SERDP
Strategic Environmental Research and Development Program
Improving Mission Readiness Through Environmental Research

Genosensor-Based Ecotoxicity Response Assessment

Cleanup
CU-1081

Background:

No technologies currently exist to directly assess biological responses to exposure of ecosystems to chemicals released into the environment. The ability to directly measure the biological response associated with genotoxic exposure would alleviate the difficult problem of determining bioavailability since measurable molecular endpoints are a direct reflection of ecologically relevant exposure.

Objective:

This project seeks to develop cost-effective methods and instrumentation for directly monitoring genotoxic exposure in a variety of natural ecosystems. The project intends to implement emerging biochip technology for in- situ monitoring of molecular endpoints of genotoxic exposure, including DNA damage-inducible gene expression pathways, in soil and water ecosystems.

Summary of Process/Technology:

Novel channel glass biosensor chips containing arrays of DNA probes will be employed to characterize and monitor the response of soil microorganisms to exposure to genotoxic agents.

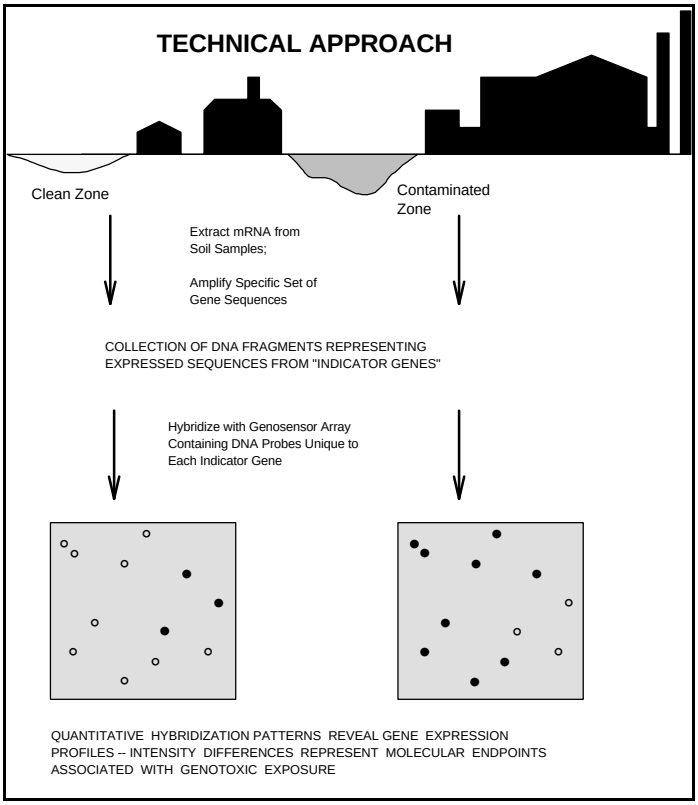
Benefit:

Capabilities for rapid, multispecies biological endpoint monitoring that is ecologically relevant to the clean up of contaminated sites should provide a rational basis for addressing the "How clean is clean?" question. This technology will greatly facilitate site characterization, risk assessment, and monitoring of the progress of remediation efforts at Department of Defense (DoD) and Department of Energy (DOE) installations.

Accomplishments:

The genosensor systems have been assembled at the Oak Ridge National Laboratory located in Tennessee. The system consists of the following components: microchannel glass wafers fabricated by the Naval Research Laboratory, a robotic fluid dispensing system for delivering oligonucleotide probe solutions to the genosensor chip surface, a fluidics module for delivery of nucleic acid analyte through the glass channels, a charged coupled devise (CCD) imaging system for acquisition of hybridization patterns, and software for interpretation of results.

Proof-of-concept initial hybridization experiments have been performed successfully on flat glass chips using a bacterial gene sequence known to be induced in response to aromatic toxicant exposure. Initially, genosensor chips were fabricated that contain probes targeted to about 30 genes known to be differentially expressed in bacteria upon exposure to toxic chemicals or other stress conditions. Methods for extraction of intact DNA and RNA from sediments and soils were evaluated. Inactivation of ribonucleases in soil samples has been demonstrated and various strategies for overcoming the sequestering of RNA by humic acids in soil and silt have been explored. Sample collection and preparation procedures are being evaluated with respect to the issue of site-to-site variability in microbial populations in environmental samples.



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