INJURIES TO AVIAN RESOURCES, LOWER FOX RIVER/GREEN BAY NATURAL RESOURCE DAMAGE ASSESSMENT

Final Report

Prepared for:

U.S. Fish and Wildlife Service
U.S. Department of the Interior
U.S. Department of Justice

Prepared by:

Stratus Consulting Inc.

Contacts:

Hector Galbraith
Mace Barron
Doug Beltman
Josh Lipton

May 7, 1999

CONTENTS

Appendices

FIGURES

TABLES

ACRONYMS

AHH	aryl hydrocarbon hydroxylase
ALAS	aminolevulinic acid synthase
CDF	Confined Disposal Facility
DOI	Department of Interior
dioxin; TCDD	2,3,7,8-tetrachlorodibenzo-p-dioxin
EROD	ethoxyresorufin-O-deethylase
LD50	lethal dose resulting in 50% mortality of the exposed population
LOEC	lowest observed effect concentration
mRNA	messenger RNA
NOAA	National Oceanic and Atmospheric Administration
NOEC	no observed effect concentration
NRDA	natural resource damage assessment
PB	phenobarbital
PCBs	polychlorinated biphenyls
PCDD	polychlorinated dibenzodioxins
PCDF	polychlorinated dibenzo-furan
PWRC	Patuxent Wildlife Research Center
TCDD-eq	TCDD equivalent concentration
TEF	toxicity equivalency factor
U.S. EPA	United States Environmental Protection Agency
USFWS	United States Fish and Wildlife Service
USGS	United States Geological Survey
WHO	World Health Organization

CHAPTER 1
INTRODUCTION

This document presents an evaluation of injuries to avian resources (birds) resulting from releases of polychlorinated biphenyls (PCBs) from paper company facilities along the Lower Fox River, Wisconsin. This evaluation has been performed as part of the natural resource damage assessment (NRDA) being performed for the Lower Fox River/Green Bay site by the U.S. Fish and Wildlife Service (USFWS, or the Service) on behalf of the U.S. Department of the Interior (the Department), the National Oceanic and Atmospheric Administration (NOAA), the Oneida Indian Tribe of Wisconsin, and the Menominee Indian Tribe. This report was prepared by Stratus Consulting Inc. under contract to the Service. The purpose of this injury evaluation is to assess:

- whether birds that use the Lower Fox River, Green Bay, and parts of Lake Michigan (the assessment area) have been exposed to PCBs
- whether birds in the assessment area have been injured as a result of exposure to PCBs.

In this chapter we introduce and define relevant terms from the Department NRDA regulations at 43 CFR Part 11, provide background information on PCB contamination of the area, and describe the overall organization of this report.

1.1 TERMS AND DEFINITIONS

The Department has promulgated regulations for the performance of NRDAs [43 CFR Part 11]. The term "injury" is defined in the Departmental regulations as "a measurable adverse change, either long or short term, in the chemical or physical quality or the viability of a natural resource resulting either directly or indirectly from exposure to a release of a hazardous substance" [43 CFR § 11.14]. The Departmental regulations also identify specific adverse changes that are defined as injuries. The relevant definitions of injury to avian resources assessed by the Trustees in this report are:

- concentrations of PCBs "sufficient to cause the biological resource or its offspring to have undergone at least one of the following adverse changes in viability: death, disease, behavioral abnormalities, cancer, genetic mutations, physiological malfunctions (including malfunctions in reproduction), or physical deformations" [43 CFR § 11.62 (f)(1)(i)]

- concentrations of PCBs sufficient to "exceed action or tolerance levels established under section 402 of the Food, Drug and Cosmetic Act, 21 U.S.C. 342, in edible portions of organisms" [43 CFR § 11.62 (f)(1)(ii)]

- concentrations of PCBs sufficient to "exceed levels for which an appropriate State health agency has issued directives to limit or ban consumption of such organism" [43 CFR § 11.62 (f)(1)(iii)].

NRDA injury determination assessment comprises two phases:

1. ***Pathway determination.*** In the pathway determination phase, pathways by which natural resources come into contact with hazardous substances are identified [43 CFR § 11.63]. The pathway is the "route or medium through which . . . a hazardous substance is or was transported from the source of the discharge or release to the injured resource" [43 CFR § 11.14 (dd)]. Thus, pathway analysis is an important component of demonstrating the linkage between the release of a hazardous substance and the injured natural resource. Other NRDA reports will specifically evaluate the pathways by which PCBs released from paper company facilities have come to be located in the Fox River, Green Bay, and parts of Lake Michigan. This report, however, presents data that describe those pathways by which birds have been exposed to PCBs. For all of the birds addressed in this report, the primary pathway is through dietary exposure.

2. ***Injury determination.*** In this phase, the trustees determine whether adverse effects that meet the definitions of injury set forth at 43 CFR § 11.62 have occurred as a result of exposure to hazardous substances.

1.2 BACKGROUND

PCBs were released into the Fox River/Green Bay system from Fox River paper company facilities that produced or processed PCB-containing carbonless copy paper waste (Wisconsin DNR, 1998). Estimates of the amount of PCBs discharged into the Fox River from paper company facilities range from 420,000 to 825,000 pounds from 1954 to the present (Wisconsin DNR, 1998). An extensive study of PCB fate and transport in the Fox River/Green Bay system demonstrated that PCBs move from the river into the bay, where they enter the food chain (DePinto et al., 1994). A mass balance study estimated that over 90% of the PCBs entering Green Bay in 1989 were from the Fox River (DePinto et al., 1994).

1.3 INJURY EVALUATION METHODS

Injuries to avian resources are determined in this report primarily through the use and interpretation of historical studies on birds in the assessment area. PCB contamination in Green Bay birds was first detected in the early 1970s (Bishop et al., 1992). Since then, multiple studies have been conducted on the exposure to and accumulation of PCBs in Green Bay birds and on adverse effects resulting from this exposure. Most of these studies have been published in the peer-reviewed literature; the evaluation presented in this report is based primarily on peer-reviewed scientific papers.

For this injury determination, the previously available information was supplemented by the collection and chemical analysis of a limited number of tern eggs (12) from the Green Bay assessment area in 1996. This data collection effort, which was outlined in the NRDA Assessment Plan published by the Service (61 Fed. Reg. 43,558), is described in detail in Appendix B.

When available, we relied on and report the statistical analyses conducted by the study authors. In cases where the study authors did not conduct their own statistical analyses, we conducted the analyses using raw data either reported in the study paper or obtained directly from the study authors. Cases where we conducted our own statistical analyses are clearly identified as such, and the statistical methods used to conduct the analyses are also identified. We used a statistical significance level of $\propto = 0.05$.

Finally, the available information was used in a weight-of-evidence approach to determine injuries to avian resources. The methods of the weight-of-evidence approach are described in Chapter 7.

As described in more detail in Chapter 8, the methods described here that were used to determine injuries to avian resources are consistent with those contained in the Departmental regulations for NRDA [43 CFR §11.64].

1.4 DOCUMENT ORGANIZATION

This document is organized as follows. Chapter 2 provides an overview of the avian resources of the Lower Fox River and Green Bay. Chapter 3 contains a review of the known toxicological effects of PCBs on birds, develops a “taxonomy” of PCB-induced injuries to birds, and develops avian toxicity thresholds. Chapter 4 summarizes information regarding exposure to PCB contamination in birds in the assessment area and evaluates the likelihood of PCB-induced injuries to these birds by comparing the exposure concentrations to the toxicity thresholds developed in Chapter 3. In Chapter 5, available site-specific biological data are reviewed by individual species to evaluate whether field data indicate that birds in the assessment area have been injured by PCBs according to injury definitions related to adverse effects on viability. Because of the availability of scientific information and studies, the bird species discussed in Chapter 5 are the double-crested

cormorant, Forster's tern, common tern, Caspian tern, tree swallow, red-breasted merganser, black-crowned night heron, and bald eagle. Chapter 6 determines injuries to ducks according to the injury definitions of exceedences of state or federal tolerance limits for PCBs in tissue, and exceedences of state or federal threshold concentrations for establishing consumption advisories. Chapter 7 then presents a weight-of-evidence evaluation of the role that PCBs have played in causing injuries to birds in the assessment area. Chapter 8 presents a determination of injuries pursuant to the Departmental NRDA regulations. Chapter 9 lists references cited.

Appendix A provides scientific names for all bird species mentioned in the text, and Appendix B provides documentation for 1996 field collection and chemical analysis of common and Forster's tern eggs. Appendix C provides the methods and results of waterfowl collection by the Service in 1987.

CHAPTER 2
ASSESSMENT AREA AVIAN RESOURCES

2.1 THE ASSESSMENT AREA

As described in the NRDA Assessment Plan published by the Service (61 Fed. Reg. 43,558), the assessment area for this NRDA includes the Lower Fox River, all of Green Bay, and parts of Lake Michigan. The assessment area is located on the northwest side of Lake Michigan (Figure 2-1) and lies within the Great Lakes ecoregion, an area that comprises a mixture of aquatic, agricultural, wetland, forested (deciduous and coniferous), and urban habitats. The main historical and current types of land use in the assessment area are agricultural, recreational, logging, and industrial/residential (largely confined to areas along the Fox River). Ecological habitats in the assessment area are primarily nonurban; while industrialization and residential development have taken place along parts of the Lower Fox River, much of the area is still dominated by low intensity agriculture, wetlands, and forests. This land use pattern has important implications for the use of the assessment area by birds, as described below.

The climate of the assessment area is highly seasonal and continental, with an average July air temperature of about 67°F and an average January air temperature of 20°F (Robbins, 1991). The average depth of soil frost in late February is about 20 inches. Annual precipitation is approximately 33 inches (Robbins, 1991). While the low winter temperatures ensure that many bird species that depend on freshwater habitats migrate out of the area, the high summer temperatures and precipitation ensure that vegetation growth is lush, with associated diverse bird habitats and communities.

2.2 AVIAN DIVERSITY IN THE ASSESSMENT AREA

Green Bay and the Lower Fox River is an important site within the Great Lakes ecoregion for breeding and migratory birds (Temple and Cary, 1987; Erdman and Jacobs, 1991; Robbins, 1991). During the five years of the Wisconsin Checklist Project, from 1982 until 1986, observers recorded over 250 bird species in the five Wisconsin counties (Door, Kewaunee, Brown, Oconto, and Marinette) immediately adjacent to Green Bay and the Lower Fox River (Table 2-1). During the Atlas of Breeding Birds of Michigan project (1983-1988), 91 bird species were found breeding in the townships adjacent to the Michigan Green Bay shore (Brewer et al., 1991). This high degree of species richness is largely due to four factors: the proximity of a major bird migration route, longitude, plant community diversity, and high quality habitat.

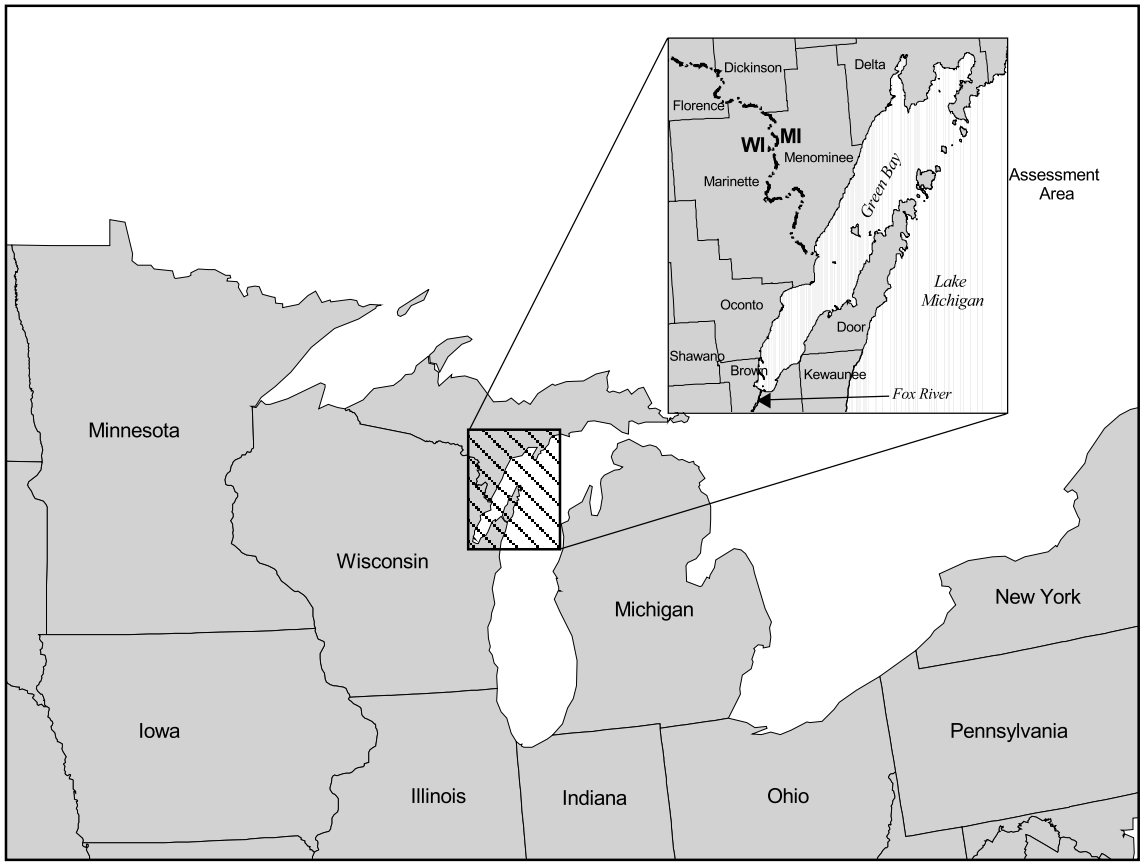


Figure 2-1. The location of the assessment area.

Green Bay and the Lower Fox River are situated on one of the major bird migration routes in North America — the Mississippi Flyway (Figure 2-2). Birds flying south during the fall from their breeding areas in Canada and flying north in the spring funnel through the Lake Michigan and Green Bay area. This results in the regular occurrence of many species that neither breed nor winter in the area (e.g., tundra swans, oldsquaw, and a large number of shorebird species).

The most spectacular of these migratory movements involves the fall influx of waterfowl species into Green Bay. Hundreds of thousands of ducks and geese traveling south from northerly breeding areas use the wetlands surrounding the bay as roosting and feeding areas. These species, which include Canada goose, mallard, teal species, scaup, goldeneye, and many others, are the basis for the intense and economically important duck hunting that takes place in the bay each fall.

Because of its longitude, the assessment area supports birds that are typical of both more western and eastern habitats. For example, both the western and the eastern meadowlark were recorded in the Green Bay area during the Wisconsin Checklist Project, as were the western marbled godwit

Table 2-1
Bird Species Recorded during the Wisconsin Checklist Project in Assessment Area Counties from 1982 until 1986

Species[a]	Breeding/ Summer Visitor	Migrant	Winter Visitor	Year Round Resident	Seasonal Status Uncertain	Federal Status[b]	State Status[b]
Red-throated loon		✔					
Common loon	✔						
Pied-billed grebe	✔						
Horned grebe		✔					
Red-necked grebe		✔					E
Double-crested cormorant	✔	✔					
American bittern	✔						
Least bittern	✔						
Great blue heron	✔						
Green-backed heron	✔						
Great egret	✔						T
Cattle egret	✔						
Black-crowned night-heron	✔						
Tundra swan		✔					
Mute swan				✔			
Snow goose		✔					
Wood duck	✔	✔					
Canada goose		✔		✔			
Green-winged teal	✔	✔					
American black duck		✔		✔			
Mallard		✔		✔			
Northern pintail	✔	✔					
Blue-winged teal	✔	✔					
Northern shoveller		✔					
Gadwall	✔	✔					
American wigeon		✔					
Canvasback		✔					
Redhead	✔	✔					
Ring-necked duck	✔	✔					
Greater scaup	✔	✔					

a. Bird species in italics obtain their food from aquatic habitats and therefore are at increased risk of exposure to PCBs.
b. T = threatened, E = endangered.

Table 2-1 (cont.)
Bird Species Recorded during the Wisconsin Checklist Project in Assessment Area Counties from 1982 until 1986

Species[a]	Breeding/ Summer Visitor	Migrant	Winter Visitor	Year Round Resident	Seasonal Status Uncertain	Federal Status[b]	State Status[b]
Lesser scaup	✔	✔					
Oldsquaw			✔				
White-winged scoter		✔					
Common goldeneye		✔		✔			
Bufflehead		✔	✔				
Hooded merganser	✔	✔					
Common merganser		✔		✔			
Red-breasted merganser	✔	✔					
Ruddy duck		✔					
Turkey vulture	✔						
Osprey	✔						T
Bald eagle				✔		T	
Northern harrier	✔	✔					
Sharp-shinned hawk		✔		✔			
Cooper's hawk		✔		✔			
Northern goshawk				✔			
Red-shouldered hawk	✔						T
Broad-winged hawk	✔	✔					
Red-tailed hawk		✔		✔			
Rough-legged hawk		✔		✔			
American kestrel		✔		✔			
Merlin	✔						
Peregrine falcon		✔				E	E
Gray partridge				✔			
Ring-necked pheasant				✔			
Ruffed grouse				✔			
Sharp-tailed grouse				✔			
Wild turkey				✔			
Northern bobwhite	✔						
Virginia rail	✔						

a. Bird species in italics obtain their food from aquatic habitats and therefore are at increased risk of exposure to PCBs.
b. T = threatened, E = endangered.

Table 2-1 (cont.)
Bird Species Recorded during the Wisconsin Checklist Project in Assessment Area Counties from 1982 until 1986

Species[a]	Breeding/ Summer Visitor	Migrant	Winter Visitor	Year Round Resident	Seasonal Status Uncertain	Federal Status[b]	State Status[b]
Sora	✔	✔					
Common moorhen	✔						
American coot	✔	✔					
Sandhill crane	✔	✔					
Black-bellied plover		✔					
Killdeer	✔						
Greater yellowlegs		✔					
Lesser yellowlegs		✔					
Solitary sandpiper	✔	✔					
Willet		✔					
Spotted sandpiper	✔	✔					
Upland sandpiper	✔	✔					
Hudsonian godwit		✔					
Marbled godwit		✔					
Ruddy turnstone		✔					
Red knot		✔					
Sanderling		✔					
Semipalmated sandpiper		✔					
Least sandpiper		✔					
White-rumped sandpiper		✔					
Baird's sandpiper		✔					
Pectoral sandpiper		✔					
Dunlin		✔					
Stilt sandpiper		✔					
Short-billed dowitcher		✔					
Long-billed dowitcher		✔					
Common snipe	✔	✔					
American woodcock	✔	✔					
Wilson's phalarope	✔	✔					
Red-necked phalarope		✔					

a. Bird species in italics obtain their food from aquatic habitats and therefore are at increased risk of exposure to PCBs.
b. T = threatened, E = endangered.

Table 2-1 (cont.)
Bird Species Recorded during the Wisconsin Checklist Project in Assessment Area Counties from 1982 until 1986

Species[a]	Breeding/ Summer Visitor	Migrant	Winter Visitor	Year Round Resident	Seasonal Status Uncertain	Federal Status[b]	State Status[b]
Franklin's gull		✔					
Bonaparte's gull	✔	✔					
Ring-billed gull				✔			
Herring gull				✔			
Glaucous gull			✔				
Caspian tern	✔	✔					E
Common tern	✔	✔					E
Forster's tern	✔	✔					E
Black tern	✔						
Rock dove				✔			
Mourning dove				✔			
Black-billed cuckoo	✔						
Yellow-billed cuckoo	✔						
Eastern screech owl					✔		
Great horned owl				✔			
Snowy owl			✔				
Barred owl				✔			
Long-eared owl	✔						
Short-eared owl				✔			
Northern saw-whet owl					✔		
Common nighthawk	✔	✔					
Whip-poor-will	✔	✔					
Chimney swift	✔	✔					
Ruby-throated hummingbird	✔	✔					
Belted kingfisher	✔	✔					
Red-headed woodpecker				✔			
Red-bellied woodpecker				✔			
Yellow-bellied sapsucker	✔	✔					
Downy woodpecker				✔			
Hairy woodpecker				✔			

a. Bird species in italics obtain their food from aquatic habitats and therefore are at increased risk of exposure to PCBs.
b. T = threatened, E = endangered.

Table 2-1 (cont.)
Bird Species Recorded during the Wisconsin Checklist Project in Assessment Area Counties from 1982 until 1986

Species[a]	Breeding/ Summer Visitor	Migrant	Winter Visitor	Year Round Resident	Seasonal Status Uncertain	Federal Status[b]	State Status[b]
Northern flicker	✔	✔		✔			
Pileated woodpecker				✔			
Olive-sided flycatcher		✔					
Eastern wood-pewee	✔	✔					
Yellow-bellied flycatcher		✔					
Alder flycatcher	✔	✔					
Willow flycatcher	✔						
Least Flycatcher	✔	✔					
Eastern phoebe	✔	✔					
Great crested flycatcher	✔	✔					
Eastern kingbird	✔	✔					
Horned lark		✔		✔			
Purple martin	✔	✔					
Tree swallow	✔						
Northern rough-winged swallow	✔	✔					
Bank swallow	✔	✔					
Cliff swallow	✔						
Barn swallow	✔	✔					
Gray jay				✔			
Blue jay				✔			
American crow				✔			
Common raven				✔			
Black-capped chickadee				✔			
Boreal chickadee					✔		
Tufted titmouse					✔		
Red-breasted nuthatch				✔			
White-breasted nuthatch				✔			
Brown creeper				✔			
House wren	✔	✔					

a. Bird species in italics obtain their food from aquatic habitats and therefore are at increased risk of exposure to PCBs.
b. T = threatened, E = endangered.

Table 2-1 (cont.)
Bird Species Recorded during the Wisconsin Checklist Project in Assessment Area Counties from 1982 until 1986

Species[a]	Breeding/ Summer Visitor	Migrant	Winter Visitor	Year Round Resident	Seasonal Status Uncertain	Federal Status[b]	State Status[b]
Winter wren				✔			
Sedge wren	✔						
Marsh wren	✔						
Blue-gray gnatcatcher		✔					
Eastern bluebird	✔						
Veery	✔	✔					
Gray-cheeked thrush		✔					
Swainson's thrush		✔					
Hermit thrush	✔	✔					
Wood thrush	✔	✔					
American robin	✔			✔			
Gray catbird	✔	✔					
Northern mockingbird	✔						
Brown thrasher	✔	✔					
Water pipit		✔					
Bohemian waxwing			✔				
Cedar waxwing	✔			✔			
Northern shrike			✔				
Loggerhead shrike	✔						E
European starling				✔			
Bell's vireo	✔						T
Solitary vireo	✔	✔					
Yellow-throated vireo	✔						
Warbling vireo	✔	✔					
Philadelphia vireo	✔	✔					
Red-eyed vireo	✔	✔					
Blue-winged warbler		✔					
Golden-winged warbler	✔	✔					
Tennessee warbler		✔					
Orange-crowned warbler		✔					

a. Bird species in italics obtain their food from aquatic habitats and therefore are at increased risk of exposure to PCBs.
b. T = threatened, E = endangered.

Table 2-1 (cont.)
Bird Species Recorded during the Wisconsin Checklist Project in Assessment Area Counties from 1982 until 1986

Species[a]	Breeding/ Summer Visitor	Migrant	Winter Visitor	Year Round Resident	Seasonal Status Uncertain	Federal Status[b]	State Status[b]
Nashville warbler	✔	✔					
Northern parula		✔					
Yellow warbler	✔	✔					
Chestnut-sided warbler	✔	✔					
Magnolia warbler		✔					
Cape May warbler		✔					
Black-throated blue warbler	✔	✔					
Yellow-rumped warbler	✔	✔					
Black-throated green warbler	✔	✔					
Blackburnian warbler	✔	✔					
Pine warbler	✔	✔					
Palm warbler		✔					
Bay-breasted warbler		✔					
Blackpoll warbler		✔					
Cerulean warbler		✔					T
Black-and-white warbler	✔	✔					
American redstart	✔	✔					
Prothonotary warbler	✔						
Ovenbird	✔	✔					
Northern waterthrush	✔	✔					
Louisiana waterthrush		✔					
Connecticut warbler		✔					
Mourning warbler	✔	✔					
Common yellowthroat	✔	✔					
Hooded warbler		✔					T
Wilson's warbler		✔					
Canada warbler		✔					
Yellow-breasted chat		✔					
Scarlet tanager	✔	✔					
Northern cardinal				✔			

a. Bird species in italics obtain their food from aquatic habitats and therefore are at increased risk of exposure to PCBs.
b. T = threatened, E = endangered.

Table 2-1 (cont.)
Bird Species Recorded during the Wisconsin Checklist Project in Assessment Area Counties from 1982 until 1986

Species[a]	Breeding/ Summer Visitor	Migrant	Winter Visitor	Year Round Resident	Seasonal Status Uncertain	Federal Status[b]	State Status[b]
Rose-breasted grosbeak	✔	✔					
Indigo bunting	✔	✔					
Dickcissel	✔						
Rufous-sided towhee	✔	✔					
American tree sparrow		✔	✔				
Chipping sparrow	✔						
Clay-colored sparrow	✔						
Field sparrow	✔						
Vesper sparrow	✔						
Lark sparrow					✔		
Savannah sparrow	✔						
Grasshopper sparrow	✔						
Le Conte's sparrow	✔						
Fox sparrow		✔					
Song sparrow	✔	✔		✔			
Lincoln's sparrow	✔	✔					
Swamp sparrow	✔	✔					
White-throated sparrow	✔	✔					
White-crowned sparrow		✔					
Harris's sparrow		✔					
Dark-eyed junco	✔	✔	✔				
Lapland longspur		✔					
Snow bunting		✔	✔				
Eastern meadowlark	✔						
Western meadowlark	✔						
Yellow-headed blackbird	✔						
Red-winged blackbird	✔						
Rusty blackbird		✔					
Brewer's blackbird	✔						
Common grackle	✔	✔		✔			

a. Bird species in italics obtain their food from aquatic habitats and therefore are at increased risk of exposure to PCBs.
b. T = threatened, E = endangered.

Table 2-1 (cont.)
Bird Species Recorded during the Wisconsin Checklist Project in Assessment Area Counties from 1982 until 1986

Species[a]	Breeding/ Summer Visitor	Migrant	Winter Visitor	Year Round Resident	Seasonal Status Uncertain	Federal Status[b]	State Status[b]
Brown-headed cowbird	✔	✔		✔			
Orchard oriole		✔					
Northern oriole	✔	✔					
Pine grosbeak			✔				
Purple finch				✔			
Red crossbill				✔			
White-winged crossbill					✔		
Common redpoll			✔				
Pine siskin		✔		✔			
American goldfinch	✔			✔			
Evening grosbeak		✔	✔	✔			
House sparrow				✔			

a. Bird species in italics obtain their food from aquatic habitats and therefore are at increased risk of exposure to PCBs.
b. T = threatened, E = endangered.

Source: Temple and Cary, 1987.

and the eastern Hudsonian godwit. The assessment area is also one of the easternmost breeding sites for the yellow-headed blackbird, and the white pelican (another western species) has recently colonized the assessment area as a breeding species (K. Stromborg, USFWS, personal communication, April 1999). This mixing of western and eastern birds adds to the avifaunal diversity of the assessment area.

Third, within Wisconsin there is a north-south shift in the major plant communities due to climate. The assessment area is located in a transitional zone called the “Tension Zone” (Curtis, 1959), where plant communities that are typical of both major ecoregions can be found. Areas north of the Lower Fox River are dominated by plant communities that are representative of higher, colder latitudes (e.g., an increased dominance of conifer forests). Northern Door County includes subarctic plant communities because of its low warmest daily average temperatures in summer, which are caused by a marked lake effect and Lake Michigan upwelling. Areas to the south have communities adapted to a warmer climate (e.g., hardwood forests). This results in the occurrence within the assessment area of bird species that are typical of both the more northern plant

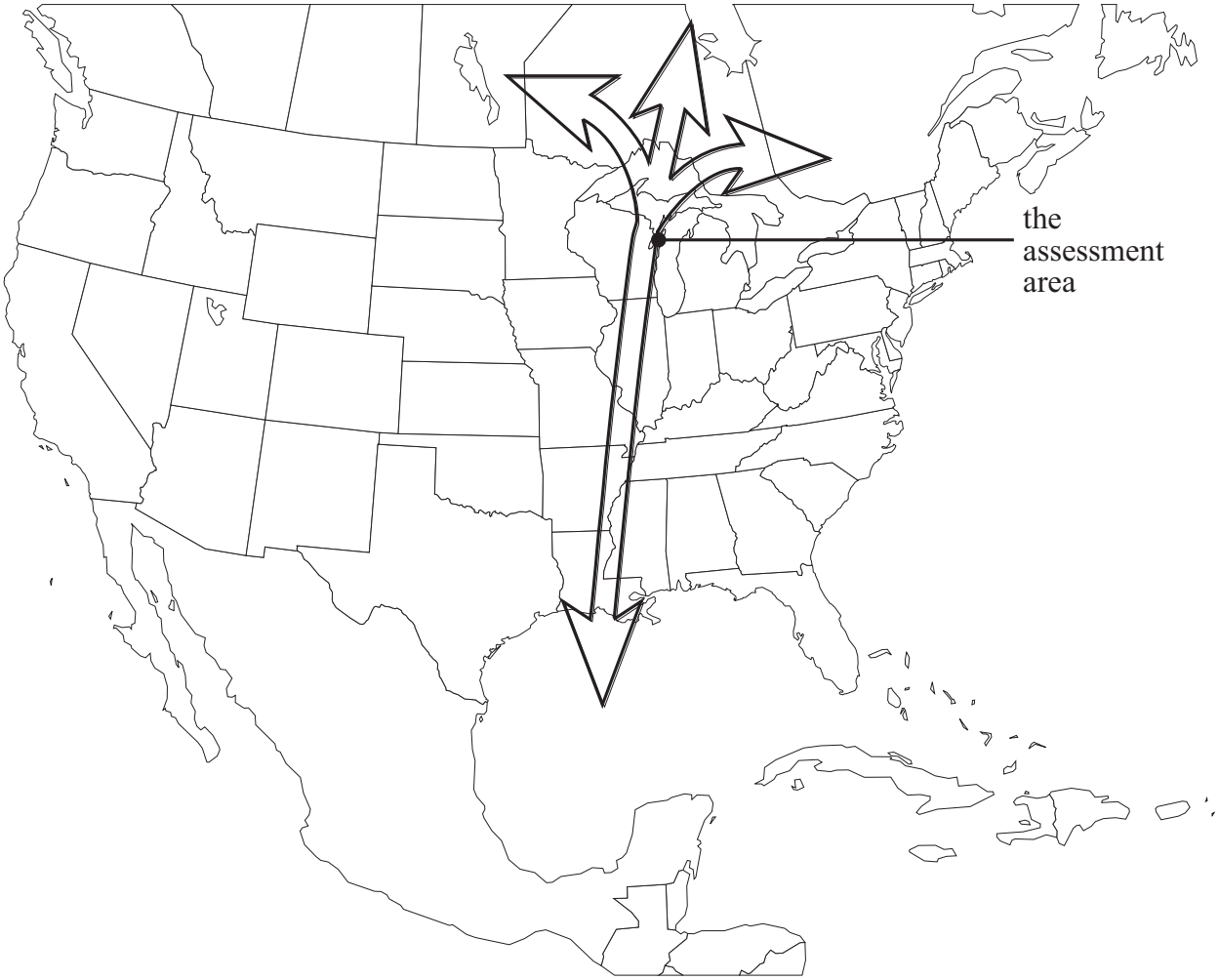


Figure 2-2. The assessment area in relation to the Mississippi Flyway.

communities (e.g., gray jay, common raven, boreal chickadee, and several *Dendroica* warbler species), together with species more characteristic of southern habitat types (e.g., turkey vulture, mourning dove, and tufted titmouse).

Last, the bird species diversity found in the assessment area is supported by the area's high quality habitats. While many of the birds that breed in or use the area as a migratory staging post can be found elsewhere in the lower, industrialized Great Lakes region, the assessment area, because of its comparatively undisturbed nature and the quality and extent of its habitats, supports more diverse bird communities.

Wetlands are an important habitat for nesting and migratory birds in the assessment area. Figure 2-3 shows that extensive tracts of the west side of the bay comprise coastal wetlands. There are over 250,000 acres of wetlands within five miles of the bay, much of which (almost 52,000 acres) is protected as national forest, state forest, state park, or state wildlife area (complied from data from USGS, 1990; Wisconsin DNR, 1998b). These extensive and contiguous tracts of wetland provide ideal habitat for migratory and nesting birds.

Other important and abundant bird habitats in the assessment area are the small uninhabited islands of Green Bay, which provide nesting sites for colonial waterbirds. Figure 2-4 shows the distribution of these nesting sites and potential nesting sites. Such sites are favored by colonial waterbirds because of their freedom from human disturbance and from mammalian predators such as raccoons, mink, foxes, and coyotes. Many of these islands are well known as waterbird breeding sites and have supported colonies for many years (e.g., herring gulls on Big Sister Island, cormorants on Hat, Spider, and Cat Islands, Caspian terns on Gravelly Island).

2.3 AVIAN RESOURCES ON THE ONEIDA RESERVATION

The proximity of the Oneida Reservation to the Lower Fox River and Green Bay means that environmental changes to the Lower Fox River/Green Bay ecosystem directly impact the reservation ecosystems. The Oneida Reservation comprises approximately 65,400 acres located 3 miles east of the lower Fox River and 2.5 miles south of Green Bay, incorporating part of the city of Green Bay. It is directly connected to the larger assessment area through waterways. All of the major reservation waters are tributaries to the Lower Fox River and Green Bay. Land use within the exterior reservation boundaries ranges from commercial and light industrial to rural agriculture to residential. The northeastern quarter of the reservation is dominated by residential and commercial land uses, while the remaining areas of the reservation are low intensity agricultural, wetlands, and forested lands.

Birds that have been sighted on the Oneida Reservation in recent years include belted kingfisher, sandhill crane, great blue heron, great horned owl, barn owl, screech owl, northern harrier, rough-legged hawk, common nighthawk, turkey vulture, red-tailed hawk, bald eagle, double-crested cormorant, and 12 different species of ducks and geese. Migratory birds that use the reservation as a stopover site include tundra and trumpeter swans and snow geese. Several species of game birds, such as wild turkey, have been successfully reintroduced to the reservation. While this is not a comprehensive list of the birds that use the reservation ecosystem, it is a representative list of the types of birds on the reservation.

The environment of the reservation is important to both the resident and the migratory bird populations that use the Mississippi Flyway. Wetlands on the reservation become waterbird breeding colonies in the spring, and many species such as sandhill cranes have been found nesting in local wetlands. Threatened species such as bald eagles have used the open waters of the

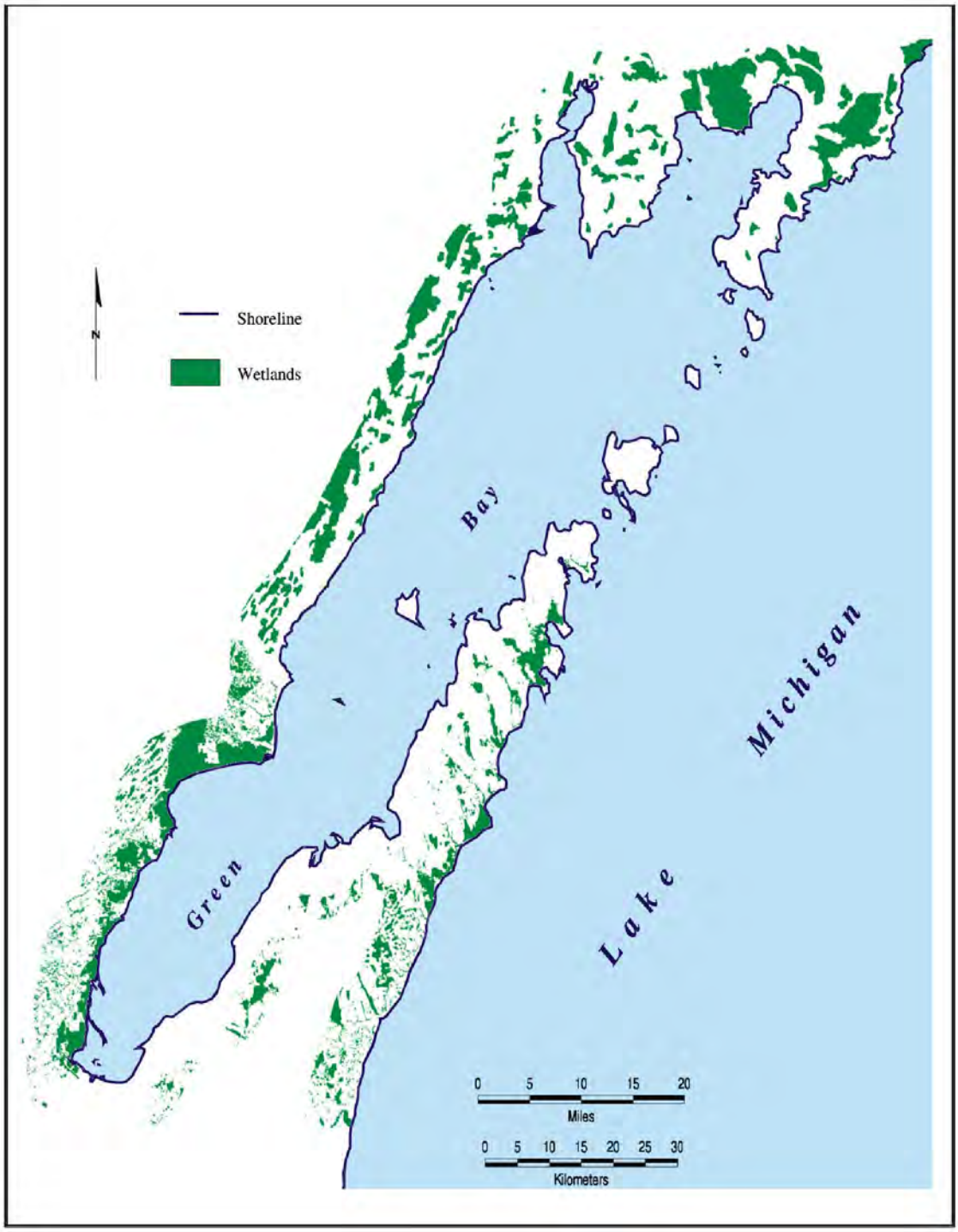


Figure 2-3. Distribution of wetland habitat within five miles of Green Bay or portions of Lake Michigan.

Source: Compiled from data from USGS, 1990, and Wisconsin DNR, 1998b.

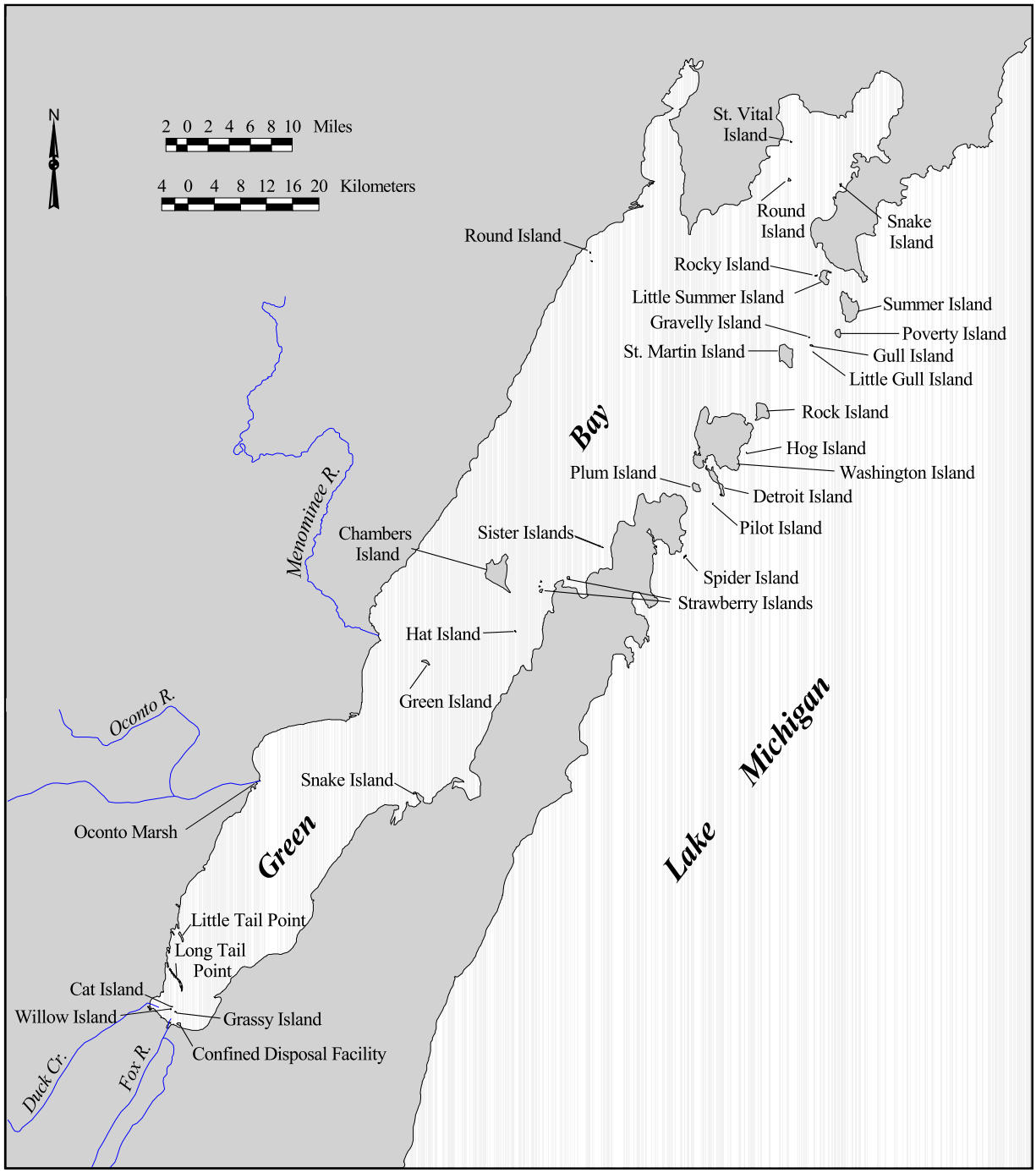


Figure 2-4. Nesting and potential nesting islands for colonial waterbirds in the assessment area.

reservation for winter feeding areas and double-crested cormorants have been seen foraging in reservation lakes and ponds. These examples illustrate the importance of the Oneida Reservation to the Lower Fox River/Green Bay environment.

The combination of agricultural fields and wetlands on the reservation provides ideal habitat for waterfowl. Reservation wetlands provide necessary cover for nesting waterfowl in close proximity to farm fields that supply food for many of the birds. Waterfowl use the many engineered ponds and wetlands on the reservation as a replacement for lost habitat along the Fox River and the shore of Green Bay.

The major waterway on the reservation, Duck Creek, received its name from the large number of ducks that use the river for nesting and rearing of their young. The Duck Creek corridor includes habitat that is ideal for the waterfowl. In the spring the water level is high enough to create nesting areas along many reaches, and the riparian zone in some areas is wide enough to provide the necessary cover for the young.

Why the Birds Are Important to the Reservation

Waterfowl and other game birds are important to the reservation as a food source. The Oneida Tribe chose this land when they were relocated from New York because of the abundant game and the similarity to lands they were leaving behind. The original name for the major river flowing through the reservation was “the place of many ducks.” Ducks and other waterfowl became an important part of the Oneida diet. Oral histories from tribal elders explain how they obtained most of their meat from the local population of game, including waterfowl, turkey, and other small game.

The local birds have always been spiritually important to the Oneida People. For example, the bald eagle was instructed by the Creator to head the bird kingdom, and appreciation for the fulfillment of these duties is expressed in the Oneida Thanksgiving Prayer. The eagle sits on top of the “Tree of Life,” ever vigilant against those who would harm the tree, and eagles carry their prayers up to the Creator. The beauty and songs of all the different birds help the Oneida people appreciate their purpose in life and remind them to enjoy their life cycle to its utmost.

Waterfowl have a special role in the creation story of protecting and safely bringing Mother Earth to the back of the Great Turtle who supports the land we walk on. Birds are also used by the tribal elders to explain many of life’s lessons to younger Oneida people. Some Oneida elders have expressed a sense of shame, loss, and sorrow because of the decreased numbers and diversity of birds. In the Oneida culture it is important to protect and preserve all of the animals, and birds have a special place in the Oneida culture. These feelings are expressed in the Oneida Thanksgiving Prayer.

> . . . Now then, I will mention . . . the water birds that swim about in the water. Now I will mention, that it is still that it can be for our good minds, good feelings, and a medicine for all people, many places the water has been polluted, and this was created by the human family, and it has created a great suffering in our minds, that we can no longer eat the fish safely, and surely it has caused great suffering to all the fish families as far as they carry on to. Now then we mention that we apologize to the waters and all her inhabitants on behalf of the humans, and pray that we will restore the waters to how it is intended to be.

Culturally, everything that the Oneida Tribe values is related to the earth's environment. The Thanksgiving Prayer implies that it is the responsibility of all Oneida people to preserve and protect their environment for future generations. The tradition of the "Seventh Generation Commitment" implies that we will honor the Creator and future generations through the protection of Mother Earth.

2.4 REGULATORY STATUS OF ASSESSMENT AREA BIRDS

Green Bay and the Lower Fox River support populations of 13 species that have been listed by the federal government or by the State of Wisconsin as either threatened or endangered (Table 2-1). The continued existence of these sensitive species in the assessment area (and the recent colonization by white pelicans) attests to the relatively undisturbed nature of the area and the quality and extent of its habitats.

2.5 BREEDING CYCLES AND LIFE HISTORIES OF ASSESSMENT AREA BIRDS

Of the birds that breed in the assessment area, colonial waterbirds, such as terns and cormorants, and bald eagles have been the most studied in terms of their PCB exposure and PCB-caused effects (see Chapter 5). All of the colonial waterbirds that breed in the assessment area are migratory, arriving at their breeding areas in the spring and leaving for their wintering areas in late fall. Thus, all of the components of their breeding cycles are contained within the short time span between approximately April and August. Figure 2-5 presents a typical breeding chronology for waterbirds within the assessment area.

The life histories of the principal colonial waterbirds of the area vary. Common and Caspian terns are exclusively ground nesters with little or no nest construction, whereas Forster's terns nest on substantial, reed nests on floating mats of vegetation. All of the terns typically nest in dense colonies with often only 2 or 3 feet between neighboring nests (Ehrlich et al., 1988). They lay clutches of two to three eggs, which are incubated over about three weeks. The young are semiprecocial in that they can leave the nest soon after hatching but depend on the adults for their food and, for the first few days after hatching, for thermoregulation (Ehrlich et al., 1988). The

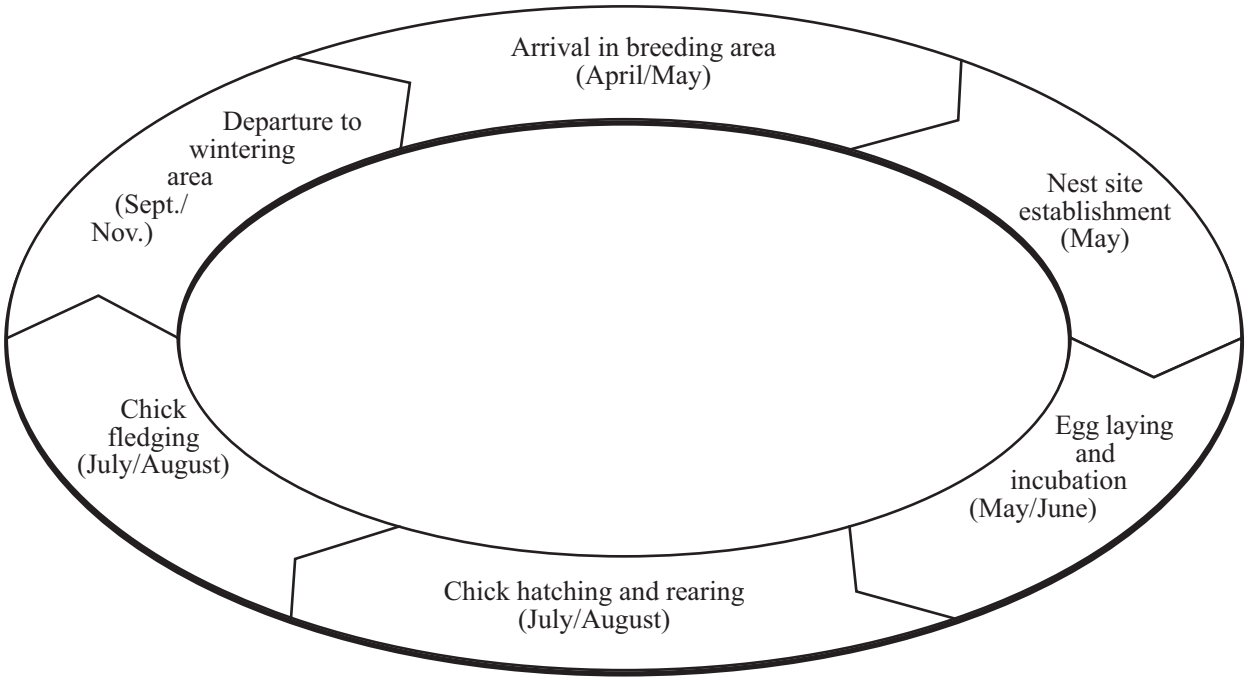


Figure 2-5. Typical breeding cycle of Green Bay colonial waterbirds.

diets of both the adults and the young are mainly forage fish (Cramp and Simmons, 1977). After about three to four weeks the young are capable of flight and leave the nesting areas in family parties. By late fall, they have left for their wintering areas in the Gulf of Mexico.

Double-crested cormorants are also summer visitors to the assessment area, arriving at their nesting colonies in April and May. They build their nests on the ground (at sites where trees are not available and mammalian predators are not a problem) or in trees (as at Cat Island, until recently). Like the terns, cormorants nest in dense colonies with only a few feet between nests. They lay three to five eggs, which are incubated for about four weeks. The nestlings are altricial, relying on their parents for their food and, in the first two weeks of life, for thermoregulation. After five to six weeks, the young are capable of flight and leave the colonies with their parents. The return to their wintering areas in the Mississippi Valley and the Gulf of Mexico begins in September. Like the three tern species, double-crested cormorants mainly eat forage fish (see Chapter 5).

Bald eagles differ substantially in their life histories from the four species discussed previously. Unlike the terns and cormorants, they are not colonial but are widely dispersed over the landscape. They may be year-round residents in the assessment area, depending on the availability of prey (Robbins, 1991). They also begin breeding much earlier in the year, with most nests being

refurbished in February and March. Nests are typically bulky structures of twigs and branches and are built high in trees. Often the same nest may be used year after year. Neighboring nests may typically be up to several kilometers apart. One to three eggs are laid in March, and the young hatch after about 35 days of incubation. The young are completely altricial and dependent on their parents for food and, in the early stages of growth, for heat. Fledging takes place about 12 weeks after hatching. The prey of bald eagles comprises fish, carrion, and other birds such as herring gulls and cormorants (see Chapter 5).

2.6 CONCLUSIONS

Because of its diversity of habitats, geographical position relative to east-west and north-south gradients in bird communities, local climate patterns, and proximity to the Mississippi Flyway, the assessment area supports a rich diversity of bird species. Over 250 species have been recorded. These include breeding birds and summer visitors, fall and spring migrants, and winter visitors. Furthermore, at least 13 species that are listed by either the State of Wisconsin or the federal government as threatened or endangered are found in the assessment area.

Many of the species in the assessment area are dependent on the large tracts of relatively undisturbed habitat in the area. This is particularly true for birds that depend on wetlands or uninhabited islands for breeding, resting, and feeding sites. The assessment area provides these critical habitats in abundance.

The Oneida Reservation provides habitat for many bird species. Furthermore, birds are an important part of the Oneida Tribe culture, as reflected in oral histories and tribal prayers.

CHAPTER 3
TOXIC EFFECTS OF PCBS ON BIRDS

As described in the NRDA Assessment Plan (61 Fed. Reg. 43,558) and as noted in Chapter 1, the Fox River/Green Bay ecosystem has been contaminated with PCBs. This chapter provides an overview of the chemistry of PCBs and the toxic effects of PCBs on birds. This information provides background for the injuries assessed in subsequent chapters.

This chapter is organized as follows. Section 3.1 presents a brief overview of PCB chemistry. Section 3.2 provides a toxicological taxonomy of PCB congeners based on their mode of action. Section 3.3 discusses the effects of PCBs on birds and relates these effects to categories of injury established in the Department's NRDA regulations. Section 3.4 discusses the toxic potency of PCB congeners in birds. Section 3.5 summarizes published studies on the concentrations of PCBs shown to cause adverse effects and includes a discussion of avian sensitivity to PCB toxicity. Section 3.6 presents conclusions.

3.1 PCB CHEMISTRY

PCBs are a class of 209 chlorinated biphenyl congeners that differ in the total number and position of chlorine atoms substituted on the biphenyl structure (Figure 3-1).

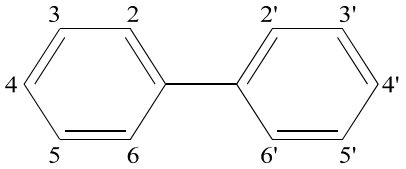


Figure 3-1. Biphenyl molecular structure.

As shown in Figure 3-1, the PCB structure is made up of two benzene (biphenyl) rings linked by a single bond. The 209 possible PCB congeners are identified by the location and number of chlorine atom substitutions on the biphenyl rings, with 10 chorine atoms being the maximum number possible. For example, the congener 2,2' dichlorobiphenyl has chlorine atoms at the 2 and 2' positions. PCB congeners are also identified by a sequential numbering system based on increasing chlorine substitution from PCB 1 (2-chlorobiphenyl; 1 chlorine) to PCB 209 (2,2',3,3',4,4',5,5',6,6'-decachlorobiphenyl; 10 chlorines).

PCBs do not occur naturally in the environment.[1] PCBs were introduced into the environment as commercial mixtures of congeners (e.g., trade names of Aroclor in the United States, Clophen in Germany, Kaneclor in Japan), with the congener composition dependent on the manufacturing process (U.S. EPA, 1980). Most commercial mixtures were differentiated by the average percentage chlorine by weight (e.g., Aroclor 1242 contained 42% chlorine). Aroclor 1242, the predominant commercial mixture involved in Fox River paper company processes (Carr et al., 1977), consists of a mixture of approximately 80 congeners (Schulz et al., 1989), with a mean number of 3.1 chlorine atoms per molecule (Eisler, 1986). Quantifiable congeners in Aroclor 1242 extend from 2,2'-dichlorobiphenyl (two chlorines; PCB 4) to 2,2',3,4,4',5,5'-heptachlorobiphenyl (seven chlorines; PCB 180) (Schulz et al., 1989).

In the environment, organisms may be exposed to mixtures of PCB congeners that no longer resemble the original commercial Aroclor. This is because physical and chemical environmental fate processes such as evaporation, transport, biodegradation, and partitioning onto sediments can alter the mixture of congeners to which biota may be exposed (Safe, 1994). Moreover, once accumulated by organisms, PCB congeners may be differentially excreted, distributed, biotransformed, or sequestered (e.g., deposition in lipids) (Rozemeijer et al., 1995; summarized by Barron et al., 1995).

3.2 TOXICOLOGICAL TAXONOMY OF PCB CONGENERS

Many individual PCB congeners have been found to cause adverse effects to biota. These effects can differ based on the specific chemical composition of the individual congener or congener mixtures. The different adverse effects caused by PCBs can be classified according to the manner in which they manifest toxicity, known as their “toxicological mode of action” (Table 3-1).

3.2.1 Dioxin-Like Toxicity

Dioxin-like congeners are known as co-planar PCBs because the biphenyl rings lie in the same two-dimensional plane, giving them a molecular configuration similar to 2,3,7,8-tetrachlorodibenzo-p-dioxin (dioxin; TCDD) (Safe, 1994). These congeners have chlorine substitutions in the 3,3',4,4' positions (meta-para substitutions) and have either zero (nonortho), one (mono-ortho), or two (di-ortho) chlorines in the 2 or 2' positions (Figure 3-2). This chlorine substitution pattern increases the structural similarity of the congeners to TCDD, inhibits metabolic transformation by organisms, and generally increases biological persistence (Safe, 1994). Dioxin-like PCBs have affinity for the same cellular receptor (the aryl hydrocarbon or Ah

1. Minor concentrations of PCBs may be produced by volcanic processes (Lamparski et al., 1990). However, data in Lamparski et al. (1990) from the Mount Saint Helen’s eruption suggest that PCBs observed in volcanic ash might be scavenged from the atmosphere rather than a product of vulcanism.

Table 3-1
Toxicological Taxonomy of PCB Congeners

Mode of Action	Toxic Effects	Example References
Dioxin-like	Edema, deformities, early life stage mortality, uroporphyrin accumulation	Safe, 1994 Bosveld, 1995 Barron et al., 1995
Phenobarbital-like	Tumor promotion, uroporphyrin accumulation	Safe, 1994 Rodman et al., 1991
Neurotoxic	Decreased dopamine, behavioral/ neuromuscular alterations	Safe, 1994 Choksi et al., 1997
Endocrine-disrupting	Estrogen mimics, metabolized to biphenylols (thyroxine mimics; Vitamin A effects)	Jansen et al., 1993 Korach et al., 1987 Walker, 1995

receptor) as TCDD. Like TCDD, dioxin-like PCBs are strong inducers of the chemical metabolizing enzyme system known as the P450 family, and exposure to a dioxin-like PCB congener can cause a substantial increase in the concentration and activity of P450 enzymes. Dioxin-like PCBs are strong inducers of the P4501A isoform, and this is the key to their toxicological characteristics. Figure 3-3 provides a simplified schematic representation of TCDD interaction with the Ah receptor showing (1) TCDD movement into a cell, (2) binding to the Ah receptor in the cytoplasm, (3) translocation of the TCDD:Ah receptor complex to the nucleus, (4) production of messenger RNA (mRNA) in the nucleus, (5) translocation of mRNA to the cytoplasm, and (6) synthesis of P450 in organelles (e.g., endoplasmic reticulum) and generation of other cellular responses.

PCBs elicit a suite of toxic effects that are similar to those of TCDD, such as pericardial and abdominal edema, and deformities of the heart, eyes, limbs, head, and body (Bosveld, 1995; Henshel et al., 1997). Although mono-ortho substituted biphenyls are less potent than nonortho congeners, they occurred at higher concentrations in commercial PCB mixtures, and thus may contribute substantially to toxicity in the environment (Braune and Norstrom, 1989; Brunstrom, 1990).

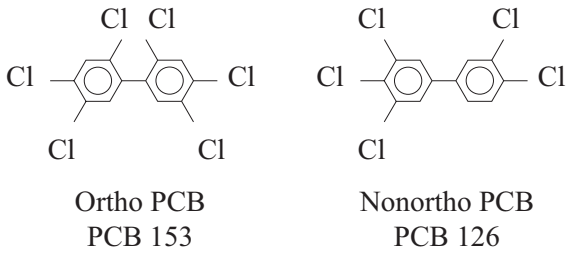


Figure 3-2. Example nonortho (coplanar) and ortho-substituted PCBs.

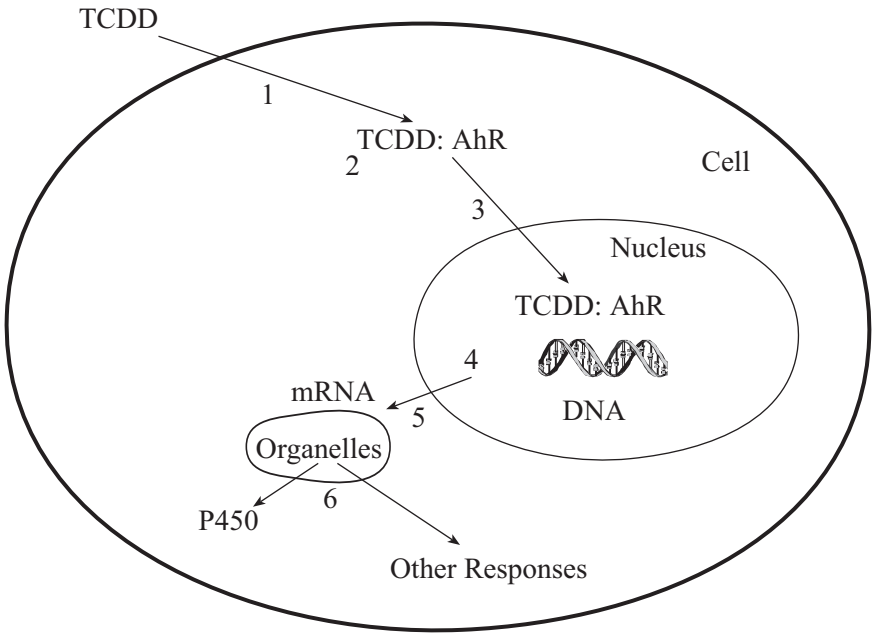


Figure 3-3. Schematic representation of TCDD interaction with the ah receptor.
(1) TCDD movement into a cell, (2) binding to the Ah receptor in the cytoplasm, (3) translocation of the TCDD:Ah receptor complex to the nucleus, (4) production of messenger RNA (mRNA) in the nucleus, (5) translocation of mRNA to the cytoplasm, and (6) synthesis of P450 in organelles (e.g., endoplasmic reticulum) and generation of other cellular responses.

Source: Simplified from Wilson and Safe, 1998.

3.2.2 Phenobarbital-Like Toxicity

The phenobarbital-like (PB-like) group of PCBs are di-ortho substituted congeners with a low affinity for the Ah receptor, and induce a different P450 isozyme (P4501B-like) in birds and other vertebrates than do the dioxin-like congeners (Safe, 1994; Van den Berg et al., 1994). The effects of PB-like PCBs include tumor promotion in rodents (Safe, 1994) and accumulation of uroporphyrin in chick embryo liver cells (hepatocytes) at high doses (Rodman et al., 1991).

3.2.3 Neurotoxicity

Neurotoxic PCBs are ortho-substituted, cause changes in neuromuscular activity and decreases in dopamine levels (Safe, 1994; Choksi et al., 1997), and are associated with behavioral changes and learning deficits (Tilson et al., 1990; Fisher et al., 1998). Heinz et al. (1980) suggested that depletion of brain neurotransmitter levels by neurotoxic PCBs may result in abnormal behavior in sensitive avian species.

3.2.4 Endocrine-Disrupting Toxicity

Current research indicates that specific PCB congeners and hydroxylated metabolites (Figure 3-4) can act as endocrine disruptors by altering normal hormonal dynamics (Sheffield et al., 1998). For example, exposure of mallards to Aroclor 1254 (20 mg/kg body weight twice per week for five weeks) caused a significant reduction in plasma levels of the thyroid hormone triiodothyronine (Fowles et al., 1997). Thyroid hormones modulate the rate of cellular metabolism (Zubay, 1983).

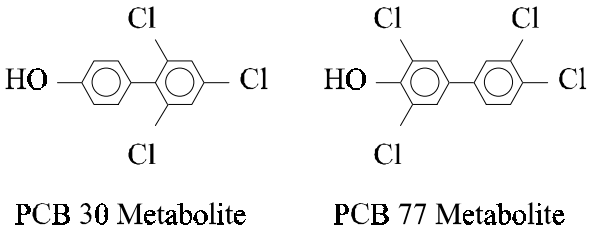


Figure 3-4. Two examples of hydroxylated PCB metabolites (biphenylols) with endocrine disrupting effects.

Biotransformation by an organism generally decreases the toxicity of PCB congeners by increasing their elimination through oxidation (e.g., hydroxylation) and/or conjugation (e.g., glutathione addition) (Figure 3-5). However, hydroxylation at specific sites on several PCB congeners produces hydroxylated biphenylol (HO group inserted into the 4 or 4' position) metabolites that can compete for and occupy hormone or vitamin binding sites (Korach et al., 1987) (Figure 3-4). These water soluble metabolites may be retained in bird eggs, exposing the developing embryo and affecting the functioning of the endocrine system (Fry, 1995). Avian endocrine disruptor effects of PCBs include hyperthyroidism (increased metabolic rate) and hypothyroidism (decreased metabolic rate) in murres (Jefferies and Parslow, 1976) and altered retinoid (vitamin A) dynamics in ring doves (Spear et al., 1989).

Fry (1995) concluded that some PCBs are estrogenic and are responsible for endocrine disruption in breeding birds and abnormalities in their offspring. However, other researchers have not found a relationship between PCBs and endocrine effects. For example, Nisbet et al. (1996) found no relationship between measured PCB congeners in common tern eggs and feminization of male embryos, although the study focused on congeners rather than their metabolites.

Estrogen mimics are thought to include PCBs 1, 9, 10, 30, 52, and 61. These PCBs have estrogenic activity (measured by increased rodent uterine weight; Jansen et al., 1993) or can be metabolized to hydroxylated biphenyls (e.g., 2',4',6'- trichloro-4-biphenylol; 2',3',4',5'-tetrachloro-4-biphenylol) with demonstrated estrogen receptor binding affinity (Korach et al., 1987). Biphenylol (one hydroxylation) and catechol (two hydroxylations) metabolites have been shown to cause endocrine disruption in several vertebrate systems (Fry, 1995; Guillette et al., 1995; Garner et al., 1999), but studies evaluating their estrogenicity in avian embryos have not been reported.

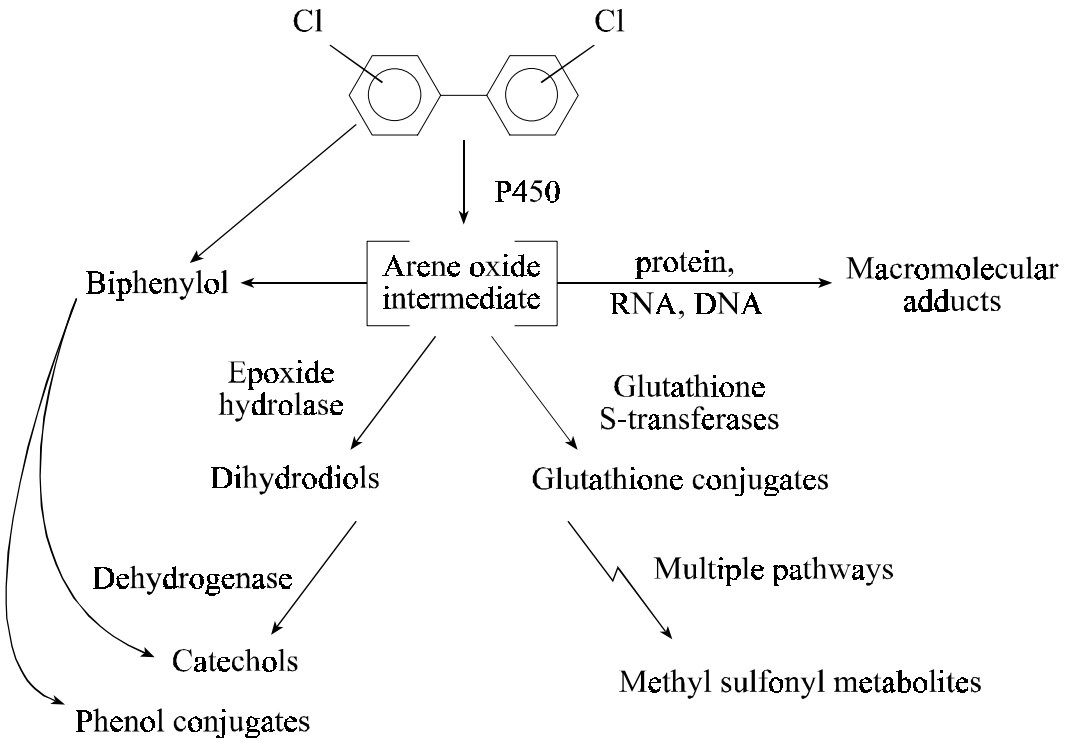


Figure 3-5. Generalized biotransformation pathway for PCBs.

Source: Safe, 1994.

In addition to these estrogen mimics, PCB 77 is thought to act as a thyroxine mimic and may modulate vitamin A levels, resulting in alteration of growth and development. This congener can be hydroxylated to biphenylol metabolites (e.g., 3,3',4,4'-tetrachloro-4-biphenylol; 3,3',4',5-tetrachloro-4-biphenylol) that compete with the binding of the thyroid hormone thyroxine to the transport protein transthyretin with subsequent alteration of growth and development (Brouwer, 1991; Walker, 1995). The activity of thyroid hormones is dependent on binding to carrier proteins in the blood, and competition with PCBs for binding sites may result in changes in metabolic rate through an increase in the “free” hormone (Zubay, 1983).

PCBs can also alter the concentrations of vitamin A in birds, which is important in vision, growth, and reproduction. Retinol is the principal natural form of vitamin A and is primarily stored in the liver as the fatty acid ester retinol palmitate (Environment Canada, 1991). Murk et al. (1994) suggested that PCBs and related contaminants may interfere with the regulation of storage and mobilization of retinoids in the livers of birds, resulting in decreased liver retinoid levels and increased plasma retinoid concentrations. Biphenylol metabolites of PCB 77 may occupy a retinol binding protein, resulting in reduction of blood vitamin A levels and thus altered growth and development (Brouwer, 1991; Walker, 1995).

3.3 PCB EFFECTS ON BIRDS

This section classifies the effects of PCBs on birds according to the injury categories identified in the Departmental NRDA regulations. These regulations define injuries to biological resources [43 CFR § 11.62 (f)], and these definitions can be used to categorize the biological effects of PCBs on birds. Table 3-2 presents, for the Department's NRDA injury categories, examples of scientific studies documenting biological effects of PCBs and concentrations causing adverse effects in birds.

3.3.1 Injury Category: Death [43 CFR § 11.62 (f)(4)(i)]

The experimental studies reported in Table 3-2 (together with many other studies not reported in Table 3-2) show that exposure to PCBs can cause death in avian embryos and juvenile and adult birds.

3.3.2 Injury Category: Disease [43 CFR § 11.62 (f)(4)(ii)]

PCBs are known to affect immune system function in mammalian systems (Safe, 1994), and may cause morphological changes in immune tissues in birds (Nikolaidis et al., 1988). Friend and Trainer (1970) reported increased mortality in mallard ducklings challenged with a duck hepatitis virus following a short-term (10 day) feeding of PCBs (25 to 100 mg/kg diet of Aroclor 1254).

3.3.3 Injury Category: Behavioral Abnormalities [43 CFR § 11.62 (f)(4)(iii)]

PCB-induced behavioral effects in birds include decreased parental incubation attentiveness in ring doves (Peakall and Peakall, 1973), and impaired courtship behavior in mourning doves (Tori and Peterle, 1983). Heinz et al. (1980) found reduced brain dopamine and norepinephrine levels in ring doves fed a 10 mg/kg diet of Aroclor 1254, and suggested that depletion of brain neurotransmitter levels may result in abnormal behavior in sensitive avian species. McCarty and Secord (1999) reported abnormal nest building behavior and lowered nest quality in tree swallows (5 to 7 mg/kg wet weight total PCBs in eggs). Subtle neurological effects such as impaired avoidance behavior in pheasants have also been reported (Dahlgren et al., 1972a).

3.3.4 Injury Category: Cancer and Genetic Mutations [43 CFR § 11.62 (f)(4)(iv)]

Long-term feeding studies of rodents have demonstrated that PCBs increase the incidence of tumors (Safe, 1994). However, our review of the literature identified only one study showing that PCBs cause tumors or genetic mutations in birds. Peakall et al. (1972) reported increased

Table 3-2
Summary of NRDA Injury Categories, Corresponding Biological Effects of PCBs on Birds, and Examples of Studies Documenting Effects

Injury Category [see 43 CFR § 11.62 (f)(4)]	Biological Response	Species Studied	Example Reference
Death	Increased adult/juvenile mortality	Mallard, pheasant, bobwhite, Japanese quail	Heath et al., 1972; Stickel et al., 1984
	Increased embryo mortality	Chicken, pheasant	Carlson and Duby, 1973; Brunstrom and Reutergårdh, 1986
Disease	Increased susceptibility to viral challenge	Mallard	Friend and Trainer, 1970
Behavioral abnormalities	Impaired mating behavior	Mourning dove	Tori and Peterle, 1983
	Impaired avoidance of visual cliff	Pheasant	Dahlgren and Linder, 1971
Cancer, genetic mutations	Chromosome alteration	Ring dove	Peakall et al., 1972
Physiological malfunctions	Reduced reproduction (reduced fecundity)	Pheasant, black- headed gull	Brunstrom and Reutergårdh, 1986
		Chicken	Carlson and Duby, 1973
		American kestrel	Lincer and Peakall, 1970
		Ring dove	Peakall et al., 1972
	Eggshell thinning	Mallard	Haseltine and Prouty, 1980
	Altered endocrine status (e.g., decreased estrogen levels)	Chicken	Chen et al., 1994
	Porphyria	Japanese quail	Elliott et al., 1990
	Enzyme induction (e.g., ED50 for P450)	Turkey	Brunstrom and Lund, 1988
Physical deformations	External malformation (e.g., small beak, eyes, unresorbed yolk sac)	Chicken, common tern, American kestrel	Brunstrom and Lund, 1988; Hoffman et al., 1998
	Skeletal deformities	Common tern, American kestrel	Hoffman et al., 1998
	Histopathological lesions	American kestrel nestlings	Hoffman et al., 1996b

chromosomal aberrations in the embryos of ring doves fed 10 mg/kg wet weight of Aroclor 1254 in their diet for three months. Chromosomal aberrations were 0.8% (range of 0 to 2%) in the control group and 1.8% (range of 0 to 9.4%) in the PCB treated group. A separate NRDA report (Barron et al., 1999) documents the increased frequency of liver tumors in fish (walleye; *Stizostedion vitreum vitreum*) exposed to elevated concentrations of PCBs in the assessment area.

3.3.5 Injury Category: Physiological Malfunctions [43 CFR § 11.62 (f)(4)(v)]

Physiological malfunctions caused by PCBs include reduced reproductive success, eggshell thinning, altered endocrine status, porphyria, altered vitamin A status, and enzyme induction (see Table 3-2).

Reduced Reproductive Success

A number of field and laboratory studies have associated PCB contamination in bird eggs with impaired reproduction, reduced fecundity and fertility, embryotoxicity, and reduced hatchling growth and development (summarized by Gilbertson et al., 1991; Barron et al., 1995; Hoffman et al., 1996a).

Eggshell Thinning

Eggshell thinning has been caused by high concentrations of PCBs in the maternal diet (summarized by Peakall and Lincer, 1996). For example, Haseltine and Prouty (1980) reported eggshell thinning (8.9% thickness reduction) in mallards fed a 105 mg/kg diet of Aroclor 1242. No effects on reproduction were observed. Peakall and Lincer (1996) concluded that PCBs do not cause significant eggshell thinning at environmentally realistic doses.

Altered Endocrine Status

The current understanding of PCB effects on bird endocrine systems is limited. However, reported avian endocrine effects of PCBs have included feminization, lowered estrogen levels, and changes in thyroid function (Colborn et al., 1993). For example, American kestrels fed a 33 mg/kg diet of Aroclor 1254 had reduced semen quality (Bird et al., 1983), and chickens fed a 250 mg/kg diet of Aroclor 1254 had significant reductions in comb and testicle weights (Platonow and Funnell, 1971). PCBs and dioxin-like compounds were associated with gonadal abnormalities in common terns, including the presence of ovarian tissue in the testes of male embryos (Hart et al., 1998). Lincer and Peakall (1970) observed a dose-dependent increase in microsomal metabolism of estradiol in American kestrels fed Aroclor 1254 or Aroclor 1262. Chickens orally administered 10 mg Aroclor 1254 daily for 5 days had reduced plasma estradiol and calcium levels, reduced egg production, decreased liver weight, and increased hepatic P450 content (Chen et al., 1994). Connor et al. (1997) reported that the estrogenicity of hydroxylated PCB congeners determined in

multiple in vitro assays was complex and response-specific, with some assays indicating estrogenicity, no activity, or antiestrogenicity.

Porphyria

Porphyrins are precursors of heme (a component of hemoglobin) and normally occur in small quantities in the body (Environment Canada, 1991). Porphyria, evidenced by increased formation and excretion of porphyrins and precursors, has been reported in several avian species (Goldstein et al., 1976; Fox et al., 1988; Elliott et al., 1997). Accumulation of highly carboxylated porphyrins in herring gull livers has been linked to environmental exposures of birds to PCBs (Environment Canada, 1991). Rodman et al. (1991) demonstrated that specific PCB congeners with three or four ortho chlorines caused increased uroporphyrin in chicken embryo hepatocyte cultures, and Elliott et al. (1990) reported a significant accumulation of liver porphyrins in Japanese quail fed 0.05 mg/kg PCB 126 daily.

Altered Vitamin A (retinoid) Status

Retinol is the principal natural form of vitamin A and is primarily stored in the liver as the fatty acid ester retinol palmitate (Environment Canada, 1991). Murk et al. (1994) suggested that PCBs and related contaminants may interfere with the regulation of storage and mobilization of retinoids in the livers of birds, resulting in decreased liver retinoid levels and increased plasma retinoid concentrations. For example, ring doves exposed to PCB 77 exhibited altered retinoid dynamics, and females laying viable eggs exhibited compensatory retinoid mobilization from the liver and transfer to the eggs (Spear et al., 1989). Murk et al. (1994) concluded that hepatic and yolk sac retinoids may be suitable indicators of early effects of dioxin-like contaminants in common terns and other fish-eating birds.

Enzyme Induction

PCBs have been reported to increase the content or activity of several enzymes in birds, including P450 isozymes (Hoffman et al., 1996a). For example, planar PCBs strongly induce the P450 isozyme CYP1A [measured by increases in aryl hydrocarbon hydroxylase (AHH) or ethoxyresorufin-O-deethylase (EROD) activity]. Each isozyme has a slightly different affinity and capacity for metabolizing contaminants and endogenous biomolecules (e.g., steroids), and may be differentially induced by exposure to PCBs.

Numerous studies have reported EROD induction by dioxin-like PCBs, and some have suggested EROD induction as a sensitive measure of exposure. EROD induction may also be indicative of adverse PCB effects because P450 isozymes may activate or deactivate endogenous biomolecules (e.g., hormones). For example, Lincer and Peakall (1970) reported increased microsomal metabolism of estradiol in American kestrels fed a 0.5 mg/kg diet of Aroclor 1254. Recent in vitro studies demonstrate that EROD activity in bird tissues is suppressed at higher PCB concentrations (e.g., Lorenzen et al., 1997).

3.3.6 Injury Category: Physical Deformations [43 CFR § 11.62 (f)(4)(vi)]

Avian physical deformations caused by PCBs include external deformations, organ and tissue malformations, skeletal deformities, and histopathological lesions (Table 3-2). Physical deformations attributed to PCBs in birds include pericardial and subcutaneous edema, cardiovascular malformation, liver lesions, microphthalmia, beak and limb deformities, brain asymmetry, thymic hypoplasia, and inhibition of lymphoid development (Gilbertson et al., 1991; Bosveld and Van den Berg, 1994; Henshel, 1998; Hoffman et al., 1998). Dioxin-like PCBs may cause deformities in turkey embryos (e.g., microphthalmia and beak deformities) at egg doses lower those causing mortality**,** but may not in other bird species (Nosek et al., 1993). Additionally, exposure of birds to PCB congeners (e.g., PCB 77) and commercial mixtures (e.g., Aroclor 1254) may cause an increase in liver weights (Elliott et al., 1997).

3.4 TOXIC POTENCY OF ENVIRONMENTAL MIXTURES OF PCBS

Different PCB congeners have different potencies in producing toxic responses in birds, as do different mixtures of congeners. Since congener mixtures in the environment are complex and can vary over space, time, and environmental media, several methods have been developed to assess the toxicity of PCB mixtures. These methods are based either on applying information on the potency of individual congeners to congener concentration measurements in media, or on using bioassays to evaluate the toxic potency of the environmental mixture as a whole.

The potency of individual PCB congeners in birds can be determined by their toxicity to avian embryos (e.g., hatching success following injection into bird eggs; Brunstrom, 1990) or from the magnitude of P450 induction caused by a congener (e.g., in vitro induction in avian embryo hepatocytes; Kennedy et al., 1996a, b). Toxicity-based congener potency is derived from studies in which small quantities of graded doses of a PCB congener are injected into the yolk sac, albumin, or air cell of an egg and hatching success or some other response is measured.

Table 3-3 summarizes the potency of selected PCB congeners in inducing P450 in chicken hepatocyte cells. Of the tested congeners, PCB 126 typically is the most potent, followed by PCB 81, PCB 77, and PCB 169. A similar relative order of potency is observed in egg injection studies

Table 3-3
Relative Potency of Selected PCB Congeners in Inducing Chicken Hepatocyte P450

PCB Congener	Concentration Causing 50% P450 Induction (mM)
77	0.51
81	0.094
105	3.3
118	19
126	0.052
169	0.79

Sources: Kennedy et al., 1996b; Lorenzen et al., 1997.

with embryomortality as the endpoint in different bird species (Brunstrom and Andersson, 1988; Brunstrom 1989, 1990; Powell et al., 1996). The general order of potency for congeners is nonortho > mono-ortho > di-ortho > tri-ortho > tetra-ortho.

The toxic potencies of PCB congeners are dependent on both the test species and the toxicity endpoint (e.g., P450 induction versus embryotoxicity). Responsiveness of P450 induction is dependent on the reporter system (e.g., enzyme activity, enzyme content, specific isozyme) and the assay system (e.g., rat versus avian tissue). For example, Kennedy et al. (1996a) concluded that P4501A induction in chicken hepatocyte cultures was more responsive to mono- and di-ortho-PCB congeners than the H4IIE rat hepatoma system, suggesting that birds are more sensitive to these congeners than mammals.

The potency of a PCB congener can be expressed relative to the potency of TCDD (generally the most toxic planar halogenated environmental contaminant) by estimating its toxicity equivalency factor (TEF). The TEF of a congener is determined by dividing the potency response (e.g., concentration at which 50% mortality or 50% P450 induction occurs) by the potency response of TCDD:

$$\text{TEF} = \frac{\text{Potency of PCB congener}}{\text{Potency of TCDD}}$$

Avian TEFs for 12 PCB congeners have been derived by the World Health Organization (WHO) and are summarized in Table 3-4. These values are "generic" in that they are not specific to an individual bird species. The WHO avian TEFs were derived from the results of multiple studies and a variety of experimental data (e.g., embryotoxicity studies, P450 induction), and represent the consensus opinion of an international group of toxicology experts.

TEFs can be used to calculate a TCDD equivalent concentration (TCDD-eq) of PCBs in an environmental sample. A TCDD-eq is the concentration of TCDD that has the same potency as the PCB congener mix and concentration in an environmental sample. A TCDD-eq is calculated by summing the

Table 3-4
Toxic Equivalency Factors (TEFs) of PCB Congeners in Birds Relative to TCDD

Congener Number	WHO[a] Avian TEF
77	0.05
81	0.1
105	0.0001
114	0.0001
118	0.00001
123	0.00001
126	0.1
156	0.0001
157	0.0001
167	0.00001
169	0.001
189	0.00001

a. Van den Berg et al., 1998.

product of the measured concentration and the TEF for each PCB congener measured in a sample (Giesy et al., 1994):

$$\text{TCDD-eq} = \sum ([\text{congener}]_i \times \text{TEF}_i) \ .$$

The TCDD-eq of environmental samples can also be determined experimentally using the H4IIE rat hepatoma system (e.g., Giesy et al., 1994) or the avian embryo hepatocyte system (e.g., Kennedy et al., 1996a,b). These in vitro systems generally require administration of small quantities of a chemical extract of an environmental sample to the bioassay system, with subsequent measurement of the P450 induction response. The response is then compared to the TCDD-induced response measured using the same method. Because of the apparent differences between mammals and birds in relative congener potency, a TCDD-eq measured using an avian bioassay is considered more relevant to the evaluation of the toxicity to birds than a TCDD-eq measured using a mammalian bioassay (U.S. EPA, 1998a).

There are two limitations to the TCDD-eq approach:

- ***Calculated values assume additive toxicity.*** An implicit assumption in the calculation of a TCDD-eq is that the contribution of individual congeners to the toxicity of a mixture is simply additive. However, both synergistic (more than additive toxicity) and antagonistic (less than additive) responses to PCBs and other contaminants have been reported (Petersen et al., 1993; Van den Berg et al., 1994). For example, Lorenzen et al. (1997) concluded that common terns may be more susceptible to CYP1A inducing effects of complex mixtures of dioxin-like contaminants than indicated by their response to individual contaminants. Synergistic and antagonistic interactions are not incorporated into the calculation of TCDD-eq because of uncertainty in the type and magnitude of contaminant interactions. There is some consensus (U.S. EPA, 1998a) that nonadditive effects may be relatively minor (i.e., toxicity of dioxin-like PCBs is approximately additive).

- ***Only dioxin-like toxicity is considered.*** The TCDD-eq approach generally accounts for only dioxin-like toxicity, whether determined directly from an in vitro bioassay system (e.g., EROD induction) or calculated using measured analyte concentrations and TEFs (derived from in vitro responses or acute in ovo exposures). The endpoints and modes of action used in determining TCDD-eq do not typically incorporate neurotoxicity, PB-like effects, endocrine disruption, or long-term responses such as cancer.

Despite these limitations, the TCDD-eq approach provides a generally accepted method for assessing the toxicity of mixtures of PCBs and other contaminants (U.S. EPA, 1998b).

3.5 PCB CONCENTRATIONS CAUSING TOXICITY TO BIRDS

This section summarizes literature studies on the concentrations of PCBs shown to cause adverse effects to birds. First, the sensitivity of avian life stages and species is discussed, after which toxicity studies using total PCBs dosed into bird eggs are summarized. Lastly, studies of the toxicity of individual PCB congeners, TCDD, or TCDD-eq are summarized.

3.5.1 Avian Sensitivity to PCBs

Embryos appear to be the life stage most sensitive to PCB toxicity, followed by nestlings, then adults (e.g., Hoffman et al., 1998). Embryotoxicity in birds has been studied primarily by direct injection of PCB congeners or commercial mixtures into the egg (air cell or yolk sac), and subsequent monitoring of embryo mortality and hatching success. Eggs injected after the period of organ development experience substantially less mortality and greater chick growth than eggs injected before completion of organ development (Carlson and Duby, 1973).

The relative sensitivity of the tested bird species to PCBs can be estimated by comparing concentrations of individual congeners that cause the same adverse effects. For example, Table 3-5 lists concentrations of PCB 77 and PCB 126 causing 50% embryo mortality when injected into the eggs of different bird species. The results presented in Table 3-5 show that the chicken is the most sensitive species tested to both PCB 77 and 126, with LD50 concentrations approximately two orders of magnitude less than those for other bird species. The higher sensitivity of chickens to TCDD-like toxicity has been documented in numerous studies (Eisler, 1986).

Table 3-5
PCB Congeners: Relative Species Sensitivity Based on Embryo Mortality

Species	Embryo Mortality (LD50; µg/kg egg)	
	PCB 77	PCB 126
Herring gull	>1,000[a]	—
Black-headed gull	<1,000[a]	—
Common tern	—	104[b]
Double-crested cormorant	—	158[c]
Mallard	>5,000[a]	—
Goldeneye, domestic goose	>1,000[a]	—
Kestrel	680[b]	65[b]
Pheasant; quail	100-1000[a]	—
Turkey	800[a]	—
Bobwhite	—	24[b]
Chicken	8.6[a]	0.4

a. Brunstrom and coworkers (summarized in Barron et al., 1995).
b. Hoffman et al., 1995, 1998.
c. Powell et al., 1997.

Bosveld and Van den Berg (1994) suggested that the general order of sensitivity to the embryotoxic effects of PCBs is chicken > pheasant/turkey > ducks > gulls. Kennedy et al. (1996b) similarly concluded that the general order of sensitivity was chicken > pheasant > turkey ≥ duck ≥ herring gull, based on EROD induction in primary

hepatocyte cultures by a variety of planar PCBs. Brouwer (1991) concluded that herring gulls were insensitive to PCB exposure because of Ah receptor nonresponsiveness. Based on the results of egg injection studies with PCB 126, Hoffman et al. (1998) concluded that species responsiveness to P450 induction was chicken > common tern > American kestrel > bobwhite. Based on EROD induction by PCBs 77, 126, and 169 in primary hepatocyte cultures, common terns appeared to be of similar sensitivity to ducks and herring gulls, and 50 to >1600 times less sensitive than chickens (Lorenzen et al., 1997). However, common tern embryo hepatocytes were only 3.5 to 15 times less sensitive than chicken embryo hepatocytes to contaminant extracts of field collected tern eggs (Lorenzen et al., 1997). Lorenzen et al. (1997) suggested that hepatocyte cultures from common tern chicks indicated that this species may be only 6 to 79 times less sensitive than chickens, and may be more susceptible to CYP1A inducing effects of complex mixtures of dioxin-like contaminants than is indicated by their response to individual contaminants. However, common tern embryo hepatocytes were not sensitive to the commercial PCB mixture Aroclor 1254 (Lorenzen et al., 1997).

In conclusion, a limited number of bird species have been evaluated for their sensitivity to PCBs and dioxin-like toxicity. Early life stages (embryos and hatchlings) appear to more sensitive to PCB toxicity than adult or juvenile birds. Of the tested bird species, chickens are consistently the most sensitive to PCB congeners and mixtures, whereas gulls appear to be the least sensitive. The relative rankings of other tested species appear to vary based on congener (or congener mixture) and toxic endpoint studied. Most of the 87 bird species that obtain their food from the Green Bay aquatic environment have not been tested. It is likely that the sensitivity of these species varies and some may be sensitive to PCBs.

3.5.2 Adverse Effect Concentrations of PCBs

As discussed above, adverse effects of PCBs include adult and embryo mortality, impaired reproductive behavior, deformities, decreased female or male fertility, lower hatching success, impaired egg production, and reductions in population size or reproductive success. These effects have been determined in the following types of studies:

- ***Egg injection experiments.*** These studies typically use graded doses of PCBs injected into the yolk sac, air cell, or albumen of the eggs. Measurement endpoints may include embryo mortality, malformations, hatching success, and chick growth.

- ***Dietary toxicity tests.*** These studies involve administration of PCBs in the diet of the bird or by gavage. Measurement endpoints may include effects on behavior (e.g., courtship or parental attentiveness, avoidance behaviors); disease resistance; various measures of reproductive success (e.g., egg production, fertility, hatching success); and chick mortality and growth. Study duration may range from a few days to many weeks. However, studies

that evaluate the effects of PCB exposure over a complete life cycle of birds have not been conducted.

- ***Field assessments.*** Field studies typically involve determining any differences in reproductive success of wild birds from contaminated sites and birds from selected reference locations. Measurement endpoints may include hatching success, chick mortality and growth, and fledgling success.

Adverse effect concentrations are typically expressed as a median effect concentration (e.g., LC50, the concentration causing 50% mortality in the test population, derived from the relationship between dose and toxic response) or as a toxicity threshold value (derived by a statistical assessment of control and treatment groups, e.g., tested concentration causing a significant increase in mortality). Toxicity thresholds are typically reported as a NOEC (no observed effect concentration) or LOEC (lowest observed effect concentration). Note that NOECs and LOECs are statistically determined, but they do not represent absolute thresholds because they are reflective of the experimental design and the doses used. For example, a LOEC of 10 mg PCB/kg egg may not represent the lowest toxicity threshold for a species because lower PCB concentrations were not tested. Traditionally, NOECs and LOECs have been used by the U.S. EPA and others to derive thresholds for chronic toxicity to protect sensitive species.

Total PCB Concentrations in Eggs Causing Toxicity

Table 3-6 presents NOECs and LOECs for total PCB concentrations in bird eggs. LOEC values are presented for the most sensitive reproductive effect measured. Excluding chickens, NOECs range upward from 1.3 mg total PCBs/kg egg (wet weight), and LOECs range upward from about 5-10 mg/kg. It should be noted that there are, apparently, large differences between species in their sensitivities to PCBs. For example, the mallard LOEC reported in Table 3-6 exceeds that of the most sensitive species, chicken, by a factor of more than 50. However, many of the study results listed in Table 3-6 may be confounded by the fact that they are based on field studies in which parameters other than PCBs (e.g., other contaminants, hatching and rearing conditions) could not be controlled.

PCB Congeners and TCDD-eq Concentrations in Eggs Causing Toxicity

Table 3-7 presents LD50, NOEC, and LOEC values for individual PCB congeners, TCDD, and TCDD-eq reported in the literature. The data presented in Table 3-7 indicate that, excluding chickens, toxicity (NOECs, LOECs) for most of the tested bird species occurs at TCDD or TCDD-eq concentrations in the range of approximately 0.2 µg/kg egg (wet weight) to 10 µg/kg. Estimated LD50s for TCDD-eq are also consistent with this range of values. However, it should be noted that because LD50s are concentrations causing effects to 50% of the test organisms; they are *not* effects *thresholds*. Effects thresholds typically will be lower.

Table 3-6
Total PCB No Observed and Lowest Observed Adverse Effect Concentrations (NOEC/LOEC) in Bird Eggs

Species	PCB	NOEC (mg/kg ww)	LOEC (mg/kg ww)	Adverse Effect	Laboratory (L) or Field (F) Study	Reference
Chicken	Total PCB	0.36	2.5	H	L	Scott, 1977
	Total PCB	0.95	1.5	H	L	Britton and Huston, 1973
	Total PCB	<5	5	P, F	L	Platonow and Reinhart, 1973
	Total PCB	—	4	D, H	L	Tumasonis et al., 1973
	A1242	0.67	6.7	G	L	Gould et al., 1997
	A1254	0.67	6.7	G	L	Gould et al., 1997
Tree swallow	Total PCB	—	5-7	B	F	McCarty and Secord, 1999
Common tern	Total PCB	7	8	S	F	Bosveld and Van den Berg, 1994
	Total PCB	4.8	[a]10	D, H	L	Hoffman et al., 1993
	Total PCB	5.2-5.6	7	H	F	Becker et al., 1993
Bald eagle	Total PCB	—	4	S	F	Ludwig et al., 1993
	Total PCB	1.3	7.2	S	F	Wiemeyer et al., 1984
	Total PCB	—	13	S	F	Bosveld and Van den Berg, 1994
Ringed turtle dove	A1254	—	16	H	L	Peakall and Peakall, 1973
Forster's tern	Total PCB	4.5	22.2	H	F/L	Kubiak et al., 1989
	Total PCB	7	[a] 19	S	F	Bosveld and Van den Berg, 1994
Caspian tern	Total PCB	—	4.2	S	F	Yamashita et al., 1993
Mallard	A1242	—	105	T	F	Haseltine and Prouty, 1980

a. Based on no apparent adverse effects in field population.

For adverse effect, A = adult mortality; B = reproductive behavior; D = deformities; F = female fertility; G = chick growth; H = hatching success; M = male fertility; P = egg production; S = population size or reproductive success; T = egg shell thinning. Data are organized by the general rank order of sensitivity (most to least sensitive species based on reported NOECs and LOECs).

Table 3-7
No Observed and Lowest Observed Adverse Effect Concentrations (NOEC/LOEC) and Median Lethal Concentrations (LD50s) for PCB Congeners, TCDD, and TCDD-eq (µg/kg egg wet weight) in Birds

Species	Toxicant	Measurement	Reported µg/kg egg (ww)	TCDD-eq[a] Toxicity Value	TCDD-eq[a] µg/kg egg (ww)	Adverse Effect	Laboratory (L) or Field (F) Study	Reference
Chicken	TCDD	NOEC	0.1	NOEC	0.1	H	L	Janz and Bellward, 1996
			0.2	NOEC	0.2	I	L	Peden-Adams et al., 1998
		LD50	0.15	LD50	0.15	H	L	Powell et al., 1996
	PCB 77	LD50	8.6	LD50	0.43	H	L	Brunstrom and Andersson, 1988
	PCB 105	LD50	2200	LD50	0.22	H	L	Brunstrom, 1990
	PCB 118	LD50	8000	LD50	0.08	H	L	Brunstrom, 1989
	PCB 126	LD50	3.2	LD50	0.32	H	L	Brunstrom and Andersson, 1988
		LD50	2.3	LD50	0.23	H	L	Powell et al., 1996
		LD50	0.4	LD50	0.04	H	L	Hoffman et al., 1995
	PCB 156	LD50	1500	LD50	0.15	H	L	Brunstrom, 1990
	PCB 157	LD50	2500	LD50	0.25	H	L	Brunstrom, 1990
	PCB 167	LD50	>4,000	LD50	>0.04	H	L	Brunstrom, 1990
	PCB 169	LD50	170	LD50	0.17	H	L	Brunstrom and Andersson, 1988
Osprey	TCDD-eq	NOEC	0.14	NOEC	0.14	S	F	Woodford et al., 1998
Bald eagle	TCDD-eq	NOEC	0.2	NOEC	0.2	S	F	Elliott et al., 1996

Table 3-7 (cont.)
No Observed and Lowest Observed Adverse Effect Concentrations (NOEC/LOEC) and Median Lethal Concentrations (LD50s) for PCB Congeners, TCDD, and TCDD-eq (µg/kg egg wet weight) in Birds

Species	Toxicant	Measurement	Reported µg/kg egg (ww)	TCDD-eq[a]		Adverse Effect	Laboratory (L) or Field (F) Study	Reference
				Toxicity Value	µg/kg egg (ww)			
Bobwhite	PCB 126	LD50	24	LD50	2.4	H	L	Hoffman et al., 1995
Caspian tern	TCDD-eq	NOEC	0.75	NOEC	0.75	H	F	Ludwig et al., 1993
Domestic pigeon	TCDD	LOEC	3	LOEC	3	G, H	L	Janz and Bellward, 1996
Eastern bluebird	TCDD	NOEC	1	NOEC	1	B	F	Thiel et al., 1988
		LOEC	10	LOEC	10	B	F	
Common tern	PCB 126	LD50	104	LD50	10.4	H	L	Hoffman et al., 1998
	TCDD-eq	NOEC	<4	NOEC	<1 (assuming 25% lipid)	H	L	Bosveld and Van den Berg, 1994
Double-crested cormorant	PCB 126	LD50	158	LD50	16	H	L	Powell et al., 1997
	TCDD	LD50	4	LD50	4	H	L	Powell et al., 1997
	TCDD-eq	LD50	~0.55	LD50	0.55	H	F	Tillitt et al., 1992

Table 3-7 (cont.)
No Observed and Lowest Observed Adverse Effect Concentrations (NOEC/LOEC) and Median Lethal Concentrations (LD50s) for PCB Congeners, TCDD, and TCDD-eq (µg/kg egg wet weight) in Birds

Species	Toxicant	Measurement	Reported µg/kg egg (ww)	TCDD-eq[a]		Adverse Effect	Laboratory (L) or Field (F) Study	Reference
				Toxicity Value	µg/kg egg (ww)			
Forster's tern	TCDD-eq	NOEC	2.2	NOEC	2.2	H	L/F	Kubiak et al., 1989
Great blue heron	TCDD	NOEC	2	NOEC	2	H	F	Janz and Bellward, 1996
	TCDD-eq	NOEC	0.02	NOEC	0.02	G, D	F	Hart et al., 1991
		LOEC	0.245	LOEC	0.245			
Ring-necked pheasant	TCDD	NOEC	1 (yolk sac injected)	NOEC	1	H	L	Nosek et al., 1993
		LOEC	1 (albumen injected)	LOEC	1	H	L	
	77	NOEC	100	NOEC	5	H	L	Brunstrom and Reutergårdh, 1986
Wood duck	TCDD-eq	NOEC	≤5	NOEC	≤5	H	F	White and Hoffman, 1995
		LOEC	>20-50	LOEC	>20-50	H	F	
American kestrel	PCB 77	LD50	680	LD50	34	H	L	Hoffman et al., 1998
	PCB 126	LD50	65	LD50	6.5	H	L	
Turkey	PCB 77	LD50	~800	LD50	40	H	L	Brunstrom and Lund, 1988
Black-headed gull	PCB 77	LD50	<1000	LD50	<50	H	L	Brunstrom, 1988

Table 3-7 (cont.)
No Observed and Lowest Observed Adverse Effect Concentrations (NOEC/LOEC) and Median Lethal Concentrations (LD50s) for PCB Congeners, TCDD, and TCDD-eq (µg/kg egg wet weight) in Birds

Species	Toxicant	Measurement	Reported µg/kg egg (ww)	TCDD-eq[a]		Adverse Effect	Laboratory (L) or Field (F) Study	Reference
				Toxicity Value	µg/kg egg (ww)			
Herring gull	PCB 77	LD50	>1000	LD50	1-2	H	L	Brunstrom, 1988
	TCDD-eq	NOEC	1-2	NOEC	1-2	H	F	Ludwig et al., 1993
Domestic goose	PCB 77	LD50	>1000	LD50	>50	H	L	Brunstrom, 1988
Goldeneye	PCB 77	LD50	>1000	LD50	>50	H	L	Brunstrom and Reutergårdh, 1986
Mallard	PCB 77	LD50	>5000	LD50	>250	H	L	Brunstrom, 1988

a. Calculated using WHO TEFs (U.S. EPA, 1998b).

For adverse effect, A= adult mortality; B = reproductive behavior; D = deformities; F = female fertility; G = chick growth; H = hatching success; I = immunological changes; M = male fertility; P = egg production; S = population size or reproductive success; T = egg shell thinning. Data are organized by the general rank order of sensitivity as TCDD-eq toxicity values (most to least sensitive species).

3.6 CONCLUSIONS

The results of this review support the following conclusions.

- ***PCBs cause a number of adverse effects in birds that meet the NRDA definitions of injury.*** PCB-caused adverse changes in viability in birds can include death, disease, behavioral abnormalities, physiological malfunctions, and physical deformities. Increasing PCB exposure results in an increase in the number and severity of effects from EROD induction to embryotoxicity to adult mortality (Figure 3-6).

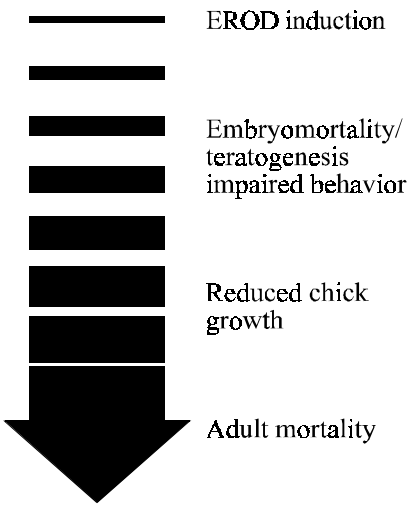


Figure 3-6. Increasing PCB exposure results in an increase in the number and severity of adverse effects in birds.

  Laboratory and field studies have shown that exposure of birds to PCBs causes a suite of toxic effects:

  - At high doses, PCBs may cause death in adult and juvenile birds (summarized by Prestt et al., 1970; Dahlgren et al., 1972b; Barron et al., 1995).
  - At lower exposures, PCBs may cause death in avian embryos (Barron et al., 1995).
  - Sublethal effects of PCBs can include reproductive and developmental toxicity: (1) altered reproductive behavior, (2) reduced fertility and egg production, (3) reduced or delayed chick growth. PCB effects also include subtle neurological effects such as impaired avoidance behavior (Dahlgren et al., 1972b).
  - P450 induction is the most consistently sensitive in vitro measure of PCB exposure, but P450 activity (e.g., EROD) can be inhibited at higher exposure concentrations (Lorenzen et al., 1997).
  - Eggshell thinning does not appear to be caused by PCB exposure (Peakall and Lincer, 1996), except at high dietary concentrations (e.g., 105 mg/kg wet weight; Haseltine and Prouty, 1980).
  - Depending on the dose and exposure scenario, PCBs and related contaminants may act as estrogen or thyroxine agonists or antagonists or may alter circulating hormone levels (McKinney et al., 1985; Gilbertson et al., 1991).

- Field studies have associated PCB exposure in birds with increased EROD, decreased thyroxine in plasma, decreased hepatic retinoid levels, increased relative liver weight, decreased head and femur size in hatchlings, reduced embryo growth, and delayed hatching (Hoffman et al., 1986; Van den Berg et al., 1994; Bosveld et al., 1995).

- ***PCBs in eggs cause toxicity at low parts-per-million concentrations of total PCBs (Table 3-6):***

  - Although there is much variability in species sensitivity, toxicity thresholds for total PCBs in the eggs of sensitive bird species range upward from 5 to 10 mg/kg egg, resulting in reproductive malfunctions, embryo mortality, and embryo deformities.

- ***PCBs in eggs cause toxicity at low, or sub parts per billion, concentrations as TCDD-eq in eggs (Table 3-7):***

  - Toxicity thresholds for TCDD-eq in the eggs (NOECs, LOECs) of many bird species range from 0.2 to 10 µg/kg egg, resulting in reproductive malformations, embryo mortality, and deformities.

In conclusion, PCBs cause multiple adverse effects in bird species, including death, deformities, and reproductive malfunctions. Low mg/kg wet weight concentrations of total PCBs in eggs and low ppb concentrations of TCDD-eq in eggs can cause embryo mortality, malformations, and impaired reproduction.

CHAPTER 4
PCB EXPOSURE IN ASSESSMENT AREA BIRDS

4.1 INTRODUCTION

This chapter presents information on the exposure of assessment area birds to PCBs. Exposure is characterized using data on PCB concentrations measured in the tissues of bird species nesting in the Fox River/Green Bay assessment area. The purpose of this chapter is to determine if assessment area birds have been exposed to PCBs and the likelihood that their exposure levels may have been sufficient to result in injuries. The occurrence of injuries to birds in the assessment area is assessed in the following chapters of this report.

Our approach in this chapter is as follows:

- We compare PCB tissue residues in assessment area birds to those from reference areas to determine whether assessment area birds have been exposed to elevated PCB concentrations.

- We evaluate PCB tissue residues in assessment area birds over time to characterize temporal trends in PCB exposure.

- We compare PCB tissue residues in assessment area birds to ranges of PCB concentrations shown to cause toxicity in laboratory or field studies. These comparisons provide information regarding the likelihood that PCBs have caused adverse effects in assessment area birds.

Figure 4-1 depicts the pathways by which assessment area birds can be exposed to PCBs. Because PCBs accumulate in biota and "biomagnify" in the food chain, the dietary pathway is the primary route by which birds are exposed. Also, of the birds that nest and feed on and near the assessment area, piscivorous species (i.e., those that consume fish) and predatory species (i.e., those that consume other birds) are expected to be most highly exposed to PCBs, since their food items are more highly contaminated with PCBs.

Exposure analysis is a fundamental component of pathway determination. The NRDA regulations indicate that confirmation of biological pathways can be characterized by direct measurement of the hazardous substance in tissues of exposed organisms [43 CFR §11.63(f)(4)(i)]. Thus, using measurements of PCBs accumulated in bird tissue is the most direct method of confirming exposure, as it takes into account such factors as contaminant bioavailability, foraging areas,

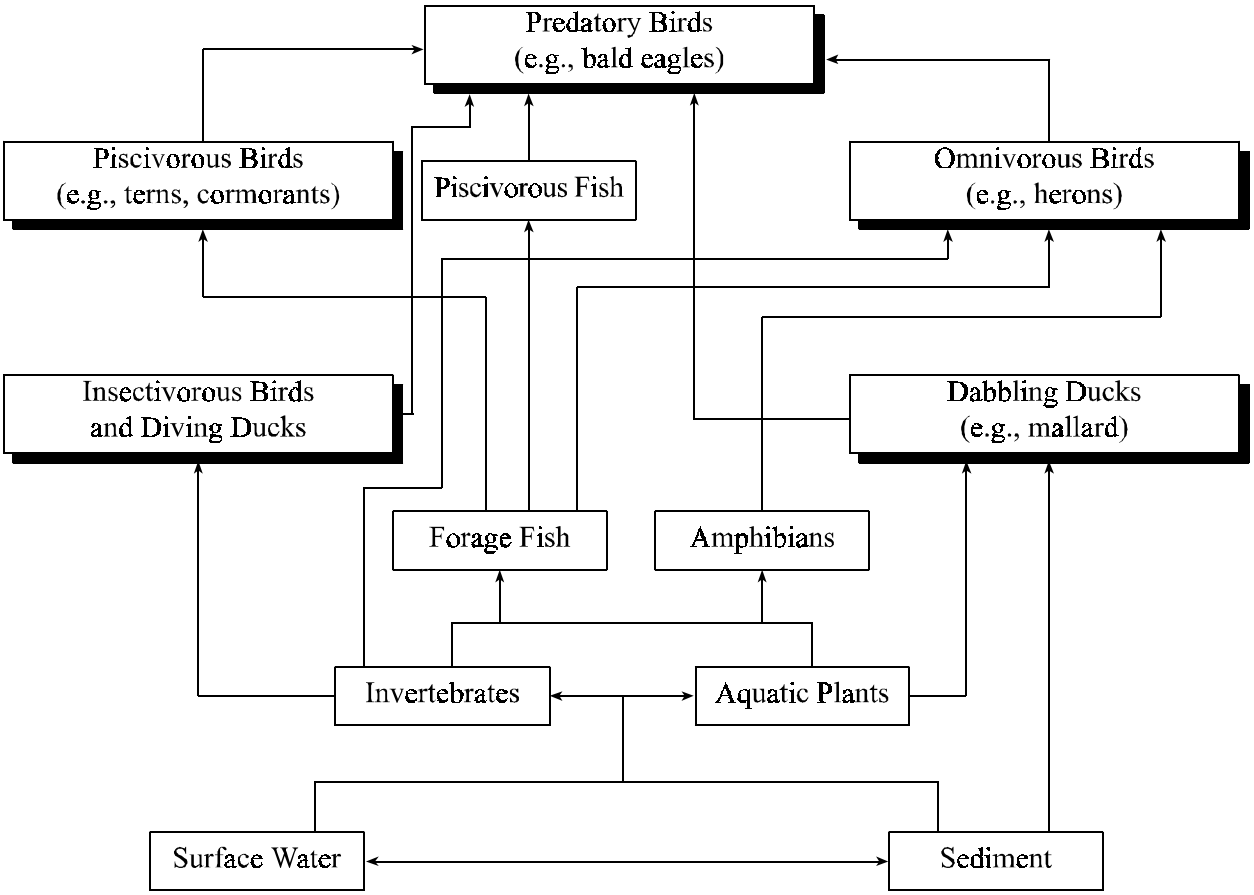


Figure 4-1. Generalized PCB exposure pathways for assessment area birds.

and contamination of prey items. More detailed pathway evaluations for each bird species assessed are presented in Chapter 5.[1]

Numerous studies have been conducted on PCB concentrations in Green Bay bird tissues. Figure 4-2 shows the locations in the assessment area at which bird tissues have been collected for PCB analysis, and Table 4-1 lists the species and tissues (egg, adult, or chick) that have been measured. Table 4-1 shows that more PCB concentration data are available for eggs than for adult or chick tissue, and since egg PCB data can be used to assess the potential for embryo toxicity (a sensitive PCB toxic endpoint), this chapter focuses on PCB concentration data in eggs.

1. The pathways by which PCBs are transported from Fox River paper company facility sources through the Fox River, into Green Bay, and into assessment area fish tissue are described in a separate NRDA report.

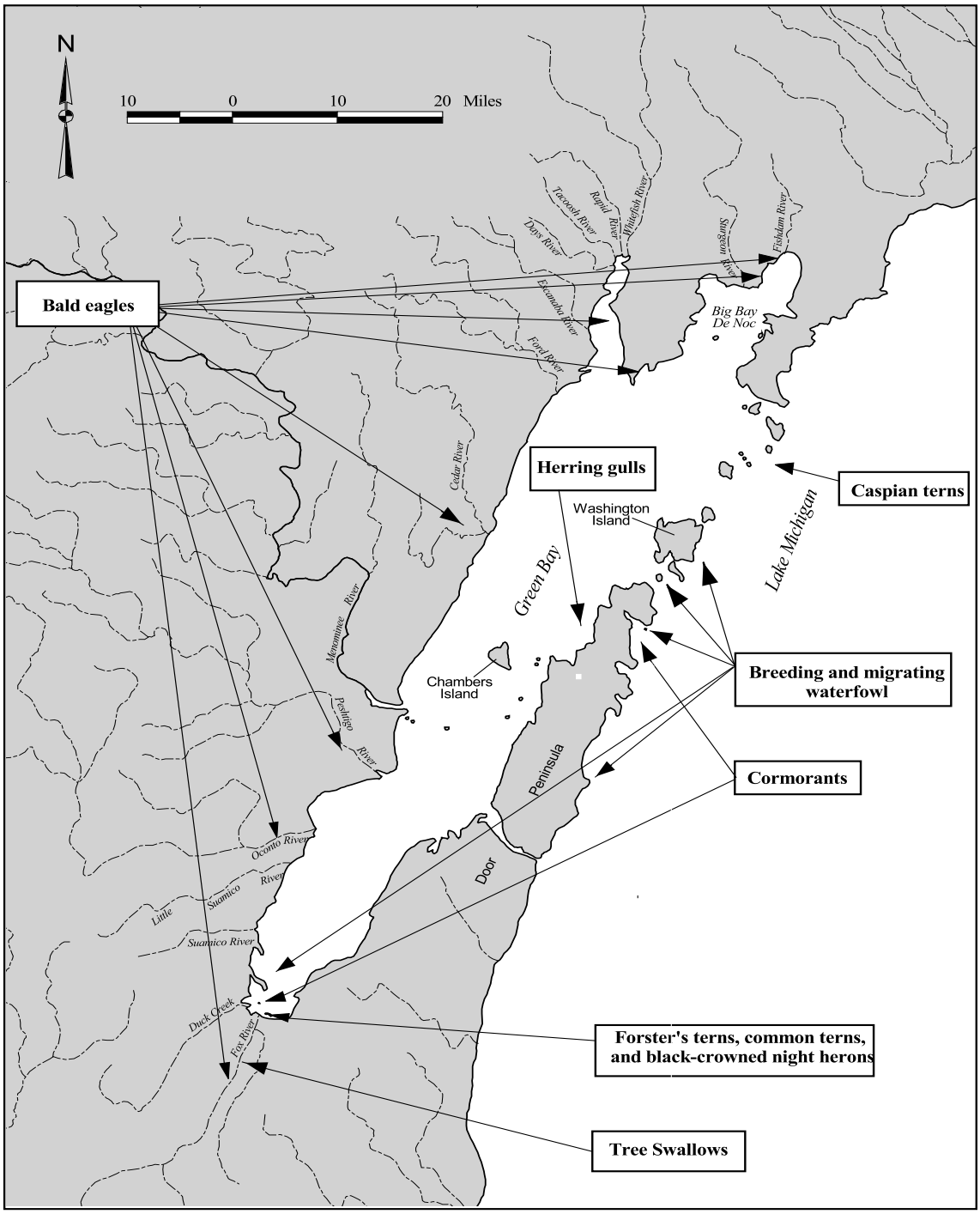


Figure 4-2. Selected locations at which bird tissues have been collected for PCB analysis.

Table 4-1
Green Bay Assessment Area Birds and Their Life Stages in Which PCBs Have Been Measured

	Eggs/Embryos	Adults	Chicks
Double crested cormorant	✓[a]	✓[b]	✓[c]
Black-crowned night heron	✓[d]		✓[d]
Green heron	✓[e]		
Canada goose	✓[b]		
Mallard	✓[b]	✓[b]	
Pintail		✓[b]	
Gadwall	✓[b]	✓[b]	
Lesser scaup		✓[q]	
Greater scaup		✓[q]	
Common goldeneye		✓[q]	
Ruddy duck		✓[q]	
Bufflehead		✓[q]	
White-winged scoter		✓[q]	
Canvasback		✓[q]	
Common merganser		✓[f]	
Red-breasted merganser	✓[g]	✓[q]	
Bald eagle	✓[h]		✓[i]
Herring gull	✓[j]		
Ring-billed gull	✓[e]		
Little gull	✓[e]		
Common tern	✓[e]		✓[k]
Forster's tern	✓[l]		✓[m]
Caspian tern	✓[n]	✓[o]	
Black tern	✓[e]		
Tree swallow	✓[p]		✓[p]
Red-winged blackbird	✓[k]		
Yellow-headed blackbird	✓[r]		✓[r]
Marsh wren	✓[s]		

a. Heinz et al., 1985; Tillitt et al., 1992; Williams et al., 1995a; Larson et al., 1996; T. Custer, USGS, pers. comm., 1998; Custer et al., in press.
b. USFWS, 1993.
c. Custer et al., 1997.
d. Custer and Custer, 1995; Rattner et al., 1993.
e. Heinz et al., 1985; Ankley et al., 1993; USFWS, 1993; Hoffman et al., 1993; Stratus Consulting unpublished data.
f. Amundson, undated.
g. White and Cromartie, 1977; Haseltine et al., 1981; USFWS, 1993; Williams et al., 1995b; Heinz et al., 1994.
h. Dykstra and Meyer, 1996.
i. W. Bowerman, Lake Superior State University, pers. comm., June 1998.
j. Bishop et al., 1992; Pekarik et al., 1998.
k. Ankley et al., 1993.
l. Heinz et al., 1985; Kubiak et al., 1989; USFWS, 1993; Harris et al., 1993; Jones et al., 1993; Stratus Consulting unpublished data.
m. Harris et al., 1993.
n. Yamashita et al., 1993.
o. Mora et al., 1993.
p. C. Custer et al., 1998.
q. USFWS, unpublished data.
r. Rattray, 1997.
s. B. Harris, University of Wisconsin, Green Bay, pers. comm., 1999.

4.2 COMPARISON OF ASSESSMENT AREA BIRD PCB CONCENTRATIONS TO REFERENCE AREA CONCENTRATIONS

In many of the studies of PCB concentrations in assessment area birds, the investigators also collected and analyzed eggs from reference areas. Within each study, similar collection, handling, storage, preparation, and analysis methods were used for both the assessment area and reference areas. Therefore, these studies can be used for direct comparison of PCB concentrations between the assessment area and reference areas.

Table 4-2 presents a comparison of bird tissue PCB concentrations in assessment and reference areas from studies in which both were measured. The table shows that for all species and studies where a statistical comparison was made between PCB concentrations in assessment and reference area tissues, concentrations were significantly greater in tissues from the assessment area. Mean assessment area PCB concentrations were up to approximately eight times greater than reference area concentrations for species such as double-crested cormorant, black-crowned night heron, and bald eagle. PCB concentrations in other species were two to five times greater in the assessment area than in reference areas.

Many of the studies listed in Table 4-2 used different reference areas for comparison. Reference areas used in studies of Caspian terns, common terns, and herring gulls are in northern Great Lakes areas where no PCB point sources such as those of the Fox River paper companies are present. Reference areas used in studies of Forster's terns, mallards, bald eagles, tree swallows, and red-winged blackbirds represent PCB exposure in inland Wisconsin. Reference areas used in studies of black-crowned night herons and double-crested cormorants are distant from the Great Lakes, reflecting lower PCB exposure in areas not influenced by Great Lakes PCB releases. Regardless of the reference area used, Table 4-2 demonstrates that PCB concentrations in birds from the Fox River/Green Bay assessment area exceed those in the reference areas.

4.3 TEMPORAL TRENDS IN PCB EXPOSURE

Because releases of PCBs into the Fox River/Green Bay environment have not been constant over time, exposures of assessment area birds to PCBs have also varied over time. Characterizing temporal trends in PCB exposure of assessment area birds helps define the time span over which injuries have occurred. The Canadian Wildlife Service has collected herring gull eggs from Big Sister Island in Green Bay (along the eastern shore; see Figure 2-4) almost every year since 1972 as part of regular monitoring of contaminant concentrations in the Great Lakes (Bishop et al., 1992; Pettit et al., 1994; Hughes et al., 1998; Pekarik et al., 1998). This dataset is the most complete dataset available with which to evaluate temporal trends in Green Bay bird PCB exposure.

Figure 4-3 plots the PCB concentrations measured in Big Sister Island herring gull eggs from 1971 through 1996 (Hughes et al., 1998). Also included in the plot is an estimate of PCB

Table 4-2
Comparison of PCB Concentrations in Bird Tissues from the Assessment and Reference Areas

Species	Tissue	Mean PCB Concentration (mg/kg wet weight, wet weight)		Statistically Higher in Assessment Area?[a]	Reference Area	Source
		Assessment Area	Reference Area			
Double-crested cormorant	Egg	7.8	1.0	Yes	Lake Winnipegosis, Manitoba	Larson et al., 1966
	Egg	5.3-14.8	0.8	Yes	Lake Winnipegosis, Manitoba	Tillitt et al., 1992
Black-crowned night heron	Whole-body	9.3	1.1	Yes	Chincoteague NWR, Virginia	Rattner et al., 1993
Herring gull	Egg	104.2	51.4	Yes	Lake Superior	Bishop et al., 1992
Forster's tern	Egg	19.2	4.6	Yes	Lake Poygan, WI	Kubiak et al., 1989
Common tern	Egg	10.0	4.0-4.7	Yes	N. Lake Michigan	Hoffman et al., 1993
Caspian tern	Egg	36.2	18.5-30.9	Yes	N. Lake Huron	Struger and Weseloh, 1985
	Egg	10.8	5.6-10.0	NA	N. Lake Huron	Yamashita et al., 1993
	Egg	15.8	8.6-14.5	NA	N. Lake Huron	Ewins et al., 1994
	Plasma	3.5	1.0-1.4	Yes	N. Lake Huron	Mora et al., 1993
Mallard	Muscle	0.43	0.19	Yes	Inland Wisconsin	Amundson, undated
Bald eagle	Egg	35	4.3	Yes	Inland Wisconsin	Dykstra and Meyer, 1996
Tree swallow	Egg	3.2	0.3	Yes	Lake Poygan, WI	Custer et al., 1998
Red-winged blackbird	Egg	1.1	0.3	NA	Inland Wisconsin	Ankley et al., 1993

a. Statistical significance as reported by study authors. In all cases significance was determined at $p < 0.05$.

NA = study authors did not conduct statistical tests, and raw data are not available to use in conducting tests.

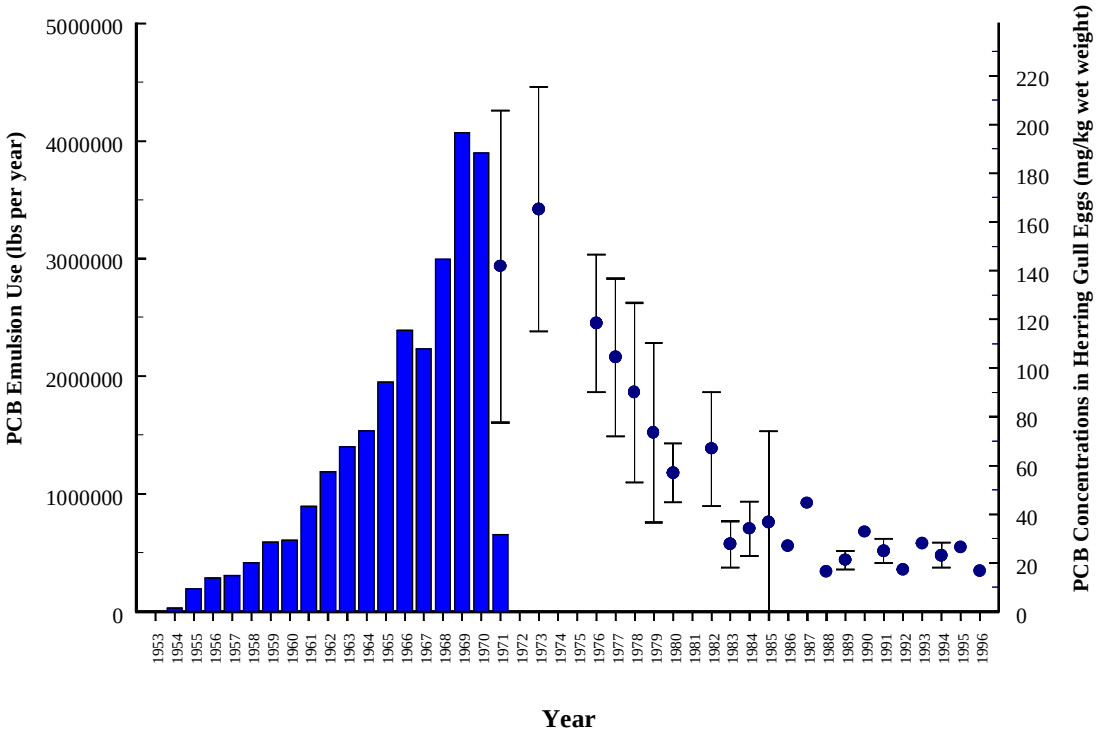


Figure 4-3. Estimated PCB emulsion use at Appleton Paper (solid bars) and mean PCB concentrations in herring gull eggs (plus or minus 1 SD) from Big Sister Island, Green Bay. PCB emulsion use is expected to closely match the temporal pattern of direct PCB discharges from paper companies into the Fox River (G. Amendola, Amendola Engineering, Inc., personal communication, 1999). PCB emulsion use data from G. Amendola (personal communication, 1999). Herring gull data from Hughes et al. (1998).

emulsion use by Appleton Paper, which is on the Fox River. The timeframe for direct PCB discharges into the Fox River is expected to closely match the timeframe of PCB emulsion use by Appleton Paper (G. Amendola, Amendola Engineering, Inc., personal communication, 1999). Direct PCB releases increased from 1954 to 1969 and dropped dramatically from 1970 to 1971, when PCB use in carbonless copy paper was discontinued (G. Amendola, Amendola Engineering, Inc., personal communication, 1999).[2] PCB concentrations in Big Sister Island herring gull eggs were highest when they were first measured in the early 1970s, with mean concentrations of approximately 170 mg/kg wet weight. Concentrations dropped from the early 1970s through the mid-1980s, reaching a mean concentration of approximately 30 mg/kg wet weight. Since the mid-

2. Direct releases into the Fox River did continue after 1971, although the estimated mass of PCBs released was much less than that released before 1971 (Wisconsin DNR, 1998a). In addition, re-releases of PCBs from contaminated river and bay sediments continue (DePinto et al., 1994).

1980s, PCB concentrations have stabilized or are declining only very slightly, with concentrations varying within the approximate range of 15 to 40 mg/kg wet weight.

Figure 4-4 shows the Big Sister Island herring gull egg PCB data broken into two time segments: 1971 to 1982, and 1983 to 1996. A comparison of the two plots shows that before 1983, PCB concentrations were clearly declining. Since approximately 1983, the decline has reached a plateau, although there is an almost significant negative trend ($r = 0.5$, $p = 0.07$).

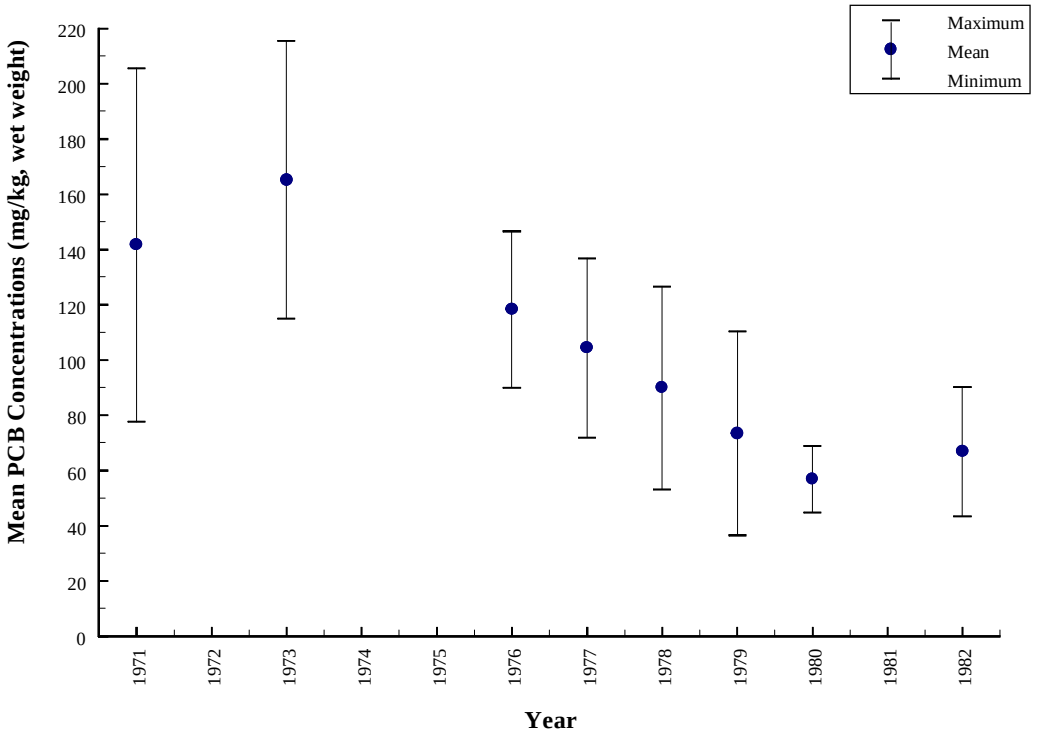
Comparison of the PCB loadings to the herring gull data (Figure 4-3) indicates that the decline in herring gull PCB concentrations followed the sharp reduction in paper company releases in 1971. Following the decline in paper company direct releases, herring gull PCB concentrations decreased, although not as rapidly. Since PCBs do not degrade readily in the environment (Erickson, 1997), they remain in the system for many years following initial release. This is reflected in the fact that the decline in herring gull PCB concentrations lagged behind the drop in loadings to the system, and that since the initial decline, concentrations have stabilized or are declining at a much lower rate. Figures 4-3 and 4-4 indicate that it took approximately 15 years for PCB concentrations in Green Bay herring gulls to respond fully to the sharp reductions in direct PCB loadings. Following the initial 15-year response, PCB concentrations have now stabilized at levels that reflect a state where direct PCB loadings are much lower than in the past, but PCBs stored in the system continue to result in exposure to biota.

Piscivorous birds in the assessment area, including herring gulls, feed on a variety of forage fish species (Ludwig and Ludwig, undated report a; Ewins et al., 1994). Figures 4-5 and 4-6 show PCB concentrations measured over time in yellow perch and alewife, respectively, in Green Bay. These forage fish data, although not as complete as the herring gull data, show a temporal pattern similar to that observed in Green Bay herring gulls. PCB concentrations were higher in the 1970s and have remained relatively constant or have declined slightly since the mid-1980s. Therefore, PCB exposure and accumulation for other assessment area piscivorous birds is expected to follow the pattern observed for herring gulls: a decline through the 1970s until approximately the early 1980s, and a stabilization of PCB concentrations since then. For example, Figure 4-7 shows such a pattern for red-breasted mergansers in the assessment area.

4.4 COMPARISON TO TOXIC EFFECTS RANGES

In this section, PCB concentrations measured in eggs of Green Bay birds are compared with toxicity threshold values obtained from the literature. The purpose of the comparison is to evaluate whether the PCB concentrations measured in Green Bay bird eggs are at or above concentrations shown to cause adverse effects. Because of uncertainties in applying toxic thresholds obtained from literature studies, such as differences in species studied, mode and timing of PCB dosing, differences in environmental mixtures of PCBs, toxic endpoints examined,

(a)



(b)

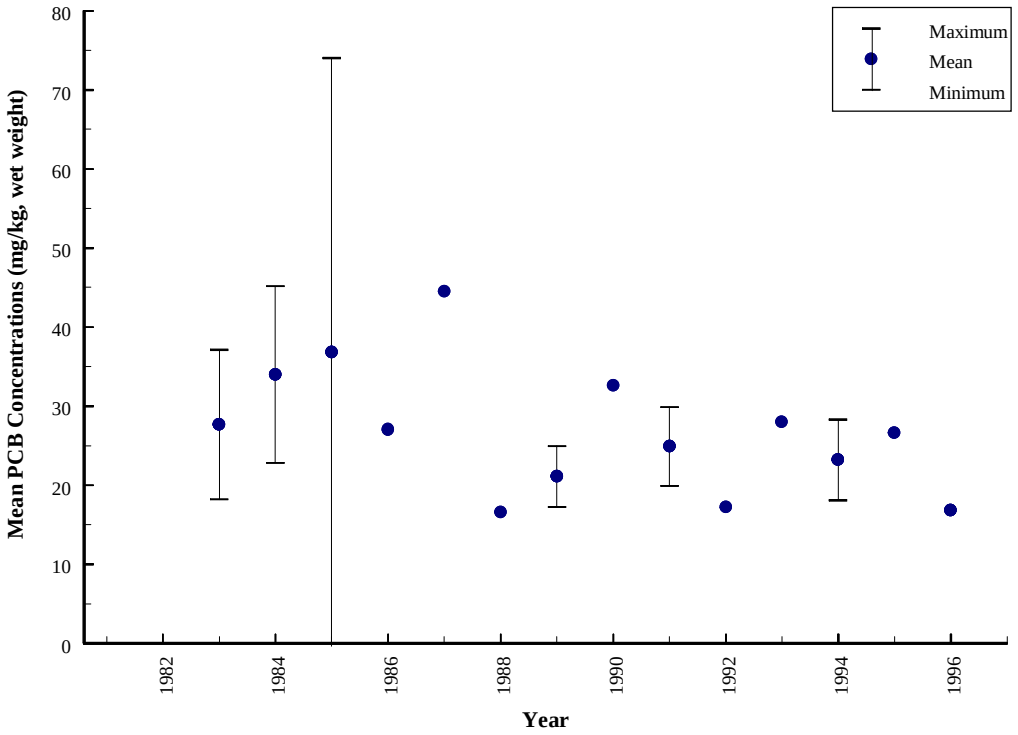


Figure 4-4. Mean and maximum and minimum PCB concentrations in Big Sister Island (Green Bay) herring gull eggs from (a) 1971 to 1982 and (b) 1983 to 1996.

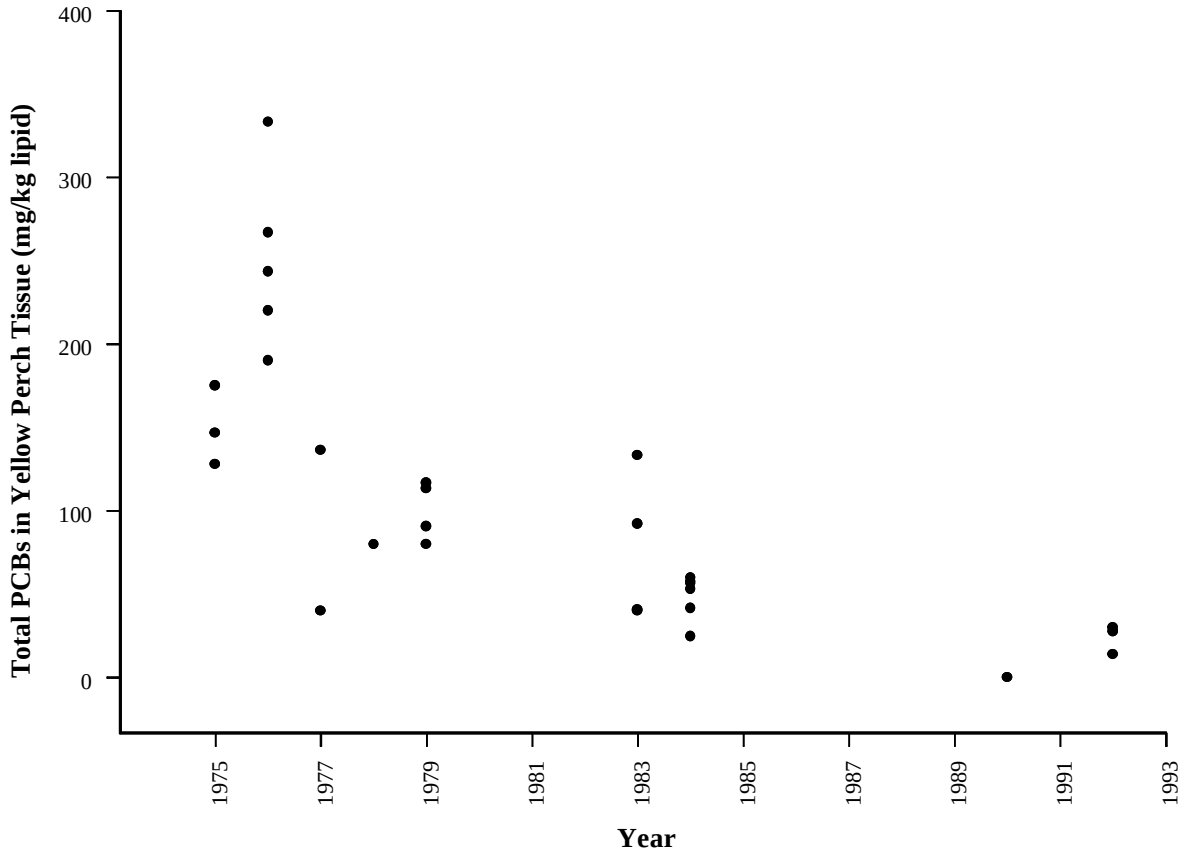


Figure 4-5. Total PCB concentrations measured in Green Bay yellow perch tissue. Each point represents a separate fish sample.
Data sources: Connolly et al., 1992; Wisconsin DNR, 1971-1995.

and the presence or absence of other stressors (e.g., other contaminants, environmental stressors), this comparison is not, in itself, a definitive determination of injury. However, it provides an indication as to whether PCB concentrations in Green Bay birds may be sufficient to cause adverse effects. A detailed evaluation of field studies examining actual adverse effects in Green Bay birds is presented in Chapter 5.

The comparison presented in this section is similar to the hazard quotient approach used in ecological risk assessment (e.g., U.S. EPA, 1998a). Hazard quotients are calculated as the ratio of exposure concentrations to toxicity threshold concentrations. Hazard quotients greater than one mean that contaminant exposure is at levels above toxic thresholds, thereby indicating risk. Although we do not actually calculate hazard quotients in this analysis, our approach is analogous to the hazard quotient approach.

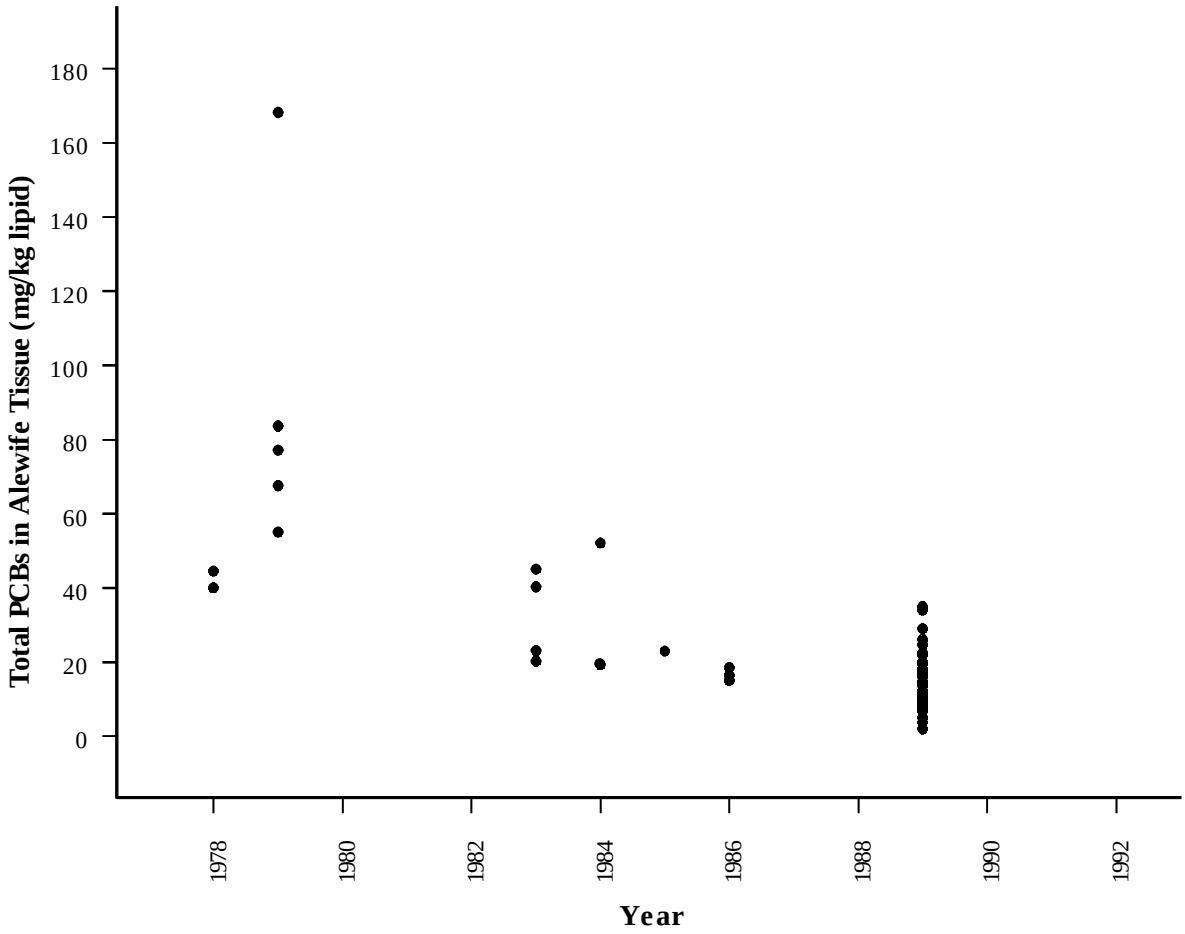


Figure 4-6. Total PCB concentrations measured in Green Bay alewife tissue. Each point represents a separate fish sample.
Data sources: Connolly et al., 1992; Wisconsin DNR, 1971-1995.

Green Bay bird egg PCB exposure data are available for both total PCBs and individual congeners. In the following analysis, total PCB concentrations measured in eggs are compared directly with the results of laboratory and field studies that quantified egg exposure as total PCBs. Measured PCB congener concentrations are converted to TCDD-eq and compared with toxicity studies that expressed exposure as TCDD or TCDD-eq concentration. Because individual PCB congeners can vary greatly in both their potency and relative concentrations in environmental samples, using TCDD-eq accounts for variations in congener potency and concentration that are not considered with total PCBs. WHO avian TEFs, which are TEFs developed by an international group of toxicology experts for use in avian risk assessments, were used to calculate TCDD-eq from congener concentrations (Van den Berg et al., 1998) (Table 3-4). Not all of the PCB congeners that have measurable TCDD-like effects (i.e., have nonzero TEFs) were measured in all of the Green Bay bird egg samples, leading to an underestimation of TCDD-eq. On the other

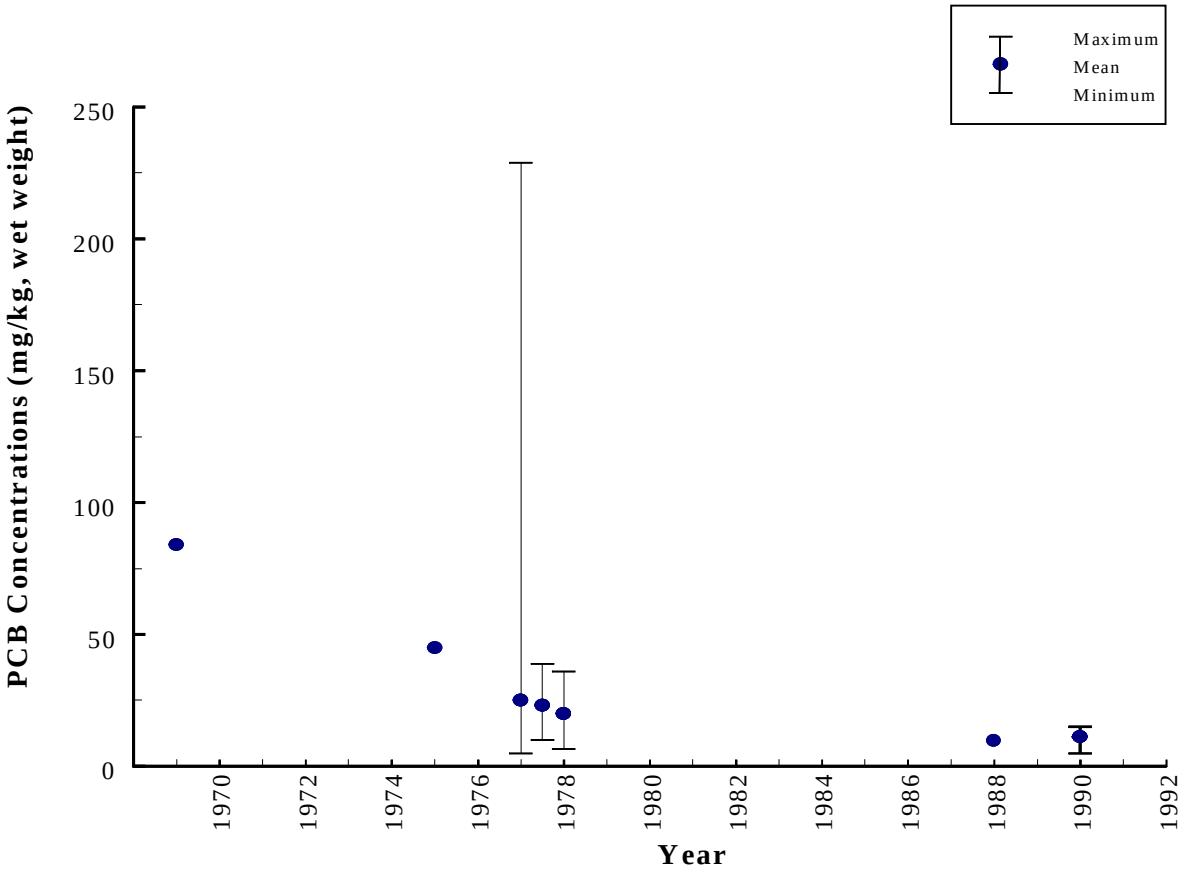


Figure 4-7. Mean PCB concentrations in red-breasted merganser eggs from Green Bay between 1968 and 1990.

Data sources: White and Cromartie, 1977; Haseltine et al., 1981; Heinz et al., 1983; Williams et al., 1995b.

hand, the calculation of TCDD-eq from congener concentrations assumes strict additivity of TCDD-like congener effects and does not take into account possible antagonism (Bosveld, 1995), although effects appear to be close enough to additive to justify the TEF approach in risk assessment (Van den Berg et al., 1998). Finally, only PCB concentrations measured in Green Bay bird eggs since 1983 are included, since the annual survey of Green Bay herring gull eggs and data on PCBs in Green Bay red-breasted mergansers (Figures 4-3, 4-4, and 4-7) show that PCB concentrations prior to the mid-1980s were still declining. Therefore this analysis underestimates pre-1983 effects.

Based on the values shown in Tables 3-6 and 3-7, egg total PCB concentrations of between approximately 5 and 10 mg/kg egg (wet weight) may result in adverse effects in sensitive wild bird species. This range is used as an overall estimate of the range at which toxic effects may begin to be seen in wild birds. Below 5 mg/kg wet weight total PCBs in eggs, effects appear to be unlikely. At and above this range, adverse effects are likely for at least sensitive wild bird species.

The adverse effects to birds documented as occurring at and above this range include reduced reproductive success, deformities, and behavioral abnormalities. It should be noted that concentrations of less than 5 mg/kg wet weight total PCBs in eggs have been shown to cause reduced hatching success in the domestic chicken. However, because the chicken is more sensitive to PCB toxicity than any wild bird species tested to date (Bosveld, 1995), it was not included in the derivation of the toxic threshold range so that the threshold is more relevant for bird species of concern in Green Bay.

From the information presented in Table 3-7, 200-10,000 pg TCDD-eq/kg egg (wet weight) is a representative toxic effects range. As with total PCBs, this range represents concentrations below which toxicity appears to be unlikely, and within and above which adverse effects to many species have been documented. Again, this range does not incorporate data for the chicken, which is much more sensitive than any wild bird species tested to date.

Data on total PCB exposure in assessment area bird eggs are shown in Figure 4-7 for red-breasted mergansers, Figure 4-8 for double-crested cormorants, Figure 4-9 for common terns, Figure 4-10 for Forster's terns, Figure 4-11 for Caspian terns, and Figure 4-12 for bald eagles. Data shown are for eggs collected from Green Bay, ranging from the inner bay (common and Forster's terns) to the outer bay (Caspian terns and bald eagles). In all the figures the mean total PCB concentrations reported are presented, and in some cases the minimum and maximum values were also available and are plotted.

Figures 4-7 through 4-12 show that for all six of these species, *average* total PCB concentrations measured in eggs after 1983 are within or above the 5-10 mg/kg range. These data indicate that the total PCB concentrations measured in eggs of red-breasted mergansers, double-crested cormorants, common terns, Forster's terns, Caspian terns, and bald eagles within the assessment area are within or, in some cases, exceed the range where adverse reproductive effects have been reported in sensitive species.

Figure 4-13 shows TCDD-eq concentrations calculated from PCB congener measurements made in assessment area bird eggs. PCB congener measurements (including coplanar congeners) are available for red-breasted mergansers, double-crested cormorants, common terns, and Forster's terns. PCB congener data were converted to TCDD-eq using both the WHO Avian TEFs (Van den Berg et al., 1998). For each species, all assessment area congener data since 1983 are combined. The sources of the congener data used in Figure 4-13 are listed in Table 4-3.

Figure 4-13 shows that the average TCDD-eq concentrations in eggs of all of the species are within or exceed the 200-10,000 pg TCDD-eq/kg egg (wet weight) toxic effects range for sensitive species derived from Table 3-7. These data are consistent with the total PCB data, and indicate that the mixture of PCB congeners in assessment area bird eggs is of sufficient potency and concentration to potentially cause adverse effects.

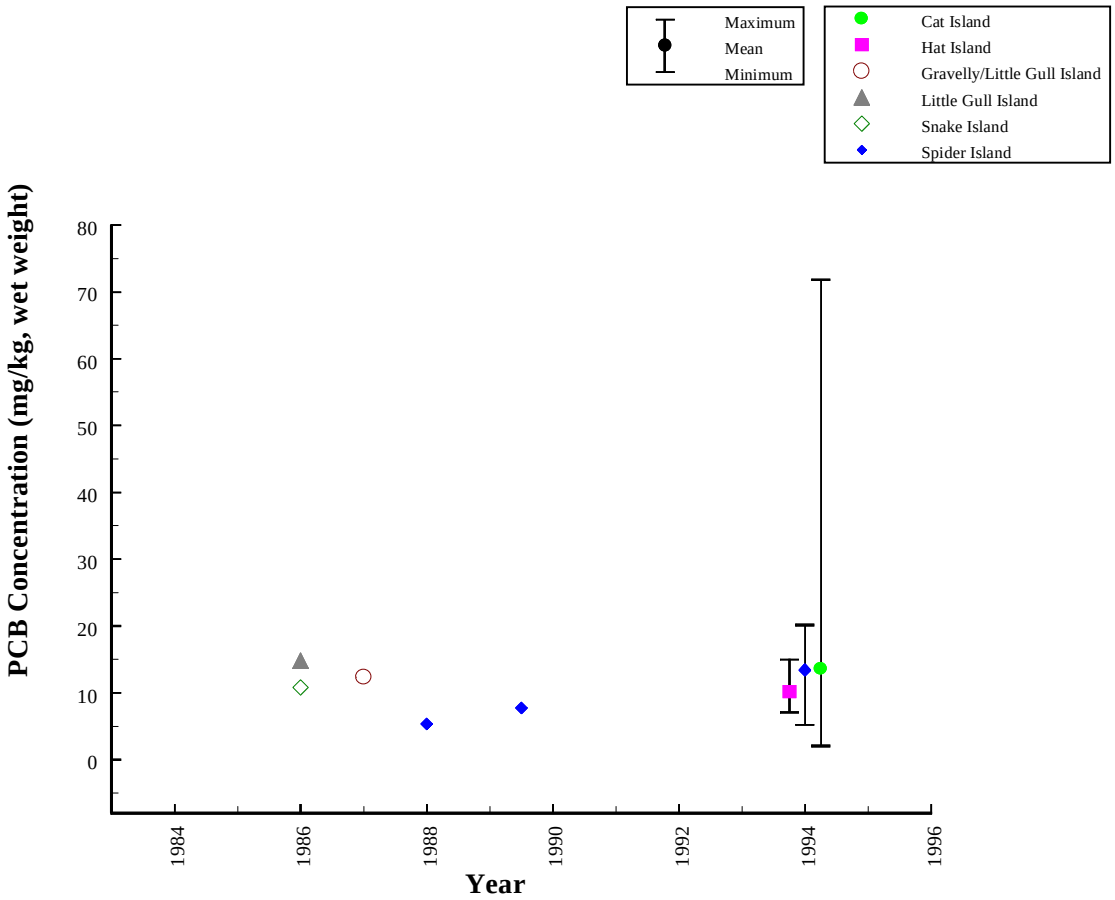


Figure 4-8. Total PCB concentrations measured in assessment area double-crested cormorant eggs, 1983-1996. See Table 4-1 for data sources.

4.5 CONCLUSIONS

The information presented and evaluated in this chapter supports the following conclusions:

- Numerous species of birds throughout the assessment area are exposed to PCBs. The primary route of exposure for most assessment area bird species is dietary.

- PCB concentrations measured in the tissues of assessment area bird species are statistically significantly greater than concentrations measured in reference areas. Every species tested has been found to have greater concentrations in the assessment area, including double-crested cormorant, black-crowned night heron, herring gull, Forster's tern, common tern, Caspian tern, mallard, bald eagle, tree swallow, and red-winged blackbird.

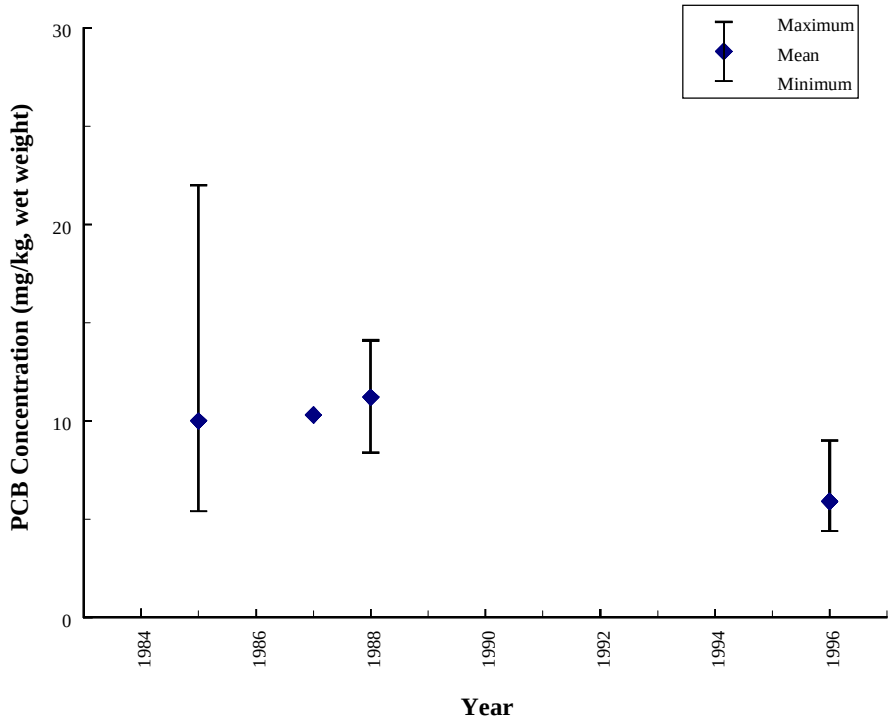


Figure 4-9. Total PCB concentrations measured in assessment area common tern eggs, 1983-1996. See Table 4-1 for data sources.

- PCB exposure of assessment area birds, as measured by PCB accumulation in bird tissue, was greatest in the early 1970s (the first dates for which data are available), declined through the 1970s and through the early 1980s, and has remained relatively stable since then.

- Total PCB concentrations measured in eggs of assessment area red-breasted mergansers, double-crested cormorants, common terns, Forster's terns, Caspian terns, and bald eagles from 1983 to 1996 are within or, in many cases, exceed the range where adverse reproductive effects have been reported in sensitive species.

- TCDD-eq concentrations calculated from PCB congener concentrations measured in assessment area bird eggs are within or exceed a TCDD-based toxicity threshold range. These data indicate that assessment area bird eggs contain a mixture of PCB congeners of sufficient potency and concentration to cause adverse effects.

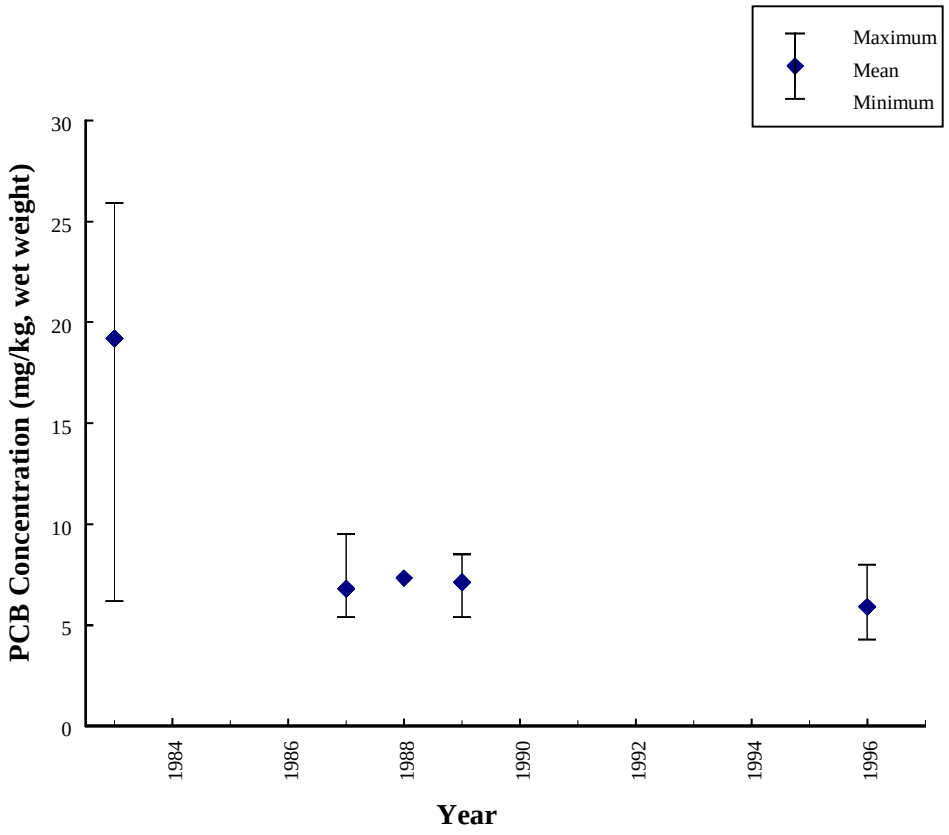


Figure 4-10. Total PCB concentrations measured in assessment area Forster's tern eggs, 1983-1996. See Table 4-1 for data sources.

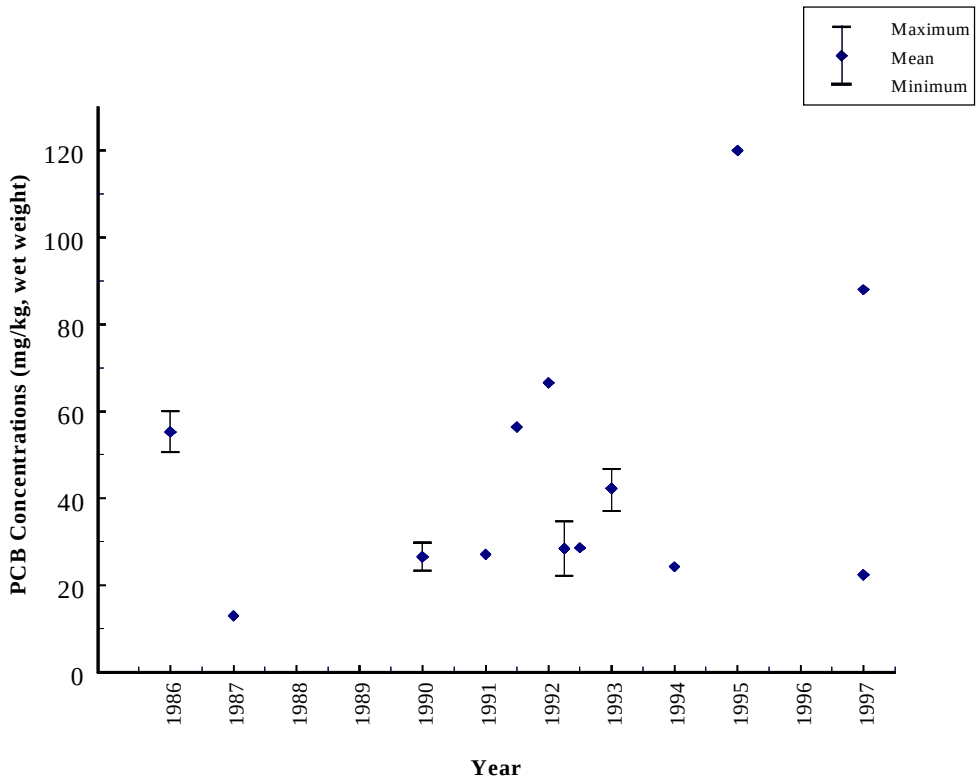


Figure 4-11. Total PCB concentrations measured in assessment area Caspian tern eggs, 1983-1996. See Table 4-1 for data sources.

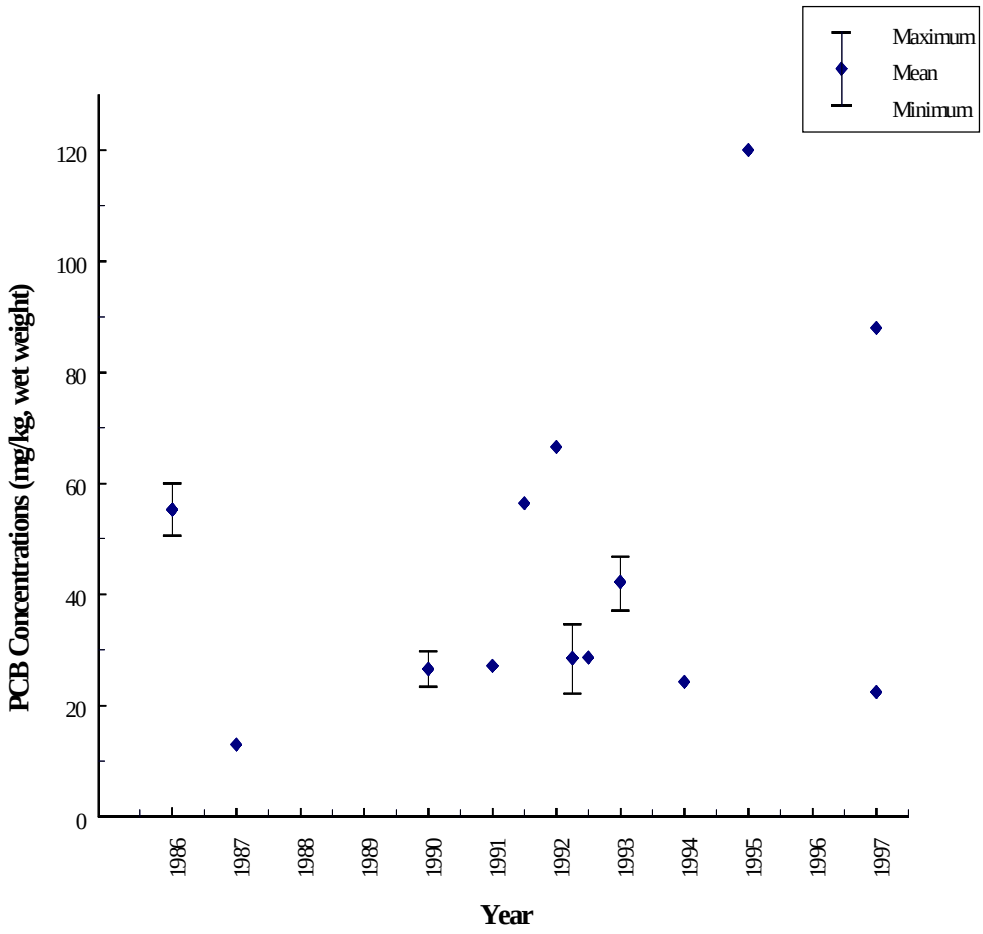


Figure 4-12. Total PCB concentrations measured in assessment area bald eagle eggs, 1986-1997. See Table 4-1 for data sources.

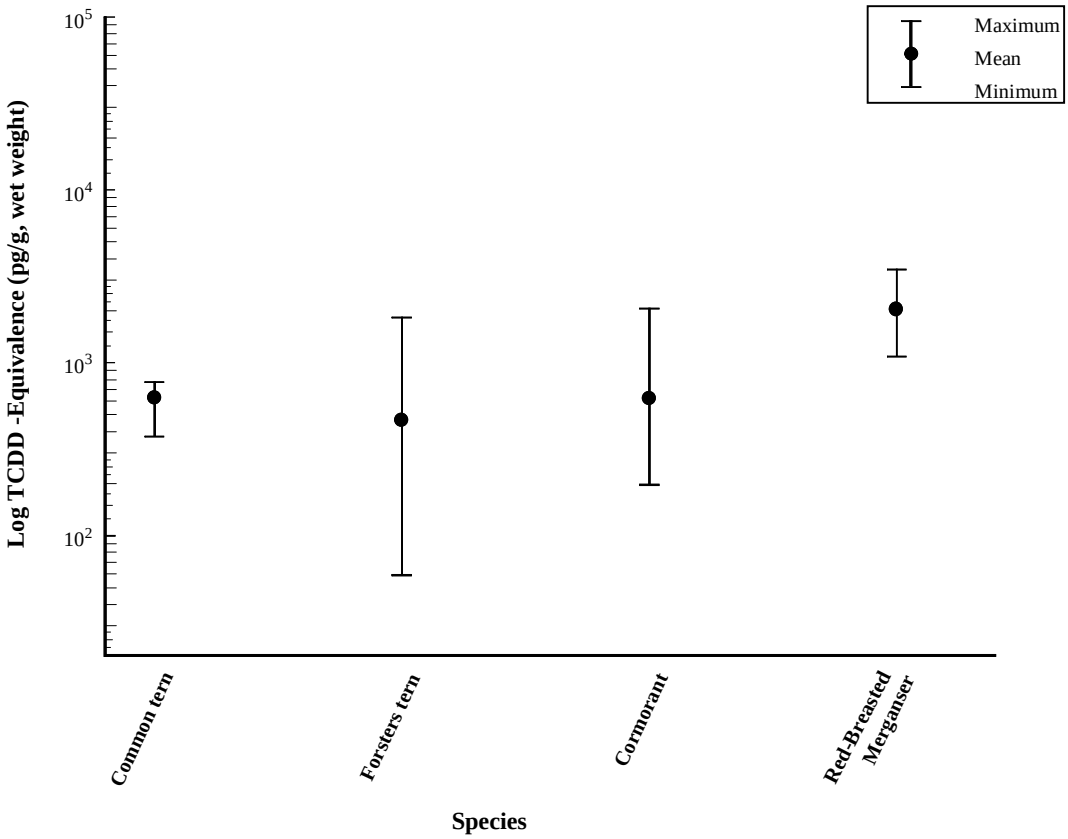


Figure 4-13. PCB TCDD-eq concentrations in assessment area bird eggs, 1983-1996.
TCDD-eq concentrations are calculated from measured PCB congener concentrations using the WHO Avian (U.S. EPA, 1998b) and Kennedy et al. (1996a) TEFs. See Table 4-3 for data sources.

Table 4-3
Sources of Assessment Area PCB Congener Data in Bird Eggs[a]

Species	Year of Collection	Number of Samples	Source
Red-breasted merganser	1990	12	Williams et al., 1995a
Double-crested cormorant	1988 1989 1994-1995	pool of 18 eggs 11 pools of 33 eggs 10	Yamashita et al., 1993 Williams et al., 1995a Custer, pers. comm., 1998
Common tern	1988 1996	2 6	Ankley et al., 1993 Appendix B of this report
Forster's tern	1982 1983 1988 1989 1996	2 6 5 5 6	Smith et al., 1990 Kubiak et al., 1989 Harris et al., 1993 Jones et al., 1993 Appendix B of this report

a. Only studies that included analysis of nonortho congeners (e.g., PCB 77, PCB 81, PCB 126, PCB 169) were used.

CHAPTER 5
INJURY EVALUATION

The previous chapter demonstrated that birds in the assessment area have been exposed to PCBs and that concentrations of PCBs measured in their tissues have exceeded concentrations that are reported to result in toxicological effects in sensitive species. In this chapter, we evaluate evidence from field studies that birds in the assessment area have suffered adverse effects as a result of PCB exposure. The species for which injuries are assessed are Forster's, common, and Caspian terns (Section 5.1); double-crested cormorant (Section 5.2); black-crowned night heron (Section 5.3); tree swallow (Section 5.4); red-breasted merganser (Section 5.5); and bald eagles (Section 5.6). These species are evaluated because of the field data available on adverse effects in the assessment area. However, it is emphasized that, with the exception of tree swallows, these birds are representative of a broader guild of birds for which fish are an important dietary component. This guild also includes other species that inhabit Green Bay, such as great blue herons, green-backed herons, white pelicans, ospreys, and gulls.

Assessment of injury to waterfowl according to the injury definitions related to exceedences of PCB tolerance levels or establishment of waterfowl consumption advisories is addressed in Chapter 6.

5.1 FORSTER'S, COMMON, AND CASPIAN TERNS

5.1.1 Status and Ecology in Green Bay

Forster's, common, and Caspian terns arrive at their nesting colonies in Green Bay in April and May and depart for their winter habitats in the southern United States and Central and South America in September and October (Ludwig, 1965; Burger and Gochfeld, 1991). Their nesting areas are usually on islands, where they are safe from land-based predators such as raccoons, foxes, and mink (Burger and Gochfeld, 1991). In the assessment area, the primary nesting areas for Forster's terns currently are the Confined Disposal Facility (CDF) near the mouth of the Fox River, Long Tail Point (approximately 3 miles from the mouth of the Fox River along the western shore of the bay), and Oconto Marsh (at the mouth of the Oconto River on the western shore of the bay). Common terns nest on the CDF, and Caspian tern colonies are located on Gravelly and Gull islands (between northern Green Bay and Lake Michigan) (see map, Figure 2-4).

Population Status in Green Bay

In 1935, Forster's terns were rare breeders in Wisconsin (Mossman, 1988). The first annual statewide census in 1978 found 136 pairs nesting in Green Bay. This total increased to 435 pairs by 1987. The lack of rigorous census data before the late 1970s makes it difficult to evaluate long-term population changes of Forster's terns in Green Bay. The only conclusion that is supported by the data is that the population increased between the late 1970s and the late 1980s. The State of Wisconsin listed the Forster's tern as endangered in 1979 (Mossman, 1988).

The Green Bay breeding population of common terns also increased over the same time period (late 1970s until the late 1980s). In 1979 there were 60 pairs breeding in Green Bay, and in 1986 there were 600 pairs (Matteson, 1988). Most of this increase took place between the 1984 and 1985 censuses (66 to 427 pairs). This rapid rate of increase could not have been supported by local productivity alone and must have been at least partly caused by immigration from outside of the area. Data on Green Bay common tern populations before this period of increase are sparse, but there is some evidence for a breeding population of several hundred pairs in the 1940s (Matteson, 1988). The State of Wisconsin listed the common tern as endangered in 1979 (Matteson, 1988).

Caspian tern breeding numbers in Green Bay and Lake Michigan have also increased over the last 20 years. In 1977 and 1978 there were 602 nests on Gravelly and Gull islands; by 1991, there were over 1,000 nests (Ewins et al., 1994). This increase is part of a general increase in the Great Lakes Caspian tern metapopulation, which has grown by at least 90% since the late 1970s (Ewins et al., 1994). The State of Wisconsin listed the Caspian tern as endangered in 1989 (Matteson, 1993).

5.1.2 Pathway and Exposure Analysis

Data presented in Chapter 4 of this report show that Forster's, common, and Caspian terns in the assessment area have been exposed to elevated concentrations of PCBs relative to birds collected from reference areas. The purpose of the supplemental pathway analysis presented in this section is to identify the environmental components through which this exposure has occurred. Specifically, we address the following questions: What are the principal prey items of the three species? Where do the species feed? Are their prey items contaminated with PCBs?

Diets and Foraging Areas

Forster's, common, and Caspian terns are mainly piscivorous (Salt and Willard, 1971; Cramp, 1985; Burger and Gochfeld, 1991; Fraser, 1994). Although few data quantitatively describe their diets in Green Bay, several studies carried out elsewhere in the Great Lakes provide evidence of their probable diets in the assessment area.

Fraser (1994) found that during courtship feeding and chick provisioning, Forster's terns at Lake Osakis, Minnesota, mainly ate yellow perch, shiners, and sunfish. Most of these fish were 7 cm or less in length. Trick (1982) reported that Forster's terns in Green Bay generally forage in littoral areas (i.e., areas of shallow water) adjacent to marshes or coastlines. This was also true at Lake Osakis, where Fraser (1994) found that Forster's terns generally foraged over shallow water within 5-20 m of the shore. At Lake Osakis, Forster's terns generally foraged within 5 km of the breeding colony (Fraser, 1994). While foraging distance is likely to be affected by site-specific factors such as the size of the water body, shoreline configuration, and the spatial distributions of feeding and nesting sites, foraging close to the colony is also likely to apply to Foster's terns in Green Bay.

In Lake Ontario, 90% of the diet of breeding common terns was alewives (*Alosa pseudoharengus*) and smelt (*Osmerus mordax*), whereas in Lake Erie, smelt, emerald shiners (*Notropis atherinoides*), and trout-perch (*Percopsis omiscomaycus*) were the main items (Courtney and Blokpoel, 1980). During 1990 and 1991, the diet of breeding common terns on Lake Erie was dominated by smelt and emerald shiner (Burness et al., 1994). Common terns also typically forage within a few kilometers of the breeding colony (Cramp, 1985; Burness et al., 1994). Birds nesting on the Green Bay CDF would probably obtain most of their food locally and within a few kilometers of the mouth of the Fox River.

Data have been reported on the diets of Caspian terns in Green Bay. Ewins et al. (1994) collected 31 regurgitated pellets from the vicinities of nests on Gravelly Island in 1991. All pellets contained the remains of alewives, 10% contained smelt, and 3% contained centrarchid remains. Using pellets to investigate avian diet can be difficult (e.g., Ewins et al., 1994); however, it seems likely that the diets of adult Caspian terns (the pellets were collected before chick hatching) nesting at Gravelly Island in 1991 comprised, in large part, alewives. Alewives and smelt have been shown in other studies to be important components of Caspian tern diet in Lake Michigan waters (Ludwig, 1965). No data have been reported on the foraging ranges of Caspian terns breeding in the assessment area, or elsewhere in the Great Lakes. However, given their larger body size, their foraging ranges may be larger than those of common or Forster's terns.

PCBs in Prey Items

Whole-body PCB concentrations in alewives, gizzard shad, and smelt were measured in 1989 as part of the development of the Green Bay Mass Balance Model (Connolly et al., 1992). Fish were collected from six zones (Figure 5-1) within the assessment area: the Fox River, the eastern and western halves of the inner bay from the Fox River mouth to Little Tail Point (approximately 10 miles north of the Fox River mouth), the eastern and western halves of the inner bay from Little Tail Point to Chambers Island, and the outer bay (beyond Chambers Island).

Figures 5-2, 5-3, and 5-4 show mean total PCB concentrations measured in the three forage fish species. In general, mean concentrations were higher for gizzard shad and alewives, which are

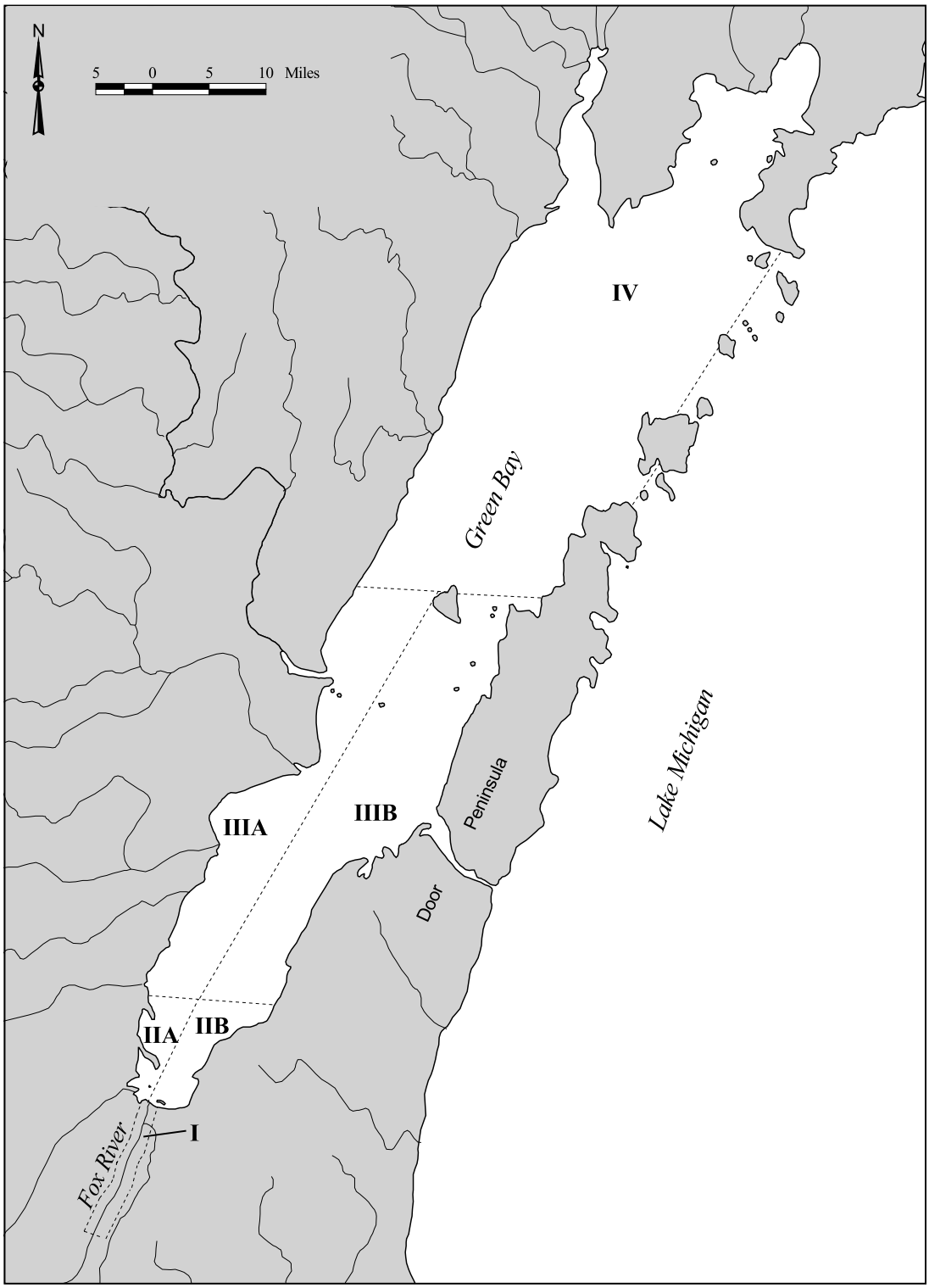


Figure 5-1. Forage fish sampling zones in 1989.

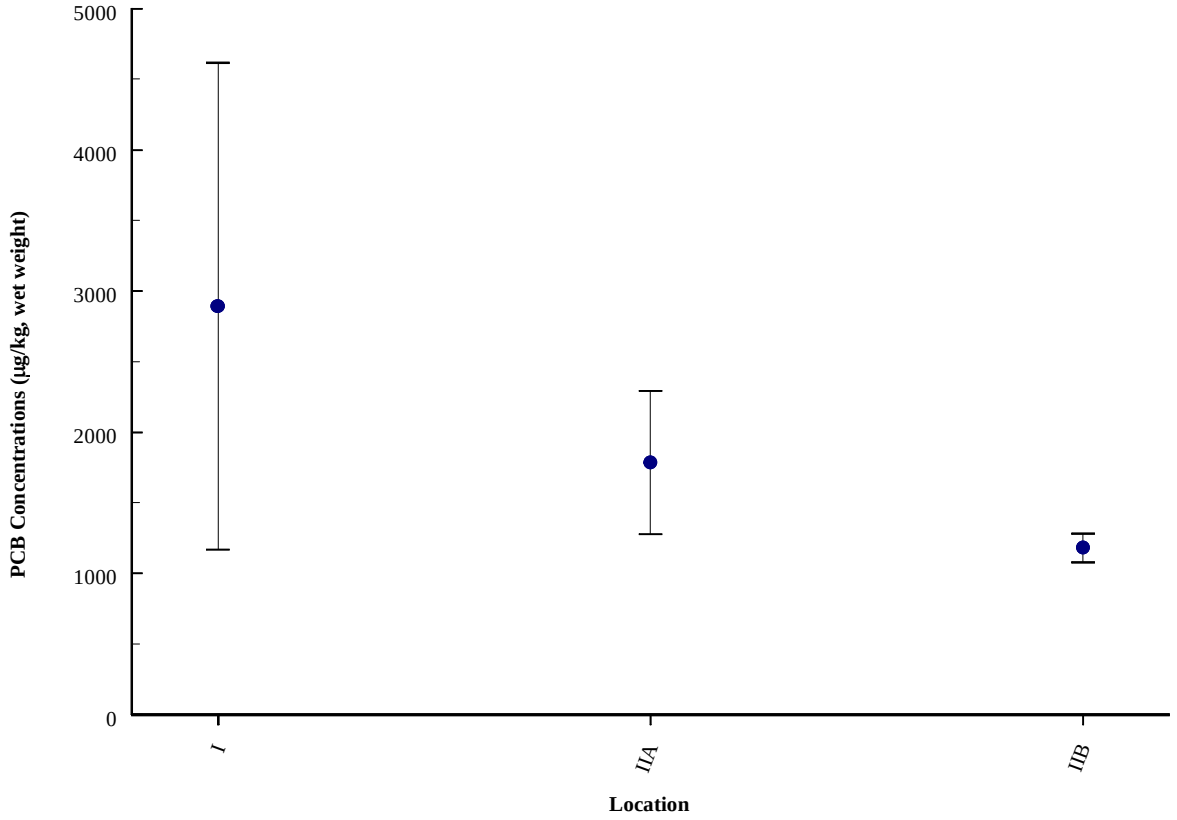


Figure 5-2. Total PCB concentrations in Green Bay gizzard shad, 1989. Bars equal means plus or minus 1 standard deviation. Data from Green Bay Mass Balance Model (Connolly et al., 1992).

relatively lipid-rich (Rottiers and Tucker, 1982; Oliver and Niimi, 1988), than for smelt. A general spatial pattern of decreasing concentrations with increasing distance from the Fox River is also evident (i.e., from zones I to IV). Concentrations in alewives also appear to be higher along the eastern shore of the bay (zones IIB and IIIB) than along the western shore (zones IIA and IIIA). This spatial pattern of PCBs in forage fish is consistent with that observed in sediment and is indicative of the Fox River being the primary source of PCBs to the bay (Manchester-Neesvig et al., 1996). A similar PCB concentration gradient has been observed in young-of-the-year littoral fishes collected from wetlands and beaches along Green Bay (Bruzner and DeVita, 1998). The forage fish data indicate that piscivorous bird exposure to PCBs in prey items tends to decrease with distance from the Fox River, yet is elevated throughout the bay. These data confirm that piscivorous birds in the assessment area are exposed to PCBs in their diet.

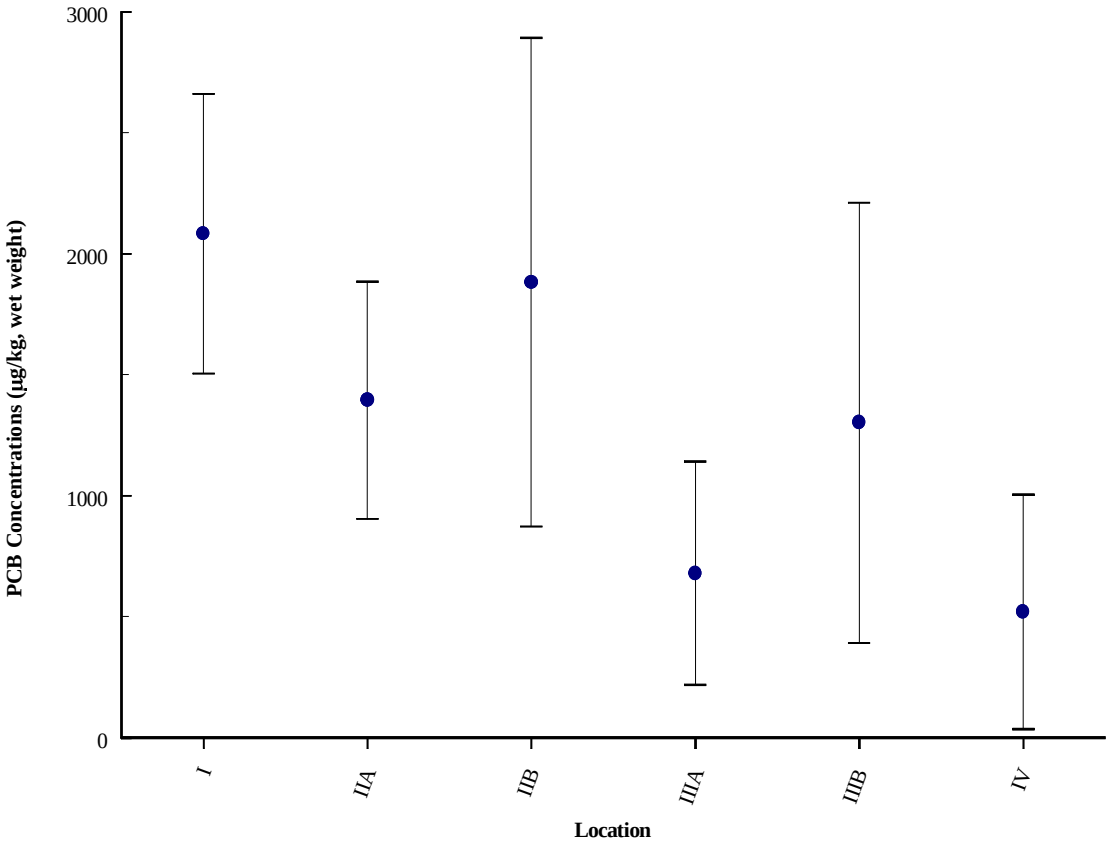


Figure 5-3. Total PCB concentrations in Green Bay alewives, 1989. Bars equal means plus or minus 1 standard deviation. Data from Green Bay Mass Balance Model (Connolly et al., 1992).

Evidence of PCB Uptake

Harris et al. (1993) monitored PCB concentrations in Forster's tern chicks on the Kidney Island CDF from hatching to fledgling. They found that the PCB concentration during this rapid growth period remained relatively constant, showing that the chicks were ingesting PCBs at a rate sufficient to keep pace with the increase in body weight. These data demonstrate that Forster's tern chicks were being fed PCB-contaminated food.

5.1.3 Field Studies of Injuries to Green Bay Terns

Forster's Tern

Field study descriptions. Two sets of studies of the potential effects of contaminants on the reproduction of Forster's terns in Green Bay have been performed.

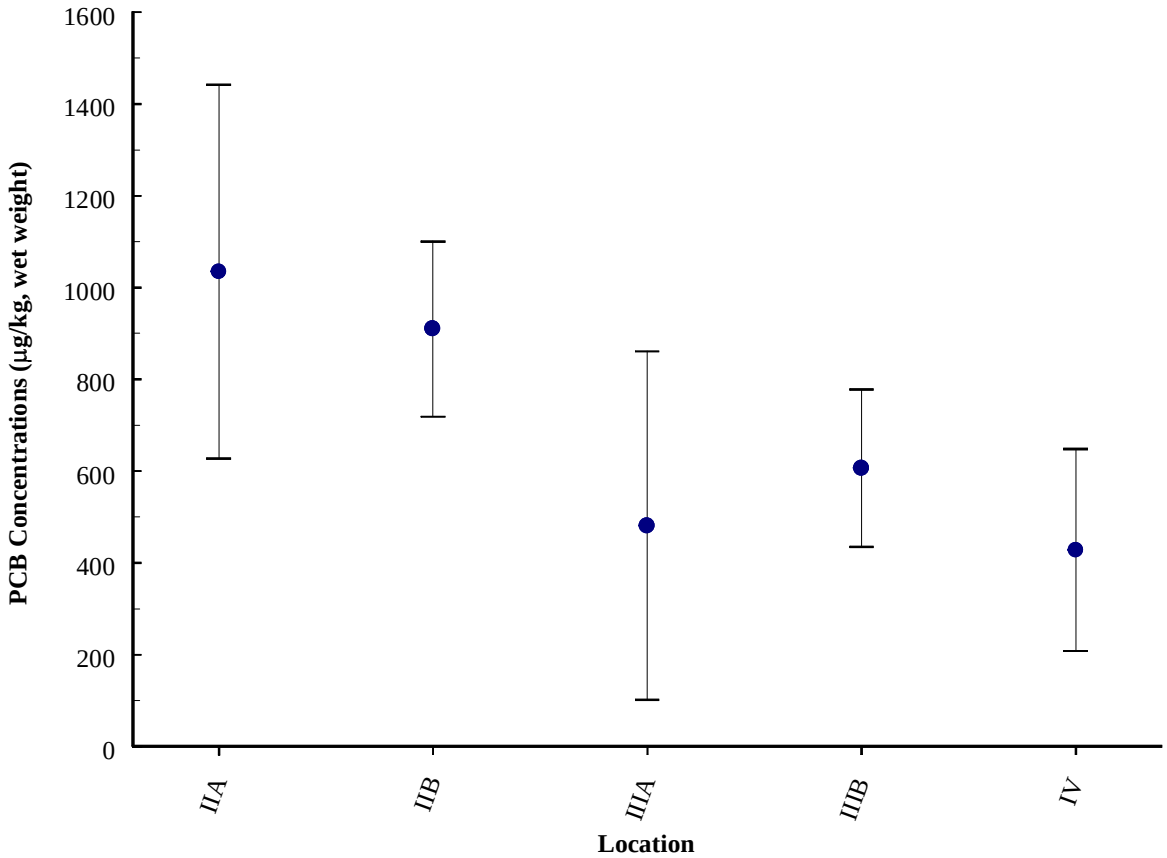


Figure 5-4. Total PCB concentrations in Green Bay smelt, 1989. Bars equal means plus or minus 1 standard deviation. Data from Green Bay Mass Balance Model (Connolly et al., 1992).

Hoffman et al. (1987) and Kubiak et al. (1989). In these companion studies, contaminant concentrations, reproductive performance, deformity rates, and biochemical responses were compared between Forster's terns nesting in Oconto Marsh, Green Bay (at the mouth of the Oconto River) and Forster's terns nesting at Lake Poygan, Wisconsin, an inland lake located in the Fox River drainage upstream of paper company PCB sources. Reproductive performance (but not contaminants, deformity rates, or biochemistry) was also monitored in Forster's terns nesting on Long Tail Point in inner Green Bay. The field work was performed in 1983.

Six tern eggs were analyzed for contaminants from both the Oconto Marsh (Green Bay) and Lake Poygan (reference) colonies. Eggs from the Oconto Marsh colony had a mean PCB concentration of 19.2 mg/kg wet weight (median of 22.2 mg/kg wet weight), and the Lake Poygan eggs had a mean of 4.6 mg/kg wet weight (median 4.5 mg/kg wet weight). The mean PCB concentrations between eggs from the two colonies were reported as being significantly different ($p < 0.05$). No egg contaminant data were collected for the Long Tail Point (Green Bay) colony.

In a companion paper to Kubiak et al. (1989), Tillitt et al. (1993) reported results of H4IIE bioassays on Green Bay and Lake Poygan Forster's tern eggs. The Green Bay eggs averaged 214.5 pg/g TCDD-EQ, compared to 23.4 pg/g TCDD-EQ at Lake Poygan. This difference was reported as being statistically significant.

The reproductive successes of the colonies are summarized in Table 5-1. Egg hatching rates were significantly lower in the Green Bay colonies than in the Lake Poygan colony. Of the eggs monitored, 40% and 55% hatched successfully at the Oconto Marsh and Long Tail Point colonies, respectively, whereas 88% of the eggs laid at the Lake Poygan site hatched. The percentage of nesting pairs that produced at least one fledgling was also lower in the Green Bay colonies, as was the average number of fledglings produced per nest. The reproductive success of terns at the Long Tail Point colony was intermediate between that of the Oconto Marsh and Lake Poygan colonies.

Table 5-1
Reproductive Success of Green Bay and Reference Colonies of Forster's Terns

Colony	Mean PCB Concentration (n = 6, mg/kg, wet weight)	Percent of Eggs that Hatched	Percent of Nesting Pairs that Were Successful[a]	Number of Fledglings/Nest
Oconto Marsh (Green Bay)	19.2	40% (14/35)	0% (0/12)	0
Long Tail Point (Green Bay)	— [b]	55% (18/33)	42% (5/12)	0.58
Lake Poygan (reference)	4.6	88% (30/34)	91% (10/11)	1.55

a. Successful means producing at least one fledgling. Only pairs from which no eggs were removed or exchanged are included.
b. No chemistry measurements were made at this colony.

Source: Kubiak et al., 1989.

The causes of the reduced hatching success in the Green Bay colonies were investigated using a combination of laboratory incubation of eggs collected at Oconto Marsh and Lake Poygan and field experimentation in which eggs were transferred between the colonies. In laboratory incubators, only 37% of the 19 Oconto Marsh eggs hatched, compared with 75% of the 20 Lake Poygan eggs. This statistically significant difference indicates that factors that were intrinsic to the eggs themselves affected hatchability under controlled conditions. Our own statistical analysis of the Kubiak et al. (1989) data showed that the hatching success rates in the incubators did not

differ significantly from those in the natal colonies in the field (χ^2 = 1.45 and 1.64, respectively; $p > 0.25$, 1 df).

In the egg transfer experiment, Kubiak et al. (1989) found that eggs removed from the Oconto Marsh colony and incubated by Lake Poygan adults had a significantly higher hatching success rate (94%) than Oconto Marsh eggs incubated in their natal colonies (55%), or in the laboratory (37%). This indicates that factors extrinsic to the eggs themselves were also important in reducing hatching success in Green Bay. This conclusion is supported by the fact that eggs transferred from the Lake Poygan colony to Oconto Marsh had a significantly lower hatching success (11%) than Lake Poygan eggs incubated by Lake Poygan adults (88%) or in the incubator (75%). These results are summarized in Table 5-2. The most plausible explanation for this extrinsic effect is that the reproductive behavior of the Oconto Marsh adults was less likely to result in a successful reproductive outcome than the reproductive behavior of Lake Poygan adults.

Table 5-2
Percent Hatching Success Results of Forster's Tern Egg Transfer and Laboratory Hatching Study

Source Colony	Colony Where Eggs Incubated		
	Lake Poygan	**Oconto Marsh**	**Laboratory**
Lake Poygan	88%	11%	75%
Oconto Marsh (Green Bay)	94%	55%	37%

Source: Kubiak et al., 1989.

Additional evidence that the reproductive performance was poorer in the Green Bay colonies was provided by data on incubation periods and nest abandonment rates. Oconto Marsh eggs took 4.6 days longer than Lake Poygan eggs to hatch in the laboratory. In the field, Oconto Marsh eggs incubated by their own parents took significantly longer to hatch (by 8.2 days) than Lake Poygan eggs incubated by their own parents. There was no difference in the time required for incubation between Lake Poygan eggs hatched in the natal colony and those hatched in the laboratory incubator. From these data, Kubiak et al. (1989) concluded that "about half of the longer incubation period for dirty eggs in the field . . . must have been due to intrinsic factors and about half to extrinsic factors."

In a companion study to Kubiak et al. (1989), Hoffman et al. (1987) reported incidences of deformities and liver microsomal aryl hydrocarbon hydroxylase (AHH) activity in embryos from the Forster's tern eggs from the Oconto Marsh and Lake Poygan colonies. In addition to lower body weights of hatchlings (also found by Kubiak et al., 1989), Hoffman et al. (1987) found that the Oconto Marsh eggs had significantly higher AHH activity (by a factor of 3), significantly greater liver-to-body weight ratios, and significantly shorter femurs. Three instances of structural

deformities were also found in the Oconto Marsh embryos. These were one embryo with a crossed bill, one with a poorly ossified foot and a short lower mandible, and one with an incompletely ossified ilium. No deformities were found in Lake Poygan embryos. In total, 16.7% of Oconto Marsh hatchlings and embryos had structural deformities, compared with 0% of the Lake Poygan neonates. This difference was reported as being statistically significant. Also, 27.7% of Oconto Marsh hatchlings and embryos had edema, compared with 13.3% of Lake Poygan neonates, although this difference was not statistically significant.

The total PCB concentrations reported by Kubiak et al. (1989) in Oconto Marsh Forster's tern eggs exceed concentrations shown to cause toxicity (Chapter 3) and are significantly higher than concentrations measured in Lake Poygan eggs. Kubiak et al. (1989) found that in addition to PCBs, several other contaminants were also higher in Oconto Marsh tern eggs than in Lake Poygan eggs. These contaminants were oxychlordane + heptachlorepoxide (median of 0.20 mg/kg wet weight in Oconto Marsh eggs vs. 0.04 mg/kg wet weight in Lake Poygan eggs); *p,p'* DDE (median of 1.8 mg/kg wet weight vs. 0.45 mg/kg wet weight); cis-nonachlor + *p,p'* DDD (median of 0.12 mg/kg wet weight vs. 0.01 mg/kg wet weight); hexachlorobenzene (median of 0.10 mg/kg wet weight vs. 0.02 mg/kg wet weight); heptachlor (median of 0.09 mg/kg wet weight vs. 0.02 mg/kg wet weight); toxaphene (median of 1.10 mg/kg wet weight vs. 0.37 mg/kg wet weight); dioxins (median of 101.5 pg/g vs. 25.0 pg/g); and furans (median of 18.5 pg/g vs. 9.0 pg/g). Based on the TEF approach, Kubiak et al. concluded that dioxins and furans contributed less than 10% of the total TCDD-eq in the eggs, with nonortho and mono-ortho PCB congeners contributing the rest. Similarly, Kubiak et al. concluded that the measured concentrations of toxaphene and hexachlorobenzene in the tern eggs were below toxic thresholds. Therefore, Kubiak et al. (1989) concluded that PCBs were the primary cause of the toxic effects observed in Green Bay Forster's terns.

Harris et al. (1993) and Ankley et al. (1993). These companion studies monitored the reproductive success and measured egg contaminant concentrations for a Forster's tern colony on the Kidney Island CDF located at the mouth of the Fox River (Figure 2-4). No reference colonies were evaluated. The field work was conducted in 1988.

The mean total PCB concentration measured in eggs from the CDF was 7.3 mg/kg wet weight (n = 5, median of 7.4) (Harris et al., 1993). The Kidney Island CDF Forster's terns had an egg hatching rate of 81% (65 of 80), similar to that found by Kubiak et al. (1989) at the Lake Poygan reference colony (88%). However, only 65% of the pairs monitored at the CDF produced at least one fledgling, whereas 91% of the pairs at the Lake Poygan colony monitored by Kubiak et al. (1989) produced at least one fledgling. Similarly, the average number of fledglings per nest was 1.0 for Forster's terns at the CDF and 1.5 for those at Lake Poygan in the Kubiak et al. study.

Harris et al. (1993) found that many of the CDF Forster's tern chick deaths occurred at a comparatively late stage of development (>20 days after hatching). These deaths were, in many cases, preceded by weight loss. Harris et al. (1993) noted that the pattern of weight loss was

characteristic of the "wasting syndrome" caused by organochlorine compounds. Furthermore, Harris et al. noted that as many chicks from nests with one and two young died as did those from nests with three young, and suggested that this implied that starvation was not the cause of the deaths. However, chick mortality due to food shortages and starvation, even late in development, is not uncommon in tern colonies (Langham, 1972; Burger and Gochfeld, 1991). Absent a breakdown of the hatch order of the young that died, it is not possible to exclude starvation due to local food shortage.

Ankley et al. (1993) showed that nesting Forster's terns on the CDF accumulated PCB residues during growth, indicating that they were obtained from local sources.

Conclusions from Forster's tern field studies. The types of adverse effects observed in the field and their relationship to measured egg PCB concentrations show that Green Bay Forster's terns have been adversely affected by exposure to PCBs. Effects and PCB exposure were most severe at the Oconto Marsh colony, where hatching success and number of fledglings per nest were lower than those of a reference colony (Kubiak et al., 1989). Specific effects included embryonic deformations, skeletal deformities, and edema, all of which can be caused by PCBs, as discussed in Chapter 3. Reproductive success (percentage of eggs hatching and number of fledglings per nest) was also lower at the Long Tail Point colony (Kubiak et al., 1989). At the Kidney Island CDF colony, where egg PCB concentrations were lower than at Oconto Marsh, egg hatching was not reduced (Harris et al., 1993). The number of fledglings per nest also was reduced, although the cause of the reduction is not clear.

The controlled egg switching experiments by Kubiak et al. (1989) show that extrinsic factors, e.g., decreased parental attentiveness, contributed to the lowered reproductive success. The adverse behavioral effects of PCBs on nesting adults have been documented in several studies. In a laboratory study using ring doves, Peakall and Peakall (1973) found that PCBs caused decreased parental attentiveness during incubation. Fox et al. (1978) showed that Lake Ontario herring gulls, which had higher PCBs and a lower rate of reproductive success than those from reference areas, also showed increased time away from nests and decreased nest defense during egg incubation. These documented adverse effects of PCBs on adult behavior during nesting are consistent with the findings of Kubiak et al. (1989) that extrinsic factors contributed to the reduced reproductive success of Green Bay Forster's terns.

Contaminants other than PCBs measured in the eggs were not significant contributors to the observed toxicity. Kubiak et al. (1989) determined that dioxins and furans, which can cause effects similar to those observed, accounted for less than 10% of the TCDD-eq in the eggs compared with PCBs. Other contaminants present in the eggs (e.g., DDE) are not known to cause the behavioral abnormalities or deformities that were observed, or were not present at concentrations sufficient to cause the observed effects (Kubiak et al., 1989).

Further evidence that DDE was not responsible for the adverse effects observed in Green Bay Forster's terns is provided by a study performed by King et al. (1991) in Texas. In this study the DDE concentrations in Forster's tern eggs from the contaminated and reference colonies were similar to the levels reported by Kubiak et al. (1989). PCB concentrations in the Texas Forster's tern eggs were low relative to the Green Bay eggs (1.2-2.3 mg/kg wet weight). Neither PCBs nor DDE were correlated with any measure of breeding success.

The field studies show that within the assessment area PCB exposure and effects were most severe at the Oconto Marsh colony and lowest at the Kidney Island CDF colony, which is consistent with the PCBs causing the observed effects. The reason for the lower PCB exposure and severity of effects observed in Forster's terns at the Kidney Island CDF compared with those at the Oconto Marsh is most likely a combination of both spatial and temporal variability. Based on the reproductive success endpoints, Forster's terns nesting at Long Tail Point, which is approximately 3 miles north of the Kidney Island CDF, may also have had lower contaminant exposure than did those at Oconto Marsh (Kubiak et al., 1989) (contaminants were not measured in Long Tail Point eggs). Data on PCBs in herring gull eggs from the Big Sister Island colony in Green Bay support the conclusion that temporal variability could also contribute to the observed variation in PCB concentrations of the Kidney Island CDF and Oconto Marsh eggs. Although PCB concentrations in herring gull eggs show no long-term trend from 1983 through 1996, concentrations vary from year to year by over a factor of two. Sample sizes of Forster's tern eggs for PCB analysis were smaller (six for Oconto Marsh and five for CDF) than those for herring gull eggs (10), which would increase between-year variability in the tern data. The herring gull egg PCB data also support the conclusion that the lower PCB concentration in Forster's tern eggs in 1988 at the CDF may not be indicative of a trend from 1983 to 1988 of declining PCB exposure for fish-eating birds in Green Bay.

Common Tern

Field study description. One study has been performed that is relevant to evaluating the potential effects of PCBs on the reproductive biology of common terns in Green Bay.

Hoffman et al. (1993). In this study, 35 newly laid, unincubated eggs of common terns were collected in 1985 from a colony situated on the Kidney Island CDF in Green Bay. Eggs were also collected from two reference colonies in nonindustrialized areas of northern Lake Michigan, Cut River, and Pointe aux Chenes. All eggs were artificially incubated in the laboratory, and hatching success, neonate morphology, biomarker activity, and contaminant concentrations were compared between colonies. No monitoring of reproductive success in the field was conducted as was done in the Forster's tern studies.

The Green Bay eggs had higher PCB concentrations (geometric mean of 10.0 mg/kg, wet weight; n = 10) than did eggs from the Cut Island (geometric mean PCB concentration of 4.7 mg/kg, wet weight) or Pointe aux Chenes (geometric mean PCB concentration of 4.0 mg/kg, wet weight)

colonies. Mean liver AHH activity, a measure of exposure to TCDD-like contaminants, was also significantly higher in Green Bay eggs (mean of 23 pmol/min/mg protein, n = 22) than in either reference colony (10 and 9 pmol/min/mg protein and n = 22 and n = 12 at Cut River and Point aux Chenes, respectively).

Table 5-3 summarizes the parameters measured in the study that differed in the Green Bay and the reference area eggs, as well as the mean PCB concentrations. Hatching success of eggs from the Green Bay CDF (71%) was significantly lower than that of eggs from the Cut River colony (85%), but not statistically different from that of eggs from the Point aux Chenes colony (73%). Similarly, the femur length to body weight ratio in 1-day-old chicks was lower for Green Bay chicks than for Cut River chicks, but was not different from Point aux Chenes chicks. Four of the 35 (11%) Green Bay embryos or chicks were deformed, whereas no deformities were observed in any of the 55 reference embryos or chicks. Other morphological parameters measured, including hatching weight, liver weight, liver weight to body weight ratio, crown-rump length, and femur length, were not different between eggs from the different colonies.

Table 5-3
Differences between Eggs from Green Bay and Reference Common Tern Colonies When Incubated in the Laboratory

Source of Eggs	PCB Concentration (geometric mean, mg/kg wet weight)	Percent Hatching Success	Femur Length to Body Weight Ratio (x 100)	Percent Deformed Embryos and Hatchlings
Kidney Island CDF, Green Bay	10.0[a]	71%[b]	93.5[b]	11%[b]
Cut River, Michigan	4.7	85%	108.9	0%
Point aux Chenes, Michigan	4.0	73%	101.0	0%

a. Reported as statistically significantly different from Cut River and Point aux Chenes colonies.
b. Reported as statistically significantly different from Cut River colony.

Source: Hoffman et al., 1993.

There were no significant differences between the areas in egg concentrations of DDE, indicating that DDE was not the cause of the observed differences between colonies. Mercury was measured at significantly higher concentrations in the Green Bay eggs (0.76 mg/kg, wet weight) than in Cut River eggs (0.33 mg/kg, wet weight) or Pointe aux Chenes eggs (0.37 mg/kg, wet weight). However, mercury does not induce AHH activity, which was higher in Green Bay chicks than in reference area chicks.

Conclusions from common tern field study. As shown in Table 5-3, the single field study that has been conducted in the assessment area demonstrated that common terns had elevated tissue residues of PCBs, increased deformity rates, and perhaps reduced egg hatching success.

Caspian Tern

Field study descriptions. Five field studies have been reported that are relevant to evaluating the potential effects of contaminants on Caspian terns in the assessment area.

Ludwig and Ludwig (undated report b). This field study was performed on Gravelly and Gull islands in northern Green Bay, at three colonies in northern Lake Michigan, and at colonies in Thunder Bay and Saginaw Bay, Lake Huron, in 1986. The study compared clutch sizes, hatching success, productivity, and the incidences of developmental defects among the colonies. Mean clutch sizes were similar in the colonies on Gravelly and Gull islands and in the three Lake Michigan colonies (2.1, 1.9, 2.0, 2.1, and 2.0, respectively). Hatching success on Gravelly and Gull islands was 72% and 71%, respectively, compared to 81%-84% reported for the three Lake Michigan colonies. Productivity on Gravelly and Gull islands was 0.73 and 0.95 young fledged per nest, respectively, and 0.8-0.91 in the three Lake Michigan colonies. No developmental defects were found in chicks in any of the colonies.

Yamashita et al. (1993). Yamashita et al. collected 18 Caspian tern eggs from Gravelly and Gull islands in 1988. Of these, 13 (72%) contained “live normal” embryos with mean total PCB and DDE concentrations of 11 and 4 mg/kg wet weight, respectively. Three eggs (17%) were infertile, with mean total PCB and DDE concentrations of 10 and 3.2 mg/kg wet weight, respectively, and two eggs (11%) contained deformed embryos and mean total PCB and DDE concentrations of 11 and 6.3 mg/kg wet weight, respectively. The results of this study indicated no clear relationship between PCB and DDE concentrations and egg or embryo viability at the concentrations found in the two colonies.

Ludwig et al. (1996). In this study, live and dead Caspian tern eggs and chicks from five colonies throughout the Great Lakes (including from Green Bay) were examined between 1987 and 1991 for egg death rates and embryonic abnormalities. Egg mortality varied among the five study areas (North Channel of Lake Huron 25%, northern Lake Michigan 27%, Georgian Bay 27%, Green Bay 34%, and Saginaw Bay 42%). Egg mortality rates were highly correlated with TCDD-eq ($r = 0.8$), but not with total PCBs.

Of the 601 Green Bay dead eggs opened and examined, 124 (20.6%) of the embryos had developmental abnormalities. This compares with 17.3% in northern Lake Michigan, 13.2% in the North Channel of Lake Huron, 14.5% in Georgian Bay, and 22.8% in Saginaw Bay (which also is contaminated with PCBs). Of the abnormalities recorded in embryos from dead Green Bay eggs, 19% were edema, 39.2% were gastroschisis, 14.2% were bill defects, 4.7% were foot deformities, and 8.2% were other skeletal deformations.

Of the 162 Green Bay fertile live eggs opened and examined by Ludwig et al. (1996), 94% contained normal embryos and 5.5% contained deformed embryos. This compares with deformity rates of 11.8% in fertile live eggs from northern Lake Michigan and 30.4% in Saginaw Bay fertile live eggs. Five of the Green Bay deformities were club feet, three were gastroschisis.

In this study, 12,124 live Caspian tern chicks were also examined in the five study areas. Of these, 29 (0.02%) had deformities. The deformity rate varied little between Green Bay and the other areas (0.16% Green Bay, 0% Georgian Bay, and 0.18% North Channel of Lake Huron). Green Bay chick deformities comprised 62% clubbed feet and 38% gastroschisis. Ludwig et al. (1996) stated that no cross bills have been recorded in 26,819 Caspian tern chicks banded in the Great Lakes since 1960.

Thus, elevated rates of deformities were observed in dead eggs in Green Bay, whereas eggs and chicks that survived had lower deformity rates. This suggests that the deformities in Green Bay were associated with the viability of the embryos.

Mora et al. (1993). In this study performed in 1990, organochlorine concentrations in adult Caspian tern plasma were compared with age, productivity, and site fidelity (i.e., the proportion of birds that return to the natal area to breed) at eight colonies in Lakes Huron and Michigan (including Gravelly and Gull islands in Green Bay).

Mean total PCB concentrations varied from 0.91 mg/kg wet weight to 3.5 mg/kg wet weight among the study colonies, with the highest concentration in Green Bay. There were no significant intercolony differences in clutch size, hatching success, or fledging success. Of the 4,075 chicks examined at the Green Bay colonies, 0.17% had deformities (four had club feet, three had gastroschisis). This compares with 0.23% in colonies in northern Lake Michigan and 0.94% at Saginaw Bay.

On the basis of recapture rates of banded terns, Mora et al. (1993) argued that Caspian terns hatched in the Green Bay colonies displayed less site fidelity than terns in other regions. Mora et al. (1993) attributed this difference to contaminants, particularly PCBs.

Ewins et al. (1994). In this study, Caspian tern eggs were collected from 10 colonies across the Great Lakes (including Gravelly Island in Green Bay) and analyzed for organochlorine contaminants. Total PCB and DDE concentrations were highest in eggs from Gravelly Island and from Saginaw Bay. Nevertheless, hatching and fledging success were not significantly different at Gravelly Island compared with other areas (Table 5-4).

Conclusions from field studies on Caspian terns. Overall, less evidence exists for depressed reproductive rates among Green Bay Caspian terns than for Forster's and common terns. Of the four studies that examined reproductive injuries among Green Bay Caspian terns (Ludwig and Ludwig undated report b; Mora et al., 1993; Ewins et al., 1994; Ludwig et al., 1996) only one

Table 5-4
Hatching Success and Productivity among Great Lakes Caspian Terns

Study Site	Nests Studied	Egg PCB Concentration (mg/kg, wet weight)	% Hatching Success	Young Fledged/Nest
Gravelly Island, Green Bay	59	15.8	79%	1.07
High Island, northern Lake Michigan	56	not reported	85%	1.13
Cousins Island, North Channel of Lake Huron	28	14.6	47%	0.79
South Watcher Island, Georgian Bay	41	10.2	52%	0.83
Source: Ewins et al., 1994.				

(Ludwig et al., 1996) found reduced reproduction relative to reference conditions. The other three studies found no evidence of adverse effects on reproduction (though Mora et al. report possible behavioral effects among adult Caspian terns in Green Bay). Ludwig et al. (1996) found higher rates of deformities in Green Bay Caspian terns than in colonies not exposed to point source releases of PCBs. Other studies that investigated deformities did not find differences, although the Ludwig et al. study was the most detailed and comprehensive of the studies.

The available studies do not provide strong evidence that the reproductive success of Caspian terns nesting on Gravelly and Gull Islands has been adversely affected by PCB exposure. However, there is some evidence of increased deformity rates in Green Bay Caspian terns.

5.2 DOUBLE-CRESTED CORMORANT

5.2.1 Status and Ecology in Green Bay

Double-crested cormorant population trends in the Great Lakes can be divided into three temporal phases.

Initial colonization and early increases. Before the beginning of the 20th century, double-crested cormorants were unknown as a breeding bird in the Great Lakes. The colonization of the area began between 1913 and 1920 in Lake Superior (Environment Canada, 1995; Weseloh et al., 1995a, b), and probably involved birds spreading from colonies farther west. From this initial bridgehead, double-crested cormorants spread rapidly throughout the region until about 1950, when approximately 1,000 pairs bred in the Great Lakes, and control measures were initiated in an effort to protect fish stocks (Weseloh et al., 1995a).

Mid-century population declines. After 1950, the initial increases in cormorant numbers were followed by spectacular population reductions; by 1972, the Great Lakes population had been reduced by more than 80% (Weseloh et al., 1995a, b). From 1970 through 1974, double-crested cormorants had disappeared, or were close to disappearing, as a breeding species on Lake Michigan, and the total Great Lakes population was reduced to fewer than 150 pairs (Ludwig, 1984). In Wisconsin, the number of cormorants had decreased to 66 pairs by 1972 and the species was listed by the state as endangered (Hatch, 1995). These precipitous declines were accompanied by significant eggshell thinning and breakage. By 1970, eggshells in Ontario colonies were 30% thinner than normal, and in 1972, 95% of the eggs in Lake Huron colonies either disappeared or were broken (Environment Canada, 1995). Because of these losses, productivity in Great Lakes colonies had fallen to about 0.1 to 0.24 fledglings per breeding pair; 0.5 to 1.0 fledglings are required to maintain a breeding population (Ludwig, 1984; Ludwig et al., 1995). Based on the widespread and severe eggshell thinning and breakage, it is likely that the population decreases of the 1950s through early 1970s were caused by the toxic effects of DDE.

Post-1960s population resurgence. In the 1970s, following the ban on the use of DDT in North America, DDE levels in cormorant eggs in the Great Lakes began decreasing. By the late 1980s, egg DDE residues had decreased by more than 80% (Environment Canada, 1995), and populations of double-crested cormorants again increased. By 1992, approximately 3,000 pairs were breeding in Green Bay (Hatch, 1995). Thus, in only 20 years, the Green Bay population increased by a factor of at least 45 (assuming that the 66 Wisconsin pairs in the early 1970s were all in Green Bay). In the Great Lakes over this same period the increase was even greater, about 250-fold from about 150 nests to 38,000 nests (Weseloh et al., 1995a), a doubling time of about 2.5 years. This increase continued into 1994, when Weseloh et al. (1995b) estimated a total Great Lakes population of 60,000 pairs. As a result of this rapid rate of increase, approximately 60% of the world's population of double-crested cormorants currently breed in the Great Lakes (Hatch, 1995).

Residence Patterns and Migrations

Double-crested cormorants breeding in Green Bay are migratory, and most winter in the lower Mississippi Valley and the Gulf of Mexico (Dolbeer, 1991). In his analysis of band recoveries, Dolbeer found a high degree of mixing of midwestern nesting populations during winter; birds from Lakes Huron and Ontario and from Saskatchewan all wintered in the same areas of the lower Mississippi and coastal Texas.

The main breeding colonies of double-crested cormorants in the assessment area are on Cat, Jack, Hat, and Snake islands in Green Bay, and on Spider Island on the east side of the Door Peninsula (see Figure 2-4). Breeding cormorants arrive in Green Bay in April and remain in the area until September/October, when the return migration to the wintering area begins. First year and second year (nonbreeding) birds either remain in their wintering areas during their first summer or return later in the season than the breeding adults (Dolbeer, 1991).

5.2.2 Pathway and Exposure Analysis

A number of the ecological traits of double-crested cormorants predispose them to being potentially highly exposed to contaminants. First, double-crested cormorants begin to arrive in their breeding areas in Green Bay in April, approximately 3-4 weeks before the beginning of egg laying. There is no published information on the length of time it takes double-crested cormorants to form and lay a clutch of eggs. However, the closely related European shag is similar in size (hence metabolic rate), lays similarly sized eggs and clutches, and takes about 22 days from the beginning of egg formation to laying the last egg of a three-egg clutch (Grau, 1996). It is likely that the double-crested cormorant requires a similar time span. Thus, the birds arriving back in Green Bay in April, 3-4 weeks before egg laying begins, have sufficient time to form their eggs using food obtained locally rather than relying on reserves built up in the wintering area. Also, if cormorants forage close to their colonies during the pre-laying period, as they do during incubation and chick rearing, it is likely that the majority of females undergoing oogenesis will obtain their food from inner Green Bay, in the case of the Cat Island birds, or from the northeastern coast of the Door Peninsula, in the case of Spider Island birds. These ecological traits render the Green Bay double-crested cormorants vulnerable to exposure to local contaminants during the formation of the most sensitive life stage, the embryo.

As fish-eating predators, double-crested cormorants feed high in aquatic food chains. This renders them vulnerable to exposure to bioaccumulative contaminants. Also, alewives, one of the cormorants' major prey species in Green Bay and the Great Lakes (Ludwig and Ludwig, undated report a; Weseloh and Ewins, 1994; Neuman et al., 1997), are richer in lipid than other forage fish species (Oliver and Niimi, 1988; Rottiers and Tucker, 1982). By consuming lipid-rich prey, Green Bay cormorants increase their exposure to lipophilic contaminants such as PCBs.

Data reviewed in Chapter 4 show that double-crested cormorants in Green Bay have elevated PCB residues in their tissues. In this section we identify the environmental components through which Green Bay cormorants have been exposed to these PCBs. Specifically, we address three questions: What organisms constitute the principal diet of cormorants in the assessment area? Where do Green Bay cormorants feed? Are the prey of cormorants in Green Bay contaminated with PCBs?

Diet

A number of studies have shown that double-crested cormorants in the Great Lakes and adjacent areas eat mainly fish, in particular forage fish such as alewives and smelt (Ludwig and Ludwig, undated report a; Belonger, 1983; Hobson et al., 1989; Neuman et al., 1997). Of these, Neuman et al., Ludwig and Ludwig, and Hobson et al. showed that crayfish (*Orconectes* spp.) are also regularly found in small numbers in double-crested cormorant food samples; however, this could be due to secondary consumption (i.e., from the stomachs of fish that had been consumed).

The specific composition of cormorant diets can vary spatially and temporally (Neuman et al., 1997). This variability may reflect differences in the availability of prey species. For example, in Green Bay in 1983, alewives and yellow perch comprised more than 90% of 1,073 identifiable fish obtained from regurgitates of nestling cormorants on Willow Island (alewives, 51.6%; yellow perch 39.3%) (Belonger, 1983). At Gravel, Fish, and Spider islands, alewives similarly comprised a high proportion of the diet (69.2% of identifiable fish); however, yellow perch comprised only 1.9% (Belonger, 1983). Yellow perch were replaced in the cormorant diet at these locations by sculpin (*Cottus sp*.) (11.8% of identifiable fish), ninespine stickleback (*Pungitius pungitius*) (8.1% of identifiable fish), Johnny darter (*Etheostoma nigrum*) (5.2% of identifiable fish), and spottail shiner (*Notropis hudsonius*) (1.9% of identifiable fish). The few yellow perch in the latter samples was probably because the shallow water habitat preferred by this fish species is not readily available at Gravel, Fish, and Spider islands. Alewives and smelt were the most frequent food items in regurgitates from adult and young cormorants in northern Green Bay colonies from 1986 to 1988 (Ludwig and Ludwig, undated report a).

Foraging Areas

Custer and Bunck (1992) tracked the foraging flights of cormorants from the Cat and Spider Island colonies in Green Bay and found that the birds typically obtain their fish prey from waters relatively close to the colonies. Foraging flights from Cat Island were restricted to within 40 km of the colony, and the mean foraging flight distance was 2 km. Most of the foraging flights from Cat Island ended with the birds landing in the central and western inner bay area (Figure 5-5). Many foraging flights ended at the confluence of the Fox River with Green Bay, and less than 1% of birds flew up the river. Cormorants from Cat Island tended to forage in shallow water areas (less than 1.8 m deep) and avoided deeper water.

Double-crested cormorants from Spider Island also tended to forage close to the colony (Custer and Bunck, 1992). The maximum distance flown from the colony was 12 km, and the mean was 2.4 km. The majority of the Spider Island birds foraged off of the east coast of the Door Peninsula (Figure 5-6). They preferred water depths of less than 9.0 m, but avoided depths of less than 1.8 m (the preferred depth for the Cat Island birds). No Spider Island cormorants were recorded flying into Green Bay to forage during the study.

Prey Contamination

USFWS (1993) collected stomach contents from adult cormorants on Cat Island during the 1988 breeding season and found concentrations of total PCBs averaging 3.3 mg/kg (wet weight). These data show that, at least during the breeding season, double-crested cormorants in Green Bay ingest prey contaminated with PCBs. USFWS (1993) also showed that adult cormorant PCB body residues approximately doubled during the 1988 breeding season (Figure 5-7), indicating a local source of the PCBs. These data confirm that exposure to PCBs is dietary.

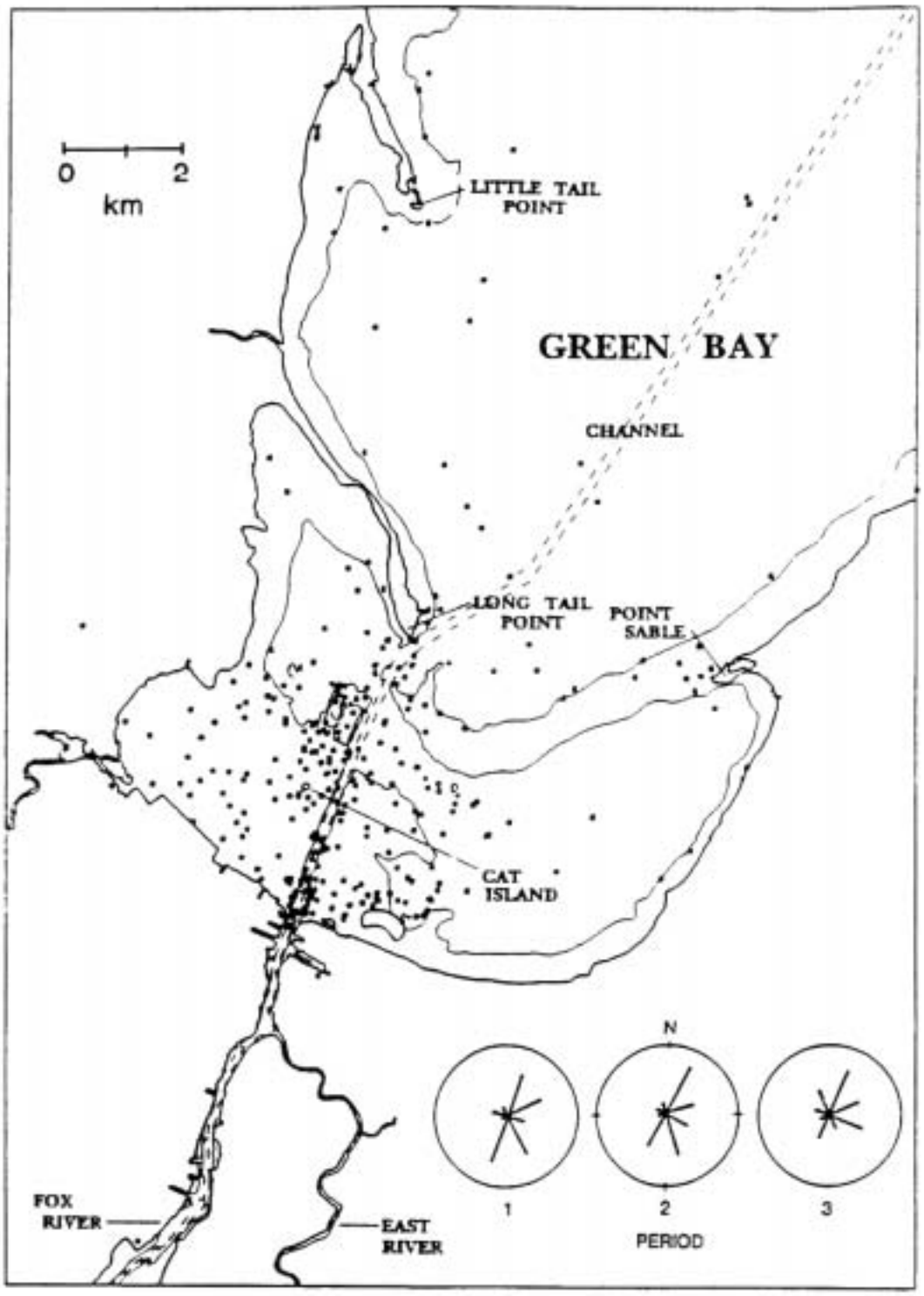


Figure 5-5. Foraging sites of double-crested cormorants from the Cat Island Colony in 1990. Each point represents the foraging destination of a bird tracked from the colony.
Source: Custer and Bunck, 1992.

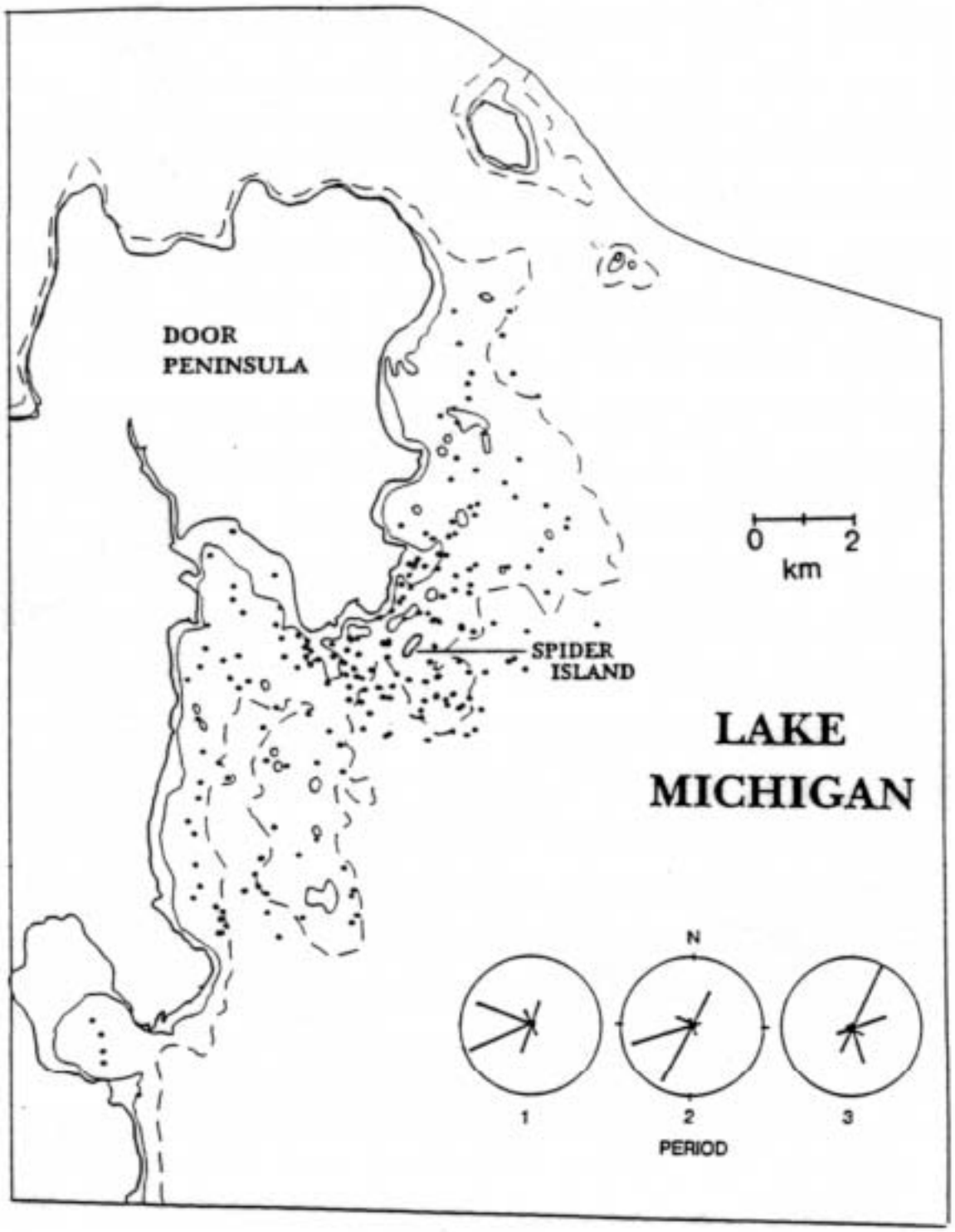


Figure 5-6. Foraging sites of double-crested cormorants from the Spider Island Colony in 1990. Each point represents the foraging destination of a bird tracked from the colony.
Source: Custer and Bunck, 1992.

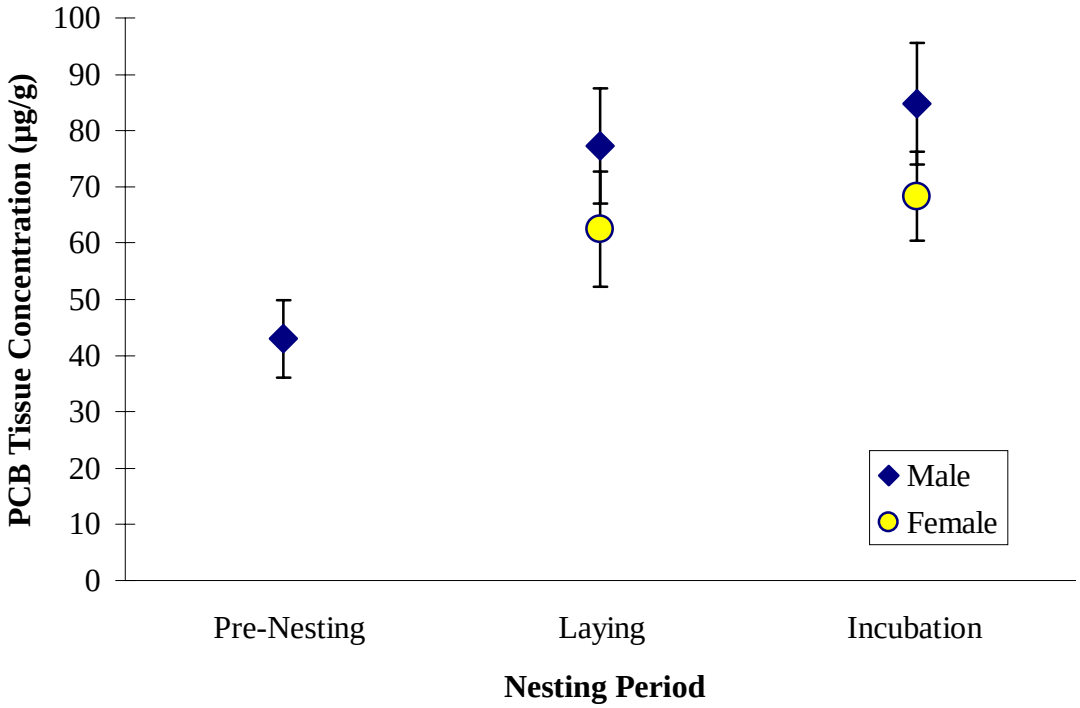


Figure 5-7. Whole-body concentrations of PCBs in cormorants during three phases of the nesting cycle in 1988. Bars equal means plus or minus 1 standard error.
Source: USFWS, 1993.

As already shown, the fish diet of double-crested cormorants in Green Bay is restricted to a relatively small number of forage fish species. These are mainly alewives and yellow perch at the Cat Island colony, and alewives and sculpin at the Spider Island colony. Sampling data show that alewives in Green Bay are contaminated with PCBs (Figure 5-3).

Thus, the PCB pathway documentation for Green Bay double-crested cormorants includes observations that during the incubation and chick-rearing phases of the breeding cycle, adult double-crested cormorants forage in areas of Green Bay that contain PCB-contaminated fish, cormorants ingest Green Bay fish that are contaminated with PCBs, and cormorant PCB tissue residues increase during the breeding season.

5.2.3 Field Studies of Injuries to Green Bay Double-Crested Cormorants

This section describes field studies addressing reproductive malfunctions and physical deformations in Green Bay double-crested cormorants.

Malfunctions in Reproduction

Reproductive success in double-crested cormorants nesting in Green Bay has been compared with that in reference colonies in three studies. In two of these studies, reproductive success was found to be lower in Green Bay colonies.

Ludwig and Ludwig (undated report b). This 1986 study compared hatching success rates in double-crested cormorant colonies in Lakes Huron, Superior, and Michigan, and northern Green Bay (Gravelly, Little Gull, and Snake islands). Hatching success varied from 63% of eggs laid to 74% of eggs laid (Table 5-5). Neither the proportions of eggs that failed to hatch nor hatching success differed between colonies.

Table 5-5
Hatching Success of Double-Crested Cormorants in Great Lakes in 1986[a]

Colony Location	Number of Eggs Studied[b]	Number (%) of Eggs Hatched	Number (%) of Eggs Disappeared	Number (%) of Eggs Dead
All Lake Huron colonies	126	96 (76)	8 (6)	22 (17)
Lake Michigan, Beaver Island	196	142 (72)	23 (12)	31 (16)
Northern Green Bay	173	114 (66)	20 (12)	39 (22)
Lake Superior colonies	65	45 (69)	6 (9)	14 (21)

a. Adapted from data in Ludwig and Ludwig, undated report b.
b. Excludes eggs that were accidentally pierced by parent birds.

Tillitt et al. (1992). This study compared hatching success rates between 1986 and 1988 in 12 double-crested cormorant colonies in Lakes Huron, Michigan, Superior, Ontario, and Winnipegosis and in Green Bay, and investigated the relationships between egg mortality, PCB concentrations, and egg H4IIE activation. Egg mortality varied between 8% and 39%, with the highest rates in Green Bay colonies (Little Gull, Snake, Gravelly, and Spider islands) and the lowest at Lake Winnipegosis. Regression analysis revealed a significant, though relatively modest, positive relationship between total PCB concentrations in eggs and egg mortality ($r = 0.319$, $p = 0.045$). However, when the analysis compared H4IIE bioassay-derived TCDD-eq in eggs with hatching success, the relationship was strengthened ($r = 0.703$, $p = 0.0003$). The H4IIE sample preparation process used in this investigation screened out both dioxins and furans (H4IIE is not sensitive to DDE). The authors of the study concluded that the elevated egg mortality rates and reduced hatching success in the more contaminated colonies were caused by the effects of dioxin-like PCBs.

Larson et al. (1996). This study compared the hatching success of double-crested cormorant eggs at Spider Island in Green Bay in 1988, 1989, and 1990 with the hatching success at Lake Winnipegosis in 1989 and 1990. Hatching success at Spider Island for the three consecutive years was 65.4% (1988), 55.2% (1989), and 57.7% (1990), compared with 75.7% (1989) and 64.1% (1990) at Lake Winnipegosis. Hatching success was significantly greater in larger clutches, and Lake Winnipegosis clutches were, on average, 0.2 eggs larger than Spider Island clutches. However, covariance analysis (in which clutch size was included as a categorical variable) revealed that hatching success was significantly lower at Spider Island in 1989 and 1990, even when clutch size was controlled for. Of 5,759 chicks examined at the Spider Island colony, 0.8% had bill deformities, compared with 0.06% at Lake Winnipegosis. This more than ten-fold difference was reported as statistically significant.

Total PCB concentrations and TCDD-eq were significantly higher in Spider Island eggs (7.8 mg/kg and 138 pg/g, respectively) than in Lake Winnipegosis eggs (1.0 mg/kg and 19 pg/g, respectively). However, *within* the Spider Island colony, neither PCBs nor TCDD-eq were significantly correlated with hatching success or the incidence of deformities among nestlings.

Depredation of seabird nests by gulls following disturbance by observers is a potential problem at many seabird colonies. In general, the greater the disturbance, the greater the opportunity for gulls to depredate eggs. To minimize this effect, the investigators visited the Spider Island colony only after dark (a time when gulls are less active). Nevertheless, the Spider Island colony was visited more frequently (13 visits in 1989) than the Lake Winnipegosis colonies (4 visits in 1989), and the success of the attempt to minimize nest predation by nocturnal visits was not evaluated. Thus, the contribution of observer disturbance to the observed differences in hatching success cannot be determined.

In addition to the above three studies, another study evaluated double-crested cormorant reproduction in Green Bay only (i.e., no reference site data were collected).

Custer et al., in press. During 1994 and 1995, the investigators in this study examined relationships between PCB, DDE, and dieldrin concentrations and hatching success, chick deformity rates, eggshell thickness, and biomarker activity in cormorant eggs from Cat Island in Green Bay. No reference colonies were sampled. Single pipping eggs were removed from each of the study nests. A subset of these eggs were analyzed for chemical contaminants; previous measurements had shown that approximately 85% of the total egg variability in contaminant concentrations in cormorant eggs in Green Bay was between-clutch variation (USFWS, 1993). Measurements made on these eggs included PCB, DDE, and dieldrin concentrations, eggshell thickness, and EROD activity in embryo livers. The fate of the eggs remaining in the study nests was monitored, as was that of the chicks that hatched. Study nests were divided into four groups on the basis of their success: nests that contained eggs with one or more dead embryos, nests that contained one or more infertile eggs, nests in which all the eggs hatched successfully, and nests that contained eggs with deformed embryos.

Table 5-6 shows the total PCB and DDE concentrations among the four nest categories. Two-way ANOVAs performed by the study investigators determined that there were no significant differences among the PCB concentrations in the four nest categories ($p = 0.05$). However, DDE concentrations did differ significantly among the four categories ($p = 0.03$). Mean concentrations of DDE in sample eggs were significantly higher in nests that contained dead embryos than in nests in which all the eggs hatched or contained deformed embryos. Total PCBs in the sample eggs were significantly correlated with EROD activity. The overall egg hatching success on Cat Island in 1994 and 1995 was 68% (1,067 of 1,570 eggs).

Table 5-6
Geometric Mean Total PCB and DDE Concentrations (mg/kg, wet weight) in Four Categories of Cormorant Nests on Cat Island, Green Bay in 1994 and 1995

Nest Category	Number of Nests	DDE	PCBs
Dead embryos	39	3.9	11.4
Infertile eggs	5	2.8	13.6
All eggs hatched	30	2.8	12.1
Deformed embryos	6	2.2	10.2

Source: Custer et al., in press.

The study authors conducted a series of logistic regressions to evaluate whether DDE, PCBs, dieldrin, or eggshell thickness was associated with differences in percent hatching success of the cormorant eggs. Only DDE and eggshell thickness were found to have significant associations ($p < 0.002$) and < 0.008, respectively). Neither total PCBs nor dieldrin was found to have significant associations with hatching success ($p < 0.84$ and < 0.29, respectively). However, egg PCB concentrations were significantly negatively correlated with both egg volume ($r = -0.39$, $p < 0.001$) and embryo weight ($r = -0.28$, $p < 0.04$). DDE was not significantly correlated with egg volume and egg weight.

Although hatching success was significantly correlated with eggshell thickness and significantly negatively correlated with DDE concentration, these two variables explained relatively little of the total variability in hatching success (2.2% and 13%, respectively). The study authors concluded that DDE may be reducing the hatching success of only the most highly contaminated eggs, and that other factors may be responsible for the majority of the egg failure observed at the colony.

Physical Deformations

A number of studies have reported on physical deformations among Green Bay double-crested cormorants.

Langenberg (1990). In this companion study to Larson et al. (1996), the author examined 183 late-term cormorant eggs from Spider Island and 125 from Lake Winnipegosis: 95% of the Spider Island eggs contained embryos, of which 18.3% were dead, and 98% of the Lake Winnipegosis eggs contained embryos, of which 11.4% were dead. Our analysis of these data found that the differences were not statistically significant ($\chi^2 = 2.7$, 1df, $p > 0.25$). Two examples of cross bills were found, both in Lake Winnipegosis embryos. On Spider Island, 10.9% of the embryos examined had edema, similar to the 16.3% of Lake Winnipegosis embryos. Seven of the Spider Island embryos had petechial hemorrhages; none were found in the Lake Winnipegosis embryos. However, because hemorrhaging in several cases was noted only after handling, Langenberg concluded that these were a result of the examination process rather than toxicological action.

Fox et al. (1991). In this analysis of deformity rates among cormorant nestlings throughout the Great Lakes between 1979 and 1987, 31,168 chicks from 42 colonies were examined. The overall rate of head and bill deformities was 0.22%. However, the local rate varied, and the highest rate was found in Green Bay (60 of 11,520 chicks, 0.52%). The rates found in other areas were 0.03% (Lake Ontario), 0.05% (Lake Superior), 0.006% (Alberta and Saskatchewan), and 0.02% (Lake of the Woods and Lake Nipigon). The Green Bay deformity rate was significantly higher than the rates at the Lake of the Woods and Lake Nipigon, and at prairie colonies in Alberta and Saskatchewan. Head and bill defects were found in 8 of 11 (73%) Green Bay colonies, but in only 6% of reference colonies.

Yamashita et al. (1993). In this study carried out in 1988, the investigators collected late-term, incubated cormorant eggs from Little Gull Island in Green Bay and elsewhere in the Great Lakes (including from colonies not exposed to point source releases of PCBs). Eggs were examined and separated into four categories: live normal, infertile, containing a deformed embryo, and not incubated. Of the 41 Green Bay eggs examined, 26 were fertile; 78% of these contained normal young and 31% contained deformed embryos (compared with about 90% and 6%, respectively, in eggs collected elsewhere). The total PCB concentrations in Green Bay eggs were 7.3 mg/kg wet weight (live normal), 7.3 mg/kg wet weight (infertile), and 6.6 mg/kg wet weight (deformed). Total PCB concentrations in eggs from Lake Superior and the North Channel (colonies unlikely to be affected by point source releases of PCBs) varied from 3.6 to 7.3.

Larson et al. (1996). The incidences of bill deformities in cormorant nestlings were compared between Spider Island (1988, 1989, and 1990) and Lake Winnipegosis (1989, 1990). At Spider Island, 5,759 chicks were examined, and approximately 24,736 were examined at Coffee Island, Coffee Island Reef, Bachelor's Island, Sugar Island Reef, and Hay Island Reef in Lake Winnipegosis. Bill defects were significantly more frequent ($p < 0.001$) at Spider Island (0.7%) than at Lake Winnipegosis (0.06%).

*Ludwig et al. (1996).*This study was based on measurements taken in several Great Lakes cormorant colonies between 1987 and 1991. In Green Bay, 24.8% of 660 dead eggs (eggs that

were dead in the nest) contained deformed embryos. Of these, 65% had subcutaneous edema and hemorrhaging, and 18.9% had bill defects. These data are reported in Table 1 of Ludwig et al. (1996). Our statistical tests on the raw data in this table indicated significant differences in deformity rates between colonies ($\chi^2 = 31.8$, 6 df, $p < 0.001$). Our tests also showed that the embryo deformity rate in dead eggs in Green Bay (24.8%) was significantly higher than that found in colonies in the three areas that were least likely to be exposed to point source releases of PCBs: southeast Lake Superior (11.3%, $\chi^2 = 23.0$, 1 df, $p < 0.001$); Georgian Bay on northern Lake Huron (15.5%, $\chi^2 = 8.5$, 1 df, $p < 0.02$); and the North Channel of Lake Huron (18.7%, $\chi^2 = 6.6$, 1 df, $p < 0.05$). Of 315 live eggs from Green Bay, 14.3% contained deformed embryos, compared with 4% at Lake Winnipegosis. The proportions of the various types of embryo deformities found in the Green Bay live eggs were not reported. Of 7,975 Green Bay cormorant nestlings, 0.6% were deformed. These had mainly crossed bills (53% of deformities) and dwarfed appendages (20.4% of deformities).

Ryckman et al. (in press). In this study of organochlorine contamination and bill defects among cormorants nesting in the Canadian Great Lakes, no significant associations were found between regional rates of dill deformities and total PCB concentrations in eggs.

Custer et al. (in press). In addition to the results presented previously, this study also investigated relationships between deformity rates in chicks of double-crested cormorants and organochlorine residues. Eggs from nests in which one or more embryos were deformed did not have significantly higher concentrations of either PCBs or DDE than eggs from nests in which no deformed young were found. Custer et al. also reported that the frequency of bill deformities among nestlings at Cat Island in 1994-1995 (0%; 0 of 632) was generally lower than those reported from cormorant colonies in northern Green Bay during the period 1979-1990 (0.6%-0.7%). This is in spite of the fact that Cat Island cormorants had higher egg PCB concentrations [Cat Island 1994-1995: mean of 13.6 mg/kg, wet weight; Spider Island 1988 and 1989: 5.3 and 7.7 mg/kg wet weight, respectively (Tillitt et al., 1992; Larson et al., 1996)]. However, the deformity rates and egg PCB concentrations in northern Green Bay cormorant colonies in the years in which the Custer et al. study was performed at Cat Island are not known.

5.2.4 Data Evaluation

Evidence of Adverse Effects in Green Bay Double-Crested Cormorants

Reproductive malfunctions. The evidence that Green Bay cormorants have suffered adverse reproductive effects is strong. Two independent studies (Tillitt et al., 1992; Larson et al., 1996) demonstrated that hatching success rates are significantly lower in Green Bay nests than in control areas. One other study (Ludwig and Ludwig, undated report b) attempted to compare nesting success of cormorants in Green Bay with reference sites. Analyses of the data presented in that study showed no significant differences between hatching success in Green Bay and in other sites.

In addition to the field studies described above, one study, Powell et al. (1997), attempted to reproduce the impaired hatching success seen in the field by injecting cormorant eggs in the laboratory with a PCB congener. They injected cormorant eggs collected from Lake Winnipegosis (a site where cormorants are not exposed to point source releases of PCBs) with doses of PCB 126 and an extract derived from Green Bay cormorant eggs. The authors found that injections of PCB 126 significantly reduced hatching success of Lake Winnipegosis eggs but only at doses an order of magnitude greater than the highest concentrations of PCB 126 that have been found in Green Bay cormorant eggs. However, it should be noted that Powell et al. did not inject the Lake Winnipegosis eggs with the mixture of congeners found in Green Bay eggs. Also, there is uncertainty regarding how closely eggs injected with contaminants or extracts mirror the "natural" uptake and effects of contaminants in the field. Overall, however, the Powell et al. study does not support PCBs as the cause of the reduced hatching success observed among assessment area double-crested cormorants.

Physical deformations. The strength of the evidence that Green Bay cormorants have deformity rates that are elevated with respect to background is strong, depending on which deformity is addressed and what background level is assumed.

Studies have shown that crossed bills have occurred among Green Bay cormorant chicks (Fox et al., 1991; Larson et al., 1996; Ludwig et al., 1996). The rate of bill deformities found in Green Bay is substantially higher (by a factor of about 10) than that observed at reference sites. The background rate of bill deformities that is typically observed is usually less than 0.1% (Fox et al., 1991; Ryckman et al., in press). However, the evidence that such a low rate is representative of all appropriate reference colonies is not entirely unambiguous. Ross and Weseloh (1988) measured a large degree of spatial variability in bill deformity rates among Lake Winnipegosis colonies. The Sugar Island colony in 1988 had a rate of 3.9%, which is much higher than the highest deformity rate ever measured in Green Bay, or any other location. It should be noted, however, that the Ross and Weseloh study is the only study, thus far, that has found such high rates of deformities in cormorants from areas not affected by point sources of contaminants. Indeed, in their research on Lake Winnipegosis, Larson et al. (1996) included Sugar Island among their sampling locations and still found that the overall Lake Winnipegosis deformity rate was less than 0.1%. The relevance of the Ross and Weseloh study is, therefore, uncertain. All studies that have assessed bill deformity rates in both Green Bay and reference colonies have found higher rates in Green Bay.

Increased incidences of edema of the head and neck (which constitutes the majority of the deformities reported in double-crested cormorants) and hemorrhaging are less certain than crossed bills. Ludwig et al. (1996) found that 16.2% of dead eggs from Green Bay had embryos with edema (mainly of the head and neck), and only 6% of live nestlings showed hemorrhaging. These data indicate that the deformity rate among live chicks may underestimate the true population rate (since many deformed embryos may die before hatching). However, Langenberg (1990) examined live eggs from Green Bay and was unable to find abnormal occurrences of

edema. She concluded that any hemorrhaging that she recorded was an artifact of her handling the embryos.

In conclusion, bill deformity rates among cormorant embryos and nestlings in the assessment area have exceeded background rates. However, the occurrence of other types of deformities is not as conclusive.

Evidence of PCB Effects

This section evaluates the evidence that PCBs are responsible for the observed adverse effects on reproduction and bill deformity rates. Chapter 4 presented data showing that total PCB and PCB congener concentrations measured in Green Bay cormorant eggs are at or above concentrations shown to cause avian toxicity in literature studies. However, because PCB concentrations can be correlated with concentrations of other lipophilic compounds (Bosveld, 1995), the likelihood of the adverse effects being caused by other contaminants must also be evaluated.

Reproductive malfunctions. The four main groups of candidate contaminants that potentially could cause the effects seen in the Green Bay cormorants are PCDDs, polychlorinated dibenzo-furans (PCDFs), DDE, and PCBs.

Using the data presented in Yamashita et al. (1993), it is possible to evaluate the relative contributions of PCDD and PCDF to total dioxin-like toxicity. The analysis in Table 5-7 shows that the contributions to total toxicity by TCDD and TCDF are less than 5%. A similar result was obtained in the Kubiak et al. (1989) study of Forster's terns on Green Bay, where PCDDs accounted for less than 10% of total toxicity (no PCDFs were found in the tern eggs) and PCBs for more than 90%. Also, Tillitt et al. (1992) eliminated PCDD and PCDF residues when preparing Green Bay cormorant egg samples for H4IIE analysis and found significant correlations between the responses elicited by the extract and hatching success at Great Lakes colonies. These results indicate that PCDDs and PCDFs are unlikely to be important contributors to the adverse effects reported in the Green Bay cormorants.

Table 5-7
Percent Contributions to Total TCDD-EQ by PCB, TCDD, and TCDF Congeners in Green Bay Double-Crested Cormorant Eggs

TEFs Used to Estimate Percent Contribution	Percent Contribution[b]							
	PCB 77	PCB 105	PCB 118	PCB 126	PCB 156	2378 TCDD	12378 TCDD	2378 TCDF
WHO Avian TEFs[a]	47.2	2.6	<1	44.1	<1	2.4	<1	<1

a. Van den Berg et al., 1998.
b. Original data from Yamashita et al., 1993.

Concentrations of PCBs and DDE in Green Bay cormorant eggs are correlated (Custer et al., in press, $r = 0.53$, 73 df, $p < 0.001$). Also, although little research has been carried out on the effects of DDE on avian embryos at levels of exposure below those known to result in eggshell thinning and breakage, there is evidence from a field study of common terns that DDE concentrations of between about 3 and 7 mg/kg wet weight may result in embryo mortality (Fox, 1976). Thus, DDE may exert effects on embryo mortality other than those associated with eggshell breakage that are similar to those that may be caused by exposure to PCBs. Possible mechanisms for this are changes in shell microstructure that are associated with comparatively low levels of thinning, with consequent disruptions of gaseous transfer, or with direct embryotoxic effects (Fox, 1976).

The three studies that have attempted to rigorously address the potential effects of contaminants on cormorant hatching success in the field are Tillitt et al. (1992), Larson et al. (1996), and Custer et al. (in press). Tillitt et al. concluded that PCBs explained much of the observed variability in hatching success between Great Lakes cormorant colonies and were responsible for the reduced hatching success seen in Green Bay. This conclusion was based on the relationships between mean PCBs and H4IIE results and hatching success *between colonies*. In contrast, both the Larson et al. and the Custer et al. studies suggested that PCBs did not explain differences in hatching success among Green Bay cormorants. The conclusions were based on the lack of significant correlations when PCB concentrations were compared with individual nest reproductive success *within a colony*. In addition, Powell et al. (1997) was unable to reproduce embryo mortality among cormorants in the laboratory when injecting eggs with doses of PCB 126 comparable to those seen in the assessment area.

The discrepancy between the results of these studies may be at least partly a function of the different study approaches. Both Larson et al. (1996) and Custer et al. (in press) used the sample egg technique, in which the reproductive success of individual nests within a colony was measured and compared with the contaminant concentrations in an egg removed from the same nest. Because many factors other than contaminants affect the reproductive success of individual nests, such as nest abandonment, predation, accidental egg breakage, and parental experience, individual nests within a colony have a high degree of variability that is not expected to be explained by contaminant concentrations. Indeed, Custer et al. (in press) found that DDE explained only 13% of the variability in individual nest success. Therefore, in this approach the power to detect effects of contaminants on the inherently variable success of individual nests is low. In contrast, Tillitt et al. (1992) compared the mean reproductive success across colonies with the mean contaminant concentrations in eggs taken from the colony. Comparing mean colony success with mean colony contaminant concentrations across different colonies reduces the variability in the reproductive success data and allows for a greater ability to detect the effects of contaminants on reproductive success. However, it should be pointed out that Tillitt et al. (1992) did not compare DDE concentrations and reproductive success between colonies. Therefore, because of the different study approaches, the findings of Larson et al. (1996) and Custer et al. (in press) that PCBs are not correlated with reduced hatching success within the Green Bay colony are not inconsistent with the finding of Tillitt et al. (1992) that PCBs and H4IIE are correlated with mean hatching

success across colonies and that the Green Bay colony had the highest PCBs and H4IIE activity and lowest mean reproductive success.

Custer et al. also indicated that in Green Bay cormorants DDE concentrations appeared to explain a greater percentage of the variability in hatching success than PCBs. However, such a small component of the total variance in hatching success is apparently explained by contaminants that detecting the effects of individual contaminants would be difficult. The fact that a significant effect on hatching success was found by Custer et al. for DDE, but not for PCBs, might only reflect this difficulty rather than the likelihood that only DDE was affecting the cormorants. Also, it cannot be definitely concluded from the data in Custer et al. that PCBs had no effect on hatching success, since the only measure of PCB contamination that was analyzed was total PCBs. Using TCDD-eq in the analysis may also have strengthened the relationship between PCBs and hatching success, as was found in the Tillitt et al. (1992) study.

Furthermore, Custer et al. (in press) found significant negative correlations between egg PCB concentrations and egg volume and embryo weight, indicating that PCBs were exerting some effects on the breeding biology of Cat Island cormorants in 1994-1995. Although they did not find a negative relationship between PCBs and hatching success that was statistically significant at $p < 0.05$, the probability of the correlation that they did find being due to chance was 0.13.

Overall, the evidence shows that exposure to PCBs may have resulted in reduced hatching success among Green Bay cormorants. However, the Custer et al. (in press) study shows that the effects observed in the assessment area are unlikely to be due to PCBs alone and that DDE has contributed to the adverse effects.

Physical deformations. PCBs have been shown in controlled experiments to cause deformations in avian embryos. These have included deformations of the head and bill and legs. However, DDE is not known to cause such deformities in avian embryos. The other candidate contaminants that could cause such deformities (PCDD and PCDF) do not occur at concentrations that could contribute significantly to the deformity rates observed among Green Bay cormorants (see previous discussion).

Summary and Conclusions

The data reviewed in this report indicate that exposure to elevated concentrations of PCBs has most likely resulted in adverse effects to double-crested cormorants in Green Bay, including reduced reproductive success and embryonic deformations. However, the evidence for this is not as conclusive as that for Forster's terns. Also, it is likely that other contaminants complicate the attribution of effects.

5.3 BLACK-CROWNED NIGHT HERON

Black crowned night-herons are opportunistic feeders. Their diet often consists mainly of fish and other aquatic organisms, although they also eat terrestrial invertebrates and the nestlings of other colonial birds (Cramp and Simmons, 1977).

Three studies summarized in Chapter 4 of this report showed that black-crowned night herons in the assessment area have been exposed to PCBs: Heinz et al. (1985), Rattner et al. (1993), and Custer and Custer (1995).

Two studies investigated adverse effects in Green Bay night herons.

Hoffman et al. (1993). In this study, five pipping eggs were collected from the colony on the CDF in 1984, and the morphologies of their chicks were compared with others from a captive control colony at Patuxent Wildlife Research Center (PWRC) in Maryland. The two groups did not differ in egg or embryo weights. The Green Bay chicks had 36% larger livers than the PWRC chicks, but this difference was not significant. However, Green Bay chicks had significantly higher liver to body weight ratios than the PWRC chicks.

This study also investigated biomarker activity in the livers of the two groups of embryos. It found that AHH activity was significantly higher in the livers of the Green Bay chicks by a factor of three. No PCB concentration measurements were carried out to determine if the morphological and biomarker differences between the two groups of chicks were associated with differences in contaminant loads.

Rattner et al. (1993). In this study, PCB concentrations and biomarker activity were measured in black-crowned night heron chicks from Cat Island, a reference site in Virginia, and two islands in San Francisco Bay. The Green Bay chicks had the highest levels of biomarker activity (AHH, EROD, BROD, ECOD, CYP1A, and CYP2B) and the highest PCB concentrations (9.32 mg/kg wet weight, with a range of 2.4-53 mg/kg wet weight). The Green Bay PCB concentrations were significantly greater than those found in all of the other colonies. No morphological abnormalities were reported.

These studies show that black-crowned night herons in Green Bay have been exposed to PCBs at levels that exceed background concentrations. One study (Hoffman et al., 1993) also suggests that Green Bay black-crowned night herons may have been injured (enlarged livers).

5.4 TREE SWALLOW

Tree swallows are insectivorous birds that feed on the emerging adult life stages of aquatic insects. Thus, because of their diet, tree swallows nesting close to the Lower Fox River and Green

Bay might be expected to be exposed to PCBs. Only one study of contaminants and breeding success has been performed on this species in the assessment area (Custer et al., 1998). This study showed that pipping hatchlings and nestlings of tree swallows nesting close to the Lower Fox River and inner Green Bay had significantly higher PCB concentrations than pipping hatchlings and nestlings from reference areas.

The breeding success of the Green Bay and Lower Fox River tree swallows was not significantly different from that of tree swallows at the reference areas. Nor were any embryo or nestling differences in weight or body condition found. No deformities were observed. These data suggest that the PCBs measured in the tree swallow hatchlings and nestlings in the assessment area were not causing adverse effects.

5.5 RED-BREASTED MERGANSER

White and Cromartie (1977) and Haseltine et al. (1981) showed that in the 1970s PCB concentrations in red-breasted merganser eggs on islands off the Door Peninsula were high. Heinz et al. (1983), a companion study to Haseltine et al. (1981), found, however, that high PCB residues in 1977 and 1978 were not correlated with rates of nest desertion, hatching success, or duckling production. Also, Heinz et al. (1994) found no significant difference between merganser hatching success in 1977-1978 and 1990, despite egg PCB concentrations having decreased by 60%. Thus, the available data do not indicate that the elevated PCB concentrations in Green Bay red-breasted merganser eggs in the late 1970s were affecting reproduction in this species.

5.6 BALD EAGLES

5.6.1 Status and Ecology in the Lower Fox River and Green Bay

Bald eagle population trends in the Great Lakes can be divided into two phases: mid-century declines and post-1960s resurgence.

Midcentury population declines. In the middle of this century, bald eagle populations throughout the contiguous United States and much of Canada underwent drastic reductions. A chronology of these population declines was reported by Nisbet (1989):

> Reproductive impairment in the bald eagle was first reported in Florida in 1947 (Broley, 1958) and became widespread during the 1950s and 1960s (Sprunt, 1963; Sprunt and Ligas, 1966; Stickel et al., 1966; Postupalsky, 1971; Grier, 1972; Wiemeyer et al., 1972, 1984; Sprunt et al., 1973). By 1970, a number of local populations in the lower 48 states of the USA and in southern Canada had been markedly reduced or extirpated (Broley, 1958; Howell, 1963; Postupalsky, 1971;

> Grier, 1972; Sprunt et al., 1973; USDI, 1974; Kiff, 1980); populations in Alaska and parts of western and northern Canada were generally unaffected. . . . Several studies have shown the inter-relationships between eggshell-thinning, reproductive impairment, populations declines, and levels of contamination with DDE and other organochlorines (Postupalsky, 1971; Wiemeyer et al., 1972, 1984; Sprunt et al., 1973).

In the Great Lakes, bald eagles were extirpated from coastal areas and anadromous runs of Lakes Huron, Michigan (including Green Bay), Ontario, and Superior and nearly extirpated from Lake Erie by the late 1960s (Bowerman, 1993).

Post-1960s population resurgence. Since the mid-1970s, when the use of DDT, PCBs, and other organochlorine compounds was banned in North America, bald eagles have increased in number. The lessening of the eggshell-thinning effects of DDT's metabolite, p,p'-DDE, has been a major reason for the current resurgence of bald eagle populations in temperate North America (Grier, 1982; Postupalsky, 1985; Colborn, 1991; Best et al., 1994; Bowerman et al., 1995). The number of bald eagle breeding pairs within 8.0 km of the Great Lakes coasts increased from 26 in 1977 to 134 in 1993. Furthermore, the reproductive productivity of these birds increased from 0.23 young per occupied nest in 1977 to 0.87 in 1993 (Bowerman, 1993). Bald eagles breeding within 8.0 km of the Lake Michigan coast or along streams open to Great Lakes fish runs also increased over this period, from 2 pairs in 1977 to 28 pairs in 1993. The productivity of these birds increased from 0.0 young per occupied nest to a high of 0.89 in 1987, but was only 0.46 in 1993 (Bowerman, 1993).

Annual monitoring data collected by staff of Wisconsin DNR and by S. Postupalsky and W. Bowerman for the State of Michigan (M. Meyer, Wisconsin DNR; D. Best, USFWS, personal communication, March 1999) show that between 1974 and 1986 bald eagle nesting numbers on Green Bay and the eastern side of the Door Peninsula were stable at between one and two pairs (Figure 5-8). A rapid increase in nesting numbers began in 1987, and by 1997 there were 14 nesting pairs. The number of breeding pairs of eagles nesting along the Lower Fox River went from one in 1986 to three in 1994 to two since 1995. The distribution of the Green Bay and Lower Fox River nest sites is shown in Figure 5-9.

Bald eagles arrive back on their nesting territories in the assessment area in February, and the young fledge between early June and July. Depending on ice conditions, bald eagles remain in the assessment area during the winter; up to 12 have been recorded in December on the Lower Fox River (Howe et al., 1993). Thus, breeding bald eagles spend a substantial part of the year in the assessment area.

In August 1989, bald eagles were listed as threatened by the State of Wisconsin. This designation was removed in August 1997. They are currently listed as threatened by the Service.

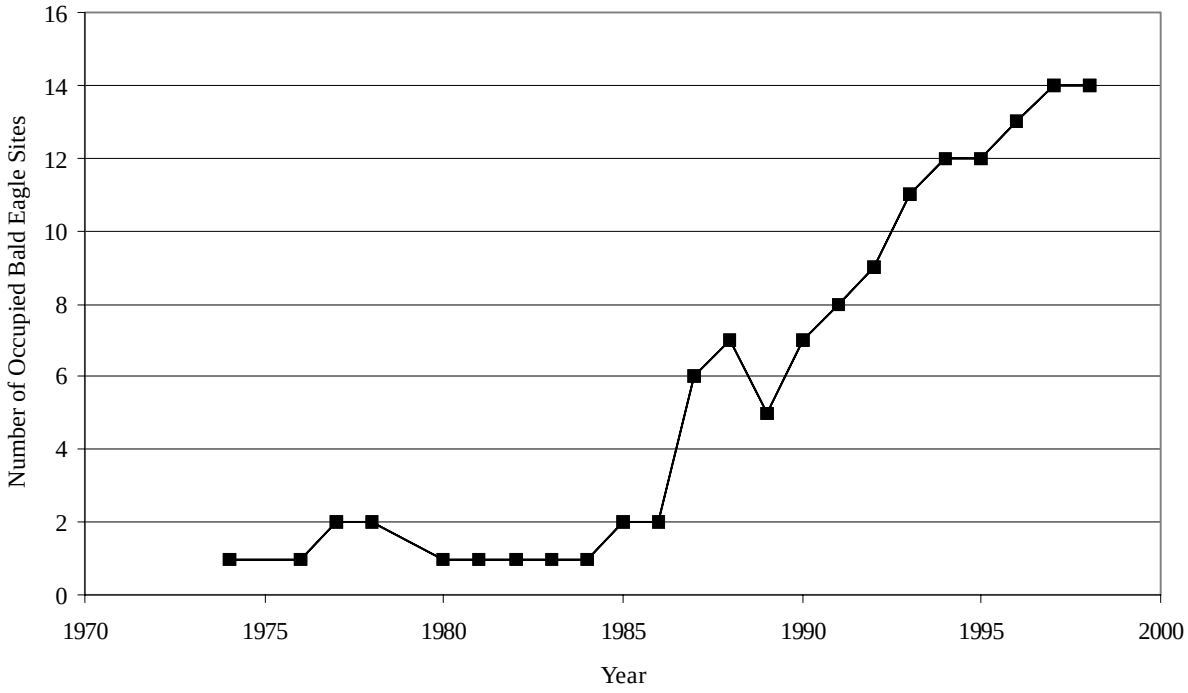


Figure 5-8. Numbers of occupied bald eagle nesting sites on Green Bay.

5.6.2 Diet and Foraging Areas around Green Bay

Diet. There are two studies that describe bald eagle diet in the assessment area. Bath (1991) quantified prey class percentages at a nest at Kaukauna on the Lower Fox River during the pre-hatching period. Dykstra and Meyer (1996) collected prey data from the entire nestling period from nests at Toft Point and Little Tail Point. The results of these studies were combined by W. Bowerman (personal communication, Lake Superior State University, April 1998) and are presented in Table 5-8. Data in Dykstra and Meyer (1996) from a nest at Blueberry Island were not used in this analysis since the nest was located along the Menominee River and might not be representative of eagles foraging around Green Bay. Also excluded were data collected by Dykstra and Meyer at Moss Lake since prey data were collected there only during the final 6 weeks of the nestling period, and prey species use changes over the nestling period (Dunstan and Harper, 1975). Prey items that were not identified in these studies were assigned identities based on the proportion of prey items that were identified by either class or species. Based on these observational and prey remains data, bald eagle prey composition on a frequency basis at Green Bay nests comprises approximately 74% fish, 23% avian prey, and 2% mammals (Table 5-8), which is similar to the diet composition of bald eagles elsewhere in the Great Lakes (Bowerman, 1993).

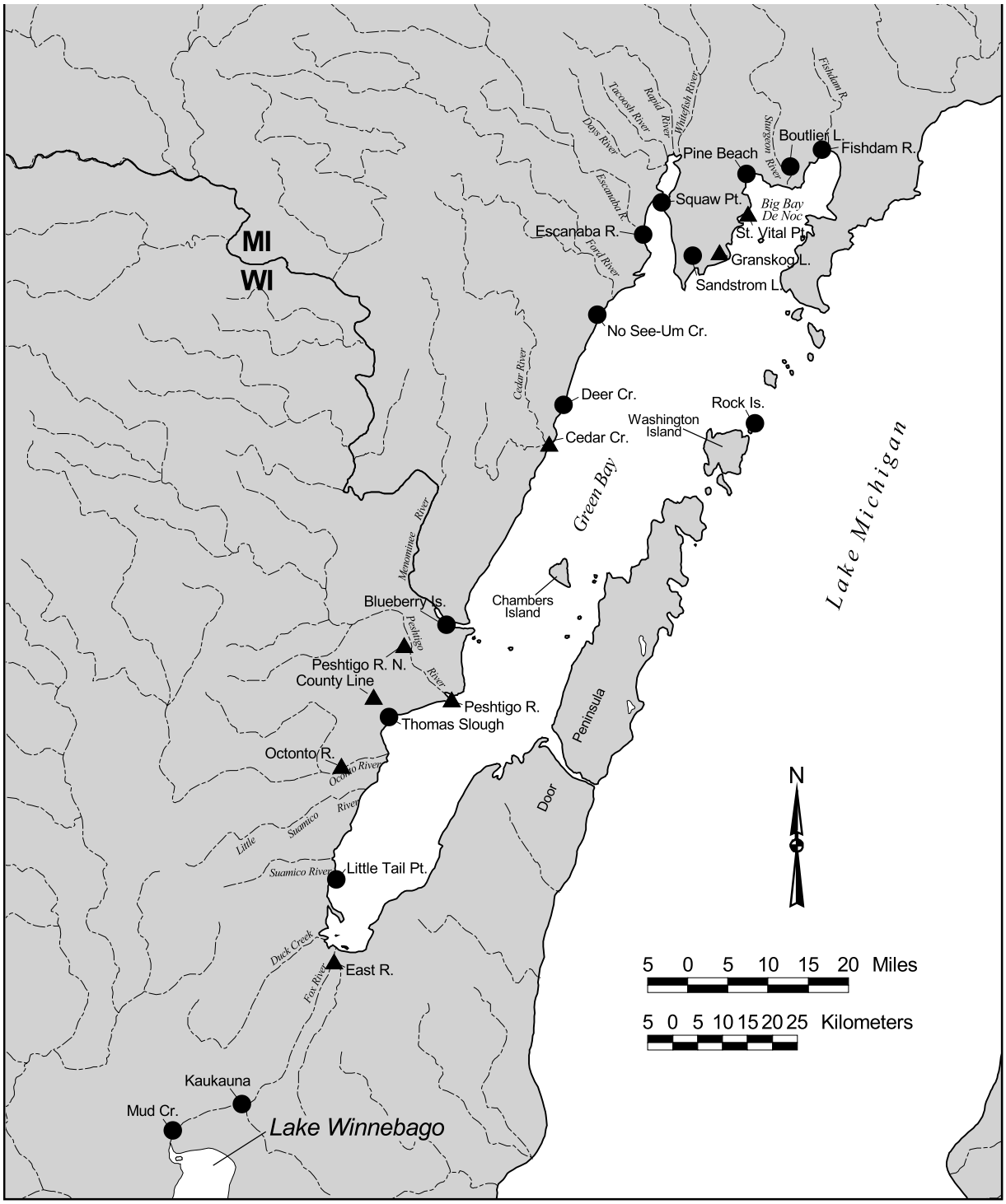


Figure 5-9. Distribution of bald eagle nest sites in Green Bay and the Lower Fox River.
Circles are sites occupied in 1998. Triangles are sites not occupied in 1998 but occupied in previous years.

Table 5-8
Prey of Bald Eagles Nesting on Green Bay and the Lower Fox River Based on Prey Remains for the Breeding Period, and Observations during the Pre-Hatch Period

Class/Species	N	Percent of Total
Fish		
Sucker	23	13.8
Bullhead	30	18.0
Northern pike	28	16.8
Bass	3	1.8
Other centrarchids	6	3.6
Walleye	2	1.2
Bowfin	11	6.6
Carp	14	8.4
Freshwater drum	2	1.2
Alewife	1	0.6
Gizzard shad	4	2.4
Subtotal	**124**	**74.4**
Birds		
Herring and ring-billed gulls	15	9.0
Mergansers	2	1.2
Other ducks	4	2.4
Double-crested cormorant	1	0.6
Common raven	1	0.6
American crow	2	1.2
Unknown heron	1	0.6
Other birds	12	7.2
Subtotal	**38**	**22.8**
Mammals		
Muskrat	2	1.2
White-tailed deer	1	0.6
Red fox	1	0.6
Subtotal	**4**	**2.4**
Reptiles		
Unknown turtle	1	0.6
Subtotal	**1**	**0.6**
Total	**167**	**100.2**

Sources: Analysis of data in Bath, 1991 and Dykstra and Meyer, 1996, by W. Bowerman, Lake Superior State University, personal communication, April 1998.

Foraging areas. Observations of bald eagles nesting at Kaukauna on the Lower Fox River showed that during February through May 1991 the adults foraged along the Fox River and generally within 0.5 km of the nest, but ranged up to 3.0 km (Bath, 1991). No data exist that allow the determination of foraging ranges at Green Bay nests; however, most previous studies of bald eagle foraging assumed a radius of 8.0 km from the nest as the likely foraging area (Bowerman et al., 1995).

5.6.3 Ecological Traits that Could Affect PCB Exposure of Bald Eagles

Bald eagles nesting around Green Bay and along the Lower Fox River have a high potential for exposure to PCBs. First, they are likely to be either year-round residents in the assessment area or present for a substantial part of the year. Second, birds nesting on the Green Bay or Lower Fox River shorelines are likely to obtain much of their food from the contaminated aquatic systems, and even those birds nesting farther inland (up to about 8 km) are also likely to be dietarily exposed to assessment area contaminants. Lastly, bald eagles are tertiary predators that include high trophic level predatory birds and fish within their diet (Table 5-8). Because of these characteristics, bald eagles are potentially liable to be exposed to high levels of lipophilic compounds that bioaccumulate through trophic levels, such as PCBs.

5.6.4 Bald Eagle Exposure Pathways

The main exposure route through which bald eagles that nest on Green Bay and the Lower Fox River are exposed to PCBs is the dietary pathway. In this section, the following questions are addressed: Do the prey species that constitute the diet of the bald eagle in the assessment area have elevated PCB concentrations, and do bald eagle tissue analyses indicate that eagles are exposed to PCBs?

PCBs in Bald Eagle Prey

Many of the fish and bird species known to be eaten by bald eagles nesting in Green Bay are contaminated with PCBs (Table 5-9). Data on PCB concentrations in alewife, gizzard shad, and rainbow smelt described in Section 5.1 show that these species, also, are contaminated with PCBs in the assessment area. These data show that bald eagles in the assessment area are exposed to PCBs in their diets.

PCBs in Bald Eagle Tissues

Table 5-10 shows the total PCB concentrations in bald eagle eggs from nests around Green Bay from 1986 (when the earliest sample was collected) until 1997 (data from Wisconsin DNR and USFWS contaminants databases provided by M. Meyer, Wisconsin DNR, and D. Best, USFWS).

Table 5-9
PCB Concentrations in Potential Bald Eagle Prey from the Assessment Area

Prey Species	Date	Locality	Tissue	Sample Size	PCB Concentrations (mg/kg wet weight) Ranges (where known) in Parentheses	Reference
Mallard	1985-1986	Lower Fox River	Muscle, skin, and fat	55	0.4[a] (0-1.5)	Amundson, undated report
Double-crested cormorant	1987-1988	Green Bay	Whole body	6	84.8[a]	USFWS, 1993
Sucker	1979	Green Bay	Whole body	4	2.6[a] (1.7-4.4)	Wisconsin DNR, 1971-1995
Bullhead	1979	Green Bay	Whole body	1	2.1	Wisconsin DNR, 1971-1995
Northern pike	1979	Green Bay	Whole body	1	10.5	Wisconsin DNR, 1971-1995
Carp	1979-1989	Green Bay	Whole body	116	4.0[a] (0.04-10.5)	Wisconsin DNR, 1971-1995; Connolly et al., 1992

a. Mean of measurements.

These data show that bald eagles nesting in the assessment area have been exposed to PCBs. DDE concentrations in bald eagles are also shown in Table 5-10 and will be discussed in Section 5.6.6.

Figure 5-10 compares the Green Bay 1986-1997 egg PCB and DDE concentrations with concentrations in eggs from inland Michigan and inland Wisconsin. PCB and DDE concentrations are significantly higher in the Green Bay eggs than in eggs from nests in inland Michigan ($t = 5.9$, $p < 0.001$, and $t = 4.9$, $p < 0.001$, respectively) and Wisconsin ($t = 6.12$, $p < 0.001$, and $t = 4.4$, $p < 0.001$, respectively).

Table 5-11 shows the total PCB concentrations in bald eagle nestling blood plasma from the assessment area from 1987 to 1995 (Dykstra and Meyer, 1996) and from inland Michigan. Although no statistical tests were carried out by Dykstra and Meyer, the plasma levels in assessment area chicks exceed those in chicks from inland Michigan.

The above data confirm that assessment area bald eagles are likely to forage in areas that contain contaminated fish and wildlife, and that their prey has highly elevated PCB concentrations. They

Table 5-10
PCB and DDE Concentrations (mg/kg fresh wet weight) in Bald Eagle Eggs from Green Bay and the Lower Fox River

Breeding Area/State/Number	Year	Total PCBs	DDE
Green Bay			
Peshtigo River/WI/MT-07	1987	13.0	2.4
Boutlier Lake/MI/DE-15	1986	55.3[a]	30.2[a]
Fishdam River/MI/DE-17	1990	26.6[a]	10.1[a]
Fishdam River/MI/DE-17	1991	27.2	7.4
Peshtigo River/WI/MT-07	1991	56.5[a]	12.0[a]
Peshtigo River/WI/MT-16	1992	66.6[a]	14.7[a]
Peshtigo River/WI	1995	120.0	21.0
Fishdam River/MI/DE-17	1992	28.5[a]	11[a]
Squaw Point/MI/DE-18	1992	28.7	12.3
Squaw Point/MI/DE-18	1993	42.3[b]	12.9[b]
Moss Lake/MI/DE-09	1994	24.3	4.3
St. Vital's Point/MI/DE-20	1997	22.4	8.3
Oconto/WI	1997	88.0	16.0
Fox River			
Kaukauna Lower Fox River/OU-1	1990	36.0	1.1
Mean of Green Bay eggs[c]	n = 13	46.1	12.5

a. Mean of two eggs.
b. Mean of three eggs.
c. Multiple eggs from the same breeding area in a given year averaged prior to determining mean.

Sources: Dykstra and Meyer, 1996; Wisconsin and USFWS contaminants monitoring databases.

also show that Green Bay bald eagle eggs and plasma are contaminated with PCBs. Furthermore, the PCB concentrations in Green Bay bald eagle tissues significantly exceed those from inland control populations.

The contaminant concentrations in the Fox River pair of bald eagles are less clearly characterized. Only one egg has been analyzed; however, egg and nestling plasma data indicate that the Fox River birds are exposed to elevated concentrations of PCBs.

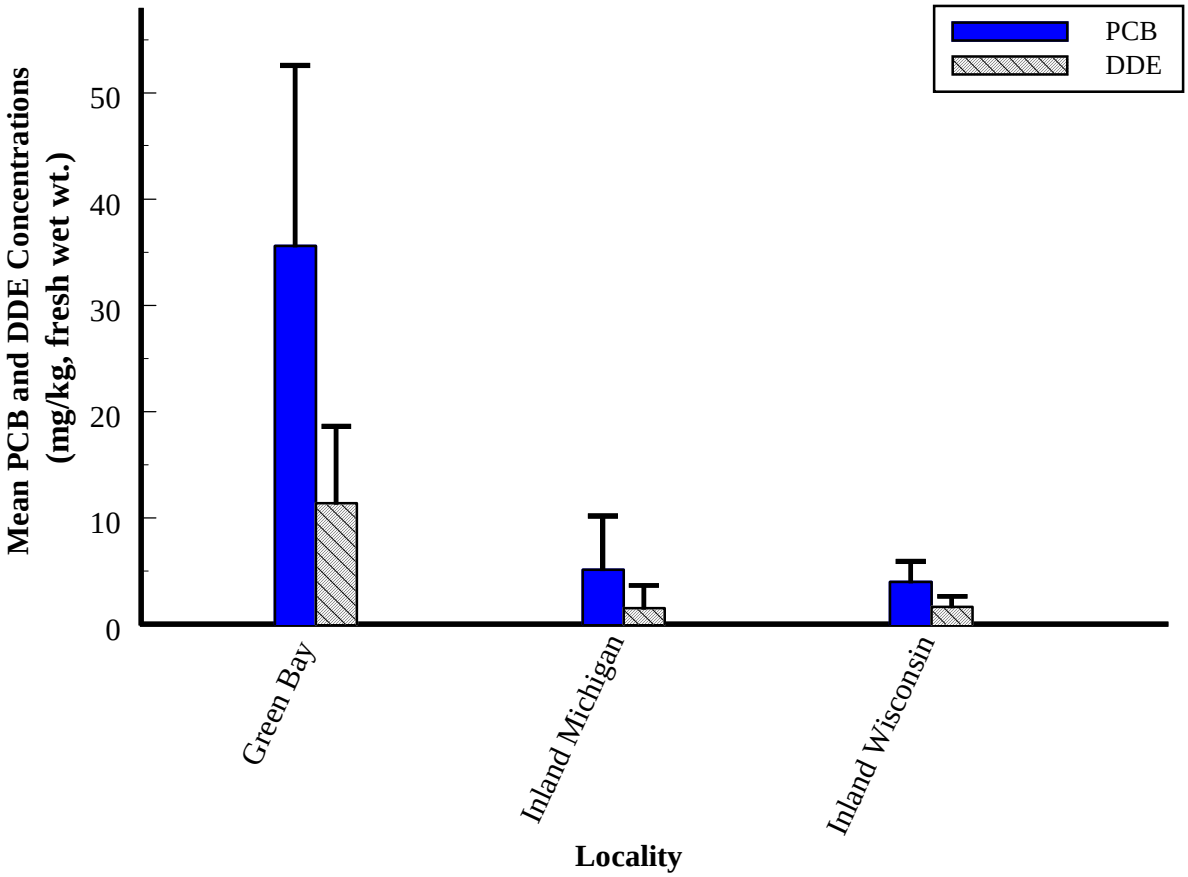


Figure 5-10. Mean PCB and DDE concentrations in bald eagle eggs from Green Bay, inland Michigan, and inland Wisconsin. Vertical lines represent one standard deviation.

5.6.5 Injuries to Assessment Area Bald Eagles

This section evaluates current evidence that assessment area bald eagles have been injured, focusing on reproductive malfunctions.[1] We then present an analysis of causality in which the main question addressed is whether observed injuries have been caused by exposure to PCBs.

Malfunctions in Green Bay Bald Eagle Reproduction

Figure 5-11 shows productivity histories of individual nests of bald eagles nesting in inland Michigan, inland Wisconsin, and Green Bay. These data show that there is much variability in

[1] Bowerman et al. (1994b) reported six instances of bill deformities among Great Lakes bald eagle nestlings. No such abnormalities have been reported among assessment area birds. As a result, this effect is not considered further for bald eagles.

Table 5-11
PCB Concentrations (μg/kg wet weight) in Plasma of Nestling Bald Eagles from Green Bay and the Lower Fox River

Breeding Area/State/Number	Year	Total PCBs[a]
Green Bay		
Granskog Lake/MI/DE-13	1987	229
Boutlier Lake/MI/DE-15	1987	319
Peshtigo River N/WI/MT-16	1992	901
Toft Point/WI/DO-01	1994	121
Oconto River-Thome/WI/OC-04	1994	393
Toft Point/WI/DO-01	1995	150
Blueberry Island	1994	83
Blueberry Island	1995	87
Fox River		
Kaukauna/WI/OU-01	1991	120
Kaukauna/WI/OU-01	1992	318
Kaukauna/WI/OU-01	1993	226
Kaukauna/WI/OU-01	1994	547
Kaukauna/WI/OU-01	1995	290
Mean Green Bay	n = 8	285.4
Mean Fox River	n = 5	300.2
Mean Inland Michigan[b]	n = 79	24

a. Data from Dykstra and Meyer, 1996.
b. Bowerman et al., 1994a.

inter-year productivity at individual nests. They also show, however, that the pattern for Green Bay nests is different from that in the two inland areas in that the Green Bay nests fail to produce young on a more consistent basis.

Figure 5-12 shows the mean annual productivity (number of large young produced) of Green Bay bald eagles compared with that of birds nesting in inland Michigan and inland Wisconsin between 1974 and 1998 (data provided by M. Meyer of Wisconsin DNR and D. Best of the USFWS). Mean annual productivity among inland Michigan and Wisconsin birds has consistently approximated or exceeded 1.0 young/nest, the productivity rate needed to maintain a healthy population (Kubiak and Best, 1991). Green Bay eagles had zero productivity during the period from 1974 until 1979. Green Bay nest productivity averaged at least 1.0 young per nest from 1980 to 1982 and from 1985 through 1987. After each of these three-year periods, productivity declined dramatically, reaching 0.0 within 1 or 2 years. However, during these periods there was only one or two pairs of eagles nesting in the assessment area (Figure 5-8). Productivity among Green Bay bald eagles has been at or near 1.0 young/year for 1995 through 1998. The

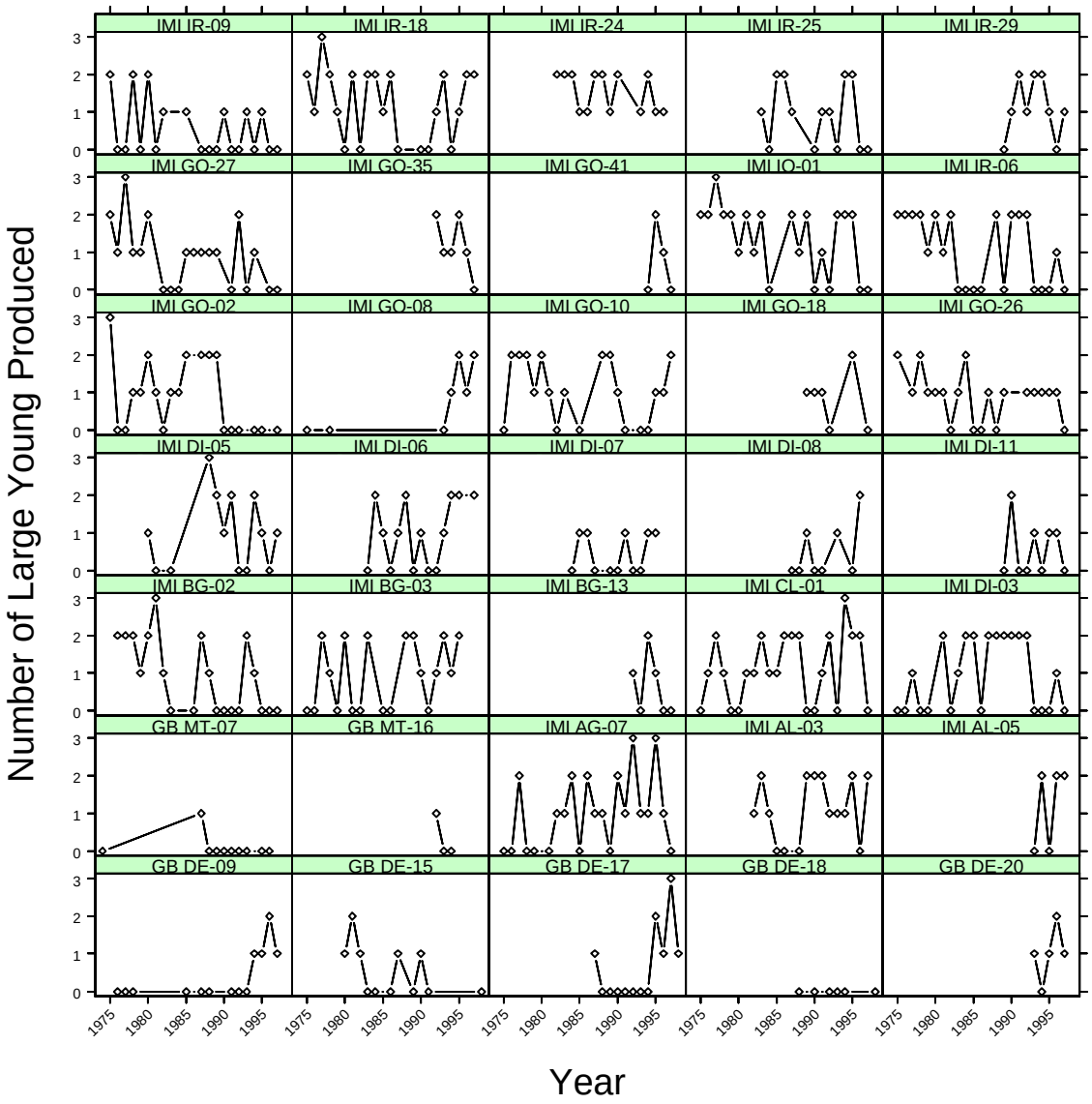


Figure 5-11. Productivity histories of individual bald eagle nests in inland Michigan (IMI), inland Wisconsin (IWI), and Green Bay (GB). Only nests for which there are both egg contaminants and productivity data are shown.

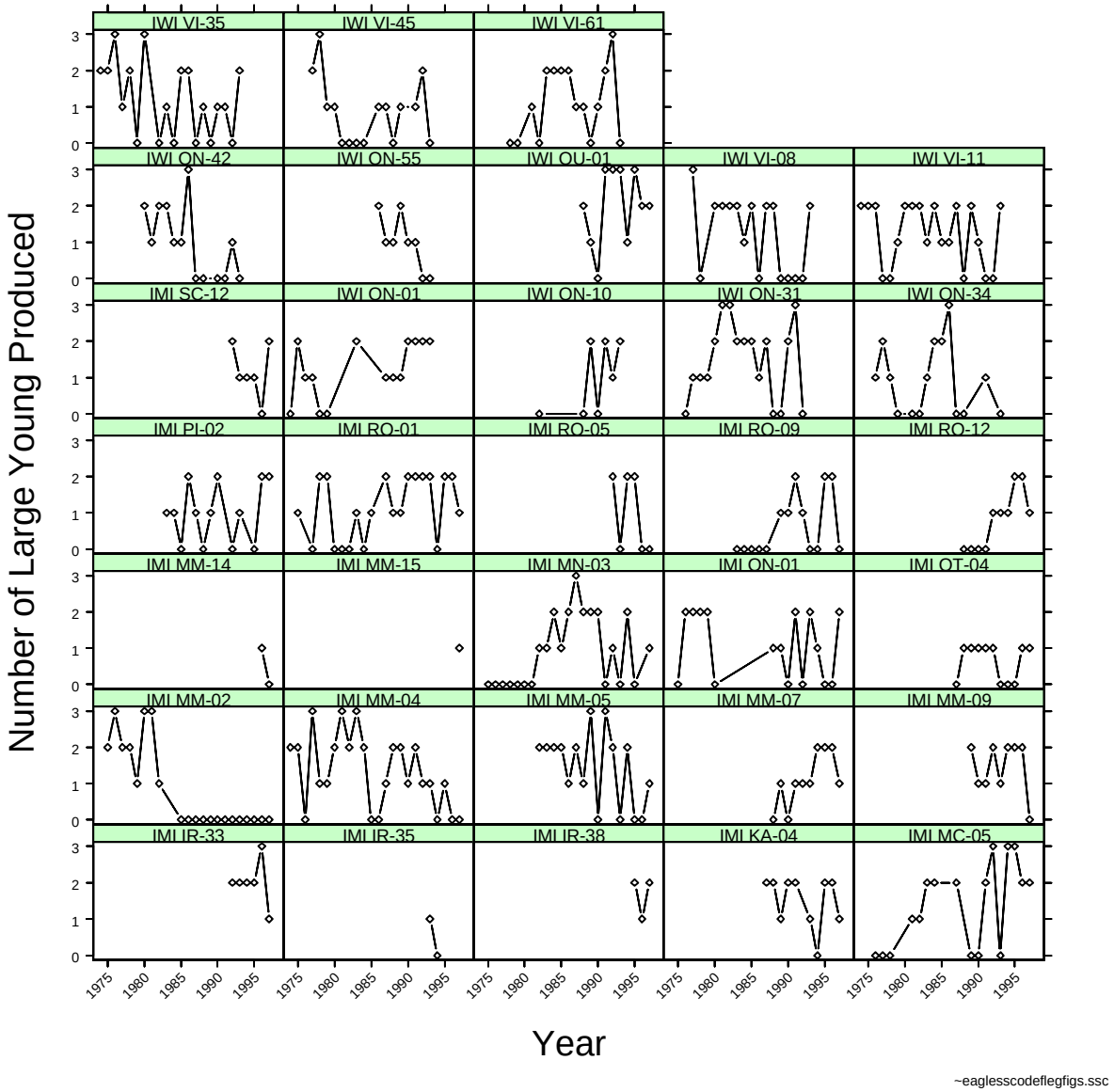


Figure 5-11 (cont.). Productivity histories of individual bald eagle nests in inland Michigan (IMI), inland Wisconsin (IWI), and Green Bay (GB). Only nests for which there are both egg contaminants and productivity data are shown.

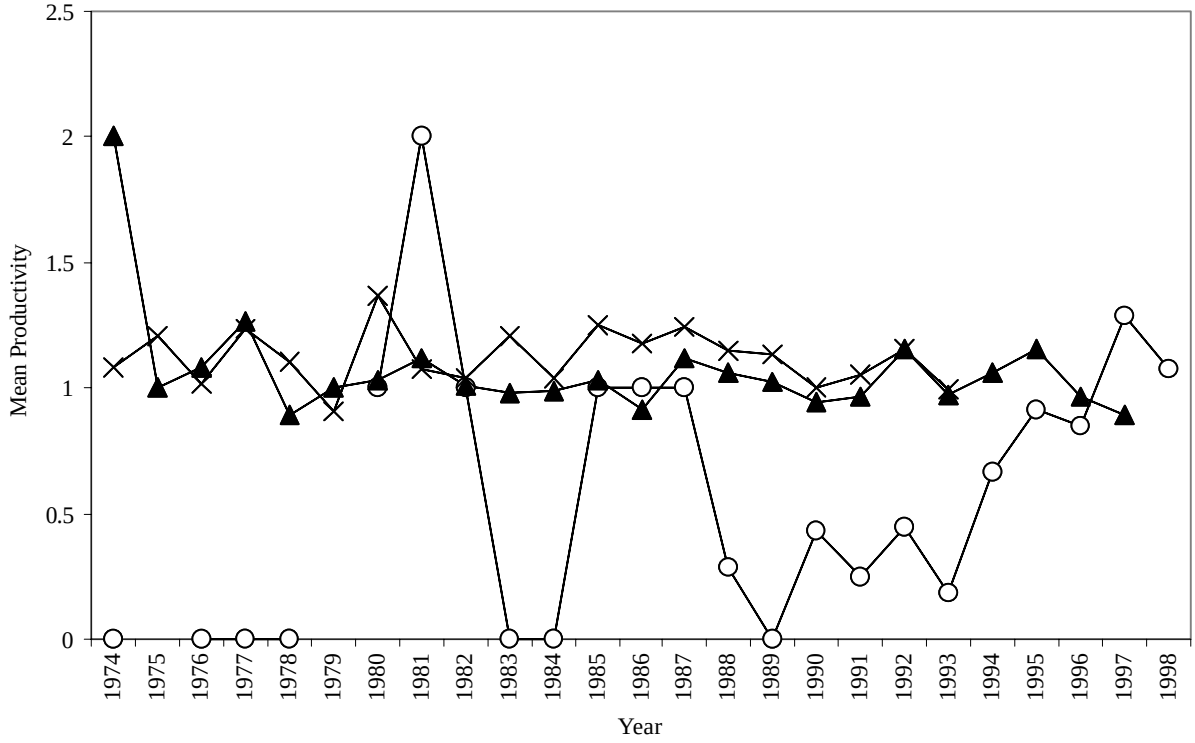


Figure 5-12. Mean annual productivity of bald eagles nesting on Green Bay (open circles), inland Michigan (triangles), and inland Wisconsin (crosses).

overall productivity rate of Green Bay bald eagles from 1974 through 1998 is significantly lower than for bald eagles in inland Wisconsin ($\chi^2 = 29.5$, 1 df, $p < 0.001$) and inland Michigan ($\chi^2 = 22.9$, 1 df, $p < 0.001$).

Table 5-12 presents the results of an analysis of the proportions of nests in Green Bay, inland Michigan, and inland Wisconsin that produced no, one, two, or three chicks during the period from 1974 to 1988. These data show that a higher proportion of bald eagle nest attempts in Green Bay resulted in no chicks being reared (0.54) than in either of the inland areas (0.36 and 0.34). Conversely, more inland nesting attempts resulted in one or more chicks being reared (0.63 and 0.66) than in Green Bay (0.46). These data support the conclusions of our previous mean productivity analysis by confirming that productivity is reduced in the assessment area.

Table 5-13 shows that the productivity of bald eagles nesting on the Fox River during the period from 1988 to 1998 was higher than in Green Bay. From 1988 to 1994 (when productivity among Green Bay eagles was low), the Fox River nests produced an average of 1.7 young/active nest. Since 1995, this productivity has been 2.4 young/active nest. The contaminants data from these sites suggest that the ratio of PCB to DDE in eggs and plasma may also be different from that for

Table 5-12
Proportions of Breeding Outcomes (0, 1, 2, or 3 chicks reared) among Green Bay, Inland Wisconsin, and Inland Michigan Bald Eagles

Area	Number of Nests and (nest years)	0 Chicks	1 Chick	2 Chicks	3 Chicks
Green Bay	23 (137)	0.54[a] (0.53)[b]	0.25 (0.20)	0.18 (0.24)	0.03 (0.03)
Inland Wisconsin	172 (1700)	0.34 (0.37)	0.25 (0.25)	0.36 (0.34)	0.05 (0.04)
Inland Michigan	251 (2664)	0.36 (0.37)	0.29 (0.31)	0.31 (0.30)	0.03 (0.02)

a. Proportions calculated for all nest/years in region without distinguishing between nests.
b. Average proportions calculated for each nest then combined in regional averages.

Table 5-13
Productivity (large young raised per active nest) of Fox River Bald Eagles from 1988 to 1998

Nest Name	88	89	90	91	92	93	94	95	96	97	98
Kaukauna, WI	2	1	0	3	3	3	1	3	2	2	3
Mud Creek, WI							2	3	1	2	3
East River, WI							0				
Productivity Summary, All Nests											
Number of active nests	1	1	1	1	1	1	3	2	2	2	2
Number of young reared	2	1	0	3	3	3	3	6	3	4	6
Young/active nest	2	1	0	3	3	3	1	3	1.5	2	3

Note: a blank cell indicates that the nesting territory was unoccupied in that year.

Source: USFWS and Wisconsin DNR bald eagle productivity databases.

the Green Bay eagles (Tables 5-10 and 5-11). However, the relatively few data that are available also suggest that the toxicity of the PCBs measured in the Fox River egg may be less than that measured in Green Bay eggs. Using the H4IIE bioassay method, two eggs from a Peshtigo Marsh nest in 1988 averaged 147.5 pg/g TCDD-EQ, while one egg from the Kaukauna nest on the Fox River in 1990 had only 34 pg/g TCDD-EQ (D. Tillitt, USFWS, unpublished data). Thus, although

the Fox River eagles may have total PCB concentrations in their eggs that are similar to those in the eggs of Green Bay birds, their toxicity may be less.

Also, bald eagles nesting on Green Bay may have less opportunity to forage in uncontaminated areas than the Kaukauna and Mud River birds, which are close to Lake Winnebago (Figure 5-9) where uncontaminated prey can be obtained. This complicates the analysis of what may be causing the increased productivity of the Fox River birds. Overall, given the small sample sizes that are available for the Fox River birds, the reason that they have higher productivity than Green Bay birds is uncertain.

The data presented above confirm that, like other Great Lakes coastal populations of bald eagles (e.g., Kubiak and Best, 1991; Best et al., 1994; Bowerman et al., 1995), eagles nesting around the Green Bay coastline have suffered decreased reproductive rates. The reduced productivity in the assessment area began in 1974, when the area was first recolonized, and continued up until at least the mid-1990s.

5.6.6 Green Bay Bald Eagle PCB and DDE Tissue Residues and Toxicity Thresholds

Kubiak and Best (1991), Wiemeyer et al. (1993), and Nisbet and Risebrough (1994) have used relationships between geospatial differences in PCB and DDE concentrations and productivity to postulate toxicity thresholds for each contaminant. The results are shown in Table 5-14. From these data, egg toxicity thresholds (concentrations at which adverse impacts on productivity become likely) may be >3.0 mg/kg wet weight for PCBs and >3.6 mg/kg wet weight for DDE. Major impacts on productivity (reductions of 50% or greater) are suggested at PCB and DDE concentrations of 13-23 mg/kg wet weight and 3.6-6.3 mg/kg wet weight, respectively.

Studies of the closely related white-tailed sea eagle in Scandinavia have also attempted to determine the contributions of PCBs and DDE to reduced hatching success (Helander et al., 1982; Helander et al., 1998; Olsson et al., 1998). These studies have not been entirely successful in determining contributions (because of the high correlation between the two contaminants in eggs). However, Olsson et al. (1998) suggested a total PCB embryo mortality LOEL of 300 mg/kg wet weight. The relevance of these studies to bald eagles is not yet clear.

All 13 of the Green Bay bald eagle eggs analyzed (Table 5-10) either equaled or exceeded 13 mg/kg wet weight PCBs, and 12 of these eggs are within or exceed the 3.6-6.3 mg/kg wet weight DDE range. Based on the thresholds in Table 5-14, the PCB concentrations in all of the Green Bay eggs are sufficient to result in major reproductive failure. The same is true for DDE for most of the eggs. Thus, based on the above thresholds, both PCBs and DDE could have been responsible for the reduced productivity observed in Green Bay bald eagles before the mid-1990s.

Table 5-14
Bald Eagle Egg Toxicity Levels Identified from Comparisons of Regional Productivities and Contaminant Concentrations

Productivity Response	Egg PCB Toxic Level (mg/kg fresh wet weight)	Egg DDE Toxic Level (mg/kg fresh wet weight)	Reference
"Normal" productivity	<3.0	<3.6	Wiemeyer et al., 1993
10% productivity reduction	3.0-5.6		Wiemeyer et al., 1993
30% productivity reduction	5.6-13.0		Wiemeyer et al., 1993
50% productivity reduction	13-23	3.6-6.3	Wiemeyer et al., 1993
70% productivity reduction	>23		Wiemeyer et al., 1993
75% productivity reduction		>6.3	Wiemeyer et al., 1993
"Healthy" reproduction	<1.7	<6.0	Kubiak and Best, 1991
No productivity reduction		<2.5	Nisbet and Risebrough, 1994
Productivity approximately halved		>5.0	Nisbet and Risebrough, 1994

To investigate potential relationships between productivity and PCBs in bald eagle eggs, the productivity data in Figure 5-11 were converted to probabilities that bald eagles in the assessment area and in the two inland reference areas will raise either no young or one or more young, and were assessed in relation to egg PCB concentrations. Productivity observations for individual nest years were omitted if they were separated by more than two years from years in which PCB concentration data were available for that nest. In cases where multiple PCB records were available for the same nest, but were separated by more than four years, independent productivity probabilities were calculated for the two or more periods. The series of productivity records that were retained by this procedure were used to calculate the relative frequency of producing a particular number of chicks, which was used to represent probabilities. These probabilities are presented in relation to the PCB concentrations measured in eggs from those nests (Figure 5-13). Figure 5-13 shows that the probability that bald eagle nests will rear no young rises steeply after egg PCB concentrations exceed 20 mg/kg fresh wet weight. Conversely, the probability that birds will raise one or more young falls after that concentration. All but one of the Green Bay bald eagle eggs that have been analyzed (Table 5-10) had PCB concentrations that exceed this threshold. This indicates that, based on the 20 mg/kg threshold, PCB concentrations in Green Bay bald eagle eggs are sufficient to result in reduced productivity.

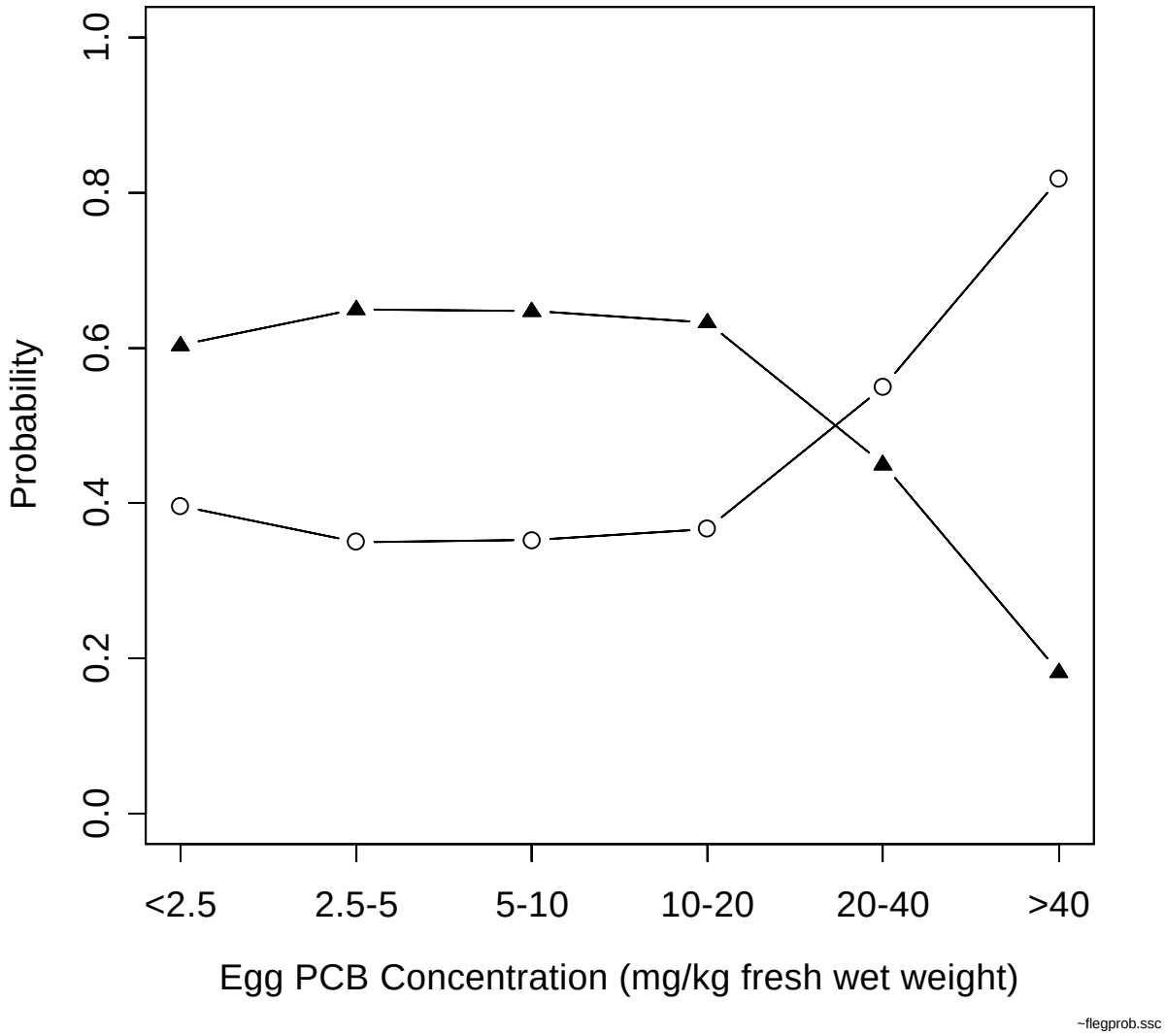


Figure 5-13. Probability of bald eagles in inland Michigan and Wisconsin and Green Bay producing no young (open circles) or one or more young (triangles) in relation to egg PCB concentrations.

5.6.7 PCBs and DDE and Reduced Reproductive Success among Green Bay Bald Eagles

The data presented in Sections 5.6.4 through 5.6.6 show that Green Bay bald eagles have elevated egg and plasma PCB and DDE concentrations. They also show that Green Bay bald eagles, during the period from 1987 until the mid-1990s, had significantly lower reproductive success than inland Wisconsin or Michigan birds and that, based on toxicity thresholds, the reduced reproduction could be attributable to the elevated PCB and DDE concentrations.

Previous studies of Great Lakes bald eagles [Kubiak and Best (1991), Bowerman (1993), Bowerman et al. (1994a), and Bowerman et al. (1995)] found that productivity among Great Lakes bald eagles was negatively correlated with both PCB and DDE concentrations in eggs and attributed the reduced reproductive success to these contaminants. Dykstra and Meyer (1996) evaluated the causes of the low productivity in Green Bay bald eagles and found that the low productivity was not attributable to either food availability (indices of food availability were similar to inland Wisconsin nests) or disturbance (adult attendance patterns at the nests were also similar to inland birds). Dykstra and Meyer (1996) concluded that the reduced productivity among Green Bay bald eagles was caused by PCBs and/or DDE. PCB and DDE concentrations are typically correlated in bald eagle eggs [Wiemeyer et al. (1993): $r = 0.76$; analysis of data in Dykstra and Meyer (1996): $r = 0.65$; analysis of mean PCB and DDE concentrations in bald eagle eggs from seven Great Lakes regions in Kubiak and Best (1991): $r = 0.9$; Clark et al. (1998): $r = 0.91$]. Figure 5-14 shows the relationship between PCB and DDE concentrations in bald eagle eggs from Green Bay. These data are from the USFWS and Wisconsin DNR contaminants monitoring databases. PCBs are positively correlated with DDE ($r = 0.67$, $p < 0.05$). Figure 5-15 shows a similar analysis but using all of the egg concentration data from Green Bay, inland Michigan, and inland Wisconsin. PCBs are again significantly correlated with DDE ($r = 0.8$, $p < 0.001$). This correlation between contaminants has proven to be a difficulty in previous attempts to quantify their relative contributions to reduced productivity in bald eagles (Wiemeyer et al., 1993; Dykstra and Meyer, 1996).

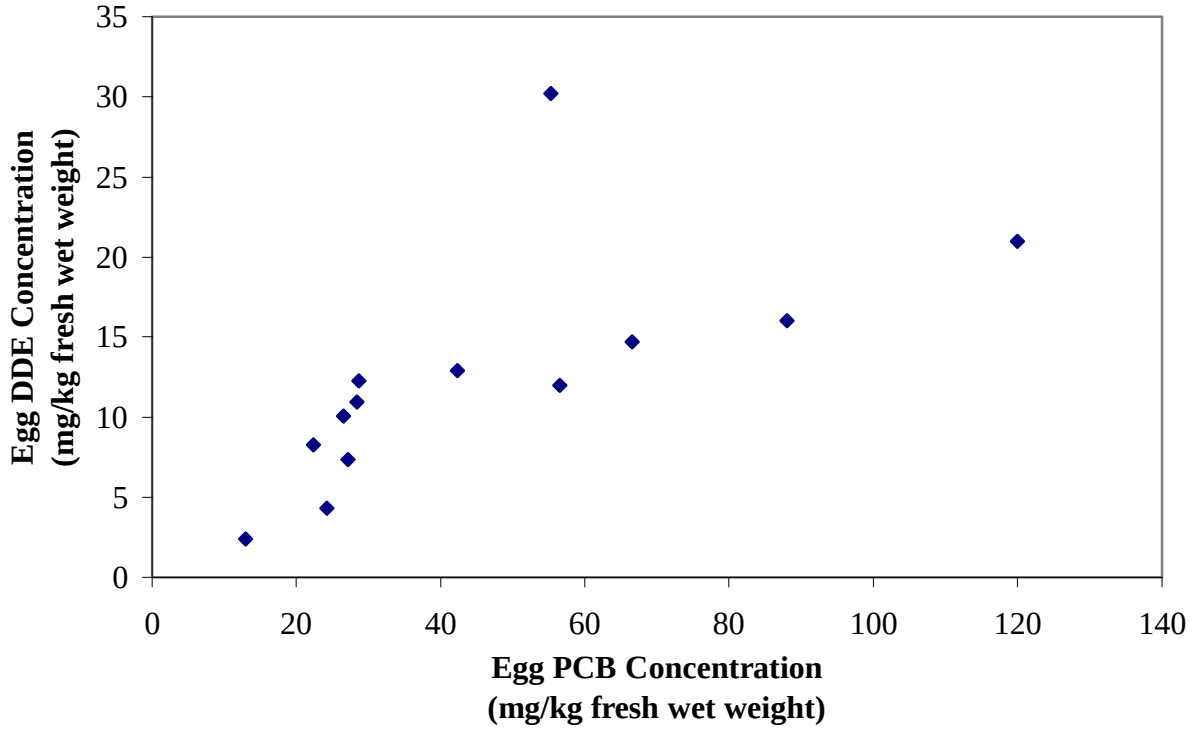


Figure 5-14. Relationship between PCB and DDE concentrations in Green Bay bald eagle eggs.

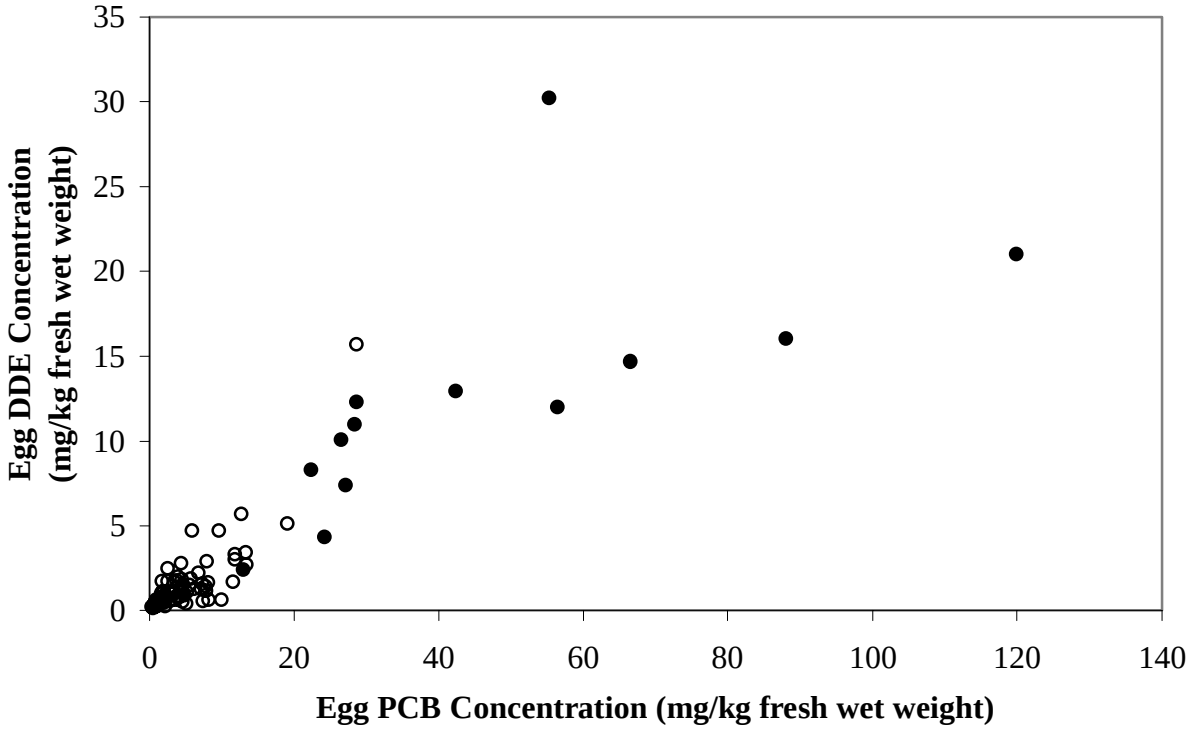


Figure 5-15. Relationship between PCB and DDE concentrations in bald eagle eggs from Green Bay (solid circles) and inland Michigan and Wisconsin (open circles).

Figures 5-16 through 5-19 show the relationships between PCB and DDE concentrations in eagle eggs from Green Bay, inland Wisconsin, and inland Michigan and two measures of productivity: the mean number of young reared at the site from which an egg was taken for chemistry analysis during the year of egg collection and the year preceding and subsequent to that event (3-year productivity), and the mean number of young reared at the site from which an egg was taken for chemistry analysis during the year of egg collection and the two years preceding and subsequent to that event (5 year productivity). These chemistry and productivity data were obtained from the USFWS and Wisconsin DNR monitoring data sets supplied by D. Best (USFWS) and M. Meyer (Wisconsin DNR).

Figures 5-16 through 5-19 show negative relationships between both PCB and DDE egg concentrations and productivity. These negative correlations are statistically significant for PCBs and 3 year productivity ($r = -0.4$, $p < 0.001$), DDE and 3 year productivity ($r = -0.36$, $p < 0.01$), PCBs and 5 year productivity ($r = -0.4$, $p < 0.001$), and DDE and 5 year productivity ($r = -0.3$, $p < 0.001$). Productivity in reference areas normally averages about 1.1 young/nest (Figure 5-11); thus, Figures 5-16 through 5-19 show that increases in egg PCB and DDE concentrations are associated with markedly reduced productivity. In contrast, in a recent study, Donaldson et al. (1999) found no significant relationships between productivity and either PCBs or DDE in eggs or

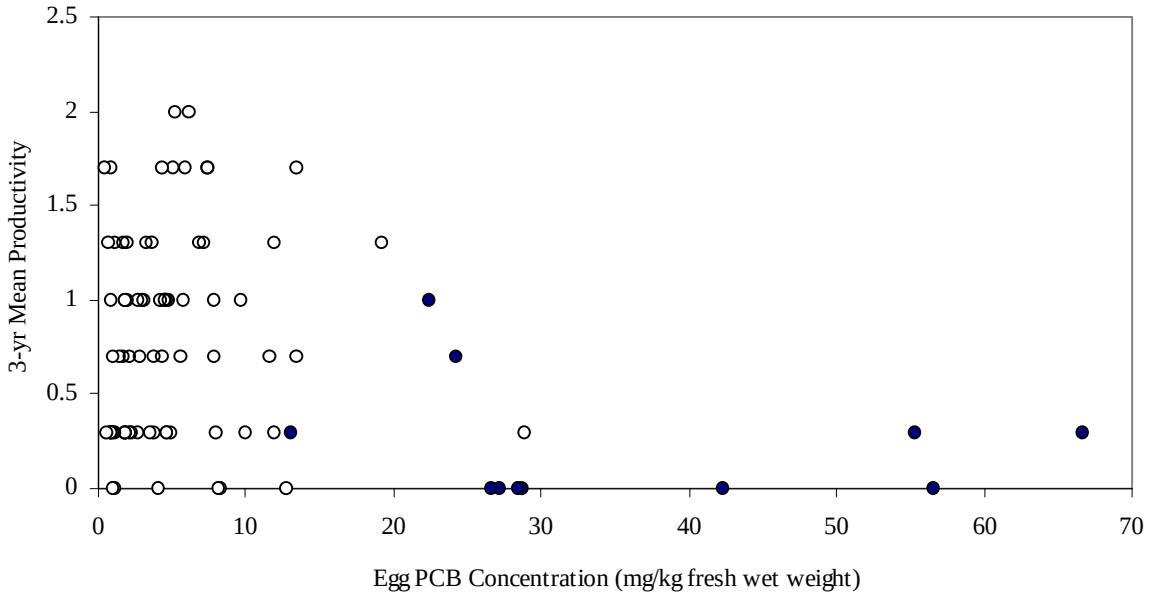


Figure 5-16. Egg PCB concentrations and 3-year mean productivity at bald eagle nests on Green Bay (solid circles) and in inland Michigan and Wisconsin (open circles).

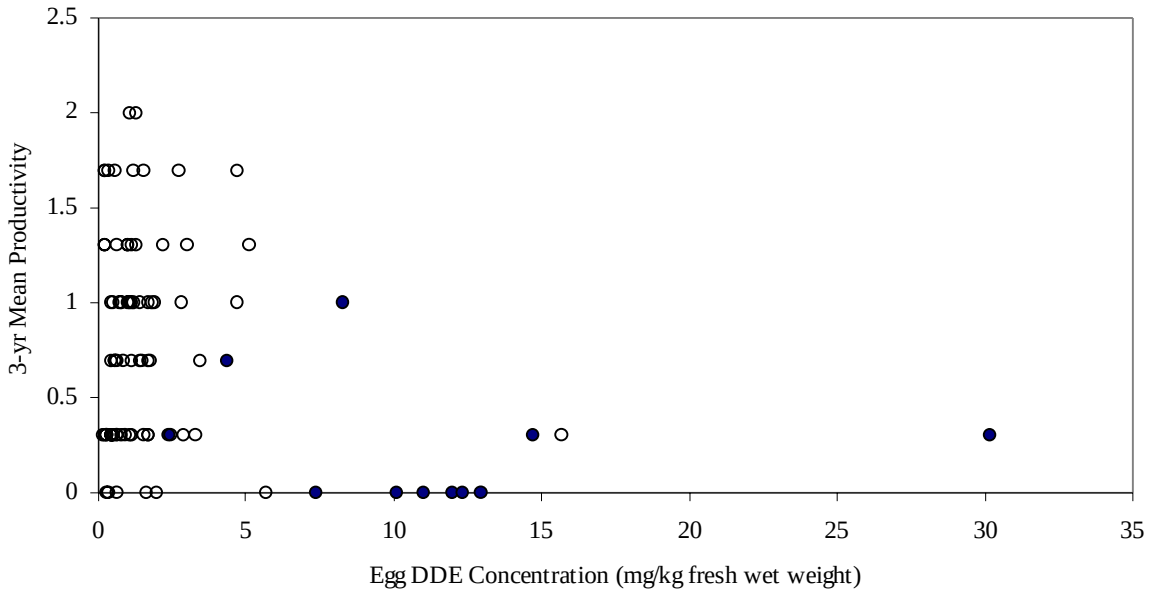


Figure 5-17. Egg DDE concentrations and mean 3-year productivity at bald eagle nests in Green Bay (solid circles) and in inland Michigan and Wisconsin (open circles).

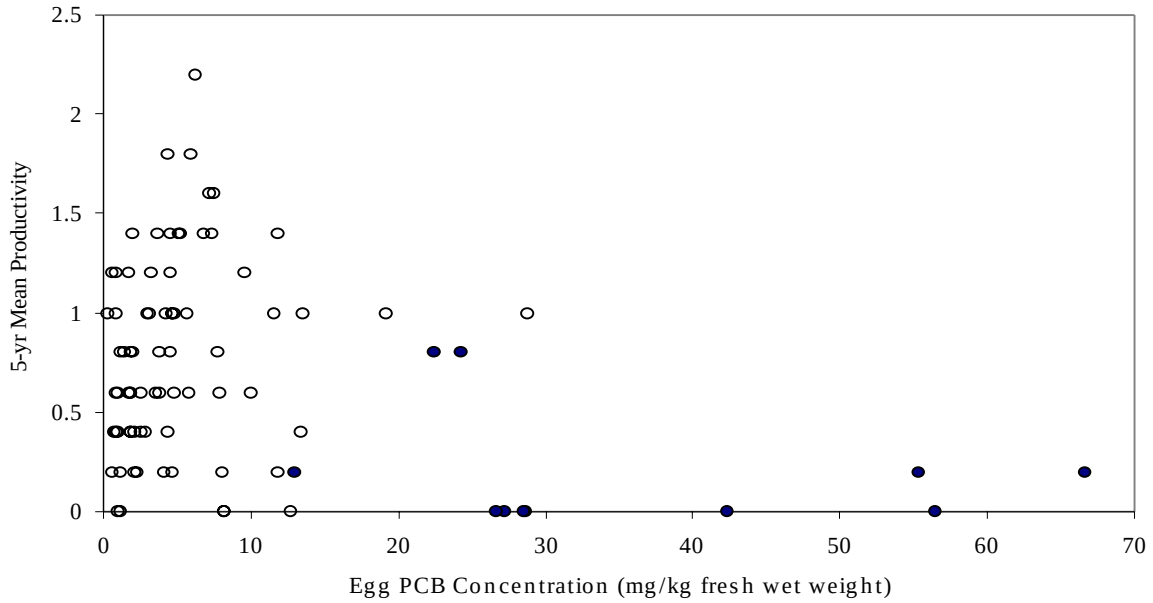


Figure 5-18. Egg PCB concentrations and mean 5-year productivity at bald eagle nests on Green Bay (solid circles) and in inland Michigan and Wisconsin (open circles).

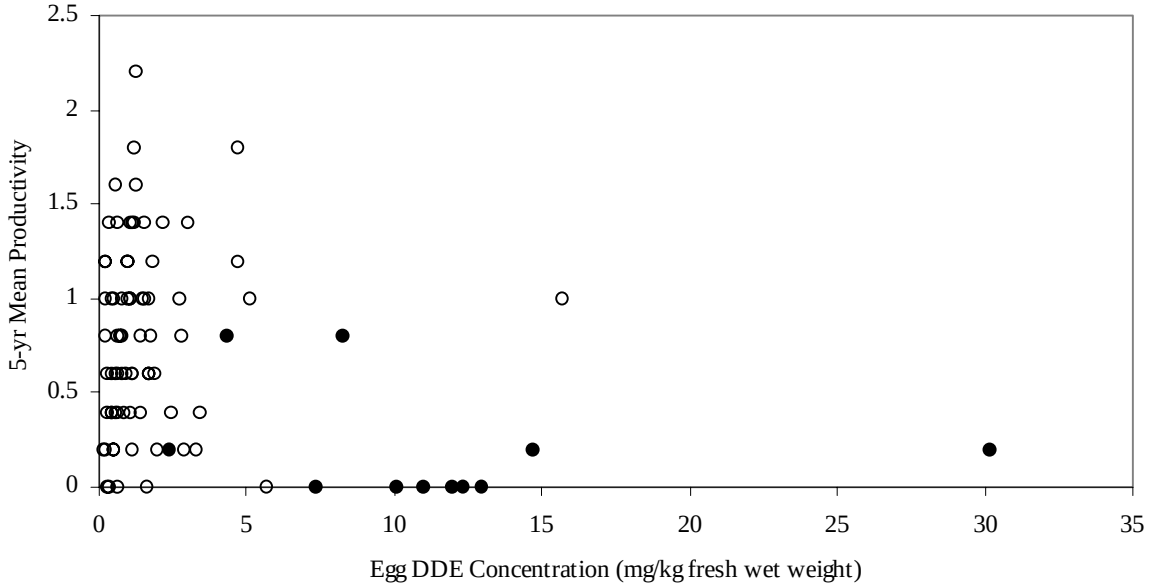


Figure 5-19. Egg DDE concentrations and mean 5-year productivity at bald eagle nests on Green Bay (solid circles) and in inland Michigan and Wisconsin (open circles).

nestling plasma from the Canadian Great Lakes. The reasons for the differences between the results of our analysis and those of Donaldson et al. are unclear.

However, the data reported in Donaldson et al. show that the period over which productivity was measured (1980-1996) was largely subsequent to a period in which PCB and DDE concentrations in Lake Erie bald eagle eggs had undergone substantial declines (1974-the early to mid 1980s). Thus, the productivities of the nests were measured after contaminants had declined (by approximately factors of 4).

5.6.8 The Relative Contributions of PCBs and DDE to Reduced Reproductive Success in Green Bay Bald Eagles

In this section we evaluate the relative contributions of PCBs and DDE to the reduced reproductive success among Green Bay bald eagles. We concentrate on PCBs and DDE because: these are the only contaminants that have been found in Great Lakes bald eagle tissues in high enough concentrations to result in adverse effects (Bowerman et al., 1995); they are the contaminants that have been most closely correlated with bald eagle reproductive success in the Great Lakes and elsewhere (Wiemeyer et al., 1984; Kubiak and Best 1991; Nisbet and Risebrough, 1994; Bowerman et al., 1995; Clark et al., 1998); and they are known to result in the types of adverse effect (embryo mortality and reduced reproductive success) observed in assessment area bald eagles.

To evaluate whether PCB effects on productivity in Green Bay can be differentiated from those of DDE, we performed partial correlation analyses. In these analyses, we partialled out DDE [making the conservative assumption that DDE is having a significant effect on productivity] to evaluate whether PCBs explain a significant amount of the residual variation. The results of these tests (Pearson and Spearman) are shown in Table 5-15. Egg PCB concentrations did not explain a significant amount of the residual variation.

Table 5-15
Partial Correlation Coefficients Obtained in Pearson and Spearman Analyses of Egg PCB Concentrations and Productivity

Test	3-Yr Productivity	5-Yr Productivity
Pearson	-0.01 (0.91)	0.00 (0.93)
Spearman	0.03 (0.77)	0.05 (0.66)
Note that p values are given in parentheses.		

In a complementary analysis, we then partialled out the effects of PCBs to evaluate whether DDE explains a significant amount of the residual variation. The results of these tests (Pearson and Spearman) are shown in Table 5-16. Egg DDE concentrations did not explain a significant amount of the residual variation.

Table 5-16
Partial Correlation Coefficients Obtained in Pearson and Spearman Analyses of Egg DDE Concentrations and Productivity

Test	3-Yr Productivity	5-Yr Productivity
Pearson	-0.15 (0.17)	-0.16 (0.15)
Spearman	-0.13 (0.24)	-0.11 (0.32)
Note that p values are given in parentheses.		

The results of the analyses described above are not sufficient to allow us to determine the relative contributions of PCBs and DDE to reductions in productivity in Green Bay bald eagles. The exceedences of the thresholds developed by Nisbet (1989), Kubiak and Best (1991), Wiemeyer et al. (1993), Nisbet and Risebrough (1994), and Stratus Consulting (Section 5.6.6), and the correlations shown above, suggest that both contaminants may be affecting productivity and that separating their effects, given the degree of correlation, is not feasible.

5.6.9 Summary

The data and analyses on bald eagles described in this section show the following:

- Green Bay bald eagles have been exposed to PCBs through their diet.
- PCB concentrations in bald eagle eggs and chick plasma in Green Bay are significantly higher than those in reference areas.
- Productivity among Green Bay bald eagles was significantly reduced relative to reference area eagles from 1974 until at least the mid-1990s.
- Exceedences of the Kubiak and Best (1991), Wiemeyer et al. (1993), and Nisbet and Risebrough (1994) thresholds and thresholds developed by Stratus Consulting, together with the negative correlations between PCB and DDE egg concentrations and productivity, indicate that PCBs and/or DDE have contributed to the reduced productivity of Green Bay bald eagles.

- Given the limitations of the chemistry and productivity data sets and of correlation among contaminants, it is not possible to determine the relative contributions of PCB and DDE to the reduced productivity of Green Bay bald eagles.

CHAPTER 6
INJURIES TO WATERFOWL: CONSUMPTION ADVISORIES

Previous chapters discussed toxicological injuries to birds caused by PCBs. In this chapter we evaluate injuries to waterfowl (ducks and geese) in Green Bay associated with PCB accumulation in bird tissue in excess of federal or state action, tolerance, or consumption advisory levels. In addition to the toxicological injuries described in previous chapters, the Departmental NRDA regulations specify that injury has occurred when concentrations of hazardous substances are sufficient to "exceed action or tolerance levels established under section 402 of the Food, Drug and Cosmetic Act" [43 CFR § 11.62(f)(1)(ii)] or "exceed levels for which an appropriate State health agency has issued directives to limit or ban consumption" [43 CFR § 11.62(f)(1)(iii)].

6.1 STATUS AND ECOLOGY OF WATERFOWL

Waterfowl are both breeding summer residents and passage migrants in the assessment area (see Chapter 2). During the summer months, surface feeding ducks and geese such as mallard, teal, gadwall, and Canada geese nest in the marshes adjacent to Green Bay, whereas red-breasted mergansers nest on many of the islands that are adjacent to the Door Peninsula (White and Cromartie, 1977; Heinz et al., 1983). In the fall, the breeding populations are augmented by large numbers of migratory ducks and geese (Robbins, 1991). These migrants, including scaup, bufflehead, goldeneye, redheads, and canvasbacks, feed in the bay until they are forced by the onset of winter to move to more southerly wintering areas. During the fall influx, the waterfowl in Green Bay and its surrounding wetlands are intensively hunted and comprise an important recreational resource (K. Stromborg, U.S. Fish and Wildlife Service, personal communication, 1998).

6.2 PATHWAY AND EXPOSURE ANALYSIS

No data have been reported on the diets of waterfowl species in Green Bay. However, based on what is known about the diets of waterfowl in general (Ehrlich et al., 1988), many of the species that inhabit the bay (e.g., mallard, teal, gadwall) are primarily herbivorous, consuming aquatic and marsh vegetation. Others (e.g., goldeneye, canvasbacks, and buffleheads) are likely to consume mainly benthivorous organisms such as molluscs, while mergansers are mainly piscivores and prey on small forage fish (Ehrlich et al., 1988).

PCB contamination in assessment area forage fish is described in Section 5.1.2. These data demonstrated that forage fish contain elevated concentrations of PCBs and serve as a component of the dietary pathway to higher trophic levels.

No data have been reported on PCBs in the diets of herbivorous or benthivorous waterfowl in the assessment area. However, given that water, sediment, phytoplankton, and zooplankton in the assessment area are contaminated with PCBs, it is likely that aquatic plants consumed by waterfowl are contaminated with PCBs. In addition, Beyer et al. (1997, 1998) found that up to 18% of the diet of swans, geese, and ducks can be incidentally ingested sediment. Thus, herbivorous and benthivorous waterfowl species are likely to be exposed to PCBs in the assessment area through ingestion of food items and incidental uptake of sediments.

Tissue analysis of various waterfowl species confirms that individuals from Green Bay have been exposed to PCBs. These data are summarized in Table 6-1.

Table 6-1
PCB Concentrations Measured in the Tissues of Waterfowl from Green Bay

Species	Diet[a]	Tissue	Site	Year	Mean PCB Conc. (mg/kg wet weight)	Reference
Red-breasted merganser	fish	eggs	Door Cty.	1975	44.7	White and Cromartie, 1977
Red-breasted merganser	fish	eggs	Door Cty.	1977/1978	20	Haseltine et al., 1981
Red-breasted merganser	fish	eggs	Door Cty.	1990	11.1	Williams et al., 1995b
Common merganser	fish	eggs	Door Cty.	1975	79.4	White and Cromartie, 1977
Mallard	plants	eggs	Door Cty.	1977/1978	2	Haseltine et al., 1981
Mallard	plants	muscle, skin, and fat	Lower Fox River	1985/1986	0.4	Amundson, undated
Mallard	plants	muscle, skin, and fat	Lower Fox River and inner Green Bay	1987	0.37	Wisconsin DNR wildlife contaminants database (supplied by K. Patnode, B. Hill of WDNR)
Mallard	plants	muscle and skin	Green Bay	1997	0.45	USFWS, unpublished data
Scaup	benthos	muscle and skin	Southern Green Bay	1997	2.0	USFWS, unpublished data

a. Assumed based on species description in Ehrlich et al., 1988.

Except for Amundson (undated), none of these studies reported PCB concentrations in waterfowl from reference areas. Amundson reported that the mean PCB concentration among 55 mallard from the assessment area in 1985-1986 (0.43 mg/kg wet weight) was significantly greater than that reported among mallard from inland areas of Wisconsin (0.19 mg/kg wet weight). Amundson also found that only 20% of inland Wisconsin mallard exceeded the PCB detection limit, compared with 64% of mallard from the assessment area.

Overall, these data confirm that waterfowl in Green Bay have been exposed to PCBs and that, at least for mallard, they have PCB body burdens that exceed those from reference areas.

6.3 INJURIES TO WATERFOWL IN GREEN BAY

In this section we show that waterfowl in the assessment area have been injured by their exposure to PCBs. These injuries comprise exceedences of "action or tolerance levels established under section 402 of the Food, Drug and Cosmetic Act" [43 CFR § 11.62(f)(1)(ii)] and exceedences of "levels for which an appropriate State health agency has issued directives to limit or ban consumption" [43 CFR § 11.62(f)(1)(iii)]. We first discuss the procedural bases for the federal tolerance level and the state advisories. We then present data that show that waterfowl in the assessment area have PCB tissue concentrations that exceed federal and state action or tolerance levels. Lastly we describe the waterfowl consumption advisory imposed in the assessment area by the State of Wisconsin in response to the measured PCB concentrations in waterfowl tissue.

6.3.1 Basis of the Federal Tolerance Level

The Food, Drug, and Cosmetic Act (21 U.S.C. 301 et seq.) authorizes the federal Food and Drug Administration (FDA) to protect the public health by regulating food shipped in interstate commerce. Sections 402 and 406 of the Act prohibit food from interstate commerce if the food contains any added poisonous or deleterious substance that is unsafe, unless the presence of the poisonous or deleterious substance cannot be avoided. Section 406 authorizes the FDA to limit the quantities of such substances by using formal rulemaking to set legal limits called tolerances. The tolerances are set at the level necessary to protect public health, taking into account the extent to which the substance is unavoidable and the ways that a consumer may be affected by the same or other deleterious substances (44 Fed. Reg. 38,330).

No tolerances have been established for waterfowl per se, but in 1972, the FDA proposed tolerances for PCBs in poultry (37 Fed. Reg. 5,705). The FDA acknowledged that there was limited knowledge of the toxicological effects of PCBs, but that PCBs appeared to be of moderate acute toxicity. The proposed temporary tolerance for poultry was 5.0 mg/kg wet weight on a fat basis. In 1973, the FDA issued regulations setting temporary tolerances for PCBs in poultry

(38 Fed. Reg. 18,096). The FDA called the PCB tolerances "temporary" because "new data may justify a further downward revision of the tolerances" (42 Fed. Reg. 17,493).

In 1977, the FDA proposed reducing the poultry tolerance from 5.0 mg/kg wet weight (fat basis) to 3.0 mg/kg wet weight (fat basis). In proposing this reduction, the FDA stated that it needed to balance protecting public health with avoiding excessive losses of food (42 Fed. Reg. 17,487). The proposal to reduce the tolerance to 3.0 mg/kg wet weight (fat basis) contained an extensive discussion of the basis for the decision based on the contaminant having become more avoidable and on new toxicity data on PCBs.

On June 29, 1979, the FDA issued a final rule reducing tolerances for PCBs (44 Fed. Reg. 38,330). The FDA also removed the designation "temporary" from the tolerances because the word was deemed not to have legal significance under Section 406 of the Food, Drug, and Cosmetic Act.

6.3.2 Basis for State Waterfowl Consumption Advisories

In 1984 Wisconsin initiated its wildlife contaminant monitoring program (Amundson, undated; Miller, 1987). This program was initiated for two reasons: first the state was cognizant that it had a responsibility to assure that game harvested by sportsmen was "healthy, wholesome, and free of contamination" (Miller, 1987); second the state wanted to monitor contaminant levels in wildlife species (Miller, 1987).

The results of the monitoring program showed that the majority of game over most of the state was relatively free of contamination. However, for certain species in certain regions, contaminants such as PCBs were elevated (Amundson, undated; Miller, 1987). Wisconsin then developed procedures for issuing consumption advisories for waterfowl (Miller, 1987). These procedures indicate that an advisory will be issued once a year, that the mechanism for public notification will be through a news release, that preparation and cooking recommendations will form part of the advisory notice, and that the advisory will be specific about areas and species covered. The threshold level that was adopted by the state, and that triggered this PCB advisory, was the federal tolerance level for poultry of 3 mg/kg wet weight PCBs on a fat basis.

6.3.3 Exceedences of FDA and State Tolerance Levels

There are two sources of data on PCB contamination of waterfowl species in the assessment area. The first is the Wisconsin DNR wildlife contaminants database (Amundson, undated; unpublished data supplied by K. Patnode and B. Hill of Wisconsin DNR). These data form the basis for the consumption advisories issued by the Wisconsin DNR and printed in the yearly hunting regulations guide. The second source of data is a study undertaken by the Service during the

summer and fall of 1997 to update, replicate, and extend the findings of the Wisconsin DNR monitoring (USFWS, unpublished data). In this study, a variety of waterfowl species were collected and edible portions were analyzed for total PCBs and lipids. Collections were made on several different dates in 1997 and at several locations. Details regarding the sample collection and analysis procedures are provided in Appendix B.

The Wisconsin DNR data show that in 1985-1986 the mean PCB concentration of 55 mallard collected from the Lower Fox River was 6.05 mg/kg fat (Amundson, undated) (Figure 6-1). In 1987 it was 9.02 mg/kg fat in 33 mallard collected from the Lower Fox River and inner Green Bay (Wisconsin DNR wildlife contaminants database) (Figure 6-1).

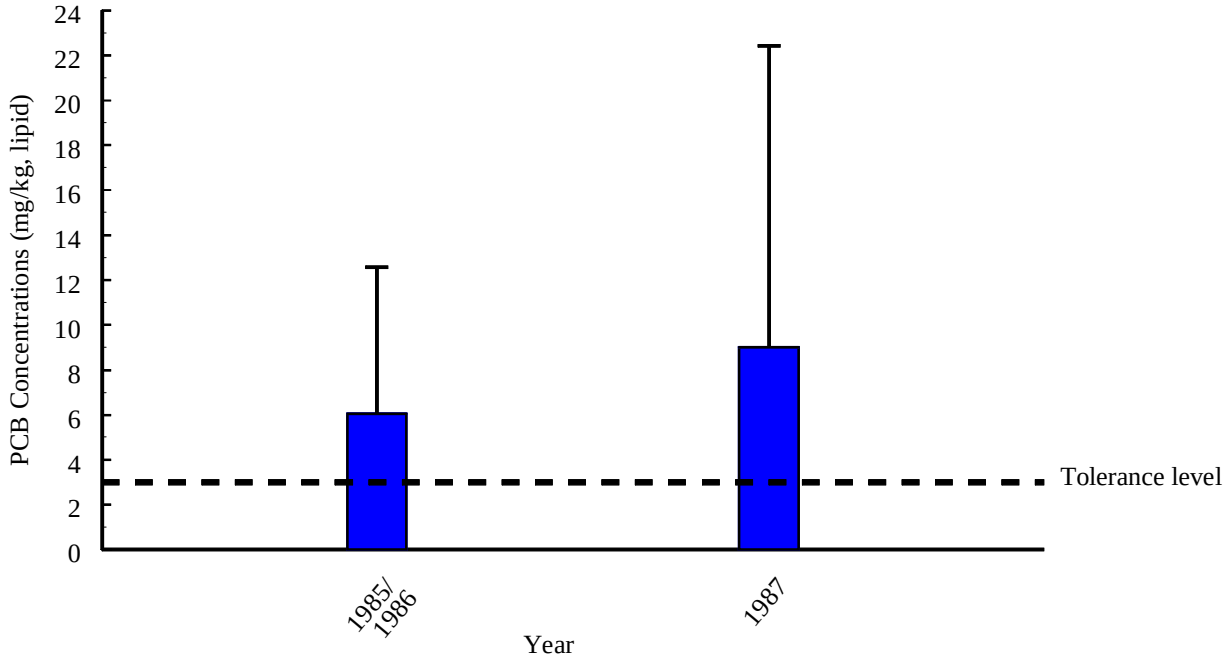


Figure 6-1. Mean PCB concentrations in mallard from the assessment area. Dashed line is the FDA and State of Wisconsin tolerance level. Vertical line represents one standard deviation.

The first USFWS collection was of 10 mallard from Lower Green Bay along the shoreline from the mouth of the Fox River eastward to the vicinity of Point au Sable in June 1997. These birds were probably summer residents. In addition to the mallard, one lesser scaup was collected. This bird apparently had not migrated to its normal breeding grounds. Eight of 10 of the mallards and the scaup exceeded the federal and the Wisconsin tolerance levels in skin plus attached muscle fillets (Table 6-2). Many of these birds also exceeded the tolerance levels in muscle tissue alone. The scaup exceeded the tolerance levels for both tissue types.

Table 6-2
PCB Concentrations in Lesser Scaup and Mallards Collected by USFWS in Southern Green Bay on June 12, 1997

Species	Sex	Age	PCBs (mg/kg wet weight lipid)[a]	
			Muscle	Muscle and Skin
Lesser scaup	male	adult	16.3[b]	23.3[b]
Mallard	male	adult	nd	2.0
Mallard	female	adult	nd	2.2
Mallard	female	adult	nd	8.0[b]
Mallard	male	adult	nd	4.5[b]
Mallard	male	adult	15[b]	19.5[b]
Mallard	male	adult	6.6[b]	15.4[b]
Mallard	male	adult	13.9[b]	17.4[b]
Mallard	female	adult	2.9	9.6[b]
Mallard	male	adult	nd	5.6[b]
Mallard	male	adult	5.9[b]	16.9[b]

a. The level of detection was 0.02 mg/kg wet weight, which is equivalent to 0.4 mg/kg wet weight on a lipid basis for a tissue sample of 5% lipid.
b. Exceeds federal and State of Wisconsin tolerance levels of 3 mg/kg wet weight, fat basis.

nd = not detected.

Another sample of waterfowl was collected by the USFWS (unpublished data) near Point au Sable during the peak of the influx of migratory ducks from northern areas in late October and November 1997. A variety of species was collected (Tables 6-3 and 6-4), spanning the range of diving ducks normally encountered by hunters in this area. Two of these ducks had PCB residues in their tissues that exceeded the federal and the Wisconsin tolerance levels.

A third set of samples was collected in September 1997 in northern Door County, Green Bay, and adjacent Lake Michigan (Table 6-5). All of these birds were diving ducks, which feed on a diet of animal rather than plant material. Of the 14 birds sampled, 13 had PCB residues that exceeded the federal and the Wisconsin tolerance levels.

The data in the Wisconsin DNR database and in Tables 6-2 through 6-5 show that many of the waterfowl collected on the Lower Fox River and Green Bay have body burdens of PCBs that

Table 6-3
PCB Concentrations in Skin and Breast Muscle of Waterfowl Collected from Point au Sable, Southern Green Bay on October 27, 1997

Species	Sex	Age	PCBs (mg/kg wet weight lipid)
Greater scaup	male	immature	1.6
Greater scaup	female	immature	3.1[a]
Greater scaup	male	immature	0.8
Greater scaup	female	immature	0.3
Greater scaup	female	immature	2.2
Greater scaup	male	immature	0.2
Lesser scaup	female	adult	2.7
Lesser scaup	male	immature	0.6
Common goldeneye	male	adult	14.1[a]

a. Exceeds federal and Wisconsin tolerance levels of 3 mg/kg wet weight, fat basis.

Table 6-4
PCB Concentrations in Skin and Breast Muscle of Waterfowl Collected from Point au Sable, Southern Green Bay on November 12-13, 1997

Species	Sex	Age	PCBs (mg/kg wet weight lipid)
Greater scaup	male	adult	3.3[a]
Lesser scaup	male	adult	2.4
Lesser scaup	male	immature	1.2
Lesser scaup	male	adult	5.1[a]
Lesser scaup	male	immature	2.5
Lesser scaup	female	immature	1.4
Lesser scaup	male	immature	0.4
Lesser scaup	male	immature	0.8
Lesser scaup	male	immature	0.9
Bufflehead	female	immature	0.4
Bufflehead	male	adult	1.4
Common goldeneye	female	immature	0.1
Common goldeneye	female	immature	1.5
Common goldeneye	male	immature	0.1

a. Exceeds federal and Wisconsin tolerance levels of 3 mg/kg wet weight, fat basis.

Table 6-5
PCB Concentrations in Skin and Breast Muscle of Waterfowl Collected from the Door Passage to Bailey's Harbor, Lake Michigan on September 16-17 and September 22 and 26, 1997

Species	Sex	Age	PCBs (mg/kg wet weight lipid)[a]
Common goldeneye	male	adult	3.5[b]
Ruddy duck	female	adult	4.6[b]
Common merganser	male	immature	8.3[b]
Common merganser	female	immature	373.9[b]
Common merganser	female	immature	36.3[b]
Common merganser	female	adult	27.4[b]
Common merganser	female	immature	25.7[b]
Common merganser	female	adult	30.3[b]
Common merganser	male	immature	10.8[b]
Common merganser	male	immature	16.8[b]
Red-breasted merganser	female	adult	36.9[b]
Red-breasted merganser	female	adult	11.4[b]
Red-breasted merganser	female	adult	nd
Red-breasted merganser	female	immature	25.3[b]

a. The lower limit of detection was 0.01 mg/kg wet weight, which is equivalent to 0.2 mg/kg wet weight lipid for a tissue sample of 5% lipid.
b. Exceeds federal and Wisconsin tolerance levels.

nd = not detected.

exceed the federal tolerance levels and the Wisconsin advisory level. The data also indicate that the PCB body burdens in waterfowl are determined by the residence time that the individual has spent in the system (summer resident mallards had higher PCB concentrations than migrant birds that had most likely recently arrived in the assessment area) and by their diet (individuals whose diet comprises fish generally had higher PCB concentrations than nonpiscivores).

6.3.4 The State of Wisconsin Waterfowl Consumption Advisory

In response to the PCB tissue concentrations measured in Green Bay waterfowl, the Wisconsin DNR and the Division of Health issued a waterfowl consumption advisory in 1987 (Wisconsin DNR, 1987). The advisory was for mallards taken in the "Lower Fox River from Lake Winnebago at Neenah and Menasha downstream, including Little Lake Butte des Morts, to the northeast city

limits of Kaukauna," and the "Lower Fox River from the DePere Dam to the river's mouth at Green Bay, and lower Green Bay south of a line from Point au Sable west to the west shore of Green Bay" (health advisory recommendations in annual Wisconsin DNR hunting pamphlets). The areas covered by the advisory are shown in Figure 6-2. The advisory advises hunters to "remove all skin and visible fat before cooking mallard ducks using these waters. Discard drippings or stuffings because they may retain fat that contains PCBs" (health advisory recommendations in annual Wisconsin DNR hunting pamphlets).

Since the first advisory was issued in 1987, the advisory has remained in place every year. The advisories are issued each year in the annual hunting guide distributed by the Wisconsin DNR.

The text for the advisory specifies that the advisory is being issued because of PCB contamination.

6.4 SUMMARY

The data reported in this chapter show that waterfowl from the Lower Fox River and from Green Bay are contaminated by PCBs. Resident species of waterfowl and species that feed relatively high in the food chain show the greatest body burdens. Apparently, migratory species newly arrived in the assessment area have relatively low levels of contamination. It is likely that these levels increase with the duration of their residence time in the Lower Fox River and Green Bay. PCB concentrations measured in waterfowl have and continue to exceed federal tolerance levels for poultry.

The Wisconsin DNR and the Division of Health issued a consumption advisory for mallards from the Lower Fox River and inner Green Bay in 1987 because of their elevated levels of PCBs. This advisory is still in force. The data reviewed in this chapter show that the Wisconsin DNR and Division of Health imposition of the consumption advisory on mallards is justified by the elevated concentrations of PCBs in the tissues of that species.

The elevated PCB tissue concentrations have resulted in waterfowl in the assessment area being injured based on the injury definitions at 43 CFR § 11.62(f)(1)(ii) (concentrations of hazardous substances sufficient to " exceed action or tolerance levels established under section 402 of the Food, Drug and Cosmetic Act" or "exceed levels for which an appropriate State health agency has issued directives to limit or ban consumption").

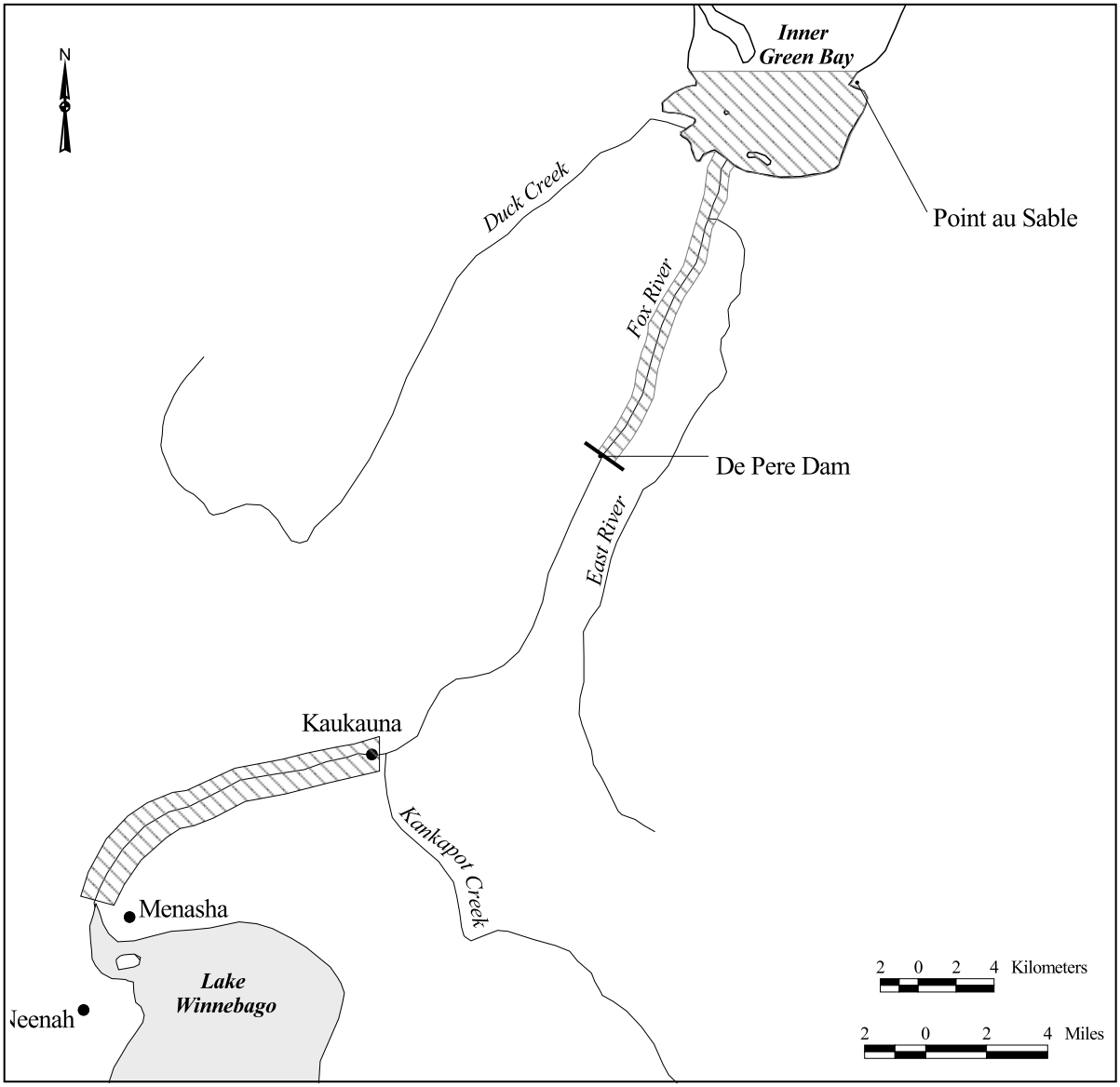


Figure 6-2. Areas covered by the Wisconsin waterfowl consumption advisory (hatched).

CHAPTER 7
WEIGHT OF EVIDENCE EVALUATION

7.1 INTRODUCTION

Chapter 5 of this report described scientific evidence that adverse effects have occurred among birds in the assessment area. As noted in that chapter, there is uncertainty regarding the extent and causes of these effects. In this chapter we carry out a weight of evidence evaluation of the scientific data to address and answer two questions:

1. ***Is it more likely than not that adverse effects that are consistent with the definitions of biological injury in the Departmental regulations [43 CFR § 11.62 (f)(1)(i)] have occurred among assessment area birds?***

We address this question by categorizing the evidence provided by each study to determine whether the case for the occurrence of each reported adverse effect is either:

- ***Highly likely.*** There is little or no doubt that the evidence reported in the study supports the conclusion that birds experienced adverse effects.

- ***Likely.*** While there may be some uncertainty associated with the evidence presented in the study, the evidence suggests that it is more likely than not that birds experienced adverse effects.

- ***Unlikely.*** The evidence indicates that it is unlikely that the reported adverse effects occurred.

- ***Indeterminate.*** Although the data in the report may indicate that adverse effects have occurred, the data do not allow an unequivocal determination. This categorization does not necessarily indicate that the adverse effects have *not* occurred.

We considered the following issues when evaluating studies of adverse effects:

- Did the study include appropriate reference areas or controls?

- How adequate were the field/laboratory methods reported in the study?

- Were sample sizes large enough to provide adequate statistical power?

- Were statistically significant differences measured between the assessment and reference/control conditions?
- Were the effects demonstrated both in the field and under controlled conditions in the laboratory?
- Do any uncertainties in the study cast doubt on the conclusions that were drawn?

2. Is it more likely than not that the adverse effects were caused by exposure to PCBs?

For those effects determined to be either highly likely or likely in the above evaluation, we evaluate causation as follows:

- ***Likely.*** While there may be some uncertainty associated with the causality, it is more likely than not that PCBs were at least a contributing factor to the adverse effect.
- ***Unlikely.*** The evidence indicates that it is not likely that PCBs caused or contributed to the adverse effect.
- ***Indeterminate.*** The data reported in the study do not allow an unequivocal determination of whether or not the adverse effects were caused by PCBs. This categorization does not necessarily indicate that the adverse effects were *not* caused by PCBs.

We considered the following when evaluating causation:

- To what extent were study results consistent with laboratory studies of the toxicology of PCBs?
- Was consistency of effects observed across studies?
- Were dose-response relationships observed?
- Were the effects consistent with definitions of injury in the Departmental NRDA regulations?
- Is there evidence that supports an alternative cause?

Lastly, we evaluated the scientific evidence from all species studied in the assessment area to determine whether there are cross-species consistencies in adverse effects that could further clarify our understanding of causation.

7.2 EVALUATION OF EVIDENCE THAT ADVERSE EFFECTS HAVE OCCURRED

7.2.1 Terns

Forster's Terns

In this analysis, we evaluate the evidence provided in two studies: Kubiak et al. (1989) and Hoffman et al. (1987). The evidence from these studies (summarized in Table 7-1 at the end of Section 7.2) supports the conclusion that it is ***highly likely*** that two of the adverse effects reported for Forster's terns in the assessment area, reduced hatching success and embryonic deformities, occurred.

The reduced hatching success was demonstrated by Kubiak et al. (1989) both in the field and under controlled conditions in the laboratory. The results of these studies leave little room for doubt that the reduced hatching success occurred. Hoffman et al. (1987) demonstrated, also under controlled conditions in the laboratory, that physical deformations also occurred in the Green Bay Forster's tern hatchlings. Statistically significant differences were found between Green Bay and reference area hatchlings for two of these deformations (femur length and liver to body weight ratios); three instances of obvious skeletal deformities were found in Green Bay chicks, but none in reference area chicks. These results provide evidence that it is highly likely that assessment area Forster's tern chicks suffered physical deformations.

The evidence for the occurrence of behavioral abnormalities in Forster's terns is deemed ***likely*** in this evaluation. The time it took Green Bay Forster's tern eggs to hatch was significantly longer than reference area Forster's tern eggs. It is very likely that this effect was caused by reduced incubation attentiveness in Green Bay adult terns. However, the incubation schedules of the terns were not measured directly and, as a result, there is uncertainty associated with this conclusion.

Common Terns

The evidence provided by Hoffman et al. (1993) was evaluated. The data supports the conclusion that it is ***likely*** that the two adverse effects reported for common terns in the assessment area, reduced hatching success and embryonic deformities, occurred (summarized in Table 7-1 at the end of Section 7.2).

Under controlled conditions in laboratory incubators Hoffman et al. (1993) found significantly lower hatching success in Green Bay eggs compared with eggs from one of the reference colonies. Hoffman et al. (1993) also demonstrated under controlled conditions in the laboratory that physical deformations occurred in the Green Bay common tern hatchlings. Statistically significant differences in femur length were found between Green Bay hatchlings and hatchlings from one of the reference colonies (the same colony that had significantly higher hatching success). The results in Hoffman et al. provide good evidence that the Green Bay common terns

had lower hatching success than eggs from the reference area. They also show that it is ***likely*** that a higher rate of deformities occurred in the Green Bay hatchlings. The only uncertainty is introduced by the fact that significant differences were found (in both metrics) for only one of the reference colonies (Cut River), but not the other (Point aux Chenes). Nevertheless, the deformity rate from the Point aux Chenes reference colony was lower (0% of hatchlings) than from Green Bay (11% of hatchlings). However, the sample size from the Point aux Chenes colony was smaller than that from Green Bay or the other reference colony (20, 35, and 35, respectively), and it is possible that this may have contributed to the lack of statistical significance.

Caspian Tern

The weight of evidence evaluation that adverse effects have occurred among Caspian terns (summarized in Table 7-1 at the end of Section 7.2) is based on data in Ludwig et al. (1996), Ludwig and Ludwig (undated report b), and Mora et al. (1993). Ludwig et al. (1996) showed that physical deformations occurred at a greater rate among Green Bay Caspian tern embryos than in embryos from reference areas. Some uncertainty is introduced into this determination by the fact that Ludwig and Ludwig (undated report b) did not find any deformities in Green Bay Caspian tern embryos. However, Ludwig and Ludwig (undated report b) focused on hatched chicks, which were shown in Ludwig et al. (1996) to have low rates of deformity. Thus, the probability of detecting effects in hatched chicks is lower. Although less conclusive than for Forster's terns, it is deemed ***likely*** that unhatched Caspian terns in the assessment area have suffered greater incidences of deformities than terns elsewhere.

The evidence that Caspian terns in the assessment area have exhibited behavioral abnormalities (Mora et al., 1993) cannot be substantiated using the data available. Although reduced site fidelity could reflect a behavioral abnormality caused by PCBs, the major effect that is measured, reduced fidelity to the nesting colony, could be a function of disturbance caused by the method used by Mora et al. (1993) to trap birds, cannon netting. Moreover, the other major method employed, analysis of band recoveries, has many potential biases, including band loss and wear, likelihood of recovery, and search effort. These biases were not adequately addressed in the study. As a result, it is concluded that the existence of behavioral effects in assessment area Caspian terns is ***indeterminate***.

7.2.2 Double-Crested Cormorants

This weight of evidence evaluation that adverse effects have occurred in double-crested cormorants in the assessment area is based on consideration of Ross and Weseloh (1988), Fox et al. (1991), Tillitt et al. (1992), Larson et al. (1996), and Ludwig et al. (1996). The results are summarized in Table 7-1 at the end of Section 7.2. These studies support the conclusion that it is ***highly likely*** that the reported adverse effects occurred in the assessment area. Of the two major studies that independently compared hatching success among Green Bay cormorants with

reference areas, both found that Green Bay hatching success was significantly lower. All three major studies that independently compared embryo/chick deformity rates in Green Bay with reference areas, found that the deformity rate in Green Bay was significantly higher. The only uncertainty concerns the "background" rate of deformities. Ross and Weseloh (1988) found anomalously high rates of head and bill defects from one small subcolony at Lake Winnipegosis. This, however, was an isolated finding, and other studies have shown that, in comparison to Green Bay, the rate of deformities at Lake Winnipegosis is typically low.

7.2.3 Bald Eagles

The weight of evidence evaluation that adverse effects have occurred in bald eagles in the assessment area is based on data in Dykstra and Meyer (1996) and in Chapter 5 of this report, and is summarized in Table 7-1 at the end of Section 7.2. Data confirm that bald eagles in the assessment area have consistently had breeding productivity that is significantly reduced compared to that in reference areas that are not exposed to point source releases of PCBs. In this evaluation, therefore, we consider it ***highly likely*** that this adverse effect occurred.

7.2.4 Black-Crowned Night Herons

The weight of evidence evaluation that adverse effects have occurred in black-crowned night herons in the assessment area is based on two studies, Hoffman et al. (1993) and Rattner et al. (1993) and is summarized in Table 7-1 at the end of Section 7.2. Only one of the studies (Hoffman et al., 1993) demonstrated physical deformations (liver to body weight ratios). A limitation of the Hoffman et al. study is that the sample size was small (n = 5). The sample size in the Rattner et al. study (in which no deformities were found) was larger (n = 18). On balance, the evidence in these studies does not provide compelling support for the conclusion that adverse effects have occurred among Green Bay black-crowned night herons and we consider the evidence ***indeterminate***.

7.2.5 Tree Swallow and Red-Breasted Merganser

No evidence that adverse effects have occurred among Green Bay tree swallows or red-breasted mergansers has been reported in the literature. These species are not considered further in this evaluation.

7.2.6 Summary

Table 7-1 summarizes the results of the weight of evidence evaluation of adverse effects. It is highly likely that adverse effects have occurred among assessment area Forster's terns, double-crested cormorants, and bald eagles. These effects comprise reproductive malfunctions (reduced hatching success), and physical deformations (head and bill deformaties). It is likely that adverse effects have occurred among assessment area common terns (reduced hatching success and physical deformations) and Caspian terns (physical deformations in unhatched chicks). It was concluded that adverse effects among black-crowned night herons, tree swallows, or red-breasted mergansers could not be substantiated using the available data.

7.3 WEIGHT OF EVIDENCE EVALUATION OF CAUSATION

In this section we evaluate whether it is more likely than not that those adverse effects identified in Section 7.2 as being highly likely or likely were caused by PCBs. The results of these evaluations are summarized in Table 7-2 at the end of Section 7.3.

7.3.1 Forster's, Common, and Caspian Terns

The data evaluated support the conclusion that it is ***likely*** that most of the adverse effects observed among assessment area Forster's and common terns have been caused, at least in part, by exposure to PCBs. This conclusion is based on the following:

- The types of effects that were observed in both species (reproductive malfunctions, deformities) are consistent with PCB toxicosis.
- Both species are likely sensitive to PCBs as discussed in Chapter 5 of this report.
- The concentrations of PCBs found in Green Bay Forster's and common tern eggs exceeded the 5-10 mg/kg toxicity range for sensitive species.
- There was a dose-response relationship established in the Kubiak et al. (1989) study.
- The study performed by Harris et al. (1993) failed to find reduced hatching success. However, at the time of this study PCB concentrations in Green Bay Forester's tern eggs were more than 50% lower than in 1983, when the Kubiak et al. (1989) study was performed. In 1988, none of the adverse effects observed in 1983 were found. Therefore, the Harris et al. study is not considered to present confounding data. Indeed, it may be suggestive of an exposure-response relationship.

Table 7-1
Weight of Evidence Evaluation that Adverse Effects Have Occurred among Assessment Area Birds

Species	Adverse Effect	Study	Support for Adverse Effect Having Occurred	Comments	Result of Evaluation
Forster's tern	Reduced hatching success	Kubiak et al., 1989	Hatching success in Green Bay significantly lower than reference colony. Hatching success of Green Bay eggs significantly lower in laboratory than reference colony eggs.	Comprehensive, rigorous study design. Reference and Green Bay eggs incubated under identical conditions in laboratory. Appropriate reference area used. Appropriate statistical tests performed.	Highly likely
	Physical deformations	Hoffman et al., 1987	Significantly shorter femurs in Green Bay hatchlings than reference colony hatchlings. Significantly greater liver to body weight ratios in Green Bay hatchlings than reference colony hatchlings. Three instances of skeletal deformities in Green Bay hatchlings but none in reference colony hatchlings.	Assessments carried out under controlled laboratory conditions. Appropriate statistical tests used to compare samples. Appropriate reference area used.	Highly likely
	Behavioral abnormalities	Kubiak et al., 1989	Significantly extended incubation periods of Green Bay clutches compared with reference area. Lake Poygan eggs incubated by Green Bay adults had low hatching success, suggesting reduced incubation attentiveness in Green Bay terns.	Appropriate reference area used. Appropriate statistical tests used to compare samples. Extended incubation periods were probably due to reduced incubation attentiveness in adult Green Bay terns. However, this is uncertain since incubation schedules were not measured.	Likely

Table 7-1 (cont.)
Weight of Evidence Evaluation that Adverse Effects Have Occurred among Assessment Area Birds

Species	Adverse Effect	Study	Support for Adverse Effect Having Occurred	Comments	Result of Evaluation
Common tern	Reduced hatching success	Hoffman et al., 1993	Hatching success of Green Bay eggs in incubators significantly lower than eggs from one of two reference colonies.	Reference and Green Bay eggs incubated under identical conditions. Assessments carried out under controlled laboratory conditions. Appropriate statistical tests performed. Uncertainty because of significant difference found for only one of the two reference colonies.	Likely
	Physical deformations	Hoffman et al., 1993	Green Bay chicks had significantly shorter femurs than chicks from one of the two reference colonies (but not the other). Four of the Green Bay neonates were deformed compared with none of the reference birds.	Assessments carried out under controlled laboratory conditions. Appropriate statistical tests performed. Uncertainty because of significant difference found for only one of the two reference colonies.	Likely/ indeterminate
Caspian tern	Physical deformations	Ludwig et al., 1996	Greater incidence of deformities found in Green Bay embryos compared with colonies in the Great Lakes not exposed to point source releases of PCBs. Deformities consistent with those observed in other species in assessment area. Age-related incidences of deformations consistent with deformations being associated with egg mortality.	More subtle deformations may be difficult to detect or correctly classify under field conditions. No abnormalities reported in other studies (Ludwig and Ludwig, undated report b).	Likely

Table 7-1 (cont.)
Weight of Evidence Evaluation that Adverse Effects Have Occurred among Assessment Area Birds

Species	Adverse Effect	Study	Support for Adverse Effect Having Occurred	Comments	Result of Evaluation
Caspian tern (cont.)	Behavioral abnormalities	Mora et al., 1993	Apparently lower site fidelity among assessment area terns than reference areas.	Study provides some evidence that Green Bay Caspian terns may have suffered behavioral abnormalities. However, two limitations on interpretation: method of capture (cannon netting) very intrusive and could, potentially, cause terns to not return to colony in future years; conclusion based on analysis of band recoveries and failed to address biases inherent in this procedures.	Indeterminate
Double-crested cormorant	Reduced hatching success	Tillitt et al., 1992 Larson et al., 1996	Hatching success at Spider Island lowest of Great Lakes colonies evaluated. Hatching success at Spider island significantly lower than at Lake Winnipegosis.	Study compared a wide range of sites, not just one reference site. Study used appropriate reference site. Used appropriate field and statistical methods.	Highly likely

Table 7-1 (cont.)
Weight of Evidence Evaluation that Adverse Effects Have Occurred among Assessment Area Birds

Species	Adverse Effect	Study	Support for Adverse Effect Having Occurred	Comments	Result of Evaluation
Double-crested cormorant (cont.)	Physical deformities	Fox et al., 1991	Highest rate of head and bill deformities found in 42 Great Lakes and other colonies was in assessment area. Rate of head and bill deformities in assessment area significantly higher than at most other colonies.	Used large number of potential reference colonies. Statistical tests appropriate. Field methods appropriate.	Highly likely
		Larson et al., 1996	Bill deformities significantly more frequent at Spider Island than Lake Winnipegosis.	Statistical tests appropriate. Field methods appropriate.	
		Ludwig et al., 1996	Deformity rate in Green Bay significantly higher than at reference colonies.	More than one reference colony evaluated. Appropriate statistical comparisons performed for this report. Some uncertainties in determination of background deformity rates (Ross and Weseloh, 1988).	

Table 7-1 (cont.)
Weight of Evidence Evaluation that Adverse Effects Have Occurred among Assessment Area Birds

Species	Adverse Effect	Study	Support for Adverse Effect Having Occurred	Comments	Result of Evaluation
Bald eagle	Reduced productivity	Dykstra and Meyer, 1996	Productivity of Green Bay bald eagles significantly lower than inland Wisconsin bald eagles.	Reference data adequate since study compared Green Bay sites with a large number of inland Wisconsin reference sites. Appropriate statistical methods used. Appropriate field methods used.	Highly likely
		This study	Productivity at Green Bay sites significantly lower than at sites in inland Wisconsin and inland Michigan.	Study compared Green Bay sites with a large number of inland Wisconsin and Michigan reference sites. Appropriate statistical methods used. Appropriate field methods used.	
Black-crowned night heron	Physical deformities	Hoffman et al., 1993	Significantly higher liver to body weight ratios in Green Bay than in reference site chicks.	Appropriate statistical methods used. Appropriate laboratory used.	Indeterminate
		Rattner at al., 1993	No deformities reported in Green Bay chicks.	Small sample size in Hoffman et al. study.	

- No alternative contaminants are likely to have caused the effects (DDE is not known to cause deformations, and PCDDs and PCDFs do not contribute substantially to the dioxin-like toxicity in the assessment area).

- Since many of the adverse effects were recorded under controlled laboratory conditions, other anthropogenic or ecological factors do not plausibly explain the results.

Although the types of deformities found in Green Bay Caspian terns by Ludwig et al. (1996) are consistent with PCB toxicosis and may have resulted from releases of PCBs into the assessment area, the evidence is equivocal, and thus we categorize it as ***indeterminate***. No relationship between PCBs and deformity rates was found in the assessment area by Yamashita et al. (1993). Also, one of the major findings of the Ludwig et al. study was that embryo deformations were likely to be associated with mortality. However, in a study that included many Caspian tern nesting sites across the Great Lakes, Ewins et al. (1994) found no relationship between egg PCB concentrations and embryo survival. Given these contradictory results from different studies, it cannot be concluded that the adverse effects observed in assessment area Caspian terns were caused by exposure to PCBs.

7.3.2 Double-Crested Cormorant

Section 7-2 concluded that it is highly likely that adverse effects (reduced hatching success and physical deformations) have occurred in assessment area double-crested cormorants. The weight of evidence evaluation in this section leads to the conclusion that it is ***likely*** that PCBs have caused, or were a significant contributing cause, of the reduced reproductive success. This conclusion is based largely on the Tillitt et al. (1992) study, which showed the following:

- Total PCB concentrations in cormorant eggs from a number of Great Lakes sites were significantly negatively correlated with hatching success.

- The correlation between contaminants in eggs and hatching success improved when H4IIE results were used as the determinate variable. This result could not have been obtained if the cause of the variation in hatching success were anything other than a dioxin-like contaminant.

- The laboratory method of sample preparation screened out PCDDs and PCDFs. Thus, these contaminants could not have contributed to the observed relationships.

- It is unlikely that any ecological or genetic factor or disease (e.g., Newcastle disease) could explain the pattern of variability in hatching success that was observed among the colonies.

The Powell et al. (1997) study failed to elicit significantly elevated embryo mortality in the laboratory by injecting Lake Winnipegosis cormorants eggs with PCB 126 at doses that exceed those observed in Green Bay. While this result is interesting and suggests that further studies need to be carried out, we consider that it does not show that PCBs do not affect hatching success in the assessment area for two reasons:

- PCB 126 is only one of the congeners that may be important in Green Bay. The contributions to toxicity by other potentially important congeners (e.g., PCB 81) were not evaluated.

- The relevance of egg injection studies to maternal transfer conditions in the field is uncertain.

Recent work by Custer et al. (in press) also demonstrates that DDE is currently affecting the hatching success of cormorants in the assessment area. However, we conclude that the Custer et al. (in press) study does not demonstrate that PCBs are *not* affecting hatching success, or did not do so in the past. In fact, the Custer et al. (in press) study did find significant relationships between egg PCB concentrations and two cormorant reproductive parameters, egg size and hatchling weight, and while the correlation between egg PCB concentrations and hatching success was not significant at $p < 0.05$, it did approach that level ($p = 0.13$). We, therefore, conclude that it is more likely than not that PCBs (possibly together with DDE) have been contributing to the reduced hatching success observed in Green Bay cormorants.

The evidence that PCBs have caused the physical deformities observed in assessment area cormorants is less certain. Although head and bill deformities are consistent with the results of laboratory studies of the effects of PCBs on birds, there are a number of other factors that could potentially cause such effects, including founder effect (reduced genetic variability due to a small colonist population with little subsequent immigration), or nutritional deficiencies. No study of deformities in Green Bay has adequately evaluated these alternative causal factors. Therefore we consider the causality to be ***indeterminate***.

7.3.3 Bald Eagles

The weight of evidence evaluation that the low productivity among bald eagles in the assessment area has been caused by PCBs is summarized in Table 7-2 at the end of Section 7.3. It is deemed ***likely*** that PCBs have contributed to the reduced productivity for the following reasons:

- The concentrations of PCBs in assessment area bald eagle eggs consistently greatly exceed the estimated toxicity range for sensitive species.

- The effect observed (embryo mortality or infertility) is consistent with PCB toxicosis.

- Human disturbance and food shortage are not contributing factors to reproductive failure in the assessment area (Dykstra and Meyer, 1996).
- There is no evidence that any other ecological factor (e.g., disease) has caused the effect, nor is it likely to do so over such a long period.
- Two alternative contaminants (PCDDs and PCDFs) are not important contributors to dioxin-like toxicity in the assessment area.

However, it cannot be concluded unequivocally that PCBs have caused the reduced productivity in assessment area bald eagles because of the potential confounding effect of DDE. While the Dykstra and Meyer (1996) study and this study have conclusively demonstrated significant negative relationships between egg PCB and DDE concentrations and productivity, neither study was able to identify the relative contributions of each. This is because PCB and DDE concentrations in Great Lakes bald eagle eggs are usually correlated. Our assessment concludes, therefore, that, based on the type of effect and the egg contaminant concentrations relative to toxicity thresholds, it is likely that both PCBs and DDE are contributing to the adverse effect, but it is not possible to identify the relative contributions.

7.4 INTER-SPECIES CONSISTENCY

The previous analyses in this chapter used a species-by-species approach to evaluate the evidence that PCB-induced adverse effects have occurred among assessment area birds. This approach, while valid, might fail to identify between-species consistencies that can further improve our understanding of the effects of contaminants in the assessment area. In this section we compare the adverse effects that have been observed among all of the species in the assessment area to determine if similarities and/or dissimilarities contribute to our understanding of causality.

The determinations presented in Table 7-3 clearly show that adverse effects caused in the laboratory and the field by PCBs were observed in every species that has been studied in detail in the assessment area. The strength of the evidence that PCBs caused these effects in birds varies from likely to indeterminate, depending on the species. However, although some studies provide only indeterminate evidence that some species may have been injured by PCBs, the consistency in effects across studies and species warns against regarding these studies as demonstrating that PCB-induced effects have *not* occurred. They only show that PCB-induced effects have not been conclusively determined.

Table 7-2
Weight of Evidence Evaluation that the Adverse Effects among Assessment Area Birds Were Caused by PCBs

Species	Adverse Effect	Study	Support for Adverse Effect Being Caused by PCBs	Result of Evaluation
Forster's tern	Reduced hatching success, physical deformations	Kubiak et al., 1989 Hoffman et al., 1987 Harris et al., 1993	Type of effects observed consistent with PCB toxicosis). Deformities not consistent with DDE toxicosis. PCB dose-response relationship established. Forster's terns likely to be sensitive to PCBs. PCB concentrations in eggs exceeded 5-10 mg/kg wet weight toxicity range for sensitive species. Low DDE concentrations in eggs (similar to concentrations in successfully reproducing Forster's terns at other sites [King et al., 1991]). Effects unlikely to be due to PCDDs or PCDFs because of their small contributions to TCDD-EQ. Harris et al. (1993) showed that reduction in PCBs in eggs associated with lack of adverse effect.	Likely
Common tern	Reduced hatching success Physical deformations	Hoffman et al., 1993	Type of effects observed consistent with PCB toxicosis. PCB dose-response relationship established. Common terns likely to be sensitive to PCBs. PCB concentrations in eggs exceeded 5-10 mg/kg toxicity range for sensitive species. No significant difference in DDE concentrations in Green Bay and reference eggs. Effects unlikely to be due to PCDDs or PCDFs because of their small contributions to TCDD-EQ.	Likely Likely/ indeterminate

Table 7-2 (cont.)
Weight of Evidence Evaluation that the Adverse Effects among Assessment Area Birds Were Caused by PCBs

Species	Adverse Effect	Study	Support for Adverse Effect Being Caused by PCBs	Result of Evaluation
Caspian tern	Physical deformations	Yamashita et al., 1993 Ludwig et al., 1996 Ewins et al., 1994	Deformities observed by Ludwig et al. consistent with PCB toxicosis, but not DDE. Effects unlikely to be due to PCDDs or PCDFs due to their small contributions to TCDD-EQ. However: No relationship between Green Bay deformities and PCB concentrations in eggs (Yamashita et al., 1993). No relationship between embryo survival and egg PCB concentrations across Great Lakes (Ewins et al., 1994).	Indeterminate

Table 7-2 (cont.)
Weight of Evidence Evaluation that the Adverse Effects among Assessment Area Birds Were Caused by PCBs

Species	Adverse Effect	Study	Support for Adverse Effect Being Caused by PCBs	Result of Evaluation
Double-crested cormorant	Reduced hatching success	Tillitt et al., 1992 Larson et al., 1996 Powell et al., 1997 Custer et al., in press	PCB dose-response relationship established. PCB concentrations in eggs exceeded 5-10 mg/kg toxicity range for sensitive species. Effects unlikely to be due to PCDDs or PCDFs. Effect shown in avian laboratory studies to be caused by PCBs but not DDE. Custer et al. (in press) found that PCBs significantly negatively correlated with egg size and hatchling weight. However: Custer et al. (in press) found significant negative correlation between DDE and hatching success but none between PCBs and hatching success (though p = 0.13).	Likely
	Physical deformations		Powell et al. (1997) unable reproduce reduction in hatching success in laboratory by injecting cormorant eggs with concentrations of PCB 126 representative of egg concentrations in Green Bay. Custer et al. (in press) showed that in 1994 and 1995 deformity rates higher among Spider Island chicks than Cat Island chicks. However, respective rates not known for any other years. Other “natural” potential causes not evaluated adequately.	Indeterminate

Table 7-2 (cont.)
Weight of Evidence Evaluation that the Adverse Effects among Assessment Area Birds Were Caused by PCBs

Species	Adverse Effect	Study	Support for Adverse Effect Being Caused by PCBs	Result of Evaluation
Bald eagle	Reduced productivity	Dykstra and Meyer, 1996 this study	PCB dose-response relationship established. PCB concentrations in eggs exceed 5-10 mg/kg toxicity range for sensitive species. PCB concentrations in eggs exceed thresholds established by Weimeyer et al. (1984) and Kubiak and Best (1991). Effects unlikely to be due to PCDDs or PCDFs due to their small contributions to TCDD-EQ. However: This and previous studies unable to separate the effects of PCBs and DDE due to their correlation in eggs.	Likely

7.5 SUMMARY

This weight of evidence evaluation of the data pertaining to the occurrence and causes of adverse effects in assessment area birds has demonstrated the following:

- Forster's, common, and Caspian terns have either suffered, or are likely to have suffered, adverse effects in the assessment area. These include low reproductive success, behavioral abnormalities, and physical deformations. The adverse effects in Forster's and common tern have more likely than not been caused by exposure to PCBs. It is uncertain whether PCBs caused the adverse effects observed in Caspian terns, though the effects are consistent with PCB toxicosis.

- Double-crested cormorants have suffered adverse effects in the assessment area. These comprise reduced hatching success and physical deformations. It is likely that PCBs have caused or contributed to the reduced reproductive success in assessment area double-crested cormorants, but the evidence linking head and bill deformities to PCBs is uncertain, although the effects are consistent with PCB toxicosis.

- Bald eagles have suffered reduced productivity in the assessment area. PCBs are likely to have caused or contributed to the reduced productivity in assessment area bald eagles. However, the relative contributions of PCBs and DDE are uncertain.

Table 7-3
Adverse Effects Documented in Assessment Area Birds and the Likelihood that They Were Caused by PCBs

Adverse Effect	Species	Evidence that Adverse Effect Occurred	Evidence that Adverse Effect Caused by PCBs
Reduced hatching success/ productivity	Forster's tern	Highly likely	Likely
	Common tern	Likely	Likely
	Double-crested cormorant	Highly likely	Likely
	Bald eagle	Highly likely	Likely
Physical deformations	Forster's tern	Highly likely	Likely
	Common tern	Likely	Likely/indeterminate
	Caspian tern	Likely	Indeterminate
	Double-crested cormorant	Highly likely	Indeterminate
	Black-crowned night heron	Indeterminate	Indeterminate
Behavioral abnormalities	Forster's tern	Likely	Likely
	Caspian tern	Indeterminate	Indeterminate

CHAPTER 8
INJURY DETERMINATION

8.1 OVERVIEW

The purpose of this chapter is to present a determination of injury for avian resources of the Lower Fox River/Green Bay assessment area. This injury determination is consistent with the components of the Departmental NRDA regulations at 43 CFR §§11.61-11.64 and is based on the data and information presented in the preceding chapters of this report. The injury determination contained herein is organized as follows:

- *Section 8.2* presents the relevant definitions of injury, as outlined in 43 CFR §11.62. These definitions of injury represent the adverse effects for which the injury determination has been conducted.

- *Section 8.3* presents the results of pathway determination, as outlined in 43 CFR §11.63. This section focuses on confirming the pathways by which assessment area birds have come to be exposed to PCBs. Separate pathway reports being prepared by the Trustees will present more detailed pathway data establishing those pathways by which PCBs have and continue to be transported throughout the environment of the assessment area.

- *Section 8.4* presents conclusions regarding the results of injury determination testing and sampling (43 CFR §11.64) and injury conclusions for the various injury definitions. Injuries to avian resources are determined in this report primarily through the use and interpretation of historical studies of birds in the assessment area. As noted previously in this report, PCB contamination in Green Bay birds was first detected in the early 1970s (Bishop et al., 1992). Since then, multiple studies have been conducted on the exposure to and accumulation of PCBs in Green Bay birds and on adverse effects resulting from this exposure. Most of these studies have been published in the peer-reviewed literature; the evaluation presented in this report is based primarily on peer-reviewed scientific papers. The previously available information was supplemented by the collection and chemical analysis of a limited number of tern eggs (12) from the Green Bay assessment area in 1996. This data collection effort, which was outlined in the NRDA Assessment Plan, is described in detail in Appendix B. Finally, the available information was evaluated using a weight-of-evidence approach (Chapter 7). The adverse effects determined to be “likely” to be caused by PCBs in Chapter 7 were considered to be injuries within the context of the injury determination.

8.2 INJURY DEFINITIONS

Chapter 3 described the types of adverse effects of PCBs on birds and discussed the relationship between these adverse effects and biological injury definitions at 43 CFR §11.62(f). Based on this information, relevant definitions of injury to avian resources of the assessment area include the following:

- ***Death.*** 43 CFR §11.62 (f)(4)(i). PCBs are known to cause embryo mortality, as manifested in reduced hatching success, reduced productivity, and embryo and chick mortality. This response is conceptually linked to the injury "reduced avian reproduction" described below.

- ***Physiological malfunctions/reduced avian reproduction.*** 43 CFR §11.62 (f)(4)(v)(B). PCBs have been found to cause reduced reproduction in various bird species. This reduced reproduction can be linked to death (through embryo or chick mortality), reduced hatching success, reduced egg fertility, reduced parental attentiveness, or other toxicological responses (see Chapter 3).

- ***Physical deformation.*** 43 CFR §11.62 (f)(4)(vi). PCBs can cause external deformations such as cross bills [43 CFR §11.62 (f)(4)(vi)(A)], skeletal deformities [43 CFR §11.62 (f)(4)(vi)(B)], and internal organ deformations [43 CFR §11.62 (f)(4)(vi)(C)].

- ***Tissue concentrations.*** 43 CFR §11.62 (f)(1)(ii-iii). Injury has occurred if concentrations of PCBs are sufficient to cause bird tissues to "exceed action or tolerance levels established under section 402 of the Food, Drug and Cosmetic Act, 21 U.S.C. 432, in edible portions of organisms" or "exceed levels for which an appropriate State health agency has issued directives to limit or ban consumption of such organism."

Injuries to birds in the assessment area are determined for each of these injury definitions in Section 8.4.

8.3 PATHWAY DETERMINATION

The purpose of the pathway determination phase is to identify the pathways by which avian resources come to be exposed to PCBs released into the assessment area.[1] As described in the Departmental regulations, pathways may be determined by demonstrating the presence of the hazardous substance in the pathway resources, or by using a model that demonstrates the routes

1. As noted previously, a separate report being prepared by the Trustees presents a complete evaluation of exposure pathways in the assessment area. The information contained in this chapter focuses on pathways by which birds are exposed to PCBs.

of exposure [43 CFR §11.63 (a)(2)]. Figure 8-1 presents the pathway diagram previously shown for avian resources in the assessment area. Table 8-1 demonstrates that PCBs have been detected at elevated concentrations in the various component pathway routes depicted in Figure 8-1, and that the spatial patterns of contamination are consistent with Fox River being the primary PCB source to the bay. Based on this information, it can be concluded that the presence of PCBs has been demonstrated in the various pathway resources that link PCB releases with avian resources.

The Green Bay Mass Balance Model also can be used to demonstrate PCB pathways to birds. The model is a multimillion dollar research effort to model the fate and transport of PCBs in the Fox River and Green Bay and their accumulation in the aquatic food chain (Connolly et al., 1992; DePinto et al., 1994). The model was constructed by numerous scientific and modeling experts from academia, government agencies, and private firms, and it has undergone extensive peer review. It provides a quantitative estimate of how PCBs move through the physical and biological compartments of Green Bay. It is based on scientific principles of PCB movement and accumulation, and was calibrated using extensive field-collected data. The model demonstrates that PCBs move through the system primarily as adherents to suspended sediment particles. Once in Green Bay, PCBs can enter the food chain through a variety of pathways, including biota ingestion of contaminated sediment and direct uptake from dissolved PCB phases in water.

Of the relevant pathway resources, the principal pathway of PCB exposure for assessment area birds is the dietary (biological) pathway. The food chain pathway is referred to as “indirect” exposure in the Departmental regulations [43 CFR §11.63(f)(2)]. Departmental regulations specify that “if indirect exposure to the biological resource has occurred . . . chemical analysis of free-ranging biological resources using one or more indicator species . . . may be performed” 43 CFR §11.63(f)(4)(ii). Thus, as demonstrated above, biological pathway determination is confirmed based both on chemical analysis of free-ranging biological resources, and on the use of a mass balance model that demonstrates the exposure routes.

In addition, Chapter 5 presented more detailed information that further confirms PCB dietary pathways to birds in the assessment area. This information included the following:

- As presented in Section 5.1.2, the diets of Forster’s, common, and Caspian terns were characterized based on known feeding behaviors and on examination of regurgitated pellets. Elevated concentrations of PCBs were measured in forage fish species that are consumed by terns. Monitoring of PCB uptake in tern chicks from hatching to fledging demonstrated that chicks reared on Kidney Island accumulated PCBs, demonstrating that chicks were being fed PCB-contaminated food and thus confirming the dietary pathway.

- As described in Section 5.2.2, the composition of cormorant diets is primarily forage fish. These prey items were shown to be contaminated with PCBs. Foraging areas were delineated, and cormorants were observed feeding in Green Bay in close proximity to their colonies. Cormorant stomachs were shown to contain fish prey that were contaminated

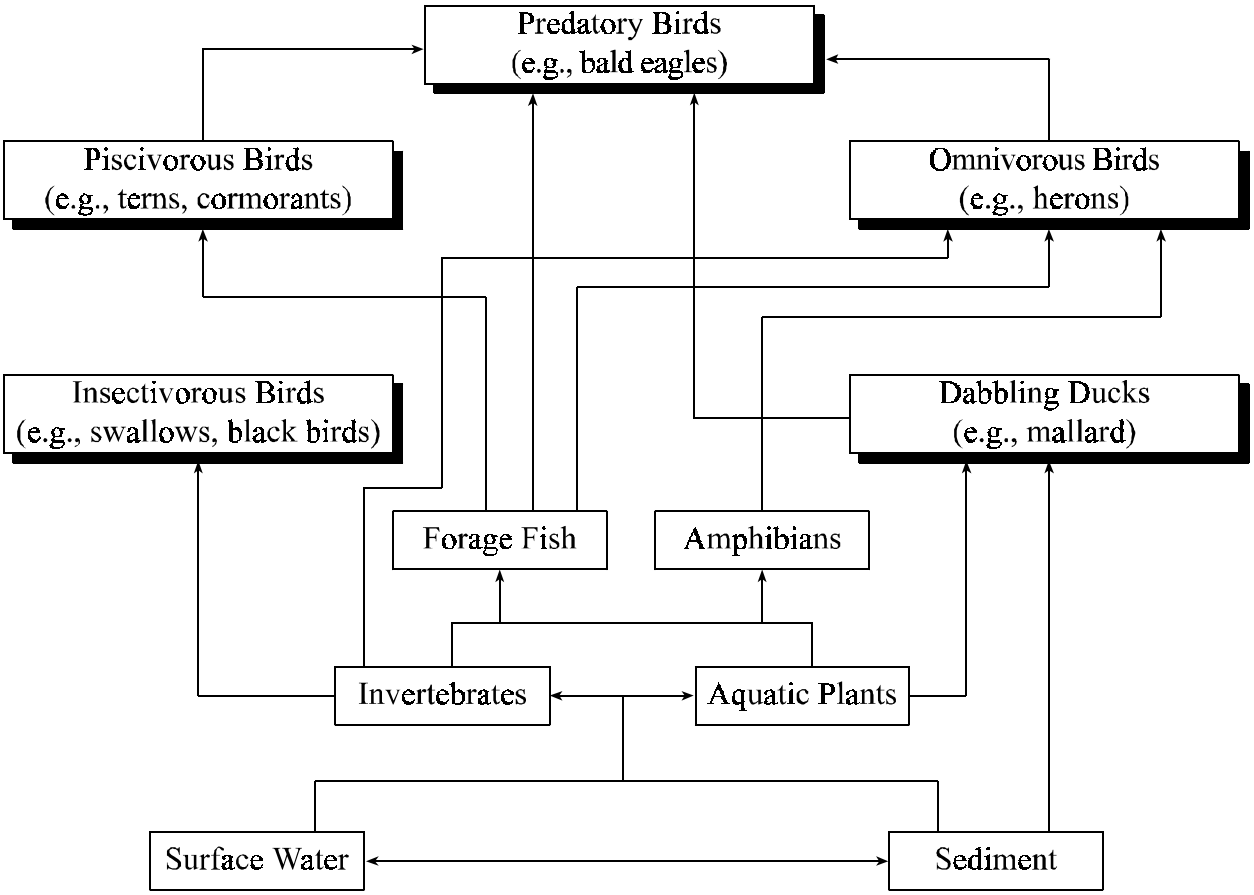


Figure 8-1. General PCB exposure pathways for assessment area birds.

with PCBs. Also, cormorant PCB tissue residues were found to increase during the breeding season while the birds were nesting in Green Bay, confirming that they were exposed to PCBs in Green Bay.

The above information — including direct measurement of PCB exposures in bird tissue and in bird prey, detailed mass balance modeling, and site-specific biological observations — is concluded to have met the requirements for pathway determination.

8.4 CONCLUSIONS OF INJURY DETERMINATION TESTING AND SAMPLING

This section summarizes the conclusions derived from the weight of evidence evaluation of those studies that comprise the injury determination testing and sampling. The methods used to

Table 8-1
Examples of PCB Concentrations Measured in Assessment Area Pathway Resources

Pathway Resource	Location	PCB Concentration	Source
Sediment	Inner bay, east side	1,600 µg/kg dry weight (averaged over top 3 cm)	Manchester-Neesvig et al., 1996
Surface water	Fox River	21.7 ng/L dissolved (mean)	Connolly et al., 1992
	Inner bay, east side	8.5 ng/L dissolved (mean)	
	Inner bay, west side	4.2 ng/L dissolved (mean)	
	Middle bay, east side	1.7 ng/L dissolved (mean)	
	Middle bay, west side	1.8 ng/L dissolved (mean)	
	Outer bay	0.6 ng/L dissolved (mean)	
Phytoplankton (aquatic plants)	Bay-wide average	~4-12 µg/kg dry weight, depending on time of year	Connolly et al., 1992
Zooplankton (invertebrates)	Fox River	~600 µg/kg dry weight (mean)	Connolly et al., 1992
	Outer bay	~60 µg/kg dry weight (mean)	
Forage fish (alewife)	Fox River	2,100 µg/kg dry weight (mean)	Connolly et al., 1992
	Inner bay, east side	1,800 µg/kg dry weight (mean)	
	Inner bay, west side	1,400 µg/kg dry weight (mean)	
	Middle bay, east side	1,300 µg/kg dry weight (mean)	
	Middle bay, west side	680 µg/kg dry weight (mean)	
	Outer bay	520 µg/kg dry weight (mean)	

determine injuries to avian resources are consistent with those contained in the Departmental regulations for NRDA [43 CFR §11.64]. Specifically, the approach relies on the use of previously collected data as outlined in the assessment plan [43 CFR §11.64 (a)(2)] and therefore is cost-effective [43 CFR §11.64 (a)(3)(ii)]. Moreover, the various studies relied upon methods that were applied to "biological responses that have satisfied the acceptance criteria of Sec. 11.62(f)(2)" and applied approaches "that have been documented and are applicable to the biological response being tested" [43 CFR §11.64(f)(2)(I-ii)]. Most of the studies relied upon in the injury evaluation have been published in the peer-reviewed literature; therefore, the methods are appropriately documented and were deemed applicable.

The conclusions derived from the evaluation of the testing and sampling data indicate that avian resources of the Lower Fox River/Green Bay assessment area have been injured. Specifically, various fish-eating birds in the assessment area, including Forster's terns, common terns, double-crested cormorants, and bald eagles have been injured as a result of exposure to PCBs. The injuries documented in the preceding chapters of this report include death [43 CFR §11.62

(f)(4)(I)] and reduced reproduction [43 CFR §11.62 (f)(4)(v)(B)],[2] as well as physical deformations [43 CFR §11.62 (f)(4)(vi)]. Waterfowl are also injured by exposure to PCBs in the assessment area. This injury comprises exceedences of tissue action or tolerance levels and waterfowl consumption advisories [43 CFR §11.62 (f)(1)(ii-iii)].

2. The injury categories "death" and "reduced avian reproduction" are effectively equivalent in this case. As noted in previous report chapters, available information suggests that mortality in assessment area birds is limited to bird embryos/chicks and this mortality contributes to reduced avian reproduction. Therefore, the two injury definitions are presented together.

CHAPTER 9
REFERENCES

Amundson, T.E. Undated. Environmental Contaminant Monitoring of Wisconsin Wild Game 1985-86. Report of Bureau of Wildlife Management, Wisconsin Department of Natural Resources, Madison.

Ankley, G.T., G.J. Niemi, K.B. Lodge, H.J. Harris, D.L. Beaver, D.E. Tillitt, T.R. Schwartz, J.P. Giesy, P.D. Jones, and C. Hagley. 1993. Uptake of planar polychlorinated biphenyls and 2,3,7,8-substituted polychlorinated dibenzofurans and dibenzo-*p*-dioxins by birds nesting in the lower Fox River and Green Bay, Wisconsin, USA. *Arch. Environ. Contam. Toxicol.* 24: 332-344.

Barron, M.G., H. Galbraith, and D. Beltman. 1995. Comparative reproductive and developmental toxicology of PCBs in birds. *Comp. Biochem. Physiol.* 112C: 1-14.

Barron, M.G., M. Anderson, D. Beltman, T. Podrabsky, W. Walsh, D. Cacela, J. Lipton, S.J. Teh, D.E. Hinton, J.T. Zelikoff, A.L. Dikkeboom, D.E. Tillitt, M. Holey, P. Bouchard, and N. Denslow. 1999. Association between PCBs, Liver Lesions, and Biomarker Responses in Adult Walleye (*Stizostedium vitreum*) Collected from Green Bay, Wisconsin. Prepared by Stratus Consulting Inc., Boulder, CO, for the U.S. Fish and Wildlife Service, Green Bay Field Office.

Bath, A.J. 1991. Kaukauna, Wisconsin Bald Eagle Nest Site — A Management Plan. Unpubl. Report. Stevens Point, WI.

Becker, P.H., S. Schuhmann, and C. Koepff. 1993. Hatching failure in common terns (*Sterna hirundo*) in relation to environmental chemicals. *Environ. Pollut.* 79: 207-213.

Belonger, B. 1983. Double-crested cormorant study results and recommendations. State of Wisconsin memorandum to C. Higgs dated December 23.

Best, D.A., W.W. Bowerman, T.J. Kubiak, S.R. Winterstein, S. Postupalsky, M.C. Shieldcastle, and J.P. Giesy. 1994. Reproductive impairment of bald eagles along the Great Lakes shorelines of Michigan and Ohio. In *Raptor Conservation Today*. B.U. Meyerburg and R.D. Chancellor (eds.), World Working Group on Birds of Prey and The Pica Press, East Sussex, Great Britain, pp. 697-701.

Beyer, W.N., L.J. Blus, C.J. Henny, and D. Audet. 1997. The role of sediment ingestion in exposing wood ducks to lead. *Ecotoxicology* 6: 181-186.

Beyer, W.N., D.J. Audet, A. Morton, J.K. Campbell, and L. Captain. 1998. Lead exposure of waterfowl ingesting Coeur d' Alene River Basin sediments. *J. Environ. Qual.* 27: 1533-1538.

Bird, D.M., P.H. Tucker, G.A. Fox, and P.C. Lague. 1983. Synergistic effects of Aroclor 1254 and mirex on the semen characteristics of American kestrels. *Arch. Environ. Contam. Toxicol.* 12: 633-640.

Bishop, C.A., D.V. Weseloh, N.M. Burgess, J. Struger, R.J. Norstrom, R. Turle, and K.A. Logan. 1992. *An Atlas of Contaminants in Eggs of Fish-Eating Colonial Birds of the Great Lakes (1970-1988).* Vols. I and II. Environment Canada. Canadian Wildlife Service. Technical Report Series No. 152.

Bosveld, A.T.C. 1995. Effects of Polyhalogenated Aromatic Hydrocarbons on Piscivorous Avian Wildlife. Doctoral thesis, University of Utrecht.

Bosveld, A.T.C. and M. Van den Berg. 1994. Effects of polychlorinated biphenyls (PCBs), dibenzo-p-dioxins (PCDDs), and dibenzofurans (PCDFs) on fish-eating birds. *Environ. Rev.* 2: 147-166.

Bosveld, A.T.C., J. Gardener, A.J. Murk, A. Brouwer, M. Van Kampen, E.H.G. Evers, and M. van den Berg. 1995. Effects of PCDDs, PCDFs and PCBs in common tern (*Sterna hirundo*) breeding in estuarine and coastal colonies in the Netherlands and Belgium. *Environ. Toxicol. Chem.* 14: 99-115.

Bowerman, W.W. 1993. Regulation of Bald Eagle (*Haliaeetus leucocephalus*) Productivity in the Great Lakes Basin: An Ecological and Toxicological Approach. PhD Dissertation, Michigan State University, East Lansing, 291 pp.

Bowerman, W.W., J.P. Giesy, D.A. Best, and V.J. Kramer. 1995. A review of factors affecting productivity of bald eagles in the Great Lakes region: Implications for recovery. *Environ. Health Perspect.* 103(Suppl 4): 51-59.

Bowerman, W.W., D.A. Best, T.J. Kubiak, M.S. Martell, S. Postupalsky, and J.P. Giesy, Jr. 1994a. The influence of environmental contaminants on bald eagle populations in the Laurentian Great Lakes, North America. In *Raptor Conservation Today*, B.U. Meyburg and R.D. Chancellor (eds.), World Working Group on Birds of Prey and The Pica Press, East Sussex, Great Britain, pp. 703-707.

Bowerman, W.W., T.J. Kubiak, J.B. Holt, D.E. Evans, R.J. Eckstein, and C.R. Sindelar. 1994b. Observed abnormalities in mandibles of nestling bald eagles. *Bull. Environ. Contam. Toxicol.* 53: 450-457.

Braune, B.M. and R.J. Norstrom. 1989. Dynamics of organochlorine compounds in herring gulls: III. Tissue distribution and bioaccumulation in Lake Ontario gulls. *Environ. Toxicol. Chem.* 8: 957-968.

Brewer, R., G.A. McPeak, and R.J. Adams. 1991. *The Atlas of Breeding Birds of Michigan.* Michigan State University Press, East Lansing.

Britton, W.M. and T.M. Huston. 1973. Influence of polychlorinated biphenyls in the laying hen. *Poult. Sci.* 32: 1620-1624.

Broley, C.L. 1958. The plight of the American bald eagle. *Audubon Mag.* 60: 162-163.

Brouwer, A. 1991. Role of biotransformation in PCB-induced alterations in vitamin A and thyroid hormone metabolism in laboratory and wildlife species. In *The Role of Enzymes in Regulating the Toxicity of Xenobiotics.* Reading University, Reading, PA, pp. 731-737.

Brunstrom, B. 1988. Sensitivity of embryos from duck, goose, herring gull, and various chicken breeds to 3,3',4',4'-tetrachlorobiphenyl. *Poultry Sci.* 67: 52-57.

Brunstrom, B. 1989. Toxicity of coplanar polychlorinated biphenyls in avian embryos. *Chemosphere* 19: 765-768.

Brunstrom, B. 1990. Mono-ortho-chlorinated chlorobiphenyls: Toxicity and induction of 7-ethoxyresorufin O-deethylase (EROD) activity in chick embryos. *Arch. Toxicol.* 64: 188-192.

Brunstrom, B. and L. Andersson. 1988. Toxicity and 7-ethoxyresorufin O-deethylase-inducing potency of coplanar polychlorinated biphenyls (PCBs) in chick embryos. *Arch. Toxicol.* 62: 263-266.

Brunstrom, B. and J. Lund. 1988. Differences between chick and turkey embryos in sensitivity to 3,3',4,4'-tetrachlorobiphenyl and in concentration/affinity of the hepatic receptor for 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Comp. Biochem. Physiol.* 91C: 507-512.

Brunstrom, B. and L. Reutergårdh. 1986. Differences in sensitivity of some avian species to the embryotoxicity of a PCB, 3,3',4,4'-tetrachlorobiphenyl, injected into the eggs. *Environ. Pollut. (Series A)* 42: 37-45.

Bruzner, J. and W. DeVita. 1998. PCBs, DDE, and mercury in young-of-the-year littoral fishes from Green Bay, Lake Michigan. *J. Great Lakes Res.* 24(1): 83-92.

Burger, J. and M. Gochfeld. 1991. *The Common Tern: Its Breeding Biology and Social Behavior.* Columbia University Press. New York, NY.

Burness, G.P., R.D. Morris, and J.P. Bruce. 1994. Seasonal and annual variation in brood attendance, prey type delivered to chicks, and foraging patterns of male common terns (*Sterna hirundo*). *Can. J. Zool.* 72: 1243-1251.

Carlson, R.W. and R.T. Duby. 1973. Embryotoxic effects of three PCBs in the chicken. *Bull. Environ. Contam. Toxicol.* 9: 261-266.

Carr, R.A., R.L. Durfee, and A.G. McKay. 1977. PCBs Involvement in the Pulp and Paper Industry. Report prepared by Versar, Inc., Springfield, VA, for U.S. EPA, Washington, DC, February 25.

Chen, S.-W., P.J. Dziuk, and B.M. Francis. 1994. Effect of four environmental toxicants on plasma Ca and estradiol 17*B* and hepatic P450 in laying hens. *Environ. Toxicol. Chem.* 13: 789-796.

Choksi, N.Y., P.R.S. Kodavanti, and R.G. Booth. 1997. Effects of polychlorinated biphenyls (PCBs) on brain tyrosine hydroxylase activity and dopamine synthesis in rats. *Fundamen. Appl. Toxicol.* 39: 76-80.

Clark, K.E., L.J. Niles, and W. Stansley. 1998. Environmental contaminants associated with reproductive failure in bald eagle eggs in New Jersey. *Bull. Environ. Contam. Toxicol.* 61: 247-254.

Colborn, T. 1991. Epidemiology of Great Lakes bald eagles. *J. Toxicol. Environ. Health.* 33: 395-453.

Colborn, T., F.S. vom Saal, and A.M. Soto. 1993. Developmental effects of endocrine-disrupting chemicals in wildlife and humans. *Environ. Health Perspect.* 101: 378-384.

Connolly, J.F., T.F. Parkerton, J.D. Quadrine, S.T. Taylor, and A.J. Thumann. 1992. Development and Application of a Model of PCBs in the Green Bay, Lake Michigan Walleye and Brown Trout and their Food Webs: 200. Report prepared for the U.S. EPA Large Lakes Research Station, Grosse lle, MI. October 2.

Connor, K., K. Ramamoorthy, M. Moore, M. Mustain, I. Chen, S. Safe, T. Zacharewski, B. Gillesby, A. Joyeu, and P. Balaguer. 1997. Hydroxylated polychlorinated biphenyls (PCBs) as estrogens and antiestrogens: Structure-activity relationships. *Toxicol. Appl. Pharmacol.* 145: 111-123.

Courtney, P. and H. Blokpoel. 1980. Food and indicators of food availability for common terns on the Lower Great Lakes. *Can. J. Zool.* 58: 1318-1323.

Cramp, S. 1985. *Handbook of the Birds of Europe, the Middle East and North Africa. Birds of the Western Palearctic. Volume IV: Terns to Woodpeckers.* Oxford University Press. Oxford, U.K.

Cramp, S. and K.E.L. Simmons. 1977. *Handbook of the Birds of Europe, the Middle East and North Africa. Birds of the Western Palearctic. Vol I.* Oxford University Press, Oxford, U.K.

Curtis, J.T. 1959. *The Vegetation of Wisconsin.* University of Wisconsin Press, Madison.

Custer, C.M., T.W. Custer, P.D. Allen, K.L. Stromborg, and M.J. Melancon. 1998. Reproduction and environmental contamination in tree swallows nesting in the Fox River drainage and Green Bay, Wisconsin, USA. *Environ. Toxicol. Chem.* 17(9): 1786-1798.

Custer, T.W. and C. Bunck. 1992. Feeding flights of breeding double-crested cormorants at two Wisconsin colonies. *J. Field Ornithol.* 63: 303-311.

Custer, T.W. and C.M. Custer. 1995. Transfer and accumulation of organochlorines from black-crowned night heron eggs to chicks. *Environ. Toxicol. Chem.* 14: 533-536.

Custer, T.W., C.M. Custer, and K.L. Stromborg. 1997. Distribution of organochlorine contaminants in double-crested cormorant eggs and sibling embryos. *Environ. Toxicol. Chem.* 16: 1646-1649.

Custer, T.W., C.M. Custer, R.K. Hines, S. Gutreuter, K.L. Stromborg, P.D. Allen, and M.J. Melancon. In press. Organochlorine contaminant effects on double-crested cormorants nesting in Green Bay, Wisconsin. *Environ. Toxicol. Chem.*

Dahlgren, R.B. and R.L. Linder. 1971. Effects of polychlorinated biphenyls on pheasant reproduction, behavior, and survival. *J. Wildl. Manag.* 35: 315-319.

Dahlgren, R.B., R.L. Linder, and C.W. Carlson. 1972a. Polychlorinated biphenyls: Their effects on penned pheasants. *Environ. Health Perspect.* 1: 89-101.

Dahlgren, R.B., R.J. Bury, R.L. Linder, and R.F. Reidinger. 1972b. Residue levels and histopathology in pheasants given polychlorinated biphenyls. *J. Wildl. Manag.* 36: 524-533.

DePinto, J.V., R. Raghunathan, P. Sierzenga, X. Zhang, V.J. Bierman, P.W. Rodgers, and T.C. Young. 1994. Recalibration of GBTOX: An Integrated Exposure Model for Toxic Chemicals in Green Bay, Lake Michigan. Draft Final Report. Prepared for the U.S. EPA, Large Lakes and Rivers Research Branch, Grosse lle, MI. March 1.

Dolbeer, R.A. 1991. Migration patterns of double-crested cormorants east of the Rocky Mountains. *J. Field Ornithol.* 62: 83-93.

Donaldson, G.M., J.L. Shutt, and P. Hunter. 1999. Organochlorine contamination in bald eagle eggs and nestlings from the Canadian Great Lakes. *Arch. Environ. Contam. Toxicol.* 36: 70-80.

Dunstan, T.C. and J.F. Harper. 1975. Food habits of bald eagles in north-central Minnesota. *J. Wildl. Manag.* 39(1): 140-143.

Dykstra, C.J.R. and M.W. Meyer. 1996. Effects of Contaminants on Reproduction of Bald Eagles on Green Bay, Lake Michigan. Report to U.S. Fish and Wildlife Service and Wisconsin Department of Natural Resources.

Ehrlich, P.R., D.S. Dobben, and D. Wildeye. 1988. *The Birder's Handbook.* Simon and Schuster, New York.

Eisler, R. 1986. Polychlorinated Biphenyl Hazards to Fish, Wildlife, and Invertebrates: A Synoptic Review. Biological Report 85(1.7), Contaminant Hazard Reviews Report No. 7. U.S. Department of the Interior, Washington, DC.

Elliott, J.E., S.W. Kennedy, and A. Lorenzen. 1997. Comparative toxicity of polychlorinated biphenyls to Japanese quail (*Coturnix c. japonica)* and American kestrels *(Falco sparverius)*. *J. Toxicol. Environ. Health.* 51: 57-75.

Elliott, J.E., S.W. Kennedy, D.B. Peakall, and H. Won. 1990. Polychlorinated biphenyl (PCB) effects on hepatic mixed function oxidases and porphyria in birds. I. Japanese quail. *Comp. Biochem. Physiol.* 96C: 205-210.

Elliott, J.E., R.J. Norstrom, A. Lorenzen, L.E. Hart, H. Philibert, S.W. Kennedy, J.J. Stegeman, G.D. Bellward, and K.M. Cheng. 1996. Biological effects of polychlorinated dibenzo-p-dioxins, dibenzofurans, and biphenyls in bald eagle (*Haliaeetus leucocephalus*) chicks. *Environ. Toxicol. Chem.* 15: 782-793.

Environment Canada. 1991. Toxic Chemicals in the Great Lakes and Associated Effects. Minister of Supply and Services Canada.

Environment Canada. 1995. The rise of the double-crested cormorant on the Great Lakes: Winning the war against contaminants. Great Lakes Fact Sheet. Environment Canada, Canadian Wildlife Service, Burlington, Ontario.

Erdman, T.C. and J.P. Jacobs. 1991. Birds of Brown County. Richter Natural History Museum, Research Report No. 3. University of Wisconsin-Green Bay.

Erickson, M.D. 1997. *Analytical Chemistry of PCBs.* Lewis Publishers, New York.

Ewins, P.J., C.V. Weseloh, R.J. Norstrom, K. Legierse, H.J. Auman, and J.P. Ludwig. 1994. Caspian terns on the Great Lakes: Organochlorine contamination, reproduction, diet, and population changes, 1972-91. Canadian Wildlife Service Occasional Paper Number 85.

Fisher, L.J., R.F. Seegal, P.E. Ganey, I.N. Pessah, and P.R.S. Kodavanti. 1998. Symposium overview: Toxicity of non-coplanar PCBs. *Toxicol. Sci.* 41: 49-61.

Fowles, J.R., A. Fairbrother, K.A. Trust, and N.I. Kerkvliet. 1997. Effects of Aroclor 1254 on the thyroid gland, immune function, and hepatic cytochrome P450 activity in mallards. *Environ. Res.* 75: 119-129.

Fox, G.A. 1976. Eggshell quality: its ecological and physiological significance in a DDE-contaminated common tern population. *Wilson Bull.* 88: 459-477.

Fox, G.A., A.P. Gilman, D.B. Peakall, and F.W. Anderka. 1978. Behavioral abnormalities of nesting Lake Ontario herring gulls. *J. Wildl. Manag.* 42: 477-483.

Fox, G.A., S.W. Kennedy, R.J. Norstrom, and D.C. Wigfield. 1988. Porphyria in herring gulls: A biochemical response to chemical contamination of Great Lakes food chains. *Environ. Toxicol. Chem.* 7: 831-839.

Fox, G.A., B. Collins, E. Hayakawa, D.V. Weseloh, J.P. Ludwig, T.J. Kubiak, and T.C. Erdman. 1991. Reproductive outcomes in colonial fish-eating birds: A biomarker for developmental toxicants in the Great Lakes food chains. II. Spatial variation in the occurrence and prevalence of bill defects in young double-crested cormorants in the Great Lakes, 1979-1987. *J. Great Lakes Res.* 17: 158-167.

Fraser, G.S. 1994. Feeding and nesting behaviors of the Forster's tern on Lake Osakis, Minnesota. Masters Thesis to North Dakota State University, Fargo. April.

Friend, M. and D.O. Trainer. 1970. Polychlorinated biphenyl: Interaction with duck hepatic virus. *Science.* 170: 1314-1316.

Fry, D.M. 1995. Reproductive effects in birds exposed to pesticides and industrial chemicals. *Environ. Health Perspect.* 103(Suppl. 7): 165-171.

Garner, C.E., W.N. Jefferson, L.T. Burka, H.B. Matthews, and R.R. Newbold. 1999. In vitro estrogenicity of the catechol metabolites of selected polychlorinated biphenyls. *Toxicol. Appl. Pharmacol.* 154: 188-197.

Giesy, J.P., J.P. Ludwig, and D.E. Tillitt. 1994. Deformities in birds of the Great Lakes region: Assigning causality. *Environ. Sci. Technol.* 28: 128A-135A.

Gilbertson, M., T. Kubiak, J. Ludwig, and G. Fox. 1991. Great Lakes Embryo Mortality, Edema and Deformities Syndrome (GLEMEDS) in colonial fish-eating birds: similarity to chick-edema disease. *J. Toxicol. Environ. Health.* 33: 455-520.

Goldstein, J.A., J.D. McKinney, G.W. Lucier, P. Hickman, H. Bergman, and J.A. Moore. 1976. Toxicological assessment of hexa chloro bi phenyl isomers and 2,3,7,8 tetra chloro di benzo furan in chicks. Part 2: Effects of drug metabolism and porphyrin accumulation. *Toxicol. Appl. Pharmacol.* 36(1): 81-92.

Gould, J.C., K.R. Cooper, and C.G. Scanes. 1997. Effects of polychlorinated biphenyl mixtures and three specific congeners on growth and circulating growth-related hormones. *Gen. Compar. Endocrinol.* 106: 221-230.

Grau, C.R. 1996. Nutritional needs for egg formation in the shag (*Phalacrocorax aristotelis*). *Ibis.* 138: 756-764.

Grier, J.W. 1972. Current status of the bald and golden eagles in North America: A summary of information and opinions from field researchers. In Transactions of the 36th Federal-Provincial Wildlife Conference, pp. 76-78.

Grier, J.W. 1982. Ban of DDT and subsequent recovery of reproduction in bald eagles. *Science* 212: 1232-1235.

Guillette, L.J., Jr., D.A. Crain, A.A. Rooney, and D.B. Pickford. 1995. Organization versus activation: The role of endocrine disrupting contaminants (EDCs) during embryonic development in wildlife. *Environ. Health Perspect.* 103(Suppl. 7): 157-164.

Harris, H.J., T.C. Erdman, G.T. Ankley, and K.B. Lodge. 1993. Measures of reproductive success and polychlorinated biphenyl residues in eggs and chicks of Forster's terns on Green Bay, Lake Michigan, Wisconsin — 1988. *Arch. Environ. Contam. Toxicol.* 25: 304-314.

Hart, C.A., M.E. Hahn, I.C.T. Nisbet, M.J. Moore, S.W. Kennedy, and D.M. Fry. 1998. Feminization in common terns (*Sterna hirundo*): Relationship to dioxin equivalents and estrogenic compounds. *Mar. Environ. Res.* 46: 174-175.

Hart, L.E., K.M. Cheng, P.E. Whitehead, R.M. Shah, R.J. Lewis, S.R. Ruschkowski, R.W. Blair, D.C. Bennett, S.M. Bandiera, R.J. Norstrom, and G.D. Bellward. 1991. Dioxin contamination and growth and development in great blue heron embryos. *J. Toxicol. Environ. Health.* 32: 331-344.

Haseltine, S.D. and R.M. Prouty. 1980. Aroclor 1242 and reproductive success of adult mallards (*Anas platyrhynchos*). *Environ. Res.* 23: 29-34.

Haseltine, S.D., Z.G.H Heinz, W.L. Reichel, and J.F. Moore. 1981. Organochlorine and metal residues in eggs of waterfowl nesting on islands in Lake Michigan off Door County, Wisconsin, 1977-78. *Pest. Monitor. J.* 15: 90-97.

Hatch, J.J. 1995. Changing populations of double-crested cormorants. *Colon. Waterbirds.* 18: 8-24.

Heath, R.G., J.W. Span, E.F. Hill, and J.F. Kreitzer. 1972. Comparative Dietary Toxicities of Pesticides to Birds. Special Scientific Report Wildlife No. 152, U.S. Department of Interior, Washington, DC.

Heinz, G.H., E.F. Hill, and J.F. Contrera. 1980. Dopamine and norepinephrine depletion in ring doves fed DDE, dieldrin, and Aroclor 1254. *Toxic. Appl. Pharmacol.* 53: 75-82.

Heinz, G.H., T.C. Erdman, S.D. Haseltine, and C. Stafford. 1985. Contaminant levels in colonial waterbirds from Green Bay and Lake Michigan, 1975-80. *Environ. Monitor. Assess.* 5: 223-236.

Heinz, G.H., S.D. Haseltine, W.L. Reichel, and G.L. Hensler. 1983. Relationships of environmental contaminants to reproductive success in red-breasted mergansers (*Mergus serrator*) from Lake Michigan. *Environ. Pollut. Ser. A.* 32: 211-232.

Heinz, G.H., D.S. Miller, B.J. Ebert, and K.L. Stromborg. 1994. Declines in organochlorines in eggs of red-breasted mergansers from Lake Michigan, 1977-1978 versus 1990. *Environ. Monitor. Assess.* 33: 175-182.

Helander, B., M. Olsson, and L. Reutergårdh. 1982. Residue levels of organochlorine and mercury compounds in unhatched eggs and the relationships to breeding success in white-tailed sea eagles *Haliaeetus albicilla* in Sweden. *Holarct. Ecol.* 5: 349-366.

Helander, B., A. Olsson, A. Bignert, K. Litzén, L. Asplund, and A. Bergman. 1998. Abnormal dehydration in eggs of the white-tailed sea eagle (*Haliaeetus albicilla*) in Sweden — a persisting effect from high exposure to organochlorines? *Organohal. Comp.* 39: 423-426.

Henshel, D.S. 1998. Developmental neurotoxic effects on dioxin and dioxin-like compounds on domestic and wild avian species. *Environ. Toxicol. Chem.* 17: 88-98.

Henshel, D.S., J.W. Martin, R.J. Norstrom, J. Elliot, K.M. Cheng, and J. DeWitt. 1997. Morphometric brain abnormalities in double-crested comorant chicks exposed to polychlorinated dibenzo-p-dioxins, dibenzofurans, and biphenyls. *J. Great Lakes Res.* 23: 11-26.

Hobson, K.A., R.W. Knapton, and W. Lysack. 1989. Population, diet and reproductive success of double-crested cormorants breeding on Lake Winnipegosis, Manitoba, in 1987. *Colon. Waterbirds.* 12: 191-197.

Hoffman, D.J., C.P. Rice, and T.J. Kubiak. 1996a. PCBs and dioxins in birds. Chapter 7 in *Environmental Contaminants in Wildlife,* W.N. Beyer, G.H. Heinz, A.W. Redmon-Norwood, eds. Lewis Publishers, Boca Raton, FL.

Hoffman, D.J., G.J. Smith, and B.A. Rattner. 1993. Biomarkers of contaminant exposure in common terns and black-crowned night herons in the Great Lakes. *Environ. Toxicol. Chem.* 17: 1095-1103.

Hoffman, D.J., M.J. Melancon, J.D. Eisemann, and P.N. Klein. 1995. Comparative developmental toxicity of planar PCB congeners by egg injection. In *Proceedings, 2nd SETAC World Congress,* Vancouver, BC, Canada, November 5-9, p. 207.

Hoffman, D.J., M.J. Melancon, J.D. Eisemann, and P.N. Klein. 1998. Comparative developmental toxicity of planar PCB congeners in chickens, American kestrels, and common terns. *Environ. Toxicol. Chem.* 17: 747-757.

Hoffman, D.J., B.A. Rattner, L. Sileo, D. Docherty, and T. Kubiak. 1987. Embryotoxicity, teratogenicity, and aryl hydrocarbon hydroxylase activity in Forster's terns on Green Bay, Lake Michigan. *Environ. Res.* 42: 176-184.

Hoffman, D.J., B.A. Rattner, C.M. Bunck, A. Krynitsky, H.M. Ohlendorf, and R.W. Lowe. 1986. Association between PCBs and lower embryonic weight in black-crowned night herons in San Francisco Bay. *J. Toxicol. Environ. Health.* 19: 383-391.

Hoffman, D.J., M.J. Melancon, P.N. Klein, C.P. Rice, J.D. Eisemann, R.K. Hines, J.W. Spann, and G.W. Pendleton. 1996b. Developmental toxicity of PCB 126 (3,3',4,4',5-pentachlorobiphenyl) in nestling American kestrels (*Falco sparverius*). *Fund. Appl. Toxicol.* 34: 188-200.

Howe, R.W., A.T. Wolf, and T.C. Erdman. 1993. A Study of Wintering Bald Eagles on the Fox River of Northeast Wisconsin. Report to REPAP, Wisconsin, Inc.

Howell, J.C. 1963. The 1961 status of some bald eagle nest sites in east-central Florida. *The Auk* 79: 716-718.

Hughes, K.D., D.V. Weseloh, and B.M. Braune. 1998. The ratio of DDE to PCB concentrations in Great Lakes herring gull eggs and its use in interpreting contaminants data. *J. Great Lakes Res.* 24(1): 12-31.

Jansen, H.T., P.S. Cooke, J. Porcelli, T.-C. Liu, and L.G. Hansen. 1993. Estrogenic and antiestrogenic actions of PCBs in the female rat: In vitro and in vivo studies. *Repro. Toxicol.* 7: 237-248.

Janz, D.M. and G.D. Bellward. 1996. In ovo 2,3,7,8-tetrachloro-p-dioxin exposure in three avian species. 1. Effects on thyroid hormones and growth during the perinatal period. *Toxicol. Appl. Pharmacol.* 139: 281-291.

Jefferies, D.J. and J.L.F. Parslow. 1976. Thyroid changes in PCB-dosed guillemots and their indication of one of the mechanisms of action of these materials. *Environ. Pollut.* 10: 295-311.

Jones, P.D., J.P. Giesy, J.L. Newstead, D.A. Verbrugge, D.L. Beaver, G.T. Ankley, D.E. Tillitt, K.B. Lodge, and G.J. Niemi. 1993. 2,3,7,8-Tetrachlorodibenzo-*p*-dioxin equivalents in tissues of birds at Green Bay, Wisconsin, USA. *Arch. Environ. Contam. Toxicol.* 24: 345-354.

Kennedy, S.W., A. Lorenzen, and R.J. Norstrom. 1996a. Chicken embryo hepatocyte bioassay for measuring cytochrome P4501A-based 2,3,7,8-tetrachlorodibenzo-p-dioxin equivalent concentrations in environmental samples. *Environ. Sci. Technol.* 30: 706-715.

Kennedy, S.W., A. Lorenzen, S.P. Jones, M.E. Hahn, and J.J. Stegeman. 1996b. Cytochrome P4501A induction in avian hepatocyte cultures: A promising approach for predicting the sensitivity of avian species to toxic effects of halogenated aromatic hydrocarbons. *Toxicol. Appl. Pharmacol.* 141: 214-230.

Kiff, L.F. 1980. Historical changes in resident populations of California islands raptors. In *The California Islands: Proceedings of a Multidisciplinary Symposium*, D.M. Power, ed. Santa Barbara Natural History Museum, Santa Barbara, CA. pp. 651-673.

King, K.A., T.W. Custer, and J.S. Quinn. 1991. Effects of mercury, selenium, and organochlorine contaminants on reproduction of Forster's terns and black skimmers nesting in a contaminated Texas bay. *Arch. Environ. Contam. Toxicol.* 20: 32-40.

Korach, K.S., P. Sarver, K. Chae, J.A. McLachlan, and J.D. McKinney. 1987. Estrogen receptor-binding activity of polychlorinated hydrodroxybiphenyls: Conformationally restricted structural probes. *Mol. Pharmacol.* 33: 120-126.

Kubiak, T.J. and D.A. Best. 1991. Wildlife Risks Associated with Passage of Contaminated, Anadromous Fish at Federal Energy Regulatory Commission Licensed Dams in Michigan. U.S. Fish and Wildlife Service Report, East Lansing, MI.

Kubiak, T.J., H.J. Harris, L.M. Smith, T.R. Schwartz, D.L. Stalling, J.A. Trick, L. Sileo, D.E. Docherty, and T.C. Erdman. 1989. Microcontaminants and reproductive impairment of the Forster's tern on Green Bay, Lake Michigan. *Arch. Environ. Contam. Toxicol.* 18: 706-727.

Lamparski, L.L., T.J. Nestrick, and S.S. Cutie. 1990. The impact on the environment of airborne particulate matter from the eruption of Mount Saint Helens in May 1980. In *Emissions from Combustion Processes: Origin, Measurement, Control.* R. Clement and R. Kager (eds.). Lewis Publishers, Chelsea, MI.

Langenberg, J. 1990. Preliminary Summary of Pathology Findings for 1989. Report to U.S. Fish and Wildlife Service, Green Bay Field Office.

Langham, N.P.E. 1972. Chick survival in terns (*Sterna spp.*) with particular reference to the common tern. *J. Anim. Ecol.* 41: 385-395.

Larson, J.M., W.H. Karasov, L. Sileo, K.L. Stromborg, B.A. Hanbidge, J.P. Giesy, D.E. Tillitt, and D. Verbrugge. 1996. Reproductive success, developmental anomalies, and environmental contaminants in double-crested cormorants (*Phalacrocorax auritus*). *Environ. Toxicol. Chem.* 15: 553-559.

Lincer, J.L. and D.B. Peakall. 1970. Metabolic effects of polychlorinated biphenyls in the American kestrel. *Nature* 228: 783-784.

Lorenzen, A., J.L. Shutt, and S.W. Kennedy. 1997. Sensitivity of common tern (*Sterna hirundo*) embryo hepatocyte cultures to CYP1A induction and porphyrin accumulation by halogenated aromatic hydrocarbons and common tern egg extracts. *Arch. Environ. Comtam. Toxicol.* 32: 126-134.

Ludwig, J.P. 1965. Biology and structure of the Caspian tern (*Hydroprogne caspia*) population of the Great Lakes from 1896-1964. *Bird Banding* 36: 217-233.

Ludwig, J.P. 1984. Decline, resurgence and population dynamics of Michigan and Great Lakes double-crested cormorants. *Jack Pine Warbler* 62: 91-102.

Ludwig, J.P. and M.E. Ludwig. Undated report a. Food Habits of the Double-Crested Cormorant in Michigan, 1986-1988: Implications for the Acquisition of Toxic Contaminants from Fish by Breeding Females. Michigan Department of Natural Resources.

Ludwig, J.P. and M.P. Ludwig. Undated report b. Productivity and Deformities in Chicks of the Michigan Great Lakes' Colonies of Double-Crested Cormorants (*Phalacrocorax auritus*) and Caspian Terns (*Hydroprogne caspia*). Michigan Department of Natural Resources.

Ludwig, J.P., H.J Auman, D.V. Weseloh, G.A. Fox, J.P. Giesy, and M.E. Ludwig. 1995. Evaluation of the effects of toxic chemicals in Great Lakes cormorants: Has causality been established? *Colon. Waterbirds* 18: 60-69.

Ludwig, J.P., H. Kurita-Matsuba, H.J. Auman, M.E. Ludwig, C.L. Summer, J.P. Giesy, D.E. Tillitt, and P.D. Jones. 1996. Deformities, PCBs and TCDD-equivalents in double-crested cormorants (*Phalacrocorax auritus*) and Caspian terns (*Hydroprogne caspia*) of the upper Great Lakes 1986-1991: Testing a cause-effect hypothesis. *J. Great Lakes Res.* 22: 172-197.

Ludwig, J.P., J.P. Giesy, C.L. Summer, W. Bowerman, S. Heaton, R. Aulerich, S. Bursian, H.J. Auman, P.D. Jones, L.L. Williams, D.E. Tillitt, and M. Gilbertson. 1993. A comparison of water quality criteria in the Great Lakes basin based on human and wildlife health. *J. Great Lakes Res.* 19: 789-807.

Manchester-Neesvig, J.B., A.W. Andren, and D.W. Edgington. 1996. Patterns of mass sedimentation and of deposition of sediment contaminated by PCBs in Green Bay. *J. Great Lakes Res.* 22(2): 444-462.

Matteson, S.W. 1988. Wisconsin Common Tern Recovery Plan. Wisconsin Endangered Resources Report 41. Bureau of Endangered Resources, Wisconsin Department of Natural Resources, Madison.

Matteson, S.W. 1993. Wisconsin Caspian Tern Recovery Plan. Bureau of Endangered Resources, Wisconsin Department of Natural Resources. Madison.

McCarty, J.P. and A.L. Secord. 1999. Nest-building behavior in PCB-contaminated tree swallows. *The Auk* 116: 55-63.

McKinney, J.D., J. Fawkes, S. Jordan, K. Chae, S. Oatley, R.E. Coleman, and W. Briner. 1985. 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) as a potent and persistent thyroxine agonist: A mechanistic model for toxicity based on molecular reactivity. *Environ. Health Perspect*. 61: 41-53.

Miller, S. 1987. Wildlife Management's procedures for issuing an advisory — wildlife consumption advisory. Memorandum from S. Miller (State of Wisconsin Bureau of Wildlife Management) to J.R. Huntoon. April 15.

Mora, M.A., H.J. Auman, J.P. Ludwig, J.P. Giesy, D.A. Verbrugge, and M.E. Ludwig. 1993. Polychlorinated biphenyls and chlorinated insecticides in plasma of Caspian terns: Relationships with age, productivity, and colony site tenacity in the Great Lakes. *Arch. Environ. Contam. Toxicol.* 24: 320-331.

Mossman, M.J. 1988. Wisconsin Forster's Tern Recovery Plan. Bureau of Endangered Resources, Wisconsin Department of Natural Resources. Madison.

Murk, A.J., A.T.C. Bosveld, M. van den Berg, and A. Brouwer. 1994. Effects of polyhalogenated aromatic hydrocarbons (PHAHs) on biochemical parameters in chicks of the common tern (*Sterna hirundo*). *Aquat. Toxicol.* 30: 91-115.

Neuman, J., D.L. Pearl, P.J. Ewins, R. Black, D.V. Weseloh, M. Pike, and K. Karwowski. 1997. Spatial and temporal variation in the diet of double-crested cormorants (*Phalacrocorax auritus*) breeding on the lower Great Lakes in the early 1990s. *Can. J. Fish. Aquat. Sci.* 54: 1569-1584.

Nikolaidis, E., B. Brunstrom, and L. Dencker. 1988. Effects of TCDD congeners 3,3',4,4'-tetrachlorobiphenyl and 3,3',4,4'-tetrachloroazoxybenzene on lymphoid development in the bursa fabricus of the chick embryo. *Toxicol. Appl. Pharmacol.* 92: 315-323.

Nisbet, I.C.T. 1989. Organochlorines, reproductive impairment and declines in bald eagle *Haliaeetus leucocephalus* populations: Mechanisms and dose-response mechanisms. In *Raptors in the Modern World,* B.-U. Meyburg and R.D. Chancellor, eds., World Working Group on Birds of Prey and Owls, Berlin, pp. 483-489.

Nisbet, I.C.T. and R.W. Risebrough. 1994. Relationship of DDE to productivity of bald eagles in California and Arizona, USA. In *Raptor Conservation Today.* B.U. Meyerburg and R.D. Chancellor, eds., World Working Group on Birds of Prey and The Pica Press, East Sussex, Great Britain.

Nisbet, I.C.T., D.M. Fry, J.J. Hatch, and B. Lynn. 1996. Feminization of male common tern embryos is not correlated to specific PCB congeners. *Bull. Environ. Contam. Toxicol.* 57: 895-901.

Nosek, J.A., J.R. Sullivan, S.R. Craven, A. Gendron-Fitzpatrick, and R.E. Peterson. 1993. Embryotoxicity of 2,3,7,8-tetrachloro-p-dioxin in the ring-necked pheasant. *Environ. Toxicol. Chem.* 12: 1215-1222.

Oliver, B.G. and A.J. Niimi. 1988. Trophodynamic analysis of polychlorinated biphenyl congeners and other chlorinated hydrocarbons in the Lake Ontario ecosystem. *Environ. Sci. Technol.* 22: 388-397.

Olsson, A., B. Helander, A. Bignert, K. Litzén, L. Asplund, and Å. Bergman. 1998. Is PCB responsible for embryo toxicity in white-tailed sea eagle (*Haliaeetus albicilla*) from the Swedish Baltic coast? *Organohal. Comp.* 39: 17-20.

Peakall, D.B. and J.L. Lincer. 1996. Do PCBs cause eggshell thinning? *Environ. Pollut.* 91: 127-129.

Peakall, D.B. and M.L. Peakall. 1973. Effects of a polychlorinated biphenyl on the reproduction of artificially and naturally incubated dove eggs. *J. Appl. Ecol.* 10: 863-868.

Peakall, D.B., J.L. Lincer, and S.E. Bloom. 1972. Embryonic mortality and chromosomal alterations caused by Aroclor 1254 in ring doves. *Environ. Health Perspect.* 1: 103-104.

Peden-Adams, M., K. Alonso, C. Godard, S. Skipper, W. Mashburn, J. Hoover, C. Charbonneau, D. Henshel, and R. Dickerson. 1998. Effects of environmentally relevant concentrations of 2,3,7,8-TCDD on domestic chicken immune function and CYP450 activity: F_1 generation and egg injection studies. *Chemosphere* 37: 1923-1939.

Pekarik, C., D.V. Weseloh, G.C. Barrett, M. Simon, C.A. Bishop, and K.E. Pettit. 1998. *An Atlas of Contaminants in Eggs of Fish-Eating Colonial Birds of the Great Lakes (1993-1997). Vol. I. Accounts by Location.* Environment Canada. Canadian Wildlife Service. Technical Report Series No. 321.

Petersen, R.E., H.M. Theobald, and G.L. Kimmel. 1993. Development and reproductive toxicity of dioxins and related compounds: Cross-species comparisons. *Crit. Rev. Toxicol.* 23: 283-335.

Pettit, K.E., C.A. Bishop, D.V. Weseloh, and R.J. Norstrom. 1994. An Atlas of Contaminants in Eggs of Fish-Eating Colonial Birds of the Great Lakes (1989-1992). Environment Canada. Canadian Wildlife Service. Technical Report Series No. 193.

Platonow, N.S. and H.S. Funnell. 1971. Anti-androgenic-like effect of polychlorinated biphenyls in cockerels. *Veterin. Record*. 83: 109-110.

Platonow, N.S. and B.S. Reinhart. 1973. The effects of polychlorinated biphenyls (Aroclor 1254) on chicken egg production, fertility and hatchability. *Can. J. Comp. Med.* 37: 341-346.

Postupalsky, S. 1971. Toxic Chemicals and Declining Bald Eagle and Cormorants in Ontario. Canadian Wildlife Service, Petic. Sec., MS Rep. 20. 70 pp.

Postupalsky, S. 1985. The bald eagles return. *Natural History* 87: 62-63.

Powell, D.C., R.J. Aulerich, J.C. Meadows, D.E. Tillitt, J.P. Giesy, K.L. Stromborg, and S.J. Bursian. 1996. Effects of 3,3',4,4',5-pentachlorobiphenyl (PCB 126) and 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) injected into the yolks of chicken (*Gallus domesticus*) eggs prior to incubation. *Arch. Environ. Contam. Toxicol.* 31: 404-409.

Powell, D.C., R.J. Aulerich, J.C. Meadows, D.E. Tillitt, J.F. Powell, J.C. Restum, K.L. Stromborg, J.P. Giesy, and S.J. Bursian. 1997. Effects of 3,3',4,4',5-pentachlorobiphenyl (PCB 126), 2,3,7,8-tetrachlordibenzo-*p*-dioxin (TCDD), or an extract derived from field-collected cormorant eggs injected into double-crested cormorant (*Phalacrocorax auritus*) eggs. *Environ. Toxicol. Chem.* 16: 1450-1455.

Prestt, I., D.J. Jefferies, and N.W. Moore. 1970. Polychlorinated biphenyls in wild birds in Britain and their avian toxicity. *Environ. Pollut.* 1: 3-26.

Rattner, B.A., M.J. Melancon, T.W. Custer, R.L. Hothem, K.A. King, L.J. LeCaptain, J.W. Spann, B.R. Woodin, and J.L. Stegeman. 1993. Biomonitoring environmental contamination with pipping black-crowned night heron embryos: Induction of cytochrome P450. *Environ. Toxicol. Chem.* 12: 1719-1732.

Rattray, K.J. 1997. The nesting success and PCB burdens of yellow-headed blackbirds found in diked marshes along Green Bay, Lake Michigan. Masters Thesis to University of Wisconsin, Green Bay. December.

Robbins, S.D. 1991. *Wisconsin Birdlife. Population and Distribution Past and Present.* The University of Wisconsin Press, Madison.

Rodman, L.E., S.I. Shedlofsky, A. Manschreck, M. Puttman, A.T. Swim, and L.W. Robertson. 1991. Differential potency of atropisomers of polychlorinated biphenyls on cytochrome P450 induction and uroporphyrin accumulation in the chick embryo hepatocyte culture. *Biochem. Pharmacol.* 41: 915-922.

Ross, P.S. and C.V. Weseloh. 1988. A Study of the Development, Occurrence and Severity of Deformities in Double-Crested Cormorants on the Canadian Great Lakes and Lake Winnipegosis, 1988. Unpublished report of Canadian Wildlife Service.

Rottiers, D.V. and R.M. Tucker. 1982. Proximate composition and caloric content of 8 Lake Michigan fishes. USFWS Technical Paper Number 108.

Rozemeijer, M.J.C., J.P. Boon, C. Swennen, A. Brouwer, and A.J. Murk. 1995. Dioxin type and mixed type induction of the cytochrome P-450 system of common eider ducklings (*Somateria mollissima*) by PCBs: With indications for biotransformation. *Aquat. Toxicol.* 32: 93-113.

Ryckman, D.P., D.V. Weseloh, P. Hamr, G.A. Fox, B. Collins, P.J. Ewins, and R.J. Norstrom. In press. Spatial and temporal trends in organochlorine contamination and bill deformities in double-crested cormorants (*Phalacrocorax auritus*) from the Canadian Great Lakes. *Env. Monitor. Assess.*

Safe, S. 1994. Polychlorinated biphenyls (PCBs): Environmental impact, biochemical and toxic responses, and implications for risk assessment. *Crit. Rev. Toxicol.* 24: 87-149.

Salt, G.W. and D.E. Willard. 1971. The hunting behavior and success of Forster's tern. *Ecology* 52: 989-998.

Schulz, D.E., G. Petrick, and J.C. Duinker. 1989. Complete characterization of polychlorinated biphenyl congeners in commercial Aroclors and Clophen mixtures by multidimensional gas chromatography-electron capture detection. *Environ. Sci. Technol.* 23: 852-859.

Scott, M.L. 1977. Effects of PCBs, DDT, and mercury compounds in chickens and Japanese quail. *Feder. Proceed.* 36: 1888-1893.

Sheffield, S.R., J.M. Matter, B.A. Rattner, and P.D. Guiney. 1998. Fish and wildlife species as sentinels of environmental endocrine disruptors. Chapter 17 in *Principles and Processes for Evaluating Endocrine Disruption in Wildlife,* R.J. Kendall (ed). SETAC Press, Pensacola, FL, pp. 369-430.

Smith, L.M., T.R. Schwartz, and K. Feltz. 1990. Determination and occurrence of AHH-active polychlorinated biphenyls, 2,3,7,8-tetrachloro-p-dioxin and 2,3,7,8-tetrachlorodibenzofuran in Lake Michigan sediment and biota. The question of their relative toxicological significance. *Chemosphere* 21(9): 1063-1085.

Spear, P.A., D.H. Bourbonnais, D.B. Peakall, and T.W. Moon. 1989. Dove reproduction and retinoid (vitamin A) dynamics in adult females and their eggs following exposure to 3,3',4,4'-tetrachlorobiphenyl. *Can. J. Zool.* 67: 908-913.

Sprunt, A., IV. 1963. Bald eagles aren't producing enough young. *Audubon Mag.* 65: 32-35.

Sprunt, A., IV and F.J. Ligas. 1966. Audubon Bald Eagle Studies — 1960-1965. In *Proceedings of the 62nd Annual Convention of the National Audubon Society,* Sacramento, CA.

Sprunt, A., IV, W.B. Robertson, Jr., S. Postupalsky, R.J. Hensel, C.E. Knoder, and F.J. Ligas. 1973. Comparative productivity of six bald eagle populations. In *Transactions of the 38th North American Wildlife & Natural Resources Conference,* pp. 98-106.

Stickel, W.H., L.F. Stickel, R.A. Dyrland, and D.L. Hughes. 1984. Aroclor 1254® residues in birds: Lethal levels and loss rates. *Arch. Environ. Contam. Toxicol.* 13: 7-13.

Stickel, L.F., N.J. Chura, P.A. Stewart, C.M. Menzie, R.M. Prouty, and W.L. Reichel. 1966. Bald eagle-pesticide relations. In *Transactions of the 31st North American Wildlife & Natural Resources Conference,* pp. 190-200.

Struger, J. and D.V. Weseloh. 1985. Great Lakes Caspian terns: Egg contaminants and biological implications. *Colon. Waterbirds* 8: 142-148.

Temple, S.A. and J.R. Cary. 1987. *Wisconsin Birds. A Seasonal and Geographical Guide.* The University of Wisconsin Press, Madison.

Thiel, D.A., S.G. Martin, J.W. Duncan, M.J. Lemke, W.R. Lance, and R.E. Peterson. 1988. Evaluation of the effects of dioxin-contaminated sludges on wild birds. In *Proceedings, 1988 TAPPI Environmental Conference,* Norcross, GA, pp. 487-506.

Tillitt, D.E., T.J. Kubiak, G.T. Ankley, and J.P. Giesy. 1993. Dioxin-like toxic potency in Forster's tern eggs from Green Bay, Lake Michigan, North America. *Chemosphere* 26(11): 2079-2084.

Tillitt, D.E., G.T. Ankley, J.P. Giesy, J.P. Ludwig, H. Kurita-Matsuba, D.V. Weseloh, P.S. Ross, C. Bishop, L. Sileo, K.L. Stromborg, J. Larson, and T. Kubiak. 1992. Polychlorinated biphenyl residues and egg mortality in double-crested cormorants from the Great Lakes. *Environ. Toxicol. Chem.* 11: 1281-1288.

Tilson, H.A., J.A. Jacobs, and W.J. Rogan. 1990. Polychlorinated biphenyls and the developing nervous system: Cross-species comparisons. *Neurotoxicol. Teratol.* 12: 239-248.

Tori, G.M. and T.J. Peterle. 1983. Effects of PCBs on mourning dove courtship behavior. *Bull. Environ. Contam. Toxicol.* 30: 44-49.

Trick, J.A. 1982. Reproductive success and foraging success as related to populations of Forster's terns (*Sterna forsteri*) on Green Bay, Wisconsin. MS Thesis, University of Wisconsin, Green Bay.

Tumasonis, C.F., B. Bush, and F.D. Baker. 1973. PCB levels in egg yolk associated with embryonic mortality and deformity of hatched chicks. *Arch. Environ. Contam. Toxicol.* 1: 312-324.

USDI. 1974. United States estimated to have 1,000 pairs of bald eagles in lower 48 states. U.S. Department of Interior, U.S. Fish and Wildlife Service, News Release, 27 February.

U.S. EPA. 1980. Ambient Water Quality Criteria for Polychlorinated Biphenyls. EPA 440/5-80-068. U.S. Environmental Protection Agency, Office of Water, Washington, DC.

U.S. EPA. 1998a. Guidelines for Ecological Risk Assessment. Final. U.S. Environmental Protection Agency, Washington, DC.

U.S. EPA. 1998b. Workshop on the Application of 2,3,7,8-TCDD Toxicity Equivalency Factors to Fish and Wildlife. U.S. Environmental Protection Agency. Chicago, IL. January 20-22.

USFWS. 1993. Reconnaissance Surveys of Contaminants Potentially Affecting Green Bay and Gravel Island National Wildlife Refuges. U.S. Fish and Wildlife Service, Ecological Services Field Office, Green Bay, WI.

USGS. 1990. USGeoData 1:250,000 and 1:100,000 Scale Land Use and Land Cover and Associated Maps Digital Data. U.S. Geological Survey, Reston, VA.

Van den Berg, M., B.L.H.J. Craane, T. Sinnige, S. van Mourik, S. Dirksen, T. Boudewijn, M. van der Gaag, I. J. Lutke-Schipholt, B. Spenkelink, and A. Brouwer. 1994. Biochemical and toxic effects of polychlorinated biphenyls (PCBs), dibenzo-p-dioxins (PCDDs) and dibenzofurans (PCDFs) in the cormorant (*Phalacrocorax carbo*) after in ovo exposure. *Environ. Toxicol. Chem.* 13: 803-816.

Van den Berg, M., L. Birnbaum, A.T.C. Bosveld, B. Brunstrom, P. Cook, M. Feely, J.P. Giesy, A. Hanberg, R. Hasegawa, S.W. Kennedy, T. Kubiak, J.C. Larsen, F.X. Rolaf van Leeuwen, A.K. Djien Liem, C. Nolt, R.E. Peterson, L. Poellinger, S. Safe, D. Schrenk, D. Tillitt, M. Tysklind, M. Younes, F. Waern, and T. Zacharewski. 1998. Toxic equivalency factors (TEFs) for PCBs, PCDDs, and PCDFs for humans and wildlife. *Environ. Health Perspect.* 106: 775-792.

Walker, C.H. 1995. Biochemical biomarkers in ecotoxicology-some recent developments. *Sci. Total Environ.* 171: 189-195.

Weseloh, D.V. and P.J. Ewins. 1994. Characteristics of a rapidly increasing colony of double-crested cormorants (*Phalacrocorax auritus*) in Lake Ontario: Population size, reproductive parameters and band recoveries. *J. Great Lakes Res.* 20(2): 443-456.

Weseloh, D.V., P.J. Ewins, and K.A. Pettit. 1995a. Continued growth of the double-crested cormorant populations on the Great Lakes. Abstract in *Bull. Colon. Waterbird Soc.* 19: 2.

Weseloh, D.V., P.J. Ewins, J. Struger, P. Mineau, C.A. Bishop, S. Postupalsky, and J.P. Ludwig. 1995b. Double-crested cormorants of the Great Lakes: Changes in population size, breeding distribution and reproductive output between 1913 and 1991. *Colon. Waterbirds* 18: 48-59.

White, D.H. and E. Cromartie. 1977. Residues of environmental pollutants and shell thinning in merganser eggs. *Wilson Bull.* 89: 532-542.

White, D.H. and D.J. Hoffman. 1995. Effects of polychlorinated dibenzo-*p*-dioxins and dibenzofurans on nesting wood ducks *(Aix sponsa)* at Bayou Meto, Arkansas. *Environ. Health Perspect.* 103(Supplement 4): 37-39.

Wiemeyer, S.N., C.M. Bunck, and C.J. Stafford. 1993. Environmental contaminants in bald eagle eggs — 1980-84 — and further interpretations of relationships to productivity and shell thickness. *Arch. Environ. Contam. Toxicol.* 24: 213-227.

Wiemeyer, S.N., T.G. Lamont, C.M. Bunck, C.R. Sindelar, F.J. Gramlich, J.D. Fraser, and M.A. Byrd. 1984. Organochlorine pesticide, polychlorobiphenyl, and mercury residues in bald eagle eggs — 1969 and 1979 — and their relationships to shell thinning and reproduction. *Arch. Environ. Contam. Toxicol.* 13: 529-549.

Wiemeyer, S.N., B.M. Mulhern, F.J. Ligas, R.J. Hensel, J.E. Mathisen, F.C. Robards, and S. Postupalsky. 1972. Residues of organochlorine pesticides, polychlorinated biphenyls, and mercury in bald eagle eggs and changes in shell thickness — 1969 and 1970. *Pest. Monitor. J.* 6(1): 50-55.

Williams, L.L., J.P. Giesy, D. Verbrugge, S. Jurzystra, and K.L. Stromborg. 1995a. Polychlorinated biphenyls and 2,3,7,8-tetrachlorodibenzo-*p*-dioxin equivalents in eggs of double-crested cormorants from a colony near Green Bay, Wisconsin, USA. *Arch. Environ. Contam. Toxicol.* 29: 327-333.

Williams, L.L., J.P. Giesy, D. Verbrugge, S. Jurzystra, H. Heinz, and K.L. Stromborg. 1995b. Polychlorinated biphenyls and 2,3,7,8-tetrachlorodibenzo-*p*-dioxin equivalents in eggs of red-breasted mergansers near Green Bay, Wisconsin, USA, in 1977-78. *Arch. Environ. Contam. Toxicol.* 29: 52-60.

Wilson, C.L. and S. Safe. 1998. Mechanisms of ligand-induced aryl hydrocarbon receptor-mediated biochemical and toxic responses. *Toxicol. Pathol.* 26(5): 657-671.

Wisconsin DNR. 1971-1995. State of Wisconsin fish contaminants monitoring database. Provided to Stratus Consulting Inc. in electronic format by J. Amrhein, Wisconsin Department of Natural Resources.

Wisconsin DNR. 1987. Press release detailing imposition of waterfowl consumption advisory. Wisconsin Department of Natural Resources, Madison. June 25.

Wisconsin DNR. 1998a. Fox River and Green Bay PCB Fate and Transport Model Evaluation: Compilation and Estimation of Historical Discharges of Total Suspended Solids and PCB from Fox River Point Sources. Wisconsin Department of Natural Resources. Draft.

Wisconsin DNR. 1998b. WISCLAND Land Cover (WLCGW930). Wisconsin Department of Natural Resources, Madison.

Woodford, J.E., Karasov, W.H., Meyer, M.W., and Chambers, L. 1998. Impact of 2,3,7,8-TCDD exposure on survival, growth, and behavior of ospreys breeding in Wisconsin, USA. *Environ. Toxicol. Chem.* 17: 1323-1331.

Yamashita, N., S. Tanabe, J.P. Ludwig, H. Kurita, M.E. Ludwig, and R. Tatsukawa. 1993. Embryonic abnormalities and organochlorine contamination in double-crested cormorants (*Phalacrocorax auritus*) and Caspian terns (*Hydroprogne caspia*) from the upper Great Lakes in 1988. *Environ. Pollut.* 79: 163-173.

Zubay, G. 1983. *Biochemistry.* Addison-Wesley Publishing Company, Reading, MA. 1268 pp.

APPENDIX A
SCIENTIFIC NAMES OF BIRD SPECIES MENTIONED IN TEXT

English Name	Scientific Name
Alder flycatcher	*Empidonax virescens*
American bittern	*Botaurus lentiginosus*
American black duck	*Anas rubripes*
American coot	*Fulica americana*
American crow	*Corvus brachyrhynchos*
American goldfinch	*Carduelis tristis*
American kestrel	*Falco sparverius*
American redstart	*Setophaga ruticilla*
American robin	*Turdus migratorius*
American tree sparrow	*Spizella arborea*
American wigeon	*Anas americana*
American woodcock	*Scolopax minor*
Baird's sandpiper	*Calidris bairdii*
Bald eagle	*Haliaeetus leucocephalus*
Bank swallow	*Riparia riparia*
Barn swallow	*Hirundo rustica*
Barred owl	*Strix varia*
Bay-breasted warbler	*Dendroica castanea*
Belted kingfisher	*Ceryle alcion*
Bell's vireo	*Vireo bellii*
Black tern	*Chlidonias niger*
Black-and-white warbler	*Mniotilta varia*
Black-bellied plover	*Pluvialis squatorola*
Black-billed cuckoo	*Coccyzus erythropthalmus*
Black-capped chickadee	*Parus atricapillus*
Black-crowned night heron	*Nycticorax nycticorax*
Black-headed gull	*Larus ridibundus*
Black-throated blue warbler	*Dendroica caerulescens*
Black-throated green warbler	*Dendroica virens*
Blackburnian warbler	*Dendroica fusca*
Blackpoll warbler	*Dendroica striata*
Blue jay	*Cyanocitta cristata*
Blue-gray gnatcatcher	*Polioptila caerulea*
Blue-winged teal	*Anas discors*
Blue-winged warbler	*Vermivora pinus*

Bobwhite	*Colinus virginianus*
Bohemian waxwing	*Bombycilla garrulus*
Bonaparte's gull	*Larus philadelphia*
Boreal chickadee	*Parus hudsonicus*
Brewer's blackbird	*Euphagus cyanocephalus*
Broad-winged hawk	*Buteo platypterus*
Brown creeper	*Certhia americana*
Brown thrasher	*Toxostoma rufum*
Brown-headed cowbird	*Molothrus ater*
Bufflehead	*Bucephala albeola*
Canada goose	*Branta canadensis*
Canada warbler	*Wilsonia canadensis*
Canvasback	*Aythya valisineria*
Cape May warbler	*Dendroica tigrina*
Caspian tern	*Hydroprogne caspia*
Cattle egret	*Bubulcus ibis*
Cedar waxwing	*Bombycilla cedrorum*
Cerulean warbler	*Dendroica cerulea*
Chestnut-sided warbler	*Dendroica pensylvanica*
Chicken	*Gallus gallus*
Chimney swift	*Chaetura pelagica*
Chipping sparrow	*Spizella passerina*
Clay-colored sparrow	*Spizella pallida*
Cliff swallow	*Hirundo pyrrhonota*
Common eider	*Somateria mollissima*
Common goldeneye	*Bucephala clangula*
Common grackle	*Quiscalus quiscula*
Common loon	*Gavia immer*
Common merganser	*Mergus merganser*
Common moorhen	*Gallinula chloropus*
Common murre	*Uria aalge*
Common nighthawk	*Chordeiles minor*
Common raven	*Corvus corax*
Common redpoll	*Carduelis flammea*
Common snipe	*Gallinago gallinago*
Common tern	*Sterna hirundo*
Common yellowthroat	*Geothlypis trichas*
Connecticut warbler	*Oporornis agilis*
Cooper's hawk	*Accipter cooperii*
Dark-eyed junco	*Junco hyemalis*
Dickcissel	*Spiza americana*
Domestic goose	*Anser anser*

Double-crested cormorant	*Phalacrocorax auritus*
Downy woodpecker	*Picoides pubescens*
Dunlin	*Calidris alpina*
Eastern bluebird	*Sialia sialia*
Eastern kingbird	*Tyrannus tyrannus*
Eastern meadowlark	*Sturnella magna*
Eastern phoebe	*Sayornis phoebe*
Eastern screech owl	*Otus asio*
Eastern wood-pewee	*Contopus virens*
European shag	*Phalacrocorax aristotelis*
Evening grosbeak	*Coccothraustes vespertinus*
Field Sparrow	*Spizella pusilla*
Forster's tern	*Sterna forsteri*
Fox sparrow	*Passerella iliaca*
Franklin's gull	*Larus pipixcan*
Gadwall	*Anas strepera*
Glaucus gull	*Larus hyperboreus*
Golden-winged warbler	*Vermivora chrysoptera*
Grasshopper sparrow	*Amodramus savannarum*
Gray catbird	*Dumetella carolinensis*
Gray jay	*Perisoreus canadensis*
Gray partridge	*Perdix perdix*
Gray-cheeked thrush	*Catharus minimus*
Great blue heron	*Ardea herodias*
Great crested flycatcher	*Myarchus crinitus*
Great egret	*Casmerodius albus*
Great horned owl	*Bubo virginianus*
Greater scaup	*Aythya marila*
Greater yellowlegs	*Tringa melanoleuca*
Green-backed heron	*Butorides virescens*
Green-winged teal	*Anas crecca*
Hairy woodpecker	*Picoides villosus*
Harris's sparrow	*Zonotrichia querula*
Hermit thrush	*Catharus guttatus*
Herring gull	*Larus argentatus*
Hooded merganser	*Lophodytes cucullatus*
Hooded warbler	*Wilsonia citrina*
Horned grebe	*Podiceps auritus*
Horned lark	*Eremophila alpestris*
House sparrow	*Passer domesticus*
House wren	*Troglodytes aedon*
Hudsonian godwit	*Limosa haemastica*

Indigo bunting	*Passerina cyanea*
Japanese quail	*Coturnix japonica*
Killdeer	*Charadrius vociferus*
Lapland longspur	*Calcarius lapponicus*
Lark sparrow	*Chondestes grammacus*
Le Conte's sparrow	*Ammodramus leconteii*
Least bittern	*Ixobrychus exilis*
Least Flycatcher	*Empidonax minimus*
Least sandpiper	*Calidris minutilla*
Lesser scaup	*Aythya affinis*
Lesser yellowlegs	*Tringa flavipes*
Lincoln's sparrow	*Melospiza lincolnii*
Little gull	*Larus minutilla*
Loggerhead shrike	*Lanius ludovicianus*
Long-billed dowitcher	*Limnodromus scolopaceus*
Long-eared owl	*Asio otus*
Louisiana waterthrush	*Seiurus motacilla*
Magnolia warbler	*Dendroica magnolia*
Mallard	*Anas platyrhynchos*
Marbled godwit	*Limosa fedoa*
Marsh wren	*Telmatodytes palustris*
Merlin	*Falco columbarius*
Mourning dove	*Zenaidura macroura*
Mourning warbler	*Oporornis philadelphia*
Mute swan	*Cygnus olor*
Nashville warbler	*Vermivora ruficapilla*
Northern cardinal	*Cardinalis cardinalis*
Northern flicker	*Colaptes auratus*
Northern goshawk	*Accipiter gentilis*
Northern harrier	*Circus cyaneus*
Northern mockingbird	*Mimus polyglottos*
Northern oriole	*Icterus galbula*
Northern parula	*Parula americana*
Northern pintail	*Anas acuta*
Northern rough-winged swallow	*Stelgidopteryx serripennis*
Northern saw-whet owl	*Aegolius acadicus*
Northern shoveller	*Anas clypeata*
Northern shrike	*Lanius excubitor*
Northern waterthrush	*Seiurus noveboracensis*
Oldsquaw	*Clangula hyemalis*
Olive-sided flycatcher	*Contopuis borealis*
Orange-crowned warbler	*Vermivora celata*

Orchard oriole	*Icterus spurius*
Osprey	*Pandion haliaetus*
Ovenbird	*Seiurus aurocapillus*
Palm warbler	*Dendroica palmarum*
Pectoral sandpiper	*Calidris melanotus*
Peregrine falcon	*Falco peregrinus*
Philadelphia vireo	*Vireo philadelphicus*
Pied-billed grebe	*Podilymbus podiceps*
Pileated woodpecker	*Dryocopus pilleatus*
Pine grosbeak	*Pinicola enucleator*
Pine siskin	*Carduelis pinus*
Pine warbler	*Dendroica pinus*
Prothonotary warbler	*Protonotaria citrea*
Purple finch	*Carpodacus purpureus*
Purple martin	*Progne subis*
Red crossbill	*Loxia curvirostra*
Red-bellied woodpecker	*Melanerpes carolinus*
Red-breasted merganser	*Mergus serrator*
Red-breasted nuthatch	*Sitta canadensis*
Red-eyed vireo	*Vireo olivaceus*
Red-headed woodpecker	*Melanerpes erythrocephalus*
Red-necked grebe	*Podiceps grisegena*
Red-necked phalarope	*Phalaropus lobatus*
Red-shouldered hawk	*Buteo lineatus*
Red-tailed hawk	*Buteo jamaicensis*
Red-throated loon	*Gavia stellata*
Red-winged blackbird	*Agelaius phoenicius*
Redhead	*Aythya americana*
Ring-billed gull	*Larus delawarensis*
Ring-necked dove	*Streptopelia risoria*
Ring-necked duck	*Aythya collaris*
Ring-necked pheasant	*Phasianus colchicus*
Rock dove	*Columba livia*
Rose-breasted grosbeak	*Pheucticus ludovicianus*
Rough-legged hawk	*Buteo lagopus*
Ruby-throated hummingbird	*Archilocus colubris*
Ruddy duck	*Oxyura jamaicensis*
Ruddy turnstone	*Arenaria interpres*
Ruffed grouse	*Bonasa umbellus*
Rufous-sided towhee	*Pipilo erythrophthalmus*
Rusty blackbird	*Euphagus carolinus*
Sanderling	*Calidris alba*

Sandhill crane	*Grus canadensis*
Savannah sparrow	*Passerculus sandwichensis*
Scarlet tanager	*Piranga olivacea*
Sedge wren	*Cistothorus platensis*
Semipalmated sandpiper	*Calidris pusilla*
Sharp-shinned hawk	*Accipter striatus*
Sharp-tailed grouse	*Tympanuchus phasianellus*
Short-billed dowitcher	*Limnodromus griseus*
Short-eared owl	*Asio flammeus*
Snow bunting	*Plectrophenax nivalis*
Snow goose	*Anser caerulescens*
Snowy owl	*Nyctea scandiaca*
Solitary sandpiper	*Tringa solitaria*
Solitary vireo	*Vireo solitarius*
Song sparrow	*Melospiza melodia*
Sora	*Porzana carolina*
Spotted sandpiper	*Actitus macularia*
Starling	*Sturnus vulgaris*
Stilt sandpiper	*Calidris himantopus*
Swainson's thrush	*Catharus ustulatus*
Swamp sparrow	*Melospiza georgiana*
Tennessee warbler	*Vermivora peregrina*
Tree swallow	*Iridoprocne bicolor*
Tufted titmouse	*Parus bicolor*
Tundra swan	*Cygnus columbianus*
Turkey	*Meleagris gallopavo*
Turkey vulture	*Cathartes aura*
Upland sandpiper	*Bartramia longicauda*
Veery	*Catharus fuscescens*
Vesper sparrow	*Phooecetes gramineus*
Virginia rail	*Rallus limicola*
Warbling vireo	*Vireo gilvus*
Water pipit	*Anthus spinoletta*
Western meadowlark	*Sturnella neglecta*
White pelican	*Pelecanus erythrorhynchos*
White-crowned sparrow	*Zonotrichia leucophrys*
Whip-poor-will	*Caprimulgus vociferus*
White-breasted nuthatch	*Sitta carolinensis*
White-rumped sandpiper	*Calidris fuscicollis*
White-tailed eagle	*Haliaeetus albicilla*
White-throated sparrow	*Zonotrichia albicollis*
White-winged crossbill	*Loxia leucoptera*

White-winged scoter	*Melanitta deglandi*
Willet	*Catoptrophorus semipalmatus*
Willow flycatcher	*Empidonax traillii*
Wilson's phalarope	*Phalaropus tricolor*
Wilson's warbler	*Wilsonia pusilla*
Winter wren	*Troglodytes troglodytes*
Wood duck	*Aix sponsa*
Wood thrush	*Hylocichla mustelina*
Yellow warbler	*Dendroica petechia*
Yellow-bellied flycatcher	*Empidonax flaviventris*
Yellow-bellied sapsucker	*Sphyrapicus varius*
Yellow-billed cuckoo	*Coccyzus americanus*
Yellow-breasted chat	*Icteria virens*
Yellow-headed blackbird	*Xanthocephalus xanthocephalus*
Yellow-rumped warbler	*Dendroica coronata*
Yellow-throated vireo	*Vireo flavifrons*

APPENDIX B
1996 TERN EGG COLLECTION AND CHEMICAL ANALYSIS PROCEDURES

Standard Operating Procedure for Collection, Transport, and Storage of Tern Eggs

Results of Chemical Analysis of Tern Eggs

Chemical Analysis Technical Procedures

Data Validation Report

STANDARD OPERATING PROCEDURE FOR THE COLLECTION, TRANSPORT, AND STORAGE OF TERN EGGS FROM GREEN BAY, WISCONSIN

1. INTRODUCTION AND STUDY OBJECTIVES

This Standard Operating Procedure (SOP) contains the objectives, methods, and approaches for the collection, transport, and storage of Common tern (*Sterna hirundo*), Forster's tern (*Sterna forsteri*), and Caspian tern (*Sterna caspia*) eggs to be collected from Green Bay, Wisconsin, for the Fox River and Green Bay Natural Resource Damage Assessment (NRDA). The collected eggs will be analyzed for contaminants by an analytical laboratory. A subsequent SOP will describe the laboratory analytical methods that will be employed.

The objective of the study is to:

- Collect eggs of the tern species listed above from colonies in the Lower Fox River and Green Bay to provide comparisons between current and historical egg contaminant concentrations.

Tern eggs will be collected during the 1996 nesting season (and, if necessary, during the 1997 nesting season) and will be analyzed for PCBs (congener-specific analyses), and potentially other contaminants. The field team leader for the egg collection will be Dr. Hector Galbraith.

2. FIELD PROCEDURES

2.1 TERN COLONY LOCATION

Suitable tern nesting colonies will be located by U.S. Fish and Wildlife Service (USFWS) personnel during the early part of the 1996 (and, if necessary, 1997) nesting season. Caspian tern eggs will be collected from the known breeding colony on Gravelly Island, Green Bay. For Foster's and common terns, the egg collections will be made from Kidney Island in the Lower Fox River. If no terns of either species nest on Kidney Island, or the numbers of nesting birds are too low to provide the required sample sizes (see below), the west shore of Green Bay will be searched for nesting colonies, and eggs will be collected from those colonies closest to the mouth of the Lower Fox River.

2.2 EGG COLLECTION

Eggs from at least 6 nests will be collected for each species. If the colony contains more than 6 nests, each nest will be located and uniquely numbered. A random number generator will then be used to identify 6 nests. Up to 2 eggs (depending on the clutch size) will be collected from each of the selected nests. If no colony of 6 or more nests is found, a number of colonies will be combined into a hypothetical colony, the nests numbered, and study nests randomly chosen.

Each collected egg will be given a unique numerical identifier in the field. This number will be written on the egg in pencil. All identification numbers will be recorded in the field logbook. The identification system for eggs samples collected for contaminant analyses consists of the following code:

TE-XX-Y-AB

where:

- **TE** is a two-letter code designating the tern egg collection effort.
- **XX** is a unique two-letter code designating the colony location
- **Y** is a tern species identifier (A = common, B = Forster's, C = Caspian)
- **##** is a unique two-number code designating the nest number. Nests will be numbered starting at "01."

2.3 FIELD DOCUMENTATION

The field team will document its sampling activities and field measurements in a dedicated, paginated, bound field logbook. Sampling locations will be clearly identified on photocopies of appropriate topographical maps and described in the field notebook. Entries in the field notebook and map marking will be done with waterproof ink, and corrections will be made with a single line through the error accompanied by the correction date and corrector's initials. The field team leader will be responsible for maintenance and proper archiving of these field notebooks.

The following information will be recorded in the field logbooks:

- site and project name
- each sampler's name and professional affiliation
- approximate numbers of nests in each colony
- clutch size in each selected nest
- date and time of egg collection, field activity, or field measurement
- identification numbers of samples collected
- number and type of samples collected

- any difficulties encountered or necessary deviations from this SOP
- any other pertinent field observations.

Maps will be marked with a sampling location code, e.g., PE for Peshtigo River, written within a circle. The field notebook page number corresponding to each sampling location will be marked adjacent to the sampling location circle. Photographs will also be taken of each colony.

Upon completion of each day's field activities, the notes will be reviewed by the field recorder and sampler and any necessary corrections made. The field recorder will sign and date each page.

2.4 PROCESSING AND STORAGE OF EGGS

The field team leader or a designated representative will transport the eggs to the USFWS laboratory in Green Bay. Immediately on returning from the field to the laboratory, the eggs will be measured and their contents transferred to chemically clean glass jars. Egg measurements will be made using a Vernier caliper and an electronic balance and will include:

- length and breadth (to the closest 0.1 mm).
- weight (to the closest 0.1g).
- egg volume using water displacement in a gravimetric flask

These measurements will be recorded in the field notebook.

After the above measurements are taken, the contents of each egg will be transferred to a pre-labeled, tared, precleaned and certified glass container and the jar plus egg contents weighed to the closest 0.1g. The jar tare weights and the jar plus contents weights will be recorded in the field log book. The jars will be stored in a freezer to await shipment to the analytical laboratory.

The tern egg shells will be labeled with the egg identifier, allowed to air dry, then stored in a sealed egg box in a dry area within the USFWS field office at Green Bay.

2.5 CHAIN OF CUSTODY

The chain of custody will start when eggs are collected from the nests. Each egg will be given a unique numerical identifier in the field. This number will be written on the egg in pencil. Once identified in this way, the eggs collected during each sampling event will be placed in a communal egg container under the custody of Dr. Hector Galbraith or a designated stand-in. On returning to the laboratory, the contents of each egg will be transferred to separate chemically clean glass jars. Each of these jars will be labeled with the appropriate sample identifier. The jars will be stored frozen in one or more shipping containers which will be sealed with custody seals (to detect

unauthorized tampering with samples after sample collection until the time of use or analysis), and contain chain of custody forms with the following information, as appropriate:

- project name
- egg identifiers (unique for each sample)
- name and signature of field recorder
- date and time of beginning of sample collection
- chain of custody seal number
- signatures of persons involved in the chain of possession
- inclusive dates and times of possession
- method and date of sample shipment.

At the appropriate time, the entire sealed container(s) will be shipped to the analytical laboratory.

The field recorder is personally responsible for the care and custody of the samples until they are transferred or properly dispatched. A sample is in the custody of an individual if any of the following occur:

- The sample is in the individual's possession.
- The sample is within view after being in possession.
- The sample is in a locked or sealed container that prevents tampering after being in possession.
- The sample is in a designated secure area.

Every transfer of custody will be noted with the date and time of transfer and signed for on the chain of custody record. The number of custody transfers will be kept to a minimum.

2.7 FIELD EQUIPMENT

The following list of equipment will be required in the field:

- SOPs (one copy for each team member)
- waders/hip boots (all crew members)
- field log books
- marking pens and pencils
- labels and labeling tape
- chain of custody forms and seals
- an egg box for sample storage and transport
- kimwipes
- camera

2.8 DEVIATIONS FROM THIS SOP

If field conditions necessitate any deviations from this SOP the Field Team Leader will document them in the field note book and in an addendum to this SOP.

ADDENDUM TO TERN EGG COLLECTION SOP

During the course of fieldwork four changes were made to the egg collection method. The decision to make these changes was made by the field team leader. The changes are:

1) Sampling Methods in Forster's Tern Colony. The method described in Section 2.2 of this SOP was changed. The method described in Section 2.2 was developed under the assumption that any Forster's tern colonies found would have relatively few widely dispersed pairs. In fact, the Kidney Island Forster's tern colony comprised about 100 pairs densely settled in a relatively small area. Also, the Kidney Island terns nested immediately adjacent to several hundred pairs of ring-billed gulls (*Larus delawarensis*). Any attempt to number each of the nests and to randomly select study nests, as described in Section 2.2, would have resulted in prolonged disturbance to the birds, with the risk of predation of unguarded eggs by gulls. For these reasons, the following method was adopted:

- the Forster's tern colony was delineated and the numbers of nests counted. Two colonies were found: the main colony comprised 65 nests distributed in an ovoid approximately 20 meters by 60 meters. Another 20 to 30 pairs of terns were nesting in a smaller colony 50 meters to the east of the main colony.

- The main colony was walked through from south to north (along the colonies 60 meter axis) and a single egg was collected from each 6th nest. This provided a sample of 10 eggs.

2) One Egg Was Collected From Each Nest. The collection permit provided by the State of Wisconsin Department of Natural Resources allowed the collection of 10 Forster's and common tern eggs only. The field team leader decided that, in the interests of characterizing the colonies most fully, 10 nest should be sampled. This entailed the collection of one egg from each nest, not the maximum of 2 described in section 2.2.

3) Common Tern Egg Collection. At the time of the collection of the Forster's tern eggs, no common terns were nesting on Kidney Island. However, a visit two weeks later revealed that common terns had by then established themselves. A total of 15 nests were found. One egg was collected from each of 10 randomly chosen nests.

4) No Caspian Tern Eggs Were Collected. No attempt was made to locate and collect the eggs of Caspian terns.

Results of chemical analysis of tern eggs

The column "field.id" represents the identification number of each egg collected. 1996 is the year of collection. "KI" denotes the collection site of Kidney Island, and the last two values (common tern) or three values (Forster's tern) are the sample number (i.e., B01, B02).

The "analyte" column identifies the PCB congener for which a value is given (i.e., c.1.ppb.wwt means PCB congener 1, measured in parts per billion wet weight).

All values of 0 denote values below the detection limit.

Batch Matrix: Tern Eggs
Region: Kidney Island, Green Bay
Reporting Unit: ng/g, wet weight

Client Reporting Sample ID	96KICT01_rep.1		96KICT03_rep.1		96KICT03_rep.2		96KICT05_rep.1		96KICT07_rep.1		96KICT09_rep.1		96KICT10_rep.1		96KICT10_rep.2		TEKIB06_rep.1	
Sample Type:	Common Tern		Common Tern		Common Tern		Common Tern		Common Tern		Common Tern		Common Tern		Common Tern		Forster's Tern	
Sample Lipid Content (%):	5.4		7.7		7.7		9.4		7.5		6.3		7.7		7.7		7.8	
Sample Moisture Content (%):	73.8		79.1		79.1		78.5		72.5		74.7		70.4		70.4		78.4	
Congener Numbers																		
c.001	0.0	U	0.0	U			0.0	U	0.0	U	0.0	U	0.0	U			0.0	U
c.003	0.0	U	0.0	U			0.0	U	0.0	U	0.0	U	0.0	U			0.0	U
c.004+010	0.0	U	0.0	U			0.0	U	0.0	U	0.0	U	0.0	U			0.0	U
c.006	0.8	U	1.1	U			1.2	U	0.0	U	0.0	U	0.0	U			5.2	U
c.007.009	1.0	J	0.3	J			0.5	J	0.5	J	0.0	U	0.0	U			3.6	J
c.008.005	0.7	J	0.7	J			0.0	U	0.0	U	0.0	U	0.0	U			4.4	J
c.012.013	0.0	U	60.8	.			0.0	U	0.0	U	0.0	U	0.0	U			23.6	.
c.016.032	4.1	J	1.7	J			1.7	J	3.2	J	3.1	J	1.9	J			8.4	.
c.017.015	2.3	J	1.0	J			0.5	J	0.0	U	0.8	J	0.0	U			4.8	J
c.018	1.5	J	0.0	UJ			0.0	UJ	0.0	UJ	1.1	J	0.0	UJ			5.5	J
c.019	0.0	U	0.0	U			0.0	U	0.0	U	0.0	U	0.0	U			0.0	U
c.021	0.0	U	0.2	J			0.0	U	0.0	U	0.0	U	0.0	U			2.1	J
c.022	10.2	.	5.5	J			3.7	J	3.1	J	11.0	.	6.1	J			11.1	.
c.024+027	0.3	U	0.0	U			0.2	U	0.0	U	0.6	J	0.0	U			2.4	J
c.025	2.4	J	1.7	J			1.1	J	1.5	J	3.2	J	1.9	J			3.7	J
c.026	1.8	J	1.5	J			0.8	J	1.1	J	4.6	J	1.7	J			3.6	J
c.028	167.8	.	181.1	.			138.5	.	59.8	.	134.6	.	165.8	.			164.2	.
c.029	1.5	J	0.8	J			0.9	J	0.0	U	0.4	J	0.0	U			4.3	J
c.031	83.4	.	56.3	.			43.1	.	20.3	.	57.1	.	51.1	.			62.8	.
c.033.020	6.0	J	3.0	J			1.2	J	0.0	U	3.0	J	3.0	J			10.2	.
c.037	0.7	J	1.3	.	1.1	.	0.6	J	0.5	J	1.4	J	0.9	.	0.9		0.2	J
c.040	0.0	U	0.0	U			0.0	U	0.0	U	2.6	J	0.0	U			16.0	.
c.041+064+071	0.0	U	0.0	U			0.0	U	0.0	U	8.6	.	2.6	J			134.0	.
c.042+037	34.6	.	12.4	.			10.3	.	12.1	.	47.7	.	16.1	.			62.9	
c.043	0.0	U	0.0	U			0.0	U	0.0	U	0.0	U	0.0	U			1.2	J
c.044	3.2	J	0.8	J			1.0	J	6.5	J	23.3	.	1.8	J			130.8	.
c.045	0.0	U	0.0	U			0.0	U	0.0	U	0.0	U	0.0	U			0.0	U
c.046	1.5	J	0.6	J			1.1	J	0.0	U	0.0	U	0.0	U			2.3	J
c.047+075	172.7	.	143.7	.			114.6	.	52.2	.	92.6	.	133.7	.			133.6	.
c.048	11.8	.	11.2	.			7.5	.	3.4	J	7.5	J	6.2	J			14.3	.
c.049	197.8	.	47.1	.			113.3	.	38.6	.	168.7	.	188.4	.			127.4	.
c.051	0.0	U	0.0	U			0.0	U	0.0	U	0.0	U	0.0	U			0.0	U
c.052	68.0	.	20.4	.			36.4	.	37.2	.	169.8	.	26.0	.			335.2	
c.053	0.0	U	0.0	U			0.0	U	0.0	U	0.0	U	0.0	U			3.9	J
c.056+060	162.2	.	130.8	.			96.5	.	54.7	.	124.9	.	105.0				104.5	
c.059	0.7	U	0.8	U			0.4	U	1.2	U	2.8	U	0.2	U			9.3	.
c.063	50.8	J	26.4	J			48.3	J	7.2	J	41.9	J	20.2	J			26.2	J
c.066	543.0	.	481.5	.			390.2	.	183.8	.	305.3	.	413.8	.			336.0	.
c.070+076	229.2	.	166.1	.			143.7	.	86.2	.	191.1	.	150.8	.			147.6	.
c.074	325.0	.	246.8	.			210.7	.	89.0	.	187.6	.	253.3	.			211.5	.
c.077	8.9	.	10.4	.	9.5	.	7.7	J	3.9	J	8.0	J	7.1	.	6.7		2.3	.
c.081	1.0	J	0.7	.	0.7	.	1.4	J	0.8	J	1.2	J	0.9	.	0.6		1.2	J
c.082	25.3	.	12.1	.			17.8	.	12.9	.	41.3	.	12.1	.			35.5	.
c.083	3.4	J	2.1	J			2.9	J	13.5	.	5.7	J	3.4	J			11.6	.
c.084	25.0	.	7.7	.			20.0	.	26.5	.	40.2	.	6.6	J			70.9	.
c.085	77.1	J	42.7	J			56.5	J	19.6	J	102.1	J	117.0	J			874.3	J
c.087+115+081	87.4	.	18.5	.			37.1	.	29.6	.	88.8	.	65.9	.			40.9	.
c.089	1.5	J	1.2	J			1.2	J	2.1	J	1.5	J	0.5	J			1.3	J
c.091	44.0	.	23.0	.			27.6	.	17.6	.	33.8	.	21.0	.			44.8	
c.092	9.0	.	3.2	J			14.8	.	3.7	J	6.2		3.3	J			59.6	
c.095	12.5	.	107.9				4.7	J	7.6	J	23.5	.	26.6	.			113.6	.

Client Reporting Sample ID:	96KICT01_rep.1		96KICT03_rep.1		96KICT03_rep.2		96KICT05_rep.1		96KICT07_rep.1		96KICT09_rep.1		96KICT10_rep.1		96KICT10_rep.2		TEKIB06_rep.1	
Sample Type:	Common Tern		Common Tern		Common Tern		Common Tern		Common Tern		Common Tern		Common Tern		Common Tern		Forster's Tern	
Sample Lipid Content (%):	5.4		7.7		7.7		9.4		7.5		5.3		7.7		7.7		7.8	
Sample Moisture Content (%):	73.8		79.1		79.1		78.5		72.5		74.7		70.4		70.4		78.4	
c.097	41.4	.	15.9	.			21.7	.	21.3		73.6	.	17.5	.			75.5	.
c.099	524.2	.	321.4	.			310.2	.	198.8	.	236.5	.	338.2	.			381.4	.
c.100	4.3	J	3.7	J			2.4	J	1.6	J	3.3	J	3.2	J			5.3	J
c.101+090	386.8	.	152.0	.			229.2	.	155.2	.	286.9	.	260.8	.			346.5	.
c.105	176.9	.	113.8	.			113.5	.	108.0	.	121.6	.	127.9	.			118.1	.
c.107+147	10.3	.	6.0	J			26.4	.	17.2	.	15.6	.	22.0	.			20.9	.
c.110+077	259.1	.	155.7	.			161.6	.	175.6	.	258.0	.	188.5	.			221.7	.
c.114	18.0	.	10.4	.			11.9	.	12.2	.	13.4		12.7	.			12.5	
c.116	688.6	.	389.3	.			399.8	.	351.4	.	359.7	.	486.8	.			421.3	
c.119	20.5	.	14.4	.			12.6	.	6.3	J	8.6	.	12.9	.			14.0	.
c.124	5.6	J	4.4	J			3.0	J	4.1	J	6.4	J	2.2	J			2.8	J
c.126	2.0	J	1.5	J	1.3	J	1.3	J	0.8	J	0.9	J	1.2	J	1.1	J	0.6	
c.128	110.6	.	54.8	.			62.7	.	61.5		53.3	.	75.8				77.5	.
c.129+126	3.4	J	1.4	J			1.7	J	1.6	J	5.0	J	1.4	J			7.0	J
c.130	25.3	.	14.8	.			13.3	.	13.3	.	21.0	.	15.1	.			38.4	.
c.131	3.6	J	2.6	J			3.2	J	3.0	J	5.6	J	1.4	J			12.5	.
c.132	141.4	J	32.7	J			30.0	J	54.4	J	39.6	J	269.0	J			42.2	J
c.134	0.0	U	0.0	U			0.0	U	0.0	U	0.0	U	0.0	U			0.0	U
c.135+144	6.5	.	0.8	J			1.4	J	3.5	J	2.7	J	4.5	J			22.0	.
c.136	0.4	J	0.0	U			0.0	U	2.0	J	0.0	U	0.2	J			5.1	J
c.137	40.7	.	16.6	.			21.7	.	23.0	.	19.1	.	25.7	.			27.4	.
c.138+160+163	824.3	.	394.8	.			461.1	.	474.1	.	346.9	.	565.3	.			528.8	
c.141+179	4.6	J	4.0	J			2.7	J	7.8	J	7.3	J	3.7	J			2.6	J
c.146	146.2	.	83.2	.			70.2	.	85.4	.	86.2	.	82.6				125.4	.
c.149+123	151.7	.	56.0	.			96.6	.	106.8	.	140.8	.	78.6	.			163.4	.
c.151	3.1	J	1.1	J			2.1	J	4.0	J	6.6	J	1.4	J			22.1	.
c.153	1026.4	.	438.6	.			558.3	.	596.5	.	439.3	.	675.3	.			641.1	.
c.156	58.0	.	26.5	.			35.8	.	30.5		26.2	.	36.9				38.0	.
c.158	55.6	.	20.7	.			27.5	.	35.1		23.4	.	38.1	.			36.1	.
c.167	45.7	.	21.2	.			27.7	.	26.6	.	21.6	.	28.6	.			28.6	.
c.169	0.2	UJ	0.2	J	0.2	J	0.1	J	0.1	J	0.1	J	0.1	J	0.1	J	0.4	UJ
c.170+190	168.5	J	54.9	J			73.1	J	73.6	J	52.6	J	116.4	J			104.9	J
c.171+202	55.0	.	20.6	.			27.1	.	28.3		21.3	.	35.0	.			37.7	
c.172	46.9	.	18.4	.			22.8	.	20.9	.	18.2	.	26.1	.			31.0	.
c.173	0.7	J	0.0	U			0.7	J	1.0	J	0.0	U	0.3	J			1.2	J
c.174	28.2	.	11.3	.			18.1	.	14.6	.	22.9	.	19.2	.			33.7	.
c.175	6.7	J	1.9	J			2.1	J	1.4	J	2.7	J	3.3	J			4.4	J
c.176	10.6	.	4.8	J			10.7	.	15.4	.	16.9	.	3.3	J			7.6	.
c.177	43.9	.	26.0	.			30.2	.	23.0	.	27.9	.	29.1	.			48.8	.
c.178	9.2	.	5.1	J			5.1	.	5.4	J	9.3		4.7	J			28.5	.
c.180	634.7	.	252.7	.			334.8	.	388.5	.	266.4	.	413.5	.			350.3	
c.183	152.0	.	56.0	.			75.5	.	93.3	.	56.9	.	104.1	.			78.9	.
c.185	2.1	J	0.0	U			1.4	J	1.1	J	1.6	J	1.7	J			2.9	J
c.187+182	283.5	.	112.5	.			143.2	.	149.2		136.6	.	168.0	.			236.9	
c.189	8.3	J	3.2	J			4.4	J	4.3	J	2.5	J	5.1	J			4.8	J
c.191	8.7	J	3.1	J			4.5	J	4.8	J	3.5	J	5.3	J			4.8	J
c.193	27.5	.	10.8	.			14.5	.	12.5	.	9.8	.	16.7	.			17.2	
c.194	77.9	.	32.6	.			41.0	.	40.7	.	31.6	.	56.0	.			50.5	
c.195+208	37.0	J	16.1	J			18.5	J	19.2	J	14.9	J	25.0	J			29.5	J
c.197	5.4	J	2.4	J			3.0	J	2.7	J	2.4	J	3.5	J			3.3	J
c.198	4.6	J	2.5	J			2.0	J	1.9	J	2.4	J	3.0	J			4.2	J
c.199	56.8	.	34.3	.			37.6	.	37.0	.	39.3	.	51.1	.			52.0	.
c.200	1.7	J	1.1	J			1.3	J	0.0	U	2.5	J	1.1	J			3.1	J
c.201+157	4.7	J	3.2	J			3.6	J	3.1	J	3.9	J	3.5	J			6.1	J
c.203+196	108.1	.	45.1	.			55.3	.	57.1	.	45.1	.	79.0				66.9	.
c.205	11.2	.	5.2	J			6.8	.	5.1	J	6.3	J	9.2	J			7.8	.
c.206	30.5	.	17.7	.			18.9	.	18.3	.	17.2	.	25.9				28.2	.
c.207	6.8	J	4.3	J			4.0	J	4.0	J	3.6	J	5.9	J			5.6	J
c.209	13.2	J	11.6	J			7.6	J	8.7	J	9.6	J	13.3	J			11.6	J
cong.sum	9010.9	.	4946.6	.			5244.7	.	4413.6	.	5390.3	.	6411.2	.			8091.7	
cong.sum.less85	8833.8	.	4903.9	.			5185.1	.	4393.8	.	5288.2	.	6294.2	.			7217.4	.

Batch Matrix	Tern Eggs	Tern Eggs						
Region	Kidney Island, Green Bay	Kidney Island, Green Bay						
Reporting Unit	ng/g, wet weight	ng/g, wet weight						
Client Reporting Sample ID:	TEKIB18_rep.1	TEKIB18_rep.2	TEKIB24_rep.1	TEKIB30_rep.1	TEKIB48_rep.1	TEKIB48_rep.2	TEKIB60_rep.1	
Sample Type:	Forster's Tern	Forster's Tern	Forster's Tern	Forster's Tern	Forster's Tern	Forster's Tern	Forster's Tern	
Sample Lipid Content (%):	6.5	6.5	6.6	5.2	7.0	7.0	6.1	
Sample Moisture Content (%):	77.3	77.3	77.9	77.6	76.1	76.1	73.3	
Congener Numbers								
c.001	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.003	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.004+010	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.006	0.0	U	3.6	U	1.0	U	1.4	U	2.1	U	0.0	U	2.1	U
c.007 009	0.0	U	1.2	J	0.4	J	0.3	J	0.4	J	0.0	U	0.4	J
c.008 005	0.0	U	2.8	J	0.0	U	0.0	U	1.1	J	0.0	U	1.1	J
c.012.013	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.016.032	3.1	J	6.3	J	3.3	J	3.0	J	5.7	J	3.1	J	5.7	J
c.017 015	0.5	J	2.4	J	1.4	J	0.9	J	1.6	J	0.5	J	1.6	J
c.018	1.9	J	3.8	J	3.0	J	2.1	J	3.2	J	1.9	J	3.2	J
c.019	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.021	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.022	7.0	J	7.1	J	4.9	J	6.7	J	4.6	J	7.0	J	4.6	J
c.024+027	0.0	U	1.7	J	0.9	J	0.6	J	1.3	J	0.0	U	1.3	J
c.025	0.6	J	1.3	J	0.0	U	1.4	J	1.1	J	0.6	J	1.1	J
c.026	5.3	J	7.2	J	8.1	J	4.9	J	1.8	J	5.3	J	1.8	J
c.028	167.9	.	252.1	.	96.7	.	140.5	.	122.8	ok	167.9	.	122.8	ok
c.029	0.0	U	1.2	J	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.031	48.3	.	73.1	.	32.1	.	38.6	.	39.5	ok	48.3	.	39.5	ok
c.033.020	1.9	J	3.5	J	1.9	J	1.9	J	1.2	J	1.9	J	1.2	J
c.037	0.3	J	0.2	J	0.2	J	0.6	J	0.3	J	0.3	J	0.3	J
c.040	7.9	J	9.7	.	10.7	.	7.0	J	9.4	ok	7.9	J	9.4	ok
c.041+064+071	3.6	J	4.5	J	0.0	U	0.0	U	0.0	U	3.6	J	0.0	U
c.042+037	39.5	.	40.6	.	34.9	.	26.0	.	31.2	ok	39.5	.	31.2	ok
c.043	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.044	78.2	.	101.0	.	82.1	.	84.5	.	86.3	ok	78.2	.	86.3	ok
c.045	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.046	0.0	U	0.0	U	1.2	J	1.2	J	3.1	J	0.0	U	3.1	J
c.047+075	123.1	.	123.1	.	68.7	.	104.5	.	100.7	ok	123.1	.	100.7	ok
c.048	6.2	J	38.0	.	7.1	J	7.8	J	6.3	J	6.2	J	6.3	J
c.049	71.7	.	91.0	.	98.0	.	58.7	.	75.8	ok	71.7	.	75.8	ok
c.051	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U	0.0	U
c.052	269.2	.	379.3	.	200.1	.	258.2	.	235.3	ok	269.2	.	235.3	ok
c.053	1.4	J	2.5	J	1.9	J	1.0	J	0.8	U	1.4	J	0.8	U
c.056+060	73.7	.	87.9	.	49.6	.	74.2	.	67.3	ok	73.7	.	67.3	ok
c.059	4.6	U	5.3	J	5.5	J	3.9	U	3.9	U	4.6	U	3.9	U
c.063	38.9	J	21.2	J	17.9	J	31.8	J	30.2	J	38.9	J	30.2	J
c.065	311.5	.	404.9	.	194.2	.	318.4		298.8	ok	311.5	.	298.8	ok
c.070+076	109.3	.	89.2	.	76.1	.	98.1	.	89.9	ok	109.3	.	89.9	ok
c.074	180.1	.	232.2	.	115.0		171.4	.	165.1	ok	180.1	.	165.1	ok
c.077	3.3	J	1.2	.	1.2	.	3.1	.	1.7	ok	3.3	J	1.7	ok
c.081	1.3	J	0.7	J	0.5	J	0.5	J	0.6	J	1.3	J	0.6	J
c.082	23.5	.	15.6	.	17.8		18.1	.	14.9	ok	23.5	.	14.9	ok
c.083	8.0	J	8.7	J	9.3	J	6.3	J	7.1	J	8.0	J	7.1	J
c.084	38.7	.	22.2	.	28.9	.	28.1	.	75.3	ok	38.7	.	75.3	ok
c.085	283.2	J	0.0	J	85.4	J	31.4	J	48.2	J	283.2	J	48.2	J
c.087+115+081	28.2	.	31.0	.	36.7	.	25.8	.	31.3	ok	28.2	.	31.3	ok
c.089	0.7	J	0.0	U	0.0	U	0.0	U	0.0	U	0.7	J	0.0	U
c.091	27.8	.	27.2	.	23.4	.	23.8	.	34.3	ok	27.8	.	34.3	ok
c.092	38.9	.	48.1	.	28.6	.	40.7	.	35.6	ok	38.9	.	35.6	ok
c.095	46.4	.	66.7	.	49.2	.	57.8	.	47.0	ok	46.4	.	47.0	ok

Client Reporting Sample ID:	TEKIB18_rep.1		TEKIB18_rep.2		TEKIB24_rep.1		TEKIB30_rep.1		TEKIB48_rep.1		TEKIB48_rep.2		TEKIB60_rep.1	
Sample Type:	Forster's Tern		Forster's Tern		Forster's Tern		Forster's Tern		Forster's Tern		Forster's Tern		Forster's Tern	
Sample Lipid Content (%):	6.5		6.5		6.6		5.2		7.0		7.0		6.1	
Sample Moisture Content (%):	77.3		77.3		77.9		77.6		76.1		76.1		73.3	
c.097	60.5	.			55.9	.	41.5	.	52.7	.			43.2	ok
c.099	246.4	.			299.4	.	163.9	.	239.8	.			336.4	ok
c.100	4.0	J			3.0	J	1.9	J	2.1	J			4.0	J
c.101+090	244.6	.			267.6	.	211.3	.	226.9	.			237.4	ok
c.105	96.9	.			108.9	.	65.8	.	90.6	.			109.3	ok
c.107+147	25.2	.			13.1	.	0.0	U	21.7	.			0.0	U
c.110+077	216.3	.			236.1	.	153.0	.	197.7	.			184.0	ok
c.114	9.8	.			10.0	.	5.9	J	9.3	.			10.5	ok
c.116	283.9	.			345.2	.	199.1	.	309.4	.			381.3	ok
c.119	9.9	.			11.8	.	7.2	J	9.1	.			12.8	ok
c.124	4.1	J			0.0	U	0.0	U	1.6	J			0.0	U
c.126	0.7	J			0.4	.	0.3	.	0.8	J			0.7	ok
c.128	55.4	.			57.2	.	45.2	.	49.0	.			77.4	ok
c.129+138	4.8	J			4.9	J	3.2	J	4.5	J			3.1	J
c.130	24.3	.			24.9	.	19.5	.	21.9	.			32.2	ok
c.131	6.9	J			6.8	J	6.0	J	7.4	J			11.1	ok
c.132	32.3	J			54.5	J	43.0	J	52.2	J			81.5	J
c.134	0.0	U			0.0	U	0.0	U	0.0	U			0.0	U
c.135+144	11.9	.			12.3	.	10.9	.	14.9	.			10.3	ok
c.136	0.0	U			1.8	J	2.2	J	3.5	J			1.8	J
c.137	17.3	.			16.7	.	14.8	.	15.1	.			25.4	ok
c.138+160+163	303.3	.			390.8	.	335.4	.	339.3	.			622.1	ok
c.141+175	3.1	J			16.0	.	19.1	.	3.4	J			1.6	J
c.146	79.8	.			92.4	.	80.5	.	79.6	.			149.1	ok
c.149+123	130.0	.			115.9	.	113.1	.	106.3	.			152.2	ok
c.151	15.6	.			20.1	.	16.8	.	18.2	.			19.9	ok
c.153	540.9	.			439.2	.	412.8	.	375.4	.			860.4	ok
c.156	2.3	J			32.5	.	24.5	.	27.7	.			40.1	ok
c.158	25.7	.			25.2	.	23.8	.	21.5	.			41.8	ok
c.167	18.3	.			19.6	.	16.2	.	20.2	.			29.5	ok
c.169	0.8	J	0.1	J	0.5	UJ	0.4	UJ	0.1	J	0.1	J	0.3	UJ
c.170+190	59.6	J			69.3	J	53.3	J	46.0	J			134.0	J
c.171+202	25.2	.			26.3	.	25.5	.	26.5	.			57.1	ok
c.172	20.9	.			22.4	.	21.5	.	19.3	.			38.1	ok
c.173	1.1	J			1.2	J	0.8	J	1.7	J			6.9	J
c.174	26.5	.			24.3	.	29.0	.	22.0	.			48.6	ok
c.175	3.3	J			3.5	J	0.0	UJ	2.7	J			7.4	J
c.176	5.3	J			4.3	J	6.0	J	10.4	.			5.0	J
c.177	30.3	.			29.7	.	32.8	.	0.0	J			45.5	ok
c.178	14.1	.			15.6	.	13.5	.	17.2	.			30.2	ok
c.180	233.1	.			257.4	.	278.8	.	194.2	.			669.9	ok
c.183	54.5	.			57.2	.	61.6	.	47.9	.			129.9	ok
c.185	2.8	J			2.5	J	2.8	J	1.8	J			3.6	J
c.187+182	135.6	.			148.8	.	156.8	.	144.3	.			314.8	ok
c.189	3.3	J			3.7	J	2.8	J	2.8	J			6.6	J
c.191	3.4	J			3.5	J	3.6	J	2.1	J			6.6	J
c.193	10.6	.			12.7	.	12.1	.	9.7	.			23.2	ok
c.194	36.3	.			39.9	.	32.5	.	32.8	.			80.8	ok
c.195+208	17.9	J			23.9	J	15.8	J	22.3	J			44.3	J
c.197	2.8	J			2.9	J	2.6	J	2.8	J			5.6	J
c.198	3.1	J			3.2	J	2.6	J	2.7	J			5.2	J
c.199	36.2	.			41.6	.	32.9	.	39.7	.			69.0	ok
c.200	1.7	J			1.9	J	1.7	J	1.7	J			2.4	J
c.201+157	7.8	J			4.1	J	4.2	J	6.4	J			10.2	ok
c.203+196	47.8	.			52.1	.	43.8	.	45.7	.			102.8	ok
c.205	5.9	J			6.9	J	4.5	J	5.1	J			12.1	ok
c.206	23.8	.			24.5	.	15.1	.	25.9	.			50.2	ok
c.207	5.4	J			6.2	J	3.6	J	5.5	J			9.4	J
c.209	15.3	J			16.9	J	9.3	J	13.3	J			29.2	J
cong.sum	5426.3	.			5798.2	.	4319.4	.	4726.0	.			7096.0	ok
cong.sum.less85	5143.1	.			5798.2	.	4234.0	.	4694.7	.			7047.9	ok

TECHNICAL PROCEDURES

INTRODUCTION

The objective of this project was to prepare and analyze approximately 123 biota tissue samples to determine concentrations of polychlorinated biphenyl (PCB), and conduct related ancillary measurements. The PCB target analytes are listed in Attachment 2. Battelle analyzed fish and eggs that were collected between the spring of 1996 and the fall of 1996. The samples were shipped to Battelle in April, May, and June, 1997 and the Battelle laboratory component of this project began in early May, 1997.

SAMPLE ANALYSIS

SAMPLE RECEIPT, STORAGE, AND HOLDING TIMES

Hagler Bailly arranged for shipment of the frozen samples to Battelle. The samples were, upon receipt, logged into the laboratory and given unique Battelle IDs. The samples were stored frozen at, or below, -20°C until laboratory preparation could begin.

The tissue samples were stored frozen until they could be homogenized and composited. Homogenized and composited tissue samples were returned to frozen storage once they had been subsampled for extraction, or upon completion of the homogenization/compositing procedures, if extraction could not begin within one day. The sample holding times were 1 year from collection to extraction, as long as they were stored frozen until sample preparation begins. Sample extracts were to be analyzed within 40 days of extraction. Table 1 presents the 1 year holding time expiration dates. All samples were extracted by these dates and the extract holding times were also consistently met.

Table 1. Fish and Egg Sample Holding Time Expiration Dates

Sample Matrix	Holding Time Expiration Date
Walleye — whole body	July 29, 1997
Walleye — liver	July 29, 1997
Brown Trout — whole body	July 29, 1997
Brown Trout — fillet	October 11, 1997
Lake Trout — whole body	October 22, 1997
Lake Trout — fillet	August 12, 1997
Lake Trout — eggs	October 22, 1997
Tern — eggs	May 29, 1997

PRELIMINARY SAMPLE COMPOSITING, SPLITTING, AND PREPARATION

The tissue was thawed and homogenized. A Hobart stainless steel grinder was used to homogenize the fillets and the whole body fish. This large-sample homogenate was collected in a stainless steel bowl, thoroughly mixed, and approximately 400 g removed for keep (the balance of the tissue homogenate was discarded). Each individual fish and fillet was homogenized and stored separately. A Tekmar Tissuemizer was used to further homogenize the fish fillet and whole body fish tissue that was used for laboratory analysis. The Tekmar Tissuemizer was also used to homogenize the livers and eggs. The homogenized sample was placed in a pre-cleaned glass jar, with Teflon lined cap, for subsequent storage. The final whole body walleye and brown trout samples were generated by compositing approximately 30 g (±0.3 g) aliquots of the homogenized tissue from several fish, and assigning this composite sample a new sample ID (in accordance with a compositing and sample ID scheme provided by Hagler Bailly). The number of samples prepared and analyzed are listed in Table 2.

Table 2. Number of Samples for Analysis

Sample Matrix	Base 106 PCB Congeners	Total PCB (as Aroclor)	Coplanar PCB Congeners
Walleye — whole body	31 [a]	0	5
Walleye — liver	0	17 [b]	0
Brown Trout — whole body	10 [c]	0	2
Brown Trout — fillet	0	14	0
Lake Trout — whole body	12 [d]	0	12
Lake Trout — fillet	0	15	0
Lake Trout — eggs	12	0	12
Tern — eggs	12	0	12
Total:	77	46	43

[a] The 31 walleye whole body samples were composited from 138 fish (3–6 fish/composite).
[b] The 17 liver samples were individual livers from 16 fish and one sample was the composite of livers from 4 fish.
[c] The 10 whole body brown trout samples were composited from 50 fish (4–6 fish/composite).
[d] The 12 lake trout whole body samples will each be of 1 fish (i.e., not composited).

SAMPLE PREPARATION

The samples were analyzed in analytical batches of no more than 20 field samples per matrix type. The following eight (8) analytical batches were analyzed:

- 1 batch of walleye liver samples
- 2 batches of brown trout and lake trout fillet samples
- 1 batch of tern egg samples (6 forsters tern and 6 common tern eggs)
- 1 batch of lake trout egg samples
- 3 batches of combinations of walleye/brown trout/lake trout whole body samples

Additionally, there was one batch with a combination of walleye liver and trout fillet samples because several of these samples had relatively low recoveries the first time they were analyzed, and they were therefore re-analyzed in one batch.

The following quality control samples were processed along with the field samples (key quality control data quality objectives are listed in Attachment 3):

- 1 procedural blank (PB)
- 1 blank spike (BS)
- 1 certified reference material (CRM). The NRC material CARP-1 was used.
- 1 sample duplicate (DUP)

Additionally, equipment/rinse blank (EB) samples were generated during the homogenization process and instrument blanks (IB) were analyzed. The EB was a solvent (hexane) rinse of the sample homogenization equipment. One EB was prepared with each batch of samples. The IB was 1 mL of hexane that was fortified with internal standards and injected onto the GC/ECD. One IB was analyzed with each batch of samples, to determine if there was any instrument "background" signal. The EB and IB samples were quantified like the PB sample, and the average sample weight of the analytical batch was used to calculate concentrations.

Tissue Extraction and Preparation

The tissue homogenate sample was thoroughly mixed and approximately 3–10 g was removed for the extraction (Table 3). The amount of tissue used for the extraction, and the eventual pre-injection volume (PIV) the sample was adjusted to, depended on the expected PCB congener concentrations (as communicated to Battelle by Hagler Bailly during the planning phase of this project). The sample was fortified with surrogate internal standards [SISs: PCB congeners Cl_3(36) and Cl_5(112)] to monitor procedural efficiency. Sodium sulfate was added to dry the sample and aid in the maceration, and the sample was serially extracted three times in a Teflon jar using hexane as the extraction solvent and a Tekmar Tissuemizer. The combined extract concentrated using a Kuderna-Danish apparatus and gentle nitrogen gas evaporation on an N-Evap.

Table 3. Target Weight and PIVs

Sample Matrix	Approximate Sample Weight Extracted (g)	Approximate Pre-Injection Volume (mL) [a]
Walleye — whole body	5	2
Walleye — liver	3	10
Brown Trout — whole body	5	4
Brown Trout — fillet	10	2
Lake Trout — whole body	5	2
Lake Trout — fillet	10	2
Lake Trout — eggs	5	4
Tern — eggs	5	10

[a] The PIV for the base congener analysis was half of this if the sample was split for coplanar congener analysis.

The extract was next purified using a chromatography column packed with 20-g, 2% deactivated F-20 alumina (a 40-g alumina column was used for the egg samples). The column eluant was concentrated using Kuderna-Danish technique and further purification was obtained by serially treating the extract with sulfuric acid until there was no visible reaction.

The alumina and sulfuric acid purified sample was concentrated using Kuderna-Danish and nitrogen evaporation techniques and adjusted to the desired PIV. If coplanar PCB analysis was to be performed, the final extract was split 50:50, with one half being submitted for coplanar PCB fractionation (see Coplanar PCB Congener Determination below) and the other half fortified with recovery internal standards [RIS: PCB congeners Cl_3(34), Cl_3(39), and Cl_6(166)] and submitted for instrumental analyses.

Ancillary Measurements

Moisture and lipid content was determined following standard gravimetric protocols. In summary, the lipid content was determined as the "hexane extractable matter" by subsampling 10 mL of the approximately 200 mL combined sample extract, allowing it to dry and weighing the material twice at least 1 hour apart to ensure complete solvent evaporation. The volume of the sample extract, from which the subsample was removed for the lipid determination, was accurately measured by marking the volume level on the outside of the glassware prior to removing the subsample for the lipid measurement. Once the balance of the extract had been transferred for concentration, the original extract volume was determined by pouring water into the glassware to the marked level and measuring the volume using a graduated cylinder.

In addition to the hexane extractable lipid determination, which was performed on all samples, three whole body trout samples were extracted separately using dichloromethane (DCM) for determination of the DCM extractable lipid content. This was performed to obtain data to compare the lipid data generated with the standard hexane extraction with that obtained using DCM.

The moisture content was determined by placing approximately 5 g of wet tissue material in a tarred weighing pan, which was then dried at least 24 hours in a drying oven at 105°C. The dry material was then removed from the oven and allowed to come to room temperature before it was again weighed. The weighing was repeated at least 6 hours later to ensure complete dryness.

Coplanar PCB Congener Determination

A sub-set of the samples analyzed for ortho substituted PCB congeners ("standard" congeners) were also analyzed for coplanar (non-ortho substituted) PCB congeners. A total of 26 samples were processed and analyzed for coplanar PCB congeners.

The final purified extract prepared for standard PCB congener analysis was split 50:50 prior to the addition of the RIS, as described above. The coplanar PCB congener analysis was performed on one of the two splits, after isolating the coplanar congeners in accordance with Draft EPA Method 1668, *Toxic PCBs by HRGC/HRMS*:

Approximately 25 ng of the coplanar PCB congener SIS [Cl4(77)–*deuterated*] was added to the coplanar extract split to monitor the efficiency of the column separation and coplanar PCB congener isolation. A 9-mm glass column was packed with 3.6 g of a 50:50 mixture of Carbopack C:Celite 545 that had been activated at 130°C for a minimum of 6 hours; the column was packed in hexane. The sample extract was loaded onto the column, rinsing the sample vial with approximately 1 mL of hexane, which was added to the column. The solvent level was brought to the top of the column and the column eluted as follows:

- 25 mL of hexane was added, eluted, and collected as the F1 (standard congeners).
- 15 mL of methanol was added, eluted, and collected as the F2 (residual polar/lipid matrix components).
- 15 mL of toluene was added, eluted, and collected as the F3 — the coplanar PCB congeners elute in this fraction.

The F3 fraction (coplanar PCB congener) was concentrated to approximately 200–250 μL using nitrogen evaporation techniques, fortified with the RIS compounds, and submitted for GC/ECD analysis.

INSTRUMENTAL ANALYSIS

GC/ECD Analysis — PCB Congener Analysis

The analysis of the target PCB congener compounds (Attachment 2) was performed by high-performance capillary gas chromatography with electron capture detection (GC/ECD) using a Hewlett-Packard 6890 or 5890-II gas chromatograph fitted with dual ^{63}Ni-electron capture detectors. The GC/ECD analysis was performed using a 60-m, 0.25-mm inner diameter, 0.25-μm film thickness, DB-5 fused silica capillary column (J&W Scientific, Inc.). A 1 µL sample extract was injected onto the instrument. The injected sample was al split to a second column (60-m, 0.25-mm inner diameter, 0.25-μm film thickness, DB-1701 column) and ECD, for simultaneous acquisition of second column GC/ECD data. The second column were acquired in case these data would be needed for review at a later time, but the analyses on the DB-1701 will not be calibrated and the data were not reduced for this project (the DB-1701 runs were, however, checked to ensure that the data were acquired).

The GC was equipped with an electronic pressure controlled (EPC) inlet for optimum sensitivity and reproducibility. Additionally, hydrogen was used as the carrier gas, and the temperature program was optimized to separate the 106 target PCB congeners. The following GC temperature program was used:

- Initial temperature 60 °C
- Initial time 1 minute
- Ramp Rate 10 °C/minute to 140 °C; 1 °C/minute to 220 °C; 5 °C/minute to 290 °C
- Final temperature 290 °C; 10 minutes

The GC/ECD system was calibrated with a multilevel calibration, with a minimum of 4 calibration points (5 points were typically be used). The analyte concentrations range from about 0.005 to 0.12 ng/μL in the calibration solutions (the concentrations of some congeners was higher, because of their lower ECD response), and the internal standard concentrations were approximately 0.05 to 0.06 ng/μL in all calibration levels. The calibration solutions were prepared with all 106 target congeners and the internal standards. For the coeluting sets of congeners (see Attachment 2), only the primary congener (the congeners listed first in Attachment 2) was used in the calibration solutions.

Each target analyte was fitted to a quadratic equation to best represent the response of the ECD. The validity of the initial calibration was monitored with a continuing calibration check analysis (a mid0level calibration standard) at least every 10 samples. Analytes concentrations were by the method of internal standards using the RIS (i.e., the internal standard added at the end of the sample processing regime) as the quantification internal standard.

Samples with target PCB congeners response above the high standard were diluted and re-analyzed. If more than 10 of the PCB congeners had a response greater than the high calibration standard, then the analytical data from only the diluted run were reduced and reported. However, if the dilution and re-analysis was performed because 10 or fewer PCB congeners had a response above the high calibration standard, then the data for all congeners were reported from the first run, and the re-analysis was only used to generate data for the congeners that were initially above the high standard (and the "E" and "D" qualifiers applied to the data, as described in Attachment 4).

Quantification of individual components was performed by the method of internal standards using the RIS compounds Cl3(39) and Cl6(166) as the quantification internal standard [Cl3(39) was used for all congeners eluting before the SIS Cl5(112), and Cl6(166) was used for the congeners eluting after this SIS]. Surrogate compound recoveries were determined for the SIS Cl3(36) versus the RIS Cl3(39), and for the SIS Cl5(112) versus the RIS Cl6(166). Target analyte concentrations were reported on a wet weight basis, and the moisture and lipid content were reported along with the PCB analytical data.

Additionally, the total PCB was estimated as the sum of the 106 congener concentrations on the spreadsheet summary tables. The sum of all congeners without congener #85 was also calculated because there was a significant interference observed with this congener and this likely biased the total PCB data when this congener was included.

GC/ECD Analysis — Total PCB Analysis (as Aroclor Equivalent)
The total PCB concentrations in the walleye liver and brown trout and lake trout fillet was determined by the Aroclor equivalent method; no individual PCB congener data were generated for these samples. The sample extraction and preparation was the same as for the PCB congener analysis, and the instrumental analysis was also as described above except for the calibration standards that was used.

The initial calibration verification was performed with a multilevel calibration containing a mixture of Aroclors 1016 and 1260 (with a concentration range of approximately 0.25 to 5 μg/mL per Aroclor). Single-point calibration standards were analyzed for the other target Aroclor formulations (Attachment 2). Additionally, a 50:50 mixture of Aroclors 1248:1254 was analyzed as a single-point calibration standard. The validity of the calibration was checked with a mid level calibration mixture of 1016 and 1260 (with a concentration of approximately 2 μg/mL) no less frequently than every 10 samples. The multilevel calibration would be used to quantify the samples that most closely resemble 1016 and 1260, and the appropriate single-point calibrations was used for the other Aroclor formulations.

Total PCB was determined as the most predominant Aroclor formulation (i.e., the analysts reviewed the chromatogram and determined which single Aroclor the PCB composition in the sample most closely resembles, and quantified the sample as the equivalent of that Aroclor). Because the Aroclor composition relatively closely resembled a 50:50 mixture of Aroclors 1248 and 1254 (ranged from about 40:60 to 60:40) in the walleye liver samples, the standard with a 50:50 of these Aroclors was used to quantify those samples, and the results were reported as "1248,1254". The PCB pattern in the trout fillets most closely resembled Aroclor 1254, and this formulation was used to quantify the fillets.

The RIS Cl6(166) was used as the quantification internal standard for Aroclors 1248, 1254, and 1260, and the RIS Cl3(39) was used for Aroclors 1221, 1016, 1232, and 1242. The RIS Cl3(39) was also used to determine the recovery of the SIS Cl3(36) and the RIS Cl6(166) was used to determine the recovery of the SIS Cl5(112).

GC/ECD Analysis — Coplanar PCB Congener Analysis
Samples selected for coplanar PCB congener analysis were processed for the isolation of these congeners, and separately submitted for GC/ECD analysis. The GC analytical conditions was the same as for the analysis of standard PCB congeners. The same calibration and quantification approach was also used for the coplanar congeners. Sample quantification will be performed versus the RIS Cl6(166) for all coplanar congeners except congener #37; the RIS Cl3(39) was used to quantify congener #37. The recovery of the column fractionation internal standard Cl4(77)-*deuterated* was determined versus the RIS Cl6(166). The recovery of Cl4(77)-*deuterated* was an indicator of the sample processing efficiency after the sample was split for coplanar PCB congener processing. The efficiency of the rest of the sample processing was indicated by the recoveries of the standard SISs [Cl3(36) and Cl5(112)], which were reported with the standard PCB congener analysis data for each sample.

GC/MS Confirmatory Analysis
The quantity of the standard PCB congeners was confirmed using quadrupole gas chromatography with mass spectrometric detection using a Hewlett-Packard Model 5972 MSD. All field samples that were analyzed for the base congeners by GC/ECD (77 samples) were also analyzed by GC/MS. However, the GC/MS data were only reduced and reported for 26 of the 77 samples; 4 tern egg, 4 lake trout egg, 10

walleye whole body, 4 brown trout whole body, and 4 lake trout whole body samples. The 26 samples were selected using criteria developed by Hagler Bailly (species, tissue type, location of capture, and PCB concentration determined in the GC/ECD analysis). GC/MS confirmation was not be performed on the coplanar PCB congener samples or the samples that were analyzed for total PCB as Aroclor equivalent. The GC/MS analysis was performed on the field samples and the PB samples — the QC data, including surrogate compound recoveries, were generated from the GC/ECD analyses.

The gas chromatograph was fitted with the same chromatography column and operated with the same oven temperature profile as that used for the primary GC/ECD analysis. However, helium was used as the carrier gas instead of hydrogen. This ensure that the peaks tentatively identified by GC/ECD had comparable chromatographic properties in the GC/MS analysis. However, because helium was used as the carrier gas instead of hydrogen, congeners #153 and #132, which were resolved on the GC/ECD, could not be resolved in the GC/MS analysis of the whole body fish samples (they were resolved during the analysis of the egg samples).

The mass spectrometer was operated in the selected ion monitoring mode (SIM) to provide the necessary sensitivity and selectivity. Each target congener was monitored using two ions — a primary ion for quantitation and a secondary ion, for structural identification and confirmation. Identifications was based on chromatographic retention time and primary/secondary ion ratio criteria (i.e., identification of the peak as a PCB congener, the level of chlorination of that PCB congener, and the known retention time characteristics of each congener from prior detailed GC/ECD retention time characterization/mapping).

The GC/MS analytical system was tuned with perfluorotributylamine (PFTBA), and calibrated with a multilevel calibration. A minimum of 4 calibration levels (but typically 5 points), was used with the analyte concentrations in a range of approximately 0.02 to 0.8 ng/μL. The calibration solutions contained all 106 target base eongeners and the internal standards. The GC/MS analytes were quantified versus the RIS C13(34).

The GC/MS confirmatory analysis was performed like a standard quantitative analysis, with the GC/MS data being reported just like the GC/ECD data. There was no quantitative comparison of the GC/ECD and GC/MS analytical results.

MDL STUDY

A method detection limit (MDL) study was performed as part of this project using "clean" (hatchery raised) trout fillet provided by Hagler Bailly. The MDL study was performed in accordance with the EPA protocol set forth in 40 CFR 136 Appendix B Method Detection Limit (MDL) Determination.

The MDL study involved fortifying eight replicate tissue homogenate sub-samples with the 106 target base PCB congeners at a concentration of approximately 3 to 5 times the expected MDL, and processing and analyzing them using the procedure that was used for the project field samples. Additionally, two non-spiked sub-samples were analyzed to determine the background PCB levels in the tissue, and a procedural blank analysis will also be included. The PCB congener concentrations were determined by GC/ECD, and the summary statistics performed to calculate the MDL for each PCB congener.

QUALITY ASSURANCE / QUALITY CONTROL

QUALITY ASSURANCE

The Quality Assurance Unit (QAU) at Battelle remains independent of all laboratory project activities. The QAU monitored Battelle's components of the project according to existing Battelle SOPs to ensure the accuracy, integrity, and completeness of the data. Additionally, the QAU monitored the project activities to ensure consistency with the applicable requirements described in the Quality Assurance Project Plan (QAPP) that was developed for this project. The QAU scope included system inspections, data audits, and reviews of documents and deliverables.

QUALITY CONTROL

Project staff were responsible for ensuring that sample tracking, sample preparation, and analytical instrument operation all met the quality control criteria detailed in the applicable analytical SOPs. The type and frequency of analysis of quality control samples for the analyses are specified in Attachment 3.

The data quality objectives (DQO) for the analyses are outlined in Attachment 3. Analytical results that did not meet the listed DQOs were submitted to and/or reviewed with the Battelle Project Manager for assessment of the potential impact of the results. Affected samples were reanalyzed at the Project Manager's discretion (e.g., a set of samples were re-extracted and re-analyzed for total PCB as Aroclor determination because surrogate recoveries fell below the DQO). Quality control sample data that were accepted outside these criteria are indicated with the appropriate data qualifier. A set of data qualifiers were applied to the final summary spreadsheet data, as indicated in Attachment 4 (e.g., quality control sample data quality objective exceedances will be qualified with a "&" on the summary spreadsheet tables). Target analyte concentrations were reported if the analyst could confidently perform the identification and determination (i.e., uncensored data were reported).

Attachment 2. Target PCB Analytes

Base PCB Congener Set [a]				
1	42/37	89	136	183
3	43	91	137	185
4/10	44	92	138/160/163	187/182
6	45	95	141/179	189
7/9	46	97	146	191
8/5	47/75	99	149/123	193
12/13	48	100	151	194
16/32	49	101/90	153	195/208
17/15	51	105	156	197
18	52	107/147	158	198
19	53	110/77	167	199
21	56/60	114 [b]	169	200
22	59	118	170/190	201/157
24/27	63	119	171/202	203/196
25	66	124	172	205
26	70/76	128	173	206
28	74	129/126	174	207
29	82	130	175	209
31	83	131	176	
33/20	84	132	177	
40	85 [b]	134	178	
41/64/71	87/115/81	135/144	180	

[a] All congeners numbers are listed using the IUPAC nomenclature.
Coeluting congeners are listed in order of abundance in Aroclors 1242/1248/1254 (most abundant listed first). The most abundant single congener will be used to calibrate the instrument for the coeluting congener sets.
[b] The pesticide 4,4-DDD coelutes with congener 114 and the pesticide 4,4-DDE coelutes with congener 85.

Attachment 2 (cont.). Target PCB Analytes

Coplanar PCB Congener Set
37
77
81
126
169
Aroclor Formulations
Aroclor 1016
Aroclor 1221
Aroclor 1232
Aroclor 1242
Aroclor 1248
Aroclor 1254
Aroclor 1260

Attachment 3. Data Quality Objectives — PCB Analysis

QC Sample	Frequency	Data Quality Objectives	Corrective Action
Procedural Blank (PB)	1 per analytical batch *	< RL, or associated samples >10 × blank concentration	Reanalyze associated samples, or justify.
Equipment Blank (EB)	1 per analytical batch	< RL, or associated samples >10 × blank concentration	Qualify data and/or describe in narrative with data reporting, or justify.
Instrument Blank (IB)	1 per analytical batch	< RL, or associated samples >10 × blank concentration	Reanalyze associated samples, or justify.
Blank Spike (BS)	1 per analytical batch *	90% of congeners to meet the following: 50—125% recovery for tri- through decachlorobiphenyls and Aroclors. 30—125% recovery for mono- and dichlorobiphenyls.	Reanalyze associated samples, or justify.
Certified Reference Material (CRM) (CARP-1)	1 per analytical batch	PD <±35% between measured and certified or consensus value for 90% of analytes; PD <±50% for all analytes. Average of PD (absolute values) <25%. Objectives apply to analytes with a certified or consensus concentration >5 × RL.	Reanalyze associated samples, or justify.
Sample Duplicate (DUP)	1 pair per analytical batch *	RPD <50% for duplicates with analyte concentrations >5 × RL. Difference <2 × RL for duplicates with analyte concentration <5 × RL.	Reanalyze associated samples, or justify.
Surrogate Compounds	Every field and QC sample	50—125% recovery	Reanalyze associated samples, or justify.
Initial Instrument Calibration (GC/ECD and GC/MS)	At initiation of analytical sequence. A minimum of 4-point calibration.	GC/ECD: Correlation coef. r >0.995 for 90% of analytes; r >0.99 for all analytes. ($r > 0.995 = r^2 > 0.99$) GC/MS: < 25% RSD in RRFs for 90% of analytes; <35% RSD for all analytes.	Recalibrate and reanalyze associated samples

* Analytical Batch: Sample set of no more than 20 field samples of the same sample matrix.

Attachment 3 (cont.). Data Quality Objectives — PCB Analysis

Continuing Instrument Calibration Check (GC/ECD and GC/MS)	No less frequently than every 10 samples	GC/ECD: determined concentration <±25% PD vs true concentration for 75% of analytes. <±35% PD for 90% of analytes; ±50% PD for all analytes. <±15% PD on average for all analytes. GC/MS: <±25% PD for RRFs versus initial calibration for 75% of analytes; <±35% PD for 90% of analytes; ±50% PD for all analytes. <±15% PD on average for all analytes.	Recalibrate and reanalyze associated samples (i.e., samples not bracketed by a passing calibration), or justify.
% Lipid determination	Replicate weighing of each sample. Sample duplicate — 1 per analytical batch [a]	<10% difference in two weighings RPD < 20%	Re-dry and re-weigh Qualify data.
% Moisture determination	Replicate weighing of each sample. Sample duplicate — 1 per analytical batch [a]	<10% difference in two weighings RPD < 20%	Re-dry and re-weigh. Re-determine moisture content for associated samples, or justify.

Attachment 4. Data Qualifiers

Data Qualifier	Purpose
Data Qualifiers *	
&	QC value outside the accuracy or precision criteria goal (SRM, BS recovery, surrogate recovery, %RPD in DUP analysis).
E	Value for analysis of compound with response above the calibration range. Sample was diluted and reanalyzed for this analyte, and the data from the diluted sample analysis are reported separately elsewhere.
D	Value for diluted analysis of compound with an original (undiluted) response above the high calibration range.
B	Analyte detected at a level above the reporting limit in the procedural blank (procedural blank value is qualified).
U	Not detected. An entry of "0" is put in the value field.
J	Estimated value. Analyte detected below the sample-specific reporting limit.
ME	Significant matrix interference — estimated value reported.
MI	Significant matrix interference — value could not be determined or estimated.
JX	Estimated value, see narrative[b].
X, Y, Z	Defined in case narrative.

* Data qualifiers that will be applied to the summary spreadsheet. Qualifying uses RLs and analyst compound identification; calculated MDLs are not used when applying the "U" or "J" qualifiers, and there is currently no qualifier for values between the MDL and the RL, or that use the MDL in any way.

[b] The JX qualifier was specifically created to qualify the congener #85 data, which in the field samples was uncharacteristically large, likely due to interference from the pesticide p,p'-DDE. This is described in more detail in a narrative in the letter data report.

Attachment 5

REPORTING LIMITS - Extended PCB Congener Set

Batch ID	97-126	97-129	97-190, 97-191, 97-192	97-191, 97-192	97-192
Matrix	*Tern eggs*	*L. Trout Eggs*	*Walleye Whole*	*B.Trout Whole*	*L.Trout Whole*
Pre-Injection Volume (uL)	5000	2000	1000	2000	1000
Lipid Analysis Split Factor	1.00	1.00	1.00	1.00	1.00
Coplanar Analysis Split Factor	2.00	2.00	1.00	1.00	1.00
Sample Dilution Factor	1.00	1.00	1.00	1.00	1.00
Sample Wet Weight (g)	5.00	5.00	5.00	5.00	10.00
Reporting Unit	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight
Analyte					
PCB1	25.6	10.2	2.6	5.1	1.3
PCB3	50.2	20.1	5.0	10.0	2.5
PCB4	25.8	10.3	2.6	5.2	1.3
PCB7	16.0	6.4	1.6	3.2	0.8
PCB6	16.0	6.4	1.6	3.2	0.8
PCB8	16.9	6.7	1.7	3.4	0.8
PCB19	9.0	3.6	0.9	1.8	0.5
PCB12	16.0	6.4	1.6	3.2	0.8
PCB18	8.4	3.4	0.8	1.7	0.4
PCB17	7.2	2.9	0.7	1.4	0.4
PCB24	9.0	3.6	0.9	1.8	0.5
PCB16	9.0	3.6	0.9	1.8	0.5
PCB29	9.0	3.6	0.9	1.8	0.5
PCB26	9.0	3.6	0.9	1.8	0.5
PCB25	9.0	3.6	0.9	1.8	0.5
PCB31	9.0	3.6	0.9	1.8	0.5
PCB28	8.4	3.4	0.8	1.7	0.4
PCB21	9.0	3.6	0.9	1.8	0.5
PCB33	9.0	3.6	0.9	1.8	0.5
PCB53	9.6	3.8	1.0	1.9	0.5
PCB51	6.2	2.5	0.6	1.2	0.3
PCB22	9.0	3.6	0.9	1.8	0.5
PCB45	7.8	3.1	0.8	1.6	0.4
PCB46	9.8	3.8	1.0	1.9	0.5
PCB52	8.4	3.4	0.8	1.7	0.4
PCB43	9.0	3.6	0.9	1.8	0.5
PCB49	9.6	3.8	1.0	1.9	0.5
PCB47	9.8	3.8	1.0	1.9	0.5
PCB48	9.6	3.8	1.0	1.9	0.5
PCB44	8.4	3.4	0.8	1.7	0.4
PCB59	9.6	3.8	1.0	1.9	0.5
PCB42	9.0	3.6	0.9	1.8	0.5
PCB41	7.4	3.0	0.7	1.5	0.4
PCB40	9.0	3.6	0.9	1.8	0.5
PCB100	9.6	3.8	1.0	1.9	0.5
PCB63	9.6	3.8	1.0	1.9	0.5
PCB74	9.6	3.8	1.0	1.9	0.5
PCB70	9.6	3.8	1.0	1.9	0.5
PCB66	8.4	3.4	0.8	1.7	0.4
PCB95	9.6	3.8	1.0	1.9	0.5
PCB91	7.4	3.0	0.7	1.5	0.4
PCB56	7.6	3.0	0.8	1.5	0.4
PCB92	6.8	2.7	0.7	1.4	0.3
PCB84	7.6	3.0	0.8	1.5	0.4
PCB89	9.6	3.8	1.0	1.9	0.5
PCB101	8.4	3.4	0.8	1.7	0.4
PCB99	9.6	3.8	1.0	1.9	0.5
PCB119	9.6	3.8	1.0	1.9	0.5
PCB83	9.6	3.8	1.0	1.9	0.5

REPORTING LIMITS - Extended PCB Congener Set

Batch ID	97-126	97-129	97-190, 97-191, 97-192	97-191, 97-192	97-192
Matrix	*Tern eggs*	*L. Trout Eggs*	*Walleye Whole*	*B.Trout Whole*	*L.Trout Whole*
PCB97	9.6	3.8	1.0	1.9	0.5
PCB87	9.6	3.8	1.0	1.9	0.5
PCB85	7.4	3.0	0.7	1.5	0.4
PCB136	9.6	3.8	1.0	1.9	0.5
PCB110	9.6	3.8	1.0	1.9	0.5
PCB82	9.6	3.8	1.0	1.9	0.5
PCB151	9.6	3.8	1.0	1.9	0.5
PCB135	7.0	2.8	0.7	1.4	0.4
PCB124	9.6	3.8	1.0	1.9	0.5
PCB107	9.6	3.8	1.0	1.9	0.5
PCB149	9.6	3.8	1.0	1.9	0.5
PCB118	8.4	3.4	0.8	1.7	0.4
PCB134	9.6	3.8	1.0	1.9	0.5
PCB114	9.6	3.8	1.0	1.9	0.5
PCB131	8.6	3.4	0.9	1.7	0.4
PCB146	7.0	2.8	0.7	1.4	0.4
PCB153	8.4	3.4	0.8	1.7	0.4
PCB132	9.6	3.8	1.0	1.9	0.5
PCB105	8.4	3.4	0.8	1.7	0.4
PCB141	9.6	3.8	1.0	1.9	0.5
PCB137	9.6	3.8	1.0	1.9	0.5
PCB176	7.8	3.1	0.8	1.6	0.4
PCB130	7.6	3.0	0.8	1.5	0.4
PCB138	8.4	3.4	0.8	1.7	0.4
PCB158	9.6	3.8	1.0	1.9	0.5
PCB129	9.6	3.8	1.0	1.9	0.5
PCB178	6.8	2.7	0.7	1.4	0.3
PCB175	9.6	3.8	1.0	1.9	0.5
PCB187	8.4	3.4	0.8	1.7	0.4
PCB183	8.4	3.4	0.8	1.7	0.4
PCB128	8.4	3.4	0.8	1.7	0.4
PCB167	9.6	3.8	1.0	1.9	0.5
PCB185	9.6	3.8	1.0	1.9	0.5
PCB174	8.0	2.4	0.6	1.2	0.3
PCB177	5.8	2.3	0.6	1.2	0.3
PCB171	9.6	3.8	1.0	1.9	0.5
PCB156	9.6	3.8	1.0	1.9	0.5
PCB173	9.6	3.8	1.0	1.9	0.5
PCB201	9.6	3.8	1.0	1.9	0.5
PCB172	9.6	3.8	1.0	1.9	0.5
PCB197	9.6	3.8	1.0	1.9	0.5
PCB180	8.4	3.4	0.8	1.7	0.4
PCB193	9.8	3.8	1.0	1.9	0.5
PCB191	9.6	3.8	1.0	1.9	0.5
PCB200	9.8	3.8	1.0	1.9	0.5
PCB169	9.6	3.8	1.0	1.9	0.5
PCB170	8.4	3.4	0.8	1.7	0.4
PCB198	9.6	3.8	1.0	1.9	0.5
PCB199	8.0	3.2	0.8	1.6	0.4
PCB203	7.6	3.0	0.8	1.5	0.4
PCB189	9.6	3.8	1.0	1.9	0.5
PCB195	8.4	3.4	0.8	1.7	0.4
PCB207	7.8	3.1	0.8	1.6	0.4
PCB194	9.8	3.8	1.0	1.9	0.5
PCB205	9.6	3.8	1.0	1.9	0.5
PCB206	6.4	2.6	0.6	1.3	0.3
PCB209	6.4	2.6	0.6	1.3	0.3

Attachment 5 (cont.)

REPORTING LIMITS - Coplanar Congeners

Batch ID	97-126	97-129	97-192	97-192	97-192
Matrix	*Tern eggs*	*L. Trout Eggs*	*Walleye Whole*	*B.Trout Whole*	*L Trout Whole*
Pre-Injection Volume (uL)	125	125	200	200	200
Lipid Analysis Split Factor	1.00	1.00	1.00	1.00	1.00
Coplanar Analysis Split Factor	2.00	2.00	2.00	2.00	2.00
Sample Dilution Factor	1.00	1.00	1.00	1.00	1.00
Sample Wet Weight (g)	5.00	5.00	5.00	5.00	10.00
Reporting Unit	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight
Analyte					
PCB37	0.24	0.24	0.38	0.38	0.19
PCB81	0.24	0.24	0.38	0.38	0.19
PCB77	0.24	0.24	0.38	0.38	0.19
PCB126	0.24	0.24	0.38	0.38	0.19
PCB169	0.25	0.25	0.40	0.40	0.20

Attachment 5 (cont.)

REPORTING LIMITS - Aroclors

Batch ID	97-124	97-127	97-128
Matrix	*Walleye Liver*	*B. Trout Fillet*	*L.Trout Fillet*
Pre-Injection Volume (uL)	10000	1000	1000
Lipid Analysis Split Factor	1.00	1.00	1.00
Coplanar Analysis Split Factor	1.00	1.00	1.00
Sample Dilution Factor	1.00	1.00	1.00
Sample Wet Weight (g)	3.00	10.00	10.00
Reporting Unit	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight
Analyte			
Aroclor1016	816.7	24.5	24.5
Aroclor1221	816.7	24.5	24.5
Aroclor1232	816.7	24.5	24.5
Aroclor1242	816.7	24.5	24.5
Aroclor1248	816.7	24.5	24.5
Aroclor1248.1254	816.7	24.5	24.5
Aroclor1254	816.7	24.5	24.5
Aroclor1260	816.7	24.5	24.5

Reporting Limit Calculation (and Spreadsheet Header Information). Sample-specific RLs are calculated directly in the Excel summary tables for application of "J" qualifiers. The reporting limits listed in the table in Attachment 5 are based on the most common PIVs for the type, lipid analysis split factor of 1.00, a dilution factor of 1.00 (no dilution), and the targeted sample weight for the sample type. Sample-specific RLs, that were actually used to qualify the data, can be calculated by using the actual factors, weights etc. listed in the spreadsheet table heading for each individual sample.

The RLs are calculated as follows:

$$RL = STD\ CONC \times PIV \times Lipid_{SF} \times Coplanar_{SF} \times Sample_{DF} \times 1/Sample_{WT}$$

RL =	Reporting limit (ng/g, wet weight)
STD CONC =	PCB concentration in low-level calibration standard (ng/µL)
PIV =	Pre-injection volume (µL). The PIV listed in the Excel data table header is used for calculating RLs, and is determined slightly differently than what is typically thought of as a PIV. Pre-injection volume in this case is the final adjusted extract volume that contains the sample for the subject analysis. This is not necessarily the same as the volume of extract that is spiked with RIS or the volume of extract placed on the GC for analysis. In the case of most analyses it is the volume the sample is adjusted to prior to analysis (and not what is removed to place on the instrument), while in the case of diluted samples it is the volume the sample is adjusted to during the dilution (and, again, not what is removed to place on the instrument). In the case of the livers, the PIV entered here is 10,000 µL because this is the adjusted volume of the entire final sample extract.
$Lipid_{SF}$ =	Lipid analysis subsampling factor. Factor that corrects for the amount removed in the lipid analysis. (total extract volume before lipid analysis) × (1 / (total extract volume before lipid analysis – extract volume removed for lipid analysis))
$Coplanar_{SF}$ =	Coplanar analysis split factor. Factor that corrects for any splitting of the extract for coplanar PCB analysis. (total extract volume before split / extract volume removed for the subject analysis)
$Sample_{DF}$ =	Sample dilution factor. Factor that corrects for any subsampling of the extract for dilution purposes (i.e., when samples were diluted and re-analyzed). This is only a factor when a portion of the extract is removed, and subsequently spiked with additional RIS. The amount of solvent added to perform the dilution is not a factor in the calculation. Additionally, this is *not* a factor if only the PIV is increased to bring the analyte response within the calibration range in a re-analysis. (total extract volume before subsampling / extract volume removed for the subject dilution and re-analysis)
$Sample_{WT}$ =	Sample weight (g, wet weight). Weight of the sample amount that was extracted.

Attachment 6

LIPID METHOD COMPARISON - Hexane vs Dichloromethane

Client Reporting ID	Matrix	Battelle ID	Analytical Batch	Lipid Content (%, wet weight) Hexane as Solvent	DCM as Solvent	PD
BTEG01CP	Brown trout whole body	VD38	97-191	11.42	12.09	5.9
BTEG02CP	Brown trout whole body	VD39	97-191	8.67	12.00	38.4
BTEG04CP	Brown trout whole body	VD40	97-191	11.20	15.76	40.7
					Average:	28.3

PD: percent difference; DCM relative to hexane.

Attachment 7

DUPLICATE MOISTURE CONTENT DETERMINATION

Client Reporting ID	Matrix	Battelle ID	Analytical Batch	% Moisture 1:st Determination	2:nd Determination	RPD
WEWG02LV	Walleye liver	VA44	97-124	61.94	41.83	38.8
BTEG01FC-1	Brown trout fillet	Z5981	97-127	77.74	77.65	0.1
LTLM01FC-1	Lake trout fillet	Z5874	97-128	67.43	61.81	8.7
LTIR08FC-1	Lake trout fillet	Z5858	97-181	67.95	61.21	10.4
TEKIB06	Tern egg	Z5797	97-126	78.44	57.51	30.8
EGLMF01FC-1	Lake trout egg	Z5958	97-129	66.78	69.41	3.9
WEFR01CP	Walleye whole body	VC53	97-190	61.04	62.46	2.3
WEEG04CP	Walleye whole body	VC73	97-191	58.70	67.42	13.8
WEFR07CP	Walleye whole body	VC59	97-192	65.38	62.22	5.0
					Average:	12.6

Attachment 8

QUANTIFICATION OF SAMPLES -- PCB by GC/ECD

Samples are quantified using the method of internal standards. The quantification internal standards are the recovery internal standards (RIS) (i.e., the internal standards added to the sample immediately prior to instrumental analysis). The concentration of target analytes is determined using the following regression equation if a linear regression calibration is used:

$$C_s = [((A_s/A_i) - b) * (Amt_i/m)] * (1/W)$$

Which is based on the linear regression equation:

$Y = mX + b$ which is equivalent to: $A_s/A_i = [(m * (C_s/Amt_i)) + b]$

where,

C_s	=	Concentration/amount of target analyte
A_s	=	Area for target analyte [e.g., PCB8]
A_i	=	Area for internal standard [e.g., PCB39]
Amt_i	=	Amount internal standard [e.g., PCB39 added to sample]
W	=	Sample size (g, dry wt)
b	=	y-intercept of linear regression equation.
m	=	slope of linear regression equation.

However, the ECD does not respond linearly and we typically calibrate with a quadratic equation for PCB target compounds. A quadratic equation was consistently used in this project (e.g, see curve type in method description on page 000073 of the bird and fish egg data package). The page references listed below for the example calculations are for the bird and fish egg GC/ECD data package.

The quadratic equation is considerably more complicated than the above listed linear regression equation, and takes a full page of calculation steps to perform. I do not think you want to subject yourself to that. We have carried out that exercise a few times to validate the data system. The method calibrates correctly as long as the correct standard amounts are put into the method (pages 000079 to 000081). The samples are correctly quantified as long as the correct recovery internal standard amounts are entered for the sample [e.g., see page 000282 where the appropriate recovery internal standards are listed with the ng amounts spiked for each sample as designated in the method]. The amount of RIS spiked into each sample can be traced to the sample preparation records [e.g. page 000013]. The two recovery internal standards are PCB39, which is used for congeners PCB1 through PCB119, and PCB166 which is used for congeners PCB112 through PCB209 (based on GC retention order and as listed on quantitation printouts).

The PCB amounts can be found on the *quantitation reports*. Quantitation reports are the data system generated reports which represent the analytes quantified with a given method, and report the result in as ng.

The PCB concentrations is calculated as follows:

$[PCB] = PCB\ Amount * Lipid_{SF} * Coplanar_{SF} * PIV_{SF} * Sample_{DF} \times 1/Sample_{WT}$

[PCB] =	PCB concentration (ng/g, wet weight)
PCB Amount =	Amount of PCB in the sample analyzed on the GC instrument, as listed on the quantitation report (ng)
$Lipid_{SF}$ =	Lipid analysis subsampling factor. Factor that corrects for the amount removed in the lipid analysis. (total extract volume before lipid analysis) × (1/(total extract volume before lipid analysis – extract volume removed for lipid analysis))
$Coplanar_{SF}$ =	Coplanar analysis split factor. Factor that corrects for any splitting of the extract for coplanar PCB analysis. (total extract volume before split / extract volume removed for the subject analysis)
PIV_{SF} =	PIV subsampling factor. Factor that corrects for subsampling of the extract prior to the addition of RIS and submission for initial instrumental. This factor only applies to liver samples which had a volume removed from the concentrated extract, and the subsample was spiked with RIS prior to analysis. (total extract volume before subsampling / extract volume removed for the subject analysis)
$Sample_{DF}$ =	Sample dilution factor. Factor that corrects for any subsampling of the extract for dilution purposes (i.e., when samples were diluted and re-analyzed). This is only a factor when a portion of the extract is removed, and subsequently spiked with additional RIS. The amount of solvent added to perform the dilution is not a factor in the calculation. Additionally, this is ***not*** a factor if only the PIV is increased to bring the analyte response within the calibration range in a re-analysis. (total extract volume before subsampling / extract volume removed for the subject dilution and re-analysis)
$Sample_{WT}$ =	Sample weight (g, wet weight). Weight of the sample amount that was extracted.

Example:

Sample ID:	Z5799, page 000305 through 000307 - quantitation report (from batch 97-126), page 000439 and 000440 (97-126 Table)
Data File Number:	pesticides,chanl_01.sa06,21,1, page 000282 (# = 21)
PCB Amount, Target Analyte:	423.7752 ng, PCB28, page 000305
$Lipid_{SF}$:	1.056, page 000010
$Coplanar_{SF}$:	2, page 000012 (10000uL was initial volume, which was then split, 5000uL to each analysis)
PIV_{SF}:	1, page 000013 (the RIS was added to the entire 5000uL)
$Sample_{DF}$:	1, page 000013 (no dilution was performed on this sample)
$Sample_{WT}$:	5.33g wet wt., page 000006

PCB28 (ng/g) = [(423.7752 * 1.056* 2* 1* 1)/ 5.33]

PCB28 (ng/g) = 167.92 ng/g (page 000439)

CALCULATION OF SURROGATE RECOVERY -- PCB by GC/ECD

Surrogate recoveries are also simply calculated using surrogate internal standard quantitation data obtained directly from the sample quantitation reports (the applicable peak areas and recovery internal standard amounts have already been incorporated into the method/equation for the quantitation report generation, as presented in the "Quantification of Samples" text). The surrogate internal standard determined amount listed on the quantitation report is a direct measure of the amount of surrogate internal standard recovered, using the quantitation method of our data system. The surrogate recoveries are calculated as detailed below:

$$SR = [(RS/Amt_i) \mid Lipid_{SF} * Coplanar_{SF} * PIV_{SF} * Sample_{DF} * 100\%]$$

where,

Amt_i =	Amount surrogate internal standard [e.g., PCB36] added to sample
$Lipid_{SF}$ =	As explained in the "Quantification of Samples" text
$Coplanar_{SF}$ =	As explained in the "Quantification of Samples" text
PIV_{SF} =	As explained in the "Quantification of Samples" text
$Sample_{DF}$ =	As explained in the "Quantification of Samples" text
RS =	Amount surrogate internal standard [e.g., PCB36] determined in sample

The two surrogate internal standards used for surrogate recoveries are PCB36 and PCB112.

There are two recovery internal standards, PCB39 and PCB166. The recovery internal standard PCB39 is used to determine the recovery of the surrogate internal standard PCB36 and the recovery internal standard PCB166 is used to determine the recovery of the surrogate internal standard PCB112.

Example:

Sample ID:	Z5799, pages 000305 through 000307 - quantitation report (from batch 97-126), page 000439 and 440 (97-126 Table)
Data File Number:	pesticides,chanl_01.sa06,21,1, page 000282 (# = 21)
Amt, Surrogate Internal Standard Spiked (ng):	803.2, page 000008 (400 uL EI17 * 2.008 ng/uL)
$Lipid_{SF}$:	1.056, page 000010
$Coplanar_{SF}$:	2, page 000012 (10000uL was initial volume, which was then split, 5000uL to each analysis)
PIV_{SF}:	1, page 000013 (the RIS was added to the entire 5000uL)
$Sample_{DF}$:	1, page 000013 (no dilution was performed on this sample)
RS, Surrogate Internal Standard Amount Determined (ng):	349.2781, page 000305

SR, PCB36 Recovery (%) = [(349.2781/803.2) * 1.056 * 2 * 1 * 1* 100%]

SR, PCB36 Recovery (%) = 92 (page 000440)

Battelle

Ocean Sciences
397 Washington Street
Duxbury, Massachusetts 02332
Telephone (617) 934-0571

August 26, 1997

Mr. Douglas Beltman
Hagler Bailly Consulting, Inc
1881 Ninth Street, #201
Boulder, CO 80302

Subject: Reporting of PCB Data for the Lower Fox River/Green Bay NRDA Project — GC/MS Data

Dear Doug:

Enclosed please find Battelle data packages for tissue sample analyses performed in support of the *Lower Fox River/Green Bay NRDA Project*. These data are for the analyses of 26 tissue samples by GC/MS and the GC/ECD MDL samples, as described in the project-specific Quality Assurance Project Plan, dated May 5, 1997 and your supplemental memorandum dated July 22, 1997 which describes the selection of samples for GC/MS analysis. The samples were analyzed for the determination of polychlorinated biphenyl (PCB) concentrations.

The GC/MS data are reported in two large 3-ring binders. One binder contains the tern egg and fish egg PCB congener data, and the other binder contains the PCB congener data for the whole body walleye, brown trout, and lake trout samples selected for GC/MS analysis. A smaller 3-ring binder is also included in this deliverable. This binder contains the data associated with the GC/ECD MDL determination for the 106 base PCB congeners.

The final data are printed out as summary spreadsheet tables in the "Tables" section of the data packages. Enclosed you will also find (1) one diskette with the Excel spreadsheet files that contain the summary data tables, (2) a table summarizing the calculated GC/ECD MDLs (Attachment 1), (3) a table with representative GC/MS reporting limits (Attachment 2), and (4) example calculations to aid the validator when reviewing the GC/MS data packages (Attachment 3).

A separate Excel spreadsheet has been prepared with transposed GC/MS field sample data (file named "Field Sum ExtendedMS.xls"), per your request and discussions with Tom Gulbransen. All 26 field samples have been pulled together into one table in this file. These data have also been compiled into a single Access data base file that is provided on a separate diskette. There are no hard copies of the transposed Excel table or the Access file because of their large size. Additionally, it should be noted that, per our discussions, the transposed data and the Access file have only received a cursory review, and I strongly recommend that your staff carefully check them against the standard deliverable tables before they are used. The standard summary spreadsheets tables, which are those that are included in the Tables section of the data packages, are the primary deliverable; these tables have all been thoroughly reviewed and validated by our independent QA Unit, as well as by staff of the chemistry department.

Analytical Information

The 26 samples for which GC/MS data are reported are a sub-set of the tissue samples that were processed and analyzed by GC/ECD. The GC/ECD data were reported on August 13, 1997 and that deliverable included the technical procedural information, and other general supporting information that are associated with all of these analyses.

General Quality Control and Other Information

- *MDL Data — GC/ECD.* The MDL sample analyses are compiled in one data package (the smaller of the three 3-ring binders). The MDL study was performed in accordance with the EPA protocol set forth in 40 CFR 136 Appendix B *Method Detection Limit (MDL) Determination*, with seven replicate analyses being used. Summary MDL data tables have been prepared for MDLs calculated the following four different ways: (1) concentrations calculated on a wet weight basis and quantification versus the recovery internal standard (i.e., no surrogate compound correction), (2) concentrations calculated on a wet weight basis and with surrogate compound correction, (3) concentrations calculated on a dry weight basis and quantification versus the recovery internal standard, (4) concentrations calculated on a dry weight basis and with surrogate compound correction. The MDL data based on sample wet weight and without surrogate correction (which is the method used for all samples in this project) are also presented in Attachment 1.

  The hatchery trout fillet that was used for the MDL study had measurable levels of PCB, as did all the hatchery samples analyzed in this project. Unfortunately, this had a significant negative impact on the results of the MDL study because there were higher levels of many PCB congeners in the sample to begin with than was added for the MDL determination. The sample used for an MDL study should ideally not contain any of the target compounds prior to fortification. Although the non-spiked sample matrix was also analyzed non-fortified (and in duplicate), it was not possible to background correct the data because the native concentrations were so high relative to the spiking levels.

  The MDLs were generally in the 0.10 to 0.15 ng/g range for the PCB congeners that were not present in the tissue material to begin with, or present at very low concentrations (Attachment 1) — these PCB congeners best represent the "true" MDLs for the method. These MDLs are consistent with our past experience, which have typically generated wet weight MDLs in the 0.02-0.05 ng/g range when there has been no sample splitting (the MDL samples in this study had a split factor of 2) and when using a sample size of about 25 g (the average sample weight was about 11 g in this study).

  The surrogate recoveries for the MDL samples ranged from 67 to 103%. There were no notable levels of PCB detected in the PB sample, and the PCB congener recoveries were near 100% in the BS sample for almost all target compounds; the apparent over-recovery of PCB41 in the BS (which is qualified with an "X") is due to coelution with coplanar congener #37 which was added to the sample at a significant level. These results indicate that the quality of the sample analyses were in control.

- *RL Data — GC/MS.* Examples of GC/MS reporting limits are tabulated in Attachment 2. Sample-specific RLs were used for qualifying the analytical data, and the RLs that were used for data reporting differ from those presented in Attachment 2 depending on sample-specific PIVs, dilution factors etc. The reporting limits are based on the PCB congener concentration in the low calibrations standard and are calculated as described in detail in the August 13, 1997 deliverable. PCB congeners could typically be confidently determined at concentrations well below the RLs, and uncensored data were reported for this work and qualified with a "J", as appropriate.

- *Quantification and Reporting of Congeners #153 and 132 — GC/MS.* Congeners #153 and #132 could not be separated in the GC/MS analysis of the whole body fish samples (analytical batches 97-190, 97-191, and 97-192),and are therefore reported as PCB153/132, indicating that the value represents the sum of these two congeners. In Hagler Bailly's original scope of work for this project separate data for these congeners was not expected, although Battelle was able to provide discrete data from the GC/ECD analysis. These congeners could be separated during the GC/MS analysis of the egg samples, and separate data are reported for those samples.

- *X Qualifier for Congener #153 in CRM Samples — GC/MS.* The CRM results for congener #153 have been qualified for the whole body fish samples (analytical batches 97-190, 97-191, and 97-192), because of the previously mentioned coelution of congeners #153 and #132. The CRM results for congener #153 are clearly elevated in these three samples, as compared to the CRM data in the two egg batches, which can be attributed to contributions from congener #132.

- *Comparison of GC/MS and GC/ECD Total PCB Data.* The data for the 26 project field samples for which both GC/ECD and GC/MS analysis was performed have been given a cursory review to assess the comparability. The GC/MS data suggest that there was interference with certain congeners in the GC/ECD analysis, although generally the comparability is quite good. The significant interference observed with congener #85 in the GC/ECD analysis (likely p,p'-DDE) was transparent to the GC/MS analysis, and the GC/MS data can be used to obtain more reliable values for congener #85. A comparison of the sum of the PCB congener values from the GC/ECD and GC/MS analyses provide good general comparability information. The average RPD in the sum of the PCB congeners determined by GC/MS and GC/ECD was just under 8% (using the sum of the congeners without congener #85 to represent the GC/ECD analysis). As could be expected, the greatest comparability was observed for the analytical batches where the GC/ECD analyst reported the "cleanest" baselines and minimal matrix contributions (the fish egg and last whole body fish batch — batches 97-129 and 97-192). The sum of the PCB congener concentrations were, on average, only 4 and 3% different, respectively, between the two analytical methods for these batches. Analytical batches with more complex GC/ECD matrix signals had somewhat greater differences in the data; the RPD in the sum of the PCB congeners averaged 11% for the bird egg batch (batch 97-126).

- *Error in Calibration Standard Table in GC/ECD Congener Package.* The spreadsheet table listing the calibration standard concentrations that was included in the GC/ECD data package for the egg samples had a few minor errors. The errors had been detected, the original spreadsheet updated in Battelle's standards records, but had not been updated in this data package. The correct standard concentrations were used in all sample quantification, so no data were affected. I am enclosing these updated pages and an additional copy that highlights where corrections were made. Please replace pages 48 and 49 in the Standard Preparation section of the GC/ECD data package that contains the bird and fish egg data with these two new pages.

Specific Quality Control Information — GC/MS Analysis
Analysis of procedural blanks was the only QC sample analysis that was required for the GC/MS work, as described in the project QAPP. Other QC samples data, and surrogate recovery information, were generated in the GC/ECD analyses. However, Battelle reduced the GC/MS data for the blank spike and certified reference material samples, and those data are included in the enclosed data packages.

- *Procedural (Method) Blanks*. A procedural blank (PB) was processed and analyzed with each of the five sample batches. There was no PCB detected in any of the PBs.

- *Blank Spike Recovery*. A blank spike (BS) sample was processed with each of the five analytical batches. Each of the 527 individual PCB congener recovery data points met the data quality objective, with the majority of the recoveries being in the 80 to 95% range.

- *CRM Recovery*. A certified reference material (CRM) was analyzed with each of the five analytical batches. This material is certified for selected PCB congeners. The CRM data are presented both non-corrected and surrogate corrected. The surrogate correction uses surrogate recoveries that were generated in the GC/ECD analysis because no surrogate recoveries were determined in the GC/MS analysis. It may not be appropriate to apply these GC/ECD surrogate recoveries to the GC/MS data since the target analytes (GC/MS data) and internal standards (GC/ECD data) may be impacted by different levels and types of analyte and matrix effects. The CRM data using non surrogate corrected GC/MS data probably provide the best data assessment, since surrogate recoveries were not determined in this analysis.

  The average PD in the CRM results consistently met the DQO. The individual congener PD values also met the DQOs, even though the measured PCB170/190 concentration was below the primary target DQO (±35 PD for analytes with concentrations >5 times the RL) by 0.2% in one analysis (analytical batch 97-192); one analyte in each sample could be up to 50 PD from the certified value. The measured concentrations were typically 5 to 20 below the certified value, which is consistent with target analyte recoveries in the 80 to 95% range (as was observed for the BS samples).

Please do not hesitate to give me a call at 617-934-0571 if you have any questions at all.

Sincerely,

Gregory S. Durell
Senior Research Scientist

Attachments:

Attachment 1: MDLs for PCB Congeners by GC/ECD
Attachment 2: PCB Analysis Reporting Limits — GC/MS
Attachment 3: Example Data Calculations — GC/MS

Attachment 1

MDLs for PCB Congeners by GC/ECD

PCB Congener	MDL (ng/g, wet weight) *
PCB1	0.85
PCB3	2.42
PCB4/10	0.37
PCB7/9	0.15
PCB6	0.17
PCB8/5	0.23
PCB19	0.13
PCB12/13	0.93
PCB18	0.19
PCB17/15	0.15
PCB24/27	0.17
PCB16/32	0.24
PCB29	0.08
PCB26	0.14
PCB25	0.13
PCB31	0.30
PCB28	0.23
PCB21	0.10
PCB33/20	0.16
PCB53	0.20
PCB51	0.09
PCB22	0.09
PCB45	0.15
PCB46	0.16
PCB52	0.64
PCB43	0.11
PCB49	0.36
PCB47/75	0.27
PCB48	0.09
PCB44	0.19
PCB59	0.06
PCB42/37	0.35
PCB41/64/71	0.64
PCB40	0.18
PCB100	0.14
PCB63	0.21
PCB74	0.32
PCB70/76	0.34
PCB66	0.25
PCB95	0.53
PCB91	0.18
PCB56/60	0.17
PCB92	0.19
PCB84	0.42
PCB89	0.25
PCB101/90	0.95
PCB99	0.67
PCB119	0.15
PCB83	0.20
PCB97	0.42
PCB87/115/81	0.56
PCB85	1.49
PCB136	0.14

PCB Congener	MDL (ng/g, wet weight) [a]
PCB110/77	0.92
PCB82	0.22
PCB151	0.42
PCB135/144	0.29
PCB124	0.12
PCB107/147	0.21
PCB149/123	0.87
PCB118	0.99
PCB134	0.10
PCB114	1.62
PCB131	0.21
PCB146	0.43
PCB153	1.70
PCB132	0.70
PCB105	0.40
PCB141/179	0.22
PCB137	0.12
PCB176	0.14
PCB130	0.18
PCB138/160/163	1.33
PCB158	0.16
PCB129/126	0.14
PCB178	1.77
PCB175	0.12
PCB187/182	0.71
PCB183	0.26
PCB128	0.28
PCB167	0.15
PCB185	0.14
PCB174	0.21
PCB177	0.27
PCB171/202	0.28
PCB156	0.12
PCB173	0.13
PCB201/157	0.18
PCB172	0.12
PCB197	0.11
PCB180	1.49
PCB193	0.13
PCB191	0.13
PCB200	0.11
PCB169	0.80
PCB170/190	0.32
PCB198	0.09
PCB199	0.31
PCB203/196	0.26
PCB189	0.16
PCB195/208	0.28
PCB207	0.12
PCB194	0.28
PCB205	0.12
PCB206	0.42
PCB209	0.25

[a] Wet weight MDL values without surrogate compound correction. Average sample weight was 11.06 g, and the split factor was 2 (i.e., only half the sample was sent to analysis).

Attachment 2

REPORTING LIMITS - Extended PCB Congener Set by GC/MS

Batch ID	97-126	97-129	97-190, 97-191, 97-192	97-191, 97-192	97-192
Matrix	*Tern eggs*	*L. Trout Eggs*	*Walleye Whole*	*B Trout Whole*	*L.Trout Whole*
Pre-Injection Volume (uL)	5000	2000	1000	2000	1000
Lipid Analysis Split Factor	1.00	1.00	1.00	1.00	1.00
Coplanar Analysis Split Factor	2.00	2.00	1.00	1.00	1.00
Sample Dilution Factor	1.00	1.00	1.00	1.00	1.00
Sample Wet Weight (g)	5.00	5.00	5.00	5.00	10.00
Reporting Unit	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight	ng/g, wet weight
Analyte					
PCB1	102.6	41.0	10.3	20.5	5.1
PCB3	201.0	80.4	20.1	40.2	10.1
PCB4	102.8	41.1	10.3	20.6	5.1
PCB7	64.2	25.7	6.4	12.8	3.2
PCB6	64.0	25.6	6.4	12.8	3.2
PCB8	66.8	26.7	6.7	13.4	3.3
PCB19	36.0	14.4	3.6	7.2	1.8
PCB12	64.0	25.6	6.4	12.8	3.2
PCB18	33.4	13.4	3.3	6.7	1.7
PCB17	28.4	11.4	2.8	5.7	1.4
PCB24	36.0	14.4	3.6	7.2	1.8
PCB16	36.0	14.4	3.6	7.2	1.8
PCB29	36.0	14.4	3.6	7.2	1.8
PCB26	36.0	14.4	3.6	7.2	1.8
PCB25	35.8	14.3	3.6	7.2	1.8
PCB31	36.0	14.4	3.6	7.2	1.8
PCB28	33.2	13.3	3.3	6.6	1.7
PCB21	36.0	14.4	3.6	7.2	1.8
PCB33	36.0	14.4	3.6	7.2	1.8
PCB53	38.6	15.4	3.9	7.7	1.9
PCB51	24.4	9.8	2.4	4.9	1.2
PCB22	36.0	14.4	3.6	7.2	1.8
PCB45	30.8	12.3	3.1	6.2	1.5
PCB46	38.6	15.4	3.9	7.7	1.9
PCB52	33.4	13.4	3.3	6.7	1.7
PCB43	36.0	14.4	3.6	7.2	1.8
PCB49	38.4	15.4	3.8	7.7	1.9
PCB47	38.6	15.4	3.9	7.7	1.9
PCB48	38.6	15.4	3.9	7.7	1.9
PCB44	33.4	13.4	3.3	6.7	1.7
PCB59	38.4	15.4	3.8	7.7	1.9
PCB42	36.0	14.4	3.6	7.2	1.8
PCB41	29.6	11.8	3.0	5.9	1.5
PCB40	36.0	14.4	3.6	7.2	1.8
PCB100	38.6	15.4	3.9	7.7	1.9
PCB63	38.4	15.4	3.8	7.7	1.9
PCB74	38.6	15.4	3.9	7.7	1.9
PCB70	38.6	15.4	3.9	7.7	1.9
PCB66	33.4	13.4	3.3	6.7	1.7
PCB95	38.6	15.4	3.9	7.7	1.9
PCB91	29.6	11.8	3.0	5.9	1.5
PCB56	30.8	12.3	3.1	6.2	1.5
PCB92	27.0	10.8	2.7	5.4	1.4
PCB84	30.8	12.3	3.1	6.2	1.5
PCB89	38.4	15.4	3.8	7.7	1.9
PCB101	33.4	13.4	3.3	6.7	1.7
PCB99	38.4	15.4	3.8	7.7	1.9
PCB119	38.6	15.4	3.9	7.7	1.9
PCB83	38.4	15.4	3.8	7.7	1.9

REPORTING LIMITS - Extended PCB Congener Sel by GC/MS

Batch ID	97-126	97-126	97-190, 97-191, 97-192	97-191, 97-192	97-192
Matrix	*Tern eggs*	*L. Trout Eggs*	*Walleye Whole*	*B Trout Whole*	*L.Trout Whole*
PCB97	38.6	15.4	3.9	7.7	1.9
PCB87	38.6	15.4	3.9	7.7	1.9
PCB85	29.4	11.8	2.9	5.9	1.5
PCB136	38.6	15.4	3.9	7.7	1.9
PCB110	38.6	15.4	3.9	7.7	1.9
PCB82	38.6	15.4	3.9	7.7	1.9
PCB151	38.6	15.4	3.9	7.7	1.9
PCB135	28.2	11.3	2.8	5.6	1.4
PCB124	38.6	15.4	3.9	7.7	1.9
PCB107	38.6	15.4	3.9	7.7	1.9
PCB149	38.4	15.4	3.8	7.7	1.9
PCB118	33.4	13.4	3.3	6.7	1.7
PCB134	38.4	15.4	3.8	7.7	1.9
PCB114	38.6	15.4	3.9	7.7	1.9
PCB131	34.6	13.8	3.5	6.9	1.7
PCB146	28.4	11.4	2.8	5.7	1.4
PCB153	33.2	13.3	3.3	6.6	1.7
PCB132	38.6	15.4	3.9	7.7	1.9
PCB105	33.4	13.4	3.3	6.7	1.7
PCB141	38.6	15.4	3.9	7.7	1.9
PCB137	38.4	15.4	3.8	7.7	1.9
PCB178	30.8	12.3	3.1	6.2	1.5
PCB130	30.8	12.3	3.1	6.2	1.5
PCB138	33.2	13.3	3.3	6.6	1.7
PCB158	38.6	15.4	3.9	7.7	1.8
PCB129	38.6	15.4	3.9	7.7	1.9
PCB178	27.0	10.8	2.7	5.4	1.4
PCB175	38.6	15.4	3.9	7.7	1.9
PCB187	33.4	13.4	3.3	6.7	1.7
PCB183	33.4	13.4	3.3	6.7	1.7
PCB128	33.4	13.4	3.3	6.7	1.7
PCB167	38.6	15.4	3.9	7.7	1.9
PCB185	38.6	15.4	3.9	7.7	1.9
PCB174	24.4	9.8	2.4	4.9	1.2
PCB177	23.0	9.2	2.3	4.6	1.2
PCB171	38.5	15.4	3.9	7.7	1.9
PCB156	38.6	15.4	3.9	7.7	1.9
PCB173	38.4	15.4	3.8	7.7	1.9
PCB201	38.4	15.4	3.8	7.7	1.9
PCB172	38.4	15.4	3.8	7.7	1.9
PCB197	38.4	15.4	3.8	7.7	1.9
PCB180	33.4	13.4	3.3	6.7	1.7
PCB193	38.4	15.4	3.8	7.7	1.9
PCB191	38.6	15.4	3.9	7.7	1.9
PCB200	38.6	15.4	3.9	7.7	1.9
PCB169	38.6	15.4	3.9	7.7	1.9
PCB170	33.4	13.4	3.3	6.7	1.7
PCB199	38.4	15.4	3.8	7.7	1.9
PCB198	32.0	12.8	3.2	6.4	1.6
PCB203	30.8	12.3	3.1	6.2	1.5
PCB189	38.4	15.4	3.8	7.7	1.9
PCB195	33.4	13.4	3.3	6.7	1.7
PCB207	30.8	12.3	3.1	6.2	1.5
PCB194	38.4	15.4	3.8	7.7	1.9
PCB205	38.6	15.4	3.9	7.7	1.9
PCB206	25.6	10.2	2.6	5.1	1.3
PCB209	25.6	10.2	2.6	5.1	1.3

Attachment 3

QUANTIFICATION OF SAMPLES – PCB by GC/MS

Samples are quantified using the method of internal standards. The quantification internal standard is the recovery internal standards (added to the sample immediately prior to instrumental analysis). The concentration of target analytes is determined using the following equation:

$$[PCB] = [(A_t/A_i) \times (Amt_i/RF_t) \times Lipid_{SF} \times Coplanar_{SF} \times Sample_{DF} \times (1/Sample_{WT})]$$

where,

[PCB]	=	Concentration target PCB analyte
A_t	=	Area quantification ion for target analyte (e.g., PCB18)
A_i	=	Area quantification ion for internal standard (PCB34)
Amt_i	=	Amount internal standard (PCB34) added to sample
Rf_t	=	Average RF for analyte (e.g., PCB18) determined from initial calibration
$Lipid_{SF}$	=	Lipid analysis subsampling factor. Factor that corrects for the amount removed in the lipid analysis. (total extract volume before lipid analysis) × (1/(total extract volume before lipid analysis – extract volume removed for lipid analysis))
$Coplanar_{SF}$	=	Coplanar analysis split factor. Factor that corrects for any splitting of the extract for coplanar PCB analysis. (total extract volume before split / extract volume removed for the subject analysis)
$Sample_{DF}$	=	Sample dilution factor. Factor that corrects for any subsampling of the extract for dilution purposes (i.e., when samples were diluted and re-analyzed). This is only a factor when a portion of the extract is removed, and subsequently spiked with additional RIS. The amount of solvent added to perform the dilution is not a factor in the calculation. Additionally, this is ***not*** a factor if only the PIV is increased to bring the analyte response within the calibration range in a re-analysis. (total extract volume before subsampling / extract volume removed for the subject dilution and re-analysis)
$Sample_{WT}$	=	Sample weight (g, wet weight). Weight of the sample amount that was extracted.

One internal standard was used for quantification (PCB34).

QUANTIFICATION OF SAMPLES – PCB by GC/MS (Continued)

The page references listed below for the example calculations are for the bird and fish egg GC/MS data package (analytical batches 97-126 and 97-129).

Example Calculation:

Sample ID:	VA89CRM, page 000208 - 210, SQB347 pg 000107
Data File Number:	B0708.d, page 000208 - 210
A, Target Analyte:	PCB128, page 000210
I, Internal Standard:	PCB34, page 000208
A_x, Target Analyte Area:	1178, page 000210
A_{is} Internal Standard Area:	11427, page 000208
Amt_{is} Internal Standard Spike Amt (ng):	235, page 000208 and page 000076 (100 μL of standard EI19 × 2.35 ng/μL)
Rf_{is} Average RF of PCB128:	0.56684, page 000113
$Lipid_{SF}$:	1.058, page 000073
$Coplanar_{SF}$:	2.0, page 000076
$Sample_{DF}$:	1.0, page 000076
$Sample_{WT}$:	5.03, page 000069

PCB128 Conc. (ng/g) = [(1178/14427) × (235/0.56684) × (1.058) × (2) × (1) × (1/5.03)]

PCB128 Conc. (ng/g) = 14.24 ng/g (page189)

Note: Please review the information in the Miscellaneous Documentation section of the data package before beginning to audit and review data; this section may contain additional information that are important to the calculation of sample analyte concentrations.

Battelle

Ocean Sciences
397 Washington Street
Duxbury, Massachusetts 02332
Telephone (617) 934-0571

December 5, 1997

Mr. Douglas Beltman
Hagler Bailly Consulting, Inc.
1881 Ninth Street, #201
Boulder, CO 80302

Subject: Reporting of PCB Data for the Lower Fox River/Green Bay NRDA Project — GC/ECD Data from Re-Quantified and Re-Analyzed Samples

Dear Doug:

Enclosed please find Battelle's data package for the *Lower Fox River/Green Bay NRDA Project* tissue sample analyses recently performed at Battelle Duxbury Operations. The samples were analyzed for the determination of polychlorinated biphenyl (PCB) concentrations. These data are from (1) the re-quantification of a set of samples that were originally quantified with a different calibration type, and (2) the re-extraction and re-analysis of a set of samples that had low surrogate recoveries in the original analysis. The original data for these samples, the data which you may wish to replace with these new data, were submitted as part of the large data delivery on August 13, 1997.

The GC/ECD data are reported in two large 3-ring binders, with the appropriate section dividers and tabs indicating the location of different data and information. The final data have been printed out as summary spreadsheet tables in the "Tables" section of the data package. Enclosed you will also find (1) one diskette with the Excel spreadsheet files that contain the summary data tables, (2) a table listing the samples that were re-quantified with the edited calibration method (Attachment 1), (3) a table listing the samples that were re-extracted and re-analyzed, and the types of analyses that was performed on them (Attachment 2), and (4) results of the lipid determination method comparison (Attachment 3). Enclosed is also a replacement page for one of the sample homogenization forms that were submitted earlier — the Battelle sample ID has been corrected for two samples (the correct ID was used in all sample preparation documentation and data deliveries).

Other relevant information such as the technical procedures, listing of target PCB analytes, data quality objectives, data qualifiers, reporting limits, method detection limits, example calculations, chain-of-custody documentation, etc. have been provided with previous data deliverables.

A sheet with transposed *field* sample data has been added to the two Excel spreadsheet files that contain field sample data (97-191 Requant_a.xls and Re-extracts_a.xls). The transposed data are in a format that can easily be accepted by data bases, and they have been compiled into an Access file (with three data tables). There are no hard copies of the transposed Excel tables or the Access file. Additionally, it should be noted that, per our discussions, the transposed data and the Access file have only received a cursory review, and I strongly recommend that your staff carefully check them against the standard deliverable tables before they are used. The standard summary spreadsheets tables, which are those included in the Tables section of the data package, are the primary deliverable; these tables have been thoroughly reviewed and validated by our QA Unit, as well as by staff of the chemistry department.

Analytical Information

Re-Quantified Samples. Re-quantified extended PCB congener data are submitted for 21 samples (Attachment 1) for which data were already submitted back on August 13, 1997. The original data for these samples were inadvertently generated using a 1/X weighted quadratic equation, and the calibrations were therefore re-generated using an non-weighted method to be consistent with all the other data; non-weighted calibrations are also more standard. Most of the affected samples were QC samples because a large number of the field samples in those analytical batches were diluted and re-analyzed, and the re-analyses were quantified with a non-weighted calibration.

Re-Analyzed Samples. Three batches of re-extractions and re-analyses are reported in this data delivery (Attachment 2). These samples were re-analyzed in the laboratory because the recoveries were lower than desirable in the original analyses. Several samples from the first re-extraction batch (97-274) were actually re-analyzed a second time in one of the other two batches because the recoveries were still low. Additionally, the field samples in the second and third re-analysis batches were processed in duplicate to obtain the best data. Both replicates were reported if the surrogate recoveries were good for both analyses, and the better of the two was reported if one or both of the replicates yielded surrogate recoveries outside the data quality objective range.

High quality data were generated for most samples, but a few surrogate recoveries remained below the data quality objective, even though they were separately analyzed up to four (4) times, indicating unique sample matrix characteristics. However, considering the large numbers of samples analyzed and reported the overall quality of the project data set is very high and only three analyses (out of 175 separate PCB sample analyses) remain with recovery results below the data quality objectives.

General Quality Control and Other Information

Several of the general reporting items listed in this section have already been communicated to Hagler Bailly, and are included here for completeness.

- *Re-Quantification*. The re-quantification of the samples listed in Attachment 1 yielded only slightly different data than what was submitted on August 13, 1997. For instance, samples VD38 (BTEG01CP) and VD40 (BTEG04CP) are now reported to have a "Sum of PCB w/o PCB85" concentration of 1,900 and 1,707 ng/g, wet weight, versus the original results of 1,955 and 1,752 ng/g, respectively. The new data for these samples are approximately 2–3% lower than the original results.

- *JX Qualifier for Congener #85*. There was significant coelution/interference with congener #85 in the field samples that appears to be caused by the presence of p,p'-DDE. Therefore, the congener #85 data have been qualified with the qualifier "JX" when this peak is clearly significantly higher than could reasonably be expected. The size of this peak was not considered when deciding on dilutions and re-analysis of samples (i.e., this peak was frequently above the high calibration standard and was often above the range of the detector, and ignored for dilution purposes). The data reported for congener #85 are not accurate, and, therefore, in addition to providing a sum of all PCB concentrations in the Excel summary tables, we are also providing a sum of all congeners with congener #85 excluded. The congener summation without congener #85 is likely a more accurate measure of the total PCB than the sum that includes congener #85.

For your information, congener #85 constitutes approximately 1% of the total PCB in mid-molecular weight Aroclor formulations, such as Aroclors 1248 and 1254 (Schultz *et al.*, 1989, *Environ. Sci. Technol.* 23, 852–859; and Battelle internal determinations). It is only reasonable to expect a similar contribution in environmental samples for this particular congener. The GC/MS data will provide more accurate congener #85 concentration data. The GC/MS data will also provide information on the relative ratio of congener #85 to other congeners that are not interfered with in the GC/ECD analysis, thus providing data that can be used to obtain a good estimate of the congener #85 concentrations in all samples.

- *X Qualifier for Congener #169*. There appeared to be a procedural contaminant in the coplanar PCB congener method that resulted in a doublet peak that interfered with congener #169; the two interfering peaks elute on either side of the congener #169 peak. If a peak was clearly present in the valley between the two contaminant it was picked as congener #169, but the contaminant most likely masked the presence of this congener under most circumstances or reduced the accuracy of any quantification of this congener when detected. The qualifier "X" was added to the congener #169 data in the coplanar analysis (whether it was detected or not) to indicate this issue.

  Battelle followed EPA Method 1668 for the coplanar PCB congener separation, and it is unclear what reagent or other component of the method contributed this interference. Congener #169 was also determined in the standard PCB congener analysis (it is the only coplanar congener that can be well resolve in that analysis), and there was no evidence of procedural interference with congener #169 in those analyses.

- *CRM Quantification*. The quality control results for the Certified Reference Material (CARP-1 CRM) were calculated and reported ***both*** surrogate corrected and not surrogate corrected; separate spreadsheet tables have been prepared. The reason is that the certified values for this CRM are based on surrogate corrected quantification (per information from the National Research Council (NRC) Canada scientists who prepared and certified this material), and surrogate correction may therefore be the most valid approach for performing data comparisons.

- *Quantification of Congener #63*. There was an error in the concentration used for congener #63 in the calibration method for the second level of the multilevel calibration, and this was discovered after the samples had been quantified (0.0196 ng/µL was entered/used rather than the correct value of 0.0192). This minor error was for one analyte in one calibration level. A field sample was requantified with the correct concentration in the calibration method to assess the impact of this error. The two methods yielded result of 23.1886 and 23.1072 ng (<0.4% difference for congener #63, with the reported value being the higher of the two) and this relatively minor discrepancy for one congener was considered so small that it did not warrant re-quantification of the data set.

- *Lipid Content Method Comparison*. Lipid content determination was performed with two different extraction solvents (hexane and dichloromethane) on seven brown trout whole body samples and seven walleye whole body samples, to assess differences caused by the two solvents. Additionally, triplicate analysis was performed on one sample of each fish type. The results from this determination are summarized in Attachment 3.

  As expected, the dichloromethane extraction method yielded higher lipid content values than the hexane extraction. The lipid content was, on average, about 43% higher with the dichloromethane method for brown trout and about 25% higher for walleye. However, these data need to be considered carefully

before they are used to generate some generic method-to-method lipid content correction factor because there is clearly significant fish-to-fish variability. The difference (percent difference) between the two methods was as low as 17% and as high as 72% for different brown trout samples, with the rest ranging from 39% to 50%. This notable fish-to-fish variability could be the result of slightly different lipid composition of different fish (i.e., the fish sample matrix). Additionally, and possibly even more importantly, variability in the *moisture* content of the fish impacts the variability in the lipid data when calculated on a wet weight basis; the lipid are primarily associated with the "dry" matrix, not the water. After normalizing the data for moisture content (i.e., calculating lipid content on a *dry*, not wet, weight basis) it is likely that the PD values will decrease. The precision in the triplicate analyses of the same sample is quite good, indicating that the observed variability is not due to the method.

Specific Quality Control Information — Re-Analyzed Samples

- *Procedural (Method) Blanks*. A procedural blank (PB) was processed and analyzed with each of the sample batches. Few congeners were detected in the PB samples in the extended PCB congener analysis, and those were consistently at very low levels — well below the reporting limits. In the coplanar PCB congener analyses there was interference with congener #169 (as discussed earlier) that contributed to a low-level signal in the PB, but none that suggested the presence of this congener. There was also a low-level signal corresponding to congener #81 in one of the three coplanar PCB congener PBs, but it too represented a concentration much below the reporting limits.

- *Blank Spike Recovery*. A blank spike (BS) sample was processed with each of the analytical batches. All extended congener target compound recoveries were acceptable for the BS sample. The BS recoveries were acceptable for the three coplanar PCB congener batches, with the exception of a slightly elevated recovery for congener #126 (135% recovery) in the BS processed with batch 97-306 and a slightly low recovery for congener #37 (48% recovery) in the BS with batch 97-312.

- *CRM Recovery*. A certified reference material (CRM) was analyzed with each of the analytical batches. This material is certified for selected "standard" PCB congeners (i.e., not for any coplanar congeners). For the coplanar PCB congener analyses the CRM was used only to track precision over several batches. CRM results are reported both non-corrected and surrogate corrected. The surrogate corrected results best represents the true native sample concentration and should be used for comparing with CRM certification values; surrogate corrected data were used by National Research Council (NRC) Canada when establishing the reference values.

  The average PD in the CRM results met the DQO, and there was only one individual congener exceedance; 43 %PD for PCB66/95, versus a DQO of ±35%. However, the CRM is not certified for PCB66/95 and a less rigorous "consensus" value is used to evaluate this parameter. The precision in the coplanar PCB analysis of the CRM was relatively good for congeners #77 and #126 (e.g., 32% RSD and 25% RSD, respectively, for the non-corrected data), considering the low concentrations of these congeners in the CRM (near or below the RL).

- *Duplicate (DUP) Precision*. Specific duplicate precision tables were not generated for all of the re-analyses because so many sets of replicate analyses were performed that no sample needed to be specifically designated as the DUP sample of the batch. A large number of replicate field samples are available to calculate analytical precision, and the template to "drop" such data in to are available as a separate sheet in the provided Excel files.

- *Surrogate Recovery*. Surrogate internal standard (SIS) compounds were added to every field and QC sample to monitor sample processing efficiency, and the recoveries of two SISs (congeners PCB36 and PCB112) were determined for each sample. Additionally, the coplanar PCB congener column separation efficiency was monitored using deuterated congener #77 for all samples that were subjected to coplanar PCB congener analysis.

  The surrogate recoveries were generally very good. The surrogate recovery DQOs (50 to 125% recovery) were met for all QC samples and almost all field samples. As discussed earlier, a few field samples had surrogate recoveries that did not meet the DQOs even though they were separately analyzed up to four (4) times. This clearly indicates a unique sample matrix for these samples — a matrix that cannot be effectively extracted using standard laboratory procedures. However, considering the large numbers of samples that were analyzed in this project it is clear that the overall quality of the data set is very high and only three analyses remain with recovery results outside the DQO range. These three samples are all lake trout whole body samples: (1) the coplanar PCB congener data for sample Z6899 (EGLMF06WC-1) is based on a sample with low standard congener surrogate recoveries, (2) the coplanar PCB congener data for sample Z6833 (EGLMF10WC-1) is based on a sample with a low coplanar congener surrogate recovery, and (3) the extended PCB congener data for sample Z6834 (EGLMF12WC-1) is based on a sample with low standard congener surrogate recoveries. The low recoveries for these three ranged from 19% to 28%.

Please do not hesitate to give me a call at 781-934-0571 if you have any questions at all.

Sincerely,

Gregory S. Durell
Senior Research Scientist

Attachments:

Attachment 1: Re-Quantified Samples
Attachment 2: Re-Analyzed Samples
Attachment 3: Lipid Method Comparison

Attachment 1

Re-Quantified Samples [a]

Client ID	Battelle ID	Batch ID
NA	VD26PB	97-190
NA	VD27BS	97-190
NA	VD28CC	97-190
NA	VD29IB	97-190
NA	VD30EB	97-190
NA	VD32PB	97-191
NA	VD33BS	97-191
NA	VD34CC	97-191
NA	VD35IB	97-191
NA	VD36EB	97-191
BTEG01CP	VD38	97-191
BTEG04CP	VD40	97-191
BTUG01CP	VD42	97-191
BTUG02CP	VD43	97-191
BTUG04CP	VD44	97-191
BTUG05CP	VD45	97-191
NA	VD48PB	97-192
NA	VD49BS	97-192
NA	VD51CC	97-192
NA	VD52EB	97-192
NA	VD53IB	97-192

[a] Re-quantified by non-weighted quadratic calibration type because they were initially quantified with a 1/x weighted quadratic calibration. Target extended PCB congener data only (surrogate recoveries were not affected because they were not calibrated by the weighted method in the original analysis).

Attachment 2

Re-Analyzed Samples [a]

Client Reporting ID	Battelle Sample ID [b]	Re-Analysis Batch ID	Original Batch ID	Congener Analysis Type [c]	Sample Type/Matrix
TEKIB18	Z5799	97-274	97-126	CP	Tern egg
96KICT05	Z5807	97-274	97-126	CP	Tern egg
96KICT07	Z5813	97-274	97-126	CP	Tern egg
96KICT09	Z5815	97-274	97-126	CP	Tern egg
WEFR07CP	VC59	97-274	97-192	CP	Walleye whole
WELG06CP	VC65	97-274	97-192	CP	Walleye whole
BTEG02CP	VD47	97-274	97-192	CP	B. trout whole body
EGLMF06WC-1	Z6899	97-274	97-192	CP	L. trout whole body
TEKIB48	Z5804	97-306	97-126	CP	Tern egg
96KICT03	Z5811	97-306	97-126	CP	Tern egg
96KICT10	Z5816	97-306	97-126	CP	Tern egg
WEWG04CP	VC69	97-306	97-192	CP	Walleye whole
EGLMF11WC-1	Z6897	97-306	97-192	CP	L. trout whole body
EGLMF07WC-1	Z6898	97-306	97-192	CP	L. trout whole body
EGLMF01FC-1	Z5958	97-306	97-129	STD	L. trout eggs
EGLMF08FC-1	Z5965	97-306	97-129	STD	L. trout eggs
EGLMF10WC-1	Z6833	97-306	97-192	CP + STD	L. trout whole body
EGLMF09WC-1	Z6901	97-306	97-192	CP + STD	L. trout whole body
EGLMF01WC-1	Z6902	97-312	97-192	CP	L. trout whole body
EGLMF02WC-1	Z6900	97-312	97-192	CP	L. trout whole body
EGLMF03WC-1	Z6881	97-312	97-192	CP	L. trout whole body
EGLMF04WC-1	Z6880	97-312	97-192	CP	L. trout whole body
EGLMF05WC-1	Z6879	97-312	97-192	CP	L. trout whole body
EGLMF08WC-1	Z6835	97-312	97-192	CP	L. trout whole body
EGLMF12WC-1	Z6834	97-312	97-192	CP	L. trout whole body
BTUG03CP	VD46	97-312	97-192	CP	B. trout whole body

[a] The listed field samples were re-extracted/re-analyzed with new QC samples (PB, BS, CRM, and DUP).
[b] The Battelle sample ID for the re-extracted and re-reported analyses has a –1 or –2 suffix as part of the ID to indicate if it is the first or second re-extraction/re-analysis of the sample. Additionally, all samples in batches 97-306 and 97-312 were re-analyzed in duplicate and the DUP designation has then also been added to the base Battelle ID for the sample tracking and data reporting (e.g., Z6901-2DUP). Both replicates were reported if both had surrogate recoveries that were well within the data quality objectives (DQOs). The sample with the better surrogate recoveries was reported if the recoveries were outside the DQOs for one or both replicates. Data for both replicates have been reported for Z5811, Z5816, VC69, Z6898, Z5958, Z5965, Z6901, Z6881, and VD46.
[c] Congener analysis type: CP; coplanar congeners. STD: standard extended list congeners.

Attachm 3

LIPID METHOD COMPARISON - Hexane vs Dichloromethane

Client Reporting ID	Matrix	Battelle ID	Lipid Content (%, wet weight) Hexane as Solvent	DCM as Solvent	PD
BTEG01CP	Brown trout whole body	VD38	7.87	11.27	43.2
BTEG03CP	Brown trout whole body	VD39	8.12	12.16	49.8
BTEG04CP	Brown trout whole body	VD40	9.82	13.81	40.6
BTEG05CP	Brown trout whole body	VD41	11.74	13.75	17.1
BTUG01CP	Brown trout whole body	VD42	8.28	14.25	72.1
BTUG03CP	Brown trout whole body	VD46	10.75	15.38	43.1
BTEG02CP	Brown trout whole body	VD47	10.33	14.33	38.7
				Average:	43.5
				%RSD:	37.3
BTEG04CP	Brown trout whole body	VD47	10.33	14.33	
BTEG04CP	Brown trout whole body	VD47-DUP	9.46	13.27	
BTEG04CP	Brown trout whole body	VD47-TRIP	11.44	13.95	
		Average:	10.4	13.9	
		%RSD:	9.5	3.9	
WEFR04CP	Walleye whole body	VA57	8.94	11.42	27.7
WEFR01CP	Walleye whole body	VC53	8.59	9.58	11.5
WEFR02CP	Walleye whole body	VC54	14.56	17.19	18.1
WEFR03CP	Walleye whole body	VC55	9.63	10.79	12.0
WEFR05CP	Walleye whole body	VC57	13.52	16.72	23.7
WEFR06CP	Walleye whole body	VC58	11.81	16.90	43.1
WEFR07CP	Walleye whole body	VC59	12.59	17.84	41.7
				Average:	25.4
				%RSD:	51.1
WEFR07CP	Walleye whole body	VC59	12.59	17.84	
WEFR07CP	Walleye whole body	VC59-DUP	14.38	15.66	
WEFR07CP	Walleye whole body	VC59-TRIP	15.97	12.39	
		Average:	14.3	15.3	
		%RSD:	11.8	17.9	

Sample Homogenization

I R
7/28/97 RLM

Project Name: Lower Fox River/Green Bay NRDA
Project Number: G003264
Homogenization Completed by: RLM — **Date:** 7/7/97
Homogenization method/equipment: Hobart Grinder
Storage Location Removed from: ERZ 18 — **Date/Time:** 7/7/97 8:00AM
Storage Location until homogenization/compositing: chemlab
Storage Location Returned to: F1220 — **Date/Time:** 7/7/97 5:00PM

Sample Matrix	Sample #	Field Sample ID *	Battelle ID (log-in) *
Lake trout whole body	1	EGLMF01WC-1	Z6902
Lake trout whole body	2	EGLMF02WC-1	Z6900
Lake trout whole body	3	EGLMF03WC-1	Z6881
Lake trout whole body	4	EGLMF04WC-1	Z6882 80
Lake trout whole body	5	EGLMF05WC-1	Z6883 79
Lake trout whole body	6	EGLMF06WC-1	Z6899
Lake trout whole body	7	EGLMF07WC-1	Z6898
Lake trout whole body	8	EGLMF08WC-1	Z6835
Lake trout whole body	9	EGLMF09WC-1	Z6901
Lake trout whole body	10	EGLMF10WC-1	Z6833
Lake trout whole body	11	EGLMF11WC-1	Z6897
Lake trout whole body	12	EGLMF12WC-1	Z6834

XIO 11/24/97 RLM

* The client Field Sample ID is the same as the Client Reporting ID for samples that are not composited, as outlined in Attachment 2 of the Project Laboratory QAPP. Similarly, the Battelle ID given at log-in is the same as the Battelle Reporting ID for samples that are not composited, such as those listed above.

DATA VALIDATION REPORT

PCB Analysis of Biota Tissues
Green Bay Natural Resource Damage Assessment

Prepared for:

Hagler Bailly
1881 9th Street, Suite 201
Boulder, CO 80302

Prepared by:

EcoChem, Inc.
405 Westland Building
100 South King Street
Seattle, Washington 98104-2885

EcoChem Project No.: C9309-3

April 10, 1998

Approved for Release:

Bob A. Olsiewski
Senior Chemist
EcoChem, Inc.

Introduction

This report summarizes the quality assurance evaluations performed and data qualifications recommended for 123 tissue samples analyzed for the Green Bay Natural Resource Damage Assessment project. Refer to the Sample Index (TABLE 1) for sample identifications and analyses.

The tissue samples were analyzed for 106 PCB congeners or seven Aroclor formations using the Battelle laboratory standard operating procedure, *Identification and Quantitation of Polychlorinated Biphenyls (by Congener and Aroclor) and Chlorinated Pesticides by Gas Chromatography/Electron Capture Detection.* Several samples that were analyzed for the standard congener list were also analyzed for five coplanar PCB congeners. A subset of 26 samples that were analyzed for the standard congener list were also analyzed by GC/MS. The analyses were performed by Battelle Ocean Sciences, 397 Washington Street, Duxbury, Massachusetts.

The surrogate percent recoveries for many of the samples were not within the acceptance limits in the initial analysis. Additionally, two sample extracts were spilled during the extraction process. For these two reasons, four samples for the standard congener analyses and 24 samples for the coplanar congener analyses were re-extracted and reanalyzed. The original results were qualified as do-not-report (DNR); the results from the re-extracted analyses should be used.

The primary data validation review was performed by Sherri Wunderlich and secondary technical review was performed by Alison Bodkin. The data validation review was based on the quality control criteria specified in the analytical methods and the data quality objectives listed in the QAPP.

Data validation and reasons for qualification are summarized in each section of the following report. Validation qualifier definitions and reason codes are listed in TABLE 2 AND TABLE 3, respectively. All data validation qualifiers appear in the database.

FULL DATA VALIDATION REPORT
PCB Analyses
Batches: 97-124, 97-126, 97-127, 97-128, 97-129, 97-181, 97-190, 97-191, 97-192, 97-274, 97-306, and 97-312

I. Data Package Completeness: ACCEPTABLE/With the following discussion.

All necessary documentation for the full validation were provided by the laboratory.

The chain-of-custody (COC) forms for six samples (96-KI-CT-01, 96-KI-CT-03, 96-KI-CT-05, 96-KI-CT-07, 96-KI-CT-09, and 96-KI-CT-10) did not list sampling dates.

For ten samples, EG-LM-F-01-FC-1 through EG-LM-F-10-FC-1, the collection date listed on the COC forms was 10/22/95. The laboratory indicated that the collection date listed on the sample bottles was 10/22/96. Since the COC forms were signed by the sampler on 10/22/96, and the COC collection date for the other two samples in the batch (Samples EG-LM-F-11-FC-1 and EG-LM-F-12-FC-1) was 10/22/96, the actual collection date was most likely 10/22/96.

Although, several internal sample custody seals were broken when received by the laboratory, all cooler seals were intact. This was probably caused by the pressure of the ice, as paper custody seals and tape can be weakened by the cold and moisture. No action was taken.

II. Sample Holding Times and Handling Conditions: ACCEPTABLE/With the following exceptions.

Qualified Data: See the Qualified Sample Results.

Discussion:

All tissue samples were stored frozen at -20 °C or below until the time of extraction. All samples were extracted within one year of the sampling date.

The analysis holding time criterion for PCBs is 40 days from extraction date to date of analysis. All samples were analyzed within the required holding time.

Batch 97-192:

Sample BTUGO3CP: During the extraction, approximately 30mL out of 200mL (or 15% of the sample extract volume) was spilled. As internal standardization was used to quantify the PCB congener concentrations, and as the standard extended list surrogate recoveries for this sample were acceptable, no qualification was performed based on the spillage.

Sample EG-LM-F-09-WC-1: During the preparation step, the vial containing the extract broke in the centrifuge. The sample was pipetted out of the rotor and put in a new vial. The laboratory re-

extracted and reanalyzed the sample by GC/ECD upon request. The reanalysis was performed with Batch 97-306. The original results were qualified as do-not-report (DNR-14); the results from the reanalysis should be used instead.

III. Calibration: ACCEPTABLE/With the following exceptions.

Qualified Data: See the Qualified Sample Results.

Discussion:

Initial Calibrations

For the GC/ECD congener and Aroclor initial calibrations, all reported coefficients of determination for the initial calibrations were greater than or equal to 0.9900. (Therefore, correlation coefficients were greater than or equal to 0.9950.) The laboratory incorrectly calculated calibration curve coefficients using 1/X weighted values for three congener Aroclor initial calibrations (that were analyzed on 7/15/97, 7/22/97 and 8/8/97). The laboratory submitted corrections for the 7/15/97 and 7/22/97 initial calibrations and recalculated all associated sample results. For example, for Sample BTEG01CP the PCB sum (without PCB 85) was originally reported as 1955 ng/g versus a new total of 1900 ng/g; this represents a percent difference of less than 3%. The 8/8/97 initial calibration was only associated with the method detection limit study. As the weighted results were only slightly different than the non-weighted results, the method detection limit study was judged as not significantly affected.

For the GC/MS initial calibrations, all percent relative standard deviation (%RSD) values were less than the 35% upper control limit. All relative response factor values were greater than the 0.050 lower control limit.

Continuing Calibrations (CCVs)

Several percent difference (%D) results (from the true values) for target analytes were outside the individual compound control limit of ±25%. Positive sample results that were associated with non-compliant %D values were qualified as estimated (J-5B). Non-detect results were judged to be not significantly affected. Qualified results are summarized in **TABLE 4.**

Several samples were not analyzed within 12 hours of the beginning CCV. Since all samples were bracketed by acceptable beginning and ending CCVs, no action was taken.

IV. Blank Analyses: ACCEPTABLE/With the following exceptions.

Qualified Data: See the Qualified Sample Results.

Discussion:

Several PCB congeners were detected at low levels by the GC/ECD in some of the procedural, instrument, and equipment blanks. Action levels were established at five times the reported blank

concentrations. Associated positive sample results less than the action levels were qualified as not detected (U-7). Qualified results are summarized in **TABLE 5**.

V. Surrogate Recovery: ACCEPTABLE/With the following exceptions.

Qualified Data: See the Qualified Sample Results.

Discussion:

Several surrogate percent recovery (%R) values were outside the 50% to 125% control limits. The %R outliers are summarized in **TABLE 6**, sample results qualified as a result of surrogate outliers are summarized in **TABLE 7**, and specific details are provided in the following text.

Standard Congener Analysis (GC/ECD)

For Sample WEEG04CP (Batch 97-191), one surrogate %R value was outside of the control limits. A laboratory duplicate analysis of this sample was performed, with acceptable %R values. The result from the field sample was qualified as do not report (DNR-13); the results from the laboratory duplicate should be used.

Sample EG-LM-F-01-FC-1 in Batch 97-129, and Samples EG-LM-F-09-WC-1 and EG-LM-F-10-WC-1 in Batch 97-192 were re-extracted and reanalyzed due to unacceptable surrogate recoveries. Sample EG-LM-F-09-WC-1 was also reanalyzed due to the sample spilling in the extraction process; see **SECTION II**. Sample EG-LM-F-08-FC-1 (Batch 97-129) had surrogate recovery values that were slightly above the lower control limit of 50% at 53% for Surrogate PCB 63 and 51% for Surrogate PCB 112. Although these recoveries were technically within the control limits, the sample was re-extraeted and reanalyzed to verify the recovery values. These re-extractions and reanalyses resulted in acceptable %R values. The results from the original analyses were qualified as do not report (DNR-13); the results from the reanalyses should be used.

Coplanar Congener Analysis (GC/ECD)

Seven samples in Batch 97-126 and 17 samples in Batch 97-192 (summarized in **TABLE 8**) were re-extracted and reanalyzed because of low surrogate percent recoveries. The results from the original analyses were qualified as do not report (DNR-13); the results from the reanalyses should be used.

For all other field samples summarized in **TABLE 7**, results were qualified as estimated (J-13/UJ-13) for %R values less 50% but greater than or equal to 10%. For %R values greater than the upper control limit, positive results were qualified as estimated (J-13); reporting limits were judged as not affected. Qualifiers were not assigned to QC samples.

Surrogate %R values less than the control limit may indicate that the sample results are biased low. The reported sample results are potentially underestimated. Surrogate %R values greater than the control limit indicate that the sample results are potentially biased high; however,

analytical interferences may be present that impact only the surrogate compounds, and these interferences may not impact the sample results.

GC/MS Analysis

As indicated in the QAPP, surrogate %R values were not calculated for the GC/MS analyses. Surrogates were evaluated based on %R values obtained from the GC/ECD analyses. The GC/MS sample results were qualified as estimated (J-13) when the recovery values from the GC/ECD analyses were not within the control limits.

The surrogate %R value ranges for all batches are summarized in **TABLE 9**.

VI. Blank Spike Sample Analysis: ACCEPTABLE/With the following exceptions.

Qualified Data: See the **Data Qualifier Summary Table**.

Discussion:

A blank spike (BS) was extracted and analyzed at the frequency requirement of one per batch. All spiked analyte recovery values were within the control limits of 50% to 125% for tri- through deca-chlorobiphenyls and 30% to 125% for mono- and dichlorobiphenyls, with the exceptions listed in **TABLE 10**.

Results associated with BS recovery values that were less than control limits were qualified as estimated (J-10/UJ-10). Positive results associated with BS recovery values that were greater than control limits were qualified as estimated (J-10). See **TABLE 11** for a summary of results qualified because of blank spike and SRM outliers.

The blank spike %R value ranges for all analytes within a batch are listed in the **TABLE 12.**

VII. Sample Duplicate Analysis: ACCEPTABLE/With the following exceptions.

Qualified Data: See the Qualified Sample Results.

Discussion:

One or more duplicate samples were extracted with each batch. The duplicate sample was analyzed by GC/ECD, but not GC/MS (as specified in the work plan). Several relative percent difference (RPD) values were greater than the control limit of 50% as listed in **TABLE 13.**

All associated sample results were qualified as estimated (J-9), with the exception of the results associated with the GC/ECD Extended PCB Congener laboratory duplicate analysis performed on Sample EG-LM-F-01-FC-1 (Batch 97-129). As mentioned in **SECTION V**, the surrogate %R values were less than the lower control limit for this field sample, but acceptable in the laboratory duplicate. The target analyte concentrations for positive results were likewise much lower in the field sample; thus, the RPD values were greater than 50%. Since the field sample was already qualified for surrogate recoveries, and the low recoveries were attributed to an isolated incident

(not indicative of a systematic problem for the batch), no qualifiers were assigned due to laboratory duplicate results for Batch 97-129. Qualified results are summarized in **TABLE 14**.

VIII. Standard Reference Material (SRM) Analysis: ACCEPTABLE/With the following exceptions.

Qualified Data: See the Qualified Sample Results.

Discussion:

SRM Carp-1 samples (acquired from the National Research Council, Canada) were extracted and analyzed at the required frequency of one per each batch. The results for the SRM were calculated and reported both surrogate-corrected and not surrogate-corrected; separate spreadsheet tables were submitted. Since the certified values for this SRM are based on surrogate-corrected quantification, only the surrogate-corrected results were evaluated for the GC/ECD analyses. The GC/ECD SRM surrogate recovery value ranges were 82% to 115% (PCB 36) and 61% to 92% (PCB 112) for all sample batches. For the GC/MS analyses, only the uncorrected values were evaluated because the surrogate-corrected values were based on surrogate %R values from the GC/ECD analyses. All results were within the established acceptance criteria, with the exceptions listed in **TABLE 15**.

For reported values that were greater than the upper acceptance criterion, positive results in associated samples were qualified as estimated (J-10). For reported values that were less than the lower acceptance criterion, associated sample results were qualified as estimated (J-10/UJ-10). See **Table 11** for a summary of results qualified because of blank spike and SRM outliers.

IX. Compound Identification and Quantitation: ACCEPTABLE/With the following exceptions.

Qualified Data: See the Qualified Sample Results.

Discussion:

As discussed in the **Calibrations Section**, several standard congener sample results were originally calculated incorrectly (using incorrect initial calibration coefficients) for the GC/ECD analyses. The laboratory submitted corrected results for the following samples: four QC samples for Batch 97-190 (the procedural blank, instrument blank, equipment blank, and blank spike); four QC samples (the procedural blank, instrument blank, equipment blank, and blank spike) and six field samples for Batch 97-191 (Samples BTEG01CP, BTEG04CP, BTUG01CP, BTUG02CP, BTUG04CP, and BTUG05CP); and four QC samples for Batch 97-192 (the procedural blank, instrument blank, equipment blank, and blank spike).

Standard Congener Analysis (GC/ECD)

The laboratory stated that there was significant coelution/interference with PCB85 in the field samples, which appeared to be caused by the presence of p,p'-DDE. Positive results for PCB85 may be biased high. All positive results for PCB85 were qualified as estimated (J-14). Since the

results for PCB85 may not be accurate, the laboratory provided a sum of all congeners with PCB85 excluded (as well as a sum of all congeners with PCB85 included). The congener summation without PCB85 is likely to be the more accurate measure of the total PCBs.

Coplanar Congener Analysis (GC/ECD)

The laboratory stated that there was a contaminant interfering with PCB169. The interference was a doublet peak on each side of the PCB169 peak. If a peak was clearly present in the valley between the two contaminant peaks, it was identified as PCB169, but the contaminant most likely masked the presence of this congener or reduced the accuracy of any quantification of this congener when detected. PCB169 results from the co-planar analyses were qualified as estimated (J-14/UJ-14).

Aroclor Analysis (GC/ECD)

The chromatograms of the walleye liver samples closely resembled a mixture of Aroclors 1248 and 1254. The results were reported as "1248.1254." The PCB pattern in the trout fillets most closely resembled Aroclor 1254, and results were reported to reflect this identification.

GC/MS Analysis

The laboratory assigned ME and MI qualifiers to several PCB22 and PCB16 results to reflect estimated positive results and estimated reporting limits, respectively . The laboratory stated that a matrix interference was present. The ME lab qualifier was applied in situations where the primary ion profile displayed somewhat of a bell-shaped curve but contained obvious saturation, while the secondary ion profile was present and clearly displayed a bell-shaped profile. The MI lab qualifier was applied when both the primary and secondary ions did not show bell-shaped profiles, or when the primary and secondary ions did not show bell-shaped profiles at the same retention time. All sample results that were flagged ME or MI by the laboratory were qualified as estimated (J-14/UJ-14). Qualified results are summarized in **Table 16.**

X. GC/ECD and GC/MS Results Comparison: ACCEPTABLE/With the following discussion.

The results of the 26 samples that were analyzed by both GC/MS and GC/ECD for standard congeners are summarized in **Table 17**. As discussed in **SECTION IX**, there was significant coelution/interference with PCB85 in the field samples for the GC/ECD standard congener analyses, which appeared to be caused by the presence of p,p'-DDE. Since the results for PCB85 are biased high for the GC/ECD analyses, the sum of all congeners with PCB85 excluded were used to compare to the GC/MS results. (For the GC/ECD analyses, the congener summation without PCB85 is likely to be a more accurate measure of the total PCB than the sum that includes PCB85. The GC/MS data provides a more accurate quantitation of PCB85.)

The RPD values for results from the GC/MS and GC/ECD analyses were all less than 20% indicating acceptable precision between the methods.

XI. Lipids Analysis: ACCEPTABLE/With the following discussion.

For each batch (excluding the re-extracted batches), percent lipids were performed in duplicate for one sample. For Batch 97-129, the percent lipid RPD values between the original and duplicate results were greater than the control limit of 20% at 43.5%. No qualification of data was necessary as the two lipid values were relatively low (the difference was 1.19%).

For Batch 197-128, the percent lipid RPD values between the original and duplicate results were greater than the control limit of 20.0% at 20.5%. For Batch 97-181, the percent lipid RPD values between the original and duplicate results were greater than the control limit at 28.3%. For Batch 97-192, the percent lipid RPD values between the original and duplicate results were greater than the control limit at 38.6%. Although these percent lipid results for these three batches were not qualified the data user should be aware of potential bias as a result of a lack of homogeneity. All other sample/duplicate percent lipid RPD values were less than the upper control limit of 20.0%.

All RPD values for consecutive weighings were less than the upper control limit of 20.0%.

Comparison of Solvents on % Lipid Values

The laboratory originally selected three samples for a comparison of lipid content using different solvents (hexane and dichloromethane). For two of the sample sets, the dichloromethane extraction method yielded %D values (dichloromethane relative to hexane) of 38.4% to 40.7% higher lipid content values. For the third sample set, the lipid content was 6% higher with the dichloromethane solvent. The sample amounts used for the comparison test were relatively small (5.10 to 7.46 grams for the hexane solvent and 0.9987 to 1.0354 grams for the dichloromethane solvent). The laboratory performed the comparison study on more samples, in order to obtain more statistically-reliable results.

The laboratory selected seven brown trout whole body samples and seven walleye whole body samples for another comparison study. The dichloromethane extraction method yielded higher lipid content values than the hexane extraction. The average lipid content was 43.5% higher with the dichloromethane method than the hexane method for the brown trout samples and 25.4% higher for the walleye samples.

The laboratory stated that the data are to be considered carefully before they are used to generate a generic method-to-method lipid content correction factor because there is clearly significant fish-to-fish variability. The %D values between the two methods ranged from 17.1% to 72.1% for the trout and 11.5% to 43.1% for the walleye. This notable fish-to-fish variability could be the result of slightly different lipid composition of different fish. Additionally, variability in the moisture content of the fish impacts the variability in the lipid data when calculated on a wet weight basis; the lipid are primarily associated with the dry matrix, not the wet. If the data were normalized for moisture content (i.e., calculating lipid content on a dry, not wet, weight basis), it is likely that the %D values between the methods will decrease.

Triplicate analyses were performed on one brown trout and one walleye sample. The %RSD values ranged from 3.9% to 17.9%, and were judged as acceptable, indicating that the observed variability between the different solvents is not due to the method.

XII Moisture Analysis: ACCEPTABLE/With the following discussion.

For each batch (excluding the re-extracted batches), percent moisture content was performed in duplicate for one sample. For Batch 97-124, the percent lipid RPD values between the original and duplicate results were greater than the control limit of 20.0% at 38.8%. For Batch 97-126, the percent lipid RPD values between the original and duplicate results were greater than the control limit at 30.8%. The laboratory stated that the percent moisture for the duplicate sample was performed several weeks after the original percent moisture. No qualifiers were assigned on this basis. All other RPD values were less than the upper control limit of 20.0%.

All RPD values for consecutive weighings were less than the upper control limit of 20.0%.

XIII. Overall Assessment of the Data

Based on this evaluation, the laboratory followed the specified method.

Accuracy was generally acceptable, as demonstrated by the %R values of the surrogate, the blank spike, and the SRM analytes, except where previously noted. Precision was generally acceptable, as demonstrated by the RPD values of the sample and laboratory duplicates, except where previously noted.

Qualifiers were assigned due to blank contamination, CCV %D outliers, blank spike results, surrogate outliers, laboratory duplicate results, SRM Carp-1 results, and chromatographic interferences.

Data that are qualified as DNR should not be used. All other data, as qualified, are acceptable for use.

Table 1
SAMPLE INDEX
CLIENT: HAGLER BAILLY
PROJECT NAME: GREEN BAY NRDA PROJECT
ECOCHEM PROJECT NO.: C9309-3

Sample ID	Aroclors by GC/ECD	Standard Congeners by GC/ECD	Co-Planar Congeners by GC/ECD	Standard Congeners by GC/MS
WEFR01LV	✓			
WELG04LV	✓			
WELG03LV	✓			
WELG02LV	✓			
WEWG02LV	✓			
WEWG04LV	✓			
WEEG04LV	✓			
WEEG02LV	✓			
WEEG01LV	✓			
WEUG01LV	✓			
WEUG02LV	✓			
WEUG03LV	✓			
TE-K1-B-06		✓	✓	✓
TE-K1-B-18		✓	✓	
TE-K1-B-24		✓	✓	
TE-K1-B-30		✓	✓	
TE-K1-B-48		✓	✓	
TE-K1-B-60		✓	✓	✓
96-KI-CT-01		✓	✓	✓
96-KI-CT-03		✓	✓	
96-KI-CT-05		✓	✓	
96-KI-CT-07		✓	✓	
96-KI-CT-09		✓	✓	
96-KI-CT-10		✓	✓	✓
BT-EG-01-FC-1	✓			
BT-EG-03-FC-1	✓			
BT-EG-04-FC-1	✓			
BT-EG-05-FC-1	✓			
BT-EG-06-FC-1	✓			
BT-EG-07-FC-1	✓			
BT-EG-09-FC-1	✓			
BT-GA-01-FC-1	✓			
BT-GA-02-FC-1	✓			
BT-GA-03-FC-1	✓			

Table 1
SAMPLE INDEX
CLIENT: HAGLER BAILLY
PROJECT NAME: GREEN BAY NRDA PROJECT
ECOCHEM PROJECT NO.: C9309-3

Sample ID	Aroclors by GC/ECD	Standard Congeners by GC/ECD	Co-Planar Congeners by GC/ECD	Standard Congeners by GC/MS
BT-GA-04-FC-1	✓			
BT-GA-05-FC-1	✓			
LT-LM-01-FC-1	✓			
LT-LM-02-FC-1	✓			
LT-LM-03-FC-1	✓			
LT-LM-04-FC-1	✓			
LT-LM-05-FC-1	✓			
LT-LM-06-FC-1	✓			
LT-LM-07-FC-1	✓			
LT-LM-08-FC-1	✓			
LT-LM-09-FC-1	✓			
LT-LM-10-FC-1	✓			
LT-IR-02-FC-1	✓			
LT-IR-06-FC-1	✓			
LT-IR-07-FC-1	✓			
EG-LM-F-01-F-C-1		✓	✓	
EG-LM-F-02-F-C-1		✓	✓	✓
EG-LM-F-03-F-C-1		✓	✓	
EG-LM-F-04-F-C-1		✓	✓	
EG-LM-F-05-F-C-1		✓	✓	✓
EG-LM-F-06-F-C-1		✓	✓	
EG-LM-F-07-F-C-1		✓	✓	
EG-LM-F-08-F-C-1		✓	✓	
EG-LM-F-09-F-C-1		✓	✓	✓
EG-LM-F-10-F-C-1		✓	✓	✓
EG-LM-F-11-F-C-1		✓	✓	
EG-LM-F-12-F-C-1		✓	✓	
LT-IR-08-FC-1	✓			
BT-EG-02-FC-1	✓			
BT-EG-08-FC-1	✓			
WELG01LV	✓			
WEWG01LV	✓			
WEWG03LV	✓			
WEEG03LV	✓			

Table 1
SAMPLE INDEX
CLIENT: HAGLER BAILLY
PROJECT NAME: GREEN BAY NRDA PROJECT
ECOCHEM PROJECT NO.: C9309-3

Sample ID	Aroclors by GC/ECD	Standard Congeners by GC/ECD	Co-Planar Congeners by GC/ECD	Standard Congeners by GC/MS
WEUG04LV	✓			
LT-IR-01-FC-1	✓			
WEFR01CP		✓		
WEFR02CP		✓		
WEFR03CP		✓		✓
WEFR04CP		✓		
WEFR05CP		✓		
WEFR06CP		✓		
WELG02CP		✓		
WELG03CP		✓		
WELG04CP		✓		
WELG05CP		✓		
WEWG01CP		✓		
WEWG02CP		✓		✓
WEWG03CP		✓		✓
WEEG01CP		✓		
WEEG03CP		✓		
WEEG04CP		✓		
WEEG05CP		✓		
WEEG06CP		✓		
WEEG07CP		✓		✓
WEEG08CP		✓		
WEEG10CP		✓		✓
WEEG11CP		✓		
WEUG01CP		✓		✓
WEUG03CP		✓		✓
BTEG01CP		✓		
BTEG03CP		✓		
BTEG04CP		✓		
BTEG05CP		✓		✓
BTUG01CP		✓		
BTUG02CP		✓		✓
BTUG04CP		✓		
BTUG05CP		✓		✓

Table 1
SAMPLE INDEX
CLIENT: HAGLER BAILLY
PROJECT NAME: GREEN BAY NRDA PROJECT
ECOCHEM PROJECT NO.: C9309-3

Sample ID	Aroclors by GC/ECD	Standard Congeners by GC/ECD	Co-Planar Congeners by GC/ECD	Standard Congeners by GC/MS
WEFR07CP		✓	✓	✓
WELG06CP		✓	✓	✓
WEWG04CP		✓	✓	
WEEG09CP		✓	✓	
WEUG02CP		✓	✓	
BTUG03CP		✓	✓	
BTEG02CP		✓	✓	✓
EG-LM-F-10-WC-1		✓	✓	✓
EG-LM-F-12-WC-1		✓	✓	
EG-LM-F-08-WC-1		✓	✓	
EG-LM-F-05-WC-1		✓	✓	✓
EG-LM-F-04-WC-1		✓	✓	
EG-LM-F-03-WC-1		✓	✓	✓
EG-LM-F-11-WC-1		✓	✓	
EG-LM-F-07-WC-1		✓	✓	
EG-LM-F-06-WC-1		✓	✓	
EG-LM-F-02-WC-1		✓	✓	
EG-LM-F-09-WC-1		✓	✓	✓
EG-LM-F-01-WC-1		✓	✓	
WELG01CP		✓		✓
WEEG02CP		✓		

Table 2
DATA VALIDATION QUALIFIER DEFINITIONS

U	The analyte was analyzed for, but was not detected above the reported sample quantitation limit.
J	The analyte was positively identified; the associated numerical value is the approximate concentration of the analyte in the sample.
UJ	The analyte was not detected above the reported sample quantitation limit. However, the reported quantitation limit is approximate and may or may not represent the actual limit of quantitation necessary to accurately and precisely measure the analyte in the sample.
R	The sample results are rejected due to serious deficiencies in the ability to analyze the sample and meet quality control criteria. The presence or absence of the analyte cannot be verified.
DNR	Do not report. A more usable set of data should be used instead.

Table 3
DATA VALIDATION QUALIFIER CODES

1	Holding Times
2	Sample Preservation
3	Sample Custody
4	Missing Deliverables
5A	Calibration (initial)
5B	Calibration (continuing)
6	Field Blanks
7	Laboratory Blanks
8	Matrix Spike
9	Precision (Duplicate, or Matrix Spike Duplicate)
10	Laboratory Control Sample
11	Detection Limit
12	Standards
13	Surrogates
14	Other
15	Furnace QC
16	ICP Serial Dilution
17	Chemical Recoveries
18	Trip Blanks
19	Internal Standards
20	Linear Range Exceeded
21	Potential False Positives

Table 4
SAMPLE RESULTS QUALIFIED AS A RESULT OF CONTINUING CALIBRATION OUTLIERS

Table 5
SAMPLE RESULTS QUALIFIED AS A RESULT OF BLANK OUTLIERS

Table 6
SURROGATE PERCENT RECOVERY OUTLIERS

Batch ID	Sample ID	Analysis	Congener	Percent Difference Value
97-124	97-124 CRM	GC/ECD Aroclor	PCB36	46%
	97-124 CRM	GC/ECD Aroclor	PCB112	35%
	WELG03LV	GC/ECD Aroclor	PCB112	127%
97-126	TE-K1-B-18	GC/ECD Congener Coplanar	PCB77-d	38%
	TE-K1-B-48	GC/ECD Congener Coplanar	PCB77-d	46%
	96-K1-CT-03	GC/ECD Congener Coplanar	PCB77-d	44%
	96-K1-CT-05	GC/ECD Congener Coplanar	PCB77-d	14%
	96-K1-CT-07	GC/ECD Congener Coplanar	PCB77-d	8%
	96-K1-CT-09	GC/ECD Congener Coplanar	PCB77-d	16%
	96-K1-CT-10	GC/ECD Congener Coplanar	PCB77-d	47%
97-128	97-128 CRM	GC/ECD Aroclor	PCB112	42%
97-129	EG-LM-F-01-FC-1	GC/ECD Congener (Standard)	PCB36	23%
	EG-LM-F-01-FC-1	GC/ECD Congener (Standard)	PCB112	17%
97-190	WEFR03CP	GC/ECD Congener (Standard)	PCB36	127%
	97-190 EB	GC/ECD Congener (Standard)	PCB112	126%
97-191	WEEG04CP	GC/ECD Congener (Standard)	PCB36	133%
	WEEG05CP	GC/ECD Congener (Standard)	PCB36	146%
	WEEG07CP	GC/ECD Congener (Standard)	PCB36	127%
97-192	WELG06CP	GC/ECD Congener (Standard)	PCB112	128%
	WEEG09CP	GC/ECD Congener (Standard)	PCB36	134%
	WEUG02CP	GC/ECD Congener (Standard)	PCB36	138%
	EG-LM-F-10-WC-1	GC/ECD Congener (Standard)	PCB36	178%
	EG-LM-F-10-WC-1	GC/ECD Congener (Standard)	PCB112	135%
	EG-LM-F-07-WC-1	GC/ECD Congener (Standard)	PCB112	48%
	EG-LM-F-09-WC-1	GC/ECD Congener (Standard)	PCB36	151%
	EG-LM-F-09-WC-1	GC/ECD Congener (Standard)	PCB112	137%
	WEFR07CP	GC/ECD Congener Coplanar	PCB77-d	17%
	WEFR06CP	GC/ECD Congener Coplanar	PCB77-d	33%
	WEFR04CP	GC/ECD Congener Coplanar	PCB77-d	48%
	BTU03CP	GC/ECD Congener Coplanar	PCB77-d	37%
	BTEG02CP	GC/ECD Congener Coplanar	PCB77-d	40%
	EG-LM-F-10-WC-1	GC/ECD Congener Coplanar	PCB77-d	13%
	EG-LM-F-12-WC-1	GC/ECD Congener Coplanar	PCB77-d	10%
	EG-LM-F-08-WC-1	GC/ECD Congener Coplanar	PCB77-d	29%
	EG-LM-F-05-WC-1	GC/ECD Congener Coplanar	PCB77-d	10%
	EG-LM-F-04-WC-1	GC/ECD Congener Coplanar	PCB77-d	10%
	EG-LM-F-03-WC-1	GC/ECD Congener Coplanar	PCB77-d	32%

Table 6
SURROGATE PERCENT RECOVERY OUTLIERS

Batch ID	Sample ID	Analysis	Congener	Percent Difference Value
	EG-LM-F-11-WC-1	GC/ECD Congener Coplanar	PCB77-d	21%
	EG-LM-F-07-WC-1	GC/ECD Congener Coplanar	PCB77-d	17%
	EG-LM-F-06-WC-1	GC/ECD Congener Coplanar	PCB77-d	19%
	EG-LM-F-02-WC-1	GC/ECD Congener Coplanar	PCB77-d	16%
	EG-LM-F-09-WC-1	GC/ECD Congener Coplanar	PCB77-d	33%
	EG-LM-F-01-WC-1	GC/ECD Congener Coplanar	PCB77-d	44%
	WEFR07CP DUP	GC/ECD Congener Coplanar	PCB77-d	43%
	97-192 BS	GC/ECD Congener Coplanar	PCB77-d	136%
97-274	WEFR07CP	GC/ECD Congener Coplanar	PCB112	47%
	WEFR07CP DUP	GC/ECD Congener Coplanar	PCB112	43%
	WELG06CP	GC/ECD Congener Coplanar	PCB112	42%
	TEKIB18	GC/ECD Congener Coplanar	PCB112	42%
	96KICT05	GC/ECD Congener Coplanar	PCB112	39%
	96KICT07	GC/ECD Congener Coplanar	PCB112	32%
	96KICT09	GC/ECD Congener Coplanar	PCB112	40%
	EGLMF06WC-1	GC/ECD Congener Coplanar	PCB36	21%
	EGLMF06WC-1	GC/ECD Congener Coplanar	PCB112	19%
97-306	97-306 BS	GC/ECD Congener Coplanar	PCB77-d	134%
	EG-LM-F-10-WC-1	GC/ECD Congener Coplanar	PCB77-d	20%
97-312	EG-LM-F-12-WC-1	GC/ECD Congener Coplanar	PCB36	28%
	EG-LM-F-12-WC-1	GC/ECD Congener Coplanar	PCB112	24%

Table 7
SAMPLE RESULTS QUALIFIED AS A RESULT OF SURROGATE PERCENT RECOVERY OUTLIERS

Table 8
SAMPLES RE-EXTRACTED AND REANALYZED FOR COPLANAR ANALYSIS

Batch 97-126:	TE-KI-B-18	TE-KI-B-48	96-KI-CT-03	96-KI-CT-05
	96-KI-CT-07	96-KI-CT-09	96-KI-CT-10	
Batch 97-192:	WEFR07CP	WELG06CP	WEWG04CP	BTUG03CP
	BTEG02CP	EG-LM-F-01-WC-1	EG-LM-F-02-WC-1	EG-LM-F-03-WC-1
	EG-LM-F-04-WC-1	EG-LM-F-05-WC-1	EG-LM-F-06-WC-1	EG-LM-F-07-WC-1
	EG-LM-F-08-WC-1	EG-LM-F-09-WC-1	EG-LM-F-10-WC-1	EG-LM-F-11-WC-1
	EG-LM-F-12-WC-1			

Table 9
SURROGATE PERCENT RECOVERY RANGES

Batch ID	Analysis	Surrogate Range
97-124	GC/ECD Aroclors	83% - 127%
97-126	GC/ECD Congener (Standard)	72% - 110%
	GC/ECD Congener (Coplanar)	8%* - 87%
97-127	GC/ECD Aroclors	73%-107%
97-128	GC/ECD Aroclors	61% - 121%
97-129	GC/ECD Congener (Standard)	17%* - 97%
	GC/ECD Congener (Coplanar)	84% - 120%
97-181	GC/ECD Aroclors	57% - 100%
97-190	GC/ECD Congener (Standard)	63% - 127%
97-191	GC/ECD Congener (Standard)	61% - 146%
97-192	GC/ECD Congener (Standard)	48%* - 178%
	GC/ECD Congener (Coplanar)	10%* - 78%
97-274	GC/ECD Congener (Coplanar)	19%* - 123%
97-306	GC/ECD Congener (Standard)	64% - 105%
	GC/ECD Congener (Coplanar)	20%* - 125%
97-312	GC/ECD Congener (Coplanar)	24%* - 108%

*As a result of these low surrogate recoveries, the samples were re-extracted and reanalyzed (See SECTION V).

Table 10
BLANK SPIKE PERCENT DIFFERENCE OUTLIERS

Batch ID	Analysis	Analyte	Percent Difference Value
97-126	GC/ECD Congener (Standard)	PCB175	44%
97-190	GC/ECD Congener (Standard)	PCB87	130%
	GC/ECD Congener (Standard)	PCB176	196%
	GC/ECD Congener (Standard)	PCB169	128%
97-191	GC/ECD Congener (Standard)	PCB176	139%
	GC/ECD Congener (Standard)	PCB169	151%
97-192	GC/ECD Congener (coplanar)	PCB169	142%
97-306	GC/ECD Congener (coplanar)	PCB126	135%
97-312	GC/ECD Congener (coplanar)	PCB37	48%

Table 11

SAMPLE RESULTS QUALIFIED AS A RESULT OF BLANK SPIKE PERCENT RECOVERY OUTLIERS

Table 12
BLANK SPIKE PERCENT RECOVERY RANGES

Batch ID	Analysis	Blank Spike %R Range
97-124	GC/ECD Aroclors	79% - 81%
97-126	GC/ECD Congener (Standard)	44% - 114%
	GC/ECD Congener (Coplanar)	63% - 84%
	GC/MS Congener (Standard)	79% - 98%
97-127	GC/ECD Aroclors	72% - 74%
97-128	GC/ECD Aroclors	75% - 75%
97-129	GC/ECD Congener (Standard)	51% - 109%
	GC/ECD Congener (Coplanar)	66% - 95%
	GC/MS Congener (Standard)	69% - 109%
97-181	GC/ECD Aroclors	75% - 76%
97-190	GC/ECD Congener (Standard)	81% - 196%
	GC/MS Congener (Standard)	77% - 90%
97-191	GC/ECD Congener (Standard)	75% - 151%
	GC/MS Congener (Standard)	77% - 100%
97-192	GC/ECD Congener (Standard)	68% - 91%
	GC/ECD Congener (Coplanar)	95% - 142%
	GC/MS Congener (Standard)	69% - 91%
97-274	GC/ECD Congener (Coplanar)	80% - 103%
97-306	GC/ECD Congener (Standard)	73% - 93%
	GC/ECD Congener (Coplanar)	86% - 135%
97-312	GC/MS Congener (Coplanar)	48% -68%

Table 13
DUPLICATE RELATIVE PERCENT DIFFERENCE OUTLIERS

Batch ID	Analysis	Sample	Analyte	RPD Value
97-126	GC/ECD Congener (Standard)	TE-KI-B-06	PCB63	74.6%
			PCB132	72.1%
	GC/ECD Congener (coplanar)		PCB37	87.6%
			PCB81	59.5%
97-129	GC/ECD Congener (Standard)	EG-LM-F-01-FC-1	PCB85	156.5%
			PCB110/77	144.3%
			PCB118	149.7%
			PCB153	170.5%
			PCB105	140.4%
			PCB138/160/163	157.4%
	GC/ECD Congener (coplanar)		PCB126	146.0%
			PCB77	169.2%
97-191	GC/ECD Congener (Standard)	WEEG04CP	PCB85	56.3%
97-192	GC/ECD Congener (Standard)	WEFR07CP	PCB85	51.2%
			PCB180	72.4%
97-274	GC/ECD Congener (coplanar)	WEFR07CP	PCB126	58.6%
97-306	GC/ECD Congener (Standard)	EG-LM-F-01-FC-1	All positive results >MDL except PCB114	> 50%
		EG-LM-F-08-FC-1	All positive results >MDL	> 50%

Table 14
SAMPLE RESULTS QUALIFIED AS A RESULT OF DUPLICATE RELATIVE PERCENT OUTLIERS

Table 15
STANDARD REFERENCE MATERIAL OUTLIERS

Batch ID	Analysis	Analyte	Acceptance Criteria (ng/g)	Reported Value (ng/g)
97-126	GC/ECD Congener (Standard)	PCB 18	13.8 - 28.8	12.56
	GC/ECD Congener (Standard)	PCB 170/190	14.3 - 29.7	10.67
97-129	GC/ECD Congener (Standard)	PCB 18	13.8 - 28.8	12.02
	GC/ECD Congener (Standard)	PCB 187/182	23.4 - 48.6	22.57
	GC/ECD Congener (Standard)	PCB 180	29.9 - 62.1	29.39
	GC/ECD Congener (Standard)	PCB 170/190	14.3 - 29.7	11.06
97-190	GC/ECD Congener (Standard)	PCB 66/95	87.1 - 180.9	191.06
	GC/ECD Congener (Standard)	PCB 118	85.8 - 178.2	269.25
	GC/ECD Congener (Standard)	PCB 153	54.0 - 112.1	138.86
	GC/MS Congener (GC/MS)	PCB 128	11.0 - 22.8	10.9
	GC/MS Congener (GC/MS)	PCB 170/190	14.3 - 29.7	13.7
97-191	GC/ECD Congener (Standard)	PCB 66/95	87.1 - 180.9	185.79
	GC/ECD Congener (Standard)	PCB 118	85.8 - 178.8	295.00
	GC/ECD Congener (Standard)	PCB 153	54.0 - 112.1	160.71
	GC/ECD Congener (Standard)	PCB 138/163/164	66.3 - 137.7	157.21
	GC/ECD Congener (Standard)	PCB 180	29.9 - 62.1	68.53
97-192	GC/ECD Congener (Standard)	PCB 118	85.8 - 178.2	246.30
97-306	GC/ECD Congener (Standard)	PCB 66/95	87.1 - 180.9	191.87

Table 16
GC/MS RESULTS QUALIFIED AS A RESULT OF POTENTIAL MATRIX INTERFERENCE

Table 17
GC/ECD - GC/MS SAMPLE RESULT RPD RANGES

SAMPLE RESULTS QUALIFIED AS A RESULT OF BLANK SPIKE AND SRM OUTLIERS (J/UJ-10)

Client_ID	Batch_ID	Matrix	Parameter	Concentration (ng/g, wet)
96KICT01	97-126	Tern Eggs	PCB18	1.54676
96KICT01	97-126	Tern Eggs	PCB175	8.67678
96KICT01	97-126	Tern Eggs	PCB170/190	168.53827
96KICT03	97-126	Tern Eggs	PCB18	0.00000
96KICT03	97-126	Tern Eggs	PCB175	1.91263
96KICT03	97-126	Tern Eggs	PCB170/190	54.94893
96KICT05	97-126	Tern Eggs	PCB18	0.00000
96KICT05	97-126	Tern Eggs	PCB175	2.10017
96KICT05	97-126	Tern Eggs	PCB170/190	73.09801
96KICT07	97-126	Tern Eggs	PCB18	0.00000
96KICT07	97-126	Tern Eggs	PCB175	1.38117
96KICT07	97-126	Tern Eggs	PCB170/190	73.61305
96KICT09	97-126	Tern Eggs	PCB18	1.13160
96KICT09	97-126	Tern Eggs	PCB175	2.70641
96KICT09	97-126	Tern Eggs	PCB170/190	52.57390
96KICT10	97-126	Tern Eggs	PCB18	0.00000
96KICT10	97-126	Tern Eggs	PCB175	3.32402
96KICT10	97-126	Tern Eggs	PCB170/190	116.38861
TEKIB06	97-126	Tern Eggs	PCB175	4.43473
TEKIB06	97-126	Tern Eggs	PCB18	5.49921
TEKIB06	97-126	Tern Eggs	PCB170/190	104.89890
TEKIB18	97-126	Tern Eggs	PCB18	1.92204
TEKIB18	97-126	Tern Eggs	PCB175	3.34540
TEKIB18	97-126	Tern Eggs	PCB170/190	59.55321
TEKIB24	97-126	Tern Eggs	PCB175	3.51152
TEKIB24	97-126	Tern Eggs	PCB18	3.83493
TEKIB24	97-126	Tern Eggs	PCB170/190	60.34811
TEKIB30	97-126	Tern Eggs	PCB175	0.00000
TEKIB30	97-126	Tern Eggs	PCB18	2.97246
TEKIB30	97-126	Tern Eggs	PCB170/190	53.32203
TEKIB48	97-126	Tern Eggs	PCB18	2.09935
TEKIB48	97-126	Tern Eggs	PCB175	2.66088
TEKIB48	97-126	Tern Eggs	PCB170/190	45.96529
TEKIB60	97-126	Tern Eggs	PCB18	3.24269
TEKIB60	97-126	Tern Eggs	PCB175	7.42168
TEKIB60	97-126	Tern Eggs	PCB170/190	133.98202
EGLMF02FC-1	97-129	Lake Trout Eggs	PCB18	0.20411
EGLMF02FC-1	97-129	Lake Trout Eggs	PCB187/182	11.26807
EGLMF02FC-1	97-129	Lake Trout Eggs	PCB180	22.98183
EGLMF02FC-1	97-129	Lake Trout Eggs	PCB170/190	5.32721
EGLMF03FC-1	97-129	Lake Trout Eggs	PCB18	0.63694
EGLMF03FC-1	97-129	Lake Trout Eggs	PCB187/182	4.94269
EGLMF03FC-1	97-129	Lake Trout Eggs	PCB180	10.82764
EGLMF03FC-1	97-129	Lake Trout Eggs	PCB170/190	2.42365
EGLMF04FC-1	97-129	Lake Trout Eggs	PCB18	0.93536
EGLMF04FC-1	97-129	Lake Trout Eggs	PCB187/182	9.85506
EGLMF04FC-1	97-129	Lake Trout Eggs	PCB180	18.21514
EGLMF04FC-1	97-129	Lake Trout Eggs	PCB170/190	4.48064
EGLMF05FC-1	97-129	Lake Trout Eggs	PCB18	1.93279
EGLMF05FC-1	97-129	Lake Trout Eggs	PCB187/182	13.36861

SAMPLE RESULTS QUALIFIED AS A RESULT OF BLANK SPIKE AND SRM OUTLIERS (J/UJ-10)

Client_ID	Batch_ID	Matrix	Parameter	Concentration (ng/g, wet)
EGLMF05FC-1	97-129	Lake Trout Eggs	PCB180	23.85250
EGLMF05FC-1	97-129	Lake Trout Eggs	PCB170/190	6.58034
EGLMF06FC-1	97-129	Lake Trout Eggs	PCB18	0.00000
EGLMF06FC-1	97-129	Lake Trout Eggs	PCB187/182	12.11489
EGLMF06FC-1	97-129	Lake Trout Eggs	PCB180	23.40895
EGLMF06FC-1	97-129	Lake Trout Eggs	PCB170/190	5.29166
EGLMF07FC-1	97-129	Lake Trout Eggs	PCB18	0.99927
EGLMF07FC-1	97-129	Lake Trout Eggs	PCB187/182	11.07883
EGLMF07FC-1	97-129	Lake Trout Eggs	PCB180	23.14357
EGLMF07FC-1	97-129	Lake Trout Eggs	PCB170/190	5.34569
EGLMF09FC-1	97-129	Lake Trout Eggs	PCB18	1.71783
EGLMF09FC-1	97-129	Lake Trout Eggs	PCB187/182	13.80636
EGLMF09FC-1	97-129	Lake Trout Eggs	PCB180	25.55064
EGLMF09FC-1	97-129	Lake Trout Eggs	PCB170/190	6.97092
EGLMF10FC-1	97-129	Lake Trout Eggs	PCB18	1.93920
EGLMF10FC-1	97-129	Lake Trout Eggs	PCB187/182	14.27640
EGLMF10FC-1	97-129	Lake Trout Eggs	PCB180	27.61000
EGLMF10FC-1	97-129	Lake Trout Eggs	PCB170/190	7.35160
EGLMF11FC-1	97-129	Lake Trout Eggs	PCB18	1.26226
EGLMF11FC-1	97-129	Lake Trout Eggs	PCB187/182	4.86918
EGLMF11FC-1	97-129	Lake Trout Eggs	PCB180	15.22107
EGLMF11FC-1	97-129	Lake Trout Eggs	PCB170/190	2.57759
EGLMF12FC-1	97-129	Lake Trout Eggs	PCB18	0.66631
EGLMF12FC-1	97-129	Lake Trout Eggs	PCB187/182	8.77747
EGLMF12FC-1	97-129	Lake Trout Eggs	PCB180	20.02061
EGLMF12FC-1	97-129	Lake Trout Eggs	PCB170/190	4.13528
WEEG01CP	97-190	Walleye Whole	PCB66	633.49489
WEEG01CP	97-190	Walleye Whole	PCB95	148.70538
WEEG01CP	97-190	Walleye Whole	PCB87/115/81	97.68867
WEEG01CP	97-190	Walleye Whole	PCB118	353.89500
WEEG01CP	97-190	Walleye Whole	PCB153	493.99963
WEEG01CP	97-190	Walleye Whole	PCB169	4.15763
WEEG03CP	97-190	Walleye Whole	PCB66	661.00181
WEEG03CP	97-190	Walleye Whole	PCB95	153.82531
WEEG03CP	97-190	Walleye Whole	PCB87/115/81	116.20330
WEEG03CP	97-190	Walleye Whole	PCB118	331.75705
WEEG03CP	97-190	Walleye Whole	PCB153	460.58435
WEEG03CP	97-190	Walleye Whole	PCB176	3.97052
WEEG03CP	97-190	Walleye Whole	PCB169	4.79962
WEFR01CP	97-190	Walleye Whole	PCB66	374.96763
WEFR01CP	97-190	Walleye Whole	PCB95	92.56827
WEFR01CP	97-190	Walleye Whole	PCB87/115/81	42.24894
WEFR01CP	97-190	Walleye Whole	PCB118	107.28683
WEFR01CP	97-190	Walleye Whole	PCB153	76.24384
WEFR01CP	97-190	Walleye Whole	PCB169	0.30085
WEFR02CP	97-190	Walleye Whole	PCB66	342.07992
WEFR02CP	97-190	Walleye Whole	PCB95	80.43008
WEFR02CP	97-190	Walleye Whole	PCB87/115/81	37.63168
WEFR02CP	97-190	Walleye Whole	PCB118	98.74544
WEFR02CP	97-190	Walleye Whole	PCB153	73.34976

SAMPLE RESULTS QUALIFIED AS A RESULT OF BLANK SPIKE AND SRM OUTLIERS (J/UJ-10)

Client_ID	Batch_ID	Matrix	Parameter	Concentration (ng/g, wet)
WEFR02CP	97-190	Walleye Whole	PCB176	1.13528
WEFR02CP	97-190	Walleye Whole	PCB169	0.43976
WEFR03CP	97-190	Walleye Whole	PCB66	444.70152
WEFR03CP	97-190	Walleye Whole	PCB95	105.24487
WEFR03CP	97-190	Walleye Whole	PCB87/115/81	56.57259
WEFR03CP	97-190	Walleye Whole	PCB118	132.92940
WEFR03CP	97-190	Walleye Whole	PCB153	106.43431
WEFR03CP	97-190	Walleye Whole	PCB176	1.62581
WEFR03CP	97-190	Walleye Whole	PCB169	0.86217
WEFR04CP	97-190	Walleye Whole	PCB66	239.63492
WEFR04CP	97-190	Walleye Whole	PCB95	50.30005
WEFR04CP	97-190	Walleye Whole	PCB87/115/81	44.05128
WEFR04CP	97-190	Walleye Whole	PCB118	153.53534
WEFR04CP	97-190	Walleye Whole	PCB153	133.95748
WEFR04CP	97-190	Walleye Whole	PCB176	0.54151
WEFR04CP	97-190	Walleye Whole	PCB169	0.34923
WEFR05CP	97-190	Walleye Whole	PCB66	373.18192
WEFR05CP	97-190	Walleye Whole	PCB95	98.29816
WEFR05CP	97-190	Walleye Whole	PCB87/115/81	46.37570
WEFR05CP	97-190	Walleye Whole	PCB118	108.33469
WEFR05CP	97-190	Walleye Whole	PCB153	79.74663
WEFR05CP	97-190	Walleye Whole	PCB176	0.75765
WEFR06CP	97-190	Walleye Whole	PCB66	479.89904
WEFR06CP	97-190	Walleye Whole	PCB95	88.36056
WEFR06CP	97-190	Walleye Whole	PCB87/115/81	58.34962
WEFR06CP	97-190	Walleye Whole	PCB118	145.70553
WEFR06CP	97-190	Walleye Whole	PCB153	124.89046
WEFR06CP	97-190	Walleye Whole	PCB176	1.61128
WEFR06CP	97-190	Walleye Whole	PCB169	1.10415
WELG02CP	97-190	Walleye Whole	PCB66	216.00470
WELG02CP	97-190	Walleye Whole	PCB95	52.68318
WELG02CP	97-190	Walleye Whole	PCB87/115/81	27.77964
WELG02CP	97-190	Walleye Whole	PCB118	65.49555
WELG02CP	97-190	Walleye Whole	PCB153	48.94966
WELG02CP	97-190	Walleye Whole	PCB176	0.72470
WELG02CP	97-190	Walleye Whole	PCB169	0.30559
WELG03CP	97-190	Walleye Whole	PCB66	304.97027
WELG03CP	97-190	Walleye Whole	PCB95	55.66528
WELG03CP	97-190	Walleye Whole	PCB87/115/81	63.26913
WELG03CP	97-190	Walleye Whole	PCB118	215.47620
WELG03CP	97-190	Walleye Whole	PCB153	193.67681
WELG03CP	97-190	Walleye Whole	PCB176	0.89256
WELG03CP	97-190	Walleye Whole	PCB169	0.56876
WELG04CP	97-190	Walleye Whole	PCB66	440.09322
WELG04CP	97-190	Walleye Whole	PCB95	82.28819
WELG04CP	97-190	Walleye Whole	PCB87/115/81	61.76615
WELG04CP	97-190	Walleye Whole	PCB118	144.76877
WELG04CP	97-190	Walleye Whole	PCB153	131.48750
WELG04CP	97-190	Walleye Whole	PCB176	0.83231
WELG04CP	97-190	Walleye Whole	PCB169	1.35028

SAMPLE RESULTS QUALIFIED AS A RESULT OF BLANK SPIKE AND SRM OUTLIERS (J/UJ-10)

Client_ID	Batch_ID	Matrix	Parameter	Concentration (ng/g, wet)
WELG05CP	97-190	Walleye Whole	PCB66	542.11292
WELG05CP	97-190	Walleye Whole	PCB95	92.25340
WELG05CP	97-190	Walleye Whole	PCB87/115/81	69.10568
WELG05CP	97-190	Walleye Whole	PCB118	173.22216
WELG05CP	97-190	Walleye Whole	PCB153	157.19023
WELG05CP	97-190	Walleye Whole	PCB176	2.18523
WELG05CP	97-190	Walleye Whole	PCB169	1.69577
WEWG01CP	97-190	Walleye Whole	PCB66	369.64548
WEWG01CP	97-190	Walleye Whole	PCB95	77.42129
WEWG01CP	97-190	Walleye Whole	PCB87/115/81	62.33032
WEWG01CP	97-190	Walleye Whole	PCB118	191.62172
WEWG01CP	97-190	Walleye Whole	PCB153	203.62350
WEWG01CP	97-190	Walleye Whole	PCB176	2.82216
WEWG01CP	97-190	Walleye Whole	PCB169	2.39732
WEWG02CP	97-190	Walleye Whole	PCB66	456.37483
WEWG02CP	97-190	Walleye Whole	PCB95	95.49359
WEWG02CP	97-190	Walleye Whole	PCB87/115/81	61.90176
WEWG02CP	97-190	Walleye Whole	PCB118	174.36441
WEWG02CP	97-190	Walleye Whole	PCB153	164.22295
WEWG02CP	97-190	Walleye Whole	PCB169	1.55698
WEWG03CP	97-190	Walleye Whole	PCB66	415.93033
WEWG03CP	97-190	Walleye Whole	PCB95	100.32650
WEWG03CP	97-190	Walleye Whole	PCB87/115/81	65.52732
WEWG03CP	97-190	Walleye Whole	PCB118	187.99572
WEWG03CP	97-190	Walleye Whole	PCB153	202.40298
WEWG03CP	97-190	Walleye Whole	PCB176	1.90026
WEWG03CP	97-190	Walleye Whole	PCB169	2.44463
BTEG01CP	97-191	B.Trout Whole	PCB95	32.74072
BTEG03CP	97-191	B.Trout Whole	PCB66	117.39217
BTEG03CP	97-191	B.Trout Whole	PCB95	30.90223
BTEG03CP	97-191	B.Trout Whole	PCB118	116.73981
BTEG03CP	97-191	B.Trout Whole	PCB153	158.43260
BTEG03CP	97-191	B.Trout Whole	PCB138/160/163	120.15413
BTEG03CP	97-191	B.Trout Whole	PCB180	42.04412
BTEG03CP	97-191	B.Trout Whole	PCB169	1.34341
BTEG04CP	97-191	B.Trout Whole	PCB95	24.87007
BTEG05CP	97-191	B.Trout Whole	PCB66	164.27513
BTEG05CP	97-191	B.Trout Whole	PCB95	35.42025
BTEG05CP	97-191	B.Trout Whole	PCB118	162.66380
BTEG05CP	97-191	B.Trout Whole	PCB153	211.58677
BTEG05CP	97-191	B.Trout Whole	PCB176	0.77516
BTEG05CP	97-191	B.Trout Whole	PCB138/160/163	160.67211
BTEG05CP	97-191	B.Trout Whole	PCB180	53.54520
BTEG05CP	97-191	B.Trout Whole	PCB169	1.27454
BTUG01CP	97-191	B.Trout Whole	PCB95	30.41482
BTUG02CP	97-191	B.Trout Whole	PCB95	23.76715
BTUG04CP	97-191	B.Trout Whole	PCB95	21.60728
BTUG05CP	97-191	B.Trout Whole	PCB95	27.97568
WEEG05CP	97-191	Walleye Whole	PCB66	672.56445
WEEG05CP	97-191	Walleye Whole	PCB95	133.67184

SAMPLE RESULTS QUALIFIED AS A RESULT OF BLANK SPIKE AND SRM OUTLIERS (J/UJ-10)

Client_ID	Batch_ID	Matrix	Parameter	Concentration (ng/g, wet)
WEEG05CP	97-191	Walleye Whole	PCB118	324.45246
WEEG05CP	97-191	Walleye Whole	PCB153	464.42202
WEEG05CP	97-191	Walleye Whole	PCB176	4.40130
WEEG05CP	97-191	Walleye Whole	PCB138/160/163	472.40318
WEEG05CP	97-191	Walleye Whole	PCB180	137.25847
WEEG05CP	97-191	Walleye Whole	PCB169	5.17204
WEEG06CP	97-191	Walleye Whole	PCB66	593.63072
WEEG06CP	97-191	Walleye Whole	PCB95	158.17253
WEEG06CP	97-191	Walleye Whole	PCB118	296.22680
WEEG06CP	97-191	Walleye Whole	PCB153	407.54164
WEEG06CP	97-191	Walleye Whole	PCB176	4.43453
WEEG06CP	97-191	Walleye Whole	PCB138/160/163	417.57820
WEEG06CP	97-191	Walleye Whole	PCB180	124.09919
WEEG06CP	97-191	Walleye Whole	PCB169	4.61514
WEEG07CP	97-191	Walleye Whole	PCB66	1061.45469
WEEG07CP	97-191	Walleye Whole	PCB95	180.11058
WEEG07CP	97-191	Walleye Whole	PCB118	574.62261
WEEG07CP	97-191	Walleye Whole	PCB153	775.61102
WEEG07CP	97-191	Walleye Whole	PCB176	5.23457
WEEG07CP	97-191	Walleye Whole	PCB138/160/163	710.73864
WEEG07CP	97-191	Walleye Whole	PCB180	187.83725
WEEG07CP	97-191	Walleye Whole	PCB169	6.24752
WEEG08CP	97-191	Walleye Whole	PCB66	769.71423
WEEG08CP	97-191	Walleye Whole	PCB95	163.65783
WEEG08CP	97-191	Walleye Whole	PCB118	466.52899
WEEG08CP	97-191	Walleye Whole	PCB153	654.24995
WEEG08CP	97-191	Walleye Whole	PCB176	3.08746
WEEG08CP	97-191	Walleye Whole	PCB138/160/163	605.35534
WEEG08CP	97-191	Walleye Whole	PCB180	166.67238
WEEG08CP	97-191	Walleye Whole	PCB169	5.52434
WEEG10CP	97-191	Walleye Whole	PCB66	1770.43495
WEEG10CP	97-191	Walleye Whole	PCB95	298.03959
WEEG10CP	97-191	Walleye Whole	PCB118	983.16688
WEEG10CP	97-191	Walleye Whole	PCB153	1072.11776
WEEG10CP	97-191	Walleye Whole	PCB176	4.52897
WEEG10CP	97-191	Walleye Whole	PCB138/160/163	877.35862
WEEG10CP	97-191	Walleye Whole	PCB180	274.01203
WEEG10CP	97-191	Walleye Whole	PCB169	7.08917
WEEG11CP	97-191	Walleye Whole	PCB66	461.80057
WEEG11CP	97-191	Walleye Whole	PCB95	114.97922
WEEG11CP	97-191	Walleye Whole	PCB118	209.97651
WEEG11CP	97-191	Walleye Whole	PCB153	257.78488
WEEG11CP	97-191	Walleye Whole	PCB176	3.21162
WEEG11CP	97-191	Walleye Whole	PCB138/160/163	252.85243
WEEG11CP	97-191	Walleye Whole	PCB180	99.79975
WEEG11CP	97-191	Walleye Whole	PCB169	4.04364
WEUG01CP	97-191	Walleye Whole	PCB66	447.25055
WEUG01CP	97-191	Walleye Whole	PCB95	130.18832
WEUG01CP	97-191	Walleye Whole	PCB118	195.12208
WEUG01CP	97-191	Walleye Whole	PCB153	236.29354

SAMPLE RESULTS QUALIFIED AS A RESULT OF BLANK SPIKE AND SRM OUTLIERS (J/UJ-10)

Client_ID	Batch_ID	Matrix	Parameter	Concentration (ng/g, wet)
WEUG01CP	97-191	Walleye Whole	PCB176	2.88719
WEUG01CP	97-191	Walleye Whole	PCB138/160/163	248.78818
WEUG01CP	97-191	Walleye Whole	PCB180	86.76450
WEUG01CP	97-191	Walleye Whole	PCB169	3.20138
WEUG03CP	97-191	Walleye Whole	PCB66	524.77593
WEUG03CP	97-191	Walleye Whole	PCB95	144.80335
WEUG03CP	97-191	Walleye Whole	PCB118	308.48427
WEUG03CP	97-191	Walleye Whole	PCB153	480.97790
WEUG03CP	97-191	Walleye Whole	PCB176	3.57025
WEUG03CP	97-191	Walleye Whole	PCB138/160/163	447.88667
WEUG03CP	97-191	Walleye Whole	PCB180	161.71550
WEUG03CP	97-191	Walleye Whole	PCB169	7.78640
BTEG02CP	97-192	B.Trout Whole	PCB118	95.87542
BTUG03CP	97-192	B.Trout Whole	PCB118	69.66250
EGLMF01WC-1	97-192	L.Trout Whole	PCB118	266.54243
EGLMF02WC-1	97-192	L.Trout Whole	PCB118	348.09430
EGLMF03WC-1	97-192	L.Trout Whole	PCB118	485.26712
EGLMF04WC-1	97-192	L.Trout Whole	PCB118	270.69644
EGLMF05WC-1	97-192	L.Trout Whole	PCB118	559.75962
EGLMF06WC-1	97-192	L.Trout Whole	PCB118	237.71889
EGLMF07WC-1	97-192	L.Trout Whole	PCB118	435.29363
EGLMF08WC-1	97-192	L.Trout Whole	PCB118	224.05929
EGLMF11WC-1	97-192	L.Trout Whole	PCB118	112.11703
EGLMF12WC-1	97-192	L.Trout Whole	PCB118	336.44410
WEEG02CP	97-192	Walleye Whole	PCB118	172.33138
WEEG09CP	97-192	Walleye Whole	PCB169	0.12292
WEEG09CP	97-192	Walleye Whole	PCB118	275.74328
WEFR07CP	97-192	Walleye Whole	PCB118	312.70665
WELG01CP	97-192	Walleye Whole	PCB118	251.21135
WELG06CP	97-192	Walleye Whole	PCB118	517.99117
WEUG02CP	97-192	Walleye Whole	PCB118	168.14965
WEWG04CP	97-192	Walleye Whole	PCB118	121.75901
96KICT03	97-306	Tern Eggs	PCB126	1.46916
96KICT03	97-306	Tern Eggs	PCB126	1.30666
96KICT10	97-306	Tern Eggs	PCB126	1.23320
96KICT10	97-306	Tern Eggs	PCB126	1.12070
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB66	50.37475
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB95	14.95517
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB95	7.24453
EGLMF07WC-1	97-306	L.Trout Whole	PCB126	1.37184
EGLMF07WC-1	97-306	L.Trout Whole	PCB126	1.96529
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB66	34.23498
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB95	9.31798
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB95	5.53896
EGLMF09WC-1	97-306	L.Trout Whole	PCB66	303.94919
EGLMF09WC-1	97-306	L.Trout Whole	PCB95	83.95568
EGLMF09WC-1	97-306	L.Trout Whole	PCB66	471.90089
EGLMF09WC-1	97-306	L.Trout Whole	PCB95	117.61421
EGLMF09WC-1	97-306	L.Trout Whole	PCB126	2.17774
EGLMF09WC-1	97-306	L.Trout Whole	PCB126	1.72864

SAMPLE RESULTS QUALIFIED AS A RESULT OF BLANK SPIKE AND SRM OUTLIERS (J/UJ-10)

Client_ID	Batch_ID	Matrix	Parameter	Concentration (ng/g, wet)
EGLMF10WC-1	97-306	L.Trout Whole	PCB66	168.86992
EGLMF10WC-1	97-306	L.Trout Whole	PCB95	44.75489
EGLMF10WC-1	97-306	L.Trout Whole	PCB126	0.20755
EGLMF11WC-1	97-306	L.Trout Whole	PCB126	0.68170
TEKIB48	97-306	Tern Eggs	PCB126	0.80491
WEWG04CP	97-306	Walleye Whole	PCB126	0.93064
WEWG04CP	97-306	Walleye Whole	PCB126	0.70704
BTUG03CP	97-312	B.Trout Whole	PCB37	0.27924
BTUG03CP	97-312	B.Trout Whole	PCB37	0.39659
EGLMF01WC-1	97-312	L.Trout Whole	PCB37	0.25142
EGLMF02WC-1	97-312	L.Trout Whole	PCB37	0.38638
EGLMF03WC-1	97-312	L.Trout Whole	PCB37	0.20341
EGLMF03WC-1	97-312	L.Trout Whole	PCB37	0.23220
EGLMF04WC-1	97-312	L.Trout Whole	PCB37	0.30943
EGLMF05WC-1	97-312	L.Trout Whole	PCB37	0.43653
EGLMF08WC-1	97-312	L.Trout Whole	PCB37	0.00000
EGLMF12WC-1	97-312	L.Trout Whole	PCB37	0.19078

SAMPLE RESULTS QUALIFIED AS A RESULT OF DUPLICATE RPD OUTLIERS (J/UJ-9)

Client ID	Batch ID	Matrix	Parameter	Concentration (ng/g, wet)
96KICT01	97-126	Tern Eggs	PCB37	0.72666
96KICT01	97-126	Tern Eggs	PCB81	0.99183
96KICT01	97-126	Tern Eggs	PCB63	50.82982
96KICT01	97-126	Tern Eggs	PCB132	141.44357
96KICT03	97-126	Tern Eggs	PCB63	26.37473
96KICT03	97-126	Tern Eggs	PCB132	32.58346
96KICT05	97-126	Tern Eggs	PCB132	29.96365
96KICT05	97-126	Tern Eggs	PCB63	48.25779
96KICT07	97-126	Tern Eggs	PCB63	7.18822
96KICT07	97-126	Tern Eggs	PCB132	54.35061
96KICT09	97-126	Tern Eggs	PCB132	39.62165
96KICT09	97-126	Tern Eggs	PCB63	41.85027
96KICT10	97-126	Tern Eggs	PCB63	29.19699
96KICT10	97-126	Tern Eggs	PCB132	269.00683
TEKIB06	97-126	Tern Eggs	PCB37	0.18066
TEKIB06	97-126	Tern Eggs	PCB81	1.22174
TEKIB06	97-126	Tern Eggs	PCB63	26.17184
TEKIB06	97-126	Tern Eggs	PCB132	42.18158
TEKIB18	97-126	Tern Eggs	PCB132	32.27604
TEKIB18	97-126	Tern Eggs	PCB63	38.92911
TEKIB24	97-126	Tern Eggs	PCB37	0.20807
TEKIB24	97-126	Tern Eggs	PCB81	0.71252
TEKIB24	97-126	Tern Eggs	PCB63	21.17520
TEKIB24	97-126	Tern Eggs	PCB132	54.53029
TEKIB30	97-126	Tern Eggs	PCB37	0.17747
TEKIB30	97-126	Tern Eggs	PCB81	0.51214
TEKIB30	97-126	Tern Eggs	PCB63	17.85565
TEKIB30	97-126	Tern Eggs	PCB132	42.98764
TEKIB48	97-126	Tern Eggs	PCB63	31.78046
TEKIB48	97-126	Tern Eggs	PCB132	52.20509
TEKIB60	97-126	Tern Eggs	PCB37	0.28751
TEKIB60	97-126	Tern Eggs	PCB81	0.63917
TEKIB60	97-126	Tern Eggs	PCB63	30.24157
TEKIB60	97-126	Tern Eggs	PCB132	81.48316
EGLMF01FC-1	97-129	Lake Trout Eggs	PCB77	0.11498
EGLMF01FC-1	97-129	Lake Trout Eggs	PCB126	0.05915
EGLMF02FC-1	97-129	Lake Trout Eggs	PCB77	1.31113
EGLMF02FC-1	97-129	Lake Trout Eggs	PCB126	0.31115
EGLMF03FC-1	97-129	Lake Trout Eggs	PCB77	0.52903
EGLMF03FC-1	97-129	Lake Trout Eggs	PCB126	0.17619
EGLMF04FC-1	97-129	Lake Trout Eggs	PCB77	0.85347
EGLMF04FC-1	97-129	Lake Trout Eggs	PCB126	0.19730
EGLMF05FC-1	97-129	Lake Trout Eggs	PCB77	1.22824
EGLMF05FC-1	97-129	Lake Trout Eggs	PCB126	0.31402
EGLMF06FC-1	97-129	Lake Trout Eggs	PCB77	1.13212
EGLMF06FC-1	97-129	Lake Trout Eggs	PCB126	0.29061
EGLMF07FC-1	97-129	Lake Trout Eggs	PCB77	1.36861
EGLMF07FC-1	97-129	Lake Trout Eggs	PCB126	0.32697
EGLMF08FC-1	97-129	Lake Trout Eggs	PCB77	0.21401

SAMPLE RESULTS QUALIFIED AS A RESULT OF DUPLICATE RPD OUTLIERS (J/UJ-9)

Client ID	Batch ID	Matrix	Parameter	Concentration (ng/g, wet)
EGLMF09FC-1	97-129	Lake Trout Eggs	PCB77	1.25099
EGLMF09FC-1	97-129	Lake Trout Eggs	PCB126	0.34722
EGLMF10FC-1	97-129	Lake Trout Eggs	PCB77	2.09536
EGLMF10FC-1	97-129	Lake Trout Eggs	PCB126	0.42840
EGLMF11FC-1	97-129	Lake Trout Eggs	PCB77	0.65290
EGLMF11FC-1	97-129	Lake Trout Eggs	PCB126	0.07803
EGLMF12FC-1	97-129	Lake Trout Eggs	PCB77	0.15594
EGLMF12FC-1	97-129	Lake Trout Eggs	PCB126	0.18373
BTEG01CP	97-191	B.Trout Whole	PCB85	309.21389
BTEG03CP	97-191	B.Trout Whole	PCB85	402.69191
BTEG04CP	97-191	B.Trout Whole	PCB85	326.33964
BTEG05CP	97-191	B.Trout Whole	PCB85	494.54958
BTUG01CP	97-191	B.Trout Whole	PCB85	378.32038
BTUG02CP	97-191	B.Trout Whole	PCB85	327.73007
BTUG04CP	97-191	B.Trout Whole	PCB85	328.96003
BTUG05CP	97-191	B.Trout Whole	PCB85	342.65271
WEEG05CP	97-191	Walleye Whole	PCB85	2162.94313
WEEG06CP	97-191	Walleye Whole	PCB85	1943.59612
WEEG07CP	97-191	Walleye Whole	PCB85	2167.03630
WEEG08CP	97-191	Walleye Whole	PCB85	2133.26683
WEEG10CP	97-191	Walleye Whole	PCB85	2205.65340
WEEG11CP	97-191	Walleye Whole	PCB85	1382.98217
WEUG01CP	97-191	Walleye Whole	PCB85	1344.32135
WEUG03CP	97-191	Walleye Whole	PCB85	2364.90738
BTEG02CP	97-192	B.Trout Whole	PCB85	781.81935
BTEG02CP	97-192	B.Trout Whole	PCB180	41.81165
BTUG03CP	97-192	B.Trout Whole	PCB85	285.18549
BTUG03CP	97-192	B.Trout Whole	PCB180	23.08871
EGLMF01WC-1	97-192	L.Trout Whole	PCB85	2613.14572
EGLMF01WC-1	97-192	L.Trout Whole	PCB180	127.56807
EGLMF02WC-1	97-192	L.Trout Whole	PCB85	2701.26525
EGLMF02WC-1	97-192	L.Trout Whole	PCB180	159.63264
EGLMF03WC-1	97-192	L.Trout Whole	PCB85	3452.65478
EGLMF03WC-1	97-192	L.Trout Whole	PCB180	221.47521
EGLMF04WC-1	97-192	L.Trout Whole	PCB85	2614.91961
EGLMF04WC-1	97-192	L.Trout Whole	PCB180	133.07266
EGLMF05WC-1	97-192	L.Trout Whole	PCB85	2079.80254
EGLMF05WC-1	97-192	L.Trout Whole	PCB180	226.42978
EGLMF06WC-1	97-192	L.Trout Whole	PCB85	2629.62183
EGLMF06WC-1	97-192	L.Trout Whole	PCB180	129.35301
EGLMF07WC-1	97-192	L.Trout Whole	PCB85	2129.52628
EGLMF07WC-1	97-192	L.Trout Whole	PCB180	179.67450
EGLMF08WC-1	97-192	L.Trout Whole	PCB85	2508.76791
EGLMF08WC-1	97-192	L.Trout Whole	PCB180	119.99790
EGLMF11WC-1	97-192	L.Trout Whole	PCB85	1213.53804
EGLMF11WC-1	97-192	L.Trout Whole	PCB180	59.45960
EGLMF12WC-1	97-192	L.Trout Whole	PCB85	2141.05995
EGLMF12WC-1	97-192	L.Trout Whole	PCB180	140.56961
WEEG02CP	97-192	Walleye Whole	PCB85	947.90876

SAMPLE RESULTS QUALIFIED AS A RESULT OF DUPLICATE RPD OUTLIERS (J/UJ-9)

Client ID	Batch ID	Matrix	Parameter	Concentration (ng/g, wet)
WEEG02CP	97-192	Walleye Whole	PCB180	62.99027
WEEG09CP	97-192	Walleye Whole	PCB85	1987.90089
WEEG09CP	97-192	Walleye Whole	PCB180	131.83004
WEFR07CP	97-192	Walleye Whole	PCB85	1123.27999
WEFR07CP	97-192	Walleye Whole	PCB180	37.85564
WELG01CP	97-192	Walleye Whole	PCB85	1297.49659
WELG01CP	97-192	Walleye Whole	PCB180	104.00942
WELG06CP	97-192	Walleye Whole	PCB85	1619.34567
WELG06CP	97-192	Walleye Whole	PCB180	155.97690
WEUG02CP	97-192	Walleye Whole	PCB85	1243.30206
WEUG02CP	97-192	Walleye Whole	PCB180	82.86603
WEWG04CP	97-192	Walleye Whole	PCB85	399.66711
WEWG04CP	97-192	Walleye Whole	PCB180	43.91320
96KICT05	97-274	Tern Eggs	PCB126	1.26048
96KICT07	97-274	Tern Eggs	PCB126	0.81603
96KICT09	97-274	Tern Eggs	PCB126	0.92498
BTEG02CP	97-274	B.Trout Whole	PCB126	0.54150
EGLMF06WC-1	97-274	L.Trout Whole	PCB126	0.29679
TEKIB18	97-274	Tern Eggs	PCB126	0.67831
WEFR07CP	97-274	Walleye Whole	PCB126	0.86155
WEFR07CP	97-274	Walleye Whole	PCB126	0.47479
WELG06CP	97-274	Walleye Whole	PCB126	1.14246
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB31	9.64066
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB28	13.92704
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB52	25.58797
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB49	18.19334
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB47/75	13.86909
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB44	17.32018
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB42/37	4.51004
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB63	5.29990
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB74	19.84732
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB70/76	36.24314
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB66	50.37475
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB95	14.95517
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB91	8.18757
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB56/60	17.17525
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB92	11.21620
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB84	17.93270
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB101/90	41.16918
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB99	38.50616
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB83	5.11670
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB97	13.16928
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB87/115/81	15.33698
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB85	21.88658
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB110/77	46.25477
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB82	6.37565
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB151	6.34463
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB135/144	8.97465
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB107/147	10.85050

SAMPLE RESULTS QUALIFIED AS A RESULT OF DUPLICATE RPD OUTLIERS (J/UJ-9)

Client ID	Batch ID	Matrix	Parameter	Concentration (ng/g, wet)
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB149/123	28.73250
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB118	60.28390
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB131	8.47346
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB146	14.31541
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB153	73.38141
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB132	7.82117
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB105	28.44523
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB141/179	6.94632
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB176	7.56233
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB130	5.42227
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB138/160/163	76.54443
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB178	4.33121
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB187/182	19.25765
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB183	7.29970
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB128	13.23936
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB174	6.91948
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB177	6.37137
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB171/202	5.98787
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB156	7.94473
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB180	41.70070
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB170/190	9.80318
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB199	6.40219
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB203/196	6.10408
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB31	5.29588
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB28	7.55210
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB52	12.73544
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB49	9.05152
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB47/75	7.18207
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB44	8.07298
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB42/37	2.13729
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB63	3.52492
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB74	9.97517
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB70/76	17.39217
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB66	25.18594
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB95	7.24453
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB91	3.72247
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB56/60	9.00404
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB92	4.48948
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB84	8.53712
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB101/90	20.13333
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB99	18.71490
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB83	2.82290
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB97	6.03594
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB87/115/81	7.67239
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB85	6.56886
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB110/77	21.50539
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB82	3.05387
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB151	2.93704
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB135/144	4.14697

SAMPLE RESULTS QUALIFIED AS A RESULT OF DUPLICATE RPD OUTLIERS (J/UJ-9)

Client ID	Batch ID	Matrix	Parameter	Concentration (ng/g, wet)
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB107/147	4.98443
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB149/123	13.07534
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB118	27.92534
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB131	3.82946
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB146	6.65185
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB153	33.04066
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB132	3.44924
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB105	13.47929
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB141/179	3.04764
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB176	3.35918
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB130	2.48316
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB138/160/163	30.51557
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB178	1.68678
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB187/182	8.83485
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB183	3.15067
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB128	6.12500
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB174	2.94966
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB177	2.83081
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB171/202	2.51793
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB156	3.37980
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB180	18.11751
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB170/190	4.78763
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB199	2.88855
EGLMF01FC-1	97-306	Lake Trout Eggs	PCB203/196	2.71288
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB31	6.31117
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB28	9.36571
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB52	16.81621
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB49	12.26067
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB47/75	9.15484
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB44	10.98063
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB74	13.68577
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB70/76	23.31798
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB66	34.23498
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB95	9.31798
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB91	5.67490
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB56/60	11.04219
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB92	7.28528
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB84	10.80069
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB101/90	29.82540
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB99	24.86206
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB97	8.34002
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB87/115/81	10.24881
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB85	7.25395
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB110/77	28.51937
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB135/144	5.69289
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB107/147	6.10899
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB149/123	18.93913
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB118	37.14209
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB131	4.41097

SAMPLE RESULTS QUALIFIED AS A RESULT OF DUPLICATE RPD OUTLIERS (J/UJ-9)

Client ID	Batch ID	Matrix	Parameter	Concentration (ng/g, wet)
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB146	8.78261
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB153	43.08300
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB132	5.67095
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB105	17.04239
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB138/160/163	44.95573
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB187/182	12.71532
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB183	4.40287
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB128	8.26789
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB174	4.42233
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB177	4.05929
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB180	26.67698
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB170/190	7.64634
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB199	4.23775
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB203/196	3.97885
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB31	3.30519
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB28	4.64253
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB52	8.96023
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB49	6.77687
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB47/75	4.97825
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB44	5.48896
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB74	7.17638
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB70/76	12.02151
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB66	17.12695
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB91	3.05990
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB56/60	5.77021
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB92	4.01769
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB84	5.56916
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB101/90	15.76696
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB99	13.29935
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB97	4.48255
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB87/115/81	5.91721
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB85	4.08101
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB110/77	15.59464
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB135/144	2.91721
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB107/147	3.50430
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB149/123	10.47938
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB118	19.71631
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB131	2.62037
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB146	4.80787
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB153	24.76250
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB132	3.01015
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB105	9.38166
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB138/160/163	25.58620
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB187/182	7.15357
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB183	2.56356
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB128	4.38571
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB174	2.52370
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB177	2.23742
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB180	14.23953

SAMPLE RESULTS QUALIFIED AS A RESULT OF DUPLICATE RPD OUTLIERS (J/UJ-9)

Client ID	Batch ID	Matrix	Parameter	Concentration (ng/g, wet)
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB170/190	4.03807
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB199	2.44245
EGLMF08FC-1	97-306	Lake Trout Eggs	PCB203/196	2.32817
EGLMF10WC-1	97-306	L.Trout Whole	PCB31	31.75677
EGLMF10WC-1	97-306	L.Trout Whole	PCB28	43.99925
EGLMF10WC-1	97-306	L.Trout Whole	PCB52	87.26316
EGLMF10WC-1	97-306	L.Trout Whole	PCB49	54.65789
EGLMF10WC-1	97-306	L.Trout Whole	PCB47/75	50.54023
EGLMF10WC-1	97-306	L.Trout Whole	PCB44	45.83421
EGLMF10WC-1	97-306	L.Trout Whole	PCB63	28.05564
EGLMF10WC-1	97-306	L.Trout Whole	PCB74	76.95602
EGLMF10WC-1	97-306	L.Trout Whole	PCB70/76	115.54286
EGLMF10WC-1	97-306	L.Trout Whole	PCB66	168.86992
EGLMF10WC-1	97-306	L.Trout Whole	PCB95	44.75489
EGLMF10WC-1	97-306	L.Trout Whole	PCB91	26.90789
EGLMF10WC-1	97-306	L.Trout Whole	PCB56/60	49.20150
EGLMF10WC-1	97-306	L.Trout Whole	PCB92	26.76278
EGLMF10WC-1	97-306	L.Trout Whole	PCB84	59.24286
EGLMF10WC-1	97-306	L.Trout Whole	PCB101/90	154.70038
EGLMF10WC-1	97-306	L.Trout Whole	PCB99	135.20865
EGLMF10WC-1	97-306	L.Trout Whole	PCB97	49.66316
EGLMF10WC-1	97-306	L.Trout Whole	PCB87/115/81	56.95226
EGLMF10WC-1	97-306	L.Trout Whole	PCB85	50.13872
EGLMF10WC-1	97-306	L.Trout Whole	PCB110/77	130.58835
EGLMF10WC-1	97-306	L.Trout Whole	PCB82	21.43308
EGLMF10WC-1	97-306	L.Trout Whole	PCB151	25.89023
EGLMF10WC-1	97-306	L.Trout Whole	PCB135/144	31.79173
EGLMF10WC-1	97-306	L.Trout Whole	PCB107/147	35.38008
EGLMF10WC-1	97-306	L.Trout Whole	PCB149/123	108.41015
EGLMF10WC-1	97-306	L.Trout Whole	PCB118	204.25977
EGLMF10WC-1	97-306	L.Trout Whole	PCB131	20.74774
EGLMF10WC-1	97-306	L.Trout Whole	PCB146	53.16015
EGLMF10WC-1	97-306	L.Trout Whole	PCB153	270.65526
EGLMF10WC-1	97-306	L.Trout Whole	PCB132	37.74962
EGLMF10WC-1	97-306	L.Trout Whole	PCB105	87.66429
EGLMF10WC-1	97-306	L.Trout Whole	PCB141/179	21.55301
EGLMF10WC-1	97-306	L.Trout Whole	PCB176	23.41654
EGLMF10WC-1	97-306	L.Trout Whole	PCB130	18.09248
EGLMF10WC-1	97-306	L.Trout Whole	PCB138/160/163	228.50902
EGLMF10WC-1	97-306	L.Trout Whole	PCB178	19.40902
EGLMF10WC-1	97-306	L.Trout Whole	PCB187/182	86.18722
EGLMF10WC-1	97-306	L.Trout Whole	PCB183	33.90677
EGLMF10WC-1	97-306	L.Trout Whole	PCB128	43.90038
EGLMF10WC-1	97-306	L.Trout Whole	PCB174	29.28872
EGLMF10WC-1	97-306	L.Trout Whole	PCB177	26.36015
EGLMF10WC-1	97-306	L.Trout Whole	PCB171/202	20.88120
EGLMF10WC-1	97-306	L.Trout Whole	PCB156	28.97030
EGLMF10WC-1	97-306	L.Trout Whole	PCB180	152.99774
EGLMF10WC-1	97-306	L.Trout Whole	PCB170/190	41.72707

SAMPLE RESULTS QUALIFIED AS A RESULT OF DUPLICATE RPD OUTLIERS (J/UJ-9)

Client ID	Batch ID	Matrix	Parameter	Concentration (ng/g, wet)
EGLMF10WC-1	97-306	L.Trout Whole	PCB199	26.92293
EGLMF10WC-1	97-306	L.Trout Whole	PCB203/196	28.33609

APPENDIX C
PROCEDURE AND RESULTS OF WATERFOWL COLLECTION BY USFWS IN THE ASSESSMENT AREA, 1997

Standard Operating Procedure for Collection, Preparation, Transport and Storage of Samples

Report by Dr. T. Custer et al. to USFWS, Green Bay Office

STANDARD OPERATING PROCEDURE FOR THE COLLECTION, PREPARATION, TRANSPORT, AND STORAGE OF WATERFOWL CARCASSES FROM GREEN BAY, WISCONSIN

1. INTRODUCTION AND STUDY OBJECTIVES

This Standard Operating Procedure (SOP) contains the objectives, methods, and approaches for the collection, preparation, transport, and storage of waterfowl carcasses to be collected from Green Bay, Wisconsin, for the Fox River/Green Bay Natural Resouree Damage Assessment (NRDA). Waterfowl tissues will be analyzed for contaminants by an analytical laboratory. A subsequent SOP will describe the laboratory analytical methods that will be employed.

The objective of the study is to:

- determine organochlorine concentrations in carcasses and breast muscle tissue of waterfowl breeding and wintering in Green Bay and the lower Fox River, Wisconsin.

Adult waterfowl will be collected during two periods in the winter of 1997/1988 (September/October and October/November), and during the 1988 nesting season and will be analyzed for organochlorines, including PCBs. The field team leader for the collections will be Dr. Thomas Custer (U.S. Geological Survey, Upper Mississippi Science Center, LaCrosse, WI).

2. FIELD PROCEDURES

2.1 WATERFOWL COLLECTION LOCATIONS

During the winter of 1997, waterfowl distribution and abundance will be measured through aerial surveys of the Fox River and Green Bay by Wiseonsin DNR. Local hunters may also assist in identifying suitable areas to collect waterfowl.

2.2 WATERFOWL COLLECTION

A variety of collection methods may be employed. These include but may not be limited to 1) shooting from a fast-moving boat, 2) shooting from a skull boat, 3) jump shooting birds from

shore, 4) or hunting from a blind or lay-out boat. The team leader or his representative will be present during all collections.

The species and numbers of birds that will be collected are shown in Table 1.

Table 1
Proposed waterfowl sampling effort for winter 1997
Numbers actually collected may be lower and/or species distributions may change, depending on availability of birds.

	Number to be Collected		Number to be Analyzed			
			Sept/Oct		Oct/Nov	
Species	**Sept/Oct**	**Oct/Nov**	**Carcass***	**Breast**	**Carcass***	**Breast**
Lesser Scaup	10	10	3	10	3	10
Common Goldeneye	0	10	0	0	3	10
Red-breasted Merganser**	0	10	0	0	3	10
Mallard**	5	0	3	5	0	0

* Carcass samples will be randomly selected from among the total sample.
** Mallards and mergansers (10 each) will also be collected in the summers of 1997 and 1998.
Summary: Maximum of 80 samples for OC analyses

On collection, each bird will be given a unique numerical identifier in the field. This number will be written on a tag and the tag tied to one leg. All identification numbers will be recorded in the field logbook. The identification system for waterfowl samples collected for contaminant analyses consists of the following code:

WF-XX-YY-00

where:

- **WF** is a two-letter code designating the waterfowl collection effort.
- **XX** is a unique two-letter code designating the collection location
- **YY** is a waterfowl species identifier (e.g.: LS = lesser scaup, etc)
- **00** is a unique two-number code designating the number assigned to this individual. Waterfowl will be numbered starting at "01."

Once uniquely identified, each bird will be placed in separate self-sealing plastic bags for transport to the USFWS Field Office in Green Bay.

2.3 FIELD DOCUMENTATION

The field team will document its sampling activities and field measurements in a dedicated, paginated, bound field logbook. Sampling locations will be clearly identified on photocopies of appropriate topographical maps and described in the field notebook. Entries in the field notebook and map marking will be done with waterproof ink, and corrections will be made with a single line through the error accompanied by the correction date and corrector's initials. The field team leader will be responsible for maintenance and proper archiving of these field notebooks.

The following information will be recorded in the field logbooks:

- site and project name
- each sampler's name and professional affiliation
- date and time of collection, field activity, or field measurement
- exact location of collection
- method of collection
- identification numbers of samples collected
- number and type of samples collected
- any difficulties encountered or necessary deviations from this SOP
- any other pertinent field observations.

Maps will be marked with a sampling location code, e.g., KI for Kidney Island, written within a circle. The field notebook page number corresponding to each sampling location will be marked adjacent to the sampling location circle.

Upon completion of each day's field activities, the notes will be reviewed by the field recorder and sampler and any necessary corrections made. The field recorder will sign and date each page.

2.4 PROCESSING AND STORAGE OF WATERFOWL TISSUES

The field team leader or a designated representative will transport the waterfowl to the USFWS Field Office in Green Bay. Immediately on returning from the field to the laboratory, the birds will be weighed and wing length measured. Measurements will be made using an electronic balance and a ruler and will include:

- wing length (to the closest 1.0 mm).
- weight (to the closest 0.1g).

- sex.
- age.

These measurements will be recorded in the field notebook.

After the above measurements are taken, the birds will be plucked, the contents of the esophagus, proventriculus and gizzard removed. The right side of the breast and associated skin will then be surgically removed.

After each dissection, the surgical equipment and the cutting board or table surface on which the dissections take place will be decontaminated according to the following procedure:

- pre-wash, using deionized water and scrub brush as necessary
- rinse thoroughly with ultra-clean acetone
- rinse thoroughly with ultra-clean hexane
- rinse again with ultra-clean acetone
- rinse thoroughly three times with deionized water.

The breast and associated skin will be weighed and wrapped in aluminum foil and sealed in an individual plastic bag. The remainder of the carcass will be weighed and wrapped in aluminum foil and sealed in an individual plastic bag. The letter 'M' for muscle or a 'C' for carcass (see below) will be attached to the labels as appropriate. The samples will be stored in a freezer before shipment to the analytical laboratory. The final identification system for waterfowl samples collected for contaminant analyses consists of the following code:

WF-XX-YY-00-T

where:

- T is a one-letter code designating the waterfowl tissue (C = carcass, M = breast muscle)

2.5 CHAIN OF CUSTODY

The chain of custody will start when waterfowl are collected. Each bird will be given a unique numerical identifier in the field. This number will be written on a tag and the tag attached to the carcass. Once identified in this way, the waterfowl collected during each sampling event will be placed (each sample within its own self-sealing plastic bag) in a communal container under the custody of Dr. Tom Custer or a designated stand-in. Each of the self sealing plastic bags will be labeled with the appropriate sample identifier. The bags will be stored frozen in one or more shipping containers which will be sealed with custody seals (to detect unauthorized tampering

with samples after sample collection until the time of use or analysis), and contain chain of custody forms with the following information, as appropriate:

- project name
- waterfowl identifiers (unique for each sample)
- name and signature of field recorder
- date and time of beginning of sample collection
- chain of custody seal number
- signatures of persons involved in the chain of possession
- inclusive dates and times of possession
- method and date of sample shipment.

At the appropriate time, the entire sealed container(s) will be shipped to the analytical laboratory.

The designated field sample eustodian will be personally responsible for the care and custody of the samples until they are transferred or properly dispatched. A sample is in the custody of an individual if any of the following occur:

- The sample is in the individual's possession.
- The sample is within view after being in possession.
- The sample is in a locked or sealed container that prevents tampering after being in possession.
- The sample is in a designated secure area.

Every transfer of custody will be noted with the date and time of transfer and signed for on the chain of custody record. The number of custody transfers will be kept to a minimum.

2.7 FIELD EQUIPMENT

The following list of equipment will be required in the field:

- SOPs (one copy for each team member)
- waders/hip boots (all crew members)
- field log books
- marking pens and pencils
- labels and labeling tape
- string
- self-sealing plastic bags
- chain of custody forms and seals
- shotguns and shells (steel shot)

2.8 DEVIATIONS FROM THIS SOP

If field conditions necessitate any deviations from this SOP the Field Team Leader will document them in the field note book and in an addendum to this SOP.

Concentrations of polychlorinated biphenyls in tissues of waterfowl from Green Bay, Wisconsin and nearby Lake Michigan

Thomas W. Custer [1], Christine M. Custer[1], Lynn A. Bartsch[1], and Kenneth L. Stromborg[2],

[1] U. S. Geological Survey, Upper Midwest Environmental Sciences Center,

2630 Fanta Reed Road, La Crosse, Wisconsin 54603

[2] U.S. Fish and Wildlife Service, 1015 Challenger Ct.,

Green Bay, Wisconsin 54311

Introduction

Green Bay is contaminated with polychlorinated biphenyls (PCBs), most of which reportedly originated from the deinking and repulping of carbonless paper at paper mills on the Fox River (Fig. 1) (Sullivan *et al.* 1983). Elevated PCB concentrations have been documented in Green Bay sediment (Sullivan *et al.* 1983, Hermanson *et al.* 1991, Ankley *et al.* 1992, Velleux and Endicot 1994, Manchester-Neesvig *et al.* 1996), fish (Sullivan *et al.* 1983), and birds (Ankley *et al.* 1993, Custer and Custer 1995, Harris *et al.* 1993, Rattner *et al.* 1993, Hoffman *et al.* 1993, Kubiak *et al.* 1989, Custer *et al.* 1998, Custer *et al.* 1999). The Wisconsin Department of Natural Resources (WDNR) has issued a consumption advisory on mallards (*Anas platyrhynchos*) obtained from Green Bay, Wisconsin because of high levels of PCBs in their tissues.

Zebra mussels (*Dreissena polymorpha*) have reached high densities in the Great Lakes, including Green Bay, since their introduction in the mid 1980s. Densities of zebra mussels over 700,000/m^2 have been reported at power plants on Lake Erie (Kovalak *et al.* 1993) and as many as 342,000m^2 on fish-spawning reefs in Lake Erie (Leach 1993). Zebra mussel biomass can be as high as 3.6 kg/m^2 (Custer and Custer 1997). The bioaccumulation capacities of zebra mussels (Brieger and Hunter 1993, Busch and Schuchardt 1991, Mersch *et al.* 1992) may enhance the transfer of contaminants to waterfowl (de Kock and Bowmer 1993). Contaminants, if high enough, can negatively affect waterfowl reproduction (de Kock and Bowmer 1993) or may have secondary effects as a contaminant source for Bald Eagles (*Haliaeetus leucocephalus*), other raptors, and humans.

Waterfowl are now migrating through and wintering in parts of the Great Lakes in larger numbers than they had immediately prior to the zebra mussel invasion (Wormington and Leach 1992). This increase has probably been due to the presence of zebra mussels, a now abundant and easily captured food source.

Zebra mussels are the primary food now for lesser scaup (*Aythya affinis*) and common goldeneye (*Bucephala clangula*) in the Great Lakes, especially in western Lake Erie (Custer and Custer 1996, Hamilton *et al.* 1994). Ninety-eight percent of lesser scaup diet, 79% of common goldeneye diet, 24% of bufflehead (*Bucephala albeola*) diet, but < 10% of canvasback (*Aythya valisineria*) diet are now zebra mussels (Custer and Custer 1996). The consequences of this food shift are mostly unknown, however, the potential for contaminant transfer may be high. The Great Lakes are an area of known contamination (Government of Canada 1991). Diving ducks collected in the Detroit River in 1980 had high organochlorine concentrations (Smith *et al.* 1985). Chlorinated hydrocarbon contaminants were still present in waterfowl from the Detroit River in the early 1990s (Mazak *et al.* 1997).

Human consumption advisory levels for PCB concentrations in edible poultry are available for Canada (0.5 µg/g lipid weight, Health and Welfare Canada 1991) and the United States (3.0 µg/g lipid weight, FDA 1979). Furthermore, PCB concentrations can be compared to the 'do not eat' category (1.9 µg/g wet weight) under proposed guidelines for a uniform Great Lakes sport fish consumption advisory (Anderson *et al.* 1993).

The objective of the study was to determine whether PCB concentrations in tissues of waterfowl breeding and wintering in Green Bay, Wisconsin exceeded human consumption advisory levels.

Methods

Waterfowl were collected by shotgun using steel shot in Green Bay and Lake Michigan during June to November 1997 under appropriate state and federal collecting permits. After collection, the birds were weighed (0.1 g) in the laboratory and in the case of lesser and greater scaup the wing length (1.0 mm) was measured. The breast of the birds was plucked and the right side of the breast and associated skin were then surgically removed. The breast and associated skin were individually weighed, wrapped in aluminum foil, sealed in an individual plastic bag, and frozen at -20 °C. Age and sex of waterfowl was determined using plumage and cloacal characteristics (Carney 1964). The remainder of the carcass was weighed, wrapped in aluminum foil, sealed in an individual plastic bag, and frozen at -20 °C.

The following organochlorines were analyzed in waterfowl muscle and skin samples by Mississippi State Chemical Laboratory, Mississippi State, Mississippi, USA: α-, β-, γ- and δ-hexachlorocyclohexane (HCH); α- and δ- chlordane; oxychlordane; *cis*-nonachlor; *trans*-nonachlor; dieldrin; endrin; hexachlorobenzene (HCB); heptachlor epoxide; mirex; toxaphene; o,p'-dichlorodiphenyldichloroethane(DDD); o,p'-dichlorodiphenyldichloroetylene (DDE); o,p'-dichlorodiphenyltrichloroethane (DDT); p,p'-DDD; p,p'-DDE; p,p'-DDT; and total PCBs. Samples were homogenized, mixed with sodium sulfate and soxhlet extracted with hexane. After the lipid determination, lipids were removed by florisil column chromatography. Following silicic acid column chromatography, pesticides and total PCBs were determined by electron capture gas chromatography. Total PCBs were estimated based on Aroclor equivalents. The nominal limit of detection for organochlorines 0.01 μg/g wet weight, except for mallards which was 0.02 μg/g. The number of spikes, duplicates and blanks was 10% of the total number

of samples analyzed. Concentrations were not adjusted for recovery which averaged 90% for all organochlorines. Organochlorine concentrations in breast muscle without skin, skin associated with the breast muscle, and breast muscle with skin are reported on a wet weight and lipid weight basis. Breast muscle from all waterfowl collected were analyzed for organochlorines. Because of budgetary constraints, not all the skins associated with breast muscle were analyzed for organochlorines.

Results and Discussion

Waterfowl were collected from three locations in Green Bay and Lake Michigan in 1997 (Fig. 1). For mallards collected in June (n=10, Tables 1 and 2) breast muscle of 5, 4, and 0 birds were above the Canadian PCB consumption advisory (0.5 µg/g lipid weight, Health and Welfare Canada 1991), United States PCB consumption advisory (3.0 µg/g lipid weight, FDA 1979), and the Great Lakes sport fish consumption advisory (1.9 µg/g wet weight, Anderson *et al.* 1993), respectively. When skin was added to the muscle, all 10 samples were above the Canadian criteria, 8 were above the United States criteria, and none were above the Great Lakes sport fish consumption advisory (Table 1). We suspect that the mallards were resident individuals that had nested earlier near or in southern Green Bay. This conclusion is based on the collection date (June 12th) which is earlier than the Fall migration. Additionally, many of the birds collected were paired.

One lesser scaup was obtained during the June 12th collection (Table 2). We suspect that this individual was injured or sick and did not migrate in the fall of 1996. If that individual was a resident in Green Bay, PCB concentrations in tissues suggest that >8 months (September 1996

to June 1997) exposure to contaminants from prey items in Green Bay brought it's muscle PCB concentrations above the Canadian and United States PCB poultry consumption advisories. Concentrations of PCBs in the breast muscle alone did not exceed the Great Lakes sport fish consumption advisory. However, when the breast muscle of this individual was analyzed with the associated skin, PCB concentrations did exceed the Great Lakes sport fish consumption advisory.

The results suggest limited PCB exposure to hunters consuming migrating diving ducks shot near Point au Sable, especially if breast muscle is consumed without skin attached. PCB concentrations in breast muscles of only two of 34 diving ducks collected from Point au Sable during October and November (Tables 1, 3, and 4) were above Canadian consumption guidelines, United States consumption guidelines, and the Great Lakes sport fish consumption advisory. When skin was added to the muscle (n=23), 13 samples were above the Canadian consumption guidelines, 4 above United States consumption guidelines, and none above the Great Lakes sport fish consumption advisory. The data suggest that the time period from arrival of diving ducks in Green Bay until collection (late-October to mid-November 1997) was too short to allow significant accumulation of PCBs.

Based on United States PCB consumption guidelines for poultry, mergansers shot in Lake Michigan in northern Door County should not be eaten. Of 14 diving ducks collected in Lake Michigan near the northern end of Door County in September and November, the breast muscle of 13 were above Canadian and United States consumption guidelines (Tables 1, 5, and 6). One individual was above the Great Lakes sport fish consumption advisory. Based on actively growing flight feathers, this immature female common merganser was raised locally.

Concentrations of total PCBs in muscle with skin attached are probably representative of PCB concentrations in whole carcasses. The ratio of PCB wet weight in muscle with skin to PCB wet weight in muscle without skin averaged 4.2 (range 1.5 to 7, n=8). This is very similar to the PCB breast muscle to carcass ratio (mean = 4.1, range =3.3 to 4.8) of sentinel mallards measured in another study (Custer *et al.* 1996).

Conclusions

These results suggest that resident waterfowl in Green Bay accumulate PCBs to concentrations above the human consumption advisory for poultry in Canada and the United States. Tissues of migrating diving ducks shot in early fall and winter in Green Bay are generally not above human consumption advisory levels for PCBs. Based on PCB concentrations in tissues, mergansers shot in Lake Michigan near Door County should not be eaten.

Acknowledgments

We thank Joel A. Trick for field assistance Paul Dummer for assistance with data analysis. This study was funded by the U.S. Fish and Wildlife Service, Green Bay Natural Resource Damage Assessment.

References

Anderson, H. A., J. F. Amrhein, P. Shubat, and J. Hesse. 1993. Protocol for a uniform Great Lakes sport fish consumption advisory. Great Lakes Fish Advisory Task Force. 81 pp

Ankley, G. T., K. B. Lodge, D. J. Call, M. D. Balcher, L. T. Brooke, P. M. Cook, R. G. Kreis, A. R. Carlson, R. D. Johnson, G. J. Niemi, R. A. Hoke, C. W. West, J. P. Giesy, P. D. Jones, and Z. C. Fuying. 1992. Integrated assessment of contaminated sediments in the lower Fox River and Green Bay, Wisconsin. Ecotoxicol Environ Safety 23:46-63.

Ankley, G. T., G. J. Niemi, K. B. Lodge, H. J. Harris, D. L. Beaver, D. E. Tillitt, T. R. Schwartz, J. P. Giesy, P. D. Jones and C. Hagley. 1993. Uptake of planer polychlorinated biphenyls and 2,3,7,8-substituted polychlorinated dibenzofurans and dibenzo-p-dioxins by birds nesting in the lower Fox River and Green Bay, Wisconsin, USA. Arch. Environ. Contam. Toxicol. 24:332-344.

Brieger, G., and R. D. Hunter. 1993. Uptake and depuration of PCB 77, PCB 169, and hexachlorobenzene by zebra mussels (*Dreissena polymorpha*). Ecotoxicol. Environ. Safety 26:153-165.

Busch, D., and B. Schuchardt. 1991. The use of the freshwater mussel Dreissena polymorpha (Pallas) for biomonitoring heavy metals in limnic ecosystems: the Weser (FRG). Verh. Int. Verein. Limnol. 24:2261-2262.

Carney, S. M. 1964. Prelininary keys to waterfowl age and sex identification by means of wing plumage. U. S. Fish and Wildl. Serv. Spec. Sci. Rpt. 82.

Custer, C. M., and T. W. Custer. 1996. Food habits of diving ducks in the Great Lakes after the zebra mussel invasion. J. Field Ornithol. 67:86-99.

Custer, C. M., and T. W. Custer. 1997. Occurrence of zebra mussels in near-shore areas of western Lake Erie. J. Great Lakes Res. 23:108-115.

Custer T. W., and C. M. Custer. 1995. Transfer and accumulation of organochlorines from black-crowned night-heron eggs to chicks. Environ Toxicol Chem 14:533-536.

Custer, T. W., D. W. Sparks, S. A. Sobiech, R. K. Hines and M. J. Melancon. 1996. Organochlorine accumulation by sentinel mallards at the Winston-Thomas sewage treatment plant, Bloomington, Indiana. Arch. Environ.Contamin. Toxicol. 30:163-169

Custer C. M., T. W. Custer, P. D. Allen, K, L. Stromborg, and M. J. Melancon. 1998. Reproduction and environmental contamination in tree swallows nesting in the Fox River drainage and Green Bay, Wisconsin, USA. Environ Toxicol Chem. 17:1786-1798.

Custer, T. W., C. M. Custer, R. K. Hines, S. Gutreuter, K. L. Stromborg, P. D. Allen, and M. J. Melaneon. 1999. Organochlorine contaminants and reproductive success of double-crested cormorants from Green Bay, Wisconsin, USA. Environmental Toxicology and Chemistry. In press.

de Kock, W. C., and C. T. Bowmer. 1993. Bioaccumulation, biological effects, and food chain transfer of contaminants in the zebra mussel (*Dreissena polymorpha*). Pp. 503-533, in T. F. Nalepa and D. W. Schloesser. eds. Zebra mussels biology, impacts, and control. Lewis Publishers, Boca Raton, Florida.

FDA, United States Food and Drug Administration (1979) Polychlorinated biphenyls (PCBs); reduction of tolerances. Fed Reg 49(Jun):38330-38340

Government of Canada. 1991. Toxic Chemicals in the Great Lakes and Associeated Effects. Vol. 1. Contaminant Levels and Trends. Environment Canada. Dept. fisheries and Oceans, Health and Welfare Canada.

Hamilton, D. J., D. Ankney, and R. C. Bailey. 1994. Predation of zebra mussels by diving ducks: an exclosure study. Ecology 75:521-531.

Harris, H. J., T. C. Erdman, G. T. Ankley, and K. B. Lodge. 1993. Measures of reproductive success and polychlorinated biphenyl residues in eggs and chicks of Forster's terns on Green Bay, Lake Michigan, Wisconsin-1988. Arch Environ Contam Toxicol 25:304-314.

Health and Welfare Canada (1991) Table 2, Division 15, Canadian Food and Drug Regulations

Hermanson M. H., E. R. Christensen, D. J. Buser, and S. M. Chen. 1991. Polychlorinated biphenyls in dated sediment cores from Green Bay and Lake Michigan. J Great Lakes Res 17:94-108.

Hoffman D. J., G. J. Smith, and B. A. Rattner. 1993. Biomarkers of contaminant exposure in common terns and black-crowned night herons in the Great Lakes. Environ Toxicol Chem 12:1719-1732.

Kovalak, W. P., G. D. Longton, and R. D. Smithee. 1993. Infestation of power plant water systems by the zebra mussel (*Dreissena polymorpha Pallas*). Pp. 359-380, in T. F. Nalepa and D. W. Schloesser, eds. Zebra mussels biology, impacts, and control. Lewis Publishers, Boca Raton, Florida.

Kubiak T. J., H. J. Harris, L. M. Smith, T. R. Schwartz, D. L. Stalling, J. A. Trick, L. Sileo, D. E. Docherty, and T. C. Erdman. 1989. Microcontaminants and reproductive impairment of the Forster's tern on Green Bay, Lake Michigan-1983. Arch Environ Contam Toxicol 18:706-727.

Leach, J. H. 1993. Impacts of the zebra mussel (*Dreissena polymorpha*) on water quality and fish spawning reefs in the western Lake Erie. Pp. 381-397, in T. F. Nalepa and D. W. Schloesser, eds. Zebra mussels biology, impacts, and control. Lewis Publishers, Boca Raton, Florida.

Manchester-Neesvig J. B., A. W. Andren, and D. N. Edgington. 1996. Patterns of mass sedimentation and of deposition of sediment contaminated by PCBs in Green Bay. J Great Lakes Res 22:444-462.

Mazak, E. J., H. J. MacIsaac, M. R. Servos, and R. Hesslein. 1997. Influence of feeding habits on organochlorine contaminant accumulation waterfowl on the Great Lakes. Ecol. Applic. 7:1133-1143.

Mersch, J., A. Jeaujean, H. Spor, and J. C. Pihan. 1992. The freshwater mussel Dreissena polymorpha as a bioindicator for trace elements, organochlorines and radionuclides . Limnologie Aktuell 4:227-244.

Rattner, B. A., M. J. Melancon, T. W. Custer, R. L. Hothem, K. A. King, L. J. LeCaptain, J. W. Spann, B. R. Woodin, and J. J. Stegeman. 1993. Biomonitoring environmental contamination with pipping black-crowned night heron embryos: Induction of cytochrome P450. Environ. Toxicol. Chem. 12:1719-1732.

Smith, V. E., J. M. Spurr, J. C. Filkins, and J. J. Jones. 1985. Organochlorine contaminants of wintering ducks foraging on Detroit River sediments. J. Great Lakes Res. 11:231-246.

Sullivan, J. R., J. J. Delfino, C. R. Buelow, and T. B. Sheffy. 1983. Polychlorinated biphenyls in the fish and sediment of the lower Fox River, Wisconsin. Bull Environ Contam Toxicol 30:58-64.

Velleux, M., and D. Endicott. 1994. Development of a mass balance model for estimating PCB export from the lower Fox River to Green Bay. J Great Lakes Res 20:416-434.

Wormington, A., and J. H. Leach. 1992. Concentrations of migrant diving ducks at Point Pelee National Park, Ontario, in response to invasion of zebra mussels, Dreissena polymorpha. Can. Field-Nat. 106:376-380.

Figure 1. Locations (hatched ellipses) in Green Bay and Lake Michigan where waterfowl were collected during June to November, 1997.

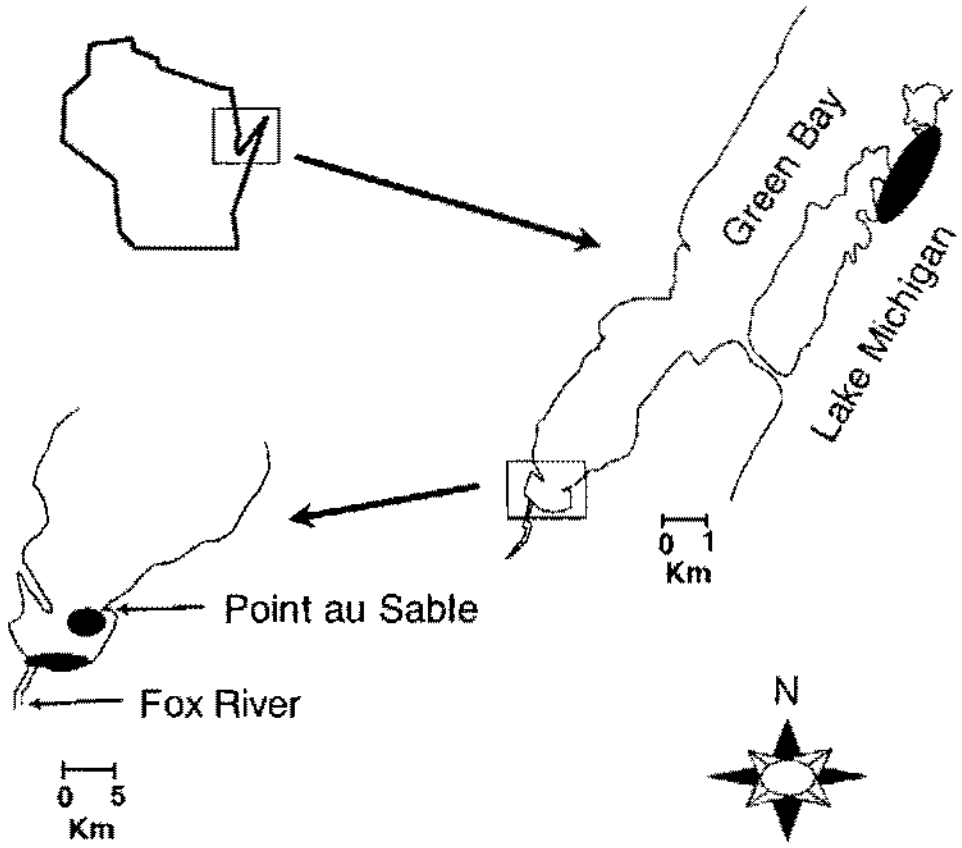


Table 1. Summary of the number of waterfowl collected near Green Bay, Wisconsin that exceeded PCB human consumption advisory levels for poultry in Canada (0.05 µg/g lipid weight), poultry in the United States (3.0 µg/g lipid weight), and fish in the Great Lakes (1.9 µg/g wet weight).

		No. of ducks with PCB concentrations exceeding human health criteria						
		Breast muscle			Breast muscle with skin attached			
Location	No.	Canada	U.S.	Great Lakes	No.	Canada	U.S.	Great Lakes
Southern Green Bay	10	5	4	0	10	10	8	0
Point au Sable	34	2	2	2	23	13	4	0
Door County	14	13	13	1	-- [1]	--	--	--

[1] -- = not measured

Table 2. PCB concetrations (µg/g lipid weight and µg/g wet weight) in skin and breast muscle of one lesser scaup and ten mallards collected in southern Green Bay on June 12, 1997. Level of detection was 0.02 µg/g wet weight.

				PCBs µg/g lipid weight			PCBs µg/g wet weight		
Cat.ID	Species	Sex	Age	Muscle	Skin	Skin + muscle	Muscle	Skin	Skin + muscle
GBMD01	Mallard	M[1]	A[2]	ND[3]	2.8	2.0	ND	1.0	0.1
GBMD02	Mallard	F	A	ND	3.3	2.2	ND	1.2	0.1
GBMD03	Mallard	F	A	ND	11.0	8.0	ND	3.4	0.3
GBMD04	Mallard	M	A	ND	6.2	4.5	ND	2.9	0.3
GBMD05	Mallard	M	A	15.0	21.2	19.5	0.2	6.6	0.8
GBMD06	Mallard	M	A	6.6	21.5	15.4	0.1	5.1	0.6
GBMD07	Mallard	M	A	13.9	18.7	17.4	0.2	5.2	0.8
GBMD08	Mallard	F	A	2.9	11.0	9.6	0.4	4.2	0.6
GBMD09	Mallard	M	A	ND	15.5	5.6	ND	1.8	0.2
GBMD10	Mallard	M	A	5.9	22.0	16.9	0.1	6.0	0.7
GBLS11	Lesser scaup	M	A	16.3	27.9	23.3	0.6	9.5	2.0

[1] M = male, F = female
[2] A = adult
[3] ND = indicates not detected

Table 3. PCB concetrations (µg/g lipid weight and µg/g wet weight) in skin and breast muscle of diving ducks collected from Point au Sable, southern Green Bay on October 27, 1997. Level of detection was 0.01 µg/g wet weight.

				PCBs µg/g lipid weight			PCBs µg/g wet weight		
Cat.ID	Species	Sex	Age	Muscle	Skin	Skin + muscle	Muscle	Skin	Skin + muscle
GBLS12	Greater scaup	M[1]	I[2]	ND[3]	2.0	1.6	ND	1.1	0.2
GBLS13	Greater scaup	F	I	ND	3.6	3.1	ND	1.7	0.3
GBLS14	Greater scaup	M	I	ND	1.2	0.8	ND	0.3	0.05
GBLS15	Greater scaup	F	I	ND	0.4	0.3	ND	0.2	0.03
GBLS16	Greater scaup	F	I	ND	2.6	2.2	ND	1.7	0.4
GBLS18	Greater scaup	M	I	ND	0.2	0.2	ND	0.2	0.04
GBLS17	Lesser scaup	F	A	ND	3.1	2.7	ND	1.9	0.4
GBLS19	Lesser scaup	M	I	ND	0.6	0.6	ND	0.5	0.2
GBCN20	Canvasback	M	I	ND	-[4]	-	ND	-	-
GBRD21	Ruddy duck	F	I	ND	-	-	ND	-	-
GBGE22	Common goldeneye	M	A	14.5	13.5	14.1	0.25	1.4	0.4

[1] M = male, F = female
[2] I = immature, A = adult
[3] ND = not detected
[4] - indicates no analysis

Table 4. PCB concetrations (µg/g lipid weight and µg/g wet weight) in skin and breast muscle of diving ducks collected from Point au Sable, southern Green Bay on November 12-13, 1997. Level of detection was 0.01 µg/g wet weight.

				PCBs µg/g lipid weight			PCBs µg/g wet weight		
Cat.ID	Species	Sex	Age	Muscle	Skin	Skin + muscle	Muscle	Skin	Skin + muscle
GBLS23	Greater scaup	M[1]	A[2]	ND[3]	3.8	3.3	ND	2.2	0.4
GBLS24	Lesser scaup	M	A	ND	2.6	2.4	ND	2.0	0.6
GBLS25	Lesser scaup	M	I	ND	1.3	1.2	ND	1.1	0.4
GBLS26	Lesser scaup	M	A	4.8	5.1	5.1	0.11	2.8	0.7
GBLS27	Lesser scaup	M	I	ND	3	2.5	ND	1.8	0.3
GBLS28	Lesser scaup	F	I	ND	1.5	1.4	ND	1.3	0.6
GBLS29	Lesser scaup	M	I	ND	0.4	0.4	ND	0.3	0.1
GBLS30	Lesser scaup	M	I	ND	0.9	0.8	ND	0.7	0.2
GBLS31	Lesser scaup	M	I	ND	1.0	0.9	ND	0.7	0.2
GBBH32	Bufflehead	F	I	ND	-[4]	-	ND	-	-
GBBH33	Bufflehead	F	I	ND	0.5	0.4	ND	0.4	0.1
GBBH34	Bufflehead	F	I	ND	-	-	ND	-	-
GBBH35	Bufflehead	F	I	ND	-	-	ND	-	-
GBBH36	Bufflehead	M	I	ND	-	-	ND	-	-
GBBH37	Bufflehead	M	I	ND	-	-	ND	-	-
GBBH38	Bufflehead	M	A	ND	1.8	1.4	ND	1.4	0.4
GBBH39	Bufflehead	M	I	ND	-	-	ND	-	-
GBRD40	Ruddy duck	F	A	ND	-	-	ND	-	-
GBGE41	Common goldeneye	F	I	ND	0.1	0.1	ND	0.04	0.01
GBGE42	Common Goldeneye	F	I	ND	1.6	1.5	ND	1.2	0.3
GBGE43	Common goldeneye	M	I	ND	0.1	0.1	ND	0.1	0.02
GBWS44	White-winged Scoter	F	I	ND	-	-	ND	-	-
GBWS45	White-winged scoter	F	I	ND	-	-	ND	-	-

[1] M = male, F = female
[2] A = adult, I = immature
[3] ND = not detected
[4] - indicates no analysis

Table 5. PCB concentrations (µg/g lipid weight and µg/g wet weight) in skin and breast muscle of diving ducks collected from Baileys Harbor and Newport Beach, Lake Michigan on September 16-17, 1997. Level of detection was 0.01 µg/g wet weight.

				PCBs µg/g lipid weight			PCBs µg/g wet weight		
Cat.ID	Species	Sex	Age	Muscle	Skin	Skin + muscle	Muscle	Skin	Skin + muscle
GBGE01	Common goldeneye	M[1]	A[2]	3.5	-[3]	0.2	-	-	
GBRD01	Ruddy duck	F	A	4.6	-	-	0.1	-	-
GBRM01	Red-breasted merganser	F	A	ND[4]	8.5	5.2	ND	2.6	0.5
GBRM02	Red-breasted merganser	F	I	25.3	-	-	1.0	-	-

[1] M = male, F = female
[2] A = adult, I = immature
[3] - indicates no analysis
[4] ND = not detected

Table 6. PCB concetrations (µg/g lipid weight and µg/g wet weight) in skin and breast muscle of diving ducks collected in northern Door County in Lake Michigan on September 22nd and September 26th, 1997. Level of detection was 0.01 µg/g wet weight.

				PCBs µg/g lipid weight			PCBs µg/g wet weight		
Cat.ID	Species	Sex	Age	Muscle	Skin	Skin + muscle	Muscle	Skin	Skin + muscle
GBPI49	Common merganser	M[1]	I[2]	8.3	-[3]	-	0.1	-	-
GBNP50	Common merganser	F	I	373.9	-	-	4.3	-	-
GBPI52	Common merganser	F	I	36.3	-	-	0.6	-	-
GBPI53	Common merganser	F	A	27.4	-	-	0.5	-	-
GBPI54	Common merganser	F	I	25.7	-	-	0.4	-	-
GBDI55	Common merganser	F	A	30.3	-	-	0.5	-	-
GBDI56	Common merganser	M	I	10.8	-	-	0.2	-	-
GBDI57	Common merganser	M	I	16.8	-	-	0.2	-	-
GBNP51	Red-breasted merganser	F	A	36.9	-	-	0.8	-	-
GBHI58	Red-breasted merganser	F	A	11.4	-	-	0.3	-	-

[1] M = male, F = female
[2] A = adult, I = immature
[3] – indicates no analysis