Abstract

Copper ions are, along with iron and zinc, one of the most commonly occurring metal ions in biological systems. Their electronic structure - especially when it comes to (coupled) polynuclear Cu centers in metalloproteins - makes them one of the most challenging systems in contemporary bioinorganic (and theoretical) chemistry.

On the other hand, the presence of copper ions leads to unique spectroscopic properties. Several spectroscopic "fingerprints" exist which are used to characterize the individual, usually short-lived intermediates in the catalytic cycles of copper metalloenzymes. By correlating experimental and theoretical data, the reaction mechanism of the copper metalloenzymes can be revealed and ultimately fully understood at the atomic and electronic levels. This not only allows us to understand the physicochemical principles underlying fundamental biological processes, but also opens up possibilities to construct artificial (biomimetic) systems that carry the same or even better function as the original biological system.

The aim of this dissertation is to characterize and understand the reaction mechanism of coupled binuclear copper (CBC) (metallo)enzymes. The redox chemistry catalyzed by CBC enzymes often employs molecular oxygen as a cofactor. This enables them to activate and subsequently catalyze a cascade of chemical processes leading to substrate hydroxylation. In particular, the reaction mechanism of CBC enzyme tyrosinase (Ty) which catalyzes the hydroxylation of tyrosine to *L*-3,4-dihydroxyphenylalanine (*L*-DOPA) and its subsequent oxidation to (DOPA-quinone), was elucidated by means of quantum and molecular mechanical (QM/MM) calculations. The complementary - experimental - part of the project was carried out in the group of Prof. Edward Solomon (Stanford University, U. S. A.) by employing various spectroscopic techniques, such as low-temperature resonance Raman and electron paramagnetic resonance spectroscopy, kinetic measurements, and biochemical experiments. It can be mentioned that throughout the “strongly correlated” experimental and computational efforts on the “strongly correlated” CBC enzyme (Ty), a comprehensive view of the Ty catalytic action has been achieved. Throughout the work on the project, it was very encouraging to find out how theory assisted in planning and designing experiments whereas the experimental data guided us through the manifolds of plausible reaction pathways, described in the literature or suggested by our QM/MM calculations.

The computational part, which comprises the main part of the dissertation, includes state-of-the-art methods of computational chemistry, including multireference *ab initio* calculations and (in our opinion still non-trivial) QM/MM modeling. Successful completion of the PhD project opens new horizons in understanding the fundamental biophysics of bioinorganic systems and helps in the development of new nature-inspired catalytic systems.

Keywords: DFT, QM/MM, [Cu_2O_2], CBC, tyrosinase