

The use of bioassays for evaluating the toxicity of sewage sludge and sewage sludge-amended soil

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Abstract

Background and Aims. In soils, the most commonly mentioned hazardous substances are metals. One of the sources of its accumulation is the application of sewage sludge. However, little information is available regarding the estimation of the toxicity of sewage sludge or soil treated with sewage sludge, even by means of a battery of bioassays. In this study an evaluation of a battery of bioassays was carried out for toxicity assessment of sewage sludge and sewage sludge-treated soil. The objectives of this study were a) to compare the sensitivity of the different bioassays for the toxicity determination of sewage sludge contaminated with metals and soil treated with this sewage sludge, b) to elaborate a procedure for the attribution of sewage sludge samples to hazard classes based on the ecotoxicological data, and c) to evaluate the suitability of elutriate bioassays and microbial toxicity tests for the assessment of sewage sludge-treated soil. **Methods.** Experiments were carried out on grey forest soil (Haplic Greyzem) treated with existent sewage sludge obtained from the Municipal Wastewater Treatment Plant of the city of Kazan, Tatarstan, Russia. Portions of sludge were preliminary spiked by adding water-soluble chlorides or nitrates containing the following metals/metalloids: As, Cd, Cr, Cu, Hg, Mn, Ni, Pb, Zn. The test battery included four organisms -bacterium *Pseudomonas putida*, protozoan *Paramecium caudatum*, water flea *Daphnia magna* and higher plant *Raphanus sativus*. In addition, the toxicity of the soils treated with sludge was measured using microbial bioassays based on the estimation of microbial respiration, microbial biomass, N₂-fixation activity and calculation of the metabolic quotient. **Results and Discussion.** The comparison of the toxicity values (expressed as the lowest dilution factor which exhibits 10% inhibition of an estimated parameter) of three model samples of sewage sludge prepared by us showed different sensitivities for all test organisms. The most sensitive was observed in the *Daphnia* test, while a similar sensitivity was noted for the *Paramecium* test. The other two tests proved to be less sensitive to the components of the sludge. The analysis of the soils treated with the sewage sludge samples, using the previously mentioned tests, showed that all metal-contaminated sludge treatments as well as the control soil were not significantly inhibitory to all elutriate tests except for that with *Daphnia*. The microbial toxicity studies including estimation of the microbial biomass C, respiration and N₂-fixating activity appear to be more sensitive than the elutriate tests for toxicity in the case of soil treated with metal-contaminated sewage sludge. **Conclusion.** An integrated cotoxicological approach to classify the sewage sludge into hazard classes is proposed based on toxicological analyses. The test battery including four bioassays is elaborated to establish the toxicity status of the sewage sludge. The proposed procedure is easy to apply, cost effective and relatively fast. The application of microbial toxicity tests can be very useful for risk assessment in the case of soil treated with metal-contaminated sewage sludge. **Recommendation and Outlook.** The application of the bioassays appears to be very promising mainly for the risk identification of highly contaminated matrices with hardly

identified composition such as sewage sludge or soil-associated contaminants. However, the future success in elaborating reliable toxicity tests can be achieved only by the progress in standardizing, national legislators and efforts of researchers.

Keywords

Bioassay, *Daphnia magna*, Metals, *Paramecium caudatum*, *Pseudomonas putida*, *Raphanus sativus*, Sewage sludge, Soil