

# Research Brief 171: An Integrated Approach to Assess Sediment Toxicity

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## Background

Sediments are the recipients of most of the organic and inorganic contaminants released into the environment. Historically, sediments are often contaminated with mixtures of polycyclic aromatic hydrocarbons (PAHs), as well as polychlorinated biphenyls (PCBs), pesticides, heavy metals and other compounds. While there are extensive data to confirm that PAHs and other sediment contaminants are toxic to aquatic receptors, information about the bioavailability and genotoxicity of complex mixtures to aquatic organisms is limited.

The results of chemical analysis generally do not predict toxicity in a biological system, particularly for complex pollutant mixtures such as PAHs or PCBs. Dr. K.C. Donnelly, Texas A&M University SBRP, believes that a battery of biomarkers provides a valuable tool to adequately characterize subtle, sublethal, or genotoxic effects of chemical mixtures. Much like different instruments or detectors may be used to detect different classes of chemicals; a battery of biological tests can be used to detect a range of toxic or genotoxic effects. A collaboration between six different university-based programs (Baylor, Duke, Michigan State, UC-Davis, UC-San Diego and Texas A&M) resulted in the formation of a National Bioassay Network to develop a protocol for analysis of sediments. The goal of the National Bioassay Network is to design an integrated biomonitoring approach using chemical analyses and a suite of biomarkers to characterize the bioavailability and genotoxicity of complex chemical mixtures in sediments. In collaboration with Dr. Bruce Duncan (USEPA Region 10), the Bioassay Network conducted a monitoring study using caged Coho Salmon in a freshwater lake near a PAH contaminated site in Region 10.

## Advances

Four approaches have been used separately to assess exposure and genotoxicity of chemical mixtures in aquatic organisms:

- *In situ* studies to monitor body burden of various contaminants in caged fish.;
- Quantification of erythrocyte micronuclei, which are small DNA-containing bodies found near the cell nucleus as a consequence of chromosome breakage and spindle dysfunction.
- Detection of DNA changes using flow cytometry (FCM), which simultaneously assays numerous cellular and molecular endpoints.
- Quantification of formation of DNA adducts, a key process in early carcinogenesis, using  $^{32}\text{P}$ -postlabeling.



Dr. Donnelly's group used all four measures, along with chemical analyses, in a field study designed to mimic the exposure scenario experienced by migratory juvenile Coho Salmon. Caged salmon were suspended in the water column at sampling stations in a freshwater lake contaminated by a former manufactured gas plant (MGP), or in a reference (control) lake. USEPA SCUBA divers collected sediment samples at each station the day before placement of salmon cages. They also collected lake water samples at each sediment station at the beginning and end of the one week long fish exposure study.

The researchers analyzed the sediment and water samples for the seven PAHs identified by EPA as probable human carcinogens. They also measured liver PAH concentrations to assess PAH uptake and bioavailability from sediments and lake water, and to assess correlations between liver PAH concentrations and *in vivo* biomarker responses.

Briefly, they found that:

- The test locations all had levels of PAH contamination significantly above typical anthropogenic levels, in proportions expected at sites impacted by MGPs.
- Liver PAH concentrations showed no correlation to concentrations at cage locations, supporting earlier studies that liver PAH concentration is not a good biomarker of exposure or effect (possibly due to rapid metabolism of PAHs in fish liver).
- Erythrocyte micronucleus counts did not show a dose-response relationship in the Coho Salmon. There was no difference in micronuclei between the hatchery fish, reference lake fish, and fish at any location in the MGP site lake.
- FCM data revealed a clear trend in genotoxic response that corresponded with the PAH concentration gradient observed in sediments.
- <sup>32</sup>P-postlabeling assays showed a dose-response effect, suggesting there was an increase in salmon hepatic DNA adducts from exposure to MGP site lake PAHs. Liver adducts were found to be a more sensitive measurement of exposure than gill adducts (which is unusual for fish).  
The <sup>32</sup>P-postlabeling data revealed a DNA adduct response gradient that corresponds to the sediment, but not water column, PAH concentration gradient.

This exposure scenario study demonstrated that PAHs in sediments and lake water are bioavailable and can have a genotoxic impact on migratory juvenile Coho Salmon.

### Significance:

When incorporated into a well-designed *in situ* biomonitoring plan, the biomarkers used in this research appear to provide sensitive measurements of the bioavailability and genotoxicity of PAH-contaminated sediments to aquatic species. Such biomarkers of exposure and effect can be used to establish quantitative correlations of the cause and effect relationship of a toxicant at any level of biological organization, from an individual organism up to a population or ecosystem. *In situ* biomonitoring has a great potential for linking biomarker data with community and ecosystem level responses.

Biomarkers respond to toxicant exposures in a time-dependent manner and have varying levels of longevity and stability. Thus, using multiple biomarkers of exposure and effect should improve the quality of the risk assessment. It may be useful to combine the measurements of genotoxicity (e.g., DNA adduct formation), as a molecular dosimeter, with an analysis of carcinogen metabolism, exposure dose, and the determination of tumor formation in order to provide insight into the dose-response relationship and the mechanisms involved in chemical carcinogenesis.

Dr. Donnelly has taken the concept of applying an integrated approach beyond his own laboratory. He brought together a group of SBRP-funded researchers who have developed a group of class-specific bioassays and biomarkers with the potential to identify degraded sediment quality at lower sediment concentrations than standard aquatic toxicity bioassays – and that may also be capable of detecting biologically significant endpoints that are not measured in standard aquatic toxicity bioassays. In collaboration with EPA Regional and Headquarters risk assessors and technical staff, they are working to develop a model to cross-walk the new bioassay and biomarker results for use in ecological risk assessment.

To date, the SBRP Bioassay Network has conducted a “Proof of Concept” exercise to document the potential of the bioassays to assess potential adverse impacts on human or ecological receptors. Each laboratory analyzed sub-samples of standard solutions of two PAHs (benzo(a)pyrene and pyrene), two PCBs (one TCDD-like, one non-TCDD-like), and one reference mixture (NIS MGPR mixture). Results were presented at the 2008 EPA Risk Assessors’ Conference and a manuscript is nearly ready for submission to Environmental Science & Technology.

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## To learn more about this research, please refer to the following sources:

Barbee G.C., J. Barich, B. Duncan, J.W. Bickham, C.W. Matson, C.J. Hintze, R.L. Autenrieth, G-D Zhou, T.J. McDonald, L. Cizmas, D. Norton and K.C. Donnelly. 2008. *In situ* biomonitoring of PAH-contaminated sediments using juvenile coho salmon (*Oncorhynchus kisutch*). [Ecotoxicology and Environmental Safety](#) **71(2)**:454-64. [doi:10.1016/j.ecoenv.2008.01.001](https://doi.org/10.1016/j.ecoenv.2008.01.001)