IMPROVING PHYTOSTABILISATION POTENTIAL OF POPLAR USING BIOCHAR AND PLANT GROWTH-PROMOTING BACTERIAL INOCULANTS

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INTRODUCTION

The use of organic and/or inorganic amendments in phytostabilisation techniques is often essential for improving the soil physicochemical properties and fertility, in order to promote plant establishment and reduce contaminant dispersion. Recent years have seen growing interest in the benefits of biochar as an amendment, due to its alkaline nature, high surface area and negatively charged functional groups, its application usually enhances soil cation exchange capacity and adsorbs positively charged metal ions (e.g. Cu^{2+}). However, it can affect the availability of nutrients (e.g. K, P) and increase soil salinity. Therefore, the use of biochar in phytostabilisation processes could be optimized by combining it with the application of other materials that can balance these negative effects.

Moreover, inoculation of plants with plant growth-promoting bacteria (PGPB) can induce beneficial effects on plant growth, health and resistance to stress, as well as improving water absorption, increasing nutrient availability and contributing to pollutant stabilization in the soil. The combination of all these options may enable successful plant establishment in contaminated soils and improve the overall efficiency of the remediation process.

OBJECTIVE

This study aimed at assessing the combined use of biochar (alone and mixed with composted sewage sludge and calcium carbonate) and inoculation with PGPB in the phytostabilisation of a copper (Cu) mine soil with poplar (*Populus maximowiczii* x *Populus trichocarpa* var. Skado).

The tested biochar was obtained from biomass of *Populus nigra* grown in the same mine soil in a previous field experiment; the aim being to revalorise the crops produced in contaminated soils.

Biochar

Composted sewage sludge

$CaCO_3$

EXPERIMENTAL DESIGN

A pot trial was carried out using mine tailings from the Touro Cu mine (NW Spain; pH <3.0; 42.5 g Al kg^{-1}; 596 mg Cu kg^{-1}).

A total of 25 treatments were carried out in pots of 1L (5 soil treatments x 5 plant inoculations) :

- Soil treatments were set up:
  i. UNT: untreated soil
  ii. CA: soil with $CaCO_3$ (at 1%)
  iii. B-CA: soil with biochar and $CaCO_3$ (at 2.5% and 1%, respectively)
  iv. CM-CA: soil with compost and $CaCO_3$ (at 2% and 1%, respectively)
  v. B-CM-CA: soil with a combination of the three amendments (at 2.5%, 2% and 1%, respectively).
- Each soil treatment was combined with the following rhizobacterial inoculants:
  i. NI: non-inoculated plants
  ii. SK2.3: *Bacillus* sp. strain SK2.3
  iii. SK12.6: *Rhodococcus* sp. strain SK12.6
  iv. SK20.12: *Streptomyces* sp. strain SK20.12
  v. P87: *Massilia niastensis* strain P87.

After the soil stabilization, previously inoculated poplar cuttings were planted. For plant inoculation, poplar cuttings were submerged in 250 mL of the corresponding inoculum (or sterile H_2O in case of non-inoculated treatments) and shaken for 12 h at room temperature before planting (Figure 1).

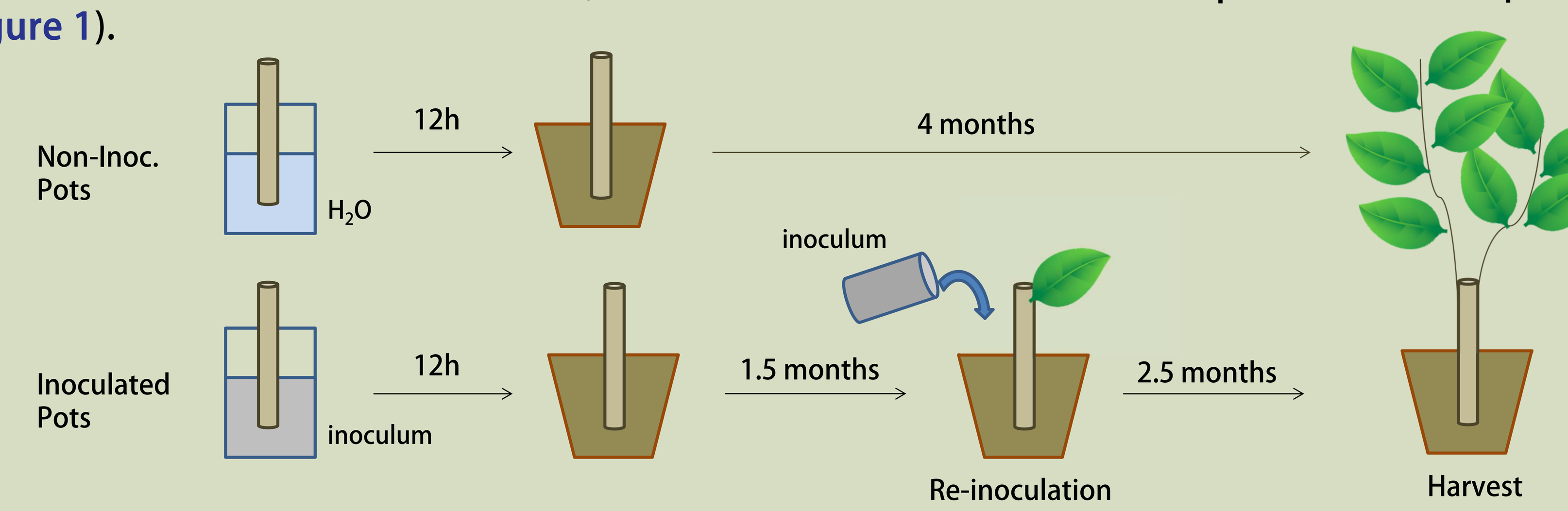

Figure 1. Experimental design.

Four months after plant transplantation, plant biomass production, number of leaves and leaf metal accumulation, as well as changes in soil properties and metal availability, were determined.

RESULTS

SOIL PROPERTIES

The amendment incorporation increased the soil pH from 3.0 to 4-4.8 (p>0.001; Figure 2), and the B-CM-CA treatment was most efficient. Additionally, soil amendments increased the cation exchange capacity of soil (p<0.001), enhancing the concentration of essential soil nutrients (Ca, K, and Mg; Figure 3).

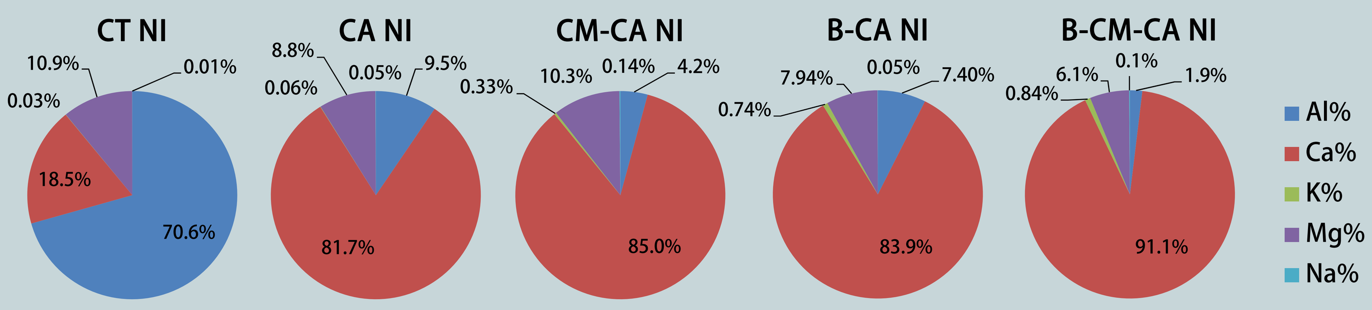

Figure 3. Proportion of cations bound to the exchange complex in the non-inoculated soils.

The untreated soil presented high extractable concentrations of Al, Cu and Zn (Figure 2). The amendment addition efficiently reduced their availability in soil (p<0.001 for all), but no inoculation effect was observed. Available-Al, -Cu and -Zn concentrations were reduced by 94-99%, 91-99% and 69-92%, respectively. The effect of the amendment treatments on metal availability followed the decreasing order: B-CM-CA > CM-CA > B-CA > CA.

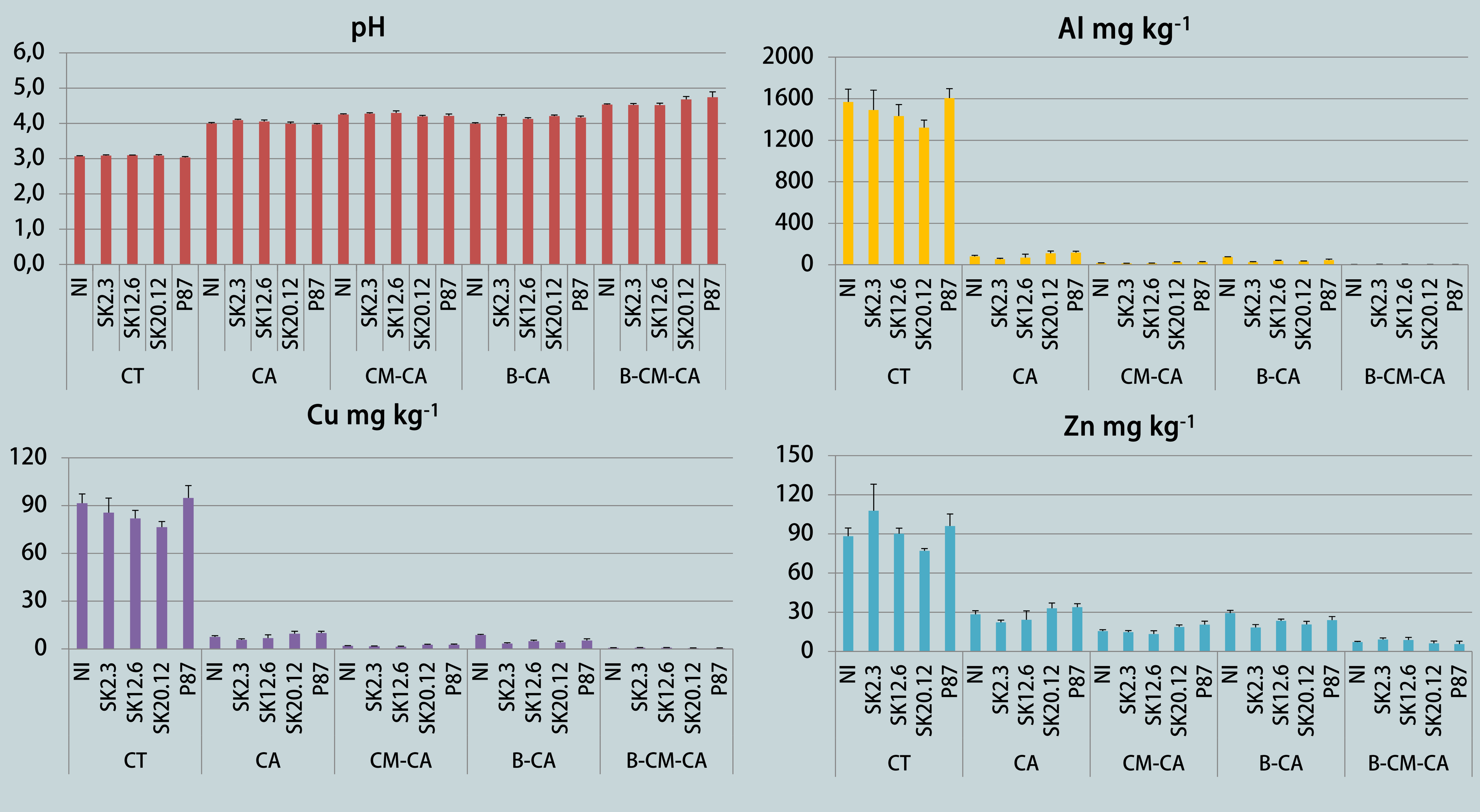

Figure 2. Soil pH and extractable concentrations of Al, Cu and Zn with $NaNO_3$..

PLANT PROPERTIES

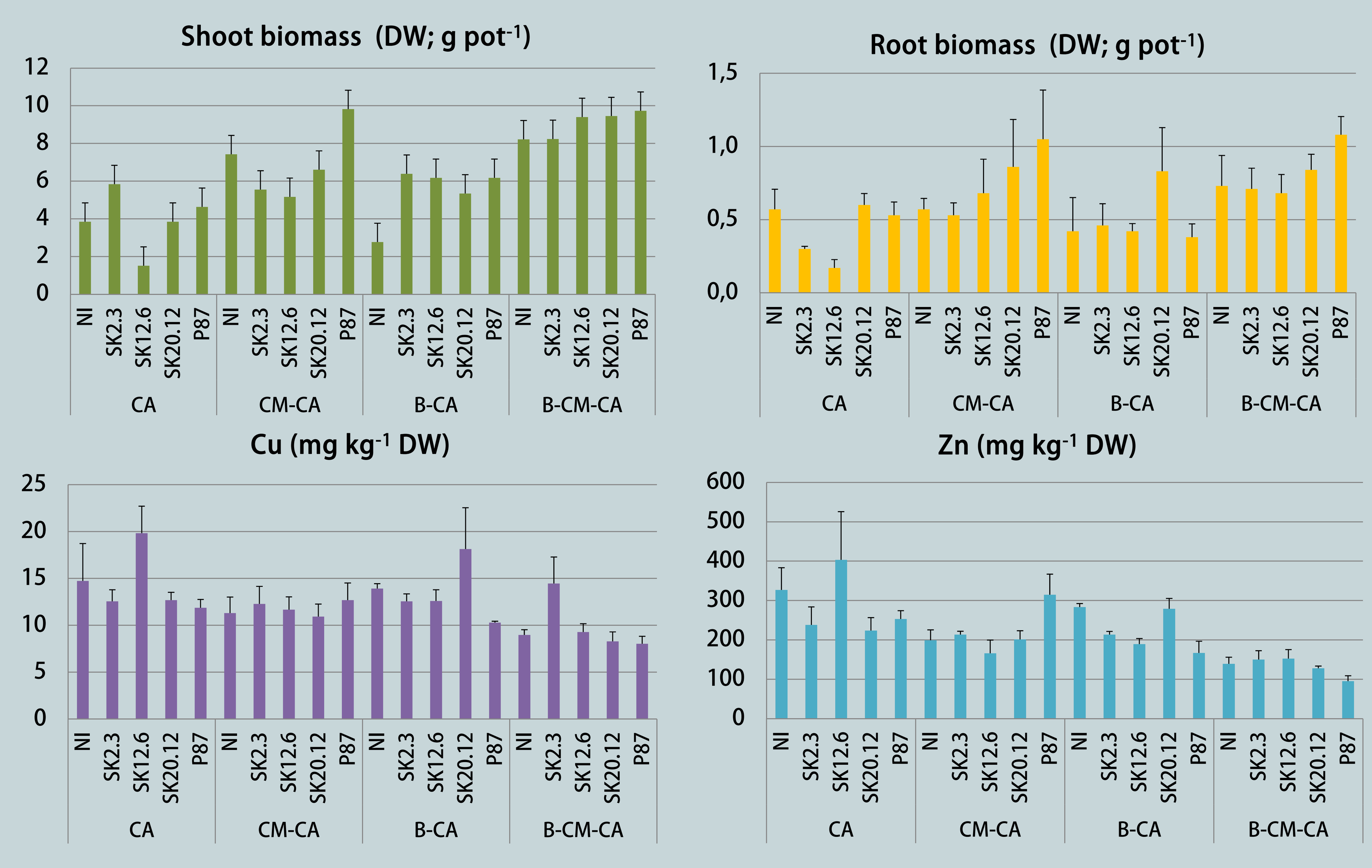

Figure 4. Shoot and root biomass production and Cu- and Zn-leaf concentrations reported in each treatment at the end of the experiment.

Plants were not able to survive in untreated soil without any amendments. The addition of the biochar- and compost-based mixtures increased the number of leaves compared to plants growing in CA-soils (data not shown), and the mixture of B-CM-CA significantly enhanced shoot and root biomasses (both p<0.05; Figure 4).

The inoculum effect on plant growth depended on the soil treatment, and was less evident when the soil received compost. Inoculation was especially effective in plants growing in B-CA-treated soils, where all strains increased the shoot biomass of poplar, and strain SK20.12 also increased root dry weight (Figure 4) .

Lower leaf concentrations of Cu and Zn were found in plants growing in compost-treated soils compared to CA-plants, especially in B-CM-CA-treated soils. In the B-CA-treatment, the inoculation of plants with the strain P87 reduced Cu and Zn concentrations in their leaves respect to non-inoculated plants, and the SK2.3 and SK12.6 strains only decreased leaf levels of Zn.

CONCLUSIONS

Application of the amendment combinations successfully improved soil conditions, reduced metal availability and allowed poplar development. Bacterial inoculants did not enhance plant growth when compost was added to soil, but were beneficial when biochar was not combined with another type of organic matter.

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