

expression of *phsABC*, *E. coli* DH5 α harboring the *phsABC* constructs showed higher thiosulfate reductase activity and produced significantly more sulfide than the control strain (*E. coli* DH5 α) under both aerobic and anaerobic conditions. Among the four constructs, *E. coli* DH5 α harboring pSB74 produced the highest level of thiosulfate reductase and removed most of heavy metals from solution under anaerobic conditions. In a mixture of 100 μ M each of cadmium, lead, and zinc, the strain could remove 99% of the total metals from solution within 10 hours. Cadmium was removed first, lead second, and zinc last. In contrast, a negative control did not produce any measurable sulfide and removed very little metals from solution. These results have important implications for removal of metals from wastewater contaminated with several metals.

SL317

Carbon Tetrachloride Dechlorination and Metal Chelating Properties of Pyridine-2,6-bis (thiocarboxylic acid) Produced from *Pseudomonas stutzeri* Strain KC

Chang-Ho Lee¹, A. Paszczynski² and Ronald L. Crawford²

¹Environmental Bioresources Lab., KRIBB, Yusong, Taejeon 305-333; ²Environmental Biotechnology Institute, University of Idaho, Moscow, Idaho 83844, U.S.A.

Pseudomonas stutzeri strain KC was originally characterized as having, under iron-limiting conditions, novel carbon tetrachloride (CCl₄) dehalogenation activity, specifically, a net conversion of CCl₄ to CO₂. The exact pathway and reaction mechanisms are unknown, but chloroform is not an intermediate and thiophosgene and phosgene have been identified as intermediates in trapping

experiments. Previous work by others using cell-free preparations has shown that cell-free culture supernatants that have been passed through a low-molecular-weight cutoff membrane can confer rapid CCl₄ transformation ability upon cultures of bacteria which otherwise show little or no reactivity toward CCl₄. We used a cell-free assay system to monitor the complete purification of compounds showing CCl₄ degradation activity elaborated by iron-limited cultures of strain KC. Electrospray tandem mass spectroscopy, NMR spectroscopy, and comparisons with synthetic material have identified pyridine-2,6-bis (thiocarboxylate) (PDTC) as a metabolite of strain KC which has CCl₄ transformation activity in the presence of chemical reductants, e.g., titanium[III] citrate or dithiothreitol, or actively growing bacterial cultures. We have tested the ability of complexes of synthetic PDTC and several transition metal ions, as well as the uncomplexed ligand, to dechlorinate CCl₄ in assays, which include either bacterial cells, chemical reducing agents, or the complex alone with CCl₄. Of the Fe, Ni, Co, and Cu complexes, only the Cu:PDTC complex showed activity under all conditions tested. The reaction between Cu:PDTC and CCl₄ took place both aerobically and anaerobically. Identification of end products and intermediates of the reaction suggested a reaction pathway that includes trichloromethyl radical coupling to one of the sulfur atoms of PDTC, and hydrolysis of the resulting thioester.

SL318

Developmental Pattern Formation Controlled by *patS* in a Cyanobacterium

Ho-Sung Yoon and James W. Golden¹

Dept. of Organismic and Evolutionary Biology,
Harvard University, MA 02138-2094, U.S.A;
¹Dept. of Biology, Texas A&M University,
College Station, Texas 77843-3258, U.S.A.

Many filamentous cyanobacteria grow as multicellular organisms that show a distinct one-dimensional developmental pattern of single nitrogen-fixing heterocysts separated by approximately ten vegetative cells. Overexpression of a 54-base-pair gene, *patS*, blocked heterocyst differentiation in *Anabaena* sp. strain PCC

7120. A *patS* null mutant showed an increased frequency of heterocysts and an abnormal pattern. Expression of a *patS::gfp* reporter was localized in developing proheterocysts. Significantly, exogenously added synthetic peptide corresponding to the last five COOH-terminal residues of PatS inhibited heterocyst development. PatS appears to control heterocyst pattern formation cell nonautonomously by serving as a diffusible inhibitor of differentiation.