Electrochemical and computational methods as a combined approach to study redox enzymes

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Keywords: Redox enzymes; O_2 inhibition; Quantum mechanics; Protein film electrochemistry

Redox enzymes such as hydrogenases and CO dehydrogenases (CODHs) catalyze hydrogen production/oxidation and CO/CO_2 redox conversion, respectively, at unique inorganic active sites, buried within the protein structure. Understanding their functioning is crucial to enlighten how nature evolved the ability to catalyze such reactions, and to use those enzymes in biotechnological devices, or as inspiration for inorganic catalysts design[1]. Protein film electrochemistry (PFE) follows the activity of redox enzymes as a current, which can be quantitatively interpreted through detailed kinetic modelling. This provides information on the steps of the catalytic cycle[2] and on the rates of small molecule (e.g. molecular oxygen) inhibition[3]. However, PFE lacks structural information about the chemical species that are formed throughout catalysis, or during inhibition reactions. This information can be obtained with computational techniques, such as Quantum Mechanical (QM) calculations[4], which allows simulating the reaction steps of both the catalytic cycle or the reaction with inhibitors.

In particular, O_2 inhibition and reactivation of CODHs are poorly understood mechanisms. Moreover, enzymes from different organisms react with O_2 in different manners, despite the 1st coordination sphere of the active site being very conserved. Using PFE, I show that O_2 inhibits CODHs incredibly fast, and in a competitive manner, binding the same site as the substrate CO (manuscript in preparation). In my current project, I aim at using QM calculations and Molecular dynamics simulations to investigate the chemical steps that follow O_2 binding at the active site of CODHs and how the protein structure controls the chemistry of O_2 inhibition and reactivation in different CODHs.

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