Potential Of Biostimulants To Increase *Sorghum Bicolor* Biomass Production And Phytoremediation Efficiency

Pietro Peroni[1], Walter Zegada-Lizarazu[1], Rossella Mastroberardino[1], Andrea Monti[1]

[1] DISTAL, Alma Mater Studiorum, Università di Bologna, pietro.peroni2@unibo.it; walter.zegadalizarazu@unibo.it; ross.mastroberardin2@unibo.it; a.monti@unibo.it

Introduction

The cultivation of biomass crops on contaminated lands is becoming more and more relevant thanks to the possibility of producing biomass with low ILUC risk, i.e. avoiding taking land away from food production, providing for the decontamination of polluted areas in an economic way and avoiding secondary pollution phenomena (Muthusaravan et al. 2018). *Sorghum bicolor* is one of the most promising species for the biomass production and it is considered capable of absorbing some widely diffused heavy metals in the aerial biomass, such as Pb, Cd, Cu and Zn (Zhuang et al. 2009) that can be precipated in transformation plants (Gong et al. 2018). Not being an hyperaccumulator species, the effectiveness of the process depends on the quantity of the biomass produced (Lima et al. 2019). Currently, in order to improve the efficiency of phytoremediation process research is particularly focused on the use of microorganism and biostimulants capable of improving the physiological well-being of crops and the bioavailability of heavy metals for roots. (Yan et al. 2020). The current study, carried out in the framework of the GOLD project, reports the results obtained by testing 6 different treatments (M, B1, B2, MB1, MB2, C) on sorghum in a greenhouse trial. The objective was to identify the best treatments able to increase the production of sorghum biomass and the relative uptake of heavy metals from the soil.

Materials and Methods

The trial was carried out between October 2021 and February 2022 in a greenhouse at the Department of Agricultural and Food Sciences of the University of Bologna. Six treatments, 3 replicates each, were tested on sorghum plants, (Bulldozer variety), grown in pots of 12l volume, after being germinated for 5 days in petri dishes (20-30 °C, 16 hours of light and 8 hours of darkness). The treatments tested were M= mycorrhizae (Symbivit, Symbiom, CZ) with a dosage of 15 g per pot applied at the start of the trial near the transplanted seedlings; B1= foliar biostimulant based on protein hydrolysates (Siapton, Agrology, GR), applied with a dosage of 3 ml*L^{-1} of application water by spraying the entire aerial part every 10 days, starting from 4 true leaves; B2= root biostimulant based on fulvic and humic acids (Lonite 80SP, Alba Milagro, IT) applied once a week in the irrigation water since plants have reached 10 cm in height, 0.5 g per pot the first 4 weeks an 0.7 g per pot thereafter; MB1= combination of M and B1; MB2 = combination of M and B2; C= untreated control. The sandy-loam soil was taken from a former landfill, “Chiarini 2” (44° 50’N, 11° 28’ E, 36 m a.s.l.) and was subjected to ICP-MS analysis to determine total concentrations of heavy metals and relative exceedances of the thresholds established by the Italian law (Table 1). Subsequently extraction with DPTA was performed to determine the bioavailable fraction (Table 1). The pots were randomly placed under the lamps (12 hours of light and darkness in the first month, 14 hours of light and 10 hours of darkness thereafter) and rotated every month. The temperature was kept between 18-26 °C and the soil humidity was kept at 75% of the pot water capacity. After 13 weeks form transplanting, all the plants were cut and their dry weight was determined. The biomass obtained was analysed by ICP-MS to detect the total content of heavy metals in each plant. The result were subjected to analysis of variance (ANOVA) and the LSD test was used for the separation of means ($p \leq 0.05$).

Table 1: Total concentration and bioavailable fraction of heavy metals exceeding the legal threshold of Italian law

Metal	Legal Threshold (mg/kg DM)	Total Concentration (mg/kg DM)	Bioavailable concentration (mg/kg DM)
Lead (Pb)	100	159	33
Tin (Sn)	1	8.8	Not detected
Zinc	150	455	62
Copper	120	137	45
Nickel	120	209	9,9

Results

The treatments applied in combination, MB1 and MB2, were the most productive for dry biomass: especially MB2 was found to be significantly more productive than B2(+ 66%), B1(+ 101%), M (+ 186%%) and C (+ 260%), while MB1 was significantly higher only than M (+118%) and C (+174 %) (Fig. 1). Among the treatments applied individually, B2 was significantly more productive than C (+116 %), while B1 and M were comparable to the control C. Zn and Cu were found in the aerial biomass and their total content closely followed the trend of biomass production indicating the most productive species such as those that have phytoextracted the greatest amount of heavy metals.

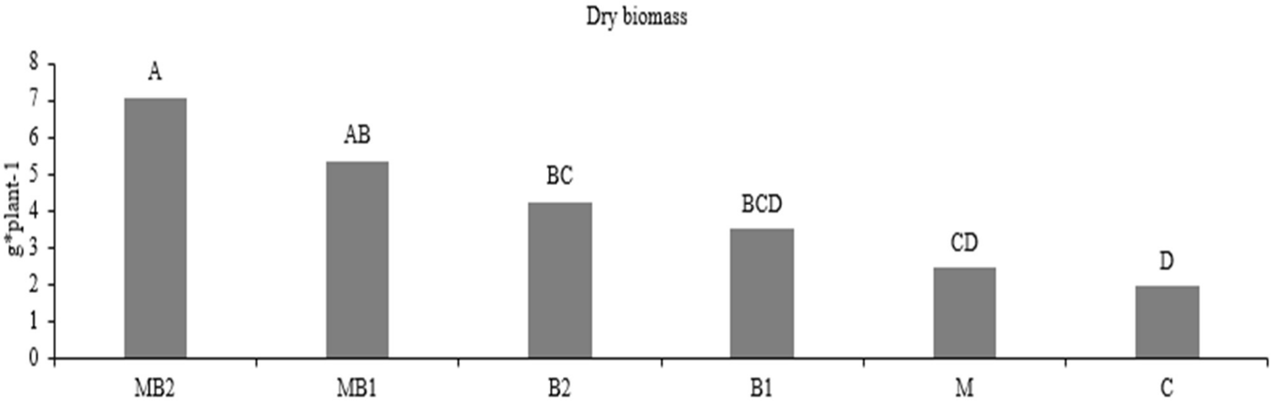

Figure 1: Dry biomass produced in each treatment

Conclusions

The combination of mycorrhizae and fulvic acids was found to be the best treatment to increase sorghum biomass production and heavy metals uptake, resulting in significant increases compared to all other treatments, apart from the combination of mychorrhizae and protein hydrolisates which was found comparable for the production of biomass. The metals absorbed were those used as micronutrients and most bioavailable in the soil used. However, the short duration of the test in a controlled environment in which the sorghum has not completed its biological cycle may have limited the adsorpiotn of other metals as the limit of quantification (> 5 mg/kg DM) prevented us to determine their actual presence (i.e. traces of Ni were detected). Further studies are ongoing to verify the results presented and the potential of the biostimulants identified in improve biomass production and heavy metal uptake in field condition.

Literature

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