Characterizing the Catalytic Potential of *Deinococcus, Arthrobacter*, and Other Robust Bacteria in Contaminated Subsurface Environments of the Hanford Site

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We have recently isolated several distinct species of *Deinococcus* and *Arthrobacter* from pristine and contaminated soils of DOE's Hanford Site in south-central Washington state. Generally, *Deinococcus* bacteria are exceptionally robust, not only surviving exposure to radiation, but also to other DNA-damaging conditions typical of DOE environments. The ability to reduce a variety of toxic metals also appears to be a characteristic shared by these organisms, and we have shown that *D. radiodurans* and *D. geothermalis* are proficient at reducing U(VI), Tc(VII) (in the presence of an electron shuttle), and Cr(VI), and have engineered both species for Hg(II) reduction. Recent experimental advances in the genetic management of *D. radiodurans* and *D. geothermalis* will facilitate our efforts to characterize the metal-reducing mechanisms of both organisms. These advances include our comprehensive analysis of the *D. radiodurans* genome, and for *D. geothermalis*, the development of a system for genetic transformation and ongoing genome sequencing, annotation, and analysis of this organism. *Arthrobacter* species are capable of free-living growth in many extreme environments, such as under chronic radiation. *Arthrobacter* spp. also have recently been reported to efficiently resist and reduce high concentrations of hexavalent chromium. These and other cultured isolates from the Hanford Site will be examined for their metal-reducing capabilities. Furthermore, we have shown that the ionizing radiation doses that yield 17% survival of *Escherichia coli* and *D. radiodurans* are higher by factors of 20 and 200, respectively, than those for *Shewanella oneidensis* MR-1, a dissimilatory metal-reducing gamma-proteobacterium that has been proposed for bioremediation of DOE waste sites. However, *S. oneidensis* is extremely sensitive to ionizing radiation, desiccation, and other oxidative stress conditions. Recent advances in understanding how *D. radiodurans* survives ionizing radiation support the possibility that the resistance characteristics of sensitive organisms such as *S. oneidensis* might be increased significantly with antioxidants, which could benefit applications in environmental biotechnology. We are examining if antioxidants can increase the efficacy of *S. oneidensis* in genotoxic environments, without the need for genetic engineering. The specific aims of this project are: (1) characterizing the genetic basis of metal reduction in *D. radiodurans* and *D. geothermalis*, and novel deinococcal isolates from the Hanford Site; (2) characterizing the microbial ecology of contaminated waste sites and the metal-reducing potential of these communities; (3) defining environmental factors that govern in situ growth, environmental robustness, metabolism, and Cr(VI)-reduction of *Deinococcus* and *Arthrobacter* spp.; (4) characterizing the genetic basis of metal reduction in *Arthrobacter aurescens* (TC1) and novel *Arthrobacter* isolates from the Hanford Site; and (5) testing the ability of antioxidants to support the growth of *S. oneidensis* under chronic ionizing radiation.