Industry Partnerships to Deploy Environmental Technology
"From Bench to Bayou: Leveraging Aquatic Model Ecosystems into a National Laboratory for Environmental Signaling and Sensors"

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INTRODUCTION

The Center for Bioenvironmental Research (CBR) at Tulane and Xavier Universities through its DOE/EM Grant #DE-FG01-93EW53023 has been doing research on "Hazardous Materials in Aquatic Environments of the Mississippi River Basin" (EM-Aquatic). During the past six years, the CBR has developed, through DOE partnership, a unique ***natural laboratory*** located in the bayou country of Louisiana to study remediation, fate and transport, toxicity, ecosystem assessment, ecology, and pollution prevention in the environment. The result is a unique model ecosystem that can provide applied holistic environmental assessment and remediation tools not only for coastal wetland ecosystems of Louisiana but for contaminated DOE sites throughout the country. Through novel approaches using receptor-based methods, exploitation of **natural remediation** processes and environmental signaling, the CBR has provided the DOE with the power to harness this knowledge and provide new solutions to truly accelerate its paths to closure on environmental management concerns.

Bayou Trepagnier, chosen as the focal sampling site for this project, not only contains hydrocarbons known to cause endocrine disruption, but it also receives large chlorine discharges from a sewage treatment plant. To date, identification, characterization, and monitoring of contaminants in Bayou Trepagnier has been conducted, as well as the characterization of histopathological, physiological, and molecular biomarkers of exposure. Already, specific research projects within the task areas of this project have developed into usable technologies for DOE and other government and private entities in

the areas of biosensor development, biomarker identification, fate and transport modeling, and remediation. Future work will include novel approaches using receptor-based methods and environmental signaling. The use of receptor-based methods, such as endocrine disruption, to perform hazard monitoring has novel advantages compared to traditional methods used for ecological and human health risk assessment. Leveraging upon the CBR's experience with its Bayou Trepagnier model ecosystem and its internationally renowned expertise on environmental signaling the CBR will collaborate with the DOE's national labs' **Environmental Health Initiative** to enhance the scientific basis for environmental decisions and prevent and predict environmental threats to human, wildlife, and ecosystem health. In addition, the CBR will continue to develop innovative technologies and transfer them through cooperative programs to the DOE and other government and commercial sectors.

The CBR and DOE are poised to take this information and technology, provide for field collections and data synthesis and apply it to DOE sites, exploiting the benefits of a "mature" model ecosystem. While using Bayou Trepagnier as the project's *'natural laboratory'*, CBR will be able to correlate the data obtained from the bayou with data from bodies of water near DOE sites that exhibit similar characteristics and contain similar contaminants. Through transitional funding, specific research projects within focused task areas will develop into usable technologies for DOE and other government and private entities, facilitating technology transfer through cooperative programs.

PROJECT DESCRIPTION

OBJECTIVES AND SIGNIFICANCE

The CBR's intent is to continually build upon the CBR's past accomplishments (Appendix A) through it's EM-Aquatic grant and provide for research applications to an aquatic model ecosystem developed in partnership with the DOE. This integrated program of basic and applied research, technology development, and technology transfer will support the Department of Energy's (DOE) Environmental Restoration and Waste Management (EM) programs relating to aquatic systems. In this effort, the project will, in general:

- Provide models to exemplify how the clean-up/technology development process can be guided through ecological and risk analyses.
- Undertake a research program for the development of new receptor based technologies related to hazard monitoring that will provide more cost-effective solutions to environmental restoration and waste management problems facing the DOE complex.
- Provide a better understanding of the problems associated with contaminated aquatic systems, including endocrine disruption, thereby providing new insights

that will lead to the development and use of highly innovative technologies to assess risk, to perform environmental monitoring, and to be used in environmental clean-up.

General Scope, Considerations and Directions

Tulane and Xavier Universities have singled out the environment as a major strategic focus for research and training for now and into the next century. The CBR was established in 1989 as the umbrella organization to coordinate environmental research at both universities. In the original DOE/EM 5-year grant awarded to the CBR in December 1992, the project was a broad research and education program aimed at elucidating the nature and magnitude of toxic materials that contaminate aquatic environments of the Mississippi River Basin. The project was intended to be aquatically oriented, have relevance to DOE, but at the same time be specific to the aquatic environment in Louisiana, and focus on the fate of contaminants in the ecosystem in terms of natural degradation.

The studies within the original project have defined: 1) the complex interactions that occur during the transport of contaminants, 2) the actual and potential impact on ecological systems and health, and 3) the mechanisms through which these impacts might be remediated. This project is particularly relevant to the U.S. Department of Energy's Environmental Restoration and Waste Management programs aimed at solving aquatic pollution problems in the DOE complex.

Based on findings in the original 5-year grant, the CBR proposes to focus the program on a robust model ecosystem to serve as a natural laboratory: Bayou Trepagnier. The bayou and spoil banks have been mapped and analyzed in terms of risks associated with the levels of hydrocarbons and metals at specific sample sites. These data, coupled with plume characterization data, indicate that Bayou Trepagnier is a model system for understanding how populations of fish, amphibians, and plants respond to long term hydrocarbon and metals contamination. The hydrocarbon plume, characterized in Bayou Trepagnier through fate and transport studies, is composed of a variety of polyaromatic hydrocarbons (PAHs) that are not only toxicologically hazardous, but also bind receptors related to endocrine functions. Such binding can cause disruptions of endocrine functions in fish and amphibians, resulting in adverse effects on development and reproduction. Studies concerning how these PAHs bind specific target receptors not only provide information about previously undefined ecotoxic effects, but also serve as the basis for new technologies for hazard monitoring and remediation.

Environmental Signaling and Receptor Based Monitoring

The CBR is pioneering research in the new area of environmental signaling. Studies are being pursued that focus on:

- how contaminants move through the environment,
- how they interact with receptors that regulate cellular functions, and
- how these receptors can be used as the basis of new technologies for hazard monitoring and remediation.

The use of receptor based methods to perform hazard monitoring has novel advantages compared to traditional methods that are used in ecological and human health risk assessment. Receptor based studies provide not only direct information about changes in molecular biomarkers used to measure exposure to contaminants, but also information about endocrine disrupting effects of specific contaminants.

Environmental exposure to endocrine disruptors is currently an area of intense worldwide interest. This area of research has been declared a national priority by the National Research Council and the National Academy of Sciences. A White House task force conducted an inventory of all federally funded endocrine-disruptor related research providing a research baseline and identifying data gaps. The White House also established an Interagency Task Force to respond to the national need for understanding both the short and long term influences that endocrine disrupters can have on the ecosystem and human health. Congress has passed a bipartisan amendment to the Clean Air and Water Act to provide funding for studying endocrine disruption. The DOE, as part of the interagency task force, has recently inventoried its funded research projects that deal with endocrine disruption. A basic science grant awarded to the CBR and work being carried out at Oak Ridge National Laboratory by CBR collaborators are the only projects in this area that are currently in the DOE's research portfolio.

Since the CBR has real expertise in this area, the CBR proposes to use this transitional funding to apply the CBR's research into the Bayou Trepagnier natural laboratory. To accomplish this, the CBR will recruit expertise in basic molecular based environmental research, as well as in engineering, at Tulane and Xavier Universities and at Oak Ridge National Laboratory. Unique collaborations will further define:

- how contaminants may or may not act as endocrine disruptors in aquatic ecosystems,
- how receptor based "molecular machines" can be designed to perform automated hazard monitoring, and
- how these constructs might be developed for use in removing contaminants through their receptor binding properties.

Bayou Trepagnier is an especially interesting sampling site on which to focus for this program because it not only contains hydrocarbons known to cause endocrine disruption, but it also receives large chlorine discharges from a sewage treatment plant. The chlorine from the sewage plant combines in the environment with the hydrocarbons to produce chlorinated molecules that are known to bind endocrine receptors. Transitional research

components will focus on the chlorinated hydrocarbons in the canal that communicates with the sewage treatment outflow and the bayou.

Correlation of Bayou Trepagnier (BT) Contaminant Data with DOE Sister-sites at Savannah River and Oak Ridge

Oak Ridge National Laboratory (ORNL) has been working closely with the project investigators since the onset of the program to provide research support and expertise in a variety of areas, including the design of Quality Assurance/Quality Control (QA/QC) protocols for sampling and analytical components of the project. Interactions have also included visits to Oak Ridge by Tulane/Xavier researchers, administrators and student interns.

Through leveraging this established partnership, the CBR will expand the scope of this program beyond the confines of the Mississippi River Basin. With Bayou Trepagnier selected as the focal sampling site, CBR proposes to correlate the data obtained from the bayou with data from bodies of water near DOE sites that exhibit similar characteristics and contain similar contaminants. Two DOE sites have been identified as "Sister-sites" to the bayou: Savannah River and Oak Ridge. Savannah River, like Louisiana, has considerable surface water (wetlands) in a similar type of climate. Although petrochemical contamination is limited in Savannah River, there is considerable contamination from chlorinated solvents, heavy metals, etc. On the other hand, Oak Ridge has mostly flowing surface water and a "pseudo tidal" effect from water flowing into fluctuating reservoirs. Contaminants at Oak Ridge include chlorinated solvents, PCBs, heavy metals, and radionuclides. Both DOE sites contain a variety of hydrocarbon and heavy metal contaminants that are also found in the water and sediments of Bayou Trepagnier.

CBR will approach the problems associated with the DOE sites from the viewpoint of environmental signaling, i.e., in understanding how contaminants influence receptors in cells, whether from animals, plants, or humans, as they are exposed to contaminants in the environment. While contaminants bind receptors that ultimately influence ecosystem and human health, they can also influence the functioning of the nervous system, immune system and genetic activity in other species. Thus, environmental signaling can predict risk to the health of the ecosystem as well as to human health.

These new data, when compared with aquatic animal health outcome data may result in more efficient and less costly methods to assess ecological risks. Connections to human health risk assessment will also be made since aquatic species, particularly fish, are now accepted as sentinel species for exposure assessment and biomedical models for human diseases such as cancer. The CBR is unique in its approach such that it can determine how waterborne and sediment-borne agents contribute to risk. Studies will integrate

receptor molecular biology and biochemical approaches in conjunction with other novel biomarker approaches developed during the initial DOE grant. Interpretation and communication of research results will target ecological and public health concerns as well as application to specific DOE risk management issues.

SCOPE OF NEW WORK

The scope of work for this transitional program has been divided into two parts: four major task areas and six cross-cutting factors for integrating the work performed in all task areas.

Task Areas for Scope of New Work

Task 1: *Contaminant: Receptor Binding as the Basis for a New Approach To Environmental Hazard Monitoring*

- To determine estrogen receptor localization and competitive receptor binding studies in reproductive (and other) tissues from wild fish species with a high prevalence of reproductive abnormalities.
- To determine the ability of a variety of hydrocarbon contaminants identified in Bayou Trepagnier to function as estrogens or antiestrogens at environmentally relevant concentrations using biotechnology screening tests.
- To utilize the progesterone, glucocorticoid, thyroid and retinoid receptors in the screening procedure to determine whether contaminants have multiple hormonal activities.
- To develop a special transgenic fish to monitor the estrogenic activity of the water in Bayou Trepagnier. Fish will glow with a green fluorescence in the presence of contaminants that act as environmental estrogens.
- To assemble and test a simpler laboratory screen for hormonal activity, that will complement the *in vivo* yeast estrogen screen.
- To construct a monitoring device for the detection and analysis of chemicals with hormonal activity in the water (or extracted sediment) from the Bayou.

Task 2: *Further Contaminant Identification, Characterization, and Monitoring in Bayou Trepagnier*

- To further identify, characterize, and monitor heavy metals and hydrocarbon contaminants in spoil bank soils and bayou bottom sediments.

- To test hypotheses concerning fate and transport mechanisms related to contaminant movements from the bayou into the surrounding marsh and into Lake Pontchartrain.
- To determine the role of dissolved organic carbon in providing a mechanism for natural removal of contaminants from the wetland system.
- To incorporate fate and transport data into a computer model that links the hydrology of the bayou with fate and transport of contaminants out of the bayou.

Task 3: *Ecotoxicity and Related Biomarkers of Exposure in Bayou Trepagnier: A Model System for Assessing Risk in Contaminated Wetlands Environments.*

- To further characterize histopathological and physiological biomarkers of exposure and aquatic animal health.
- To further characterize molecular biomarkers of exposure as indicators of toxicity in vertebrate and invertebrate animals.
- To incorporate new molecular biomarker data, including endocrine disruption data, into risk analysis models.
- To chart specific levels of molecular biomarkers for the development of less invasive tests, such as blood tests, to determine exposure in the field environment.
- To further characterize uptake of metals and hydrocarbons by plants living in the bayous and on the spoil banks and to understand its impact on the wetlands ecosystem.
- To perform laboratory studies and field studies to determine the ability of endemic bayou plants to perform remediation of wetlands contaminants.

Task 4: *Technology Development*

- To complete studies on thin film composite polyphosphazene membranes with carboxylate ion exchange groups for use in separating tritiated water from light water. These membranes could have an impact on the clean up of tritiated water and spent fuel storage basins on DOE sites near the Columbia River.
- To further characterize fungi and bacteria that degrade hydrocarbons.
- To construct a computer model describing how tidal flow from Lake Pontchartrain, rain, and flooding influence fate and transport of contaminants from the spoil banks in Bayou Trepagnier. This model will be important for understanding spoil bank leaching into aquatic ecosystems and for predicting rates of transport of contaminants away from the site.
- To further develop software that describes movements of microorganisms through different media related to bioremediation. These models should predict patterns of bacterial transport related to *in situ* bioremediation.

- To produce "molecular machines" that use bio-molecules for cell-free hazard monitoring and risk prediction.

Cross-cutting Factors for Project Integration

Information Retrieval and Analysis

All data, whether physical or biotic, field based or lab based, current or historical, are being entered into a project-wide data base. The centerpiece of the database is the Geographical Information Systems (GIS) capability for analyzing and specially plotting data. Thus, hypotheses generated concerning one class of data can be cross-tested using different types of data.

Technology Transfer

Physical and chemical remediation, bioremediation, fate and transfer studies, and biomarker studies are all integrated into an approach that is producing technologies for better risk prediction related to wetlands environments. The inclusion of endocrine disruption data in hazard monitoring and the construction of new detectors that rely on hormone receptors to assay levels of disrupting chemicals in the environment is unprecedented. These studies will provide, for the first time, a unique molecular approach to understanding the full scope of impact of contaminants produced by fossil energy production and refining in the wetlands environment. In addition, hardware and software related to removal of contaminants, for predicting the efficiency of bioremediation, and for producing cheaper and quicker field tests for heavy metal contamination will be further developed in work supported by transitional funds.

Publications, Reporting, and Program Management

An administrative team whose sole purpose is to coordinate, manage, and integrate all activities that are carried out under the project has been in place since the inception of the grant. In addition to tracking and collecting publications, producing reports, overseeing core work sponsored by the grant, and interfacing with the DOE/EM Program Manager, the management team communicates with local industries (e.g., Shell Oil) involved with the sites and also with local stake holders interested in wetlands environmental quality.

Collaboration with EM Focus Areas & Cross-Cutting Programs

CBR will actively interact with DOE/EM's Focus Areas and Cross-cutting Programs in the Field and at Headquarters to ensure the relevancy of the project's research to EM activities, especially with respect to usable technologies emerging from this project. Collaborations, currently under way, that relate to testing instrumentation developed at the Oak Ridge National Laboratory for field monitoring of hydrocarbon contaminant

levels in wetlands environments, will be expanded into the testing and design of instrumentation related to assaying unique endocrine disrupting contaminants with PNNL's Environmental Health Initiative.

Applicability of Research to Industrial and Other Government Environmental Concerns

The work in Bayou Trepagnier is currently being conducted in cooperation with Shell Oil Company. Shell is responsible for producing a remediation plan for Bayou Trepagnier. Information gathered, both by Shell and by the DOE funded project, have been shared freely. In fact, scientists from the project have been asked to advise Shell concerning the suitability of the proposed remedial action.

Research conducted under the project is applicable not only to industry and the DOE complex, but also to clean up activities at DOD and other Super fund sites. CBR will further explore these avenues to determine the potential for technology transfers.

International Activities

The Collaborative Research with IREP and CREM project is the result of a collaboration between Tulane University personnel and two Institutes in the Republic of Belarus. The project team is studying the transport and fate of radionuclides in the Iput River basin that includes modeling the movement of contaminants. This research is applicable to radionuclide contamination at DOE sites such as Hanford, Oak Ridge, and Savannah River.

FACILITIES AND RESOURCES

Center for Bioenvironmental Research

The CBR at Tulane and Xavier Universities represents a solidly established and well-developed collaboration between a major research university and one of the nation's leading historically black universities. This unique partnership combines leading-edge research with innovative math and science education programs to produce an environment that opens up science and engineering careers to minority students, and encourages the multi-disciplinary cooperation needed for a productive environmental research program.

In a joint partnership, Tulane and Xavier Universities created the CBR, an interdisciplinary research center dedicated to investigating the causes and effects of environmental problems, and developing cost-effective solutions. The CBR fosters

extensive collaborative research with such diverse disciplines as toxicology, medicine, epidemiology, cell and molecular biology, chemistry, ecology, engineering, and information sciences. With this synergistic approach, scientists in these areas can draw on each other's expertise, fostering an ideal environment for unraveling complex biomedical problems.

The CBR was founded in 1989 when the US Department of Defense awarded a grant to the two universities to develop advanced technological methods to assess, control, prevent, and clean-up environmental hazards. Additional funding for the CBR is provided by participating universities, federal and private grants and contributions, and industry. With these funds, the CBR is able to acquire and maintain equipment and facilities, as well as to recruit established scientists in the field.

Faculty associated with the CBR study both health effects and remediation techniques, with the ultimate goal of developing responsible public policy. Research focuses on understanding the basic mechanisms by which toxic substances compromise health. This allows scientists to design prevention, intervention, and therapeutic strategies to mitigate the effects of human and animal exposure to environmental contaminants. Such research will also lead to new technologies for the removal and degradation of toxic materials from contaminated areas. Most importantly, environmental research is the cornerstone of effective strategies for disease prevention.

Within the CBR mission, interdisciplinary and inter-institutional collaboration is pivotal. With the best minds and talents of a broad spectrum of scientists, physicians, engineers, and public policy researchers working to provide reliable, accurate scientific information, the CBR is a conduit for effective public policy and sound decision-making for businesses.

CBR Resources and Facilities

The CBR operates within first rate core research facilities that house sophisticated research equipment including: exposure chambers for humans and animals for respiratory disease studies and extensive computer equipment for Data Management and Geographic Information Systems (GIS) Analysis. These capabilities are further strengthened by analytical equipment and cell and molecular biology equipment, which provide resources for faculty in collaborative arrangements. The CBR has a physically and scientifically close working relationship with all the departments at both Tulane and Xavier Universities.

Core Informatics Group and Equipment Base

The CBR has established an environmental informatics team, capitalizing on existing resources to "push the envelope" on information sciences at Tulane and Xavier Universities. Furthermore, the CBR has established an Internet site on the World Wide

Web (http://www.cbr.tulane.edu); most CBR faculty are connected by e-mail; planning is done over the Internet; and the capacity for large-scale computer modeling of data sets has been developed.

State of the Art Shared Equipment Facility

CBR supports the **Coordinated Instrumentation Facility (CIF),** an important equipment facility which provides tools for analysis and synthesis of chemicals and localization of molecules through imaging technologies. These tools are made available to a wide array of scientists from many different campuses.

The CIF represents a strength both in terms of providing an equipment resource that is on-site and of assuring continuous management of that equipment by its first rate over-sight team. Importantly, it is an investment that has already been made.

APPENDIX A

EM-AQUATIC ACCOMPLISHMENTS TO DATE

During five years of funding from the DOE, the grant has supported twelve collaborative cluster projects (multi-year awards) and twenty-six initiation projects (one year awards). Over 75 faculty from Xavier University (School of Arts and Sciences and College of Pharmacy) and Tulane University (Liberal Arts and Sciences, School of Engineering, Medical School, and the School of Public Health and Tropical Medicine) have participated in the project to date. More than 50 graduate and numerous undergraduate students have received scholarships and/or worked on research problems under this project.

Through collaborations with Oak Ridge National Laboratory (ORNL), the CBR it's project investigators have been working diligently to ensure the program continued to meet the DOE's evolving needs. The DOE provided research support and expertise in a variety of areas, including the design of Quality Assurance/Quality Control (QA/QC) protocols for sampling and analytical components of the project. Interactions have also included visits to Oak Ridge by Tulane/Xavier researchers, administrators and student interns.

Establishment of Cores

Following a recommendation by the December 1995 External Review Panel, CBR set up research support cores to support the competitively funded projects under the grant: data management, field work, chemical analysis, and education. The cores which will be maintained for transitional funds include data management, field work, and chemical analysis. Participants in core groups perform work related to research and technology development projects and are expected to produce interdisciplinary publications related to the project.

Data Management Core

Is responsible for the development of an infrastructure and sample tracking system for data collected through EM-Aquatic funding. The data is collected, reviewed, and stored in a central electronic repository (database) accessible to all investigators. The database is interfaced to the Geographic Information System (GIS) and tightly links the precision of analytical instrumentation to the spatial capabilities of GIS. The Data Management Core also develops tools for electronic access to the database, digitizes geographic features in the bayou using the Global Positioning System (GPS), analyzes data, and generates maps using (GIS).

Field Work Core

Is responsible for overseeing and coordinating all field work through EM-Aquatic funding, as well as standardizing collection procedures and QA/QC. The field work core includes co-directors from both Tulane and Xavier Universities. The Core provides researchers with water, soil, or biotic samples, measures water quality parameters, and coordinates the overall plume characterization effort.

Chemical Analysis Core

Provides centralized coordination to support the chemical analyses of all work funded through DOE. Analysis of environmental samples for metals and/or organic chemicals is coordinated with analytical work performed at the Coordinated Instrumentation Facility (CIF). The Core performs organic and inorganic analyses, insures uniformity of analysis protocols, and maintains QA/QC and instrument certification programs for each of the satellite labs involved in the project.

Bayou Trepagnier Characterization

Bayou Trepagnier was chosen by the CBR's EM-Aquatic Project as a natural laboratory for studying the fate and transport of heavy metal and organic contaminants in wetlands. Specific tasks include characterization of contaminant loadings, water quality, hydrology, and biota.. As described above, cores were established to streamline and coordinate sampling, contaminant characterization, and data analysis. Over the past five years, the Core components of the project have been gathering data in the bayou and the swamp to characterize the plume in order to assess the impact of contaminants on the wetland ecosystem.

Bayou Trepagnier has been heavily impacted by the discharge of heavy metals (lead, zinc, chromium, and copper), crude oil, and refining intermediates from the Norco Manufacturing Complex over the past 80 years. Contaminants were mainly confined to the bayou channel until extensive dredging in 1951 created spoil banks mainly on the spillway side of the bayou. Over the past 45 years, the weathering of spoil bank material mobilized lead, zinc, and chromium, thus contaminating the marsh and swamp adjacent to the bayou. Monitoring of the water column indicates that lead found in sediments tends to be tightly bound as sulfides. The possibility of sporadic releases from the sediments exists due to the high content of dissolved organic carbon in the water column. Polycyclicaromatic hydrocarbons, the dominant organic contaminants in the bayou, are concentrated in the bottom sediments, whereas the spoil banks are relatively free of these compounds. Several of these compounds that occur at high concentrations are known to have carcinogenic, embryotoxic, and endocrine disrupter effects on aquatic organisms.

Work performed under the past EM-Aquatic grant characterized the distribution of heavy metal and hydrocarbon contaminants, both in Bayou Trepagnier, and in the spoil banks

that bound the bayou. The Bayou has been mapped through careful surveying, from the site of the Norco Refinery at the headwaters of the bayou to the bayou's outflow into Lake Pontchartrain. Data from contaminated sample sites have been incorporated into a large database and used in GIS analyses to track the fate and transport of heavy metals from spoil banks into the surrounding marsh. These data are crucial to understanding how heavy metals move through aquatic and wetland environments.

Initial characterization of fish populations from Bayou Trepagnier indicates that the species diversity is low. Some field validation of biomarkers of mixed chemical exposure in spotted gar developed in Devil's Swamp were verified in Bayou Trepagnier gars. Initial biomarkers results indicate that Bayou Trepagnier is an ideal site for studying endocrine disruption and reproductive/developmental impacts of mixed chemical pollution typical of DOE sites.

Technology Development

An important aspect of EM-Aquatic research is technology development and the process of transferring the technology into a usable entity. Several areas of research in the project have resulted in the development of technologies that have potential use in the DOE environment for remediation and waste management. Through transitional funding, these areas will be further explored for transfer and use by the DOE complex or by commercial concerns.

Sensitive Rapid On-Site Immunoassay for Heavy Metals

Monoclonal antibodies to specific metal-chelate complexes were developed for quick, cost-effective assessments of metal contamination at field sites. The assay used an antigen inhibition format to measure the soluble metal ions. The sensitivity of the metal ion immunoassay can be modulated by changing the structure of the inhibiting antigen. Methods are being developed to enable the extraction of heavy metals from soil and sediment samples in the field without recourse to strong acids, high temperatures, or cumbersome equipment. A prototype immunoassay for cadmium as Cd(II) has been tested in the field. Further research on the immunoassay would contribute to developments in the Metals and Radionuclide Remediation Product Line of DOE/EM's Subsurface Contaminant Focus Area as well as the Characterization, Monitoring and Sensor Technology Cross-cutting Program.

Polymeric Membranes for Separations of Contaminants from Water

Preliminary studies have shown that tritiated water can be separated from light water using a thin film composite polyphosphazene membrane with carboxylate ion-exchange groups. The separation was carried out in a low pressure reverse osmosis process. The development of these membranes could have an impact on the clean-up

of tritiated water in spent fuel storage basins on DOE sites near the Columbia River. Development of this technology would contribute to advances in the Short-Lived Radionuclide Product Line in DOE/EM's Efficient Separations and Processes Cross-cutting Program. The Hanford Site's Tri-Party Agreement currently requires that tritiated water be treated or removed.

Bioremediation Techniques that use Fungi and Bacteria to Break Down PAHs

A patent application for encapsulating fungal strains that degrade hydrocarbons has been submitted. Further work will characterize metabolic pathways and enzymes in the degradation process. This research would contribute to remediation processes in the Organics and DNAPLs Product Lines in DOE/EM's Subsurface Contaminant Focus Area.

Pore-level Microorganism Behavior Modeling for Optimizing Bioremediation

Laboratory experiments, computer models, and specific software are being developed to describe the movements of microorganisms through different media for bioremediation processes. The models include a detailed analysis of convection and diffusion within the pores, as well as chemotactic responses of swimming microorganisms to local contaminant concentration gradients. The models also include microbial adhesion to each other and to the surrounding pore structures. These models should predict patterns of bacterial transport related to *in situ* remediation. These studies relate to the development of technologies for the Subsurface Contaminant Focus Area.

Molecular Machines for Hazard Monitoring and Risk Assessment

The ultimate endpoint of contaminant**:**receptor binding studies is to produce new "molecular machines" composed of specific biological receptors linked to a reporter system that will sense cytokine or hormone disrupting contaminants in the environment. The magnitude of the response should indicate both bioavailability and risk. These constructs would replace both the contaminant measurement phase of risk assessment, and the biomarker validation phase which, in current work, is carried out by exposing animals to field sediments. Preliminary work has been initiated to explore the design of molecular machines that will incorporate the use of functional bio-molecules, usually receptor molecules, coupled to a bio sensor to detect hazardous chemical: receptor interaction. The activated biosensor responds by giving off or absorbing photons of light. These biologically engineered constructs are fixed to support systems which may be solid or flexible but can be placed in the field and monitored by light storage devices or connected to electronic monitoring systems. Tulane and Xavier have the ability and capacity to handle all stages, including

formulation of the devices. The Department of Biomedical Engineering is applying for a major grant for a Laboratory for Artificial Cell Engineering which will create truly cutting edge technologies for the combination of biological systems with artificial materials. Application of new technologies such as these might represent a considerable savings for the Department of Energy in hazard assessment work related to risk based prioritization of sites to clean up. Development of these technologies will produce a new generation of simple devices that not only will detect exposure, but will also be calibrated to predict risk and associated clean up levels. These constructs may also be adapted for use in binding contaminants to accomplish clean up.

International Collaborative Research with IREP and CREM

Tulane University personnel and two Institutes in the Republic of Belarus (the Institute of Radioecological Problems (IREP) and the Committee on Radiation and Environmental Monitoring (CREM)) are studying the transport and fate of radionuclides in the Iput River basin. The study includes modeling the movement of contaminants. A non-linear curve fitting scheme that can automatically interpolate and produce a mapping of the contaminants' distribution is being developed. In addition to utilization of traditional methods that make use of systems of differential equations, the project is developing the technique of neural networks as a way of mechanistically looking at the problem of radionuclide distribution.

Related Research Important to DOE

The research conducted through the CBR's EM-Aquatic Program has resulted in significant enhancement in fundamental knowledge specifically targeted to "real-life" environmental problems. Through support from DOE/EM, the CBR established collaborative research core groups to focus on field work, data management and chemical analysis. The work impacts human and ecosystem health on both regional and national levels. In addition, this research resulted in CBR's award of a DOE-EM Basic Science Competitive Grant for $620,000 in September 1996.

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