

# IMPACTS OF HEAVY METAL ENRICHED BIOSOLIDS ON NITROGEN MINERALIZATION IN TWO GEORGIA SOILS

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## ABSTRACT

The effects of Zn, Cu, and Ni added with biosolid in two soils (Ultisols, Cecil and Tifton series) on N mineralization were studied. Two experiments were carried out: (i) an incubation of soil-enriched biosolid mixtures where ammonification and nitrification were periodically assessed; (ii) soil-enriched biosolid mixtures were leached with water in order to observe nitrification and metal release into the soil solution over time. Nitrification was much more heavy metal sensitive than ammonification. Zinc was the metal with the strongest inhibitory effect on nitrification. Nickel was not inhibitory and the Cu effect was intermediate. Zinc thresholds were: 38.1 and 35.1 mg kg<sup>-1</sup> Zn extractable with 0.05 M CaCl<sub>2</sub> caused full inhibition, whereas 5 to 19 and 6.6 to 10.5 mg water leached Zn kg<sup>-1</sup> soil caused partial<sup>2</sup> inhibition of nitrification in the Cecil and Tifton soil, respectively. Metal release into soil solution appeared to be related with C and N cycles.

## INTRODUCTION

Biosolid application on agricultural land and landfills can lead to soil contamination with heavy metals. Nutrient cycles are affected and the effects on the N cycle are often concentrated on N fixation. Relatively less attention has been paid to N mineralization steps like ammonification or nitrification. Therefore these two steps are considered as indices of heavy metal impact in this work. Metals extractable with “weak” extractants (water being “the weakest”), which are *a priori* more strongly related to the actual bioavailable metal portion, will be the focal point of the following experiments.

## MATERIALS AND METHOD

### Experiment 1 (without water leaching)

Biosolid spiked with chlorides of Zn, Cu, and Ni was mixed with two soils from Georgia (Ultisol) at a ratio 98.5:1.5 (w/w) to obtain the following treatments: (i) a mixture (Zn + Cu + Ni) unit with 140 + 75 + 21 mg kg<sup>-1</sup> soil respectively applied as 1, 2.5, 5, 7.5, and 10 fold and, (ii) single metals as Zn at 700 and 1400, Cu at 375 and 750, and Ni at 105 and 210 mg kg<sup>-1</sup> soil. Controls with and without biosolids were prepared. Each treatment (100 g on dry basis) was placed in a Mason jar, incubated for 14 days at 25°C and water potential between - 0.021 and - 0.024 MPa. Then urea was spiked at 100 mg N kg<sup>-1</sup> soil and incubation continued as above. Net ammonification and nitrification rates were measured in time sequences, by taking soil portions from the Masons jars and analyzing for NH<sup>+</sup> and NO<sup>-</sup>. Respiration was estimated by measurement of daily CO<sub>2</sub> evolution in the 3-rd, 22-nd, 4<sup>th</sup>, and 46-th day after urea addition. For this NaOH traps placed in the tightly closed Mason jars were used, which reacted with the evolved CO<sub>2</sub>. Entrapped CO<sub>2</sub> was precipitated with BaCl<sub>2</sub> and the solution was titrated with HCl. At the end of<sup>2</sup> experiment (75<sup>2</sup> days after urea addition) the 0.05M CaCl<sub>2</sub> extractable metals were determined.

### Experiment 2 (with water leaching)

The soils, biosolids, their mixture ratio, and the incubation procedure were the same as in the Experiment 1. Three replicates were used for leaching procedure and one for periodic sequential extraction for metals. Each treatment for leaching weighed 10 g on a dry basis. The applied spiking rates created a range of treatments from 0 to 1500 mg Zn kg<sup>-1</sup> soil and from 0 to 220 mg Ni kg<sup>-1</sup> (in equal incremental steps of 150 mg Zn kg<sup>-1</sup> and 20 mg Ni kg<sup>-1</sup>, respectively). Soil-biosolid mixtures were placed in funnels and the moisture was maintained constant by daily replacement of the weight loss with water. Ammonium sulfate was added at 100 mg N kg<sup>-1</sup> soil after 14 days of soil-biosolid mixture incubation. Water was leached periodically (ratio water:soil was 1.5:1 on weight basis) and the leachate was analyzed for NO<sup>-</sup> and for metals. After six leaching sequences another portion of (NH<sup>+</sup>)<sub>2</sub>SO<sub>4</sub> was added at same amount<sup>3</sup> and followed by the same procedure as above.

<sup>4 2</sup> The fourth replicate weighed 20 g. and was used to extract sequentially the added metals, first with 0.05M CaCl<sub>2</sub>, followed by 0.05M EDTA. These extractions were done 7, 13, 30, and 50 days after the beginning<sup>2</sup> of first incubation.

## RESULTS AND DISCUSSION

### Experiment 1

Ammonification and nitrification were both adversely affected by the metal loadings, nitrification being much more sensitive than ammonification. The contour plots in Fig. 1 present these relations over time.

The estimation of ammonification is complete only if related to nitrification. Ammonium accumulation after 75 days of urea addition in both soils, showed a parabolic trend (Fig. 2). The initial increase in NH<sup>+</sup> accumulation was due to the decrease in nitrification rates at those metal levels. After the maximum NH<sup>+</sup> accumulation was reached, the inhibitory effect of added metals on ammonification began to appear<sup>4</sup>.

Respiration rates initially depended on metal levels, but later the differences between treatments were not statistically significant; therefore respiration, like ammonification, was not a better direct index than nitrification to characterize soil pollution with the applied metals.

The concentration of the fraction extractable with 0.05 M CaCl<sub>2</sub>, at which nitrification was severely inhibited, was similar for both Zn and Ni in each soil. Comparison of the effects of the single metals and their mixtures on nitrification over time (Fig. 3) showed that Zn was the strongest inhibitor. Nickel enhanced nitrification in any case, whereas Cu effect was intermediate.

### Experiment 2

Zinc and Ni released into the leachate over time were initially strongly related with the amount of added metals (Fig. 4). Water soluble fraction should be the main constituent of leached Zn and Ni at this stage. At the end of first incubation period this relation changed and another mechanism was likely to determine the metal released. Presumably, the microbial decomposition of organic matter was the main mechanism influencing the metal release at this stage. Because this decomposition is directly influenced by the amounts of metals available, "self-inhibition" of metal release is likely to occur. This explains why by the end of first incubation and in the second incubation more added Zn unexpectedly brought about less Zn in solution. The decomposition rate is controlled by the available C/N ratio. This could explain the increase of metals in solution with the second addition of ammonium sulfate (an exchange reaction here seems not to be plausible), and also the different

release pattern obtained with Ni, which did not inhibited nitrification like Zn. In the Cecil soil Zn levels with up to 5 mg leached Zn kg<sup>-1</sup> soil stimulated nitrification, 5 to 19 mg leached Zn kg<sup>-1</sup> soil caused partial inhibition, and over this range the inhibition lasted longer than the first leaching period. For the Tifton soil these ranges were narrower, as 6.6 to 10.5 mg leached Zn kg<sup>-1</sup> soil caused partial inhibition. Zinc fractions extracted with CaCl<sub>2</sub> or EDTA did not change substantially over time; therefore their use would not reveal any significant relations between Zn and nitrification.

Sequential measurements of nitrification rates in marginal Zn treatments showed the gradual way at which both inhibition and recovery of nitrification occurred. These observations indicated that the Zn effect on nitrification does not have clear cutoffs in quantity and time. This is manifested by the delays, or lag phases in nitrification, as result of temporary, or partial inhibition before the full recovery of nitrifiers.

## CONCLUSIONS

Nitrification was more Zn and Cu sensitive than ammonification. Therefore nitrification could be better used as bioindicator of soil pollution with these metals than ammonification.

Zinc had the strongest inhibitory effect on N mineralization, followed by Cu at the rates used. The Ni effect involved soil related parameters and needs further and more detailed investigations.

The Cecil soil, with more clay and organic content (the Cecil soil), was more resilient to applied heavy metals effects than the Tifton soil. There is evidence for a protective action of soil colloidal components that mitigates the toxic effect of heavy metals to nitrifiers.

Concentrations of heavy metals in the soil solution were better related to the effects on nitrification than the fractions extracted with stronger extractants.

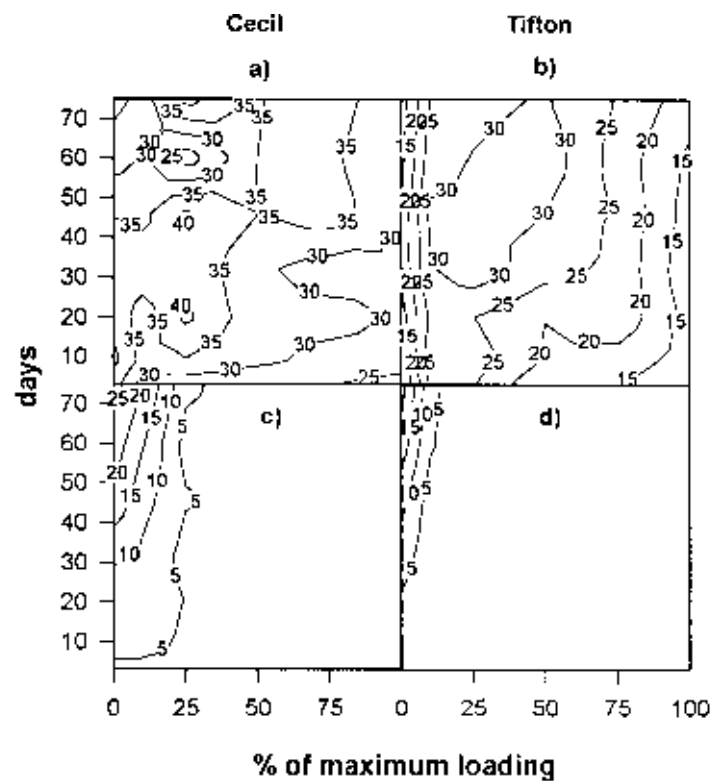
Determination of the metal thresholds for nitrification depended on the used extractants and soils. So, 5 to 19 mg initial water-leached Zn kg<sup>-1</sup> caused partial inhibition in the Cecil soil, while in the Tifton soil that interval was 6.6 to 10.5 mg kg<sup>-1</sup>. When extraction with 0.05M CaCl<sub>2</sub> was used (with no leachings) 38.1 mg Zn kg<sup>-1</sup> in the Cecil soil and 38.1 mg Zn kg<sup>-1</sup> in the Tifton soil<sup>2</sup> caused full inhibition. A unified procedure is needed to determine the thresholds of heavy metals in biosolid amended soils to substitute for the total amounts in soils, which are usually in use.

Zinc effect on nitrification needed some time to be complete in the marginal treatments dividing the full from the partial inhibition domains.

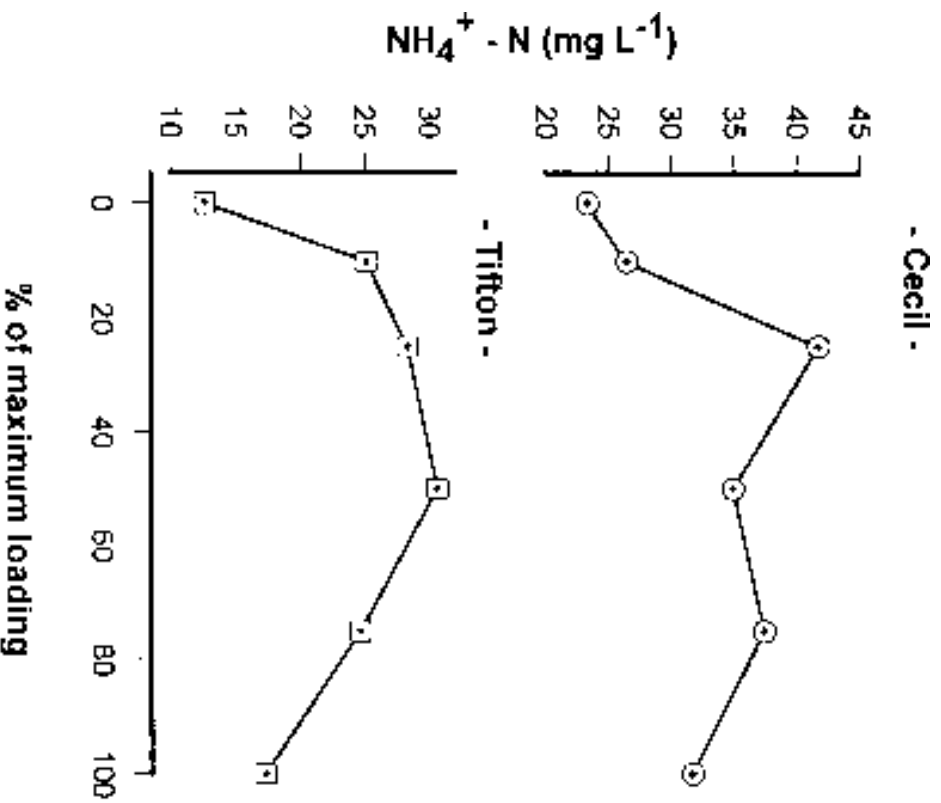
Heavy metal release into the soil solution appeared to be closely related with soil microbial activity. Therefore microbial activity inhibition results to a decrease of heavy metals in the soil solution. Applied biosolids with elevated content of heavy metals can remain for a long time a potential source for heavy metals in a latent state. Ecological functioning in biosolid amended soils becomes the last threshold concerning heavy metals. In addition, the low heavy metal concentration in the soil solution is not sufficient to consider a biosolid amended soil as safe from the adverse effects of heavy metals. It might be safe in terms of phytotoxicity, but meanwhile, in a latent way, soil microbial biomass could be under heavy metal stress. Possibly, soil microbes pay the price for such "safety". This is in accord with the conclusion drawn by Giller et al. (1997) about the long term toxic effect of heavy metals in biosolid amended soils, as revealed by C cycling parameters (Dahlin and Witter, 1998; Brookes et al., 1986).

## REFERENCE

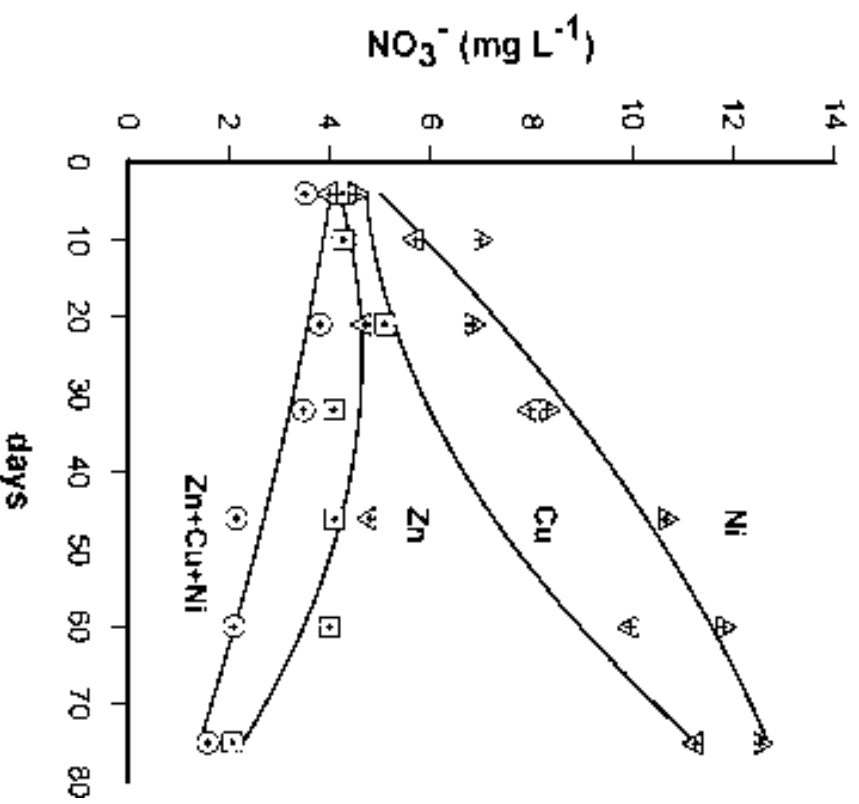
- Brookes, P. C., S. P. McGrath, and C. Heijnen. 1986. Metal residues in soils previously treated with sewage-sludge and their effects on growth and nitrogen fixation by blue-green algae. *Soil Biol. Biochem.* 18:345-353.
- Dahlin, S. and E. Witter. 1998. Can the low microbial biomass C-to-organic C ratio in an acid and a metal contaminated soil be explained by differences in the substrate utilization efficiency and maintenance requirements. *Soil Biol. Biochem.* 30:633-641.
- Giller, K. E., E. Witter, and S. P. McGrath. 1998. Toxicity of heavy metals to microorganisms and microbial processes in agricultural soils: A review. *Soil Biol. Biochem.* 30: 1389-1414.



**Fig. 1 Ammonification (a, b) and nitrification (c, d) in the Cecil and Tifton soils at different metal mixture loadings. Contours represent accumulated  $\text{NH}_4^+$  and  $\text{NO}_3^-$  -N in  $\text{mg kg}^{-1}$ , respectively.**



**Fig. 2** Ammonium accumulation at the end of incubation



**Fig. 3** Nitrification over time at 50% of metal loading in the Cecil soil

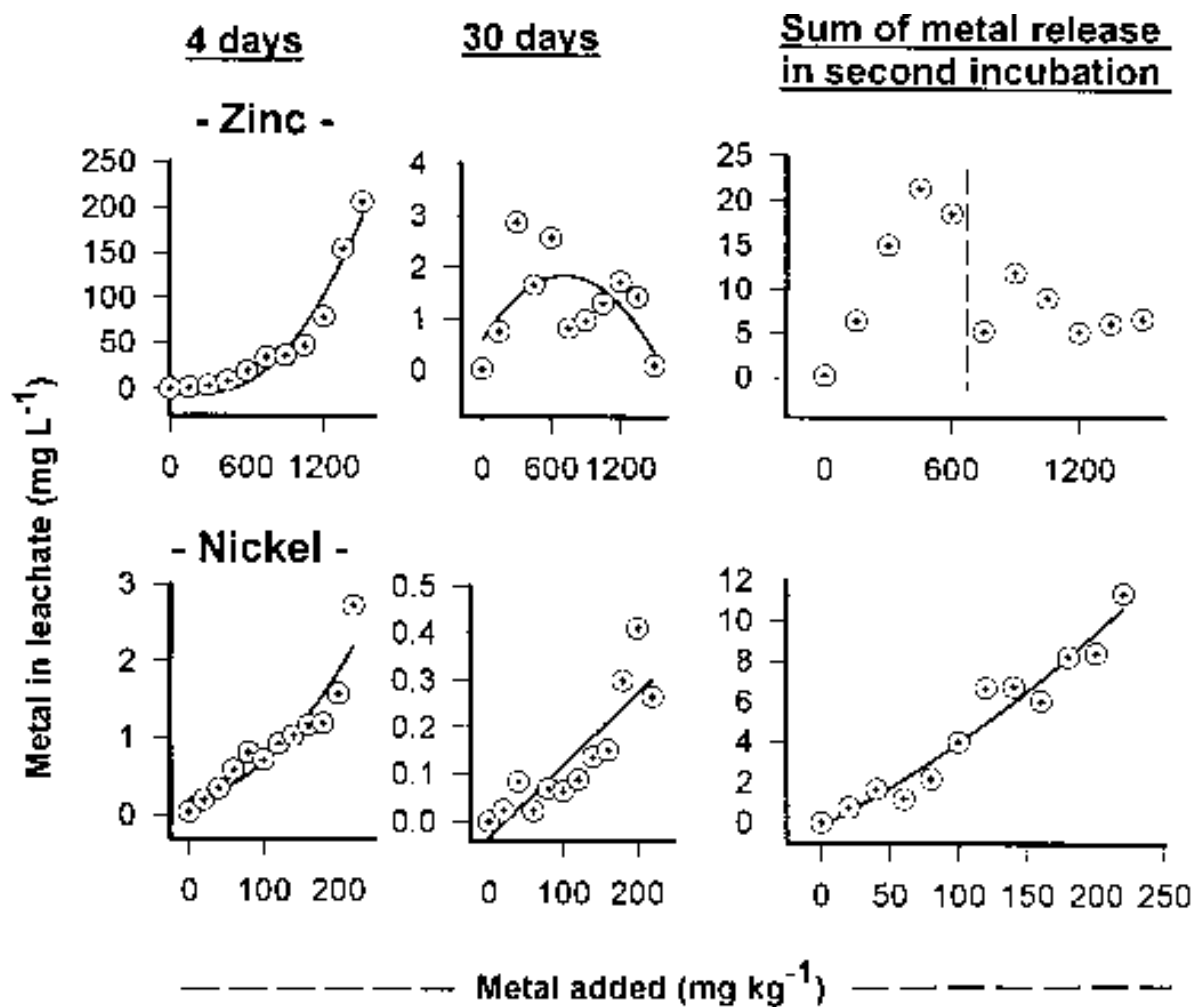


Fig. 4 Zinc and Ni released in the Cecil soil over time at different metal loadings