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Analytical solution of non-linear reaction diffusion equations in an amperometric immobilized enzyme electrode

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ABSTRACT

The boundary value problem in basic enzyme reactions is formulated and approximate expressions for mediator and substrate concentration are presented. This investigation contains a non-linear term related to Michaelis-Menten kinetics. In this paper, we obtain the approximate analytical solutions for the non-linear reaction diffusion equations that describe the diffusion and the reaction within a uniform film containing immobilized enzyme and mediator at an electrode surface. Analytical expression pertaining to the mediator and substrate concentration profiles and current response were reported for all possible values of dimensionless parameter κ, γ, η and μ. An approximate expression of flux is also derived. The obtained concentration results are compared with the numerical solution acquired using Scilab program and found to be in satisfactory agreement.

KEYWORDS

Amperometric electrodes;
Non-linear reaction-diffusion equations;
Redox enzyme;
Michaelis-Menten kinetics;
Homotopy perturbation method.

INTRODUCTION

In recent times, amperometric immobilized enzyme electrodes are finding increasing application in analytical chemistry and in electrochemical sensors[1,2]. These electrodes incorporate the specificity of the enzyme together with the rapid analysis time of the electrochemical detection. Rahamathunissa and Rajendran[3] acquired the analytical solutions for substrate concentration and transient current for both steady-state and non-steady-state amperometric polymer-modified electrodes by means of Danckwerts' relation. Andrieux et al.[4] and Albery and Hillman[5] analyzed the kinetics of reactions at polymer-modified electrodes.

During these reactions, species from the solution react with a mediator that was bound in a film at the electrode surface. The approximate analytical solutions can be acquired by linearizing the non-linear term[6]. In the case of an immobilized enzyme, the problem is further intricate by the non-linear enzyme kinetics. For the enzyme kinetics problem, approximate analytical solutions have been developed by Blaedel et al.[7], Kulys et al.[8] and Bartlett and Pratt[9] for the limiting cases (saturated and unsaturated). The relevance of numerical and approximate analytical methods can be perceived by Flexer et al.[10].

Recently, Senthamarai and Rajendran[11] derived the approximate analytical expressions for the substrate, mediator concentrations and current for the non-linear Michaelis-Menten kinetic scheme in a system of

Full Paper

coupled non-linear reaction-diffusion processes at conducting polymer-modified ultramicroelectrodes using the variational iteration method. More recently, for steady state conditions, Loghambal and Rajendran[12] obtained the approximate analytical solutions for the non-linear equations that describe diffusion and the reaction within the film by employing the Homotopy perturbation method. However, for non steady state conditions, there were no analytical results available till date that corresponds to the mediator concentration and substrate concentration for all likely values of dimensionless parameters κ, γ, η and μ. In this paper, we present the approximate analytical expressions for the concentrations of the mediator and substrate. The flux is determined corresponding to all possible values of the parameters κ, γ, η and μ. These parameters are explained below in Eq. 10. Moreover, herein we employ "Homotopy Perturbation Method" (HPM) to solve the non-linear reaction equation[13,14].

MATHEMATICAL FORMULATION OF THE PROBLEM AND ANALYSIS

Figure 1 illustrates the common kinetic design for an enzyme-membrane/electrode. *A* and *B* are the oxidized and reduced forms of the mediator. E_1 and E_2 are the oxidized and reduced forms of the enzyme. *S* and *P* are the substrate and the product of the enzymatic reaction, respectively. Diffusion of mediator *A* and substrate *S* arise within the film with diffusion coefficients D_A and D_S, respectively. Partition of the substrate between the film and the bulk solution is depicted by the partition coefficient K_S. The mediator partition is expressed by K_A. The reactions that occur within the film (Figure 1) in the kinetic scheme can be written as follows[6]:

$$A + E_2 \xrightarrow{k_A} B + E_1 \quad (1)$$

$$E_1 + S \xrightarrow{k_E} E_2 + P \quad (2)$$

and the reaction at the electrode is $B \rightarrow A$. Here, k_E and k_A are second-order rate constants that describe the reaction between the enzyme and the substrate and between the enzyme and the mediator, respectively. According to Michaelis-Menten kinetics, the following is true:

$$k_E = \frac{k_{cat}}{K_M + [S]} \quad (3)$$

where k_{cat} stand for the catalytic rate constant, and K_M denotes the Michaelis-Menten constant. The homogenous enzyme kinetics is elucidated by Eqs. 1-3 occur throughout the film from $x = 0$ to $x = l$, where l is the thickness of the membrane. We consider a situation in which the mediator is entrapped within the film. This situation does not include a separate soluble redox mediator that is re-oxidized on a conducting entrapment matrix. Here, the rate constants for a heterogeneous reaction on the supporting matrix must be considered. The differential equations that quantify the diffusion and reaction within the film may be written as follows[6,9]:

$$\frac{\partial[A]}{\partial t} = D_A \frac{\partial^2[A]}{\partial x^2} - k_A[E_2][A] \quad (4)$$

$$\frac{\partial[S]}{\partial t} = D_S \frac{\partial^2[S]}{\partial x^2} - \frac{k_{cat}[E_1][S]}{K_M + [S]} \quad (5)$$

$$\frac{\partial[E_1]}{\partial t} = k_A[A][E_2] - \frac{k_{cat}[E_1][S]}{K_M + [S]} \quad (6)$$

Assuming that the enzyme is bound within the film, is not free to diffuse and is in the steady- state $d[E_1] / dt = 0$, Eq. 6 leads to the following:

$$[E_2] = \frac{k_{cat}[E_\Sigma][S]}{k_A[A](K_M + [S]) + k_{cat}[S]} \quad (7)$$

where $[E_\Sigma] = [E_1] + [E_2]$ denotes the total concentration of the immobilized enzyme. Then, in the non steady-state, Eq. 4 and Eq. 5 are reduced to the following:

$$\frac{\partial[A]}{\partial t} = D_A \frac{\partial^2[A]}{\partial x^2} - \frac{k_A k_{cat}[A][S][E_\Sigma]}{k_A[A](K_M + [S]) + k_{cat}[S]} \quad (8)$$

$$\frac{\partial[S]}{\partial t} = D_S \frac{\partial^2[S]}{\partial x^2} - \frac{k_A k_{cat}[A][S][E_\Sigma]}{k_A[A](K_M + [S]) + k_{cat}[S]} \quad (9)$$

Eq. 8 and Eq. 9 are solved for the following boundary conditions:

$$t = 0, [A] = [A]_\infty, [S] = [S]_\infty K_S \quad (9a)$$

$$x = 0, [A] = [A]_\infty, \partial[S] / x = 0 \quad (9b)$$

and

$$x = l, \partial[A] / \partial x = 0, [S] = [S]_\infty K_S = 0 \quad (9c)$$

We make the non-linear differential Eq. 8 and Eq. 9 dimensionless by defining the following parameters:

$$a = [A] / K_A[B_\Sigma]_\infty, \; s = [S] / K_S[S]_\infty, \; \chi = x / l,$$

$$\kappa = l(k_A[E_\Sigma] / D)^{1/2}, \; \eta = \frac{k_A K_M}{k_{cat}},$$

$$\gamma = \frac{k_A K_A[B_\Sigma]_\infty K_M}{k_{cat} K_S[S]_\infty}, \; \mu = \frac{K_S[S]_\infty}{K_M}, \; \tau = \frac{Dt}{l^2} \quad (10)$$

We can assume that $D = D_A = D_S$. Here *a* is the dimensionless concentration of the mediator and *s* is the dimensionless concentration of the substrate. χ is the normalized distance from the electrode/membrane interface. κ describes the equilibrium constant between the diffusion of *B* within the film and its reaction with the enzyme. *l* is the film thickness. η denotes the relative quantity of depletion of the substrate and oxidized mediator within the film. The parameter γ represents the equilibrium constant between the two forms of the enzyme. The ratio of the substrate concentration within the film to the Michaelis constant is described by μ. The subscript ∞ denotes the concentration in the bulk solution. *a* and *s* are normalized with respect to the total concentrations $K_A[B_S]$ and $K_S[S]\infty$ of the two species within the film, where $[B_\Sigma] = [A] + [B]$, $K_A[B_\Sigma]\infty = [B_\Sigma]$, and $K_S[S]\infty = [S] + [P]$. When $\kappa << 1$, *B* can diffuse across the film before it reacts with the enzyme. For $\eta << 1$, consumption of the substrate is greater than mediator reduction, and for $\eta >> 1$, the mediator reduction is greater than consumption of the substrate. For $\gamma << 1$, all of the enzymes are in the E_2 form. For unsaturated Michaelis-Menten kinetics, $\mu << 1$. For saturated kinetics, $\mu >> 1$, Eq. 8 and Eq. 9 reduce to the following dimensionless forms[6]:

$$\frac{\partial a}{\partial \tau} = \frac{\partial^2 a}{\partial \chi^2} - \frac{\kappa^2 as}{\gamma a(1+\mu s)+s} \quad (11)$$

$$\frac{\partial s}{\partial \tau} = \frac{\partial^2 s}{\partial \chi^2} - \frac{\gamma \eta^{-1} \kappa^2 as}{\gamma a(1+\mu s)+s} \quad (12)$$

The initial and boundary conditions for Eq. 11 and Eq. 12 are as follows:

$$\tau = 0,\ a = a_\varepsilon,\ s = 1 \quad (12a)$$

$$\chi = 0,\ a = a_\varepsilon,\ \partial s / \partial \chi = 0 \quad (12b)$$

and

$$\chi = 1,\ s = 1,\ \partial a / \partial \chi = 0 \quad (12c)$$

The parameter ε is the dimensionless potential, which can be defined as

$$\varepsilon = (E - E^0)nF / RT \quad (13)$$

where E is the potential of an electrode, E^0 is the standard potential of an electrode, n is the number of electrons, F is the Faraday constant, R is the universal gas constant and *T* is the absolute temperature. Combining the Nernst equation $E = E^0 + (RT / nF) \ln ([A]_0 / [B]_0)$ and Eq. 13 gives the dimensionless oxidized mediator concentration a_ε at the electrode surface:

$$a_\varepsilon = 1 / [1 + \exp(-\varepsilon)] \quad (14)$$

Here $[A]_0$ and $[B]_0$ denote the concentration of the two forms of the mediator at the electrode surface. Eq. 14 gives the boundary condition for *a* at the electrode surface. The dimensionless flux of substrate (J_S) consumed at the electrode is considered as the following:

$$J_S = \frac{lj_S}{D_A[B_\Sigma]_\infty K_A} = \frac{\eta}{\gamma}\left(\frac{\partial s}{\partial \chi}\right)_{\chi=1} = \left(\frac{\partial a}{\partial \chi}\right)_{\chi=1} - \left(\frac{\partial a}{\partial \chi}\right)_{\chi=0} \quad (15)$$

and that of the mediator (J_{obs}) measured at the electrode is as follows:

$$J_{obs} = \frac{lj_{obs}}{D_A[B_\Sigma]_\infty K_A} = -\left(\frac{\partial a}{\partial \chi}\right)_{\chi=0} \quad (16)$$

With respect to the boundary condition, the flux of the substrate reacting within the film is equal to the observed flux: $J_{obs} = J_S$.

SOLUTION OF BOUNDARY VALUE PROBLEM USING THE HPM

Recently, many authors have applied the HPM to various problems and demonstrated the efficiency of the HPM for handling non-linear structures and solving various physics and engineering problems[15-18]. This process is a combination of homotopy in topology and classic perturbation techniques. Ji-Huan He used the HPM to solve the Lighthill equation[19], the Duffing equation[20] and the Blasius equation[21]. The idea has been used to solve non-linear boundary value problems[22], integral equations[23-25], Klein-Gordon and Sine-Gordon equation[26], Emden-Flower type equations[27] and various other problems. This wide variety of applications shows the power of the HPM to solve functional equations.

The HPM is unique in its applicability, accuracy and efficiency. More recently, Meena and Rajendran[28-31] presented an analysis of system of coupled non-linear reaction diffusions within an electroactive polymer film deposited on an inlaid microdisc electrode using HPM which uses the imbedding parameter *p* as a small parameter, and only a few iterations are needed to search for an asymptotic solution. Using this method (see Appendix B), we can obtain the following solution to Eq. 11 and Eq. 12:

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$$a = a_s + \frac{k^2 a_s \chi^2}{2\left[\gamma a_s(1+\mu)+1\right]} - \frac{k^2 a_s \chi}{\left[\gamma a_s(1+\mu)+1\right]} + \frac{16k^2 a_s}{\pi^3\left[\gamma a_s(1+\mu)+1\right]}\sum_{n=0}^{\infty}\frac{(-1)^n \cos\left(\frac{(2n+1)\pi(\chi-1)}{2}\right)e^{\frac{-(2n+1)^2\pi^2}{4}\tau}}{(2n+1)^3} \quad (17)$$

$$s = 1 + \frac{\gamma k^2 a_s \chi^2}{2\eta\left[\gamma a_s(1+\mu)+1\right]} - \frac{\gamma k^2 a_s}{2\eta\left[\gamma a_s(1+\mu)+1\right]} + \frac{16\gamma k^2 a_s}{\pi^3\eta\left[\gamma a_s(1+\mu)+1\right]}\sum_{n=0}^{\infty}\frac{(-1)^n \cos\left(\frac{(2n+1)\pi\chi}{2}\right)e^{\frac{-(2n+1)^2\pi^2}{4}\tau}}{(2n+1)^3} \quad (18)$$

Eq. 17 and Eq. 18 represents the analytical expression of the mediator concentration and the substrate concentration for all values of the parameters κ, γ, η and μ and satisfies the boundary conditions Eqs. 12*a-c*. We can obtain the dimensionless flux, which is as follows:

$$J_S = J_{obs} = \frac{\eta}{\gamma}\left(\frac{\gamma k^2 a_s}{\eta\left[\gamma a_s(1+\mu)+1\right]} - \frac{8\gamma k^2 a_s}{\pi^2\eta\left[\gamma a_s(1+\mu)+1\right]}\sum_{n=0}^{\infty}\frac{e^{\frac{-(2n+1)^2\pi^2}{4}\tau}}{(2n+1)^2}\right) \quad (19)$$

Eq. 19 is the new approximate expression of the flux.

NUMERICAL SIMULATION

The nonlinear differential equations Eq. 11 and Eq. 12 are also solved by using numerical methods. The function pdex4 in Matlab software which is a function of solving two-point boundary value problems (BVPs) is used to solve those equations. Its numerical solution is compared with the solution obtained by using Homotopy perturbation method and it gives a satisfactory result. The Scilab program is also given in Appendix C.

RESULTS AND DISCUSSION

Eq. 17 and Eq. 18 are the new and simple analytical expressions of concentrations of the mediator and the substrate for all values of parameters κ, γ, η and μ. The dimensionless analytical expressions of concentration a and s for various values of dimensionless reaction parameters versus the dimensionless time τ compared with numerical solution. From Figure 2a it is inferred that when μ increases the value of the concentration of the mediator increases for the fixed vaues of k = 10 and γ = 100. Figure 2b represents the increase of mediator for the fixed values of k = 1 and μ = 10 when γ is varied. Figure 2c indicates the gradual decline of the mediator when k increase for the fixed values η = 1, μ = 5 and γ = 100. Figure 3a represents the decrease of the substrate concentration when η increases for the fixed values of η = 1, μ = 5 and γ = 100. Figure 3b represents the increase of the substrate concentration when η increase for the fixed values μ = 0.01, k = 1 and γ = 100. Figure 3c represents the rise of the substrate concentration when μ increases for the fixed values k = 1, η = 1 and γ = 100. Figure 3d represents the decline of the substrate concentration when γ increases for the fixed values μ = 0.01, k = 1 and η = 1. Figures 4a-c represents the flux and it abruptly reaches the steady state value when τ = 1 for all values of γ, μ and k. Also, the value of flux increases when the diffusion parameter γ and μ decreases while κ increases. From the Figure 4c, it is inferred that the flux mainly depends upon the equilibrium constant κ.

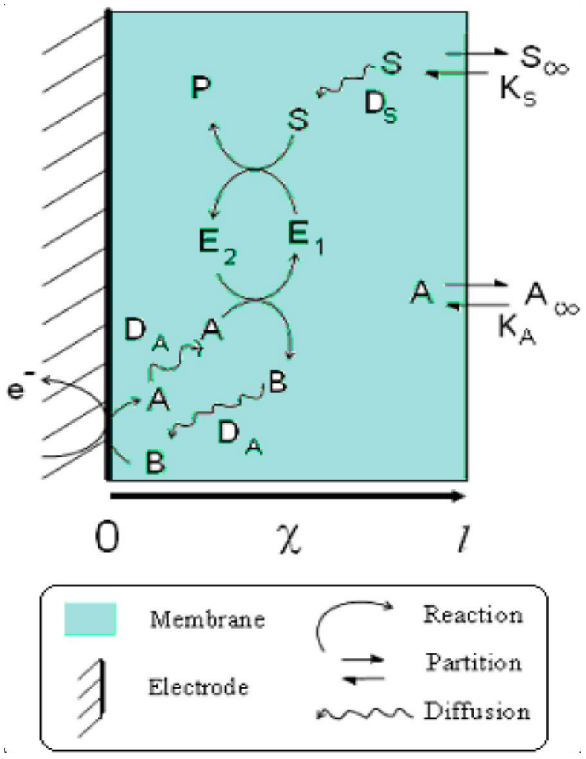

Figure 1 : Schematic representation of a typical enzyme-membrane electrode showing the processes considered in the model[6]. The homogenous enzyme kinetics that occurs throughout the film is described by Eqs. 1-3.

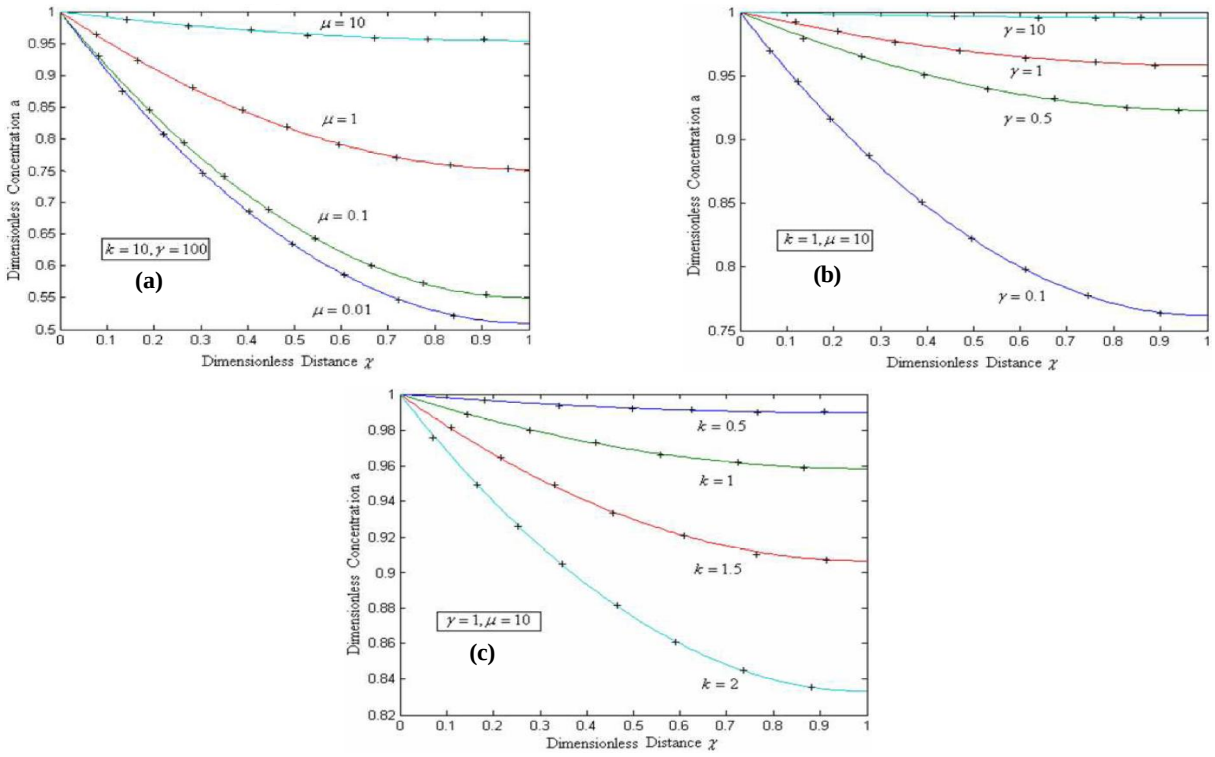

Figure 2 : Profile of the two-dimensional diagram of the normalized mediator concentration *a* versus the normalized distance χ when (a) k = 10, γ = 100 and a_s = 1 for various values of μ; (b) k = 1, μ = 10 and a_s = 1 for various values of γ; (c) γ = 1, μ = 10 and a_s = 1 for various values of k. The concentrations were computed using Eq. 17.

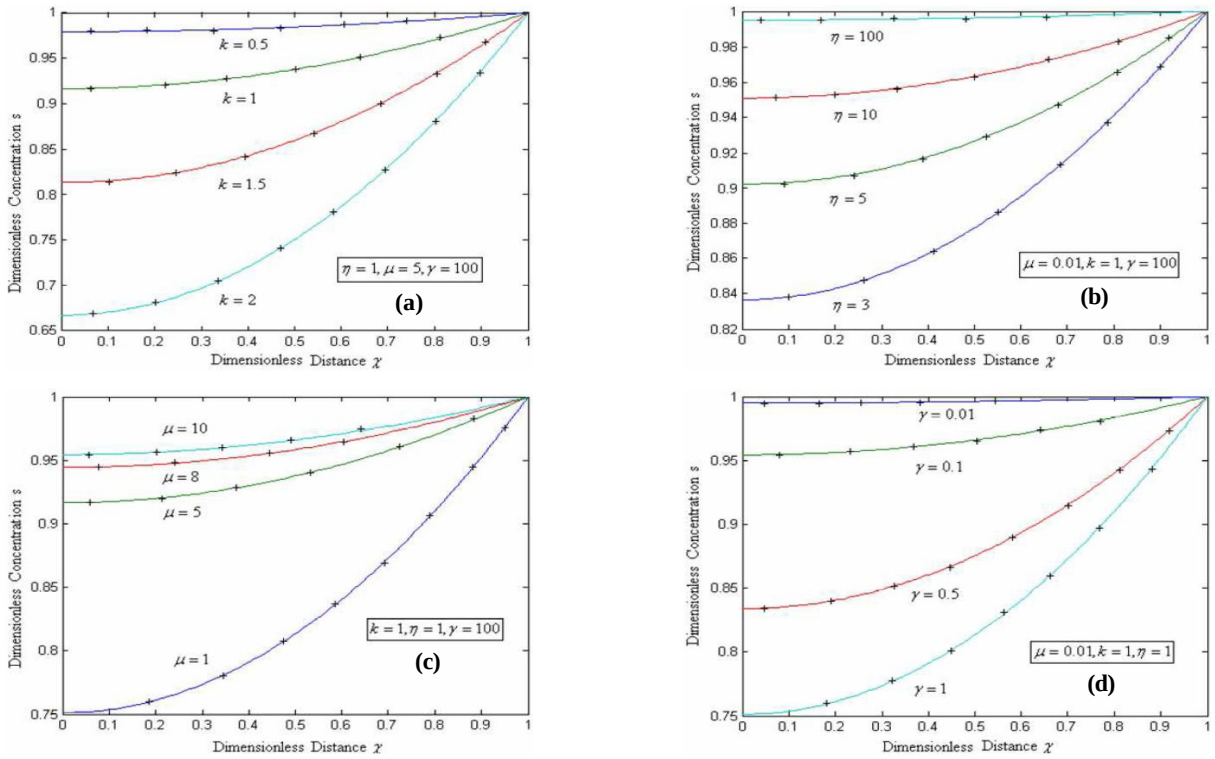

Figure 3 : Profile of the two-dimensional diagram of the normalized substrate concentration *s* versus the normalized distance χ when (a) η = 1, μ = 5, γ = 100 and a_s = 1 for various values of κ; (b) μ = 0.01, k = 1, γ = 100 and a_s = 1 for various values of η; (c) k = 1, η = 1, γ = 100 and a_s = 1 for various values of μ; (d) μ = 0.01, k = 1, η = 1 and a_s = 1 for various values of γ. The concentrations were computed using Eq. 18.

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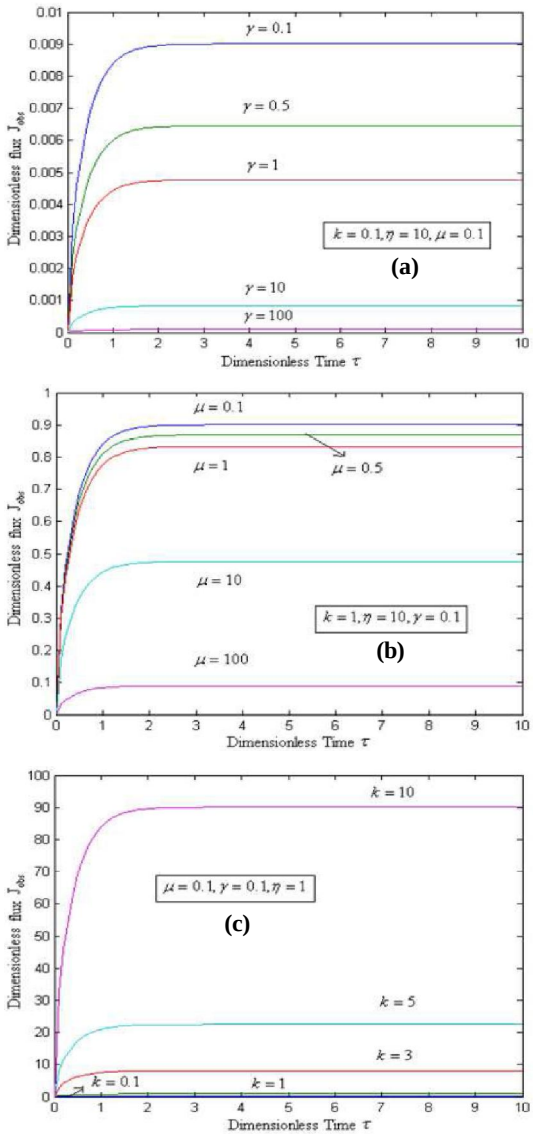

Figure 4 : Diagrammatic representation of the dimensionless flux against the dimensionless time τ when (a) k = 0.1, μ = 0.1 and η = 10 for various values of γ; (b) k = 1, γ = 0.1 and η = 1 for various values of μ; (c) μ = 0.1, γ = 0.1 and η = 1 for various values of k. The concentrations were computed using Eq. 19.

APPENDIX

Appendix A

Basic concepts of the HPM

The HPM method has overcome the limitations of traditional perturbation methods. It can take full advantage of the traditional perturbation techniques, so a considerable deal of research has been conducted to apply the homotopy technique to solve various strong non-linear equations. To explain this method, let us consider the following function:

$$\mathbf{D_o(u) - f(r) = 0, \qquad r \in \Omega} \tag{A1}$$

with the boundary conditions of

$$\mathbf{B_o\left(u, \frac{\partial u}{\partial n}\right) = 0, \qquad r \in \Gamma} \tag{A2}$$

where D_o is a general differential operator, B_o is a boundary operator, f(r) is a known analytical function and Γ is the boundary of the domain Ω. In general, the operator D_o can be divided into a linear part L and a nonlinear part N. Eq. A1 can therefore be written as

$$\mathbf{L(u) + N(u) - f(r) = 0} \tag{A3}$$

By the homotopy technique, we construct a homotopy $\nu(r, p) : \Omega \times [0,1] \rightarrow \Re$ that satisfies

$$\mathbf{H(\nu, p) = (1-p)[L(\nu) - L(u_0)] + p[D_0(\nu) - f(r)] = 0} \tag{A4}$$

$$\mathbf{H(\nu, p) = L(\nu) - L(u_0) + pL(u_0) + p[N(\nu) - f(r)] = 0} \tag{A5}$$

where $p \in [0, 1]$ is an embedding parameter, and u_0 is an initial approximation of Eq. A1 that satisfies the boundary conditions. From Eq. A4 and Eq. A5, we have

$$\mathbf{H(\nu,0) = L(\nu) - L(u_0) = 0} \tag{A6}$$

$$\mathbf{H(\nu,1) = D_0(\nu) - f(r) = 0} \tag{A7}$$

When p=0, Eq. A4 and Eq. A5 become linear equations. When p =1, they become non-linear equations. The process of changing p from zero to unity is that of $L(\nu) - L(u_0) = 0$ to $D_o(\nu) - f(r) = 0$. We first use the embedding parameter p as a "small parameter" and assume that the solutions of Eq. A4 and Eq. A5 can be written as a power series in p:

$$\mathbf{\nu = \nu_0 + p\nu_1 + p^2\nu_2 + \ldots} \tag{A8}$$

Setting p = 1 results in the approximate solution of Eq. A1:

$$\mathbf{v = \lim_{p \to 1} v = v_0 + v_1 + v_2 + \ldots} \tag{A9}$$

This is the basic idea of the HPM.

Appendix B

Approximate analytical solutions of the mediator and substrate

Using the HPM, we construct a homotopy for Eq. 11 and Eq. 12 as follows:

$$\mathbf{(1-p)\left(\frac{\partial a}{\partial \tau} - \frac{\partial^2 a}{\partial \chi^2}\right) + p\left(\frac{\partial a}{\partial \tau} - \frac{\partial^2 a}{\partial \chi^2} + \frac{\kappa^2 as}{[\gamma a(1+\mu s) + s]}\right) = 0} \tag{B1}$$

and

$$(1-p)\left(\frac{\partial s}{\partial \tau}-\frac{\partial^2 s}{\partial \chi^2}\right)+p\left(\frac{\partial s}{\partial \tau}-\frac{\partial^2 s}{\partial \chi^2}+\frac{\gamma\eta^{-1}\kappa^2 as}{[\gamma a(1+\mu s)+s]}\right)=0 \quad \textbf{(B2)}$$

The approximate solution of B1 is

$$a = a_0 + pa_1 + p^2a_2 + ... \quad \textbf{(B3)}$$

and the approximate solution of B2 is

$$s = s_0 + ps_1 + p^2s_2 + ... \quad \textbf{(B4)}$$

Substituting Eq. B3 into Eq. B1 and arranging the coefficients of *p* powers, we have

$$p^0 : \frac{\partial a_0}{\partial \tau}-\frac{\partial^2 a_0}{\partial \chi^2}=0 \quad \textbf{(B5)}$$

$$p^1 : \frac{\partial a_1}{\partial \tau}-\frac{\partial^2 a_1}{\partial \chi^2}-\frac{\kappa^2 a_0 s_0}{[\gamma a_0(1+\mu s_0)+s_0]}=0 \quad \textbf{(B6)}$$

Substituting Eq. B4 into Eq. B2 and arranging the coefficients of *p* powers, we have

$$p^0 : \frac{\partial s_0}{\partial \tau}-\frac{\partial^2 s_0}{\partial \chi^2}=0 \quad \textbf{(B7)}$$

$$p^1 : \frac{\partial s_1}{\partial \tau}-\frac{\partial^2 s_1}{\partial \chi^2}-\frac{\gamma\eta^{-1}\kappa^2 a_0 s_0}{[\gamma a_0(1+\mu s_0)+s_0]}=0 \quad \textbf{(B8)}$$

The initial approximations are as follows:

$$a_0(0) = a_\varepsilon,\ a_0'(1)=0,\ a_i'(1)=0 \text{ for all } i=1,2,3,\ldots \quad \textbf{(B9)}$$

$$s_0(1)=1,\ s_0'(0)=0,\ s_i'(0)=0 \text{ for all } i=1,2,3,\ldots \quad \textbf{(B10)}$$

From Eq. B5 we get

$$a_0 = \frac{a_\varepsilon}{s} \quad \textbf{(B11)}$$

From Eq. B7 we get

$$s_0 = \frac{1}{s} \quad \textbf{(B12)}$$

Substituting Eq. B11 and Eq. B12 into Eq. B6, we obtain the solution to Eq. B6:

$$a_1 = \frac{k^2 a_\varepsilon}{(\gamma a_\varepsilon(1+\mu)+1)}\left(\frac{\cosh(\sqrt{s(\chi-1)})}{s^2\cosh(\sqrt{s})}\right)-\frac{k^2 a_\varepsilon}{s^2[\gamma a_\varepsilon(1+\mu)+1]} \quad \textbf{(B13)}$$

Substituting Eq. B11 and Eq. B12 in Eq. B8 and then solving we get

$$s_1 = \frac{\gamma\eta^{-1}k^2 a_\varepsilon \cosh(\sqrt{s}\chi)}{s^2\cosh(\sqrt{s})[\gamma a_\varepsilon(1+\mu)+1]}-\frac{\gamma\eta^{-1}k^2 a_\varepsilon}{s^2[\gamma a_\varepsilon(1+\mu)+1]} \quad \textbf{(B14)}$$

Adding Eq. B11 and Eq. B13, we get Eq. 17 (the concentration of the mediator, *a*) in the text. Similarly, by adding Eq. B12 and Eq. B14 we get Eq. 18 (the concentration of the substrate, *s*) in the text.

Appendix C

```
function pdex4
m = 0;
x =linspace(0,1);
t =linspace(0,100);
sol = pdepe(m,@pdex4pde,@pdex4ic,@pdex4bc,x,t)
u1 = sol(:,:,1);
u2 = sol(:,:,2);
figure
plot(x,u1(end,:))
title('u1(x,t)')
figure
plot(x,u2(end,:))
title('u2(x,t)')
function [c,f,s] = pdex4pde(x,t,u,DuDx)
k = 1;
γ = 0.1;
μ = 0.01;
η = 1;
c = [1; 1];
f = [1; 1].* DuDx;
F1 = -(k^2*u(1)*u(2))/(γ*u(1)*(1+μ*u(2))+u(2));
F2 = -(γ*k^2*u(1)*u(2))/(η*(γ*u(1)*(1+μ*u(2))
+u(2)));
s = [F1; F2];
function u0 = pdex4ic(x);
u0 = [1; 1];
function [pl,ql,pr,qr] = pdex4bc(xl,ul,xr,ur,t)
pl = [ul(1)-1;0];
ql = [0;1];
pr = [0;ur(2)-1];
qr = [1;0];
```

CONCLUSIONS

This paper presents a mathematical treatment for analyzing amperometric enzymatic reactions. We have obtained a theoretical model describing the concentration of the mediator and the substrate. We have derived the flux which is described in terms of the dimensionless parameters κ, γ, η and μ. A simple closed form

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of analytical expressions of non steady -state substrate concentration profile to all possible values of the reaction/diffusion parameter κ, γ, η and μ are derived using Homotopy Perturbation Method. Furthermore, we have also presented an analytical expression for the flux in non steady state. This method is an extremely simple method and it is also a promising method to solve other non-linear equations. The solution procedure can be easily extended to all kinds of non-linear equations with various complex boundary conditions in enzyme-substrate reaction diffusion processes.

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