

## **Novel Beijerinckiaceae expand our knowledge about lanthanide-dependent metabolism**

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Methylotrophic bacteria represent the only known biological sink for methanol and related C<sub>1</sub>-compounds in carbon cycling. Lanthanides were recently identified as important co-factors for methanol dehydrogenases, which are key for methanol oxidation. We isolated Beijerinckiaceae from soft coal slags enriched in mobilizable lanthanides and demonstrated for the first time that these isolates depend on lanthanides for methylotrophy. Furthermore, we showed that methanol oxidation is only possible in the presence of low mass lanthanides (La, Ce, Nd) in nanomolar concentrations (> 10 nM). Incubation experiments and a molecular screening for C<sub>1</sub>-metabolism modules revealed that utilizable C<sub>1</sub>-substrates include methylamines and methylated sulfur species as well. An overall broad carbon utilization range including various sugars and organic acids suggested a metabolic diversity expanding beyond C<sub>1</sub>-metabolism. A preliminary genomic characterization revealed multiple specifics relating to C<sub>1</sub>-metabolism: a glutathione-dependent oxidation pathway compensates for the lack of tetrahydromethanopterin-mediated formaldehyde oxidation and carbon for biomass assimilation is channelled into the serine cycle using a tetrahydrofolate-centred pathway involving FolD. While methanol oxidation is linked to the activity of a lanthanide-dependent XoxF methanol dehydrogenase, we found that the respective gene is fragmented in one isolated soft coal slag methylotroph - a novelty among methylotrophs. We assume that XoxF activity might be complemented by one additional, lanthanide-dependent alcohol dehydrogenase, ExaF, either at the level of methanol or formaldehyde oxidation. ExaF is one of the first lanthanide-dependent enzymes not tied to methanol oxidation and thus far only known from distinctly related Methylobacteriaceae, but not Beijerinckiaceae. Different from common methylotrophic model organisms, we believe that the metabolic diversity of these novel strains provides a platform to study lanthanide-dependent metabolism in a physiological setting reaching beyond methylotrophy.