Transiently Multicellular Marine Bacteria as Self-Patterning Collectives

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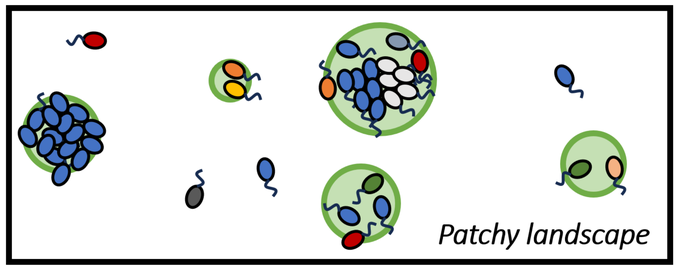
Overview

- Far from being well-mixed, marine ecosystems are often patchy (for marine heterotrophs)
- Patchiness shapes ecological dynamics:
  - Competition for resources
  - Interactions with other species and phage
  - Successive immigration and emigration events
- Life in a patchy landscape can be life at high cell density

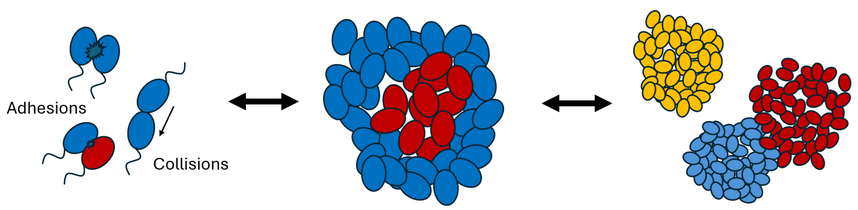


A prevalent consequence of life in a patch: ***Cells stick to each other***

- As cells interact, they form transiently multicellular groups
- Emergent multicellular states can tip the scales of microbial ecology

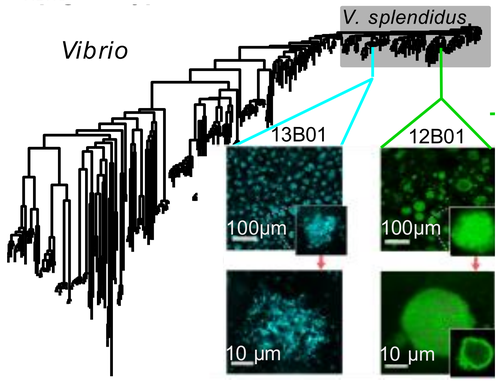
How does the feedback between cell interactions and emergent multicellular states shape the ecology of a patchy marine environment?

Cellular Interactions *Multicellular Structure* *Multicellular Interactions*

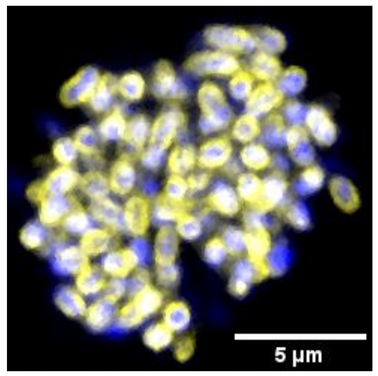


Our experimental model

- *Vibrio splendidus:* a heterotrophic Gram-negative bacteria
- Forms groups in the presence of alginate polysaccharides
- Is common and widespread in coastal oceans
- Has many closely related strains and species
- There is rampant horizontal gene transfer among heterotrophic *Vibrio*



Left: Phylogeny of *Vibrio*, using marker gene *hsp60*.
Inset: Two particular strains of *Vibrio splendidus* are highlighted. Both clump in the presence of alginate polysaccharides, with different cell and group structure.



Blue: Staining cell DNA with Syto-9 (488nm ex. 510 em.)
Yellow: Staining lipids with Nile Red (560 ex. 630 em.)

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If I'm not here at my poster but you want to talk more, come find me in person (here is what I look like) or online:
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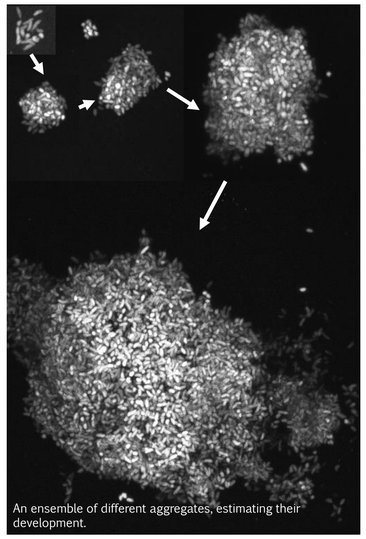
Exploring physical development within aggregates

What does group development look like?
How do cell-level traits propagate to the group level, and vice versa?

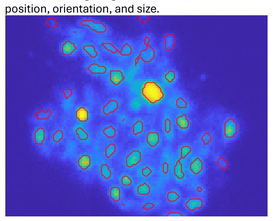
Cell-level properties ⟷ Group-level properties

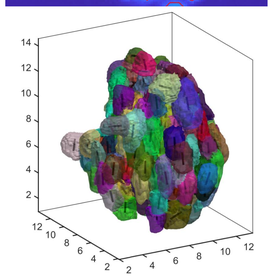
$$r(t), \psi(t), \ell(t) \longleftrightarrow \phi(t), S(t), \lambda(t)$$

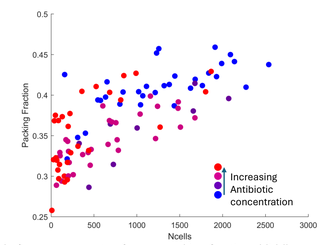
Using confocal microscopy to obtain the 3D cell assembly of individual aggregates, we can obtain a list of observables that track the aggregate's development.

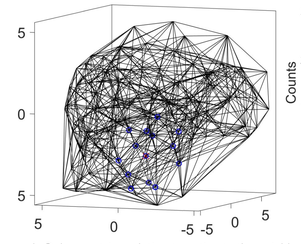


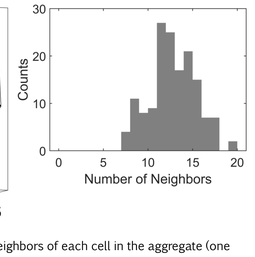
Automated image segmentation measures cell position, orientation, and size.





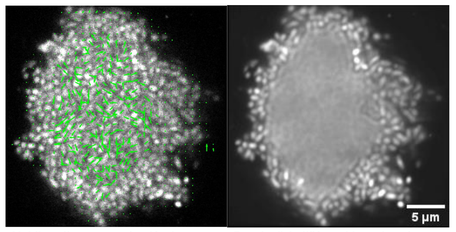






Left: measurements of group packing fraction. Middle: an example Delaunay triangulation to estimate the neighbors of each cell in the aggregate (one shown in red, neighbors in blue). Right: a histogram of the number of neighbors for one aggregate.

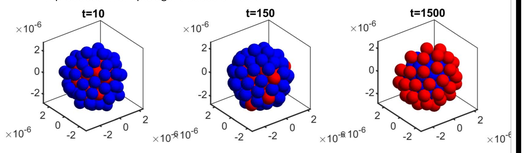
Cells sort and become phenotypically heterogeneous



Mobile cells are contained in the core. Particle Image Velocimetry shows that cells in the core move rapidly. Mean speed: 25 $[\mu m/s]$

Exterior "shell" is stable and static. Frame-averaging of a 1 min video shows that there are no blurry regions around the periphery.

This type of cell sorting is different to previously known bacterial aggregates, and is not reproduced in simple agent-based simulations.



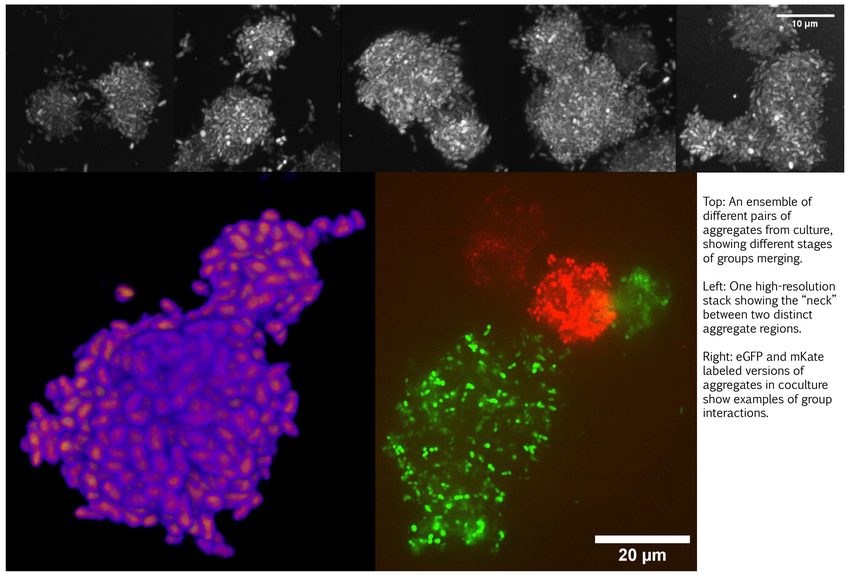
Red: Mobile
Blue: Immobile

In simulation, mobile cells are not easily retained in the interior, unlike empirical observations. This indicates that some cellular feedback may be necessary to achieve the experimentally-observed spatial pattern.

Merging of separate groups

What sets the encounter rate of groups in a patchy marine environment?
How often will groups merge and interact?

Using differentially-labeled *Vibrio* groups, we can measure frequency of combination as a function of sweeps in: Initial cell density, time in culture, species, agitation speed, nutrient concentration, and more.



Top: An ensemble of different pairs of aggregates from culture, showing different stages of groups merging.

Left: One high-resolution stack showing the "neck" between two distinct aggregate regions.

Right: eGFP and mKate labeled versions of aggregates in coculture show examples of group interactions.

The future:

How do we square ecological predictions in the context of spatially-dependent interactions?

What properties of marine microbial communities can be scaffolded by physical interactions?

Can we understand how cell-level and group-level properties of transiently multicellular organisms are connected?

How do we combine physical and metabolic approaches to interrogating group structure?