

Aerobic chromate reduction by *Thermus scotoductus* strain SA-01

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The contamination of the environment with hexavalent chromium [Cr(VI)] has become a major concern. *Thermus scotoductus* strain SA-01, a thermophile previously isolated from groundwater from a South African gold mine at a depth of 3.2 km, has shown the ability to reduce Cr(VI) under non-growth conditions using lactate as an electron donor (1). Batch growth experiments as well as cell suspensions under non-growth conditions were used to assess its ability to reduce Cr(VI) under aerobic conditions. The bacteria were able to reduce Cr(VI) when grown in a complex organic medium containing Cr(VI) concentrations up to 0.5 mM. Suspension of *T. scotoductus* SA-01 cells also reduced Cr(VI) under non-growth conditions using a variety of electron donors. The optimum temperature and pH for Cr(VI) reduction under non-growth conditions were found to be 80°C and pH7, respectively. It was also found that the Cr(VI) reduction was catalyzed by a soluble cytoplasmic, constitutively expressed enzyme. The soluble aerobic chromate reductase was purified using anion-exchange chromatography, hydrophobic interaction chromatography, dye affinity chromatography and size-exclusion chromatography, showing a monomer molecular weight of 36 kDa on SDS-PAGE analysis. The optimum of the reaction is at pH 7.5 and 65°C. The chromate reductase is extremely thermostable with more than 60% of the activity remaining after 8h exposure to 80°C. The enzyme showed hyperbolic dependence on the concentration of both chromate and NADH, catalyzing the reduction of 1 mol Cr(VI) while consuming 2 mol NADH as electron donor.

¹ Kieft, T.L., Fredrickson, J.K., Onstott, T.C., Gorby, Y.A., Konstandarithes, H.M., Bailey, T.J., Kennedy, D.W., Plymale, A.E., Spadoni, C.M. and Gray, M.S. (1999) Dissimilatory reduction of Fe(III) and other electron acceptors by a *Thermus* isolate. Appl. Environ. Microbiol 65, 1214-1221.