Mycofluidics

The Fluid Mechanics of Fungal Chimerism and Spore Dispersal

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Introduction to Mycology

Notable Fungi

- Rice blast
- Cordyceps
- White nose syndrome

Figure: Cordyceps versus an ant

Introduction to Mycology

- A **chimera** is an organism that is made up of cells with genetically different nuclei.
- Can reproduce asexually via spores
- Most fungi grow as **hyphae**, which are filamentous structures that grow at their tips.

Moran Process

A **Moran process** is a stochastic process that models processes affecting genetic diversity.The dynamics of two different populations of nuclei within a fungi can be modeled as a two species Moran process.

Roper M, Simonin A, Hickey PC, Leeder A, Glass NL (2012) Nuclear dynamics in a fungal chimera (preprint).

Previous Work

- Fungal chimerism gives rise to the internal genetic diversity of fungi. It has been found that nuclear mixing within the mycelium keeps genetically different nuclei well-mixed and in stable proportions throughout the fungus.

Roper M, Simonin A, Hickey PC, Leeder A, Glass NL (2012) Nuclear dynamics in a fungal chimera (preprint).

Our Experiment: Question

Is a fungus able to maintain its genetic diversity? If so, how?

Our Experiment: Method

- *Neurospora crassa* (*N. crassa*), a type of bread mold, with GFP or DsRed proteins
- GFP and DsRed cause nuclei to emit a green and red fluorescence, respectively.

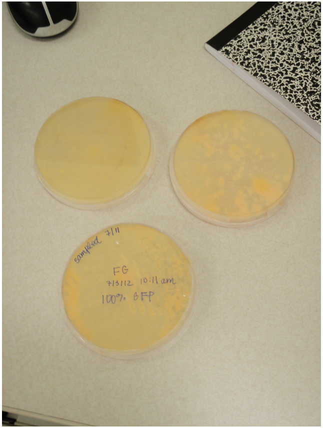


Figure: *N. crassa* Cultures

Our Experiment: Method

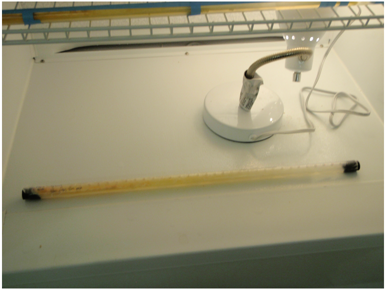


Figure: Race Tubes

- We built and innoculated race tubes with both GFP and DsRed.
- When the colonies started sporulating, we took images with a microscope.

Our Experiment:Method

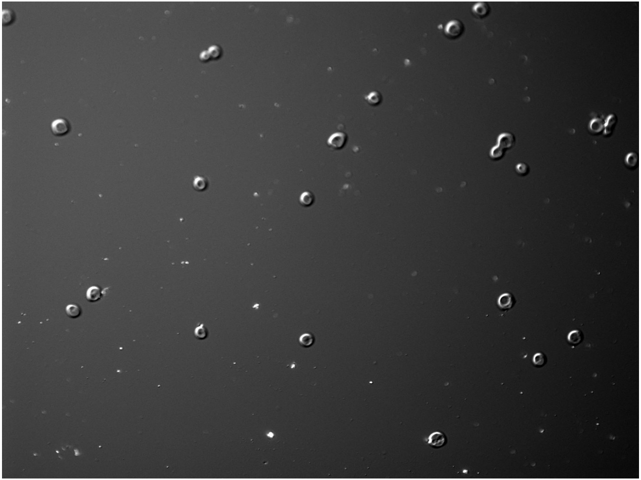


Figure: DIC Images of Spores

Our Experiment:Method

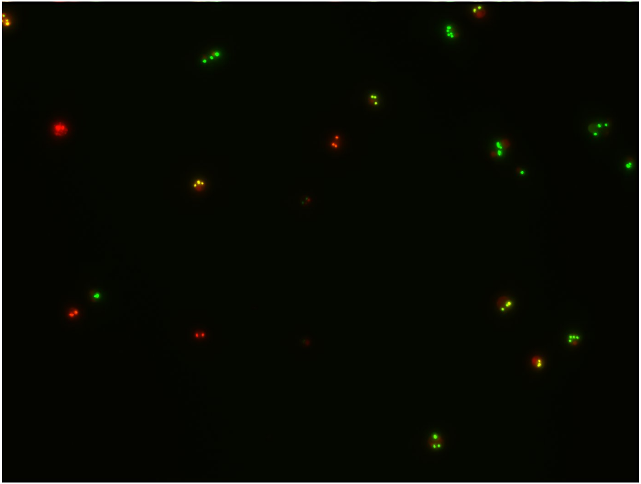


Figure: Fluorescent DsRed and GFP

Our Experiment: Method

Image Analysis

- Find the percentage of spores that have only red nuclei (p_R), only green nuclei (p_G), and both (p_Y).
- Find the frequency of the number of nuclei present in a spore to get $\mathbb{P}(N_{nucl} = n)$.
- From this, we can solve

$$\sum_n \mathbb{P}(N_{nucl} = n) p_r^n = p_R$$

$$\sum_n \mathbb{P}(N_{nucl} = n)(1 - p_r)^n = p_G$$

for p_r, the probability that a nucleus is red.

Our Experiment: Method

P_r Fitting

- Estimate p_r from likelihood function

$$\mathcal{L}(N_R, N_G, N_Y) = \frac{N!}{N_R! N_G! N_Y!} p_R^{N_R} p_G^{N_G} p_Y^{N_Y}$$

- Find maximum of log-likelihood

$$\frac{N_R}{p_R}\frac{\partial p_R}{\partial p_r} + \frac{N_G}{p_G}\frac{\partial p_G}{\partial p_r} - \frac{N_Y}{p_Y}\left(\frac{\partial p_R}{\partial p_r} + \frac{\partial p_G}{\partial p_r}\right) = 0$$

Our Experiment: Method

Image Analysis

- **Otsu's Method**–histogram-based thresholding
  - Finds optimum value in a bi-modal histogram such that the intra-class variance is minimal.

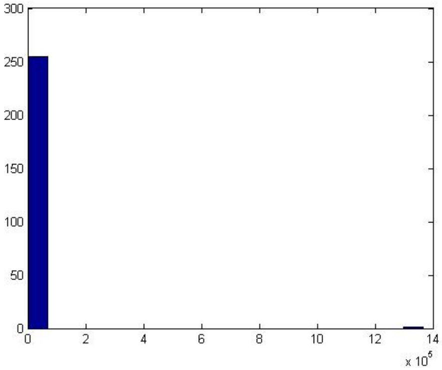


Figure: Initial Intensity Histogram

Our Experiment: Method

Image Analysis

- Modified Otsu's Method to take in the original histogram
- Used this second threshold to identify nuclei within spores

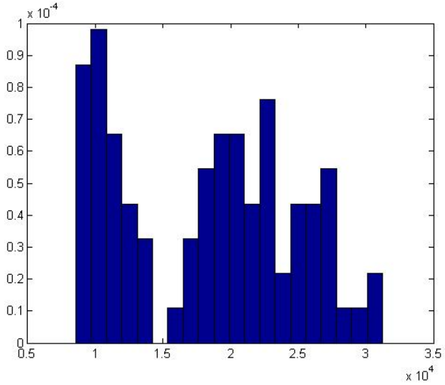
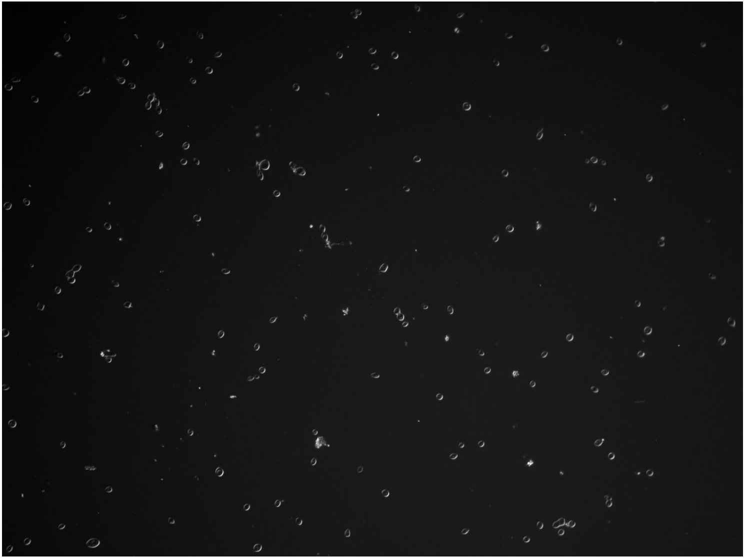


Figure: Normalized Intensity Histogram

Thresholding



Thresholding

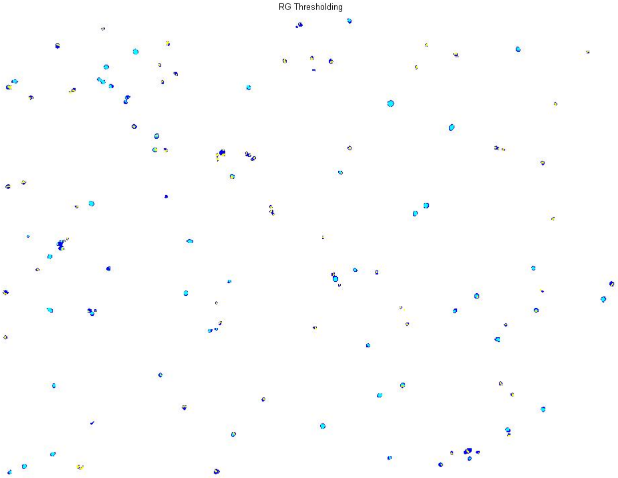


Figure: Otsu's thresholding

Our Experiment: Results

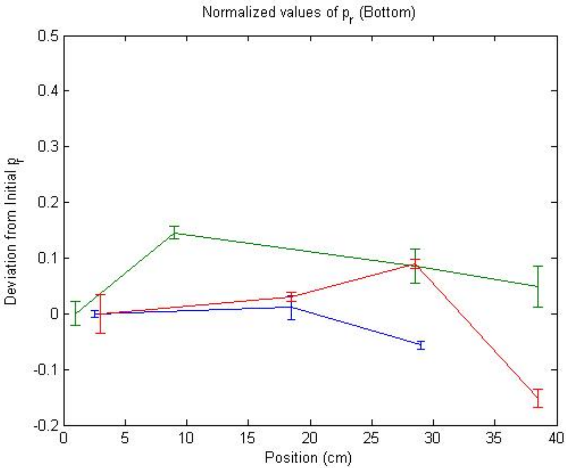


Figure: p_r values along race tube

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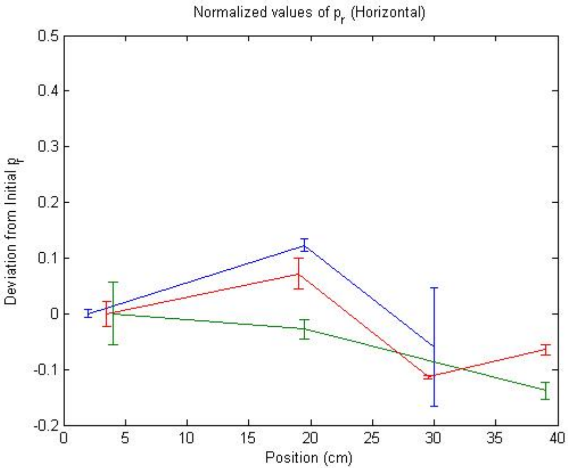

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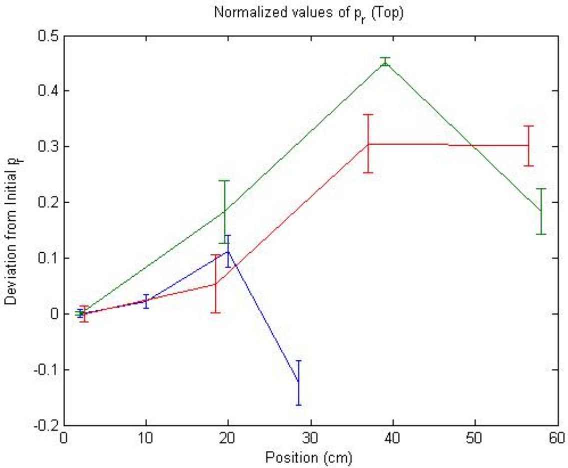

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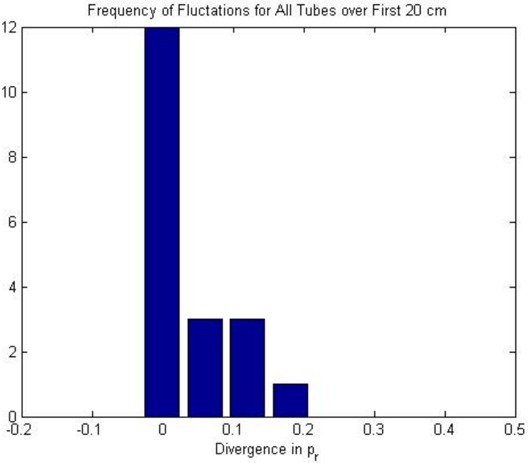

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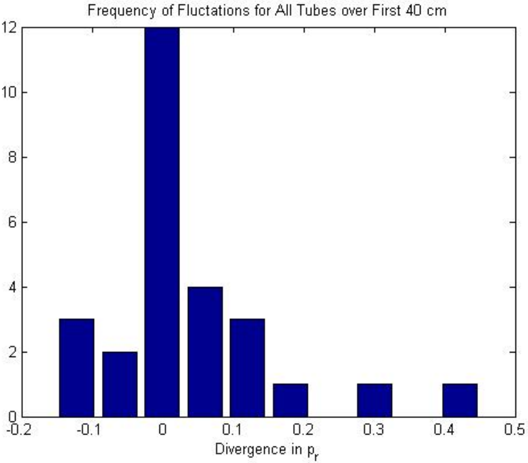

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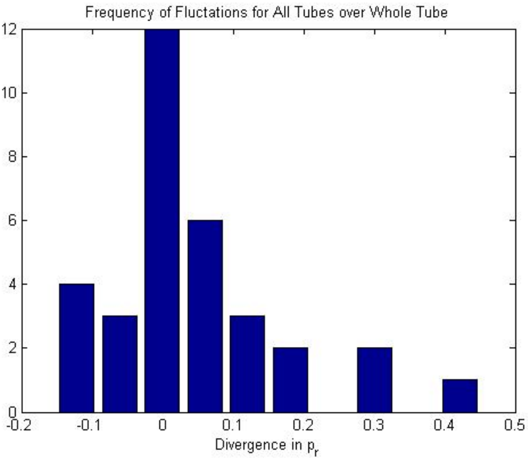

Our Experiment: Results

Interpretation of p_r Fluctuations

- Moran process
- Effect of shaking and accelerated growth

Our Experiment: Future Work

- Improve p_r fitting
  - Multinomial distribution with dependency parameters
- Segmentation issues
  - Clumping
  - Illumination/Saturation sensitivity