UNSTEADY CONDITIONS IN A POROUS REACTION-DIFFUSION MEDIUM

V. GRYTSAY

"N.N. Bogolyubov" Institute of Theoretical Physics, 14, Metrologichna St., 03143 Kyiv, Ukraine

Abstract. The paper presents a reaction-diffusion model of a flow bioreactor that uses soil bacteria *Arthrobacter globiformis* immobilized in a macroporous gel in order to transform steroids. We propose and investigate a set of nonlinear differential equations that describe biochemical processes in the presence of diffusion through porous media. Dependence of ordered and chaotic formed structures from the manipulated variables of system is investigated.

Key words: biosystem, autowave processes, self-organization, catalytic centers.

INTRODUCTION

The article discusses a mathematical model of steroid transformation by immobilized-cells of *Arthrobacter globiformis* in a macroporous gel in the flow bioreactor regime. The bioreactor is a fermenter granule, or a biosensor bioselective membrane. The model of this process was built in the complete concitation regime [4–6, 8]. The characteristics close to experimental characteristics were obtained. The articles [3, 13, 14] investigate the metabolic processes of *Arthrobacter globiformis* cells.

The mathematical model presented in this article also accounts for diffusion [1, 2, 7, 9–12]. Autowave regimes were found to influence on a catalysis process.

THE MATHEMATICAL MODEL OF FLOW BIOREACTOR

Mathematical modeling of reaction-diffusive media causes the appearance of an interesting class of problems for non-linear equations.

Together with experimentalists, we have previously constructed a model for the biotechnological process of steroids transformation, whose calculated values met experimental characteristics [4–6, 8]. This process course was studied in different conditions. This work studies the formation sequence of different dissipative and

Received October 2005;
in final form December 2006.

ROMANIAN J. BIOPHYS., Vol. 17, No. 1, P. 55–62, BUCHAREST, 2007

chaotic structures appearing in the macroporous gel of bioselective biosensor membrane depending on the diffusion ratio change.

The model is constructed in conformity to a general diagram of biochemical process course taking into consideration a diffusive mass transfer of reagents as follows [1, 2, 9–12]:

$$\frac{\partial G(x,t)}{\partial t} = D_G \frac{\partial^2 G(x,t)}{\partial^2 t} + \frac{G_0}{N_3 + G + \gamma_2 \psi} - l_1 V(E_1) V(G) - \alpha_3 G, \tag{1}$$

$$\frac{\partial P(x,t)}{\partial t} = D_P \frac{\partial^2 P(x,t)}{\partial^2 t} + l_1 V(E_1) V(G) - l_2 V(E_2) V(N) V(P) - \alpha_4 P, \tag{2}$$

$$\frac{\partial B(x,t)}{\partial t} = D_B \frac{\partial^2 B(x,t)}{\partial^2 t} + l_2 V(E_2) V(N) V(P) - k_1 V(\psi) V(B) - \alpha_5 B, \tag{3}$$

$$\begin{aligned} \frac{\partial N(x,t)}{\partial t} = {} & -l_2 V(E_2) V(N) V(P) - l_7 V(Q) V(N) + \\ & + k_{16} V(B) \frac{\psi}{K_{10} + \psi} + \frac{N_0}{N_4 + N} - \alpha_6 N, \end{aligned} \tag{4}$$

$$\begin{aligned} \frac{\partial E_1(x,t)}{\partial t} = {} & E_{10} \frac{G^2}{\beta_1 + G^2} \left(1 - \frac{P + mN}{N_1 + P + mN}\right) - \\ & - l_1 V(E_1) V(G) + l_4 V(e_1) V(Q) - \alpha_1 E_1, \end{aligned} \tag{5}$$

$$\frac{\partial e_1(x,t)}{\partial t} = -l_4 V(e_1) V(Q) + l_1 V(E_1) V(G) - \alpha_1 e_1, \tag{6}$$

$$\begin{aligned} \frac{\partial Q(x,t)}{\partial t} = {} & 6lV(Q^0 + q^0 - Q) V(O_2) V^{(1)} V(\psi) - \\ & - l_6 V(e_1) V(Q)_1 - l_7 V(Q) V(N), \end{aligned} \tag{7}$$

$$\begin{aligned} \frac{\partial O_2(x,t)}{\partial t} = {} & D_{O_2} \frac{\partial^2 O_2(x,t)}{\partial x^2} + \frac{O_{20}}{N_5 + O_2} - \\ & - lV(Q^0 + q^0 - Q) V(O_2) V^{(1)} V(\psi) - \alpha_7 O_2 \end{aligned} \tag{8}$$

$$\frac{\partial E_2(x,t)}{\partial t} = E_{20} \frac{P^2}{\beta_2 + P^2} \cdot \frac{N}{\beta + N} \left(1 - \frac{B}{N_2 + B}\right) - l_{10} V(E_2) V(N) V(P) - \alpha_2 E_2, \tag{9}$$

$$\frac{\partial\psi(x,t)}{\partial t} = l_5 V(E_1)V(G) + l_9 V(N)V(Q) - \alpha\psi \tag{10}$$

where $V(X) = X/(1+X)$, $V^1(\psi) = 1/(1+\psi^{2)})$.

The equations (1)–(10) determine the change in the concentrations (the levels) of: (1) – *hydrocortisone* (G), (2) – *prednisolone* (P), (3) – *20*β*-reduced prednisolone* (B), (4) – *NADH* (N), (5) – oxidative form of *3-ketosteroid-*Δ*-dehydrogenase* (E_1), (6) – reduced form of *3-ketosteroid-*Δ*- dehydrogenase*, (7) – oxidative form of *respiratory chain* (Q), (8) – *oxygen* (O_2), (9) – *oxysteroid-dehydrogenase* (E_2), (10) – *kinetic membrane potential* (ψ).

The function $V(X)$ characterizes the enzyme adsorption in the range of the local binding to active complexes.

$V^{(1)}(\psi)$ is the function taking into account the kinetic membrane potential effect on the redox reactions of the respiratory chain.

D_G, D_P, D_B, D_{O_2} are the diffusion coefficients of: *hydrocortisone, prednisolone, 20* β*-reduced prednisolone* and *oxygen*. These reagents have the diffusion transfer that is taken into account by introducing the partial derivatives with respect to x in (1), (2), (3), (8).

It was accepted a one-dimensional interpretation of an active portion of medium $[0,s]$. Free diffusion equations are used in border zone: $[-d,0]$, and $[s,s+d]$.

Boundary conditions have been selected, respectively:

$$\partial_x U_{(x=-d)} = \partial_x U_{(x=s+d)} = 0\,. \tag{11}$$

Parameters have been made dimensionless [1, 2, 9–12] and have the following form:
$l = l_1 = k_1 = 0.2$; $l_2 = l_{10} = 0.27$; $l_5 = 0.6$ $l_4 = l_6 = 0.5$; $l_7 = 1.2$; $l_9 = 2.4$; $k_{16} = 1.5$; $E_{10} = 3$; $\beta_1 = 2$; $N_1 = 0.03$; $m = 2.5$; $a_1 = 0.007$; $\alpha_1 = 0.0068$; $E_{20} = 1.2$; $\beta = 0.01$; $\beta_2 = 1$; $N_2 = 0.03$; $\alpha_2 = 0.02$; $G_0 = 0.019$; $N_3 = 2$; $\gamma_2 = 0.2$; $\alpha_5 = 0.014$; $\alpha_3 = \alpha_4 = \alpha_6 = \alpha_7 = 0.001$; $O_{20} = 0.015$; $N_5 = 0.1$; $N_0 = 0.003$; $N_4 = 1$; $K_{10} = 0.7$.

The nonlinear differential system (1) – (10) is a system with distributed parameters. Its solutions are obtained by the finite difference method. The nonlinear finite-difference system is solved by the Rynge-Kuta-Merson method. An accuracy of solution is 10^{-12}.

Using a numerical experiment we have found out a sequence of formation of autoperiodical regimes. Depending on the kinetic membrane potential dissipation coefficient α the order and length of period T change (Table 1), where $D = D_G = D_P = D_b = D_{O_2} = 0$.

When diminishing the dissipation ratio α from 0.056 to 0.032075, periodicity ratio of self-oscillation process is increasing from the 1 to a 14-fold one, but then again reaches the 1-fold one with a period length approximately similar to the previous condition. Transitions between conditions given in Table 1 take place *via* the appearance of strange attractors regimes.

Table 1

Dependence of auto-periodical regimes ratio on value α

Order of period	α	T	Order of period	α	T	Order of period	α	T
$1{\cdot}2^0$	0.056	218	$6{\cdot}2^0$	0.0337	1483	$11{\cdot}2^0$	0.03247	2346
$2{\cdot}2^0$	0.04	631	$7{\cdot}2^0$	0.0334	1579	$12{\cdot}2^0$	0.03238	2550
$3{\cdot}2^0$	0.036	847	$8{\cdot}2^0$	0.033	1941	$13{\cdot}2^0$	0.03227	2743
$4{\cdot}2^0$	0.03563273	1010	$9{\cdot}2^0$	0.032865	1991	$14{\cdot}2^0$	0.03212	2952
$5{\cdot}2^0$	0.0345	1398	$10{\cdot}2^0$	0.03262	2174	$1{\cdot}2^0$	0.032075	222

The difference of one-fold periodic regimes appearing at different α values consists in the fact that at high dissipation ($\alpha = 0.056$) the biosystem lies close to the thermodynamic branch and the flow of biochemical process is determined through external conditions. At low dissipation ($\alpha = 0.32075$) – the system is self-organized, and the dissipative structure is established. The activity of a biochemical process in this case is determined, as a whole, through internal self-organization of a biosystem.

Assuming as a basis each of the mentioned regimes we have carried out a study of the dependence of space-time structures of reagents on the diffusion ratio value. We have found scenarios for the formation of both dissipative and chaotic structures. One of the scenarios for 8-fold autowave process ($8{\cdot}2^0$) appearing at $D = 0$ is shown in Table 2.

The value D corresponds to values at which bifurcation appears and the most interesting conditions are noticed. At any other intermediate values the space-time structures are changed “smoothly” and they are similar to structures corresponding to the nearest lower value D.

Table 2

Scenario for the formation of space-time structures of different conditions depending on diffusion ratio *D*, at $\alpha = 0.033$, $x \in [1,24]$ ($s = 22$, $d = 2$) and $t \in [10^5 - 10^5 + 10^3]$

D	Structure	*D*	Structure	*D*	Structure
0.000100	$\approx 8 \cdot 2^0$	0.0048990	St. ($8 \cdot 2^0$) St.	0.0083000	Stable
0.001000	Chaos $8 \cdot 2^\infty$	0.0050000	St. ($1 \cdot 2^0$) St.	0.0085000	Stable
0.002000	Chaos $8 \cdot 2^\infty$	0.0055000	St. ($8 \cdot 2^0$) St.	0.0087000	Stable
0.003500	St. ($12 \cdot 2^0$) St.	0.0058000	Stable	0.0100000	Stable
0.003910	St. ($1 \cdot 2^0$) St.	0.0059000	Stable	0.0120000	Stable
0.003930	St. ($1 \cdot 2^0$) St.	0.0059500	Stable	0.0200000	Stable
0.003950	Stable	0.0059700	Stable	0.0300000	Stable
0.004000	Stable	0.0059800	Stable	0.0440600	Stable
0.004050	St. ($8 \cdot 2^0$) St.	0.0059900	$1 \cdot 2^0$–($8 \cdot 2^0$)–$1 \cdot 2^0$	0.0441000	Stable
0.004070	St. ($8 \cdot 2^0$) St.	0.0059960	$1 \cdot 2^0$–($8 \cdot 2^0$)–$1 \cdot 2^0$	0.0500000	Stable
0.004090	St. ($1 \cdot 2^0$) St.	0.0059963	$1 \cdot 2^0$– St. –$1 \cdot 2^0$	0.0600000	Stable
0.004100	Chaos $8 \cdot 2^\infty$	0.0059965	Stable	0.0630000	Stable
0.004200	St. ($9 \cdot 2^0$) St.	0.0059968	Stable	0.0650000	Stable
0.004300	$8 \cdot 2^\infty$ St. $8 \cdot 2^\infty$	0.0059970	$1 \cdot 2^0$–($8 \cdot 2^0$)–$1 \cdot 2^0$	0.0660000	Stable
0.004400	St.Chaos n$\cdot 2^\infty$St	0.0059973	St. ($1 \cdot 2^3$) St.	0.0661000	Stable
0.004410	St. ($8 \cdot 2^0$) St.	0.0059975	$1 \cdot 2^0$–($8 \cdot 2^0$)–$1 \cdot 2^0$	0.0662000	Chaos n$\cdot 2^\infty$
0.004415	St. ($1 \cdot 2^0$) St.	0.0059978	Stable	0.0663000	Chaos n$\cdot 2^\infty$
0.004420	Stable	0.0059980	St. ($1 \cdot 2^1$) St.	0.0665000	Chaos n$\cdot 2^\infty$
0.004425	$1 \cdot 2^0$ –($9 \cdot 2^0$)–$1 \cdot 2^0$	0.0060000	$1 \cdot 2^0$ –St. – $1 \cdot 2^0$	0.0666000	Chaos n$\cdot 2^\infty$
0.004435	St. ($9 \cdot 2^0$) St.	0.0061000	Stable	0.0667000	Chaos n$\cdot 2^\infty$
0.004440	Stable	0.0062000	Stable	0.0668000	Unstable Focus
0.004441	Stable	0.0063000	Stable	0.0669000	Chaos $1 \cdot 2^\infty$
0.004442	St. ($8 \cdot 2^0$) St.	0.0064000	Stable	0.0670000	Chaos n$\cdot 2^\infty$
0.004445	St. ($1 \cdot 2^0$) St.	0.0065000	St. ($8 \cdot 2^0$) St.	0.0677000	Chaos n$\cdot 2^\infty$
0.004450	St. ($1 \cdot 2^0$) St.	0.0066000	Stable	0.0680000	Chaos n$\cdot 2^\infty$
0.004500	Stable	0.0067000	Stable	0.0685000	$8 \cdot 2^0$
0.004501	St. ($1 \cdot 2^0$) St.	0.0068000	St. ($8 \cdot 2^0$) St.	0.0690000	Chaos n$\cdot 2^\infty$
0.004505	St. ($8 \cdot 2^0$) St.	0.0068300	St. ($8 \cdot 2^0$) St.	0.0700000	$8 \cdot 2^0$
0.004515	St. ($8 \cdot 2^0$) St.	0.0068500	Stable	0.0800000	$8 \cdot 2^0$
0.004518	$1 \cdot 2^0$ –St.– $1 \cdot 2^0$	0.0068800	St. ($8 \cdot 2^0$) St.	0.1000000	$8 \cdot 2^0$
0.004520	Stable	0.0070000	St. ($8 \cdot 2^0$) St.	0.3000000	$8 \cdot 2^0$
0.004530	St. ($1 \cdot 2^0$) St.	0.0080000	St. ($8 \cdot 2^0$) St.	0.5000000	$8 \cdot 2^0$
0.004600	St. ($8 \cdot 2^0$) St.	0.0081000	St. ($1 \cdot 2^0$) St.		
0.004850	St. ($8 \cdot 2^0$) St.	0.0082000	Stable		

St. is Stable, and defines steady state domain of space-time structures $G(x,t)$.

Fig. 1, a, b, c, d shows examples of space-time structures $G(x,t)$ at various D values.

The consecutive increase of the diffusion ratio from 0.0001 to 0.5 causes the destruction of old and the appearance of new different in type space-time structures. After 8-fold quasi-periodical auto-wave structure ($\approx 8 \cdot 2^0$) a chaos appears (Chaos $8{\cdot}2^{\infty}$), then another kind of chaos (Chaos $8{\cdot}2^{\infty}$), localized in a centre 12-fold periodicity dissipative structure (St.($12{\cdot}2^{0}$)St.), etc. until the end of scenario. Auto-wave or chaotic structures appeared are localized in one, two or three zones separated between them with stabilized states. If a diffusion ratio value increased up to 0.07 we shall obtain a space-time periodical structure of 8-fold period similar to event $D = 0.0001$, but its all components at x owing to a high diffusion will oscillate practically synchronously with a very low phase shift.

One of the most interesting effects occurs when a stabilized space-unhomogeneous structure following bifurcation will instantly be transferred to another kind of space-unhomogeneous stabilized structure (for example, D from 0.0058 to 0.00598 and others).

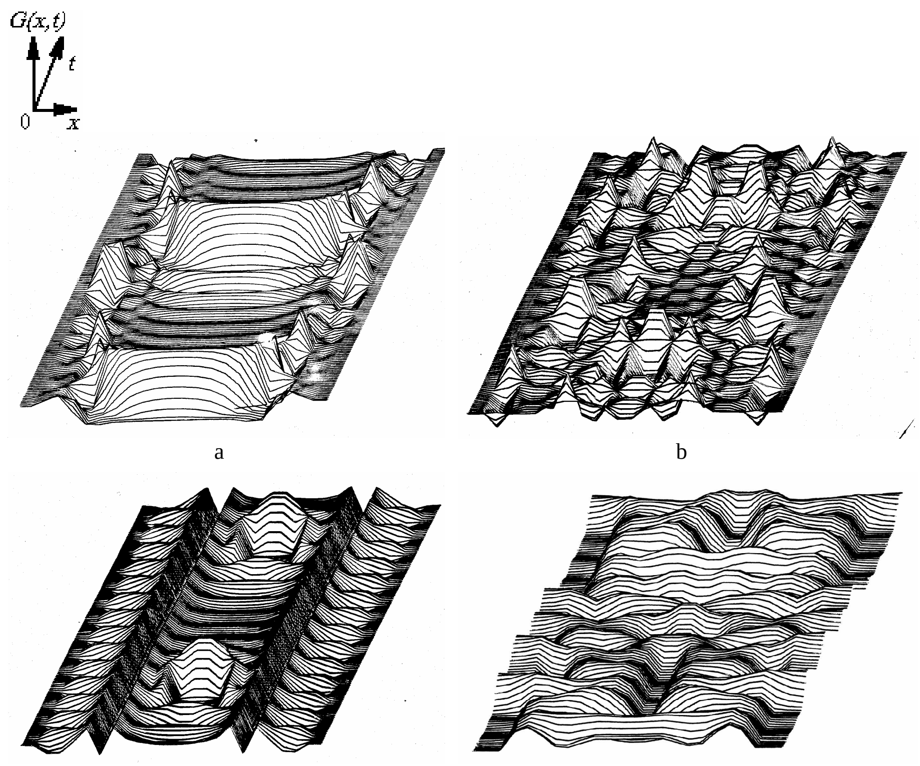

Fig. 1. Space-time structures $G(x,t)$; a. quasi-periodical auto-wave structure of 8-fold period, $D = 0.0001$, $[\min G;\ \max G]$ = $[0.1663;\ 0.5314]$; b. chaos, $D = 0.001$, $[\min G;\ \max G]$ = $[0.1677;\ 0.5042]$; c. localization of dissipative auto-wave structures in 3 zones: 9-fold periodical one – in the centre, 1-fold – at edges, $D = 0.004425$, $[\min G;\ \max G]$ = [0.1656; 0.5185]; d. chaos, $D = 0.069$, $[\min G;\ \max G]$ = [0.1795; 0.3546].

The obtained diversity of space-time structures for reagents of reaction diffusive porous medium is defined through structural-functional bonds of a biosystem and reflects changes in the kinetics of the biochemical process caused by a change of its diffusion ratio value taking place within the period of this process.

CONCLUSION

The results reveal the diffusion-dependence of the activity of catalytic reactions. In the case in which an autocatalytic process appears, autowave regimes can emerge in the bioreactor and that influences the catalysis process. Variable areas of different catalytic reactivity form in the granules of fermenter in these regimes. The biochemical process activity is not in accord with the measured signal in the bioselective membrane of biosensor. The sensitive and insensitive areas of measuring are formed.

The results can be generalized and used in the investigation of catalyst porous granules and biosensor bioselective membrane.

REFERENCES

1. ANDREEV, V.V., V.I. GRYTSAY, Inactive regions modeling in porous granules of catalyst and in biosensor (in Russian), *Matematicheskoe Modelirovanie*, 2005, **17**, 57–64.
2. ANDREEV, V.V., V.I. GRYTSAY, Passing diffusion-reactionary process effect on structure formation in porous medium (in Russian), *Matematicheskoe Modelirovanie*, 2005, **17**, 3–12.
3. ARINBASAROVA, A.Y., A.V. KARPOV, V.V. FOKINA, A.G. MEDENTSEV, K.A. KOSHCHEYENKO, Kinetic characteristics of 1-en-dehydrogenation of 6α-methylhydrocortisone by cells of *Arthrobacter globiformis* 193, *Enzyme and Microbial Technology*, 1996, **19**, 501–506.
4. GACHOK, V.P., A.YU. ARINBASAROVA, K.A. KOSHCHEYENKO, A.G. MEDENTSEV, V.K. AKIMENKO, Kinetic model of hydrocortisone 1-en dehydrogenation by *Arthrobacter globiformis*, *Appl. Biochem. Microbiol.*, 1988, **XXIV**, 380–388.
5. GACHOK, V.P., A.YU. ARINBASAROVA, V.I. GRYTSAY, A.G. MEDENTSEV, K.A. KOSHCHEYENKO, V.K. AKIMENKO, Kinetic model for the regulation of redox reactions in steroid transformation by *Arthrobacter globiformis* cells, *Appl. Biochem. Microbiol.*, 1988, **XXIV**, 389–399.
6. GACHOK, V.P., V.I. GRYTSAY, A.YU. ARINBASAROVA, A.G. MEDENTSEV, K.A. KOSHCHEYENKO, V.K. AKIMENKO, Kinetic model for the regulation of redox reactions in steroid transformation by *Arthrobacter globiformis* cells, *Biotechnology and Bioengineering*, 1989, **33**, 668–680.
7. GACHOK, V.P., V.I. GRYTSAY, Kinetic model of macroporosity granules with biochemical process control (in Russian), *Doklady AN SSSR*, 1985, **282**, 51–53.
8. GACHOK, V.P., V.I. GRYTSAY, A.YU. ARINBASAROVA, A.G. MEDENTSEV, K.A. KOSHCHEYENKO, V.K. AKIMENKO, Kinetic model of hydrocortisone 1-en dehydrogenation by *Arthrobacter globiformis*, *Biotechnology and Bioengineering*, 1989, **33**, 661–667.

9. GRYTSAY, V.I., Self-organization in macroporosity gel structure with immobilization cells. Kinetic model of bioselective membrane of biosensor (in Russian), *Reports of NAS of Ukraine*, 2000, **2**, 175–179.
10. GRYTSAY, V.I., Self-organization in reaction-diffusion environment (in Russian), *Reports of NAS of Ukraine*, 2000, **3**, 201–206.
11. GRYTSAY, V.I., V.P. GACHOK, YU.I. PRYLUTSKYY, M.E. KUCHERENKO, O.P. MATYSHEVSKA, S.V. PRYLUTYSKA, P. SCHARFF, Modeling of the biochemical processes in the bioselective membrane of biosensor using the colloidal fullerene particles, *EUROCARBON 2000, 1-st World Conference on Carbon*, Germany, Berlin, 2000, 969–970.
12. GRYTSAY, V.I., Self-organization of the immobilization cells biochemical process in the biosensor bioselective membrane (in Ukrainian), *Ukrainian Journal of Physics*, 2001, **1**, 124–127.
13. MUKAI, K., K. MARUTA, K. SATOUCHI, M. KUBOTA, S. FUKUDA, M. KURIMOTO, Y. TSUJISAKA, Cyclic tetrasaccharide-synthesizing enzymes from *Arthrobacter globiformis* A19, *Biosci. Biotechnol. Biochem.*, 2004, **68**, 2529–2540.
14. SUZUKI, K., S.-I. SAKASEGAWA, H. MISAKI, M. SUGIYAMA, Molecular cloning and expression of uricase gene from *Arthrobacter globiformis* in *Escherichia coli* and characterization of the gene product, *J. Biosci. Bioeng.*, 2004, **98**, 153–158.