A universal immuno-probe for (per)chlorate-reducing bacteria

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Recent studies in our lab have demonstrated the ubiquity, diversity and metabolic versatility of microorganisms which couple growth to the anaerobic reduction of chlorate or perchlorate ((per)chlorate). We identified two taxonomic groups, the Dechloromonas and the Dechlorosoma groups, which represent the dominant (per)chlorate-reducing bacteria (ClRB) in the environment. As part of these studies we demonstrated that chlorite dismutation is a key step in the reductive pathway of (per)chlorate that is common to all ClRB which is mediated by the enzyme, chlorite dismutase (CD). Initial studies on CD suggested that this enzyme is highly conserved amongst the ClRB, regardless of their phylogenetic affiliation. As such this enzyme makes an ideal target for a probe specific for these organisms. Polyclonal antibodies were commercially raised against the purified CD from the ClRB Dechloromonas agitata strain CKB. The obtained antisera was deproteinated by ammonium sulfate precipitation and the antigen binding activity was assessed using dot-blot analysis of a serial dilution of the antisera. The titer values obtained with purified CD indicated that the antisera had a high affinity for the CD enzyme and activity was observed in dilutions as low as $1 \times 10^{-6}$ of the original antisera. The antisera was active against both cell lysates and whole cells of D. agitata, but only if the cells were grown anaerobically with (per)chlorate. No response was obtained with aerobically grown cultures. In addition to D. agitata, dot-blot analysis employed with both whole cell suspensions and cell lysates of several diverse ClRB representing the alpha, beta, and gamma subclasses of the Proteobacteria tested positive regardless of their phylogenetic affiliation. Interestingly, the dot-blot response obtained for each of the ClRB cell lysates was different suggesting that there may be some differences in the antigenic sites of the CD protein produced in these organisms. In general, no reactions were observed with cells or cell lysates of the closely related organisms to the ClRB which could not grow by (per)chlorate reduction.

These studies have resulted in the development of a highly specific and sensitive immuno-probe based on the commonality of the chlorite dismutase enzyme in ClRB which can be used to assess dissimilatory (per)chlorate-reducing populations in environmental samples regardless of their phylogenetic affiliations.

High-resolution sulphur isotopic analysis, using laser ablation techniques, on pyritised fossils from the Hunsrück Slate, Western Germany.

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First detailed study of pyritised fossils by laser ablation techniques

High-resolution laser sulphur isotope techniques have been used to produce three-dimensional isotopic maps of in situ pyritised fossils from the Lower Devonian Hunsrückschiefere (Western Germany). Previous sulphur isotope studies of pyritised fossils have generally relied upon bulk rock analysis (Briggs et al. 1996), and the separations of different morphologies (e.g. Underwood & Bottrell 1994). Recent advances in laser technology allow more accurate and precise methods of sampling, but until now there has been no detailed study of pyritised fossils by laser ablation techniques.

Discussion

Three-dimensional micro-scale variations of sulphur isotope compositions have been found within a variety of organisms. Micro-scale (<1mm$^3$) sulphur isotopic data from a gastropod, trilobite, orthocones and furcaster, are all isotopically heavy (>0‰) and show consistent surface and depth trends that reach maximum values of +51.6‰.

Conclusions

High-resolution sulphur isotopic data can be used to identify the timing of pyritisation within different parts of the same organism. Spatial trends indicate that the process of pyritisation began on the surfaces of the skeletal tissue, replacing them inwards in the main body, before moving towards the organisms’ extremities.

References