A simple unforced oscillatory growth model in the chemostat

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Abstract

In a chemostat, oscillatory behaviors during the transients are often experimentally observed for cells growth. The aim of this paper is to propose a simple autonomous model which is able to generate these oscillations, and to investigate it analytically. Our point of view is based on the simplification of the cell cycle mechanisms, emphasizing two main steps: mature and immature stages are modeled and the transfer between these two steps depends on the resource. We use the mathematical global properties of competitive differential systems to exhibit a limit cycle. A comparison between our model and a more complex PDE model is done with the help of numerical simulations, giving qualitatively similar results.

Key words: biochemical mechanisms, structured model, ordinary differential equations, chemostat, competitive system, oscillations.

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1 Introduction

A chemostat is a laboratory apparatus, composed of a reservoir and fed by a constant liquid flow, used for experiments of controlled growth of microorganisms. The inflow feeds the culture with biochemicals called substrates (e.g. nitrate). In the vessel, microorganisms grow consuming these nutrients, then the outflow retrieves substrates and cells present in the reservoir. Usually, only one substrate is limiting in order to evaluate its influence on cells growth. This idealized and controlled apparatus allows reproducible experiments and

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gives a very good approximation of specific realistic biological mechanisms (see for example (Sciandra et al., 2003)).

Unstructured mathematical models, meaning that only one global variable is used to represent the microorganisms, are often employed to describe cell growth in chemostat. The most representative one has been proposed by Monod (1942). His approach is based on the interaction between microorganisms, more precisely bacteria, and substrates dissolved in the liquid medium. Although this model could be successfully used to fit steady state, its predictions in perturbed conditions is far from being satisfactory. In particular considering *Saccharomyces cervisae* (Parulekar et al., 1986; Porro et al., 1988) or phytoplankton cells (*e.g. Chlamydomonas reinhardii*, (Nisbet and Gurney, 1982)), it is not able to represent oscillatory transients (in cell number) observed in chemostat experiments (see section 2.2). A new modeling approach is therefore required.

One idea is to consider different uptake and growth rate functions in order to obtain more complex dynamics (Arino et al, 2003). Another one is to use *structured* models, meaning that the whole cell population is described by several variables representing some physiological states (Lemesle and Gouzé, 2005). Different models could be obtained by the choice of a structured variable: if this variable is continuous, a model with partial differential equations is obtained; if this variable is discrete, the model results in an ordinary differential equations system. For example, in (Pascual and Caswell, 1997), the structured variable is continuous and represents the cell maturation along its cycle. Moreover, the cell number density is considered and the cell cycle is divided into resource-dependent and resource-independent. The obtained model is with PDE and some numerical simulations show an oscillatory behavior. In (Cazzador, 1991), a discrete variable representing two phases of the cell maturation (unbudded and budded cells) is chosen, the biomass concentrations are the variables, and numerical simulations show autonomous oscillatory behaviour. We propose in the present paper a new model inspired by both of the above models and an analytical proof of the oscillatory behavior. Our global aim is to obtain a simple unforced growth model, retaining the main qualitative biological hypotheses of the more complex model (Pascual and Caswell, 1997) and giving the same qualitative results (concerning the oscillations). The hypotheses of the model (Cazzador, 1991) are quite different: the variables are biomass concentrations and not number density, and the oscillatory behaviour is based on the complex non-linear transfer rates between two compartments, both rates depending symmetrically on the substrate concentration. Moreover the oscillatory behaviour is not mathematically proved.

After recalling some classical results on unstructured and structured models (section 2), we build, using a discrete structured variable, a model based on biochemical grounds (section 3). This model is autonomous, meaning that there is no forcing in the inputs. The cell cycle is divided in two parts describing its main phases: the mature and the immature one. The transfer between the two phases is modeled by a function depending or not on resources which is very similar to the previous assumption done by Pascual and Caswell (1997). Moreover the cell number density is considered as state variables to precisely describe cell division. Then a competitive system is built and existence of a non trivial limit cycle can be analytically proved under some assumptions. Numerical simulations are given to illustrate our results (section 4).

2 The chemostat paradigm

2.1 The chemostat

The chemostat is a vessel crossed by a constant flow where microorganisms grow. The nutrient is provided by a constant inflow F_{in} and a blend of nutrient and of microorganisms is retrieved in the constant outflow F_{out}. In the continuous well mixed cultures we consider here, the input flow rate and the output flow rate are the same ($F_{in} = F_{out} = F$).

The physically based mathematical modeling of a component dynamics p with respect to this passing flow is simple; the variation of the total component pV in the volume V (constant), is the difference between the inflow Fp_{in} and the outflow Fp:

$$\dot{\overline{pV}} = Fp_{in} - Fp \Leftrightarrow \dot{p} = dp_{in} - dp \tag{1}$$

where d the dilution rate is equal to $\frac{F}{V}$. Generally the unit of the component p is a concentration (i.e. mass of cells by volume unit or number of cells by volume unit).

2.2 The classical Monod model

The most classical chemostat model was introduced by Monod (1942) to describe the bacteria growth. Only two variables are chosen to describe the reaction occurring in the reactor vessel: the biomass X in concentration x and the limiting substrate S in concentration s.

The mathematical model is divided in two parts: the previously mentioned physical part due to the flow and the biological part, which describes the reactions in the vessel. Then the following model is obtained:

$$\left.\begin{array}{rc|l} \dot{s} = & -\alpha\mu(s)x & -ds + ds_{in} \\ \dot{x} = & \mu(s)x & -dx \\ & \text{Biological part} & \text{Physical part} \end{array}\right. \tag{2}$$

This system (2) has been extensively studied (see Smith and Waltman (1995)). Let us recall the main properties of this system. First, $\mathbb{R}_+^2$, which is of biological interest, is invariant under (2).

Let us point some qualitative biological properties of the specific growth rate $\mu(s)$. The more substrate there is, the more cells grow (*i.e.* $\mu(0) = 0$, $\mu(s)$ increasing); this implies that cells do not "lose biomass" (*i.e.* $\mu(s) \geq 0$). Moreover, cells cannot absorb more than a given quantity of substrate during a given time (*i.e.* $\mu(s)$ bounded). These properties can be summarize as follows.

Hypothesis H 2.1 $\mu(0) = 0$ *and* $\mu(.)$ *is* $\mathcal{C}^1$*, increasing and bounded.*

We will call such a function with the qualitative features **(H2.1)** a *"Monod like function"* (*e.g.* Holling type II). This specific growth rate $\mu(s)$ is often defined by the classical function $\mu(s) = \dfrac{\mu_m s}{k + s}$.

To ensure the existence of a non trivial steady state, another hypothesis is necessary.

Hypothesis H 2.2 $\mu(s_{in}) > d$

Then there exist two equilibria: an equilibrium, such that all the microorganisms in the device disappear, referred to as the washout point $(s_{in}, 0)$ and another equilibrium such that biomass population remains alive in the vessel, called the non trivial point (designated (s^*, x^*) with $x^* > 0$).

Proposition 1 *Under the hypotheses (H2.1) and (H2.2), the washout steady state is unstable and the non-trivial steady state is globally asymptotically stable in the positive orthant.*

See for example Smith and Waltman (1995) for the proof.

This simple unstructured model is very intuitive and reproduces most of bacteria population dynamics. The main restriction is that the whole physiological description is contained in the variable x. For example, this model predicts that an oscillatory behavior cannot exist. Since oscillations have been experimentally observed, more complicated models (i.e. structured models) have to be used to describe the population behaviour: the following model of Pascual and Caswell (1997) illustrates this idea.

2.3 A PDE oscillatory model

In the literature, to describe cells growth, the most useful structured variable is the size (Metz and Diekmann, 1986). But if the cell division is taken into account, a more specific variable should be considered. In (Pascual and Caswell, 1997), this new variable is the position along the cell cycle. Moreover, to specify the description of the cell division, cells number density is modeled. A dynamical system given by partial differential equations based on the Monod hypotheses is derived and gives interesting numerical results:

$$\begin{aligned}
&\frac{\partial x}{\partial t} + \frac{s}{1+s}\frac{\partial x}{\partial p} = -(d + b(p))x \text{ for } p \in [p_0; p_c]\\
&\frac{\partial x}{\partial t} + \nu\frac{\partial x}{\partial p} = -(d + b(p))x \text{ otherwise}\\
&\frac{ds}{dt} = d(s_{in} - s) - \frac{s}{1+s}x_{tot}
\end{aligned}$$

where s is the substrate, $x_{tot} = \int_p x(t,p)dp$ the total population, p gives the position along the cell cycle, $b(p)$ the division rate, d the dilution rate, s_{in} the inflow substrate. The variable p is normalized so that $p_0 = 0$ and the average cell divides at $p = 1$. The cell cycle is divided into resource-dependent and resource-independent segments separated by a transition point p_c.

The cell division is described by a boundary condition for the newborn cell at $p = 0$

$$\nu x(0,t) = 2\int_p b(p)x(p,t)dp$$

where each cell divides into two immature cells.

Proposition 2 (see Pascual and Caswell (1997)) *For some parameters values (using the numerical method known as the escalator boxcar train (de Roos, 1988)), an oscillatory behavior is observed. Disappearance of oscillations depends on the value of* d *or* s_{in}.

Let us remark that some local mathematical results ensure the existence of exponentially periodic behavior (Webb, 1995) for such a structured model of cell population dynamics without cell division (the division rate could easily be incorporated into the analysis). Yet, the analysis of such models (specially the global analysis of non linear PDE models) is often rather difficult or even intractable.

In the following, since our purpose is to capture the essential qualitative mechanisms of the cell cycle, we want to built a model which is easy to study analytically. To do this, an approach based on ordinary differential equations is followed and a structured model is built under the same main assumptions as the above system.

3 An ODE oscillatory model

3.1 The model

Two main steps are considered in our model. The first step correspond to the immature phase and the second one to the mature one. In each stage, the cell density number is modeled: using this unit the division cell (one mature cell gives two immature cells) can be easily described.

The transfer between the two compartments is given by different maturation and division rates depending or not on the limiting substrate. The maturation rate depends on the substrate, but the division rate does not. The uptake nutrient rate function is different from the maturation rate meaning that the energy used for growth is assumed to be different from the energy furnished by consumption of substrate. Thus the model does not have a conservative form.

All these biological phenomena are described by the following diagrams.

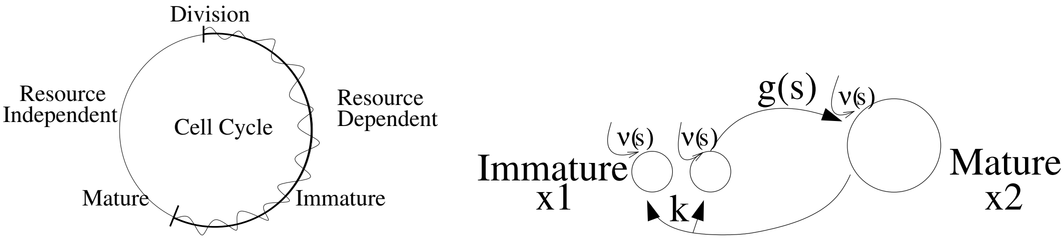

Figure 1. (left) Two steps representing the cell cycle : substrate dependence and independence. (right) Biochemical diagram of the biological mechanisms : transfer between mature and immature cell compartments.

That leads to the model :

$$\begin{array}{rr|l} \dot{x_1} = & -g(s)x_1 + 2kx_2 & -dx_1 \\ \dot{x_2} = & g(s)x_1 - kx_2 & -dx_2 \\ \dot{s} = & -\nu(s)(x_1 + x_2) & -ds + ds_{in} \\ & \text{Biological part} & \text{Physical part} \end{array} \qquad (3)$$

where x_1 is the density of the immature cells number, x_2 of the mature one;

$k > 0$ is the division rate, $g(s)$ the maturation rate , $\nu(s)$ the uptake rate function.

We choose initial conditions which have a biological meaning $x_1(0) > 0$, $x_2(0) > 0$ and $s \geq 0$.

As in the Monod model, some mathematical and qualitative hypotheses on the specific maturation rate $g(s)$ and the uptake rate $\nu(s)$ have to be done.

Hypotheses H 3.1

(a) $g(s) \geq 0$.
(b) $g(s)$ *is an increasing function.*
(c) There exists $\underline{s}$ *and* $\overline{s}$ *such that* $g'(s) \gg 1$ *for* $\underline{s} \leq s \leq \overline{s}$.
(d) $g(s)$ *is bounded.*
(e) $g(0) = 0$.
(f) $\nu(0) = 0$ *and* $\nu(.)$ *is* $\mathcal{C}^1$*, bounded, positive and increasing.*
(g) The domain of study of the system (3) is $\mathbb{R}^3_+$.

Thus, the function $g(s)$ is assumed to be sigmoïdal. For example, we can take $g(s) = \dfrac{\alpha s^a}{1 + s^a}$ with $a > 1$ and $\alpha > 0$. This formulation for the maturation rate means that cells need to have enough substrate to maturate, with a threshold effect. This is often observed in cells growth experiments. Moreover, the fact that the slope $g'(s) \gg 1$ means that we are free to choose the slope as large as desired; that will be useful in the convergence proofs.

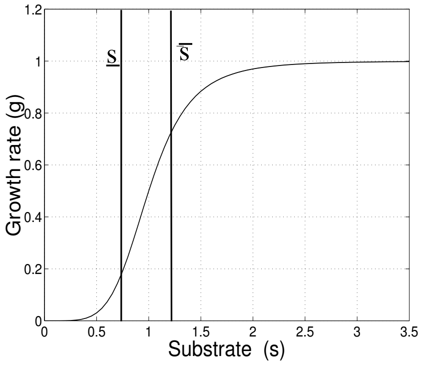

Figure 2. Maturation rate function ($\alpha = 1$)

We add two useful hypotheses:

Hypothesis H 3.2 $k - d > 0$ *and* $\dfrac{d(k + d)}{k - d} < g(s_{in})$.

Hypothesis H 3.3 $\underline{s} < s^* < \overline{s}$

The first hypothesis ensures the existence and the uniqueness of a non trivial equilibrium point. The second one implies that $g'(s^*) \gg 1$ meaning that the slope of the maturation function is very stiff at equilibrium.

3.2 Existence of steady states, boundedness and invariant set

The set of study of the system (3) is $\mathbb{R}^3_+$ since it is of biological interest.

Proposition 3 *Under (H3.2) two steady states exist for the system (3):* $(0, 0, s_{in})$ *referred to as the washout point (i.e. whole population disappearance),* (x_1^*, x_2^*, s^*) *referred to as the non trivial point (i.e. population persists).*

PROOF. Computing the steady states, we find that :

- $(0, 0, s_{in})$ is always a solution.

- a positive equilibrium (x_1^*, x_2^*, s^*) such that $x_1^* > 0$, $x_2^* > 0$, $s^* > 0$ exists and is unique. Indeed,

$g(s^*) = \dfrac{d(k+d)}{k-d}$ has a unique solution under the hypothesis (H3.2) and because g is strictly increasing.

Let us remark that $g(s_{in}) > g(s^*)$, and since g is strictly increasing $s_{in} > s^*$.□

Proposition 4 *Under the hypotheses (H3.1), the closed domain of study* $\mathbb{R}^3_+$ *is invariant for (3).*

PROOF. Computing the dynamical equations $\dot{x}_1$, $\dot{x}_2$ and $\dot{s}$ at $x_1 = 0$, $x_2 = 0$ and $s = 0$ respectively and showing that the edges are repulsive, we can prove the invariance of the set $\mathbb{R}^3_+$ for the system (3). □

Remark 5 *Consider the subset* $U = \mathbb{R}^2_+ \times [0, s_{in}]$. *We can prove that this set is invariant for the system (3). Indeed, computing the dynamical equation* $\dot{s}$ *at* s_{in}, *we obtain* $\dot{s}_{(s=s_{in})} = -\nu(s_{in})(x_1 + x_2) < 0$.

Proposition 6 *In the domain* $\mathbb{R}^3_+$, *the state variables are bounded.*

PROOF. Let $c > 0$ be defined such that for all $s \geq 0$, $c > \dfrac{g(s)}{\nu(s)}$ (see the next remark). Let $V = x_1 + 2x_2 + cs$ be a positive definite function. Then we obtain:

$$\dot{V} = -dV + (g(s) - c\nu(s))x_1 - \nu(s)x_2 + dcs_{in} < -dV + dcs_{in}$$

so that

$$V(t) < (V(0) - cs_{in})e^{-dt} + cs_{in}.$$

Then for $V(0) > cs_{in}$, $V(t) < V(0)$ and for $V(0) \leq cs_{in}$, $V(t) \leq cs_{in}$. Thus the variables x_1, x_2, s are bounded. $\square$

Remark 7 *The existence of a positive constant c is always possible even near* $s = 0$. *Indeed since* $g(s)$ *is a sigmoïdal function and* $\nu(s)$ *a Monod like function, near* 0, $g(s) \sim ks^a$ *and* $\nu(s) \sim k's$ *with* $k > 0$, $k' > 0$, $a > 1$. *Thus* $\dfrac{g(s)}{\nu(s)}$ *exists on* $\mathbb{R}_+$ *and can be bounded.*

3.3 Local Analysis and persistence

In order to study the local behavior of (3), some classical techniques of first order linearization will be used. We prove that the washout point is unstable and then a detailed study of its stable and unstable manifolds will be done.

Proposition 8 *Under the hypothesis (H3.1), the washout point* $(0, 0, s_{in})$ *is unstable.*

PROOF. Computing the associated Jacobian matrix at the washout point, we find that $-d$ is an eigenvalue and the other two verify the system :

$$\lambda_1 + \lambda_2 = -g(s_{in}) - d - k - d$$
$$\lambda_1 \lambda_2 = g(s_{in})(-k + d) + d(d + k)$$

Using the hypothesis (H3.2), we prove that one eigenvalue is positive and the other is non positive. We can conclude that the washout point is unstable. $\square$

In the following a more detailed local study of its stable and unstable manifolds is proposed. Thus the invariance of the interior of $\mathbb{R}^3_+$ will be ensure (for more details, see the definition of persistence and the theorem of Butler and MacGehee in (Smith and Waltman, 1995) and see (Lemesle, 2004)).

Proposition 9 *The only eigenvector intersecting* $\mathbb{R}^3_+$ *is the vector associated with the positive eigenvalue. The stable manifold of the washout equilibrium does not intersect the positive orthant.*

PROOF. The eigenvectors are computed solving the following system:

$$J_{wo} v_i = \lambda_i v_i \text{ for } i = 1, 2, 3$$

with $\lambda_1 > 0$, $\lambda_2 < 0$ and $\lambda_3 = -d < 0$. Then we obtain:

$$v_{\lambda_1} = \begin{pmatrix} \dfrac{k+d+\lambda_1}{g(s_{in})} \\ 1 \\ -\dfrac{\nu(s_{in})}{d+\lambda_1}(v_1+1) \end{pmatrix} \quad v_{\lambda_2} = \begin{pmatrix} \dfrac{k-g(s_{in})-\sqrt{\Delta}}{2g(s_{in})} \\ 1 \\ \dfrac{\nu(s_{in})(k+g(s_{in})-\sqrt{\Delta})}{g(s_{in})(g(s_{in})+k+\sqrt{\Delta})} \end{pmatrix} \quad v_{-d} = \begin{pmatrix} 0 \\ 0 \\ 1 \end{pmatrix}$$

with $\Delta = (g(s_{in}) + 2d + k)^2 + 4(g(s_{in})(k-d) - d(d+k))$.

We find that the local stable manifold of the washout point and $\mathbb{R}^3_+$ intersect over the set $x_1 = 0$ and $x_2 = 0$. Moreover, the local unstable manifold intersects the domain $\mathbb{R}^3_+$. □

Corollary 10 *Under the hypothesis (H3.1), the system (3) is dissipative in the open positive orthant, and is uniformly persistent.*

PROOF. Using the proposition 6, all the variables are bounded. Uniform persistence follows from the above proposition and from the fact that there is no heteroclinic cycle in the boundary (see (Smith and Waltman, 1995)). □

3.4 Global Analysis

To study the stability of the non trivial point when it exists, we make the change of variables $z = ln(x_1 + x_2)$, $u = \frac{x_1}{x_1 + x_2}$, $s = s$. Because of the dissipativity and the uniform persistence of the original system, the new system is well defined in the open set $D_1 = \mathbb{R} \times \mathbb{R}^*_+ \times \mathbb{R}^*_+$, and, moreover, there exists a compact subset $B \in D_1$ which attracts all solutions starting in D_1. From a biological point of view, we remark that this change of variables amounts to take the total number of cell, and the proportion of mature cells, as the new variables.

We obtain the new dynamical system:

$$\begin{aligned} \dot{z} &= k(1-u) - d \\ \dot{u} &= -g(s)u + k(1-u)(2-u) \\ \dot{s} &= -\nu(s)e^z - ds + ds_{in} \end{aligned} \tag{4}$$

with its associated Jacobian matrix

$$J = \begin{pmatrix} 0 & -k & 0 \\ 0 & -g(s) - k - 2k(1-u) & -g'(s)u \\ -\nu(s)e^z & 0 & -\nu'(s)e^z - d \end{pmatrix}$$

This new system is competitive in D_1 since all the off-diagonal terms of the Jacobian matrix are non positive (Smith, 1995). Moreover this system is irreducible in D_1 since J is an irreducible matrix.

Proposition 11 *Under the hypothesis (H3.3), the non trivial equilibrium* (z^*, u^*, s^*) *is unstable. Moreover, the stable manifold is one-dimensional.*

PROOF. Let J^* the associated Jacobian matrix computed at (z^*, u^*, s^*) which verifies $k(1-u^*) = d$.

The number of positive eigenvalues are computed using the Routh criterion for the J^* characteristic polynomial, see (Hofbauer and Sigmund, 1988). We compute the number of sign changes in the first Routh column. This first column is given by

$$1, \quad a_1, \quad \frac{a_1a_2 - a_3}{a_1}, \quad a_3$$

with $a_1 = -\mathrm{tr}(J^*)$, a_2 the sum of the three principal 2×2 minors of J^* and $a_3 = -\det(J^*)$. We obtain:

$$\begin{aligned} a_1 &= g(s^*) + k + 2d + \nu'(s^*)e^{z^*} + d > 0 \\ a_2 &= (g(s^*) + k + 2d)(\nu'(s^*)e^{z^*} + d) > 0 \\ a_3 &= g'(s^*)u^*\nu(s^*)ke^{z^*} > 0 \end{aligned}$$

Computing $a_1a_2 - a_3$:

$$(g(s^*) + k + 2d + \nu'(s^*)e^{z^*} + d)(g(s^*) + k + 2d)(\nu'(s^*)e^{z^*} + d) - g'(s^*)u^*\nu(s^*)ke^{z^*} \tag{5}$$

and using (H3.3), since $g'(s^*) \gg 1$, the second part of equation (5) can be made very large non positive, and thus $a_1a_2 - a_3$ will be non positive.

Thus since there are two sign changes in the Routh column, there are two eigenvalues with a non negative real part meaning that the stable manifold is one-dimensional. $\square$

To prove the existence of a stable limit cycle, the following theorem (Zhu and Smith, 1994) using competitive property applies:

Theorem 12 (see Zhu and Smith (1994)) *Let* $\dot{x} = f(x)$ *be a dissipative, irreducible and competitive system in an open subset* $D \subset \mathbb{R}^3$*. Moreover, let* D *be a* $p-$*convex system of* $\mathbb{R}^3$*. Assume that* D *contains a unique equilibrium point* ξ^* *and* $det(J^*) < 0$*. Then either*

(i) ξ^* *is stable, or*
(ii) there exists a non trivial orbitally stable periodic orbit in D*.*

Let us denote the non trivial steady state $(x_1^*, x_2^*, s^*) = \xi$ and consider (4) in the open set D_1.

Proposition 13 *Under the hypotheses (H3.2), a non trivial stable periodic orbit exists for (4) in* D_1*.*

PROOF. To prove this proposition, let us verify the hypotheses of the above theorem 12. We compute the Jacobian matrix J at ξ.

(a) The system is dissipative, irreducible and competitive using proposition 6 and corollary 10.

(b) D_1 is $p-$convex by definition, contains a unique equilibrium point ξ^*.

(c) Since J^* has two non negative eigenvalues, xi^* is unstable and moreover $det(J^*) < 0$ (see proof of proposition 11).

Thus since all the hypotheses of theorem 12 are verified, a non trivial stable positive orbit exists for (4), and therefore for system (3). □

Remark 14 (The dilution rate as a bifurcation parameter) *An important hypothesis ensuring the existence of the limit cycle is* $g'(s^*) \gg 1$*. Recall that the non trivial point is defined such that:*

$$g(s^*) = \frac{d(k+d)}{k-d} = f(d)$$

where $f(d)$ *is an increasing function of* d*. When the dilution rate* d *varies,* s^* *can leave the interval* $[\underline{s}, \overline{s}]$ *and* $g'(s*)$ *may become smaller (see Figure 3). Thus for smaller or higher dilution rate, the limit cycle may disappear and the non trivial equilibrium become stable.*

4 Simulations studies

We choose for parameters values $k = 0.6$, $s_{in} = 10$, $g(s) = \dfrac{s^{18}}{1+s^{18}}$ and $\nu(s) = \dfrac{s}{1+s}$, which verify *(H3.3)* for $d = 0.02$, 0.1 and 0.25. This values correspond to classical order values usually taken in growth phytoplankton chemostat experiments. Notice that the exponent 18 put in the formula of $g(s)$ has to be large so that all the mathematical hypotheses (in particular inequality 5) are verified.

A limit cycle can be observed for $d = 0.1$ and disappears for higher dilution values. Indeed, an approximative study of the function $g(s)$ and its derivative provide $\underline{s} \sim 0.9$, $\overline{s} \sim 1$; then $g(0.9) = 0.07$, $g(1) = 0.5$ and therefore if $0.07 \gtrsim f(d) \gtrsim 0.5$, there is no limit cycle. With the chosen values we have $f(0.02) \sim 0.02$, $f(0.1) \sim 0.14$ and $f(0.25) \sim 0.6$. Thus for $d = 0.02$ and $d = 0.25$, the limit cycle disappears.

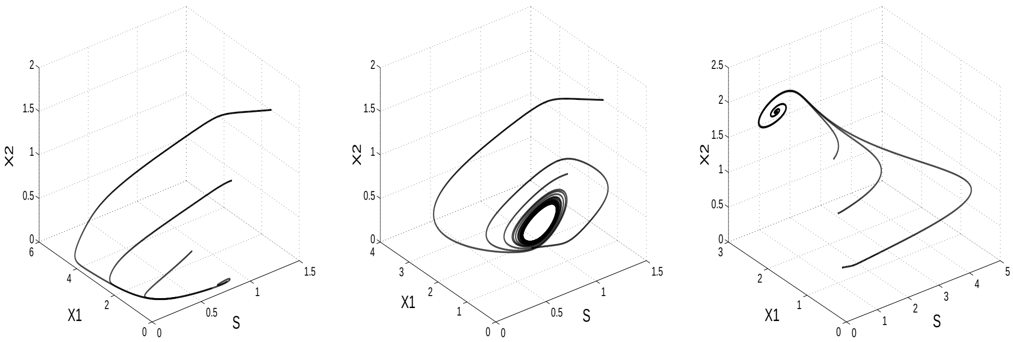

Figure 3. Phase diagram of the model. Disappearance of the limit cycle for different values of d ($d = 0.02$, $d = 0.1$, $d = 0.25$)

4.1 Qualitative comparison with the PDE model

Our model is qualitatively able to reproduce the the results obtained by (Pascual and Caswell, 1997) with the PDE model recalled in section 2.3. For similar parameters value of (d and s_{in}), the same qualitative results can be observed. Indeed, figure 4.1 shows the evolution of the total cells number for our model and we can see that as the dilution value increases (*resp.* decreases), the limit cycle disappears and as the s_{in} value increases (*resp.* decreases), the limit cycle disappears. These two figures have to be compared with the figures 3 and 6 of (Pascual and Caswell, 1997), and are qualitatively the same.

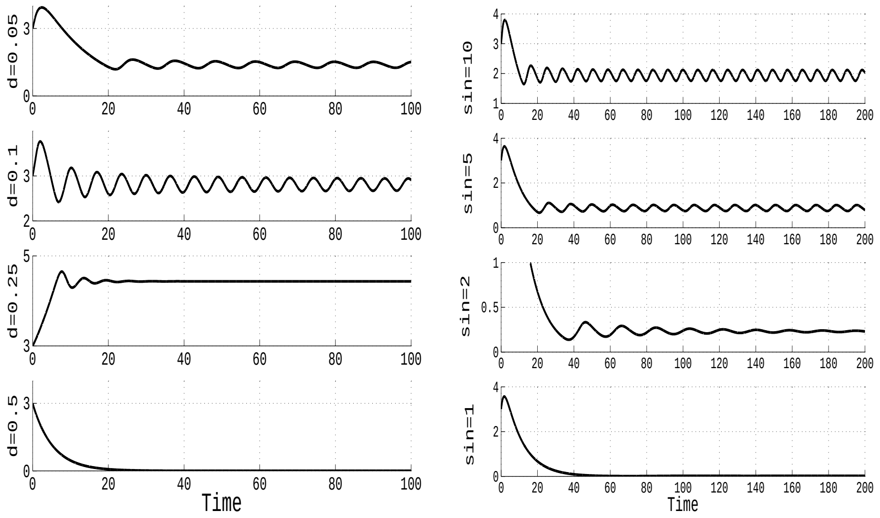

Figure 4. Total cells number (mature + immature). On the left part, different values of d are taken for $s_{in} = 10$. On the right part, different values of s_{in} are taken for $d = 0.1$. For $s_{in} = 2$, we scaled the figure to see the damped oscillations.

5 Conclusion

The classical unstructured Monod model is not able to explain all biological observations of continuous cultures of cells. Various modifications have been proposed to improve its accuracy by introducing a new descriptive continuous variable: for example, the continuous position of the cell along its cycle. This structure has been proposed to explain the oscillatory behavior observed during experiments. In this paper a different approach is considered. The structured model is based on a discrete variable describing the two main stages of cell cycle: mature and immature stages.

Finally a comparison with some simulations of the model in PDE shows similarities: we obtain oscillatory behavior depending on the dilution rate d or on the inflow value s_{in}. However our model based on a discrete structured variable is very simple and can be analytically studied.

The mathematical study of this system proves existence of a limit cycle using some properties of competitive systems. More work is needed to be able to prove the uniqueness of the limit cycle, that was clear in the simulations.

All the properties will be the same if we add a constant mortality term in the system. This mortality rate could be due to the death of cells which occurs before leaving the chemostat. Let m denote the mortality rate, the model becomes:

$$\begin{array}{rl|l}
\dot{x}_1 = & -g(s)x_1 + 2kx_2 - mx_1 & -dx_1 \\
\dot{x}_2 = & \;\; g(s)x_1 - kx_2 - mx_2 & -dx_2 \\
\dot{s} = & \qquad -\nu(s)(x_1 + x_2) & -ds + ds_{in}
\end{array}$$

If m is constant and not too large, the properties remain the same as above. More complicated behaviors may appear if this mortality rate depends on the substrate (for more details see (Lemesle, 2004)).

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