

## **Anaerobic oxidation of metal sulfides in the Iberian Pyrite Belt**

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Until recently, the extreme conditions of pH and high concentration of toxic heavy metals identified in the Río Tinto basin were considered the consequence of over 5000 years of mining activities. However, recent geological, geophysical and hydrological information does not support this assumption. The recharge area of Peña de Hierro aquifer, considered the origin of Río Tinto, was detected northwest of the pit lake at a depth ranging from 100 to 400mbs. Its groundwater moves southwards along the fracture network and when it reaches the massive sulfide bodies of the Iberian Pyrite Belt (IPB) it activates the metabolism of chemolithotrophic microorganisms, generating acidic waters with high concentration of iron and sulfate. To demonstrate this hypothesis two devoted drilling projects to intersect this subsurface bioreactor and obtain information on the oxidation of metal sulfides in anaerobic conditions were carried out. Different complementary methodologies were been used to identify the biodiversity existing in the subsurface of the IPB. Of those, fluorescence in situ hybridization allowed to visualize the distribution of microorganisms in microniches in the solid rock matrix and to detect microorganisms-mineral interactions. Enrichment cultures under controlled conditions strongly suggest the implication of nitrate reducing bacteria, such as *Acidovorax* spp. or *Tessaracoccus lapidicaptus*, in the generation of high concentrations of oxidized iron from pyrite in the subsurface of the IPB. These results confirm the hypothesis that anaerobic microbial metabolisms are responsible of the extreme conditions detected in Río Tinto.