<u>ABSTRACT</u>

The quest for a rapid bioassay for oil-contaminated soil was prompted by the expense and turn-around time of tests prescribed by current regulations, which aim to quantify the toxicity of such materials towards organisms that represent various soil ecosystem levels.

In Alberta Environment's Eco-Contact Guideline, the Weight of Evidence approach requires, as well as tests with other species, earthworm reproduction bioassays which run for several months and can be costly, depending on the number of samples required per site.

The rapid bioassay developed in the course of this project requires only two working days, and will cost less than \$1,000 per sample. It involves shaking soil with aqueous cyclodextrin (CD), which extracts bio-available petroleum hydrocarbons (PHC) by forming an association complex.

After centrifugation, the supernatant CD extract is pasteurized to suppress natural soil enzyme activity, then a portion is treated with amylase enzyme and incubated in darkness at room temperature for 20 h to uncouple CD from any associated PHC.

A standard Microtox bioassay using the test organism *Vibrio fischeri* is then done, to assess extract toxicity.

Eight PHC-contaminated soils were donated to the project by CAPP member companies and their contractors, and a control soil was spiked with various rates of weathered diesel fuel.

Soils with total PHC concentrations between 1,000 - 20,000 mg/kg gave CD-extracts with 20 - 400 mg/L. After amylase treatment these extracts, and CD solutions spiked with diesel in that same mg/L range, were toxic towards *V. fischeri* in proportion to the level of PHC contained.

Earthworm bioassays were also done on all soils but, unfortunately, no correlations could be drawn between these bioassay results and CD-extract toxicity to *V. fischeri*. The donated soils inhibited earthworm reproduction independent of PHC level; this effect may be related to soil texture. Organic amendments to subsoils to eliminate this confounding factor will be investigated in follow-up work (Phase 2), in which we plan to do similar bioassays on a range of true Tier-2 candidate soils (see below).