A Thin-Film Extensional Flow Model for Biofilm Expansion by Sliding Motility

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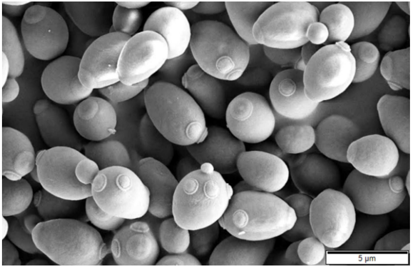
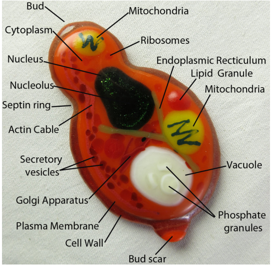
Yeast

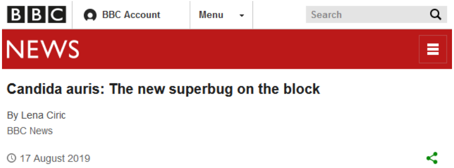
- Single-cell fungi ($\sim 4\,\mu m$ diameter)
    - Food and drink
    - Waste management and biofuel production
- Baker's yeast (*S. cerevisiae*) is a common model organism
    - Similar to plant and animal cells
    - First eukaryotic genome to be sequenced
    - Helps develop anti-fungals and understand (cancer) cell division

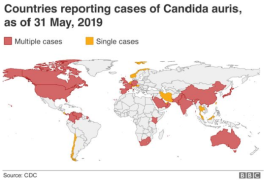




Fungal Infections

- Pathogenic yeasts colonise medical devices and cause infections
  - Resist antimicrobial therapy — surgery often needed
  - Dangerous to immunocompromised people
  - Affects 1–2% of ICU patients, with up to 40% mortality rate[1]
- Emerging pathogen *C. auris*: Japan 2009, 5 continents since
  - Highly resistant and difficult to diagnose



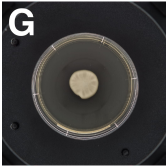


- We seek common mechanisms underlying yeast biofilm growth

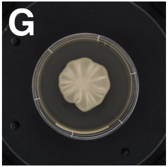
[1]P. G. Pappas et al., *Nat. Rev. Dis. Primers* 4 (2018), 18026.

Yeast Biofilms

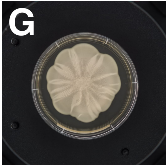
- Pathogenic yeasts form **biofilms**: sticky communities of cells and fluid existing on surfaces
  - Assist nutrient transport
  - Provide physical barrier to anti-fungals
- Lab-grown biofilms of baker's yeast form a floral pattern



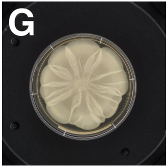
(a) Day 3



(b) Day 5



(c) Day 7

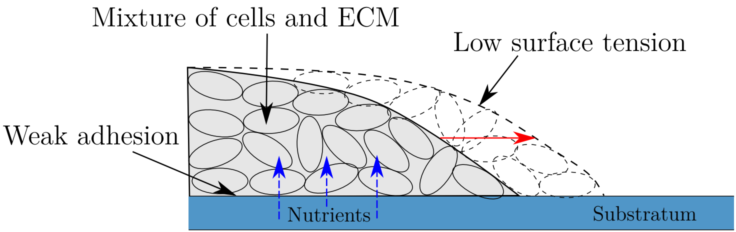


(d) Day 10

- Mechanisms of growth only understood qualitatively

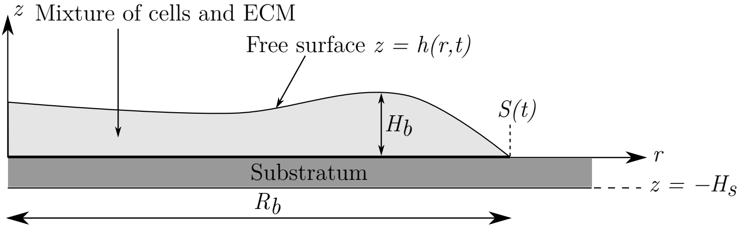
Sliding Motility

- **Hypothesis**: yeast biofilms expand by sliding motility[2]
  - Yeast adheres weakly to substratum — enables radial growth as cells proliferate
  - Biofilm takes up nutrients from the substratum
  - Nutrient consumption produces new cells and extracellular fluid
  - Cells and fluid spread passively as a unit



[2] T. B. Reynolds and G. R. Fink, *Science* 291 (2001), pp. 878–881.

Two-Phase Fluid Model



- Axisymmetric cylindrical geometry.
  - Biofilm occupies $0 \leq r \leq S(t)$ and $0 \leq z \leq h(r, t)$
- Biofilm is a mixture of two Newtonian viscous fluid phases:
  - Living cells $\phi_n(r, z, t)$ and ECM $\phi_m(r, z, t)$, with $\phi_n + \phi_m = 1$
  - Similar physical properties: $\rho_n = \rho_m$, $\mu_n = \mu_m$, *etc.*
  - Large interphase drag: $\boldsymbol{u}_n = \boldsymbol{u}_m$
- Thin aspect ratio

$$\frac{H_s}{R_b} = \varepsilon \ll 1, \qquad \frac{H_b}{R_b} = \mathcal{O}(\varepsilon)$$

Governing Equations

- Mass balance (fluid phases)

$$\frac{\partial \phi_n}{\partial t} + \nabla \cdot (\phi_n \boldsymbol{u}) = \psi_n \phi_n g_b - \psi_d \phi_n$$

$$\frac{\partial \phi_m}{\partial t} + \nabla \cdot (\phi_m \boldsymbol{u}) = \psi_m \phi_n g_b + \psi_d \phi_n$$

- Mass balance (nutrients in the substratum and biofilm)

$$\frac{\partial g_s}{\partial t} = D_s \nabla^2 g_s$$

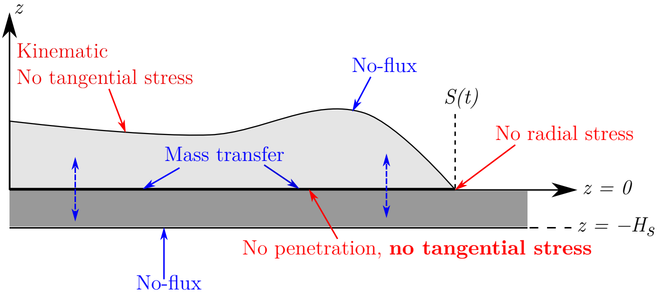
$$\frac{\partial g_b}{\partial t} + \nabla \cdot (g_b \phi_m \boldsymbol{u}) = D_b \nabla^2 g_b - \eta \phi_n g_b$$

- Momentum balance (fluid mixture), $\mathrm{Re} \approx 0.001$

$$\nabla \cdot \boldsymbol{\sigma} = \boldsymbol{0}$$

Boundary Conditions

- Boundary conditions for nutrients and fluids close the model



- Nutrient transfer conditions on $z = 0$:

$$D_s \frac{\partial g_s}{\partial z} = -Q\left(g_s - g_b\right), \quad D_b \frac{\partial g_b}{\partial z} = -Q\left(g_s - g_b\right)$$

- **No tangential stress** on substratum models weak adhesion
- Free surface normal stress proportional to local curvature:

$$\widehat{\boldsymbol{n}} \cdot (\phi_\alpha \boldsymbol{\sigma} \cdot \widehat{\boldsymbol{n}}) = -\gamma\kappa \quad \text{on} \quad z = h$$

Extensional Flow Scaling

- Scaling based on relevant physics
    - Thin biofilm (aspect ratio $\varepsilon \ll 1$)
    - Low surface tension
    - Nutrient-limited growth
- Variables

$$(r, z) = (R_b\hat{r}, \varepsilon R_b\hat{z}), \quad (u_r, u_z) = (\psi_n G R_b \hat{u}_r, \varepsilon \psi_n G R_b \hat{u}_z),$$

$$t = \frac{\hat{t}}{\psi_n G}, \quad g_s = G\hat{g}_s, \quad g_b = G\hat{g}_b, \quad p = \psi_n G \mu \hat{p}$$

- Parameters (estimated based on experiments)

$$\Psi_m = \frac{\psi_m}{\psi_n} = 0.11, \quad \Psi_d = \frac{\psi_d G}{\psi_n} = 0, \quad \gamma^* = \frac{\varepsilon\gamma}{\Psi_n G R_b \mu} = 0,$$

$$D = \frac{D_s}{\psi_n G R_b^2} = 4.34, \quad \mathrm{Pe} = \frac{\psi_n G R_b^2}{D_b} = 0.95, \quad \Upsilon = \frac{\eta R_b^2}{D_b} = 3.15,$$

$$Q_s = \frac{Q R_b}{\varepsilon D_s} = 2.09, \quad Q_b = \frac{Q R_b}{\varepsilon D_b} = 8.65$$

Thin-Film Model

- Expand variables

$$h \sim h_0(r,t) + \varepsilon^2 h_1(r,t), \quad \phi_n \sim \phi_{n0}(r,z,t) + \varepsilon^2 \phi_{n1}(r,z,t), \qquad etc.$$

- Dimensionless model (dropping hats)

$$\frac{1}{r}\frac{\partial}{\partial r}(r u_r) + \frac{\partial u_z}{\partial z} = (1 + \Psi_m)\,\phi_n g_b$$

$$\frac{\partial \phi_n}{\partial t} + \frac{1}{r}\frac{\partial}{\partial r}(r u_r \phi_n) + \frac{\partial}{\partial z}(u_z \phi_n) = \phi_n g_b - \Psi_d \phi_n$$

$$\frac{\partial g_s}{\partial t} = D\left[\frac{1}{r}\frac{\partial}{\partial r}\left(r\frac{\partial g_s}{\partial r}\right) + \frac{1}{\varepsilon^2}\frac{\partial^2 g_s}{\partial z^2}\right]$$

$$\mathrm{Pe}\left(\frac{\partial g_b}{\partial t} + \boldsymbol{\nabla}\cdot[(1-\phi_n)g_b\boldsymbol{u}]\right) = \frac{1}{r}\frac{\partial}{\partial r}\left(r\frac{\partial g_b}{\partial r}\right) + \frac{1}{\varepsilon^2}\frac{\partial^2 g_b}{\partial z^2} - \Upsilon \phi_n g_b$$

$$-\frac{\partial p}{\partial r} + \frac{2}{r}\frac{\partial}{\partial r}\left(r\frac{\partial u_r}{\partial r}\right) - \frac{2}{3}\frac{\partial}{\partial r}\left[\frac{1}{r}\frac{\partial}{\partial r}(r u_r) + \frac{\partial u_z}{\partial z}\right] + \frac{\partial}{\partial z}\left(\frac{\partial u_z}{\partial r} + \frac{1}{\varepsilon^2}\frac{\partial u_r}{\partial z}\right) - \frac{2}{r^2}u_r = 0$$

$$-\frac{\partial p}{\partial z} + 2\frac{\partial^2 u_z}{\partial z^2} - \frac{2}{3}\frac{\partial}{\partial z}\left[\frac{1}{r}\frac{\partial}{\partial r}(r u_r) + \frac{\partial u_z}{\partial z}\right] + \frac{1}{r}\frac{\partial}{\partial r}\left[r\left(\frac{\partial u_r}{\partial z} + \varepsilon^2\frac{\partial u_z}{\partial r}\right)\right] = 0$$

Thin-Film Model

- Expand variables

$$h \sim h_0(r,t) + \varepsilon^2 h_1(r,t), \quad \phi_n \sim \phi_{n0}(r,z,t) + \varepsilon^2 \phi_{n1}(r,z,t), \qquad etc.$$

- Simplified leading-order model

$$\frac{1}{r}\frac{\partial}{\partial r}(r u_{r0}) + \frac{\partial u_{z0}}{\partial z} = (1+\Psi_m)\,\phi_{n0} g_{b0}$$

$$\frac{\partial \phi_{n0}}{\partial t} + \frac{1}{r}\frac{\partial}{\partial r}(r u_{r0}\phi_{n0}) + \frac{\partial}{\partial z}(u_{z0}\phi_{n0}) = \phi_{n0} g_{b0} - \Psi_d \phi_{n0}$$

$$\frac{\partial^2 g_{s0}}{\partial z^2} = 0$$

$$\frac{\partial^2 g_{b0}}{\partial z^2} = 0$$

$$\frac{\partial^2 u_{r0}}{\partial z^2} = 0$$

$$-\frac{\partial p_0}{\partial z} + \frac{1}{3}\frac{\partial}{\partial z}\left[\frac{1}{r}\frac{\partial}{\partial r}(r u_{r0}) + \frac{\partial u_{z0}}{\partial z}\right] + \frac{\partial^2 u_{z0}}{\partial z^2} = 0$$

Thin-Film Model

- Integrating across biofilm depth eliminates z dependence

$$\bar{\phi}_n = \frac{1}{h}\int_0^h \phi_n \, \mathrm{d}z.$$

- Applying BCs gives a 1D system for $r \in [0, S(t)]$

$$\frac{\partial h_0}{\partial t} + \frac{1}{r}\frac{\partial}{\partial r}\left(r u_{r0} h_0\right) = \left(1+\Psi_m\right)\bar{\phi}_{n0} g_{b0} h_0$$

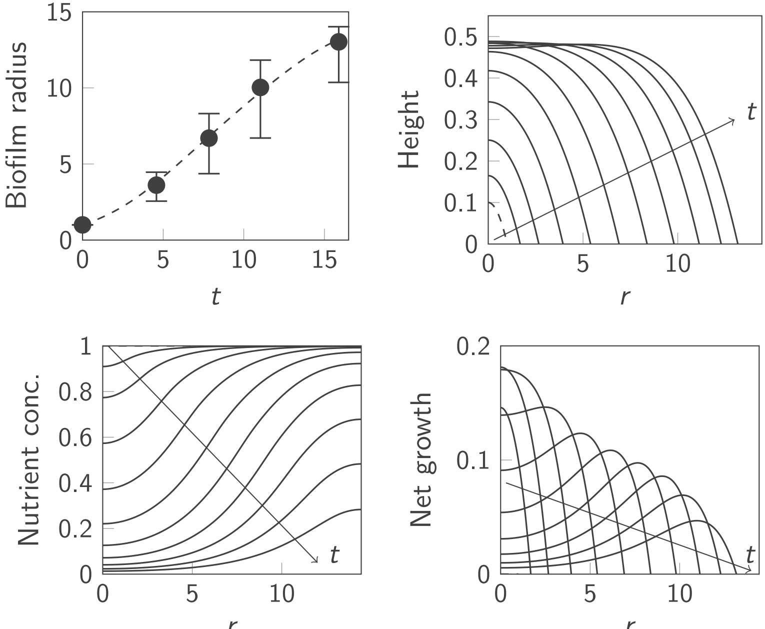
$$\frac{\partial \bar{\phi}_{n0}}{\partial t} + u_{r0}\frac{\partial \bar{\phi}_{n0}}{\partial r} = \bar{\phi}_{n0}\left[g_{b0} - \Psi_d - \left(1+\Psi_m\right)\bar{\phi}_{n0} g_{b0}\right]$$

$$\frac{\partial g_{s0}}{\partial t} = D\left[\frac{1}{r}\frac{\partial}{\partial r}\left(r\frac{\partial g_{s0}}{\partial r}\right) - Q_s\left(g_{s0} - g_{b0}\right)\right]$$

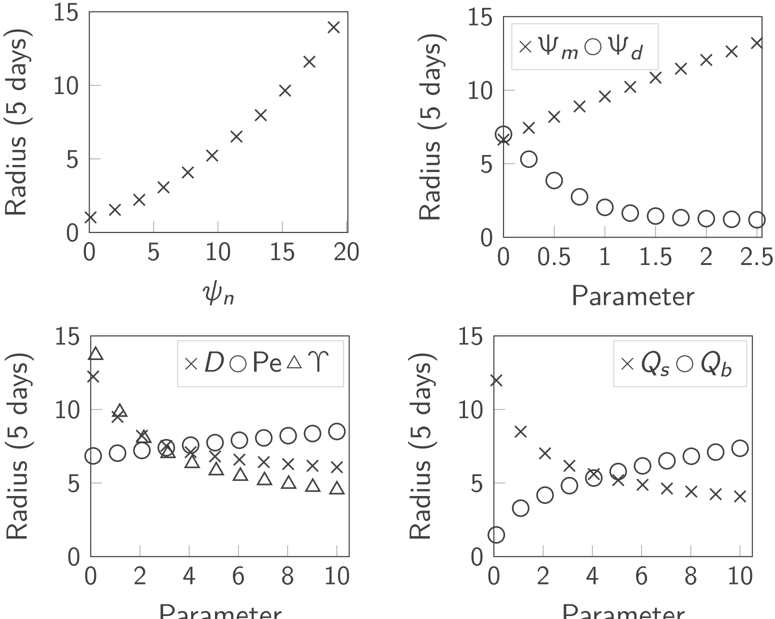
$$\mathrm{Pe}\left[h_0\frac{\partial g_{b0}}{\partial t} + \frac{1}{r}\frac{\partial}{\partial r}\left(r u_{r0}\left(1-\bar{\phi}_{n0}\right) g_{b0} h_0\right)\right] = \frac{1}{r}\frac{\partial}{\partial r}\left(r h_0 \frac{\partial g_{b0}}{\partial r}\right) + Q_b\left(g_{s0} - g_{b0}\right) - \Upsilon \bar{\phi}_{n0} g_{b0} h_0$$

$$4\frac{\partial}{\partial r}\left[\frac{h_0}{r}\frac{\partial}{\partial r}\left(r u_{r0}\right)\right] - 2\frac{u_{r0}}{r}\frac{\partial h_0}{\partial r} = 2\left(1+\Psi_m\right)\frac{\partial}{\partial r}\left(\bar{\phi}_{n0} g_{b0} h_0\right) - \gamma^* h_0 \frac{\partial}{\partial r}\left[\frac{1}{r}\frac{\partial}{\partial r}\left(r\frac{\partial h_0}{\partial r}\right)\right]$$

Numerical Solutions

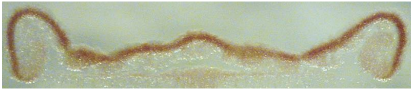


Effect of Parameters on Expansion Speed

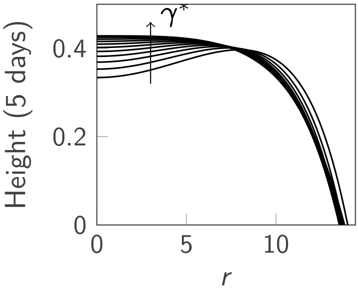


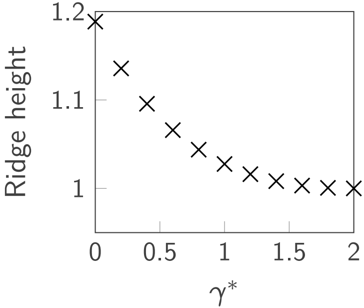
What About Surface Tension?

- In different experiments, yeast colonies can contain ridges[3]



- Surface tension represents cell–cell adhesion strength[4]
- Non-zero γ^* does not affect size, but inhibits ridge formation





[3] J. Maršíková et al., *BMC Genom.* 18 (2017), pp. 1–16.

[4] G. Forgacs et al., *Biophys. J.* 74 (1998), pp. 2227–2234.

Summary

- Yeast biofilms are a leading cause of bloodstream infections
- Two-phase thin-film extensional flow model describes expansion by sliding motility[5]
- Model shows how to enhance/inhibit growth and ridge formation
- Future work: strong adhesion model, pattern formation

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[5] A. Tam et al., *Proc. Royal Soc. A* 475 (2019), 20190175.