Molecular Mechanism of Functional Regulation for the Heme-Based Sensor Proteins

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Hemeproteins show a wide variety of functions including storage and transport of molecular oxygen, electron transfer, and redox catalysis of various substrates. Besides these traditional functions, a new function of hemeproteins has been found recently, in which the heme acts as a sensor of diatomic gas molecules such as oxygen, nitric oxide, and carbon monoxide. In this work, the structure and function relationships of an oxygen sensor protein (HemAT-Bs), a CO-sensor protein (CooA), and a redox sensor (DcrA) are discussed.

HemAT-Bs is a signal transducer protein responsible for aerotaxis of *B. subtilis*. The recombinant HemAT-Bs shows similar spectroscopic properties to Mb. However, HemAT-Bs shows a unique resonance Raman spectrum in the O_2-bound form suggesting a unique hydrogen bonding network between the heme-bound oxygen and distal amino acid residues in the distal heme pocket. Thr95 is involved in the hydrogen bonding to the heme-bound oxygen. Resonance Raman spectroscopy reveals that there are three different conformations in the O_2-bound form of HemAT-Bs. I will discuss spectroscopic properties of wild type and some mutants including T95 A HemAT-Bs.

CooA is a CO sensing transcriptional activator, which contains a b-type heme as the active site for sensing its physiological effector, CO. CooA from *Rhodospirillum rubrum* (Rr-CooA) shows some unique properties for the coordination structure of the heme. However, it is not clear if these unique properties are essential for CooA function. To determine the essential elements for CooA function, we have characterized a CooA homologue from *Carboxydothermus hydrogenoformans* (Ch-CooA) by spectroscopic and mutagenesis studies in this work. Comparing the properties of Ch-CooA and Rr-CooA provides that the essential elements for CooA function will be that (i) the heme is six-coordinate in the Fe(III), Fe(II), and Fe(II)-CO forms; (ii) the N-terminus is coordinated to the heme as an axial ligand; and (iii) CO replaces the N-terminus bound to the heme upon CO binding.

DcrA is a putative methyl accepting chemotaxis protein from a sulfate reducing bacterium *Desulfovibrio vulgaris*. The periplasmic domain of DcrA (DcrA-N) contains a c-type heme that shows a redox-dependent ligand exchange between a water and an endogenous amino acid. The redox potential of DcrA-N (-250 mV vs. NHE) is very low compared with that of typical cytochrome c. I will discuss the physiological function of DcrA deduced from spectroscopic and mutagenesis studies.