MSc Computational Science
joint programme UvA/VU

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

INTERMEDIATE PRESENTATION DECEMBER

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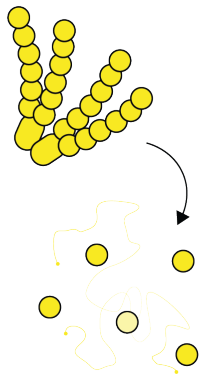
OUTLINE

INTRODUCTION

Hypothesis

- Upon inoculation in an aqueous medium, a germination inhibitor diffuses away from the conidium.
  - Experiments have highlighted **1-octen-3-ol** as a likely candidate [2, 3, 4]
- Once its concentration at the spore falls below a certain threshold, the conidium breaks dormancy and enters a swelling phase.
  - It has been observed that in a **externally non-inhibited** spore swelling begins around **4 hours** from inoculation
- An increased density of spores drives the local inhibitor concentration high, reducing overall germination.
  - Densities above 10^5 sporesl/mL exhibit germination inhibition[4, 5]

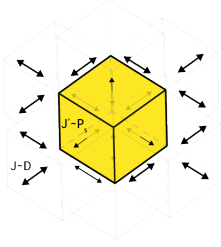


OUTLINE

3D DIFFUSION

Experiment assumptions

- Assumptions as in **Experiment 2: Slow release**, but in 3 dimensions
- Diffusion/permeation can occur over a larger surface area, in more spatial directions
- For memory considerations, a smaller lattice edge length is taken: $L = 128 \times 5\,\mu\text{M} = 640\,\mu\text{M}$
- Total lattice volume: $V = 2.62 \times 10^{-4}$ mL (larger than in 2D system)



3D DIFFUSION

Results: comparison

- Dimensionality of diffusion plays a role, but not as decisive as rate of permeation.

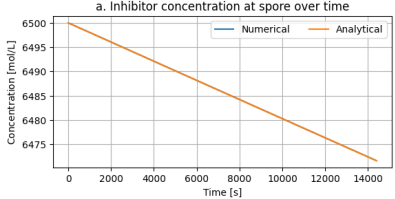


Figure: Results from Experiment 2
(2D system with permeation)

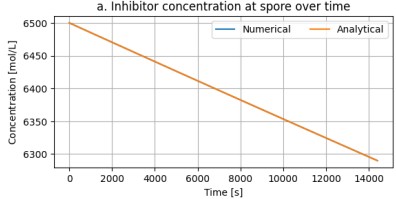


Figure: Results from Experiment 2
(3D system with permeation)

OUTLINE

DISCUSSION

Estimations

- Some quantities can be inferred by fixing specific parameters.
- If swelling begins at $t = 4\,\text{h}$ immediately upon inhibitor drop below the threshold of $10\,\text{mM}$ and

1. if **cell membrane is limiting permeation** from within the spore, then the initial concentration is:

$$c_0 \approx 1.033 \times 10^{-5}\,\text{mol/mL} \tag{1}$$

2. if **spore is completely filled with 1-octen-3-ol**, then the permeation coefficient through the barrier is:

$$P_s = \frac{125\,\mu\text{m}^3}{14\,400\,\text{s} \cdot 150\,\mu\text{m}^2} \ln \frac{6.5 \times 10^3\,\text{mol/mL}}{10^{-5}\text{mol/mL}} \approx 0.001\,174\,\mu\text{m/s} \tag{2}$$

3. or if **cell wall is completely filled with 1-octen-3-ol**, then a diffusion coefficient through the cell wall can be computed

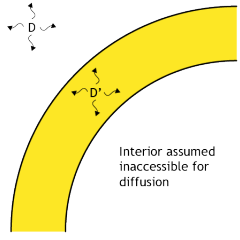
- $$c_0 = c_{\text{out}} - \frac{c_{\text{out}} - c_{\text{in}}(t)}{\exp\left(-\frac{P_s A t}{V}\right)} \tag{3}$$

- $$P_s = \frac{V_s}{tA} \ln\left(\frac{\Delta c(0)}{\Delta c(t)}\right) \tag{4}$$

DISCUSSION

Estimations

- Option 1 means that the concentration drop until break of dormancy is very small. Can the system be this sensitive?
- Estimating P_s like in Option 2 needs to be done with a more realistic initial concentration - hard to determine proportion of cell volume filled with inhibitor.
- Option 3 should consider that the cell wall is **not a barrier**, but a **source region with a different diffusion constant D'**.



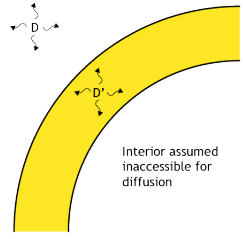
DISCUSSION

Cell wall diffusivity

- An equation describing the relationship between D and P_s is:

$$P_s = \frac{K}{d} D, \tag{5}$$

  where K is a partition coefficient and d is the thickness of the permeating barrier.
- But P_s cannot be known without the assumption that the concentration originates from **beyond** the barrier.
- Therefore, a different mathematical relation needs to be derived or numerical simulations need to be fitted to find D'.

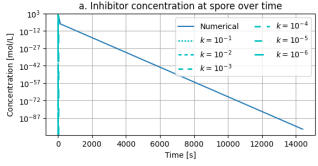


OUTLINE

NEXT STEPS

Educated guess of D'

- The diffusion coefficient is not always known but it can be related to comparable systems.
- Diffusion of glucose through a cellulose film has $D \approx 1.7 \times 10^{-11}\,\mathrm{m}^2/\mathrm{s} = 17\,\mu\mathrm{m}^2/\mathrm{s}$[1]
- Results of 3D simulation with this diffusivity show a concentration drop to 10^{-12}mol/mL in the first 15 minutes.



NEXT STEPS

Physical derivation of D'

- The diffusion coefficient can be estimated e.g. using the Einstein relation:

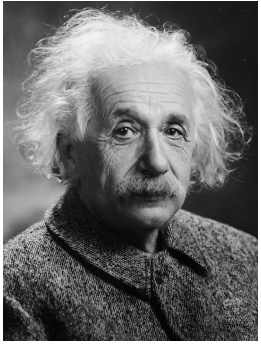
$$D = \mu k_B T, \tag{6}$$

where μ is the mobility of a particle (a factor relating the drift velocity to an applied force $\langle \vec{v} \rangle = \mu \vec{f}$,

- of the Stokes-Einstein relation:

$$D = \frac{k_B T}{6\pi\eta a}, \tag{7}$$

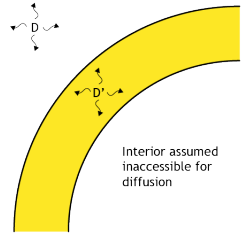
where η is the viscosity of the medium and a is the radius of the particle.



NEXT STEPS

Mathematical derivation of D'

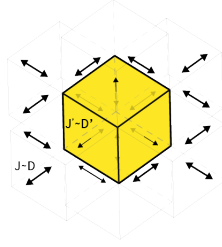
- An attempt at an analytical derivation can be done using Fick's laws and Random Walk models.
- Fixing a starting concentration c_0, the diffusivity of the surrounding medium D, the end concentration in the cell wall $c(t_i)$ and the time for inhibitor release t_i — D' can be expressed.
- Might be non-trivial to solve depending on what is assumed at the interface.



NEXT STEPS

Numerical estimation of D'

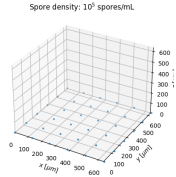
- The diffusion coefficient can be approximated by stochastic simulations.
- Running for various D' candidates, recording the threshold release time t_i' and optimising towards $|t_i' - t_i|$, where $t_i = 4$ h
- The resulting D' can be related to other physical properties, verifying consistency with the properties of 1-octen-3-ol and the cell wall layers.
- D' may have a more complex interpretation, e.g. the cumulative effect of different cell wall layers.

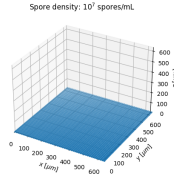


NEXT STEPS

The effect of spore density

- Assume we find a reasonable D'.
- Is it consistent with spore-density-dependent inhibition?
- Different spore densities need to be simulated to observe saturation effects (10^4, 10^5, ... 10^9 spores/mL).
- Start with a regular grid of spores, then try other distributions.
- Results should be related to observed decrease in germination percentage.
- **If there is a slow-release system that reaches germination threshold for 1 spore, does it explain increased rates of inhibition for high spore densities?**





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[2] Gilma Silva Chitarra et al. “1-Octen-3-ol inhibits conidia germination of Penicillium paneum despite of mild effects on membrane permeability, respiration, intracellular pH, and changes the protein composition.”. In: *FEMS microbiology ecology* 54 1 (2005), pp. 67–75. URL: https://api.semanticscholar.org/CorpusID:24273006.

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[5] Maryam Ijadpanahsaravi et al. “The impact of inter- and intra-species spore density on germination of the food spoilage fungus Aspergillus niger.”. In: *International journal of food microbiology* 410 (2023), p. 110495. URL: https://api.semanticscholar.org/CorpusID:265268197.