1. MICROTOX® ACUTE TOXICITY TEST

B. THOMAS JOHNSON

Environmental Microbiology
Columbia Environmental Research Center
U. S. Geological Survey
4200 New Haven Road
Columbia, Missouri 65201 USA
btjohnson@usgs.gov

1. Objective, development, and scope

The Microtox Acute Toxicity Test, usually identified as Microtox, has played a leading and pivotal role in developing minimalistic microscale toxicity testing. “Speed, simplicity, reproducibility, precision, sensitivity, standardization, cost effectiveness, and convenience” (Isenberg, 1993) were features sought and developed in Microtox. This test uses a specific clonal strain of bioluminescent bacteria prepared in a unique lyophilized vial format. This approach is rapid, simple, cost-effective, and sensitive with large sample throughput capabilities. Microtox is a screening tool and provides an alternate to traditional, complex, and more costly whole animal testing with invertebrates and fish; the manufacturer’s suggested applications are listed in Table 1. Microtox uses very few elements: the Reagent (a specific bacterial strain of Vibrio fischeri), the test sample in compatible carrier solution, the Diluent test solutions, a duo-function Analyzer that includes an incubator and luminometer, a personal computer, and a data capturing and analyzing MicrotoxOmni software package.

“A simple rapid method for monitoring the toxicity of aquatic samples has been developed” (Bulich, 1979); thus in 1979, in this short statement, the bacterial toxicity bioassay known as Microtox® ushered in a new far-reaching revolution in bioassays and a paradigm shift in test organisms and, most importantly, introduced a new

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1 Use of specific products by USGS and its laboratories does not constitute an endorsement. Columbia Environmental Research Center (CERC) uses Microtox materials and equipment sold by Strategic Diagnostics Inc. (SDI) in Newark, DE, to preserve the Microtox protocol. SDI provides comprehensive instructive guides, manuals and computer software to operate the Microtox test at their Web site (www.azurenv.com). The Microtox protocol described here is a standard USGS SOP.

2 The USGS as well as others (Environment Canada, 1992) adopted Microtox terminology to reduce confusion. Specific Microtox products are printed in italics with the initial letter in upper case.
microscale biomonitoring tool in environmental toxicology. Over the last twenty-five years bacterial toxicity bioassays have emerged as important screening tools for toxicity assessments, for regulatory compliance, and for use in a battery of tests to rapidly monitor the health hazards and risks of chemicals that enter the nation’s aquatic environment (Wells et al., 1998). This chapter describes Microtox, an ecotoxicological screening tool designed to detect aquatic toxicity, to detect changes in toxicity, and to predict expectations of other toxicity tests. The advantages, new and old applications, and limitations of Microtox are explored.

Table 1. Recommended applications for Microtox (SDI Web site, 2003).

- Wastewater treatment plant influent testing for protection of activated sludge.
- Wastewater treatment plant effluent testing for protection of receiving waters.
- Toxicity Reduction Evaluations (TREs) and Toxicity Identification Evaluations (TIEs).
- Surface water monitoring for identification of point source and non-point source pollution.
- Monitoring raw drinking water to detect contamination due to point source or non-point source pollution.
- Bioterrorism.
- Sediment and soil testing.
- Monitoring of remediation processes.
- Biocide monitoring of industrial processed waters.

Water by its very nature is a universal solvent, a natural repository, and a carrier of both biogenic and xenogenic chemicals. The magnitude of this problem is expressed in part in the U. S. Chemical Industry’s Statistical Handbook (1998) that states the industry annually produces 70,000 chemical products in 12,000 plants. The broad ecological impact of these and other chemicals on the health and well being of aquatic communities presents a very complex problem of hazard and risk assessment for both ecotoxicologists and resource managers.

In the last century analytical chemists have made amazing strides in collecting, separating, and identifying waterborne chemicals at nano- and picogram concentrations (Manahan, 1989). However, ecotoxicologists have only begun to make similar strides in the detection and characterization of environmental toxicants (Wells et al. 1998; Ostrander, 1996; Rand et al., 1995). The unraveling of contaminants (chemicals "out of place") and toxicants (chemicals injurious to ecosystem health) centers on three basic questions: What is the toxicant (qualitative)? How toxic is it (quantitative)? And how does the toxicant move (bioavailability)?