

Bacterial mutualism as new strategy of survive extreme acidic and microaerophilic conditions

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The Iberian Pyrite belt contains some of the most diverse and extreme acidic ecosystems. In order to study the biogeochemical processes supporting the biome under acidic and oxygen limiting conditions, a biofilm was sampled from an abandoned mine pithead. Physico-chemical conditions of the microenvironments ranged from microaerophilic (pH, Eh, and DO of 2.76, 508 mV and 16.9 μ M, respectively) to anaerobic (pH, Eh, and DO of 2.35, 555 mV and 3.8 μ M respectively). Fe and Al showed the highest concentrations (600 and 108 mg/L, respectively) followed by other trace metals, mainly Cu and Zn (14 and 15 mg/L). Analysis by SEM-EDS, XRD, XPS and Py-GC/MS-CSIA revealed that the SO_x species present in the biofilm were originated from oxidative processes ($\delta^{18}\text{O}$ and $\delta^{34}\text{S}$) and it was associated with Cu⁰. The copper interaction is interesting as it acts as cofactor for enzymes such as cytochrome oxidases or superoxide dismutases because of its ability to cycle between Cu²⁺ and Cu⁺. One consideration is the accumulation of Cu⁺²⁺ ions as it contributes to oxidative stress due its participation in Fenton-type reactions. Next generation microbial diversity analysis showed the coexistence of oxido-reductive microorganisms such as *Alicyclobacillus sp.* and *Desulfosporosinus sp.*. This special coexistence and data suggested novel strategies for ROSs (Radical Oxygen Species) and copper homeostasis. Thus, the oxidation processes by *Alicyclobacillus sp.* generates ROSs that oxidize CuS precipitated by *Desulfosporosinus sp.* metabolism. This oxidation releases SO_x and Cu⁰, which accumulate in the biofilm. This process could regulate the ROSs in the system and could allow the accumulation of Cu⁰ as a neoformed mineral phase now possibly not toxic for the biofilm.