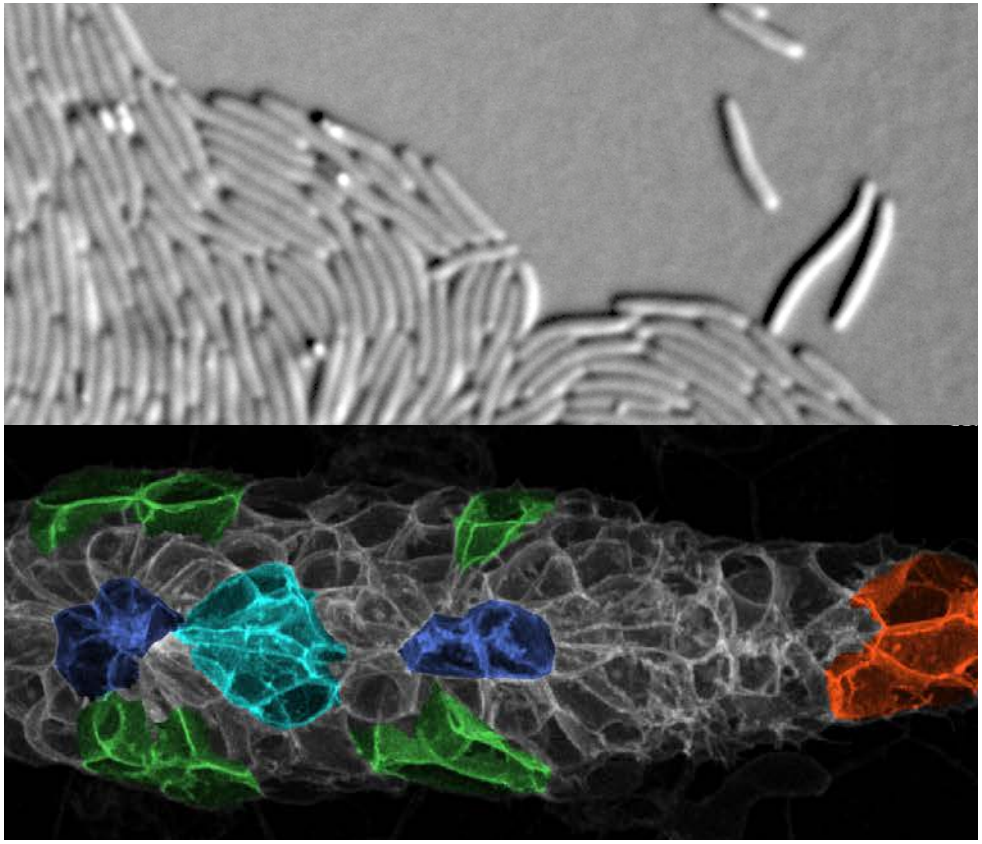
Collective Cellular Motility



Course 1: Introduction

Thomas Lecuit

chaire: Dynamiques du vivant

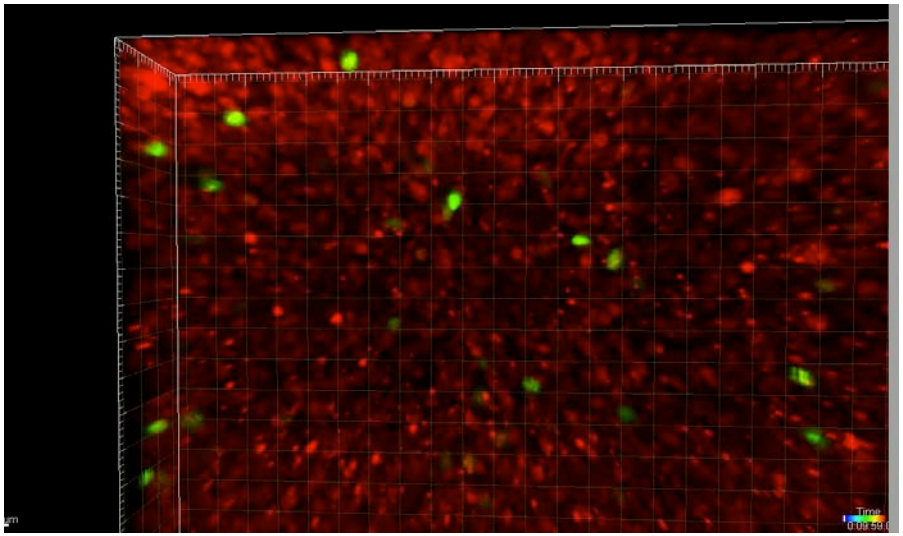
COLLÈGE DE FRANCE 1530

Individual and collective dynamics

- Cells exhibit individual motility (Courses 2021)
- Yet, in vivo, cells are part of tissues:

Depending of cell connectedness (eg. adhesion, cortical contractility) tissues exhibit liquid/fluid or solid (elastic) states. (see course#1–17 Oct 2017).

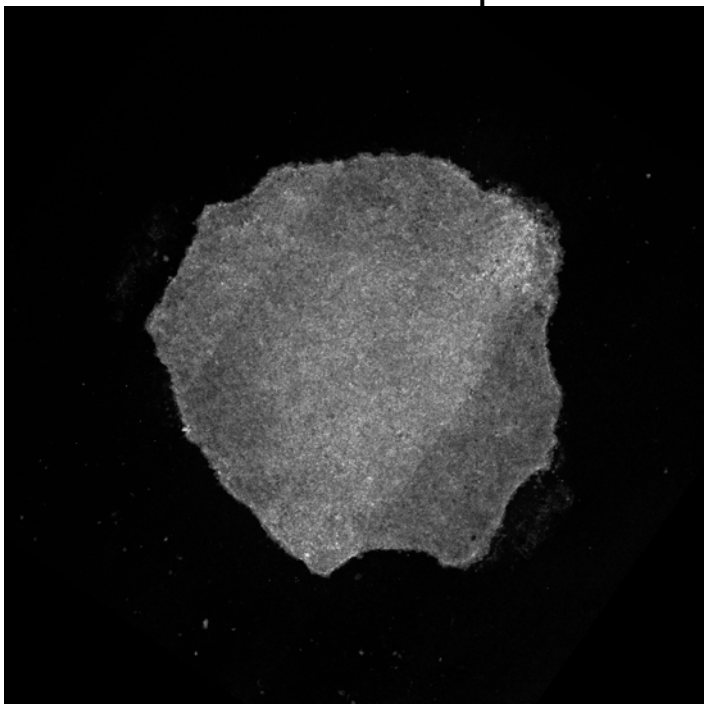
« Gaz-like », fluid state: 3D mesenchyme



Chick embryo axis elongation (Pre Somitic Mesoderm)

Bénazéraf B. et al, Pourquie O. *Nature.* 466:248 (2010)

Viscoelastic fluid: 2D epithelium



Quail embryo gastrulation (Epiblast)

Firmino J. et al, and Gros J. *Dev. Cell.* 36:249 (2016)

Saadaoui M, et al., Corson F, Gros J. *Science* 367(6476):453-458 (2020)

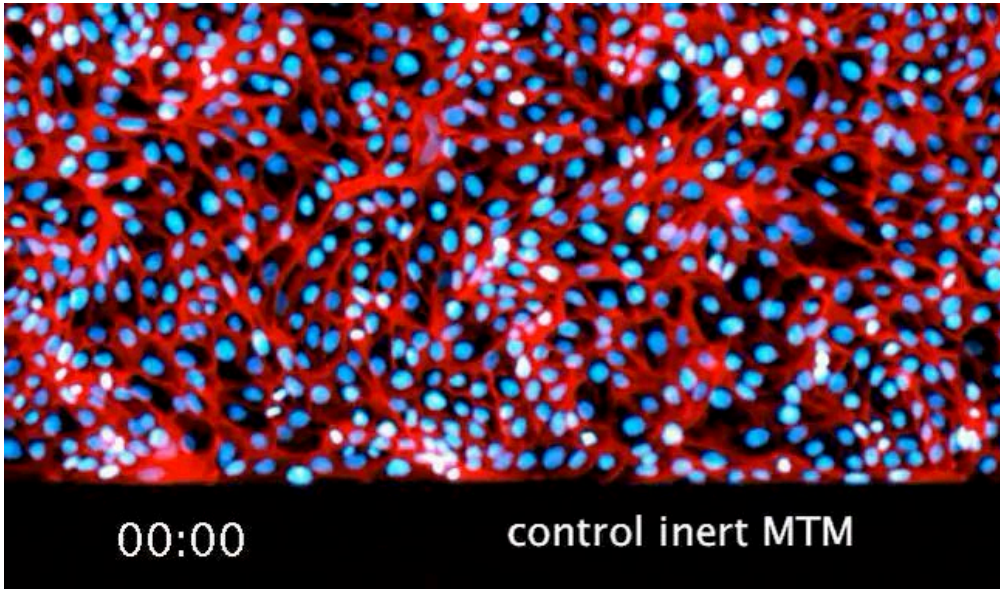
Individual and collective dynamics

- Question:

– is motility affected by high density/connectedness between cells? Seemingly not.

– can cells exhibit collective dynamics?

Hi Chi Minh city - Vietnam

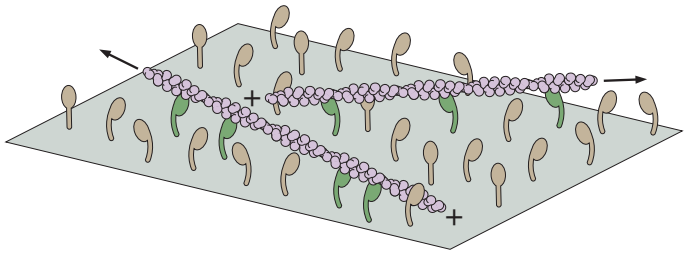


Confluent MDCK cell monolayer

D. Cohen et al *PNAS* 113: 14698–14703 (2016)
www.pnas.org/cgi/doi/10.1073/pnas.1612208113

Collective motility across scales

Microtubule filaments and Kinesin motors gliding assay: l = <1µm ; L = 100s µm



In vitro experiment

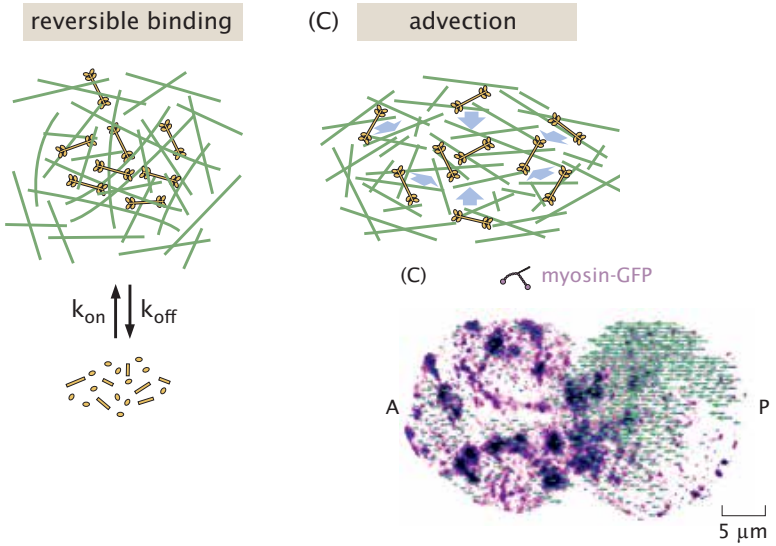
www.youtube.com/watch?v=lms2jQeHnVs

Simulation

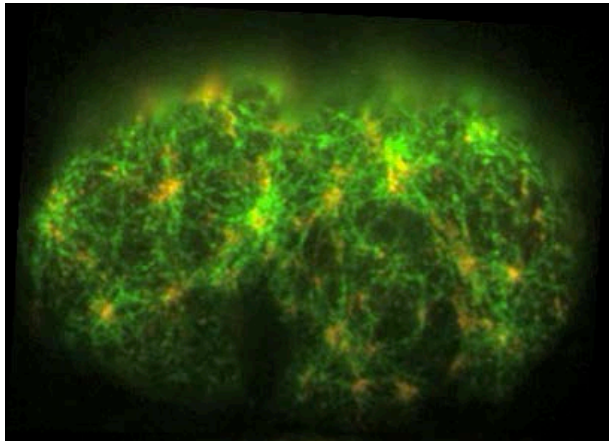
Collective motility across scales

Actin filaments and Myosin motors: l = 0.1-1µm; L = 10-100µm

Actomyosin flow

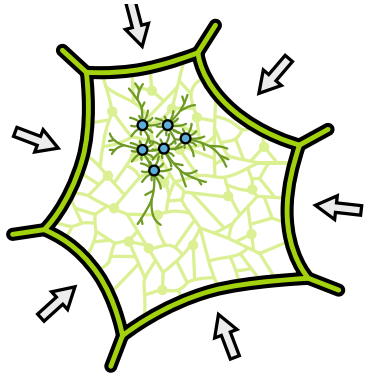


C. elegans

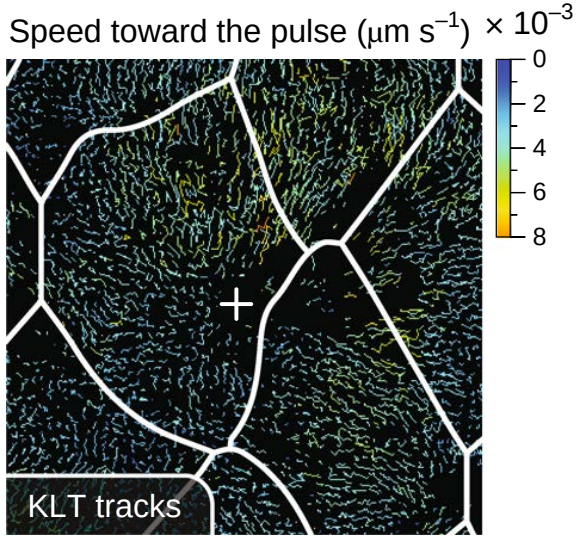


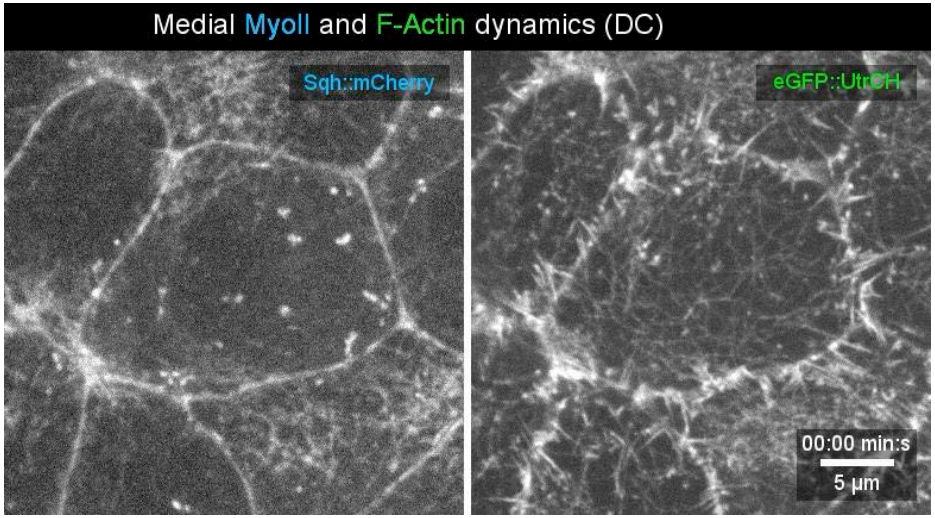
Michaux, Robin et al, E. Munro. *J Cell Biol*. 217(12):4230-4252 (2018)

M. Mayer et al and SW Grill, *Nature* 467, 617 (2010)



Drosophila

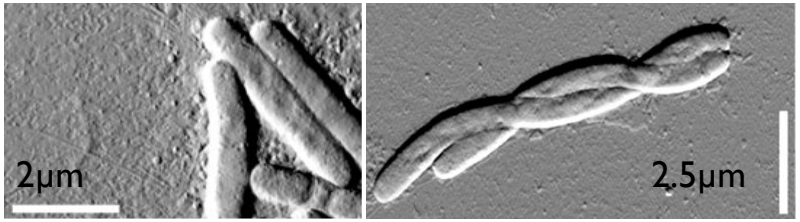




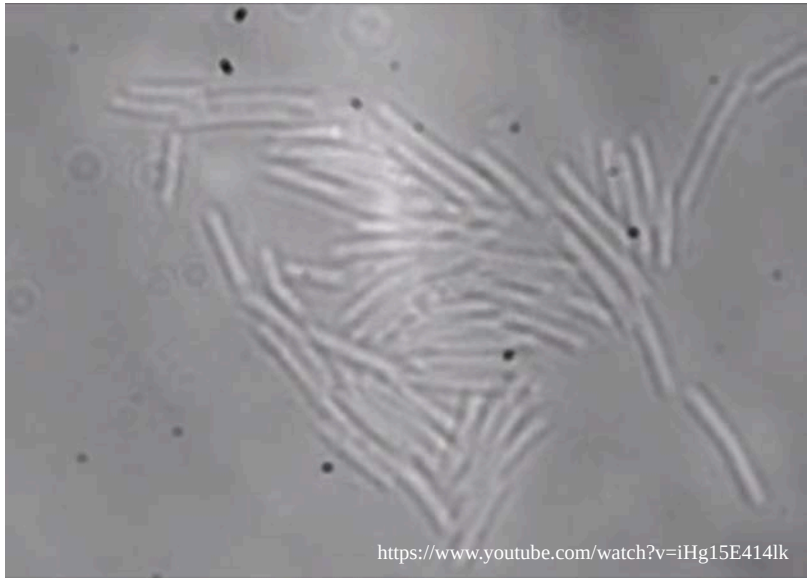
B. Dehapiot et al, and T. Lecuit. *Nat Cell Biol.* 22(7):791-802 (2020)

Collective motility across scales

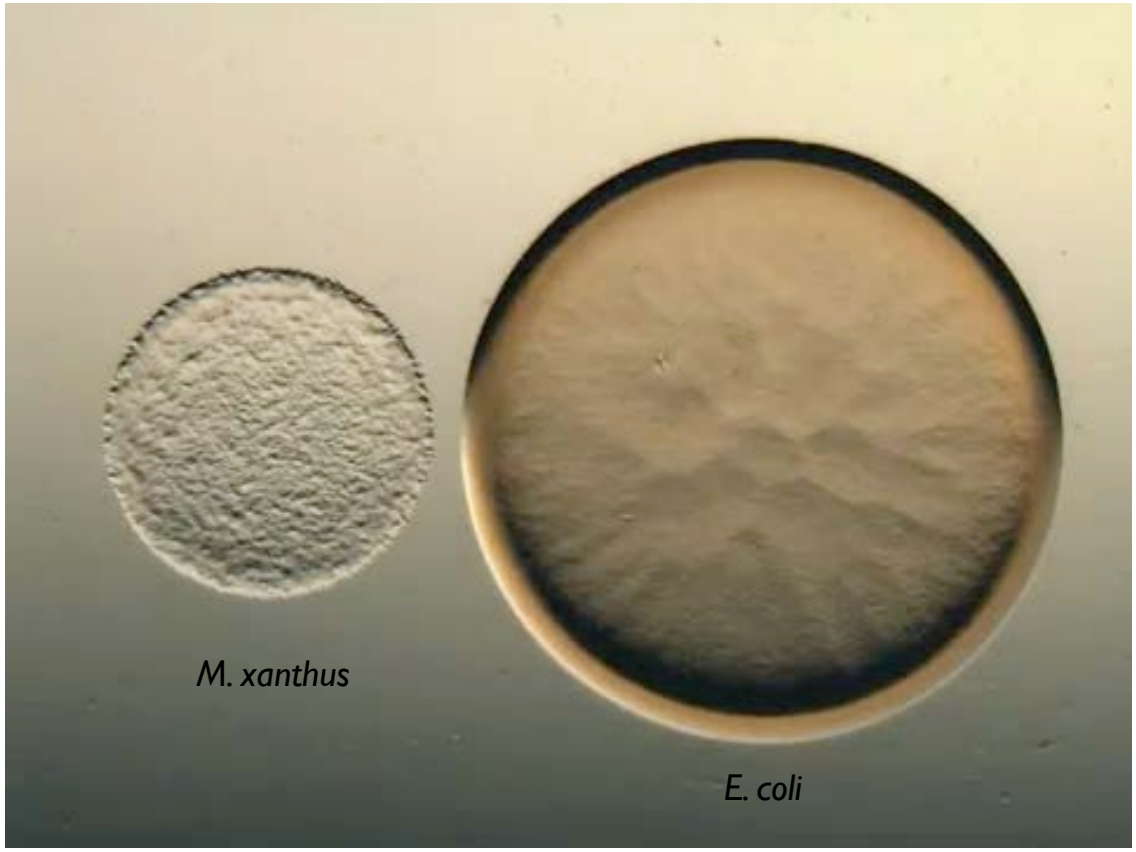
Bacteria: *Myxococcus xanthus:* L = 5µm; L ~ 1-2 cm



A. Pelling et al. PNAS, 102: 6484–6489 (2005)



speed: 2-4 µm/min

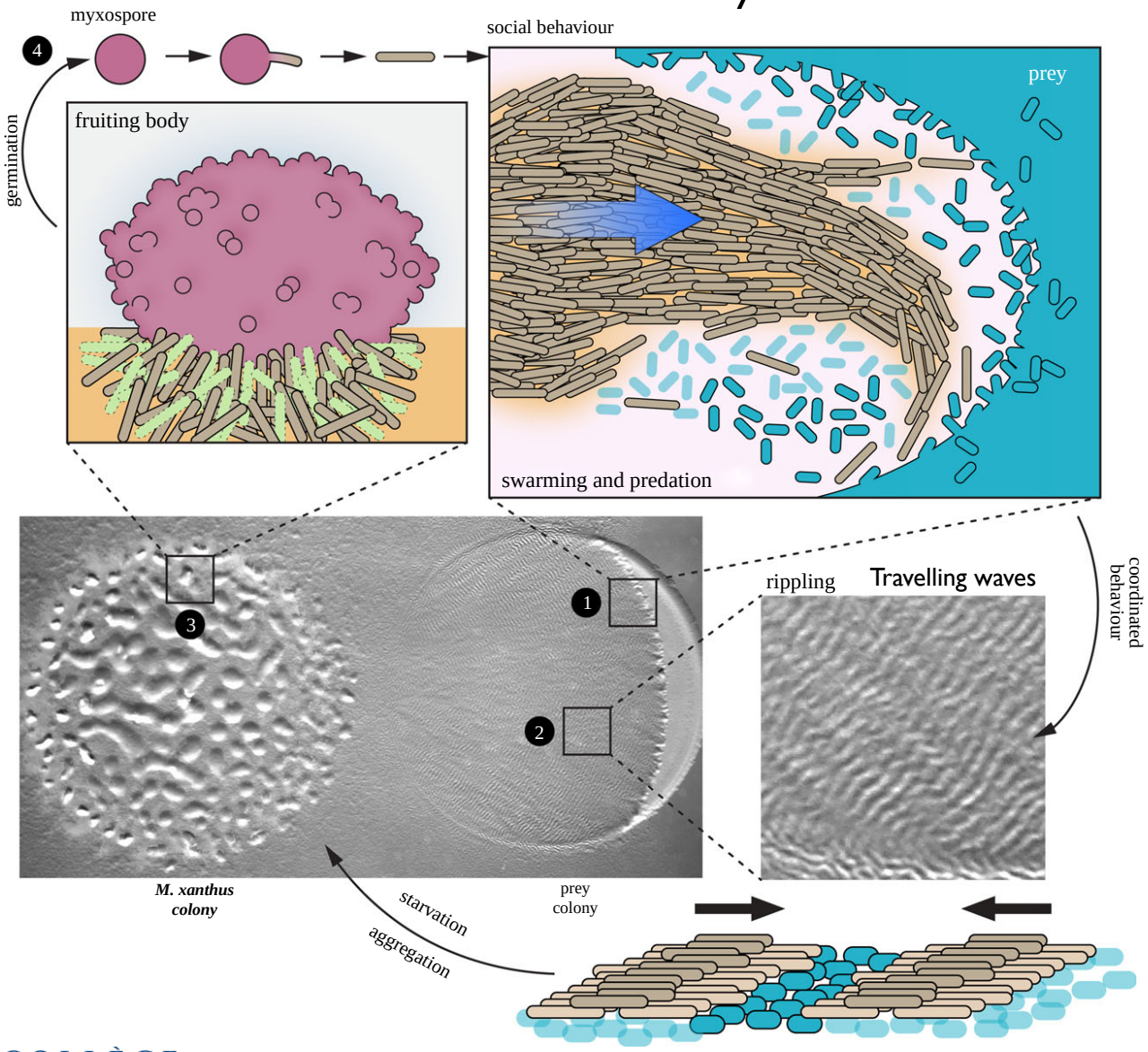


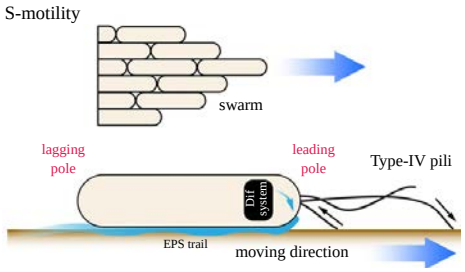
www.youtube.com/watch?v=tstc6doiNCU

Treuner-Lange, MPI Marburg

Collective motility across scales

Bacteria: *Myxococcus xanthus*: L = 5µm; L ~ 1-2 cm



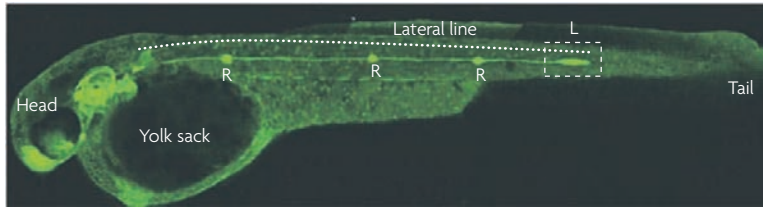


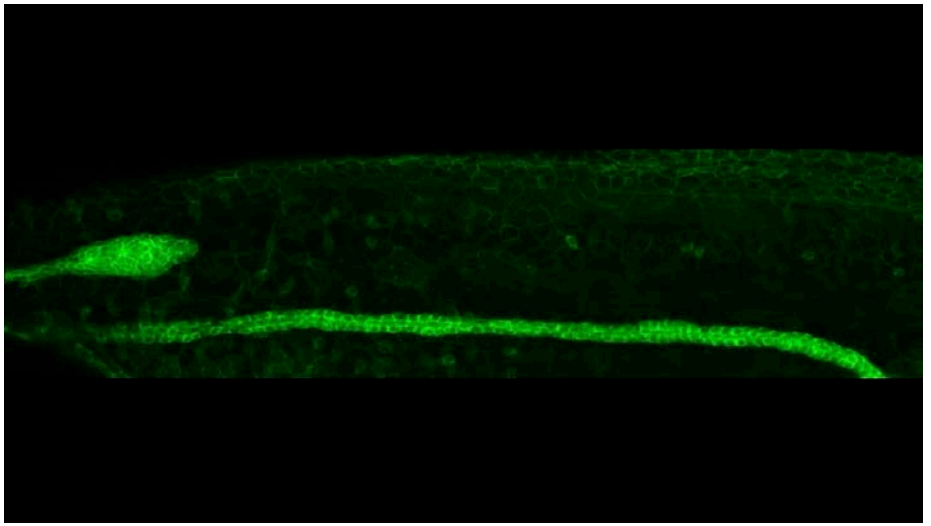
Dinet C, Michelot A, Herrou J, Mignot T. 2021 *Phil. Trans. R. Soc. B* 376: 20190755.

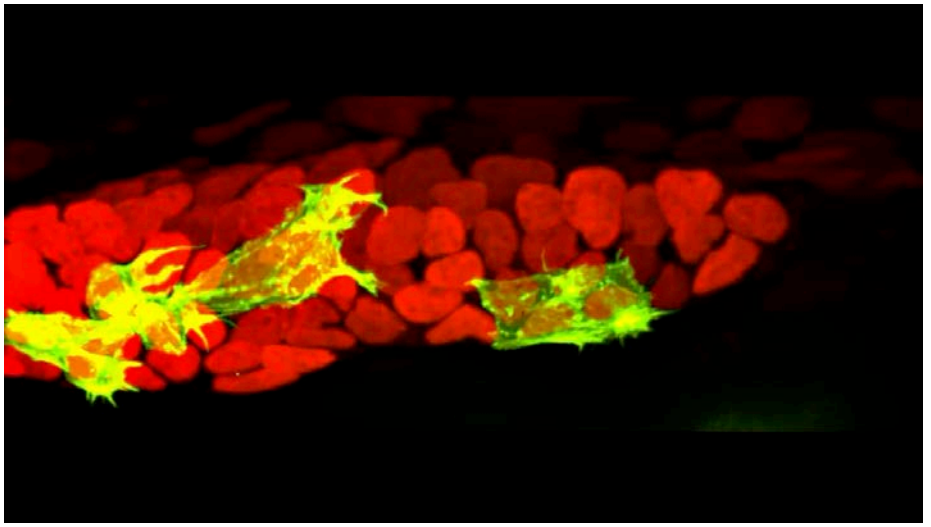
Collective motility across scales

Eukaryotic cells: Lateral line in zebrafish

l = 10-15µm; L = 1-10 mm



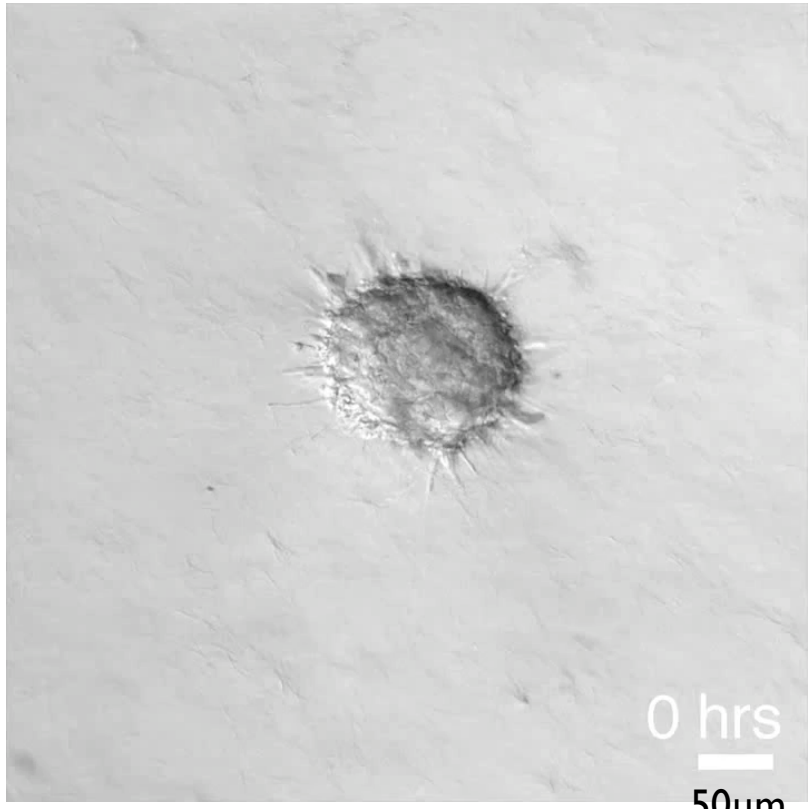




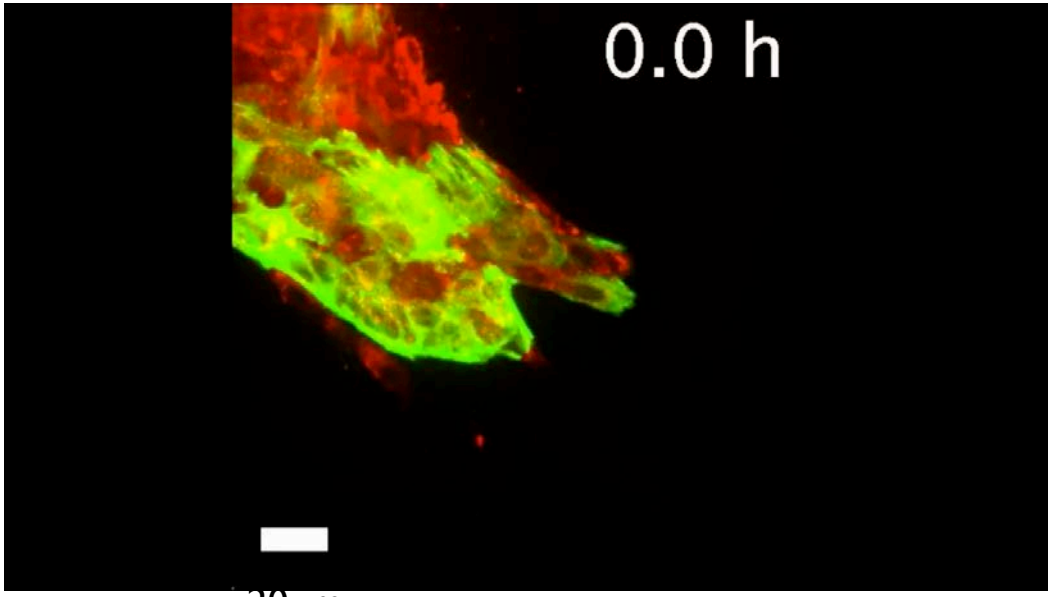
Darren Gilmour (University of Zurich)
Principles of Development
(Oxford Univ. Press). 2015
L. Wolpert, C. Tickle, A Martinez-Arias

Collective motility across scales

Invasive tumors $l \sim 20\mu m$ $L \sim$ few cm?



Invasive mammary tumor organoid



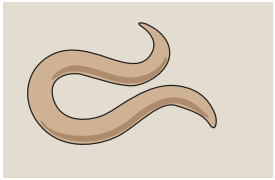
20µm
Invasive mammary tumor organoid
Keratin14+ cells in green

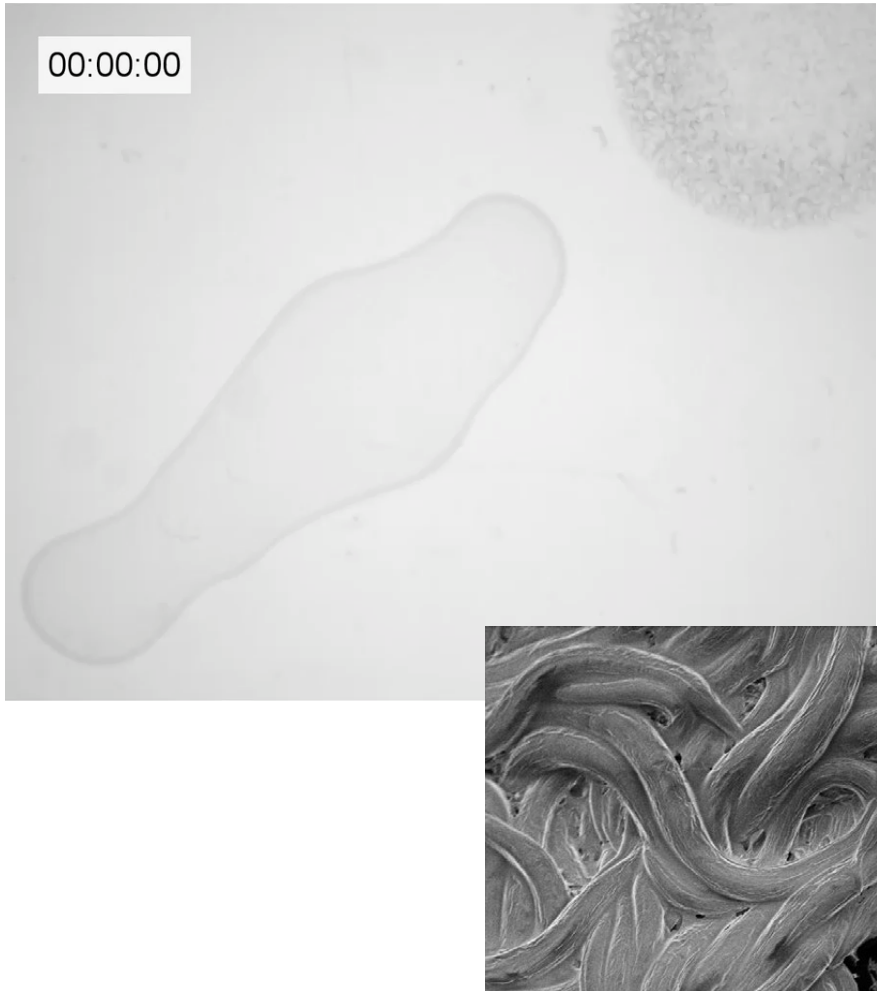
KJ. Cheung et al. And A. Ewald. *Cell* 155(7):1639-51 (2013)

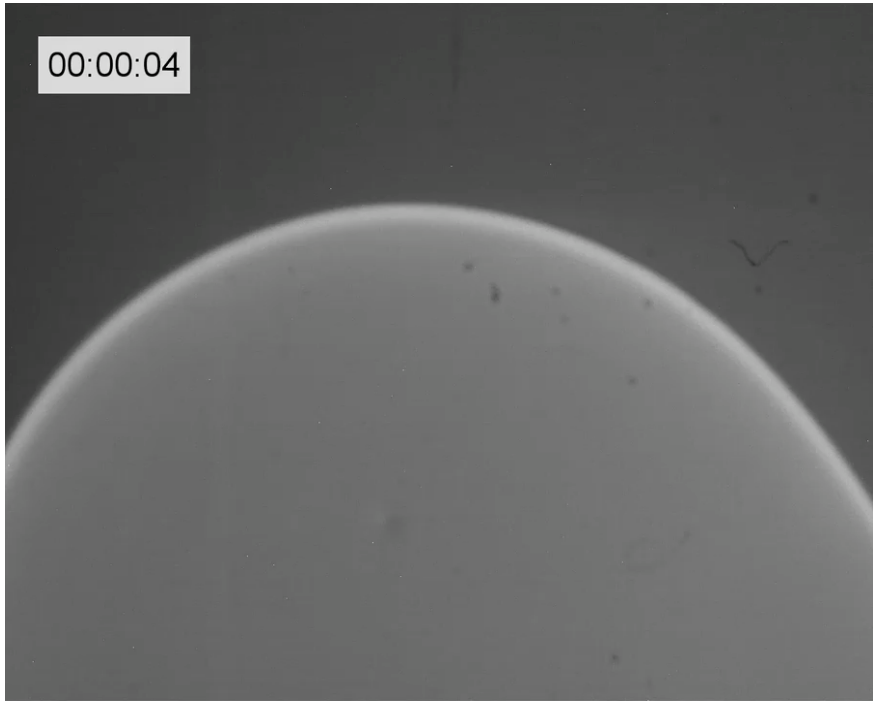
Collective motility across scales

Swarming in nematode *Caenorhabditis elegans*

l = 500µm; L = 10cm







Demir et al. A. Kocabas. *eLife* 2020;9:e52781.

Collective motility across scales

Fish schooling l = 1cm-1m; L= 10s m

www.youtube.com/watch?v=vcPlPAAsfP0

Alex Kydd

Collective motility across scales

Bird flocking (murmuration of dunlin)

$l \sim 0.2\text{m}$; $L = 100\text{s m}$

(*Calidris*)

www.youtube.com/watch?app=desktop&v=ObDlvBLPxas

Port Susan Bay, Washington

Collective motility across scales

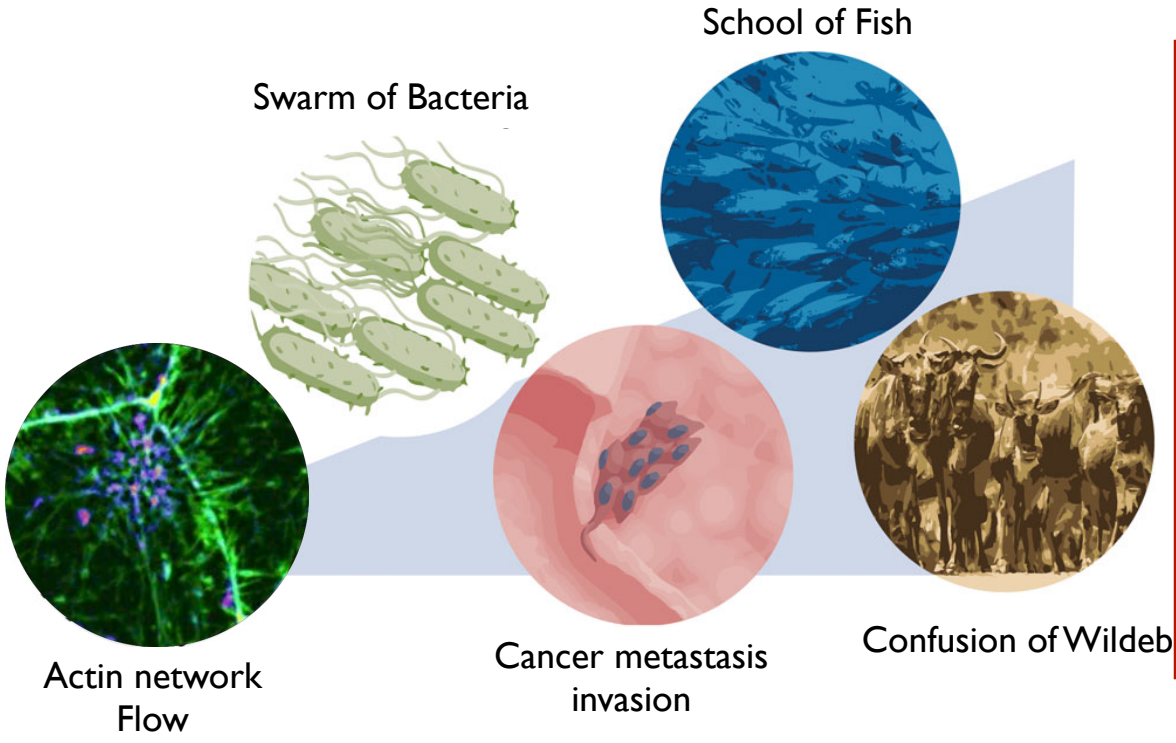
Sheep herding $l \sim 1\text{m}; L = 100\text{s m}$

Lior Patel, photographer

www.youtube.com/watch?v=cI8wV9B2VN0

Collective motion across scales

– General properties of swarming living systems

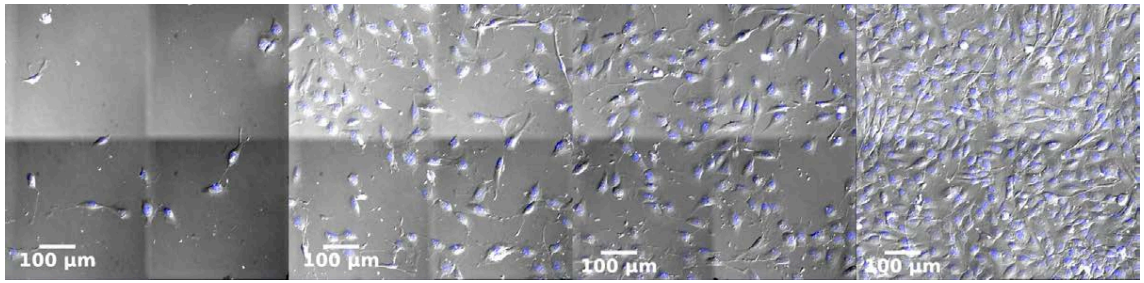


- Active motion: self-propelled
- Interactions: attraction and repulsion (density)
- Alignement (ordering)
- Boundary conditions

Adapted from Shellard A, Mayor R. *Phil.Trans. R. Soc. B* 375: 20190387. (2020)

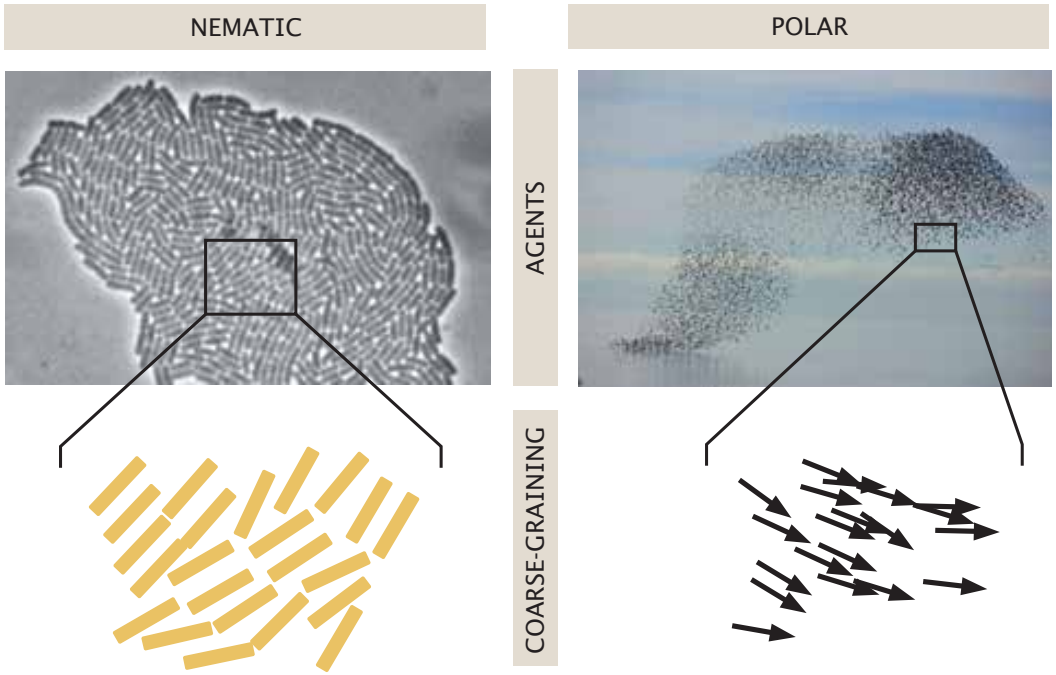
Collective motion across scales

- Active motion (energy flux): density dependent collective motion



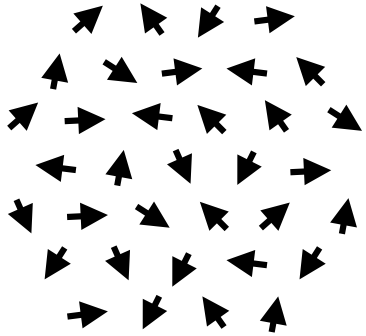
CA. Burger et al. *Front. Cell Dev. Biol.* 10:854721

- Interactions: attraction and repulsion allow density regulation
- Alignement: axis (nematic order, tensorial) or direction (polarity, vectorial)

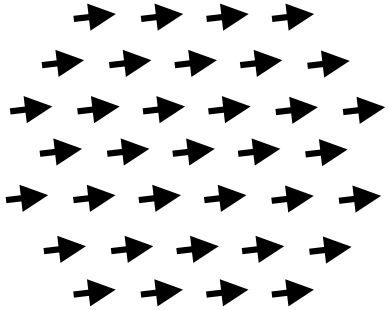


Collective motility: emergence of ordered state

- Emergence of ordered state (eg. alignement) from local interactions
- Emergence of *local* polarity
- Emergence of *global* polarity with leader: intrinsic or extrinsic

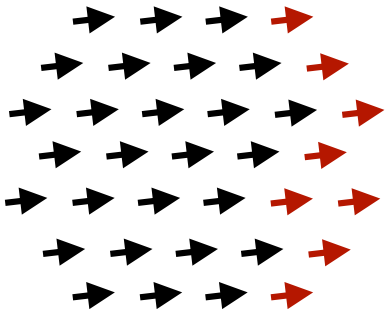


- No leader

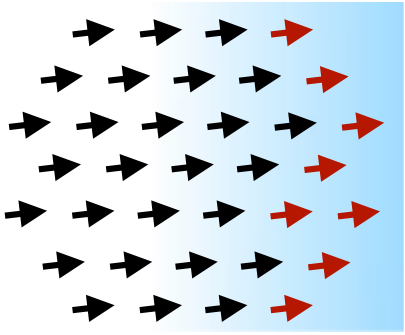


- LOCAL symmetry breaking
- Cell-cell coupling

- Leaders



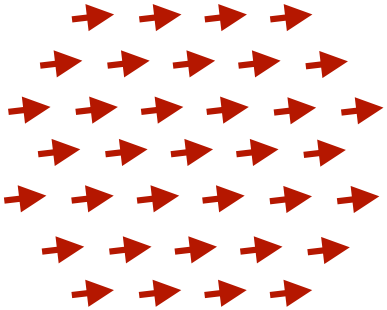
- Intrinsic, GLOBAL symmetry breaking
- Cell-cell coupling



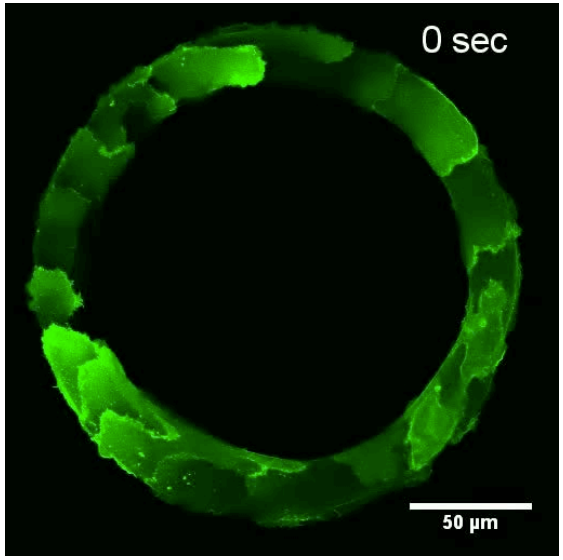
- Extrinsic GLOBAL symmetry breaking
- Cell-cell coupling

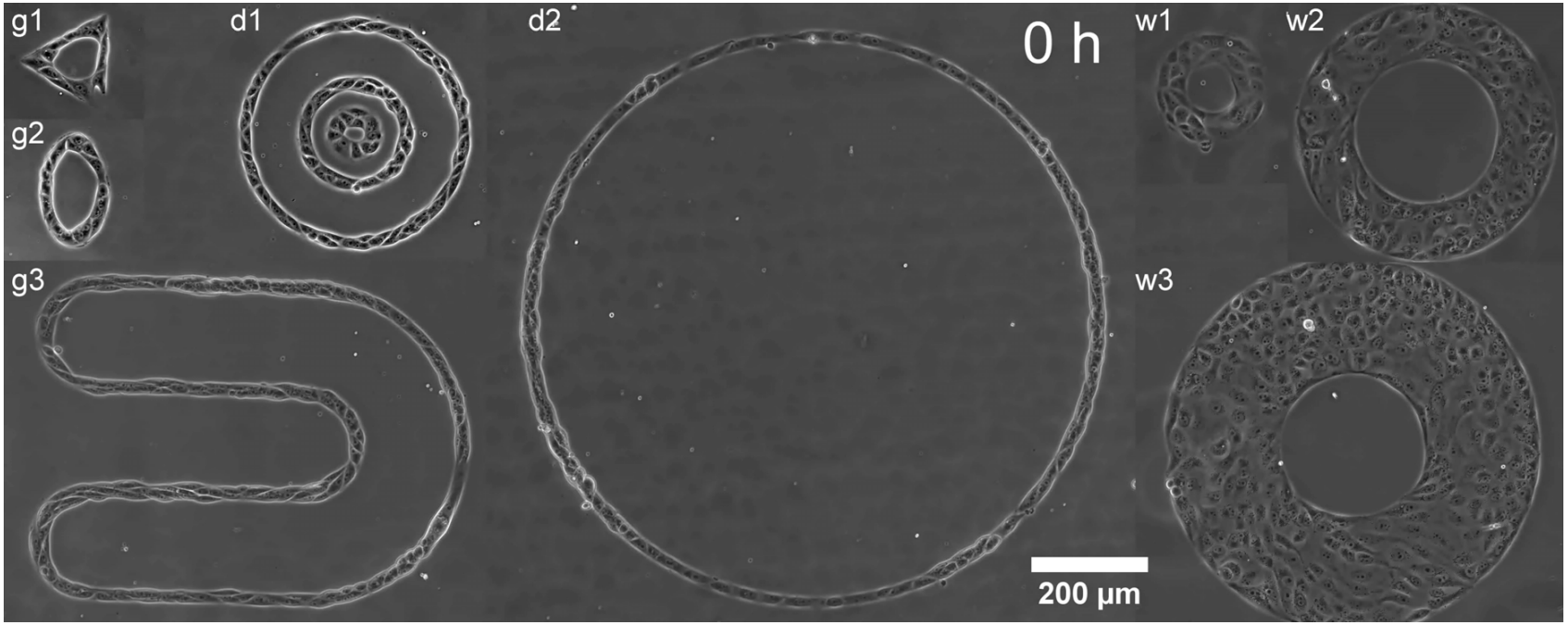
Collective motility: emergence of ordered state

- Without leader



- Local symmetry breaking
- Cell-cell coupling

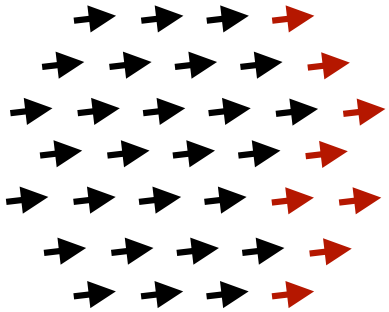




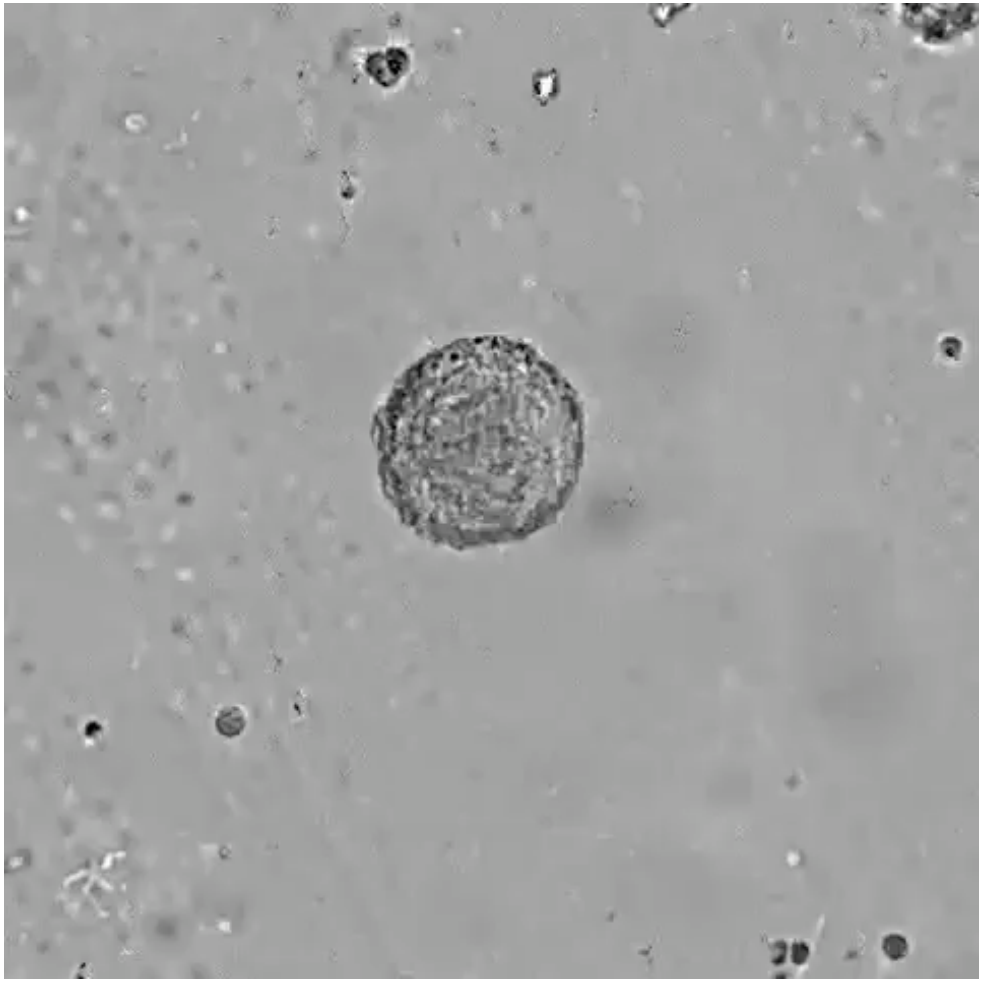
Jain, S. et al. and B. Ladoux *Nat. Phys.* 16, 802–809 (2020).

Collective motility: emergence of ordered state

- With leader cells



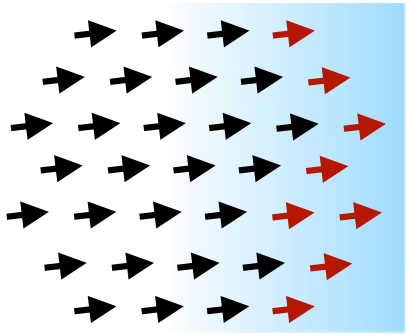
- Intrinsic, GLOBAL symmetry breaking
- Cell-cell coupling



G. Beaune et al, …, and F. Brochard-Wyart. *PNAS*, 115: 12927 -12931 (2018)

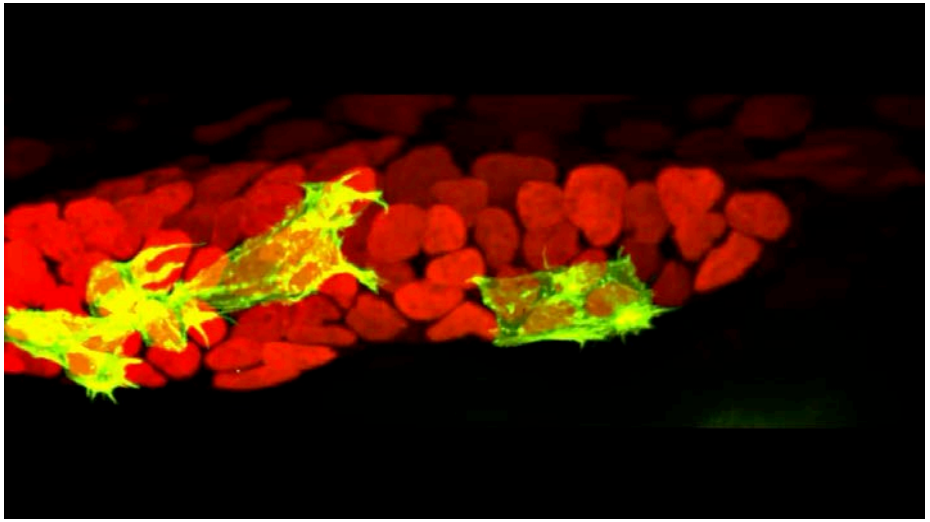
Collective motility: emergence of ordered state

- With leader cells



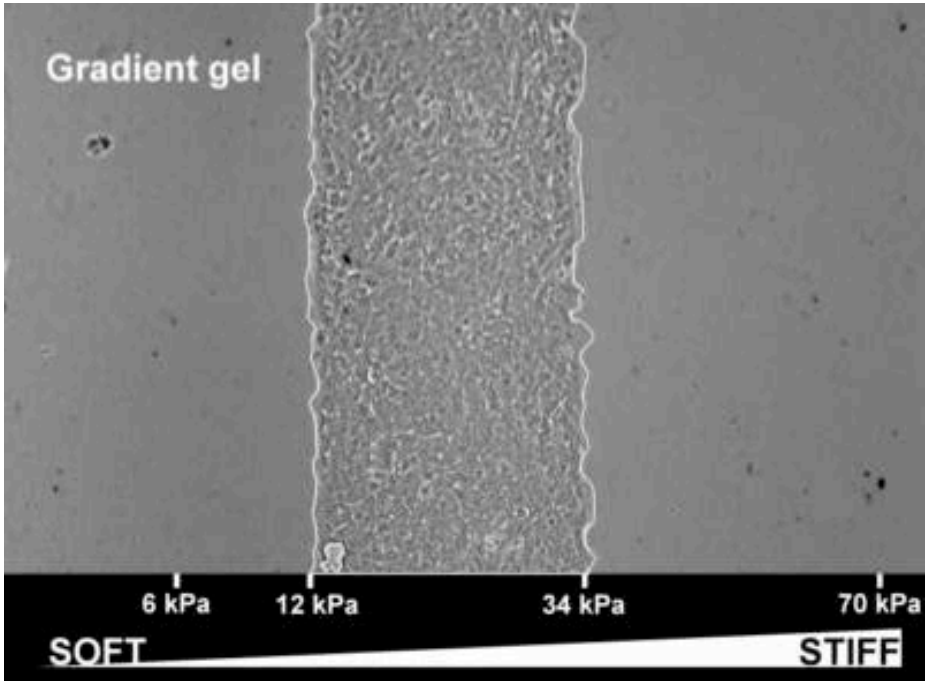
- Extrinsic GLOBAL symmetry breaking
- Cell-cell coupling

- Chemical Gradient



E. Dona et al. and D. Gilmour. *Nature*, 503(7475):285-9 (2013)

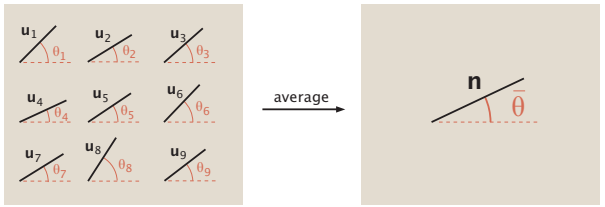
- Stiffness Gradient

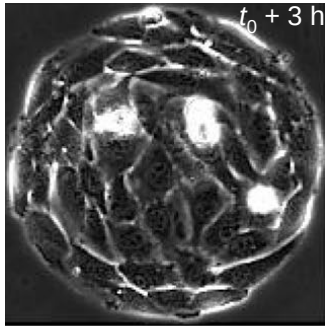


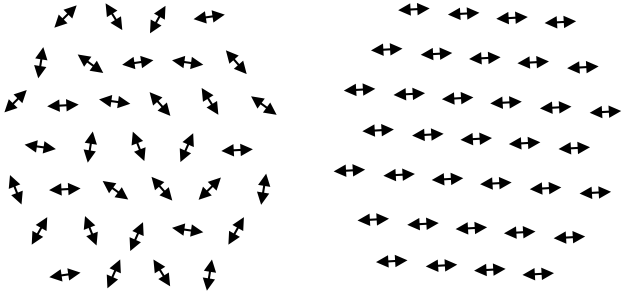
Sunyer, R., et al and Roca-Cusachs, P., and X. Trepat. *Science 353*, 1157–1161 (2016)

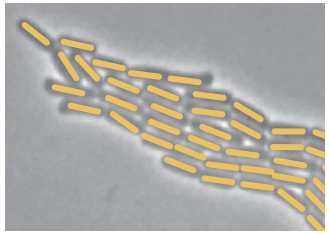
Collective motility: emergence of ordered state

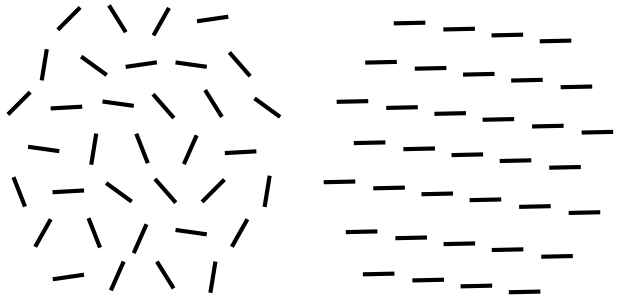
- Quantifying ordering of axis: Nematic order









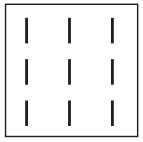
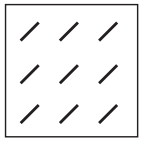
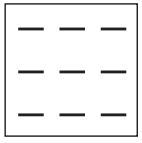


- Probability distribution $\psi(\mathbf{u})$
- Nematic tensor order (in 2D)

$$Q_{ij} = \langle u_i u_j \rangle - \frac{1}{2}\delta_{ij}$$ (=0 for disordered state)

2nd statistical moment

- Scalar order $S = 1 - \langle \delta\theta^2 \rangle$ $\theta_i = \bar{\theta} + \delta\theta_i$

coarse-graining region	director	scalar order parameter	tensor order parameter
	$\mathbf{n} = (0,1)$	$S = 1$	$\mathbf{Q} = \begin{bmatrix} -\frac{1}{2} & 0 \\ 0 & \frac{1}{2} \end{bmatrix}$
	$\mathbf{n} = \left(\frac{1}{\sqrt{2}}, \frac{1}{\sqrt{2}}\right)$	$S = 1$	$\mathbf{Q} = \begin{bmatrix} 0 & \frac{1}{2} \\ \frac{1}{2} & 0 \end{bmatrix}$
	$\mathbf{n} = (1,0)$	$S = 1$	$\mathbf{Q} = \begin{bmatrix} \frac{1}{2} & 0 \\ 0 & -\frac{1}{2} \end{bmatrix}$
	$\mathbf{n} = (0,1)$	$S = \frac{1}{3}$	$\mathbf{Q} = \begin{bmatrix} -\frac{1}{6} & 0 \\ 0 & \frac{1}{6} \end{bmatrix}$

A model of collective motility in animals

- Theory of flocking and herding (Toner-Tu)

Rate of change of velocity is a function of animal density, velocities and spatial gradients of velocities

$$\left(\frac{\partial \mathbf{v}(\mathbf{r},t)}{\partial t}\right)_{\text{total}} = \sum \mathbf{f}(\rho, v_j, \frac{\partial v_k}{\partial x_l})$$

Continuity equation: conservation of animals $\quad \frac{\partial \rho}{\partial t} + \nabla \cdot (\rho \mathbf{v}) = 0.$

Minimal version

$$\frac{\partial \mathbf{v}}{\partial t} = \left(\alpha(\rho - \rho_c) - \beta|\mathbf{v}|^2\right)\mathbf{v} - \sigma\nabla\rho + D\nabla^2\mathbf{v} - \lambda\mathbf{v}\cdot\nabla\mathbf{v}.$$

preferred speed — preferred density — neighbor coupling — advection

Homeostasis — Homogeneity

Toner, J and Tu, Y, (1998) Flocks, herds, and schools: A quantitative theory of flocking, *Phys. Rev. E* 58, 4828-4858.

Christina Hueschen and Rob Phillips. The restless cell, *in press*

A model of collective motility in animals

- Preferred velocity

$$\frac{\partial \mathbf{v}}{\partial t} = \underbrace{(\alpha\,(\rho - \rho_c) - \beta|\mathbf{v}|^2)\mathbf{v}}_{\text{preferred speed}}$$

Self-propelled entities have a preferred velocity v_0
(Active matter property)

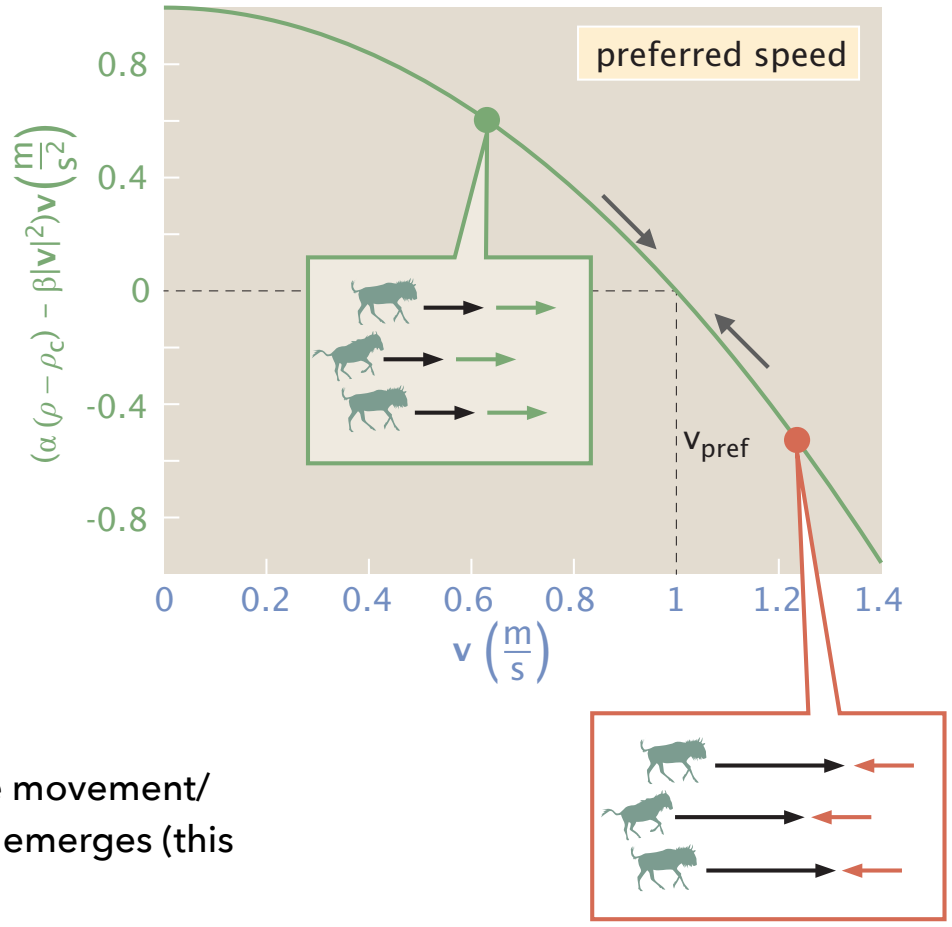
Phenomenological equation:

$$\left(\frac{\partial v_i}{\partial t}\right)_{\text{preferred speed}} = U(|\mathbf{v}|, \rho)v_i,$$

$$U(|\mathbf{v}|, \rho) = \alpha(\rho - \rho_c) - \beta(\rho)|\mathbf{v}|^2$$

Preferred velocity: $v_0 = \sqrt{\alpha(\rho - \rho_c)/\beta}$

ρ_c critical density above which collective movement/ ordered state with preferred velocity emerges (this term conveys activity in the theory)



Christina Hueschen and Rob Phillips. The restless cell, *in press*

A model of collective motility in animals

- **Preferred density:**

Animals show a preferred density ρ_0

This amounts to having an effective pressure that represses/penalizes any local increase in density

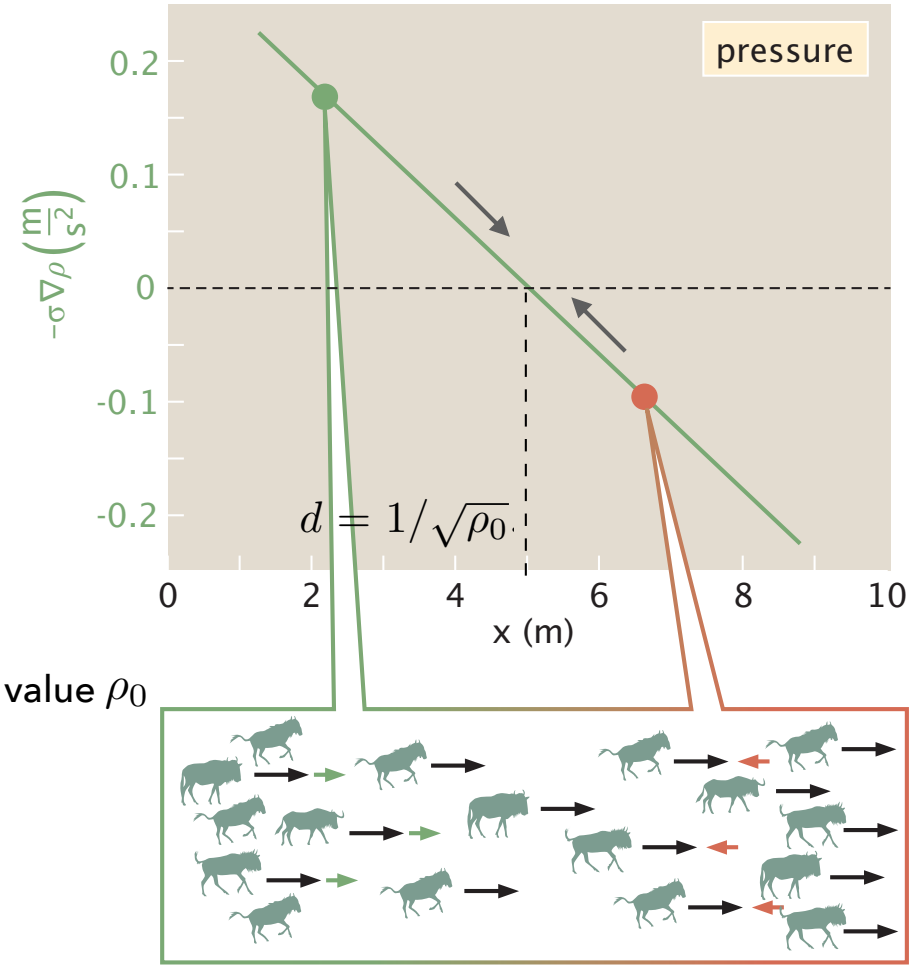
General form: $P(\rho) = \sum_n \sigma_n (\rho - \rho_0)^n$

First order term: $P(\rho) = -\sigma \nabla \rho$

(Any gradient in density is associated with a pressure)

The pressure gradient keeps the density to its preferred value ρ_0

$$\left(\frac{\partial v_i}{\partial t}\right)_{\text{preferred density}} = -\sigma \nabla \rho$$



When the density is above ρ_0 , the pressure is positive, so density decreases by increasing velocity

Christina Hueschen and Rob Phillips. The restless cell, *in press*

A model of collective motility in animals

- **Homogeneity of speed/neighbor coupling:**

Animals tend to adopt a speed that is adjusted to their neighbors, resulting in effective neighbor coupling.

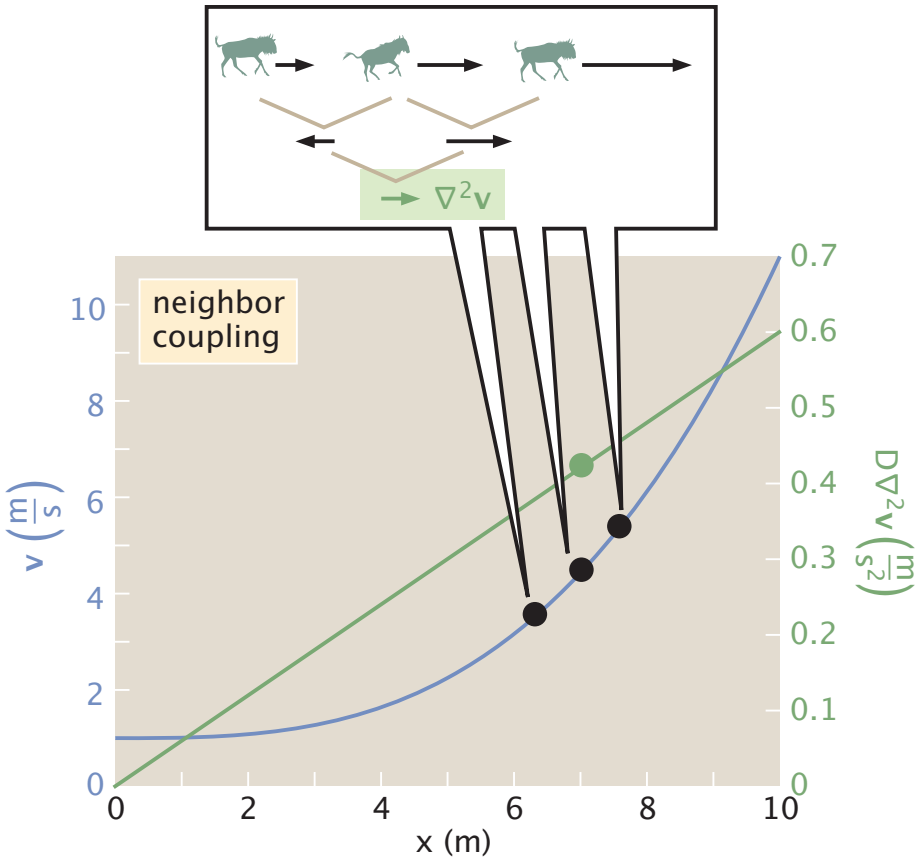
–If a neighbor goes faster, it will want to increase its speed to catch up.

–If a neighbor goes slower, it will want to slow down.

Animals sample their environment and update their velocity based on the gradient of velocity differences

This has the effect of *smoothing* any gradient in velocity and has the form of a *diffusion equation of velocity*.

$$\left(\frac{\partial v_i}{\partial t}\right)_{smoothing} = \mathrm{D}\,\nabla^2 v_i$$



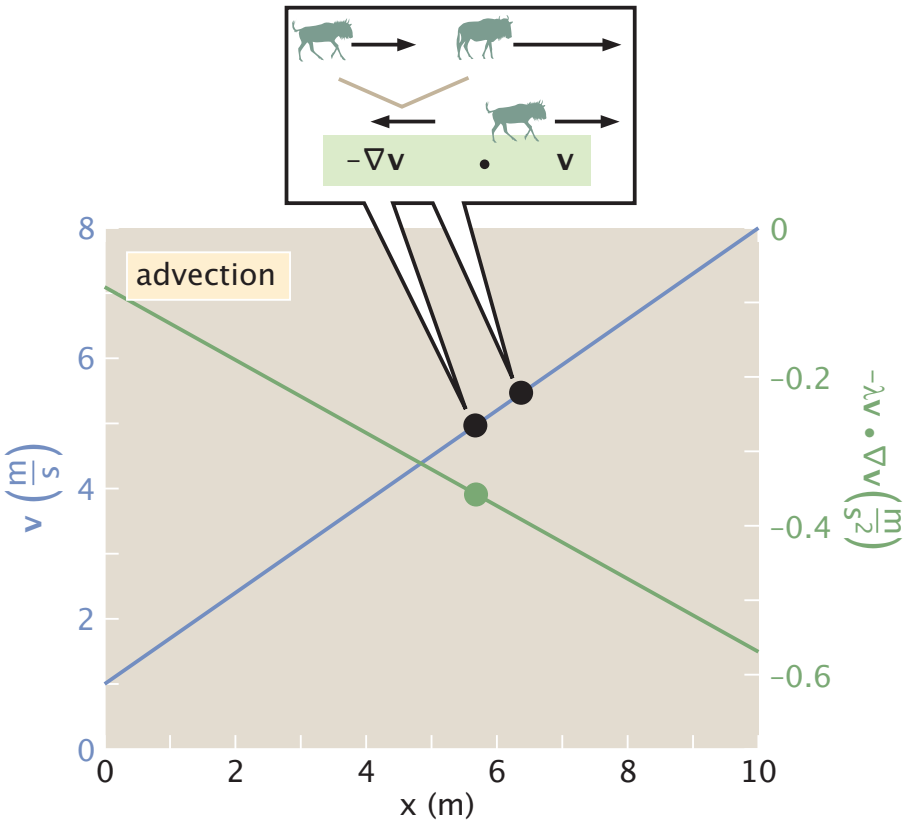
Christina Hueschen and Rob Phillips. The restless cell, *in press*

A model of collective motility in animals

- Penalizing gradient of velocity

A positive gradient of velocity (a mismatch in velocity) has the effect of reducing the velocity over time.
This is akin to an *advection of velocity*

$$\left(\frac{\partial v_i}{\partial t}\right)_{\text{gradient penalty}} = - \lambda v_j \frac{\partial v_i}{\partial x_j}$$



Christina Hueschen and Rob Phillips. The restless cell, *in press*

Collective motility: emergence of ordered state

- Prediction of velocity profiles in a 1-dimensional channel:

Governing equations: $\frac{\partial \mathbf{v}}{\partial t} = \left(\alpha(\rho - \rho_c) - \beta|\mathbf{v}|^2\right)\mathbf{v} - \sigma\nabla\rho + D\nabla^2\mathbf{v} - \lambda\mathbf{v}\cdot\nabla\mathbf{v}.$ $\qquad \frac{\partial \rho}{\partial t} + \nabla\cdot(\rho\mathbf{v}) = 0.$

The parameters can be determined from data such that velocity and density changes can be predicted in simulations

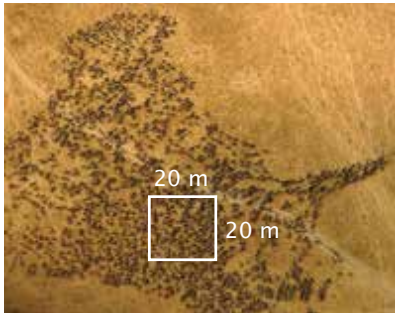
$\alpha = 0.5\ \mathrm{m}^2/\mathrm{s}$ $\qquad \rho_c \approx \frac{1}{20}\frac{1}{\mathrm{m}^2}$

$\beta = 0.1\ \mathrm{s}/\mathrm{m}^2.$

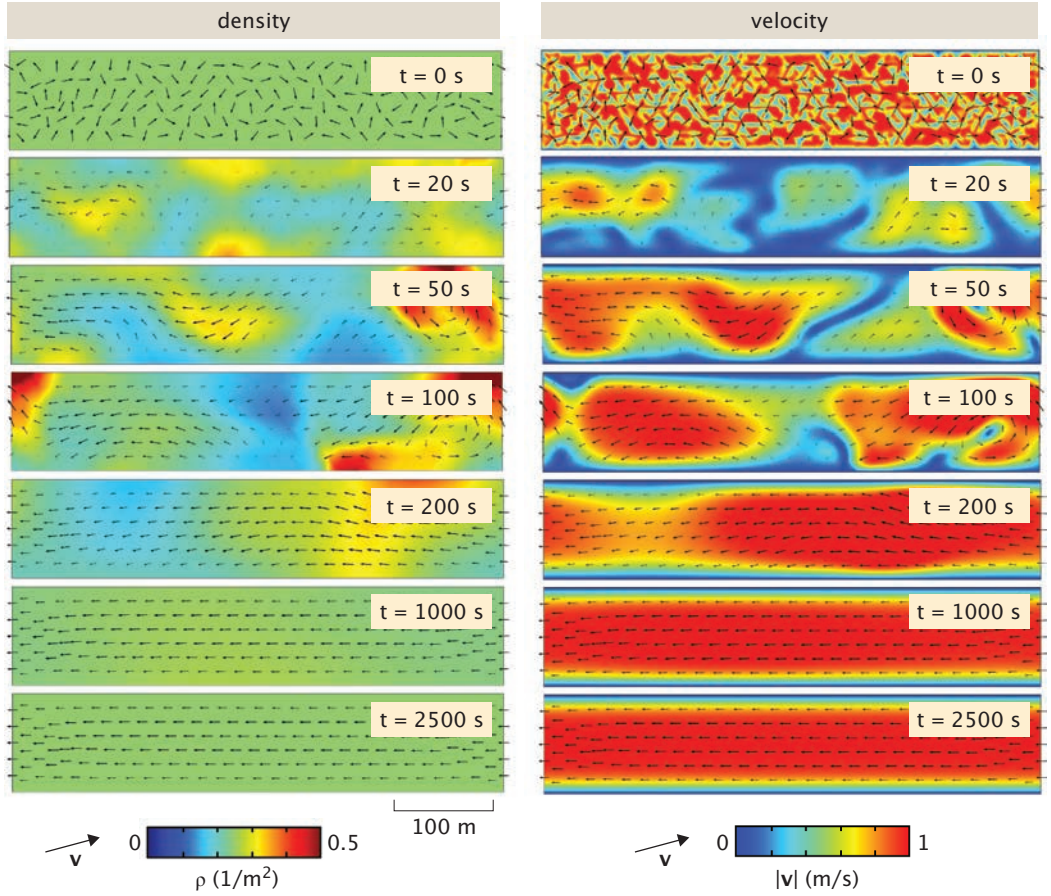
$\sigma = 10\ \mathrm{m}^4/\mathrm{s}^2.$ $\qquad \rho_0 = \frac{1}{4}\frac{1}{\mathrm{m}^2}$

$D = 10\ \mathrm{m}^2/\mathrm{s}$

$\lambda = 1$



- A random distribution of velocities among uniformly distributed animals converges towards a stationary large scale flow at uniform density



Christina Hueschen and Rob Phillips. The restless cell, *in press*

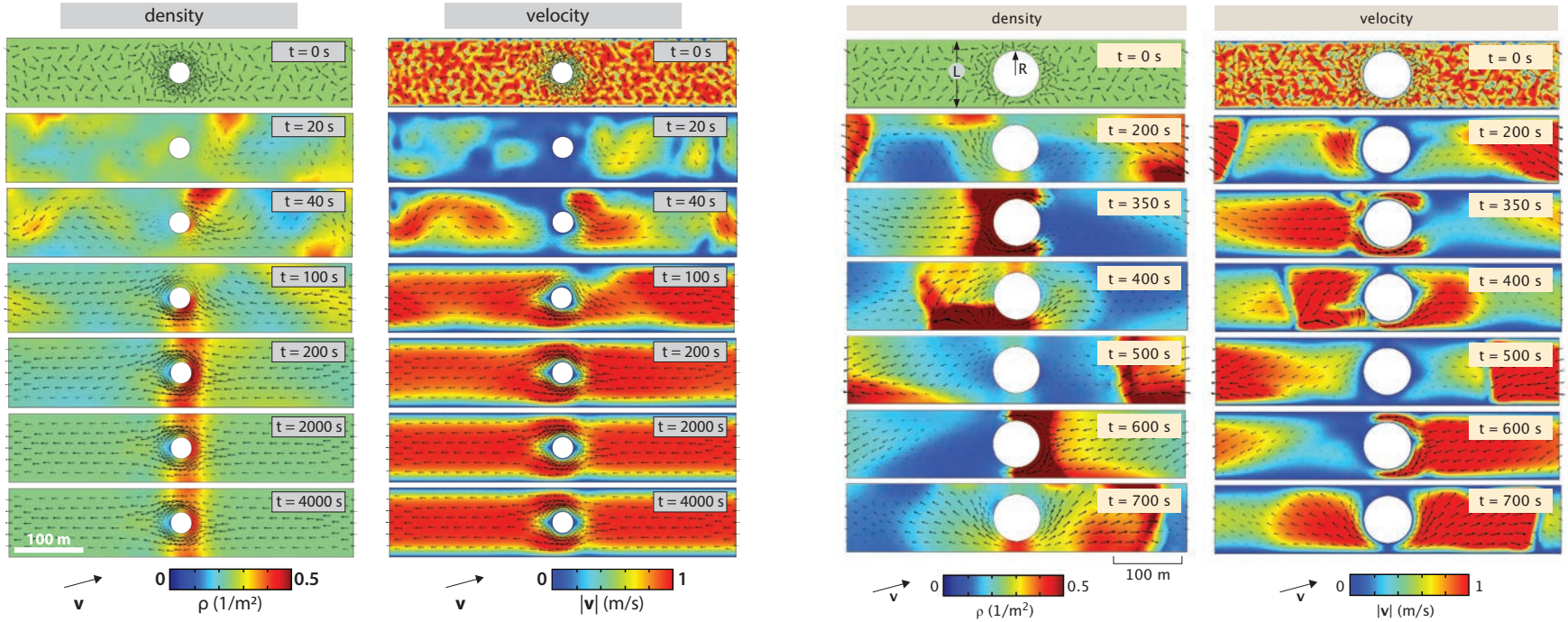
Collective motility: emergence of ordered state

- Prediction of velocity profiles in a 1-dimensional channel with an obstacle:

An obstacle causes some local increase in velocity a density.

Yet this is penalized in the Toner-Tu model by the preferred speed term (as well as preferred density term) so if the obstacle becomes too large, the flow will not persist at the edge of the obstacle and the herd will reflect on the obstacle

$$\frac{\partial \mathbf{v}}{\partial t} = \left(\alpha(\rho - \rho_c) - \beta|\mathbf{v}|^2\right)\mathbf{v} - \sigma\nabla\rho + D\nabla^2\mathbf{v} - \lambda\mathbf{v}\cdot\nabla\mathbf{v}.$$



Christina Hueschen and Rob Phillips. The restless cell, *in press*

Collective motility: emergence of ordered state

Flocking across scales: Actin filaments, birds and wildebeest

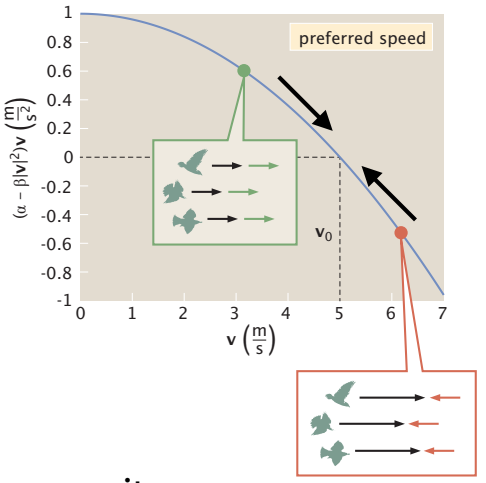
Sandpipers (*Calidris)*

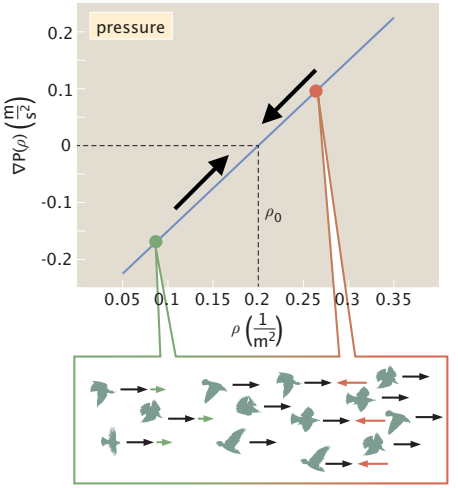
ebird.org

Ocean Shores, Washington by Peggy Dolane.

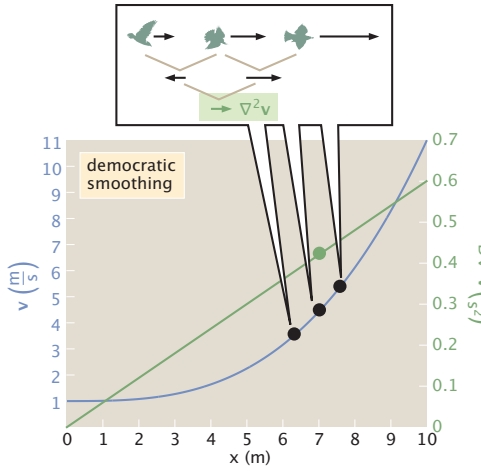
$$\frac{\partial \mathbf{v}}{\partial t} = \underbrace{(\alpha - \beta|\mathbf{v}|^2)\mathbf{v}}_{\text{preferred speed}} - \underbrace{\nabla P}_{\text{pressure}} + \underbrace{D\nabla^2\mathbf{v}}_{\text{smoothing}} - \underbrace{\lambda \mathbf{v}\cdot\nabla\mathbf{v}}_{\text{advection}}$$

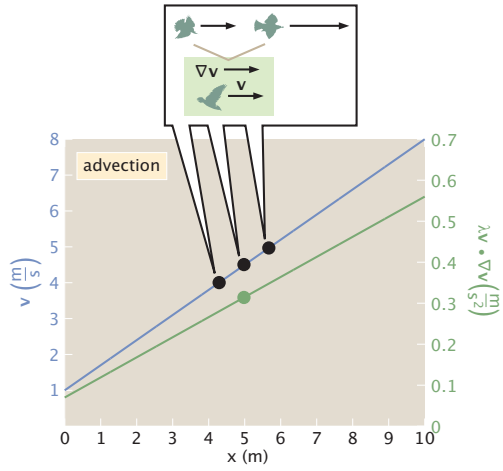
Homeostasis





Homogeneity

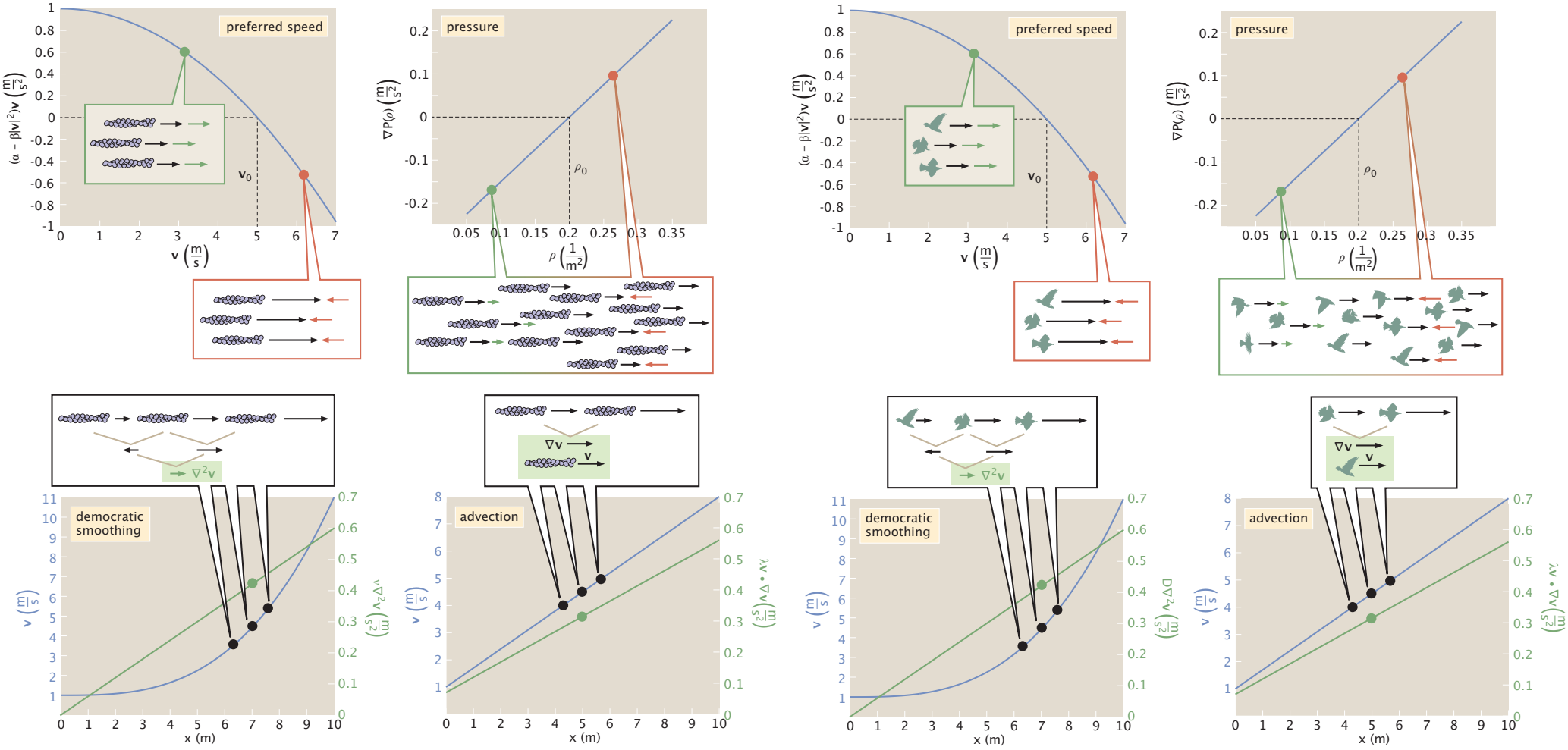




Christina Hueschen and Rob Phillips. The restless cell, *in press*

Collective motility: emergence of ordered state

Flocking across scales: Actin filaments, birds and wildebeest



Christina Hueschen and Rob Phillips. The restless cell, *in press*

Summary: Self-organisation of collective motility

- Active motion (self-propelled dynamics)
- Density dependent collective motion: collective motion requires critical density

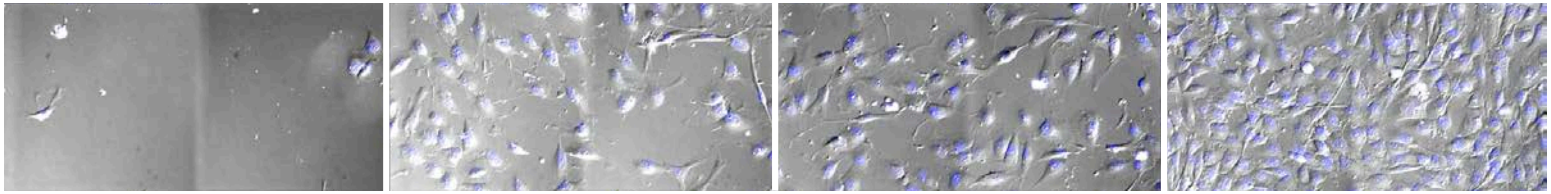
- Interactions: attraction and repulsion allow density regulation and preferred speed.

- Alignement: axis (nematic order, tensorial) or direction (polarity, vectorial) emerges with preferred speed orientation

- Boundary conditions (eg. chemical, mechanical or geometric cue): influence self-organized dynamics

- General phenomenological model
- Different underlying physics: high vs low Reynolds number, contact vs no contact between agents

What about **collective cell motility?**



- Increased cell density is associated with cell contacts and cell adhesion
- Antagonism between cell motility and cell-cell adhesion
  (Johannes Holtfreter and Paul Weiss)
- Collective motility yet exists: new collective phenomenon whereby cells maintain collective integrity (direct contacts) while being motile.

Cell motility versus cell-cell adhesion: cell sorting

- « selective adhesiveness » controls cellular migration

SIGNIFICANCE OF THE CELL MEMBRANE IN EMBRYONIC PROCESSES

By JOHANNES HOLTFRETER*
Biology Department, University of Rochester, Rochester, N. Y.

Annals: New York Academy of Sciences, 709-760. 1948

Based mainly upon observations on amphibian material, the attempt has been made to show that many embryological phenomena may be better understood if we take into consideration the properties and functions of the interfacial membranes which separate the cells from each other and from the external medium. While all cells are furnished with a liv-

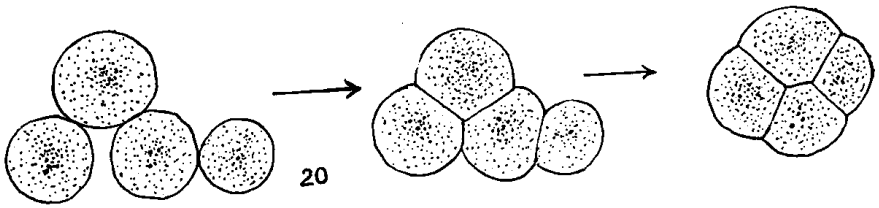


FIGURE 20. Aggregation of myelin vesicles in a 10^{-2}M solution of $CaCl_2$.

The direction of cellular migration, and the histotypical groupings and regroupings exhibited by the various types of cells in a developing organism, appear to be controlled by a selective adhesiveness of the cell membrane, which varies with the developmental stage and with the kind of cells involved. Cellular adhesiveness depends both on the chemical constitution of the contacting cell surfaces and on the composition of the immersion fluid. From the observed antagonistic effects of hydrating

Johannes Holtfreter , *circa* 1950
1901- 1992

Cell motility versus cell-cell adhesion: cell sorting

- « selective adhesiveness » controls cellular migration and cell sorting.

DIRECTED MOVEMENTS AND SELECTIVE ADHESION OF EMBRYONIC AMPHIBIAN CELLS [1]

PHILIP L. TOWNES [2] AND JOHANNES HOLTFRETER
Departments of Biology and Anatomy, The University of Rochester, Rochester, New York

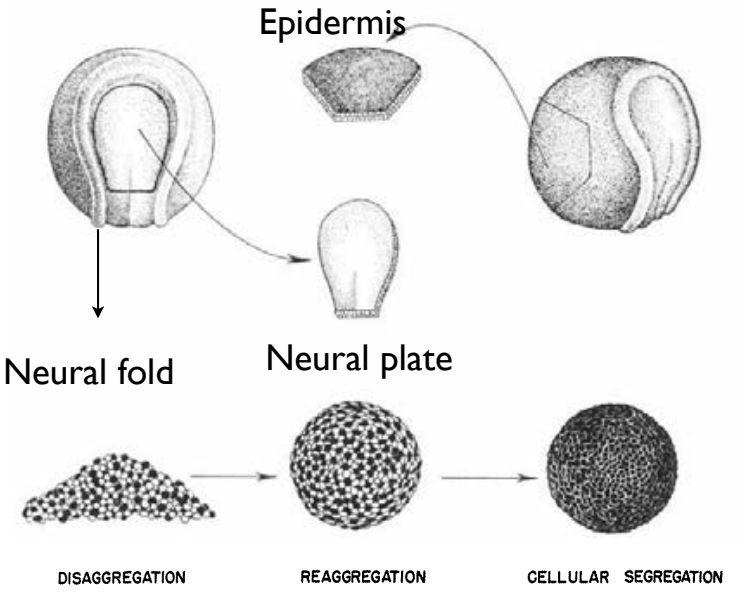


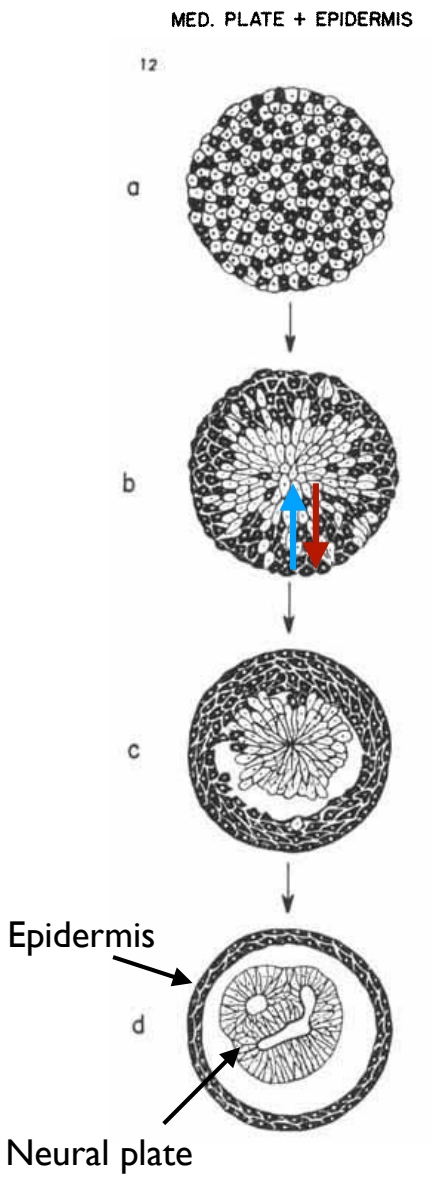
Fig. 10 A piece of the medullary plate and a piece of prospective epidermis are excised and disaggregated by means of alkali. The free cells are intermingled (epidermal cells indicated in black). Under re-adjusted conditions the cells re-aggregate and subsequently segregate so that the surface of the explant becomes entirely epidermal.

Ambystoma punctatum

Rana pipiens

Townes P. and Holtfreter J. *J. Exp. Zool.* 53-120. 1955

Cell motility versus cell-cell adhesion: cell sorting



Observations:

- Rapid cell aggregation
- Elongation of prospective neural cells
- Centripetal migration of prospective neural cells
- Centrifugal migration of epidermal cells
- Sorting of distinct cell populations according to identity
- Clear separation between two tissues

- 2 forms of cell adhesion:
  - not specific at early stages (does not affect motility)
  - specific at late stages, following prolonged contact

- Sorting relies on sequential contributions of 1) Migration and 2) Specific adhesion
- It is implied that selective adhesion adhesion blocks migration and thereby stabilises cell configuration

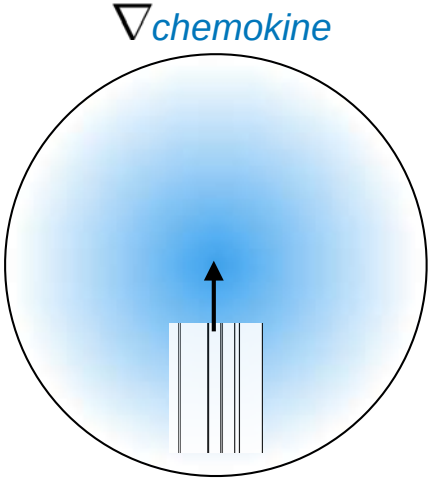
Townes P. and Holtfreter J. *J. Exp. Zool.* 53-120. 1955

Cell motility versus cell-cell adhesion: cell sorting

- Central role of cell-specific directed motility
- Gradient of chemokine
- Suggests possible role of surface tension at cellular level
- Cell-specific adhesion stabilises final configuration

- Uncouples motility and adhesion

course#2–31 Oct 2017



5. In consequence of directed movements, the different cell types in a composite aggregate are sorted out into distinct homogeneous layers, the stratification of which corresponds to the normal germ layer arrangement. The tissue segregation becomes complete because of the emergence of a selectivity of cell adhesion

Townes P. and Holtfreter J. *J. Exp. Zool.* 53-120. 1955

Antagonism between motility and adhesion

Paul A Weiss
1898-1989

PERSPECTIVES IN THE FIELD OF MORPHOGENESIS

By PAUL WEISS
Department of Zoology, University of Chicago

Cell coaptation (« affinities relations ») is based on surface recognition by molecules

Cell acquire stable configuration or are motile depending on coaptation configuration

In order to become settled, a cell must be fully adjusted and equilibrated with the conditions prevailing along its various surfaces, including its neighbors of the same kind, adjacent cell layers, the intercellular matrix, and the liquid media bathing its free portion. This relation may be designated as "coaptation."

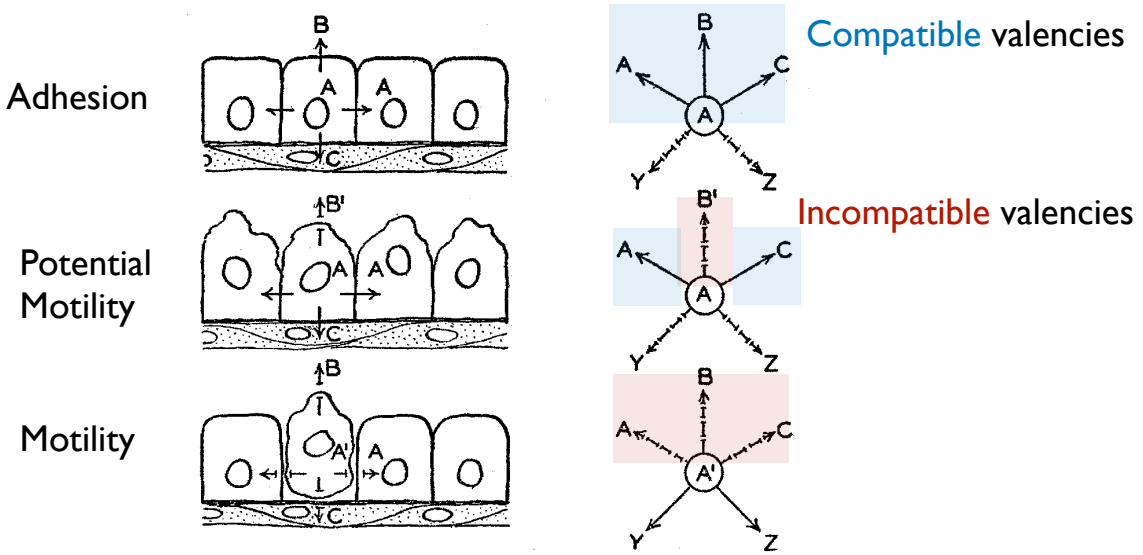


FIG. 2. AFFINITY RELATIONS IN EPITHELIUM (LEFT) WITH RELATION DIAGRAMS (RIGHT)

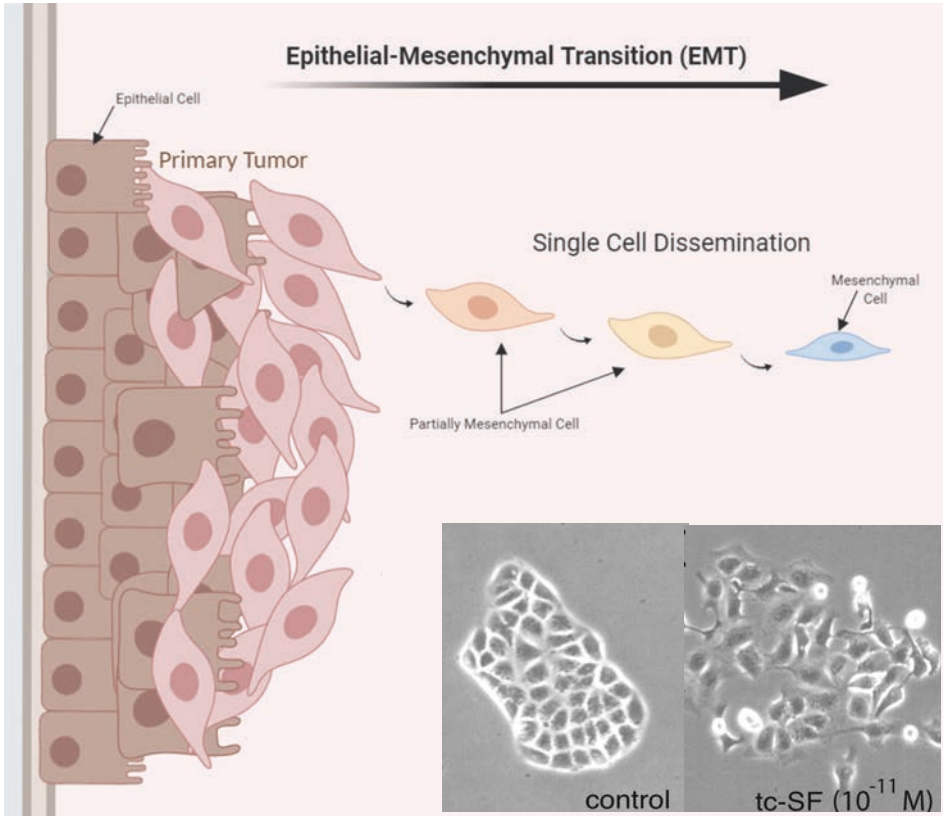
Full arrows indicate conformance of conditions (active "bonds"); interrupted arrows, non-conformance.

Paul Weiss. *The Quarterly Review of Biology*, 25, 2. (1950) https://doi.org/10.1086/397540

Antagonism between motility and adhesion

Epithelial to mesenchymal transition (EMT)

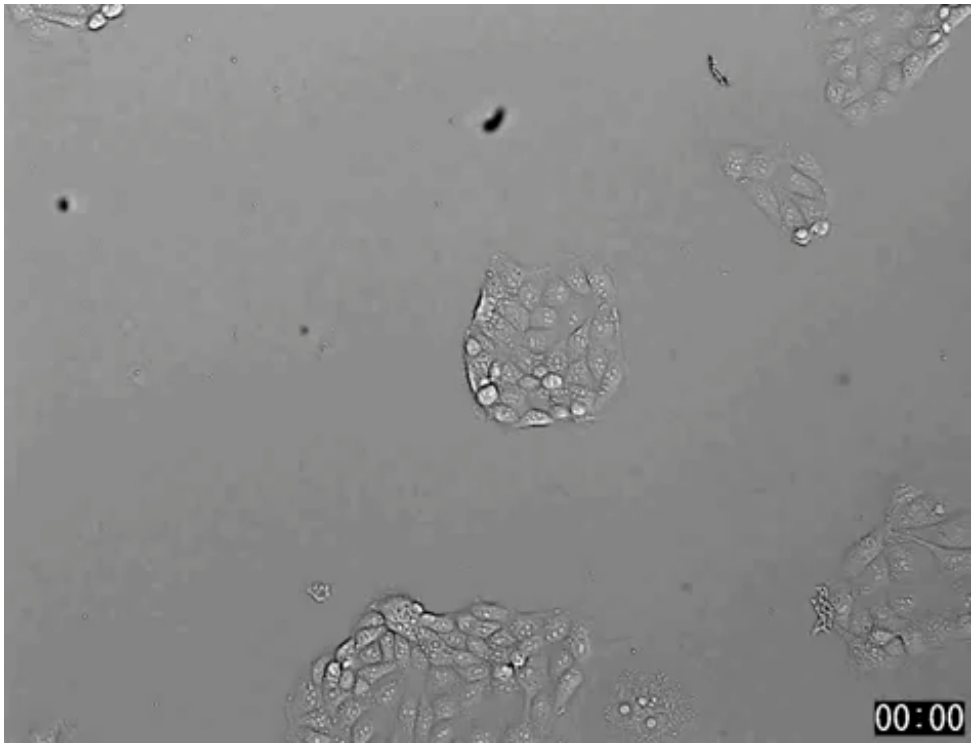
Fluid state of motile cells due to loss of adhesion and reduced cell density



J. Fares et al. *Signal Transduction and Targeted Therapy* (2020)5:28
Molecular principles of metastasis: a hallmark of cancer revisited

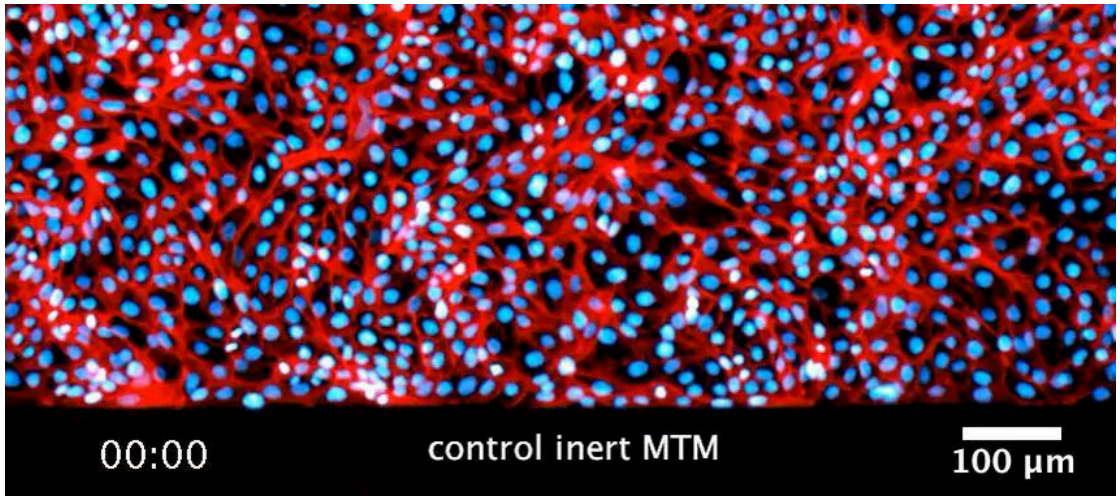
E. Gherardi et al. (2005) www.pnas.org/cgi/doi/10.1073/pnas.0509040103

HGF induced cell scattering
Loss of adhesion leads to motility (MDCK cells)



S. Nakahara et al *J Cell Sci* 128 (11): 2047–2056 ((2015).

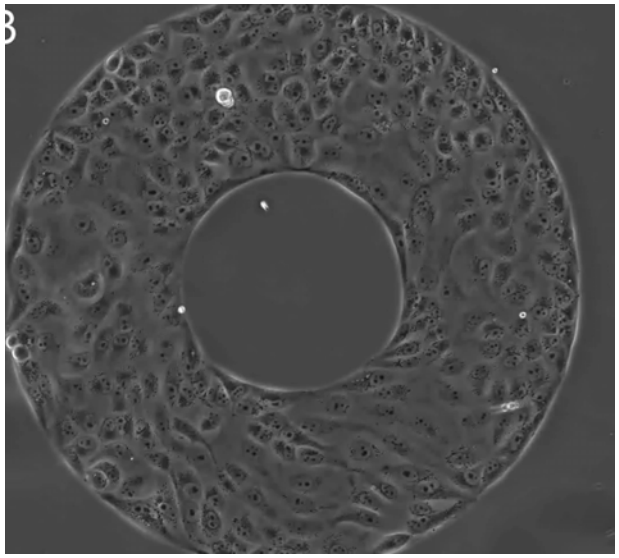
Yet, Emergence of tissue fluidity at high density among adhesive cells



MDCK cell monolayer

D. Cohen et al *PNAS* 113: 14698–14703 (2016)
www.pnas.org/cgi/doi/10.1073/pnas.1612208113

But not all epithelial layers are dynamic!

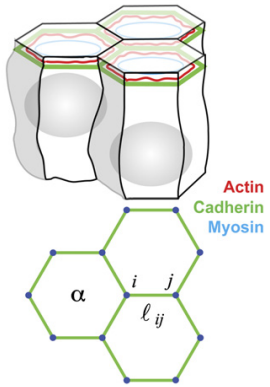


Jain, S. et al. and B. Ladoux *Nat. Phys.* 16, 802–809 (2020).

Adhesive cells can exhibit fluidity

Density-*independent* rigidity transition

- 2D Vertex model of epithelial tissue:



Force balance at vertices, and all forces derive from energy functional:

Energie function: $\boldsymbol{F_i} = -\dfrac{\partial \boldsymbol{E}}{\partial \boldsymbol{R_i}}$.

$$\boldsymbol{E}(\boldsymbol{R_i}) = \sum_\alpha \frac{\boldsymbol{K}_\alpha}{2}\left(\boldsymbol{A}_\alpha - \boldsymbol{A}_\alpha^{(0)}\right)^2 + \sum_{<i,j>} \Lambda_{ij}\ell_{ij} + \sum_\alpha \frac{\Gamma_\alpha}{2}\boldsymbol{L}_\alpha^2$$

Area elasticity | Line tension | Contractility

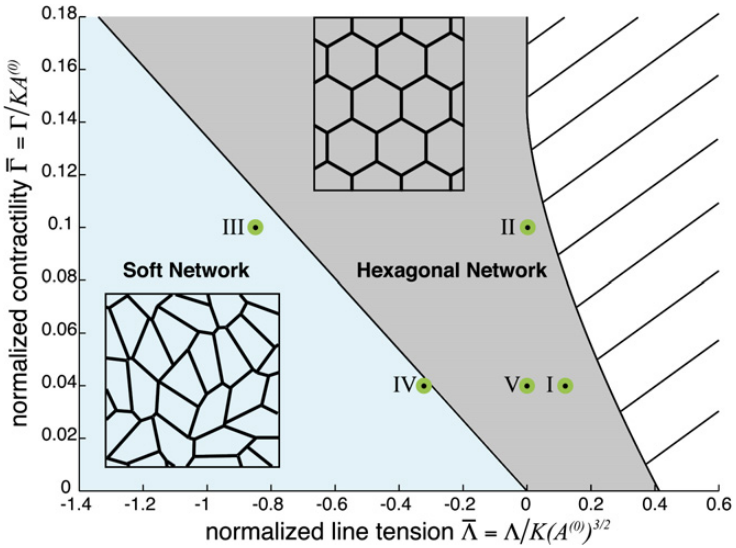
Λ_{ij} sets the competition between contractile tension and adhesion

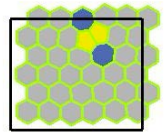
- Phase diagram of ground states:

Grey: hexagonal network with non-zero bulk and shear modulus

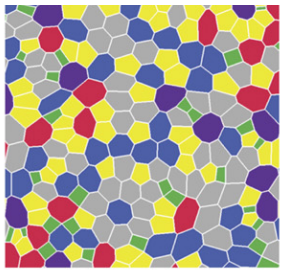
Blue: Soft network is degenerate, ie. different configurations have the same energy and transitions between configurations do not require work. Requires negative normalized line tension. All cells have the same area $A^{(0)}$ and junctions have length $L_0 = -\Lambda/2\Gamma$

This network exhibits a fluid state.

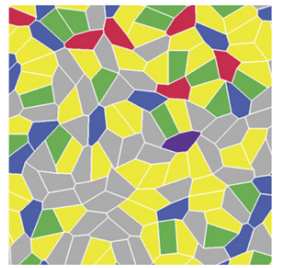


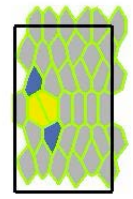


case I



case III





R. Farhadifar et al, S. Eaton and F. Jülicher. *Current Biology* 17, 2095–2104 (2007)

Adhesive cells can exhibit fluidity

Density-independent rigidity transition: vertex model

- epithelial packing describe by equilibrium model (Vertex Model in 2D)

$$E_i = K_{A_i}(A_i - A_{i0})^2 + \xi_i P_i^2 + \gamma_i P_i$$

$$E_i = K_{A_i}(A_i - A_{i0})^2 + \xi_i (P_i - P_{i0})^2$$

Where $P_{i0} = -\gamma_i/(2\xi_i)$ is the effective target perimeter

Total mechanical energy of tissue with N cells with identical properties

$$\varepsilon = \frac{1}{K_A A_0^2}\sum_{i=1}^{N} E_i = \sum_{i=1}^{N}\left[(\tilde{a}_i - 1)^2 + \frac{(\tilde{p}_i - p_0)^2}{r}\right]$$

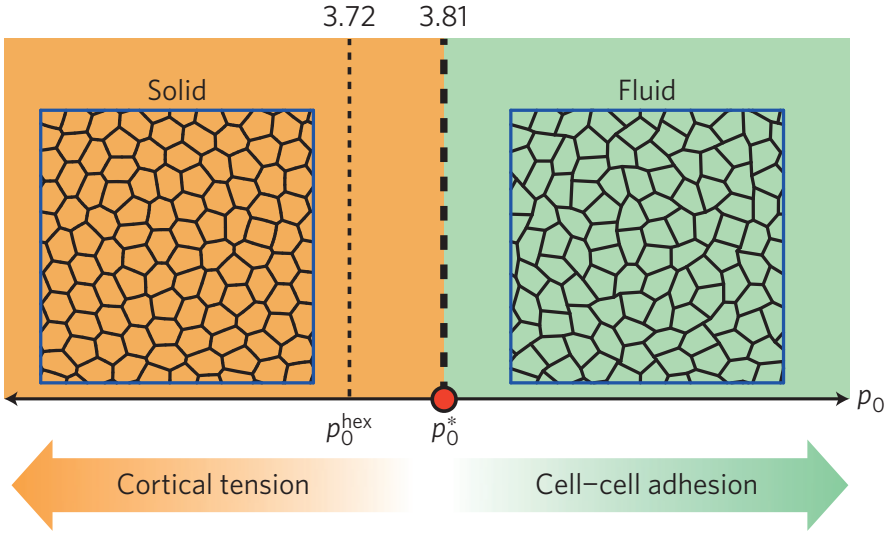
Rescaled shape functions: $\tilde{a}_i = A_i/A_0 \quad \tilde{p}_i = P_i/\sqrt{A_0}$

Inverse perimeter modulus: $r = K_A A_0/\xi$

Target shape factor or preferred perimeter to area ratio:

$$p_0 = P_0/\sqrt{A_0}$$

- Shape factor is order parameter of rigidity transition

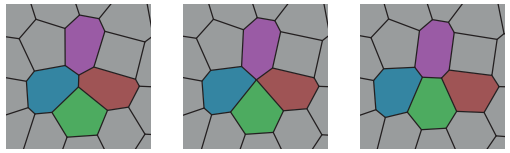


D. Bi et al. and L. Manning. *Nature Physics* 11 1074-1079 (2015)

Adhesive cells can exhibit fluidity

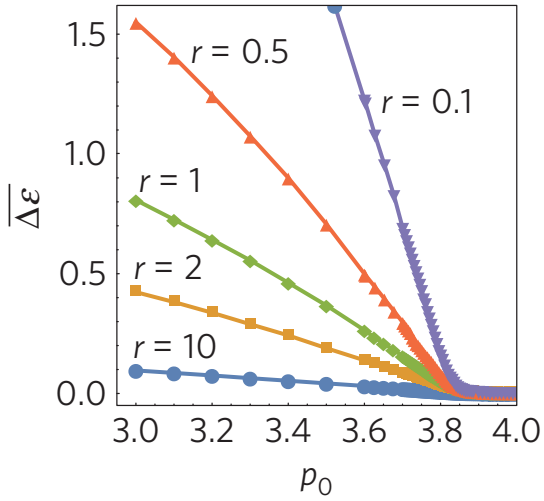
Density-independent rigidity transition: vertex model

- In a confluent adhesive tissues cells are self-propelled and move via cell rearrangements or neighbor exchange (ie. dynamics of adhesive cell contacts)

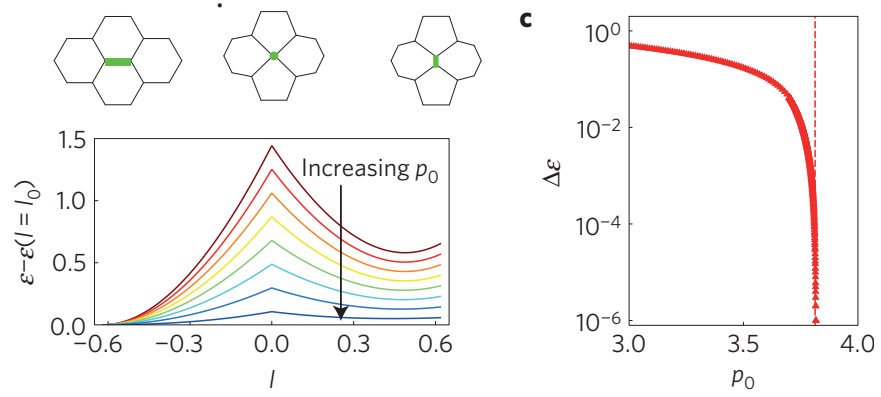


- Energy barrier of rearrangements sets the rate of cell rearrangements (T1 exchange):
- Explore dependency on model parameters, namely shape factor p_o and inverse perimeter modulus r
- Mean energy barrier height: $\overline{\Delta\varepsilon}$

  p_o sets the distance from transition to fluid state: above ~3.81, the energy barrier is zero so rearrangements occur without energy cost. This is the fluid state.
  r sets the magnitude of the energy barrier.



Calculation of p_0^* in a 4 cell T1

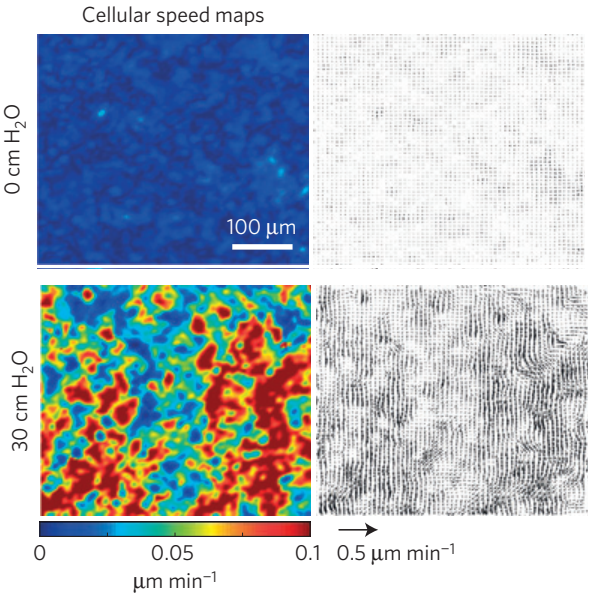


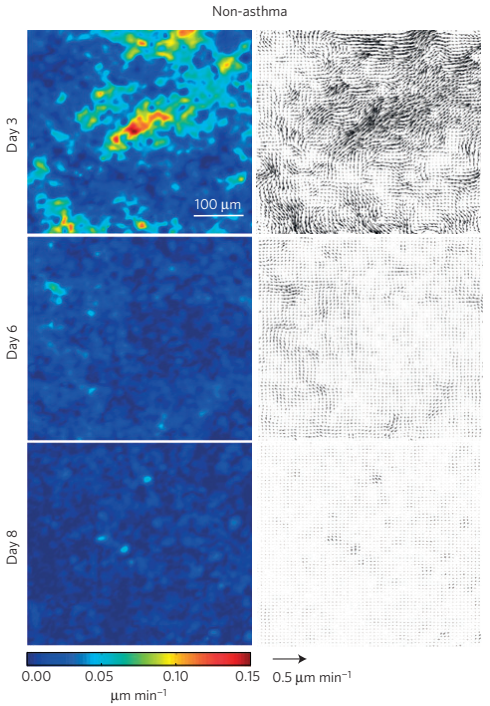
D. Bi et al. and L. Manning. *Nature Physics* 11 1074-1079 (2015)

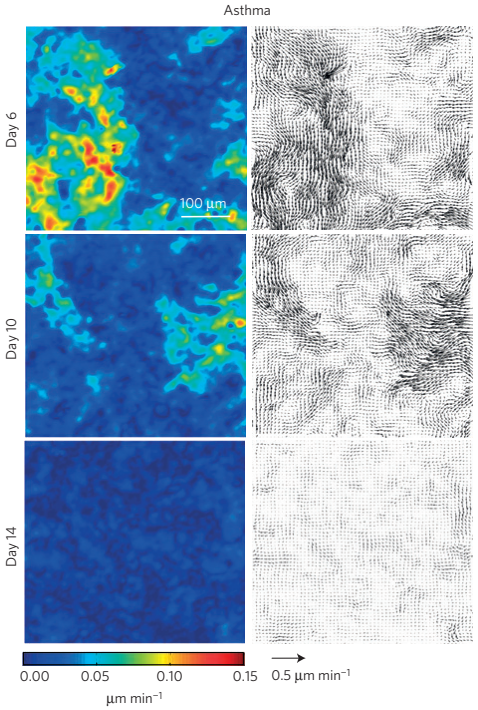
Adhesive cells can exhibit fluidity

Density-independent rigidity transition: experiments

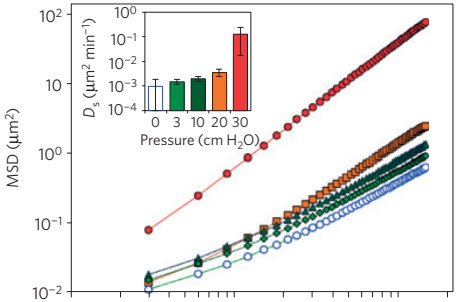
- Airway epithelial cells under compression (that mimic compressive effect of bronchospasm):
- Unjamming transition (increased effective diffusivity of cells)

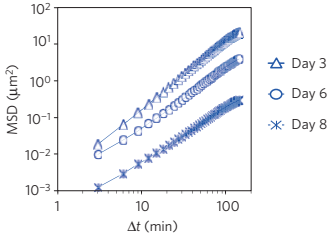






- As cell culture grows, tissue layer jams
- This is delayed in cells from asmathic patients





J-A. Park et al. and J. Fredberg. *Nature Material*, 10:1040-8. (2015) doi: 10.1038/nmat4357.

Adhesive cells can exhibit fluidity and cell flows

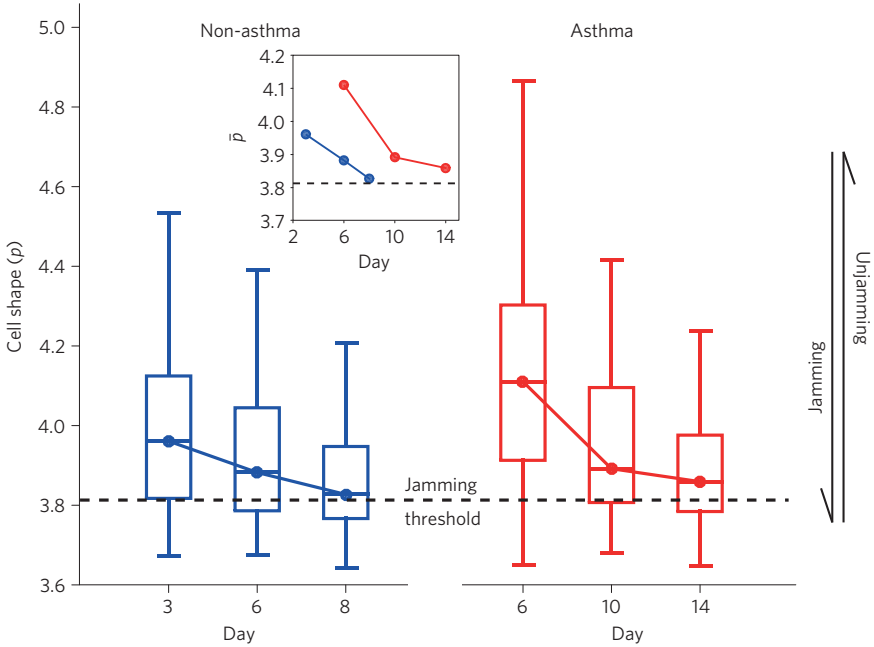
Density-independent rigidity transition: experiments

- Measure of cell shape index as unjamming transition is observed:

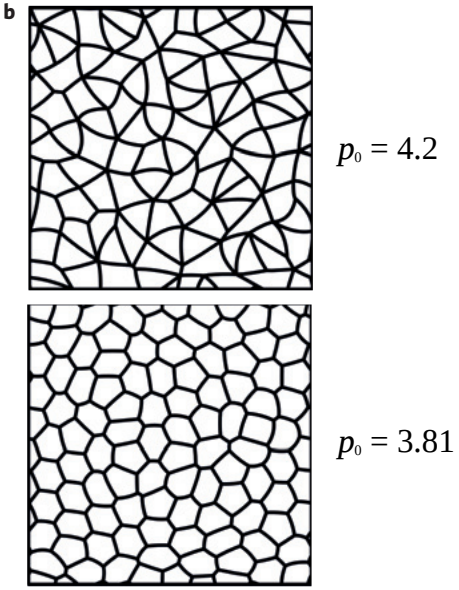
$$p_0 = P_0/\sqrt{A_0}$$

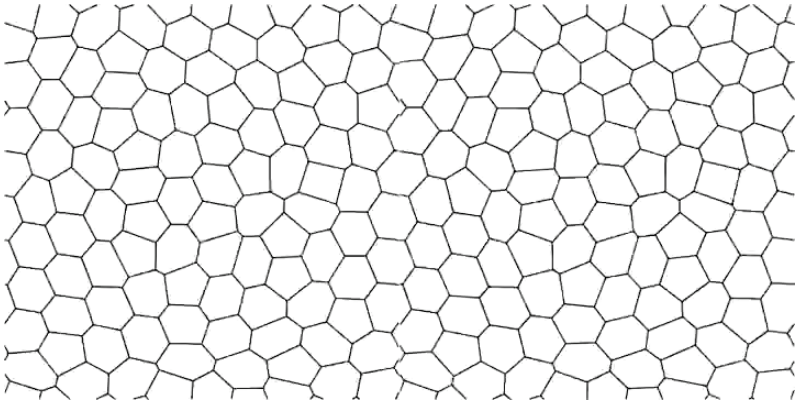
- Shape factor reduces and approaches 3.8 at the transition:

- Experiments



- Simulations





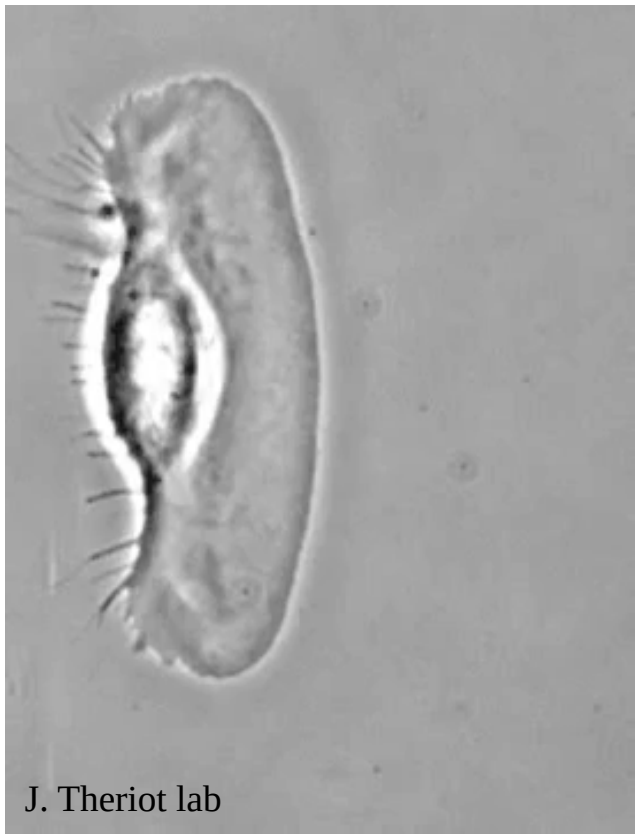
J-A. Park et al. and J. Fredberg. *Nature Material*, 10:1040-8. (2015)
doi: 10.1038/nmat4357.

Conclusions

- Density dependent collective dynamics (critical density sets a preferred speed, eg. Toner Tu model and phenomenology)
- Density independent collective dynamics
- Cell adhesion does not block fluidity in self-propelled population of cells (different in particulate adhesion where adhesion causes jamming such as in colloids)
- Fluidity is associated with collective dynamics
- Collective dynamics requires symmetry breaking

From cell to cell cluster motility: 2D

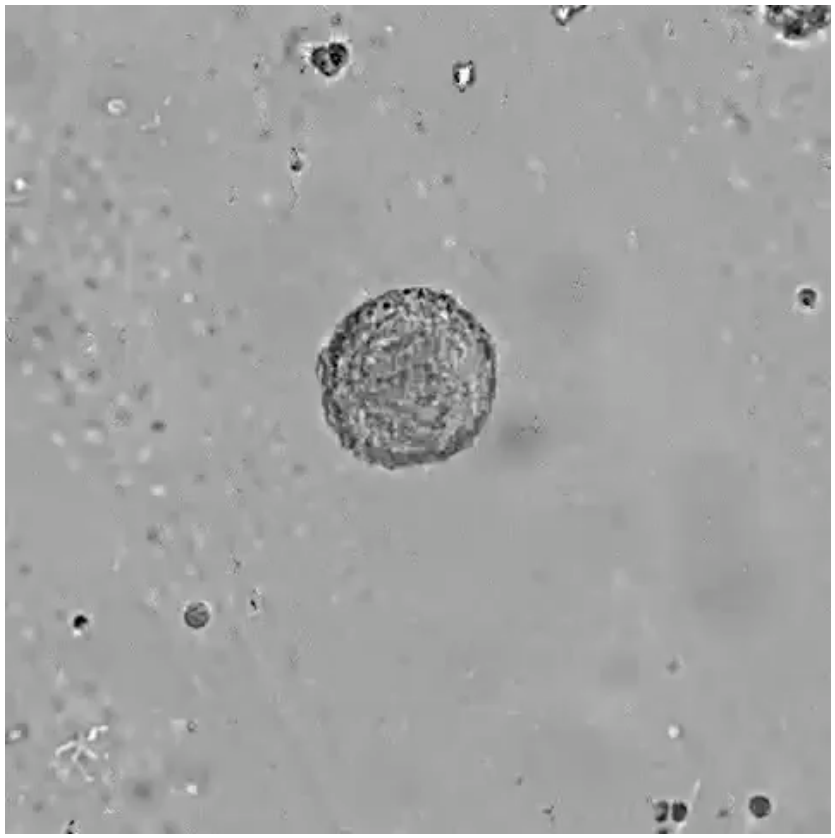
Cell

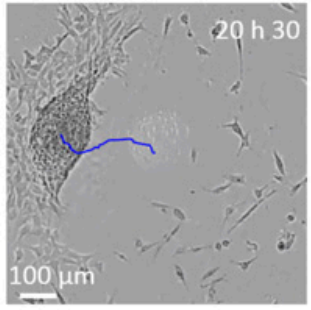


Keratocyte

1 cell — L=35µm, U= 0.7µm.s^{-1}

SupraCell



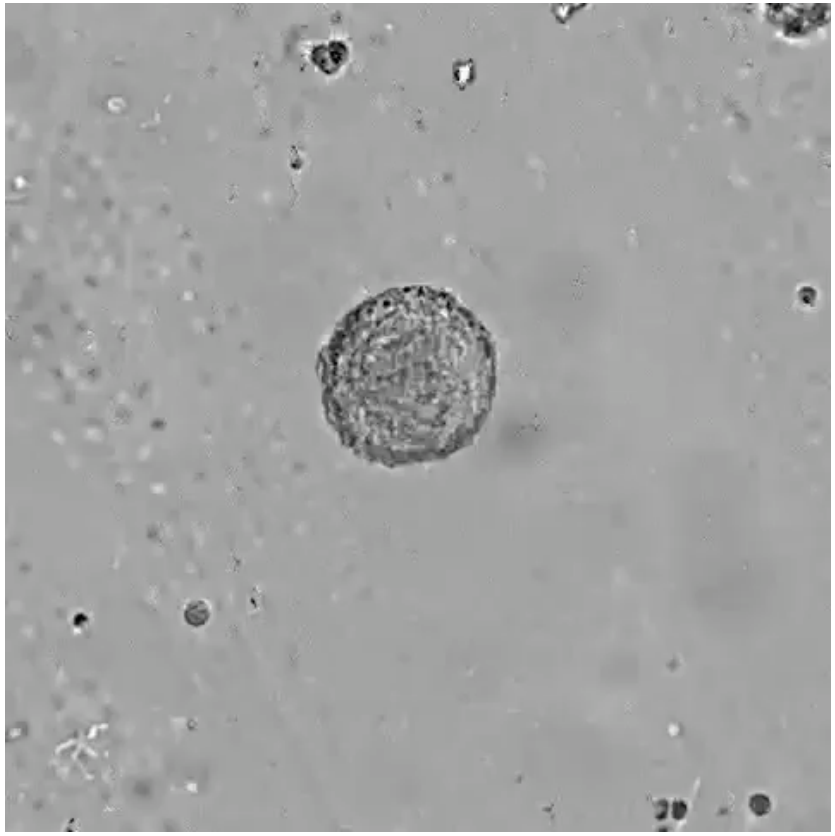


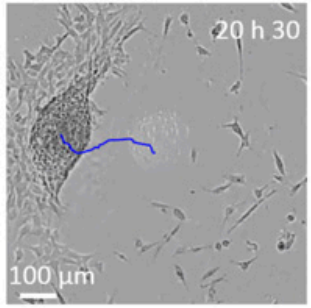
« Keratocyte-like » cell aggregate

>10.000 cells — L>500µm, U= 10^{-2}µm.s^{-1}

G. Beaune et al, …, and F. Brochard-Wyart. *PNAS*, 115: 12927 -12931 (2018)

From cell to cell cluster motility: 2D





J. Theriot lab

Keratocyte

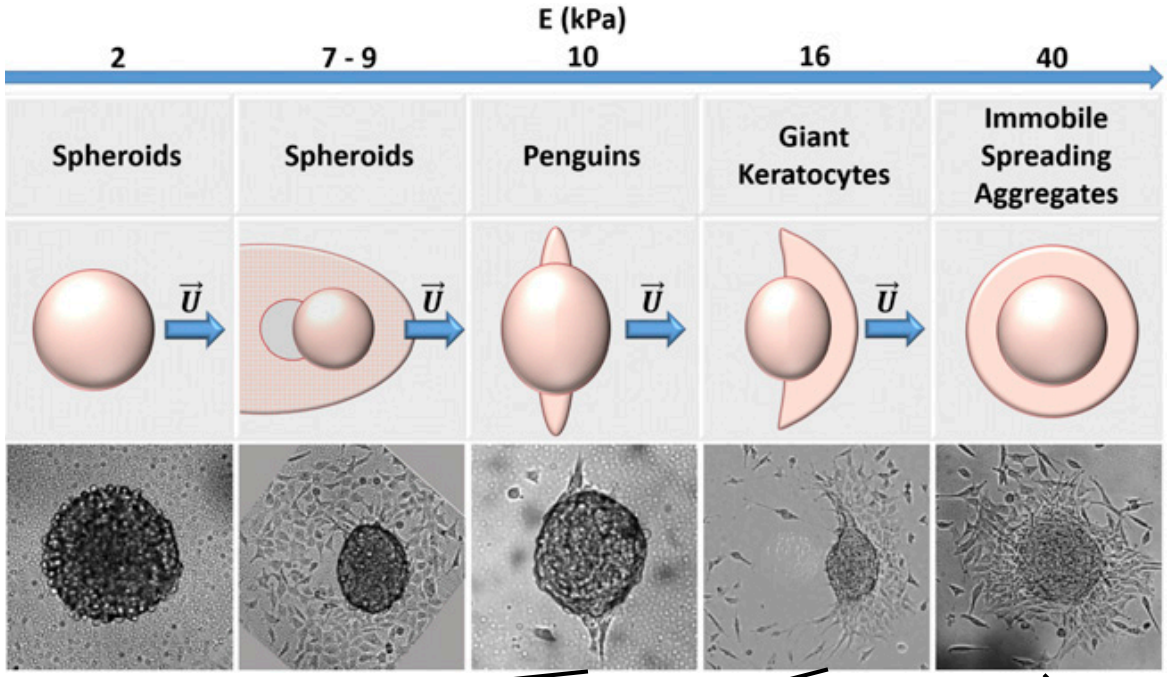
1 cell — L=35µm, U= 0.7µm.s^{-1}

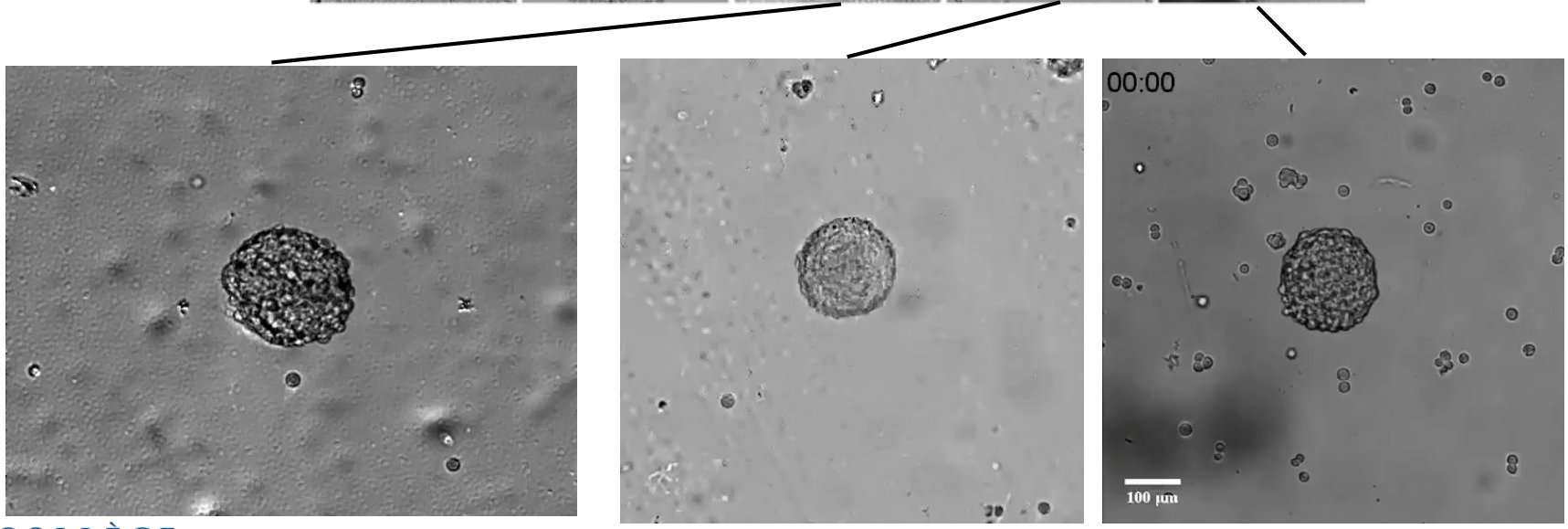
« Keratocyte-like » cell aggregate

>10.000 cells — >500µm, U= 10^{-2}µm.s^{-1}

G. Beaune et al, …, and F. Brochard-Wyart. *PNAS*, 115: 12927 -12931 (2018)

From cell to cell cluster motility: 2D





G. Beaune et al, …, and F. Brochard-Wyart. *PNAS*, 115: 12927 -12931 (2018)

From cell to cell cluster motility: 2D

- Model of cluster spreading as Liquid wetting:

Wetting parameter: difference between substrate adhesion and cell-cell adhesion energy

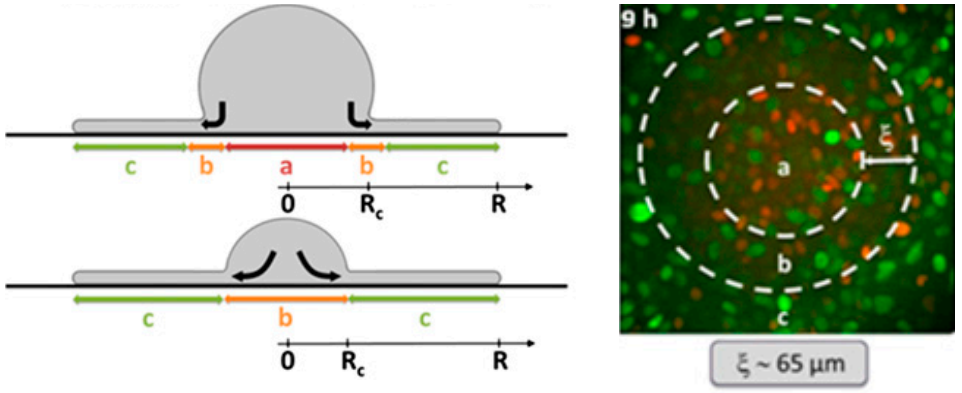
$$S = W_{CS} - W_{CC}$$

Diffusive expansion of cell: $R^2 = D\ t$

Diffusion coefficient: depends on spreading velocity and cluster size: $D = V^* R_a$

Spreading velocity is ratio between wetting energy and tissue viscosity $V^* = S/\eta$

>Therefore cell diffusion depends on substrate stiffness via the wetting parameter



Permeation at boundary between cell cluster and cell layer

G. Beaune et al, …, and F. Brochard-Wyart. *PNAS* 111: 8055–8060 (2014)

- Cell cluster motility:

Force balance: net driving force in layer $\boldsymbol{F}$ and frictional forces across area A associated with cell velocity U

$$\boldsymbol{F} = \oint S(s)\boldsymbol{n}ds = Ak\boldsymbol{U},$$

G. Beaune et al, …, and F. Brochard-Wyart. *PNAS*, 115: 12927 -12931 (2018)

From cell to cell cluster motility: 2D

- Cell cluster motility:

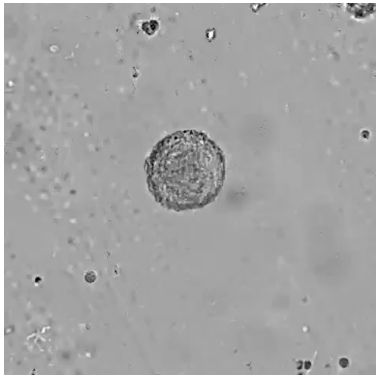
  - 1D model: asymmetry in wetting forces across cluster
    Difference between advancing and receding fronts.

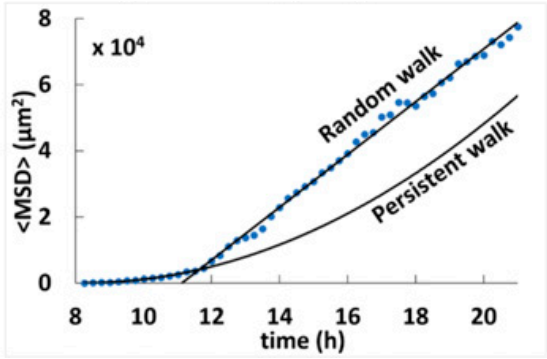
$$S_a - S_r \approx kRU,$$

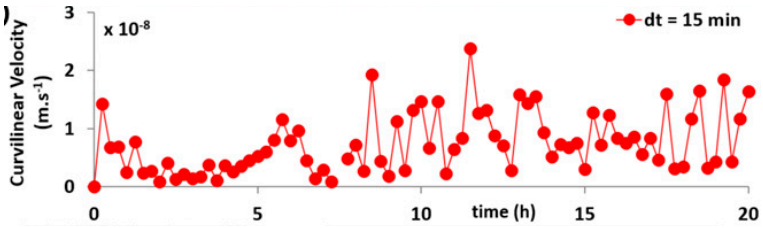
On stiff substrates, spreading is isotropic around cell cluster

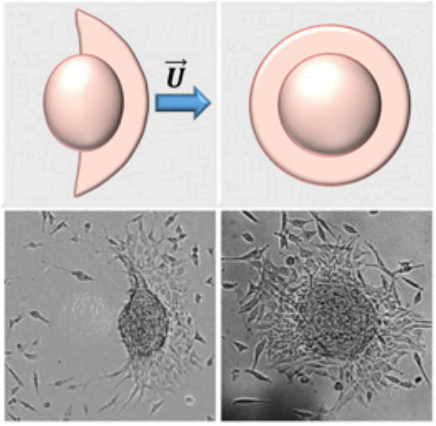
On softer substrates, spontaneous holes arise in layer near cluster edge because cell-cell adhesion cannot hold tension arising from traction forces at the edge (cell-cell adhesion is itself reinforced by substrate stiffness)

Once a hole is formed, symmetry breaking arises $S_r \sim 0$







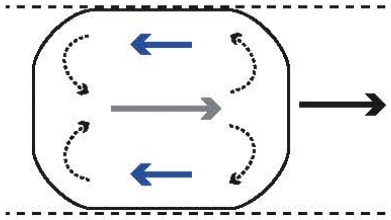


- Mechanism for persistance of motility:
— modification of substrate and wettability (« reactive » droplet/cluster)
— cell polarisation in response to forces (« active » droplet/cluster)

G. Beaune et al, …, and F. Brochard-Wyart. *PNAS*, 115: 12927 -12931 (2018)

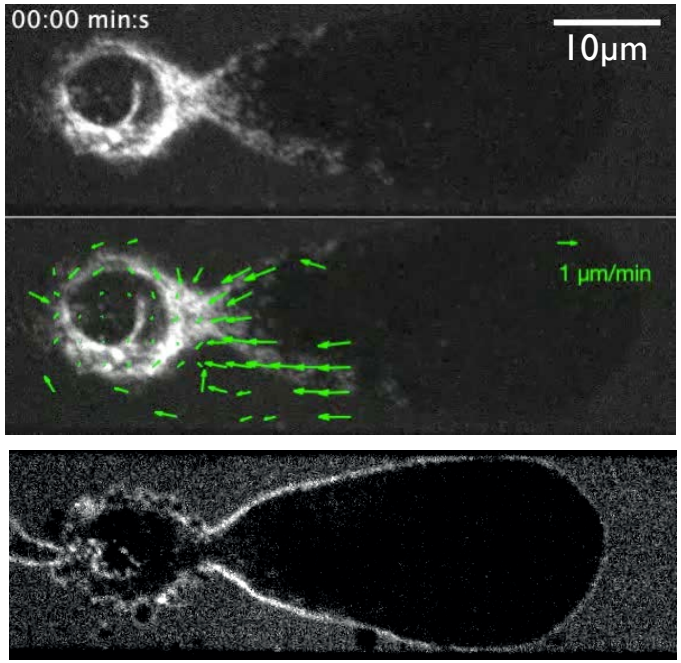
From cell to cell cluster motility: 3D

Retrograde flows
(of myosin or cells)



Cell

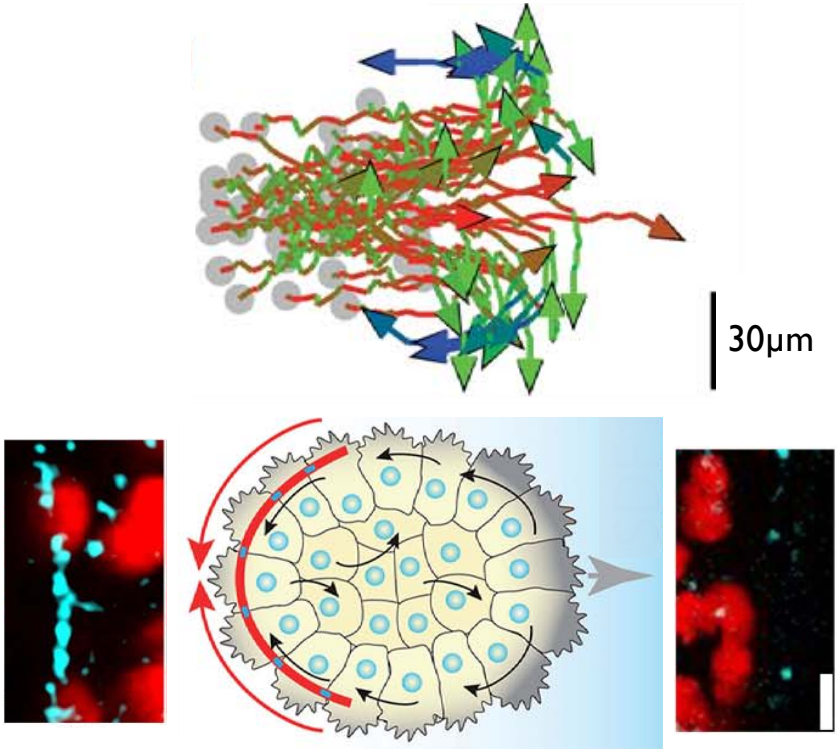
Myosin flow



Myosin polarity

1 cell — L=30µm, U= 0.7µm.s^{-1}

SupraCell



100 cells — L=120µm, U= 2-3$10^{-2}$µm.s^{-1}

DL. Pagès et al., and R. Voituriez, M. Piel, F. Jaulin *Sci. Adv.* **8**, eabp8416 (2022)

Shellard et al., and R. Mayor. *Science* 362, 339–343 (2018)

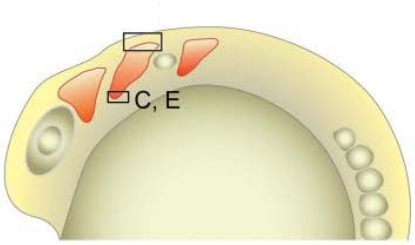
From cell to cell cluster motility: 3D

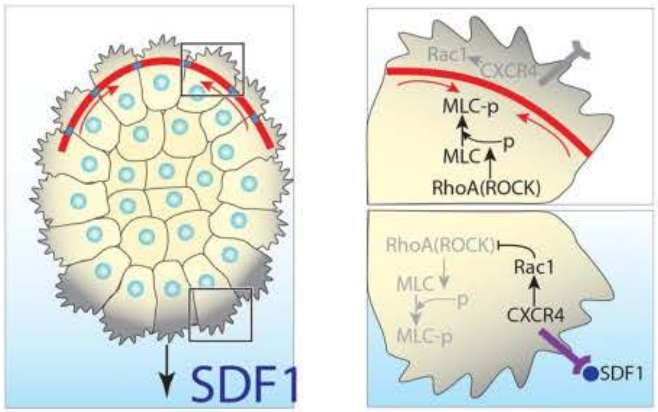
- Clusters of neural crest cells exhibit global front-back polarity:

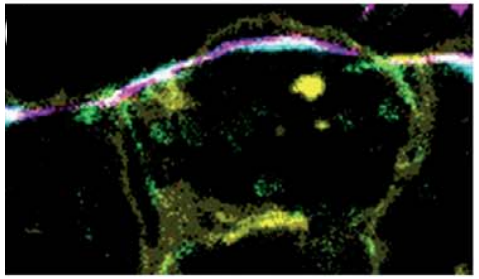
—Myosin is enriched at the rear of the cluster

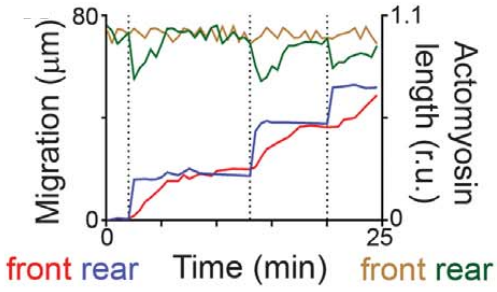
—Cell contractions at the rear are induced by a gradient of SDF1

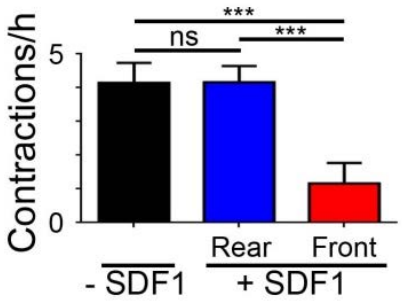
—Actomyosin cell contractions coincide with steps of forward movement of the rear







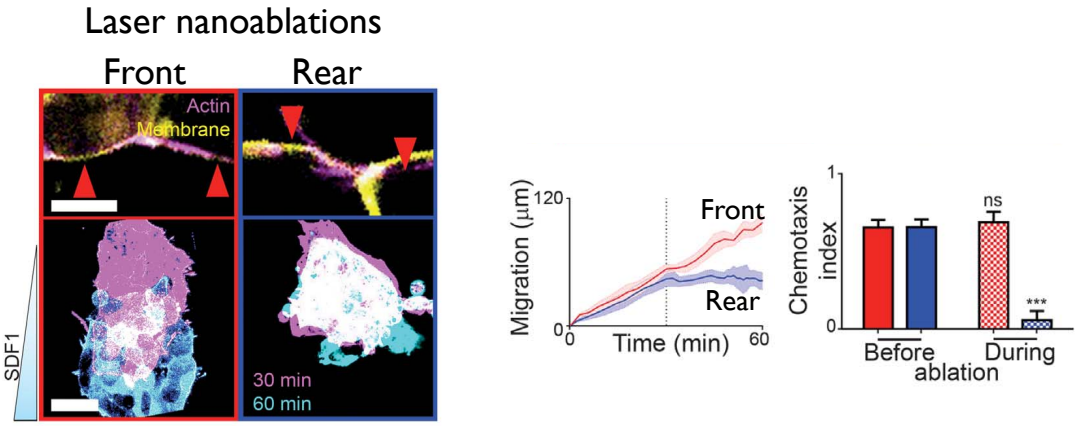




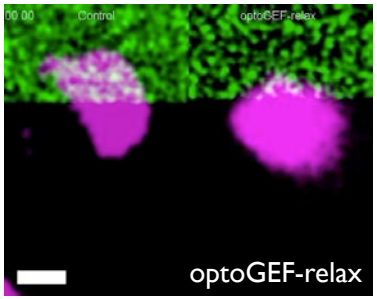
Shellard et al., and R. Mayor. *Science* 362, 339–343 (2018)

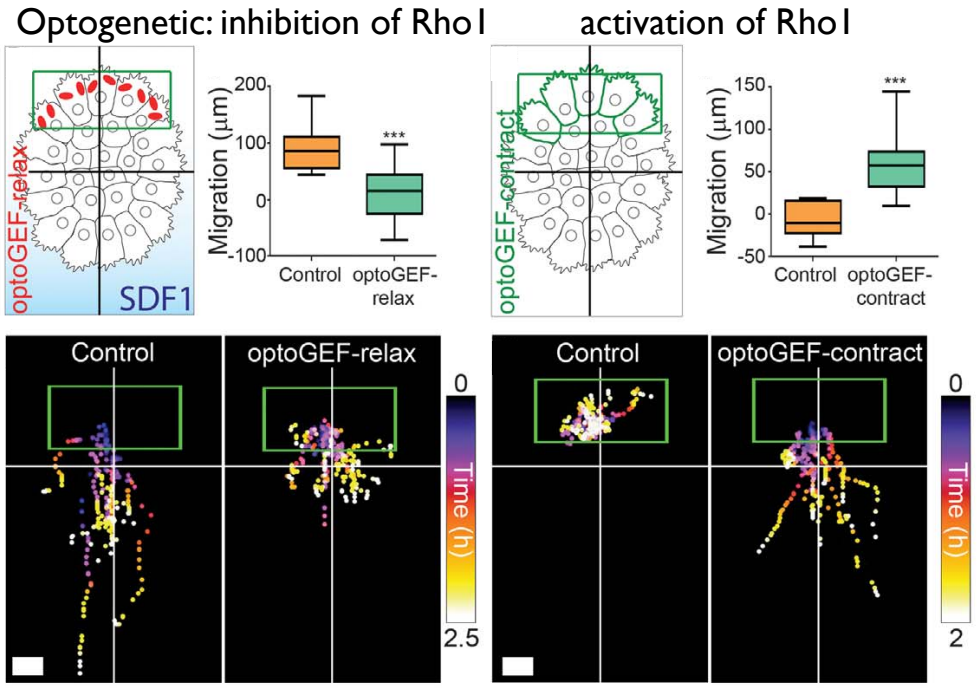
From cell to cell cluster motility: 3D

- Posterior contractility induced by the actomyosin supracellular cortex is required for forward cell cluster migration



- Polarized contractility is sufficient to induce cluster migration:
- Activation of contractility at the rear of the cell cluster rescues motility in the absence of an SDF1 gradient





Shellard et al., and R. Mayor. *Science* 362, 339–343 (2018)

From cell to cell cluster motility: 3D

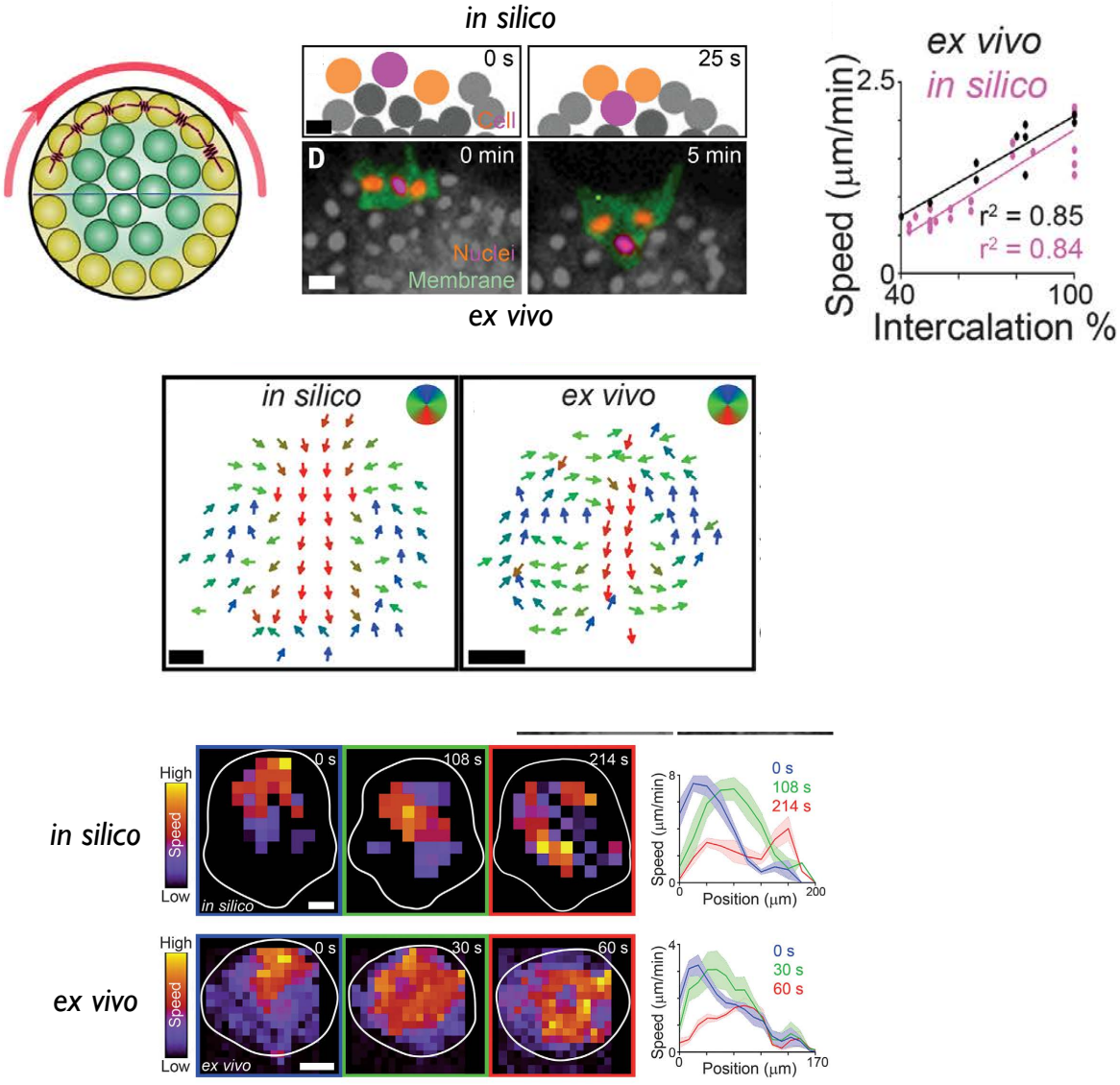
- Computational model:

Soft core repulsion, midrange attraction and periodic attraction forces to model MyosinII contraction.

In silico:

- Cell intercalation at the rear that correlates with cluster migration speed.
- Forward cell movement at the core that propagates as a wave towards the front
- Retrograde cell flow at the periphery.

Ex vivo: similar observations



Shellard et al., and R. Mayor. *Science* 362, 339–343 (2018)

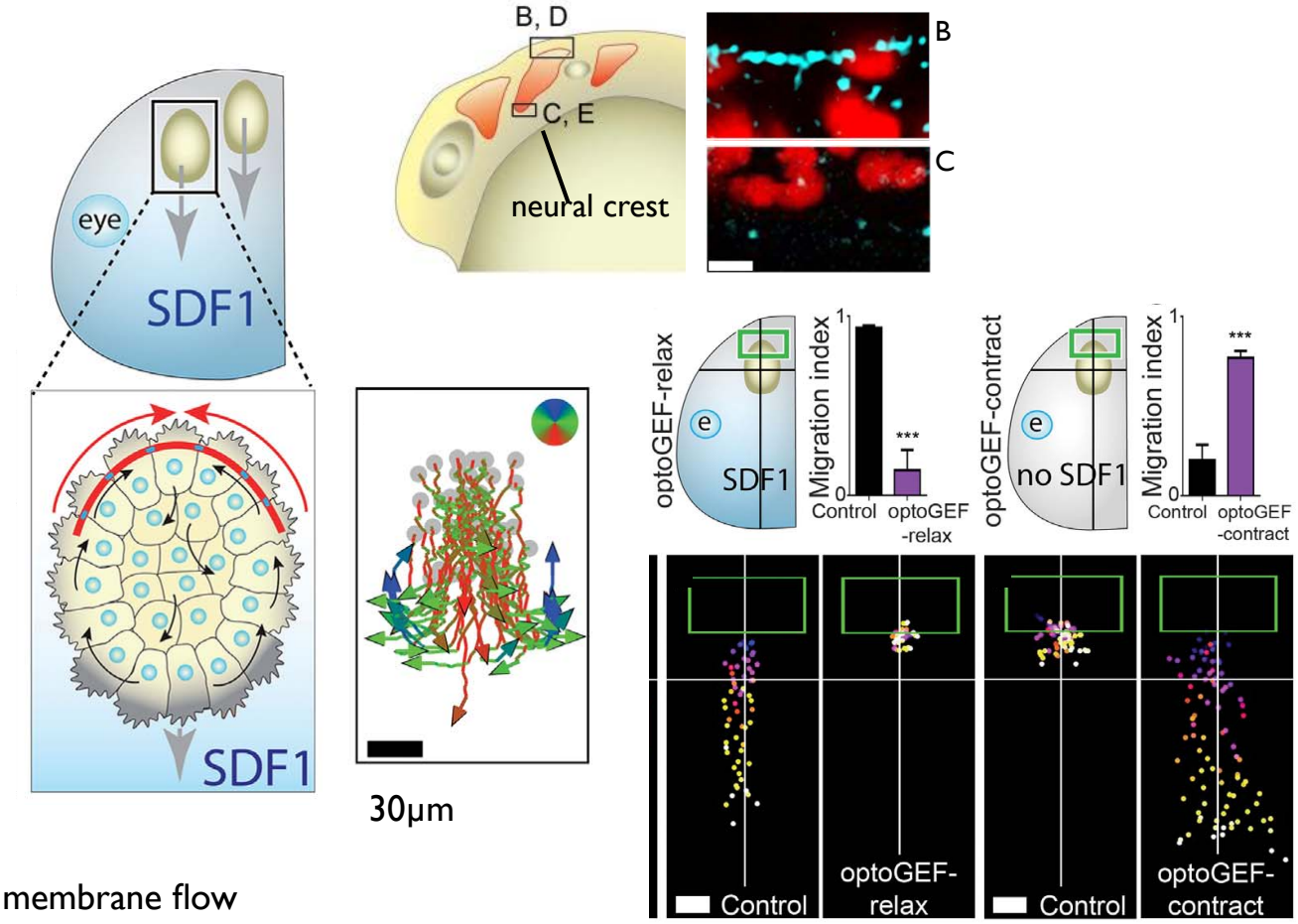
From cell to cell cluster motility: 3D

Rearward flow of cells induced by polarized contractility

- ***In vivo:*** neural crest cell clusters migrate as a whole along a gradient of SDF1
- Collective cell motility results from:

—polarized contractility of cell cluster

—convective flow of cells induced by contractility



Cell scale: Rearward plasma membrane flow generates tangential viscous forces at the cell-liquid interface to drive the cell forward

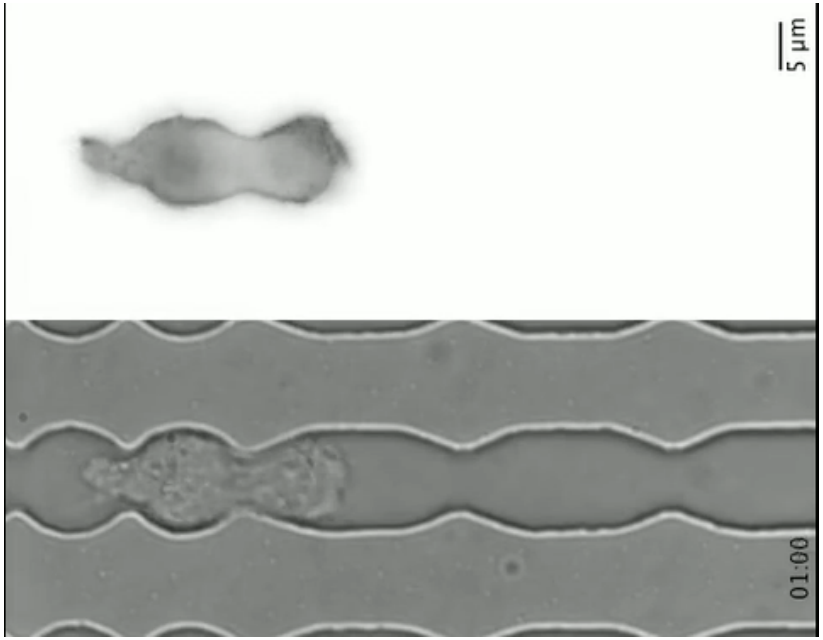
P. R. O'Neill et al., *Dev. Cell* 46, 9–22.e4 (2018).

Shellard et al., and R. Mayor. *Science* 362, 339–343 (2018)

From cell to cell cluster motility: 3D

Non-adhesive motility under confinement

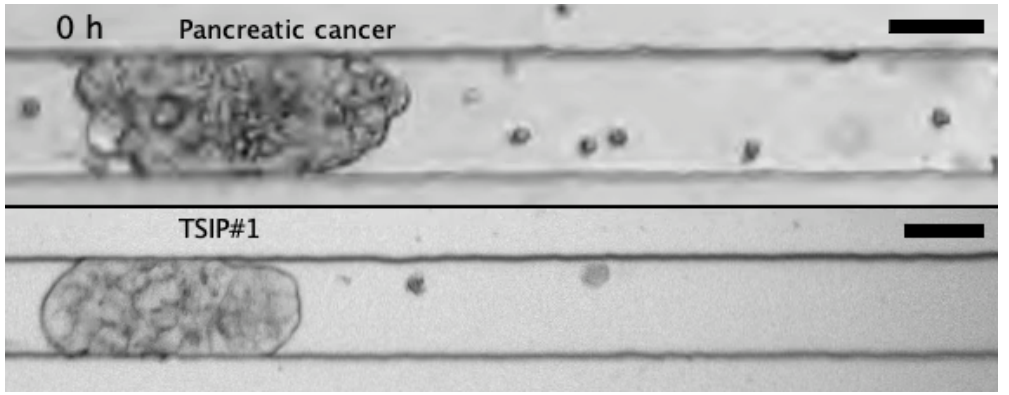
Cell



1 cell — L=20µm, U= 0.7µm.s^{-1}

A. Reversat et al. and R. Voituriez and M. Sixt. *Nature*. 582(7813):582-585. (2020)

SupraCell



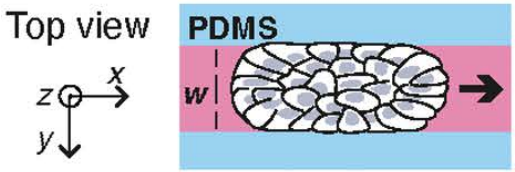
100 cells — L=100µm, U= 1-7 10^{-3}µm.s^{-1}

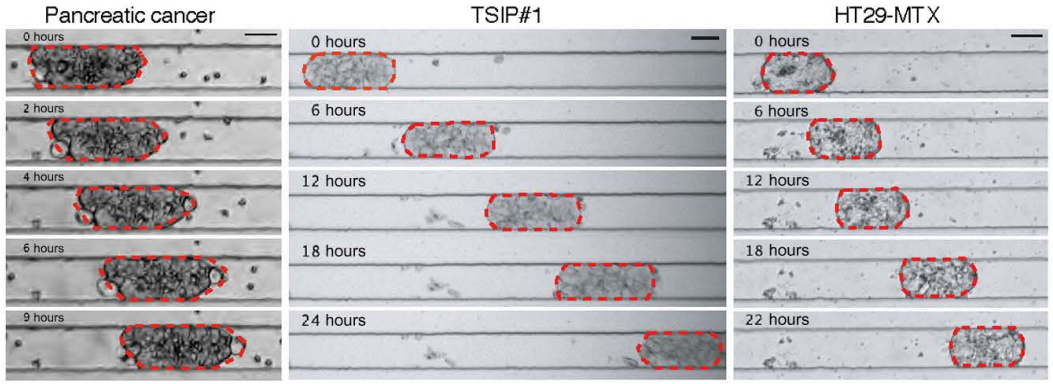
DL. Pagès et al., and R. Voituriez, M. Piel, F. Jaulin *Sci. Adv.* **8**, eabp8416 (2022)

From cell to cell cluster motility: 3D

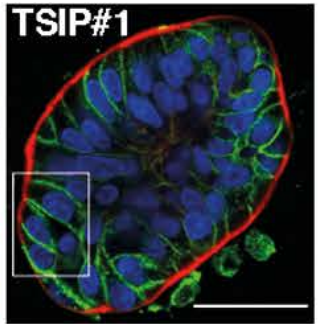
- Clusters of cancer cells of many origins form clusters with the apical surface outside

  (tumor spheres with inverted polarity - TSIP)

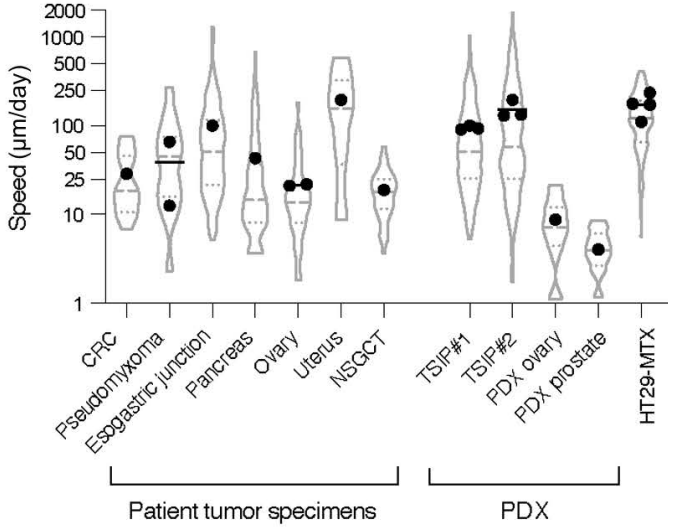




- Cancer cell clusters in microchannels coated with PEG exhibit spontaneous motility.
- Slow motility: 20-25µm/hour



F-actin ß1-intergrin



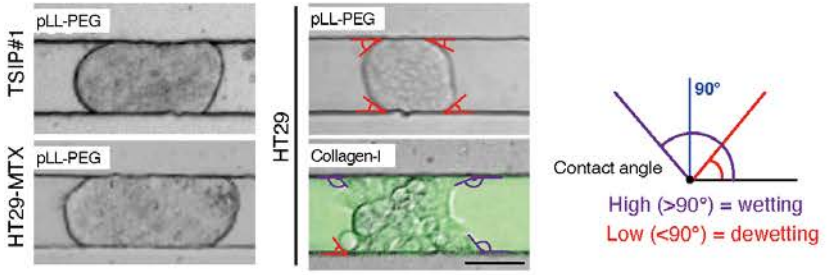
DL. Pagès et al., and R. Voituriez, M. Piel, F. Jaulin *Sci. Adv.* **8**, eabp8416 (2022)

From cell to cell cluster motility: 3D

Non-adhesive motility of cell clusters in micro channels

- Dewetting behavior:

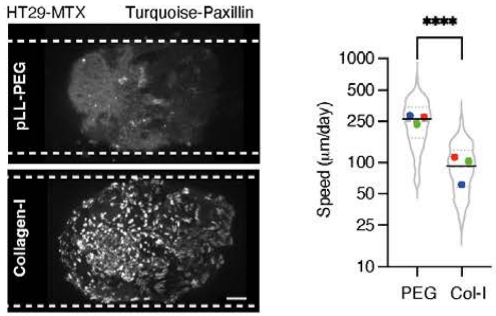
  Clusters of cancer cells show no wetting behavior on PEG, but do so on collagen



- Hallmarks of non-adhesive motility under confinement

—There is no focal adhesion mediated by Integrins on PEG substrates.

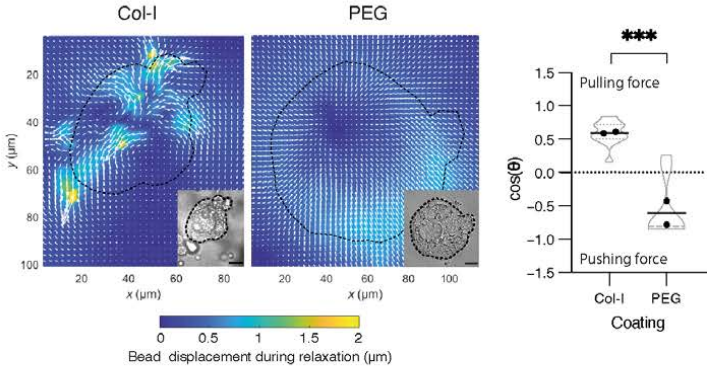
—No traction forces on PEG



- Role of friction in motility:

—Non specific friction mediated by BSA increases speed

—Integrin mediates non specific friction

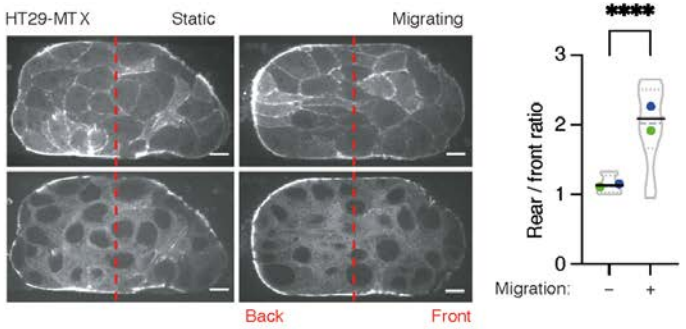


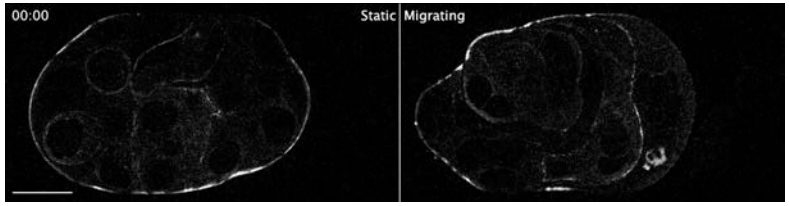
DL. Pagès et al., and R. Voituriez, M. Piel, F. Jaulin *Sci. Adv.* **8**, eabp8416 (2022)

From cell to cell cluster motility: 3D

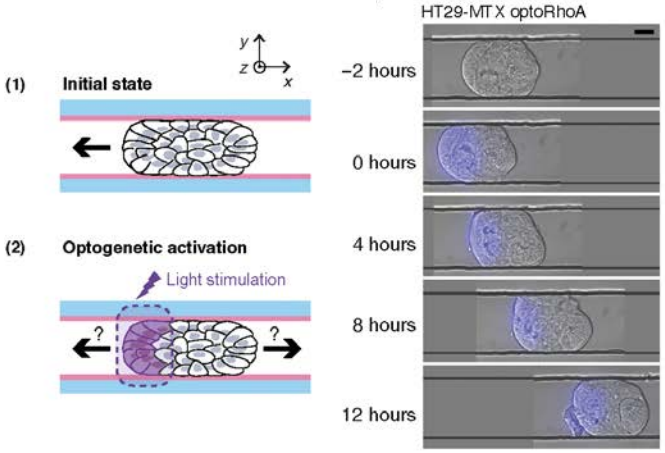
Polarized contractility of cell clusters drive collective motility

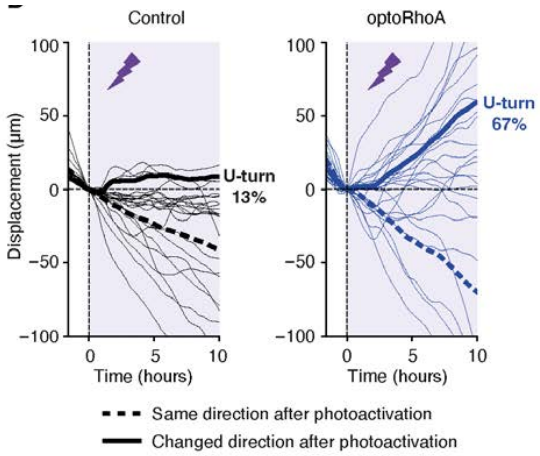
- Polarized distribution of actomyosin contractility ***correlates with*** cluster motility



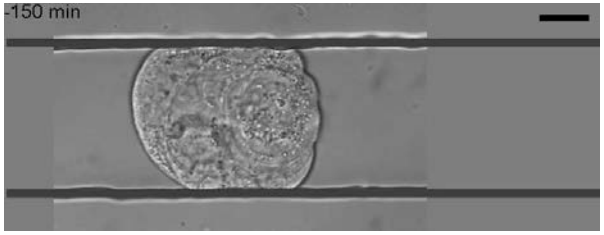


- Polarized distribution of actomyosin contractility is ***required for*** collective motility





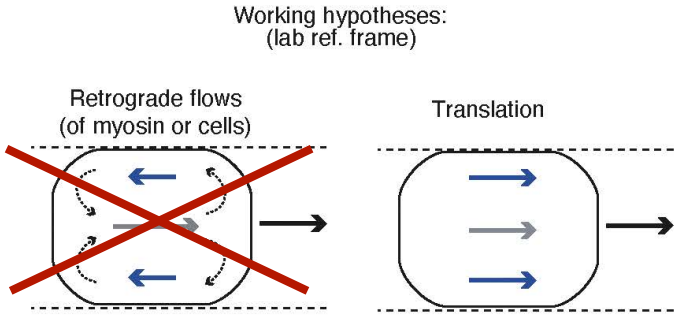
Optogenetic activation of Rho1-MyosinII



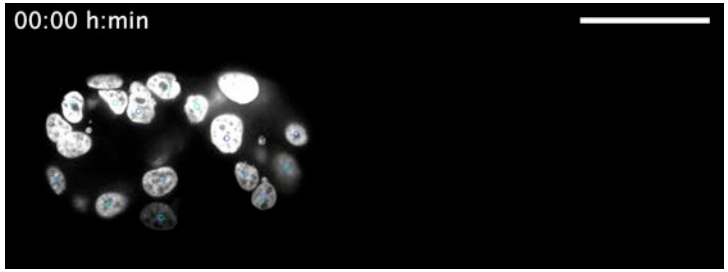
DL. Pagès et al., and R. Voituriez, M. Piel, F. Jaulin *Sci. Adv.* **8**, eabp8416 (2022)

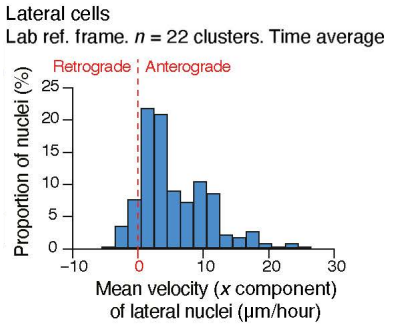
From cell to cell cluster motility: 3D

Cluster motility: Rigid body motility without retrograde cell flows

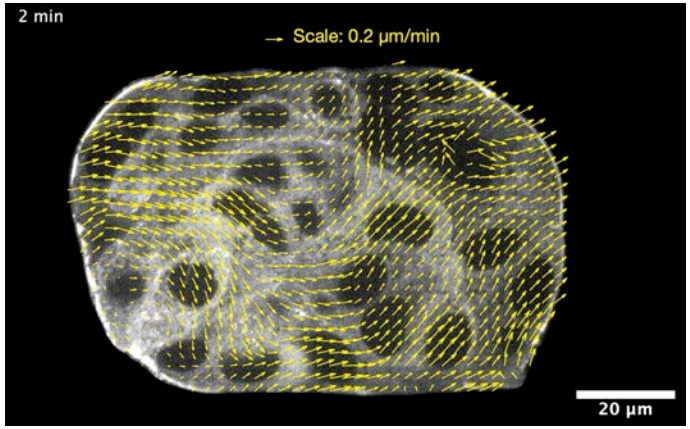


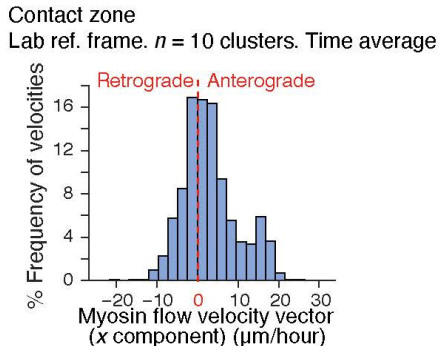
- Forward movement of nuclei





- Forward cytoplasmic flow (using PIV)





DL. Pagès et al., and R. Voituriez, M. Piel, F. Jaulin *Sci. Adv.* **8**, eabp8416 (2022)

From cell to cell cluster motility: 3D

- Model of cluster self-propulsion:
- Jiggling, polarized, elastic solid.

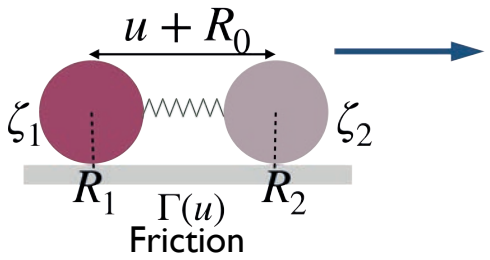
—Gradient of fluctuating contractile stress ζ induced by Myosin

—Induces gradient of stochastic cell deformations

—And asymmetry of frictional forces

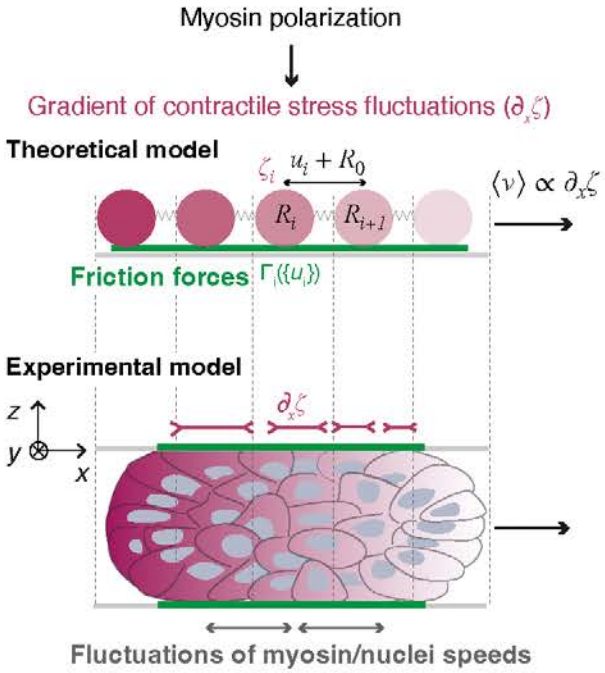
—Hence, cluster movement

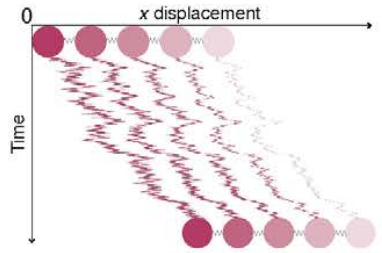
2-cell 1D model:

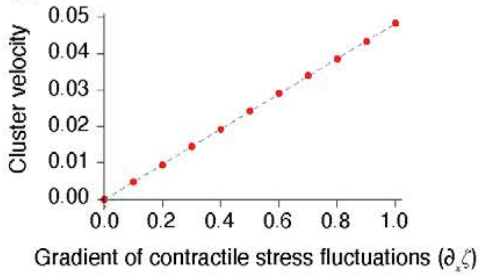


Mobility $M = 1/\Gamma$

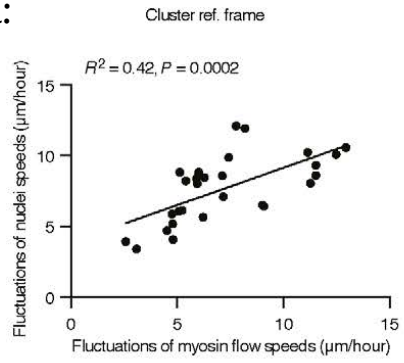
$$\langle v \rangle = \frac{\zeta_2 - \zeta_1}{2} \partial_u M(u)$$

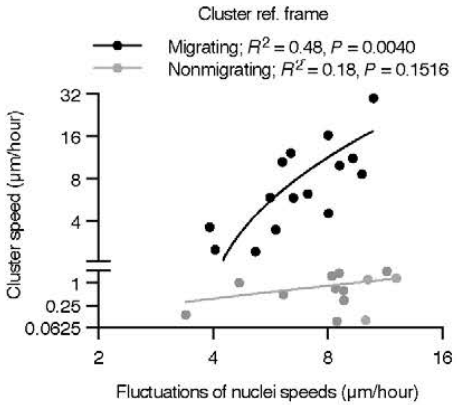






- Data:



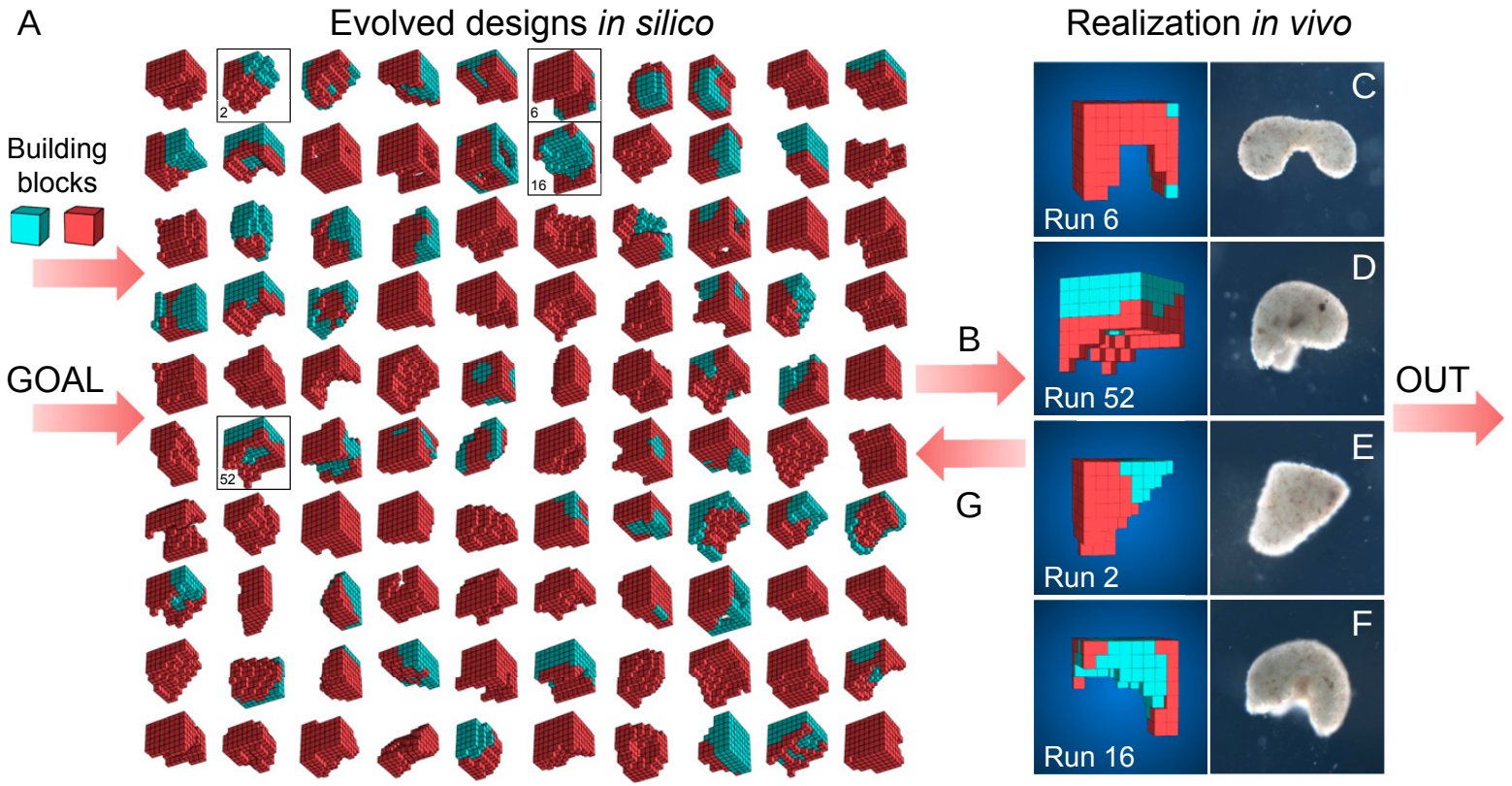


DL. Pagès et al., and R. Voituriez, M. Piel, F. Jaulin *Sci. Adv.* **8**, eabp8416 (2022)

From cell to cell cluster motility: 3D

Engineered biological « robots » and emergence of aggregate motility

Combination of two cell states: passive cells (blue) and fluctuating active contracting cells (red)

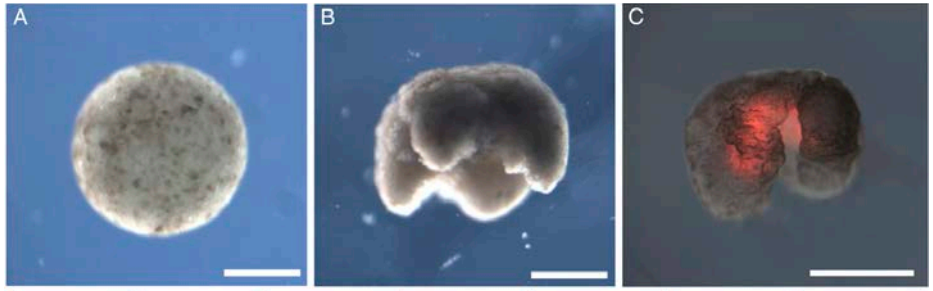


S. Kriegman et al, — M. Levin and J. Bongard. *PNAS*. 117: 1853–1859 (2019)

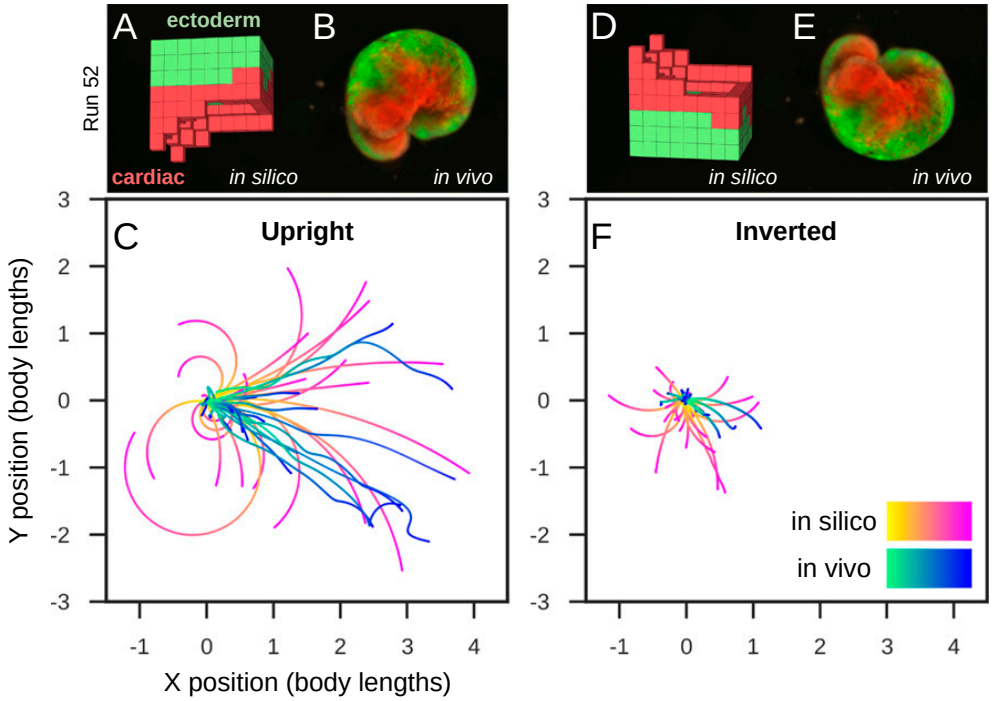
From cell to cell cluster motility: 3D

Engineered biological « robots » and emergence of aggregate motility

Realisation of *Xenopus* engineered robot



Motility is sensitive to orientation of *Xenopus* robot with respect to the substrate



S. Kriegman et al, — M. Levin and J. Bongard. *PNAS*. 117: 1853–1859 (2020)

Summary Course #1

- Density dependent collective dynamics (critical density sets a preferred speed, eg. Toner Tu model and phenomenology)

- Density independent collective dynamics

- Cell adhesion does not block fluidity in self-propelled, actively contracting population of cells (different in particulate adhesion where adhesion causes jamming such as in colloids)

- Fluidity is associated with collective dynamics

- Collective dynamics requires symmetry breaking
- Symmetry breaking is self-organized or induced by external cue

Summary Course #1

Symmetry breaking: From cell to cell collective motility

- Emergent dynamics reflects general principles of underlying mechanics that operate across different scales

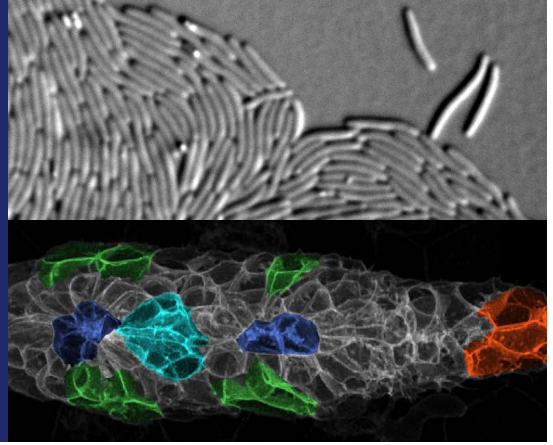
- Questions:
- How do cells generate front back polarity?
- What are the intrinsic, self-organizing mechanisms?
- What are the extrinsic, guidance mechanisms?

Thomas LECUIT

CHAIRE DYNAMIQUES DU VIVANT

Motilité cellulaire collective

15 novembre > 13 décembre



COURS

Les mardis de 10 h 00 à 11 h 30 – Amphithéâtre Guillaume Budé

15 novembre 2022
Introduction : de la cellule à la supra-cellule

22 novembre 2022
Tactisme mécanique — barotaxie

29 novembre 2022
Tactisme mécanique — durotaxie individuelle et collective

6 décembre 2022
Motilité collective au cours du développement

13 décembre 2022
Motilité collective des bactéries

COLLOQUE

Amphithéâtre Maurice Halbwachs

Lundi 19 Juin 2023
Growth and Form

COLLÈGE DE FRANCE 1530

Les cours et séminaires sont gratuits, en accès libre, sans inscription préalable.

Thomas Römer
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