

From Non-equilibrium Physics to Biology

The case of bacterial colony formation

Herbert Levine – UCSD

- Introduction to pattern formation - systems+questions
- Why turn biological? Why do theory?
- Bacterial colonies under stress
 - Branching structures and diffusion-limited growth
 - Solvability theory - how to control a pattern
 - External vs. internal control - the chiral transition
 - Cooperation to enhance flexibility - vortex structure
- General lessons and new challenges

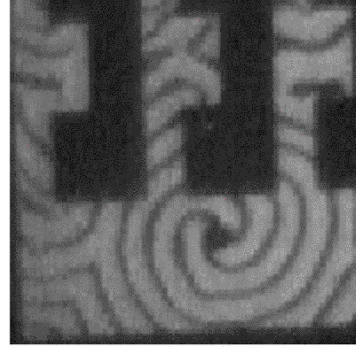
Thanks to E. Ben-Jacob; also I. Cohen, D. Kessler, W. Rappel

Pattern-formation Physics



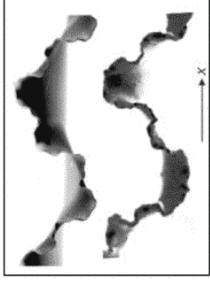
-Applications to CM physics,
fluid mechanics, materials science,
geophysics, catalytic chemistry

- Exciting progress over the past two decades
- Cooperative endeavor utilizing theory, experiment, and computer simulation

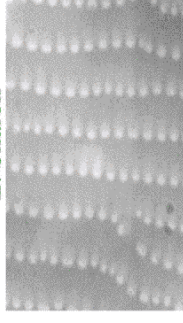


Issues/systems

- Fracture - ductile vs. brittle behavior, how to incorporate microstructural physics into macroscopic elastodynamics
 - Experiments on PMMA, glass, crystalline silicon, rubber
- Chemical waves and fronts - role of fluctuations, effects of spatial heterogeneity, waves with complex periodicity
 - Experiments on BZ reaction, microemulsions
- Spatiotemporal Chaos - defect dynamics, measures of pattern complexity and predictability, fluctuation effects in bifurcations
 - Experiments on convection in fluids, liquid crystals



E. Sharon



I. Epstein



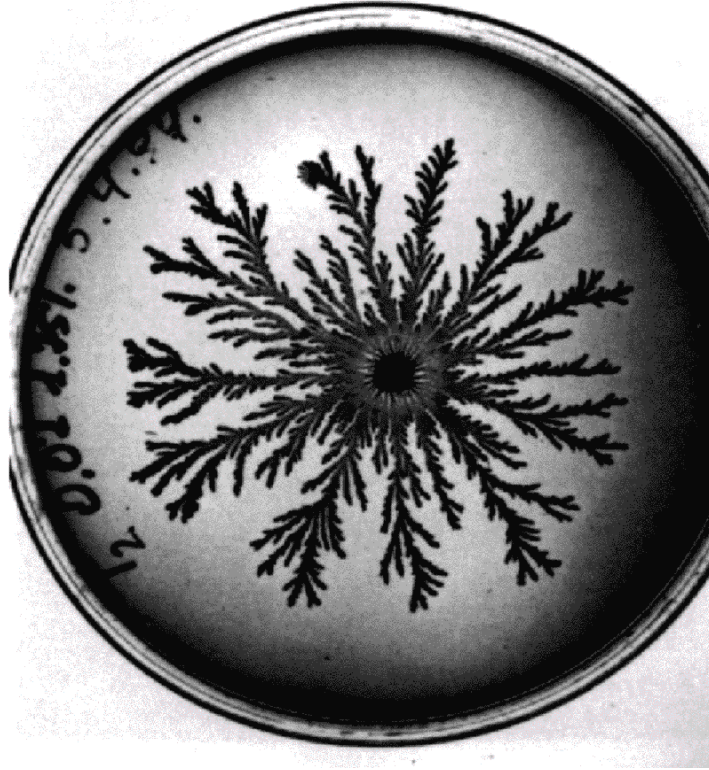
K. Daniels

Why study patterns in biological systems?

Biological systems offer new challenges and opportunities for non-equilibrium physics

1. Biology is in the midst of a tremendous revolution towards a more quantitative and predictive science. Can physics play a critical role?
2. Genetic engineering allows for a degree of control over the microscopic interactions not possible in non-living systems
3. Biological constituents (organisms, cells, protein complexes) are much more complex than simple molecules. Does this allow for more complex pattern-forming behavior? Is this behavior merely quantitatively different or are there new principles of pattern formation at work?

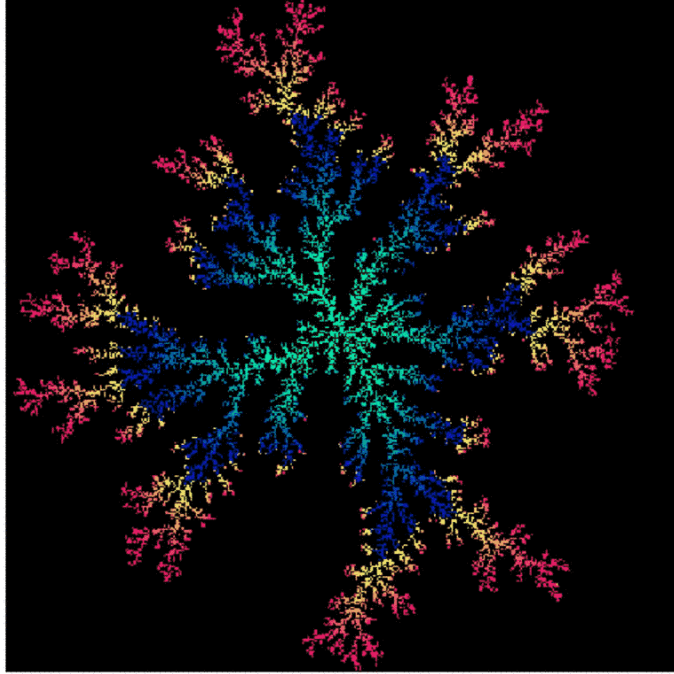
Branched growth in Bacillus*



From lab of E. Ben-Jacob;
following earlier work by
Japanese group

Growth is limited by the
diffusion of nutrient

* and related species!



Bacterial branching
patterns has physical
equivalent

One random walker
attaches wherever it
hits and another one is
released

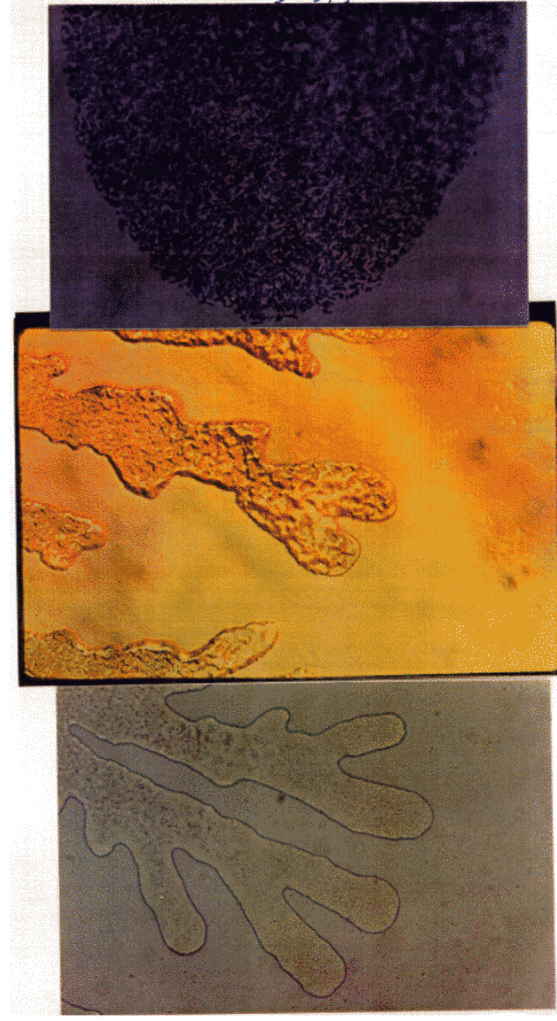
DLA – Witten and Sander (1981) - picture courtesy of L. Sander

Why do theory?

- Identification of pattern-forming schemata
 - The first step in analyzing a biological pattern is to place it within a specific schema
 - Here, bacterial colonies appear to fall within the diffusion-limited growth class (n.b. one can be fooled! - Turing pattern fiasco)

First task is to understand why?

A closer look at the branches



Note: wetting envelope, discrete cells.

Models of Bacterial Patterns

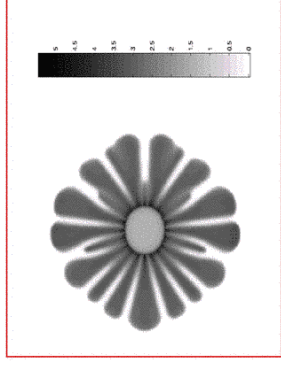
Diffusive instability in the simplest reaction-diffusion model of bacterial pattern formation

Bacterial growth (A) limited by nutrient (B) which is diffusion-limited

$$\frac{\partial c_A}{\partial t} = c_A c_B + \nabla \cdot (D_A(c_A) \nabla c_A)$$

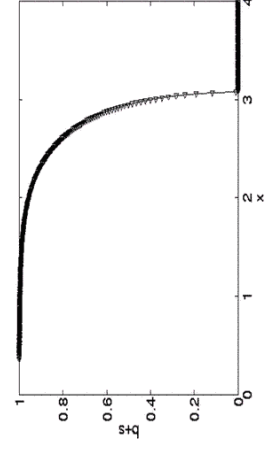
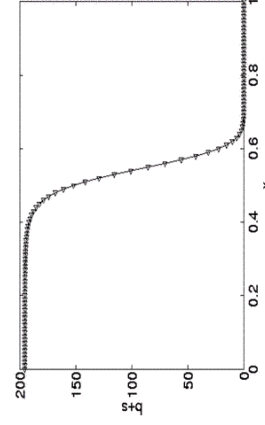
$$\frac{\partial c_B}{\partial t} = -c_A c_B + D_B \nabla^2 c_B$$

The non-linear diffusion of the bacteria is due to the need for making liquid film. One can then show that this system exhibits a diffusive instability which drives the branching pattern formation .
(NB : similar effects arise from discreteness)



Physics of the bacterial instability

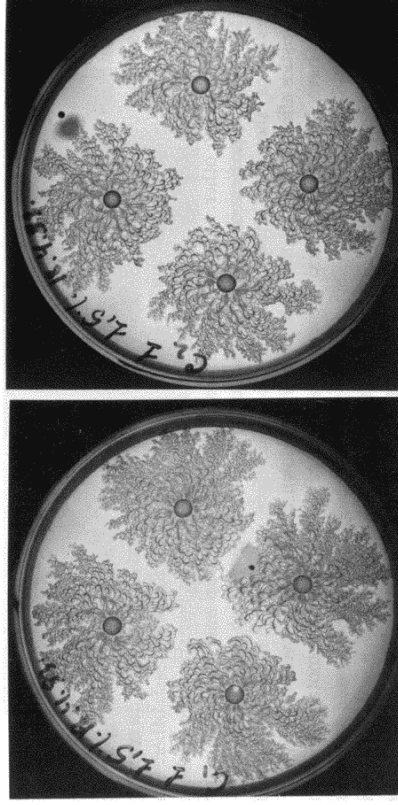
- Steady-state front solutions (separating the colony from the surrounding region) exist for all velocities
- Localized initial conditions lead to a unique “marginally stable” selected solution
- Nonlinear diffusion changes the problem from “linear-type I” in which only the leading edge matters to “nonlinear-type II” where the full equation determines a unique front (with compact support) which is selected
- Nutrient diffusion can bias the local growth velocity of a nonlinear front and thereby cause instability



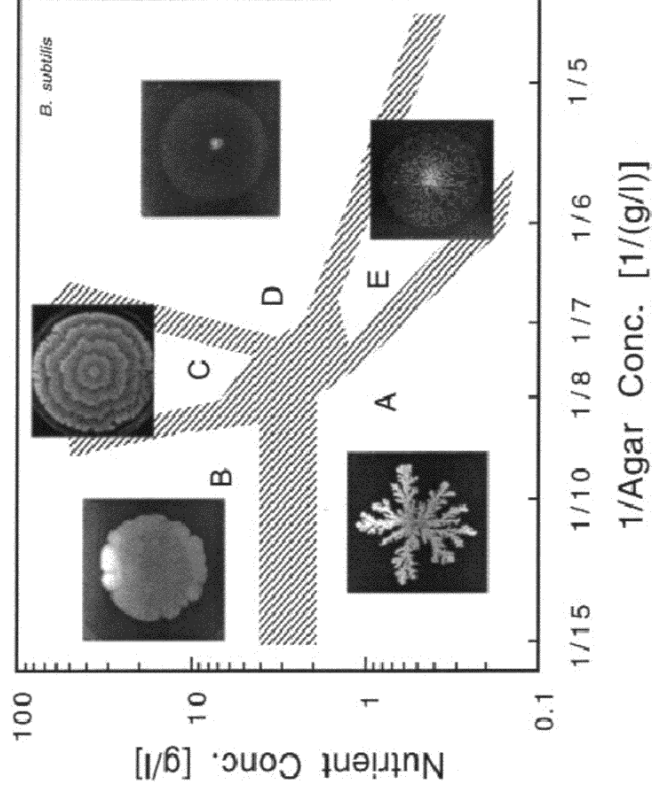
Why do theory?

- Identification of pattern-forming schemata
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 - Here, bacterial colonies fall within the diffusion-limited growth class
- Detailed understanding of the generic behavior of this type of process
 - Predictions which can be verified independent of knowing the precise underlying model
 - Identifies which features are unique to the biological system and hence require regulatory feedback

COMPLEXITY and REPRODUCIBILITY



Phase diagram for Bacillus



Microscopic Solvability

- How can a small perturbation control a large pattern?
- One possibility is afforded by the fact that microscopic physics is often needed to cutoff singularities in macroscopic equations (e.g. string theory might get rid of space-time singularities)
- In diffusion-limited growth, this idea has been worked out in detail for a number of model equations - simplest is geometrical model of crystal growth

$$v_n = (1 + \epsilon \cos 4\theta) \left(\kappa + \gamma \frac{\partial^2 \kappa}{\partial s^2} \right)$$

If $\gamma=0$, we have family of steady-state solutions; but the initial value problem is ill-posed, due to unbounded stability spectrum

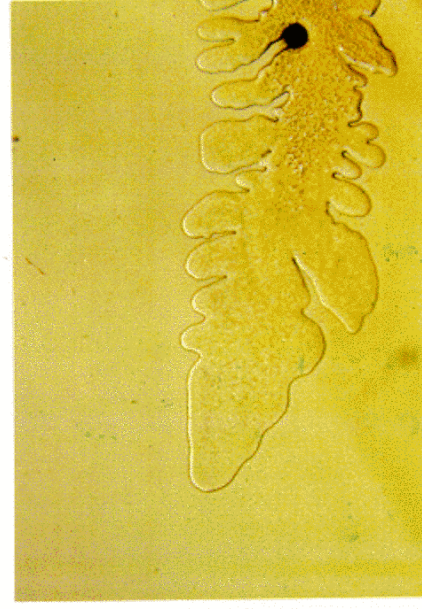
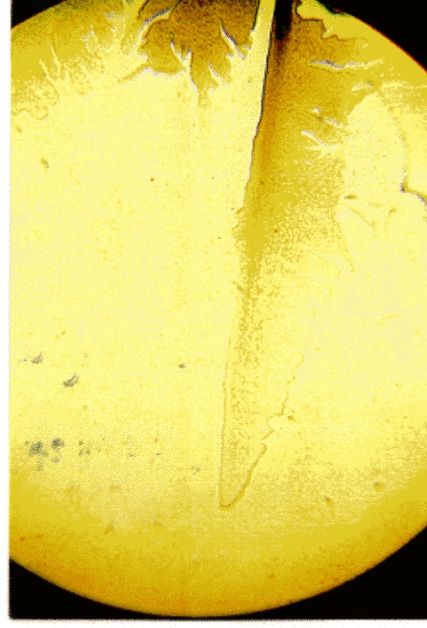
$$\frac{v \cos \theta}{1 + \epsilon \cos 4\theta} = \frac{\partial \theta}{\partial s} \quad \theta(0) = 0 \rightarrow \theta(\infty) = \pi/2$$

When we introduce a microscopic cutoff on the spectrum, we get

$$\frac{v \cos \theta}{1 + \epsilon \cos 4\theta} = \frac{\partial \theta}{\partial s} + \gamma \frac{\partial^3 \theta}{\partial s^3}$$

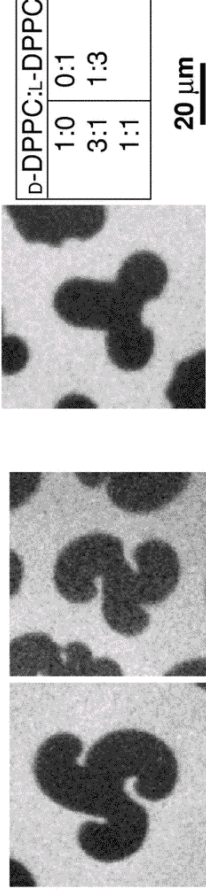
Now, there are new allowed modes at the tail which are non-analytic in the microscopic regulation parameter γ ; we then have to fix $\theta'(0)$ and $\theta''(0)$ to find unique smooth trajectory

$$\theta'(0) = g\left(\frac{v}{\epsilon^{7/4}}\right) e^{-\frac{c}{\sqrt{\gamma}}}$$



Hierarchy of pattern control

- Adding a perturbation extrinsically (grooves)
- Control via an intrinsic but decoupled physical process
 - Crystal growth with a fixed underlying lattice
 - Nonequilibrium domain growth with chiral molecules



Non-equilibrium pattern of LC phase in phospholipid monolayer

Control, cnt'd

- Control via intrinsic process coupled to the macroscopic pattern-formation dynamics
 - Viscous fingering into nematic liquid crystals where anisotropy is created by flow-induced ordering



FIG. 1. Viscous fingering patterns in the liquid-crystal-8CB-air system. (a) $T = 46.0^\circ\text{C}$, (b) $T = 37.0^\circ\text{C}$, (c) $T = 34.0^\circ\text{C}$. The applied pressure is 6 kPa. The diameter of the envelope of the patterns is 4 cm.

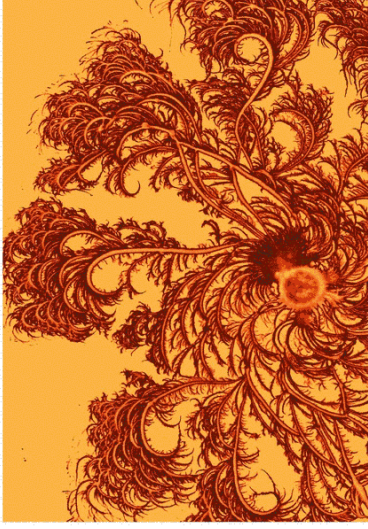
also, active
chiral

- Do the bacteria make use of this more powerful control strategy?

SELF-ORGANIZATION OF NEW BACTERIAL STRAIN (Ben-Jacob et al)



Branching morphotype



Chiral morphotype

Peainibacillus dendritiformis

Transition to faster growing morphology!

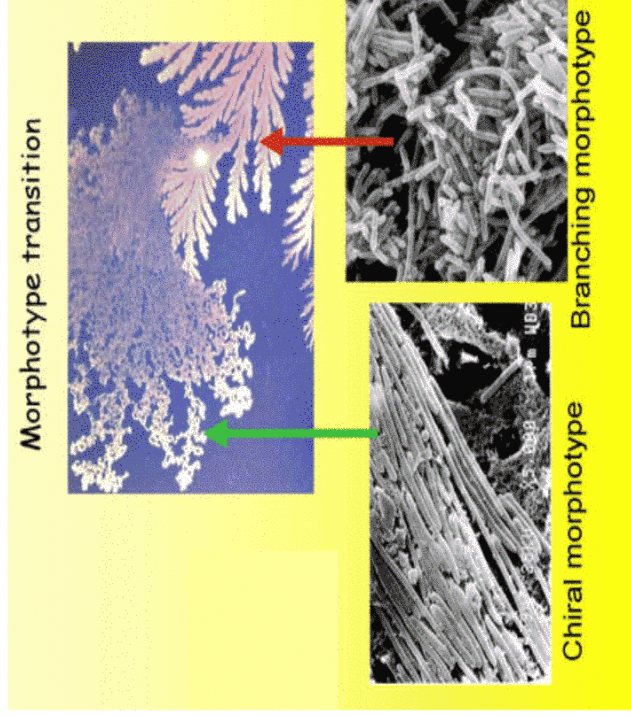


Heritable change

From branching to chiral

Chirality due to the flagellum

How do the bacteria become chiral?



One can build model of the chiral pattern using local orientational order.

Transition is inherited!

Recent evidence that there is some chemical factor which induces the transition

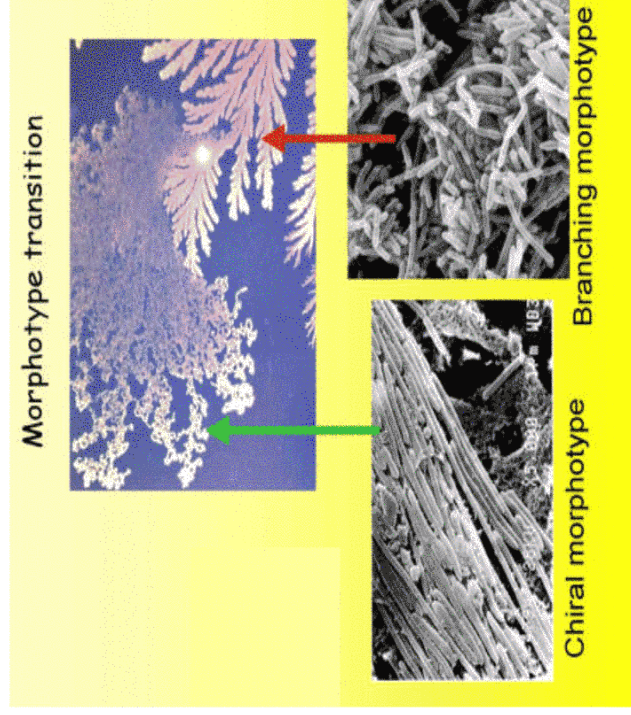
Direct coupling between colony pattern and gene expression dynamics!



Model requires introduction of an orientational degree of freedom coupled to the motion

$$\theta'_i = P(\theta_i, \Theta(r_i)) + c_0 + \eta$$

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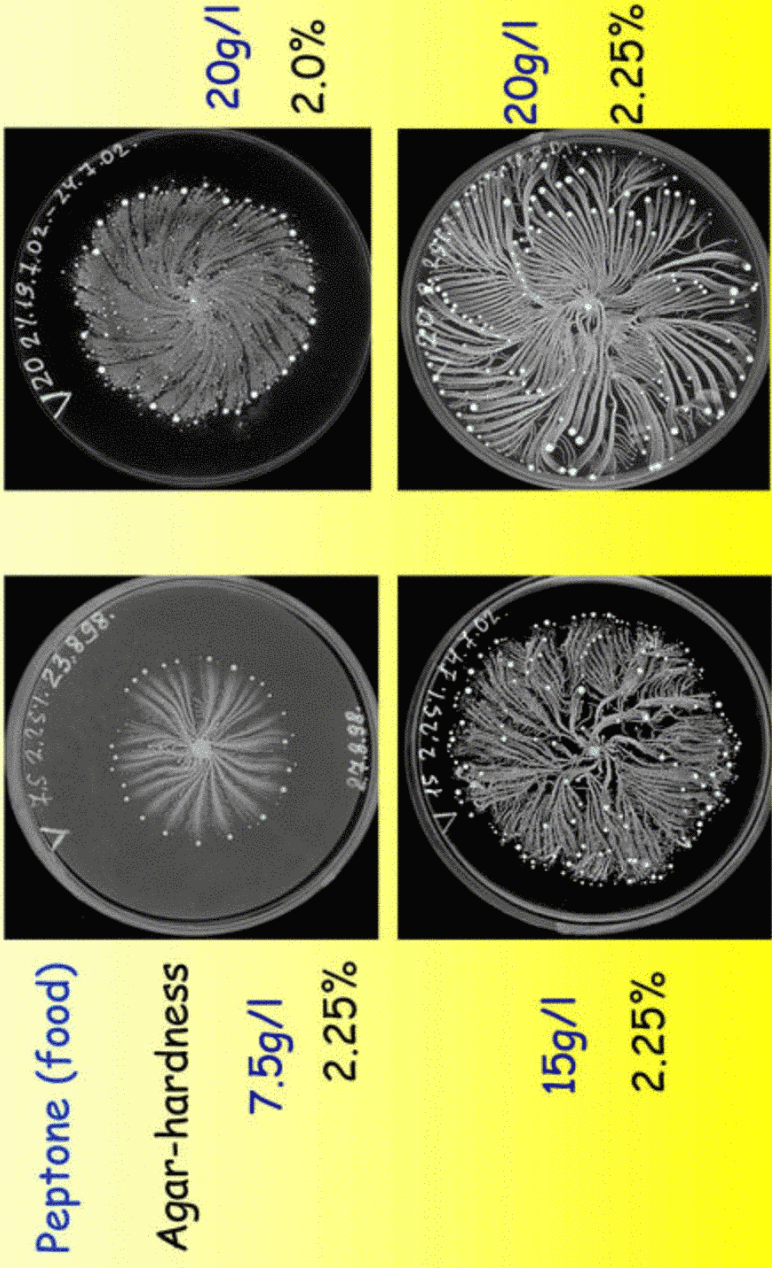
Could this have been predicted??

Patterns of patterns

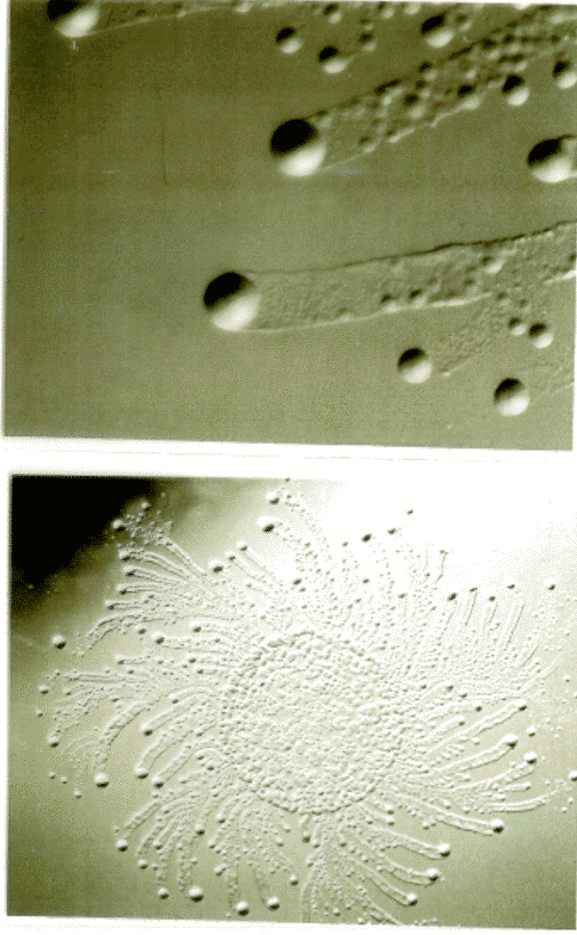
- We have seen how the bacteria use coupling between the single cell and colony levels to optimize spreading on the agar substrate
- Are there examples involving coupling between three levels of dynamical variables?
- Most intriguing possibility to date involves the use of chemotactic signaling to organize into mesoscale vortices

Three-level pattern formation hierarchy

The same *P. vortex* bacteria under different growth conditions

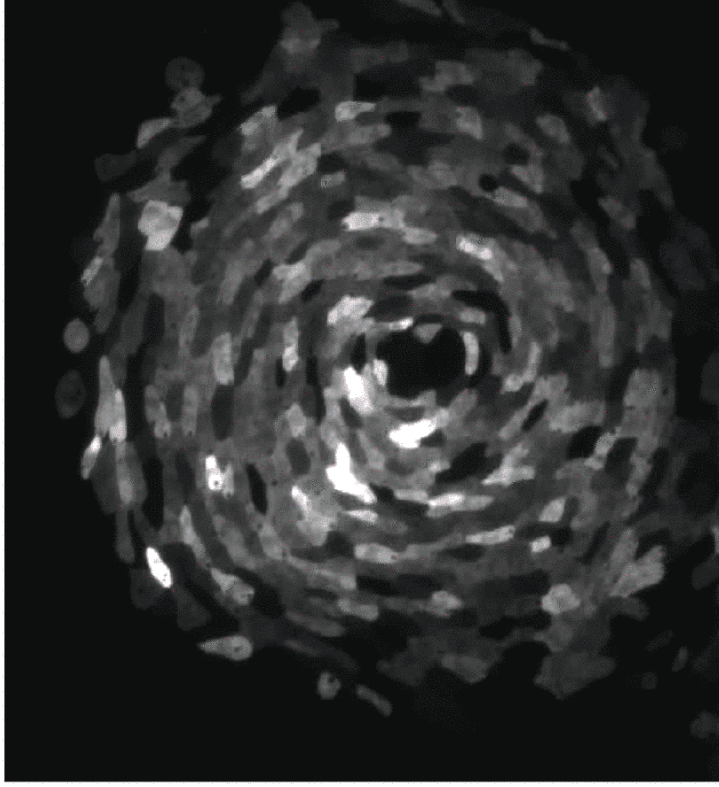


Vortex structures in bacteria



This represents a new pattern-formation schema

Self-organized motion - a new schema!



Bodenschatz lab
Cornell Univ

2d rotating mound of
Dicty amoebae

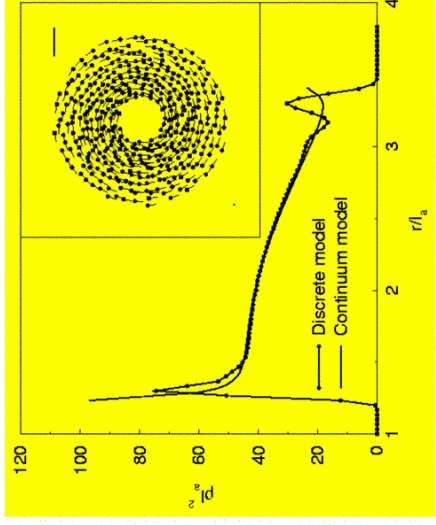
Simple point-particle flocking

$$m_i \partial_t \vec{v}_i = \alpha \hat{f}_i - \beta \vec{v}_i - \nabla U$$

$$U = U_a + U_r + U_{hc}$$

Repulsive and attractive forces are exponential functions of the inter-particle separation; alignment force \hat{f}_i is just the (exponentially) averaged velocity direction; Hard-core is typically a power-law, form is not crucial

Formation of the vortex state



Continuum equation:
$$\int_0^{2\pi} d\phi \int_0^{\infty} dr' \rho(r') U(r, r'; \phi) = D - \frac{\alpha^2}{\beta^2} \ln r$$

Understanding the vortex pattern

- Much more work is needed to fully understand the self-organized motion schema
 - Other examples such as Dicty, zooplankton
 - Related to work on traffic patterns
- Can the macroscopic pattern be understood solely in terms of the interactions of vortices of fixed structure/behavior or is there important feedback from the colony level?
- Is there feedback from the colony level to the behavior of a single bacterium or can these be treated as fixed constituents?
 - Answers are unknown at this time

Turtles all the way down

the scientist, after giving a lecture on astronomy, is approached by a little old lady who says that he's got it all wrong and that the world in fact rests on the back of a giant turtle. The scientist then asks the lady what the turtle is standing on, and she answers: on the back of a second, even larger turtle. But, asks the scientist, what does that turtle stand on? To which the lady triumphantly answers: "You're very clever, young man, but it's no use -- it's turtles all the way down!".

How many interacting levels can there be in a complex biological system? It seems almost unlimited!

- Sensors (such as those for bacterial communication and chemotaxis) are composed of interacting protein receptors which have multiple chemical states with transitions coupled to the received signals



- Genomes are not static memory but can have a variety of dynamical features which re-model the DNA (NB: strange case of bacterial genetic competence; TE's etc.)

Robustness vs. flexibility



Perhaps this type multi-level organization of the dynamics will help us understand how biological systems can be robust (insensitive to the inevitable noise in its components and surroundings) and yet at the same time exhibit flexible responses to real-world challenges

After all, the bacteria have flourished for billions of years; we need them to survive, but they can get along perfectly well without us.

Why do theory?

- Identification of pattern-forming schemata
- Detailed understanding of the generic behavior of this type of process
- Making sense of all these squishy notions
 - Experimental measures of robustness/flexibility?
 - What is complexity, anyway?
 - Bridging the gap between bacteria and non-living systems; is there a non-living 3T system like the bacterial vortex pattern?, e.g.

Summary/General Lessons

- Biological systems offer a new challenge - how to account for pattern purpose as it shapes the underlying dynamical interactions.
- Experiments should provide conditions under which systems can be induced to reveal new levels of feedback giving rise to new flexibility.
- A fully quantitative model is a practical impossibility - often, trying to put in more detail obscures the mechanisms being used by the system
- Schematic models can make reliable predictions as long as the biological system is not “pushed” to involve additional mechanisms not explicitly incorporated in the model.
- Schematic models are the only way to try to come to grips with the overall functional architecture of living systems

