

## **OVERVIEW**

### **Environmental Technology**



The health of marine ecosystems is affected by societal activities including pollution, global warming, fishing, dredging, and transportation. In 1999 the National Research Council recommended a renewed effort to understand the health of the ocean, its effects on humans, and possible human health threats. Equally important is to gain a better understanding of the beneficial use of marine biodiversity to human health and the exploration and development of biomedical products from the sea. Consistent with the objectives of the NOAA Oceans and Human Health Initiative, Woods Hole Sea Grant has focused its efforts in Environmental Technology on developing innovative ways to monitor environmental changes in coastal habitats specifically linked to consequences related to harmful algal blooms, toxicology, climate change and eutrophication.

#### **Why Environmental Technologies?**

Over the past three decades, Woods Hole Sea Grant has invested in research, extension, and outreach in the programmatic theme area of Environmental Technology. With the increasing concern of the role of the oceans in human health and the need to monitor global scale changes in climate and biodiversity, this investment has resulted in the development of new tools for assessing the distribution of pathogens and harmful algal blooms in coastal waters, for understanding the relationship between toxic chemicals and reproductive success of marine animals, and for remotely monitoring the marine environment.

A recent study conducted by the Donahue Institute at the University of Massachusetts values the marine science technology industry in New England at \$4.8 billion, with specific applications directed at homeland security, oceanographic and atmospheric monitoring systems, and environmental monitoring. The marine science industry in New England has grown into a robust economic cluster that stretches across five coastal New England states, with 481 marine science firms employing nearly 40,000 people. The report suggests that continued investment



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in research partnerships and funding is the catalyst necessary to continue the growth of the marine science technology industry in New England. Sea Grant's investment in Environmental Technologies is a critical part of the research enterprise, especially for Southeastern Massachusetts.



#### **Woods Hole Sea Grant Involvement**

Woods Hole Sea Grant's Environmental Technology theme focuses on the initiation of research projects that will develop and deploy innovative technologies to address specific problems in coastal ecosystems, and the development of extension and outreach activities to foster information transfer, education, and development of new monitoring and treatment technologies. *Thematic components include:* 

- Marine biotechnology, including the development of molecular markers for understanding contaminant effects in the environment and probes for application to ecological processes;
- Remediation technologies, including the development of new approaches to understanding the degradation of contaminants in the environment; and
- Remote technologies for monitoring the marine environment.



## **INVESTMENT**

## Woods Hole Sea Grant's Investment, 2000–2006

Research Project Title	P.I.(s)	Years Funded
Detection and Quantification of Live Acanthamoeba in Natural Marine Ecosystems Using Molecular Genetic Methods (R/B-147)	Becky Gast, Woods Hole Oceanographic Institution	2000–2001
Reducing the Risk of Open Ocean Aquaculture Facilities to Protected Species (R/M-43)	Walter Paul, Woods Hole Oceanographic Institution	2000–2001
Estradiol Dynamics: A Molecular Basis for Potential Endocrine Disruption in Marine Mammals (R/B-162)	John Stegeman, Woods Hole Oceanographic Institution	2000–2002
Impact of Environmental Contaminants on Aquatic Birds: The Molecular Basis of Differential Dioxin Sen- sitivity (R/P-64)	Mark Hahn, Woods Hole Oceanographic Institution	2000–2002
Environmental Contaminants in Aquatic Birds: Novel Biomarkers of Dioxin Effects (R/P-64)	Mark Hahn, Woods Hole Oceanographic Institution	2000–2002
Bivalve Dispersal as Indicated by Shell Trace Element Composition (R/0-32)	Lauren Mullineaux and Stan Hart, Woods Hole Oceanographic Institution	2000–2002
Ligand Screen for Orphan Receptors in Marine Animals (R/P-66)	Mark Hahn and Sibel Karchner, Woods Hole Oceanographic Institution	2002–2004
Contaminants and Aquatic Animals: A Biomarker to Assess Species Differences in Susceptibility to Di- oxin-like Chemicals (R/P-67)	Mark Hahn, Woods Hole Oceanographic Institution	2002–2004
Estrogen- and Aryn Hydrocarbon-receptor Mediated Reproductive Effects and Adaptions in the Marine En- vironment (R/P-68)	Gloria Gallard, Boston University	2002–2004
Detection and Enumeration of Harmful Algal Bloom Species Using Fiber Optic Microarrays (R/P-69)	Don Anderson, Woods Hole Oceano- graphic Institution; and David Walt, Tufts University	2003–2005
A Proteomics Approach to the Study of Endocrine Disruption in Fish (R/B-169)	John Stegeman and Joanna Wilson, Woods Hole Oceanographic Institution	2004–2006

In addition to the formal projects identified above, program development funds ("New Initiatives") have also been used to initiate new projects, provide opportunities for students to attend conferences to present their work, respond to emergency funding requests, jump-start a research program or test a new technique, and support workshops relevant to the research conducted under this theme.



#### Research Support, 2000–2006

 Sea Grant Funds:
 \$1,474,587

 Matching Funds:
 \$ 838,492

TOTAL: \$2,313,079

#### Student Support, 2000-2006

Graduate Student Support: 8 Students • 59 Months' Student Support





Scott Gallager

Don Anderson and Deanna Erdner



## **IMPACTS**

#### Research Dividends, 2000-2006

#### Research Highlights

Research projects supported in 2000-2006 have yielded numerous advances and discoveries within the scientific community. A few highlights include:

- Woods Hole Sea Grant researchers created a tissue bank of marine mammal samples that has allowed scientists around the world to evaluate the pervasive problem of persistent organic contaminants in cetaceans. In Woods Hole, researchers are exploring the contaminant burden and effects of organo-halogen compounds on endangered species of cetaceans, providing a better understanding of the mechanisms underlying endocrine disruption in cetaceans.
- Molecular probes that differentiate and enumerate species responsible for harmful algal blooms (HABs)
  have been developed facilitates the rapid identification of bloom conditions and the potential threat
  of toxin accumulation in shellfish stocks. The probes have been commercialized by Saigene, Inc., and
  were very effective in predicting the extensive red tide conditions experienced off the New England
  coast during the spring and summer of 2005.
- Advances in biotechnology have resulted in the development of sensitive indicators of environmental
  risk of contaminant exposure in marine animals. This approach has been applied to the study of aquatic
  birds, marine mammals, fish and marine invertebrates. These studies have given scientists tools with
  which they can assess the comparative sensitivity of different species within an ecosystem and the
  sensitivity of endangered species to chemical contaminants in their habitats.



## The Link Between Environmental Contaminants and Animal Susceptibility by Tracey Crago

With Sea Grant support, WHOI toxicologist Mark Hahn and former graduate student Brenda Jensen examined the potential toxicity of dioxin, a planar halogenated aromatic hydrocarbon (PHAH), in beluga whales.

Beluga tissues are known to accumulate PHAH. Exposure may result in immuno-suppression, reproductive problems, and cancers. Beluga whales from the St. Lawrence Estuary are among the most affected marine mammals; dioxin contamination has been blamed for the failure of the population to recover from hunting-related declines at the beginning of the 20th century.

Yet until scientists can provide a mechanism and show a cause and effect relationship, said Jensen, the



Mark Hahn and Sibel Karchner

presence of pollutants is merely coincident with observed health problems. "Many feel that chemicals may contribute," she said, "but the trick is to establish that link."

Using the aryl hydrocarbon receptor (AhR), present in most vertebrate species, the team sought to understand the cellular-level processes that make animals sensitive to PHAH exposure. The more tightly a species' AhR binds to a contaminant, the greater its sensitivity. Measuring the binding affinity of beluga AhR to dioxin, Hahn and Jensen's results suggest that belugas are highly sensitive to contaminant exposure, much more so than humans.

"The levels of PHAH contaminants in the marine environment are rarely high enough to kill an animal outright," said Jensen, but the findings are consistent with high disease incidence in St. Lawrence belugas. "These contaminants exert their effects by interrupting other processes and, sometimes, cause them to fail." Their work, said Jensen, "may be a key for understanding the susceptibility of a species to PHAH, which, in turn, may help us better understand the health ramifications of the contaminant burdens we see in animals and in the environment."



### A Shell is Worth a Thousand Maps

by Tracey Crago

A project involving WHOI biologists and geochemists has resulted in a promising method for identifying the source habitat for soft-shell clams-right down to the specific geographic location of their spawning grounds. Like many of Nature's wonders, the answer to where clamsand presumably other bivalves—originate has been there all along, in their shells.

Biologists Lauren Mullineaux and Susan Mills incubate hatchery-spawned soft-shell clam embryos (Mya arenaria) in clean seawater (the control) and seawater spiked with trace elements. Within 36 hours, larval shells form. Once collected—a lengthy and painstaking process—the



Microscopic laser tracks on a juvenile Mya arenaria clam. A mass spectrometer is used to determine the chemical signature of the ablated shell material.

shells can be analyzed for evidence of trace metals. That analysis is made possible using two tools: the Inductively Coupled Plasma Mass Spectrometer (ICP/MS for short), and the ion microprobe. Each has a specialty: the ICP/MS can measure bulk samples and gives results for different elements at the same time, whereas the ion microprobe can more precisely detect the location in the shell of each element, though it can only look for one element at a time.

Investigators documented location-specific elemental signatures in the shells of clams collected from multiple field sites in New England and found barium, strontium, cobalt, magnesium, manganese, and lead most useful for determining trace-element fingerprints. In one experiment, researchers compared clams from a known polluted area, Neponset Harbor, just south of Boston, to clams from a relatively clean harbor, Barnstable Harbor on Cape Cod. They saw lead levels 10 times higher in Neponset clams than those from Barnstable, and a cobalt reading twice as high in Neponset as Barnstable. As a bonus, their results dovetailed nicely with a U.S. Geological Survey map of trace element distributions in sedi-

ments along the Massachusetts coastline.

Investigators say direct economic benefit is likely to result from two applications of their results. One, using the findings to calculate connectivity among soft-shell clam populations in geographically separate bays in New England, "will allow local shellfish managers to determine the potential effects of regional populations on their local stocks, and vice versa," says Mullineaux. Another benefit, she says, "is a deeper understanding of the physical and biological processes that control exchange of larvae and recruits among marine populations," information she says is needed for critical evaluation of plans for marine reserves.



Lauren Mullineaux (left) and Susan Mills

Currently, doctoral student Carly Strasser is working on a follow-up project estimating the maternal contribution of trace elements in the larval shell. The Sea Grant project also helped investigators secure funding for a large bi-coastal, multidisciplinary study of connectivity in coastal bivalve populations, now underway.





#### **PROFILE - Bill Martin**

When 1998 presidential candidate George H. W. Bush declared Boston Harbor to be "the filthiest in America," its sediments and water quality were already under scientific scrutiny. Today, nearly four years after the region's sewage discharge was diverted from the harbor through an outfall pipe extending nine miles into Massachusetts Bay, scientists studying the harbor wonder if the situation has improved.

What do the sediments near the original discharge site look like today? And what about sediments near the new discharge site?

To find out, Bill Martin is heading a WHOI Sea Grant-supported team of investigators sampling harbor and bay sediments. Martin's group, including WHOI colleague Roger Francois and MIT–WHOI Joint Program graduate student Linda Kalnejais, joined forces with a USGS Woods Hole team. The partnership brought historic baseline information to the project, showing decreasing heavy metal concentrations in harbor sediments from the late 1970s through the early 1990s.

"Our recent data show that the trend continues," says Martin. But are the polluted sediments simply being diluted, or are the metals going elsewhere? "That's something our Sea Grant study should be able to quantify."

Sediment cores were taken from Hull Bay, in Boston Harbor, and a Massachusetts Bay site just west of the outfall; both solids and sediment pore waters were analyzed. Both, Martin hopes, will lead to a better understanding of heavy metal cycling in sediments.

Kalenjais undertook the time-consuming analysis of sediment samples. Initially, investigators focused on the exchange of dissolved metals between sediments and seawater, measuring heavy metal concentrations in the pore waters and in sediment incubators on the seafloor, known as in situ benthic flux chambers. This method allows for direct estimates of the transport of metals from sediments to the overlying water.

Next, Martin's team looked at metal cycling within the sediments themselves. They found metals concentrating near the sediment surface, possibly allowing them to be re-suspended and transported to other sites. Recently, sediment sampling began in Cape Cod Bay. Based on storm, wind, and sediment transport patterns, Martin believes the bay could accumulate metals remobilized from the Boston Harbor site. "What we find out will help us project the effects of anthropogenic metal release in the region," says Martin. "And that information should be applicable to other urban coastal areas—a key goal of this project."



Bill Martin (right) and Mike Bothner (USGS) recover push cores from a Sea Grant-supported study of trace metals in surface sediments. Since 2000, sewage discharge re-routed from Boston Harbor has been pumped 14 km offshore into Massachusetts Bay. Baseline data collected prior to the outfall completion will be compared to data from this study to determine possible links between sewage output and metal accumulation near the outfall site.



#### Research Publications, 2000–2006

# Isolation and Characterization of Two Cytochrome P450 Aromatase Forms in Killifish (*Fundulus heteroclitus*): Differential Expression in Fish from Polluted and Unpolluted Environments

Greytak, S.R., D. Champlin, and G.V. Callard *Aquatic Toxicology*, Vol. 71, pp. 371-389, 2005 WHOI-R-05-001

# Cytochrome P450 1A1 and Aromatase (CYP19) in Cetaceans: Enzyme Expression and Relationship to Contaminant Exposure Wilson, J.Y.

Doctoral Dissertation, MIT/WHOI Joint Program in Oceanography/Applied Ocean Science and Engineering, 260 pp., 2003 WHOI-X-03-003

#### Biomarkers and Bioassays for Detecting Dioxin-Like Compounds in the Marine Environment Hahn, M.E.

Elsevier, The Science of the Total Environment, Vol. 289, pp. 49-69, 2002 WHOI-R-02-001

# cDNA Cloning and Characterization of an Aryl Hydrocarbon Receptor from the Harbor Seal (*Phoca vitulina*): A Biomarker of Dioxin Susceptibility?

Kim, E.-Y. and M.E. Hahn *Elsevier, Aquatic Toxicology*, Vol. 58, pp. 57-73, 2002 WHOI-R-02-002

### cDNA Cloning of an Aryl Hydrocarbon Receptor from Baikal Seals (*Phoca sibirica*)

Kim, E.-Y., M.E. Hahn, H. Iwata, S. Tanabe, and N. Miyazaki

Marine Environmental Research, 5 pp., 2002 WHOI-R-02-004

# Use of Stable Isotopes to Investigate Individual Differences in Diets and Mercury Exposures Among Common Terns *Sterna hirundo* in Breeding and Wintering Grounds

Nisbet, I.C.T., J.P. Montoya, J. Burger, and J.J. Hatch

Marine Ecology Progress Series, Vol. 242, pp. 267-274, 2002 WHOI-R-02-005

#### cDNA Cloning and Characterization of a High Affinity Aryl Hydrocarbon Receptor in a Cetacean, the Beluga, *Delphinapterus leucas*

Jensen, B.A. and M.E. Hahn *Toxicological Sciences*, Vol. 64, pp. 41-56, 2001 WHOI-R-01-007

#### Development of an Acanthamoeba-specific Reverse Dot-Blot and the Discovery of a New Ribotype

Gast, R.J.

*J. Eukaryot. Microbiol.*, Vol. 48, No. 6, pp. 609-615, 2001 WHOI-R-01-009

# Towards Molecular Understanding of Species Differences in Dioxin Sensitivity: Initial Characterization of Ah Receptor cDNAs in Birds and an Amphibian

Karchner, S.I., S.W. Kennedy, S. Trudeau, and M.E. Hahn

Marine Environmental Research, Vol. 50, pp. 51-56, 2000 WHOI-R-00-001

#### Relative Contributions of Affinity and Intrinsic Efficacy to Aryl Hydrocarbon Receptor Ligand Potency

Hestermann, E.V., J.J. Stegeman, and M.E. Hahn *Toxicology and Applied Pharmacology*, Vol. 168, pp. 160-172, 2000 WHOI-R-00-002

# Cytochrome P4501A Induction and Porphyrin Accumulation in PLHC-1 Fish Cells Exposed to Sediment and Oil Shale Extracts

Huuskonen, S.E., A. Tuvikene, M. Trapido, K. Fent, and M.E. Hahn

*Arch. Environ. Contam. Toxicol.*, Vol. 38, pp. 59-69, 2000 WHOI-R-00-003

# Serum Alters the Uptake and Relative Potencies of Halogenated Aromatic Hydrocarbons in Cell Culture Bioassays

Hesterman, E.V., J.J. Stegeman, and M.E. Hahn *Toxicological Sciences*, Vol. 53, pp. 316-325, 2000 WHOI-R-00-004



Identification of Cytochrome P450 1B-like Sequences in Two Teleost fish Species (Scup, Stenotomus chrysops and Plaice, Pleuronectes platessa) and in a Cetacean (Striped Dolphin, Stenella Coeruleoalba)

Godard, C.A.J., M.J. Leaver, M.R. Said, R.L. Dickerson, S. George, and J.J. Stegeman *Marine Environmental Research*, Vol. 50, pp. 7-10, 2000 WHOI-R-00-008

Cellular Localization of CYP3A Proteins in Various Tissues from Pilot Whale (*Globicephala melas*) Celander, M.C., M.J. Moore, and J.J. Stegeman *Environmental Toxicology and Pharmacology*, Vol. 8, pp. 245-253, 2000 WHOI-R-00-009

# Identification, Functional Characterization, and Regulation of a New Cytochrome P450 Subfamily, the CYP2Ns

Oleksiak, M.F., S. Wu, C. Parker, S.I. Karchner, J.J. Stegeman, and D.C. Zeldin *The Journal of Biological Chemistry,* Vol. 275, No. 4, pp. 2312-2321, 2000 WHOI-R-00-010

# The Ah Receptor: Comparative Biochemistry and Possible Role as a Biomarker of Susceptibility to PHAH

Hahn, M.E., B.A. Jensen, and E.-Y. Kim In: Endocrine Disruptors in the Marine Environment: Impacts on Marine Wildlife and Human Health. Proceedings of the Atlantic Coast Contaminants Workshop, pp. 120-126, 2000 WHOI-R-00-012

#### Mechanisms of Action for Aryl Hydrocarbon Receptor Ligands in the PLHC-1 Cell Line

Hestermann, E.V.

Doctoral Dissertation, MIT/WHOI Joint Program in Oceanography/Applied Ocean Science and Engineering, 193 pp., 2000 WHOI-Y-00-001

# Characterization of an Aryl Hydrocarbon Receptor from a Cetacean: An Approach for Assessing Contaminant Susceptibility in Protected Species Jensen, B.A.

Doctoral Dissertation, MIT/WHOI Joint Program in Oceanography/Applied Ocean Science and Engineering, 199 pp., 2000 WHOI-Y-00-002

## Extension and Outreach Publications, 2000–2006

New Tools for Assessing Water Quality: Stable Isotope Analysis of Nutrients

Woods Hole Sea Grant Focal Point, 3 pp., 2002 WHOI-G-02-001

Theme Booklet: Environmental Technologies Woods Hole Sea Grant 4 pp., 2000 WHOI-G-00-006