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Preliminary findings of contaminant screening of Maine bird eggs

2007 Field Season

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Preliminary findings of contaminant screening of Maine bird eggs:

2007 Field Season

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1. EXECUTIVE SUMMARY AND PRIMARY FINDINGS

Starting in May 2007, BioDiversity Research Institute (BRI) and collaborators initiated a broad-based contaminant study on Maine birds, measuring both historical and emerging chemicals. This comprehensive project measured 192 synthetic contaminants in 23 species across Maine to determine in which species, habitats, and locations these anthropogenic compounds are concentrating. The compounds we analyzed in 60 egg composites were mercury (Hg), polychlorinated biphenyls (PCB), polybrominated diphenyl ethers (PBDE), perfluorinated compounds (PFCs), and organochlorine pesticides (OCs). Our preliminary findings are:

- Hg, PCBs, PBDEs, PFCs, and OCs are found in all species sampled across marine, estuarine, riverine, lacustrine (lake), and terrestrial ecosystems; these are the first records of PFCs in Maine birds.
- Hg, PCBs, PFCs are all found at levels that may cause adverse effects—there are currently no established adverse effects thresholds established for PBDEs in bird eggs. OCs are all significantly below adverse effects thresholds.
- Our Hg, PCB, and OC levels were generally consistent with levels recorded around the country. Certain species had PBDEs higher than other locations, while other species had lower levels. PFOS have not been widely studied in eggs; therefore, we could not directly compare our results to other areas.
- The total PCBs levels we recorded are lower than those in the past, indicating a continued decline in PCBs.
- Bald eagles have the highest overall contaminant load of the 23 species measured.
- We found all of the compounds across the entire state, but overall contaminant loading tends to be highest in southern coastal Maine. This geographic pattern suggests that these compounds are entering the environment both through atmospheric deposition, because they are found across the entire state, and through local point sources, because we detected higher levels in urban and industrial areas.
- PCBs, PBDEs, PFCs, and OCs levels are positively correlated, indicting that birds with high levels of one compound tend to have higher levels of the others. PBDEs and PCB have the strongest relationship.
- Birds that feed on terrestrial prey accumulated higher brominated PBDEs; DecaBDE is found in eight species with gulls and peregrine falcon having the highest levels.
- Of the samples we analyzed, birds feeding in estuaries have the lowest contaminant levels.
- The mouth of the Kennebec and Isles of Shoals tended to have high concentrations of contaminants.

2. INTRODUCTION

2.1 Project overview

Starting in May 2007, BioDiversity Research Institute (BRI) and collaborators initiated a broad-based contaminant study on Maine birds, measuring both historical and emerging chemicals. This comprehensive project measured 192 synthetic contaminants in 23 species across Maine to determine in which species, habitats, and locations these anthropogenic compounds are concentrating. The chemicals we analyzed in 60 egg composites were mercury, polychlorinated biphenyl (PCB) congeners, polybrominated diphenyl ether (PBDE) congeners, perfluorinated compounds (PFCs; e.g., PFOS, PFOSA, PFHxS, PFOA, PFNA, PFDA, PFDoDA, PFUnDA, PFHxA, PFHpA), and organochlorine pesticides (OCs) (DDTs, HCHs, chlordanes, HCB).

The project had two components. The first was evaluating geographic differences by analyzing eggs of seven marine species from six sites near the outflows of Maine's largest rivers (Figure 1). Since studies indicate that levels of PCBs and other organics in eagles are higher along the coast than inland (Matz 1998), and contaminants bioaccumulate[1] in coastal cormorants (Mower 2006), terns, and plovers (Mierzykowski and Carr 2004), we focused geographic contaminant screening along the coast. We selected sites near the largest river outflows and areas of high population density. The sites were: 1) Isles of Shoals (Piscataqua River, Kittery); 2) Casco Bay (Portland); 3) Popham Beach and Sheepscot Bay (Androscoggin, Kennebec, and Sheepscot rivers, Phippsburg); 4) Penobscot Bay (Penobscot River, Islesboro); and 5) Cobscook Bay (St. Croix River, Eastport) (Figure 1, Table 2).

We evaluated geographic variation in freshwater ecosystems with common loon and bald eagle eggs (Figure 1). The species we selected have a broad range of foraging strategies and represent most of Maine's primary ecosystems.

The second component evaluated exposure in major habitat types through analyzing eggs from multiple species in the same area. In the Portland, Maine area we collected eggs from marine, estuarine, riverine, lake, and terrestrial habitats, focusing on high trophic[2] level predators (Figure 2). Species include insectivores, piscivores, and bird and mammal predators. Additionally, to ensure direct comparison among habitats, we collected eggs from tree swallows—a low trophic level insectivore. Collectively, this sampling effort provided a baseline and initial screening of contaminant levels, and helped determine if contaminants are concentrating in certain areas.

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[1] Increase in an organism over time because they take in more than they can expel.

[2] How high in the food web a bird eats (i.e. eagles are high trophic level and eiders are low).

2.2 Chemical Interaction

Researchers have studied the effects of many of the contaminants analyzed in this study on behavior, reproductive success, organ function, and acute toxicity. However, a number of studies have also attempted to determine if multiple compounds interact to create physiological effects greater than their sum. Researchers found that organochlorine pesticides can interact with each other to create either an additive or synergistic effect (Blus 2003). Epidemiological studies on human children (Grandjean et al. 2001, Stewart et al. 2003, Roegge et al. 2004), and laboratory studies on animals (Bemis and Seegal 1999, Costa et al. 2007) indicate that PCBs and methylmercury may act synergistically or additively. Additionally, researchers have found that PCB 52 can interact with PBDE 99 to enhance neurobehavioral defects in mice (Eriksson et al. 2006) . These studies suggests that many of the compounds analyzed in this study can interact to create an effect greater than one contaminant alone.

2.3 Review of compounds measured

2.3.1 Hg

Mercury is a naturally occurring heavy metal that has been mobilized into the environment by anthropogenic activities. Due to its unique properties, mercury is used in many products such as thermostats and dental fillings. It is also used in mining, and is released to the environment through the combustion of fossil fuels.

Generally attributed to anthropogenic input (Lockhart et al. 1998), mercury (Hg) levels in the North Atlantic have doubled over the last 100 years (Asmund and Nielsen 2000) and are increasing by nearly 1.5% a year (Slemr and Langer 1992) with peak levels in Maine recorded after 1970 (Perry et al. 2005). This historical increase has been documented in North Atlantic seabirds (Thompson and Furness 1992, Monteiro and Furness 1997), Canadian Arctic seabirds (Braune 2007) with local Hg deposition causing high rates of increase in biota (Frederick et al. 2004, Evers et al. 2007). This increase of global Hg levels since the 1900s is of concern because mercury is a persistent toxic heavy metal that both bioaccumulates and biomagnifies[3] in wildlife, and has neurological and reproductive impacts (Wolfe et al. 2007).

Researchers have documented Hg in the Maine sediment (Perry et al. 2005), water (Dennis et al. 2005), crayfish (Pennuto et al. 2005), fish (Kamman et al. 2005), salamanders (Bank et al. 2005), birds (Evers et al. 2005), and mammals (Yates et al. 2005). In addition Hg hot spots have been documented in Maine (Evers et al. 2007).

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[3] Builds up exponentially when one organism eats another.

2.3.2 PCBs

Polychlorinated biphenyls (PCBs) are synthetic chlorinated aromatic hydrocarbons that were first created in 1881; between 1930 and 1975 680 million kilograms were manufactured in the United States (Hoffman et al. 1996). Because of PCBs unique chemical properties they were used in a many industrial processes such as heat transfer agents, lubricants, dielectric agents, flame retardants, plasticizers, water proofing material, and most notably for cooling in electrical transformers (Hoffman et al. 1996). They are resistant to chemical breakdown, and have high thermal stability, low vapor pressure, flammability, and solubility (Niimi 1996). PCBs consist of two benzene (phenyl) rings connected by a carbon bond to which chlorine atoms are connected. The number of chlorine atoms provide the base for the 209 PCB congeners (Rice et al. 2003).

Originating from industrial leaks, sewage runoff, landfills, and incinerators, researchers have detected PCBs worldwide in the atmosphere, water, fish, birds, mammals, and humans (Hoffman et al 1996). Because of PCBs chemical structure, they are extremely persistent in the environment and resist being broken down by bacteria or chemicals. However, PCBs are easily absorbed into the fat of plankton and enter the wood web (Hoffman et al 1996) and are eventually consumed by wildlife and humans.

In wildlife, PCBs both bioaccumulate and biomagnify. Piscivorous (fish eating) birds are most exposed to PCBs, and eagles and other top trophic level predators are particularly vulnerable to accumulating elevated levels. PCBs are extremely toxic to biota, causing wasting, immune effects, reduced reproduction, and liver damage (Hoffman et al 1996). In birds PCBs reduce egg hatchability, increase liver size, and affect thyroid and spleen function (Hoffman et al 1996). Researchers have observed similar effects in mammals with PCBs reducing reproductive success, and at high levels can lead to death (Kamrin and Ringer 1996). Because of these known effects, PCBs were banned in the United States in 1979 (Rice et al. 2003). Today in Maine PCBs are still widely detected in wildlife. They have been detected in mussels (Chase et al. 2001), seabirds, shorebirds (Mierzykowski and Carr 2004), eagle (Matz 1998), porpoise (Westgate et al. 1997), dolphin, and pilot whale (Weisbrod et al. 2001).

2.3.3 PBDEs

Polybrominated diphenyl ethers (PBDEs) are brominated flame retardants that are used in both commercial and residential textiles and electronics. They work by slowing combustion by releasing hydrogen bromide gas, which interferes with the chemical reaction that spreads fire (Janssen 2005). PBDEs consist of two benzene rings linked by an oxygen atom and can have up to ten attached bromine atoms (Hellstrom 2000). This stable structure causes the molecules to be lipophilic (fat loving) and consequently subject to bioaccumulation (Karlsson et al. 2006). The three primary types of PBDEs are penta-BDE, octa-BDE, and deca-BDE. Penta has been primarily used in polyurethane foam (up to 30% in weight) that is used in couches, carpets, and mattresses; octa is used in computer monitor plastics; and deca, which makes up 83% of global PBDE production, is used in electronic equipment (Johnson-Restrepo et al. 2005). Deca-BDE is an off-white crystalline powder that is usually 10-15% of the weight of the host material and is an additive flame retardant that does not chemically bond to its host material. Consequently, deca-BDE migrates into the environment (DEP 2007). PBDEs enter the environment through atmosphere deposition, wastewater treatment facilities, and runoff (Anderson and MacRae 2006).

PBDEs are found globally in humans, wildlife, and the environment. They have been found in whales, Tasmanian devils, fish, and falcons in Australia (Symons et al. 2004); terns in San Francisco Bay (She et al. 2004); guillemots in the Baltic Sea (Sellstrom et al. 2003); peregrine falcons in Sweden (Sellstrom et al. 2001); marine fish in Florida (Johnson-Restrepo et al. 2005); seabirds in Norway (Murvoll 2006); birds of prey in Belgium (Voorspoels et al. 2004); birds of prey in China (Chen et al. 2007); fish in Maine's Penobscot River (Anderson and MacRae 2006); and Arctic fox in Greenland and Russia (Lifgren 2005).

Laboratory studies have documented health effects of PBDEs, generally at levels higher than currently observed in the environment. Rats fed penta-BDE had reduced growth, diarrhea, reduced activity, tremors, red stained eye edges, and chewed continuously. Those animals that received repeated doses had changes in hepatic and thyroid size and histology as well as immunological effects. Rats fed octa-BDE had enlarged livers, and fetuses with bent ribs, limp bones, and rear limb malformations. Although health effects were observed at higher doses, animals dosed with deca-BDE had enlarged livers, and hyaline degeneration in kidneys. Those fed deca-BDE for 103 weeks at high doses developed tumors as well as an increase in thyroid, hepatic and pancreatic adenomas (Darnerud 2003). A dosing study on kestrels found changes in thyroid levels and concludes: "Concentrations of PBDE congeners in wild birds may alter thyroid hormone and vitamin A concentrations, glutathione metabolism and oxidative stress (Fernie et al. 2005)." Because of these effects, penta and octa were voluntarily phased out in 2004 (EPA website), and deca was partially banned in Maine and Washington State in 2007.

2.3.4 PFCs

Perflorinated chemicals (PFCs) have been produced for over 50 years for their repellant properties and are used as stain repelents, cleaning agents, floor polish, fire-fighting foam, and in photography (Tao et al. 2006). Most commonly used PFCs are derived from perfluorooctanesulfonyl fluoride (POSF), which have extremely strong carbon-fluorine bonds. These strong bonds make the PFCs highly resistant to environmental and metabolic degradation (Butenhoff et al. 2006) and are consequently environmentally persistent (Kannan et al. 2002). Of the PFC congeners, perfluorooctanesulfonate (PFOS) and perfluorooctanoate (PFOA) are of greatest concern because of their global abundance and bioaccumulation (Giesy and Kannan 2001, Kannan et al. 2002, and Tao et al. 2006).

Annual estimated production of POSF in 2000 was greater than 5000 tons (Tao et al. 2006), but by 2002 the 3M Company—the primary manufacture of POSF—discontinued production (Butenhoff et al. 2006). However, some PFOS is still produced outside of the United States for applications where there are no alternatives (Butenhoff et al. 2006) and other PFC are still produced and used in the United States (Kannan pers. com.). PFCs are transported in the environment through ocean currents and the atmospheric circulation (Toa et al. 2006) and may enter the environment through similar pathways as PBDEs.

Although there has been no analysis of PFCs in Maine, they have been documented in wildlife in the Southern Ocean and Antarctica (Toa et, 2006), Artic, North America, Pacific Ocean, Japan, Europe (Giesy and Kannan 2002), seaotters in California (Kannan et al. 2006), birds in Japan and Korea (Kannan et al. 2002), and in fish and pelicans in Columbia (Olivero-Verbel et al. 2006).

PFOS are documented to have health effects in wildlife. Hen eggs injected with PFOS had significantly lower hatching success (Molina et al. 2006). Quail exposed to PFOS through diet had increased liver weight and, at high levels, died (Newsted et al. 2007). In California, diseased sea otters were positively associated with elevated PFOS levels (Kannan et al. 2006).

2.3.5 OCs

Organochlorine pesticides (OCs) are used primarily for insect control, are extremely persistent in the environment, and bioaccumulate in wildlife (Blus 2003). The five major groups are dichlorodiphenyltrichloroethane (DDT), hexachlorocyclohexane (HCH), cyclodienes, toxephene, and chlordecone.

2.3.6 HCH

Hexachlorocyclohexane (HCH) is an insecticide that is currently used in agriculture—the most wildly used form is lindane. Unlike other OCs pesticides, lindane has a short half-life and rapidly degrades after use. Consequently, lindane is rarely found in wildlife. However, in some laboratory studies lindane has reduced hatching success, increased embryo mortality, and caused egg shell thinning in chickens. In other studies researchers documented little effects (Blus 2003).

2.3.7 HCB

Hexachlorobenzene (HCB) is a fungicide used most commonly on seed grains, is an industrial waste product, and is used in the manufacture of tire rubber (Wiemeyer 1996). HCB is persistent in the environment and experimental studies have documented death and significant effects in birds. Quail fed high doses of HCB had weight loss, ruffling of feathers, and tremors. Birds fed a lower does had reduced hatchability of eggs and sterile eggs (Wiemeyer 1996).

2.3.8 Chlordane

Chlordane is composed of number of OCs and has been used since the 1940s (Blus 2003). In 1978 most chlordane was restricted in the United States; all chlordanes are now banned (Wiemeyer 1996). The most toxic metabolite is oxychlordane (Wiemeyer 1996). In the past chlordane was used extensively on lawns, golf courses, and crops, and is persistent in the environment. The most measured effect in experimental settings is death. As recently as 1997 over 400 birds died from eating beetles with high chlordane residues in an area that had been treated in the past (Blus 2003).

2.3.9 DDT

Dichlorodiphenyltrichloroethane (DDT) was first synthesized in 1874, used as an insecticide in 1939, used extensively in agriculture after World War II (Blus 1996), and banned in the United States in 1972 (Blus 2003). Despite the well-documented effects on wildlife, DDT is still used in a number of countries. After application DDT breaks down to DDE. DDE has been well documented to cause egg shell thinning, which causes eggs to break during incubation. Because of the persistent nature of DDE, it is still widely detected in birds although at levels generally below effects thresholds (Blus 2003).

2.4 Birds as bioindicators of the environmental contaminants

Birds are commonly used as indicators of Hg and other contaminants in the environment (Scheuhammer 1987, Furness and Camphuysen 1997, Wolfe et al. 1998, Cifuentes et al. 2003, Braune 2007, Evers et al. 2005, and Sheuhammer et al. 2007, and Wolfe et al. 2007). The species we selected for this contaminant screening represent distinct foraging guilds and ecosystems across Maine. Additionally, some of the species we selected are high trophic level predators that may accumulate contaminants at higher levels. In total the 23 species of birds in our study indicate the contaminants other biota, and people—through consuming fish and game—may be exposed to.

2.5 Eggs as indicators of local contaminants

Eggs are used extensively for contaminant studies (Wiemeyer 1996, Kannan et al. 2001, Braune et al. 2002, Evers et al. 2003, and Braune 2007) because female birds depurate lipophilic contaminants into their eggs. For most species, all of the egg nutrients are allocated from exogenous (i.e. recent dietary uptake) rather than endogenous (reserves acquired during migration and on winter grounds) sources (Bond et al. 2007, Hobson 2006, Hobson et al. 2000, and Hobson et al. 1997). Consequently, egg contaminant residues represent the contaminants present in the bird's breeding territory diet (Hobson et. al 1997). These findings are supported by Evers et al. (2003), which found a strong relationship between common loon egg Hg levels and female Hg blood levels (blood represents recent dietary uptake). The exception is species that arrive on the breeding ground and immediately lay eggs (Hobson 2006). The species in our study are all present at their breeding site for at least two weeks prior to laying eggs (Table 1). Therefore, the results presented in this report represent contaminant levels of the birds within their foraging range during the breeding season in Maine.

2.6 Species selected for this study

2.6.1 Tree swallows (multiple habitats)

In order to be able to directly compare contaminant levels between habitats, we collected swallow eggs from birds nesting in the marine, estuarine, lacustrine (lake), and terrestrial environments. We were not able to collect samples from a river. Although we only analyzed one composite per habitat, each composite represented multiple clutches. Tree swallows feed on flying insects (Robertson et al. 1992) close to their nesting boxes and feed at a low trophic level; therefore, they act as a bioindicators of their nesting habitat.

2.6.2 Marine

In order to seek geographic variation along the coast, we analyzed eggs of eight species of seabirds at six locations at multiple trophic levels. Below is a description of the habitat each species represents.

- Herring gulls are scavengers (Perotti and Good 1994) and will be exposed to contaminants through multiple sources such as invertebrates, fish, birds, small mammals, and garbage. Therefore, they act as bioindicators of multiple coastal habitats.

- Double-crested cormorants are piscivores and forage on mid-water and benthic fish 3-40cm long (Hatch and Weseloh 1999). These fish tend to be highly mobile, consequently cormorants act as bioindicators of a broad coastal region (Goodale et al. 2007).

- Common eiders feed primarily on mollusks (Goudie et al. 2002) and in Maine feed extensively on blue mussels. Therefore, eiders act as bioindicators of the contaminants that mussels are accumulating through filter feeding.

- Leach's storm-petrel feed 100-200km offshore (Huntington et al. 1996) and feed primarily on mesopelagic fish and crustaceans (Watanuki 1985, Hedd and Montevecchi 2006). Since storm-petrels feed in the offshore food web, their contaminant levels reflect a global signal rather than one influenced by point sources (Goodale et al. 2007). Therefore, storm-petrels act as bioindicators of global contaminant levels.

- Black guillemot provide a contaminant signal of benthic dwelling biota because they feed primarily on rock gunnels (a small fish) in the Gulf of Maine (Preston 1968, Hayes 1993, and Bulter and Buckley 2002). Since guillemots feed close to their breeding colony and rock gunnels have low mobility during the summer (Vallis et al. 2007), they indicate contaminants close to their breeding colony. Therefore, guillemots act as bioindicators of the coastal benthic system.

- Terns feeding on small fish and invertebrates close to the ocean surface (Hatch 2002, Nisbet 2002, Thompson et al. 1997) which tend to be mobile. Therefore, they act as bioindicators of the coast as a whole, but at a lower trophic level than cormorants.

- Atlantic puffin provide a mid-trophic level signal as they feed on larger fish than the terns but smaller than cormorants (Lowther et al. 2002). Therefore, they act as bioindicators at a mid-trophic level.

- Piping plovers are not a seabird, but they feed on invertebrates in the intertidal zone (Haig 2004) along Maine beaches. Therefore, they act as bioindicators of Maine's beaches.

- Osprey provide a similar signal to many of the seabirds along the coast. They feed on live fish (Poole et al. 2002) on the surface by plunging into the water. Therefore, they act as a similar bioindicator as cormorants, but for raptors.

2.6.3 Estuarine

We focused on one estuary, Scarborough Marsh. Within this estuary, in addition to tree swallows, we collected samples from four species: Virginia rail, willet, glossy ibis, and snowy egret. Collectively these species provide an indication of bioaccumulation in estuarine invertebrates.

- Virginia rail feed on aquatic invertebrates (Conway 1995).

- Willets on insects, invertebrates, and fish (Lowther et al. 2001).

- Glossy ibis also feed on invertebrates (Davis and Krichner 2002), but likely at a higher trophic level than the rail and willet.

- Snowy egret have greater variety in their diet and, in addition to invertebrates, feed on fish, amphibians, and reptiles (Parsons and Master 2000).

2.6.4 Riverine

From rivers we collected eggs from kingfishers and eagles.

- Kingfisher feed on small fish (Hamas 1994) by plunging into the water. They build their nests by burrowing into river banks (Albano 2002) and feed in the adjacent river. Consequently, they provide a direct contaminant signal of the river where they are nesting.

- Bald eagles are scavengers and have a varied diet of fish, birds, and mammals, but when nesting close to rivers will feed primarily on fish. They feed on larger fish than kingfishers and consequently are indicators of a higher trophic level.

2.6.5 Lacustrine (lake)

From lakes, in addition to tree swallows, we collected samples from red-winged blackbirds, loons, and eagles.

- Blackbirds provide a contaminant signal of insects (Yasukawa and Searcy 1995) along the lake edges.

- Loons provide a signal of mid-sized fish, and eagles mid-sized fish, birds, and mammals that are associated with lakes.

2.6.6 Terrestrial

In addition to tree swallows, we collected samples from peregrine falcons and American kestrels for the terrestrial environment. Together these species provide a terrestrial contaminant signal at two trophic levels.

- Kestrels feed at a lower trophic level than peregrines, feeding primarily on terrestrial arthropods and small vertebrates (Smallwood and Bird 2002).

- Peregrines feed on birds (White et al. 2002) and in the area that we collected samples, many of these prey birds feed in the terrestrial environment.

3. METHODS

3.1 Field

We collected viable and nonviable three-egg composites and single eggs from each species (Table 2). Species scientific names are displayed in Table 2. Members of Gulf of Maine Seabird Contaminant Network (GOMSCAN)[4] collected seabird eggs from remote seabird islands, and BRI staff collected the additional samples. In the field eggs were handled with polyethylene gloves, placed whole in polyethylene bags, and placed in chemically clean jars, and frozen. The eggs were sent with dry ice to the Wadsworth Center (New York State Department of Health) for analysis (see below for methods). Whole egg was analyzed. BRI currently has state and federal collection permits.

3.2 Statistics

We performed statistics with JMP (SAS Institute Inc., 2001). Each egg composite was treated as a sample size of one. We log10 transformed the data to increase normality and homoscedasticity. We tested for interaction between habitat and species: we found no interaction for all compounds, indicating that we could independently test for habitat differences.

We sought spatial trends by mapping contaminant levels in eight species where we had three samples or more. The species we mapped were: herring gull, double-crested cormorant, common eider, black guillemot, piping plover, common loon, osprey, and bald eagle. The range of each contaminant was displayed in three categories determined by natural breaks within the contaminant range for each species. Trends were evaluated qualitatively.

3.3 Egg morphometric measurements

An hand-held caliper, capable of recording the 0.1 of a mm was used to determine the length and width. The egg length was measured from tip to tip of the egg. The width was measured from the widest point of the egg. A digital balance capable of weighing to the 0.1 of a gram was used to measure weight of the eggs with shell (whole egg) and without shell (content weight). Graduated measuring cylinders with Milli-Q water was used to determine the volume of eggs, determined as the volume of water displaced (recorded in ml). Developmental stage of the eggs were recorded as a ranking of the developmental stage of the embryo. An embryological development scale used for common loon and

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[4] GOMSCAN is comprised of researchers from U.S. Fish and Wildlife Service, Maine Department of Inland Fisheries and Wildlife, National Audubon, Maine Coast Island National Wildlife Refuge, Shoals Marine Lab, and Canadian Wildlife Service. This group is currently preparing a paper for publication on seabird mercury levels in the Gulf of Maine.

waterfowl eggs was used to assess the developmental stage and ranked as NA, 0,1,2,3,4,and 5 as below:

NA (not assessable): Developmental stage could not be determined. Contents were gray or yellowish-tan in color and typically had a foul smell. A darker color suggested some degree of development had occurred, whereas a yellow homogeneous liquid may be sifted through and if no dark spots or hardened areas were found we classified the egg as infertile (0).

0: No development was evident. Egg had a yellow/orange or yellow/tan yolk (intact or broken down into a liquid). A translucent jelly-like mass surrounded the yolk sac and showed no sign of embryonic development (e.g. mass not dark or hardened).

1: Embryo was viable (length was up to 1.5 cm). The jelly like mass (embryo) was dense and hardened. Small dark (red) eyespots may be visible at this stage.

2: Developing embryo (length was 1.5 – 2.0) has an apparent central nervous system. Cranial development and visible eyes are apparent. Feathers are absent.

3: The embryo shows advanced development (length was 2-3 cm). Bill was developed (e.g. egg tooth present but soft). Legs and wings were visible but not fully developed. Some feathers were present (first seen in tail).

4: The fully developed embryo was completely covered by feathers. Appendages were completely developed. Vent, preen gland was visible. A small portion of yolk sac remained attached to belly.

3.4 Analysis of egg moisture and lipid contents

After the determination of morphometric parameters on each of the eggs, some samples collected from the same location and same species were pooled and homogenized using a homogenizer and composites were prepared. The composites were used the analysis of trace organic contaminants and mercury. Homogenized egg samples (in most cases 10-11 g; for some samples only 5 g was used due to the availability) were extracted with dichloromethane and hexane (1:3; 400 mL) in a Soxhlet apparatus for 16 h after spiking the samples with surrogate standards (PCB-30 and PCB204). The extracts were concentrated to 10 mL and 1 ml of the aliquot was taken for the analysis of lipid content by gravimetry. An aliquot of the egg homogenate (approximately 2 g) was also taken and freeze-dried to measure the moisture content.

3.5 Analysis of PCBs, PBDEs and organochlorine pesticides

Details of the analytical methods have been described elsewhere (Kannan et al., 2005; 2007). An aliquot of the sample extract was spiked with ^{13}C-labeled PCB congeners 3, 15, 31, 52, 118, 153, 180, 194, 206, 209, and ^{13}C-labeled PBDE congeners 3, 15, 28, 47, 99, 100, 118, and 153 as internal standards. PCB congeners 30 (2,4,6-triCB) and 204 (2,2',3,4,4',5,6,6'-octaCB) were spiked as surrogate standards. The sample extracts was then purified by passage through a series of layers of silica gel (Davisil, 100-200 mesh, Aldrich, WI; 1 g of silica gel, 2 g of 40% acidic-silica gel, 2 g of 20% acidic-silica gel, and 1 g of silica gel at the top). The analytes were then eluted using 150 mL of 20% dichloromethane in hexane. The extracts were then concentrated using a rotary evaporator and treated with sulfuric acid (5 mL) and further concentrated to 1 mL for the analysis of PCBs and PBDEs. Another portion of the extract was passed through silica gel (2 g) by elution with 20% dichloromethane in hexane; it was then treated with sulfuric acid, for the analysis of organochlorine pesticides.

Extracts were injected into a gas chromatograph (Hewlett-Packed 6890) coupled with a mass-selective detector (Hewlett-Packed, series 5973) for the determination of PCBs and PBDEs. A capillary column coated with RTX-5MS (30 m x 0.25 mm i.d. x 0.25 µm film thickness; Restek Corp, Bellefonte, PA) was used for the separation of individual isomers. The column oven temperature was programmed from 100°C (1 min) to 160°C (3 min) at a rate of 4°C/min, and then to 250°C at 3°C/min, with a final hold time of 5 min for PCBs. For PBDEs, the column temperature was programmed from 100 °C (1 min) to 160°C (3 min) at a rate of 10°C/min, and then to 260°C at 2°C/min, with a final hold time of 5 min. The MS was operated in an electron impact (70 eV), selected ion monitoring mode. An equivalent mixture of Kanechlor (KC300, 400, 500, and 600) with known PCB composition was used in the identification of PCB congeners. One hundred and fifty four isomers of PCBs with 35 coleuting pairs (IUPAC number in the order of GC-MS elution: 4+10, 9+7, 6, 5+8, 19, 18, 17, 15, 24+27,16+32 ,26,25,28+31, 20+33+53, 22, 36, 37, 54, 50,53,51,45, 52+73, 46+69, 49+43, 47+48+75, 44, 59+42, 41+64, 40+57, 67, 63, 74+61, 70+76, 66+80, 60+56, 77, 104, 98+102, 93+95, 91, 92, 84, 90+101+89, 99, 86+97, 97+113, 87+117+125+116+111+115, 85+120, 110, 82, 124, 107, 118+106, 114+122, 105+127, 126, 155, 136, 151, 135+144, 149+139, 134, 133, 146+161, 153, 132+168, 141, 137, 130, 138+164+163, 158, 129, 128, 167, 156, 157, 169, 188, 179, 176, 178, 187+182, 183, 185, 174, 177, 171, 173, 172+192, 180, 193, 191, 170, 190, 189, 202, 201, 197, 200, 198, 199, 196+203, 195, 194, 205, 208, 207, 206, and 209), including mono-*ortho* PCB congeners (105, 118, 189) were analyzed. Quantification of PCB congeners was based on external calibration standards containing known concentrations of di- through deca-CB congeners. Concentrations of individually resolved peaks of PCB isomers were summed to obtain total PCB concentrations. PBDE congeners were monitored at molecular ion clusters, $[M]^+$ and $[M+2]^+$ or $[M+4]^+$. Tri- through hexa-PBDE congeners analyzed in this study were 28, 30, 47, 66, 85, 99, 100, 138, 153, and 154 were targeted for analysis. Hepta- through deca-BDE congeners (183, 203, and 209) were analyzed using a Agilent Technologies 6890N gas chromatograph-electron capture detector (GC-ECD). PBDE congeners were quantified using an external calibration standard. Organochlorine pesticides were analyzed using a Agilent Technologies 6890N gas chromatograph-electron capture detector (GC-ECD; for HCH isomers) and a gas chromatograph (Hewlett-Packed 6890) coupled with a mass-selective detector (Hewlett-Packed, series 5973) for DDTs, chlordanes and HCB. A capillary column coated with DB-5 (30 m x 0.25 mm i.d. x 0.25 µm film thickness) was used for the separation of pesticides. Concentrations were calculated from the peak area of the sample to that of the corresponding external standard. DDTs refers to the sum of *p,p'*-DDE, *p,p'*-DDT and *p,p'*-DDD; chlordanes to the sum of *cis*-chlordane, *cis*-nonachlor, *trans*-nonachlor, and oxychlordane; HCHs to the sum of α-,β-, and γ-isomers. PCB and PBDE congeners are represented by their IUPAC numbers.

3.6 PCB and PBDE quality assurance and quality control

The extraction, clean-up, and fractionation steps were evaluated by measurement of the absolute recoveries of the compounds spiked and passed through the entire analytical procedure. Mean (± standard deviation) recoveries of ^{13}C-labeled PCB congeners #30, 118, 153, and 194 spiked into the samples were 80 ± 14%, 82 ± 17%, 89 ± 12%, and 91 ± 14%, respectively. Recoveries of surrogate PCB congeners CB-30 and CB-204 spiked into the egg samples prior to extraction were 72±10%. Mean (± standard deviation) recoveries of ^{13}C-labeled PBDE congeners 28 and 47 were 92 ± 14% and 91 ± 14%, respectively. Overall recoveries of PBDEs ranged from 82 to 103%. The reported concentrations of PCBs, PBDEs and pesticides were corrected for the recoveries of surrogate standards (CB-30 and CB-204). Recoveries of organochlorine pesticides through the analytical procedure ranged from 85 to 110%. Procedural blanks were analyzed for every set of 10 samples, as a check for interferences. Calculated concentrations were reported as below the limit of detection, if either the observed isotope ratio was not within ±20% of the theoretical-ratio, or the peak area was not greater than the specified threshold (3 times the noise). Known concentrations of PCBs, PBDEs, and organochlorine pesticides were spiked into selected samples (matrix spikes) and passed through the entire analytical procedures to calculate the recoveries. Recoveries of all of the target compounds spiked into egg matrixes were between 84 and 106% with a standard deviation of <15%. The quantitation limits of individual PBDE congeners varied from 10 to 500 pg/g, wet wt. The quantitation limit for organochlorine pesticides varied from 50 to 1000 pg/g, wet wt.

3.7 Analysis of perfluorinated compounds:

Potassium salts of PFOS (86.4%), PFOA (98%), PFOSA (95%), PFHxS (99.9%), and PFBS (99%) were provided by the 3M Company (St. Paul, MN). PFHpA, PFNA, PFDA, and PFUnDA were from Fluorochem Ltd (≥95% purity, Derbyshire, UK). $^{13}C_4$-PFOS, $^{13}C_4$-PFOA (99% purity, Wellington Laboratories, Guelph, ON, Canada), $^{13}C_4$-PFNA and $^{13}C_4$-PFDA were used as internal standards and were spiked into egg samples prior to the addition of reagents for extraction.

PFCs in eggs were analyzed following the method described elsewhere (Tao et al., 2007). Egg homogenates (0.3-0.5 g) were taken in 15-mL polypropylene (PP) tubes and 5 ng of internal standards ($^{13}C_4$-PFOS, $^{13}C_4$-PFOA, $^{13}C_2$-PFDA, and $^{13}C_2$-PFNA), 2 mL of 0.25 M sodium carbonate buffer, and 1 mL of 0.5 M tetrabutylammonium hydrogensulfate solution (adjusted to pH 10) were mixed. Sample was then extracted with 5 mL of methyl-tert-butyl ether (MTBE) by shaking vigorously for 45 min. The MTBE layer was separated by centrifugation at 3500 rpm for 5 min and then transferred into another PP tube. The extraction was repeated twice with another 3 mL of MTBE. The MTBE extract was combined and evaporated to near-dryness under a gentle stream of nitrogen and then reconstituted with 1 mL of methanol. The sample was vortexed for 30 sec and filtered through a 0.2-µm nylon filter into an autosampler vial. Matrix-matched calibration standards (seven points ranging from 0.5 ng/mL to 75 ng/mL) were prepared by spiking different amounts of calibration standards into a sample that contained no quantifiable amount of the target analytes; these standards were passed through the entire analytical procedure along with the samples.

Analytes were detected and quantified using an Agilent 1100 series high-performance liquid chromatography (HPLC) coupled with an Applied Biosystems API 2000 electrospray triple-quadrupole mass spectrometer (ESI-MS/MS). Ten microliters of the extract were injected onto a 50 x 2 mm (5 µm) Keystone Betasil® C18 column. The mobile phase was 2 mM ammonium acetate/methanol starting at 10% methanol, at a flow rate of 300 µL/min. The gradient increased to 100% methanol at 10 min and was held for 2 min, and then reversed back to 10% methanol. The MS/MS was operated in electrospray negative ion mode. Target compounds were determined by multiple reaction monitoring (MRM). The MRM transitions were 299>80 for PFBS, 399>80 for PFHS, 499>99 for PFOS, 503>99 for $^{13}C_4$-PFOS, 599>99 for PFDS, 498>78 for PFOSA, 363>169 for PFHpA, 369>169 for PFOA, 372>172 for $^{13}C_4$-PFOA, 463>219 for PFNA, 513>219 for PFDA, 563>169 for PFUnDA, and 613>169 for PFDoDA. Samples were injected twice, to monitor sulfonates and carboxylates separately, and PFBS was monitored in both of the injections. A mid-point calibration standard was injected after every 10 samples to check for the instrumental response and drift. Calibration standards were injected daily before and after the analysis.

The egg samples were quantified with the quadratic regression fit analysis weighted by 1/x of a matrix-extracted calibration curve. The limit of quantitation (LOQ) was determined as the lowest acceptable standard in the calibration curve that is defined as a standard within ±30% of the theoretical value, and that has a peak area twice as great as the analyte peak area in blanks. LOQs for PFCs were 0.28 to 0.6 ng/g, wet wt, except for PFDS and PFBS, for which the LOQs were 0.94 and 1.12 ng/g, wet wt, respectively.

3.8 PFC quality assurance and quality control

Matrix spikes (6 egg composites) were performed for egg samples. Known amounts of mixed PFC standards (20 ng each) were spiked into sample matrices before extraction and were passed through the entire analytical procedure. Recoveries of PFCs spiked into egg homogenates and passed through the entire analytical procedure are shown in Table 3. The recoveries of all the PFCs were acceptable except for PFBS, which had a low recovery; however, PFBS does not bioaccumulate in tissues and also had not been detected in biological samples. Four 13C-labeled internal standards were spiked into all samples before the extraction, and the recoveries of internal standards are also shown in Table 3. Reported concentrations of PFCs in egg samples were not corrected for the recoveries of internal standards. Blanks were analyzed by passing Milli-Q water and reagents through the whole analytical procedure. Blanks contained trace levels of PFOA (<100 pg). Reported concentrations for PFOA in egg samples were subtracted from the mean blank values. A midpoint calibration standard was injected after every 10 samples to check for instrumental stability, response and drift. Calibration standards were injected daily before and after the analysis.

3.9 Mercury analysis

Egg composites were freeze-dried and homogenized; an aliquot (~0.1 g) of the sample was weighed in a vial lined with Teflon®. Samples were digested overnight in concentrated nitric acid (2 mL). Samples were then further digested in a microwave oven for 7 min at 200 W; this step was repeated three times. Concentrations of Hg were determined by a cold vapor atomic absorption spectrometer (Model HG-3000; Sanso, Tsukuba, Japan). The limit of quantification was 50 ng/g, dry wt. Accuracy of the analysis was examined by analyzing Certified Reference Materials: dogfish muscle (DORM2; National Research Council, Ottawa, ON, Canada) and bovine liver (SRM1577b; National Institute of Standards and Technology, Gaithersburg, MD, USA) along with the samples. The overall analytical scheme used for the analysis of egg samples is shown in Figure 3.

4. RESULTS AND DISCUSSION

4.1 Relationship between compounds (Figure 4, Figure 5)

We found that PCBs, PBDEs, chlordane, and DDE all significantly increase simultaneously ($p < 0.0001$). This finding indicates that birds with high PCB levels also tend to have high PBDE, chlordane, and DDE levels. This is consistent in studies conducted with OCs, which show that the pesticides are positively correlated in animal tissue (Blus 2003). This is of particular interest because in mice PCBs and PBDEs are demonstrated to interact, and together, at low doses can enhance developmental neurobehavioral defects (Eriksson et al. 2006). Additionally, researchers have also found that organochlorine pesticides (both DDE and chlordane are OCs) interact (Blus 2003).

The simultaneous increase in these compounds may be caused by a number of factors, including the similar chemical structure of PCBs and PBDEs, and their similar pattern of bioaccumulation. PCBs, PBDEs, and DDE are all composed of two benzene rings, but in PCBs the benzene rings are connected with a carbon bond, while in PBDEs there is an oxygen atom. PBDEs have attached bromine atoms, while PCBs have attached chlorines. This similar structure may mean that they move through the environment in a similar pattern.

PCBs and OCs have been extensively studied (Hoffman et al. 1996, Wiemeyer 1996, Blus 2003), but only recently have PBDEs been studied in wildlife. The positive relationship between these compounds suggests that species and geographic areas that have been documented to have high PCB levels may also have elevated PBDEs.

Although not as strongly correlated, we also found a significant relationship between PFOS and PBDE and PCB and PFOS. This indicates that as PCB and PBDE levels increase, that PFOS levels also tend to increase.

4.2 Total Contaminants (Figure 6)

By combining the ranks (e.g. the species with the lowest PCB levels receives a rank of "1" while the highest receives a rank of "23") of the major contaminant groups (Hg, total PCB, total PBDE, PFOS, total chlordane, HCB, and DDE, PFOS) for all species we are able to determine which species have the highest contaminant load overall. Bald eagle have the highest or second highest level in almost all contaminants. Note: this method of ranking does not account for the relative level of the contaminants (e.g. eagles in many cases had contaminant levels multiple times higher than other species).

As expected, high trophic level predators have the highest levels (bald eagle, peregrine falcon, great black-backed gull), while species specializing on invertebrates (Virginia rail, willet) and mollusks (common eider) have the lowest levels. However, two species do not fall into expected levels, specifically: belted kingfisher and piping plover.

Belted kingfisher may have higher than expected levels because their eggs (from to two nests) were collected from an industrialized Presumscot River (Portland). This river potentially has point source pollution as well as higher atmospheric deposition rates than other sites because of its proximity to Portland and a municipal incinerator.

The reason for the elevated contaminants in piping plover is not clear. Like willets and Virginia rails, plovers feed on small invertebrates in the intertidal zone. However, in contrast to the plovers, these two species consistently have some of the lowest contaminant levels. The plovers may be feeding upon large or higher trophic level invertebrates than the other invertivores, and from mud flats that may have higher pollution levels. Note: although plovers ranked fifth overall, in general their contaminant loads are four to six times lower than the species with the highest levels.

4.3 Hg (Figure 7)

4.3.1 Comparison to known effects thresholds

Four samples are close to or above the known effects threshold of 1.3 µg/g (ww, ppm) (Evers et al. 2003, Evers et al. 2007a). Two loon eggs from northwestern Maine are close to or significantly above this threshold—Aziscohos Reservoir, 1.26 µg/g, and Flagstaff, 3.32 µg/g—and one egg from Long Pond in Acadia National Park, 1.55 µg/g. The eggs from Aziscohos and Flagstaff were collected at sites known to have high Hg levels (BRI unpublished data) and in an area documented as a mercury hotspot (Evers et al. 2007b). Salamanders Hg levels are higher than other sites in Acadia National Park (Banks et al. 2005). One eagle sample from a fresh water lake is close to the effects threshold, 1.20 µg/g.

4.3.2 Comparison with other studies

Our results are consistent with other studies (levels from other studies are bold and in brackets). Common loon eggs consistently have the highest Hg levels in multi-species studies (Evers et al. 2005). However, our mean 1.4 µg/g, is higher than the regional mean **[0.78 µg/g (ww)(Evers et al. 2005)]**; this is the result of sampling at areas known to be high. We collected samples at these sites to determine if other contaminants would also be elevated. Our eagle mean, 0.67 µg/g, is also slightly above the regional mean **[0.45 µg/g (ww)(Evers et al. 2005)]**.

The seabird results are also consistent with those established for the Gulf of Maine. Black guillemot and Leach's storm petrel consistently have the highest level among seabirds in Maine (Goodale et al. 2007). The storm-petrel Hg level we recorded, 0.60 µg/g, is close to the Gulf of Maine mean **[0.62 µg/g (Goodale et al. 2007)]**. The mean guillemot Hg levels we recorded, 0.69 µg/g, are slightly higher than those of the region **[0.52 µg/g (Goodale et al. 2007)]**.

Other species also have Hg levels consistent with regional means. Our results compare to other studies as follow: common tern in our study, 0.02 µg/g **[0.13 µg/g (Goodale et al. 2007)]**; piping plover in our study, 0.23 µg/g **[0.17 µg/g (fww) (Mierzykowski et al. 2003)]**; least tern in our study, 0.16 µg/g **[0.12 µg/g (Evers et al. 2005)]**; osprey in our study 0.18 µg/g, **[0.19 µg/g (Evers et al. 2005)]**; and tree swallow in our study 0.11 µg/g **[0.19 µg/g (Evers et al. 2005)]**.

4.3.3 *Spatial Variation (Figure 19)*

Mercury accumulates in the environment in hot spots, influenced by deposition patterns, watershed chemistry, food web dynamics, reservoirs, and point sources (Evers et al. 2007). Our results are consistent with this pattern. As discussed above, the two elevated Hg levels in loons in northwestern Maine are in an established hot spot (Evers et al. 2007). The high levels in Isles of Shoals are consistent with other studies (Goodale et al. 2007) and are likely influenced by the Piscataqua River and proximity to deposition from southern New England. The elevated levels in mid-coast Maine—Penobscot Bay, Mount Desert Island—are also associated with an area of concern for high Hg availability (Evers et al. 2007). The elevated levels are due in part to the former HoltraChem plant, which dumped Hg into the Penobscot River. The moderate levels in Cobscook Bay may be influenced by being downstream from a Hg area of concern (Evers et al. 2007). Although not consistent across species, piping plover, herring gull, and common eider all have higher levels at the mouth of the Kennebec. However, cormorants and osprey levels are low at this site.

4.3.4 Habitat (Figure 26)

Our data demonstrated Hg levels are highest in lakes and lowest in estuaries. We observed this trend both in the over all data set (ANOVA Tukey HSD, F = 8.5, df = 4, 53, $p < 0.0001$) and in the tree swallow data. This trend is also observed with the eagle data: marine < riverine < lake.

Evers et al. (2005) found a similar trend, with eagle and belted kingfisher data demonstrating that lakes had the greatest Hg availability. They attribute the higher Hg levels in lakes to the presence of sulfur reducing bacteria, which methylates elemental Hg (convert inorganic Hg to an organic form that enters the food web), and the dilution in marine, estuarine, and riverine environments.

4.4 PCB (Figure 8)

4.4.1 Comparison to known effects thresholds

The effects of PCBs on wildlife have been well studied (Blus 2003). Studies on bird eggs have shown chickens are particularly sensitive to total PCB levels and can show effects at 1,000-5,000 ng/g (ww) (Hoffman et al. 1996). In the field, total PCB levels have shown effects ranging from 8,000 – 20,000 ng/g in terns and other species (Hoffman et al 1996). Our results indicate that the eagle egg collected on the coast may have PCB levels high enough to cause effects. Osprey, herring and great black-backed gulls, peregrine falcon, and common loon all have PCB levels between 1,000 and 5,000 ng/g (ww). Past studies indicate that these species may have a higher threshold for effects of PCBs (Welch, 1994, Matz 1998, Hoffman et al. 1996), and may not experience effects at the levels we detected. However, the PCB congeners that dominated the samples are not considered the most toxic (Figure 14 &15). Most species total PCBs are dominated by the following co-elutes: 153/132/168, 138/164/163/158, and 180. The exceptions were Virginia Rail and willet. We did not test for 81, 126, or 169. However, we did find 77 in 27 of the composites (ranging from 0.16 – 1.82 ng/g ww), with osprey from Bug Light in Portland (1.82), bald eagle (1.35) from Quakish Lake in T3 Indian Purchase, and piping plover (1.15) from Popham Beach in Phippsburg having the highest levels.

Piping plovers at two sites, Ferry Beach (Saco) and Popham Beach (Phippsburg), also have PCB levels greater than 1,000 and may be more sensitive to contaminants. Although effects thresholds have not been determined for piping plovers, a study conducted on a close relative, snowy plovers, indicates that contaminants could be among a number of stressors leading to the decline of least terns and snowy plovers (Hothem and Powell 2000). The authors did conclude, however, that the levels they recorded were not sufficiently elevated to cause concern. The level they recorded for total PCBs (330 – 2,360 ng/g, ww) in snowy plover are similar to our results.

4.4.2 Comparison with other studies

Our total PCB, 2,810-11,138 ng/g (ww), are lower than those detected in eagles in Maine between 1994-1996 (levels from other studies are bold and in brackets) **[330-45,398 ng/g (fww)(Matz 1998)]**, but similar to levels detected along the Penobcot River, Maine **[6,230 – 11,410 ng/g (fww)(Mierzykowski and Carr 2002)]**. However, our results indicate that eagles in Maine have higher levels than those in British Columbia **[1,108 – 7,140 ng/g (ww) Elliot et al. 1996)]**. Our total PCBs in osprey ranged from 501-2,666 ng/g, which is slightly lower than levels observed in New Jersey osprey in 1998 **[1,090-4,450 ng/g (Clark et al. 2001)]**.

Our seabird results are similar to those detected in Maine by Mierzykowski and Carr (2004) **[560 ng/g (fww) in piping plovers, 560 in common terns, and 430 in least tern].** We found the piping plover range is 160-1,876 ng/g (ww), the common tern level

183, and the least tern level 263. Our double-crested cormorant results 1,403-2,413 are within the range reported in neotropic cormorants in Texas **[464-5,720 ng/g (ww) (Frank et al. 2001)]**.

Our results indicate that PCB levels in Maine have decreased since 1977. Szaro et al. (1979) found the mean total PCB in common eiders in 1977 was 1,600 ng/g (ww); our means is 217 ng/g. Similarly, in herring gull eggs they found a mean of 7,760 ng/g (ww) on Appledore Island; we also collected herring gull eggs on Appledore and found 1,176 ng/g (ww) in a three-egg composite. These results are consistent with other studies that have found a decrease in PCBs over time (Williams et al. 1995, Clark et al. 2001, Braune 2007).

4.4.3 Spatial variation (Figure 20)

Spatially, PCBs are distributed across Maine in a similar pattern as PBDEs, PFOS, and HCB. Generally, for the species for which we had more than three samples, highest levels are in the southern coastal region of the state. This trend is likely caused by the greater level of development along the coast and in the southern portion of the state. It also may reflect contaminants being transported to the coast through developed rivers.

Across species there are several locations that are consistently high. Isles of Shoals herring gull, cormorant, and eider PCB levels tended to be higher than other areas. This may be the result of the contaminants coming from the Piscataqua River, Portsmouth, and Kittery. Additionally, this area may have greater atmosphere deposition from southern New England. Similarly, the mouth of the Kennebec River tended to have higher PCB levels in eider, osprey, herring gull, cormorant, and plover. The mouth of the Kennebec drains Merrymeeting Bay, which is at the confluence of both the Androscoggin and Kennebec rivers, and consequently may be where contaminants are being concentrated. Another site with consistently high PCB levels is Seal Island in Penobscot Bay. The higher levels on this island may be the result of the island being used as a bombing range in the past.

4.4.4 Habitat (Figure 26)

Overall the dataset show little variation between habitats. Estuaries have significantly lower PCB levels than marine habitat (ANOVA Tukey HSD, F = 3.4, df = 4,55, p = 0.0147). The tree swallow results showed little variation between habitats, although lake levels are the highest. However, the eagle samples show that the egg from a marine nest has PCB levels more than three times greater than the river and lake samples. This is consistent with eagle studies in Maine which have found significantly higher PCB levels in coastal eagles (Welch, 1994, Matz 1998)

4.5 PBDEs (Figure 9)

4.5.1 Comparison to known effects thresholds

Laboratory study in kestrels found negative physiological effects in chicks that had 1,500 ng/g total PBDE injected into their egg and were fed 100 ng/g per day (Fernie et al. 2005). Our egg total PBDE residues, ranging 3-782 ng/g, are not as high as the kestrel dosing study. Consequently, we do not know if the levels we recorded are having a negative effect.

4.5.2 Comparison with other studies

The range of our results is not consistent with other studies (levels from other studies are bold and in brackets). With some species our levels are significantly higher than other studies, while others are lower. Our black-guillemot results, 50 ng/g (ww), are significantly higher than guillemots eggs in Greenland **[2.5 ng/g (ww) (Vorkamp et al. 2004)]**, and higher than PBDEs in Brunnich's guillemot in Norway **[27 ng/g (ww) (Murvoll 2006)]**. Contaminants in yolk have greater concentrations than whole eggs. In our study we analyzed whole egg.

Our herring gulls results, 234 ng/g (ww), are higher than the slightly larger glaucous gull (*Larus hyperboreus*) in Norway **[52.9 ng/g, ww (Verreault et al. 2004)]**, but lower than herring gull eggs from the Great Lakes **[662 ng/g, (ww) (Norstom et al. 2002)]**. Our tern results, however, on a lipid weight basis are an order of magnitude lower than tern samples from San Francisco Bay, an area that has recorded some of the highest PBDE levels in women in the world (She et al. 2004). Our eider results, 5.82 ng/g (ww), are higher than those in egg yolk in Norway **[0.4 ng/g (ww)(Murvoll 2006)]**. Our cormorants results, 31.12 ng/g (ww), are similar to European shag (*Phalacrocorax aristotelis*) in Norway **[17 ng/g (ww, yolk)]**, and double-crested cormorant in British Columbia (Elliot et al. 2005). Our Leach storm-petrel results, 13.35 ng/g (ww), are slightly higher than those in British Columbia **[3.38 ng/g (ww) (Elliot et al. 2005)]**.

Elliot et al. (2005) reports that osprey PBDE levels have risen from 7.84 & 18.4 in 1991 to 162 & 185 ng/g (ww) in 2000. Our osprey results are nearly identical, 185 ng/g (ww), to Elliot et al. 2000 levels, and similar to Norwegian osprey **[103 ng/g (ww) (Herzke et al. 2005)]**. Our peregrine falcon level, 149.06 ng/g (ww) are also consistent with Norwegian **[155 ng/g (ww) (Herzke et al. 2005)]**, and Swedish peregrines (Sellstrom et al. 2004), but were generally lower than southern New England (Chen et al. 2007). However, our bald eagle levels trended higher, 440.10 ng/g (ww) than white-tailed sea eagle in Norway **[184 ng/g (ww) (Herzke et al. 2005)]**, but were consistent with eggs analyzed in Maine **[mean 577 ng/g (ww), range 226-952 (USFWS unpublished data)]**.

4.5.3 Spatial variation (Figure 21)

PBDEs are distributed across Maine in a similar pattern as PCBs and PFOS, tending to be higher in coastal Maine between Mount Desert Island and Isles of Shoals. Penobscot Bay in particular has higher levels in osprey, guillemot, and eider. These higher levels likely reflect PBDEs that are transported down the Penobscot River. Another area of generally higher levels is the Portland Area. This pattern is expected, because PBDE can be transported into the environment from atmospheric deposition, in household dust, incinerators, and water treatment facilities. Like many of the other contaminants, Isles of Shoals PBDE levels are higher than other locations for reasons describes above. Additionally, like PCBs the loon egg from Aziscohos Lake (western Maine) has higher PBDE levels, possibly the result of historical dumping.

4.5.4 Habitat (Figure 26)

Overall there are no statistical differences between habitats (ANOVA, F = 2.3, df = 4, 55, p = 0.06). However, estuaries are lower than marine habitats and the kingfisher and eagle residues from riverine habitat tended to be higher than the other habitats. Generally, the tree swallow data showed little variation between habitats, although the terrestrial birds tended to have higher levels.

4.5.5 Congener patterns (Figure 16, 17)

Although BDE 47, 99, 100 made up the majority of the samples for most species, there is great variation in the pattern from species to species. This indicates that PBDEs may be entering environment, dispersing, and bioaccumulating in different patterns between food webs and habitats. Research has demonstrated that bacteria can cause deca to breakdown into the more toxic lower brominated congeners (He et al. 2006); consequently the levels of tetra- and octa-BDE that we recorded may have originated from deca.

Terrestrial predators have a dramatically different congener patterns than the other habitats. Specifically, BDE47 composed a lower percentage of the samples while the higher brominated congeners 196, 197, 207, and 209 have a higher percentage. The terrestrial samples are composed of peregrine falcon, American kestrel, and tree swallow. This shows conclusively that higher brominated congeners can bioaccumulate in wildlife.

4.6 PFC (Figure 10)

4.6.1 Comparison to known effects thresholds

PFCs have only recently been identified as a persistent bioaccumulative contaminant of concern. Consequently, few studies have been conducted on effects in bird eggs.

However, a study that injected perfluorooctane sulfonate (PFOS) in white leghorn chicken eggs—known to be particularly sensitive to contaminants—determined, based on reduced hatchability, that the lowest-observed adverse-effects level (LOAEL) was 0.1 ug/g or 100 ng/g (ww). The species we studied may be either more or less sensitive than the chickens.

Twenty-three of our composite samples have PFOS values above the LOAEL of 100 ng/g (ww) including the following species: Atlantic puffin, bald eagle, belted kingfisher, common eider, common loon, double-crested cormorant, great black-backed gull, osprey, peregrine falcon, piping plover, and tree swallow. Two samples are considerably higher than the rest: eagle, 710.53 ng/g (ww) and kingfisher, 954.76 ng/g (ww).

4.6.2 Comparison with other studies

Only one study looked specifically at PFOS in bird eggs and our results are comparable (levels from other studies are bold and in brackets) to double-crested cormorants in the Great Lake region **[157 ng/g (ww), yolk]** and ring-billed gull **[67 ng/g (ww, yolk) (Kannan et al. 2001)]**. As noted above (section 4.5.2) whole egg contaminant levels are lower than yolk levels. In our study, cormorants have a mean PFOS level of 126.13 ng/g, herring gulls 40.66, and great black-backed gulls 78.68.

Kannan et al. (2001) screened the livers of many of the same species as we did in our study. Liver generally has higher contaminant levels than eggs in organochlorines (Mason et al. 1997), in DDE (Norstrom et al. 1986), and in Hg (Evers et al. 2005), but liver and egg have similar PCB levels (Hoffman et al. 1996). Kannan et al. (2002) liver results compare to our eggs as follows: eagles in our study 103.82-710.53 **[24-648 ng/g, ww]**, in loons 30.90-186.06 **[8.6-595]**, in cormorants 96.59-177.95 **[51-288]**, in great black-backed gull 46.19 - 111.18 **[187-608]**, in osprey 60.27-441.18 **[42-959]**, in herring gull 11.67-95.65 **[16-353]**, and snowy egret 88.50 **[43]**. Although the liver and egg values cannot be directly compared, the overlap between our values and the liver results, indicate that Maine PFOS levels are similar to other locations in the U.S., and potentially that our results are higher than other locations.

Our results also are higher or comparable to PFOS in liver of fulmar (*Fulmarus glacialis*) in the Faroe Islands **[24 ng/g (ww) (Bossi et al. 2005)]**; shorebirds, seabirds, waterfowl, and raptors in Japan **[10-650 ng/g (ww) (Kannan et al. 2002)]**; pelicans in Columbia, **[36.65 ng/g (ww) (Oliver-Verbal et al. 2006)]**; and sea otter on California **[<1 – 884 ng/g (ww) (Kannan et al. 2006)]**.

4.6.3 Spatial variation (Figure 22)

Similar to PCBs, PBDEs, and HCBs, PFOS tend to be higher in southern coastal Maine. As with the other compounds the mouth of the Kennebec has higher levels in herring gull, cormorant, eider, osprey, and piping plover. Similarly, the Portland area and Isles of

Shoals have relatively higher levels for reasons described above. Also similar to PCBs and PBDEs, we detected high PFOS in the coastal eagle egg (only second to the kingfisher sample).

4.6.4 Habitat (Figure 26)

Like other compounds, the estuary has statistically significantly lower PFOS than other habitats (ANOVA, Tukey HSD, F = 2.43, df = 4, 59, p = 0.058). Similar to the PBDE findings, the riverine habitat has the highest levels, which are driven by the belted kingfisher and eagle results. The kingfisher eggs collected from the Presumscot River have the highest levels recorded in this study, and are high compared to other studies (see above). Proximity to urban development and industrial areas may causes these higher PFOS levels. The tree swallow data showed a variation between habitats: terrestrial is the highest, while estuary is the lowest.

4.6.5 Congener patterns (Figure 18)

Initially we have focused on PFOS because research has documented that this congener bioaccumulates (Kannan et al. 2002). Our results indicate that for most species greater than 50% of total PFC are comprised of PFOS. Arctic and common tern and common loon deviated from this pattern and have a greater portion of PFUnDA. These differences suggest that PFCs may be inputted into the environment through different mechanisms (i.e. atmosphere deposition, point source).

4.7 Organochlorine pesticides (Figure 11, 12, 13)

4.7.1 Comparison to known effects thresholds

Although the OCs tested are present in all species (except HCH), the samples are well below known effects thresholds. HCH are not detected in any samples. This is consistent with other studies that have not detected HCH, because it has a short half-life (Blus 2003).

Our HCB residues range for all species is 0.75 – 20.33 ng/g (ww), which is significantly below the effects threshold of 35,000 ng/g (ww) (Wiemeyer 1996). Our chlordane residues range for all species is 1.81 – 259.51 ng/g (ww), which is significantly below the effects threshold of 2,000 ng/g (ww) (Blus 2003). Our DDE residues range for all species is 9.91 – 2,072.44 ng/g (ww), which is significantly below the effects threshold of 3,000 - 30,000 ng/g (ww) (Blus 2003); however, studies indicate that slight egg shell thinning is possible at lower levels. Depending on the species, no eggshell thinning is seen below 100 to 2000 ng/g (Blus 1996).

4.7.2 Comparison with other studies

The levels of OC measured in our study are generally in the range detected in other studies (levels from other studies are bold and in brackets). Our HCB and DDE levels in piping plover, least tern, and common tern are nearly identical to residues detected in these species in a 2003 Maine study (Mierzykowski and Carr 2004). Our HCB levels are similar to those of British Columbia bald eagles **[1-25 ng/g, ww (Elliot et al. 1996)]**. Our chlordane and DDE residues are at similar or below levels recorded in Arctic seabirds (Braune et al. 2002, Braune 2007) and our chlordane Norwegian birds of prey (Herzke et al. 2002). The DDE levels of New Jersey osprey are higher than our osprey findings **[930 ng/g ww]**, but comparable to our eagle and peregrine falcon levels (Clark et al. 2001). Bald eagle eggs in British Columbia from 1990-92 have higher mean DDE levels **[2,170 to 5,140]** as well as higher chlordane residues than our eagle samples (Elliot et al. 1996).

4.7.3 Spatial variation (Figure 23, 24, 25)

Although not consistent amongst all species, coastal Maine—from Kittery to Penobscot Bay—generally have higher HCB, chlordane, and DDE levels than the rest of the state. HCB levels showed a strong trend towards higher levels between Casco Bay and Mount Desert Island. This trend may be caused by the major river flowages transporting historical chemical into the marine system in the more developed portion of Maine. This area may also receive higher levels of atmospheric deposition than other areas in Maine.

4.7.4 Habitat (Figure 26)

Marine birds have significantly higher HCB levels than estuarine birds (ANOVA, Tukey HSD, $F = 3.1$; df, 4, 55, $p = 0.02$, Figure 18); there is no difference between the other habitats. This trend is followed with the tree swallow results although the lake habitat has the highest levels. The reason for this trend is not clear.

Riverine birds have significant higher chlordane levels than marine birds (ANOVA, Tukey HSD, $F = 3.4$; df, 4, 55, $p = 0.01$, Figure 18); there is no difference between the other habitats. Since we were unable to collect riverine tree swallow samples we cannot directly compare to the overall results. However, marine swallows did have the lowest chlordane residues. The riverine samples are dominated by belted kingfisher and bald eagle samples. The birds may be exposed to higher chlordane levels because chlordane residues are still present in terrestrial environments and continue to wash into rivers. The higher levels in terrestrial birds supports this supposition.

Riverine birds have significantly higher DDE residues than estuarine birds (ANOVA, Tukey HSD, $F = 2.8$; df, 4, 55, $p = 0.03$, Figure 18); there is no difference between the other habitats. Since we were unable to collect riverine tree swallow samples we cannot directly compare to the overall results. However, like chlordane, DDE may still be bound up in soils and be leaching into riverine systems. The higher levels in terrestrial birds support this supposition.

4.8 Portland area break out (Figure 27)

By collecting multiple species within the same area, we are able to gain insight into bioaccumulation and various pathways through which the contaminants are entering the environment. A clear trend for all of the contaminants is that the higher trophic level predators such as peregrine falcon, common loon, great black-backed gull, and osprey tend to have higher levels than the other species. However, the species with the highest levels are not uniform across the different contaminates. Since the birds have different foraging strategies, these differences provide insight into contaminant sources.

4.8.1 Hg

As stated above, our results are consistent with known patterns of Hg availability. The top trophic level predators such as loons, which forage exclusively in the lacustrine (lakes) ecosystem, have the highest Hg levels. This is consistent with studies that show higher rates of methylation in freshwater systems because they provide ideal habitat for sulfur reducing bacteria. An interesting finding is that piping plovers have similar Hg levels to osprey and cormorants.

4.8.2 PCB

Matz (1998) and Welch (1994) found that coastal eagles had higher PCB levels than birds feeding from rivers and lakes. The samples from Portland also follow this pattern. Of the species sampled, those feeding in the marine system have higher levels than those on lakes and rivers. The one exception is peregrine falcon, which also has higher levels; this is likely explained by their higher trophic status. Of note is that the piping plovers have PCB levels greater than herring gulls.

4.8.3 PBDEs

These results follow a similar pattern to the PCBs and are consistent with the high correlation between PCBs and PBDEs. Great black-backed and herring gull both have high levels, perhaps associated with foraging close to urban development. Osprey also have high levels and since they feed exclusively on fish this indicates that PBDEs are bioaccumulating in near-shore marine fish. Of note is that piping plovers have higher levels than the other coastal invertivores.

4.8.4 PFOS

Most species have similar levels, the belted kingfisher being the notable exception. As described above, these samples were collected from an industrialized river, the Presumscot. Since their levels are more than twice those of other species, their PFOS residues indicate that fish in the river may be bioaccumulating PFOS at a greater rate than the fish in the marine system.

4.8.5 OCs

These follow a similar pattern as the dataset as a whole.

4.9 Overall conclusions

We found both established (Hg, PCBs, chlordane, HCB, DEE) and emerging (PBDEs, PFCs) bioaccumaltive toxic pollutants of concern in all the bird eggs we analyzed. Our results are the first records of PFCs in Maine birds. Since the birds we selected act as bioindicators of multiple ecosystems across the state, our results indicate that the compounds we measured are present in the offshore marine, coastal marine, estuarine, riverine, lacustrine, and terrestrial ecosystems. Although we found most of the compounds across the entire state, there tended to be higher levels in coastal southern Maine. This geographic pattern suggests that these compounds are entering the environment both through atmospheric deposition, because they are found across the entire state, and through local point sources, because we detected higher levels in urban and industrial areas. In particular, several areas consistently have higher levels than the rest of the state: Isles of Shoals, Portland, and the mouth of the Kennebec.

As expected, a number of loon eggs have Hg levels above known effects thresholds. One eagle egg has PCB levels within a range of known effects, although the congener pattern is not dominated by the most toxic PCBs. Since no effects threshold have been established for PBDEs in bird eggs, the residues we detected may or may not have negative effects. Twenty-three of our samples have PFOS levels above effects threshold established for chicken eggs—the species we studied may be more or less sensitive than the chickens. OCs are all substantially below effects thresholds.

Our Hg results are consistent with other studies conducted in the region. Our PCB results are also consistent with those across the United States, and when compared to earlier studies in Maine, herring gull, common eider, and bald eagle all have lower levels than in the past. Overall our PBDE result are not consistently higher or lower than other areas across the globe, but some species had higher or lower PBDEs than in other areas. Like recent studies on terrestrial birds we detected higher brominated PBDEs, including decaBDE, in terrestrial predators: American kestrel and peregrine falcon. Only one study has analyzed PFOS in bird eggs of two species in the Great Lakes region. Our results are similar to these. Moreover, although a direct comparison is not possible, our results are consistent with PFOS in the liver of multiple species around the world. Our OC results are generally within the range of other studies.

Our results show that many of the compounds we measured increase in concert with each other. The strongest relationship we found is between PCBs and PBDEs, indicating that species and areas with high PCB levels may also have high PBDE levels. These relationships suggest that some species may have higher levels simultaneously of multiple compounds, which together may have greater negative impact on reproductive success, the neurological system, endocrine function, and overall physiology. Consequently, high trophic level predators may have a combined negative effect of these compounds despite having individual contaminants below known effects thresholds.

In general our results followed the expected pattern- with high trophic level predators having the highest overall contaminant levels. Bald eagle in particular have PCB, PBDEs, PFCs, chlordane, and DDE multiple times higher than other species. Two species did not follow the expected pattern: belted-kingfisher and piping plover. The kingfisher eggs were collected from an urbanized river that may have overall higher pollution levels than other sites where we collected samples; if we were able to collect samples from other species at this site, their levels also may have been higher. The reason for the higher contaminant levels in plovers in not clear.

Estuaries consistently have lower levels of all the compounds. However, these results may be confounded by the lower trophic level of species we collected samples from in estuaries. As expected, lakes have higher levels of Hg than other habitats, and PCBs have higher levels along the coast.

In summary, our results indicate that both historical and emerging chemicals of concern are accumulating in birds that forage in diverse ecosystems across the entire state of Maine.

5. ACKNOWLEDGEMENTS

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6. FIGURES

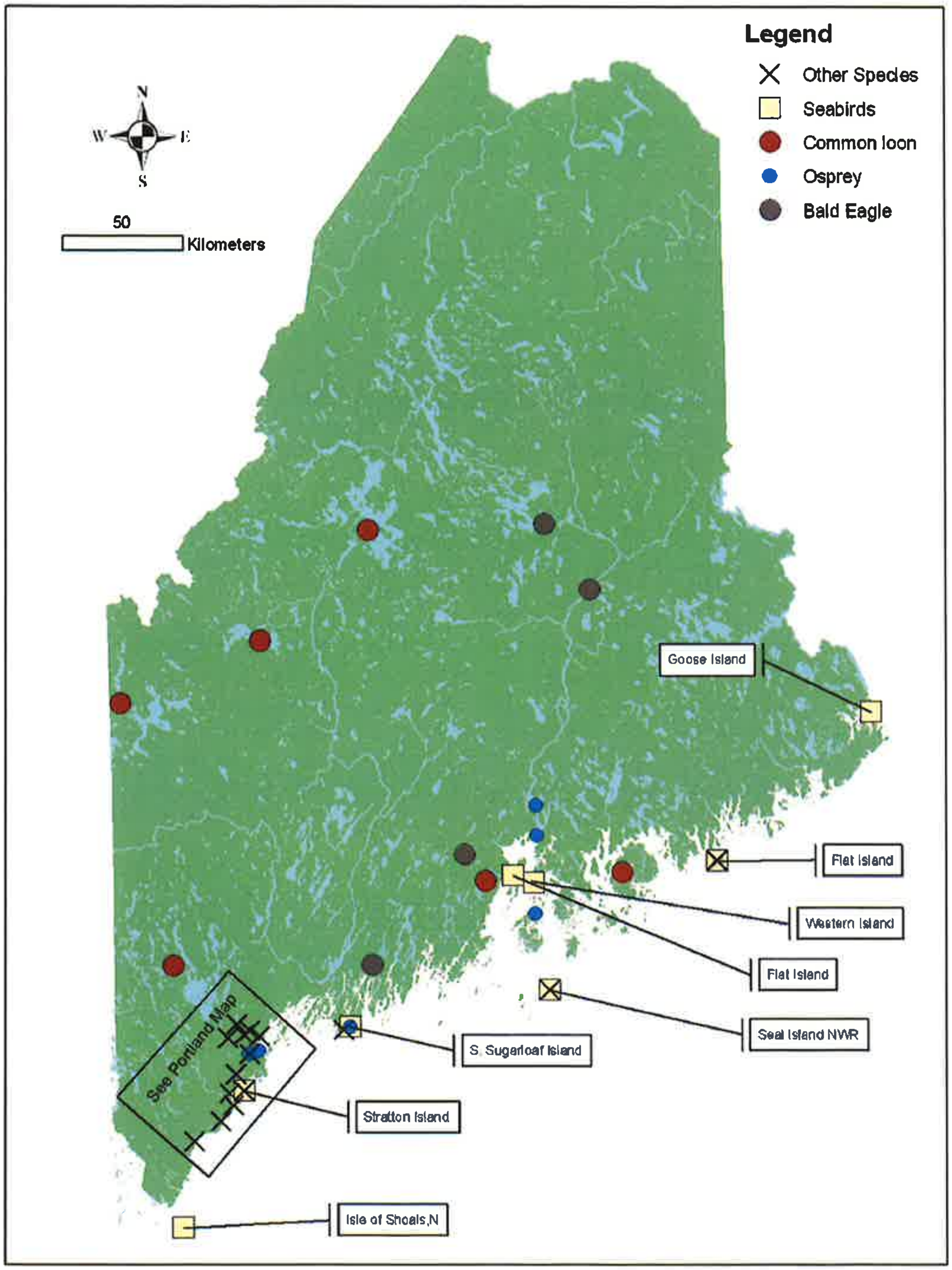


Figure 1. Maine sampling effort

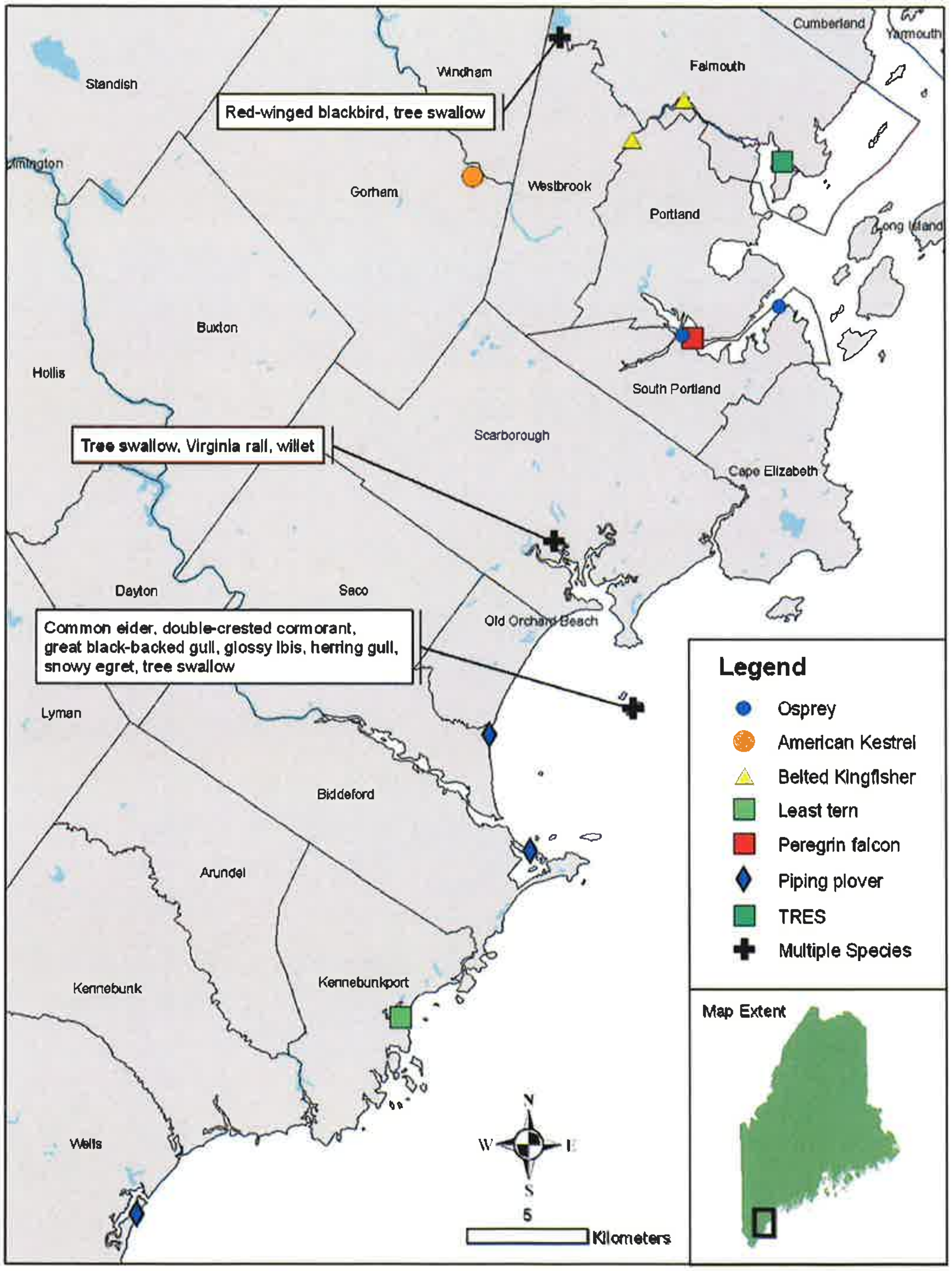


Figure 2. Portland sampling effort

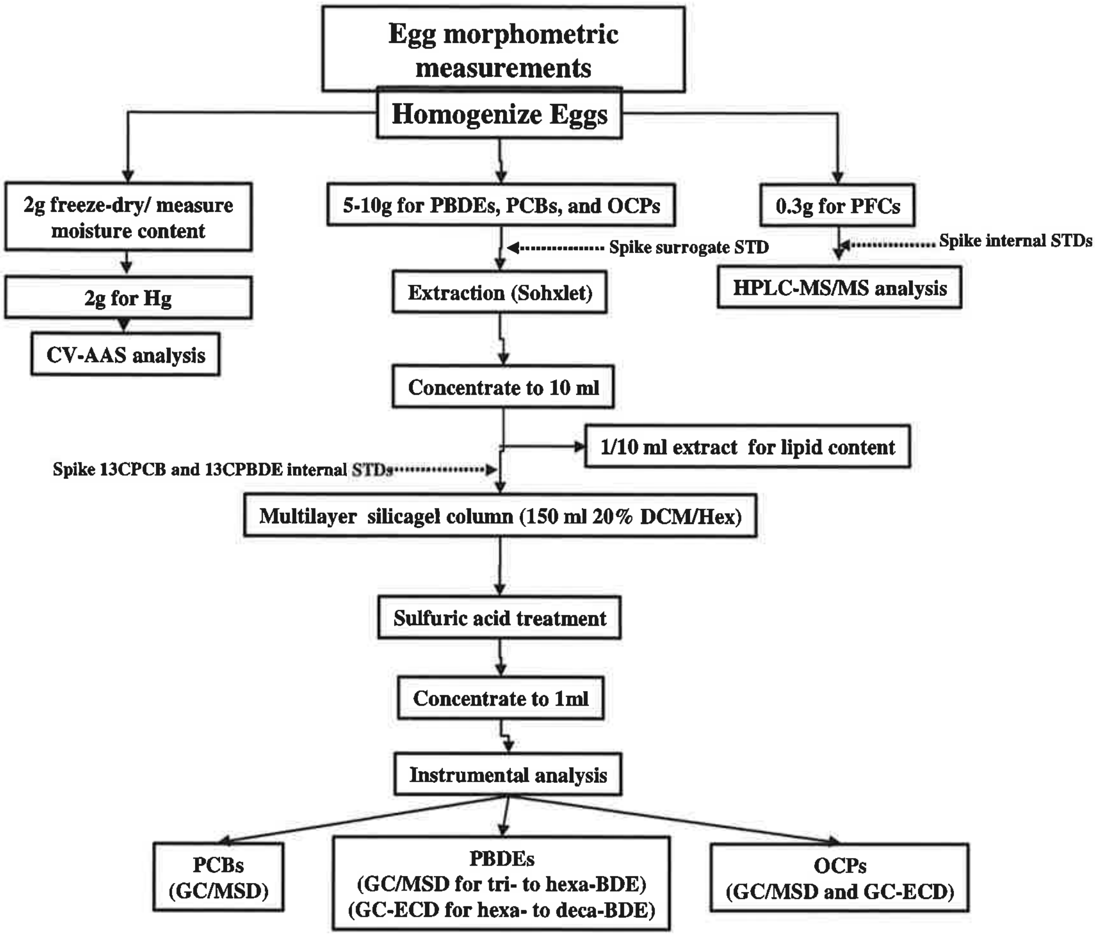
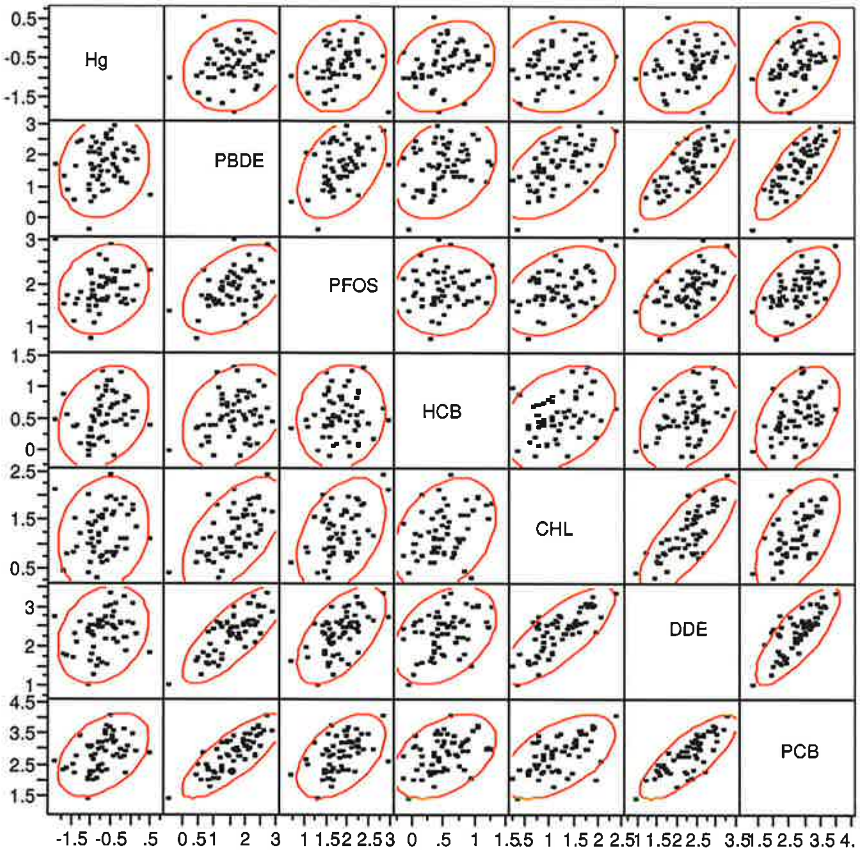
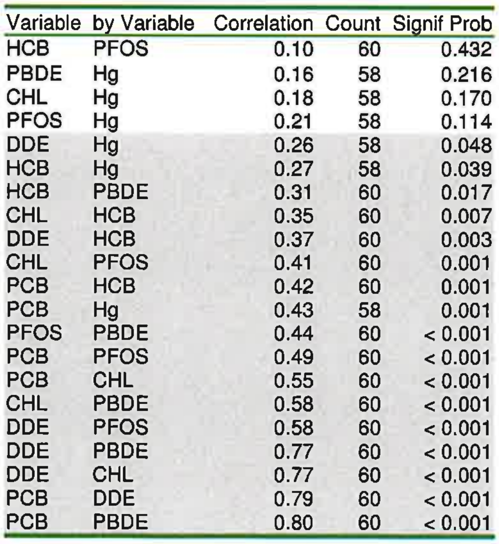


Figure 3. Analytical scheme for the determination of PCBs, PBDEs, organochlorine pesticides, perfluorinated compounds and mercury in the eggs of birds from Maine, USA, 2007.





Variable	by Variable	Correlation	Count	Signif Prob
HCB	PFOS	0.10	60	0.432
PBDE	Hg	0.16	58	0.216
CHL	Hg	0.18	58	0.170
PFOS	Hg	0.21	58	0.114
DDE	Hg	0.26	58	0.048
HCB	Hg	0.27	58	0.039
HCB	PBDE	0.31	60	0.017
CHL	HCB	0.35	60	0.007
DDE	HCB	0.37	60	0.003
CHL	PFOS	0.41	60	0.001
PCB	HCB	0.42	60	0.001
PCB	Hg	0.43	58	0.001
PFOS	PBDE	0.44	60	< 0.001
PCB	PFOS	0.49	60	< 0.001
PCB	CHL	0.55	60	< 0.001
CHL	PBDE	0.58	60	< 0.001
DDE	PFOS	0.58	60	< 0.001
DDE	PBDE	0.77	60	< 0.001
DDE	CHL	0.77	60	< 0.001
PCB	DDE	0.79	60	< 0.001
PCB	PBDE	0.80	60	< 0.001

Figure 4. Correlation between compounds. In the graph, the stronger relationships have tight ovals while poor relationships have circles. The closer the correlation value is to 1 the stronger the relationship. Rows highlighted in gray are significantly related.

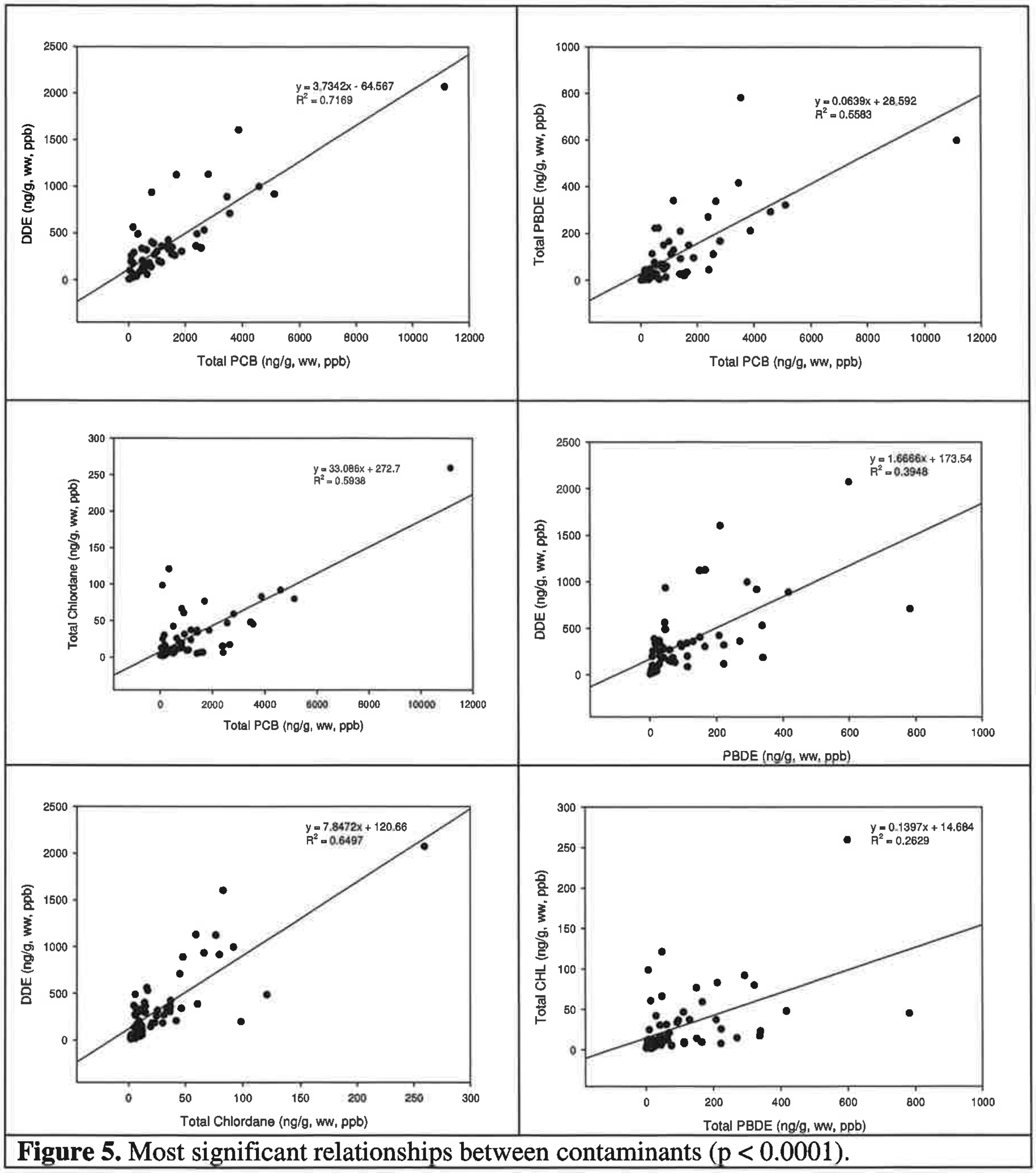


Figure 5. Most significant relationships between contaminants ($p < 0.0001$).

Figure 6. Rank of total contaminants. Species key and sample size below.

Species	Species Code	N
American kestrel	AMKE	1
Arctic tern	ARTE	1
Atlantic puffin	ATPU	1
Bald eagle	BAEA	4
Belted kingfisher	BEKI	2
Black guillemot	BLGU	3
Common eider	COEI	6
Common loon	COLO	6
Common tern	COTE	1
Double-crested cormorant	DCCO	5
Great black-backed gull	GBBG	2
Glossy ibis	GLIB	1
Herring Gull	HERG	6
Least tern	LETE	1
Leach's storm-petrel	LHSP	1
Osprey	OSPR	6
Peregrine falcon	PEFA	1
Piping plover	PIPL	4
Red-winged blackbird	RWBL	1
Snowy egret	SNEG	1
Tree swallow	TRES	4
Virginia rail	VIRA	1
Willet	WILL	1

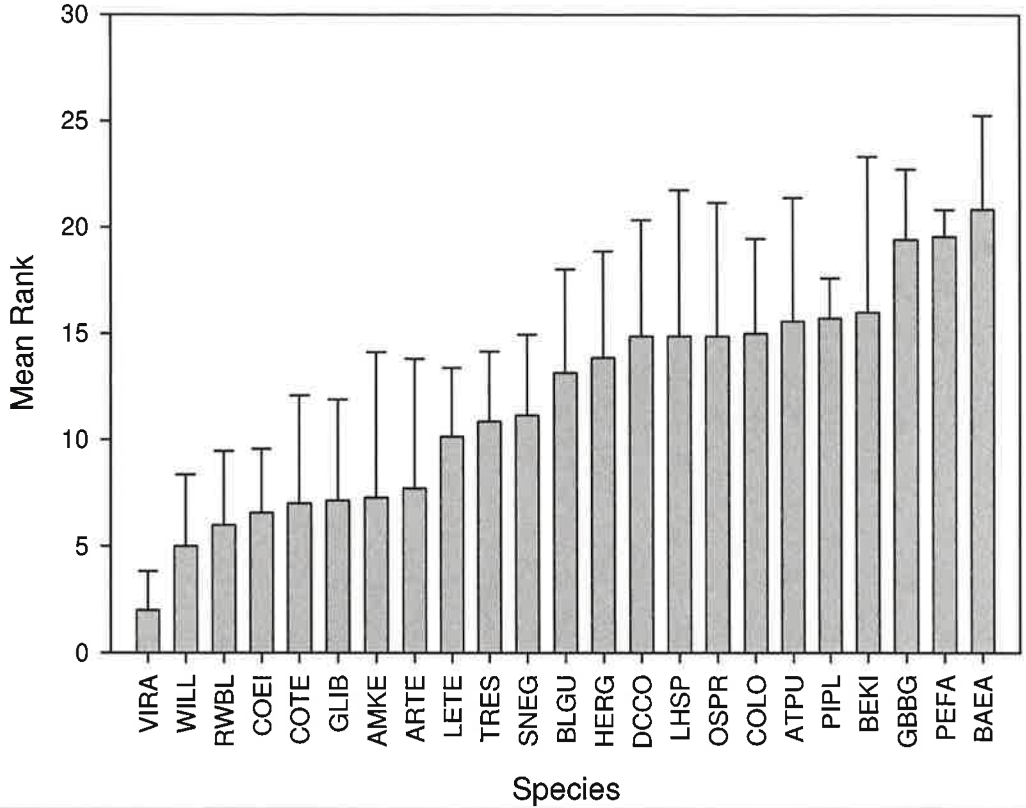
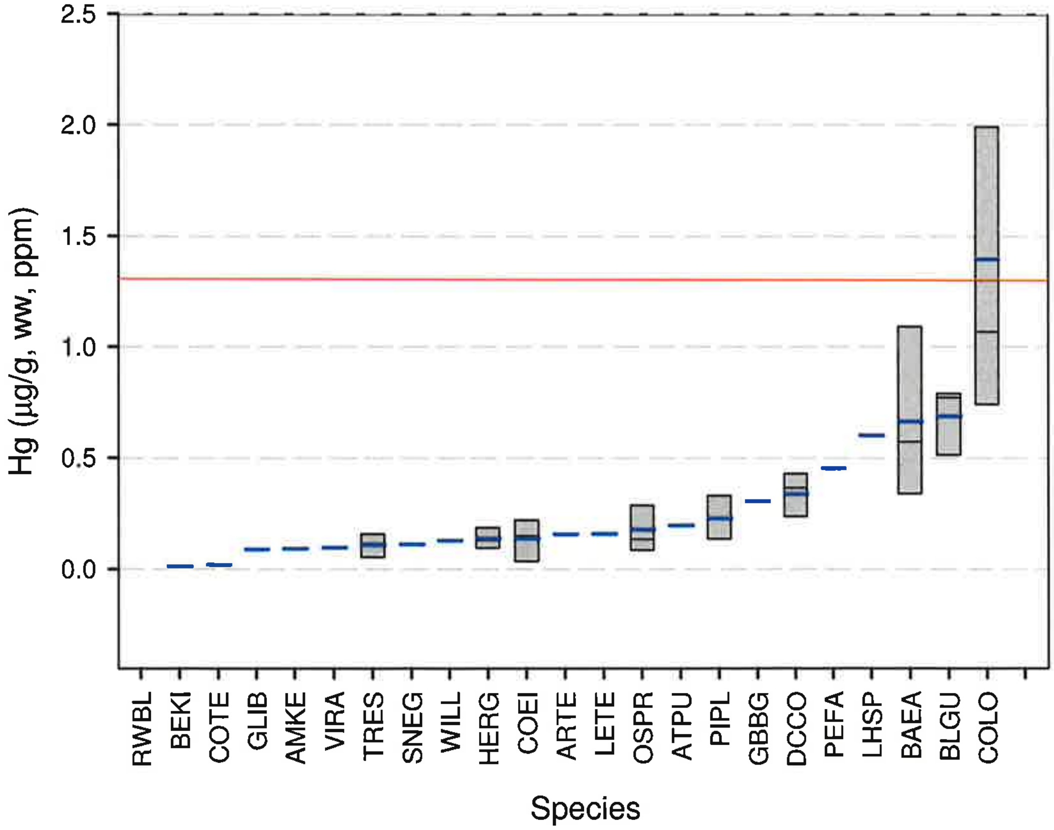


Figure 7. Hg levels by species. Blue line is mean, black line is the median, and box boundary is data range. Red line is adverse effects threshold for loon eggs, 1.3 µg/g, wet weight (Evers et al. 2003).

Species Key (sample size): **American kestrel**, AMKE, (n = 1); **Arctic tern**, ARTE, (n = 1); **Atlantic puffin**, ATPU, (n = 1); **bald eagle**, BAEA, (n = 4); **belted kingfisher**, BEKI, (n = 2); **black guillemot**, BLGU, (n= 3); **common eider**, COEI, (n = 6); **common loon**, COLO, (n = 6); **common tern**, COTE, (n = 1); **double-crested cormorant**, DCCO, (n = 5); **great black-backed gull**, GBBG, (n = 2); **glossy ibis**, GLIB, (n = 1); **herring gull**, HERG, (n = 6); **least tern**, LETE, (n = 1); **Leach's storm-petrel**, LHSP, (n = 1); **osprey**, OSPR, (n = 6); **peregrine falcon**, PEFA, (n = 1); **piping plover**, PIPL, (n = 4); **red-winged blackbird**, RWBL, (n = 1); **snowy egret**, SNEG, (n = 1); **tree swallow**, TRES, (n = 4); **Virginia rail**, VIRA, (n = 1); and **willet**, WILL, (n = 1).



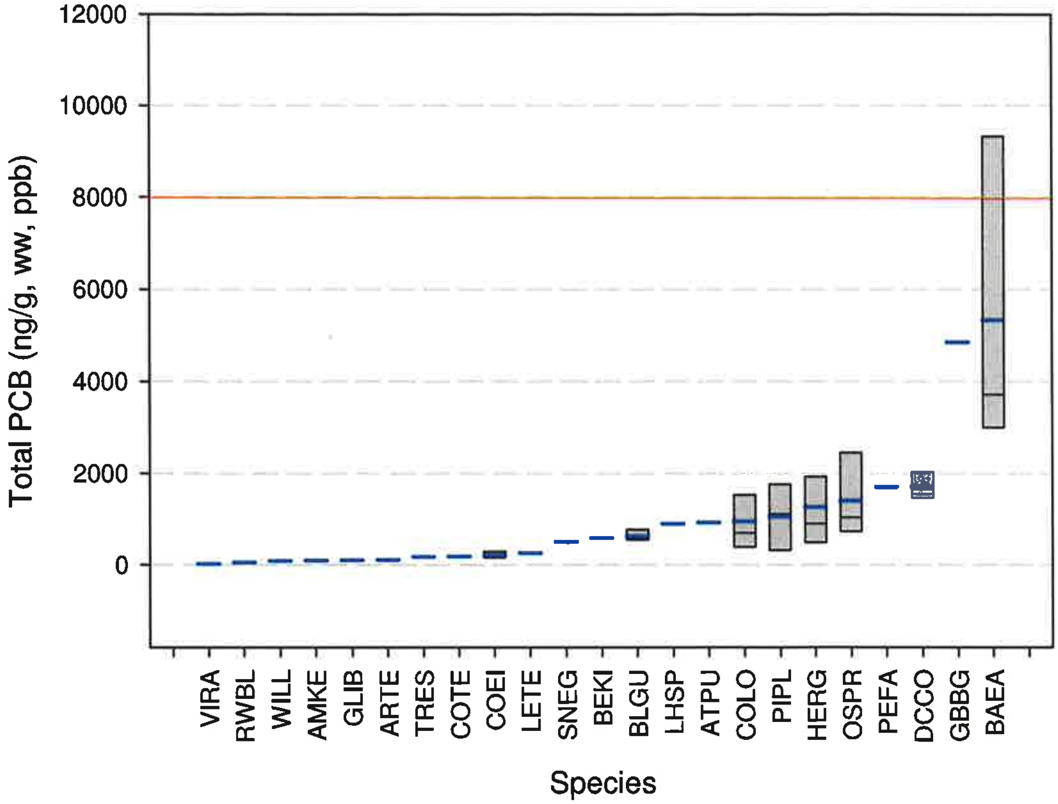


Figure 8. Total PCB levels by species. Blue line is mean, black line is the median, and box boundary is data range. Red line is adverse effects threshold.

Species Key (sample size): **American kestrel**, AMKE, (n = 1); **Arctic tern**, ARTE, (n = 1); **Atlantic puffin**, ATPU, (n = 1); **bald eagle**, BAEA, (n = 4); **belted kingfisher**, BEKI, (n = 2); **black guillemot**, BLGU, (n= 3); **common eider**, COEI, (n = 6); **common loon**, COLO, (n = 6); **common tern**, COTE, (n = 1); **double-crested cormorant**, DCCO, (n = 5); **great black-backed gull**, GBBG, (n = 2); **glossy ibis**, GLIB, (n = 1); **herring gull**, HERG, (n = 6); **least tern**, LETE, (n = 1); **Leach's storm-petrel**, LHSP, (n = 1); **osprey**, OSPR, (n = 6); **peregrine falcon**, PEFA, (n = 1); **piping plover**, PIPL, (n = 4); **red-winged blackbird**, RWBL, (n = 1); **snowy egret**, SNEG, (n = 1); **tree swallow**, TRES, (n = 4); **Virginia rail**, VIRA, (n = 1); and **willet**, WILL, (n = 1).

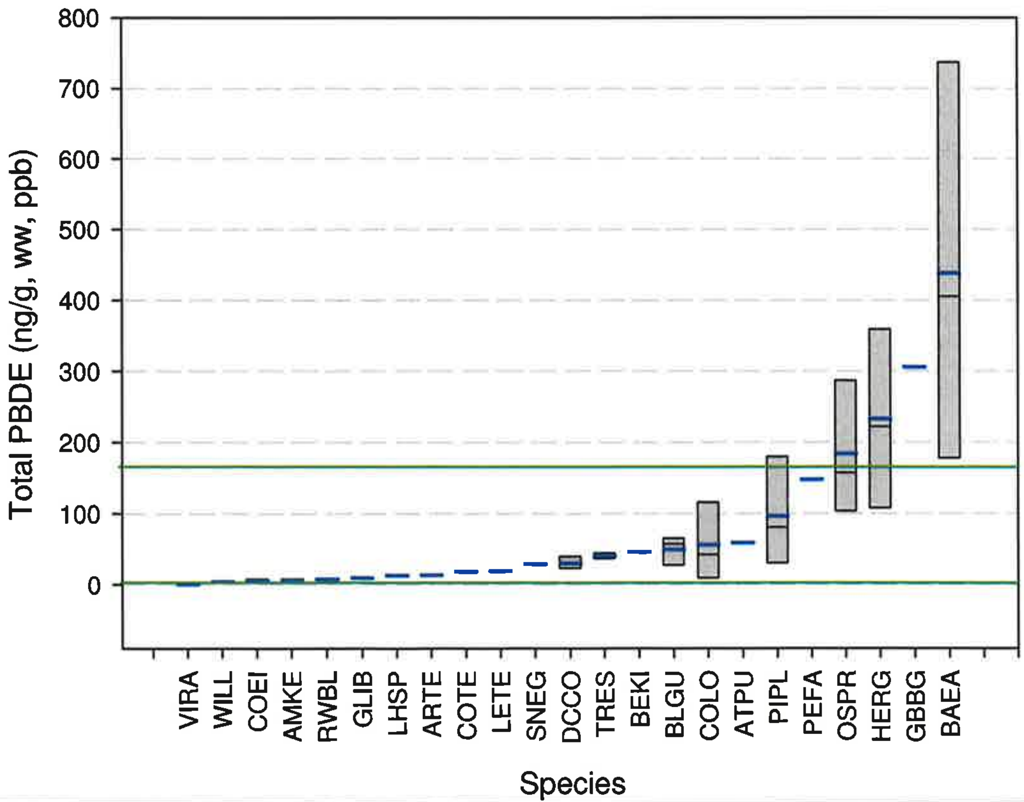


Figure 9. Total PBDE by species. Blue line is mean, black line is the median, and box boundary is data range. Between green lines is mean range of total PBDE in six species of Norwegian predatory bird eggs (Herzke et al. 2005).

Species Key (sample size): **American kestrel**, AMKE, (n = 1); **Arctic tern**, ARTE, (n = 1); **Atlantic puffin**, ATPU, (n = 1); **bald eagle**, BAEA, (n = 4); **belted kingfisher**, BEKI, (n = 2); **black guillemot**, BLGU, (n= 3); **common eider**, COEI, (n = 6); **common loon**, COLO, (n = 6); **common tern**, COTE, (n = 1); **double-crested cormorant**, DCCO, (n = 5); **great black-backed gull**, GBBG, (n = 2); **glossy ibis**, GLIB, (n = 1); **herring gull**, HERG, (n = 6); **least tern**, LETE, (n = 1); **Leach's storm-petrel**, LHSP, (n = 1); **osprey**, OSPR, (n = 6); **peregrine falcon**, PEFA, (n = 1); **piping plover**, PIPL, (n = 4); **red-winged blackbird**, RWBL, (n = 1); **snowy egret**, SNEG, (n = 1); **tree swallow**, TRES, (n = 4); **Virginia rail**, VIRA, (n = 1); and **willet**, WILL, (n = 1).

Figure 10. PFOS levels by species. Blue line is mean, black line is the median, and box boundary is data range. Red line is lowest-observed-adverse-effects level for leghorn chicken, 100 ng/g, wet weight (Molina et al. 2006).

Species Key (sample size): **American kestrel**, AMKE, (n = 1); **Arctic tern**, ARTE, (n = 1); **Atlantic puffin**, ATPU, (n = 1); **bald eagle**, BAEA, (n = 4); **belted kingfisher**, BEKI, (n = 2); **black guillemot**, BLGU, (n= 3); **common eider**, COEI, (n = 6); **common loon**, COLO, (n = 6); **common tern**, COTE, (n = 1); **double-crested cormorant**, DCCO, (n = 5); **great black-backed gull**, GBBG, (n = 2); **glossy ibis**, GLIB, (n = 1); **herring gull**, HERG, (n = 6); **least tern**, LETE, (n = 1); **Leach's storm-petrel**, LHSP, (n = 1); **osprey**, OSPR, (n = 6); **peregrine falcon**, PEFA, (n = 1); **piping plover**, PIPL, (n = 4); **red-winged blackbird**, RWBL, (n = 1); **snowy egret**, SNEG, (n = 1); **tree swallow**, TRES, (n = 4); **Virginia rail**, VIRA, (n = 1); and **willet**, WILL, (n = 1).

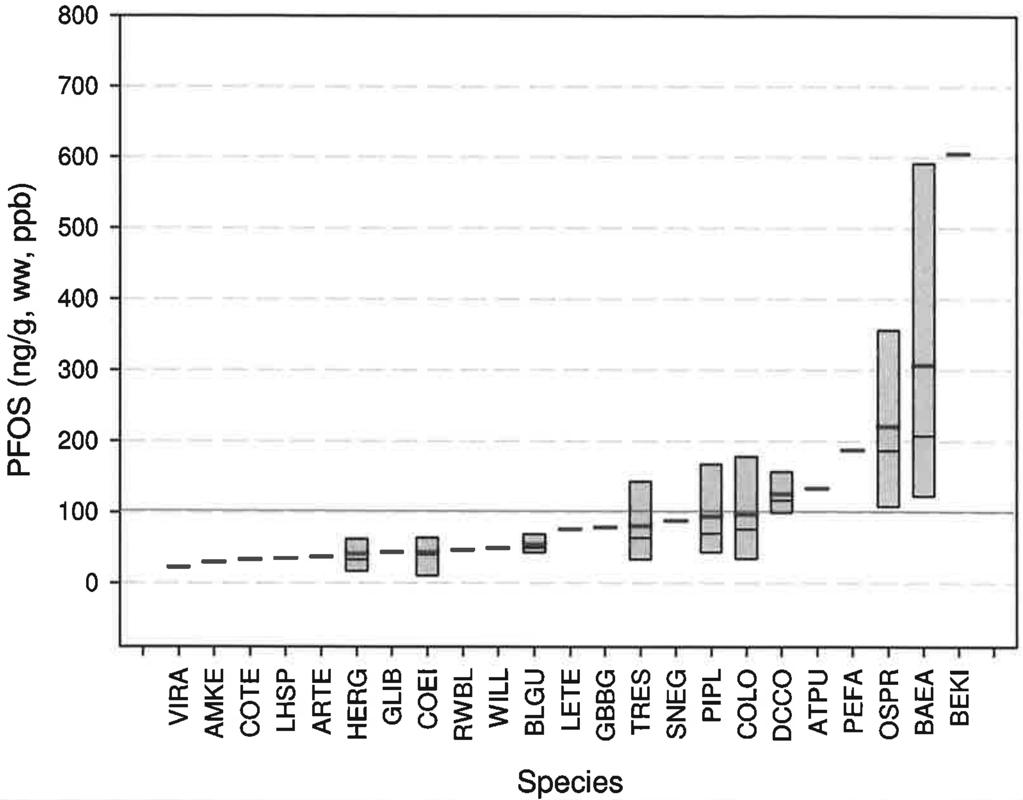


Figure 11. HCB levels by species. Blue line is mean, black line is the median, and box boundary is data range. All levels fall well below the 35,000 (ng/g, ww) effects threshold for Japanese quail (Wiemeyer 1996).

Species Key (sample size): **American kestrel**, AMKE, (n = 1); **Arctic tern**, ARTE, (n = 1); **Atlantic puffin**, ATPU, (n = 1); **bald eagle**, BAEA, (n = 4); **belted kingfisher**, BEKI, (n = 2); **black guillemot**, BLGU, (n= 3); **common eider**, COEI, (n = 6); **common loon**, COLO, (n = 6); **common tern**, COTE, (n = 1); **double-crested cormorant**, DCCO, (n = 5); **great black-backed gull**, GBBG, (n = 2); **glossy ibis**, GLIB, (n = 1); **herring gull**, HERG, (n = 6); **least tern**, LETE, (n = 1); **Leach's storm-petrel**, LHSP, (n = 1); **osprey**, OSPR, (n = 6); **peregrine falcon**, PEFA, (n = 1); **piping plover**, PIPL, (n = 4); **red-winged blackbird**, RWBL, (n = 1); **snowy egret**, SNEG, (n = 1); **tree swallow**, TRES, (n = 4); **Virginia rail**, VIRA, (n = 1); and **willet**, WILL, (n = 1).

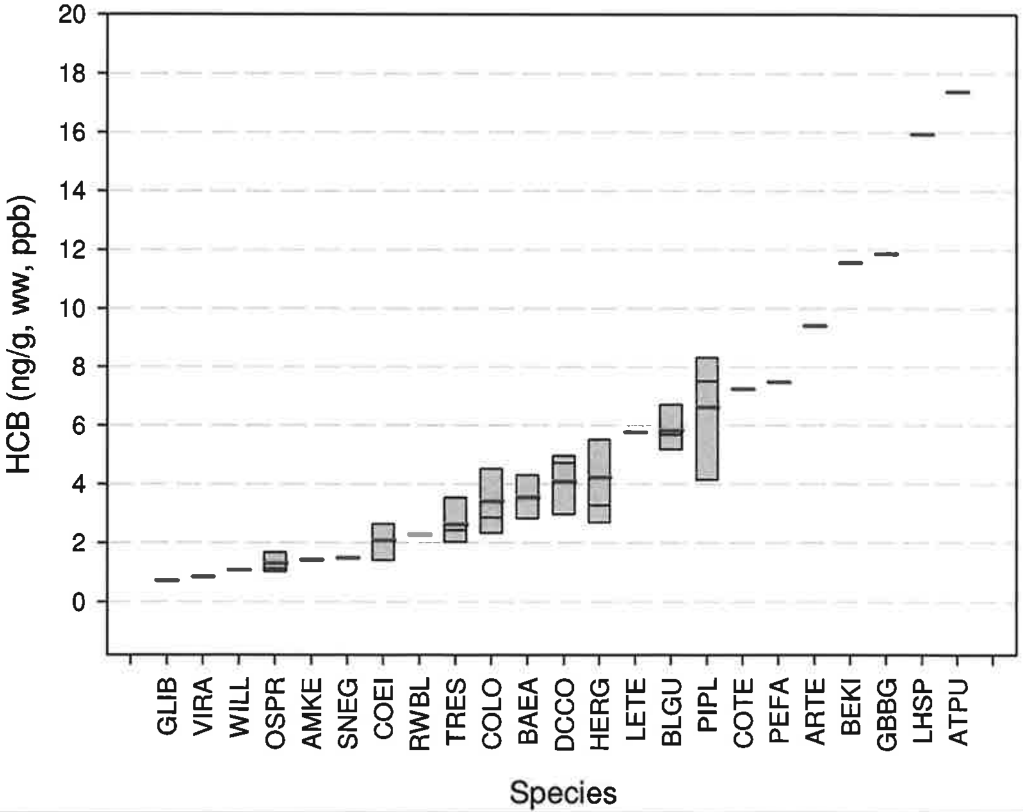


Figure 12. Total chlordane by species. Blue line is mean, black line is the median, and box boundary is data range. All sample fall below the 2,000 (ng/g) lethal levels measured in bird brains (Blus 2003).

Species Key (sample size): **American kestrel**, AMKE, (n = 1); **Arctic tern**, ARTE, (n = 1); **Atlantic puffin**, ATPU, (n = 1); **bald eagle**, BAEA, (n = 4); **belted kingfisher**, BEKI, (n = 2); **black guillemot**, BLGU, (n= 3); **common eider**, COEI, (n = 6); **common loon**, COLO, (n = 6); **common tern**, COTE, (n = 1); **double-crested cormorant**, DCCO, (n = 5); **great black-backed gull**, GBBG, (n = 2); **glossy ibis**, GLIB, (n = 1); **herring gull**, HERG, (n = 6); **least tern**, LETE, (n = 1); **Leach's storm-petrel**, LHSP, (n = 1); **osprey**, OSPR, (n = 6); **peregrine falcon**, PEFA, (n = 1); **piping plover**, PIPL, (n = 4); **red-winged blackbird**, RWBL, (n = 1); **snowy egret**, SNEG, (n = 1); **tree swallow**, TRES, (n = 4); **Virginia rail**, VIRA, (n = 1); and **willet**, WILL, (n = 1).

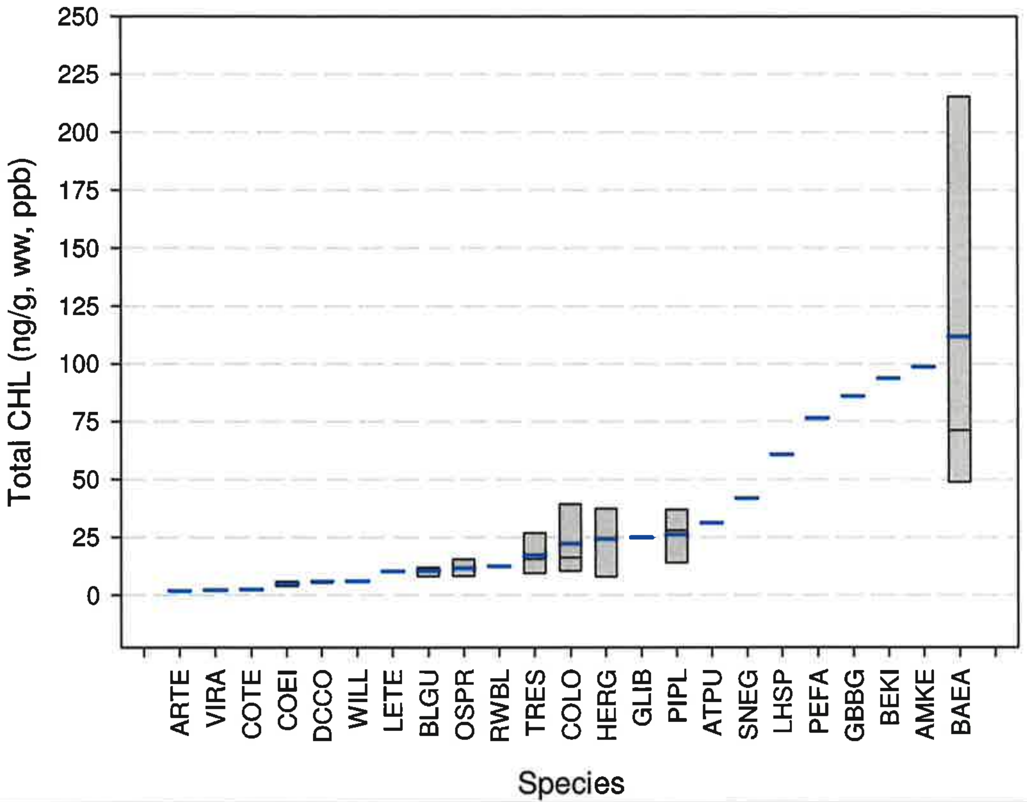
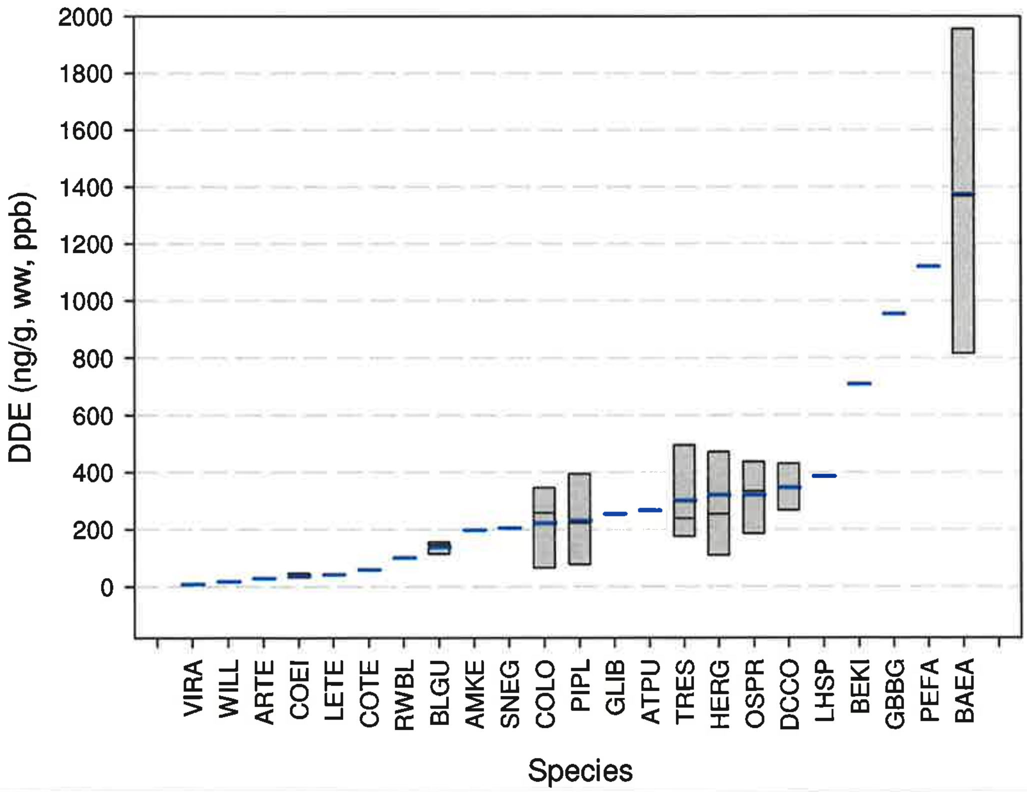


Figure 13. DDE levels by species. Blue line is mean, black line is the median, and box boundary is data range. All levels fall below the 15,000 (ng/g, ww) effects threshold for peregrine falcons (Blus 2003).

Species Key (sample size): **American kestrel**, AMKE, (n = 1); **Arctic tern**, ARTE, (n = 1); **Atlantic puffin**, ATPU, (n = 1); **bald eagle**, BAEA, (n = 4); **belted kingfisher**, BEKI, (n = 2); **black guillemot**, BLGU, (n= 3); **common eider**, COEI, (n = 6); **common loon**, COLO, (n = 6); **common tern**, COTE, (n = 1); **double-crested cormorant**, DCCO, (n = 5); **great black-backed gull**, GBBG, (n = 2); **glossy ibis**, GLIB, (n = 1); **herring gull**, HERG, (n = 6); **least tern**, LETE, (n = 1); **Leach's storm-petrel**, LHSP, (n = 1); **osprey**, OSPR, (n = 6); **peregrine falcon**, PEFA, (n = 1); **piping plover**, PIPL, (n = 4); **red-winged blackbird**, RWBL, (n = 1); **snowy egret**, SNEG, (n = 1); **tree swallow**, TRES, (n = 4); **Virginia rail**, VIRA, (n = 1); and **willet**, WILL, (n = 1).



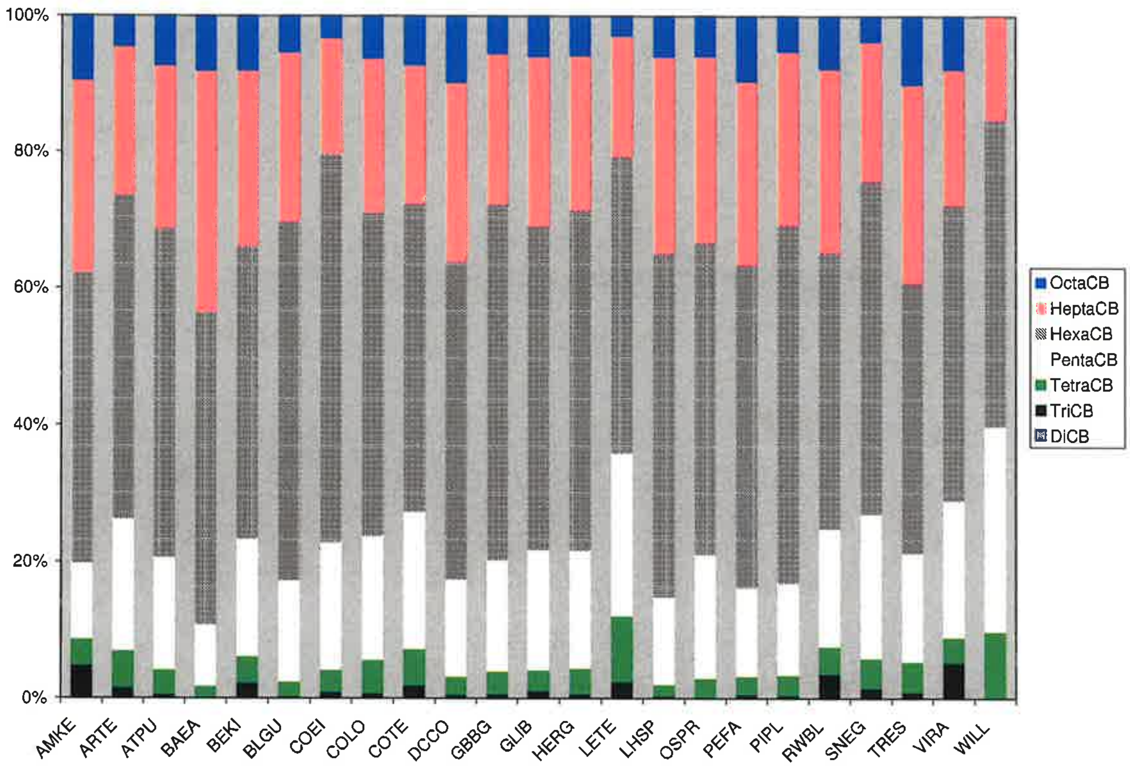


Figure 14. Percentage of PCB homologue by species.

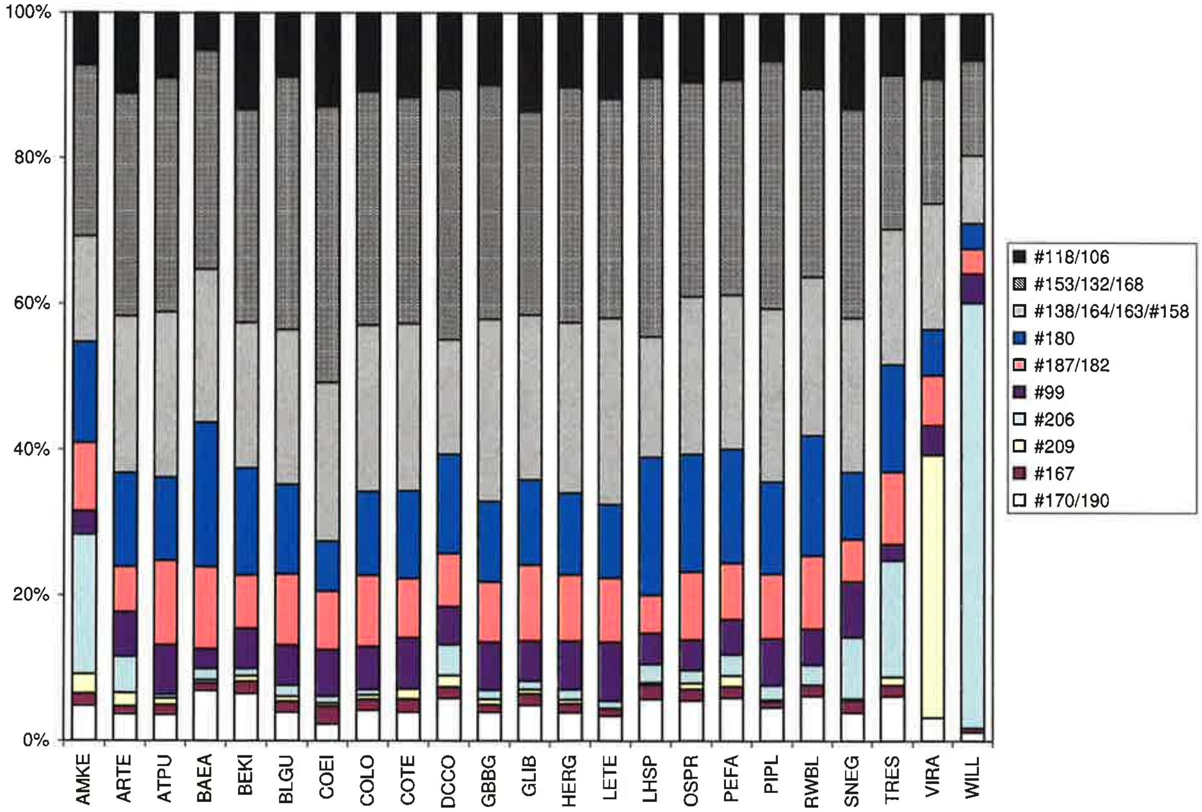


Figure 15. Percentage of PCB isomers (composing > 5% of total PCB) by species.

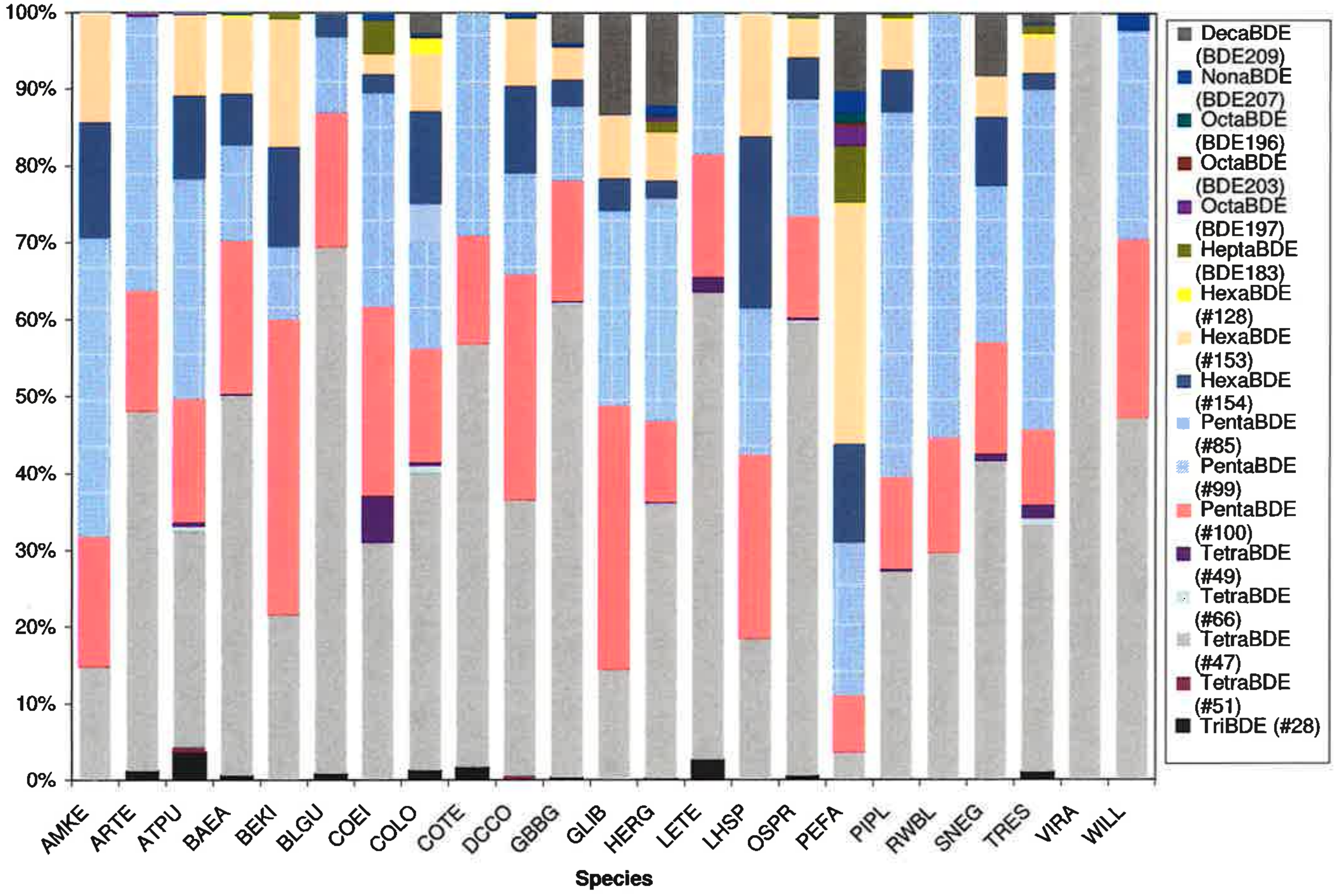


Figure 16. Percentage of PBDE congener by species.

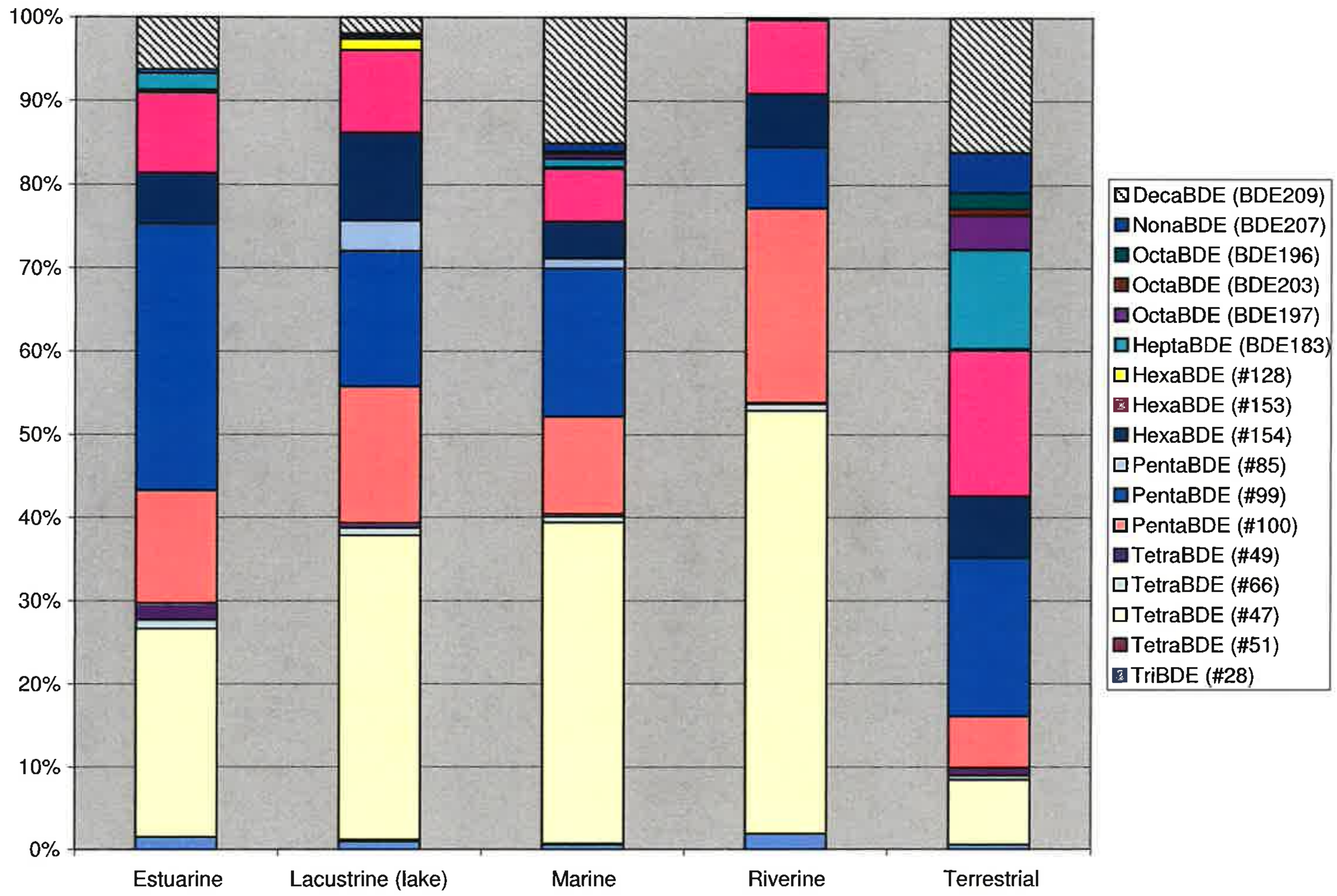


Figure 17. PBDE congener pattern across habitat type.

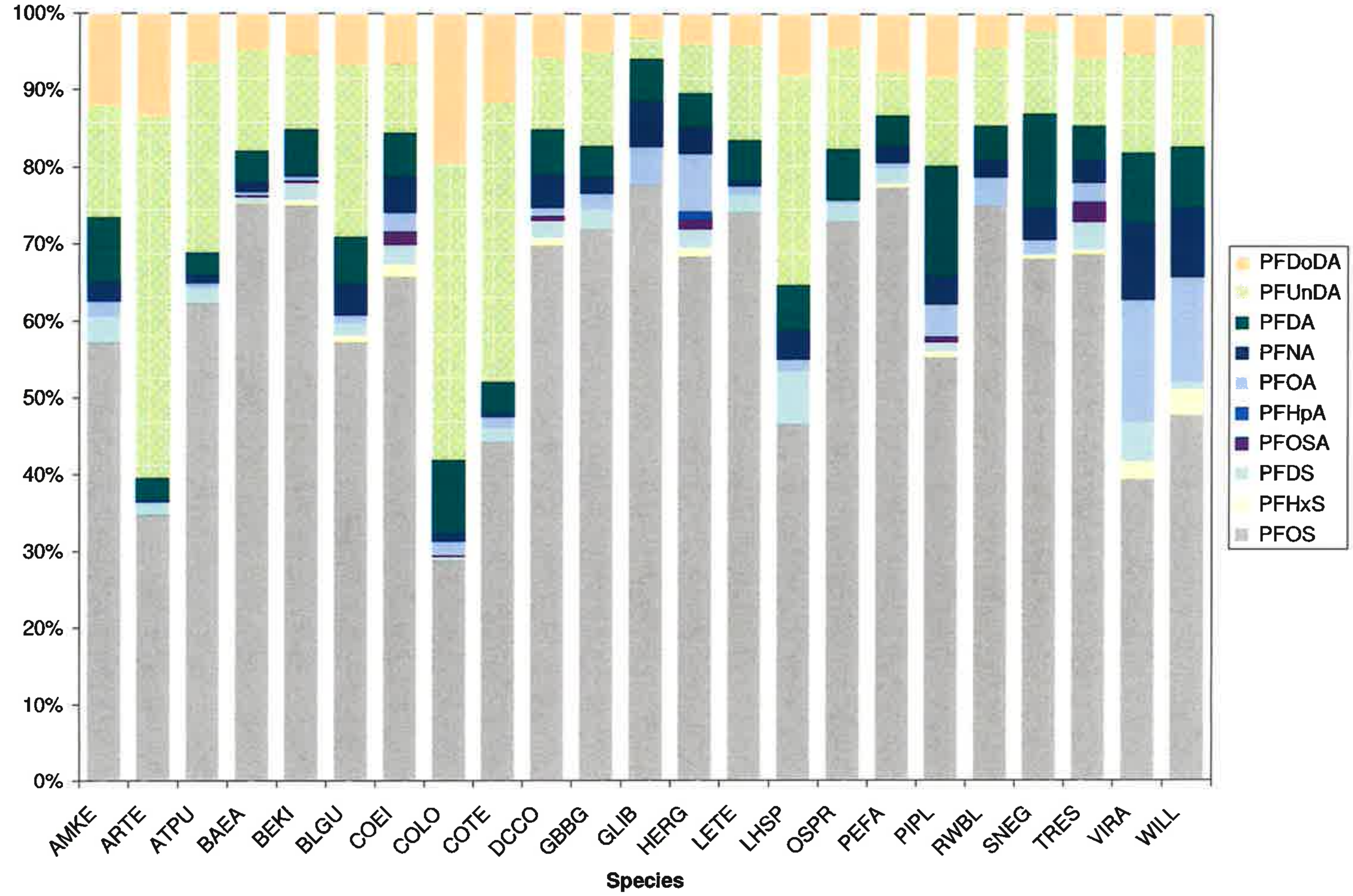


Figure 18. Percentage of PFC congener by species. Note PFBS and PFHxA are below detection limits in all species.

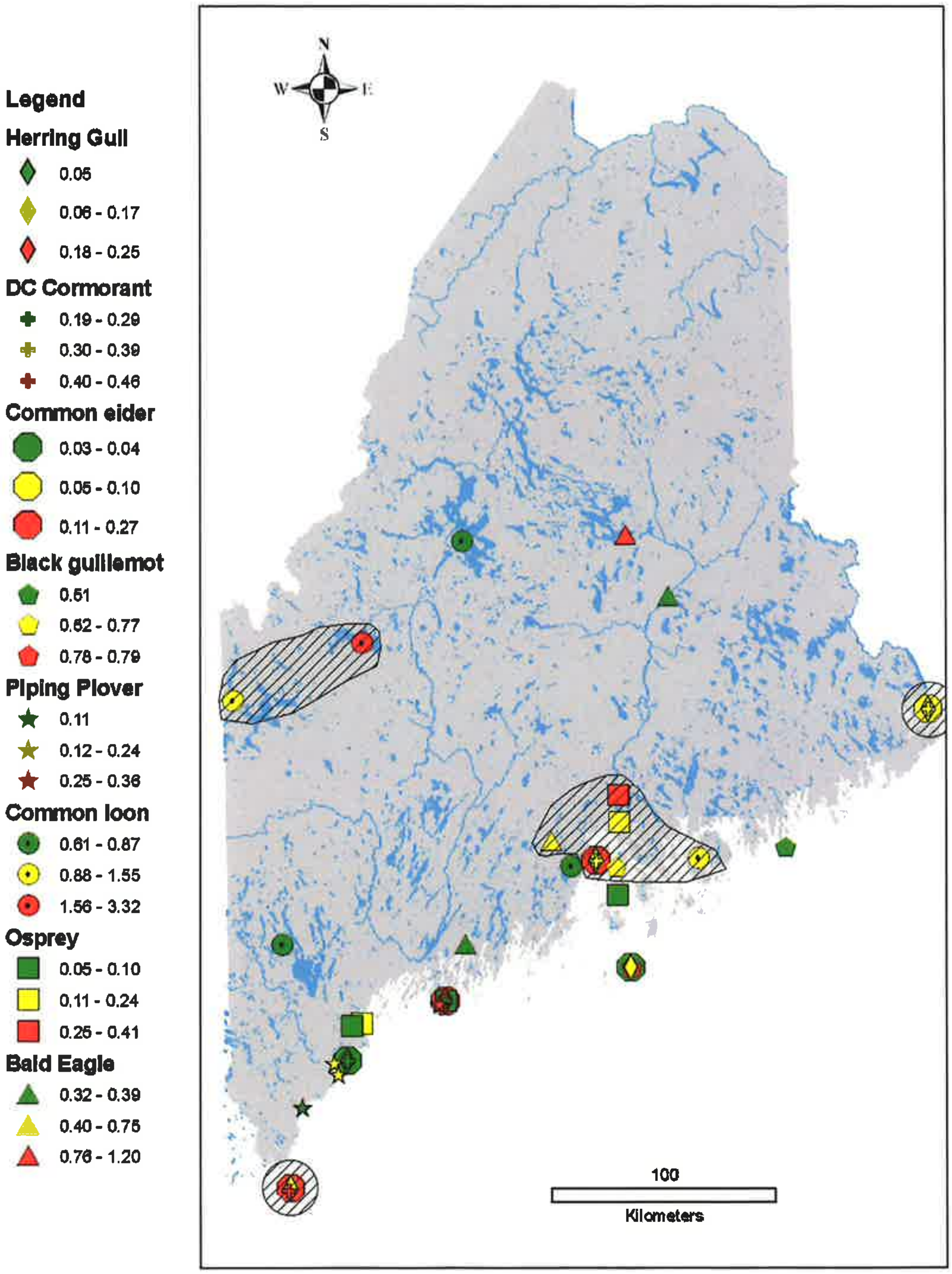


Figure 19. Hg geographic variation (µg/g, ww, ppm).

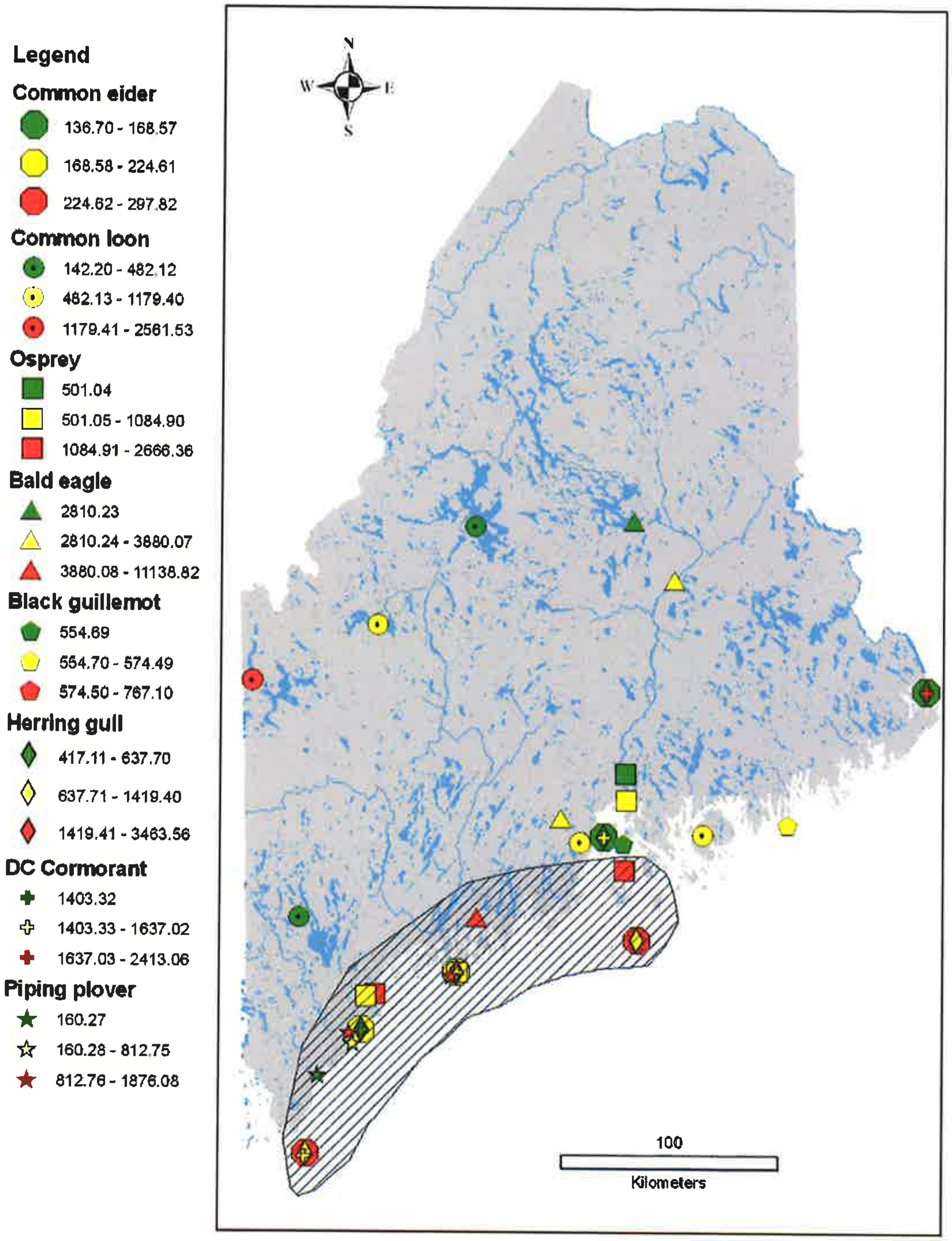


Figure 20. Total PCB geographic variation (ng/g, ww, ppb). Categories are relative to each species. Hatched area represents area generally high in most species (note: within this area there are herring gull, cormorant, and plover levels that do not follow the trend).

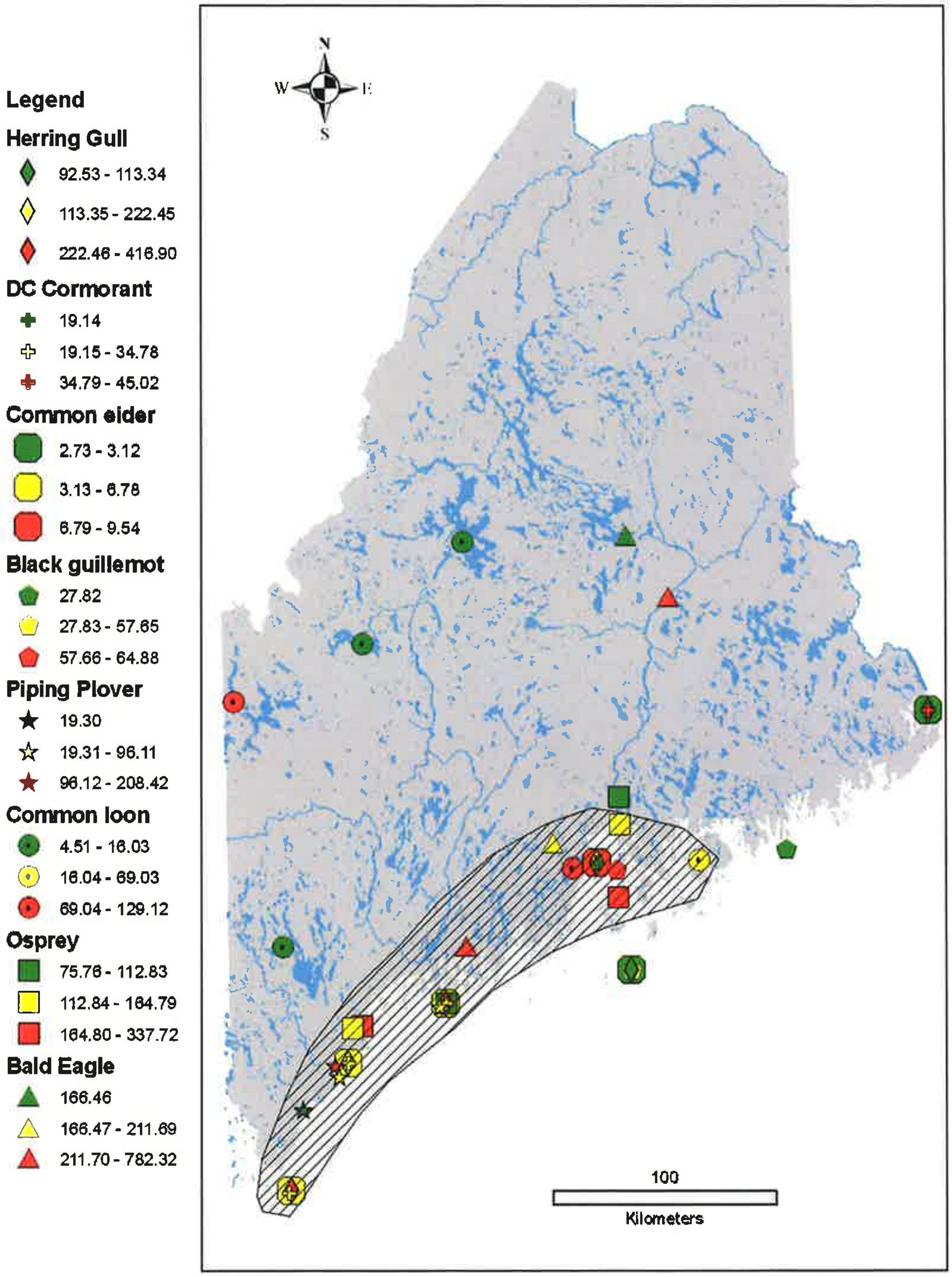


Figure 21. PBDE geographic variation (ng/g, ww, ppb). Categories are relative to each species. Hatched area represents area generally high in most species (note: within this area there are plover, cormorant, and osprey levels that do not follow the trend).

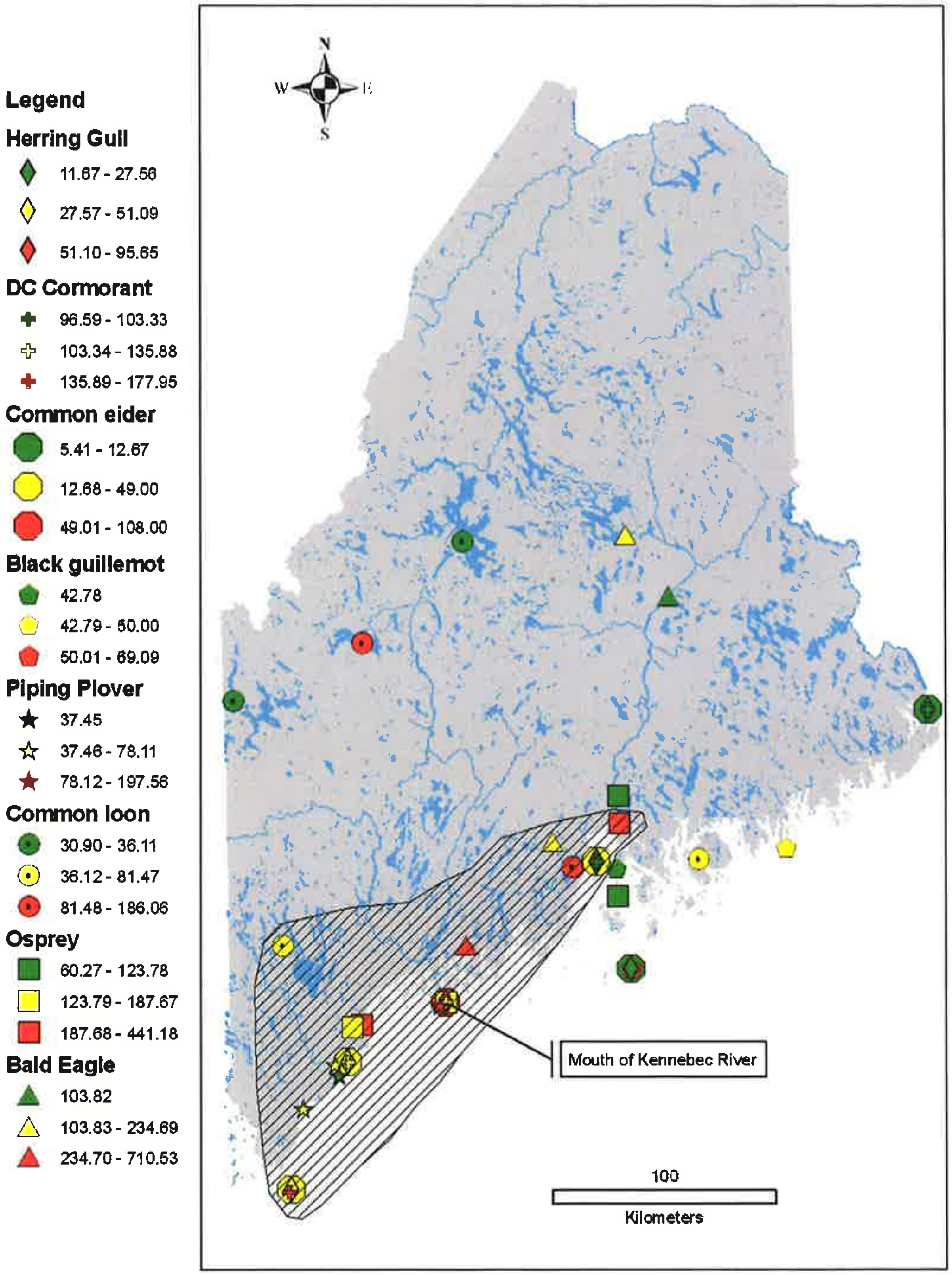


Figure 22. PFOS geographic variation (ng/g, ww, ppb). Categories are relative to each species. Hatched area represents area generally high in most species (note: within this area there are plover, cormorant, and herring gull levels that do not follow the trend).

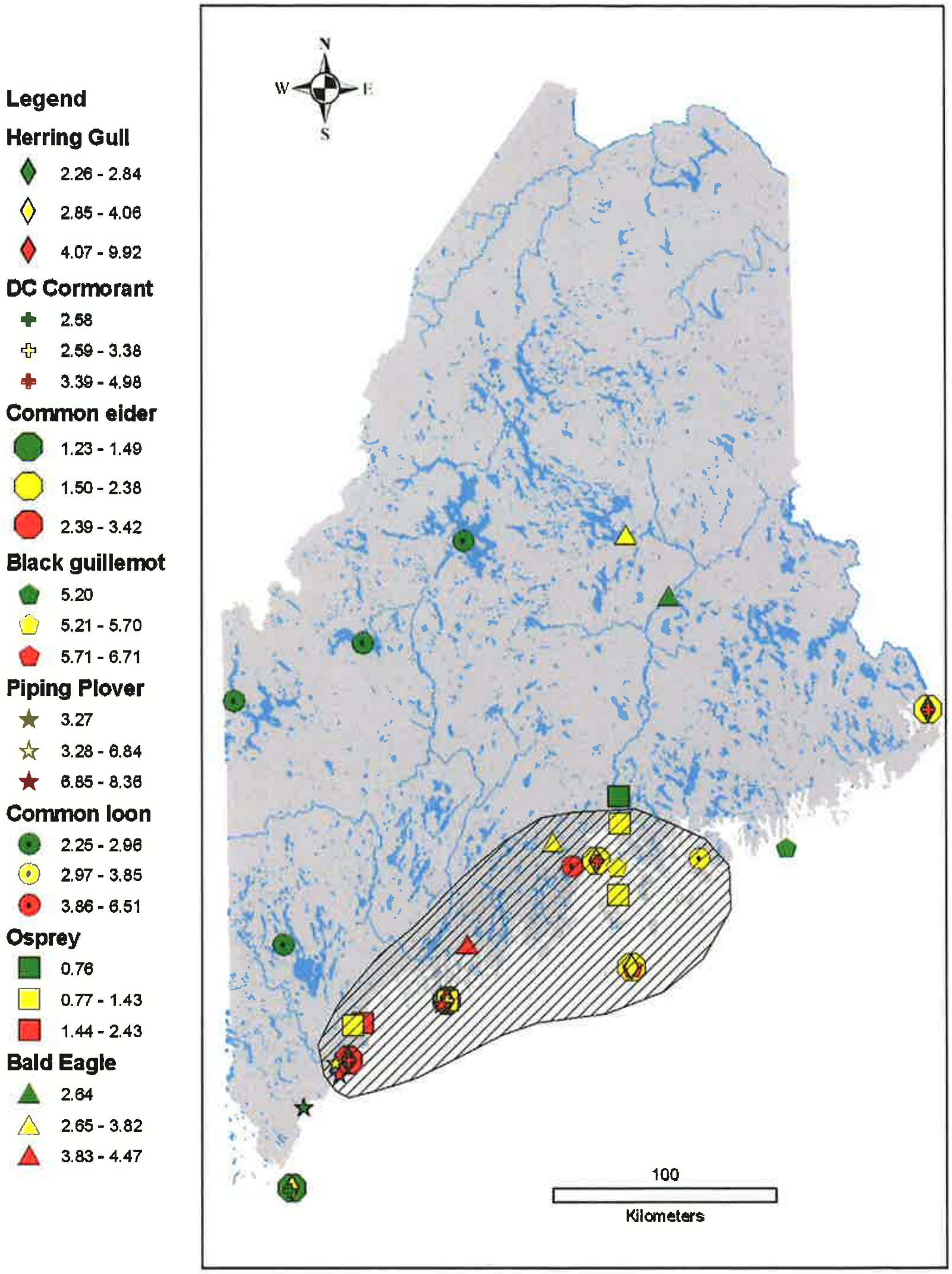


Figure 23. HCB geographic variation (ng/g, ww, ppb). Categories are relative to each species. Hatched area represents area generally high in most species (note: within this area there are eider and herring gull levels that do not follow the trend).

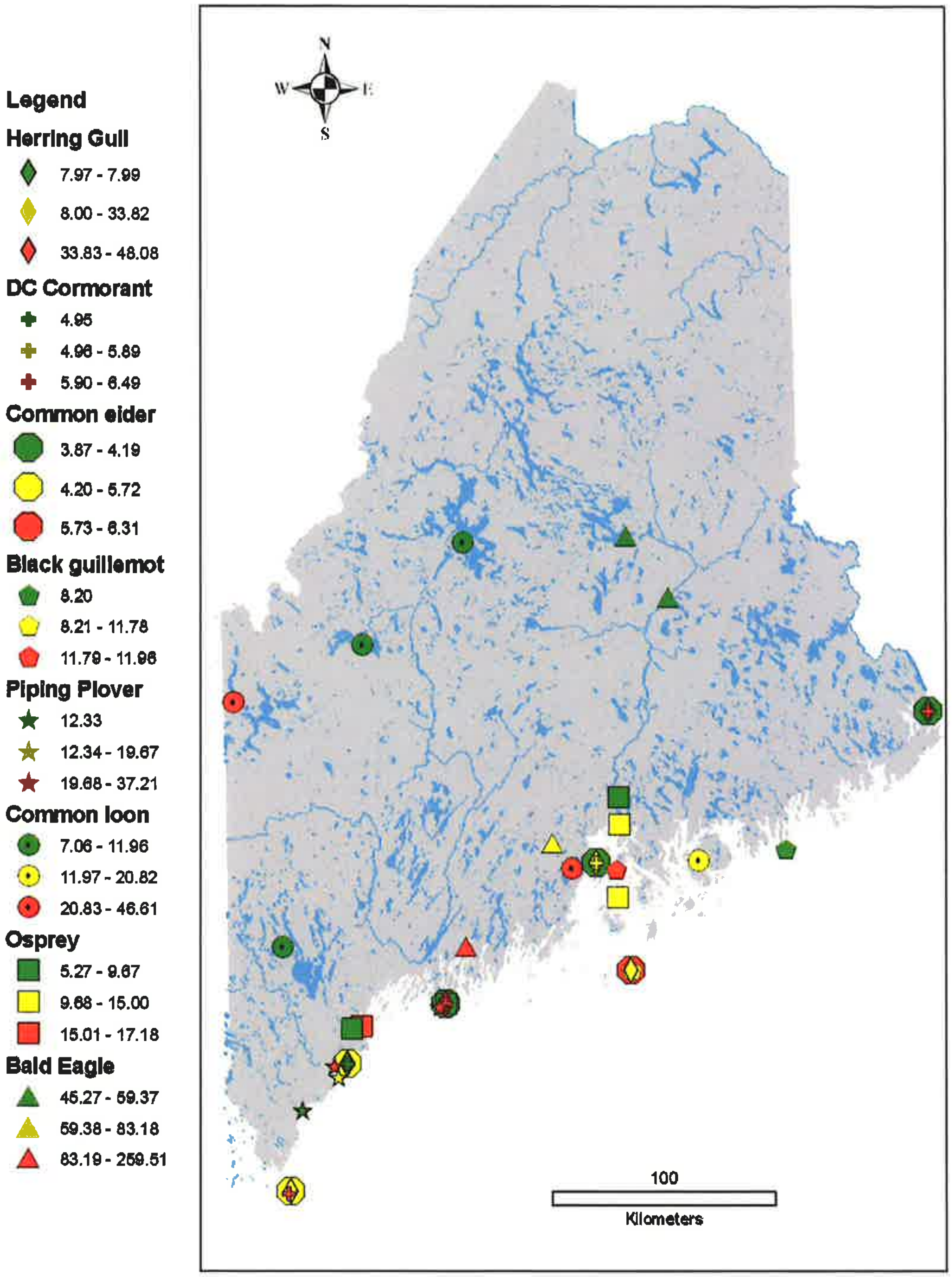


Figure 24. Chlordane geographic variation (ng/g, ww, ppb). Categories are relative to each species.

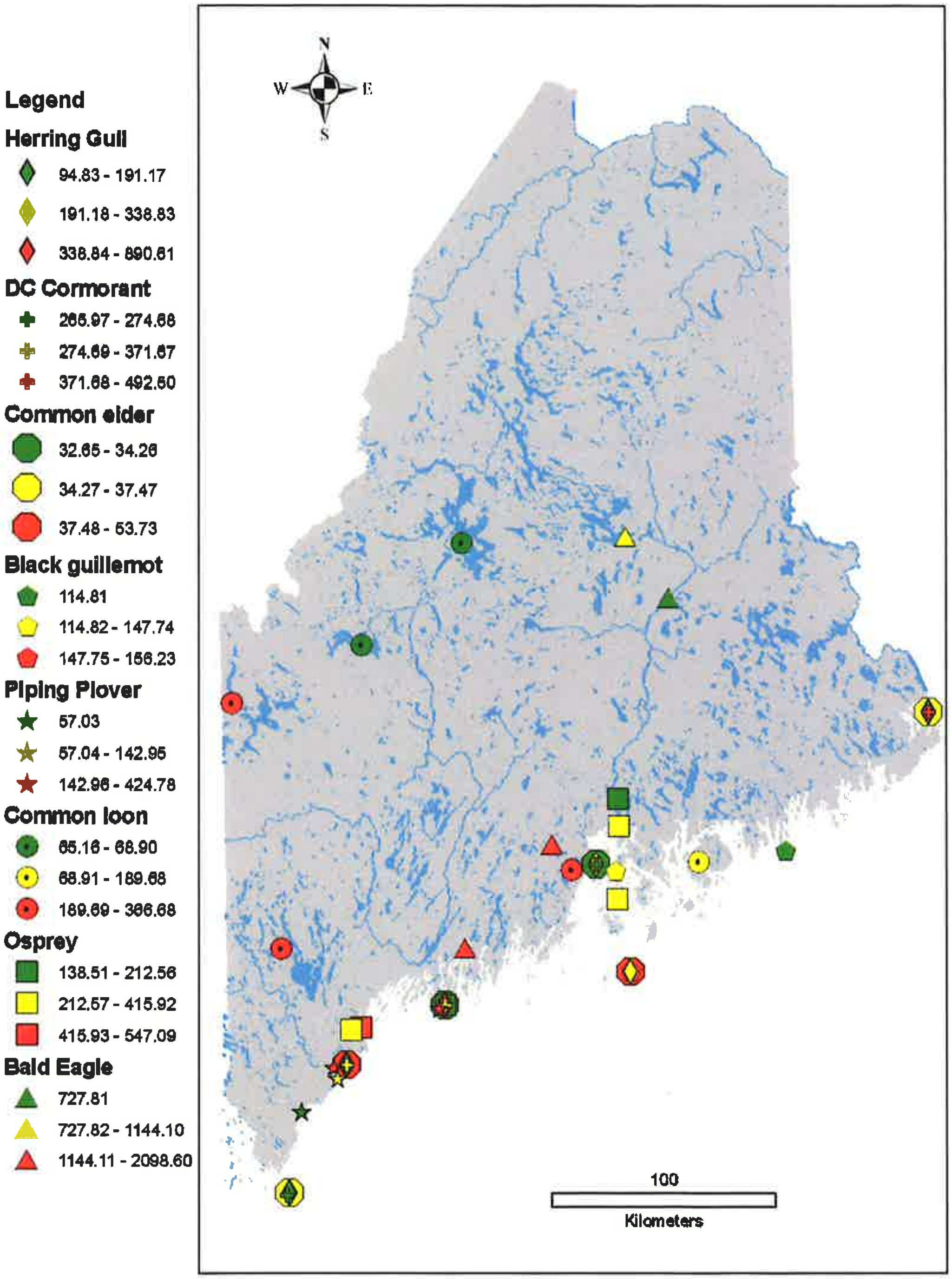


Figure 25. Total DDT geographic variation (ng/g, ww, ppb).

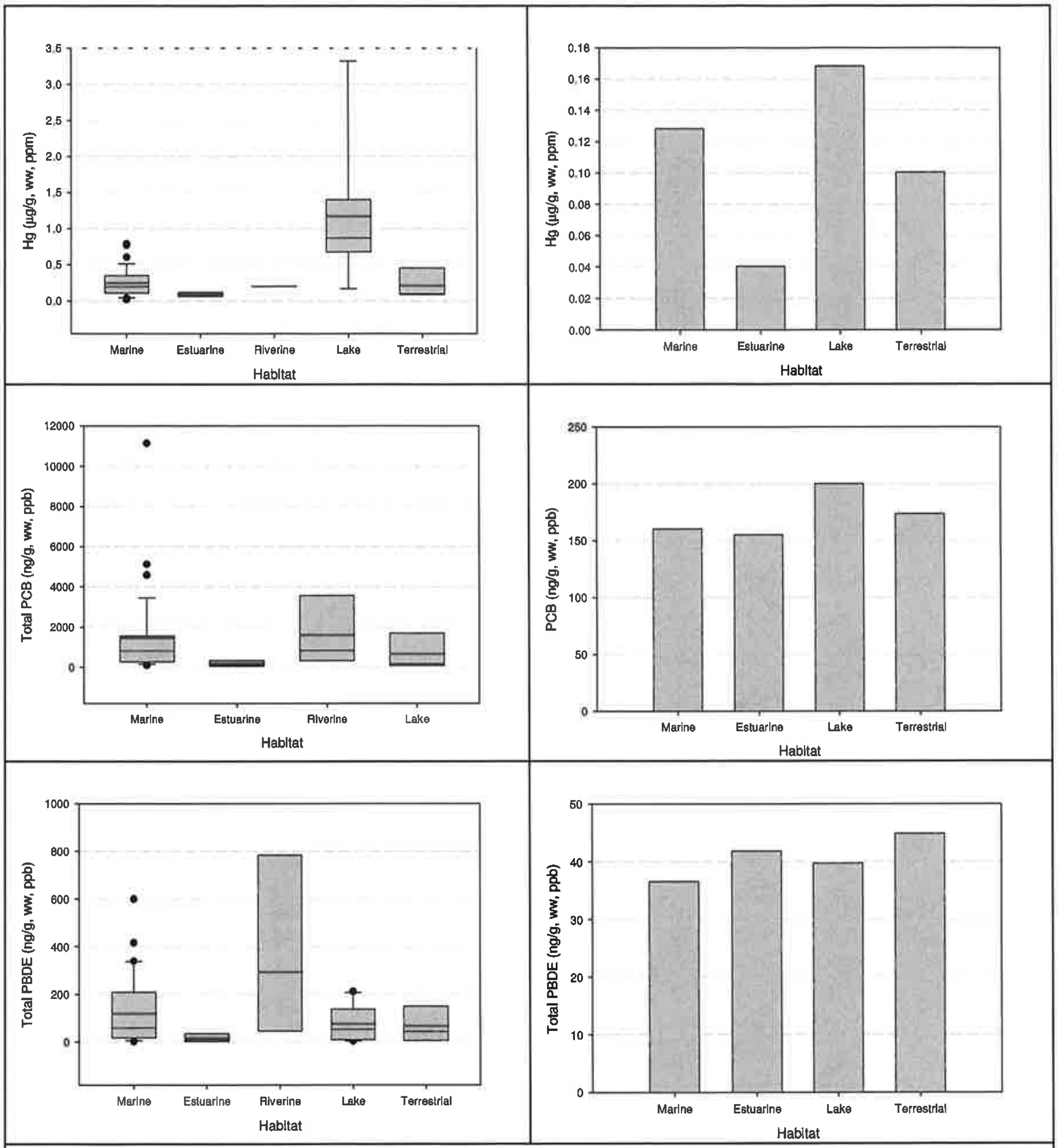


Figure 26. Contaminants by habitat type. Entire data set on left, tree swallows on right.

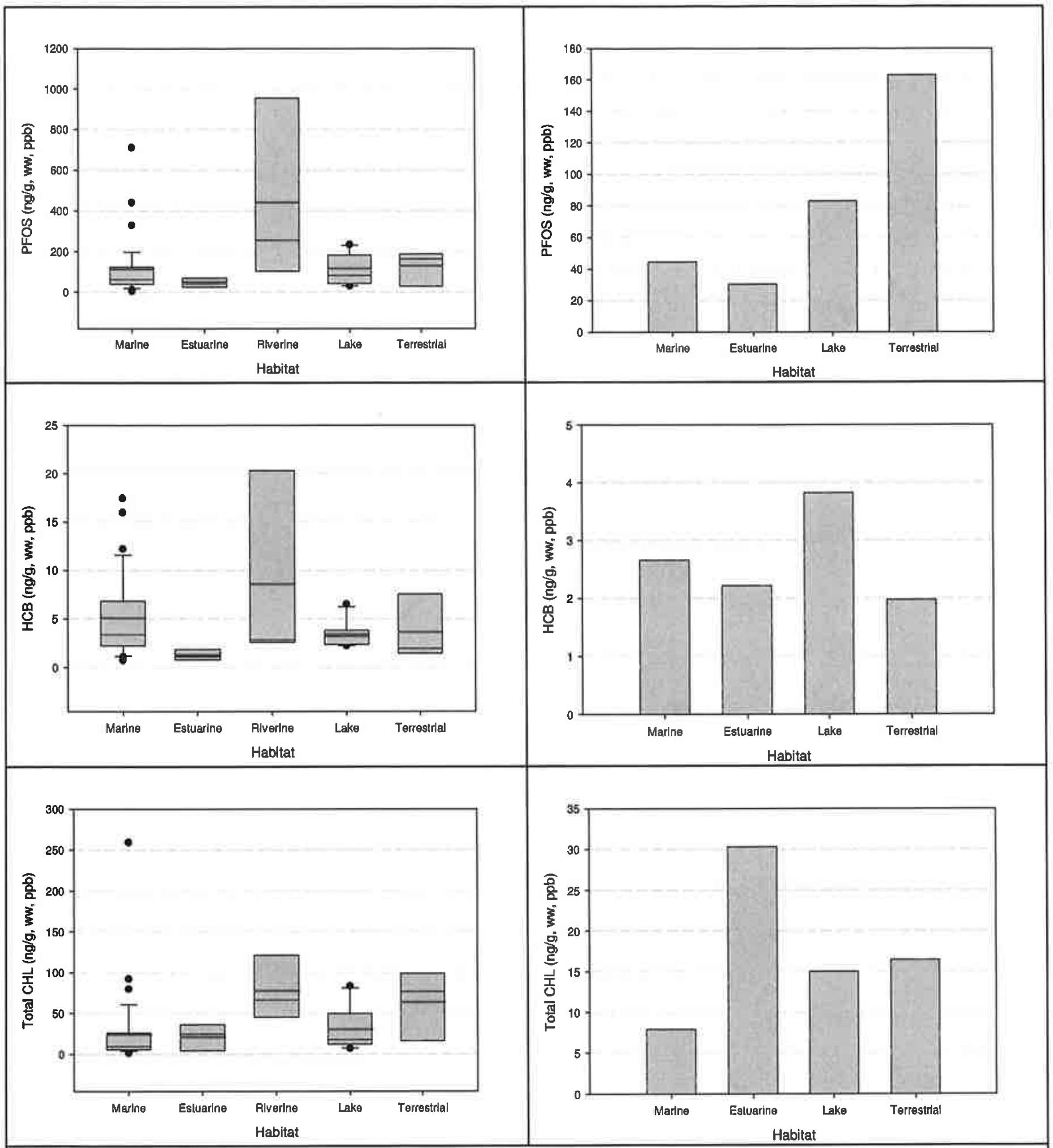


Figure 26 (con.). Contaminants by habitat type. Entire data set on left, tree swallows on right.

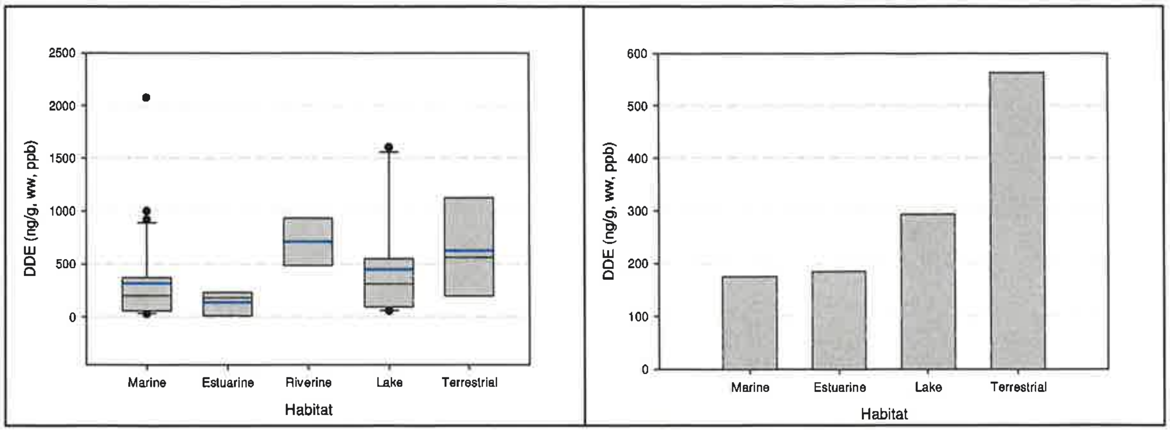


Figure 26 (con). Contaminants by habitat type. Entire data set on left, tree swallows on right.

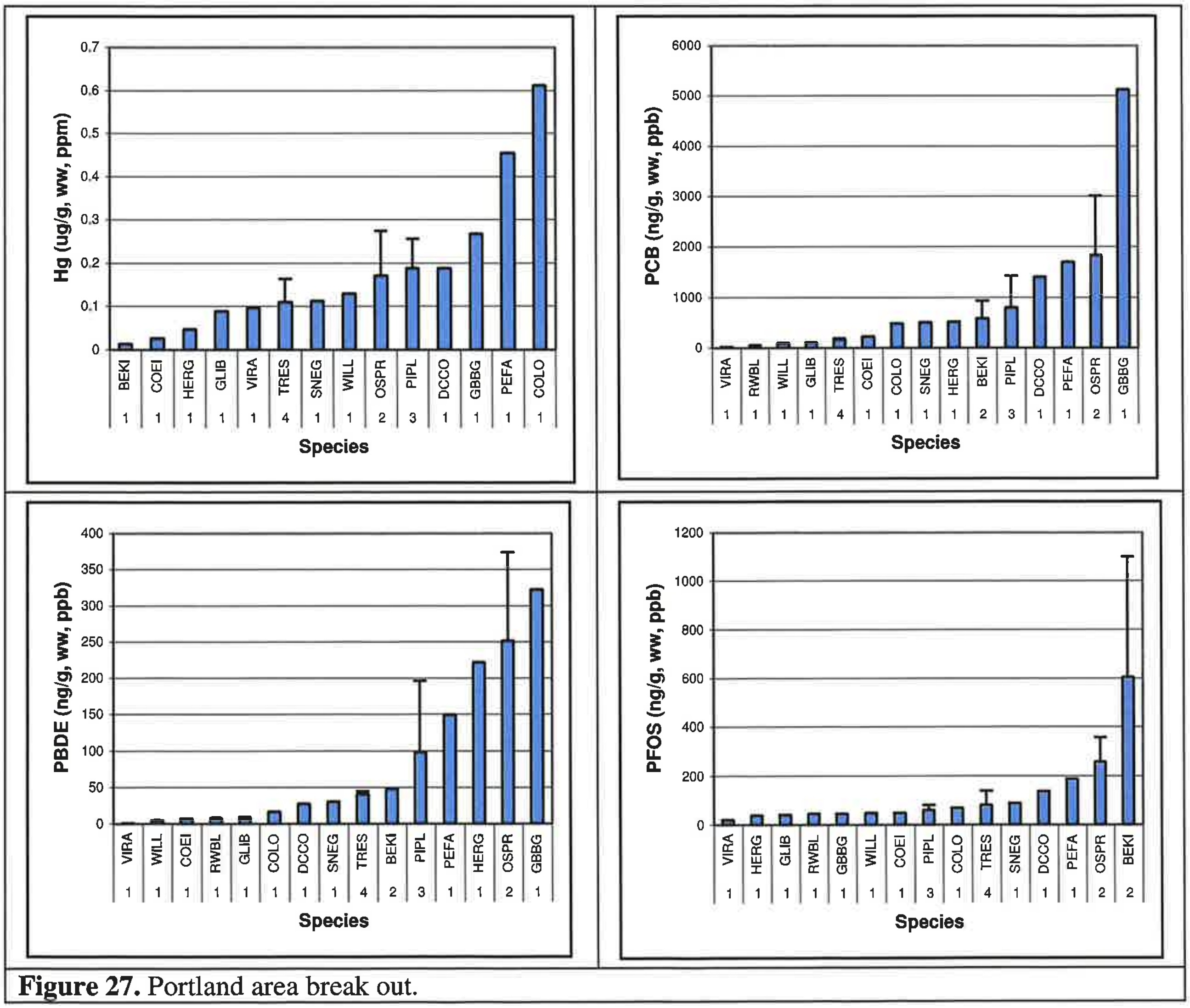


Figure 27. Portland area break out.

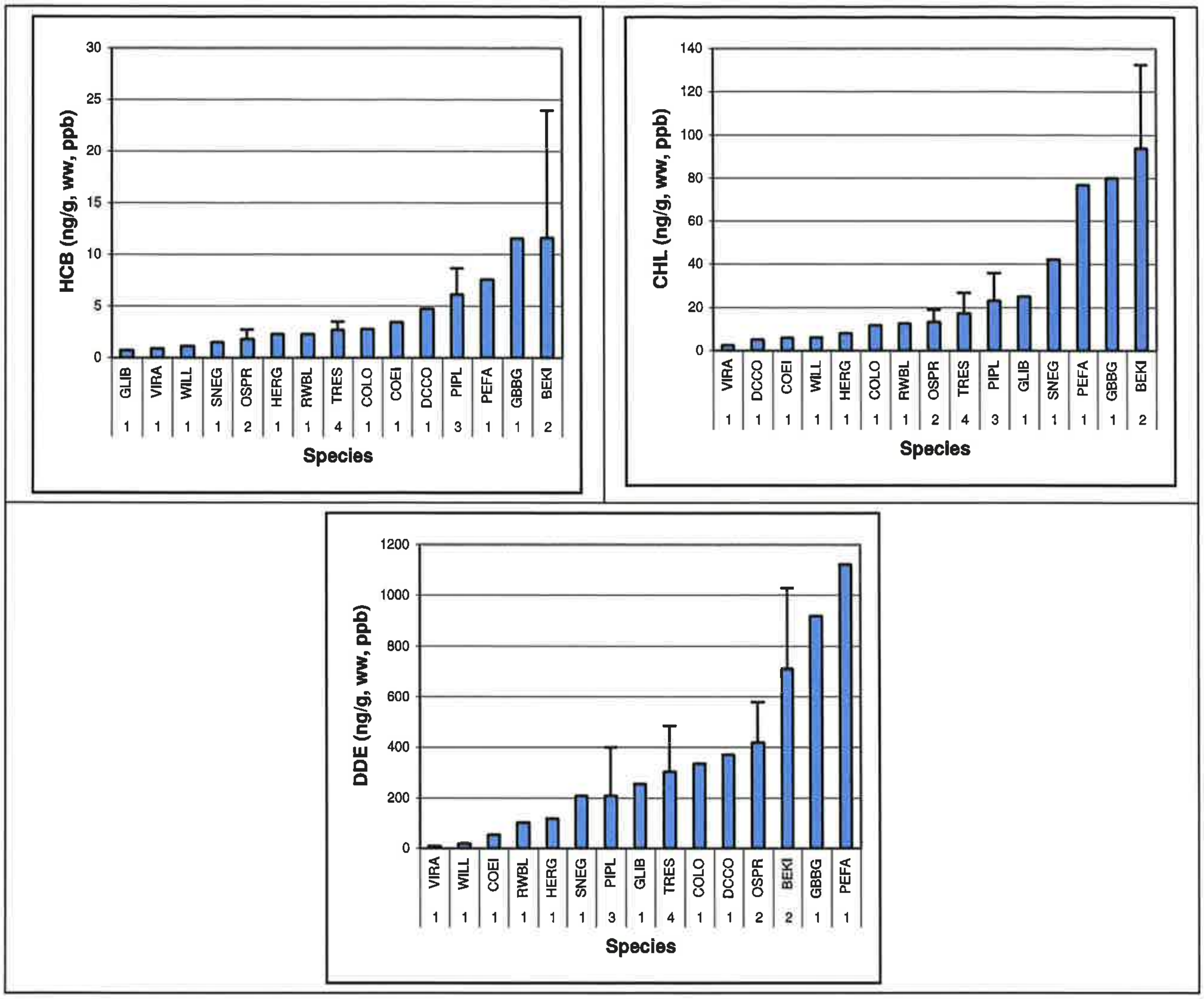


Figure 27 (con.). Portland area break out.

7. TABLES

Table 1. Migration, breeding phenology, and diet of study species.

Species	Time on breeding ground before egg laying	Diet	Citation
American kestrel	2-3 weeks	Terrestrial arthropods and small vertebrates	Smallwood and Bird 2002
Arctic tern	2-4 weeks	Small fish, crustaceans, insects	Hatch 2002
Atlantic puffin	2-3 weeks	Small to mid-sized schooling fish	Lowther et al. 2002
Bald eagle	2-3 months	Fish, birds, mammals	C. Desorbo per. Com.
Belted kingfisher	4-6 weeks	Small fish	Hamas 1994, Albano 2002
Black guillemot	Year round	Benthic and pelagic fish	Butler and Buckley 2002
Common eider	2-4 weeks	Benthic invertebrates	Goudie et al. 2002
Common loon	4 weeks	Small, medium sized fish	BRI unpublished data
Common tern	15-25 days	Small fish	Nisbet 2002
Double-crested cormorant	2-4 weeks	Fish 4-40cm	Hatch and Weseloh 1999
Great black-backed gull	Year round	Fish, invertebrates, birds, mammals	Good 1998
Glossy ibis	2-4 weeks	Invertebrates	Davis and Kricher 2002
Herring Gull	Year round	Invertebrates, fish, birds	Perotti and Good 1994
Least tern	2-3 weeks	Small fish, invertebrates	Thompson et al. 1997
Leach's storm-petrel	< 9 weeks	Myctophid fish, invertebrates	Huntington et al. 1996
Osprey	4 weeks	Live fish	Poole et al. 2002
Peregrine falcon	2 weeks to 2 months	Birds	White et al. 2002
Piping plover	2-4 weeks	invertebrates	Haig 2004
Red-winged blackbird	2-4 weeks	Insects, seeds	Yasukawa and Searcy 1995
Snowy egret	3-4 weeks	Invertebrates, fish, frogs, snakes	Parsons and Master 2000
Tree swallow	2-4 weeks	Flying insects	Robertson et al. 1992
Virginia rail	2-4 weeks	Aquatic invertebrates	Conway 1995
Willet	3 weeks	Insects, invertebrates, small fish	Lowther et al. 2001

Table 2 Samples Collected

Species	Latin	Location	Town	Lat	Long	Viable	Habitat	# Clutch	Total eggs
American kestrel	*Falco sparverius*	Gorham	Gorham	43.700470	-70.398850	No	Terrestrial	3	3
Arctic tern	*Sterna paradisaea*	Petit Manan	Millbridge	44.366980	-67.866060	Yes	Marine	2	2
Atlantic puffin	*Fratercula arctica*	Petit Manan	Millbridge	44.366980	-67.866060	No	Marine	1	1
Bald eagle	*Haliaeetus leucocephalus*	Penobscot River	Chester	45.384722	-68.518111	No	Riverine	1	1
		Tibbet island	Boothbay	43.978810	-69.662128	No	Marine	1	1
		Quakish Lake	T3 Indian Purchase	45.630750	-68.758278	No	Freshwater	1	1
		Quantabcook	Searsmont	44.395444	-69.180389	No	Freshwater	1	1
Belted kingfisher	*Ceryle alcyon*	Presumscott R.	Falmouth	43.715360	-70.317300	Yes	Riverine	1	2
			Westbrook	43.730470	-70.290800	Yes	Riverine	1	2
Black guillemot	*Pandion haliaetus*	Petit Manan	Millbridge	44.366980	-67.866060	Yes	Marine	3	3
		Seal Island NWR	Criehaven TWP	43.887590	-68.740823	Yes	Marine	3	3
		Western Island	Deer Isle	44.291217	-68.822417	Yes	Marine	3	3
Common eider	*Somateria mollissima*	Isles of Shoals	Kittery	42.988010	-70.610950	Yes	Marine	3	3
		Flat Island	Islesboro	44.317640	-68.932160	Yes	Marine	3	3
		Goose Island	Eastport	44.913610	-67.041310	Yes	Marine	3	3
		S. Sugarloaf Island	Phippsburg	43.748330	-69.771790	Yes	Marine	3	3
		Seal Island NWR	Criehaven TWP	43.887590	-68.740823	Yes	Marine	3	3
		Stratton Island	Old Orchard Beach	43.504310	-70.312640	Yes	Marine	3	3
Common loon	*Gavia immer*	Aziscohos Lake	Lincoln TWP	44.944051	-70.994675	No	Freshwater	1	1
		Coleman Pond	Lincolnville	44.295339	-69.073618	No	Freshwater	1	1
		Flagstaff Lake	Dead River TWP	45.187117	-70.267309	No	Freshwater	1	1
		Forest Ingalls Pond	Bridgton	43.966739	-70.686541	No	Freshwater	1	1
		Moosehead Lake	Spalding, TWR	45.604519	-69.701706	No	Freshwater	1	1
		Long Pond,	Mount Desert Island	44.325997	-68.361031	No	Freshwater	1	1
Common tern	*Sterna hirundo*	Petit Manan	Millbridge	44.366980	-67.866060	Yes	Marine	3	3
Double-crested cormorant	*Phalacrocorax auritus*	Flat Island	Islesboro	44.317640	-68.932160	Yes	Marine	3	3
		Goose Island	Eastport	44.913610	-67.041310	Yes	Marine	3	3
		Isles of Shoals	Kittery	42.975960	-70.625630	Yes	Marine	3	3
		S. Sugarloaf Island	Phippsburg	43.748330	-69.771790	Yes	Marine	3	3
		Stratton Island	Old Orchard Beach	43.504310	-70.312640	Yes	Marine	3	3
Great black-backed gull	*Larus marinus*	S. Sugarloaf Island	Phippsburg	43.748330	-69.771790	Yes	Marine	3	3
		Stratton Island	Old Orchard Beach	43.504310	-70.312640	Yes	Marine	3	3

Species	Latin	Location	Town	Lat	Long	Viable	Habitat	# Clutch	Total eggs
Glossy ibis	*Plegadis falcinellus*	Stratton Island	Old Orchard Beach	43.504310	-70.312640	No	Estuarine	3	3
Herring Gull	*Larus argentatus*	Isles of Shoals	Kittery	42.988010	-70.610950	Yes	Marine	3	3
		Flat Island	Islesboro	44.317640	-68.932160	Yes	Marine	3	3
		Goose Island	Eastport	44.913610	-67.041310	Yes	Marine	3	3
		S. Sugarloaf Island	Phippsburg	43.748330	-69.771790	Yes	Marine	3	3
		Seal Island NWR	Criehaven TWP	43.887590	-68.740823	Yes	Marine	3	3
		Stratton Island	Old Orchard Beach	43.504310	-70.312640	Yes	Marine	3	3
Least tern	*Sterna antillarum*	Crescent Surf	Kennebunk	43.387931	-70.429053	No	Marine	2	2
Leach's storm-petrel	*Oceanodroma leucorhoa*	Seal Island NWR	Seal Island NWR	43.887590	-68.740823	Yes	Marine	3	3
Osprey	*Pandion haliaetus*	Bug Light	South Portland	43.654510	-70.240360	No	Marine	1	1
		Fore River	Portland	43.643426	-70.289968	No	Marine	1	1
		Fort Point	Stockton Springs	44.469305	-68.803208	No	Marine	1	1
		Hog Island	North Haven	44.177492	-68.815976	Yes	Marine	1	1
		S. Sugarloaf Island	Phippsburg	43.748330	-69.771790	Yes	Marine	1	1
		Verso Mill	Bucksport	44.579019	-68.809802	No	Marine	1	1
Peregrine falcon	*Falco peregrinus*	Portland	Portland	43.642255	-70.284844	No	Terrestrial	1	1
Piping plover	*Charadrius melodus*	Ferry Beach	Saco	43.493666	-70.385325	No	Estuarine	1	3
		Hills Beach	Biddeford	43.451048	-70.363724	No	Estuarine	1	2
		Popham Beach	Phippsburg	43.735246	-69.807936	No	Estuarine	1	4
		Wells Beach	Wells	43.313573	-70.561277	No	Estuarine	1	2
Red-winged blackbird	*Agelaius phoeniceus*	Highland Lake	Falmouth	43.752700	-70.354640	Yes	Freshwater	1	2
Snowy egret	*Egretta thula*	Stratton Island	Scarborough	43.504310	-70.312640	No	Estuarine	2	2
Tree swallow	*Tachycineta bicolor*	Gilsland Farm	Falmouth	43.708157	-70.239675	Yes	Terrestrial	2	6
		Highland Lake	Falmouth	43.752700	-70.354640	Yes	Freshwater	2	9
		Scarborough marsh	Scarborough	43.565970	-70.354060	Yes	Estuarine	6	14
		Stratton Island	Old Orchard Beach	43.504310	-70.312640	Yes	Marine	3	12
Virginia rail	*Rallus limicola*	Scarborough marsh	Scarborough	43.565970	-70.354060	No	Estuarine	1	5
Willet	*Catoptrophorus semipalmatus*	Scarborough marsh	Scarborough	43.565970	-70.354060	Yes	Estuarine	1	1

Table 3. Recoveries of PFCs and labeled internal standards spiked into egg matrixes

	Matrix Spike Recovery (%) n=6	
Compound	Mean	STDEV
PFOS	70	35
PFDS	81	12
PFOSA	60	25
PFHpA	80	13
PFOA	103	17
PFNA	102	15
PFDA	131	15
PFUnDA	124	22
PFDoDA	130	24
	Internal Standard Recovery (%) n=60	
	Mean	STDEV
13C4PFOS	107	23
13C4PFOA	122	27
13C4PFNA	101	24
13C4PFDA	134	34

Table 4. Species code, sample size, % lipid, and % moisture of egg composites. Sample size applies to all figures that follow.

Species	Species Code	N	Mean Lipid (%)	S.D. Lipid content (%)	Mean Moisture (%)	S.D. Moisture content (%)
American kestrel	AMKE	1	8.28		78.05	
Arctic tern	ARTE	1	9.76		74.02	
Atlantic puffin	ATPU	1	10.39		71.57	
Bald eagle	BAEA	4	5.09	2.06	77.77	3.19
Belted kingfisher	BEKI	2	6.74	0.13	77.97	0.04
Black guillemot	BLGU	3	11.43	0.85	69.61	3.68
Common eider	COEI	6	19.72	1.38	61.92	0.80
Common loon	COLO	6	8.43	2.10	72.54	3.27
Common tern	COTE	1	11.16		70.24	
Double-crested cormorant	DCCO	5	5.24	0.69	81.10	1.39
Great black-backed gull	GBBG	2	8.33	1.41	74.46	1.77
Glossy ibis	GLIB	1	6.41		78.20	
Herring Gull	HERG	6	10.02	1.83	72.62	2.96
Least tern	LETE	1	13.60		68.00	
Leach's storm-petrel	LHSP	1	11.77		69.79	
Osprey	OSPR	6	4.42	1.59	79.62	3.53
Peregrine falcon	PEFA	1	5.84		78.37	
Piping plover	PIPL	4	15.09	2.11	68.56	2.06
Red-winged blackbird	RWBL	1	4.59		84.00	
Snowy egret	SNEG	1	7.59		77.18	
Tree swallow	TRES	4	6.59	1.33	77.59	4.11
Virginia rail	VIRA	1	9.09		75.36	
Willet	WILL	1	13.10		64.02	

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