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Spatial variation in polycyclic aromatic hydrocarbon exposure in Barrow's goldeneye (*Bucephala islandica*) in coastal British Columbia

Megan Willie [a,*], Daniel Esler [b], W. Sean Boyd [c], Philip Molloy [d], Ronald C. Ydenberg [e]

[a] Centre for Wildlife Ecology, Simon Fraser University, 8888 University Drive, Burnaby, BC V5A 1S6, Canada
[b] US Geological Survey, Alaska Science Center, 4210 University Drive, Anchorage, AK 99508, USA
[c] Science and Technology Branch, Environment and Climate Change Canada, 5421 Robertson Road, Delta, BC V4K 3N2, Canada
[d] Stantec Consulting Ltd., 2042 Mills Rd W, Sidney, BC V8L 5X4, Canada
[e] Centre for Wildlife Ecology, Simon Fraser University, 8888 University Drive, Burnaby, BC V5A 1S6, Canada

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ABSTRACT

Barrow's goldeneyes are sea ducks that winter throughout coastal British Columbia (BC). Their diet consists primarily of intertidal blue mussels, which can accumulate PAHs; accordingly, goldeneyes may be susceptible to exposure through contaminated prey. In 2014/15, we examined total PAH concentrations in mussels from undeveloped and developed coastal areas of BC. At those same sites, we used EROD to measure hepatic CYP1A induction in goldeneyes. We found higher mussel PAH concentrations at developed coastal sites. Regionally, goldeneyes from southern BC, which has relatively higher coastal development, had higher EROD activity compared to birds from northern BC. Our results suggest goldeneyes wintering in coastal BC were exposed to PAHs through diet, with higher exposure among birds wintering in coastal areas with greater anthropogenic influence. These results suggest the mussel-goldeneye system is suitable as a natural, multi-trophic-level indicator of contemporary hydrocarbon contamination occurrence and exposure useful for establishing oil spill recovery endpoints.

1. Introduction

Oil pollution in the marine environment is a significant cause of bird mortality in Canada (Wiese et al., 2004; O'Hara et al., 2009; Calvert et al., 2013). Among marine wildlife, birds are considered especially sensitive to lethal or sub-lethal effects resulting from direct contact, inhalation, absorption, or ingestion of oil (Leighton, 1993). Birds that come into direct contact with oil will readily adsorb it onto their feathers. Adsorbed oil reduces the waterproofing, insulating, and buoyancy properties that feathers provide; loss of these functions can result in death due to starvation or hypothermia (Leighton, 1993; Wiese, 2002). Similarly, oil that is inhaled, absorbed, or ingested can exert debilitating or lethal toxicity on internal tissues and organs (Eisler, 1987; Leighton, 1993; Piatt and Anderson, 1996; Franci et al., 2014).

Catastrophic oil spills have resulted in sizeable marine bird mortality events. Mortality estimates following the *Amoco Cadiz*, *Exxon Valdez*, *Prestige*, *Selendang Ayu*, and *Deep Water Horizon* oil spills range from 100,000 to 300,000 birds (Piatt et al., 1990; Munilla and Velando, 2010; Haney et al., 2014). In some marine ecosystems, however, chronic oil discharges are reported to contribute larger inputs of hydrocarbons over time than catastrophic spills, with subsequent, enduring effects on birds (Leighton, 1993; Wiese, 2002; Camphuysen, 2007). Chronic discharges are regularly-occurring hydrocarbon releases typically associated with marine industrial activities (e.g., discharges of oily waste water from vessels) but also can be sourced from naturally occurring oil seeps, atmospheric emission and deposition, surface water runoff, and recreational, or commercial marine activities (Eisler, 1987; Camphuysen, 2007; Nikolaou et al., 2009; Yunker et al., 2011).

Among classes of hydrocarbons, polycyclic aromatic hydrocarbons (PAHs), in particular, have potential to exert toxic effects on marine birds (Eisler, 1987). Both the United States (US) and Canadian Environmental Protection Agencies (EPAs) have classified PAHs as priority pollutants due to their persistence in the environment, ability to act as carcinogens or mutagens, and consequently, their potential to cause adverse physiological effects in vertebrates (Canadian Environmental Protection Agency, 1994; Bojes and Pope, 2007; Pampanin and Sydnes, 2013). Chronic PAH exposure has been attributed to a wide range of effects in marine birds, including immune suppression, oxidative stress in the liver and kidneys, depressed reproductive performance, embryotoxicity, susceptibility to disease, and death (Eisler, 1987; Leighton, 1993; Canadian Environmental Protection Agency, 1994; Piatt and Anderson, 1996; Esler et al., 2010). Collectively, effects

* Corresponding author at: Stantec Consulting Ltd., 500-4730 Kingsway, Burnaby, BC V5H 0C6, Canada.
E-mail addresses: megan.willie@stantec.com (M. Willie), desler@usgs.gov (D. Esler), sean.boyd@canada.ca (W.S. Boyd), philip.molloy@stantec.com (P. Molloy), ydenberg@sfu.ca (R.C. Ydenberg).

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from chronic exposure can have wide-ranging demographic consequences on marine bird populations that exceed lethal effects of acute exposure experienced during catastrophic spills (Szaro, 1977; Leighton, 1993; Nur et al., 1997; Wiese and Robertson, 2004; O'Hara et al., 2009; Esler and Iverson, 2010; Henkel et al., 2014).

Birds that live and forage in industrialized coastal ecosystems are particularly vulnerable to sustained exposure to hydrocarbons (Miles et al., 2007). In western North America, British Columbia is heralded as the "Gateway to the Pacific". Several regions of the British Columbia coast operate as major departure points for the transport of commercial and industrial commodities to primarily Asian markets. Deep-water ports located in Vancouver, Kitimat, and Prince Rupert serve as confluences of several major shipping routes. Shipping activity in British Columbia includes tankers, bulk carriers, tugs, barges, ferries, cruise ships, and recreational and commercial fishing boats. Through these activities, an estimated 110 million m^3 of petroleum are transported along the British Columbia coast each year (Nuka Research and Planning Group LLC, 2013). Recent proposed industrial expansion could see the addition of >20 coastal facilities, including several oil and liquefied natural gas terminals. Collectively, these developments could result in an associated increase of approximately 1,000 tankers and bulk carriers transiting British Columbia waters each year (Nuka Research and Planning Group LLC, 2013). Existing and future marine industries have the potential to result in chronic or catastrophic PAH inputs into coastal ecosystems. However, despite a long history of industrial activity in the province, the ecological risk of contemporary oil exposure to marine birds is poorly quantified. Little is known of the relationship between PAH concentrations in marine bird prey and evidence of exposure in birds. The degree of contemporary PAH exposure among marine birds occupying industrialized coastlines is similarly undocumented. These knowledge gaps limit efforts to predict and manage effects from existing and future hydrocarbon contamination.

In British Columbia, the potential for contemporary chronic PAH exposure in marine birds is well characterized by Barrow's goldeneyes (*Bucephala islandica*). Barrow's goldeneyes are sea ducks that winter in the Pacific Northwest; an estimated 60% of the global population resides in coastal British Columbia during winter months (Campbell et al., 1997; Eadie et al., 2000). Large concentrations of Barrow's goldeneyes occur throughout coastal British Columbia, representing a wide range of industrialization and human activity (Campbell et al., 1997; Horwood, 1992; Important Bird Areas Canada, 2016). Barrow's goldeneyes show strong seasonal associations with intertidal habitats where they feed almost exclusively on blue mussels (*Mytilus* spp.) (Eadie et al., 2000). The risk of toxicological effects from sustained consumption of contaminated prey is greatest for species feeding on organisms with high PAH burdens (Newell, 1989; Boehm et al., 1996). Among mollusc species, blue mussels are generally thought to be poor metabolizers of PAHs and as filter-feeders are highly susceptible to, and capable of accumulating high PAH burdens in their lipid-rich tissues from their immediate surroundings (Boehm et al., 1996; Meador, 2003; Galgani et al., 2011). However, partitioning and depuration of PAHs in mussel tissue is influenced by the bioavailability of individual PAHs but is also regulated by the foraging behaviour and physiology of the organism and exogenous environmental factors (Fossato and Canzonier, 1976; Miles and Roster, 1999; Meador, 2003). Consequently, goldeneyes are expected to be particularly prone to toxicological effects of dietary sources of PAH and, hence, constitute a simple, sensitive vertebrate indicator of PAH exposure in coastal ecosystems in British Columbia.

The objectives of this study were to evaluate the degree to which Barrow's goldeneyes wintering in coastal British Columbia exhibit exposure to PAHs and to identify whether ingestion of contaminated prey serves as a pathway for exposure. To investigate this, non-lethal liver biopsies were obtained from goldeneyes captured from sites representing varying degrees of industrialization. Because vertebrates rapidly metabolize hydrocarbon compounds, it is difficult measure PAH concentrations in bird tissues directly. The cytochrome P4501A (CYP1A) system is reliably induced by metabolism of a small suite of contaminants, including PAHs, polychlorinated dibenzo-p-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs), and polychlorinated biphenyls (PCBs). 7-Ethoxyresorufin-O-deethylase (EROD) activity from liver samples was used to examine CYP1A expression. Previous studies have established EROD as an effective biomarker of exposure to PAHs and other contaminants (e.g., Trust et al., 2000; Miles et al., 2007; Esler et al., 2011; Franci et al., 2014). Elevated EROD activity can indicate both recent and chronic contaminant exposure (Altenburger et al., 2003; Miles et al., 2007). Measures of EROD activity were compared to sex, age, and body mass attributes from sampled goldeneyes to explore potential physiological relationships in birds exhibiting evidence of higher hydrocarbon exposure. Patterns of exposure also were hypothesized to correspond with PAH burdens in goldeneye prey. Accordingly, we measured the concentration of total PAHs in blue mussels obtained from goldeneye wintering sites in northern and southern British Columbia. Because PCDDs, PCDFs, and PCBs also serve as potential inducing agents, and previous studies indicate that these compounds are bioavailable to marine wildlife in British Columbia (e.g., Elliott and Martin, 1998; Harris et al., 2003; Jacques Whitford Axys, 2010), mussels also were evaluated for the potential occurrence of other CYP1A inducing agents. We predicted that birds wintering in industrialized coastal areas would demonstrate higher levels of EROD activity as a result of ingesting prey that are more likely to be chronically exposed to PAHs (Iverson and Esler, 2006; Miles et al., 2007; Oros et al. 2007; Kimbrough et al., 2008). This pattern was expected to occur across industrialized coastal habitats in northern and southern British Columbia.

2. Materials and methods

2.1. Study areas

To investigate PAH contamination in blue mussels and exposure in Barrow's goldeneyes, sampling was conducted in coastal regions of northern and southern British Columbia where large congregations of goldeneyes occur (d'Entremont, 2010; Horwood, 1992; Important Bird Areas Canada, 2016). In both regions, samples were collected at industrialized, intermediate, and reference sites. Industrialized sites were characterized by the historical and contemporary presence of industrial, commercial, and recreational shore-based facilities and associated vessel traffic. These sites serve as known or putative sources of PAHs. Intermediate sites were identified as areas without major shore-based infrastructure (with the exception of small residential communities) and support a lower intensity of marine-based activity. Samples collected from industrialized and intermediate sites were compared to those obtained from reference locations. For this study, reference sites were defined as lacking shore-based infrastructure and encompassed areas where marine activity was entirely from recreational watercraft (i.e., small motorized and non-motorized vessels).

2.1.1. Northern British Columbia: Kitimat Arm, Douglas Channel, and Kiskosh Inlet

In northern British Columbia, goldeneye and mussel sampling was conducted in the Douglas Channel fjord system, extending 150 km inland from the Pacific Ocean to Kitimat Arm (Fig. 1). Industrial activity in the Kitimat Estuary dates back to the early 1950′s with the operation of the Rio Tinto Alcan aluminum smelter, the Eurocan pulp and paper mill, and the Methanex methanol production plant (Levings, 1976; MacDonald and Shepherd, 1983). PAH loads in marine sediments in Kitimat Arm are highest in northern parts of the estuary (Simpson et al., 1996; Yunker et al., 2011). The south end of Douglas Channel marks the confluence of several major shipping channels supporting tankers, bulk carriers, ferries, cruise ships, tugs, fishing boats, and recreational vessel traffic. Two notable shipwrecks have occurred in this area. The M/V *USAT Brigadier General M.G. Zalinski* was a US Army transport ship that sank in southern Grenville Channel in 1946, containing 700 t