

E0078

Time-resolved Crystallographic Studies of the Heme-based Sensor FixLH. Jason Key¹, Vukica Srajer^{1,2}, Reinhard Pahl², Keith Moffat^{1,2}, ¹Dept. of Biochemistry and Molecular Biology, ²Consortium for Advanced Radiation Sources, Univ. of Chicago, Chicago IL USA.

The protein FixL, a heme-based oxygen sensor found in nitrogen-fixing *Rhizobia*, is responsible for the regulation of nitrogen fixation genes in response to molecular oxygen. It is a modular protein composed of two domains: an N-terminal PAS domain which contains covalently bound heme (FixLH), and a C-terminal histidine kinase domain. The N-terminal sensor domain regulates the enzymatic activity of the kinase in response to heme-bound ligand. FixLH is a member the emerging family of heme-based PAS domain gas sensor proteins, distinct from globins in structure and function. In order to investigate the early structural events of signal transduction in FixLH, we are conducting time-resolved crystallographic experiments on the photolabile CO complex of FixLH from the organism *Bradyrhizobium japonicum* using time-resolved Laue diffraction. Static structures of the protein have been determined at room temperature to investigate structural changes upon CO binding as well as to provide phase information for room temperature time-resolved studies. These structures reveal a shift of the residues distal to the heme in the H β and I β strands of the protein upon CO binding. Time-resolved experiments and subsequent analysis of time-resolved difference Fourier maps by singular value decomposition (SVD) show the structural relaxation of CO induced change in the protein to the deoxy conformation following CO photolysis.