***Understand* - Impacts of the *Deepwater Horizon* oil spill on the health and growth of estuarine fish and ecosystem functionality**

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**Background:**

Recreational fishing in the Gulf of Mexico is a billion dollar industry with over six million saltwater anglers, taking over 45 million fishing trips each year (NOAA/NMFS Statistical Data 2008). Neither the short- nor long-term impacts of the Deepwater Horizon oil spill on fishery resources are known at present; and it is a research priority to clarify this question for management. Two sportfish which represent the focus of many of the anglers are spotted seatrout (*Cynoscion nebulosus*) and red drum (*Sciaenops ocellatus*). These fish have different life history strategies, with spotted seatrout often found in shallow waters, rarely moving far from home estuaries (Patillo 1997, Hendon et al. 2002), while red drum are a more mobile species that spend a portion of their life off-shore in deeper waters with some migrating as far as 150 nautical miles (Wenner 1992). Understanding the impacts of environmental perturbations on population stability is important for both species. Yet, the different life history strategies of these two species make them a useful comparative tool, as they should react to the introduction of a potentially toxic substance in different ways.

There is also a need to improve our understanding of ecosystem-level effects of events such as the Deepwater horizon oil spill (DpH; Peterson et al. 2003). This is best accomplished by first making the link between impacts at the individual level and population-level responses in ecologically important species. The 2010 DpH oil spill and subsequent cleanup efforts introduced large quantities of crude oil and dispersant into coastal marshes of the northern Gulf of Mexico. The chronic impacts of weathered, dispersed oil on coastal fisheries are potentially very important. We initiated a study in September of 2010 to look for molecular evidence of crude oil exposure in spotted sea trout and red drum at multiple sites in MS and LA. We propose here to continue this study in 2011 in order to look for growth responses in these two species using marginal increment analysis and a comparison of growth in the spring -summer of 2010 to historical data on fish growth in the same area. These data will be valuable information for assessing ecosystem-level effects on coastal fishery production using population growth models. We will look at both short-term biomarkers and medium-term effects on growth of individual fish. We will then use the information obtained to model the potential population-level effects of the DpH oil spill.

**Objectives:**

* Measure exposure to crude oil of individual spotted sea trout and red drum at sites in Mississippi and Louisiana using molecular biomarkers (initiated in September 2010).
* Quantify the biomarker responses of spotted seatrout and red drum to weathered dispersed oil in a laboratory setting and compare to levels found in nature.
* Determine if short-term exposure to oil and dispersant have medium-term effects on estuarine fish growth and health.
* Estimate population wide effects extrapolated from individual effects.

**Approach**

**Measure fish exposure to crude oil (September 2010-May 2011)**

Spotted sea trout (n=42) and red drum (n=23) were collected at two sites in Mississippi (Grand Bay and Western Mississippi Sound) and two sites in Louisiana (Barataria Bay) in September 2010. These sites represent a range of oil exposure over the spring and summer of 2010. Sampling will be repeated in May and July of 2011 both for re-examination of molecular biomarkers and for collection of otoliths for growth measurements (See below). To assess short-term exposure to crude oil, tissue burden of hydrocarbons (PAHs), morphometric indices of development, as well as biomarker stress responses will be evaluated in multiple tissue types for all captured fish. Biomarker stress will be evaluated by quantifying the expression of Cytochrome P450-1A (CYP1A) levels and EROD activity. CYP1A is often used as a bioindicator of waterborne contaminants as its expression is a result of the fish breaking down toxic substances via the Phase 1 biotransformation pathway. Ethoxyresorufin-o-deethylase (EROD) activity is another measure of CYP1A levels and has been found to be an accurate measure of fish exposure to PAHs (Ramachandran et al. 2004). Water samples were also collected at each site on each sample day using approved methods for analysis of hydrocarbon and dispersant content.

**Measuring medium-term effects of crude oil exposure in 2011**

***Field estimates of growth and condition*** - The continuation of this work in 2011 will be focused on quantifying the relationship between hydrocarbon exposure and biomarker expression in the laboratory and investigating growth responses of both target species in the wild using otolith analysis. Growth rates of individual fish will be estimated in both species and at all four previously sampled sites in the spring and summer of 2011 based on marginal increment analysis (MIA). Marginal Increment Analysis involves the examination of growth ring width during previous growth periods (i.e., Spring-Summer 2010). We will use MIA to examine growth of age-0 and age-1 fish during the 2010 growth ring season when fish were potentially exposed to oil and dispersant. These data will be compared across sites in MS and LA, as well as to historical data to test the null hypothesis of no difference between pre and post-DpH growth rates. Additionally, various morphometric indices will be used to evaluate fish health and condition in 2011.

 ***Laboratory assays of response to oil exposure*** - Laboratory assays of survival and growth as a function of dispersed oil concentration will be conducted at Gulf Coast Research Laboratory (GRCL) using spotted seatrout from the GCRL spotted seatrout hatchery and red drum from the CCA/CPL Marine Development Center in Corp us Christi, TX. Crude oil and dispersant (Corexit 9500) will be obtained from British Petroleum. Dispersed oil will be produced for experiments by mixing crude oil with Corexit 9500 in seawater for 96 hours creating a chemically enhanced water accommodated fraction (CEWAF). Juvenile fish will be exposed to dilutions of produced CEWAF (initially 1.0% and 0.1%) for 14 days in a flow-through system (N = 4 tanks per treatment, 10 individuals per tank). Individuals will be measured and weighed prior to being randomly assigned to one of the exposure tanks. At the conclusion of the experiment, individuals will be removed, weighed and measured, sacrificed by immersion in MS222, and liver, brain, and gonad removed. Individual organs will be frozen at -80C for storage. Expression of several genes expected to be responsive to hydrocarbon exposure including CYP1A, CYP19, and Vitellogenin will be assessed bu qPCR. Where necessary, the relevant sequences will be obtained by cloning and sequencing following standard procedures.

**Expected outcomes**

The results of this study will be a test of the hypothesis that spotted sea trout and red drum exposed to waterborne hydrocarbons, exhibited depressed growth in 2010 and decreased health as compared to fish from clean environments. The results of the laboratory assays will be analyzed in conjunction with the field results to establish a link between individual exposure and population-level response to the oil spill. These data will be an important piece for understanding the impacts of the DpH spill on spotted seatrout and red drum sustainability, as well as overall estuarine ecosystem functionality. Completion of this research will contribute to the body of knowledge pertaining to the ecological effects of the oil spill. This information is critical for the development and implementation of an informed and scientifically sound response to the disturbance of sensitive habitats globally. These outcomes will be important for fishery managers across the Gulf of Mexico, as well as in other regions of the world where oil drilling may pose a potential threat to healthy fisheries.

This project will also facilitate the training of a graduate student (MS, R. Brewton) who is conducting the work as her thesis research.

**Literature Cited:**

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