brief communications

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Phosphite oxidation by sulphate reduction

Bacterial metabolism

iological phosphorus occurs almost exclusively as phosphate in the redox state of + V, although a few phosphonic (+III) and phosphinic (+I) acids are found as secondary metabolites1 or as constituents of phosphonolipids. Here we show that a culture of a lithoautotrophic bacterium purified from marine sediments in Venice can grow by anaerobic oxidation of phosphite (+III) to phosphate (+V) while simultaneously reducing sulphate to hydrogen sulphide. To our knowledge, this is the first description of a redox reaction involving phosphorus in microbial energy metabolism, an activity that might have operated on the early Earth and which could represent an ancient evolutionary trait.

The reduction of phosphate to phosphine (PH₃) has to proceed through steps of extremely low redox potential² (HPO₄²⁻/HPO₃²⁻, -690 mV; HPO₃²⁻/H₂PO₂⁻, -913 mV; H₂PO₂⁻/P, -922 mV; P/PH₃, -525 mV, at pH 7.0), so it is unlikely to drive the coupling of a respiratory process to biomass oxidation^{3,4}. Traces of phosphine have been detected in sediments⁵, paddy fields⁶ and manure samples⁴, but their origin is unclear.

Phosphite oxidation to phosphate should provide a good electron source for microbial energy metabolism, however, as the electrons are released at a very low redox potential. Phosphite and hypophosphite can serve as a phosphorus source for aerobic^{7,8} and anaerobic bacteria⁹, but so far there has been no evidence for quantitative phosphite oxidation as a type of energy metabolism.

We enriched phosphite-oxidizing, anaerobic bacteria in a sulphide-reduced mineral medium¹⁰ with sulphate as the elec-



Figure 1 Phylogenetic tree based on gene sequences encoding the small subunit of ribosomal RNA (16S rRNA), showing the relation of the phosphite-oxidizing, sulphate-reducing strain FiPS-3 to members of the genus *Desulfobacter, Desulfobacula, Desulfospira,* and other representatives of major lineages of the δ -subclass of Proteobacteria. Scale bar represents 10% estimated difference in nucleotide sequences. DNA from strain FiPS-3 was extracted and purified using standard protocols and the rDNA was sequenced and analysed by using the ARB software package (TU Munich). The topology of the phylogenetic tree was derived from distance-matrix analyses and constructed using a neighbour-joining algorithm. *Escherichia coli* and *Holophaga foetida* were used as outgroup references.

tron acceptor, by using marine or freshwater sediments as inoculum. Growing cultures developed within 2–4 months and were transferred repeatedly into subcultures. Control untreated cultures, or cultures containing autoclaved cells, showed no evidence of phosphite oxidation. We isolated a pure culture of a strictly anaerobic, phosphite-oxidizing sulphate-reducing bacterium, strain FiPS-3, which grew with phosphite plus sulphate and a doubling time of three days according to

 $4\text{HPO}_3^{-} + \text{SO}_4^{-} + \text{H}^+ \rightarrow 4\text{HPO}_4^{-} + \text{HS}^$ where Δ*G*[°] = − 364 kJ per mol sulphate, or −91 kJ per mol phosphite.

The strain also oxidized fumarate and malate to CO_2 , and slowly oxidized hydrogen and formate. Lithotrophic growth with phosphite or hydrogen was not enhanced by addition of acetate, indicating that this strain covers all its carbon assimilation through autotrophic CO_2 fixation.

Analysis of 16S ribosomal RNA gene sequences revealed that strain FiPS-3 shares strong sequence similarities with sulphate-reducing Proteobacteria of the δ -subclass and is most closely related to an as-yet undescribed benzene-mineralizing clone SB-9 and to *Desulfospira joergensenii, 'De-sulfobacterium phenolicum'* and *Desulfobacula toluolica* (Fig. 1).

The ecological contribution of dissimilatory phosphite oxidation is unclear; on today's oxygen-exposed Earth, phosphite and other reduced inorganic phosphorus compounds are unstable. Phosphite may be an intermediate in the microbial degrada-

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tion of organophosphonates in anoxic habitats, but there is no evidence for this. In prebiotic and archaean times, reduced phosphorus compounds might have been important as precursors of biochemical phosphorus compounds¹¹, for example, or they may have been introduced by meteorites¹². Anaerobically respiring microorganisms like the sulphate-reducing bacterium described here could have thrived on a litho(auto)trophic metabolism involving the oxidation of reduced phosphorus compounds, in which case dissimilatory phosphite oxidation by sulphate-reducing bacteria may represent an ancient evolutionary trait.

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