

Continuous Selection of the Fastest Growing Species in the Chemostat

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Abstract: This paper proposes control laws for the continuous culture of microorganisms, which make it possible to select species which maximize a criterion. In particular, by controlling the dilution rate and the input substrate concentration, the species with the fastest growth rate in chosen environmental conditions can be selected.

In a first step a control is proposed for Monod and Droop models in order to achieve periodic substrate stresses, and a closed loop control is proposed to regulate the total biomass concentration. We show that this biomass regulation causes the selection of the fastest growing species if the system has a periodic behavior, and derive new selection criteria. Finally, the method is simulated using the Droop model for selecting species which maximize these criteria.

Keywords: chemostat; microorganisms; competition; selection

1. INTRODUCTION

The chemostat is an open bioreactor where a microorganism can grow in suboptimal conditions of substrate limitation. The chemostat model supports several ecological theories (Jessup et al. [2005]) that were then extrapolated and tested in real life. Among these theories, the competition theory is one of the most famous. It states that if n competing species are introduced in a chemostat, generically (in the adequate working modes) only one species will stay in the chemostat, while the $n - 1$ other will disappear. This principle was validated with real experiments in Hansen and Hubell [1980] where the species that "wins" the competition could be predicted; it was the one that could grow at a constant rate (equal to the dilution rate as detailed further on) with the smallest amount of limiting substrate s^* . This theoretical result could be used to select among a blend of species, the ones of interest. This idea has been used for diverse applications, such as Directed Evolution for strain improvement (see the reviews of Dykhuizen and Hartl [1983], Zelder and Hauer [2000] and Percival Zhang et al. [2006]). By adjusting a particular stress such as an inhibitor's concentration, a substrate limitation, or the dilution rate, it can lead to the isolation of species with optimal yield. This is of particular importance since it is a rather simple way of making a particular species emerge within a population. However, in most biotechnological applications, the selection criterion based on the idea of "optimal yield" is not appropriate. A selection that would e.g. select the microorganism with the highest growth potential would be preferable, especially if one wants to identify organisms that grow in hostile conditions. More generally, the objective of this paper is to propose new selection criteria. For this, the chemostat is not run in open loop, but a control law is proposed to run the system in closed loop. We show how the competition outcome is

modified and we propose new criteria that could be used for species separation.

The paper is structured as follows. In a first part we recall two classical models of microorganisms in the chemostat, and the classical competition principle. Then we propose control laws to generate periodic substrate stresses, and to put the chemostat into a turbidostat mode. In the third part we show that this last control causes the selection of the fastest growing species if the system has a periodic behavior, and infer new selection criteria. A simulation example illustrates the benefit of the approach and shows how three species can be separated on this principle.

2. SHORT REVIEW OF COMPETITION ON A SINGLE SUBSTRATE IN THE CHEMOSTAT

2.1 Basic model for microorganisms in the chemostat

Monod model is the basic model for describing microorganisms growth on a single substrate in a chemostat.

$$\begin{cases} \dot{s} = D(s_{in} - s) - \sum_{i=1}^N \frac{1}{y_i} \mu_i(s) x_i \\ \dot{x}_i = (\mu_i(s) - D) x_i \end{cases} \quad (1)$$

where s stands for the substrate concentration in the chemostat, s_{in} its input concentration, and x_i the i th species biomass concentration. The μ_i functions represent the growth rates of these species, and the y_i constants are their substrate conversion yields. D is the input/output dilution rate. We control the system with s_{in} and D .

In this model the growth rate functions μ_i are taken as positive monotonic increasing functions

$$s_a < s_b \Leftrightarrow \mu_i(s_a) < \mu_i(s_b) \quad (2)$$

They are bounded by their supremum values μ_{m_i} :

$$\mu_i(s) < \mu_{m_i} \quad \text{and} \quad \lim_{s \rightarrow +\infty} \mu_i(s) = \mu_{m_i} \quad (3)$$

2.2 Variable yield model

Droop model (Droop [1968]) is more complex, it describes the internal substrate storage q of the microorganisms :

$$\begin{cases} \dot{s} = D(s_{in} - s) - \sum_{i=1}^N \rho_i(s)x_i \\ \dot{q}_i = \rho_i(s) - \mu_i(q_i)q_i \\ \dot{x}_i = (\mu_i(q_i) - D)x_i \end{cases} \quad (4)$$

In this model the ρ_i functions represent the substrate absorption rates, while the μ_i functions represent the growth rates. These functions are positive monotonic increasing functions. They are upper-bounded by the supremum values ρ_{m_i} and $\bar{\mu}_i$. The minimum absorption and growth rate are $\rho_i(0) = 0$ and $\mu_i(q_i) = 0$ for $q_i \in [0, K_{q_i}]$.

Internal substrate storage behavior : For each fixed substrate concentration s , the \dot{q}_i equation indicates that q_i goes toward $Q_i(s)$, defined as the unique solution of

$$\mu_i(Q_i(s))Q_i(s) = \rho_i(s) \quad (5)$$

The uniqueness of $Q_i(s)$ and its attractivity for a fixed s are straightforward since $\mu_i(q_i)q_i$ is increasing in q_i .

Let us define the maximum internal substrate storage

$$Q_{m_i} = \lim_{s \rightarrow +\infty} Q_i(s) \quad (6)$$

Q_{m_i} is thus the solution of $\mu_i(Q_{m_i})Q_{m_i} = \rho_{m_i}$.

$Q_i(s)$ is bounded in $[K_{q_i}, Q_{m_i}]$, and K_{q_i} and Q_{m_i} are lower and upper-bounds for q_i along the solutions of (4). Indeed for $q_i(t) = K_{q_i}$ (resp. $q_i(t) = Q_{m_i}$) then $\dot{q}_i \geq 0$ (resp. $\dot{q}_i \leq 0$), so that the interval $[K_{q_i}, Q_{m_i}]$ is invariant for q_i . The fact that there is a minimum and a maximum internal substrate storage for each species is biologically relevant.

Corresponding to the maximum internal substrate storage, there is a maximum growth rate μ_{m_i} for each species

$$\mu_{m_i} = \mu_i(Q_{m_i}) \quad (7)$$

2.3 The Competitive Exclusion Principle (CEP)

Under constant D and s_{in} , for some x_i species, there exists a substrate concentration s_i^* for which the growth rate μ_i at equilibrium is equal to the dilution rate D :

$$\begin{cases} \mu_i(s_i^*) = D & \text{in the Monod model} \\ \mu_i(Q_i(s_i^*)) = D & \text{in the Droop model} \end{cases} \quad (8)$$

If this substrate concentration does not exist for a given species, it means that $\mu_i(s)$ or $\mu_i(Q_i(s)) < D \quad \forall s$, and the species will be washed out of the chemostat. If $\mu_i(s_{in})$ or $\mu_i(Q_i(s_{in})) < D$ for a particular species, that species will also be washed out of the chemostat because we will have $s(t) < s_{in}$ after some finite time t_0 , and then $\mu_i(s(t))$ or $\mu_i(Q_i(s(t))) < D$ for all $t > t_0$.

With no loss of generality, we order the species so that their s_i^* satisfy : $s_1^* < s_2^* < \dots < s_N^*$.

Hypothesis 1. $s_1^* < s_{in}$ and $x_1(0) > 0$

The CEP stipulates that in a chemostat with single substrate growth limitation, constant controls D and s_{in} , and under hypothesis 1, only species 1 stays in the chemostat:

$$\begin{aligned} \lim_{t \rightarrow +\infty} x_1(t) &> 0 \\ \forall i \in [2, N], \lim_{t \rightarrow +\infty} x_i(t) &= 0 \end{aligned} \quad (9)$$

Criterion 2. CEP's Competitiveness Criterion

Under hypothesis 1, the species with smallest s_i^* , who needs less substrate than the others for obtaining an equilibrium growth rate $\mu_i(s)$ or $\mu_i(Q_i(s))$ equal to the dilution rate D , wins the competition and excludes all others from the chemostat.

Proof : A demonstration can be found in Smith and Waltman [1995] for the Monod model with N species and for the Droop model with 2 species. It has been validated with several species (Hansen and Hubell [1980] - Ducobu et al. [1998]). \square

The CEP is crucial for the understanding of natural ecosystems. Some authors (Li and Smith [2003] - Hessler et al. [2006] - Lobry and Harmand [2006]) developed alternative models for which the CEP is not verified, in order to explore some coexistence cases.

Motivations for this work

Some works have already been done to control the competition in the chemostat, generally to enable the coexistence of several species (Rao and Roxin [1990] - de Leenheer et al. [2003] - Gouzé and Robledo [2005]). Here we aim at finding controls which change the result of the competition keeping a single species. More precisely, we want to select species of interest who maximize a criterion other than the smallest s_i^* , by imposing a periodic behavior to the system.

Bernard et al. [1996] have studied the effect of periodic substrate stresses, which is a realistic ecological situation. In section 3.1 we propose controls which permit to reproduce such stresses, and in section 4.3 we show that this can cause a new selection criterion.

3. CONTROLS FOR SELECTING SPECIES

3.1 Periodic substrate stresses

We propose an approach for the generation of periodic substrate stresses. It consists in periodically imposing a phase of rising substrate concentration, followed by a phase of falling substrate concentration. The reference period is $T = t_{rise} + t_{fall}$.

Rising phase during time t_{rise}

We choose s_{in} to obtain the dynamics $\dot{s} = \lambda(s_M - s)$:

$$s_{in} = s + \frac{1}{D} \left(\lambda(s_M - s) + \sum_{i=1}^N \rho_i(s)x_i \right) \quad (10)$$

Control s_{in} is positive because $s_M > s$.

We obtain $s(t) = s_M(1 - e^{-\lambda t})$ with $s(0) = 0$

Notice that $\sum_{i=1}^N \rho_i(s)x_i$ is generally not available from measurements so that an observer may be built.

Falling phase during time t_{fall}

Let us define the total biomass concentration $X_T = \sum_{i=1}^N x_i$. We choose s_{in} such that $\dot{s} = -\gamma\phi(s)$.

$$s_{in} = s + \frac{1}{D} \left(X_T \left(\sum_{i=1}^N \frac{x_i}{X_T} \rho_i(s) \right) - \gamma\phi(s) \right) \quad (11)$$

For s_{in} to be positive, we need $X_T \sum_{i=1}^N \frac{x_i}{X_T} \rho_i(s) \geq \gamma\phi(s)$. Since $\sum_{i=1}^N \frac{x_i}{X_T} \rho_i(s)$ is a convex combination of the $\rho_i(s)$, we have for all s : $X_T \sum_{i=1}^N \frac{x_i}{X_T} \rho_i(s) \geq X_T \min_i \rho_i(s)$. Then $\phi(s) \leq \min_i \rho_i(s)$ for $s \in [0, s_M]$ and $\gamma \leq X_T$ are sufficient conditions for the positivity of s_{in} .

If γ is high enough and t_{fall} is long enough, $s(kT)$ can reasonably be considered null, which ensures the periodicity of $s(t)$ when periodically imposing control (10) followed by (11).

Periodic substrate stresses cause periodic growth rates

In this subsection we show that periodic $s(t)$ causes periodic $q_i(t)$, and thus periodic growth rates $\mu_i(q_i(t))$.

Let us remind

$$\dot{q}_i = \rho_i(s) - \mu_i(q_i)q_i \quad (12)$$

Lemma 3. Under any T -periodic $s(t)$, there exists a unique periodic solution $\bar{q}_i(t)$ to (12). This solution is attractive for any initial condition $q_i(0)$ inside $[K_{q_i}, Q_{m_i}]$

Proof : First we demonstrate that $\bar{q}_i(t)$ exists. Then we prove its attractivity.

As the q_i -attractive $Q_i(s)$ is bounded in $[K_{q_i}, Q_{m_i}]$, initial condition $q_i(0) = K_{q_i}$ causes $q_i(T) \geq q_i(0)$, and $q_i(0) = Q_{m_i}$ causes $q_i(T) \leq q_i(0)$. Thus by continuity of $q_i(T)$ with regard to the initial condition, there exists an initial condition $q_i(0) = \bar{q}_i(0)$ in $[K_{q_i}, Q_{m_i}]$ such that $\bar{q}_i(T) = \bar{q}_i(0)$ and q_i is T -periodic under controls (10) and (11)

For the attractivity, $V_i(t) = |q_i(t) - \bar{q}_i(t)|$ is decreasing along the solutions : $\dot{V}_i(t) = \text{sign}(q_i(t) - \bar{q}_i(t))(\dot{q}_i(t) - \dot{\bar{q}}_i(t)) < 0$ because $\text{sign}(\dot{q}_i - \dot{\bar{q}}_i) = \text{sign}(-\mu_i(q_i)q_i + \mu_i(\bar{q}_i)\bar{q}_i) = -\text{sign}(q_i - \bar{q}_i)$. V_i being a Lyapunov function converging to zero, $\lim_{t \rightarrow +\infty} q_i(t) = \bar{q}_i(t)$. \square

Therefore, under controls (10) and (11) q_i converges towards a T -periodic behavior, and so does $\mu_i(q_i(t))$. We will see in section 4.3 that this periodic behavior, caused by the periodic stresses, can lead to a new selection criterion.

3.2 Regulation of the total biomass concentration

To achieve the effective selection of one species, X_T must be lower bounded so that at least one microorganisms species remains in the chemostat, and it must be upper bounded to avoid saturation of the chemostat by microorganisms. That is why we propose a control D to obtain the dynamics

$$\dot{X}_T = D(X_T^* - X_T) \quad (13)$$

so that X_T converges towards a chosen concentration X_T^* .

In both models this leads us to $\sum_{i=1}^N \mu_i(\cdot)x_i - DX_T = D(X_T^* - X_T)$ and

$$D = \sum_{i=1}^N \frac{x_i}{X_T^*} \mu_i(\cdot) \quad (14)$$

For the positivity of (11) we need $\gamma \leq X_T$. It is now possible to choose $\gamma \leq \min(X_T^*, X_T(0))$.

Implementation : In practice $\sum_{i=1}^N \mu_i(\cdot)x_i$ is often an indicator of the microorganisms activity that can be measured through the evolution of influent and effluent gas composition and flow rate. For example in the case of anaerobic digestion, this would be the CH_4 flow rate; in the case of microalgae, this would be the O_2 production (or CO_2 uptake).

Remark : It is also possible to regulate $K_T = \sum_{i=1}^N k_i x_i$ with k_i coefficients who can represent turbidity coefficients. This is what is approximatively done in a particular family of chemostat, the turbidostat.

4. SELECTION OF THE FASTEST GROWING SPECIES

4.1 Selection theorem in the X_T -regulated chemostat

We have shown how to obtain periodic growth rates, and regulated X_T . In this section we will suppose that these controls or others are applied to the system so that X_T is bounded in a fixed positive interval $[X_0, X_m]$, and the growth rates are T_i -periodic functions $\mu_i(t)$.

Hypothesis 4. X_T is bounded in $[X_0, X_m]$ with $X_0 > 0$, and the $\mu_i(t)$ are T_i -periodic functions.

We use the following notation for the mean growth of the species :

$$\mu_{mean_i} = \frac{1}{T_i} \int_0^{T_i} \mu_i(t) dt \quad (15)$$

Hypothesis 5. Let us assume that $\exists j \in [1; N]$ such that $\mu_{mean_j} > \mu_{mean_i} \quad \forall i \neq j$

Theorem 6. Selection Theorem for any periodic behavior Under hypotheses 4 and 5, species j with highest mean growth μ_{mean_j} is selected and excludes all others.

Proof : We denote $d_i = \ln \left(\frac{x_j}{x_i} \right)$. With $\dot{x}_i = (\mu_i - D)x_i$ we obtain $\dot{d}_i(t) = \frac{\dot{x}_j(t)}{x_j(t)} - \frac{\dot{x}_i(t)}{x_i(t)} = \mu_j(t) - \mu_i(t)$ and

$$d_i(t) = d_i(0) + \int_0^t \mu_j(\tau) d\tau - \int_0^t \mu_i(\tau) d\tau$$

We then use the following notations :

$$\begin{aligned} p_i(t) &= \frac{t - \text{mod}(t, T_i)}{T_i} \\ r_i(t) &= \text{mod}(t, T_i) \\ t &= p_i(t)T_i + r_i(t) \end{aligned} \quad (16)$$

where $p_i(t)$ is the number of T_i -periods for species i until time t . which leads us to

$$\begin{aligned} \int_0^t \dot{d}_i(\tau) d\tau &= p_j(t) \int_0^{T_j} \mu_j(\tau) d\tau - p_i(t) \int_0^{T_i} \mu_i(\tau) d\tau + R_i^j(t) \\ &= p_j(t) \left(\int_0^{T_j} \mu_j(\tau) d\tau - \frac{p_i(t)}{p_j(t)} \int_0^{T_i} \mu_i(\tau) d\tau \right) + R_i^j(t) \\ &= p_j(t) \left(T_j \mu_{mean_j} - \frac{p_i(t)}{p_j(t)} T_i \mu_{mean_i} \right) + R_i^j(t) \end{aligned}$$

with notation $R_i^j(t) = \int_0^{r_j(t)} \mu_j(\tau) d\tau - \int_0^{r_i(t)} \mu_i(\tau) d\tau$.

Since $\frac{p_i(t)}{p_j(t)} = \frac{t - \text{mod}(t, T_i)}{t - \text{mod}(t, T_j)} \frac{T_j}{T_i}$, $\lim_{t \rightarrow +\infty} \frac{p_i(t)}{p_j(t)} = \frac{T_j}{T_i}$ and $\lim_{t \rightarrow +\infty} d_i(t) = d_i(0) + p_j(t) T_j (\mu_{mean_j} - \mu_{mean_i}) + R_i^j(t)$. Then as $d_i(0)$ and $R_i^j(t)$ are bounded, $\lim_{t \rightarrow +\infty} d_i(t) = +\infty$ because $\lim_{t \rightarrow +\infty} p_i(t) = +\infty$ and $\mu_{mean_j} > \mu_{mean_i}$.

Thus $\lim_{t \rightarrow +\infty} \frac{x_j(t)}{x_i(t)} = +\infty$ and, as x_j is upper bounded by the upper-bound on X_T , then $\lim_{t \rightarrow +\infty} x_i(t) = 0$. As X_T is also lower bounded by $X_0 > 0$, then $\liminf_{t \rightarrow +\infty} x_j(t) \geq X_0 > 0$ and the proof is complete. \square

4.2 Selection of the species with highest μ_{m_i}

We have shown that it is possible to select a species with fastest mean growth in given environmental conditions. Here we determine conditions for selecting the species with highest μ_{m_i} . Bennett and Boraas [1988] have realized such a selection by using a turbidostat culture.

Criterion 7. μ_{m_i} Selection Criterion

With a bounded total biomass concentration X_T , and with $\mu_i(t) \approx \mu_{m_i} \quad \forall t$, the species with highest μ_{m_i} is selected

Proof : The demonstration of Criterion 7 is the same as for Theorem 6, with $\mu_{mean_i} = \mu_{m_i}$. \square

Implementation : In order to have $\mu_i(t) \approx \mu_{m_i}$, we regulate s at a large value s_0 (so that $\mu_i(s_0)$ or $\mu_i(Q_i(s_0)) \approx \mu_{m_i}$) by imposing $\dot{s} = D(s_0 - s)$ which is achieved through $s_{in} = s_0 + \frac{1}{D} \sum_{i=1}^N \rho_i(s) x_i$.

Remark : From a practical point of view, as $\sum_{i=1}^N \rho_i(s) x_i$ is not always measurable, we have imagined a simpler way to realize $\mu_i(s)$ or $\mu_i(Q_i(s)) \approx \mu_{m_i}$: with a high constant s_{in} and a small X_T^* we have $\dot{s} \approx D(s_{in} - s)$ and the substrate concentration will converge close to s_{in} , which is chosen high enough to have $\mu_i(s_{in})$ or $\mu_i(Q_i(s_{in})) \approx \mu_{m_i}$.

4.3 Periodic stresses and new selection criterion

In this section we present the results that we obtained with controls (10), (11), and the Selection Theorem 6.

Functions used for the study

We have used Michaelis-Menten absorption rates $\rho_i(s)$ and Droop growth rates $\mu_i(q_i)$:

$$\begin{aligned} \rho_i(s) &= \rho_{m_i} \frac{s}{s + K_{s_i}} \\ \mu_i(q_i) &= \bar{\mu}_i \left(1 - \frac{K_{q_i}}{q_i} \right) \end{aligned} \quad (17)$$

Rectangular shaped stresses

With functions (17) and controls (10) and (11), we have obtained pseudo-rectangular shaped stresses of Figure 1.

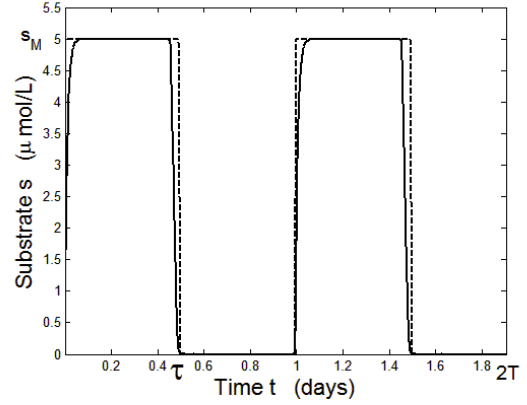


Fig. 1. Pseudo-rectangular shaped regular periodic stresses

The way to approach this regular rectangular shape as much as possible is described in section 5.

It is then reasonable (with $s_M \gg K_{s_i}$ such that $\rho_i(s_M) \approx \rho_{m_i}$) to approximate the stresses by (18)

$$\rho_i(s(t)) = \begin{cases} \rho_{m_i} & \text{if } \text{mod}(t, T) \leq \tau \\ 0 & \text{if } \text{mod}(t, T) > \tau \end{cases} \quad (18)$$

The ratio τ/T is the proportion of time the microorganisms are fed, and $(T-\tau)/T$ is the proportion of time during which they are stressed.

Determination of a new selection criterion

Under control (18) we have calculated the periodic behavior of the internal substrate storage $\bar{q}_i(t)$ presented in Appendix A, with $\lim_{t \rightarrow +\infty} q_i(t) = \bar{q}_i(t)$ (see Lemma 3)

We have then calculated :

$$\mu_{mean_i} = \frac{1}{T} \frac{\mu_{m_i}}{\bar{\mu}_i} \ln \left(1 + \frac{Q_{m_i}/K_{q_i}}{\frac{1}{e^{\bar{\mu}_i \tau} - 1} + \frac{Q_{m_i}/K_{q_i} - 1}{e^{\bar{\mu}_i T} - 1}} \right) \quad (19)$$

This criterion can be simplified in the specific case where τ is small compared to T :

Hypothesis 8. $\tau/T \ll \frac{1}{Q_{m_i}/K_{q_i} - 1}$ and $\tau \ll \frac{1}{\bar{\mu}_i \cdot Q_{m_i}/K_{q_i}}$

Criterion 9. Selection Criterion under periodic stresses

Under control (18) and hypothesis 8, the species with highest $\frac{T}{\tau} \mu_{mean_i} \approx \mu_{m_i} \frac{Q_{m_i}}{K_{q_i}}$ is selected.

where $\frac{Q_{m_i}}{K_{q_i}}$ represents the capacity of the species to increase its internal substrate storage.

Consequently, periodic stresses with a small τ make it possible to select a species which can both grow fast and increase significantly its internal substrate storage.

5. SIMULATIONS WITH THREE SPECIES

These simulations have been carried out on the Droop model (4) with control (14) so that the total biomass concentration converges toward X_T^* .

We have used (17) with the parameters of Table 1 for the species, whose values come from Vatcheva et al. [2006].

species	1	2	3
K_s ($\mu\text{mol}/L$)	0.01	0.15	0.10
ρ_m ($10^{-9} \cdot (\mu\text{mol}/(\mu\text{m})^3)/\text{day}$)	9	14	8
K_q ($10^{-9} \cdot \mu\text{mol}/(\mu\text{m})^3$)	1.5	5	2
$\bar{\mu}$ (1/day)	1.5	6	3
μ_m (1/day)	1.2	1.91	1.71
$\mu_m \cdot Q_m/K_q$ (1/day)	6	2.8	4

Table 1. Parameter values of the species

5.1 Selection of the highest μ_{m_i} with a X_T -regulation

For this simulation we have chosen a large s_{in} and a small X_T^* to ensure a large s , and thus we meet the conditions of Criterion 7 ($s_{in} = 10\mu\text{mol}/L$, $X_T^* = 1 \cdot 10^9 \cdot (\mu\text{m})^3/L$)

Figure 2 shows the result of the simulation, where species 2 with highest μ_{m_i} excludes all others.

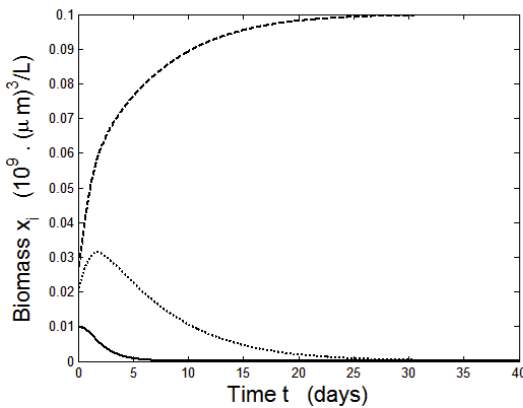


Fig. 2. Selection of the species with highest μ_{m_i}
 Lines & species : Solid-1, Dashed-2, Dotted-3

5.2 Selection under periodic substrate stresses

For these simulations we have used controls (10) and (11) with $T = 1$ day, and $s_M \gg K_{s_i}$ so that $\rho_i(s_M) \approx \rho_{m_i}$ ($s_M = 5\mu\text{mol}/L$)

Approaching rectangular shaped stresses

We chose a high λ so that s rises fast ($\lambda = 1000$). In the presence of perturbations, λ should not be taken that high, in order to avoid amplifying them.

For a fast fall we have chosen a high X_T^* , and a $X_T(0)$ close to X_T^* (this can be achieved by using control (14) long enough before starting the s_{in} control), which allowed us to choose a high $\gamma \leq \min(X_T^*, X_T(0))$:

$$X_T^* = 400 \cdot 10^9 (\mu\text{m})^3/L, X_T(0) = 220 \cdot 10^9 (\mu\text{m})^3/L, \gamma = 100 \cdot 10^9 (\mu\text{m})^3/L.$$

We also used $\phi(s) = \rho_{m_\phi} \frac{s}{s+K_{s_\phi}}$ with $\rho_{m_\phi} = 5 \cdot 10^9 \mu\text{mol}/(\mu\text{m})^3 < \rho_{m_i}$ and $K_{s_\phi} = 1\mu\text{mol}/L > K_{s_i}$ so that $\phi(s) < \rho_i(s) \quad \forall i \in \{1, \dots, N\}, \forall s$.

The method to obtain s_M -phases with duration τ is presented in Appendix B.

Selecting a species by choosing a τ value

Figure 3 shows that different species can be selected with different environmental condition τ , because for each species i here (see Table 1 for their parameter values),

there exists a τ value such that μ_{mean_i} is higher than the other species' mean growth.

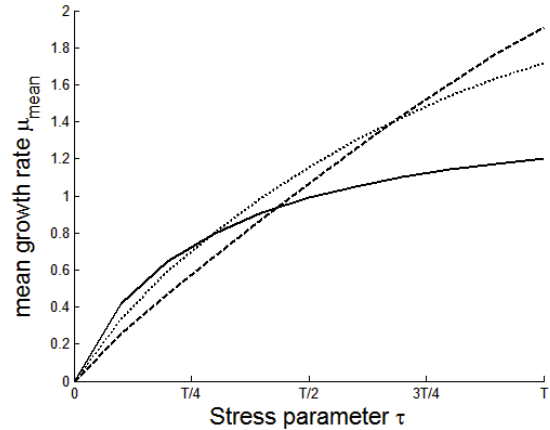


Fig. 3. $\mu_{mean_i}(\tau)$ for the three species. The species with highest μ_{mean_i} will be selected. It is thus possible to select each of the three species, by using different τ
 Lines & species : Solid-1, Dashed-2, Dotted-3

Finally, Figure 4 illustrates the possibility of selecting any of the three species by choosing an adequate τ .

6. CONCLUSION

This work shows that it is in theory possible to select species exhibiting a desired feature in the chemostat : under turbidostat-like control in constant or periodic conditions, the species with the fastest mean growth rate is selected. We have proposed two possible applications : the selection of a species that can grow faster than any other, and the selection of a species which can both grow fast and increase its internal substrate storage.

This method makes it possible to select quite easily a species for its study or culture. It could be used to identify species which grow in hostile conditions, to amplify and verify the presence of a species in a medium, to determine conditions which permit a species to be resistant to invaders, to select a species which is interesting for a specific biotechnological objective, or to improve strains by directed evolution. The selection criterion, which is the maximum mean growth rate in chosen environmental conditions, seems very promising.

This work could be extended by using other controls such as an inhibitor's concentration. This would lead to other selection criteria.

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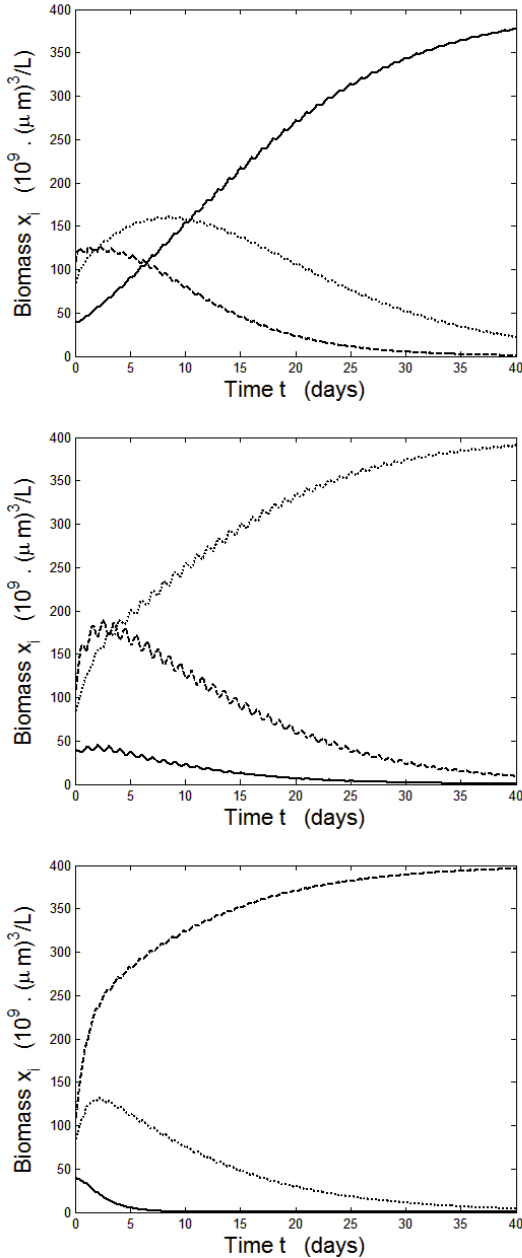


Fig. 4. Simulations of selections in the Droop model, under controls (14), (10) and (11).
top : $\tau = 0.1T$, middle : $\tau = 0.5T$, bottom : $\tau = 0.9T$
Lines & species : Solid-1, Dashed-2, Dotted-3

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Appendix A. PERIODIC INTERNAL SUBSTRATE STORAGE CAUSED BY PERIODIC STRESSES (18)

We have demonstrated that (A.1) is the unique solution of (12) under periodic substrate stresses (18)

$$\bar{q}_i(t) = \begin{cases} Q_{m_i} (1 - e^{-\bar{\mu}_i t}) + \bar{q}_i(0) e^{-\bar{\mu}_i t} & \text{if } \text{mod}(t, T) \leq \tau \\ K_{q_i} (1 - e^{-\bar{\mu}_i (t-\tau)}) + \bar{q}_i(\tau) e^{-\bar{\mu}_i (t-\tau)} & \text{else} \end{cases}$$

$$\text{with } \bar{q}_i(0) = Q_{m_i} - (Q_{m_i} - K_{q_i}) \frac{1 - e^{-\bar{\mu}_i (T-\tau)}}{1 - e^{-\bar{\mu}_i T}} \quad (\text{A.1})$$

$$\text{and } \bar{q}_i(\tau) = K_{q_i} + (Q_{m_i} - K_{q_i}) \frac{1 - e^{-\bar{\mu}_i \tau}}{1 - e^{-\bar{\mu}_i T}}$$

Appendix B. OBTAINING τ FOR (18)

To obtain regular stresses (18), we need to predict τ_{fall} , the time needed, starting from $s = s_M$, to obtain $s = s_0$ where s is negligible ($s_0 = 0.005 \mu\text{mol/L}$).

$\frac{ds}{dt} = -\gamma\phi(s) = \gamma\rho_{m_\phi} \frac{s}{s+K_{s_\phi}}$ leads us to $dt = -\frac{1}{\gamma\rho_{m_\phi}} (1 + \frac{K_{s_\phi}}{s}) ds$ and to $\tau_{fall} = \frac{1}{\gamma\rho_{m_\phi}} \left(s_M - s_0 + K_{s_\phi} \ln \left(\frac{s_M}{s_0} \right) \right)$. If the stresses are rectangular shaped ($s \approx s_M$ during most of the time interval $[0, t_{rise}]$), it is thus possible to choose t_{rise} so that $\tau = t_{rise} + \tau_{fall}$, and to approach control (18).

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