Mercury and marine birds in Arctic Canada: pathways, effects, and interactions in a model species

by

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Abstract

Mercury (Hg) is an element naturally found both in the environment, and released through a number of industrial processes. Importantly, Hg is a toxin known to cause a wide variety of effects from changes in endocrine and neurological pathways, to impaired reproduction and decreased survival. Avian species are widely used as indicators of environmental Hg contamination as they are sensitive to its effects and multiple endpoints can be easily assessed.

My thesis aims to review and explore the pathways and effects of Hg on Arctic marine birds. First, I review the current levels, known effects and trends in eight marine bird species in northern Canada. Following this review I use a model avian species (the common eider, Somateria molissima) to better understand how Hg may affect marine birds independently and jointly with our other environmental stressors. In Chapter 2 I examine how sex, diet and individual size can influence both Hg and parasites as they share common host pathways. My results indicate that several different extrinsic factors can influence parasite and Hg distributions within a population, emphasizing the importance in assessing these factors when examining contaminant burdens in wildlife.

In Chapter 3 I present an experimental manipulation of wild female eider ducks used to examine the effects of Hg on reproduction and survival under natural and reduced parasite burdens. My findings indicate parasites, but not Hg burdens, influence breeding decisions in this species. In Chapter 4 I investigate how Hg and lead (Pb, another natural metal known to have neurotoxic effects on wildlife) are associated directly and indirectly with known breeding pre-cursors (immunoglobulin Y, corticosterone levels, arrival date and arrival condition) in female eider ducks. While female eider ducks were found to
have low levels of both Hg and Pb, I found that both varied significantly with the physiological and phenological indices examined indicating that these contaminants do influence breeding in marine birds even at levels often considered below toxic thresholds. My thesis results demonstrate that to fully understand how Hg affects wildlife, both direct and indirect effects must be considered throughout the annual cycle.
Preface

Co-author statement

I contributed and lead the research presented in this thesis in several ways. First, I proposed and developed the research questions in collaboration with H.G. Gilchrist and M.R. Forbes. Second, I was primarily responsible for carrying out, supervising and directing all the laboratory work that was done in conjunction with this research. This included field work, as well as several different laboratory components (i.e. necropsy of birds, tissues collection preparation, parasite enumeration, chemical analyses).

Throughout this project I supervised a number of students who contributed to the project by assisting with the examination of both birds and parasites. Further, I worked with Dr. J.D. McLaughlin to preserve and identify parasite specimens. All stable isotope analyses were completed by the G.G. Hatch Stable Isotope Laboratory at the University of Ottawa. All of the contaminant analyses were done at RPC Laboratories in Fredericton, New Brunswick (blood samples), or at the National Wildlife Research Centre in Ottawa, Ontario (breast muscle samples). Third, I managed and analyzed all the data presented within the thesis. I also wrote all the first drafts of the chapters/manuscripts and was responsible for incorporating co-author feedback into the final version. Lastly, I was responsible for all correspondence and administration required to submit each manuscript/chapter for peer-reviewed publication.

I have chosen to use the integrated thesis format, and therefore each data chapter is presented as an independent research manuscript that is either already published, or has been submitted to a peer-reviewed journal upon the submission of this thesis. As a result, there is some repetition in the introduction and discussion sections, however, I have
cross-referenced between chapters to reduce repetition in the method sections. While each chapter has several contributing authors I always played a major role in the design of the research, the analysis of the data, the preparation of each manuscript and in handling each article throughout the editing and publishing process. To this end, I must acknowledge the guidance and support from my co-authors that have been involved throughout my PhD work.

I have permission from each publisher to reproduce published manuscripts in my thesis. I have also received permission from each of my co-authors to use the collaborative work in my thesis. For a specific chapter that has been published elsewhere, all citations must be of the published journal article with the information provided below. However, to reference the entire thesis as a whole, I recommend using the following citation:


**Status of chapters at the time of thesis submission**


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sizes, and the students helped to dissect the birds, prepare tissues for contaminant analysis and examine birds for gastro-intestinal parasites. I could not have completed this work without the help of the students involved.

Throughout my work I was fortunate to have access to Environment and Climate Change Canada’s National Wildlife Research Center (NWRC). While the staff at the NWRC are incredibly supportive and helpful there are two groups that were integral to my work. First, the northern seabird team, including Amie Black, Birgit Braune, Michael Janssen, Sam Iverson, Christie Macdonald, Nik Clyde, and Frankie Jean-Gagnon were always a source of encouragement and support, both in the field and in the office. Second, the personnel working in the Specimen Bank (Caroline Roberts, Robyn Lima, Christine McClelland, Guy Savard, Michelle Zanuttig, Kyle Seabrook) were welcoming and patient as I learned to navigate the specimen bank methods and procedures over the years. I would also like to extend this thank you to Mark Mallory at Acadia University. Mark has been a constant supporter of my work over the years, both during his time with the Canadian Wildlife Service, and more recently at Acadia. I consider both Mark Mallory and Birgit Braune as unofficial committee members who have been great enablers of my research.

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1 Chapter: General Introduction

Reproduction and survivorship are thought to be under substantial selective pressure in animals breeding in seasonal environments (Garant et al., 2007; Lepage et al., 2000). For such species, individuals that are able to time their breeding to take advantage of resources and optimal rearing conditions can often improve their reproductive success in any given year, and thus their overall fitness (Lepage et al., 2000; Love et al., 2010). For example, female northern common eider ducks (*Somateria mollissima borealis*) arriving earlier in the breeding season laid larger clutch sizes, and females arriving in better condition laid earlier (Descamps et al., 2011a). Importantly, both large clutch sizes and earlier laying are associated with higher nest success (Descamps et al., 2011a). While there is a genetic component that codes for many traits associated with reproduction and survival, these are likely significantly influenced by factors external to the organism. Factors that affect body condition are therefore expected to explain variation in breeding propensity, onset and productivity. Understanding how external factors, such as contaminants\(^1\), influence individual reproduction is crucial to understanding how these factors might drive changes in populations (Reed et al., 2008; Wayland et al., 2002).

1.1 Mercury as an external factor influencing reproduction and survival

Although numerous contaminants are of toxicological concern in birds, Mercury (Hg) is of particular interest in northern ecosystems due to high levels detected in wildlife in polar environments, and its negative impacts on organisms (Dietz et al., 2013; Riget et al., 2011). Most of the Hg deposited in the terrestrial, aquatic and marine ecosystems from atmospheric sources is in the inorganic form, but through a number of pathways

\(^1\) See Appendix B for operational definitions for all text in bold. Bold appears the first time a term is used.
methylmercury (MeHg) is formed, which is toxic to biota (Liu et al., 2012). Metabolism of inorganic Hg to MeHg can take place within wetlands, aquatic sediments or in the water column by microbial methylation (Kidd et al., 2011). Aquatic organisms, such as plankton and fish, can absorb MeHg directly from surrounding waters, while terrestrial organisms and marine birds mainly accumulate MeHg through their diet (Liu et al., 2012). Although Hg is found naturally, anthropogenic inputs of Hg are increasing environmental levels on timescales that are much shorter than evolutionary timescales to levels known to cause toxic effects in organisms (Dietz et al., 2011).

Animals exposed to MeHg store it in their tissues at higher levels than found in the surrounding environment (Wiener et al., 2003). Methylmercury is also one of the few metals that is known to biomagnify through food webs resulting in animals at upper trophic positions having higher tissue concentrations than those species lower in food webs (Campbell et al., 2005). Although MeHg represents less than 1% of the total Hg (THg) found in marine and freshwater systems, essentially all Hg found in most marine bird tissues examined is the toxic MeHg form as MeHg accumulates in organisms and most marine birds occupy relatively high trophic levels (Bond and Diamond, 2009a).

Once in the body, MeHg acts as a neurotoxin (Liu et al., 2012). Methylmercury can act as a competitor to selenium based enzymes (Farina et al., 2011). As a result, Hg is an enzyme inhibitor and may irreversibly disrupt a number of metabolic processes (Hoffman et al., 2003). Mercury exposure also has been shown to cause oxidative cellular injury, in which it disrupts receptor signaling pathways associated with the endocrine system, and disrupts gene regulation of the body’s protective mechanisms (Liu et al., 2012; Tartu et al., 2014). Thus, Hg not only causes direct damage to cells, it can have an
indirect effect on metabolic pathways leaving cells open to damage from other chemicals as well. These physiological disruptions at the cellular level can manifest in a number of ways including sensory disturbances and loss of muscle coordination (Heinz, 1979, 1975; Liu et al., 2012).

Wild birds have been found to exhibit neurological and physical changes with increasing Hg levels. In wild adult birds, Scheuhammer et al. (2008) found neurological changes in brain receptors and enzymes in common loons (Gavia immer) when birds had high concentrations of Hg. Thus, Hg can potentially impact a range of behaviours including foraging and migration because Hg affects the central nervous system of an animal, and this can lead to an overall decline in body condition. In both clapper rails (Rallus longi; Ackerman et al., 2012) and eider ducks (Wayland et al., 2002) body mass has been found to decrease with increasing Hg levels. Such changes in physiology and body condition can then manifest in a bird as a reduced condition, which can directly or indirectly lead to reduced breeding success or output. For example, wild tree swallows (Tachycineta bicolor) exposed to high environmental Hg concentrations had smaller clutch sizes than those with minimal Hg contamination (Brasso and Cristol, 2008). Thus, Hg can affect wildlife indirectly through declines in health and condition, because of its negative impacts on biota, which can then impact breeding effort and success.

1.2 Common eider ducks as a model species

The northern common eider duck (hereafter referred to as eider duck) is a useful model species for investigations that examine factors affecting breeding ecology, including Hg. Eider ducks are well studied in the circumpolar Arctic, and are relatively easy to mark, track and monitor over time while breeding (Goudie et al., 2000).
Importantly, eider duck breeding ecology has been intensively studied in relation to reproductive trade-offs and optimal reproductive strategies, as they relate to individual and parental characteristics (Bustnes et al., 2010; Descamps et al., 2011a, 2009; Hanssen, 2006; Hanssen et al., 2003; Hennin et al., 2013; Legagneux et al., 2014).

Specifically, eider ducks are useful models to study how extrinsic factors may affect reproduction as it has been shown that body condition plays an important role in eider duck reproduction during several different phases of the breeding season. First, the condition of an individual is important in their ability to reach the breeding areas. Eider ducks wintering in Greenland have high levels of endogenous reserves throughout the non-breeding season (Jamieson et al., 2006). Subsequently, following the breakup of the sea ice in Hudson Strait, eider ducks move from Greenland, across the Atlantic and into the Arctic Archipelago (Mosbech et al., 2006). Following their spring migration, eiders carry little to no fat reserves; a time of energetic low (S. Jamieson unpubl data) indicating that during the post-migration period body condition is likely to greatly influence the ability of individuals to undertake a breeding attempt.

Second, body condition at the commencement of breeding is particularly important to incubating female eiders. While incubating their eggs, female eider ducks do not forage and typically only leave the nest occasionally to drink from local freshwater ponds. Female eider ducks must sustain themselves during this time on their fat reserves acquired post migration (Bustnes et al., 2010). Females with low reserves may not be able to incubate eggs to completion, or flush easily when approached by predators. Thus, any factors that influence body condition may affect reproductive commitment and success, both at the arrival stage and at the incubation stage when resources are low.
Third, eider ducks are also a useful model species for Hg concentrations because eiders have been assessed in several geographic regions, including the Canadian Arctic (Bustnes et al., 2010; Fenstad et al., 2014, 2016; Franson et al., 2004; Mallory et al., 2004; Wayland et al., 2001b). While Hg concentrations in eider ducks is often among the lowest levels reported in marine birds (Provencher et al., 2014b), negative effects associated with Hg levels in eiders have been reported. In adult eider ducks, high Hg levels are associated with reduced abdominal fat, and with increased intensity of parasitic nematodes (Wayland et al., 2001b); both of which affect body condition. Wayland et al. (2008) also found that wild king eider ducks (Somateria spectabilis; a related species to common eider ducks) with higher levels of Hg had a lower possibility of being recaptured the following year, which suggests a reduced survival rate in birds with higher concentrations of contaminants. This growing body of evidence suggests that even low levels of Hg can affect northern eider duck species.

Lastly, eider ducks are an ideal species to undertake this work because of the collaborative nature in which they can be sampled in Arctic Canada. It is difficult to address ecological questions regarding contaminants for many bird species because of ethical and practical limitations on acquiring sufficiently large sample sizes (Mallory et al., 2006c). Because eider ducks are commonly harvested by Inuit for food throughout Nunavut and Greenland, we can access large numbers of birds collected by hunters for sub-sampling. As a result of working directly with hunters in northern communities the sample sizes presented within this thesis were made possible. Thus, I investigated how Hg may contribute to survival and reproduction in a large sample of birds, adding to the power of the results presented here. Another advantage of my research is the use of a
free-living population of eider ducks. While this has limited the types of manipulations possible, the results presented are more applicable to discussions involving the potential for individual and population level effects in wildlife as compared to experimentally induced contaminant concentrations alone.

1.3 Cumulative effects of Hg and other extrinsic factors

Importantly, Hg (or any other environmental contaminants) does not act in isolation when influencing wildlife. While Hg may show inter-annual variation, other sources of stress (any factor that may limit an individual’s ability to gain resources, grow, survive and/or reproduce) are influencing wildlife at the same time. Food shortages, weather, parasites, inter- and intra-species competition and infectious disease can influence both condition and health (Daoust et al., 1998; Forrester et al., 1997). When wildlife are exposed to contaminants in association with other ecological variables, the relationship between pollutants and host often become more complex (Marcogliese and Pietrock, 2011). For example, both parasites and contaminants can influence how a host is affected by the other through altered immune functions (Owen et al., 2010; Sonne, 2010). In addition, contaminants, such as Hg, that are known to induce cellular oxidative stress may alter the immune response a host can mount in the face of parasite exposure. In turn, when a host is exposed to parasites, the host mounts an immune response which may be energetically costly (Bulté et al., 2012). Changes in exposure rates to parasites can affect the host immune response, and thus the impact that contaminants may have on the host (Bulté et al., 2012; Schulenburg et al., 2009). In turn the presence of parasites, which can interfere with the host’s immune response, can change how a host responds to contaminant exposure (Marcogliese and Pietrock, 2011). Such interactions are difficult
to predict, but important when investigating what may influence reproductive success of wild organisms.

1.4 Aims of this thesis

Arguably, it is the examination of cumulative effects that are the most critical in regions such as the Arctic where environmental change is happening at an unprecedented rate (IPCC, 2007). Changing climatic conditions inducing shifting levels of parasitism, predation and contaminants alongside increasing development and industrial shipping all make the Arctic ecosystems particularly sensitive to cumulative effects (ACIA, 2004; Iverson et al., 2014; Provencher et al., 2014b). Within this context this thesis sets out to examine how Hg burdens influence reproduction and survival a marine bird model species in connection with other environmental stressors present in northern Canada (Fig 1).

An overview of what is known of Hg concentrations in marine birds from Arctic Canada is presented first in Chapter 2. I discuss how Hg may affect northern marine bird species in general, and review the current levels and trends of Hg concentrations in eight marine bird species that have been sampled in the region. Within this chapter I also identify current research gaps in our understanding of how Hg affects Arctic marine birds in Canada, and why Hg in northern marine birds is an important research area that warrants continued attention even after decades of study. I specifically review why harvested species should be the focus of several research questions.

In chapter 3 I examine patterns of Hg concentrations in eider ducks (collected with hunters near Cape Dorset, Fig 2) to better tease apart the factors that contribute to how Hg may be distributed in this species, and how this relates to parasites which are
also trophically transmitted. The goal of this work is to examine how Hg may be influenced by diet and sexual dimorphism to inform future interpretations of Hg concentrations in eiders.

With the insights gained from the previous chapters, Chapter 4 goes on to examine how Hg may affect breeding female eider duck survival and reproduction, independently and jointly with burdens of gastro-intestinal helminthes. For this work I use an experimental approach to manipulate the parasite burdens in a group of breeding females at East Bay Island in northern Hudson Bay following work done by Hanssen et al. (2003) and Bustnes et al. (2006). Blood Hg concentrations and parasite treatment were examined with arrival condition, arrival timing, reproduction (propensity, clutch size, nest attendance and nest survival) and survival (year to year return rates).

In my 5th Chapter I further examine how Hg may directly or indirectly influence reproductive success in eider duck females via the immune and endocrine systems. Blood Hg and lead (Pb) concentrations in females from East Bay Island were examined in relation to known breeding pre-cursors, including two physiological metrics (immunoglobulin Y and corticosterone). This work aims to explore how both Hg and Pb may vary with phenological and physiological traits known to be strongly associated with eider reproductive success.

Finally, in the general discussion (Chapter 6) I summarize my key findings of each data chapter, and examine how the information presented in each section contributes to our understanding of how Hg may affect marine birds in Arctic ecosystems. I also discuss several factors that were considered throughout my research program but are not
presented in the main body of the thesis, and future research directions stemming from the thesis work presented here.
Figure 1. Map of Nunavut indicating sampling locations of common eider ducks (*Somateria mollissima*) from 2011 to 2014.
Chapter: Mercury and marine birds in Arctic Canada: effects, current trends and why we should be paying closer attention

2.1 Abstract

Mercury is a naturally occurring trace element that is also a by-product of anthropogenic activities and, in its methylated form it is a neurotoxin that can have adverse effects on wildlife. The toxicity of Hg for humans, wildlife and ecosystem health merits monitoring of its concentrations by various sampling means. Marine birds are widely used as indicators of ecosystem health, including biomonitoring of Hg in the Arctic. Since the mid-1970s, Hg concentrations in marine birds have been monitored across the Canadian North. Current Hg burdens in most northern marine bird species are below levels associated with health concerns, but several species have concentrations that are at or near levels associated with impaired reproduction. Arctic marine birds in Canada may be particularly at risk from increasing Hg levels associated with changing climatic conditions and long term Hg deposition patterns. Research on marine birds should therefore continue to focus on spatial and temporal patterns of Hg contamination, assessing levels and biological effects in species that are experiencing high concentrations, and among species that are widely harvested due to the possible implications for human health and population management.

2.2 Introduction

Mercury has been identified by the United Nations Environment Programme (UNEP) as a major concern to human and ecosystem health (UNEP, 2013). Over the last 150 years, natural sources of Hg have changed little while anthropogenic sources have increased dramatically due to industrialization (Dietz et al., 2009). As the release of Hg
into the environment has increased, mostly as elemental Hg from the burning of fossil fuels, accumulation of Hg has outpaced natural removal processes causing higher levels of Hg in the environment (Stow et al., 2011).

Elemental Hg may be converted to methylmercury (MeHg) in both freshwater and marine habitats (Lehnerr et al., 2011; Zdanowicz et al., 2013). Methylmercury is taken up by organisms through the food chain and biomagnifies, resulting in markedly high Hg concentrations in top predators (Campbell et al., 2005). Methylmercury is a concern as it acts as a neurotoxin and can have deleterious effects on organisms (Dietz et al., 2013; Sonne, 2010). A major concern with MeHg is that it can cross the mammalian placenta and is also incorporated into bird eggs during their formation, posing developmental risks to vertebrate young during critical periods of development (Stow et al., 2011). Thus, Hg can affect organisms directly exposed to Hg pollution, and through biomagnification, can affect organisms that are higher up the food chain as well as their future offspring (Heinz, 1979; Stow et al., 2011). Given the potential toxicity of Hg, monitoring of this trace metal in the environment is critical and of international importance (AMAP, 2011).

Since the 1970s, marine birds have served as useful bioindicators for monitoring Hg concentrations and biological effects (Dietz et al., 2011; Mallory and Braune, 2012). The ecology of marine birds is well studied, which has allowed documentation of patterns of Hg bioaccumulation across food webs and trophic levels (Campbell et al., 2005), and at different spatial and temporal scales (Braune, 2007; Point et al., 2011). Such studies are possible because marine birds are abundant, widespread, and are part of planned sustainable harvests allowing for relatively easy and ethical sampling to occur (Braune et al., 2005). Importantly, studies of marine birds offer a number of useful endpoints,
including reproductive output in relation to Hg burden at the individual level, to changes in demography over periods of decades (Mallory and Braune, 2012). Although marine birds are likely exposed to many different forms of Hg (i.e. elemental Hg, MeHg etc.) studies indicate that at least 95% of the total Hg in many seabird tissues is MeHg, allowing total Hg to be a useful indicator of the more toxic organic form (Wiener et al., 2003).

Both field and laboratory investigations of marine and non-marine birds have played a critical role in understanding how Hg affects animals. In wild bird species, elevated tissue concentrations of Hg have been associated with both biochemical and physical effects that lead to lower overall fitness (Scheuhammer et al., 2007). In adult common eiders, for example, increased Hg levels were associated with reduced abdominal fat, and with increased levels of parasitic intestinal worms (Wayland et al., 2001b). In wild tree swallows (Tachycineta bicolor), a reduction in reproductive output was found when adults were exposed to Hg contamination (Brasso and Cristol, 2008). In common loons (Gavia immer), individuals with high concentrations of Hg may be subject to neurological changes in brain receptors and enzymes (Scheuhammer et al., 2008), and may fledge fewer young, attend nests less and spend less time foraging (Evers et al., 2004).

Similarly, exposure to Hg may negatively influence avian growth and development in laboratory experiments. Eggs of thick-billed murreas (Uria lomvia) and Arctic terns (Sterna paradisaea) injected with increasing levels of MeHg had reduced egg hatchability and survival at higher dose levels (Braune et al., 2012). Across a number of bird species, embryos exposed to MeHg in the lab often develop deformities (Braune
et al., 2012; Heinz et al., 2011). Earlier work has shown that young birds of mothers exposed to MeHg also showed erratic behaviour and a lower response to maternal calls (Heinz, 1979). Overall, marine birds appear susceptible to Hg as both adults and developing young, which can affect birds at the individual level, and possibly, at the population level.

As a result of captive and field studies, lowest observed adverse effect levels of Hg have been developed for avian eggs and other tissues along with other toxicity thresholds (Shore et al., 2011; Thompson, 1996). A Hg concentration from 0.5 to 2.0 µg/g wet weight (ww) in eggs appears to interfere with hatchability or proper embryo development (Dietz et al., 2013). Similarly, from a review of the literature, Shore et al. (2011) suggest that values greater than 20 µg/g ww in hepatic tissue are hazardous in 5% of the population for non-marine bird species. In the absence of more species-specific values, Hg concentrations in marine birds have been compared against these toxicological thresholds, as done in a recent review by Dietz et al. (2013). This review examined the available Hg data for marine biota and Inuit populations against known threshold levels. With the work by Dietz et al. (2013) in mind, the aim of this review is to synthesize known Hg concentrations in marine Arctic bird tissues within the Canadian Arctic, identify knowledge gaps and provide suggestions for future research that will help bridge gaps in our knowledge of marine birds and Hg. In addition, I review mechanisms and ecosystem changes that have been proposed to influence Hg in the future, and thus potentially affect marine bird exposure to Hg in the Arctic region.
2.3 Current trends in mercury in Canadian Arctic marine birds

In the Canadian Arctic, nine marine bird species have been studied for Hg concentrations at varying spatial and temporal scales: Arctic tern, glaucous gull (*Larus hyperboreus*), ivory gull (*Pagophila eburnea*), black guillemot (*Cepphus grylle*), black-legged kittiwake (*Rissa tridactyla*), common eider, long-tailed duck (*Clangula hyemalis*), northern fulmar (*Fulmarus glacialis*), and thick-billed murre (*Braune et al.*, 2005; Mallory and Braune, 2012; Mallory et al., 2004). These species represent overlapping but different geographical areas, and occupy a variety of niches within Arctic food webs making them ideal for examining questions about Hg within the Canadian Arctic marine ecosystem (Mallory and Braune, 2012).

Long term monitoring of Hg in marine birds in Arctic Canada is primarily through collection of eggs from breeding colonies. Egg collection provides an ideal sampling technique that is both sensitive to change and limits impact on breeding birds (Dietz et al., 2013; Goodale et al., 2008). Based on total Hg in eggs, ivory gulls have the highest mean Hg concentration and maximum detected value of the species examined in the Canadian Arctic (mean = 6.37 ± 5.17 µg/g dry weight (dw); range = 1.59 -15.3 µg/g dw), and common eiders have the lowest mean egg Hg concentration reported (mean = 0.73 ±0.25 µg/g dw; Table 1). Of note is that the three related gull/tern species have the three highest mean egg Hg concentrations, indicating that this group may either accumulate more dietary Hg, be less capable of physiologically shedding Hg as has been suggested for other marine birds, or overwinter in areas different than the other species (Kim et al., 1996a). Similarly, the two duck species (common eider and long-tailed duck) have among the lowest reported mean concentrations, suggesting that duck eggs may be
either less exposed to Hg through diet, or be less susceptible to Hg accumulation than compared to other marine bird species from the same area.

Mercury concentrations in eggs are related to the trophic level of the species (Akearok et al., 2010; Fig 2). Eggs of marine birds show a wide range of Hg concentrations between species, with two orders of magnitude between the highest and lowest levels reported (15.3 µg/g dw in an ivory gull eggs, and 0.73 µg/g dw in common eider eggs). Diet and depuration are most likely the primary sources of the differences in egg concentrations of Hg between species. However, there are some species, such as the black-legged kittiwake, which have a lower egg Hg concentration than would be predicted by trophic level. The reasons for low Hg concentrations in kittiwake eggs may be due to differences in maternal intake (dietary items low in Hg), depuration rates, or metabolism (increased demethylation capabilities leading to excretion); all three are possible mechanisms. Alternatively, low levels in black-legged kittiwakes may be the result of them overwintering farther south along the coast of North America where Hg emissions have been reduced over the past few decades (Munthe et al., 2011).

A recent review of the ecological effects of Hg in Arctic wildlife identified ivory gull, glaucous gull and black guillemot eggs from the Canadian Arctic as falling within the range of published lowest observed adverse effect levels (Dietz et al., 2013). More recent data suggest that Arctic terns should also be added to the list of marine birds in the Canadian Arctic that have egg Hg concentrations within that range, suggesting they may also be at risk for adverse effects (Provencher et al., 2014a). Although avian embryos show a range of sensitivities to Hg exposure (Heinz et al., 2009), Braune et al. (2012) showed that average levels found in both thick-billed murre and Arctic tern eggs from
wild populations are within the same order of magnitude as the median lethal concentration (LC50) for the species. Also of note is that three of the species with the highest Hg concentrations (ivory gull, glaucous gull and Arctic tern) show declining population trends (Gilchrist and Mallory, 2005; Gilchrist and Robertson, 1999; Mallory and Braune, 2012). It is important that the scales at which declines are observed are also the scale from which egg data were collected to make inferences about possible linkages between Hg contamination and population declines (Bond et al., 2015). Notwithstanding the high levels in some species, these data suggest that Hg concentrations in eggs are not likely a threat to most species and populations, but Hg concentrations in the eggs of gulls and terns (family Laridae) are within the range of toxicological effects and merit monitoring in these declining species’ populations. Particular attention might be paid to egg laying order and Hg concentrations as laying order is known to affect many other biological attributes in asynchronously hatching eggs such as is seen with larids (Akearok et al., 2010).

Temporal trends of egg Hg concentrations have been determined for four of the nine species examined (Table 1). For two of these species, the ivory gull and northern fulmar in the high Arctic, increases in egg Hg concentrations have been detected (Braune et al., 2007; Riget et al., 2011). During the same time period, black-legged kittiwakes showed no change in Hg levels, while one colony of thick-billed murres shows no significant trend in Hg concentrations in the low Arctic, and another murre colony in the high Arctic shows an increase in Hg (Riget et al., 2011). Differing trends in thick-billed murres from different colonies over the same period of time indicate that Hg deposition and uptake in the Canadian Arctic is not uniform, with Hg accumulation in wildlife
occurring at different rates in the high Arctic versus the low Arctic (Braune unpub data). Here, more replication is needed at a variety of different sites to strengthen inferences about Hg accumulation in relation to geographical location.

Total Hg in liver tissue of the same nine species has been examined, although the data are not as current as the egg data (Table 2). Glaucous gulls have the highest reported mean hepatic Hg concentrations (mean = 9.73 µg/g dw), with long-tailed ducks having the lowest mean concentrations reported (mean = 0.9 µg/g dw). Again, glaucous gulls and Arctic terns have higher values than most species examined, but common eiders also have relatively high hepatic Hg concentrations (Table 2). Such high levels in common eiders may be due to local exposure or timing of sampling (Mallory and Braune, 2012; Mallory et al., 2004). Black-legged kittiwakes, black guillemots and long-tailed ducks have relatively low concentrations of Hg in the liver, but the most recent reported hepatic Hg values for the Canadian Arctic are more than a decade old, suggesting that some species should be re-examined to assess current levels.

Mercury concentrations are higher in liver than in eggs for all species examined, which follows the general pattern for birds (Wiener et al., 2003). Hepatic Hg concentrations associated with death (>20 µg/g ww HC5 = ~66 µg/g dw assuming 70% moisture; Shore et al., 2011) are much higher than those observed in any species discussed here, indicating that Hg exposure is not a direct threat to the health of adult Arctic marine birds. Of note, glaucous gulls have hepatic Hg levels in the range of concentrations known to cause impaired reproduction (>2 µg/g ww = ~6.6 µg/g dw). As outlined above, with broad declines in glaucous gulls populations across the Arctic
(Gaston et al., 2009b), such high concentrations of Hg in glaucous gulls are cause for concern.

Only a single reported mean value for hepatic Hg exists for ivory gulls based on a sample size of two individuals (Campbell et al., 2005), therefore the reported value may not be representative. Consistently high Hg concentrations in ivory gull eggs (Braune et al., 2006), much higher than in other species, suggest that this species may experience even higher hepatic Hg concentrations than values reported in the literature for this species.

2.4 Why should we be paying closer attention

Mercury is widely recognized as damaging to biological organisms, and biomonitoring of Hg in the environment is key to understanding its sources, pathways and biological effects (Dietz et al., 2013). The following are eight reasons why we should be paying closer attention to Hg in Canada’s Arctic marine bird community.

2.4.1 Mercury is on the rise in some parts of the Canadian Arctic, despite regional reductions and an international convention.

Reductions in North American Hg emissions have occurred over the last 20 years, but the amount of Hg in the atmosphere over North America is influenced by global patterns. Elemental Hg released in Asia typically spends six to 24 months in the atmosphere during which time air currents can carry it across the Pacific to the North American Arctic (Jaffe et al., 2005; Munthe et al., 2011; Schuster et al., 2002). While Hg concentrations in wildlife are declining in northern Europe where air masses primarily originate from North America, some top predators in the North American Arctic and Greenland continue to show increasing Hg concentrations (Riget et al., 2011; St Louis et
The rate of industrial development in Asia is not likely to decrease in the coming years, and thus continued Hg deposition in the Canadian Arctic via atmospheric long-range transport is expected (Kirk et al., 2012; Pacyna et al., 2010; Riget et al., 2011).

In practice, the Minamata Convention, a legally binding agreement between 128 countries to manage the mining, use and disposal of Hg, was negotiated to limit the release of Hg into the atmosphere by 2020 (UNEP, 2013). Although the convention has been ratified now by 28 countries, and is much needed, the current concentrations of Hg in the environment will persist for decades to come, and in some areas are predicted to increase. Indeed, the global oceans have not reached equilibrium with the current atmospheric levels (Sunderland and Mason, 2007). As a result, oceanic Hg levels are likely to continue to increase for centuries whereas atmospheric levels may continue to decrease in the coming decades until equilibrium is attained (Cole and Steffen, 2010; Munthe et al., 2011). Additionally, the polar regions are susceptible to continued Hg accumulation through atmospheric mercury depletion events (ADMEs); polar spring events that enhance the deposition of elemental and particulate Hg (Kirk et al., 2012). These relationships ensure that Arctic marine ecosystems will continue to be exposed to elevated levels of Hg for years to come, despite expected reductions in atmospheric inputs as expected with the ratification of the Minamata Convention.

### 2.4.2 Changing climatic conditions are altering natural Hg sinks in the Arctic.

Anthropogenic activities are increasing Hg emissions, but they are not the only source of environmental Hg levels. Several natural sources are becoming more important as climatic conditions change. Glaciers and ice caps are known to act as transient
reservoirs for Hg (Stern et al., 2012). Under historic climatic conditions, glaciers and ice caps accumulated Hg for decades or millennia, but under the current warming climatic trend these terrestrial ice bodies are releasing sequestered Hg directly into aquatic environments as they melt, with the potential to affect both marine and aquatic ecosystems (Stern et al., 2012).

Melting permafrost is another potential source of increasing concentrations of Hg for biota (Stern et al., 2012). Continuous permafrost is the layer of soil that stays frozen for more than two consecutive years, and underlies most of the terrestrial areas in the Arctic region (IPCC, 2007). In the circumpolar Arctic region, it is estimated that a 13% to 28% reduction in permafrost area will occur by 2050 (IPCC, 2007). The amount of Hg being released by this process is unknown, but it may be more significant than changes in atmospheric deposition (Stern et al., 2012). Although the focus of this paper is on the Canadian Arctic, melting permafrost will have implications for Arctic regions of other countries, as well.

2.4.3 Interactions between Hg and other ecological stressors are not well understood.

Mercury and other environmental contaminants do not act on an organism in isolation. For example, although often considered separately, the effects that contaminants and parasites have on an organism may be connected (Marcogliese and Pietrock, 2011). Both parasites and contaminants can influence how a host is affected by the other through altered immune functions. In addition to changes in immunology, parasites may alter host contaminant uptake and retention. Parasites can alter the energy budget of a host causing an increase or decrease in metabolic and/or foraging rates.
(Bergey et al., 2002). In turn, this may increase or decrease a host’s exposure to trophically transmitted contaminants, such as Hg. Once contaminants are in the host, parasites, such as helminths, may take up contaminants thereby lowering exposure of hosts to contaminants (Bergey et al., 2002; Robinson et al., 2010); all contributing to complex interactions between parasites and contaminants such as Hg.

Water pH has also been shown to influence how organisms accumulate Hg (Jardine et al., 2013). Some fish species are sensitive to changes in pH and accumulate more Hg in their tissues in environments where pH is lower (Burgess and Meyer, 2008; Jardine et al., 2013). For fish-eating birds this could lead to an increased intake of Hg in more acidic environments. Loons nesting in lakes with lower pH values have been found to have increased blood Hg concentrations (Meyer et al., 1995). With the polar seas particularly sensitive to ocean acidification (McNeil and Matear, 2008; Steinacher et al., 2009), increased Hg accumulation may occur in fish consumed by marine birds, thus potentially exposing Arctic marine birds to even higher levels than currently being experienced. Again, this problem is not specific to the Canadian Arctic.

2.4.4 Changes in sea ice cover and northern food webs may alter exposure to Hg.

As local conditions change and food webs shift, so do the mechanisms through which Hg accumulates and magnifies up the trophic levels (Stern et al., 2012). Mercury accumulation in predatory species can be altered through changes in their prey if some prey are Hg accumulators, or through shifts in trophic levels. *Daphnia* spp., a small zooplankton found in freshwater lakes, has been identified as accumulating higher Hg compared to other zooplankton occupying similar trophic positions (Pickhardt et al., 2002). A range expansion of such species or an increase in similar ‘super-accumulator’
species in the north due to changing climatic conditions may result in Arctic birds and fish-eating species that carry concentrations of Hg higher than historical levels in similar biota. This increase in a Hg-rich prey species may lead to a higher consumption of Hg without any apparent change in trophic position (Stern et al., 2012).

Alternatively, top predators, such as marine birds, may shift trophic position due to changing prey populations as a result of changing climatic conditions. Thick-billed murres in Hudson Bay have shifted from a diet dominated by the ice-associated Arctic cod (*Boreogadus saida*) to capelin (*Mallotus villosus*) as the extent of summer sea ice around breeding colonies has declined (Provencher et al., 2012). Importantly, Arctic cod have lower Hg concentrations than capelin (Braune et al., 2014a). As a result of such diet shifts, the rate of Hg accumulation in top predators may change, and thus affect how long-term data should be interpreted (Braune et al., 2015).

Changes in sea ice in the Arctic Ocean can also directly influence MeHg degradation. A recent study examining Hg species concentrations in Arctic seabird eggs indicates that sea ice cover can impede the breakdown of MeHg (Point et al., 2011). Thus, with sea ice cover decreasing over time, photochemical degradation of MeHg is likely to increase, thus changing its propensity to accumulate and magnify in marine food webs (Point et al., 2011). Such processes will be important to understand as the Arctic Ocean shifts from an ice-dominated habitat to an open water region during the summer season.
2.4.5 The less productive ecosystems of the Arctic are vulnerable to exacerbated bioaccumulation.

When organisms grow rapidly, the Hg concentration within their tissues decreases; this is known as growth dilution, but the opposite can also be true (Kidd et al., 2011). In less productive ecosystems where primary production is low, such as the Arctic, slow growth rates can lead to higher levels of Hg in tissues as compared to more productive areas (Kidd et al., 2011). As a result of less productive ecosystems exacerbating Hg accumulation in top predators, Arctic marine birds feeding at the top of the food web may be particularly susceptible to acquiring large Hg concentrations as compared to their lower latitude counterparts (Fig. 3). If Hg continues to increase in the Arctic it will be important to monitor Hg levels across food webs to be able to predict and interpret changes.

2.4.6 Large data gaps still exist for many marine bird species in the Canadian North.

There continues to be limited data for a number of marine bird species in the Canadian Arctic (Table 3). Although some species have been assessed over several time periods and/or geographic regions, little to no studies of Hg burdens exist for many species (Table 3). Importantly, there has been little investigation into Hg burdens in predators of marine birds, such as peregrine falcons (*Falco peregrinus*) and snowy owls (*Bubo scandiacus*). Northern birds of prey warrant special attention as some, such as the snowy owl, are known to prey on marine birds at sea during the winter (Robertson and Gilchrist, 2003; Therrien et al., 2012). In a study examining trends in birds of prey from Greenland, the bird species with the most marine diet had the highest Hg tissue
concentrations (Dietz et al., 2006). Similar marine specialists in the Canadian Arctic may also have high levels of Hg.

Spatial comparisons of Hg concentrations in marine birds in the Canadian Arctic have shown a latitudinal gradient in some species, while others show no regional patterns (Braune et al., 2014b, 2005). The causes of such regional patterns may be linked to Hg deposition, as well as dietary differences within species, but it is not clear whether latitudinal gradients exist for all species, or just certain groups due to foraging behaviours and migration patterns.

Additionally, sources of inter-annual variation in Hg concentrations within species are still not understood. Shorebirds on Southampton Island in northern Hudson Bay have an order of magnitude difference in egg Hg concentrations within only a few years of comparative sampling (McCloskey et al., 2013). Similarly, large inter-annual variation has been found in seabird eggs in Lancaster Sound (Braune, 2007). These differences may be due to annual differences in the Hg cycle caused by the magnitude of ADMEs that occur each year at during the polar spring, a time when many migratory birds are returning to Arctic regions (Kirk et al., 2012). Differences in the timing of spring melt, which releases accumulated Hg from the snow pack (Zdanowicz et al., 2013) into the marine environment, may also affect the rate, timing and amount of Hg taken up by regional food webs.

2.4.7 **Marine birds in Canada have some of the highest reported levels for Hg in the pan-Arctic region.**

Although studies differ by sampling season, and occur over the course of decades, some of the highest Hg concentrations reported for bird populations from the pan-Arctic
region are from Arctic Canada (Table 4 and 5). When marine bird Hg tissue concentrations from the Canadian Arctic are compared to other studies that also sampled birds foraging in the low and high Arctic oceanographic zone (Salomonsen, 1965), the maximum reported values for six of the eight species in both liver and muscle tissue occurred in birds from Canadian waters. Consequently, marine birds from the Canadian Arctic may act as a bellwether for wider ecosystem trends in the north.

2.4.8 In the Arctic, marine birds are an important local food, tied to both human health and food security.

In addition to being useful indicator species, many marine bird species in the North are harvested, and are mainstay dietary items in northern communities (Mallory and Braune, 2012). A number of Arctic communities consume large quantities of locally harvested birds, potentially resulting in high exposure to dietary Hg. In short, the health of ecosystems influences the health of human communities. The more we understand about Hg pollution within ecosystems the better informed human health recommendations and related policy can be.

2.5 Where future research efforts should be focused

Mercury emissions have been reduced in many regions, but environmental levels continue to rise in some areas of the Arctic (Kirk et al., 2012; Stow et al., 2011). With changing climatic conditions and shifting food webs altering how Hg is accumulated and magnified, northern ecosystems are at risk from increasing Hg exposure. Marine birds provide a unique opportunity to track changes in Hg over time in the Canadian Arctic, investigate how Hg concentrations affect wildlife, and link ecosystem contamination to local communities. Taking into account the prior studies, data gaps as discussed above,
and with a future outlook, I have identified three areas of study that should be a focus in the Canadian Arctic.

First, studies should continue to focus on harvested species to track patterns and assess Hg levels in biota that are consumed by humans. Harvested species are useful indicators for biomonitoring, and Hg concentrations in these species have direct implications for human health (Kirk et al., 2012). In addition, population level effects of Hg could influence population size of harvested species, which in turn could influence their availability and rates of harvest. Research efforts should focus on tracking trends in Hg in particular areas and species where long-term data exist that provide the potential for temporal comparisons.

Second, species with particularly high levels of Hg, such as glaucous gulls, ivory gulls and Arctic terns, should be the focus of studies for biological or behavioural effects of Hg exposure. These three species have some of the highest egg and hepatic Hg concentrations in the Arctic, and show overall population declines in Canada (e.g. Allard et al., 2010; Braune et al., 2006; Gilchrist and Mallory, 2005). Although these declines are most likely multi-factorial, it is alarming that these declining populations also have the highest Hg levels. To better understand how Hg is affecting marine bird populations and biodiversity in the Arctic, future studies should focus on these species to yield results concerning Hg impacts on birds. Additionally, as discussed above, very little is known about many predatory species that feed on marine birds. These species may have very high levels of Hg concentrations, and may be among those bird species with the highest Hg concentrations in northern Canada.
Third, since marine birds in Canada exhibit large spatial and inter-annual variation in Hg levels, future sampling efforts should focus on understanding the cause of such variation. The causal mechanisms are not likely limited to marine birds, and thus understanding these patterns will help develop better insights to Hg cycling in northern ecosystems. Specifically for birds, there is also a need to develop a better understanding of which sub-populations or groups of birds may be at highest risk of Hg exposure, both from the perspective of human consumption and conservation.

In addition to the above described Canadian specific research areas, there are several topics of more broad scope where future research should be focused. Since Hg is a neurotoxin, research investigating Hg concentrations in brain tissue is also needed. Values in eggs and other tissues, such as liver and muscle, are common because of ease of collection and potential human health implications, but assessing Hg burdens in brain tissue, where concentrations may be more directly linked with potential impacts, is needed. Few studies of Arctic marine birds examined Hg concentrations in brain tissues. Provencher et al. (2014a) found that brain Hg concentrations in Arctic terns were considered high, while hepatic concentrations were below threshold values. This suggests Hg concentrations in the brain may be more indicative of long term exposure to Hg, as seasonal exposure may differ in migratory birds, and thus in tissues that have a higher turnover rate (Provencher et al., 2014a).

Of wide importance to marine bird and Hg studies is that the interpretation of how Hg burdens impact marine birds will remain uncertain as long as threshold values are based upon unrelated, non-marine species. Some studies have focused on understanding
how Hg affects seabirds in particular (i.e. Braune et al., 2012), but more needs to be done to develop our understanding of how marine birds exposed to Hg are affected. Overall, collections of marine birds in the Arctic as part of new and ongoing research projects should be co-ordinated in an effort to assess Hg levels in this group. Such efforts would greatly increase sample sizes, geographic coverage of the Hg information available, as well as disperse geographic coverage within a given year to help understand spatial patterns. Collective efforts that increase the resolution and detail of how Hg is affecting marine bird populations should be encouraged and supported. Continued monitoring of Hg in marine birds is also warranted as the accumulation of Hg in ecosystems may be long-lasting due to continued melting of the sea ice as well as other environmental changes, especially if trophic shifts occur as expected (Stern et al., 2012).

2.6 Conclusions

Even though the regulation of Hg released into the environment is progressing (UNEP, 2013), environmental levels in some areas continue to increase. As a result, Hg levels in the Canadian Arctic are predicted to increase for decades to come (Kirk et al., 2012), and have been identified as particularly sensitive to such changes (Douglas et al., 2012). Although Hg concentrations in most Arctic Canada marine birds are currently below levels of concern, some species show Hg concentrations that may have the potential of impairing biological processes (Thompson, 1996), and those species are thus at growing risk. In particular, Hg levels are notably high in three species (ivory gull, glaucous gull and Arctic tern) which are currently experiencing population declines. I conclude that the possibility of causal, synergistic effects merits investigation.
Canada has a long history of monitoring Hg in the North, and providing leading information on these issues (e.g. Douglas et al., 2012; Kirk et al., 2012; Mallory and Braune, 2012). Marine birds constitute an important part of northern ecosystems, and are an important food resource to dozens of communities across the north. This interest in bird health, as it relates to both human, and bird population health, makes Canadian marine birds of particular value in tracking patterns in Hg, as well as understanding how it might be affecting ecosystems worldwide.
Figure 2. Mean (± SD) total mercury levels in eggs with mean δ 15N values (± SD) for Arctic marine bird eggs. Data from Braune et al. (2006) and Akearok et al. (2010). Values are only presented for samples from the high Arctic (Prince Leopold Island, Seymour Island and Tern Island) where birds are likely to be exposed to similar environmental pollution levels within the region (colonies within 300 km of each other). δ 15N values indicate trophic position, with greater δ 15N values representing birds that eat at a higher trophic position.
Figure 3. Schematic of how a system with low productivity with slow growing biota may lead to exacerbated mercury burdens in top predators as compared with more productive, faster growing systems.
Table 1. Recent total mercury (THg) concentrations in eggs, and population trend data where known for eight marine bird species studied in Arctic Canada. Concentration mean, standard deviation and range in eggs are reported. Wet weight values are also included. Range of reported values is given where available, with mean values reported with standard deviations given when range was not provided. Population trend data from Mallory & Braune (2012), mercury trend data from Riget et al (2011). Arrows indicate an increasing or decreasing trend, ↔ denotes no change, and ? indicates trend data lacking.

<table>
<thead>
<tr>
<th>Species</th>
<th>Hg trend in Canada</th>
<th>Population trend in Canada</th>
<th>Year</th>
<th>Reported egg THg levels (dry weight, mean ± sd; value range)</th>
<th>Reported egg THg levels (wet weight; value range)</th>
<th>Source of data</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ivory gull</td>
<td>↑</td>
<td>↓</td>
<td>2004</td>
<td>6.37 ± 5.17; 1.59 -15.3</td>
<td>0.23 - 3.85</td>
<td>(Braune et al., 2006)</td>
</tr>
<tr>
<td>Glaucous gull</td>
<td>?</td>
<td>↓</td>
<td>2008</td>
<td>1.80 ± 1.03; 0.53 - 3.94</td>
<td>0.12 - 0.95</td>
<td>(Braune et al., 2016)</td>
</tr>
<tr>
<td>Arctic Tern</td>
<td>?</td>
<td>↓</td>
<td>2008</td>
<td>2.11 ± 0.53; 1.125 - 3.08</td>
<td>0.32 - 0.70</td>
<td>(Akearok et al., 2010)</td>
</tr>
<tr>
<td>Black-legged kittiwake</td>
<td>↔</td>
<td>↑</td>
<td>2008</td>
<td>0.80 ± 0.14; 0.62 - 1.16</td>
<td>0.15 - 0.26</td>
<td>(Braune et al., 2016)</td>
</tr>
<tr>
<td>Black guillemot</td>
<td>?</td>
<td>?</td>
<td>2008</td>
<td>1.79 ± 0.50; 1.10 - 2.36</td>
<td>0.30 - 0.56</td>
<td>(Braune et al., 2016)</td>
</tr>
<tr>
<td>Thick-billed murre</td>
<td>↑, ↔</td>
<td>↔</td>
<td>2008</td>
<td>1.14 ± 0.39; 0.56 - 2.15</td>
<td>0.14 - 0.57</td>
<td>(Braune et al., 2016)</td>
</tr>
<tr>
<td>Northern fulmar</td>
<td>↑</td>
<td>↔</td>
<td>2008</td>
<td>1.15 ± 0.29; 0.61 - 1.80</td>
<td>0.17 - 0.44</td>
<td>(Braune et al., 2016)</td>
</tr>
<tr>
<td>Common eider</td>
<td>?</td>
<td>↔</td>
<td>2008</td>
<td>0.73 ± 0.25; 0.37 - 1.45</td>
<td>0.13 - 0.48</td>
<td>(Akearok et al., 2010)</td>
</tr>
<tr>
<td>Long-tailed duck</td>
<td>?</td>
<td>↔</td>
<td>2008</td>
<td>0.85 ± 0.50; 0.41 - 1.56</td>
<td>0.13 - 0.46</td>
<td>(Akearok et al., 2010)</td>
</tr>
</tbody>
</table>
Table 2. Recent hepatic total mercury (THg) concentrations in μg/g dry weight (unless otherwise indicated) for marine birds from Arctic Canada. * Indicates a dry weight value calculated from reported wet weight value (assuming 70% moisture).

<table>
<thead>
<tr>
<th>Species</th>
<th>Year</th>
<th>Dry weight hepatic THg (mean ± sd; range of means given when available)</th>
<th>Source of data</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ivory gull</td>
<td>1998</td>
<td>2.67*</td>
<td>(Campbell et al., 2005)</td>
</tr>
<tr>
<td>Glaucous gull</td>
<td>1998</td>
<td>9.73*</td>
<td>(Campbell et al., 2005)</td>
</tr>
<tr>
<td>Arctic Tern</td>
<td>2007</td>
<td>4.15 ± 1.00</td>
<td>(Provencher et al., 2014a)</td>
</tr>
<tr>
<td>Black-legged kittiwake</td>
<td>1998</td>
<td>3.50*</td>
<td>(Campbell et al., 2005)</td>
</tr>
<tr>
<td>Black guillemot</td>
<td>1998</td>
<td>3.97*</td>
<td>(Campbell et al., 2005)</td>
</tr>
<tr>
<td>Thick-billed murre</td>
<td>2007-08</td>
<td>1.62 ± 0.12 - 4.41 ± 0.33</td>
<td>(Braune et al., 2014b)</td>
</tr>
<tr>
<td>Northern fulmar</td>
<td>2007-08</td>
<td>3.42 ± 0.53 - 6.99 ± 1.13</td>
<td>(Braune et al., 2014b)</td>
</tr>
<tr>
<td>Common eider</td>
<td>2008</td>
<td>2.68 ± 0.69 - 4.16 ± 1.62</td>
<td>Mallory unpublished</td>
</tr>
<tr>
<td>Long-tailed duck</td>
<td>1993-</td>
<td>0.9 ± 0.1 - 8.0 ± 3.4</td>
<td>(Braune et al., 2005)</td>
</tr>
<tr>
<td></td>
<td>1994</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Table 3. Availability of reported Hg values for bird species that feed within the marine environment, or prey on marine bird species, in the Canadian Arctic. Where data are available the date of last published data available is included in brackets.

<table>
<thead>
<tr>
<th>Species with Hg assessments within the last 10 years and/or in multiple locations</th>
<th>Species with limited Hg reports</th>
<th>Species currently with no reported Hg values</th>
</tr>
</thead>
<tbody>
<tr>
<td>ivory gull (<em>Pagophila eburnean</em>; 2004)&lt;sup&gt;1&lt;/sup&gt;</td>
<td>Thayer's gull (<em>Larus thayeri</em>; 1998)&lt;sup&gt;6&lt;/sup&gt;</td>
<td>Iceland gull (<em>Larus glaucooides</em>)</td>
</tr>
<tr>
<td>glaucous gull (<em>Larus hyperboreus</em>; 2008)&lt;sup&gt;2&lt;/sup&gt;</td>
<td>herring gull (<em>Larus argentatus</em>; 1992)&lt;sup&gt;7&lt;/sup&gt;</td>
<td>Sabine's gull (<em>Xema sabini</em>)</td>
</tr>
<tr>
<td>Arctic tern (<em>Sternula paradisaea</em>; 2008)&lt;sup&gt;3&lt;/sup&gt;</td>
<td>dovekies (<em>Alle alle</em>; 1998)&lt;sup&gt;8&lt;/sup&gt;</td>
<td>long-tailed jaeger (<em>Stercorarius longicaudus</em>)</td>
</tr>
<tr>
<td>black-legged kittiwake (<em>Rissa tridactyla</em>; 2008)&lt;sup&gt;2&lt;/sup&gt;</td>
<td>king eiders (<em>Somateria spectabilis</em>; 1997)&lt;sup&gt;9&lt;/sup&gt;</td>
<td>parasitic jaeger (<em>Stercorarius parasiticus</em>)</td>
</tr>
<tr>
<td>black guillemot (<em>Cepphus grille</em>; 2008)&lt;sup&gt;2&lt;/sup&gt;</td>
<td>common loon (<em>Gavia immer</em>; 1992)&lt;sup&gt;7&lt;/sup&gt;</td>
<td>pomarine jaeger (<em>Stercorarius pomarinus</em>)</td>
</tr>
<tr>
<td>thick-billed murre (<em>Uria lomvia</em>; 2008)&lt;sup&gt;4&lt;/sup&gt;</td>
<td>red-throated loon (<em>Gavia stellata</em>; 1992)&lt;sup&gt;7&lt;/sup&gt;</td>
<td>Atlantic puffin (<em>Fratercula arctica</em>)</td>
</tr>
<tr>
<td>northern fulmar (<em>Fulmarus glacialis</em>; 2008)&lt;sup&gt;4&lt;/sup&gt;</td>
<td></td>
<td>gyrfalcon (<em>Falco rusticolus</em>)</td>
</tr>
<tr>
<td>common eider (<em>Somateria mollissima</em>; 2008)&lt;sup&gt;5&lt;/sup&gt;</td>
<td></td>
<td>snowy owl (<em>Bubo scandiacus</em>)</td>
</tr>
<tr>
<td>long-tailed duck (<em>Clangula hyemalis</em>; 2008)&lt;sup&gt;3&lt;/sup&gt;</td>
<td></td>
<td>peregrine falcon (<em>Falco peregrinus</em>)</td>
</tr>
</tbody>
</table>

<sup>1</sup>Braune et al. 2006; <sup>2</sup>Dietz et al. 2013; <sup>3</sup>Akearok et al. 2010; <sup>4</sup>Braune et al. 2016; <sup>5</sup>Mallory unpub; <sup>6</sup>Campbell et al. 2005; <sup>7</sup>Braune et al. 1999; <sup>8</sup>Atwell et al. 1998; <sup>9</sup>Wayland et al. 2001.
Table 4. Hepatic mercury levels in Arctic marine birds from different areas. * Indicates dry weight values converted from wet weight values (Dietz et al. 1996). Italics represent highest values for the species where data are available for more than one region.

<table>
<thead>
<tr>
<th>Species</th>
<th>Sampling location and year</th>
<th>N</th>
<th>Figure</th>
<th>THg (µg/g dry weight)</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ivory gull</td>
<td>Northwater Polynya, 1998</td>
<td>2</td>
<td>mean</td>
<td>2.67*</td>
<td>Campbell et al., 2005</td>
</tr>
<tr>
<td>Glauco gull</td>
<td>Svalbard, 1980</td>
<td>11</td>
<td>mean</td>
<td>5.38</td>
<td>Norheim, 1987</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>range</td>
<td>2.69 – 7.73</td>
<td>Braune and Scheuhammer, 2008</td>
</tr>
<tr>
<td></td>
<td>Ungava Bay, 1983</td>
<td>5</td>
<td>mean</td>
<td>9.44</td>
<td>Braune and Scheuhammer, 2008</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>sd</td>
<td>4.74</td>
<td></td>
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<tr>
<td></td>
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<td>2</td>
<td>mean</td>
<td>6.78</td>
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<td>mean range with sd</td>
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<td></td>
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<td>1</td>
<td>mean</td>
<td>3.28</td>
<td>Kim et al. 1996</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>sd</td>
<td>3.66</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Svalbard, 2005-2006</td>
<td>10</td>
<td>mean</td>
<td>3.90*</td>
<td>Jaeger et al., 2009</td>
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<td>Arctic tern</td>
<td>Barents Sea, 1991-1992</td>
<td>10</td>
<td>mean</td>
<td>1.08</td>
<td>Savinov et al. 2003</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>sd</td>
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<td></td>
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<td></td>
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<td>10</td>
<td>mean</td>
<td>4.84</td>
<td>Kim et al. 1996</td>
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<td></td>
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<td></td>
<td>sd</td>
<td>3.66</td>
<td></td>
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<tr>
<td></td>
<td>Canadian high Arctic, 2007</td>
<td>41</td>
<td>mean</td>
<td>4.15</td>
<td>Provencher et al. 2014</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>sd</td>
<td>1.00</td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td>sd</td>
<td>1.43</td>
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<td></td>
<td>Barents Sea, 1991-1992</td>
<td>41</td>
<td>mean range with sd</td>
<td>0.45 ± 0.05 - 1.95 ± 0.44</td>
<td>Savinov et al. 2003</td>
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<tr>
<td>Location</td>
<td>Species</td>
<td>Year(s)</td>
<td>Sample Size</td>
<td>Mean ± SD</td>
<td>Reference</td>
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<tr>
<td>Canadian high Arctic, 1993</td>
<td>Black guillemot</td>
<td></td>
<td></td>
<td></td>
<td>Braune and Schehammer, 2008</td>
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<td>Campbell et al. 2005</td>
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<td>Thick-billed murres</td>
<td></td>
<td></td>
<td></td>
<td>Braune et al., 2014b</td>
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<tr>
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<td>Thick-billed murres</td>
<td></td>
<td></td>
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<td>Braune et al., 2014b</td>
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<td>Baffin Bay, 2007</td>
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<td></td>
<td></td>
<td></td>
<td>Braune et al., 2014b</td>
</tr>
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<td>Lancaster Sound, 2007</td>
<td>Thick-billed murres</td>
<td></td>
<td></td>
<td></td>
<td>Braune et al., 2014b</td>
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### Northern fulmar

<table>
<thead>
<tr>
<th>Location</th>
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<th>Measure</th>
<th>Mean</th>
<th>Range</th>
<th>Source</th>
</tr>
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<tbody>
<tr>
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<td>10</td>
<td>mean</td>
<td>7.06</td>
<td></td>
<td>Norheim, 1987</td>
</tr>
<tr>
<td></td>
<td></td>
<td>range</td>
<td>2.02 – 14.10</td>
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</tr>
<tr>
<td></td>
<td></td>
<td>sd</td>
<td>4.44</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Barents Sea, 1991-1992</td>
<td>15</td>
<td>mean range with sd</td>
<td>1.95 ± 1.29 - 5.72 ± 0.83</td>
<td>Savinov et al. 2003</td>
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<td>mean</td>
<td>8.12</td>
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<td>Braune and Schehammer, 2008</td>
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<tr>
<td>Northwater Polynya, 1998</td>
<td>10</td>
<td>mean</td>
<td>11.37*</td>
<td></td>
<td>Campbell et al. 2005</td>
</tr>
<tr>
<td>Lancaster Sound, 2007</td>
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<td>mean</td>
<td>6.99</td>
<td></td>
<td>Braune et al., 2014b</td>
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<td></td>
<td></td>
<td>sd</td>
<td>1.13</td>
<td></td>
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<td>Baffin Bay, 2007</td>
<td>10</td>
<td>mean</td>
<td>3.42</td>
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<td>Braune et al., 2014b</td>
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### Common Eider

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<th>Measure</th>
<th>Mean</th>
<th>Range</th>
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<td>mean</td>
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<td>Lande, 1977</td>
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<td>Svalbard, 1980</td>
<td>9</td>
<td>mean</td>
<td>3.36</td>
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<td>Norheim, 1987</td>
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<tr>
<td></td>
<td></td>
<td>range</td>
<td>1.68 – 5.71</td>
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<td></td>
</tr>
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<td></td>
<td></td>
<td>sd</td>
<td>2.02</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Barents Sea, 1991-1992</td>
<td>3</td>
<td>mean</td>
<td>1.8</td>
<td></td>
<td>Savinov et al. 2003</td>
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<tr>
<td></td>
<td></td>
<td>sd</td>
<td>0.22</td>
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<tr>
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<td>60</td>
<td>mean</td>
<td>3.15</td>
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<td>Wayland et al. 2001</td>
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<td></td>
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<td>range</td>
<td>0.86 - 9.81</td>
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<td>Victoria Island, Canada, 1997</td>
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<td>mean</td>
<td>1.01</td>
<td></td>
<td>Wayland et al. 2001</td>
</tr>
<tr>
<td></td>
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Table 5. Mercury concentration means in the breast muscle of Arctic marine birds from different regions. * Indicates dry weight values converted from wet weight values (Dietz et al. 1996). Italics represent highest values for the species where data are available from more than one region.

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3 Chapter: Direct and indirect causes of sex differences in mercury concentrations and parasitic infections in a marine bird

3.1 Abstract

In many animal species, males and females differ in their levels of contaminants and/or parasitic infections. Most contaminants and gastro-intestinal parasites are obtained through prey ingestion, and thus the causes of sex differences in the distribution of these factors might follow similar pathways. I studied the northern common eider duck as an avian model, and used directed-separation path analysis to explore the causes of sex differences in Hg and gastro-intestinal helminths. Two trophically transmitted helminths were examined: a cestode (*Lateriporus* sp.) and an acanthocephalan (*Profillicolus* sp). I found that the number of *Lateriporus* sp. varied positively with stable isotope values (as indicated by δ15N in eider breast muscle tissue), and negatively with crustaceans being present in the short term diet. I also found that *Profillicolus* sp. varied positively with eider tissue stable isotope values, indicating that birds at higher trophic levels had more helminthes. However, *Profillicolus* sp. varied negatively with sex indirectly through condition and liver mass. Similarly, Hg concentrations also varied negatively with sex indirectly through condition and liver mass, with both *Profillicolus* sp. abundance and Hg concentrations significantly higher in males. I found that model fit increased when a negative relationship between the two helminth species was included, suggesting a yet unknown causal mechanism linking these parasites. Our findings suggest that although Hg and gastro-intestinal parasites are both trophically transmitted through the eider’s prey items, the factors that contribute towards bioaccumulation of these two burdens
differ in source, likely caused by several different factors and may potentially influence each other.

3.2 Introduction

Two important factors that affect fitness in a wide range of wildlife taxa are parasites and contaminants (Marcogliese and Pietrock, 2011). In birds, for example, higher prevalence of parasites has been linked to reduced reproductive fecundity (Hudson and Dobson, 1991). Also in birds, high contaminant levels can cause neurological and physical changes (Ackerman et al., 2012; Scheuhammer et al., 2008). In many taxa, parasites and contaminants may differ between the sexes, resulting in patterns of sex bias within the same population. For example, Poulin (1996) found a consistent male bias in parasite infection in birds, fish and mammals in his review of male and female parasitism. Interestingly, sex differences in parasitism are not consistent across wildlife or parasite species. While some parasite species may show higher prevalence or abundance in male hosts, others are more prevalent in females of the same host species. In Richardson’s ground squirrels (Urocitellus richardsonii), adult males had higher levels of nematode infestations, whereas females had a higher prevalence of fleas within the same population (Waterman et al., 2013). Similarly, some heavy metals in Franklin’s gulls (Larus pipixcan) have been found in greater concentrations in males (Hg and chromium), whereas others showed higher concentrations in females (cadmium, lead, selenium and manganese; Burger and Gochfeld, 1996). As a result, there has been interest in the causes and implications of sex-specific differences in both parasites and contaminants given that both may negatively affect wildlife health and reproduction either additively or synergistically (Marcogliese and Pietrock, 2011).
The heterogeneous nature of parasite and contaminant distribution in wildlife species has led to many hypotheses on why such sex-related differences may occur, including variation in exposure. For trophically-transmitted parasites and contaminants, diets that differ between the sexes could result in differential exposure. Specifically, males and females may feed on different prey species that have unequal infection status and/or Hg concentrations (Burger et al., 2004; Robinson et al., 2009). Higher levels of Hg were found in female lizards (*Anolis sagrei*) when compared to male lizards of the same age and size occurring sympatrically (Burger et al., 2004). These differences were attributed to microhabitat preferences between the sexes which ultimately resulted in different prey being consumed by males and females.

Variation also can be caused by differences in the defensive responses to parasite and/or contaminants burdens. Differences in physiology may result in altered retention of contaminants and/or altered host defenses against parasites. For example, the sex with larger livers may have an increased capacity for Hg demethylation, a process that converts toxic MeHg to the less harmful inorganic Hg (Wiener et al., 2003). Sexual dimorphism in body size, and thus energetics, may also lead to differences in exposure and response to contaminants and parasites. The larger sex may consume more food (Key and Ross, 1999), leading to higher intake and exposure to both contaminants and parasites. Additionally, the reasons for variation in parasites and contaminants also likely vary from parasite to parasite, from contaminant to contaminant and from each other because of differences in host interactions with their environment and internal dynamics.

To examine external factors that may contribute to sex-related patterns in parasites and Hg I used a model species, the common eider duck, and a path analytic
approach (Shipley, 2009, 2000). Importantly, eider ducks are a part of a widespread subsistence hunt in eastern North America and southwest Greenland allowing for easy access to large sample sizes of birds (e.g. Jamieson et al., 2006). From these harvest collected samples diet variation can be studied through analysis of stomach contents (which indicate recent feeding bouts of their hard-bodied benthic prey; Piatt et al., 2007a) and stable isotopes analysis of eider duck breast muscle tissue (which indicate trophic level; Hobson et al., 1994) allowing for diet studies alongside parasitological and contaminants sample collections. Previous work has also shown that eider ducks carry varying levels of both gastro-intestinal parasites (Wayland et al., 2001b), and Hg contamination (Mallory et al., 2004), making them an excellent model species for this analysis.

I examined Hg in eider ducks as well as a large cestode (Lateriporus sp.) and acanthocephalan (Profillicolus sp.) helminths which are common gastro-intestinal tract helminths of eiders that are transmitted through their prey (Borgsteede et al., 2005; Camphuysen et al., 2002; McLaughlin, 2008). Our study was designed to test for sex differences in either contamination or parasitism or both, and then assess possible pathways leading to those sex differences using a form of path analysis. I had four subsidiary objectives. First, I tested if either or both of the two parasites examined would vary with the presence of crustaceans but not bivalves in the bird’s diet (crustaceans are intermediate hosts for the parasites). Our second objective was to test whether body condition was positively related to either or both parasites and Hg within either sex (noting that males are larger bodied than females in this species). Third, I also tested how
relative liver mass varied with Hg. Lastly, I tested whether Hg and parasites varied positively with trophic level.

3.3 Methods

3.3.1 Background and Rationale

I examined Hg and two gastro-intestinal parasites in eider ducks to explore patterns in these variables related to sex, condition and diet. Both helminth species are trophically transmitted to eiders who are the **definitive hosts**, whereas the crustaceans are the **intermediate hosts** (Brattey and Campbell, 1986; McLaughlin, 2008; Nikolov et al., 2008; Richardson and Nichol, 2008). Thus, I expected these helminth parasites to vary positively with the presence of crustaceans in the stomach. Additionally, since the known intermediate hosts of the parasites (crustaceans) often feed at a higher trophic level than bivalves themselves (Kharlamenko et al., 2013), I expected both helminth species to positively vary with eider duck tissue δ15N (which gives a longer timeline for eider diet than stomach content analysis alone).

Male eiders are generally larger than females (Goudie et al., 2000), as I also found in this study. Thus, I expected that sex might influence diet due to higher energy needs of males during the pre-breeding season (egg formation versus mate-guarding). Due to potential differences in energy content (in terms of calories) between crustaceans and bivalves, I hypothesized that eider duck body condition could be influenced by diet (both short term as reflected by stomach contents and long term as indicated by eider duck tissue δ15N). I also expected that eider duck structural body size would vary positively with parasites as larger birds eat more prey, and thereby potentially ingest more infective stages of parasites.
The liver is the body’s filtering mechanism, therefore, liver size can influence a bird either through direct filtering of the blood (i.e. trapping mercury), or indirectly through immunity which has been negatively linked with toxins in the blood stream (Grasman et al., 1996; Wiener et al., 2003). As a result, I tested whether liver mass varied negatively with parasite burden whose presence in a host can be affected by host immunity. Additionally, I expected birds with larger livers relative to their body size, to have lower concentrations of Hg. Importantly, I did not expect liver size to vary with sex when body size and diet was controlled for as a sex bias in organ size is not known in this species. I also did not expect liver size to vary with diet. For these reasons, linkages between liver size and either diet or sex were not explicit paths in our analysis.

As Hg is also transmitted to wildlife through their diet (Atwell et al., 1998), I expected Hg concentrations to positively vary with eider duck body size since larger individuals likely consume more prey. I expected Hg to vary with diet (positively with eider duck tissue δ15N and more likely to be positive with crustaceans versus bivalves) as Hg biomagnifies with increasing trophic level (Atwell et al., 1998). Lastly, I expected Hg to vary either directly or indirectly with sex as Hg has been shown to have a male bias in other marine birds (Robinson et al., 2012). Our analysis aims to simultaneously examine the direct and indirect effects of diet and size on the relationship between sex, Hg and the two parasites studied to help better understand what influences sex bias patterns in these two environmental burdens.

3.3.2 Collections

Eider ducks were sampled in Cape Dorset, Nunavut (64° 14’N, 76° 33’W), in collaboration with the local Aiviq Hunters’ and Trappers’ Association as part of the
annual aboriginal hunt in May 2011 (n = 165; males = 94, females = 71). Eiders were collected by hunters using 12-gauge shotguns and steel shot along the coastal areas within 25 km of Cape Dorset. All collections were done with the appropriate territorial and federal permits. Most birds were frozen within 12 h of collection, with a small number dissected in the community. Dissections of whole birds were completed in collaboration with the Nunavut Arctic College (Provencher et al., 2013). Prior to dissection each bird was measured for total dry body mass using Mettler Toledo PB5001-S scale. Total head (mm) was also measured on each individual using General digital calipers. From each bird, the gastro-intestinal tract was removed from the esophagus to the cloaca and temporally stored and sealed in a polyethylene bag for transport. In addition, the left breast muscle was removed and placed in an acetone-hexane rinsed glass jar using acid-rinsed chemically clean instruments. The liver was also removed, weighed and placed in an acetone-hexane rinsed glass jar. All tissues were then shipped to the National Wildlife Research Centre (NWRC) in Ottawa, Canada and stored at -40°C until processed which is outlined more specifically below in the relevant sections.

3.3.3 Stable isotopes

Analysis of stable carbon and nitrogen isotopes were performed University of Ottawa’s G.G. Hatch Stable Isotope Laboratory, Ottawa, Canada. Muscle tissues were homogenized and freeze-dried. Samples/standards were weighed in 1 mg subsamples of homogenized tissue and placed in tin capsules and loaded into an isotope ratio mass spectrometer where they were combusted at 1800°C. The resultant gases were carried by helium through columns of oxidizing/reducing chemicals optimised for N2. The gases were then separated by a "purge and trap" adsorption column and sent to the interface
before being measured by the isotope ratio mass spectrometer. Replicate measurements of an internal standards indicated errors were less than 0.2‰.

3.3.4 Gastro-intestinal parasites and diet items

All eider duck gastro-intestinal tracts were examined for prey and parasites in the intestine at the NWRC. Eider gizzards were opened and examined for both helminthes and prey items. Presence or absence of bivalves and crustaceans were recorded based on the prey remains found in each gizzard. As expected, the fragmented nature of the prey items within the gastro-intestinal tract I was unable to identify prey items to a more specific taxonomic level.

The intestine was separated from the gizzard and sliced down its entire length, while taking care not to cut any worms present. The intestine and its contents were rinsed with 0.85% saline solution, and any visible helminths removed using gentle suction or by removing the attachment site and leaving the tissue to soak for 24 h, and later removing the parasite scolices and proboscises from the softened intestine wall. The intestines were rinsed and the solution collected and examined for helminths using a binocular dissecting microscope. Parasites were enumerated by counting scolices and proboscises, and placed in 95% ethanol for identification. I used parasite abundance, which includes all hosts examined (Rozsa et al., 2000), in our model because of the associated other data I had from each individual and because I was interested in the casual mechanisms underlying the distributions.

3.3.5 Mercury analysis

Muscle samples were homogenized and prepared for chemical analysis at the NWRC in Ottawa. Muscle homogenates were weighed out into acid-washed plastic vials
and freeze-dried. Samples were weighed into nickel combustion boats, and analyzed for total Hg using an Advanced Mercury Analyzer (AMA-254) equipped with an ASS-254 autosampler for solid samples (see EPA Method 7473; Salvato and Pirola, 1996). This method uses direct combustion of the samples into an oxygen-rich atmosphere. Analytical accuracy was determined by using standard reference materials (Dolt-4, and Tort-2 from National Research Council and Oyster Tissue 1566b obtained from the National Institute of Standards and Technology), and 34 random muscle samples were analyzed in replicate. Both reference and replicate materials returned values within the confidence interval of certified reference values. The nominal detection limit for total Hg was 0.006 μg/g dry weight. Although total Hg was measured, I take this as a proxy of MeHg, the toxic form of Hg. It has been shown that in aquatic birds total Hg consists mostly of MeHg (Houserova et al., 2007).

### 3.3.6 Data treatment and statistical analysis

To explore direct and indirect effects of sex on gastro-intestinal helminths and Hg, I used a directed separation path modeling framework (Shipley, 2009, 2000). This method was chosen for several reasons. First, path modeling can easily accommodate the non-normal distribution of some of our response variables (e.g., parasite counts; Shipley, 2009, 2000). Second, it allows for the user to draw stronger inferences from correlational data than linear models (LM) and generalized linear models (GLM) do on their own (Shipley, 2009, 2000). Lastly, this method was chosen because it allowed us to assess and conceptualize both direct and indirect effects (Shipley, 2009, 2000). All data were analysed using R3.1.1 statistical software (R Development Core Team, 2013). All statistical tests were evaluated at α = 0.05 and means are presented ± SD.
I constructed our global path model based on previous knowledge of how both diet and condition are believed to influence gastro-intestinal helminths and Hg (see Methods and Fig 4). For our purposes condition was defined as body mass/total head length. I use this metric based on previous work done in the species that has shown this as a useful variable in modeling (Descamps et al., 2011a, 2010). The fit of the model was evaluated through directed separation (d-sep) tests (Shipley, 2009). In brief, for each path model there is a set of independence tests (the basis set) that must be true if the hypothesized causal structure of the model matches the correlational structure of the data. Global model fit was evaluated using Fisher’s C statistic: $C = -2 \sum \ln (p)$, which follows a chi-squared distribution with $2k$ df, where $p$ is the null probability of each d-sep test ($n = k$; Shipley, 2009). The global model is rejected if the $C$ value is below a significant $p$-value ($p < 0.05$), which is interpreted as the hypothesized causal structure of the model differs significantly from the correlational structure in the data. Each path was fit as appropriate to the data. This included transforming some metrics (log transforming Hg concentrations and liver mass), using a logit link function for the binominal response variables (e.g. presence of bivalves in the diet), and using hurdle models with a negative binomial distribution for the parasite data to account for the data being zero-inflated and over-dispersed.

Prior to fitting the path model, variables were assessed for correlations through a covariance-variance matrix of each individual equation making up the path model. All correlation coefficients were $\leq 0.6$. In order to calculate relative path coefficients for each path (i.e., standardized partial regression coefficients), each variable was standardized using z-score scaling ($V - \text{mean of } V / \text{standard deviation of } V$), except the parasite count
data. Total effect sizes for a given variable, where appropriate, were calculated by summing the direct effect and the products of the indirect effect sizes (Shipley, 2000).

3.4 Results

3.4.1 Model fit

Data from 165 eider ducks were used to test and parameterize our path model (Fig 4). Although δ13C was originally included in the model this variable was dropped from the final model as it was highly correlated with δ15N, and made the global model insignificant. Our final hypothesized model (Fig 4) was consistent with the correlational structure of the data (Fisher’s $C$ statistic = 7.40, df = 6, $p = 0.29$). Gastro-intestinal parasites and Hg appeared to differ in eider ducks according to sex, diet or body condition, but not necessarily in the expected directions (Fig. 5).

3.4.2 Body metrics

The average mass of female eider ducks was 1751 ± 146 g while the average mass of male eider ducks was 1941 ± 159 g. Male eiders were significantly heavier than females (GLM; $z_{164} = 61.4$, $p < 0.0001$). Body condition also varied with sex, being larger for males relative to females (GLM; $z_{164} = 4.19$, $p > 0.0001$; Fig. 5). Eider ducks in better body condition had greater liver mass (GLM; $z_{164} = 7.25$, $p > 0.0001$; Fig. 5). I did not find any evidence for a relationship between liver mass with sex, suggesting that liver mass varies indirectly with sex through body condition (Fig. 5). I did not find any evidence to suggest that condition varied directly with Hg concentration nor with either parasite burden (Fig. 5).
3.4.3 Stable Isotopes

The mean δ13C value for eider ducks was -19.00 ± 0.91‰ (n = 165). The mean δ15N value for the eider ducks examined was 11.93 ± 0.78‰ (n = 165). δ15N values did not vary either directly or indirectly with sex (GLM $z_3$, 161 = -0.10, $p = 0.58$; males: 11.91 ± 0.67‰, females: 11.95 ± 0.90‰; Fig 5).

3.4.4 Diet

The contribution of bivalves to the prey items in the gut contents consumed by male and female eider ducks at the time of collections differed, with males having bivalves present in their gastro-intestinal tracts more often than females (Fig 5; GLM $z_{164}$ = 2.20, $p = 0.03$). Interestingly, neither bivalves nor crustaceans significantly varied with long term diet as was predicted (as indicated by δ15N; Fig 5), suggesting that long term diet (as indicated from stable isotopes) and short term diet (indicated by gut contents) may not be consistent in nature over time.

3.4.5 Gastrointestinal parasites

In total, 6611 individual Lateriprous sp. and Profillicolus sp. helminths were found in our sample of 165 eiders sampled in 2011, with 37% of this total were Lateriporus sp, and the remaining 63% made up of Profillicolus sp. For Lateriporus sp, the median intensity was 4.0 helminths/host, with a mean intensity and standard deviation of 14.7 ± 30.0 helminths/host. A quarter of the birds examined (24%) of the eiders examined showed no evidence of infection by Lateriporus sp. The median intensity of Profillicolus sp. was 5 helminths/host, and the mean intensity was 25.4 ± 53.5 helminths/host. Similarly, a quarter (26%) of the eiders examined showed no infection of
Profillicolus sp. Overall, only 6% of the eiders examine showed no infections by Lateriporus sp. or Profillicolus sp.

The abundance of both the gastrointestinal helminths examined in the path model did not vary directly with sex. However, the parasite Profillicolus sp. varied indirectly with sex through body condition and liver mass, whereby females had higher parasitism when the other variables were controlled for (Fig. 5). Specifically, males had a higher condition index than females, liver mass varied positively with condition, and the abundance of Profillicolus sp. varied negatively with liver mass (hurdle; \( F_{164} = -0.43, p = 0.04 \); Fig 5). I also found that eider ducks feeding at a higher trophic level (as indexed by \( \delta^{15}N \)) had higher levels of both Profillicolus sp. acanthocephalans (effect size of 0.54; hurdle \( z_{164} = 2.90, p = 0.003 \); Fig 5) and Lateriporus sp. cestodes (effect size of 0.35; hurdle \( z_{164} = 2.19, p = 0.03 \); Fig 5).

During our initial evaluation of model fit, I found that not including a path between Profillicolus sp. and Lateriporus sp. resulted in poor congruence between our hypothesized model structure and the correlational structure of the data. However, I had no a priori reason to assume one direction was more biologically relevant than the other and therefore include paths for both directions in our final model. Specifically, I found that Lateriporus sp. varied negatively with Profillicolus sp. (hurdle; \( z_{164} = -2.46, p = 0.01 \); effect size = 0.010), and vice versa (hurdle; \( z_{164} = -3.38, p < 0.001 \); effect size = 0.014; Fig 5).

3.4.6 Mercury concentrations in breast muscle tissue

The average Hg concentration in eider duck breast muscle was 0.63 ± 0.24 µg/g. As predicted, our model showed Hg concentrations in eider ducks varied with sex; male
eider ducks had significantly higher concentrations of Hg in their breast muscle (0.69 ± 0.25 µg/g) as compared to females (0.56 ± 0.20 µg/g; GLM \( z_{3,161} = 3.94, p = 0.0001 \); Fig. 5; effect size 0.64). Moreover, our path model revealed both direct and indirect effects on Hg concentrations associated with sex. Specifically, while approximately 14% of the variation in Hg was explained directly by sex, the hypothesized model also showed an indirect effect of sex on Hg through condition and liver mass, where condition varied positively with sex (effect size = 0.07), liver mass varied positively with condition (effect size = 4.30) and liver mass had a negative relationship with Hg concentrations (effect size = -0.18; GLM \( z_{3,161} = -1.93, p = 0.05 \); Fig 5). Together our results suggest that higher mercury loads in males are only partially offset by having larger livers.

I also found some evidence (marginally insignificant \( p \) values) to suggest that both the presence of crustaceans in the stomach contents (short term diet; LM \( F_{164} = 1.91, p = 0.06 \)) and \( \delta^{15}N \) (long term diet; LM \( F_{164} = 1.91, p = 0.06 \)) varied positively with Hg (Fig 5).

### 3.5 Discussion

Patterns of sex differences in parasitic infection and contaminant levels have been observed in a range of taxa, leading to a number of hypotheses explaining why one sex may have a higher level of a particular burden than the other (Cordoba-Aguilar and Munguia-Steyer, 2013; Poulin, 1996; Schalk and Forbes, 1997). In this study, I used the eider duck to investigate the potential reasons for sex differences in two common eider parasites (a cestode, *Lateriporus* sp., and an acanthocephalan *Profillicolus* sp.) as well as Hg concentrations using directed path analysis. Both parasites and Hg are trophically-transmitted through diet in marine birds which allows these three factors to be examined
using a single model. As a consequence, sex bias in gastrointestinal helminths and Hg concentrations may be related to a number of attributes of an animal’s ecology associated with condition, diet and trophic level.

3.5.1 Parasites

I found no direct or indirect causal relationship between sex and the cestode *Lateriporus* sp. This suggests males and females have similar infections of *Lateriporus* sp. However, *Lateriporus* sp. abundance varied significantly with both short and long-term diet, indicating that diet is an important driver of parasite abundance. More specifically, *Lateriporus* sp. abundance varied negatively with the presence of crustaceans, indicating that eiders which ate more crustaceans over the short term paradoxically had lower levels of cestodes. This finding was counter to our prediction as I expected crustaceans in the diet to vary positively with *Lateriporus* sp., because crustaceans are known intermediate hosts for the parasite (McLaughlin, 2008; Schiller, 1954). This unexpected result may come from our lack of ability to identify stomach contents beyond genus. I could identify that crustaceans were present, but due to the rapid digestion of even very hard prey items within eiders, obtaining a level identification to genus was not possible. Therefore, I have no definitive way of identifying whether the prey were species known to carry *Lateriporus* sp. as intermediate hosts. Alternatively, this negative relationship between crustacean in the recent diet and lower *Lateriporus* sp. cestodes in the host may be related to differential infection status of prey intermediate hosts between foraging regions. For example, eiders may have ingested highly infected crustacean in their wintering grounds, as indicated by higher δ15N values. During their migration to the Canadian Arctic (where they were collected), eiders may be ingesting
crustaceans with lower parasite loads while still carrying the high parasite burdens acquired during the winter. This would lead to the inverse relationship that I found. More work on the intermediate hosts in the different regions is needed to test this hypothesis.

For the acanthocephalan *Profillicolus* sp. I found no direct causal relationship with sex, but I did find an indirect effect of sex on the abundance of *Profillicolus* sp. through condition and liver mass. Males that had a higher condition index generally had a larger liver mass, and lower levels of *Profillicolus* sp. There are several possible explanations for this finding. First, larger males may be feeding on prey that have lower levels of infection from *Profillicolus* sp. (i.e., larger males have less exposure to infection). Differential infection status in prey size classes targeted by smaller versus larger eiders could lead to this discrepancy in infection. Alternately, larger hosts with larger livers may have lower levels of toxins due to the filtering function of the liver (i.e., lower Hg concentrations were associated with larger liver mass). Since toxins negatively affect immunity (Sollome and Fry, 2015), hosts with larger livers, and thus fewer toxins, may maintain better immunity against parasites leading to lower infection levels.

Although no direct relationship was found between short term diet and acanthocephalans, the indirect negative relationship between bivalves in bird diets and acanthocephalans does suggest that large male hosts may select prey items that lead to lower infection levels.

Additionally, our model indicates that birds feeding at higher trophic levels have higher levels of both acanthocephalans and cestodes. Interestingly, the total indirect effect size of sex on *Profillicolus* sp. (-0.13) was smaller than the direct effect size of δ15N on *Profillicolus* sp. (0.54; Fig. 5). This suggests that infections of *Profillicolus* sp.
may be partially counter-balanced among the sexes if their trophic level at which they feed differs by gender. For example, larger birds eating at a lower trophic level have a lower *Profillicolus* sp. infection level than another bird of similar size eating at a higher trophic level. This illustrates how multiple factors of wildlife ecology may be driving these patterns in parasitism.

Originally, I did not include a path between the two parasite species considered in this analysis as I had no a priori knowledge that would lead to a hypothesis that one would directly affect the other. However, through the iterative process of the analysis I found that global model fit increased with the inclusion of this path. Due to the lack of a specific hypothesis of a causal relationship between the two parasites, I fit path coefficients in both directions. The negative correlation between both parasites indicates that as one parasite increases in abundance, the other decreases. This relationship may be driven by a direct relationship between the two parasites, or indirectly by another as yet unknown causal mechanism. With respect to a direct interaction, there may be direct competition between these two gastro-intestinal helminths. One species may make the local environment in the gastro-intestinal tract less suitable to other species. Further work should be done to explore the spatial relationships of these parasites in the guts of marine birds to better understand their interactions.

### 3.5.2 Mercury

Consistent with our predictions, Hg concentrations in eider ducks varied both directly and indirectly with sex. Our model suggests that although males may have higher concentrations of Hg, those that are in better condition and have larger livers actually have lower concentrations of Hg than their smaller counterparts. This indicates that both
condition and liver mass (perhaps through increased demethylation capabilities) potentially play a role in Hg concentrations of eider ducks as has been shown in other species (Eagles-Smith et al., 2009; Kim et al., 1996b).

I found that the direct relationship (0.64) between sex and mercury was larger than the total indirect effect (-0.05), indicating that some other variable or variables not considered, which are associated with sex, are larger drivers of Hg than condition and liver mass alone. Beyond size and diet composition, there are a number of factors that may be leading to male eiders having higher concentrations of Hg. I acknowledge that some reasons may indirectly relate to diet that I was unable to quantify. For example, males may eat the same prey items, but eat more of them thus increasing their exposure to Hg. Alternatively, males may be eating in a different region from the females over the winter and spring months (Mosbech et al., 2006), which could lead to differential exposure. Another possible factor influencing sex bias among contaminants in birds is the depuration to eggs by females (Donaldson and Braune, 1999; Mallory et al., 2006a). However, the ducks in our study were sampled in May, prior to egg formation by females. Thus, seasonal depuration of Hg to the eggs by females had not yet occurred, and cannot explain the differences in Hg concentrations found (see also Robinson et al., 2012). Nonetheless, depuration over the lifetime of a female may contribute to a lower body burden of Hg as compared with the males in the study. Interestingly, no differences in Hg concentrations from breast muscle have been found in two other Arctic marine bird species (the northern fulmar (Fulmarus glacialis) and the thick-billed murre (Uria lomvia)) suggesting that egg formation by females does not necessarily lead to the presence of a sex bias in Hg concentrations (Braune et al., 2014b). Variation in sex bias
patterns in Hg between marine bird species suggests there are different mechanisms which influence lifetime Hg load.

Other factors related to bird movement may play a role in patterns of sex bias observed among trophically-transmitted burdens such as sex-specific segregation during migration. Tracking data from northern common eider ducks shows that during the post-breeding season, males and females in the Canadian Arctic often move to different areas; males leave the breeding colonies to moult before female incubation at the colony is complete. Thus males travel farther from colonies before initiating their moult (Mosbech et al., 2006). This may occur on a local scale, such as differences in habitat use within a region (Schwab et al., 2005), or during migration (Fort et al., 2014; Leat et al., 2013). Such differences in migration patterns could lead to variation in contaminant burdens acquired as there may be regional differences in the contaminant concentrations of prey species (Baert et al., 2013). For example, adult great skuas (*Stercorarius skua*) have significantly different organochlorine concentrations in blood plasma depending on where they winter (Bourgeon et al., 2013). These types of variations in migration patterns could lead to the sex bias in Hg and gastrointestinal helminths as observed among male and female common eiders.

### 3.5.3 Common causal relationships between parasites and mercury

Interestingly, both *Profillicolus* sp. and Hg concentrations showed similar indirect effects to each other (i.e., an indirect negative effect through condition and liver mass). While reduced Hg concentrations with larger liver mass may be attributable to the demethylation that occurs in the organ (Kim et al., 1996b), the relationship between liver mass and lower abundance of gastro-intestinal acanthocephalans is less clear and may be
related to toxins and immunity. These relationships should be further explored as parasites and contaminants are known to interact in complex ways (Marcogliese and Pietrock, 2011).

Typically in marine ecosystems, a higher δ15N value indicates a higher trophic level, which generally has a positive correlation with Hg concentrations (Atwell et al., 1998). Although a positive correlation was found between δ15N and Hg concentrations in eider breast muscle, the relationship was not quite significant (p=0.06). One reason for this finding may be linked to the low variability in both the δ15N values and the Hg concentrations observed in eider ducks. The positive relationship between δ15N and Hg concentrations may only become apparent when compared across large ranges of δ15N and Hg as done with multispecies comparisons (Akearok et al., 2010; Atwell et al., 1998) versus within species as I have done here. Although the positive relationship between δ15N and Hg concentrations in breast muscle was not quite significant, it is important to note that eider ducks with higher δ15N values showed higher values of both parasites and Hg in our model. Together this suggests that typically birds that feed at higher trophic levels have higher burdens of parasites and Hg. This finding has implications for studies interested in examining the effects of parasites and contaminants. If individuals who have higher concentrations of parasites also have higher levels of Hg, any effects study must account for the shared effects (Morrill et al., 2014). Similarly, high levels of multiple burdens may lead to cumulative effects, which must also be taken into account when assessing how burdens impact wildlife (Marcogliese and Pietrock, 2011).

It is important to note that our study was limited to examining the diet of individuals through the stomach contents and stable isotopes from breast muscle. While
the stomach contents likely indicates prey items take in the hours before collections (Piatt et al., 2007b), stable isotope values likely reflect a diet signature from approximately 23 days before collections (Hobson and Clark, 1992a). Therefore, the patterns observed here likely result from the eider’s diet shortly before and during the migratory period into the Canadian Arctic, but Hg is accumulated over much longer periods of time (Wiener et al., 2003), and likely so are parasites. This suggests that the patterns of Hg and parasite accumulation reflect relationships between Hg, parasites, prey and eiders during the spring season. Whether these relationships hold true for the rest of the annual cycle for eiders warrants further investigation.

I acknowledge that one metric I was unable to measure in the sample was age of the birds. Host age can play a strong role in the likelihood of infection by some parasites (Lopez et al., 2013). Although all the eider ducks examined in this study were adults (at least two years old), more specific age information was not available. It is possible that older birds (eiders can live as long as 20 years but on average live 7.5 years; Goudie et al., 2000) accumulate more Hg and gastrointestinal helminths. If age structure within a population differs between males and females, this could also contribute to a sex bias being present in either Hg and/or helminths.

3.6 Conclusions

Collectively, I found that sex, diet, and condition can directly and indirectly influence parasite and Hg burdens among eider ducks. Our results indicate that the causes of sex differences in parasitic infections and contaminant levels in a marine bird such as the eider duck can be independent of each other. They may be related to sex, condition and diet but also potentially to other factors not measured here such as local
environmental conditions and migration strategies. Importantly, these patterns in sex biases may be extended to a population, which highlights the need to examine species of interest across large geographic scales when investigating possible biases in parasite and contaminants levels as well as their interactions. Furthermore, where multiple burdens share common causal relationships, studies interested in the effects of burdens such as parasites and contaminants need to consider how to account for other stressors with similar impacts and distribution patterns within wildlife hosts.
Figure 4. Full path model showing the hypothesized relationships between sex, diet, body metrics, Hg concentration and two gastro-intestinal helminthes (Lateriporus sp. and Profillicolus sp.) in common eider ducks (Somateria molissima) during their spring migration. Solid black arrows represent significant relationships ($p < 0.05$). Numbers associated with each line represent the standardized path coefficients. Dashed black lines represent two significant independent paths that were found to increase global model fit, but without a priori or post hoc rationale for one direction to be fitted over the other. Solid gray lines represent slightly non-significant lines ($p = 0.06$) and dashed gray lines represent hypothesized relationships that were found to be non-significant ($p > 0.05$).
Figure 5. Simplified path model showing the relationships between sex, diet, body metrics, Hg concentration and two parasitic helminths (Lateriporus sp. and Profillicolus sp.) in common eider ducks (Somateria molissima) during their spring migration through the Canadian Arctic. Only paths that were significant ($p < 0.05$) were maintained. Numbers associated with each line represent the standardized path coefficients. Green arrows represent positive relationships, red arrows represent negative relationships and grey arrows represent significant differences between the sexes.
Chapter: Anti-parasite treatment, but not mercury burdens, influence nesting propensity dependent on arrival time or body condition in a marine bird

4.1 Abstract

Arctic wildlife can be exposed to high Hg levels, and are also naturally exposed to gastrointestinal parasites that can reduce condition and negatively affect reproductive output and/or survival in similar ways. Importantly, both Hg and parasites are increasing in wildlife in some Arctic regions. I studied the northern common eider duck to explore how Hg in association with both natural levels and experimentally reduced parasitic infections, affect reproduction and survival. Female eiders were measured, banded, and blood sampled to determine blood Hg burdens, prior to breeding. Propensity to nest, clutch size, nest survival, nest attendance, and return rates were assessed in relation to both Hg burden and parasite treatment. Neither reproduction nor return rates of females varied with Hg concentrations, but females arriving late to the colony, or in low body condition, showed increased nesting propensity when given the anti-parasite treatment as compared to placebo treatment. Our results suggest that parasites can play a critical role in decisions to invest in avian breeding annually, particularly among individuals with a late onset to breeding, and in poor condition.

4.2 Introduction

Schedules of breeding often show marked variation within a population, which may be driven by a number of biotic and abiotic factors. In migratory birds, body condition (e.g., adipose tissue) can be a key driver of inter-individual variation in the likelihood that individuals nest as well as the number of eggs they lay (Ankney, 1992; Descamps et al.,
Both parasitism and contaminant burdens have been found to independently influence body condition in birds (Daoust et al., 1998; Sheldon and Verhulst, 1996), and therefore may be expected to explain variation in breeding propensity and productivity (Hanssen et al., 2003). Understanding how contaminants and parasites influence individual reproduction, both independently and jointly has been examined only in a few studies to date, but is crucial to understanding their potential as drivers of population dynamics (Bustnes et al., 2006; Marcogliese and Pietrock, 2011; Reed et al., 2008; Wayland et al., 2002).

Contaminants acquired through diet have the potential to deleteriously affect wildlife health and condition in several ways. In both wild birds and mammals, individuals with high Hg concentrations may exhibit neurological, physical and hormonal changes (Scheuhammer et al., 2008; Sonne, 2010; Van Hoomissen et al., 2015). This suggests that Hg can potentially influence a range of behaviours, including foraging and migration, which, in turn can lead to an overall decline in body condition. For example, in clapper rails (Rallus longi; Ackerman et al., 2012) and common eider ducks (Wayland et al., 2001a), body mass was negatively associated with Hg burdens. Mercury also influences reproduction and survival in wild birds. Black-legged kittiwakes with higher blood Hg concentrations were more likely to skip breeding than those with low Hg (Tartu et al., 2013). White-winged scoters (Melanitta fusca) with higher levels of Hg showed lower annual survival rates (Wayland et al., 2008). Collectively, these findings suggest that Hg can induce changes in physiology, which can then manifest in a bird through reductions in body condition, reproductive output and/or survival.
To assess the manifold effects of Hg on wildlife, it is important to study Hg in concert with other factors expected to affect individual or population health. Parasites can affect an animal’s reproduction and condition because they deplete host resources and/or compete for nutrients with their hosts. For example, red-legged partridges (*Alectoris rufa*) in Spain with higher intensities of parasitic helminth infestations (*Capillaria contorta*) had reduced body condition (Calvete et al., 2003). Parasites also can negatively influence the demography of host populations. Female red grouse (*Lagopus lagopus scoticus*) infected with nematodes (*Trichostrongylus tenuis*) had smaller clutches which led to cyclical patterns in grouse population levels (Hudson and Dobson, 1991). In comparison, higher levels of parasitism were associated with lower survival in young female snow geese (*Chen caerulescens*), illustrating that parasites can have long term effects on host demographics (Souchay et al., 2013). Of note is that parasite infections in wild birds may not be associated with any decline in reproduction or survival (Perez-Orella and Schulte-Hostedde, 2005; Shutler et al., 2012), and reducing parasites has been shown to alter avian survival (Hanssen et al., 2003).

Importantly, the relationships between contaminants and parasites, and their direct and indirect influence on host condition, survivorship and reproduction of fish and wildlife can be complex (Bulté et al., 2012; Marcogliese and Pietrock, 2011; Provencher, 2013). For instance, the host typically mounts an energetically-costly immune response when exposed to parasites, which can vary with parasite exposure rates (Schulenburg et al., 2009). The presence of parasites and their effect on the host immune response can influence how a host responds to contaminant exposure by altering host contaminant uptake and retention (Marcogliese and Pietrock, 2011). Parasites also can alter the energy
budget of a host causing an increase or decrease in metabolic and/or foraging rates (Bergey et al., 2002). In turn, this may increase or decrease a host’s exposure to trophically-transmitted contaminants such as Hg.

To investigate how Hg affects reproduction and survival in wildlife under conditions of natural and reduced parasitism, I undertook an experimental anti-parasitic manipulation of female eider ducks breeding at a colony in northern Canada. Eider ducks are well-studied in the circumpolar Arctic, and are relatively easy to mark, track and monitor over time and while breeding (Descamps et al., 2009; Goudie et al., 2000; Love et al., 2010). Eider ducks are also useful models for studies that examine parasite and contaminant interaction because the prevalence of parasites is often high (Wayland et al., 2001b). To experimentally broaden the range of parasite burdens in wild eider ducks a sample of breeding females were given either an anti-parasite treatment (PANACUR® Hoechst Roussel Vet GmbH) or a placebo of distilled water following Hanssen et al. (2003). A combination of direct colony observations and remote logging devices were then used to assess female behaviour and productivity throughout the breeding season in relation to their parasite treatment and naturally occurring blood Hg burden.

Our general hypothesis was that variation in intensities of parasite infections and in contaminant burdens would be associated jointly or independently with eider duck reproduction and survival. Such studies of free-ranging wildlife pose challenges for two main reasons: first, one can experimentally manipulate parasitism, but not easily manipulate Hg burden; and second, wildlife come ’pre-packaged’ with their contaminant burdens which vary by region and year. From this perspective, I generated several predictions. First, I expected if the range of Hg concentrations was great, higher blood
concentrations of Hg in females would be associated with decreased measures of reproduction or survival. Since eider hens go through a fasting period during incubation, losing up to a third of their body mass (Bustnes et al., 2010), I also expected nest attendance to be negatively related to their Hg burden. Mercury can reduce condition so that females take more incubation recesses. Therefore, I expected that females with lower levels of blood Hg to have fewer absences from their nests as measured through automated nest data loggers. I also expected individuals given the anti-parasite treatment to have higher measures of reproduction and survival, and fewer absences from the nest as compared with those receiving the placebo. I anticipated that an experimental decline of intestinal parasites would enhance a female’s ability to acquire and retain energy as compared with birds only given a placebo treatment. Lastly, I expected that eider ducks with higher levels of both Hg (higher blood concentrations) and parasites (placebo treated birds) would show the lowest measures of reproduction and survival, and have the most absences from the nest during incubation.

4.3 Methods

4.3.1 Capture of birds

Eiders were caught at the East Bay Migratory Bird Sanctuary, Nunavut (64°01’04’N, 82°07’49’W) in northern Hudson Bay as they returned to the Arctic during their spring migration. Hens were caught using monofilament nets as they were arriving at the colony in June, 2013 and 2014. More detailed information on capture and handling can be found in Hennin et al. (2014). Birds were caught prior to laying their eggs on the island, which continues for several weeks when the weather allows, until most females have arrived and have initiated nesting. The colony is surrounded by sea ice during the
pre-breeding season so that all birds are caught in flight as they arrive on the island. Band reading for mark-recapture studies occurs daily during and after the banding season, so that the arrival date of females to the colony can be estimated.

At the time of capture, a metal US Fish and Wildlife Service band was placed on the right tarsus of each bird, as well as individually coded plastic colour bands on both legs. Females were also fitted with a unique combination of nasal disks allowing for individual identification during incubation. The disks were attached to the nares using UV-degradable surgical monofilament so that the disks fall off shortly after the breeding season and prior to fall migration. Upon capture, weight (body mass in g) and total head length (in mm) was taken for each female following banding. Birds captured as pairs were released as pairs.

4.3.2 Anti-parasite and placebo treatments

During capture in 2013 and 2014, females were also given an oral dose of either an anti-parasite treatment (PANACUR®) or a placebo treatment (distilled water). PANACUR® is a broad spectrum anti-parasite treatment (active ingredient is fenbendazole) that effectively removes nematodes, lungworms and cestodes in birds, as well as acanthocephalans in monkeys (Norton et al., 1991; Weber and Junge, 2000; Yazwinsky et al., 1993, 1992). I alternated giving either the anti-parasite treatment or the placebo treatment to captured females. Those females designated for the anti-parasite treatment were given 0.5 ml (50 mg fenbendazole, 26 mg/kg body mass) oral dose of 10% PANACUR®, following procedures used in other wild bird species, including common eiders (Bustnes et al., 2006; Hanssen et al., 2003). Females treated with the placebo were given 0.5 ml of distilled water.
4.3.3 **Blood sampling**

Following capture, a blood sample (0.5-1 ml) was taken from the jugular vein using a 23G thin wall, 1-inch (c. 25 mm) needle attached to a heparinized 3 ml syringe. Whole blood samples were placed in acid-rinsed vials, kept at 4 °C, and subsequently frozen within 6 h of collection for analysis of individual Hg concentrations.

4.3.4 **Colony observations**

I consider arrival date on the island to be the first time an individual female was caught or seen each spring. The colony on East Bay Island is monitored annually from observation blinds so that disturbance to the nesting eiders is minimized. During the years of this study, observers scanned the breeding colony in search of nasal-tagged females for 10 to 11 h/d. All nasal-tagged females were recorded, their nesting sites mapped, and the fate of their nests monitored. Band reading was also conducted during daily plot checks, in which observers recorded any banded bird they observed. Combined, these methods provided daily coverage of the eider colony, regardless of weather conditions.

Once eiders established nests and the rate of new arrivals declined, observers visited the nests of most nasal-tagged females by slowly crawling to minimize disturbance. Where possible, clutch size was determined and eggs were candled to confirm clutch lay date. In 2013 and 2014 temperature and light loggers (HOBO® pendant loggers) were also placed in some nests of nasal-tagged females that had been experimentally manipulated (both those given PANACUR® and placebo treatments). These devices allowed us to continually monitor nest attendance patterns in relation to treatment. Loggers were set to record the light and temperature every minute until the memory filled (21 days) at which time recording stopped. Unfortunately due to high rates
of polar bear (*Ursus maritimus*) depredation of eider nests on East Bay Island, the number of chicks and number of fledglings in relation to female treatment could not be studied.

### 4.3.5 Laboratory work

Analysis of Hg concentrations in eider blood was done by RPC Laboratories. Each sample was prepared by microwave assisted digestion in nitric acid before analysis. Mercury was analyzed by cold vapour Atomic Absorption Spectroscopy (AAS), and the detection limit was 0.01 μg/g. Quality assurance/quality control (QA/QC) procedures included analysis of three reagent blanks, two certified biological reference tissues (DORM-4 and DOLT-4; National Research Council, Canada), two standard samples to assess between batch reproducibility, and two randomly selected duplicate samples per batch of 35 samples. All QA/QC measures were in compliance with normal laboratory operating procedures at the time of analysis. All Hg blood results are presented in wet weights (ww).

### 4.3.6 Mercury data

Mercury data were tested for normal distribution (Shapiro-Wilks) and equal variances (Levene’s test). Mercury was square root transformed to achieve the assumptions of parametric analysis. All females had Hg concentrations above detection rates.

### 4.3.7 Statistical Analysis

I use reproduction principally to refer to propensity to breed, nest survival, attendance and clutch size, whereas survival refers to return to the colony the year after the treatment (a reasonably good measure of survival for the philopatric eider; Wayland
et al., 2008). I classified a female eider as “breeding” when she was observed to occupy a nest cup for a minimum of three consecutive days. Breeding propensity was defined as an individual’s tendency to breed in a given year, while nest initiation was the date when a hen first occupied a nest. The last day of incubation for each female was determined by observing all nests twice daily and at least one day following the last sighting of the female on the nest to confirm her absence. Previous studies have shown that arrival date and condition can also affect individual breeding metrics at this colony (Descamps et al., 2011a). Therefore, arrival date and condition index were also examined as predictor variables along with treatment. I used size corrected mass as an index of condition. To test if the experiment itself had an influence on the birds regardless of treatment I used a Pearson’s Chi² test to compare the frequency of resighting and nesting propensity of all nasal tagged females post-banding in the experimental years (2013 and 2014) with two years prior to (2011 and 2012), and one year after the experiment (2015).

I used several methods to examine if treatment level and Hg burdens influenced female resight, propensity, and clutch size in the treatment year. First, a generalized linear model (GLzM, binomial distribution and logit function) was used to test for differences in probability of resighting PANACUR vs. placebo individuals in association with their Hg burdens. Second, I tested for an effect of treatment on nesting propensity in relation to combinations of predictor variables (including Hg concentrations and parasite treatment) using a GzLM, with a binomial distribution and breeding propensity as the response variable. Third, I used general additive models (GAM) with a smoothing function when variables such as arrival date and a condition index (mass/total head) and the response variables (e.g., propensity to nest) were expected to vary non-linearly across
the range of values. Lastly, I examined how both parasite treatment and Hg burden affected clutch size for a subset of birds where data were available. Clutch size was determined from direct observations when nests were visited for data logger placement of 64 available nests (2013 – 29; 2014 – 35). Clutch size data were examined using a GLzM (Poisson distribution, log function).

To further explore how parasite treatment affects nest survival I used the program MARK to calculate an average daily nest survival estimate for the two treatment groups (Dinsmore et al., 2002). I first examined daily nest survival in relation to several potential factors known to influence nest survival, including year, age of the nest and time of season. The latter was tested using linear and quadratic functions of date, which examine whether daily nest survival shows a linear or curvilinear trend respectively over the course of the nesting season. Prior to testing treatment effects, I used Akaike’s Information Criterion (AIC; Burnham and Anderson, 2002) to examine support for a set of candidate models containing effects of year, nest age and time of season. Support for the different models was evaluated using AIC for small sample sizes (AICc) and Akaike weights (wi). Once I had identified a top model containing these other effects, I included treatment to test whether daily nest survival differed between PANACUR treated and control birds. I present all candidate models.

Additionally, I investigated if experimental removal of parasites and Hg burdens influenced year to year return rate (resight in n + 1). I used a GzLM (binomial distribution, logit function) with probability of return the year after females were treated as the response variable. I also examined the probability of return in the year following
treatment in relation to arrival date and condition index because these two variables could
influence how treatment affected individuals.

All statistical approaches were performed in R (R Development Core Team, 2013), unless otherwise indicated, and tests were assessed with the significance level of 0.05. When appropriate for each model I present the β estimate, the standard error (se), the appropriate test statistic and the p value.

4.3.8 Nest logger data

Only logger devices that were found in the nest cup upon retrieval and in nests that were not depredated were used for analysis as some loggers were displaced from the nests during the incubation period. The light readings were 0 lux when females were sitting on the nest (confirmed through comparing logger data with nest observations). Thus, nest disturbances were detected when light levels rose above 0 lux, and also confirmed with temperature readings (greater than ambient temperatures from camp weather station within 0.5 km of all nests). The return time for birds was calculated as the time between the deployment of the logger until the hen had returned to her nest and the light reading registered again at 0 lux. Once the bird had returned to the nest, disturbances of a single minute were classified as ‘shuffles’; those that lasted more than one minute were considered ‘multi-minute disturbances’ (or mmd). Activity at the nest included the total number of shuffles, average number of shuffles/day, the total number of mmd, average number of mmd/day, average length of mmd, cumulative time over the incubation period of mmd/by the number of days, and the cumulative time of mmd and shuffles/by the number of days. All nest attendance metrics were determined from the logger data and compared between the two treatment groups as well as individual Hg
burdens. Linear models (LM) were used to compare nest attendance metrics with parasite treatment and blood Hg concentrations.

4.3.9 Anti-parasite treatment in captive males

In 2013 a sample of male eider ducks was caught concurrently with nasal-tagged females to test the efficacy of the PANACUR® anti-parasite treatment on parasite burdens. Lone males were caught using monofilament nets as they were arriving at the colony during the same collection period as the females. Similar to the females, males were alternatively given either the anti-parasite treatment (0.5 ml of 10% PANACUR®; 50 mg fenbendazole, 26 mg/kg body mass) or the placebo (0.5 ml of distilled water). Males were subsequently kept in outdoor cages (wooden structure surrounded by wire mesh, 0.4 x 1.4 x 1.4 m) for 24 h. After this time males were euthanized using cervical dislocation in accordance with animal care protocols. Carcasses were frozen immediately, dissected at the Nunavut Arctic College (Provencher et al., 2013), and all gastro-intestinal tracts were subsequently shipped to the National Wildlife Research Center in Ottawa for parasitological examination. A more detailed description of parasite enumeration and identification can be found in Provencher et al. (2016; or above in Chapter 3).

Species specific and overall helminths prevalence (using unconditional exact test), mean intensity (bootstrap-t test), median intensity (Mood's median test), and mean abundance (bootstrap-t test) were then compared between the two male treatment groups using the Quantitative Parasitology program (Reiczigel et al., 2013), as suggested for small sample sizes (Rozsa et al., 2000). I expected that those treated with PANACUR® would have lower prevalence, intensity and abundances of helminths than those given the
placebo treatment of distilled water. All data and metadata from this study is available via the Polar Data Catalogue (CCIN reference number 11810).

4.4 Results

A total of 231 females were captured, sampled and treated as part of the experimental manipulation in 2013 and 2014 (Table 6). The average blood Hg concentrations of female eiders was significantly higher in 2014 as compared with 2013 (GLM $z_{1,219} = 20.38, p < 0.001$; Table 6). Clutch size did not differ significantly in 2014 and 2013 (GLzM $z_{63} = 0.24, p = 0.12$; Table 6). I found no significant difference between treatment groups in mass (LM $F_{1,219} = 0.21, p = 0.65$), Hg (LM $F_{1,219} = 1.32, p = 0.26$), or condition (LM $F_{1,219} = 3.6, p = 0.55$) at capture. I therefore attribute any differences between treatment groups in breeding propensity, nest survival, nest attendance or return rates as treatment effects. Only one female was sampled in both years, and was discarded from the analysis in the second year.

4.4.1 Effects of the experimental manipulation

In 2013, 87% ($n = 106$) of the females involved in the experimental manipulation were observed again on the island at least once following capture and release. In 2014, 71% ($n = 77$) of the experimental individuals were resighted. These resight levels were similar to other years within the long term study at the colony. I found significant, within-year resight differences among years (Table 7; $\chi^2 = 34.7$, df = 4, $p < 0.001$), including between the two experimental years (2013 and 2014, $\chi^2 = 10.5$, df = 1, $p = 0.001$). This suggests that there are inter-annual differences in within-year resight rates outside of this experimental manipulation that likely influence whether birds remained on the colony (and thus were re-sighted) after initial capture and release each year (e.g., weather, local
foraging conditions due to ice cover). Given the difference in within-year resight rates between 2013 and 2014, year was included as a covariate for the subsequent return rate analyses. Resight rates in the year after treatment did not vary between the treatment years (GzLM \( z_{218} = 1.24, p = 0.22 \)), so year was not considered further.

Overall nesting propensity of female eiders did not differ in the experimental years (2013 and 2014) as compared with other years (2011-2012 and 2015). In 2013 and 2014, respectively, 41\% (n=53) and 45\% (n=50) of the females that were captured, treated and banded went on to nest on the East Bay colony. This proportion did not differ significantly to other years at the East Bay colony (Table 7; \( \chi^2 = 2.72, \text{df} = 3, p = 0.59 \)), suggesting that the experimental treatment did not alter the proportion of females that went on to nest. Given that nesting propensity within-year did not differ between the two treatment years, I considered both years together in further analysis.

4.4.2 Effects of Hg

The probability that females eiders were resighted during the year of treatment was not significantly correlated with blood Hg concentrations sampled at capture (GzLM \( z_{218} = 0.84, p = 0.40 \)). Female eiders tended to have greater nesting propensity if they had higher blood Hg concentrations (GzLM \( z_{219} = 1.77, p = 0.08 \)), but the relationship was not significant. Clutch size was also not significantly related to maternal Hg blood concentrations in either year (GzLM \( z_{63} = 0.09, p = 0.92 \)). Moreover, the probability of females returning the year after treatment was not influenced by Hg concentrations in the blood (GzLM \( z_{218} = 0.16, p = 0.88 \)).

4.4.3 Effects of the anti-parasite treatment
As expected, within-year resighting of female eiders on the breeding colony did not differ with treatment type (anti-parasite vs. placebo) in either year (GzLM $z_{218} = -0.04, p = 0.97$). Resight of females in the year following the treatment, an indication of survival, were similar regardless of whether they received PANACUR® or a placebo ($GzLM z_{218} = -0.73, p = 0.46$). This suggests that the anti-parasite treatment showed no carry-over effects into the next breeding season. Furthermore, female eiders had similar return rates in the year following treatment after accounting for their body condition in the treatment year ($GzLM z_{218} = 0.63, p = 0.53$). Female eiders that arrived earlier in the season tended to have a higher return rate in the following year ($GzLM z_{218} = -1.88, p = 0.06$).

Contrary to our expectations, nesting propensity did not differ significantly between the two treatment groups ($GzLM z_{218} = 1.50, p = 0.14$). I also found that condition of eiders upon their arrival did not vary significantly with either arrival date ($GzLM z_{218} = -0.44, p = 0.66$), or by year ($GzLM z_{218} = 1.24, p = 0.22$; Table 6). Therefore, I tested both condition and arrival date as separate predictor variables. The nesting propensity of the placebo treatment group declined significantly as the breeding season progressed ($GAM \chi^2 = 16.2, p < 0.0001$; Fig. 6A). By contrast birds with experimentally reduced parasite burdens showed no difference in breeding propensity in relation to the arrival date ($GAM \chi^2 = 12.4, p = 0.10$; Fig. 6B). When the arrival condition of the eiders was considered with treatment, females given placebos exhibited greater nesting propensity if they were in better condition ($GAM \chi^2 = 7.5, p = 0.006$; Fig. 6C), suggesting that birds arriving in poor condition were less likely to initiate breeding. The breeding propensity of birds that were given the anti-parasite treatment did not
significantly differ across condition (GAM $\chi^2 = 1.36, p = 0.24$; Fig. 6D), indicating that individuals arriving in poor condition but then had their parasite burdens reduced were significantly more likely to initiate nesting. I found no significant difference in clutch size between the two treatment group (GzLM $\beta = 0.15, se = 0.15, z63 = 0.98, p = 0.32$).

### 4.4.4 Joint effects of parasites and Hg

No significant interactions were detected when both treatment and Hg were considered together. During each treatment year, I resighted individual female eiders at similar rates regardless of which treatment they received or their concentration of blood Hg (GzLM $z_{218} = -1.33, p = 0.18$). Nesting propensity of female eiders did not vary with treatment type or blood Hg (GzLM $z_{218} = -1.57, p = 0.12$), and neither did clutch size (GzLM $z_{63} = -0.53, p = 0.59$). Similarly, resight rate in the year following treatment did not vary (GzLM $z_{218} = 0.19, p = 0.85$) when both parasite treatment and blood Hg concentrations were considered.

### 4.4.5 Nest attendance

Twenty-one and 13 loggers were collected from nest cups in 2013 and 2014, respectively. I found that the number of minutes until a hen returned to her nest after the loggers were deployed varied significantly with year (LM $F_{1,27} = 10.72, p = 0.003$), but not significantly with treatment, or blood Hg concentrations (LM all $p > 0.05$). I found no significant relationships in nest activity between treatment, or blood Hg concentrations (total number of mmd, total number of shuffles, average length of mmd, average number of mmd/day, average number of shuffles/day, cumulative time of mmd and total cumulative time; LM all $p > 0.05$). There was a significant year effect in total number of mmd (LM $F_{1,30} = 16.16, p < 0.001$), total number of shuffles (LM $F_{1,30} = 30.4, p <$
and average number of shuffles/day (LM $F_{1,30} = 21.12, p < 0.0001$), with 2013 nests showing lower levels of nest activity than 2014, suggesting that in 2014 the entire colony experienced higher levels of disturbance on the nest than in 2013.

4.4.6 Nest survival

I found strong temporal variation in daily nest survival (DNS) with higher DNS as nest age increased ($\beta_{\text{nest age}} = 0.19$, se $= 0.03$, Table 8), and a curvilinear seasonal pattern with lower DNS early and late in the breeding season ($\beta_{\text{time}} = 0.19$, se $= 0.09$, $\beta_{\text{time}^2} = -0.008$, se $= 0.002$). There was little difference in daily nest survival between the two years ($\beta = -0.10$, se $= 0.40$) with AICc 1.96 units higher than a model with only nest age and time of season. After accounting for influential temporal effects I did not find higher DNS of hens treated with PANACUR® ($\beta = 0.48$, se $= 0.35$, 95% CI: -0.204, 1.16) and the AICc of this model was 0.1 units higher than the reduced model with only age, year and time of season.

4.4.7 Anti-parasite treatment trial

In total 10 males captured between June 16th and 25th 2013 were used in the captive trial portion of the experiment (PANACUR® n = 5, Placebo n = 5). Four species of parasites were found in the gastrointestinal tracts of both treatment groups (Table 9). Although on average males treated with the anti-parasite medicine showed lower prevalence, intensity and abundance of most helminths, none of these relationships were significant (Table 9). While there was less change in the level of two species of helminths ($Corynosoma$ sp. and $Microsomacanthus$ sp.), I did observe that the two most abundant helminth species ($Lateriporus$ sp., $Profillicolus$ sp.) were reduced in males given the PANACUR® treatment (Table 9). There was also an overall reduction in all helminths in
the PANACUR® treated group. The power to detect any differences here was likely hindered by the small sample sizes.

4.5 Discussion

Understanding how different intrinsic and extrinsic environmental factors influence wildlife health and behaviour, either together or independently, is complex. This study is unique because it investigated how blood Hg burdens influence the reproduction and survival of birds whose gastrointestinal parasites burdens were naturally occurring or experimentally reduced in a free-living species. Both Hg and gastrointestinal parasites have the potential to negatively influence body condition of wildlife (Calvete et al., 2003; Wayland et al., 2001b), and thus could have consequences for reproduction and survival. Both Hg and parasites are increasing in wildlife from some Arctic regions (Davidson et al., 2011; Riget et al., 2011), so understanding how these two stressors may interact to affect wildlife health is important when predicting how ecosystems respond to environmental change.

While I found that Hg blood burdens in female eiders did not influence reproduction or return rates, I did find that experimentally altering the parasite loads of females influenced breeding propensity during the treatment year. Female eiders which arrived later in the season, or in poorer condition throughout the breeding season, appear to have benefited from receiving the anti-parasite treatment given upon their arrival. Normally, eider ducks that arrive on the colony late or in low condition are less likely to initiate nesting (Descamps et al., 2011a). Females receiving the placebo treatment in this study confirmed this pattern, with breeding propensity significantly decreasing with both arrival date, and for birds in lower condition. For birds that received the anti-parasite
treatment, however, I found that breeding propensity did not decline with arrival time, or condition. This suggests that birds arriving early and individuals in good condition are able to breed, regardless of their parasite burden, whereas birds that arrive late, or in poor condition benefit from experimentally reduced parasite burdens. The reduction in parasite burdens in females is perhaps enhancing their ability to acquire resources during the pre-breeding phase to initiate laying. Our findings illustrate that eiders are constrained by their arrival time or body condition to breed (Hennin et al., 2014; Love et al., 2010), and that these dynamics can be influenced by their gastrointestinal parasites.

I did not find an effect of parasite treatment on either nest survival or clutch size of eiders, similar to Hanssen et al. (2003) who also conducted an anti-parasite experiment on eider ducks. However, breeding constraints among birds in the Arctic may be particularly important given the very short summer, and that environmental conditions can be extremely variable (Hennin et al., 2013; Legagneux et al., 2013). Individuals that can gain energy rapidly during the pre-breeding season can successfully breed. In contrast, those that fail to rapidly gain resources during the short Arctic summer are at a disadvantage and often cannot undertake the costly task of breeding. Parasites are just one element of the phenotypic make-up of these female eiders. Females with higher levels of triglycerides, an indicator of how fast an individual can acquire fat reserves, have higher breeding success (Hennin et al., 2013). Our study demonstrates that gastrointestinal parasite burdens can negatively influence the ability of eiders to nest likely because they influence ability to retain energy during the critical pre-laying period, and should be considered when examining state-dependent breeding decisions.
Although Hanssen et al. (2003) found unsuccessful females in low condition benefited from the anti-parasite treatment in the year following treatment, I found that females in poor condition benefitted in the year of treatment. Importantly, these studies suggest that individuals which arrive late and/or in poor condition may be in need of energetic resources; something that can be limited by parasites. Therefore, when considering how changing parasite conditions under warming conditions in the Arctic may influence wildlife (Brooks and Hoberg, 2007; Kutz et al., 2005), it may be most important to consider how these burdens affect individuals not just on the margins of breeding success, but to consider how individuals may be susceptible to even minor changes. While I did find that treating females with the anti-parasite increased propensity in birds in low condition and which arrived later in the year, I found no difference in nest attendance or daily nest survival between the two treatments. This suggests that decisions to undertake a breeding attempt are made early in the year reflect an individual’s ability to see the breeding attempt through to the end. The strength and uniqueness of these findings stem from the fact that free-living wildlife were experimental manipulated in large sample sizes, and show deleterious effects that may be small in effect but significant to the reproductive outcomes.

Due to the complex nature of the potential interactions, few studies have been carried out in wild birds that aim to address questions relating to contaminants and parasitism and effects on avian reproduction or health. One study that did this examined a wild population of glaucous gulls, where males with naturally-occurring levels of both parasites and high levels of organochlorines had lower nest success as compared to males who experienced the same organochlorine levels, but were treated with anti-parasitic
drugs to reduce intestinal parasites (Bustnes et al., 2006). Similar to previous studies, I did not find any evidence that the experimental procedure negatively influenced individual breeding success, nor that breeding females treated with the anti-parasite treatment had higher reproductive success than non-treated females (Bustnes et al., 2006; Hanssen et al., 2003). In our study nest success seemed to be mainly driven by external factors, particularly polar bear (*Ursus maritimus*) predation (Iverson et al., 2014), which females cannot control regardless of their condition. Again, this consistent finding among several studies suggests that parasite burdens likely influence decisions to undertake a breeding attempt, but in the case of marine birds may not influence female condition which leads to success once the breeding attempt is underway.

While I could not determine the prevalence of parasitic infestation present in female eiders on their arrival to the breeding colony, nor that our treatment reduced these levels, I am confident that most eiders had some level of burden which was lowered by PANACUR® based on our experimental treatment of males. A study at this colony in the late 1990s found that 100% of the common eiders sampled (*n*=10) at the East Bay breeding colony were host to several species of gastrointestinal parasites (Wayland et al., 2001b). Similarly, recent studies of the parasites in the hunted birds from the nearby community of Cape Dorset found that 98% of eiders examined (*n*=185) over a two year period (2011/12) hosted gastrointestinal parasites (Provencher et al., 2016; Chapter 3). Furthermore, previous studies have shown that PANACUR® is effective in reducing nematodes, cestodes and acanthocephalans, the main gastrointestinal parasites carried by this population of eiders (Norton et al., 1991; Weber and Junge, 2000; Yazwinsky et al., 1993, 1992). Although our captive trial of males did not find that the PANACUR® was
100% effective at removing all parasite species and individuals, the total parasite load was reduced by about 50% in treated males. The lack of significant differences between the male groups may be attributable to insufficient time of the treatment to reduce the parasites significantly, or simply the small sample size.

Contrary to our predictions, I found no relationship between blood Hg levels and a female’s propensity to nest, clutch size, nest attendance, nor with return rate in the year following treatment. While eider ducks are benthic feeders that prey mostly upon bivalves and echinoderms, and have low levels of contaminants as compared with other Arctic marine birds (Mallory et al., 2004; Provencher et al., 2014b; Chapter 2), effects of Hg burdens on seabirds feeding at trophic levels slightly higher than eider have been found to have altered reproduction when Hg burdens were elevated (Fort et al., 2014). Moreover, the lack of significant relationships found between blood Hg and parasite treatment differed from other studies that found that contaminants and parasites jointly influenced breeding success (Bustnes et al., 2006). One explanation may be that this study focused on eiders, a species that has relatively low concentrations of Hg due to their low trophic level in the Arctic (Provencher et al., 2014b; Chapter 2), even though other studies have found reduced condition with higher Hg levels (Wayland et al., 2001b). Importantly, the levels of Hg as measured in circulating blood reflect Hg contamination acquired in the previous days, versus other tissues reflect longer term deposition which may influence breeding and survival more (Hobson and Clark, 1992b). Additionally, eider contaminant levels are known to increase over the breeding season as tissues are metabolized during incubation when females are fasting (Bustnes et al., 2010; Wayland et al., 2005), so potentially our early season measure of Hg was not indicative
of the Hg levels eider hens experienced throughout the breeding period which may have a
greater influence on breeding metrics. Another explanation may be that eiders may
actually benefit, at least in the short term, from consuming high levels of prey (and thus
Hg) during times of high energetic demand such as post-migration/pre-breeding season.

One important relationship I found was that eider blood Hg concentrations differed
substantially between the two sampling years (2013 and 2014). This inter-annual
difference in Hg concentrations was also found among shorebirds from East Bay
(McCloskey et al., 2013). Collectively these findings suggest that Hg uptake by organisms
occupying higher trophic levels (e.g., marine birds) in Arctic ecosystems can change
drastically from year to year. Since environmental Hg levels in the spring months can be
affected by local Hg depletion events (Zdanowicz et al., 2013), snow fall, and the duration
of spring melt these factors may all influence avian Hg concentrations during the pre-
breeding season when birds are acquiring energy.

4.6 Conclusions

Although both parasites and contaminants are commonly found in hosts, they
often have sub-lethal or non-detectable effects, and both factors may have varying
influence on the host. Therefore it follows that the effects of parasites and contaminants
on host condition may only become apparent during annual periods of energetic stress
each year. For example, contaminants may influence host condition during winter months
when lower temperatures lead to reduced foraging rates and increased thermoregulatory
costs (Lemly, 1993). Parasites may have a negative impact during a time of low energetic
reserves like migration (van de Crommenacker et al., 2012). Therefore it is important to
consider the effects of such burdens during times of energetic constraints such as
breeding. I considered the breeding period to be a key time during the annual cycle of the eider duck when effects of environmental factors might be most likely to manifest themselves, given that females already experience considerable breeding stress (Korschgen, 1977). There are other times when eider ducks and other wildlife may be under considerable stress that may result in exacerbated effects from contaminants and parasites, such as times of limited prey (Robertson and Gilchrist, 1998), or increased predation levels (Iverson et al., 2014). This study illustrates the importance of understanding how different environmental stressors affect wildlife not under average conditions, but marginal conditions as these may play a larger role in influencing wildlife populations.
Table 6. Eider duck (*Somateria molissima*) females treated and sampled in 2013 and 2014 as part of the anti-parasite experimental treatment manipulation. *ww* = wet weight for blood Hg samples. Body mass divided by total head length is used as the condition index.

<table>
<thead>
<tr>
<th>Year</th>
<th>PANACUR®</th>
<th>Placebo</th>
<th>Total</th>
<th>n</th>
<th>Blood sampling Hg (µg/g; ww)</th>
<th>n</th>
<th>Clutch size</th>
<th>n</th>
<th>Condition index</th>
</tr>
</thead>
<tbody>
<tr>
<td>2013</td>
<td>62</td>
<td>61</td>
<td>123</td>
<td>112</td>
<td>0.43 ± 0.06</td>
<td>29</td>
<td>2.4 ± 0.9</td>
<td>122</td>
<td>18.6 ± 1.7</td>
</tr>
<tr>
<td>2014</td>
<td>53</td>
<td>56</td>
<td>109</td>
<td>109</td>
<td>0.47 ± 0.06</td>
<td>35</td>
<td>3.1 ± 0.9</td>
<td>108</td>
<td>19.0 ± 1.7</td>
</tr>
</tbody>
</table>
Table 7. Number of banded female eider ducks (*Somateria molissima*) at the East Bay Island colony with annual re-sight and nesting rates.

<table>
<thead>
<tr>
<th>Year</th>
<th>Number of females banded</th>
<th>% re-sighted on colony in banding year</th>
<th>% nested in banding year</th>
</tr>
</thead>
<tbody>
<tr>
<td>2011</td>
<td>226</td>
<td>91</td>
<td>42</td>
</tr>
<tr>
<td>2012</td>
<td>77</td>
<td>91</td>
<td>44</td>
</tr>
<tr>
<td>2013</td>
<td>123</td>
<td>87</td>
<td>41</td>
</tr>
<tr>
<td>2014</td>
<td>109</td>
<td>71</td>
<td>45</td>
</tr>
<tr>
<td>2015</td>
<td>74</td>
<td>65</td>
<td>34</td>
</tr>
</tbody>
</table>
Table 8. AIC model results examining how nest survival varied with treatment in common eider ducks (*Somateria molissima*) in northern Hudson Bay, Canada. Treat = categorical parasite treatment (placebo or anti-parasite treatment given), year = experimental year (2013 or 2014), T = linear seasonal effect (lay date), T2 = quadratic seasonal effect (quadratic lay date), nestage = nest survival through breeding season.

<table>
<thead>
<tr>
<th>Model</th>
<th>AICc</th>
<th>ΔAICc</th>
<th>Akaike weights (wi)</th>
<th>Model Likelihood</th>
<th>Num. Par</th>
<th>Deviance</th>
</tr>
</thead>
<tbody>
<tr>
<td>{S(T2+nestage)}</td>
<td>314.2639</td>
<td>0</td>
<td>0.37404</td>
<td>1</td>
<td>4</td>
<td>306.2254</td>
</tr>
<tr>
<td>{S(treat+T2+nestage)}</td>
<td>314.3627</td>
<td>0.0988</td>
<td>0.35602</td>
<td>0.9518</td>
<td>5</td>
<td>304.3049</td>
</tr>
<tr>
<td>{S(year+T2+nestage)}</td>
<td>316.2229</td>
<td>1.959</td>
<td>0.14045</td>
<td>0.3755</td>
<td>5</td>
<td>306.165</td>
</tr>
<tr>
<td>{S(treat+year+T2+nestage)}</td>
<td>316.3855</td>
<td>2.1216</td>
<td>0.12949</td>
<td>0.3462</td>
<td>6</td>
<td>304.3044</td>
</tr>
<tr>
<td>{S(treat+year+T2)}</td>
<td>346.4682</td>
<td>32.2043</td>
<td>0</td>
<td>0</td>
<td>4</td>
<td>336.4103</td>
</tr>
<tr>
<td>{S(treat+T2)}</td>
<td>347.3245</td>
<td>33.0606</td>
<td>0</td>
<td>0</td>
<td>5</td>
<td>339.286</td>
</tr>
<tr>
<td>{S(treat+year)}</td>
<td>352.9101</td>
<td>38.6462</td>
<td>0</td>
<td>0</td>
<td>3</td>
<td>346.887</td>
</tr>
<tr>
<td>{S(.)}</td>
<td>354.2823</td>
<td>40.0184</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>352.2784</td>
</tr>
<tr>
<td>{S(treat+year+T)}</td>
<td>354.7922</td>
<td>40.5283</td>
<td>0</td>
<td>0</td>
<td>4</td>
<td>346.7537</td>
</tr>
<tr>
<td>{S(treat)}</td>
<td>356.2884</td>
<td>42.0245</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>352.2769</td>
</tr>
</tbody>
</table>
Table 9. Helminth infection levels in captive male eider ducks (*Somateria molissima*) treated with the anti-parasite PANACUR® and a placebo treatment, and euthanized after a 24 hour captive period.

Prevalence (using unconditional exact test), mean intensity (bootstrap-t test), median intensity (Mood’s median test), and mean abundance (bootstrap-t test) with *p*-values given for each test (na represents where statistical tests could not be completed due to low sample sizes).

<table>
<thead>
<tr>
<th>Parasite</th>
<th>PANACUR®</th>
<th>Placebo</th>
<th>Associated p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n = 5</td>
<td>n = 5</td>
<td></td>
</tr>
<tr>
<td><em>Lateriporus sp.</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>prevalence</td>
<td>20</td>
<td>60</td>
<td>0.30</td>
</tr>
<tr>
<td>mean intensity</td>
<td>13 (na)</td>
<td>32.7 (29)</td>
<td>na</td>
</tr>
<tr>
<td>median intensity</td>
<td>13</td>
<td>38</td>
<td>1.00</td>
</tr>
<tr>
<td>mean abundance</td>
<td>2.6 (5.8)</td>
<td>19.6 (27.4)</td>
<td>na</td>
</tr>
<tr>
<td><em>Profillicolus sp.</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>prevalence</td>
<td>40</td>
<td>100</td>
<td>0.11</td>
</tr>
<tr>
<td>mean intensity</td>
<td>32.5 (40)</td>
<td>16.8 (10.9)</td>
<td>na</td>
</tr>
<tr>
<td>median intensity</td>
<td>32.5</td>
<td>20</td>
<td>1.00</td>
</tr>
<tr>
<td>mean abundance</td>
<td>13 (26.9)</td>
<td>16.8 (10.9)</td>
<td>0.76</td>
</tr>
<tr>
<td><em>Corynosoma sp.</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>prevalence</td>
<td>60</td>
<td>60</td>
<td>1</td>
</tr>
<tr>
<td>mean intensity</td>
<td>5.3 (4.9)</td>
<td>1.7 (0.6)</td>
<td>na</td>
</tr>
<tr>
<td>median intensity</td>
<td>3</td>
<td>2</td>
<td>0.33</td>
</tr>
<tr>
<td>mean abundance</td>
<td>3.2 (4.6)</td>
<td>1 (1.0)</td>
<td>0.40</td>
</tr>
<tr>
<td><em>Microsomacanthus sp.</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>prevalence</td>
<td>40</td>
<td>40</td>
<td>1.00</td>
</tr>
<tr>
<td>All helminths</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>prevalence</td>
<td>80</td>
<td>100</td>
<td>0.30</td>
</tr>
<tr>
<td>mean intensity</td>
<td>23.5 (26.4)</td>
<td>37.4 (37.8)</td>
<td>0.53</td>
</tr>
<tr>
<td>median intensity</td>
<td>12</td>
<td>23</td>
<td>0.48</td>
</tr>
<tr>
<td>mean abundance</td>
<td>18.8 (25.2)</td>
<td>37.4 (31.8)</td>
<td>0.38</td>
</tr>
</tbody>
</table>
Figure 6. A – arrival date (presented in ordinal date with January 1\textsuperscript{st} = 1) of female common eider ducks (\textit{Somateria mollissma}) versus their breeding propensity in placebo treated birds; B - arrival date (presented in Julian date with January 1\textsuperscript{st} = 1) of females versus their breeding propensity in females given the anti-parasite treatment, PANACUR®; C – condition index (mass/total head length) of females in relation to their breeding propensity in placebo treated birds; and D – condition index versus breeding propensity in birds treated with the anti-parasite. Red line on each graph indicates the zero value for breeding propensity, indicating where breeding propensity changes between positive and negative. ★ indicate where a significant difference was found in the breeding propensity between the two treatments.
5 Chapter: Implications of mercury and lead concentrations on breeding physiology and phenology in an Arctic sea duck

5.1 Abstract

Although both physiological traits and phenology are thought to be evolved traits, they often show marked variation within populations, which may be related to extrinsic factors. For example, trace elements such as Hg and lead (Pb) alter biochemical processes within wildlife that may affect migration and breeding. Although there is a growing understanding of how trace elements and other contaminants may influence wildlife physiology, studies addressing these interactions in free-living species are still limited. In this study, I examined how two non-essential trace elements of concern (Hg and Pb) interacted with physiological and breeding measures known to influence breeding in a free-living population of common eider ducks. I collected blood from female eiders as they arrived at a breeding colony in northern Hudson Bay. Blood samples were subsequently assessed for baseline corticosterone (CORT), immunoglobulin Y (IgY), Hg, and Pb. I used path analysis to assess how Hg and Pb varied with arrival date and arrival condition and pre-breeding measures of IgY and CORT also suspected of influencing breeding. I found that arrival date, blood Hg and IgY showed significant inter-annual variation. While blood Pb concentrations were quite low, blood Pb levels significantly increased with later arrival date of the birds, and varied negatively with eider body condition, suggesting that even at low blood concentrations, Pb may be related to lower investment in reproduction in eiders. In contrast to our predictions, blood Hg concentrations were positively correlated with eider body condition, indicating that fatter birds also had higher Hg burdens. Overall, our results suggest that although blood Hg and
Pb concentrations were below no-effect levels, these low level concentrations of known toxic metals show significant relationships with breeding onset and condition in female eider ducks, factors that could influence reproductive success in this species.

5.2 Introduction

There is increasing interest in determining how measures of immunity and stress relate to survival and reproduction (Hennin et al., 2012; Humborstad et al., 2016; Madeira et al., 2016). Since there is an expectation that environmental contaminants may influence the condition and reproductive success of an individual directly or indirectly by altering physiological traits (Blévin et al., 2014; Tartu et al., 2015), there is a need for integration of toxicology into this expanding field. Mercury is of particular concern for marine birds in the Arctic, due to high levels of this trace element in some marine bird species (Provencher et al., 2014b; Riget et al., 2011). Importantly, Hg has been shown to be an endocrine disruptor in both captive (Jayasena et al., 2011) and wild birds (Tartu et al., 2016, 2013), even when exposure is low level. In addition to Hg, marine birds are also exposed to other trace elements that are known to be toxic. For example, marine birds are exposed to lead (Pb) through shot from hunting activities, which can also have negative impacts (Merkel et al., 2006; Scheuhammer, 2009), making it is important to consider trace metals in relation to each other to investigate possible interactions (Sarigiannis and Hansen, 2012). As a result, research exploring the links between physiological traits, contaminants, survival and reproduction are needed to better understand how individuals, and populations, may respond to changes in predation and disease, among other ecological interactions, in sensitive environments (Guindre-Parker et al., 2013; Harms et al., 2015; Hennin et al., 2016).
The impacts of Hg and Pb may be linked directly to behaviours associated with breeding. For example, female mallard ducks (*Anas platyrhynchos*) with higher Hg exposure laid fewer eggs and produced fewer ducklings compared with hens with lower Hg exposure (Heinz, 1979). Importantly, Hg and Pb poisoning may also cause changes in attributes that have negative subsequent effects on reproduction, such as reduced adult health and body condition. Increased levels of Pb in mallard ducks in the Mediterranean were correlated with increased humoral immune response and negatively correlated with cellular immune response (Vallverdú-Coll et al., 2015). In free-living Arctic black-legged kittiwakes higher levels of Hg have been linked with decreased levels of prolactin, an important hormone in reproduction (Tartu et al., 2016). Thus, even chronic moderate exposure to some trace elements can potentially impact physiological traits that have been linked with individual condition and fitness. While there are an increased number of studies are demonstrating the links between contaminants and physiological traits, few studies have examined whether potential deleterious, synergistic effects of trace elements and physiological states can influence condition, reproduction and survival (although see Tartu et al., 2013).

The overall objective of this study was to investigate two biologically-important trace metals (Hg and Pb) in relation to the health and condition of a free-living bird, the common eider duck. Long-term monitoring of populations in Canada have yielded a wealth of information about eider contaminants, physiology and breeding metrics (Hennin et al., 2013; Love et al., 2010; Mallory and Braune, 2012). Importantly, in eider ducks both large clutch sizes and earlier laying are associated with higher nest success (Descamps et al., 2011a). Thus, eider ducks are an excellent species to investigate how
trace elements interact with physiological parameters and their subsequent relations to breeding parameters.

To examine how Hg and Pb vary with physiological traits that have been shown to influence reproductive onset and success (IgY, CORT, arrival date and arrival condition), I used a path analytic approach that assessed both the direct and indirect relationships among these variables (Shipley, 2009). In assessing candidate paths for our model, I relied on what has been demonstrated in previous studies. As Hg and Pb are transferred trophically (Franson and Pain, 2011; Shore et al., 2011), I predicted by analogy that both elements may show inter-annual variation as has been described in other avian species in the region (McCloskey et al., 2013), due to differing environmental deposition each year (Kirk et al., 2012). I also expected that arrival date and arrival condition of female eiders would show inter-annual variation between the two sampling years, since these metrics are influenced by a variety of environmental conditions that often differ annually (Descamps et al., 2011a; Love et al., 2010). I expected inter-annual variation in IgY as found in other northern eider species (Counihan et al., 2015), and inter-annual variation in CORT as several species of marine birds have highly variable CORT levels from year to year (Ninnes et al., 2011; Satterthwaite et al., 2012). I predicted that Hg would show a negative relationship with IgY, as Hg has been associated with depressed immune responses (Lewis et al., 2013a). Conversely, I predicted that Pb would be positively correlated with IgY, as Pb poisoning can lead to increases in humoral immune responses (Vallverdú-Coll et al., 2015). I also expected that Hg and Pb would be negatively related to body condition, as higher concentrations of these metals are related to lower body condition in other bird species (Vallverdú-Coll et
I also included paths between IgY, arrival date, arrival condition and CORT to assess how Hg and Pb may influence CORT indirectly. I used IgY as a general indicator of health and immune status in the arriving eider ducks (Apanius and Nisbet, 2006; Counihan et al., 2015). Based on this framework, I predicted that IgY would be negatively correlated with arrival condition, with birds in better condition arriving with lower IgY levels as compared with birds with poorer condition (Bourgeon et al., 2010). I also expected IgY to be negatively correlated with arrival date, based on work by Counihan (2015), demonstrating that reproductive timing and IgY can be negatively correlated in eider ducks. I measured baseline CORT levels in this study, using it as a metric of general physiological condition; higher CORT levels relate to greater energetic demand (Hennin et al., 2016, 2015). Based on this interpretation, I predicted that arrival condition would be positively correlated with CORT, with birds arriving in better condition having higher baseline CORT levels (Hennin et al., 2014). I also expected arrival date to positively correlate with CORT, with birds arriving earlier in the year having lower CORT levels, and those arriving later, and thus closer to breeding, having higher baseline CORT levels (Hennin et al., 2014). A summary of our predictions upon which our framework was based is provided in Figure 7.
5.3 Methods

5.3.1 Capture

Female eider ducks were caught at the breeding colony on East Bay Island in northern Hudson Bay in 2013 (n= 98) and 2014 (n=92; Table 10) as part of the experimental experiment presented in Chapter 4 above. For more details on capture see Methods Chapter 4.

5.3.2 Blood sampling

In addition to the sample used to assess blood trace elements (see Chapter 4 methods above for more details) a small blood sample (maximum 1ml) was taken immediately after capture at the net and within 3 min of the birds hitting the net using a 23G thin wall, 1-inch (c.25-mm) needle attached to a heparinized 1-mL syringe. This sample was used to assess baseline CORT (Hennin et al., 2014; Romero and Reed, 2005; Wingfield et al., 1982) and immunoglobulin Y (IgY; Legagneux et al., 2014). All blood samples were kept at 4°C and centrifuged at 10,000 rpm for 10 min within 6h of collection. Upon separation, the plasma component of the blood collected within 3 mins was collected and stored at −20°C for further analysis.

5.3.3 Trace element analysis

Analyses were conducted by RPC Laboratories for total Hg and Pb (Table 11; also shows detection limits). The same analytical methods were used for this study as in Chapter 4 above, with the inclusion of Pb in the analysis. All trace element concentrations are presented in wet weight μg/g, unless otherwise indicated for comparison purposes (Table 11).
5.3.4 Physiological assays

Baseline plasma CORT was analysed using an enzyme-linked immunoassay (EIA; Assay Designs, Ann Arbor, MI, USA), previously validated in common eiders (Hennin et al., 2015). Samples were run in triplicate at a 1:20 dilution with 1.5 % of kit-provided steroid displacement buffer (Hennin et al., 2015). Plates were run with a kit-provided standard curve by serially diluting a 200,000 pg mL\(^{-1}\) CORT standard and a control of laying hen plasma (Sigma-Aldrich Canada, Oakville, ON, Canada). Assay plates were read on a spectrophotometer plate reader at 405 nM and the mean inter- and intra-assay coefficients of variation across all plates were 8.17 and 7.99 %, respectively. All CORT samples were run in the year of sampling, and are presented in pg/mm.

The level of plasma IgY was used as an indicator of overall humoral adaptive immune function (Bourgeon et al., 2009, 2006). A sensitive, in-house, ELISA (enzyme-linked immunosorbent assay) method previously validated in common eiders (Bourgeon et al., 2006) was used to determine the amount of total IgY in eider plasma (diluted to 1/32,000 in carbonate–bicarbonate buffer; Bourgeon et al., 2006). The mean intra-assay coefficient of variation across all plates was 3.08%. All IgY levels are presented in absorbance units.

5.3.5 Statistical approaches

Levels of the two trace elements (Hg and Pb) were examined in female eider blood. The arithmetic means with standard deviation (SD) and the geometric means with 95% confidence intervals along with the range of values are presented (Table 11). When values of less than the detection limit occurred, values were set at 10% below the detection limit to complete the multivariate analysis and geometric mean calculations.
Levene's test was used to confirm homogeneity of variances and the Shapiro-Wilk test was used to confirm data normality. Data were log (Pb and CORT) or square –root (Hg) transformed to approach the assumptions for parametric multivariate analysis where necessary. I used body mass divided by the total head length as an index of body condition, as this index suitably scores condition adjusted for structural size in this species (Descamps et al., 2011a). Since samples were collected over two breeding seasons, I also included sampling year as a predictor variable. I used a path analytic approach (Shipley, 2009, 2000) to explore the relationships between trace elements (Hg and Pb) and the physiological variables (CORT, IgY), in relation to arrival date and body condition. The benefits of using a path analytic approach applied here follow the benefits as outlined in Chapter 3 above. Briefly, this method was also chosen because it allows us to conceptualize and evaluate both direct and indirect affects that Hg and Pb may have on other variables known to be associated with breeding in eider ducks. All statistical tests were evaluated at $\alpha = 0.05$ and means are presented $\pm$ SD.

The global path model was constructed based on previous knowledge of how trace elements, arrival date, condition metrics and physiological parameters are believed to interact (Fig. 7). The fit of the model was evaluated through directed separation (d-sep) tests (Shipley, 2009). See Chapter 3 above for more details regarding the application of the path analytic approach to global model testing. Upon acceptance of the final global model each path was fit as appropriate to the data. To calculate relative path coefficients for each path (i.e., standardized partial regression coefficients), each variable (V) was standardized using z-score scaling, except for paths that related to year. All year paths were scored using $d$ statistics which allow for better estimates when a predictor variable
is categorical (Nakagawa and Cuthill, 2007). Total effect sizes were calculated by summing the direct effect and the products of the indirect effect sizes (Shipley, 2000).

5.4 Results

5.4.1 Model Fit

In 2013 all birds \((n = 98)\) were caught between June 15\(^{th}\) and July 3\(^{rd}\), and in 2014 \((n = 92)\) birds were caught between June 11\(^{th}\) and July 1\(^{st}\). Data from these female eider ducks were used to test and parameterize our path model (Fig 7). Our final hypothesized model was consistent with the correlational structure of the data (Fisher’s C statistic = 5.01, \(df = 6, p = 0.54\)), suggesting the global model constructed significantly fit the data. I found that Hg and Pb varied significantly with several variables in the hypothesized model, and while some relationships followed our predictions, not all relationships did (Fig. 8).

5.4.2 Trace element concentrations

A total of 154 female eiders (or 81%) had blood Pb concentrations above the detection level, with concentrations on average below the normal range reported for waterfowl (0.02-0.50 ppm ww; Puls, 1994; Table 12). All females had blood Hg concentrations above the detection level, with concentrations within normal ranges reported for waterfowl and seabirds, and below values reported to be associated with impaired reproduction (Evers et al., 2004; Puls, 1994; Shore et al., 2011). Female eider ducks had similar Hg blood concentrations to other reports for this species in North America (Meattey et al., 2014), including previous studies at East Bay Island (Wayland et al., 2001b; Table 12). While I found that blood Pb showed no significant inter-annual variation (GLM \(F_{1,188}= 0.16, p = 0.68\); Table 11; Fig. 8), blood Hg concentrations showed
a significant difference between the two sampling years (GLM $F_{1,188} = 17.8, p < 0.0001$; Table 11; Fig.8).

5.4.3 Variables related with IgY

IgY was measured in the blood of all females examined, and levels were significantly higher in females in 2013 compared with 2014 (GLM $F_{3,186} = 14.56, p < 0.0001$; Table 10; Fig 8). Contrary to our predictions, I found that IgY did not vary significantly with blood concentrations of either Hg or Pb (Hg - GLM $F_{3,186} = 14.56, p < 0.30$; Pb - GLM $F_{3,186} = 14.56, p = 0.24$; Fig 8).

5.4.4 Variables related to arrival condition

Mean (SD) female mass was 2171 ± 185 g, with no difference in arrival condition between the two sampling years (GLM $F_{4,185} = 8.97, p = 0.79$; Table 10; Fig. 8). Contrary to our predictions, I found no relationship between IgY and arrival condition (GLM $F_{4,185} = 8.97, p = 0.15$; Fig 8). Interestingly, both Hg and Pb varied significantly with arrival condition, but in opposite directions. Female eiders with higher blood Pb concentrations had lower arrival condition (GLM $F_{4,185} = 8.97, p < 0.0001$; Fig. 8). Female eiders with higher blood Hg concentrations had higher arrival condition (GLM $F_{4,185} = 8.97, p = 0.04$; Fig 8), which was contrary to our prediction.

5.4.5 Relationships with arrival date

Median arrival date of female eiders was earlier in 2013 than 2014 (GLM $F_{4,185} = 8.97, p = 0.04$), although the effect was small (Fig. 8). Concentrations of Hg (GLM $F_{4,185} = 8.97, p = 0.88$) or IgY (GLM $F_{4,185} = 8.97, p = 0.82$) were not related to female arrival date. However, there was a positive correlation between blood Pb levels and
arrival date of female eiders (GLM $F_{4,185} = 8.97, p > 0.0001$), with eiders arriving later in the season having higher blood Pb concentrations (Fig. 8).

Variables related with Baseline CORT

Baseline CORT was detected in all 190 female eiders sampled (Table 10), and females sampled in 2014 had higher CORT than those caught in 2013 (GLM $F_{6,183} = 3.37, p = 0.03$; Table 2; Fig. 8), although the effect size was small (Fig. 8). While no significant relationship between Pb and CORT was detected (GLM $F_{6,183} = 3.37, p = 0.64$), there was a trend for females with higher Hg to have lower CORT (GLM $F_{6,183} = 3.37, p = 0.05$; Table 10; Fig. 8). Additionally, female eiders with higher CORT arrived at the colony later (GLM $F_{6,183} = 3.37, p = 0.02$) and in better body condition (GLM $F_{6,183} = 3.37, p = 0.04$, Fig. 8).

5.5 Discussion

The purpose of this study was to examine how Hg and Pb concentrations are directly or indirectly related to physiology, condition and reproduction in a free-living Arctic bird. Overall our findings suggest that blood concentrations of Hg and Pb in breeding female eider ducks in northern Hudson Bay are below reported levels associated with toxic effects (Puls, 1994), and similar or lower to what has been reported for this species at this location (Mallory et al., 2014; Wayland et al., 2001b), as well as in other regions (Franson et al., 2004, 2000; Meattey et al., 2014; Table 10). While I found that generally blood concentrations of Pb and Hg were low, I did find that these blood concentrations significantly varied with several parameters known to influence breeding success in the species.
5.5.1 Trace Elements and Corticosterone

I detected a significant relationship between Hg and baseline levels of CORT. Our findings support the growing body of literature that suggests that Hg may disrupt endocrine capabilities in birds (Herring et al., 2012; Pollock and Machin, 2008; Tartu et al., 2016, 2013). While the size of the direct relationship between Hg and CORT was small (-0.14), the total indirect effect of year and Hg on CORT was more than five times greater (-0.75). This suggests that interannual variation in eider Hg blood concentrations had significant effects on CORT, and likely contributes variation in endocrine system outputs in eider ducks. This is important as baseline CORT, among other physiological parameters dependent on endocrine systems, strongly influences reproductive success in eiders (Hennin et al., 2016, 2014) and other species (Lattin et al., 2016; Love et al., 2005).

Our findings should be interpreted within the larger rubric of endocrine biology when applying them to even relatively simple ecological systems. I chose to construct our hypothesized global path model with CORT downstream of arrival date and condition, but there are other interpretations of this complex and interrelated system. I recognize that CORT likely influences condition both before individuals arrive, and afterwards (e.g. Lovvorn et al., 2012). While altering the position of CORT in the model in relation to arrival date and condition alters the effect size slightly, the overall significance of the global model is not affected because the number and identity of the individual paths are not altered.

Interestingly, while our results show that Hg is related to lower levels of CORT (direct effect size - 0.14; Fig. 8), I also found that Hg is positively, but indirectly
associated with CORT via arrival condition, with an indirect effect size that is twice as large as the direct relationship (0.30; Fig. 8). This suggests that although Hg and CORT may be directly negatively correlated, the drive to increase in condition via increased foraging (and thus uptake of Hg), after arrival and in preparation for reproduction may have a larger positive effect on CORT secretion than the associated Hg that comes with it. This demonstrates how relationships between condition and trophically transferred factors (i.e. contaminants, parasites) can be complex and should be considered simultaneously in terms of their relations with endocrine levels that affect resource acquisition (Bulté et al., 2012; Marcogliese and Pietrock, 2011).

I note that our sample included females that were at different physiological stages within the pre-breeding period. Recent work has demonstrated that female eiders that are captured and sampled during the rapid follicle growth period physiologically differ from females that are still in the pre-recruitment phase of breeding (Hennin et al., 2015). Thus, it is likely that any influence both Hg and Pb have on physiological parameters could affect these two groups differently, and thus breeding outcomes. While I recognize breeding stage as important, I was unable to test for these differences in a rigorous way due to limited sample size. Future work should take breeding stage into consideration to explore more fully how contaminants may differentially affect wildlife throughout their annual cycle.

5.5.2 Trace Elements and Immune Function

Although trace elements, such as Hg, are often associated with immunosuppressive effects in wildlife even at sublethal levels (Fallacara et al., 2011; Kenow et al., 2007; Lewis et al., 2013b), I found no evidence of a relationship between
IgY levels and either Hg or Pb concentrations. Similar to our findings, a study examining immunity metrics in great skuas (Stercorarius skua) nesting in the Shetland Islands, Iceland and Bjørnøya also found no significant relationship between IgY and organochlorines or PBDEs (Bourgeon et al., 2012). While several captive studies have shown negative effects of Hg on immune metrics (Lewis et al., 2013a) they often use a suite of immune metrics to determine systemic responses within individuals. To date, studies of free-living species using a wide selection of immune metrics are few. I found that IgY did not vary with arrival date or condition even though IgY showed significant interannual variation. This suggests that general immune status may not be a large driver of timing of and condition at arrival on the breeding grounds in female eiders. Further, interannual IgY variation has been showed in other studies (Counihan et al., 2015; Hegemann et al., 2012; Staszewski et al., 2007), but there is little consensus on what causes this variation. While immunity likely plays some role in reproduction, IgY on its own may not be specific enough to detect any subtle differences in relation to contaminants and breeding metrics. To investigate further how Hg may influence northern birds’ immunity, a broad suite of immune metrics including immune responses to novel immunological challenges may be required.

### 5.5.3 Trace elements, timing and condition

While Pb levels in females eiders did not vary with IgY or CORT, later arriving birds and birds in poorer condition did have higher blood Pb concentrations. Typically, eiders arriving on the breeding colonies earlier often perform better reproductively (Descamps et al., 2011a), and thus our findings suggest that eider ducks with higher Pb levels would likely have had lower reproductive success. Interestingly, the effect of Pb on
arrival condition was the largest effect detected in this study (-1.52; Fig. 8), indicating that this relationship warrants further consideration to understand the underlying causes and mechanisms. Taken together, these two findings suggest that while blood Pb levels may be very low in eider ducks in the Canadian Arctic in June, there are likely sublethal effects of Pb on eider reproduction. While Pb is thought to be both trophically transferred and originating from point sources of Pb in the environment (Finkelstein et al., 2003), high Pb concentrations in waterfowl have also been linked with embedded Pb shot (Johansen et al., 2001; Merkel et al., 2006; Sanderson et al., 1998). Although Pb shot was banned for shooting waterfowl in North America since 1999, Pb shot is still used in Greenland where many eider ducks from Arctic Canada spend the non-breeding season (Mosbech et al., 2006). Migratory birds may also consume spent shot in the environment that is still used for hunting non-migratory bird species, mistaking it for grit, which is commonly ingested (Schummer et al., 2011). Additionally, waterfowl have been reported in ingest Pb fishing weights, another source of Pb in the aquatic environments (Schummer et al., 2011).

5.6 Conclusions

Regardless of the source, our results suggest wild birds may be susceptible to deleterious effects from Pb even at blood levels below toxic concentrations. This work highlights how research interesting in exploring the effects of contaminants must consider both the direct and indirect effects on wildlife to get an accurate picture of how wildlife may be affected. This work also highlights the need to consider the cumulative effects of contaminants on individual traits in wildlife. I found that both Hg and Pb significantly varied with CORT, directly and indirectly, illustrating that while some
contaminants can alter traits within individuals linked with health and reproduction it may not be easily detectable when the relationships are indirect. Thus, research that utilized a variety of tools and approaches (i.e. biochemical, breeding colony observations, endocrine metrics, contaminants, phenology etc.) to on wild populations are best equipped to further inform how environmental contaminants affect biota.
Table 10. Biometric values for female common eider ducks (*Somateria molissima, n = 193*) caught during the pre-breeding season (June to July) in 2013 and 2014 at East Bay Island, in northern Hudson Bay. Condition index is the total mass of the individual/total head length.

<table>
<thead>
<tr>
<th></th>
<th>Total n</th>
<th>Corticosterone (pg/mm)</th>
<th>Immunoglobulin Y (absorbance units)</th>
<th>Condition Index</th>
</tr>
</thead>
<tbody>
<tr>
<td>2013</td>
<td>98</td>
<td>9.93 ± 14.09</td>
<td>0.73 ± 0.17</td>
<td>7.71 ± 0.70</td>
</tr>
<tr>
<td>2014</td>
<td>92</td>
<td>11.98 ± 12.70</td>
<td>0.59 ± 0.15</td>
<td>7.78 ± 0.64</td>
</tr>
<tr>
<td>Overall</td>
<td>190</td>
<td>10.94 ± 13.43</td>
<td>0.66 ± 0.18</td>
<td>7.75 ± 0.69</td>
</tr>
</tbody>
</table>
Table 11. Trace element concentrations in female common eider ducks (*Somateria molissima*) at a breeding colony in northern Hudson Bay (*n* = 193). Arithmetic mean with standard deviation (SD), geometric mean with 95% confidence intervals (CI), minimum detected values and maximum detected values are given along with the detection limits for each element. All concentrations given in µg/g wet weights (ww).

<table>
<thead>
<tr>
<th>Element</th>
<th>Detection limit dw (µg/g)</th>
<th>Mean (SD)</th>
<th>Geometric mean (95% CI) (µg/g)</th>
<th>Minimum, Maximum values (µg/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lead (Pb)</td>
<td>0.005</td>
<td>0.009 (0.006)</td>
<td>0.008 (0.001)</td>
<td>0.005, 0.043</td>
</tr>
<tr>
<td>Overall</td>
<td>2013</td>
<td>0.009 (0.006)</td>
<td>0.008 (0.001)</td>
<td>0.0045, 0.043</td>
</tr>
<tr>
<td>2014</td>
<td>0.009 (0.006)</td>
<td>0.008 (0.001)</td>
<td>0.0045, 0.039</td>
<td></td>
</tr>
<tr>
<td>Mercury (Hg)</td>
<td>0.01</td>
<td>0.21 (0.06)</td>
<td>0.20 (0.008)</td>
<td>0.08, 0.43</td>
</tr>
<tr>
<td>Overall</td>
<td>2013</td>
<td>0.19 (0.05)</td>
<td>0.18 (0.010)</td>
<td>0.08, 0.34</td>
</tr>
<tr>
<td>2014</td>
<td>0.22 (0.06)</td>
<td>0.21 (0.012)</td>
<td>0.09, 0.43</td>
<td></td>
</tr>
</tbody>
</table>
Table 12. Blood concentrations of trace elements in common eiders (*Somateria molissima*) from published studies. *Indicates the average reported value in the paper, AM = arithmetic mean, GM = geometric mean, dw = dry weight, ww = wet weight. All values are presented in µg/g.

<table>
<thead>
<tr>
<th>Study</th>
<th>Location</th>
<th>Year</th>
<th>Hg</th>
<th>Pb</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>AM</td>
<td>GM</td>
</tr>
<tr>
<td>Franson et al. 2000</td>
<td>Finland</td>
<td>1997</td>
<td>0.31ww</td>
<td>0.084*ww</td>
</tr>
<tr>
<td>Wayland et al. 2001</td>
<td>Hudson Bay</td>
<td>1997/98</td>
<td>0.23*ww</td>
<td>0.37ww</td>
</tr>
<tr>
<td>Franson et al. 2004</td>
<td>Beaufort Sea</td>
<td>2000</td>
<td>0.80*dw</td>
<td>1.19dw</td>
</tr>
<tr>
<td>Goodale et al. 2008</td>
<td>Gulf of Maine</td>
<td>2001-06</td>
<td>0.11ww</td>
<td>0.20ww</td>
</tr>
<tr>
<td>Bond and Diamond 2009</td>
<td>New Brunswick</td>
<td>2005/06</td>
<td>0.051ww</td>
<td></td>
</tr>
<tr>
<td>Meattey et al. 2014</td>
<td>New England</td>
<td>2013</td>
<td>0.293*ww</td>
<td>1.75ww</td>
</tr>
<tr>
<td>This study</td>
<td>Hudson Bay</td>
<td>2013/14</td>
<td>0.21ww</td>
<td>0.20ww</td>
</tr>
</tbody>
</table>
Figure 7. Hypothesized global path model between two trace metals (mercury and lead) and known breeding precursors in eider ducks (*Somateria molissima*; year, arrival date, arrival condition, immunoglobulin Y and corticosterone) over two years of sampling (2013 and 2014). + signs indicate a predicted positive relationship between the variables and - indicates a predicted negative relationship. ✓ indicates were an inter-annual differences was expected.
Figure 8. Simplified path model showing the significant relationships between two trace metals (mercury and lead) and known breeding precursors in eider ducks (*Somateria molissima*; year, arrival date, arrival condition, immunoglobulin Y and corticosterone) over two years of sampling (2013 and 2014; $n = 165$). Only significant paths are shown ($p < 0.05$). Standardized path coefficients are given for each path, with arrow size scaled to illustrate relative effect sizes.
6 Chapter: General Discussion

Wildlife populations and ecosystems are facing numerous challenges including changing climatic conditions, emerging diseases, habitat destruction and introduced predators. Some of these are acute and often receive more attention, while others are more subtle and difficult to integrate into our understanding of wildlife ecology. While contaminants in general have received a lot of attention globally, there is still much we do not understand about how northern wildlife are exposed to, and affected by, contaminants that originate from distant sources.

In this thesis I aimed to increase our understanding of how Hg contamination can influence marine bird reproduction and survival through the use of eider ducks as a model species. My work focused on addressing key gaps in our understanding of how the patterns and distribution of Hg in marine birds, and how Hg interacts with other environmental burdens to affect marine birds. I strived to achieve this through a combination of field and laboratory work that utilized samples collected from hunters, and existing long-term research programs. The research presented in this thesis is novel and informative as it considers Hg in a wild bird species allowing the assessment of effects to occur using naturally varying levels. My work is fundamental in its scope because I worked with communities and team members to achieve large sample sizes that would allow rigorous analytical approaches to be applied to the ecological questions of interest. As both contaminant and parasite research is often limited by sample sizes, this component adds strength to my findings. Below I discuss the results of each thesis chapter, how each informs our greater understanding of Hg as a contaminant in marine birds, and research questions that warrant more study.
I first set out to synthesis and summarize the state of knowledge of Hg contaminant studies in northern marine birds in Canada. Numerous studies have investigated Hg in this group, but the literature lacked an overall view of the research to date that could be used to outline research gaps and directions for the future work. In Chapter 2, I present a synthesis paper where I explore the available data for Hg concentrations in eight northern marine birds, and how these levels relate to known effect levels. Importantly, this review allowed me to discuss and explore what external factors likely influence Hg concentrations in northern marine birds, including poorly understood interactions with other environmental factors and knowledge gaps which helped to guide the rest of my thesis. I also outline areas of future research for both the Canadian Arctic and for wildlife Hg research in general, namely assessing interannual variation in Hg concentrations, and effects in northern bird populations. Beyond my own thesis work, I also identify research priorities that should be undertaken to increase our understanding of how Hg can affect wildlife.

Following my findings in Chapter 2 I set out to explore the distribution of Hg concentrations in my model species, the northern common eider duck (Chapter 3). I examine Hg concentrations in breast muscle alongside two parasite species within a path analysis framework to investigate shared pathways between Hg and parasites in eider ducks, and how other ecological factors. I was able to take this statistical approach due to the large sample sizes of birds collected in collaboration with the hunters in Cape Dorset, Nunavut. I found that while male eiders are more likely to have higher Hg concentrations than females, condition and liver size of eider ducks can also influence Hg, suggesting that liver size may moderate Hg concentrations. My results suggest that when assessing
the effects of Hg in wild birds, condition and sex should be considered. Importantly, my results also show that Hg and parasites can follow similar distribution patterns in eider ducks. I found that both parasite species examined had significant positive relationships with $\delta^{15}N$, indicating that eider ducks feeding at higher trophic had greater intensities of both helminthes examined. Additionally, I found that Hg also varied positively with $\delta^{15}N$, although the relationship was slightly non-significant. This common pattern between higher levels of parasites and Hg with increasing trophic levels demonstrates how these two environmental stressors may co-vary in wildlife. As a result, correlational studies that attribute biological effects to differences in Hg without accounting for parasites may produce spurious results given that both stressors can lead to similar biological effects such as changes in body condition and decreased reproduction.

Building on my findings from Chapter 3, in Chapter 4 I examine how Hg may affect eider duck reproduction and survival while also considering parasite burden. I did this by experimentally manipulating the parasite loads of female eider ducks with naturally varying Hg burdens (Chapter 4). For this work I administrated an oral anti-parasite treatment to breeding female eider ducks following Hanssen et al. (2003) and Bustnes et al. (2006). While this approach has been undertaken previously on wild eider ducks (Hanssen et al., 2003), my research is novel in testing how the experimental manipulation of eider duck parasites affected reproduction and survival in conjunction with Hg burdens. I also paired this work with a captive experiment to test for the efficacy of the anti-parasite oral treatment. While the captive experiment using males was not significant or conclusive, my research is one of the few examples that tests for how
contaminants and parasites may influence wildlife, and tests the assumptions of the experiment in a wild population.

Interestingly, I found that Hg concentrations did not influence eider duck reproduction or survival even when parasite burdens were considered, but did detect that modifying parasite burdens significantly altered breeding propensity in eider ducks that arrived late, or in poor condition. These results suggest that differing degrees of parasitism in eiders may have a larger influence on annual breeding outcomes than current Hg burdens do. This suggests that future work aimed at understanding the dynamics of eider reproduction and survival should focus on how parasites may be distributed within this species, how this may change over time, and on examining the mechanistic links between parasites and reproductive choices made by females.

In Chapter 5 I continue to examine how Hg concentrations may influence female eider ducks, but focused on potential sub-lethal effects of Hg on known breeding precursors such as immunoglobulin Y (IgY), corticosterone (CORT), arrival date and arrival condition. Based on preliminary results from analyzing 32 trace elements in these birds I also included lead (Pb) in this analysis as I found it to consistently be an important when assessing how trace elements vary with the physiological and phenological variables. I expected to find that both Hg and Pb had deleterious effects on IgY, CORT, arrival date and arrival condition (either directly or indirectly). While I did find that Hg negatively varied with CORT (which is linked with reproductive success in eiders; Hennin et al., 2016), blood Hg concentrations positively varied with eider duck condition. This demonstrates that while Hg may be a contaminant with negative effects on wildlife, the ecological relationships between pathways and effects are complex. This finding
illustrates the importance of critically examining the potential trade-offs in condition and effects when stressors are trophically transferred.

My results in Chapter 5 also highlight that there is significant interannual variation in blood Hg concentrations in eider ducks, which can influence the CORT levels in female eider ducks. This illustrates how Hg can indirectly, but significantly, influence a range of endpoints through altering the endocrine system. Interestingly, I also found that higher blood Pb concentrations were associated with decreases in body condition and later arrival dates, suggesting that Pb negatively influences eider duck onset and propensity. These findings suggest that Hg does influence eider ducks, even at low concentrations that are well below known toxicity thresholds, but that other contaminants such as Pb also negatively influence breeding in this species and should be considered when assessing the influence of contaminants on wildlife.

Overall, my thesis explores how Hg, alone and in concert with other environmental factors, can influence reproduction and survival in a marine bird model species. Collectively, my findings illustrate that while Hg itself may negatively affect wildlife, interactions with both parasites and other contaminants are complex and important to consider. My work also demonstrates that Hg may vary with condition and other biological metrics in unexpected ways, and research on effects must consider species across a range of Hg burdens to better understand how wildlife are influenced by this contaminant. Lastly, my research also highlights the importance of examining contaminant concentrations that align with the ecological endpoints of interest to fully assess the effects of Hg throughout an individual’s annual cycle.
6.1 Why it is important to continue to study Hg

In Chapter 2 I outline eight reasons why Hg in northern marine birds is an important research area to pursue. While I discuss how Hg concentrations detected in many Arctic wildlife species in Canada have shown to be increasing over the last few decades (Riget et al., 2011), more recent studies have demonstrated that Hg concentrations in some marine bird species may be stabilizing (Braune et al., 2015, 2016). These recent trends are encouraging, but there is continued need to examine how Hg population may affect wildlife as industrial outputs, and thus environmental deposition, continue to change over the coming decades (Provencher et al., 2014b and Chapter 2 above). A recent review of Hg concentrations in birds in western North America also highlighted northern Canada (and Nunavut specifically) as a hotspot for high levels of Hg in birds as compared to other regions in North America (Ackerman et al., 2016). This highlights the need to continue to understand what dynamics influence Hg burdens in marine birds in northern Canada as marine birds. Important to remember is that marine birds in northern Canada are both important apex predators, and harvested species, making this group both ecologically and culturally important in the region.

6.2 Hg and MeHg in seabirds

As outlined in Chapter 1 and 2, the most toxic form of Hg to wildlife is MeHg (or organic Hg). While marine birds are exposed to different forms of Hg, previous research has shown that in several seabird tissues upwards of 95% of the total Hg is MeHg (Bond and Diamond, 2009b; Kim et al., 1996b). This allows for total Hg (THg) to be a useful indicator of the more toxic organic form (Wiener et al., 2003). Importantly, while the ratio between MeHg and THg in breast muscle has been demonstrated to be between 66%
to 94% in birds (Houserova et al., 2007; Kim et al., 1996b), it has been shown that THg and MeHg have a significant linear correlation in muscle for most species examined, including seaducks (Houserova et al., 2007; Kim et al., 1996b). While there is less information on THg and MeHg correlations in seabird blood, blood THg is regularly used as a proxy for Hg burdens in marine birds (Ackerman et al., 2016; Fort et al., 2014; Hawley et al., 2009).

In this thesis I chose to analyze THg in all tissues. This was based primarily on the trade-off between getting more samples done for THg (a more inexpensive analysis) and assessing fewer samples for the more toxic form of MeHg (a more expensive analysis). As I moved through my PhD I decided that the strength of the work was in the large sample sizes I could obtain from northern hunters that allowed me to apply robust statistical analyses. Nonetheless, the use of THg as a proxy for MeHg may be a contributing factor to the lack of significant results presented here between THg and reproduction and survival. Future work should be undertaken to investigate the relationship between THg and MeHg in blood and other tissues in northern birds to validate the use of easily sampled tissues (i.e. feather, blood, etc.) on the primary target tissues of Hg (i.e. brain).

6.3 Contaminants throughout the annual cycle

Most of the work examining contaminants and their effects in migratory marine birds has been undertaken during the breeding season when birds are easy to access (Mallory and Braune, 2012; Piatt et al., 2007b; Riget et al., 2011). With the increase in techniques available to examine bird physiology and movements throughout the year, there is growing interest in investigating what influences marine bird contaminant
burdens across the annual cycle (Fort et al., 2014; Leat et al., 2013). Recent research has demonstrated how seasonal movements can influence contaminants burdens (Fort et al., 2014; Leat et al., 2013), and how contaminant burden can vary within an individual over both the short term (Bustnes et al., 2012, 2010) and long term (Lavoie et al., 2014). Importantly, this type of variation has implications for individual reproduction and survival.

One of the goals of this work was to investigate the flux and patterns of Hg burdens in eider ducks throughout their annual cycle. I found that Hg concentrations in the early pre-breeding season did negatively vary with CORT (which is linked with reproductive success; Hennin et al., 2016), I detected no effect of pre-breeding blood Hg concentrations on breeding propensity, clutch size, nest attendance or survival. Additionally, I found evidence that suggests that there may be a seasonal flux of Hg in eider ducks (Appendix D). Other research has shown that female eider ducks experience changing contaminant levels throughout the incubation period (Bustnes et al., 2012, 2010). Cumulatively, these findings suggest that eiders, and likely other marine birds, may experience changing levels of contaminants throughout their annual cycle leading to varying susceptibility to contaminants.

Of specific concern is when contaminants have effects on wildlife that vary throughout the year, with effects being potentially exacerbated during specific time periods of energetic stress. For example, during the winter months lower temperatures may coincide with reduced foraging rates, or changes in metabolism which pose specific energetic constraints (Lemly, 1993). In eider ducks the breeding season has been shown to be an energetically demanding period (Descamps et al., 2009; Hanssen, 2006).
Important to consider is that in wildlife relative contaminant concentrations may increase or become mobilized (for lipophilic contaminants), as body mass decreases during energetically demanding times, notably breeding and migration (Bustnes et al., 2010). Thus, studies examining how sub-lethal effects of contaminants on wildlife need to focus on times of energetic stress rather than just times of resource abundance in trying to elucidate potential impacts of contaminants on hosts. While breeding is an energetically demanding time period, migration and the pre-breeding season as examined in this thesis are also periods that contaminants may have differential effects as compared with other seasonal cycles.

6.4 The importance of direct and indirect effects of contaminants

The vast majority of contaminant effect research focuses on the direct effects of contaminants, often in species with high exposure rates (Braune et al., 2012; Heinz, 1974). While this work has greatly informed our understanding of how contaminants affect wildlife, I believe that there is great value in examining how contaminants may affect species in a variety of ways along a larger gradient of tissue concentrations. Species feeding at lower trophic levels (i.e. eiders and kittiwakes) may have lower Hg concentrations than upper trophic species (i.e. gulls and terns), and thus experience the effects of contaminants differently. For example, while reduced hatching success may occur in some species as higher levels (Braune et al., 2012), there are likely a multitude of effects from Hg and other contaminants occurring at the lower exposure levels that influence other processes such as the endocrine systems hypothalamic-pituitary-adrenal (Tartu et al., 2016, 2014, 2013; Chapter 5). The significant relationship between higher
levels of blood Hg and lower levels of CORT found in this thesis demonstrates how Hg may alter a suite of traits in marine birds.

Importantly, understanding what extrinsic factors influence individual physiology is of interest to wildlife researchers, but is also of interest to a broader research community (Hennin et al., 2016, 2012; Love et al., 2010). Additionally, it is the greater understanding of sub-lethal effects that allows researchers to understand the true impact of contaminants. While many species may have low exposure to both Hg and Pb, my research suggests that sub-lethal levels of trace metals from the environment may be associated with reduced reproduction. Such sub-lethal effects are critical to understanding the cumulative impacts of contaminants on wildlife. Recent work on captive birds has shown that low level sub-lethal concentrations of Hg are linked with increased loss of body mass, and altered foraging behaviour (Kobiela et al., 2015). Future work on wild bird species should focus on these types of effects and interactions to inform this growing body of contaminant research.

6.5 Eiders as a model species

I chose eider ducks as a model species due to several reasons, including access to samples, and relevance as a managed wildlife species. While seabirds are widely used as bio-indicators of the environment (Piatt et al., 2007b), the usefulness of eider ducks in particular at times may not be as apparent. Based on the work presented in the previous chapters I feel that indeed eiders are useful model species for cumulative effects research in northern Canada. Importantly, there is a plethora of research on eider duck survival, reproduction and physiology that make this species highly useful in examining how different environmental factors influence this species’ biology (Descamps et al., 2009;
Hennin et al., 2014; Legagneux et al., 2013; Steenweg et al., 2015). This body of research allows for a more in depth examination of traits in relation to contaminants that can be extrapolated out to a wider set of species where less is known about their physiology. Additionally, while the low levels of contaminants in this species may be seen as a disadvantage when examining the potential effects of environmental pollution, there is merit to understanding how low concentrations of contaminants may affect species. Research on the low levels effects of contaminants are more applicable to a wider range of organisms when considering how cumulative effects of contaminants may affect other species because a larger number of species may experience low to moderate levels of contaminants as compared with species with acute exposure.

6.6 Use of condition metric

While condition is a commonly used metric, it used throughout the literature in different ways in regards to wildlife. In Chapters 3 and 5 I use eider condition as a variable related to the fatness of an individual in relation to its overall size (total head length/body mass). The use of this condition metric was used based on the work with this eider population by Jamieson et al. (2006) and Descamps et al. (2011a). Alternative condition metrics can be used that include a suite of size metrics, and/or physiological measures that indicate if an individual is in good or poor condition such as lipid content in the blood. Future work should explore more how both physical and physiological measures may be used to measure the plumpness or fatness of a bird to explore the relationships discussed above in more detail.
6.7 Hg and Selenium interactions

Chapter 2 of this thesis describes some of the interactions Hg can have with other ecological processes (i.e. parasites, water acidity). Not presented here but considered within the toxicology literature as extremely important when considering Hg concentrations is the relationship between Hg and selenium (Se). While I do not present any data or work in this thesis considering Se, it was considered with Hg concentrations at each step in the process but was found to be insignificant. Based on these cumulative findings I decided to not include Se as a component within the main thesis framework. Although correlations between Hg and Se have been found more consistently in mammals, our results agree with those of Ohlendorf and Heinz (2011) and other studies from the Canadian Arctic (Provencher et al., 2014a) that no clear pattern between these two elements exists in marine birds.

6.8 Future research directions

My thesis aims to add to our understanding of how Hg affects Arctic marine birds. Through my different approaches I have investigated both the pathways and effects of Hg, but as many theses do, this has led to more questions that warrant further study.

6.8.1 Seasonal and interannual flux in Hg

I found significant interannual variation in blood Hg concentrations in female eider ducks (Chapter 3), and evidence that male eider ducks may also experience seasonal differences in Hg burdens (Appendix D). This phenomena has been found in other bird species in this region (McCloskey et al., 2013). While we know that Hg is trophically transferred to birds via their prey, we do not fully understand how fluxes in Hg in the environment may lead to annual variations in wildlife Hg levels. Given that low levels of
Hg may have sub-lethal but significant effects on wildlife, these interannual variations may contribute to individuals having reduced reproductive success (as demonstrated in Chapter 5), and are thus important to understand how extrinsic factors influence populations.

In the East Bay region in particular where much of this thesis work was done, interannual variation in Hg seems to be common among several species (McCloskey et al., 2013; Chapter 5). Based on these findings I believe that local geography and climate can influence Hg uptake in marine birds. For example, it has been shown that Hg is released into the meltwater during Arctic thaw events (Douglas et al., 2012; Zdanowicz et al., 2013). In areas where colonies of birds are situated within a watershed with large catchment areas (i.e. East Bay Island), Hg deposition to the freshwater system (where shorebirds feed), and eventually the river mouths (where eiders feed), may be highly determined by the rate and timing of snow melt. In years where the snow pack is minimal, or spring thaw occurs over a long period, the Hg deposited on the snow (via atmospheric depletion events) may enter the terrestrial system in a more gradual way, and be dispersed over time. On the contrary, in years where the snow pack is large, or the spring thaw happens rapidly, there is a potential for the freshwater and marine environments nearby to experience a Hg pulse in the freshwater environment. If the timing of phytoplankton blooms (and subsequently zooplankton and benthic invertebrates) match the thaw and pulse of Hg from the melting snow, this could lead to an increase in Hg throughout the food chain. Under the proposed circumstances, differing snow and thaw conditions each year could therefore lead to significant interannual variation in Hg concentrations in birds as observed in the East Bay region. Recent work
examining Hg in shorebirds in Alaska showed that species feeding in areas with higher moisture content had higher concentrations of blood Hg (Perkins et al., 2016). These results also suggest that wildlife Hg burdens may be influenced by local hydrology of a region.

In my future research program I will focus efforts on investigating how Hg moves through the Arctic ecosystems in relation to environmental conditions. I will take an ecosystem approach to study how Hg moves from the snow to the freshwater systems, and eventually to invertebrates and marine birds in areas with different hydrological conditions. By tracking the flow of Hg in the ecosystem and at different trophic levels, under differing annual conditions I hope to inform how seasonal conditions can influence marine bird contaminant levels, and thus the potential effects regional birds may experience.

6.8.2 Timing and effects of Hg on physiological processes

My final chapter (Chapter 5) investigated the indirect links between Hg and known breeding pre-cursors. One of the unexpected results was a positive correlation between increased blood Hg concentrations and individual arrival condition. While it is well known that Hg has a deleterious effect on many physiological processes (see Chapter 1), I found that in female eider ducks a positive relationship between condition and Hg. As discussed in Chapter 5, this positive correlation between condition and Hg may be driven by the need for eiders to increase their condition prior to the breeding season via increased foraging.

While increased foraging may lead to an immediate increase in condition, it can also lead to increased Hg burdens. It is possible that because eider ducks Hg
concentrations are so low, that the benefits of increased condition outweigh the increased Hg burden. Another explanation is that the benefits of the increased condition are short term, and the negative effects of increased Hg burdens either affect a parameter we did not measure in this study, or that appear later. For example, this increase in circulating Hg may be deposited into a hen’s eggs (depuration), and influence hatching success, a factor we were unable to measure. Alternatively, the increase in overall body Hg concentrations may not manifest until later in the year when the Hg is deposited in other tissues where Hg is known to cause damage (i.e. the brain) as discussed in Chapter 2. Either explanation suggests that marine birds experience fluxes in their contaminant loads that should be considered when examining possible effects on a species.

In my future research I will further explore this question of contaminant flux in marine birds as a necessary tool to understand the true impacts on contaminants on wildlife. I would like to continue to work with hunters and collaborators to gain access to samples year around and examine how contaminants loads may change over time in different tissues targeted by Hg and other contaminants. I believe that assessing how contaminant measures are biologically linked with the assess endpoints is critical to addressing the full impact of contaminants on wildlife.

6.8.3 Further investigation of the effects of parasites on birds

While my thesis work considered both Hg and parasites as drivers of eider duck reproduction and survival, I was limited in my examination of how parasites affect the same biometric and physiological metrics. I designed my research to investigate the effects of Hg, along with other various environmental factors, but my results from Chapter 4 (experimental manipulation of eider parasites in relation to Hg, reproduction
and survival) demonstrate that in this population of eiders, endo-parasites may have a
greater influence on eider reproduction then Hg. Similarly, the experimental reduction of
parasites in other bird species has led to altered immune function in other two species of
northern raptors (Hanssen et al., 2013). While my thesis was limited to examining the
effects of Hg, I believe that my findings along with other research that parasites can have
significant effects on a variety of traits in wild birds; an area of great research potential.

6.8.4 Contaminant cocktails

While I choose to focus on Hg, Pb and parasites for this study, Arctic eider ducks
are exposed to numerous other contaminants in the Canadian Arctic, including a suite of
persistent organic pollutants (POPs; Mallory et al., 2004). In the future, I will focus more
efforts on exploring how other contaminants in conjunction with Hg affect marine bird
health, reproduction and survival. The consideration of contaminants as a suite of
chemicals that can have cumulative effects on species is a growing field (Bustnes et al.,
2015; Nordstad et al., 2012; Sarigiannis and Hansen, 2012), but more work needs to be
done in this area that also considers the flux of contaminants and within the context of
other environmental stressors as outlined above.

6.9 Conclusions

As the field of ecotoxicology grows, there is an increased understanding of how
wildlife are affected by contaminants and other environmental factors. While research to
date has strived to assess the impacts of contaminants on wildlife, my PhD research has
led me to believe that more work is needed before we are able to estimate the combined
effects of contaminants and other environmental stressors are on species across the
spectrum of contaminant exposure. By continuing to study how contaminants may affect
species throughout their annual cycle, both directly and indirectly, including sub-lethal effects we can compile a more complete picture of how contaminants affect wildlife.
Appendices

Appendix A - How wildlife research can be used to promote wider community participation in the North

A.1 Abstract

Each year many research programs are undertaken in the territory of Nunavut, Canada. Although community consultation requirements have greatly increased the role of communities in research conducted in the region, current practices typically engage only a small portion of the population. This leaves many community members unconnected to research programs. Wildlife studies and related community based laboratory studies are one way that research programs can attract the attention of a wider community audience, thereby introducing wildlife research topics in a meaningful way. Here I describe a collaborative workshop facilitated by Environment Canada and the Nunavut Arctic College that engaged Northern students to develop skills related to Northern job opportunities in the field of wildlife research while using local knowledge. I describe the program as it was developed to involve Northern students between 2007 and 2011, and how program refinements may be applied to other research projects who aim to involve wider participation by Northern residents.

A.2 Introduction

Worldwide, researchers are being asked to better communicate their studies and findings to the public (Lubchenco, 1998). In Canada’s Arctic dozens of research projects take place each year ranging from natural science studies to social science investigations (Table 13), and in Nunavut, Canada community consultation has been legally mandated
pursuant to the Nunavut Land Claims Agreement (between the Government of Canada and the Inuit of Nunavut; Sections 5.1.2(h), 5.2.37, 5.2.38, and Article 33 as it extends to field research; Indian and Northern Affairs, 1993). Now, each research project that takes place in Nunavut must engage in community review as it passes through several layers of permitting and consultation before any work is authorized.

The permitting and consultation process in Nunavut is complex for those unfamiliar with the existing protocols, and varies with the scientific approach and duration of a project. Briefly, each research project must acquire a variety of permits from various regulatory agencies which may include the Canadian Wildlife Service, Department of Fisheries and Oceans, and the Nunavut Research Institute. Most of those permits require evidence of consultation with the community that are geographically closest to the study area prior to permit issuance. Experience has shown that this process is greatly facilitated when the research team has first contacted and consulted with the community on the proposed work and, if relevant, discussed results of previous studies. Given the importance of consultation, some government departments have initiated broader consultation processes where local organizations are given the opportunity to comment on any research plan in their jurisdiction, as well as being updated on territory-wide efforts (Gearheard and Shirley, 2007). As part of this procedural review (see Gearheard and Shirley, 2007 and Nickels et al., 2007 for more detail), the community groups that are involved in the numerous review boards have the opportunity to comment on projects, and to request more information. The primary purpose of this extensive application, review and consultation process is to provide Northern residents the opportunity to comment on how and where research is conducted near their communities,
and to identify potential risks to the local flora and fauna. Unfortunately, although great efforts have been made to rectify the ‘fly in, fly out’ approach that has sometimes been practiced by researchers in the past (Gearheard and Shirley, 2007; Korsmo and Graham, 2002), many Northern community members still report feeling disconnected from much of the research that occurs.

There are several practical, ethical and regulatory reasons for the involvement of local communities in research (Pearce et al., 2009). On a practical level, research is often dependent on community resources including local knowledge, guides and equipment, in order to complete work either within the community or out on the land (Gilchrist et al., 2005; Pearce et al., 2009). Researchers also have an ethical obligation to engage communities in the work as it is the members of the community that are living in the area being studied. In doing so, research findings may be more relevant to local management decisions (Pearce et al., 2009). In Canada’s North, where land claim agreements have been signed, researchers also have a legal obligation to engage and involve communities in local studies.

Community involvement in science programs has also been identified as a priority to improve local community decision-making, and is a recognized key element of successful co-management of resources (Fazey et al., 2006; Korsmo and Graham, 2002; Lebel et al., 2006). Involvement of communities increases local investment and support in completing the project. A recent study by the Ecological Monitoring and Assessment Coordinating Office and the Canadian Nature Federation (Ecological Monitoring and Assessment Network, 2002) examined ways to improve decision-making in local communities across Canada. They described how timely, relevant and accessible
information delivered as part of a two-way dialogue can greatly increase community involvement and understanding of findings across a variety of disciplines. This in turn has the potential to increase community capacity, stewardship and public participation in local governance related to the environment (Ecological Monitoring and Assessment Network, 2002). This highlights the need for research programs to not only involve groups in phases of data collection, but also to be active in sharing results with (and among) community members.

Despite the obligations and benefits of involving local community members, many researchers by their own admission simply do not have the contacts, skills, or resources to actively engage community members beyond the required permitting process (Gearheard and Shirley, 2007). At the same time, many educators in the North would like to incorporate more integrated learning experiences into their teaching curricula and, while researchers are often keen to contribute to educational programs, the resources to do so are lacking (Salmon et al., 2011). As a result, although legislation and consultation practices have been implemented and many community members are able to assist, inform and learn from research activities (usually hunters, Hunter’s and Trappers’ Organization members and guides), a large portion of Northern community members still feel uninvolved in local research programs, although many have a desire to be. In Nunavut for example, this is particularly true of students and educators at the elementary school to the post-secondary school levels. Given that 41% of Nunavut’s population is under the age of 20 years old (as of 2010; Nunavut Bureau of Statistics www.eia.gov.nu.ca/stats/), this suggests that there is a gap in community engagement among almost half of the Northern population. Thus, the next generation of hunters,
conservation officers and resource managers are not being fostered, encouraged and trained within many current research efforts, a missed opportunity.

Here I describe a series of outreach programs led by Environment Canada that involved elementary schools, high schools, and college students, with special focus on a collaboration developed with the Nunavut Arctic College (from here forward referred to as the college). This program overcame many of the above-mentioned challenges, and brought together students, educators and wildlife researchers in mutually beneficial learning experiences. I offer a review of this program as a tangible example of how wildlife researchers can interact with community members, and offer a set of lessons learned that I hope will help educators, researchers and managers work together to create similar programs.

A.3 The Research Program

Arctic marine birds are an important component of Northern ecosystems, both as harvested species for eggs, meat and down, and as indicator species used to detect changes in marine environments (Gilliland et al., 2009; Mallory et al., 2006c; Provencher et al., 2012). Consequently, marine bird studies are important to local communities, researchers and wildlife managers. Environment Canada, specifically the Canadian Wildlife Service and the Science and Technology Branch, is tasked with monitoring and studying migratory bird populations in Canada for the purpose of their conservation. As a result, several long-term monitoring programs have been established to study marine birds in Canada’s North since the 1970s (Gaston et al., 2009a).

Marine bird research in Nunavut often includes cooperation with the northern communities. Local Hunter and Trappers Organization are consulted in hiring guides to
travel in the area of colonies, and to collect hunted birds to study avian diet (Provencher et al., 2012), population and disease dynamics (Descamps et al., 2011a), contaminants (Braune, 2007), marine pollution (Provencher et al., 2010, 2009) and parasites (Mallory et al., 2007). When birds are collected with hunters they are later dissected with multiple tissue samples being archived within the National Wildlife Specimen Bank (Braune et al., 2010). This collection of samples is labour intensive in the lab after the field season is finished, and these activities provide an opportunity to engage people not directly involved with the collections themselves.

A.4 Chronology of the Outreach Program

Community involvement in the Environment Canada northern marine bird scientific programs has occurred for decades; including assistance from local guides and community consultations. More recently, a new education initiative was recently developed during the International Polar Year (2007-2008; IPY) as part of the Canadian IPY project titled “How Seabirds Can Help Detect Ecosystem Change in the Arctic” (see Provencher et al., 2012 for more details). Environment Canada partnered with the Nunavut Arctic College in Iqaluit to provide hands-on experience for students to dissect birds as part of the project’s science outreach initiative. In the first year of the program a PhD student spent two days working with 12 students from the college’s Environmental Technology Program (referred to as at the science class) in Iqaluit, and an Environment Canada researcher provided an introduction to marine birds in general and background for the project specifically through lectures. Students then dissected thick-billed murres and northern fulmars collected for the IPY science project (Mallory et al., 2010;
Provencher et al., 2010, 2009). The workshop was intended to provide hands-on training in dissection skills, and to provide an overview of marine bird research in Arctic Canada.

In 2008 and 2009, the workshop was held again and involved more students each year. In these years students were also offered a short-term employment opportunity after the workshop to help finish the dissection and related tissue preparation of remaining birds. Following the 2009 workshop, two students were also given the opportunity to travel to Ottawa to work at the National Wildlife Research Centre for an additional week to gain experience. This exchange was valuable to the students in adding to their skill base, and in building their own resumes and contacts within the larger research community, both of whom continue to work in northern research.

In 2011, Environment Canada initiated a new study examining avian disease, contaminants and internal parasites among both eider ducks (*Somateria mollissima*) and snow geese (*Chen caerulescens*). Birds were collected by field researchers and local hunters in Cape Dorset, Nunavut (GN Permit #WL-2011-029), and were used to again hold the workshop in Iqaluit and to expand the program. This time researchers from Environment Canada, the Nunavut Arctic College, Carleton University, University of Saskatchewan and the Government of Nunavut Department of Health and Social Services developed a program building on previous program successes. Specifically, the workshop was expanded to include concerns regarding wildlife and human health. A question-and-answer format was used to create an open dialogue in which students raised questions about human health and the nutrition of country foods, and both health and bird researchers discussed how their concerns were, or were not, being addressed through ongoing research.
In 2011 the Fur Production and Design class (hereafter, the design class) also joined the workshop to learn about marine bird research in Nunavut, and to both teach and learn about how eider skins are used in traditional design. One of the design students taught workshop participants about how to cut an eider skin in the traditional way used for making baskets and slippers (Fig 9). The science students were then given the option of dissecting their practice birds using either the scientific cut or the traditional cut. Finally, the remaining bird meat was shared through the Inuit Studies Program, providing food for students in need.

Although the traditional use and hunting of marine birds had always been discussed in the workshop, the involvement of the design class in 2011 allowed the students to showcase their own traditional skills in the context of a research program and teach their fellow students, instructors and researchers about how to process an eider skin for traditional basket- and slipper-making. This new component of the program greatly added to the workshop with several of the science students choosing to process their bird specimens in the traditional way and donate the skins to the design class for their own clothing projects.

In addition to the formal workshops, several informal visits to local schools were also made by researchers and graduate students as travel opportunities presented themselves. When researchers had birds in hand and time was available, they contacted local schools to inquire whether teachers would be interested in participating in a marine bird dissection demonstration. Over the course of the college collaboration, six demonstrations also took place at elementary schools and high schools in Cape Dorset and Sanikiluaq, Nunavut, and at both the high school and the French school in Iqaluit.
Through these additional school demonstrations, researchers reached young students within the community where the birds are harvested. With the growth of this program, the team of educators and researchers have gained extensive experience on the collective needs of students and education programs, and what resources are required to ensure success. Based on five years of workshops and demonstrations, I have identified eight key elements that were required to develop a successful program of this magnitude.

A.5 Key elements for program success

An integrated educational experience

To create a meaningful experience for students, a workshop requires adequate time and resources. A contributing factor to the success of many outreach programs during the IPY was the financial and logistical support of programs alongside ongoing science projects (Salmon et al., 2011). The marine bird dissection workshop has regularly been an educational component of a larger ongoing research program concerning the health of marine birds. Graduate students and researchers intentionally secured additional funding and integrated the needs of the existing college curriculum and course calendar. Topics that were highlighted in the workshop did not always align strictly with scientific priorities. Rather than try to present a complete and comprehensive view of the entire scientific program, which includes breeding phenology, physiology, disease pathology and epidemiology, population dynamics, toxicology, parasitology a few key messages were communicated instead. This included linkages between studying bird health and human health and what the diet of marine birds can tell us about changing environments; topics that were connected to the college curriculum.
The importance of relevant material

Classrooms require learning materials that focus on locally relevant topics. Further, it is more interesting to students when classroom topics are presented in a local context as it helps to connect with their own priorities and values (Simmons, 2000). Wildlife studies are often interesting to Northern residents as many people in Nunavut actively fish, hunt, trap, and regularly spend extended lengths of time camping. For example, harvested wildlife maintains key dietary and cultural relevance to Inuit (Kinloch et al., 1992), and people are interested in the state and health of local wildlife. Wildlife presents an instant and interesting conversation starter. Programs that also reflect the use of local ecological and societal systems, helps to encourage a personal connection with the material being presented (Simmons, 2000).

Understanding mismatched timelines

Research program timelines are often quite different from those of educational institutions. In the North, wildlife field work and collections typically occur in the summer (May to August), when students and instructors are not in school. By contrast, the best time for educational workshops to occur is during the late fall and early winter months when student programs are well underway and instructors are looking for educational resources to enhance their curricula. As a result, timing for an educational program can be a challenge. Clearly, effective workshops can rarely be ‘tacked on’ to the end of a field season (despite the economic advantage and logistical simplicity this offers for researchers). In light of this timing mismatch, research programs must plan prior to the field season how samples and collections will be stored for use in subsequent outreach and education programs.
For example, the collection of the birds used in the marine bird dissection workshops occurred in the spring months during the birds’ migration. Knowing that these birds had to be prepared and stored well in advance of the autumn school year meant that the carcasses had to be frozen and stored in Iqaluit annually (which required local facilities and collaboration with Environment and Climate Change Canada staff), rather than shipping them to southern labs, or dissecting them immediately. Initially, this organization of samples was a challenge, but I found that the benefits of the student help during the dissections greatly outweighed the early organizational time investment.

*Dedicated budgets*

The cost of working in the North can be prohibitive to many projects, including both scientific and outreach projects. Successful outreach programs typically require dedicated funding of their own to ensure equipment purchase, and to cover shipping, travel and accommodation costs (Salmon et al., 2011). One of the reasons this workshop has been successful and continues to grow is that it has been identified as a program priority by both Environment and Climate Change Canada and the Nunavut Arctic College. Each year, ancillary grants and funding have been secured to carry out the workshop and its associated costs. Without this funding, the workshops would not have been possible. Few science budgets can support such large outreach projects, and thus within science programs, establishing collaborations with other agencies that support complementary outreach and education programs is essential. In our program, the Nasivvik Centre for Inuit Health and Changing Environments has been a key funding source for this annual workshop. Often, programs with such targeted funding are able to achieve multiple objectives and unplanned successes that arise through sharing of
common priorities (Pokiak and Pokiak, 2011). I have found that these successes cannot be achieved without dedicated funding that supports direct interaction.

The importance of logistics and appropriate space

The availability of suitable facilities greatly helps learning activities. Students, researchers and educators alike are more stimulated and comfortable in rooms well lit with space that meets their needs. While it is true that dedicated spaces are not a necessity for engaging educational experiences, having a room with the proper equipment, lighting and space for people to interact, participate and easily view demonstrations easily, is a great benefit.

In the early years of the workshop, the dissections were held in a variety of venues; none that were directly associated with the college program, as the college did not have the appropriate facilities to carry out marine bird dissections. Although these venues allowed us to hold the workshop, they often had poor lighting and ventilation, were small, and lacked enough space to allow students to gather around to observe and subsequently try the dissection techniques (Fig 10A).

In 2009-2010, the college constructed new buildings that were purposely designed for science education through funding provided by the Government of Canada’s Arctic Research Infrastructure Fund. As a result, in 2011 the workshop was held in these new facilities where laboratories were fully equipped (Fig 10B). The new space provided good lighting, ventilation, and ample space. Having a dedicated space also allowed the researchers and instructors to expand number of participants where previous space constraints prevented this.
Face-to-face time

Many northern outreach events and projects are conducted successfully through the internet-based platforms in which researchers and students are linked (e.g. the PolarConnect webinar series sponsored by PolarTREC, http://www.polartrec.com/; the Ice Stories: Dispatches from Polar Scientists blog series, http://icestories.exploratorium.edu/dispatches/; and the Association of Polar Early Career Scientist’s Career Development Webinar Series, http://apecs.is.careers/career-development-webinars). However, the importance of interpersonal interactions cannot be underestimated (Gearheard and Shirley, 2007; Pokiak and Pokiak, 2011). This is especially true for applied programs where practical skills are highly (ex. environmental education; Ecological Monitoring and Assessment Network, 2002).

One of the strengths of the marine bird dissection workshop is that students, educators and researchers spend considerable time learning together. The workshop centers on dissecting marine birds, so the focus is on students practicing the skills needed to complete the dissection task. As such, the students are offered the time and space to highlight their own skills, ask questions, and interact with researchers on a one-to-one basis as they dissect their own practice bird. This time and interaction allows connections to form, and for two-way learning to occur. Students also come to realize how their efforts in processing the samples contributes to the overall scientific program. This helps instil among students the will to successfully complete the project, and forges a social contract that encourages researchers to return the next year to share results of the team work. These face-to-face benefits increase student engagement and comradery, and I
found helped to create a sense of community that included both researchers and community members in a joint venture of shared interest.

*Educator burn out*

Educator burn out must also be recognized as a challenge not only in the North, but in schools everywhere. Although science outreach programs help teachers meet some of their curricular needs, it must be understood that these interactions are in addition to their regular activities and therefore almost always generate extra work for them. Teachers have a complex job, and must cover the territorial curriculum, and respond to the changing and varied needs of their students. Not every teacher is open to working with researchers. In small communities there may be only a few key teachers willing or able to take on the extra work associated with extra-curricular outreach programs. For instance, the science program at the Nunavut Arctic College is contacted by numerous researchers wanting to work with the students each year. However, because of limited resources, and especially instructor time in particular, the college can only commit to a few. As a result, many researchers likely go away thinking the college is not interested, whereas in reality the real problem is a lack of time and staff to take on new projects. Educators and teachers are more likely to dedicate what little extra time they have to collaborations who are organized, require very little extra commitment from them, and that better enable them to cover aspects of the curriculum they are required to teach.

The marine bird dissection workshop and other classroom visits were highly successful for teachers in part because the researchers and graduate students deliver the program with very little additional commitment from the teachers other than time set aside in the class and space. When researchers first approached the schools and teachers
they asked if they could come into the classroom to share their research, clearly stated that they would provide the instruction and equipment. The researchers also sought insights from the teachers to assess what the students were learning and subsequently adapted their material to better fit the existing curriculum.

Recognition of education and outreach efforts is needed

A recent assessment of education and outreach during the 2007-2008 IPY (Salmon et al., 2011) identified that, although many researchers enjoyed doing science education and outreach programs, the lack of professional recognition for these types of activities within the academic community discouraged them. This was especially true when resources and time were increasingly limited (Salmon et al., 2011). Recognition can include letters of support, professional recommendations and awards. However, the simple lack of meaningful recognition by academia for outreach remains problematic (Salmon et al., 2011).

In small northern communities, positive or negative ‘word of mouth' and informal reports can make a great difference for returning research programs passing through the permitting and funding processes (as demonstrated in Gearheard and Shirley, 2007). During IPY, outreach was mandated as part of science program funding. Similarly, funding bodies such as the Northern Contaminants Program and the Nunavut Wildlife Management Board assess applicants not only on their scientific merit and productivity, but also on the basis of their outreach achievements in the North. As such, some of the long-standing programs that have continued on past the IPY were those with both clear science and outreach objectives (e.g. this project, many ArcticNET initiatives, the Coastlines bulletin). Even in the North where informal and formal outreach programs are
strongly encouraged, greater recognition of programs that provide relevant skills, knowledge, and meaningful interactions is needed.

A.6 Conclusions

Overall the marine bird dissection workshop brought students from two very different programs at the college together with researchers and educators from five different agencies, institutions and programs working in the North and South. The marine bird dissection workshop has been a positive experience for the students, educators and researchers. The students benefit from the skills and hands-on experience, the educators gain knowledge and context for their own work with the students. The researchers benefited both by having an able-bodied group of people to help process samples, and by interacting with people that have a wide range of experiences with bird species.

Community involvement in science is most often valued for its utility in addressing science questions (Huntington, 2000; Mallory et al., 2006b). However, research programs can and should be encouraged to broaden their scope. Research programs can create opportunities related to technical skill development and scientific literacy, often involving more people in the process. Although there are challenges to creating programs that benefit local students and researchers, wildlife studies can help to engage more Northerners beyond those that are directly involved in the permitting and consultation process. Understanding these challenges, where they fall within the larger research program, and how to manage them is essential in designing and implementing successful educational programs (Fig 11).

Long-term programs that have successfully integrated wildlife monitoring and a variety of community needs recognize that continuity of programs is directly linked to
“energetic collaboration among diverse partners and a unified approach that responds to identified needs” (Brook et al., 2009, p. 266). Although successful educational and research relationships can occur over short time periods (e.g. a one year project), engaging students, and the community at large probably occurs more often among long-term programs (Gagnon and Berteaux, 2009; Pokiak and Pokiak, 2011).

Fundamentally, the most important theme that runs through all of the key components is that meaningful outreach projects need to be made a priority. This requires devoted space, committed time to develop relevant educational material, and dedicated planning for how to collect and process samples in anticipation of student assistants. Without devoted time and energy even well-intentioned and planned programs often fall short of their potential, but with targeted resources generated by both researchers and educators, many research programs can expand to include wider community participation.

In the larger context, true community involvement in research programs needs to be promoted beyond those that are already involved in research consultation (e.g. Hunter’s and Trappers’ Organizations, hunters, etc.), and recognized as a way for a wider range of goals to be achieved. For example, by actively engaging groups that are outside this normal consultation stream, such as those that consist of mostly women (i.e. design programs), one can also engage a group that plays a critical role in promoting sustainable development and fostering changes in behaviour and attitude (Gregoire and Lebner, 2001).

Although great improvements have been made in the last few decades on how Northern communities are involved in the research that is conducted in their local area,
research programs are still not reaching their full capacity to engage wider community audiences. The ‘fly in, fly out’ approach has been greatly reduced in many ways, but programs that incorporate local skills and knowledge from the communities beyond those formally involved in the application review and consultation process are lacking. The experiences of this workshop support the idea that modern, western views of science and traditional knowledge can be integrated in a science curriculum that enhances both Northern communities and research programs (Van Eijck and Roth, 2007). Furthermore, younger members of the community need to be more actively involved in Northern research in order to foster the sense of connectedness to the research being undertaken and the desire to be more involved. As most of Nunavut’s population is under the age of 20 years old, research programs should be encouraged to move beyond just consulting, interviewing and hiring elders and hunters. They must create programs that train, engage and foster skills in the next generation of community leaders, thus meeting the long-term and future needs of Northern communities and Northern science.
Table 13. Number of permits issued each year by permitting bodies in Nunavut.

<table>
<thead>
<tr>
<th>Permitting Body</th>
<th>2007</th>
<th>2008</th>
<th>2009</th>
<th>2010</th>
<th>2011</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nunavut Research Institute</td>
<td>130</td>
<td>138</td>
<td>179</td>
<td>133</td>
<td>120</td>
</tr>
<tr>
<td>Department for Fisheries and Oceans</td>
<td>54</td>
<td>54</td>
<td>47</td>
<td>45</td>
<td>52</td>
</tr>
<tr>
<td>Canadian Wildlife Service(^1)</td>
<td>7</td>
<td>12</td>
<td>8</td>
<td>8</td>
<td>15</td>
</tr>
</tbody>
</table>

\(^1\) Many of these are multi-year permits, unlike other organizations which provide annual permits.
Figure 9. A Fur Production and Design student at the Nunavut Arctic College teaches researchers, and biology and design students how to remove the skin of a common eider duck in the traditional way used for making slippers and baskets; B – The demonstration of removing the eider skin (*Somateria molissima*) with an ulu, a traditional Inuit knife and; C – an eider duck basket made by the Fur Production and Design Class after their participation in the marine bird dissection workshop in 2011.
Figure 10. In 2007, 08 and 09 the spaces used for the workshop the NAC lacked dedicated workshop space. The rooms that were loaned to the program were often cramped, lacked good lighting and resulted in crowding and uncomfortable working conditions. B – With federal infrastructure funding in 2010 the Nunavut Arctic College was able to build new laboratory facilities with dedicated dissection areas with better lighting, storage space, sinks and equipment that greatly improved student and research working conditions.
Figure 11. Timeline and the associated effort required when undertaking meaningful long-term outreach collaborations in the North.
## Appendix B - Operational definitions of terms in bolded in the text

<table>
<thead>
<tr>
<th>Term</th>
<th>Operational Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abundance</td>
<td>Used to describe total number of parasites within all hosts examined, including uninfected hosts (Bush et al., 1997; Rozsa et al., 2000)</td>
</tr>
<tr>
<td>Acanthocephalan</td>
<td>Referred to as thorny-headed worms, in the Phylum Acanthocephala. These helminthes use at least two hosts in their life cycle and commonly found in several classes of invertebrates (Bush et al., 1997)</td>
</tr>
<tr>
<td>Bioaccumulation</td>
<td>The increase in concentrations of a contaminant in an organism from all sources (Newman, 2010)</td>
</tr>
<tr>
<td>Biomagnification</td>
<td>An increase in the concentration of a contaminant from one trophic level to the next (i.e. from prey to predator) attributable to accumulation via the food chain (Newman, 2010)</td>
</tr>
<tr>
<td>Cestode</td>
<td>Commonly referred to as a flatworm, in the Phylum Platyhelminthes and Class Cestoda. All members are parasitic and many are commonly found in several classes of vertebrates (McLaughlin, 2008)</td>
</tr>
<tr>
<td>Condition</td>
<td>Used to refer to the state of an individual indicative of overall health, often uses a combination of mass, size and fat indices</td>
</tr>
<tr>
<td>Contaminant</td>
<td>A natural substance, or released by human activities that makes something less pure and potentially poisonous (Hoffman et al., 2003; Newman, 2010)</td>
</tr>
<tr>
<td>Definitive host</td>
<td>The host in which a parasite achieves sexual maturity (Roberts and Janovy, 2005)</td>
</tr>
<tr>
<td>Helminth</td>
<td>A parasitic worm that lives within a host</td>
</tr>
<tr>
<td>Immunosuppression</td>
<td>Reduction in an individual’s immune function</td>
</tr>
<tr>
<td>Intensity</td>
<td>Used to describe the total number of parasites in hosts that were infected, excluding uninfected hosts (Bush et al., 1997; Rozsa et al., 2000)</td>
</tr>
<tr>
<td>Intermediate host</td>
<td>The host of a parasite where a parasite spends a portion of its lifecycle but does not reach sexual maturity (Roberts and Janovy, 2005)</td>
</tr>
<tr>
<td>Prevalence</td>
<td>Percentage of all hosts infected based on total number of hosts examined (Bush et al., 1997)</td>
</tr>
<tr>
<td>Stressor</td>
<td>An external factor that may limit an individual’s ability to grow, survive or reproduce</td>
</tr>
<tr>
<td>Survival</td>
<td>Used to describe apparent year to year survival of an individual</td>
</tr>
<tr>
<td>Trophically transmitted</td>
<td>A route by which both parasites and contaminants may be transmitted from prey to predator to reach organisms higher in the food chain</td>
</tr>
<tr>
<td>Trophic level</td>
<td>A position in a food chain or web (Mittelbach, 2012)</td>
</tr>
</tbody>
</table>
Appendix C - Reports submitted to the Northern Contaminants Program

C.1 Background on the Northern Contaminants Program

All the Hg analysis for this project was funded by the Northern Contaminants Program (NCP) under the department of Indigenous and Northern Affairs Canada (http://www.aadnc-aandc.gc.ca/eng/1100100035611/1100100035612). The aim of the NCP is to support research and work to reduce, and when possible eliminate, contaminants in traditionally harvested foods while providing information to northerners to assist in decision making regarding country food and contaminants.

Each year the NCP funds contaminants research in northern Canada. In 2013-14 and 2015-16 NCP funded two projects in support of the thesis work presented here. While some of the objectives of the NCP projects are presented in Chapters 3-5 above, there are several objectives addressed within my work that are related to NCP priorities (temporal and spatial trends of Hg), that were not included in the main data chapters of the thesis. As part of the funding structure of the NCP annual synopsis reports are submitted following each funding cycle. This appendix summarizes the NCP supported work examining patterns of Hg in eider ducks related to this thesis, but not directly presented in the previous chapters (Gilchrist and Provencher, 2016, 2014).

C.2 Introduction and Objectives

Due to global atmospheric wind currents, the main source of elemental Hg in Arctic Canada from industrial activities is from North America and Asia (Munthe et al., 2011; Pacyna et al., 2010). As changes in emissions in these two regions differ, the deposition of Hg in different Arctic regions is complex (Kirk et al., 2012; Munthe et al.,
Thus, the deposition of Hg across Arctic Canada is not uniform (Kirk et al., 2012; Munthe et al., 2011). This has implications for Hg studies in wildlife as Hg concentrations may differ within species across time and space. Thus, to fully understand how ecosystems and wildlife are exposed to Hg contamination, it is important consider both temporal and spatial trends.

In order to understand the broader findings presented in this thesis, I have several secondary objectives related to the patterns of Hg concentrations in eider ducks (Somateria molissima). First, I compare Hg concentrations in two related eider duck species which breed in northern Canada. S. m. borealis (hereafter migratory) is a migratory subspecies of eider duck found throughout the eastern Canadian Arctic and western Greenland. Migratory eider ducks breed in northern Canada and migrate to the open waters of Greenland and Atlantic Canada each autumn where they overwinter until the following spring (Goudie et al., 2000). The subspecies S. m. sedentaria (hereafter resident) is a subspecies of eider duck that does not migrate, and is found all year in southern Hudson Bay where it overwinters in areas of open water among the sea ice (Goudie et al., 2000). Although, the two sub-species differ in their migration ecology, they have otherwise similar in life history traits (e.g., timing of breeding, clutch size, chick rearing habits; Goudie, Robertson and Reed 2000).

Second, I examine how Hg concentration in eider ducks may vary between years. This was done using male eider breast muscle from Cape Dorset and female eider blood from East Bay Island, both sampling locations where the work was carried out in multiple years. Third, I compare Hg concentrations in female eider blood at East Bay Island collected for this project with historical samples collected in the 1990s (Wayland et al., 2011).
2001a) to assess for changes in Hg burdens over time. Lastly, I compare breast muscle Hg concentrations within a single population of eider ducks in two different seasons to investigate if this species experiences seasonal variation in Hg burdens. The overall aim of these objectives was to assess how Hg concentrations may differ in time and space within a species.

C.3 Methods

In addition to the breast muscle samples collected from eider ducks in Cape Dorset (Chapter 3; migratory), I also collected eider ducks from East Bay (n = 11; migratory) and with hunters in Sanikiluaq (n = 20, resident; Table 14). The eider ducks sampled for breast muscle Hg concentrations from East Bay Island were the subset of males used to test the efficacy of the anti-parasite treatment (see Chapter 4). Additionally, resident eider ducks were collected in Sanikiluaq in 2012, 2013 and 2014 in collaboration with local hunters in March of each year when they were concentrated in regions around polynyas (open water areas surrounded by sea ice) in the Sanikiluaq region. Birds were either frozen or dissected in the community within 36 h of collection. Similar to other hunted birds presented in this thesis, all birds that were frozen were later dissected using the same protocols as outlined above at the NWRC or the Nunavut Arctic College (Provencher et al., 2013).

In 2013 and 2014 blood samples were taken from migratory female eider ducks as they arrived at the colony on East Bay Island (as outlined in Chapter 5). These values were then compared to individual female Hg concentrations sampled in 1997 and 1998 by Wayland et al. (2001a) to examine for changes in Hg burdens in eider ducks at this colony over time.
A general linear model (GLM) approach was used to assess the differences in Hg concentrations between sub-species (migratory vs. resident), sampling year (interannual variation), time periods (1990s vs 2010s) and season (migration vs. breeding). Mercury concentration values in breast muscle were log transformed to meet parametric test assumptions, while Hg concentrations in blood were square root transformed for our analyses. Based on the differences between the sexes found in Chapter 3 I included only male eiders to examine differences in Hg concentrations in eider ducks when breast muscle tissue is used.

C.4 Results

There was no significant difference detected in the breast muscle Hg concentrations in adult male eider ducks from the two different sub-species (S.m. borealis n = 20, S.m. sedentaria n = 141; GLM $F_{1,159} = 0.173, p = 0.678$; Gilchrist and Provencher, 2014). I found no interannual variation in breast muscle Hg concentrations when just the birds from Cape Dorset were examined from 2011 (n = 98) and 2012 (n = 43; GLM $F_{1,139} = 0.77, p = 0.38$; Fig 12), therefore I grouped all the males from Cape Dorset together for other tests. As already presented in Chapter 5, I did detect significant interannual variation in the blood Hg concentrations in female eider ducks (GLM $F_{1,188} = 17.8, p < 0.0001$). Over the longer time period examined I found that the blood Hg concentrations in females sampled in 1997/1998 (n = 28) were significantly lower than those sampled at the same breeding colony in 2013/2014 (n = 201; GLM $F_{1,229} = 7.01, p = 0.008$; Fig 12; Gilchrist and Provencher, 2016). I found that in general the eider ducks sampled during migration in Cape Dorset had lower breast muscle Hg concentrations as compared with the breeding eider ducks at East Bay (Fig 13), but these differences were
not statistically significant (Cape Dorset n = 141, East Bay n = 11; GLM $F_{1,151} = 3.42, p = 0.07$).

C.5 Discussion and Conclusions

Previous studies have found large variation in eider duck contaminant concentrations within the Canadian Arctic (Mallory et al., 2004), but we detected no difference in breast muscle (indicative of the months before sampling) Hg concentrations between the two sub-species of eiders which utilize different regions within the Canadian Arctic. This suggests that the two sub-species of eider ducks in northern Canada has similar exposure to environmental Hg even though they overwinter in very different regions.

While we found no interannual in the Hg burdens in eider duck breast muscle, we did find that the blood Hg concentrations from breeding females at East Bay Island were significantly different between sampling years. These results highlight the importance of sampling Hg and other contaminant concentrations in wildlife over time to account for interannual variation which may produce spurious results in short term studies. Since blood is likely indicative of mercury intake over the period of days or weeks (Hobson and Clark, 1992b), the blood Hg levels likely indicate that females in East Bay took in lower levels of Hg in 2013 versus 1997/98 and 2014. This could be indicative of differing annual mercury deposition rates in the region. The difference in concentration between the two sampling time periods may also be due to differences in local conditions in Hg availability in the ecosystem at this time of year related to seasonal snow melt and release of Hg (Provencher et al., 2014b; Zdanowicz et al., 2013). Other marine birds in the area
have shown similar inter-annual differences in Hg concentrations over very short time spans (McCloskey et al., 2013), suggesting that local drivers may be influencing Hg in marine birds in the region.

While we did find that contemporary eiders had lower levels of blood Hg than individuals sampled in the late 1990s, the significant interannual variation that we also detected makes the interpretation of these relatively few data points difficult. Our findings emphasize the need for long term monitoring of Hg using regular intervals that allow for inter-annual differences and long term trends to be considered (Riget et al., 2011).

Mercury concentrations in male eider duck breast were in general higher in the migrating birds as compared with breeding male eiders. While this relationship was slightly non-significant, it suggests that more work is needed to better understand how Hg concentrations in eider ducks may change over the seasons leading to different expected toxicological impacts throughout a bird’s annual cycle. Overall, this work shows that there may be differences in Hg concentrations in eider ducks related to year and season that differ by tissue type. This is important to consider when long term trends are being examined. Additionally, as ecotoxicology explores the impacts of contaminants on wildlife, it is important to consider the flux of Hg and other contaminants within wildlife in relation to their annual cycle.
Figure 13. Breast muscle Hg concentrations in male eider ducks (*Somateria mollissima borealis*) during the migratory and breeding seasons. Hg values are dry weight measures.
Appendix D - Data Archiving

This thesis has discussed how changes in both contaminants and parasites in wildlife can be used to study ecosystem changes across time and space. Therefore, the archiving of data and tissues related to this work is extremely important. With hundreds of birds and thousands of tissues collected as a part of this thesis (Table 14), management of both tissues and the data associated has been a critical component to my research program.

For posterity I include in this appendix an overview of all eider ducks (both common and king eiders as they are often collected by hunters while flying in mixed flocks) collected and processed for this project. Data and tissue archiving has occurred in several steps. First, tissue samples from all birds are stored at the NWRC in Ottawa as part of Environment and Climate Change Canada’s Wildlife Specimen Bank (Braune et al., 2010). This bank is publically accessible to researchers beyond Environment and Climate Change Canada, and samples can be requested with a project submission to the specimen bank manager. While breast muscle tissue was used in this project, numerous other tissues (feathers, liver, kidney, bone etc.) have been archived and can be made available to future projects (Table 14). Parasites collected as part of this work have been stored in both 90% ethanol and frozen in chemically clean vials. All parasites are stored at the NWRC. Voucher specimens have also been prepared by J.D. McLaughlin and submitted to the New Brunswick Museum along with his own collection.

Second, all metadata from this project are archived and available through the Polar Data Catalogue (CCIN reference number: 11810, 11553, and 11809). Additionally, all the data associated with Chapters 2, 3 and 4 are archived in the Polar Data Catalogue
(CCIN reference number: 11810, 11553, and 11809). All biometric data collected for this project (including all blood trace element concentrations, IgY, CORT, and breeding data) from females at East Bay Island is also archived in Grant Gilchrist’s East Bay database at the NWRC. The above tissues and data are archived with the hopes that they may inform future work, and that while this thesis project has resulted in large sample sizes and datasets there is still much to be done with the data available from these birds.
Table 14. Summary of samples from common eider (*Somateria mollissima*, including sub species *S.m. borealis* and *S.m. sedentaria*) ducks and King eiders (*Somateria spectabilis*) collection in Nunavut, Canada as part of this thesis research. Information on tissues archived and stored at the National Wildlife Research Centre and analyses completed to date is included along with sample size (n).

<table>
<thead>
<tr>
<th>Year</th>
<th>Location</th>
<th>Species</th>
<th>Tissues banked at NWRC</th>
<th>Completed analyses</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>2011</td>
<td>Cape Dorset</td>
<td><em>S.m. borealis</em></td>
<td>breast muscle, liver, kidney, leg bone, wing, head, helminths</td>
<td>GIT helminth identification and enumeration; stomach contents assessed; Hg and stable isotopes (C and N) in breast muscle</td>
<td>176</td>
</tr>
<tr>
<td></td>
<td>Cape Dorset</td>
<td><em>S.spectabilis</em></td>
<td>breast muscle, liver, kidney, leg bone, wing, head, helminths</td>
<td>GIT helminth identification and enumeration; stomach contents assessed</td>
<td>14</td>
</tr>
<tr>
<td>2012</td>
<td>Cape Dorset</td>
<td><em>S.m. borealis</em></td>
<td>breast muscle, liver, kidney, leg bone, wing, head, helminths</td>
<td>GIT helminth identification and enumeration; stomach contents assessed; Hg and stable isotopes (C and N) in breast muscle</td>
<td>66</td>
</tr>
<tr>
<td></td>
<td>Cape Dorset</td>
<td><em>S.spectabilis</em></td>
<td>breast muscle, liver, kidney, leg bone, wing, head, helminths</td>
<td>GIT helminth identification and enumeration; stomach contents assessed</td>
<td>8</td>
</tr>
<tr>
<td></td>
<td>Belcher Islands</td>
<td><em>S.m. sedentaria</em></td>
<td>breast muscle, liver, kidney, leg bone, wing, head, helminths</td>
<td>GIT helminth identification and enumeration; stomach contents assessed; Hg and stable isotopes (C and N) in breast muscle</td>
<td>13</td>
</tr>
<tr>
<td>2013</td>
<td>Belcher Islands</td>
<td><em>S.m. sedentaria</em></td>
<td>breast muscle, liver, kidney, leg bone, wing, head, helminths</td>
<td>GIT helminth identification and enumeration; stomach contents assessed; Hg and stable isotopes (C and N) in breast muscle</td>
<td>62</td>
</tr>
<tr>
<td></td>
<td>East Bay</td>
<td><em>S.m. borealis</em></td>
<td>breast muscle, liver, kidney, leg bone, wing, head, helminths</td>
<td>GIT helminth identification and enumeration; stomach contents assessed; 32 trace element concentrations from blood</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>East Bay</td>
<td><em>S.m. borealis</em></td>
<td>na</td>
<td>32 trace element concentrations from blood; Immunoglobulin and corticosterone</td>
<td>92</td>
</tr>
<tr>
<td>2014</td>
<td>Belcher Islands</td>
<td><em>S.m. sedentaria</em></td>
<td>breast muscle, liver, kidney, leg bone, wing, head, helminths</td>
<td>na</td>
<td>58</td>
</tr>
<tr>
<td></td>
<td>East Bay</td>
<td><em>S.m. borealis</em></td>
<td>na</td>
<td>32 trace element concentrations from blood; Immunoglobulin and corticosterone</td>
<td>92</td>
</tr>
</tbody>
</table>

**TOTAL** 597
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