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ON

VARIATIONAL METHODS IN ANALYSIS AND MATHEMATICAL PHYSICS

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MATHEMATICAL MODELING OF IMMOBILIZED ENZYME SYSTEMS

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5.1. *Statement of the Problems.*

When a model or a biochemical reactor is realized or designed, it is important to know how it is possible to control its behavior.

5.1.1. Substrate Flux Control by a Boundary Inhibitor Concentration. This system was described in 1.2.3. I.

The boundary concentration of inhibitor is written $v(t) = (v_0(t), v_1(t))$.

$$\begin{cases} \dfrac{\partial y}{\partial t} - \dfrac{\partial^2 y}{\partial x^2} + F(y,i) = 0 \qquad \text{where } F(y,i) = \dfrac{y}{1+y+\lambda i} \\ y(0,t) = \alpha,\ y(1,t) = \beta,\ y(x,0) = 0 \\ \dfrac{\partial i}{\partial t} - \dfrac{\partial^2 i}{\partial x^2} = 0,\ i(0,t) = v_0(t),\ i(1,t) = v_1(t),\ i(x,0) = 0 \end{cases} \tag{5.1}$$

In the compartments, $v_0(t)$ and $v_1(t)$ are at our disposal. We can change $v = (v_0, v_1)$ by adding inhibitor or by creating a reaction of disappearance of inhibitor.

The constraints on v are

$$0 \leq v_0(t) \leq M,\ 0 \leq v_1(t) \leq M,\ M \geq 0. \tag{5.2}$$

We define

$$\mathscr{U}_{ad} = \{v \in (L^2(0,T))^2; 0 \leq v_0 \leq M, 0 \leq v_1 \leq M\} \tag{5.3}$$

Then, $v \in \mathscr{U}_{ad}$.

By changing v, we want to minimize the difference between a desired flux and the substrate flux entering the membrane. We define the cost function J by

$$J(v) = \int_0^T \left[-\frac{\partial y}{\partial x}(0,t) - z_d^0(t) \right]^2 dt + \int_0^T \left[\frac{\partial y}{\partial x}(1,t) - z_d^1(1,t) \right]^2 dt \tag{5.4}$$

and we are looking for some $u \in \mathscr{U}_{ad}$ such that

$$J(u) = \inf_{v \in \mathscr{U}_{ad}} J(v) \tag{5.5}$$

A similar problem is the substrate concentration control in the middle of a membrane by a boundary inhibitor concentration. It is studied in C. M. Brauner and P. Penel (1972).

The observation is now

$$t \to y(\tfrac{1}{2}, t) \qquad \text{(electrode)}$$

and we wish to minimize

$$J(v) = \int_0^T |y(\tfrac{1}{2},t) - z_d(t)|^2 dt \tag{5.6}$$

5.1.2. Active Transport Control. This bienzyme system has been described in 1.3.1.

It would be interesting to control the active transport velocity against the concentration gradient.

It is possible to perform this control because the presence of an activator (M_g^{++}) is a necessity for the enzyme reaction in the first layer.

Equations of this system:

(y_1, y_2, y_3 are substrate, product and activator concentrations).

$$\begin{cases} \dfrac{\partial y_1}{\partial t} - \dfrac{\partial^2 y_1}{\partial x^2} + F(y_1,y_2,y_3) = 0 \\ \qquad\qquad\qquad\qquad\qquad\qquad x \in]0,1[\\ \dfrac{\partial y_2}{\partial t} - \dfrac{\partial^2 y_2}{\partial x^2} - F(y_1,y_2,y_3) = 0 \\ \qquad\qquad\qquad\qquad\qquad\qquad t \in]0,T[\\ \dfrac{\partial y_3}{\partial t} - \dfrac{\partial^2 y_3}{\partial x^2} = 0 \quad \text{where} \\ F(y_1,y_2,y_3) = \sigma \dfrac{y_3}{1+y_3} \dfrac{y_1}{1+y_1+y_2} \\ \text{if } 0 < x < \tfrac{1}{2} \qquad (\sigma > 0) \\ F(y_1,y_2,y_3) = -\sigma \dfrac{y_2}{1+y_2} \qquad \text{if } \tfrac{1}{2} < x < 1 \end{cases} \tag{5.7}$$

Boundary Conditions:

$$y_1(0,t) = \alpha \qquad \alpha > 0 \tag{5.8}$$

$$\begin{cases} \dfrac{\partial y_1}{\partial t} + \dfrac{\partial y_1}{\partial \nu} = 0 \qquad \text{for } x = 1 \\ y_1(1,0) = \beta \qquad \beta \geq 0 \end{cases} \tag{5.9}$$

$$\frac{\partial y_2}{\partial \nu} = 0 \qquad \text{for } x = 0 \text{ and } x = 1 \tag{5.10}$$

$$y_3(0,t) = 0, \qquad y_3(1,t) = v(t) = \textit{control}. \tag{5.11}$$

Initial Conditions:

$$y_1(x,0) = 0, \quad y_2(x,0) = 0, \quad y_3(x,0) = 0 \tag{5.12}$$

The difference between the boundary conditions for y_1 in $x = 0$ and $x = 1$ is linked to the compartment relative sizes. The first compartment is large enough to maintain a constant concentration, the second one is smaller by far.

The control is the activator concentration in the second compartment.

This control is in the set of admissible controls:

$$\mathscr{U}_{ad} = \{v; v \in L^2(0,T), \quad 0 \leq v(t) \leq M\} \tag{5.13}$$

The cost function J is defined by

$$J(v) = \int_0^T (y_1(1,t) - z_d)^2 dt \tag{5.14}$$

A variant has been studied in J. P. Yvon (1973a, 1973b), where the boundary films are selective only for a concentration of product smaller than h:

$$\frac{\partial y_2}{\partial \nu} = \begin{cases} 0 & \text{if } y_2 < h \\ -ay_2 \text{ if } y_2 > h & (a > 0) \end{cases} \tag{5.15}$$

5.1.3. Identification Problems. For these problems one is referred to

G. Joly (1974)
G. Joly *et al.* (to appear)
G. Joly *et al.* (to appear)

5.2. *Mathematical Part.*

5.2.1. Control of the Flux of Substrate Entering an Enzymatic Membrane by an Inhibitor Concentration at the Boundary. The equations are (5.1) and the cost function

$$J(v) = \int_0^T t^{2\gamma}\left(-\frac{\partial y}{\partial x}(0,t) - z_d^0(t)\right)^2 dt + \int_0^T t^{2\gamma}\left(\frac{\partial y}{\partial x}(1,t) - z_d^1(t)\right)^2 dt \tag{5.16}$$

instead of (5.4) because, as we shall see later in this §, $t^\gamma \frac{\partial y}{\partial x}(0,t)$ and $t^\gamma \frac{\partial y}{\partial x}(1,t)$ are defined and belong to $L^2(0,T)$ for $\gamma > \frac{1}{2}$.

a) *Existence of an optimal control*

i The inhibitor concentration i is given by

$$i(x,t) = \sum_{j=1}^{\infty} c_j(t)\, w_j(x) \tag{5.17}$$

in $L^2(\Omega)$ where

$$w_j(x) = \sqrt{2} \sin j\pi x \tag{5.18}$$

$$c_j(t) = \sqrt{2}\pi j \int_0^t e^{-\pi^2 j^2 (t-\sigma)} (v_0(\sigma) + (-1)^{j+1} v_1(\sigma)) d\sigma. \tag{5.19}$$

Since, from (5.3), $0 \le v_0, v_1 \le M$,

$$c_j(t) \le \frac{2\sqrt{2}M}{\pi j}(1 - e^{-\pi^2 j^2 t}) < \frac{2\sqrt{2}M}{\pi j}$$

and if we call

$$\lambda_j = \pi^2 j^2 \quad \text{we have} \tag{5.20}$$

$$\sum_{j=0} \lambda_j^{1/2-\epsilon} \int_0^T c_j^2(t) dt < \infty \tag{5.21}$$

Let us show that this is equivalent to

$$i \in L^2(0,T;H^{1/2-\epsilon}(\Omega)) \qquad (\Omega =]0,1[) \tag{5.22}$$

Let $w_j(x) = \sqrt{2} \sin j\Pi x$. If $\phi \in L^2(\Omega)$, $\phi = \sum_{j=1}^{\infty} \phi_j w_j$ and the application $\Pi : \phi \to (\phi_j)$ is an isomorphism from $H^0(\Omega) = L^2(\Omega)$ to $l_0^2 = l^2 = \{(\phi_j) | \Sigma\phi_j^2 < \infty\}$

A denoting $-\frac{d^2}{dx^2}$, Π is also an isomorphism from

$$H^2(\Omega) \cap H_0^1(\Omega) = D(A) \to l_\lambda^2 = \{(\phi_j) | (\lambda_j \phi_j) \in l^2\}$$

$$\Pi \in \mathscr{L}(D(A);l_\lambda^2) \cap \mathscr{L}(L^2(\Omega);l^2)$$

so that

$$\Pi \in \mathscr{L}([D(A);L^2(\Omega)]_\theta; [l_\lambda^2;l^2]_\theta)$$

$$[l_\lambda^2;l^2]_\theta = \{(\phi_j) | (\lambda_j^{1-\theta}\phi_j) \in l^2\}$$

$$[D(A);L^2(\Omega)]_\theta = \begin{cases} H^{2(1-\theta)} \text{ for } 2(1-\theta) < \frac{1}{2},\ \theta > \frac{3}{4} \\ \{\phi | \phi \in H^{2(1-\theta)},\ \phi_{|\Gamma} = 0\} \text{ for } \theta \le \frac{3}{4} \end{cases}$$

Let us apply that to (5.21): $1-\theta = \frac{1}{4} - \frac{\epsilon}{2}$, $\theta = \frac{3}{4} + \frac{\epsilon}{2}$, $2(1-\theta) = \frac{1}{2} - \epsilon$ whence (5.22).

ii Let us recall also that, $i(x,t)$ being known $y(x,t)$ is the solution of

$$\frac{\partial y}{\partial t} - \frac{\partial^2 y}{\partial x^2} + F(y,i) = 0, \qquad F(y,i) = \sigma \frac{y}{1+y+i} \tag{5.23}$$

$$y(0,t) = \alpha, \qquad y(1,t) = \beta, \qquad y(x,0) = 0 \tag{5.24}$$

and in particular $y \in L^\infty(Q)$.

To be able to give a correct definition of $\frac{\partial y}{\partial \nu}$ we are going to define a new function z:

$$z = t^\gamma (y - \Phi) \tag{5.25}$$

where Φ is the affine function

$$\Phi : x \to (\alpha - \beta)x + \alpha$$

(5.23) and (5.24) become

$$z' - \frac{\partial^2 z}{\partial x^2} = \gamma t^{\gamma-1}(y - \Phi) - t^\gamma F(y,i) = G \tag{5.26}$$

$$z(0,t) = z(1,t) = 0 \tag{5.27}$$

$$z(x,0) = 0 \tag{5.28}$$

and, for $\gamma > \frac{1}{2}$, $G \in L^2(0,T;L^2(\Omega)) = L^2(Q)$.

The initial and boundary conditions for z are compatible and z, element of $L^2(0,T;H_0^1(\Omega))$, satisfies

$$z', Az \in L^2(Q) \tag{5.29}$$

$$z \in H^{2,1} = \{\phi | \phi, \frac{\partial \phi}{\partial t}, \frac{\partial \phi}{\partial x}, \frac{\partial^2 \phi}{\partial x^2} \in L^2(Q)\} \tag{5.30}$$

since

$$\begin{cases} z' + Az = G \in L^2(Q) \\ z(0) = 0 \end{cases} \tag{5.31}$$

We can then define $\frac{\partial z}{\partial \nu} \in L^2(0,T;H^{1/2}(\Gamma)) = L^2(0,T)$, the application $z \to \frac{\partial z}{\partial \nu}$ being linear continuous from $H^{2,1}(Q) \to L^2(0,T)$.

At last

$$\frac{\partial z}{\partial \nu} = t^\gamma \frac{\partial y}{\partial \nu} \tag{5.32}$$

iii) *A priori estimations*

So we have the functional (5.16) and let $v_n = ((v_0)_n, (v_1)_n)$ be a minimizing sequence

$$v_n \in \mathscr{U}_{ad} = \text{bounded set of } (L^\infty(0,T))^2 \tag{5.33}$$

$$i_n \in \text{bounded set of } L^2(0,T;H^{1/2-\epsilon}(\Omega)) \qquad \text{(from i)} \tag{5.34}$$

$$\frac{\partial i_n}{\partial t} \in \text{bounded set of } L^2(0,T;H^{-3/2-\epsilon}(\Omega)) \tag{5.35}$$

from the last equation in (5.1)

$$y_n - \Phi \in \text{bounded set of } L^\infty(0,T;L^2(\Omega)) \cap L^2(0,T;H_0^1(\Omega)) \tag{5.36}$$

from

$$\frac{\partial}{\partial t}(y_n - \Phi) - \frac{\partial^2}{\partial x^2}(y_n - \Phi) + F(y_n,i_n) - F(\Phi,i_n) = -F(\Phi,i_n)$$

$$G_n = \gamma t^{\gamma-1}(y_n - \Phi) - t\gamma F(y_n,i_n) \in \text{bounded set of } L^2(Q). \tag{5.37}$$

but

$$z \to \frac{\partial z}{\partial t} - \frac{\partial^2 z}{\partial x^2} \text{ is an isomorphism from } Z \to L^2(Q)$$

where

$$Z = \{z \mid z \in H^{2,1}(Q), \quad z_{|\Sigma} = 0, \quad z(0) = 0\} \tag{5.38}$$

so that

$$z_n \in \text{bounded set of } Z \tag{5.39}$$

$$v_n \in \text{bounded set of } (L^2(0,T;H^{1/2}(\Gamma))^2 = (L^2(0,T))^2 \tag{5.40}$$

iv) *Taking the limit*

We can extract a subsequence, always denoted v_n, and such that

$$v_n \to v \text{ in } (L^\infty(0,T))^2 \text{ weak star} \tag{5.41}$$

$$i_n = i(v_n) \to i(v) = i \text{ in } L^2(Q) \text{ strongly and a.e.} \tag{5.42}$$

$$y_n - \Phi = y(v_n) - \Phi \to y(v) - \Phi = y - \Phi \text{ in } L^2(Q) \text{ strongly and a.e.} \tag{5.43}$$

(this last point results from $F(y_n,i_n) \to F(y,i)$ in $L^2(Q)$ strongly).

$$G_n \to G \text{ in } L^2(Q) \text{ strongly} \tag{5.44}$$

$$z_n \to z \text{ in } Z \text{ strongly} \tag{5.45}$$

$$\left(t^\gamma \frac{\partial y_n}{\partial x}(0,t), t^\gamma \frac{\partial y_n}{\partial x}(1,t)\right) \to \left(t^\gamma \frac{\partial y}{\partial x}(0,t), t^\gamma \frac{\partial y}{\partial x}(1,t)\right) \text{ in } (L^2(0,T))^2 \text{ strongly} \tag{5.46}$$

$$J(v_n) \to J(v) \tag{5.47}$$

v_n being a minimizing sequence, v is an optimal control

$$J(v) = \inf_{w \in \mathscr{U}_{ad}} J(w) \quad \square \tag{5.48}$$

b) *Equations giving the gradient*

We are going to prove the

Theorem 5.1. *The system being governed by the equations (5.1), the applications $v \to y(v)$ from $(L^\infty(0,T))^2$ to $L^2(0,T;H^1(\Omega))$ and $v \to i(v)$ from $(L^\infty(0,T))^2$ to $L^2(Q)$ are Gateaux-differentiable. If we call $\delta = (y'(v),\phi)$, $\epsilon = (i'(v),\phi)$ $(\phi \in (L^\infty(0,T))^2)$ then δ and ϵ are such that*

$$\begin{cases} \delta \in L^2(0,T;H_0^1(\Omega)) & (5.49) \\ \frac{\partial \delta}{\partial t} + A\delta + F_y'(y,i)\delta + F_i'(y,i)\epsilon = 0 & (5.50) \\ \delta(0) = 0 & (5.51) \end{cases}$$

$$\begin{cases} \epsilon \in L^2(0,T;H^\theta(\Omega)), \quad \theta < \frac{1}{2} & (5.52) \\ \frac{\partial \epsilon}{\partial t} + A\epsilon = 0 & (5.53) \\ \epsilon_{|\Sigma} = \phi & (5.54) \\ \epsilon(0) = 0 & (5.55) \end{cases}$$

Remark 5.1. $A = -\frac{d^2}{dx^2}$ and $F(y,i) = \sigma y/(1+y+i)$ but the theorem is true for A second order elliptic operator, F Lipschitz-continuous with respect to y and i, increasing with y, continuously differentiable with respect to y and i.

Proof of the theorem 5.1. Let us call

$$\delta_\theta = \frac{1}{\theta}(y(v+\theta\phi) - y(v)), \qquad \epsilon_\theta = \frac{1}{\theta}(i(v+\theta\phi) - i(v)). \tag{5.56}$$

We have at once (5.52)-(5.55), and $\epsilon_\theta = \epsilon$ is independent of θ. We have in plus

$$\|\epsilon\|_{L^2(Q)} \le C\left(\|v_0\|_{L^\infty(0,T)} + \|v_1\|_{L^\infty(0,T)}\right) \tag{5.57}$$

We deduce from (5.1)

$$\frac{\partial \delta_\theta}{\partial t} + A\delta_\theta + X_\theta = Y_\theta \tag{5.58}$$

where

$$\begin{cases} X_\theta = \dfrac{1}{\theta}\left(F(y(v+\theta\phi),\, i+\theta\epsilon) - F(y, i+\theta\epsilon)\right) \\ Y_\theta = -\dfrac{1}{\theta}\left(F(y, i+\theta\epsilon) - F(y,i)\right) \qquad (y(v) = y,\ i(v) = i) \end{cases}$$

In (5.58) we do the scalar product with δ_θ

$$\begin{cases} \dfrac{1}{2}\dfrac{d}{dt}\,|\delta_\theta|^2 + a(\delta_\theta) + (\text{term} \geq 0) \leq L|\epsilon|\,|\delta_\theta| < \dfrac{L}{2}\left(|\epsilon|^2 + |\delta_\theta|^2\right) \\ \frac12\,|\delta_\theta(t)|^2 + \displaystyle\int_0^t a(\delta_\theta(\tau))d\tau < \frac{L}{2}\,\|\epsilon\|^2_{L^2(Q)} + \frac{L}{2}\int_0^t |\delta_\theta(\tau)|^2\, d\tau \end{cases}$$

Taking account of (5.57) we have

$$\delta_\theta \in \text{bounded set of } L^\infty(0,T;L^2(\Omega)) \tag{5.59}$$

$$\delta_\theta \in \text{bounded set of } L^2(0,T;H_0^1(\Omega)) \tag{5.60}$$

$$X_\theta \in \text{bounded set of } L^\infty(0,T;L^2(\Omega)) \tag{5.61}$$

$$Y_\theta \in \text{bounded set of } L^\infty(0,T;L^2(\Omega)) \tag{5.62}$$

(these two last properties because F is Lipschitz-continuous and because of (5.59)).

$$\frac{\partial \delta_\theta}{\partial t} \in \text{bounded set of } L^2(0,T;H^{-1}(\Omega)) \tag{5.63}$$

Now, let θ_n be a sequence of positive numbers converging towards 0. We can extract a subsequence, always called θ_n, such that

$$\delta_{\theta_n} \to \delta \text{ in } L^2(Q) \text{ strongly and a.e.} \tag{5.64}$$

$$\delta_{\theta_n} \rightharpoonup \delta \text{ in } L^\infty(0,T;L^2(\Omega)) \text{ weak star} \tag{5.65}$$

$$\delta_{\theta_n} \rightharpoonup \delta \text{ in } L^2(0,T;H_0^1(\Omega)) \text{ weakly} \tag{5.66}$$

$$X_{\theta_n} \rightharpoonup F_y'(y,i)\delta \text{ in } L^2(Q) \text{ weakly} \tag{5.67}$$

$$Y_{\theta_n} \rightharpoonup -F_i'\,\epsilon \text{ in } L^2(Q) \text{ weakly} \tag{5.68}$$

and we have (5.50) for δ.

Because of the unicity of a solution for (5.49)–(5.51), the whole initial sequence δ_{θ_n} is converging, and that for every sequence θ_n converging to 0. □

c) *Adjoint system. Necessary condition of optimality.*

u is an optimal control $\Rightarrow (J'(u), v-u) \geq 0 \quad \forall v \in \mathcal{U}_{ad}$ where $(\,,\,)$ means the scalar product in $(L^2(0,T))^2$.

$$\frac{1}{2}(J'(v),\phi) = \int_0^T t^{2\gamma}\left[\left(-\frac{\partial y}{\partial x}(0,t) - z_d^0\right)\left(-\frac{\partial \delta}{\partial x}(0,t)\right) + \left(\frac{\partial y}{\partial x}(1,t) - z_d^1\right)\frac{\partial \delta}{\partial x}(1,t)\right]dt \tag{5.69}$$

Let us define the adjoint state by

$$-\frac{\partial p}{\partial t} + A^*p + F_p'p = 0 \qquad p_{|\Sigma} = t^{2\gamma}\left(\frac{\partial y}{\partial \nu_A} - z_d\right), \qquad p(T) = 0 \tag{5.70}$$

$$-\frac{\partial q}{\partial t} + A^*q + F_i'p = 0 \qquad q_{|\Sigma} = 0, \qquad q(T) = 0 \tag{5.71}$$

Let us write the Green's formula

$$\begin{aligned} 0 &= \int_Q p\left(\frac{\partial \delta}{\partial t} + A\delta + F_y'\delta + F_i'\epsilon\right)dxdt - \int_Q \delta\left(-\frac{\partial p}{\partial t} + A^*p + F_y'p\right)dxdt \\ &= \int_\Omega \left(p(T)\delta(T) - p(0)\delta(0)\right)dx - \int_\Sigma p\,\frac{\partial \delta}{\partial \nu_A}\,d\Sigma + \int_\Sigma \delta\,\frac{\partial p}{\partial \nu_{A^*}}\,d\Sigma \\ &+ \int_Q pF_i'\epsilon\, dxdt = -\int_\Sigma t^{2\gamma}\left(\frac{\partial y}{\partial \nu_A} - z_d\right)\frac{\partial \delta}{\partial \nu_A}\,d\Sigma + \int_Q \left(\frac{\partial q}{\partial t} - A^*q\right)\epsilon\, dxdt \\ &= -\frac{1}{2}(J'(v),\phi) + \int_\Omega \left(q(T)\epsilon(T) - q(0)\epsilon(0)\right)dx + \int_\Sigma \epsilon\,\frac{\partial q}{\partial \nu_{A^*}}\,d\Sigma \\ &- \int_\Sigma q\,\frac{\partial \epsilon}{\partial \nu_A}\,d\Sigma = -\frac{1}{2}(J'(v),\phi) + \int_\Sigma \phi\,\frac{\partial q}{\partial \nu_{A^*}}\,d\Sigma \end{aligned}$$

A necessary condition of optimality is then

$$\int_\Sigma \frac{\partial q}{\partial \nu_{A^*}}(v-u)\,d\Sigma \geq 0 \qquad \forall v \in \mathcal{U}_{ad} \tag{5.72}$$

In the precise case of the problem this is equivalent to the *local conditions*

$$-\frac{\partial q}{\partial x}(0,t;u)\,(\xi - u_0(t)) \geq 0, \qquad \frac{\partial q}{\partial x}(1,t;u)\,(\xi - u_1(t)) \geq 0 \tag{5.73}$$

$$\forall \xi, \quad 0 \leq \xi \leq M$$

That is itself equivalent to (5.74)

$$\begin{cases} u_0(t) = P_K\left(u_0(t) + \rho\,\dfrac{\partial q}{\partial x}(0,t;u)\right) \\ u_1(t) = P_K\left(u_1(t) - \rho\,\dfrac{\partial q}{\partial x}(1,t;u)\right) \end{cases} \tag{5.75}$$

where $K = [0,M]$, $\rho > 0$

P_K = projection of $\mathbb{R}$ on K.

Remark 5.2. In general the inhibitor with which the control is made is cheap. However it is not always the case. The functional becomes then

$$J(v) = \int_0^T t^{2l}\left[\left(-\frac{\partial y}{\partial x}(0,t)-z_d^0\right)+\left(\frac{\partial y}{\partial x}(1,t)-z_d^1\right)^2\right]dt + a\int_0^T (|v_0(t)|^2+|v_1(t)|^2)dt \tag{5.76}$$

and a necessary condition of optimality is

$$\int_\Sigma \left(\frac{\partial q}{\partial \nu_{A^*}}+au\right)(v-u)d\Sigma \geq 0 \qquad \forall v \in \mathscr{U}_{ad} \tag{5.77}$$

or, equivalently

$$\begin{cases} u_0(t) = P_K\left(u_0(t)-\rho\left(-\dfrac{\partial q}{\partial x}(0,t;u)+au_0(t)\right)\right) & \text{a.e. } t \\ u_1(t) = P_K\left(u_1(t)-\rho\left(\dfrac{\partial q}{\partial x}(1,t;u)+au_1(t)\right)\right) & \text{a.e. } t \end{cases} \tag{5.78}$$

5.2.2. Optimal Control of the Substrate Concentration at x = 1 in the Active Transport System. *The existence of at least one optimal control* can be proved as in §5.2.1 for the preceding problem.

a) *Equations giving the gradient*

Let us call

$$\begin{cases} \dfrac{d}{d\xi}y_1(v+\xi\phi)_{|\xi=0} = \hat{y}_1 \\ \dfrac{d}{d\xi}y_2(v+\xi\phi)_{|\xi=0} = \hat{y}_2 \\ \dfrac{d}{d\xi}y_3(v+\xi\phi)_{|\xi=0} = \hat{y}_3 \end{cases} \tag{5.79}$$

We can verify, as in §5.2.1, using *a priori* estimations similar to those in §2.3., that

$$(B\hat{y}_1)'+A_1\hat{y}_1+\frac{\partial F}{\partial y_1}\hat{y}_1+\frac{\partial F}{\partial y_2}\hat{y}_2+\frac{\partial F}{\partial y_3}\hat{y}_3 = 0 \tag{5.80}$$

$$\hat{y}_2'+A_2\hat{y}_2-\frac{\partial F}{\partial y_1}\hat{y}_1-\frac{\partial F}{\partial y_2}\hat{y}_2-\frac{\partial F}{\partial y_3}\hat{y}_3 = 0 \tag{5.81}$$

$$\int_Q \hat{y}_3\left(-\frac{\partial q_3}{\partial t}+A_3^*q_3\right)dx\,dt = -\int_{\Sigma_1}\phi\frac{\partial q_3}{\partial \nu_{A_3^*}}d\Sigma \qquad \forall q_3 \in Q_3 \tag{5.82}$$

where Ω, Q, Γ, Σ are defined as usually

$(\Omega =]0,1[, \ldots)$, with $\Gamma = \Gamma_0 \cup \Gamma_1$, $\Gamma_0 = \{0\}$,
$\Gamma_1 = \{1\}$, $\Sigma_i = \Gamma_i \times]0,T[$, $i = 0,1$

We define

$$V = \{v \mid v \in H^1(\Omega),\ v_{|\Gamma_0} = 0\} \tag{5.83}$$

$$(Bu,v) = \int_\Omega uv\,dx + \frac{1}{K}\int_{\Gamma_1}\gamma_0 u\gamma_0 v\,d\Gamma \tag{5.84}$$

B so defined is in $\mathscr{L}(V,V')$.

- *definition of A_1* $(\in \mathscr{L}(V,V'))$

$$(A_1u,v) = a(u,v)\left(=\int_0^1 \frac{\partial u}{\partial x}\frac{\partial v}{\partial x}dx\right) \qquad \forall u,v \in V \tag{5.85}$$

- *definition of A_2*

$$(A_2u,v) = a(u,v) \qquad \forall u,v \in H^1(\Omega) \tag{5.86}$$

- *definition of A_3*

$$(u,A_3^*v) = a(u,v) \qquad \forall u,v \in H_0^1(\Omega) \tag{5.87}$$

- *definition of Q_3*

$$Q_3 = \{q_3 \mid q_3 \in L^2(0,T;H_0^1(\Omega)),\ -q_3'+A_3^*q_3 \in L^2(Q),\ q_3(T) = 0\} \tag{5.88}$$

We have the initial conditions

$$(B\hat{y}_1)(0) = 0 \qquad \hat{y}_2(0) = 0 \tag{5.89}$$

In fact (5.80) and the first equation of (5.89) define $\hat{y}_1$ in

$$Y_1 = \{\hat{y}_1 \mid \hat{y}_1 \in L^2(0,T;V),\ (B\hat{y}_1)'+A\hat{y}_1 \in L^2(Q),\ (B\hat{y}_1)(0) = 0\} \tag{5.90}$$

It is easy to check that Y is a Banach space for the norm

$$\|\hat{y}_1\|_{Y_1} = \|\hat{y}_1\|_{L^2(0,T;V)}+\|(B\hat{y}_1)'+A_1\hat{y}_1\|_{L^2(Q)} \tag{5.91}$$

and that the application

$$L_1 : \hat{y}_1 \to (B\hat{y}_1)'+A_1\hat{y}_1+\frac{\partial F}{\partial y_1}\hat{y}_1 \tag{5.92}$$

is an isomorphism from Y_1 on $L^2(Q)$.

In the same way (5.81) and the second equation (5.89) define $\hat{y}_2$ in

$$Y_2 = \{\hat{y}_2 \mid \hat{y}_2 \in L^2(0,T;H^1(\Omega)),\ \hat{y}_2'+A_2\hat{y}_2 \in L^2(Q),\ \hat{y}_2(0) = 0\} \tag{5.93}$$

and the application

$$L_2 : \hat{y}_2 \to \hat{y}_2'+A_2\hat{y}_2-\frac{\partial F}{\partial y_2}\hat{y}_2 \tag{5.94}$$

is an isomorphism from Y_2 on $L^2(Q)$.

At last (5.82) define a unique $\hat{y}_3$ in $L^2(Q)$.

b) *The adjoint system*

We define the adjoint system (p_1,p_2,p_3) in the following way:

i/ since L_1 is an isomorphism from Y_1 on $L^2(Q)$, L_1^* is an isomorphism from $L^2(Q) = (L^2(Q))'$ on Y_1', and in particular it exists in $L^2(Q)$ a unique function p_1 such that:

$$(L_1^* p_1, \psi) = \int_Q \frac{\partial F}{\partial y_1} p_2 \psi dxdt - 2 \int_{\Sigma_1} (y_1 - z_d)\psi d\Sigma \qquad \forall \psi \in Y_1 \tag{5.95}$$

(5.95) can be written also

$$\int_Q p_1 L_1 \psi dxdt = \int_Q \frac{\partial F}{\partial y_1} p_2 \psi dxdt - 2 \int_{\Sigma_1} (y_1 - z_d)\psi d\Sigma \qquad \forall \psi \in Y_1 \tag{5.96}$$

ii/ in the same way p_2 is the unique element in $L^2(Q)$ such that

$$\int_Q p_2 L_2 \psi dxdt = - \int_Q \frac{\partial F}{\partial y_2} p_1 \psi dxdt \qquad \forall \psi \in Y_2 \tag{5.97}$$

iii/ thirdly p_3 is the unique element in Q_3 such that

$$\begin{cases} -p_3' + A_3^* p_3 + \dfrac{\partial F}{\partial y_3}(p_1 - p_2) = 0 \\ p_3(T) = 0 \end{cases} \tag{5.98}$$

c) *Computation of* $\hat{J} = \dfrac{d}{d\xi} J(v + \xi\phi)_{|\xi=0}$

Using (5.80) and (5.85) we get

$$\begin{aligned} 0 &= \int_Q p_1 \left(L_1 \hat{y}_1 + \frac{\partial F}{\partial y_2} \hat{y}_2 + \frac{\partial F}{\partial y_3} \hat{y}_3\right) dxdt \\ &- \left[\int_Q p_1 L_1 \hat{y}_1 dxdt - \int_Q \frac{\partial F}{\partial y_1} p_2 \hat{y}_1 dxdt + 2 \int_{\Sigma_1} (y_1 - z_d)\, \hat{y}_1 \, d\Sigma\right] \\ &= \int_Q \left(p_1 \frac{\partial F}{\partial y_2} \hat{y}_2 + p_1 \frac{\partial F}{\partial y_3} \hat{y}_3 + \frac{\partial F}{\partial y_1} p_2 \hat{y}_1\right) dxdt - \hat{J} \end{aligned} \tag{5.99}$$

In the same way, using (5.81) and (5.96)

$$\begin{aligned} 0 &= \int_Q p_2 \left(L_2 \hat{y}_2 - \frac{\partial F}{\partial y_1} \hat{y}_1 - \frac{\partial F}{\partial y_3} \hat{y}_3\right) dxdt \\ &- \int_Q p_2 L_2 \hat{y}_2 dxdt + \int_Q \frac{\partial F}{\partial y_2} p_1 \hat{y}_2 dxdt \\ &= \int_Q \left(-p_2 \frac{\partial F}{\partial y_1} \hat{y}_1 - p_2 \frac{\partial F}{\partial y_3} \hat{y}_3 - \frac{\partial F}{\partial y_2} p_1 \hat{y}_2\right) dxdt \end{aligned} \tag{5.100}$$

Adding (5.99) and (5.100) we get

$$\hat{J} = \int_Q \hat{y}_3 \frac{\partial F}{\partial y_3}(p_1 - p_2)\, dxdt \tag{5.101}$$

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But from (5.101), (5.98) and (5.82)

$$\hat{J} = - \int_Q \hat{y}_3(-p_3' + A_3^* p_3) dxdt = \int_{\Sigma_1} \phi \frac{\partial p_3}{\partial \gamma_{A_3^*}} d\Sigma \tag{5.102}$$

Whence the result

$$\frac{d}{d\xi} J(v + \xi\phi)_{|\xi=0} = \int_{\Sigma_1} \frac{\partial p_3}{\partial \gamma_{A_3^*}} \phi d\Sigma \tag{5.103}$$

which can be expressed

$$J'(v) = \frac{\partial p_3}{\partial \gamma_{A_3^*}} \tag{5.104}$$

d) *A necessary condition of optimality and the associated algorithm of research of the optimal control (simple gradient algorithm)*

A necessary condition of optimality for u is

$$(J'(u), v - u) \geq 0 \qquad \forall v \in \mathscr{U}_{ad} \tag{5.105}$$

or, which is equivalent

$$u = P_{\mathscr{U}_{ad}}(u - \rho J'(u)), \qquad \rho > 0 \tag{5.106}$$

$$P_{\mathscr{U}_{ad}} = \text{projection on the convex } \mathscr{U}_{ad} \tag{5.107}$$

We deduce from (5.106) the algorithm

$$u^{(0)} \text{ given in } \mathscr{U}_{ad}, \; u^{n+1} = P_{\mathscr{U}_{ad}}(u^n - \rho J'(u^n)). \tag{5.108}$$

Remark 5.3. It is easy to prove existence and uniqueness of (p_1, p_2) defined by (5.96), (5.97):

i/ We define the application

$$A : (\psi_1, \psi_2) \to \left(L_1 \psi_1 + \frac{\partial F}{\partial y_2} \psi_2, L_2 \psi_2 - \frac{\partial F}{\partial y_1} \psi_1\right) \tag{5.109}$$

from

$$Y_1 \times Y_2 \to L^2(Q) \times L^2(Q).$$

ii/ It is an isomorphism. Indeed, f_1 and f_2 being given in $L^2(Q)$, there exists (ψ_1, ψ_2), unique in $Y_1 \times Y_2$, such that

$$\left(\frac{d}{dt}(B\psi_1), v_1\right) + \left(\frac{d}{dt}\psi_2, v_2\right) + a_1(\psi_1, v_1; \psi_2, v_2) = \int_\Omega (f_1 v_1 + f_2 v_2) dx \tag{5.110}$$

$$\forall (v_1, v_2) \in (H^1(\Omega))^2$$

$$\begin{aligned} a_1(\psi_1, v_1; \psi_2, v_2) &= \int_\Omega (\text{grad}\psi_1 \, \text{grad} v_1 + \text{grad}\psi_2 \, \text{grad} v_2) dx \\ &+ \int_\Omega \left(\frac{\partial F}{\partial y_1} \psi_1 v_1 + \frac{\partial F}{\partial y_2} \psi_2 v_1 - \frac{\partial F}{\partial y_2} \psi_2 v_2 - \frac{\partial F}{\partial y_1} \psi_1 v_2\right) dx \end{aligned} \tag{5.111}$$

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Using

$$0 \le \frac{\partial F}{\partial y_1} < \sigma, \qquad -\sigma < \frac{\partial F}{\partial y_2} < 0$$

We have

$$a_1(v_1,v_1;v_2,v_2) \ge \int_\Omega (|\mathrm{grad}\, v_1|^2 + |\mathrm{grad}\, v_2|^2)dx - \sigma \int_\Omega (|v_1|^2 + |v_2|^2)dx \tag{5.112}$$

iii) Consequently there exists a unique couple (p_1,p_2) such that

$$\int_Q \left[p_1\left(L_1\psi_1 + \frac{\partial F}{\partial y_2}\psi_2\right) + p_2\left(L_2\psi_2 - \frac{\partial F}{\partial y_1}\psi_1\right)\right] dx\,dt = -(J'_y,\psi_1) \tag{5.113}$$

$$\forall (\psi_1,\psi_2) \quad Y_1 \times Y_2$$

which is equivalent to (5.96), (5.97).

5.3. *Numerical Methods and Results.*

5.3.1. Optimal Control. (Substrate flux control by inhibitor).

Numerical Algorithm

First step: Assuming u^n to be known, we define y^{n+1} and i^{n+1} by the solution of

$$\partial y^{n+1}/\partial t - \partial^2 y^{n+1}/\partial x^2 + F(y^{n+1}, i^{n+1}) = 0$$

$$\partial i^{n+1}/\partial t - \partial^2 i^{n+1}/\partial x^2 = 0$$

$$y^{n+1}(0,t) = \alpha, \quad y^{n+1}(1,t) = \beta$$

$$i^{n+1}(0,t) = u_0^n(t), \quad i^{n+1}(1,t) = u_1^n(t),$$

$$y^{n+1}(x,0) = 0, \quad i^{n+1}(x,0) = 0$$

Second step: We define next p^{n+1} and q^{n+1} by the solution of

$$-\partial p^{n+1}/\partial t - \partial^2 p^{n+1}/\partial x^2 + \frac{\partial F}{\partial y}(y^{n+1}, i^{n+1})p^{n+1} = 0$$

$$-\partial q^{n+1}/\partial t - \partial^2 q^{n+1}/\partial x^2 + \frac{\partial F}{\partial i}(y^{n+1}, i^{n+1})q^{n+1} = 0$$

$$p^{n+1}(0,t) = 2\left[-\frac{\partial y^{n+1}}{\partial x}(0,t) - z_d^0(t)\right]$$

$$p^{n+1}(1,t) = 2\left[\frac{\partial y^{n+1}}{\partial x}(1,t) - z_d^1(t)\right]$$

$$q^{n+1}(0,\cdot) = 0, \quad q^{n+1}(1,t) = 0$$

$$p^{n+1}(x,T) = 0, \quad q^{n+1}(x,T) = 0$$

Of course, each step is solved by numerical approximation. We define the new value of u by

$$u_0^{n+1}(t) = P_K\left(u_0^n(t) + \rho \frac{\partial q^{n+1}}{\partial x}(0,t)\right)$$

$$u_1^{n+1}(t) = P_K\left(u_1^n(t) - \rho \frac{\partial q^{n+1}}{\partial x}(1,t)\right)$$

where P_K = projection of R on $K = [0,M]$.

Numerical Results

We solved using the explicit method, taking the following values: $\Delta x = 0.1$ = space mesh size, $\Delta t = 0.004$ = time mesh size, $T = 120\,\Delta t$, $\sigma = 36$, $\alpha = \beta = 1$, $M = 12$, z_d obtained by

$$z_d(t) = \frac{\partial y}{\partial x}(1,t) = -\frac{\partial y}{\partial x}(0,t)$$

for $v_0(t) = v_1(t) = 6$; initial estimation of v: $v_0^0(t) = v_1^0(t) = 12$. When ρ is properly chosen the results are very satisfactory.

Figure 18 shows the value of the cost function J_9 at the iteration number 9, according to ρ. The initial value of J was $J_0 = 940 \times 10^{-4}$ and, for the best value of p, which is 15×10^3, $J_9 = 3 \times 10^{-4}$.

Table 1 shows for $p = 15 \times 10^3$, the decreasing of J and of the gradient

$$G = \left(\int_0^T \left|\frac{\partial q}{\partial x}(1,t)\right|^2 dt\right)^{1/2}$$

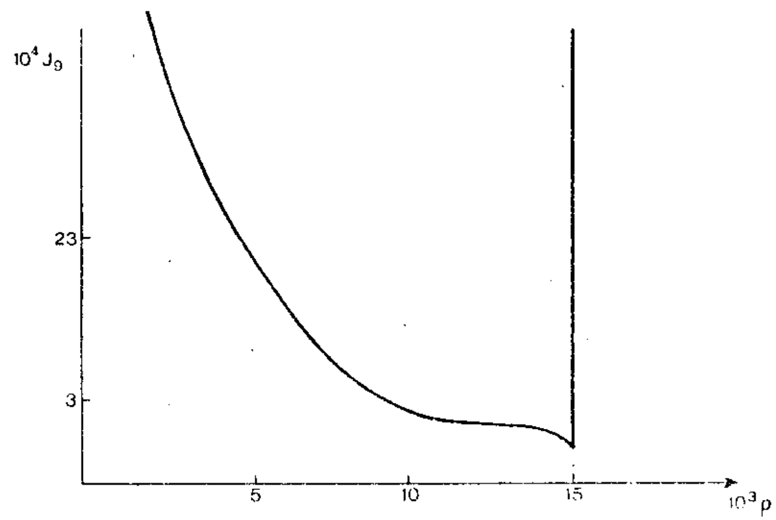

Figure 18. J_9 as a function of $\rho (J^0 = 940 \times 10^{-4})$.

Table 1. Cost Function J and Gradient G.

Iteration number	Cost function	Gradient
0	0.075 997	0.001 952
5	0.000 829	0.000 135
10	0.000 263	0.000 064
20	0.000 079	0.000 030
30	0.000 037	0.000 017
50	0.000 013	0.000 007
75	0.000 006	0.000 003
99	0.000 003	0.000 002

Conclusion

This work is the result of a collaboration between Biochemists and Mathematicians. The above experimental systems are performed in the "E.R.A. N° 338 du C.N.R.S.—Université de Technologie de Compiègne". The discussion about the interest of these systems at both fundamental and applied points of view can be found in Thomas and Caplan (1974).

At the mathematical and numerical point of view it is possible to get more details from: Kernevez (1972) for a general survey of the mathematical problems, Kernevez *et al.* (1973) for a stochastic feedback control, Brauner and Penel (1972) for systems without michaelian assumption, Dubus (1972) for a multienzyme system and Yvon (1973) for optimal control of systems governed by variational inequalities.

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Hysteresis, Oscillations, and Pattern Formation in Realistic Immobilized Enzyme Systems

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Summary. Hysteresis, oscillations, and pattern formation in realistic biochemical systems governed by P.D.E.s are considered from both numerical and mathematical points of view. Analysis of multiple steady states in the case of hysteresis, and bifurcation theory in the cases of oscillations and pattern formation, account for the observed numerical results. The possibility to realize these systems experimentally is their main interest, thus bringing further arguments in favor of theories explaining basic biological phenomena by diffusion and reaction.

Key words: Immobilized enzyme systems—Diffusion-reaction—Hysteresis, oscillations—Pattern formation

1. Introduction

The aim of this paper is to consider some immobilized enzyme systems with hysteresis, oscillations, or pattern formation, which are simple, can be realized experimentally, and which illustrate the theories explaining short term memory, biological clocks, or morphogenesis by diffusion and reaction phenomena.

a) Hysteresis. It has been suggested that hysteresis effects in biochemical systems are adequate to account for short-term memory (Changeux and Thiery, 1968; Katchalsky and Oplatka, 1966; Katchalsky and Spangler, 1968).

Such an hysteresis effect was found experimentally (Naparstek, Romette, Kernevez, and Thomas, 1974) in an uricase system governed by the following P.D.E.

$$s_t - s_{xx} + \sigma s/(1 + s + ks^2) = 0, \quad 0 < x < 1, \qquad (1)$$
$$s(0, t) = s(1, t) = s_0.$$

In Section 2 we find the numerical solution of the initial boundary value problem (1) for a succession of values s_0 and use at each step the steady state of the preceding step as a starting value. In a first phase, s_0 is increasing, then in a second phase it is decreasing. For a given s_0, the steady states in each phase are found to be distinct.

These numerical results are explained by a mathematical analysis of the solutions of the ordinary differential equation boundary value problem:

$$-s'' + \sigma s/(1 + s + ks^2) = 0, \quad 0 < x < 1, \qquad s(0) = s(1) = s_0. \tag{2}$$

b) Oscillations. From the biological point of view, sustained oscillations have been experimentally observed and established beyond doubt for glycolysis (Hess, 1962; Betz and Chance, 1965).

Models to represent the observed oscillations have been worked out (Sel'kov, 1968; Higgins, 1964). A model taking explicitly into account the allosteric effects which is free from phenomenological factors has been constructed (Goldbeter and Lefever, 1972). Oscillation phenomena were already experimentally produced with artificial membranes (Naparstek, Thomas, and Caplan, 1973).

The system proposed in Section 3 is still simpler:

$$\begin{aligned} &s_t - s_{xx} + \sigma as/(1 + s + ks^2) = 0, \quad 0 < x < 1, t > 0 \\ &a_t - \beta a_{xx} + \sigma as/(1 + s + ks^2) = 0 \\ &s(0, t) = s(1, t) = s_0; \qquad a(0, t) = a(1, t) = a_0. \end{aligned} \tag{3}$$

A bifurcation analysis explains, at least in a neighborhood of the bifurcation points, the oscillatory behavior obtained by numerical simulations.

c) Pattern Formation. It has been suggested that pattern formation in biochemical systems is adequate to account for the morphogenesis of a developing tissue (Turing, 1952; Gmitro and Scriven, 1966; Meinhardt, 1977; Nicolis and Prigogine, 1977; Kauffman, Shymko, and Trabert, 1978). In Section 4 we present an immobilized enzyme system in which a sequence of patterns arises. It is governed by two coupled P.D.E.s:

$$\begin{aligned} &s_t - \Delta s + \lambda(F(s, a) - (s_0 - s)) = 0 \\ &a_t - \beta\,\Delta a + \lambda(F(s, a) - \alpha(a_0 - a)) = 0 \\ &\text{with no-flux boundary conditions} \\ &\text{and } F(s, a) = \rho as/(1 + s + ks^2). \end{aligned} \tag{4}$$

We give numerical results for simulations in one and two dimensions. We solve the initial boundary value problem (4) for a succession of values λ, starting at each step from the slightly perturbed steady state of the preceding step. Sequential pattern formation is observed, in accordance with a linear stability analysis. A non-linear stability analysis using bifurcation theory and referring to earlier work (Boa and Cohen, 1976; Meurant and Saut, 1977) explains these numerical results, at least in certain neighborhoods of the bifurcation points.

Our systems are artificial membranes where the uricase enzyme is immobilized by polymerization with glutaraldehyde (Thomas, 1976). The uricase enzyme is a catalyst of the reaction:

uric acid + oxygen → allantoïn + other products.

The reaction rate is:

$$J_R = [V_M A/(K_A + A)]S/(K_M + S + S^2/K_{SS}), \tag{5}$$

S and A denoting respectively uric acid and oxygen concentrations.

Within the active membrane, substrate S and cosubstrate A diffuse (diffusion coefficients D_S and D_A) and react under the catalytic action of the immobilized enzyme.

In the model discussed in Section 2, A is in excess and consequently we can take the reaction rate:

$$J_R = V_M S/(K_M + S + S^2/K_{SS}). \tag{6}$$

In the other models (Sections 3 and 4), A is small with respect to K_A and we can take:

$$J_R = (V_M/K_A)AS/(K_M + S + S^2/K_{SS}). \tag{7}$$

2. Hysteresis

The enzyme membrane separates two reservoirs where A is in excess and S is at a fixed concentration S_0. A and S diffuse in the membrane and within it react together, the reaction rate being given by (6). The system evolution is governed by (1) if we take as units for concentration, length, and time, K_M, L (thickness of the membrane), and θ ($\theta = L^2/D_S$, D_S diffusion coefficient of S) respectively. And finally $\sigma = \theta V_M/K_M$, $k = K_M/K_{SS}$.

It is important to note that by using dimensionless quantities s, x, t the only significant parameter is σ. σ comprises the membrane diffusion time θ and the enzyme reaction characteristic time K_M/V_M.

Numerical simulations were performed following the procedure indicated in the first section. For s_0 values first increasing, then decreasing, we found the succession of profiles reported in Figure 1.

These profiles obey Eq. (2).

Before proving the possibility of multiple solutions for (2), let us see, using a simpler system with the same qualitative behavior, why multiple steady states may appear when diffusion is coupled to substrate inhibited kinetics.

This simpler system can be viewed as an inactive membrane (i.e. without enzyme) separating an outside reservoir with fixed concentration s_0 and a well-stirred reactor with concentration s. At the steady state, s is a solution of the algebraic equation:

$$s_0 - s = \rho s/(1 + s + ks^2). \tag{8}$$

The state s of the system is a (multi-valued) function of the two parameters s_0 and ρ and is represented by a catastrophe surface as shown in Figure 2.

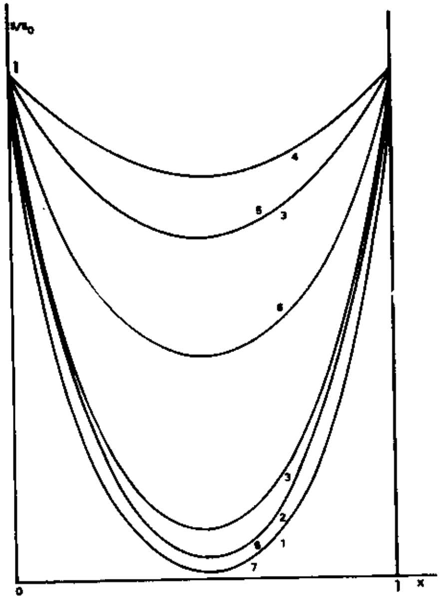

Fig. 1. The profiles 1–2–3–3–4–5–6–6–7 represent s/s_0 as a function of x. Low profiles 1–2–3 correspond to increasing s_0 values, until a jump to the high profile 3. High profiles 4–5–6 correspond to decreasing s_0 values, until a jump to the low profile 6. $\sigma = 1200$, profiles 3 and 6 correspond to $s_0 \simeq 68$, $s_0 \simeq 61$ respectively

Suppose that ρ is kept fixed and that s_0 varies, first increasing, then decreasing. The representative point on the surface follows a path 1–2–3–3–4–5–6–6–7 with jumps at points 3 and 6. For s_0 in a suitable range there exist two stable states. The state in which the system lies depends in fact upon its past history. There is an hysteresis effect.

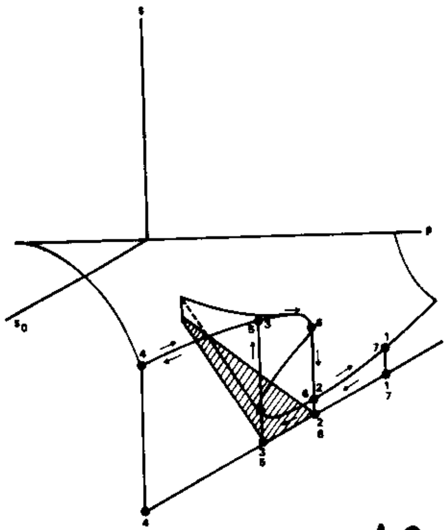

Fig. 2. Path followed by the point (s_0, ρ, s) on the catastrophe surface $s_0 - s = \rho s/(1 + s + ks^2)$ when ρ is kept fixed and s_0 is varying as in Figure 1

Coming back to the distributed system (2), a first integral for (2) is:

$$-s'^2(x) + 2\sigma G(s(x)) = 2\sigma G(\mu), \quad 0 < x < 1$$

where G is some primitive function of F and μ $(= s(1/2))$ is the minimum of $s(x)$ for $x \in (0, 1)$.

$$s'(x) = (2\sigma)^{1/2}(G(s(x)) - G(\mu))^{1/2} \quad \text{for } x \in (1/2, 1)$$

$$(2\sigma)^{1/2}(x - 1/2) = \int_\mu^{s(x)} \frac{d\xi}{(G(\xi) - G(\mu))^{1/2}} \tag{9}$$

and the condition $s(1) = s_0$ is equivalent to:

$$\left(\frac{\sigma}{2}\right)^{1/2} = \int_\mu^{s_0} \frac{d\xi}{(G(\xi) - G(\mu))^{1/2}} \tag{10}$$

(9) defines a solution $s(x)$ if μ is a solution of:

$$f(\mu) = (\sigma/2)^{1/2}, \tag{11}$$

where

$$f(\mu) = \int_\mu^{s_0} \frac{d\xi}{(G(\xi) - G(\mu))^{1/2}} = \int_0^{s_0-\mu} \frac{d\eta}{(G(\mu + \eta) - G(\mu))^{1/2}} \tag{12}$$

$$f'(\mu) = \frac{-1}{(G(s_0) - G(\mu))^{1/2}} - \frac{1}{2}\int_\mu^{s_0} \frac{F(\xi) - F(\mu)}{(G(\xi) - G(\mu))^{3/2}}\, d\xi. \tag{13}$$

Our purpose is to show that we can find s_0 and σ such that (10) admits at least 3 solutions.

Let us choose μ^* such that $F'(\mu^*) < 0$ $(\mu^* > 0)$. When $s_0 \to +\infty$ the first term in (13) tends towards 0 and the second one towards a limit $l < 0$. Let us take s_0 large enough to have $f'(\mu^*) > 0$. For such an s_0 the graph of f looks like (c) in Figure 3. (It is easy to check that $f(\mu) \to +\infty$ when $\mu \to 0+$ and $f(\mu) \to 0$ when $\mu \to s_0$.)

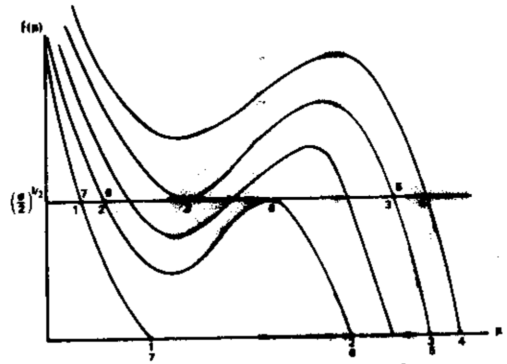

Fig. 3. Sketch of function $f(\mu)$ for s_0 values similar to those in Figure 1, and, for given value of σ, determination of the corresponding values of μ

If we take a value of σ for which (11) can have 3 solutions, and assume that s_0 is varying continuously, first increasing, then decreasing, then we again find a path 1–2–3–3–4–5–6–6–7 with two jumps (Fig. 3), and a range of values s_0 for which there are multiple steady states. In fact, from Sattinger (1972) we know that the minimal and maximal steady states are stable. Thus the profiles of Figure 1 are explained.

3. Oscillations

Equations (3) come from the modelling of an enzyme membrane separating two compartments which contain S and A. These substrates diffuse throughout the membrane and react because of E. The reaction rate to be taken is (7). By using the same dimensionless quantities as in Section 2 for concentrations, space and time, we find equations (3), where $\beta = D_A/D_S$ is the ratio of A and S diffusion coefficients in the membrane. s and a boundary values are fixed.

In order to simplify the analysis we choose s_0 and a_0 such that $s_0 = \beta a_0$.

As a consequence Eqs. (3) admit a trivial steady state solution, $(\tilde{s}, \tilde{a})$, defined by:

$$\begin{aligned} & -\tilde{s}''(x) + (\sigma/\beta)\tilde{s}^2(x)/(1 + \tilde{s}(x) + k\tilde{s}^2(x)) = 0, \quad 0 < x < 1 \\ & \tilde{s}(0) = \tilde{s}(1) = s_0 \\ & \tilde{a}(x) = \tilde{s}(x)/\beta. \end{aligned} \tag{14}$$

Linearization of (2) around $(\tilde{s}, \tilde{a})$ gives a linear system obtained by substituting $s = \tilde{s} + u$, $a = \tilde{a} + v$, and retaining only terms up to first order in u and v in a Taylor expansion of $F(s, a)$:

$$\begin{aligned} & u_t - u_{xx} + \sigma F_s u + \sigma F_a v = 0 \\ & v_t - \beta v_{xx} + \sigma F_s u + \sigma F_a v = 0 \\ & u(0, t) = u(1, t) = v(0, t) = v(1, t) = 0 \end{aligned} \tag{15}$$

where F_s and F_a are defined by:

$$\begin{aligned} & F(s, a) = as/(1 + s + ks^2) \\ & F_s(x) = \frac{\partial F}{\partial s}(s(x), a(x)), \qquad F_a(x) = \frac{\partial F}{\partial a}(s(x), a(x)). \end{aligned} \tag{16}$$

In a condensed form (15) can be written:

$$\frac{dU}{dt} + L_\sigma U = 0 \tag{17}$$

if we define:

$$U = \begin{bmatrix} u \\ v \end{bmatrix}, \qquad L_\sigma U = \begin{bmatrix} -u_{xx} + \sigma F_s u + \sigma F_a v \\ -\beta v_{xx} + \sigma F_s u + \sigma F_a v \end{bmatrix} \tag{18}$$

whereas (3) can be written:

$$\frac{dU}{dt} + L_\sigma U + M_\sigma(U) = 0 \tag{19}$$

with:

$$M_\sigma(U) = \sigma[F(\tilde{s} + u, \tilde{a} + v) - F(\tilde{s}, \tilde{a}) - F_s u - F_a v]\begin{bmatrix} 1 \\ 1 \end{bmatrix}. \tag{20}$$

For the parameter values indicated in Figure 4 the leading eigenvalues of L_σ are complex conjugate. The plot of these eigenvalues is given in Figure 4, where it can be seen that for 2 critical values of σ, σ_1, and σ_2, the spectrum of L_σ crosses the imaginary axis by the two purely imaginary simple eigenvalues, the remaining part of the spectrum lying in the half space $Re z > 0$. In that situation there is an exchange of stability between the trivial steady state $(\tilde{s}, \tilde{a})$ and a periodic bifurcated solution (Iooss, 1973; Meurant and Saut, 1977). More precisely the trivial steady state is stable when the spectrum of L_σ lies in the half space $Re z > 0$ and unstable when at least one eigenvalue lies in the left half plane $Re z < 0$. Moreover in a right (resp. left) neighborhood of the bifurcation point σ_1 (resp. σ_2) a family of bifurcated stable periodic solutions is defined. In fact from the numerical simulations it appears that these oscillations exist in the whole interval (σ_1, σ_2).

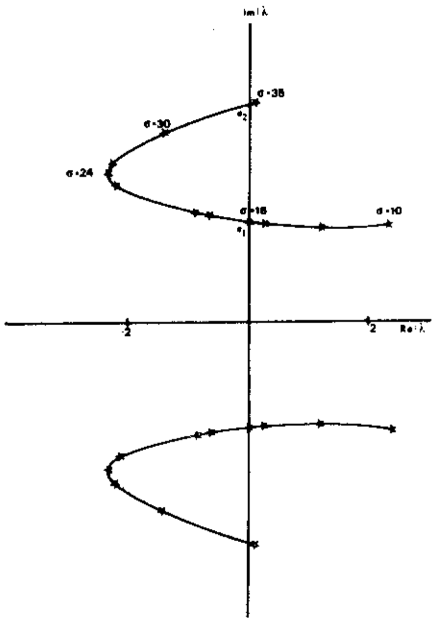

Fig. 4. Leading eigenvalues of L_σ for σ varying from 10 to 35, $s_0 = 100$, $a_0 = 500$, $\beta = 0.2$, $k = 0.1$

4.1. *Modelling of the Uricase System with an Inactive Layer*

A coating in a Petri dish (Fig. 5) is a superposition of 2 layers: the inner one is an active layer, with a thickness L_1 of about 50 μ ($L_1 = 5\cdot10^{-3}$ cm), which is in fact an enzyme membrane as previously described in Section 1, and is stuck on the Petri dish. Just above is spread an inactive layer with possibly a lipido proteic layer. This inactive layer (thickness L_2) is in contact with a well stirred solution in a reservoir with fixed concentrations of substrate S_0 and cosubstrate A_0.

The enzyme membrane can be considered as a 2-dimensional region Ω where s and a, the normalized S and A concentrations, ($s = S/K_M$, $a = A/K_M$), functions of space $r = (x, y)$, and time t, are governed by Eqs. (4). The space coordinates x and y are measured with the Petri dish diameter L, as a unit of length.

α (resp. β) is the ratio of A and S diffusion coefficients in the inactive (resp. active) layer:

$$\alpha = D'_A/D'_s, \qquad \beta = D_A/D_S.$$

The time unit θ is the diffusion time of S in Ω:

$$\theta = L^2/D_S.$$

Two characteristic times, plus θ, play a role in the definition of ρ and λ: $\theta'_S = L_1L_2/D'_S$, characteristic of diffusion transport through the inactive layer from the reservoir to the active membrane, and K_A/V_M, characteristic of reaction in the enzyme membrane Ω: ρ and λ are now defined:

$$\rho = \theta'_S/(K_A/V_M) \quad \text{and} \quad \lambda = \theta/\theta'_S.$$

It is interesting to note that λ, which will be in the following a bifurcation parameter, is proportional to L^2, and therefore to the area of the coating, and to D'_S.

The insertion of a lipido proteic layer within the inactive layer acts as a barrier and lowers D'_S. On the other hand by using a larger Petri dish L^2 is increased.

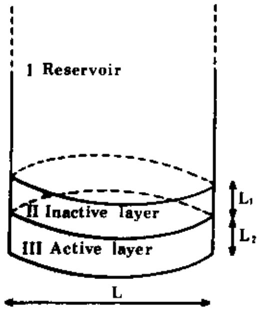

Fig. 5. Substrates S and A diffuse through II from I to III where they react and diffuse

4.2. *Numerical Experiments and Results*

Two series of numerical experiments were carried out:

4.2.1. *1-Dimensional Case.* In the case of a ribbon-like coating s and a are functions of only one space coordinate x so that in Eqs. (4) $\Omega =]0, 1[$, $\Delta = \partial^2/\partial x^2$, and the no flux boundary conditions are $\partial s/\partial x = \partial a/\partial x = 0$ for $x = 0$ and $x = 1$. By using the numerical procedure outlined in Section 1 we found the steady-state profiles indicated in Figure 6.

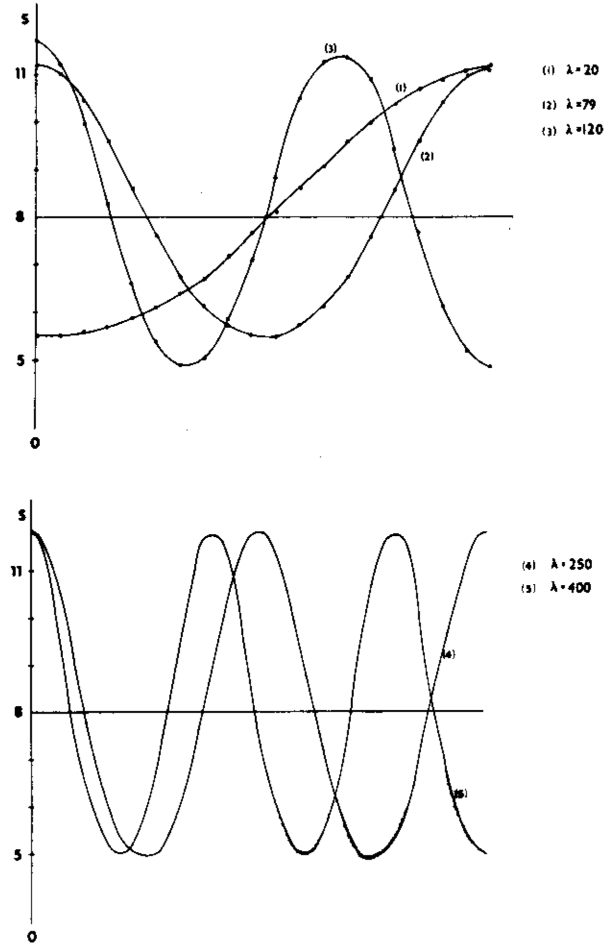

Fig. 6. Steady-state patterns obtained for the parameter values of Figure 8, $\beta = 5$ and λ as indicated

4.2.2. *2-Dimensional Case.* In the case of an ellipsoidal domain Ω a similar numerical procedure gave the patterns shown in Figure 7.

In both series of experiments the parameter values were:

$$s_0 = 102.5, \quad a_0 = 79.5, \quad \alpha = 1.45, \quad \rho = 13, \quad k = 0.1, \quad \beta = 5,$$

satisfying the conditions (H1)–(H6) given in the next section.

We used the finite difference method in the 1-dimensional case, and the finite element method in the 2-dimensional case. The latter can be employed without any change to a geometrical domain of arbitrary shape, and the ellipsoidal shape

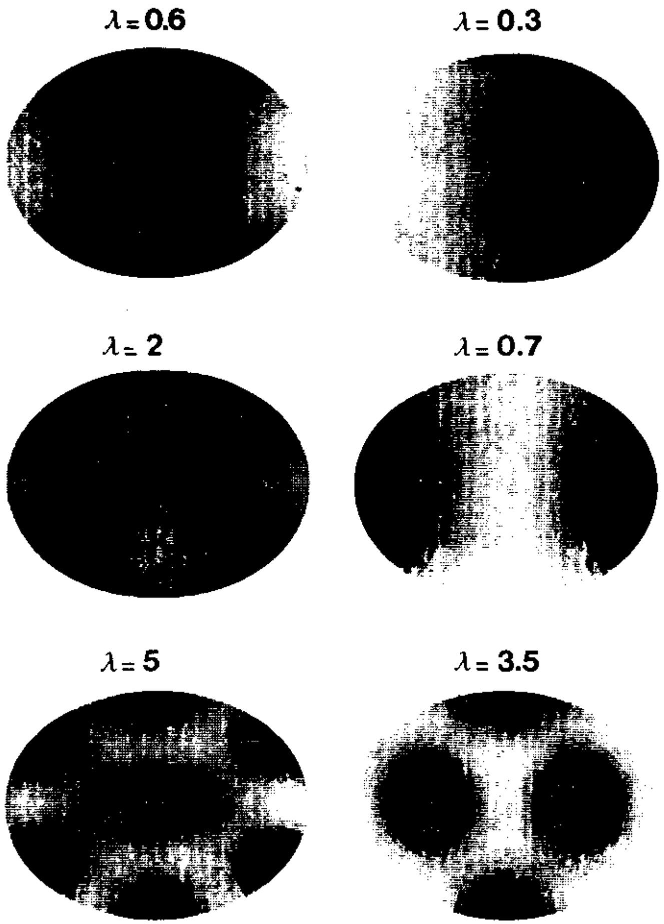

Fig. 7. Steady-state patterns obtained for the parameter values of Figure 8, $\beta = 5$, and λ as indicated

was chosen only because of the possible comparison with the predictions of Kauffman et al., and the corresponding experimental observations on sequential compartment formation in Drosophila.

In both series of experiments, every time λ was changed, the preceding steady-state was triggered off by random disturbances in order to have the whole range of spatial wavelengths in the Fourier expansion of the initial disturbance.

In fact these environmental conditions, so that the system may undergo its sequential alterations, seem to be a good simulation of the ever present thermal noise in Nature.

4.3. *Assumptions and Notations*

4.3.1. *Statement of the Assumptions and their Meaning*

(H1) There is (Fig. 8) an unique solution $(\bar{s}, \bar{a})$ for the algebraic system:

$$F(s, a) - (s_0 - s) = 0, \qquad F(s, a) - \alpha(a_0 - a) = 0 \tag{21}$$

(H2) $\alpha > 1, \quad \beta > 1$
(H3) $F_s + 1 + F_a + \alpha > 0.$

Here

$$F_s = \frac{\partial F}{\partial s}(\bar{s}, \bar{a}), \qquad F_a = \frac{\partial F}{\partial a}(\bar{s}, \bar{a}).$$

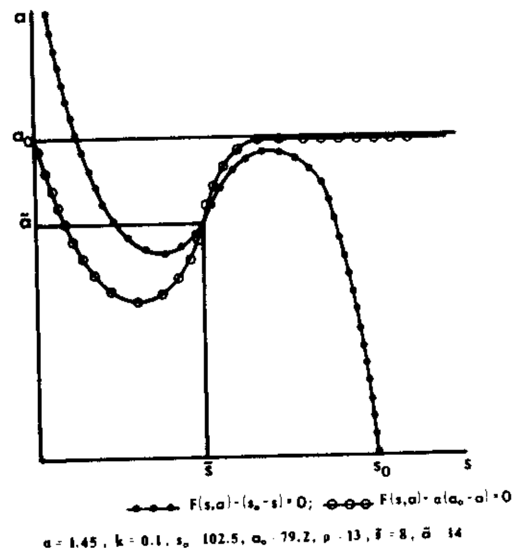

Fig. 8. Values as indicated

(H4) $\alpha F_s + F_a + \alpha > 0$
(H5) $\beta(F_s + 1) + F_a + \alpha < 0$
(H6) $(F_a + \alpha - \beta(F_s + 1))^2 > -4\beta F_s F_a$.

The hypotheses (H3)–(H6) are similar to those given by Kauffman et al.

From (H1) it results that there exists one and only one spatially uniform steady-state solution to (4), the trivial one $s \equiv \bar{s}$, $a \equiv \bar{a}$.

In Section 4.4 we shall study the stability of this trivial steady-state solution $(\bar{s}, \bar{a})$ according to the value of λ. In Section 4.5 we shall see that when $(\bar{s}, \bar{a})$ loses its stability by λ crossing some critical value, then its symmetry is broken and its stability is transferred to a steady-state space-dependent regime. (H3) and (H4) mean that $(\bar{s}, \bar{a})$ is a stable steady-state solution for the 0-dimensional dynamical system.

$$\frac{ds}{dt} + F(s, a) - (s_0 - s) = 0, \qquad \frac{da}{dt} + F(s, a) - \alpha(a_0 - a) = 0.$$

Finally (H6) means that the quadratic polynomial:

$$T(z) = \beta z^2 + [\beta(F_s + 1) + F_a + \alpha]z + \alpha(F_s + 1) + F_a$$

has 2 real roots z' and z'' and (H4) and (H5) that these roots are positive: $0 < z' < z''$.

4.3.2. *Choice of the Parameters.* Parameters k and β are given

It is not obvious that the other parameters α, β, s_0, a_0, ρ can be chosen so that (H1)–(H6) hold. Here is one way to obtain a set of such parameters. We use the fact that hypothesis (H4), (H5), (H6) are equivalent to the fact that the polynomial T can be written:

$$T(z) = \beta(z - z')(z - z'') \quad 0 < z' < z'',$$

which itself is equivalent to:

$$\beta(F_s + 1) + F_a + \alpha = -\beta(z' + z''), \qquad \alpha(F_s + 1) + F_a = \beta z'z''$$

or

$$F_s = -(z' + 1)(z'' + 1)(1 - \alpha/\beta), \qquad F_a = (z' + \alpha/\beta)(z'' + \alpha/\beta)(1 - \alpha/\beta). \tag{22}$$

As a by-product we see that we must have $\alpha < \beta$ in order that $F_s < 0$ (implied by (H5)). From Eqs. (4) we have:

$$F(s, a) = \rho a G(s), \quad \text{with } G(s) = s/(1 + s + ks^2).$$

Now take values for z', z'', α/β, ρ, $\bar{s}$ such that $0 < z' < z''$, $1 < \alpha < \beta$, $\rho > 0$, $G'(\bar{s}) < 0$.

From Eqs. (22) we know the value of $F_a = \rho G(\bar{s})$, whence ρ, and from the first one the value of $F_s = \rho\bar{a}G'(\bar{s})$, whence $\bar{a}$. By using Eqs. (21) with $s = \bar{s}$ and $a = \bar{a}$

we obtain s_0 and a_0. Then we check whether (H3) is satisfied and at last by considering the plots of:

$$a = (s_0 - s)/(\rho G(s)), \qquad a = \alpha a_0/(\alpha + \rho G(s))$$

we readily see whether (H1) holds.

4.3.3. *Other Notations.* The following notations will be used hereafter: $s = \bar{s} + u$, $a = \bar{a} + v$,

$$U = \begin{bmatrix} u \\ v \end{bmatrix}, \quad L_\lambda U = \begin{bmatrix} -\Delta u + \lambda(F_s + 1) + \lambda F_a v \\ -\beta\,\Delta v + \lambda F_s + \lambda(F_a + \alpha)v \end{bmatrix}$$

$$M_\lambda(U) = \lambda(F(\bar{s} + u, \bar{a} + v) - F(\bar{s}, \bar{a}) - F_s u - F_a v)\begin{bmatrix} 1 \\ 1 \end{bmatrix}$$

(μ_n, w_n), $n \geqslant 0$, will be the eigenpairs of $-\Delta$:

$$-\Delta w_n = \mu_n w_n \quad \text{on} \quad \Omega$$

subject to no-flux boundary conditions:

$$\frac{\partial w_n}{\partial \gamma} = 0 \quad \text{on} \quad \Gamma = \partial\Omega.$$

Here γ is the direction of the outward normal to the boundary Γ.

4.4. *Stability Analysis of* $(\bar{s}, \bar{a})$

It is well known (Iooss, 1973; Marsden and McCracken, 1976) that if the spectrum of L_λ lies in the right half plane, then $(\bar{s}, \bar{a})$ is asymptotically stable, whereas if there exists an eigenvalue k such that $Re k < 0$, then $(\bar{s}, \bar{a})$ is unstable. In fact the eigenpairs of L_λ are such that:

$$L_\lambda \Phi_n = k_n \Phi_n \qquad \Phi_n = w_n\begin{bmatrix} 1 \\ M_n \end{bmatrix},$$

k_n satisfying the dispersion equation:

$$\begin{vmatrix} \mu_n + \lambda(F_s + 1) - k_n & \lambda F_a \\ \lambda F_s & \beta\mu_n + \lambda(F_a + \alpha) - k_n \end{vmatrix} = 0 \tag{23}$$

and the corresponding M_n being such that:

$$\mu_n + \lambda(F_s + 1) - k_n + \lambda F_a M_n = 0.$$

(23) may be rewritten in the form of the quadratic equation:

$$k_n^2 - \text{tr}\,(n)k_n + \det\,(n) = 0. \tag{24}$$

Here tr (n) and det (n) are given by:

$$\begin{aligned} \text{tr}\,(n) &= (\beta + 1)\mu_n + \lambda(F_s + 1 + F_a + \alpha), \\ \det\,(n) &= \beta\mu_n^2 + \lambda(\beta(F_s + 1) + F_a + \alpha)\mu_n + \lambda^2(\alpha(F_s + 1) + F_a). \end{aligned} \tag{25}$$

We have the:

Proposition 4.1. *Under hypothesis H1–H6:*

i) *The eigenvalues* k_n *are real:* $k_n^- < k_n^+$, $n \geqslant 0$

ii) *For* $n = 0$, $0 < k_0^- < k_0^+$

iii) *For every* $n \geqslant 1$ *there is an interval* $I_n = (\mu_n/z'', \mu_n/z')$ *such that if* $\lambda \in I_n$, *then* $k_n^- < 0 < k_n^+$, *so that* $(\bar{s}, \bar{a})$ *is unstable. On the other hand if* $\lambda \notin I_n$, *then* $0 < k_n^- < k_n^+$ *and if* λ *is outside all the* I_n, *then* $(\bar{s}, \bar{a})$ *is stable.*

Proof.

i) the discriminant of the polynomial expression in (24) is:

$$\mathrm{tr}^2(n) - 4\det(n) = [(\beta - 1)\mu_n + \lambda(F_a^{1/2} - (-F_s)^{1/2})^2 + \lambda(\alpha - 1)] \times [(\beta - 1)\mu_n + \lambda(F_a^{1/2} + (-F_s)^{1/2})^2 + \lambda(\alpha - 1)]$$

(from (H5) $F_s < 0$).
From (H2) this discriminant is positive.

ii) for $n = 0$ we already know that (24) has 2 real solutions k_0^- and k_0^+. From (H3) and (H4) $k_0^- + k_0^+ > 0$, $k_0^- k_0^+ > 0$, whence $0 < k_0^- < k_0^+$.

iii) for $n \geqslant 1$ we always have:

$k_n^- + k_n^+ = \mathrm{tr}(n) > 0$, from expression (25) and (H3). Hence at least $k_n^+ > 0$. We also have $k_n^- > 0$, except if $\det(n) < 0$. But:

$$\det(n) = \lambda^2 T(\mu_n/\lambda) = \lambda^2\left(\frac{\mu_n}{\lambda} - z'\right)\left(\frac{\mu_n}{\lambda} - z''\right)$$

so that:

$$\det(n) < 0 \Leftrightarrow z' < \mu_n/\lambda < z'' \Leftrightarrow \lambda \in (\mu_n/z'', \mu_n/z') = I_n.$$

4.5. Existence and Stability of the Bifurcated Branches

Let λ_0 be a critical value such that when λ enters I_n by crossing λ_0, $(\bar{s}, \bar{a})$ loses its stability. Then this instability is symmetry breaking in the sense that there appear 2 branches of stable bifurcating steady-state solutions of (4). This is a consequence of operators L_λ and M_λ properties, as they have been established in detail by Meurant and Saut for the Prigogine model, and of bifurcation theory results (Iooss, 1973; pp. VIII.19 and IX.12).

The same methods may be applied to our system. Of course it is only possible to claim the existence and stability of bifurcating branches provided that the parameter λ is in an unknown neighborhood of λ_0.

4.6. Formal Calculation of the Bifurcated Branches

By a two time-scales method already used by Matkovsky (1970), Kogelman and Keller (1971), Boa (1974), Boa and Cohen (1976), we can show that if λ is entering I_{n0}, then for λ near λ_0 and $\lambda \geqslant \lambda_0$, $\lambda = \lambda_0 + \varepsilon^2$ and:

$$\begin{bmatrix} s \\ a \end{bmatrix} = \begin{bmatrix} \bar{s} \\ \bar{a} \end{bmatrix} + \varepsilon\left(\xi(\varepsilon^2 t)w_{n_0}\begin{bmatrix} 1 \\ M_{n_0}^- \end{bmatrix} + e.d.\right) + O(\varepsilon^2).$$

Here *e.d.* means exponentially decreasing terms and $\xi(\tau)$ takes the form of:

$$\xi(\tau) = \xi(0)|\xi(\infty)|e^{\mu\tau}/(\xi(0)^2(e^{\mu\tau} - 1) + \xi(\infty)^2)^{1/2}.$$

When $t \to \infty$

$$\begin{bmatrix} s \\ a \end{bmatrix} \to \begin{bmatrix} \bar{s} \\ \bar{a} \end{bmatrix} + (\lambda - \lambda_0)^{1/2}\xi(\infty)w_{n_0}(r)\begin{bmatrix} 1 \\ M_{n_0}^- \end{bmatrix} + 0(\lambda - \lambda_0).$$

5. Conclusion

The simplicity of the models studied in this paper is such that most probably similar situations currently arise in living cells. Uricase is just one example among many other common enzymes having the same kinetic properties. An enzyme layer separating two solutions, or an enzyme layer separated from a solution by a boundary layer, are frequent situations. Consequently it is to be hoped that more and more biological phenomena will be explained by considering spatially and temporally ordered solutions of reaction and diffusion equations.

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ANALYSIS AND OPTIMIZATION OF SYSTEMS WITH MULTIPLE STEADY STATES

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ABSTRACT

This communication presents an algorithm for optimization of systems with multiple steady states. This algorithm is a variation of the generalized gradient method, with the possibility to deal with bifurcating sheets of steady states. A motivation for studying such systems is the behavior of an enzyme system modeled by reaction-diffusion equations. This system can show pattern formation, and the problem is to maximize this pattern. The phenomenon of pattern formation in immobilized enzyme systems is explained, and its relation with morphogenesis is pointed out. Pattern optimization is a problem of the form: minimize $J(z,v)$, where the state and control variables z and v are related by a state equation $f(z,v) = 0$ with possibly multiple states z for each control v. It is shown how methods for continuation and bifurcation can be adapted to find sequential points (z_n,v_n) with decreasing values of $J(z_n,v_n)$.

1. INTRODUCTION

Diffusion-reaction systems have been extensively studied during the last decades, one of the motivations being that such systems could account for phenomena occuring in living organisms. In particular it has been suggested that morphogenesis in embryos could be of the same nature as pattern formation in simple model chemical systems where diffusion and reaction interact. The pioneer in this field was Turing in 1952 [1], who was followed, among others, by Prigogine and coworkers [2]. Their Bruxellator has been the most popular model for many numerical and mathematical developments.

Immobilized enzyme systems are other examples of dissipative structures, i.e. of structures in which temporal or spatial order may occur as a consequence of the dissipation of energy or of substrates coming from the external medium. We will focus on the system governed by the following equations:

$$(1.1)\quad \begin{cases} \frac{\partial s}{\partial t} - \Delta s + \lambda[\rho a F(s) - (s_o - s)] = 0 \quad \text{in } \Omega, \\ \frac{\partial a}{\partial t} - \beta\Delta a + \lambda[\rho a F(s) - \alpha(a_o - a)] = 0, \\ \text{with no-flux boundary conditions} \\ F(s) = s/(1 + s + ks^2) \end{cases}$$

Here s and a are the concentrations of two substrates reacting together and diffusing through a membrane Ω. The reaction is catalyzed by enzyme molecules uniformly distributed in Ω. Ω is a planar disk (Fig. 1.1). The substrates consumed in Ω are transported across a boundary layer from a well-stirred reservoir where those substrates are at fixed concentrations s_o and a_o. In the first equation of (1.1) the terms of diffusion through Ω, reaction within Ω and transport across the boundary layer are respectively $-\Delta s$, $\lambda\rho a F(s)$, and $(s_o - s)$. In the 2nd equation α(resp. β) is the ratio of diffusion coefficients in the boundary layer (resp. Ω). λ and ρ are the ratios of characteristic times:

$$(1.2)\qquad \lambda = \theta_D/\theta_T \text{ and } \rho = \theta_T/\theta_R$$

where θ_D, θ_T, and θ_R are the characteristic times for diffusion within Ω, transport across the boundary layer and reaction in Ω. This system and its properties have been studied in detail in [3]. Its main interest is that it admits, in addition to a trivial, spatially uniform, steady state solution $(\tilde{s},\tilde{a})$, defined by:

$$(1.3)\quad \begin{cases} \tilde{s} - s_o - \rho\tilde{a}F(\tilde{s}) = 0 \\ \alpha(\tilde{a} - a_o) - \rho\tilde{a}F(\tilde{s}) = 0, \end{cases}$$

other (stable) steady state solutions which are patterned (Fig. 1.2). By steady-state solution of (1.1) we mean a solution of:

$$(1.4)\quad \begin{cases} -\Delta s + \lambda[\rho a F(s) - (s_o - s)] = 0, \\ -\beta\Delta a + \lambda[\rho a F(s) - \alpha(a_o - a)] = 0 \\ \frac{\partial s}{\partial \nu} = 0, \quad \frac{\partial a}{\partial \nu} = 0, \end{cases}$$

and its stability is relative to the "dynamical" system (1.1). Figure 1.2 shows the patterns of s for such a solution (s,a). The higher the concentration, the greyer the level.

The background of this communication is the following optimal control problem: the state of the system is a solution of the reaction-diffusion equations (1.4). The control $v = (s_o, a_o, \lambda, \rho)$ is a vector of 4 positive and bounded parameters. The cost function is:

$$(1.5)\quad J(z,v) = -\int_\Omega (|\nabla s|^2 + |\nabla a|^2)dx + M(a_o^2 + s_o^2 + \lambda^2 + \rho^2),$$

where z = (s,a). The problem to find

$$(1.6)\quad \inf J(z,v), \quad (z,v) \text{ related by } (1.4),$$

corresponds to the goal of achieving patterns with gradients as large as possible, at the lowest price for v. Problem (1.4), (1.5), (1.6) is just an example of the following:

A dynamical system:

$$(1.7)\quad \frac{dz}{dt} + f(z,v) = 0$$

depends upon a vector of parameters v, and admits, when v is conveniently chosen, multiple steady states:

$$(1.8)\quad f(z,v) = 0.$$

The aim is to find a pair (z,v) satisfying (1.8) and minimizing a cost function J(z,v).

Thus after explaining pattern formation in enzyme systems in Section 2, relating it to morphogenesis in Section 3, we give, in Sections 4 and 5, general methods for optimization and, respectively, continuation and bifurcation.

2. PATTERN FORMATION IN IMMOBILIZED ENZYME SYSTEMS

Maybe one of the simplest dissipative structures is the one we are going to introduce now. At first, consider the two substrates S and A reacting together in a homogeneous solution of these substrates and of the enzyme E which catalyzes their reaction. The evolution of their concentrations is governed by the O.D.E.s

$$(2.1)\quad \begin{cases} \frac{ds}{dt} = \frac{da}{dt} = -\frac{1}{\theta_R} aF(s) \\ F(s) = s/(1 + s + ks^2). \end{cases}$$

This system is not very interesting - both concentrations tend to 0 as time evolves.

Let us remark at this stage that the velocity term, increasing with a and tending to 0 as $s \to \infty$, is representative of a large class of enzyme reactions, which are said to be activated by A and inhibited by exceeding S.

Now let us complicate a little our system, by introducing an inactive (i.e. without enzyme) membrane between the reactor containing the enzyme and an external reservoir containing the substrates at concentrations s_o and a_o. The governing equations for the concentrations of S and A in the reactor are now:

$$(2.2)\quad \begin{cases} \frac{ds}{dt} = -\frac{1}{\theta_R} aF(s) + \frac{1}{\theta_T}(s_o - s) \\ \frac{da}{dt} = -\frac{1}{\theta_R} aF(s) + \frac{\alpha}{\theta_T}(a_o - a). \end{cases}$$

At equilibrium we have:

$$(2.3)\quad \begin{cases} \rho a F(s) - (s_o - s) = 0 \\ \rho a F(s) - \alpha(a_o - a) = 0 \end{cases} \Leftrightarrow \begin{cases} a = (s_o - s)/\rho F(s) \\ a = \alpha a_o/(\alpha a_o + \rho F(s)), \end{cases}$$

where $\rho = \theta_T/\theta_R$. This defines a point $(\tilde{s},\tilde{a})$ which, for convenient values of s_o, a_o, α, and ρ, is unique and stable with respect to the dynamical system (2.2).

Again the behavior of (2.2) is not very interesting: s(t) and a(t) tend towards $\tilde{s}$ and $\tilde{a}$ as $t \to \infty$.

The situation becomes much more interesting if the reactor vessel is separated into 2 compartments by a membrane imposing diffusional constraints on the transport of S and A from each compartment to the other. The state variables are now s_1, a_1 and s_2, a_2, concentrations of S and A in both compartments. s_1 and a_1, for example, are governed by:

$$(2.4)\quad \begin{cases} \dfrac{ds_1}{dt} = -\dfrac{1}{\theta_R} a_1 F(s_1) + \dfrac{1}{\theta_T}(s_o - s_1) + \dfrac{1}{\theta_D}(s_2 - s_1) \\ \dfrac{da_1}{dt} = -\dfrac{1}{\theta_R} a_1 F(s_1) + \dfrac{\alpha}{\theta_T}(a_o - a_1) + \dfrac{\beta}{\theta_D}(a_2 - a_1) \end{cases}$$

and we have analogous equations for s_2 and a_2.

It is easy to check that $s_1 = s_2 = \tilde{s}$, $a_1 = a_2 = \tilde{a}$ is still an equilibrium solution that one could still think of as stable. However this is not true. The steady states are the solutions of:

$$(2.5)\quad \begin{cases} s_1 - s_2 + \lambda[\rho a_1 F(s_1) - (s_o - s)] = 0, \\ \beta(a_1 - a_2) + \lambda[\rho a_1 F(s_1) - \alpha(a_o - a_1)] = 0, \\ -s_1 + s_2 + \lambda[\rho a_2 F(s_2) - (s_o - s_2)] = 0, \\ \beta(-a_1 + a_2) + \lambda[\rho a_2 F(s_2) - \alpha(a_o - a_2)] = 0 \end{cases}$$

where $\lambda = \theta_D/\theta_T$ and $\rho = \theta_T/\theta_R$.

The representation of s_1 for example as a function of λ looks like Figure 2.1, where full (resp. dotted) lines represent stable (resp. unstable) steady states. Thus when λ is small or large the trivial steady state $(\tilde{s},\tilde{a})$ is stable, which can be easily understood since in the first case we are approaching the condition where there is no diffusional constraint ($\theta_D = 0$) and in the second we are approaching the condition where both compartments are separated by an impermeable wall ($\theta_D = \infty$). But there is an interval (λ', λ'') such that, for each value of λ between λ' and λ'', the trivial steady state is unstable and the two steady states which are stable are non-symmetric (i.e. $s_1 \neq s_2$ and $a_1 \neq a_2$). Usually diffusion has a smoothing effect and tends to make the concentrations uniform. Here it is the contrary: diffusional constraints may cause a gradient of concentrations.

This two-compartment model is simple enough to be realized experimentally. The problem is to find optimal values of the parameters, i.e. values minimizing:

$$(2.6)\quad J(z,v) = -(s_1 - s_2)^2 - (a_1 - a_2)^2 + M(a_o^2 + s_o^2 + \lambda^2 + \rho^2),$$

where $z = (s_1, s_2, a_1, a_2)$ and $v = (a_o, s_o, \lambda, \rho)$ are verifying (2.5). This is a discrete version of problem (1.4), (1.5), (1.6), which is posed by biochemists for their experiments.

We must point out the analogy with polarity in biology. In the preceding model, diffusion constraints, interacting with a very common enzyme activity, may cause a differentiation between 2 compartments.

In biology too, polarity phenomena (i.e. for example anterior-posterior differentiation) maybe due to a similar cause.

3. MORPHOGENESIS

Insect imaginal discs are assemblages of cells which metamorphose into the different adult appendages (wing, leg, genital, haltere, eye-antenna, ...). During the development of a disc, say the Drosophila wing disc for example, there is a sequential formation of compartments: anterior-posterior, dorsal-ventral, wing-thorax, ... Kauffman [4] observed that the compartment lines resemble the nodal lines of the mode shapes:

$$(3.1)\quad \begin{cases} -\Delta w = \mu w \quad \text{in } \Omega, \\ \dfrac{\partial w}{\partial \nu} = 0 \quad \text{on } \Gamma = \partial\Omega \end{cases}$$

the nodal lines being those points (x,y) such that $w(x,y) = 0$. Whence the idea that diffusion of "morphogens"" was playing a rôle, together with some reaction. Kauffman's theory is that a reaction-diffusion system:

$$(3.2)\quad \begin{cases} \dfrac{\partial s}{\partial t} - \Delta s + \lambda f(s,a) = 0 \text{ in } \Omega, \\ \dfrac{\partial a}{\partial t} - \beta\Delta a + \lambda g(s,a) = 0, \\ + \text{ no-flux B.C.s} \end{cases}$$

acts throughout development and generates a sequence of differently shaped chemical patterns. More precisely (3.2) passes, as λ varies, through a sequence of stable spatially non uniform steady states, inducing one commitment (for example anterior) in cells where s (or a) is above some threshold level, and the alternate commitment (posterior) in cells below threshold.

Increasing values of λ correspond to increasing sizes of the disc Ω. Since our S - A system (1.1) is of the same form as (3.2), it was interesting to check whether it was possible to find sequential patterns as λ varies. It is indeed what was found, as shows Figure 1.2. A complete diagram of the steady states of (1.1), either stable or unstable, is shown in Figure 3.1, which was obtained by Sharan [5] by employing the method of Kubicek [6] for continuation and of Keller [7] for bifurcation. This bifurcation diagram was obtained for the following values of the parameters:

(3.3) $$a_o = 79.2,\ k = 0.1,\ s_o = 102.5,\ \alpha = 1.45,\ \beta = 5, \text{ and } \rho = 13.$$

On this diagram the abscissa is λ and the ordinate is the value of s at some point of Ω. Full (resp. dotted) lines correspond to stable (resp. unstable) solutions of (1.4). The straight line A - I corresponds to the trivial steady-state $(\tilde{s},\tilde{a})$, and the points A - I are bifurcation points from this branch. Points J - Q are secondary bifurcation points. The path followed by system (1.4) as λ increases (we assume the variation of λ slow enough for sequential steady states to settle, i.e. we make the quasi-steady-state hypothesis) can be inferred from this bifurcation diagram. Before attaining point A, the system follows the trivial branch. Then it follows an arc AB of the first loop. Between B and C, $(\tilde{s},\tilde{a})$ again is stable, but after C it becomes unstable, and the system passes smoothly to steady states which are structured in space, between C and J, then jumps from J to one of the 2 branches of stable steady states existing for $\lambda > \lambda_J$, say the upper one, until N jumps again to, say, the upper line and follows it until R, ... Of course, if λ was permitted to decrease, the system would describe arc RP, jump from P to arc NL, jump from L to arc JC, ..., so that an hysteresis phenomenon would be observed: for a given value of λ the system can occupy different steady states according to its past history.

Now we see the importance of being able to control systems like (1.4), if diffusion and reaction play such a role in morphogenesis.

4. OPTIMIZATION AND CONTINUATION

It can be shown that the optimal control problem (1.4), (1.5), (1.6) admits a solution. We refer for the proof to [8], where other optimal control problems arising in biology are studied. We limit ourselves here to the numerical approximation of such problems. After discretization, we are faced with a problem of the

form:

(4.1) $$\begin{cases} \text{minimize } J(z,v) \\ \text{subject to } f(z,v) = 0 \\ v \in R^M,\ z \in R^N,\ f(z,v) \in R^N. \end{cases}$$

The numerical method is a variation of the generalized reduced gradient method employing techniques used in continuation and bifurcation methods.

Continuation of a branch of solutions of $f(z,v) = 0$ towards a point minimizing $J(z,v)$ (Fig. 4.1):

We employ the Kubicek method. We parametrize by arc-length s :

(4.2) $$\begin{cases} f(z(s),v(s)) = 0 \\ |\dot{z}(s)|^2 + |\dot{v}(s)|^2 = 1, \end{cases}$$

or:

(4.3) $$\begin{cases} f_z\dot{z} + f_v\dot{v} = 0 \\ |\dot{z}|^2 + |\dot{v}|^2 = 1 \\ z(0) = z_o,\ v(0) = v_o \end{cases}$$

where $\dot{z} = \frac{dz}{ds}$ and $\dot{v} = \frac{dv}{ds}$, while $[f_z \vdots f_v] =$

$$\left[\begin{array}{ccc:ccc} \frac{\partial f_1}{\partial z_1} & \cdots\cdots & \frac{\partial f_1}{\partial z_N} & \frac{\partial f_1}{\partial v_1} & \cdots\cdots & \frac{\partial f_1}{\partial v_M} \\ \vdots & & \vdots & \vdots & & \vdots \\ \frac{\partial f_N}{\partial z_1} & \cdots\cdots & \frac{\partial f_N}{\partial z_N} & \frac{\partial f_N}{\partial v_1} & & \frac{\partial f_N}{\partial v_M} \end{array}\right].$$

i) Total pivoting strategy is applied to the N x (N + M) array $[f_z \vdots f_v]$. This determines the "independent" variable w: it is that M-dimensional vector whose components correspond to the columns which have not been selected. Let y be the N-dimensional new "dependent" variable:

(4.4) $(z,v) = (y,w)$

As a by-product we have β such that:

(4.5) $f_y\beta + f_w = 0$

ii. Descent direction: Let:

(4.6) $\tilde{J}(w) = J(y(w),w).$

Then the reduced gradient is:

(4.7) $g = \tilde{J}'(w) = J_y^T\beta + J_w.$

We decide that:

(4.8) $\dot{w} = -\alpha_o g, \ \alpha_o > 0.$

Since

(4.9) $f_y\dot{y} + f_w\dot{w} = 0$

with f_y nonsingular, it results:

(4.10) $\dot{y} = -\alpha_o \beta g$

with

(4.11) $\alpha_o = 1/\sqrt{(|\beta g|^2 + |g|^2)}.$

iii. Prediction: (Fig. 4.2)

(4.12)
$$\hat{y} = y(s) + h\dot{y}(s)$$
$$\hat{w} = w(s) + h\dot{w}(s)$$

iv. Corrector step: (Fig. 4.2). Let $x = (z,v) = (y,w)$.

$$x^{(0)} = (\hat{y},\hat{w})$$
$$f_{y^{(k)}}(x^{(k)})(y^{k+1} - y^{(k)}) = -f(x^{(k)})$$

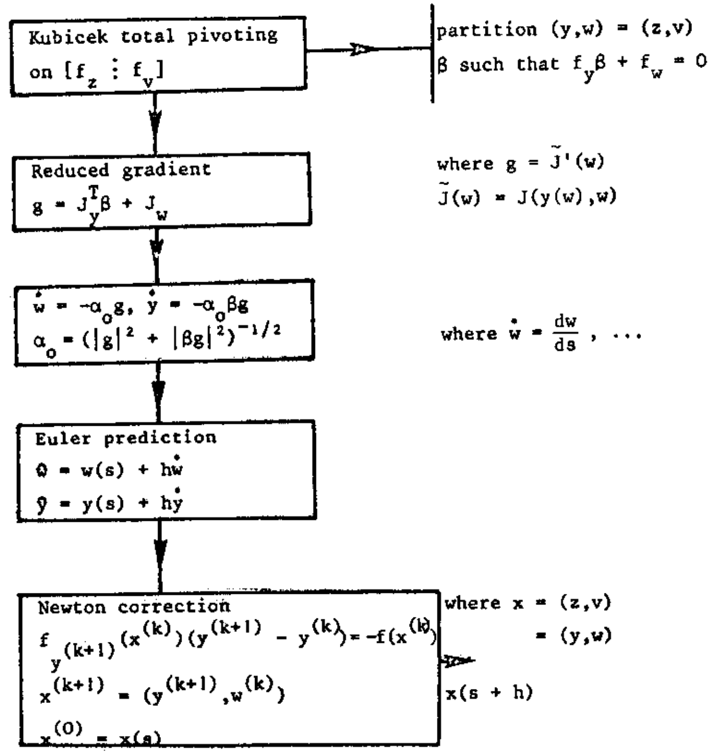
(#(4.13) continued)

(4.13)
$$x^{(k+1)} = (y^{(k+1)},w^{(k)})$$
$$\text{stop test: } |y^{(k+1)} - y^{(k)}| \leq \epsilon |y^{(k+1)}|$$
$$x(s+h) = \text{last found } x^{(k+1)}.$$

At k^{th} iteration, Gaussian elimination with total pivoting on $f_x(x^{(k)})$ selects the basic N-vector $y^{(k)}$ to be modified whereas the non-basic vector $w^{(k)}$ stays unchanged.

The important feature is that the selected columns may vary from iterations k to k + 1.

Design of Continuation for Minimizing J(z,v) Subject to f(z,v) = 0



5. BIFURCATION OF A BRANCH OF SOLUTIONS OF f(z,v) = 0 TOWARDS ANOTHER POINT MINIMIZING J(z,v) (Fig. 5.1)

Assumptions:

Let B be the bifurcation point (z_o, v_o).

(i) $f(z_o, v_o) = 0$,

(ii) $\dim N(f_z^0) = \operatorname{codim} R(f_z^0) = 1$,

$N(f_z^0) = \operatorname{span}\{\Phi\}$, $N(f_z^{0*}) = \operatorname{span}\{\Psi^*\}$

(iii) $R(f_v^0) \subset R(f_z^0)$

$(\Leftrightarrow \forall i,\ f_{v_i}^0 \in R(f_z^0) \Leftrightarrow \Psi^* f_{v_i}^0 = 0)$,

(iv) the roots of the bifurcation equation are distinct:

(5.1) $$a\alpha_1^2 + 2b\alpha_o\alpha_1 + c\alpha_o^2 = 0$$

where:

(5.2) $$a = \Psi^* f_{zz}^0 \Phi\Phi \text{ and } b = \Psi\,(f_{zz}^0 \Phi_o \Phi + f_{zv}^0 \Phi d)$$

(5.3) $$\begin{cases} f_z^0 \Phi_o + f_v^0 d = 0, & d = \dot{v}/|\dot{v}|, \\ \Psi^* \Phi_o = 0. \end{cases}$$

With these assumptions the tangent to the first curve (Fig. 5.1) is:

(5.4) $$\begin{cases} \dot{v} = \alpha_o d \\ \dot{z} = \alpha_o \Phi_o + \alpha_1 \Phi_1 \end{cases}$$

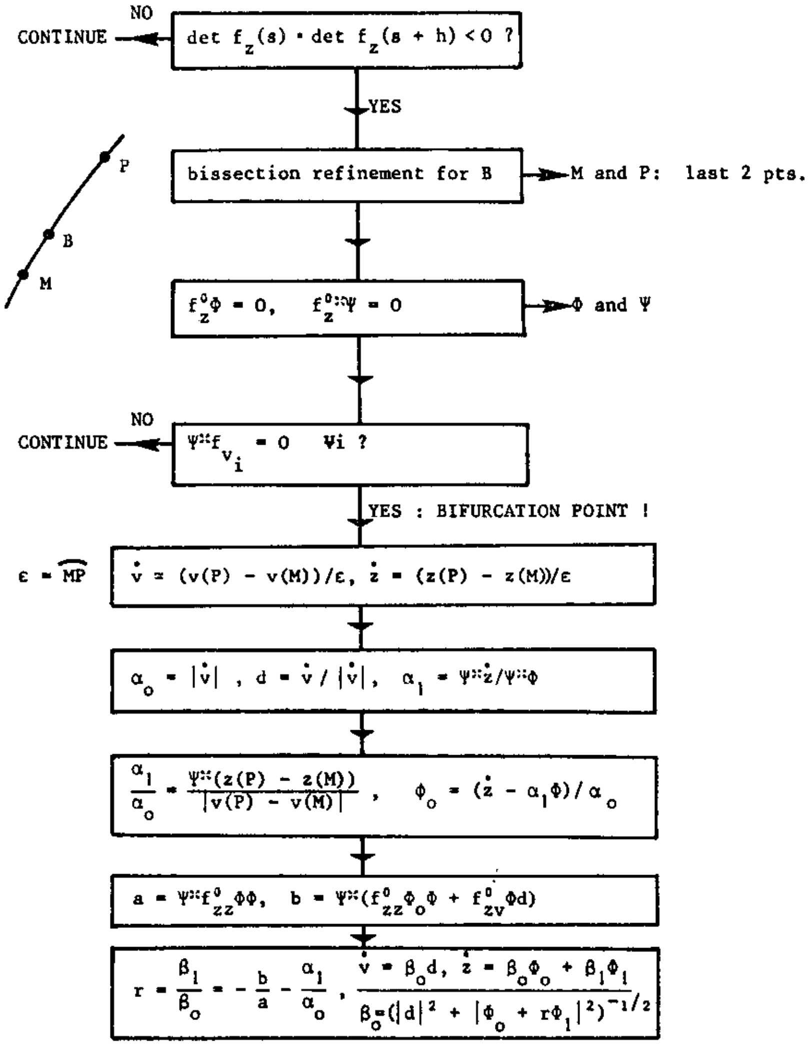
and the tangent to the second curve

(5.5) $$\begin{cases} \dot{v} = \beta_o d \\ \dot{z} = \beta_o \Phi_o + \beta_1 \Phi_1 \end{cases}$$

where both (α_o, α_1) and (β_o, β_1) satisfy the bifurcation equation (5.1), whence

(5.6) $$\frac{\alpha_1}{\alpha_o} + \frac{\beta_1}{\beta_o} = -\frac{b}{a}.$$

Design for Bifurcation



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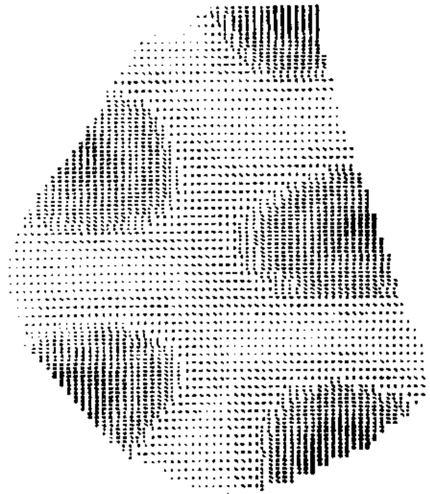

fig 1.2

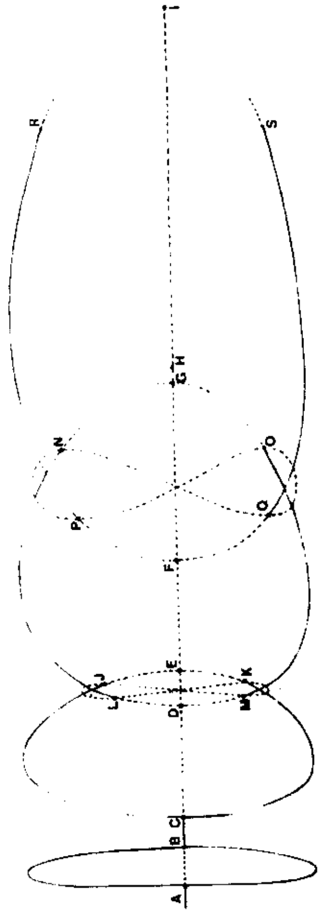

fig 3.1

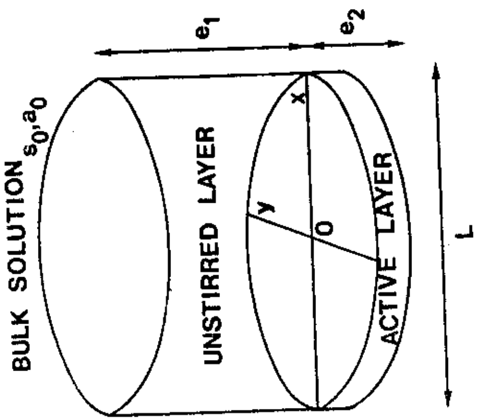

fig 1.1

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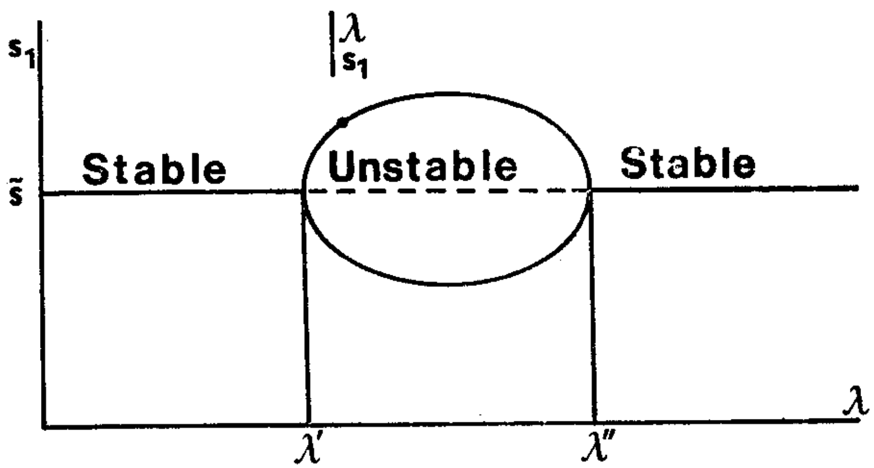

fig 2·1

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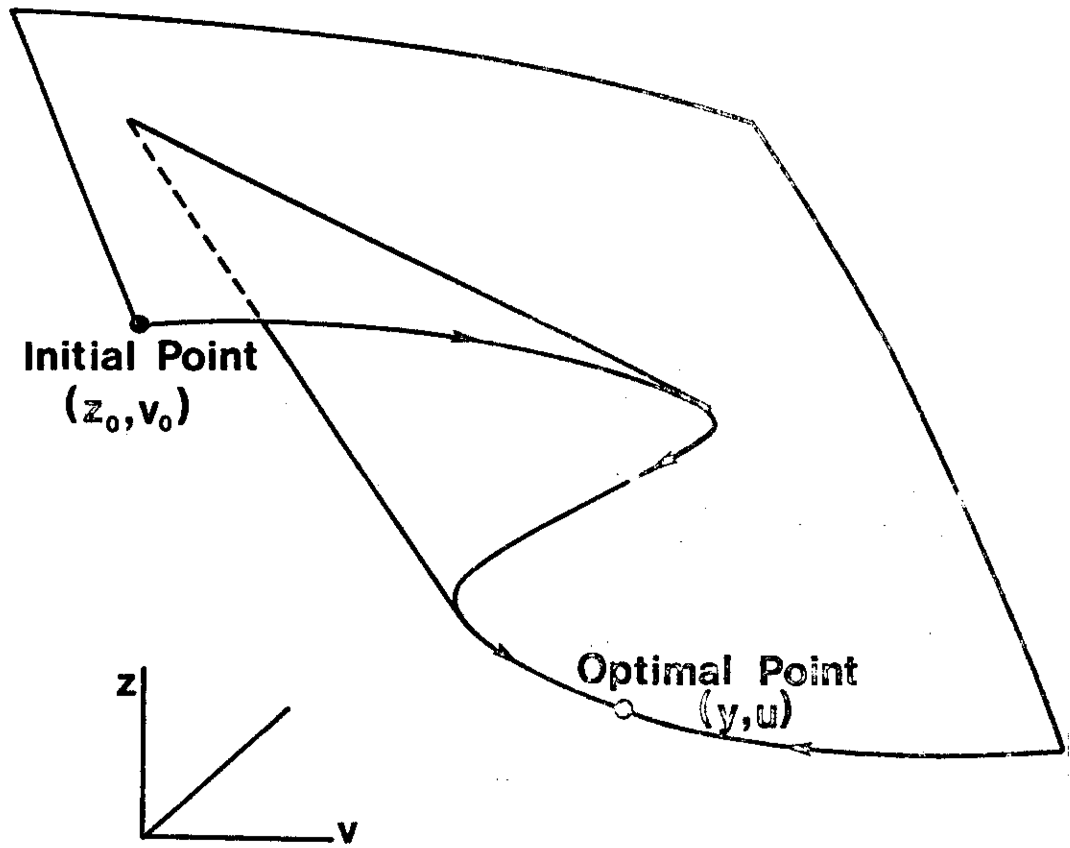

fig 4·1

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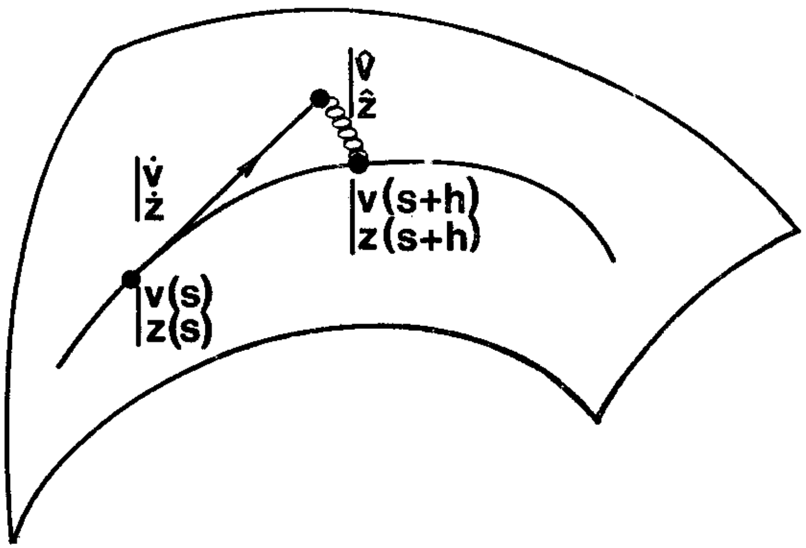

fig 4.2

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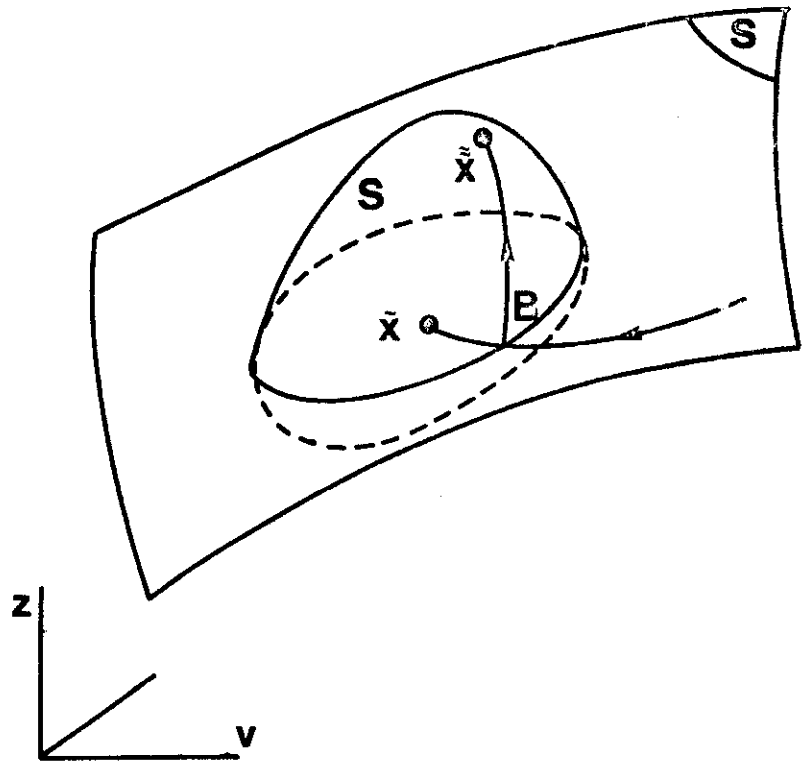

fig 5.1