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Factors influencing individual variability of PCB body burdens in fish populations

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By

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Factors influencing individual variability of PCB body burdens in fish populations

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DECLARATION OF CO-AUTHORSHIP AND PREVIOUS PUBLICATION

I. Co-Authorship Declaration

I hereby declare that this thesis incorporates material that is result of joint research under the supervision of Dr. Doug Haffner and Dr. Ken Drouillard. Chapter 2 contains material from an article titled "Characterizing variability of POPs bioaccumulation within forage fish communities of the Detroit River, Ontario, Canada" which has been submitted to *Environmental Toxicology and Chemistry*. This article was co-authored by McLeod, A., Paterson, G., Drouillard, K., and Haffner, G. The main ideas, experimental design, and data analysis were performed by the author. The contributions of co-authors were through guidance with field and laboratory work and revising manuscript drafts.

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II. Declaration of Previous Publication

This thesis includes one original paper that has been previously submitted for publication in peer reviewed journals, as follows:

Thesis Chapter	Publication title/full citation	Publication status*
Chapter 2	McLeod, A., Paterson, G., Drouillard, K., and Haffner, G. Characterizing variability in POPs bioaccumulation within forage fish communities of the Detroit River, Ontario, Canada. Submitted September 2013. *Environmental Toxicology and Chemistry*.	* submitted

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ABSTRACT

I investigated the contributions of hydrophobicity, species differences, and spatial and temporal variation to individual variability in PCB concentrations using three species of cyprinids, bluntnose minnows (*Pimephales notatus*), emerald shiners (*Notropis atherinoides*), and spottail shiners (*Notropis hudsonius*). I then investigated the influence of variation in chemical, physiological, and ecological characteristics on trophic magnification factors (TMFs), a food-web bioaccumulation metric commonly used by regulators. PCB concentrations are influenced most notably by hydrophobicity which explains 14% of the variability. When drivers are examined on a K_{OW}-specific basis physiological and ecological factors have differing importance, for instance species differences account for twice as much variation for PCBs with log KOW > 6.0. Finally, I used a food-web biomagnification model to investigate the sensitivity of TMFs to chemical and ecological perturbations demonstrating the importance of spatial and temporal variation in contaminant concentrations and the need to incorporate top predator foraging ranges into sampling strategies.

DEDICATION

To Doug, for the challenge.

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CHAPTER 1
GENERAL INTRODUCTION

1.1 Introduction

A basic tenet in ecotoxicology and population ecology has been that all individuals within a population display similar characteristics, and variations between individuals can be disregarded as statistical noise (Ringler, 1983). Statistical analyses are based on the assumption that the average individual is representative of the population. Recent research, however, shows that a range of terrestrial and aquatic species consistently exhibit behavioural differences among individuals (Gosling 2001; Sih *et al.* 2004). Research within these fields is now beginning to recognize the ecological ramifications that using average individuals to describe populations may have. For example, Tinker *et al*. (2009) noted individuals within a population display variability in most measurable behavioural traits including aggression (e.g. Riechert and Hedrick 1993), activity (e.g. Sih *et al.* 2003), exploration (Dingemanse *et al.* 2002), risk-taking (e.g. Fraser *et al.* 2001), fearfulness (Boissy 1995), and reactivity (Koolhaas *et al.* 1997). Moreover, this variability often has a non-random distribution suggesting that it is subject to selection with significant ecological and evolutionary consequences (e.g. Dall *et al*. 2004). This has important implications for evolution because a population that consists of multiple behavioural types will be better able to respond to change, e.g. bolder individuals may locate new resources and aggressive individuals may be better competitors as current resources become limited (Sih *et al.* 2004). Finally, a high degree of variability results in three things: a greater population stability when challenged by competition or predation (e.g. Lomnicki 1984), the exertion of multiple stressors on prey species (Sherratt and MacDougall 1995), and a greater ability to diversify (e.g. Doebeli and Dieckmann 2000).

While the causes of variability can have implications for the ecology of organisms, individual variability can have eco-toxicological implications as well. For instance, large amounts of variation in contaminant body burdens between organisms can have broader repercussions for hazard assessments and monitoring programs. Emergent chemicals are screened using a variety of bioaccumulation metrics including bioconcentration factors (BCFs, or the amount of chemical in an organism compared to that in the water), biomagnification factors (BMFs, or the increase in chemical concentration between trophic levels), and trophic magnification, or foodweb magnification, factors (TMFs or FWMFs, or the increase in chemical concentration across the whole foodweb) (Gobas et al. 2009; Weisbrod et al. 2009). The inherent variability in these metrics can cause persistent chemicals to be mis-categorized as biodiluting chemicals, or can cause biodiluting chemicals to be mis-characterized as biomagnifying. An example of this would be the chemical of concern, D5, a cyclic volatile methylsiloxane (or cVMS), where studies have reported its TMF as being >1, and less than 1, essentially biomagnifying, and biodiluting (Borgå et al. 2012). Furthermore, when the above parameters are measured empirically, i.e. via laboratory or field assessment, statistical considerations related to potential sampling strategies often come into play. However, such considerations tends to focus on interpretation of the mean exposure metric relative to government mandated guidelines and objectives. It is much less common to interpret variation in measured metrics in light of regulating factors contributing to the distributions observed across samples.

The inherent variation in natural systems can affect the applicability of models. Mathematical models have widespread use in ecology, including the prediction of predator-prey cycles, population dynamics, growth curves, and bioaccumulation. The accuracy of these models serves as both a metric of how well we understand a species or process, and a way of extrapolating our knowledge. If, however, individuals within a population vary extensively across any number of traits, it calls into question the reliability of these models. In today's world of high computing power, it becomes possible to incorporate individual variability into these models.

One model which appears to be suited for this purpose deals with the accumulation of persistent organic pollutants (POPs) in aquatic systems. Decades of research have gone into studying attributes of one class of POPs, polychlorinated biphenyls (PCBs), and several models have been created simulating their biomagnification in aquatic systems (e.g. Morrison *et al.* 1996; Nfon and Cousins 2007). Unfortunately, these models were created to estimate the average body burden within a species leaving much of the natural variation in these systems unexplored. Examining inter-individual differences in contaminant accumulation is important not just for a more accurate prediction of individual body burdens, but also because it represents a well modeled system that can be broken down allowing each term to be studied more carefully to determine the source of variation.

PCBs are the ideal POP reference chemical to explore variation in chemical exposures. They represent a class of closely related congeners spanning a wide range of hydrophobicities (measured by their octanol-water partition coefficient, or K_{OW}) that have been studied for decades, examining most facets of their chemistry and accumulation. It is now understood that PCBs accumulate through bioaccumulation, or a combination of bioconcentration (uptake through water alone) and biomagnification (uptake through diet) (Gobas et al 1993). Furthermore, the importance of each process is K_{OW} dependent. Likewise, the elimination of PCBs is driven by gill ventilation, fecal egestion, and growth dilution (Fisk et al. 1998; Paterson et al. 2007). Both the accumulation and elimination dynamics of PCBs are tied to an organism's physical properties as well as exposure dynamics. This allows different PCB congeners to be used as tracers for various biological, ecological, and physiological processes.

Both field based studies and models can play roles to help tease out regulators of chemical exposures to PCBs in aquatic organisms. Field based studies are important because the use of variance partitioning structure in chemical, spatial, temporal, and ecological factors in population data sets can be demonstrated. On the other hand, modeling simulations can provide insights to parameters regulating variation in community-wide food webs.

In this thesis I investigate the sources of variability in PCB congener concentrations in fish including chemical and ecological factors, such as hydrophobicity, species, and temporal and spatial variability, and food-web level drivers such as migration, diet choices, species specific growth rates, and environmental concentrations of PCBs in both sediments and water.

In my second chapter I consider the sources of variability in PCB congener concentrations in feral fish populations in the Detroit River. More specifically, I investigate the role that chemical hydrophobicity (measured by a chemicals octanol-water partition coefficient, called K_{OW}), species, season, site, and weight differences have on PCB congener variability. Since uptake of PCBs occurs through a combination of bioconcentration and biomagnification, the importance of each process being K_{OW} dependent (Gobas et al 1993), congeners should have K_{OW} dependent differences in the importance of those ecological and physiological drivers. Here I propose that hydrophobicity is the largest driver of variability followed by interspecific feeding differences, and that these species specific differences should be highest in the mid and high K_{OW} congeners. Since uptake of mid and high KOW congeners is driven mainly by biomagnification (Hebert and Haffner, 1990; Russell et al 1999; Gobas et al 1993), and hence reflect predominantly differences in diet choices, these congeners should have lower variability in benthivores, when compared to generalist feeders. This chapter explores the relationship between PCB variability, hydrophobicity, and a variety of ecological and physiological factors using a subset of 9 PCB congeners in which 3 represent low K_{OW} congeners, mid K_{OW} congeners, and high K_{OW} congeners. It uses three species of cyprinid minnows, bluntnose minnows (*Pimephales notatus*), emerald shiners (*Notropis atherinoides*) and spottail shiners (*Notropis hudsonius*), representing a benthic specialist and two generalist species, respectively. By using closely related, similar sized, and aged fish many extraneous sources of variability can be controlled. Furthermore, by collecting these minnows at three different sites along the Detroit River and over two different seasons we can determine the influence of spatial and temporal variability. Specifically, this chapter investigates five potential factors regulating PCB variability including hydrophobicity, weight, species, site, and season to quantify the variation explained by these drivers. Further, it hypothesizes that interspecific variation in bioaccumulated PCB residues should be the lowest for specialized feeders (the bluntnose minnow) which forage exclusively on the benthos and highest for generalist feeders (spottail shiner and emerald shiner) which forage both pelagically and benthically.

In my third chapter I consider the sources of variability in trophic magnification factors (TMFs), a common regulatory metric which measures the relationship between chemical concentration and trophic level. I use stochastic food web model simulations and sensitivity analyses to investigate the influences of environmental, ecological, and physiological parameters on TMFs assessing their validity as a metric of bioaccumulation potential. Here I propose that TMFs are highly affected by the heterogeneity of chemicals in their systems, influenced strongly by both background sediment and water concentrations and fish movement between high and low contaminated areas. This chapter builds on previous bioaccumulation sensitivity analyses by Selck et al. (2011) incorporating extensive diet variability and movement of organisms into a food-web level response. It uses physical characteristics, as well as, sediment and water concentrations obtained from the Detroit River, a highly heterogeneous area to evaluate the role of background sediment and water concentrations, species-specific growth rate, lipid content, feeding choice, movement, and assimilation efficiencies on TMFs. Here I test which bioaccumulation parameters TMFs are most sensitive too, with the hypothesis that it would be diet choice and we determine what effect spatial variability in PCB sediment and water concentration data has on TMFs with the hypothesis that the more spatially heterogeneous a system is the more variable a TMF is.

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CHAPTER 2

CHARACTERIZING VARIABILITY OF POPS BIOACCUMULATION WITHIN FORAGE FISH COMMUNITIES OF THE DETROIT RIVER, ONTARIO, CANADA

2.1 Introduction

Understanding the bioaccumulation and potential hazards of persistent organic pollutants (POPs) exposure has been a focus of ecotoxicological research. It is currently understood that POP bioaccumulation is most strongly influenced by three major factors: (1) physico-chemical factors of the contaminant such as hydrophobicity, measured as the octanol-water partition coefficient (or K_{OW}) (Veith and Lee, 1971; Chiou and Shoup, 1985); (2) the partitioning capacity of the organism for the contaminant of study, often associated with the whole body lipid content of the organism (Pastor et al 1996; Drouillard et al. 2004); and (3) the concentration of chemical in the organism's diet, often associated with trophic position of the organism over appropriate temporal and spatial scales (Connolly and Pedersen 1988; Clark et al 1990; Borgå et al 2004). While these factors influence bioaccumulation potentials, differences in chemical body burdens accumulated by individuals are certainly not limited to them. Differences in growth performance (Madenjian et al. 1994), ontogenetic diet shifts (Paterson et al. 2006), foraging ecology (Madenjian et al 1993; Burtnyk et al 2009), offloading of contaminants to offspring (Fisk et al 1998), and seasonal weight loss (Paterson et al 2007; Daley et al 2013) can also be responsible for much of the inter-specific and inter-individual differences observed in biota contaminant levels.

While the mechanisms of bioaccumulation are largely understood, less attention has been paid towards characterizing factors regulating individual variation in contaminant burdens. Efforts aimed at understanding bioaccumulation processes tend to distribute sampling across a variety of species within a food-web rather than focusing on high resolution sampling within a given species, particularly across spatial and temporal gradients associated with a given species' life cycle. This is especially true for forage fish species which typically receive a minimum sampling effort compared with the larger, more economically relevant sport fish (Borga et al. 2012). For instance, in a survey of

125 Lake Michigan lake trout (*Salvelinus namaycush*), fish of 620 mm total length exhibited PCB concentrations ranging from 1.5 – 6.0 mg/kg (Madenjian et al 1993). This range of concentrations observed in a narrow size class of individuals encompassed 30 % of the total range in PCB concentrations found in all surveyed lake trout (350 mm to 900 mm) from the lake (Madenjian et al 1993). In this case, individual variation in POP bioaccumulation observed in lower trophic levels is likely propagated up the food web becoming amplified in top predators, as was predicted by Individual Based Models (IBMs) (Madenjian et al 1994). However, larger long-lived top predators, such as lake trout, are likely to have incorporated chemical signatures over the course of their lives thereby integrating large temporal and spatial exposure gradients with individual differences in growth and reproductive output potentially confounding the variability in POP exposure solely associated with diet choice (Madenjian et al 1993, Lopes et al 2011). Thus, investigating variation in POP concentrations in feral fish populations requires the ability to contrast ecological factors such as habitat use, foraging ecology and diet while minimizing potential differences in species life history characteristics related to foraging range, age and growth performance.

Cyprinid species are one of the largest and most diverse groups of fishes in freshwater ecosystems and represent an ideal system for examining differences in variability in POP bioaccumulation among ecologically similar species. In contrast to top predators such as lake trout, cyprinids are small bodied, short-lived lower trophic level consumers. These characteristics minimize potential long-term variability in POP exposure propagated by factors such as ontogenetic diet shifts and larger scale habitat integration. Importantly, owing to their high diversity, multiple cyprinid species coexist within the same regions but through resource partitioning mechanisms can exploit substantially different habitat and food resources (Starrett, 1950; Scott and Crossman 1973). For example, bluntnose minnows (*Pimephales notatus*), spottail shiners (*Notropis hudsonius*) and emerald shiners (*Notropis atherinoides*) are philopatric species of similar trophic level, size, age and life history yet have distinct feeding ecologies (Scott and Crossman 1973; Keast and Webb 1966; Johnson and Dropkin 1993). The bluntnose minnow has a sub-terminal mouth evolved for benthic feeding (Starrett 1950; Scott and Crossman, 1993; Keast and Webb, 1966; Johnson and Dropkin, 1993) while

spottail shiners and emerald shiners have terminal mouths and exhibit more a generalist feeding strategy (Starrett, 1950; Johnson and Dropkin, 1993; Muth and Busch, 1989). Hebert and Haffner (1991) demonstrated differences in bioaccumulated residues between these species that were attributed to differences in their feeding ecology (Hebert and Haffner, 1991). The differences in POPs bioaccumulation among these cyprinid species were found to be K_{OW} dependent (Hebert and Haffner, 1991). These observations verified that subtle differences in species ecology were an important factor contributing to POPs bioaccumulation observed in short lived, lower trophic level fish.

Previous studies have shown that for chemicals with log K_{OW} between 3.0 - 6.0, the dominant exposure route is through bioconcentration, or uptake from water (Hebert and Haffner, 1991; Gobas et al. 1993; Haffner et al 1994; Qiao et al 2000). This leads to the prediction that individual variability of bioaccumulated residues between cyprinid species would be lower for these compounds than that of increasingly hydrophobic congeners. The prediction is made because all individuals from a similar location are exposed to a common water source and variability associated with feeding behaviour would not be propagated into intra- and interspecific exposure differences. This assumption implies that any spatial variability observed in the bioaccumulation of low to moderately hydrophobic PCBs congeners results from environmental factors affecting baseline chemical residues (point and non-point source loadings), chemical bioavailability (dissolved organic carbon and suspended particulate matter) or bioaccumulation rates (temperature).

In contrast, congeners with a log(K_{OW}) > 6.0 are bioaccumulated by a combination of exposure to water and diet (biomagnification), with diet becoming increasingly more import to exposures with increasing chemical hydrophobicity (Hebert and Haffner, 1991; Gobas et al. 1993; Russell et al. 1999). For these chemicals, both the magnitude of bioaccumulation as well as variation in bioaccumulated residues between species and among individuals are expected to increase due to time integrated differences in diet choice. Here it is anticipated that inter- and intraspecific differences in foraging habitat (i.e. pelagic versus benthic feeding) and foraging breadth (i.e. generalist versus specialist) will strongly dictate variability in bioaccumulation residues. Finally, for super hydrophobic chemicals (i.e. log K_{OW} >7), exposures to water are likely to be negligible

with food and sediment exposures (via consumption of benthic invertebrates) dominating uptake (DiPinto and Coull, 1997). Furthermore, K_{OW} dependent reductions in chemical bioavailability from food and sediments are likely to place constraints on the magnitude and variation of bioaccumulated residues for such highly hydrophobic substances (Gobas et al. 1988; Gobas et al. 1989). Given that K_{OW} dependent bioavailability reductions for super hydrophobic chemicals are more significant for sediments compared to food items (Liu et al 2010; Lamoureux and Brownawell 1999), it is anticipated that benthivores will show stronger reductions in intra-specific variation for such highly hydrophobic pollutants relative to pelagic species.

This examines three cyprinid species to determine how different feeding strategies regulate individual variability in PCB congener bioaccumulation. Furthermore, this study investigates five potential factors regulating PCB variability, one chemical (hydrophobicity) and four ecological (weight, species, site, and season), to quantify how much variation is explained by these potential drivers. Finally, the study tests the hypothesis that interspecific variation in bioaccumulated PCB residues should be minimized for specialized benthic feeders, and maximized for generalist feeders foraging both pelagically and benthically. The above hypotheses were tested by collecting the three species of cyprinids at three different sites in the Detroit River and over two seasons to contrast intra- and interspecific variation in bioaccumulated residues across time and space.

2.2 Methods

Study sites

Fish were collected by seine netting and electro-shocking from three sites in the Detroit River: (A) Peche Island; (B) Fighting Island; and (C) Boblo Dock (Figure 1). The sites were chosen from the head to the mouth of the river in order to capture the

contaminant gradient of the entire river (Drouillard et al. 2006), such that each site is distant enough to minimize migration of fish between sites.

Fish sampling

Three cyprinid species were chosen because (i) they are of similar size and age, so growth effects could be assumed negligible; (ii) they are abundant, so large numbers could be readily collected to characterize variation in chemical residues; and (iii) they have different diets and exploit different habitats. Specimens were collected twice, once in spring (June 2012), and once in the fall (September 2011) (Table 1). At each sample date specimens were collected by seine netting or electro-shocking. To minimize variability associated with growth, fish were graded into similar size classes (3.5-11.5 cm) during collections. All field work was carried out in accordance with University of Windsor's Animal Care Guidelines.

Sample analysis

Immediately following field collection, samples were wrapped in hexane rinsed aluminum foil, and frozen in food grade freezer bags until they could be processed. Processing included taking length and weight measurements followed by the homogenization of whole fish samples. Lipid and PCB concentrations were determined using a microextraction method (Daley et al. 2009). In brief, 0.5 g of homogenate was ground with mortar and pestle with 10 g of sodium sulfate. The mixture was then wet packed into a glass chromatography column with 15 mL of a dichloromethane:hexane (50:50, v/v) mixture and 50 μL of PCB 34, used as an internal standard to determine sample recoveries. After one hour and following elution of solvent, the extract was eluted with an additional 15 mL of the 50:50 extraction solutions. The extracts were evaporated under vacuum to approximately 2 mL and brought to 10 mL in a volumetric flask. From this, 1 mL was removed and neutral lipid content was determined gravimetrically (Drouillard et al. 2004). Sample cleanup was performed with 6 g of Florisil topped with approximately 1 g of sodium sulfate (Lazar et al 1992). The eluent, consisting of 50 mL of hexane, was evaporated under vacuum brought up to 1 mL final volume with iso-octane and prepared for analysis by gas chromatography-electron capture detector (GC-ECD) as per [34]. All samples were analyzed for the following PCB congeners (IUPAC #): 18/19, 31/28, 33, 52, 49, 44, 74, 70, 95, 101, 99, 87,

110, 151/82, 149, 118, 153, 105/132, 138, 158, 187, 183, 128, 177, 156/171, 180, 191, 170, 199, 195/208, 194, 205, 206, and 209.

For every extraction batch of 6 samples, a method blank and an in house reference tissue homogenate (Detroit River carp) was extracted simultaneously for quality assurance. All homogenate results were in compliance with the Great Lakes Institute for Environmental Research's s organic analytical laboratory quality assurance guidelines (PCBs in reference homogenates were always within 2 standard deviations of the laboratory control chart values). Additional details of the extraction, clean-up, and instrument conditions for analysis are provided by Daley et al. 2009 and Lazar et al. 1992. Recoveries of the internal standard averaged 89% ± 1.04% (Standard Error). Sample concentrations were not recovery corrected.

Data analysis

PCB concentrations were lipid normalized to account for differences in partitioning capacities among the species and across the temporal and spatial study design [35]. Congeners were divided into three categories based on their K_{OW} as obtained from Hawker and Connell (1988). Two congeners having high detection frequencies were selected from each hydrophobicity category to examine the general trends for that group. PCB 44 and 52 were selected from the low K_{OW} congeners (log K_{OW} of 5.75 and 5.84), PCB 138 and 153 were selected from the mid K_{OW} congeners (log K_{OW}s of 6.83 and 6.92), and PCB 194 and 199 were selected from the high K_{OW} congeners (log K_{OW} of 7.2 and 7.8). A Levene's test of homogeneity of variances was used to determine if the concentration variances of these congeners differed across the species and within the species, both temporally and spatially. All data analyses were performed using the R statistical computing program (R Core Team 2012) and a significance level of $\alpha < 0.05$ was used.

2.3 Results

Total body length of minnows were 6.9 ± 0.33 (average ± SE), while body weight and percent lipid content of collected fish were 2.6 ± 0.09, and 2.8 ± 0.15 (average ± SE), respectively. Body weights were statistically similar across species in most sites and seasons, as well as across season for most sites and species (see Table 2.1). There were very few statistical differences in lipid content among species across sites and seasons, and between seasons across sites and species (see Table 2.1).

Significant differences were observed in PCB body burdens across species, sites, and seasons (Figure 2.2). These differences were more pronounced in the spring sampling period and in most cases, bluntnose minnows had significantly lower concentrations than emerald shiners and spottail shiners. Furthermore, as can be seen in the error bars (Figure 2.2), there was significant variation in body burdens across sites, species, and seasons.

In order to first examine chemical and ecological factors explaining variability in bioaccumulated PCB residues, congeners, sites, species, and seasons were pooled together. Sums of squares were then computed on the data to determine the percent of the total variation explained by these parameters. As can be seen in Figure 2.3, chemical hydrophobicity was the largest contributor to variability (~13%), followed by species at 4%, season at 1.6%, site at 1.3%, and weight at 0.01%.

PCB congeners were then divided into hydrophobicity categories to include low K_{OW} mid K_{OW}, and high K_{OW} congener pairs and variability contrasted among the ecological factors within each hydrophobicity group.

Species accounted for the highest amount of variation among ecological factors for each hydrophobicity category. However, the amount of variability explained by species was considerably higher for mid- and high K_{OW} congeners (8.6%) compared to low K_{OW} congeners (4.6%). For the low K_{OW} congeners, variation explained by site (3.6%) approached the variation explained by species whereas the difference in variation explained by these two ecological factors was much larger for mid- and high K_{OW} chemicals. Site explained a greater amount of bioaccumulation variation than season for low K_{OW} chemicals, but these two factors were approximately equivalent for mid- and high- K_{OW} compounds. Weight accounted for less than 0.5% of the variation and was similar for all hydrophobicity categories.

Finally, samples were separated into species and hydrophobicity categories, but pooled across sites and season, in order to evaluate interspecific differences in variation of bioaccumulated residues within each hydrophobicity category (Figure 2.5). Significant interspecific differences were observed using Levene's test of homogeneity of variances in the magnitude of PCB concentration variability as estimated using coefficients of variation. Specifically, PCB congener concentrations were significantly less variable for bluntnose minnows relative to emerald and spottail shiners. No significant differences in PCB congener coefficients of variation were determined between emerald and spottail shiners. In this case, the inter-specific differences among coefficients of variation were similar across hydrophobicity categories.

2.4 Discussion

Hebert and Haffner (1991) showed that similar sized and aged cyprinid species within the same trophic level could have different PCB concentrations due to feeding differences. In the present study, data analysis focused on examining differences in the variation in PCB bioaccumulation between and within species as influenced by chemical and ecological factors. While this study utilized similar sampling strategies as described by Hebert and Haffner (1991), there were some conflicting observations compared to the previous work. Hebert and Haffner (1991) found that bluntnose minnows had the highest levels of contamination followed by the two shiner species. In the present study, there were few interspecific differences in PCB concentrations in the spring, while during the fall, bluntnose minnows had significantly lower concentrations than either shiner species (Figure 2.2). However, when sites and seasons were pooled the present results mirror those found by Hebert and Haffner (1991). This result, combined with the significantly lower coefficient of variation observed for bluntnose minnow (Figure 2.5), suggests that the shiner species were subdividing within their populations into pelagic feeders and benthic feeders. Bluntnose minnows have specific morphology to feed on benthos (Starrett 1950; Scott and Crossman, 1973; Keast and Webb, 1966; Johnson and Dropkin, 1993) and all individuals within the population are expected to track sediment-associated

contamination. Hence, their individual variability would be predicted to be lower than the spottail and emerald shiners which have a generalists feeding morphology and feed opportunistically in the water column or on benthos (Scott and Crossman 1973; Johnson and Dropkin, 1993; Muth and Busch 1989). The significantly lower variability in PCB concentrations in bluntnose minnows when compared to spottail and emerald shiners demonstrates that resource specialists have lower individual variability in bioaccumulated PCB residues than generalists. It shows that the majority of the shiners had lower overall PCB body burdens which was demonstrated when all sites and seasons were pooled, but that a small number of individuals from each site had much higher body burdens, especially in the fall, causing significantly higher concentrations in shiners, and causing the higher variability in PCB residues within the shiner species.

Intraspecific variability in contaminant body burdens has been observed in many studies (Madenjian et al. 1993; Madenjian et al. 1994; Lopes et al 2011; Borga et al 2012) and individual based bioaccumulation models have been available for almost two decades (e.g. Madenjian et al. 1993; Madenjian et al. 1994). However, studies addressing intraspecific variation in PCB burdens (e.g. Lopes et al. 2011; Selck et al 2011) have investigated causes of variability in sum PCB concentrations negating investigation of the effect of chemical hydrophobicity. In addition, the use of long-lived fish, or top predators, to explore intraspecific variation can be confounded by long time periods and/or spatial scales of integrated exposures over which individual differences in performance and behavior attributes can change (Madenjian et al. 1993; Madenjian et al. 1994). The present study investigated variability in congener-specific PCB residues spanning a broad range of hydrophobicities using shorter lived fish populations to tease out chemical and ecological factors responsible for contributing to variation within and between closely related sympatric species. Furthermore, by characterizing factors regulating variation in bioaccumulated residues in key forage fish species, the propagation of uncertainty in exposures to upper trophic level piscivores, via variation in ingested diet items, can be better understood and related to physical-chemical properties as well as food web and ecological characteristics of the study system.

While previous studies have suggested that variability in PCB body burdens in piscivores resulted from interspecific differences in diet choice (Madenjian et al. 1993;

Madenjian et al. 1994), variability in contaminant body burden is regulated by both chemical and biological processes, including seasonal weight loss (Paterson et al. 2007), amount of lipids (Hebert and Keenleyside, 1994), offloading of contaminants to offspring (Fisk et al. 1998), differences in growth performance (Madenjian et al. 1994), foraging differences (Madenjian et al. 1993) and variation in toxicokinetic parameters such as chemical assimilation efficiencies and/or elimination rate coefficients (Drouillard et al. 2009; Liu et al. 2010). By size grading animals at sampling, targeting of short lived species and lipid-normalizing the data, the influences of growth performance and lipid levels on differences in PCB body burdens were minimized. This explains why body weight was observed to contribute to very little of the PCB variability (<0.5% of the differences). The study findings indicate that approximately 20% of the total intra- and interspecific differences in bioaccumulated PCBs residues among three species of Detroit River cyprinids can be explained by chemical properties (13%), followed by species specific physiological and ecological traits (4%), site differences (1.6%), and seasonal differences related to sampling (1.3%).

An extensive body of work exists suggesting that uptake and elimination dynamics depend strongly on congener hydrophobicity (Paterson et al. 2007, Gobas et al. 1988; Gobas et al. 1989; Liu et al. 2010). It has been demonstrated that congeners with a log K_{OW} > 6.5 are poorly eliminated, especially at low temperatures (Paterson et al. 2007; Drouillard et al. 2009). However, much less is known about the interaction between chemical K_{OW} and variability in toxicokinetic parameters that contributed to variation in bioaccumulated residues. The results from the present study reveal that K_{OW} is an important factor regulating not only the magnitude of bioaccumulation, but also individual variation of bioaccumulated contaminant concentrations in fish populations. This has important implications to risk assessment. For example, safety factors are often used to account for uncertainty in exposure and toxicity when extrapolating laboratory to field effects in risk assessments used for wildlife hazards and/or generating fish consumption advice information. The present study suggests that safety factors should be scaled according to chemical hydrophobicity when applied to POPs as opposed to applying a uniform safety factor (commonly 10) for all chemicals.

Across chemical hydrophobicities, low K_{OW} congeners had the least amount of variation explained by species differences. The low variation observed for low- K_{OW} congeners across species is consistent with bioconcentration, i.e. uptake from water, playing a larger role to chemical exposures for these compounds with individual variation tracking small spatial/temporal scale differences in water concentration and/or chemical bioavailability in water. Alternatively, mid- and high K_{OW} congeners exhibited greater amounts of variation explained by differences among species. Since the three cyprinid species chosen were of similar sizes, ages, and trophic levels, the predominant differences observed among them were likely attributed to dietary differences (Starrett, 1950; Scott and Crossman, 1973; Hebert and Haffner, 1991). Biomagnification represents a more complex exposure pathway that is dependent on the feeding history of individuals, hence propagating variation in individual diet choices at intra- and interspecific scales. This suggests that the greater degree of variability observed for the mid- and high-range K_{OW} congeners is due, predominantly, to variation in biomagnification, and hence variation in dietary sources.

Results from this study also demonstrated smaller amounts of variation contributed by spatial and temporal differences in sampling. Site specific differences in variation are likely related to heterogeneity in contaminant concentrations within water and sediments as previously characterized for the Detroit River (Drouillard et al. 2006; Drouillard et al. 2013). Seasonal differences in variation may be explained by temperature/toxicokinetic parameter interactions. Changes in temperature have significant effects on fish metabolism limiting their ability to eliminate contaminants (Paterson et al. 2007). While temporal and spatial differences did not account for as much of the variation as inter-specific differences, they accounted for significantly more variability than body weight.

Based on the present study findings, it is concluded that variability in PCB bioaccumulation within cyprinids is driven largely by chemical hydrophobicity, followed by species specific foraging strategies, sampling location and sampling season. Individual variability in bioaccumulated PCB residues was species dependent with bluntnose minnows, a benthic specialist, demonstrating less variability than the opportunistic emerald and spottail shiner species, supporting a role of sediment bioavailability as a

factor regulating intra- and interspecific variation in chemical exposures in forage fish. To fully understand the bioaccumulation process, and in particular individual variability in bioaccumulated residues, one has to begin to understand habitat use and feeding ecology of the species in question, and should take these into consideration when constructing sensitivity analyses for congener specific bioaccumulation models and uncertainty propagation of bioaccumulated residues generated from food web bioaccumulation models.

2.5 References

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Table 2.1. Sample size (n) and average (± 1 SD) length, mass and lipid content data for bluntnose minnow and spottail and emerald shiners collected during fall (2011) and spring (2012) from Boblo Dock, Fighting Island and Peche Island locations within the Detroit River where (* and †) indicate significant differences in mass and lipids between seasons within site and species, and the superscript a and b indicate differences in mass and lipids between species within site and season ($p < 0.05$).

	Site	Season	n	Mass (g)	Lipid (%)
Bluntnose minnow	Boblo Dock	Fall	24	3.3 ± 1.5^{a}	2.9 ± 1.3^{a}
		Spring	12	3.3 ± 0.7	3.8 ± 1.9^{ab}
	Fighting Island*	Fall	20	1.7 ± 0.7^{a}	3.8 ± 3.7
		Spring	12	3.7 ± 0.7^{a}	3.6 ± 1.4^{ab}
	Peche Island*†	Fall	16	1.7 ± 1.9	1.8 ± 1.1
		Spring	12	3.7 ± 0.6^{a}	3.1 ± 1.9
Spottail shiner	Boblo Dock*	Fall	7	3.2 ± 2.4^{b}	1.9 ± 1.3^{a}
		Spring	12	2.7 ± 1.3	1.9 ± 1.3^{ab}
	Fighting Island*	Fall	4	1.3 ± 0.8^{ab}	1.6 ± 1.2
		Spring	12	2.0 ± 0.5^{b}	1.8 ± 1.0^{ab}
	Peche Island	Fall	7	1.6 ± 0.7	1.9 ± 1.3
		Spring	12	2.4 ± 0.4^{a}	4.3 ± 2.3
Emerald Shiner	Boblo Dock	Fall	13	0.9 ± 0.4^{a}	1.7 ± 1.3^{a}
		Spring	12	2.8 ± 1.3	2.5 ± 1.6^{a}
	Fighting Island	Fall	4	0.8 ± 0.3^{ab}	2.5 ± 0.7
		Spring	12	2.9 ± 0.9^{b}	3.3 ± 2.3^{ab}
	Peche Island*†	Fall	7	1.8 ± 0.8	1.9 ± 1.1
		Spring	12	4.0 ± 0.4^{b}	4.0 ± 4.0

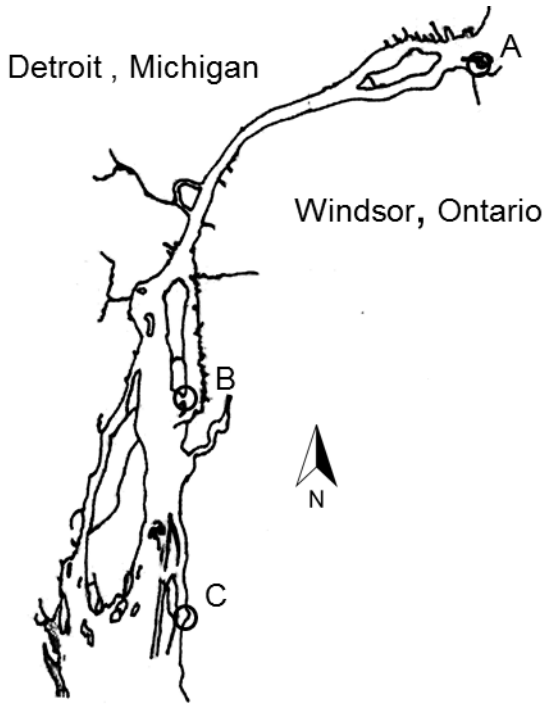

Figure 2.1. Locations of sampling sites where (A) is Peche Island, (B) is Fighting Island, and (C) is Boblo Dock.

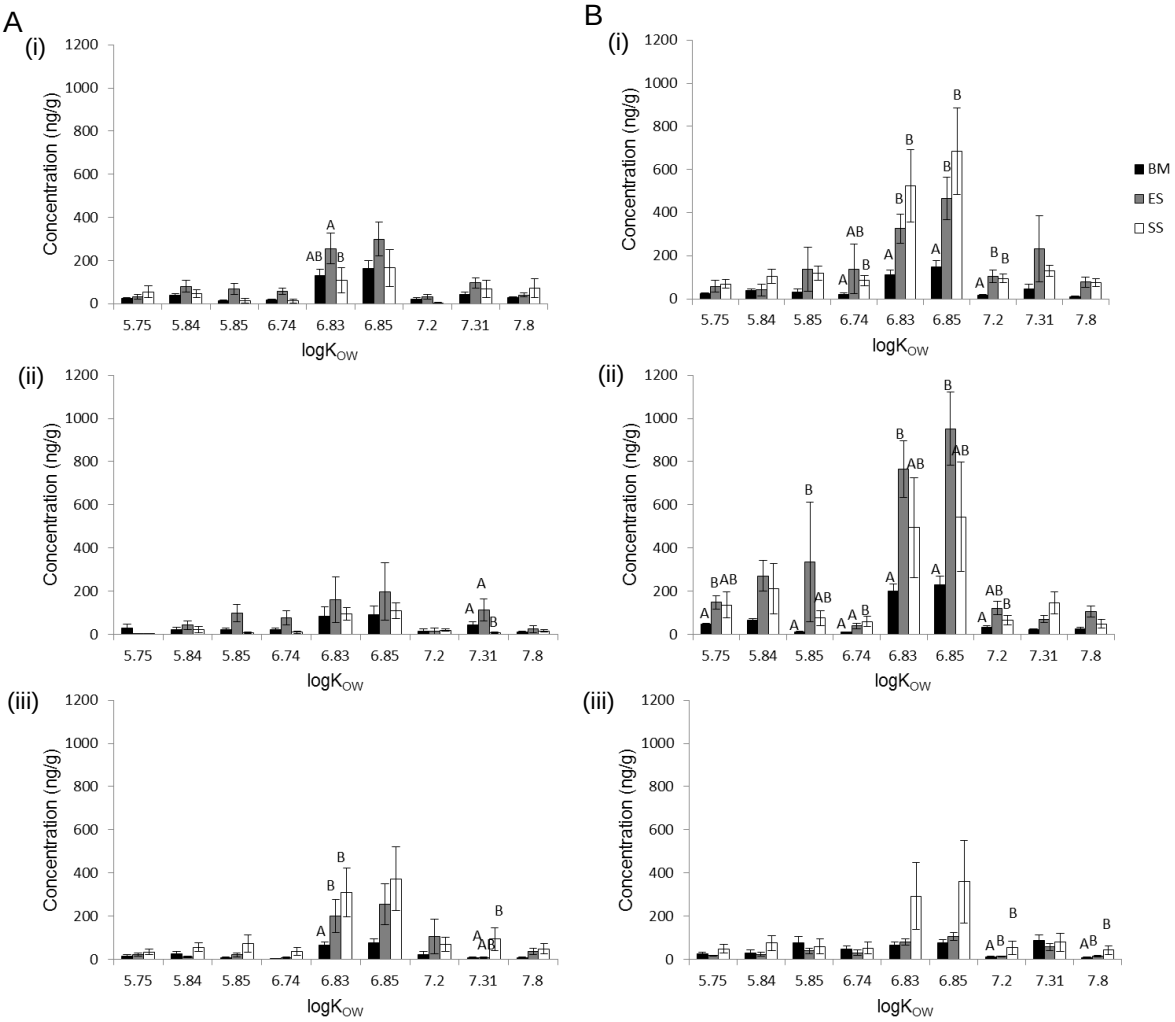

Figure 2.2. Concentrations of individual PCB congeners (PCB 44, 52, 138, 153, 194, 199, respectively; in ng/g ± standard error) broken down by site, species, and season, where (A) is fall sampling, and (B) is spring sampling, for (i) Boblo Dock, (ii) Fighting Island, and (iii) Peche Island. The letters indicate significant interspecific differences from an ANCOVA with weight as the covariate ($p < 0.05$).

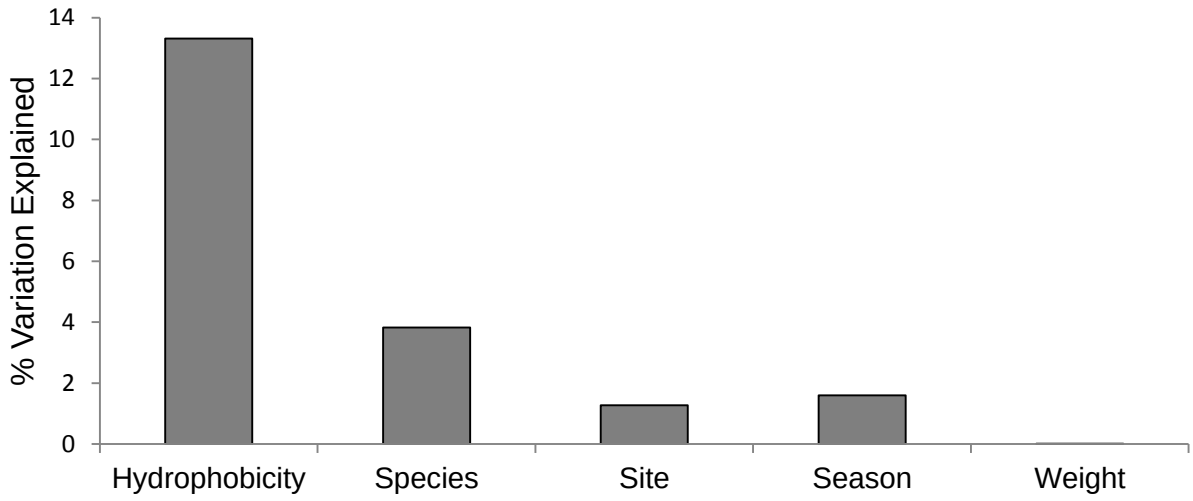

Figure 2.3. Amount of variation in PCB concentrations explained by hydrophobicity, species, site, season, and weight.

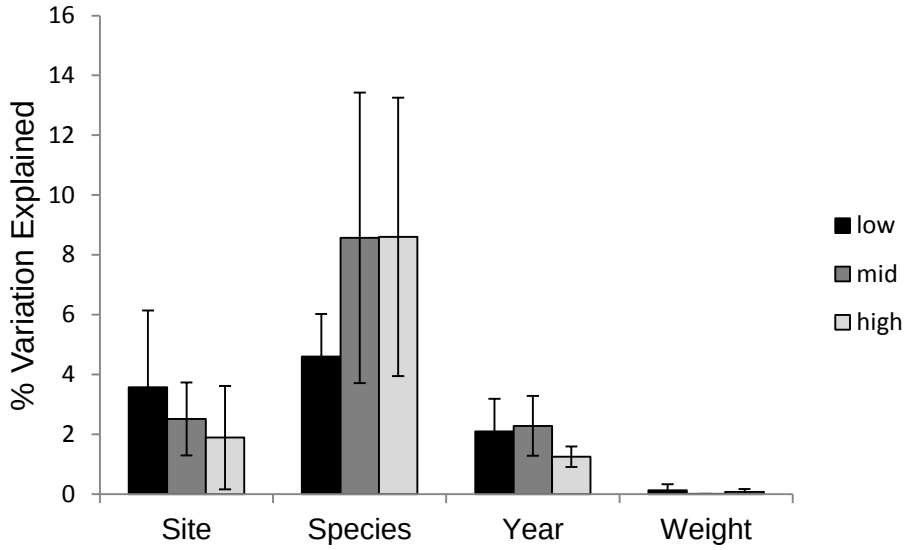

Figure 2.4. Amount of variation explained (in percentage of total variation) for site, species, year, and weight compared between the three different hydrophobic groups (low, medium, and high KOW).

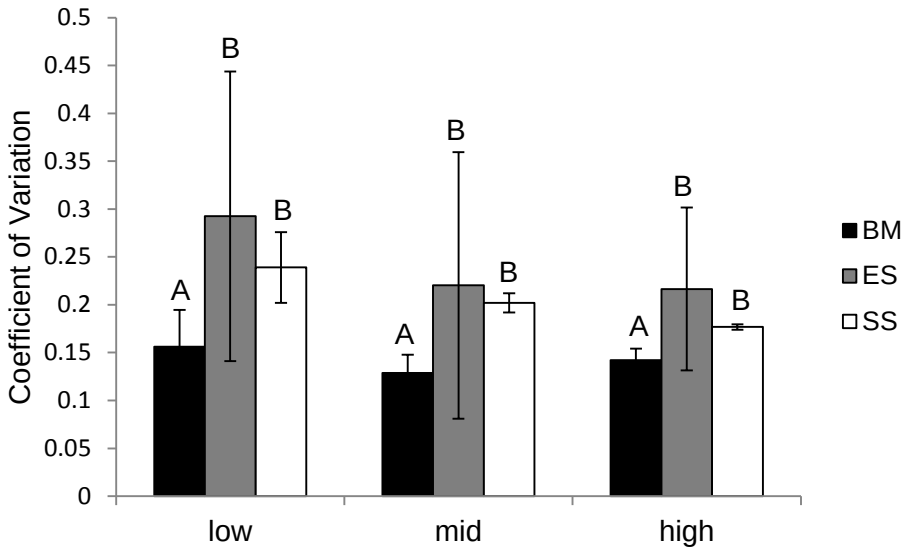

Figure 2.5. Average coefficient of variation (CV) of PCB congeners quantified in bluntnose minnows (BM) and emerald (ES) and spottail shiners (SS) collected from three locations within the Detroit River where error bars indicate standard deviation. Letters indicate significant differences between congener concentration variability as indicated by Levene's test of homogeneity of variances ($p < 0.05$).

CHAPTER 3
USE OF MODELS TO SIMULATE UNCERTAINTY AND STATISICAL POWER IN THE TMF APPROACH

3.1 Introduction

Trophic magnification factors (TMFs) provide a method of assessing food web biomagnification through integration of bioaccumulation processes occurring in individuals, species, and trophic levels in a given ecosystem. They are increasingly being used by policy makers to screen emergent chemicals as they provide a metric of an average food-web biomagnification factor (FWBMF) (Fisk et al. 2001; Jardine et al. 2006) giving an empirically definitive measure of bioaccumulation potential; especially relative to other metrics of bioaccumulation such as bioconcentration factors (BCFs) and biomagnification factors (BMFs) (Gobas et al. 2009). For example, there are classes of compounds, such as the phthalate esters, which have high BCFs (exceeding 1000) and high K_{OW}s ($\log K_{OW} > 5$), but do not biomagnify. This can be seen by their TMFs which are less than 1, indicating that biotransformation attenuates food web biomagnification processes especially at upper trophic levels (Mackintosh et al. 2004).

The TMF approach assumes that diet is the major contaminant exposure route, and more specifically, that organism trophic level is directly correlated to chemical concentrations present in an animal's diet and that biomagnification within organismal tissue occurs (Borgå et al. 2012). Unfortunately, empirical testing of TMFs may be confounded by many factors influencing chemical toxicokinetics within organisms. While organism diet and trophic level are important factors governing persistent organic pollutant (POP) bioaccumulation (Connolly and Pedersen, 1988; Clark et al. 1990; Borgå et al. 2004), chemical accumulation is also affected by other processes. Species and individual specific characteristics including differences in metabolic biotransformation capacity (Borgå et al. 2004); growth performance (Madenjian et al. 1994), offloading of contaminants to offspring (Fisk et al 1998), seasonal weight loss (Paterson et al 2007; Daley et al 2013), omnivorous feeding and ontogenetic diet shifts (Paterson et al. 2006), age, and size can all influence chemical toxicokinetics and observed bioaccumulation

potentials in individuals and populations, contributing to variation and lower statistical power of detecting TMFs greater than one (Borgå et al. 2012).

Moreover, TMF calculations can be further complicated by different foraging ranges among species included in the study; larger foraging ranges create open foodwebs instead of closed ones. This could be of particular importance when there exists a high degree of spatial heterogeneity of chemical concentrations in water and sediments (e.g. the Detroit River (Szalinska et al 2011), and the Hudson River (Bopp et al. 1981)). In this case, organisms exhibiting a high degree of spatially integrated chemical exposures, typical of mobile top predators, can contribute to either under- or overestimates of TMF when paired with spatially restricted prey items collected over small spatial scales. Indeed, Borgå et al. (2012) highlighted a common sampling issue in TMF studies, whereby the collection of lower trophic level organisms is typically under represented with respect to sampling effort and replication afforded to upper trophic level organisms. This contributes to issues of statistical power related to detecting TMFs significantly greater than a value of 1, but also has the potential to contribute to spatial sampling artifacts associated with TMF estimates. For example, when all organisms used in a TMF study are collected at a highly contaminated location, TMFs can be underestimated due to a raise in the TMF intercept value. This occurs because benthic organisms are more likely to exhibit constant exposures to the contaminated location where they were collected over their life spans (Zstolt et al 1989) whereas top predators will be exposed to a combination of clean and contaminated areas within the study system as reflected by their overall foraging range.

Recent studies have attempted to characterize the individual effects of chemical, physiological, ecological, and environmental characteristics on bioaccumulation (Selck et al. 2012) and TMFs (Borgå et al. 2012). Reviews of the TMF approach have suggested the use of bioaccumulation models to examine the aforementioned factors, and establish TMF simulations as screening tools for emerging chemicals of interest. Alternatively, use of food web bioaccumulation models make it possible to propagate uncertainty in TMF calculations and examine the influence of variation in model inputs, toxicokinetic parameters, animal bioenergetic performance, and ecological characteristics on the magnitude and statistical power of TMFs.

This study used a general food-web bioaccumulation model (Arnot and Gobas, 2004) to explore the relative importance of various model parameters including model inputs (chemical concentrations in water and sediments), physiological factors (species specific biological attributes) and ecological factors (diet matrix, effective trophic position and foraging range) on TMFs. The model was adapted to explore the above attributes as applied to a hypothetical food web in the Detroit River, which was chosen because it represents a well-known, well sampled system demonstrating spatial heterogeneity in contaminated water and sediments (Drouillard et al. 2013; Drouillard et al. 2006) and having shown to exhibit food web biomagnification of various POPs (Russell et al. 1999). The objectives for this study were to; (1) determine the sensitivity of TMFs to model inputs, physiological and ecological parameters included in the model, (2) determine the effects of these parameters on uncertainty in TMFs, specifically to pinpoint which parameters need more precise estimation given realistic constraints on model input and parameter uncertainty, and (3) to determine the influence of spatial heterogeneity in water and sediment contamination and how this interacts with different foraging ranges of fish on estimated TMFs.

3.2 Methods

Food web model description

Model simulations were performed using the general one-compartment food-web bioaccumulation model developed for organic contaminants by Arnot and Gobas (2004). This model builds off of the general concepts provided by Thomann and Connolly (1984) integrating predictive algorithms and parameter estimates provided by numerous subsequent studies (e.g. Clark et al 1990; Gobas 1993; Morrison et al. 1997). The basic model equation is as follows:

(eq 1.) $$\frac{dC_{org}}{dt} = k_w \times C_w + k_{(p,w)} \times C_{(p,w)} + k_{diet} \times C_{diet} - \left(k_2 + k_{ex} + k_m + k_g\right)C_{org}$$

where the C_w, $C_{(p,w)}$, C_{diet}, and C_{org} represent chemical concentration in water, pore water, diet, and organism respectively and the k_i's are first order rate coefficients defined as: the uptake rate coefficient from water (k_w; $mL \cdot g^{-1}_{wet\ wt} \cdot d^{-1}$), the uptake rate coefficient from ingested diet items (k_{diet}; $g_{food} \cdot g^{-1}_{wet\ wt} \cdot d^{-1}$), elimination rate coefficient to water (k_2; d^{-1}), fecal elimination rate coefficient (k_{ex}; d^{-1}), metabolic biotransformation rate coefficient (k_m; d^{-1}), and the growth dilution rate coefficient (k_g; d^{-1}). Model inputs include chemical concentrations in water (ng/mL), pore water (ng/mL) and sediments (ng/g dry sediments). Model outputs consist of wet weight chemical concentrations (ng/g) in tissues of all animals included within the food web simulation.

Since the model required integration of an entire food-web, multiple diet items were included and for some simulations these varied for each iteration when simulations were performed stochastically. This does not, however, change the steady-state equation, and when equation 1 is expanded and solved for steady-state assumptions it is as follows:

(eq 2.) $$C_{ss} = \frac{\sum_{i=1}^{n}\left(p_{food,i} \times G_d x E_d \times\right) + G_v \times E_w \times C_{(p,w)} \times p_{(p,w)} + G_v \times E_w \times C_{w(o,w)} \times p_{(o,w)}}{\frac{E_w \times G_v}{K_{BW}} + \sum_{i=1}^{n}\left(G_{f,i} \times E_d \times \frac{p_{f,lipid,i} \times K_{OW} + p_{f,NLOM,i} \times K_{OW} \varphi_{NLOM} + p_{f,w,i}}{K_{BW}}\right)}$$

where the Gs are the organism's feeding, gill ventilation, fecal production rates and growth rates (G_d (in $g_{food} \cdot g_{BW} \cdot d^{-1}$), G_v (in $mL \cdot g_{BW} \cdot d^{-1}$), G_f($g_{BW} \cdot g_{BW} \cdot d^{-1}$)), the E's represent the organism's chemical assimilation efficiency for water and food (E_w and E_d, respectively), the p's represent the proportion of pore water, overlying water, diet item i consumed, lipid in feces for diet item i, NLOM (non-lipid organic matter) in feces for diet item i, and water in feces for diet item i ($p_{(p,w)}$, $p_{(o,w)}$, $p_{food,i}$, $p_{f,lipid,i}$, $p_{f,NLOM,i}$, and $p_{f,water,i}$, respectively), and the Cs represent organism concentration at steady-state, concentration in diet item i, concentration in pore water, and concentration in overlying water (C_{SS} ($ng \cdot g^{-1}$ BW), $C_{f,i}$ ($ng \cdot g^{-1}$ food), $C_{(p,w)}$ ($ng \cdot mL^{-1}$), and $C_{w(o,w)}$ ($ng \cdot mL^{-1}$), respectfully). It is assumed that E_d is constant for different types of diet items consumed by an individual, that $p_{(p,w)}$ and $p_{(o,w)}$ sum to one, and $\sum p_{food,i} = 1$. For fish, $p_{(p,w)}$ is set zero and assigned a value greater than zero for benthic invertebrates. K_{BW} represents the

biota water partitioning coefficient, and φ_{NLOM} is the NLOM partitioning equivalent in the organism compared to octanol. C_{ss} is subsequently expressed on a lipid equivalent basis ($C_{ss(leq)}$) according to:

(eq. 3) $$C_{ss(leq)} = \frac{C_{SS}}{p_{org,lipid} + p_{org,NLOM} \times \varphi_{NLOM}}$$

where $p_{org,lipid}$, $p_{org,NLOM}$ refer to the proportion of lipid and non-lipid organic matter in the organism, respectively.

Equation 1 differs for phytoplankton. In this case uptake from food (k_{diet}), elimination to feces (k_{ex}), metabolism (k_m) and growth (k_g) are omitted. k_w and k_2 are calculated by the following submodels:

(eq. 4) $$k_w = (A + \left(\frac{B}{K_{OW}}\right))^{-1}$$

(eq. 5) $$k_2 = \frac{k_w}{K_{BW}}$$

where A and B are constants assigned values of 6.0 x 10^{-5} and 5.5 respectively.

K_{BW} is the biota-water partition coefficient and is calculated in the following way;

(eq. 6) $$K_{BW} = p_{lip} \times K_{OW} + p_{NLOM} \times K_{OW} \times \varphi_{NLOM} + p_w$$

where the ps are proportion lipid, NLOM, and water in the organism (p_{lip}, p_{NLOM}, and p_w, respectively), and φ_{NLOM}, the NLOM partitioning equivalent in the organism compared to octanol, is set at 0.05 for all organisms and diet items (Debruyn and Gobas, 2007).

The $G_{f,i}$, the fecal production rate of dietary item i, is modeled in the following submodel;

(eq 7.)

$$G_{f,i} = G_d \times p_{food,i} \times \left[\left(1 - AE_{lip}\right) \times p_{lip,diet\ i} + (1 - AE_{NLOM}) \times p_{NLOM,diet\ i} + (1 - AE_w) \times p_{w,diet\ i}\right]$$

where the AEs represent the dietary assimilation efficiencies of lipid, non-lipid organic matter (NLOM), and water (AE_{lip}, AE_{NLOM}, and AE_w, respectively) from a given ingested food item (i), and the p's represent the proportion of lipids, NLOM, and water in diet item i ($p_{lip,diet\ i}$, $p_{NLOM,diet\ i}$, $p_{w,diet\ i,}$), respectively. The exception to Eq. 6 is for ingested sediment (G_{fsed}) which is calculated according to:

(eq 8.) $$G_{f,sed} = (p_{sed} \times (1 - AE_{sed}))$$

where p_{sed} is the proportion of sediment in the diet, and AE_{sed} is the bulk sediment assimilation efficiency. Elimination of chemical to feces generated by sediment ($K_{ex,sed}$) is modeled according to:

(eq 9.) $$K_{ex,sed} = G_{f,sed} \times \frac{f_{OC,sed} \times K_{OC}}{K_{BW}}$$

where $f_{oc,sed}$ represents the fraction of organic carbon in egested sediments, assumed to be similar to bulk sediments and K_{OC} is the organic carbon water partition coefficient. K_{OC} is estimated to be $0.35 \cdot K_{OW}$ (Boethling and Mackay, 2000).

Benthic invertebrates are able to accumulate PCBs through gill ventilation of the pore water associated with the sediment. Pore water concentrations are assumed to be in equilibrium with sediment and estimated as:

(eq 10.) $$C_{(p,w)} = \frac{C_{sed}}{K_{OC} \times D_{sed}}$$

where K_{OC} is the organic carbon – water partition coefficient, D_{sed} is the sediment density, and C_{sed} is the concentration of contaminant in the sediment.

TMFs were calculated using the food web bioaccumulation model outputs related to PCB concentrations achieved in animal tissues ($C_{ss,leq}$) against organism trophic level. The trophic level of organism included in the model was calculated using the following equation:

(eq 11.) $$TL = 1 + \sum_{i=1}^{n}(p_i \times TL_i)$$

where p_i is proportion of diet item I, and TL_i is trophic level of organism i. Sediment and phytoplankton were assigned TL's of 1, while zooplankton was assigned a TL of 2. TMFs were then estimated by performing linear regressions on the log-transformed, lipid-normalized PCB concentrations against assigned organism trophic levels:

(eq 12.) $$\log(PCB_{lipid}) = b + (m \times TL)$$

(eq 13.) $$TMF = 10^{m}$$

where b and m are the intercept and slope, respectively, of the regression line.

Most of the parameters in the model were allowed to vary during stochastic simulations, or varied because components in their submodel were allowed to vary (Table 1). Chemical partition coefficients K_{OW}, K_{OC}, and φ_{NLOM} were fixed because they are considered physical constants. E_w and E_d were allowed to vary by adding a variable constant a and b, respectively. Constant a caused E_w to vary according to a triangular distribution between 0.11 and 0.6 with the empirically calculated E_w as the peak. Constant b caused E_d to vary according to a triangular distribution between 0.23 (or the empirically calculated E_d if it was lower) and 1.01 with the empirically calculated E_d as the peak. Finally, temperature was allowed to vary on a lognormal distribution, causing C_{O2}, G_d, and G_v to vary.

Model Simulations

Four different model simulations were used. The first used the river wide average water and sediment concentrations for the Detroit River and the best estimates for each model parameter to provide a deterministic estimate of the TMF for 35 PCB congeners where input data on river-wide water and sediment concentrations were available. This simulation is referred to as the baseline simulation and forms the basis on which to compare TMF estimates and variation of TMF under probabilistic simulations.

The second set of simulations served as a sensitivity analysis where model inputs and parameters were allowed to vary in a consistent way under Monte Carlo simulations to provide a probabilistic estimate of TMF for comparison with the baseline TMF value. In each case, simulations were performed for 1000 iterations in which the specified parameter(s) were allowed to vary randomly within specified constraints. In this scenario, model inputs (river wide mean water and sediment concentrations) and model parameters were given a lognormal probability distribution with a standard deviations equal to 25% of the mean (referred to hereafter as the 25% simulation). A series of model simulations were run where only a single input or parameter was allowed to vary under each model iteration to determine how each model input or parameter contributes to variation in the estimated TMF and to specify which parameters the model is most sensitive to. Finally, a combined sensitivity simulation was generated such that all model inputs and parameters were allowed to vary during each model iteration using a 25% standard deviation rule to contrast variation in TMF against the baseline value. Here, random values for individual model inputs and parameters were chosen in the Monte Carlo analysis, i.e. the choice of one value for a given parameter in the model did not affect the choice of another parameter or input used within the simulation iteration. Model sensitivity analysis was restricted to a selected set of PCB congeners (PCBs #31/28, 153, and 194) reflecting different hydrophobicities over the log K_{OW} range of 5.67-7.8.

The third set of simulations attempted to utilize more realistic constraints on model inputs and model parameters during Monte Carlo simulations. Here, the actual standard deviations for river wide Detroit River water and sediment concentrations were utilized as well as best estimates of model parameter variation as described in Selck et al. (2012). This set of simulations is referred to as the model uncertainty analysis. Similar to

the sensitivity analysis, the model was run first by altering only a single input or parameter at a time to test the influence of each variable on model TMF predictions. This was followed by a combined simulation where all inputs/parameters were allowed to vary simultaneously to estimate uncertainty in model predicted TMFs against the baseline prediction. Similar to sensitivity analysis, model uncertainty analysis was restricted to the selected PCB congeners.

Finally, the last set of simulations was performed to address the interaction between spatial variation in model inputs and fish movements on estimates of TMFs. For this set of simulations, the Detroit River was divided into six food web zones (Figure 1) demonstrated to exhibit differences in sediment and water contamination (Drouillard et al. 2006; Drouillard et al. 2013). Baseline deterministic simulations were performed to contrast river wide and zone-specific TMF estimates without allowing for organism movement between zones or parameter uncertainty. Probabilistic simulations were subsequently performed such that plankton, zooplankton and benthos were assumed to remain within a given food web zone over their entire lifespans, whereas fish were allowed to move as established through a literature review of species specific foraging ranges described in Kashian et al. (2010). In the latter simulations, the model input (zone specific water and sediment concentrations) and parameter uncertainties were established according to the same constraints used in the uncertainty simulation trials. Fish foraging ranges were incorporated into the model by considering a weighted average PCB concentration in diet items in a given species and food web zone based on species specific foraging coefficients. Specifically, organisms consumed, on average, the same proportion of different diet items that they did for the river wide mean, and multi-zone models, however, the allocations of these proportions differed for each organism based on species specific foraging proportions derived using estimates provided by Kashian et al. (2010). For each food item consumed by a given fish species, the locations, and hence PCB concentration, of contaminant in that food item was spread across food web zones depending on the species foraging potentials. In this case, the weighted average concentration of a given ingested food item was calculated based on the sum of proportions of time spent in each food web zone multiplied by the concentration of chemical in the food item from each zone. In these simulations the sum proportion of diet

items was allowed to vary as described previously, and the species specific foraging potentials were allowed to vary on a lognormal distribution ± 25% of the mean foraging potential. Hence, for these simulations the amount of a specific diet item consumed in each zone varies as well as the foraging potential of that organism in each zone.

3.3 Results and Discussion

Under the baseline simulation scenario, the model predicted TMFs were greater than 1 for the majority of PCB congeners. Figure 2 presents lipid-equivalent log(concentration) for representative low, medium, and high $\log K_{OW}$ congeners (PCBs 31/28, 153, and 194 respectively) against animal trophic level. A plot of TMFs against chemical K_{OW} for all PCB congeners ($\log K_{OW}$ ranging from 5.67-8.09) under the baseline scenario is provided in Figure 3. The relationship between model predicted TMF and $\log K_{OW}$ does not resembles those observed in the literature (e.g. Hoekstra et al. 2003; Walters et al. 2011). In this case, the baseline model predicts TMFs to be highest for mid-range K_{OW} congeners and lowest for congeners with very high K_{OW}s (Figure 3). In field studies comparing hydrophobicity with TMFs, there is a general positive correlation between TMF and chemical K_{OW} and little evidence for subsequent declines in TMF at very high K_{OW}s (e.g. Hoekstra et al. 2003; Walters et al. 2011). The baseline model predicted trend in TMF against K_{OW} is strongly influenced by the algorithm specifying organism chemical assimilation efficiency from food which significantly drops for highly hydrophobic chemicals. However, Liu et al. 2010 demonstrated large variation in both the dietary assimilation efficiency of PCBs in fish across different types of food items as well as differences in the E_D vs. K_{OW} relationship. Importantly, for some diet items, model estimated E_D's for high K_{OW} chemicals tended to be underestimated compared with measured values reported by Liu et al. (2010). TMFs vary in magnitude in different lake systems, however, our predictions were within range for those PCBs with log K_{OW}s < 6.5. For instance, TMFs for different food-webs in lakes across North America ranged between 1.0 – 4.5 for PCB 52 to 1.5 – 6.0 for PCB 153 congeners (Houde et al. 2008), while ours were 1.8 and 2.8 respectively.

The sensitivity analysis performed on the river-wide model simulations suggests that the model is most sensitive to changes in the dietary assimilation efficiency of

chemical for each of the selected PCB congeners (Figure 4). Mean TMF estimates generated when this parameter was allowed to vary according to a triangle distribution resulted in underestimates the TMF relative to the baseline value by as much as 20% for the mid K_{OW} congener and 15% for the high and low K_{OW} congeners, respectively. The sensitivity analyses further demonstrates that degree of effect differs between congeners. For instance, in low K_{OW} congeners $p_{(o,w)}$ has a much stronger effect than diet choice as compared to the mid and high K_{OW} congeners. Interestingly, all the parameters influence TMFs in the same directions for each congener with the exception of concentrations in the water and sediment which have K_{OW} dependent effects. This result reflects differences in accumulation pathways between congeners of differing hydrophobicities and highlights the importance of treating PCBs as individual chemicals instead of as a sum. Furthermore, the congeners differ in the degree of error generated for TMF estimates when individual parameters were allowed to vary. For instance, E_d has a much larger standard deviation for the high K_{OW} congeners than the low and mid K_{OW} congeners during sensitivity trials (Figure 4). Variation in the assimilation efficiencies of non-lipid organic matter and water had a less than 1% effect on TMF so they were omitted from Figure 4.

The model uncertainty simulation results suggest differing importance of parameters compared to sensitivity trials (Figure 4). For the low K_{OW} congeners, variation in $p_{(o,w)}$ has the largest effect on TMFs, followed closely by E_w. For the mid and high K_{OW} congeners, E_d is the most influential parameter, followed by diet choice while $p_{(o,w)}$ has a very low influence on TMFs. The uncertainty scenarios typically had similar or larger errors associated with them than the sensitivity analyses. This occurs because the error ranges of model inputs and several parameters exceeded the 25% standard deviation used in the sensitivity analysis. The exceptions were for the following parameters, organism lipid content, and AE of lipid and $p_{(o,w)}$ which had lower uncertainty than the 25% standard deviation used sensitivity simulations.

Similar to sensitivity trials, allowing for model input and parameter variation in model uncertainty simulations were shown to have different effects on TMF estimates depending on the PCB congener examined. The simulations also demonstrate that while TMFs are most sensitive to assimilation efficiency of lipids under sensitivity analysis,

realistically, organisms vary little in respect to this parameter (Arnot and Gobas, 2004; Selck et al 2011), especially in contrast to other parameters such as E_d (Liu et al 2010) and diet proportion which had a greater impact to TMF variation under the uncertainty simulations. These observations emphasize the need for thorough knowledge of these ecological and physiological parameters when establishing model simulations of TMF using food web bioaccumulation models. Species specific and diet specific information related to organisms included in the simulation would be preferred over general parameter estimates.

While this study is unique in examining uncertainty and sensitivity of TMFs to parameter variation, there have been previous studies examining sensitivity and uncertainty for food web biomagification. These studies demonstrate that model sensitivity changes depending on the organism and trophic position; however, there are similarities between the driving parameters. For instance, chemical assimilation from food is a key driver of uncertainty in TMFs and also in food web biomagnification studies done on Lake Ontario (MacLeod et al. 2001). However, concentration in the sediments and water is a very large driver in these studies (e.g. MacLeod et al. 2001; de Laender et al. 2010), while it describes only a small fraction of variability in ours (Figure 4).

The sensitivity simulations predicted TMFs closer to the baseline TMF than the uncertainty simulations for low, medium, and high K_{OW} congeners (see Figure 5). The sensitivity simulations, however, tended to underestimate the TMF, while the uncertainty simulations overestimated them. The 95% confidence intervals (CIs) for the uncertainty simulations were much larger than those for the combined sensitivity trials. These results demonstrate that incorporation of realistic parameter variation have a net positive effect on TMF, with large associated errors, while utilization of a default 25% variation across parameters and model inputs results in TMFs being skewed lower by the strong effect of assimilation efficiency of lipids underestimating the TMF, but with much smaller variation across simulation iterations.

For the three PCBs studied in the present research, the probability that the stochastically generated TMF simulations exceed the baseline TMF differs according to chemical hydrophobicity. The probability of over-estimating the TMF against the

baseline value for PCB 31/28 for the combined sensitivity trial is $p > 0.85$, while the probability of over-estimating the TMF of PCB 153 and 194 are $p< 0.05$, and $p > 0.4$, respectively. The probability that the stochastic generated TMF simulations exceed the baseline TMF for the combined uncertainty trial are much higher with $p > 0.9$ for PCB 31/28, $p > 0.9$ for PCB 153 and $p = 1$ for PCB 194. The elevated TMF estimate for the high K_{OW} congener (PCB 194) generated by the uncertainty simulations is more consistent with the empirical data on PCB TMFs (Hoekstra et al. 2003; Walters et al. 2011) which demonstrate a positive correlation between TMFs and PCB hydrophobicity. Although the baseline TMF approximation did not follow this pattern, as described previously, the baseline predictions are strongly influenced by K_{OW} trends in E_D as estimated from the model algorithm. Under realistic perturbations of E_D, the parabolic relationship observed for TMF against chemical K_{OW} is lost.

All simulations indicated that PCB TMFs for the three selected PCB congeners would be greater than 1. For stochastic simulations the probability estimates that either the combined sensitivity or uncertainty simulation would generate a TMF less than the critical value of 1 for any congener is 0. This suggests that even though TMFs are likely to have variation around their mean estimates, the likelihood of generating a false conclusion that the TMF < 1 remains low even for the least and most hydrophobic PCB congeners of study.

The results from deterministic and stochastic multi-zone simulations contrasting simulations with and without fish movement are presented in Figure 6. The multi-zone model deterministic output was similar to the base-line river-wide mean output across all congeners. All zones produced similar estimates of TMFs compared to the river wide baseline value when fish movement was not included in the model simulations. The only difference between zones in these model simulations is concentration of contaminants in the water and the sediment. Hence, these results support the common assumption that changes in background contaminant concentrations do not affect a system's TMF (Broman et al. 1992). The simulations which allow for between zone movement of fish yielded different results. The lower contaminated zones (zones 2, 4, and 6) on the south-eastern side of the Detroit River exhibit higher TMFs than either the river-wide mean output or the multi-zone output. This contrasts with the more contaminated zones (zones

1, 3, 5) on the north western side of the river which have somewhat lower TMFs than the river-wide mean output or the multi-zone output.

Based on the probability distributions generated for TMFs in the simulation series with fish movements, the probability that the TMF of PCB 153 would be over-predicted by one TMF unit over the baseline value when all organisms are sampled exclusively in the clean areas of the river was $p > 0.8$. The probability of over-prediction of TMFs for this congener by two TMF units was $p > 0.6$. When all fish are collected in the contaminated areas of the river, the probability of under-predicting the TMF of PCB 153 by one TMF unit was low at < 0.05. The probability of under-predicting the TMF of PCB 153 by two TMF units was negligible, $p<0$. For the high K_{OW} congener, PCB 194, the probability of over-estimating the TMF by one unit was $p > 0.9$, and by two TMF units was $p > 0.7$ when all samples are collected from the clean regions of the river. When all organisms are collected from the contaminated regions of the river, the probability of under-estimating the TMF by one TMF unit is $p = 0$. The low K_{OW} congener, PCB 31/28, under the fish-movement scenario, generated TMFs that were closer to the baseline value compared to other congeners. For this chemical, the probability of over-estimating the TMF by one TMF unit was $p > 0.55$ and the probability of overestimating the TMF by two TMF units was $p>0.25$ when organisms were sampled in the clean area of the river. When organisms are sampled exclusively at contaminated regions of the river, there is zero probability, $p = 0$, that the TMF will be under-estimated by one or more TMF units.

Model simulations incorporating model sensitivity, uncertainty and fish movement scenarios suggest that TMFs are likely to vary as a result of physiological and ecological attributes of organisms and their differential ability to track spatial contamination patterns within the system. This has implications to the development of appropriate sampling strategies for establishing empirical TMFs in real systems. In the present simulation system, allowing for fish movement contributed to the greatest effect on TMF estimates. In this case, there was a distinct interaction between degree of heterogeneity in water and sediment contamination with organism foraging movements, such that TMFs in clean areas were underestimated, while TMFs in contaminated areas were overestimated compared to riverwide baseline values.

From a regulatory perspective, a TMF greater than 1 has been suggested for use as a criterion to designate a contaminant as capable of undergoing food web biomagnification (Condor, et al. 2011). In present set of simulations, the likelihood of mis-categorizing PCBs as non-biomagnifying chemicals in the Detroit River system was low, even if organisms are exclusively sampled in the most contaminated region of the river where underestimates of TMFs due to sampling artifacts are likely to be maximized. As chemical K_{OW} decreases below that of PCB28/31, however, the chance of mischaracterizing the TMF increases. For instance, a non-biotransformed PCB-like chemical with a hypothetical log K_{OW} of 5.0 is predicted to have a TMF of approximately 1, and simulation results show that the probability of mis-characterizing it as non-biomagnifying would be $p > 0.4$ when samples are exclusively collected in contaminated regions. Moreover, the spread of the TMFs for a hypothetical chemical of K_{OW} 5.0 is ranges from 0.5-2.2. Alternatively, over estimates of TMF can occur as a result of sampling all individuals within a clean zone in the system. While this has limited implications concerning the categorization of PCBs and other well-known biomagnifying chemicals, the observations present the possibility that non-biomagnifying chemicals have a greater chance of being mis-classified as biomagnifying due to sampling artifacts compared to the mis-categorization of biomagnifying chemicals. For example, if a chemical had a true TMF = 0.5, the probability that it would be misclassified as having a TMF = 1 or more is $p < 0.35$ under the fish movement scenario where all organisms are sampled exclusively from clean regions of the river. Furthermore, this could result in gross-overestimations of the hypothetical chemical's TMF as it ranges from 0.3-5.0, where a TMF of 5.0 is usually reserved for the persistent, highly hydrophobic contaminants with $\log K_{OW} > 7.0$ (e.g. Houde et al. 2008; Walters et al. 2011)

Additional simulation trials were performed where the spatial heterogeneity of water and sediment contamination in the system was increased to determine how this could cause further perturbations in TMF estimates. In this case chemical concentrations in water and sediments were increased in the American zones (zones 1, 3, 5) by factors of 2, 10, and 100, while they stayed constant in the Canadian zones (zones 2, 4, 6). Across the above simulations, the minimum TMF values tended to stay relatively constant. In all cases, the chance that TMFs would falsely predict bio-dilution, i.e. TMF<1, remained

slight ($p < 0.01$) when organisms were exclusively sampled in the contaminated regions of the river. The stability of TMFs with increasing perturbation of water and sediment spatial heterogeneity was somewhat surprising and is likely to be a system specific characteristic related to the foraging range coefficients of the different species included in the model. It is possible that systems which demonstrate larger differences in spatial movements between species may show different interactions between TMF and spatial scale heterogeneity of model inputs.

The present results underscores the need to consider the spatial scale of sampling efforts to provide a best estimate of the chemical TMF in a given system. Rather than focusing efforts on collecting organisms from a single area, forage fish, and organisms with small foraging radii should be collected in multiple regions within a system. Ideally, lower trophic level organisms with a smaller foraging footprint should be collected across the foraging range of the most mobile top predator being sampled and included in TMF calculations. These spatial design elements should be adopted in addition to the recommended increase in replication efforts for lower trophic animals suggested by Borgå et al. (2012).

Alternatively, the results from this research support the use of a chemical benchmarking approaches to address potential sampling artifacts associated with TMF estimates of emerging contaminants of concern (Adolfsson-Erici et al. 2012). Although simulations indicate that PCBs will exhibit variation in TMF estimates due to sampling artifacts, the consistently high TMF value of mid-K_{OW} congeners, such as PCB 153, are unlikely to be mis-categorized as non-biomagnifying even under a high degree of sampling bias and system-wide spatial heterogeneity of sediment and water contamination. Hence determination of PCB 153 TMFs in conjunction with those of emergent contaminants of concern could provide a valuable quality control check against sampling artifacts. For instance, by agreeing upon a standard TMF value for each PCB across systems, a TMF correction factor for a range of K_{OW}s could be obtained by comparing the standard values to the observed values in the system in question.

3.4 Conclusion

The model simulations demonstrate that TMFs are much more robust to parameter perturbations then hypothesized previously (Borgå et al. 2012). Food web bioaccumulation models used to estimate TMFs are most sensitive to variation in the assimilation efficiency of chemical from diet and the assimilation of lipids in diet. However, when realistic values for parameter variation are incorporated into stochastic model simulations, chemical assimilation from diet tends to dominate estimates of TMF variability. The importance of dietary food item proportions to TMF variation was relatively minor compared to physiological and toxicokinetic parameters included in the model for low K_{OW} congeners, but had a lot more significance for congeners with $\log K_{OW} > 6.0$. Model uncertainty analysis revealed nearly equal contributions of water and sediment contamination to TMF variability estimates. Finally, spatial scale heterogeneity of water and sediment contamination have a greater potential to modify TMF estimates under conditions where fish movement is allowed to occur between contaminated and clean zones of the system. Although for PCBs, the likelihood of misclassifying these compounds as non-biomagnifying (i.e. TMF < 1) was <1%, the likelihood of misclassifying emerging chemicals whose true TMF approaches the regulatory criteria of 1 becomes greater. These simulations suggest that care in the sampling strategy for collecting organisms should be taken. Ideally, lower trophic level organisms should be sampled from across the foraging range of top predators included in the food web sampling efforts. Alternatively, benchmarking approaches to TMF calculation using well established biomagnifying compounds such as PCBs can be used to evaluate and/or adjust for sampling strategy bias.

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Table 3.1. List of model parameters, descriptions, whether the parameter was allowed to vary in model simulations or held fixed, and values.

Parameter	Parameter Description	Varied or Fixed	Value
C_{O2}	concentration of oxygen in water	Varied	$= -0.24 \times T + 14.04 \times 0.9$
T	Detroit River Temperature	Varied	10.34 ± 8.39
BW	organism body weight	Varied	Table A.3
C_w	concentration of chemical in water	Varied	Table A.1
C_{sed}	concentration of chemical in sediment	Varied	Table A.1
f_{OC}	fraction organic carbon in sediment	Varied	Table A.1
D_{sed}	sediment density	Fixed	1.2
G_d	organism feeding rate	Varied	$= 0.022 \times BW^{0.85} \times e^{0.06 \times T}$
G_v	organism gill ventilation rate	Varied	$= \frac{1400 \times BW^{0.65}}{C_{O_2}}$
E_w	organism's chemical assimilation efficiency for water	Varied	$= a \times \frac{1}{1.857 + \left(\frac{155}{K_{OW}}\right)}$
E_d	organism's chemical assimilation efficiency for food	Varied	$= b \times \frac{1}{0.0000003 \times K_{OW} + 2}$
$p_{(o,w)}$	fraction of respired overlying water	Varied	Table A.2
$p_{(p,w)}$	fraction of respired pore water	Varied	Table A.2
p_i	proportion of diet item i's	Varied	Table A.5
$C_{w(p,w)}$	concentration of contaminant in pore water	Varied	$= \frac{C_{sed}}{K_{OC} \times D_{sed}}$
AE_{lip}	organisms chemical assimilation efficiency for lipid	Varied	Table A.2
AE_{NLOM}	organisms chemical assimilation efficiency for NLOM	Varied	Table A.2
AE_w	organisms chemical assimilation efficiency for water	Varied	Table A.2
$p_{lip,\ diet\ i}$	proportion of lipid in diet item i	Varied	Table A.3
$p_{NLOM,\ diet\ i}$	proportion of NLOM in diet item i	Varied	Table A.3
$p_{w,\ diet\ i}$	proportion of water in diet item i	Fixed	Table A.3
φ_{NLOM}	NLOM partitioning equivalent in the organism compared to octanol	Fixed	0.05

p_{lip}	proportion of lipid in the organism	Varied	Table A.3	
p_{NLOM}	proportion of NLOM in the organism	Varied	Table A.3	
p_w	proportion of water in the organism	Fixed	Table A.3	
K_{OW}	the octanol-water partitioning coefficient	Fixed	Table A.1	
K_{OC}	the organic carbon-water partitioning coefficient	Fixed		$= 0.35 \cdot K_{OW}$

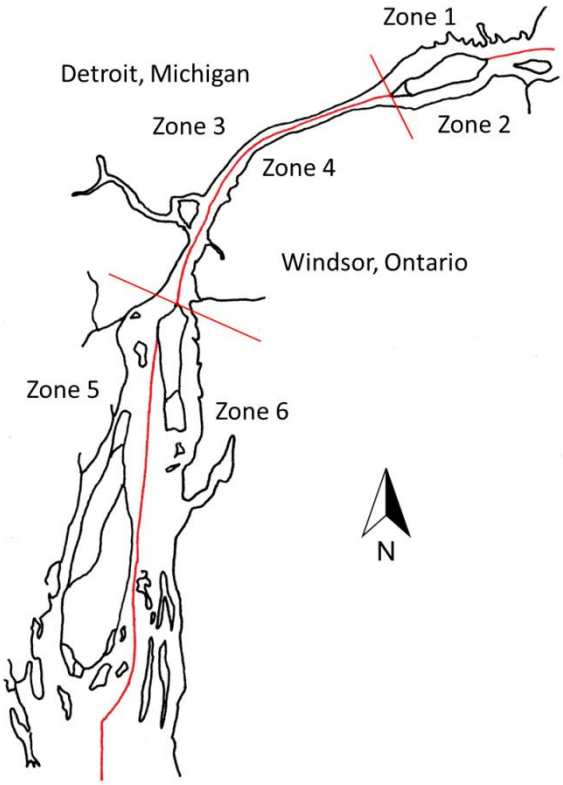


Figure 3.1. Map of the Detroit River divided into the six zones used in the model simulations.

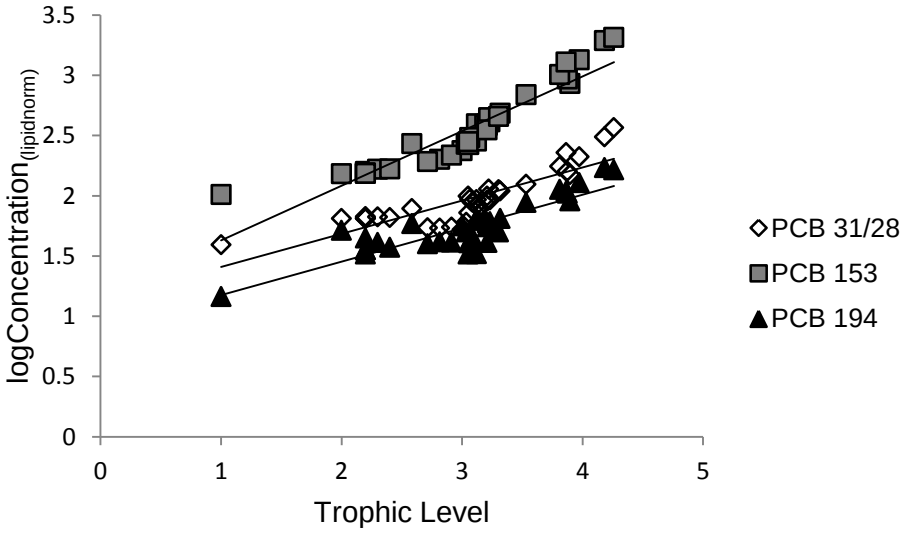


Figure 3.2. Model predictions for lipid-normalized PCB concentrations for a Detroit River food web. Each slope has an R^2 greater than 0.7 (0.7283, 0.8683, and 0.7484 for PCB 31/28, 153, and 194, respectively).

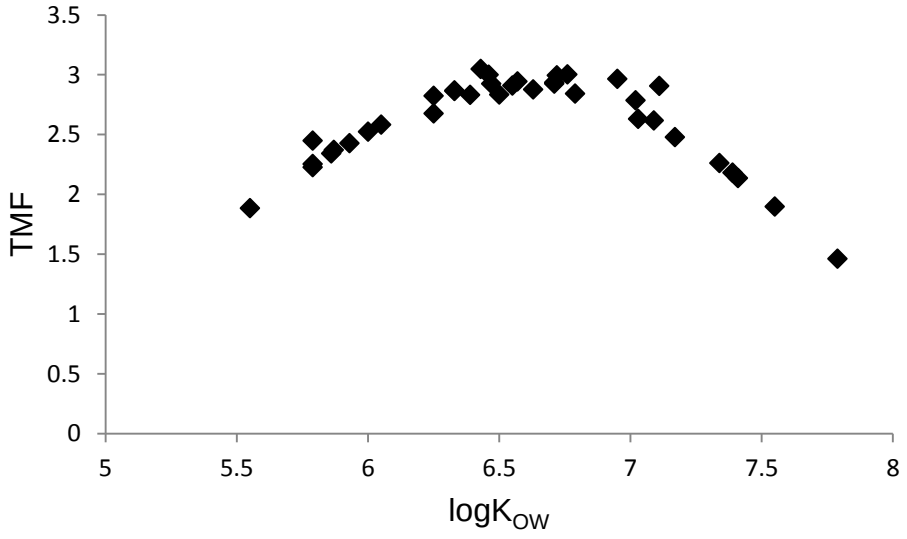
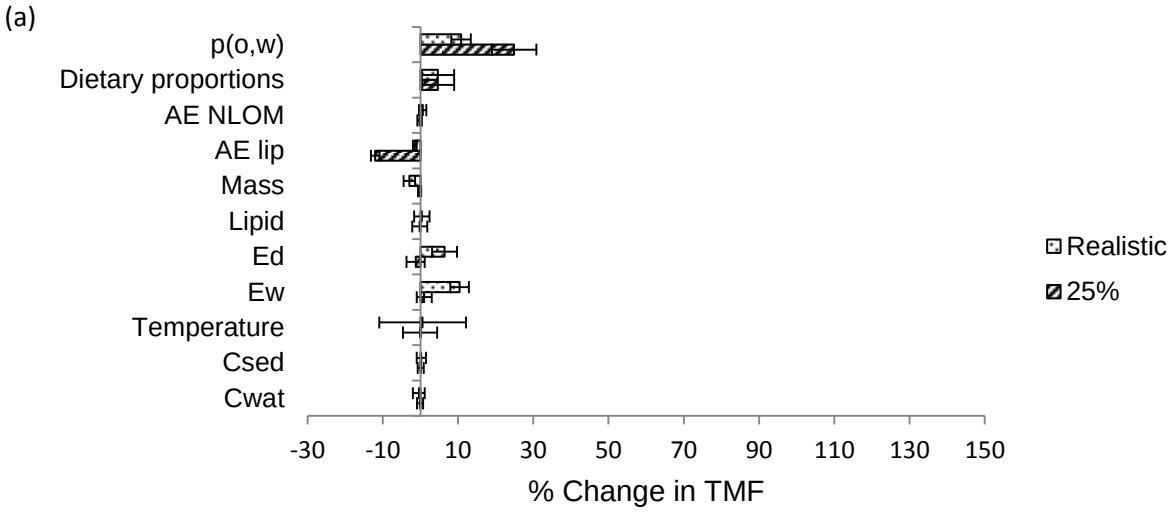
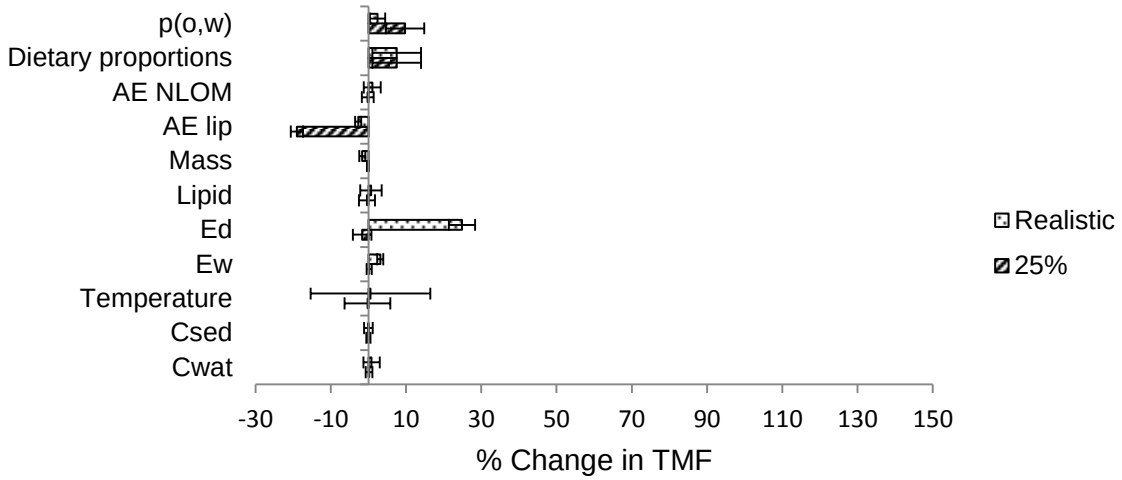


Figure 3.3. Relationship between model predicted TMFs vs. congener log(K_{OW}).



(b)



(c)

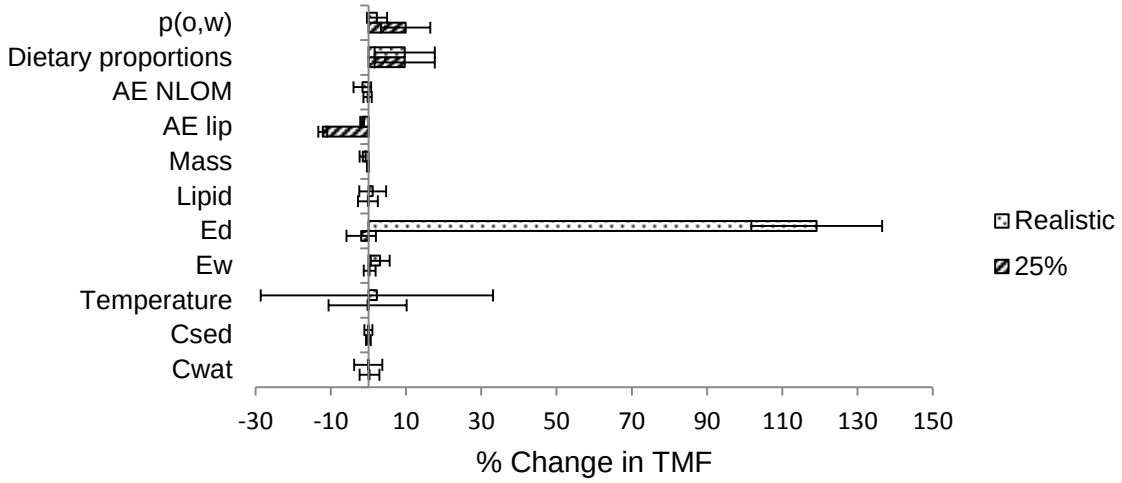


Figure 3.4. Comparison between simulation where chosen parameters varied by ±25%, and one where they varied by realistic amounts for (a) a low K_{OW} PCB (PCB 31/28), (b) a medium K_{OW} congener (PCB 153), and (c) a high K_{OW} congener (PCB 194). Each bar represents the percent increase or decrease in TMF and the error bars represent the corresponding percent standard deviation around those values.

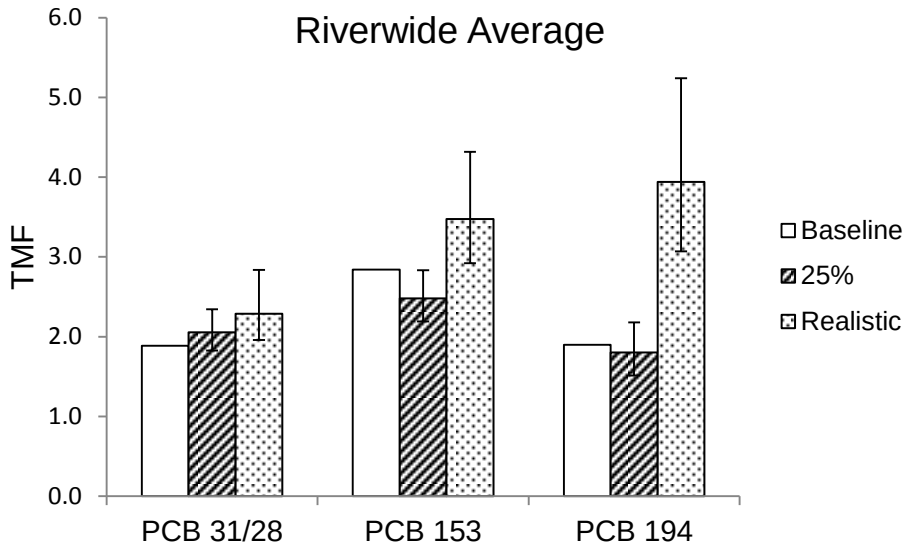
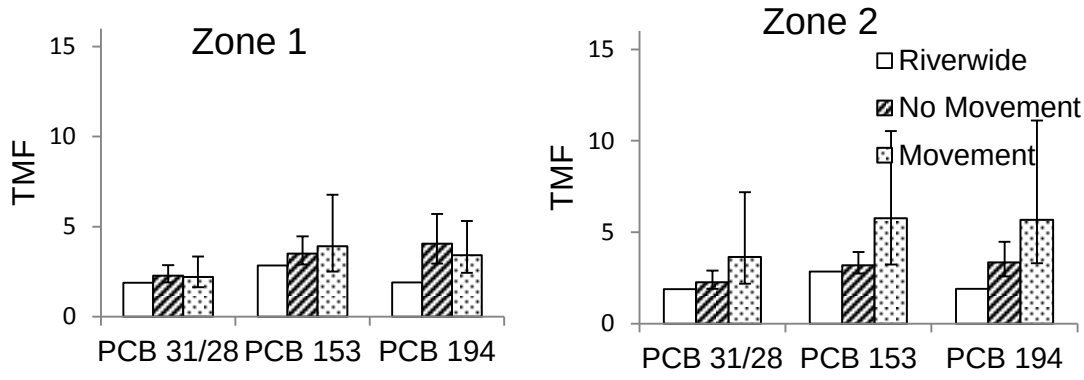


Figure 3.5. Comparison between baseline TMFs, a simulation where chosen parameters were varied by ±25%, and a simulation where chosen parameters were varied by realistic amounts for a low, medium, and high K_{OW} PCB (PCB 31/28, PCB 153, and PCB 194, respectively) for the Detroit River. The error bars represent 95% CIs for the simulations.



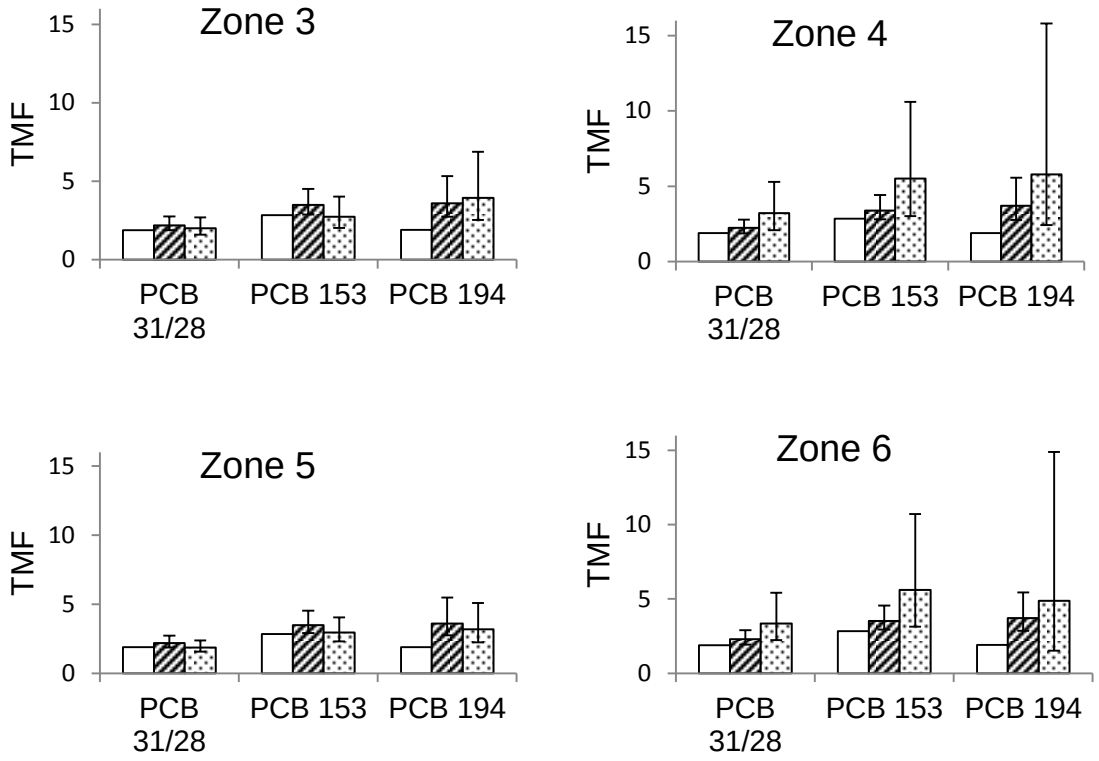


Figure 3.6. Comparison between baseline TMFs for the river-wide average, the deterministic multi-zone model, and the deterministic movement model for low, medium, and high K_{OW} PCBs (PCB 31/28, PCB 153, and PCB 194, respectively) in individual Detroit River zones. The error bars represent 95% CIs for the deterministic simulations.

CHAPTER 4
GENERAL DISCUSSION

5.1 Discussion

Understanding the sources of variability of contaminant concentrations in fish and wildlife is important from an ecological perspective as well as for quantifying hazard and risk assessment of bioaccumulating chemicals. This thesis examined various factors regulating variability in PCB congener concentrations in forage fish and food webs. Specifically, the thesis examined the role of chemical and ecological factors, such as hydrophobicity, species, temporal and spatial variability, food-web level drivers (migration, diet choices, and species specific growth rates) and environmental drivers such as concentrations of PCBs in both sediments and water and temperature. Both statistical assessment of variation derived from empirical measurements in populations of forage fish and food web bioaccumulation modeling tools were used to explore PCB variability. This included causes and consequences of environmental characteristics, species level physiology and ecology and toxicokinetics factors on chemical bioaccumulation, model uncertainty and observed variation in field collected animals.

In my field study, I quantified the relative importance of chemical and ecological drivers on variability of POP body burdens in forage fish species. I focused on three similar sized and aged cyprinid species common in the Detroit River and known to exploit different habitats and diet compositions. These species include the bluntnose minnow (*Pimephales notatus*), a benthic specialist, and two generalist shiner species, the spottail shiner (*Notropis hudsonius*), and emerald shiner (*Notropis atherinoides*) (Starrett, 1950; Keast and Webb, 1966; Scott and Crossman, 1973; Muth and Busch 1989; Johnson and Dropkin, 1993). In particular, I highlighted the importance of feeding strategy and its role in regulating POP concentrations and intraspecific variation in bioaccumulated residues. For example, variability in bioaccumulated PCB residues was lowest for bluntnose minnows (with an average coefficient of variation (CV) of ~ 0.15 for all three classes of congeners), a benthic specialist when compared to the two opportunistic shiner species (with average CVs ranging from 0.2-0.3). Furthermore, this study demonstrated

that not only was hydrophobicity the largest driver of variability in PCB bioaccumulation residues (explaining ~14% of the variability), the other drivers of variability differed in importance once hydrophobicity was accounted for. This underlines the need for scientists and regulators to recognize individual PCBs as unique contaminants instead of as a broad group. While chemical hydrophobicity and feeding ecology have long been recognized as critical factors regulating the magnitude of chemical bioaccumulation by fish (Hebert and Haffner, 1991; Madenjian et al. 1993; Madenjian et al. 1994), this study provided a unique perspective on how the above factors also contribute to enhanced variation in accumulated residues between individuals of the same species. Finally, this study concludes that it is critical to incorporate individual-specific habitat use and feeding ecology into congener specific bioaccumulation models and uncertainty propagation generated from food web bioaccumulation models in order to fully understand the bioaccumulation process.

Modeling is an important tool for both hazard assessment and for extrapolating our knowledge by asking 'what if' questions that are too complicated for an experimental approach. I took the conclusions of the field study about the importance of incorporating individual ecology into congener specific bioaccumulation models and applied these to a food web bioaccumulation model in my third chapter. Here, I used a common hazard assessment metric, the trophic magnification factor (TMF) (Gobas et al. 2009; Weisbrod et al. 2009), and used sensitivity analyses to determine the influence of a wide variety environmental, ecological, and physiological parameters on TMFs. This study further demonstrated that individual PCB congeners have differential degrees of sensitivity to bioaccumulation parameters that are dependent on K_{OW}. For instance, TMFs for mid and high K_{OW} congeners are most sensitive to perturbations in E_d (organism's chemical assimilation efficiency for food), while for the TMFs of low K_{OW} congeners, this has very little effect. The most interesting conclusion from the uncertainty simulations, however, is the influence of spatial heterogeneity and fish movement on TMFs. Spatial heterogeneity and sampling resolution are key drivers of TMFs often causing regulators to under or over-estimate TMFs. In clean areas the foraging of larger predatory fish in neighbouring highly contaminated areas can cause a gross over-estimation of the TMF, resulting in a mis-characterization of a chemical as being one that biomagnifies. The

reverse is also true, where a biomagnifying contaminant in a highly contaminated area gets mis-calculated as attenuating through the foodweb. Although food web bioaccumulation models reflect a commonly used tool in PCB hazard assessments and to assessment environmental mitigation options, this chapter was the first to apply the food web bioaccumulation model to specifically evaluate TMFs under a different set of ecological scenarios (i.e. incorporating variation in diet composition and fish movement). The chapter also provided the first evaluation of model sensitivity and uncertainty of the Arnot and Gobas (2004) model to explore plausible ranges of TMFs in a highly heterogeneous system such as the Detroit River.

Examination of individual variability in POP concentrations is important, especially as the number of novel chemicals being released into the environment is increasing at an alarming rate (Binetti et al. 2008). Not only has intraspecific variability in contaminant body burdens been observed in many studies (Madenjian et al. 1993; Madenjian et al 1994; Lopes et al. 2011; Borgå et al. 2012), it has been addressed by some individual based bioaccumulation models (Madenjian et al. 1993; Madenjian et al 1994). Unfortunately, these models have addressed variability in sum PCB concentrations, despite chemical-specific hydrophobicity being the largest driver in bioaccumulated PCB residue variability. This thesis started out by examining variability at a low trophic level, the forage fish, scaling up to a whole food-web level approach. It begins to answer some questions about the major drivers of variability associated which organic chemical bioaccumulation by fish. More research is needed, however, before this issue can fully be addressed and more accurate bioaccumulation models created. Further experiments include studies to determine inter-specific differences in PCB elimination rates, and the relationship between these and K_{OW}. This is important not just from a modeling perspective to create more accurate bioaccumulation models, but as well from a ecotoxicological perspective. Previous research (Paterson et al. 2007, Drouillard et al. 2009) has suggested relationships between elimination rates and organism metabolism. By examining differential inter and intra specific elimination rates the degree of variability in a species metabolic rate could be determined quantitatively. Also, further experiments should be performed on the variability in assimilation efficiencies among different food items both within and across species. As the sensitivity analyses from

Chapter 3 points out, TMF, as well as bioaccumulation in general (see Selck et al. 2011), are very sensitive to variation in assimilation efficiencies, but the inherent variability in this metric is not yet resolved and limited to a narrow range of laboratory studies (e.g. Liu et al. 2010)

Other factors related to improvements and changes in food web bioaccumulation models can also be considered in the future. For example, there is a growing recognition that non-steady state bioaccumulation predominates in temperate fish as a result of seasonal temperature changes that both alter fish metabolic rates as well as chemical toxicokinetics (Burtnyk et al. 2009; Drouillard et al. 2009). In addition, non-steady state bioaccumulation processes are shown to manifest themselves across different life stages of animals as a result of seasonal and age-related changes in growth (Paterson et al. 2007; Daley et al. In Press) and weight loss (Paterson et al. 2007; Norstrom et al. 2007; Daley et al. 2009; 2011). To date, all food web bioaccumulation models for persistent organic compounds are solved under steady state assumptions. Yet, incorporating non-steady state processes within such a framework will provide more accurate, i.e. age- and size related differences in chemical bioaccumulation, but also provide an important venue for exploring inter-individual differences in organism response to changing environmental conditions between and within years of simulations. Thus, future bioaccumulation modeling efforts should strive to combine attributes of non-steady state population models (Drouillard et al. 2009; Daley et al. In Press) into a food web model construct and further explore how changes in environmental parameters, ecological parameters and physiological characteristics contribute to additional error propagation and variation under more dynamic model scenarios that better reflect the heterogeneous environment that fish inhabit.

5.2 References

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APPENDICES

Appendix A

Table A.1. Parameter values for concentration of contaminant in water (C_w) and sediment (C_{sed}), where each parameter had a lognormal distribution with the standard deviations (SDs) presented below.

	Parameter	Zone	Mean	25 % SD	Realistic SD
PCB 31/28	**C_w**	River-wide	0.0978924	0.0244731	66.03375
($logK_{OW}$ =5.67)		Zone 1	0.110905	0.02772626	104.311
		Zone 2	0.03415323	0.008538308	32.75071
		Zone 3	0.1383108	0.03457771	59.81413
		Zone 4	0.04748052	11.87013	42.8977
		Zone 5	0.2075277	51.88192	124.2109
		Zone 6	0.04897709	12.24427	32.21804
	C_{sed}	River-wide	150523.6	37630.91	248038.9
		Zone 1	218900.9	54725.22	455467.6
		Zone 2	63198.72	15799.68	112603.8
		Zone 3	206103.2	51525.79	379199.1
		Zone 4	72955.01	18238.75	87426.66
		Zone 5	262675.4	65668.85	254206.7
		Zone 6	79308.56	19827.14	199329.3
PCB 153	**C_w**	River-wide	15.93981	3.984953	12.60555
($logK_{OW}$ = 6.92)		Zone 1	16.60719	4.151797	16.49383
		Zone 2	10.32325	2.580814	8.182842
		Zone 3	29.2838	7.320949	24.39329
		Zone 4	7.987657	1.996914	7.979944
		Zone 5	24.77021	6.192552	14.22762
		Zone 6	6.666764	1.666691	4.355774
	C_{sed}	River-wide	220976.9	55244.23	381860.6
		Zone 1	389190.1	97297.52	1030062
		Zone 2	21320.7	5330.174	25050.02

		Zone 3	485191	121297.7	785774.1
		Zone 4	31863.47	7965.868	32190.28
		Zone 5	354107.8	88526.94	345445.9
		Zone 6	44188.45	11047.11	72641.15
PCB 194	$\mathbf{C_W}$	River-wide	0.369997	0.092499	0.393821
($logK_{OW}$ =7.8)		Zone 1	0.410808	0.102702	0.436082
		Zone 2	0.301044	0.075261	0.219835
		Zone 3	0.305716	0.076429	0.201021
		Zone 4	0.331297	0.082824	0.836707
		Zone 5	0.567511	0.141878	0.388305
		Zone 6	0.303604	0.075901	0.280976
	$\mathbf{C_{sed}}$	River-wide	54112.1	13528.03	92065.58
		Zone 1	139657.4	34914.35	348223.4
		Zone 2	8024.241	2006.06	8169.163
		Zone 3	59586.01	14896.5	59012.67
		Zone 4	9648.059	2412.015	12514.44
		Zone 5	99309.41	24827.35	112061
		Zone 6	8447.485	2111.871	12412.87

Table A.2. Assimilation efficiency (AE) and pore and overly water values used in model perturbations.

Parameter	Mean	25 % SD	Realistic SD	Min	Max	Notes
$\mathbf{AE_{Lip,\ invert}}$	75	18.75		40	80	Triangular distribution for Realistic perturbations
$\mathbf{AE_{Lip,\ fish}}$	92	23		80	100	Triangular distribution for Realistic perturbations
$\mathbf{AE_{NLOM,\ sed}}$	30	7.5		0	40	Triangular distribution for Realistic perturbations
$\mathbf{AE_{NLOM,\ invert}}$	75	18.75		40	80	Triangular distribution for Realistic perturbations
$\mathbf{AE_{NLOM,\ fish}}$	60	15		40	80	Triangular distribution for Realistic perturbations
$\mathbf{AE_w}$	25	6.25	6.25			Lognormal distribution for both 25% and Realistic perturbations

$p_{(o,w),\ invert}$	0.95	0.85	1	Triangular distribution for Realistic perturbations
$p_{(o,w),\ fish}$	1	0.85	1	Triangular distribution for Realistic perturbations

Table A.3. Species specific model parameters where the 25% and realistic simulations both had lognormal distributions for both body weight and lipid content parameter selections. The organism moisture content was held constant, and the organism`s non-lipid organic matter content was the difference.

	Body Weight			Lipid Content			% Moisture
	Mean	25 % SD	Realistic SD	Mean	25 % SD	Realistic SD	
Zebra Mussel	0.00011	0.000028	0.00001034	1.3	0.325	0.325	78.7
Caddisfly	0.000044	0.00001	0.0000376	1.7	0.425	0.425	78.3
Oligochaetes	0.000004	0.000001	0.00000376	1	0.25	0.25	79
Chironomids	0.000004	0.000001	0.00000376	1	0.25	0.25	79
Gammerus	0.00001	0.000003	0.0000094	2.1	0.525	0.525	77.9
Mayfly	0.0001	0.000025	0.0000094	2	0.5	0.5	78
Crayfish	0.0018	0.00045	0.001692	1.9	0.475	0.475	78.1
YOY Fish	0.0004	0.0001	0.000376	2.1	0.525	0.525	77.9
Brook Silverside	0.0015	0.000375	0.00141	4.5	1.125	0.934	75.5
Emerald Shiner	0.0025	0.000625	0.00235	4.7	1.175	0.934	75.3
Spottail Shiner	0.002	0.0005	0.00188	4.5	1.125	0.934	75.5
Round Goby	0.0025	0.000625	0.00235	4	1	0.934	76
Alewife	0.05	0.01250	0.047	7.4	1.85	1.931	72.6
Smelt	0.05	0.01250	0.047	4	1	0.934	76
Small White Sucker	0.029	0.00725	0.02726	3.5	0.875	0.934	76.5
Bluegill	0.085	0.02125	0.02555836	4	1	0.934	76
Black Crappie	0.0696	0.0174	0.4903	5.7	1.425	0.252	74.3
Gizzard Shad	0.585	0.14625	0.88	7.2	1.8	1.931	72.8
White Perch	0.187	0.04675	0.129683	5.6	1.4	2.279	74.4
White Bass	0.449	0.11225	0.444275	6.5	1.625	1.119	73.5
Rock Bass	0.249	0.06225	0.233506	5.7	1.425	0.252	74.3
Yellow Perch	0.217	0.05425	0.159683	5.5	1.375	0.335	74.5
Walleye	1.238	0.3095	1.267458	9.5	2.375	0.828	70.5
Smallmouth Bass	0.715	0.17875	0.917586	7.6	1.9	1.682	72.4
Largemouth Bass	1.028	0.257000	1.028	7	1.75	0.4	73
Northern Pike	1.667	0.41675	1.929015	8	2	0.057	72
Gar Pike	0.63	0.1575	0.993254	8	2	3.108	72
Muskellunge	6.597	1.64925	6.712084	11	2.75	1.344	69

Bowfin	1.546	0.3865	1.5982365	11	2.75	1.344	69
Redhorse Sucker	0.6	0.15	0.88	12	3	0.766	68
White Sucker	0.87	0.2175	0.917586	8.7	2.175	3.108	71.3
Carp	2.98	0.745	2.785723	12	3	3.849	68
Freshwater Drum	1.245	0.31125	1.22778	6.5	1.625	5.037	73.5
Brown Bullhead	0.49	0.1225	0.444275	10	2.5	0.220	70
Stonecat	0.673	0.16825	0.672167	10	2.5	4.327	70

Table A.4. Proportion of mean foraging time spent in each zone for each species, for movement model simulations each was log normally distributed with the means presented here ±25% of mean as a standard deviation. The home zone probability was calculated as 1 – sum(probability in other zones). Finally, the fraction organic carbon had a lognormal distribution with standard deviations presented here for the realistic simulations, and ±25% of mean as a standard deviation for the 25% simulations.

	Species	Zone1	Zone 2	Zone 3	Zone 4	Zone 5	Zone 6
	f_{OC}	3.7±1.5	3.4±0.8	3.6±2.4	5.3±1.0	5.3±1.4	4.7±1.8
Zone 1							
	Zebra Mussel	1	0	0	0	0	0
	Caddisfly	1	0	0	0	0	0
	Oligochaetes	1	0	0	0	0	0
	Chironomids	1	0	0	0	0	0
	Gammerus	1	0	0	0	0	0
	Mayfly	1	0	0	0	0	0
	Crayfish	1	0	0	0	0	0
	YOY Fish	1	0	0	0	0	0
	Brook Silverside	0.76	0.16	0.04	0.04	0	0
	Emerald Shiner	0.76	0.16	0.04	0.04	0	0
	Spottail Shiner	0.76	0.16	0.04	0.04	0	0
	Round Goby	0.82	0.12	0.03	0.03	0	0
	Alewife	0.42	0.26	0.12	0.12	0.04	0.04
	Smelt	0.42	0.26	0.12	0.12	0.04	0.04
	Small White Sucker	0.66	0.24	0.06	0.06	0	0

	Bluegill	0.66	0.24	0.06	0.06	0	0
	Black Crappie	0.66	0.24	0.06	0.06	0	0
	Gizzard Shad	0.46	0.26	0.11	0.11	0.03	0.03
	White Perch	0.58	0.28	0.07	0.07	0	0
	White Bass	0.46	0.26	0.11	0.11	0.03	0.03
	Rock Bass	0.58	0.28	0.07	0.07	0	0
	Yellow Perch	0.42	0.26	0.12	0.12	0.04	0.04
	Walleye	0.3	0.24	0.15	0.15	0.08	0.08
	Smallmouth Bass	0.42	0.26	0.12	0.12	0.04	0.04
	Largemouth Bass	0.54	0.28	0.08	0.08	0.01	0.01
	Northern Pike	0.48	0.28	0.1	0.1	0.02	0.02
	Gar Pike	0.58	0.28	0.07	0.07	0	0
	Muskellunge	0.52	0.28	0.09	0.09	0.01	0.01
	Bowfin	0.46	0.26	0.11	0.11	0.03	0.03
	Redhorse Sucker	0.44	0.24	0.12	0.12	0.05	0.05
	White Sucker	0.44	0.24	0.12	0.12	0.05	0.05
	Carp	0.38	0.24	0.13	0.13	0.06	0.06
	Freshwater Drum	0.44	0.24	0.12	0.12	0.05	0.05
	Brown Bullhead	0.48	0.28	0.1	0.1	0.02	0.02
	Stonecat	0.38	0.24	0.13	0.13	0.06	0.06
Zone 2							
	Zebra Mussel	0	1	0	0	0	0
	Caddisfly	0	1	0	0	0	0
	Oligochaetes	0	1	0	0	0	0
	Chironomids	0	1	0	0	0	0
	Gammerus	0	1	0	0	0	0
	Mayfly	0	1	0	0	0	0
	Crayfish	0	1	0	0	0	0
	YOY Fish	0	1	0	0	0	0
	Brook Silverside	0.16	0.76	0.04	0.04	0	0
	Emerald Shiner	0.16	0.76	0.04	0.04	0	0
	Spottail Shiner	0.16	0.76	0.04	0.04	0	0

	Round Goby	0.12	0.82	0.03	0.03	0	0
	Alewife	0.26	0.42	0.12	0.12	0.04	0.04
	Smelt	0.26	0.42	0.12	0.12	0.04	0.04
	Small White Sucker	0.24	0.66	0.06	0.06	0	0
	Bluegill	0.24	0.66	0.06	0.06	0	0
	Black Crappie	0.24	0.66	0.06	0.06	0	0
	Gizzard Shad	0.26	0.42	0.11	0.11	0.03	0.03
	White Perch	0.28	0.58	0.07	0.07	0	0
	White Bass	0.26	0.46	0.11	0.11	0.03	0.03
	Rock Bass	0.28	0.58	0.07	0.07	0	0
	Yellow Perch	0.26	0.42	0.12	0.12	0.04	0.04
	Walleye	0.24	0.3	0.15	0.15	0.08	0.08
	Smallmouth Bass	0.26	0.42	0.12	0.12	0.04	0.04
	Largemouth Bass	0.28	0.54	0.08	0.08	0.01	0.01
	Northern Pike	0.48	0.28	0.1	0.1	0.02	0.02
	Gar Pike	0.28	0.58	0.07	0.07	0	0
	Muskellunge	0.52	0.28	0.09	0.09	0.01	0.01
	Bowfin	0.26	0.46	0.11	0.11	0.03	0.03
	Redhorse Sucker	0.24	0.44	0.12	0.12	0.05	0.05
	White Sucker	0.24	0.44	0.12	0.12	0.05	0.05
	Carp	0.24	0.38	0.13	0.13	0.06	0.06
	Freshwater Drum	0.24	0.44	0.12	0.12	0.05	0.05
	Brown Bullhead	0.28	0.48	0.1	0.1	0.02	0.02
	Stonecat	0.24	0.38	0.13	0.13	0.06	0.06
Zone 3							
	Zebra Mussel	0	0	1	0	0	0
	Caddisfly	0	0	1	0	0	0
	Oligochaetes	0	0	1	0	0	0
	Chironomids	0	0	1	0	0	0
	Gammerus	0	0	1	0	0	0
	Mayfly	0	0	1	0	0	0
	Crayfish	0	0	1	0	0	0

	YOY Fish	0	0	1	0	0	0
	Brook Silverside	0.04	0.04	0.7	0.14	0.04	0.04
	Emerald Shiner	0.04	0.04	0.7	0.14	0.04	0.04
	Spottail Shiner	0.04	0.04	0.7	0.14	0.04	0.04
	Round Goby	0.03	0.03	0.78	0.1	0.03	0.03
	Alewife	0.1	0.1	0.38	0.22	0.1	0.1
	Smelt	0.1	0.1	0.38	0.22	0.1	0.1
	Small White Sucker	0.06	0.06	0.56	0.2	0.06	0.06
	Bluegill	0.06	0.06	0.56	0.2	0.06	0.06
	Black Crappie	0.06	0.06	0.56	0.2	0.06	0.06
	Gizzard Shad	0.09	0.09	0.4	0.24	0.09	0.09
	White Perch	0.07	0.07	0.48	0.24	0.07	0.07
	White Bass	0.09	0.09	0.4	0.24	0.09	0.09
	Rock Bass	0.07	0.07	0.48	0.24	0.07	0.07
	Yellow Perch	0.1	0.1	0.38	0.22	0.1	0.1
	Walleye	0.12	0.12	0.28	0.24	0.12	0.12
	Smallmouth Bass	0.1	0.1	0.38	0.22	0.1	0.1
	Largemouth Bass	0.08	0.08	0.46	0.22	0.08	0.08
	Northern Pike	0.28	0.48	0.1	0.1	0.02	0.02
	Gar Pike	0.07	0.07	0.48	0.24	0.07	0.07
	Muskellunge	0.28	0.52	0.09	0.09	0.01	0.01
	Bowfin	0.09	0.09	0.4	0.24	0.09	0.09
	Redhorse Sucker	0.1	0.1	0.36	0.24	0.1	0.1
	White Sucker	0.1	0.1	0.36	0.24	0.1	0.1
	Carp	0.11	0.11	0.31	0.25	0.11	0.11
	Freshwater Drum	0.1	0.1	0.36	0.24	0.1	0.1
	Brown Bullhead	0.09	0.09	0.42	0.22	0.09	0.09
	Stonecat	0.11	0.11	0.31	0.25	0.11	0.11
Zone 4							
	Zebra Mussel	0	0	0	1	0	0
	Caddisfly	0	0	0	1	0	0
	Oligochaetes	0	0	0	1	0	0

Chironomids	0	0	0	1	0	0
Gammerus	0	0	0	1	0	0
Mayfly	0	0	0	1	0	0
Crayfish	0	0	0	1	0	0
YOY Fish	0	0	0	1	0	0
Brook Silverside	0.04	0.04	0.14	0.7	0.04	0.04
Emerald Shiner	0.04	0.04	0.14	0.7	0.04	0.04
Spottail Shiner	0.04	0.04	0.14	0.7	0.04	0.04
Round Goby	0.03	0.03	0.1	0.78	0.03	0.03
Alewife	0.1	0.1	0.22	0.38	0.1	0.1
Smelt	0.1	0.1	0.22	0.38	0.1	0.1
Small White Sucker	0.06	0.06	0.2	0.56	0.06	0.06
Bluegill	0.06	0.06	0.2	0.56	0.06	0.06
Black Crappie	0.06	0.06	0.2	0.56	0.06	0.06
Gizzard Shad	0.09	0.09	0.24	0.4	0.09	0.09
White Perch	0.07	0.07	0.24	0.48	0.07	0.07
White Bass	0.09	0.09	0.24	0.4	0.09	0.09
Rock Bass	0.07	0.07	0.24	0.48	0.07	0.07
Yellow Perch	0.1	0.1	0.22	0.38	0.1	0.1
Walleye	0.12	0.12	0.24	0.28	0.12	0.12
Smallmouth Bass	0.1	0.1	0.22	0.38	0.1	0.1
Largemouth Bass	0.08	0.08	0.22	0.46	0.08	0.08
Northern Pike	0.48	0.28	0.1	0.1	0.02	0.02
Gar Pike	0.07	0.07	0.24	0.48	0.07	0.07
Muskellunge	0.8	0.08	0.24	0.44	0.08	0.08
Bowfin	0.09	0.09	0.24	0.4	0.09	0.09
Redhorse Sucker	0.1	0.1	0.24	0.36	0.1	0.1
White Sucker	0.1	0.1	0.24	0.36	0.1	0.1
Carp	0.11	0.11	0.25	0.31	0.11	0.11
Freshwater Drum	0.1	0.1	0.24	0.36	0.1	0.1
Brown Bullhead	0.09	0.09	0.22	0.42	0.09	0.09
Stonecat	0.11	0.11	0.25	0.31	0.11	0.11

Zone 5

Zebra Mussel	0	0	0	0	1	0
Caddisfly	0	0	0	0	1	0
Oligochaetes	0	0	0	0	1	0
Chironomids	0	0	0	0	1	0
Gammerus	0	0	0	0	1	0
Mayfly	0	0	0	0	1	0
Crayfish	0	0	0	0	1	0
YOY Fish	0	0	0	0	1	0
Brook Silverside	0	0	0.02	0.02	0.8	0.16
Emerald Shiner	0	0	0.02	0.02	0.8	0.16
Spottail Shiner	0	0	0.02	0.02	0.8	0.16
Round Goby	0	0	0.01	0.01	0.86	0.12
Alewife	0.04	0.04	0.1	0.1	0.44	0.28
Smelt	0.04	0.04	0.1	0.1	0.44	0.28
Small White Sucker	0	0	0.04	0.04	0.68	0.24
Bluegill	0	0	0.04	0.04	0.68	0.24
Black Crappie	0	0	0.04	0.04	0.68	0.24
Gizzard Shad	0.03	0.03	0.09	0.09	0.46	0.3
White Perch	0	0	0.05	0.05	0.62	0.28
White Bass	0.03	0.03	0.09	0.09	0.46	0.3
Rock Bass	0	0	0.05	0.05	0.62	0.28
Yellow Perch	0.04	0.04	0.1	0.1	0.44	0.28
Walleye	0.08	0.08	0.14	0.14	0.34	0.22
Smallmouth Bass	0.04	0.04	0.1	0.1	0.44	0.28
Largemouth Bass	0	0	0.06	0.06	0.58	0.3
Northern Pike	0.02	0.02	0.08	0.08	0.5	0.3
Gar Pike	0	0	0.05	0.05	0.62	0.28
Muskellunge	0.01	0.01	0.07	0.07	0.54	0.3
Bowfin	0.03	0.03	0.09	0.09	0.46	0.3
Redhorse Sucker	0.05	0.05	0.11	0.11	0.44	0.24
White Sucker	0.05	0.05	0.11	0.11	0.44	0.24

	Carp	0.06	0.06	0.12	0.12	0.4	0.24
	Freshwater Drum	0.05	0.05	0.11	0.11	0.44	0.24
	Brown Bullhead	0.02	0.02	0.08	0.08	0.5	0.3
	Stonecat	0.06	0.06	0.12	0.12	0.4	0.24
Zone 6							
	Zebra Mussel	0	0	0	0	0	1
	Caddisfly	0	0	0	0	0	1
	Oligochaetes	0	0	0	0	0	1
	Chironomids	0	0	0	0	0	1
	Gammerus	0	0	0	0	0	1
	Mayfly	0	0	0	0	0	1
	Crayfish	0	0	0	0	0	1
	YOY Fish	0	0	0	0	0	1
	Brook Silverside	0	0	0.02	0.02	0.16	0.8
	Emerald Shiner	0	0	0.02	0.02	0.16	0.8
	Spottail Shiner	0	0	0.02	0.02	0.16	0.8
	Round Goby	0	0	0.01	0.01	0.12	0.86
	Alewife	0.04	0.04	0.1	0.1	0.28	0.44
	Smelt	0.04	0.04	0.1	0.1	0.28	0.44
	Small White Sucker	0	0	0.04	0.04	0.24	0.68
	Bluegill	0	0	0.04	0.04	0.24	0.68
	Black Crappie	0	0	0.04	0.04	0.24	0.68
	Gizzard Shad	0.03	0.03	0.09	0.09	0.3	0.46
	White Perch	0	0	0.05	0.05	0.28	0.62
	White Bass	0.03	0.03	0.09	0.09	0.3	0.46
	Rock Bass	0	0	0.05	0.05	0.28	0.62
	Yellow Perch	0.04	0.04	0.1	0.1	0.28	0.44
	Walleye	0.08	0.08	0.14	0.14	0.22	0.34
	Smallmouth Bass	0.04	0.04	0.1	0.1	0.28	0.44
	Largemouth Bass	0	0	0.06	0.06	0.3	0.58
	Northern Pike	0.02	0.02	0.08	0.08	0.3	0.5
	Gar Pike	0	0	0.05	0.05	0.28	0.62

Muskellunge	0.01	0.01	0.07	0.07	0.3	0.54
Bowfin	0.03	0.03	0.09	0.09	0.3	0.46
Redhorse Sucker	0.05	0.05	0.11	0.11	0.24	0.44
White Sucker	0.05	0.05	0.11	0.11	0.24	0.44
Carp	0.06	0.06	0.12	0.12	0.24	0.4
Freshwater Drum	0.05	0.05	0.11	0.11	0.24	0.44
Brown Bullhead	0.02	0.02	0.08	0.08	0.3	0.5
Stonecat	0.06	0.06	0.12	0.12	0.24	0.4

Table A.5. Species specific diet matrix where each percentage has a lognormal distribution (± 25% of mean) for parameter selections, the numbers in bold were assigned the unaccounted for percentage each model iteration, and BW stands for body weight.

	BW	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V	W	X	Y
D	0.00011	40	**40**	20																						
E	0.00004	**40**	30	30																						
F	0.000004	**60**	20	20																						
G	0.000004	**60**	20	20																						
H	0.00001	**40**	20	40																						
I	0.0001	**60**	20	20																						
J	0.0018	28	25		**35**	1			9	2																
K	0.0004			**100**																						
L	0.0015		20	**72**				8																		
M	0.0025	9		**90**				1																		
N	0.002	2	30	**51**			15	2																		
O	0.0025	3		**75**	12				10																	
P	0.05			**80**		3			7	10																
Q	0.05			**65**		10			10	10		5														
R	0.029	5		**40**		10			25	20																
S	0.085			**40**	5	10	10	10	10	10		5														
T	0.0696			**40**		10			40	10																
U	0.585			**65**	5	5	7.5	7.5	5	5																
V	0.187			**54**	2	3			18	10		10		3												
W	0.449			**35**	2	3			18	20	2	10		5	5											
X	0.249					10		8	5	10	**50**	5	3	3	3	3										
Y	0.217			**40**	25	1	6	6	6	6		7		3												
Z	1.238			10								**50**		5	10	5	5	5	5							5
AA	0.715			5							5	**30**		20	20		10	10								

AB	1.028		5							**25**	10		20	15		10	10	5											
AC	1.667									3		3	3	3	3	3	3	5	10	8	8	12	12	12	**12**					
AD	0.63				3				3	5	5	10	12	**12**	5	5	5	5	5	5		5	5	5	5					
AE	6.597																	5	5	10	10	15	15	15	**25**					
AF	1.546	5								**30**			5	5	5	5	5	5	5	5	5	5	5	5	5					
AG	0.6	5	**25**	10	10	10	15	10	15																					
AH	0.87	5	**50**	10	5	5	5	10	10																					
AI	2.98	10	**25**	10	10	15	15	5	10																					
AJ	1.245	5	15	**20**	5	10	10	15	10		5		5																	
AK	0.49	5	10	10	10	**15**	15	10	15	5	5																			
AL	0.673	5	8	10	10	10	10	10	**15**	5	5		3	3	3			3												

A	Sediment	G	Chironomids	M	Emerald Shiner	S	Bluegill	Y	Yellow Perch
B	Phytoplankton	H	Gammerus	N	Spottail Shiner	T	Black Crappie	Z	Walleye
C	Zooplankton	I	Mayfly	O	Round Goby	U	Gizzard Shad	AA	Smallmouth bass
D	Zebra Mussel	J	Crayfish	P	Alewife	V	White Perch	AB	Largemouth bass
E	Caddisfly	K	YOY Fish	Q	Smelt	W	White Bass	AC	Northern Pike
F	Oligochaetes	L	Brook Silverside	R	Small White Sucker	X	Rock Bass	AD	Gar Pike
AE	Muskellunge	AF	Bowfin	AG	Redhorse Sucker	AH	White Sucker	AI	Carp
AJ	Freshwater Drum	AK	Brown Bullhead	AL	Stonecat				

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