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European Starling (Sturnus Vulgaris): Avian Model and Monitor of Contaminant and Remedial Effects at Crab Crab Orchard National Wildlife Refuge

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RR-E84
January 1998
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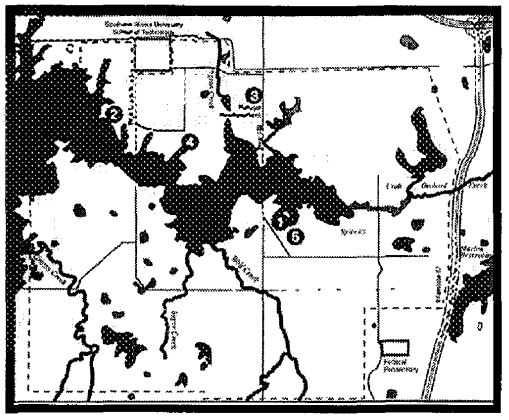

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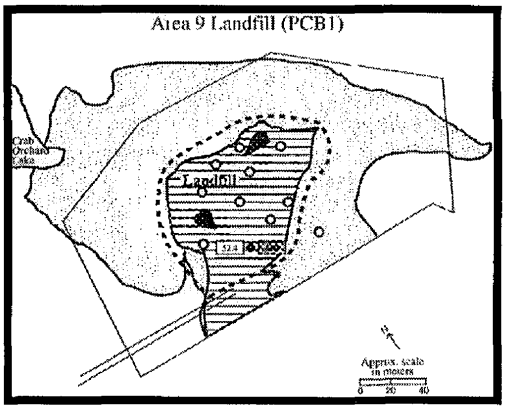

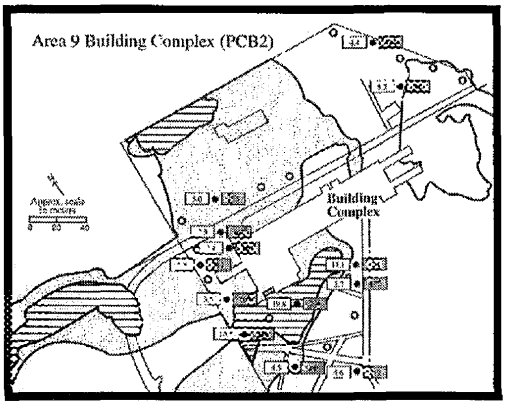

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Submitted to the
Illinois Waste Management and Research Center
One E. Hazelwood Drive
Champaign, Illinois 61820

Printed January 1998

Printed by the Authority of the State of Illinois
98/150

ACKNOWLEDGMENTS

We would like to thank MaryJo Woodruff, Hui-Lin Jun, and the analytical staff at the Cooperative Wildlife Research Laboratory for their laboratory and field assistance with this project. We also would like to thank Mark Sattelberg and the staff at Crab Orchard National Wildlife Refuge, Ron Hliwa at Olin Corporation, Inc., and Robert Hammack at Flour Daniel GTC for assistance in coordinating study efforts on the refuge.

Financial support for this project was provided by the Waste Management and Research Center of the Illinois Department of Natural Resources Project #94-120. Additional support was provided by the Cooperative Wildlife Research Laboratory, the Zoology Department, and the Graduate School at Southern Illinois University at Carbondale.

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ABSTRACT

Accumulation and effects of polychlorinated biphenyl (PCB) and heavy metal concentrations in avian species were evaluated at a Superfund site located at Crab Orchard National Wildlife Refuge, Illinois. European starlings (*Sturnus vulgaris*) were monitored at 12 nest boxes constructed at each of 4 study and 2 reference sites. Adult starlings were observed in the field to assess possible contaminant effects on nest attentiveness behavior, and the number of eggs and chicks surviving to 15 days per nest were recorded. Fifteen-day-old chicks were necropsied and kidney tissue was analyzed for mercury, cadmium, chromium, and lead concentrations. Ethoxyresorufin-O-deethylase (EROD) activity was measured in liver tissue, and PCB (Aroclor 1254) and 34 chlorinated biphenyl congener concentrations were measured in whole body carcass samples using gas chromatography. Differences ($P < 0.05$) among study and reference sites were found for Aroclor 1254, quantified CBs, and EROD activity. Pre-remediation cadmium concentrations at the metals site differed from concentrations at reference sites, but post-remediation cadmium concentrations did not. Effects included a reduction ($P < 0.05$) in nest attentiveness behavior and increased chick mortality between PCB and reference sites. Nest attentiveness behavior also was reduced at the metals site compared to reference sites. There were no differences among study and reference sites in number of eggs laid and percent of eggs hatched. Greater tissue concentrations of contaminants in starlings collected at study sites compared to those collected at reference sites suggest that other avian species utilizing these sites may also have greater body burdens of metals and PCBs, and may suffer adverse nesting and behavior effects demonstrated similar to those observed in this study. This study also demonstrates that starling nestlings are good indicators of local contamination, as well as, of the effectiveness of remediation. Additional research is needed to further evaluate some effects identified in the current study including: determination of adult versus chick contribution to decreased feeding activity, objective measurement of the increased boldness observed in adults at contaminated sites, and investigation into environmental contaminants near the Annex reference site as potential causes of observed reductions in hematocrit values and increases in liver weights at that site.

EXECUTIVE SUMMARY

The 1988 Remedial Investigation (RI) of Crab Orchard National Wildlife Refuge (CONWR) indicated that past manufacturing activities had contaminated specific areas within the Refuge with polychlorinated biphenyls (PCBs) and various metals. The RI identified 7 sites as posing a potential risk to wildlife and recommended them for remedial action. The identified sites were separated into PCB and Metals Operable Units (OU) based on the major contaminants of concern. Potential risks to wildlife were considered high at the Job Corps Landfill (PCB OU) and Old Refuge Shop (Metals OU) and moderate at the Area 9 Landfill (PCB OU) and Area 9 Building Complex (PCB OU). Exposure and effects data were available at some of these sites for mammalian species, however, only limited data were available for avian species. Preliminary studies revealed statistically significant accumulations of contaminants and effects in starlings (*Sturnus vulgaris*) nesting at one of these sites.

The goal of this project was to provide pre-remediation base-line exposure and effects data on an avian model, the starling, and to provide a benchmark for assessing the effectiveness of remediation. The objectives were (1) to measure PCB and selected metal (cadmium, chromium, lead, and mercury) exposure, (2) to determine physiological, reproductive, and biomarker effects, and (3) to monitor nest attentiveness and abandonment behavior in starlings at the 4 contaminated sites identified as posing moderate to high risk to wildlife. Prior to the beginning of the nesting season, 12 nest boxes were located at each of the 4 contaminated sites and at 2 reference sites. During the breeding season, starling productivity (eggs/nest, chicks/nest, chicks surviving to 15 days) and adult nest attentiveness behavior (incubation, chick feeding, and abandonment behavior) were monitored. At 15 days post-hatch, chicks and adults were collected for contaminant and biomarker analyses. Eggs that failed to hatch also were collected. Differences in PCB concentrations in eggs, liver, and homogenized carcasses, differences in metal concentrations in eggs, kidney and feathers, and behavior differences in adults were compared among sites. In addition, liver ethoxyresorufin-O-deethylase (EROD) activity was measured as a biomarker of contaminant exposure.

During 1995 and 1996, starlings initiated 158 nests among 5 of the 6 study areas (no nesting was initiated at the Job Corps Landfill). Ninety-five nests were completed and eggs were laid in 89 nests. Chicks hatched in 83 nests and survived to 15 days post-hatch in 54 nests. Only 1 of 11 (9%) nests initiated at the Area 9 Landfill was completed compared to 57-69% among the other sites studied. Seventy-four eggs, 82 pre-15-day-old chicks, 198 15-day-old chicks, and 20 adults were collected for contaminant and biomarker analyses.

Mean clutch size (5.0 ± 0.1) and hatch rates (4.2 ± 0.1) did not differ among sites. Mean fledging success was not different between reference (76%) and metals (65%) sites but was significantly lower at the PCB (42%) sites. One hundred percent chick mortality occurred in second nesters at PCB sites (N = 44 chicks) compared to 67% mortality at the metals site (N = 18 chicks), and 60 % at reference sites (N = 25 chicks). Chicks from PCB sites appeared anemic and had lower mean hematocrit values (7%) compared to reference chicks (15%).

Adult incubation behavior did not differ among sites but adult attentiveness behavior did. The number of times adults came to the nest when chicks were present was lower ($P < 0.001$) at PCB sites (11 times/h) compared to reference sites (15 times/h).

Mean Aroclor 1254 and quantified PCB congener concentrations were greater in eggs, pre-15-day- and 15-day-old chicks, and adults collected from the PCB sites (Area 9 Landfill and/or Area 9 Building Complex) compared to those collected from other sites studied. PCB concentrations measured in samples collected from the PCB sites are similar to concentrations previously associated with adverse effects in avian species.

Except for cadmium in kidney and lead in feathers, metal concentrations were low and quantified in few of the egg and chick kidney samples analyzed. Low concentrations of cadmium were quantified in > 69% of the chick kidney samples at all sites and low lead concentrations were quantified in > 60% of chick feather samples from all sites. A greater percentage of adult kidney and feather samples had quantified concentrations of metals but concentrations were also low in adult tissues. Metal concentrations measured in starling eggs and tissues collected from the sites studied were below those associated with adverse effects in other avian species.

Following the first year of this study, the metals site (Old Refuge Shop) was remediated. Pre-remediation kidney cadmium concentrations in 15-day-old chicks collected from the metals site were significantly greater than concentrations in 15-day-old chicks collected from this site following remediation. Furthermore, kidney cadmium concentrations in post-remediation 15-day-old chicks were similar to concentrations measured in chicks collected from reference sites.

Chick and adult morphometric measurements and EROD activity were similar among sites except that 15-day-old chick spleen, liver, and kidney weights differed among sites and liver EROD activity was greater in 15-day-old chicks collected from the Area 9 Landfill compared to those collected from other sites. Egg measurements were similar among sites except that shell thickness was greater in eggs collected from the Area 9 Building Complex compared to eggs collected from reference sites.

Results of this study demonstrate that starlings are useful avian models for measuring exposure to environmental contaminants and may serve as a biological indicator of contaminant effects. Although it is doubtful that starlings nesting at the contaminated sites studied will experience any population level effects, the relationship between observed effects and increased PCB concentrations measured in eggs and tissues of starlings collected from PCB sites suggests sufficient reason for concern that less prolific avian species feeding at contaminated sites on CONWR may experience adverse effects that influence population level productivity. This study also demonstrates that starlings are useful as biological monitors of remediation and provides contaminant concentration data that resource managers may use as a biological benchmark for evaluating the effectiveness of remedial decisions.

INTRODUCTION

In the early 1800's, settlers moved into the southern Illinois region, established farms, and utilized the area extensively for agriculture. However, the soil was infertile and badly eroded causing the farming industry to fail. In 1936, the Resettlement Administration acquired 17,250 hectares of these depleted lands with the intent to construct 3 lakes for use as an industrial water supply and to provide recreation. However, the onset of World War II shifted ownership of the project to the War Department who established the Illinois Ordnance Plant, one of the largest producers of ammunition in the nation at that time (O'Brien and Gere 1988). In addition, several other industries were established near the Plant at the eastern end of Crab Orchard Lake; they manufactured printing inks, electrical components, metal fabrication, and plating. After the war, Public Law 61, Statute 770, which was enacted on August 5, 1947, transferred management of this area from the U.S. Army to the Department of the Interior for wildlife, recreational, industrial, and related purposes (USDI 1992). Thus, Crab Orchard National Wildlife Refuge (CONWR) was established, and is unique in that it is the only refuge in the U. S. National Wildlife Refuge System managed for industry as well as wildlife. As wartime industries left, new companies moved onto CONWR and produced automobile parts, fiberglass boats, corrugated boxes, plated metal parts, tape, flares, and jet engine starters; however, munitions continued to be the primary industry. Electrical capacitors and transformers containing polychlorinated biphenyls (PCBs) also were produced until the early 1960's. Many of these companies used landfills and dumps for disposal of industrial waste (O'Brien and Gere 1988).

During routine monitoring, Hite and King (1977) found elevated concentrations of mercury in fish from Crab Orchard Lake. Livers from hunter harvested white-tailed deer (*Odocoileus virginianus*) killed on CONWR contained significantly greater concentrations of nickel and lead compared to deer collected off CONWR (Woolf et al. 1983). Additionally, Kohler et al. (1990) reported PCB concentrations greater than FDA safety limits (2 ppm) in 38% of fish collected from the eastern portion of Crab Orchard Lake. These findings prompted investigations into the source of these contaminants.

In their 1988 Remedial Investigation, O'Brien and Gere (1988) recommended 7 sites within CONWR for remediation due to potential risk to wildlife. These sites were divided into either PCB or Metals Operable Units based on major contaminants of concern. The PCB Operable Units included the Job Corps Landfill, Water Tower Landfill, Area 9 Landfill, and Area 9 Building Complex; the Metals Operable Units included the Area 7 Plating Pond, Fire Station Landfill, and Old Refuge Shop. Risk to wildlife from exposure to cadmium and cyanide at the Old Refuge Shop and PCBs, cadmium, and lead at the Job Corps Landfill was considered high. Risk to wildlife from exposure to PCBs and lead at the Area 9 Landfill, and PCBs and trichloroethene at the Area 9 Building Complex was considered moderate (O'Brien and Gere 1988).

Proposed remedial action at the PCB Operable Units included excavation and incineration of PCB contaminated soil, stabilization/fixation of the resulting ash if significant

metal concentrations were detected, and on-site landfill disposal. Proposed remediation of the Metals Operable Units included excavation of contaminated soil, stabilization/fixation, and on-site landfill disposal (USEPA 1989). Despite various public meetings addressing these procedures, members of surrounding communities remained concerned about the PCB incineration process and its potential to further contaminate the area (Rezanka 1993, Rowell 1994). Movement of contaminants from polluted sites via wild species has been documented (McKee 1995), although not all sites have been thoroughly studied. McKee (1995) found significantly greater concentrations of PCBs (Aroclor 1254) in European starlings (*Sturnus vulgaris*) collected at the Area 9 Landfill compared to those collected from a reference area. He also noted behavioral effects such as nest abandonment, but definitive conclusions could not be drawn due to small sample size.

Because CONWR is home to a variety of avian species, it is important that contaminant effects among these populations are studied. This project will provide biological and contaminant baseline data that can be used to evaluate avian exposure and effects. It also will provide a benchmark for evaluating the effectiveness of remedial actions at CONWR. Biological and analytical data will demonstrate whether or not avian species at CONWR have been exposed to hazardous concentrations of contaminants and may help reduce public anxiety by measuring the success of remediation. These issues will be addressed through the following objectives:

1) Monitor adult starlings for evidence of contaminant effects in the form of nest attentiveness and abandonment behavior at 12 nest boxes constructed at each of 4 Crab Orchard National Wildlife Refuge contaminated sites and 2 reference sites.

2) Measure physiological and biomarker responses in starling chicks and adults collected from nests at study and reference sites as indicators of effects and exposure to environmental contaminants.

3) Measure environmental contaminant exposure (PCBs and cadmium, chromium, lead, and mercury) in starling chicks and adults collected from nests at study and reference sites.

STUDY AREA

In 1984, several landfill sites at CONWR were added to the National Priorities List under the Comprehensive Environmental Recovery, Compensation, and Liabilities Act (CERCLA) (USEPA 1989). Four of these sites (Job Corps Landfill, Old Refuge Shop, Area 9 Landfill, and Area 9 Building Complex) were chosen for investigation and 2 reference sites were chosen for comparison to contaminated sites: one located on-CONWR (Area P) and one located off-CONWR (Annex) (Figure 1).

JOB CORPS LANDFILL (PCB3)

The Job Corps Landfill consists of a 4-ha pond created in the 1960's and the adjacent 0.4-ha landfill north of the pond. Widespread debris including bottles, cans, mica flakes, small electrical contacts, and a few small capacitors have surfaced from the landfill. The site was first discovered in 1985 when 30 or more goose carcasses in varying degrees of decay were found floating on the water and littering the shore of the pond. Although chemical analyses were run on the carcasses, no causative agent was identified at that time (O'Brien and Gere 1988).

O'Brien and Gere (1988) found PCB (Aroclor 1254) concentrations ranging from 0.077 to 69,042 mg/kg (dry wt) in surface soil (0-0.3 m), but concentrations diminished to <1 mg/kg (dry wt) at 0.8 m in depth. Lead, cadmium, and mercury also were found at the site with surface concentrations of 6-17,410, 1-57, and 0.19 mg/kg (dry wt), respectively. Below 0.8 m all metal concentrations were substantially decreased, with lead ranging from 6 to 219 mg/kg (dry wt) and cadmium and mercury below detection levels (O'Brien and Gere 1988).

Risk to humans was considered low because the site is closed to the public; however, even a single exposure could be hazardous. O'Brien and Gere (1988) estimated a PCB cancer risk factor of 1.1×10^{-3}, which is higher than the 10^{-7}-10^{-4} range accepted by public health agencies. Possible cadmium inhalation also was an acute hazard since cadmium is a carcinogen via this route of exposure. Possible exposure to lead was determined to be 0.088 µg/kg/day which is well below the accepted exposure limit of 1.43 µg/kg/day (O'Brien and Gere 1988).

Risk to wildlife was assessed as being high. Direct contact, inhalation, and ingestion were all determined to be viable exposure routes with possible chronic and acute effects. Both migratory and resident wildlife use this area and are subject to exposure. Resident populations of small mammals, as well as pond life including turtles, panfish, frogs, and aquatic insects may suffer lethal or sub-lethal effects (O'Brien and Gere 1988).

Chronic PCB exposure concentrations were determined to exceed 300 µg/kg/day, a level that can cause reproductive failure in mink. Lead exposure concentrations of 47.5 mg/kg/day were thought to have potential to cause adverse effects and cadmium posed substantial risk to wildlife due to its tendency for biomagnification in soil-ingesting animals such as earthworms (O'Brien and Gere 1988).

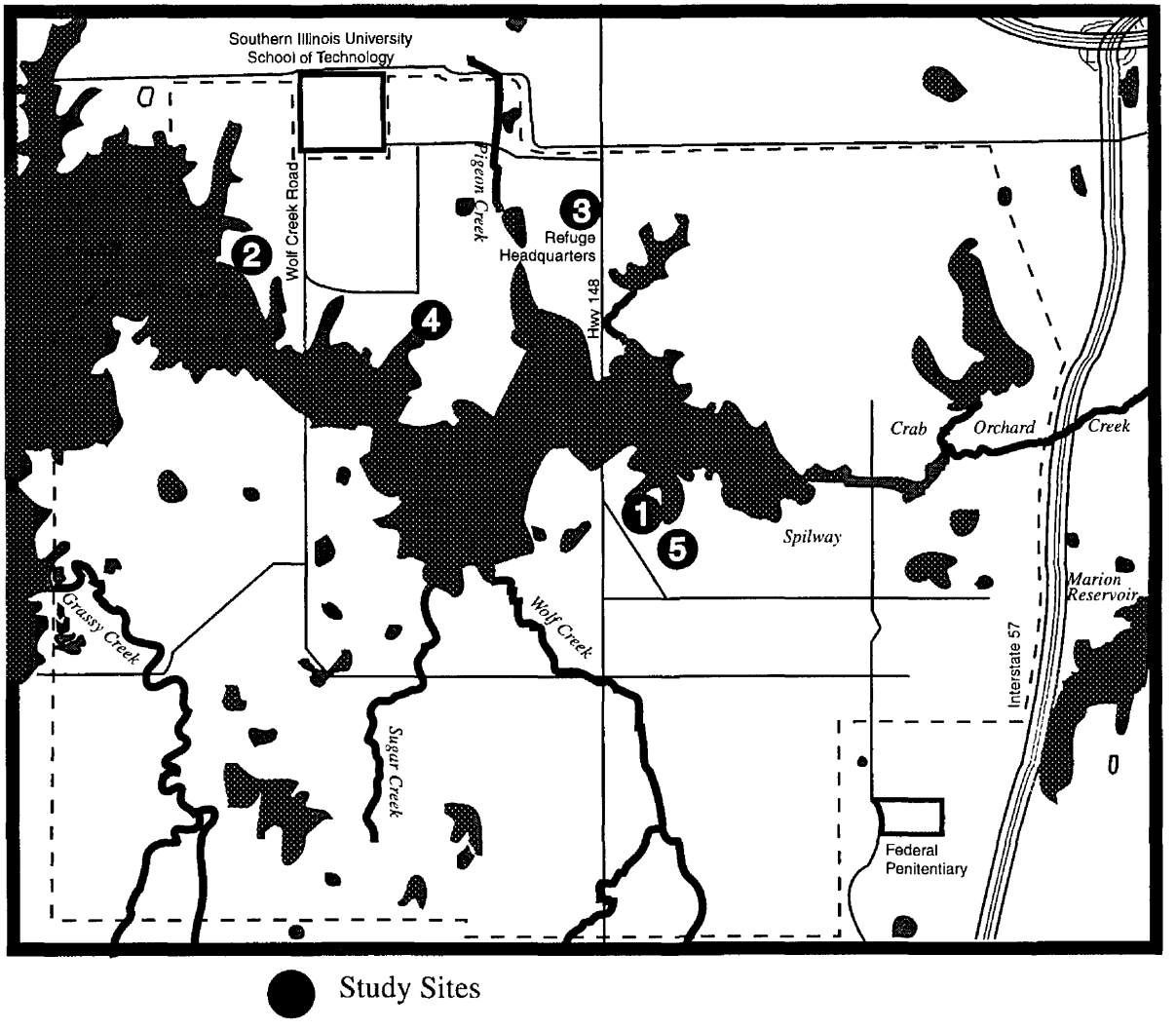


Figure 1. Location of study sites at Crab Orchard National Wildlife Refuge (CONWR). 1) Area 9 Landfill (PCB1); 2) Job Corp Landfill; 3) Old Refuge Shop (MET); 4)Area P Reference site (REF1); and 5) Area 9 Building Complex (PCB2). Annex reference site (REF2) located 32 km west of CONWR not shown.

OLD REFUGE SHOP (MET)

The Old Refuge Shop is located at the north end of CONWR along Wolf Creek Road. Behind the old refuge headquarters building, pine wood poles were treated with the wood preservative, pentachlorophenol, resulting in contaminated soil. A small drainage pool north of the building drains northwest through a wooded area to Crab Orchard Lake (O'Brien and Gere 1988).

Surface soils contained elevated dry weight concentrations of cadmium (0.68-780 ppm), chromium (10-889 ppm), and lead (93-166 ppm). The Resource Conservation and Recovery Act (RCRA) lists cadmium as a hazardous waste at 1.0 ppm. Three samples contained 9.1, 7.1, and 3.8 ppm cadmium, which are well above the RCRA classification (O'Brien and Gere 1988). Because cadmium soil concentrations at this site reach up to 780 mg/kg, it is of primary concern (O'Brien and Gere 1988).

The most common route of cadmium exposure is through ingestion; however, inhalation of wind-borne dust can occur. Risk to humans was determined to be low because the site is in a closed area. Wildlife, particularly small burrowing mammals, are at high risk for developing reproductive effects and other systemic toxicity (O'Brien and Gere 1988).

AREA 9 LANDFILL (PCB1)

Area 9 was a manufacturing site leased to Sangamo Electric Company, Capacitor Division from 1946 to 1962. Currently it is leased by Olin Corporation, a munitions manufacturer. Sangamo Electric Co. manufactured capacitors utilizing aluminum, electrolytes, mica, silver, lead foil, and PCBs. The landfill was used from the 1950's to 1964 for the disposal of wastes from capacitor manufacturing. These wastes were burned, compacted in a swale, and covered. The landfill currently covers 1 ha and is 2-3 m deep in the middle and 2-4 m deep at the edges and contains approximately 9,300-20,400 m^3 of waste. The area is covered with thick herbaceous vegetation except for bare patches where waste materials are exposed. Runoff through intermittent ditches flows into the eastern region of Crab Orchard Lake, approximately 80 m from the site (O'Brien and Gere 1988).

PCB concentrations in surface soil samples were greatest in the north and west sectors of the landfill (2,100-13,000 mg/kg [wet wt]). Soil lead concentrations ranged from 205 to 8,270 mg/kg (dry wt), and groundwater chromium concentrations were above the Illinois Public Water Safety standard of 50 mg/L. The primary concern at this site was PCB ($\bar{x}$ = 3,200 mg/kg [wet wt]) and lead contamination of soil (O'Brien and Gere 1988).

Potential human exposure to PCBs via inhalation, ingestion, and dermal routes was determined to be below the level of concern (7 µg/kg/visit), but potential exposure to lead (8.7 µg/kg/visit) was above the acceptable chronic exposure rate of 1.4 µg/kg/day. Wildlife had the greatest risk from chronic exposure to PCBs. This exposure could result in behavioral, immunological, or other subtle effects that may decrease competitiveness and survival.

Additionally, wildlife exposure to lead (up to 38 mg/kg/day) could result in behavioral and/or reproductive effects (O'Brien and Gere 1988).

AREA 9 BUILDING COMPLEX (PCB2)

The Area 9 Building Complex lies approximately 80 m west of the Area 9 Landfill, and consists of several buildings and adjacent land. It was used by Sangamo Weston, Inc. from 1946 to 1962 for the manufacturing of capacitors and is currently leased to Olin Corporation for munitions production. The area is fenced and has limited access. The total area consists of 5.3 ha, but contamination is greatest next to 2 buildings at the north end of the complex (O'Brien and Gere 1988).

PCB concentrations were >50 mg/kg (wet wt) in most surface soil samples and ranged up to 120,000 mg/kg. Primary risk was attributed to high concentrations of PCBs and trichloroethene. Short-term and long-term risks to wildlife and short-term risks to humans were assessed to be similar to that found at the Area 9 Landfill; however, long-term exposure risks for humans via inhalation, ingestion, and dermal routes at the building complex was considered to be below effects levels (O'Brien and Gere 1988).

AREA P REFERENCE SITE (REF1)

The Area P reference site is located on CONWR across from Olin Corporation building complex Area P. It is approximately 1.2 ha, consists of a large field bordered on 3 sides by woods and a road on the fourth side. Topography at this site is similar to that at contaminated sites and no known contaminants have been reported.

ANNEX REFERENCE SITE (REF2)

The Annex reference site is adjacent to the Cooperative Wildlife Research Laboratory Annex building located on the campus of Southern Illinois University at Carbondale, Illinois. This site is approximately 32 km west of CONWR, is approximately 1.2 ha in size, and consists of an open field bordered by woods to the south and the annex building to the north. No known contaminants have been reported at this site.

METHODOLOGY

NEST PRODUCTIVITY

During 1995, twelve nest boxes were constructed at each of 4 study sites located on CONWR that were identified as posing moderate to high risk to wildlife. These included the Job Corps Landfill, the Old Refuge Shop, the Area 9 Landfill, and the Area 9 Building Complex. Twelve nest boxes also were constructed at each of 2 reference sites, one located on CONWR and the other located adjacent to the Cooperative Wildlife Research Laboratory Annex building. During the 1996 field season, 12 additional boxes were added to the Annex reference site, 10 additional boxes were added to the Area 9 Building Complex, and all boxes were removed from the Area 9 Landfill due to remediation activities. Additionally, all boxes were removed from the Job Corps Landfill because they were not utilized by starlings in 1995, and because of remediation activities.

Reproductive parameters assessed included number of nests constructed, number of eggs laid per nest, number of eggs that hatch per nest, and number of chicks that survive 15 days post-hatch per nest. Chick weights also were measured in the field with an analytical scale at days 3, 9, and 15 post-hatch. These data were collected between 1030-1530 hours, because disturbances outside this time block could increase the probability of nest abandonment (Kessel 1957, Bogucki 1972, Feare 1984, Kendall et al. 1989). Nest boxes were checked 3 times per week until egg-laying began; every other day until egg-hatching began; daily until hatching was completed; then 3 times per week until chicks and adults were collected.

MORPHOMETRIC MEASUREMENTS

Chicks were collected from nests when the average nestling age for the clutch was 15 days post-hatch. Attempts to capture one or both of the adults were made prior to collection of chicks. Adults were captured using a pull down flap to cover the nest hole (Figure 2) and chicks were collected by hand. Live chicks and adults were transported to the Wildlife Annex where they were euthanized by asphyxiation with CO_2 and necropsied. During the 1996 season 3 hematocrit samples were collected from the brachial vein of chicks and adults prior to euthanization.

Eggs that failed to hatch were collected 5 days after the last hatching occurred and were taken to the Wildlife Annex for examination. Eggs were weighed and measured, (length, width, and greatest circumference), and contents were emptied into a large petri dish. Contents were weighed and shell thickness and weight were measured. Evidence of development was recorded and embryos were examined for abnormalities (deformed limbs or crossed bill). The embryo and other egg contents were then homogenized and separated into 2 aliquots of approximately equal size, and these stored at -80°C prior to PCB and metal analyses.

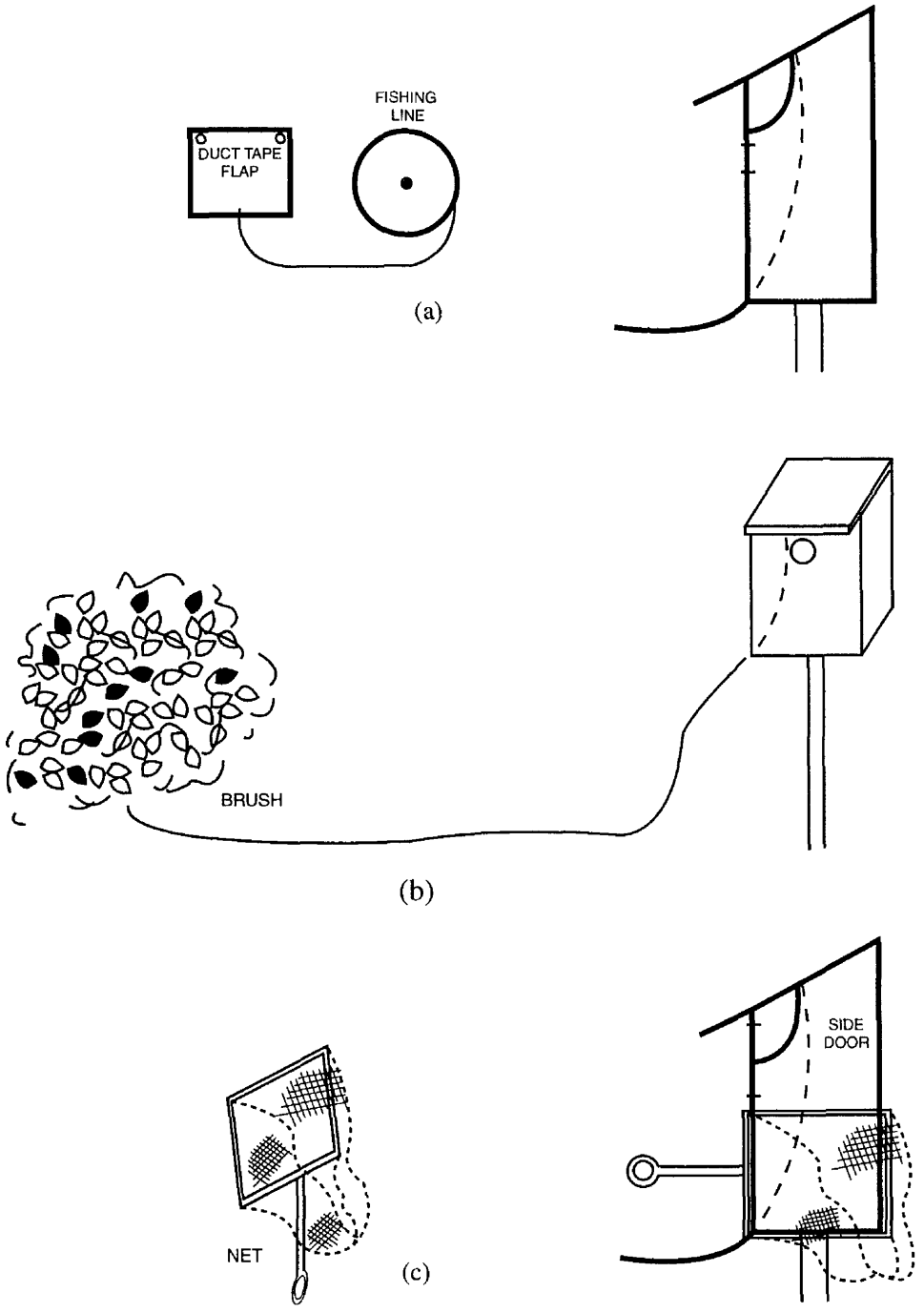


Figure 2. Method used to trap adult starlings in the nest box for collection: a) a duct tape flap was tacked inside the nest box so that it was not visible from the outside; b) fishing line was used to pull the flap over the hole from a remote location when the adult entered the box; c) the adult was then captured with a small net from the side opening of the box.

Following euthanasia, chick and adult measurements (total weight, total length, wing length, and tarsus length) were recorded. Feathers from the entire carcass were collected and, if wet, dried for 24 hours at 37°C. The carcass was opened and examined for gender determination. Kidneys, liver, and spleen were collected and weighed and the heart was weighed and returned to the carcass. A small piece of liver tissue was collected for histological analysis, and the rest of the liver was archived prior to PCB and EROD analyses. The entire digestive system was removed and archived.

All tissues and organs were wrapped in aluminum foil, labeled, and archived at -80°C prior to analysis. Samples collected for histological analysis were placed in a 10% neutral buffered formalin solution, labeled, and stored at room temperature. Feathers were stored in a paper envelope at room temperature. Kidney tissue and feathers were analyzed for Cd, Cr, Pb, and Hg, and the remainder of the carcass was homogenized and lipid content and PCB and 34 PCB congener (CB) concentrations were determined.

BEHAVIOR

Nest attentiveness was determined through observations of adult starling behavior during nesting. Sampling methods included a combination of frequency and scan sampling as described by Lehner (1979). Adult behavior, described in Appendix A, was observed for 30 minutes and recorded at 1 minute intervals. The total number of times the adult(s) came to the nest also was recorded during this 30 minute period.

Three sites were randomly selected for the first day of observations and were alternated daily with the remaining 3 sites. Weather conditions were noted and schedules altered to prevent weather induced biases. Nest boxes were divided into 2 groups of 6 each and one observer monitored 3 boxes in 1 group for 30 minutes and the other 3 boxes in that group for the remaining 30 minutes of the hour. A second observer monitored the second group of 6 boxes in a similar manner. Observations were repeated 3 times per week at each site.

PCB AND PCB CONGENERS

PCB concentrations were determined in whole carcass homogenates following EPA methods 3541, 3640A, and 8081 for extraction, clean-up, and quantification, respectively (USEPA 1992). Samples were homogenized with anhydrous sodium sulfate at a ratio of 5:1, Na_2SO_4 to tissue, and extracted with hexane using a Soxtec continuous extractor. Clean-up was accomplished by solvent partitioning with Gel Permeation Chromatography (GPC) and the hexane fraction reduced using rotary evaporation. A Hewlett-Packard 5890 II Gas Chromatograph (Hewlett Packard Co., Wilmington, Del.) equipped with a Ni-63 electron capture detector and DB5 fused silica capillary column was used for extract analysis. A thermal gradient from 60°C to 250°C was used for elution of congeners, and helium and nitrogen were used as the carrier and make-up gas, respectively. Duplicate samples were run through the entire analytical procedure and spike recoveries in uncontaminated samples were used to assess analytical

efficiency. Quantification was accomplished by summing the values of all peaks in the retention time range of 29 to 60 minutes, which was determined using a standard Aroclor 1254 preparation (Aroclor 1254 is the dominant PCB in soil at CONWR). In addition to total Aroclor 1254, 34 individual CB congeners were quantified (Table 1). Final concentrations are expressed as μg 1254 (or CB)/g tissue (wet wt).

Biomarker responses in chick and adult starlings were quantified by measuring EROD activity in liver tissue following methods of Mazel (1971) and Lubet et al. (1985). Microsomal fractions were obtained by centrifugation followed by O-dealkylation of ethoxyresorufin and measurement of resorufin formation using an Hitachi F-2000 Fluorescence spectrophotometer (Hitachi, Ltd., Tokyo, Japan).

METALS

Cadmium, chromium, and lead concentrations in kidney, feather, and egg tissue were quantified following EPA Methods 213.2, 218.2, and 239.2 (USEPA 1987), respectively, using atomic absorption spectrophotometry. Samples were prepared according to EPA Method 200.3 (McDaniel 1991). Duplicate aliquots were dried and digested with nitric acid and metal concentration determined using a Perkin-Elmer Model 4100ZL Graphite Furnace Atomic Absorption Spectrometer (Perkin-Elmer Corp., Cupertino, Calif.). Mercury concentrations were determined by cold vapor analysis (EPA Method 245.6, Lobring and Potter 1991) using a Bacharach Coleman Model 50B Mercury Analyzer System (Bacharach, Inc., Pittsburgh, Pa.).

QUALITY CONTROL/QUALITY ASSURANCE

Field Sampling

Nest boxes were uniquely numbered. A registered notebook and sequentially numbered forms were used for recording field observations, reproductive data, and necropsy data. Duplicate copies of recorded data were made on the day information was entered and stored at separate locations.

Eggs were uniquely marked in permanent ink, and chicks and adults had a uniquely numbered leg band attached to 1 leg. Eggs were labeled with a unique 6-character alpha-numeric identification. On the day of collection, chicks from each nest were kept separate from chicks from other nests.

Laboratory Samples

One duplicate tissue sample was analyzed per sample batch. Additionally, standards (sample with a known concentration of the contaminant), spiked samples (tissue of unknown concentration to which a known concentration of the contaminant is added) and blanks (matrix only) were analyzed prior to beginning sample analysis and along with every sample batch.

Table 1. Chlorine substitution patterns of PCB congeners[1] measured in the current study.

CB	Chlorine Substitution	CB	Chlorine Substitution
18	2,2',5-trichlorobiphenyl	153	2,2',4,4',5,5'-hexachlorobiphenyl
31	2,4',5-trichlorobiphenyl	[b]156	2,3,3',4,4',5'-hexachlorobiphenyl
40	2,2'3,3'-tetrachlorobiphenyl	[c]167	2,3,4,4',5,5'-hexachlorobiphenyl
44	2,2'3,5'-tetrachlorobiphenyl	169	3,3',4,4',5,5'-hexachlorobiphenyl
49	2,2'4,5'-tetrachlorobiphenyl	170	2,2',3,3',4,4',5-heptachlorobiphenyl
52	2,2'5,5'-tetrachlorobiphenyl	180	2,2',3,4,4',5,5'-heptachlorobiphenyl
77	3,3'4,4'-tetrachlorobiphenyl	183	2,2',3,4,4',5',6-heptachlorobiphenyl
87	2,2',3,4,5'-pentachlorobiphenyl	[c]185	2,2',3,4,5,5',6-heptachlorobiphenyl
99	2,2',4,4',5-pentachlorobiphenyl	194	2,2',3,3',4,4',5,5'-octachlorobiphenyl
101	2,2',4,5,5'-pentachlorobiphenyl	195	2,2',3,3',4,4',5,6-octachlorobiphenyl
105	2,3,3',4,4'-pentachlorobiphenyl	[b]200	2,2',3,3',4,5',6,5'-octachlorobiphenyl
110	2,3,3',4',6-pentachlorobiphenyl	201	2,2',3,3',4',5,5',6-octachlorobiphenyl
118	2,3',4,4',5-pentachlorobiphenyl	203	2,2',3,4,4',5,5',6-octachlorobiphenyl
126	3,3',4,4',5-pentachlorobiphenyl	205	2,3,3',4,4',5,5',6-octachlorobiphenyl
128	2,2',3,3',4,4'-hexachlorobiphenyl	206	2,2',3,3',4,4',5,5',6-nonachlorobiphenyl
129	2,2',3,3',4,5-hexachlorobiphenyl	[a]207	2,2',3,3',4,4',5,6,6'-nonachlorobiphenyl
138	2,2',3,4,4',5'-hexachlorobiphenyl	208	2,2',3,3',4,5,5',6,6'-nonachlorobiphenyl
[a]141	2,2',3,4,5,5'-hexachlorobiphenyl	209	2,2',3,3',4,4',5,5',6,6'-decachlorobiphenly
151	2,2',3,5,5',6-hexachlorobiphenyl		

[1]Numbers as suggested in Ballschmiter and Zell (1980)
[a]Congeners with the same letter co-elute together and one value is reported for the pair.

Aliquots of 10 randomly selected homogenized chick and kidney tissue samples each were sent to the Illinois Department of Natural Resources, Waste Management and Research Center (WMRC), One East Hazelwood Drive, Champaign, Illinois 61820, for PCB and metal analyses. Comparison of Cooperative Wildlife Research Laboratory (CWRL) results and WMRC results revealed an overall RPD of 63, 81, 63, and 114 for PCBs, lead, cadmium, and chromium, respectively. PCB, cadmium, and chromium RPDs were improved to 35, 33, and 94 when excluding samples with concentrations below 1 ppm (wet wt).

Mean recovery rates for fortified blanks were 104, 86, 100, 84, and 105% for PCBs, lead, cadmium, chromium, and mercury, respectively. Mean recovery rates for fortified matrix (accuracy) were 113, 100, 94, 87, and 99% for PCBs, lead, cadmium, chromium, and mercury, respectively. Additionally, mean relative percent differences (RPD) between duplicate samples (precision) were 19, 9, 5, 13, and 6 RPD for PCBs, lead, cadmium, chromium, and mercury, respectively. Precision was greater in samples with greater concentrations of PCBs (RPD of 5 % or less in samples with Aroclor 1254 concentrations >1 ppm [wet wt]).

DATA ANALYSIS

Reproductive success (eggs/nest, chicks/nest), physiological measurements (body and organ measurements), biomarker response (EROD activity), contaminant concentrations (Aroclor, congeners, and metals), and behavioral data (number of times adults came to nest per hr and percent time spent in the nest) were compared among sites using an ANOVA test. A Student's *t*-test was used to compare results between reference sites and, if appropriate, these data were combined prior to comparisons among sites. Normality was verified by the absence of extreme positive or negative kurtosis and skewness in the sample populations. EROD activity data also was correlated with PCB data using a Pearson's correlation test. Statistical probabilities ≤ 0.05 were considered significant. Statistical analysis of lipid adjusted PCB concentrations did not differ from non-lipid adjusted analysis; therefore, all PCB values are presented as non-lipid adjusted concentrations.

For PCB congener analysis, if $\geq$50% of samples from a site had detectable concentrations of a particular congener, then one-half the minimum detection limit was used in place of non-detects to allow for statistical comparisons. Additionally, if <50% of samples from a site had detectable concentrations of a particular congener, then that congener was excluded when determining profiles (% of total congeners) and was treated separately in the results and discussion.

RESULTS

NEST PRODUCTIVITY

Starlings initiated nest building around 1 April, and the first egg was laid at reference sites on 10 April and 17 April in 1995 and 1996, respectively. At contaminated sites, the first egg was laid on 13 April in 1995 and 20 April in 1996. During 1995 and 1996, starlings initiated 158 nests among 5 of the 6 study areas (no nesting was initiated at the Job Corps Landfill). From these, 95 nests were completed and eggs were laid in 89 nests. Chicks hatched in 83 nests and survived to 15 days post-hatch in 54 nests (Table 2). Of the number of nests initiated, the percent that went to full cup was similar among sites except for PCB1. At this site, only 1 of 11 initiated nests (9%) went to full cup compared to 57-69% among the other 4 sites (Table 2). Biological samples collected included 74 eggs, 82 pre-15-day-old chicks, 198 15-day-old chicks, and 20 adults (Table 3).

Mean clutch size and hatch rates did not differ among sites (Table 4). Mean percent fledging success at reference and MET sites did not differ ($P = 0.362$) and produced approximately 3 chicks per nest; however, percent fledging success at PCB sites was lower ($P = 0.017$) compared to reference sites and only 2 chicks per nest were produced (Table 4).

During the 1995 and 1996 breeding seasons, 44 (100%) chicks from second nesters at the PCB sites and 18 (67%) chicks at the metals site died prior to 15 days post-hatch. Evidence of anemia was observed in these chicks. During 1996, hematocrits were collected from a sample of 1-day-old chicks at PCB and reference sites. Mean hematocrit value for 1-day-old chicks from PCB2 was 7% (6-10%, n=5) compared to 15% (9-19%, n=4) in 1-day-old chicks from reference locations. A single 1-day-old chick hematocrit from MET had a value of 11.5%. Hematocrits in 1-day-old chicks were not compared statistically due to small sample size, however, there was a trend towards lower hematocrits at PCB2.

SEX RATIOS AND MORPHOMETRIC MEASUREMENTS

Although sex ratios among fledglings varied (29% females at PCB sites (PCB1 and PCB2 combined) and REF1 to 57% females at MET), these were not significant ($P = 0.201$). Three-day body weights differed ($P = 0.036$) between reference and MET sites, but not between PCB and reference or MET sites (Table 5). Nine-day body weights, 15-day body weights and lengths, relative wing and tarsus lengths (percent of body length), relative heart weight (percent of body wt), and percent lipid did not differ among sites. Relative spleen weights were larger ($P < 0.001$) at reference and MET sites compared to PCB sites (Table 5). Relative liver weights were larger ($P = 0.003$) at reference sites compared to PCB sites; however, liver weights at MET did not differ from either reference or PCB sites. Relative kidney weights were larger ($P = 0.003$) at reference sites than at MET; however, kidney weights at PCB sites did not differ from either reference or MET sites. Mean hematocrit values in 15-day-old chicks were reduced ($P =$

Table 2. Number of active starling nests recorded during different nesting stages from contaminated sites (PCB1, PCB2, and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites[2], 1995-1996.

Site	Initiated	Full Cup	Eggs	Chicks	15 Day Chicks
REF2	41	27	24	22	17
REF1	30	17	16	13	9
MET	27	16	16	16	12
PCB2	49	34	32	31	15
PCB1[3]	11	1	1	1	1

[1]PCB1 = Area 9 Landfill, PCB2 = Area 9 Building Complex, and MET = Old Refuge Shop.
[2]One reference site was located on CONWR (REF1 = Area P reference site) and 1 was located 32 km west of CONWR (REF2 = Annex reference site).
[3]Not sampled in 1996 due to remediation.

Table 3. Number of starling samples collected and number of chicks predated or missing from contaminated sites (PCB1, PCB2, and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites[2], 1995-1996.

Site	Eggs	Pre-15 Day	Chicks P/M[3]	15 Day	Adults
REF2	14	21	8/4	69	3
REF1	16	11	8/2	34	3
MET	16	15	0/6	37	6
PCB2	28	34	13/24	55	7
PCB1[4]	0	1	0/0	3	1

[1]PCB1 = Area 9 Landfill, PCB2 = Area 9 Building Complex, and MET = Old Refuge Shop.
[2]One reference site was located on CONWR (REF1 = Area P reference site) and 1 was located 32 km west of CONWR (REF2 = Annex reference site).
[3]P/M = Predated/Missing
[4]Not sampled in 1996 due to remediation.

Table 4. Starling nest productivity at contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

	REF (n = 28 nests[3])		MET (n = 14 nests)		PCB (n = 29 nests)		
PARAMETER	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	P-Value[4]
# of Eggs	4.9	0.1	5.1	0.1	5.0	0.1	0.352
# Hatched	4.4	0.2	4.1	0.3	4.1	0.3	0.627
% Hatched	90.1	3.9	79.5	6.0	80.9	4.3	0.208
# of Chicks	3.4[a]	0.4	2.6[ab]	0.5	1.9[b]	0.4	0.025
% Fledged	75.9[a]	7.3	65.2[ab]	11.5	42.4[b]	8.5	0.015

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]Nests affected by predation were excluded in statistical analyses.
[4]ANOVA
[a]Means with the same letters are not different ($P > 0.05$) as determined by Scheffe post-hoc separation.

Table 5. Morphometric measurements from 15-day-old starling chicks for starlings nesting at contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

	REF ($n = 98$)		MET ($n = 37$)		PCB ($n = 58$)		
PARAMETER	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	P-Value[3]
3 Day Body Wt (g)	22.08[a]	0.87	17.54[b]	1.63	20.18[ab]	0.88	0.036
9 Day Body Wt (g)	62.87	2.00	61.46	2.74	60.09	1.96	0.631
15 Day Body Wt (g)	65.07	0.83	64.21	1.10	62.55	0.93	0.139
% Carcass Lipid	2.6	0.2	2.5	0.3	2.5	0.2	0.882
Body Length (mm)	191	1	187	2	190	2	0.331
PCV	34[a]	1	38[b]	1	36[ab]	1	0.004
Wing (%)[4]	44.45	0.30	43.74	0.44	43.57	0.31	0.117
Tarsus (%)	24.96	0.23	24.95	0.32	24.59	0.23	0.505
Spleen (%)[5]	0.21[a]	0.01	0.19[a]	0.01	0.14[b]	0.01	<0.001
Liver (%)	5.76[a]	0.10	5.7[ab]	0.20	5.2[b]	0.12	0.003
Kidney (%)	1.53[a]	0.02	1.37[b]	0.04	1.46[ab]	0.03	0.003
Heart (%)	1.08	0.02	1.06	0.02	1.05	0.02	0.307

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]ANOVA.
[4]Wing and tarsus represented as % of body length.
[5]Spleen, liver, kidney, and heart represented as % of body wt.
[a]Means with the same letters are not different ($P > 0.05$) as determined by Scheffe post-hoc separation.

0.004) at reference sites compared to MET (38%); however, hematocrit values at the PCB sites were not different from those at reference or MET sites (Table 5).

One chick with a lethal bill deformity (Figure 3) was collected from a MET site nest with low hatching (40%) and fledging (25%) rates. Metals and PCB concentrations in this chick were below effects levels, and mass spectrometry analysis did not reveal any other contaminants in concentrations of concern.

Adult morphometric and hematocrit measurements did not differ among sites (Table 6). Overall mean body, spleen, liver, kidney, and heart weights were 70.25, 0.07, 2.84, 1.04, and 0.85 g, respectively; and overall mean body, wing, and tarsus lengths were 233, 125, and 48 mm, respectively. Mean hematocrit values for adults were 52, 50, and 46% for reference, MET, and PCB sites, respectively.

Shell thickness was the only egg measurement that differed among sites. Egg shells were thinner ($P = 0.014$) at reference sites compared to PCB sites; however, shell thickness at MET did not differ from shell thickness at either reference or PCB sites (Table 7). Overall mean length, width, and greatest circumference were 29, 21, and 71 mm, respectively; and overall mean egg content and shell weights were 5.2 and 0.4 g, respectively.

BEHAVIOR

Maintenance behavior was the only behavior that differed among sites prior to the beginning of egg laying. Adult starlings at the PCB sites spent a greater ($P = 0.018$) percent of time (0.40%) performing maintenance behaviors, such as preening, than those at the metals site (0.02%). Adult maintenance behavior at reference sites, however, did not differ from that observed at PCB or metals sites.

When eggs were present in the nest, adult incubation behavior (IN) did not differ ($P = 0.687$) among sites. Time spent perching in the doorway (XIN) was greater ($P = 0.006$) at the metals site (8.7%) compared to PCB sites (5.1%), but this behavior at reference sites (6.2%) did not differ from either metals or PCB sites. No other monitored adult behaviors differed during egg incubation.

While chicks were in the nest, adult nest attentiveness behavior (number of times per hr adults came to the nest box (TCN) to feed chicks) was reduced ($P < 0.001$) from 15 ± 1 counts per hour at reference sites to 11 ± 1 counts per hour at PCB sites, but was not different between reference and metals sites (13 ± 1 counts per hr). Foraging trips per hour made by adult starlings during days 1-5 post-hatch did not differ between PCB and reference sites; however, foraging trips per hour made during days 6-10 post-hatch were significantly greater at reference sites (18 trips/hr) compared to PCB (12 trips/hr) sites (Figure 4). Additionally, there was a 73% increase in the average number of foraging trips per hour from days 1-5 to days 6-10 post-hatch at reference sites, while only a 26% increase between these 2 periods was observed at

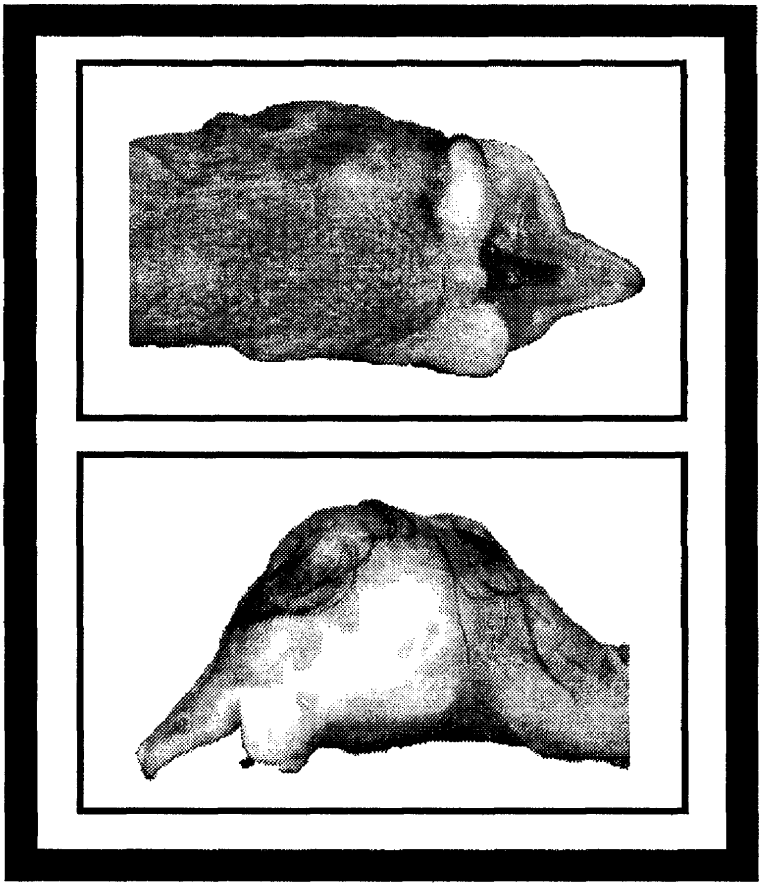


Figure 3. Shortening of the lower bill in a 1-day-old starling chick collected from a site with metals and dioxin contamination (MET = Old Refuge Shop) at Crab Orchard National Wildlife Refuge, 1996.

Table 6. Morphometric measurements for adult starlings nesting at contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

	REF ($n = 5$)		MET ($n = 7$)		PCB ($n = 8$)		
PARAMETER	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	*P*-Value[3]
Body Weight (g)	70.25	1.82	69.85	0.90	70.44	1.66	0.943
Body Length (mm)	236	2	229	3	236	3	0.166
PCV	52	3	50	3	46	1	0.194
Wing (%)[4]	52.94	0.43	54.19	0.71	53.15	0.91	0.504
Tarsus (%)	20.53	0.47	20.74	0.54	20.15	0.44	0.672
Spleen (%)[5]	0.08	0.01	0.12	0.03	0.08	0.01	0.319
Liver (%)	3.96	0.33	4.25	0.29	3.88	0.29	0.656
Kidney (%)	1.46	0.10	1.51	0.11	1.46	0.09	0.900
Heart (%)	1.19	0.04	1.22	0.04	1.21	0.04	0.942

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]ANOVA.
[4]Wing and tarsus represented as % of body length.
[5]Spleen, liver, kidney, and heart represented as % of body wt.

Table 7. Egg measurements for starlings nesting at contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

	REF ($n = 27$)		MET ($n = 11$)		PCB($n = 27$)		
PARAMETER	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	P-Value[3]
Weight (g)	5.94	0.15	5.47	0.31	5.61	0.19	0.264
Length (mm)	29.3	0.3	29.3	0.4	29.2	0.3	0.961
Width (mm)	20.8	0.2	21.9	1.0	21.0	0.2	0.141
Circumference (mm)	71	1	72	1	71	1	0.779
Content Wt (g)	4.89	0.19	4.51	0.31	4.71	0.17	0.524
Shell Thickness (mm)	0.26[a]	0.01	0.28[ab]	0.01	0.29[b]	0.01	0.014
Shell Wt (g)	0.45	0.01	0.44	0.03	0.45	0.01	0.898

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]ANOVA.
[a]Means with the same letters are not different ($P > 0.05$) as determined by Scheffe post-hoc separation.

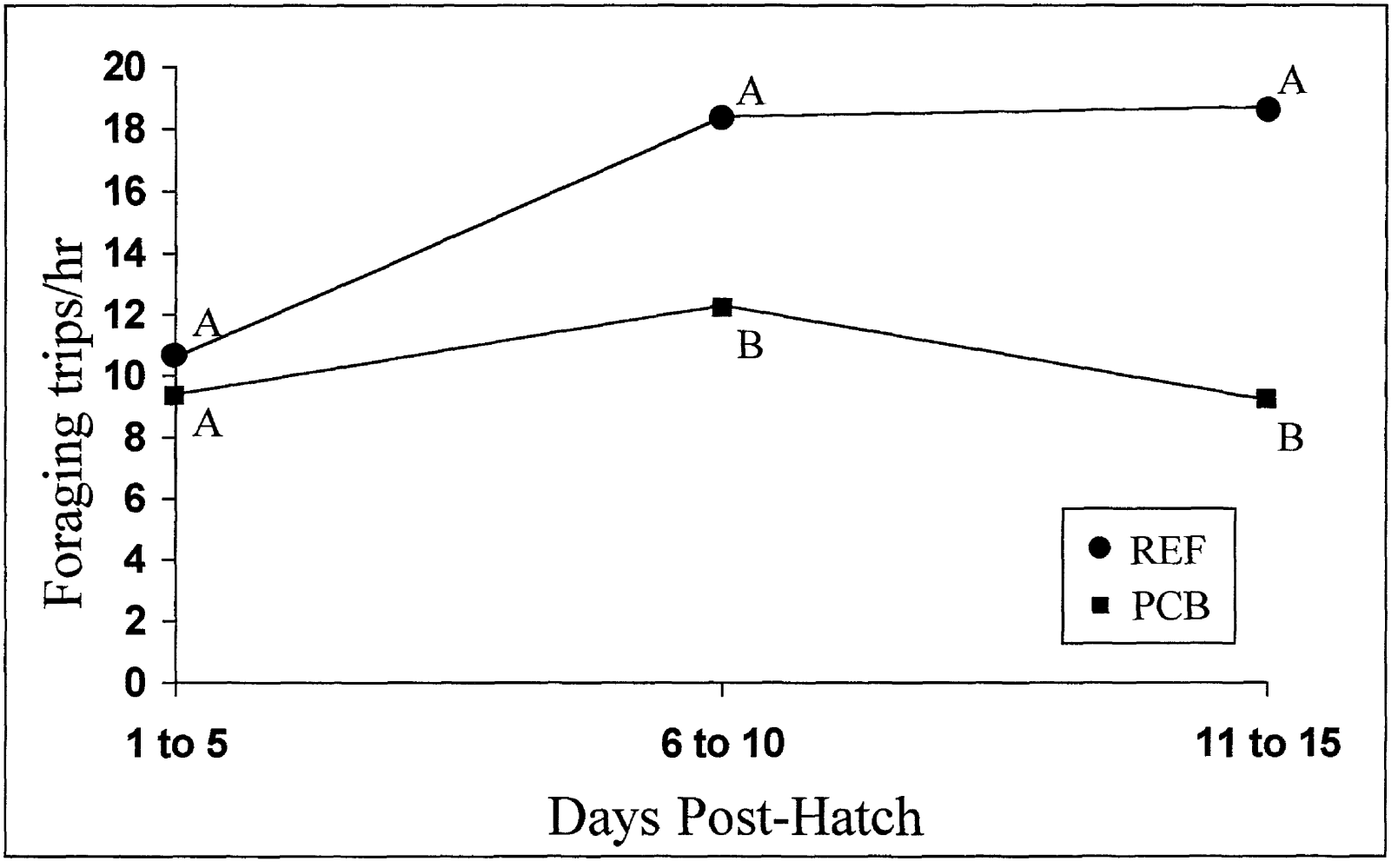


Figure 4. Foraging activity (number of foraging trips per hr) in adult starlings nesting at the Area P and Annex reference sites (REF) and the Area 9 Landfill and Area 9 Building Complex (PCB). Different letters indicate statistical differences ($P \leq 0.05$) between REF and PCB.

PCB sites (Figure 4). There also was a 25% decrease in the number of foraging trips per hour from days 6-10 to days 11-15 post-hatch at the PCB sites compared to a 2% increase in feeding activity of reference adults between these 2 periods (Figure 4). Additionally, maintenance (M) behavior while chicks were in the nest, was significantly greater at PCB sites (0.34%) compared to reference sites (<0.01%). No other adult behaviors that were monitored when chicks were present in the nest differed among sites.

PCB AND PCB CONGENERS

15-Day-Old Chicks

In 15-day-old chicks, 47, 38, and 18% of the 34 CBs analyzed were detected in ≥50% of the samples collected from PCB1, PCB2, and MET, respectively. There were no CBs detected in ≥50% of 15-day-old chicks collected from the reference sites. CBs 138, 153 and 180 predominated (CBs that accounted for ≥10% of total CBs) in 15-day-old chicks collected at MET and CBs 99, 101, 118, 138 and 153 were predominant at PCB1 and PCB2 (Table 8). Two coplanar CBs (110 and 118) were quantified in ≥50% of 15-day-old chicks from the PCB sites and 1 coplanar CB (118) was quantified in ≥50% of 15-day-old chicks collected from MET.

Mean concentrations of CBs, Aroclor 1254, and EROD activity were approximately 10 times greater in chicks collected from PCB1 compared to all other sites; however, small sample size (n = 3) prevented statistical comparisons that included this site (Table 8). Mean Aroclor 1254 concentrations were greater ($P < 0.001$) at PCB2 (6.0 ppm) compared to references sites (0.5 ppm) and EROD activity was positively correlated ($r = 0.383$, $P < 0.001$) with Aroclor 1254 concentrations. EROD activity differed ($P = 0.002$) among sites and was greater at MET compared to reference sites; however, EROD activity at PCB2 did not differ from reference sites.

Mean Aroclor 1254 concentrations for 15-day-old chicks from individual nests at PCB1 and PCB2 were positively correlated ($r = 0.667$, $P < 0.001$) with the contaminant concentration in the soil where the nest box was located. Chicks from nest boxes located on category 2 soil (Soil with Aroclor 1254 ≥ 25, Pb < 450, and Cd < 25 ppm [dry wt]) had greater Aroclor 1254 concentrations compared to chicks from nest boxes located in category 1 soil (Soil with Aroclor 1254 < 25, Pb < 450, and Cd < 25 ppm [dry wt]) (Figures 5 and 6). EROD activity was greater in 75% of chicks from nests located in category 2 soil compared to chicks from nests located in category 1 soil (Figures 5 and 6). Mean chick Aroclor 1254 concentrations in chicks from nests at MET and reference sites did not differ.

Eggs

In eggs, 44, 18, 6, and 9% of the 34 CBs analyzed were detected in ≥50% of the samples collected from PCB2, MET, REF1, and REF2, respectively. No eggs were collected from PCB1. As in 15-day-old chicks, CBs 99, 101, 118, 138, and 153 were the predominant CBs at PCB2 and CBs 138, 153, and 180 were dominant at MET (Table 9). Two coplanar CBs (110 and 118) were

Table 8. Mean (± SE) Aroclor 1254 and PCB congener (CB) concentrations (ppm [wet wt]) in whole body homogenates of 15-day-old starling chicks collected from contaminated sites (PCB1, PCB2, and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

CB	PCB1 ($n = 3$) x̄±SE (% of Sum of CBs)		PCB2 ($n = 55$) x̄±SE (% of Sum of CBs)		MET ($n = 36$) x̄±SE (% of Sum of CBs)		REF ($n = 99$)	P-Value[3]
52	1 49 ± 0.13	(5)	0.16 ± 0 02	(4)	N Q [4]		N.Q.	N.A.[5]
49	0.53 ± 0 06	(2)	0 07 ± 0.01	(2)	N Q		N.Q.	N A
44	0.20 ± 0.04	(1)	N.Q		N Q		N Q	N.A
101	3.58 ± 0 27	(13)	0.54 ± 0 06	(13)	0 03 ± 0.00	(7)	N Q	N.A.
99	3.21 ± 0.23	(12)	0 42 ± 0.05	(10)	0.02 ± 0 00	(7)	N Q.	N.A.
87	1 05 ± 0 12	(4)	0.09 ± 0.01	(2)	N Q		N.Q	N A.
110	1.57 ± 0.16	(6)	0.16 ± 0 02	(4)	N Q		N Q.	N A
118	3 80 ± 0 20	(14)	0.56 ± 0 06	(14)	0.03 ± 0 00	(7)	N.Q.	N.A.
153	3.86 ± 0.20	(14)	0.67 ± 0.08	(16)	0 06 ± 0 01	(15)	N.Q.	N A
105	1.14 ± 0 03	(4)	0 18 ± 0 02	(4)	N Q		N.Q.	N A
138	3 95 ± 0 22	(14)	0.75 ± 0 09	(18)	0 04 ± 0 00	(11)	N.Q.	N.A
128	1 05 ± 0.08	(4)	0 15 ± 0.02	(4)	N Q		N Q	N.A.
185/167	0 22 ± 0 01	(1)	N.Q		N Q		N Q	N.A
156/200	0.58 ± 0 03	(2)	N.Q.		N Q		N Q.	N A
180	0.96 ± 0.05	(4)	0 14 ± 0.02	(4)	0.04 ± 0.00	(10)	N.Q.	N A
170	0.50 ± 0 04	(2)	0.06 ± 0.01	(2)	N Q		N.Q.	N A.
AR1254	52.5 ± 3.5		5.9 ± 0 8[a]		0.6 ± 0 1[b]		0 5 ± 0.1[b]	<0.001
EROD	399 7 ± 11 1		155 8 ± 10 7[ab]		179 4 ± 3.6[a]		123 8 ± 6 1[b]	0.002

[1]PCB1 = Area 9 Landfill, PCB2 = Area 9 Building Complex, and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference site).
[3]ANOVA (PCB1 not included due to small sample size).
[4]N.Q. = <50% of samples had quantifiable concentrations of the CB.
[5]N.A. = low percentage of samples with detectable concentrations precluded meaningful statistical comparisons.
[a]Means with the same letters are not different ($P > 0.05$).

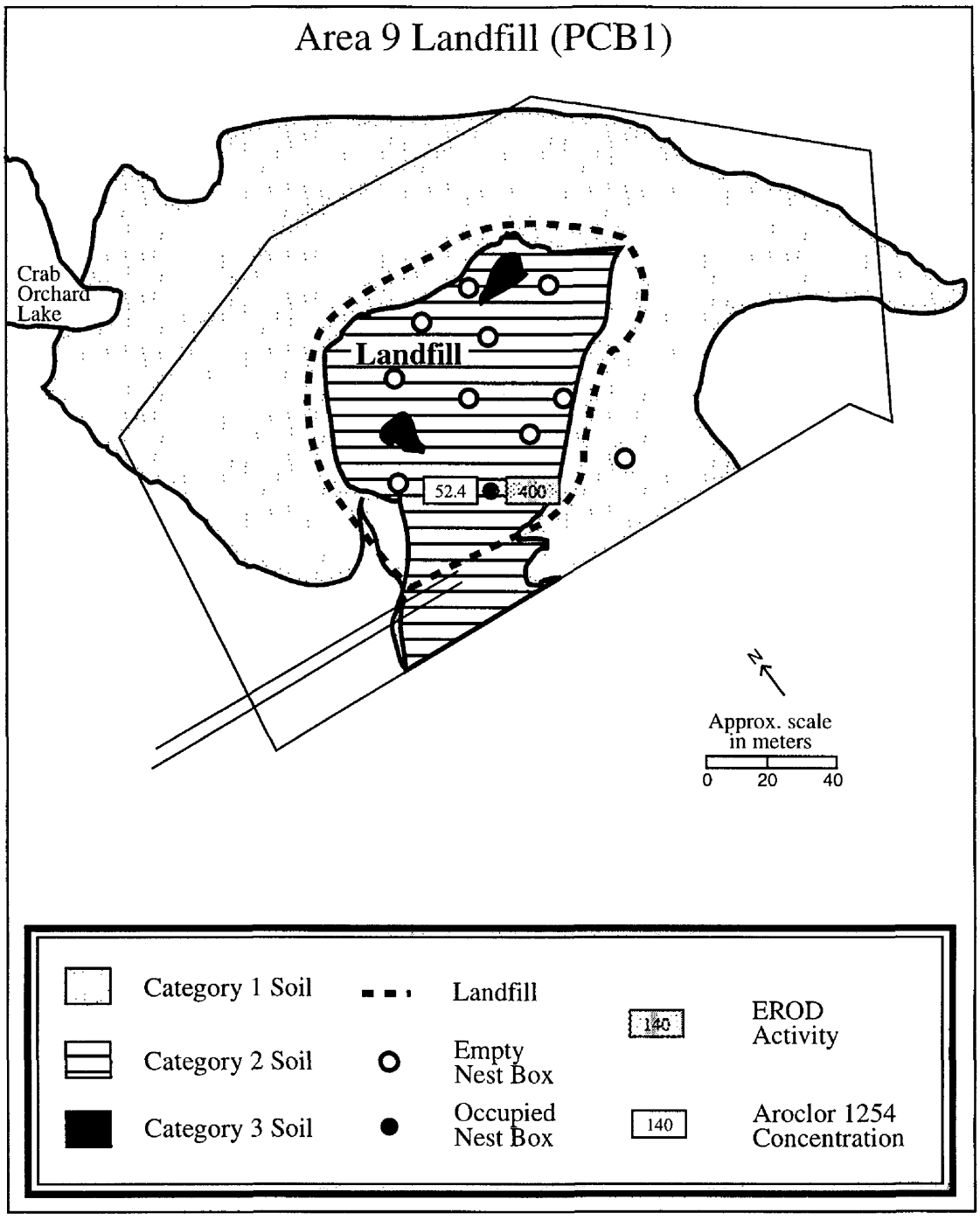


Figure 5. Aroclor 1254 concentrations (ppm [wet wt]) and EROD activity (pmol/mg protein/min) in 15-day-old starling chicks collected at the Area 9 Landfill (PCB1) located at Crab Orchard National Wildlife Refuge, 1995-1996. Category 1 soil = Aroclor 1254 < 25, Pb < 450, and Cd < 25 ppm (dry wt), Category 2 soil = Aroclor 1254 ≥ 25, Pb < 450, and Cd < 25 ppm (dry wt), and Category 3 soil = Aroclor 1254 ≥ 25, Pb ≥ 450, and Cd ≥ 25 ppm (dry wt).

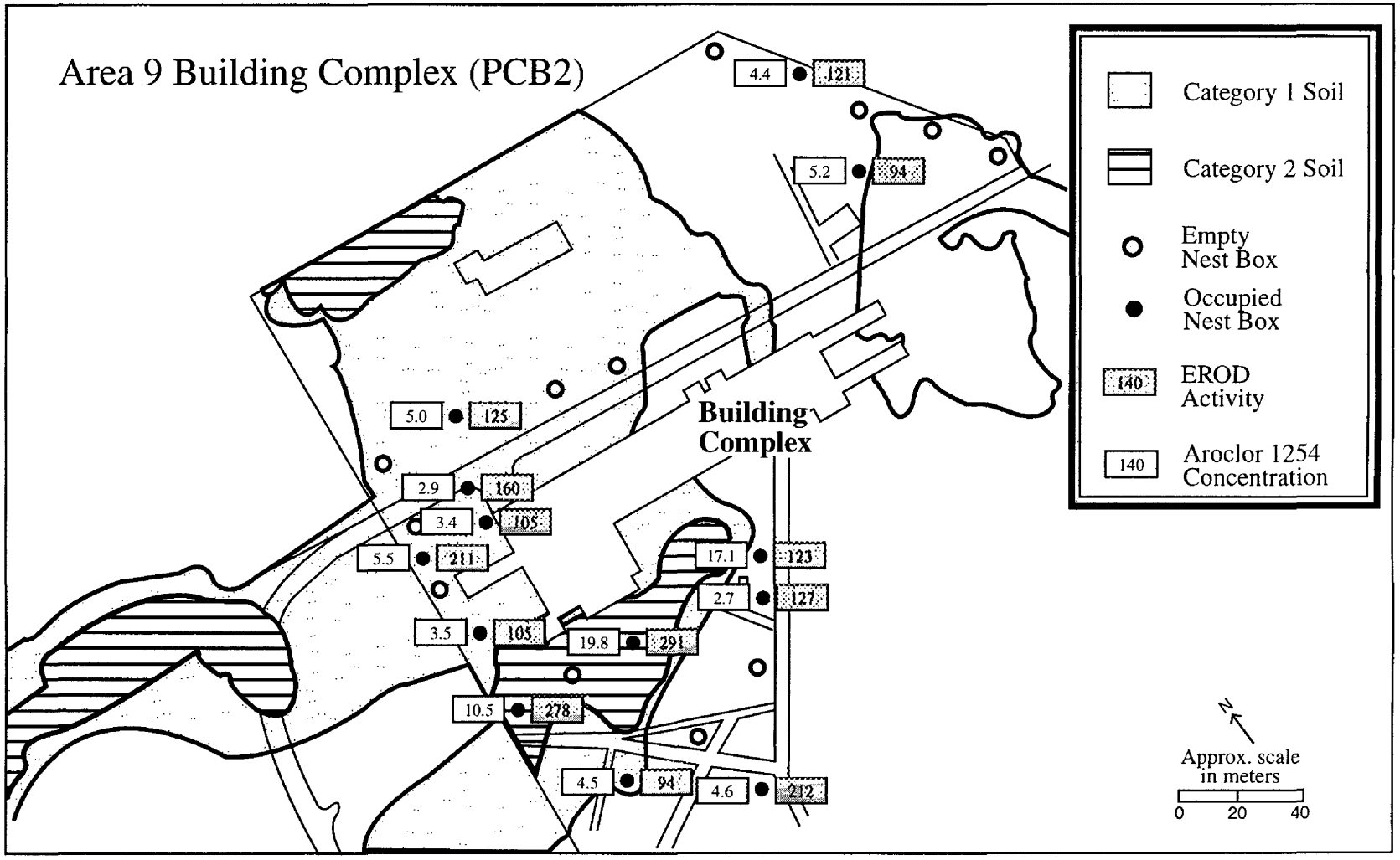


Figure 6. Aroclor 1254 concentrations (ppm [wet wt]) and EROD activity (pmol/mg protein/min) in 15-day-old starling chicks collected at the Area 9 Building Complex (PCB2) located at Crab Orchard National Wildlife Refuge, 1995-1996. Category 1 soil = Aroclor 1254 < 25, Pb< 450, and Cd < 25 ppm (dry wt), Category 2 soil = Aroclor 1254 $\geq$ 25, Pb < 450, and Cd < 25 ppm (dry wt).

Table 9. Mean (± SE) Aroclor 1254 and PCB congener (CB) concentrations (ppm [wet wt]) in homogenated starling eggs collected from contaminated sites (PCB2 and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

CB	PCB2 ($n = 27$) $\bar{x}$±SE (% of Sum of CBs)		MET ($n = 10$) $\bar{x}$±SE (% of Sum of CBs)		REF ($n = 27$) $\bar{x}$±SE (% of Sum of CBs)		P-Value[3]
31	0.05 ± 0.01	(1)	N.Q.[4]		N.Q.		N.A.[5]
52	0.32 ± 0.08	(4)	N.Q.		N.Q.		N.A.
49	0.10 ± 0.02	(1)	N.Q.		N.Q.		N.A.
44	0.07 ± 0.02	(1)	N.Q.		N.Q.		N.A.
101	1.12 ± 0.29	(13)	0.04 ± 0.01	(8)	N.Q.		N.A.
99	1.03 ± 0.22	(12)	0.03 ± 0.01	(7)	0.03 ± 0.01	(7)	<0.001
87	0.27 ± 0.09	(3)	N.Q.		N.Q.		N.A.
110	0.38 ± 0.12	(4)	N.Q.		N.Q.		N.A.
118	0.98 ± 0.27	(11)	0.03 ± 0.01	(6)	N.Q.		N.A.
153	1.74 ± 0.34	(20)	0.09 ± 0.02	(19)	0.04 ± 0.01	(8)	<0.001
105	0.37 ± 0.11	(4)	N.Q.		N.Q.		N.A.
138	1.44 ± 0.27	(16)	0.07 ± 0.02	(15)	0.02 ± 0.00	(4)	<0.001
183	0.07 ± 0.01	(1)	N.Q.		N.Q.		N.A.
128	0.36 ± 0.08	(4)	N.Q.		N.Q.		N.A.
180	0.34 ± 0.06	(41)	0.05 ± 0.02	(11)	0.03 ± 0.00	(5)	<0.001
AR1254	13.6 ± 2.9[a]		1.1 ± 0.2[b]		1.6 ± 0.3[b]		<0.001[6]

[1]PCB2 = Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]Student's t-test between PCB2 and REF (value of ½ detection limit used if concentration below the detection limit of 0.02 ppm).
[4]N.Q. = <50% of samples had quantifiable concentrations of the CB.
[5]N.A. = low percentage of samples with detectable concentrations precluded meaningful statistical comparisons.
[6]ANOVA.
[a]Means with the same letters are not different ($P > 0.05$).

quantified in ≥50% of eggs from the PCB2 and 1 coplanar CB (118) was quantified in ≥50% of eggs collected from MET (Table 9). CBs 138, 153, and 180 accounted for 34% of the total congeners at REF1 with CBs 153 and 180 being predominant. CBs 99 and 153 were the only CBs quantified in ≥50% of eggs collected from REF2, but these were not considered predominant because they each accounted for <10% of the total CBs (Table 9). Mean concentrations of Aroclor 1254 and CBs 99, 138, 153, and 180 were greater ($P \leq 0.05$) in eggs collected from PCB2 compared to the reference sites (Table 9).

Pre-15-Day-Old Chicks

Of the 34 CBs analyzed, 41, 6, and 3% were detected in ≥50% of pre-15-day-old chicks collected from PCB sites, MET, and REF1, respectively. None of the CBs quantified were detected in ≥50% of pre-15-day-old chicks collected from REF2 (Table 10). One pre-15-old chick was collected from PCB1 and Aroclor 1254 and CB concentrations in this chick were similar to those observed in pre-15-day-old chicks collected from PCB2, therefore concentrations in PCB1 and PCB2 were pooled. CBs 99, 101, 138, 153, and the coplanar CB 118 accounted for 69% of the total congeners at PCB sites (Table 10). CBs 153 and 138 at MET and CB 44 at REF1 were quantified in ≥50% of pre-15-day-old chicks collected from these sites, but only CB 153 was predominant, accounting for >10% of total CBs in pre-15-day-old chicks at MET (Table 10). Mean Aroclor 1254 concentrations were greater ($P = 0.001$) in pre-15-day-old chicks collected from PCB sites compared to reference and MET sites (Table 10).

Adults

In adults, 50, 21, and 21% of the 34 CBs analyzed were detected in ≥50% of the samples collected from PCB, MET, and REF1, respectively. None of the CBs quantified were detected in ≥50% of adults collected from REF2. One adult was collected from PCB1 and Aroclor 1254 and CB concentrations in this adult were similar to those observed in adults collected from PCB2; therefore concentrations in PCB1 and PCB2 were pooled. CBs 99, 101, 138 and 153 were predominant at PCB sites, and CBs 138, 153 and 180 were predominant at REF1 and MET sites (Table 11).

Seven CBs were quantified in ≥50% of adults collected from both PCB and reference sites, and all but CB 52 were greater ($P \leq 0.05$) at PCB sites (Table 11). Mean Aroclor 1254 concentrations also were greater ($P < 0.001$) at PCB sites compared to references and MET sites (Table 11). EROD activity was not correlated ($r = -0.058$, $P = 0.808$) with Aroclor 1254 concentrations, and did not differ ($P = 0.113$) among sites (Table 11).

Table 10. Mean (± SE) Aroclor 1254 and PCB congener (CB) concentrations (ppm [wet wt]) in whole body homogenates of pre-15-day-old starling chicks collected from contaminated sites (PCB2 and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

CB	PCB ($n = 25$) x̄±SE	(% ∑ CBs)	MET ($n = 13$) x̄±SE	(% ∑ CBs)	REF ($n = 25$) x̄±SE (% CBs)		P-Value[3]
52	0.13 ± 0.02	(4)	N.Q.[4]		N.Q.		N.A.[5]
49	0.05 ± 0.01	(2)	N.Q.		N.Q.		N.A.
44	N.Q.		N.Q.		0.02 ± 0.01	(8)	N.A.
101	0.39 ± 0.08	(13)	N.Q.		N.Q.		N.A.
99	0.30 ± 0.07	(10)	N.Q.		N.Q.		N.A.
87	0.09 ± 0.02	(3)	N.Q.		N.Q.		N.A.
110	0.14 ± 0.03	(5)	N.Q.		N.Q.		N.A.
151	0.03 ± 0.01	(1)	N.Q.		N.Q.		N.A.
118	0.37 ± 0.07	(12)	N.Q.		N.Q.		N.A.
153	0.51 ± 0.13	(17)	0.05 ± 0.02	(14)	N.Q.		N.A.
105	0.11 ± 0.03	(4)	N.Q.		N.Q.		N.A.
138	0.50 ± 0.13	(17)	0.02 ± 0.00	(6)	N.Q.		N.A.
128	0.07 ± 0.03	(2)	N.Q.		N.Q.		N.A.
180	0.08 ± 0.03	(3)	N.Q.		N.Q.		N.A.
170	0.05 ± 0.02	(2)	N.Q.		N.Q.		N.A.
AR1254	5.0 ± 1.3		0.7 ± 0.2		0.6 ± 0.1		0.001

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]ANOVA.
[4]N.Q. = <50% of samples had quantifiable concentrations of the CB.
[5]N.A. = low percentage of samples with detectable concentrations precluded meaningful statistical comparisons.
[a]Means with the same letters are not different ($P > 0.05$).

Table 11. Mean (± SE)Aroclor 1254 and PCB congener (CB) concentrations (ppm [wet wt]) in whole body homogenates of starling adults collected from contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

CB	PCB (n = 8) x̄±SE	(% ∑ CBs)	MET (n = 7) x̄±SE	(% ∑ CBs)	REF (n = 5) x̄±SE	(% ∑ CBs)	P-Value[3]
52	0.27 ± 0.11	(3)	N.Q.[4]		0.02 ± 0.01	(3)	0.057
49	0.07 ± 0.03	(1)	N.Q.		N.Q.		N.A.[5]
101	1.06 ± 0.42	(12)	0.04 ± 0.01	(6)	0.02 ± 0.01	(4)	0.044
99	1.19 ± 0.42	(13)	0.06 ± 0.02	(8)	0.02 ± 0.01	(5)	0.026
87	0.14 ± 0.07	(2)	N.Q.		N.Q.		N.A.
110	0.17 ± 0.07	(2)	N.Q.		N.Q.		N.A.
118	0.58 ± 0.17	(6)	0.03 ± 0.01	(5)	0.01± 0.00	(3)	0.012
153	2.02 ± 0.59	(22)	0.17 ± 0.04	(24)	0.08 ± 0.05	(19)	0.013
105	0.09 ± 0.02	(1)	N.Q.		N.Q.		N.A.
138	1.99 ± 0.60	(22)	0.13 ± 0.04	(19)	0.06 ± 0.03	(13)	0.014
183	0.07 ± 0.02	(1)	N.Q.		N.Q.		N.A.
128	0.40 ± 0.13	(4)	0.03 ± 0.01	(4)	N.Q.		N.A.
156/200	0.08 ± 0.05	(1)	N.Q.		N.Q.		N.A.
180	0.48 ± 0.13	(5)	0.08 ± 0.03	(12)	0.08 ± 0.05	(18)	0.020
170	0.21 ± 0.06	(2)	N.Q.		N.Q.		N.A.
201	0.04 ± 0.01	(1)	N.Q.		N.Q.		N.A.
203	0.04 ± 0.01	(1)	N.Q.		N.Q.		N.A.
AR1254	15.9 ± 5.3[a]		1.5 ± 0.4[b]		0.9 ± 0.3[b]		0.014[6]
EROD	275.0 ± 68.3		508.4 ± 113.3		279.6 ± 41.2		0.113

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]Students t-test between PCB and REF (value of ½ detection limit used if concentration below the detection limit of 0.02 ppm).
[4]N.Q. = <50% of samples had quantifiable concentrations of the CB.
[5]N.A. = low percentage of samples with detectable concentrations precluded meaningful statistical comparisons.
[6]ANOVA.
[a]Means with the same letters are not different (P > 0.05).

METALS

15-Day-Old Chicks

Mean Cd concentrations in kidney tissue collected during 1995 and 1996 from 15-day-old chicks were not different ($P = 0.282$) among sites (Table 12). Pb was detected in 70% of kidney samples from MET, but only 24 and 33% of kidney samples from reference and PCB sites, respectively. Because <50% of samples collected from reference and PCB sites had quantifiable concentrations of Pb in kidney tissues, concentrations were not statistically compared among sites (Table 12). Cr and Hg were quantified in <50% of samples, precluding meaningful comparisons for these contaminants (Table 12).

Feather tissue exhibited a different pattern of accumulation than kidney tissue in 15-day-old chicks (Table 13). Pb concentrations in feathers collected from chicks at the reference sites were significantly greater ($P = 0.019$) than concentrations in chicks from PCB sites, but did not differ from feather Pb concentrations in 15-day-old chicks collected from MET (Table 13). Fifty-nine and 53% of samples at reference and PCB sites, respectively, contained quantifiable concentrations of Cr, and means at these sites did not differ ($P = 0.076$) (Table 13). Cd was only detected in 13 samples and Hg was absent in all feathers precluding comparison of these metals among sites (Table 13).

Remediation of MET occurred during the summer between 1995 and 1996 breeding seasons. MET kidney Cd concentrations differed ($P < 0.001$) from reference and PCB sites in 1995, but not in 1996 (Table 14). Kidney Cd concentrations in MET 15-day-old chicks were reduced ($P < 0.001$) from 0.083 to 0.037 ppm (wet wt) from 1995 to 1996 (Table 14), while no between year differences were detected in Pb, Cr, and Hg levels.

Eggs

Very few metals were detected in collected eggs. One sample from PCB2 had a quantifiable concentration of Pb (0.42 ppm [wet wt]), and another egg from this site contained Cr (0.10 ppm [wet wt]). Cr also was detected in 1 reference site egg (0.11 ppm [wet wt]), and Cd and Hg were not detected in any eggs.

Pre-15-Day-Old Chicks

No metals were detected in ≥50% of kidneys collected from pre-15-day-old chicks at study sites, which precluded statistical comparisons among sites (Table 15). Of the metals analyzed, Cd was detected in the greatest percentage of pre-15-day-old chicks (20, 33, and 36% of samples from reference, MET, and PCB sites, respectively) (Table 15). Visual comparison of means (0.04, 0.05, and 0.05 at reference, MET, and PCB sites, respectively) suggests that accumulation of Cd was similar among sites. Hg was not detected in any pre-15-day-old chicks (Table 15).

Table 12. Mean metals concentrations (ppm [wet wt]) in kidney tissue of 15-day-old chicks collected at contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

	REF (n = 101)		MET (n = 37)		PCB (n = 54)		
METAL	$\bar{x} \pm$ SE[3] (range)	% Detect[4]	$\bar{x} \pm$ SE (range)	% Detect	$\bar{x} \pm$ SE (range)	% Detect	P-Value[5]
Pb	0.47 ± 0.09	24	0.44 ± 0.07	70	0.43 ± 0.13	33	N.A.[6]
	(0.09-1.59)		(0.19-1.64)		(0.19-2.67)		
Cd	0.03 ± 0.00	75	0.05 ± 0.01	100	0.05 ± 0.03	69	0.282
	(0.017-0.075)		(0.019-0.150)		(0.017-0.990)		
Cr	0.17 ± 0.04	15	--	0	1.66 ± 1.11	15	N.A.
	(0.08-0.70)		(--)		(0.08-8.96)		
Hg	0.18 ± 0.05	3	0.11	3	0.24 ± 0.03	6	N.A.
	(0.11-0.27)		(0.11)		(0.20-0.30)		

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]Means calculated for samples with concentrations above the minimum detection limit (Pb = 0.185, Cd = 0.017, Cr = 0.077, and Hg = 0.1005 ppm [wet wt]).
[4]Percent of samples with detectable concentrations.
[5]ANOVA
[6]N.A. = low percentage of samples with detectable concentrations precluded meaningful statistical comparisons.

Table 13. Mean metals concentrations (ppm [dry wt]) in feather tissue of 15-day-old chicks collected at contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

	REF (n = 101)		MET (n = 37)		PCB (n = 54)		
METAL	$\bar{x} \pm$ SE[3] (range)	% Detect[4]	$\bar{x} \pm$ SE (range)	% Detect	$\bar{x} \pm$ SE (range)	% Detect	P-Value
Pb	4.27 ± 0.32^{a}	88	3.66 ± 0.44^{ab}	60	2.97 ± 0.26^{b}	86	0.019[5]
	(1.16-16.82)		(1.24-9.65)		(1.05-8.83)		
Cd	2.79 ± 2.61	2	0.71 ± 0.12	29	--	0	N.A.[6]
	(0.18-5.40)		(0.31-1.70)		(--)		
Cr	0.51 ± 0.08	59	0.32 ± 0.09	23	0.33 ± 0.07	58	0.076[7]
	(0.04-2.54)		(0.10-0.80)		(0.07-2.00)		
Hg	--	0	--	0	--	0	N.A.
	(--)		(--)		(--)		

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex references sites).
[3]Means calculated for samples with concentrations above the minimum detection limit (Pb = 0.172, Cd = 0.134, Cr = 0.145, Hg = 0.928 ppm [dry wt]).
[4]Percent of samples with detectable concentrations.
[5]ANOVA
[6]N.A. = low percentage of samples with detectable concentrations precluded meaningful statistical comparisons.
[7]Student's t-test between REF and PCB.
[a]Means with the same letters are not different ($P > 0.05$).

Table 14. Comparison of cadmium concentrations (ppm [wet wt]) between 1995 and 1996 in kidney tissue from 15-day-old starlings collected from contaminated sites (MET and PCB)[1] located on Orchard National Wildlife Refuge (CONWR), and reference sites (REF)[2].

	1995			1996			
Site	$\bar{x}$	SE	% Detect	$\bar{x}$	SE	% Detect	*P*-Value[3]
MET	0.083[a]	0.008	100	0.037[b]	0.006	100	<0.001
REF	0.028[b]	0.002	100	0.034[b]	0.003	72	0.052
PCB	0.031[b]	0.005	86	0.059[b]	0.005	66	0.378
ANOVA	P<0.001			P = 0.435			

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]Students *t*-test.
[a]Means with same letters are not different ($P > 0.05$).

Table 15. Mean metals concentrations (ppm [wet wt]) in kidney tissue of pre-15-day-old chicks collected at contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

	REF ($n = 20$)		MET ($n = 9$)		PCB ($n = 14$)		
METAL	$\bar{x} \pm$ S.[3] (range)	% Detect[4]	$\bar{x} \pm$ SE (range)	% Detect	$\bar{x} \pm$ SE (range)	% Detect	*P*-Value
Pb	0.41 ± 0.11	35	0.20	11	2.29	7	N.A.[5]
	(0.21-1.02)		(0.20)		(2.29)		
Cd	0.04 ± 0.01	20	0.05 ± 0.03	33	0.05 ± 0.02	36	N.A.
	(0.02-0.06)		(0.02-0.10)		(0.02-0.12)		
Cr	0.17 ± 0.06	10	1.27	11	0.09 ± 0.01	14	N.A.
	(0.11-0.23)		(1.27)		(0.09-0.10)		
Hg	--	0	--	0	--	0	N.A.
	(--)		(--)		(--)		

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]Means calculated for samples with concentrations above the detection limit (Pb = 0.185, Cd = 0.017, Cr = 0.077, and Hg = 0.1005 ppm [wet wt]).
[4]Percent of samples with detectable concentrations.
[5]N.A. = low percentage of samples with detectable concentrations precluded meaningful statistical comparisons.

Adults

Cd and Hg were quantified in >65% of adult kidney tissue samples; however, mean concentrations did not differ among sites (Table 16). Pb was detected in <50% of samples from PCB and reference sites, but in >85% of adult samples from MET (Table 16). Cr was quantified in <50% of samples from all sites (Table 16).

Pb was detected in >50% of adult feather samples collected from each study site, however concentrations did not differ ($P = 0.907$) among sites (Table 17). Cd was detected in <50% of adult feathers collected at reference and PCB sites, and 71% of samples collected at MET (Table 17). Cr was detected in fewer than 50% of adult feather samples at reference and MET sites, but was detected in 75% of samples collected at PCB sites (Table 17). Hg was not quantified in any adult feathers collected (Table 17).

Table 16. Mean metals concentrations (ppm [wet wt]) in kidney tissue of adult starlings collected at contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

	REF (n = 5)		MET (n = 7)		PCB (n = 8)		
METAL	$\bar{x} \pm$ SE[3] (range)	% Detect[4]	$\bar{x} \pm$ SE (range)	% Detect	$\bar{x} \pm$ SE (range)	% Detect	P-Value[5]
Pb	0.36 ± 0.04	40	0.45 ± 0.15	86	0.58 ± 0.29	25	N.A.[6]
	(0.32-0.41)		(0.20-1.14)		(0.29-0.87)		
Cd	0.56 ± 0.17	100	0.82 ± 0.13	100	0.81 ± 0.13	100	0.428
	(0.03-1.00)		(0.48-1.47)		(0.39-1.64)		
Cr	0.16 ± 0.05	40	--	0	0.70 ± 0.41	25	N.A.
	(0.10-0.21)		(--)		(0.28-1.10)		
Hg	0.18 ± 0.01	75	0.15 ± 0.00	100	0.18 ± 0.04	67	0.700
	(0.15-0.20)		(0.14-0.16)		(0.10-0.24)		

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]Means calculated for samples with concentrations above the detection limit (Pb = 0.185, Cd = 0.017, Cr = 0.077, and Hg = 0.1005 ppm [wet wt]).
[4]Percent of samples with detectable concentrations.
[5]ANOVA
[6]N.A. = low percentage of samples with detectable concentrations precluded meaningful statistical comparisons.

Table 17. Mean metals concentrations (ppm [dry wt]) in feather tissue of adult starlings collected at contaminated sites (PCB and MET)[1] located on Crab Orchard National Wildlife Refuge (CONWR) and 2 reference sites (REF)[2], 1995-1996.

	REF ($n = 5$)		MET ($n = 7$)		PCB ($n = 8$)		
METAL	$\bar{x} \pm$ SE[3] (range)	% Detect[4]	$\bar{x} \pm$ SE (range)	% Detect	$\bar{x} \pm$ SE (range)	% Detect	*P*-Value
Pb	5.11 ± 0.88	80	4.62 ± 0.97	71	5.54 ± 1.79	88	0.907[5]
	(2.92-7.20)		(2.65-6.95)		(2.07-15.69)		
Cd	0.15 ± 0.03	40	0.42 ± 0.11	71	0.24 ± 0.04	25	N.A.[6]
	(0.12-0.17)		(0.18-0.74)		(0.20-0.28)		
Cr	0.98 ± 0.83	40	1.48 ± 0.25	29	0.47 ± 0.21	75	N.A.
	(0.15-1.81)		(1.23-1.73)		(0.08-1.49)		
Hg	--	0	--	0	--	0	N.A.
	(--)		(--)		(--)		

[1]PCB = Area 9 Landfill and Area 9 Building Complex and MET = Old Refuge Shop.
[2]One reference site was located on CONWR and 1 was located 32 km west of CONWR (REF = Area P and Annex reference sites).
[3]Means calculated for samples with concentrations above the detection limit (Pb = 0.172, Cd = 0.134, Cr = 0.145, Hg = 0.928 ppm [dry wt]).
[4]Percent of samples with detectable concentrations.
[5]ANOVA
[6]N.A. = low percentage of samples with detectable concentrations precluded meaningful statistical comparisons.

DISCUSSION

PHYSIOLOGICAL AND BIOMARKER RESPONSES

In the current study, mean fledging success at reference and MET sites (76 and 65%, respectively) was similar to that reported by Kessel (1957) (76%) and DeHaven and Guarino (1970) (71%) for starlings nesting at uncontaminated sites, but was reduced at PCB sites (42%). Reference and MET nests fledged approximately 3 chicks per nest, while PCB nests fledged an average of 2 chicks per nest. Similarly, Kubiak et al. (1989) reported that the average fledged young per pair of Forster's terns was 1 bird less at PCB contaminated sites in Green Bay compared to their reference area. McArthur et al. (1983) dosed ring doves with a mixture of organochlorines (low dose: 8 ppm Aroclor 1254, 1.64 ppm DDE, 0.297 ppm mirex, and 0.0954 ppm photo-mirex; high dose: 23.03 ppm Aroclor 1254, 4.61 ppm DDE, 0.897 ppm mirex, and 0.324 ppm photo-mirex) and reported a dose related decrease (15% in low dose and 50% in high dose groups) in the number of ring dove chicks fledged per nest compared to untreated birds.

In addition to reduced fledging success, chicks from second broods at contaminated sites were heavily infested with mites and suffered anemia and high mortality (100% at PCB2 and 67% at MET). Although heavy mite infestation also was noted at reference sites during second broods, chicks from reference nests did not show anemia. DeHaven and Guarino (1970) reported an 18% decrease in nesting success from first to second clutches in starlings nesting in an uncontaminated area near Denver, Colorado, and attributed this decline to infestation by the chicken mite (*Dermanyssus gallinae*). Kessel (1957) reported a 13% decrease in fledging success for second nest attempts by starlings in an uncontaminated area near Ithaca, New York, but attributed this decline in success to decreased fitness of second broods. Results of the current study indicated that although mite infestation and a reduction in fitness of second broods may have contributed to a decrease in nesting success at contaminated sites, they were not the only factors involved in this decrease. The marked contrast between contaminated and reference fledging success suggest that contaminants may have been a major contributing factor in the observed decrease in nest success. Grassman et al. (1996) reported immunosuppression (suppressed T-cell-mediated immunity) in pre-fledgling Caspian terns and herring gulls exposed to organochlorine (PCBs, dioxins, and DDE) contamination around the Great Lakes area (Saginaw Bay, western Lake Erie, and Hamilton Harbor). This immunosuppression was most closely associated with PCB concentrations in Caspian tern (14.18-27.45 ppm [wet wt]) and herring gull (6.57-7.73 ppm [wet wt]) eggs. Therefore, mortality in young starling chicks observed in the current study may have been associated with sub-lethal exposure that resulted in impaired immunofunction and increased susceptibility to effects of ectoparasites. However, data from the current study are inconclusive and additional study would be necessary to substantiate this speculation.

Despite these deleterious effects, the largest impact of contaminant exposure was observed in second nests, and previous reports have indicated that fledglings from second nests add little to the total population of starlings (Kessel 1957, DeHaven and Guarino 1970).

Therefore effects seen in the current study will most likely have little effect on the starling population. However, other less prolific avian species exposed to contamination at the sites studied may experience adverse population effects.

Clutch size in the current study (5 eggs per nest) is similar to that reported by others for the starling (Kessel 1957, DeHaven and Guarino 1970, Grue et al. 1986). McKee (1995) reported a significant decrease in the percent starling eggs that hatched at the PCB1, however, no difference among sites was observed in the current study. Decreased hatchability has been reported in Forster's terns nesting near Green Bay, Wisconsin, and was correlated with PCB concentrations (Kubiak et al. 1989), however Grue et al. (1986) reported no differences in either hatching or fledging success in starlings with elevated lead concentrations in tissues compared to reference starlings. Hatching success in the current study (84% overall hatch rate) are consistent with Kessel (1957) who reported an 86% hatching success in starlings nesting at uncontaminated sites in Ithaca, New York, and DeHaven and Guarino (1970) who reported an 84% hatching success in starlings nesting at uncontaminated sites in Denver, Colorado.

In addition to decreased fledging success at PCB sites, differences in liver EROD activity also were noted. EROD activity was greater at PCB sites compared to reference sites, but not the MET site , and was positively associated ($r = 0.383$, $P < 0.001$) with Aroclor 1254 concentrations. This correlation was slightly weaker than that reported by Melancon et al. (1994) ($r = 0.537$) in 15-day-old starling nestlings collected in 1990 from the PCB1 ($n = 31$) and the REF1 ($n = 39$) sites. Reference 15-day-old starlings analyzed by Melancon et al. (1994) had a mean EROD activity of 68.35 pmol/mg protein/min, that increased approximately 1.6-fold in chicks collected at PCB1 (108.10 pmol/mg protein/min). Fifteen-day-old starling chicks ($n = 3$) collected from a nest located on PCB1 in the current study had a greater mean Aroclor 1254 concentration (52.5 ppm [wet wt]) than that reported by McKee (1995) (2.65 ppm [wet wt]) and a correspondingly greater EROD activity (400 pmol/mg protein/min) than reported by Melancon et al. (1994) (170 pmol/mg protein/min).

Studies of other avian species also reported associations between EROD activity and PCB concentrations, although these reports are conflicting. EROD induction in pipping black-crowned night-heron (*Nycticorax mycticorax*) embryos was significantly correlated ($r^2 = 0.42$) with PCB concentrations (Rattner et al. 1994). This observation, coupled with no observed effects in night-heron embryos with the greatest PCB concentrations ($\bar{x} = 9.32$, range = 2.4-53 ppm [wet wt]), caused Rattner et al. (1994) to conclude that P450 induction precedes other deleterious responses such as impaired growth or reproductive failure, and is therefore a good early warning biomarker of environmental exposure to PCBs. Conversely, EROD activity was not correlated with PCB concentration in a later study with black-crowned night heron nestlings from Green Bay, Wisconsin, and San Francisco Bay, California (Rattner et al. 1996), or in 21-day-old nestling herring gulls from Great Island, Newfoundland, Canada (Peakall et al. 1986). This led Rattner et al. (1996) to conclude that cytochrome P450 monooxygenase activity was not a robust biomarker of organochlorine exposure in heron nestlings. In the current study, EROD activity in 15-day-old chicks was greater at PCB sites compared to reference sites; however, the

correlation between EROD activity and PCB concentrations was relatively weak. Additionally, many samples from MET and reference sites with low PCB concentrations had large values for EROD activity. Although it is possible that these chicks were exposed to another EROD inducer, this weak correlation may be evidence that EROD is not a robust indicator of PCB contamination in some nestling bird species as has been previously suggested by Rattner et al. (1996).

EROD activity in adult starlings collected in the current study did not differ among sites, and was highly variable (range 54-1,062 pmol/mg protein/min). This was consistent with highly variable liver enzyme activity reported in adult yellow-legged herring gulls (*Larus cachinnaus*) during periods of sexual activity that differed from the variability in liver enzyme activity measured during sexually inactive periods (Fossi et al. 1988).

Although some variation was observed in organ weights, little differences were observed for most morphometric measurements in 15-day-old chicks, adults, and eggs. Elevated PCB concentrations have been associated with enlarged liver to body weight ratios (McArthur et al. 1983, Kubiak et al. 1989, McKee 1995); however, in the current study, mean liver to body weight ratio in 15-day-old chicks was significantly reduced at PCB sites (5.2% body wt) compared to ratios at reference sites (5.8% body wt). McKee (1995) also reported decreased tarsus length in 15-day-old chicks collected from PCB1 compared to REF1 chicks, but tarsus length did not vary among sites in the current study.

Mean clutch weights at day 3 and day 9 in nestling starlings were greater in the current study (20 and 62 g, respectively) than those reported by Kessel (1957) (14 and 50 g, respectively). However, mean clutch weight at day 15 (65 g) in the current study was similar to that reported by Kessel (64 g, 1957). In most other studies (McArthur et al. 1983, Kubiak et al. 1989), reduction in reproductive success was accompanied by a decrease in fledgling body weight. In the current study and in McKee's (1995) study, 15-day-old starling chick weights were not reduced at sites characterized by decreased nesting success.

Egg size in the current study (21.2 x 29.2 mm) is consistent with Kessel (1957) who reported that North American starling eggs averaged 21.1 x 29.2 mm; however, mean egg weight (5.7 g) was less than that reported by Kessel (1957) (7.0 g) and Ricklefs (1977) (7.2 g). This difference in egg weight is likely due to the time of collection. Ricklefs (1977) collected newly laid eggs from incomplete clutches, whereas eggs in the current study were collected 5 days after the last chick in the clutch hatched. Kessel (1957) reported a 10-12% loss in weight of starling eggs through the course of incubation, which accounts for much of this discrepancy. The 7% reduction in egg shell thickness at reference sites compared to PCB sites in the current study is largely unexplained, however there were no observed detrimental effects associated with egg shell thinning (e.g., egg cracking) in the current study.

Fifteen-day-old chick sex ratios at the contaminated sites (59% females) were similar to reported starling sex ratios (57% females) (Kessel 1957) with a predominance of females at

fledging; however, reference sites had either more males than females or equal numbers of males and females at fledging. Kessel (1957) indicated that female nestlings as well as female fledglings have a higher mortality rate resulting in an adult population consisting of 42% females. Similarly, Feare (1984) reported that the sex ratio in adult starlings favored males (2:1). Kessel (1957) convincingly argued that sex ratio estimates in adult starling populations are biased because they are usually collected in the winter when starlings are flocking or in early spring at the beginning of the breeding season. Male and female starlings show little plumage differentiation during the winter making sex determination difficult, and males begin arriving on the breeding grounds prior to the females creating periods of unbalanced sex ratios in the breeding population (Kessel 1957).

BEHAVIORAL EFFECTS

In the current study, the number of nests initiated, the percent of nests completed, and the number of nests with eggs laid were similar among 4 of the 5 sites. The 1 exception was PCB1, in which only 9% of nests initiated went to full cup, indicating abandonment in the early stages of nest construction. Koval et al. (1987) reported similar nest abandonment in the early stages of nest construction in PCB-treated (Aroclor 1254) mourning doves (*Zenaida macroura*). McKee (1995) also reported nest abandonment behavior in starlings nesting at PCB1 in 1991, although this abandonment occurred after eggs were laid.

In addition to behavioral effects observed during nest initiation, adult starlings nesting at PCB and MET sites made fewer foraging trips per hour when feeding chicks than reference adults. Reference adults averaged foraging trips every 4 min, but adults at PCB sites only made foraging trips every 5.5 min. Foraging trips at reference sites were similar to those reported by Bogucki (1972) for nesting starlings (1 foraging trip per 3.1 min). Bogucki (1972) also reported that foraging trips are at their lowest during the first few days post-hatch. To test this suggestion, all observations in the current study were divided into 3 groups; observations made during the first 5 days post-hatch, observations made during days 6-10 post-hatch, and observations made on days 11-15 post-hatch. Results from this analysis (foraging trips every 6.1 min during days 1-5, every 4.0 min during days 6-10, and every 3.4 min during days 11-15) support Bogucki's suggestion.

It also is interesting to note that the number of foraging trips per hour made by adult starlings during the first 5 days post-hatch did not differ between reference and PCB sites; however, foraging trips per hour made during days 6-10 post-hatch were significantly greater at reference sites compared to PCB sites. The number of foraging trips per hour actually decreased from days 6-10 to days 11-15 post-hatch at the PCB sites. The depressed foraging activity at PCB sites suggests that PCB contamination at these sites may induce reductions in parental care. Additionally, parent foraging activity in avian species is primarily stimulated by begging intensity of the nestlings (Bogucki 1972); therefore, reduced foraging behavior in adult starlings in the current study also may be attributable to decreased begging behavior in PCB contaminated chicks.

Behavior effects similar to those observed in the current study (decrease in number of times per hr adults came to nest box to feed chicks) have previously been reported in starlings dosed with an organophosphate (OP) pesticide. Grue et al. (1982) observed that OP-dosed female starlings made fewer sorties to feed their young and remained away from their boxes for longer periods of time than controls. They concluded that parental care may be significantly reduced in songbirds due to sub-lethal exposure to OPs. Similarly, results from the current study suggest that sub-lethal exposure to PCBs (use of OPs is not allowed at CONWR) may significantly reduce parental care in songbirds.

McKee (1995) noted nest abandonment behavior during the incubation period in adult starlings nesting at PCB1, which resulted in reduced hatching success. Similarly, Peakall and Peakall (1973), Fox et al. (1978), and Kubiak et al. (1989) reported decreased hatching success in captive ring doves, Lake Ontario herring gulls, and Green Bay Forster's terns, respectively, attributable to nest abandonment or decreased parental attentiveness by organochlorine contaminated adults. Peakall and Peakall (1973) reported lower mean egg temperatures during incubation by ring doves treated with 10 ppm Aroclor 1254 compared to untreated ring doves, and observed highly variable daily egg temperature patterns in PCB-treated birds. Peakall and Peakall (1973) and Fox et al. (1978) reported greater hatching success in artificially incubated dove and herring gull eggs, respectively, compared to eggs incubated by contaminated adults.

Adult behavior prior to egg laying did not appear to differ among the 5 sites, with the exception of slightly increased maintenance behavior at PCB sites compared to the MET site. The importance of this behavior is not known. It is worth noting that in all stages of nesting (prior to egg laying, during incubation, and during the rearing of the young) adults at the PCB2 and MET sites were observed sitting on the roof of the nest box more frequently than reference adults and also appeared to demonstrate increased boldness. In fact, one adult at the PCB2 site came to feed the young during a nest check, and another frequently returned to the nest while the observer was within 2-3 m of the nest box. Adults at the MET site also were largely undisturbed by human presence, many returning to sit on the nest box by the time the observer had reached the next box (approx 15 m). Adults at the MET and PCB2 sites also were easier to trap. Many adults at these sites were captured within 30 minutes of setting up the trap, while it often took 1-2 hours to capture reference adults. Additionally, fewer adults were captured at the REF1 ($n = 2$) and REF2 ($n = 3$) sites compared to the PCB1 ($n = 7$) and MET ($n = 7$) sites, which may indicate a lack of wariness among the adult starlings at contaminated sites. Coturnix quail (*Coturnix coturnix*) chicks dosed with 200 ppm Aroclor 1254 had impaired avoidance to visual and audio stimuli compared to control chicks (Kreitzer and Heinz 1974). In addition, these chicks failed to recover normal avoidance behavior when returned to untreated food. These results and the subjective observations noted in the current study suggest that PCB exposure may impair normal avoidance behavior, that could potentially cause increased susceptibility to predation.

CONTAMINANT ACCUMULATION

The current study indicates that starlings nesting at PCB contaminated sites accumulated greater concentrations of PCBs than starlings nesting at reference sites. In nationwide surveys, starlings captured from Illinois, Kentucky, and Missouri had whole body total PCB concentrations of 0.25-3.13 ppm in 1970 and 0.042-0.23 ppm in 1974 (Martin and Nickerson 1972, White 1976). Whole body Aroclor 1254 concentrations in 15-day-old starlings (range <0.01-2.73) and adults (range 0.14-1.36) at REF1, REF2, and MET sites approximate background values estimated for Illinois, Kentucky, and Missouri. Mean Aroclor 1254 concentrations recorded in 15-day-old chicks (27.7 and 2.6 ppm at PCB1 and PCB2, respectively) and adults (6.8 ppm) collected at both PCB sites were considerably greater than reference concentrations.

McKee (1995) provided evidence that insects at PCB1 contained elevated concentrations of PCBs. Beetles (*Phyllophaga fervida* and *P. bipartita*), collected directly on the landfill, 50 m from the landfill, and 500 m from the landfill had Aroclor 1254 concentrations of 10.6 and 8.3 ppm (wet wt), 1.2 and 2.6 ppm, and 0.3 and 0.1 ppm, respectively, indicating a pattern of decreasing accumulation of PCBs in insects with increasing distance from the contaminated area (McKee 1995). Furthermore, mean starling clutch Aroclor 1254 concentrations in individual nests at PCB sites were significantly correlated ($r = 0.667$) with Aroclor 1254 concentrations in the soil where the nest box was located. Starlings generally forage within 500 meters of the nesting site (Feare 1984, Kendall et al. 1989), and in the current study, were observed feeding on the contaminated areas and in adjacent fields during the formal observation period. Therefore, results of the current study and McKee (1995) indicate that adult starlings were feeding on contaminated insects foraged from the contaminated sites, and that starling nestlings are good indicators of local (contaminated site) and specific (soil where nest box is placed) PCB contamination.

In the current study, the mean Aroclor 1254 concentration in 15-day-old chicks collected at the PCB1 site was almost 20 times greater than that reported by McKee (1995) (3 ppm [wet wt]); however, this concentration was below concentrations previously associated with mortality. Stickel et al. (1984) associated mortality of 4 bird species, including immature female starlings, with whole body (including liver) Aroclor 1254 concentrations of 172-1,120 ppm and brain concentrations in dead starlings of 179 ppm.

Mean Aroclor 1254 concentrations in eggs also were greater at the PCB2 site (13.6 ppm [wet wt]; range <0.24-57.5 ppm) compared to reference sites (1.6 ppm [wet wt]; range <0.24-5.7 ppm). Reproductive effects, primarily reduced hatching success, have been associated with egg PCB wet weight concentrations of 16 ppm in ring doves (Peakall and Peakall 1973), >5 ppm Aroclor 1254 in chickens (Platonow and Reinhart 1973), 22.2 ppm in Forster's terns (Kubiak et al. 1989), and 5.3 ppm in double-crested cormorants (*Phalacrocorax auratus*) (Larson et al. 1996). Additionally, concentrations ≥15 ppm (wet wt) in eggs resulted in increased embryotoxicity accompanied by edema, growth retardation, and deformities of the leg, bill, and

neck in chickens (McLaughlin et al. 1963, Platonow and Reinhart 1973, Tumasonis et al. 1973, Cecil et al. 1974), and Forster's terns (Kubiak et al. 1989). Bill deformities were observed in cormorants from Lake Michigan, Wisconsin (egg PCB concentrations 5.3 ppm); however, other factors, such as Vitamin D deficiency, could not be ruled out as potential causes (Larson et al. 1996). Thus, PCB concentrations in starling eggs from the PCB2 site (as well as some samples at reference sites) were within the range (5.3-22.2 ppm) associated with reproductive effects in prior studies, although reduced hatching success was not observed. However, increased embryo mortality was evident at PCB2. One nest in which the entire clutch (n = 5) failed to hatch had a mean Aroclor 1254 egg concentration of 29.9 ppm (range 0.3-57.6 ppm). It is not known whether these elevated PCB concentrations or potential adult incubation anomalies were the cause of the embryo death, but similar embryo death was observed in eggs collected from the MET site that had significantly lower PCB concentrations (1.1 ppm [wet wt]) than observed at the PCB2 site (13.6 ppm [wet wt]). Dioxins, which were reported in some soil samples collected at the MET site (O'Brien and Gere 1988), may also cause embryotoxicity (Varrett 1976); however, dioxins were not detected by mass spectrometry analysis in a deformed chick, or in a pooled sample of chicks that died 1-2 days post-hatch, collected from this site. Therefore, it appears that parental care may be a primary factor in decreased embryo survival compared to contaminant concentrations within the egg; however, additive effects of contaminant concentrations in eggs and reduced parental care cannot be ruled out.

Analysis of CB patterns may be useful in determining potential adverse effects, because individual CBs elicit varying toxic responses. Coplanar CBs (e.g., mono-ortho-substituted CB 118) exhibit a toxicodynamic action by binding to the aryl hydrocarbon (Ah) -receptor and blocking estrogen binding substances (Soontornchat et al. 1994). This is an endocrine-disrupting mechanism similar to that reported for 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (Safe 1990, Safe et al. 1991) and, therefore, these CBs are often responsible for dioxin-like effects such as embryotoxicity (McFarland and Clarke 1989, Brunstrom 1990). Non-coplanar CBs (e.g., di-ortho-substituted CBs 138 and 153) are considered estrogenic (Ah receptor-independent), and cause endocrine disruption through toxicokinetic interactions with estrogen receptors and induction of biotransformation activity (Soontornchat et al. 1994, Li and Hansen 1996). These CBs can cause decreases in uterine weights (Li and Hansen 1996) and increases in pentoxyresorufin-O-deethylase (PROD) activity that may affect estrogen levels in the blood (Soontornchat et al. 1994).

CBs 138, 153, and 180 were predominant CBs in starling eggs in the current study, as they were in previous studies of bird eggs (Ormerod and Tyler 1994, Mora 1996). These CBs accounted for 45, 45, 5, and 34% of the total congeners at the PCB2, MET, REF1, and REF2 sites, respectively. CBs 138, 153, and 180 also were common in 15-day-old chicks collected from the PCB1, PCB2, and MET sites (31, 37, and 36%, respectively, of the total congeners) and adult starlings collected at PCB, MET, and REF1 sites (49, 55, and 45%, respectively, of the total congeners). These 3 CBs are resistant to atmospheric transport and environmental degradation and have previously been reported in wildlife species (Harrad et al. 1994, Mora 1996).

In general, accumulation of heavy metals in starlings collected in the current study was inconsistent, and concentrations were below those associated with adverse effects. Kidney lead concentrations were similar to those reported as normal background levels in adult common terns (*Sterna hirundo*), ring doves, and American kestrels (*Falco sparverius*) collected from uncontaminated areas or maintained under laboratory conditions (Connors et al. 1975, Kendall and Scanlon 1981, Custer et al. 1984), and were far below lead concentrations associated with mortality (200-500 ppm [dry wt]) (Longcore et al. 1974, Benson et al. 1976). Wet weight kidney lead concentrations in pre-15-day-old (0.2-2.3 ppm) and 15-day-old starlings (0.4-0.5 ppm) in the current study were below those associated with impaired growth (>6 ppm) and survival (>15 ppm) in nestling kestrels (Custer et al. 1984, Hoffman et al. 1985).

Greater lead concentrations were recorded in feathers from 15-day-old starlings collected at reference sites compared to 15-day-old chicks collected at PCB sites. Overall lead concentrations in feathers of adults and 15-day-old starlings were below those reported by Grue et al. (1986) in starlings nesting in the median of a well-traveled Maryland highway. However, 15-day-old starlings in the current study had greater overall lead concentrations in feathers than control starlings in Maryland, but adults did not (Grue et al. 1986). Grue et al. (1986) reported decreased ALAD aminolevulinic (aminolevulinie acid dehydrase) activity and depressed hematocrits in 18-day-old starlings, as well as increased nest abandonment behavior in adults in the Maryland study. Hematocrit values at reference sites (34%) in the current study were significantly reduced compared to hematocrit values at the MET site (38%), but did not differ from hematocrit readings at PCB sites (36%). Hematocrit values at all sites in the current study were greater than those reported by Grue et al. (1986) in 18-day-old starlings from contaminated and reference locations (range 27-32%) in Maryland.

Kidney cadmium concentrations in the current study ranged from 0.03 to 0.08 ppm (wet wt) in 15-day-old starling chicks, and from 0.56 to 0.82 ppm (wet wt) in starling adults. These concentrations are below background cadmium concentrations (<10 ppm [wet wt] in combined kidney and liver tissue) reported for canvasback ducks (*Aythya valisineria*) and ruddy ducks (*Oxyura jamaicensis*) (White and Kaiser 1976, White et al. 1979), and whole body cadmium concentrations in free-ranging adult starlings captured during 1971 and 1973 in Illinois, Kentucky, and Missouri (range <0.05-0.13 ppm, wet wt; Martin and Nickerson 1973, and White et al. 1977). Because information on the relationship between whole body and kidney cadmium concentrations could not be found, starling kidney concentrations in the current study cannot be accurately compared to whole body concentrations reported in previous starling studies. Greater kidney cadmium concentrations in adults were expected because cadmium has a long half-life (up to 20 years) (Friberg et al. 1974) and, therefore, accumulates with age, even in wildlife species exposed to low background concentrations (Woolf et al. 1982).

Whereas cadmium was detected in the majority of kidney samples (>69% in 15-day-old starlings at all sites and 100% of all collected adults), it was only detected in feathers from 13 of 192 15-day-old chicks. More adults (9 of 20) had detectable concentrations of cadmium in feathers; however, in all cases, these concentrations were below those recorded from adult kidney

tissue. This is consistent with reports that feathers are not good indicators of cadmium concentrations in other tissues (Gochfeld et al. 1996). No eggs in the current study had detectable cadmium concentrations. This was expected because little Cd is transferred to eggs (Sell 1975, White and Finley 1978).

Cadmium concentrations of 130-140 ppm (wet wt) in combined liver and kidney tissue have been associated with renal tubular necrosis and testicular atrophy in mallards dosed with 200 ppm dietary cadmium (White et al. 1978). Cain et al. (1983) reported decreased hematocrits (anemia) in mallard ducklings with liver cadmium concentrations of approximately 40 ppm (wet wt). Cadmium ingestion inhibits intestinal absorption of calcium, which could cause reproductive effects in laying females (Hamilton and Smith 1978). In fact, egg production was suppressed in mallards at dietary cadmium concentrations of 200 ppm (White and Finley 1978) and in chickens at concentrations of 60 (Sell 1975) and 48 ppm (Leach et al. 1979). Cadmium concentrations reported in the current study are well below those associated with adverse effects.

Mercury (total mercury) in kidney tissue was detected in 5 (3%) 15-day-old starling chicks and 11 (79%) adults, with concentrations in chicks and adults ranging from 0.1 to 0.3 (overall $\bar{x}$ = 0.057 ppm [wet wt]). These concentrations are similar to those reported in laughing gulls (*Larus atricilla*) collected from uncontaminated areas in New York which had a mean kidney mercury concentration of 0.05 ppm (wet wt) (Gochfeld et al. 1996). Kidney mercury concentrations were greater than whole body concentrations detected in free ranging adult starlings sampled in Illinois, Kentucky, and Missouri during 1970-1973 (<0.01-0.1 ppm [wet wt]) (Martin 1972, Martin and Nickerson 1973, White et al. 1977).

Mercury was not detected in eggs or in feathers collected from adults or 15-day-old starlings in the current study. This is unusual because mercury has been reported to accumulate at greater concentrations in feather tissue compared to kidney or liver tissue (Heinz 1976, Finley and Stendell 1978, Gochfeld et al. 1996). Mercury concentrations in feathers represent exposure to mercury during feather formation (Appelquist et al. 1984, Goede and deBruin 1984); therefore it is possible that adults accumulated kidney mercury concentrations while nesting at the study sites and were not exposed during the pre-breeding season molt. However, 15-day-old chicks did not accumulate quantifiable feather mercury despite active feather development. A plausible explanation for the different accumulation pattern may be related to the detection limits of the assay. In the current study, the minimum detection limits for mercury in kidney and feather tissue were 0.1 ppm and 0.93 ppm (dry wt), respectively. Therefore, detected kidney mercury concentrations in adult and 15-day-old starlings (0.11-0.24 ppm [wet wt]) were close to the minimum detection limit in kidney and were likely present, but unquantifiable, in feather tissue.

Kidney chromium concentrations were detected in <13% of all samples collected in the current study. Two kidney samples from 15-day-old chicks collected from PCB2 had chromium concentrations of 9 and 3.2 ppm (wet wt); however, all other samples ranged from 0.08 to 1.27 ppm (wet wt). Mean chromium concentrations in feathers collected from 15-day-old chicks were greater than concentrations in kidney tissue excluding the extreme outliers in kidney samples

listed above. A similar trend was observed in adults collected from reference sites; however, adults collected at PCB sites had greater chromium concentrations in kidney tissue than those in feathers. Gochfeld et al. (1996) reported concentrations of 0.02 (wet wt) and 0.07 ppm (dry wt) in kidney and feather tissue, respectively, in laughing gulls collected from an uncontaminated area in New York. He concluded that chromium (like mercury and lead) accumulates in greater concentrations in feathers than in kidney. The converse of this pattern, however, was observed in adult starlings collected at PCB sites and may have resulted from exposure to chromium after the molting stage as discussed above. Few studies describing chromium effects in wildlife have been reported; therefore, the significance of the exposure observed in the current study are unknown.

Little information is available on the combined effects of mixtures of contaminants in avian species; however, combined effects of cadmium and PCBs have been documented. Leonzio et al. (1992) reported increased accumulation of PCBs in muscle tissue and decreased PCB accumulation in liver when Japanese quail were dosed with a mixture of cadmium (100 ppm) and PCBs (100 ppm Aroclor 1260). They suggested that decreased lipid reserves in liver after exposure to cadmium caused mobilization of PCBs from this organ. In the current study, combinations of contaminants were reported in soil at the MET site (mercury, cadmium, and some dioxin) and PCB (PCBs, lead, chromium, and cadmium) sites (O'Brien and Gere 1988). Behavioral and reproductive effects were demonstrated at all contaminated sites; however, greater effects were observed at PCB sites suggesting that effects of PCBs and heavy metals may be additive.

CONCLUSIONS

Greater PCB concentrations were documented in 15-day-old and pre-15-day-old starling chicks, adults, and eggs collected at PCB sites compared to reference sites. Observed effects included an overall decrease in nesting productivity, increased mortality in second brood nestlings, increased late-stage embryo mortality, and abnormalities in adult behavior (decreased foraging trips, early stage nest abandonment, and loss of fear in adults).

Despite these deleterious effects, little effect on the starling population would be expected; however, less prolific avian species feeding at contaminated sites may experience population level declines. The current study also demonstrates that nestling starlings are effective indicators of local contamination, particularly for PCBs.

EROD activity was only weakly correlated with PCB tissue concentrations, therefore it was not an effective biomarker of PCB exposure in the current study. However, increased EROD activity at the MET site, as well as a trend towards decreased nesting and fledging success and decreased feeding activity, suggest that EROD may be indicating exposure to a contaminant not measured at this site.

Lead, cadmium, and mercury concentrations were below those associated with adverse effects in avian species. Although chromium concentrations were greater in some samples than those reported in other free-ranging birds, no known adverse effects have been reported at the observed concentrations. Greater concentrations of lead and chromium were recorded in feathers compared to kidney tissue, and greater concentrations of cadmium were recorded in kidney tissue compared to feathers. These accumulation patterns are similar to those reported in laughing gulls. Mercury was not detected in feathers, but was present in kidney tissues, which is contrary to accumulation patterns previously described for mercury accumulation in avian species. However, this was most likely related to differences in detection limits between the 2 tissue types.

Pre-remediation kidney cadmium concentrations in 15-day-old starlings collected from the MET site during 1995 were significantly greater than kidney cadmium concentrations in 15-day-old starlings collected from the MET site following remediation in 1996. Furthermore, post-remediation kidney cadmium concentrations in chicks collected from the MET site were similar to those in reference chicks. This suggests that metals remediation at this site was successful and that starlings are a good monitor of the effectiveness of remediation.

Observed nesting activity, adult starling behavior, and contaminant results in the current study indicate that starlings are useful avian models for demonstrating potential effects of exposure to environmental contaminants. Additional research is needed to substantiate some of the effects observed in the current study, including determination of adult versus chick contribution to decreased feeding activity, and objective measurement of the decreased fear observed in adults nesting at contaminated sites.

REFERENCES

Appelquist, H. S. Asbirk, and I. Drabaek. 1984. Mercury monitoring: mercury stability in bird feathers. Mar. Pollut. Bull. 15:22-24.

Ballschmiter, K., and M. Zell. 1980. Baseline studies of global pollution. I. Occurrence of organohalogens in pristine European and Antarctic environments. Int. J. Environ. Anal. Chem. 8:15-35.

Benson, W. W., D. W. Brock, J. Gabica, and M. Loomis. 1976. Swan mortality due to certain heavy metals in the Mission Lake area, Idaho. Bull. Environ. Contam. Toxicol. 15:171-174.

Bogucki, Z. 1972. Studies on the activity of starlings, *Sturnus vulgaris* Linnaeus, 1758, in the breeding season. Acta Zool. 17:97-121.

Brunstrom, B. 1990. Embryotoxicity of polycyclic aromatic hydrocarbons (PAHs) in three domestic avian species, and of PAHs and coplanar polychlorinated biphenyls (PCBs) in the common eider. Environ. Pollut. 67:133-143.

Cain, B. W., L. Sileo, J. C. Franson, and J. Moore. 1983. Effects of dietary cadmium on mallard ducklings. Environ. Res. 32:286-297.

Cecil, H. C., J. Bitman, R. J. Lillie, G. F. Fries, and J. Verrett. 1974. Embryotoxic and teratogenic effects in unhatched fertile eggs from hens fed polychlorinated biphenyls (PCBs). Bull. Environ. Contam. Toxicol. 11:489-495.

Connors, P. G., V. C. Anderlini, R. W. Risebrough, M. Gilbertson, and H. Hays. 1975. Investigations of metals in common tern populations. Can. Field-Nat. 89:157-162.

Custer, T. W., J. C. Franson, and O. H. Pattee. 1984. Tissue lead distribution and hematologic effects in American kestrels (*Falco sparverius* L.) fed biologically incorporated lead. J. Wildl. Dis. 20:39-43.

DeHaven, R. W., and J. L. Guarino. 1970. Breeding of starlings using next-boxes at Denver, Colorado. Colorado Field Ornithol. No. 8. 10pp.

Feare, C. 1984. The starling. Oxford University Press, New York, N.Y. 315pp.

Finley, M. T., and R. C. Stendell. 1978. Survival and reproductive success of black ducks fed methyl mercury. Environ. Pollut. 16:51-64.

Fossi, C., C. Leonzio, S. Focardi, and A. Renzoni. 1988. Seasonal variations in aldrin epoxidase (MFO) activity of yellow-legged herring gulls: the relationship to breeding and PCB residues. Bull. Environ. Contam. Toxicol. 41:365-370.

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

Friberg, L., M. Piscator, G. F. Nordberg, and T. Kjellstrom. 1974. Cadmium in the environment II. CRC Press. Cleveland, Oh. 248pp.

Gochfeld, M., J. L. Belant, T. Shukla, T. Benson, and J. Burger. 1996. Heavy metals in laughing gulls: gender, age and tissue differences. Environ. Toxicol. Chem. 15:2275-2283.

Goede, A. A., and M. deBruin. 1984. The use of bird feather parts as a monitor for metal pollution. Environ. Pollut. 8:281-289.

Grassman, K. A., G. A. Fox, P. F. Scanlon, and J. P. Ludwig. 1996. Organochlorine-associated immunosuppression in prefledgling Caspian terns and herring gulls from the Great Lakes: an ecoepidemiological study. Environ. Health Perspect. 104:829-842.

Grue, C. E., D. J. Hoffman, W. N. Beyer and L. P. Franson. 1986. Lead concentrations and reproductive success in European starlings nesting within highway roadside verges. Environ. Pollut., Ser. A. 42:157-182.

___, G. V. N. Powell and M. J. McChesney. 1982. Care of nestlings by wild female starlings exposed to an organophosphate pesticide. J. Appl. Ecol. 19:327-335.

Hamilton, D. L., and M. W. Smith. 1978. Inhibition of intestinal calcium uptake by cadmium and the effect of low calcium diet on cadmium retention. Environ. Res. 15:175-184.

Harrad, S. J., A. P. Stewart, R. Alcock, R. Boumphrey, V. Burnett, R. Duarte-Davidson, C. Halsall, G. Sanders, K. Waterhouse, S. J. Wild, and K. C. Jones. 1994. Polychlorinated biphenyls (PCBs) in the British environment: sinks, sources and temporal trends. Environ. Pollut. 85:131-146.

Heinz, G. 1976. Methylmercury: second-year feeding effects on mallard reproduction and duckling behaviour. J. Wildl. Manage. 40:82-90.

Hite, R. L., and M. King. 1977. Biological investigation of the crab orchard creek basin, summer 1975. Ill. Environ. Prot. Agency, Springfield, Ill. 141pp.

Hoffman, D. J., J. C. Franson, O. H. Pattee, and C. M. Bunck. 1985. Survival, growth, and accumulation of ingested lead in nestling American kestrels (*Falco sparverius*). Arch. Environ. Contam. Toxicol. 14:89-94.

Kendall, R. J., L. W. Brewer, T. E. Lacher, Jr., B. T. Marden, and M. L. Whitten. 1989. The use of starling nest boxes for field reproductive studies: Provisional guidance document and technical support document. 600/8-89/056. Environ. Prot. Agency. Natl. Tech. Inf. Serv. Public. No. PB89 195 028/AS. 82pp.

___, and P. F. Scanlon. 1981. Effects of chronic lead ingestion on reproductive characteristics of ringed turtle doves (*Streptopelia risoria*) and on tissue lead concentrations of adults and their progeny. Environ. Pollut, Ser. A 26:203-213.

Kessel, B. 1957. A study of the breeding biology of the European starling in North America. Am. Midl. Nat. 58:257-331.

Kohler, C.C., R. C. Heidinger, and T. Call. 1990. Levels of PCBs and trace metals in crab orchard lake sediment, benthos, zooplankton and fish. Hazardous Waste Research and Information Center, HWRIC RR-043. Champaign, Ill. 130pp.

Koval, P. J., HT. J. Peterle, and J. D. Harder. 1987. Effects of polychlorinated biphenyls on mourning dove reproduction and circulating progesterone levels. Bull. Environ. Contam. Toxicol. 39:663-670.

Kreitzer, J. F., and G. H. Heinz. 1974. The effect of sublethal dosages of five pesticides and a polychlorinated biphenyl on the avoidance response of Coturnix quail chicks. Environ. Pollut. 6:21-29.

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.

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

Leach, R. M., Jr., K. W. Wang, and D. E. Baker. 1979. Cadmium and the food chain: the effect of dietary cadmium on tissue composition in chicks and laying hens. J. Nutr. 109:437-443.

Lehner, P.N. 1979. Handbook of ethological methods. Garland STPM Press. New York, N.Y. 403pp.

Leonzio, C., L. Marsili, and S. Focardi. 1992. Influence of cadmium on PCB congener accumulation in quail. Bull. Environ. Toxicol. 49:686-693.

Li, M. H., and L. G. Hansen. 1996. Enzyme induction and acute endocrine effects in prepubertal female rats receiving environmental PCB/PCDF/PCDD mixtures. Environ. Health Perspect. 104:712-722.

Lobring, L. B., and B. B. Potter. 1991. Determination of mercury in tissues by cold vapor atomic absorption spectrometry. Method 245.6, rev 2.3. 600-4-91-010. U.S. Environ. Prot. Agency, Washington, D.C. 13pp.

Longcore, J. R., L. N. Locke, G. E. Bagley, and R. Andrews. 1974. Significance of lead residues in mallard tissues. U.S. Dept. of Interior, Fish and Wildl. Serv. Special Scientific Report - Wildlife No. 182.

Lubet, R.A., R.W. Nims, R.T. Mayer, J.W. Cameron, and L.M. Schechtman. 1985. Measurement of cytochrome P-450 dependent dealkylation of alkoxyphenoxazones in hepatic S9s and hepatocyte homogenates: effects of dicumarol. Mut. Res. 142:127-131.

Martin, W. E. 1972. Mercury and lead residues in starlings - 1970. Pestic. Monit. J. 6:27-32.

___, and P. R. Nickerson. 1972. Organochlorine residues in starlings - 1970. Pestic. Monit. J. 6:33-40.

___, ___. 1973. Mercury, lead, cadmium, and arsenic residues in starlings - 1971. Pestic. Monit. J. 7:67-72.

Mazel, P. 1971. Experiments illustrating drug metabolism *in vitro*. Pages 546-560 *in* B.N. La Du, H. G. Mandel, and E. L. Way, eds. Fundamentals of drug metabolism and drug disposition. Williams and Wilkins, Baltimore, Md.

McArthur, M. L. B., G. A. Fox, D. B. Peakall, and B. J. R. Philogene. 1983. Ecological significance of behavioral and hormonal abnormalities in breeding doves fed and organochlorine chemical mixture. Arch. Environ. Contam. Toxicol. 12:343-353.

McDaniel, W. 1991. Sample preparation procedure for spectrochemical determination of total recoverable elements in biological tissues. Method 200.3, rev 1.0. U.S. Environ. Prot. Agency, Washington, D.C. 7pp.

McFarland, V. A., and J. U. Clarke. 1989. Environmental occurrence, abundance, and potential toxicity of polychlorinated biphenyl congeners: considerations for a congener-specific analysis. Environ. Health Perspect. 81:225-229.

McKee, M. J. 1995. Biological monitoring of chemical contamination at crab orchard national wildlife refuge. Final Report, Prepared for U.S. Dept. of Interior, Fish and Wildl. Serv. 86pp.

McLaughlin, J., Jr., J. P. Marliac, M. J. Verrett, M. K. Mutchler, and O. G. Fitzhugh. 1963. The injection of chemicals into the yolk sac of fertile eggs prior to incubation as a toxicity test. Toxicol. Appl. Pharmacol. 5:760-771.

Melancon, M. J., J. D. Eisemann, M. E. Maxey, and M. J. McKee. 1994. Monooxygenase activity in dosed and field-collected nestling starling *(Sturnus vulgaris)*. Page 92 *in* Abstracts, 15th Annual Meeting, Society of Environmental Toxicology and Chemistry, Denver, Colo. October 30 - November 3.

Mora, M. A. 1996. Congener-specific polychlorinated biphenyl patterns in eggs of aquatic birds from the lower Laguna Madre, Texas. Environ. Toxicol. Chem. 15:1003-1010.

O'Brien and Gere Engineers, Inc. 1988. Remedial Investigation report, Volume 1: Report. O'Brien and Gere Engineers, Inc., Syracuse, N.Y. 178pp.

Ormerod, S. J., and S. J. Tyler. 1994. Inter- and intra-annual variation in the occurrence of organochlorine pesticides, polychlorinated biphenyl congeners, and mercury in the eggs of a river passerine. Arch. Environ. Contam. Toxicol. 26:7-12.

Peakall, D. B. and M. L. Peakall. 1973. Effect of a polychlorinated biphenyl on the reproduction of artificially and naturally incubated dove eggs. J. Appl. Ecol. 10:863-868.

___, R. J. Norstrom, A. D. Rahimtula, and R. D. Butler. 1986. Characterization of mixed function oxidase systems of the herring gull and its implications for bio-effects monitoring. Environ. Toxicol. Chem. 5:379-385.

Platonow, N. S., and B. S. Reinhart. 1973. The effects of polychlorinated biphenyls (Aroclor 1254) on chicken egg production, fertility and hatchability. Can. J. Compar. Med. 37:341-346.

Rattner, B. A., J. S. Hatfield, M. J. Melancon, T. W. Custer, and D. E. Tillitt. 1994. Relation among cytochrome P450, Ah-active PCB congeners and dioxin equivalents in pipping black-crowned night-heron embryos. Environ. Toxicol. Chem. 13:1805-1812.

___, M. J. Melancon, T. W. Custer, and R. L. Hothem 1996. Cytochrome P450 and contaminant concentrations in nestling black-crowned night-herons and their interrelation with sibling embryos. Environ. Toxicol. Chem. 15:715-721.

Rezanka, J. 1993. Incinerator's safety concerns congressman. Daily Egyptian, 13 September:1, 5.

Ricklefs, R. E.. 1977. Variation in size and quality of the starling egg. Auk 94:167-168.

Rowell, R. 1994. Stop Crab Orchard incinerator. Daily Egyptian, 24 January:4.

Safe, S. 1990. Polychlorinated biphenyls (PCBs) dibenzo-*p*-dioxins (PCDDs), dibenzofurans (PCDFs) and related compounds: environmental and mechanistic considerations which support the development of toxic equivalency factors (TEFs). Crit. Rev. Toxicol. 21:51-88.

___, B. Astroff, M. Harris, T. Zacharewski, R. Dickerson, R. Romkes, and L. Biegel. 1991. 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) and related compounds as antiestrogens: characterization and mechanism of action. Pharmacol. Toxicol. 69:400-409.

Sell, J. L. 1975. Cadmium and the laying hen: apparent absorption, tissue distribution and virtual absence of transfer into eggs. Poult. Sci. 54:1674-1678.

Soontornchat, S., M. H. Li, P. S. Cooke, and L. G. Hansen. 1994. Toxicokinetic and toxicodynamic influences on endocrine disruption by polychlorinated biphenyls. Environ. Health Perspect. 102:568-571.

Stickel, F., 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.

Tumasonis, C. F., B. Bush, and F. D. Baker. 1973. PCB levels in egg yolks associated with embryonic mortality and deformity of hatched chicks. Arch. Environ. Contam. Toxicol. 1:312-324.

USDI. 1992. Digest of federal resource laws of interest to the U.S. Fish and Willdlife Service. Office of Legislative Services, U.S. Dept. of Interior and U.S. Fish and Wildl. Serv. Washington, D.C. 4pp.

USEPA. 1987. Atomic absorption furnace technique. User's guide to the contract laboratory program (CLP-M), cadmium determination (Method 212.2), chromium determination (Method 218.2), lead determination (Method 239.2), U.S. Environ. Prot. Agency, Washington, D.C. 4pp.

___. 1989. U.S. EPA proposed cleanup action: Crab Orchard National Wildlife Refuge superfund site. U.S. Environ. Prot. Agency Fact Sheet, Office of Public Affairs, Region 5, Chicago, Ill., August. 8pp.

___. 1992. Test methods for evaluating solid waste, physical/chemical methods, SW-846, Third ed. Organic Section. Automated soxhlet extraction (Method 3541), gel-permeation clean-up (Method 3640A), and organochlorine pesticide, halowaxes, and PCBs as Aroclors by gas chromatography: capillary column technique (Method 8081). U.S. Environ. Prot. Agency, Washington, D.C. 108pp.

Varrett, M. J. 1976. Investigation of the toxic and teratogenic effects of halogenated dibenzo-*p*-dioxins and dibenzofurans in the developing chick embryo. Memorandum Report. U.S. Food and Drug Admin., Washington, D.C.

White, D. H. 1976. Nationwide residues of organochlorines in starlings, 1974. Pestic. Monit. J. 10:10-17.

___, J. R. Bean and J. R. Longcore. 1977. Nationwide residues of mercury, lead, cadmium, arsenic, and selenium in starlings, 1973. Pestic. Monit. J. 2:35-39.

___, and M. T. Finely. 1978. Uptake and retention of dietary cadmium in mallard ducks. Environ. Res. 17:53-59.

___, and T. E. Kaiser. 1976. Residues of organochlorines and heavy metals in ruddy ducks from the Delaware River, 1973. Pestic. Monitor. J. 9:155-156.

___, R. C. Stendel, and B. M. Mulhern. 1979. Relations of wintering canvasbacks to environmental pollutants - Chesapeake, Maryland. Wilson Bull. 91:279-287.

Woolf, A., C. Gremillion-Smith, and C.O. Frank. 1983. Regional variation in metals in livers of white-tailed deer in Illinois. Trans. Ill. State Academy Sci. 1-2:305.310.

___, J. R. Smith, and L. Small. 1982. Metals in livers of white-tailed deer in Illinois. Bull. Environ. Contam. Toxicol. 28:189-194.

APPENDICES

Appendix A. Starling ethogram utilized during observation periods to identify behaviors.

NEST ATTENTIVENESS

In the Nest (IN) - When bird is actually inside the nest. It is assumed that bird is being attentive by incubating or brooding and feeding at this time.

Coming to Nest(CN)- Bird returns to the nest from another location presumably to take care of the young (e.g., deliver food, incubate eggs).

Leaving Nest (LN) - When bird flies off of nest and lands in another area away from the nest. Simply flying up and landing back on the box does not count. Can assume that bird lands somewhere else if it flies out of your sight.

MATING BEHAVIOR (MB)

Singing - Considered mating behavior when male sits on nest box making patterned chirps and whistles which increase in vigor when another starling flies by; often accompanied by wing flailing (Kessel 1957).

Wing Flailing - Wings are half extended and rotated above the horizontal; looks like a slowed down flap used in flying; usually occurs at the end of a singing bout; seems to be used primarily for mate attraction stimulated by another bird flying by (Feare 1984).

Sexual Chase - Female leaves every time male approaches; he may follow her over a long distance across branches, a field, or flight; usually small hops or short flights (Feare 1984).

Shoulder Pecking - Female pecks the neck or shoulder region of the male. The male responds by immediate mounting and copulation follows. Kessel (1957) feels that this behavior incites the male to mate - it was never omitted in her 5 year study.

AGGRESSION (AG)

Stare - Often done with the head held higher than in resting position and the height may indicate intensity of aggression; can be accentuated by raising the crown feathers; easiest and initial form of aggression (Feare 1984).

Open-Bill Threat - The bill is opened slightly as the bird stares at the other with head raised; signals a higher degree of aggression than just the stare (Feare 1984).

Fly-up - Two birds fly up together calling at each other and threatening with stabs and kicks; the most extreme form of aggression (Feare 1984).

INTERSPECIES AGGRESSION (IAG)

Same behaviors as listed above, but directed at a different species.

SUBMISSION (SB)

Departure -	Bird leaves (walks away or flies away) after a stare, open bill threat or fly up (Feare 1984).
Submissive Crouch -	Bird lowers its body and sleeks back its head feathers (Feare 1984).
Bill-wiping -	Bird wipes both sides of the bill on a twig or other structure; usually occurs when feeding is finished and is thought to signal to other birds that it is no longer competing for food resources; this is employed as a sign of backing down during a threat and is also seen in birds who submit during a contest for nest site (Feare 1984).

MAINTENANCE (M)

Self-preen
Dust Bath
Bill Wiping (only if done while/after feeding)
Defecation

STATIONARY (ST)

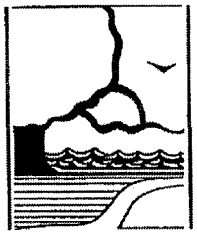
Bird is on the nest box, but is not exhibiting any other behavior; the head can be moving.

NOT VISIBLE (NV)

When the bird or birds for a particular nest box are not in sight.

OTHER (OT)

Any behavior observed that is not listed in this ethogram. Be sure to give a detailed description of the behavior in the comments section of the observation form.



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