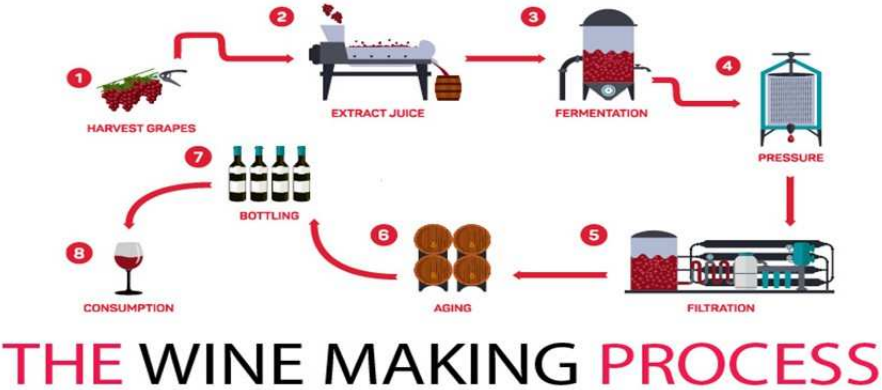
Studying the long time dynamics of fermentation models: production of dry and sweet wine

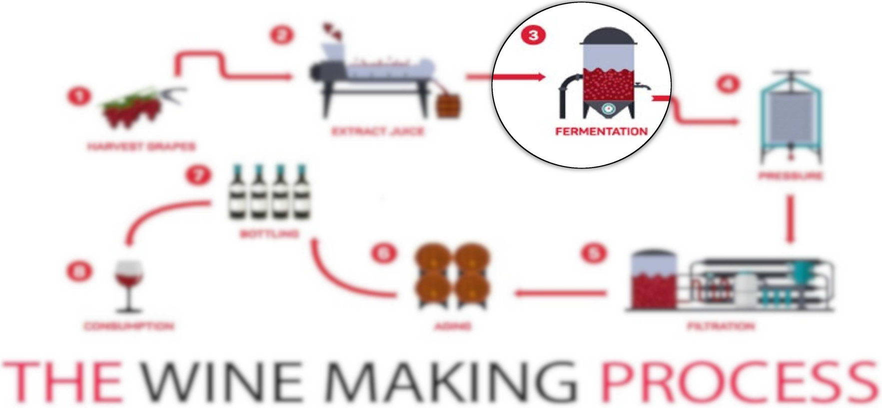
Javier López de la Cruz

in collaboration with Renato Colucci

ICMC Summer Meeting São Carlos

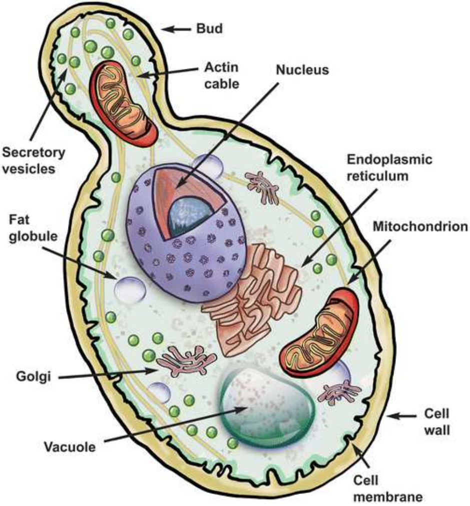
Table of Contents


1
HARVEST GRAPES
2
EXTRACT JUICE
3
FERMENTATION
4
PRESSURE
5
FILTRATION
6
AGING
7
BOTTLING
8
CONSUMPTION
THE WINE MAKING PROCESS

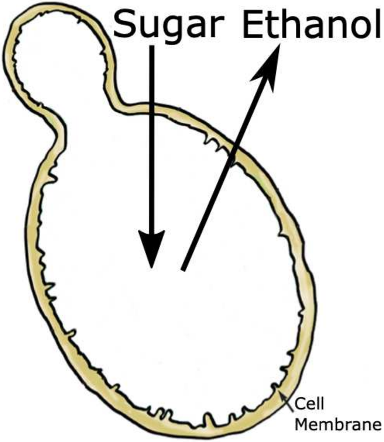

3
FERMENTATION
THE WINE MAKING PROCESS

Introduction

- **Fermentation**: bio-chemical process by means of which sugar in grape juice is transformed into ethanol in presence of nutrients.
- **Transformation**: thanks to yeast in the must which allows to degrade sugar into ethanol.
- **Different wines**: dry or sweet.
- **Bioreactor (batch)**: substrate provided at the beginning and no remove.



- **Fermentation**: bio-chemical process by means of which sugar in grape juice is transformed into ethanol in presence of nutrients.
- **Transformation**: thanks to yeast in the must which allows to degrade sugar into ethanol.
- **Different wines**: dry or sweet.
- **Bioreactor (batch)**: substrate provided at the beginning and no remove.



Introduction

- **Fermentation**: bio-chemical process by means of which sugar in grape juice is transformed into ethanol in presence of nutrients.
- **Transformation**: thanks to yeast in the must which allows to degrade sugar into ethanol.
- **Different wines**: dry or sweet.
- **Bioreactor (batch)**: substrate provided at the beginning and no remove.

Literature dedicated to derive mathematical models

S. Aiba, M. Shoda and M. Nagatani,
Kinetics of product inhibition in alcohol fermentation,
Biotechnology and Bioengineering, 10 (1968), 845–864.

R. Boulton,
The prediction of fermentation behavior by a kinetic model,
Am J Enol Vitic, 31 (1980), 40–45.

I. Caro, L. P'erez and D. Cantero,
Development of a kinetic model for the alcoholic fermentation of must,
Biotechnology and Bioengineering, 38 (1991), 742–748.

A. C. Cramer, S. Vlassides and D. E. Block,
Kinetic model for nitrogen-limited wine fermentations,
Biotechnology and Bioengineering, 77 (2002), 49–60.

Few works dedicated to study the mathematical models!

Table of Contents

Table of Contents

Mathematical model

$$
\begin{aligned}
\frac{dx}{dt} &= \mu(n)x, \\
\frac{dn}{dt} &= -\mu(n)x, \\
\frac{de}{dt} &= \beta(s)\gamma(e)x, \\
\frac{ds}{dt} &= -\beta(s)\gamma(e)x
\end{aligned}
$$

- $x = x(t)$: yeast conc.
- $n = n(t)$: nitrogen conc.
- $e = e(t)$: ethanol conc.
- $s = s(t)$: sugar conc.

Mathematical model

$$\begin{aligned}\frac{dx}{dt} &= \mu(n)x, \\ \frac{dn}{dt} &= -\mu(n)x, \\ \frac{de}{dt} &= \beta(s)\gamma(e)x, \\ \frac{ds}{dt} &= -\beta(s)\gamma(e)x\end{aligned}$$

Growth rate

$$\mu(n) = \frac{\mu_{\max} n}{k_n + n}$$

Rate of sugar utilization

$$\beta(s) = \frac{\beta_{\max} s}{k_s + s}$$

Inhibition of sugar consumption

$$\gamma(e) = \frac{k_e}{k_e + e}$$

- $x = x(t)$: yeast conc.
- $n = n(t)$: nitrogen conc.
- $e = e(t)$: ethanol conc.
- $s = s(t)$: sugar conc.

- $\mu_{\max}$, $\beta_{\max}$: max. specific growths
- k_n: nitrogen limited growth
- k_s: sugar transport across cell membrane
- k_e: ethanol inhibition

Mathematical model

$$\frac{dx}{dt} = \mu(n)x, \quad (1)$$

$$\frac{dn}{dt} = -\mu(n)x, \quad (2)$$

$$\frac{de}{dt} = \beta(s)\gamma(e)x, \quad (3)$$

$$\frac{ds}{dt} = -\beta(s)\gamma(e)x \quad (4)$$

Observe that

$$\frac{dx}{dt} + \frac{dn}{dt} = 0 \qquad \frac{de}{dt} + \frac{ds}{dt} = 0.$$

Then

$$x(t) + n(t) = x(0) + n(0) := \gamma > 0$$

$$e(t) + s(t) = e(0) + s(0) := \lambda > 0.$$

Thus, we can rewrite system (1)-(4) as a two dimensional one

$$\frac{dx}{dt} = \frac{\mu_{\max}(\gamma - x)}{k_n + \gamma - x}x, \quad (5)$$

$$\frac{de}{dt} = \frac{\beta_{\max}(\lambda - e)}{k_s + \lambda - e}\frac{k_e}{k_e + e}x. \quad (6)$$

Theorem 1

For any initial value $(x_0, e_0) \in [0, \gamma] \times [0, \lambda]$, *system* (5)-(6) *possesses a unique global solution which is, in addition, positive and bounded. Moreover, as long as* $(x_0, e_0) \in (0, \gamma] \times [0, \lambda]$, *the solutions of system* (5)-(6) *approach the fixed point* $P = (\gamma, \lambda)$ *as* t *goes to infinity. As a consequence, solutions of system* (1)-(4) *converge to* $(\gamma, 0, \lambda, 0)$.

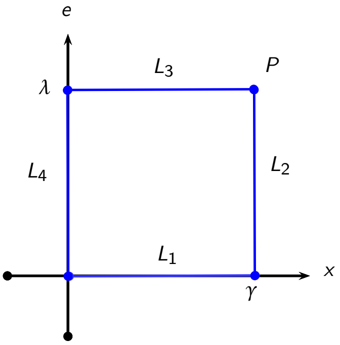
Proof. By classical theory of ODE's it is easy to obtain local existence and uniqueness of the solutions of system (5)-(6).

Moreover, the positive cone $\mathscr{X} = \{(x, e), x \geq 0, e \geq 0\}$ is positive invariant since $x = 0$ is an invariant plane and on $e = 0$ we have

$$\left.\frac{de}{dt}\right|_{e=0} = \frac{\beta_{\max}\lambda}{k_s + \lambda} x \geq 0.$$

Hence, we obtain the positiveness of solutions.

Dynamics of the model



- **Side** L_1**:** $e = 0$, $x \in (0, \gamma)$.

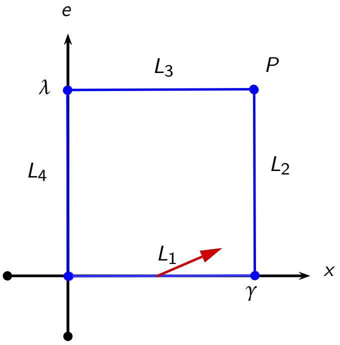
$$\frac{dx}{dt} > 0 \quad \text{and} \quad \frac{de}{dt} > 0.$$

$$\frac{dx}{dt} = \frac{\mu_{\max}(\gamma - x)}{k_n + \gamma - x} x,$$

$$\frac{de}{dt} = \frac{\beta_{\max}(\lambda - e)}{k_s + \lambda - e} \frac{k_e}{k_e + e} x.$$

- **Side** L_2**:** $x = \gamma$, $e \in [0, \lambda)$. Set $x = \gamma$ invariant and $\frac{de}{dt} > 0$ for $e \in [0, \lambda)$.
- **Side** L_3**:** $e = \lambda$, $x \in (0, \gamma)$. Set $e = \lambda$ is invariant while $\frac{dx}{dt} > 0$ for $x \in (0, \gamma)$.
- **Side** L_4**:** $x = 0$, $e \in [0, \lambda)$. This side consists of a segment of fixed points which are unstable.

Dynamics of the model



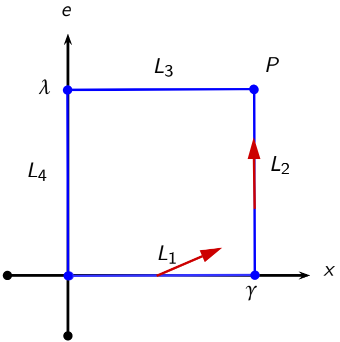
- **Side** L_1**:** $e = 0,\ x \in (0, \gamma)$.

$$\frac{dx}{dt} > 0 \quad \text{and} \quad \frac{de}{dt} > 0.$$

$$\begin{aligned} \frac{dx}{dt} &= \frac{\mu_{\max}(\gamma - x)}{k_n + \gamma - x} x, \\ \frac{de}{dt} &= \frac{\beta_{\max}(\lambda - e)}{k_s + \lambda - e} \frac{k_e}{k_e + e} x. \end{aligned}$$

- **Side** L_2**:** $x = \gamma,\ e \in [0, \lambda)$. Set $x = \gamma$ invariant and $\frac{de}{dt} > 0$ for $e \in [0, \lambda)$.
- **Side** L_3**:** $e = \lambda,\ x \in (0, \gamma)$. Set $e = \lambda$ is invariant while $\frac{dx}{dt} > 0$ for $x \in (0, \gamma)$.
- **Side** L_4**:** $x = 0,\ e \in [0, \lambda)$. This side consists of a segment of fixed points which are unstable.

Dynamics of the model



- **Side** L_1**:** $e = 0$, $x \in (0, \gamma)$.

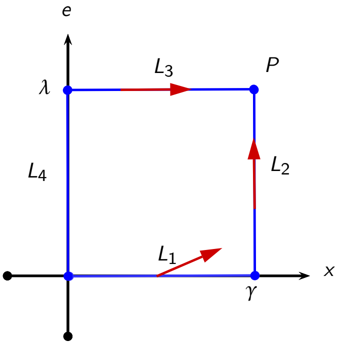
$$\frac{dx}{dt} > 0 \qquad \text{and} \qquad \frac{de}{dt} > 0.$$

$$\frac{dx}{dt} = \frac{\mu_{\max}(\gamma - x)}{k_n + \gamma - x} x,$$

$$\frac{de}{dt} = \frac{\beta_{\max}(\lambda - e)}{k_s + \lambda - e} \frac{k_e}{k_e + e} x.$$

- **Side** L_2**:** $x = \gamma$, $e \in [0, \lambda)$. Set $x = \gamma$ invariant and $\frac{de}{dt} > 0$ for $e \in [0, \lambda)$.
- **Side** L_3**:** $e = \lambda$, $x \in (0, \gamma)$. Set $e = \lambda$ is invariant while $\frac{dx}{dt} > 0$ for $x \in (0, \gamma)$.
- **Side** L_4**:** $x = 0$, $e \in [0, \lambda)$. This side consists of a segment of fixed points which are unstable.

Dynamics of the model



$$\frac{dx}{dt} = \frac{\mu_{\max}(\gamma - x)}{k_n + \gamma - x} x,$$

$$\frac{de}{dt} = \frac{\beta_{\max}(\lambda - e)}{k_s + \lambda - e} \frac{k_e}{k_e + e} x.$$

- **Side** L_1**:** $e = 0$, $x \in (0, \gamma)$.

$$\frac{dx}{dt} > 0 \quad \text{and} \quad \frac{de}{dt} > 0.$$

- **Side** L_2**:** $x = \gamma$, $e \in [0, \lambda)$. Set $x = \gamma$ invariant and $\frac{de}{dt} > 0$ for $e \in [0, \lambda)$.
- **Side** L_3**:** $e = \lambda$, $x \in (0, \gamma)$. Set $e = \lambda$ is invariant while $\frac{dx}{dt} > 0$ for $x \in (0, \gamma)$.
- **Side** L_4**:** $x = 0$, $e \in [0, \lambda)$. This side consists of a segment of fixed points which are unstable.

Dynamics of the model

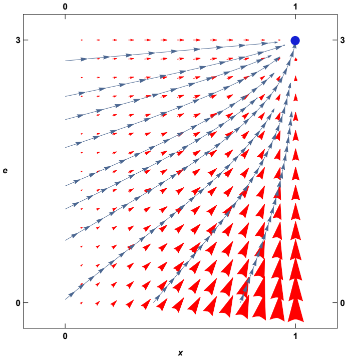


Figure: Vector field of system (5)-(6) with $\gamma = 1$ and $\lambda = 3$.

Dynamics of the model

Then, $B = [0,\gamma] \times [0,\lambda]$ is positively invariant. Moreover, solutions starting on B are positive, bounded and global in time.

Now, we study the asymptotic behavior of solutions starting on B.

- Since $\frac{dx}{dt} > 0$ for $x \in (0,\gamma)$, no periodic orbits in B.
- Then, invariant sets on B are the unstable fixed points on the side L_4 and the fixed point P.
- Stability of P: compute the eigenvalues of the Jacobian matrix

$$\lambda_1 = -\frac{\gamma}{k_n}, \qquad \lambda_2 = -\frac{\beta_{max}}{k_s(k_e + \lambda)},$$

- λ_1 and λ_2 are both negative, then P is locally stable.

Finally, every solution of system (5)-(6) with initial value in $(0,\gamma] \times [0,\lambda]$ converges to P . ■

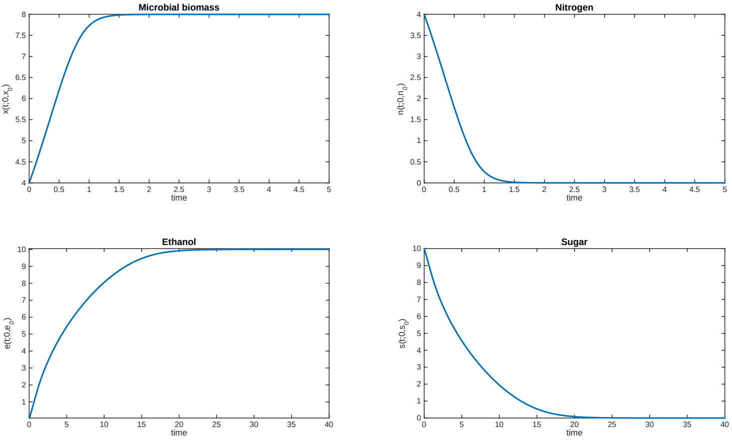
Dynamics of the model

Thanks to Theorem 1, since every solution of system (5)-(6) with initial value in $(0,\gamma] \times [0,\lambda]$ converges to $P = (\gamma, \lambda)$, every solution of system (1)-(4) converge to $(\gamma, 0, \lambda, 0)$.

Remark: Theorem 1 consistent with real fermentation process

- In this case, dry wine is obtained.
- Sum of sugar and ethanol concentrations $s(t) + e(t) = s(0) = \lambda$ remains constant.
- Total quantity of sugar transformed into ethanol since $e(t) \to s(0)$ while $s(t) \to 0$.
- Sum of microbial biomass and nitrogen concentrations $x + n$ remains constant with $n(t) \to 0$ and $x(t) \to x(0) + n(0)$.

Numerical simulations



$$x_0 = 4, n_0 = 4, e_0 = 0, s_0 = 10, \mu_{max} = 1.5, k_n = 2, \beta_{max} = 0.4, k_s = 1.2, k_e = 2.$$

Table of Contents

Mathematical model

$$\begin{aligned}
\frac{dx}{dt} &= x(\mu(n) - ke), \\
\frac{dn}{dt} &= -\mu(n)x, \\
\frac{de}{dt} &= \beta(s)\gamma(e)x, \\
\frac{ds}{dt} &= -\beta(s)\gamma(e)x
\end{aligned}$$

- $x = x(t)$: yeast conc.
- $n = n(t)$: nitrogen conc.
- $e = e(t)$: ethanol conc.
- $s = s(t)$: sugar conc.

Mathematical model

$$\frac{dx}{dt} = x(\mu(n) - ke),$$

$$\frac{dn}{dt} = -\mu(n)x,$$

$$\frac{de}{dt} = \beta(s)\gamma(e)x,$$

$$\frac{ds}{dt} = -\beta(s)\gamma(e)x$$

- $x = x(t)$: yeast conc.
- $n = n(t)$: nitrogen conc.
- $e = e(t)$: ethanol conc.
- $s = s(t)$: sugar conc.

Growth rate

$$\mu(n) = \frac{\mu_{\max} n}{k_n + n}$$

Rate of sugar utilization

$$\beta(s) = \frac{\beta_{\max} s}{k_s + s}$$

Inhibition of sugar consumption

$$\gamma(e) = \frac{k_e}{k_e + e}$$

- $\mu_{\max}$, $\beta_{\max}$: max. specific growths
- k_n: nitrogen limited growth
- k_s: sugar transport across cell membrane
- k_e: ethanol inhibition
- k: sensitivity of yeast to ethanol

Similarly to the dry wine case we can rewrite the previous system

$$\frac{dx}{dt} = x\left[\frac{\mu_{\max} n}{k_n + n} - ke\right], \tag{7}$$

$$\frac{dn}{dt} = -\frac{\mu_{\max} n}{k_n + n} x, \tag{8}$$

$$\frac{de}{dt} = \frac{\beta_{\max}(\lambda - e)}{k_s + \lambda - e} \frac{k_e}{k_e + e} x. \tag{9}$$

where $e(t) + s(t) = s(0) := \lambda > 0$.

From now on we will denote

$$\mathscr{X} = \{(x, n, e) \in \mathbb{R}^3 \, : \, x \geq 0, n \geq 0, e \geq 0\}$$

the positive cone.

Dynamics of the model

Theorem 2

All solutions of system (7)-(9) *with initial data in*

$$C := [0, +\infty) \times [0, +\infty) \times [0, \lambda),$$

are defined for all $t \in [0, +\infty)$. *Moreover, they are positive and bounded.*

Proof. By classical theory of ODE's we obtain local existence and uniqueness of solutions.

Observe that $x = 0$, $n = 0$ and $e = \lambda$ are invariant plane while on $e = 0$ the vector field points inside C. Then, we also have that $e(t)$ is globally defined and bounded.

Dynamics of the model

From

$$\frac{dn}{dt} = -\frac{\mu_{\max} n}{k_n + n} x$$

we have that $n(t)$ is decreasing, then $n(t)$ is bounded for any $n_0 \in C$ and defined for all $t \geq 0$.

Moreover, we have

$$\frac{dx}{dt} = x \left[\frac{\mu_{\max} n}{k_n + n} - ke \right] \leq \frac{\mu_{\max} n}{k_n + n} x \leq \frac{\mu_{\max} n(0)}{k_n + n(0)} x =: \rho x, \tag{10}$$

where we used that $\mu(n(t))$ is decreasing with respect to t.

Then,

$$x(t) \leq x(0) e^{\rho t}, \quad \text{for } t \geq 0,$$

whence $x(t)$ is defined for all $t \in [0, +\infty)$.

Dynamics of the model

It remains to prove the boundedness of $x(t)$. Suppose by contradiction

$$\lim_{t\to+\infty} x(t) = +\infty, \qquad \text{and} \qquad \lim_{t\to+\infty} n(t) = n^* > 0,$$

then

$$\frac{dn}{dt} = -\frac{\mu_{\max} n}{k_n + n} x \quad \Longrightarrow \quad \lim_{t\to+\infty} \frac{dn}{dt} = -\infty$$

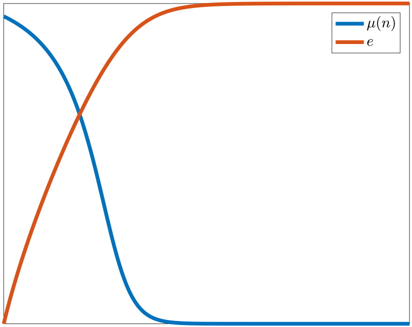
Now suppose that

$$\lim_{t\to+\infty} x(t) = +\infty, \qquad \text{and} \qquad \lim_{t\to+\infty} n(t) = 0. \tag{11}$$

We recall that

- the nitrogen concentration n is decreasing from $n(0) > 0$ to zero.
- μ is monotonic, then $\mu(n)$ goes to zero.
- the ethanol concentration e is increasing from zero.

Dynamics of the model



There exists $T > 0$ such that

$$\mu(n(T)) - ke(T) = 0.$$

Then

$$\frac{dx}{dt} = x\left[\frac{\mu_{\max} n}{k_n + n} - ke\right] \leq 0,$$

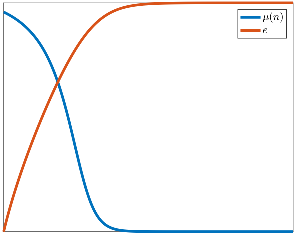
for all $t > T$.

This contradicts (11) and then the biomass concentration x is bounded and defined for all $t \geq 0$.

■

Corollary 3

For any $x_0 > 0$, there exists $T > 0$ such that the biomass concentration $x(t)$ is increasing for every $t \in [0, T]$. Then, it attains its maximum at $t = T$ and decreases for every $t > T$.



There exists $T > 0$ such that

$$\mu(n(T)) - ke(T) = 0.$$

Then

$$\frac{dx}{dt} = x\left[\frac{\mu_{\max} n}{k_n + n} - ke\right] \begin{cases} > 0, & t < T \\ = 0, & t = T \\ < 0, & t > T \end{cases}$$

Crucial effect of inhibition

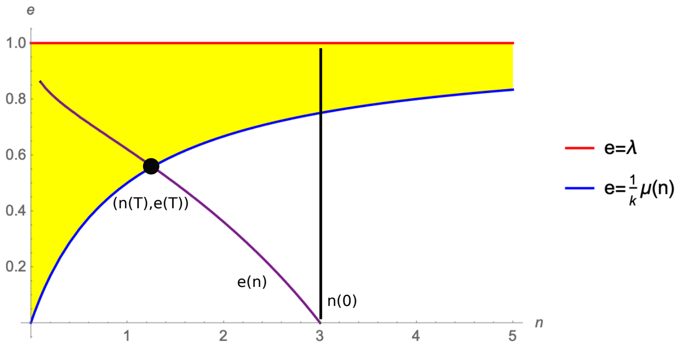
Dynamics of the model

Theorem 4

The set

$$A = \{(x,n,e) \in C : \lambda_1(n,e) := \mu(n) - ke < 0\}.$$

is positively invariant.



Theorem 5

Every solution of system (7)-(9) with initial value in $C \setminus \{x = 0\}$ converges to a fixed point in the plane $x = 0$.

Proof. Suppose that there exists a strictly positive constant $L > 0$ such that

$$\lim_{t \to +\infty} x(t) = L > 0,$$

then we have

$$\lim_{t \to \infty} \frac{dx}{dt} = \lim_{t \to \infty} x \left[\frac{\mu_{\max} n}{k_n + n} - ke \right] = L(\mu(n*) - ke^*), \tag{12}$$

where n^* and e^* denote the limit values of the nitrogen and ethanol concentrations. Such limits satisfy

$$0 \le n^* \le n(0), \qquad \text{and} \qquad 0 < e^* \le \lambda.$$

From Theorem 4, we have that the limit points are in

$$A = \{(x, n, e) \in C : \lambda_1(n, e) := \mu(n) - ke < 0\}.$$

and as a consequence

$$\lim_{t\to\infty} \frac{dx}{dt} = L(\mu(n*) - ke^*) < 0, \qquad (13)$$

Thus, we conclude that

$$\lim_{t\to+\infty} x(t) = 0. \qquad (14)$$

As a result, every solution of system (7)-(9) with initial value in $C \setminus \{x = 0\}$ converges to a fixed point in the plane $x = 0$.

■

Theorem 6

The nitrogen concentration is not completely consumed at the end of the process, i.e.,

$$\lim_{t\to+\infty} n(t) > 0.$$

Proof. Assume by contradiction that

$$\lim_{t\to+\infty} n(t) := n_\infty = 0.$$

Since the nitrogen concentration n remains positive and decreasing for $t \geq 0$, it is possible to define a diffeomorphism from $[0,+\infty)$ to $(n_\infty, n_0]$, where $n_0 = n(0)$. Then, the microbial biomass concentration x can be expressed as a function of n:

$$\frac{dx}{dn} = \frac{x(\mu(n)-ke)}{-\mu(n)x} = -1 + \frac{ke}{\mu(n)}.$$

Dynamics of the model

Hence, for $n < n(T)$ with $T > 0$, we have

$$\frac{dx}{dn} > -1 + \frac{ke(T)}{\mu(n)}. \tag{15}$$

We observe that $\mu(n) \leq \frac{\mu_{\max}}{k_n} n$.

Hence, we have

$$\frac{dx}{dn} > -1 + ke(T)\frac{k_n}{\mu_{\max}}\frac{1}{n} := -1 + \frac{\sigma}{n},$$

where $\sigma > 0$.

Finally, by integrating the last inequality between n_∞ and n we obtain

$$x(n) > x(n_\infty) - n + n_\infty + \sigma(\log n - \log n_\infty).$$

Then, if $n_\infty = 0$ we have that $x > \infty$.

Dynamics of the model

Theorem 7

Let $F : C \to \mathbb{R}$ be the function defined as

$$F(e,n) := -k_s(k_e+\lambda)\log\left(\frac{\lambda-e}{\lambda}\right) + (k_e-k_s)e + \frac{1}{2}e^2 + \nu k_n \log\left(\frac{n}{n_0}\right) + \nu(n-n_0),$$

where

$$\nu := \beta_{max}\frac{k_e}{\mu_{max}}.$$

Then, the ethanol and nitrogen concentrations satisfy $F(e,n) = 0$.

Theorem 8

The ethanol concentration $e(t)$ does not tend to the initial sugar concentration λ.

Proof. Suppose $e(t) \to \lambda$. Then $n \to 0$. ∎

Theorem 9

Suppose that $\rho = \frac{\mu_{max} n(0)}{k_n + n(0)} < k\lambda$. *Then the unique positive solution of the following equation provide an upper bound for the limit value of the ethanol concentration*

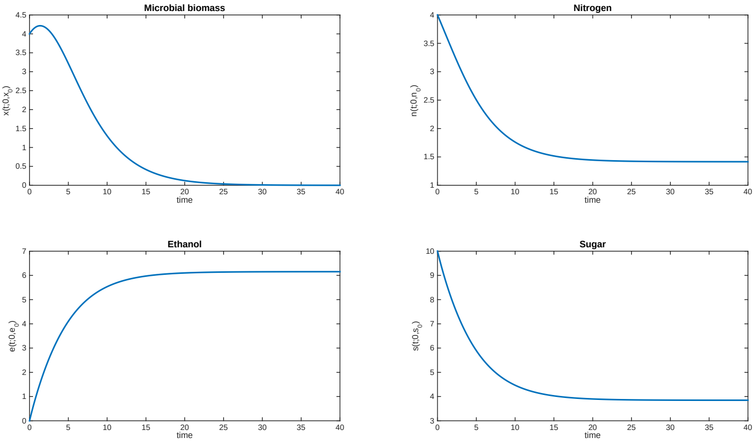
$$-\delta x(0) = \alpha_3 e^3 + \alpha_2 e^2 + \alpha_1 e + \alpha \log\left(\frac{\lambda - e}{\lambda}\right), \tag{16}$$

where

$$\delta = \frac{\beta_{max} k_e}{k}, \quad \alpha_3 = -\frac{1}{3}, \quad \alpha_2 = \frac{1}{2}\left(\frac{\rho}{k} - k_e + k_s\right),$$

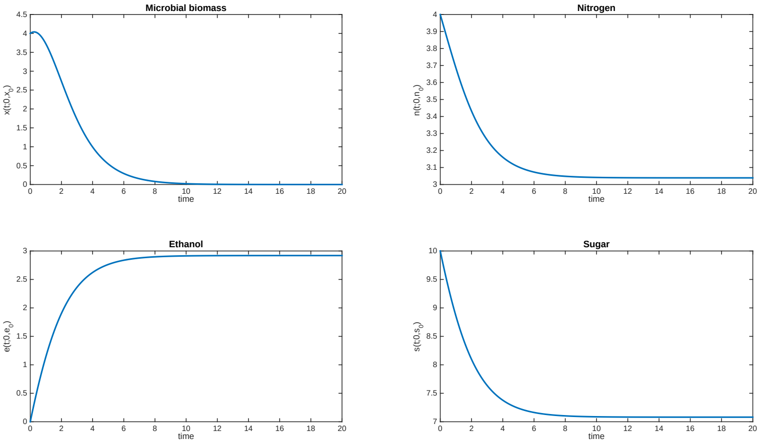
$$\alpha_1 = k_s\left(\lambda - \frac{\rho}{k}\right) + k_e\left(k_s + \frac{\rho}{k}\right), \quad \alpha = k_s\left(\lambda - \frac{\rho}{k}\right)(k_e + \lambda).$$

Remark: upper bound for limit value of ethanol!



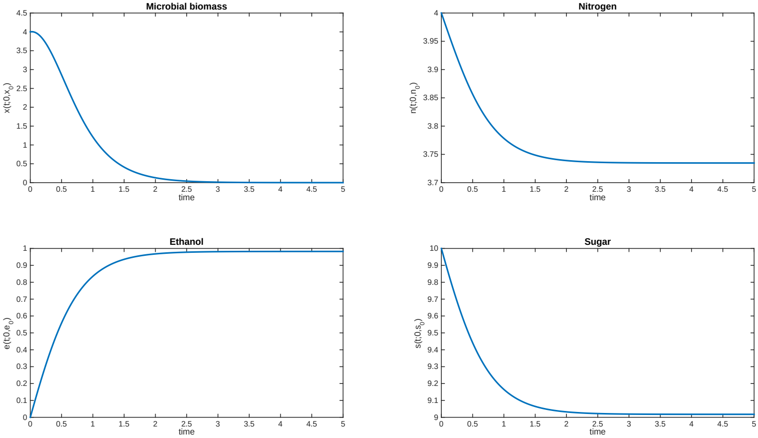
$$x_0 = 4, n_0 = 4, e_0 = 0, s_0 = 10, \mu_{\max} = 0.1, k_n = 1, \beta_{\max} = 0.4, k_s = 2, k_e = 4.$$

Numerical simulations. $k = 0.25$ $e^* = 3.19$ $e^* = 2.92$



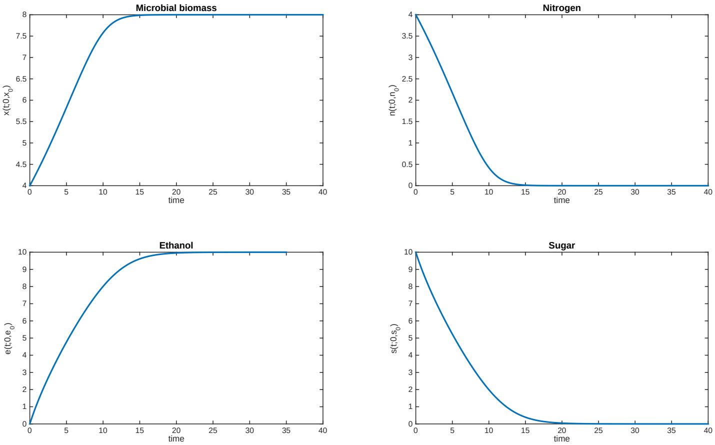
$$x_0 = 4, n_0 = 4, e_0 = 0, s_0 = 10, \mu_{max} = 0.1, k_n = 1, \beta_{max} = 0.4, k_s = 2, k_e = 4.$$

Numerical simulations. $k = 2.5$ $e^* = 1.07$ $e^* = 0.98$



$$x_0 = 4, n_0 = 4, e_0 = 0, s_0 = 10, \mu_{max} = 0.1, k_n = 1, \beta_{max} = 0.4, k_s = 2, k_e = 4.$$

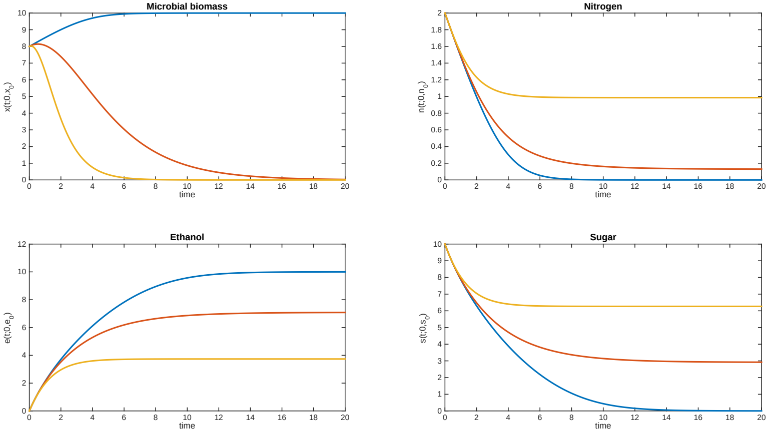
Numerical simulations. $k = 0$



$$x_0 = 4, n_0 = 4, e_0 = 0, s_0 = 10, \mu_{\max} = 0.1, k_n = 1, \beta_{\max} = 0.4, k_s = 2, k_e = 4.$$

Table of Contents

Numerical simulations. $k = 0.05$ and $k = 0.25$



$$x_0 = 8, n_0 = 2, e_0 = 0, s_0 = 10, \mu_{\max} = 0.1, k_n = 1, \beta_{\max} = 0.4, k_s = 2, k_e = 4.$$

Table of Contents

Conclusion

- Studied two models for wine production.
  - Dry wine: total quantity of sugar transformed into ethanol.
  - Sweet wine: parameter to interrupt.
- Proved existence, uniqueness, boundedness and positiveness of solution of both models.
- Studied in details the asymptotic behavior of state variables: yeast, nitrogen, sugar, ethanol.
- However... results in this work not only clarify the dynamics of the model.
- We provide useful tools to control the fermentation process and produce wine with the desired sugar.

Here you can see practical guide for producers ;)

Reference

Renato Colucci and Javier López-de-la-Cruz,
Dynamics of fermentation models to study the production of dry and sweet wine,
Communications on pure and applied analysis, vol. 19, 4 (2020) 2015–2034.

Happy birthday... and thank you very much for everything!