Slides are available on my website.
`http://lukaseigentler.github.io`

Competition and coexistence in bacterial biofilms: the role of founder cells

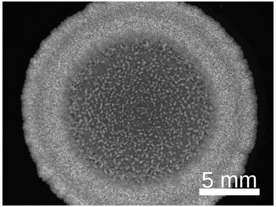
Warwick-Cambridge Quantitative Cell Biology Symposium

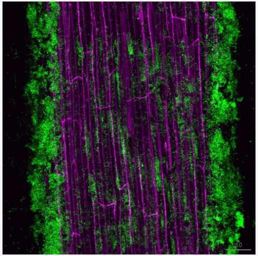
16/05/2024

Lukas Eigentler (University of Warwick)
joint with Margarita Kalamara, Graeme Ball, Nicola R. Stanley-Wall, Fordyce A. Davidson (all Dundee), Cait E. MacPhee (Edinburgh)

Biofilms

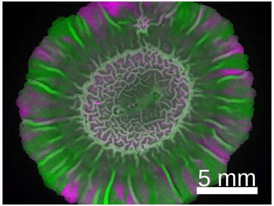
- Bacterial biofilms are surface-adhering multicellular collectives embedded in a self-produced extracellular matrix.
- Biofilms can have both beneficial and detrimental effects on the surrounding environment.
- Example: the soil-dwelling bacterium *Bacillus subtilis* forms biofilms on the roots of plants, where some strains promote the growth of plants.
- To fully realise their potential as biocontrol agents, strains need to be capable of coexisting with (or outcompeting) other biofilm-forming strains in the rhizosphere.

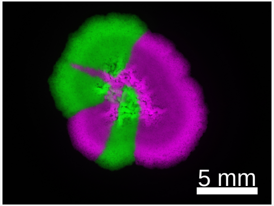




Competition within biofilms

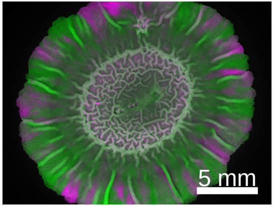
- Competition in biofilms is underpinned by kin discrimination.
- Many mechanisms of kin discrimination require spatial co-location of strains.
- Take a step back: need to understand the role of spatial structure first.

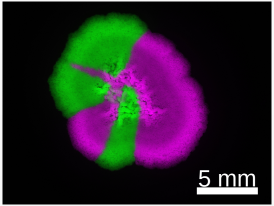




Competition within biofilms

- Spatial structure is best studied using isogenic strains: all other competitive mechanisms (e.g. kin discrimination) are excluded from the model system by design.
- Isogenic strains: Low founder densities promote spatial segregation and formation of spatial sectors.[1,2]
- Questions: How does spatial structure arise and how does it affect competitive interactions?

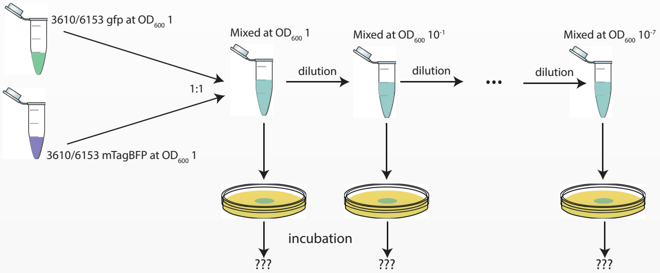




[1] van Gestel, J. et al.: *ISME J.* 8.10 (2014)
[2] Martinez-Garcia, R. et al.: *PLoS Comput. Biol.* 14.4 (2018)

Methods

Experimental assay:



Tested for

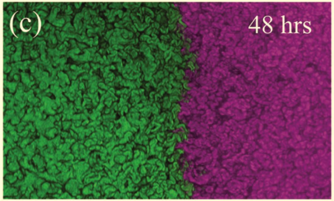
- *B. subtilis* NCIB3610 (“standard” lab strain).
- *B. subtilis* NRS6153 (isolated from garden soil in Tayport, Fife, UK).

Methods

Mathematical model for isogenic strain pair:

$$
\begin{aligned}
\frac{\partial B_1}{\partial t} &= \nabla \cdot \left(\left(1 - (B_1 + B_2)\right) \nabla B_1 \right) + B_1 \left(1 - (B_1 + B_2)\right), \\
\frac{\partial B_2}{\partial t} &= \nabla \cdot \left(\left(1 - (B_1 + B_2)\right) \nabla B_2 \right) + B_2 \left(1 - (B_1 + B_2)\right).
\end{aligned}
$$

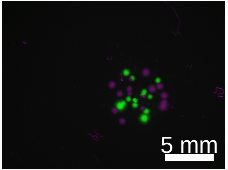
- Circular domain $\Omega = \{x \in \mathbb{R}^2 : \|x\| \leq R\}$.
- Logistic growth
- Diffusion with density-dependent diffusion coefficient, motivated by experimental observation that initially separated colonies abut rather than merge[1].

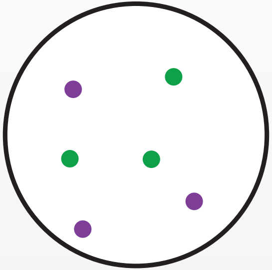


[1] Matoz-Fernandez, D. et al.: *Soft Matter* 16.13 (2020)

Initial conditions

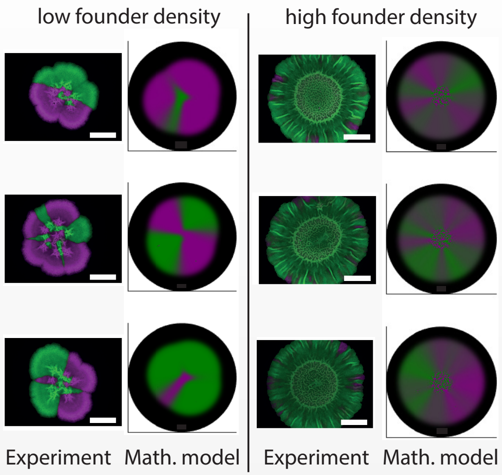
- What are appropriate initial conditions?
- In experiments, cells settle at random locations within the initial spot and grow to small micro-colonies.
- In the model, we position initial "cell patches" at random locations in the domain centre.
- Each model patch represents 1 microcolony $\Rightarrow$ tool to modulate founder density.





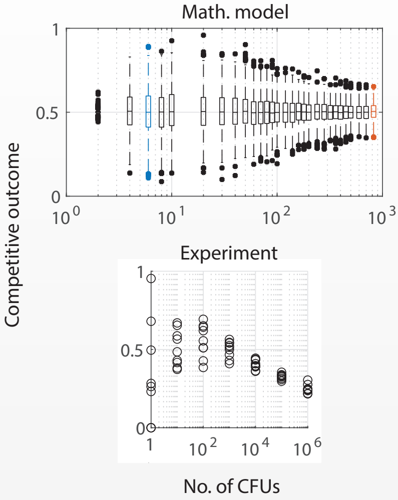
Variability in competitive outcome

- High founder density: no spatial structure and initial strain ratio consistently determines competitive outcome.
- Low founder density: spatial segregation occurs. Large variability in competitive outcome for fixed initial strain ratio.



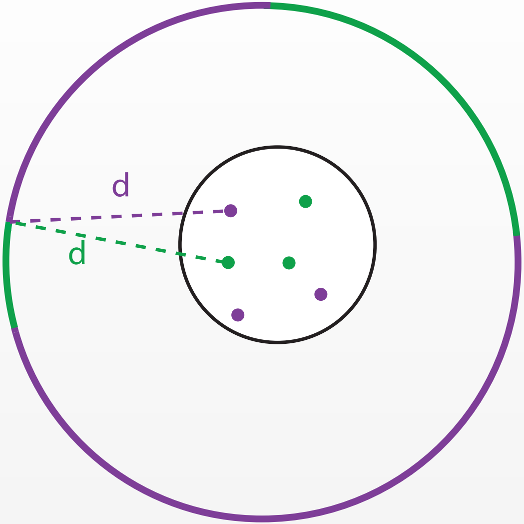
Variability in competitive outcome

- Founder density significantly affects phenotype and variability in competitive outcome.
- Variability increases with decreasing founder density.



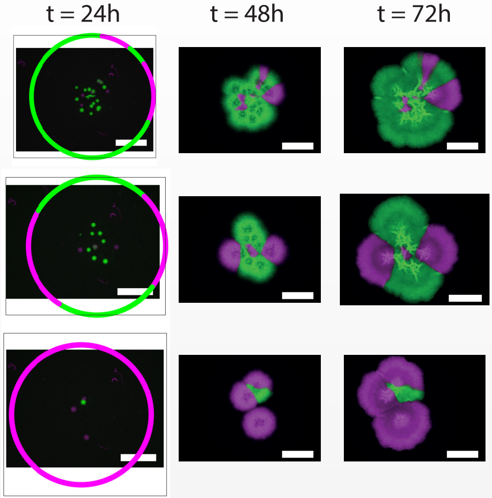
Disentangling variability

- Hypothesis: only initial patches that can drive the biofilm's radial expansion contribute to outcome density.
- We define a quantity that, based on the initial cell locations, measures a strain's "access to free space"



Disentangling variability

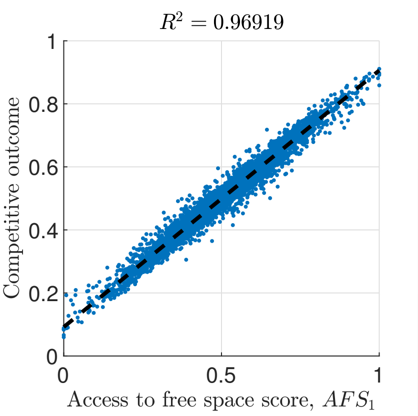
- Hypothesis: only initial patches that can drive the biofilm's radial expansion contribute to outcome density.
- We define a quantity that, based on the initial cell locations, measures a strain's "access to free space"



Access to free space predicts outcome

- Access to free space determines competitive outcome.

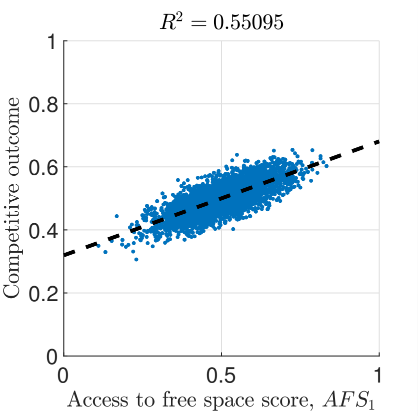
low founder density



Access to free space predicts outcome

- Access to free space determines competitive outcome.
- Slope of relation between access to free space and competitive outcome depends on founder density.

high founder density



Non-isogenic strains

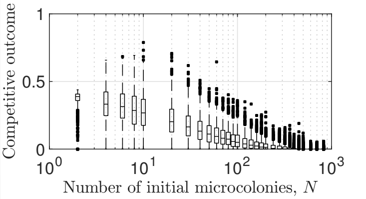
- Do these results also hold if strains are non-isogenic and interact through local antagonisms?

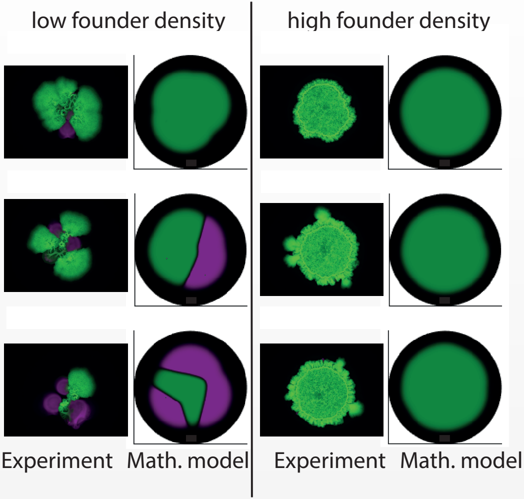
$$\frac{\partial B_1}{\partial t} = \nabla \cdot \left(\left(1 - \frac{B_1 + B_2}{k}\right) \nabla B_1 \right) \quad + B_1 \left(1 - \frac{B_1 + B_2}{k}\right) - B_1 B_2,$$
$$\frac{\partial B_2}{\partial t} = \nabla \cdot \left(d \left(1 - \frac{B_1 + B_2}{k}\right) \nabla B_2 \right) + r B_2 \left(1 - \frac{B_1 + B_2}{k}\right) - c B_2 B_1.$$

Nondimensional model, i.e. d, r, c are ratios of corresponding dimensional parameters.

Non-isogenic strains

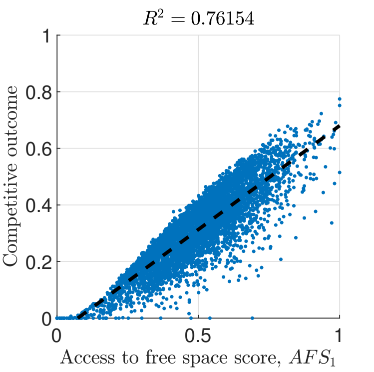
- High founder density: competitive exclusion.
- Low founder density: spatial segregation enables coexistence.
- Decreases in founder density cause (i) increased variability in competitive outcome, (ii) higher (on average) densities of weaker strain.





Access to free space predicts outcome

- Access to free space remains a reliable predictor of competitive outcome for low founder densities.
- Competition for space is the dominant mode of interaction for low founder densities.



Conclusions

- Large variability in competitive outcome occurs for biofilms inoculated at low founder density, induced by the random positions of founder cells within the inoculum.
- Competition for space is the dominant mode of competition for low founder densities.
- Antagonistic interactions determine competitive outcome for high founder densities.

Acknowledgements

Collaborators:

- Margarita Kalamara (Univ. of Dundee)
- Graeme Ball (Univ. of Dundee)
- Cait E. MacPhee (Univ. of Edinburgh)
- Nicola R. Stanley-Wall (Univ. of Dundee)
- Fordyce A. Davidson (Univ. of Dundee)

Funding:

UKRI Biotechnology and Biological Sciences Research Council

University of Dundee

References

Slides are available on my website.

`http://lukaseigentler.github.io`

[1] Eigentler, L., Davidson, F. A. and Stanley-Wall, N. R.: 'Mechanisms driving spatial distribution of residents in colony biofilms: an interdisciplinary perspective'. *Open Biol.* 12.220294 (2022).

[2] Eigentler, L., Kalamara, M., Ball, G., MacPhee, C. E., Stanley-Wall, N. R. and Davidson, F. A.: 'Founder cell configuration drives competitive outcome within colony biofilms'. *ISME J.* 16.6 (2022), pp. 1512–1522.

[3] Eigentler, L., Stanley-Wall, N. R. and Davidson, F. A.: 'A theoretical framework for multi-species range expansion in spatially heterogeneous landscapes'. *Oikos* 2022.8 (2022), e09077.