

MSc Computational Science
joint programme UvA/VU



A DIFFUSION-BASED MODEL OF SPATIAL INTERACTIONS IN *ASPERGILLUS* SPP. GERMINATION



FIRST EXPERIMENTS

Presented by Boyan Mihaylov

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Supervisor: Prof. dr. Han Wösten, Utrecht University

Examiner: Dr. Jaap Kaandorp, University of Amsterdam

OUTLINE



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GENERAL ASSUMPTIONS



Simulation space

» 2D lattice \equiv thin layer of medium clamped between 2 impermeable sheets

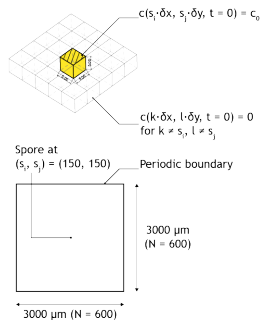
- $L = 3000 \mu\text{m}$
- Discrete blocks of size $\delta x = \delta y = \delta z = 5 \mu\text{m}$
- $N = 600$ subdivisions
- Lattice volume $V_L = 4.5 \times 10^7 \mu\text{m}^3 = 4.5 \times 10^{-5} \text{ mL}$
- Assuming water as medium

» Spore \equiv single block in the center of the lattice, inhibitor source

- Volume $V_s = 125 \mu\text{m}^3$
- Top and bottom side of spore are sealed

» Inhibitor - initial concentrations $c(x, y, t = 0)$:

- At spore: $c(x_s, y_s, t = 0) = c_0$
- Elsewhere: $c(x, y, t = 0) = 0$



GENERAL ASSUMPTIONS



Inhibitor

» Initial concentration is assumed to be contained within the cell wall

- Assumed cell wall thickness: $d = 10 \text{ nm}$ [6]
- Total cell wall volume: $V_{\text{cw}} \approx 6 \cdot 25 \mu\text{m}^2 \cdot 0.01 \mu\text{m} = 1.5 \mu\text{m}^3$
- This concentration is non-replenishable.



» Assumed to have known molecular properties of 1-octen-3-ol:

- Molecular weight: $m_M = 128.21 \text{ g/mol}$
- Density: $\rho = 0.837 \text{ g/mL}$

Figure: 1-octen-3-ol

» Assumed to have similar diffusion properties as glucose:

- Diffusion constant in water at temperature $T = 293.15\text{K}$:
 $D = 600 \mu\text{m}^2/\text{s}$ [2]
(i.e. a molecule wanders on average $\langle x \rangle = \sqrt{6 \cdot D} = 60 \mu\text{m}$ per second)
- Permeability constant in an artificial lipid bilayer:
 $P_s = 1.9 \times 10^{-6} \mu\text{m/s}$ (average from [3] and [1], close to [7])
- Octanol-water partition coefficient: $K \approx 10^{-3}$



Figure: D-Glucose

GENERAL ASSUMPTIONS



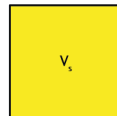
Initial concentration check

» Full spore volume

- Homogeneous distribution, standard temperature and pressure:

$$c_{\max} = \frac{\rho}{m_M} = \frac{0.837 \text{ g/mL}}{128.21 \text{ g/mol}} \approx 6.5 \text{ mol/mL} \quad (1)$$

- Equivalently, $N = N_A c_{\max} V_s = 4.89 \times 10^{14}$ molecules.

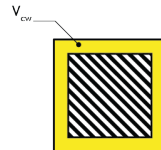


» Cell wall volume only

- The maximum possible concentration is:

$$c_{\text{mcw}} = \frac{\rho V_{\text{cw}}}{m_M V_s} = 6.5 \text{ mol/mL} \cdot 0.012 = 0.078 \text{ mol/mL} \quad (2)$$

- Equivalently, $N = N_A c_{\text{mcw}} V_{\text{cw}} = 7.04 \times 10^{10}$ molecules.
- The experiments use an initial concentration $c_0 = c_{\max, \text{cw}}$.



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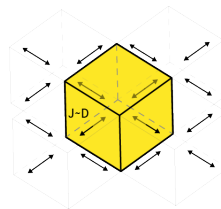
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EXPERIMENT 1 SETUP



Assumptions

- » At $t = 0$, all the inhibitor is contained in a cell wall at the interface between the spore and the medium, having the same diffusion coefficient as water.
- » To prevent numerical error, the concentration in the simulations is normalised by a factor $\lambda = \frac{1}{c_0}$.
- » Diffusion is simulated using the diffusion equation (Fick's Law):
$$\frac{\partial c}{\partial t} = D \nabla^2 c \quad (3)$$
- » Simulation run for $t_{\max} = 3600$ s
- » The times for reaching threshold concentrations of $c_t = k c_0$ ($k = \{10^{-1}, 10^{-2}, \dots, 10^{-6}\}$) are recorded.

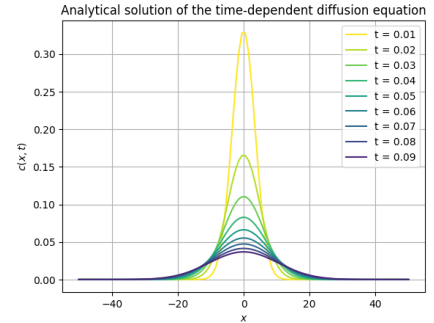


- » Assuming the initial condition is a delta pulse of concentration at coordinates (x_0, y_0)
- » The concentration at an arbitrary position (x, y) at time t can also be computed through the analytical formula[5]:

$$c(x, y, t) = \frac{N^{2/3}}{4\pi Dt} \exp\left(-\frac{(x - x_0)^2 + (y - y_0)^2}{4Dt}\right) \quad (4)$$

- » At the spore site, this equation becomes:

$$c(x_0, y_0, t) = \frac{(V_s c_0)^{2/3}}{4\pi Dt} \quad (5)$$



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EXPERIMENT 2 SETUP



Assumptions

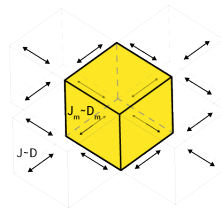
- » All the inhibitor is contained within in a cell wall with the properties of a lipid bilayer at the spore-medium interface.
- » Diffusion through the medium as before
- » Diffusion at the interface follows the flux equation

$$J_m = -D_m \nabla c \quad (6)$$

- » where J_m is the flux through the cell wall such that

$$D_m = \frac{d}{K} P_s = 1.9 \times 10^{-5} \mu\text{m}^2/\text{s} \quad (7)$$

- » Simulation run for $t_{\max} = 7200 \text{ s}$
- » The times for reaching threshold concentrations of $c_t = kc_0$ ($k = \{10^{-1}, 10^{-2}, \dots, 10^{-6}\}$) are recorded.



- » Since the concentration outside (c_{out}) the spore with a volume V and a surface A diffuses much faster than inside (c_{in}), $c_{\text{out}} \approx \text{const.}$

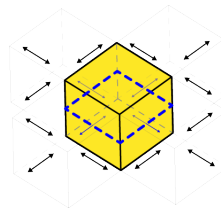
- » Then, an analytical solution is:

$$c_{\text{in}}(t) = c_{\text{out}} - \Delta c(0)e^{-t/\tau}, \quad (8)$$

where τ is a decay constant:

$$\tau = \frac{V}{AD_m} \quad (9)$$

- » Only 2D communication in the numerical simulation $\rightarrow A$ is actually the **circumference** of the cross section!



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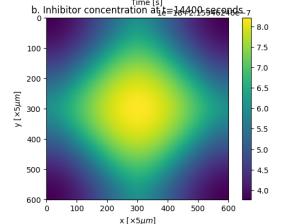
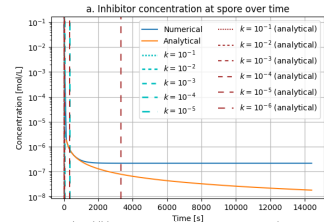
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RESULTS



Experiment 1

- » Results after 4 hours
- » Numerical vs. analytical
 - Alignment of numerical and analytical solution up to $t \approx 1000$ s, then finite size starts to take effect.
 - Under the assumed c_0 , the concentration in this period is low enough to make this deviation insignificant (few thousands of molecules).
- » Threshold times follow a power law
- » Under the given c_0 (maximum that the cell wall can fit), the results seem unrealistic \rightarrow diffusion is too fast

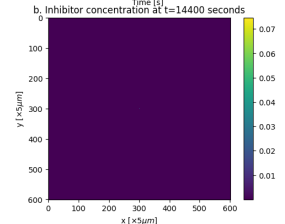
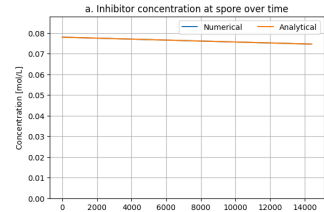


RESULTS



Experiment 2

- » Results after 4 hours
- » Numerical vs. analytical
 - Good alignment of numerical and analytical solution (because medium does not saturate in the given time)
- » Linear decrease in concentration
- » Diffusion appears too slow, threshold of one tenth never reached



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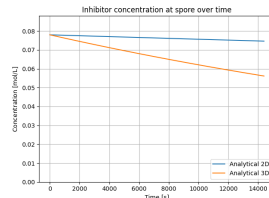
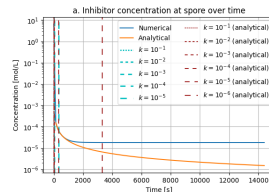
Experiment 2

5 Discussion

DISCUSSION



- » The documented threshold for 1-octen-3-ol inhibition is $10 \text{ mM} = 1 \times 10^{-5} \text{ mol/mL}$ [4].
 - Diffusion through water: threshold immediately reached
 - Diffusion through membrane: threshold not reached
- » The resulting concentrations can be rescaled to different c_0 :
 - If the starting concentration is based on the entire spore volume V_s , then Experiment 1 would reach the 1-octen-3-ol threshold within 30 minutes.
 - More reasonable result, but unrealistic initial concentration.
 - Perhaps the initial concentration is only slightly above threshold?
- » The truth lies in between:
 - Permeation through the membrane is facilitated by proteins (e.g. glucose transporters) \rightarrow increased P_s ?
 - Cell wall permeability modulated during transition from dormancy.
 - In three dimensions: more surface area for diffusion



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