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Modulation of Axial Methionine Coordination in Type-1 Copper Sites. Iain MacPherson, Michael E. P. Murphy, Biochemistry and Molecular Biology, Univ. of British Columbia, Vancouver, BC.

Type-1 copper sites are characterized by their strong absorbance at 600 nm and sometimes 460 nm, giving rise to an intense blue-to-green color. A typical type-1 copper site contains three strong ligands, two histidines and one cysteine, and one weaker axial methionine. Whether the four-coordinate copper site lies in the blue or green end of the spectrum (ratio of A_{600}/A_{460}) depends largely on the geometry of the axial methionine with respect to the other three ligands and the copper, affecting the ratio of A_{600}/A_{460}. The type-1 copper protein, nitrite reductase from the soil bacteria Alcaligenes faecalis (AfNiR), is characterized by its intense absorbance at 589 and 460 nm, giving rise to a strong green color. Random mutagenesis and screening resulted in two variants of nitrite reductase, H60Y and H60R. Their electronic spectra differ significantly from the native enzyme with A_{600}/A_{460} ratios of 0.85 and 1.18, respectively, compared to 0.81 from native AfNiR, leading to a marked blue shift in color. X-ray structural analysis of the H60Y and H60R variants show a change in the β-strand containing non-coordinating residues Met62 and Phe64, whose side chains flank the axial Met150. The changes observed in the structures of H60Y and H60R are discussed and related to the close homologue, nitrite reductase from Alcaligenes xylosoxidans, which differs from AfNiR with its axial methionine coordination and intense blue color.