Project Rationale

Environmental contaminants and a changing global environment threaten the stability of natural systems, from individual physiology to the ecosystem level. These changes include increases in atmospheric CO2, toxic loading, and temperature fluctuations that lead to widespread environmental phenomena. The impacts of these anthropogenic influences are already apparent through direct and indirect effects on marine organism physiology. The following outlined research would provide necessary data for mitigating and understanding human impacts on natural ecosystems through bioindicators.

Heightened contaminant load, especially in natural systems near dense urban populations, have become increasingly problematic for marine and estuarine biota. The Puget Sound Partnership communicated a need for increased studies of toxic chemical loading in Puget Sound in their Action Agenda (Puget Sound Partnership 2009). It has recently been discovered, however, that simply monitoring water and sediment levels of toxins is not sufficient to understand the impacts they have on Puget Sound biota (WA Dept. of Ecology 2010). Unanticipated impacts on animal physiology have occurred that were not predicted by standard environmental monitoring techniques. This dearth of information points to a necessity for biological monitoring of Puget Sound and its biota (WA Dept. of Ecology 2010). An efficient and broad-scale method for biological monitoring is to assay organism physiological response to the environment. Through gene expression and epigenetic analyses of oysters, the specific stressors that are affecting Puget Sound’s inhabitants will be revealed.

The purpose of this proposed research is to ***develop biomarkers of ecosystem health that coincidentally elucidate population health and response to environmental changes***.

Oysters provide an ideal system through which to study the effects of environmental change on the environment.Oysters are well-designed to be environmental monitors of ecosystem health and the Pacific oyster, *Crassostrea gigas,* has a wealth of genomic resources, making it feasible to characterize biological effects and identify environmental threats in a controlled setting. Work is already underway to characterize *C. gigas* physiological responses to a number of environmental stressors via controlled laboratory stress challenges to create a panel of “response genes”. From this panel of genes, the *C. gigas* information will be extrapolated to environmental monitoring of ecosystem health through data from outplanted groups of sibling oysters.

Pacific oysters are sessile marine filter-feeders, so they accurately portray the contaminant and environmental regime of their habitat. In the wild, they can furnish site-specific information on environmental factors and in the laboratory their physiological responses correspond to the type and amount of stressor with which they are challenged (Todgham and Hofmann 2009). Since the physiological stress response is highly conserved evolutionarily, the response mechanisms uncovered in oysters can be extrapolated to other taxa.

The stress response as a reaction to environmental perturbation has been well documented in many organisms and is conserved across taxa (Rodriguez-Lanetty et al. 2009, Shaw et al. 2007). Populations evolve to survive and reproduce through interactions with their environment and their species community. If a perturbation causes the environment or species composition to change, a population will respond through physiological adaptations, range shifts, or extinction. Range shifts and extinction are the more extreme reactions, but there are limits to physiological adaptations based on the plasticity of the response in the individual and within the population. I propose to explore the limits of the physiological compensations and adaptations to certain environmental factors associated with climate change and environmental contaminants in order to assess future population viability. Anthropogenic impacts threaten the integral relationship between a species and its environment and thus affect overall ecosystem homeostasis.

Stress causes an energy-intensive response that deprives other physiological functions of resources. Long-term exposure to a stress could result in a population’s decreased ability to perform basic physiological functions. Since the marine environment is dynamic and susceptible to a variety of abrupt environmental changes and pollutants, there has been an increased interest in assessing the effects of multiple stresses on physiology. Eastern oysters, *C. virginica* demonstrated significantly elevated disease mortality after being exposed to multiple stressors (Anderson et al. 1998). For both hatchery practices and native population conservation, it is useful to understand the effects of synergistic stressors on population physiology.

Working Hypothesis

Contaminants and aquatic environmental regimes associated with human influence have significant effects on endemic fauna. These effects can be qualified according to varying level of impacts using gene expression assays and genetic markers.

Materials & Methods

About 100 seed *C. gigas* will be placed in mesh bags at 3 locations around Puget Sound: Samish Bay, the Seattle Aquarium, and Big Beef Creek. Over a period of 12 months, oysters will be sampled every other month. Sampling will occur at the same time on the tidal cycle and will include pulling the bags out of the water to assess incurred mortalities and remove 10 oysters for genetic analysis (gill tissue & hemocytes). At the endpoint of the experiment, all remaining oysters will be sampled for genetic analysis.

Gill tissue will be extracted for RNA and DNA. Gene expression analysis on the RNA will include a candidate gene approach based on previous laboratory trials and gene discovery. Epigenetic analysis on the oyster DNA will provide information on the environmental effects on the epigenetic profile that could change due to environmental effects.

Timeline

The experiment will run from September 2010 to September 2011. Sampling dates will be at low tide in Sept. ’10, November ’10, January ’11, March ’11, May ’11, July ’11, and Sept. ’11. Gene expression and epigenetic analyses will be ongoing during the experiment and will continue for up to a year after the last sampling date.

References

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