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PCB concentrations of lake whitefish (*Coregonus clupeaformis*) vary by sex

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ABSTRACT

We determined whole-fish polychlorinated biphenyl (PCB) concentrations in 26 female lake whitefish (*Coregonus clupeaformis*) and 34 male lake whitefish from northern Lake Huron. In 5 of the 26 female lake whitefish, we also determined PCB concentrations in the somatic tissue and ovaries. In addition, bioenergetics modeling was used to determine the contribution of the growth dilution effect to the observed difference in PCB concentrations between the sexes. Whole-fish PCB concentrations for females and males averaged 60 ng/g and 80 ng/g, respectively; thus males were 34% higher in PCB concentration compared with females. Based on the PCB determinations in the somatic tissue and ovaries, we predicted that PCB concentration of females would increase by 2.5%, on average, immediately after spawning due to release of eggs. Thus, the change in PCB concentration due to release of eggs did not explain, to any degree, the higher PCB concentrations observed in males compared with females. Bioenergetics modeling results indicated that the growth dilution effect could account for males being only 0.7% higher in PCB concentration compared with females. Thus, the growth dilution effect contributed very little to the observed difference in PCB concentrations between the sexes. We conclude that males were higher than females in PCB concentration most likely due to a higher rate of energy expenditure, stemming from greater activity and a greater resting metabolic rate. A higher rate of energy expenditure leads to a higher rate of food consumption, which, in turn, leads to a higher PCB accumulation rate.

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Introduction

Recent research has revealed an apparent pattern in the relative difference between whole-fish polychlorinated biphenyl (PCB) concentrations of mature males and whole-fish PCB concentrations of mature females (Madenjian, 2011; Madenjian et al., 2014). Specifically, mature males have been found to be 15–45% higher in PCB concentration than similarly aged mature females, and this difference has been primarily attributed to a higher rate of energy expenditure in mature males due to greater activity and a higher resting metabolic rate (or standard metabolic rate [SMR]). To date, higher whole-fish PCB concentrations in males compared with females have been documented in walleye (*Sander vitreus*), lake trout (*Salvelinus namaycush*), coho salmon (*Oncorhynchus kisutch*), burbot (*Lota lota*), sea lamprey (*Petromyzon marinus*), and cisco (*Coregonus artedi*). A higher rate of energy expenditure results in a higher rate of food consumption, which, in turn, results in a higher PCB accumulation rate. A faster growth rate by females compared with males can also contribute toward males exhibiting higher PCB concentrations than females, however this growth dilution effect has been estimated to be relatively unimportant in most cases (Madenjian, 2011; Madenjian et al., 2014). Typically, bioenergetics modeling has been used to assess the contribution of the growth dilution effect to the observed difference in PCB concentrations between the sexes.

To be certain that observed differences in PCB concentrations between the sexes are true indicators of the above-mentioned behavioral and physiological differences between the sexes, whole-fish PCB determinations are required (Madenjian, 2011). The difference in muscle tissue PCB concentrations between the sexes or in liver tissue PCB concentrations between the sexes may or may not accurately reflect the difference in whole-fish PCB concentrations between the sexes. Unfortunately, most of the studies addressing differences in PCB concentrations between the sexes of fish have been based on muscle tissue or liver tissue PCB determinations. Consequently, results from these studies based on parts of fish rather than whole fish may not, in some cases, provide a true indication of the differences in behavior and physiology between the sexes of fish.

In general, results from muscle tissue and liver tissue determinations supported the contention that male fish exceeded female fish in PCB concentrations. For example, Bodiguel et al. (2009) reported significantly higher PCB concentrations in both the muscle tissue and liver tissue of adult male European hake (*Merluccius merluccius*) compared with adult female European hake from the Mediterranean Ocean. Similarly, Kammann et al. (1993) observed substantially greater PCB concentrations in the livers of male dab (*Limanda limanda*), a species of flatfish,

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compared with female dab from the North Sea. Likewise, Rypel et al. (2007) documented significantly greater muscle PCB concentrations in males compared with females in both largemouth bass (*Micropterus salmoides*) and spotted bass (*Micropterus punctulatus*) from a southeastern USA reservoir. Moreover, Gewurtz et al. (2011) showed that muscle PCB concentrations were significantly greater in males than in females for most of the walleye populations surveyed in Canada. However, in a number of cases, significant differences in muscle tissue or liver tissue PCB concentrations between the sexes were not detectable. Further, in a few cases, muscle tissue PCB concentration was found to be significantly greater in females than in males. For example, Marthinsen et al. (1991) did not detect significant differences in liver tissue PCB concentrations between the sexes of Atlantic cod (*Gadus morhua*) or European flounder (*Platichthys flesus*) caught in the Glomma River, a tributary to the North Sea. Similarly, significant differences in muscle PCB concentrations were rarely detected for six species of freshwater fishes (other than walleye) in Canada (Gewurtz et al., 2011). In addition, muscle PCB concentrations of channel catfish (*Ictalurus punctatus*) from a southeastern USA reservoir were found to be significantly greater in females than in males (Rypel et al., 2007). These cases of failing to detect a significant difference in PCB concentrations between the sexes, or finding significantly higher PCB concentrations in females compared with males, may have simply been due, at least in part, to determining PCB concentrations in muscle tissue or liver tissue rather than in whole fish.

Although lake whitefish (*Coregonus clupeaformis*) is the most valuable commercial fish species in the Upper Great Lakes (Ebener et al., 2008; Brenden et al., 2013), to the best of our knowledge, differences in whole-fish PCB concentrations between the sexes of lake whitefish have not been examined. Lake whitefish is not considered a top predator in Great Lakes food webs (Ebener et al., 2008). Rather, lake whitefish is a generalist feeder, preying predominately on invertebrates but capable of some degree of piscivory (Pothoven and Madenjian, 2013). Thus, the trophic position of lake whitefish is intermediary between the top predators, such as walleye, lake trout, coho salmon, burbot, and sea lamprey, and the planktivorous fishes, such as cisco. Documentation of males exceeding females in whole-fish PCB concentrations of lake whitefish would mark another important step in establishing the pervasiveness of this apparently general characteristic of higher energy expenditure rate in males.

The overall goal of our study was to evaluate the difference in whole-fish PCB concentrations between the sexes of the Detour spawning stock of lake whitefish from northern Lake Huron. If a difference was established, then we utilized modeling to identify the most likely explanation for the observed difference. Specific objectives of our study included: (1) quantify the difference in whole-fish PCB concentrations between the sexes of lake whitefish captured from a spawning aggregation of the Detour stock in northern Lake Huron, (2) use bioenergetics modeling to quantify the growth dilution effect on the difference in whole-fish PCB concentrations between the sexes, (3) compare somatic tissue PCB concentration with ovary PCB concentration in female lake whitefish, and (4) estimate the change in whole-fish PCB concentration of female lake whitefish associated with the release of eggs at spawning. In addition, we were especially interested in whether our findings for lake whitefish supported the emerging pattern of higher whole-fish PCB concentrations in males compared with females across various species of fish.

Methods

Field methods

Adult lake whitefish were captured using a commercial trap net fished in northern Lake Huron in the vicinity of Cedarville, MI, which is located at the center of the spawning area used by the Detour stock of lake whitefish (Ebener et al., 2010). The trap net was set in waters between 5 and 15 m deep. On 6 November 2010, the trap net catch was retrieved, and 60 lake whitefish were randomly selected from the catch. Typically, spawning by lake whitefish in northern Lake Huron peaks in mid-November (Ebener et al., 2008). Total length of each lake whitefish was measured to the nearest mm, and each lake whitefish was weighed to the nearest 10 g. Sex and maturity of each lake whitefish were determined by visual inspection of the gonads. Each lake whitefish was individually bagged with a cardboard tag marked with a unique identification number and then frozen at −20 °C. During January 2011, the frozen lake whitefish were transported to the Great Lakes Science Center (GLSC) in Ann Arbor, MI for further processing.

PCB determinations

At the GLSC, 5 female lake whitefish were randomly selected for PCB determinations of both somatic tissue and ovaries. For each of these 5 females, the lake whitefish was partially thawed, and otoliths were then removed for aging purposes. Ovaries were removed and weighed to the nearest 0.1 g. Likewise, the remaining somatic tissue was weighed to the nearest 0.1 g. Ovaries and somatic tissue were homogenized separately in appropriately sized blenders. About 100 g of the homogenate was placed in a contaminant-free glass jar, sealed with a lid, and then stored at −20 °C. For the 55 other lake whitefish, the fish were partially thawed, and otoliths were removed for aging purposes. Each whole fish was homogenized using appropriately sized blenders, and then approximately 100 g of the homogenate was placed in a contaminant-free glass jar, sealed with a lid, and stored at −20 °C. The frozen homogenates were shipped to the State Environmental Laboratory of the Oklahoma Department of Environmental Quality (DEQ) in Oklahoma City, OK for PCB determinations. The otoliths were shipped to the Chippewa Ottawa Resource Authority (CORA) fishery research facility in Sault Ste. Marie, MI for aging. Aging was accomplished via enumeration of annuli on the otoliths.

At the State Environmental Laboratory of the Oklahoma DEQ, total PCB concentration in each of the homogenates was determined by following Protocol 303 of the U. S. Food and Drug Administration (FDA) (1999). Approximately 50 g of the homogenate was extracted with 250 mL acetonitrile by blending in an industrial blender. The acetonitrile extract was filtered using a vacuum filter flask apparatus and partitioned with 150 mL petroleum ether (or hexane) in a separatory funnel. The petroleum ether (or hexane) extract was then passed through a Florisil® (Frederick, MD) cleanup column using 150 mL of 100% petroleum ether (or hexane), followed by 150 mL of 6% diethyl ether/petroleum ether (or hexane) and 150 mL of 15% diethyl ether/petroleum ether (or hexane) fractions. The three co-solvent fractions were combined and concentrated to a volume of 1 mL during a solvent exchange with hexane. The concentrate was brought to a final volume of 5 mL in hexane. Separation, identification, and quantification of total PCBs was accomplished by dual injection, dual capillary column gas chromatography (GC) with dual electron capture detectors (ECD) using an Agilent 6890 GC/MS (Agilent Industries, Palo Alto, CA). Two chromatographic columns with dissimilar stationary phases were used for analyte identification confirmation. The columns used for primary and confirmatory analyses were (5%-phenyl)-methylpolysiloxane with dimensions of 30 m × 0.53 mm ID × 0.5 μm phase thickness, and (35%-phenyl)-methylpolysiloxane with dimensions of 30 m × 0.53 mm ID × 0.5 μm. The mobile phase for each column was helium at 4.0 mL/min constant flow. Inlets were purged packed adapted for megabore capillary columns with isothermal temperatures of 250 °C each. Micro-electron capture detectors used 5% methane/argon mixture as the makeup gas at 60.0 mL/min flow with isothermal temperatures of 250 °C each. The initial oven temperature was 45 °C ramping at 8 °C/min to hold at 200 °C for 3 min; then ramping at 5 °C/min to hold at 280 °C for 7.5 min for simultaneous dual inlet of 2 μL injections. The data collection rate was 20 Hz. Pentachloronitrobenzene was used an internal standard. Matrix spikes were performed using Aroclors 1232 and 1260