Modelling of immobilized cell systems

J. C. VINCENT, J. LEFEBVRE
URA 500 CNRS "Polymères, Biopolymères, Membranes", Université de Rouen, Faculté des Sciences, BP 118, 76134 Mont Saint Aignan Cedex, France

A new model for the dynamic evolution of a membrane system containing immobilized cells is designed and theoretically studied. The analysis is based on the diffusion–reaction theory in which both the diffusion and reaction components are space and time dependent. The numerical treatment gives the time evolution of the system which tends toward a U-form cell distribution in the membrane, depending on its transport characteristics.

1. Introduction

There are many ways to consider immobilized cell systems [1]. The simplest theoretical approach is to study the system under steady-state conditions without taking into account its dynamic behaviour. The growth of the microorganisms in the gel is not considered, indeed the biocatalytic phase, i.e. the biomass, remains constant. The first studies developed simple diffusion–reaction systems [2], either by micro–macro models [3, 4] or by intrinsic models [5]. The new trend is to develop dynamic models by combining diffusion, reaction and also cellular growth [6–10], in order to describe how the rate of substrate consumption, the diffusion and the cell growth vary in the gel as a function of space and time. Both theoretical and experimental results show that the dynamical evolution of the systems leads to a heterogeneous distribution of the biomass, only the layer near the surface of the biocatalytic particle being active at the steady-state.

For the last few years, our laboratory has been engaged in the design and improvement of bioreactors [11] which, nowadays, require modelling. The aim of this paper is to present a new model which describes the dynamic evolution of membranes containing immobilized cells in order to reach a better understanding of the phenomena which control cell reactors.

2. Theory

The model is composed of two compartments separated by a gel slab containing the immobilized cells. A schematic representation of the system is shown in Fig. 1. The reaction occurring in the reactor corresponds to both the metabolic reaction which transforms substrate S into product P and the cell growth which increases the cell concentration inside the gel. The reaction can be represented by the equation:

\[ n \text{ cells} + \text{substrate } S \rightarrow m \text{ cells} + \text{product } P \] (1)

While cells are immobilized in the gel slab and not diffusing, substrate and product are involved in two coupled phenomena: the reaction and the diffusion.

The main assumptions on which the model is formulated are: (1) cells are homogeneously distributed inside the gel slab at the outset of the reaction; (2) no cell leakage will occur; (3) because of the geometry of the membrane, the diffusion phenomenon will be considered only in the direction perpendicular to the membrane; (4) complete mixing occurs in the reactor, hence the substrate concentration at the surface of the membrane is equal to the bulk substrate concentration; (5) the reaction catalysed by cells is assumed to be of the Michaelis–Menten type and the cell growth is assumed to follow a law of the Monod type without inhibition and death. The reaction parameters of the immobilized cells are equal to those of the free cells; and (6) the diffusion coefficients in the gel are dependent on the local cell concentration.

On the basis of these assumptions, the unsteady-state balance equation inside the membrane for a species Z is given, as for immobilized enzyme systems [12], by the classical diffusion–reaction law

\[
\frac{\partial Z(x,t)}{\partial t} = \frac{\partial^2 Z(x,t)}{\partial x^2} - \nabla \cdot \mathbf{q} + \rho Z(x,t)
\] (2)

where

\[
\mathbf{q} = \frac{\partial Z(x,t)}{\partial t} \text{diffusion} = D_Z \frac{\partial^2 Z(x,t)}{\partial x^2}
\] (3)

\[
\rho Z(x,t) = \varepsilon \left( \frac{V_m S(x,t)}{K_m + S(x,t)} B(x,t) \right)
\] (4)

\[
\varepsilon = \begin{cases} 
+1 & \text{when } Z = P \\
-1 & \text{when } Z = S 
\end{cases}
\]

\(V_m\) and \(K_m\) are the equivalent Michaelis–Menten constants corresponding to the immobilized cell, \(x\) is the abscissa coordinate along the thickness of the membrane.
membrane, \( t \) the time and \( D_z \) the diffusion coefficient of species \( Z \) inside the membrane. In our model, \( D_z \) is assumed to be a hyperbolic function of the local cell concentration (Fig. 2).

For cell growth, the Monod equation is used

\[
\frac{\partial B(x, t)}{\partial t} = \mu B(x, t)
\]

in which

\[
\mu = \frac{S(x, t)}{K_s + S(x, t)} \mu_m
\]

The initial and boundary conditions are:

\[
t = 0 : S(x, 0) = 0, \text{ for } 0 < x < e \\
P(x, 0) = 0, \text{ for } 0 \leq x \leq e \\
B(x, 0) = B_0, \text{ for } 0 < x < e
\]

where \( e \) is membrane thickness

\[
t > 0 : S(0, t) = S(e, t) = S_0
\]

where \( S_0 \) is initial concentration

Owing to the complexity of the partial derivative equations, no analytical solutions can be calculated. A numerical simulation is required and the explicit scheme will be used leading to

\[
B_{x+\Delta t} = B_x + \Delta t \mu_m \left[ \frac{S_x'}{K_s + S_x'} \right] B_x'
\]

and

\[
S_{x+\Delta t} = S_x' + \frac{\Delta t}{\Delta x^2} D_z \left( B_x'(S_x + S_x') + S_x'' \Delta x \\
- 2 S_x'' \right) + \Delta t V_m \left[ \frac{S_x'}{K_m + S_x'} B_x' \right]
\]

3. Results

Equations 8 and 9 give the time evolution of the system which shows the simultaneous substrate consumption and cell growth. These two phenomena are coupled: the cell growth is dependent on the local substrate concentration inside the membrane and the diffusion (Equation 3) and reaction (Equation 4) parts of the global substrate equation are functions of the local cell concentration. This explains why the system tends toward a stabilization of the concentration profiles, which we call steady-state distribution. Three types of result will be shown: (i) the shape of the steady-state cell distribution inside the membrane, (ii) the different steps which lead the system to its steady-state, and (iii) the influence of diffusion and reaction parameters on the steady-state cell distribution.

3.1. Intramembrane cell concentration profiles

Because of the cell growth, the catalyst concentration increases with time inside the membrane. Owing to the diffusion constraints which affect the uptake of substrate, the best conditions for cell growth will be obtained near the membrane–solution interfaces and the increase in cell concentration will be maximum in these regions. The heterogeneity in the cell distribution inside the membrane will be reinforced by the increase of the diffusion constraints in these regions due to the fact that the cells progressively occupy the free space in the membrane. The result is strong heterogeneity of the cell concentration inside the membrane as shown in Fig. 3 where the cell concentration is plotted as a function of membrane thickness. The greatest part of the biomass is located near the membrane–solution interfaces.

3.2. Time evolution of the system

Because of the cell growth, the catalyst concentration and thus the reaction rate inside the membrane is not constant as a function of time. Three periods can thus be distinguished.

(i) The first period is characterized by a low cell concentration leading to a diffusion–reaction balance in favour of the diffusion. The substrate uptake by the membrane can be seen both in the substrate concentration profile (Fig. 4) and in the reaction rate profile (Fig. 5). The substrate is consumed as soon as it diffuses into the gel, leading to a cell growth limited to the borders of the membrane. Conversely, the cell growth is very slow in the centre of the membrane because of the low concentrations of substrate and cells.

(ii) When the membrane is filled by the substrate, the reaction rate increases and the net rate becomes