

Environment Canada's National Wildlife Specimen Bank: A Valuable Resource for Monitoring and Research

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Abstract—Environment Canada manages two environmental specimen banks; the National Aquatic Biological Specimen Bank, which contains mostly fish tissues collected as part of Environment Canada's Fish Contaminants Monitoring Program, and the National Wildlife Specimen Bank, which holds primarily bird tissues from a number of monitoring programs and research studies across Canada. Both specimen banks originated in the 1960s and 1970s at a time of emerging concern over persistent toxic substances in the environment. They are the largest repositories of frozen environmental specimens in Canada and are among the longest running formal environmental specimen banking programs in the world. This paper will focus on the monitoring and research activities supported by the National Wildlife Specimen Bank. Over the years, archived tissues have been used to carry out retrospective analyses of new and emerging chemical contaminants as well as other compounds of interest. Specimens have also been used to address a number of research and policy-related questions.

Keywords: specimen banking, environmental monitoring, wildlife

INTRODUCTION

Concerns over persistent organic chlorinated pollutants in the environment led to the development of fish and wildlife monitoring programs in Canada in the 1960s and 1970s. Concerns about toxic chemicals were originally focussed on the Great Lakes. Fish have been collected annually at 12 locations throughout the Great Lakes since 1977 and archived in the Great Lakes Fisheries Specimen Bank, now known as the National Aquatic Biological Specimen Bank. Over the years, the Bank's holdings have grown to almost 19,000 specimens representing 53 fish species.

During the early 1970s, observations of poor reproductive success in colonial

waterbirds nesting near and feeding in the Great Lakes (Mineau *et al.*, 1984; Bishop *et al.*, 1992) led to the development of the Great Lakes Herring Gull Monitoring Program, which is now one of the longest-running biomonitoring programs in the world (Hebert *et al.*, 1999). Eggs of herring gulls (*Larus argentatus*) have been sampled on an annual basis at 13 colonies throughout the Great Lakes since 1974 and placed in frozen storage. Programs to monitor contaminants in seabird eggs were also established for Canada's Atlantic and Pacific coastal environments as well as for the Canadian Arctic (Noble and Elliott, 1986). The goal of these programs was to understand the behavior of chemical pollutants in natural systems, their effects on wildlife, and to provide a baseline from which to assess the effectiveness of emission reductions. The history of the relationship of these monitoring programs and specimen banking has been described by Wakeford and Kasserra (1997). Samples collected for these monitoring programs were, and continue to be, archived in what has evolved into the National Wildlife Specimen Bank (NWSB). Archived collections have also allowed us to investigate the role of contaminants in the population decline of some species. This paper will focus on the monitoring and research activities supported by the National Wildlife Specimen Bank, whose holdings have grown to include approximately 90,000 specimens representing about 820 species of birds, mammals, amphibians, reptiles, and other organisms collected from various locations across Canada.

METHODS

Collection

Wildlife specimens are normally collected for annual chemical or biochemical monitoring rather than being banked solely for some unspecified future use. Specimens are collected via three routes: long term monitoring projects, short term or regional studies, and opportunistic collections (Wakeford and Kasserra, 1997).

Storage

Initially, collected samples were stored in -20°C chest freezers. In 1981, a larger and more permanent storage facility was built at the National Wildlife Research Centre (NWRC) in Hull, Quebec, for the emerging Specimen Bank. It consisted of two -40°C freezer rooms for the permanent storage of samples, a smaller room operating at -25°C for current-year samples which required processing, and one -80°C chest freezer. In 2003, the Bank was moved to the new NWRC facility located at Carleton University in Ottawa. The Specimen Bank's current capacity includes three extra-large -40°C walk-in freezer rooms for long-term storage of samples, seven ultra-low temperature (-85°C) freezers which provide better stability for samples intended for analyses of fatty acids and some biochemical assays (e.g. vitamin A), and three liquid nitrogen freezers (N_2 gas phase temperature: -150°C) to ensure sample viability for most biochemical

assays. The freezers are located inside a room equipped with a full yellow light system for the handling of samples sensitive to normal light. Sample locations and processing status are tracked using a computerized Laboratory Information Management System (LIMS). All freezers are hooked to Emergency Back-up Power, have their temperatures logged daily, and are remotely monitored for temperature fluctuations which can activate an alarm system should temperatures deviate outside acceptable ranges.

Access to specimens

Tissue samples from the National Wildlife Specimen Bank are frequently shared for collaborative purposes. In 1998, an Access Policy was instituted which is administered by a Specimen Bank Committee. Priority access to samples is maintained for the first five years by the principal investigator responsible for making the collection. Specimens belong to the NWSB once the time elapsed from original collection has exceeded five years. Specimens are then released for research purposes under the following conditions:

- The research proposed has been reviewed for scientific merit through an internal review process led by the Specimen Bank Committee.
- There is sufficient tissue to issue without unduly depleting the total specimen. If a request will deplete the remaining tissue, then the rationale for issuing it will be more stringent.
- Other means of obtaining the information have been explored and are not possible.
- Publications resulting from the research acknowledge the National Wildlife Specimen Bank as the source of the tissues.

RESULTS AND DISCUSSION

Samples from the National Wildlife Specimen Bank have enabled a number of monitoring programs and research studies over the years. Changes in analytical methodology have also been monitored using archived samples to ensure the integrity and continuity of the quality of the monitoring data sets over time (Turle *et al.*, 1991). Temporal monitoring of contaminants in seabird eggs has occurred and continues on Canada's three coasts: Atlantic (Elliott *et al.*, 1989a), Pacific (Elliott *et al.*, 1989b), and Arctic (Braune, 2007), as well as in the Great Lakes (Hebert *et al.*, 1999). In the Canadian Arctic, eggs of thick-billed murre (*Uria lomvia*), northern fulmars (*Fulmarus glacialis*) and black-legged kittiwakes (*Rissa tridactyla*) from Prince Leopold Island have been monitored for contaminants since 1975 (Braune, 2007) to provide an index of contamination of the arctic marine ecosystem and possible implications for seabird health. Most of the legacy persistent organic pollutants or POPs (e.g. PCBs, DDT) have been declining (Braune, 2007) as shown in Table 1.

More recent uses of the Bank include the use of archived samples to: i) carry out retrospective analyses for emerging compounds of interest, ii) determine the exposure of species at risk to contaminants, and iii) to investigate food web changes. Examples of these uses are discussed below.

Table 1. Temporal trends of concentrations of total PCBs (Σ PCB), DDT metabolites (Σ DDT), total chlorobenzenes (Σ CBz), total chlordanes (Σ CHL), dieldrin and total mirex (Σ Mirex = mirex + photo-mirex) in eggs of thick-billed murres, northern fulmars and black-legged kittiwakes collected from Prince Leopold Island, 1975–2008. ↓ indicates a significant decrease in concentrations. Regression analyses indicate that Σ Mirex concentrations in thick-billed murres are decreasing although the decrease is not statistically significant ($p = 0.08$).

	Σ PCB	Σ DDT	Σ CBz	Σ CHL	Dieldrin	Σ Mirex
Murre	↓	↓	↓	↓	↓	↔
Fulmar	↓	↓	↓	↓	↓	↓
Kittiwake	↓	↓	↓	↓	↓	↓

Retrospective analyses

Archived arctic seabird samples have been screened for newer compounds such as the perfluorinated compounds (PFCs) (Martin *et al.*, 2004), polychlorinated naphthalenes (Muir, 2004) and brominated flame retardants such as the polybrominated diphenyl ethers (PBDEs) and hexabromocyclododecane (Braune and Muir, 2005). Once the presence of these chemical compounds was confirmed in wildlife tissues, retrospective analyses were carried out using archived samples to establish time trends. For example, time trends have been established for PFCs (Butt *et al.*, 2007) and PBDEs in arctic seabirds (Braune, 2008). The concentrations of total PBDEs in seabird eggs from Prince Leopold Island increased significantly between 1975 and 2003 (Fig. 1) which agrees well with the pattern of increase documented for North America during that time period (Hale *et al.*, 2003; Law *et al.*, 2003). The subsequent decline in concentrations reflects the phasing out of Penta-mix BDE usage in North America in recent years (de Wit *et al.*, 2006).

Exposure of species at risk to contaminants

Numerous bird species which breed in Canada are experiencing population declines. One such species is the ivory gull (*Pagophila eburnea*) which is an arctic marine bird which has been listed as “*Endangered*” in Canada (COSEWIC, 2006). In order to determine whether or not chemical contaminants were playing a role in the population decline of this species in Canada, archived egg samples, which had been collected from Seymour Island in the Canadian Arctic in 1976, 1987 and 2004, were retrieved from the Specimen Bank and analyzed for a variety of contaminants.

Concentrations of the legacy organochlorines as well as PCDDs and PCDFs either decreased or showed little change between 1976 and 2004, whereas concentrations of PBDEs steadily increased over this time period (Braune *et al.*, 2007). Measured concentrations of all of these persistent chlorinated compounds were below published toxicological threshold values for wild birds. However, mercury concentration in eggs of ivory gulls were determined to be among the highest ever recorded in arctic seabird eggs, with mercury in some eggs exceeding

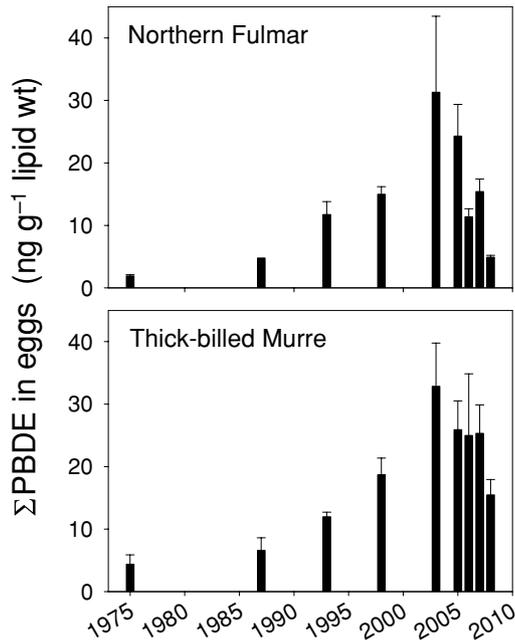


Fig. 1. Concentrations (ng g⁻¹ lipid wt) of total polybrominated diphenyl ethers (Σ PBDE) in eggs of northern fulmars and thick-billed murres from Prince Leopold Island in the Canadian Arctic, 1975–2008.

published toxicological threshold levels for impaired reproductive success in birds (Fig. 2). These findings merit concern and further investigation (Braune *et al.*, 2006).

Effects of food web changes on contaminant trends

The development of ecological tracers to track the flow of energy and nutrients through food webs has provided insights into factors affecting diet composition in wildlife. For example, ecological tracers have allowed for the quantitative evaluation of dietary changes in Great Lakes herring gulls (Hebert *et al.*, 2006). Stable nitrogen isotopes (¹⁵N/¹⁴N) measured in archived samples of herring gull eggs from the Great Lakes were used to estimate herring gull trophic position, and fatty acid data provided information on the degree to which aquatic foods, namely fish, were being consumed by the birds (Hebert *et al.*, 2006). Hebert and Weseloh (2006) showed how interpretation of contaminant temporal trends can be affected by changes in diet composition which may affect exposure to contaminants. Significant declines in trophic position were determined for 10 of 15 herring gull colonies in the Great Lakes due to a shift from fish to a terrestrial-based diet (Hebert *et al.*, 2008). At colonies where significant declines

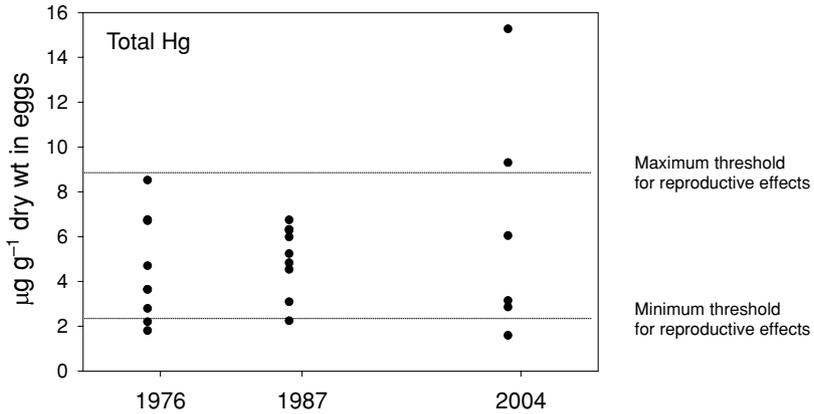


Fig. 2. Total mercury concentrations ($\mu\text{g g}^{-1}$ dry weight) in individual ivory gull eggs collected in 1976, 1987 and 2004 (modified from Braune *et al.*, 2006). Minimum and maximum threshold levels for reproductive effects based on data from Thompson (1996).

in trophic position were found, temporal trends in contaminants adjusted for trophic position, as indicated by $\delta^{15}\text{N}$, showed a less pronounced decline (Hebert and Weseloh, 2006). These results have major ramifications for all programs monitoring contaminants in wildlife.

SUMMARY

The National Wildlife Specimen Bank is integral to the wildlife monitoring and research activities of Environment Canada. Archived samples have found many uses over the years including screening for new chemical compounds which is often followed by retrospective analyses to establish time trends, investigating the exposure of species at risk to contaminants, and more recently, the use of archived samples to investigate food web changes. Many of our monitoring programs and research studies would not be possible without the use of archived samples.

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