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Analytical expression for the steady state concentration of the species of an enzyme containing polymer modified electrode

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Abstract: In this article the mathematical analysis of non-linear differential equation in the action of an enzyme containing polymer modified electrode is discussed. The approximate analytical expressions of the steady state concentration and current of the species for all values of the dimensionless rate constants have been derived using the Homotopy perturbation method. Our analytical results are compared with the previous work and a satisfactory agreement is note. The present approach is less computational and is applicable for solving other strongly non-linear initial and boundary value problems in chemical and biological sciences.

Keywords: Polymer modified electrode; Bio sensor; Non-linear differential equation; Current; Homotopy analysis method.

I. INTRODUCTION

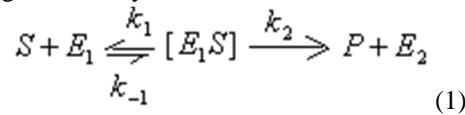
The first model for enzyme electrodes was proposed by Clark and Lyons [1]. The stable enzyme electrode for urea was reported by Guilbault and Montalvo [2]. The model provides a way to evaluate pertinent kinetic and diffusion properties of the system. Recently many techniques of enzyme immobilization have been developed. The biggest advantage of using immobilized enzymes is the reusability. Most enzymes are expensive, and routine analysis requires large amounts of these materials, which leads to the high cost. Immobilized enzymes can be reused, up to several thousand times, thus representing a tremendous cost saving. Immobilized enzymes are used at higher temperatures and they are known to be more stable [3]. The behavior of GO_x immobilized in poly (*N*-methylpyrrole) films and the immobilization of GO_x at platinum electrodes in electro polymerized films have been investigated by Bartlett and coworkers [4]. Mechanisms of enzyme entrapment and applications of the entrapped enzyme have been described by Bartlett and Cooper [5]. Dong and Che [6] obtained the current equation of the electro catalytic reaction at a microdisc electrode modified with redox species. Earlier, mathematical expressions pertaining to approximate analytical concentration and current for limiting cases at enzyme electrodes were calculated by Bartlett and Whitaker [7]. Recently, Meena and Rajendran [8] presented an analysis of system of coupled non-linear reaction diffusions within an electro active polymer film deposited on an inlaid microdisc electrode. Approximate analytical expression for substrate concentration for an amperometric biosensor at mixed enzyme kinetics have analyzed by Manimozhi et al. [9]. More recently, Senthamarai and Rajendran [10] have derived the approximate analytical expressions for the substrate, mediator concentrations and current response for the non-linear Michaelis-Menten kinetic scheme at conducting polymer-modified ultra-microelectrodes. The diffusion and kinetics in amperometric immobilized enzyme electrodes for reactions of the enzyme and substrate have reported by Loghambal and Rajendran [11].

Recently, Zhu Kan and Wu Hui-huang [12] have described a model for enzyme-entrapped conducting polymer modified electrodes and employ the model to investigate the steady-state current response of the enzyme electrode. However, to the best of our knowledge, to date, no analytical expressions corresponding to the substrate concentration and current for all values of parameters α and K at polymer modified electrode have been reported. The purpose of this communication is to derive an analytical expression for concentration and current of electrodes for all values of parameters using Homotopy perturbation method.

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II. MATHEMATICAL FORMULATION OF THE PROBLEM

The general reaction scheme for at reagentless enzyme electrodes



The corresponding governing equations in cartesian coordinates of the planar diffusion and reaction in the enzyme electrode is

$$D_s \frac{d^2 s}{dx^2} - R' = 0 \quad (3)$$

$$\text{when } R' = \frac{k_2 e_{\Sigma}}{1 + K_M/s + k_2/k_5} \quad (4)$$

Where D_s is the diffusion coefficient, s is the concentrations of species, k_2, k_5 reactions rate constants and, K_M is the Michaelis – Menten constant . The boundary conditions are

$$x = 0; \quad \frac{ds}{dx} = 0; \quad (5)$$

$$x = L; \quad D_s \left(\frac{ds}{dx} \right) = h_s (s_{\infty} - k_s s) \quad (6)$$

The current response is

$$I = nFD_s \left(\frac{ds}{dx} \right)_{x=L} \quad (7)$$

We make the above non-linear differential eqn.(3) in dimensionless form by defining the following dimensionless parameters

$$\alpha = \left(\frac{k_2 e_{\Sigma} L^2}{K_M D_s} \right)^{1/2}, \quad \beta_s = \frac{D_s}{h_s k_s L}, \quad K = \frac{(k_2 + k_5)}{K_M k_5} \quad (8)$$

The eqns. (2) and (3) reduces to the following dimensionless form:

$$\frac{d^2 s}{dx^2} - \frac{\alpha^2 s}{L^2 (1 + Ks)} = 0 \quad (9)$$

where s represents the dimensionless concentration. Now the boundary conditions become as

$$x = 0; \quad \frac{ds}{dx} = 0; \quad (10)$$

$$x = L; \quad \left(\frac{ds}{dx} \right) = \frac{(s_{\infty} - k_s s)}{\beta_s k_s L} \quad (11)$$

III. ANALYTICAL EXPRESSIONS OF THE CONCENTRATION AND CURRENT USING THE HOMOTOPY PERTURBATION METHOD

Linear and non-linear phenomena are of fundamental importance in various fields of science and engineering. Most models of real – life problems are still very difficult to solve. Therefore, approximate analytical solutions such as Homotopy perturbation method (HPM) were introduced [13-20]. This method is most effective and convenient ones for both linear and non-linear equations.

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Perturbation method is based on assuming a small parameter. Majority of non-linear problems, especially those having strong non-linearity, have no small parameters at all. Approximate solutions obtained by perturbation methods are valid only for small values of small parameter. Perturbation solutions are uniformly valid as long as scientific system parameter is small. We cannot rely fully on approximations, because there is no criterion on which the small parameter should exist. It is essential to check validity of approximations numerically and/or experimentally. To overcome these difficulties, HPM has been proposed recently.

Recently, many authors have applied the Homotopy perturbation method (HPM) to solve non-linear problem in physics and engineering sciences [13-16]. This method is used to solve non-linear problem in physical sciences [17-19]. This method is a combination of Homotopy in topology and classic perturbation techniques. Ji-Huan He solved Lighthill equation [17], the Duffing equation [18] and Blasius equation [19-20]. HPM is unique in applicability, accuracy and efficiency. HPM uses imbedding parameter as small parameter. Only a few iterations are needed to search for asymptotic solution. Using this Homotopy perturbation method (refer Appendix B), we obtain the normalized concentrations of the substrate as follows:

$$s(x) = \left\{ \begin{aligned} & \frac{s_{\infty} \cosh(\alpha x/L)}{k_s [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]} + \frac{K s_{\infty}^2}{2 k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^2} \\ & - \frac{K s_{\infty}^2 \cosh(\alpha x/L)}{2 k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^3} - \frac{k s_{\infty}^2 \cosh\left(\frac{2\alpha x}{L}\right)}{6 k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^2} \\ & + \frac{k s_{\infty}^2 \cosh\left(\frac{\alpha x}{L}\right) [\cosh(2\alpha) + 2\alpha \beta_s \sinh(2\alpha)]}{6 k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^3} \end{aligned} \right\} \quad (12)$$

The current expression is

$$I = n F D_s \left\{ \begin{aligned} & \frac{\alpha s_{\infty} \sinh(\alpha)}{L k_s [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]} - \frac{k \alpha s_{\infty}^2 \sinh(\alpha)}{2 L k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^3} \\ & + \frac{\alpha k s_{\infty}^2 \sinh(\alpha) [\cosh(2\alpha) + 2\alpha \beta_s \sinh(2\alpha)]}{6 L k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^3} \\ & - \frac{\alpha k s_{\infty}^2 \sinh(2\alpha)}{3 L k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^2} \end{aligned} \right\} \quad (13)$$

A. Unsaturated (Zero order) catalytic kinetics

When $K_s \ll 1$ eqn.(4) reduces to $R' = k_2 e_{\Sigma} / [1 + (k_2 / k_5)]$

In this case the concentration of species is:

$$s(x) = \frac{s_{\infty}}{k_s} - \frac{\alpha^2 (1/2 + \beta_s)}{k} + \frac{\alpha^2 x^2}{2 l^2 k} \quad (14)$$

The current expression is:

$$I = n F \int_0^L k_5 e_{\Sigma} dx = \frac{n F e_{\Sigma} L k_2 k_5}{k_2 + k_5} = n F D_s \left(\frac{\alpha^2}{L K} \right) \quad (15)$$

B. Saturated (First order) catalytic kinetics

When $K_s \gg 1$, the eqn.(4) reduces to $R' = (k_2 e_{\Sigma} s / K_M)$

In this case the concentration of species is:

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$$s(x) = \frac{s_{\infty} \cosh\left(\frac{\alpha x}{l}\right)}{k_s [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]} \quad (16)$$

The current expression is

$$I = nFD_s \left\{ \frac{\alpha s_{\infty} \tanh(\alpha)}{Lk_s [1 + \alpha \beta_s \tanh(\alpha)]} \right\} \quad (17)$$

IV. RESULTS AND DISCUSSION

The eqn.(12) represents the simple approximate analytical expression of the concentration of substrate s for all the parameter $\alpha, \beta_s, k_s, s_{\infty}$ and k . The concentration of substrate s using the eqn. (12) and compared with previous available analytical results using the eqn.(16) are represented in Figs.(2-11). In Figs.(2-6), the concentration of substrate reach the constant value for $\alpha = 0.1-1$ and for $\alpha > 1$, it decreases slowly and reaches the constant value for some fixed values $k = 0.1, \beta_s = 0.01$ to 10.

In Fig.(7), the concentration of substrate reach the constant for all values of β_s and for some fixed values of $k = 0.1, \alpha = 0.1$. In Figures (8-11), the concentration of substrate decreases slowly for $\beta = 0.1-5$ and reaches the constant value when $\beta_s > 5$ and for some fixed values of $k = 0.1, \alpha = 1$ to 100.

Fig. (12) shows the dimensionless current I (eqns. (13) and (17)) versus dimensionless parameter β_s . From this figure, it is apparent that the value of the current I decreases abruptly and reaches the constant value for small values of α and the current I becomes steady for large values of α . In Fig. (13), the value of the current I increases slowly and reaches the constant value for small values of β_s and the current I becomes steady for large values of β_s .

Fig.(14) shows the dimensionless current I (eqns.(13) and (15)) versus dimensionless parameter α . In this Figure, the value of the current I increases slowly and reaches the constant value for small values of L and the current I becomes steady for large values of L . Figs.(15-17) gives us the confirmation for the above discussion in 3 dimensional graphs also.

Table:1 represents the analytical result with Bartlett result of concentration of substrate s for $\alpha = 0.1, 0.2, 0.3$. The average relative error for s is 0.44%, 1.833% and 4.0778% when $\alpha = 0.1, 0.2, 0.3$.

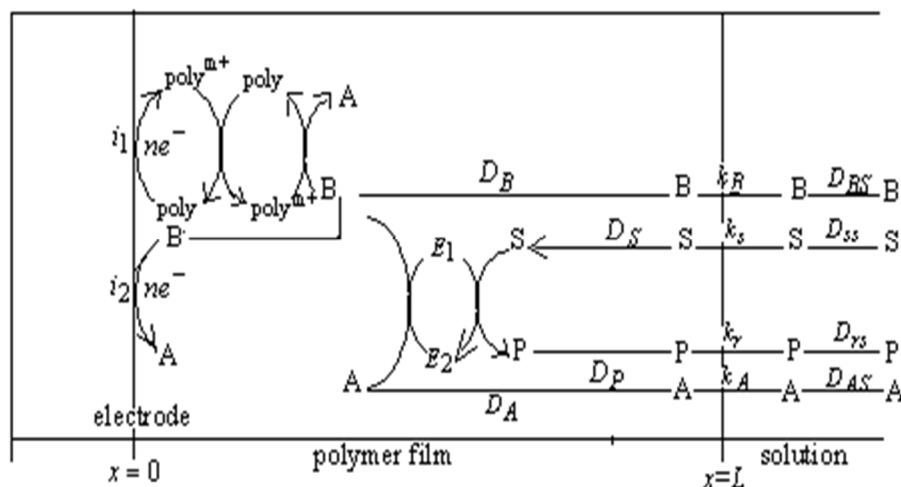


Fig. 1. The general kinetic scheme for the enzyme-entrapped conducting polymer modified electrode.

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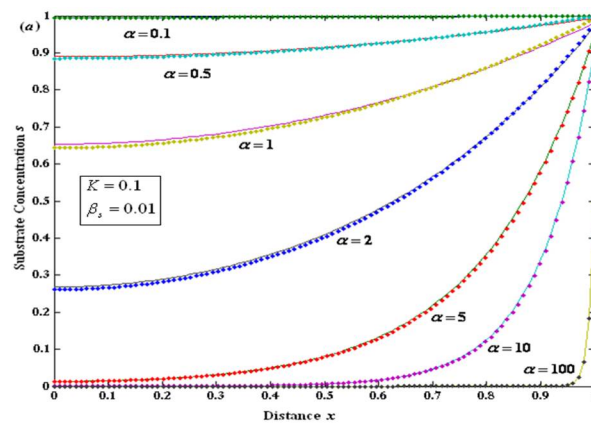


Fig. 2: The normalized concentration profiles for the substrate s . The curves are plotted using the eqnn.(12) for various values of reaction – diffusion parameter α when $k = 0.1, \beta_s = 0.01$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

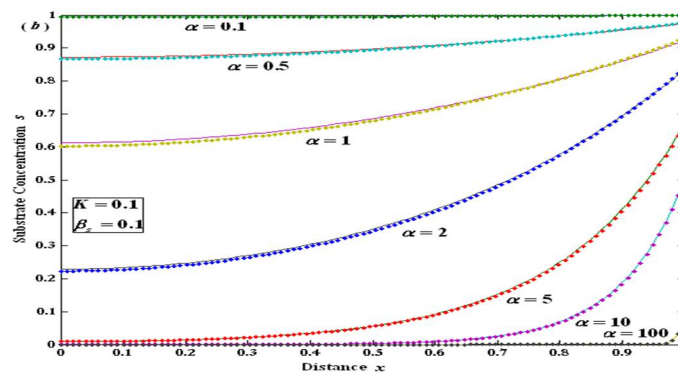


Fig. 3: The normalized concentration profiles for the substrate s . The curves are plotted using Eq.(3.12) for various values of reaction – diffusion parameter α when $k = 0.1, \beta_s = 0.1$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

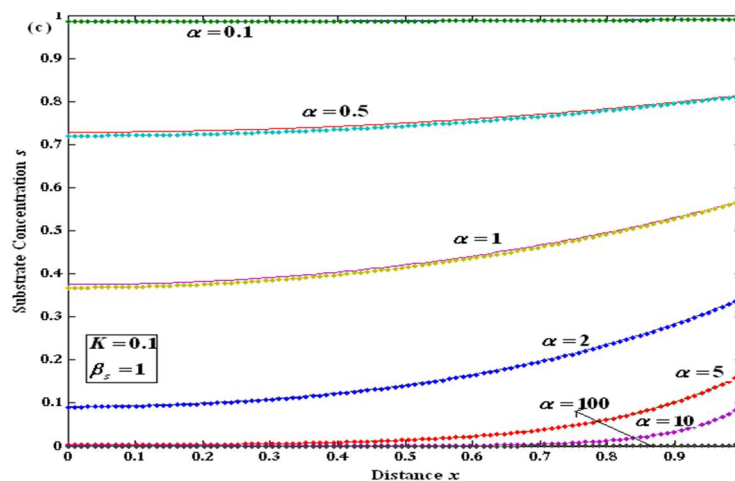


Fig. 4: The normalized concentration profiles for the substrate s . The curves are plotted using Eq. (3.12) for various values of reaction – diffusion parameter α when $k = 0.1, \beta_s = 1$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

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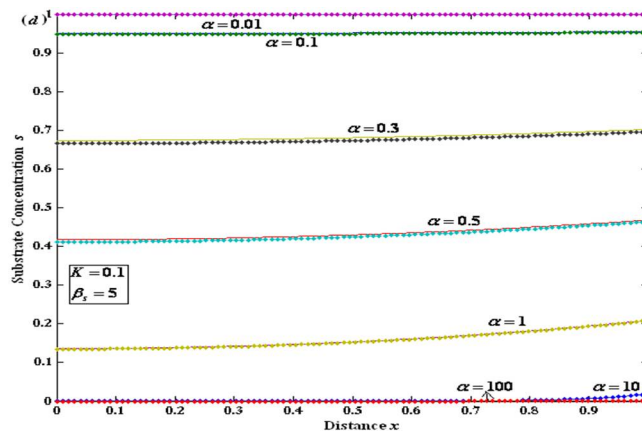


Fig. 5: The normalized concentration profiles for the substrate s . The curves are plotted using the eqn.(12) for various values of reaction – diffusion parameter α when $k = 0.1, \beta_s = 5$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

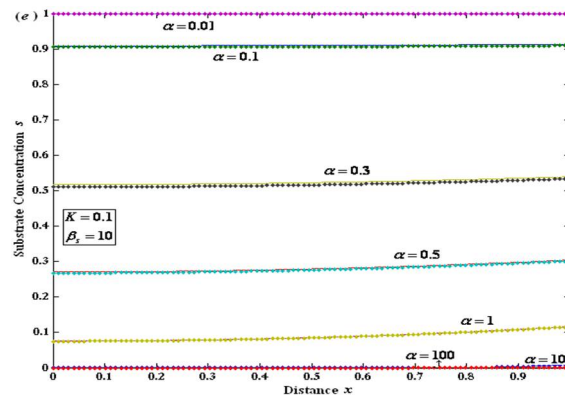


Fig. 6: The normalized concentration profiles for the substrate s . The curves are plotted using the eqn.(12) for various values of reaction – diffusion parameter α when $k = 0.1, \beta_s = 10$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

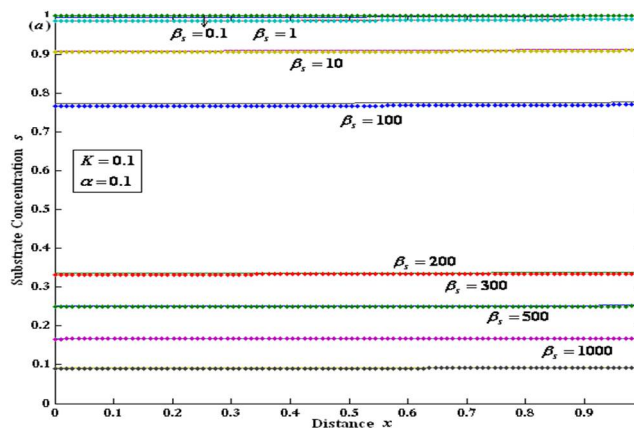


Fig. 7: The normalized concentration profiles for the substrate s . The curves are plotted using the eqn.(12) for various values of reaction – diffusion parameter α when $k = 0.1, \alpha = 0.1$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

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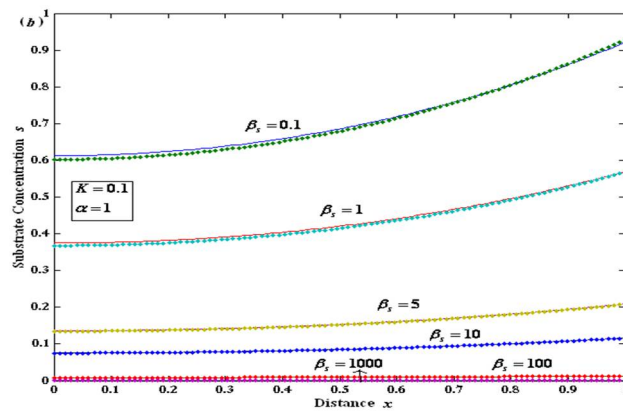


Fig. 8: The normalized concentration profiles for the substrate s . The curves are plotted using the eqn.(12) for various values of reaction – diffusion parameter α when $k = 0.1, \alpha = 1$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

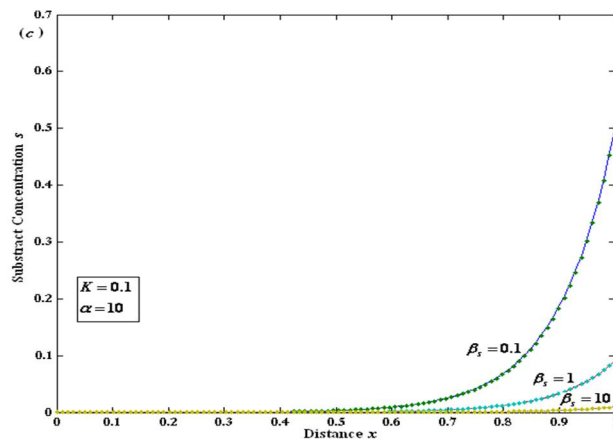


Fig. 9: The normalized concentration profiles for the substrate s . The curves are plotted using the eqn.(12) for various values of reaction – diffusion parameter α when $k = 0.1, \alpha = 10$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

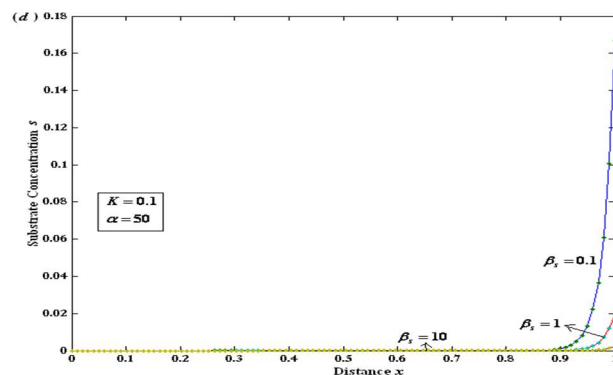


Fig. 10: The normalized concentration profiles for the substrate s . The curves are plotted using the eqn.(12) for various values of reaction – diffusion parameter α when $k = 0.1, \alpha = 50$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

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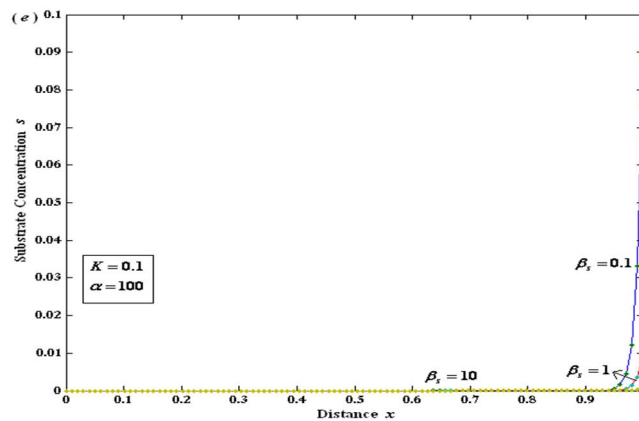


Fig. 11: The normalized concentration profiles for the substrate s . The curves are plotted using the eqn.(12) for various values of reaction – diffusion parameter α when $k = 0.1, \alpha = 100$. Symbols (-) denotes the present work (eqn.(12)) and (...) denotes the previous work (eqn.(16)).

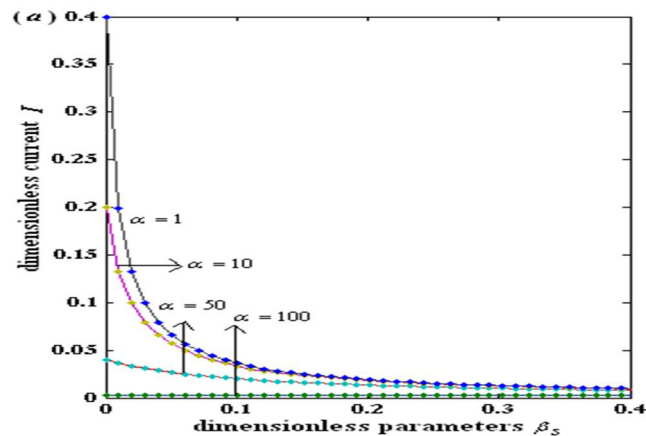


Fig. 12: Diagrammatic representation of current I vs dimensionless parameter β_s . The current were computed using the eqn.(13). The key to the graph (-) represents the present work (eqn. (13)) and (...) represents the the previous work (eqn. (17)).

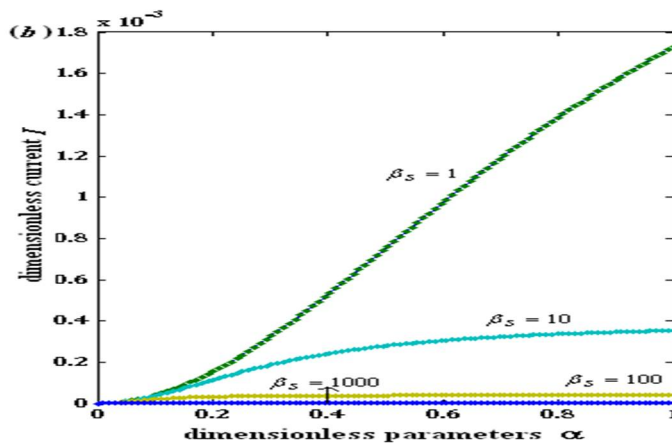


Fig. 13: Diagrammatic representation of current I versus the dimensionless parameter α . The current were computed using the eqn.(13). The key to the graph (-) represents the present work (eqn. (13)) and (...) represents the the previous work (eqn. (17)).

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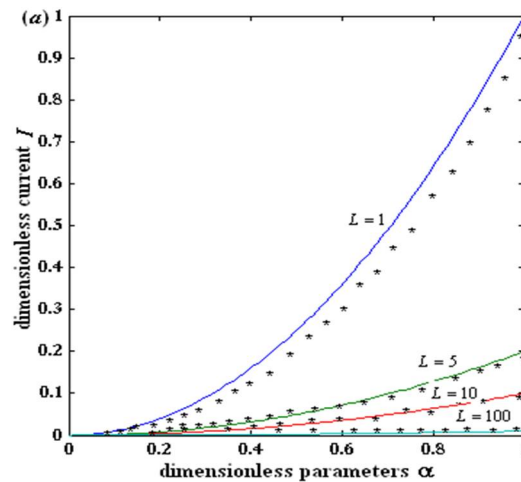


Fig. 14: Diagrammatic representation of current I versus dimensionless parameter α . The current were computed using the eqn. (13). The key to the graph (-) represents the eqn. (13) and (*) represents the eqn. (15).

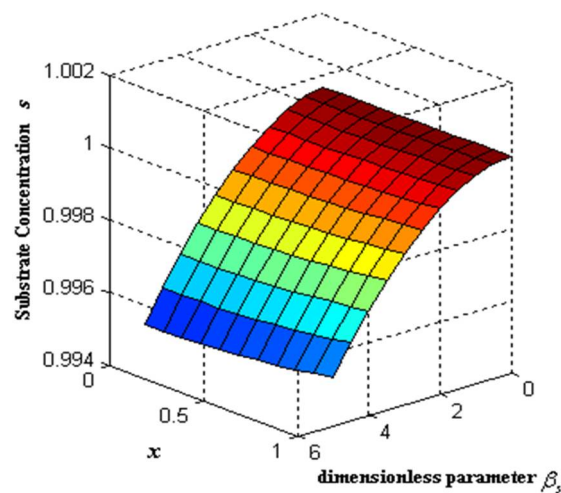


Fig.15: The normalized three-dimensional concentration of substrate s (eqn. (12)) for $x = 0$ to 1.

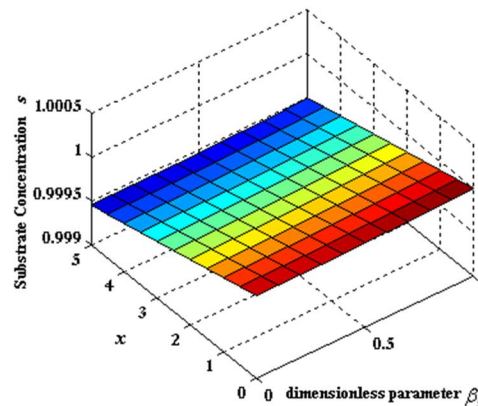


Fig. 16: The normalized three-dimensional concentration of substrate s (eqn. (12)) for $x = 0$ to 1.

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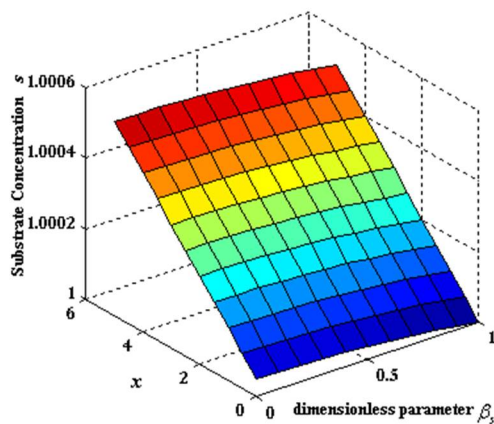


Fig. 17: The normalized three-dimensional concentration of substrate s (eqn. (12)) for $x = 0$ to 1 .

V. CONCLUSION

A non-linear differential equation in the action of an enzyme-containing polymer modified electrode has been solved using Homotopy perturbation method. In this first part of the paper, we have derived the steady-state analytical expression of the concentrations of the species for all values of rate constants. In the second part of the paper we have presented an approximate analytical expression corresponding to the species s in terms of the kinetic parameters k_2, k_5 , and K_M based on the Homotopy perturbation method for non steady-state. In addition, we have also presented an analytical expression for the non-steady state current. The kinetics of this electrochemical step can be principle be studied by observing how the limiting current responses strongly rely on the kinetics at the enzyme electrode including the rates of the enzymatic reactions.

VI. ACKNOWLEDGEMENTS

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Appendix A:

Basic concepts of the Homotopy perturbation method

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 [12]. To explain this method, let us consider the following function:

$$D_o(u) - f(r) = 0, \quad r \in \Omega \quad (\text{A.1})$$

with the boundary conditions of

$$B_o(u, \frac{\partial u}{\partial n}) = 0, \quad r \in \Gamma \quad (\text{A.2})$$

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 . Generally speaking, the operator D_o can be divided into a linear part L and a nonlinear part N . The eqn. (A.1) can therefore be written as

$$L(u) + N(u) - f(r) = 0 \quad (\text{A.3})$$

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

$$H(v, p) = (1-p)[L(v) - L(u_0)] + p[D_o(v) - f(r)] = 0. \quad (\text{A.4})$$

$$H(v, p) = L(v) - L(u_0) + pL(u_0) + p[N(v) - f(r)] = 0. \quad (\text{A.5})$$

where $p \in [0,1]$ is an embedding parameter, and u_0 is an initial approximation of the eqn.(A.1) that satisfies the boundary conditions. From the eqn. (A.4) and (A.5), we have

$$H(v, 0) = L(v) - L(u_0) = 0 \quad (\text{A.6})$$

$$H(v, 1) = D_o(v) - f(r) = 0. \quad (\text{A.7})$$

When $p=0$, the eqns.(A.4) and (A.5) become linear equations. When $p=1$, they become non-linear equations. The process of changing p from zero to unity is that of $L(v) - L(u_0) = 0$ to $D_o(v) - f(r) = 0$. We first use the embedding parameter p as a small parameter and assume that the solutions of the eqns. (A.4) and (A.5) can be written as a power series in p :

$$v = v_0 + pv_1 + p^2v_2 + \dots \quad (\text{A.8})$$

Setting $p = 1$ results in the approximate solution of the eqn.(A.1)

$$u = \lim_{p \rightarrow 1} v = v_0 + v_1 + v_2 + \dots \quad (\text{A.9})$$

This is the basic idea of the HPM.

Appendix B:

Solution of non-linear differential eqns.(9)-(11) using the Homotopy perturbation method

In this Appendix, we indicate how the eqns. (15) and (16) in this paper are derived. Furthermore, a Homotopy was constructed to determine the solution of the eqns. (9)-(11).

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$$(1-p) \left[\frac{d^2 s}{dx^2} - \frac{\alpha^2}{L^2} s \right] + p \left[\frac{d^2 s}{dx^2} + ks \frac{d^2 s}{dx^2} - \frac{\alpha^2}{L^2} s \right] = 0 \quad \text{and the initial} \quad (\text{B.1})$$

approximations are as follows:

$$x = 0, \quad \frac{ds}{dx} = 0; \quad (\text{B.2a})$$

$$x = L, \quad D_s \left(\frac{ds}{dx} \right) = h_s (s_\infty - k_s s); \quad (\text{B.2b})$$

$$x = 0, \quad \frac{ds_1}{dx} = 0; \quad (\text{B.2c})$$

$$x = L, \quad \frac{ds_1}{dx} = \frac{-s_1}{\beta_s L}; \quad (\text{B.2d})$$

The approximate solutions of the eqn. (9) is

$$s = s_0 + p s_1 + p^2 s_2 + p^3 s_3 + \dots \quad (\text{B.3})$$

Substituting the eqn. (B.3) into an eqn. (B.1) and comparing the coefficients of like powers of p

$$p^0 : \frac{d^2 s_0}{dx^2} - \frac{\alpha^2}{L^2} s_0 = 0 \quad (\text{B.4})$$

and

$$p^1 : \frac{d^2 s_1}{dx^2} - \frac{\alpha^2}{L^2} s_1 + ks_0 \frac{d^2 s_0}{dx^2} = 0 \quad (\text{B.5})$$

Solving the equations (B.4) and (B.5), and using the boundary conditions eqns.(B.2a)-(B.2d), we can obtain the following results:

$$s_0(x) = \frac{s_\infty \cosh\left(\frac{\alpha x}{L}\right)}{k_s [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]} \quad (\text{B.6})$$

and

$$s_1(x) = \left\{ \begin{aligned} & \frac{-ks_\infty^2 \cosh\left(\frac{\alpha x}{L}\right)}{2k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^3} + \frac{ks_\infty^2 \cosh(2\alpha) \cosh\left(\frac{\alpha x}{L}\right)}{6k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^3} \\ & + \frac{k\alpha \beta_s s_\infty^2 \sinh(2\alpha) \cosh\left(\frac{\alpha x}{L}\right)}{3k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^3} \\ & + \frac{ks_\infty^2}{2k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^2} - \frac{ks_\infty^2 \cosh\left(\frac{2\alpha x}{L}\right)}{6k_s^2 [\cosh(\alpha) + \alpha \beta_s \sinh(\alpha)]^2} \end{aligned} \right\} \quad (\text{B.7})$$

According to the HPM, we can conclude that

$$s(x) = \lim_{p \rightarrow 1} s(x) = s_0 + s_1 \quad (\text{B.8})$$

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Using the eqns. (B.6) and (B.7) in to the eqn. (B.8), we obtain the solution in the text eqn.(12).

Appendix C:

Nomenclature

Symbol	Meaning
s	Concentration of substrate (mM)
s_{∞}	Substrate concentration in bulk solution (mM)
x	Dimensionless distance
k_2, k_5	Rate constants ($M^{-1} s^{-1}$)
e_{Σ}	Total enzyme concentration in the film (mole cm^{-3})
D_s	Diffusion coefficient of the substrate ($\text{cm}^2 \text{sec}^{-1}$)
K_M	Michealis-Mental constant (mole cm^{-3})
α	Dimensionless parameter ($\text{mole}^{-1} \text{cm}^3$)
β	Dimensionless parameter ($\text{mole}^{-1} \text{cm}^3$)
L	Bulk
n	Number of electrons
F	Faraday constant ($C \text{mole}^{-1}$)
I	Current density at a enzyme electrode (ampere/cm^2)

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Table 1:

Comparison of normalized concentration of substrate s with Bartlett results for various values α and for fixed value of

$$k = 1, \beta = 0.1, k_s = 1, s_\infty = 1.$$

x	Concentration of substrate $s(x)$								
	s (when $\alpha = 0.1$)			s (when $\alpha = 0.2$)			s (when $\alpha = 0.3$)		
	This work eqn. (12)	Bartlett et. al eqn. (14) (previous work)	% of deviation	This work eqn. (12)	Bartlett et. al eqn. (14) (previous work)	% of deviation	This work eqn. (12)	Bartlett et. al eqn. (14)	% of deviation
0	0.9999	1.006	0.6100	0.9991	1.024	2.4922	0.9956	1.054	5.8658
0.2	0.9999	1.006	0.6100	0.9991	1.023	2.3921	0.9958	1.052	5.6437
0.4	0.9999	1.005	0.5100	0.9992	1.021	2.1817	0.9963	1.047	5.0888
0.6	1	1.004	0.4000	0.9994	1.017	1.7610	0.9971	1.038	4.1018
0.8	1	1.003	0.3000	0.9996	1.011	1.1404	0.9982	1.025	2.6848
1	1	1.001	0.1000	0.9999	1.004	0.4100	0.9982	1.009	1.0819
	Average % of deviation		0.4400	Average % of deviation		1.833	Average % of deviation		4.0778



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