Mathematical Model behind the Protein-Film Voltammetry of a Surface EC' Catalytic Mechanism under Butler–Volmer Kinetics

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Protein-film voltammetry is a simple and powerful electrochemical methodology designed to provide direct access to electron-transfer reactions of redox-active proteins immobilized at the electrode surface. In such systems, the reacting redox centers are confined to the interfacial region and no diffusional transport of the protein from the bulk solution toward the electrode is involved. Consequently, the voltammetric response is governed by the amount of electroactive material adsorbed at the electrode, by the heterogeneous electron-transfer kinetics, and by possible chemical reactions coupled to the electrode process. The surface EC' mechanism considered here is a regenerative catalytic mechanism. In this mechanism, the reduced form of the adsorbed protein undergoes a one-electron oxidation at the electrode surface, whereas the oxidized form is regenerated chemically back to the reduced form by reaction with another adsorbed or interfacially available species, denoted as $Y(ads)$. Such mechanisms are highly relevant in protein-film voltammetry, because many redox proteins and enzymes operate through catalytic cycles in which the electrochemically generated state is rapidly converted back to the initial redox form by a chemical reaction. This type of regeneration is particularly important for redox enzymes, metalloproteins, and surface-confined catalytic systems, where electron transfer and chemical turnover are strongly coupled.

The mechanism can be written as follows:

$$\mathrm{Red}_{ads} \rightleftharpoons \mathrm{Ox}_{ads} + e^- \qquad \text{(I)}$$

$$\mathrm{Ox}_{ads} + \mathrm{Y}_{ads} \rightarrow \mathrm{Red}_{ads} \qquad \text{(II)}$$

The first reaction (I) is the electrode reaction. It is characterized by the standard heterogeneous electron-transfer rate constant, k_s, and by the electron-transfer coefficient, α. The second reaction (II) is the regenerative catalytic homogeneous chemical reaction. It is characterized by a second-order surface chemical rate constant, k_{cat}, if the concentration of $Y(ads)$ changes during the experiment. When $Y(ads)$ is present in large excess, or when its interfacial concentration is effectively constant, the catalytic reaction can be treated as a pseudo-first-order step with the apparent catalytic rate constant:

$$k_c = k_{cat}\Gamma_Y^*$$

where Γ_Y^* is the constant surface concentration of the catalytic co-reactant. Under this condition, the regenerative reaction is described as:

$$\mathrm{Ox}_{ads} \overset{k_c}{\rightarrow} \mathrm{Red}_{ads}$$

The total surface concentration of the protein redox couple is conserved:

$$\Gamma_{Red} + \Gamma_{Ox} = \Gamma^*$$

In this equation, Γ^* is the total surface coverage of the immobilized protein, while Γ_{Red} and Γ_{Ox} are the surface concentrations of the reduced and oxidized forms, respectively. This conservation condition is fundamental for protein-film voltammetry, because the redox-active protein does not leave the electrode surface. The chemical species $Y(ads)$ may either be treated as a constant excess reagent or, in a more general formulation, as an additional surface species with its own kinetic equation.

The potential-dependent electron-transfer kinetics are described by the Butler–Volmer formalism. The oxidation rate constant (k_{ox}) is:

$$k_{ox} = k_s \exp\left[\frac{\alpha F(E - E^0)}{RT}\right]$$

and the reduction rate constant (k_{red}) is:

$$k_{red} = k_s \exp\left[-\frac{(1-\alpha)F(E - E^0)}{RT}\right]$$

In these last two equations, E is the applied electrode potential, E^0 is the formal potential of the immobilized redox couple, F is the Faraday constant, R is the gas constant, and T is the absolute temperature. The standard rate constant k_s determines how rapidly the immobilized redox center exchanges electrons with the electrode at the formal potential. Large values of k_s correspond to nearly reversible electron transfer, whereas small values lead to quasireversible behavior, peak broadening, peak displacement, and suppression of the backward component of the voltammetric response.

The transfer coefficient α describes the symmetry of the activation barrier. When $\alpha = 0.5$, the oxidation and reduction barriers respond symmetrically to changes in electrode potential. If α differs from 0.5, the anodic and cathodic branches of the Butler–Volmer response become asymmetric, which can influence both the position and the shape of the voltammetric peaks.

For the pseudo-first-order catalytic case, the kinetic equations for the surface concentrations are:

$$\frac{d\Gamma_{Red}}{dt} = -k_{ox}\Gamma_{Red} + k_{red}\Gamma_{Ox} + k_c\Gamma_{Ox}$$

$$\frac{d\Gamma_{Ox}}{dt} = k_{ox}\Gamma_{Red} - k_{red}\Gamma_{Ox} - k_c\Gamma_{Ox}$$

The first equation describes the loss of $Red(ads)$ through electrochemical oxidation and its regeneration through both electrochemical reduction and catalytic chemical reaction. The second equation describes the formation of $Ox(ads)$ by oxidation of $Red(ads)$, followed by its consumption through electrochemical reduction and through the regenerative catalytic chemical step. Addition of the two equations gives:

$$\frac{d}{dt}(\Gamma_{Red} + \Gamma_{Ox}) = 0$$

which confirms that the total amount of immobilized protein is conserved during the experiment.

For the more general case in which the concentration of $Y(ads)$ is not constant, the full second-order kinetic model is:

$$\frac{d\Gamma_{Red}}{dt} = -k_{ox}\Gamma_{Red} + k_{red}\Gamma_{Ox} + k_{cat}\Gamma_{Ox}\Gamma_{Y}$$

$$\frac{d\Gamma_{Ox}}{dt} = k_{ox}\Gamma_{Red} - k_{red}\Gamma_{Ox} - k_{cat}\Gamma_{Ox}\Gamma_{Y}$$

$$\frac{d\Gamma_{Y}}{dt} = -k_{cat}\Gamma_{Ox}\Gamma_{Y}$$

This system describes catalytic regeneration accompanied by consumption of the adsorbed co-reactant $Y(ads)$. In many protein-film voltammetric experiments, however, Y is present in large excess in solution or is continuously supplied to the interface. Under those conditions, Γ_Y can be considered constant and the pseudo-first-order formulation is preferable.

At the beginning of a voltammetric experiment, the initial conditions are usually selected according to the starting potential. If the experiment starts at a sufficiently negative potential, the immobilized protein film is assumed to be fully reduced:

$$\Gamma_{Red}(0) = \Gamma^*$$

$$\Gamma_{Ox}(0) = 0$$

$$\Gamma_{Y}(0) = \Gamma_Y^*$$

These initial conditions are appropriate when the starting potential stabilizes the reduced protein form and when the catalytic co-reactant is already present at the interface. If the experiment starts from a more positive potential, a different initial distribution may be required, for example a partially oxidized film:

$$\Gamma_{Red}(0) = \theta\Gamma^*$$

$$\Gamma_{Ox}(0) = (1-\theta)\Gamma^*$$

where $0 \leq \theta \leq 1$ defines the initial fraction of the reduced form.

The boundary conditions for a strictly surface-confined protein-film system are different from those of diffusion-controlled voltammetry. Since $Red(ads)$ and $Ox(ads)$ are immobilized, no flux condition for the protein species is required. Instead, the essential boundary condition is the conservation of surface coverage:

$$\Gamma_{Red}(t) + \Gamma_{Ox}(t) = \Gamma^*$$

If Y is supplied from the bulk solution in large excess, its interfacial concentration remains effectively constant:

$$\Gamma_Y(t) = \Gamma_Y^*$$

and therefore:

$$k_c = k_{cat}\Gamma_Y^* = \text{constant}$$

This boundary condition is important because it converts the catalytic reaction into a pseudo-first-order regeneration process.

Using the conservation equation, one may write:

$$\Gamma_{Red} = \Gamma^* - \Gamma_{Ox}$$

Substitution into the kinetic equation for Γ_{Ox} gives:

$$\frac{d\Gamma_{Ox}}{dt} = k_{ox}(\Gamma^* - \Gamma_{Ox}) - (k_{red} + k_c)\Gamma_{Ox}$$

or:

$$\frac{d\Gamma_{Ox}}{dt} = k_{ox}\Gamma^* - (k_{ox} + k_{red} + k_c)\Gamma_{Ox}$$

At a fixed potential, where k_{ox} and k_{red} are constant, the solution of this equation is:

$$\Gamma_{Ox}(t) = \Gamma_{e,Ox} + \left[\Gamma_{Ox}(0) - \Gamma_{e,Ox}\right]\exp[-(k_{ox} + k_{red} + k_c)t]$$

where the stationary, or instantaneous equilibrium-like, surface concentration of the oxidized form is:

$$\Gamma_{e,Ox} = \frac{k_{ox}\Gamma^*}{k_{ox} + k_{red} + k_c}$$

The corresponding surface concentration of the reduced form is:

$$\Gamma_{e,Red} = \Gamma^* - \Gamma_{e,Ox}$$

or explicitly:

$$\Gamma_{e,Red} = \frac{(k_{red} + k_c)\Gamma^*}{k_{ox} + k_{red} + k_c}$$

These expressions are very useful because they directly show how the catalytic regeneration step modifies the distribution of the adsorbed redox forms. When $k_c = 0$, the stationary distribution is determined only by the Butler–Volmer oxidation and reduction rates. When k_c increases, the oxidized form is more rapidly converted back into the reduced form, and the stationary concentration of $Red(ads)$ increases. Thus, catalytic regeneration maintains a larger population of electroactive reduced species available for further oxidation.

For a surface-confined system, the faradaic current is proportional to the net rate of electron transfer between the electrode and the adsorbed film:

$$I = nFA(k_{ox}\Gamma_{Red} - k_{red}\Gamma_{Ox})$$

where n is the number of transferred electrons and A is the electrode area. The first term, $k_{ox}\Gamma_{Red}$, represents the anodic oxidation flux of $Red(ads)$ into $Ox(ads)$. The second term, $k_{red}\Gamma_{Ox}$, represents the cathodic reduction flux of $Ox(ads)$ back to $Red(ads)$. Therefore, the measured current is the difference between the forward and backward electron-transfer rates.

By incorporating the evaluated stationary surface concentrations into the Butler–Volmer current expression, one obtains:

$$I_e = nFA\left(k_{ox}\Gamma_{e,Red} - k_{red}\Gamma_{e,Ox}\right)$$

Substituting $\Gamma_{e,Red}$ and $\Gamma_{e,Ox}$ gives:

$$I_e = nFA\left[\frac{k_{ox}(k_{red}+k_c)\Gamma^*}{k_{ox}+k_{red}+k_c} - \frac{k_{red}k_{ox}\Gamma^*}{k_{ox}+k_{red}+k_c}\right]$$

After simplification, the catalytic current becomes:

$$I_e = nFA\Gamma^*\frac{k_{ox}k_c}{k_{ox}+k_{red}+k_c}$$

This equation is central for understanding the surface EC' mechanism. It shows that the current depends directly on the electron-transfer rate constant for oxidation and on the catalytic regeneration rate constant. If $k_c = 0$, the stationary catalytic current disappears. If k_c is large, $Ox(ads)$ is rapidly regenerated to $Red(ads)$, which sustains the anodic current and leads to catalytic amplification of the voltammetric signal.

When the Butler–Volmer expressions are inserted explicitly, the catalytic current can be written as:

$$I_e = nFA\Gamma^*\frac{k_s\exp\left[\frac{\alpha F(E-E^0)}{RT}\right]k_c}{k_s\exp\left[\frac{\alpha F(E-E^0)}{RT}\right] + k_s\exp\left[-\frac{(1-\alpha)F(E-E^0)}{RT}\right] + k_c}$$

This final expression links the measured catalytic current directly to the applied potential, heterogeneous electron-transfer kinetics, catalytic regeneration rate, temperature, transfer coefficient, and total surface coverage. It is particularly useful for analyzing catalytic protein-film voltammograms, because it explicitly couples Butler–Volmer electron transfer with the regenerative chemical reaction.

For numerical simulations of dynamic voltammetric techniques, such as square-wave voltammetry or cyclic staircase voltammetry, the differential equations are solved stepwise. At a given time step j, the potential is E_j, and the rate constants are:

$$k_{ox,j} = k_s\exp\left[\frac{\alpha F\left(E_j - E^0\right)}{RT}\right]$$

$$k_{red,j} = k_s\exp\left[-\frac{(1-\alpha)F\left(E_j - E^0\right)}{RT}\right]$$

The surface concentrations are then updated according to:

$$\Gamma_{Red,j+1} = \Gamma_{Red,j} + \Delta t\left[-k_{ox,j}\Gamma_{Red,j} + k_{red,j}\Gamma_{Ox,j} + k_c\Gamma_{Ox,j}\right]$$

$$\Gamma_{Ox,j+1} = \Gamma_{Ox,j} + \Delta t\left[k_{ox,j}\Gamma_{Red,j} - k_{red,j}\Gamma_{Ox,j} - k_c\Gamma_{Ox,j}\right]$$

After each update, the current is calculated from:

$$I_j = nFA\left(k_{ox,j}\Gamma_{Red,j} - k_{red,j}\Gamma_{Ox,j}\right)$$

This algorithm is especially important in square-wave voltammetry, where the potential changes rapidly between forward and backward pulses. Since k_{ox} and k_{red} depend exponentially on the applied potential, they must be recalculated at every potential step.

Several limiting cases can be obtained from the same model. If the catalytic regeneration is absent, then:

$$k_c = 0$$

and the mechanism reduces to a simple surface-confined electron-transfer reaction:

$$\mathrm{Red}_{ads} \rightleftharpoons \mathrm{Ox}_{ads} + e^-$$

Under this condition, the voltammetric response is controlled only by k_s, α, and the total surface coverage. No catalytic amplification occurs.

If the catalytic reaction is very fast, then:

$$k_c \gg k_{red}$$

and $Ox(ads)$ is rapidly converted back to $Red(ads)$. In this case, the anodic current can be strongly enhanced, because the electroactive reduced form is continuously regenerated at the electrode surface. This is the characteristic behavior of a regenerative catalytic EC' mechanism.

If electron transfer is fast:

$$k_s \to \infty$$

then the surface concentrations of $Red(ads)$ and $Ox(ads)$ tend to follow the applied potential almost reversibly, while the catalytic step determines the extent of current amplification. Conversely, when electron transfer is slow:

$$k_s \to 0$$

then the overall response becomes limited by heterogeneous electron transfer, and catalytic regeneration may have only a weak observable effect because $Ox(ads)$ is produced slowly.

Thus, the surface EC' mechanism treated with Butler–Volmer kinetics provides a flexible theoretical framework for protein-film voltammetry. It allows simultaneous evaluation of heterogeneous electron-transfer kinetics, catalytic regeneration, and redistribution of surface concentrations. The most important feature of this formalism is that the experimentally measured current is directly linked to the dynamic surface concentrations of $Red(ads)$ and $Ox(ads)$, which are controlled by both electrode potential and regenerative catalytic chemistry. Entire MATHCAD simulation protocols of this

mechanism, ready for simulating square-wave and cyclic voltammograms, can be implemented using the differential equations and finite-difference expressions given above. Under conditions of cyclic voltammetry, the corresponding simulation Mathcad file is available at:

From theory to simulation: Open interactive MATHCAD simulation protocols for exploring common electrode mechanisms in cyclic voltammetry | Macedonian Journal of Chemistry and Chemical Engineering

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