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Extracellular elemental copper bioaccumulation

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Heavy metals are essential for the survival or inhibition of life in extreme environments as they can act as energy sources (electron donors/acceptors) or, in high concentrations, be lethal. Under anaerobic conditions, electron acceptors such as Fe^{3+}, U^{6+}, and As^{5+}, among others, are used as a source of energy by several extremophiles. In contrast, the biological reduction of other metals (e.g., Cu, Zn) has been reported as thermodynamically unfavorable [1]. Although, intracellular reduction of copper to its elemental form under oxic conditions has been reported [2]. Here we describe a novel strategy of extracellular copper reduction to its elemental form in a particular biofilm. A pink biofilm was collected from acid mine drainage with oxidative (pH, Eh, and DO of 2.76, 508 mV, and 16.9 µM, respectively) and microoxic conditions from an abandoned mine pithead. Fe and Al showed the highest concentrations (600 and 108 mg/L, respectively), followed by other trace metals, such as Cu and Zn (14 and 15 mg/L). Analysis by SEM-EDS, XRD, and XPS of the precipitates embedded in the biofilm showed that the precipitates are composed of Cu^0 and bacteria forms. Analysis by µXRF confirmed the presence of Cu^0 and trace minerals such as CuO in the biofilm. High concentrations of quinone<MK-8<MK-7 were detected in the biofilm where Cu^0 was present. Shotgun metagenomics revealed the presence of a complex bacteria community dominated, mainly, by *Rhodanobacter*, *Acidimicrobium*, and *Gallionella*. The functional annotation confirmed the capability of these bacteria to produce several quinones. Chemical experiments demonstrated that synthetic quinone reduces Cu^{2+} ($CuCl_2$) to Cu^0. Since the nature of Cu is to cause multiple damages at the cellular level in bacterial systems, the production of quinones by the consortium could control the mobility of Cu^{2+} in the biofilm, inducing its accumulation of Cu^0 as a neoformed mineral, not toxic for the bacterial consortium.

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