Electron Transfer in Active Enzyme Molecules Adsorbed on Electrodes: Studies and Exploits of the Coupling to Proton Transfer and Catalysis.

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Enzyme molecules can be bound at electrodes such that interfacial electron transfer (ET) is sufficiently fast to study and exploit all the exquisite chemistry that these complex catalysts normally perform. There are many ongoing efforts to develop suitable electrodes, which include materials ranging from graphite to metals modified with surfactants and self-assembled monolayers of organo-thiols [1]. The nature and details of the interactions between protein and electrode are far from clear [2].

Faradaic electrochemical studies on adsorbed enzymes can be divided into two types – nonturnover and catalytic [3]. Non-turnover studies address the signals that arise from electron exchange between the enzyme's active sites and the electrode. The exchange is induced by modulating the potential, as in cyclic voltammetry, or using square-wave voltammetry which can enhance resolution of complex signals and ET kinetics [4,5]. The coverage of active enzyme must be sufficiently high to obtain well-defined and reproducible peak shapes: notably, detectability is greatly improved for a n = 2 electron center such as a flavin, due to the nernstian n^2 – dependence of peak current [6,7]. A simple and informative procedure is to analyze the oxidation and reduction peak potentials as a logarithmic function of scan rate. The resulting plot depicts how ET properties of an active site vary over different time domains and we can establish how ET is coupled to proton transfer and other processes [8,9].

Catalytic studies involve the addition of substrate to the solution, so that ET is now coupled to enzyme turnover. To control substrate supply and product removal, the electrode is usually rotated. Analysis of the catalytic waves now yields detailed information on how rates depend on potential [6,10-12]. Depending on the properties of the enzyme being investigated, the shape and potential of the wave may reflect the properties of the enzyme's intramolecular relay system (if interfacial ET is fast relative to intramolecular ET) or of the catalytic active site (if both interfacial and intramolecular ET are fast) [10]. The 'potential dimension' is a relatively unexplored variable in enzyme kinetics. The experiments yield potentials and rates for different intermediates in the catalytic cycle and highlight interesting effects such as potential optima – the counterpart of long-established pH optima for enzyme activity. Another appealing aspect is the ability to modulate the potential at a rate exceeding the turnover number, from which properties of the Enzyme-Substrate complex are extracted [6,7].

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