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## THE EFFECTS OF ENVIRONMENTAL CONTAMINANTS ON IMMUNE FUNCTION AND HEALTH IN FREE-RANGING PINNIPEDS

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### Abstract

A number of marine mammal populations in Alaska have decreased in recent decades. The western stock of the Steller sea lion (*Eumetopias jubatus*) has undergone a severe decline resulting in listing as an endangered species. The northern fur seal (*Callorhinus ursinus*), with 80% of the world population breeding on the Pribilof Islands, has been designated as a depleted stock. The cause(s) of these population declines have not been discovered and several areas of investigation have yet to be thoroughly explored including the role of environmental contaminant exposure on health. Organochlorine (OC) contaminants have recently been identified to be present in the tissues of marine mammals in Alaska at concentrations higher than expected. Organochlorine contaminant exposures have been linked to immune suppression and reproductive dysfunction in marine mammals.

In field studies conducted during live-capture operations from 1995 to 2001, we investigated OC contaminant and mercury exposure along with the general health and development of immune function in juvenile northern fur seals and Steller sea lions. We optimized and validated multiple immune functional assays for use in these species, starting with the northern fur seal. These assays were then used to define each of the components of the immune system quantitatively and qualitatively in relation to age. Our approach included lymphocyte function assays [lymphoproliferative assays (specific T-cell and B-cell function), flow cytometry, IL-2 receptor expression, immunoglobulins, and specific antigen stimulation (B-cell function)]; and less specific white blood cell differential counts to demonstrate perturbations in leukocyte subpopulations and inflammatory/stress responses. These assays mainly utilized peripheral blood of free-ranging animals live-captured in Alaska, complemented with some captive animal validation.

By examining multiple cohorts of Steller sea lions from different stocks as well as repeat sampling of fur seals from birth to weaning, we documented baseline individual, stock, age-related, and stress-induced variation in responses in immune function in growing animals over time; thereby validating the use of these assays to assess the health of free-ranging otariids. We established reference ranges for normal leukocytes subpopulations for different age groups of free-ranging juveniles. Additionally, in Steller sea lions, we conducted expanded health surveys including serology,

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parasitology, bacterial cultures, viral cultures and viral PCR, fungal cultures, testing for Chlamydia by culture and PCR as well as detailed physical examinations.

These investigations detected significant correlations between OC exposure and impaired immune function at several levels including T-cell-mediated B-cell responses. Antibody production responses in fur seal pups to primary and secondary tetanus toxoid vaccinations were negatively correlated to circulating blood levels of selected polychlorinated biphenyl congeners at the time of vaccination. Developmental age could not explain this effect. Responses to mitogen stimulation using lymphoproliferative assays in fur seals and Steller sea lions were negatively correlated to PCB levels but the effects of developmental age had an impact on these results in fur seals. Total mercury concentrations in fur of fur seal pups were significantly higher than in similarly aged and older juvenile Steller sea lions. Although the total mercury concentrations were within the toxic range of terrestrial mammals, no impact of mercury exposure on health could be demonstrated in either species.

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