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## Bioaccumulation of Zn and Ag Nanoparticles in the Earthworms (*Eisenia fetida*)

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Many studies are carried out to evaluate environmental effects of engineered nanoparticles (ENPs). Most of the previous studies primarily focused on the effects of nanoparticles into the aquatic environment and human. Model studies predict that ENPs released into environment would transferred primarily to the soil of the terrestrial environment. Despite this prediction, biogeochemical behavior of ENPs in soil environment as well as bioavailability of ENPs to soil-dwelling organisms such as earthworm, springtail, isopod and nematodes are poorly understood.

The main goal of this study was to compare the bioaccumulation factor (BAFs) and subcellular partitioning of nanoparticles in the soil-dwelling earthworm (*Eisenia fetida*) from ENP (ZnO and Ag nanoparticles) or ionic metal (Zn<sup>2+</sup>, Ag<sup>+</sup>) contaminated soil. And the sequential extraction was also used to determine the mobility of metals in soil which could be used as to predict bioavailability and compare that with bioaccumulation factor. The radiotracer method was employed to trace the transfer of ENPs and ionic metal among different environmental media and animals. Radiolabeled  $^{65}$ ZnO,  $^{110m}$ AgNPs coated with PVP or citrate were synthesized in the laboratory and their chemical and biological behavior was compared to ionic  $^{65}$ Zn and  $^{110m}$ Ag.

The BAFs of Zn and Ag in the earthworms were determined after animals exposed to the contaminated soils. After the 7 days of elimination phase, subcellular partitioning of metals were also obtained. BAF for ZnO(0.06) was 31 times lower than that for Zn ion (1.86), suggesting that ZnO was less bioavailable than its ionic form from contaminated soil. On the other hands, BAFs for AgNPs coated with PVP (0.12) or with citrate (0.11) were comparable to those for Ag ion (0.17), indicating that Ag from contaminated soil was bioavailable in a similar rate regardless of chemical forms. The subcellular partitioning results showed that bioaccumulated Zn from Zn ion and ZnO contaminated soil were present mainly in HSP (heat-sensitive protein) while cellullar Ag from Ag ion and AgNPs (Ag/PVP, Ag/citrate) treatments were found mostly in cellular debris. No statistical difference in partitioning of metals among different subcelluar pools was found between the metal forms. Zn from ZnO contaminated solis was found largely in carbonate fraction (41%), while Zn from Zn ion treatment was found in Fe-Mn Oxide (29%). Association of Zn to mobile fractions (ZnO; 65%, Zn ion; 35%) suggest that Zn from ZnO contaminated soil would be more bioavailable than that from Zn ion treatment. However, the BAFs for Zn in the animals did not follow this prediction. Majority of Ag from AgNPs or Ag ion contaminated soil was bound mainly to biologically inert fractions mainly in organic matter, surphide fractions, and residual fractions. Consistent with these findings, the BAFs of Ag in the worms exposed to Ag contaminated soils were generally lower than those for Zn treatments.