

# Designing a Highly Efficient Process for Sequential Removal of Heavy Metals in Wastewater Treatment

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The presence of heavy metals in wastewater poses significant threats to human health and the environment, necessitating the development of effective treatment methods. This study focuses on isolating and utilizing heavy metal-resistant bacteria from wastewater to selectively and efficiently remove heavy metals. Sixteen heavy metal-resistant strains, including copper-resistant (CuV1-4), zinc-resistant (ZnX1-4), nickel-resistant (NiX1-4), and cobalt-resistant (CoV1-4) bacteria, were isolated using heavy metal-supplemented culture medium. These bacteria were identified to the genus level through 16S rRNA sequencing and demonstrated varying degrees of resistance. CuV1-4 and CoV1-4 strains tolerated 5 mM Cu<sup>2+</sup> and Co<sup>2+</sup>, respectively, while ZnX1-4 and NiX1-4 tolerated 20 mM of the corresponding heavy metals. These resistant bacteria were tested for their heavy metal removal ability through bioaccumulation and biosorption. While only strain CuV2 can efficiently eliminate Cu<sup>2+</sup> through bioaccumulation, all tested strains showed a strong ability to remove corresponding heavy metals through biosorption. The removal efficiency through biosorption ranged from 32% to 91%, with a biosorption capacity of 0.3-11 mg of heavy metal per gram of dry biomass. This project also revealed that these microbial biosorbents can be recycled and reused for heavy metal removal. Based on these results, a specialized, sequential, and reusable process for heavy metal removal was designed for wastewater treatment. The process was experimentally validated using NiX4 and CuV4 to remove Ni<sup>2+</sup> and Cu<sup>2+</sup>. Therefore, this research presents a cost-effective and sustainable approach for heavy metal removal, which can be easily adopted and applied globally, including in economically disadvantaged areas.