Abstract

Screening of Autochthonous Microbial Strains for the Bioremediation of Heavy Metals †

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Keywords: bioremediation; soil; heavy metals

Numerous economic and industrial activities release significant amounts of heavy metals into the environment, with varying levels of toxicity depending on the concentration of exposure. Based on their physiological functionality, heavy metals may be essential (Fe, Zn, Mn, Co), vital in trace amounts for metabolic processes, or non-essential (Pb, Hg, As, Cr, Cd), and highly toxic even in low concentrations, causing mutations and multiple organ damage [1]. Common remediation methods can require high costs and energy, as well as generate high amounts of toxic wastes. Thus, biological methods have been intensively researched for their potential ecological and economic benefits [2]. The aim of this study was to isolate and identify metal-tolerant indigenous microbial strains that efficiently reduce the concentration of heavy metals for the purpose of bioremediating contaminated environments.

Soil samples were collected from Bucharest. The Cr, Pb and Zn content in soil was determined by ICP-OES. Microorganisms were isolated by serial dilutions on agar media supplemented with a low concentration of K₂Cr₂O₇, ZnSO₄ and Pb (NO₃)₂, respectively. The purified cultures were further identified using the Biolog MicroStation™. The tolerance of the isolated strains to Cr, Pb and Zn was assessed in broth and agar culture media containing 1000 mg L⁻¹ of the respective metal salts. Growth was observed after 24 h for bacteria and after 7 days for fungi. The samples were subsequently compared to the control, representing the microbial strain inoculated on unamended media. Values of the ratio ≥ 1 indicated a very high tolerance to the tested heavy metals [3]. Following the qualitative screening, one multi-metal tolerant strain was further selected from each group.
for the bioremediation assay. Bioremediation efficiency was tested in broth culture media supplemented with 100 mg L\(^{-1}\) of K\(_2\)Cr\(_2\)O\(_7\), ZnSO\(_4\) and Pb (NO\(_3\))\(_2\) individually. Samples were centrifuged after 72 h for the bacterial strain and 120 h for the fungal strain. The efficiency in reducing the heavy metal concentration was verified using electrochemical sensors modified with nanomaterials and amperometry techniques by comparing the initial and final concentration of the heavy metals in the supernatant.

The soil was confirmed to contain concentrations between 40 and 400 mg/kg of Cr, Pb and Zn, the latter being the most abundant. A total of seven Gram-positive bacteria, nine filamentous fungi and one yeast were isolated, 90% of the strains displaying a high tolerance to Pb. All bacterial strains were sensitive to Cr, while 30% of them displayed a high tolerance to Zn. On the other hand, 30% of the fungi were moderately tolerant to Cr and 40% of them were highly tolerant to Zn. In terms of remediation potential, the fungal strain, belonging to the genera *Trichoderma* sp., was efficient in reducing more than 80% of the concentration of the three metal salts tested.

Through our study, we isolated microbial strains from soil contaminated with heavy metals displaying various levels of tolerance to Pb, Cr and Zn. We observed the overall higher metal-tolerance of fungi compared to bacteria, as well as the efficiency of *Trichoderma* sp. in reducing the concentration of Cr, Pb and Zn in solution. Future studies will seek to optimize the bioremediation process.

**Author Contributions:** C.F., I.R., M.-L.J. (Maria-Lorena Jinga), L.C., D.P., A.M.B., L.-G.Z., A.-M.G., M.-L.J. (Maria-Luiza Jecu), M.D., R.C.F., M.C. and T.E.S. contributed equally to this paper. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research was funded by Ministry of Research and Digitalization of Romania and through Core Program PN 23.06.01.01.

**Institutional Review Board Statement:** Not applicable.

**Informed Consent Statement:** Not applicable.

**Data Availability Statement:** Not applicable.

**Acknowledgments:** The authors thank to the Ministry of Research, Innovation and Digitization through Program 1—Development of the national research and development system, Subprogram 1.2—Institutional performance—Projects to finance excellence in RDI, Contract no. 15PFE/2021.

**Conflicts of Interest:** The authors declare no conflict of interest.

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