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## Gulf Oil Spill: UAB Department of Biology's Approach

Toral Patel

The 2010 Gulf of Mexico Oil Spill, also known as the Deep-water Horizon Oil Spill, lasted 3 months and resulted in over 185 million gallons of crude oil escaping into the ocean. The spill has caused damage to the Gulf's fishing and tourism industries, as well as to marine and wildlife habitats. The three fundamental strategies to address the spilled oil were to contain it on the surface, to dilute and disperse it, and to remove it from the water. By July 2010, oil on the surface of the Gulf had largely dissipated, but the concern still remains for underwater oil and ecological damage.

Factors such as petroleum toxicity, oxygen depletion, dispersant compounds, and crude oil components threaten the environment and wildlife. Researchers have begun to monitor turtles, coral reef, microbes, crustaceans, fish, mollusks, and birds. The UAB Department of Biology has proposed to investigate the effect of the crude oil and remnants on organisms ranging from sea grass and microbes to fish and turtles.

### Oil Degrading Bacteria

Dr. Asim Bej, from the Department of Biology, and Dr. Elizabeth Gardner, from the Department of Justice Sciences, are studying the effect of the oil spill on the response of oil degrading bacteria. While marine biology will consume the remaining oil over the next years, Nutrient Enhanced Bioremediation (NEB) can accel-

erate this process without negatively impacting the environment. Bioremediation is the process of using microorganism, in this case oxidative hydrocarbon degraders (OHD), and their biological systems to destroy contaminants in soil and water. In order to obtain data on the effectiveness of NEB, Dr. Bej and Dr. Gardner will be establishing baseline measurements from the affected areas, determining the optimal nutrient concentration to culture beneficial OHD, and applying the OHD to the affected coastal areas. Finally, they will monitor the OHD populations and degradation of crude oil.

Concurrently, Dr. Bej and Dr. Gardner will investigate the role of oil contamination and dispersant on pathogenic *Vibrios* (PV). Some of these PVs are oil consuming bacteria that cause seafood born infections and are found in oysters, shrimp, and crabs. The use of the dispersants has increased the surface area of oil micelles, allowing oil degrading bacteria to enhance the degradation process. The contamination increases pathogens that affect the Gulf of Mexico ecosystem as well as human health. PVs are studied using the *Bacterial Analytical Manual* (BAM) with molecular methodologies from oil affected sediments and oysters that will be compared to samples collected from unaffected locations. This data will allow scientists to assess the ecological impact of the oil and dispersants, provide information to guide the recovery process to natural conditions in the Gulf, and prevent human health consequences.

## Turtlegrass

*Thalassia testudinum* (turtlegrass) is the largest of the seagrasses and a vital component of the Gulf of Mexico's unique ecosystems. Turtlegrass is very effective in stabilizing sediments, inhibiting erosion, producing oxygen and organic matter, regenerating nutrients, serving as a food source, and providing a nursery for many fish and shellfish. The hydrophobic oil molecules can limit the growth and survival of the seagrass by multiple mechanisms. Dr. Karolina Mukhtar, from the Department of Biology, and collaborators Drs. Shahid Mukhtar and Stephen Watts (Department of Biology), and Dr. Stephen Barnes (Department of Pharmacology and Toxicology), are evaluating the molecular response of the turtlegrass to sublethal concentrations of crude oil and dispersants. They will evaluate bioaccumulation patterns in plant tissues and the immunological stress response to the contaminants, dispersants, and byproducts by comparing samples from unaffected areas to samples from heavily impacted areas by the spill.

In order to determine the extent of the impact of petroleum hydrocarbons and dispersant components on turtlegrass systems, Dr. Mukhtar et al, will evaluate the biomass production of the turtlegrass as well as the photosynthetic rates correlated to the bioaccumulation of toxicants. Petrochemicals are able to penetrate through the lipid bilayer and accumulate on the thylakoid membranes of the chloroplasts, reducing photosynthetic efficiency contents of anthocyanins, ascorbic acid, and riboflavin; expression levels of genes coding for enzymes involved in their biosynthesis will be analyzed to reflect the degree of environmental pollution. These experiments will provide necessary information on how the seagrasses respond to oil and dispersants, insights into the long-term effects of the toxicant exposure, and possible remediation strategies.

## Zebrafish

To screen for low level toxicity of dispersants and residue oil, Dr. Mickie Powell from the Department of Biology and collaborators Dr. Stephen Watts and Dr. Vithal Ghanta (Department of Biology), Dr. Alex Szalai (Department of Medicine), and Dr. Bob Peters (Department of Engineering) have proposed the use of molecular biomarkers in zebrafish. Non-lethal levels of crude oil and bioremediation by-products can negatively impact organisms through physiological, biochemical, and pathological changes. Animals respond to environmental stresses by changes in the expression of different stress proteins, such as the C-reactive protein (CRP). Increased serum levels of CRP can identify a response to bacterial toxins and other inflammatory agents. Dr. Powell et al, have previously identified several stress-inducible mRNAs in zebrafish, including CRP, vitellogenin, and serum amyloid A (SAA). They have also confirmed the increased expression of their mRNAs in response to injection of bacterial endotoxin.

The levels of CRP, SAA, and vitellogenin gene expression will

be measured in zebrafish exposed to crude oil contaminants and dispersants. Dose-dependent response curves will be generated to quantify the response to environmentally relevant exposure levels. These same proteins will be analyzed in fish exposed to remediated water to test the effectiveness of various bioremediation techniques. The data collected will form the basis for the development of techniques to monitor wild populations of fish for possible toxin exposure (acute, chronic, and recovery post) in the future.

## Blue Crab

In order to understand the biological consequences of the 2010 Deepwater Horizon oil spill, it is important to determine the effects of endocrine disrupting compounds (EDCs) present in crude oil and dispersant on aquatic animals. EDCs are environmental pollutants that mimic or block the effects of naturally occurring hormones. This has adverse effects on the organisms, their offspring, or both. Constituents of oil spilled into the Gulf of Mexico and chemical dispersants used in remediation are known and are potential EDCs. Dr. Douglas Watson from the Department of Biology, along with collaborators Dr. Helen Kim (Department of Pharmacology and Toxicology) and Dr. Christine Duarte (Department of Biostatistics), are testing the effects of constituents of crude oil (polycyclic aromatic hydrocarbons, PAHs) and chemical dispersant (Corexit 9500A) singly and in combination on markers of endocrine disruption in blue crabs (*Callinectes sapidus*).

They hypothesize that these constituents of oil and dispersant will act individually as endocrine disruptors and that dispersant will enhance the uptake and bioavailability of oil and oil byproducts, thus making animals more susceptible to the effects of the compounds. The overall approach by Dr. Watson and colleagues will be to assess established markers of endocrine disruption and proteomic changes that may represent previously unrecognized responses to the oil and dispersants. The blue crab is both economically and ecologically significant to the Gulf of Mexico. The research is designed to determine the effects of oil constituents and dispersant on this key sentinel species in the Gulf of Mexico and, in future experiments, to permit assessment of the impacts of the oil spill in wild populations during remediation and recovery.

## Turtles

To lessen the impact of the oil spill on ecosystems, the National Oceanic and Atmospheric Association (NOAA) and the U.S. Fish and Wildlife Service (USFWS) decided to relocate all loggerhead sea turtle eggs from the beaches in Alabama and Florida panhandle. All eggs were moved to the mid-Atlantic coast of Florida so that the hatchlings could enter the Atlantic Ocean. Dr. Thane Wibbels and Jenny Estes (Ph.D. student) from the Department of Biology plan to evaluate the genetic implications of translocating the loggerhead eggs and relate it to future ecological and conservational translocations. DNA sequence analysis on the tissues will be conducted and compared

to previously published data on loggerheads nesting along the mid-Atlantic coast of Florida. By performing genetic analysis of tissue samples from hatchlings, Dr. Wibbels and Estes will be able to evaluate if the translocation strategy is altering the genetic composition of loggerhead subpopulations inhabiting the coastal waters of southeastern U.S.

A key species in salt marsh habitats of the Gulf of Mexico are diamond back terrapins. These turtles are pivotal in the diversity of animals and plants found in the salt marsh. Terrapin populations are sensitive to contamination in the salt marsh. They also consume prey that accumulate PAHs (i.e. clams and mussels)

and store the PAHs in their fatty tissues for prolonged periods, increasing their toxicological exposure. Dr. Wibbels and Dr. Ken Marion (Department of Biology) propose to evaluate physiological markers that document the impact of oil contamination on the terrapins. PAHs will be analyzed in both tissue and blood samples and hormone systems that respond to chronic stress. Terrapins will also be marked in order to be repeatedly recaptured and resampled over a 1-year period to assess chronic oil exposure effects. Data from these experiments will generate a baseline on the impact of oil on the terrapins, as well as facilitate long-term studies on the effects of the oil spill on the ecology and survival status of the terrapins.