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Ion homeostasis and interrenal stress responses in juvenile Pacific herring, *Clupea pallasi*, exposed to the water-soluble fraction of crude oil

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Abstract

Juvenile Pacific herring, *Clupea pallasi*, were exposed both acutely (96 h) and chronically (9 weeks) to three concentrations of the water-soluble fraction (WSF) of North Slope crude oil. Mean (± S.E.) total PAH (TPAH) concentrations at the beginning of the acute exposure experiment were: 9.7 ± 6.5, 37.9 ± 8.6 and 99.3 ± 5.6 μg/L. TPAH concentrations declined with time and the composition of the WSF shifted toward larger and more substituted PAHs. Significant induction of hepatic cytochrome P450 content, ethoxyresorufin *O*-deethylase and glutathione-*S*-transferase activities in WSF-exposed fish indicated that hydrocarbons were biologically available to herring. Significant but temporary, elevations in plasma cortisol (4.9-fold and 8.5-fold increase over controls in the 40 and 100 μg/L groups, respectively), lactate (2.2-fold and 3.1-fold over controls in the 40 and 100 μg/L groups) and glucose (1.3-fold, 1.4-fold and 1.6-fold over controls in the 10, 40 and 100 μg/L groups) occurred in fish exposed acutely to WSF. All values returned to baseline levels by 96 h. Similar responses were seen with the first of several sequential WSF pulses in the chronic exposure study. Subsequent WSF pulses resulted in muted cortisol responses and fewer significant elevations in both plasma lactate and glucose concentrations. Hematocrit, leucocrit, hemoglobin concentration and liver glycogen content were not affected by acute or chronic WSF exposure. Plasma [Cl^-], [Na^+] and [K^+] were significantly higher in the 100 μg/L WSF-exposed group by 96 h compared to control fish, and continued to be elevated through the entire chronic exposure period. Unlike the measured stress parameters, ionoregulatory dysfunction was not modulated by WSF pulses. The results of this study suggest that chronic exposure to WSF affects at least two important physiological systems in herring: the ability of fish to maintain ion homeostasis and the interrenally-mediated organismal stress response.

Keywords: *Clupea pallasi*; Cortisol; Fish; Ions; Oil; Osmoregulation; Pacific herring; Stress; Toxicity

1. Introduction

Petroleum-derived hydrocarbons are a major contributor to the contamination of aquatic environments.

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Approximately 5 million tons of crude oil from a variety of sources enters the marine environment each year (Neff, 1990). Typical concentrations of total hydrocarbons in contaminated marine coastal waters can be as high as 80 μg/L, with occasional reports of up to 500 μg/L in the Arabian Gulf (Badawy and Al-Harthy, 1991; Madany et al., 1994; Alkindi et al., 1996). Much attention has been paid to large crude petroleum spills and their visible surface effects, however, of more recent concern are the potential effects of dissolved hydrocarbons, which are the most available to marine biota (Neff and Anderson, 1981). Of particular interest are the polycyclic aromatic hydrocarbons (PAHs), which are known to produce a myriad of lethal and sublethal effects in a wide range of biota.

The potential effects of hydrocarbons on marine benthic and intertidal organisms have been the primary focus of research to date. Notwithstanding, organisms that spend some or most of their lifecycle in the pelagic environment, such as the Pacific herring (*Clupea pallasi*), may also be negatively impacted by exposure. The acute toxicity of oil and its components have been well documented for several teleosts (Anderson et al., 1974; Rice et al., 1987) and reported effects in larval and juvenile stages include morphological, histopathological and genetic damage (Brown et al., 1996; Hose et al., 1996; Kocan et al., 1996; McGurk and Brown, 1996; Norcross et al., 1996; Carls, 1987, 1999; Heintz et al., 1999). Recently, work on the potential mechanisms underlying the common suite of PAH-induced developmental abnormalities in fish have been undertaken (Incardona et al., 2004). Still, more information is certainly needed on sublethal effects to further predictions regarding the risks of exposure to pelagic populations.

The exposure of fish to sublethal concentrations of contaminants can disturb homeostasis and impose considerable stress on physiological systems. The stress responses in teleosts is well documented and involves a series of cellular (e.g. heat shock protein production), neuroendocrine (e.g. catecholamines and corticosteroid release), biochemical (hyperlacticemia and hyperglycemia) and organismal responses (e.g. reduced growth, predisposition to disease, impaired reproduction and a reduced capacity to tolerate subsequent stress [Adams, 1990]), depending on the stressor and duration of its imposition.

Several reasons prompted an examination of the neuroendocrine and biochemical stress responses of juvenile Pacific herring exposed acutely and chronically to the WSF of crude oil. First, the paradigm of the neuroendocrine stress response is well documented in teleosts, and generally yields a consistent pattern for xenobiotic stressors. Second, fish are exposed to dissolved pollutants via an extensive respiratory surface and, in seawater, also by drinking. The high bioavailability of many chemicals in water, in combination with a variety of highly sensitive perceptive mechanisms in the integument, typically generate an integrated stress response in fish in addition to toxic effects. The ability of fish to mount an appropriate stress response, and the negative consequences associated with chronic stress, give its measurement both evolutionary and ecological significance.

2. Materials and methods

2.1. Fish

Juvenile Pacific herring (8.2 to 13.8 g) were obtained through a local supplier in West Vancouver, BC. Fish were transported to facilities at the Fisheries and Oceans Canada, West Vancouver Laboratory, BC, with a minimal use of nets to reduce trauma to the young fish. Fish were held in 500-L fiberglass tanks supplied with flowing filtered seawater, salinity 31 ppt, water temperature 11.0 ± 0.5 °C and dissolved O_2 content above 95% saturation. Following transfer, mortality in the first week was approximately 5.2%, which declined to less than 0.25% per week. When the mortality rate had stabilized, fish were acclimated for a further 4 weeks in experimental tanks before an experiment was performed. Fish were fed twice daily ad libitum with frozen krill until one day before an experiment.

2.2. Chemicals and exposure

Herring were exposed to 3 concentrations of the water-soluble fraction (WSF) of North Slope crude oil in 500-L fiberglass tanks. All exposures were performed in duplicate. WSFs were generated by seawater continuously passing through a modified apparatus developed by Carls et al. (1995, 1998),