3MP-C	0.06	< 0.19	< 0.15	< 0.01	0.18	< 0.02	< 0.02	0.07	0.17	0.01	0.10	0.02	0.05	30
Mean	**0.06**				**0.17**			**0.06**	**0.16**	**0.01**	**0.10**	**0.02**	**0.05**	**30**
SD	**0.00**				**0.00**			**0.00**	**0.01**	**0.00**	**0.00**	**0.01**	**0.00**	**0.02**
Rising Pond, RP-A	99	110	12	13	220	9.6	21	30	210	16	37	3.3	7.7	18,000
Rising Pond, RP-B	100	110	12	13	220	9.4	21	30	210	16	37	3.3	7.7	19,000
Rising Pond, RP-C	100	100	12	13	230	9.5	21	31	210	17	38	3.4	7.9	18,000
Mean	**100**	**110**	**12**	**13**	**220**	**9.5**	**21**	**30**	**210**	**16**	**37**	**3.3**	**7.7**	**18,000**
SD	**0.5**	**5.8**	**0.18**	**0.07**	**5.8**	**0.12**	**0.03**	**0.56**	**0.0**	**0.28**	**0.34**	**0.07**	**0.11**	**580**
Woods Pond, WP-A	110	300	42	28	550	46	62	85	420	34	90	12	29	37,000
Woods Pond, WP-B	120	300	40	27	540	45	60	84	420	33	89	12	29	37,000
Woods Pond, WP-C	120	290	40	26	540	45	60	88	420	33	89	12	29	37,000
Mean	**120**	**300**	**41**	**27**	**540**	**45**	**61**	**86**	**420**	**33**	**89**	**12**	**29**	**37,000**
SD	**5.8**	**5.8**	**1.3**	**1.0**	**5.8**	**0.54**	**0.95**	**1.7**	**0.0**	**0.18**	**0.30**	**0.16**	**0.35**	**0**
Deep Reach DRP-A	360	610	93	43	1,000	100	110	150	760	63	150	26	81	61,000
Deep Reach DRP-B	300	570	89	48	990	99	110	150	760	63	150	26	78	61,000
Deep Reach DRP-C	360	590	93	43	1,000	100	110	150	760	61	150	26	84	63,000
Mean	**340**	**590**	**92**	**45**	**1,000**	**100**	**110**	**150**	**760**	**62**	**150**	**26**	**81**	**62,000**
SD	**30**	**20**	**2.4**	**2.6**	**10**	**0.4**	**0**	**0**	**0**	**1**	**0**	**0.5**	**3.21**	**1,200**

Table 5. Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**001**	**003**	**004**	**005**	**006**	**007**	**008**	**009**	**010**	**015**	**016**	**017**	**018**	**019**
Three-Mile Pond, 3MP-A	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.10	< 0.01	< 0.02	< 0.07	< 0.14
Three-Mile Pond, 3MP-B	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.10	< 0.01	< 0.02	< 0.07	< 0.14
Three-Mile Pond, 3MP-C	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.10	< 0.01	< 0.02	< 0.07	< 0.14
Mean										**0.10**				
SD										**0.00**				
Rising Pond, RP-A	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	1.2	1.8	1.6	1.6
Rising Pond, RP-B	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	1.9	1.6	2.0
Rising Pond, RP-C	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	1.8	1.5	1.6
Mean												**1.8**	**1.6**	**1.7**
SD												**0.1**	**0.1**	**0.2**
Woods Pond, WP-A	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	12	10.2	9.9
Woods Pond, WP-B	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	11	9.1	9.4
Woods Pond, WP-C	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	12	9.4	14.0
Mean												**12**	**9.6**	**11.1**
SD												**0**	**0.6**	**2.5**
Deep Reach DRP-A	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	19	14	14
Deep Reach DRP-B	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.60	< 0.01	19	14	12
Deep Reach DRP-C	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	16	11	32
Mean												**18**	**13**	**19**
SD												**2**	**2**	**11**

Table 5 (Cont.). Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**020**	**022**	**024**	**025**	**026**	**027**	**028**	**031**	**032**	**033**	**034**	**035**	**037,059**	**040**
Three-Mile Pond, 3MP-A	< 0.01	< 0.04	< 0.05	< 0.01	< 0.01	< 0.04	< 0.04	< 0.02	< 0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02
Three-Mile Pond, 3MP-B	< 0.01	< 0.04	< 0.05	< 0.01	< 0.01	< 0.04	< 0.04	< 0.02	< 0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02
Three-Mile Pond, 3MP-C	< 0.01	< 0.04	< 0.05	< 0.01	< 0.01	< 0.04	< 0.04	< 0.02	< 0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02
Mean														
SD														
Rising Pond, RP-A	< 0.01	0.40	< 0.05	1.7	3.0	0.6	6.3	0.80	1.7	< 0.04	< 0.17	< 0.10	0.60	0.50
Rising Pond, RP-B	< 0.01	0.40	< 0.05	1.7	3.1	0.7	6.1	1.00	4.0	< 0.04	< 0.17	< 0.10	0.70	0.60
Rising Pond, RP-C	< 0.01	0.30	< 0.05	1.9	3.1	0.8	6.2	1.0	4.3	< 0.04	< 0.17	< 0.10	0.60	0.60
Mean		**0.40**		**1.8**	**3.1**	**0.7**	**6.2**	**0.90**	**3.3**				**0.60**	**0.60**
SD		**0.10**		**0.1**	**0.1**	**0.1**	**0.1**	**0.10**	**1.4**				**0.10**	**0.10**
Woods Pond, WP-A	< 0.01	1.7	< 0.05	13	22	4.5	29	6.2	25	2.5	< 0.17	< 0.10	2.7	1.8
Woods Pond, WP-B	< 0.01	2.0	< 0.05	12	23	4.4	29	6.2	24	2.0	< 0.17	0.20	1.7	1.8
Woods Pond, WP-C	< 0.01	2.4	< 0.05	12	22	4.1	29	6.5	23	2.4	< 0.17	< 0.10	2.7	1.5
Mean		**2.0**		**12**	**22**	**4.3**	**29**	**6.3**	**24**	**2.3**			**2.4**	**1.7**
SD		**0.4**		**1**	**0**	**0.2**	**0**	**0.2**	**1**	**0.3**			**0.6**	**0.2**
Deep Reach DRP-A	< 0.01	2.3	< 0.05	8.8	23	6.4	24	10	51	1.2	< 0.17	1.2	< 0.02	2.2
Deep Reach DRP-B	< 0.01	2.8	< 0.05	7.7	26	7.2	25	7.7	50	2.2	3.9	< 0.10	2.8	2.0
Deep Reach DRP-C	< 0.01	3.2	< 0.05	9.5	23	7.0	27	10	39	0.60	0.60	< 0.10	3.8	2.9
Mean		**2.8**		**8.7**	**24**	**6.9**	**25**	**9**	**46**	**1.3**				**2.4**
SD		**0.5**		**0.9**	**2**	**0.4**	**2**	**1**	**7**	**0.8**				**0.5**

Table 5 (Cont.). Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**041**	**042**	**043**	**044**	**045**	**046**	**047**	**048**	**049**	**051**	**052**	**053**	**054**	**055**
Three-Mile Pond, 3MP-A	< 0.02	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.10	< 11.97	0.10	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02
Three-Mile Pond, 3MP-B	< 0.02	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.10	< 11.97	0.10	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02
Three-Mile Pond, 3MP-C	< 0.02	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.10	< 11.97	0.10	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02
Mean							**0.10**		**0.10**					
SD							**0.00**		**0.00**					
Rising Pond, RP-A	< 0.02	14	1.4	15	0.50	0.50	220	65	120	11	92	10	< 0.02	< 0.02
Rising Pond, RP-B	< 0.02	16	1.7	15	0.60	0.50	220	14	120	11	90	9.9	< 0.02	< 0.02
Rising Pond, RP-C	< 0.02	14	3.7	15	0.60	0.40	220	24	120	11	92	10	< 0.02	< 0.02
Mean		**15**	**2.3**	**15**	**0.60**	**0.50**	**220**	**35**	**120**	**11**	**92**	**10**		
SD		**1**	**1.3**	**0**	**0.10**	**0.10**	**0**	**27**	**0**	**0**	**1**	**0**		
Woods Pond, WP-A	< 0.02	110	5.7	55	0.50	1.8	590	310	380	47	300	43	0.90	< 0.02
Woods Pond, WP-B	0.70	120	4.0	54	1.3	1.8	600	330	380	46	310	42	0.90	< 0.02
Woods Pond, WP-C	< 0.02	110	4.5	55	1.1	1.5	580	310	380	45	300	42	1.1	< 0.02
Mean		**113**	**4.7**	**55**	**1.00**	**1.7**	**590**	**317**	**380**	**46**	**303**	**42**	**1.00**	
SD		**6**	**0.9**	**1**	**0.40**	**0.2**	**10**	**12**	**0**	**1**	**6**	**0**	**0.10**	
Deep Reach DRP-A	< 0.02	120	7.0	62	2.2	2.2	740	790	410	53	360	48	0.50	< 0.02
Deep Reach DRP-B	< 0.02	120	8.6	66	< 0.12	1.5	730	770	410	56	360	50	2.0	< 0.02
Deep Reach DRP-C	< 0.02	120	8.8	65	< 0.12	2.3	760	900	410	56	370	50	0.60	< 0.02
Mean		**120**	**8.1**	**64**		**2.0**	**743**	**820**	**410**	**55**	**363**	**50**	**1.00**	
SD		**0**	**1.0**	**2**		**0.4**	**15**	**70**	**0**	**2**	**6**	**1**	**0.80**	

Table 5 (Cont.). Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**056,060**	**057**	**058**	**063**	**064**	**066**	**067**	**069**	**070**	**071**	**072**	**074**	**075**	**082**
Three-Mile Pond, 3MP-A	< 1.69	< 0.02	< 0.01	< 0.01	< 0.02	0.10	< 0.06	< 0.01	0.20	< 0.23	< 0.01	< 0.13	< 0.01	< 0.01
Three-Mile Pond, 3MP-B	< 1.69	< 0.02	< 0.01	< 0.01	< 0.02	0.10	< 0.06	< 0.01	0.20	< 0.23	< 0.01	< 0.13	< 0.01	< 0.01
Three-Mile Pond, 3MP-C	< 1.69	< 0.02	< 0.01	< 0.01	< 0.02	0.10	< 0.06	< 0.01	0.20	< 0.23	< 0.01	< 0.13	< 0.01	< 0.01
Mean						**0.10**			**0.20**					
SD						**0.00**			**0.00**					
Rising Pond, RP-A	3.8	0.50	1.1	2.2	5.1	37.9	0.60	0.50	13	72	5.3	13	2.4	13
Rising Pond, RP-B	4.3	0.10	1.0	2.2	5.0	37.6	0.70	0.60	13	71	5.1	12	2.9	13
Rising Pond, RP-C	3.8	0.20	1.0	2.2	5.2	37.2	0.60	0.60	14	72	5.2	12	2.8	13
Mean	**4.0**	**0.30**	**1.0**	**2.2**	**5.1**	**37.6**	**0.60**	**0.60**	**13**	**71**	**5.2**	**12**	**2.7**	**13**
SD	**0.3**	**0.20**	**0.1**	**0.0**	**0.1**	**0.4**	**0.10**	**0.10**	**0**	**0**	**0.1**	**0**	**0.3**	**0**
Woods Pond, WP-A	30	2.5	3.2	4.8	17	110	1.4	1.8	61	170	12	41	6.4	22
Woods Pond, WP-B	28	2.2	3.2	6.2	18	100	2.4	1.6	52	170	15	41	8.0	22
Woods Pond, WP-C	26	1.3	2.1	5.6	17	110	1.7	2.2	60	170	14	41	5.2	21
Mean	**28**	**2.0**	**2.8**	**5.5**	**17**	**107**	**1.8**	**1.9**	**58**	**170**	**14**	**41**	**6.5**	**22**
SD	**2**	**0.6**	**0.6**	**0.7**	**1**	**6**	**0.5**	**0.3**	**5**	**0**	**1**	**0**	**1.4**	**0**
Deep Reach DRP-A	68	< 0.02	< 0.01	3.2	22	110	2.2	0.50	75	190	8.6	57	3.8	30
Deep Reach DRP-B	68	0.50	< 0.01	3.6	21	100	2.5	3.6	76	190	8.6	55	4.1	31
Deep Reach DRP-C	69	2.9	< 0.01	2.9	22	110	< 0.06	1.2	78	190	9.4	57	4.1	33
Mean	**68**			**3.2**	**22**	**107**		**1.80**	**76**	**190**	**8.9**	**56**	**4.0**	**31**
SD	**1**			**0.4**	**0**	**6**		**1.60**	**2**	**0**	**0.5**	**1**	**0.2**	**1**

Table 5 (Cont.). Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**083**	**084**	**085**	**086**	**087**	**090**	**091**	**092**	**095**	**096**	**097**	**099**	**101**	**102**
Three-Mile Pond, 3MP-A	< 0.01	< 0.17	28.1	< 0.01	< 0.29	0.10	0.10	0.20	< 0.19	< 0.01	< 0.29	0.40	0.50	< 0.02
Three-Mile Pond, 3MP-B	< 0.01	< 0.17	28.0	< 0.01	< 0.29	0.10	0.10	0.20	< 0.19	< 0.01	< 0.29	0.40	0.50	< 0.02
Three-Mile Pond, 3MP-C	< 0.01	< 0.17	27.8	< 0.01	< 0.29	0.10	0.10	0.20	< 0.19	< 0.01	< 0.29	0.40	0.50	< 0.02
Mean			**28.0**			**0.10**	**0.10**	**0.20**				**0.40**	**0.50**	
SD			**0.2**			**0.00**	**0.00**	**0.00**				**0.00**	**0.00**	
Rising Pond, RP-A	3.9	21	130	2.1	120	110	83	170	180	1.1	73	350	580	4.3
Rising Pond, RP-B	3.9	21	120	2.0	120	110	82	160	180	1.0	71	350	570	4.0
Rising Pond, RP-C	3.9	21	120	2.1	120	110	85	170	180	1.1	72	360	580	3.6
Mean	**3.9**	**21**	**123**	**2.1**	**120**	**110**	**84**	**167**	**180**	**1.1**	**72**	**353**	**577**	**4.0**
SD	**0.0**	**0**	**6**	**0.1**	**0**	**0**	**1**	**6**	**0**	**0.1**	**1**	**6**	**6**	**0.4**
Woods Pond, WP-A	13	70	210	4.5	360	180	190	460	610	4.1	180	640	1,500	3.4
Woods Pond, WP-B	13	69	210	5.1	360	180	180	450	610	4.0	180	640	1,500	0.70
Woods Pond, WP-C	13	68	210	3.9	360	170	180	440	610	4.5	180	640	1,500	3.0
Mean	**13**	**69**	**210**	**4.5**	**360**	**177**	**183**	**450**	**610**	**4.2**	**180**	**640**	**1,500**	**2.4**
SD	**0**	**1**	**0**	**0.6**	**0**	**6**	**6**	**10**	**0**	**0.3**	**0**	**0**	**0**	**1.5**
Deep Reach DRP-A	15	93	260	3.3	530	180	230	710	830	3.8	240	680	2,000	3.8
Deep Reach DRP-B	16	93	250	6.2	530	180	230	680	840	4.6	240	680	1,900	4.1
Deep Reach DRP-C	16	94	260	6.4	550	180	240	740	850	2.9	250	690	2,000	5.3
Mean	**16**	**94**	**257**	**5.3**	**537**	**180**	**233**	**710**	**840**	**3.8**	**243**	**683**	**1,967**	**4.4**
SD	**1**	**1**	**6**	**1.7**	**12**	**0**	**6**	**30**	**10**	**0.9**	**6**	**6**	**58**	**0.8**

Table 5 (Cont.). Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**105**	**109**	**110**	**112**	**113**	**114**	**115**	**117**	**118**	**119**	**122**	**123**	**128**	**129**
Three-Mile Pond, 3MP-A	0.30	0.10	0.40	< 0.01	0.10	< 0.01	< 0.02	< 0.02	0.80	0.10	< 0.01	< 0.01	0.20	< 0.01
Three-Mile Pond, 3MP-B	0.30	0.10	0.40	< 0.01	0.10	< 0.01	< 0.02	< 0.02	0.80	0.10	< 0.01	< 0.01	0.20	< 0.01
Three-Mile Pond, 3MP-C	0.30	0.10	0.40	< 0.01	0.10	< 0.01	< 0.02	< 0.02	0.90	0.10	< 0.01	< 0.01	0.20	< 0.01
Mean	**0.30**	**0.10**	**0.40**		**0.10**				**0.80**	**0.10**			**0.20**	
SD	**0.00**	**0.00**	**0.00**		**0.00**				**0.10**	**0.00**			**0.00**	
Rising Pond, RP-A	110	140	350	1.1	7.0	3.3	3.6	37	310	64	0.90	7.7	180	25
Rising Pond, RP-B	110	140	340	1.1	6.6	4.2	3.5	37	330	63	1.00	7.6	170	27
Rising Pond, RP-C	110	140	350	1.0	6.8	4.0	3.6	37	310	64	1.00	7.7	170	26
Mean	**110**	**140**	**347**	**1.1**	**6.8**	**3.8**	**3.6**	**37**	**317**	**64**	**1.00**	**7.7**	**173**	**26**
SD	**0**	**0**	**6**	**0.1**	**0.2**	**0.5**	**0.1**	**0**	**12**	**1**	**0.10**	**0.1**	**6**	**1**
Woods Pond, WP-A	210	310	850	0.60	20	16	9.8	40	740	140	1.8	8.9	360	58
Woods Pond, WP-B	200	320	830	2.4	19	12	9.6	41	750	140	1.6	8.3	360	58
Woods Pond, WP-C	200	310	830	0.50	21	12	9.7	39	730	140	1.5	8.7	360	56
Mean	**203**	**313**	**837**	**1.20**	**20**	**13**	**9.7**	**40**	**740**	**140**	**1.6**	**8.6**	**360**	**57**
SD	**6**	**6**	**12**	**1.10**	**1**	**2**	**0.1**	**1**	**10**	**0**	**0.2**	**0.3**	**0**	**1**
Deep Reach DRP-A	320	320	1,100	2.6	22	26	14	69	1,100	140	2.2	11	500	110
Deep Reach DRP-B	320	310	1,200	2.5	23	25	13	68	1,100	140	2.5	11	490	110
Deep Reach DRP-C	340	320	1,300	0.70	23	25	13	75	1,100	150	2.3	13	540	120
Mean	**327**	**317**	**1,200**	**1.9**	**23**	**26**	**13**	**70**	**1,100**	**143**	**2.3**	**12**	**510**	**113**
SD	**12**	**6**	**100**	**1.1**	**1**	**0**	**0**	**4**	**0**	**6**	**0.2**	**1**	**27**	**6**

Table 5 (Cont.). Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**130**	**131**	**132**	**133**	**134**	**136**	**137**	**138**	**139**	**141**	**144**	**146**	**147**	**149**
Three-Mile Pond, 3MP-A	0.10	< 0.02	< 0.33	0.10	0.10	0.10	0.10	1.60	< 0.02	0.20	< 0.01	0.50	< 0.01	0.50
Three-Mile Pond, 3MP-B	0.10	< 0.02	< 0.33	0.10	0.10	0.10	0.10	1.60	< 0.02	0.20	< 0.01	0.50	< 0.01	0.50
Three-Mile Pond, 3MP-C	0.10	< 0.02	< 0.33	0.10	0.10	0.10	0.10	1.60	< 0.02	0.20	< 0.01	0.50	< 0.01	0.50
Mean	**0.10**			**0.10**	**0.10**	**0.10**	**0.10**	**1.60**		**0.20**		**0.50**		**0.50**
SD	**0.00**			**0.00**	**0.00**	**0.00**	**0.00**	**0.00**		**0.00**		**0.00**		**0.00**
Rising Pond, RP-A	99	5.6	240	81	43	56	47	1,700	4.7	470	100	660	19	1,300
Rising Pond, RP-B	98	5.8	240	80	42	56	46	1,600	6.7	460	100	650	17	1,400
Rising Pond, RP-C	98	5.7	240	80	43	56	46	1,600	5.6	460	100	650	19	1,400
Mean	**98**	**5.7**	**240**	**80**	**43**	**56**	**46**	**1,633**	**5.7**	**463**	**100**	**653**	**18**	**1,367**
SD	**0**	**0.1**	**0**	**1**	**0**	**0**	**0**	**58**	**1.0**	**6**	**0**	**6**	**1**	**58**
Woods Pond, WP-A	190	16	1000	150	120	230	90	4,000	16	1,200	330	1,200	28	3,500
Woods Pond, WP-B	190	16	970	150	120	230	90	4,000	15	1,200	330	1,200	25	3,400
Woods Pond, WP-C	190	16	950	140	120	220	90	3,900	15	1,200	320	1,200	26	3,400
Mean	**190**	**16**	**973**	**147**	**120**	**227**	**90**	**3,967**	**15**	**1,200**	**327**	**1,200**	**26**	**3,433**
SD	**0**	**0**	**25**	**6**	**0**	**6**	**0**	**58**	**0**	**0**	**6**	**0**	**1**	**58**
Deep Reach DRP-A	230	27	1,600	170	180	350	120	6,100	30	1,800	430	1,600	37	5,100
Deep Reach DRP-B	230	27	1,500	170	170	350	120	6,100	28	1,800	440	1,600	39	5,200
Deep Reach DRP-C	250	28	1,700	180	190	360	130	6,600	32	1,900	440	1,700	43	5,400
Mean	**237**	**28**	**1,600**	**173**	**180**	**353**	**123**	**6,267**	**30**	**1,833**	**437**	**1,633**	**40**	**5,233**
SD	**12**	**1**	**100**	**6**	**10**	**6**	**6**	**289**	**2**	**58**	**6**	**58**	**3**	**153**

Table 5 (Cont.). Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**151**	**153**	**156**	**157**	**158**	**163**	**164**	**166**	**167**	**170**	**171**	**172**	**173**	**174**
Three-Mile Pond, 3MP-A	0.20	2.50	0.10	< 0.04	0.10	0.40	0.10	< 0.01	0.10	0.60	0.10	0.10	< 0.02	0.10
Three-Mile Pond, 3MP-B	0.20	2.50	0.10	< 0.04	0.10	0.40	0.10	< 0.01	0.10	0.60	0.10	0.10	< 0.02	0.10
Three-Mile Pond, 3MP-C	0.20	2.50	0.10	< 0.04	0.10	0.30	0.10	< 0.01	0.10	0.70	0.10	0.10	< 0.02	0.10
Mean	**0.20**	**2.50**	**0.10**		**0.10**	**0.40**	**0.10**		**0.10**	**0.60**	**0.10**	**0.10**		**0.10**
SD	**0.00**	**0.00**	**0.00**		**0.00**	**0.10**	**0.00**		**0.00**	**0.10**	**0.00**	**0.00**		**0.00**
Rising Pond, RP-A	470	3,500	93	22	190	580	170	7.7	72	1,200	190	180	8.4	460
Rising Pond, RP-B	470	3,500	95	22	190	590	150	7.6	72	1,200	190	180	8.0	470
Rising Pond, RP-C	470	3,500	97	22	190	580	160	7.5	72	1,200	190	180	8.4	470
Mean	**470**	**3,500**	**95**	**22**	**190**	**583**	**160**	**7.6**	**72**	**1,200**	**190**	**180**	**8.3**	**467**
SD	**0**	**0**	**2**	**0**	**0**	**6**	**10**	**0.1**	**0**	**0**	**0**	**0**	**0.2**	**6**
Woods Pond, WP-A	1,400	7,000	180	29	450	1,200	400	11	130	2,200	500	390	34	1,600
Woods Pond, WP-B	1,400	7,000	180	31	450	1,200	420	11	130	2,200	500	380	32	1,500
Woods Pond, WP-C	1,300	6,900	180	30	440	1,200	410	11	130	2,200	490	380	37	1,600
Mean	**1,367**	**6,967**	**180**	**30**	**447**	**1,200**	**410**	**11**	**130**	**2,200**	**497**	**383**	**34**	**1,567**
SD	**58**	**58**	**0**	**1**	**6**	**0**	**10**	**0**	**0**	**0**	**6**	**6**	**3**	**58**
Deep Reach DRP-A	1,700	9,600	200	32	680	1,700	350	14	180	3,300	760	620	60	2,500
Deep Reach DRP-B	1,700	9,600	210	35	670	1,600	530	13	180	3,400	760	620	62	2,500
Deep Reach DRP-C	1,800	10,100	210	34	730	1,800	410	14	200	3,400	760	630	65	2,500
Mean	**1,733**	**9,767**	**207**	**34**	**693**	**1,700**	**430**	**14**	**187**	**3,367**	**760**	**623**	**62**	**2,500**
SD	**58**	**289**	**6**	**2**	**32**	**100**	**92**	**1**	**12**	**58**	**0**	**6**	**3**	**0**

Table 5 (Cont.). Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**175**	**176**	**177**	**178**	**179**	**180**	**183**	**185**	**187**	**189**	**190**	**191**	**193**	**194**
Three-Mile Pond, 3MP-A	< 0.10	< 0.01	0.10	0.10	< 0.06	1.4	0.40	< 0.01	0.90	< 0.02	0.20	< 0.01	0.10	0.20
Three-Mile Pond, 3MP-B	< 0.10	< 0.01	0.10	0.10	< 0.06	1.4	0.40	< 0.01	0.90	< 0.02	0.20	< 0.01	0.10	0.20
Three-Mile Pond, 3MP-C	< 0.10	< 0.01	0.10	0.10	< 0.06	1.4	0.50	< 0.01	0.90	< 0.02	0.20	< 0.01	0.10	0.20
Mean			**0.10**	**0.10**		**1.4**	**0.40**		**0.90**		**0.20**		**0.10**	**0.20**
SD			**0.00**	**0.00**		**0.0**	**0.10**		**0.00**		**0.00**		**0.00**	**0.00**
Rising Pond, RP-A	39	14	340	170	69	2,200	650	77	1,400	37	270	35	140	320
Rising Pond, RP-B	40	16	330	170	70	2,100	650	78	1,400	37	270	36	140	330
Rising Pond, RP-C	40	11	330	180	69	2,200	660	79	1,400	37	270	36	140	330
Mean	**39**	**14**	**333**	**173**	**69**	**2,167**	**653**	**78**	**1,400**	**37**	**270**	**36**	**140**	**327**
SD	**1**	**3**	**6**	**6**	**1**	**58**	**6**	**1**	**0**	**0**	**0**	**0**	**0**	**6**
Woods Pond, WP-A	94	130	840	430	320	4,500	1,600	210	3,000	75	680	87	300	730
Woods Pond, WP-B	94	130	850	420	320	4,600	1,600	210	3,000	75	680	85	300	730
Woods Pond, WP-C	94	130	840	420	320	4,500	1,600	210	3,000	75	680	88	300	730
Mean	**94**	**130**	**843**	**423**	**320**	**4,533**	**1,600**	**210**	**3,000**	**75**	**680**	**87**	**300**	**730**
SD	**0**	**0**	**6**	**6**	**0**	**58**	**0**	**0**	**0**	**0**	**0**	**1**	**0**	**0**
Deep Reach DRP-A	65	260	1,300	630	450	7,400	2,700	330	5,300	130	1,300	150	510	1,300
Deep Reach DRP-B	87	220	1,300	630	450	7,400	2,700	320	5,300	130	1,300	150	510	1,300
Deep Reach DRP-C	64	240	1,300	620	450	7,500	2,700	320	5,200	140	1,300	150	520	1,300
Mean	**72**	**240**	**1,300**	**627**	**450**	**7,433**	**2,700**	**323**	**5,267**	**133**	**1,300**	**150**	**513**	**1,300**
SD	**13**	**20**	**0**	**6**	**0**	**58**	**0**	**6**	**58**	**6**	**0**	**0**	**6**	**0**

Table 5 (Cont.). Concentrations (ng/µL) of PCB congeners in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	IUPAC Congener Number													
Site / Sample ID	**195**	**196**	**197**	**198**	**199**	**200**	**201**	**202**	**203**	**205**	**206**	**208**	**209**	**Total PCBs**
Three-Mile Pond, 3MP-A	0.10	< 0.19	< 0.15	< 0.01	0.30	< 0.02	< 0.02	0.10	0.20	< 0.01	0.20	< 0.26	< 2.5	46
Three-Mile Pond, 3MP-B	0.10	< 0.19	< 0.15	< 0.01	0.30	< 0.02	< 0.02	0.10	0.20	< 0.01	0.20	< 0.26	< 2.5	46
Three-Mile Pond, 3MP-C	0.10	< 0.19	< 0.15	< 0.01	0.30	< 0.02	< 0.02	0.10	0.30	< 0.01	0.20	< 0.26	< 2.5	46
Mean	**0.10**				**0.30**			**0.10**	**0.20**		**0.20**			**46**
SD	**0.00**				**0.00**			**0.00**	**0.10**		**0.00**			**0**
Rising Pond, RP-A	130	140	16	17	290	12	27	38	270	21	48	4.2	9.8	22,000
Rising Pond, RP-B	130	140	15	17	290	12	27	39	270	21	48	4.2	9.8	22,000
Rising Pond, RP-C	130	130	15	17	290	12	28	40	270	21	48	4.4	10	22,000
Mean	**130**	**137**	**15**	**17**	**290**	**12**	**27**	**39**	**270**	**21**	**48**	**4.3**	**9.9**	**22,000**
SD	**0**	**6**	**0**	**0**	**0**	**0**	**0**	**1**	**0**	**0**	**0**	**0.1**	**0.2**	**0**
Woods Pond, WP-A	160	430	60	39	780	65	88	120	590	48	130	17	42	53,000
Woods Pond, WP-B	170	420	57	38	770	64	86	120	590	48	130	17	41	53,000
Woods Pond, WP-C	170	410	56	36	770	64	85	120	590	47	130	17	41	52,000
Mean	**167**	**420**	**58**	**38**	**773**	**64**	**86**	**120**	**590**	**47**	**130**	**17**	**41**	**52,667**
SD	**6**	**10**	**2**	**1**	**6**	**1**	**1**	**0**	**0**	**0**	**0**	**0**	**1**	**577**
Deep Reach DRP-A	470	790	120	56	1,300	130	140	200	990	82	190	34	100	79,000
Deep Reach DRP-B	390	740	120	62	1,300	130	140	190	990	82	200	33	100	79,000
Deep Reach DRP-C	470	770	120	56	1,300	130	140	200	990	80	190	34	110	81,000
Mean	**443**	**767**	**120**	**58**	**1,300**	**130**	**140**	**197**	**990**	**81**	**193**	**34**	**103**	**79,667**
SD	**46**	**25**	**0**	**3**	**0**	**0**	**0**	**6**	**0**	**1**	**6**	**1**	**6**	**1,155**

Table 6. Concentrations (ng/g) of organochlorine pesticides in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

Site / Sample ID	HCB	PCA	alpha-BHC	Lindane	delta-BHC	Heptachlor	Heptachlor Epoxide	Oxy-chlordane	cis-Chlordane	trans-Chlordane	cis-Nonachlor
Three-Mile Pond, 3MP-A	< 0.05	< 0.03	< 0.01	< 0.01	< 0.01	< 0.01	0.07	0.06	0.04	0.01	0.06
Three-Mile Pond, 3MP-B	< 0.05	< 0.03	< 0.01	< 0.01	< 0.01	< 0.01	0.07	0.06	0.04	0.01	0.06
Three-Mile Pond, 3MP-C	< 0.05	< 0.03	< 0.01	0.01	< 0.01	< 0.01	0.07	0.06	0.04	0.01	0.06
Mean	**< 0.05**	**< 0.03**	**< 0.01**		**< 0.01**	**< 0.01**	**0.07**	**0.06**	**0.04**	**0.01**	**0.06**
SD							**0.00**	**0.00**	**0.00**	**0.00**	**0.00**
Rising Pond, RP-A	0.44	0.08	< 0.01	< 0.01	< 0.01	< 0.01	0.51	0.20	< 0.02	< 0.01	0.33
Rising Pond, RP-B	0.41	0.07	< 0.01	< 0.01	< 0.01	< 0.01	0.52	0.19	< 0.02	0.09	0.37
Rising Pond, RP-C	0.42	0.08	< 0.01	< 0.01	< 0.01	< 0.01	0.46	0.19	< 0.02	0.10	0.48
Mean	**0.42**	**0.08**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**0.50**	**0.19**	**< 0.02**		**0.40**
SD	**0.01**	**0.00**					**0.04**	**0.01**			**0.08**
Woods Pond, WP-A	1.6	0.71	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.02	< 0.01	< 0.01
Woods Pond, WP-B	1.2	0.53	0.18	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.02	< 0.01	< 0.01
Woods Pond, WP-C	1.4	0.34	0.34	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.02	0.27	< 0.01
Mean	**1.4**	**0.53**		**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.02**		**< 0.01**
SD	**0.18**	**0.18**									
Deep Reach DRP-A	2.8	0.46	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	3.6	< 0.01	< 0.01
Deep Reach DRP-B	2.6	0.87	0.43	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	3.6	< 0.01	< 0.01
Deep Reach DRP-C	3.0	1.5	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	3.5	< 0.01	< 0.01
Mean	**2.8**	**0.94**		**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**3.6**	**< 0.01**	**< 0.01**
SD	**0.19**	**0.52**							**0.06**		

Table 6 (Cont.). Concentrations (ng/g) of organochlorine pesticides in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

Site / Sample ID	trans-Nonachlor	o,p'-DDE	o,p'-DDD	o,p'-DDT	p,p'-DDE	p,p'-DDD	p,p'-DDT	Endosulfan I	Endosulfan II	Endosufate	Mirex
Three-Mile Pond, 3MP-A	0.21	< 0.01	< 0.01	< 0.01	14	0.67	0.06	0.18	< 0.01	< 0.06	< 0.01
Three-Mile Pond, 3MP-B	0.22	< 0.01	< 0.01	< 0.01	15	0.68	0.06	0.18	< 0.01	< 0.06	< 0.01
Three-Mile Pond, 3MP-C	0.22	< 0.01	< 0.01	< 0.01	14	0.68	0.06	0.18	< 0.01	< 0.06	< 0.01
Mean	**0.22**	**< 0.01**	**< 0.01**	**< 0.01**	**14**	**0.67**	**0.06**	**0.18**	**< 0.01**	**< 0.06**	**< 0.01**
SD	**0.00**				**0.09**	**0.00**	**0.00**	**0.00**			
Rising Pond, RP-A	2.6	< 0.01	< 0.01	< 0.01	36	4.2	0.64	0.40	< 0.01	< 0.06	< 0.01
Rising Pond, RP-B	2.6	< 0.01	< 0.01	< 0.01	34	4.0	0.59	0.40	< 0.01	< 0.06	< 0.01
Rising Pond, RP-C	2.6	< 0.01	< 0.01	< 0.01	35	4.0	0.67	0.39	< 0.01	0.32	< 0.01
Mean	**2.6**	**< 0.01**	**< 0.01**	**< 0.01**	**35**	**4.1**	**0.63**	**0.40**	**< 0.01**		**< 0.01**
SD	**0.01**				**0.57**	**0.10**	**0.04**	**0.01**			
Woods Pond, WP-A	1.8	< 0.01	< 0.01	< 0.01	31	1.3	< 0.01	< 0.01	< 0.01	1.8	< 0.01
Woods Pond, WP-B	1.9	< 0.01	< 0.01	< 0.01	31	2.0	< 0.01	< 0.01	< 0.01	1.1	< 0.01
Woods Pond, WP-C	1.9	< 0.01	< 0.01	< 0.01	32	0.8	< 0.01	< 0.01	< 0.01	0.27	< 0.01
Mean	**1.9**	**< 0.01**	**< 0.01**	**< 0.01**	**31**	**1.4**	**< 0.01**	**< 0.01**	**< 0.01**	**1.1**	**< 0.01**
SD	**0.04**				**0.49**	**0.62**				**0.78**	
Deep Reach DRP-A	2.8	< 0.01	< 0.01	< 0.01	52	8.3	< 0.01	< 0.01	< 0.01	< 0.06	< 0.01
Deep Reach DRP-B	2.2	< 0.01	< 0.01	< 0.01	39	2.9	< 0.01	< 0.01	< 0.01	< 0.06	< 0.01
Deep Reach DRP-C	2.8	< 0.01	< 0.01	< 0.01	37	3.9	< 0.01	< 0.01	< 0.01	< 0.06	< 0.01
Mean	**2.6**	**< 0.01**	**< 0.01**	**< 0.01**	**42**	**5.0**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.06**	**< 0.01**
SD	**0.33**				**8.2**	**2.85**					

Table 7. Concentrations (ng/µL) of organochlorine pesticides in dosing solutions prepared from extracts of Housatonic River largemouth bass.

Site / Sample ID	HCB	PCA	alpha-BHC	Lindane	delta-BHC	Heptachlor	Heptachlor Epoxide	Oxy-chlordane	cis-Chlordane	trans-Chlordane	cis-Nonachlor
Three-Mile Pond, 3MP-A	<0.08	<0.05	<0.02	<0.02	<0.02	<0.02	0.10	0.09	0.06	0.02	0.09
Three-Mile Pond, 3MP-B	<0.08	<0.05	<0.02	<0.02	<0.02	<0.02	0.10	0.09	0.06	0.02	0.09
Three-Mile Pond, 3MP-C	<0.08	<0.05	<0.02	<0.02	<0.02	<0.02	0.10	0.10	0.06	0.02	0.09
Mean	**<0.08**	**<0.05**	**<0.02**	**<0.02**	**<0.02**	**<0.02**	**0.10**	**0.09**	**0.06**	**0.02**	**0.09**
SD								**0.00**			**0.00**
Rising Pond, RP-A	0.56	0.10	<0.01	<0.01	<0.01	<0.01	0.66	0.26	<0.03	<0.01	0.43
Rising Pond, RP-B	0.52	0.10	<0.01	<0.01	<0.01	<0.01	0.67	0.24	<0.03	0.12	0.48
Rising Pond, RP-C	0.53	0.10	<0.01	<0.01	<0.01	<0.01	0.58	0.25	<0.03	0.12	0.62
Mean	**0.54**	**0.10**	**<0.01**	**<0.01**	**<0.01**	**<0.01**	**0.64**	**0.25**	**<0.03**		**0.51**
SD	**0.02**	**0.00**					**0.05**	**0.01**			**0.10**
Woods Pond, WP-A	2.27	1.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.03	<0.01	<0.01
Woods Pond, WP-B	1.75	0.75	0.25	<0.01	<0.01	<0.01	<0.01	<0.01	<0.03	<0.01	<0.01
Woods Pond, WP-C	1.96	0.49	0.49	<0.01	<0.01	<0.01	<0.01	<0.01	<0.03	0.39	<0.01
Mean	**1.99**	**0.75**		**<0.01**	**<0.01**	**<0.01**	**<0.01**	**<0.01**			**<0.01**
SD	**0.26**	**0.26**									
Deep Reach DRP-A	3.60	0.60	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	4.6	<0.01	<0.01
Deep Reach DRP-B	3.39	1.13	0.56	<0.01	<0.01	<0.01	<0.01	<0.01	4.7	<0.01	<0.01
Deep Reach DRP-C	3.87	1.94	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	4.6	<0.01	<0.01
Mean	**3.62**	**0.00**		**<0.01**	**<0.01**	**<0.01**	**<0.01**	**<0.01**	**4.7**	**<0.01**	**<0.01**
SD	**0.24**	**0.00**							**0.1**		

Table 7 (Cont.). Concentrations (ng/µL) of organochlorine pesticides in dosing solutions prepared from extracts of Housatonic River largemouth bass.

Site / Sample ID	trans-Nonachlor	o,p'-DDE	o,p'-DDD	o,p'-DDT	p,p'-DDE	p,p'-DDD	p,p'-DDT	Endosulfan I	Endosulfan II	Endosufate	Mirex
Three-Mile Pond, 3MP-A	0.21	<0.02	<0.02	<0.02	22	1.0	0.09	0.27	<0.02	<0.09	<0.02
Three-Mile Pond, 3MP-B	0.22	<0.02	<0.02	<0.02	22	1.0	0.09	0.28	<0.02	<0.09	<0.02
Three-Mile Pond, 3MP-C	0.22	<0.02	<0.02	<0.02	22	1.0	0.09	0.28	<0.02	<0.09	<0.02
Mean	**0.22**	**<0.02**	**<0.02**	**<0.02**	**22**	**1.0**	**0.09**	**0.28**	**<0.02**	**<0.09**	**<0.02**
SD	**0.00**					**0.0**	**0.00**	**0.00**			
Rising Pond, RP-A	2.61	<0.01	<0.01	<0.01	46	5.4	0.81	0.51	<0.01	<0.08	<0.01
Rising Pond, RP-B	2.58	<0.01	<0.01	<0.01	44	5.2	0.76	0.52	<0.01	<0.08	<0.01
Rising Pond, RP-C	2.60	<0.01	<0.01	<0.01	45	5.2	0.86	0.49	<0.01	0.41	<0.01
Mean	**2.60**	**<0.01**	**<0.01**	**<0.01**	**45**	**5.2**	**0.81**	**0.51**	**<0.01**		**<0.01**
SD	**0.01**				**1**	**0.1**	**0.05**	**0.01**			
Woods Pond, WP-A	1.83	<0.01	<0.01	<0.01	44	1.8	<0.01	<0.01	<0.01	2.59	<0.01
Woods Pond, WP-B	1.90	<0.01	<0.01	<0.01	45	2.9	<0.01	<0.01	<0.01	1.55	<0.01
Woods Pond, WP-C	1.90	<0.01	<0.01	<0.01	45	1.2	<0.01	<0.01	<0.01	0.39	<0.01
Mean	**1.88**	**<0.01**	**<0.01**	**<0.01**	**45**	**1.9**	**<0.01**	**<0.01**	**<0.01**	**1.51**	**<0.01**
SD	**0.04**				**1**	**0.9**				**1.10**	
Deep Reach DRP-A	2.76	<0.01	<0.01	<0.01	67	10.8	<0.01	<0.01	<0.01	<0.08	<0.01
Deep Reach DRP-B	2.19	<0.01	<0.01	<0.01	50	3.8	<0.01	<0.01	<0.01	<0.08	<0.01
Deep Reach DRP-C	2.75	<0.01	<0.01	<0.01	48	5.1	<0.01	<0.01	<0.01	<0.08	<0.01
Mean	**2.57**	**<0.01**	**<0.01**	**<0.01**	**55**	**6.6**	**<0.01**	**<0.01**	**<0.01**	**<0.08**	**<0.01**
SD	**0.33**				**11**	**3.7**					

Table 8. Concentrations (pg/g) of non-*ortho*-chloro substituted polychlorinated biphenyls in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

	Non-*ortho*-chloro Polychlorinated Biphenyls			
	Tetra:		Penta:	
Site/Sample ID	3,4,4',5-TCB (81)	3,3',4,4'-TCB (77)	3,3',4,4',5-PeCB (126)	3,3',4,4',5,5'-HxCB (169)
Three-Mile Pond, 3MP-A	0.4	8.0	3.0	0.4
Three-Mile Pond, 3MP-B	0.5	7.7	3.0	0.5
Three-Mile Pond, 3MP-C	0.5	8.1	3.0	0.5
Mean	**0.5**	**7.9**	**3.0**	**0.5**
SD	**0.0**	**0.2**	**0.0**	**0.0**
Rising Pond, RP-A	36 IF	870	1,500	220
Rising Pond, RP-B	31 IF	830	1,500	210
Rising Pond, RP-C	43	730	1,400	210
Mean	**37**	**810**	**1,500**	**210**
SD	**5.8**	**72**	**58**	**5.9**
Woods Pond, WP-A	110 IF, LQ	1,600 LQ	2,300	320
Woods Pond, WP-B	62 IF, LQ	1,800	2,400	270 LQ
Woods Pond, WP-C	77 IF	1,600	2,400	310
Mean	**83**	**1,700**	**2,400**	**300**
SD	**24**	**120**	**58**	**26**
Deep Reach, DRP-A	210 IF, LQ	2,000	2,900	310 LQ
Deep Reach, DRP-B	215 IF, LQ	1,600 LQ	2,500 LQ	390
Deep Reach, DRP-C	156 IF	2,500	2,000 LQ	380 LQ
Mean	**194**	**2,000**	**2,500**	**360**
SD	**33**	**450**	**450**	**44**

IF = Low Values of PCBs #81 and #77 that were adjusted for interference from partially co-eluting PCBs #87 and #110, respectively.

LQ = Less than Method Quantification Limit due to Incomplete Ion Cluster or Inaccurate Ion Ratio (Outside +/- 15% Tolerances).

Table 9. Concentrations (pg/µL) of non-*ortho*-chloro substituted polychlorinated biphenyls in dosing solutions prepared from extracts of Housatonic River largemouth bass.

	Non-*ortho*-chloro Polychlorinated Biphenyls			
	Tetra:		Penta:	
Site/Sample ID	3,4,4',5-TCB (81)	3,3',4,4'-TCB (77)	3,3',4,4',5-PeCB (126)	3,3',4,4',5,5'-HxCB (169)
Three-Mile Pond, 3MP-A	0.7	12	4.6	0.7
Three-Mile Pond, 3MP-B	0.7	12	4.6	0.7
Three-Mile Pond, 3MP-C	0.8	12	4.5	0.8
Mean	**0.7**	**12**	**4.6**	**0.7**
SD	**0.0**	**0.3**	**0.1**	**0.0**
Rising Pond, RP-A	47 IF	1100	1900	280
Rising Pond, RP-B	40 IF	1100	1900	270
Rising Pond, RP-C	55	930	1800	270
Mean	**47**	**1000**	**1900**	**270**
SD	**7.4**	**93**	**74**	**7.5**
Woods Pond, WP-A	156 IF, LQ	2300 LQ	3300	450
Woods Pond, WP-B	88 IF, LQ	2600	3400	380 LQ
Woods Pond, WP-C	109 IF	2300	3400	440
Mean	**118**	**2400**	**3400**	**430**
SD	**35**	**160**	**82**	**38**
Deep Reach, DRP-A	270 IF, LQ	2600	3800	400 LQ
Deep Reach, DRP-B	280 IF, LQ	2100 LQ	3200 LQ	500
Deep Reach, DRP-C	200 IF	3200	2600 LQ	490 LQ
Mean	**250**	**2600**	**3200**	**470**
SD	**42**	**590**	**590**	**57**

IF = Low Values of PCBs #81 and #77 that were adjusted for interference from partially co-eluting PCBs #87 and #110, respectively.

LQ = Less than Method Quantification Limit due to Incomplete Ion Cluster or Inaccurate Ion Ratio (Outside +/- 15% Tolerances).

Table 10. Concentrations (pg/g) of chlorinated dibenzo-p-dioxins and dibenzofurans in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

		Three-Mile Pond						
	TEF Value*	3MP-A	3MP-B	3MP-C	Mean	SD	Mean TEQ	TEQ (% Total)
Dioxins								
2,3,7,8-Tetrachloro	1.0	0.5	0.4 LQ	0.4 LQ	0.4	0.1	0.4	33
1,2,3,7,8-Pentachloro	1.0	0.4	0.4 LQ	0.4	0.4	0.0	0.4	31
1,2,3,4,7,8-Hexachloro	0.5	0.2 LQ	0.1 LQ	0.2	0.1	0.0	0.1	6
1,2,3,6,7,8-Hexachloro	0.01	0.3 LQ	0.3 LQ	0.4 LQ	0.3	0.1	0.0	0
1,2,3,7,8,9-Hexachloro	0.01	0.2 LQ	0.2	0.2 ND	0.2	0.0	0.0	0
1,2,3,4,6,7,8-Heptachloro	0.001	0.8	0.9 LQ	0.8 LQ	0.8	0.1	0.0	0
Octachloro	0.0001	6.0	6.0 LQ	5.3	5.8	0.4	0.0	0
Furans								
2,3,7,8-Tetrachloro	0.05	2.4	2.5	2.4	2.4	0.0	0.1	10
1,2,3,7,8-Pentachloro	0.05	0.2 LQ	0.1 LQ	0.1 LQ	0.2	0.0	0.0	1
2,3,4,7,8-Pentachloro	0.5	0.4 LQ	0.3 LQ	0.3	0.3	0.0	0.2	14
1,2,3,4,7,8-Hexachloro	0.1	0.1 LQ	0.1 LQ	0.0 LQ	0.1	0.0	0.0	1
1,2,3,6,7,8-Hexachloro	0.1	0.1 LQ	0.1 LQ	0.0 LQ	0.1	0.0	0.0	0
1,2,3,7,8,9-Hexachloro	0.1	0.1	0.2 LQ	0.1 LQ	0.1	0.1	0.0	1
2,3,4,6,7,8-Hexachloro	0.1	0.1LQ	0.1 LQ	0.1 ND	0.1	0.0	0.0	1
1,2,3,4,6,7,8-Heptachloro	0.01	0.2 LQ	0.2 LQ	0.2 LQ	0.2	0.0	0.0	0
1,2,3,4,7,8,9-Heptachloro	0.01	0.2 ND	0.2 ND	0.2 ND	0.2	0.0	0.0	0
Octachloro	0.0001	0.2	0.2 ND	0.2 LQ	0.2	0.0	0.0	0
Total D/F TEQs		1.3	1.1	1.1	1.2			

*Dioxin Toxicity Equivalent Factors from van den Berg et al. (1998).
Values not annotated have a percent of variation of about 10%.
LQ Less than method quantification limit due to incomplete ion cluster or ion ratio outside of +/- 15% tolerances.
ND Not detected at specified detection limit.

Table 10 (Cont.). Concentrations (pg/g) of chlorinated dibenzo-p-dioxins and dibenzofurans in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

		Rising Pond						
	TEF Value*	RP-A	RP-B	RP-C	Mean	SD	Mean TEQ	TEQ (% Total)
Dioxins								
2,3,7,8-Tetrachloro	1.0	3.1	3.1	2.9	3.0	0.1	3.0	12
1,2,3,7,8-Pentachloro	1.0	1.5	1.5	1.5	1.5	0.0	1.5	6
1,2,3,4,7,8-Hexachloro	0.5	0.5 LQ	0.2	0.2	0.3	0.2	0.1	1
1,2,3,6,7,8-Hexachloro	0.01	1.3 LQ	1.1	0.9	1.1	0.2	0.0	0
1,2,3,7,8,9-Hexachloro	0.01	0.3 LQ	0.1 ND	0.1 ND	0.2	0.1	0.0	0
1,2,3,4,6,7,8-Heptachloro	0.001	0.6 LQ	0.6 LQ	0.5	0.6	0.1	0.0	0
Octachloro	0.0001	2.5	1.4	1.5	1.8	0.6	0.0	0
Furans								
2,3,7,8-Tetrachloro	0.05	9.1	9.3	9.3	9.2	0.1	0.5	2
1,2,3,7,8-Pentachloro	0.05	14	14	14	14	0.0	0.7	3
2,3,4,7,8-Pentachloro	0.5	35	36	36	36	0.6	17.9	73
1,2,3,4,7,8-Hexachloro	0.1	1.8	1.8	1.8	1.8	0.0	0.2	1
1,2,3,6,7,8-Hexachloro	0.1	2.1	1.7	1.8	1.9	0.2	0.2	1
1,2,3,7,8,9-Hexachloro	0.1	0.5	0.1 LQ	0.1 LQ	0.2	0.2	0.0	0
2,3,4,6,7,8-Hexachloro	0.1	3.2	3.1	2.9	3.1	0.1	0.3	1
1,2,3,4,6,7,8-Heptachloro	0.01	2.0	1.7	1.8	1.8	0.2	0.0	0
1,2,3,4,7,8,9-Heptachloro	0.01	0.9	0.6	0.8 LQ	0.8	0.1	0.0	0
Octachloro	0.0001	1.9	0.8	1.1 LQ	1.3	0.5	0.0	0
Total D/F TEQs		24	25	25	25			

*Dioxin Toxicity Equivalent Factors from van den Berg et al. (1998).
Values not annotated have a percent of variation of about 10%.
LQ Less than method quantification limit due to incomplete ion cluster or ion ratio outside of +/- 15% tolerances.
ND Not detected at specified detection limit.

Table 10 (Cont.). Concentrations (pg/g) of chlorinated dibenzo-p-dioxins and dibenzofurans in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

		Woods Pond						
	TEF Value*	WP-A	WP-B	WP-C**	Mean	SD	Mean TEQ	TEQ (% Total)
Dioxins								
2,3,7,8-Tetrachloro	1.0	2.1 LQ	1.9	NA	2.0	0.1	2.0	16
1,2,3,7,8-Pentachloro	1.0	0.6	1.0 LQ	NA	0.8	0.3	0.8	6
1,2,3,4,7,8-Hexachloro	0.5	0.2 LQ	0.2 LQ	NA	0.2	0.0	0.1	1
1,2,3,6,7,8-Hexachloro	0.01	0.4	0.3	NA	0.4	0.1	0.0	0
1,2,3,7,8,9-Hexachloro	0.01	0.1 LQ	0.1 ND	NA	0.1	0.0	0.0	0
1,2,3,4,6,7,8-Heptachloro	0.001	0.3	0.4	NA	0.3	0.1	0.0	0
Octachloro	0.0001	0.7	0.9 LQ	NA	0.8	0.1	0.0	0
Furans								
2,3,7,8-Tetrachloro	0.05	8.2	8.4	NA	8.3	0.1	0.4	3
1,2,3,7,8-Pentachloro	0.05	12	12	NA	12	0.1	0.6	5
2,3,4,7,8-Pentachloro	0.5	16	16	NA	16	0.4	8.1	65
1,2,3,4,7,8-Hexachloro	0.1	1.4 LQ	1.5	NA	1.5	0.1	0.1	1
1,2,3,6,7,8-Hexachloro	0.1	1.2	1.2 LQ	NA	1.2	0.0	0.1	1
1,2,3,7,8,9-Hexachloro	0.1	0.1	0.1 LQ	NA	0.1	0.0	0.0	0
2,3,4,6,7,8-Hexachloro	0.1	2.0 LQ	2.6 LQ	NA	2.3	0.4	0.2	2
1,2,3,4,6,7,8-Heptachloro	0.01	1.1 LQ	1.2	NA	1.2	0.1	0.0	0
1,2,3,4,7,8,9-Heptachloro	0.01	0.4 LQ	0.4 LQ	NA	0.4	0.0	0.0	0
Octachloro	0.0001	0.2 LQ	0.2 LQ	NA	0.2	0.0	0.0	0
Total D/F TEQs		12	13	NA	12			

*Dioxin Toxicity Equivalent Factors from van den Berg et al. (1998).
** Sample processing error – No recovery of ^{13}C-surrogates.
Values not annotated have a percent of variation of about 10%.
LQ Less than method quantification limit due to incomplete ion cluster or ion ratio outside of +/- 15% tolerances.
ND Not detected at specified detection limit.
NA Data not available

Table 10 (Cont.). Concentrations (pg/g) of chlorinated dibenzo-p-dioxins and dibenzofurans in dosing solutions, normalized to the initial mass of carcasses of Housatonic River largemouth bass.

		Deep Reach						
	TEF Value*	DRP-A	DRP-B	DRP-C	Mean	SD	Mean TEQ	TEQ (% Total)
Dioxins								
2,3,7,8-Tetrachloro	1.0	1.6	1.7	1.7	1.7	0.1	1.7	12
1,2,3,7,8-Pentachloro	1.0	1.0	1.1	1.2 LQ	1.1	0.1	1.1	8
1,2,3,4,7,8-Hexachloro	0.5	0.2 LQ	0.1 LQ	0.1	0.3	0.0	0.1	1
1,2,3,6,7,8-Hexachloro	0.01	0.5 LQ	0.5	0.5 LQ	1.1	0.0	0.0	0
1,2,3,7,8,9-Hexachloro	0.01	0.1	0.1 ND	0.1 LQ	0.1	0.0	0.0	0
1,2,3,4,6,7,8-Heptachloro	0.001	0.4 LQ	0.4	0.4 LQ	0.4	0.0	0.0	0
Octachloro	0.0001	1.9	3.9	1.4	2.4	1.3	0.0	0
Furans								
2,3,7,8-Tetrachloro	0.05	11	11	11	11	0.4	0.5	4
1,2,3,7,8-Pentachloro	0.05	13	2.5 LQ	13	9.8	6.3	0.5	3
2,3,4,7,8-Pentachloro	0.5	19	18	19	19	0.7	9.4	67
1,2,3,4,7,8-Hexachloro	0.1	2.0	1.8	2.1	2.0	0.2	0.2	1
1,2,3,6,7,8-Hexachloro	0.1	1.5 LQ	1.4	1.6	1.5	0.1	0.2	1
1,2,3,7,8,9-Hexachloro	0.1	0.1	0.1 ND	0.1 LQ	0.1	0.0	0.0	0
2,3,4,6,7,8-Hexachloro	0.1	3.8	2.6	3.6 LQ	3.3	0.6	0.3	2
1,2,3,4,6,7,8-Heptachloro	0.01	2.1 LQ	1.7	1.9	1.9	0.2	0.0	0
1,2,3,4,7,8,9-Heptachloro	0.01	0.5	0.2 LQ	0.6 LQ	0.5	0.2	0.0	0
Octachloro	0.0001	0.4 LQ	0.2 LQ	0.4 LQ	0.3	0.1	0.0	0
Total D/F TEQs		14	13	15	14			

*Dioxin Toxicity Equivalent Factors from van den Berg et al. (1998).
Values not annotated have a percent of variation of about 10%.
LQ Less than method quantification limit due to incomplete ion cluster or ion ratio outside of +/- 15% tolerances.
ND Not detected at specified detection limit.

Table 11. Concentrations (pg/µL) of chlorinated dibenzo-p-dioxins and dibenzofurans in dosing solutions prepared from extracts of Housatonic River largemouth bass.

		Three-Mile Pond						
	TEF Value*	3MP-A	3MP-B	3MP-C	Mean	SD	Mean TEQ	TEQ (% Total)
Dioxins								
2,3,7,8-Tetrachloro	1.0	0.7	0.6 LQ	0.5 LQ	0.6	0.1	0.6	33
1,2,3,7,8-Pentachloro	1.0	0.6	0.5 LQ	0.6	0.6	0.0	0.6	32
1,2,3,4,7,8-Hexachloro	0.5	0.3 LQ	0.1 LQ	0.3	0.2	0.1	0.1	6
1,2,3,6,7,8-Hexachloro	0.01	0.5 LQ	0.4 LQ	0.5 LQ	0.5	0.1	0.0	0
1,2,3,7,8,9-Hexachloro	0.01	0.3 LQ	0.3	0.2 ND	0.3	0.0	0.0	0
1,2,3,4,6,7,8-Heptachloro	0.001	1.1	1.4 LQ	1.2 LQ	1.2	0.1	0.0	0
Octachloro	0.0001	9.2	9.0 LQ	8.1	8.8	0.6	0.0	0
Furans								
2,3,7,8-Tetrachloro	0.05	3.7	3.7	3.7	3.7	0.0	0.2	10
1,2,3,7,8-Pentachloro	0.05	0.3 LQ	0.2 LQ	0.2 LQ	0.2	0.1	0.0	1
2,3,4,7,8-Pentachloro	0.5	0.6 LQ	0.5 LQ	0.5	0.5	0.1	0.3	14
1,2,3,4,7,8-Hexachloro	0.1	0.1 LQ	0.1 LQ	0.1 LQ	0.1	0.0	0.0	1
1,2,3,6,7,8-Hexachloro	0.1	0.1 LQ	0.1 LQ	0.1 LQ	0.1	0.0	0.0	0
1,2,3,7,8,9-Hexachloro	0.1	0.1	0.3 LQ	0.1 LQ	0.2	0.1	0.0	1
2,3,4,6,7,8-Hexachloro	0.1	0.1 LQ	0.1 LQ	0.1 ND	0.1	0.0	0.0	1
1,2,3,4,6,7,8-Heptachloro	0.01	0.3 LQ	0.3 LQ	0.3 LQ	0.3	0.0	0.0	0
1,2,3,4,7,8,9-Heptachloro	0.01	0.3 ND	0.3 ND	0.3 ND	0.3	0.0	0.0	0
Octachloro	0.0001	0.3	0.3 ND	0.3 LQ	0.3	0.0	0.0	0
Total D/F TEQs		2.0	1.7	1.7	1.8			

*Dioxin Toxicity Equivalent Factors from van den Berg et al. (1998).
Values not annotated have a percent of variation of about 10%.
LQ Less than method quantification limit due to incomplete ion cluster or ion ratio outside of +/- 15% tolerances.
ND Not detected at specified detection limit.

Table 11 (Cont.). Concentrations (pg/µL) of chlorinated dibenzo-p-dioxins and dibenzofurans in dosing solutions prepared from extracts of Housatonic River largemouth bass.

		Rising Pond						
	TEF Value*	RP-A	RP-B	RP-C	Mean	SD	Mean TEQ	TEQ (% Total)
Dioxins								
2,3,7,8-Tetrachloro	1.0	4.0	4.0	3.7	3.9	0.2	3.9	12
1,2,3,7,8-Pentachloro	1.0	1.9	2.0	2.0	2.0	0.0	2.0	6
1,2,3,4,7,8-Hexachloro	0.5	0.6 LQ	0.3	0.3	0.4	0.2	0.2	1
1,2,3,6,7,8-Hexachloro	0.01	1.7 LQ	1.4	1.2	1.4	0.2	0.0	0
1,2,3,7,8,9-Hexachloro	0.01	0.4 LQ	0.1 ND	0.1 ND	0.2	0.2	0.0	0
1,2,3,4,6,7,8-Heptachloro	0.001	0.8 LQ	0.7 LQ	0.6	0.7	0.1	0.0	0
Octachloro	0.0001	3.2	1.8	1.9	2.3	0.8	0.0	0
Furans								
2,3,7,8-Tetrachloro	0.05	12	12	12	12	0.2	0.6	2
1,2,3,7,8-Pentachloro	0.05	18	18	18	18	0.1	0.9	3
2,3,4,7,8-Pentachloro	0.5	45	46	47	46	0.7	23	73
1,2,3,4,7,8-Hexachloro	0.1	2.3	2.3	2.3	2.3	0.0	0.2	1
1,2,3,6,7,8-Hexachloro	0.1	2.7	2.2	2.3	2.4	0.2	0.2	1
1,2,3,7,8,9-Hexachloro	0.1	0.6	0.1 LQ	0.1 LQ	0.3	0.3	0.0	0
2,3,4,6,7,8-Hexachloro	0.1	4.0	4.0	3.7	3.9	0.2	0.4	1
1,2,3,4,6,7,8-Heptachloro	0.01	2.6	2.2	2.2	2.3	0.2	0.0	0
1,2,3,4,7,8,9-Heptachloro	0.01	1.1	0.8	1.0 LQ	1.0	0.2	0.0	0
Octachloro	0.0001	2.4	1.1	1.4 LQ	1.6	0.7	0.0	0
Total D/F TEQs		31	32	32	32			

*Dioxin Toxicity Equivalent Factors from van den Berg et al. (1998).
Values not annotated have a percent of variation of about 10%.
LQ Less than method quantification limit due to incomplete ion cluster or ion ratio outside of +/- 15% tolerances.
ND Not detected at specified detection limit.

Table 11 (Cont.). Concentrations (pg/µL) of chlorinated dibenzo-p-dioxins and dibenzofurans in dosing solutions prepared from extracts of Housatonic River largemouth bass.

		Woods Pond						
	TEF Value*	WP-A	WP-B	WP-C**	Mean	SD	Mean TEQ	TEQ (% Total)
Dioxins								
2,3,7,8-Tetrachloro	1.0	3.0 LQ	2.8	NA	2.9	0.1	2.9	16
1,2,3,7,8-Pentachloro	1.0	0.8	1.4 LQ	NA	1.1	0.4	1.1	6
1,2,3,4,7,8-Hexachloro	0.5	0.2 LQ	0.2 LQ	NA	0.2	0.0	0.1	1
1,2,3,6,7,8-Hexachloro	0.01	0.6	0.4	NA	0.5	0.1	0.0	0
1,2,3,7,8,9-Hexachloro	0.01	0.1 LQ	0.1 ND	NA	0.1	0.0	0.0	0
1,2,3,4,6,7,8-Heptachloro	0.001	0.4	0.5	NA	0.4	0.1	0.0	0
Octachloro	0.0001	1.0	1.3 LQ	NA	1.1	0.2	0.0	0
Furans								
2,3,7,8-Tetrachloro	0.05	12	12	NA	12	0.2	0.6	3
1,2,3,7,8-Pentachloro	0.05	17	18	NA	17	0.2	0.9	5
2,3,4,7,8-Pentachloro	0.5	22	23	NA	23	0.5	12	65
1,2,3,4,7,8-Hexachloro	0.1	2.0 LQ	2.2	NA	2.1	0.1	0.2	1
1,2,3,6,7,8-Hexachloro	0.1	1.7	1.7 LQ	NA	1.7	0.0	0.2	1
1,2,3,7,8,9-Hexachloro	0.1	0.1	0.1 LQ	NA	0.1	0.0	0.0	0
2,3,4,6,7,8-Hexachloro	0.1	2.9 LQ	3.7 LQ	NA	3.3	0.6	0.3	2
1,2,3,4,6,7,8-Heptachloro	0.01	1.6 LQ	1.7	NA	1.6	0.1	0.0	0
1,2,3,4,7,8,9-Heptachloro	0.01	0.6 LQ	0.6 LQ	NA	0.6	0.0	0.0	0
Octachloro	0.0001	0.3 LQ	0.3 LQ	NA	0.3	0.0	0.0	0
Total D/F TEQs		17	18	NA	17			

*Dioxin Toxicity Equivalent Factors from van den Berg et al. (1998).
** Sample processing error – No recovery of ^{13}C-surrogates.
Values not annotated have a percent of variation of about 10%.
LQ Less than method quantification limit due to incomplete ion cluster or ion ratio outside of +/- 15% tolerances.
ND Not detected at specified detection limit.
NA Data not available

Table 11 (Cont.). Concentrations (pg/µL) of chlorinated dibenzo-p-dioxins and dibenzofurans in dosing solutions prepared from extracts of Housatonic River largemouth bass.

		Deep Reach						
	TEF Value*	DRP-A	DRP-B	DRP-C	Mean	SD	Mean TEQ	TEQ (% Total)
Dioxins								
2,3,7,8-Tetrachloro	1.0	2.1	2.3	2.2	2.2	0.1	2.2	12
1,2,3,7,8-Pentachloro	1.0	1.3	1.4	1.5 LQ	1.4	0.1	1.4	8
1,2,3,4,7,8-Hexachloro	0.5	0.2 LQ	0.2 LQ	0.1	0.2	0.0	0.1	1
1,2,3,6,7,8-Hexachloro	0.01	0.7 LQ	0.6	0.7 LQ	0.7	0.0	0.0	0
1,2,3,7,8,9-Hexachloro	0.01	0.1	0.1 ND	0.1 LQ	0.1	0.0	0.0	0
1,2,3,4,6,7,8-Heptachloro	0.001	0.6 LQ	0.5	0.5 LQ	0.6	0.0	0.0	0
Octachloro	0.0001	2.4	5.1	1.8	3.1	1.7	0.0	0
Furans								
2,3,7,8-Tetrachloro	0.05	14	14	15	14	0.5	0.7	4
1,2,3,7,8-Pentachloro	0.05	17	3.2 LQ	18	13	8.2	0.6	3
2,3,4,7,8-Pentachloro	0.5	25	23	25	24	1.0	12	67
1,2,3,4,7,8-Hexachloro	0.1	2.6	2.3	2.8	2.6	0.2	0.3	1
1,2,3,6,7,8-Hexachloro	0.1	2.0 LQ	1.8	2.0	2.0	0.1	0.2	1
1,2,3,7,8,9-Hexachloro	0.1	0.1	0.1 ND	0.1 LQ	0.1	0.0	0.0	0
2,3,4,6,7,8-Hexachloro	0.1	4.9	3.3	4.6 LQ	4.3	0.8	0.4	2
1,2,3,4,6,7,8-Heptachloro	0.01	2.7 LQ	2.2	2.5	2.5	0.3	0.0	0
1,2,3,4,7,8,9-Heptachloro	0.01	0.6	0.3 LQ	0.8 LQ	0.6	0.3	0.0	0
Octachloro	0.0001	0.5 LQ	0.3 LQ	0.5 LQ	0.4	0.1	0.0	0
Total D/F TEQs		18	17	20	18			

*Dioxin Toxicity Equivalent Factors from van den Berg et al. (1998)
Values not annotated have a percent of variation of about 10%.
LQ Less than method quantification limit due to incomplete ion cluster or ion ratio outside of +/- 15% tolerances.
ND Not detected at specified detection limit.

Table 12. TEQs (pg/g) and Total PCBs (ng/g) in dosing solutions prepared from extracts of largemouth bass from the Housatonic River and a reference site. Concentrations are reported as normalized to the initial mass of carcass tissue (mass/g) and normalized to the final volume of the extract (mass/uL).

Location	Sample ID	non-o-PCB TEQs# (pg/g)	% nPCB	Total PCDD TEQs# (pg/g)	% PCDD	Total PCDF TEQs# (pg/g)	% PCDF	Dosing Solution			
								Total TEQs# (pg/g)	Total TEQ pg/µL	Total PCB (ng/g)	Total PCB (ng/µL)
Three-Mile Pond	3MP-A DOSE	0.016	1	0.9	71	0.4	27	1.3	2.0	30	46
	3MP-B DOSE	0.016	1	0.8	69	0.3	30	1.1	1.7	30	46
	3MP-C DOSE	0.016	1	0.8	72	0.3	27	1.2	1.8	30	46
							Mean	**1.2**	**1.8**	**30**	**46**
							SD	**0.1**	**0.2**	**0.02**	**0.0**
Rising Pond	RP-A DOSE	7.8	24	4.9	15	20	61	32.3	41.3	18,000	22,000
	RP-B DOSE	7.8	24	4.8	15	20	61	32.4	41.4	19,000	22,000
	RP-C DOSE	7.3	23	4.5	14	20	63	31.8	40.8	18,000	22,000
							Mean	**32.2**	**41.2**	**18,000**	**22,000**
							SD	**0.3**	**0.4**	**580**	**0.0**
Woods Pond	WP-A DOSE	12	50	2.1	9.0	9.4	41	23.2	32.9	37,000	53,000
	WP-B DOSE	12	51	1.9	8.0	9.8	41	24.0	34.1	37,000	53,000
	WP-C DOSE	*	---	*	---	*	---	*	*	*	*
							Mean	**23.6**	**33.5**	**37,000**	**53,000**
							SD	**0.6**	**0.8**	**0.0**	**0.0**
Deep Reach	DRP-A DOSE	15	51	2.7	9.0	11	39	29.1	37.8	61,000	79,000
	DRP-B DOSE	13	49	3.0	12	10	39	26.0	33.8	61,000	79,000
	DRP-C DOSE	10	47	0.0	0.0	12	53	22.0	28.5	63,000	81,000
							Mean	**25.5**	**33.2**	**62,000**	**79,661**
							SD	**3.6**	**4.6**	**1,200**	**1,155**

***No measured value. # Using fish TEFs from van den Berg et. al (1998).**

5.3 Responses of Largemouth Bass to Standards and Extracts

Survival and Growth:

Survival in negative controls was generally low, but ranged from 0 to 80%. Mortality in the controls at this stage was believed to be related to the failure of largemouth bass to successfully survive the transition from endogenous to exogenous feeding. Despite our attempts to provide food of the correct size, quantity, and at a suitable frequency, average survival for largemouth bass at swim-up was 51 ± 20 % for the uninjected fish and 53 ± 21 % for the triolein injected fish. At fifteen days post swim-up, survival had decreased to 22 ± 13 % and 22+ ± 16% for uninjected and triolein injected fish, respectively. Mortality for Three-mile Pond, Rising Pond, and Deep Reach fish was below 50% at the highest dose and therefore, no LD50s were estimated for these extracts. Only PCB 126 and 2,3,7,8-TCDD at concentrations of 1200 ng/g egg (6000 pg TEQ/g egg) and 6.0 ng/g egg (6000 pg TEQ/g egg), respectively, were observed to cause significantly higher mortality at this lifestage as compared to the negative controls (Table 13). The LD_{50} estimated for Woods Pond extract Trial 3 was 178 pg TEQ/g egg, a factor of nearly 30 times lower than for PCB 126 (5217 pg TEQ/g egg) and 2,3,7,8-TCDD (4572 pg TEQ/g egg; Table 14). Slopes of the linear portions of the dose-response curves (Figure 5) for these chemicals were similar (Table 14). Neither weight nor length was affected by exposure to the extracts, PCB 126 or 2,3,7,8-TCDD (Tables 15-20).

Table 13. Percent survival at swim-up of largemouth bass exposed in ovo to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Values are means of 3 replicate samples of 10 eggs each (standard deviations in parentheses). Trials are temporally distinct dose-response experiments.

	% Survival															
	Three-mile Pond		Rising Pond		Woods Pond			Deep Reach	PCB 126					2,3,7,8-TCDD		
Dose	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 3	Trial 1	Trial 1	Trial 2	Trial 3	Trial 4	Trial 5	Trial 1	Trial 2	Trial 3
Uninjected	80 (10)	60 (10)	50 (30)	40 (20)	70 (20)	50 (30)	50 (40)	60 (20)	50 (40)	70 (20)	0 (0)	50 (20)	60 (20)	10 (10)	50 (10)	60 (20)
Triolein	60 (10)	60 (10)	70 (10)	30 (20)	80 (10)	40 (10)	60 (30)	60 (20)	70 (30)	80 (10)	20 (10)	40 (10)	---	30 (10)	70 (20)	20 (10)
1	---	60 (10)	50 (10)	20 (20)	60 (20)	20 (10)	60 (20)	60 (10)	80 (10)	50 (40)	10 (20)	40 (40)	30 (30)	10 (10)	70 (20)	20 (10)
2	---	50 (10)	60 (10)	20 (10)	60 (30)	60 (20)	40 (10)	60 (20)	70 (20)	70 (20)	10 (10)	50 (10)	50 (10)	10 (10)	40 (40)	40 (20)
3	---	40 (0)	40 (20)	10 (10)	80 (20)	40 (20)	50 (20)	60 (30)	70 (10)	70 (20)	20 (10)	50 (10)	30 (0)	20 (10)	70 (10)	40 (10)
4	---	40 (20)	40 (10)	10 (10)	80 (10)	50 (20)	40 (30)	60 (10)	---	90 (10)	20 (10)	40 (20)	10 (10)	20 (10)	80 (10)	30 (20)
5	60 (10)	60 (10)	70 (20)	10 (10)	70 (10)	30 (20)	30 (30)	60 (10)	30* (40)	30* (0)	0 (0)	20 (20)	30 (10)	20 (10)	20* (20)	40 (20)

*** Significantly different from Triolein controls.**
--- High or complete mortality due to fungal infection.

Figure 5. Dose-response curves for lethality of largemouth bass at swim-up from exposure to increasing concentrations of Woods Pond extracts, PCB 126, or TCDD. Concentrations are represented by TEQs. The inverted triangle shows the mean mortality of Woods Pond offspring in the Phase I study at measured egg concentrations.

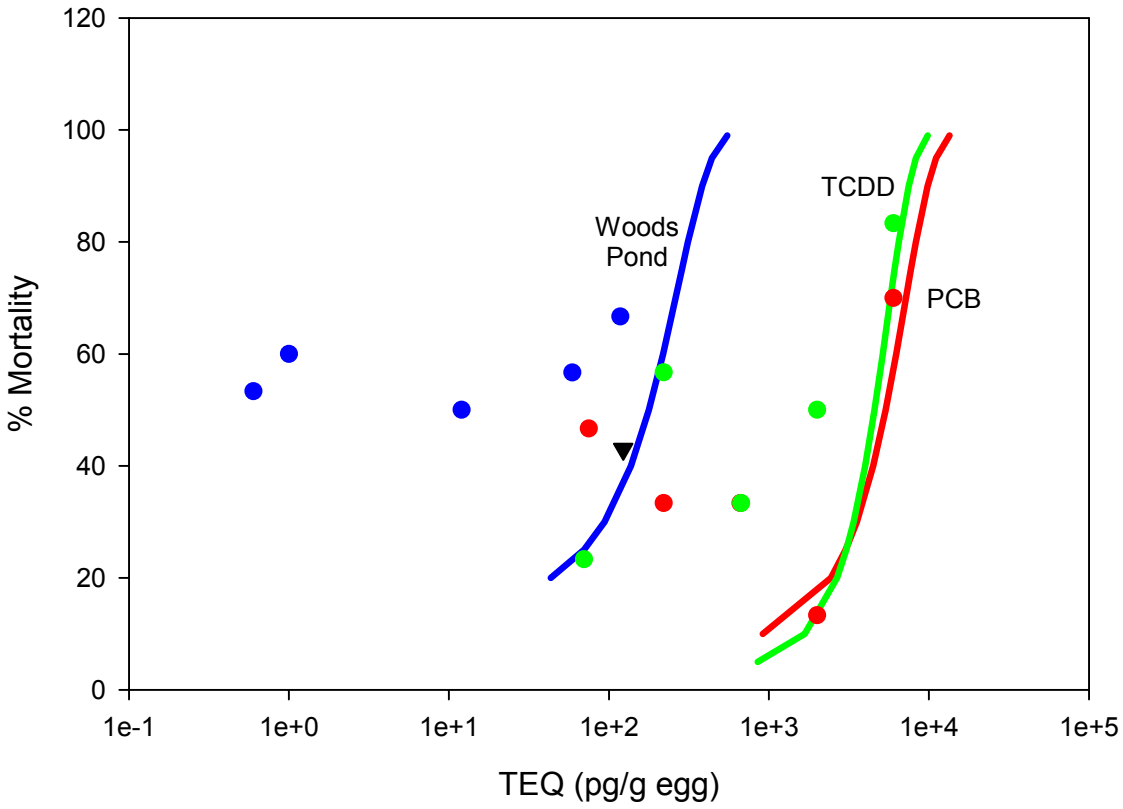

Table 14. LD50 values for largemouth bass at swim-up following in ovo injection with Woods Pond extracts, PCB 126, and TCDD. Values are expressed as pg TEQs per g egg.

	LD50	95% CI	Slope
Woods Pond (Trial 3)	178	68-287	101
PCB 126 (Trial 2)	5217	3610-6824	127
2,3,7,8-TCDD (Trial 2)	4572	3761-5382	154

Table 15. Mean lengths and weights (± standard deviations) of largemouth bass exposed to Three-Mile Pond extracts. Trials are temporally distinct dose-response experiments.

	Trial 1		
Dose	**n**	**Length (cm)**	**Weight (g)**
Uninjected	11	1.1 ± 0.9	0.0144 ± 0.0043
Triolein	8	1.1 ± 0.1	0.0173 ± 0.0018
1	11	1.1 ± 0.1	0.0168 ± 0.0033
2	12	1.3 ± 0.1	0.0156 ± 0.0031
3	12	1.1 ± 0.1	0.0154 ± 0.0040
4	10	1.1 ± 0.1	0.0146 ± 0.0027
5	14	1.1 ± 0.1	0.0133 ± 0.0036

No significant differences between treatments and controls were detected.

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Table 16. Mean lengths and weights (± standard deviations) of largemouth bass exposed to Rising Pond extracts. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2		
Dose	**N**	**Length (cm)**	**Weight (g)**	**N**	**Length (cm)**	**Weight (g)**
Uninjected	13	1.1 ± 0.1	0.0135 ± 0.0053	9	1.1 ± 0.1	0.0099 ± 0.0026
Triolein	9	1.1 ± 0.1	0.0146 ± 0.0027	8	1.0 ± 0.2	0.0091 ± 0.0042
1	12	1.1 ± 0.1	0.0160 ± 0.0036	7	1.1 ± 0.1	0.01 ± 0.0023
2	13	1.1 ± 0.1	0.0152 ± 0.0033	5	1.0 ± 0.1	0.0095 ± 0.0039
3	8	1.2 ± 0.1	0.0174 ± 0.0038	1	1.2	0.0148
4	6	1.1 ± 0.1	0.0143 ± 0.0049	3	1.1 ± 0.1	0.0126 ± 0.0045
5	15	1.1 ± 0.1	0.0146 ± 0.0027	8	1.0 ± 0.2	0.0091 ± 0.0042

No significant differences between treatments and controls were detected

Table 17. Mean lengths and weights (± standard deviations) of largemouth bass exposed to Woods Pond extracts. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2			Trial 3		
Dose	**N**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	5	0.90 ± 0.05	0.0085 ± 0.0017	6	1.4 ± 0.1	0.0732 ± 0.0030	8	1.0 ± 0.1	0.0107 ± 0.0034
Triolein	12	0.85 ± 0.1	0.0066 ± 0.0026	7	1.4 ± 0.1	0.0476 ± 0.0023	8	1.1 ± 0.1	0.0104 ± 0.0036
1	5	0.90 ± 0.1	0.0080 ± 0.0028	4	1.4 ± 0.1	0.1080 ± 0.0055	6	1.1 ± 0.1	0.0126 ± 0.0039
2	3	0.90 ± 0.1	0.0076 ± 0.0028	10	1.2 ± 0.1	0.1313 ± 0.0061	8	1.1 ± 0.1	0.0124 ± 0.0037
3	5	0.85 ± 0.1	0.0067 ± 0.0024	9	1.2 ± 0.1	0.1732 ± 0.0054	7	1.0 ± 0.1	0.0094 ± 0.0028
4	5	1.00 ± 0.05	0.0097 ± 0.0008	9	1.3 ± 0.1	0.0866 ± 0.0037	8	1.0 ± 0.1	0.0098 ± 0.0038
5	0	ND	ND	7	1.3 ± 0.1	0.1035 ± 0.0034	4	1.0 ± 0.05	0.0090 ± 0.0014

No significant differences between treatments and controls were detected.
ND - Eggs were not injected at this dose.

Table 18. Mean lengths and weights (± standard deviations) of largemouth bass exposed to Deep Reach extracts. Trials are temporally distinct dose-response experiments.

	Trial 1		
Dose	**n**	**Length (cm)**	**Weight (g)**
Uninjected	15	1.1 ± 0.1	0.0124 ± 0.0034
Triolein	12	1.1 ± 0.1	0.0140 ± 0.0032
1	13	1.1 ± 0.1	0.0140 ± 0.0032
2	9	1.1 ± 0.1	0.0155 ± 0.0021
3	10	1.1 ± 0.1	0.0159 ± 0.0030
4	15	1.1 ± 0.1	0.0130 ± 0.0028
5	15	1.0 ± 0.1	0.0125 ± 0.0036

No significant differences between treatments and controls were detected.

Table 19. Mean lengths and weights (± standard deviations) of largemouth bass exposed to PCB 126. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2			Trial 3		
Dose	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	3	1.0 ± 0.1	0.0087 ± 0.0027	6	1.0 ± 0.0	0.0085 ± 0.0015	3	1.0 ± 0.05	0.0133 ± 0.0025
Triolein	5	1.0 ± 0.1	0.0078 ± 0.0016	3	1.0 ± 0.0	0.0095 ± 0.0021	0	ND	ND
1	4	1.0 ± 0.1	0.0089 ± 0.0016	3	1.0 ± 0.0	0.0080 ± 0.0018	1	1.3	0.0221
2	1	0.90	0.01	5	1.0 ± 0.1	0.0103 ± 0.0022	2	1.2 ± 0.05	0.0187 ± 0.0023
3	6	0.95 ± 0.05	0.0075 ± 0.0006	6	1.0 ± 0.1	0.0088 ± 0.0017	3	1.2 ± 0.1	0.0194 ± 0.0041
4	3	1.0 ± 0.1	0.0077 ± 0.0020	6	1.0 ± 0.1	0.0082 ± 0.0021	3	1.1 ± 0.1	0.0127 ± 0.0029
5	0	ND	ND	1	1.0	0.0122	0	ND	ND

No significant differences between treatments and controls were detected.
ND - Eggs were not injected at this dose.

Table 19 (Cont.). Mean lengths and weights (± standard deviations) of largemouth bass exposed to PCB 126. Trials are temporally distinct dose-response experiments.

	Trial 4			Trial 5		
Dose	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	4	1.1 ± 0.1	0.0136 ± 0.0021	5	1.1 ± 0.2	0.0169 ± 0.0052
Triolein	3	1.2 ± 0.1	0.0189 ± 0.0051	2	1.1 ± 0.2	0.0133 ± 0.0074
1	1	1.3	0.0221	5	1.1 ± 0.2	0.0145 ± 0.0077
2	6	1.1 ± 0.1	0.0160 ± 0.0031	5	1.2 ± 0.1	0.0167 ± 0.0030
3	4	1.2 ± 0.1	0.0191 ± 0.0034	8	1.1 ± 0.1	0.0152 ± 0.0033
4	3	1.1 ± 0.1	0.0127 ± 0.0029	6	1.0 ± 0.1	0.0137 ± 0.0036
5	0	ND	ND	0	ND	ND

No significant differences between treatments and controls were detected.
ND - Eggs were not injected at this dose.

Table 20. Mean lengths and weights (± standard deviations) of largemouth bass exposed to TCDD. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2			Trial 3		
Dose	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	6	0.90 ± 0.10	0.0082 ± 0.0025	4	1.0 ± 0.20	0.0119 ± 0.0045	7	1.3 ± 0.1	0.0229 ± 0.0037
Triolein	2	0.95 ± 0.05	0.0091 ± 0.0015	1	1.0	0.0098	17	1.2 ± 0.1	0.0159 ± 0.0042
1	2	0.90 ± 0.15	0.0074 ± 0.0053	5	0.95 ± 0.10	0.0101 ± 0.0036	6	1.3 ± 0.1	0.0270 ± 0.0046
2	1	1.1	0.0150	3	1.0 ± 0.05	0.0104 ± 0.0009	10	1.3 ± 0.1	0.0222 ± 0.0063
3	1	1.0	0.0125	6	1.0 ± 0.05	0.0117 ± 0.0020	10	1.2 ± 0.1	0.0208 ± 0.0066
4	2	0.90 ± 0.15	0.0084 ± 0.0047	8	0.95 ± 0.2	0.0089 ± 0.0036	8	1.4 ± 0.1	0.0221 ± 0.0039
5	3	0.80 ± 0.10	0.0060 ± 0.0030	1	1.1	0.0140	11	1.3 ± 0.1	0.0215 ± 0.0041

No significant differences between treatments and controls were detected.

Pathology:

Hatch- Overall, pathologies in largemouth bass at this developmental stage were low and those observed in extract- or chemical-exposed bass were generally similar in nature and extent to negative controls (Tables 21 and 24). No clear dose-related patterns in pathologies were evident in exposed largemouth bass embryos at this stage. Pathologies classified as "Other" included occurrence of a circular-shaped yolk sac and pale coloration.

Swim-up- All largemouth bass at the swim-up stage in these studies generally experienced a high incidence of uninflated or partially inflated swim bladders and spinal deformities. Largemouth bass at swim-up injected *in ovo* with PCB and TCDD also showed signs of craniofacial deformities, difficulty inflating swim bladder, and pericardial and peritoneal edema (Tables 22 and 25). The abnormalities in these treatment groups generally followed an increasing dose-response pattern. These same abnormalities were also observed in Housatonic River extract-exposed fish, and at much reduced rate in Three-Mile Pond fish at the highest dose (Figure 6). Swim-up fry exposed to Three-Mile Pond and Rising Pond extracts had deformities of the eye. Swim-up fry exposed to Deep Reach extract uniquely had fin deformities and seven individuals with the swim bladder external to the body. This latter condition was also observed in one triolein-exposed fish. Observations included in the "Other" category included brain hemorrhages, and small size.

15d post swim-up- Fifteen days after swim-up, the pathologies most frequently seen in largemouth bass fry exposed to PCB 126 were craniofacial, and eye deformities, peritoneal edema, hemorrhaging, and problems associated with inflation and position of the swim bladder (Tables 23 and 26). TCDD-exposed fish experienced craniofacial, eye, and spinal deformities, as well as problems associated with inflation of the swim bladder. These fish also uniquely showed signs of delayed development. Housatonic River extract-exposed fish uniquely displayed fin deformities (Figure 7). The only pathologies observed in Three-Mile pond fish were craniofacial anomalies at the highest dose tested. Rising Pond fish experienced peritoneal edema, hemorrhaging, and uninflated swim bladders (Figure 8). Woods Pond extract-exposed

fish had craniofacial deformities at similar levels to those observed in PCB-exposed fish and peritoneal edema. The Woods Pond extract-dosed fish also had one individual with an external swim bladder, a condition also observed in two PCB-treated fish (Figures 9 and 10). In addition to fin deformities, fish treated with the Deep Reach extract experienced problems inflating their swim bladders. Pathological observations made and encompassed in the "Other" category for the 15-d post swim-up largemouth bass included spherical shape to yolk, opaque swim bladder, slightly protruding intestine, air bubble at vent, and cerebral edema.

The percentage of fish with one or more pathological anomaly at swim-up tended to increase with increasing dose of Housatonic River extract, PCB, or TCDD (Figures 11-16). Fish exposed to PCB 126 were the most affected followed by TCDD and the Woods Pond extract. Based on the cumulative pathological effects measured at this stage of development, the geometric mean of the no observed effect level (NOEL; 59 pg TEQ/g egg) and lowest observed effect level (LOEL; 18 pg TEQ/g egg) for Woods Pond was estimated to be 83 pg TEQ/g egg. No NOEL was established for PCB 126 in this study during which we measured an LOEL of 75 pg TEQ/g egg). Dose-response curves were also developed for the effect of Woods Pond and PCB 126 on the combination of mortality and pathologies (Figure 17). The estimated ED50 from these combined dose-response curves was 90 pg TEQ/g egg for the Woods Pond extract, approximately 40 times lower than the ED50s for PCB 126 (3750 pg TEQ/g egg; Table 27). ED50s could not be estimated for Three-mile Pond, Rising Pond, or Deep Reach extracts, because effects at the highest doses were less than 50%.

Table 21. Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth bass at hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 2,118.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Craniofacial Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
			Eye Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	14
1	0	0	16	0	0	15
2	0	0	0	0	13	16
3	0	0	14	0	0	0
4	0	0	14	0	0	0
5	0	0	0	0	0	15
			Vertebral/Spinal Deformity			
Uninjected	44	19	0	91	133	56
Triolein	49	49	14	43	121	71
1	125	22	48	0	221	45
2	43	21	153	53	184	63
3	0	27	56	45	91	16
4	105	44	85	136	121	121
5	63	61	80	43	103	108
			Hemorrhage			
Uninjected	0	0	13	0	24	0
Triolein	24	49	0	0	0	0
1	42	0	63	0	52	61
2	0	0	0	0	0	31
3	0	0	0	0	30	48
4	53	0	0	0	30	0
5	0	0	13	0	34	62

Table 21 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth bass at hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 2,118.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Pericardial Edema			
Uninjected	22	38	0	48	24	19
Triolein	49	49	14	48	0	14
1	42	22	48	0	52	45
2	87	21	68	136	66	63
3	0	0	28	0	15	48
4	105	0	56	87	91	103
5	21	0	67	0	92	77
			Peritoneal Edema			
Uninjected	44	0	0	0	0	19
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	31
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
			Yolk Sac Edema			
Uninjected	22	0	0	0	0	19
Triolein	0	0	0	48	0	14
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	15	17
5	0	0	0	0	0	0
			Delayed Development			
Uninjected	0	0	13	0	0	19
Triolein	49	0	0	0	0	29
1	0	22	63	0	26	0
2	0	21	17	0	13	31
3	0	0	14	0	15	0
4	53	0	14	48	0	0
5	0	0	27	0	23	15

Table 21 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth bass at hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 2,118.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Other			
Uninjected	0	0	0	53	12	19
Triolein	73	49	14	45	0	14
1	42	22	48	0	0	30
2	43	21	153	0	13	31
3	67	0	28	95	15	32
4	158	0	42	105	15	34
5	0	0	40	0	23	15

Table 22. Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth bass at swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 1,942.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Craniofacial Deformity			
Uninjected	0	0	17	0	11	0
Triolein	0	0	17	0	14	0
1	0	34	22	0	38	70
2	0	0	52	0	13	48
3	0	0	17	0	0	19
4	0	80	52	0	14	93
5	190	0	0	0	876	220
			Eye Deformity			
Uninjected	0	0	0	0	0	27
Triolein	50	0	0	0	0	0
1	48	0	43	0	0	0
2	0	0	0	0	13	0
3	0	0	17	0	0	0
4	0	0	0	0	0	0
5	95	0	0	0	11	0
			Fin Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	48	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
			Vertebral/Spinal Deformity			
Uninjected	0	0	0	0	114	81
Triolein	50	0	51	59	125	132
1	48	0	43	0	125	0
2	0	0	86	50	128	48
3	0	0	102	48	67	74
4	77	0	52	50	82	93
5	95	0	19	56	22	60

Table 22 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth bass at swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 1,942.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			External Swim Bladder			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	19
1	0	0	0	0	0	0
2	0	0	0	100	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	278	0	0
			Uninflated Swim Bladder			
Uninjected	59	421	400	0	466	135
Triolein	500	455	356	118	389	604
1	95	483	500	59	500	279
2	105	550	345	150	526	524
3	83	583	407	48	520	444
4	154	800	483	100	466	465
5	238	571	547	0	798	780
			Partially-Inflated Swim Bladder			
Uninjected	294	0	33	111	0	135
Triolein	0	91	17	0	14	19
1	190	69	22	118	13	23
2	53	0	0	0	0	24
3	83	42	51	0	13	222
4	0	0	52	0	14	163
5	95	0	94	0	0	40
			Hemorrhage			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	13	0
2	0	0	0	0	0	0
3	0	125	0	0	0	37
4	0	0	0	0	0	0
5	0	0	0	0	0	0

Table 22 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth at swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 1,942.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Pericardial Edema			
Uninjected	0	0	17	0	0	0
Triolein	50	0	0	0	0	19
1	0	0	43	0	25	23
2	0	0	34	0	0	0
3	0	0	0	0	0	0
4	0	40	34	0	0	47
5	48	0	19	0	652	340
			Peritoneal Edema			
Uninjected	0	0	17	0	0	0
Triolein	50	0	0	0	0	19
1	0	34	22	0	38	0
2	0	50	69	0	0	24
3	0	0	0	48	27	0
4	0	0	0	0	123	0
5	48	0	0	0	944	340
			Larval Weakness			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	23
2	0	0	0	0	0	0
3	0	0	0	0	0	19
4	0	0	0	0	0	0
5	0	0	0	0	0	0
			Delayed Development			
Uninjected	0	0	0	0	0	0
Triolein	50	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	13	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	11	0

Table 22 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth bass at swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 1,942.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Other			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	22	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	19
4	0	0	0	0	0	0
5	0	0	0	0	0	0

Table 23. Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth bass at 15d post swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 520.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Craniofacial Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	45	0	0	0
3	0	0	59	0	0	0
4	0	0	45	0	176	0
5	45	0	91	0	0	133
			Eye Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	77
2	0	0	0	0	59	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
			Fin Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	53	0	0	0	0
2	0	0	91	111	0	0
3	0	0	176	0	0	0
4	0	0	45	0	0	0
5	0	0	273	0	0	0
			Vertebral/Spinal Deformity			
Uninjected	0	0	0	0	67	59
Triolein	0	0	37	0	0	0
1	0	0	67	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	42	0
4	0	0	0	0	59	111
5	0	0	0	0	0	133

Table 23 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth bass at 15d post swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 520.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			External Swim Bladder			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	67	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	59	0
5	0	0	0	0	0	0
			Uninflated Swim Bladder			
Uninjected	0	0	0	0	0	0
Triolein	0	59	37	0	0	0
1	0	0	0	0	0	77
2	0	0	0	0	59	0
3	0	0	0	0	0	0
4	0	111	0	0	0	0
5	0	0	0	67	0	0
			Partially-Inflated Swim Bladder			
Uninjected	0	0	0	0	0	59
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	59	0
3	0	0	0	0	42	0
4	0	0	0	0	118	111
5	0	0	0	0	0	0
			Hemorrhage			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	53	0	0	0	0
2	0	0	0	0	59	0
3	0	0	0	0	0	0
4	0	0	0	0	59	0
5	0	0	0	0	1000	0

Table 23 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in largemouth bass at 15d post swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 520.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Peritoneal Edema			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	56	0	0	0	0
3	0	0	0	0	0	0
4	0	111	45	0	59	0
5	0	0	0	0	0	0
			Delayed Development			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	56
5	0	0	0	0	0	0
			Other			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	77	0
2	0	56	0	0	0	0
3	0	0	294	0	0	0
4	0	0	45	0	0	0
5	45	0	0	0	0	0

Table 24. Estimated occurrence of gross pathologies (per 1000 individuals) at hatch in largemouth bass exposed to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials and all doses combined.

	Treatment						
Observation	**Three-Mile Pond**	**Rising Pond**	**Woods Pond**	**Deep Reach**	**PCB 126**	**2,3,7,8-TCDD**	**Control**
Craniofacial Deformity	0	0	0	0	0	0	0
Eye Deformity	0	0	9	0	3	10	2
Fin Deformity	0	0	0	0	0	0	0
Vertebral/Spinal Deformity	70	33	82	56	145	70	59
External Swim Bladder	0	0	0	0	0	0	0
Uninflated Swim Bladder	0	0	0	0	0	0	0
Partially-Inflated Swim Bladder	0	0	0	0	0	0	0
Tubular Heart	0	0	0	0	0	0	3
Hemorrhage	16	0	15	0	30	41	9
Pericardial Edema	47	10	53	47	65	67	22
Peritoneal Edema	0	0	0	0	0	6	5
Yolk Sac Edema	0	0	0	0	3	3	6
Larval Weakness	0	0	0	0	0	0	0
Delayed Development	8	10	26	9	16	10	9
Other	47	10	59	37	13	29	17

Table 25. Estimated occurrence of gross pathologies (per 1000 individuals) at swim-up in largemouth bass exposed to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials and all doses combined.

	Treatment						
Observation	**Three-Mile Pond**	**Rising Pond**	**Woods Pond**	**Deep Reach**	**PCB 126**	**2.3.7.8-TCDD**	**Control**
Craniofacial Deformity	47	21	29	0	210	91	8
Eye Deformity	35	0	11	0	5	0	4
Fin Deformity	0	0	0	10	0	0	0
Vertebral/Spinal Deformity	47	0	62	42	84	56	66
External Swim Bladder	0	0	0	73	0	0	2
Uninflated Swim Bladder	140	589	453	73	570	504	381
Partially-Inflated Swim Bladder	93	21	44	21	8	99	39
Tubular Heart	0	0	0	0	0	0	0
Hemorrhage	0	21	0	0	3	9	0
Pericardial Edema	12	7	26	0	152	86	6
Peritoneal Edema	12	21	18	10	248	78	6
Yolk Sac Edema	0	0	0	0	0	0	0
Larval Weakness	0	0	0	0	0	9	0
Delayed Development	0	0	0	0	5	0	2
Other	0	0	4	0	0	4	0

Table 26. Estimated occurrence of gross pathologies (per 1000 individuals) at 15d post swim-up in largemouth bass exposed to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials and all doses combined.

	Treatment						
Observation	**Three-Mile Pond**	**Rising Pond**	**Woods Pond**	**Deep Reach**	**PCB 126**	**2,3,7,8-TCDD**	**Control**
Craniofacial Deformity	15	0	46	0	42	26	0
Eye Deformity	0	0	0	0	14	13	0
Fin Deformity	0	14	103	16	0	0	0
Vertebral/Spinal Deformity	0	0	11	0	28	51	14
External Swim Bladder	0	0	11	0	14	0	0
Uninflated Swim Bladder	0	14	0	16	14	13	9
Partially-Inflated Swim Bladder	0	0	0	0	56	26	5
Tubular Heart	0	0	0	0	0	0	0
Hemorrhage	0	14	0	0	42	0	0
Pericardial Edema	0	0	0	0	0	0	0
Peritoneal Edema	0	28	11	0	14	0	0
Yolk Sac Edema	0	0	0	0	0	0	0
Larval Weakness	0	0	0	0	0	0	0
Delayed Development	0	0	0	0	0	13	0
Other	14	14	69	0	14	0	0

Figure 6. Largemouth bass exhibiting craniofacial deformity, peritoneal edema, and missing eyes at hatch following in ovo exposure to Woods Pond extracts.

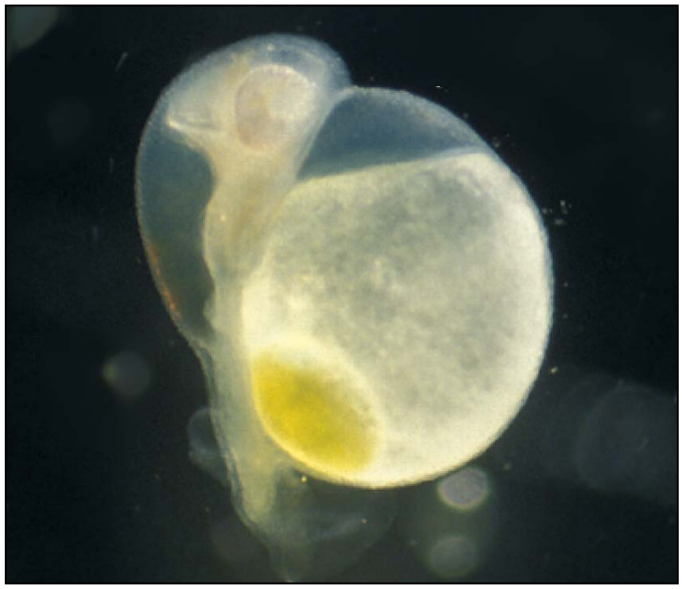


Figure 7. Largemouth bass exhibiting anal fin deformity at 15d post swim-up following in ovo exposure to Woods Pond extracts.

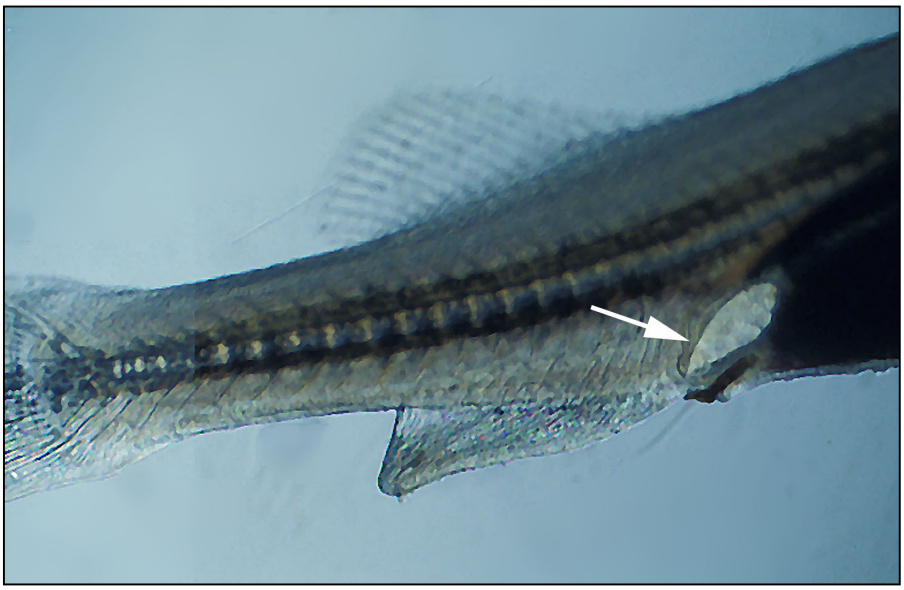


Figure 8. Largemouth bass (top exhibiting underinflated swim bladder at 15d post swim-up following in ovo exposure (bottom fish is normal).

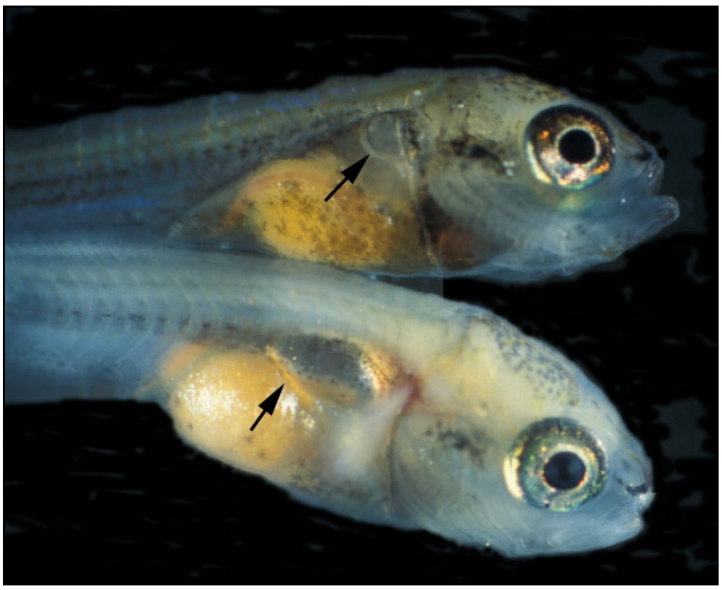


Figure 9. Largemouth bass exhibiting partially external swim bladder at 15d post swim-up following in ovo exposure to PCB 126.

Figure 10. Histological slide of partially external swim-bladder (arrow) in largemouth bass at 15d post swim-up following in ovo exposure to Woods Pond extracts (125X magnification).

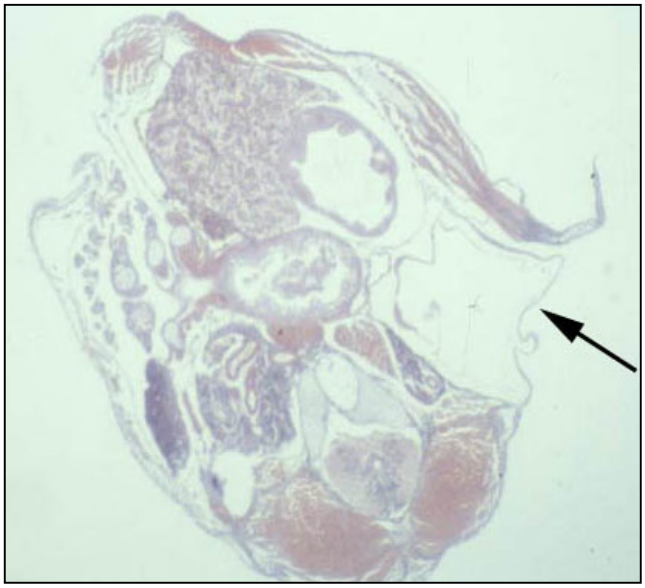


Figure 11. Effects of in ovo exposure to increasing doses of Three-Mile Pond extracts on largemouth bass at swim-up. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

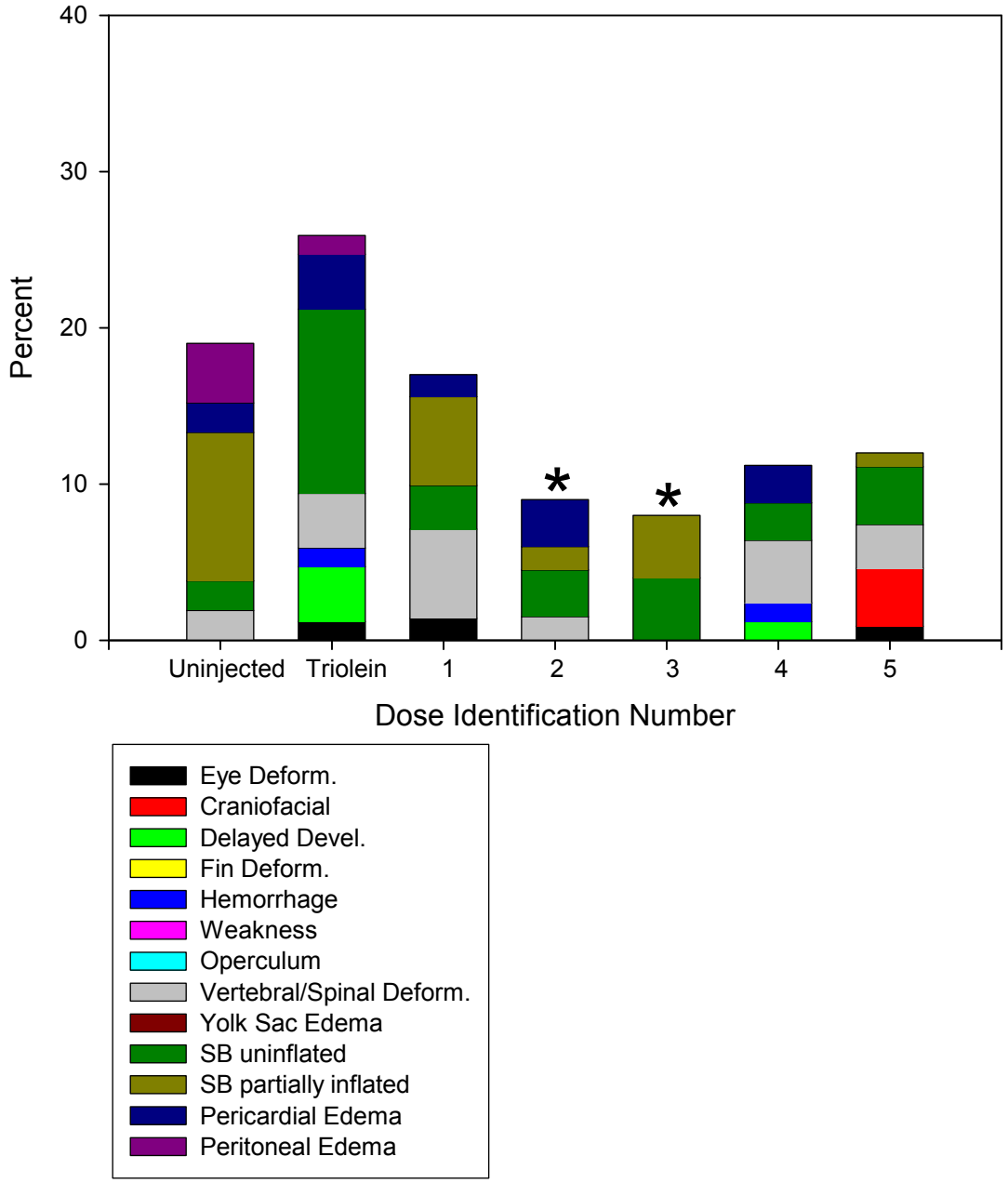


Figure 12. Effects of in ovo exposure to increasing doses of Rising Pond extracts on largemouth bass at swim-up. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

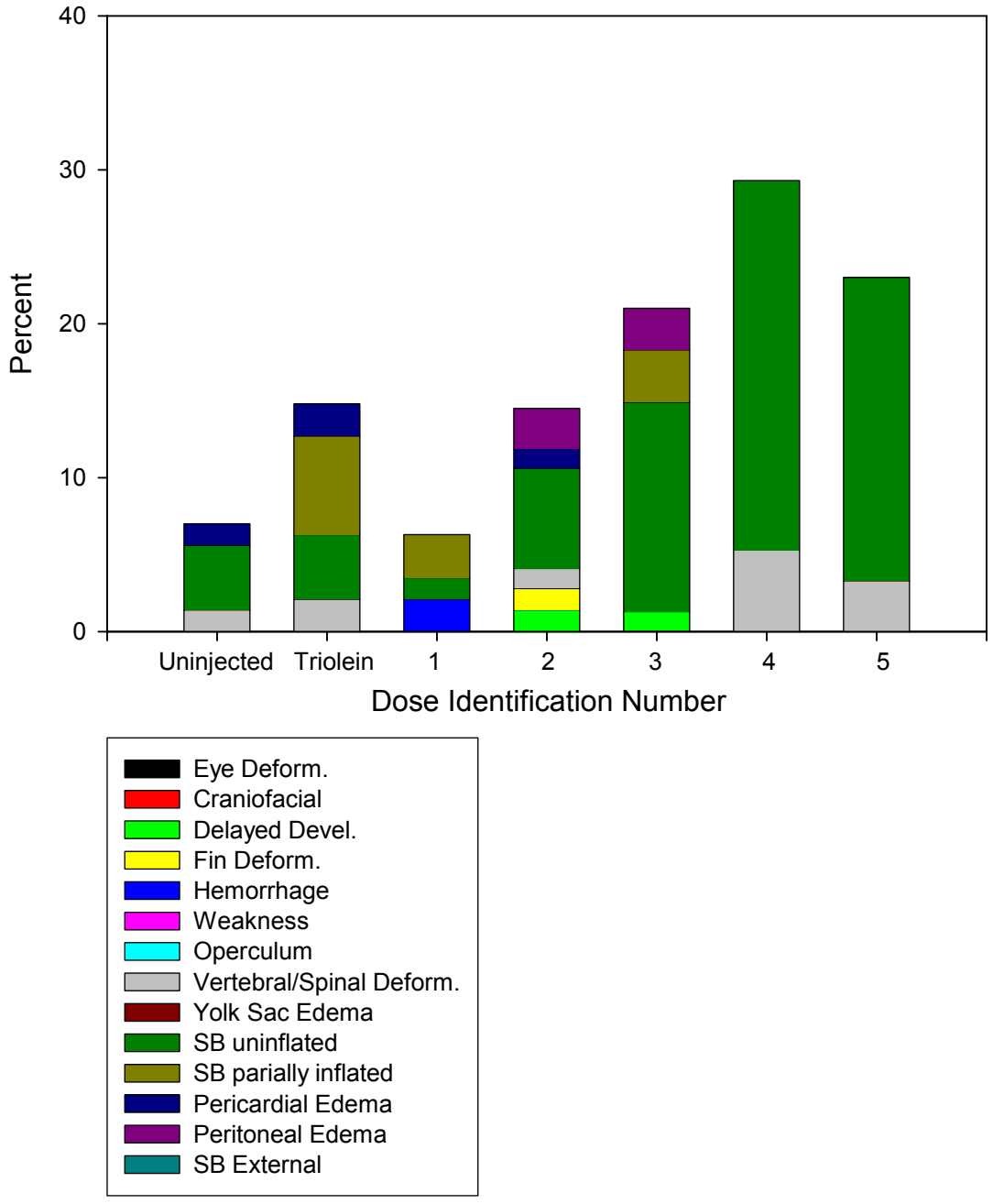


Figure 13. Effects of in ovo exposure to increasing doses of Woods Pond extracts on largemouth bass at swim-up. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

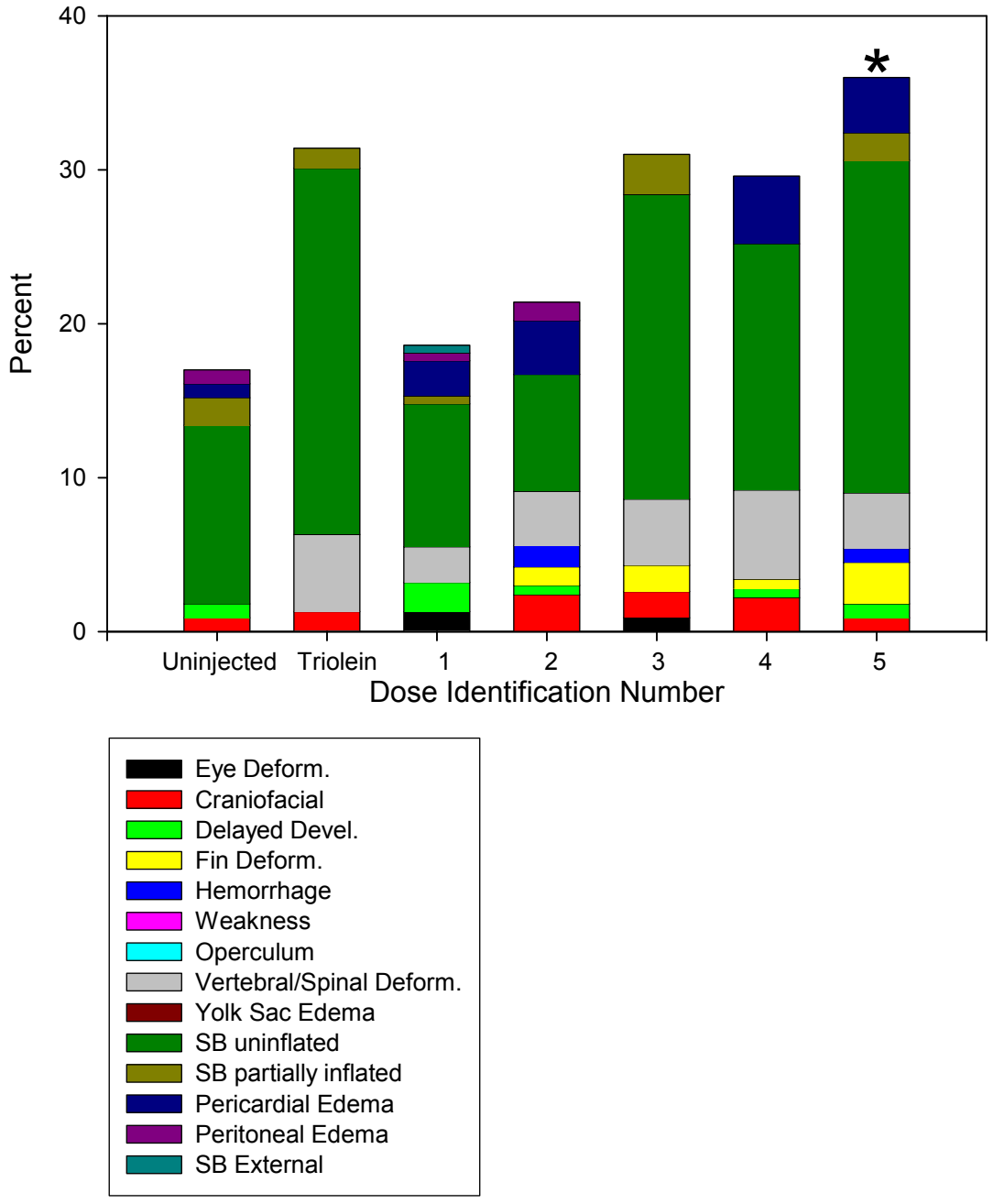


Figure 14. Effects of in ovo exposure to increasing doses of Deep Reach extracts on largemouth bass at swim-up. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

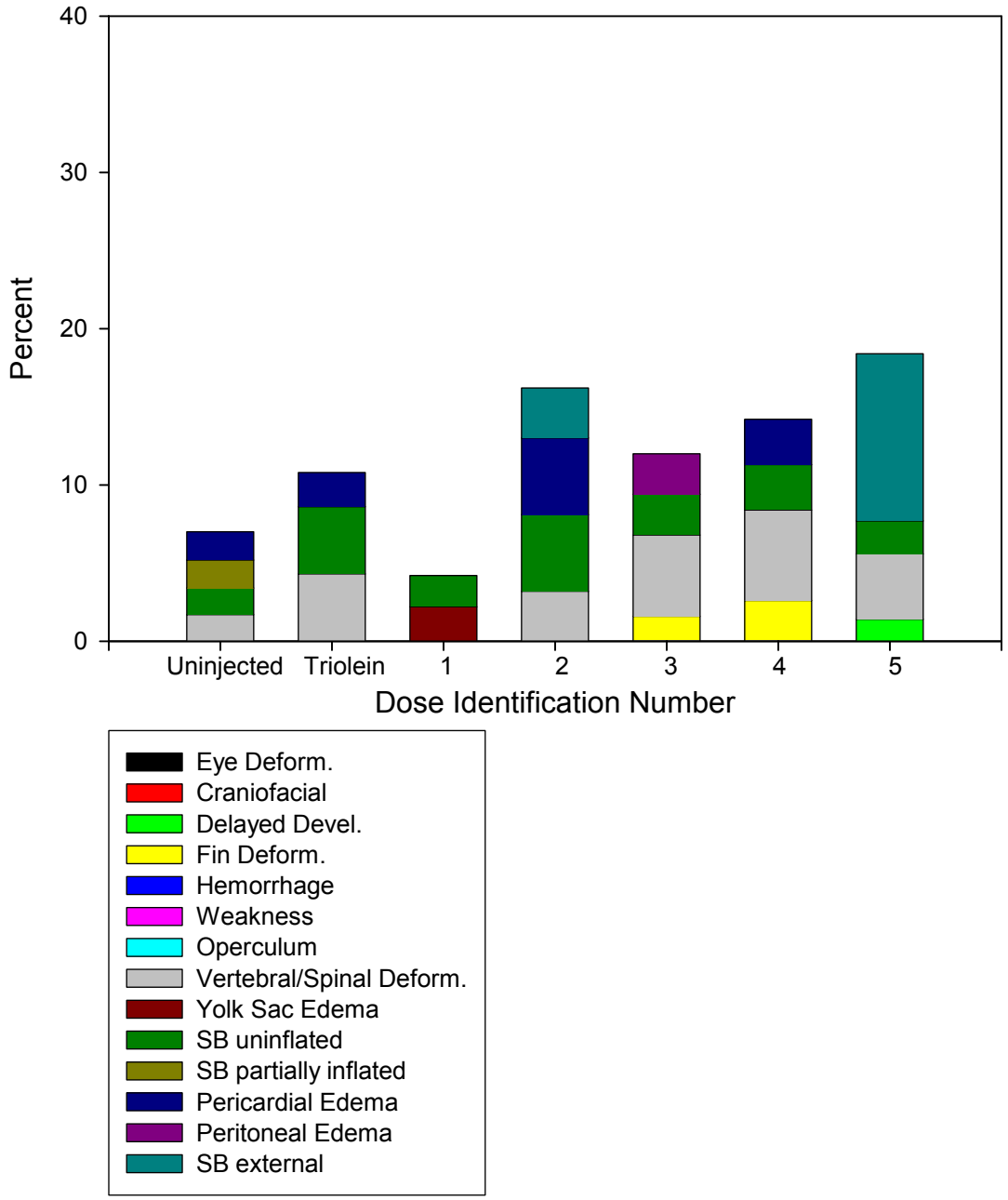


Figure 15. Effects of in ovo exposure to increasing doses of PCB 126 on largemouth bass at swim-up. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

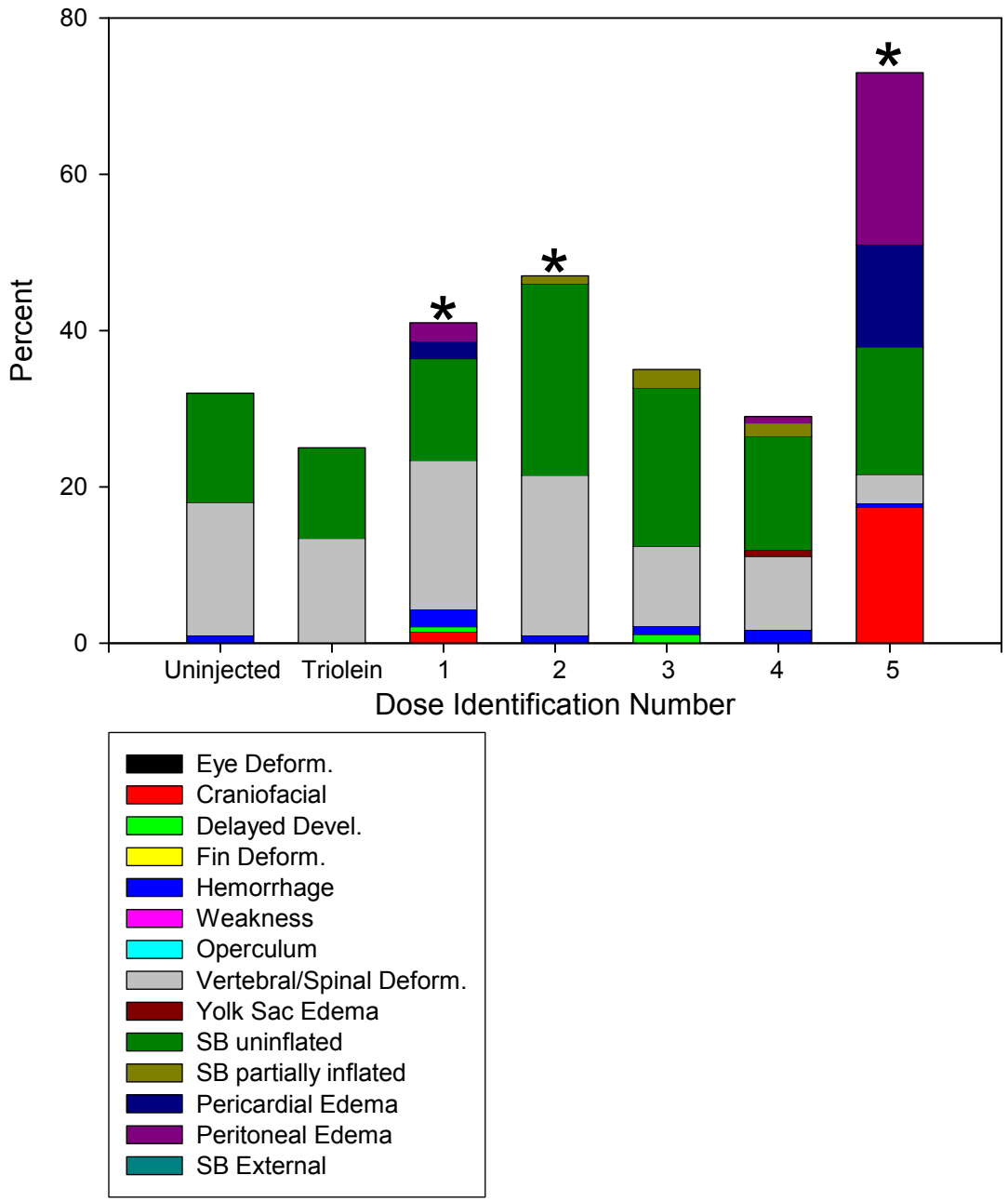


Figure 16. Effects of in ovo exposure to increasing doses of TCDD on largemouth bass at swim-up. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

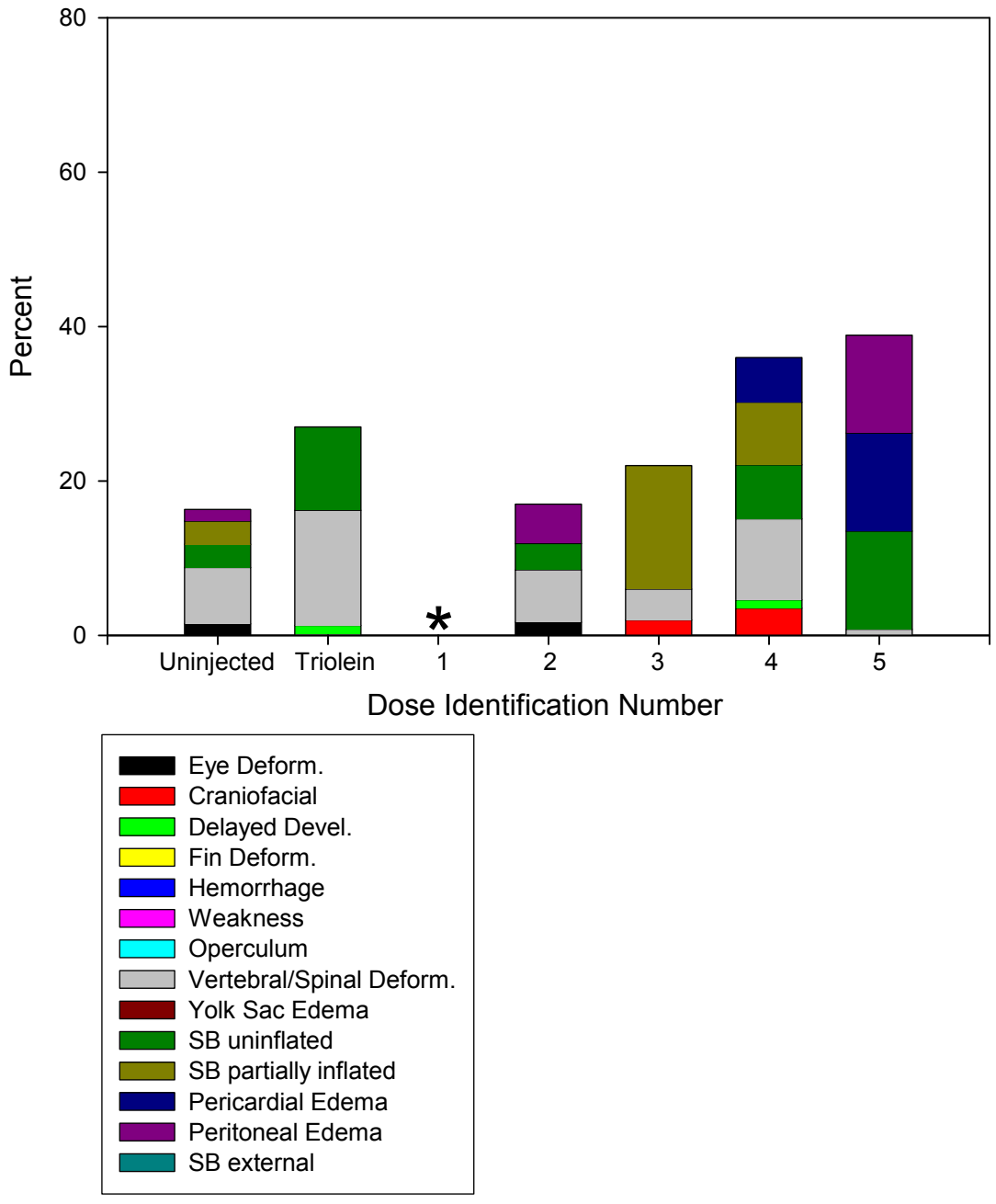


Figure 17. Dose-response curves of effects on largemouth bass at swim-up from exposure to increasing concentrations of Woods Pond extracts or PCB 126. Concentrations are represented by TEQs. The inverted triangle shows the mean percentage of Woods Pond offspring experiencing effects in the Phase I study at measured egg TEQ concentrations.

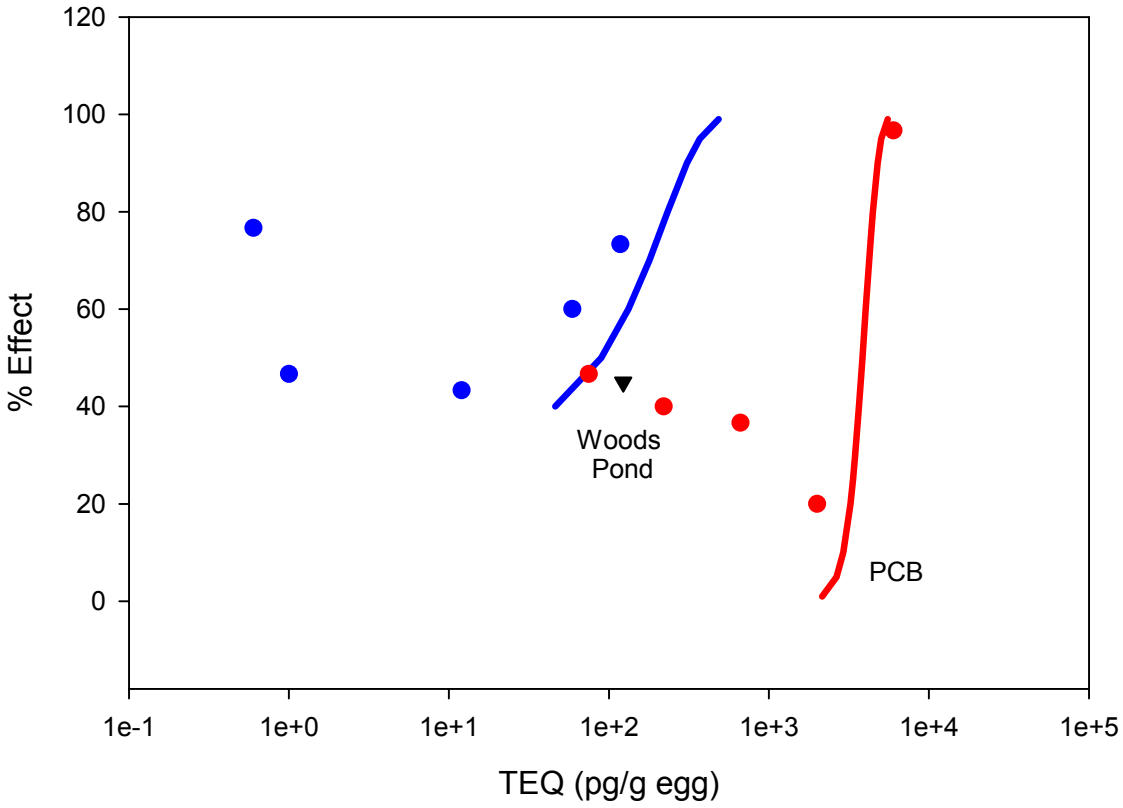


Table 27. ED50 values for largemouth bass at swim-up following in ovo injection with Woods Pond extracts, PCB 126, and TCDD. Values are expressed as pg TEQs per g egg.

	ED50	95% CI	Slope
Woods Pond (Trial 1)	90	39-141	71
PCB 126 (Trial 2)	3750	2462-5036	115

Biochemistry:

Cytochrome P450 1A induction in largemouth bass 15d post swim-up fish was low and showed no clear dose-response pattern (Table 28). Positive reactions to the antibody were observed in gill and kidney tissues of Three-Mile Pond, Woods Pond, PCB, and TCDD injected fish.

Table 28. Qualitative assessment of induction of cytochrome P450 1A in histologically prepared tissues of whole largemouth bass at 15d post swim-up. Results are presented for three doses representing low, medium, and high exposures. Asterisks represent the extent and intensity of the response (* = low, ** = moderate, and * = high).**

			Dose		
	n	**Triolein**	**1**	**3**	**5**
Three-Mile Pond	19	None	*	None	None
Rising Pond	13	None	None	None	None
Woods Pond	17	None	None	*	None
Deep Reach	9	None	None	None	None
PCB 126	9	None	*	ND	None
2,3,7,8-TCDD	13	None	None	None	*

ND – No data.

5.4 Responses of Medaka to Standards and Extracts

Survival and Growth:

100h post hatch -- Survival in negative controls was generally greater than 70% (Table 29). It ranged from 10% to 90%, and averaged 81±16% for uninjected fish and 72±19% in triolein-injected fish. At 100h post hatch only PCB at 400 and 1200 ng/g egg caused significantly higher mortality than negative controls. Dose-response curves for lethality were developed (Figure 18). Mortality data derived from fish treated with extracts of largemouth bass from Woods Pond (Trial 2) and Deep Reach (Trial 1) met the test acceptance criteria, but only Deep Reach (Trial 1) data was suitable for statistical analysis and determination of LD50s. The LD50 for Deep Reach (95 pg TEQ/g egg) was approximately 6 and 60 times lower than for PCB (580 pg TEQ/g egg) and TCDD (5481 pg TEQ/g egg), respectively (Table 30). Slopes of the linear portions of the curves were similar (Table 30).

15d post swim-up -- At 15d post swim-up, all treatment doses of the extract from Deep Reach fish (0.6-128 pg TEQ/g egg), the highest dose tested of the extract from Woods Pond fish (118 pg TEQ/g egg), PCB 126 at doses ≥ 665 pg TEQ/g egg, and TCDD at doses ≥ 670 pg/g egg all had significantly greater mortality than controls (Table 31). Survival for Rising Pond and Three-Mile Pond fish exposed to the highest concentrations of those extracts did not differ from the negative controls. Dose-response curves for mortality were developed from these data (Figure 19). The LD50 for the Deep Reach extract (47 pg TEQ/g egg) was lower than for that for PCB 126 (241 pg TEQ/g egg) and TCDD (1579 pg TEQ/g egg). Slopes of linear portions of all the dose-response curves were similar (Table 32).

Neither weight nor length at this stage of development was affected by exposure to the extracts, PCB 126 or 2,3,7,8-TCDD (Tables 33–38).

Table 29. Percent survival at 100h post hatch of medaka exposed in ovo to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Values are means of 3 replicate samples of 10 eggs each (standard deviations in parentheses). Trials are temporally distinct dose-response experiments.

	% Survival																		
	Three-mile Pond			Rising Pond				Woods Pond			Deep Reach			PCB 126			2,3,7,8-TCDD		
Dose	Trial 1	Trial 2	Trial 3	Trial 1	Trial 2	Trial 3	Trial 4	Trial 1	Trial 2	Trial 3	Trial 1	Trial 2	Trial 3	Trial 1	Trial 2	Trial 3	Trial 1	Trial 2	Trial 3
Uninjected	80 (0)	90 (0)	90 (0)	20 (30)	70 (10)	80 (0)	90 (10)	80 (0)	90 (10)	70 (0)	90 (10)	90 (10)	90 (10)	80 (0)	80 (10)	90 (10)	---	90 (10)	90 (0)
Triolein	80 (10)	90 (0)	80 (20)	10 (10)	60 (10)	80 (10)	90 (10)	---	70 (10)	60 (10)	90 (10)	50 (10)	70 (10)	70 (10)	60 (10)	80 (10)	90 (0)	80 (10)	80 (0)
1	80 (10)	100 (10)	70 (10)	10 (10)	70 (10)	80 (10)	90 (10)	80 (10)	40 (0)	80 (10)	80 (10)	80 (10)	70 (10)	70 (10)	ND	ND	90 (10)	80 (10)	ND
2	80 (10)	90 (10)	60 (20)	0 (0)	60 (10)	90 (10)	90 (10)	90 (0)	50 (10)	80 (10)	100 (10)	40 (10)	90 (10)	70 (10)	70 (10)	80 (0)	---	90 (10)	80 (10)
3	60 (0)	90 (10)	80 (0)	30 (0)	40 (10)	80 (10)	80 (10)	100 (10)	20* (0)	90 (10)	60 (10)	50 (10)	80 (10)	80 (10)	60 (10)	30 (20)	---	90 (10)	70 (0)
4	80 (10)	70 (10)	80 (10)	0 (0)	90 (10)	70 (10)	90 (10)	80 (10)	50 (10)	80 (0)	70 (10)	40 (10)	80 (10)	---	10 (10)	30 (10)	70 (10)	90 (10)	80 (10)
5	80 (10)	90 (10)	70 (20)	0 (0)	80 (10)	80 (10)	80 (10)	80 (10)	40 (10)	90 (10)	30 (20)	50 (0)	70 (0)	---	0* (0)	0* (0)	---	100 (10)	100 (10)
6														10* (10)	0* (0)	0* (0)	60 (10)	ND	40 (10)

*** Significantly different from negative controls.**
--- High or complete mortality due to fungal infection.
ND - Eggs were not injected at this dose.

Figure 18. Dose-response curves for lethality of medaka at 100 h post hatch from exposure to increasing concentrations of Deep Reach extracts, PCB 126, or TCDD. Concentrations are represented by TEQs.

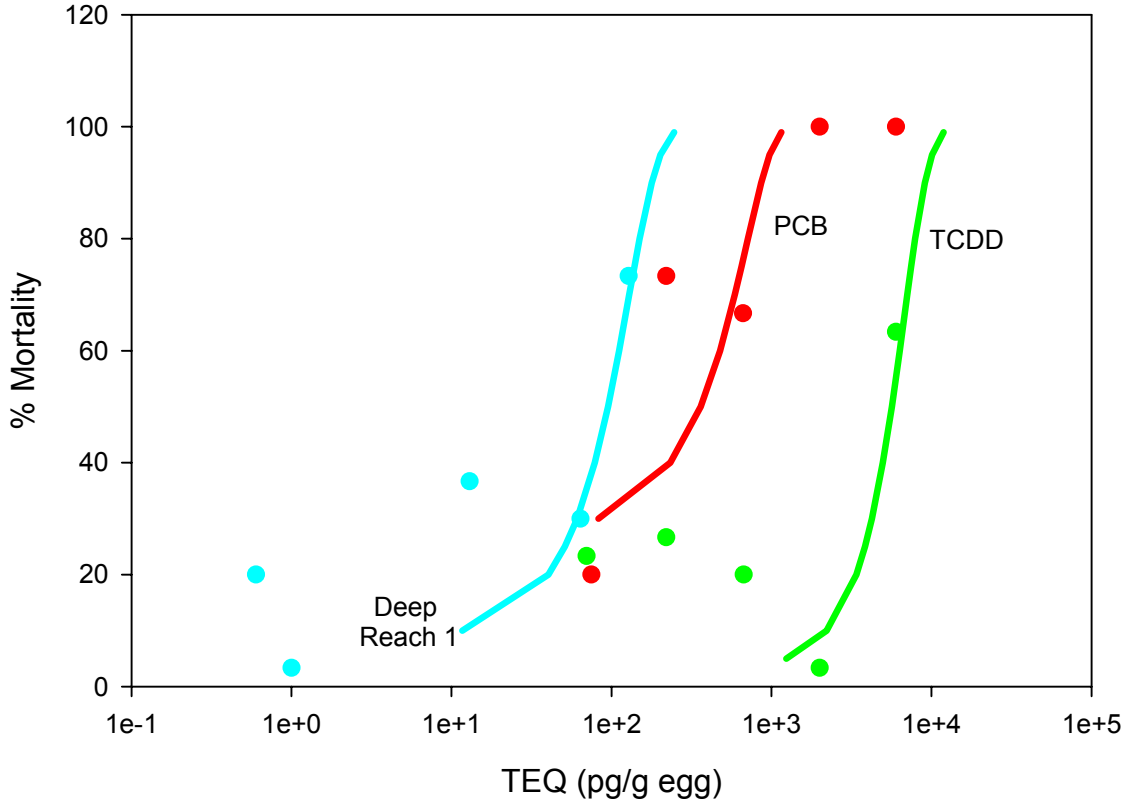


Table 30. LD50 values for medaka at 100h post hatch following in ovo injection with Deep Reach extracts, PCB 126, and TCDD. Values are expressed as pg TEQs per g egg.

	LD50	95% CI	Slope
Deep Reach (Trial 1)	95	75-116	114
PCB 126 (Trial 3)	580	379-782	104
2,3,7,8-TCDD (Trial 3)	5481	3950-7013	142

Table 31. Percent survival at 15d post swim-up of medaka exposed in ovo to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Values are means of 3 replicate samples of 10 eggs each (standard deviations in parentheses). Trials are temporally distinct dose-response experiments.

	% Survival																		
	Three-mile Pond			Rising Pond				Woods Pond			Deep Reach			PCB 126			2,3,7,8-TCDD		
Dose	Trial 1	Trial 2	Trial 3	Trial 1	Trial 2	Trial 3	Trial 4	Trial 1	Trial 2	Trial 3	Trial 1	Trial 2	Trial 3	Trial 1	Trial 2	Trial 3	Trial 1	Trial 2	Trial 3
Uninjected	70 (10)	20 (20)	90 (10)	0 (1)	70 (10)	80 (0)	80 (10)	80 (0)	80 (10)	70 (10)	80 (10)	90 (10)	80 (0)	80 (0)	60 (10)	80 (10)	---	60 (10)	0[a] (10)
Triolein	60 (30)	90 (0)	80 (20)	1 (2)	60 (10)	80 (10)	70 (10)	---	60 (20)	60 (10)	90 (20)	50 (10)	70 (10)	70 (10)	50 (10)	80 (10)	80 (10)	50 (20)	80 (10)
1	60 (10)	80 (10)	70 (10)	0 (0)	70 (10)	80 (10)	80 (10)	70 (10)	40 (10)	70 (10)	70* (10)	80 (10)	70 (10)	70 (10)	ND	ND	80 (10)	80 (10)	ND
2	70 (20)	80 (10)	60 (20)	0 (0)	60 (10)	90 (10)	60 (20)	90 (0)	50 (10)	50 (30)	70* (10)	40 (10)	80 (10)	50 (20)	70 (10)	80 (10)	---	90 (10)	70 (10)
3	50 (10)	40 (10)	70 (20)	0 (1)	40 (0)	70 (10)	50 (10)	90 (10)	20 (0)	70 (10)	20* (10)	50 (10)	0* (0)	0* (0)	10 (10)	0* (10)	---	70 (20)	70 (10)
4	70 (0)	60 (0)	80 (10)	0 (0)	80 (10)	70 (10)	90 (10)	60 (10)	50 (0)	40 (20)	50* (10)	40 (10)	40 (10)	---	0 (0)	0* (10)	70 (10)	70 (10)	0* (0)
5	80 (10)	70 (0)	70 (10)	0 (0)	70 (10)	70 (10)	70 (10)	40* (20)	20 (10)	40 (20)	10* (20)	50 (0)	0* (10)	---	0 (0)	0* (10)	---	0 (0)	70 (0)
6														0* (0)	0 (0)	0* (0)	20* (10)	ND	0* (10)

[a] Fish escaped, uninjected data not used in analyses.
* Significantly different from negative controls.
--- High or complete mortality due to fungal infection.
ND - Eggs were not injected at this dose.

Figure 19. Dose-response curves for lethality of medaka at 15d post swim-up from exposure to increasing concentrations of Deep Reach extracts, PCB 126, or TCDD. Concentrations are represented by TEQs.

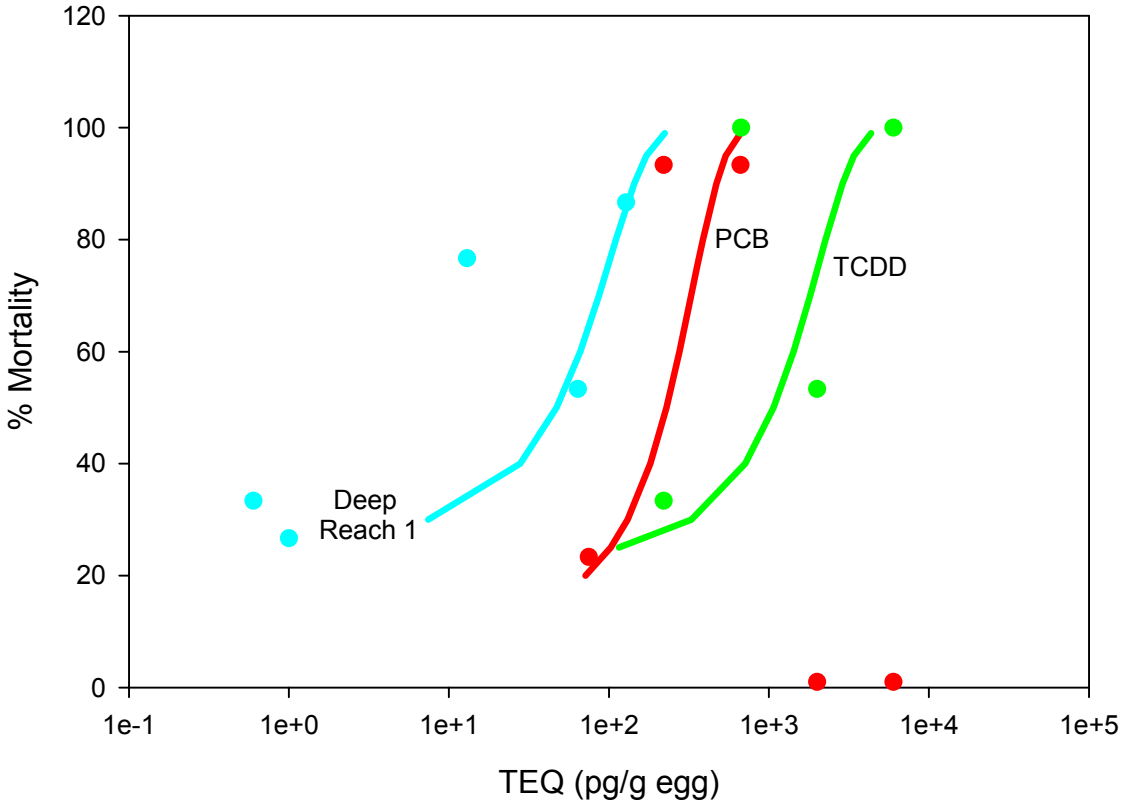


Table 32. LD50 values for medaka at 15d post swim-up following in ovo injection with Deep Reach extracts, PCB 126, and TCDD. Values are expressed as pg TEQs per g egg.

	LD50	95% CI	Slope
Deep Reach (Trial 1)	47	30-64	75
PCB 126 (Trial 3)	241	131-352	103
2,3,7,8-TCDD	1579	927-2230	82

Table 33. Mean lengths and weights (± standard deviations) of medaka exposed to Three-Mile Pond extracts. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2			Trial 3		
Dose	**N**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	21	8.5 ± 1.0	0.0046 ± 0.0015	5	9.0 ± 2.5	0.0052 ± 0.0031	18	9.0 ± 0.00	1.182 ± 0.0017
Triolein	19	8.5 ± 1.0	0.0043 ± 0.0016	27	9.0 ± 1.0	0.0044 ± 0.0017	24	9.0 ± 0.0	0.7614 ± 0.0016
1	17	8.0 ± 1.0	0.0038 ± 0.0016	24	9.0 ± 1.0	0.0044 ± 0.0012	16	9.5 ± 0.0	0.4755 ± 0.0012
2	20	8.5 ± 1.0	0.0043 ± 0.0020	24	8.5 ± 1.0	0.0042 ± 0.0014	16	9.5 ± 0.0	0.6124 ± 0.0018
3	14	9.0 ± 1.5	0.0050 ± 0.0022	11	8.0 ± 1.5	0.0035 ± 0.0021	19	9.5 ± 0.0	0.7504 ± 0.0016
4	21	9.5 ± 1.0	0.0059 ± 0.0022	18	9.0 ± 0.5	0.0044 ± 0.0012	22	9.0 ± 0.0	0.4990 ± 0.0012
5	25	9.0 ± 1.0	0.0057 ± 0.0017	14	9.0 ± 1.5	0.0046 ± 0.0017	19	9.5 ± 0.0	0.5590 ± 0.0009

No significant differences between treatments and controls were detected.

Table 34. Mean lengths and weights (± standard deviations) of medaka exposed to Rising Pond extracts. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2			Trial 3		
Dose	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	23	8.5 ± 1.0	0.0036 ± 0.0017	25	9.0 ± 0.5	0.0046 ± 0.0011	24	9.0 ± 1.0	0.0050 ± 0.0014
Triolein	19	9.0 ± 1.0	0.0045 ± 0.0015	24	9.0 ± 0.5	0.0045 ± 0.0010	20	9.0 ± 0.5	0.0055 ± 0.0010
1	21	9.5 ± 0.5	0.0049 ± 0.0014	23	8.0 ± 1.0	0.0037 ± 0.0017	25	9.0 ± 1.0	0.0054 ± 0.0018
2	18	8.0 ± 1.5	0.0030 ± 0.0015	27	8.5 ± 0.5	0.0040 ± 0.0009	18	8.5 ± 1.0	0.0050 ± 0.0019
3	12	8.5 ± 1.0	0.0033 ± 0.0014	22	9.0 ± 0.5	0.0047 ± 0.0011	16	9.5 ± 0.5	0.0063 ± 0.0020
4	25	8.0 ± 1.0	0.0029 ± 0.0014	21	9.0 ± 0.5	0.0049 ± 0.0009	26	9.0 ± 1.0	0.0053 ± 0.0017
5	20	8.0 ± 1.0	0.0027 ± 0.0014	21	8.5 ± 0.5	0.0037 ± 0.0010	22	9.0 ± 0.5	0.0057 ± 0.0014

No significant differences between treatments and controls were detected.

Table 35. Mean lengths and weights (± standard deviations) of medaka exposed to Woods Pond extracts. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2			Trial 3		
Dose	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	24	10.0 ± 0.5	0.0069 ± 0.0014	24	9.5 ± 1.0	0.0037 ± 0.0014	20	9.0 ± 1.0	0.0056 ± 0.0013
Triolein	0	ND	ND	17	8.0 ± 1.0	0.0030 ± 0.0011	17	9.0 ± 1.0	0.0056 ± 0.0017
1	23	9.0 ± 0.5	0.0055 ± 0.0013	11	8.5 ± 1.0	0.0041 ± 0.0016	22	9.0 ± 0.5	0.0048 ± 0.0014
2	26	9.0 ± 0.5	0.0062 ± 0.0017	14	8.5 ± 1.5	0.0034 ± 0.0017	16	8.0 ± 1.5	0.0036 ± 0.0020
3	28	9.0 ± 1.0	0.0055 ± 0.0016	1	10	0.0058	21	9.0 ± 0.5	0.0049 ± 0.0015
4	19	9.0 ± 1.0	0.0053 ± 0.0022	15	8.5 ± 1.0	0.0034 ± 0.0013	12	8.0 ± 1.0	0.0039 ± 0.0022
5	11	9.0 ± 9.0	0.0052 ± 0.0016	7	7.5 ± 1.0	0.0028 ± 0.0011	13	9.0 ± 0.5	0.0058 ± 0.0014

No significant differences between treatments and controls were detected.
ND - Eggs were not injected at this dose.

Table 36. Mean lengths and weights (± standard deviations) of medaka exposed to Deep Reach extracts. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2			Trial 3		
Dose	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	24	9.0 ± 1.0	0.0058 ± 0.0018	26	8.5 ± 2.0	0.0044 ± 0.0025	24	9.0 ± 0.5	0.0057 ± 0.0012
Triolein	26	9.5 ± 0.5	0.0059 ± 0.0013	16	9.5 ± 1.5	0.0057 ± 0.0014	20	9.0 ± 1.0	0.0054 ± 0.0018
1	20	9.0 ± 0.5	0.0053 ± 0.0016	26	9.0 ± 1.0	0.0054 ± 0.0014	20	9.0 ± 0.5	0.0051 ± 0.0014
2	22	9.0 ± 0.5	0.0056 ± 0.0011	11	9.0 ± 0.5	0.0058 ± 0.0012	23	9.0 ± 0.5	0.0047 ± 0.0010
3	7	7.5 ± 1.5	0.0042 ± 0.0034	18	9.0 ± 1.5	0.0055 ± 0.0023	0	---	---
4	14	9.0 ± 0.5	0.0051 ± 0.0013	13	10 ± 0.5	0.0076 ± 0.0016	12	9.0 ± 1.0	0.0055 ± 0.0014
5	4	9.0 ± 0.5	0.0051 ± 0.0010	21	8.5 ± 1.5	0.0049 ± 0.0024	0	---	---

No significant differences between treatments and controls were detected.

Table 37. Mean lengths and weights (± standard deviations) of medaka exposed to PCB 126. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2			Trial 3		
Dose	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	24	9.5 ± 1.0	0.0061 ± 0.0016	18	7.5 ± 1.5	0.0031 ± 0.0020	25	9.0 ± 1.0	0.0051 ± 0.0014
Triolein	21	9.0 ± 0.5	0.0055 ± 0.0013	16	8.6 ± 0.70	0.0045 ± 0.0009	23	9.5 ± 0.5	0.0055 ± 0.0011
2	21	9.0 ± 0.5	0.0055 ± 0.0011	20	8.4 ± 1.2	0.0043 ± 0.0017	23	9.0 ± 0.5	0.0052 ± 0.0013
3	17	8.5 ± 1.0	0.0042 ± 0.0016	2	6.5 ± 2.1	0.0025 ± 0.0026	2	7.5 ± 3.0	0.0035 ± 0.0041
4	0	ND	ND	0	ND	ND	2	10 ± 0.5	0.0085 ± 0.0004
5	0	ND	ND	0	ND	ND	0	ND	ND
6	2	10 ± 0.0	0.0061 ± 0.0009	0	ND	ND	0	ND	ND

No significant differences between treatments and controls were detected.
ND - Eggs were not injected at this dose.

Table 38. Mean lengths and weights (± standard deviations) of medaka exposed to TCDD. Trials are temporally distinct dose-response experiments.

	Trial 1			Trial 2		
Dose	**n**	**Length (cm)**	**Weight (g)**	**n**	**Length (cm)**	**Weight (g)**
Uninjected	11	8.5 ± 0.5	0.0040 ± 0.0011	17	8.0 ± 1.5	0.0037 ± 0.0017
Triolein	24	8.0 ± 0.5	0.0040 ± 0.0014	14	8.0 ± 1.5	0.0037 ± 0.0017
1	25	8.5 ± 0.5	0.0042 ± 0.0009	24	8.5 ± 1.5	0.0041 ± 0.0017
2	13	8.5 ± 0.5	0.0038 ± 0.0011	26	9.0 ± 1.0	0.0048 ± 0.0015
3	3	8.0 ± 0.5	0.0036 ± 0.0009	21	9.0 ± 1.0	0.0046 ± 0.0017
4	22	8.5 ± 0.5	0.0045 ± 0.0013	22	8.5 ± 1.0	0.0044 ± 0.0017
5	2	9.5 ± 0.5	0.0051 ± 0.0004	0	ND	ND
6	6	7.5 ± 1.5	0.0034 ± 0.0015	ND	ND	ND

No significant differences between treatments and controls were detected.
ND - Eggs were not injected at this dose.

Pathology:

Hatch – Recently-hatched medaka exposed to Housatonic River extracts experienced a greater number of pathologies than did those fish exposed to Three-Mile Pond extract or the negative controls (Tables 39 and 42). The only pathology observed in negative control fish was yolk sac edema. Three-Mile pond fish showed low levels of pericardial, peritoneal and yolk sac edema. The three Housatonic River extracts induced craniofacial anomalies, hemorrhaging, and problems associated with inflating the swim bladder (Figure 20). Deep Reach and Rising Pond exposed fish also showed signs of spinal deformities, edema, and weakness. Additionally, fish treated with the Deep Reach extract had eye deformities and delayed development, that were not apparent in the negative control hatchlings. No data were collected for PCB and TCDD exposed fish at this stage. Observations made and included in the "Other" category included pale coloration, abnormal swimming behavior, not eating, small size, spherical yolk sac, and incomplete hatch.

100h post hatch -- All swim-up medaka had difficulty inflating swim bladders, although this condition was most pronounced for PCB, TCDD and Deep Reach exposed fish (Tables 40 and 43). Larval weakness was observed in all but the Woods Pond-exposed fish, and was most severe in PCB and TCDD fish. Medaka exposed to PCBs had highest frequency of edemas and only those exposed to TCDD showed signs of craniofacial and eye deformities. Pathological observations made at this stage of development in the medaka and included in the "Other" category included not eating, pale coloration, abnormal swimming behavior, and emaciation.

15d post swim-up -- At 15d post swim-up, only a few pathologies were observed in medaka (Tables 41 and 44). Any of the Housatonic extracts, PCB- or TCDD-exposed fish continued to have problems inflating swim bladders. Medaka exposed to extracts derived from Rising Pond fish showed spinal deformities, and both Rising Pond and Woods Pond extracts produced hemorrhage in exposed medaka at 15d post swim-up. Woods Pond extract treatment also led to weak and lethargic medaka swim-up fry. Fish exposed to the extract derived from Deep Reach fish or those exposed to TCDD had craniofacial deformities. TCDD-exposed medaka were also retarded in their development at 15d post swim-up. Pathological observations made at the 15d

post swim-up stage in medaka and included in the "Other" category included anemia, emaciation, pale coloration, and lump at base of caudal peduncle.

The percentage of fish with one or more pathologies tended to increase with increased exposure concentrations of extracts or the chemical standards, except for those fish exposed to the Three-Mile Pond extract (Figures 21-26). Predicted NOELs and LOELs were variable for all extracts and standards. No NOEL or LOEL was established for Three-Mile Pond because they would have been greater than the highest dose administered. The geometric mean of the NOEL (81 pg TEQ/g egg) and LOEL (161 pg TEQ/g egg) for the Rising Pond extract dosed in ovo to medaka was 114 pg TEQ/g egg. The geometric mean of the NOEL (12 pg TEQ/g egg) and the LOEL (59 pg TEQ/g egg) for the Woods Pond extract dosed in ovo to medaka was 27 pg TEQ/g egg, while the geometric mean for the NOEL (0.6 pg TEQ/g egg) and the LOEL (1.0 pg TEQ/g egg) was the lowest for the Deep Reach extract at 0.8 pg TEQ/g egg. The geometic mean of the PCB 126 NOEL (75 pg TEQ/g egg) and LOEL (220 pg TEQ/g egg) in medaka was 128 pg TEQ/g egg, and that for the NOEL and LOEL (220 and 670 pg TEQ/g egg, respectively) for TCDD was 384 pg TEQ/g egg. Dose-response curves for cumulative effects (mortalities and pathologies) were developed for the medaka exposures (Figure 27). Slopes calculated from the linear portions of these curves were similar (Table 45). The potency of extracts developed from Deep Reach fish (ED50: average of two trials, 48 pg TEQ/g egg) to cause deformities and mortality in medaka was approximately 10 times greater than PCB 126 (ED50: 448 pg TEQ/g egg) and approximately 60 times greater than 2,3,7,8-TCDD (ED50: 2719 pg TEQ/g egg). Estimates of ED50s for Three-mile and Rising Ponds could not be made because effects were not greater than 50% at the highest doses. Effects data derived from medaka treated with extracts of largemouth bass from Woods Pond (Trial 2) met the test acceptance criteria, but data were not suitable for statistical analysis and determination of ED50s.

Table 39. Estimated occurrence of gross pathologies (per 1000 individuals) in medaka at hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 3,084.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Craniofacial Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	15	0	0	0
2	0	0	0	13	0	0
3	0	58	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Eye Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	13	0	0
3	0	0	0	0	0	0
4	0	0	0	15	0	0
5	0	0	0	0	0	0
6					0	0
			Vertebral/Spinal Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	24	0	0
2	0	0	0	0	0	0
3	0	11	0	0	0	0
4	0	19	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Hemorrhage			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	15	0	0	0
2	0	0	0	0	0	0
3	0	35	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	13	0	0
6					0	0

Table 39 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in medaka at hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 3,084.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Pericardial Edema			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	12	0	0	0	0	0
2	0	0	0	13	0	0
3	0	0	0	13	0	0
4	0	19	0	0	0	0
5	12	0	0	0	0	0
6					0	0
			Peritoneal Edema			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	13	0	0
3	0	58	0	13	0	0
4	0	19	0	0	0	0
5	12	0	0	0	0	0
6					0	0
			Yolk Sac Edema			
Uninjected	0	70	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	9	0	13	0	0
3	0	105	0	13	0	0
4	0	19	0	15	0	0
5	12	50	0	0	0	0
6					0	0
			Larval Weakness			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	20	0	0	0	0
2	0	9	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	15	0	0
5	0	25	0	0	0	0
6					0	0

Table 39 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in medaka at hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 3,084.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Delayed Development			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	13	0	0
3	0	0	0	0	0	0
4	0	0	0	15	0	0
5	0	0	0	0	0	0
6					0	0
			Other			
Uninjected	0	11	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	13	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	37	0	0	0	0
6					0	0

Table 40. Estimated occurrence of gross pathologies (per 1000 individuals) in medaka at 100h post hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 2,486.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Craniofacial Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	14
5	0	0	0	0	0	0
6					0	0
			Eye Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	14
5	0	0	0	0	0	0
6					0	0
			Vertebral/Spinal Deformity			
Uninjected	0	0	0	13	14	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	85	0	15	0	0	0
3	0	0	0	0	0	0
4	0	0	0	17	0	0
5	0	0	0	0	0	0
6					0	0
			Uninflated Swim Bladder			
Uninjected	106	13	14	121	14	87
Triolein	64	13	27	44	15	0
1	154	68	16	26	10	0
2	85	56	78	111	14	31
3	72	68	33	216	47	156
4	14	40	0	120	500	304
5	45	56	37	75	0	322
6					0	190

Table 40 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in medaka at 100h post hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 2,486.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Partially-Inflated Swim Bladder			
Uninjected	15	0	0	27	14	0
Triolein	0	0	0	14	0	0
1	0	0	16	0	0	0
2	28	0	0	15	0	31
3	14	0	0	100	0	19
4	0	0	0	120	0	0
5	0	28	0	242	0	0
6					0	142
			Hemorrhage			
Uninjected	0	0	0	0	14	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	17	0	0
5	0	0	0	15	0	0
6					0	0
			Pericardial Edema			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	14	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	23	0
4	0	0	0	0	125	0
5	0	0	0	0	0	0
6					0	0
			Peritoneal Edema			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	47	0
4	0	0	0	0	125	0
5	0	0	0	0	0	0
6					0	0

Table 40 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in medaka at 100h post hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 2,486.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Yolk Sac Edema			
Uninjected	0	0	0	0	14	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	95	0
4	0	0	0	0	125	14
5	0	0	0	0	0	0
6					0	47
			Larval Weakness			
Uninjected	0	13	0	13	0	17
Triolein	0	0	0	0	15	0
1	28	41	0	0	0	19
2	28	14	0	0	0	15
3	28	34	0	200	0	0
4	14	13	0	34	500	28
5	0	70	0	0	0	322
6					0	333
			Delayed Development			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	17	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Other			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	19
2	0	0	0	31	0	0
3	0	0	33	300	47	0
4	0	0	222	120	500	260
5	0	0	148	242	0	0
6					0	0

Table 41. Estimated occurrence of gross pathologies (per 1000 individuals) in medaka at 15d post swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 2,017.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Craniofacial Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	40	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Eye Deformity			
Uninjected	0	0	0	15	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Vertebral/Spinal Deformity			
Uninjected	0	0	0	0	14	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	31	0	0	0	0
3	0	19	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Uninflated Swim Bladder			
Uninjected	43	0	0	106	29	35
Triolein	28	0	0	0	33	32
1	33	14	22	15	0	0
2	16	31	74	107	0	33
3	66	0	0	40	100	22
4	45	13	65	34	0	0
5	33	0	32	0	0	62
6					0	142

Table 41 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in medaka at 15d post swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 2,017.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Partially-Inflated Swim Bladder			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	13	0	0	0	0
5	0	15	0	0	0	0
6					0	0
			Hemorrhage			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	45	0	0	0
2	0	15	18	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Peritoneal Edema			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Larval Weakness			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	111	0	0	0
3	0	0	0	0	0	0
4	0	0	21	0	0	0
5	0	0	0	0	0	0
6					0	0

Table 41 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in medaka at 15d post swim-up after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 2,017.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Delayed Development			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	80	0	0
4	0	0	0	0	0	0
5	33	0	0	0	0	0
6					0	142
			Other			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	16	0	16
1	0	0	0	15	0	0
2	0	0	0	0	0	0
3	0	0	0	40	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0

Table 42. Estimated occurrence of gross pathologies (per 1000 individuals) at hatch in medaka exposed to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials and all doses combined.

	Treatment						
Observation	**Three-Mile Pond**	**Rising Pond**	**Woods Pond**	**Deep Reach**	**PCB 126**	**2,3,7,8-TCDD**	**Control**
Craniofacial Deformity	0	1	3	3	ND	ND	0
Eye Deformity	0	0	0	5	ND	ND	0
Vertebral/Spinal Deformity	0	6	0	5	ND	ND	0
Uninflated Swim Bladder	0	0	3	14	ND	ND	0
Partially-Inflated Swim Bladder	0	29	37	0	ND	ND	0
Hemorrhage	0	6	3	3	ND	ND	0
Pericardial Edema	5	4	0	5	ND	ND	0
Peritoneal Edema	3	15	0	5	ND	ND	0
Yolk Sac Edema	3	33	0	8	ND	ND	9
Larval Weakness	0	11	0	3	ND	ND	0
Delayed Development	0	0	0	5	ND	ND	0
Other	0	6	0	3	ND	ND	2

ND - Eggs were not injected at this dose.

Table 43. Estimated occurrence of gross pathologies (per 1000 individuals) at 100h post hatch in medaka exposed to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials and all doses combined.

	Treatment						
Observation	**Three-Mile Pond**	**Rising Pond**	**Woods Pond**	**Deep Reach**	**PCB 126**	**2,3,7,8-TCDD**	**Control**
Craniofacial Deformity	0	0	0	0	0	3	0
Eye Deformity	0	0	0	0	0	3	0
Vertebral/Spinal Deformity	17	0	3	3	0	0	3
Uninflated Swim Bladder	75	58	33	106	59	152	44
Partially-Inflated Swim Bladder	9	6	3	93	0	21	6
Hemorrhage	0	0	0	6	0	0	1
Pericardial Edema	3	0	0	0	17	0	0
Peritoneal Edema	0	0	0	3	25	0	0
Yolk Sac Edema	0	0	0	0	42	7	1
Larval Weakness	20	35	0	43	84	73	5
Delayed development	0	3	0	0	0	0	0
Other	0	0	80	134	50	66	0

Table 44. Estimated occurrence of gross pathologies (per 1000 individuals) at 15d post swim-up in medaka exposed to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials and all doses combined.

	Treatment						
Observation	**Three-Mile Pond**	**Rising Pond**	**Woods Pond**	**Deep Reach**	**PCB 126**	**2,3,7,8-TCDD**	**Control**
Craniofacial Deformity	0	0	0	5	0	0	0
Eye Deformity	0	0	0	0	0	0	1
Vertebral/Spinal Deformity	0	9	0	0	0	0	1
Uninflated Swim Bladder	37	13	41	45	23	23	3
Partially-Inflated Swim Bladder	0	6	0	0	0	0	0
Hemorrhage	0	3	14	0	0	0	0
Peritoneal Edema	0	0	0	0	0	0	0
Larval Weakness	0	0	32	0	0	0	0
Delayed Development	7	0	0	10	0	5	0
Other	0	0	0	10	0	0	1

Figure 20. Medaka exhibiting craniofacial deformity, hemorrhages, and yolk sac edema at hatch following in ovo exposure to Woods Pond extracts.

Figure 21. Effects of in ovo exposure to increasing doses of Three-Mile Pond extracts on medaka at 100h post hatch. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

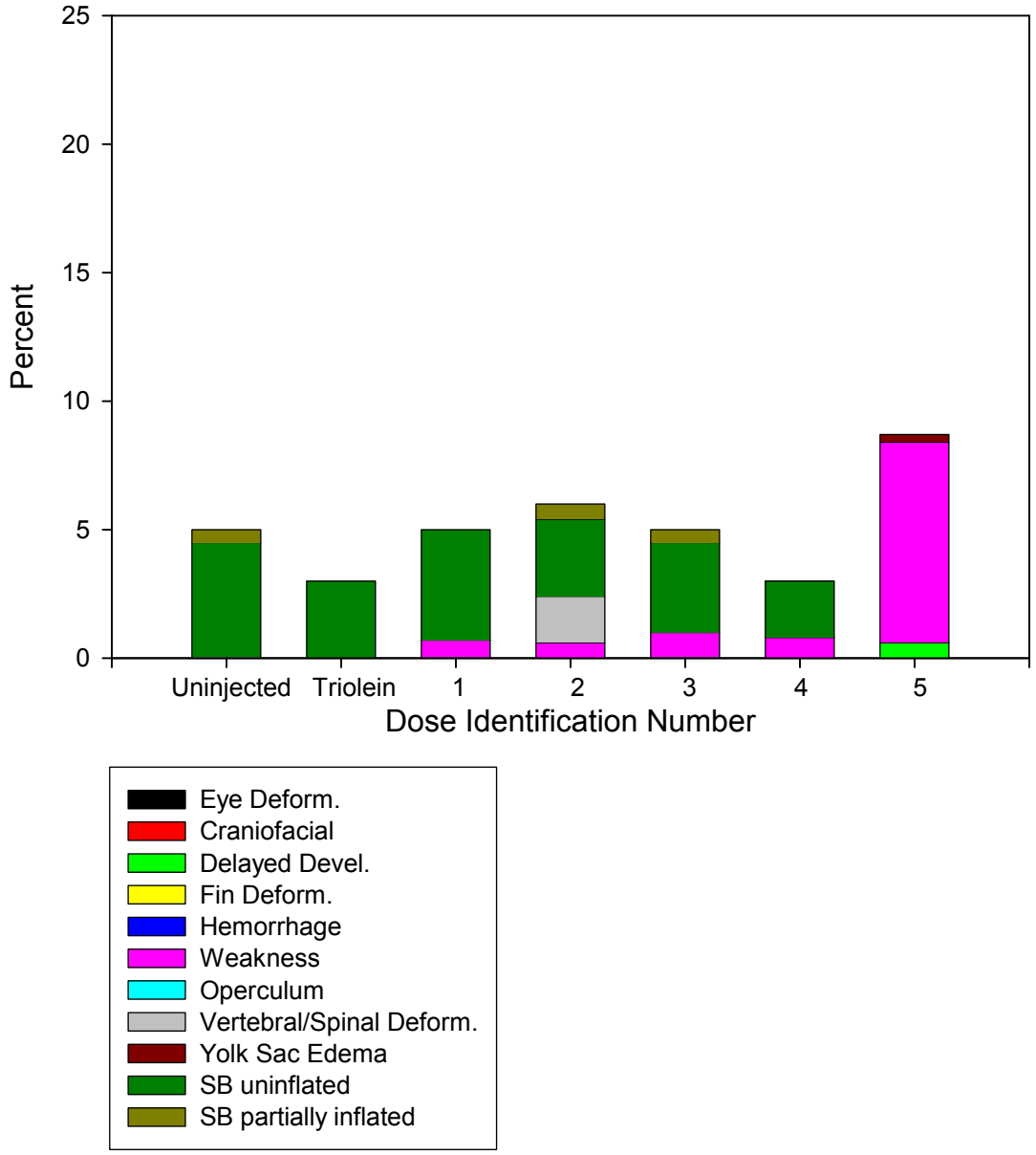


Figure 22. Effects of in ovo exposure to increasing doses of Rising Pond extracts on medaka at 100h post hatch. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

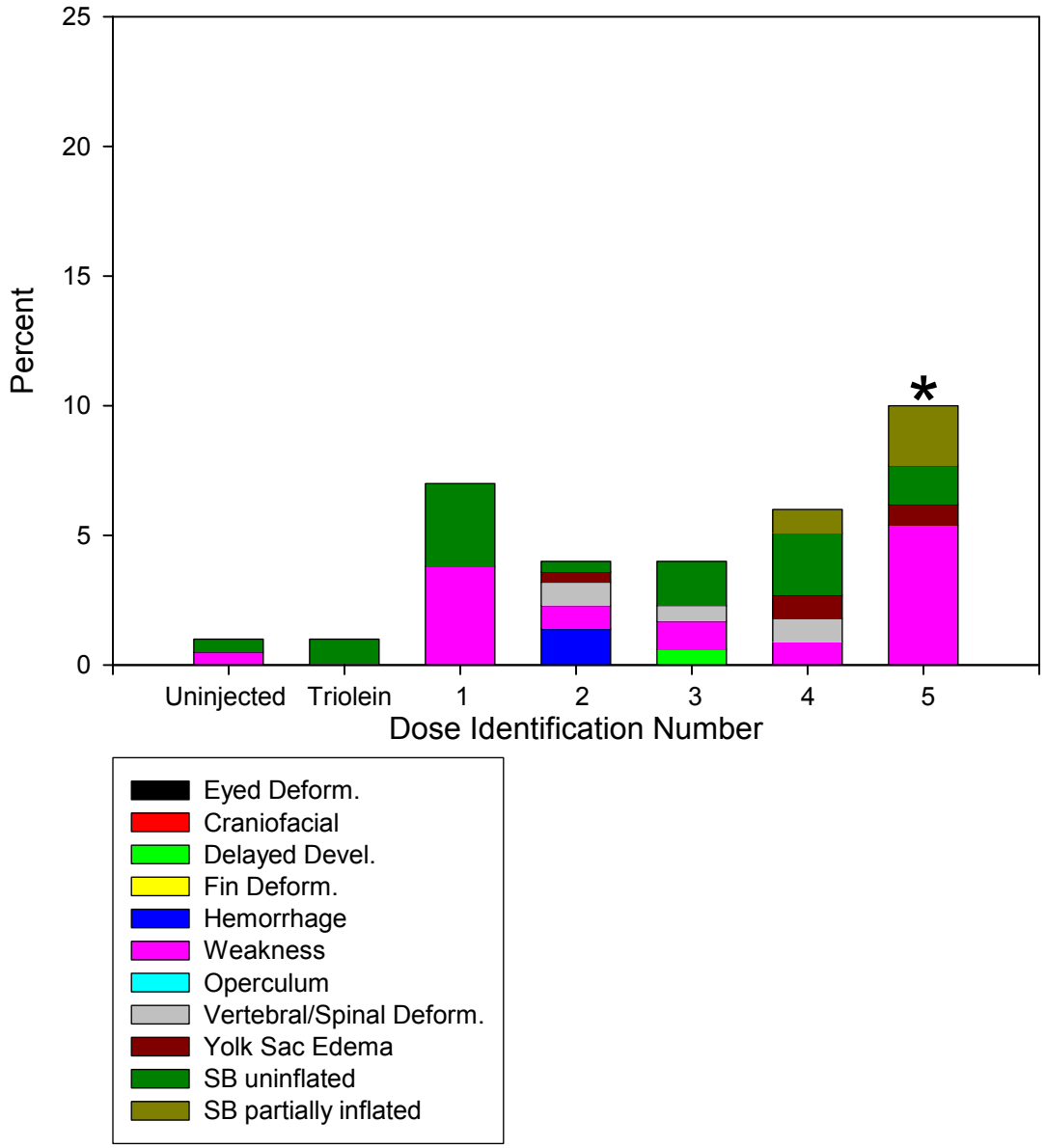


Figure 23. Effects of in ovo exposure to increasing doses of Woods Pond extracts on medaka at 100h post hatch. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

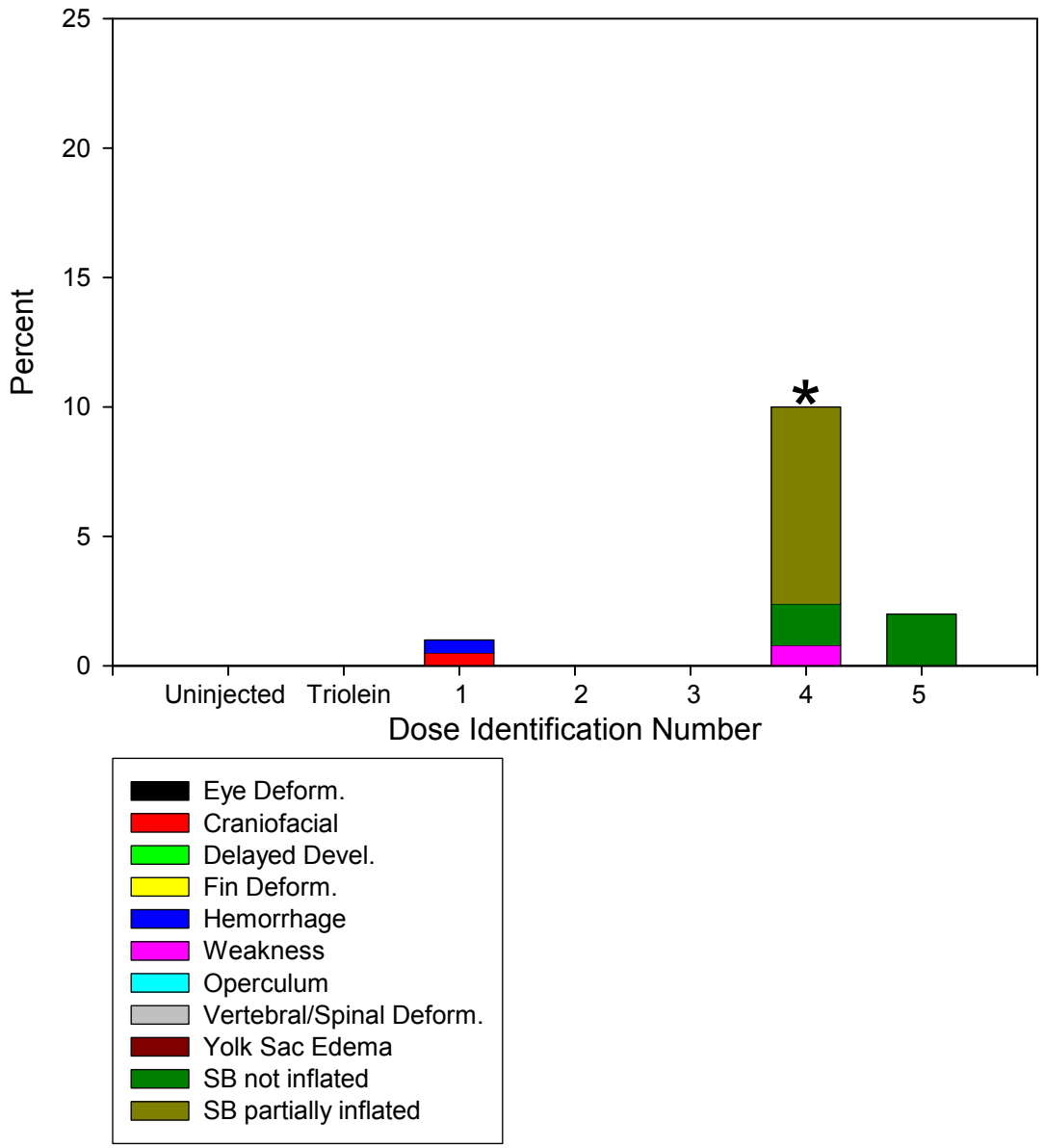


Figure 24. Effects of in ovo exposure to increasing doses of Deep Reach extracts on medaka at 100h post hatch. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

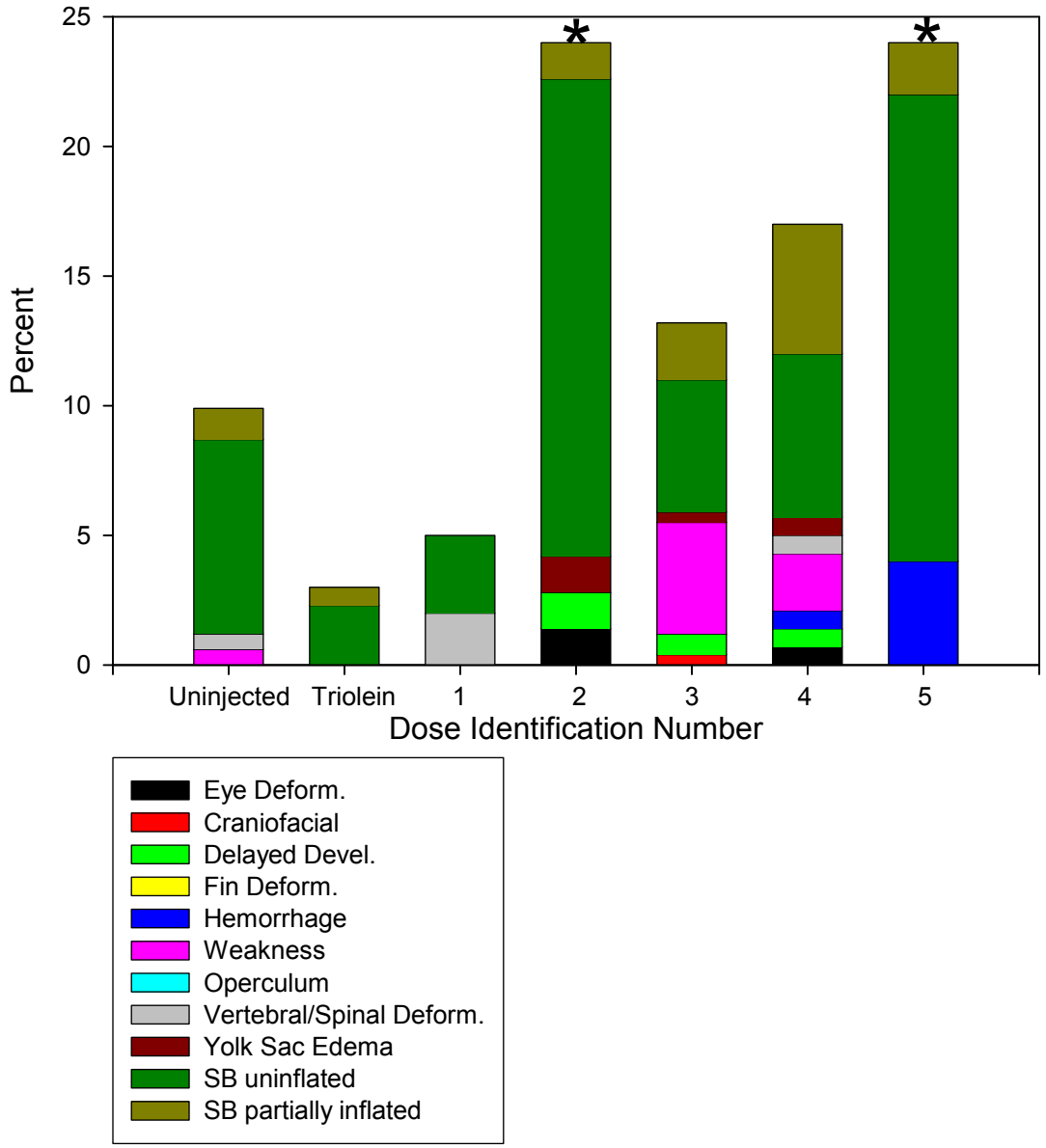


Figure 25. Effects of in ovo exposure to increasing doses of PCB 126 on medaka at 100h post hatch. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

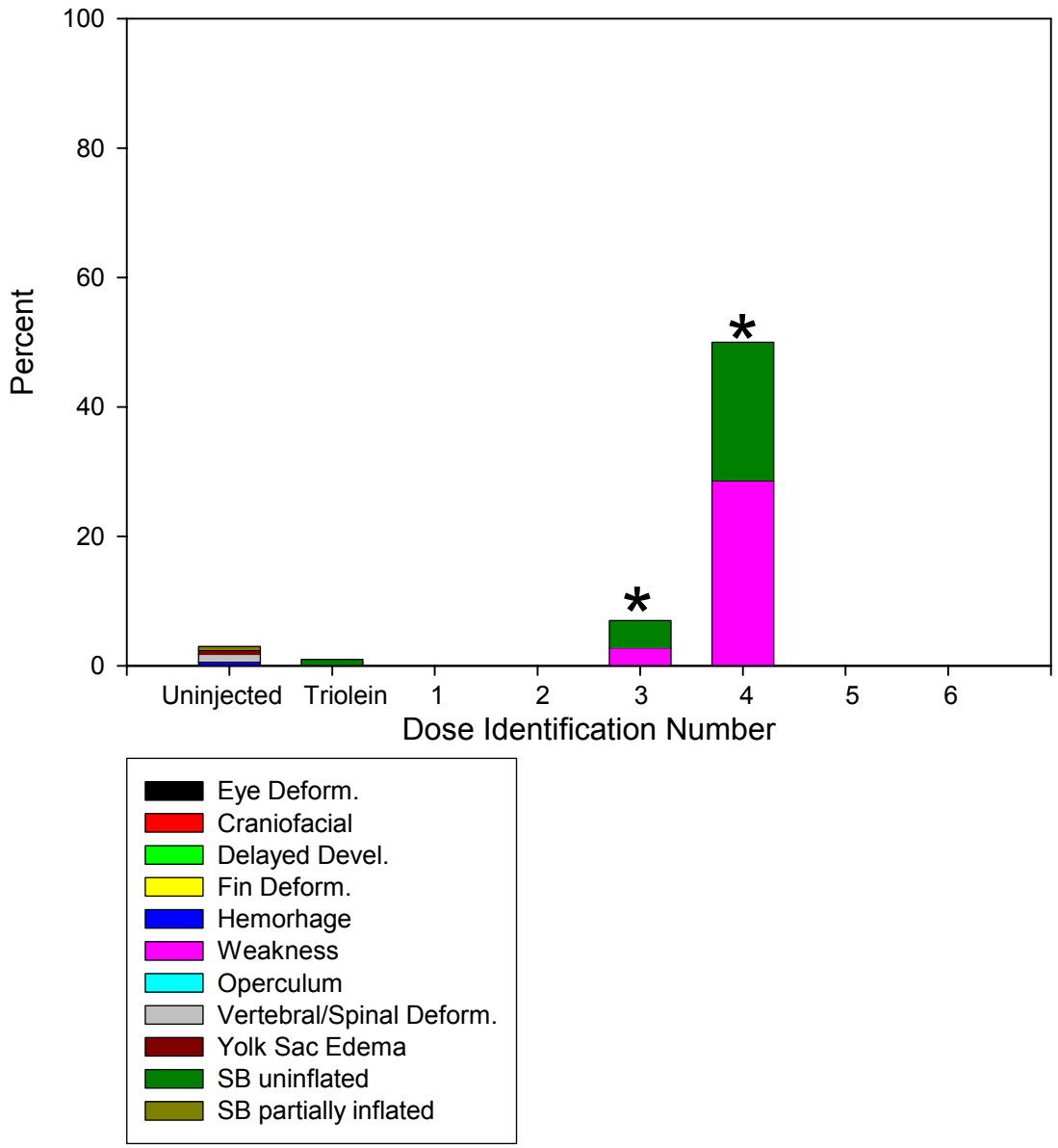


Figure 26. Effects of in ovo exposure to increasing doses of TCDD on medaka at 100h post hatch. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

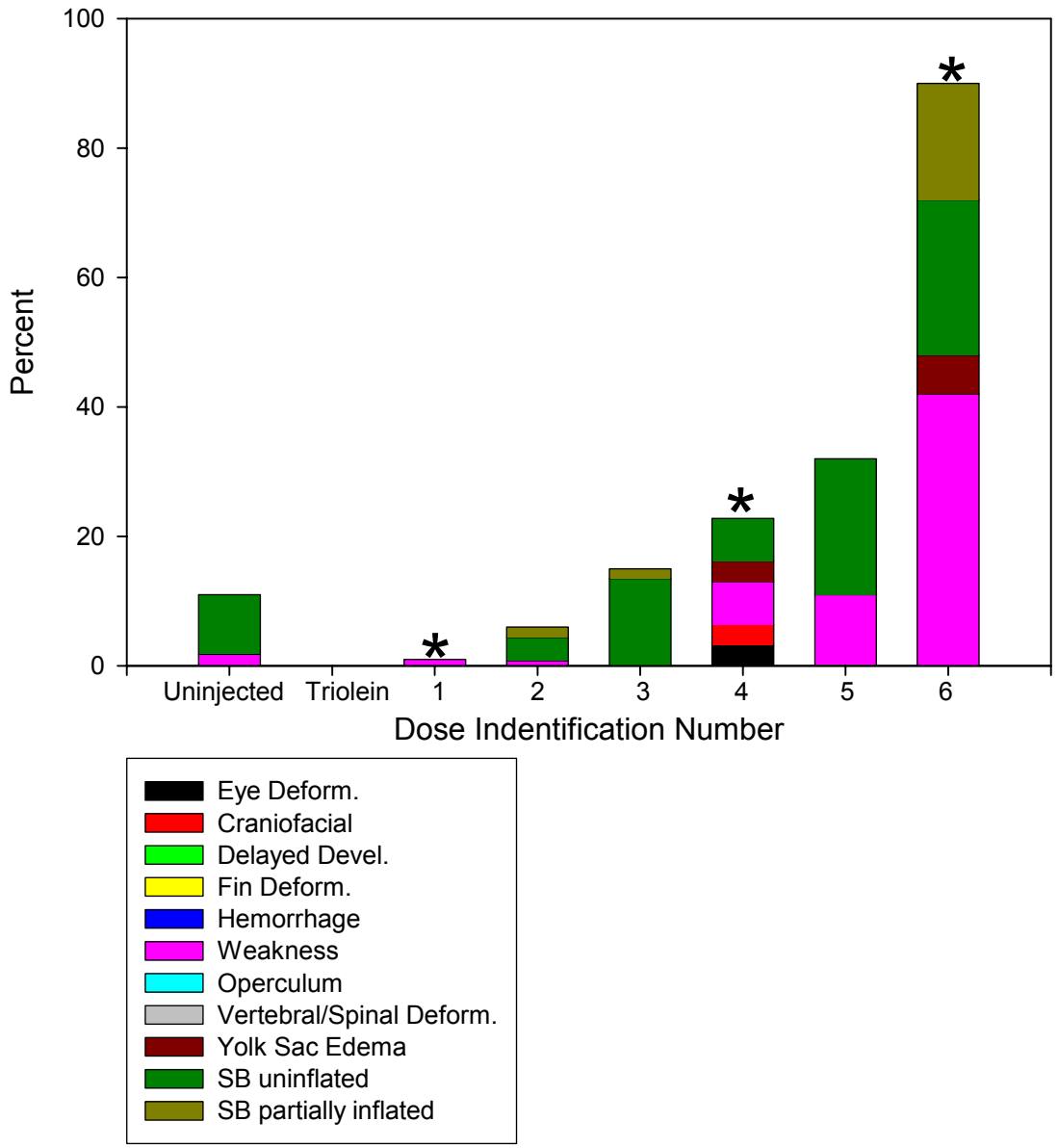


Figure 27. Dose-response curves of effects on medaka at 100h post hatch from exposure to increasing concentrations of Deep Reach extracts, PCB 126, or TCDD. Concentrations are represented by TEQs.

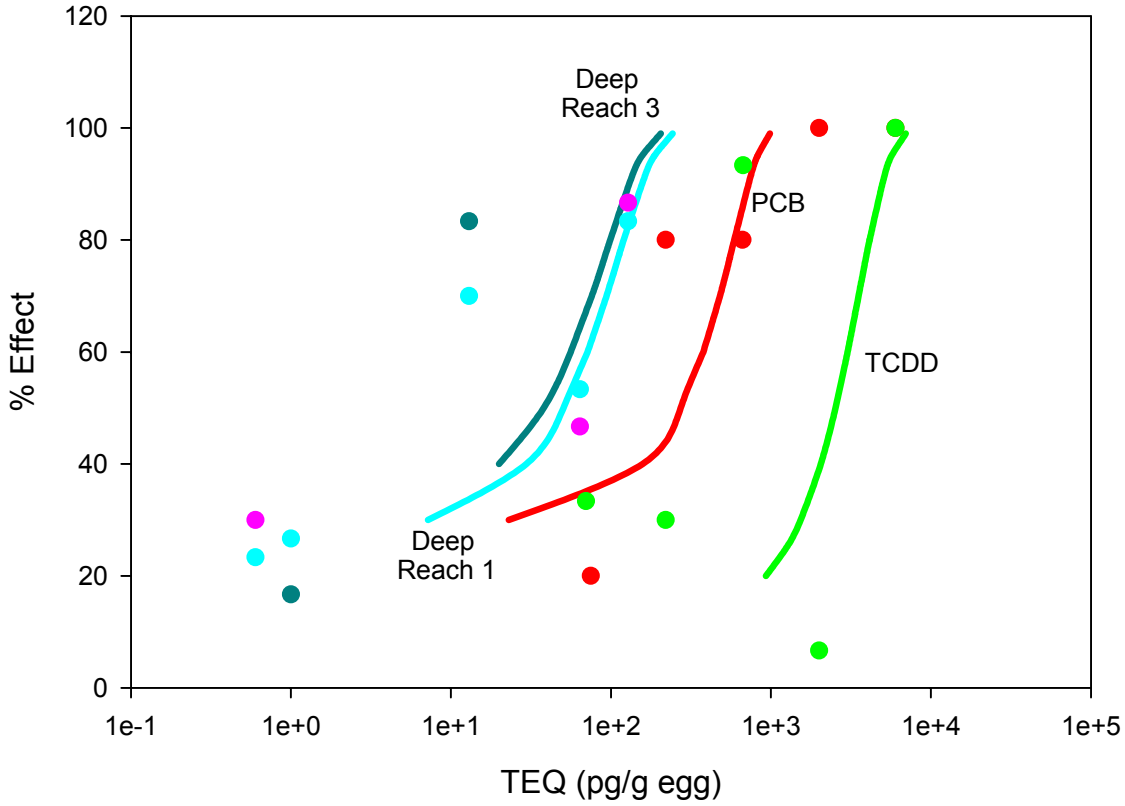


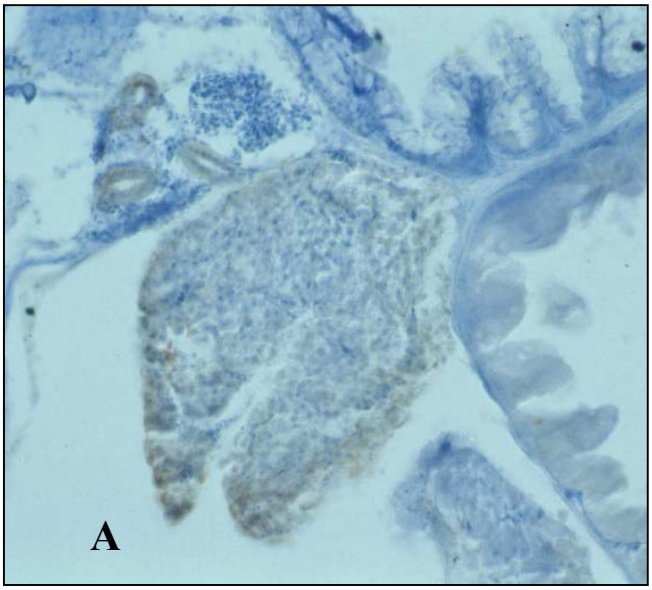
Table 45. ED50 values for medaka at 100h post hatch following in ovo injection with Deep Reach extracts, PCB 126, and TCDD. Values are expressed as pg TEQs per g egg.

	ED50	95% CI	Slope
Deep Reach (Trial 1)	51	33-70	75
Deep Reach (Trial 3)	45	41-73	95
PCB 126 (Trial 3)	448	268-629	96
2,3,7,8-TCDD (Trial 3)	2719	1950-3488	108

Biochemistry:

Cytochrome P450 1A (CYP 1A) was detected at moderate levels primarily in brain, kidney, and liver tissues of TCDD-exposed fish at Dose 3 and above and also in Three-Mile Pond exposed fish at the highest dose (Figure 28). Low levels of CYP 1A were seen in Rising Pond fish (Dose 3), all doses of Woods Pond fish, and in PCB-exposed fish (Dose 3). CYP 1A was strongly detected in Deep Reach fish at Dose 3 (Table 46).

Figure 28. Cytochrome P450 1A staining in medaka at 15d post swim-up following in ovo exposure (500X magnification). A – liver and kidney (Three-Mile Pond extracts), B – liver and kidney (TCDD).



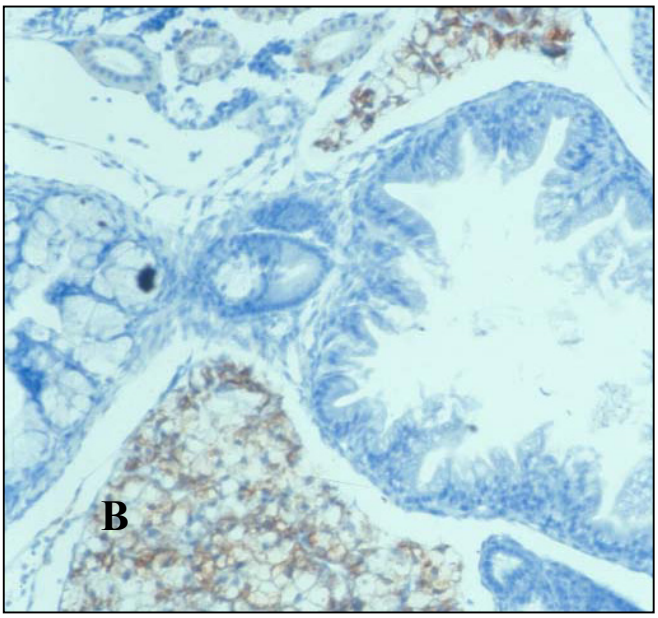


Table 46. Qualitative assessment of induction of cytochrome P450 1A in histologically prepared tissues of whole medaka at 15d post swim-up. Results are presented for three doses representing low, medium, and high exposures. Asterisks represent the extent and intensity of the response (* = low, ** = moderate, and * = high).**

			Dose		
	n	**Triolein**	**1-2**	**3-4**	**5-6**
Three-Mile Pond	28	None	None	ND	**
Rising Pond	32	None	None	*	None
Woods Pond	30	None	*	*	*
Deep Reach	14	None	**	***	*
PCB 126	9	None	ND	*	ND
2,3,7,8-TCDD	23	None	None	**	**

ND – No data.

5.5 Responses of Rainbow Trout to Standards and Extracts

Survival:

Rainbow trout survival at 600 DTUs averaged 88±13% for uninjected embryos and 87±10% for triolein injected embryos across trials. Mortality was significantly greater than controls only at the highest doses of PCB and TCDD (1800 pg TEQ/g egg) and at the 200 pg TEQ/g egg dose of TCDD (Table 47). Dose-response curves for PCB and TCDD are shown in Figure 29. The estimated LD50 for PCB 126 (average of two trials, 999 pg TEQ/g egg) was similar to that of 2,3,7,8-TCDD (average of two trials, 1073 pg TEQ/g egg) and the slopes for the predicted response curves were similar (Table 48).

Table 47. Percent survival at 600 DTU of rainbow trout exposed in ovo to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Values are means of 3 replicate samples of 24 eggs each (standard deviations in parentheses). Trials are temporally distinct dose-response experiments.

	% Survival													
	Three-mile Pond		Rising Pond		Woods Pond		Deep Reach				PCB 126		2,3,7,8-TCDD	
Dose	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 3	Trial 4	Trial 1	Trial 2	Trial 1	Trial 2
Uninjected	94 (7)	100 (0)	97 (2)	96 (3)	98 (3)	94 (5)	63 (18)	67 (8)	67 (2)	100 (0)	86 (6)	95 (1)	80 (20)	89 (5)
Triolein	94 (7)	99 (3)	93 (2)	92 (5)	99 (3)	86 (9)	77 (28)	69 (19)	78 (1)	99 (2)	72 (20)	94 (3)	80 (10)	85 (10)
1	97 (3)	100 (0)	91 (5)	91 (4)	95 (5)	93 (3)	45 (15)	52 (4)	70 (11)	98 (3)	77 (3)	97 (3)	80 (6)	84 (9)
2	90 (5)	100 (0)	90 (6)	87 (7)	94 (10)	83 (8)	62 (32)	58 (13)	79 (11)	97 (3)	78 (7)	89 (10)	83 (10)	89 (9)
3	88 (2)	99 (2)	83 (15)	93 (6)	96 (8)	91 (8)	59 (8)	71 (12)	77 (6)	96 (4)	79 (13)	91 (1)	86 (7)	80 (13)
4	94 (2)	97 (2)	94 (5)	92 (3)	97 (3)	86 (5)	50 (15)	68 (23)	78 (4)	90 (5)	71 (22)	93 (7)	77 (5)	46* (7)
5	90 (4)	93 (3)	90 (10)	87 (5)	100 (0)	91 (8)	82 (22)	62 (9)	83 (13)	94 (7)	50 (8)	83 (3)	61 (20)	81 (15)
6											0* (0)	5* (6)	23* (6)	9* (9)

* Significantly different from negative controls.

Figure 29. Dose-response curves for lethality of rainbow trout at 600 DTU from exposure to increasing concentrations of PCB 126 or TCDD. Concentrations are represented by TEQs.

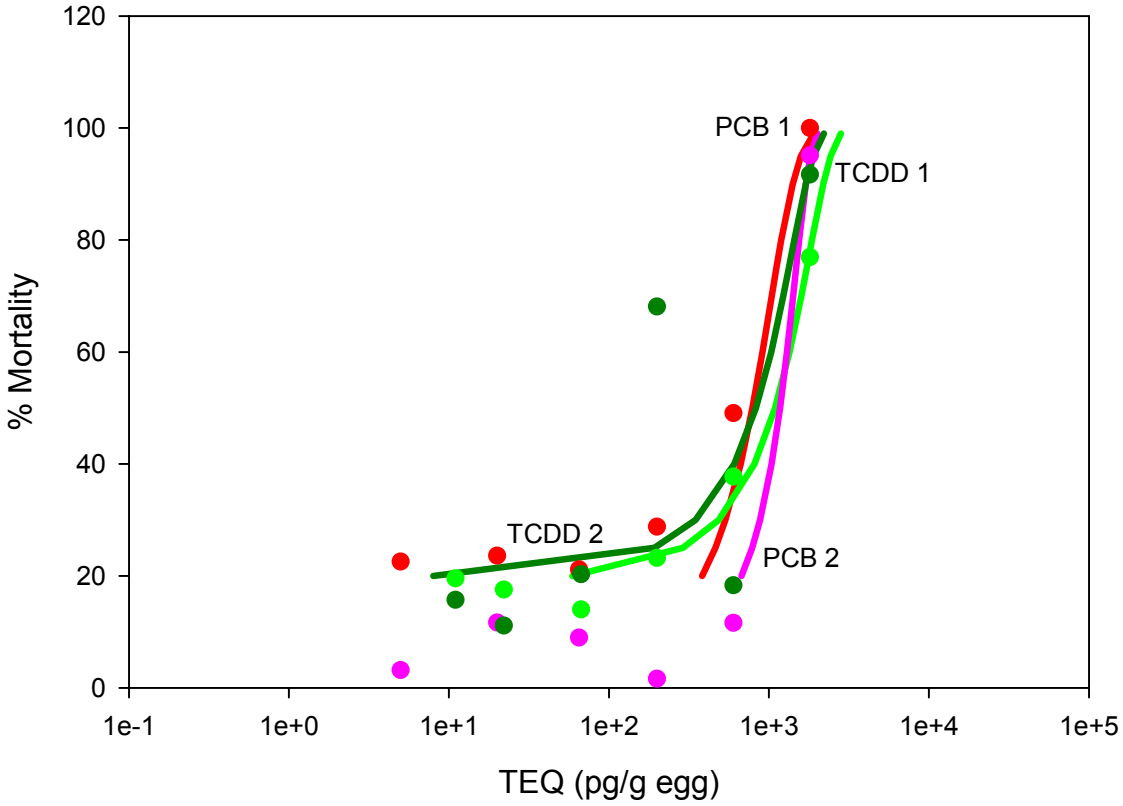


Table 48. LD50 values for rainbow trout at 600 DTU following in ovo injection with PCB 126 and TCDD. Values are expressed as pg TEQs per g egg.

	LD50	95% CI	Slope
PCB 126 (Trial 1)	813	665-963	121
PCB 126 (Trial 2)	1185	1095-1276	208
2,3,7,8-TCDD (Trial 1)	1257	894-1620	124
2,3,7,8-TCDD (Trial 2)	889	673-760	121

Pathology:

Hatch -- Recently hatched rainbow trout larvae exposed to PCB and TCDD showed elevated signs of craniofacial deformities, hemorrhaging, and yolk sac edema (Tables 49 and 51). Fin deformities were also observed in PCB fish but not TCDD fish. Delayed development was the most prominent pathology observed in Housatonic River extract-exposed trout. Deep Reach fish also had an elevated incidence of craniofacial and opercular deformities, and hemorrhaging (Figure 30). Additional pathological observations that were compiled for the "Other" category in newly hatched rainbow trout included incomplete closure of the abdomen, and darkened coloration.

600 DTU -- At 600 DTU rainbow trout sac fry exposed to PCB and TCDD continued to experience craniofacial deformities, and yolk sac edema (Tables 50 and 52; Figure 31). Delayed development was also observed and was severe at higher doses. Craniofacial and opercular deformities along with delayed development characterized the pathologies observed in fish exposed to Housatonic River extracts (Figures 32-34). Additional pathological observations in rainbow trout at 600 DTUs included under the "Other" category were twinning, deformed yolk sac, malformed anus, raised tissues on head, enlarged yolk sac, hardened yolk sac, and small size.

The percentage of fish with one or more pathologies tended to increase with exposure concentrations of either extracts or positive standard chemicals (Figures 35-40). NOELs and LOELs for the all extracts were similar and much lower than for the standards on a TEQ basis. No NOEL was established for Deep Reach within our study design where the LOEL was estimated to be 0.6 pg TEQ/g egg. The geometric mean of the NOEL (0.7 pg TEQ/g egg) and LOEL (1.0 pg TEQ/g egg) for Rising Pond was 0.8 pg TEQ/g egg and 3.3 pg TEQ/g egg for Woods Pond (NOEL and LOEL were 1.0 and 11.0 pg TEQ/g egg, respectively). The geometric mean for the PCB 126 NOEL (65 pg TEQ/g egg) and LOEL (200 pg TEQ/g egg) was 114 pg TEQ/g egg and 15 pg TEQ/g egg for TCDD (NOEL and LOEL were 11 and 22 pg TEQ/g egg, respectively). Dose-response curves for cumulative effects (mortalities plus pathologies) were developed (Figure 41). Slopes calculated from the linear portions of these curves were similar

(Table 53). The ED50 value for the Woods Pond extract (18 pg TEQ/g egg) was lower than the ED50 value for the Deep Reach extract (49 pg TEQ/g egg). Additionally, both of these extracts had greater potency to cause deformities and mortality than the positive control compounds PCB 126 (ED50: average of two trials, 358 pg TEQ/g egg) or 2,3,7,8-TCDD (ED50:average of two trials, 412 pg TEQ/g egg). No ED50s could be estimated for Three-Mile or Rising Ponds because effects were less than 50% at the highest dose tested.

Table 49. Estimated occurrence of gross pathologies (per 1000 individuals) in rainbow trout at hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 5,139.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Craniofacial Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	6	10	0
2	8	0	0	0	0	0
3	0	0	0	6	0	0
4	0	0	0	6	0	0
5	0	0	0	11	0	0
6					1000	906
			Eye Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	6	0	0
2	0	8	0	0	0	0
3	8	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Fin Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					1000	0
			Vertebral/Spinal Deformity			
Uninjected	15	8	8	30	0	0
Triolein	0	0	0	23	9	0
1	0	17	0	19	10	0
2	0	8	8	12	9	0
3	8	0	8	6	9	0
4	0	8	8	12	0	0
5	24	0	8	6	23	0
6					333	0

Table 49 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in rainbow trout at hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 5,139.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Hemorrhage			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	28	0
1	0	8	0	6	0	0
2	0	8	0	12	0	0
3	0	0	0	6	0	0
4	0	8	0	12	0	0
5	0	0	0	17	0	0
6					1000	969
			Opercular Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	13	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	6	0	0
5	0	0	0	0	0	0
6					0	0
			Pericardial Edema			
Uninjected	0	0	0	0	0	0
Triolein	0	8	0	0	0	0
1	0	8	0	0	0	0
2	0	0	8	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6					0	0
			Peritoneal Edema			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	8	0	0	0
3	0	0	0	0	0	0
4	0	0	0	6	0	0
5	0	0	0	0	0	0
6					0	0

Table 49 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in rainbow trout at hatch after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 5,139.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Yolk Sac Edema			
Uninjected	0	0	0	12	0	0
Triolein	0	0	0	6	28	0
1	0	17	0	0	40	0
2	0	8	8	0	19	0
3	0	0	0	0	0	0
4	8	8	0	0	0	261
5	8	9	0	0	11	35
6					1000	1000
			Larval Weakness			
Uninjected	7	8	0	30	0	0
Triolein	8	8	8	11	9	0
1	0	8	0	13	20	0
2	0	16	8	12	9	0
3	8	9	8	17	0	0
4	23	8	8	6	0	0
5	8	0	16	11	11	0
6					0	0
			Delayed Development			
Uninjected	0	8	0	18	0	0
Triolein	0	8	0	0	0	0
1	8	8	0	20	0	0
2	0	0	0	6	0	0
3	15	20	0	0	0	0
4	8	20	0	61	0	0
5	0	20	0	166	0	0
6					0	0
			Other			
Uninjected	16	0	0	6	0	10
Triolein	0	0	0	6	21	0
1	0	0	0	6	0	0
2	8	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	0	0	28	11	0
6					0	0

Table 50. Estimated occurrence of gross pathologies (per 1000 individuals) in rainbow trout at 600 DTU after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials combined. Total number of fish observed is 5,433.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Craniofacial Deformity			
Uninjected	16	16	0	25	9	0
Triolein	0	0	0	34	31	0
1	0	0	0	20	10	0
2	16	33	0	25	0	0
3	0	0	16	29	0	0
4	0	24	16	6	10	40
5	0	19	33	22	146	88
6					0	200
			Eye Deformity			
Uninjected	0	8	8	0	9	0
Triolein	0	0	0	11	10	0
1	8	0	0	7	0	0
2	8	0	0	0	0	0
3	0	0	8	0	0	0
4	0	8	0	12	0	0
5	0	0	0	11	11	0
6					0	0
			Vertebral/Spinal Deformity			
Uninjected	0	0	0	13	0	0
Triolein	0	0	0	17	10	0
1	0	17	0	39	10	0
2	8	41	0	12	0	0
3	0	0	8	29	9	0
4	23	8	8	0	0	13
5	33	10	8	73	11	11
6					0	0
			Opercular Deformity			
Uninjected	0	0	0	6	0	0
Triolein	0	0	0	11	0	0
1	0	0	0	7	0	0
2	0	0	26	0	0	0
3	0	9	8	29	0	0
4	0	24	32	37	10	0
5	0	19	50	34	11	0
6					0	0

Table 50 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in rainbow trout at 600 DTU after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls. Data are for all trials combined. Total number of fish observed is 5,433.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Yolk Sac Edema			
Uninjected	16	8	8	0	0	0
Triolein	8	0	0	6	10	0
1	0	0	17	13	20	0
2	8	0	9	0	0	0
3	0	0	24	6	0	0
4	16	8	16	0	10	53
5	8	10	17	0	157	22
6					0	533
			Larval Weakness			
Uninjected	0	0	0	0	0	0
Triolein	0	8	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	0	0	0
5	0	10	0	6	0	0
6					0	0
			Delayed Development			
Uninjected	24	40	45	38	18	0
Triolein	8	32	89	29	31	0
1	8	25	84	52	30	0
2	32	66	86	12	126	45
3	0	54	619	29	83	173
4	54	40	266	86	327	227
5	25	87	620	212	371	692
6					0	1000

Table 50 (Cont.). Estimated occurrence of gross pathologies (per 1000 individuals) in rainbow trout at 600 DTU after in ovo exposure to Housatonic River extracts, PCB 126, TCDD, and negative controls. Data are for all trials combined. Total number of fish observed is 5,433.

Dose	Three-Mile Pond	Rising Pond	Woods Pond	Deep Reach	PCB 126	2,3,7,8 TCDD
			Hemorrhage			
Uninjected	16	0	0	0	0	0
Triolein	8	0	0	6	21	10
1	0	0	8	0	10	0
2	8	8	0	6	0	0
3	46	0	0	6	0	0
4	8	0	0	8	0	13
5	0	10	0	22	101	11
6					0	67
			Fin Deformity			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	6	0	0
3	0	0	0	6	0	0
4	0	0	0	0	0	0
5	0	0	0	6	0	0
6					0	0
			Peritoneal Edema			
Uninjected	0	0	0	0	0	0
Triolein	0	0	0	0	0	0
1	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
4	0	0	0	10	0	0
5	0	0	0	0	0	0
6					0	0
			Other			
Uninjected	20	0	0	0	0	0
Triolein	0	0	0	10	20	0
1	0	0	0	0	10	0
2	24	0	0	0	0	0
3	0	9	8	6	0	0
4	8	0	24	12	10	13
5	0	10	25	45	101	11
6					0	67

Table 51. Estimated occurrence of gross pathologies (per 1000 individuals) at hatch in rainbow trout exposed to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials and all doses combined.

	Treatment						
Observation	**Three-Mile Pond**	**Rising Pond**	**Woods Pond**	**Deep Reach**	**PCB 126**	**2,3,7,8-TCDD**	**Control**
Craniofacial Deformity	2	0	0	6	20	92	0
Eye Deformity	2	2	0	1	0	0	0
Fin Deformity	0	0	0	0	8	0	0
Vertebral/Spinal Deformity	6	7	6	11	12	0	9
Opercular Deformity	0	0	0	4	0	0	0
Hemorrhage	0	5	0	11	6	98	2
Pericardial Edema	0	2	2	0	0	0	0
Peritoneal Edema	0	0	0	0	0	0	0
Yolk Sac Edema	3	8	2	0	33	146	4
Larval Weakness	8	8	8	12	8	0	8
Delayed Development	6	20	11	52	0	0	3
Other	2	0	0	7	2	0	1

Table 52. Estimated occurrence of gross pathologies (per 1000 individuals) at 600 DTU in rainbow trout exposed to Housatonic River extracts, PCB 126, TCDD, and negative controls (uninjected and triolein). Data are for all trials and all doses combined.

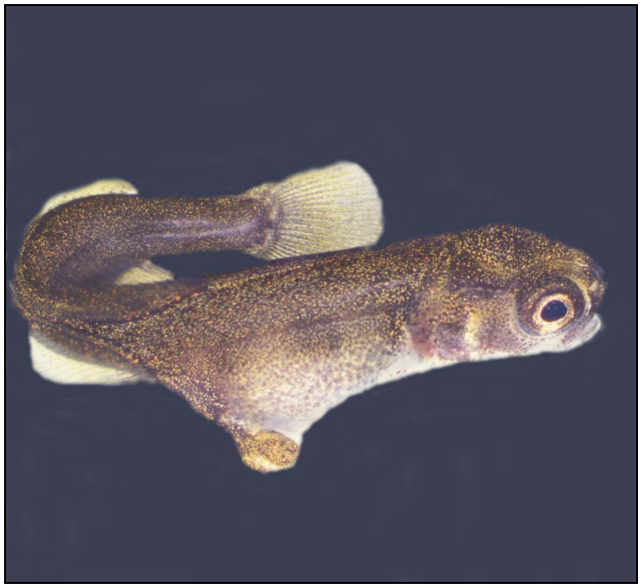
	Treatment						
Observation	**Three-Mile Pond**	**Rising Pond**	**Woods Pond**	**Deep Reach**	**PCB 126**	**2,3,7,8-TCDD**	**Control**
Craniofacial Deformity	3	16	13	21	30	29	12
Eye Deformity	3	2	2	6	2	0	2
Fin Deformity	0	0	0	4	0	0	0
Vertebral/Spinal Deformity	13	16	5	31	6	4	6
Opercular Deformity	0	10	23	22	4	0	2
Hemorrhage	13	3	3	10	20	6	5
Pericardial Edema	0	0	0	0	0	0	1
Peritoneal Edema	0	0	0	1	0	0	0
Yolk Sac Edema	6	3	17	4	34	29	4
Larval Weakness	0	2	0	1	0	0	1
Delayed Development	24	54	340	81	182	245	31
Other	6	3	12	13	22	6	5

Figure 30. Rainbow trout exhibiting craniofacial deformity, hemorrhages, pericardial edema, and yolk sac rupture at hatch following in ovo exposures.

Figure 31. Rainbow trout (bottom) exhibiting craniofacial deformity, yolk sac edema, and delayed development at 600 DTU following in ovo exposure to PCB 126 (top fish is normal).

Figure 32. Rainbow trout (bottom) exhibiting craniofacial deformity, vertebral/spinal deformity, yolk sac edema, and delayed development at 600 DTU following in ovo exposure to Deep Reach extracts (top fish is normal).

Figure 33. Rainbow trout exhibiting craniofacial deformity, vertebral/spinal deformity, anal fin deformity, and delayed development at 600 DTU following in ovo exposure to Deep Reach extracts.



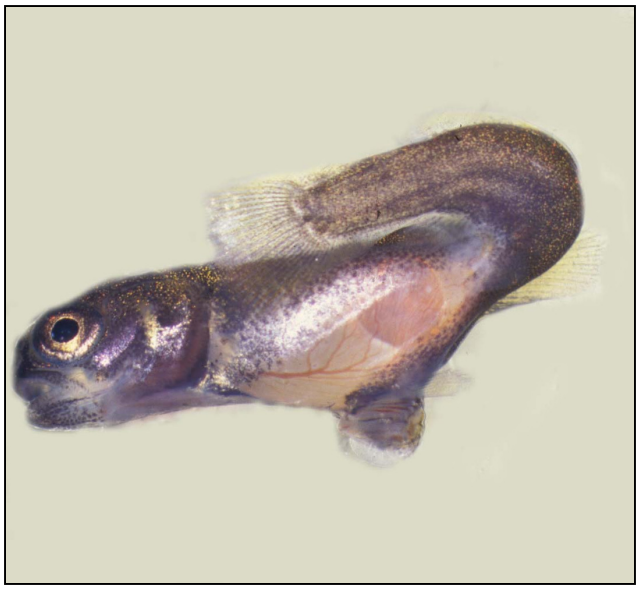


Figure 34. Rainbow trout (top) exhibiting craniofacial and operular deformities at 600 DTU following in ovo exposure to Deep Reach extracts (bottom fish is normal).

Figure 35. Effects of in ovo exposure to increasing doses of Three-Mile Pond extracts on rainbow trout at 600 DTU. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

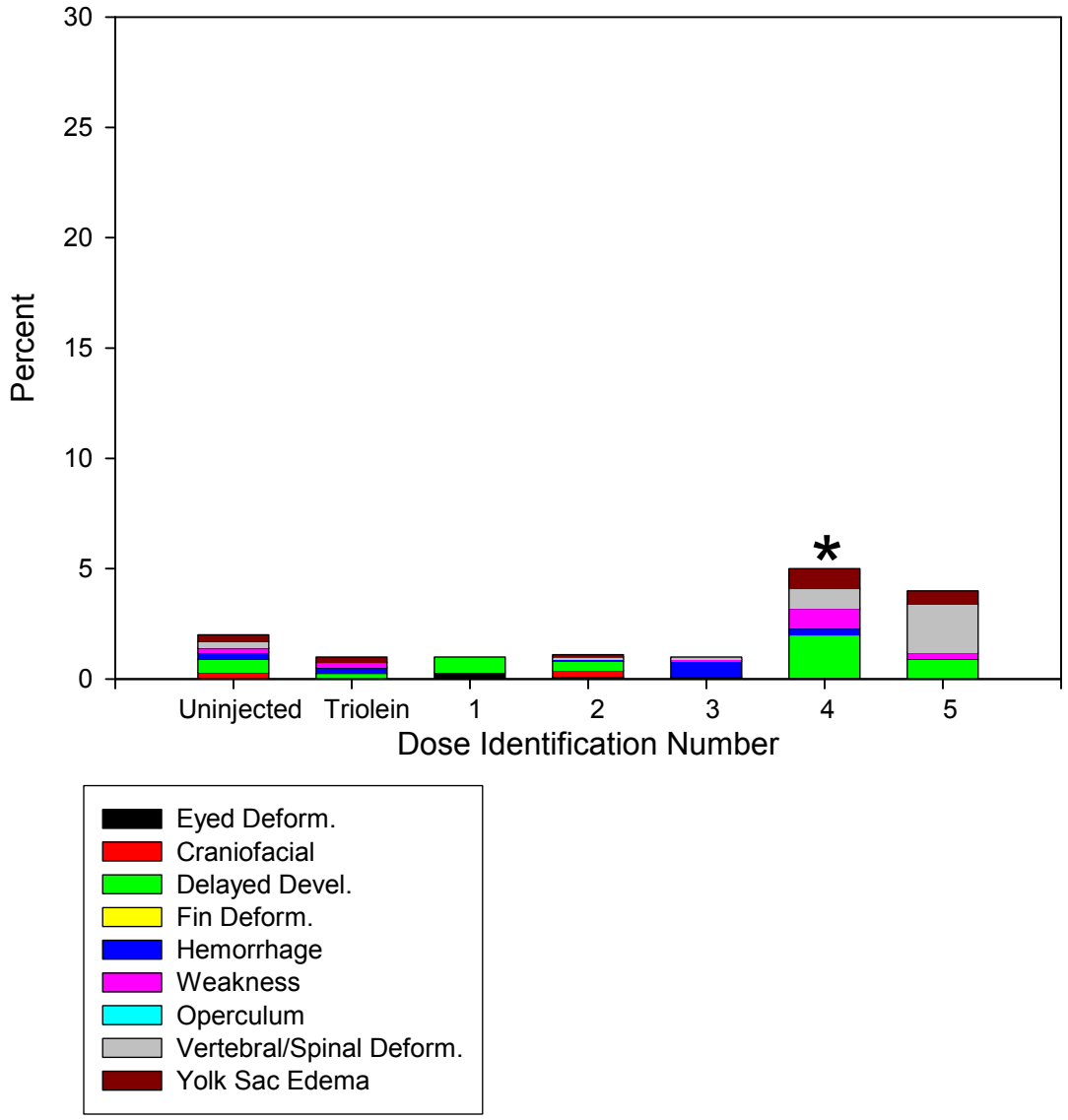


Figure 36. Effects of in ovo exposure to increasing doses of Rising Pond extracts on rainbow trout at 600 DTU. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

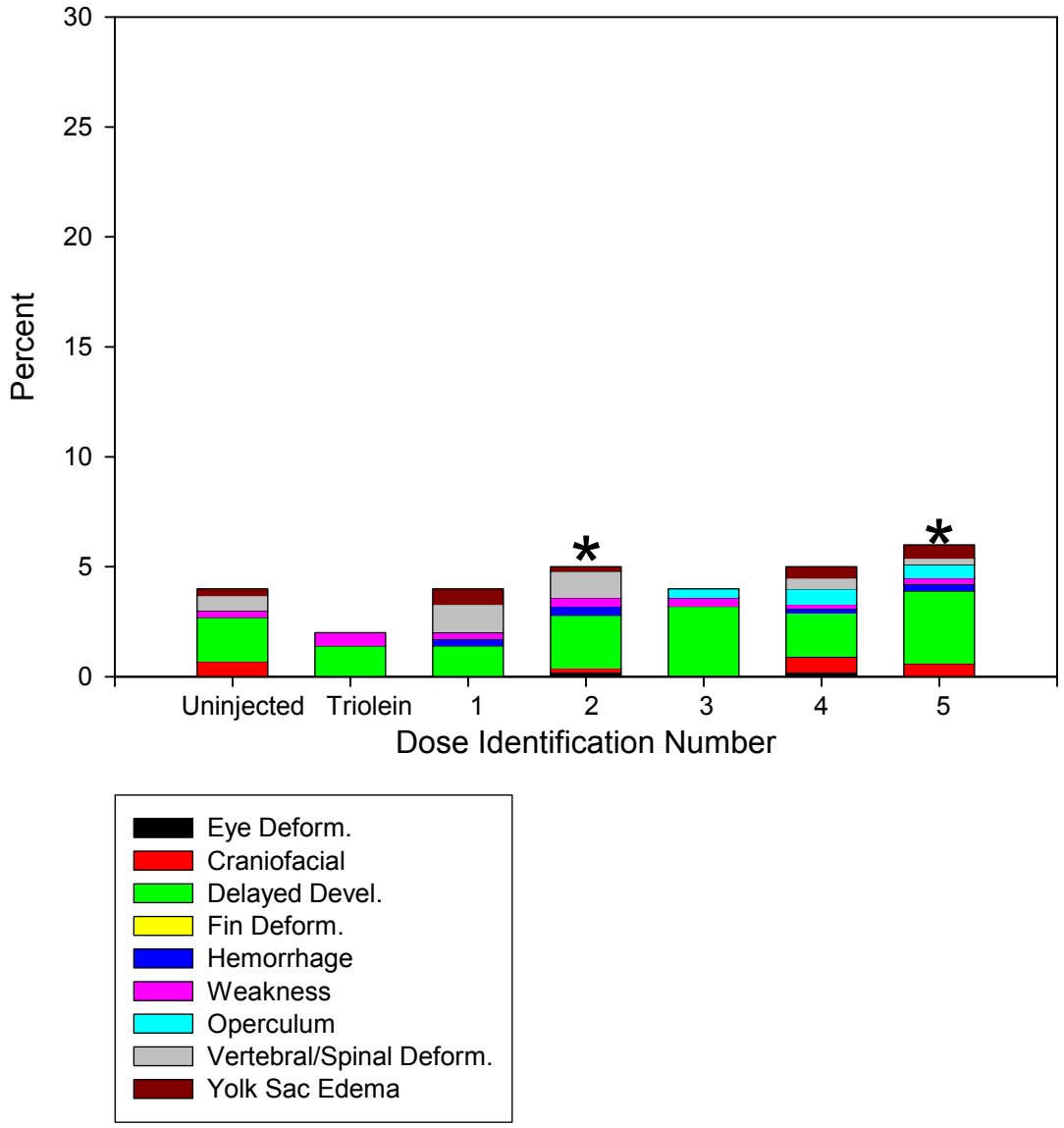


Figure 37. Effects of in ovo exposure to increasing doses of Woods Pond extracts on rainbow trout at 600 DTU. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

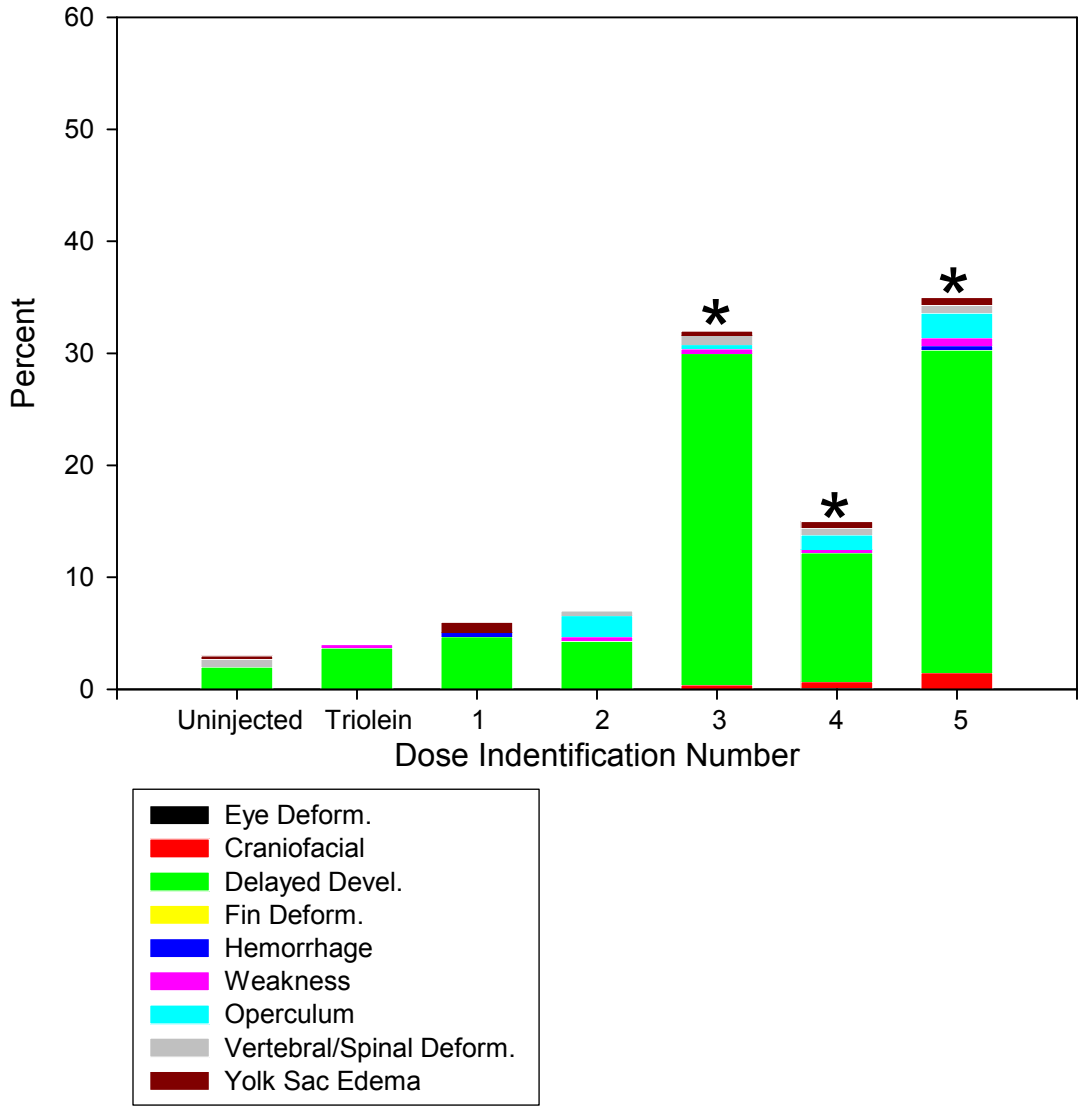


Figure 38. Effects of in ovo exposure to increasing doses of Deep Reach extracts on rainbow trout at 600 DTU. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

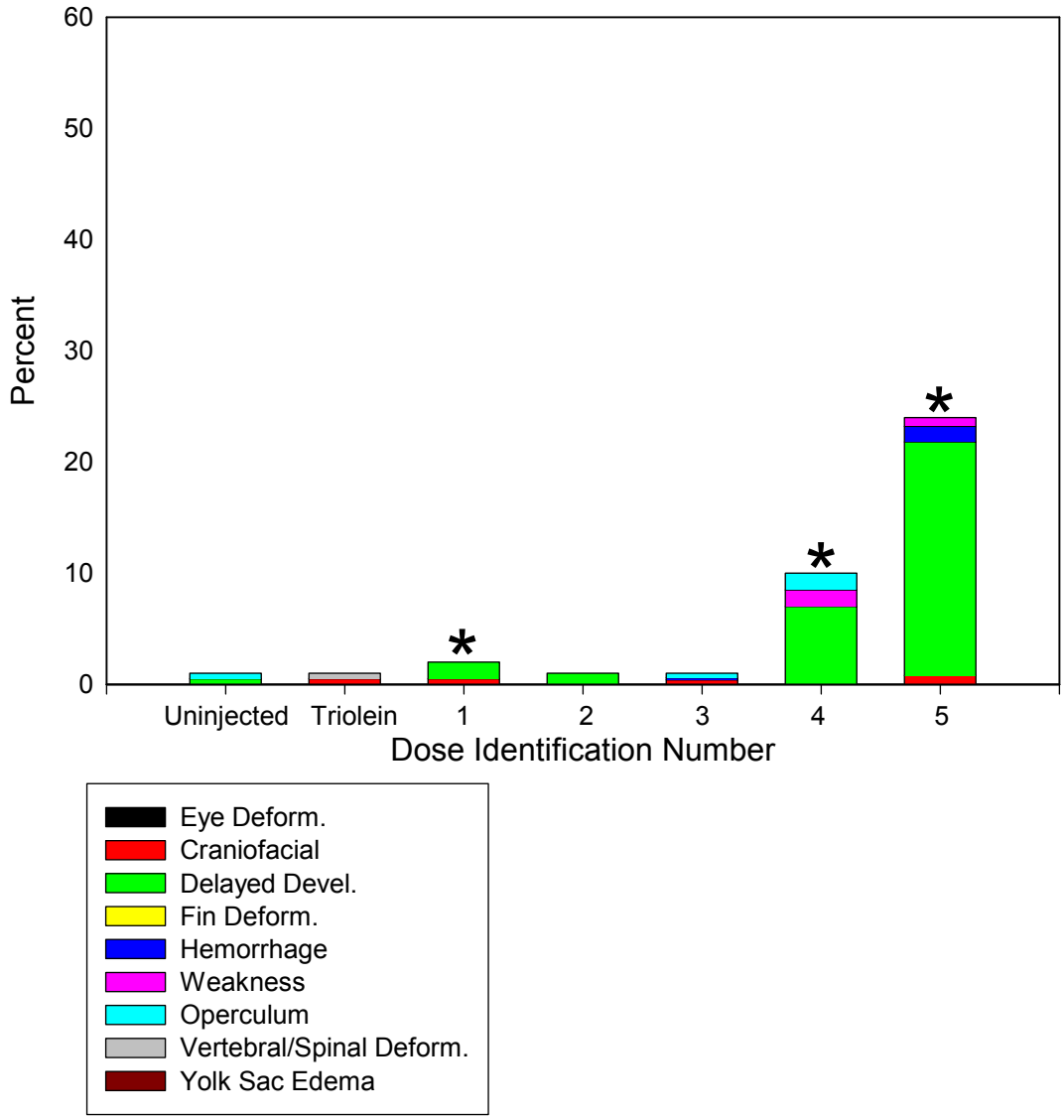


Figure 39. Effects of in ovo exposure to increasing doses of PCB 126 on rainbow trout at 600 DTU. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

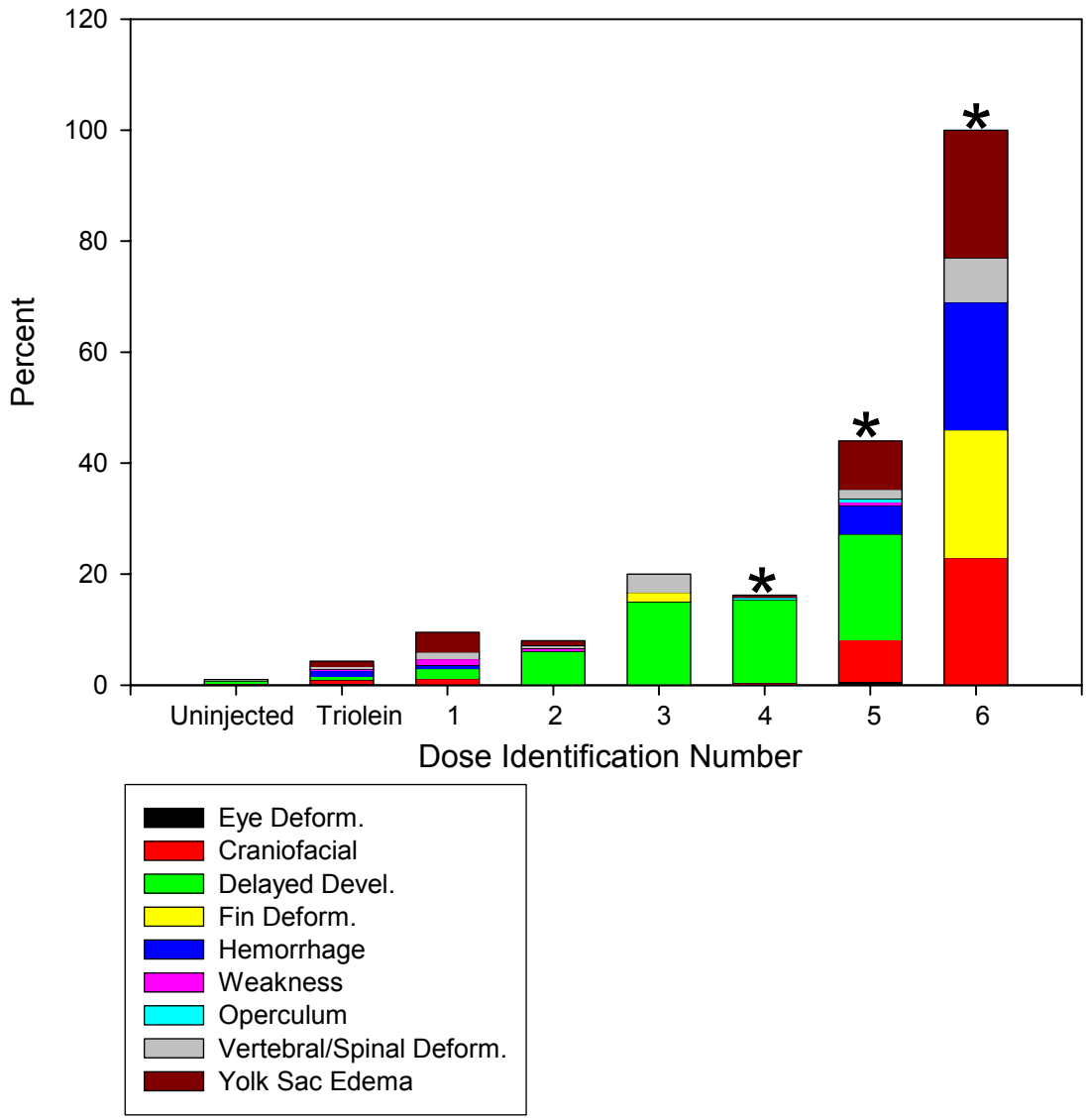


Figure 40. Effects of in ovo exposure to increasing doses of TCDD on rainbow trout at 600 DTU. Bar height indicates percentage of fish affected with one or more pathologies. Bands on bars indicate relative proportions of specific pathologies observed. Asterisks indicate significant differences from negative controls (uninjected and triolein).

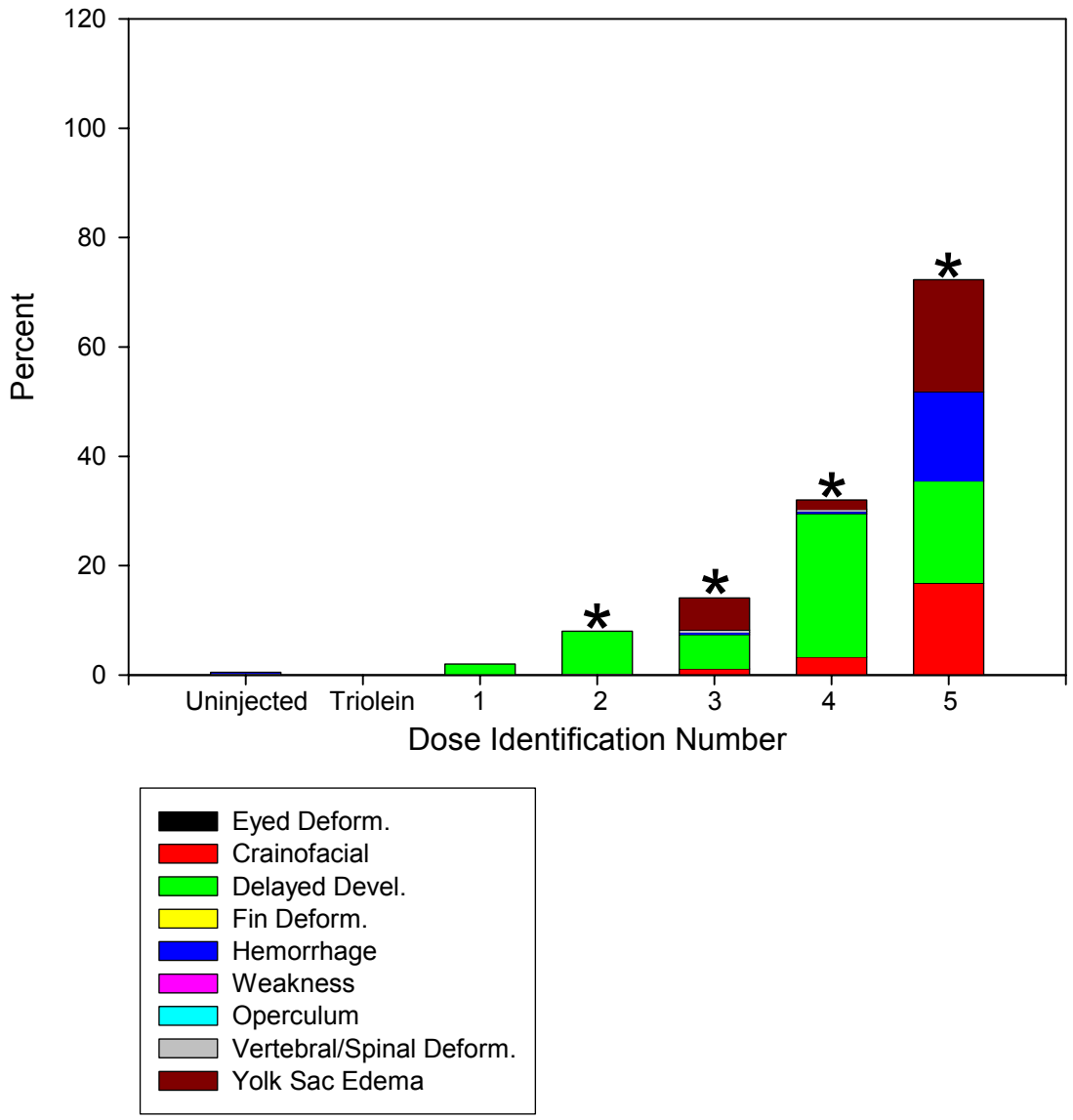


Figure 41. Dose-response curves of effects on rainbow trout at 600 DTU from exposure to increasing concentrations of Woods Pond extracts, Deep Reach extracts, PCB 126, or TCDD. Concentrations are represented by TEQs.

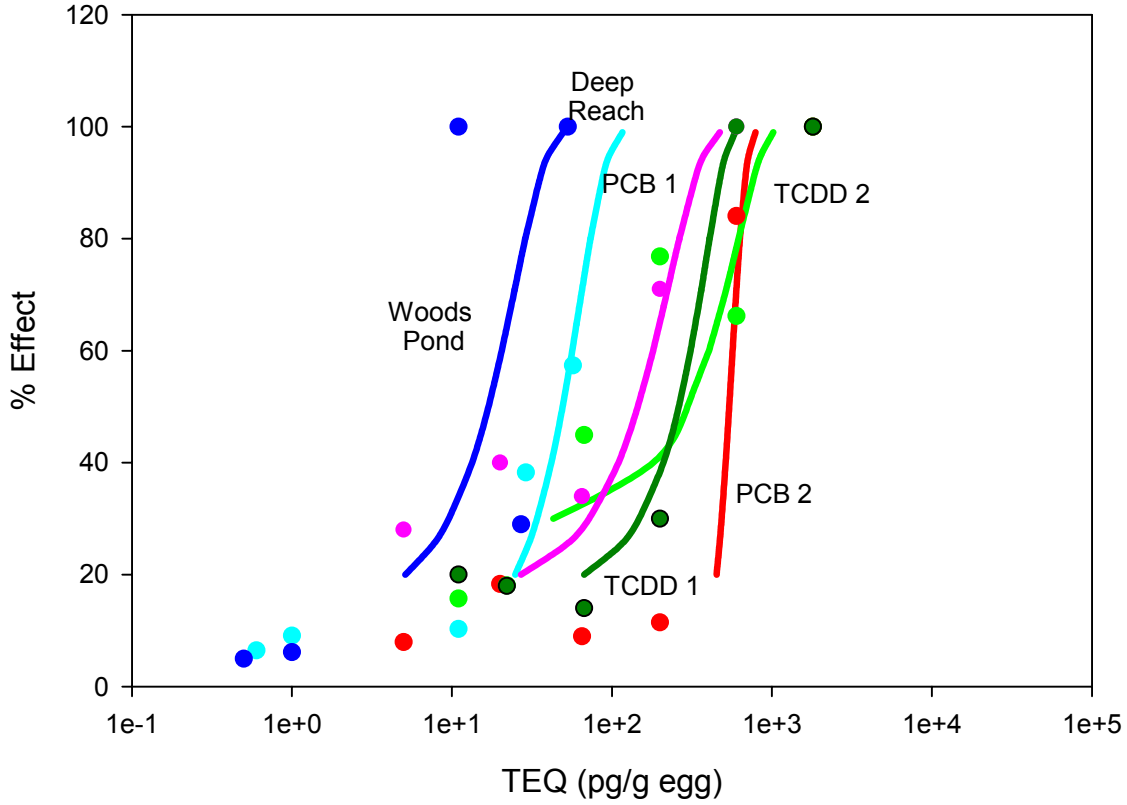


Table 53. ED50 values for rainbow trout at 600 DTU following in ovo injection with Woods Pond extracts, Deep Reach extracts, PCB 126, and TCDD. Values are expressed as pg TEQs per g egg.

	ED50	95% CI	Slope
Woods Pond (Trial 1)	18	15-21	107
Deep Reach (Trial 4)	49	35-64	136
PCB 126 (Trial 1)	194	153-235	96
PCB 126 (Trial 2)	521	432-610	315
2,3,7,8-TCDD (Trial 1)	360	296-423	136
2,3,7,8-TCDD (Trial 2)	464	347-581	80

Biochemistry:

Cytochrome P450 1A (CYP1A) induction occurred mainly in liver, brain, and gill tissue in rainbow trout (Figure 42). An increase in staining intensity and frequency was observed along an increasing dose gradient for those fish exposed to PCB 126. PCB, TCDD and Deep Reach fish all showed strong staining for the CYP 1A antibody at the highest dose tested. Also at the highest dose, Woods Pond showed moderate induction, while Rising Pond and Three-Mile Pond showed low induction (Table 54).

Figure 42. Cytochrome P450 1A staining in gills of rainbow trout at 600 DTU following in ovo exposure (500X magnification). A – Three-Mile Pond extracts, B – Woods Pond extracts, C – Deep Reach extracts, D – TCDD.

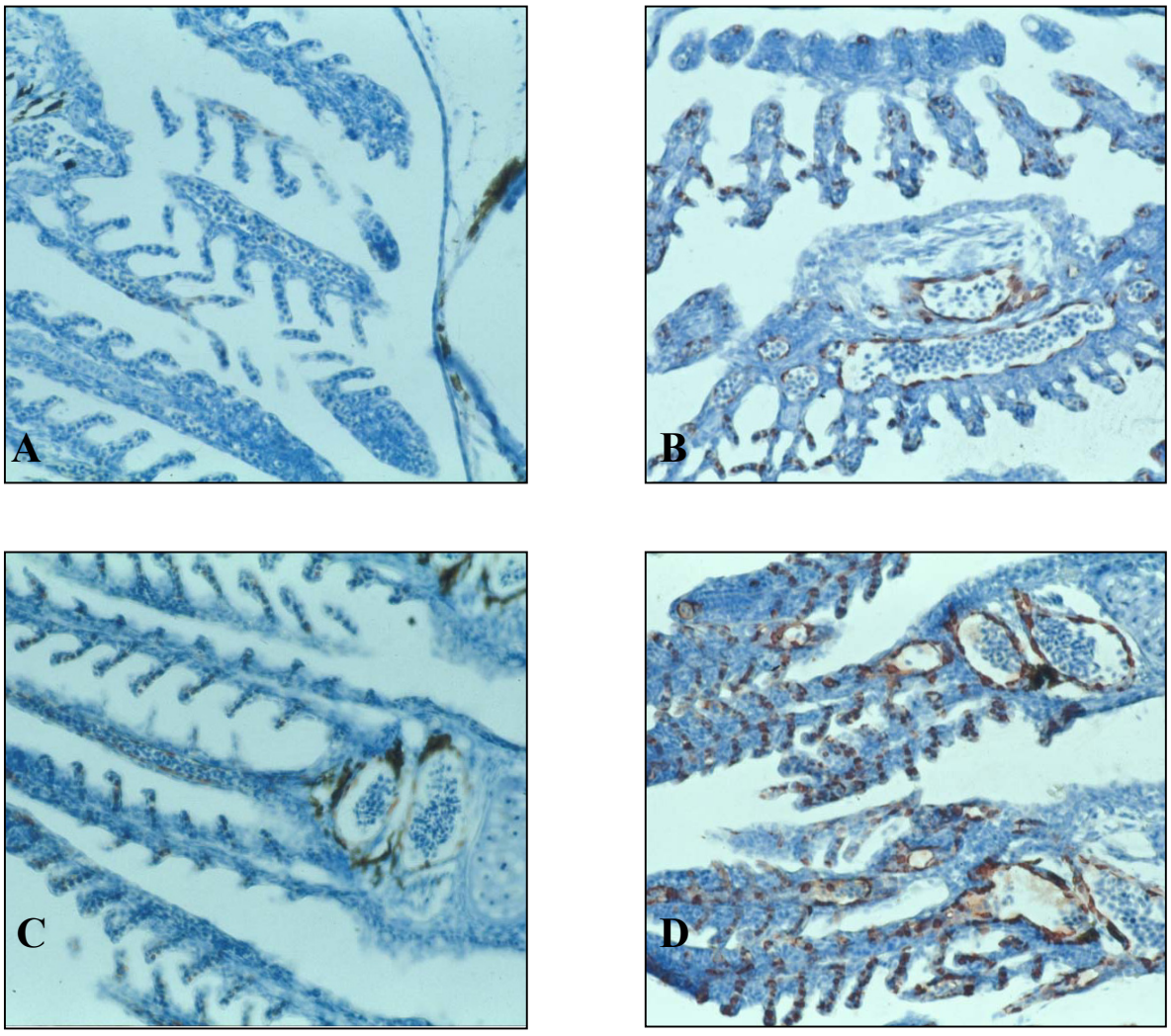


Table 54. Qualitative assessment of induction of cytochrome P450 1A in histologically prepared tissues of whole rainbow trout at 600 DTU. Results are presented for three doses representing low, medium, and high exposures. Asterisks represent the extent and intensity of the response (* = low, ** = moderate, and * = high).**

			Dose		
	n	**Triolein**	**1-2**	**3-4**	**5-6**
Three-Mile Pond	2	None	ND	ND	*
Rising Pond	6	None	None	*	*
Woods Pond	12	None	None	*	**
Deep Reach	6	None	None	***	***
PCB 126	5	None	*	**	***
2,3,7,8-TCDD	7	None	None	None	***

ND – No data.

6.0 SUMMARY AND CONCLUSIONS

6.1 Comparison of Results for Standards with the Literature

Point estimators (LD50s and ED50s) of mortality or dioxin-related toxicity were always greater for PCB 126 than TCDD across all species that were tested, indicative of the greater toxic potency of TCDD. The LD50 values for rainbow trout developed from this set of studies are greater than, but still consistent with other estimates reported in the literature (Walker and Peterson 1991; Zabel et al. 1995). The LD50 values for TCDD in early life stages of rainbow trout were in the range of 0.17 – 0.49 ng TCDD/g egg, dependent on the strain of rainbow trout (Walker and Peterson 1991; Zabel et al. 1995). The strain used in this set of experiments, Fish Lake strain, had not been tested previously. Our results indicated that the Fish Lake strain of rainbow trout was the least sensitive strain of trout examined to date. The LD50 value of TCDD in the Fish Lake strain was 1.1 ng TCDD/g egg (average of two values in Table 48), which is three-fold greater value than the LD50 for TCDD in the Arlee strain of rainbow trout (0.37 ng PCB 126/g egg; Walker and Peterson 1991). Similarly, the LD50 value for PCB 126 we determined in the Fish Lake strain of rainbow trout (average of two values, 200 ng PCB 126/g egg) was approximately three-fold greater than was observed in the Arlee strain (74 ng PCB 126/g egg; Walker and Peterson 1991). Thus, it appears that the Fish Lake strain of rainbow trout used for these studies was less sensitive than any other strain of rainbow trout tested to date with dioxin-like compounds. The fact that the LD50 values for both PCB 126 and TCDD were consistently greater (ie. Fish Lake strain is less sensitive) than other strains of rainbow trout, provides evidence that the LD50 values generated from these experiments were valid and not artifactual.

The validity of the LD50 values generated in these studies is further confirmed by evaluation of the relative potency difference between PCB 126 and TCDD. The relative potency factor (RPF) between PCB 126 and TCDD was calculated to be 0.005 (ie. TCDD LD50/PCB126 LD50 = (1.1 ng/g)/(200 ng/g) = 0.005). This relative potency difference between PCB 126 and TCDD is the same as has been observed previously in other strains of rainbow trout (Walker and Peterson 1991; Zabel et al. 1995) and equal to the TEF value proposed for fish by the World Health

Organization (WHO, van den Berg et al. 1998). The RPF for PCB 126 in largemouth bass was 0.0044, which is also fairly consistent with the TEF proposed for fish by the WHO. The RPFs for PCB 126 in medaka from this study at 100h post hatch and 15d post swim-up were 0.033 and 0.047, a factor of approximately ten less than the RPF observed in rainbow trout or largemouth bass. However, these RPF values are consistent with the literature value for medaka of 0.032 (Kim and Cooper 1998). The shapes of the dose-response curves for the compounds we tested were very similar both within and across species.

As expected, PCB 126 and TCDD induced cytochrome P450 in brain, gill, kidney and liver, although there were differences in strength of induction depending upon species.

In general, pathologies observed in PCB 126-exposed fish were also seen in TCDD-exposed fish and have been previously reported by others (Walker and Peterson 1991; Zabel et al. 1995; Henry et al. 1997; Cantrell et al. 1998; Kim and Cooper 1998; Toomey et al. 2001). These included: craniofacial deformities, edemas, hemorrhaging, weakness, delayed development, and problems associated with filling the swim bladder. Craniofacial deformities and edema seemed to occur more frequently in PCB 126-treated fish while delayed development was more commonly observed in TCDD-exposed fish. Fin and spinal deformities were associated with PCB 126 exposure, but not TCDD exposure. PCB 126-exposed largemouth bass uniquely exhibited incidences of the swim bladder being external to the body.

6.2 Species Comparisons

Overall, swim-up largemouth bass were the least sensitive to the acutely toxic effects of the reference toxicant standards and the extracts. Rainbow trout were the most sensitive to TCDD (LD50 of 1.1 ng/g egg), while largemouth bass and medaka had similar sensitivities (LD50s of 4.6 and 5.5 ng/g egg, respectively). Medaka were the most sensitive to PCB 126 (LD50 of 116 ng/g egg) followed by rainbow trout (LD50 of 200 ng/g egg), while largemouth bass were the least sensitive (1043 ng/g egg).

Largemouth bass were less sensitive to the effects of the standards and the Woods Pond extracts than were rainbow trout, based on LD50 and ED50 values estimated near the swim-up stage. The threshold value for effects that we estimated from the NOEL and LOEL for Woods Pond extracts is consistent with this lower sensitivity. The concentrations of PCB 126 and Deep Reach extracts required for induction of effects in medaka were similar to those for rainbow trout (when expressed as TEQs per g egg); medaka were less sensitive than rainbow trout to 2,3,7,8-TCDD. NOEL and LOEL estimated threshold values were consistent with these results for TCDD and PCB 126 (threshold values for medaka and rainbow trout were similar) and Deep Reach (no threshold value estimated for rainbow trout, but trout LOEL was similar to medaka threshold level). Similar to rainbow trout, the ED50 for PCB 126 for medaka was less than that for largemouth bass. The largemouth bass threshold value for Woods Pond was greater than for medaka. No NOEL and therefore no threshold value could be estimated for largemouth bass exposed to PCB 126, however the PCB 126 LOEL for bass was lower than the threshold value estimated for medaka exposed to PCB 126.

Cytochrome P450 induction in largemouth bass showed the weakest positive response with the CYP1A antibody of the three species we tested. Rainbow trout demonstrated both the strongest positive responses and a dose-dependent response to PCB 126. All species were induced somewhat by the standards, and all extracts induced a positive response in medaka and rainbow trout. Only Three-Mile Pond and Woods Pond extracts induced a positive response (albeit low) in largemouth bass.

Pathologies consistently observed in all species in response to reference toxicants or the extracts included craniofacial deformities, edema, and hemorrhaging. Largemouth bass and medaka both had a high incidence of problems associated with inflating the swim bladder. Rainbow trout and largemouth bass shared an elevated incidence of fin deformities not seen in medaka, while delayed development was commonly observed in rainbow trout and medaka. Opercular deformities were unique to rainbow trout, as the occurrence of the swim bladder outside the body was unique to largemouth bass.

6.3 Site Comparisons

Mortality was consistently low at swim-up for all species at the highest doses tested with Three-Mile and Rising Pond extracts. Mortality was also too low to calculate an LD50 after exposure of rainbow trout and largemouth bass to Deep Reach extracts, and after exposure of rainbow trout to Woods Pond extracts. Mortality data for medaka exposed to Woods Pond extracts met the test acceptance criteria, but were not suitable for calculation of an LD50. The incidence of pathologies was similar across sites and was below 40% at the highest dose for largemouth bass and rainbow trout, and below 25% for medaka. A dose-dependent response for pathologies was observed with all extracts and species except for largemouth bass and medaka exposed to Three-Mile Pond extracts. ED50 estimates for rainbow trout exposed to Woods Pond and Deep Reach extracts ranged from 18 to 49 pg TEQ per g egg. All LD50s and ED50s estimated for Woods Pond and Deep Reach extracts were lower than those for PCB 126 and 2,3,7,8-TCDD.

CYP1A induction was strongest in rainbow trout and medaka exposed to Deep Reach extract. A site-related response was observed in trout as follows: Deep Reach>Woods Pond>Rising and Three-Mile Ponds.

With a few notable exceptions, the same pathologies were observed at all sites. External swim bladder was never observed in fish exposed to Three-Mile or Rising Pond extracts. Additionally, Three-Mile Pond fish were never observed to have deformities of the fins or opercula. In general, fish exposed to Three-Mile Pond extract experienced the fewest pathological effects, followed by those fish exposed to Rising Pond extract. The incidence of pathologies was similar for fishes exposed to either Woods Pond and Deep Reach extracts.

A comparison of the induced pathologies resulting from exposure to PCB 126 and common to those induced by exposure to the extracts at equivalent TEQ concentrations for the three species is presented in Tables 55–57. Concordance with PCB 126 pathologies was greatest for largemouth bass and rainbow trout exposed to Woods Pond and Rising Pond extracts. Deep Reach-exposed fish shared comparatively fewer pathologies with PCB 126-exposed fish.

6.4 Consistency of Findings with Phase I Studies

Survival in swim-up largemouth bass in Phase II studies was generally higher than survival in Phase I studies. Differences in site-specific mortality were not notable in swim-up largemouth bass from Phase II studies, while in Phase I studies Deep Reach offspring showed poorer survival. However, mean mortality for Woods Pond offspring of females which averaged 128 pg TEQ/g ovarian tissue (Phase I results) fall on the predicted dose-response curve from the in ovo injection studies in Phase II (Figure 5) and estimated mean effects exhibited in the Phase I Woods Pond offspring are close to the corresponding predicted dose-response curve from Phase II (Figure 17).

Growth was not affected by exposure to extracts or standards in Phase II studies, but Woods Pond and Deep Reach offspring weighed less than Three-Mile or Rising Pond offspring in Phase I studies.

Positive cytochrome P450 induction was observed with all Housatonic River extracts and all Housatonic River sites in Phase II and Phase I studies. However, some differences in patterns off induction occurred. Induction in largemouth bass in Phase I was more consistent with medaka and rainbow trout in Phase II than with largemouth bass.

Pathologies observed in Phase II studies were consistent with pathologies observed in Phase I studies. Craniofacial, eye, fin, and opercula deformities, edema, and problems associated with swim bladder inflation were all observed in largemouth bass in Phase I studies and in largemouth bass, as well as in medaka and trout, in Phase II studies. Three-Mile Pond-exposed fish never experienced fin or opercula deformities and, in general, the types and incidence of pathologies were low for Three-Mile Pond fish in both studies. It is notable that external swim bladder was observed in Woods Pond- and Deep Reach-exposed fish in Phase II studies and in fish from all Housatonic River sites in Phase I studies.

The consistencies among effects observed in Phase I and Phase II studies strongly suggest that components of the dosing solutions prepared from tissues of largemouth bass collected from Housatonic River sites contributed to the observed toxicity in both studies. The correspondence between effects in fish exposed to the dosing solutions and effects in fish exposed to PCB 126 and TCDD in Phase II studies, and the consistency of those results with literature values, strongly suggests that compounds with dioxin-like properties contributed to the toxicity observed in both studies. The high levels of PCBs present in tissues of largemouth bass collected from Housatonic River sites, and subsequently present in the dosing solutions prepared from those tissues, strongly suggest that PCBs substantially contributed to the toxicity observed in both studies.

Table 55. Concordance of gross pathologies observed in largemouth bass exposed to Housatonic River extracts and TCDD with those observed in largemouth bass exposed to PCB 126 at comparable TEQs*. Phase I results shown for comparison.

	PCB 126						
Treatment	**Craniofacial Deformity**	**Vertebral/Spinal Deformity**	**Uninflated Swim Bladder**	**Hemorrhage**	**Pericardial Edema**	**Peritoneal Edema**	**Delayed Development**
Three-Mile Pond**							
Three-Mile Pond (Phase I)		X			X		
Rising Pond		X	X			X	
Rising Pond (Phase I)	X	X	X		X	X	
Woods Pond	X	X	X	X	X		
Woods Pond (Phase I)	X	X	X		X		
Deep Reach			X				
Deep Reach (Phase I)	X	X	X		X		
2,3,7,8-TCDD			X		X	X	X

* **TEQs based upon PCB contributions only for Housatonic River extracts and sites.**
** **Since no comparable dose for Three-Mile Pond exists, results at the highest dose (6.0 pgTEQ/g egg) are shown.**

Table 56. Concordance of gross pathologies observed in medaka exposed to Housatonic River extracts and TCDD with those observed in medaka exposed to PCB 126 at comparable TEQs*.

Treatment	PCB 126		
	Uninflated Swim Bladder	Partially-Inflated Swim Bladder	Larval Weakness
Three-Mile Pond			
Rising Pond	X	X	X
Woods Pond	X		
Deep Reach	X	X	X
2,3,7,8-TCDD	X	X	

*** TEQs based upon PCB contributions only for Housatonic River extracts and sites.**
**** Since no comparable dose for Three-Mile Pond exists, results at the highest dose (6.0 pg TEQ/g egg) are shown.**

Table 57. Concordance of gross pathologies observed in rainbow trout exposed to Housatonic River extracts and TCDD with those observed in rainbow trout exposed to PCB 126 at comparable TEQs*.

Treatment	PCB 126				
	Craniofacial Deformity	Vertebral/Spinal Deformity	Opercular Deformity	Yolk Sac Edema	Delayed Development
Three-Mile Pond		X			
Rising Pond	X	X	X	X	
Woods Pond	X	X		X	X
Deep Reach	X				X
2,3,7,8-TCDD	X	X		X	X

*** TEQs based upon PCB contributions only for Housatonic River extracts and sites.**
**** Since no comparable dose for Three-Mile Pond exists, results at the highest dose (5 pgTEQ/g egg) are shown.**

7.0 PERSONNEL AND ACKNOWLEDGMENTS

7.1 Project Personnel

The following individuals participated in conducting the studies described in this report.

Alan Allert	*Technical Assistance*
Mandy Annis	*Histology*
Jesse Arms	*Chemical Analysis*
Sean Birke	*EROD Analysis*
James Candrl	*Fish Culture*
Paulo Carvalho	*Technical Assistance*
Rachel Claunch	*Technical Assistance*
Tricia Dietrich	*Technical Assistance*
Kathy Echols	*Chemical Analysis*
Eugene Greer	*Fish Culture*
Chris Ivey	*Fish Culture*
Allen Johnson	*Fish Culture*
John Meadows	*Chemical Analysis*
Tim McTague	*Chemical Analysis*
Eric Nelson	*Technical Assistance*
Diane Nicks	*EROD Analysis*
Carl Orazio	*Chemical Analysis*
Lori Patton	*Technical Assistance*
Aaron Roberts	*Technical Assistance*
Shawn Sanders	*Fish Culture*
Mike Tanner	*Chemical Analysis*
Jeff Whyte	*EROD Analysis*

7.2 Acknowledgments

The authors would like to thank all of the project personnel (listed above) for their high level of performance over the course of these studies. The success of these studies is a direct result of each of their contributions to this team effort. In addition to the persons directly involved in this project, there were a number of other people from the CERC that we would like to thank for their important contributions to the success of this project. Julia Towns conducted literature searches and collected numerous library references. Chris Schmitt, Jim Fairchild and the Ecology Section provided information and resources for the outdoor pond aspects of this study, as well as the loan of equipment. The Maintenance Section, including Larry Farley, provided valuable assistance in construction during the course of these studies.

Critical assistance was also provided by people from outside of our laboratory. Any success which has come from this study, began with the efforts of Joe McKeon, U.S. Fish and Wildlife Service, Laconia Office of Fisheries Assessment. We also are thankful for the assistance of Craig Gemming and Robin Tillitt of the Missouri Department of Conservation for collection of largemouth bass brood stock. Personnel of the Genoa National Fish Hatchery, U.S. Fish and Wildlife Service, also assisted by providing brood stock. We also thank John Stegeman of the Woods Hole Oceanographic Institute for providing CYP1A antibody.

Funding for this work came from the U.S. Environmental Protection Agency, through a Cooperative Agreement with the U.S. Fish and Wildlife Service and we thank Ken Carr and Susan Svirsky for their special effort to include these studies in the ecological assessment of the Housatonic River.

8.0 LITERATURE

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CERC SOP P.195. Capillary gas chromatography with electron capture detection procedure for congener specific polychlorinated biphenyl analysis. Columbia Environmental Research Center, USGS, Columbia, Missouri.

CERC SOP P.459. Organochlorine pesticide analysis by high resolution capillary gas chromatography with electron capture detection. Columbia Environmental Research Center, USGS, Columbia, Missouri.

CERC SOP P.461. Extraction of animal tissues for residue analysis and percent lipid determination. Columbia Environmental Research Center, USGS, Columbia, Missouri.

CERC SOP P.464. Use of high performance gel permeation chromatography in sample preparatory applications. Columbia Environmental Research Center, USGS, Columbia, Missouri.

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CERC SOP P.525. Culture of brine shrimp, *Artemia sp.*, from dehydrated cysts as a food source for larval fish. Columbia Environmental Research Center, USGS, Columbia, Missouri.

CERC SOP P.483. Quality control guidelines and criteria for gas chromatographic data processing. Columbia Environmental Research Center, USGS, Columbia, Missouri.

CERC SOP P.535. Procedures for injection of newly fertilized eggs with whole fish extracts and dioxin PCB standards. Columbia Environmental Research Center, USGS, Columbia, Missouri.

CERC SOP P.539. Hematoxylin and eosin (H&E) staining of paraffin sections. Columbia Environmental Research Center, USGS, Columbia, Missouri.

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9.0 APPENDIX

Residue analysis of largemouth bass brood fish (see Tables below).
PCBs, PCDDs, PCDFs and Pesticides in Dosing Solutions Created from Tissues of Largemouth Bass from the Housatonic River (see attached report).

Inorganic Chemicals in Brood Fish (mg/Kg, wet weight) (detection limits in parentheses)								
Analyte	**Little Dixie 1**	**Little Dixie 2**	**Little Dixie 2 Duplicate**	**Little Dixie 3**	**Little Dixie 3 Duplicate**	**Genoa 1**	**Genoa 2**	**Genoa 3**
% Moisture	15	45	45	67	67	53	64	65
Al	14.318 (0.871)	13.303 (0.528)	13.645 (0.546)	342.474 (0.328)	335.619 (0.332)	772.796 (0.464)	947.117 (0.357)	821.333 (0.344)
As	0.914 (0.436)	0.507 (0.264)	0.696 (0.273)	0.792 (0.164)	0.798 (0.166)	0.902 (0.232)	1.008 (0.179)	0.860 (0.172)
B	1.062 (0.436)	1.120 (0.264)	0.746 (0.273)	0.776 (0.164)	1.262 (0.166)	1.139 (0.232)	0.914 (0.179)	0.940 (0.172)
Ba	0.871 (0.871)	0.528 (0.528)	0.546 (0.546)	6.490 (0.328)	6.674 (0.332)	6.311 (0.464)	7.235 (0.357)	5.988 (0.344)
Be	0.026 (0.026)	0.016 (0.016)	0.016 (0.016)	0.010 (0.010)	0.010 (0.010)	0.014 (0.014)	0.013 (0.011)	0.016 (0.010)
Cd	0.087 (0.087)	0.053 (0.053)	0.055 (0.055)	0.036 (0.033)	0.033 (0.033)	0.070 (0.046)	0.084 (0.036)	0.074 (0.034)
Cr	1.201 (0.087)	0.682 (0.053)	0.692 (0.055)	0.759 (0.033)	0.758 (0.033)	772.796 (0.046)	2.383 (0.036)	2.212 (0.034)
Cu	5.286 (0.087)	2.706 (0.053)	2.656 (0.055)	1.787 (0.033)	1.757 (0.033)	0.902 (0.046)	2.234 (0.036)	1.997 (0.034)
Fe	53.833 (4.357)	26.031 (2.638)	26.574 (2.732)	295.032 (1.640)	290.262 (1.660)	592.062 (2.322)	728.506 (1.786)	638.804 (1.722)
Hg	0.044 (0.044)	0.026 (0.026)	0.029 (0.027)	0.016 (0.016)	0.017 (0.017)	0.023 (0.023)	0.018 (0.018)	0.017 (0.017)
Mg	722.077 (4.357)	518.664 (2.638)	511.576 (2.732)	347.519 (1.640)	354.115 (1.660)	585.667 (2.322)	450.690 (1.786)	425.399 (1.722)
Mn	8.952 (0.261)	9.183 (0.158)	9.065 (0.164)	19.563 (0.098)	19.806 (0.100)	65.603 (0.139)	72.687 (0.107)	55.950 (0.103)
Mo	0.871 (0.871)	0.528 (0.528)	0.546 (0.546)	0.328 (0.328)	0.332 (0.332)	0.464 (0.464)	0.357 (0.357)	0.344 (0.344)
Ni	0.957 (0.087)	0.463 (0.053)	0.453 (0.055)	0.867 (0.033)	0.898 (0.033)	2.257 (0.046)	1.689 (0.036)	1.515 (0.344)
Pb	0.044 (0.044)	0.026 (0.026)	0.027 (0.027)	0.016 (0.016)	0.017 (0.017)	0.061 (0.023)	0.109 (0.018)	0.160 (0.017)
Se	2.708 (0.436)	1.835 (0.264)	1.611 (0.273)	0.609 (0.164)	0.626 (0.166)	1.117 (0.232)	0.751 (0.179)	0.743 (0.172)
Sr	0.839 (0.087)	0.491 (0.053)	0.490 (0.055)	6.779 (0.033)	6.817 (0.033)	4.993 (0.046)	4.931 (0.036)	4.241 (0.034)
V	0.087 (0.087)	0.053 (0.053)	0.055 (0.055)	0.652 (0.033)	0.670 (0.033)	1.491 (0.046)	1.808 (0.036)	1.548 (0.034)
Zn	113.691 (0.871)	58.756 (0.528)	57.849 (0.546)	23.339 (0.328)	335.619 (0.332)	37.336 (0.464)	27.264 (0.357)	26.073 (0.344)

Organic Chemicals in Brood Fish (mg/Kg, wet weight) (detection limits in parentheses)							
Analyte	**Little Dixie 1**	**Little Dixie 2**	**Little Dixie 3**	**Little Dixie 3 Duplicate**	**Genoa 1**	**Genoa 2**	**Genoa 3**
% Moisture	74.5	74.9	80.4	80.4	76.6	76.2	74.3
% Lipid	5.49	6.48	5.71	5.71	6.33	5.98	6.06
HCB	0.000983 (0.000546)	0.000639 (0.000392)	0.000644 (0.000311)	0.000407 (0.000284)	0.000343 (0.000248)	0.000456 (0.00024)	0.00034 (0.000118)
alpha BHC	0.000501 (0.000501)	0.000502 (0.000502)	0.000356 (0.000356)	0.000349 (0.000349)	0.000254 (0.000254)	0.000242 (0.000242)	0.0002 (0.0002)
gamma BHC	0.00063 (0.00063)	0.000632 (0.000632)	0.000751 (0.000448)	0.00044 (0.00044)	0.000319 (0.000319)	0.000305 (0.000305)	0.000251 (0.000251)
oxychlordane	0.00192 (0.00192)	0.00283 (0.00283)	0.0017 (0.0017)	0.000747 (0.000747)	0.000994 (0.000994)	0.00113 (0.00113)	0.000813 (0.000813)
gamma chlordane	0.000362 (0.000174)	0.000364 (0.000254)	0.000345 (0.000138)	0.000377 (0.000221)	0.000236 (0.0000653)	0.000197 (0.000119)	0.000326 (0.0000744)
alpha chlordane	0.00172 (0.000203)	0.00152 (0.000297)	0.00155 (0.000161)	0.00147 (0.000258)	0.00104 (0.0000761)	0.00115 (0.000138)	0.00104 (0.0000868)
trans-nonachlor	0.00731 (0.000102)	0.00304 (0.000104)	0.00605 (0.0000858)	0.00625 (0.0000884)	0.00204 (0.0000875)	0.00246 (0.000134)	0.00272 (0.0000734)
o,p'-DDE	0.000249 (0.000249)	0.000249 (0.000249)	0.00016 (0.00016)	0.0002 (0.0000955)	0.000141 (0.000141)	0.000116 (0.000116)	0.000153 (0.0000996)
P,p'-DDE	0.0262 (0.0000499)	0.012 (0.000229)	0.0236 (0.0000502)	0.0236 (0.0000248)	0.0126 (0.0000248)	0.0156 (0.0000233)	0.0169 (0.0000322)
O,p'-DDD	0.000201 (0.000119)	0.000131 (0.000125)	0.000122 (0.0000859)	0.000161 (0.000161)	0.000135 (0.0000901)	0.000197 (0.0000717)	0.000166 (0.000103)
P,p'-DDD	0.00145 (0.000123)	0.000868 (0.000129)	0.00136 (0.0000892)	0.00141 (0.000167)	0.00162 (0.0000936)	0.00227 (0.0000745)	0.00228 (0.000107)
O,p'-DDT	0.000369 (0.000136)	0.000143 (0.000143)	0.000283 (0.0000985)	0.000311 (0.000185)	0.000122 (0.000103)	0.000167 (0.0000822)	0.000192 (0.000118)
P,p'-DDT	0.000816 (0.000168)	0.000176 (0.000176)	0.00102 (0.000122)	0.000894 (0.000228)	0.000265 (0.000128)	0.000266 (0.000102)	0.000312 (0.000146)
mirex	0.000329 (0.000329)	0.00034 (0.00034)	0.000163 (0.000163)	0.000252 (0.000252)	0.000163 (0.000163)	0.000178 (0.000178)	0.000103 (0.000103)
PCB-1242	0.00163 (0.00163)	0.00139 (0.00139)	0.000703 (0.000703)	0.000804 (0.000804)	0.00158 (0.000696)	0.00179 (0.000877)	0.00237 (0.000505)
PCB-1248	0.00486 (0.000961)	0.00197 (0.000927)	0.00523 (0.000668)	0.00578 (0.000605)	0.00932 (0.000578)	0.0126 (0.000655)	0.0139 (0.000272)
PCB-1254	0.0336 (0.0029)	0.0139 (0.00241)	0.0291 (0.00128)	0.0262 (0.00249)	0.0424 (0.00125)	0.053 (0.00179)	0.0614 (0.00123)
PCB-1260	0.0732 (0.000626)	0.0313 (0.000664)	0.0575 (0.000327)	0.054 (0.000704)	0.0214 (0.000579)	0.0267 (0.000403)	0.0315 (0.00042)
PCB-TOTAL	0.112 (0.0029)	0.0472 (0.00241)	0.0918 (0.00128)	0.0859 (0.00249)	0.0746 (0.00125)	0.0941 (0.00179)	0.109 (0.00123)
toxaphene	0.0148 (0.0148)	0.0141 (0.0141)	0.00698 (0.00698)	0.00825 (0.00825)	0.00408 (0.00408)	0.00513 (0.00513)	0.00593 (0.00593)
heptachlor epoxide	0.000731 (0.000115)	0.000796 (0.0000944)	0.000669 (0.0000663)	0.000538 (0.0000673)	0.000157 (0.0000629)	0.000114 (0.0000522)	0.000195 (0.0000471)
dieldrin	0.0078 (0.000133)	0.00821 (0.000109)	0.00787 (0.0000763)	0.00754 (0.0000774)	0.000839 (0.0000724)	0.000728 (0.00006)	0.00101 (0.0000542)
endrin	0.000224 (0.000224)	0.000183 (0.000183)	0.000128 (0.000128)	0.00013 (0.00013)	0.000122 (0.000122)	0.000101 (0.000101)	0.0000913 (0.0000913)

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April 10, 2002

PCBs, PCDDs, PCDFs and Pesticides in Dosing Solutions Created from Tissues of Largemouth Bass from the Housatonic River

USGS Report
Section 31 Project # 3307-70L1E

Analytical Investigation by the Organic Chemistry Section

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Submitted to

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Fish Reproductive Health Assessment in PCB Contaminated Regions of the Housatonic River, MA, USA; Investigation of Causal Linkages Between PCBs and Fish Health

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I. Project Summary:

The Housatonic River runs from Vermont though Massachusetts and then into Connecticut, and Long Island Sound where it empties into the Atlantic Ocean. For about 50 years, a General Electric facility in Pittsfield MA, adjacent to the river, produced approximately 39,000 pounds of PCBs for use in transformers. PCB-contaminated wastewater from GE was released into the Housatonic River. The sediments at downstream sites, including those at Woods Pond and Deep Reach Pond remain contaminated with PCBs despite the factory being closed since the 1977 PCB ban.

The contaminant residue information provided in this report is part of a larger study concerned with assessing fish reproductive health in PCB contaminated regions of the Housatonic River. The larger study investigates causal linkages between polyhalogenated hydrocarbons (PHHs) and fish health. Levels of PHHs were determined in the fish tissue, ovaries, and dosing solutions to complement the information gained from the embryotoxicity studies.

Reported herein are the results of the trace-level analyses of the following contaminants in dosing solutions created from largemouth bass tissue:

Persistent organochlorine pesticides (OC pesticides),
Polychlorinated dibenzo-p-dioxins and furans (PCDDs/PCDFs),
Polychlorinated biphenyls (PCBs), and
Non-*ortho* PCBs (n-PCBs)

II. Summary of Analytical Methods

1. Sample Preparation

A mass of 15-18 kg of largemouth bass (*Micropterus salmoides*) was collected just prior to spawning from each of the following four sites on the Housatonic River: Three-Mile Pond (reference area), Rising Pond, Woods Pond, and Deep Reach Pond. Figure 1 shows the procedure used to prepare the dosing solution extract. After removal of the ovaries from the females, composites of ground fish were created for each site. The tissue samples from each site were dehydrated by addition of anhydrous sodium sulfate. No method recovery compounds were added. The masses of tissue used to create the dosing solutions are reported in the attached tables of data. The four large composites were column extracted with methylene chloride and the solvent was removed from the lipid by rotoevaporation. Contaminants were isolated from the lipids using large-scale dialysis (LSD) through a polyethylene membrane. Dialytic recoveries, as checked by ^{14}C-labeled 2,5,2',5'-tetrachlorobiphenyl and mirex, averaged 77%. Dialysates were composited and a small portion of the extract (0.1%) was used to determine percent lipid (3). Any remaining co-dialyzed lipids were removed from the extracts by reactive column cleanup on acid- and base-treated silica gels and adsorbent chromatography on activated silica gel (4). These were further purified by high performance gel permeation chromatography (HPGPC) (1,2,5). Dosing solutions were

prepared containing graded doses of the isolated contaminants using triolein as the diluent and injection vehicle (3). The dosing solutions were provided to the Biochemistry Group whereby they were injected into large mouth bass eggs for subsequent observation of toxic responses.

The procedure described above produces purified dosing solutions containing 40% of the mass of PCBs, PCDDs, and PCDFs present in the original tissue samples. Triplicate portions of the final purified dialysates from HPGPC were removed for analysis using procedures outlined in Figure 1. Appropriate dilutions using estimates of total PCBs from HPGPC with UV detection were made in order to prevent overloading the porous graphitic carbon (PGC) column. These diluted samples were fractioned on PGC (6) into the following fractions:

PGC 1 *ortho*-chlorinated PCB congeners
- Analysis by GC/ electron-capture detection (ECD)

PGC 2 non-*ortho*-chlorinated PCBs
- Analysis by GC/ high-resolution mass spectrometry (GC/HRMS)

PGC 3 polychlorinated dibenzo-*p*-dioxins and -furans (PCDDs/PCDFs)
- Analysis by GC/ high-resolution mass spectrometry (GC/HRMS)

PGC 3 was further purified on basic alumina before the GC/HRMS analysis (7).

The following quality control samples were incorporated into the various analyses:

1 Procedural Blank
1 Procedural Spike

The final purified dialysates and QC samples had the following method recovery compounds added to them to monitor method recoveries through the HPLC carbon and alumina cleanup steps.

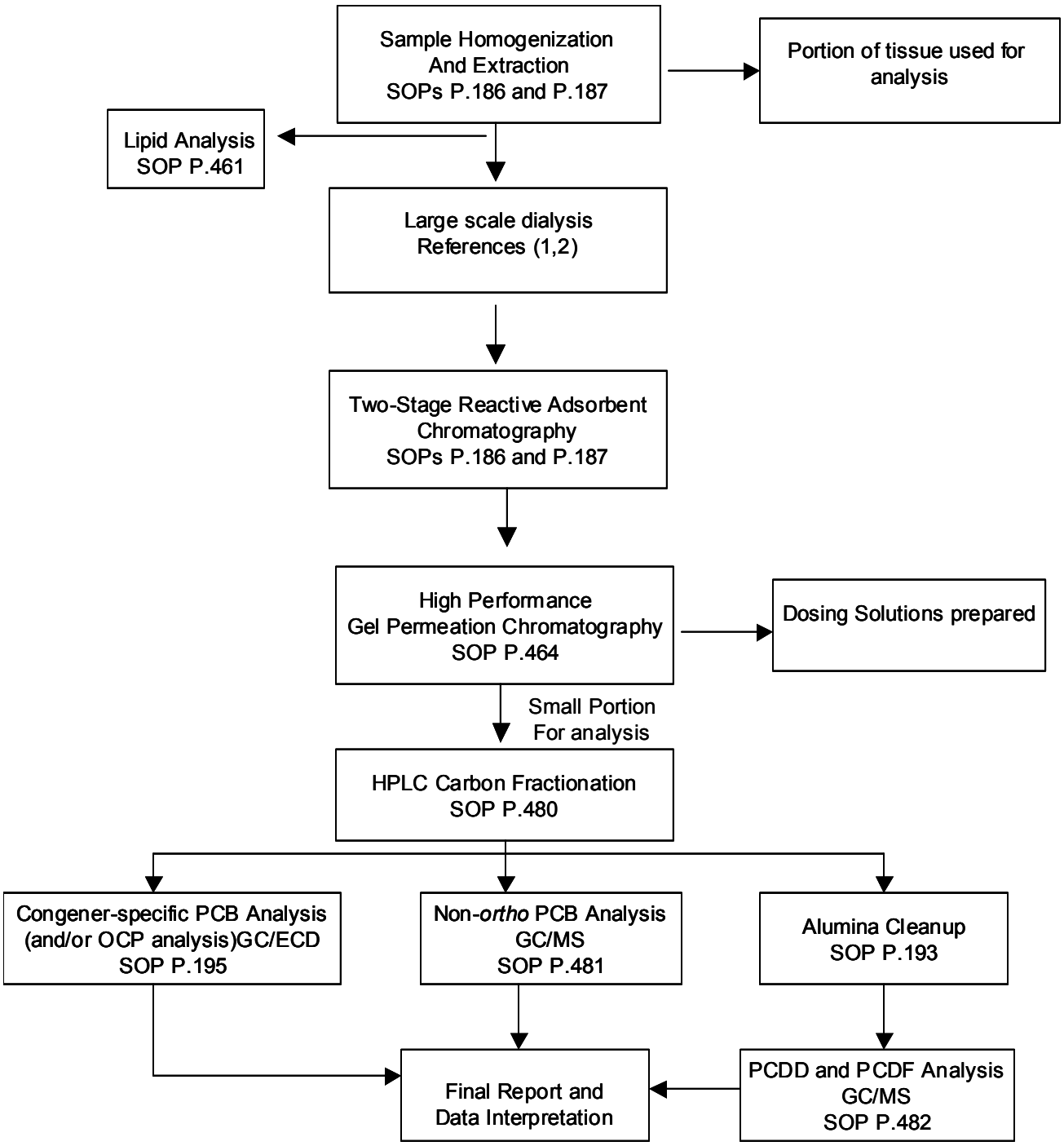
PCB 029 (2,4,6-trichlorobiphenyl)
PCB155 (2,2',4,4',6,6'-hexachlorobiphenyl)
PCB 204 (2,2',3,4,4',5,6,6'-octachlorobiphenyl)
Four ^{13}C-labeled non-*ortho* PCB congeners
Seventeen ^{13}C-labeled 2,3,7,8 substituted dioxin/furans

Procedural spikes in the PHH portion of the analysis also received:

PCBs (mixed Aroclors 1242, 1248, 1254, 1260)
Native (^{12}C) dioxin and furan congeners

PCB 029, a trichlorobiphenyl, is representative of more volatile early eluting PCBs (Cl_1 - Cl_3); PCB 155, a hexachlorobiphenyl, is representative of mid-range eluting congeners (Cl_4 - Cl_6); and PCB 204, an octachlorobiphenyl, is less volatile and representative of later eluting PCBs (Cl_7 - Cl_{10}).

Figure 1. Analytical Scheme for, PCB Congeners, OC Pesticides, PCDDs, and PCDFs in Housatonic Dosing Solutions



OC pesticides were quantified in PGC1 fraction (No other cleanup or fractionation steps such as silica gel fractionation were performed on the PGC/OC portions).

2. Summary of gas chromatographic method for PCB congeners

The purified dosing solution fractions containing PCB congeners (PGC1) were adjusted to a final volume of 10 mL, to which two instrumental internal standards were added: PCB congeners 030 and 207 (400 ng each). Individual PCB congeners were measured in PGC1 fractions by GC/ECD. Analyses were performed using Hewlett-Packard 5890 Series II GCs with cool on-column capillary injection systems and Hewlett-Packard model 7673 autosamplers (10). For all analyses, a 3-m section of 0.53 mm i.d. uncoated and deactivated capillary retention gap guard column (J&W Scientific/Agilent, Wilmington, DE) was attached to the front of each analytical column by a Press-Tight® connector (Restek Corp., Bellefonte, PA). The analytical columns were a 60-m x 0.25-mm DB-5 (0.25µm 5% phenyl-, 95% methylsilicone, J&W Scientific/Agilent, Folsom, CA) and a 60-m x 0.25-mm DB-17 (0.25µm 50% phenyl-, 50% methylsilicone, J&W Scientific/Agilent, Folsom, CA). The H_2-carrier gas was pressure regulated at 25 psi. The temperature program for the PCB analysis was as follows: initial temperature 60°C, immediately ramped to 150°C at 15°C/min, then ramped to 250°C at 1°C/min, and finally ramped to 320°C at 10°C/min, and held for 1 min. The electron capture detector temperature was 330°C.

Capillary GC/ECD data were collected, archived in digital form, and processed using a Perkin-Elmer chromatography data system, which included the model 970 interface and version 6.1 of Turbochrom Workstation™ chromatography software (PerkinElmer Corp., Norwalk, CT), on a Pentium III microcomputer (11). Six levels of PCB standards, a combination of Aroclors 1242, 1248, 1254, 1260 in 1:1:1:1 w/w/w/w ratio (designated A1111), were used for PCB congeners calibration, with total PCB concentrations ranging from 50 to 8000 ng/mL. The elution (on two capillary columns) and concentrations of individual PCB congeners in the A1111 calibration standards have been verified against certified individual PCB standards (Accustandard, New Haven CT) using the methods of Frame and others (12-14).

The method detection limits (MDLs) and method quantitation limits (MQL) for individual PCB congeners and for total PCBs were based on procedural blank (PB) results according to the method outlined by Keith *et al.* (15,16). Briefly, an average and standard deviation are determined. The MDL and MQL values (ng) are calculated using the following formulas:

$$MDL = (PB\ AVG) + 3(PB\ SD)$$
$$MQL = (PB\ AVG) + 10(PB\ SD)$$

Both the MDL and MQL are then expressed in units of concentration, e.g. mass of analyte per mass of sample. An average mass for the set is used.

3. Summary of gas chromatographic method for OC pesticides

Organochlorine pesticides were analyzed with the PCBs. Since reactive cleanup was used and silica gel/ODS fractionation was not used the individual organochlorine pesticides were measured in the PGC1 fraction by GC/ECD (17). Instrumental components were the same as for the PCB congener analysis discussed above. The analytical column used was the DB-17 column (see congener PCB section above for details).

Capillary GC/ECD data were collected, archived in digital form, and processed using a Perkin-Elmer chromatography data system, which included the model 970- interface and version 6.1 of Turbochrom Workstation™ chromatography software, on a Pentium III microcomputer (11). Six levels of OC pesticide standards were used for calibration, with each pesticide at concentrations ranging from 0.5 to 80 ng/mL. Organochlorine pesticide results are presented in tables and are designated by their CERC database number and are cross-referenced to their field identification number. Concentrations are expressed as nanograms of analyte per gram of sample (wet weight). Detection limits were calculated as discussed above for PCB congeners.

4. Summary of GC/HRMS method for non-*ortho*-PCBs

The non-*ortho*-PCB fraction (PGC-2) contained 5 ng of instrumental internal standard (^{13}C-labeled 2,2',4,5,5'-PeCB (PCB #101). At a final volume of 50 µL, the non-*ortho*-PCBs were determined by GC/HRMS, monitoring two sequential mass windows during the chromatographic separation (18,19). GC/HRMS analysis was performed with a HP 5890A capillary gas chromatograph interfaced to a VG 70-250AS high-resolution mass spectrometer. An HP 7673 autosampler was used to introduce 2 µL of the extract onto a 2.5 m x 320 µm deactivated fused silica retention gap via heated (285 °C) direct on-column injection with a Restek spiral Uniliner. The analytical column was a 50 m x 200 µm x 0.11 µm Ultra-1 capillary column. The GC oven was held at 120 °C for 1 min, programmed to 240 °C at 2.2 °C/min, then ramped to 310 °C at 5 °C/min, and a final hold of 5 min. Helium carrier gas was maintained at 45 psig with an initial linear velocity of 27 cm/s.

The VG GC/HRMS system was tuned to a resolution of 10,000 and calibrated using perfluorodecalin. Mass windows were established for two groups of non-*ortho*-PCBs. Within each mass window, two most abundant ions were measured for positive identification and quantitation of each analyte. The ion responses were quantified and averaged. Also, additional ions monitored the responses of higher chlorinated, potentially interfering PCB congeners, Cl_{4-8} naphthalenes (PCNs), Cl_{3-5} terphenyls (PCTs), Br_5- and Cl_6-diphenyl ethers (residual carryover from PGC-1), and Cl_4-PCDF (to ensure no breakthrough of PCDFs).

A calibration curve describing the response of each native congener (0.25 to 2,500 pg/µL) to that of its ^{13}C-labeled surrogate was used. Quantification is inherently

corrected by the ^{13}C-isotopically labeled surrogates, which account for analytical losses during isolation procedures and variations in the instrumental analysis.

Molecular ion responses of certain PCB congeners are measured to ensure that their fragment ion responses do not contribute an interference >10% to the responses of the respective non-*ortho*-PCB. Column performance is verified by analyzing standards of individual congeners, labeled congeners, and congeners from Aroclor mixtures. Because non-*ortho*-Cl_5-PCB 126 is only minimally resolved from Cl_6-PCB 129, PCB 129's molecular ion response is monitored to assure that its fragment ion response (3.5% abundance) does not contribute an interference of >10% to the response of PCB 126. PCB 129's molecular ion response must not exceed three times that of PCB 126. Adequate mass resolution is verified while monitoring $Cl_{4\text{-}8}$ ions of PCNs.

Criteria for Confirmation: For the positive identification and quantitation of each congener, the following criteria were established and met in this study:

1. Peak areas for the selected ion responses must be greater than three times background noise.
2. Native ion peaks must occur at retention times from -1 to +3 sec of that for the corresponding ^{13}C-labeled ion peaks, which generally elute about 1 sec earlier.
3. The ion ratio for the two principal ion responses must be within the acceptable range (±15%).

5. Summary of GC/HRMS method for 2,3,7,8-Cl substituted PCDDs/PCDFs

The PCDD/PCDF fractions from PGC (PGC-3) were eluted through basic alumina to remove potential co-contaminants such as chlorinated diphenyl ethers (PCDEs) and residual PCNs and PCBs (6). A total of 1 ng of the internal standard, ^{13}C-labeled 1,2,3,4-TCDD (tetrachlorodibenzo-*p*-dioxin), was added and the volume reduced to ~25 µL. The GC/HRMS analysis was performed using a HP 5890A capillary gas chromatograph interfaced to a VG 70-AS high-resolution mass spectrometer. Five sequential selected ion windows were monitored. (20). An HP 7673 autosampler injected 2µL of the extract into the GC/HRMS. A spiral uniliner, 2.5m x 320µm deactivated fused silica retention gap, and heated (285 °C) direct inlet were employed. The analysis was conducted with a 50m x 200µm x 0.11µm Ultra-2 (Agilent Technologies, Wilmington, DE) capillary column, with an initial hold of 1 min at 120 °C followed by a ramp to 200 °C at 20 °C/min, another ramp to 300 °C at 2.3 °C/min, and a final hold of 5 min. The helium carrier gas was maintained at 44 psig with an initial linear velocity of 25 cm/s.

The VG GC/HRMS system was tuned to a resolution of 10,000 and calibrated using perfluorokerosene. Mass windows were established for five ion groups to measure $Cl_{4\text{-}8}$ PCDFs and PCDDs. Within each mass window, the two most abundant ions were measured for positive identification and quantitation of each analyte. The ion responses

were quantified and averaged. Additional ions monitored any responses from potentially interfering Cl_{5-9}-PCDEs and Cl_{5-7}-polychlorinated terphenyls (PCTs), and dioxin-like Cl_{6-7}-PCNs, Cl_{3-8} dibenzothiophenes (PCDTs), and Cl_{3-8} phenanthrene and anthracenes. A calibration curve describing the response of each native congener to that of a ^{13}C-labeled surrogates congener was used for quantification.

Column performance was verified by analyzing standards of individual components, and observing the chromatographic resolution of the TCDDs, HxCDDs (hexachlorodibenzodioxins), and HxCDFs (hexachlorodibenzofurans). Similarly, relative retention times for all other congeners of interest were evaluated with respect to labeled analogs. Adequate mass resolution was verified while monitoring ions Cl_{6-7} PCNs vs. ion responses of ^{13}C-TCDDs and of native TCDD versus ^{13}C-TCDF. Lock-mass and lock-mass-check ions were used to maintain and verify the accuracy of mass measurement.

For the positive identification and quantitation of a particular congener, the following criteria were met:

1. The peak areas for the selected ion responses must be greater than three times the background noise (S/N > 3)
2. For congeners with isotopically-labeled analogs, the ion peaks for the native must occur at retention times from -1 to +3 sec that for the corresponding ^{13}C-labeled ion peaks, which elute about 1 sec earlier than the native ion peaks;
3. For OCDF (without an isotopically-labeled analog), ion responses in sample analyses must occur at RRTs from -0.2 to 0.5% of ^{13}C-labeled OCDD, analogous to the window above;
4. For the two principal ion responses, the ion ratio must be within the acceptable range (±15%).

III. Results and Discussion

1. PCB Congeners

The results of the analysis of the samples for 139 PCB congeners and total-PCBs are presented in Table 1. Note that the units are nanograms of analyte per gram (ng/g) of fish tissue, referring back to the original masses of the fish tissue extracted for the dosing solution (Rising Pond 12,800 g, Woods Pond 14,200 g, Deep Reach Pond 13,000 g, Three Mile Pond 15,200 g). The average concentrations of total-PCBs in the dosing solutions: were 18,000 ng/g for Rising Pond; 37,000 ng/g for Woods Pond; 62,000 ng/g for Deep Reach Pond and 30 ng/g for Three Mile Pond (reference site).

The concentrations in the dosing solutions are corrected for recovery through the HPLC carbon fractionation step. Recoveries are presented in Table 2. Recoveries through HPLC-carbon (the only step monitored) for 029, 155 and 204 averaged 61±7%, 67±3%

and 78±1%, respectively. The QC consisted of a procedural blank (injected in triplicate), and a procedural spike and method triplicates of dosing solutions from each site. Precision of the method was better than 3% RSD for total-PCBs at the four sites. The procedural blank's low background of PCBs was insignificant when compared with samples. Detection limits for total PCBs were 20 ng/g.

Dioxin toxic equivalency factors (TEFs) for fish were used to evaluate the toxic equivalents (TEQs) in the dosing solutions (21). The mono-*ortho* PCB TEQs were tabulated and the total TEQs were reported in Table 8. A conservative approach was used in calculating TEQs: any PCB reported as not detected (ND) was considered present at one-half the specified minimum detection limit (MDL). PCB 118 contributes most of the mono-*o*-PCB TEQ concentrations.

2. Organochlorine Pesticides

The results for triplicate analyses of each of the four dosing solutions for organochlorine pesticides (OCPs) are presented in Table 3. Because these extracts were treated by reactive cleanup, a number of the OC pesticides (beta-BHC, dacthal, dieldrin, endrin, methoxychlor) were destroyed, and are thus not shown in Table 3. Since no silica gel fractionation was performed on these extracts to separate OC pesticides from PCBs the OC pesticides were analyzed in the same fraction as the PCBs. The chromatographic resolution was sufficient to distinguish most of the pesticides from the PCBs. MDLs for the OCPs ranged from 0.01 ng/g to 1.1 ng/g. Most of the pesticides were below detection however, low amounts of p,p'-DDE, p,p'-DDD, and p,p'-DDT; *trans*- and *cis*-nonaclor; hexachlorobenzene and pentachloroanisole were found at detectable levels in the dosing solutions.

3. Non-*ortho*-PCB Congeners

The concentrations of non-*ortho*-PCBs (81, 77, 126, and 169) found in the dosing solutions are presented in Table 4. Note that the units are picograms of analyte per gram (pg/g) of fish tissue (wet weight). In samples from the three non-reference sites—Rising Pond, Woods Pond, Deep Reach Pond--PCB 126 was present at the highest concentration, followed by PCB 77 and PCB 169.

Dioxin toxic equivalency factors (TEFs) for fish were used to evaluate the toxic equivalents (TEQs) in the dosing solutions (21). The non-ortho PCB TEQs were tabulated and the total TEQs are reported in Table 8. A conservative approach was used in calculating TEQs: any PCB reported as not detected (ND) was considered present at one-half the specified minimum detection limit (MDL). Any values listed as "LQ", however, were used as shown in the table. The "LQ" indicates that the compound was detected, but is less than the minimum quantification limit (MQL) due to incomplete ion cluster or ion ratio outside of ±15% tolerance. PCB 126 contributes virtually all of the non-*o*-PCB TEQ concentrations.

Quality control was within guidelines, except where noted. Precision of the analysis of dosing solution triplicates for non-*ortho* PCBs had coefficients of variation of 2 to 16% for Rising Pond solutions; 3 to 29% for Woods Pond; 12 to 23% for Deep Reach Pond dosing solutions. Ion ratios were within the expected QC tolerance (±15%). Surrogate recoveries (these went through HPLC-carbon and alumina steps only), listed in Table 5, averaged 69±9% for ^{13}C PCB 81, 72±7% for ^{13}C PCB 77, 78±7% for ^{13}C PCB 126, and 88±14% for ^{13}C PCB 169.

Concentrations of each non-*ortho*-PCB in the procedural blank were all low (less than 20 pg, on a total sample basis). In Table 4, some values for PCB 81 and PCB 77 are designated (IF) to indicate that they were corrected for interference from the di-*ortho*-PCB whenever it contributed more than 10% of the value (PCB 87 for PCB 81, PCB 110 for PCB 77).

4. PCDDs and PCDFs

Concentrations of 2,3,7,8-substituted PCDFs and PCDDs in the dosing solutions are presented in Table 6. Note that the units are picograms of analyte per gram (pg/g) of fish tissue in the dosing solution (wet weight). Also shown in Table 6 are mean concentrations for each triplicate, percent coefficients of variation, as well as the geometric mean. As expected, PCDD/PCDF concentrations were low (< 1 pg/g) in field control dosing solution (Three Mile Pond) for all but OCDD and 2378-TCDF, and higher in samples from the other sites. The highest analyte for all sample sites was 2,3,4,7,8-pentachlorodibenzofuran (PeCDF), followed by 2,3,4,6,7,8-hexachloro- (HxCDF) and 2,3,7,8-tetrachlorodibenzofurans (TCDF).

Dioxin toxic equivalency factors (TEFs) for fish were used to evaluate the toxic equivalents (TEQs) in the dosing solutions (21). The dioxin/furan TEQs are tabulated for each replicate at the bottom of Table 6 and the total TEQs were reported in Table 8. A conservative approach was used in calculating TEQs: any PCDDs/PCDFs reported as not detected (ND) was considered present at one-half the specified minimum detection limit (MDL). Any values listed as “LQ”, however, were used as shown in the table. The “LQ” indicates that the compound was detected, but is less than the minimum quantification limit (MQL) due to incomplete ion cluster or ion ratio outside of ±15% tolerance. The 2,3,4,7,8-PeCDF contributed most of the TEQ concentrations resulting from PCDDs and PCDFs.

Quality control was within our guidelines, except where noted. Ion ratios for analytes were within the expected QC tolerance (±15%). Surrogate recoveries, listed in Table 7, were within QC guidelines, except for a few samples that had low recoveries of the hexa- and hepta-PCDFs because of earlier elution from alumina. Concentrations of each PCDF/PCDD in the procedural blank were all low (less than 0.2 pg/g, normalized to a typical 10-g basis). The quality control blank concentrations were within the same range as the lowest concentrations in the solutions.

5. Summary

The dosing solution concentrations of total-PCBs (based on grams of fish tissue extracted) in Table 1 were 18,000 ng/g, 37,000 ng/g, 62,000 ng/g, and 30 ng/g for Rising Pond, Woods Pond, Deep Reach Pond, and Three Mile Pond, respectively. Organochlorine pesticides (Table 3) were not found at significant levels in the dosing solutions. Average levels of p,p'-DDE were generally found to be < 40 ng/g. Non-*o*-PCB concentrations for Rising Pond, Woods Pond, Deep Reach Pond and Three Mile Pond were, respectively: 35±6, 81±21, 180±40, and 0.45±0.06 pg/g for congener 81; 820±61, 1600±130, 2100±380, 7.8±0.4 pg/g for congener 77; 1500±50, 2400±180, 2700±560, 2.9±0.2 pg/g for congener 126; 220±6, 300±22, 360±36, 0.5±0.04 pg/g for congener 169. Dioxin and furans were at or near background level in dosing solutions from all sites.

Total TEQs, presented in Table 8 show that TEQs (using fish TEFs from WHO reference 21) for these dosing solutions range from 10 to 37 pg/g. The TEQs from the Three Mile Pond reference site are low, averaging < 2 pg/g. The contribution of non-*ortho*-PCBs dominates the TEQs for Woods Pond and Deep Reach Pond dosing solutions, whereas, furans predominate (55-58%) the TEQs for Rising Pond.

IV. References

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PCBs, PCDD/PCDFs and Pesticides in Dosing Solutions Created from Largemouth Bass Tissue from the Housatonic River

Final Report: April 10, 2002

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Table 1. Housatonic Large Mouth Bass--Dosing Solutions--PCB Concentrations (ng/g) and Quality Control Data

Sample ID	Field ID	Sample Type	Gram-equivalents for Analysis (g)	001	003	004	005	006	007	008	009	010	015	016	017	018
RP LMB	Rising Pond	Dosing Solution	12,800	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	1.5	1.3
RP-A DOSE	Rising Pond	Dosing Solution	12,800	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	0.92	1.4	1.3
RP-B DOSE	Rising Pond	Dosing Solution	12,800	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	1.5	1.2
RP-C DOSE	Rising Pond	Dosing Solution	12,800	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	1.4	1.2
Average															1.4	1.2
SD(n-1)															0.05	0.05
%RSD															4	4
WP LMB	Woods Pond	Dosing Solution	14,200	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	8.1	6.5
WP-A DOSE	Woods Pond	Dosing Solution	14,200	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	8.2	7.2
WP-B DOSE	Woods Pond	Dosing Solution	14,200	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	7.8	6.4
WP-C DOSE	Woods Pond	Dosing Solution	14,200	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	8.2	6.6
Average															8.1	6.7
SD(n-1)															0.20	0.37
%RSD															3	6
DRP LMB	Deep Reach Pond	Dosing Solution	13,000	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	13	11
DRP-A DOSE	Deep Reach Pond	Dosing Solution	13,000	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	14	11
DRP-B DOSE	Deep Reach Pond	Dosing Solution	13,000	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.4	< 0.01	14	11
DRP-C DOSE	Deep Reach Pond	Dosing Solution	13,000	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	< 0.02	< 0.01	12	8.8
Average															14	10
SD(n-1)															1.3	1.1
%RSD															9	11
3MP LMB	Three Mile Pond	Dosing Solution	15,200	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.05	< 0.01	< 0.02	< 0.07
3MP-A DOSE	Three Mile Pond	Dosing Solution	15,200	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.05	< 0.01	< 0.02	< 0.07
3MP-B DOSE	Three Mile Pond	Dosing Solution	15,200	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.05	< 0.01	< 0.02	< 0.07
3MP-C DOSE	Three Mile Pond	Dosing Solution	15,200	< 0.01	< 0.01	< 0.29	< 0.01	< 0.01	< 0.07	< 0.09	< 0.01	< 0.01	0.05	< 0.01	< 0.02	< 0.07
Average													0.05			
SD(n-1)													0.00			
%RSD													0			
PS 113000 PCB*	Procedure Spike	Na_2SO_4	---	2.2	1.4	12	0.51	6.2	0.83	22	2.0	0.45	3.5	15	17	42
Percent Recovery				***52***	***75***	***72***	***66***	***69***	***67***	***68***	***67***	***86***	***75***	***75***	***60***	***74***
PB 113000 GCR1	Procedure Blank	Na_2SO_4	---	0.00	0.00	0.15	0.00	0.00	0.07	0.07	0.00	0.00	0.01	0.02	0.00	0.01
PB 113000 GCR2	Procedure Blank	Na_2SO_4	---	0.00	0.00	0.01	0.00	0.00	0.07	0.08	0.00	0.00	0.00	0.01	0.01	0.02
PB 113000 GCR3	Procedure Blank	Na_2SO_4	---	0.00	0.00	0.02	0.00	0.00	0.07	0.07	0.00	0.00	0.00	0.00	0.00	0.04
Average				0.00	0.00	0.06	0.00	0.00	0.07	0.07	0.00	0.00	0.00	0.00	0.00	0.02
Standard Deviation				0.00	0.00	0.08	0.00	0.00	0.00	0.01	0.00	0.00	0.01	0.00	0.01	0.02
MDL		*Method Detection Limit = PB Average +*		0.00	0.00	0.29	0.00	0.00	0.07	0.09	0.00	0.00	0.02	0.00	0.02	0.07
MDL	Mass normalized to 1 g			0.01	0.01	0.29	0.01	0.01	0.07	0.09	0.01	0.01	0.02	0.01	0.02	0.07
Note: Values are rounded to 2 significant figures.																
MDL < 0.01 set to 0.01 instrument detection limit																
***Average of injection triplicate.**																

Table 1. Housatonic Large Mouth Bass--Dosing Solutions--PCB Concentrations (ng/g) and Quality Control Data

Sample ID	Field ID	019	020	022	024	025	026	027	028	031	032	033	034	035	037,059	040	041
RP LMB	Rising Pond	1.3	< 0.01	0.31	< 0.01	1.5	2.4	0.66	5.2	0.78	3.0	< 0.04	0.39	< 0.10	0.62	0.48	< 0.02
RP-A DOSE	Rising Pond	1.3	< 0.01	0.31	< 0.01	1.4	2.3	0.51	4.9	0.66	1.3	< 0.04	< 0.17	< 0.10	0.47	0.42	< 0.02
RP-B DOSE	Rising Pond	1.5	< 0.01	0.29	< 0.01	1.3	2.4	0.55	4.8	0.77	3.1	< 0.04	< 0.17	< 0.10	0.51	0.49	< 0.02
RP-C DOSE	Rising Pond	1.2	< 0.01	0.26	< 0.01	1.5	2.5	0.59	4.8	0.82	3.3	< 0.04	< 0.17	< 0.10	0.48	0.43	0.03
Average		1.3		0.29		1.4	2.4	0.55	4.9	0.75	2.6				0.49	0.45	
SD(n-1)		0.16		0.03		0.08	0.06	0.04	0.07	0.08	1.1				0.02	0.03	
%RSD		12		9		5	3	8	1	11	43				4	8	
WP LMB	Woods Pond	5.8	< 0.01	1.8	< 0.01	8.5	15	3.0	21	4.5	15	1.7	1.7	< 0.10	1.8	1.4	< 0.02
WP-A DOSE	Woods Pond	7.0	< 0.01	1.2	< 0.01	8.9	16	3.1	21	4.4	17	1.7	< 0.17	< 0.10	1.9	1.3	< 0.02
WP-B DOSE	Woods Pond	6.6	< 0.01	1.4	< 0.01	8.2	16	3.1	20	4.4	17	1.4	< 0.17	0.17	1.2	1.3	0.47
WP-C DOSE	Woods Pond	9.9	< 0.01	1.7	< 0.01	8.7	15	2.9	21	4.6	16	1.7	< 0.17	< 0.10	1.9	1.1	< 0.02
Average		7.8		1.4		8.6	16	3.1	21	4.4	17	1.6			1.7	1.2	
SD(n-1)		1.8		0.24		0.37	0.19	0.14	0.19	0.13	0.74	0.19			0.39	0.12	
%RSD		23		17		4	1	5	1	3	4	12			23	10	
DRP LMB	Deep Reach Pond	24	0.49	2.4	< 0.01	7.8	19	5.8	20	7.8	39	2.4	< 0.17	0.49	2.9	2.3	< 0.02
DRP-A DOSE	Deep Reach Pond	11	< 0.01	1.8	< 0.01	6.8	18	5.0	18	7.7	39	0.90	< 0.17	0.90	< 0.02	1.7	< 0.02
DRP-B DOSE	Deep Reach Pond	9.3	< 0.01	2.1	< 0.01	5.9	20	5.5	19	5.9	38	1.7	3.0	< 0.10	2.1	1.6	< 0.02
DRP-C DOSE	Deep Reach Pond	24	< 0.01	2.4	< 0.01	7.3	18	5.4	21	7.8	30	0.49	0.49	< 0.10	2.9	2.3	< 0.02
Average		15		2.1		6.7	18	5.3	20	7.1	36	1				1.8	
SD(n-1)		8.3		0.32		0.7	1.2	0.3	1.1	1.0	5.2	0.6				0.38	
%RSD		56		15		10	7	5	6	15	15	60				21	
3MP LMB	Three Mile Pond	< 0.14	< 0.01	< 0.04	< 0.01	0.01	< 0.01	< 0.04	< 0.04	< 0.02	< 0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02	< 0.02
3MP-A DOSE	Three Mile Pond	< 0.14	< 0.01	< 0.04	< 0.01	< 0.01	< 0.01	< 0.04	< 0.04	< 0.02	< 0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02	< 0.02
3MP-B DOSE	Three Mile Pond	< 0.14	< 0.01	< 0.04	< 0.01	< 0.01	< 0.01	< 0.04	< 0.04	< 0.02	0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02	< 0.02
3MP-C DOSE	Three Mile Pond	< 0.14	< 0.01	< 0.04	< 0.01	< 0.01	< 0.01	< 0.04	< 0.04	< 0.02	0.01	< 0.04	< 0.17	< 0.10	< 0.02	< 0.02	< 0.02
Average																	
SD(n-1)																	
%RSD																	
PS 113000 PCB*	Procedure Spike	4.0	1.8	14	0.39	1.5	7.0	1.7	34	30	17	20	< 0.17	0.16	3.1	6.8	4.6
Percent Recovery		***73***	***68***	***73***	***77***	***70***	***73***	***78***	***62***	***71***	***69***	***67***	***---***	***86***	***84***	***80***	***75***
PB 113000 GCR1	Procedure Blank	0.05	0.00	0.00	0.00	0.00	0.00	0.02	0.02	0.00	0.00	0.03	0.04	0.00	0.00	0.01	0.00
PB 113000 GCR2	Procedure Blank	0.09	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.03	0.01	0.05	0.01	0.00	0.01
PB 113000 GCR3	Procedure Blank	0.08	0.00	0.02	0.00	0.00	0.00	0.00	0.02	0.00	0.00	0.02	0.09	0.00	0.01	0.00	0.00
Average		0.07	0.00	0.01	0.00	0.00	0.00	0.01	0.01	0.00	0.00	0.03	0.05	0.02	0.01	0.00	0.00
Standard Deviation		0.02	0.00	0.01	0.00	0.00	0.00	0.01	0.01	0.01	0.00	0.01	0.04	0.03	0.01	0.01	0.01
MDL		0.14	0.00	0.04	0.00	0.00	0.00	0.04	0.04	0.02	0.00	0.04	0.17	0.10	0.02	0.02	0.02
MDL	Mass normalized to 1 g	0.14	0.01	0.04	0.01	0.01	0.01	0.04	0.04	0.02	0.01	0.04	0.17	0.10	0.02	0.02	0.02

Note: Values are rounded to 2 significant figures.

MDL < 0.01 set to 0.01 instrument detection limit

***Average of injection triplicate.**

Table 1. Housatonic Large Mouth Bass--Dosing Solutions--PCB Concentrations (ng/g) and Quality Control Data

Sample ID	Field ID	042	043	044	045	046	047	048	049	051	052	053	*054*	055	*056,060*	*057*	*058*
RP LMB	Rising Pond	13	3.2	12	0.38	0.31	180	50	97	8.8	74	7.9	< 0.02	< 0.02	3.2	0.41	0.83
RP-A DOSE	Rising Pond	11	1.1	12	0.39	0.35	170	51	96	8.4	72	7.9	< 0.02	< 0.02	3.0	0.39	0.85
RP-B DOSE	Rising Pond	13	1.4	12	0.49	0.36	170	< 12	93	8.5	70	7.8	< 0.02	< 0.02	3.3	0.07	0.78
RP-C DOSE	Rising Pond	11	2.9	12	0.47	0.33	170	19	95	8.5	72	7.8	< 0.02	< 0.02	2.9	0.13	0.76
Average		11	1.8	12	0.45	0.35	170		90	8.5	70	7.9			3.1	0.20	0.80
SD(n-1)		1.1	0.96	0.20	0.05	0.01	0.0		1.6	0.04	1.1	0.08			0.22	0.17	0.05
%RSD		10	54	2	12	4	0		2	1	2	1			7	87	6
WP LMB	Woods Pond	83	3.5	39	0.60	1.1	420	230	270	33	220	30	0.75	< 0.02	19	1.1	2.1
WP-A DOSE	Woods Pond	80	4.0	39	0.32	1.3	420	220	260	33	210	30	0.64	< 0.02	21	1.8	2.3
WP-B DOSE	Woods Pond	82	2.8	38	0.94	1.3	430	230	270	32	220	30	0.63	< 0.02	19	1.6	2.2
WP-C DOSE	Woods Pond	81	3.2	39	0.76	1.1	410	210	260	32	210	30	0.76	< 0.02	18	0.91	1.5
Average		81	3.3	38	0.67	1.2	420	220	260	32	210	30	0.68		20	1.4	2.0
SD(n-1)		0.64	0.61	0.70	0.32	0.12	10	10	5.8	0.59	5.8	0.26	0.07		1.3	0.45	0.43
%RSD		1	18	2	47	10	2	5	2	2	3	1	11		6	32	21
DRP LMB	Deep Reach Pond	100	8.3	54	< 0.12	1.8	630	700	340	47	300	40	0.92	< 0.02	53	< 0.02	< 0.01
DRP-A DOSE	Deep Reach Pond	93	5.4	47	1.7	1.7	570	610	310	41	280	37	0.42	< 0.02	52	< 0.02	< 0.01
DRP-B DOSE	Deep Reach Pond	92	6.6	51	< 0.12	1.2	560	590	310	43	280	39	1.6	< 0.02	52	0.39	< 0.01
DRP-C DOSE	Deep Reach Pond	92	6.8	50	< 0.12	1.8	580	690	320	43	280	39	0.45	< 0.02	53	2.3	< 0.01
Average		92	6.3	49		1.5	570	630	310	42	280	38	0.81		52		
SD(n-1)		0.3	0.75	1.8		0.33	10	53	6	1	0	1	0.65		0.50		
%RSD		0	12	4		22	2	8	2	3	0	3	80		1		
3MP LMB	Three Mile Pond	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.08	< 12	< 0.08	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02	< 1.7	0.03	< 0.01
3MP-A DOSE	Three Mile Pond	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.08	< 12	< 0.08	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02	< 1.7	0.03	< 0.01
3MP-B DOSE	Three Mile Pond	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.09	< 12	< 0.08	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02	< 1.7	0.03	< 0.01
3MP-C DOSE	Three Mile Pond	< 0.13	< 0.01	< 0.07	< 0.12	< 0.01	0.08	< 12	< 0.08	< 0.01	< 0.13	< 0.01	< 0.02	< 0.02	< 1.7	0.03	< 0.01
Average							0.08									0.03	
SD(n-1)							0.00									0.00	
%RSD							0									1	
PS 113000 PCB*	Procedure Spike	13	3.2	40	7.5	3.3	12	<12	27	1.8	53	9.0	0.03	0.44	26	< 0.02	< 0.01
Percent Recovery		***74***	***84***	***70***	***73***	***67***	***70***	***---***	***70***	***67***	***69***	***62***	***83***	***71***	***79***	***---***	***---***
PB 113000 GCR1	Procedure Blank	0.01	0.00	0.06	0.00	0.00	0.04	10	0.01	0.00	0.11	0.00	0.00	0.00	0.37	0.00	0.00
PB 113000 GCR2	Procedure Blank	0.07	0.00	0.06	0.06	0.00	0.01	8.8	0.04	0.00	0.09	0.00	0.00	0.00	0.38	0.01	0.00
PB 113000 GCR3	Procedure Blank	0.02	0.00	0.05	0.00	0.00	0.03	7.9	0.00	0.00	0.10	0.00	0.01	0.01	1.0	0.01	0.00
Average		0.03	0.00	0.06	0.02	0.00	0.03	8.9	0.02	0.00	0.10	0.00	0.00	0.00	0.59	0.01	0.00
Standard Deviation		0.03	0.00	0.01	0.03	0.00	0.01	1.0	0.02	0.00	0.01	0.00	0.01	0.01	0.37	0.01	0.00
MDL		0.13	0.00	0.07	0.12	0.00	0.07	12	0.08	0.00	0.13	0.00	0.02	0.02	1.7	0.02	0.00
MDL	Mass normalized to 1 g	0.13	0.01	0.07	0.12	0.01	0.07	12	0.08	0.01	0.13	0.01	0.02	0.02	1.7	0.02	0.01

Note: Values are rounded to 2 significant figures.

MDL < 0.01 set to 0.01 instrument detection limit

***Average of injection triplicate.**

Table 1. Housatonic Large Mouth Bass--Dosing Solutions--PCB Concentrations (ng/g) and Quality Control Data

Sample ID	Field ID	063	064	066	067	*069*	070	071	*072*	074	075	082	083	084	085	*086*	087
RP LMB	Rising Pond	1.7	4.1	31	0.55	0.58	11	59	4.4	10	2.6	11	3.2	17	100	1.8	100
RP-A DOSE	Rising Pond	1.7	3.9	30	0.49	0.39	10	56	4.1	9.7	1.9	10	3.1	17	99	1.6	95
RP-B DOSE	Rising Pond	1.7	3.9	29	0.55	0.46	10	55	4.0	9.6	2.3	10	3.0	16	97	1.5	94
RP-C DOSE	Rising Pond	1.7	4.0	29	0.50	0.47	11	56	4.0	9.6	2.2	10	3.0	17	98	1.7	95
Average		1.7	4.0	29	0.52	0.44	10	60	4.0	9.6	2.1	10	3.0	16	98	1.6	90
SD(n-1)		0.01	0.06	0.27	0.03	0.04	0.10	0.35	0.08	0.07	0.22	0.07	0.02	0.21	0.90	0.06	0.80
%RSD		1	1	1	6	10	1	1	2	1	10	1	1	1	1	4	1
WP LMB	Woods Pond	3.8	13	76	1.5	1.2	44	120	11	30	4.8	15	8.8	49	160	2.8	260
WP-A DOSE	Woods Pond	3.4	12	75	1.0	1.3	43	120	8.5	29	4.5	15	9.2	49	150	3.1	260
WP-B DOSE	Woods Pond	4.4	13	73	1.7	1.1	36	120	10	29	5.6	15	8.9	49	150	3.6	250
WP-C DOSE	Woods Pond	3.9	12	74	1.2	1.5	43	120	9.9	29	3.6	15	9.0	48	150	2.8	250
Average		3.9	12	74	1.3	1.3	41	120	9.6	29	4.6	15	9.0	49	150	3.2	250
SD(n-1)		0.51	0.42	1.1	0.39	0.21	3.7	0.0	0.94	0.29	1.0	0.19	0.13	0.53	0.0	0.42	5.8
%RSD		13	3	2	30	16	9	0	10	1	22	1	1	1	0	13	2
DRP LMB	Deep Reach Pond	2.3	14	87	1.8	0.46	57	150	6.9	46	4.1	26	14	77	210	3.4	450
DRP-A DOSE	Deep Reach Pond	2.5	17	81	1.7	0.42	57	150	6.6	44	2.9	23	12	71	200	2.5	410
DRP-B DOSE	Deep Reach Pond	2.7	16	80	2.0	2.7	59	150	6.6	42	3.1	24	12	72	190	4.8	410
DRP-C DOSE	Deep Reach Pond	2.3	17	82	< 0.06	0.90	60	150	7.2	44	3.2	25	13	73	200	5.0	420
Average		2.5	17	81	1.8	1.3	59	150	6.8	43	3.1	24	12	72	200	4.1	410
SD(n-1)		0.24	0.32	0.83	0.20	1.2	1.3	0.0	0.33	1.0	0.13	1.0	0.50	1	5.8	1.3	6
%RSD		10	2	1	11	90	2	0	5	2	4	4	4	1	3	33	1
3MP LMB	Three Mile Pond	< 0.01	< 0.02	< 0.08	< 0.06	< 0.01	< 0.20	< 0.23	< 0.01	< 0.13	< 0.01	0.01	< 0.01	< 0.17	17	< 0.01	< 0.29
3MP-A DOSE	Three Mile Pond	0.01	< 0.02	0.08	< 0.06	< 0.01	< 0.20	< 0.23	< 0.01	< 0.13	< 0.01	0.02	< 0.01	< 0.17	18	< 0.01	< 0.29
3MP-B DOSE	Three Mile Pond	0.01	< 0.02	0.08	< 0.06	< 0.01	< 0.20	< 0.23	< 0.01	< 0.13	< 0.01	0.02	< 0.01	< 0.17	18	< 0.01	< 0.29
3MP-C DOSE	Three Mile Pond	0.01	< 0.02	0.08	< 0.06	< 0.01	< 0.20	< 0.23	< 0.01	< 0.13	< 0.01	0.02	< 0.01	< 0.17	18	< 0.01	< 0.29
Average		0.01		0.08								0.02			18		
SD(n-1)		0.00		0.00								0.00			0.07		
%RSD		1		0.0								0.00			0.4		
PS 113000 PCB*	Procedure Spike	0.95	13	24	1.0	0.02	44	14	0.10	20	0.52	5.9	0.78	15	9.0	0.25	28
Percent Recovery		***80***	***72***	***73***	***75***	***83***	***76***	***71***	***---***	***69***	***76***	***85***	***82***	***73***	***78***	***126***	***77***
PB 113000 GCR1	Procedure Blank	0.00	0.00	0.01	0.03	0.00	0.12	0.09	0.00	0.03	0.00	0.01	0.00	0.08	0.05	0.00	0.22
PB 113000 GCR2	Procedure Blank	0.00	0.01	0.00	0.03	0.00	0.16	0.12	0.00	0.08	0.00	0.01	0.00	0.11	0.06	0.00	0.24
PB 113000 GCR3	Procedure Blank	0.00	0.00	0.04	0.01	0.00	0.13	0.02	0.00	0.04	0.00	0.01	0.00	0.12	0.07	0.00	0.19
Average		0.00	0.00	0.02	0.02	0.00	0.14	0.08	0.00	0.05	0.00	0.01	0.00	0.10	0.06	0.00	0.22
Standard Deviation		0.00	0.01	0.02	0.01	0.00	0.02	0.05	0.00	0.03	0.00	0.00	0.00	0.02	0.01	0.00	0.03
MDL		0.00	0.02	0.08	0.06	0.00	0.20	0.23	0.00	0.13	0.00	0.01	0.00	0.17	0.09	0.00	0.29
MDL	Mass normalized to 1 g	0.01	0.02	0.08	0.06	0.01	0.20	0.23	0.01	0.13	0.01	0.01	0.01	0.17	0.09	0.01	0.29

Note: Values are rounded to 2 significant figures.

MDL < 0.01 set to 0.01 instrument detection limit

***Average of injection triplicate.**

Table 1. Housatonic Large Mouth Bass--Dosing Solutions--PCB Concentrations (ng/g) and Quality Control Data

Sample ID	Field ID	090	091	092	095	096	097	099	101	102	105	109	110	*112*	*113*	114	115
RP LMB	Rising Pond	93	70	140	150	0.89	59	300	480	3.1	92	110	290	0.87	5.5	3.2	2.9
RP-A DOSE	Rising Pond	88	65	130	140	0.85	57	280	450	3.4	87	110	270	0.90	5.5	2.6	2.8
RP-B DOSE	Rising Pond	87	64	130	140	0.78	56	280	450	3.1	85	110	270	0.83	5.2	3.3	2.8
RP-C DOSE	Rising Pond	87	66	130	140	0.83	56	280	450	2.8	86	110	270	0.77	5.3	3.1	2.8
Average		90	70	130	140	0.82	60	280	450	3.1	90	110	270	0.83	5.3	3.0	2.8
SD(n-1)		0.46	1.0	0.00	0.00	0.04	0.72	0.00	0.0	0.27	1.0	0.0	0.0	0.06	0.16	0.36	0.02
%RSD		1	1	0	0	4	1	0	0	9	1	0	0	8	3	12	1
WP LMB	Woods Pond	130	130	320	440	3.0	130	460	1,100	2.6	150	220	600	0.55	15	8.0	7.1
WP-A DOSE	Woods Pond	130	130	320	430	2.9	130	450	1,000	2.4	150	220	600	0.39	14	11	6.9
WP-B DOSE	Woods Pond	120	130	320	430	2.8	130	450	1,000	0.5	140	220	600	1.7	14	8.4	6.7
WP-C DOSE	Woods Pond	120	130	320	430	3.2	120	450	1,000	2.1	140	220	580	0.37	15	8.2	6.8
Average		120	130	320	430	3.0	130	450	1,000	1.7	140	220	590	0.82	14	9.3	6.8
SD(n-1)		5.8	0.0	0.0	0.0	0.20	5.8	0.0	0.0	1.0	5.8	0.0	12	0.77	0.62	1.7	0.09
%RSD		5	0	0.0	0.0	7	4	0.0	0.0	63	4.1	0.0	2	93	4	18	1
DRP LMB	Deep Reach Pond	150	190	600	690	3.2	200	570	1,600	< 0.02	270	250	1,100	3.3	19	19	10
DRP-A DOSE	Deep Reach Pond	140	170	540	640	2.9	180	530	1,500	2.9	250	240	880	2.0	17	20	10
DRP-B DOSE	Deep Reach Pond	140	180	530	640	3.5	180	520	1,500	3.1	240	240	890	1.9	17	20	10
DRP-C DOSE	Deep Reach Pond	140	180	570	660	2.3	190	530	1,500	4.1	260	240	1,000	0.5	18	19	9.9
Average		140	180	550	650	2.9	180	530	1,500	3.4	250	240	920	1.5	17	20	10
SD(n-1)		0	6	21	10	0.63	6	6	0	0.6	10	0	70	0.8	0.5	0.3	0.2
%RSD		0	3	4	2	21.7	3	1	0	18	4	0	8	55	3	1	2
3MP LMB	Three Mile Pond	0.04	< 0.08	< 0.17	< 0.19	< 0.01	< 0.29	0.25	0.34	< 0.02	< 0.24	0.08	< 0.40	0.01	0.07	0.01	< 0.02
3MP-A DOSE	Three Mile Pond	0.04	< 0.08	< 0.17	< 0.19	< 0.01	< 0.29	0.26	0.35	< 0.02	< 0.24	0.09	< 0.40	0.01	0.08	0.01	< 0.02
3MP-B DOSE	Three Mile Pond	0.04	< 0.08	< 0.17	< 0.19	< 0.01	< 0.29	0.27	0.36	< 0.02	< 0.24	0.09	< 0.40	0.01	0.08	0.01	< 0.02
3MP-C DOSE	Three Mile Pond	0.04	< 0.08	< 0.17	< 0.19	< 0.01	< 0.29	0.26	0.36	< 0.02	< 0.24	0.09	< 0.40	0.01	0.08	0.01	< 0.02
Average		0.04						0.26	0.36			0.09		0.01	0.08	0.01	
SD(n-1)		0.00						0.00	0.00			0.00		0.00	0.00	0.00	
%RSD		1						0	1			1		8	2	1	
PS 113000 PCB*	Procedure Spike	0.90	6.0	14	40	0.35	15	15	42	2.1	24	3.2	46	0.07	< 0.07	1.2	1.0
Percent Recovery		***73***	***78***	***83***	***72***	***80***	***80***	***74***	***71***	***73***	***83***	***80***	***76***	***40***	***---***	***78***	***74***
PB 113000 GCR1	Procedure Blank	0.00	0.04	0.08	0.18	0.00	0.02	0.11	0.23	0.00	0.21	0.01	0.36	0.00	0.03	0.01	0.00
PB 113000 GCR2	Procedure Blank	0.00	0.01	0.00	0.17	0.00	0.12	0.13	0.27	0.01	0.21	0.01	0.33	0.00	0.00	0.01	0.00
PB 113000 GCR3	Procedure Blank	0.00	0.04	0.00	0.18	0.00	0.14	0.10	0.22	0.00	0.19	0.03	0.32	0.00	0.02	0.01	0.01
Average		0.00	0.03	0.03	0.17	0.00	0.09	0.11	0.24	0.00	0.20	0.02	0.34	0.00	0.02	0.01	0.00
Standard Deviation		0.00	0.02	0.05	0.01	0.00	0.06	0.02	0.03	0.01	0.01	0.01	0.02	0.00	0.02	0.00	0.01
MDL		0.00	0.08	0.17	0.19	0.00	0.29	0.16	0.32	0.02	0.24	0.05	0.40	0.00	0.07	0.01	0.02
MDL	Mass normalized to 1 g	0.01	0.08	0.17	0.19	0.01	0.29	0.16	0.32	0.02	0.24	0.05	0.40	0.01	0.07	0.01	0.02

Note: Values are rounded to 2 significant figures.

MDL < 0.01 set to 0.01 instrument detection limit

***Average of injection triplicate.**

Table 1. Housatonic Large Mouth Bass--Dosing Solutions--PCB Concentrations (ng/g) and Quality Control Data

Sample ID	Field ID	117	118	119	122	123	128	129	130	131	132	133	134	136	137	138	139
RP LMB	Rising Pond	31	260	53	0.82	6.5	150	23	85	5.0	200	67	36	47	39	1,400	5.7
RP-A DOSE	Rising Pond	29	250	50	0.67	6.0	140	19	77	4.4	190	63	34	44	36	1,300	3.7
RP-B DOSE	Rising Pond	29	260	49	0.75	5.9	140	21	77	4.6	190	62	33	44	36	1,300	5.2
RP-C DOSE	Rising Pond	29	250	50	0.77	6.0	140	20	77	4.4	190	62	33	44	36	1,300	4.4
Average		29	250	50	0.73	6.0	140	20	80	4.5	190	60	33	40	36	1,300	4.4
SD(n-1)		0.25	5.8	0.34	0.05	0.07	0.00	0.84	0.18	0.09	0.0	0.52	0.31	0.06	0.20	0.0	0.77
%RSD		1	2	1	7	1	0	4	0	2	0	1	1	0	1	0	17
WP LMB	Woods Pond	28	540	96	1.2	6.2	260	40	140	11	680	100	86	160	62	2,800	11
WP-A DOSE	Woods Pond	28	520	96	1.3	6.3	260	41	130	12	700	100	86	160	62	2,800	11
WP-B DOSE	Woods Pond	29	530	96	1.1	5.9	250	41	130	11	690	100	85	160	62	2,800	11
WP-C DOSE	Woods Pond	27	510	95	1.1	6.2	250	39	130	11	670	100	84	160	62	2,800	11
Average		28	520	96	1.1	6.1	250	40	130	11	690	100	85	160	62	2,800	11
SD(n-1)		0.58	10	0.56	0.12	0.21	5.8	0.82	0.0	0.24	15	0.0	0.74	0.0	0.33	0	0.14
%RSD		2	2	1	10	3	2	2	0	2	2	0	1	0	1	0	1
DRP LMB	Deep Reach Pond	58	890	120	2.3	10	430	97	200	24	1,500	140	150	290	100	5,300	26
DRP-A DOSE	Deep Reach Pond	53	870	110	1.7	8.5	390	87	180	21	1,300	120	140	270	91	4,700	23
DRP-B DOSE	Deep Reach Pond	52	830	110	2.0	8.6	380	83	180	21	1,200	130	130	270	89	4,700	22
DRP-C DOSE	Deep Reach Pond	57	860	110	1.8	9.9	420	95	190	22	1,300	140	140	280	97	5,100	24
Average		54	850	110	1.8	9.0	400	88	180	21	1,300	130	140	270	92	4,800	23
SD(n-1)		2.9	21	0	0.14	0.79	21	5.8	5.8	0.45	60	10	5.8	5.8	4.0	230	1.3
%RSD		5	2	0	8	9	5	7	3	2	5	8	4	2	4	5	5
3MP LMB	Three Mile Pond	< 0.02	0.52	< 0.04	< 0.01	< 0.01	0.13	0.02	0.05	< 0.02	< 0.33	0.03	0.05	0.05	0.05	0.99	< 0.02
3MP-A DOSE	Three Mile Pond	< 0.02	0.56	0.04	< 0.01	< 0.01	0.14	0.02	0.06	< 0.02	< 0.33	0.04	0.06	0.05	0.05	1.0	< 0.02
3MP-B DOSE	Three Mile Pond	< 0.02	0.55	< 0.04	< 0.01	< 0.01	0.14	0.02	0.06	< 0.02	< 0.33	0.04	0.05	0.05	0.05	1.1	< 0.02
3MP-C DOSE	Three Mile Pond	< 0.02	0.57	< 0.04	< 0.01	< 0.01	0.14	0.02	0.06	< 0.02	< 0.33	0.04	0.05	0.05	0.05	1.0	< 0.02
Average			0.56				0.14	0.02	0.06			0.04	0.05	0.05	0.05	1.0	
SD(n-1)			0.01				0.00	0.00	0.00			0.00	0.00	0.00	0.0	0.0	
%RSD			1				1	2	1			1	1	1.1	0.21	0.71	
PS 113000 PCB*	Procedure Spike	1.4	31	0.62	0.29	0.48	8.4	2.4	2.8	0.84	34	0.79	3.7	9.3	2.5	57	0.65
Percent Recovery		***94***	***75***	***92***	***82***	***98***	***82***	***89***	***79***	***81***	***82***	***75***	***79***	***77***	***79***	***81***	***88***
PB 113000 GCR1	Procedure Blank	0.01	0.32	0.02	0.00	0.00	0.03	0.00	0.01	0.00	0.02	0.00	0.00	0.02	0.00	0.30	0.00
PB 113000 GCR2	Procedure Blank	0.00	0.29	0.00	0.00	0.00	0.05	0.00	0.01	0.01	0.15	0.00	0.01	0.01	0.01	0.24	0.01
PB 113000 GCR3	Procedure Blank	0.00	0.33	0.00	0.00	0.01	0.01	0.00	0.00	0.00	0.15	0.00	0.01	0.01	0.01	0.29	0.01
Average		0.00	0.31	0.01	0.00	0.00	0.03	0.00	0.01	0.00	0.11	0.00	0.01	0.01	0.01	0.28	0.01
Standard Deviation		0.01	0.02	0.01	0.00	0.00	0.02	0.00	0.01	0.01	0.07	0.00	0.01	0.01	0.01	0.03	0.01
MDL		0.02	0.38	0.04	0.00	0.01	0.09	0.00	0.02	0.02	0.33	0.00	0.02	0.03	0.02	0.37	0.02
MDL	Mass normalized to 1 g	0.02	0.38	0.04	0.01	0.01	0.09	0.01	0.02	0.02	0.33	0.01	0.02	0.03	0.02	0.37	0.02
Note: Values are rounded to 2 significant figures.																	
MDL < 0.01 set to 0.01 instrument detection limit																	
***Average of injection triplicate.**																	

Table 1. Housatonic Large Mouth Bass--Dosing Solutions--PCB Concentrations (ng/g) and Quality Control Data

Sample ID	Field ID	141	144	146	147	149	151	153	156	157	158	163	164	166	167	170	171
RP LMB	Rising Pond	390	88	550	16	1,200	390	3,000	83	19	160	510	130	6.5	61	1,000	160
RP-A DOSE	Rising Pond	370	82	510	15	1,000	370	2,800	73	17	150	450	130	6.0	57	920	150
RP-B DOSE	Rising Pond	360	81	500	13	1,100	360	2,700	74	17	150	460	110	6.0	56	930	150
RP-C DOSE	Rising Pond	360	82	510	15	1,100	370	2,800	76	17	150	450	130	5.9	56	940	150
Average		360	80	510	14	1,100	370	2,800	70	17	150	450	120	5.9	60	930	150
SD(n-1)		5.8	0.43	5.8	0.73	58	5.8	58	1.8	0.04	0.0	5.8	12	0.06	0.25	10	0.0
%RSD		2	1	1	5	5	2	2	3	0	0	1	10	1	0	1	0
WP LMB	Woods Pond	860	230	870	19	2,500	970	5,000	140	23	320	850	290	8.2	92	1,600	360
WP-A DOSE	Woods Pond	860	230	870	19	2,500	960	4,900	120	21	320	840	280	7.7	92	1,600	350
WP-B DOSE	Woods Pond	850	230	870	18	2,400	960	4,900	130	22	320	820	290	7.8	92	1,600	350
WP-C DOSE	Woods Pond	840	230	860	18	2,400	950	4,800	130	21	310	820	290	7.6	90	1,600	350
Average		850	230	870	18	2,400	960	4,900	130	21	320	830	290	7.7	91	1,600	350
SD(n-1)		10	0.0	5.8	0.96	58	5.8	58	5.8	0.74	5.8	12	5.8	0.11	1.1	0.0	0.0
%RSD		1	0	1	5	2	1	1	4	4	2	1	2	1	1	0	0
DRP LMB	Deep Reach Pond	1,500	360	1,400	36	4,400	1,400	8,200	180	29	590	1,500	290	11	160	2,900	640
DRP-A DOSE	Deep Reach Pond	1,400	330	1,200	28	3,900	1,300	7,400	160	25	520	1,300	270	10	140	2,500	590
DRP-B DOSE	Deep Reach Pond	1,400	340	1,200	30	4,000	1,300	7,400	160	27	520	1,300	410	10	140	2,600	590
DRP-C DOSE	Deep Reach Pond	1,500	340	1,300	33	4,100	1,400	7,800	160	26	560	1,400	320	11	150	2,600	590
Average		1,400	340	1,200	30	4,000	1,300	7,500	160	26	530	1,300	330	10	140	2,600	590
SD(n-1)		60	6	60	2.4	100	60	230	0	1	23	60	70	0.3	6	60	0
%RSD		4	2	5	8	3	5	3	0	5	4	5	21	3	4	2	0
3MP LMB	Three Mile Pond	0.13	0.03	0.29	< 0.01	0.29	0.11	1.5	0.04	< 0.04	0.08	0.22	< 0.04	< 0.01	0.04	0.40	0.06
3MP-A DOSE	Three Mile Pond	0.14	0.03	0.31	< 0.01	0.31	0.12	1.6	0.04	< 0.04	0.09	0.23	0.04	< 0.01	0.04	0.42	0.06
3MP-B DOSE	Three Mile Pond	0.14	0.03	0.31	< 0.01	0.31	0.12	1.6	0.04	< 0.04	0.09	0.24	0.04	< 0.01	0.04	0.41	0.06
3MP-C DOSE	Three Mile Pond	0.13	0.03	0.31	< 0.01	0.31	0.11	1.6	0.06	< 0.04	0.09	0.23	0.04	< 0.01	0.04	0.43	0.06
Average		0.13	0.03	0.31		0.31	0.12	1.6	0.05		0.09	0.23	0.04		0.04	0.42	0.06
SD(n-1)		0.00	0.00	0.00		0.00	0.00	0.01	0.01		0.00	0.00	0.0		0.0	0.01	0.00
%RSD		0.5	1.4	0.4		0.5	1.1	0.5	16		2	1	2		1	4	5
PS 113000 PCB*	Procedure Spike	16	4.7	6.9	0.20	45	15	51	3.0	0.78	7.1	12	4.3	0.17	1.5	19	5.7
Percent Recovery		***80***	***79***	***80***	***65***	***77***	***80***	***78***	***85***	***68***	***76***	***81***	***79***	***76***	***75***	***83***	***80***
PB 113000 GCR1	Procedure Blank	0.06	0.00	0.15	0.00	0.22	0.02	1.1	0.02	0.01	0.00	0.02	0.00	0.00	0.00	0.00	0.00
PB 113000 GCR2	Procedure Blank	0.05	0.00	0.13	0.00	0.24	0.04	1.2	0.02	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00
PB 113000 GCR3	Procedure Blank	0.06	0.00	0.14	0.00	0.21	0.03	1.0	0.01	0.00	0.04	0.01	0.00	0.00	0.00	0.00	0.00
Average		0.06	0.00	0.14	0.00	0.22	0.03	1.1	0.02	0.00	0.02	0.02	0.01	0.00	0.00	0.00	0.00
Standard Deviation		0.01	0.00	0.01	0.00	0.02	0.01	0.09	0.01	0.00	0.02	0.01	0.01	0.00	0.00	0.00	0.00
MDL		0.07	0.00	0.17	0.00	0.27	0.06	1.4	0.03	0.01	0.08	0.03	0.04	0.00	0.00	0.00	0.00
MDL	Mass normalized to 1 g	0.07	0.01	0.17	0.01	0.27	0.06	1.4	0.03	0.04	0.08	0.03	0.04	0.01	0.01	0.01	0.01

Note: Values are rounded to 2 significant figures.

MDL < 0.01 set to 0.01 instrument detection limit

***Average of injection triplicate.**

Table 1. Housatonic Large Mouth Bass--Dosing Solutions--PCB Concentrations (ng/g) and Quality Control Data

Sample ID	Field ID	172	173	174	175	176	177	178	179	180	183	185	187	189	190	191	193
RP LMB	Rising Pond	150	7.3	390	32	15	280	140	58	1,800	540	65	1,100	31	230	30	120
RP-A DOSE	Rising Pond	140	6.5	360	30	11	260	140	54	1,700	510	60	1,100	29	210	28	110
RP-B DOSE	Rising Pond	140	6.2	360	31	13	260	140	55	1,700	510	61	1,100	29	210	28	110
RP-C DOSE	Rising Pond	140	6.6	370	31	8.6	260	140	54	1,700	510	61	1,100	29	210	28	110
Average		140	6.4	360	31	11	260	140	54	1,700	510	61	1,100	29	210	28	110
SD(n-1)		0.00	0.20	5.8	0.42	2.0	0.0	0.00	0.57	0.0	0.0	0.47	0.0	0.19	0.0	0.17	0.00
%RSD		0	3	2	1	19	0	0	1	0	0	1	0	1	0	1	0
WP LMB	Woods Pond	270	21	1,100	68	86	610	310	230	3,300	1,200	150	2,200	55	490	62	210
WP-A DOSE	Woods Pond	270	24	1,100	66	93	590	300	230	3,200	1,100	150	2,100	53	480	61	210
WP-B DOSE	Woods Pond	270	22	1,100	66	88	600	300	230	3,200	1,100	150	2,100	53	480	60	210
WP-C DOSE	Woods Pond	260	26	1,100	66	89	590	300	220	3,200	1,100	150	2,100	53	480	62	210
Average		270	24	1,100	66	90	590	300	230	3,200	1,100	150	2,100	53	480	61	210
SD(n-1)		5.8	1.9	0.0	0.08	2.8	5.8	0.0	5.8	0.0	0.0	0.0	0.0	0.08	0.0	0.81	0.0
%RSD		2	8	0	0	3	1	0	3	0	0	0	0	0	0	1	0
DRP LMB	Deep Reach Pond	520	53	2,100	53	220	1,100	520	380	6,200	2,300	270	4,400	110	1,100	120	420
DRP-A DOSE	Deep Reach Pond	480	46	1,900	50	200	1,000	490	350	5,700	2,100	250	4,100	100	1,000	120	390
DRP-B DOSE	Deep Reach Pond	470	48	2,000	67	170	1,000	490	340	5,700	2,100	250	4,100	100	990	110	390
DRP-C DOSE	Deep Reach Pond	480	50	1,900	49	180	1,000	480	350	5,800	2,100	250	4,000	100	990	120	400
Average		480	48	1,900	55	180	1,000	490	350	5,700	2,100	250	4,100	100	990	120	390
SD(n-1)		6	2	60	10	20	0	10	10	60	0	0	60	0.0	10	5.8	5.8
%RSD		1	4	3	19	11	0	2	3	1	0	0	1	0	1	5	1
3MP LMB	Three Mile Pond	0.07	< 0.02	0.09	< 0.10	0.02	0.08	0.04	< 0.06	0.86	0.27	0.02	0.56	< 0.02	0.17	< 0.01	0.06
3MP-A DOSE	Three Mile Pond	0.08	< 0.02	0.09	< 0.10	0.02	0.08	0.04	< 0.06	0.91	0.28	0.02	0.59	0.02	0.14	< 0.01	0.06
3MP-B DOSE	Three Mile Pond	0.08	< 0.02	0.09	< 0.10	0.02	0.08	0.04	< 0.06	0.91	0.28	0.02	0.60	0.02	0.14	< 0.01	0.06
3MP-C DOSE	Three Mile Pond	0.08	< 0.02	0.10	< 0.10	0.02	0.08	0.04	< 0.06	0.90	0.30	0.02	0.60	0.02	0.16	< 0.01	0.06
Average		0.08		0.09		0.02	0.08	0.04		0.91	0.29	0.02	0.60	0.02	0.15		0.06
SD(n-1)		0.0		0.00		0.00	0.00	0.00		0.00	0.01	0.00	0.01	0.00	0.01		0.00
%RSD		0		4		5	1	1		1	4	5	1	2	10		1
PS 113000 PCB*	Procedure Spike	3.5	0.65	20	0.71	2.7	11	3.7	7.6	48	16	2.3	20	0.90	9.1	0.60	2.3
Percent Recovery		***78***	***86***	***80***	***88***	***88***	***82***	***87***	***81***	***84***	***81***	***80***	***81***	***87***	***84***	***91***	***79***
PB 113000 GCR1	Procedure Blank	0.00	0.00	0.00	0.04	0.00	0.03	0.00	0.00	0.03	0.04	0.00	0.06	0.01	0.00	0.00	0.00
PB 113000 GCR2	Procedure Blank	0.00	0.01	0.01	0.05	0.00	0.01	0.00	0.00	0.02	0.01	0.00	0.05	0.00	0.00	0.00	0.00
PB 113000 GCR3	Procedure Blank	0.00	0.01	0.00	0.01	0.00	0.00	0.01	0.03	0.02	0.02	0.00	0.05	0.00	0.02	0.00	0.00
Average		0.00	0.01	0.00	0.03	0.00	0.01	0.00	0.01	0.02	0.02	0.00	0.05	0.00	0.01	0.00	0.00
Standard Deviation		0.00	0.01	0.01	0.02	0.00	0.02	0.01	0.02	0.01	0.02	0.00	0.01	0.01	0.01	0.00	0.00
MDL		0.00	0.02	0.02	0.10	0.00	0.06	0.02	0.06	0.04	0.07	0.00	0.07	0.02	0.03	0.00	0.00
MDL	Mass normalized to 1 g	0.01	0.02	0.02	0.10	0.01	0.06	0.02	0.06	0.04	0.07	0.01	0.07	0.02	0.03	0.01	0.01
Note: Values are rounded to 2 significant figures.																	
MDL < 0.01 set to 0.01 instrument detection limit																	
***Average of injection triplicate.**																	

Table 1. Housatonic Large Mouth Bass--Dosing Solutions--PCB Concentrations (ng/g) and Quality Control Data

Sample ID	Field ID	194	195	196	197	198	199	200	201	202	203	205	206	208	209	Total PCBs	
RP LMB	Rising Pond	270	120	100	13	15	240	9.9	23	33	230	18	40	3.5	7.9	19,000	ng/g
RP-A DOSE	Rising Pond	250	99	110	12	13	220	9.6	21	30	210	16	37	3.3	7.7	18,000	ng/g
RP-B DOSE	Rising Pond	250	100	110	12	13	220	9.4	21	30	210	16	37	3.3	7.7	19,000	ng/g
RP-C DOSE	Rising Pond	250	100	100	12	13	230	9.5	21	31	210	17	38	3.4	7.9	18,000	ng/g
Average		250	100	110	12	13	220	9.5	21	30	210	16	40	3.3	7.7	18,000	ng/g
SD(n-1)		0.00	0.5	5.8	0.18	0.07	5.8	0.12	0.03	0.56	0.0	0.28	0.34	0.07	0.11	580	ng/g
%RSD		0	0	5	2	0	3	1	0	2	0	2	1	2	1	3	
WP LMB	Woods Pond	530	140	300	40	27	560	46	61	87	430	31	92	12	30	38,000	ng/g
WP-A DOSE	Woods Pond	520	110	300	42	28	550	46	62	85	420	34	90	12	29	37,000	ng/g
WP-B DOSE	Woods Pond	520	120	300	40	27	540	45	60	84	420	33	89	12	29	37,000	ng/g
WP-C DOSE	Woods Pond	520	120	290	40	26	540	45	60	88	420	33	89	12	29	37,000	ng/g
Average		520	120	300	41	27	540	45	61	86	420	33	89	12	29	37,000	ng/g
SD(n-1)		0.0	5.8	5.8	1.3	1.0	5.8	0.54	0.95	1.7	0.0	0.18	0.30	0.16	0.35	0	ng/g
%RSD		0	5	2	3	4	1	1	2	2	0	1	0	1	1	0	
DRP LMB	Deep Reach Pond	1,100	370	640	100	46	1,100	110	120	160	830	71	170	29	85	67,000	ng/g
DRP-A DOSE	Deep Reach Pond	990	360	610	93	43	1,000	100	110	150	760	63	150	26	81	61,000	ng/g
DRP-B DOSE	Deep Reach Pond	970	300	570	89	48	990	99	110	150	760	63	150	26	78	61,000	ng/g
DRP-C DOSE	Deep Reach Pond	990	360	590	93	43	1,000	100	110	150	760	61	150	26	84	63,000	ng/g
Average		980	340	590	92	45	1,000	100	110	150	760	62	150	26	81	62,000	ng/g
SD(n-1)		10	30	20	2.4	2.6	10	0.4	0	0	0	1	0	0.5	3.21	1,200	ng/g
%RSD		1	9	3	3	6	1	0.4	0	0	0	1	0	2	4	2	
3MP LMB	Three Mile Pond	0.13	0.05	< 0.19	< 0.15	< 0.01	0.16	< 0.02	< 0.02	0.06	0.16	0.01	0.10	0.02	0.04	28	ng/g
3MP-A DOSE	Three Mile Pond	0.13	0.06	< 0.19	< 0.15	< 0.01	0.17	< 0.02	< 0.02	0.06	0.15	0.01	0.10	0.02	0.05	30	ng/g
3MP-B DOSE	Three Mile Pond	0.14	0.06	< 0.19	< 0.15	< 0.01	0.17	< 0.02	< 0.02	0.06	0.16	0.01	0.10	0.03	0.05	30	ng/g
3MP-C DOSE	Three Mile Pond	0.14	0.06	< 0.19	< 0.15	< 0.01	0.18	< 0.02	< 0.02	0.07	0.17	0.01	0.10	0.02	0.05	30	ng/g
Average		0.13	0.06				0.17			0.06	0.16	0.01	0.10	0.02	0.05	30	ng/g
SD(n-1)		0.00	0.00				0.00			0.00	0.01	0.00	0.00	0.01	0.00	0.02	ng/g
%RSD		1	2				1			3	6	2	0.3	25	1	0.1	
PS 113000 PCB*	Procedure Spike	7.3	4.4	5.5	0.78	0.37	7.0	2.1	1.1	2.2	5.1	0.68	1.7	0.50	< 2.5	1,400	ng
Percent Recovery		***84***	***80***	***82***	***81***	***80***	***84***	***89***	***82***	***86***	***82***	***86***	***88***	***88***	***---***	***74***	%
PB 113000 GCR1	Procedure Blank	0.00	0.00	0.16	0.08	0.00	0.02	0.01	0.00	0.00	0.01	0.00	0.01	0.20	2.4	18	ng
PB 113000 GCR2	Procedure Blank	0.01	0.01	0.14	0.01	0.00	0.02	0.00	0.00	0.01	0.03	0.00	0.01	0.15	2.4	17	ng
PB 113000 GCR3	Procedure Blank	0.00	0.00	0.13	0.05	0.00	0.00	0.01	0.01	0.00	0.03	0.00	0.01	0.19	2.4	17	ng
Average		0.00	0.00	0.14	0.05	0.00	0.01	0.01	0.00	0.00	0.02	0.00	0.01	0.18	2.4	17	ng
Standard Deviation		0.01	0.01	0.02	0.04	0.00	0.01	0.01	0.01	0.01	0.01	0.00	0.00	0.03	0.04	0.79	ng
MDL		0.02	0.02	0.19	0.15	0.00	0.05	0.02	0.02	0.02	0.06	0.00	0.01	0.26	2.5	20	ng
MDL	Mass normalized to 1 g	0.02	0.02	0.19	0.15	0.01	0.05	0.02	0.02	0.02	0.06	0.01	0.01	0.26	2.5	20	ng/g

Note: Values are rounded to 2 significant figures.

MDL < 0.01 set to 0.01 instrument detection limit

***Average of injection triplicate.**

Table 2. Recoveries of Procedural Standards in Housatonic Dosing Solutions

Sample ID	Field ID	Sample Type	029 % Recovery	155 % Recovery	204 % Recovery
RP LMB	Rising Pond	Dosing Solution	***63***	***71***	***80***
RP-A DOSE	Rising Pond	Dosing Solution	***63***	***69***	***76***
RP-B DOSE	Rising Pond	Dosing Solution	***67***	***75***	***82***
RP-C DOSE	Rising Pond	Dosing Solution	***66***	***73***	***79***
WP LMB	Woods Pond	Dosing Solution	***59***	***65***	***75***
WP-A DOSE	Woods Pond	Dosing Solution	***56***	***61***	***72***
WP-B DOSE	Woods Pond	Dosing Solution	***56***	***63***	***74***
WP-C DOSE	Woods Pond	Dosing Solution	***58***	***64***	***75***
DRP LMB	Deep Reach Pond	Dosing Solution	***57***	***60***	***69***
DRP-A DOSE	Deep Reach Pond	Dosing Solution	***61***	***66***	***74***
DRP-B DOSE	Deep Reach Pond	Dosing Solution	***65***	***70***	***79***
DRP-C DOSE	Deep Reach Pond	Dosing Solution	***56***	***61***	***73***
3MP LMB	Three Mile Pond	Dosing Solution	***74***	***77***	***78***
3MP-A DOSE	Three Mile Pond	Dosing Solution	***74***	***75***	***77***
3MP-B DOSE	Three Mile Pond	Dosing Solution	***73***	***74***	***75***
3MP-C DOSE	Three Mile Pond	Dosing Solution	***78***	***79***	***80***
PS 113000 PCB GCR1	Procedure Spike	Na_2SO_4	***68***	***69***	***78***
PS 113000 PCB GCR2	Procedure Spike	Na_2SO_4	***67***	***69***	***78***
PS 113000 PCB GCR3	Procedure Spike	Na_2SO_4	***67***	***70***	***78***
PB 113000 GCR1	Procedure Blank	Na_2SO_4	***55***	***64***	***79***
PB 113000 GCR2	Procedure Blank	Na_2SO_4	***55***	***64***	***79***
PB 113000 GCR3	Procedure Blank	Na_2SO_4	***53***	***64***	***79***
Average			**61**	**67**	**78**
SD			**7**	**3**	**1**

Table 3. Organochlorine Pesticides in Dosing Solutions (ng/g)

Sample ID	Site ID	Sample Type	Gram-equivalents for Analysis (g)	HCB	PCA	alpha-BHC	Lindane	delta-BHC	Heptachlor	Heptachlor Epoxide	Oxychlordane
RP LMB	**Rising Pond**	Dosing Solution	12,800	**0.44**	**0.12**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**0.48**	**0.22**
RP-A DOSE	Rising Pond	Dosing Solution	12,800	0.44	0.08	< 0.01	< 0.01	< 0.01	< 0.01	0.51	0.20
RP-B DOSE	Rising Pond	Dosing Solution	12,800	0.41	0.07	< 0.01	< 0.01	< 0.01	< 0.01	0.52	0.19
RP-C DOSE	Rising Pond	Dosing Solution	12,800	0.42	0.08	< 0.01	< 0.01	< 0.01	< 0.01	0.46	0.19
Average				**0.42**	**0.08**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**0.50**	**0.19**
SD(n-1)				0.01	0.00					0.04	0.01
%RSD				3	3					7	4
WP LMB	**Woods Pond**	Dosing Solution	14,200	**1.5**	**0.17**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**
WP-A DOSE	Woods Pond	Dosing Solution	14,200	1.6	0.71	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
WP-B DOSE	Woods Pond	Dosing Solution	14,200	1.2	0.53	0.18	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
WP-C DOSE	Woods Pond	Dosing Solution	14,200	1.4	0.34	0.34	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
Average				**1.4**	**0.53**		**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**
SD(n-1)				0.18	0.18						
%RSD				13	35						
DRP LMB	**Deep Reach Pond**	Dosing Solution	13,000	**3.5**	**1.0**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**
DRP-A DOSE	Deep Reach Pond	Dosing Solution	13,000	2.8	0.46	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
DRP-B DOSE	Deep Reach Pond	Dosing Solution	13,000	2.6	0.87	0.43	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
DRP-C DOSE	Deep Reach Pond	Dosing Solution	13,000	3.0	1.5	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
Average				**2.8**	**0.94**		**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**
SD(n-1)				0.19	0.52						
%RSD				7	55						
3MP LMB	**Three Mile Pond**	Dosing Solution	15,200	**< 0.05**	**< 0.03**	**< 0.01**	**< 0.01**	**< 0.01**	**< 0.01**	**0.06**	**0.06**
3MP-A DOSE	Three Mile Pond	Dosing Solution	15,200	< 0.05	< 0.03	< 0.01	< 0.01	< 0.01	< 0.01	0.07	0.06
3MP-B DOSE	Three Mile Pond	Dosing Solution	15,200	< 0.05	< 0.03	< 0.01	< 0.01	< 0.01	< 0.01	0.07	0.06
3MP-C DOSE	Three Mile Pond	Dosing Solution	15,200	< 0.05	< 0.03	< 0.01	0.01	< 0.01	< 0.01	0.07	0.06
Average				**< 0.05**	**< 0.03**	**< 0.01**		**< 0.01**	**< 0.01**	**0.07**	**0.06**
SD(n-1)										0.00	0.00
%RSD										1	1
PB 113000 GCR1	Procedure Blank	Na_2SO_4	---	0.04	0.02	0.00	0.00	0.00	0.00	0.00	0.00
PB 113000 GCR2	Procedure Blank	Na_2SO_4	---	0.03	0.01	0.00	0.00	0.00	0.00	0.00	0.00
PB 113000 GCR3	Procedure Blank	Na_2SO_4	---	0.04	0.01	0.00	0.00	0.00	0.00	0.00	0.00
Average				0.04	0.01	0.00	0.00	0.00	0.00	0.00	0.00
Standard Deviation				0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00
Method Detection Limit	*MDL=*	PB Average + 3 (SD)		0.05	0.03	0.00	0.00	0.00	0.00	0.00	0.00
MDL				**0.05**	**0.03**	**0.01**	**0.01**	**0.01**	**0.01**	**0.01**	**0.01**
Note: Values are rounded to 2 significant figures.											
MDL < 0.01 set to 0.01 instrument detection limit											

Table 3. Organochlorine Pesticides in Dosing Solutions (ng/g)

Sample ID	Site ID	cis-Chlordane	trans-Chlordane	cis-Nonachlor	trans-Nonachlor	o,p'-DDE	o,p'-DDD	o,p'-DDT	p,p'-DDE	p,p'-DDD	p,p'-DDT	Endosulfan 1
RP LMB	**Rising Pond**	**< 0.02**	**< 0.01**	**0.35**	**2.6**	**< 0.01**	**< 0.01**	**< 0.01**	**36**	**4.7**	**0.73**	**0.44**
RP-A DOSE	Rising Pond	< 0.02	< 0.01	0.33	2.6	< 0.01	< 0.01	< 0.01	36	4.2	0.64	0.40
RP-B DOSE	Rising Pond	< 0.02	0.09	0.37	2.6	< 0.01	< 0.01	< 0.01	34	4.0	0.59	0.40
RP-C DOSE	Rising Pond	< 0.02	0.10	0.48	2.6	< 0.01	< 0.01	< 0.01	35	4.0	0.67	0.39
Average		**< 0.02**		**0.40**	**2.6**	**< 0.01**	**< 0.01**	**< 0.01**	**35**	**4.1**	**0.63**	**0.40**
SD(n-1)				0.08	0.01				0.57	0.10	0.04	0.01
%RSD				19	1				2	2	7	3
WP LMB	**Woods Pond**	**< 0.02**	**0.27**	**< 0.01**	**2.0**	**< 0.01**	**< 0.01**	**< 0.01**	**43**	**3.3**	**< 0.01**	**< 0.01**
WP-A DOSE	Woods Pond	< 0.02	< 0.01	< 0.01	1.8	< 0.01	< 0.01	< 0.01	31	1.3	< 0.01	< 0.01
WP-B DOSE	Woods Pond	< 0.02	< 0.01	< 0.01	1.9	< 0.01	< 0.01	< 0.01	31	2.0	< 0.01	< 0.01
WP-C DOSE	Woods Pond	< 0.02	0.27	< 0.01	1.9	< 0.01	< 0.01	< 0.01	32	0.8	< 0.01	< 0.01
Average		**< 0.02**		**< 0.01**	**1.9**	**< 0.01**	**< 0.01**	**< 0.01**	**31**	**1.4**	**< 0.01**	**< 0.01**
SD(n-1)					0.04				0.49	0.62		
%RSD					2				2	45		
DRP LMB	**Deep Reach Pond**	**3.8**	**< 0.01**	**< 0.01**	**3.3**	**< 0.01**	**< 0.01**	**< 0.01**	**48**	**5.0**	**< 0.01**	**< 0.01**
DRP-A DOSE	Deep Reach Pond	3.6	< 0.01	< 0.01	2.8	< 0.01	< 0.01	< 0.01	52	8.3	< 0.01	< 0.01
DRP-B DOSE	Deep Reach Pond	3.6	< 0.01	< 0.01	2.2	< 0.01	< 0.01	< 0.01	39	2.9	< 0.01	< 0.01
DRP-C DOSE	Deep Reach Pond	3.5	< 0.01	< 0.01	2.8	< 0.01	< 0.01	< 0.01	37	3.9	< 0.01	< 0.01
Average		**3.6**	**< 0.01**	**< 0.01**	**2.6**	**< 0.01**	**< 0.01**	**< 0.01**	**42**	**5.0**	**< 0.01**	**< 0.01**
SD(n-1)		0.06			0.33				8.2	2.85		
%RSD		2			13				19	56		
3MP LMB	**Three Mile Pond**	**0.04**	**0.01**	**0.05**	**0.20**	**< 0.01**	**< 0.01**	**< 0.01**	**14**	**0.63**	**0.06**	**0.18**
3MP-A DOSE	Three Mile Pond	0.04	0.01	0.06	0.21	< 0.01	< 0.01	< 0.01	14	0.67	0.06	0.18
3MP-B DOSE	Three Mile Pond	0.04	0.01	0.06	0.22	< 0.01	< 0.01	< 0.01	15	0.68	0.06	0.18
3MP-C DOSE	Three Mile Pond	0.04	0.01	0.06	0.22	< 0.01	< 0.01	< 0.01	14	0.68	0.06	0.18
Average		**0.04**	**0.01**	**0.06**	**0.22**	**< 0.01**	**< 0.01**	**< 0.01**	**14**	**0.67**	**0.06**	**0.18**
SD(n-1)		0.00	0.00	0.00	0.00				0.09	0.00	0.00	0.00
%RSD		0	0	0	1				1	1	0	1
PB 113000 GCR1	Procedure Blank	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00
PB 113000 GCR2	Procedure Blank	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.10	0.00	0.00	0.00
PB 113000 GCR3	Procedure Blank	0.01	0.00	0.00	0.01	0.00	0.00	0.00	0.01	0.00	0.00	0.00
Average		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00
Standard Deviation		0.01	0.00	0.00	0.01	0.00	0.00	0.00	0.05	0.00	0.00	0.00
Method Detection Limit	*MDL=*	0.02	0.00	0.00	0.02	0.00	0.00	0.00	0.19	0.00	0.00	0.00
MDL		**0.02**	**0.01**	**0.01**	**0.02**	**0.01**	**0.01**	**0.01**	**0.19**	**0.01**	**0.01**	**0.01**

Note: Values are rounded to 2 significant figures.

MDL < 0.01 set to 0.01 instrument detection limit

Table 3. Organochlorine Pesticides in Dosing Solutions (ng/g)

Sample ID	Site ID	Endosulfan II	Endosulfate	Mirex
RP LMB	**Rising Pond**	**< 0.01**	**< 0.06**	**< 0.01**
RP-A DOSE	Rising Pond	< 0.01	< 0.06	< 0.01
RP-B DOSE	Rising Pond	< 0.01	< 0.06	< 0.01
RP-C DOSE	Rising Pond	< 0.01	0.32	< 0.01
Average		**< 0.01**		**< 0.01**
SD(n-1)				
%RSD				
WP LMB	**Woods Pond**	**< 0.01**	**0.27**	**< 0.01**
WP-A DOSE	Woods Pond	< 0.01	1.8	< 0.01
WP-B DOSE	Woods Pond	< 0.01	1.1	< 0.01
WP-C DOSE	Woods Pond	< 0.01	0.27	< 0.01
Average		**< 0.01**	**1.1**	**< 0.01**
SD(n-1)			0.78	
%RSD			73	
DRP LMB	**Deep Reach Pond**	**< 0.01**	**0.4**	**< 0.01**
DRP-A DOSE	Deep Reach Pond	< 0.01	< 0.06	< 0.01
DRP-B DOSE	Deep Reach Pond	< 0.01	< 0.06	< 0.01
DRP-C DOSE	Deep Reach Pond	< 0.01	< 0.06	< 0.01
Average		**< 0.01**	**< 0.06**	**< 0.01**
SD(n-1)				
%RSD				
3MP LMB	**Three Mile Pond**	**< 0.01**	**< 0.06**	**< 0.01**
3MP-A DOSE	Three Mile Pond	< 0.01	< 0.06	< 0.01
3MP-B DOSE	Three Mile Pond	< 0.01	< 0.06	< 0.01
3MP-C DOSE	Three Mile Pond	< 0.01	< 0.06	< 0.01
Average		**< 0.01**	**< 0.06**	**< 0.01**
SD(n-1)				
%RSD				
PB 113000 GCR1	Procedure Blank	0.00	0.00	0.00
PB 113000 GCR2	Procedure Blank	0.00	0.00	0.00
PB 113000 GCR3	Procedure Blank	0.00	0.03	0.00
Average		0.00	0.01	0.00
Standard Deviation		0.00	0.02	0.00
Method Detection Limit	*MDL=*	0.00	0.06	0.00
MDL		**0.01**	**0.06**	**0.01**
Note: Values are rounded to 2 significant figures.				
MDL < 0.01 set to 0.01 instrument detection limit				

Table 4. Non-*o*-Chloro-Substituted PCBs (pg/g-Equiv.) in Dosing Solutions of Large Scale Extraction of Fish Tissue from the Housatonic River

Revised: 11/20/2001
N46pcb-dosingsolns.xls

GC/MS Set: N46PCB
Dates: Jan. 12-14, 2000

NFCR Number:	Field Number:	Sample Description:	GC/MS Run No.	Non-o-Polychlorinated Biphenyls: Tetra: 3,4,4',5-TCB (81)	Tetra: 3,3',4,4'-TCB (77)	Penta: 3,3',4,4',5-PeCB (126)	Hexa: 3,3',4,4',5,5'-HxCB (169)
		Procedure Blank (152 g Equiv. Basis) for 3MP Dosing Solns:		**0.03 IF,LQ**	**0.1 IF**	**0.05 LQ**	**0.01 LQ**
		Procedure Blank (10 g Equiv.) for Comparative 10g Fish Samples		**0.4 IF,LQ**	**7.1**	**4.1**	**0.6 LQ**
	3MP-LMB	3 Mile Pond,3MP-LMB,Dilution Factor 100 (152 g)	46-7	0.4	7.2	2.6	0.4 LQ
	3MP-A	3 Mile Pond, 3MP-A, Dilution Factor 100 (152 g)	46-9	0.4	8.0	3.0	0.4
		(from 10g Fish, 3MP-B)		**2.2 IF**	**52**	**15**	**2.1** LQ
	3MP-B	3 Mile Pond, 3MP-B, Dilution Factor 100 (152 g)	46-10	0.5	7.7	3.0	0.5
		(from 10g Fish, 3MP-C)		**1.9 IF**	**32**	**13**	**1.8**
	3MP-C	3 Mile Pond, 3MP-C, Dilution Factor 100 (152 g)	46-11	0.5	8.1	3.0	0.5
		(from 10g Fish, 3MP-D)		**0.6 IF,LQ**	**27**	**12**	**1.6**
		Procedure Blank (0.41 g Equiv. Basis) for RP Dosing Solns:		**12 IF,LQ**	**48 IF**	**17 LQ**	**3.9 LQ**
	RP-LMB	Rising Pond, RP-LMB, Dilutn Factor 31,250 (0.41g)	46-12	31 IF,LQ	840	1,500	220
	RP-A	Rising Pond, RP-A, Dilution Factor 31,250 (0.41g)	46-14	36 IF	870	1,500	220
		(from 10g Fish, RP-A)		**53**	**1,800**	**3,100**	**540**
	RP-B	Rising Pond, RP-B, Dilution Factor 31,250 (0.41g)	46-15	31 IF	830	1,500	210
		(from 10g Fish, RP-B)		**74**	**2,200**	**3,200**	**570**
	RP-C	Rising Pond, RP-30-9, Dilution Factor 31,250 (0.41g)	46-16	43	730	1,400	210
		(from 10g Fish, RP-C)		**66**	**2,000**	**3,300**	**600**
		Procedure Blank (0.102 g Equiv. Basis) for WP Dosing Solns:		**49 IF,LQ**	**190 IF**	**69 LQ**	**16 LQ**
	WP-LMB	Woods Pond, WP-LMB, D.F. 138,889 (0.102g)	46-17	73 IF,LQ	1,500	2,600	310
	WP-A	Woods Pond, WP-A, Dilution Factor 138,889 (0.102g)	46-19	110 IF,LQ	1,600 LQ	2,300	320
		(from 10g Fish, WP-A)		**350**	**10,000**	**10,000**	**1,600**
	WP-B	Woods Pond, WP-B, Dilution Factor 138,889 (0.102g)	46-20	62 IF,LQ	1,800	2,400	270 LQ
		(from 10g Fish, WP-B)		**290**	**7,200**	**8,400**	**1,300**
	WP-C	Woods Pond, WP-C, Dilution Factor 138,889 (0.102g)	46-21	77 IF	1,600	2,400	310
		(from 10g Fish, WP-C)		**210 IF**	**6,200**	**8,200**	**1,300**

Table 4. Non-*o*-Chloro-Substituted PCBs (pg/g-Equiv.) in Dosing Solutions of Large Scale Extraction of Fish Tissue from the Housatonic River

Revised: 11/20/2001
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GC/MS Set: N46PCB
Dates: Jan. 12-14, 2000

NFCR Number:	Field Number:	Sample Description:	GC/MS Run No.	Non-o-Polychlorinated Biphenyls: Tetra: 3,4,4',5-TCB (81)	Tetra: 3,3',4,4'-TCB (77)	Penta: 3,3',4,4',5-PeCB (126)	Hexa: 3,3',4,4',5,5'-HxCB (169)
		Procedure Blank (0.036 g Equiv. Basis) for DRP Dosing Solns:		**140 IF,LQ**	**540 IF**	**200 LQ**	**45 LQ**
	DRP-LMB	Deep Reach Pond, DRP-LMB, D.F. 357,143 (0.036g)	46-22	140 IF,LQ	2,200 LQ	3,300 LQ	350
	DRP-A	Deep Reach Pond, DRP-A, Dilution F. 357,143 (0.036g)	46-24	210 IF,LQ	2,000	2,900	310 LQ
		(from 10g Fish, DRP-A)		**480**	**11,000**	**12,000**	**2,000**
	DRP-B	Deep Reach Pond, DRP-B, Dilution F. 357,143 (0.036g)	46-25	215 IF,LQ	1,600 LQ	2,500 LQ	390
		(from 10g Fish, DRP-B)		**300** IF	**6,900**	**8,100**	**1,300**
	DRP-C	Deep Reach Pond, DRP-C, Dilution F. 357,143 (0.036g)	46-26	156 IF	2,500	2,000 LQ	380 LQ
		(from 10g Fish, DRP-C)		**220**	**4,600**	**5,600**	**1,000**
		Quality Control Samples:					
Proc. Blk #1, 11/30/2000		**Procedure Blank, 11/30/2000 (TOTAL Basis)**	**46-4**	**5.0 IF,LQ**	**20 IF**	**7 LQ**	**1.6 LQ**
		(Based on 152 g Equiv.)		0.03 IF,LQ	0.1 **IF**	0.05 LQ	0.01 LQ
		(Based on 0.41 g Equiv.)		**12 IF,LQ**	**48 IF**	**17 LQ**	**3.9 LQ**
		(Based on 0.102 g Equiv.)		49 IF,LQ	191 **IF**	69 LQ	16 LQ
		(Based on 0.036 g Equiv.)		**140 IF,LQ**	**542 IF**	**200 LQ**	**45 LQ**
Proc. Spike 11/30/2000		Procedure Spike, 11/30/2000 (TOTAL BASIS) (Spiked with 2 µg Aroclors total)	46-5	180	2,600	73	0.5 LQ

LQ Less than Method Quantification Limit due to Incomplete Ion Cluster or Inaccurate Ion Ratio (Outside +/- 15% Tolerances)
IF Low Concentrations of PCBs #81 and #77 that were adjusted for interference from partially co-eluting PCBs #87 and #110, respectively.

Table 5. Percent Recoveries of ^{13}C-Non-*o* -Chloro-Substituted PCBs in Dosing Solutions of Large-Scale Extraction of Fish from the Housatonic R.

Revised: 11/20/2001
N46pcb-dosingsolns.xls

GC/MS Set: N46PCB
Dates: Jan. 12-14, 2000

NFCR Number:	Submitter Number:	Sample Description:	GC/MS Run No.	^{13}C-Non-*o* -Polychlorinated Biphenyls (% Recovery) Tetra: 3,4,4',5-TCB (^{13}C-PCB #81)	Tetra: 3,3',4,4'-TCB (^{13}C-PCB #77)	Penta: 3,3',4,4',5-PeCB (^{13}C-PCB #126)	Hexa: 3,3',4,4',5,5'-HxCB (^{13}C-PCB #169)
	3MP-LMB	3 Mile Pond, 3MP-LMB, 152 g (Dilution Factor 100)	46-7	71	74	74	78
	3MP-A	3 Mile Pond, 3MP-A, 152 g (Dilution Factor 100)	46-9	64	65	70	71
	3MP-B	3 Mile Pond, 3MP-B, 152 g (Dilution Factor 100)	46-10	66	71	71	74
	3MP-C	3 Mile Pond, 3MP-C, 152 g (Dilution Factor 100)	46-11	71	76	79	82
	RP-LMB	Rising Pond, RP-LMB, 0.41 g (Dil. Factor 31,250)	46-12	75	79	83	88
	RP-A	Rising Pond, RP-A, 0.41 g (Dil. Factor 31,250)	46-14	68	70	74	80
	RP-B	Rising Pond, RP-B, 0.41 g (Dil. Factor 31,250)	46-15	75	78	82	78
	RP-C	Rising Pond, RP-30-9, 0.41 g (Dil. Factor 31,250)	46-16	67	81	85	102
	WP-LMB	Woods Pond, WP-LMB, 0.102 g (Dil. F. 138,889)	46-17	57	61	89	116
	WP-A	Woods Pond, WP-A, 0.102 g (Dil. Factor 138,889)	46-19	61	74	85	85
	WP-B	Woods Pond, WP-B, 0.102 g (Dil. Factor 138,889)	46-20	85	76	76	89
	WP-C	Woods Pond, WP-C, 0.102 g (Dil. Factor 138,889)	46-21	78	77	78	96
	DRP-LMB	Deep Reach Pond, DRP-LMB, 0.036g (D.F. 357,143)	46-22	77	70	76	95
	DRP-A	Deep Reach Pond, DRP-A, 0.036 g (Dil. F. 357,143)	46-24	69	76	76	87
	DRP-B	Deep Reach Pond, DRP-B, 0.036 g (Dil. F. 357,143)	46-25	47	54	88	122
	DRP-C	Deep Reach Pond, DRP-C, 0.036 g (Dil. F. 357,143)	46-26	70	75	72	83
		Quality Control Samples:					
Proc. Blk #1, 11/30/2000		Procedure Blank, 11/30/2000	46-4	70	70	82	88
Proc. Spike 11/30/2000		Procedure Spike, 11/30/2000 (Spiked with 2 µg Aroclors total	46-5	63	65	65	75

Table 6. 2,3,7,8-Substituted Polychlorinated Dibenzo-*p*-dioxin and Dibenzofuran Concentrations (pg/g) in Dosing Solutions for Fish-egg Injections

File: DF39-housatonic-dosingsolns-rev.xls
Date Revised: Nov. 19, 2001
Date Analyzed: August 20-22, & September 14-15, 2001

		Toxicity Equiv. Factors: (Birds):	(Fish):	Humans/ Mammals:	3 Mile Pond Dosing Solutions					
Sample Site/Matrix:										
CERC Number:					3MP-1	3MP-2	3MP-3	**Mean**	**% CV**	**Geometric**
GC/HRMS Sets: DF39- or DF41- Injection No.					41-6	41-7	41-8		**(Coefficient of Variation)**	**Mean**
Sample Submitter No.										
Sample Mass Extracted (grams):					5.0	5.0	5.0			
DIOXINS	2,3,7,8-Tetrachloro	1.0	1.0	1.0	**0.5**	**0.4 LQ**	**0.4 LQ**	**0.4**	**15**	**0.4**
	1,2,3,7,8-Pentachloro	1.0	1.0	1.0	**0.4**	**0.4 LQ**	**0.4**	**0.4**	**5**	**0.4**
	1,2,3,4,7,8-Hexachloro	0.05	0.5	0.1	**0.2 LQ**	**0.1 LQ**	**0.2**	**0.1**	**31**	**0.1**
	1,2,3,6,7,8-Hexachloro	0.01	0.01	0.1	**0.3 LQ**	**0.3 LQ**	**0.4 LQ**	**0.3**	**17**	**0.3**
	1,2,3,7,8,9-Hexachloro	0.1	0.01	0.1	**0.2 LQ**	**0.2**	**0.2 ND**	**0.2**	**15**	**0.2**
	1,2,3,4,6,7,8-Heptachloro	0.001	0.001	0.01	**0.8**	**0.9 LQ**	**0.8 LQ**	**0.8**	**10**	**0.8**
	Octachloro	0.0001	0.0001	0.0001	**6.0**	**6.0 LQ**	**5.3**	**5.8**	**7**	**5.8**
FURANS	2,3,7,8-Tetrachloro	1.0	0.1	0.1	**2.4**	**2.5**	**2.4**	**2.4**	**0.3**	**2.4**
	1,2,3,7,8-Pentachloro	0.1	0.05	0.05	**0.2 LQ**	**0.1 LQ**	**0.1 LQ**	**0.2**	**27**	**0.2**
	2,3,4,7,8-Pentachloro	1.0	0.5	0.5	**0.4 LQ**	**0.3 LQ**	**0.3**	**0.3**	**13**	**0.3**
	1,2,3,4,7,8-Hexachloro	0.1	0.1	0.1	**0.1 LQ**	**0.1 LQ**	**0.0 LQ**	**0.1**	**34**	**0.1**
	1,2,3,6,7,8-Hexachloro	0.1	0.1	0.1	**0.1 LQ**	**0.1 LQ**	**0.0 LQ**	**0.1**	**31**	**0.1**
	1,2,3,7,8,9-Hexachloro	0.1	0.1	0.1	**0.1**	**0.2 LQ**	**0.1 LQ**	**0.1**	**47**	**0.1**
	2,3,4,6,7,8-Hexachloro	0.1	0.1	0.1	**0.1 LQ**	**0.1 LQ**	**0.1 ND**	**0.1**		**0.1**
	1,2,3,4,6,7,8-Heptachloro	0.01	0.01	0.01	**0.2 LQ**	**0.2 LQ**	**0.2 LQ**	**0.2**	**2**	**0.2**
	1,2,3,4,7,8,9-Heptachloro	0.01	0.01	0.01	**0.2 ND**	**0.2 ND**	**0.2 ND**	**0.2**	**ND**	**0.2**
	Octachloro	0.0001	0.0001	0.0001	**0.2**	**0.2 ND**	**0.2 LQ**	**0.2**	**13**	**0.2**
	Total Dioxin-Toxicity Equivalents (Fish):				**1.3**	**1.1**	**1.1**	**1.2**	**9**	**1.2**

* Dioxin Toxicity Equivalent Factors (Birds and Fish) from:
Van den Berg et.al, Environ. H. Persp, 106 (12), 775-791 (1998).

TEQs = Σ (Conc * TEF) except 0.5 Conc * TEF for ND

LQ Less than Method Quantification Limit due to Incomplete Ion Cluster or Ion Ratio Outside of +/- 15% Tolerances
ND Not Detected at Specified Detection Limit

Table 6. 2,3,7,8-Substituted Polychlorinated Dibenzo-*p*-dioxin and Dibenzofuran Concentrations (pg/g) in Dosing Solutions for Fish-egg Injections

File: DF39-housatonic-dosingsolns-rev.xls
Date Revised: Nov. 19, 2001
Date Analyzed: August 20-22, & September 14-15, 2001

Sample Site/Matrix:					Rising Pond Dosing Solutions					
CERC Number:					RP-1	RP-2	RP-3	**Mean**	**% CV**	**Geometric**
GC/HRMS Sets: DF39- or DF41- Injection No.					39-43	39-44	39-46			**Mean**
Sample Submitter No.										
Sample Mass Extracted (grams):					6.4	6.4	6.4			
		Toxicity Equiv. Factors: (Birds):	(Fish):	Humans/ Mammals:						
DIOXINS	2,3,7,8-Tetrachloro	1.0	1.0	1.0	**3.1**	**3.1**	**2.9**	**3.0**	**5**	**3.0**
	1,2,3,7,8-Pentachloro	1.0	1.0	1.0	**1.5**	**1.5**	**1.5**	**1.5**	**1**	**1.5**
	1,2,3,4,7,8-Hexachloro	0.05	0.5	0.1	**0.5 LQ**	**0.2**	**0.2**	**0.3**	**51**	**0.3**
	1,2,3,6,7,8-Hexachloro	0.01	0.01	0.1	**1.3 LQ**	**1.1**	**0.9**	**1.1**	**17**	**1.1**
	1,2,3,7,8,9-Hexachloro	0.1	0.01	0.1	**0.3 LQ**	**0.1 ND**	**0.1 ND**	**0.2**	**--**	**0.1**
	1,2,3,4,6,7,8-Heptachloro	0.001	0.001	0.01	**0.6 LQ**	**0.6 LQ**	**0.5**	**0.6**	**12**	**0.6**
	Octachloro	0.0001	0.0001	0.0001	**2.5**	**1.4**	**1.5**	**1.8**	**33**	**1.7**
FURANS	2,3,7,8-Tetrachloro	1.0	0.1	0.1	**9.1**	**9.3**	**9.3**	**9.2**	**1**	**9.2**
	1,2,3,7,8-Pentachloro	0.1	0.05	0.05	**14**	**14**	**14**	**14**	**0.3**	**14**
	2,3,4,7,8-Pentachloro	1.0	0.5	0.5	**35**	**36**	**36**	**36**	**2**	**36**
	1,2,3,4,7,8-Hexachloro	0.1	0.1	0.1	**1.8**	**1.8**	**1.8**	**1.8**	**1**	**1.8**
	1,2,3,6,7,8-Hexachloro	0.1	0.1	0.1	**2.1**	**1.7**	**1.8**	**1.9**	**10**	**1.9**
	1,2,3,7,8,9-Hexachloro	0.1	0.1	0.1	**0.5**	**0.1 LQ**	**0.1 LQ**	**0.2**	**103**	**0.2**
	2,3,4,6,7,8-Hexachloro	0.1	0.1	0.1	**3.2**	**3.1**	**2.9**	**3.1**	**4**	**3.1**
	1,2,3,4,6,7,8-Heptachloro	0.01	0.01	0.01	**2.0**	**1.7**	**1.8**	**1.8**	**8**	**1.8**
	1,2,3,4,7,8,9-Heptachloro	0.01	0.01	0.01	**0.9**	**0.6**	**0.8 LQ**	**0.8**	**18**	**0.8**
	Octachloro	0.0001	0.0001	0.0001	**1.9**	**0.8**	**1.1 LQ**	**1.3**	**43**	**1.2**
	Total Dioxin-Toxicity Equivalents (Fish):				**24**	**25**	**25**	**25**	**0.3**	**25**

* Dioxin Toxicity Equivalent Factors (Birds and Fish) from:
Van den Berg et.al, Environ. H. Persp, 106 (12), 775-791 (1998).

TEQs = Σ (Conc * TEF) except 0.5 Conc * TEF for ND

LQ Less than Method Quantification Limit due to Incomplete Ion Cluster or Ion Ratio Outside of +/- 15% Tolerance
ND Not Detected at Specified Detection Limit

Table 6. 2,3,7,8-Substituted Polychlorinated Dibenzo-*p*-dioxin and Dibenzofuran Concentrations (pg/g) in Dosing Solutions for Fish-egg Injections

File: DF39-housatonic-dosingsolns-rev.xls
Date Revised: Nov. 19, 2001
Date Analyzed: August 20-22, & September 14-15, 2001

					Woods Pond Dosing Solutions					
Sample Site/Matrix:										
CERC Number:					WP-1	WP-2	WP-3**	**Mean**	**% CV**	**Geometric**
GC/HRMS Sets: DF39- or DF41- Injection No.					39-47	39-48	39-49			**Mean**
Sample Submitter No.										
Sample Mass Extracted (grams):					7.1	7.1	7.1			
		Toxicity Equiv. Factors: (Birds):	(Fish):	Humans/ Mammals:						
DIOXINS	2,3,7,8-Tetrachloro	1.0	1.0	1.0	**2.1 LQ**	**1.9**	**NA**	**2.0**	5	**2.0**
	1,2,3,7,8-Pentachloro	1.0	1.0	1.0	**0.6**	**1.0 LQ**	**NA**	**0.8**	37	**0.8**
	1,2,3,4,7,8-Hexachloro	0.05	0.5	0.1	**0.2 LQ**	**0.2 LQ**	**NA**	**0.2**	0	**0.2**
	1,2,3,6,7,8-Hexachloro	0.01	0.01	0.1	**0.4**	**0.3**	**NA**	**0.4**	25	**0.4**
	1,2,3,7,8,9-Hexachloro	0.1	0.01	0.1	**0.1 LQ**	**0.1 ND**	**NA**	**0.1**	21	**0.1**
	1,2,3,4,6,7,8-Heptachloro	0.001	0.001	0.01	**0.3**	**0.4**	**NA**	**0.3**	29	**0.3**
	Octachloro	0.0001	0.0001	0.0001	**0.7**	**0.9 LQ**	**NA**	**0.8**	18	**0.8**
FURANS	2,3,7,8-Tetrachloro	1.0	0.1	0.1	**8.2**	**8.4**	**NA**	**8.3**	2	**8.3**
	1,2,3,7,8-Pentachloro	0.1	0.05	0.05	**12**	**12**	**NA**	**12**	1	**12**
	2,3,4,7,8-Pentachloro	1.0	0.5	0.5	**16**	**16**	**NA**	**16**	2	**16**
	1,2,3,4,7,8-Hexachloro	0.1	0.1	0.1	**1.4 LQ**	**1.5**	**NA**	**1.5**	4	**1.5**
	1,2,3,6,7,8-Hexachloro	0.1	0.1	0.1	**1.2**	**1.2 LQ**	**NA**	**1.2**	0	**1.2**
	1,2,3,7,8,9-Hexachloro	0.1	0.1	0.1	**0.1**	**0.1 LQ**	**NA**	**0.1**	10	**0.1**
	2,3,4,6,7,8-Hexachloro	0.1	0.1	0.1	**2.0 LQ**	**2.6 LQ**	**NA**	**2.3**	17	**2.3**
	1,2,3,4,6,7,8-Heptachloro	0.01	0.01	0.01	**1.1 LQ**	**1.2**	**NA**	**1.2**	6	**1.2**
	1,2,3,4,7,8,9-Heptachloro	0.01	0.01	0.01	**0.4 LQ**	**0.4 LQ**	**NA**	**0.4**	7	**0.4**
	Octachloro	0.0001	0.0001	0.0001	**0.2 LQ**	**0.2 LQ**	**NA**	**0.2**	13	**0.2**
	Total Dioxin-Toxicity Equivalents (Fish):				**12**	**13**	**Data Not Available**	**12**	4	**12**

* Dioxin Toxicity Equivalent Factors (Birds and Fish) from:
Van den Berg et.al, Environ. H. Persp,
106 (12), 775-791 (1998).

TEQs = Σ (Conc * TEF) except 0.5 Conc * TEF for ND

** Sample Processing Error -- No Recovery of ^{13}C-surrogates

LQ Less than Method Quantification Limit due to Incomplete Ion Cluster or Ion Ratio Outside of +/- 15% Tolerances

ND Not Detected at Specified Detection Limit

NA Data Not Available

Table 6. 2,3,7,8-Substituted Polychlorinated Dibenzo-*p*-dioxin and Dibenzofuran Concentrations (pg/g) in Dosing Solutions for Fish-egg Injections

File: DF39-housatonic-dosingsolns-rev.xls
Date Revised: Nov. 19, 2001
Date Analyzed: August 20-22, & September 14-15, 2001

		Toxicity Equiv. Factors: (Birds):	(Fish):	Humans/ Mammals:	Deep Reach Pond Dosing Solutions DRP-1	DRP-2	DRP-3	**Mean**	**% CV**	**Geometric Mean**
Sample Site/Matrix:										
CERC Number:										
GC/HRMS Sets: DF39- or DF41- Injection No.					39-51	39-52	39-53			
Sample Submitter No.										
Sample Mass Extracted (grams):					6.5	6.5	6.5			
DIOXINS	2,3,7,8-Tetrachloro	1.0	1.0	1.0	**1.6**	**1.7**	**1.7**	**1.7**	**4**	**1.7**
	1,2,3,7,8-Pentachloro	1.0	1.0	1.0	**1.0**	**1.1**	**1.2 LQ**	**1.1**	**7**	**1.1**
	1,2,3,4,7,8-Hexachloro	0.05	0.5	0.1	**0.2 LQ**	**0.1 LQ**	**0.1**	**0.1**	**20**	**0.1**
	1,2,3,6,7,8-Hexachloro	0.01	0.01	0.1	**0.5 LQ**	**0.5**	**0.5 LQ**	**0.5**	**4**	**0.5**
	1,2,3,7,8,9-Hexachloro	0.1	0.01	0.1	**0.1**	**0.1 ND**	**0.1 LQ**	**0.1**	**13**	**0.1**
	1,2,3,4,6,7,8-Heptachloro	0.001	0.001	0.01	**0.4 LQ**	**0.4**	**0.4 LQ**	**0.4**	**5**	**0.4**
	Octachloro	0.0001	0.0001	0.0001	**1.9**	**3.9**	**1.4**	**2.4**	**56**	**2.2**
FURANS	2,3,7,8-Tetrachloro	1.0	0.1	0.1	**11**	**11**	**11**	**11**	**4**	**11**
	1,2,3,7,8-Pentachloro	0.1	0.05	0.05	**13**	**2.5 LQ**	**13**	**9.8**	**65**	**7.7**
	2,3,4,7,8-Pentachloro	1.0	0.5	0.5	**19**	**18**	**19**	**19**	**4**	**19**
	1,2,3,4,7,8-Hexachloro	0.1	0.1	0.1	**2.0**	**1.8**	**2.1**	**2.0**	**9**	**2.0**
	1,2,3,6,7,8-Hexachloro	0.1	0.1	0.1	**1.5 LQ**	**1.4**	**1.6**	**1.5**	**5**	**1.5**
	1,2,3,7,8,9-Hexachloro	0.1	0.1	0.1	**0.1**	**0.1 ND**	**0.1 LQ**	**0.1**	**10**	**0.1**
	2,3,4,6,7,8-Hexachloro	0.1	0.1	0.1	**3.8**	**2.6**	**3.6 LQ**	**3.3**	**19**	**3.3**
	1,2,3,4,6,7,8-Heptachloro	0.01	0.01	0.01	**2.1 LQ**	**1.7**	**1.9**	**1.9**	**11**	**1.9**
	1,2,3,4,7,8,9-Heptachloro	0.01	0.01	0.01	**0.5**	**0.2 LQ**	**0.6 LQ**	**0.5**	**47**	
	Octachloro	0.0001	0.0001	0.0001	**0.4 LQ**	**0.2 LQ**	**0.4 LQ**	**0.3**	**28**	**0.3**
	Total Dioxin-Toxicity Equivalents (Fish):				**14**	**13**	**15**	**14**	**6**	**14**

* Dioxin Toxicity Equivalent Factors (Birds and Fish) from:
Van den Berg et.al, Environ. H. Persp, 106 (12), 775-791 (1998).

TEQs = Σ (Conc * TEF) except 0.5 Conc * TEF for ND

LQ Less than Method Quantification Limit due to Incomplete Ion Cluster or Ion Ratio Outside of +/- 15% Tolerance
ND Not Detected at Specified Detection Limit

Table 6. 2,3,7,8-Substituted Polychlorinated Dibenzo-*p*-dioxin and Dibenzofuran Concentrations (pg/g) in Dosing Solutions for Fish-egg Injections

File: DF39-housatonic-dosingsolns-rev.xls
Date Revised: Nov. 19, 2001
Date Analyzed: August 20-22, & September 14-15, 2001
Sample Site/Matrix:

CERC Number:
GC/HRMS Sets: DF39- or DF41- Injection No.

Sample Submitter No.

Sample Mass Extracted (grams):

		Toxicity Equiv. Factors: (Birds):	(Fish):	Humans/ Mammals:	Quality Assurance Samples: Procedure Blank 39-41 3/15/2001 **Conc. (pg/g-eq) based on sample wgts 7 g**	Procedure Blank 41-5 8/30/2001 **Conc. (pg/g-eq) based on sample wgts 7 g**
DIOXINS	2,3,7,8-Tetrachloro	1.0	1.0	1.0	**0.1 LQ**	**0.1 ND**
	1,2,3,7,8-Pentachloro	1.0	1.0	1.0	**0.1 ND**	**0.2 ND**
	1,2,3,4,7,8-Hexachloro	0.05	0.5	0.1	**0.1 ND**	**0.2 ND**
	1,2,3,6,7,8-Hexachloro	0.01	0.01	0.1	**0.1 ND**	**0.2 ND**
	1,2,3,7,8,9-Hexachloro	0.1	0.01	0.1	**0.1 ND**	**0.2 ND**
	1,2,3,4,6,7,8-Heptachloro	0.001	0.001	0.01	**0.1 LQ**	**0.2 LQ**
	Octachloro	0.0001	0.0001	0.0001	**0.2 LQ**	**0.3 LQ**
FURANS	2,3,7,8-Tetrachloro	1.0	0.1	0.1	**0.2 LQ**	**0.3**
	1,2,3,7,8-Pentachloro	0.1	0.05	0.05	**0.1**	**0.1 ND**
	2,3,4,7,8-Pentachloro	1.0	0.5	0.5	**0.1 LQ**	**0.1 LQ**
	1,2,3,4,7,8-Hexachloro	0.1	0.1	0.1	**0.1 ND**	**0.1 ND**
	1,2,3,6,7,8-Hexachloro	0.1	0.1	0.1	**0.1 ND**	**0.1 ND**
	1,2,3,7,8,9-Hexachloro	0.1	0.1	0.1	**0.1 LQ**	**0.2 ND**
	2,3,4,6,7,8-Hexachloro	0.1	0.1	0.1	**0.1 ND**	**0.2 ND**
	1,2,3,4,6,7,8-Heptachloro	0.01	0.01	0.01	**0.1 ND**	**0.2 LQ**
	1,2,3,4,7,8,9-Heptachloro	0.01	0.01	0.01	**0.1 ND**	**0.2 ND**
	Octachloro	0.0001	0.0001	0.0001	**0.2 ND**	**0.2 ND**
	Total Dioxin-Toxicity Equivalents (Fish):				**0.2**	**0.3**

* Dioxin Toxicity Equivalent Factors (Birds and Fish) from:
Van den Berg et.al, Environ. H. Persp, 106 (12), 775-791 (1998).

TEQs = Σ (Conc * TEF) except 0.5 Conc * TEF for ND

LQ Less than Method Quantification Limit due to Incomplete Ion Cluster or Ion Ratio Outside of +/- 15% Tolerances
ND Not Detected at Specified Detection Limit

Table 7. Percent Recovery of ^{13}C-Substituted Polychlorinated Dibenzo-*p*-dioxins and Dibenzofurans in Dosing Solutions for Fish-egg Injections

File: DF39-housatonic-dosingsolns.xls
Date Revised: Nov. 19, 2001
Date Analyzed: August 20-22, & September 14-15, 2001

Sample Site/Matrix:	3 Mile Pond Dosing Solutions			Rising Pond Dosing Solutions		
CERC Number:	3MP-1	3MP-2	3MP-3	RP-1	RP-2	RP-3
GC/HRMS Sets: DF39- or DF41- Inj. No.	41-6	41-7	41-8	39-43	39-44	39-46
Sample Submitter No.						
Sample Mass Extracted (grams):	5.0	5.0	5.0	6.4	6.4	6.4
				(% Recovery)		
DIOXINS 2,3,7,8-Tetrachloro	62	66	67	71	71	73
1,2,3,7,8-Pentachloro	63	65	64	72	69	70
1,2,3,4,7,8-Hexachloro	64	64	68	71	73	69
1,2,3,6,7,8-Hexachloro	61	61	60	67	66	60
1,2,3,7,8,9-Hexachloro	56	58	58	65	65	61
1,2,3,4,6,7,8-Heptachloro	65	66	66	71	70	66
Octachloro	52	53	54	58	56	53
FURANS 2,3,7,8-Tetrachloro	63	65	63	78	78	77
1,2,3,7,8-Pentachloro	60	62	61	70	71	70
2,3,4,7,8-Pentachloro	62	60	59	70	73	71
1,2,3,4,7,8-Hexachloro	61	62	62	68	67	66
1,2,3,6,7,8-Hexachloro	50	52	51	56	57	55
1,2,3,7,8,9-Hexachloro	48	47	47	53	52	51
1,2,3,4,6,7,8-Heptachloro	47	46	48	50	50	48
1,2,3,4,7,8,9-Heptachloro	57	58	55	60	60	56

Table 7. Percent Recovery of ^{13}C-Substituted Polychlorinated Dibenzo-*p*-dioxins and Dibenzofurans in Dosing Solutions for Fish-egg Injections

File: DF39-housatonic-dosingsolns.xls
Date Revised: Nov. 19, 2001
Date Analyzed: August 20-22, & September 14-15, 2001

Sample Site/Matrix:	Woods Pond Dosing Solutions			Deep Reach Pond Dosing Solutions		
CERC Number:	WP-1	WP-2	WP-3**	DRP-1	DRP-2	DRP-3
GC/HRMS Sets: DF39- or DF41- Inj. No.	39-47	39-48	39-49	39-51	39-52	39-53
Sample Submitter No.						
Sample Mass Extracted (grams):	7.1	7.1	7.1	6.5	6.5	6.5
				(% Recovery)		
DIOXINS 2,3,7,8-Tetrachloro	79	79	-- NA	45	58	70
1,2,3,7,8-Pentachloro	95	80	-- NA	44	69	74
1,2,3,4,7,8-Hexachloro	79	86	-- NA	43	79	74
1,2,3,6,7,8-Hexachloro	76	77	-- NA	40	72	66
1,2,3,7,8,9-Hexachloro	74	76	-- NA	38	70	64
1,2,3,4,6,7,8-Heptachloro	81	83	-- NA	42	72	73
Octachloro	65	72	-- NA	32	58	57
FURANS 2,3,7,8-Tetrachloro	87	86	-- NA	48	70	74
1,2,3,7,8-Pentachloro	79	85	-- NA	45	32	73
2,3,4,7,8-Pentachloro	80	86	-- NA	45	55	73
1,2,3,4,7,8-Hexachloro	79	84	-- NA	43	62	70
1,2,3,6,7,8-Hexachloro	67	72	-- NA	36	34	63
1,2,3,7,8,9-Hexachloro	62	63	-- NA	33	53	56
1,2,3,4,6,7,8-Heptachloro	62	65	-- NA	33	49	55
1,2,3,4,7,8,9-Heptachloro	71	77	-- NA	38	30	64

** Sample Processing Error -- No Recovery of ^{13}C-surrogates

NA Data Not Available

Table 7. Percent Recovery of ^{13}C-Substituted Polychlorinated Dibenzo-*p*-dioxins and Dibenzofurans in Dosing Solutions for Fish-egg Injections

File: DF39-housatonic-dosingsolns.xls
Date Revised: Nov. 19, 2001
Date Analyzed: August 20-22, & September 14-15, 2001

Sample Site/Matrix:	Quality Assurance Samples	
CERC Number:	Procedure Blank	Procedure Blank
GC/HRMS Sets: DF39- or DF41- Inj. No.	39-41	41-5
	3/15/2001	8/30/2001
Sample Submitter No.		
Sample Mass Extracted (grams):		
		(% Recovery)
DIOXINS 2,3,7,8-Tetrachloro	71	68
1,2,3,7,8-Pentachloro	74	69
1,2,3,4,7,8-Hexachloro	74	69
1,2,3,6,7,8-Hexachloro	69	66
1,2,3,7,8,9-Hexachloro	66	62
1,2,3,4,6,7,8-Heptachloro	78	71
Octachloro	57	55
FURANS 2,3,7,8-Tetrachloro	83	65
1,2,3,7,8-Pentachloro	77	64
2,3,4,7,8-Pentachloro	80	63
1,2,3,4,7,8-Hexachloro	73	66
1,2,3,6,7,8-Hexachloro	61	58
1,2,3,7,8,9-Hexachloro	57	52
1,2,3,4,6,7,8-Heptachloro	54	53
1,2,3,4,7,8,9-Heptachloro	66	63

Table 8. Calculated TEQs for Housatonic Dosing Solutions (pg/g)

Sample ID	Total mono-o-PCB TEQs#	% mPCB	Total non-o-PCB TEQs#	% nPCB	Total PCDD TEQs#	% PCDD	Total PCDF TEQs#	% PCDF	Total Dosing Solution TEQs#
RP LMB	2.8	*36*	7.6	*100*	*	---	*	---	7.6
RP-A DOSE	2.6	*8*	7.8	*24*	5.3	*16*	20	*60*	32.7
RP-B DOSE	2.7	*8*	7.8	*24*	5.2	*16*	20	*61*	32.8
RP-C DOSE	2.6	*8*	7.3	*23*	4.9	*15*	20	*62*	32.2
WP LMB	5.1	*39*	13	*100*	*	---	*	---	13.2
WP-A DOSE	4.9	*20*	12	*49*	2.9	*12*	9.4	*39*	24.0
WP-B DOSE	4.9	*20*	12	*49*	3.1	*12*	9.8	*39*	25.2
WP-C DOSE	4.8	*39*	12	*100*	*	---	*	---	12.2
DRP LMB	8.3	*50*	17	*100*	*	---	*	---	16.7
DRP-A DOSE	7.9	*27*	15	*51*	2.9	*10*	11	*39*	29.3
DRP-B DOSE	7.6	*29*	13	*49*	3.1	*12*	10	*39*	26.1
DRP-C DOSE	7.9	*32*	10	*41*	3.2	*13*	12	*47*	25.1
3MP LMB	0.004	*27*	0.014	*100*	*	---	*	---	0.0
3MP-A DOSE	0.004	*0.3*	0.016	*1*	1.0	*73*	0.4	*26*	1.4
3MP-B DOSE	0.004	*0.3*	0.016	*1*	0.9	*71*	0.3	*28*	1.2
3MP-C DOSE	0.004	*0.3*	0.016	*1*	0.9	*74*	0.3	*25*	1.2

**na- no measured value*

#Using Fish TEFs from ref (19)