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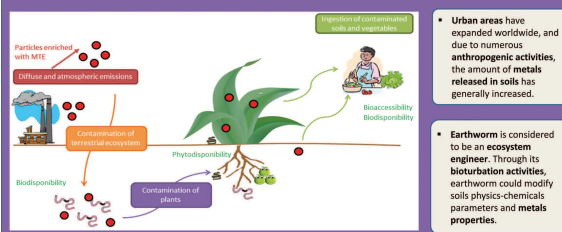
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Earthworm's influence on phytoavailability and Human gastric bioaccessibility of metals

Scientific context



Objectives

- Assess the impact of earthworm's bioturbation on 2 primordial notions governing the level of exposure of human population to metals: Phytoavailability and Human gastric Bioaccessibility.
- Improve the understanding of the mechanisms involved.

Materials & Methods

Soil preparation

- Sampling of a highly contaminated soil diluted with an uncontaminated soil
- 4 MTE concentrations in soils (C0, C1, C2, C3)
- Earthworm *L. terrestris* used to create 3 different conditions of soils:
 - Control soil (SNB)
 - Bioturbated soil (SB)
 - Earthworms casts (T)

Culture experiment

- Lettuce (*Lactuca sativa*)
- Culture with RHYZOTest[®] in climatized chamber
- 9 replicates on each soil condition and concentration = 108 plants

Mechanisms understanding

- XANES (X-ray Absorption Near Edge Structure spectrometry) and EXAFS (Extended X-Ray Absorption Fine Structure) spectrometry of Pb in the different soils conditions in Grenoble synchrotron: **Pb speciation analysis.**

Results and Discussion

Table 1: MTE concentrations in soils in mg.kg⁻¹

(ppm)	Pb	Cd	Cu	Zn
C ₀	20.4	0.11	13.1	72.5
C ₁	755.9	1.39	18.4	94.4
C ₂	2886.6	5.15	30.8	126.8
C ₃	4383.2	7.5	39.1	147.7

- Expect for Zn, no significant difference was observed on the MTE Human gastric bioaccessibility in the different soils conditions.
- For Zn bioaccessibility, the same trend is observed as for MTE phytoavailability with SB > T > SNB.
- Earthworm's bioturbation increase the Human gastric bioaccessibility of Zn in soils.
- Human gastric Bioaccessibility of MTE in lettuce's shoots is not impacted by earthworm's bioturbation.

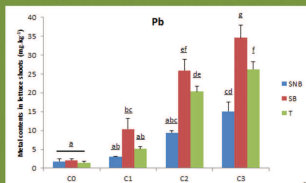


Figure 1: MTE concentrations in shoots of lettuce grown on the different soils conditions (Control soil: SNB, Bioturbated soil: SB and earthworms casts: T) and concentrations of MTE (C0, C1, C2, C3). Bars sharing the same letter are not significantly different (p<0.01 Tukey's HSD test).

- MTE contents in lettuce shoots are correlated with soils concentrations for pollutants like Pb and Cd ($R^2=0.88$ & $R^2=0.83$, $p<0.01$ respectively) but it is poorly correlated for essential elements like Cu and Zn ($R^2=0.35$ & $R^2=0.39$, $p<0.05$ respectively). Meaning that plants can hardly regulate their internal concentration of pollutants.
- Significant differences have been observed for MTE contents in lettuce's shoots cultivated on the different soil conditions. Generally, MTE contents in lettuce's ranged in that order SB> T> SNB.
- Earthworm's bioturbation increase phytoavailability of pollutants like Pb and Cd but also essential elements like Cu and Zn.
- Bioturbated soil (SB) is the soil where the earthworms lived (but without the earthworm's casts removed to create condition T). Meaning that soil pathway through the digestive system of the earthworms was not the only cause for MTE phytoavailability modifications.
- MTE speciation changes could be one of the numerous mechanisms responsible of the MTE phytoavailability modifications induced by earthworm's bioturbation. Other mechanisms: Macroporosity creation; Casts MTE enriched; Soil pH modification; Soil organic matter content changes, Symbiosis with microbial communities etc...

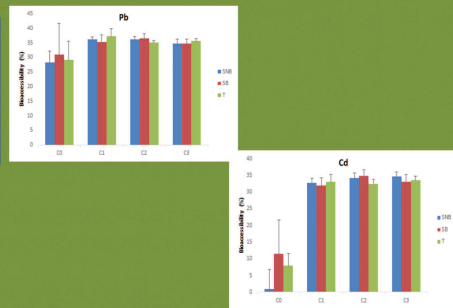
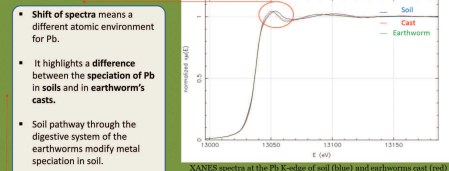
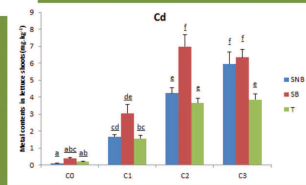


Figure 2: MTE Human gastric bioaccessibility in the different soils conditions (Control soil: SNB, Bioturbated soil: SB and earthworms casts: T) and concentrations of MTE (C0, C1, C2, C3).



- Shift of spectra means a different atomic environment for Pb.
- It highlights a difference between the speciation of Pb in soils and in earthworm's casts.
- Soil pathway through the digestive system of the earthworms modify metal speciation in soil.

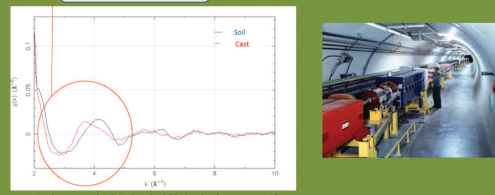


Figure 3: XANES and EXAFS spectra at the Pb K-edge of soil (blue) and earthworm's casts (red).

- Earthworm's bioturbation ↑ phytoavailability of pollutants like Pb and Cd but also essential elements like Cu and Zn.
- Bioturbated soils have more impact on MTE phytoavailability than earthworm's casts alone.
- Numerous mechanisms involved: MTE speciation changes etc.
- Earthworm's bioturbation increase Human gastric Zn bioaccessibility.
- Earthworm's bioturbation do not impact MTE bioaccessibility in lettuces.