Appendix D

Other Research

EPA has a number of grant programs that support research in areas of special significance to the Agency's mission. EPA's National Center for Environmental Research in the Office of Research and Development runs competitions for Science to Achieve Results, or STAR grants, in numerous environmental science and engineering disciplines through a competitive solicitation process and independent peer review. The program engages scientists and engineers in targeted research that complements EPA's own intramural research program.

The Regional Applied Research Effort (RARE) provides the Regions with a mechanism to address near term research needs through an ORD Laboratory/Center. Applied research projects are funded to meet information needs that a Region identifies as necessary and that an ORD laboratory has the expertise to carry out.

The EPA is also one of 11 federal agencies that participate in the Small Business Innovation Research Program established by the Small Business Innovation Development Act of 1982. The purpose of this Act was to strengthen the role of small businesses in federally funded R&D and help develop a stronger national base for technical innovation. Through the SBIR Program, EPA makes awards to small, high-tech firms to help develop and commercialize cutting-edge environmental technologies.

Science to Achieve Results (STAR) Grants

Data Collection and Modeling of Enteric Pathogens, Fecal Indicators and Real-Time Environmental Data at Madison, WI (EPA Grant Number: R829339)

The City of Madison, Wisconsin contains three recreational lakes with over 20 miles of shoreline within the city limits. The lakes are heavily used for recreational activities including sail boating, power boating, wind surfing, water skiing, swimming, scuba diving, canoeing, kayaking, fishing and jet skiing. The Madison Department of Public Health (MDPH) developed beach-closing criteria using testing results, combined with physical observations at the beach sites. There was a concern that the criteria might not reflect the actual risk to swimmers because the occurrence of pathogenic microorganisms during periods of high indicator levels had never been determined.

The objectives of this research are to:

1. Expand the current city beach monitoring program to include use of improved indicators that index changes in the microbial quality of the beach water including sensitive gene probe technologies to discriminate between human and animal sources of fecal pollution.

2. Determine the correlations between microbial indicator data, occurrence of pathogens, and meteorological, physical and water quality data collected by remote monitoring stations.

3. Consider mathematical constructs for modeling pathogen occurrence.

4. Create innovative partnerships with community groups and agencies to facilitate dissemination of water quality data and beach closure decisions, including development of a water quality web-based database with dynamic query capacity for the public.

For more information visit: http://cfpub.epa.gov/ncer_abstracts/index.cfmfuseaction/display.abstractDetail/abstract/5843/report/0

Real-Time Water Quality Monitoring and Modeling for Equitable Recreation on the Mystic River (EPA Grant Number: R829338)

The city of Somerville, Massachusetts, in collaboration with Tufts University and the Mystic River Watershed Association, proposed a project that combines advanced technology for real-time water quality and meteorological monitoring with sampling of bacterial levels to develop a model that anticipates river conditions, especially after heavy rains. The real-time data, water quality indices, and model-generated water quality predictions will be made available to the public via the Internet as well as color-coded flags at riverfront sites. The predictive model will enhance standard water quality monitoring by providing a way to anticipate bacterial levels that ordinarily require 24 hours to assess, leaving citizens in this dense and heavily polluted river basin with inadequate information about water safety.

The project objectives include collection of water quality indicator data (e.g., fecal coliform, enterococcus, DO, turbidity) along with data on depth, temperature, pH, conductivity, and meteorological conditions in real-time to develop an "early warning" water quality forecasting model. The data presentations will interpret the data into indices useful for everyday decisions about contact with the water.

For more information visit: http://cfpub.epa.gov/ncer_abstracts/index.cfm/fuseaction/display.abstractDetail/abstract/5858/report/0

Prevalence and Survival of Microorganisms in Shoreline Interstitial Waters: A Search for Indicators of Health Risks (EPA Grant Number: R828830)

Researchers felt there was some suggestion in the literature that the microbiological quality of beach sand may constitute a health risk to bathers, particularly children who spend time in the "swash zone." Sand could act as a filter to trap and concentrate bacteria, spores, and cysts because it has a large surface area for microbial attachment, ample oxygen levels, higher temperatures, and a constant resupply of nutrients through wave action and tides. Pathogenic organisms could potentially accumulate in interstitial space. Organisms could then be periodically swept from surfaces and transported to the surf zone where they pose a health risk aggravated by the abrasive nature of sand, the ingestion of contaminated waters, and the inhalation of aerosols rich in microbes. Thus, individuals exposed to the surf zone of populated beaches may show a higher incidence of illnesses from either enteric or nonenteric pathogens.

The study will document the number of "classic" fecal indicators in sand (*E. coli*, enterococci, and fecal coliforms), paying attention to whether they are free in interstitial space or attached to sand particles. Consideration will be given to the possibility that some of these organisms are lofted into the air. Other non-indigenous microorganisms in sand including non-enterics, coliphage and several eukaryotic microbes will be enumerated. A laboratory-based microcosm will be enumerated to study the survival (and growth) of indicator organisms in sand relative to overlying open water. Additionally, correlations of microbial abundance data to incidences of beach-related indicators will be compiled.

For more information visit:http://cfpub.epa.gov/ncer_abstracts/index.cfm/fuseaction/display.abstractDetail/abstract/1009/report/0

Near-Real Time Monitoring of Inland Suburban Waterways: Application to Three Critical Environmental Issues Facing the North Shore/Metro Boston (EPA Grant Number: R828582)

Local citizen groups are engaged in environmental monitoring of two major tributaries that empty into Plum Island Sound estuary, the Ipswich and Parker Rivers. In addition, commonwealth, federal agency and research/academic groups have carried out focused studies on these watersheds.

The proposed work assembles a consortium of eleven partner groups drawn from the public, academic, and private sectors. Through this partnership the group wants to link several, ongoing, but currently uncoordinated, environmental monitoring efforts. The existing environmental data sets will serve as a historical benchmark by which to assess future change detected by the

near real-time monitoring system that will be installed. Pooled data will be evaluated with models and Web tools to observe the changing character of the Ipswich and Parker watersheds. Although the focus is on monitoring, the consortium also has technical expertise to interpret and draw scientifically sound conclusions from the emerging data sets. An active public outreach program is included in the proposal.

The focal point for this work will be a geospatial Web-based information system, the Ipswich/Parker Suburban Watershed Channel (I/PS-WATCH), akin to the "Weather Channel" but reporting on suburban watershed environmental variables. The interface will represent extension of an existing system (see: http://www.gm-wics.sr.unh.edu/ for prototype). I/PS-WATCH will be applied to three sub-projects, already identified by the partners as of high public relevance, cast in near real-time.

For more information visit: http://cfpub.epa.gov/ncer_abstracts/index.cfm/fuseaction/display.abstractDetail/abstract/408/report/0

Molecular Detection of Anaerobic Bacteria as Indicator Species for Fecal Pollution in Water (EPA Grant Number: R827639)

Fecal contamination of aquatic environments is a continuing problem. Yet some of the standard indicators for fecal pollution do not distinguish between human and animal sources. A novel indicator system was developed based on the anaerobic gut bacterial group Bacteroides/Prevotella. Molecular markers, amplified from bacteria filtered from the water, are measured. This method can already distinguish human from cow fecal pollution in both estuarine and river waters. The proposed study will focus on a small, nutrient-rich, fecally polluted estuary, Tillamook Bay, Oregon, and its tributary rivers.

The objectives of this proposal are:

1. To develop additional markers from other biologically important polluting species, such as waterfowl.
2. Identify the indicator strains or species that are host-specific.
3. To allow quantitative estimation of both the amount of total pollution in the water and the proportions of different sources of fecal pollution.

For more information visit: http://cfpub.epa.gov/ncer_abstracts/index.cfm/fuseaction/display.abstractDetail/abstract/279/report/0

Community Recreational Water Risk Assessment and Public Outreach (EPA Grant Number: R827063)

A consortium was formed with several scientific and community organizations for the purpose of more effectively collecting and disseminating to the public recreational water quality data from several beaches in Milwaukee and Racine. This project is focusing on reporting E. *coli* levels to the public in a time-relevant and meaningful format.

Project objectives include:

- To improve documentation and dissemination of environmental data specifically related to health risk associated with the recreational use of public beaches.
- To improve the type, quantity and quality of environmental data collected at and around public beaches in both Milwaukee and Racine Counties in development of a public health risk model.
- To improve coordination and collaboration of environmental data collected between Local Public Health Agencies (LPHAs), other organizations and community stakeholders, and standardize data collection.

For more information visit: http://cfpub.epa.gov/ncer_abstracts/index.cfm/fuseaction/display.abstractDetail/abstract/427/report/0

Regional Applied Research Effort (RARE) Grants

Development of Guidance on Decision-Making When Using Microbial Source Tracking Methods

Molecular biology methods (e.g., DNA fingerprinting) are now commonplace in public health monitoring

programs. One of the more common uses is microbial source tracking (MST), i.e., identifying the sources of pathogens in water. The potential value of these methods is illustrated in a recent report published by the Natural Resources Defense Council. The report noted that 87 percent of the 13,000 beach closings and water advisories in 2001 were due to high levels of bacteria associated with fecal contamination, but in 54 percent of the cases, the exact source of the contamination could not be identified.

The methods that have been used include genotypic (e.g., ribotyping, rep-PCR, and pulsed-field gel electrophoresis) and phenotypic (antibiotic resistance testing and carbon utilization) approaches. These methods have already been used in recreational water and shellfish bed closure programs, TMDLs, and source water protection for both surface water and ground water in several coastal states in the United States. Although already in use by state and local governments, these methods have not been fully validated, and no single best method for all situations has been discovered. Integration of the vast array of information on these methods and publishing of this guidance as an EPA document will support the Regions in the appropriate use of these methods and inform potential users of issues that need to be considered when selecting a method.

For more information visit: http://intranet.ord.epa.gov:9876/OSP/RARE.nsf/7523fef8d5be8b05852569fa00619181/0540a0d5a97be2fa85256f6600589621?OpenDocument

Proof of Concept Demonstration for Near Real-Time In Situ Detection of Fecal Contamination in Fresh and Marine Waters

A key component of monitoring is the detection and timely reporting of concentrations of bacteria that are associated with human gastroenteritis and indicate fecal contamination. Current monitoring methods require incubation periods between 24 hours and 3 days before public health decisions can be made. The development of new in situ instruments with the capability for rapid, near real-time, quantification of bacterial densities would provide a more effective and better warning system for both environmental managers and the public.

For more information visit: http://intranet.ord.epa.gov:9876/OSP/RARE.nsf/7523fef8d5be8b05852569fa00619181/a0c21dbd217c5e6485256c83006cdae3?OpenDocument

Small Business Innovation Research (SBIR) Program

Portable Pathogenic Predictor for Storm water (EPA Contract Number: 68D99028)

The key objective of this phase of the project is to develop a novel dual-wavelength radiometric fluorescence method and sensor for the detection of coliform bacteria in water samples. The radiometric technology will be rapid, robust, and suitable for the development of online sensors for monitoring the effluents of urban storm water and sewage treatment plants. This novel approach, unlike previous single-wavelength intensity-based methods, will be highly immune to light source and detector instabilities, temperature effects, optical density, and turbidity variability in the sample, spurious quenching, and photobleaching. In contrast to current methods that take 24 hours or more to generate results, the proposed technology will provide reliable results in minutes to hours, depending on bacterial concentration.

The overall potential market for coliform testing of water encompasses several segments: drinking water, municipal sewage, coastal fisheries, shellfish aquaculture, and beach and recreational waters. Products will include a portable instrument, reagent kits, and online sensors.

For more information visit: http://cfpu b.epa.gov/ncer_abstracts/index.cfm/fuseaction/display.abstractDetail/abstract/1267/report/0

A New Biosensor for Rapid Identification of Bacterial Pathogens (EPA Contract Number: 68D02051)

Rapid, handheld, or portable instrumentation for determining the quality of natural waters, recreational waters, and distributed and treated supplies does not currently exist. Echo Technologies, Inc., completed a Phase I project that demonstrated a new approach for identifying bacteria in aqueous systems. The approach uses bacteriophage as the molecular recognition

element. Bacteriophage are virus particles that generally attach to, and infect, a narrow range of host cells. Biosensors based on this molecular recognition offer a rapid, selective, and potentially very sensitive method to detect bacteria and bacterial pathogens in potable and recreational waters.

Several experiments were conducted with a customized detection system to demonstrate the feasibility of making a small in-line instrument capable of high-sensitivity detection.

Application of the fabricating fluorescently labeled virus probes (FLVP) technology to solid-state optical sensing represents a new approach to real-time detection of bacterial pathogens. This approach will minimize the need for culturing to identify pathogens and is an important departure from immunoassay- or DNA-based sensing concepts. The miniature probes are perfectly suited for incorporation in a sensor array for the simultaneous detection of many bacterial pathogens.

For more information visit: http://cfpub.epa.gov/ncer_abstracts/index.cfm/fuseaction/display.abstractDetail/abstract/5182/report/0

Automated Human Fecal Pollution Detection (EPA Contract Number: EPD05036)

Public health departments have set strict standards for the quantity of coliform bacteria allowed in the water. Because of the requirement to detect very low levels of these bacteria, rapid automated detection is very difficult. Culture techniques take 24 hours, and the more rapid DNA amplification techniques still require DNA purification and the use of unstable enzymes and nucleotides as well as elaborate instrumentation, all of which are difficult and expensive to automate. In this research project, an automated biosensor capable of detecting low levels of fecal microorganisms without the need for bacterial culture or DNA amplification techniques will be developed. The biosensor should be sensitive enough to detect fecal microorganisms at a level of 1 coliform cell per 100 mL of water within 30 minutes. The sensing element will be reusable to allow for long-term, unattended, cost-effective analysis. The sensor also should distinguish human from farm livestock sources of fecal pollution.

The final product is envisioned as an automated buoy placed in drinking water inlet sources, swimming waters, and shellfish or other aquaculture growing waters. The buoy will report fecal microorganism levels at set time intervals by remote telemetry communication. A portable version of the instrument also will be developed.

For more information visit: http://cfpub.epa.gov/ncer_abstracts/index.cfm/fuseaction/display.abstractDetail/abstract/7485/report/0