Influence of topology on bacterial social interaction

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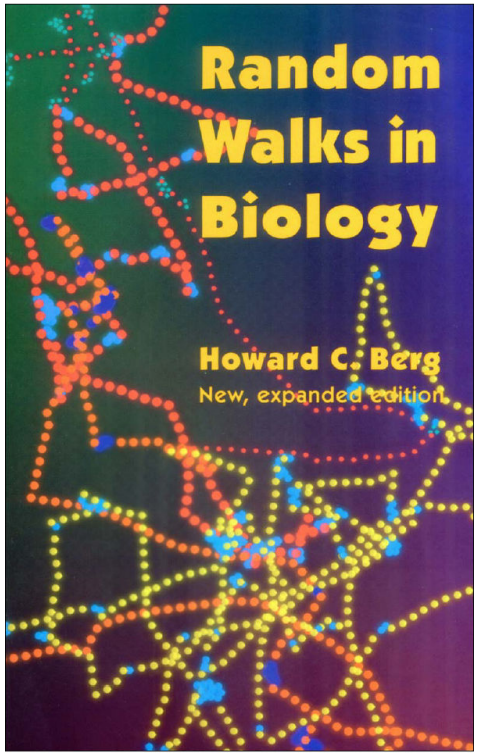
Outline

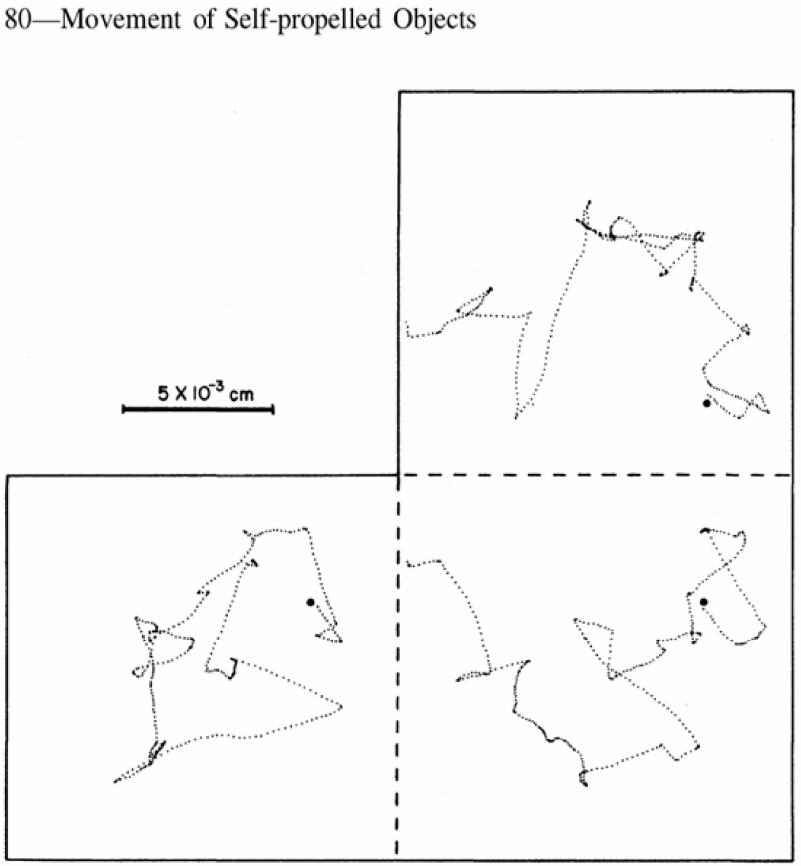
1. A set of experiments that explore the motion of bacteria in a topologically nontrivial environment.
2. Physical explanation of our experiments.
3. Biological implications of our findings.

S. Park, P. M. Wolanin, E. A. Yuzbashyan , P. Silberzan, J. B. Stock, R. H. Austin, *Science*, 301, 188, 2003; *PNAS*, 100, 13910, (2003).

How are bacteria different from Brownian particles?

A bacterium in a homogeneous medium behaves as a Brownian particle.





A wild-type bacterium E. coli executing a random walk.

Is many different?

1. Many Brownian particles or a gas.

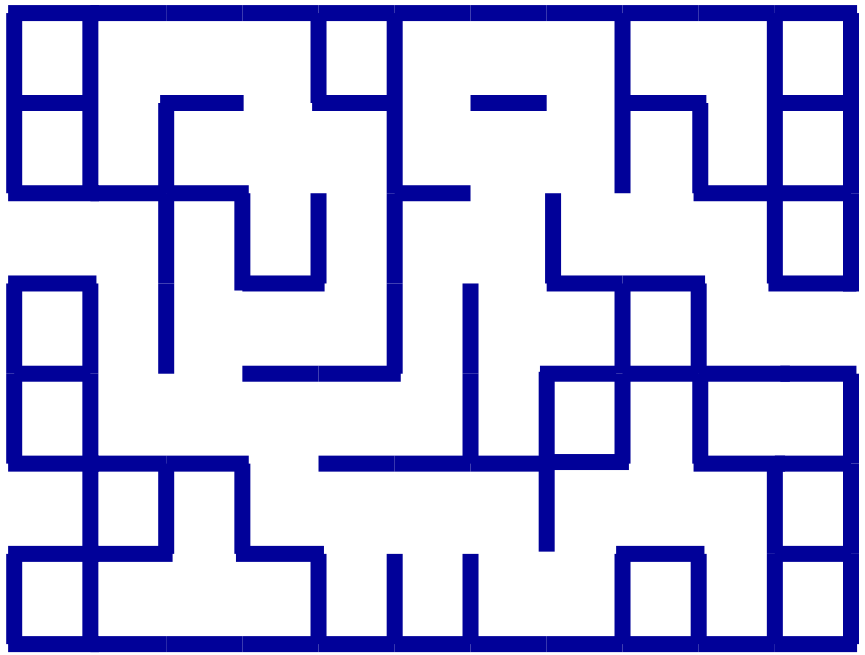
2. Many bacteria.

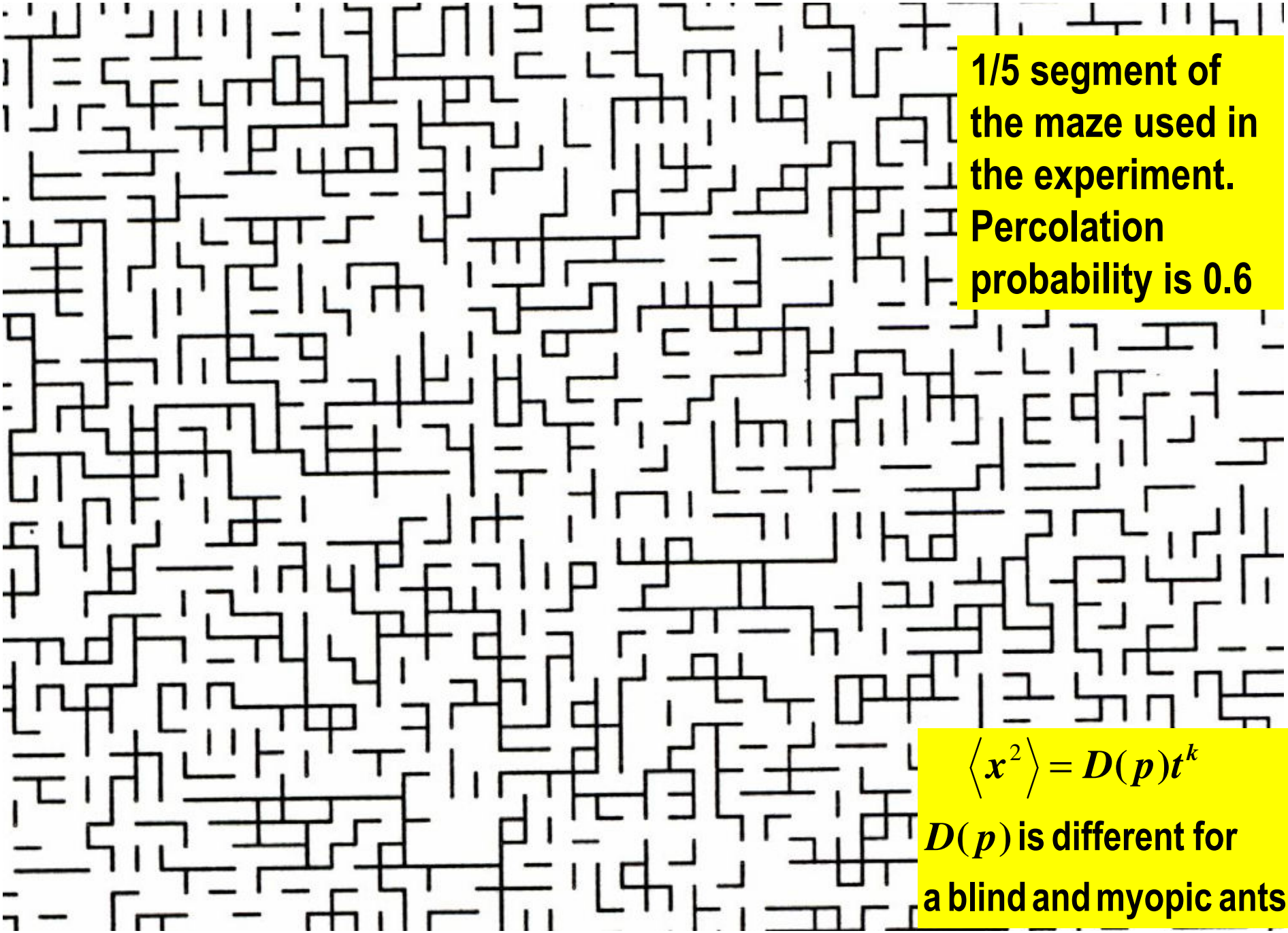
Why would a CM theorist do a biophysics experiment?

1. “Condensed bacterial matter” can be viewed as a strongly correlated system with unusual interactions. These interactions can produce new physical effects.

2. Experimental project at Princeton. Labyrinth for bacteria.

Bob Austin



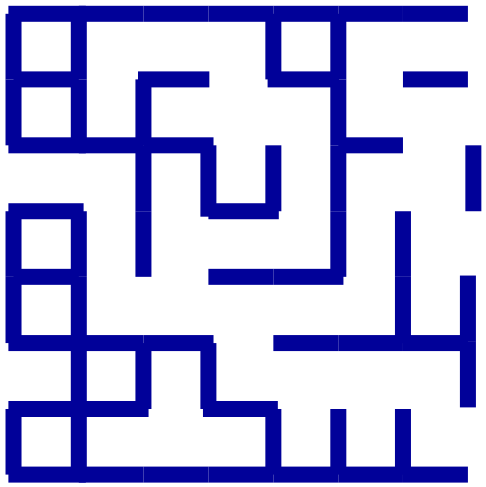


1/5 segment of the maze used in the experiment. Percolation probability is 0.6

$$\left\langle x^2 \right\rangle = D(p)t^k$$

$D(p)$ is different for a blind and myopic ants.

Fabrication process



Photolithography

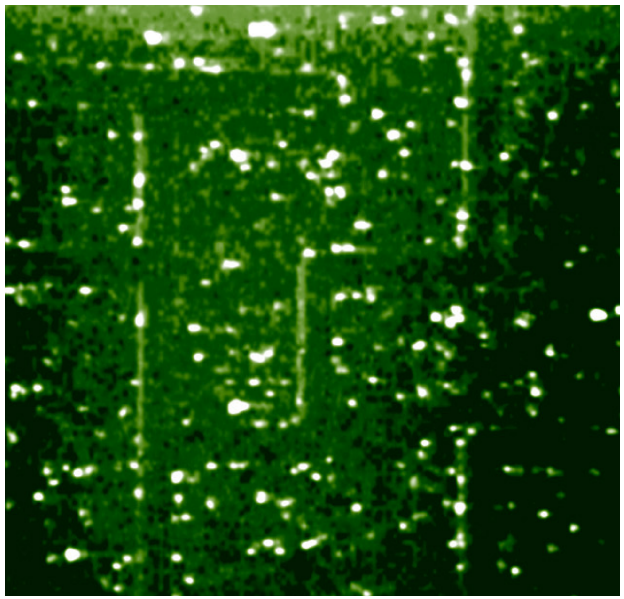
PDMS

2 μm

E. coli

20 x 20 x 200 μm

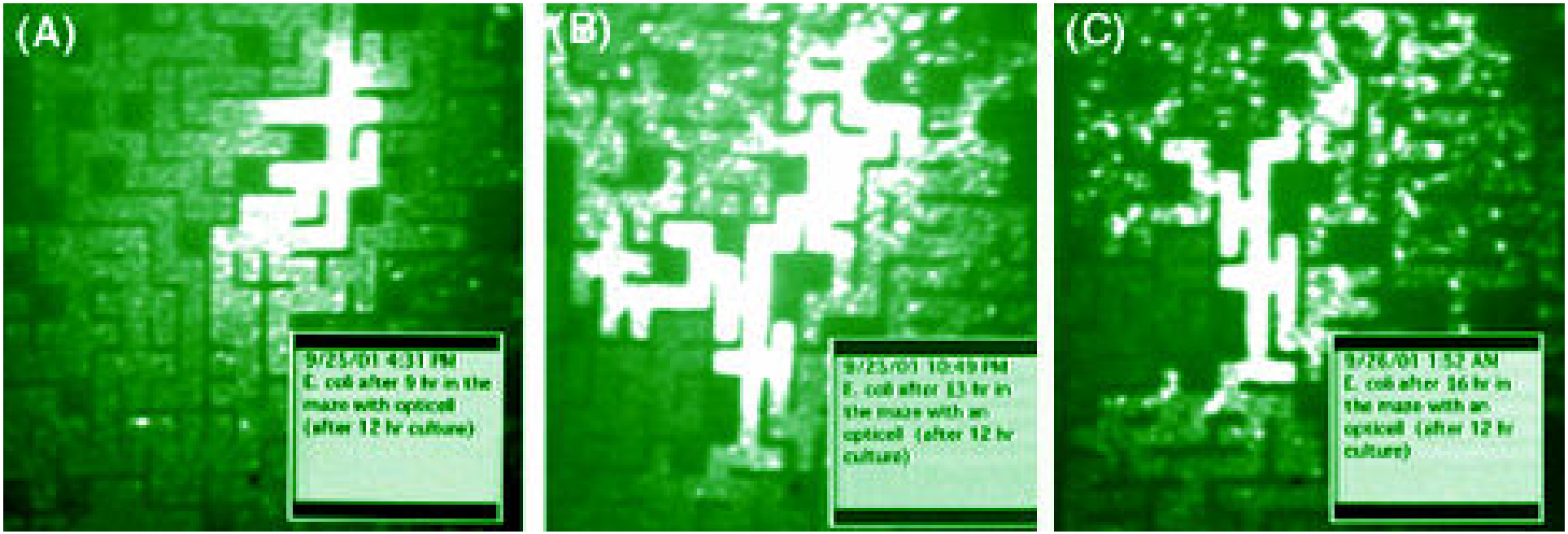
Glass or OptiCell

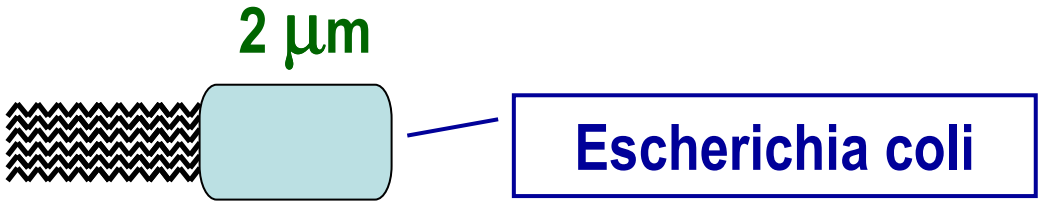


GFP-expressing E.coli in a maze

Congregation of E. coli

Initially uniform distribution of bacteria becomes inhomogeneous with time as bacteria cluster into certain spots in the maze.





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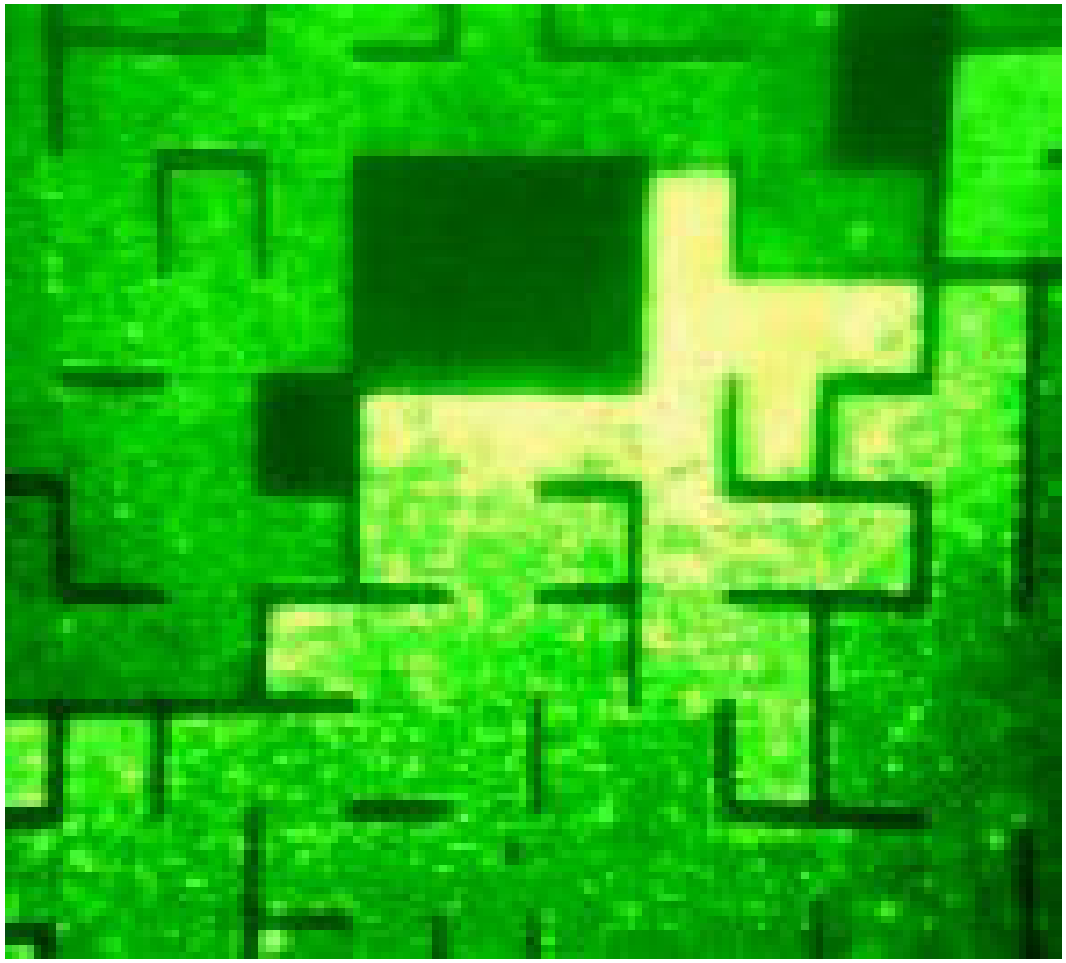
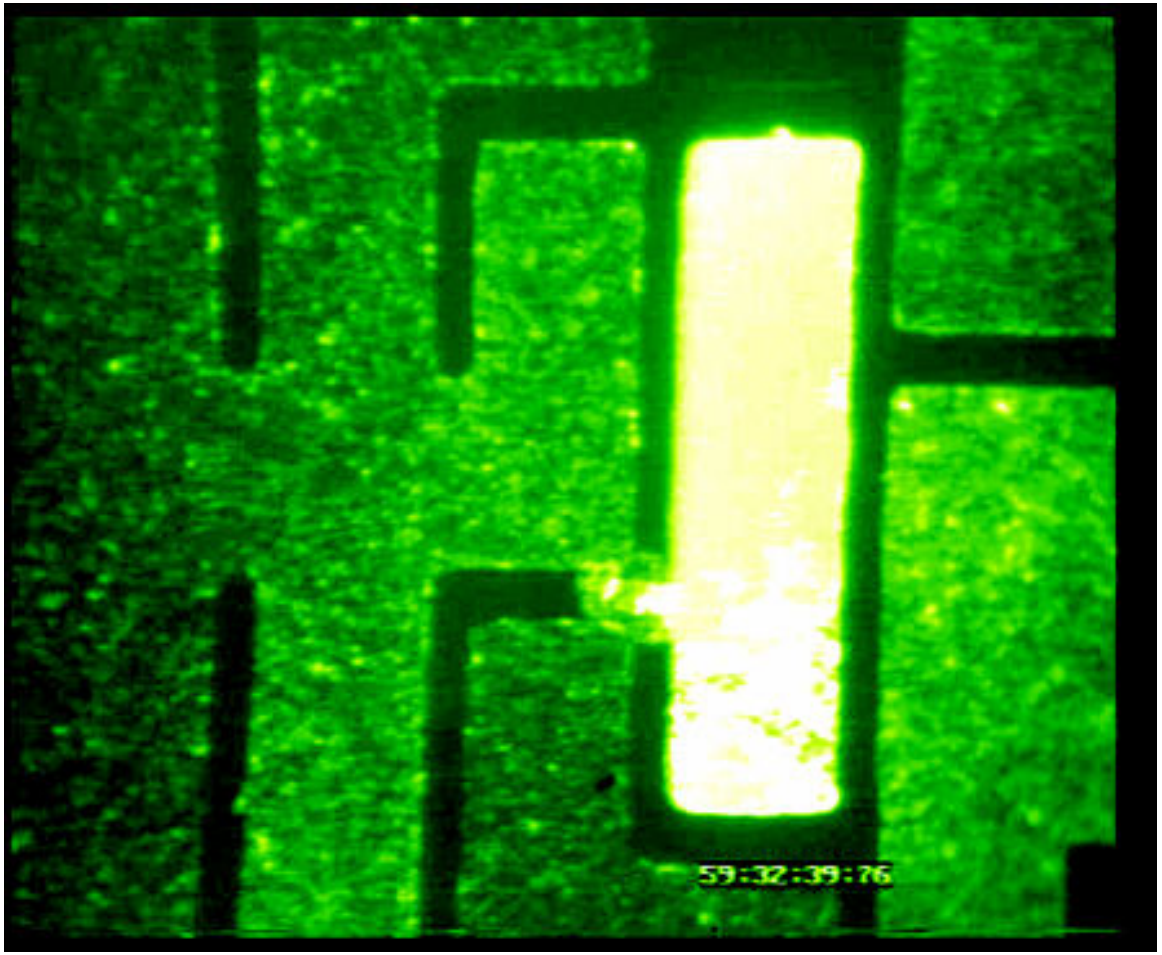


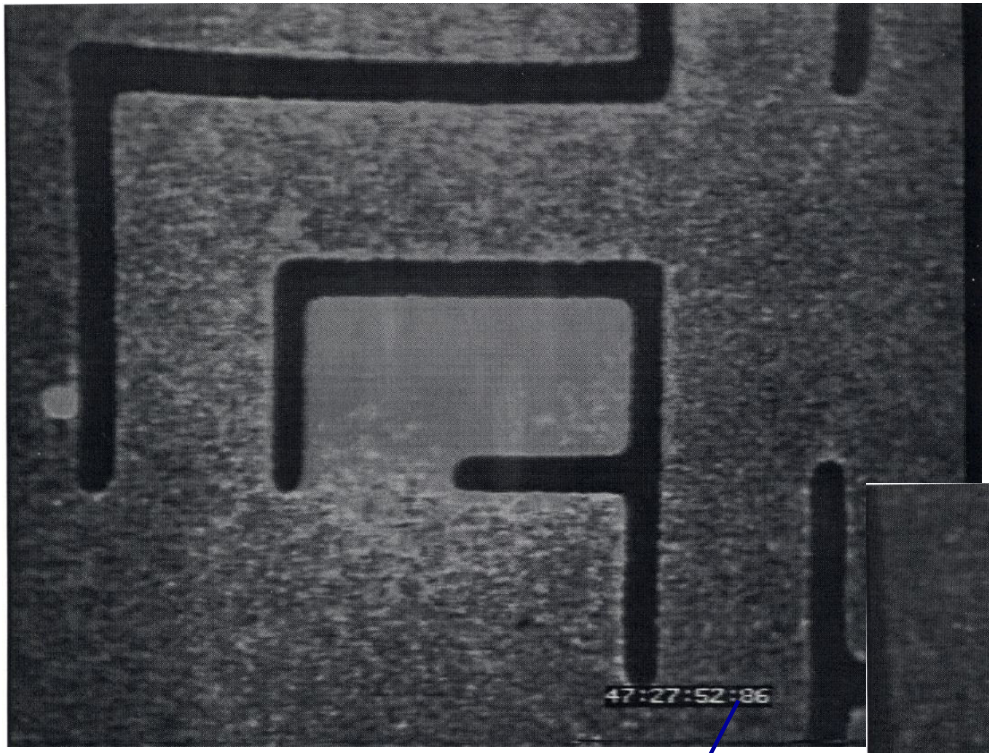
Wild-type E.coli (RP437) in a random maze. Dynamical accumulation in a “dead-end” part of the maze 2 hours after loading.

Congregation of E. coli

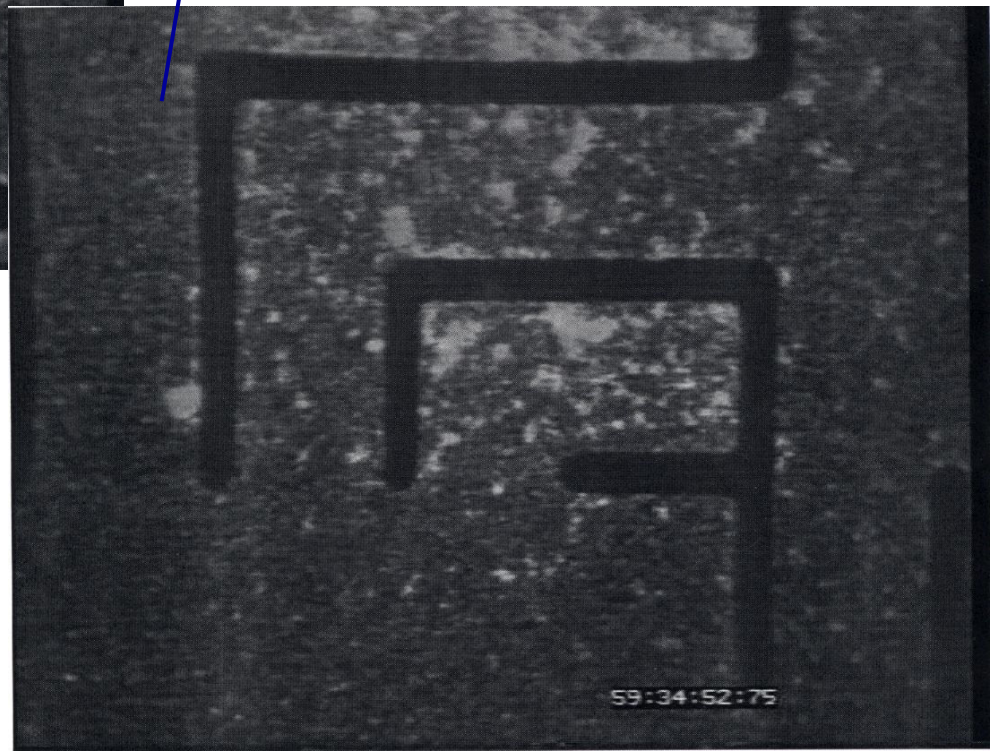


E. Coli are actively swimming into a rectangle through a small opening created by a sealing defect. The density of cells inside is about 10 times greater.

“Crystallization” of the E. coli “liquid”.

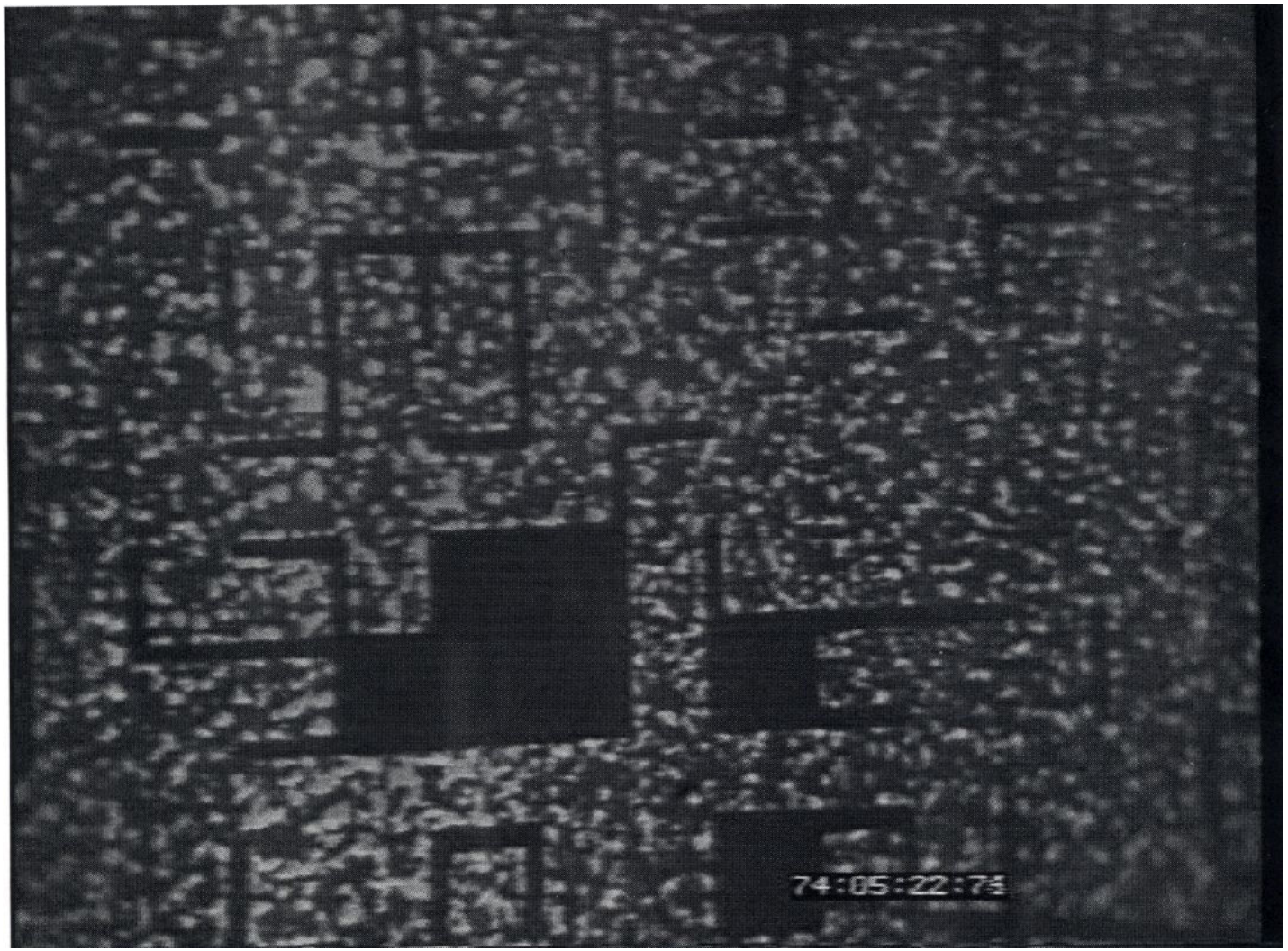


10 hours later bacterial density further collapses into ~20 μm clusters

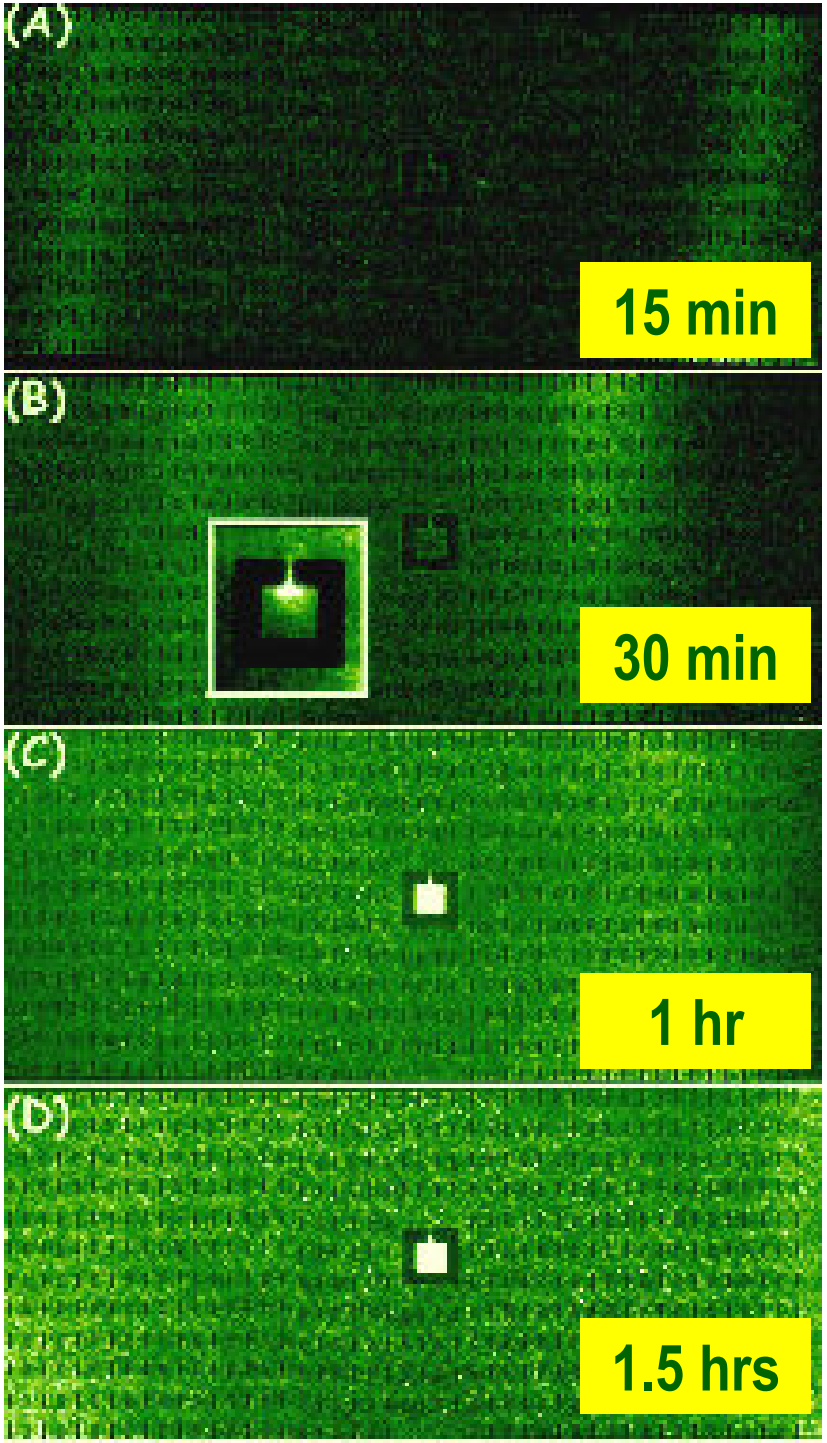


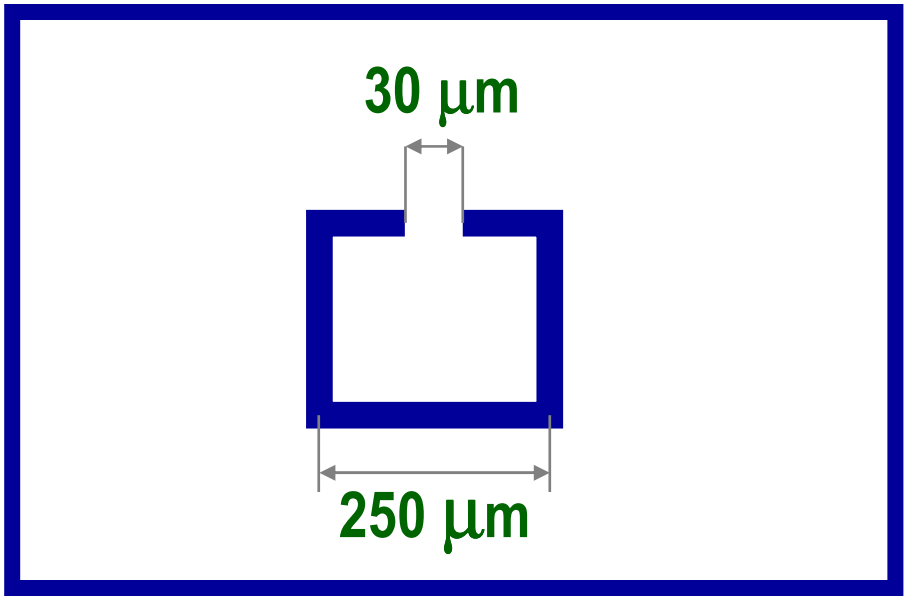
Congregation of E. coli into a confined area several hours after loading.

“Crystallization” of the E. coli “liquid”.



Eventually the clustering occurs throughout the maze. The cells are still motile.

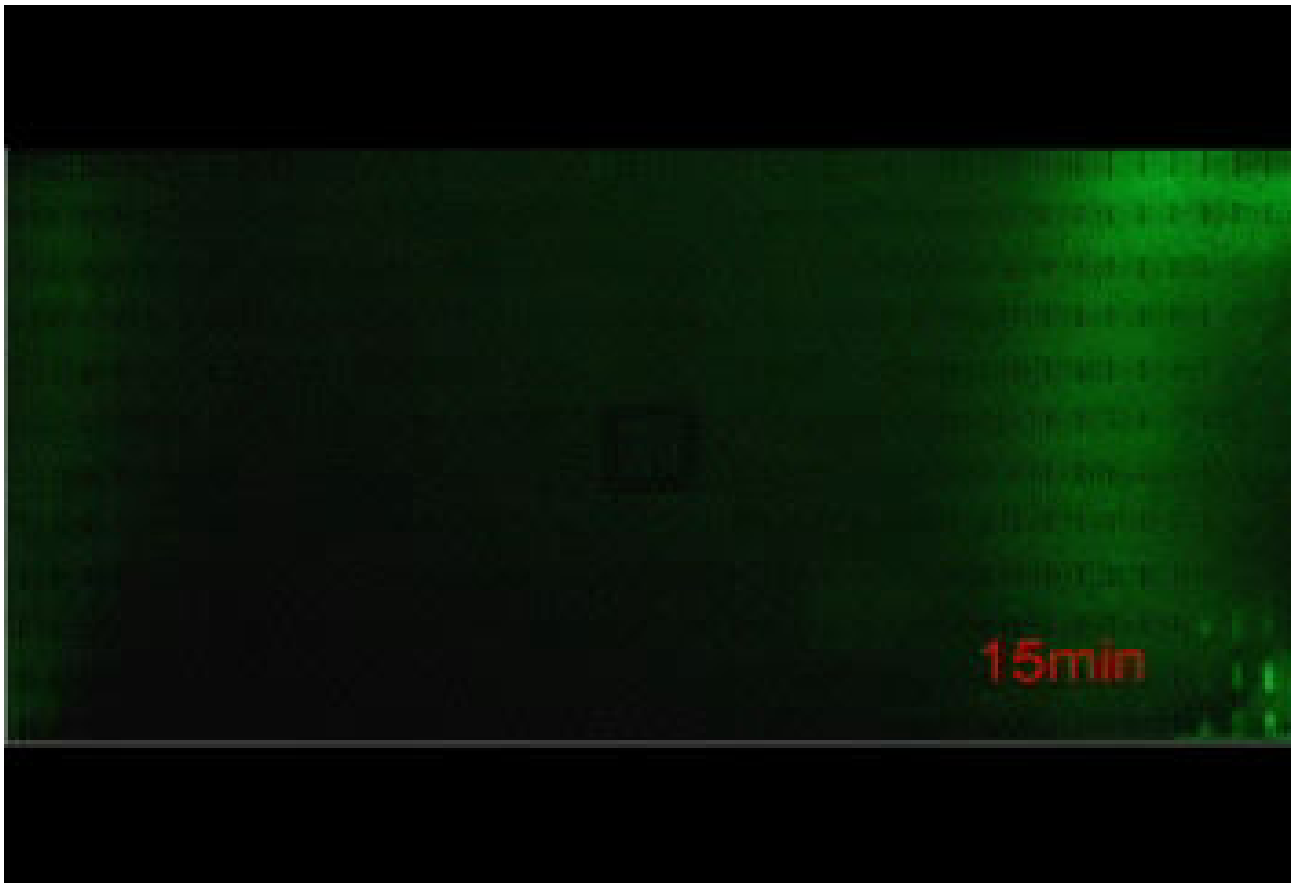




Note long wavelength bacterial density waves in the larger rectangle.

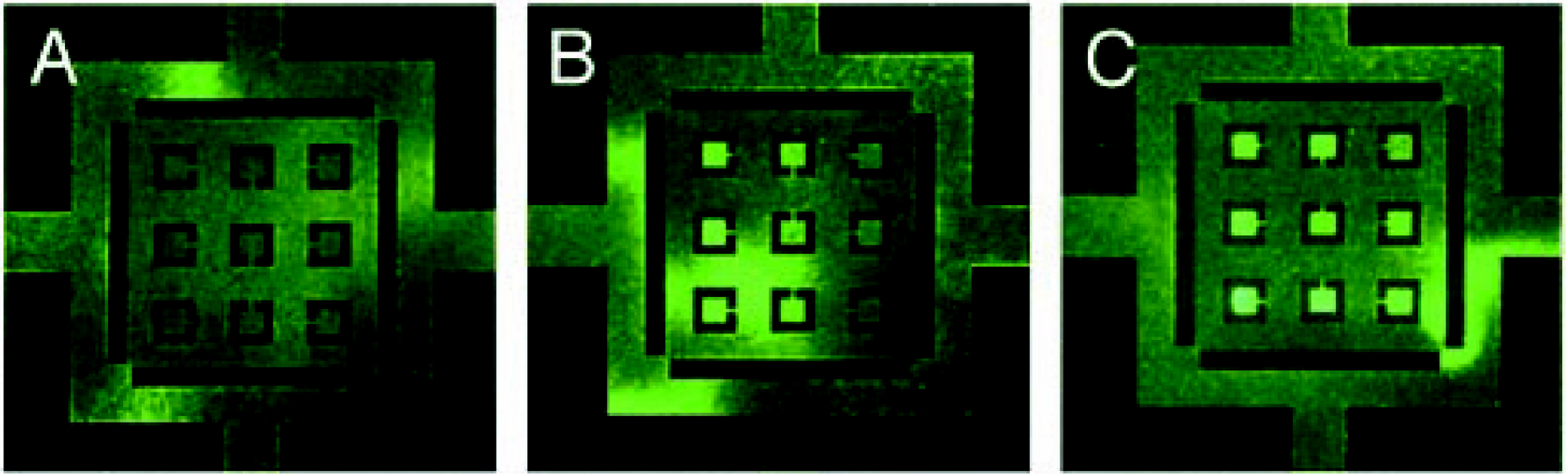
Collapse of wild-type E. coli in LB media into a center square.

Congregation of E. coli

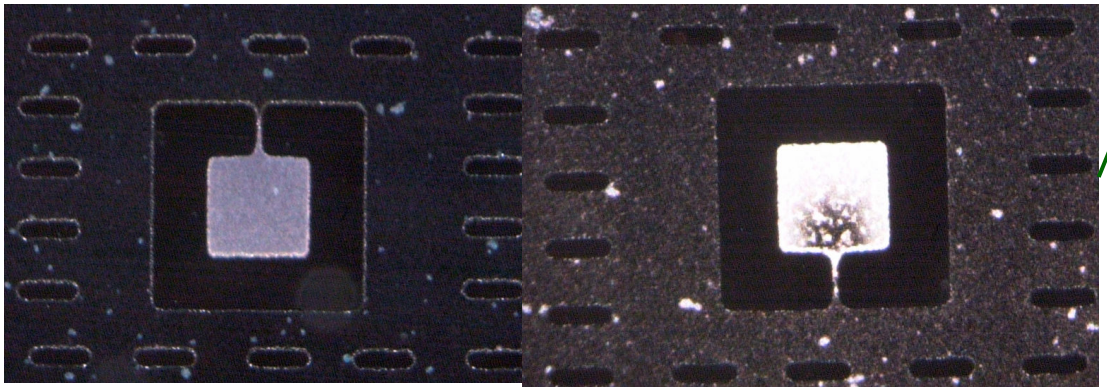


E. coli in M9 minimal media as the accumulate into a central enclosure. After three hours the density of cells is more than seven times greater inside than outside.

Congregation of E. coli and V. harveyi



Congregation into an array of squares. Each frame is separated by 1 hour.



Congregation is also seen in other bacteria such as Vibrio harveyi.

Minimal model of chemotaxis

Congregation must be produced by some sort of attractive interaction between bacteria.

Chemotaxis – motion of bacteria in response to gradients of amino acids secreted by other bacteria.

$$\frac{\partial \rho}{\partial t} + \nabla J = a\rho$$

$$J = k\rho\nabla c - D_b \nabla \rho$$

Chemotaxis

Diffusion.

J – current of bacteria, ρ – bacterial density, c – attractant concentration, a – growth rate, D_b – bacterial diffusion coefficient.

Need another equation for the attractant concentration c.

$$\frac{\partial c}{\partial t} = D_c \nabla^2 c + \alpha\rho$$

α – attractant production rate,

D_c – attractant diffusion coefficient.

Minimal model of chemotaxis – Keller-Segel equations

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$$\frac{\partial \rho}{\partial t} = D_b \nabla^2 \rho - \nabla(k \rho \nabla c) + a\rho$$

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Chemotactic collapse into confined spaces

$$J = J_{diff} + J_{chem}$$

$$J_{diff} = -D_b \nabla \rho$$

$$J_{chem} = k \rho \nabla c$$

J – current of bacteria,

ρ – bacterial density,

c – attractant concentration.

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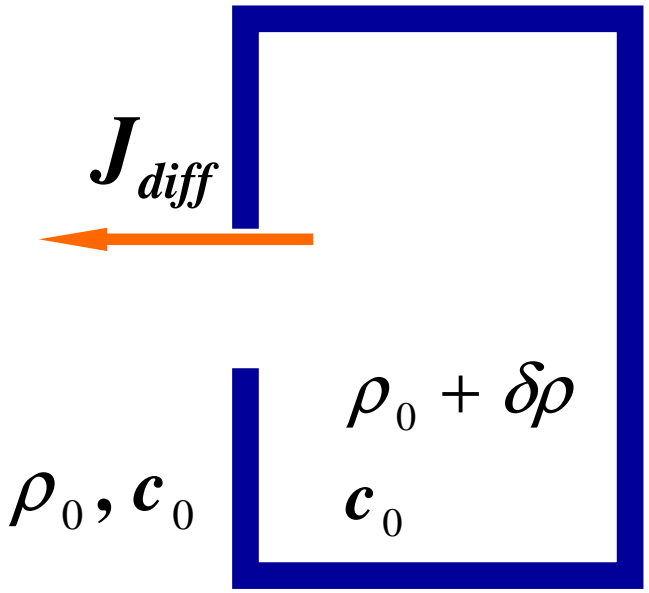

Density fluctuation

Normally a density fluctuation would be washed out by diffusion.

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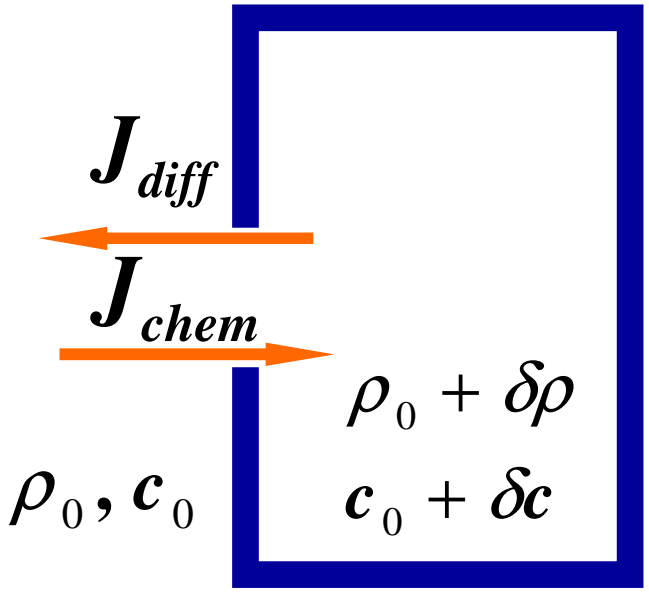

Attractant gradient

However, since there are more cells inside, they produce an excess of attractant and set up an attractant gradient.

Chemotactic collapse into confined spaces

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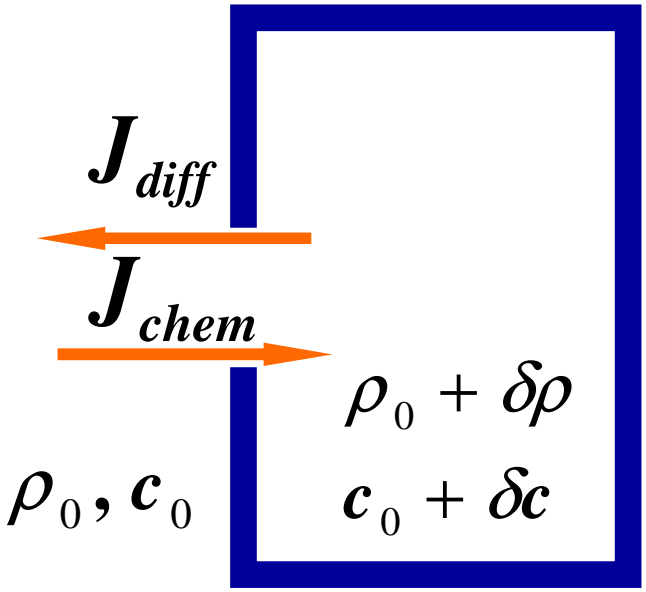
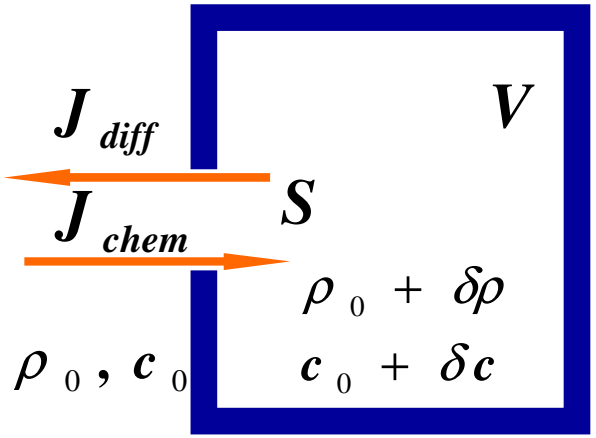


If the time to wash out the density fluctuation is large enough, chemotaxis overwhelms the diffusion and an irreversible collapse occurs.

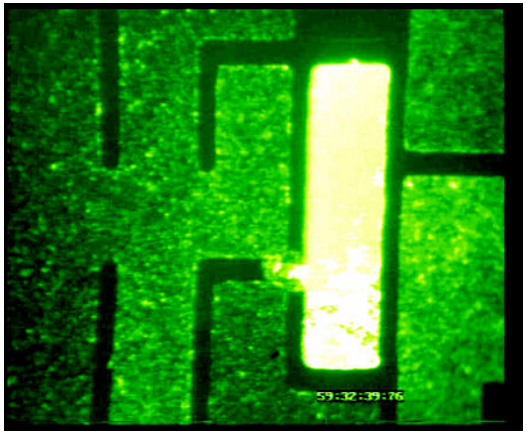
Competition between chemotaxis and diffusion.

Chemotactic collapse into confined spaces



J – current of bacteria,
ρ – bacterial density,
c – attractant concentration.

$$J = k\rho\nabla c - D_b\nabla\rho$$



$$V\frac{\partial(\delta\rho)}{\partial t} = -D_b S\frac{\delta\rho}{l_b} + kS\rho_0\frac{\delta c}{l_c}$$

$$V\frac{\partial(\delta c)}{\partial t} = -D_c S\frac{\delta c}{l_c} + \alpha V(\delta\rho)$$

l_b & l_c – characteristic length of gradient decays,
S – opening cross-section, V – volume.

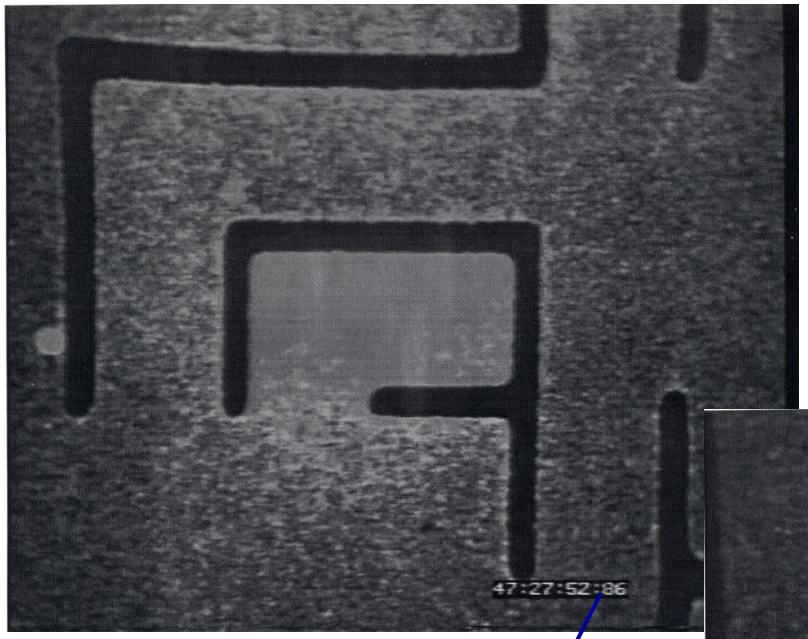
Above the critical density an irreversible collapse begins.

$$\rho_c = \left(\frac{D_b D_c}{k\alpha l_b}\right)\frac{S}{V}$$

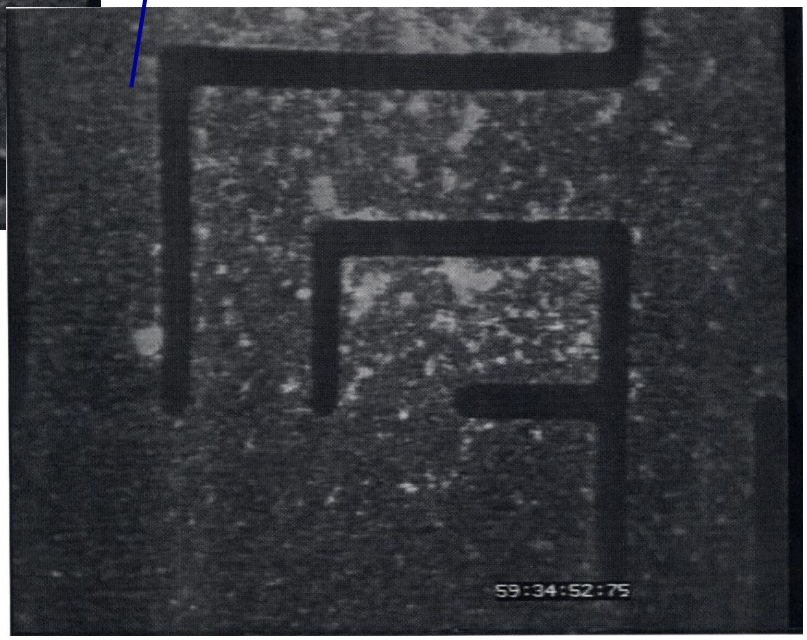
The collapse first occurs in relatively large volumes with small openings.

“Crystallization” of E. coli “liquid”

With the increase of density the instability eventually occurs even in the “open” spaces.

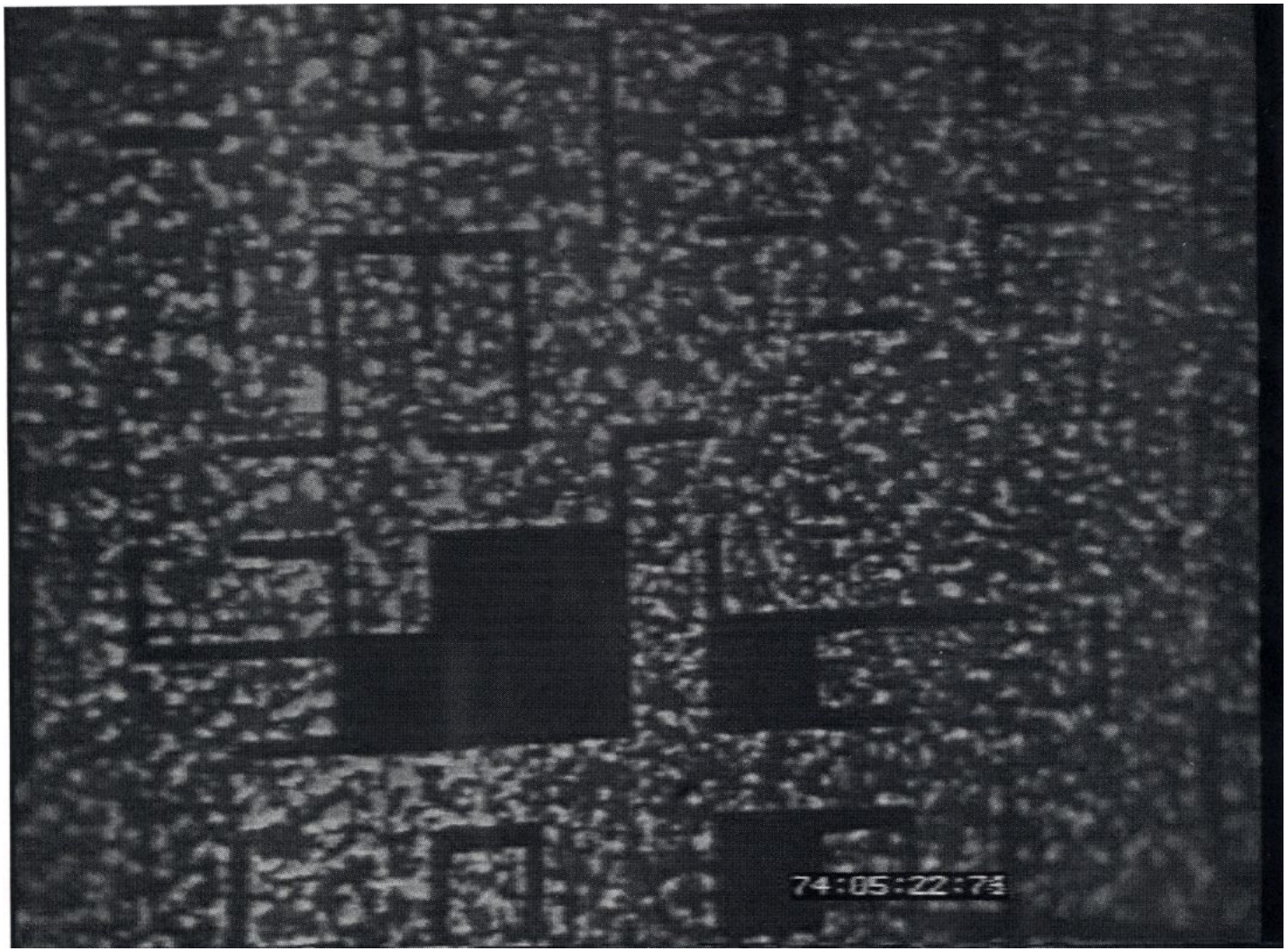


10 hours later bacterial density further collapses into ~20 μm clusters



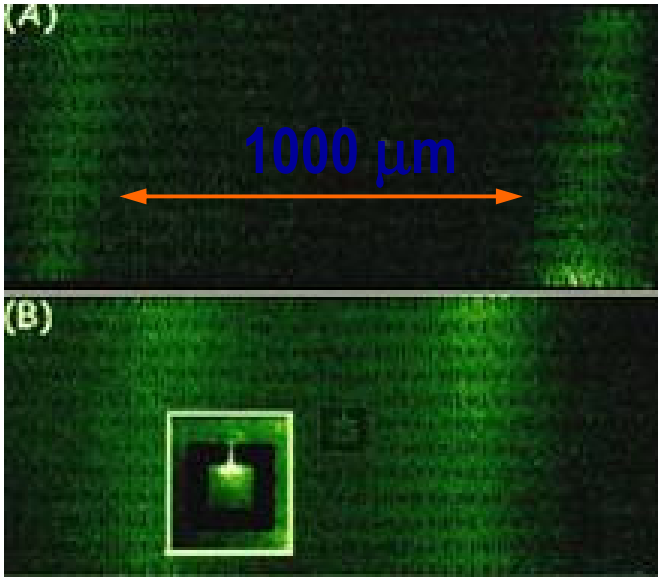
Congregation of E. coli into a confined area .

“Crystallization” of the E. coli “liquid”.



Eventually the clustering occurs throughout the maze. The cells are still motile.

Large scale instability



$$\frac{\partial \rho}{\partial t} = -D_b \nabla^2 \rho + \nabla(k\rho \nabla c) + a\rho$$

$$\frac{\partial c}{\partial t} = D_c \nabla^2 c + \alpha\rho$$

ρ – bacteria,
c – attractant.

Stability of a uniform state

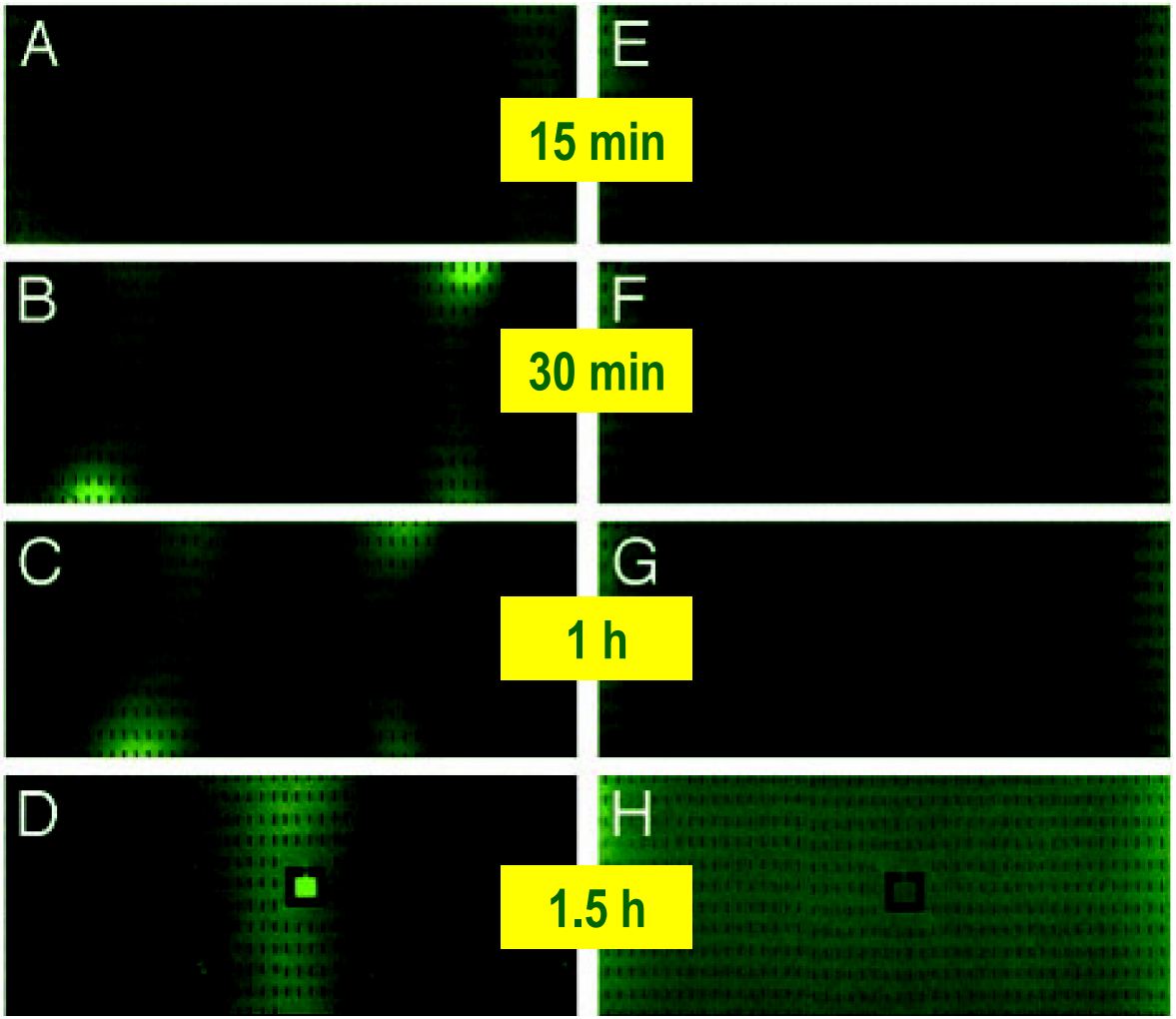
$$\rho = \rho_0 + \varepsilon e^{\omega t + iqx} \qquad \rho = \rho_0 + \delta e^{\omega t + iqx}$$

Fastest growing mode $\lambda^* = \dfrac{2\pi(D_c + D_b)}{\sqrt{\alpha k \rho_0}} \sim 500\ \mu\text{m}$

All modes with $\lambda > \lambda^*/2$ grow.

M. Brenner, L. S. Levitov & E. O. Burdene, *Biophys. J.* 74: 1677 (1998)

Nonchemotactic cells do not congregate

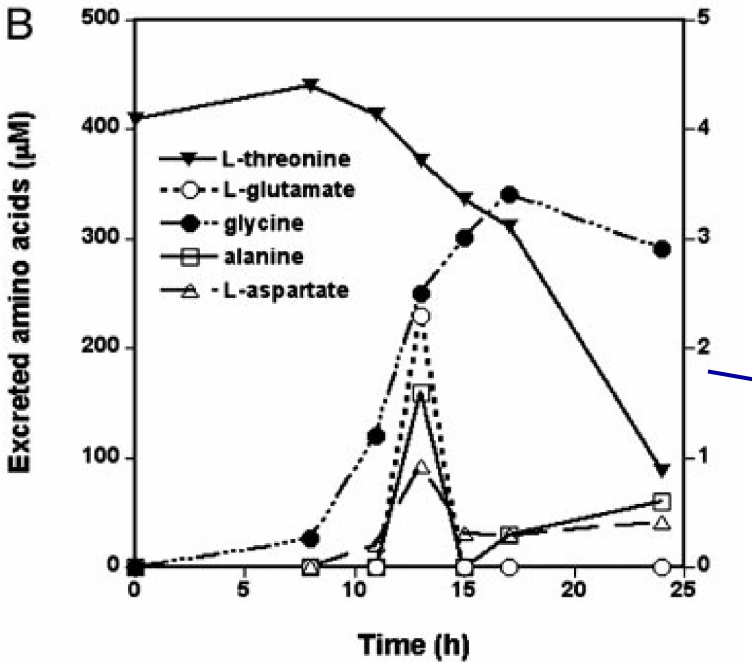


Comparison between chemotactic and nonchemotactic strains.

Nonchemotactic cells do not congregate

Mutant strains that are motile but deficient in chemotaxis (e.g. PS2002) do not congregate.

Congregation	No congregation
RP437 (wild type), RP437 + L-aspartate, RP2361 (Δtar), , KX1485 (ΔluxS), UU117 (Δaer).	RP437 + L-serine, RP5700 (Δtsr), HCB317 (Δtsr), PS2002 (ΔcheA-Z).

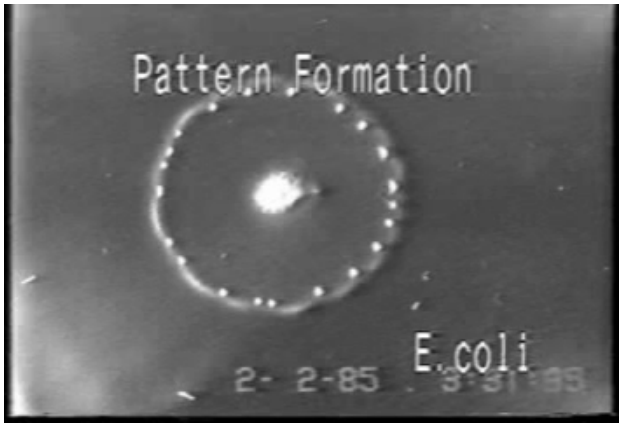


The serine receptor, Tsr, is involved.

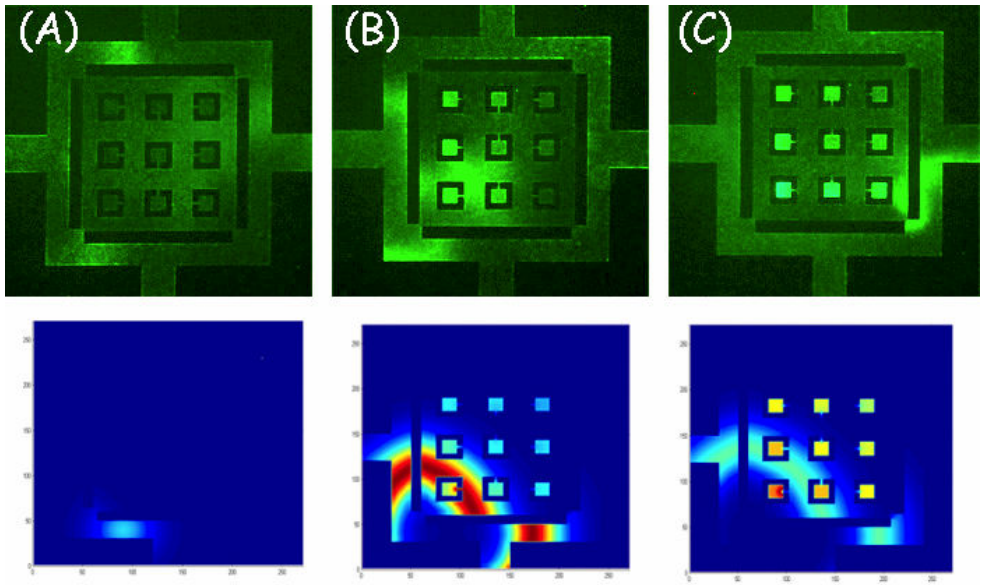
The signaling amino acid (chemoattractant) is glycine.

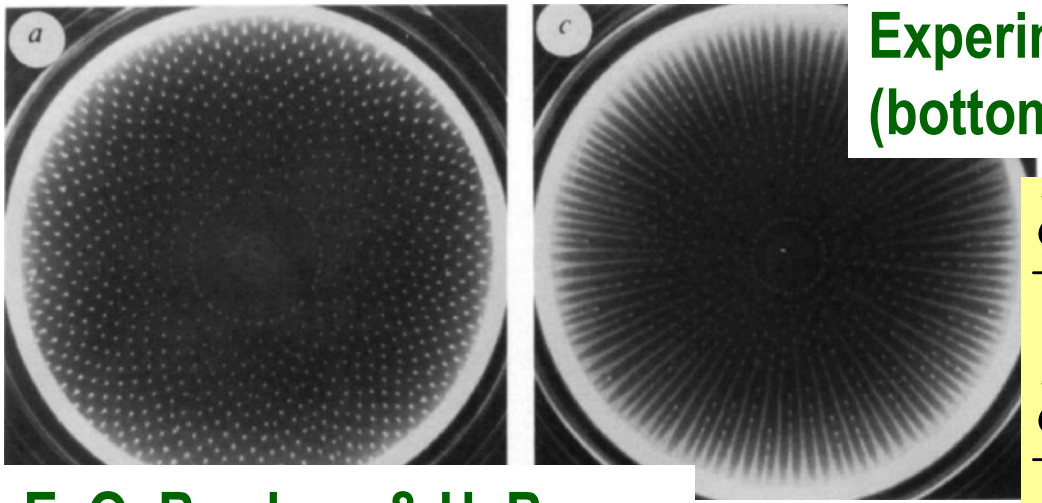
Extracellular amino acid concentration as a function of time for RP437 cells grown in M9 glycerol medium

Nonlinear phenomena in chemotaxis



From H. Berg's website.





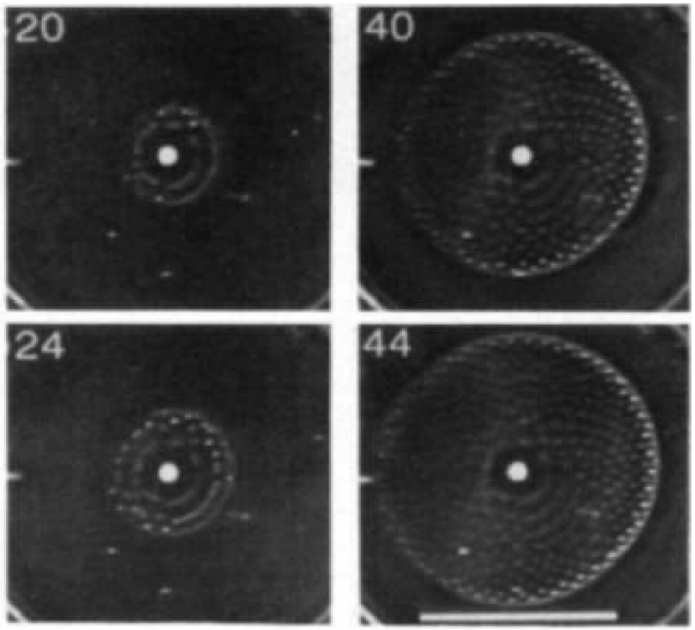
E. O. Burdene & H. Berg, *Nature*, 349, 630 (1991)

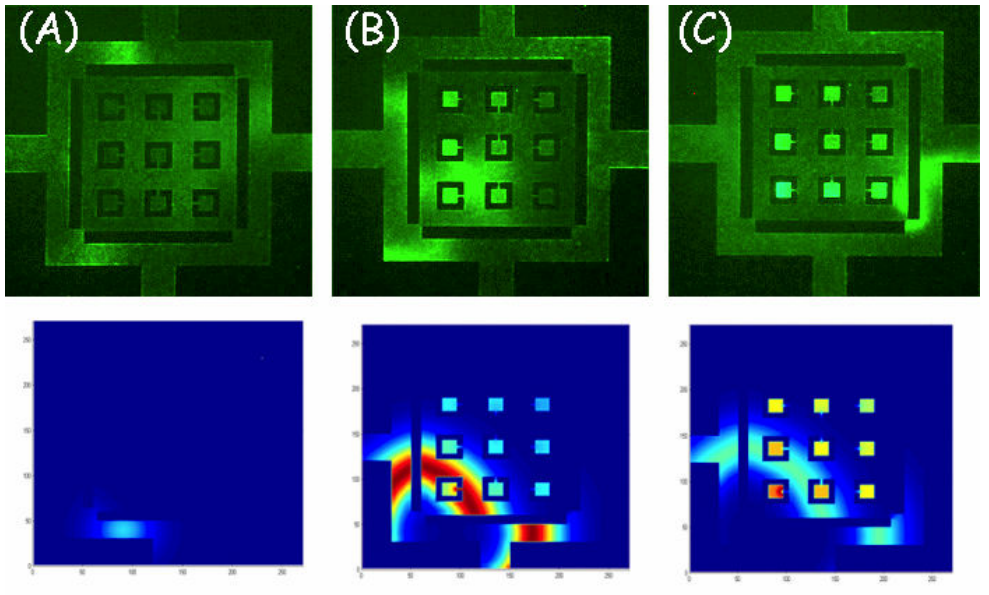
Experiment (top) versus simulation (bottom) based on KS equations.

$$\frac{\partial \rho}{\partial t} = -D_b \nabla^2 \rho + \nabla(k\rho \nabla c) + a\rho$$

$$\frac{\partial c}{\partial t} = D_c \nabla^2 c + \alpha\rho$$

Nonlinear phenomena in chemotaxis





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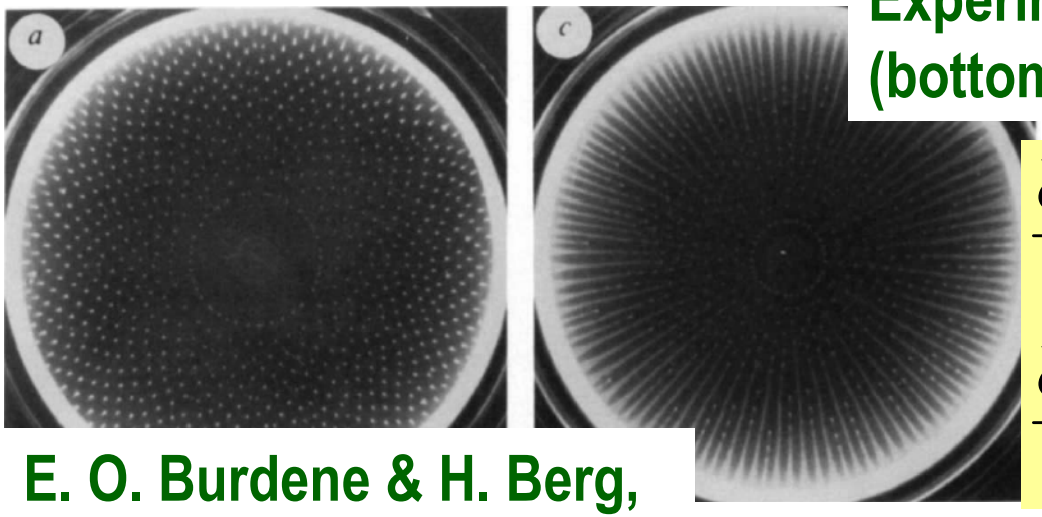

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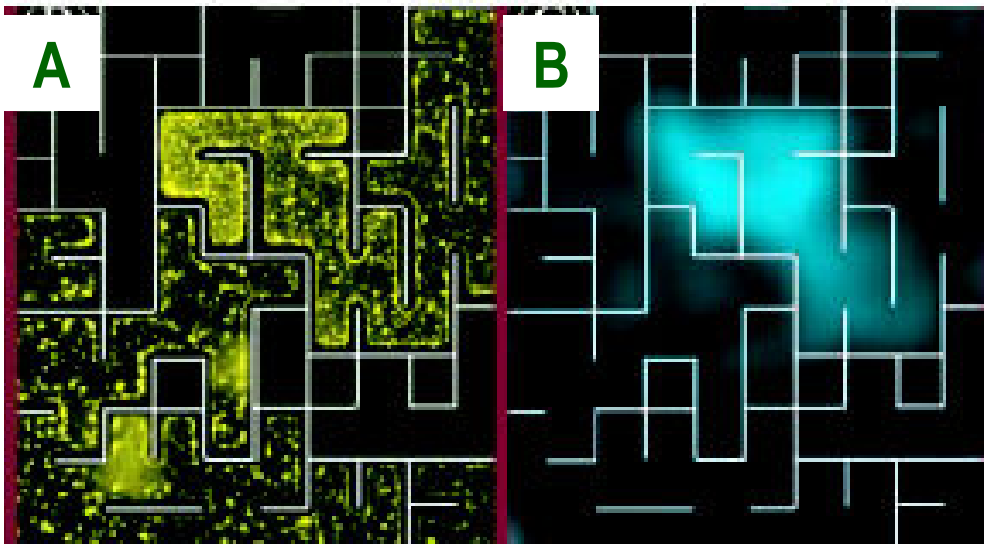
Quorum sensing

Upon reaching a critical population density ("quorum") bacteria can produce a response such as luminescence, virulence or biofilm formation.



A. V. Harveyi after 8 hrs in a maze.

B. B. Photon-counting image of the intrinsic luminescence.

Therefore, chemotaxis provides an important mechanism for establishing a quorum.

The study "brings up a lot of intriguing questions," says L. L. Kiessling … For example, "drugs that inhibit chemotaxis might inhibit biofilm formation," ...

CHEMICAL & Engineering News

Summary

1. Given appropriate surface topologies, bacteria can dynamically confine themselves to highly enclosed spaces.
2. The physical mechanism responsible for this is chemotaxis that leads to an effective attraction between bacteria.
3. This behavior can be explained theoretically by a minimal, Keller-Segal, model of chemotaxis.
4. Chemotaxis provides an important mechanism for achieving high local cell densities required for quorum-dependent interactions.

S. Park, P. M. Wolanin, E. A. Yuzbashyan , P. Silberzan, J. B. Stock, R. H. Austin, *Science*, 301, 188, 2003; *PNAS*, 100, 13910, (2003).