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THE CONSTRUCTION AND STUDY OF THE SYSTEM OF DIFFERENTIAL EQUATIONS THAT DESCRIBES BIOCHEMICAL PROCESSES RATES

Губаль Г.М. Побудова та дослідження системи диференціальних рівнянь, яка описує швидкості біохімічних процесів. У статті зроблено математичний аналіз деяких ферментативних реакцій. Досліджено швидкості біохімічних процесів.
Ключові слова: система диференціальних рівнянь, швидкість біохімічних процесів, субстрат, фермент, інгібітор.

Губаль Г.Н. Построение и исследование системы дифференциальных уравнений, описывающей скорости биохимических процессов. В статье сделано математический анализ некоторых ферментативных реакций. Исследовано скорости биохимических процессов.
Ключевые слова: система дифференциальных уравнений, скорость биохимических процессов, субстрат, фермент, ингибитор.

Hubal H.M. The construction and study of the system of differential equations that describes biochemical processes rates. Mathematical analysis of some enzymatic reactions is made in this article. Biochemical processes rates are studied.
Keywords: system of differential equations, biochemical processes rate, substratum, enzyme, inhibitor.
Bibl. 3.

Introduction. While studying rates of various biological and biochemical processes, it is important to describe a complex system using a small amount of differential equations [1] that represents one of the main tasks of mathematical modeling. General laws for rates are the same for both chemical reactions and biochemical reactions and reproduction of individuals of coexisting species.

Problems of studying reaction rates of various substances are rather difficult and their solution requires application of many sciences including quantum mechanics.

One of the main rules of kinetics is this: in order to interact, one needs to meet. In chemical reactions, it is necessary that two molecules and atoms meet; in the problem of coexistence of species, it is necessary that, for example, a lynx and a hare meet (this accompanying by an absorption reaction). However, meetings are usually random and it is impossible to foresee each of them. Therefore, we consider the processes that consist of many meetings in which we can talk about the probability of a meeting and the average number of reactive objects in a reaction, i.e. about the concentration of objects. In the case of chemical reactions, they are just usual concentrations; in the case of interaction of microorganisms with each other or with molecules of nutrient, by concentrations we mean the number of microorganisms (or the amount of organic substance contained in them) per unit volume. In ecology, for example, in coexistence of species, concentration is the number of individuals per unit area.

Main part. Consider at what rate the reaction of interaction between two molecules M and N of different substances goes. This reaction (the second-order reaction) can result in the creation of the complex molecule P or two new molecules P_1 and P_2:

$$M + N \to P \quad \text{or} \quad M + N \to P_1 + P_2.$$

The reaction rate depends primarily on the probability of collision of the molecules M and N. The probability of collision is proportional to the product of concentrations C_M and C_N. The reaction rate of their interaction

$$v_{MN} = k_{MN} C_M C_N,$$

where the coefficient k_{MN} is absolute reaction rate. This coefficient takes into account the average efficiency of the collision, i.e. the probability that a collision will be efficient. This coefficient depends on a number of factors, for example, on temperature determining the rate of motion of molecules or on rate that can develop a hare running away from a predator.

If the reaction involves three objects M, N, Q (the third-order reaction):

$$M + N + Q \to P,$$

then the rate of formation of the product P is proportional to the product of all three concentrations:

$$v_{MNQ} = k_{MNQ} C_M C_N C_Q. \quad (1)$$

In the case, if all three molecules are identical, for example, in the polymerization reaction, the rate is proportional to the cube of concentration.

However, as a rule, similar reactions go in two stages:

$$M + N \to [MN],$$

$$[MN] + Q \to P,$$

i.e. first, the complex [*MN*] of two molecules is formed, and then the third molecule joins this complex (each of these reactions is the second-order reaction). Then

$$v_{MNQ} = k_{[MN]Q} C_{[MN]} C_Q. \quad (2)$$

If the intermediate compound [*MN*] is unstable and quickly decays into components, then the concentration $C_{[MN]}$ becomes proportional to the product of concentrations of the initial substances: $C_{[MN]} \sim C_M C_N$ and formula (2) transforms to formula (1).

Processes of lower order (first-order reactions) are possible. For example, in the process of decay of a complex molecule into two simpler molecules, the decay rate is proportional to the concentration of the substance decaying:

$$v_{M \to N+Q} = k_M C_M$$

but not to the probability of their meeting.

There are also possible zero-order reactions which rate is independent of concentrations, for example, processes in which substance flows into (or flows out) the reaction region (or from the reaction region) at a constant rate.

We can also use similar formulas for rates when the number of reacting molecules is small. The limiting case is case if there is only one molecule in the reaction region. If this molecule reacts irreversibly turning into another, then the process ends, such case being not interesting. However, if this molecule is a catalyst, i.e. it reacts with another molecule which is a substratum (there being a lot of substrata in the reaction region), it processes a substratum, it turns a substratum into a final product, a catalyst itself recovering to previous form (regenerating), then for sufficient time, this one molecule of a catalyst may process a lot of molecules of a substratum. For this, it is necessary that processing time, i.e. time during which the catalyst and substratum are in bound state, may be much less than monitoring time. Then we can speak about the probability p to find the molecule of the catalyst in a free state or the probability $q = 1 - p$ in a bound state. In the kinetics of the formation of the catalyst-substratum complex, the probability p would play the same role as the concentration of free molecules of a catalyst if there were a lot of them.

The example given is not an abstraction. In the application of chemical kinetics to biological objects, we often encounter such a situation. The number of molecules of some specific substances, i.e. enzymes, in a cell, is often calculated with unities. The application of equations of chemical kinetics to describe such systems is very necessary.

Any real process consists of many separate stages as a rule. Thus, the mathematical model of the process (the model of a chain of reactions) involves many elementary acts. To construct the full mathematical model of the process, it is necessary to express rates of change of concentrations of different substances in terms of rates of separate elementary reactions. The rate of change of concentration of each substance $\frac{dC_i}{dt}$ is a derivative of the concentration C_i with respect to time t. It is equal to the difference between rates of formation and disappearance of this substance in separate reactions. If we write differential equations for all the components of the process (let their number be equal to n), then we get the system of differential equations:

$$\begin{cases} \dfrac{dC_1}{dt} = f_1(C_1, C_2, \ldots, C_n), \\ \dfrac{dC_2}{dt} = f_2(C_1, C_2, \ldots, C_n), \\ \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \\ \dfrac{dC_n}{dt} = f_n(C_1, C_2, \ldots, C_n). \end{cases} \quad (3)$$

The number of differential equations in this system is equal to the number of variables, i.e. the number of various substances taking part in the process. Then the system is always closed.

The functions $f_i(C_1, C_2, \ldots, C_n)$ are algebraic sums of rates of separate reactions; as a rule, these functions are rational and often are polynomials of low degrees determining by the order of corresponding reactions.

Consider the reaction catalyzing by a biological catalyst (an enzyme), that is by a large protein molecule (i.e. by macromolecule which molecular weight is of the order of hundreds of thousands), often containing a special group of non-protein nature, i.e. coenzyme [2], [3]. The mechanism of catalysis is as follows: first, the molecule that turns, so called substratum, as a rule, is a small molecule that joins enzyme and forms a complex. Then the enzyme processes the molecule of the substratum: either breaks it down or interchanges or replaces some groups of atoms. This usually takes place in several stages. The enzyme produces the molecule (product) formed ready, i.e. the substratum *S* connecting with the enzyme *F*, forms the complex [*FS*]:

$$S + F \underset{k_{-1}}{\overset{k_{+1}}{\rightleftarrows}} [FS] \xrightarrow{k_{+2}} P + F. \quad (4)$$

The coefficient k_{+1} is a constant of a rate (or an absolute rate) of the reaction of synthesis of the complex. The reverse arrow indicates that the reaction is reversible; the absolute rate of decay of the complex is k_{-1}. The arrow on the right of [*FS*] means that the complex decays into the product *P* and the enzyme *F* at the absolute rate k_{+2}.

For simplicity, we consider this reaction to be irreversible which is in most cases. The process of synthesis of the complex is a second-order reaction and the process of its decay is a first-order reaction.

Denoting, for simplicity, concentrations of substances by the same symbols which were denoted substances by, according to the reaction scheme (4) and taking into account (3), we write the system of equations:

$$\begin{cases} \dfrac{dS}{dt} = -k_{+1}FS + k_{-1}[FS], \\ \dfrac{dF}{dt} = -k_{+1}FS + k_{-1}[FS] + k_{+2}[FS], \\ \dfrac{d}{dt}[FS] = k_{+1}FS - k_{-1}[FS] - k_{+2}[FS], \\ \dfrac{dP}{dt} = k_{+2}[FS]. \end{cases} \quad (5)$$

Positive terms in the system of equations (5) describe the grow of corresponding concentrations and negative ones describe their decline.

The condition of conservation of enzyme molecules in the reaction simplifies the system of equations (5). Indeed, if we add the third differential equation of the system (5) to the second one, we obtain:

$$\frac{d}{dt}(F + [FS]) = 0$$

or

$$F + [FS] = F_0 = \text{const}, \quad (6)$$

where F_0 is initial concentration of the enzyme, F is concentration of a free enzyme, $[FS]$ is concentration of a bound enzyme.

Thus, one of the differential equations of the system (5), for example, the second one can be replaced by the algebraic relation (6) which expresses the law of conservation of the enzyme in the reaction.

We separately write the third differential equation of the system (5) taking into account (6):

$$\frac{d}{dt}[FS] = k_{+1}(F_0 - [FS])S - k_{-1}[FS] - k_{+2}[FS]$$

or

$$\frac{d}{dt}[FS] = k_{+1}F_0S - [FS](k_{-1} + k_{+2} + Sk_{+1}). \qquad (7)$$

Consider a stationary (concerning $[FS]$) solution of the differential equation (7), i.e. put $\frac{d}{dt}[FS] = 0$ where $[FS] = \text{const} = [\overline{FS}]$. Then for the stationary (constant) concentration of the complex $[\overline{FS}]$, from the differential equation (7), we obtain:

$$[\overline{FS}] = \frac{F_0S}{K_m + S} \quad \text{or} \quad [\overline{FS}] = F_0\frac{S}{K_m + S}, \qquad (8)$$

where $K_m = \frac{k_{-1} + k_{+2}}{k_{+1}}$. As $S = K_m$, then from the formula (8), we obtain $[\overline{FS}] = \frac{F_0}{2}$, namely in this case, a half of the enzyme molecules is in the state of the complex.

The concentration of the enzyme F being much less than the concentration of the substratum S, during the "turnover" time of the enzyme, the concentration of the substratum changes very slightly. As a rule, characteristic concentrations of substrata and products, during biochemical reactions, are of the order $S \approx P \approx$ from 10^{-2} to 10^{-3} mole per litre and the concentration of the enzymes $F \approx$ from 10^{-5} to 10^{-6} mole per litre. It is necessary for the enzyme "to work" for a long time to substantially change the initial concentration of the substratum. It is such situations that are in studies of enzymatic processes in vitro. Processes occur similarly in a live cell. The substratum enters the cell from the environment.

Since for the stationary mode $\frac{d}{dt}[FS] = 0$, then from the third differential equation of the system (5), we get:

$$-k_{+1}FS + k_{-1}[\overline{FS}] = -k_{+2}[\overline{FS}]. \qquad (9)$$

Thus, for the stationary (concerning the concentration of the bound enzyme $[\overline{FS}]$) mode the system of equations (5) taking into account (9) and (8), takes on the form:

$$\begin{cases} \frac{dS}{dt} = -k_{+1}FS + k_{-1}[\overline{FS}], \\ \frac{dP}{dt} = k_{+2}[\overline{FS}] \end{cases} \Rightarrow \begin{cases} \frac{dS}{dt} = -k_{+2}[\overline{FS}], \\ \frac{dP}{dt} = -\frac{dS}{dt} \end{cases} \Rightarrow \begin{cases} \frac{dS}{dt} = -k_{+2}F_0\frac{S}{K_m + S}, \\ \frac{dP}{dt} = k_{+2}F_0\frac{S}{K_m + S}. \end{cases} \qquad (10)$$

As it is seen in (10), $\frac{dP}{dt} = -\frac{dS}{dt}$, namely if the substratum does not enter additionally from outside, the rate of decrease of the concentration of the substratum $-\frac{dS}{dt}$ is equal to the rate of increase of the concentration of the product $\frac{dP}{dt}$, which corresponds to the law of conservation of substance.

The system of equations (10) reflects the main property of enzymatic reactions, that is saturation. Indeed, as we see from the second equation of the system (10), as concentrations of the substratum are

low, the rate of product synthesis depends strongly on S and as $S \to \infty$, the rate of product synthesis takes a constant independent of the concentration of the substratum value $k_{+2}F_0$ where $k_{+2}F_0$ is a maximum rate of the enzymatic reaction and a constant k_{+2} is an enzyme turnover number indicating how many acts of catalysis the enzyme can make per unit time when fully saturated with the substratum.

The characteristic time of the enzymatic reaction τ_F depends on the enzyme turnover number k_{+2} (or on the time $\tau = \frac{1}{k_{+2}}$ for which one enzyme turnover is occurred) but it is much greater than τ. Indeed, the denotation τ_F means such time for which the enzyme manages to process a great amount of the substratum. Then taking into account the first equation of the system (10) and assuming $\frac{S}{K_m + S} \approx 1$, we can determine τ_F:

$$\tau_F \approx \frac{S}{\left|\frac{dS}{dt}\right|} \approx \frac{S}{k_{+2}F_0}.$$

Thus,

$$\tau_F \approx \frac{S}{F_0}\tau \approx 10^3\tau.$$

Similar to formulas (5)–(10), we can obtain formulas describing stopping of enzymatic reactions by special substances, i.e. inhibitors. It is very important because it enables us to control biochemical processes.

In nature, as a rule, enzymes rarely work at "maximum power" they are often taken with a "reserve". Regulation of biochemical processes is carried out by inhibiting activities of enzymes and managerial regulatory apparatus of a cell is an apparatus of "violence and inhibition". To speed up the process, the cell decreases stopping and to slow the process, it increases stopping. Biochemical processes regulate themselves in such a way.

As a rule, inhibitors are relatively small molecules often similar in structure to the molecules of substrata or products.

There are two types of stopping (inhibition): competitive (isosteric) and non-competitive (allosteric).

In the first case, the inhibitor is similar to the substratum and can take the place of the substratum in the active center; the inhibitor and substratum seem to compete for the same place on the enzyme, namely it is "the competitive stopping".

In the second case, the inhibitor is not similar to the substratum and joins the enzyme molecule in another place. However, it does not interfere with the formation of the complex but paralyzes the work of the enzyme, i.e. interferes with the formation of the product.

In the case of competitive (isosteric) stopping, the second equation of the system (10) takes on the form:

$$\frac{dP}{dt} = k_{+2}F_0 \frac{S}{K_m + S + \frac{I_0}{K_i}}. \qquad (11)$$

In the case of non-competitive (allosteric) stopping, the second equation of the system (10) takes on the form:

$$\frac{dP}{dt} = k_{+2}F_0 \frac{S}{(K_m + S)\left(1 + \frac{I_0}{K_i}\right)}. \qquad (12)$$

In the formulas (11), (12), I_0 is a concentration of the inhibitor, K_i is an inhibitor constant determining by rates of formation and dissociation of the enzyme and inhibitor complex.

Along with the inhibitors are substances which can enhance the activity of the enzyme and speed up its work. This regulation method can be called the encouragement method.

The essence of the process is as follows: special substance, i.e. the activator *A* connects with the enzyme *F* or with the existing enzyme and substratum complex; along with the common reaction

$$[FS] \xrightarrow{k_{+2}} P + F$$

occurs the decay of the triple complex:

$$[FAS] \xrightarrow{k^a_{+2}} P + [FA] \tag{13}$$

where $k^a_{+2} \gg k_{+2}$, namely the decay constant of the active complex is much greater than of non-active one. In this case (if we neglect k_{+2} compared to k^a_{+2}), the rate of release of the product (taking into account the second equation of the system (10) and according to the scheme of the reaction (13)) can be written in the form:

$$\frac{dP}{dt} = k^a_{+2} F_0 \frac{S}{K_m + S} \cdot \frac{A}{K_a + A} \tag{14}$$

where K_a is an activation constant.

Conclusions. Thus, mathematical analysis of some types of enzymatic reactions is made in this article. Obtained formulae (10)–(12), (14) enable us to determine the rate of release of a separate reaction product.

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