Earthworm Bioassay

Contaminant bioaccumulation and toxicity are often poorly related to total soil contaminant content. Contaminant bioavailability may be a better predictor. Soil properties and site remediation may affect metal bioavailability to ecological receptors. Therefore contaminant bioavailability varies among soil types and may depend on one or a combination of soil properties. A single total contaminant level can result in multiple contaminant exposure doses across different soils due to modification by soil properties or modification due to in situ remediation.

During the bioassay, it is important to ensure, as far as possible, that variation in earthworm response is due to contaminant bioavailability and not due to uncontrolled variables such as problems with soil quality (extremes of pH or salinity, water availability etc.) or variation in earthworm food (organic carbon) availability. If these variables are not controlled, it will be impossible to assess the effect of contaminant bioavailability on the earthworm endpoint. Thus, control of soil quality and earthworm food supply is essential to make meaningful comparisons between or among different soils or even within a single soil for before and after remediation comparisons. Therefore, before the bioassay is conducted:

- Remove excess salt and/or adjust the pH of the contaminated soil
- Ensure soil has adequate moisture
- Ensure sufficient earthworm food availability
- For heavy or compacted soils add inert bulking agent (i.e., vermiculite, perlite) to improve soil aeration and drainage.

Depurated vs. Non-Depurated

For contaminant bioavailability studies, earthworms should be depurated and rinsed prior to analysis. For contaminant toxicity studies, earthworms should not be depurated prior to analysis.

Calibrated In Vitro Surrogate Methods for Earthworm Bioassay

Several tests that measure direct exposure include both in vivo bioassays (i.e., earthworm, other soil invertebrate, or swine—human surrogate) and in vitro soil tests (i.e., laboratory soil chemical methods correlated with in vivo models). In vivo bioassay tests are more expensive and require more time to conduct (i.e., weeks to months) than in vitro soil tests. However, a critical prerequisite for use of in vitro methods to predict in vivo endpoints is documentation demonstrating the in vitro method is strongly correlated with the in vivo endpoint. On a limited budget, more soil samples can be tested by in vitro soil tests than in vivo
bioassays. Analysis of greater numbers of samples allows a more thorough characterization of contaminant potential bioavailability/toxicity or for evaluating remedial success. Grid sampling for bioavailability/toxicity testing is possible using \textit{in vitro} methods but not feasible using bioassays. A strong relationship between \textit{in vitro} and \textit{in vivo} is a prerequisite for using \textit{in vitro} to predict \textit{in vivo} (as a surrogate method) while limiting the onus and expense of multiple bioassays.

A "calibrated bioassay" is a test where soils have paired \textit{in vivo} and \textit{in vitro} measures in order to determine if a robust relationship between them can be established. \textit{In vitro} extractions that may be predictive of earthworm endpoints include weak neutral salt extractions such as 0.01 M CaCl$_2$, KNO$_3$, or NH$_4$NO$_3$ which mimic soil solution (pore water) contaminant concentration. This may provide a measure of the contaminant dose the earthworm is directly exposed to and is often highly related to earthworm response. Other soil extraction methods correlated with earthworm biological endpoints include those based on solutions containing chelates, such as diethylenetriaminepentaacetic acid (DTPA). Once a robust relationship between \textit{in vivo} and \textit{in vitro} measures the regression equation can be used to predict contaminant bioavailability/toxicity across a site.

The extensive use of such methods to estimate trace element availability in residual-treated soils has been reviewed by McLaughlin (2002) and Pierzynski (1998).

\textbf{References}


