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Pre-oil spill baseline profiling for contaminants in Southern Resident killer whale fecal samples indicates possible exposure to vessel exhaust

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ABSTRACT

The Southern Resident killer whale population (*Orcinus orca*) was listed as endangered in 2005 and shows little sign of recovery. Exposure to contaminants and risk of an oil spill are identified threats. Previous studies on contaminants have largely focused on legacy pollutants. Here we measure polycyclic aromatic hydrocarbons (PAHs) in whale fecal (scat) samples. PAHs are a diverse group of hazardous compounds (e.g., carcinogenic, mutagenic), and are a component of crude and refined oil as well as motor exhaust. The central finding from this study indicates low concentrations of the measured PAHs (< 10 ppb, wet weight), as expected; however, PAHs were as high as 104 ppb prior to implementation of guidelines mandating increased distance between vessels and whales. While causality is unclear, the potential PAH exposure from vessels warrants continued monitoring. Historical precedent similarly emphasizes the importance of having pre-oil spill exposure data available as baseline to guide remediation goals.

1. Introduction

Marine traffic of oil tankers and other vessels to ports in Washington State and British Columbia, as well as oil transportation by rail and pipeline, increases the vulnerability of inland marine waters to a catastrophic event such as a spill or grounding. Current shipping lanes transect areas designated as critical habitat for wildlife listed as endangered and threatened under the U.S. Endangered Species Act, including the Southern Resident killer whales (SRKWs) (Ecology, 2015). This area of inland waters is partially separated from the open Pacific Ocean, with few places for the oil to disperse. Historic events (e.g., "*Exxon Valdez*" oil spill in Alaska and "*Deepwater Horizon*" oil spill in the Gulf of Mexico) have demonstrated that oil spills in the marine environment can have population-level consequences for aquatic species. Specifically, exposure to oil in marine mammals has been associated with adrenal dysfunction and increased lung disease (Schwacke et al., 2013; Venn-Watson et al., 2015), cardiac, pulmonary, adrenal and gastric lesions (Stimmelmayr et al., 2018), reduced reproductive success (Kellar et al., 2017), immune system impairment (De Guise et al., 2017), and population decline (Matkin et al., 2008). A common component of oil is polycyclic aromatic hydrocarbons (PAHs), a diverse group of compounds known to be both carcinogenic and mutagenic that rank in the top 10 hazardous substances by the United States Agency for Toxic Substances and Disease Registry (ATSDR, 2018). Marine mammal exposure to PAHs and other compounds following a spill can initially occur through inhalation as volatile components of the oil slick evaporate, followed by contact and ingestion in the water column, and persistence in the marine environment once in sediment (Rosenberger et al., 2017).

Combustion of fuel from boat motors is another source of PAHs, and an additional risk for the Southern Resident killer whales because of shipping and recreational boat traffic, ferries, fishing vessels, etc.

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commonly found in the inland waters. Marine mammals are inherently vulnerable to poor air quality due to their extended time spent at the water's surface, and their deep breaths. A previous estimate demonstrated safe pollutant (PAH) levels of vessel exhaust exposure in Southern Resident killer whales are exceeded under certain conditions (Lachmuth et al., 2011). Concerns related to vessel traffic as a risk factor for Southern Resident killer whales is not a new concept (NMFS, 2008); however much of the focus to date has been on foraging behavior (Lusseau et al., 2009) and the effects of vessel noise (Holt et al., 2009).

Previous studies on the health risks of exposure to toxic contaminants on the killer whale population have focused on legacy persistent organic pollutants (Krahn et al., 2007; Lundin et al., 2016; Ross et al., 2000), with few studies that have evaluated exposure to other environmental contaminants (Lachmuth et al., 2011; Rayne et al., 2004; Rosenberger et al., 2017). The objective of this study was to measure PAH concentrations in fecal (scat) samples from this endangered killer whale population. Floating killer whale fecal samples were collected and analyzed using gas chromatography–mass spectrometry. Results from this study indicate low current levels of PAH exposure in the whales. Prior to the change in guidelines that increased the protective distance of boats from this population, the PAH exposure may have been higher.

2. Methods

2.1. Sample collection

A total of 263 SRKW fecal samples were collected May through October from 2010 to 2013; of these, 70 samples were > 2 g, which was the minimum mass for the chemical analysis used in this study. All samples were collected within 30 linear miles of Mosquito Bay, San Juan Island, Washington, USA (Fig. 1). Samples were located by detection dogs trained to locate SRKW fecal floating on the water surface (Ayres et al., 2012; Lundin et al., 2016; Wasser et al., 2004). Samples were scooped off the surface of the water using a 1 L polypropylene beaker, collected in a 50 mL polypropylene tube, immediately centrifuged using a small field centrifuge, and all sea-water was decanted. The remaining pellet was kept on ice, frozen the day of collection, and then transferred to a −20 °C freezer until processed in the lab. Collections occurred from mid-May through mid-October, the time period when the SRKWs appear with regularity in the waters around the San Juan Islands and Puget Sound of Washington state, collectively referred to as the Salish Sea (Fig. 1).

2.2. Life history data

Age, sex, family lineage, and reproductive status of whales genotyped in this study (described below) were determined using annual population census data collected through photo-identification since 1976 by the Center for Whale Research (CWR, 2018). Age-sex class was defined as juveniles (either sex, < 10 years), adult males (≥10 years), reproductive-age females (≥10 to < 40 years) (CWR, 2018; Robeck et al., 2004), and post-reproductive females (40+ years).

2.3. Laboratory methods

Samples were thawed, homogenized, and sub-sampled for high throughput genotyping to determine individual identification (Ford et al., 2011) and for toxicant analysis. Individual identification was linked with the population census data to determine pod and age-sex class (CWR, 2018). Seventy samples were analyzed for PAHs and, as reported previously, persistent organic pollutants (POPs) (Lundin et al., 2016). These samples were extracted, purified, and analyzed for PAH concentrations using the procedures of Sloan et al. (2014). In brief, samples were extracted with dichloromethane using an accelerated

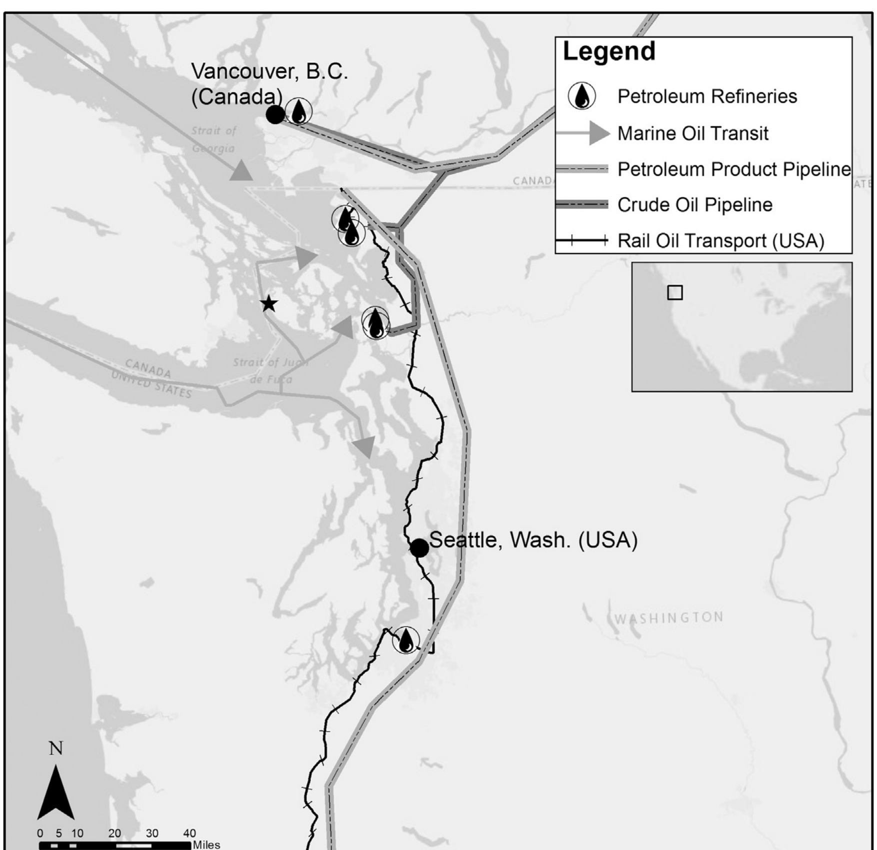

Fig. 1. Map of Southern Resident killer whale habitat within the Salish Sea, showing locations of petroleum refineries together with areas of pipeline, marine, and rail transport of oil. The star (★) marks Mosquito Bay, San Juan Island, Washington, USA; all fecal samples were collected within 30 linear miles of this point. Service layer credits: Esri, HERE, DeLorme, MapmyIndia, © OpenStreetMap contributors, and the GIS user community; Data source for map layers: https://www.eia.gov/mapslayer_info-m.php, http://www.wsdot.wa.gov/mapsdata/geodatacatalog/.