and also on the copper(II) ion in relation to the chromophore.

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EXAFS and X-ray Crystallographic Studies of Hemerythrin

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Hemerythrin is a respiratory protein in several phyla of marine invertebrates. The active site of the protein contains a binuclear iron center which cycles from the ferrous to the ferric state upon oxygen binding, with a concomitant reduction of oxygen to peroxide [1]. The structure of the binuclear iron center in the oxidized, metazide form of hemerythrin has been revealed by X-ray crystallography [2]. In the 2.2 Å resolution structure the two iron atoms are coordinated to 2 and 3 protein histidine residues, respectively. In addition, the irons are bridged by 2 protein carboxylates and an oxo group derived from solvent. The remaining octahedral coordination site on one of the iron atoms is occupied by the exogenous ligand, azide.

The present of a μ-oxo-bridged binuclear iron center in methemerythrins was predicted on the basis of spectroscopic and magnetic behavior [1]. Additional evidence has been obtained from the EXAFS above the iron K edge [3]. For metazidohemerythrin the first coordination shell data is well fit by 5 nitrogens plus oxygens at an average distance of 2.15 ± 0.05 Å and an additional oxygen at 1.80 ± 0.05 Å. The short Fe–O bond is consistent with observed Fe–O(μ-oxo) distances in model compounds and also with the X-ray crystallographic data for metazidohemerythrin.

Oxyhemerythrin shows many similarities to methemerythrins, particularly in the strong antiferromagnetic coupling of the iron atoms comprising the binuclear iron center [1]. A comparison of the EXAFS data for oxyhemerythrin and metazidohemerythrin [3] reveals even closer similarity

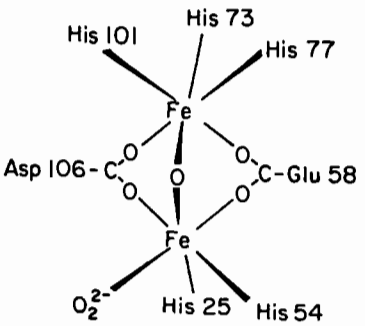

Fig. 1. Proposed structure of the binuclear iron center in oxyhemerythrin based on EXAFS and X-ray crystallographic results.

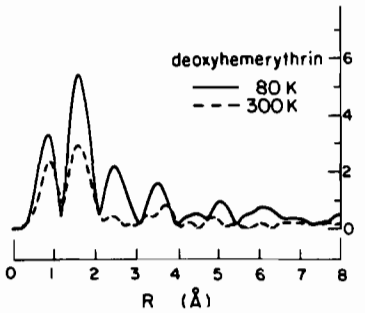

Fig. 2. Fourier transforms of the EXAFS above the iron K edge. Fe–Fe peak is at ~2.5 Å.

between the two forms than had been expected. They appear to have the same number and types of iron ligands, the same iron–ligand distances, and similar iron–iron distances. The proposed active site structure for oxyhemerythrin is analogous to that of metazidohemerythrin [2], but with peroxide in place of azide at the exogenous ligand site (Fig. 1).

Deoxyhemerythrin is distinguished by its loss of antiferromagnetic coupling between the iron atoms [1]. The decreased iron–iron interaction in deoxyhemerythrin is also apparent from the absence of the iron–iron peak in the Fourier transforms of the EXAFS data obtained at 300 K. The iron–iron peak can be observed by lowering the temperature to 80 °K (Fig. 2). The increased relative thermal motion of the two iron atoms in deoxyhemerythrin is apparently due to the absence of a μ-oxo bridge in this form of the protein.

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