Research article

Mutual inhibition in presence of a virus in continuous culture

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Abstract: In this paper, we consider two species competing for a limiting substrate such that each species impedes the growth of the other one (Mutual inhibition) in presence of a virus inhibiting one bacterial species. A system of ordinary differential equations is proposed as a mathematical model for this competition. A detailed local qualitative analysis of the system is carried out. We proved that for a general nonlinear growth rates, the Competitive Exclusion Principle still valid, that at least one species goes extinct. For some cases where we have two locally stable equilibrium points, initial species concentrations are important in determining which is the winning species. Obtained results were confirmed by some numerical simulations using Matlab software.

Keywords: chemostat; competition; reversible inhibition; virus; local analysis; competitive Exclusion Principle

1. Introduction

A chemostat is a special type of biological continuous stirred tank reactor in which microorganisms (*Phytoplankton*, *Yeasts* ...) are placed in the presence of a limiting nutrient and other elements in non-limiting quantity. We can thus, from the variations of the limiting nutrient, know the influence of the latter on the cultivated population. The chemostat is therefore a model of a controlled ecosystem in which we can quantify precisely the relationships between a nutrient and an organism [1]. In ecology, it refers to an artificial lake for bacterial continuous culture where we can analyse inter-specific interactions between bacteria. A huge number of mathematical studies were published (see, for example, the recent monograph by Smith and Waltman [1] and the references therein). The most used mathematical system modelling the bacterial competition for a single obligate limiting substrate predicts the competitive exclusion principle [2–4], that at least one competitor bacteria loses the competition [1]. Hsu et al. [5] are among the first, in 1977, to study the problem of competition in the chemostat. They consider *n* populations in competition for the same nutrient, and show that the competitive exclusion is verified: that of the competitors who uses the better the substrate in small quantity survives, the others

are extinguished. In the case of nonmonotonic growth functions, Butler and Wolkowicz [6] show in 1985 that the competitive exclusion principle is also verified. In 1992, Wolkowicz and Lu [7] use Lyapunov functions to show that, again in the case of general shape-growth functions, but with different mortality rates. For each species, the competitive exclusion principle is further checked (the resulting equilibrium being globally stable). Li [8] has recently extended this result to an even wider class of growth functions. Finally, Smith and Waltman [9] verify in 1994 this principle for the model of Droop. This theoretical result was confirmed by Hansen and Hubbell, experimentally [10].

In many cases, the competing bacteria can produce a plethora of secondary metabolites to increase their competitiveness against other bacteria. For example, the production of *Nisin* by a number of strains of *Lactococcus lactis* to exert a high antibacterial activity against Gram-positive bacteria has been widely studied [11, 12]. This inter-specific interaction is classified as an inhibition relationship. In the same time, viruses are the most abundant and diverse form of life on Earth. They can infect all types of organisms (*Vertebrates, Invertebrates, Plants, Fungi, Bacteria, Archaea*). Viruses that infect bacteria are called *bacteriophages* or *phages*.

In this work, we extend the chemostat model [1] to general growth rates taking into account the reversible inhibition between species (mutual inhibition, i.e., each species impedes the growth of the other.) as in [2, 13–17] but in presence of a virus associated to the first species. As our study is qualitative, we suppose that the two species are feeding on a nonreproducing limiting substrate that is essential for both species. We suppose also that the chemostat is well-mixed so that environmental conditions are homogenous. We proved that with general nonlinear response functions, the mutual inhibitory relationship in presence of two species confirms the competitive exclusion principle. It is proved that at least one species goes extinct and that for some cases where we have more than one locally stable equilibrium point, the initial species concentrations are important in determining which is the winning species (see Figure 6).

The rest of the paper is structured as follows. In Section 2, we proposed a mathematical model describing two species competing for a limiting substrate with reversible inhibition in presence of a virus associated to the first species and we recall some useful results of the chemostat theory. In Section 3, the main results of the local stability analysis are presented. Finally, in Section 4, some numerical examples were presented for illustrating the obtained results confirming the competitive exclusion principle.

2. Mathematical model

Consider a mathematical system of ordinary differential equations describing two species (x_1 and x_2) competing for a limiting substrate (s) with reversible inhibition in presence of a virus (v) associated to the species x_1. We ignore all species-specific death rates and only consider the dilution rate.

$$
\begin{cases}
\dot{s} &= D(s^{in} - s) - f_1(s, x_2)x_1 - f_2(s, x_1)x_2, \\
\dot{x}_1 &= f_1(s, x_2)x_1 - Dx_1 - \alpha v x_1, \\
\dot{x}_2 &= f_2(s, x_1)x_2 - Dx_2, \\
\dot{v} &= \kappa \alpha v x_1 - Dv.
\end{cases}
\tag{2.1}
$$

Here s^{in} is the input concentration of substrate into the chemostat. D is the dilution rate and α is the rate of infection and κ is the production yield of the virus.
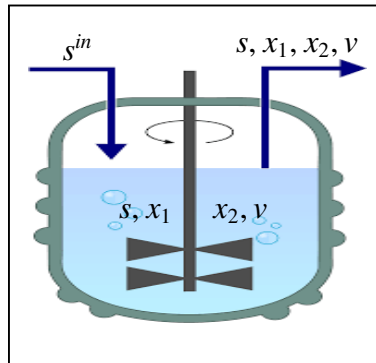
Figure 1. A chemostat is a bioreactor to which a limiting substrate (s^{in}) is continuously added, while culture liquid (s, x_1, x_2, v) are continuously removed at the same flow rate (D) [2, 18].

We can see from fourth equation of (2.1) that the condition $s^{in} > \dfrac{D}{\kappa \alpha}$ must be fulfilled in order to permits the existence of equilibrium points where the species 1 can survive with the virus.

$s(t)$ is the concentration of substrate in the chemostat at time t. $x_i(t)$ is the ith species concentration in the chemostat at time t. $v(t)$ is the virus concentration in the chemostat at time t. $f_i(s, x_j)$: is the species growth rate depending on substrate and the concentration of the other species.

For each species, the response function $f_i : \mathbb{R}_+^2 \to \mathbb{R}_+$, $i = 1, 2$ is of class C^1, and satisfies

A1 $f_1(0, x_2) = 0$ and $f_2(0, x_1) = 0$, $\quad \forall \, x_1, x_2 \in \mathbb{R}_+$,

A2 $\dfrac{\partial f_1}{\partial s}(s, x_2) > 0$, $\quad \forall \, (s, x_2) \in \mathbb{R}_+^2$ $\quad \dfrac{\partial f_2}{\partial s}(s, x_1) > 0$, $\qquad \forall \, (s, x_1) \in \mathbb{R}_+^2$.

A3 $\dfrac{\partial f_1}{\partial x_2}(s, x_2) < -\kappa \alpha < 0$, $\quad \forall \, (s, x_2) \in \mathbb{R}_+^2$, $\quad \dfrac{\partial f_2}{\partial x_1}(s, x_1) < 0$, $\qquad \forall \, (s, x_1) \in \mathbb{R}_+^2$.

Hypothesis **A1** expresses that the substrate is essential for the bacteria growth; hypothesis **A2** reflects that the growth rate increases with substrate. Hypothesis **A3** means that species inhibit each other and that the species 1 is more sensitive to the other species than to the produced virus.

The system (2.1) plus **A1**-**A3** is not a realistic model for a considered biological system. To be more realistic, we should introduce two other variables describing intermediair proteins. Each protein produced by species x_i inhibits the growth of species j where $i, j = 1, 2$ and $i \neq j$. In this case the model will be huge ($\mathbb{R}^6$) and then difficult to study.

In El Hajji [2], the author considers two species feeding on limiting substrate in a chemostat considering a mutual inhibitory relationship between both species. The proposed model is the same as the one we proposed here but with $\alpha = 0$ (no virus associated to the first species). It is proved in [2] that at most one species can survive which confirms the competitive exclusion principle. The author proved also in the case where there is two equilibrium points locally stable, the initial concentrations of species have great importance in determination of which species is the winner.

Proposition 1. *1. For every initial condition $\left(s(0), x_1(0), x_2(0), v(0) \right) \in \mathbb{R}_+^4$, the corresponding solution admits positive and bounded components and then is definite for all $t \geq 0$.*

2. $\Omega = \left\{(s, x_1, x_2, v) \in \mathbb{R}_+^4 \; / s + x_1 + x_2 + \dfrac{v}{\kappa} = s^{in}\right\}$ is an invariant set and it is an attractor of all solution of system (2.1).

Proof. 1. The solution components are positive.

If $s = 0$ then $\dot{s} = Ds^{in} > 0$ and if $x_i = 0$ then $\dot{x}_i = 0$ for $i = 1, 2$. If $v = 0$ then $\dot{v} = 0$.

Next we have to prove the boundedness of solutions of (2.1). By adding all equations of system (2.1), one obtains, for $T(t) = s(t) + x_1(t) + x_2(t) + \dfrac{v(t)}{\kappa} - s^{in}$, a single equation :

$$\dot{T}(t) = \dot{s}(t) + \dot{x}_1(t) + \dot{x}_2(t) + \frac{\dot{v}(t)}{\kappa} = D\left(s^{in} - s(t) - x_1(t) - x_2(t) - \frac{v(t)}{\kappa}\right) = -DT(t).$$

Then $T(t) = T(0)e^{-Dt}$ which means that

$$s(t) + x_1(t) + x_2(t) + \frac{v(t)}{\kappa} = s^{in} + (s(0) + x_1(0) + x_2(0) + \frac{v(0)}{\kappa}) - s^{in})e^{-Dt}. \tag{2.2}$$

Since all terms of the sum are positive, then the solution of system (2.1) is bounded.

2. The second point is simply a direct consequence of equality (2.2)

$\square$

3. Local stability analysis

In this section, the equilibria are determined and their local stability properties are established. Define the parameters $\bar{x}_1, \bar{x}_2, \bar{v}, \bar{\bar{x}}_1, \bar{\bar{x}}_2, x_2^*$ and v^* as the following:

- $\bar{x}_1$ the solution of the equation $f_1(s^{in} - \bar{x}_1, 0) = D$.

- $\bar{x}_2$ the solution of the equation $f_2(s^{in} - \bar{x}_2, 0) = D$.

- $\bar{v}$ the solution of the equation $f_1(s^{in} - \dfrac{D}{\kappa\alpha} - \bar{v}, 0) = D + \alpha\bar{v}$.

- $(\bar{\bar{x}}_1, \bar{\bar{x}}_2)$ the solution of the equations $f_1(s^{in} - \bar{\bar{x}}_1 - \bar{\bar{x}}_2, \bar{\bar{x}}_2) = f_2(s^{in} - \bar{\bar{x}}_1 - \bar{\bar{x}}_2, \bar{\bar{x}}_1) = D$.

- (x_2^*, v^*) the solution of the equations $f_1(s^{in} - \dfrac{D}{\kappa\alpha} - x_2^* - v^*, x_2^*) = D + \alpha v^*$ and $f_2(s^{in} - \dfrac{D}{\kappa\alpha} - x_2^* - v^*, \dfrac{D}{\kappa\alpha}) = D$.

Then the system (2.1) admits $F_0 = (s^{in}, 0, 0, 0), F_1 = (s^{in} - \bar{x}_1, \bar{x}_1, 0, 0), F_2 = (s^{in} - \bar{x}_2, 0, \bar{x}_2, 0), F_3 = (s^{in} - \dfrac{D}{\kappa\alpha} - \bar{v}, \dfrac{D}{\kappa\alpha}, 0, \bar{v}), F_4 = (s^{in} - \bar{\bar{x}}_1 - \bar{\bar{x}}_2, \bar{\bar{x}}_1, \bar{\bar{x}}_2, 0)$ and $F^* = (s^{in} - \dfrac{D}{\kappa\alpha} - x_2^* - v^*, \dfrac{D}{\kappa\alpha}, x_2^*, v^*)$ as equilibrium points.

Let $D_1 = f_1(s^{in}, 0), \; D_2 = f_2(s^{in}, 0), D_3 = f_1(s^{in} - \dfrac{D}{\kappa\alpha}, 0), D_4 = f_1(s^{in} - \bar{x}_2, \bar{x}_2), D_5 = f_2(s^{in} - \bar{x}_1, \bar{x}_1),$ $D_6 = f_2(s^{in} - \dfrac{D}{\kappa\alpha} - \bar{v}, \dfrac{D}{\kappa\alpha}), D_7 = f_1(s^{in} - \bar{v} - \dfrac{D}{\kappa\alpha}, \bar{v})$. Note that $D_7 < D_3 < D_1, D_4 < D_1$ and $D_5, D_6 < D_2$.

The conditions of existence of the equilibria are stated in the following lemmas.

Lemma 1. *F_0 exists allways. F_0 is a saddle point if $D < \max(D_1, D_2)$. It is a stable node if $D > \max(D_1, D_2)$.*

Proof. The proof is given in Appendix 5. □

Lemma 2. *The equilibrium point F_1 exists if and only if $D < D_1$. If $D > \max(D_3, D_5)$ then F_1 is a stable node however if $D < D_3$ or $D_3 < D < D_5$ then F_1 is a saddle point.*

Proof. The proof is given in Appendix 5. □

Lemma 3. *The equilibrium point F_2 exists if and only if $D < D_2$. If $D > D_4$ then F_2 is a stable node however if $D < D_4$ then F_2 is a saddle point .*

Proof. The proof is given in Appendix 5. □

Lemma 4. *F_3 exists if and only if $D < D_3$. If $D_5 < D < D_3$, then F_3 is then locally asymptotically stable. If $D < \min(D_3, D_5)$, then F_3 is unstable.*

Proof. The proof is given in Appendix 5. □

Lemma 5. *The situation $D < \min(D_4, D_5)$ is impossible.*

Proof. The proof is given in Appendix 5. □

Lemma 6. *An equilibrium F_4 exists if and only if $\max(D_4, D_5) < D < \min(D_1, D_2)$. If it exists then F_1 and F_2 exist and satisfy $\bar{\bar{x}}_1 < \bar{x}_1$ and $\bar{\bar{x}}_2 < \bar{x}_2$. F_4 is always a saddle point.*

Proof. The proof is given in Appendix 5. □

Lemma 7. *F^* exists if and only if $\max(D_6, D_7) < D < \min(D_2, D_3)$. If it exists then it is always unstable.*

Proof. The proof is given in Appendix 5. □

We summarize the lemmas given above in the following theorem.

Theorem 1. *A) If $\min(D_4, D_5) < D < \max(D_4, D_5)$ then*

(i) if $D_5 < D_4$ then

     1. if $D_5 < D < \min(D_2, D_4, D_7)$ then system (2.1) admits four equilibria F_0, F_1, F_2 and F_3. F_3 is a stable node however F_0, F_1 and F_2 are saddle points.

     2. if $\max(D_5, D_7) < D < \min(D_3, D_4, D_6)$ then system (2.1) admits four equilibria F_0, F_1, F_2 and F_3. F_3 is a stable node however F_0, F_1 and F_2 are saddle points.

     3. if $\max(D_5, D_6, D_7) < D < \min(D_2, D_3, D_4)$ then system (2.1) admits five equilibria F_0, F_1, F_2, F_3 and F^*. F_3 is a stable node however F_0, F_1, F_2 and F^* are saddle points.

     4. if $\max(D_3, D_5) < D < \min(D_2, D_4)$ then system (2.1) admits three equilibria F_0, F_1 and F_2. F_1 is a stable node however F_0 and F_2 are saddle points.

     5. if $D_2 < D < \min(D_3, D_4)$ then system (2.1) admits three equilibria F_0, F_1 and F_3. F_3 is a stable node however F_0 and F_1 are saddle points.

6. *if* $\max(D_2, D_3) < D < D_4$ *then system (2.1) admits two equilibria* F_0 *and* F_1. F_1 *is a stable node however* F_0 *is a saddle point.*

(ii) *if* $D_4 < D_5$ *then*

1. *if* $D_4 < D < \min(D_3, D_5)$ *then system (2.1) admits four equilibria* F_0, F_1, F_2 *and* F_3. F_2 *is a stable node however* F_0, F_1 *and* F_3 *are saddle points.*

2. *if* $\max(D_3, D_4) < D < \min(D_1, D_5)$ *then system (2.1) admits three equilibria* F_0, F_1 *and* F_2. F_2 *is a stable node however* F_0 *and* F_1 *are saddle points.*

3. *if* $D_1 < D < D_5$ *then system (2.1) admits two equilibria* F_0 *and* F_2. F_2 *is a stable node however* F_0 *is a saddle point.*

4. *if* $\max(D_4, D_6, D_7) < D < \min(D_3, D_5)$ *then system (2.1) admits five equilibria* F_0, F_1, F_2, F_3 *and* F^*. F_2 *is a stable node however* F_0, F_1, F_3 *and* F^* *are saddle points.*

B) *If* $\max(D_4, D_5) < D < \min(D_1, D_2)$ *then*

(i) *if* $\max(D_4, D_5) < D < \min(D_3, D_6)$ *then system (2.1) admits five equilibria* F_0, F_1, F_2, F_3 *and* F_4. F_2 *and* F_3 *are stable nodes however* F_0, F_1 *and* F_4 *are saddle points.*

(ii) *if* $\max(D_3, D_4, D_5) < D < \min(D_1, D_6)$ *then system (2.1) admits four equilibria* F_0, F_1, F_2 *and* F_4. F_1 *and* F_2 *are stable nodes however* F_0 *and* F_4 *are saddle points.*

(iii) *if* $\max(D_4, D_5, D_6) < D < \min(D_2, D_7)$ *then system (2.1) admits five equilibria* F_0, F_1, F_2, F_3 *and* F_4. F_2 *and* F_3 *are stable nodes however* F_0, F_1 *and* F_4 *are saddle points.*

(iv) *if* $\max(D_4, D_5, D_6, D_7) < D < \min(D_2, D_3)$ *then system (2.1) admits six equilibria* F_0, F_1, F_2, F_3, F_4 *and* F^*. F_2 *and* F_3 *are stable nodes however* F_0, F_1, F_4 *and* F^* *are saddle points.*

(v) *if* $\max(D_3, D_4, D_5, D_6) < D < \min(D_1, D_2)$ *then system (2.1) admits four equilibria* F_0, F_1, F_2 *and* F_4. F_1 *is a stable node however* F_0, F_2 *and* F_4 *are saddle points.*

C) *If* $\min(D_1, D_2) < D < \max(D_1, D_2)$ *then*

(i) *If* $D_1 < D < D_2$ *then system (2.1) admits two equilibria* F_0 *and* F_2. F_2 *is a stable node however* F_0 *is a saddle point.*

(ii) *If* $D_2 < D < D_1$ *then*

1. *if* $D_2 < D < D_3$ *then system (2.1) admits three equilibria* F_0, F_1 *and* F_3. F_3 *is a stable node however* F_0 *and* F_1 *are saddle points.*

2. *if* $\max(D_2, D_3) < D < D_1$ *then system (2.1) admits two equilibria* F_0 *and* F_1. F_1 *is a stable node however* F_0 *is a saddle point.*

D) *If* $\max(D_1, D_2) < D$ *then model (2.1) admits only* F_0 *as equilibrium point.* F_0 *is a stable node.*

4. Numerical Simulations

In this section, we validated the obtained results by some numerical simulations on a system that uses classical Monod growth rates and takes into account the reversible inhibition between species:

$f_1(s, x_2) = \dfrac{s}{(1 + s)(1 + x_2)}$, and $f_2(s, x_1) = \dfrac{s}{(2 + s)(1 + x_1)}$ with $\alpha = 0.1$ and $\kappa = 1.5$. One can readily check that the functional responses satisfy Assumptions **A1** to **A3**.

In Figure 2, if the dilution rate $D = 1$ satisfying $D_2 = 0.9 < D_1 \approx 0.95 < D = 1$, each solution with initial condition inside the whole domain converges to the equilibrium F_0 from where the extinction of the two species (point D of Theorem 1).
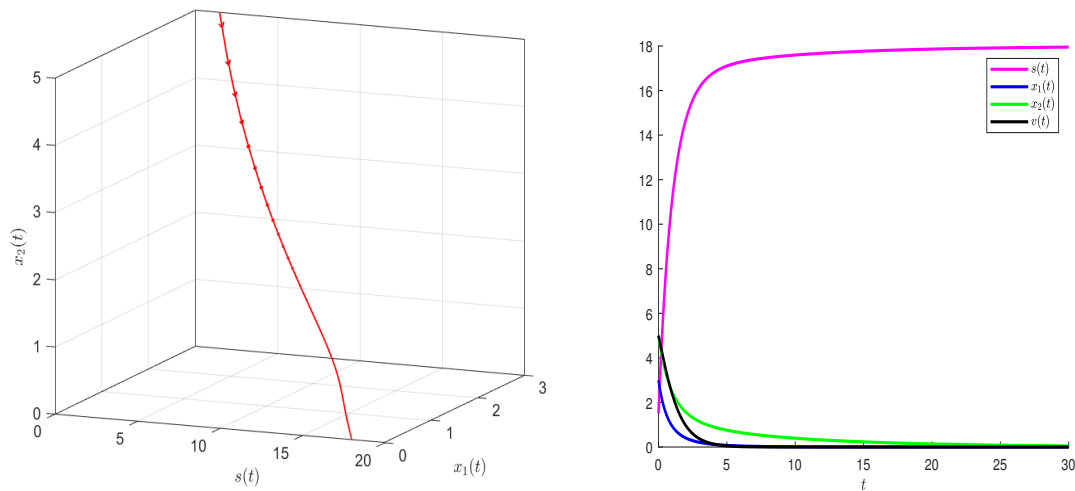
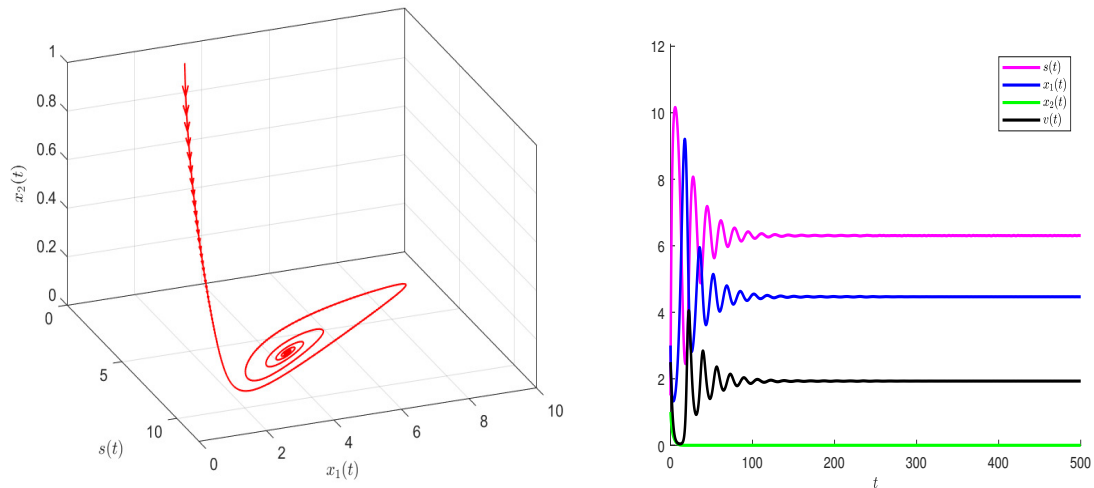**Figure 2.** Behaviour for $D = 1$, $s^{in} = 18$.

In Figure 3, if $D = 0.92$ which satisfies $\max(D_4 \approx 0.1, D_5 \approx 0.14, D_6 \approx 0.84, D_7 \approx 0.89) < D_2 \approx 0.892 < D = 0.92 < D_3 \approx 0.942 < D_1 \approx 0.943$, the solution with initial condition $(1.5, 3, 1, 2.5)$ converges to the equilibrium F_1. This confirms the point C(ii)-1 of Theorem 1. Only species 1 persists and the competitive exclusion principle is fulfilled.
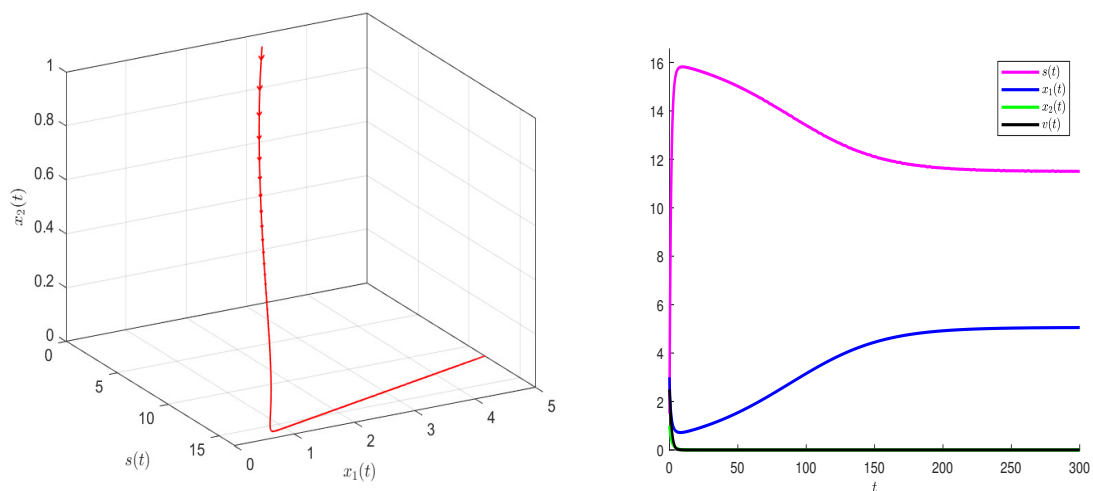


Figure 3. Behaviour for $D = 0.92$, $s^{in} = 16.56$.

In Figure 4, if $D = 0.67$ which satisfies $\max(D_4 \approx 0.09, D_5 \approx 0.05) < D = 0.67 < \min(D_3 \approx$

$0.92, D_6 \approx 0.79)$, the solution with initial condition $(1.5, 3, 1, 2.5)$ converges to the equilibrium F_3. This confirms the point B(i) of Theorem 1. The competitive exclusion principle is fulfilled here since that at least one species goes extinct.

Figure 4. Behaviour for $D = 0.67$, $s^{in} = 12.06$ and an initial condition $(1.5, 3, 1, 2.5)$.

In Figure 5, we use the same values as in Figure 4 but with different initial condition $(1.5, 3, 5, 5)$ then the solution converges to the equilibrium F_2. Again, this confirms the point B(i) of Theorem 1.
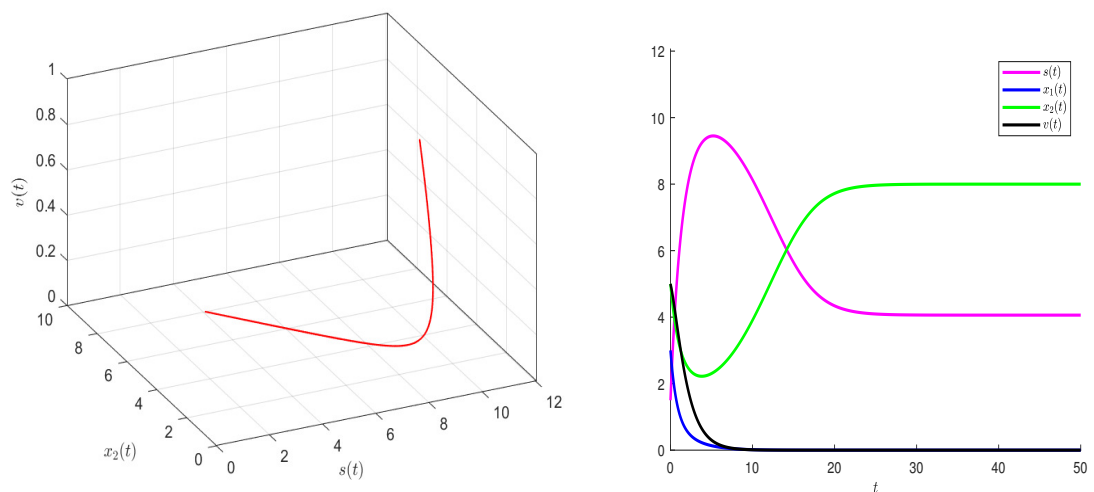

Figure 5. Behaviour for $D = 0.67$, $s^{in} = 12.06$ and an initial condition $(1.5, 3, 5, 5)$.

In the case where we have two equilibrium points which are locally stable (Figure 6), the initial concentrations of species have great importance in determination of which species is the winner. If the initial concentration is inside the attraction domain of the equilibrium point corresponding to the persistence of species 1, then species 2 goes extinct and if the initial concentration is inside the attraction domain of the equilibrium point corresponding to the persistence of species 2, then species 1 goes

extinct.
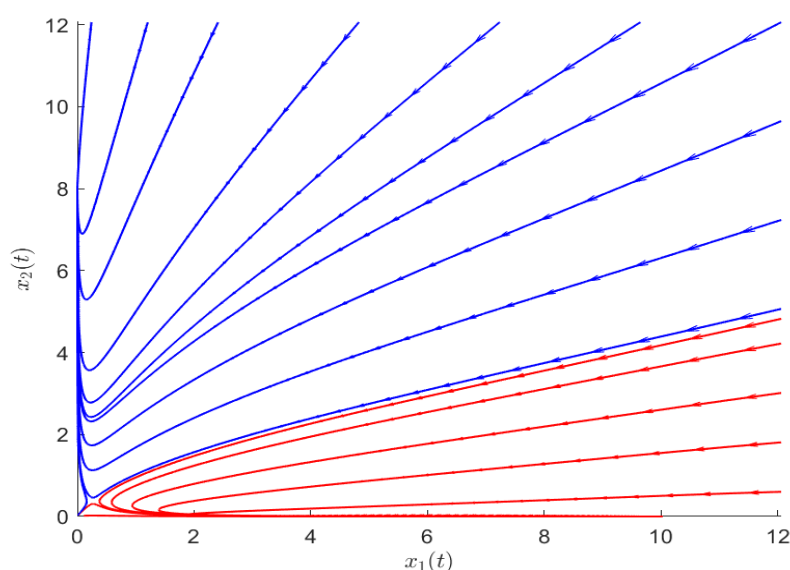
Figure 6. Behaviour in the (x_1, x_2)-plane for $D = 0.67$, $s^{in} = 12.06$. The trajectories filling the whole blue domain are converging to the equilibrium F_2 and the trajectories filling the whole red domain are converging to the equilibrium F_3

5. Conclusion

The competitive exclusion principle (CEP) has been widely studied in the scientific literature not only from a biological point of view but also from a mathematical modeling point of view. Some experiments were realized by Gause in 1932 on the growth of yeasts and paramecia [19]. It is deduced that the most competitive species consistently wins the competition. In 1960, this principle became quite popular in ecology: in fact, the CEP still valid for many kinds of ecosystems [4]. Hsu et al. [5] are among the first, in 1977, to study the problem of competition in the chemostat. They consider n populations in competition for the same nutrient, and show that the competitive exclusion is verified: that of the competitors who uses the better the substrate in small quantity survives, the others are extinguished. In this paper, we proposed a mathematical model (2.1) describing a reversible inhibition relationship between two competing bacteria for one resource in presence of a virus associated to the first species. We locally analysed the system (2.1). We proved that in a continuous reactor and under nonlinear general functional responses f_1 and f_2, the competitive exclusion principle is still fulfilled, that at least one species goes extinct. In the situation where we have two equilibrium points which are locally stable, initial species concentrations are important in determining which is the winning species.

Conflict of interest

The authors declare no conflict of interest.

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Proofs of lemmas

The Jacobian matrix J of system (2.1) on a point (s, x_1, x_2, v) is given by :

$$
J = \begin{bmatrix}
-D - x_1 \dfrac{\partial f_1}{\partial s} - x_2 \dfrac{\partial f_2}{\partial s} & -f_1 - x_2 \dfrac{\partial f_2}{\partial x_1} & -f_2 - x_1 \dfrac{\partial f_1}{\partial x_2} & 0 \\[2mm]
x_1 \dfrac{\partial f_1}{\partial s} & f_1 - D - \alpha v & x_1 \dfrac{\partial f_1}{\partial x_2} & -\alpha x_1 \\[2mm]
x_2 \dfrac{\partial f_2}{\partial s} & x_2 \dfrac{\partial f_2}{\partial x_1} & f_2 - D & 0 \\[2mm]
0 & \kappa \alpha v & 0 & \kappa \alpha x_1 - D
\end{bmatrix}
\tag{.1}
$$

Proof of lemma 1

The Jacobian matrix J_0 of system (2.1) on F_0 is then given by :

$$
J_0 = \begin{bmatrix}
-D & 0 & 0 & 0 \\[2mm]
0 & D_1 - D & 0 & 0 \\[2mm]
0 & 0 & D_2 - D & 0 \\[2mm]
0 & 0 & 0 & -D
\end{bmatrix}.
$$

Their eigenvalues are given by $\lambda_1 = D_1 - D, \lambda_2 = D_2 - D$ and $\lambda_3 = \lambda_4 = -D < 0$. Therfore, if $D < \max(D_1, D_2)$ then F_0 is a saddle point and if $D > \max(D_1, D_2)$ then F_0 is a stable node.

Proof of lemma 2

An equilibrium F_1 exists if and only if $\bar{x}_1 \in]0, s^{in}[$ is a solution of

$$
f_1(s^{in} - \bar{x}_1, 0) = D.
\tag{.2}
$$

Let $\psi_1(x_1) = f_1(s^{in} - x_1, 0) - D$. Since $\psi_1'(x_1) = -\dfrac{\partial f_1}{\partial s}(s^{in} - x_1, 0) < 0, \psi_1(0) = D_1 - D, \psi_1(s^{in}) = -D < 0$, equation (.2) admits a positive solution if and only if $D < D_1$. If this condition is satisfied then (.2) admits a unique solution since the function $\psi_1(.)$ is decreasing.

Assume that F_1 exists. One has

- If $D < D_3$ then $f_1(s^{in} - \bar{x}_1, 0) = D < D_3 = f_1(s^{in} - \dfrac{D}{\kappa \alpha}, 0)$ then $\bar{x}_1 > \dfrac{D}{\kappa \alpha}$.

- If $D > D_3$ then $f_1(s^{in} - \bar{x}_1, 0) = D > D_3 = f_1(s^{in} - \dfrac{D}{\kappa\alpha}, 0)$ then $\bar{x}_1 < \dfrac{D}{\kappa\alpha}$.

The Jacobian matrix J_1 of system (2.1) at F_1 is given by :

$$J_1 = \begin{bmatrix} -D - \bar{x}_1 \dfrac{\partial f_1}{\partial s} & -D & -f_2 - \bar{x}_1 \dfrac{\partial f_1}{\partial x_2} & 0 \\ \bar{x}_1 \dfrac{\partial f_1}{\partial s} & 0 & \bar{x}_1 \dfrac{\partial f_1}{\partial x_2} & -\alpha \bar{x}_1 \\ 0 & 0 & f_2 - D & 0 \\ 0 & 0 & 0 & \kappa\alpha \bar{x}_1 - D \end{bmatrix} = \begin{bmatrix} -D - \bar{x}_1 \dfrac{\partial f_1}{\partial s} & -D & -f_2 - \bar{x}_1 \dfrac{\partial f_1}{\partial x_2} & 0 \\ \bar{x}_1 \dfrac{\partial f_1}{\partial s} & 0 & \bar{x}_1 \dfrac{\partial f_1}{\partial x_2} & -\alpha \bar{x}_1 \\ 0 & 0 & D_5 - D & 0 \\ 0 & 0 & 0 & \kappa\alpha \bar{x}_1 - D \end{bmatrix}.$$

J_1 admits four eigenvalues given by $\lambda_1 = -(D - D_5)$ and $\lambda_2 = \kappa\alpha\left(\bar{x}_1 - \dfrac{D}{\kappa\alpha}\right)$. The other two eigenvalues are nonpositive and solution of the polynomial

$$\lambda^2 + a\lambda + b = 0$$

where $a = D + \bar{x}_1 \dfrac{\partial f_1}{\partial s} > 0$ and $b = D\bar{x}_1 \dfrac{\partial f_1}{\partial s} > 0$. It follows that

- F_1 is a saddle point if $D < D_3$.

- F_1 is a stable node if $D > D_3$ and $D > D_5$.

- F_1 is a saddle point if $D > D_3$ and $D < D_5$.

Proof of lemma 3

An equilibrium F_2 exists if and only if $\bar{x}_2 \in]0, s^{in}[$ is a solution of

$$f_2(s^{in} - \bar{x}_2, 0) = D. \tag{.3}$$

Let $\psi_2(x_2) = f_2(s^{in} - x_2, 0) - D$. Since $\psi_2'(x_2) = -\dfrac{\partial f_2}{\partial s}(s^{in} - \bar{x}_2, 0) < 0$, $\psi_2(0) = D_2 - D$, $\psi_2(s^{in}) = -D < 0$, equation (.3) admits a positive solution if and only if $D < D_2$. If this condition is satisfied then (.3) admits a unique solution since the function $\psi_2(.)$ is decreasing.

Assume that F_2 exists ($D < D_2$). The Jacobian matrix J_2 of system (2.1) at F_2 is given by :

$$J_2 = \begin{bmatrix} -D - \bar{x}_2 \dfrac{\partial f_2}{\partial s} & -f_1 - \bar{x}_2 \dfrac{\partial f_2}{\partial x_1} & -D & 0 \\ 0 & f_1 - D & 0 & 0 \\ \bar{x}_2 \dfrac{\partial f_2}{\partial s} & \bar{x}_2 \dfrac{\partial f_2}{\partial x_1} & 0 & 0 \\ 0 & 0 & 0 & -D \end{bmatrix} = \begin{bmatrix} -D - \bar{x}_2 \dfrac{\partial f_2}{\partial s} & -f_1 - \bar{x}_2 \dfrac{\partial f_2}{\partial x_1} & -D & 0 \\ 0 & D_4 - D & 0 & 0 \\ \bar{x}_2 \dfrac{\partial f_2}{\partial s} & \bar{x}_2 \dfrac{\partial f_2}{\partial x_1} & 0 & 0 \\ 0 & 0 & 0 & -D \end{bmatrix}.$$

J_2 admits four eigenvalues given by $\lambda_1 = -(D - D_4)$ and $\lambda_2 = -D < 0$. The other two eigenvalues are nonpositive and solution of the polynomial

$$\lambda^2 + a\lambda + b = 0$$

where $a = D + \bar{x}_2 \dfrac{\partial f_2}{\partial s} > 0$ and $b = D\bar{x}_2 \dfrac{\partial f_2}{\partial s} > 0$. It follows that

- If $D > D_4$ then F_2 is a stable node.

- If $D < D_4$ then F_2 is a saddle point.

Proof of lemma 4

An equilibrium F_3 exists if and only if $\bar{v} \in]0, s^{in} - \dfrac{D}{\kappa\alpha}[$ is a solution of

$$f_1(s^{in} - \frac{D}{\kappa\alpha} - \bar{v}, 0) = D + \alpha\bar{v}. \tag{.4}$$

Let $\psi_3(v) = f_1(s^{in} - \dfrac{D}{\kappa\alpha} - v, 0) - D - \alpha v$. Since $\psi_3'(v) = -\dfrac{\partial f_1}{\partial s}(s^{in} - \dfrac{D}{\kappa\alpha} - v, 0) - \alpha < 0$, $\psi_3(0) = D_3 - D$ and $\psi_3(s^{in} - \dfrac{D}{\kappa\alpha}) = -D - \alpha(s^{in} - \dfrac{D}{\kappa\alpha}) < 0$, equation (.4) admits a positive solution if and only if $D < D_3$. If this condition is satisfied then (.4) admits a unique solution since the function $\psi_3(.)$ is decreasing.

If F_3 exists, the Jacobian matrix J_3 of system (2.1) at F_3 is given by :

$$J_3 = \begin{bmatrix} -D - \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial s} & -D - \alpha\bar{v} & -f_2 - \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial x_2} & 0 \\ \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial s} & 0 & \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial x_2} & -\dfrac{D}{\kappa} \\ 0 & 0 & f_2 - D & 0 \\ 0 & \kappa\alpha\bar{v} & 0 & 0 \end{bmatrix} = \begin{bmatrix} -D - \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial s} & -D - \alpha\bar{v} & -f_2 - \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial x_2} & 0 \\ \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial s} & 0 & \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial x_2} & -\dfrac{D}{\kappa} \\ 0 & 0 & D_5 - D & 0 \\ 0 & \kappa\alpha\bar{v} & 0 & 0 \end{bmatrix}.$$

J_3 admits three eigenvalues given by $\lambda_1 = -(D - D_5)$ and three others eigenvalues associated to the following matrix

$$J_3^s = \begin{bmatrix} -D - \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial s} & -D - \alpha\bar{v} & 0 \\ \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial s} & 0 & -\dfrac{D}{\kappa} \\ 0 & \kappa\alpha\bar{v} & 0 \end{bmatrix}.$$

These eigenvalues are solutions of

$$-(D + \lambda)(\lambda^2 + a\lambda + b) = 0$$

where $a = \dfrac{D}{\kappa\alpha}\dfrac{\partial f_1}{\partial s} > 0$ and $b = D\alpha\bar{v}(1 + \dfrac{1}{\kappa\alpha}\dfrac{\partial f_1}{\partial s}) > 0$. It follows that

- If $D_5 < D < D_3$, then F_3 is then locally asymptotically stable.

- If $D < \min(D_3, D_5)$, then F_3 is a saddle point.

Proof of lemma 5

Assume that $0 < D < \min(D_4, D_5)$. From Lemmas 2 and 3, F_1 and F_2 exist.

1. If $\bar{x}_1 \geq \bar{x}_2$ then $D = f_2(s^{in} - \bar{x}_2, 0) \geq f_2(s^{in} - \bar{x}_1, 0) > f_2(s^{in} - \bar{x}_1, \bar{x}_1) = D_5 > D$ which is impossible.

2. If $\bar{x}_1 \leq \bar{x}_2$ then $D = f_1(s^{in} - \bar{x}_1, 0) \geq f_1(s^{in} - \bar{x}_2, 0) > f_1(s^{in} - \bar{x}_2, \bar{x}_2) = D_4 > D$ which is impossible.

Proof of lemma 6

Assume that F_4 exists. One has

$$\psi_1(\bar{\bar{x}}_1) = 0 = f_1(s^{in} - \bar{\bar{x}}_1, 0) - D > f_1(s^{in} - \bar{x}_1 - \bar{x}_2, \bar{x}_2) - D = 0 = \psi_1(\bar{x}_1)$$

then $\psi_1(\bar{\bar{x}}_1) > \psi_1(\bar{x}_1)$ since the function $\psi_1(.)$ is decreasing, $\bar{x}_1 > \bar{\bar{x}}_1$.

$$\psi_2(\bar{\bar{x}}_2) = f_2(s^{in} - \bar{\bar{x}}_2, 0) - D > f_2(s^{in} - \bar{x}_1 - \bar{x}_2, \bar{x}_1) - D = 0 = \psi_2(\bar{x}_2)$$

then $\psi_2(\bar{x}_2) < \psi_2(\bar{\bar{x}}_2)$ since the function $\psi_2(.)$ is decreasing, $\bar{x}_2 > \bar{\bar{x}}_2$.

Since the functions $x_2 \to f_1(s^{in} - x_1 - x_2, x_2)$ and $x_2 \to f_2(s^{in} - x_1 - x_2, x_1)$ are decreasing, one deduces immediatly that the isoclines are the graphs of two functions $x_2 = \varphi_1(x_1)$ and $x_2 = \varphi_2(x_1)$ and then $0 = \varphi_1(\bar{x}_1)$ and $\bar{x}_2 = \varphi_2(0)$. $\bar{x}_1$ is solution of $\psi_4(\bar{x}_1) = 0$ where $\psi_4(x_1) = \varphi_2(x_1) - \varphi_1(x_1)$. The derivatives of φ_1 and φ_2 are given by $\varphi_2'(x_1) = -1 + \frac{\partial f_2}{\partial x_1} / \frac{\partial f_2}{\partial s} < -1 < \varphi_1'(x_1) = -1 + \frac{\partial f_1}{\partial x_2} / (\frac{\partial f_1}{\partial x_2} - \frac{\partial f_1}{\partial s}) < 0$. one deduces that $\psi_4'(x_1) = \varphi_2'(x_1) - \varphi_1'(x_1) < 0$. $\psi_4(0) = \varphi_2(0) - \varphi_1(0) = \bar{x}_2 - \varphi_1(0)$ and $\psi_4(\bar{x}_1) = \varphi_2(\bar{x}_1)$ then $\bar{x}_1$ exists and is unique if and only if $\bar{x}_2 > \varphi_1(0)$ and $\varphi_2(\bar{x}_1) < 0$ and this is satisfied only if $D = f_1(s^{in} - \varphi_1(0), \varphi_1(0)) > f_1(s^{in} - \bar{x}_2, \bar{x}_2) = D_4$ and $D = f_2(s^{in} - \bar{x}_1 - \varphi_2(\bar{x}_1), \bar{x}_1) > f_2(s^{in} - \bar{x}_1, \bar{x}_1) = D_5$. The existence and the uniqueness of $\bar{x}_2 = \varphi_1(\bar{x}_1) = \varphi_2(\bar{x}_1)$ is easily deduced since the two function $\varphi_1(.)$ and $\varphi_2(.)$ are increasing.

Assume that F_4 exists. The Jacobian matrix J_4 of system (2.1) at $F_4 = (s^{in} - \bar{x}_1 - \bar{x}_2, \bar{x}_1, \bar{x}_2, 0)$ is given by :

$$J_4 = \begin{bmatrix} -D - \bar{x}_1 \frac{\partial f_1}{\partial s} - \bar{x}_2 \frac{\partial f_2}{\partial s} & -D - \bar{x}_2 \frac{\partial f_2}{\partial x_1} & -D - \bar{x}_1 \frac{\partial f_1}{\partial x_2} & 0 \\ \bar{x}_1 \frac{\partial f_1}{\partial s} & 0 & \bar{x}_1 \frac{\partial f_1}{\partial x_2} & -\alpha \bar{x}_1 \\ \bar{x}_2 \frac{\partial f_2}{\partial s} & \bar{x}_2 \frac{\partial f_2}{\partial x_1} & 0 & 0 \\ 0 & 0 & 0 & \kappa\alpha\bar{x}_1 - D \end{bmatrix}.$$

J_5 admits four eigenvalues given by $\lambda_1 = \kappa\alpha(\bar{x}_1 - \frac{D}{\kappa\alpha})$ and three other eigenvalues associated to the following matrix

$$J_4^s = \begin{bmatrix} -D - \bar{x}_1 \frac{\partial f_1}{\partial s} - \bar{x}_2 \frac{\partial f_2}{\partial s} & -D - \bar{x}_2 \frac{\partial f_2}{\partial x_1} & -D - \bar{x}_1 \frac{\partial f_1}{\partial x_2} \\ \bar{x}_1 \frac{\partial f_1}{\partial s} & 0 & \bar{x}_1 \frac{\partial f_1}{\partial x_2} \\ \bar{x}_2 \frac{\partial f_2}{\partial s} & \bar{x}_2 \frac{\partial f_2}{\partial x_1} & 0 \end{bmatrix}.$$

These eigenvalues are solutions of

$$-(\lambda + D)(\lambda^2 + a\lambda + b) = 0$$

where $a = \bar{x}_1 \frac{\partial f_1}{\partial s} + \bar{x}_2 \frac{\partial f_2}{\partial s} > 0$ and $b = \bar{x}_1 \bar{x}_2 \left[-\frac{\partial f_1}{\partial x_2} \frac{\partial f_2}{\partial x_1} + \frac{\partial f_1}{\partial x_2} \frac{\partial f_2}{\partial s} + \frac{\partial f_1}{\partial s} \frac{\partial f_2}{\partial x_1} \right] < 0$. It follows that F_4 is a saddle point.

Proof of lemma 7

Since the functions $x_2 \to f_1(s^{in} - x_2 - \frac{D}{\kappa\alpha} - v, x_2) - \alpha v$ and $x_2 \to f_2(s^{in} - x_2 - \frac{D}{\kappa\alpha} - v, \frac{D}{\kappa\alpha})$ are decreasing, one deduces immediatly that the isoclines are the graphs of two functions $x_2 = \varphi_3(v)$ and $x_2 = \varphi_4(v)$ and then $0 = \varphi_3(\bar{v})$. v^* is solution of $\psi_5(v^*) = 0$ where $\psi_5(v) = \varphi_4(v) - \varphi_3(v)$. The derivatives of φ_3 and φ_4 are given by $\varphi_4'(v) = -1 < \varphi_3'(v) = -1 + \left(\frac{\partial f_1}{\partial x_2} + \alpha\right)/(\frac{\partial f_1}{\partial x_2} - \frac{\partial f_1}{\partial s}) < 0$. One deduces that $\psi_5'(v) = \varphi_4'(v) - \varphi_3'(v) < 0$. $\psi_5(0) = \varphi_4(0) - \varphi_3(0)$ and $\psi_5(\bar{v}) = \varphi_4(\bar{v})$ then v^* exists and is unique if and only if $\varphi_3(0) < \varphi_4(0)$ and $\varphi_4(\bar{v}) < 0$ and this is satisfied only if $D = f_2(s^{in} - \varphi_4(\bar{v}) - \bar{v} - \frac{D}{\kappa\alpha}, \frac{D}{\kappa\alpha}) > f_2(s^{in} - \bar{v} - \frac{D}{\kappa\alpha}, \frac{D}{\kappa\alpha}) = D_6$.

Now, since $D = f_2(s^{in} - \varphi_4(0) - \frac{D}{\kappa\alpha}, \frac{D}{\kappa\alpha}) > f_2(s^{in} - \bar{v} - \frac{D}{\kappa\alpha}, \frac{D}{\kappa\alpha}) = D_6$ then $\varphi_4(0) < \bar{v}$ and hence $\varphi_3(0) < \varphi_4(0) < \bar{v}$. Then $D = f_1(s^{in} - \varphi_3(0) - \frac{D}{\kappa\alpha}, \varphi_3(0)) > f_1(s^{in} - \bar{v} - \frac{D}{\kappa\alpha}, \bar{v}) = D_7$.

Assume that F^* exists. One has

$$\psi_2(x_2^*) = f_2(s^{in} - x_2^*, 0) - D \geq f_2(s^{in} - x_2^* - \frac{D}{\kappa\alpha} - v^*, \frac{D}{\kappa\alpha}) - D - \alpha v^* = 0 = \psi_4(\bar{x}_2)$$

then $\psi_2(\bar{x}_2) < \psi_2(x_2^*)$ since the function $\psi_2(.)$ is decreasing, $\bar{x}_2 > x_2^*$.

The characteristic polynomial of the Jacobian matrix of system (2.1) at $F^* = (s^{in} - \frac{D}{\kappa\alpha} - x_2^* - v^*, \frac{D}{\kappa\alpha}, x_2^*, v^*)$ is given by :

$$P^* = \begin{vmatrix} -X - D - \frac{D}{\kappa\alpha}\frac{\partial f_1}{\partial s} - x_2^*\frac{\partial f_2}{\partial s} & -D - \alpha v^* - x_2^*\frac{\partial f_2}{\partial x_1} & -D - \frac{D}{\kappa\alpha}\frac{\partial f_1}{\partial x_2} & 0 \\ \frac{D}{\kappa\alpha}\frac{\partial f_1}{\partial s} & -X & \frac{D}{\kappa\alpha}\frac{\partial f_1}{\partial x_2} & -\frac{D}{\kappa} \\ x_2^*\frac{\partial f_2}{\partial s} & x_2^*\frac{\partial f_2}{\partial x_1} & -X & 0 \\ 0 & \kappa\alpha v^* & 0 & -X \end{vmatrix} = \begin{vmatrix} -(X+D) & -(X+D) & -(X+D) & -\frac{(X+D)}{\kappa} \\ \frac{D}{\kappa\alpha}\frac{\partial f_1}{\partial s} & -X & \frac{D}{\kappa\alpha}\frac{\partial f_1}{\partial x_2} & -\frac{D}{\kappa} \\ x_2^*\frac{\partial f_2}{\partial s} & x_2^*\frac{\partial f_2}{\partial x_1} & -X & 0 \\ 0 & \kappa\alpha v^* & 0 & -X \end{vmatrix}.$$

By developing, we obtain

$$P^*(X) = (X+D)\left(X^3 - \frac{D}{\kappa\alpha}x_2^*\frac{\partial f_1}{\partial x_2}\frac{\partial f_2}{\partial x_1}X + D\alpha v^* X + \frac{D}{\kappa\alpha}\frac{\partial f_1}{\partial s}X^2 + \frac{D}{\kappa\alpha}x_2^*\frac{\partial f_1}{\partial x_2}\frac{\partial f_2}{\partial s}X\right)$$
$$+ (X+D)\left[X\left(\frac{D}{\kappa\alpha}x_2^*\frac{\partial f_1}{\partial s}\frac{\partial f_2}{\partial x_1} + x_2^*\frac{\partial f_2}{\partial s}X\right) + D\alpha v^* x_2^*\frac{\partial f_2}{\partial s} + v^*\left(\frac{D}{\kappa}\frac{\partial f_1}{\partial s}X + \frac{D}{\kappa}x_2^*\frac{\partial f_1}{\partial x_2}\frac{\partial f_2}{\partial s}\right)\right].$$

Then

$$P^*(X) = (X+D)(X^3 + b_2 X^2 + b_1 X + b_0)$$

with

$$b_2 = \frac{D}{\kappa\alpha}\frac{\partial f_1}{\partial s} + x_2^*\frac{\partial f_2}{\partial s} > 0,$$
$$b_1 = -\frac{D}{\kappa\alpha}x_2^*\frac{\partial f_1}{\partial x_2}\frac{\partial f_2}{\partial x_1} + D\alpha v^* + \frac{D}{\kappa\alpha}x_2^*\frac{\partial f_1}{\partial x_2}\frac{\partial f_2}{\partial s} + \frac{D}{\kappa\alpha}x_2^*\frac{\partial f_1}{\partial s}\frac{\partial f_2}{\partial x_1} + \frac{D}{\kappa}v^*\frac{\partial f_1}{\partial s},$$
$$b_0 = D\alpha v^* x_2^*\frac{\partial f_2}{\partial s} + \frac{D}{\kappa}v^* x_2^*\frac{\partial f_1}{\partial x_2}\frac{\partial f_2}{\partial s} = \frac{D}{\kappa}v^* x_2^*\left(\kappa\alpha + \frac{\partial f_1}{\partial x_2}\right)\frac{\partial f_2}{\partial s} < 0.$$

It follows that F^* is always a saddle point.