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Homotopy perturbation method for kinetic analysis of thermal inactivation of jack bean urease

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Abstract

In this work, theoretical modeling and determination of molar concentration of the native and denatured jack bean urease (EC 3.5.1.5) are presented. A three-reaction kinetic model of thermal inactivation of urease is analyzed using homotopy perturbation method. The obtained analytical solutions are used to study the kinetics of thermal inactivation of the enzyme as applied in biotechnology. From the results, it is established that the molar concentration of native enzyme decreases as the time increases while the molar concentration of the denatured enzyme increases as the time increases. The time taken to reach the maximum value of the molar concentration of native enzyme is the same as the time taken to reach the minimum value of the molar concentration of the denature enzyme. The molar concentration of the denatured enzyme reaches the steady state value when reaction time is less than or equal to 5s. Also, the molar concentration of the denatured enzyme becomes zero when rate constant of dissociation reaction of the native form of the enzyme into a denatured form, is less than or equal to 0.01 s^{-1}. The analytical solutions are verified with numerical solutions using Runge–Kutta with shooting method and good agreements are established between the solutions. The information given in this theoretical investigation will assist in the kinetic analysis of the experimental results over handling rate constants and molar concentrations.

Keywords: Kinetics; Thermal activation; Jack bean urease; Enzyme; Homotopy perturbation method

1. Introduction

Functionally, urease (urea amidohydrolase E.C.3.5.1.5) is a part of the superfamily of amidohydrolases and phosphotriesterases. It is a highly efficient catalyst for the hydrolysis of urea into carbon dioxide and ammonia. It catalyzes at a rate approximately 10^{14} times faster than the rate of the non-catalyzed reaction [1–12]. The hydrolysis of urea is catalyzed by urease to produce ammonia and carbamate and the carbamate produced is subsequently degraded by spontaneous hydrolysis to produce another ammonia and carbonic acid. Urease activity tends to increase the pH of its environment as it produces ammonia.

Jack bean urease, which is the most widely used plant urease, is a nickel containing oligomeric enzyme exhibiting a high degree of specificity to urea [2]. The importance and the various applications of the urease as

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a good catalyst for hydrolysis of urea have attracted several research interests [2−21] especially in biotechnology and biomedical engineering studies. Also, the thermostability of jack bean urease has often been a subject of investigation [22−25]. However, there are few studies where the temporal loss of enzyme activity and the kinetic analysis of heat induced decay of enzyme activity were presented. Moreover, none of these studies involved consistent evaluation of kinetics of the urease inactivation. Most of the past studies described the complex mechanisms of thermal deactivation of enzymes as a "one step − two states" process where the native (active) form is transformed in the denaturated (inactive) form by a first order unimolecular irreversible reaction [24]. This unifying simplification is of interest for people focusing attention to phenomenological rather than mechanistic description of the kinetics of heat induced enzyme deactivation. However, the multitemperature evaluation revealed that an adequate kinetic model had to incorporate at least three reaction steps [24]. While the three-step mechanism model of inactivation of the enzyme has been developed by Illeova et al. [24], there is no provision of analytical solutions (except by Ananthi et al., [26]) for the predictions of model concentrations of the native enzyme, denature enzyme and temperature for thermal inactivation of urease. Ananthi et al., [26] applied homotopy analysis method to develop approximate analytical solutions for the analysis of kinetic and thermal inactivation of the enzyme. Although, the homotopy analysis method (HAM) is a reliable and efficient semi-analytical technique, but it suffers from a number of limiting assumptions such as the requirements that the solution ought to conform to the so-called rule of solution expression and the rule of coefficient ergodicity. Also, the use of HAM in the analysis of linear and nonlinear equations requires the determination of auxiliary parameter which will increase the computational cost and time. Also, the lack of rigorous theories or proper guidance for choosing initial approximation, auxiliary linear operators, auxiliary functions, and auxiliary parameters limit the applications of HAM. Moreover, such method requires high skill in mathematical analysis and the solution comes with large number of terms. In practice, analytical solutions with large number of terms and conditional statements for the solutions are not convenient for use by designers and engineers [26]. The determination of Adomian polynomials as carried out in Adomian decomposition method (ADM), the need for small perturbation parameter as required in traditional PMs, the rigor of the derivations of differential transformations or recursive relation as carried out in differential transformation method (DTM), the lack of rigorous theories or proper guidance for choosing initial approximation, auxiliary linear operators, auxiliary functions, auxiliary parameters, and the requirements of conformity of the solution to the rule of coefficient ergodicity as done in HAM, the search Langrange multiplier as carried out in variational iteration method (VIM), and the challenges associated with proper construction of the approximating functions for arbitrary domains or geometry of interest as in Galerkin weighted residual method (GWRM), least square method (LSM) and collocation method (CM) are some of the difficulties that are not experienced in HPM. Furthermore, in the class of the newly developed approximate analytical methods, homotopy perturbation method is considered to relatively simple with fewer requirements for mathematical rigor or skill. HPM solves differential equations, difference equation, differential-difference equations, fractional differential equation, pantograph equation and integro-differential equation. It solves nonlinear integral and differential equations without linearization, discretization, closure, restrictive assumptions, perturbation, approximations, round-off error and discretization that could result in massive numerical computations. It does not require small parameter in the algebraic or differential equation as done in the other traditional perturbation methods (Regular and singular perturbation). It provides excellent approximations to the solution of non-linear equation with high accuracy. Also, most of the above methods are limited to small domains. Applying the methods to large or infinite domain problems are often carried out with the applications of before-treatment techniques such as domain transformation techniques, domain truncation techniques and conversion of the boundary value problems to initial value problems or with the use of after-treatment techniques such as Pade-approximant, basis function, cosine after-treatment techniques, sine-after-treatment techniques and domain decomposition techniques. Indisputably, such additional computations through the before- and after-treatment techniques increase the computational cost and time. Furthermore, the search for a particular value that will satisfy second the boundary condition in DTM, HAM, ADM, and VIM necessitated the use of software and such could result in additional computational cost in the generation of solution to the problem. This drawback in the other approximation analytical methods is not experienced in HPM. HPM is a powerful method that gives acceptable analytical and accurate results with

convenient convergence and stability [26–31]. Therefore, in finding approximate analytical solutions to linear and nonlinear differential equations, HPM has fast gained ground as it appeared in many engineering and scientific research papers. Although, the improved HPM such as optimal homotopy asymptotic method (OHAM), optimal homotopy perturbation method (OHPM), homotopy analysis method (HAM), Optimal homotopy analysis method (OHAM) give higher accurate results than HPM but this comes with increased computational cost and time. Therefore, in this work, homotopy perturbation method is applied to the kinetics analysis of thermal inactivation of enzyme. The developed analytical solutions are used to study the effects of the models parameters on the molar concentration of the native and denatured enzyme.

2. Model formulation

The three − step mechanism of inactivation with a dissociation reaction of the native form of the enzyme, N, into a denatured form, D, and with two parallel association reactions of the native and denatured forms into irreversible denatured enzymes forms I_1 and I_2, respectively.

$$N \underset{k_{-1}}{\overset{k_{+1}}{\rightleftharpoons}} 2D \quad 2D \xrightarrow{k_2} I_1 \quad 2N \xrightarrow{k_3} I_2 \tag{1}$$

where k_{-1}, k_{+1}, k_2 and k_3 represent the rate constants of individual reactions. The material balances equations for N, D and temperature are given as follows [24,26]:

$$\frac{dc_N}{dt} = -k_{+1}c_N + k'_{-1}c_D^2 - 2k'_3c_N^2 \tag{2a}$$

$$\frac{dc_D}{dt} = 2k_{+1}c_N - 2\left(k'_{-1} + k'_2\right)c_D^2 \tag{2b}$$

$$\frac{dT}{dt} = K(T - T_B) \tag{2c}$$

Initial conditions are

$$t = 0,\ c_N = 1,\ c_D = 0,\ \ T_B - T = 30 \tag{3}$$

K is coefficient of enthalpy balance given as $4.44 \times 10^{-2}/s$ and TB is the bath temperature taken as $55–87.5\ ^{\circ}C$.

Eq. (2a and b) are the kinetic models which are formulated by the material balances of native form of the enzyme, N, and the denatured form, D. Eq. (2c) represents enthalpy balance equation which describes the initial heating period.

Let c_N, c_D, k_{+1}, k_{-1}, k_2 and k_3 by X,Y, a, b, c *and* d, respectively, Eq. (2a and b) become

$$\frac{dX}{dt} = -aX + bY^2 - 2dX^2 \tag{4a}$$

$$\frac{dY}{dt} = 2aX - 2(b + c)Y^2 \tag{4b}$$

$$t = 0,\ X = 1,\ Y = 0 \tag{5}$$

While the exact solution of Eq. (2c) is given as

$$T(t) = T_B - 30e^{-Kt}$$

3. Method of solution by homotopy perturbation method

It is very difficult to develop a closed-form solution for the above non-linear Eq. (19). Therefore, recourse has to be made to either approximation analytical method, semi-numerical method or numerical method of solution. In this work, homotopy perturbation method is used to solve the equation.

3.1. The basic idea of homotopy perturbation method

In order to establish the basic idea behind homotopy perturbation method as given by He [27–34], consider a system of nonlinear differential equations given as

$$A(U) - f(r) = 0, \quad r \in \Omega \tag{6}$$

With the boundary conditions

$$B\left(u, \frac{\partial u}{\partial \eta}\right) = 0, \quad r \in \Gamma \tag{7}$$

where A is a general differential operator, B is a boundary operator, $f(r)$ a known analytical function and Γ is the boundary of the domain Ω. It should be noted that the general differential operator A containing unknown function *f(r)* has the same spatial and temporal independent variables as known analytical function.

The operator A can be divided into two parts, which are L and N, where L is a linear operator, N is a nonlinear operator. Eq. (6) can be therefore rewritten as follows

$$L(u) + N(u) - f(r) = 0 \tag{8}$$

By the homotopy technique, a homotopy $U(r, p) : \Omega \times [0, 1] \rightarrow R$ can be constructed, which satisfies

$$H(U,p) = (1-p)[L(U) - L(U_o)] + p[A(U) - f(r)] = 0, \quad p \in [0,1] \tag{9}$$

Or

$$H(U,p) = L(U) - L(U_o) + pL(U_o) + p[N(U) - f(r)] = 0 \tag{10}$$

In the above Eqs. (9) and (10), $p \in [0,1]$ is an embedding parameter, u_o is an initial approximation of equation of Eq. (6), which satisfies the boundary conditions.

Also, from Eqs. (9) and (10), we will have

$$H(U,0) = L(U) - L(U_o) = 0 \tag{11}$$

$$H(U,0) = A(U) - f(r) = 0 \tag{12}$$

The changing process of p from zero to unity is just that of $U(r,p)$ from $u_o(r)$ to $u(r)$. This is referred to homotopy in topology. Using the embedding parameter p as a small parameter, the solution of Eqs. (9) and (10) can be assumed to be written as a power series in p as given in Eq. (13)

$$U = U_o + pU_1 + p^2U_2 + \ldots \tag{13}$$

It should be pointed out that of all the values of p between 0 and 1, $p = 1$ produces the best result. Therefore, setting $p = 1$, results in the approximation solution of Eq. (6)

$$u = \lim_{p \to 1} U = U_o + U_1 + U_2 + \ldots \tag{14}$$

The basic idea expressed above is a combination of homotopy and perturbation method. Hence, the method is called homotopy perturbation method (HPM), which has eliminated the limitations of the traditional perturbation methods. On the other hand, this technique can have full advantages of the traditional perturbation techniques. The series Eq. (14) is convergent for most cases.

3.2. Application of the homotopy perturbation method to the present problem

According to homotopy perturbation method (HPM), one can construct an homotopy for Eq. (4a and b) as

$$(1-p)\left[\frac{dX}{dt} - \frac{dx_0}{dt}\right] + p\left[\frac{dX}{dt} + aX - bY^2 + 2dX^2\right] = 0 \tag{15a}$$

$$(1-p)\left[\frac{dY}{dt} - \frac{dy_0}{dt}\right] + p\left[\frac{dY}{dt} - 2aX + 2(b+c)Y^2\right] = 0 \tag{15b}$$

The perturbation expressions of X and Y are

$$X = X_0 + pX_1 + p^2X_2 + p^3X_3 + p^4X_4 + p^5X_5 + p^6X_6 + p^7X_7 + p^8X_8 + p^9X_9 + p^{10}X_{10} + \ldots \tag{16a}$$

$$Y = Y_0 + pY_1 + p^2Y_2 + p^3Y_3 + p^4Y_4 + p^5Y_5 + p^6Y_6 + p^7Y_7 + p^8Y_8 + p^9Y_9 + p^{10}Y_{10} + \ldots \tag{16b}$$

Substituting Eq. (16a and b) into Eq. (15a and b) and the initial conditions in Eq. (5), we have

$$(1-p)\left[\frac{d(X_0 + pX_1 + p^2X_2 + p^3X_3 + p^4X_4 + p^5X_5 + p^6X_6 + p^7X_7 + p^8X_8 + p^9X_9 + p^{10}X_{10} + \ldots)}{dt} - \frac{dx_0}{dt}\right]$$
$$+p\left[\begin{array}{l} \dfrac{d(X_0 + pX_1 + p^2X_2 + p^3X_3 + p^4X_4 + p^5X_5 + p^6X_6 + p^7X_7 + p^8X_8 + p^9X_9 + p^{10}X_{10} + \ldots)}{dt} \\ +a(X_0 + pX_1 + p^2X_2 + p^3X_3 + p^4X_4 + p^5X_5 + p^6X_6 + p^7X_7 + p^8X_8 + p^9X_9 + p^{10}X_{10} + \ldots) \\ -b(Y_0 + pY_1 + p^2Y_2 + p^3Y_3 + p^4Y_4 + p^5Y_5 + p^6Y_6 + p^7Y_7 + p^8Y_8 + p^9Y_9 + p^{10}Y_{10} + \ldots)^2 \\ +2d(X_0 + pX_1 + p^2X_2 + p^3X_3 + p^4X_4 + p^5X_5 + p^6X_6 + p^7X_7 + p^8X_8 + p^9X_9 + p^{10}X_{10} + \ldots)^2 \end{array}\right] = 0 \tag{17a}$$

$$(1-p)\left[\frac{d(Y_0+pY_1+p^2Y_2+p^3Y_3+p^4Y_4+p^5Y_5+p^6Y_6+p^7Y_7+p^8Y_8+p^9Y_9+p^{10}Y_{10}+\ldots)}{dt}-\frac{dy_0}{dt}\right]$$
$$+p\left[\begin{array}{l}\dfrac{d(Y_0+pY_1+p^2Y_2+p^3Y_3+p^4Y_4+p^5Y_5+p^6Y_6+p^7Y_7+p^8Y_8+p^9Y_9+p^{10}Y_{10}+\ldots)}{dt}\\ -2a\left(X_0+pX_1+p^2X_2+p^3X_3+p^4X_4+p^5X_5+p^6X_6+p^7X_7+p^8X_8+p^9X_9+p^{10}X_{10}+\ldots\right)\\ +2(b+c)\left(Y_0+pY_1+p^2Y_2+p^3Y_3+p^4Y_4+p^5Y_5+p^6Y_6+p^7Y_7+p^8Y_8+p^9Y_9+p^{10}Y_{10}+\ldots\right)^2\end{array}\right]=0 \quad (17b)$$

The initial conditions are

$$X(0)=X_0(0)+pX_1(0)+p^2X_2(0)+p^3X_3(0)+p^4X_4(0)+p^5X_5(0)+p^6X_6(0)+p^7X_7(0)+p^8X_8(0)+p^9X_9(0)+p^{10}X_{10}(0)+\ldots=1 \quad (18a)$$

$$Y(0)=Y_0(0)+pY_1(0)+p^2Y_2(0)+p^3Y_3(0)+p^4Y_4(0)+p^5Y_5(0)+p^6Y_6(0)+p^7Y_7(0)+p^8Y_8(0)+p^9Y_9(0)+p^{10}Y_{10}(0)+\ldots=0 \quad (18b)$$

On expanding the Eq. (17a and b) and collecting all terms with the same order of p together, the resulting equation appears in form of polynomial in p. On equating each coefficient of the resulting polynomial in p to zero, we arrived at a set of differential equations and the corresponding initial conditions.

For X

$$p^0:\frac{dX_0}{dt}-\frac{dx_0}{dt}=0 \quad (19a)$$

$$p^1:\frac{dX_1}{dt}+aX_0-bY_0^2+2dX_0^2=0 \quad (19b)$$

$$p^2:\frac{dX_2}{dt}+aX_1-2bY_0Y_1+4dX_0X_1=0 \quad (19c)$$

$$p^3:\frac{dX_3}{dt}+aX_2-2bY_0Y_2-bY_1^2+4dX_0X_2+2dX_1^2=0 \quad (19d)$$

$$p^4:\frac{dX_4}{dt}+aX_3-2bY_1Y_2-2bY_0Y_3+4dX_0X_2+4dX_0X_3=0 \quad (19e)$$

$$p^5:\frac{dX_5}{dt}+aX_4-2bY_1Y_3-2bY_0Y_4-bY_2^2+4dX_0X_4+4dX_1X_3+2dX_2^2=0 \quad (19f)$$

$$p^6:\frac{dX_6}{dt}+aX_5-2bY_1Y_4-2bY_0Y_5-2bY_2Y_3+4dX_1X_4+4dX_0X_5+4dX_2X_3=0 \quad (19g)$$

$$p^7:\frac{dX_7}{dt}+aX_6-2bY_1Y_5-2bY_0Y_6-2bY_2Y_4-bY_3^2+4dX_1X_5+4dX_0X_6+4dX_2X_4+4dX_3^2=0 \quad (19h)$$

$$p^8:\frac{dX_8}{dt}+aX_7-2bY_1Y_6-2bY_0Y_7-2bY_2Y_5-2bY_3Y_4+4dX_1X_6+4dX_0X_7+4dX_2X_5+4dX_3X_4=0 \quad (19i)$$

$$p^9:\frac{dX_9}{dt}+aX_8-2bY_1Y_7-2bY_0Y_8-2bY_2Y_6-2bY_3Y_5-2bX_4^2+4dX_1X_7+4dX_0X_8+4dX_2X_6+4dX_3X_5+2dX_4^2=0 \quad (19j)$$

Initial conditions are

$$X_0(0)=1,\ X_1(0)=X_2(0)=X_3(0)=X_4(0)=X_5(0)=X_6(0)=X_7(0)=X_8(0)=X_9(0)=0 \quad (20)$$

For Y

$$p^0:\ \frac{dY_0}{dt}-\frac{dy_0}{dt}=0 \quad (21a)$$

$$p^1:\ \frac{dY_1}{dt}-2aX_0-2(b+c)Y_0^2=0 \quad (21b)$$

$$p^2:\ \frac{dY_2}{dt}-2aX_1-4(b+c)Y_0Y_1=0 \quad (21c)$$

$$p^3:\ \frac{dY_3}{dt}-2aX_2-4(b+c)Y_0Y_2-2(b+c)Y_1^2=0 \quad (21d)$$

$$p^4: \quad \frac{dY_4}{dt} - 2aX_3 - 4(b+c)Y_0Y_3 - 4(b+c)Y_1Y_2 = 0 \tag{21e}$$

$$p^5: \quad \frac{dY_5}{dt} - 2aX_4 - 4(b+c)Y_0Y_4 - 4(b+c)Y_1Y_3 - 2(b+c)Y_2^2 = 0 \tag{21f}$$

$$p^6: \quad \frac{dY_6}{dt} - 2aX_5 - 4(b+c)Y_0Y_5 - 4(b+c)Y_1Y_4 - 4(b+c)Y_2Y_3 = 0 \tag{21g}$$

$$p^7: \quad \frac{dY_7}{dt} - 2aX_6 - 4(b+c)Y_0Y_6 - 4(b+c)Y_1Y_5 - 4(b+c)Y_2Y_4 - 2(b+c)Y_2^2 = 0 \tag{21h}$$

$$p^8: \quad \frac{dY_8}{dt} - 2aX_7 - 4(b+c)Y_0Y_7 - 4(b+c)Y_1Y_6 - 4(b+c)Y_2Y_5 - 4(b+c)Y_3Y_4 = 0 \tag{21i}$$

$$p^9: \quad \frac{dY_9}{dt} - 2aX_8 - 4(b+c)Y_0Y_8 - 4(b+c)Y_1Y_7 - 4(b+c)Y_2Y_6 - 4(b+c)Y_3Y_5 - 2(b+c)Y_4^2 = 0 \tag{21j}$$

The initial conditions are

$$Y_0(0) = Y_1(0) = Y_2(0) = Y_3(0) = Y_4(0) = Y_5(0) = Y_6(0) = Y_7(0) = Y_8(0) = Y_9(0) = 0 \tag{22}$$

Solving the above Eqs. (19a–j) and (21a–j) with the initial conditions in Eqs. (20) and (22), we have

$$X_0 = x_0 = 1 \quad Y_0 = y_0 = 0$$

$$X_1 = -(a+2d)t, \quad Y_1 = 2at$$

$$X_2 = \frac{(a+2d)(a+4d)t^2}{2}, \quad Y_2 = -a(a+2d)t^2$$

$$X_3 = \frac{1}{6}\left(-a^3 + 8a^2b - 14a^2d - 4ad^2 - 48d^3\right)t^3,$$

$$Y_3 = \frac{a}{3}\left(a^2 - 8ab - 8ac + 6ad + 8d^2\right)t^3$$

$$X_4 = \frac{1}{24}\left(a^4 - 32a^3b + 30a^3d - 80a^2bd + 200a^2d^2 + 480ad^3 + 384d^4\right)t^4 \tag{17a}$$

$$Y_4 = -\frac{1}{12}a\left(a^3 - 32a^2b - 24a^2c + 14a^2d - 48abd - 48acd + 48ad^2 + 48d^3\right)t^4 \tag{18b}$$

$$X_5 = \frac{1}{120}\begin{pmatrix} -a^5 + 88a^4b - 256a^3b^2 - 256a^3bc - 62a^4d + 624a^3bd - 720a^3d^2 + 928a^2bd^2 \\ -312a^2d^3 - 576ad^4 - 384d^5 \end{pmatrix}t^5 \tag{19a}$$

$$Y_5 = \frac{1}{60}a\begin{pmatrix} a^4 - 88a^3b + 256a^2b^2 - 56a^3c + 512a^2bc + 256a^2c^2 + 30a^3d - 368a^2bd - 288a^2cd \\ +200a^2d^2 - 352abd^2 - 352acd^2 + 480ad^3 + 384d^4 \end{pmatrix}t^5 \tag{20b}$$

$$X_6 = \frac{1}{720}\begin{pmatrix} a^6 - 208a^5b + 217a^4b^2 + 1856a^4bc + 126a^5d - 313a^4bd + 422a^3b^2d + 422a^3bcd \\ 2408a^4d^2 - 11744a^3bd^2 + 16800a^3d^3 - 12672a^2bd^3 + 53760a^2d^4 + 8064ad^5 \\ +46080d^6 \end{pmatrix}t^6 \tag{21a}$$

$$Y_6 = -\frac{1}{360}a\begin{pmatrix} a^5 - 208a^4b + 2176a^3b^2 - 120a^4c + 3776a^3bc + 1600a^3c^2 + 62a^4d - 1824a^3bd \\ +3200a^2b^2d - 1200a^3cd + 6400a^2bcd + 3200a^2c^2d + 720a^3d^2 - 4448a^2bd^2 \\ -3520a^2cd^2 + 3120a^2d^3 - 3200abd^3 - 3200acd^3 + 5760ad^4 + 3840d^5 \end{pmatrix} t^6 \quad (22b)$$

$$X_7 = \frac{1}{5040}\begin{pmatrix} -a^7 + 456a^6b - 1152a^5b^2 + 17408a^4b^3 - 8704a^5bc + 34816a^4b^2c + 17408a^4bc^2 \\ -254a^6d + 12960a^5bd - 60416a^4b^2d - 50816a^4bcd - 7728a^5d^2 + 91648a^4bd^2 \\ -67840a^3b^2d^2 - 67840a^3bcd^2 - 81648a^4d^3 + 234752a^3bd^3 - 403200a^3d^4 \\ 199424a^2bd^4 - 1021440a^2d^5 - 1290240ad^6 - 645120d^7 \end{pmatrix} t^7 \quad (23a)$$

$$Y_7 = \frac{1}{2520}a\begin{pmatrix} a^6 - 456a^5b + 11520a^4b^2 - 17408a^3b^3 - 248a^5c + 18048a^4bc - 52224a^3b^2c \\ +6848a^4c^2 - 52224a^3bc^2 - 17408a^3c^3 + 126a^5d - 7456a^4bd + 43008a^3b^2d \\ -4320a^4cd + 76032a^3bcd + 33024a^3c^2d + 2408a^4d^2 - 34624a^3bd^2 + 38656a^2b^2d^2 \\ -22880a^3cd^2 + 77312a^2bcd^2 + 38656a^2c^2d^2 + 16800a^3d^3 - 60672a^2bd^3 \\ -48000a^2cd^3 + 53760a^2d^4 - 35072abd^4 - 35072acd^4 + 80640ad^5 + 46080d^6 \end{pmatrix} t^7 \quad (24b)$$

$$X_8 = \frac{1}{40320}\begin{pmatrix} a^8 - 960a^7b + 49152a^6b^2 - 25395a^5b^3 + 33792a^6bc - 457728a^5b^2c \\ -203776a^5bc^2 + 510a^7d - 48048a^6bd + 516864a^5b^2d - 442368a^4b^3d \\ +378624a^5bcd - 884736a^4b^2cd - 442368a^4bc^2d + 24200a^6d^2 - 570432a^5bd^2 \\ +1459968a^4b^2d^2 + 1217280a^4bcd^2 + 372960a^5d^3 - 260121a^4bd^3 + 1192448a^3b^2d^3 \\ +1192448a^3bcd^3 + 2669184a^4d^4 - 5064704a^3bd^4 + 10160640a^3d^5 - 3550208a^2bd^5 \\ +21288960a^2d^6 + 23224320ad^7 + 10321920d^8 \end{pmatrix} t^8 \quad (25a)$$

$$Y_8 = \frac{1}{20160}a\begin{pmatrix} a^7 - 960a^6b + 49152a^5b^2 - 253952a^4b^3 - 504a^6c + 7142a^5bc - 694272a^4b^2c \\ +25088a^5c^2 - 626688a^4bc^2 - 186368a^4c^3 + 254a^6d - 27408a^5bd + 352512a^4b^2d \\ -372736a^3b^3d - 14448a^5cd + 562432a^4bcd - 1118208a^3bcd + 219520a^4c^2d \\ -1118208a^3bc^2d - 372736a^3c^3d + 7728a^5d^2 - 214848a^4bd^2 + 759552a^3b^2d^2 \\ -123200a^4cd^2 + 1354496a^3bcd^2 + 594944a^3c^2d^2 + 81648a^4d^3 - 682752a^3bd^3 \\ +512512a^2b^2d^3 - 448000a^3cd^3 + 1025024a^2bcd^3 + 512512a^2cd^3 + 403200a^3d^4 \\ -935936a^3bd^4 - 736512a^2cd^4 + 1021440a^2d^5 - 451584abd^5 - 451584acd^5 \\ +1290240ad^6 + 645120d^7 \end{pmatrix} t^8 \quad (26b)$$

$$X_9 = \frac{1}{362880}\begin{pmatrix} -a^9 + 1976a^8b - 185856a^7b^2 + 2195456a^6b^3 - 2031616a^5b^4 - 118656a^7bc \\ +3634176a^6b^2c - 6094848a^5b^3c + 1477120a^6bc^2 - 6094848a^5b^2c^2 - 2031616a^5bc^3 \\ -1022a^8d + 166608a^7bd - 3449856a^6b^2d + 9785344a^5b^3d - 2258688a^6bcd \\ +17512448a^5b^2cd + 7727104a^5bc^2d - 74640a^7d^2 + 3114336a^6bd^2 \\ -18216192a^5b^2d^2 + 9615360a^4b^3d^2 - 13099264a^5bcd^2 + 19320720a^4b^2cd^2 \\ +9615360a^4bc^2d^2 - 1637040a^6d^3 + 22479360a^5bd^3 - 3589785a^4b^2d^3 \\ -29738496a^4bcd^3 - 16329600a^5d^4 + 75234816a^4bd^4 - 23255040a^3b^2d^4 \\ -23255040a^3bcd^4 - 87655680a^4d^5 + 118098944a^3bd^5 - 270950400a^3d^6 \\ +70469632a^2bd^6 - 483840000a^2d^7 - 46448640ad^8 - 185794560d^9 \end{pmatrix} t^9 \quad (27a)$$

$$Y_9 = \frac{1}{181440}a\begin{pmatrix} a^8 - 1976a^7b + 185856a^6b^2 - 2195456a^5b^3 + 2031616a^4b^4 - 1016a^7c + 255360a^6bc \\ -5575680a^5b^2c + 8126464a^4b^3c + 8486a^6c^2 - 4653568a^5bc^2 + 12189696a^4b^2c^2 \\ -1273344a^5c^3 + 8126464a^4bc^3 + 2031616a^4c^4 + 510a^7d - 94416a^6bd + 2282496a^5b^2d \\ -7753728a^4b^3d - 46368a^6cd + 3345408a^5bcd - 21420032a^4b^2cd + 1201152a^5c^2d \\ -19578880a^4bc^2d - 5912576a^4c^3d + 24200a^6d^2 - 1169184a^5bd^2 + 8904960a^4b^2d^2 \\ -6731776a^3b^3d^2 - 598752a^5cd^2 + 14323456a^4bcd^2 - 20195328a^3b^2cd^2 + 5661184a^4c^2d^2 \\ -20195328a^3bc^2d^2 - 6731776a^3c^3d^2 + 372960a^5d^3 - 5961216a^4bd^3 + 13920768a^3b^2d^3 \\ -3360000a^4cd^3 + 24920576a^3bcd^3 + 10999808a^3c^2d^3 + 2669184a^4d^4 - 14393856a^3bd^4 \\ +7606272a^2b^2d^4 - 9329152a^3cd^4 + 15212544a^2bcd^4 + 7606272a^2c^2d^4 + 10160640a^3d^5 \\ -16194560a^2bd^5 - 12644352a^2cd^5 + 21288960a^2d^6 - 6690816abd^6 - 6690816acd^6 \\ +23224320ad^7 + 10321920d^8 \end{pmatrix} t^9 \tag{28b}$$

$$X_{10} = \frac{1}{3628800}\begin{pmatrix} a^{10} - 4016a^9b + 652416a^8b^2 - 14729216a^7b^3 + 45285376a^6b^4 + 393024a^8bc \\ -22726656a^7b^2c + 125829120a^6b^3c - 8588800a^7bc^2 + 115802112a^6b^2c^2 \\ +35258368a^6bc^3 + 2046a^9d - 55299a^8bd + 19870080a^7b^2d - 125067264a^6b^3d \\ +74579968a^5b^4d + 1186137a^7bcd - 203614208a^6b^2cd + 223739904a^5b^3cd \\ -81081344a^6bc^2d + 223739904a^5b^2c^2d + 74579968a^5bc^3d + 228008a^8d^2 \\ -15631008a^7bd^2 + 173158656a^6b^2d^2 - 300335104a^5b^3d^2 + 110142208a^6bcd^2 \\ -534751232a^5b^2cd^2 - 234416128a^5b^2cd^2 - 234416128a^5bc^2d^2 + 6996000a^7d^3 \\ -167505024a^6bd^3 + 611667456a^5b^2d^3 - 213356544a^4b^3d^3 + 434254336a^5bcd^3 \\ -426713088a^4b^2cd^3 - 213356544a^4bc^2d^3 + 94744320a^6d^4 - 856747008a^5bd^4 \\ +930398208a^4b^2d^4 + 766973952a^4bcd^4 + 689230080a^5d^5 - 2258736128a^4bd^5 \\ +500871168a^3b^2d^5 + 500871168a^3bcd^5 + 2948520960a^4d^6 - 2968922112a^3bd^6 \\ +7664025600a^3d^7 - 1542111232a^2bd^7 + 11921817600a^2d^8 + 10218700800ad^9 \\ +3715891200d^{10} \end{pmatrix} t^{10} \tag{29a}$$

$$Y_{10} = -\frac{1}{1814400}a\begin{pmatrix} a^9 - 4016a^8b + 652416a^7b^2 - 14729216a^6b^3 + 45285376a^5b^4 - 2040a^8c + 859584a^7bc \\ -35260416a^6b^2c + 169082880a^5b^3c + 274368a^7c^2 - 27681280a^6bc^2 + 235536384a^5b^2c^2 \\ -7111680a^6c^3 + 144965632a^5bc^3 + 33226752a^5c^4 + 1022a^8d - 311808a^7bd \\ +12859776a^6b^2d - 94658560a^5b^3d + 6645350a^4b^4d - 145200a^7cd + 17574144a^6bcd \\ -243341312a^5b^2cd + 265814016a^4b^3cd + 5905536a^6c^2d - 206285824a^5bc^2d \\ +398721024a^4b^2c^2d - 57603072a^5c^3d + 265814016a^4bc^3d + 66453504a^4c^4d \\ +74640a^7d^2 - 5849376a^6bd^2 + 80992512a^5b^2d^2 - 190267392a^4b^3d^2 - 2735040a^6cd^2 \\ +118961920a^5bcd^2 - 528789504a^4b^2cd^2 + 43086336a^5c^2d^2 - 486776832a^4bc^2d^2 \\ -148254720a^4c^3d^2 + 1637040a^6d^3 - 44722560a^5bd^3 + 221326848a^4b^2d^3 \\ -122990592a^3b^3d^3 - 22243200a^5cd^3 + 356894208a^4bcd^3 - 368971776a^3b^2cd^3 \\ +141726720a^4c^2d^3 - 368971776a^3bc^2d^3 - 122990592a^3c^3d^3 + 16329600a^5d^4 \\ -168035328a^4bd^4 + 273917952a^3b^2d^4 - 92800512a^4cd^4 + 490967040a^3bcd^4 \\ +217049088a^3c^2d^4 + 87655680a^4d^5 - 326730752a^3bd^5 + 126173184a^2b^2d^5 \\ -208631808a^3cd^5 + 252346368a^2bcd^5 + 126173184a^2c^2d^5 + 270950400a^3d^6 \\ -311339008a^2bd^6 - 240869376a^2cd^6 + 483840000a^2d^7 - 112214016abd^7 \\ -112214016acd^7 + 464486400ad^8 + 185794560d^9 \end{pmatrix} t^{10} \tag{29b}$$

From the definition, the solutions in HPM domain are

$$X = X_0 + pX_1 + p^2X_2 + p^3X_3 + p^4X_4 + p^5X_5 + p^6X_6 + p^7X_7 + p^8X_8 + p^9X_9 + p^{10}X_{10} + \ldots \quad (30)$$

And

$$Y = Y_0 + pY_1 + p^2Y_2 + p^3Y_3 + p^4Y_4 + p^5Y_5 + p^6Y_6 + p^7Y_7 + p^8Y_8 + p^9Y_9 + p^{10}Y_{10} + \ldots \quad (31)$$

Setting $p = 1$, results in the approximation solutions of Eqs. (30) and (31) as

$$x = \lim_{p\to 1} X = X_0 + X_1 + X_2 + X_3 + X_4 + X_5 + X_6 + X_7 + X_8 + X_9 + X_{10} + \ldots \quad (32)$$

and

$$y = \lim_{p\to 1} Y = Y_0 + Y_1 + Y_2 + Y_3 + Y_4 + Y_5 + Y_6 + Y_7 + Y_8 + Y_9 + Y_{10} + \ldots \quad (33)$$

Hence, the solutions can therefore be written as

$$x = X_0 + X_1 + X_2 + X_3 + X_4 + X_5 + X_6 + X_7 + X_8 + X_9 + X_{10} + \ldots \quad (32)$$

and

$$y = Y_0 + Y_1 + Y_2 + Y_3 + Y_4 + Y_5 + Y_6 + Y_7 + Y_8 + Y_9 + Y_{10} + \ldots \quad (33)$$

Eq. (32) and (33) form the approximate analytical solutions of concentrations of native and denatured enzyme. The analytical solutions are simulated and the results are shown below.

4. Results and discussion

Tables 1 and 2 show the comparison between the results of HPM and NM. The obtained results of velocity distributions using HPM as compared with the numerical procedure using Runge-Kutta method coupled with shooting method are in good agreements. The high accuracy of HPM gives high confidence about validity of the method in providing solutions to the problem.

Table 1
Comparison of results.

The results of HPM and Numerical methods for *X(t)* for			
$a = 1$,	$b = 0.01$,	$c = 0.001$,	$d = 0.05$
X(t)			
X	HPM	NUM	Error of HPM
0.00	1.000000	1.000000	0.000000
0.10	0.896320	0.896320	0.000000
0.20	0.804239	0.804239	0.000000
0.30	0.722362	0.722362	0.000000
0.40	0.649479	0.649479	0.000000
0.50	0.584542	0.584542	0.000000
0.60	0.526638	0.526637	0.000001
0.70	0.474968	0.474965	0.000003
0.80	0.428836	0.428824	0.000012
0.90	0.387641	0.387599	0.000042
1.00	0.350878	0.350748	0.000130

Table 2
Comparison of results.

The results of HPM and Numerical methods for *X(t)* for			
$a = 1$,	$b = 0.01$,	$c = 0.001$,	$d = 0.05$
Y(t)			
X	HPM	NUM	Error of HPM
0.00	0.000000	0.000000	0.000000
0.10	0.189399	0.189399	0.000000
0.20	0.359101	0.359101	0.000000
0.30	0.511178	0.511178	0.000000
0.40	0.647477	0.647477	0.000000
0.50	0.769644	0.769644	0.000000
0.60	0.879150	0.879150	0.000000
0.70	0.977312	0.977311	0.000001
0.80	1.065300	1.065300	0.000000
0.90	1.144180	1.144170	0.000010
1.00	1.214880	1.214840	0.000040

Fig. 1 shows variation of the molar concentration of native and denatured enzyme with time when $k_{-1} = 1$, $k_{+1} = 0.01$, $k_2 = 0.001$, $k_3 = 0.05$. As depicted in the figure, the molar concentration of native enzyme decreases as the time increases while the molar concentration of the denatured enzyme increases as the time increases. The time taken to reach the maximum value of the molar concentration of native enzyme is the same as the time taken to reach the minimum value of the molar concentration of the denature enzyme. The steady values of molar concentrations of native and denatured enzyme depend upon the rate constants.

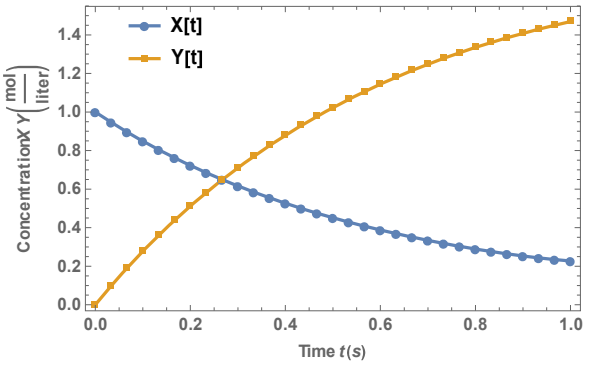

Fig. 1. Molar concentrations of native and denatured enzyme when $k_{-1} = 1, \quad k_{+1} = 0.01, \quad k_2 = 0.001, \quad k_3 = 0.05$.

Fig. 2 show the effects of dissociation native rate constant (k_{-1}) on molar concentration of denatured enzyme while Fig. 3 depict the effects of dissociation native rate constant (k_{-1}) on molar concentration of native enzyme when $k_{+1} = 0.01, \quad k_2 = 0.001, \quad k_3 = 0.001$.

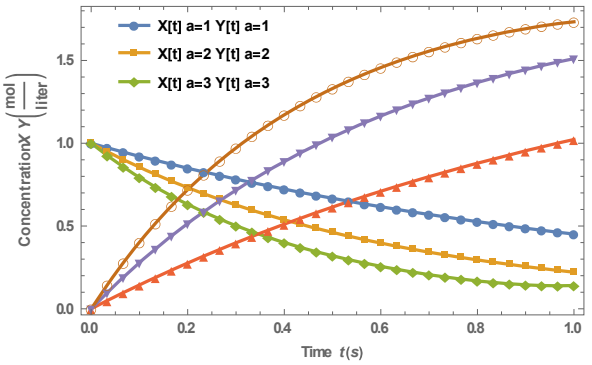

Fig. 2. Effects of dissociation native rate constant (k_{-1}) on molar concentration of denatured enzyme.

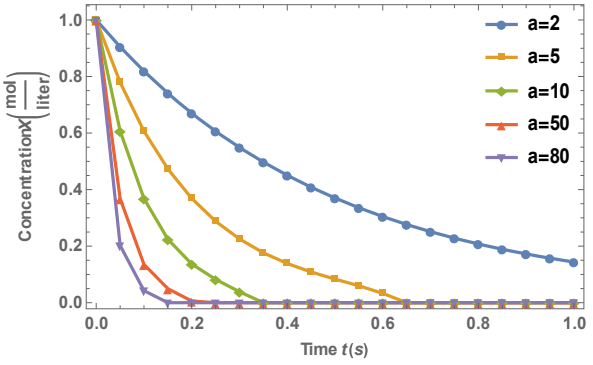

Fig. 3. Effects of dissociation native rate constant (k_{-1}) on mmolar concentration of native enzyme when $k_{+1} = 0.01, \quad k_2 = 0.001, \quad k_3 = 0.001$.

From these figures, it is found that, the value of molar concentration of the denatured enzyme initially increases and reaches the steady state value when $t \geq 5$. Also, the molar concentration of the denatured enzyme increases when 5 increases and the molar concentration becomes zero when $5_{+1} \leq 0.01\ s^{-1}$. Fig. 4 presents the effects of dissociation native rate constant (k_{+1}) on molar concentration of native enzyme when $k_{-1} = 0.88, \quad k_2 = 0.001, \quad k_3 = 0.00028$ while Fig. 5 shows the effects of dissociation native rate constant (k_{-1}) on molar concentration of denatured enzyme when $k_{+1} = 0.1, \quad k_2 = 0.00026, k_3 = 0.001$. Effects of dissociation native rate constant (k_{-1}) on molar concentration of native enzyme when $k_{-1} = 1, \quad k_2 = 0.1, \quad k_3 = 0.001$ are shown in Fig. 6.

Fig. 7 show the temperature history of the enzyme when $k_{-1} = 1, \ k_2 = 0.1, \quad k_3 = 0.001$. Also, effects of bath temperature on the temperature history are depicted in the figure. The temperature of the enzyme

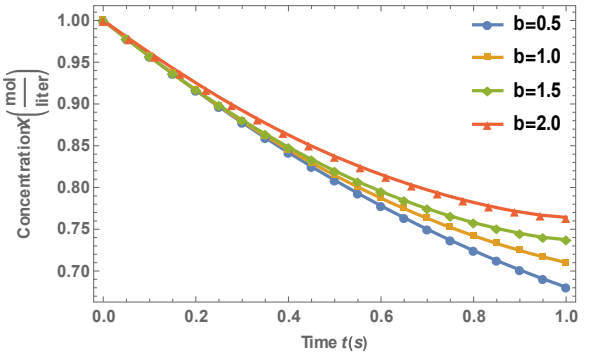

Fig. 4. Effects of dissociation native rate constant (k_{+1}) on molar concentration of native enzyme when $k_{-1} = 0.88, \quad k_2 = 0.001, \quad k_3 = 0.00028$.

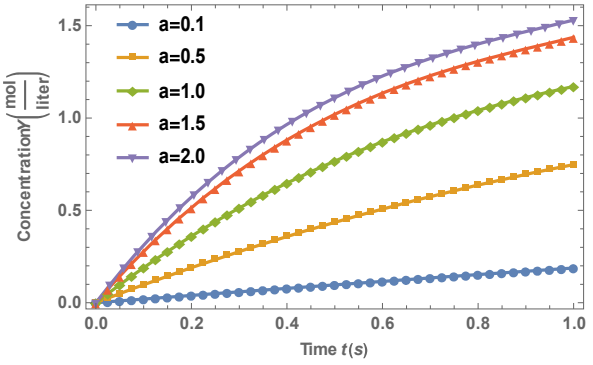

Fig. 5. Effects of dissociation native rate constant (k_{-1}) on molar concentration of denatured enzyme when $k_{+1} = 0.1, \quad k_2 = 0.00026, \quad k_3 = 0.001$.

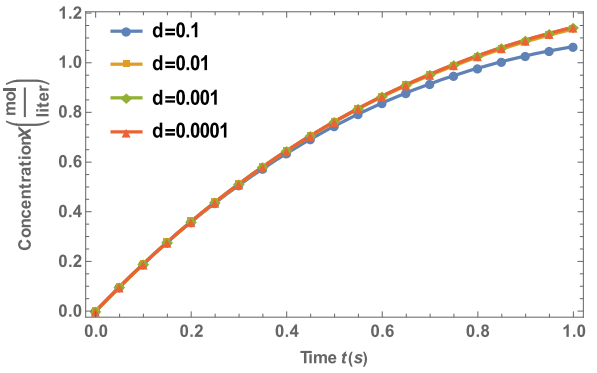

Fig. 6. Effects of dissociation native rate constant (k_{-1}) on molar concentration of native enzyme when $k_{-1} = 1$, $k_2 = 0.1$, $k_3 = 0.001$.

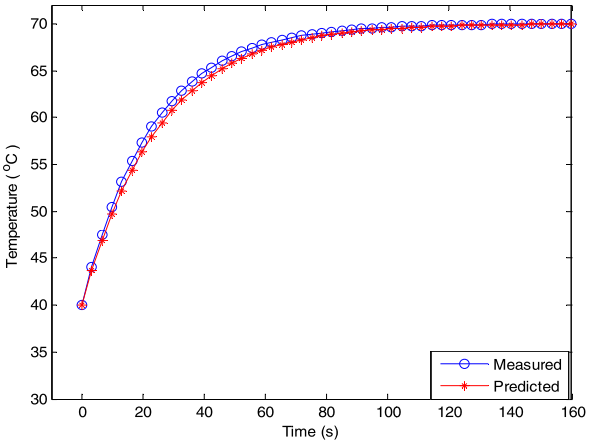

Fig. 8. Comparison between the experimental and model results at $T_B = 70$ °C.

decreases linearly with time. It could be seen that as the bath temperature, 5_5 increases, as the temperature of the enzyme increase.

Fig. 8 shows the comparison of the experimental data with the results predicted by HPM as applied in our work. Also, this result validates the earlier results in Fig. 1 of the earlier work carried out by Illeova et al. [24].

5. Conclusion

In this work, approximate analytical solutions for the analysis of kinetic model of thermal inactivation of the jack bean urease (E.C.3.5.1.5) have been developed using homotopy perturbation method. From the results, it was established that the molar concentration of native enzyme decreases as the time increases while the molar concentration of the denatured enzyme increases as the time increases. The time taken to reach the maximum value of the molar concentration of native enzyme is the same as the time taken to reach the minimum value of the molar concentration of the denature enzyme. The molar concentration of the denatured enzyme reaches the steady state value when reaction time is less than or equal to 5s. Also, the molar concentration of the denatured enzyme becomes zero when rate constant of dissociation reaction of the native form of the enzyme into a denatured form, is less than or equal to 0.01 s^{-1}. The analytical solutions were verified with numerical solution using

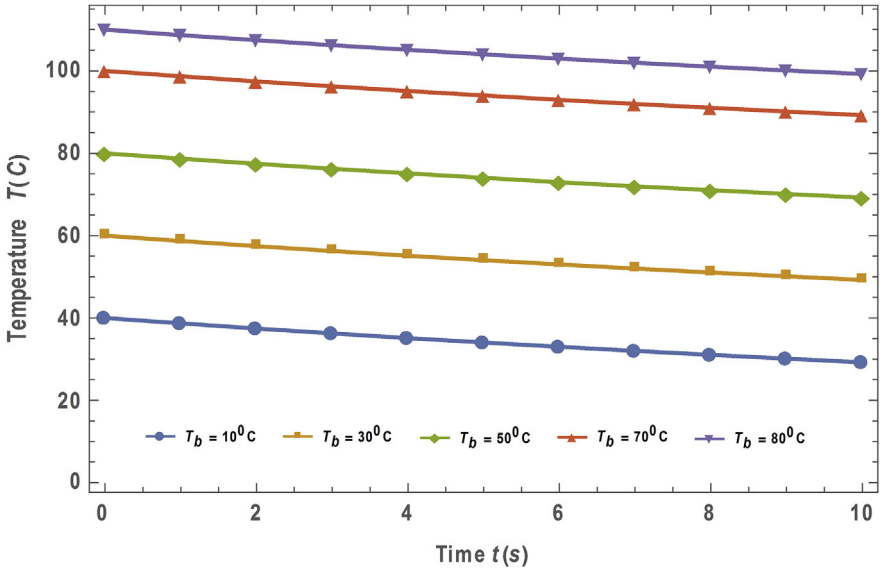

Fig. 7. Temperature variation with time of the enzyme when $k_{-1} = 1$, $k_2 = 0.1$, $k_3 = 0.001$.

Runge−Kutta with shooting method and good agreements were established. The information given in this theoretical investigation will assist in the kinetic analysis of the experimental results over handling rate constants and molar concentrations.

Nomenclature

c_N molar concentration of the native enzyme form (mole/cm)
c_D molar concentration of the denatured enzyme form (mole/cm)
k_{-1}, k_{+1}, k_2, k_3 rate constants of individual reaction (s^{-1})
k'_{-1}, k'_2, k'_3 modified rate constants (s^{-1})
K coefficient in the enthalpy balance (s)
T_B bath temperature (K)
T temperature (K)
t time (s)

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