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Examination of the Nickel Site Structure in *Streptomyces seoulensis* Superoxide Dismutase by EPR and X-ray Absorption Spectroscopy

Jin-Won Lee*, Yang-In Yim, Michael J. Maroney[1] and Sa-Ouk Kang
Laboratory of Biophysics, Department of Microbiology, College of Natural Sciences, and Research Center for Molecular Microbiology, Seoul National University, [1]Department of Chemistry and the Program in Molecular and Cellular Biology, University of Massachusetts

Superoxide dismutases are metalloenzymes catalyzing the dimutation of superoxide anion radical to hydrogen peroxide and molecular oxygen. Examples of enzymes containing Cu, Mn and Fe as the redox-active metal have been characterized. Recently, an SOD containing one Ni atom per subunit was reported. The structural gene *sodN* from *Streptomyces seoulensis* was composed of 131 amino acids and post-translationally modified to 117 amino acids subunit which had a molecular mass of 13.2 kDa. For the confirmation of active site nickel and nitrogen, NiSODs enriched in ^{61}Ni or ^{15}N were studied by EPR spectroscopy. The nitrogen isotope (N^{15}), which has a nuclear spin of 1/2, splitted the original g_z line at g=2.012 into 2 lines. And the nickel isotope (N^{61}), which has a nuclear spin of 3/2, splitted the g_z line into 6 lines. And the X-ray absorption spectroscopic studies showed that the Ni in the oxidized enzyme is in a five-coordinate site composed of three S-donor ligands, one N-donor and one other O- or N-donor. This unique coordination environment is modified by loss of one N- (or O-) donor ligand in the reduced enzyme. These results characterize another class of metal center active in catalyzing the redox chemistry of superoxide dismutase.