High-resolution Emission Study of the Binding Mode of Aminoacids in Cu Bioinorganic Enzyme Models of the 2-his-1-carboxylate Triad

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Galactose oxidase(GO): the Cu metal site catalyzes the oxidation of primary alcohols with the reduction of dioxygen into hydrogen peroxide.

In developing bio-inspired catalysis, Cu-His in aqua solutions have been studied with a multi-technique approach. The results suggest the coexistence of several species in different proportions for different pH values [1]. The imidazole binds at low pH in a monodentate manner to the metal, from the carboxylate end, and with increasing pH the coordination turns to bidentate. This is rather different from the normal binding mode of Cu-His in proteins (by any of the nitrogens of the imidazole ring). In another study, Cu-His was bound to a zeolite, and the species appearing for different pH values have been studied [2]. The modeled compounds are proposed to have a tridentate histidine coordination, at difference of the bidentate coordination proposed for Cu–His in solution [1]. In recent GO model compounds [3], using pro-phenoxyl ligands, the deprotonation of the axial phenol forces the metal to move out of the square plane towards the oxygen. The order of the deprotonation during titration (benzimidazolium ligand/phenol ligand vs. coordinated water) is important for the understanding of the fine tune between protonation and pKa and ultimately, the determinants of the mechanism of GO.

X-ray emission data on four biomimics of GO were collected using synchrotron radiation at NSRRC SP12U1 beamline. The $K\beta_{2,5}$ emission peak as using to monitor the radiation damage. Cryogenic temperatures as well as short exposure avoided any damage on the samples.

The crossover peak Kβ" at the lower energy side of the $K\beta_{2,5}$ emission peak has been assigned to the ligand 2s band energy and is caused by transitions from the metal 4p-character in this band to the metal 1s. This implies that the strength of the metal 4p-ligand 2s hybridization is directly visible in the crossover peak. It turns out that the crossover peak is sensitive to the difference in protonation of a solvent ligand, even in the presence of coordinated nitrogen (from imidazoles), as shown in carbonic anhydrase (CA) and in the CA bioinorganic model compounds [4]. Also the crossover peak is sensitive to the difference between N and O bound to a metal [5].

The experimental results show differences in the crossover peak for the two samples analyzed. Due to the low count rate achievable in molecular systems, duplicates of the samples were measured in 2009. The result of 2007 was successfully repeated. XANES simulations with FEFF8 agree with the experiments.

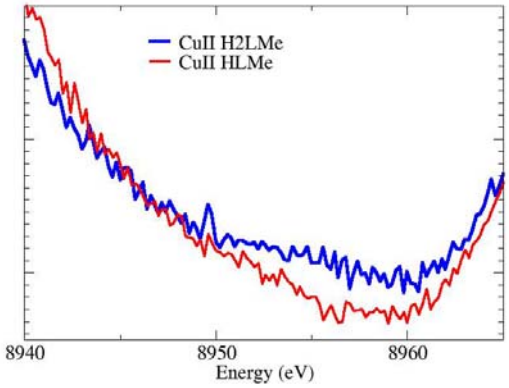

Fig. 1: (left). Crossover peak for the HLMe (Red) and H2LMe (Blue) compounds (experiment 2009).

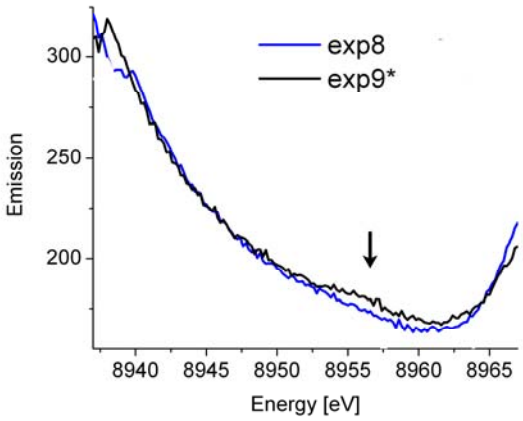

Fig. 2: (right). Crossover peak for the HLMe (Blue) and H2LMe (Black) compounds (experiment 2007).

References

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