Genomic insights into the metabolic flexibility of the acid-tolerant Fe(II)-oxidizer *Sideroxydans* sp. CL21

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Chemolithotrophic, microaerophilic, Fe(II)-oxidizing bacteria (FeOB), including *Sideroxydans* sp., found within the family *Gallionellaceae* are known to oxidize Fe(II) and to fix CO_2 via the RubisCO pathway. To elucidate the metabolic potential of *Sideroxydans* sp. CL21, a gram-negative FeOB isolated from the slightly acidic, minerotrophic Schlöppnerbrunnen fen, we used the PacBio RS II platform to sequence and analyze the genome. The genome has a single chromosome of 3.77 Mb encoding 3795 genes [1]. Genome-resolved characterization of *Sideroxydans* sp. CL21 revealed genes encoding homologs of the Fe(II) oxidation genes *mtoAB* and *cyc2*. We also identified genes encoding an acetate permease and transporter, as well as a lactate permease and dehydrogenase, suggesting strain CL21 can also use organic carbon, thus revealing an atypical mixotrophic metabolism. In addition, the genome contains multiple gene clusters encoding NiFe hydrogenases, homologs of the sulfur oxidation (*sox*) genes, and genes involved in sulfate uptake and assimilation. These findings suggest strain CL21 is capable of using H_2 or sulfur compounds as an alternative electron donor. To validate the metabolic versatility of *Sideroxydans* sp. CL21, we used increasing concentrations of H_2 (0-5%) and $S_2O_3^{2-}$ (0-5mM), with or without FeS, to determine growth and electron donor consumption rates under autotrophic or mixotrophic conditions (0-5mM lactate or acetate). Incubations amended with 1mM lactate, 5mM $S_2O_3^2$, and FeS resulted in higher 16S rRNA gene copies over 17 d, compared to incubations with FeS and 1mM lactate or 1mM acetate. Interestingly, maximal16S rRNA gene copies were highest in incubations amended with both H_2 and FeS plus an organic carbon source. 16S rRNA gene copies in incubations with organic carbon and either H_2 or FeS were similar to each other, albeit slightly lower than in incubations with both donors. Additionally, the rate of H_2 consumption was faster in the absence of Fe(II), suggesting H_2 may be the preferred electron donor when organic carbon is present. Taken together, our combined genome-resolved and ongoing experimental results will help to better assess the impact of *Sideroxydans* sp. CL21-mediated Fe(II) oxidation on the biogeochemical iron cycle in environments where inorganic and organic carbon sources, as well as alternative electron donors, are abundant.

[1] Cooper, *et al*. (2020) *MRA*, **9**:e01444-19.