A Theoretical Study on the T1Cu Site in Bilirubin Oxidase (BOD): Relation between Structural Changes and Redox Potentials

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Bilirubin oxidase (BOD) catalyzes the chemical reaction (2 Bilirubin + O_2 → 2 Biliverdin + $2H_2O$) [1]. The wild type (WT) BOD possesses a characteristic cross-link between Trp396 and His398 near the T1Cu atom. A M467Q mutant of BOD lacks the cross-link [2], and the catalytic activity of the M467Q becomes quite low (~0.3%) compared to the WT [3]. In this study, the role of the T1Cu site was studied to elucidate a relation between the structural changes and the redox potentials around the T1Cu atom (Cu(II) and Cu(I) states) by means of the QM/MM method using the NWChem program [4].

QM/MM results well reproduced the X-ray crystal structures, where the root-mean-square deviation (RMSD) values for heavy atoms were 0.082-0.094 Å for WT and 0.148-0.181 Å for M467Q, respectively. In particular, the covalent C-N bond length between Trp396 and His398 in WT BOD obtained by the QM/MM calculation was the same as the X-ray structure (R = 1.40 Å), and the corresponding interatomic distance in the M467Q BOD (R_{calc} = 2.88 Å) was also consistent with the X-ray structure (R_{exp} = 3.08 Å) [2]. The calculated redox potentials, ΔG (= $G_{Cu(II)}$ - $G_{Cu(I)}$), were 3.85 eV and 3.26 eV for WT and M467Q, respectively. Therefore, the change in the redox potentials ($\Delta\Delta G = \Delta G_{M467Q} - \Delta G_{WT}$) was the negative value [5], which is consistent with the tendency of the observed redox potential decrease in the M467Q BOD [3].

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