

On the Estimation of the Coefficients in One Stochastic Model of an Enzymic Reaction

Besarion Dochviri*, Elizbar Nadaraya**, Grigol Sokhadze*,
Guram Tkemaladze*

* I. Javakhishvili Tbilisi State University

** Academy Member, I. Javakhishvili Tbilisi State University

(Presented by Academy Member Nugzar Aleksidze)

ABSTRACT. A stochastic model of an enzymic reaction is considered. Using statistical methods, formulas are derived for calculating the coefficients contained in this model. In particular, estimators of the parameters of the Michaelis-Menten equation are obtained. © 2011 Bull. Georg. Natl. Acad. Sci.

Key words: enzyme, mathematical model, statistical estimator, disorder, stochastic equation.

In the investigation of the enzymic reaction kinetics, an important role is played by various forms of a Michaelis-Menten type equation which establish the relation between the substrate concentration and the initial rate of an enzymic reaction. These equations also contain two unknown parameters - a maximal reaction rate and the so-called Michaelis-Menten constant [1-4]. These constants can be defined in each concrete case by means of the available heuristic and statistical methods [5, 6].

The classical Michaelis-Menten equation has the form

$$v = v(S) = \frac{V_{\max} S}{K + S}, \quad (1)$$

where v is the initial reaction rate, S is the substrate concentration, $V = V_{\max}$ is a maximal reaction rate, $K = K_M$ is the Michaelis-Menten constant for which $v(K) = 0.5V$.

We denote by $u(t, S)$ the optical density of the suspension at a moment of time t . Note that the values V and K are naturally time-dependent $V = V(t)$, $K = K(t)$. In other words, $u(t, S)$ describes the reaction course, i.e. the evolution of optical density in time. Using this notation, the

expression in the right-hand part of (1) will take the form

$$u'(0, S) = \left. \frac{\partial u(t, S)}{\partial t} \right|_{t=0},$$

which implies

$$u'(0, S) = \frac{V_{\max}(0) \cdot S}{K(0)} = V_{\max}(0) - \frac{K(0) \cdot V_{\max}(0)}{K(0) + S}.$$

Let us now iterate this function with respect to time. As a result of the first approximation we can write

$$u'(t, S) = V_{\max}(0) - \frac{K \cdot V_{\max}(0)}{K + S} + A(S)t,$$

where $A(S)$ is some non-negative function of S . If in this expression we take into account random deviations, which are produced by factors and which cannot be measured because of a great number of factors and the smallness of each individual factor, then by the central limit theorem we can write with good approximation

$$du(t, S) = \left(V_{\max}(0) - \frac{K \cdot V_{\max}(0)}{K + S} + tA(S) \right) \chi(t - \theta) dt + \delta dw(t). \quad (2)$$

In this formula $\delta > 0$, $w(t)$ is a standard Brownian motion for which $w(0) = Ew(t) = 0$, $Dw(t) = t$, θ is a random variable independent of $w(t)$, having values in $[0, \infty)$ and distributed by the law

$$P\{\theta = 0\} = \gamma, \quad P\{\theta > 0\} = e^{-\lambda t}, \quad t > 0, \quad (3)$$

where $0 < \lambda < \infty$ and $0 \leq \gamma \leq 1$ are the known parameters, $\chi(t) = \begin{cases} 1, & t \geq 0 \\ 0, & t < 0 \end{cases}$. The parameter λ in (3) depends

on mixture and on reaction temperature. This parameter can be assumed constant because for a concrete substance and the known temperature value the reaction duration is known.

The stochastic equation (2) describes the evolution of the suspension density in an enzymic reaction and the presence of the function $\chi(t)$ in it is due to the fact that in any reaction of such kind there sooner or later occurs a disorder of the reaction process. This depends on temperature and, to a lesser extent, on other factors. The unknown random parameter θ denotes the moment of time of the disorder onset and we have to estimate θ by means of observations of the process $u(t, S)$.

To the equation (2) we should add the initial conditions

$$u(0, S) = 0, \quad u(t, 0) = \alpha, \quad (4)$$

where α shows the reaction course in the absence of enzymes.

Assume that we observe the solution $u(t, S)$ of the problem (2) with the initial condition (4). It is required to estimate the parameters contained in this question at the moment of disorder.

In such a general formulation, we can pose various problems connected with the reaction process. In that case, the main tasks are the estimation of the parameters, the function $A(S)$ and the moment θ of the disorder onset. Mathematically, this is the so-called disorder problem which is solved using the sufficiently well-developed methods from the theory of random processes [7].

In this paper we will consider the problem of estimation of the parameters for the process

$$du(t, S) = \left(V_{\max} - \frac{K \cdot V_{\max}}{K + S} + rtS \right) dt + \delta dw(t) \quad (5)$$

with the initial conditions (4).

From (4)-(5) we write

$$u(t, S) = \left(V_{\max} - \frac{K \cdot V_{\max}}{K + S} \right) t + \frac{r}{2} t^2 S + \delta w(t).$$

Here $u(t, S)$ is a Gaussian Markov process for which

$$Eu(t, S) = \left(V_{\max} - \frac{K \cdot V_{\max}}{K + S} \right) t + \frac{r}{2} t^2 S, \\ Du(t, S) = \delta^2 t.$$

Therefore

$$\left. \frac{\partial Eu(t, S)}{\partial t} \right|_{t=0} = V_{\max} - \frac{K \cdot V_{\max}}{K + S} \quad \text{and} \quad \frac{\partial Du(t, S)}{\partial t} = \delta^2. \quad (6)$$

Hence it follows that for estimating the parameters V_{\max} and K it suffices to apply the least squares method provided that we have the observed data on the initial reaction rate dynamics depending on a concentration S (this also follows from the meaning of the parameters V_{\max} and K in the Michaelis-Menten equation).

Let us derive formulas for calculation of these parameters. Assume that we have n observations organized as shown in the following table:

Table 1

S	S_1	S_2	...	S_n
X	X_1	X_2	...	X_n

Here S_k denotes the concentrations and $X_k = v(S_k)$, $k=1, 2, \dots, n$, denotes the respective values of the initial reaction rate.

It is more convenient to apply the least squares method

to the inverse values $Y_k = \frac{1}{X_k}$, $f_k = \frac{1}{S_k}$. We write the

relation (1) in the Linuiver-Bark form

$$Y = af + b,$$

where $a = \frac{K}{V_{\max}}$, $b = \frac{1}{V_{\max}}$, $Y = \frac{1}{v} = \frac{1}{X}$ and $f = \frac{1}{S}$. Accordingly, our table will be rewritten as follows:

Table 2

f	$\frac{1}{S}$	f_1	f_2	...	f_n
Y	$\frac{1}{X}$	f_1	Y_2	...	Y_n

According to the principle of least squares, we are to find such values of the coefficients a and b that the expression

$$\sum_{k=1}^n (Y_k - af_k + b)^2$$

would take the smallest value. By standard analytical methods we find

$$\hat{a} = \frac{\frac{1}{n} \sum_{k=1}^n f_k Y_k - \bar{f} \cdot \bar{Y}}{\frac{1}{n} \sum_{k=1}^n f_k^2 - \bar{f}^2} \quad \text{and}$$

$$\hat{b} = \frac{\bar{Y} - \frac{1}{n} \sum_{k=1}^n f_k^2 - \bar{f} \cdot \frac{1}{n} \sum_{k=1}^n f_k Y_k}{\frac{1}{n} \sum_{k=1}^n f_k^2 - \bar{f}^2},$$

where $\bar{f} = \frac{1}{n} \sum_{k=1}^n f_k$ and $\bar{Y} = \frac{1}{n} \sum_{k=1}^n Y_k$. Returning to the initial variable, we can write

$$\hat{V}_{\max} = \frac{n \sum_{k=1}^n \frac{1}{S_k^2} - \left(\sum_{k=1}^n \frac{1}{S_k} \right)^2}{\sum_{k=1}^n \frac{1}{X_k} \sum_{k=1}^n \frac{1}{S_k^2} - \sum_{k=1}^n \frac{1}{S_k} \sum_{k=1}^n \frac{1}{X_k S_k}},$$

$$\hat{K} = \frac{n \sum_{k=1}^n \frac{1}{X_k S_k} - \sum_{k=1}^n \frac{1}{S_k} \sum_{k=1}^n \frac{1}{X_k}}{\sum_{k=1}^n \frac{1}{X_k} \sum_{k=1}^n \frac{1}{S_k^2} - \sum_{k=1}^n \frac{1}{X_k S_k}}.$$

As seen from the formula (6), the coefficient δ^2 is the dispersion of the process $\frac{\partial u(t, S)}{\partial t}$ and does not depend on t . Therefore it can be estimated from the above tables since this is the dispersion of the initial reaction rate. Thus we can write the estimate

$$\hat{\delta}^2 = \frac{1}{n} \sum_{k=1}^n \left(X_k - \frac{\hat{V}_{\max} \cdot \hat{K}}{\hat{K} + S_k} \right)^2.$$

It remains to indicate the estimation procedure for the coefficients c and r . For this we note that for an average rate of the enzymic reaction we have the relation

$$\frac{\partial Eu(t, S)}{\partial t} = V_{\max} - \frac{K \cdot V_{\max}}{K + S} + rtS = \frac{S \cdot V_{\max}}{K + S} + ct + rtS.$$

Hence it follows that we can use the table of observation data which relate the reaction rate to the concentrations S and time t .

So, we have

Table 3

$t \backslash S$	S_1	S_2	...	S_m
t_1	v_{11}	v_{12}	...	v_{1m}
t_2	v_{21}	v_{22}	...	v_{2m}
\vdots	\vdots	\vdots	...	\vdots
t_n	v_{n1}	v_{n2}	...	v_{nm}

Using the experimental results (given in Table 3) and the formula (5), we compose the sum

$$\sum_{i=1}^n \sum_{j=1}^m \left(v_{ij} - \frac{S_j \cdot \hat{V}_{\max}}{\hat{K} + S_j} - rt_i S_j \right)^2 \quad (7)$$

and choose the parameters c and r so that the function (7) would have a minimal value. Applying again the standard optimization methods, we find

$$\hat{r} = \frac{\sum_{i=1}^n t_i a_i - \hat{V}_{\max} bc}{\Delta},$$

where

$$a_i = \sum_{j=1}^m S_j v_{ij}, \quad b = \sum_{i=1}^n t_i, \quad c = \sum_{j=1}^m \frac{S_j^2}{\hat{K} + S_j}.$$

$$\Delta = \sum_{i=1}^n t_i^2 \sum_{j=1}^m S_j^2.$$

The obtained formulas for estimation of the parameters of the equation (5) can be used in the analysis of enzymic reactions as well as in practical computations.

Acknowledgement. This work is supported by the Georgia National Science Foundation (Project # GNSF/ST09_383_3-106).

ბიოქიმია

ფერმენტაციული რეაქციის ერთი სტოქასტური მოდელის კოეფიციენტების შეფასების შესახებ

ბ. დოჭვირი*, ე. ნადარაია**, გ. სოხაძე*, გ. ტყემალაძე*

* ი. ჯავახიშვილის სახ. თბილისის სახელმწიფო უნივერსიტეტი

** აკადემიის წევრი, ი. ჯავახიშვილის სახ. თბილისის სახელმწიფო უნივერსიტეტი

(წარმოდგენილია აკადემიის წევრის ნ. ალექსიძის მიერ)

ნაშრომში განხილულია ფერმენტაციული რეაქციის სტოქასტური მოდელი. სტატისტიკური მეთოდების გამოყენებით მიღებულია მოდელში შემავალი კოეფიციენტების გამოსათვლელი ფორმულები. კერძოდ, მოცემულია მიხაელის-მენტენის განტოლების პარამეტრების შეფასების ფორმულები.

REFERENCES

1. *L. Michaelis, M.L. Menten* (1913), *Biochem. Z.*, 49: 333.
2. *G.E. Briggs, J.B.S. Haldane* (1925), *Biochem. J.*, 19: 338.
3. *E.A. Dawes* (1980), *Quantitative problems in biochemistry*. Longman, London-New York.
4. *A. Lehninger* (1983), *Osnovy biokhimii* (v 3 tomakh), M. (in Russian)
5. *B. Dochviri, G. Sokhadze, G. Tkemaladze* (2010), *Bulletin of St. Andrew the First-Called Georgian University of the Patriarchy of Georgia*, 2.
6. *A. B. Dochviri, G. Sokhadze, G. Tkemaladze* (2010), *Bulletin of St. Andrew the First-Called Georgian University of the Patriarchy of Georgia*, 2.
7. *Shiryayev* (1978), *Optimal stopping rules*. Springer-Verlag, New York–Heidelberg.

Received December, 2010