TROPHIC TRANSFER OF CONTAMINANTS IN TREE SWALLOWS (*TACHYCINETA BICOLOR*) NESTING NEAR LAKE CALUMET, ILLINOIS

BY

SUSAN ELAINE GALLO

THESIS

Submitted in partial fulfillment of the requirements
for the degree of Master of Science in Natural Resources and Environmental Sciences
in the Graduate College of the
University of Illinois at Urbana-Champaign, 2011

Urbana, Illinois

Adviser:
Doctor David J. Soucek

ABSTRACT

Tree swallow (*Tachycineta bicolor*) nestlings, eggs, and diet and sediment grab samples were used to quantify risks of exposure to15 trace elements, 31 polychlorinated biphenyl (PCB) congeners, 15 polybrominated diphenyl ether (PBDE) congeners and 13 organochlorine pesticides in the Calumet area of northeastern Illinois, USA. Nesting success and clutch size were measured in tree swallows to determine whether local contaminants reduced tree swallow fitness. Overall nesting success was not reduced when compared among sites and to range averages; 71-90% of clutches started had at least one nestling fledge. Likewise there were no differences among sites in the proportion of eggs that hatched and nestlings that fledged. Generally, contaminant concentrations in the media were considered low or not elevated, although sediment concentrations of cadmium, chromium, and nickel at some sites were higher than the “probable effects concentration” or the “probable effects level” for sediment dwelling organisms, and lead, manganese, and zinc were above the “severe effects levels” at some sites. Calumet nestlings in 2005 were fed between 51 and 64% aquatic insects by mass. Terrestrial insects in the nestling tree swallow diet contained significantly greater concentrations of lead than aquatic insects consumed by the nestling tree swallows. Mean mercury concentrations in nestlings ranged from 0.10 to 0.18 mg/kg dry weight (dw) and egg concentrations ranged from 0.11 to 0.23 mg/kg dw and approximately 5% of the total mercury mass in nestlings came from the eggs. Egg mercury concentrations, which are acquired directly from the mother, were positively correlated with the timing of nesting, and negatively correlated with brood size. Nestlings at Indian Ridge in 2004 and Powderhorn in 2005 accumulated the greatest mass of mercury. Mean sum PCB concentrations in tree swallow eggs ranged from 463 to 830 ng/g wet weight (ww) and from 105 to 208 ng/g ww in nestlings. Egg concentrations contributed approximately 48% of the total PCB mass in nestlings. Nestlings at Big Marsh in both years, and Indian Ridge in 2004 accumulated the greatest mass of PCBs. Nestlings from both Big Marsh and Indian Ridge in 2005 accumulated the most PBDEs, with approximately 21% of the total mass in nestlings coming from the eggs. Mean sum PBDE concentrations in eggs ranged from 47 to 78 ng/g ww and from 20 to 62 ng/g ww in nestlings, and these results appear to be among the first reported PBDE concentrations in tree swallows. Powderhorn had no record of

sediment contamination that was found, however low levels of contaminants were in the sediment and biota there. Tree swallow nestlings accumulated a variety of contaminants from the Calumet sites though their diet, though eggs contributed significant amounts for some compounds like PCBs. Understanding contaminant presence and uptake in wetlands of the Calumet area is particularly useful due to the loss of wetland habitat in this region.

ACKNOWLEDGMENTS

This study was funded by the Illinois Sustainable Technology Center, a Division of the Prairie Research Institute at the University of Illinois at Urbana-Champaign. John Scott, Gary Bordson, Luann Wiedenmann, Joan Esarey, Amy Dickinson, Marshall McDaniel, Molly Tranel, Nina Hill, Guillermo Arce, John Shaw, Heidi Garbe and Erik Chatroop provided technical assistance. Much thanks to my advisor, David Soucek, for patient mentoring and for rapidly reading countless drafts. Thanks also to my committee members, Jeff Levengood and Walter Hill, for their guidance. Lastly much thanks to my husband, parents, sister, and friends who quietly supported me through this long process.

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CHAPTER 1

LITERATURE REVIEW

Tree Swallow Natural History

Tree swallows (*Tachycineta bicolor*) are mid-level predators that are often used to assess contaminant exposure via trophic transfer from aquatic systems to terrestrial systems (Table 1.1). Tree swallows have a large breeding range in North America extending from Tennessee to California and north to Labrador and Alaska. They are common near water, provided there is structure for roosting and nesting (National Geographic Society, 1987). They possess a number of qualities that make them particularly easy to use in ecological studies: they readily exploit nest boxes (Robertson et al., 1992), are tolerant of disturbances, and are of a manageable size for easy handling. Not surprisingly, they have long been used in field research studies, and there is a tremendous body of knowledge about them (Robertson et al., 1992; McCarty 2002). They are sometimes referred to as the 'avian equivalent of the white rat' of avian ecology (Jones 2003, pg 596). Because tree swallows are so well studied, and easy to work with, they have frequently been used as biomonitors (Table 1.1).

Tree swallows have historically arrived at nesting sites in northern Illinois in mid April (Graber et al., 1972) with males arriving up to a week prior to the females to defend a nest site (Robertson et al., 1992) and yearling swallows arriving later than others (Robertson et al., 1992). Females generally lay one egg per day (Robertson et al., 1992; Cornell University, 2004), and their total clutch size is usually between four and seven eggs (Cornell University, 2004; Gallo personal observation). Egg mass averages 1.9 g but varies with clutch size (Robertson et al., 1992), and increases with laying order (Wiggins, 1990; Robertson et al., 1992). Eggs are typically incubated for 11 to 19 days, with most requiring only 14 or 15 days (Robertson et al., 1992).

When nesting, tree swallows are primarily aerial insectivores and typically feed in open areas 0-50 m above ground (Robertson et al., 1992) and within 300 m of their nesting sites (Quinney and Ankney, 1985; Dunn and Hannon, 1992; McCarty 2002), but have been observed to fly over 100 km to forage (Robertson et al., 1992). Both parents feed the nestlings and the diet typically consists of insects caught on the wing (Robertson et al., 1992). Nestling diet varies with site and insect availability, but swallows appear to be selective of prey size that they feed to their young

(Quinney and Ankney 1985). Holroyd (1983) found that swallows at sites in southern Ontario fed their young primarily nematocerid flies (60-97% of the proportion of their daily diet). Elsewhere, dipterans have been found to comprise from 46 to 94% of tree swallow diets, but odonates (up to 87%) and homopterans (up to 26%) have been observed to be important as well (Blancher et al., cited in Robertson et al., 1992; McCarty and Winkler, 1991; Menglekoch et al., 2004; Quinney and Ankney, 1985). Aquatic insects are often the majority of the diet (Holroyd, 1983; Quinney and Ankney, 1985; Menglekoch et al., 2004; Neigh et al., 2006c) but this varies with habitat (McCarty, 2001; Johnson and Lombardo, 2000) and insect availability (Smits et al., 2005).

Nestlings fledge between 15 to 25 days after hatching and nesting success (defined as at least one nestling departing the nest) is almost 79% (Robertson et al., 1992). Nesting success has been observed to be up to 14% lower for later nesting birds (Robertson et al., 1992). Tree swallows winter in the southern United States, Mexico and Central America (Cornell University, 2004). They have been documented to live up to 11 years, but the average is 2.7 years (Robertson et al., 1992). Estimates indicate that only about 20% of fledged swallows survive the first year of life (Robertson et al., 1992).

The use of tree swallows to assess contaminant exposure started in the 1980s, and became common in the late 1990s (Table 1.1). A number of studies have clearly demonstrated that tree swallow nestlings accumulate contamination from local sediment (Bishop et al., 1995; Froese et al., 1998; Dods et al., 2005; Maul et al., 2006) or soil (Smits et al., 2001; Maul et al., 2006) through the insects they consume. Tree swallows are migratory, and because of slow depuration times in maternal tissues and subsequent deposition in the egg, there is uncertainty regarding the extent to which contaminant burdens in eggs and nestlings can be attributed to a particular locality (Custer and Custer, 1995; Dauwe et al., 2006; Maul et al., 2010). To overcome these concerns, some studies use both nestlings and eggs to calculate local uptake rates (e.g. Ankley et al., 1993; Custer et al., 1998; Custer et al., 2000), or to normalize nestling concentrations by subtracting egg contributions from the total nestling contaminant mass (this study; Maul et al., 2010).

There is growth dilution from a two gram egg to a 20 gram nestling and the proportion of maternally derived contaminant mass in nestlings will vary. One study found eggs contributed between 2.4 to 23.9% of the mercury found in tree swallow nestlings (Longcore et al., 2007a). PCB contribution from eggs was 14.3 to 16.2% of the nestling mass (Maul et al., 2010) at a highly contaminated site in southern Illinois.

In addition to documenting exposure, tree swallows have been used to investigate effects of contamination using various endpoints including reproduction (e.g. Froese et al., 1998; Wayland, 1998; McCarty and Secord, 1999b; Harris and Elliott, 2000; Neigh et al., 2006a), development (e.g. McCarty and Secord, 1999b; Yorks, 1999; Longcore et al., 2007) parental care (e.g. Bishop et al., 2000), genetic mutation rates (Stapleton et al., 2001), histopathological changes (Bishop et al., 1998; Yorks, 1999; Gentes et al., 2007), and immunological response (Bishop et al., 1998b; Smits, 2000; Mayne, 2004; Dods, 2005; Franceschini et al., 2009; Hawley et al., 2009).

For the purpose of this review, studies are separated by the primary contaminants that were assessed, and only results relevant to tree swallows are discussed. Trace Element results are listed in parts per billion (ppb) dry weight (dw) and organic compound results are in ppb wet weight (ww) unless otherwise indicated. This presentation facilitates comparisons with the literature. Feather concentrations are listed in the form of the original study as no trustworthy conversion was found, (Custer et al., 2007a listed a moisture content of 46 to 77%, but they attribute this large range to condensation due to thawing). All other media concentrations were transformed using values from the literature. (liver: 70% moisture from Erry et al., 1999; Adrian, 1979; Franson, 1984; for other species; Blood: 80% Santolo et al., 1999) or using values from this study (nestlings: 30.8% moisture and 8.5% lipid, eggs: 82.7% moisture and 5.4% lipid).

Trace elements

Tree swallow eggs and nestlings have been demonstrated to contain various toxic and non-essential trace elements (Kraus, 1989; Gerrard and St Louis, 2001; Custer et al., 2001; Custer et al., 2003; Custer et al., 2005; Tsipoura et al., 2007; Longcore et al., 2007a). Most studies evaluate multiple contaminants, fewer examine metals exclusively, and fewer still analyze only one trace element, usually mercury (Hg) (Table 1.2).

Body burdens

Tsipoura and colleagues (2007) measured 5 metals in bird blood, feathers and eggs in the Hackensack Meadowlands of New Jersey in 2006, furthering work in that region by Kraus (1989). They found relatively high Cr concentrations in blood, whereas Cr feather concentrations were lower than those found in other birds. Blood lead levels were higher than the 2,000 ppb dw (400 ppb ww) that may have adverse physiological effects in birds (Tsipoura et al., 2007), and one tree swallow had feather lead concentrations above the adverse effects threshold of 4,000 ppb dw (Tsipoura et al., 2007). Lastly, the mercury concentrations in feathers were similar to those of birds from higher trophic levels, but blood concentrations were lower than other species, likely due to the depuration to the feathers. Other studies found trace element concentrations that were similar to background concentrations (Bishop, 1995; Custer et al., 2005), were equal to reference site values (Custer et al., 2001), or were considered to be “low” (Custer et al., 2003).

Gerrard and St. Louis (2001) assessed bioaccumulation of methylmercury (MeHg) in tree swallow eggs, nestlings and adults before and after the creation of a reservoir. Along with increased concentrations of MeHg in swallow nestlings and feathers after flooding, they found a 1:1 ratio of MeHg to Total Hg (THg) in all nestling tissue concentrations. Two other studies evaluated Hg concentrations before and after hydrologic drawdowns and found no differences in mercury concentrations relative to water level (Custer et al., 2006; Custer et al., 2007d).

Mercury concentrations in tree swallow eggs, nestlings, feathers and diet, from Maine and Massachusetts were measured as were its effects on growth (Longcore et al., 2007a). The researchers found a lower rate of mass gain for 2- to10-day-old swallows with higher feather concentrations of mercury. They suggested, based on as yet unpublished injection studies by Heinz, that tree swallow embryos may be more sensitive to Hg than are mallards, stating that Heinz observed effects at “somewhat less than 1 ppm ww” (approximately 5,780 ppb dw) (Longcore et al., 2007a pg 138). Eleven clutches had an egg with concentrations of THg that exceeded the threshold of 4,624-5,780 ppb dw for embryo toxicity, and four of these 11 nests had other eggs that failed to hatch (Longcore et al., 2007a). Nonetheless, their study sites had rates of hatch success above 89%. Additionally, a higher maximum concentration of 2,000 ppb ww (11,560 dw) is currently listed for the range of impaired reproduction for some bird species (Eisler, 2000). A different study around the Carson River in Nevada (Custer et al., 2007b) yielded no impact on hatch success at egg concentrations in the 4,624-5,780 ppb dw range.

In an attempt to determine mercury concentrations in nestlings that likely came from the egg rather than the nestling diet, one study calculated that the egg contributed between 2.4 ± 0.9 and 23.9 ± 3.8 percent of the 14-day-old nestling burden of mercury (Longcore et al., 2007b). The authors calculated that these nestlings transferred between 80 to 92% of their Hg body burden to their growing feathers (Longcore et al., 2007b). A similar finding in adult Bonaparte's gulls (*Larus Philadelphia*) determined that new feathers contained about 93% of the mercury body burden, after completion of molt (Braune & Gaskin 1987).

Effects

Bioindicator response and reproductive success were measured relative to trace element concentrations in tree swallows in Minnesota, before and after a wetland drawdown (Custer et al., 2006). These authors found low concentrations of elements, and observed no differences among sites or years in bioindicator responses or nesting success. Similarly, no significant differences in bioindicator response or hatching success were observed in a second study measuring a drawdown along the Mississippi River (Custer et al 2007d), though trace element concentrations were found to be low. Nesting success was not reduced with maximum average mercury mass in fresh eggs of 1,280 ng, nor with maximum average mercury mass of 13,590 ng in whole nestlings (Longcore et al., 2007a). There were a number of unhatched eggs with mercury concentrations over 1 ppm ww, but either these were part of successful clutches, or the sample size was too small to detect reduced success (Longcore et al., 2007a). Mercury concentrations in feathers were associated with reduced weight gain in 2-10 day old nestlings (Longcore et al., 2007b).

Elevated mercury concentrations in tree swallows have been correlated with immunological responses. Higher blood mercury concentrations were associated with higher baseline corticosterone levels in nestlings (Franceschini et al., 2009), and lower mitogen-induced swelling response to phytohaemagglutinin in females (Hawley et al., 2009). Neither study was able to draw conclusions as to the impact of these responses on tree swallow survival. A recent study utilized mark-recapture techniques and modeled survival relative to THg concentrations, and determined that adult tree swallows with blood mercury concentrations approximately 2,600 ppb greater than the reference swallows had approximately 1% reduced survival (Hallinger et al., 2011).

Organic contaminants

Tree swallows have often been used to assess exposure to organic contaminants (Table 1.1), but similar to elemental studies, most organics studies evaluate multiple contaminants. Polychlorinated biphenyls (PCBs) were assessed in the bulk of studies investigating organic contaminants in tree swallows. Many studies evaluated dioxins, furans or pesticides, and a few measured polycyclic aromatic hydrocarbons (PAHs), aliphatic hydrocarbons (ALHs), oil sand reclamation products and contaminants from pulp mills or wastewater treatment plant effluents. Most studies documented tissue burdens in tree swallow eggs or nestlings; fewer evaluated effects of exposure. Such studies assessed effects on reproductive success, (e.g. Custer, 1998; Smits et al., 2000; Neigh et al., 2006a), behavior (McCarty and Secord, 1999a; Bishop et al., 2000), expression of endocrine disruption by aberrant plumage color (McCarty and Secord, 2000) changed biomarker status (e.g. Burgess et al., 1999; Dods, 2005; Custer, 2006), reduced immunological function (Bishop et al., 1998a) or histological changes (Bishop et al., 1998b). Other studies have generated contaminant uptake models (Nichols et al., 1995; Nichols et al., 2004).

PCBs

The Hudson River, below Hudson Falls, New York is the location of the highest recorded concentrations of PBCs yet found in tree swallow eggs. Concentrations of the sum of over 116 PCBs were between 9,000 and 24,000 ppb ww in eggs and between 32,000 and 96,000 ppb in 15-day-old nestlings (Echols et al., 2004). Further studies along the Hudson River determined there were no adverse reproductive effects at these concentrations (McCarty and Secord, 1999b), but suggested that there may be endocrine disruption as observed through altered nest building behavior (McCarty and Secord, 1999a), and plumage maturation (McCarty and Secord, 2000). Total PCB concentrations of 100,880 and 44,660 ppb for over 90 congeners in tree swallow pipers (egg surrogates) and 12-day-old nestlings, respectively, were found along the Housatonic River in Massachusetts (Custer et al., 2003). The concentrations found in the Housatonic nestlings are the highest recorded in tree swallows. There was a negative relationship between total PCBs and hatching success for two of the three years of the Housatonic study (Custer et al., 2003), though no correlation was found between adult survival and egg PCB concentration (Custer et al., 2007). Studies in other locations evaluating PCBs found highest means in eggs or pipers ranging between 180 (Elliott et al., 1994) to 29,500 ppb (cited in Secord and McCarty, 1999b) and between 4.3 (Neigh et al., 2006b) and 96,000 ppb for nestlings (Echols et al., 2004; Table 1.3).

Uptake rates of PCBs were used to prompt a re-evaluation of the remediation process of a superfund site in southern Illinois. A recent study of nestling tree swallow PCB accumulation in the Sangamo National Priorities List Site (SNPLS) found dilution rates in the SNPLS chicks ranged from 92.4 to -203 ng/g/d (Spears et al., 2008). At SNPLS, diet appeared to be the main contributor to overall PCB uptake (Maul et al., 2006). A similar finding from a different study suggested that tissue concentrations of PCBs were more closely related to prey consumed than local sediment or soil contamination (Smits et al., 2005).

Organochlorine pesticides

Many studies measured concentrations of multiple organochlorine (OC) pesticides (eg. see Table 1.1). The compounds DDE and DDD are consistently detected most frequently and in the highest concentrations in these studies. Approximately five years after DDT was banned for general use in North America, a study in Alberta, Canada analyzed tree swallow eggs and nestlings for eight OC pesticides to determine whether the contaminants originated from the natal area (Shaw, 1983). Concentrations of DDE were between 860 and 2,230 ppb in eggs and nestling carcasses contained between 1.06 and 21.9 μg DDE (Shaw, 1983). The author concluded that some of the OC pesticide and PCB burdens were obtained close to the natal area (Shaw, 1983). Studies decades later are finding residues in southern Ontario nestlings up to 16 ppb DDT, 309 ppb DDE and 8 ppb DDD (Smits et al., 2005). The authors determined that sum DDT tissue residues were positively correlated with terrestrial prey (and PCBs were correlated with aquatic prey) (Smits et al., 2005). Tree swallow eggs in southern Ontario orchards contained a mean of 1,140 ppb *p,p'*-DDE, even though no applications of its parent product had occurred for decades (Mayne et al., 2005). Lastly, a study examining 11 OCs in tree swallows nesting in orchards in British Columbia found DDE and DDD concentrations in eggs of up to 11,200 ppb and 749 ppb, respectively (Elliott et al., 1994). There was a high DDE to DDT ratio, implying that DDT in the tree swallow's food web was breaking down and there were no new sources of it.

Concentrations of OCs were not elevated in tree swallows from the Great Lakes and St. Lawrence River basin (Bishop et al., 2000), or pool 15 of the Mississippi River (Custer et al., 2000). Likewise, OCs were either not detected or present at low concentrations in eggs and nestlings of tree swallows nesting along the Housatonic River (Custer et al., 2003). Ten OC pesticides were detected in 16-day-old tree swallows collected from nests near wastewater treatment plants around Vancouver, British Columbia (Dods et al., 2005). They also found heavier livers in nestlings below the wastewater outflows, but attributed this to 4-nonylphenol rather than OCs and other contaminants (Dods et al., 2005). Tree swallow nestlings from old farm land in southern Ontario had low dieldrin residues of up to 18 ppb (Smits et al., 2005).

Organophosphates, carbamates, pyrethroids and other non-persistent pesticides commonly used in apple orchards were examined for effects on thyroid function in conjunction with DDE (Mayne et al., 2005). Tree swallow eggs contained a mean of 1,140 ppb *p,p'*-DDE, and there were higher plasma T_4 concentrations and thyroid follicular epithelial cell height in tree swallow nestlings from sprayed sites. The study also documented changes in thyroid physiology associated with greater applications of non-persistent pesticides, though the long-term significance of this is unknown. A number of studies evaluated pesticides with respect to behavioral, immunological, and histopathological effects (eg Bishop et al., 2000, Bishop et al., 1998a, Bishop et al., 1998b). They found that after cholinesterase (ChE)-inhibiting insecticide applications, parental visits were reduced in a manner that is not consistent with reduced food availability but rather with ChE inhibition. A separate study found that the use of four ChE-inhibiting pesticides significantly reduced mean plasma ChE levels by 41% within 12 hours of a second application of specific pesticides, but did not detect effects on survival (Burgess et al., 1999).

Dioxins and furans

Most studies with dioxins and furans merely document the concentrations found in the biota (Table 1.4). Moreover, almost all include analyses of other contaminants so any effects information must be interpreted as the result of mixtures.

Tree swallow exposure to and uptake of PCBs, dioxins and furans was demonstrated in the Green Bay area of Wisconsin, and the need to use both eggs and nestlings to demonstrate local uptake was highlighted (Ankley et al.,1993). Tree swallows nesting near pool 15 along the Mississippi River acquired 2,3,7,8-TCDD and 2,3,7,8 TCDF in their eggs (Custer et al., 2000). The TCDD concentrations were 2 to 5 times higher than those found in Green Bay, WI (Ankley et al., 1993) (Table 1.4). Custer et al., (2002) calculated dioxin TEQs and accumulation rates in another exposure study along the Wisconsin River as well.

Total dioxin/furans and their TEQs were significant variables in separate models explaining hatch success in tree swallows along the Housatonic River (Custer et al., 2003). Tree swallows nesting along the Woonasquatucket River (Custer et al., 2005) produced some of the highest avian tissue TCDD concentrations yet reported (Table 1.4) and hatching success was negatively associated with these concentrations in eggs. Additionally, along the Fraser and Thompson Rivers, British Columbia, lower nesting success was observed downstream from the pulp and paper mills than upstream; however the difference was thought to be caused primarily by nest abandonment and the study was unable to link this to effluent exposure (Harris and Elliott, 2000). Martinovic et al., (2003b) found total PCDF concentrations in 16 day old nestlings negatively correlated with basal corticosterone levels, and hypothesized that these concentrations may be interfering with the glucocorticoid component of the endocrine system in tree swallows, and may contribute to an inability to physiologically respond to stress.

Conclusions

Tree swallows have been used extensively as biomonitors to assess exposure to or effects of many environmental contaminants including mercury, PCBs, pesticides and dioxins and other contaminants not detailed here like PAHs (e.g. Custer et al., 2001, Custer et al., 2003), reclaimed oil sand mine residues (Smits et al., 2000) and radiation (reviewed in McCarty, 2002). Because tree swallows are predominantly aerial insectivores, they appear to be excellent surrogates for swifts, nighthawks, other swallow species and possibly even some bats, but care should be taken with extrapolating their uptake to birds with other foraging methods. Exposure studies clearly document that tree swallow nestlings accumulate contaminants on site (e.g. Ankley et al., 1993); however, the exclusive use of eggs or nestlings to document uptake is problematic. Moreover, if scientists hope to draw conclusions about contaminant transfer from aquatic to terrestrial systems with tree swallows, it is important to clearly understand the diet of the swallows at the study site as there is much variation in the proportion of aquatic to terrestrial insects from different locations (e.g. 95% aquatic by mass (Menglekoch et al., 2004) to 85% terrestrial by count (Johnson and Lombardo 2000). The number of studies evaluating toxicological effects on tree swallows is growing; nonetheless, our understanding of behavioral endpoints, population effects and indirect effects is still in its infancy. Lastly, more toxicokinetic and experimental studies should be conducted to better understand the uptake and fate of contaminants in tree swallows and, indeed to learn how tree swallows function with exposure to specific concentrations and combinations of contaminants.

Table 1.1. List of published tree swallow (*Tachycineta bicolor*) ecotoxicological studies. Abbreviations are in appendix 1.

Author	#	Location	Tissue analyzed	Contaminant and/or endpoint studied
Ankley et al., 1993	1	Lower Fox R and Green Bay WI	Eggs, nestlings	PCBs; PCDFs; PCDDs; TCDD-EQ
Bishop et al., 1995	2	Great Lakes St. Lawrence River Basin (GLSLRB)	Eggs nestlings	CHCs; Hg
Bishop et al., 1998a	3	Ontario	Eggs, nestlings	OC pesticides; PCBs; Immunological parameters; EROD; Histopathology
Bishop et al., 1998b	4	Ontario	Nestlings, adult blood	OC pesticides; PCBs; histological; hormone
Bishop et al., 1999	5	GLSTRB	Eggs, nestlings	OC pesticides, PCBs; productivity; biochemical indicators; vitamin A; EROD
Bishop et al., 2000	6	Ontario	Nestling, adult (behavior)	ChE inhibiting insecticides; nestling calling and mass; adult behavior
Brasso et al., 2008	7	Shenandoah R. headwaters, VA	Female blood, feather	Accumulation and effects of Hg found reduced fledge success for year old females at contaminated site
Brasso et al., 2010	8	Shenandoah R. headwaters, VA	Eggs	Hg and laying order
Burgess et al., 1999	9	Ontario	Nestlings, adult blood	OP pesticides; ChE inhibition
Custer et al., 1998	10	Fox R. and Green Bay, WI	Pipers, nestlings	PCBs; DDE; cytochrome P450; EROD; BROD
Custer et al., 2000	11	Pool 15 Mississippi R., IA	Eggs, nestlings	PCBs; OC pesticides; TCDD; TCDF
Custer et al., 2001	12	North Platte R., WY	Eggs, nestlings	PAHs; ALHs; 18 trace elements; Hg; monooxygenase
Custer et al., 2002	13	Wisconsin R., WI	Eggs, nestlings	PCBs; dioxins
Custer et al., 2003	14	Housatonic R., MA	Pipers, nestlings	90+ PCBs; 16 dioxins and Furans; 26 OC pesticides; 39 PAHs; 27 ALHs; 19 trace elements

Table 1.1. Cont.

Custer et al., 2005	15	Woonasquatucket R., RI	Eggs, nestlings	TCDD; 10 PCDFs; 7PCDDs; 107 PCBs; 21 trace elements; EROD; HPCV;
Custer & Read, 2006	16	Housatonic R., MA	Pipers, nestlings	PCBs
Custer et al., 2006	17	Agassiz NWR, MN	Eggs, nestling livers	Oxidative stress; genetic damage; 19 trace elements
Custer et al., 2007b	18	Carson R., NV	Eggs, nestling livers	Hg; 18 other trace elements
Custer et al., 2007c	19	Housatonic R., MA	Eggs, adults	PCBs; 21 OCs; Survival in adults
Custer et al., 2007d	20	Pool 8, Mississippi R., WI	Eggs, Nestling liver, carcass	Total PCBs, 24 OC pesticides, Hg, 18 trace elements; biomarkers
Custer et al., 2008	21	Lostwood NWR, ND	Nestling liver and carcass	Hg and 18 other elements, oxidative stress and reproductive success
Custer et al., 2009	22	Summit Co., CO	Nestling kidney, liver	31 metals and metalloids
Custer et al., 2010a	24	Hudson R., NY	Eggs	160 PCB congeners, 17 PCDD-Fs, 28 OC pesticides
Custer et al., 2010b	23	Housatonic R., Berkshire Co., MA	Eggs	Egg order for 21 OCs, total PCBs, Hg and 18 other elements
DeWeese et al., 1985	25	Colorado	Eggs, adult females (brain and carcass)	OC pesticides; DDE; PCBs
Dewitt et al., 2006	26	Monroe Co., IN	Nestlings	PCB and external heart morphology
Dods et al., 2005	27	Vancouver, BC	Nestling livers and carcass	4-nonylphenol; 56 PCBs; 21 OC pesticides; immune status; body composition
Echols et al., 2004	28	Hudson R., NY	Eggs, nestlings, adults	PCBs
Elliott et al., 1994	29	Okanagan Valley BC	Eggs	OC pesticides; PCBs
Franceschini et al., 2009	30	SE ME, and Eastern MA	Nestlings, adults	Baseline and stress induced plasma corticosterone; Hg
Froese et al., 1998	31	Saginaw Bay, MI	Eggs, nestlings	110 PCBs

Table 1.1. Cont.

Gentes et al., 2007	32	Poplar Creek Reservoir, AB	Nestlings	Naphthenic acids (Nas), histopathology, biomarkers; Dosing study
Gerrard and St Louis, 2001	33	Experimental Lakes Area Research Project, NW ON	Eggs, nestlings	MeHg
Hallinger et al., 2011	34	South River, VA	Adults	Hg, survival
Harris and Elliott, 2000	35	Fraser and Thompson Rivers, BC	Nestlings	PCBs; PCDDs; PCDFs; pesticides; chlorophenols; chloroguaiacols
Hawley et al., 2009	36	South River, VA	Adult females	Mitogen induced swelling in response to phytohaemagglutinin and antibody response to sheep red blood cells; Hg
Jayaraman et al., 2009	37	New Bedford Harbor, MA	Eggs, nestlings	PCBs, DDE
Jones et al., 1993	38	Green Bay, WI	Eggs, nestlings	PCBs; PCDF; PCDD
Kraus, 1989	39	Hackensack River Estuary, NJ	Eggs, nestlings	Metals
Longcore et al., 2007a	40	Acadia NP ME, Ayer, MA	Eggs, nestlings, feathers	Hg; success
Longcore et al., 2007b	41	Acadia NP ME, Ayer, MA	Eggs, nestlings, feathers	Hg; growth
Martinovic et al., 2003a	42	St. Lawrence R. Basin (SLRB), ON	Nestlings	CHCs (67 PCBs PCDD PCDF); Vitamin A
Martinovic et al., 2003b	43	SLRB, ON	Nestlings	CHCs (PCBs PCDD PCDF); stress plasma
Maul et al., 2006	44	Crab Orchard NWR, IL	Nestlings	PCBs
Maul et al., 2010	45	Crab Orchard NWR, IL	Eggs, nestlings	PCBs
Mayne et al., 2004	46	Southern ON	Eggs, nestlings	Non-persistent pesticides; DDE; stress and immune response
Mayne et al., 2005	47	Southern ON	Nestling	Non-persistent pesticides; DDE; thyroid function

Table 1.1. Cont.

McCarty and Secord, 1999a	48	Hudson R., NY	Adults	Nest building behavior; PCB
McCarty and Secord, 1999b	49	Hudson R., NY	Nestling	Productivity, mass and survival; PCBs
McCarty and Secord, 2000	50	Hudson R., NY	Sub adult females	Plumage color; PCBs
McCarty, 2001/2002	51			Review paper
Neigh et al., 2006a	52	Kalamazoo R., MI	Eggs, nestling	PCBs, productivity
Neigh et al., 2006b	53	Kalamazoo R., MI	Eggs, nestling	PCBs
Neigh et al., 2006c	54	Kalamazoo R., MI	Eggs, nestling	PCBs (Risk assessment)
Nichols et al., 1995	55	Saginaw Bay, MI	Eggs, nestlings	PCBs
Nichols et al., 2004	56	Hudson R., NY	Nestlings	PCBs
Papp et al., 2007	57	Point Pelee N P (PPNP), ON	Nestlings	PCBs
Secord et al., 1999	58	Hudson R., NY	Eggs, nestlings, adults	PCBs
Shaw, 1984	59	Alberta	Eggs nestlings	OC pesticides, PCBs
Smits et al., 2000	60	Athabaska R. Basin, AB	Nestling	Reclaimed oil sand mine sites; Nestling immune function, growth and survival
Smits et al., 2005	61	PPNP, ON	Nestlings	OC pesticides, PCBs
Spears et al., 2008	62	Crab Orchard NWR, IL	Eggs, nestlings	PCBs
Stapleton et al., 2005	63	Hudson R., NY and sites in AB ON WI MI	Nestlings	PCBs; minisatellite DNA mutation rates
Tsipoura et al., 2007	64	Meadowlands, NJ	Eggs, nestlings (carcass and feather), a few adults	5 Metals
Wayland et al., 1998	65	Alberta and Saskatchewan	Nestlings	Biomarkers; pulp mill and sewage effluent
Yorks, 1999	66	Maryland	Eggs, nestlings	PCBs biomarkers and reproductive parameters; dosing study

Table 1.2. Trace elements in tree swallow studies, listing highest mean Hg concentration and effects where appropriate. THg = Total mercury, MeHg = Methylmercury

Study	Trace Elements	Highest Hg Mean ppb dw unless noted otherwise	Effects Finding
1	THg	Eggs: 456	NA
7	Hg	Female blood: 17800 Feather: 13.55 ppm ww	Reduced fledge success for year old females at contaminated site
8	Hg	Eggs: 1965 Blood: 6150	No interclutch variability, MeHg 96.5% of egg Hg, female blood Hg positively correlated with average hg of eggs in her nest, sampling any egg would give an estimate that was ~10% of the true mean 100% of the time
12	THg and 18 other trace elements	Eggs: 300	NA
14	THg and 17 other trace elements	Pipers: 640 Nestlings: 310	NA (Reduced hatch success in some years relative to PCBs)
15	THg, MeHg and 19 other trace elements	Nestling liver: Hg 143 Nestling liver: MeHg 117	Nesting success differences, however, attributed to dioxins
17	THg and 18 other trace elements	Eggs: 250 Nestling liver: 240 Nestling: 270	Elements generally at background concentrations, no increased Hg due to drawdown
18	THg and 18 other trace elements	Eggs: 9230 Nestling liver: 4210	Reduced hatching but not significantly different
20	THg and 18 other trace elements	Egg: 310 Nestling liver: 190	Drawdown did not influence element concentrations or bioindicator response
21	THg and 18 other trace elements	Eggs: 204 Nestling liver: 160 Nestling: 180	Element concentrations not elevated, reproductive success normal, difference in [Hg] relative to wetland type.
22	THg and 30 other trace elements	Nestling liver: 80 (results pooled with ~30% other insectivorous passerines)	Elevated Pb liver tissue concentrations, Fecal samples were not a good indication of what elements were ingested

Table 1.2. Cont.

23	THg and 18 other trace elements	Hg detected in only 44% of eggs	Difference in egg order among and within clutches for suite of 7 elements detected in >50% of eggs, Mn, Zn increased with egg order, B decreased
30	THg	Egg: 1457	"No relationship b/t blood, egg or feather Hg and stress induced corticosterone" in adults
		Adult blood: 4980	
33	MeHg	Egg: 518	Nested earlier after flooding, no success differences
		Nestling (no feather): 188	
34	THg	Blood: 3600	Model-averaged survival 1% lower in adults breeding at contaminated sites
36	THg	Blood: 16250	Females at more contaminated sites had weaker PHA-induced swelling responses, but this was not predicted by blood Hg concentrations
39	Cr, Cu, Pb, Ni, Cd	NA	NA
40	THg	Egg: 1280 ng	No success differences, suggests MeHg toxicity threshold for TRES eggs of 1000 ppb ww
		Nestling: 13590 ng	
41	THg	Feathers: 3000	Feather MeHg negatively correlated with linear growth rate weight b/t 2-10 days
64	As Cd, Cr, Pb, Hg	Blood: 97	Elevated blood concentrations of Pb, Hg concentrations elevated, but below ecological thresholds

Table 1.3. Highest mean sum PCB concentrations. DO = day old.

Study	Number of PCB congeners	Medium	Highest Mean ppb ww
1	Total	Eggs	4120
		Nestlings (b/t 4-17 DO)	2970
5	Total	Nestling	5469
10	15 and total	Pipers	3290
		11-13 DO nestlings	3770
11	Total	Eggs	540
13	91 and total	Eggs	330
		11-13 DO nestlings	300
15	134+ and total	Eggs	1130
		9-13 DO nestling	1720
14 - 19	90+ and total	Pipers	100880
		12 DO nestlings	44660
20	Total	Nestling	215
23	Total	Eggs	161000
24	160	Eggs	6800
25	Not listed	Adult carcass	370
		Eggs	330
26	70 and total	Nestling	16400
27	Sum of 56	16 DO nestlings (less liver and testis tissue)	104
28	116+ and total	Eggs	25000
		15 DO nestlings	96000
29	Total	Eggs	180
31	Sum of 110	Eggs	810
		15 DO nestlings	2272
35	42+ (but only 5 in sum)	16 DO nestlings	32
37	Total as sum of 18	Eggs	11200
		9-13 DO nestlings	16800
38	Total	Eggs	10800
		16 DO nestlings	13100
44	Sum of 15	15 DO nestlings	2828
45	Sum of 55	Egg	4604
		Nestling	2297
		Local source	43252 ng
58	Not listed	Eggs	29500
		Nestlings	62200
52-54	Approximately 100	Eggs	8100
		12 DO nestling	4300
57	85 and 66 for sum	13 DO nestlings	473

Table 1.3. Cont.

61	sum of 85	12 DO nestling less digestive tract	786
62	112	Eggs	4452
		12-17 DO nestlings	3994
66	Total	Eggs	948
		12 DO nestlings	18460

Table 1.4. Concentrations of polychlorinated dibenzo-p-dioxin (PCDDs), polychlorinated dibenzofurans (PCDFs), Furans and OC pesticide contaminants. This lists only the highest concentrations and the concentration for 2,3,7,8-TCDD, the most toxic dioxin and 2,3,4,7,8-PeCDF the most toxic furan. ≤ indicates congener was coeluted, thus actual concentration of individual congener is a concentration less than or equal to the listed value.

Study	Medium	Contaminant	Highest Mean ppb ww
1	Eggs	2,3,7,8-PCDD	0.0052
		OCDD	0.140
		2,3,4,7,8-PCDF	≤0.0065
		2,3,7,8-PCDF	≤0.040
	4-17 DO nestlings	2,3,7,8-PCDD	0.0028
		1,2,3,4,6,7,8-PCDD	0.071
		2,3,4,7,8-PCDF	≤0.0021
		2,3,7,8-PCDF	≤0.026
3	Eggs	DDE	2290
5	Nestling	DDE	1484
10	Pippers	*p,p'*-DDE	200
11	Eggs	2,3,7,8-TCDD	0.026
		2,3,7,8 TCDF	0.22
		DDE	170
13	Eggs	2,3,7,8-TCDF	0.0057
		DDE	0.099
	11-13 DO nestlings	2,3,7,8-TCDD	0.003
		2,3,7,8-TCDF	0.028
		DDE	0.008
14	Pipers	1,2,3,4,6,7,8-HpCDD	0.049
		1,2,3,7,8-PeCDF	1.01
		2,3,4,7,8-PeCDF	0.09
	12 DO nestlings	1,2,3,4,6,7,8-HpCDD	0.022
		2,3,7,8-TCDF	0.507
		2,3,4,7,8-PeCDF	0.114
15	Eggs	2,3,7,8-TCDD	1.013
		OCDD	0.568
		2,3,4,7,8-PeCDF	0.016
		1,2,3,4,6,7,8-HpCDF	0.057
	12 DO nestlings	2,3,7,8-TCDD	0.99
		1,2,3,4,6,7,8-HpCDD	0.038
		2,3,7,8-TCDF	0.008
		2,3,4,7,8-PeCDF	0.003
19	Eggs	*p,p'*-DDE	460
20	12 DO Nestling	*p,p'*-DDE	32
24	Eggs	OCDD	0.213
25	Eggs	*p,p'*-DDE	1300

Table 1.4. Cont.

29	Eggs	DDT	40
		DDE	11200
		DDD	749
35	16 DO nestlings	1,2,3,4,6,7,8-HpCDD	0.036
		1,2,3,4,6,7,8-HpCDF	0.010
		DDE	178.3
37	Eggs	DDE	526
	9-13 DO nestlings	DDE	235
42 - 43	16 day old nestlings	total PCDDs	0.080
		total PCDFs	0.121
46 - 47	Eggs	*p,p'*-DDE	1820
59	Eggs, viable	DDE	1010
	Eggs, unhatched	DDE	2230
	11 DO nestling	DDE	310
61	12 DO nestlings	DDE	309

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CHAPTER 2

TRACE ELEMENTS
AND
ECOLOGICAL PARAMETERS

Introduction

Biomonitors are frequently used to assess the extent of local contamination and to determine the ecological risks to local wildlife presented by those contaminants. This is particularly useful for assessing whether an area of concern will act as a population 'sink' for species that utilize it. The selection of a useful biomonitor requires an understanding of the species' natural history and how those habits interact with questions of interest. Tree swallow (*Tachycineta bicolor*) ecology is relatively well understood as they have been extensively studied in a variety of sub-disciplines (e.g. Kuerzi, 1941; Davis, 1982; Zach, 1982; Quinney et al., 1986; Robertson et al., 1992; Jones, 2003; Ardia, 2005; Winkler et al., 2005). This comparatively large body of ecological knowledge combined with their ease of study and their tendency to feed in close proximity to their nest sites (Quinney et al., 1985; Mengelkoch et al., 2004) has contributed to an increase in the use of tree swallows as biomonitors for North American wetland ecosystem health (Secord et al., 1999; McCarty, 2002; Jones, 2003; Maul et al., 2006; Neigh et al., 2006).

Tree swallow nestlings have accumulated high concentrations of organic pollutants and physiological or behavioral changes have been associated with these contaminant burdens. For example, high polychlorinated biphenyl (PCB) concentrations along the Hudson River were found to affect nest building behavior (McCarty and Secord, 1999), plumage coloration, and reproductive success (McCarty and Secord, 2000). Organophosphorus insecticide applications in Ontario orchards tended to reduce parent to nest feeding visits by tree swallows in a manner suggestive of cholinesterase-inhibiting insecticide exposure (Bishop et al., 2000). Swallows have been shown to accumulate dichlorodiphenyldichloroethylene (DDE) (Neigh et al., 2006), polycyclic aromatic hydrocarbons (Custer et al., 2001), as well as polychlorinated dibenzo-p-dioxins and dibenzofurans at various locations throughout their summer range (Harris and Elliot, 2000). Tree swallows have also been used to investigate the movement and effects of mercury (Bishop et al., 1995; Gerrard and St Louis, 2001; Custer et al., 2007a; Longcore et al., 2007a; Longcore 2007b), and other trace elements (Kraus, 1989; Tsipoura et al., 2007; Custer et al. 2009) in the environment.

Inorganic mercury (Hg) is naturally converted to the more toxic methylmercury (MeHg) in aquatic sediments (Peakall, 1972; Eisler, 2000). MeHg is a known teratogen, carcinogen and mutagen and has been shown to adversely affect growth and reproduction in both terrestrial and aquatic organisms (Eisler, 2000). It is a strong immune suppressor in vertebrates (Hawley et al., 2009). Methyl mercury bioaccumulates and biomagnifies in animals, and tree swallows are primarily exposed through diet and maternal transfer to the eggs (Eisler, 2000). In birds, dietary exposure to mercury is primarily to MeHg (Burger and Gochfeld, 1997), and almost 100 percent of what is consumed is absorbed rather than excreted (Wolfe et al., 1998). Total mercury (hereafter "mercury" or "Hg") in tree swallows has been shown to be almost entirely comprised of MeHg (Gerrard and St. Louis, 2001), as has been demonstrated with other passerines (Rimmer et al., 2005). Methylmercury was 84% of Hg with red-winged blackbird (*Agelaius phoeniceus*), Brewer's blackbird (*Euphagus cyanocephalus*), and cliff swallow (*Hirundo pyrrhonota*) (Wolfe and Norman 1998). Similarly, egg Hg content has been shown to be 96.5% MeHg (Braune & Gaskin, 1987). Mercury is mainly depurated through molting, and new feathers in gulls contain up to 93% of the Hg body burden after molting (Braune & Gaskin, 1987), and it has been estimated that feather growth in tree swallow nestlings accounted for 80-92% of body depuration of Hg (Longcore et al., 2007b). Egg-laying is also a depuration route for females (Eisler, 2000; Brasso et al., 2010). Mercury burdens in birds are influenced by species specific absorption rates, trophic level of diet, exposure differences, sex, age, and molt (Burger and Gochfeld, 1997).

Tree swallows are migratory, and this may affect their use as biomonitors. The half-life of MeHg in blood is rather long: 84 days in mallard ducks (*Anas platyrhynchus*) (Heinz and Hoffman, 2004), 40 to 60 days in non-molting adult Cory's shearwaters (*Calonectris diomedea*) (Monteiro and Furness, 2001), and 60 days in seabirds (Wolfe et al., 1998). In dosing studies with chickens, Hg was present in newly laid eggs long after the dosing was halted. Elevated Hg concentrations were found in eggs laid 29 (Kiwimae et al., 1969), 46 (Kambamanouli et al., 1991), and 60 days after dosing stopped (Sell et al., 1974) and all of these time frames were the maximum studied. There is only a 2-4 week period when swallows arrive at their nesting sites and begin to lay eggs (Robertson et al., 1992), so this should be considered when determining the proportion of Hg burden that is from local sources and the proportion originating from sources outside the study site (Rimmer et al., 2005). Moreover, because tree swallows are opportunistic predators, assumptions that the prey is primarily of aquatic origin should be confirmed (Maul et al., 2006; personal observation).

In this study I investigated Hg and trace element exposure at three wetlands in the Lake Calumet area of northeastern Illinois. I quantified trace element concentrations in sediments, diet items, eggs and nestlings. I attempted to account for off-site accumulation of Hg by tree swallow mothers, and I collected food boluses to determine diet composition in an attempt to understand terrestrial and aquatic sources of contaminants. Tree swallow productivity was assessed as well. Understanding local contaminants and their effects on productivity is of particular interest in the Lake Calumet region as it is an area that was once dominated by wetlands. Now, however, the study sites offer some of the only wetland habitat for reproductive animals for miles. In such situations, the few remaining wetlands can act as population sources or sinks, depending on a variety of factors, including the extent of contamination.

Methods

Study sites and habitat

The Lake Calumet area of Illinois is highly industrialized, and has a long history of industry and waste disposal (USEPA, 2011; Sprenger et al., 2001; USACE and Tetra Tech, 2001). Three wetlands in the area were selected to provide a spectrum of contamination. Big Marsh (41° 41' 30", 87° 34' 24") and Indian Ridge Marsh (41° 40' 51", 87° 33' 50"), hereafter referred to as Indian Ridge, were chosen for the high levels of Aluminum (Al), cadmium (Cd), chromium (Cr), copper (Cu), lead (Pb), Hg, selenium (Se), zinc (Zn), PCBs, and Dichlorodiphenyldichloroethane (DDD)/DDE, contamination in sediments (Sprenger et al., 2001) and Powderhorn Lake (41° 38' 53", 87° 31' 37") (Figure 2.1), was selected as a reference site because it had no known sediment contamination. In March of 2004, 30 tree swallow nest boxes (9.5 x 14 x 20 cm interior size, 3.8 cm diameter entry hole) were placed on posts at approximately 2 m above the ground, and at a minimum of 15 m apart at each of the sites. Boxes with no obscuring vegetation between them were placed at greater distances to avoid intraspecific competition. Approximately one-meter-long segments of polyvinyl chloride (PVC) pipe filled with expandable foam insulation (Great

Stuff©, Dow Chemical) were installed below each box to reduce predator access. All boxes were placed within 20 m of water, with the vast majority of boxes within 5 m of water. In some cases a wide band of emergent aquatic vegetation existed between the nest box and open water, increasing the distance between the box and open water. Nest box sites were evaluated for habitat openness by measuring canopy cover using a convex spherical densiometer and estimating percent cover, as well as four measurements of the distance to nearest woody neighbor taller than 2 m at 90^{o} intervals. Additional measurements of the distance to open water and nearest water were recorded as well as accessibility in front of nest box which measured the horizontal angle of gap created by the vegetation below box height within 5 m.

Nest measurements

In 2004, nest boxes were visited periodically starting on April 20 and then on alternate days from May 9 until all nestlings had fledged in early July. In 2005, twice weekly box visits commenced March 19, and alternate day visits began on May 11 and continued through July 1. Box visits involved opening the box and observing nest development, parent coloration, egg number, nestling number and appearance, and looking for signs of predation or competition from other species, like muddy raccoon prints on the predator guard or twig house wren nests in a box previously occupied by tree swallows. In 2005 nesting material from competitor species found in nest boxes was removed.

Fourteen day-old nestlings were weighed to the nearest quarter gram using a Pesola spring scale. Proportion hatch was defined as the proportion of eggs in a clutch hatching, and proportion fledge was defined as the proportion of nestling fledging from a successfully hatched clutch. Nesting success was defined as having one nestling fledge from a clutch of eggs. Nests with raccoon predation (14 attempts, 1 successful) or human interference (eg: 5 stolen or vandalized boxes) were excluded from success and proportion calculations, depending on the timing of the occurrence.

Media collection

Grab samples of sediments were collected in acid washed, 500 ml glass jars from the sediment surface at three or four locations that were accessible and close to the nest boxes within each wetland during the summer of 2004. The samples were frozen, and then air-dried. When a stable weight was achieved, the samples were ground using a tabletop Retsch Laboratory Mortar Grinder.

Aquatic insects were collected qualitatively in both years from each site with a kick net. Netting was performed along vegetated shorelines, near nest boxes. Larger genera were collected disproportionately to ensure there was enough mass for contaminant analysis, and genera not regularly seen in bolus samples were minimally included. All insects were frozen, identified to order or family, sorted and rinsed in de-ionized water before submission for contaminant analysis.

Food boluses were collected from all 12- to 14-day-old tree swallow nestlings in 2004 and from all 6- and 10-day-old nestlings in 2005. Boluses were collected using the ligature method (Orians, 1966, Orians and Horn, 1969; Quinney and Ankney, 1985; Yorks, 1999; Menglekoch et al., 2004) with 89 mm, black Ty-Rap© zip ties. One hundred thirty-six ligature attempts were made in 2004 and 376 attempts were made in 2005. Ligatures were left on for 30 to 45 minutes. Boluses were preserved with 95% ethanol in 2004 but were frozen in 2005. Bolus contents were cleaned of mucus, and insects were identified to family or lower. The 2005 insects were dried at 30 ºC and submitted for contaminant analysis.

Five 0.3 square meter insect emergence traps were placed on the surface of the water near the nest boxes at each site on May 20, 2004 and May 3, 2005. The traps were checked and insects collected every other day until the traps were removed on June 30. Insects were frozen within 48 hours of emergence, then identified to order or family, sorted and rinsed in de-ionized water before submission for contaminant analysis. In 2004, insects from each emergence trap were identified and recorded daily for a one week sub-sample. In 2005 insects were identified and recorded after each trap check. All insect samples for contaminant analysis were ground under liquid argon and sample aliquots were stored at –20ºC.

In 2004, the first two eggs in each of ten nests from each site were marked and collected. In 2005 the largest two eggs in each of ten nests at each site were collected to ensure sufficient biomass for analyses. Previous studies of other bird species, have suggested that laying order is not related to organic contaminant burdens in eggs of other species (Reynolds et al., 2004; Van den Steen et al., 2006) and Brasso et al., (2010) reported that the mercury concentration in any one tree swallow egg is representative of the other eggs in the same nest. Whole eggs were stored at 4ºC until the contents could be removed from the shell, combined with another egg from the same nest and homogenized with an Omni ES Mixer. Egg contents were then digested and analyzed as indicated below.

In both years, the heaviest 14 day-old nestling from each of the same nests from which eggs were collected was euthanized (University of Illinois Institutional Animal Care and Use Committee protocol number 03028). Carcass were placed in separate clean 500ml glass jars and frozen. The digestive tracks were excised from partially thawed carcasses. The carcasses were then homogenized using liquid argon and a blender, before being returned to storage at –20ºC.

Sample analysis

Aluminum (Al), arsenic (As), barium (Ba), Cd, Cr, Cu, iron (Fe), Pb, manganese (Mn), Hg, nickel (Ni), Se, silver (Ag), and Zn were analyzed in all sediment, insect, egg and nestling samples, and cobalt (Co) was analyzed in insect and nestling samples as well. Nitric acid microwave digestion procedures equivalent to US EPA Method 3051 (USEPA, 2003) for eggs, US EPA Method 3052 (USEPA, 2003) for nestlings and all insects, and a modified version of USEPA Method 3051 (USEPA, 2003) using only nitric acid for sediments were used prior to analysis. Quality control (QC) samples were prepared with each type of samples in each of the digestion batches. Batch QC measures for insects and eggs included a digested reagent blank, digested duplicate, analytical duplicate, matrix spike, analytical spike, and a Dogfish Liver Standard Reference Material (SRM). The same QC procedures were used for sediments but with Montana Soil #2710 SRM. In general, the overall QC for these samples ranged from acceptable to excellent. Duplicates were reproducible, and spikes and SRMs recovered well with few exceptions. Sample results for most metals pose a maximum uncertainty of $\pm$30% except for Cr which may be biased high in 2004 eggs as recovery was consistently approximately 190%. Mercury recovery in matrix and analytical spikes, and standard reference material averaged 88%, and all media samples except one egg sample had Hg concentrations above the reporting limit (0.06 mg/kg dry weight [dw] for eggs).

Results for most metals were obtained by Inductively Coupled Plasma Mass Spectrometry (ICP-MS) using scandium, yttrium, niobium, rhodium, lanthanum, and thorium as internal standards. Mercury analyses were conducted using a VG Elemental PQ ExCell ICP-MS and a PS Analytical Millennium Fluorescence System. All results are reported as total mass of element per unit dry weight, unless noted otherwise.

Statistical analysis

Differences in ecological endpoints and mean elemental concentrations in all media were compared using Analysis of Variance (ANOVA) in SAS (SAS System for Windows V8.2, © 1999-2001 SAS Institute Inc, Cary NC, USA) or with Kruskal-Wallis in SAS. Differences between years were first evaluated with ANOVA, and data from both years were pooled if results indicated no significant differences between years using $p<0.05$. Differences in mean element concentrations among sites were determined using ANOVA and individual comparisons were performed with contrast statements in SAS. Ecological endpoints and mean elemental concentrations in all media were assessed for parametric statistic suitability with Levene's test for homogeneity of variance and Shapiro-Wilks test for normality using a significance level of 0.1. Results failing normality tests were either log-transformed to normality or analyzed with nonparametric Kruskal-Wallis tests using SAS and individual site differences were determined with Dunn's method in Sigma Stat (© 2004 Systat Software, Inc). Means and standard deviations were calculated in SAS and values greater than two standard deviations were considered extreme outliers and excluded from statistical analyses. Relationships among variables were evaluated with Pearson's Correlation via SAS. Analysis of nesting and fledgling success parameters were made using a z test in Sigma Stat. Statistics for a given element were not performed if greater than half of the samples had concentrations below the detection or reporting limit. When values below the detection or reporting limit were included in analyses, the value equal to half of the limit was used.

To make a more accurate estimation of the Hg burden nestlings acquired on site, I calculated "local-sourced contaminant mass" as follows. The Hg concentration (in dry weight) in a nestling was multiplied by the dry weight of that nestling to obtain Hg mass. The same process was done with the eggs. Finally, the average mass of the contaminant in the eggs was subtracted from the mass in the nestling from the same nest. A generalized egg shell mass of 0.1g (DeWeese et al., 1985) was subtracted from the fresh egg mass, and all wet weights were transformed to dry mass using individual percent solids for nestlings and mean solid proportions from Calumet eggs for individual years (17.3% solid for 2004 and 17.6% solid for 2005). Differences between nestling and egg contaminant mass that were less than zero were changed to zero. I reported trace element concentrations in dw. Comparisons to values reported in the literature as wet weight were converted to dw using values from this study (nestlings 30.8% moisture and eggs 82.7% moisture).

Results

Habitat and Productivity

Including data from all three sites, tree swallow adults completed nests at significantly different dates in 2004 and 2005 (Table 2.1). In 2004, there was a significant difference among sites in the date of nest completion, with nests completed significantly earlier at Big Marsh when compared to Powderhorn (means in Julian date 128, 131, 134 for Big Marsh, Indian Ridge and Powderhorn, respectively; Table 2.1, Table 2.2). Again in 2005, there was a significant difference among sites in the date of nest completion; however, that year Indian Ridge was the site with the earliest completed nests (Julian date means 127, 125, and 130 for Big Marsh, Indian Ridge and Powderhorn, respectively; Table 2.1, Table 2.2). In 2005, nest building at the three sites took between 3 and 34 days, calculated from the first appearance of nesting material in boxes to the time of observation of feathers lining the cup (I do not have observations early enough to document this for 2004).

Late nesting tree swallows laid fewer eggs. Julian date of nest completion was positively correlated with date of the first egg being laid ($r = 0.96$, $p < 0.001$), but negatively correlated with number of eggs per nest (r = -0.57, p<0.001) with sites and years combined. The number of eggs per nest was not significantly different between the years (Table 2.1). Tree swallows laid significantly different numbers of eggs per nest among the three sites with the years pooled, and the late arriving swallows at Powderhorn had significantly fewer eggs per nest than swallows at Big Marsh (Table 2.1, Table 2.2 for individual years). Nestlings were heavier in 2004, and had significantly different masses among sites in both years (Table 2.1, Table 2.2). Nestlings were significantly heavier at Powderhorn than at either of the other two sites in 2004 and nestlings at Indian Ridge were heavier than those at Big Marsh (Table 2.1). In 2005, nestlings were again heavier at Powderhorn than the other two sites (Table 2.1). There were no differences in proportion hatch or fledge between years nor were there significant differences in proportion hatch or fledge among sites when years were combined (Table 2.1, Table 2.2).

Physical habitat among sites was slightly variable (Table 2.3). The percent closed overstory and distance to open water were the only parameters that differed among sites (Table 2.1). Boxes at Powderhorn had the greatest canopy cover and boxes at Indian Ridge were furthest from open water. Indian Ridge had a ring of emergent aquatic vegetation in the shallow water, accounting for the different distance to open water. There were no statistical differences among sites for other habitat variables including distance to vegetated water, nearest woody neighbor taller than 2 m, or openness/accessibility in front of nest box (all p>0.05). None of the measured habitat parameters differed significantly when comparing used and unused boxes across all 3 sites (p>0.05 for each).

Nest box use suggested that tree swallows had more competition for nesting sites at Powderhorn from other species. One Black-capped Chickadee (*Poecile atricapillus*), three house sparrows (*Passer domesticus*) and 12 house wrens (*Troglodytes aedon*) established nests in the nest boxes, compared to nine house wren nests at Indian Ridge and seven house wren nests at Big Marsh in 2004.

Combining data from all sites and both years, Hg concentration in eggs was positively correlated with Julian Date of nest completion (Figure 2.2) and negatively correlated with brood size (r= -0.53, p<0.001).

Diet

Total insect dry mass collected in emergence traps at each site was between 0.87 and 2.64 g for the two years (Table 2.4). The week long sub-sample of emergence trap insect mass in 2004 indicated that there was no significant difference in capture among sites (Table 2.5). A more thorough evaluation of emergence trap insect mass capture in 2005 indicated significantly different capture mass among sites, with significantly less emergence trap insect mass collected from Big Marsh than the other two sites (Table 2.4, Table 2.5). Likewise, insect counts from emergence traps were significantly different among sites in 2005 (Table 2.5). Powderhorn had significantly higher numbers of zygopterans and anisopterans in emergence traps than did the other two sites (Table 2.5). Indian Ridge had the greatest count of sum dipteran, coleopteran, and other non-odonate insects among the three sites (Table 2.5).

Forty-three boluses were collected from nestling tree swallows in 2004 and 121 were collected in 2005. Odonata, Diptera, and Coleoptera were most abundant by count (Table 2.6). Of these orders most prominent in boluses, there were significant differences in numbers of insects per bolus between years ($p<0.05$ for each); however, the boluses did not have significantly different numbers of the less numerous insect orders between years, so these insects were pooled by year and by insect type for further analysis. The sum of homopterans, trichopterans and miscellaneous insects in boluses were not different among sites (Table 2.7). In bolus samples from 2004, there were no significant differences in numbers of dipterans, coleopterans or hymenopterans per bolus among sites; however, there were significant differences among the sites in the number of odonates found in boluses (Table 2.7), with boluses at Powderhorn containing more odonates than those at the other two sites. Similarly, in 2005, there were no significant differences in numbers of dipterans, coleopterans or hymenopterans per bolus among sites, but odonate count in boluses was again significantly different among sites in 2005, with significantly more odonates in boluses from Powderhorn than both Big Marsh and Indian Ridge (Table 2.7). In 2004, aquatic insects were 34% of the total bolus insect count and 63% in 2005, assuming all Diptera were aquatic (along with the Odonata, and Trichoptera). All Coleoptera, Hemiptera, Hymenoptera, Lepidoptera and other insects were considered terrestrial. In 2005 boluses were identified to genus, and a more thorough identification determined dry weight biomasses of 52, 51 and 64% aquatic origin for Big Marsh, Indian Ridge and Powderhorn, respectively.

Mercury

Mean sediment mercury concentrations were not significantly different among sites (Table 2.8; mean sediment concentration 0.192, 0.084, and 0.102 mg/kg for Big Marsh, Indian Ridge and Powderhorn, respectively). Similarly, mercury concentrations in aquatic insects were not significantly different when comparing different sampling methods (Table 2.8), so emergence trap insects, benthic insects and the aquatic portion of bolus insects were pooled for site analysis. Mean concentrations of total mercury in all insects were 0.026, 0.027, and 0.033 for Big Marsh, Indian Ridge and Powderhorn, respectively. There was no significant difference in Hg concentrations in aquatic insects from different sites (Table 2.8; individual means in Table 2.9). Terrestrial bolus insects did not have significantly different Hg concentrations compared to aquatic bolus insects (Table 2.8), but the sample size was low. There were no site differences when aquatic and terrestrial bolus insects were pooled (Table 2.8), but again there was a low sample size.

Total mercury concentrations in eggs ranged from 0.08 to 0.32 mg/kg dry weight (Figure 2.3). There was no difference in Hg concentration in eggs between years; however, there was a significant difference in concentrations among sites with the years pooled (Table 2.8). Both Big Marsh and Indian Ridge had eggs with lower concentrations of Hg than eggs at Powderhorn and eggs at Big Marsh had lower concentrations than eggs at Indian Ridge (Table 2.8, Figure 2.3). Mean concentrations of Hg in eggs were 0.12, 0.19, and 0.23 mg/kg for Big Marsh, Indian Ridge and Powderhorn, respectively with years pooled.

One swallow nestling sample at Big Marsh had Hg concentrations of 1 mg/kg which was greater than two standard deviations from the mean so it was excluded from statistical analysis, though it is indicated in the figures. Concentrations in nestlings were not significantly different between years (Table 2.8); however, the concentration pattern among the three sites was different between the two years (Figure 2.3), so years were not pooled to better illustrate these differences. Total mercury concentrations in swallow nestlings were significantly different among sites in

2004, with nestlings from Indian Ridge having significantly greater concentrations of Hg than nestlings at Powderhorn and Big Marsh (Table 2.8). Concentrations in nestlings in 2005 were significantly different among sites as well, with nestlings at Big Marsh and Indian Ridge both having significantly lower Hg concentrations than nestlings at Powderhorn (Table 2.8).

Local-sourced total Hg mass in nestlings was analyzed excluding the aforementioned outlier. Local-sourced Hg mass was significantly different between years (Table 2.8, Figure 2.4). Mercury mass in nestlings from 2004 was significantly different among sites, with nestlings from Indian Ridge having greater Hg than nestlings at both Big Marsh and Powderhorn (Table 2.8). Nestlings from 2005 also had significantly different masses of local-sourced Hg among sites. Powderhorn nestlings had significantly greater local-sourced Hg than nestlings at Big Marsh and Indian Ridge (Table 2.8). Hg from the eggs contributed 5 (± 2 s.d.)% to the Hg mass in the Calumet nestlings. Mercury is being accumulated locally, as indicated from the difference between the mass in the eggs and the mass in the nestlings, but one might expect that different proportions would be accumulated considering what is available from the nestling diet and the local sediment (Figure 2.5). Sample sizes in insect and sediment samples were too small to permit comparisons, unfortunately.

Other Trace Elements

All trace elements were detected in all sediment samples (Table 2.10). Trace element concentrations in sediments did not differ among sites (all values $p>0.05$).

Barium, Cd, Cr,Cu, Mn, Se, and Zn concentrations did not differ by benthic, bolus, and emergent insect type and were pooled ($p>0.05$, Table 2.11); Ag was not included in analyses as most of the samples were below the detection or the reporting limit. For the pooled insect samples only copper concentrations differed among sites, with Indian Ridge having lower concentrations of copper in insects than Powderhorn (Table 2.12). When insect sample types that could not be pooled are analyzed separately, only Fe in bolus insects was significantly different among sites (Table 2.12). Concentrations of Fe in bolus insects at Big Marsh were significantly greater than concentrations at Powderhorn (Table 2.12; means: Big Marsh, 355; Indian Ridge, 210; Powderhorn, 120 mg/kg dw). Trace element concentrations in benthic insect samples that were not pooled were not significantly different among sites ($p>0.05$). All bolus insect trace element concentrations tested were not significantly different between aquatic and terrestrial origin, ($p>0.05$) except Pb concentrations, which were significantly greater in terrestrial insect samples compared to aquatic insect samples (Table 2.12; means 0.53, 0.74 mg/kg dw for aquatic and terrestrial bolus insects, respectively).

Tree swallow egg samples were compared among sites for Ba, Cr, Cu, Fe, Mn, Se, and Zn. Copper, Fe, Mn, and Se concentrations in eggs did not differ between years, and none were significantly different among sites when analyzed with years pooled (all values $p>0.05$). Barium, Cr, and Zn concentrations in eggs did not differ among sites in 2004 or 2005 (Table 2.13 for means).

Concentrations of Ag, As, Cd and Ni in nestlings were largely below the reporting or detection limit in both years, detections of Al were rare in 2004 nestlings, and Pb was mostly below the reporting limit in 2005 (Table 2.14 for means). Concentrations of Ba, Co, and Cu in nestlings did not differ between years ($p>0.05$, Table 2.14). When these concentrations were pooled by year, there were no significant differences among sites for Co and Cu, but nestling Ba concentrations were significantly different and nestlings at Indian Ridge had higher Ba concentrations than nestlings from Big Marsh and Powderhorn (Table 2.12). Nestling concentrations of Se and Cr from 2004 were significantly different among sites (Table 2.12). Nestlings at Indian Ridge had significantly greater Cr concentrations than did nestlings at Big Marsh whereas concentrations of Se were significantly greater in nestlings from Big Marsh than in those from Powderhorn (Table 2.12). In 2005, there were no significant differences in nestling trace metal concentrations among sites for elements that were not pooled by year ($p>0.05$ for all, Table 2.14).

Discussion

Mercury

There was no specific pattern in Hg concentrations among different sample media at the sites. Mean Hg in sediment samples from Big Marsh were twice that of the other sites; however, there was no statistical difference likely due to the small sample size. Insect samples, likewise did not have statistically different mean Hg concentrations among sites, but insects at Powderhorn tended to have the greatest mercury concentration. Eggs at Powderhorn had significantly greater concentrations of mercury than the other sites, and nestling Hg concentrations and local source nestling mass of Hg was greatest at Indian Ridge in 2004, but greatest at Powderhorn in 2005. Trophic transfer is occurring as the nestlings are not acquiring much Hg from their eggs, and Hg concentrations in nestlings are greater than concentrations in their diet items.

Mercury concentrations in Big Marsh sediments were higher than the threshold effects concentration (TEC) and threshold effects level (TEL) standards of 0.18 and 0.174, respectively (MacDonald et al., 2000; Buchman, 2008), but sediments at Indian Ridge and Powderhorn were below these ecological screening standards. This means that sediments at Indian Ridge and Powderhorn would be considered unlikely to have toxic effects on sediment organisms. Sediments at Big Marsh were above this standard, suggesting they may cause toxic effects to some sediment dwelling organisms, but they were below the probable effects standard so effects may be somewhat less likely.

Relative to other tree swallow studies, insect Hg concentrations at the Calumet sites were rather low. Mean Calumet insect mercury concentrations were higher than concentrations in diet items from mine tailing contaminated sites in Colorado (Custer et al., 2009), but lower than concentrations found in Agassiz National Wildlife Refuge (NWR, Custer et al., 2006) and in the some Canadian experimental lakes (Gerrard and St. Louis, 2001).

Egg concentrations at the Calumet sites were low to intermediate and nestling concentrations were low, relative to other tree swallow studies. Concentrations of Hg in Calumet eggs were similar to concentrations from Agassiz NWR (Custer et al., 2006) and pool eight of the

Mississippi River (Custer et al., 2007b), though far lower than concentrations near an old mine in Nevada (Custer et al., 2007a) likely the highest Hg concentrations found in tree swallow eggs. Mercury concentrations in nestlings from Calumet were similar to concentrations from contaminated sites from a number of studies (Gerrard and St Louis, 2001; Custer et al., 2003a; Custer et al., 2006; Custer et al., 2008) and were much lower than reported by Longcore et al.(2007a) and Custer et al. (2007a).

Local-source mercury mass in Calumet nestling was variable between the two years, and among the sites. This may have been the result of the variability of mercury concentrations in the sediment samples and perhaps it underscores the opportunistic nature of tree swallow foraging. It would be insightful to have large numbers of terrestrial and aquatic insect bolus samples to more accurately determine whether differences exist in insect burdens relative to insect habitat.

Habitat and Productivity

Mercury is known to reduce hatchability and impair reproduction (Eisler, 2000), and Heinz and Hoffman (2003) observed reduced hatchability in eggs of mallards at concentrations as low as 2.1 mg/kg dw (Heinz and Hoffman 2003, assuming 64.5% moisture from Ricklefs, 1977); however, mercury concentrations in the eggs I examined were an order of magnitude lower than that found to impair hatchability in mallards. The hatch and fledge proportion at the sites (71%-90%) is similar to the mean of 78.8% from almost 3500 tree swallow nests throughout North America (Robertson et al., 1992; Custer et al., 2007a). The reproductive ecology of tree swallows near Lake Calumet was consistent with that of tree swallows across their range. Mercury concentrations in tree swallow eggs and nestlings in the Lake Calumet area were below avian levels affecting egg and nestling success and well below a general concentration for reduced hatching in tree swallow eggs of approximately 5.7 mg/kg dw (1 mg/kg ww) suggested by Longcore et al (2007a).

Swallows prefer to colonize nest boxes that are in areas of relative openness, rather than densely shrubby or canopied areas (Rendell and Robertson, 1990), and the swallows at the sites nested at the more open sites 3 to 6 days earlier than at the less open one, Powderhorn. This difference in timing among the sites may have had an effect on ecological fitness, since tree swallows, like many migratory passerines, are well documented to have larger broods the earlier they nest (Robertson et al., 1992; Stutchbury and Robertson, 1987; Winkler and Allen, 1996). I observed a difference in brood size among sites that could be the result of the timing of nesting (Robertson et al 1992), parent age and experience (Robertson et al., 1992; Robertson and Rendell, 2001) or possibly food availability (Murphy, 2000). I attempted to quantify female age in 2005 by observing coloration, but the sample size of confirmed aged females was small, and was confounded by the inability to similarly estimate male age. Greater food availability at Powderhorn might suggest it to be a preferred site, contrary to colonization dates.

I believe the reduced brood size for nestlings at Powderhorn was associated with later nesting (based on Robertson et al., 1992). Nonetheless, the relationship in Calumet-area tree swallows between mercury egg content, viewed as a surrogate for female body burden of mercury, and nest timing, which can impact productivity, is intriguing. There may be numerous factors why birds arrive and nest at different times, and the study was not designed to address these. However, the effects of compounds like mercury, in conjunction with stressful events like migration need to be better understood. This study suggests that understanding of ecological effects of contamination would benefit from investigating indirect reproductive affects such as timing of nesting or migration success. The tree swallows that nested later do not necessarily accumulate greater concentrations of mercury on site than do the early arrivals, but they do appear to experience greater interspecific competition for nest sites and have fewer young.

Trace Elements

Arsenic, Cd, Cr, Cu, Ni, Pb, and Zn concentrations in Calumet sediments were above the TEC, the benchmark at which toxicity becomes more likely (Ba, Mn, and Se are not listed) (MacDonald et al., 2000). Sediment manganese concentrations were above the lowest effects level (LEL) (Buchman, 2008), which function similarly to the TEC, and below which sediment toxicity is unlikely. Cadmium, Cr, and Ni concentrations at Big Marsh, a few samples from Powderhorn were higher than the probable effects concentration (PEC) or probable effects level (PEL), both of which are generally predictive of sediment toxicity (MacDonald et al., 2000). Concentrations of Pb at Big Marsh and Powderhorn, Mn at Big Marsh, and Zn at Big Marsh were higher than the severe effects level (SEL) indicating the sediments were severely polluted and that adverse effects on sediment dwelling organisms would be expected to occur (MacDonald et al., 2000). These effects values are general guidelines and definitive classification of sediment toxicity would require further evaluation.

Mean Al, As, Ba, Cu, Pb, Mn, Ni, and Se concentrations in Calumet insects were greater than the (geometric) mean concentrations found in tree swallow dietary items from mine tailing contaminated sites in Colorado (Custer et al., 2009), but mostly were similar in concentration to the contaminated sites in the few other studies found (Custer et al., 2002, Custer et al., 2003b, Custer et al., 2007a). Concentrations of Pb were far greater in Calumet insects than those in the diet of tree swallows in the Lostwood NWR, ND (Custer et al., 2008); however, concentrations in the diet of starlings (*Sternus vulgaris*) nesting near highways when Pb additives were still used in gasoline were about ten fold higher (Grue et al., 1986).

Generally, mean concentrations of elements in tree swallow eggs in this study were similar to concentrations found in tree swallow eggs along the Housatonic River in MA (Custer et al., 2003a). Mean Cu and Fe concentrations are a little higher in some Calumet sites than the geometric mean from the Housatonic study. Calumet egg trace element concentrations were generally similar to or slightly higher than concentrations in tree swallow eggs from along the North Platte River, WY (Custer et al., 2001), Agassiz NWR, MN (Custer et al., 2006), pool 8 of the Mississippi River (Custer et al., 2007b), Lostwood NWR, ND (Custer et al., 2008) and along the Carson River, NV (Custer et al., 2007a). Egg Se concentrations were lower than barn swallow egg concentrations at both the control and Se contaminated sites in Texas (King et al., 1994).

Comparative results for nestlings are not as abundant in the literature as they are for eggs, because different tissues are often analyzed. Calumet nestling trace element concentrations were generally similar to those of nestlings along the Housatonic River, MA and from the Lostwood NWR, ND. Calumet nestlings had higher Ba concentrations than nestlings from the Housatonic study, and higher Fe and Pb concentrations than nestlings from the Lostwood study, but overall these concentrations are considered as background or ‘no effects’ concentrations (Custer et al., 2003a).

Elevated concentrations of Pb, Mn and Zn at some Calumet sites may be problematic for sediment dwelling organisms. Manganese and Zn are considered 'essential' or 'beneficial' elements and are fairly well regulated by birds (as cited in Custer et al., 2009) and were not elevated in the Calumet bird and egg tissues. Lead has deleterious impacts on metabolism, growth, development, reproduction, and survival of most species, but unless large doses occur, such as through ingestion of lead shot for hunting, lead toxicosis is unlikely (Eisler, 2000). Lead has been shown to impact other passerines. Barn swallows (*Hirundo rustica)* nesting along roads had reduced red blood cell (RBC) δ-aminolevulinic acid dehydratase (ALAD) of 31% in adults with mean Pb concentrations of 5.1 mg/kg (Grue et al., 1984). Starlings (*Sturnus vulgaris*) nesting in a road right of way had nestling carcass Pb concentrations of 4 mg/kg and significantly reduced brain mass, haemoglobin concentration, percent haematocrit, and RBC ALAD activity (Grue et al., 1986). These reductions do not impact nestling survival in the nest, but their success after fledging is unknown (Grue et al., 1986). These concentrations are well above the mean concentration in Calumet nestlings, but within the observed range.

Diet origin

Bolus samples from the tree swallow nestlings at the Calumet area sites over the two years indicated that their diet comprised of Diptera (44%), Coleoptera (19%), Hymenoptera (14%), Homoptera (11%), and Odonata (8%) by count. Calumet nestlings in 2005 were fed between 51 and 64% aquatic insects by mass, which is useful in understanding contaminant uptake since terrestrial insects had greater concentrations of Pb than aquatic insects at the Calumet sites. Sample size was low, and other differences between aquatic and terrestrial insects may have been missed. Many tree swallow studies assume the birds are primarily eating emergent aquatic insects, but Calumet swallows consume both terrestrial and aquatic insects in fairly equal proportions. The fact that tree swallows are opportunistic insectivores underscores the importance of understanding diet source to provide greater insight into local contaminant movements. The use of eggs and nestlings were integral in determining the extent of local-source contaminant uptake. Egg Hg from unknown maternal sources contributed approximately 5% of the Hg mass in the Calumet nestlings indicating that the vast majority of nestling Hg accumulation is by local dietary uptake. Another study calculated that the egg contributed between 2.4 ± 0.9 and 23.9 ± 3.8% of the 14-day-old nestling burden of mercury (Longcore et al., 2007b).

Conclusions

Tree swallows nesting at three sites in the Calumet area of Chicago, Illinois experienced no detectable reproductive effects, with Hg concentrations in the eggs as high as 0.32 mg/kg dry weight, and maximum concentrations in nestlings of 1 mg/kg dry weight. Local diet and sediment Hg concentrations, along with the timing of tree swallow migration, egg development and Hg pharmacokinetics in birds reinforced the understanding that tree swallow egg contaminant values should be interpreted as indications of maternal contaminant burden obtained from local nesting sites *and* previous unknown sources. Rosten et al., (1998) came to similar conclusions, reporting that correlations of mercury in passerine eggs to concentrations in nesting site vegetation and insects ‘seemed to depend mainly on whether the bird was migratory or resident’ (Rosten et al., 1998).

Eggs and nestlings are useful for calculating an estimate of locally derived contaminant mass. In the Lake Calumet area, tree swallow eggs contributed only about 5% of the nestling Hg mass. We found that concentrations of different contaminants in eggs from the Calumet region contributed up to 50% of the nestling contaminant mass, so understanding the egg-nestling relationship was very useful in understanding local contaminant uptake.

Total Pb concentrations in sediment at Powderhorn, the reference site, were above probable effect levels, which could have had deleterious effects on sediment dwelling organisms. Overall concentrations in biotic media were considered to be at ‘no effects’ levels, but physiological and physical changes have been demonstrated at concentrations within the range of Calumet observations (Grue et al., 1984; Grue et al., 1986), though their effects on survival or success are unknown.

Mercury and other contaminants of concern may indirectly affect fitness. This combined with the fact of habitat degradation in the Calumet region that has left a fraction of the original wetland size, underscores the importance of monitoring remaining nesting habitat for the future of wildlife populations. Biomonitors as thoroughly studied as tree swallows may be well suited to study this. However, greater understanding is only possible with detailed knowledge of the local habitat, food web, and the habits of the biomonitor.

I attempted to understand local sources of contamination by the use of sediment and diet samples and by quantifying terrestrial and aquatic sources of diet. Moreover, by collecting both eggs and nestlings I could distinguish whether contaminants burdens were transferred from the mother from external sources or were accumulated on site. The continued and thoughtful use of bio-monitors can shed more light on these issues and will be imperative for understanding the role habitat contamination plays in regions with increasing habitat loss in areas such as Calumet, Illinois.

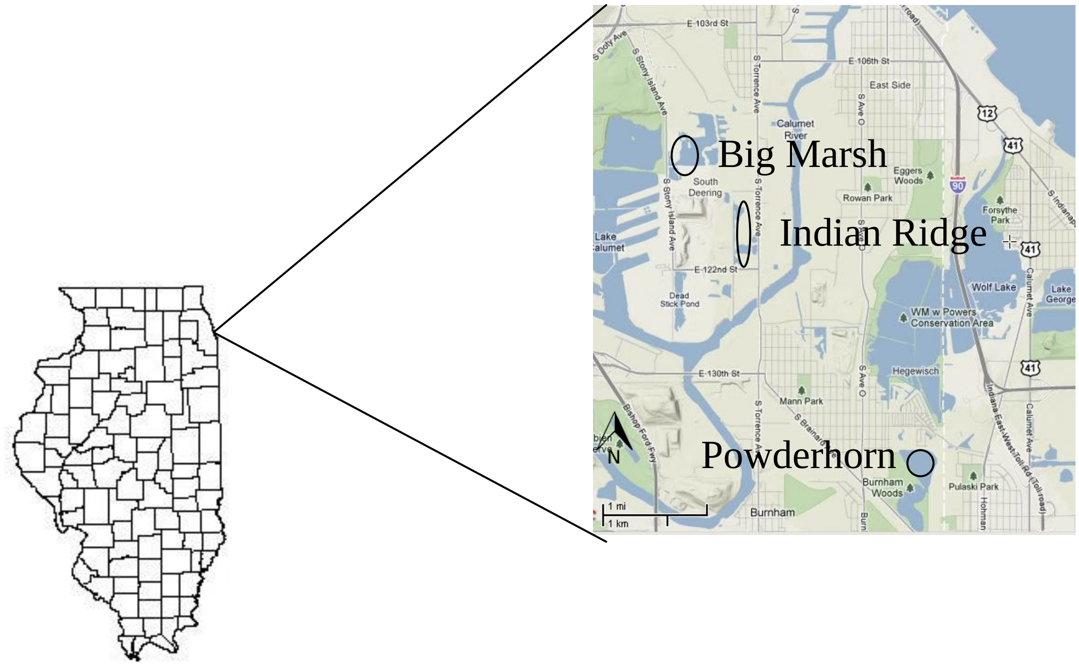

Figure 2.1: Map of Illinois, USA with expanded view of study sites.

Table 2.1. Test statistics and p values for habitat and productivity statistical comparisons.

Variable	Comparison	Test statistic	p
J.D. completed nest	years	H=13.56	<0.001
J.D. completed nest 2004	sites	H=15.84	<0.001
BM vs PL	sites	Q=3.67	<0.05
J.D. completed nest 2005	sites	H=9.89	0.007
BM vs IR	sites	Q=2.45	<0.05
IR vs PL	sites	Q=2.68	<0.05
Number of eggs	years	H=0.01	0.94
Number of eggs	sites	H=9.18	0.01
BM vs PL	sites	Q=2.75	<0.05
Nestling mass	years	H=32.01	<0.001
Nestling mass 2004	sites	F=10.83	<0.001
BM vs PL	sites	F=19.65	<0.001
IR vs PL	sites	F=4.38	0.039
BM vs IR	sites	F=7.06	0.009
Nestling mass 2005	sites	F=3.65	0.028
BM vs PL	sites	F=4.44	0.036
IR vs PL	sites	F=7.29	0.008
Proportion hatch	years	H=0.28	0.597
Proportion hatch	sites	H=0.93	0.628
Proportion fledge	years	H=0.82	0.365
Proportion fledge	sites	H=0.40	0.818
Percent overstory cover	sites	H=12.30	0.002
Distance to open water	sites	H=18.89	<0.001

Table 2.2. Mean ± standard deviation tree swallow nesting ecology parameters at three wetlands in the Calumet region, IL. Numbers in parentheses = sample size. Letter indicates mean is significantly different from that of different letters (p>0.05). Hatch and fledge proportion includes second nest attempts, nest success does not.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
J.D. completed nest	$128.1^{a} \pm 2.2$ (18)	$127.4^{A} \pm 4.1$ (25)	$130.9^{ab} \pm 4.6$ (13)	$124.8^{B} \pm 2.9$ (20)	$133.9^{b} \pm 2.6$ (8)	$130.5^{A} \pm 7.2$ (13)
# eggs/ nest[1]	$5.7^{a} \pm 0.7$ (18)	$5.3^{a} \pm 0.7$ (27)	$5.1^{ab} \pm 0.8$ (15)	$5.6^{ab} \pm 0.8$ (21)	$4.9^{b} \pm 0.7$ (11)	$4.9^{b} \pm 1.0$ (13)
proportion hatch^	0.8 ± 0.3 (21)	0.8± 0.4 (26)	0.8 ± 0.4 (17)	0.9± 0.2 (21)	0.7 ± 0.5 (11)	0.7± 0.4 (13)
proportion fledge^	0.9 ± 0.3 (19)	0.9 ± 0.2 (22)	0.8 ± 0.4 (11)	0.8 ± 0.3 (19)	1.0 ± 0.0 (8)	0.7 ± 0.4 (10)
nest success^	86% (21)	88% (24)	71% (17)	86% (21)	73% (11)	90% (10)
nestling mass	$21.3^{a} \pm 1.7$ (67)	$20.5^{A} \pm 2.3$ (93)	$22.3^{b} \pm 1.9$ (38)	$20.1^{A} \pm 2.4$ (76)	$23.2^{c} \pm 1.2$ (21)	$21.6^{B} \pm 1.7$ (21)

J.D. = Julian Date
^ indicates nests that failed through known causes – predation, human influence - were excluded from the calculation.
[1] indicates that variable was pooled by year for statistical evaluations of site differences.

Table 2.3. Habitat variables measured ± standard deviation. All values are in meters except orientation and angle of openness (degrees), percent overstory cover (percent) and densiometer value for shrubby cover (count out of 96). Distance to nearest woody neighbor over 2 m (DNN) 0 is the quadrat encompassing 45 degrees to either side of the box opening, DNN represents the quadrat to the east of the opening, DNN 180 to the rear, DNN 270 to the west. Orientation is the degree representation of the box opening.

Habitat variable	Big Marsh	Indian Ridge	Powderhorn
DNN 0	8 ± 9	9 ± 9	12 ± 9
DNN 90	5 ± 7	5 ± 7	3 ± 5
DNN 180	7 ± 8	7 ± 9	2 ± 4
DNN 270	5 ± 7	6 ± 7	2 ± 4
Densiometer value for cover	58 ± 32	60 ± 26	49 ± 27
Percent overstory cover	26 ± 27	21 ± 27	40 ± 29
Orientation	179	230	217
Angle of openness	130 ± 109	155 ± 106	146 ± 70
Distance to open water	5.3 ± 1.7	50.4 ± 44	5.6 ± 2.5
Distance to vegetated water	5 ± 2	5 ± 3	6 ± 3

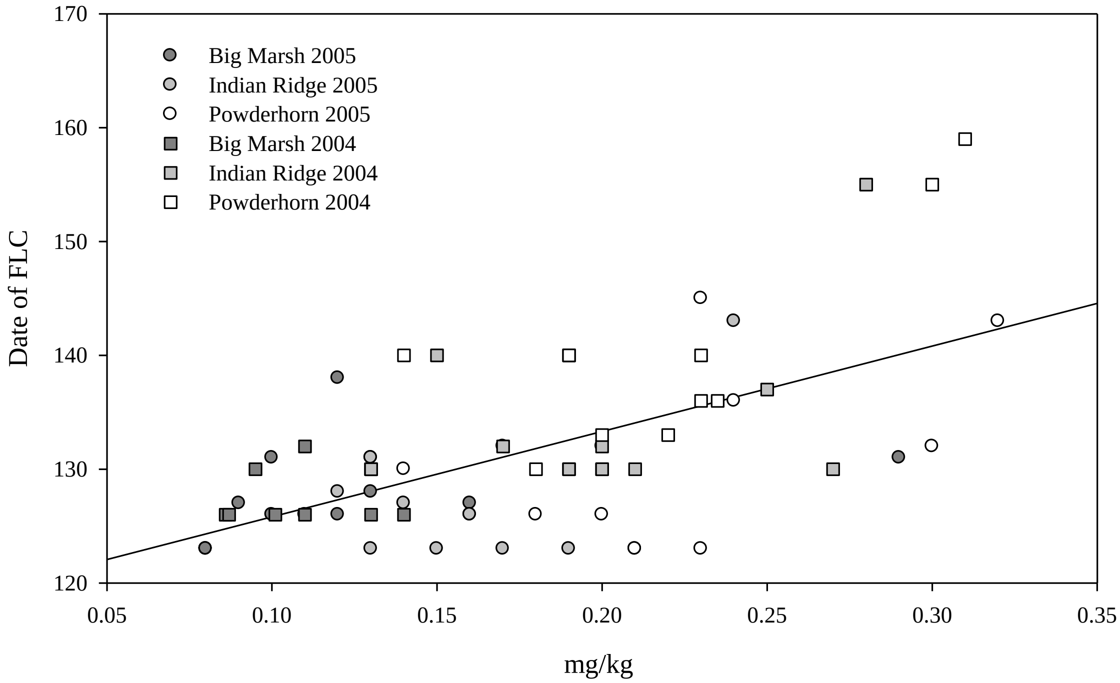

Figure 2.2. Nesting timing as indicated by the Julian date that feathers were first found in the nest, plotted with the concentration of total mercury in the eggs from those nests ($r=0.60$, $p<0.0001$). Circles are 2004 values and squares are 2005. FLC = feather lined cup.

Table 2.4. Total insect dry mass in grams from three wetlands in the Calumet region, IL. Emergent traps in 2005 were deployed two weeks longer than in 2004.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
Emergent trap insects	2.03	0.87	2.64	1.86	1.60	2.12
Bolus insects A	nm	1.15	nm	0.51	nm	0.76
Bolus insects T	nm	0.95	nm	0.49	nm	0.27

T = terrestrial
A = aquatic
nm = not measured

Table 2.5. Test statistics and p values for emergent trap insect statistical comparisons.

Variable	Comparison	Test statistic	p
Insect mass 2004	sites	F=0.14	0.870
Insect mass 2005	sites	F=7.25	0.009
BM vs IR	sites	F=8.06	0.015
BM vs PL	sites	F=13.08	0.004
Zygopterans 2005 count	sites	H=123.02	<0.001
BM vs PL	sites	Q=7.73	<0.05
IR vs PL	sites	Q=8.87	<0.05
Anisopterans 2005 count	sites	H=118.56	<0.001
BM vs PL	sites	Q=8.06	<0.05
IR vs PL	sites	Q=8.73	<0.05
Non-Odonate 2005 count	sites	F=15.54	<0.001
IRvs PL	sites	F=17.94	<0.001
IR vs BM	sites	F=27.62	<0.001

Table 2.6. Number of insects per bolus at three wetlands in the Calumet region, IL.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
Odonata	1.1	0.6	0.7	0.3	5.5	3.7
Diptera	4.6	11.7	5.7	9.3	4.3	10.2
Coleoptera	11.1	0.7	35.3	0.9	5.5	1.2
Homoptera	1.4	2.9	2.7	1.1	0.6	3.3
Hymenoptera	0.8	5.3	2.8	1.9	0.2	2.0
Other	0	0.5	0.5	0.4	0.3	1.1

Table 2.7. Test statistics and p values for bolus insect statistical comparisons.

Variable	Comparison	Test statistic	p
Odonata 2004	sites	H=8.80	0.012
BM vs PL	sites	Q=2.27	<0.05
IR vs PL	sites	Q=2.42	<0.05
Odonata 2005	sites	H=38.28	<0.001
BM vs PL	sites	Q=5.24	<0.05
IR vs PL	sites	Q=5.03	<0.05
Homoptera+Hymenoptera+other	sites	H=0.52	0.769

Table 2.8. Test statistics and p values for Mercury concentration statistical comparison for all media from three wetlands in the Calumet area, IL.

Variable	Comparison	Test statistic	p
Sediment	sites	F=0.97	0.424
Aquatic insects	sampling methods	F=0.62	0.547
Aquatic insects	sites	F=0.68	0.203
Bolus insects	terrestrial vs aquatic	F=1.50	0.289
Bolus insects (all)	sites	F=2.84	0.203
Eggs	years	F=2.36	0.129
Eggs	sites	F=30.79	<0.001
BM vs PL	sites	F=60.10	<0.001
IR vs PL	sites	F=8.92	0.004
BM vs IR	sites	F=25.69	<0.001
Nestlings	years	H<0.01	0.987
Nestlings 2004	sites	F=4.09	0.028
BM vs IR	sites	F=6.86	0.014
IR vs PL	sites	F=4.96	0.035
Nestlings 2005	sites	F=13.51	<0.001
BM vs IR	sites	F=20.38	0.001
IR vs PL	sites	F=20.09	0.002
Local Source	years	F=13.84	0.001
Local Source 2004	sites	F=4.25	0.026
BM vs IR	sites	F=6.72	0.016
IR vs PL	sites	F=5.40	0.029
Local Source 2005	sites	F=11.81	<0.001
BM vs PL	sites	F=16.44	<0.001
IR vs PL	sites	F=18.86	<0.001

Table 2.9. Total mercury concentrations in insect samples from three wetlands in the Calumet region, IL. Sample size = 1 for all values. All concentrations mg/kg dry weight. Benthic insect composite samples include Odonata, Diptera, Ephemeroptera.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
Benthic Anisoptera	nm	0.016	nm	0.015	nm	0.040
Benthic Zygoptera	nm	0.010	nm	0.011	nm	0.028
Benthic insect composite	0.033	0.038	0.062	0.025	0.054	0.035
Emergent insect	0.018	nm	0.042	nm	0.034	nm
Bolus insect A	nm	0.037	nm	0.025	nm	0.020
Bolus insect T	nm	0.028	nm	0.011	nm	0.017

T = terrestrial
A = aquatic
nm = not measured

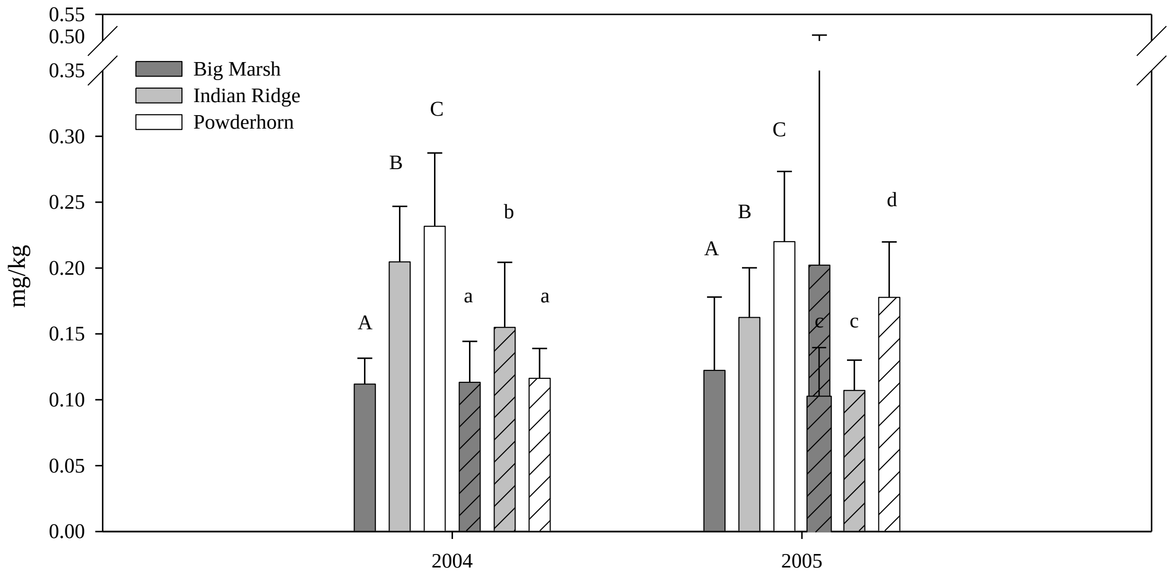

Figure 2.3. Mercury in tree swallow eggs (open bars) and nestling (cross hatch), 2004 and 2005. Error bars are standard deviation, significant differences indicated by different letter. Mercury concentration in 2005 nestlings at Big Marsh are depicted as two values with and without the extreme outlier. Note the break in the Y-axis. n=10 except n=9 for Big Marsh 2005, lower value which had an extreme outlier removed.

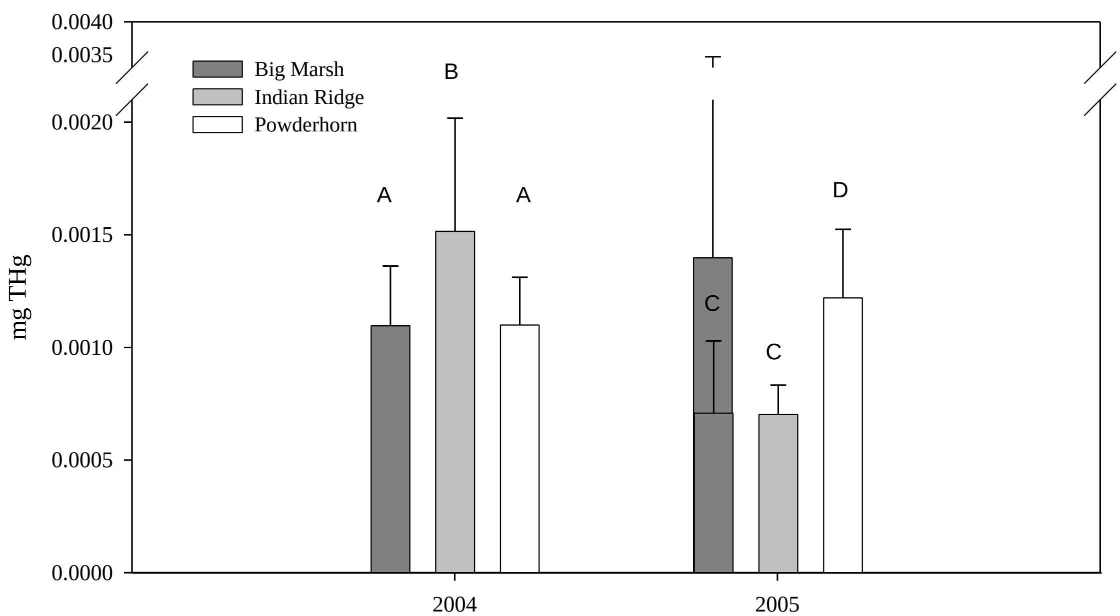

Figure 2.4. Mean local-sourced mercury in swallow nestlings. Local –sourced Hg mass = mass of Hg in nestlings minus mass of Hg in eggs from same nest box. Error bars are standard deviation, significant differences indicated by different letter. Local-sourced mercury mass in 2005 nestlings at Big Marsh are depicted as two values with and without the extreme outlier. Note the break in the Y-axis. n=10 except n=9 for Big Marsh 2005, lower value.

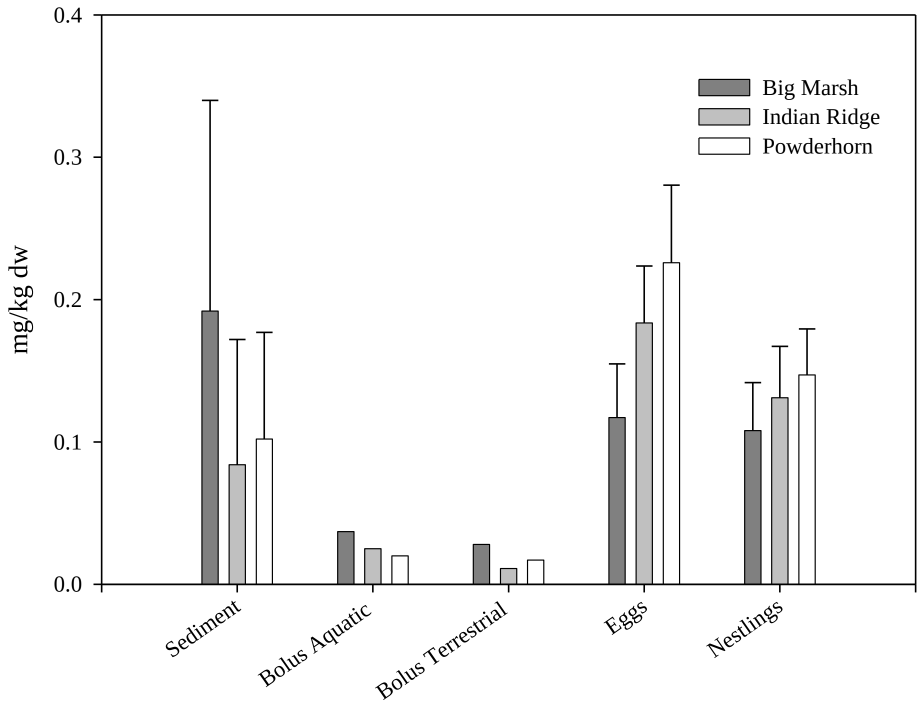

Figure 2.5. Mercury concentrations in sediments, boluses, eggs and nestlings. Error bars, where present are standard deviation. Big Marsh nestling value does not include outlier from 2005.

Table 2.10. Trace metal concentrations in sediment samples from three wetlands in the Calumet region, IL, in 2004. All concentrations mg/kg dry weight.

	Big Marsh	Indian Ridge	Powderhorn
n	3	4	3
Al	17267 ± 9482	19625 ± 14540	20633 ± 14808
As	13 ± 5	7 ± 5	15 ± 10
Ba	176 ± 49	132 ± 102	157 ± 56
Cd	3.0 ± 2.6	0.9 ± 0.9	3.7 ± 1.2
Cr	94 ± 55	51 ± 29	64 ± 34
Cu	107 ± 25	72 ± 74	79 ± 41
Fe	117033 ± 149280	34025 ± 22562	50567 ± 25452
Pb	400 ± 315	87 ± 60	250 ± 61
Mn	2370 ± 2629	510 ± 332	517 ± 431
Ni	50 ± 27	26 ± 17	31 ± 10
Se	1.2 ± 0.6	1.7 ± 0.7	1.5 ± 0.9
Ag	0.5 ± 0.4	0.3 ± 0.3	0.3 ± 0.2
Zn	2081 ± 2898	278 ± 260	362 ± 102

Table 2.11. Concentrations in pooled insect samples (benthic, bolus, and emergent insects) at three sites in the Calumet region, IL, years pooled. All concentrations mg/kg dry weight. Data for Al, As, Co, Fe, Pb, Ni, and Ag not shown due to low detections.

	Big Marsh	Indian Ridge	Powderhorn
pooled n	7	7	7
Ba	9.1 ± 5.8	6.0 ± 3.8	14.3 ± 18.4
Cd	0.4 ± 0.6	0.1 ± 0.1	0.3 ± 0.1
Cr	1.7 ± 0.7	1.2 ± 0.6	2.0 ± 1.0
Cu	21.8[ab] ± 5.4	16.3[a] ± 4.0	26.5[b] ± 7.6
Mn	88.2 ± 117.3	42.8 ± 49.2	175.4 ± 189.7
Se	2.0 ± 1.4	1.4 ± 0.4	1.3 ± 0.4
Zn	110.1 ± 23.4	89.3 ± 15.9	100.1 ± 14.3

Letter indicates mean is significantly different from that of different letters ($p<0.05$).

Table 2.12. Test statistics and p values for trace element concentration statistical comparisons for all media from three wetlands in the Calumet area, IL. Only significant results ($p<0.05$) are reported.

Element	Variable	Comparison	Test statistic	p
Cu	Pooled insect types	sites	F=5.33	0.015
	IR vs PL	sites	F=10.53	0.004
Fe	Bolus insects	sites	F=11.32	0.040
	BM vs PL	sites	F=22.24	0.018
Pb	Bolus	origin	F=8.59	0.043
Ba	Nestlings	sites	F=4.61	0.014
	BM vs IR	sites	F=6.37	0.015
	IR vs PL	sites	F=7.13	0.010
Se	Nestling 2004	sites	H=7.45	0.024
	BM vs PL	sites	Q=2.65	<0.05
Cr	Nestling 2004	sites	H=9.90	0.007
	BM vs IR	sites	Q=3.29	<0.05

Table 2.13. Trace metal concentrations in eggs of tree swallows nesting at three wetlands in the Calumet region, IL. All concentrations mg/kg dry weight.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
n	10	13	15	12	12	11
Ba	1.6 ± 1.1	2.0 ± 0.9	0.2 ± 0.0	2.9 ± 1.1	2.7 ± 1.8	2. 5 ± 1.2
Cr	4.1 ± 2.5	1.5 ± 0.4	3.7 ± 2.0	1.9 ± 0.9	2.9 ± 0.9	2.1 ± 1.6
Cu	3.8 ± 1.1	4.8 ± 1.4	6.0 ± 3.5	4.1 ± 1.5	6.8 ± 2.9	5.0 ± 3.2
Fe	196.6 ± 77.3	139.2 ± 27.5	137.6 ± 75.3	134.7 ± 31.0	104.5 ± 30.7	171.6 ± 71.5
Mn	2.8 ± 1.0	3.0 ± 0.9	2.2 ± 0.9	2.9 ± 0.9	2.6 ± 1.2	2.8 ± 0.6
Se	2.4 ± 0.3	2.3 ± 0.4	2.4 ± 0.5	2.2 ± 0.5	2.4 ± 0.7	2.5 ± 0.6
Zn	48.1 ± 13.1	58.6 ± 9.9	40.9 ± 14.5	54.4 ± 7.8	46.4 ± 16.3	56.0 ± 8.6

Table 2.14. Trace metal concentrations in 14-day old tree swallow nestlings from three wetlands in the Calumet region, IL. All concentrations mg/kg dry weight.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
n	11	9	11	10	8	9
Al	~	6.1 ± 2.6	~	6.8 ± 2.4	~	10.6 ± 11.6
Ba[1]	$2.5^{a} \pm 0.7$	$2.6^{a} \pm 1.0$	$3.1^{b} \pm 1.2$	$4.3^{b} \pm 2.2$	$2.3^{a} \pm 0.9$	$2.7^{a} \pm 1.3$
Cr	$1.0^{a} \pm 0.6$	1.1 ± 0.9	$2.5^{b} \pm 1.9$	0.9 ± 0.2	$1.6^{ab} \pm 0.3$	0.9 ± 0.4
Co	0.1 ± 0.0	0.1 ± 0.0	0.1 ± 0.0	0.1 ± 0.1	0.1 ± 0.0	0.1 ± 0.0
Cu	8.0 ± 1.9	7.5 ± 1.2	7.4 ± 1.2	6.9 ± 1.3	8.2 ± 1.3	7.6 ± 2.2
Fe	155.5 ± 38.6	246.7 ± 61.6	133.6 ± 26.2	256.0 ± 43.3	131.3 ± 35.2	285.6 ± 126.8
Pb	0.2 ± 0.1	~	0.4 ± 0.4	~	1.0 ± 2.0	~
Mn	2.2 ± 0.5	2.0 ± 0.4	2.3 ± 0.5	1.8 ± 0.3	2.3 ± 0.5	1.9 ± 0.6
Se	$2.7^{a} \pm 0.9$	2.2 ± 0.7	$2.1^{ab} \pm 0.4$	1.8 ± 0.6	$1.8^{b} \pm 0.4$	1.4 ± 0.8
Zn	84.9 ± 14.7	69.2 ± 10.9	78.1 ± 11.1	63.1 ± 7.2	74.1 ± 8.9	66.7 ± 10.0

~ majority of samples under detection or reporting limit.
Letter indicates mean is significantly different from that of different letters ($p<0.05$).
[1] Samples were pooled by year for statistical analysis

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CHAPTER 3

ORGANIC CONTAMINANTS

Introduction

Aquatic stages of insects (e.g., midge larvae, mayfly nymphs, etc.) accumulate sediment-bound contaminants, including polychlorinated biphenyls (PCBs), organochlorine pesticides (OCs), metals (Cd and Hg), and polycyclic aromatic hydrocarbons (PAHs), and then transport the contaminants into terrestrial food webs upon emergence (Mauck and Olson, 1977; Menzie, 1980; Larsson, 1984; Ciborowski and Corkum, 1988; Kovats and Ciborowski, 1989; Dukerschein et al., 1992; Corkum et al., 1997; Reinhold et al., 1999, Maul et al., 2006). In fact, in a laboratory study, adult chironomids had higher PCB concentrations than the larvae that were exposed to sediment containing PCBs (Larsson, 1984). Therefore, aquatic insects that emerge as adults from polluted wetlands may present a substantial risk of contaminant exposure to aerial insectivores such as bats and birds.

Tree swallows are commonly used as monitors of sediment contaminants in part due to their preference for feeding on insects over open water near their nesting site (Menzie, 1980; McCarty 1997; McCarty and Winkler, 1999; Mengelkoch et al., 2004), and for their utilization of nest boxes (Kuerzi, 1941; Graber et al., 1972). The natural history and ecology of tree swallows is relatively well understood (e.g., Kuerzi, 1941; Robertson et al., 1992), and there has been much work using them to assess contamination throughout their range (see McCarty, 2002; Chapter 1 of this thesis). Secord and colleagues have shown nestlings to accumulate high concentrations of PCBs along the Hudson River (Secord et al., 1999) and suggested this affects nest building behavior (McCarty and Secord, 1999a), plumage color and hatch success (McCarty and Secord, 2000). Others have observed that tree swallow exposure to organophosphorus (OP) insecticide spray events in Ontario orchards tended to reduce feeding visits by parents in a manner suggestive of cholinesterase-inhibiting insecticide exposure (Bishop et al., 2000). Swallows have also been shown to accumulate dichlorodiphenyldichloroethylene (DDE) (Custer et al., 1998), PAHs (Custer et al., 2001), polychlorinated dibenzo-p-dioxins (PCDDs), and dibenzofurans (PCDFs) (Harris and Elliott, 2000).

Many studies have characterized accumulation of PCBs and other organic contaminants in tree swallows and other birds (eg. Ankley et al., 1993; Secord et al., 1999; Custer et al., 2003; Smits et al., 2005), but fewer researchers have looked for polybrominated diphenyl ethers (PBDEs) in birds. PBDEs are flame-retardants that are widely used in household electrical appliances, foams, bedding material and textiles. Congeners with fewer bromines have been found to be highly persistent and bioaccumulative, and in 2004 penta-brominated formations were banned in the European Union (cited in Van den Steen et al., 2009a). Recent studies provide evidence that higher brominated congeners break down to lower brominated forms (Rayne et al., 2003; Stapleton et al., 2004; Van den Steen et al., 2007). Voorspoels et al. (2007) demonstrated that PBDEs move through simple terrestrial food chains in Europe, with measurable concentrations in some common bird species including great tits (*Parus major*), common buzzards (*Buteo buteo*), and sparrowhawks (*Accipiter nisus*). In North America, PBDEs have been found in eggs of peregrine falcons (*Falco peregrinus*) (Holden et al., 2009; Park et al., 2009), ospreys (*Pandion haliaetus*) (Henny et al., 2009), herring gulls (*Larus argentatus*) (Gauthier et al., 2008), and a few other piscivores or birds of prey (see Chen and Hale, 2010). PBDEs are neurotoxic and alter endocrine function (Darnerud, 2003).

Tree swallows are both migratory and opportunistic insectivores, so it is useful to understand the proportion of contaminant burden that is maternally transferred to nestlings from sources outside the study site, as well as the fraction of their diet that is terrestrial or aquatic in origin (Custer et al., 1998; Maul et al., 2006). Diet is particularly relevant because research suggests that terrestrial feeding birds may be exposed to greater concentrations of higher brominated PBDE congeners than aquatic feeding birds (Law et al., 2003; Lindberg et al., 2004; Jaspers et al., 2006; see Chen and Hale, 2010). My research goals were to determine concentrations of organic contaminants in tree swallow nestlings, eggs, and diet and sediment samples at three locations within the Calumet region of Illinois, and to determine organic contaminant loads accumulated by nestlings on site via their diet by comparing contaminant burdens in nestlings to those in eggs from the same nest. To do so, tree swallow eggs and nestlings, insect populations, insects collected from actual food bolus samples, and sediments were analyzed for selected organic contaminants.

Methods

Study sites

The Lake Calumet area of Illinois is highly industrialized, and has a long history of industry and waste disposal (USEPA, 2011; Sprenger et al., 2001; USACE and Tetra Tech, 2001). Three study sites were selected to provide a spectrum of contamination around the Calumet area of Illinois. Big Marsh (41° 41' 30", 87° 34' 24") and Indian Ridge Marsh (41° 40' 51", 87° 33' 50" hereafter Indian Ridge) were chosen for the high levels of 8 metals, PCBs, and Dichlorodiphenyldichloroethane (DDD)/DDE, contamination in sediments (Sprenger et al., 2001) and Powderhorn Lake (41° 38' 53", 87° 31' 37" hereafter Powderhorn) was selected as a reference site because it had no known sediment contamination (Figure 3.1). In March of 2004, 30 nest boxes of 14 by 10 by 20 cm interior size, and 38 mm diameter entry hole were placed on posts at approximately 2 m in height, and at a minimum of 15 m apart at each of the sites. Boxes with no woody cover between them were placed at greater distances. Approximately 1-m long segments of polyvinyl chloride (PVC) pipe filled with expandable foam insulation (Great Stuff© by Dow Chemical) were positioned below each box to reduce predator access. All boxes were placed within 20 meters of water, with the vast majority of boxes within 5 meters of water.

Sample collection and preparation

Grab samples of sediment were collected in acid washed, 500 ml glass jars in 2004 from the sediment surface at three or four locations adjacent to the nest boxes at each wetland. The samples were kept frozen until processed at the lab where they were air-dried and ground using a tabletop Retsch Laboratory Mortar Grinder. A sub-sample was removed, oven dried at 105 °C, and percent moisture was determined. Approximately 10-g portions of the air-dried sediment samples were weighed for sample preparation following the sample extraction, cleanup and fractionation details below, and subsequently analyzed. Total organic carbon (TOC) was measured in sediment samples with a carbon analyzer. Samples were treated with sulfurous acid and dried at low temperature, then analyzed for TOC.

Aquatic insects were collected in both years from each site by kick netting with a D-frame net. Larger genera were disproportionately retained to ensure there was enough mass for contaminant analysis, and genera not regularly seen in bolus samples were minimally included. All insects were frozen, sorted and rinsed in de-ionized water before submission for contaminant analysis. Prior metal contaminant analyses (Chapter 2, of this thesis) reduced sample mass available for organic analyses, leaving only one sample from Indian Ridge and Powderhorn for 2004 and Big Marsh for 2005 and two samples each from Indian Ridge and Powderhorn for 2005.

Food boluses were collected from 12- or 14-day-old tree swallow nestlings in 2004 and from six- and10-day-old nestlings in 2005. Boluses were collected using the ligature method (Orians, 1966; Wilson, 1966; Orians and Horn, 1969; Bryant and Turner, 1982; Yorks, 1999; Mengelkoch et al., 2004) with 89 mm, black Ty-Rap© zip ties. One hundred thirty-six ligature attempts were made in 2004 and 376 attempts were made in 2005. Ligatures were left on for 30 to 45 minutes. Nests were inspected for loose insects and fallen boluses before and after ligatures were applied, and all insects were collected. Boluses were preserved with 95% ethanol in 2004, but were frozen in 2005. Boluses were cleaned of mucus, contents were identified to family or to the lowest relevant level to determine aquatic or terrestrial origin, dried and weighed. Samples from 2005 were submitted for isotope and contaminant analysis as pooled terrestrial or aquatic insects from each site.

In 2004, the first two eggs in each of ten nests from each site were marked and then collected after there were four or more eggs in the nest. In 2005 the largest two of the first four eggs in each of ten nests were collected to maximize sample mass. Studies of laying order effects of contaminant concentration in passerines generally suggest there is either no effect or the among-clutch concentration variance was too large to make specific eggs necessary (Ormerod and Tyler, 1992; Bryan et al., 2003; Reynolds et al., 2004; Van den Steen et al., 2006; Longcore et al., 2007; Van den Steen et al., 2009b; Custer et al., 2010a;). Whole eggs were placed in individual, pre-weighed scintillation vials with chlorine-free padding and stored at 4 ºC. Prior to analysis, the two eggs from a given nest were allowed to come to room temperature, opened, combined in clean glass vials and homogenized with an Omni ES Mixer. All sample aliquots were stored at minus 20 ºC for up to 12 months.

The nestling with the greatest mass was collected from each of the same nests from which eggs were collected. Nestlings were decapitated (University of Illinois Institutional Animal Care and Use Committee protocol number 03028) and whole carcasses were placed in individual, clean 500ml glass jars and frozen. Digestive tracts were removed from the carcasses prior to contaminant analysis. Insect and nestling samples were homogenized under liquid argon and stored at –20 ºC until analysis.

Chemical analyses

Concentrations of 31 PCBs, 15 PBDEs, dichloro-diphenyl-trichloroethane (DDT), its break-down products (DDD and DDE) and ten other chlorinated pesticides were evaluated. The following PBDE congeners (named according to congener number) were measured: 17, 28, 49, 71, 47, 66, 100, 99, 85, 154, 153, 138, 183, 190, and 209. The PCB congeners measured (named according to the Ballschmiter–Zell (BZ) numbering system with international union of pure and applied chemistry (IUPAC) recommendations) include: 5&8, 18, 28, 31, 33, 44, 49, 52, 66, 70, 74, 77, 84&101, 95, 99, 105, 110, 118, 126, 128, 138&163, 149, 153, 180, 183, 187, 194, and 200/201. Numbers joined with an ampersand were co-eluted. The organochlorine (OC) pesticides measured include: alpha-chlordane, beta-chlordane, trans-nonachlor, alpha-hexachlorocyclohexane (a-BHC), beta-hexachlorocyclohexane (b-BHC), gamma-hexachlorocyclohexane (g-BHC or Lindane), heptachlor epoxide, isomer B (HPX), oxychlordane (OXC), dieldrin, heptachlor, DDD, DDE and DDT.

Aliquots of the prepared samples were extracted using a Dionex Accelerated Solvent Extractor (ASE) following a modified *Pressurized Fluid Extraction (PFE)* (USEPA, 1996a). Following the ASE extraction, the samples were taken through two cleanup procedures: a modified *Gel Permeation Cleanup* (USEPA, 1994), and a modified *Silica Gel Cleanup* (USEPA, 1996b). Two silica gel fractions were collected for instrumental analysis, the first contained the PCBs, 4,4'-DDE, Heptachlor, and a small portion of trans-nonachlor, and the fraction second contained the remainder of the chlorinated pesticides. Polybrominated diphenyl ether (PBDE) flame retardants were contained in both fractions.

The sample fractions were analyzed for PCBs and chlorinated pesticides using a Varian 3400 gas chromatograph (GC) equipped with an electron capture detector (GC/ECD) and a Restek Rtx®-5 Integra guard column. The column was 30m x 0.25 mm ID with a 0.25-μm df coating of 5% diphenyl-95% dimethyl polysiloxane. The instrumental analysis followed a modification of USEPA methods, *Organochlorine Pesticides by Gas Chromatography* (USEPA, 2003d), combined with *Polychlorinated Biphenyls (PCBs) by Gas Chromatography* (USEPA, 2003b). To confirm the GC/ECD results and in some cases, to differentiate and quantify some analytes that co-eluted on the GC/ECD, the same fractions were then analyzed on a Varian 3800 GC with Saturn 2000 ion trap mass spectrometer using a modified version of *Semivolatile Organic Compounds by Gas Chromatography/Mass Spectrometry (GC/MS)* (USEPA, 2003c). The same type of GC column was used in the GC/MS analysis. Both silica gel fractions were analyzed for PBDEs using a Micromass Autospec NT High Resolution mass spectrometer equipped with a Hewlett-Packard 6890 gas chromatograph. The GC/MS set up followed a modification of Brominated *Diphenyl Ethers in Water, Soil, Sediment and Tissue by HRGC/HRMS* (USEPA, 2007). A Restek Stx®-500 (15 m x 0.25 mm ID x 0.15 μm df) column was used in the PBDE analyses. Identification and confirmation of the individual PBDE congeners was accomplished using peak retention time and the abundance ratios of selected ion fragments (eg USEPA, 2003a). Quality control (QC) samples were processed and evaluated with each type of samples in each of the digestion batches. Batch QC measures for all samples included a digested reagent blank, digested duplicate, analytical duplicate, matrix spike, and an analytical spike.

Statistical analyses

When possible, I calculated the concentrations of sum PBDE and sum PCB by adding the concentrations of all the individual congeners analyzed in a given media type. Sum PCB was calculated as the total of all individual PCB congeners excluding PCB 126, which was not detected in any media. Sum PBDE or PCB concentrations were only calculated when greater than 50% of congeners were above detection limits in greater than 50% of the samples. When the above criteria were met for a given medium, I used a value equal to half of the detection or reporting limit for samples with values below detection limits. With a few exceptions, in media where these criteria were not met, I generally did not conduct statistical analyses and report only maximum detected concentrations of individual congeners. All media concentrations except sediment are presented in ng/g wet weight (ww); sediment concentrations are reported as dry weight (dw) as these formats are most common in the literature. Nestling, insect and most 2004 egg contaminant concentrations were calculated from dry weight concentrations using individually obtained wet:dry ratios. Some 2004 and all 2005 eggs concentrations were adjusted with a mean solid content for the eggs from that year due to lack of individual ratios. Values were not lipid normalized because individual lipid concentrations were not available for all media and most of the variables did not correlate with lipid concentrations for their respective media type (Herbert et al., 1995).

Yearly differences in contaminant concentrations were compared across all three sites for each media type with one-way analysis of variance (ANOVA) using SAS (V8.2 and V9, 1999 and 2002, SAS Institute, Inc.) to determine whether the data could be pooled for further analyses. Homogeneity of variance was confirmed with Levene's test using alpha 0.1. Normality was assessed visually and with Shapiro-Wilk's test using an alpha value of 0.1 since ANOVA is robust with non-normal data (Zar, 1984). Media pooled between years were compared for contaminant differences among sites with a one-way ANOVA. To minimize comparisons, contrast statements were used to determine if mean concentrations at a specific site were different from those measured at the reference site. Data that lacked homogeneity of variance or normal distribution, were log transformed or analyzed with nonparametric Kruskal-Wallis tests in SAS or in Sigma Stat (© 2004 Systat Software, Inc) and individual comparisons verses the control (Powderhorn) were made with Dunn's method. A Satterthwaite t-test was performed using SAS when there were only two means to compare, such as terrestrial and aquatic insects from food boluses.

When applicable, differences among sites in PCB or PBDE congener suites were investigated using analysis of similarity (ANOSIM) on generated, log transformed, Bray-Curtis Similarity matrices using PRIMER (V5.2.4, 2001, Primer-E Ltd). Congener suites included the same individual congeners that were used for sum values. ANOSIM is a multivariate, non-parametric test of significance, evaluating within and among group distances for multiple variables (congeners) with a single comparison. Using ANOSIM allowed us to determine site or year differences among the suite of congeners without having to create a 'sum' variable of the individual congeners. 'Sum' variables are widely used (e.g., Custer et al., 1998; Dods et al., 2005; Maul et al., 2006); however, different studies may use different congeners in their sum values, ranging from 5 PCB congeners (e.g., Harris and Elliott, 2000) to all 209 (e.g., Custer et al., 2007b) and thus are not readily comparable. In addition, different congener profiles can yield similar sum values, masking some differences.

To make a more accurate estimation of the true concentration of contaminants nestlings acquired on site, I calculated "local-sourced contaminant mass" as follows. The concentration (in dry weight) of a given contaminant in a nestling was multiplied by the dry weight of that nestling to obtain the mass of the contaminant. The same process was done with the contaminant in eggs from the same nest. Finally, the mass of the contaminant in the eggs was subtracted from the mass in the nestling to obtain local-sourced contaminant mass. A generalized egg shell mass of 0.1g (DeWeese et al., 1985) was subtracted from the fresh egg mass, and all wet weights were transformed to dry mass using individual percent solids for nestlings and mean solid proportions from the eggs for individual years (17.3% solid for 2004 and 17.6% solid for 2005). Local-sourced contaminant mass differences were determined by subtracting total sum mass of the relevant congeners in eggs from the total sum mass of the relevant congeners in nestlings the same nests. Differences in nestling and egg contaminant mass that were less than zero were changed to zero (96 out of 128 occurrences for Sum PCBs and 15 out of 608 occurrences for sum PBDEs).

Results

PBDEs

The PBDEs were detected too rarely in sediments to generate sum PBDE values, and therefore, statistical analyses were not conducted. Of the 15 congeners analyzed, only BDEs 71, 47, 99, and 209 were detected, and each of these was detected in less than half of the samples. The maximum concentrations measured for each of these congeners were 0.34 (Indian Ridge), 0.66 (Indian Ridge), 0.52 (Indian Ridge), and 82 (Powderhorn) ng/g, respectively. While BDE 209 was detected in two sediment samples, it was rarely detected in the other media tested (a total of 18 detections out of 139 samples).

Detection of PBDEs in benthic insects was rare as well. Of seven samples analyzed over both years, each PBDE congener was found in at least two samples; however, only BDEs 47, 66, 100, 99, 85, 153, and 183 were detected in over four samples. Therefore, sum PBDE values were not generated for benthic insects, and these data were not statistically analyzed. The maximum concentrations measured for each of these congeners were 2.73 (Big Marsh), 1.52 (Powderhorn), 0.84 (Big Marsh), 2.80 (Big Marsh), 0.88 (Big Marsh), 1.63 (Powderhorn) and 2.85 (Powderhorn) ng/g, respectively.

Five bolus insect samples were analyzed and only BDEs 71, 47 and 99 were found in three or more samples. I did not analyze samples from 2004 and I did not generate sum PBDE values due to the low frequency of congener detection. Maximum concentrations for BDE 71, 47 and 99 in bolus samples were 0.70 (Indian Ridge), 6.60 (Big Marsh), and 2.28 (Indian Ridge) ng/g, respectively. No site had consistently higher concentrations of these congeners than the others, and comparing insects of aquatic and terrestrial origin did not reveal differences in mean sum PBDEs when pooling data from all three sites (Table 3.1).

In tree swallow eggs, ten PBDE congeners were detected in a majority of samples; congeners 17, 71, 138, 190, and 209 were below detection and/or reporting limits in the majority of samples. Mean concentrations of sum PBDEs in eggs at the various sites were analyzed with the years pooled, as concentration means were not significantly different between years (Table 3.1). Mean pooled sum PBDE concentrations in eggs at Big Marsh, Indian Ridge, and Powderhorn were 51, 69, and 68 ng/g, respectively, and there were no significant differences in mean sum PBDE concentrations among sites (Table 3.1, Table 3.2). The suites of all the congener concentrations among the three sites were compared using ANOSIM, and were significantly different between years but not significantly different among sites (Table 3.1, Figure 3.2).

In tree swallow nestlings, concentrations of five PBDE congeners were below reporting or detection limits in a majority of samples (BDEs 17, 49, 66, 190, and 209, though each of these was detected in at least 3 samples). The mean sum PBDE concentrations in nestlings were significantly different between years and among sites in 2005, but not in 2004 (Table 3.1, Table 3.2). Concentrations of sum PBDEs in nestling carcasses from 2005 at both Big Marsh and

Indian Ridge were significantly greater than concentrations in nestlings from Powderhorn (Table 3.1). The suite of PBDE congener concentrations in nestling carcasses was significantly different between the two years (Figure 3.3) and the suite of PBDE concentration profiles were significantly different among sites in both 2004 and 2005 (Table 3.1). In 2004 both Big Marsh and Indian Ridge nestlings had significantly different suites of PBDE congener concentrations than those at Powderhorn, and again in 2005 (Table 3.1).

Local-sourced sum-PBDE mass, (the difference in sum PBDE mass between the nestlings and the eggs from the same nest) were significantly different between years (Table 3.1, Table 3.2). Analyzing the years separately indicated that local-sourced sum PBDE mass in nestlings was not significantly different among sites in 2004, but was significantly different in 2005 ($F=7.87$, $p=0.003$ Table 3.1, Table 3.2). Both Big Marsh and Indian Ridge nestlings had significantly greater local-sourced sum PBDE mass than the nestlings at Powderhorn (Table 3.1). The congeners with the greatest local-sourced sum PBDE masses were 47, 99, 153, and 183 (Figure 3.4). On average, eggs contributed 21 (± 33 s.d.)% of PBDE mass to the nestlings.

PCBs

PCB congener detection in sediment samples was highly variable, with all but 3 congeners (153, 180 and 187) having at least one result below the reporting or detection limit. There were no differences among sites in the concentration of sum PCBs (Table 3.3, Table 3.4). Similarly, the suite of PCB congener concentrations were not significantly different among sites.

Seventeen PCB congeners were detected in >50% of the benthic insect samples (44, 49, 52, 70, 84&101, 95, 99, 105, 110, 118, 128, 138&163, 149, 153 and 187); however, there was only one sample at Big Marsh (due to prior metals analyses), so a t-test comparing Indian Ridge and Powderhorn was performed. Sum PCB concentrations in benthic insects at Indian Ridge and Powderhorn were not significantly different (Table 3.3). Sum PCB concentrations in benthic insects for Big Marsh, Indian Ridge and Powderhorn with years pooled were 44, 11 and 15 ng/g, respectively (Table 3.4).

Seventeen congeners were detected in 3 or more of the five bolus samples (44, 70, 84&101, 95, 99, 105, 110, 118, 128, 138&163, 149, 153, 187, 194 and 200/201). Mean sum PCB concentrations in boluses were not compared because there was only one sample at Powderhorn. The sum PCB means were 34, 64, and 57 ng/g at Big Marsh, Indian Ridge and Powderhorn, respectively (Table 3.4). Suites of PCB congeners in both benthic and bolus insect samples were not tested with ANOSIM due to low replication.

The sum PCB concentrations in the five bolus samples did not differ by origin (terrestrial vs aquatic Table 3.3). The means were 51 (± 10 s.d.) ng/g for aquatic insects, and 49 (± 31) ng/g for terrestrial insects. Similarly, the suite of PCBs were not significantly different by origin (Table 3.3).

All but three of the 30 PCB congeners analyzed (5&8, and 18) were detected in the majority of tree swallow eggs. Mean sum PCB concentrations in eggs were not significantly different between years, and means were significantly different among sites when the years were pooled (Table 3.3, Table 3.4, yearly means shown). Eggs at Indian Ridge had significantly lower sum PCB concentrations than those at Powderhorn (Table 3.3, means 541, 463, and 830 ng/g for Big Marsh, Indian Ridge and Powderhorn, respectively). The suites of PCB congeners in eggs were significantly different between years (Table 3.3, Figure 3.5). The suite of PCB congeners in eggs were not significantly different among sites in 2004, but were significantly different among sites in 2005 ($R=0.192$, $p=0.002$). Eggs from 2005 at both Big Marsh and Indian Ridge had significantly different suites of PCB congeners from eggs at Powderhorn (Table 3.3, Figure 3.4).

Most PCB congeners were detected in the majority of nestlings with the exception of 5&8, 18 and 33. Mean sum PCB concentrations in nestlings were not significantly different between years; therefore, years were pooled for site analysis (Table 3.3, Table 3.4). Mean sum PCB nestling concentrations were significantly different among sites (Table 3.3). Big Marsh nestlings had significantly greater sum PCB concentrations than those at Powderhorn (Table 3.3). The suites of PCBs congeners in nestlings were significantly different between years, and were also significantly different among sites in both years (Figure 3.6, Table 3.3). In 2004 both Big Marsh and Indian Ridge nestlings had significantly different suites of PCB congener concentrations than those at Powderhorn, and again in 2005 (Table 3.3).

Local-sourced sum-PCB mass in nestlings differed among years and among sites for both years (Table 3.3). In both years, local-sourced sum-PCB mass in nestlings from Big Marsh was greater than in nestlings from the reference site, Powderhorn (Table 3.3). Indian Ridge nestlings had significantly greater mass of PCBs than did nestlings at Powderhorn in 2004 (Table 3.3, Table 3.4, Figure 3.7). Eggs contributed 48 (± 77 s.d.)% of the nestling PCB mass.

Organochlorine Pesticides

Thirteen organochlorine pesticides were measured in all media; however, only DDD and DDE were detected in a majority of sediment samples. DDT was found in five sediment samples (Table 3.5). There were no significant differences in DDD or DDE concentrations in sediments among sites (Table 3.6).

DDE was detected in all benthic and bolus insect samples, while DDD was detected in only one bolus sample and 2 benthic samples. DDT and alpha chlordane were each only detected in one benthic sample (Table 3.5). The remaining OCs were not detected in either of these two media types. DDE concentrations in benthic insects were not significantly different between years, but there was only one sample from Big Marsh so concentrations of DDE in benthic insects at Indian Ridge were compared to concentrations at Powderhorn, which were not significantly different (Table 3.5, Table 3.6). Similarly, there was only a single bolus insect sample from the reference site so site means were not compared (Table 3.5). There were no significant differences in DDE concentrations between the aquatic or terrestrial origin of the insects in the bolus samples (Table 3.6). However, the sample size was very low, and aquatic bolus insects had 2.5 times the concentration of DDE than did terrestrial insects (means of 12 and 5 ng/g respectively).

In tree swallow eggs, DDD, DDE, DDT, trans-nonachlor, dieldrin and the sum of HPX and OXC were detected frequently enough for statistical analyses (Table 3.5). Concentrations of trans-nonachlor, DDD, and sum HPX and OXC in eggs were significantly different between years (Table 3.6), and therefore data from each year were analyzed separately. For the rest of the analytes with sufficient detections, years were pooled. DDE concentrations in eggs differed among sites, with eggs at Big Marsh having greater mean concentrations of DDE than the eggs at Powderhorn (Table 3.5, Table 3.6). Concentrations of dieldrin and DDT in eggs were significantly different among sites (Table 3.6). Dieldrin concentrations in eggs at Big Marsh and Indian Ridge were significantly lower than concentrations at the reference site and concentrations of DDT in eggs from Big Marsh were significantly lower than those at Powderhorn (Table 3.6). Concentrations of sum HPX and OXC and DDD in eggs from 2004 were significantly different among sites, though concentrations of trans-nonachlor in eggs were not ($F=0.77$, $p=0.4728$, Table 3.6). Eggs from Big Marsh and Indian Ridge in 2004 had significantly greater DDD concentrations than eggs at Powderhorn; however, the eggs from Powderhorn in 2004 had significantly greater concentrations of sum HPX and OXC than eggs from Big Marsh and Indian Ridge (Table 3.6). Eggs differed in concentrations of sum HPX and OXC and DDD and trans-nonachlor among the three sites in 2005 (Table 3.6). Eggs from Big Marsh in 2005 had greater concentrations of DDD than eggs from Powderhorn (Table 3.6). Eggs from both Big Marsh and Indian Ridge in 2005 had significantly lower concentrations of trans-nonachlor and sum HPX and OXC than eggs at Powderhorn (Table 3.6).

In nestlings DDD, DDE, dieldrin and trans-nonachlor, were detected frequently enough for statistical analyses (Table 3.5) Concentrations of dieldrin, trans-nonachlor and DDE in nestlings differed between years (Table 3.6). In both 2004 and 2005 DDE concentrations in nestlings were significantly different among sites, with the nestlings at Big Marsh having a significantly greater concentration of DDE than nestlings at Powderhorn (Table 3.6). Concentrations of dieldrin and trans-nonachlor in 2004 nestlings were both significantly different among sites (Table 3.6). Nestlings from Big Marsh in 2004 had significantly lower concentrations of dieldrin and trans-nonachlor than nestlings at Powderhorn (Table 3.6). Nestling concentrations of dieldrin in 2005 were significantly different among sites (Table 3.6 and trans-nonachlor $H=2.9228$, $p=0.2319$). In 2005, Indian Ridge nestlings had significantly lower concentrations of dieldrin than nestlings from the reference site (Table 3.6). Pooled by year, concentrations of DDD were significantly different among sites, and the nestlings at Big Marsh and Indian Ridge had significantly greater concentrations of DDD than the nestlings at Powderhorn (Table 3.5, Table 3.6).

Local-sourced DDD and trans-nonachlor mass in nestlings were not significantly different between years (H=0.0646, p=0.7993; H=0.8891, p=0.3457, respectively, Table 3.7); however, DDE and dieldrin were (Table 3.6). DDD mass was significantly different among sites with nestlings at Big Marsh and Indian Ridge having significantly greater local-source mass than nestlings at Powderhorn (Table 3.6). Local-source trans-nonachlor mass in nestlings was not significantly different among sites (H=3.6243, p=0.1633). In 2004 there were differences in local-sourced DDE and dieldrin mass (Table 3.6, Table 3.7), and no significant differences in DDE mass in nestlings when comparing to the reference site (F=3.30, p=0.0820; F=0.58, p=0.4553, comparing Big Marsh and Indian Ridge to Powderhorn respectively). Local-source dieldrin mass in nestlings from Big Marsh was significantly lower than the mass in Powderhorn nestlings (though there was no difference for nestlings from Indian Ridge (Q=0.673, p>0.05) Table 3.6). Local-sourced mass of DDE in 2005 was significantly different among sites (Table 3.6, Table 3.7) with nestlings from Big Marsh having significantly greater local-sourced DDE mass than nestlings at Powderhorn (Table 3.6; Q=1.626, p>0.05 for Indian Ridge and Powderhorn). Local-sourced mass of dieldrin in nestlings was not significantly different among sites in 2005 (H=1.0268, p=0.5985, Table 3.7). Egg mass contributed 42 (± 95 s.d.), 35 (± 34 s.d.), 31 (± 34 s.d.) and 17 (± 54 s.d.)% of nestling contaminant mass for DDD, DDE, trans-nonachlor, and dieldrin, respectively.

Diet

Forty-three boluses were collected from nestling tree swallows in 2004 and 121 were collected in 2005. The insects identified from the bolus samples ranged from 13 (Indian Ridge, 2004) to 69 (Indian Ridge, 2005) % insects of aquatic origin by count. These proportions were calculated defining all Odonata and Diptera as aquatic and all Hemiptera, Hymenoptera, Lepidoptera and Arachnida of terrestrial origin (Table 3.8). In 2005 boluses were identified to genus to determine origin, and weighed. Dry-weight biomass from 2005 was of slightly different proportions but all sites had greater than 50% aquatic biomass.

Discussion

PBDEs

Polybrominated diphenyl ethers are an emerging contaminant of concern that are neurotoxic, alter endocrine function and have been shown to impair fetal development in kestrels (*Falco sparverius*) (McKernan et al., 2009). Reference information for passerines is sparse at this time and the only reference to tree swallows I found was a sum of 81 ng/g ww (assuming 5.4% lipid in converting to ww units) in tree swallow eggs nesting near New Bedford Harbor, MA., with the majority of the PBDE sum comprised of BDEs 99 and 47 (Jayaraman et al.2008). Maximum mean concentrations of PBDEs in the eggs at the Calumet sites were similar (Powderhorn 2004: 78 ng/g) with lowest concentrations being about half the New Bedford value (47 ng/g at Big Marsh in 2004). Similar to the findings of Jayaraman et al. (2008), the greatest proportion of the sum PBDEs at the Calumet sites was comprised of BDEs 99 and 47. Studies in Europe with a similar species, the great tit (*Parus major*), yielded far lower egg concentrations for the sum of 7 PBDE congers: 4.19 to 6.85 ng/g ww (Dauwe et al., 2006), 0.4 to 13.6 ng/g (Van den Steen et al., 2009b), 20.4 ng/g ww (Van den Steen et al., 2009a) and 22 ng/g ww for the sum of 8 congeners (Voorspoels et al., 2007, assuming 10% egg lipid proportion from Voorspoels et al., 2007, and 5.4% nestling lipid from this study). One study had a range of 3 – 79 ng/g ww for the sum of 7 congeners (Van den Steen et al., 2008), the maximum of which is similar to the concentrations observed in New Bedford and Calumet swallows. Sum PBDE concentrations in bird eggs tend to be far greater in North America than Europe, and insectivorous species tend to have lower concentrations than piscivorous species. The concentrations in eggs of Calumet tree swallows were 19 times lower than sum PBDE concentrations in Lake Michigan herring gull (*Larus argentatus*) eggs (Gautier et al., 2008). These concentrations from Lake Michigan gulls may be the highest recorded in birds eggs (Chen and Hale, 2010) though when lipid normalized the greatest sum PBDE concentrations in bird eggs were found in osprey (*Pandion haliaetus*) eggs along the Willamette River (Henny et al., 2009), but wet weight geometric mean concentrations are about 14 times greater than those of Calumet swallow eggs.

Fifteen-day-old great tit nestlings were found to contain PBDE concentrations (the sum of 7 congeners) between 0.79 – 0.82 ng/g ww (Dauwe et al., 2006), which is approximately 20-70 times lower than the values in Calumet nestlings. Voorspoels and colleagues (2007) sampled great tit body fat and found mean concentrations of 117 ng/g ww for the sum of 8 PBDE congeners, about twice the concentration of PBDEs from whole nestlings at Indian Ridge in the study. Calumet nestling PBDE concentrations were comparable to the median concentration of the sum of about 10 PBDEs in muscle tissue of seven aquatic and terrestrial predatory birds in Belgium (range: 2.73 ng/g ww for kestrels, *Falco tinnunculus* to 50.4 ng/g ww for barn owls, *Tyto alba*) (Jaspers et al., 2006), but far below muscle concentrations of 141 ng/g ww in European sparrowhawks *Accipiter nisus* (Voorspoels et al., 2007). Concentrations were converted from lipid weight using lipid proportions listed in respective studies.

The only documentation I found of PBDEs in insects were for BDE 47, with concentrations in caterpillars in South Antwerp of 0.027 and 0.030 ng/g (Dauwe et al., 2006), being much lower than the mean concentrations of BDE 47 in terrestrial Calumet bolus insects (1.00 ng/g) and Calumet benthic insects (1.04 ng/g all sites) and far less than the mean of 3.09 ng/g in aquatic bolus insects.

Comparisons among different species are problematic because of different physiology, habits, and toxicokenetics. Moreover, when comparing the Calumet PBDE concentrations to concentrations from Europe it should be kept in mind that Europe banned penta and octa BDE compounds in 2004 (Van den Steen et al., 2009a). The urban location of the Calumet sites should be noted as well, since higher sum PBDE concentrations have been found in passerine eggs in urban areas (Van den Steen et al., 2009c). Lastly, the aquatic nature of a portion of the tree swallow diet may provide for different congener exposure than is provided by terrestrial insects (Law et al., 2003; Lindberg et al., 2004; Jaspers et al., 2006; Chen and Hale, 2010).

PCBs

Although PCB levels were highest in the “contaminated” sites it seems unlikely that these levels interfered with nesting and fledgling success. A number of other studies have reported tree swallow nestling sum PCB concentrations that are approximately ten fold greater than those I observed in the Calumet nestlings (Ankley et al., 1993; Custer et al., 1998; Custer et al., 2005; Maul et al., 2006; Neigh et al., 2006; Spears et al., 2008; Maul et al., 2010) and some studies reported concentrations 100 fold greater (Jones et al., 1993; Yorks, 1999; Custer et al., 2007b; Jayaraman et al., 2009). Calumet nestling concentrations were roughly 500 times lower than those found in nestlings living along the Hudson River (96,000 ng/g; Echols et al., 2004), which at this time are the highest recorded in tree swallow nestlings. Sum PCB nestling concentrations at Calumet were far lower than those of 2,272 ng/g ww in 15 day old nestlings in Saginaw Bay, MI., (Froese et al., 1998), and half that of 473 ng/g ww for 13 do nestlings in Southern Ontario (Papp et al., 2007) (assuming 14% lipid). Calumet nestling values were somewhat similar to those of nestlings along the Wisconsin River WI. (300 ng/g ww) (Custer et al., 2002) and concentrations found in nestlings sampled near Vancouver BC waste water treatment plants (104ng/g ww) (Dods et al., 2005).The maximum mean sum PCB concentrations in the Calumet nestlings were about 7 times that of nestlings roosting downstream of pulp mills in western Canada 31.6 ppb ww (Harris and Elliott, 2000). Effects on hatch success, such as there being more dead embryos or infertile eggs in a clutch with sum PCBs over 62,800 ng/g ww in eggs, (Custer et al., 2003) or endocrine disruption (114,000 ng/g in adults, McCarty and Secord, 1999a; McCarty and Secord, 2000) in tree swallows has been shown to occur at much greater concentrations than were found in the nestlings at Calumet. Therefore, Calumet swallows probably suffered few effects from the PCB loads they experienced.

The PCB concentrations in Calumet area tree swallows were relatively low. Sum PCB concentrations in tree swallow eggs in the Calumet region were more than two orders of magnitude less than the mean sum concentrations in eggs at the most contaminated sites along the Housatonic River, which are the highest yet found in tree swallow eggs (161,000 ng/g eggs; Custer et al., 2010a and 100,880ng/g ww for pipers; Custer et al., 2003). Calumet egg concentrations were about one order of magnitude lower than those found in the Hudson River (Echols et al., 2004), a Massachusetts superfund site (Jayaraman et al., 2009), a Michigan superfund site (Neigh et al., 2006), and near Green Bay, Wisconsin (Jones et al., 1993). Calumet concentrations in eggs were 4 to 10 times lower than found along the Hudson River (Custer et al., 2010b), or in a superfund site in southern Illinois (Spears et al., 2008; Maul et al., 2010) or near Green Bay WI (Ankley et al., 1993; Custer et al., 1998). Concentrations of sum PCBs in eggs from the Calumet sites were similar to or greater than concentrations found in a number of other studies (DeWeese et al., 1985; Elliott et al., 1994; Froese et al., 1998; Yorks, 1999; Custer et al., 2000; Custer et al., 2002; Custer et al., 2005). Ecological effects, such as higher levels of nest abandonment (McCarty and Secord, 1999b) were not observed in the Calumet nests (Chapter 2, this thesis for overall success; personal observation).

Tree swallows accumulate PCBs from their diet, so understanding their exposure through the concentrations found in local insects is useful for evaluating risks from contaminated local sediments. Total PCB concentrations documented in insects from other tree swallow studies were far greater than the range of 7 to 64 ng/g I found in bolus and benthic insects in the Calumet. Concentrations between 115 (caddisflies) to 630 (general gut contents) ng/g were found in southern Illinois (Maul et al., 2006), whereas samples from Michigan yielded a range between 90 – 682 ng/g (Nichols et al., 1995), but the greatest PCB concentrations in insects were between 70 and 18,800 ng/g and were collected in the Housatonic River study where the highest PCB concentrations have been found in tree swallow eggs (Custer et al., 2003).

Mean sediment concentrations of sum PCBs at the Calumet sites are greater than the National Oceanic and Atmospheric Administration's (NOAA) derived upper effects threshold (UET) (Buchman, 2008) which is the "lowest reliable concentration" from microtox bioassays below which no effects were observed (Buchman, 2008). Calumet sediment concentrations are also greater than the probable effects level (PEL) which delineates a PCB concentration above which adverse effects to sediment dwelling organisms are expected to frequently occur. These are screening values and should be interpreted as indications of potential sediment toxicity that requires further evaluation.

Local source nestling concentrations of 43,252 ng were found at a superfund site in southern Illinois (Maul et al., 2010) which were over an order of magnitude higher than local source concentrations found in the Calumet.

Observed PCB concentrations are low in the media collected from the Calumet area sites. This may help explain why the egg contribution to nestling PCB mass was approximately 50%, if local concentrations could not dilute the PCB mass in the egg. Although concentrations are low, uptake is occurring, but the risk to tree swallows is low.

Organochlorine Pesticides

Although DDT has been banned for general use in North America since the early 1970s, its breakdown products DDD and DDE were the major pesticide residues in sediments, insects, tree swallow eggs and nestlings at the Calumet sites. Mean DDD concentrations in the Calumet nestlings were generally much higher than the range of concentrations found in old agricultural areas within Point Pelee National Park, Canada (4 – 8 ng/g) (Smits et al., 2005). Maximum DDE concentrations in Point Pelee nestlings were higher than Calumet concentrations by about 40%, as were concentrations in nestlings sampled from a various wetlands in southern Ontario (22-559ng/g) (Bishop et al., 1995). Concentrations of DDD and DDE in tree swallow nestlings have been associated with terrestrial prey (Smits et al., 2005); however, I did not find a difference in bolus concentrations relative to terrestrial or aquatic origin at the Calumet sites.

The DDD and DDE concentrations in tree swallow eggs from the Calumet area were generally low and were much lower than in those collected from orchards in British Columbia (Elliott et al., 1994). Maximum concentrations of DDE in tree swallow eggs in BC orchards were almost two orders of magnitude greater than those of Calumet eggs. Concentrations of DDD in tree swallow eggs in Calumet were within the range found in BC orchard eggs; however, the maximum concentrations in BC eggs were approximately three times greater than the largest mean DDD concentration in Calumet swallow eggs. Concentrations of DDE in tree swallow eggs in Southern Ontario orchards were two to ten times greater than the concentrations I found in Calumet tree swallow eggs (Mayne et al., 2005). Mean maximum DDE concentrations in pipers (1 or 2- day old nestlings) collected along the Housatonic River were twice as high as mean maximum concentrations from the Calumet area (Custer et al., 2003).

The DDE concentrations in diet samples from the literature range as high as 65 ng/g along the Housatonic R. (Custer et al., 2003) to concentrations of 0.37 ng/g in caterpillars from Belgium (Dauwe et al., 2006). Calumet insects sampled contained DDE concentrations ranging from 1 to 56 ng/g.

Of the five sediment samples in which DDT was detected, two samples had DDT concentrations greater than 100 ng/g dw, which is greater then the PEL, above which adverse effects on sediment dwelling organisms are expected to occur (Buchman, 2008). All detected sediment DDD sample concentrations exceeded the PEL, and two exceed the severe effects level (SEL), above which sediments are considered severely polluted, and adverse effects on the majority of sediment dwelling organisms is to be expected. Observed DDE concentrations exceeded PEL concentrations in three samples and probable effects concentrations (PEC) in two samples (MacDonald et al., 2000). Calumet sediment samples did not uniformly exceed effects threshold levels, and these variable sediment concentrations help explain the lower concentrations in Calumet insect and tree swallow samples.

Eggs vs Nestlings

Sum PBDE concentrations in the Calumet tree swallow eggs were consistently dominated by BDEs 47 and 99 in both years and among the three sites. These two congeners are most abundant in 2004 nestlings, but some of the nestlings at Powderhorn from 2004 also have a preponderance of BDE 183 in addition to 47 and 99. Nestlings from 2005 had a slightly different profile with BDEs 47, 99 and 183 as most abundant, but with BDE 153 as a strong contributor to overall sum PBDE mass as well. This difference of profiles between eggs and nestlings suggests active uptake of higher congeners by nestlings at the Calumet sites and metabolism of the higher congeners to lower ones in the eggs. PCB profiles were similar through both years and among sites as well as between eggs and nestlings. Congeners 84&101, 118, 138&163, 153 and 180 were dominant. OC pesticides did not follow a general pattern when comparing between egg and nestling profiles.

LocalSource

Bolus insect count and biomass suggested that tree swallows in the Calumet area consumed a variable diet of insects from both aquatic and terrestrial sources. When there was an emergence of weevils (Curculionidae), the terrestrial beetles were found in high numbers in boluses, reinforcing how opportunistic the swallows are with their food choice. Diet samples are an accurate “snapshot”, of the actual food the nestling is eating; however, they only provide periodic confirmation of diet items so some caution should be exercised in drawing conclusions based on these snapshot results.

The proportion of contaminant mass in Calumet nestlings that is attributable to the egg varied tremendously depending on the compound. Eggs accounted for approximately one fifth of nestling PBDE mass and about one half of the nestling PCB mass. There was a great deal of variation in this measurement and is likely due in part to the fact that the eggs are merely from the same nest as the nestling that was analyzed as well as the mathematical inaccuracies of using mean egg moisture values in conversions. Nonetheless, these proportion estimates are useful in providing an estimate regarding the source of persistent contaminants found in nestlings of migratory species. Examining local source contaminant mass in nestlings highlighted that PBDEs in Calumet nestlings are mostly acquired locally. The PCB results differed between nestling and local source as did those for the OCs. Eggs contributed greater than 30% of the mass of these contaminants, altering the interpretation of the nestling results alone. These differences between nestling and local source results should be considered when researchers are interested in site specific understanding of contaminant availability.

Local risk

Approximately 20% of the PBDE contaminant mass in nestlings was attributed to the egg, confirming local uptake in the Calumet region. Eggs had fairly similar concentrations and profiles, but nestlings acquired different amounts in their local diet depending on which site they were collected from. Big Marsh and Indian Ridge both posed the greatest risk of uptake among the Calumet sites, but, concentrations in tree swallow nestlings in the Calumet region were low, compared to piscivorous or predatory species of North American birds. Comparing Calumet nestlings to insectivorous and predatory birds in Europe, concentrations may be considered intermediate. Concentrations of PBDEs have been noted to cause growth effects, oxidative stress and thyroid, vitamin A, glutathione homeostasis and immunomodulatry changes in developing kestrels with egg BDE 71 concentrations of 18,700ng/g (cited in Chen and Hale, 2010), though a recent study with chicken (*Gallus gallus*), mallard (*Anas platyrhyncho*s), and kestrel (*Falco sparverius*) eggs suggested a lowest-observed-effect level (LOEL) of pentaBDEs, to be 1,800 ng/g egg wet weight (McKernan et al., 2009). This is approximately 45 times the measured pentaBDE concentrations in the Calumet tree swallow eggs. In contrast, it was suggested that a concentration of about half the LOEL reported by McKernan et al. may impact reproductive success of wild osprey (*Pandion haliaetus*) eggs in the Pacific northwest of the

USA (Henny et al. 2009); however, this concentration is still far higher than what is seen in Calumet tree swallow eggs.

Concentrations of PCBs in nestlings at the Calumet sites were acquired equally from maternal egg input and from their diet. Big Marsh posed the greatest risk for uptake among the three sites, but the concentrations were nonetheless low. Laboratory studies suggest LD50s in various bird species with diet PCB concentrations ranging from mid 60 to 6,000 ng/g diet (Rice et al., 2003). Calumet bolus insects had sum PCB concentrations of 64 ng/g. The concentrations available at the Calumet sites may pose problems for sensitive birds; however, there is a wide range of toxicity of different PCB congeners, and PCB toxicity of 60 ng/g would likely depend which congeners contributed to the sum value. The most toxic congener (e.g. 126) was not found in the Calumet nestlings. Brain concentrations of 310,000 ng/g were considered diagnostic of PCB induced mortality in altricial birds (Rice et al., 2003). This is about 2000 times greater than the concentrations in Calumet swallow nestlings.

The OC pesticide exposure varied among the sites, with greatest risks from DDE at Big Marsh, and from DDD at Big Marsh and Indian Ridge, whereas nestlings at Powderhorn were exposed to more dieldrin and trans-nonachlor. Concentrations of DDE in tree swallow eggs were about a tenth of the concentration which causes almost complete reproductive failure in brown pelicans (*Pelecanus occidentalis*), the avian species most sensitive to DDE (Blus, 2003). Concentrations of DDD in eggs that were associated with greatly reducing a population of western grebes (*Aeschmorphorus occidentalis*) in California (Blus, 2003) were about 250 times that of the Calumet swallow eggs. Reduced productivity was found in shags (*Phalacrocorax aristotelis*) when egg dieldrin concentrations were 100 times that of Powderhorn tree swallow egg concentrations (Blus, 2003).

Conclusions

Tree swallow eggs and nestlings in the Calumet area of Illinois had low concentrations of a variety of organic pollutants. Sediment samples from the Calumet sites exceeded a number of contaminant screening criteria. Nonetheless, existing literature does not indicate that observable reproductive effects would occur in tree swallows at the concentrations of these Calumet contaminants. Diet concentrations of sum PCBs may be considered high for sensitive species. Diet and sediment sample analyses did not confirm terrestrial or aquatic contaminant origin in the Calumet tree swallows. Using a normalizing function to determine local contaminant mass yielded different results and more insight into local uptake than only evaluating nestling contaminant concentrations. Localized contaminant uptake occurred and could be an issue for PBDEs, concentrations of which are trending higher through time in bird species in North America (Chen and Hale, 2010). As far as I have determined, this is the first published account of PBDE concentrations in tree swallow eggs and nestlings.

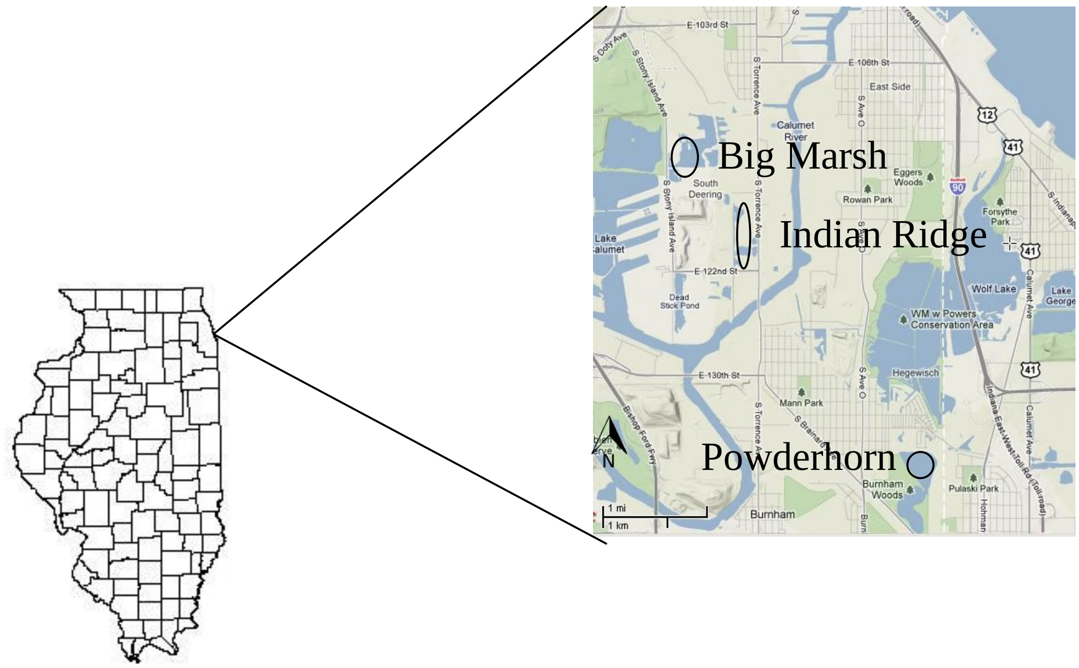

Figure 3.1: Map of Illinois, USA with expanded view of study sites.

Table 3.1. Test statistics and p values for PBDE concentration statistical comparisons for all media from three wetlands in the Calumet area, IL.

Variable	Comparison	Test statistic	p
Bolus BDE 71	origin	t=2.96	0.060
Bolus BDE47	origin	t=1.27	0.296
Bolus BDE99	origin	t=0.55	0.636
Egg sum	years	F=0.83	0.367
Egg sum 2004 and 2005	site	F=1.85	0.166
Egg suite	years	R=0.10	0.021
2004	sites	R=0.04	0.166
2005	sites	R<0.01	0.385
Nestling sum	years	H=10.42	0.001
2004	sites	H=5.67	0.059
2005	sites	F=6.81	0.005
BM vs PL	sites	F=8.62	0.007
IR vs PL	sites	F=12.05	0.002
Nestling suite	years	R=0.55	<0.001
2004	sites	R=0.26	0.001
BM vs PL	sites	R=0.55	0.001
IR vs PL	sites	R=0.27	0.005
2005	sites	R=0.32	0.001
BM vs PL	sites	R=0.45	0.003
IR vs PL	sites	R=0.60	0.001
Local-sourced sum	years	F=8.34	0.006
2004	sites	F=2.34	0.117
2005	sites	F=7.87	0.003
BM vs PL	sites	F=10.14	0.004
IR vs PL	sites	F=13.61	0.001

Table 3.2. Mean (± SD when available) sum PBDE concentrations (ng/g wet weight), maximum, % lipid in nestlings[1] and mean local-sourced[2] sum PBDE mass (ng) in nestlings, and sample size (n) at three sites in Calumet, Illinois, for 2004 and 2005.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
eggs[1,3]	46.8 ± 13.3	55.7 ± 44.8	74.0 ± 56.9	63.4 ± 45.7	78.5 ± 44.6	60.0 ± 25.7
max (n)	71.6 (10)	160.3 (11)	213.0 (10)	164.9 (10)	176.7 (9)	105.2 (11)
nestlings	23.3 ± 3.9	51.0* ± 26.4	30.0 ± 14.7	62.5* ± 56.7	20.4 ± 7.9	22.5 ± 10.1
max	28.8 (11)	100.3 (9)	60.5 (11)	218.7 (10)	38.9 (8)	42.6 (9)
% lipid	8.9 ±2.1	20.3 ± 5.6	7.2 ± 1.9	19.7 ± 6.0	9.9 ± 1.4	19.0 ± 3.8
Local-sourced	452 ± 95	1062* ± 611	519 ± 241	1212* ± 1016	363 ± 211	389 ± 229
max (n)	601 (10)	2280 (9)	1084 (10)	3962 (9)	845 (7)	824 (9)

[1] the pooled % lipid value for eggs across all years and sites was 5.41
[2]Local –sourced sum PBDE mass = mass of sum PBDE in nestlings minus mass of sum PBDE in eggs from same nest box.
[3]Yearly values were pooled for statistical analysis.
* Indicates values are significantly different from the reference site, Powderhorn; p<0.05. Values below mean and SD is maximum concentrations observed, percent lipid or TOC and sample size.

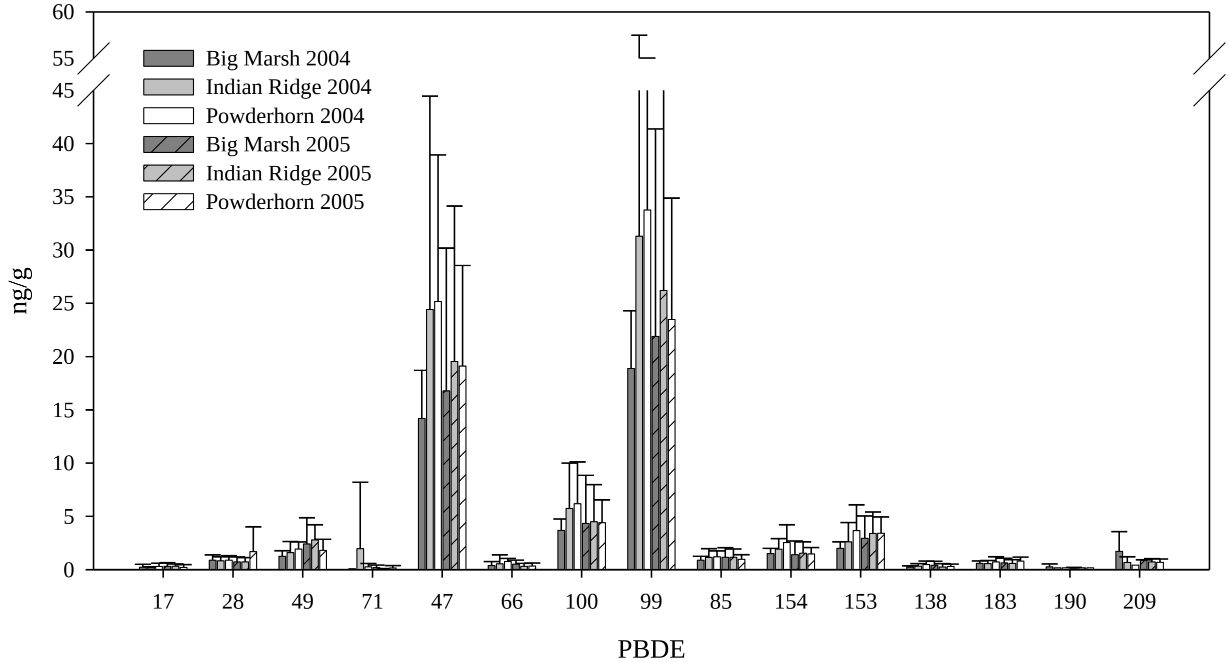

Figure 3.2. PBDE congener concentrations in tree swallow eggs (ng/g wet weight) for 2004 and 2005 at three sites in Calumet, Illinois.

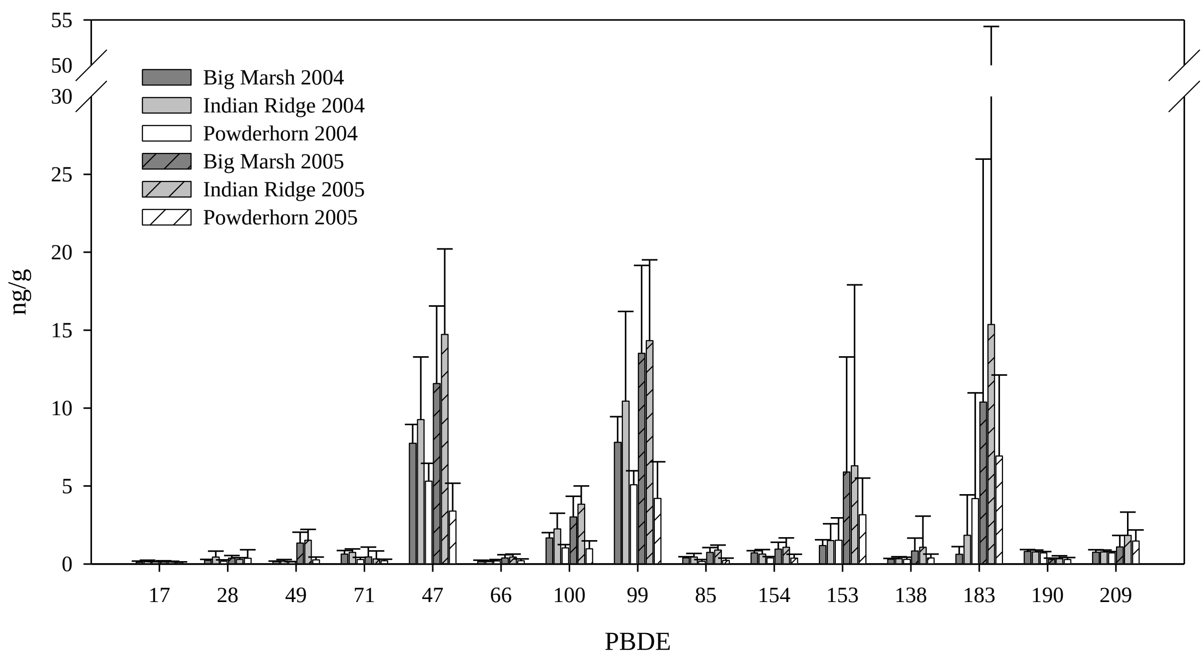

Figure 3.3. PBDE congener concentrations in tree swallow nestlings (ng/g wet weight) for 2004 and 2005 at three sites in Calumet, Illinois.

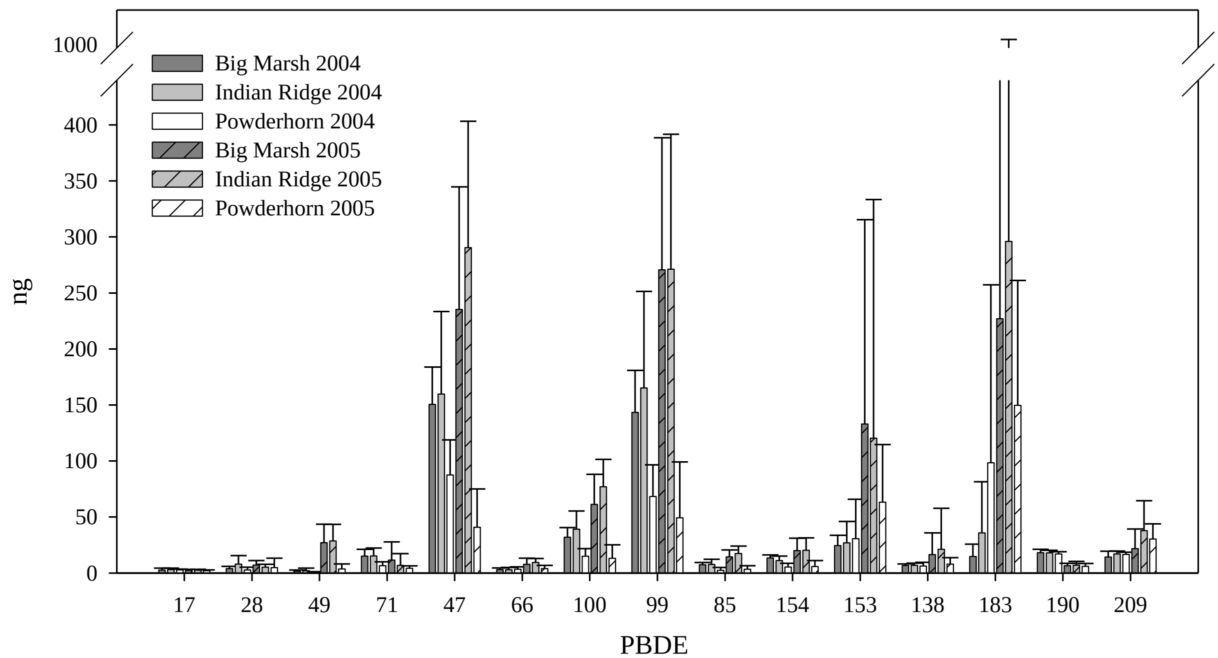

Figure 3.4. Local-sourced PBDE mass in nestlings for 2004 and 2005 at three sites in Calumet, Illinois. Local –sourced PBDE mass = mass of PBDE congener in nestlings minus mass of PBDE congener in eggs from same nest box.

Table 3.3. Test statistics and p values for PCB concentration statistical comparisons for all media from three wetlands in the Calumet area, IL.

Variable	Comparison	Test statistic	p
Sediment sum	sites	H=0.60	0.741
Sediment suite	sites	R=-0.15	0.859
Benthic sum IR vs PL	sites	t=-0.63	0.571
Bolus sum	origin	F=0.01	0.916
Bolus suite	origin	R=-0.17	0.600
Eggs sum	years	H=0.52	0.470
Eggs sum pooled	sites	H=6.38	0.041
IR vs PL	sites	Q=2.52	<0.05
Eggs suite	years	R=0.30	0.001
2004	sites	R<0.00	0.446
2005	sites	R=0.192	0.002
BM vs PL	sites	R=0.26	0.003
IR vs PL	sites	R=0.26	0.008
Nestling sum	years	F=2.65	0.109
Nestling sum pooled	sites	F=10.65	<0.001
BM vs PL	sites	F=19.23	<0.001
Nestling suite	years	R=0.19	0.001
2004	sites	R=0.43	0.001
BM vs PL	sites	R=0.80	0.001
IR vs PL	sites	R=0.24	0.014
2005	sites	R=0.16	0.006
BM vs PL	sites	R=0.16	0.040
IR vs PL	sites	R=0.16	0.020
Local-source	years	F=4.72	0.034
Local-source 2004	sites	F=6.14	0.007
BM vs PL	sites	F=12.22	0.002
IR vs PL	sites	F=5.10	0.033
Local-source 2005	sites	F=8.86	0.001
BM vs PL	sites	F=15.77	<0.001

Table 3.4. Mean (± SD when available) sum PCB concentrations (ng/g wet weight or ng/g dry weight for sediment), % lipid[1], and mean local-sourced[2] sum PCB mass (ng) in nestlings at three sites in Calumet, Illinois, for 2004 and 2005. Values below mean and SD are maximum concentrations observed, the number of samples (n) and percent lipid or TOC.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
sediment	50 ± 17	nm	877 ± 1674	nm	62 ± 58	nm
TOC (n)	5 ± 4 (3)		9 ± 8 (4)		11 ± 7 (3)	
ben. ins.	nm	42.7	17.2	7.6 ± 2.8	27.2	9.3 ± 4.0
% lipid (n)		3 (1)	4 (1)	2 ± 0 (2)	3 (1)	2 ± 1 (2)
bol. ins.	nm	33.6 ± 9.1	nm	64.3 ± 9.9	nm	56.5
% lipid (n)		9.0 ± 0 (2)		13.5 ± 3.5 (2)		8.0 (1)
eggs[1,3]	605 ± 307	482 ± 128	498 ± 287	428 ± 199	519 ± 250	1084 ± 748
max (n)	1215 (10)	715 (11)	1226 (10)	955 (10)	947 (9)	2731 (11)
nestlings[3]	207.6 ± 53.4	179.1 ± 69.5	164.7 ± 77.7	109.8 ± 25.6	104.5 ± 17.9	132.8 ± 63.8
max (n)	311 (11)	325 (9)	379 (11)	154 (10)	129 (8)	244 (9)
% lipid	8.9 ±2.1	20.3 ± 5.6	7.2 ± 1.9	19.7 ± 6.0	9.9 ± 1.4	19.0 ± 3.8
Local-sourced[2]	3472* ± 809	3434* ± 1714	2904* ± 1153	1578 ± 566	1866 ± 707	1128 ± 1138
in nestlings (max)	4827 (10)	6999 (9)	5083 (10)	2345 (9)	3112 (7)	3194 (9)

nm = not measured. ben. ins = benthic insects. bol. ins. = bolus insects.
* Indicates values are significantly different from the reference site, Powderhorn; $p<0.05$.
[1] the pooled % lipid value for eggs across all years and sites was 5.41
[2] Local –sourced sum PCB mass = mass of sum PCB in nestlings minus mass of sum PCB in eggs from same nest box.
[3] Yearly values were pooled for statistical analysis.

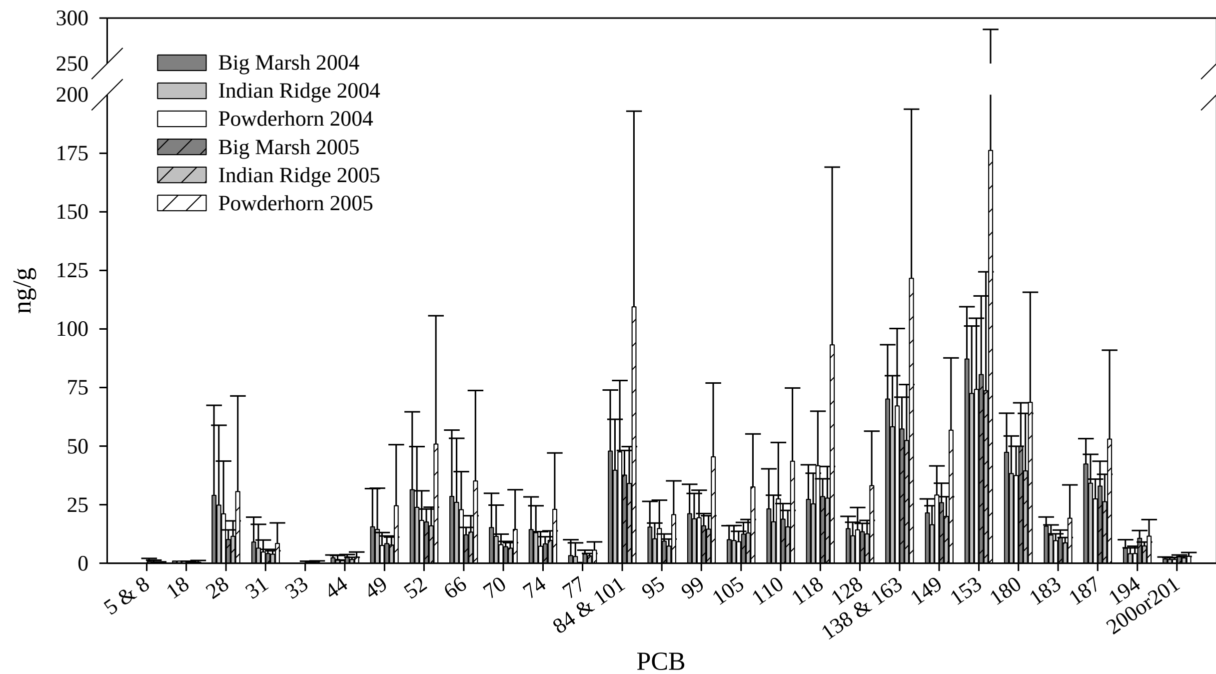

Figure 3.5. Profile of PCB congener concentrations in tree swallow eggs (ng/g wet weight) for 2004 and 2005 at three sites in Calumet, Illinois.

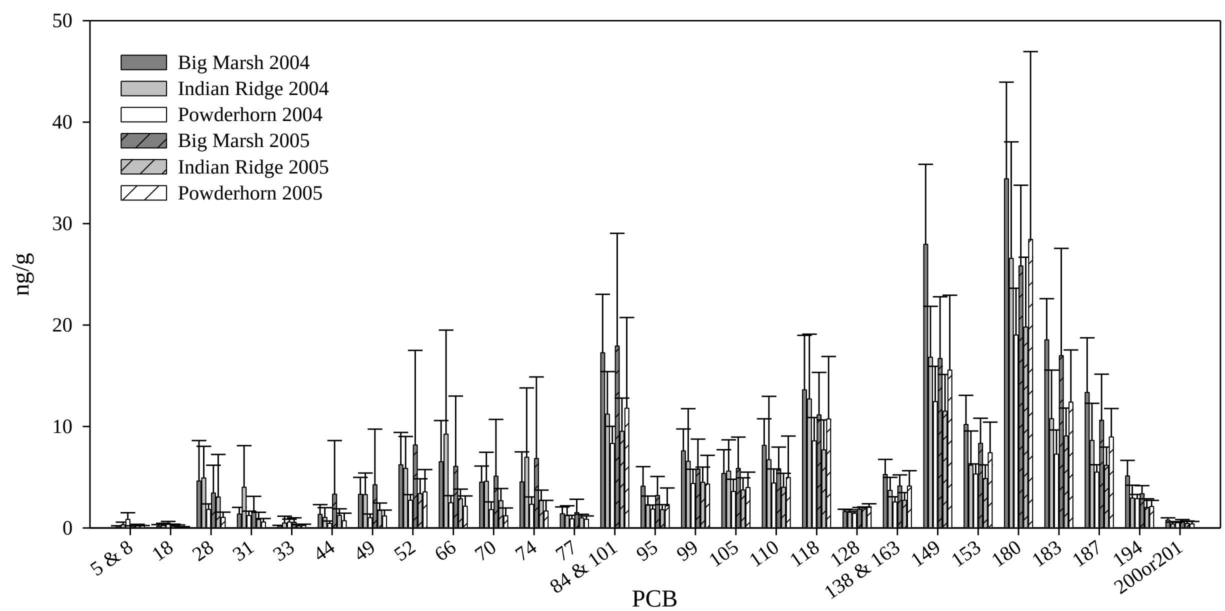

Figure 3.6. Profile of PCB congener concentrations in 14-day-old nestling carcasses in 2004 and 2005 at three sites in Calumet, Illinois.

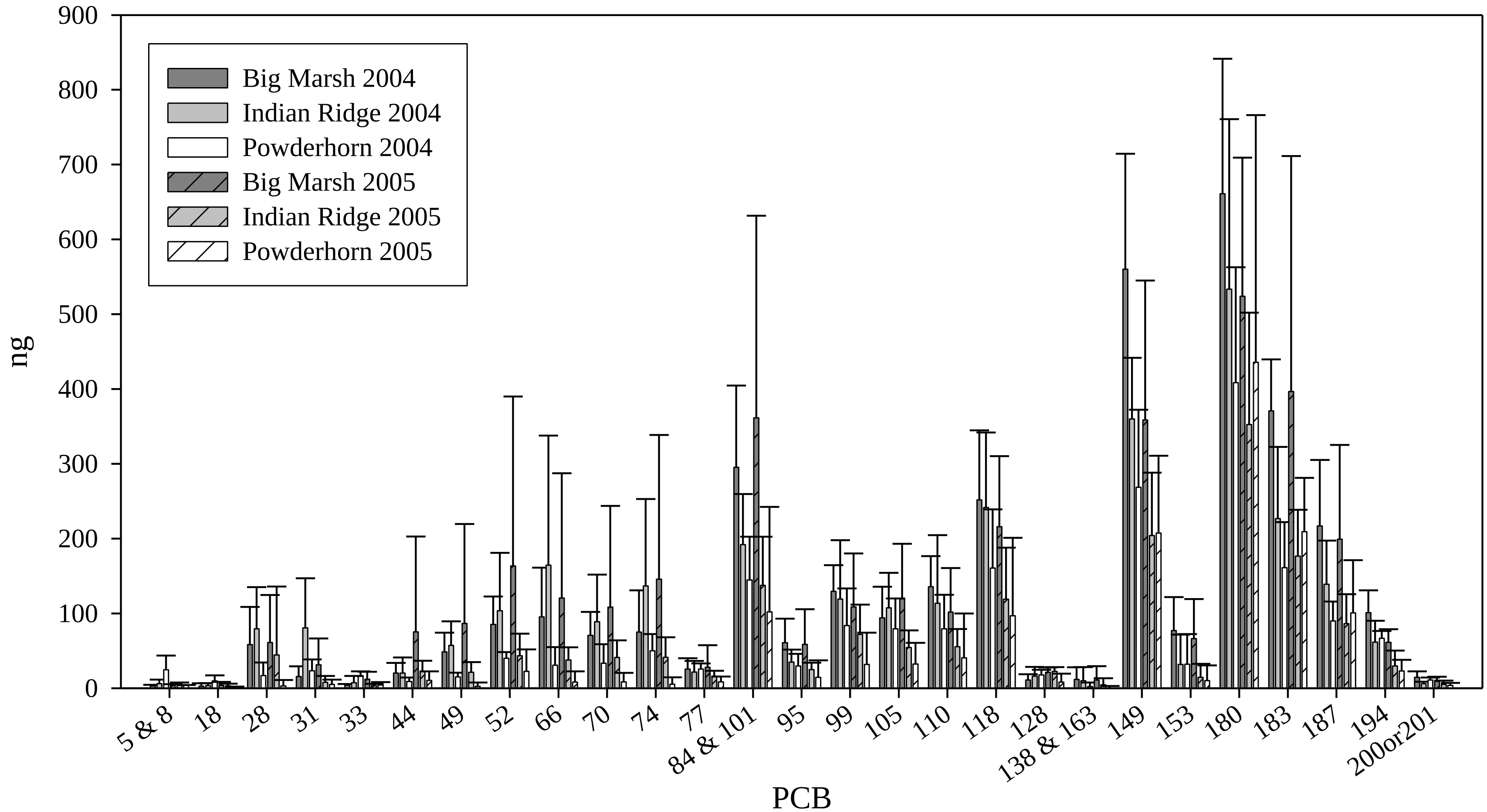

Figure 3.7. Local-sourced PCB mass in nestlings for 2004 and 2005 at three sites in Calumet, Illinois. Local –sourced PCB mass = mass of PCB congener in nestlings minus mass of PCB congener in eggs from same nest box.

Table 3.5. Mean (±SD) / maximum DDD, DDE and DDT concentrations (ng/g ww or ng/g dw for sediment) in select media from three sites at Lake Calumet area wetlands in 2004 and 2005.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
sediments (n)	3		4		3	
4,4'-DDD	~	NM	189.8 ± 361.5	NM	113.0 ±155.4	NM
	13		732		292	
4,4'-DDE	6.4 ± 8.5	NM	13.3 ± 21.2	NM	20.7 ± 31.5	NM
	16		45		57.0	
benthic insects (n)	0	1	1	2	1	2
4,4'-DDE	NS	56.0	13.1	7.4 ± 7.9	6.5	4.1 ± 2.0
				13.0		5.5
bolus insects (n)		2		2		1
4,4'-DDD	NM	4.5± 5.8[2]	NM	26.7 ± 21.4	NM	5.4
		8.6		41.8		
4,4'-DDE	NM	9.2 ± 8.8	NM	12.4 ± 8.1	NM	1.0
		15.4		18.1		
eggs (n)	10	11	10	10	9	11
4,4'-DDD	205* ± 1718	218* ± 132	64* ± 54	109 ± 937	18 ± 16	74 ± 81
	501	510	169	334	51	282
4,4'-DDE[1]	371 ± 99	414 ± 180	271 ± 104	261 ± 121	182 ± 56	368 ± 252
	565	862	452	581	298	845
4,4'-DDT[1]	1.0 ± 1.0	1.2 ± 1.0	1.8± 2.5	1.3 ± 0.7	1.5 ± 0.7	2.1 ± 1.3
	3.1	3.3	7.6	2.9	2.7	5.4
dieldrin[1]	2.7 ± 3. 5	7.1 ± 7.2	4.8 ± 2.6	7.1 ± 6.4	13.8 ± 8.9	34.2 ± 33.5
	9.2	26.4	8.4	22.9	28.8	103.8
tnonachlor	8.4 ± 10.1	11.7* ± 10.9	8.5 ± 5.4	11.0* ± 8.2	9.7 ± 4.3	63.5 ± 121.6
	36.5	38.7	19.1	24.64	14.8	422.4
HPX+OXC	19.4* ± 6.9	29.5* ± 17.7	24.8* ± 10.6	31.6* ± 19.7	36.3 ± 17.4	66.9 ± 46.3
	34.1	61.6	40.7	66.9	67.9	193.6

Table 3.5. (cont.)

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
nestlings (n)	11	9	11	10	8	9
4,4'-DDD[1]	230 ± 184	231 ± 165	73 ± 70	71 ± 48	3.4 ± 3.4	53 ± 147
	623	580	234	171	11	444
4,4'-DDE	179* ± 93	116* ± 53	79 ± 32	66 ± 20	71 ± 35	51 ± 19
	340.0	226.3	140.4	105.4	129.0	81.4
dieldrin	3.6* ± 1.3	18.9 ± 35.6	36.5 ± 68.0	6.3* ± 2.1	6.4 ± 1.5	15.3 ± 10.6
	5.8	113.4	180.9	9.9	8.7	37.13
trans-nonachlor	2.0* ± 0.5	15.6 ± 36.7	8.0 ± 12.6	4.1 ± 2.2	4.05 ± 1.02	21.2 ± 45.8
	2.7	113.4	38.8	8.8	5.3	142.8

[1]Years were pooled for statistical analysis
[2]One sample was lost and value was replaced with half of detection limit.
* Indicates mean is significantly different from reference site, Powderhorn; $p<0.05$.
NM = not measured

Table 3.6. Test statistics and p values for OC pesticide concentration statistical comparisons for all media from three wetlands in the Calumet area, IL. Most comparisons that are not significant are not shown.

Variable	Comparison	Test statistic	p
Sediment DDD	sites	F=4.07	0.067
Sediment DDE	sites	F=0.43	0.669
Benthic DDE IR vs PL	sites	t=1.12	0.362
Bolus DDE	origin	t=1.19	0.336
Eggs DDE	sites	F=5.80	0.005
BM vs PL	sites	F=10.01	0.003
Eggs dieldrin	sites	H=23.15	<0.001
BM vs PL	sites	Q=4.56	<0.05
IR vs PL	sites	Q=3.62	<0.05
Eggs DDT	sites	F=4.82	0.012
BM vs PL	sites	F=9.60	0.003
Eggs trans-nonachlor	years	H=4.18	0.041
2005	sites	F=6.04	0.006
BM vs PL	sites	F=8.06	0.008
IR vs PL	sites	F=6.75	0.015
Eggs DDD	years	F=5.06	0.028
2004	sites	F=19.31	<0.001
BM vs PL	sites	F=8.63	<0.001
IR vs PL	sites	F=10.60	0.003
2005	sites	F=7.90	0.002
BM vs PL	sites	F=15.72	<0.001
Eggs Sum HPX+OXC	years	F=4.89	0.031
2004	sites	F=4.72	0.018
BM vs PL	sites	F=9.15	0.006
IR vs PL	sites	F=4.23	0.005
2005	sites	F=4.72	0.018
BM vs PL	sites	F=9.97	0.004
IR vs PL	sites	F=7.89	0.009

Table 3.6. Cont.

Variable	Comparison	Test statistic	p
Nestling dieldrin	years	H=6.76	0.009
2004	sites	H=8.67	0.013
BM vs PL	sites	Q=2.73	<0.05
2005	sites	H=7.12	0.029
IR vs PL	sites	Q=2.63	<0.05
Nestling Trans-nonachlor	years	H=4.95	0.026
2004	sites	H=10.37	0.006
BM vs PL	sites	Q=3.21	<0.05
Nestling DDE	years	F=4.36	0.041
2004	sites	F=9.43	<0.001
BM vs PL	sites	F=14.71	<0.001
2005	sites	F=10.44	<0.001
BM vs PL	sites	F=20.33	<0.001
Nestling DDD	sites	H=32.81	<0.001
BM vs PL	sites	Q=5.72	<0.05
IR vs PL	sites	Q=3.38	<0.05
Local-source DDE	years	H=7.10	<0.001
2004	sites	F=3.80	0.037
2005	sites	H=14.40	<0.001
BM vs PL	sites	Q=3.81	<0.05
Local-source dieldrin	years	H=4.58	0.032
2004	sites	H=7.72	0.021
BM vs PL	sites	Q=2.65	<0.05
Local-source DDD	sites	H=27.39	<0.001
BM vs PL	sites	Q=5.22	<0.05
IR vs PL	sites	Q=3.08	<0.05

Table 3.7. Mean (± SD) / maximum ng local-sourced pesticide mass of select pesticides in nestlings for 2004 and 2005 at three sites in Calumet, Illinois.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
4,4'-DDD[1]	4771 ± 4655 15086	5052 ± 3966 13366	1436 ± 1600 5105	1551 ± 1251 4139	200 ± 377 1109	763 ± 2229 6706
4,4'-DDE	3215 ± 2251 8097	1952* ± 1172 4509	1313* ± 743 2429	989 ± 443 1650	1578 ± 755 2649	443 ± 444 1221
dieldrin	74* ± 25 110	456 ± 897 2836	143 ± 113 333	353 ± 775 2685	608 ± 1340 3922	241 ± 174 554
tnonachlor[1]	32 ± 18 69	371 ± 922 2825	47 ± 28 85	99 ± 114 420	177 ± 263 824	318 ± 745 2295

[1]Yearly values were pooled for statistical analysis.
* indicates mean is significantly different from reference site, Powderhorn; $p<0.05$.

Table 3.8. Mean ± SD number of individuals per bolus of dominant invertebrate orders in 2004 bolus samples. *mostly araneae.

	Big Marsh		Indian Ridge		Powderhorn	
	2004	2005	2004	2005	2004	2005
	19	68	6	28	18	25
Diptera	4.6 ± 7.9	13 ± 16.4	5.7 ± 5.1	9.8 ± 11.1	4.3 ± 5.4	10.2 ± 16.1
Odonata	1.1 ± 1.3	0.5 ± 0.8	0.7 ± 1.1	0.3 ± 0.5	5 ± 5.7	3.7 ± 2.7
Coleoptera	11.1 ± 19.0	0.8 ± 1.4	35.3 ± 47.9	1.5 ± 3.7	5.5 ± 11.2	1.2 ± 1.7
Hemiptera	1.4 ± 2.8	3.2 ± 6.1	2.7 ± 2.6	1.1 ± 1.9	0.6 ± 1.1	3.3 ± 4.5
Hymenoptera	0.8 ± 1.6	4.9 ± 11.8	2.8 ± 5.0	2.0 ± 3.7	0.2 ± 0.5	2.0 ± 4.4
Other	na	1.4 ± 4.6*	0.2 ± 0.5	0.5 ± 1.6	0.1 ± 0.4	1.1 ± 5.2
Aquatic, by count, %	30	54	13	69	58	65
Aquatic, by mass, %	na	52	na	51	na	64

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APPENDIX

ABBREVIATIONS

Abbreviations

AB = Alberta
ALHs aliphatic hydrocarbon
BROD = benzyloxyresorufin O-dealkylase
Cd = cadmium
CHC = chlorinated hydrocarbons
ChE = cholinesterase
Co = county
CO = Colorado
Cr = chromium
Cu = copper
DDD = dichlorodiphenyldichloroethane
DDE = dichlorodiphenyldichloroethylene
DDT = dichlorodiphenyltrichloroethane
DNA = deoxyribonucleic Acid
DO = day old
ELARP = experimental Lakes Area Research Project
EROD = ethoxyresorufin-O-deethylase ethoxyresorufin O-deethylase
GLSLRB = Great Lakes St Lawrence River Basin
Hg= mercury
HpCDD = heptachlorodibenzodioxin
HpCDF = heptachlorodibenzofuran
HPCV = half-peak coefficient of (nuclear DNA) variation
HxCDD = hexachlorodibenzodioxin
HxCDF = hexachlorodibenzofuran
Ni = nickel
NP = national park
OC = organochlorine
OCDD = octachlorodibenzodioxin
OCDF = octachlorodibenzofuran
OP = organophosphorus
NWR = national wildlife refuge
PAH = polycyclic aromatic hydrocarbon
Pb = lead
PCB = polychlorinated biphenyl
PCDD = polychlorinated dibenzo-p-dioxin
PCDF = polychlorinated dibenzofurans

Abbreviations (continued)

PeCDD = pentachlorodibenzodioxin
PeCDF = pentachlorodibenzofuran
SLRB = Saint Lawrence River Basin
TCDD = tetrachlorodibenzo-p-dioxin
TCDD-EQ = tetrachlorodibenzo-p-dioxin equivalent
TCDF = tetrachlorodibenzofuran