Title: Microtox Assessment of Anaerobic Bacterial Toxicants

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Objectives:

Anaerobic treatment processes have recently received increased attention owing to their advantages over conventional aerobic biological treatment. These include the production of smaller amounts of waste sludge and the production of methane gas as a utilizable energy source. One concern about the usage of this treatment method is the process reliabiilty regarding waste materials containing constituents which are toxic to the methanogenic microorganisms. Bioassay methods have therefore been developed to measure the presence or absence of inhibitory substances in wastewaters being treated by such anaerobic processes.

Techniques have been developed to monitor the possible toxicity of constituents in feed sources to anaerobic treatment processes. This batch anaerobic toxicity assay (ATA) measures the adverse effect of a complex or complex mixture on the rate of the total gas production from an easily, utilized, methanogenic substrate. While simple and inexpensive, the method is time consuming and tedious. Alternatively, Beckman Instruments has developed a toxicity analyzer utilizing bioluminescent bacteria to determine the toxicity of certain chemicals. The assay is based upon changes in light output of the bacteria measured photometrically. Thus, on exposure to toxic substances, the light intensity of luminescent bacterial suspensions is rapidly diminished in direct proportion to the toxicant concentration. The categories of chemicals which have been observed to most actively diminish bacterial luminescence are surface agents, organic solvents, heavy metals and antibotics.

The Microtox bioluminescence toxicity analyzer is a fast, relible, and reproducible method. Correlations between acute fish and Daphnia toxicity with Microtox have already been established.

The objective of the proposed research, is to investigate Microtox as a possible surrogate parameter to the ATA in evaluating the toxicity of chemicals which inhibit the growth or metabolism of anaerobic bacteria. Such a method would influence current procedures for monitoring toxicant plugs to anaerobic methanogenic unit proesses in wastewater treatment.

Procedure:

Various compounds including some heavy metals and organic solvents will be monitored for possible anaerobic toxicity using both the ATA and "Microtox" methods. Defined media, containing nutrients and vitamins for mixed anaerobic cultures and containing a methanogenic bacterial innoculum will be equilibrated to assay temperature and transferred to serum bottles, containing test toxicant compounds, for ATA. Bacterial inhibition will be monitored by measuring gas production relative to a toxicant-free

control, over the incubation test period. Alternatively 5EC50 values (concentration of toxicant causing a 50 percent reduction in light intensity after five minutes) will be determined for these same chemicals using the "Microtox" toxicity analyzer.

Following the ATA results, the Microtox will be correlated with conventional process control parameters such as gas production and organic acids concentrations. This will be done with laboratory fill and draw units.

Expected Results:

The correlation between ATA results and "Microtox" EC50 values for chemicals known to inhibit anaerobic biological treatment processes, will be established and presented in a technical report.

Cost: $28,000