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

INTERMEDIATE PRESENTATION JANUARY

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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 [1, 2, 3]

» 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
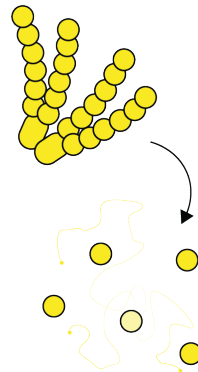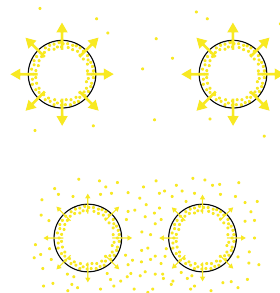
INTRODUCTION

━━━ Hypothesis

» An increased spore density reduces the average probability of germination.

   – Densities above 10^5 spores/mL exhibit germination inhibition[3, 4]

» This could be caused by a reduction of the concentration gradient between the spore and the medium, which causes more inhibitor to stay in the spore.
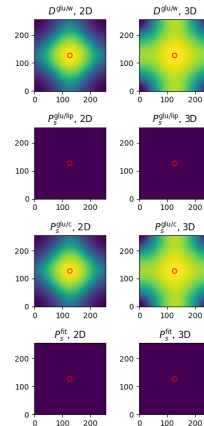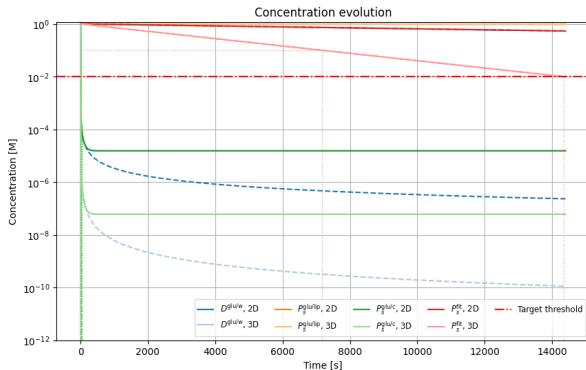
OUTLINE

SINGLE-SPORE EXPERIMENTS

Overview

» Lattice size of $L = 256 \times 5\,\mu\text{m} = 1280\,\mu\text{m}$

» Initial concentration of $c_0 = 1.018\,\text{M}$

» $t_{\max} = 4\,\text{h}$

» List of experiments

1. Superficial release with D of glucose in water (2D)
2. Superficial release with D of glucose in water (3D)
3. Slow release with P_s of a lipid bilayer membrane (2D)
4. Slow release with P_s of a lipid bilayer membrane (3D)
5. Slow release with P_s of a CNF film (2D)
6. Slow release with P_s of a CNF film (3D)
7. Slow release with analytically fitted P_s (2D)
8. Slow release with analytically fitted P_s (3D)

» Fitted permeation constant is $P_s = 2.675 \times 10^{-8}\,\text{cm/s}$

SINGLE-SPORE EXPERIMENTS

Results

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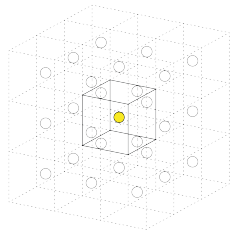
MULTI-SPORE EXPERIMENTS

Assumptions

» Spores assumed to be arranged in a regular three-dimensional grid.

» Assuming a large inoculum, in which the central part exhibits characteristic germination behaviour and the boundaries are irrelevant.

» Therefore, the grid can be considered infinite.

» Since the spores **repeat periodically**, it suffices to simulate a **single spore** in a **volume of variable size** L and a triply periodic boundary.
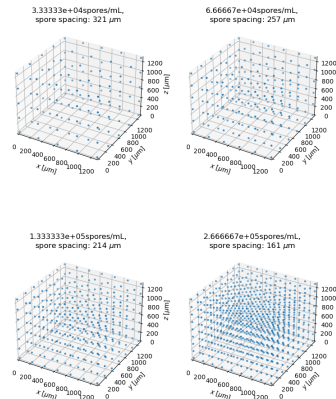
MULTI-SPORE EXPERIMENTS

Experiment setup

» The size L of the simulation volume depends on the spore density.

» In [4] a difference in germination is observed between 5000 spores and 40000 spores in a $150\,\mu$L medium.

» 4 densities are chosen for the experiment:

  – $5000\,\text{spores}/150\,\mu\text{L} \approx 0.33 \times 10^4\ \text{spores/mL}$
  – $10\,000\,\text{spores}/150\,\mu\text{L} \approx 0.67 \times 10^4\ \text{spores/mL}$
  – $20\,000\,\text{spores}/150\,\mu\text{L} \approx 1.33 \times 10^5\ \text{spores/mL}$
  – $40\,000\,\text{spores}/150\,\mu\text{L} \approx 1.33 \times 10^5\ \text{spores/mL}$

» Spore spacings of 315, 250, 200 and 160 μm respectively.

» The simulations are run for $t_{\max} = 4\,$h.
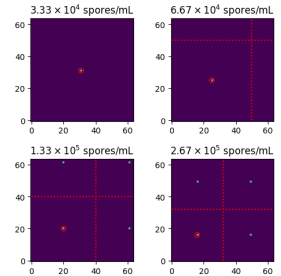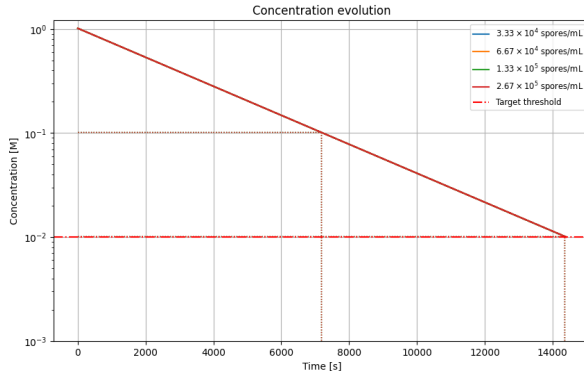
MULTI-SPORE EXPERIMENTS

Results

Figure: Concentrations at $t = 4$ h. The boundaries are aligned with the largest lattice for comparison, i.e. smaller lattices are repeated.

MULTI-SPORE EXPERIMENTS

▬▬▬ Discussion

» No observable saturation effects at $t = 4\,\mathrm{h}$ for any of the given spore densities → the space around each spore is too large to become saturated.

» To observe saturation, higher local densities are necessary. These are present when:

   – the spores densify at the bottom of the medium due to gravity;

   – isolated pockets form between spores due to their irregular distribution / aggregation.

» Next experiments:

   – Rectangular grids of spores at the bottom of a 3D volume;

   – Irregular distributions / aggregation studies with spherical spores.

2.666667e+05 spores/mL, spore spacing: 161 μm
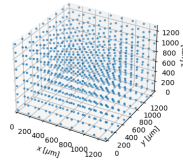
2.666667e+05 spores/mL, spore spacing: 39 μm

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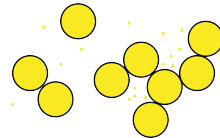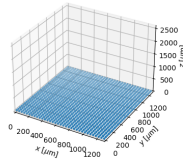
[1] Gilma Silva Chitarra et al. "1-Octen-3-ol inhibits conidia germination of Penicillium paneum despite of mild effects on membrane permeability, respiration, intracellular pH, and changes the protein composition.". In: *FEMS microbiology ecology* 54 1 (2005), pp. 67–75. URL: https://api.semanticscholar.org/CorpusID:24273006.

[2] Gilma Silva Chitarra et al. "Germination of Penicillium paneum Conidia Is Regulated by 1-Octen-3-ol, a Volatile Self-Inhibitor". In: *Applied and Environmental Microbiology* 70 (2004), pp. 2823 –2829. URL: https://api.semanticscholar.org/CorpusID:19828197.

[3] Erika Herrero-García et al. "8-Carbon oxylipins inhibit germination and growth, and stimulate aerial conidiation in Aspergillus nidulans.". In: *Fungal biology* 115 4-5 (2011), pp. 393–400. URL: https://api.semanticscholar.org/CorpusID:33687383.

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[4]   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.