# Integrating of Common Mechanisms in Protein-Film Voltammetry-MATHCAD Simulation Protocol

***Milkica Janeva, Pavlinka Kokoskarova, Sanja Lazarova, Rubin Gulaboski***

**Faculty of Medical Sciences, Goce Delcev University, Stip, Macedonia**

## Abstract

Protein-film voltammetry (PFV) provides a powerful methodology for investigating electron-transfer processes and coupled chemical reactions of surface-confined proteins and enzymes. In this work, for the firt time, a comprehensive theoretical framework is presented for the surface ***CirrE$_1$CrevE$_2$C′ mechanism***, integrating a preceding irreversible chemical transformation, two consecutive electron-transfer steps interconnected by a reversible chemical reaction, and a final regenerative step. Owing to its multistep architecture, the model encompasses several commonly encountered PFV mechanisms as limiting cases, providing a unified platform for their systematic analysis. Simulated cyclic voltammetric patterns demonstrate how the kinetics of individual chemical and electron-transfer steps, the equilibrium constant of the intermediate reversible reaction, and catalytic regeneration govern the shape, magnitude, reversibility, and potential position of the voltammetric responses. Particular attention is given to systems with closely spaced electron-transfer potentials, where catalytic kinetics can assist in resolving otherwise overlapping redox processes. Importantly, the *original Mathcad simulation protocol* for the CirrE$_1$CrevE$_2$C′ mechanism is provided, enabling straightforward variation of relevant kinetic and thermodynamic parameters. The presented framework offers a practical computational platform for simulation, mechanistic exploration, and interpretation of complex reaction pathways encountered in protein-film voltammetry.

# MATHCAD File of Surface CirrE1CrevE2C’ Mechanism in Cyclic Voltammetry

$EfI := 0.8 \quad EfII := .4 \quad \tau := 0.05$

$EsI := -0.4 \quad \Delta E := 1.2 \quad dE := 0.01 \quad EsII := -0.8 \quad r := 1..1$

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

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

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

u, λ and z
are defined as
u = Kc1x τ
λ = kc2xτ and
z = kcat x τ

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

$KII = 0.1$

$u := 01.50$

u is Kchem,1

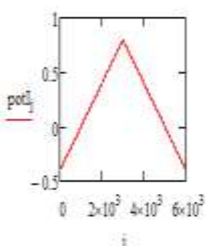
$$potII_j := if\left[j \le \frac{\Delta E}{dE}\cdot 25, EsII + \left(ceil\left(\frac{j}{25}\right)\cdot dE - dE\right), EfII - \left[ceil\left[\frac{j - \left(\frac{\Delta E}{dE}\cdot 25\right)}{25}\right]\cdot dE - dE\right]\right]$$

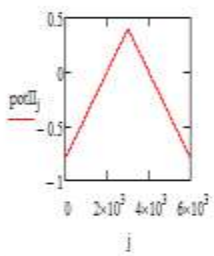
$\lambda := 0.500$

λ is Kchem,2 $\quad Keq := 00.50000605100$

$z := 0.14208102062$

z is Kcat




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

$$L_j := e^{-u\cdot\frac{j}{25}} - e^{-u\cdot\frac{j+1}{25}} \qquad A_j := e^{-\lambda\cdot\frac{j}{25}} - e^{-\lambda\cdot\frac{j+1}{25}} \qquad B_j := e^{-z\cdot\frac{j}{25}} - e^{-z\cdot\frac{j+1}{25}}$$

$$\Psi I_1 := \frac{KI\cdot e^{\alpha 1\cdot\Phi I_1}}{1 - KI\cdot e^{\alpha 1\cdot\Phi I_1}\cdot\left[\frac{-L_1}{u} + \frac{-1}{25}\cdot\left(\left(1 + \frac{e^{-\Phi I_1}}{1 + Keq}\right)\right) - \frac{1\cdot e^{-\Phi I_1}\cdot A_1}{\lambda\cdot(Keq + 1)}\right]}$$

$$\Psi II_1 := \frac{KII\cdot\frac{1}{25}\cdot e^{\alpha 2\cdot\Phi II_1}}{1 + \frac{1\cdot B_1}{(z)}KII\cdot e^{\Phi II_1\cdot\alpha 2} + \frac{1\cdot B_1}{(z)}KII\cdot e^{-\Phi II_1\cdot(1-\alpha 2)}}$$

$Cirr :=$

Cirr.. A--->Red (kchem,1)

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

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

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

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

**Surface CirrE1CrevE2C' in Cyclic Voltammetry**

PROTEIN-FILM
CYCLIC VOLTAMMETRY

MODEL ASSUM
ALL ADSORBEC
redox species

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

z e kataliticki parametar vo ovoj model povrzan so vtor cekor

$$\Psi II_1 = 3.007 \times 10^{-14}$$

$$\Psi I_1 = 3.045 \times 10^{-6}$$

$$\Psi I_j := \frac{KI \cdot e^{\alpha 1 \cdot \Psi I_j} \cdot \left[1 - \frac{1}{u} \left[\sum_{i=1}^{j-1} \left(\Psi I_i \cdot L_{j-i+1}\right)\right] - \frac{1}{25} \cdot \left(1 + \frac{e^{-\Psi I_j}}{1 + Keq}\right) \cdot \sum_{i=1}^{j-1} \left(\Psi I_i\right) - \frac{1 \cdot e^{-\Psi I_j}}{(1 + Keq) \cdot \lambda} \cdot \sum_{i=1}^{j-1} \left(\Psi I_i \cdot A_{j-i+1}\right)\right]}{1 - KI \cdot e^{\alpha 1 \cdot \Psi I_j} \cdot \left[\frac{-L_1}{u} - \frac{1}{25} \cdot \left(\left(1 + \frac{e^{-\Psi I_j}}{1 + Keq}\right)\right) - \frac{1 \cdot e^{-\Psi I_j} \cdot A_1}{\lambda \cdot (Keq + 1)}\right]}$$

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

$p := 1 .. 2 \cdot \frac{\Delta EI}{dE} - 1$ p is serial number of potential steps

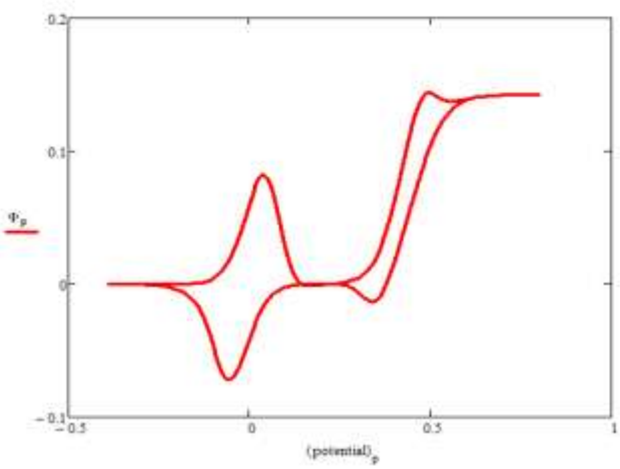
$\Psi I_p := \Psi I_{\left(\frac{\tau}{d \cdot 25} + p\right) \cdot 25}$

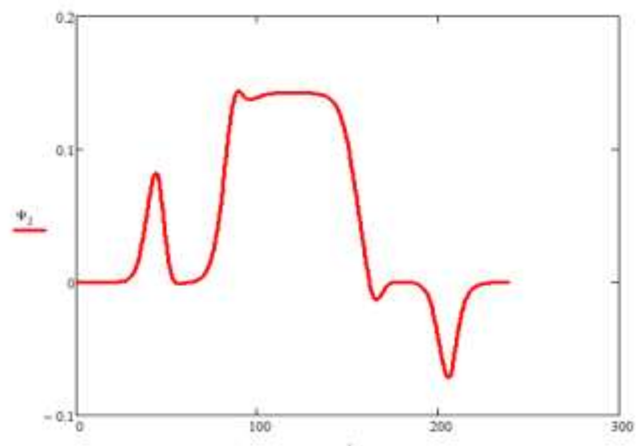
$\Psi II_p := \Psi II_{\left(\frac{\tau}{d \cdot 25} + p\right) \cdot 25}$

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

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

$\text{potential}_p := \text{if}\left[p \le \frac{\Delta EI}{dE}, Esl + p \cdot dE, Efl - \left(p - \frac{\Delta EI}{dE}\right) \cdot dE\right]$ potential value of each potential step in V



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