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**METAL - *Pseudomonas extremaustralis* 2E-UNGS INTERACTIONS
FOR THE IMPROVEMENT OF BIOTECHNOLOGICAL APPLICATIONS:
A GENOMIC STUDY**

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During the last 20 years *P. extremaustralis* 2E-UNGS, a native and non-pathogenic bacterium isolated from a polluted environment, was applied in the development of environmental biotechnologies such as metal biotreatments and biosensing. The complete circular 6,372,594 bp chromosome was annotated in NCBI GenBank as NZ_CP091043.1. In previous studies, the maximum tolerance concentrations (MTC) for different metals were determined. The presence of different genes associated with metal tolerance, observed in the development of bioremediation processes, was explored using bioinformatics tools. The aim of this work was to improve the understanding of metal-biomass interactions and their consequences in future bioreactors for metal removal treatments. For that purpose, bioinformatic tools such as Rapid Annotation using Subsystems Technology Server (RAST), the *Pseudomonas* Genome Database, Proksee-Genome Analysis and NIH/NCBI Basic Local Alignment Search Tool (BLAST) or Kyoto Encyclopedia of Genes and Genomes (KEGG) were applied. Specific proteins such as ABC transporters, efflux RND transporters, resistance-specific proteins and two component system response regulators were found. Interestingly, several of these proteins were implicated in multidrug resistance. Performing the traditional Kirby-Bauer disk diffusion susceptibility test revealed that *P. extremaustralis* 2E-UNGS is sensitive to imipenem. A higher sensitivity to imipenem was detected when *P. extremaustralis* 2E-UNGS was previously grown in Cu(II) or Cd(II) containing broth. In contrast, non-growth inhibition was observed in free-metal or in Cu(II), Cd(II) or Zn(II) supplemented Mueller Hinton Agar when this microorganism was precultured in presence of Zn(II). Imipenem resistance was not induced when *P. extremaustralis* 2E-UNGS was precultured in a Cu(II) or Cd(II) supplemented broth. Looking inside the genome, the imipenem resistance may be associated with the negative effects on the expression of *OprD* by the presence of Zn(II) decreasing the imipenem translocation and the expression of MexAB-OprM pump efflux that reduces the antibiotic intracellular concentration. Results on metal-activation of antibiotic resistance were only reported for *P. aeruginosa* and not for other environmentally relevant *Pseudomonas*. In conclusion, the presence of metals can enable or disable resistance mechanisms to other metals or even more induce antibiotic resistance. In this case Zn(II) induces imipenem resistance. This behaviour could

be replicated in indigenous microorganisms of metal contaminated environments, promoting the activation of antibiotic resistance.

Palabras clave: *Pseudomonas extremaustralis* - bioremediation - waste biotreatment - environmental biotechnology