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DYNAMIC BEHAVIOR OF A MEMBRANE PERMEABILITY SENSITIVE MODEL FOR A CONTINUOUS BIO-REACTOR EXHIBITING CULTURE RHYTHMICITY

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ABSTRACT

A modified Monod model of a continuous microbial culture in which the yield term depends linearly on the substrate concentration is extended to incorporate the effect of external forces on the cell membrane permeability. Bifurcation analysis of the new mathematical model, which consists of three non-linear ordinary differential equations, shows that the model can simulate the oscillatory behavior observed in experimental data for certain ranges of the system parameters. Computer simulation of the model is presented in support of our theoretical predictions.

INTRODUCTION

Sustained oscillations in the patterns of microbial growth and product formation have been frequently observed in continuous cultures when the feed conditions and the culture conditions remain constant [1, 2]. According to Yerushalmi *et al.* [2], these oscillations are even more pronounced in the long term fermentations or in the cell-retention fermentations where the cells stay in the bio-reactor for long periods of time.

Although the mechanism for these oscillations is not yet fully understood, it is clear that occurrence of such oscillatory behavior has adverse effects on the efforts to optimize the operation of continuous bio-reactors. It also effects productivity of the process and complicates its proper design. It is therefore most important to investigate in depth the factors that cause such rhythmicities, the explanations for which range from experimental errors to the changing microbial physiological behavior often attributed to changes in the cellular metabolic pathway under certain conditions. Recent studies of the parameter affecting the cell physiology of *C. acetobutylicum* showed a high sensitivity of growth and solvent production to the cytoplasmic membrane permeability [2]. A high permeability of the cytoplasmic membrane promotes the growth of the microbial culture, the utilization of the substrate and the biosynthesis of the solvents. The opposite result is obtained with a low permeability of the cell membrane.

The controlling action of the cellular membrane permeability on the activities in many anaerobic processes has been frequently observed. Examples include the influence of plasma-membrane lipid composition and membrane fluidity on growth and solute accumulation by *S. cerevisiae* [3], growth of *Clostridium thermocellum* [4], and growth and production of ethanol and glycerol by yeast cultures [5].

In this paper, we consider a mathematical model which incorporates this sensitivity to the cellular membrane permeability, the specific rate of change of which is assumed to vary in a sinusoidal fashion. One physical controlling factor which has been proposed to exert its biological effect on the cytoplasmic membrane permeability is the geomagnetic field variation. This concept has been extensively investigated and is well supported by experimental evidence [6, 7]. Attempts to incorporate such effects into a model of the continuous microbial culture was carried out by Yerushalmi *et al.* [2]. We consider a modification of their model based on an adaptation of the Monod model in which the yield term is assumed to vary linearly with the substrate concentration. Through bifurcation analysis, the model is shown to simulate different oscillatory behavior observed in experimental data.

SYSTEM MODEL

Basically, microbial kinetics have varied in diverse ways from a model due to Monod fashioned after Michaelis-Menten kinetics for single enzyme-substrate reactions. This simple but valuable model views microbial growth as conversion of a fixed amount of substrate (or nutrient) to biomass occurring autocatalytically in the presence of preexisting biomass [8]. The yield coefficient Y in the Monod's model is constant. The most obvious departure of the predictions of Monod's model, apparently, is in the variation of the stoichiometric coefficient Y. Theoretical studies of models in which the yield term varies linearly with the substrate concentration can be found in the work of Agrawal *et al.* [8] and that of Lenbury *et al.* [9]. In [8], Agrawal *et al.* carried out an extensive theoretical investigation of the dynamic behavior of isothermal continuous stirred tank biological reactors modelled by the following mass balance equations on cells and the limiting substrate:

$$\frac{dS}{dt} = -\sigma(S)X + D(S_0 - S) \qquad (1)$$

$$\frac{dX}{dt} = \mu(S)X - DX \qquad (2)$$

where X denotes the cells concentration; S the substrate concentration; $\mu(S)$ the specific growth rate; $\sigma(S)$ the specific substrate consumption rate; S_0 the feed substrate concentration; and D the dilution rate.

In their work, the function $\sigma(S)$ was assumed to have the form

$$\sigma(S) \equiv \frac{\mu(S)}{Y(S)} \equiv \frac{\mu_m S}{(K_m + S)Y(S)} \qquad (3)$$

where μ_m is the maximum specific growth rate and K_m is the Monod constant while the yield term Y(S) has the form

$$Y(S) = \frac{\text{amount of biomass formed}}{\text{amount of substrate consumed}} \equiv aS + b \qquad (4)$$

which reflects the increase in the yield in response to an increase in the substrate concentration S. This also includes the case of constant yield when $a = 0$.

The model equations (1) and (2) do not take into account the variation of the membrane permeability with time. Since studies have confirmed high sensitivity of culture growth and production to membrane permeability, it is suggested in [2] that the influence is incorporated into the system model so that the mass balance equation on the limiting substrate is given by

$$\frac{dS}{dt} = -\frac{n'SX}{S+K_m} + D(S_0 - S) \qquad (5)$$

where $n' = kn$, with k a proportionality constant, and n the number of active nutrient transport sites. According to Yerushalmi *et al.* [2], permeation dynamics is the major factor responsible for the formation of the active sugar (nutrient) transport sites, especially in the aging cells. This is in turns due to the accumulation of the non-active deposits in the cytoplasm which make the permeation control the incorporation of the protein in the lipid skeleton of the cytoplasmic membrane. This relationship may be described by the equation:

$$\frac{d}{dt}(nX) = k_p \frac{d}{dt}(PX) \qquad (6)$$

where P measures the membrane permeability and k_p is a constant of variation. Integrating equation (6), we obtain the relation

$$nX = k_p XP + k_1 \qquad (7)$$

where k_1 is a constant of integration.

Using (7), equation (5) may be cast in the following form:

$$\frac{dS}{dt} = -\frac{(C_1XP + C_2)S}{(S+K_m)Y} + D(S_0 - S) \qquad (8)$$

where $C_1 = kk_pY$ and $C_2 = kk_1Y$ are constants. In other words, assuming that the yield term is constant, the specific growth rate has the form

$$\mu = \frac{(C_1P + C_2 / X)S}{(S + K_m)} \qquad (9)$$

so that the mass balance equation for X becomes

$$\frac{dX}{dt} = \frac{(C_1XP + C_2)S}{(S + K_m)} - DX \qquad (10)$$

in which the effect of permeability variation has been taken into account. On the other hand, it is reasonable to expect the yield coefficient Y to reflect the varying amount of nutrient mass required to produce a unit of biomass, as has been argued in [8] and [9] for example. We therefore combine both effects by letting Y assume the form in (4) so that the mass balance equation for S becomes

$$\frac{dS}{dt} = -\frac{(C_1XP + C_2)S}{(S + K_m)(aS + b)} + D(S_0 - S) \qquad (11)$$

Experimental evidence has shown that external forces such as electrical or magnetic fields can contribute to permeability by introducing an 'order' in the composition of the cytoplasmic membrane (see [2] for more detail). As a result, the cellular membrane permeability can follow an oscillatory pattern which can be described by the following equation:

$$\frac{dP}{dt} = -K\cos(\omega_0 t)P \qquad (12)$$

where K is a proportionality constant. Equation (12) describes the periodic changes in the cytoplasmic membrane permeability when there is no cells growth. If there is cells growth, the newly formed cells posses thin cell membrane with high permeability which contributes to an increase in the apparent permeability of the cells population. In the case of influence from the geomagnetic field variations, the period is found to be approximately 24 hours, so that $\omega_0 = 2\ /24$. However, to include other factors which may effect membrane permeability in the similar manner, we let ω_0 be an arbitrary constant frequency of oscillation of the applied field.

Thus, the variation in the permeability of the cells population, based on the overall cells mass, can be described by the following equation:

$$\frac{d}{dt}(PX) = -\gamma_1\cos(\omega_0 t)PX + \gamma_2\frac{dX}{dt}$$

in which the first term on the right was directly obtained from equation (12), describing the periodic changes in the membrane permeability, while the second term describes the increase in the apparent permeability of the cells population due to the growth of the culture and the formation of new cells, assuming that the inhibitory effect of other factors such as the butanol level is neglegible.

Eliminating X from both sides of the above equation results in the following expression:

$$\frac{dP}{dt} = -\gamma_1\cos(\omega_0 t)P + (\gamma_2 - P)\mu \qquad (13)$$

where μ is given by equation (9).

Therefore, our system model consists of equations (10), (11), and (14) with (9). We are interested in the dynamic behavior and , in particular, the existence of different types of oscillatory behavior in the system described by these three equations.

BIFURCATION ANALYSIS

For the following analysis, it is convenient to introduce new variables. Namely, we define $T = Dt$, $x = X/a$, $y = PC_1/D$, $z = S$, $\rho = C_2/aD$, $M = k_m$, $d = b/a$, $z_0 = S_0$, $\alpha = 1/D$, $\beta = \gamma_2 C_1/D$, $u = \cos(\omega_0 t)$, $v = \gamma_1\sin(\omega_0 t)$, and $\omega = \omega_0/D$.

In these variables, our model equations becomes

$$\frac{dx}{dT} = (xy+\rho)\frac{z}{M+z} - x \qquad (14)$$

$$\frac{dy}{dT} = -\alpha uy + (\beta - y)\left[y + \frac{\rho}{x}\right]\frac{z}{M+z} \qquad (15)$$

$$\frac{dz}{dT} = -(xy+\rho)\frac{z}{(M+z)(z+d)} + (z_0 - z) \qquad (16)$$

$$\frac{du}{dT} = -\omega v \qquad (17)$$

$$\frac{dv}{dT} = \omega u \qquad (18)$$

The above system has a steady state solution $(x_s, y_s, z_s, u_s, v_s)$ obtained from equating the right sides of equations (14) - (18) to zero, namely

$$y_s = \beta \tag{19}$$

$$-(\beta x_s + \rho)\frac{z_s}{(M + z_s)(z_s + d)} + (z_0 - z_s) = 0 \tag{20}$$

$$x_s = (z_s + d)\,(z_0 - z_s) \tag{21}$$

and

$$u_s = 0, \quad v_s = 0 \tag{22}$$

If we let

$$\theta = \frac{z_s}{M + z_s} \tag{23}$$

$$\delta = \frac{(\beta x_s + \rho)M}{(M + z_s)^2} \tag{24}$$

then the Jacobian matrix J of the system of equations (14) - (18) evaluated at the steady state $(x_s, y_s, z_s, u_s, v_s)$ can be written as

$$J = \begin{bmatrix} \beta\theta - 1 & \theta x_s & \delta & 0 & 0 \\ 0 & -1 & 0 & -\alpha y_s & 0 \\ \dfrac{-\theta\beta}{z_s + d} & \dfrac{-x_s\theta}{z_s + d} & \dfrac{\delta(z_s^2 - Md)}{M(z_s + d)^2} - 1 & 0 & 0 \\ 0 & 0 & 0 & 0 & -\omega \\ 0 & 0 & 0 & \omega & 0 \end{bmatrix}$$

The 5 eigenvalues of J are found to be

$$\lambda_{1,2} = \frac{1}{2}\Gamma(\delta) \pm \frac{1}{2}\Lambda^{1/2}(\delta) \qquad (25)$$

$$\lambda_3 = -1$$

$$\lambda_{4,5} = \pm\, i\omega$$

where

$$\Gamma(\delta) = \beta\theta + \frac{\delta(z_s^2 - Md)}{M(z_s + d)^2} - 2 \qquad (26)$$

$$\Lambda(\delta) = \Gamma^2(\delta) - 4\{(\beta\theta - 1)\left[\frac{\delta(z_s^{\,2} - Md)}{M(z_s + d)^2} - 1\right] + \frac{\theta\beta\delta}{z_s + d}\} \qquad (27)$$

Due to the complex conjugate eigenvalues ± $i\omega$, therefore, the model will have a periodic solution for appropriate parametric values. In particular, by the theory of ordinary differential equations, if the parametric values are such that all eigenvalues other tha $\alpha_{4,5}$ have negative real parts, then the simulated solution trajectories close to the steady state will approach a closed cycle surrounding the critical point (x_s, y_s, z_s, u_s, v_s) in the five dimensional phase space. In this case the profile of $x(T)$ will be periodic with time closely resembling the regular rhythmicity found in many experimental data. However, such closed cycles lying on a plane in the phase space cannot simulate more irregular oscillatory patterns also observed in other data, such as that taken from the work of Paruleka *et al.*[10] presented in Figure 1. Here, alternatively low and high peaks can be observed in the growth pattern. Such characteristics appear in all their runs under different operating parameters.

To investigate the possibility of such higher dimensional oscillations in our model, we consider the system of equations (14) - (16) with $\alpha = 0$, and let

$$\delta_b \equiv (\beta\theta - 1)^2 (z + d) / \beta\theta \qquad (28)$$

$$\delta_c \equiv (2 - \beta\theta)(z_s + d)\gamma \qquad (29)$$

where

$$\gamma \equiv \frac{(z_s + d)M}{z_s^{\,2} - Md} \qquad (30)$$

According to Hopf bifurcation theory [11], if a value δ_c can be found such that

i) Re $\lambda_1(\delta_c) = 0$,
ii) $\lambda_1(\delta_c)$ and $\lambda_2(\delta_c)$ are complex conjugates,
iii) Im $\lambda_1(\delta_c) \neq 0$,
iv) Re $\lambda'_1(\delta_c) \neq 0$, where λ' denotes the derivative of λ,
v) all other eigenvalues have negative real parts,

then the system of equations (14) - (16) with $\alpha = 0$ will have a family of periodic solutions for values of δ in some open interval ($\delta_c, \delta_c+\varepsilon$). The result is stated in the following theorem.

Theorem If

$$\gamma > 0 \qquad (31)$$

$$\beta \geq 1 \qquad (32)$$

$$1/\beta > \theta > \frac{1-\sqrt{\gamma/(\gamma+1)}}{\beta} \qquad (33)$$

and

$$\gamma > M > \frac{1-\theta}{\theta} \qquad (34)$$

then the system of equations (14) - (16) with $\alpha = 0$ will have periodic solutions bifurcating from a non-washout steady state for values of δ in some open interval ($\delta_c, \delta_c+\varepsilon$) where is given by equation (29).

Proof First, we show that with θ so chosen, $\delta_b < \delta_c$ by considering the equation

$$F(\theta) \equiv (\beta\theta)^2 - 2(\beta\theta) + \frac{1}{\gamma+1} = 0$$

The function $F(\theta)$ is quadratic in θ and has two real roots:

$$\theta_{1,2} = \frac{1 \pm \sqrt{\gamma/(\gamma+1)}}{\beta} \qquad (35)$$

Thus, for $\theta_1 > \theta > \theta_2$, we have $F(\theta) < 0$, that is

$$(\beta\theta)^2 - 2(\beta\theta) + \frac{1}{\gamma+1} < 0 \qquad (36)$$

Rearranging (36), we find

$$(\beta\theta)^2 - 2(\beta\theta) + 1 < (2\beta\theta - \beta^2\theta^2)\gamma \qquad (37)$$

Multipying both sides by z_s+d, we have

$$\frac{(\beta\theta - 1)^2 (z_s + d)}{\beta\theta} < (2 - \beta\theta)(z_s + d)\gamma \qquad (38)$$

That is, we have

$$\delta_b < \delta_c \qquad (39)$$

if $\theta_1 > \theta > \theta_2$. However,

$$\theta_1 = \frac{1 + \sqrt{\gamma / \gamma + 1}}{\beta} > 1/\beta$$

so that if θ satisfies inequality (33) then

$$\theta_1 > 1/\beta > \theta > \theta_2$$

which implies (39) as claimed.

Now, we observe that

$$\Gamma(\delta_c) = 0 \qquad (40)$$

and $$\Lambda(\delta_c) = -4\left[-(\beta\theta - 1)^2 + \frac{\theta\beta\delta_c}{z_s + d}\right] \qquad (41)$$

which is negative because of inequality (39). Thus,

$$\operatorname{Re} \lambda_1 (\delta_c) = \Gamma (\delta_c)/2 = 0$$

and $\lambda_1 (\delta_c)$ and $\lambda_2 (\delta_c)$ are complex conjugates. Also, since we have strict inequality in (39),

$$\operatorname{Im} \lambda_1(\delta_c) = \frac{1}{2}\left[-\Lambda(\delta_c)\right]^{1/2} \neq 0$$

These are requirements i), ii), and iii), respectively.

Moreover, from (26) we have

$$\Gamma'(\delta_c) = \frac{(z_s^2 - Md)}{M(z_s + d)^2} = \frac{1}{\gamma(z_s + d)} \neq 0$$

and therefore $\operatorname{Re} \lambda'_1 (\delta_c) \neq 0$ which is requirement iv). Finally, the remaining eigenvalue is $\lambda_3 = -1 < 0$.

Thus, all requirements for Hopf bifurcation are met. For δ in some open interval $(\delta_c , \delta_c + \varepsilon)$, the system of equations (14) - (16) with $\alpha = 0$ will have a periodic solution bifurcating from its steady state (x_s, y_s, z_s). For the system of equations (14) - (18) with $\alpha \neq 0$, this means that if conditions (31) - (34) are satisfied a Hopf bifurcation occurs on top of the existing periodic solution (due to the eigenvalues $\pm i\omega$) giving rise to solution trajectory on a 2-torus in the five dimensional phase space.

With the above choice of parametric values, Hopf bifurcation occurs at a non-washout steady state (x_s, y_s, z_s), namely $y_s = \beta \geq 0$ and from (23),

$$z_s = \frac{M\theta}{1-\theta} > 0 \qquad (42)$$

since $\frac{M\theta}{1-\theta} > 0$, with θ chosen to be less than $1/\beta \quad 1$. Then, the value of d can be determined from (30) as

$$d = \frac{\gamma z_s^2 - z_s M}{M(\gamma + 1)} \qquad (43)$$

Since

$$(\gamma z_s^2 - z_s M) \qquad (M z_s^2 - z_s M) \;=\; M\,(z_s^2 - z_s)$$

and $z_s > 1$ by the second inequality in (34), we have $d > 0$.

With these values of γ, β, θ, z_s, and d, the critical value δ_c can be found from (29). It is important to note that with our choice of γ ,

$$\theta < \theta_1 = \frac{1+\sqrt{\gamma/(\gamma+1)}}{\beta} < \frac{2}{\beta}$$

since $\frac{\gamma}{(\gamma+1)} < 1$. Therefore $2 - \theta\beta > 0$ so that the value of δ_c given by (29) will be positive.

The parametric value $\delta > 0$ is then chosen to be in the interval $(\delta_c, \delta_c+\varepsilon)$ for some small $\varepsilon > 0$ so that Hopf bifurcation may occur. Then, x_s can be determined from (20) and (24) as

$$x_s = \delta\,(M + z_s)\,z_s/M > 0 \qquad (44)$$

Then, from (20) and (21) we find that

$$\rho = \frac{x_s(M+z_s)}{z_s} - \beta x_s$$

That is,

$$\rho = x_s\,(1 - \theta\beta)\,/\,\theta \qquad (45)$$

which is positive since $\theta < 1/\beta$.

Finally, from (21), we have

$$z_0 = \frac{x_s}{z_s+d} + z_s > 0 \qquad (46)$$

using the values of x_s, y_s, z_s and d found previously.

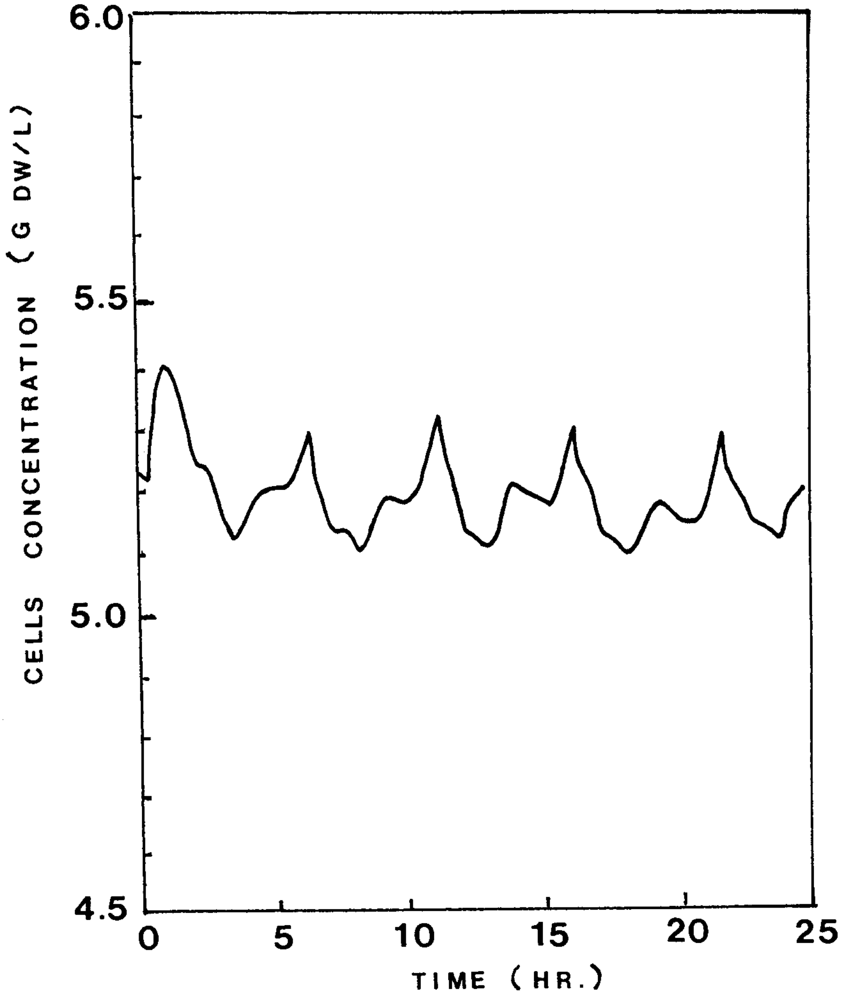

Fig. 1. Alternatively low and high peaks can be observed in the profile of cells concentration (x), for which the data points have been taken from reference [10] of continuous culture with fixed dilution rate: D = 0.2 hr^{-1}, pH = 5.5, Temp. = 30° C.

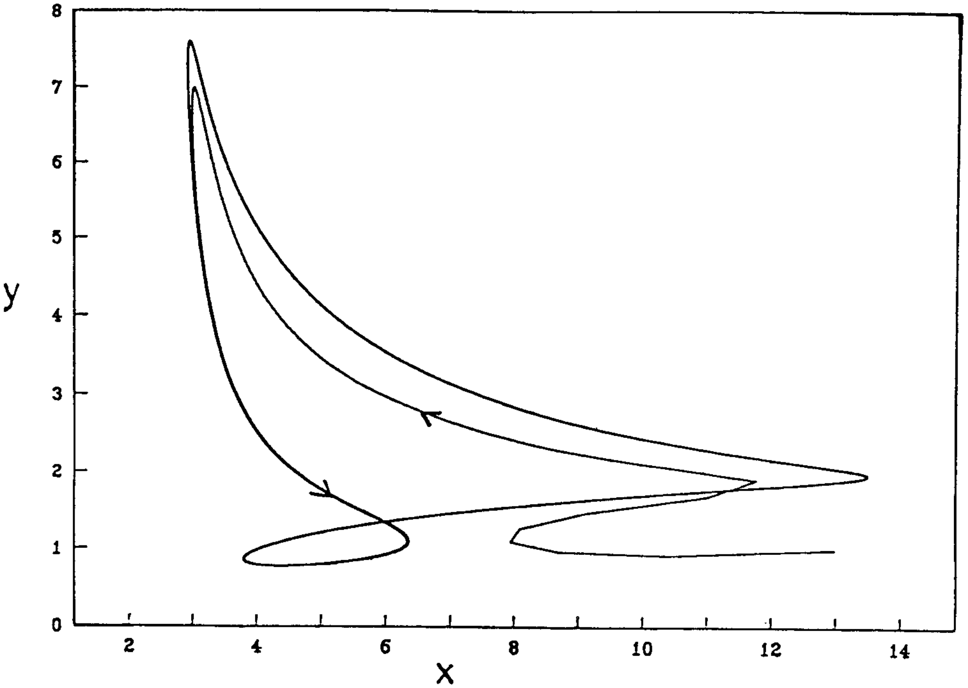

Fig. 2. Computer simulation of the model system of equations (14) - (18) with parametric values chosen so that bifurcation occurs: $M = 1$, $\gamma = 1$, $\beta = 1.5$, $\theta = 0.6$, $y_s = 1.5$, $d = 0.375$, $\delta = 2.1$, $x_s = 7.875$, $\rho = 1.3125$, $Z_0 = 5.7$, $\omega = /12$ and $\alpha = 1$. The solution trajectory, projected onto the (x, y)-plane, is seen to approach the closed curve on a torus surrounding the steady state $(x_s, y_s, z_s, u_s, v_s) = (7.875, 1.5, 1.5, 0, 0)$

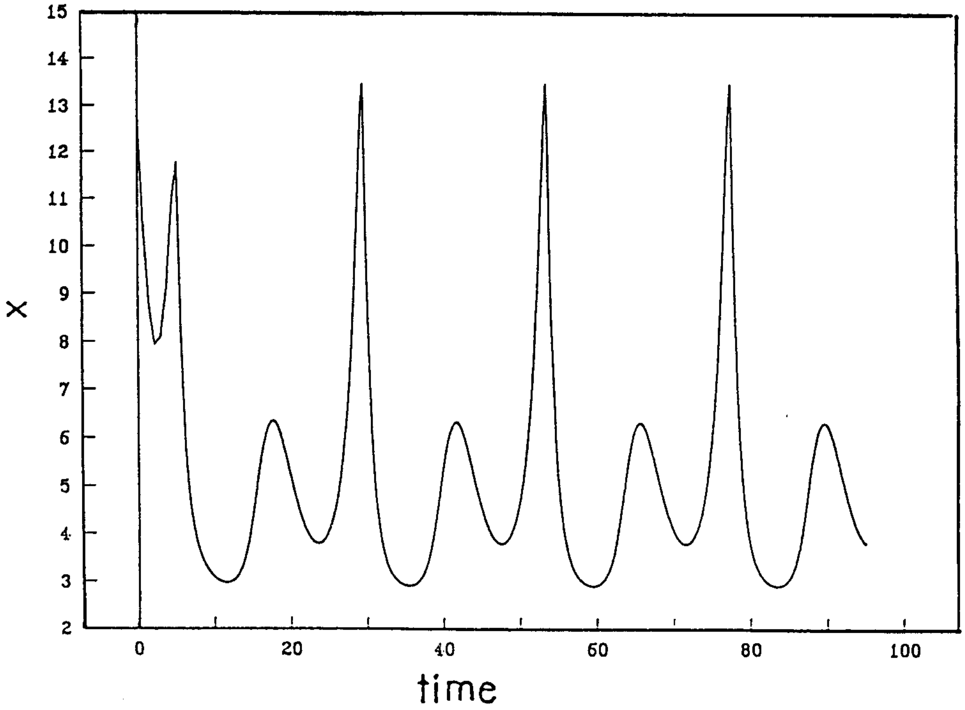

Fig. 3. The simulated time course of cells concentration x of Fig. 2 exhibiting alternatively low and high peaks resembling those observed in experimental data.

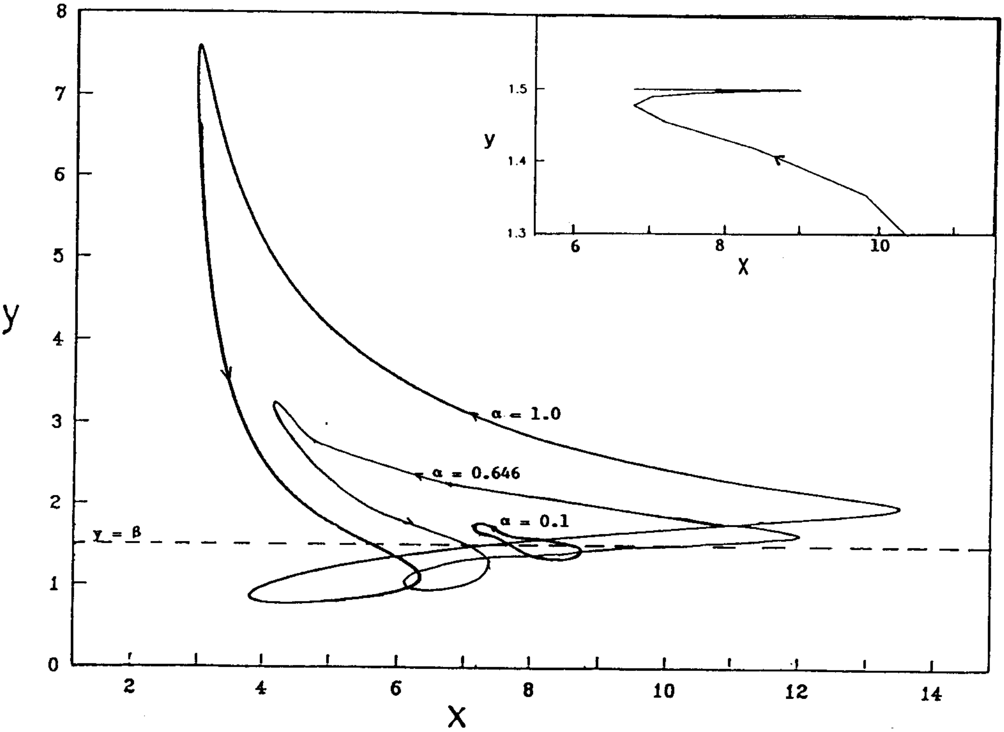

Fig. 4. The effect of varying the field density constant α. In the inset, where α = 0, the solution trajectory is seen to approach and lie on the plane y = β as time progresses.

In Figure 2, we present a computer simulation of the model equations (14) - (18) with $\alpha \neq 0$ and parametric values chosen to satisfy the bifurcation requirements (31) - (34). The solution trajectory is seen to approach the closed curve on the surface of a 2-torus surrounding the steady state $(x_s, y_s, z_s, u_s, v_s) = (7.875, 1.5, 1.5, 0, 0)$ in the 5-dimensional phase space, seen here projected onto the (x, y) - plane. The time course of cells concentration is shown in Figure 3 exhibiting alternatively low and high peaks which compares well with experimental data mentioned earlier (Figure 1). When different parametric values were tried, we have been able to generate different oscillatory patterns resembling those observed in experimental data of continuous cultures under different operating parameters [1, 10].

ASYMPTOTIC BEHAVIOR AND STABILITY ANALYSIS

On multiplying equation (14) by y, equation (15) by x, and adding, we obtain the equation

$$\frac{dw}{dT} = (\alpha u-1)w + \beta(w+\rho)\frac{z}{M+z} \qquad (47)$$

where $w = xy$. We see that equations (16) and (47) involve only the two variables w and z , and therefore can be solved without the help of equation (14). Letting $(\hat{w} (T), \hat{z} (T))$ be the solution to equations (16) and (47), equation (14) may then be written as

$$\frac{dx}{dT} = F(T) - x \qquad (48)$$

where $F(T) = (\hat{w}(T) + \rho)\frac{\hat{z}T}{M+\hat{z}T}$ is a known function of T. Equation (48) can be solved directly for the solution $x = \hat{x} (T)$.

Moreover, on substituting $z = 0$ in (16), we find that

$$\left.\frac{dz}{dT}\right|_{z=0} = z_0 \quad > \quad 0$$

which means that

$$z(T) \geq 0 \text{ for all } T \geq 0 \qquad (49)$$

Considering equation (47) with $w = 0$, we also have

$$\left.\frac{dw}{dT}\right|_{w=0} = \frac{\beta\rho z}{M+z} \geq 0$$

for positive parametric values. Thus,

$$x(T)y(T) \geq 0 \quad \text{for all } T \geq 0 \tag{50}$$

Using (49) and (50) in (48), we again have

$$x(T) \geq 0 \quad \text{for all } T \geq 0 \tag{51}$$

Therefore, we conclude that all solutions to our system model remain in the positive octant of the (x, y, z) space.

Further, with (49), (50) and (51), equation (15) can be written as

$$\frac{dy}{dT} = -\alpha u(y-\beta) - (y-\beta)G(T) - \alpha\beta u \tag{52}$$

where $G(T) \equiv \dfrac{(\hat{x}(T)\hat{y}(T)+\rho)\hat{z}(T)}{(M+\hat{z}(T))\hat{x}(T)}$ is a known function which satisfies

$$G(T) \geq 0 \quad \text{for all } T \geq 0 \tag{53}$$

Using the Liebnitz' formula to solve equation (52), we obtain

$$y(T) - \beta = e^{-\alpha v(T) - h(T)}\left\{c - \alpha\beta\int_0^T e^{\alpha v(\tau)+h(\tau)}u(\tau)d\tau\right\} \tag{54}$$

where $h(T) \equiv \int_0^T G(\tau)d\tau$ and c is a constant of integration. Since (53) holds $h(T)$ is increasing with T. Also, $e^{\pm\alpha v(T)} \leq e^{\alpha}$ since $-1 \leq v(T) \leq 1$. Thus, we have

$$\left|e^{-h(T)}\int_0^T e^{h(\tau)}u(\tau)d\tau\right| \leq e^{2\alpha - h(T)}e^{h(T)}\left|\int_0^T u(\tau)d\tau\right| \leq e^{2\alpha}$$

Thus, letting $T \to \infty$ in (54) we find

$$y(T) - \beta \to \alpha\beta e^{2\alpha} y_p(T) \text{ as } T \to \infty$$

where $y_p(T)$ is a bounded function. In other words, with $\alpha = 0$, all solutions to the system of equations (14)-(16) approach and lie, as time passes, on the plane $y = \beta$ in the (x, y, z) space.

Figure 4 shows the effect of varying the field density constant α on the position and shape of the solution trajectory. The solution trajectories for smaller α are closer to the plane $y = \beta$

With regards to the stability of these periodic solutions, one can apply various stability criteria (see, for example, [11]) on the system of equations (16) and (47) with $\alpha = 0$ which describes the solution curve $(\hat{w}(T), \hat{z}(T))$. It turns out to be very laborous calculation if one allows complete generality for the system parameters. However, for the case $\rho = 0$ and $\beta = 1$, equations (16) and (47) may be written as

$$\frac{dx_1}{dT} = \Pi(x_2)x_1 - x_1 \qquad (55)$$

$$\frac{dx_2}{dT} = -\Sigma(x_2)x_1 + x_2 \qquad (56)$$

where $x_1 = \frac{W}{z_0}$, $x_2 = 1 - \frac{z}{z_0}$,

$$\Pi(x_2) \equiv \frac{(1-x_2)}{1+\phi-x_2} \qquad (57)$$

and

$$\Sigma(x_2) \equiv \frac{\Pi(x_2)}{1+\psi-x_2} \qquad (58)$$

with $\phi = \frac{M}{z_0}$, and $\psi = \frac{d}{z_0}$

By making use of the Poincare's criterion and Friedrichs' bifurcation theory, the following condition for orbitally stable periodic solution of equations (55) and (56) can be found [8]:

$$3\Sigma'''(x_{2s}^*)\, x_{2s}^* < \Sigma''(x_{2s}^*)\left[1 + \frac{4\Pi''(x_{2s}^*)\, x_{2s}^*}{3\Pi'(x_{2s}^*)}\right] \qquad (59)$$

where x_{2s}^* is the value of $x_{2s} = 1 - \frac{z_s}{z_0}$ at the critical value δ_c of δ. That is, from (24) and (29),

$$x_{s2}^* = 1 - \frac{1}{z_0}\left[\frac{\delta}{\gamma(2-\theta)} - d\right]$$

Using (57) and (58) in (59), we find that the bifurcated periodic solution will be stable if

$$F(\phi, \psi) \quad = \quad \left[1 - x_{2s}^* - \phi\theta(1+\theta)\right]\left\{9\frac{x_{2s}^*}{\theta} - 1 - \frac{8}{3}x_{2s}^*\right\} - 9\phi x_{2s}^*\theta^2 \quad < \quad 0 \qquad (60)$$

where

$$\theta = \frac{(1 + \psi - x_{2s}^*)}{(1 + \phi - x_{2s}^*)}$$

Therefore, the bifurcation originating at the critical value δ_c of δ is stable if $F < 0$ and unstable if $F > 0$. Moreover, it can be shown that a stable bifurcated periodic solution surrounds an unstable critical point. If it surrounds a stable critical point, it is unstable.

CONCLUSIONS

A model of three ordinary differential equations is used to describe, under certain simplifying hypotheses, a membrane permeability sensitive chemostat system. Depending on the values of the system parameters, the model system may exhibit sustained regular oscillation in the form of a one frequency limit cycle, or a more irregular oscillation in the form of a solution trajectory on the surface of a torus surrounding a non-washout steady state. Thus, by incorporating the effect of membrane permeability variation, the model is shown to be capable of exhibiting oscillatory behavior which compares well with observed experimental data. A stability investigation shows that if the quantity $F(\phi, \psi)$ has positive value then the bifurcated solutions are repelling and if it is negative then the solutions are attracting.

Factors such as electric and magnetic forces have been proposed to have significant effects on cytoplasmic membrane permeability inducing oscillatory pattern in permeability which in turn causes the rhythmicity in the microbial growth patterns. Some investigations have been carried out in that direction [2, 7]. Nontheless, relatively little efforts have been made, up to date, to model such effects of rhythmic variation in membrane permeability on microbial culture, in order that their biochemical impact may be better understood and appreciated. More in depth studies of the causes and mechanism of the rhythmicities are clearly needed, the repercussions of these kind of studies in the large scale fermentation industry being significant indeed.

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