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Article Title: Mechanism of methane monooxygenase enzymatic chemical reactions

Article Author: Liu, A. M.; Li, S. B

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ABSTRACTS OF PAPERS

Part 1

205th ACS National Meeting

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American Chemical Society

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452. KINETIC AND STRUCTURAL CHARACTERIZATION OF CARBONIC ANHYDRASES OF PLANT ORIGIN. Roger S. Rowlett, Jill R. Royal, Rajib Saha, Melanie Woodroffe, Michael G. Lam, Department of Chemistry, Colgate University, Hamilton NY 13346; Mark R. Chance and Michael D. Wirt, Department of Physiology and Biophysics, Albert Einstein College of Medicine, Bronx, NY 10461.

Carbonic anhydrase has been purified to homogeneity from leaves of *Spinacea oleracea* (spinach) and *Zea mays* (corn). Like carbonic anhydrases of animal origin, these enzymes have subunit molecular weights of approximately 30 kDa, and contain a catalytically essential Zn^{2+} ion. Kinetic studies of the spinach enzyme using stopped-flow spectrophotometry and ^{13}C-NMR exchange are consistent with a zinc-hydroxide mechanism of action with a rate-determining intramolecular H^+ transfer, similar to that of animal carbonic anhydrases. But the structural organization of carbonic anhydrases is very different from known animal varieties. The spinach holoenzyme appears to be an octamer, and the corn holoenzyme a dimer--all animal carbonic anhydrases are monomeric. Zn-EXAFS studies of the spinach enzyme indicate the presence of one or more sulfur ligands to the catalytically essential Zn^{2+}--the analogous animal enzymes have only nitrogen-based ligands. Plant carbonic anhydrases are functionally equivalent to, but structurally different from, animal carbonic anhydrases. Thus, plant and animal carbonic anhydrases appear to be convergently evolved enzymes.

453. X-RAY ABSORPTION SPECTROSCOPY OF FERRIC SUPEROXIDE DISMUTASE. David L. Tierney, Anita L. Metzger, Martha L. Ludwig and James E. Penner-Hahn, Departments of Chemistry and Biophysics, University of Michigan, Ann Arbor, MI 48109

Superoxide dismutase (SOD) catalyzes the disproportionation of superoxide to hydrogen peroxide and dioxygen. The structure of Fe(III) SOD from *E. Coli* has been determined crystallographically. The crystal structure shows the Fe(III) site to have trigonal bipyramidal geometry (an equatorial plane of two histidines and an unusually close [1.76 Å] aspartate; axially coordinated solvent and histidine). Azide, an inhibitor of SOD, is believed to bind analogously to superoxide. The structure of the azide derivative has been previously determined by Ludwig et. al., and shows no loss of protein ligands upon binding of azide. Two different structural schemes of the azide complex have been advanced - one in which the azide displaces water, and a second where iron coordination increases from five to six. Extended X-ray absorption fine structure (EXAFS) has been employed to distinguish between these two schemes. The increase in average bond length upon addition of azide and the decreased intensity of the 1s-3d transition are consistent with Fe(III) going from five to six coordinate. The average Fe-ligand bond lengths support the five coordinate crystallographic model. However, at the current resolution, the EXAFS data provide no evidence regarding the short iron-aspartate bond.

454. METHANE MONOOXYGENASE STRUCTURE AND MECHANISM — WHERE TO NOW? Ai-Min LIU†, Shu-Ben LI‡, Hui-Lin WAN†, and Khi-Rui TSAI†, † Department of Chemistry, Xiamen University, Xiamen 361005; ‡ Lanzhou Institute of Chemical Physics, Chinese Academy of Sciences, Lanzhou, 730000, P. R. China.

The previous investigations of methane monooxygenase systems have revealed many aspects of the structure and function, however, MMO is still natural substance known to catalyze the oxidation of hydrocarbons through the insertion of one of oxygen atom of O_2 at atmospheric pressure and room temperature in a single step. The prospects and problems of the isolation, purification, structural characterization, phsico-chemical properties, and biosynesis of MMO will be viewed with emphasis on the structural organization and mechanistic implication for the active site and the cluster environment, especially the evolution of the inconsistent results from various labs including the difference of soluble and particulate MMO systems in nature among methanotrophs, the existence or non-existence of μ-oxo(or hydroxo)-bridge-Fe center, etc. By regarding all the known exogenous substrates and inhibitors of MMO as chemical probes and based on coordination approach, a chemical model of the enzyme system will be proposed and some ideas following the example set by studies of the mechanistic aspects of the nitrogenase will be suggested.