

EVOSTC ANNUAL PROJECT REPORT

Recipients of funds from the *Exxon Valdez* Oil Spill Trustee Council must submit an annual project report in the following format by **Sept. 1** of each fiscal year for which project funding is received (with the exception of the final funding year in which a final report must be submitted). Please help ensure that continued support for your project will not be delayed by submitting your report by **Sept. 1**. Timely receipt of your report allows more time for court notice and transfer, report review and timely release of the following year's funds.

Satisfactory review of the annual report is necessary for continuation of multi-year projects. Failure to submit an annual report by **Sept. 1** of each year, or unsatisfactory review of an annual report, will result in withholding of additional project funds and may result in cancellation of the project or denial of funding for future projects. **PLEASE NOTE:** Significant changes in a project's objectives, methods, schedule, or budget require submittal of a new proposal that will be subject to the standard process of proposal submittal, technical review, and Trustee Council approval.

Project Number: 10100839

Project Title: *Evaluating injury to harlequin ducks (Histrionicus histrionicus) caused by sublethal hydrocarbon exposure in Prince William Sound using species-specific cell lines*

PI Name:..... *Hollmen, T. and Springman, K.*

Time period covered: *October 1, 2011 - August 31, 2012*

Date of Report:..... *September 1, 2012*

Report prepared by: *Hollmen, T., Springman, K., and Riddle, A.*

Project website (if applicable): *N/A*

Work Performed:

Objective 1: *Develop harlequin duck and surrogate (mallard) cell lines to evaluate injury from site-specific hydrocarbons in harlequin ducks*

FY12 Tasks: This task was completed in FY11

Objective 2: *Develop bioassays using harlequin duck and surrogate (mallard) cell lines to assess and quantify injury due to lingering oil in PWS*

FY12 Tasks: Endpoint bioassay development

Progress:

The preliminary testing on DNA/adduct assays has been conducted. Twenty-five biopsies from harlequin ducks (field samples) were submitted. Of the 25 liver samples, 19 were from oiled sites in PWS and 6 from unoiled areas. The 19 samples are from the same or are near sites of SPMD samples taken for this project and used in EROD analyses with harlequin duck cell lines.

The biopsies were matched with seven samples produced in the lab with mallard cell lines for DNA/adduct analysis. Mallard cell pellets consisted of 2 cell controls (no dose), 1 reagent control, 1 positive control, 1 Alaska north slope crude dose, and 2 PWS SMPD doses. Cell pellets have been processed and adduct analysis of pellets and field biopsies will be conducted during fall 2012. Results should be available before the end of 2012.

Objective 3: *Evaluate injury due to site-specific lingering oil in PWS in harlequin ducks at the cellular level*

FY12 Tasks: Test PWS samples in cell lines, data analysis

Progress:

Data analysis is underway. The results to date are similar to earlier studies with fish (Springman et al., 2008) with parallel methodology and confirm earlier results concerning HADU sensitivity to EROD induction.

Objective 4: *Link analytical chemistry results from known oil-contaminated sites to injury assessments in harlequin ducks at the cellular level*

FY12 Tasks: Data analysis

Progress:

Chemical analyses of SPMD extracts are similar to those from samples taken in 2004 with the same methods (Short et al., 2008). The availability of DNA adduct analyses will allow the linkage to be made between induction and injury.

We anticipate one of the products of this research will be mathematical model(s) that describe the interaction between bioavailable PAH and resulting induction and possibly adduct formation.

Objective 5: *Develop methods to link injury due to site-specific lingering oil in PWS in harlequin duck cell lines to harlequin duck population parameters and population level impact*

FY12 Tasks: Data analysis

Progress:

This task will be completed once all laboratory data is available from bioassays.

Future Work:

We will continue data analysis as described in our work plan and proposal. We will complete work on one of our bioassays during the fall 2012. According to our study plan, we will complete the final report in 2013, and plan to prepare two manuscripts for publication in peer reviewed journals.

Coordination/Collaboration:

We continued coordination and collaboration with NCI/NIH on validation of bioassays of genetic toxicity, and with Dr Dan Esler on testing of samples from PWS. These results can be linked to Dr. Esler's work as the biopsies taken for his research in March 2009 were shared with us and used for DNA analysis.

Information Transfer:

The following abstract was presented during the reporting period:

Riddle AE, Holmén TE, and Springman K. (2011) Developing Cell Culture Methods and Bioassays to Assess Toxicological Responses in Harlequin Ducks (*Histrionicus histrionicus*). In proceedings of the 4th International Sea Duck Conference, Seward, AK September 12-16, 2011.

Literature Cited

Jeffrey W. Short, Kathrine R. Springman, Mandy Lindeberg, Jacek Maselko, Colin Khan, Peter Hodson, Margaret Krahn, Stanley D. Rice (2008). Semipermeable membrane devices link site-specific contaminants to effects: Part II - a comparison of lingering *Exxon Valdez* oil with other potential sources of cytochrome P4501A inducers in Prince William Sound, Alaska. *Marine Environmental Research* 66 (5): 487-498.

Kathrine R. Springman, Jeffrey W. Short, Mandy R. Lindeberg, Jacek Maselko, Colin Khan, Peter V. Hodson, Stanley D. Rice (2008). Semipermeable membrane devices link site-specific contaminants to effects: Part I - Induction of CYP1A in rainbow trout from contaminants in Prince William Sound, Alaska. *Marine Environmental Research* 66 (5): 477-486 .