A Unified Framework of ECrevEC′ Mechanism in Protein-Film Cyclic Voltammetry: A Master Model for Mechanistic, Kinetic, and Thermodynamic Elucidation of Complex Redox Systems whose Electrode Transformation Takes Place from Adsorbed State

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Abstract

The interpretation of cyclic voltammetric responses of complex redox systems remains a major challenge, particularly for processes involving coupled chemical steps and catalytic regeneration. In this work, we introduce a comprehensive surface-confined ECrevEC′ mechanism that serves as a *master theoretical framework* capable of unifying a wide range of classical electrochemical mechanisms within a single simulation platform. The model describes two sequential electron-transfer steps separated by a reversible chemical transformation, followed by a regenerative catalytic step, all occurring under surface-controlled conditions.

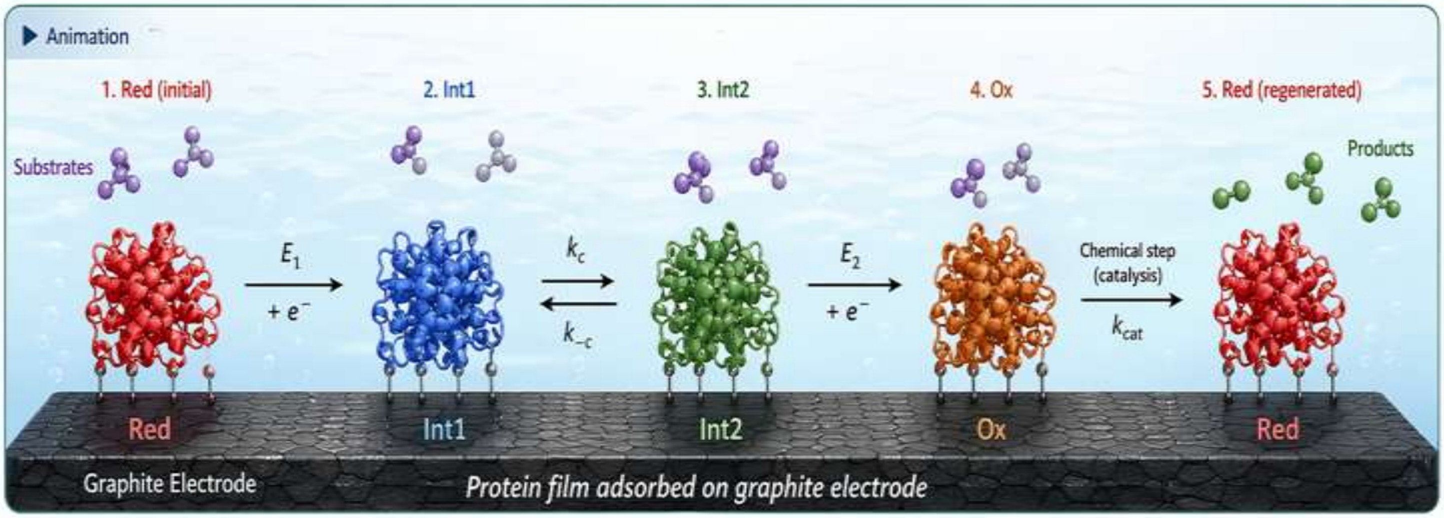
Through systematic numerical simulations, we demonstrate that this generalized mechanism converges, under well-defined limiting cases, to simpler mechanisms including **E, EE, ECrev, EC′, ECrevE, CrevE, and EEC′**. The resulting voltammetric patterns exhibit rich features such as peak splitting, asymmetry, quasireversible maxima, and catalytic amplification, providing direct access to kinetic parameters (ks, kc, kcat), equilibrium constants (Keq), and thermodynamic constraints of the system.

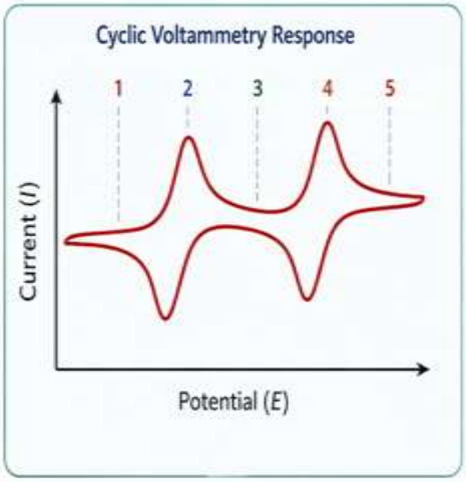
Particular emphasis is placed on the applicability of the model in protein-film voltammetry (PFV)**,** where immobilized redox-active biomolecules often undergo multi-step electron-transfer processes coupled with intramolecular rearrangements and catalytic cycles. The proposed framework enables rigorous interpretation of voltammetric data from lipophilic enzymatic systems, including quinone-based, flavin-based, and NAD(P)H-dependent redox pathways, offering new insights into their mechanistic complexity and interfacial behavior.

Surface EC*revEC′* Mechanism in Protein-Film Voltammetry

All protein forms are adsorbed on the graphite electrode surface and submerged in aqueous solution containing substrates.

$EC_{rev}EC'$





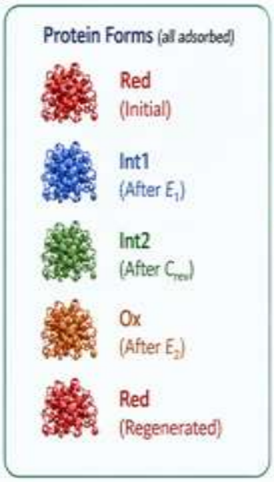
Mechanism Summary

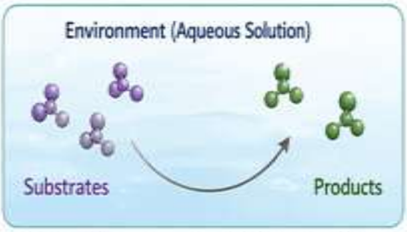
E_1 (Electron transfer 1)
$\mathrm{Red} + e^- \rightleftharpoons \mathrm{Int1}$

C_{rev} (Reversible chemical step)
$\mathrm{Int1} \rightleftharpoons \mathrm{Int2}$
(k_c, k_{-c})

E_2 (Electron transfer 2)
$\mathrm{Int2} + e^- \rightleftharpoons \mathrm{Ox}$

C' (Chemical catalytic step)
$\mathrm{Ox} + \mathrm{Substrates} \rightarrow \mathrm{Red} + \mathrm{Products}$
(k_{cat})





Common in PFV Systems

- Redox enzymes (e.g., dehydrogenases, oxidases)
- Quinoproteins, flavoproteins
- Lipophilic and membrane-associated enzymes
- Catalytic turnover within protein film

The $EC_{rev}EC'$ mechanism unifies multiple pathways in cyclic voltammetry and reveals mechanistic, kinetic and thermodynamic insights into complex enzymatic redox systems at electrode–solution interfaces.

MATHCAD SIMULATION FILE of SURFACE ECrevEC’ Mechanism in Cyclic Voltammetry

$EfI := 0.7$ $EfII := .4$ $\tau := 0.05$

$EsI := -0.5$ $\Delta E := 1.2$ $dE := 0.01$ $EsII := -0.8$ $\varepsilon := 1..1$

$n := 1$ $F := 96500$ $R := 8.314$ $T := 298.15$ $\Delta EI := EfI - EsI$ $KI := 10^{-1}$ $kc1 := 100$

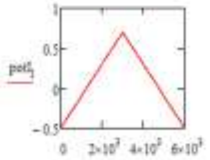
$M := 25$ $\Delta EII := EfII - EsII$ $KII := 10^{-1}$ $kc2 := 200$

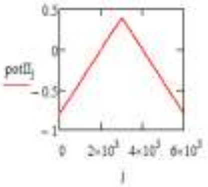
$j := 1..2\frac{\Delta E}{dE}\cdot 25$ $d := \frac{\tau}{M}$ $\alpha 2 := 0.5$ $\alpha 1 := 0.5$

λ i z se definirani kako odnos na Kc1x τ i kc2xτ

$$potI_j := if\left[j \le \frac{\Delta E}{dE}\cdot 25, EsI + \left(ceil\left(\frac{j}{25}\right)\cdot dE - dE\right), EfI - \left[ceil\left[\frac{j - \left(\frac{\Delta E}{dE}\cdot 25\right)}{25}\right]\cdot dE - dE\right]\right]$$

$$potII_j := if\left[j \le \frac{\Delta E}{dE}\cdot 25, EsII + \left(ceil\left(\frac{j}{25}\right)\cdot dE - dE\right), EfII - \left[ceil\left[\frac{j - \left(\frac{\Delta E}{dE}\cdot 25\right)}{25}\right]\cdot dE - dE\right]\right]$$





$\Phi I_j := n\cdot\frac{F}{R\cdot T}\cdot potI_j$ $\Phi II_j := n\cdot\frac{F}{R\cdot T}\cdot potII_j$

$$\Psi I_1 := \frac{KI\cdot e^{\alpha 1\cdot \Phi I_1}}{1 - KI\cdot e^{\alpha 1\cdot \Phi I_1}\left[\frac{-1}{50}\left(\left(1 + \frac{e^{-\Phi I_1}}{1 + Keq}\right)\right) - \frac{1\cdot e^{-\Phi I_1}\cdot A_1}{\lambda\cdot(Keq + 1)}\right]}$$

$$\Psi II_1 := \frac{KII\cdot\frac{1}{50}\cdot e^{\alpha 2\cdot \Phi II_1}}{1 + \frac{1\cdot B_1}{(z)}KII\cdot e^{\Phi II_1\cdot\alpha 2} + \frac{1\cdot B_1}{(z)}KII\cdot e^{-\Phi II_1\cdot(1-\alpha 2)}}$$

$\Psi II_1 = 1.112\times 10^{-11}$

$\Psi I_1 = 1.038\times 10^{-7}$

$$\Psi I_j := \frac{KI\cdot e^{\alpha 1\cdot \Phi I_j}\left[1 - \frac{1}{50}\left(1 + \frac{e^{-\Phi I_j}}{1 + Keq}\right)\sum_{i=1}^{j-1}\left(\Psi I_i\right) - \frac{1\cdot e^{-\Phi I_j}}{(1 + Keq)\cdot\lambda}\sum_{i=1}^{j-1}\left(\Psi I_i\cdot A_{j-i+1}\right)\right]}{1 - KI\cdot e^{\alpha 1\cdot \Phi I_j}\left[\frac{-1}{50}\left(\left(1 + \frac{e^{-\Phi I_j}}{1 + Keq}\right)\right) - \frac{1\cdot e^{-\Phi I_j}\cdot A_1}{\lambda\cdot(Keq + 1)}\right]}$$

$$\Psi II_j := \frac{\frac{KII\cdot e^{-\Phi II_j\cdot(1-\alpha 2)}}{(1 + Keq)\cdot\lambda}\left[\sum_{i=1}^{j}\left(\Psi I_i\cdot A_{j-i+1}\right)\right] - KII\cdot e^{\alpha 2\cdot \Phi II_j}\left[-1 + \frac{1 + e^{-\Phi II_j}}{z}\cdot\sum_{i=1}^{j-1}\left(\Psi II_i\cdot B_{j-i+1}\right)\right]}{1 + \frac{KII\cdot e^{-\Phi II_j\cdot(1-\alpha 2)}}{(1 + Keq)\cdot\lambda}\cdot A_1 + \frac{KII\cdot e^{\alpha 2\cdot \Phi II_j}}{z}\cdot B_1\left(1 + e^{-\Phi II_j}\right) + KII\cdot e^{\alpha 2\cdot \Phi II_j}\cdot B_1} = \ldots$$

$$E_1:\ \mathrm{Red} \rightleftarrows \mathrm{Int1} + e^{-}\quad (k_{s,1;}\alpha_1) \qquad \text{(a)}$$

$$C_{rev}:\ \mathrm{Int1} \rightleftarrows \mathrm{Int2}\quad \left(k_f, k_b;\ K_{eq} = \frac{k_f}{k_b};\ k_{chem} = k_f + k_b\right) \qquad \text{(b)}$$

$$E_2:\ \mathrm{Int2} \rightleftarrows \mathrm{Ox} + e^{-}\quad (k_{s,2}, \alpha_2) \qquad \text{(c)}$$

$$C'(\mathrm{regenerative}):\ \mathrm{Ox} + \mathrm{Y} \rightarrow \mathrm{Int2}\quad (k_c) \qquad \text{(d)}$$

λ e kineticki parametar na reverzibilna hemiska reakcija (follow up) povrzana so prv elektroden cekor

MODEL ASSUMES
ALL ADSORBED
redox species

$KII = 0.1$

PROTEIN-FILM
CYCLIC VOLTAMMETRY

$\lambda := 00.017550000010007510000$

$Keq := .200000006051000$

$z := 222.500001$

z e katalticki parametar vo ovoj model povrzan so vtor cekor

$A_j := e^{-\lambda\cdot\frac{j}{50}} - e^{-\lambda\cdot\frac{j+1}{50}}$ $B_j := e^{-z\cdot\frac{j}{50}} - e^{-z\cdot\frac{j+1}{50}}$

$p := 1..2\cdot\frac{\Delta EI}{dE} - 1$

serial number of potential steps

$\Psi I_p := \Psi I_{\left(\frac{\tau}{4\cdot 25}+p\right)\cdot 25}$

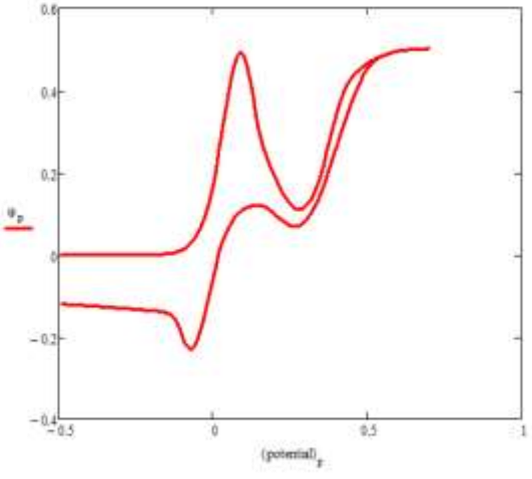
$\Psi II_p := \Psi II_{\left(\frac{\tau}{4\cdot 25}+p\right)\cdot 25}$

$\Psi_p := \Psi I_p + \Psi II_p$

dimesnioless current at the end of each potential step of first electrode reaction ΨI and of second selectrode reaction ΨII Ψp is symbol the cumulative current measured as final output

$\text{potential}_p := \text{if}\left[p \le \frac{\Delta EI}{dE}, EsI + p\cdot dE, EfI - \left(p - \frac{\Delta EI}{dE}\right)\cdot dE\right]$

potential value of each potential step in V

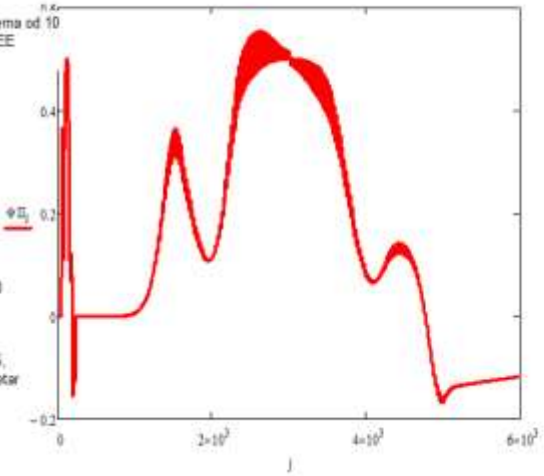


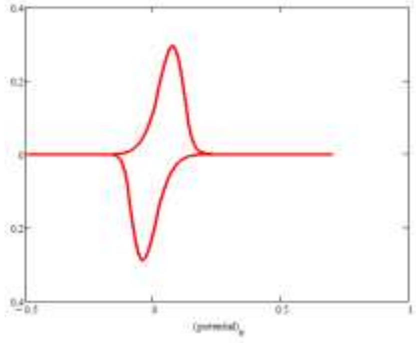
Pri Keq od 0.01 0.1 i 1 lambda pogolema od 10
konstantni i isti se dvata pikovi kako EE
to e OK

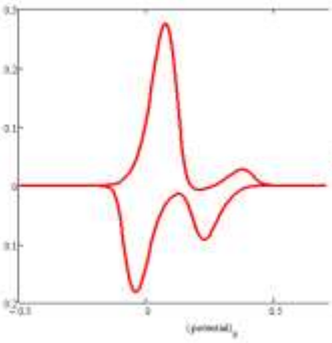
OVOJ e TOCHEN MODEL
ECrevEC' SURFACE
vo ciklicna
18 04 2026ta

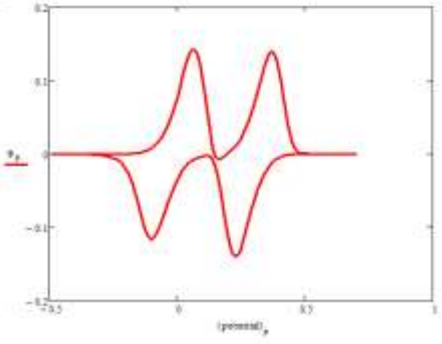
Keq od 1 and lambda of 100
equalts to EE mechanism

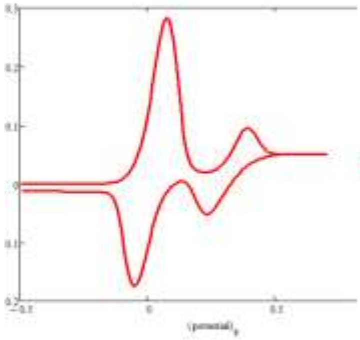
Vazni Fenomeni ima pri
Keq 0.01 i pri malo lambda od 0.05,
da se zgolemuva katalticki parametar
od 0.01 do 1...

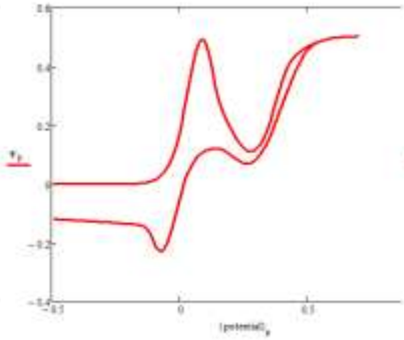


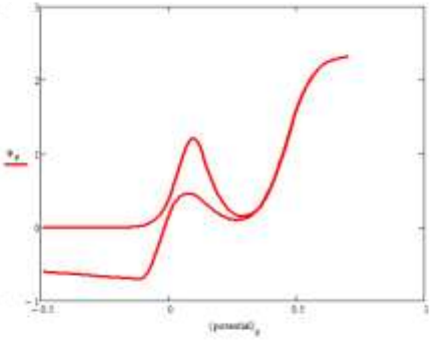












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