

Week 07: Mechanobiology of cell clusters

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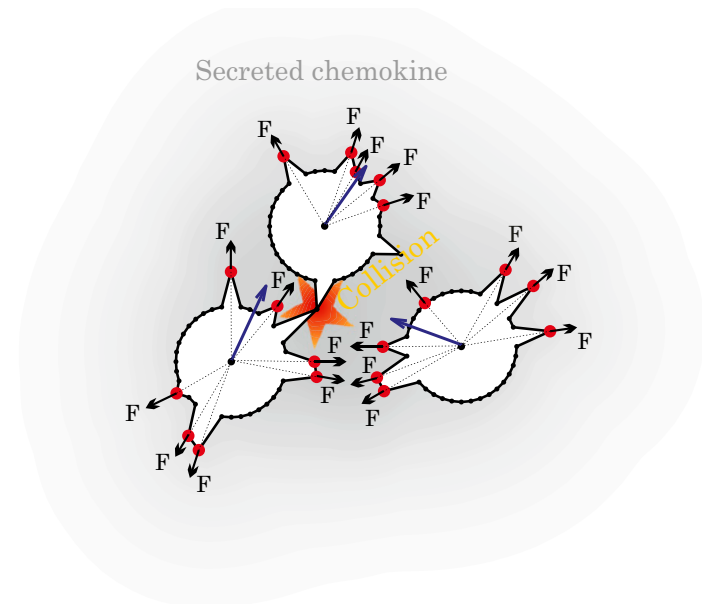
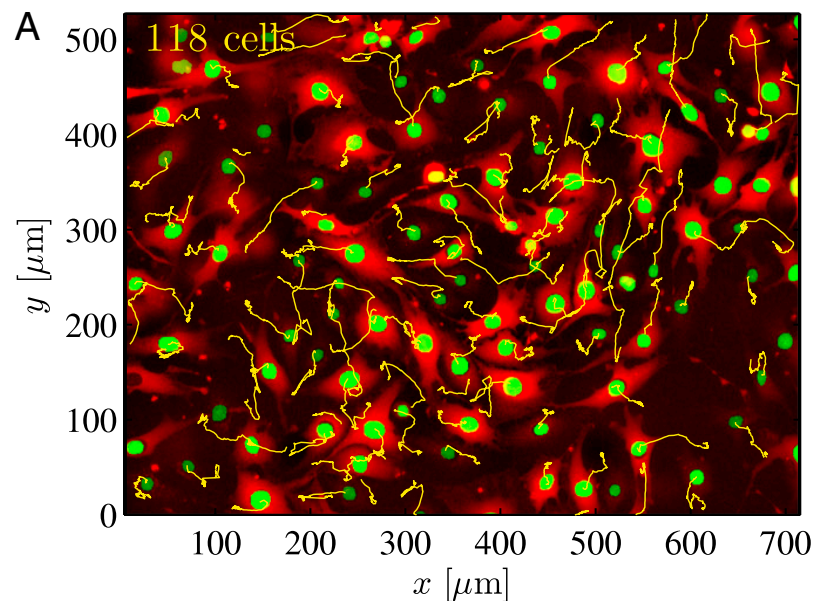
- Cell-cell adhesion
- Epithelial sheets
- Collective cell migration

Contact inhibition

- A tissue culture is often referred to as a colony of cells, thereby implying that a cell can in some circumstances be regarded as a social organism
 - Abercrombie and Heaysman, 1952
- The velocity of the bodily displacement of a cell tends to be affected by its contacts with other cells – Weiss, 1945
- A mutual restriction of movement...

Cellular traffic rules

- Movies of single cells moving around other cells
- Statistical measures: maximum path distance, persistence, contact time, directional autocorrelation, density, etc.
- Pseudopod formation biased by secreted chemicals
- Pseudopod collapse following collisions

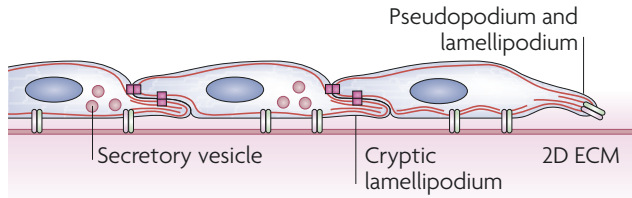


Cell-cell adhesion

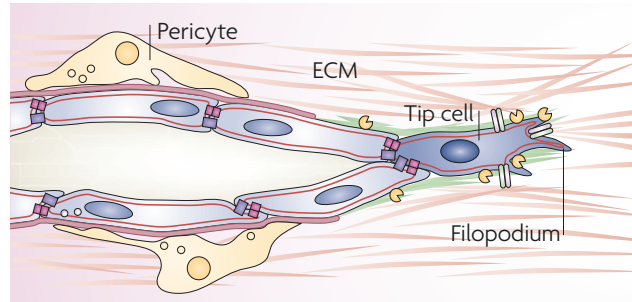
- Cells can recognize identical or different cell types
- They can adhere to each other in a selective manner
- Cell adhesion molecules
 - Cadherin (E-cadherin, N-cadherin, P-cadherin)
- Adherens junctions link the cytoskeleton of adjacent cells via clusters of cadherins
- Adherens junctions grow and strengthen when
 - an external force is applied (outside-in)
 - Actomyosin contraction is enhanced (inside-out)
- Desmosomes are in the same class as adherens junctions: the cytoskeletal anchor is intermediate filaments
- Other cell junctions: tight junctions, gap junctions

Collective cell migration

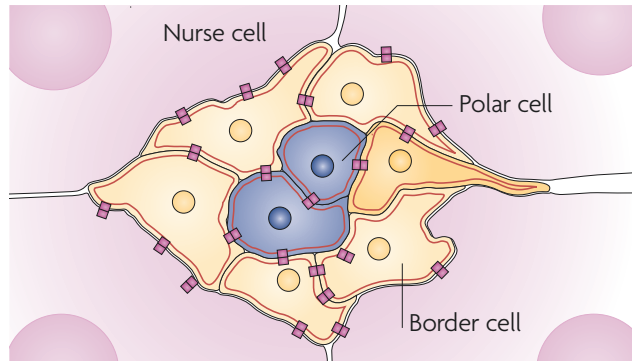
a 2D sheet



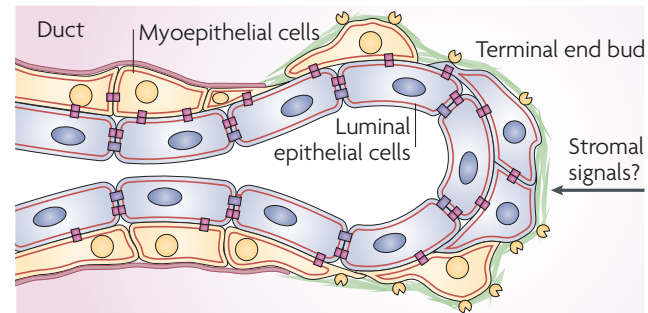
c Vascular sprouting



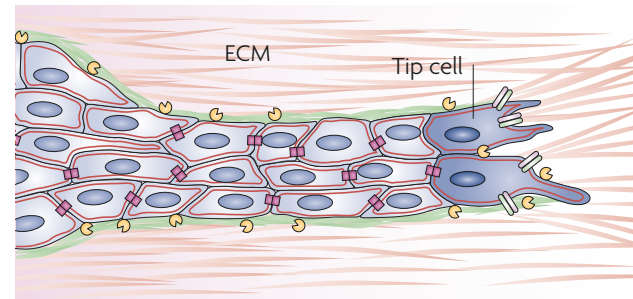
e Border cells



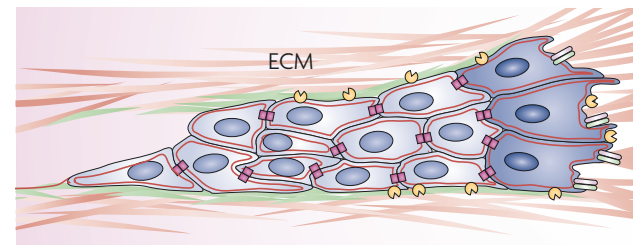
b Branching morphogenesis (mammary gland)



d Multicellular 3D invasion strands

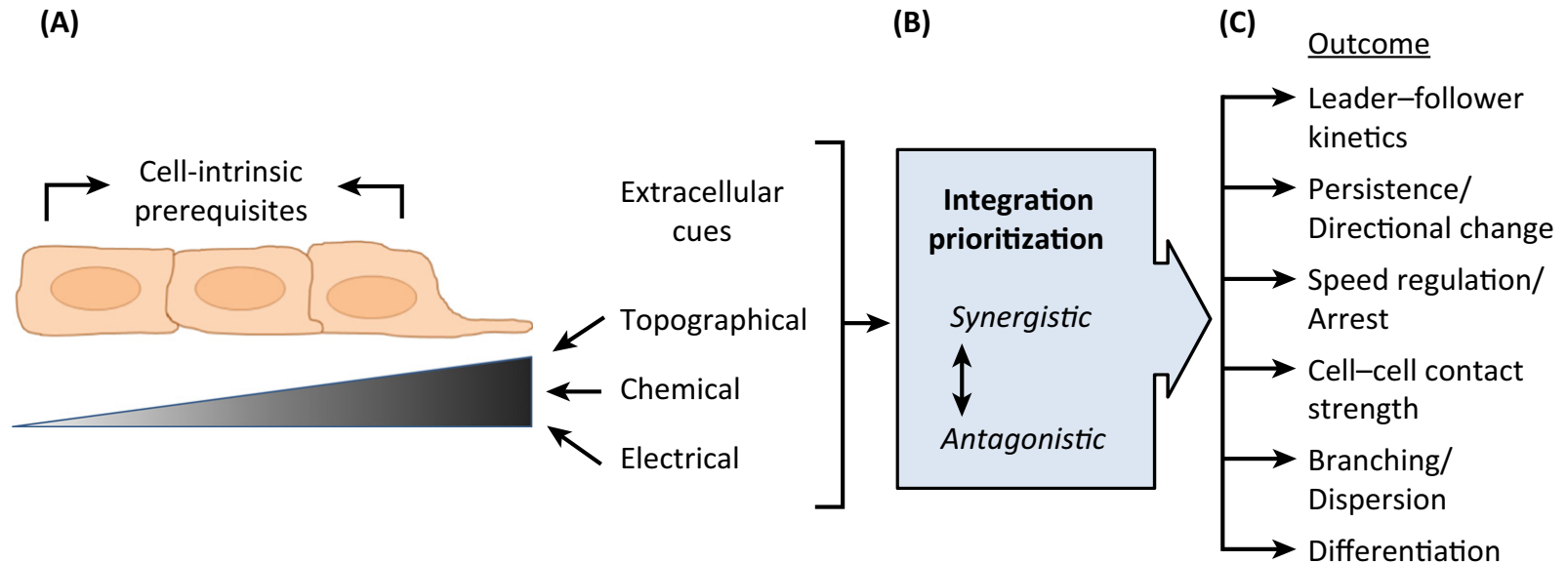


f Detached cluster



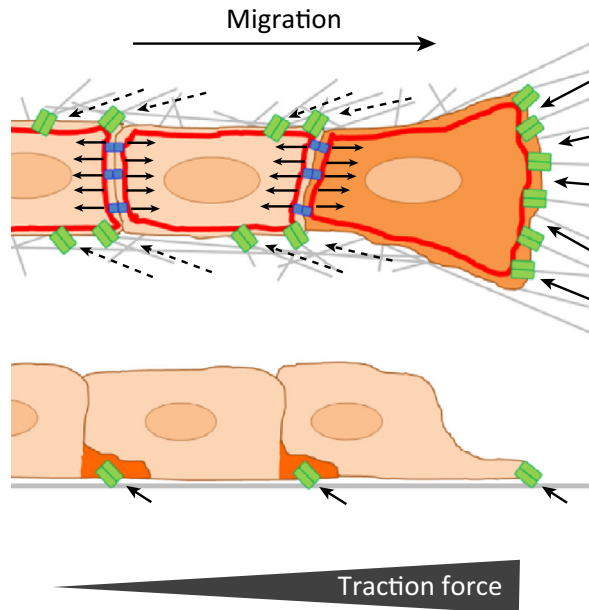
Basement membrane	Adherens junction	Proteolytically remodelled ECM	Integrins ($\beta 1$ and $\beta 3$) in focal contacts	Proteases (MMPs, MTIMMP and UPA)
Cortical F-actin	Tight junction			

Collective cell migration

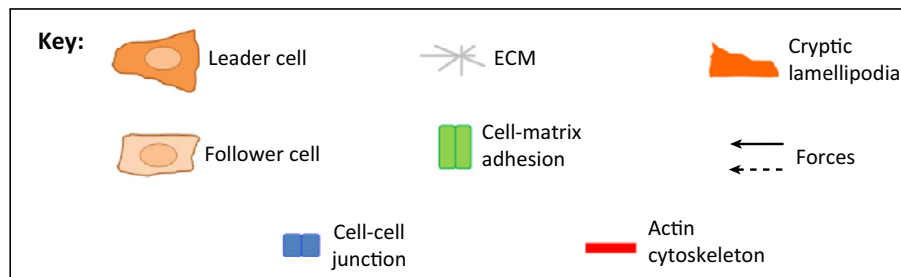
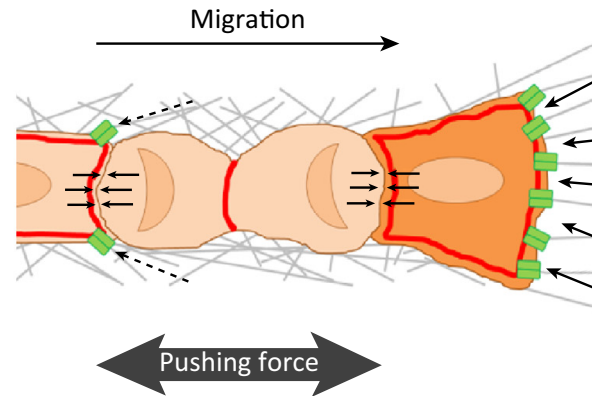


Collective cell migration

(A) Strain generated by leader cells and cryptic lamellipodia



(B) Pushing by dividing cells



Cell-cell adhesion

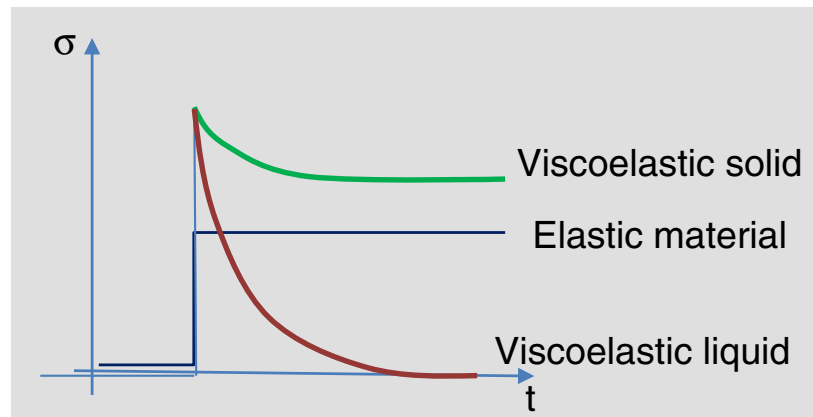
- Intercellular contacts can be
 - transient interactions that regulate collective migration of mesenchymal cells
 - stable junctions that allow epithelia and endothelia to form tissue barriers in the body
- Cell adhesion systems resist tensile forces that would otherwise tend to tear the tissue apart.
- As well as contributing to morphogenesis and tissue integrity, mechanical forces at junctions may provide a mode of cell-cell communication
 - they can propagate very fast and very far

Viscoelasticity

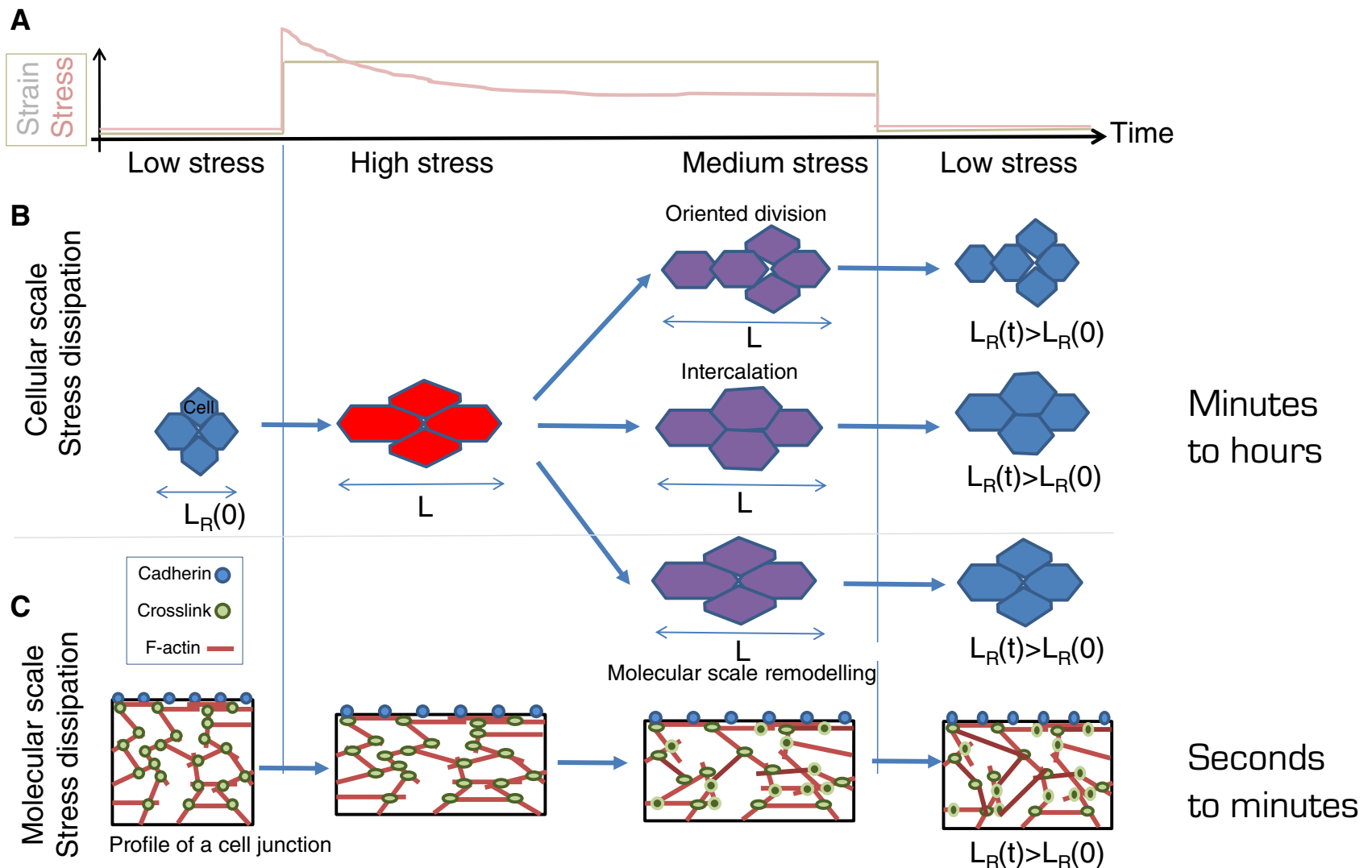
- On second-to-minute time-scales, most biological tissues behave as viscoelastic solids, which signifies that they are elastic-like solids at minute time-scales but also display viscous properties at shorter time-scales.
- When strain rates are **low**, loading is **quasi-static**, signifying that only the tissue's **elastic** properties are solicited.
- When strain rates are **high**, the tissue's **viscous** properties are solicited in addition to its elastic properties, leading to additional stress. As a result, tissues are subjected to **transiently higher peak stresses** when deformed at **high strain rate** and adhesions must be able to withstand this.

Viscoelasticity

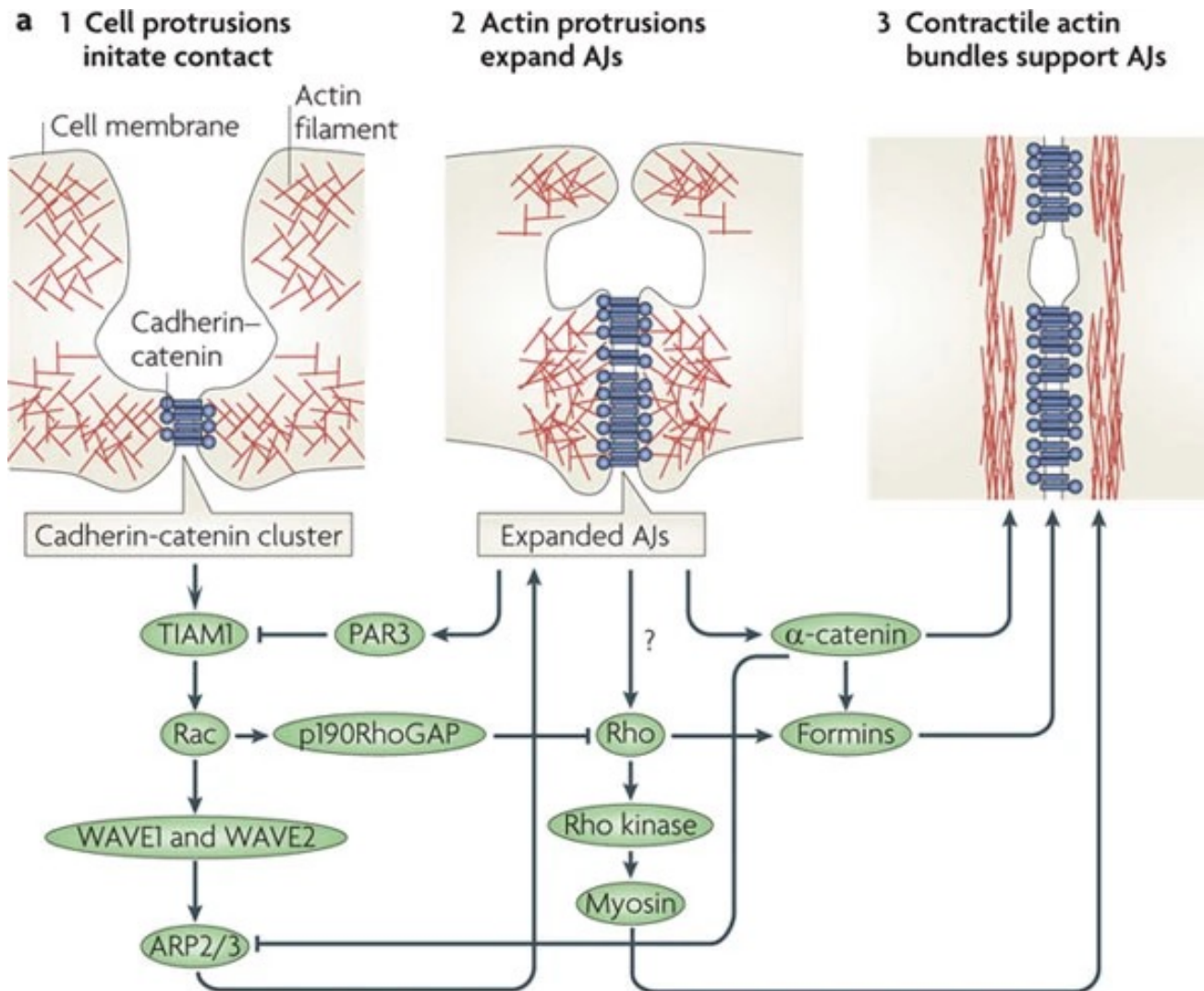
- The duration of the applied load plays a role in the final steady state. Indeed, on minute to hour time-scales, tissues behave as viscoelastic fluids, which can bear stresses at minute time scales but completely dissipate stress at hour-long time scales.
- When subjected to **short pulses of contractility**, tissues behave as **elastic solids** but, in response to **longer pulses of contractility**, they behave as **viscous fluids** and adopt the imposed shape as their rest shape.



Stress dissipation

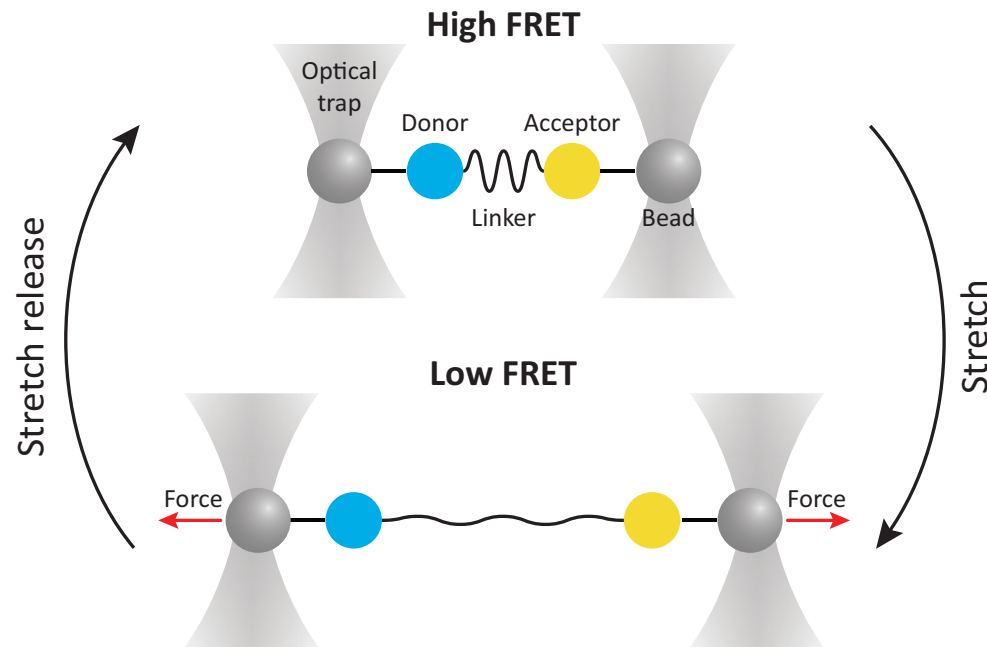


Maturation of adherens junctions



Molecular tension sensors

- Förster resonance energy transfer (FRET): radiation-free energy transfer from a fluorescent donor to an acceptor molecule
- Two fluorophores undergoing efficient energy transfer are connected by a spring-like linker peptide that stretches or unfolds in response to mechanical forces.
- The increase in fluorophore separation distance leads to a decrease in energy transfer rates so that mechanical tension can be determined by measuring FRET

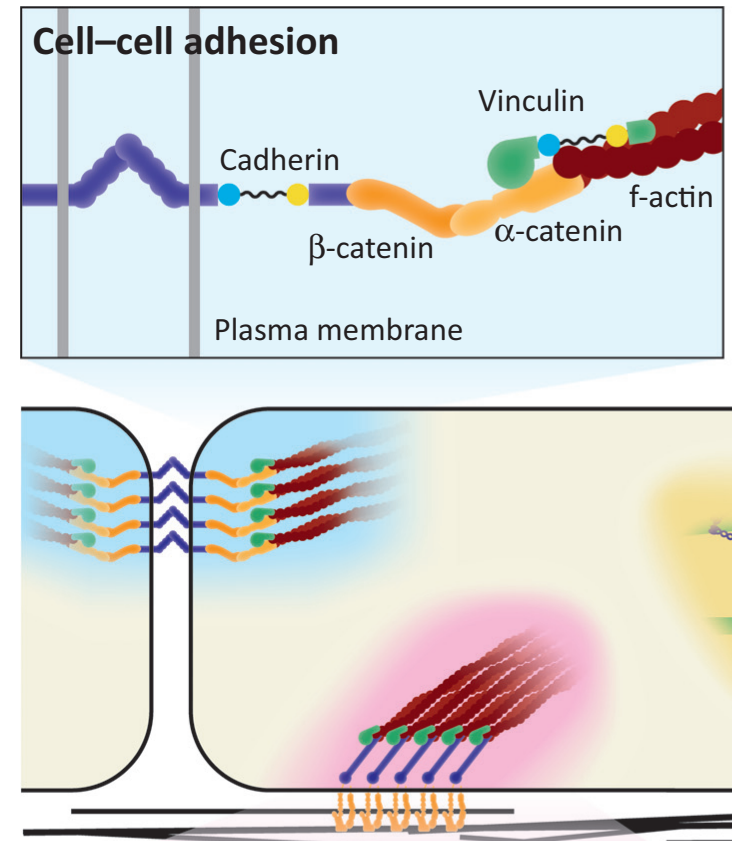


Molecular tension sensors

- Requirements for precise force measurements
 - the linker peptide has to respond to very small forces, such as those experienced by individual molecules in cells (pN force sensitivity)
 - the sensor element must quickly return to its original conformation once forces decrease (reversibility)
 - the peptide unfolding should be insensitive to the velocity at which mechanical force is applied (loading-rate insensitivity)
 - the linker must display similar stretch/relaxation transitions (absence of hysteresis)

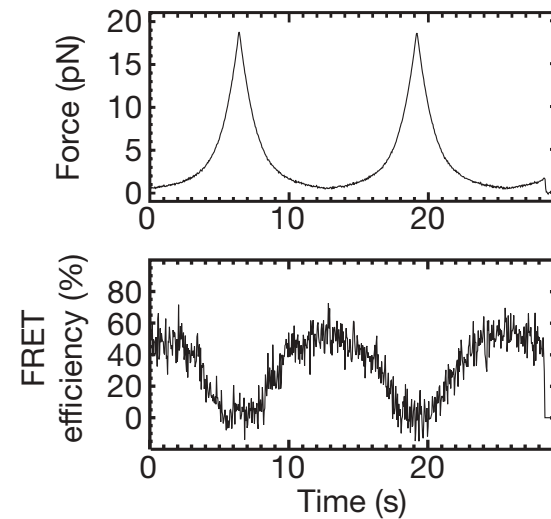
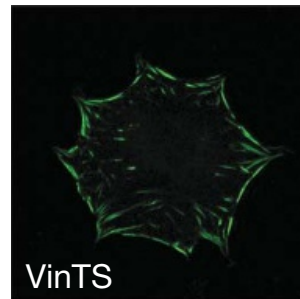
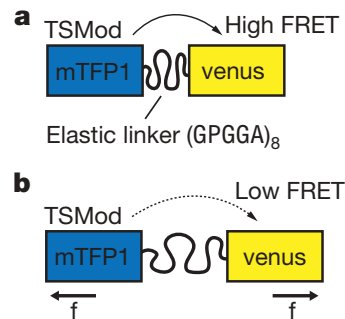
Genetically encoded tension sensors

- E-cadherin, VE-cadherin, and vinculin
- F40 was derived from spider silk protein
- Sensitive to forces of about 1-6 pN



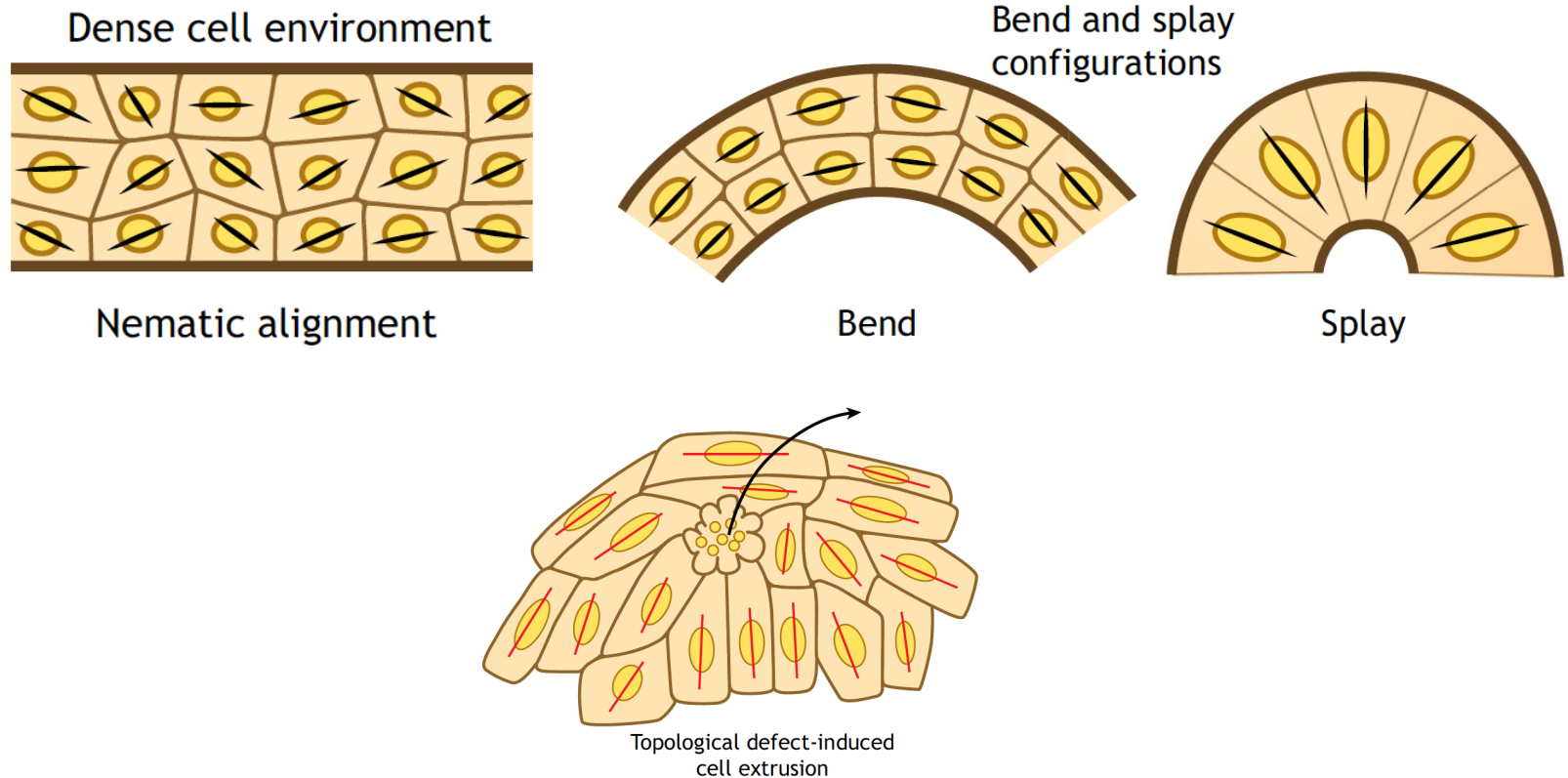
Genetically encoded tension sensors

- Vinculin connects integrins to actin filaments
- Tension across vinculin in stable focal adhesions 2.5 pN
- High force leads to adhesion assembly and enlargement

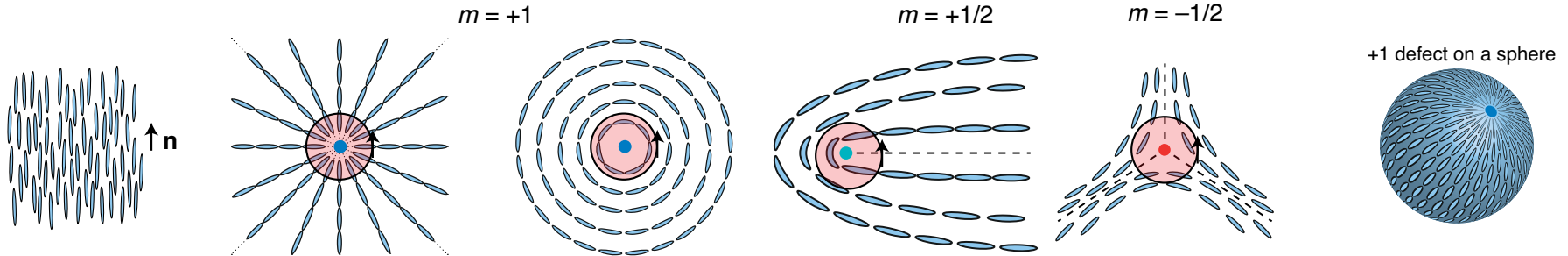
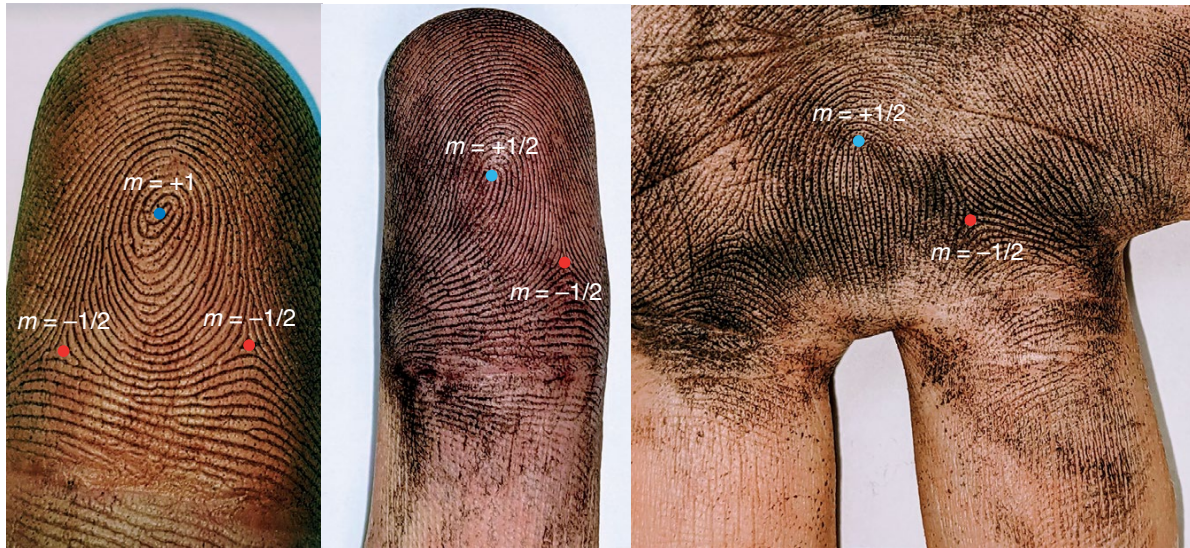


Cell clusters as active nematics

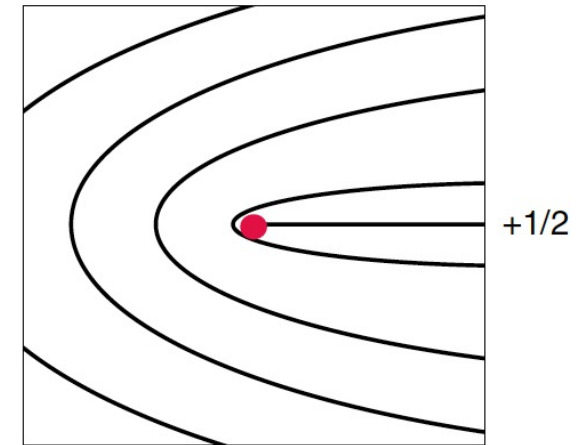
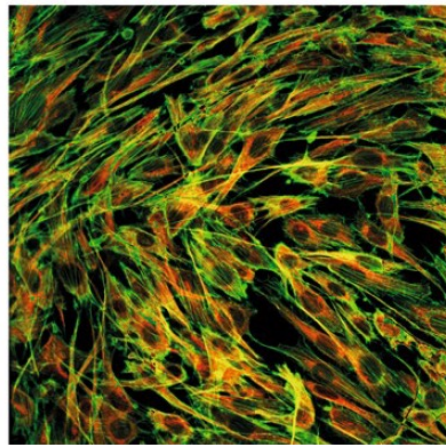
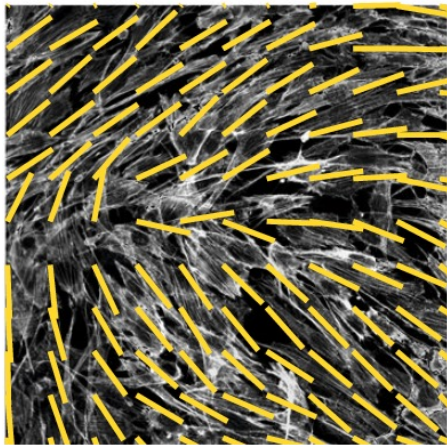
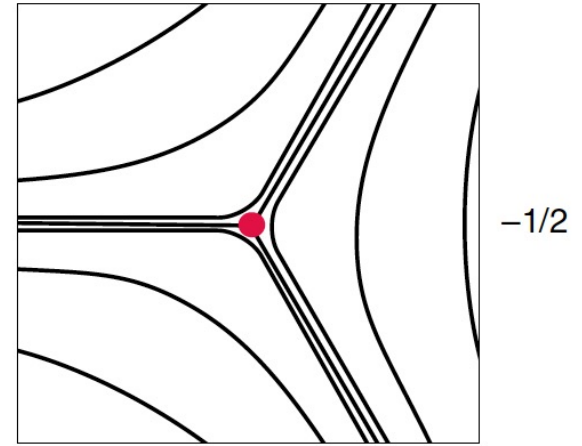
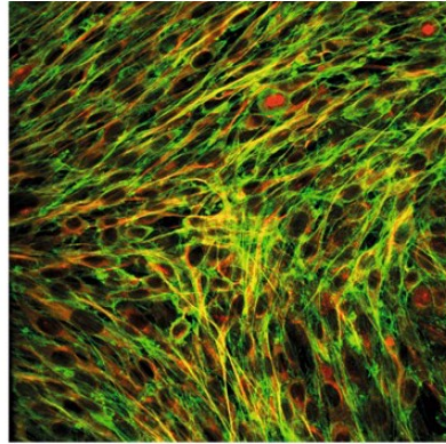
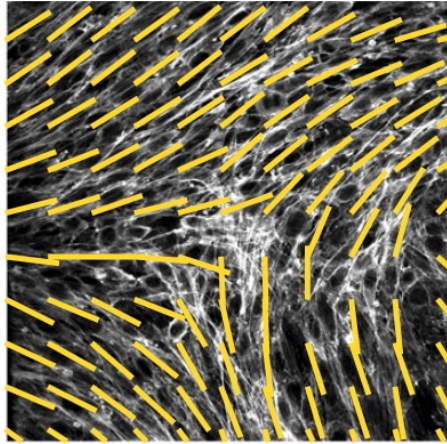
- Liquid crystals are made up of anisotropic molecules
- Flow like particles in liquid but can orient in a crystal-like way
- Long range orientation order



Topological defects



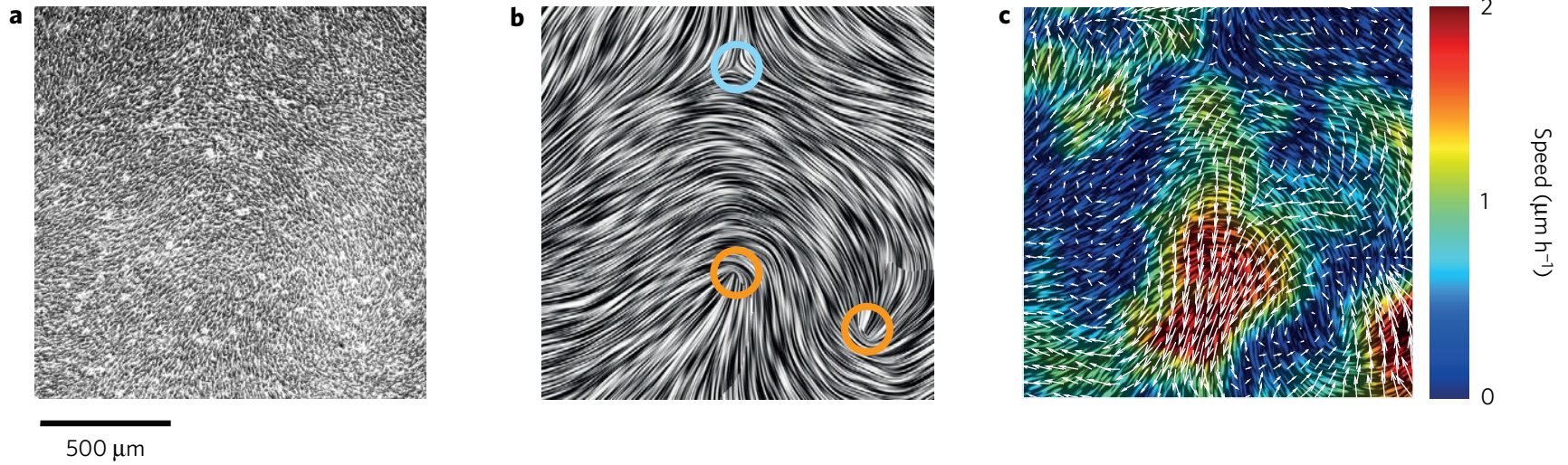
Cell clusters as active nematics (fibroblasts)



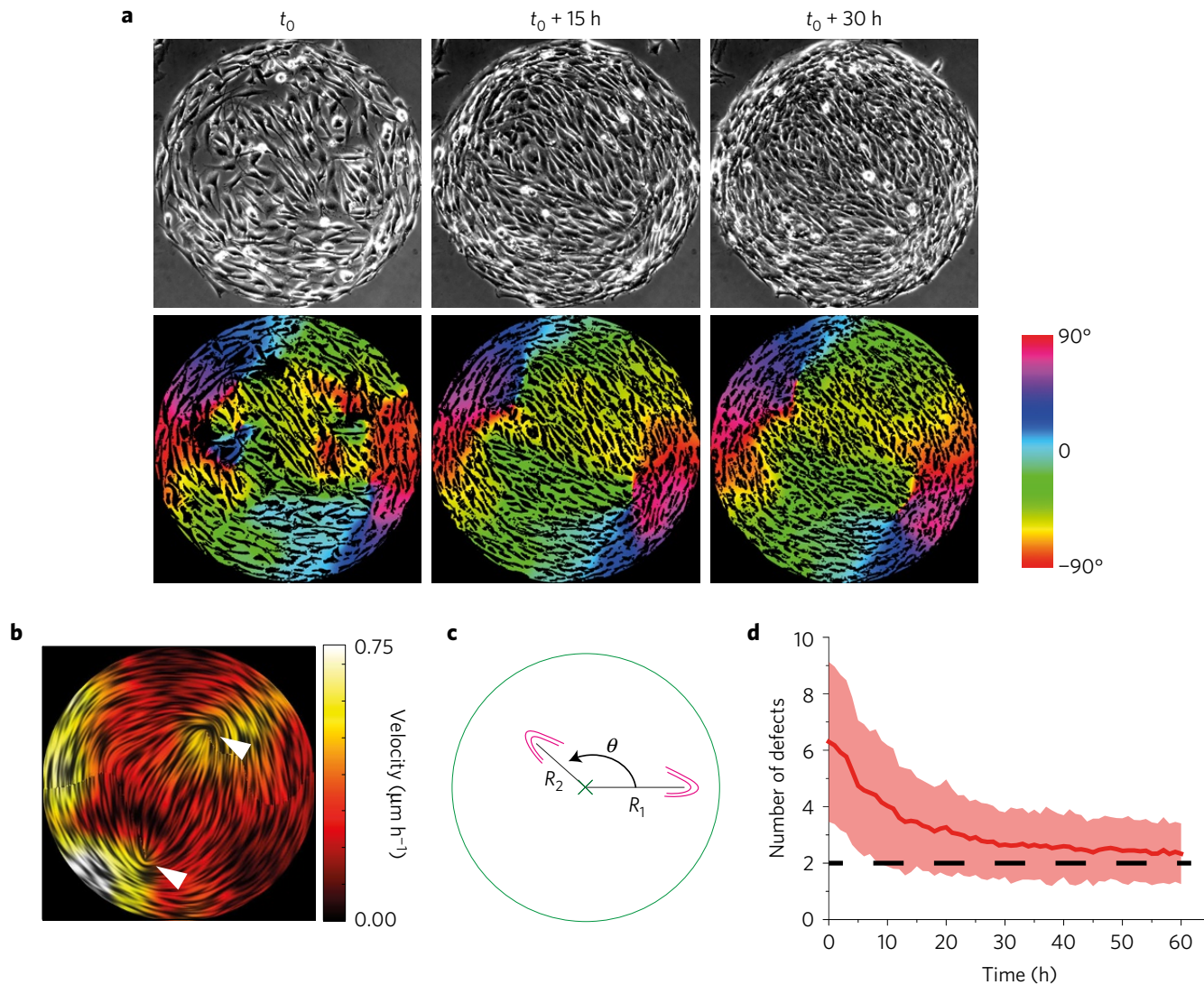
100 μm

Actin/tubulin

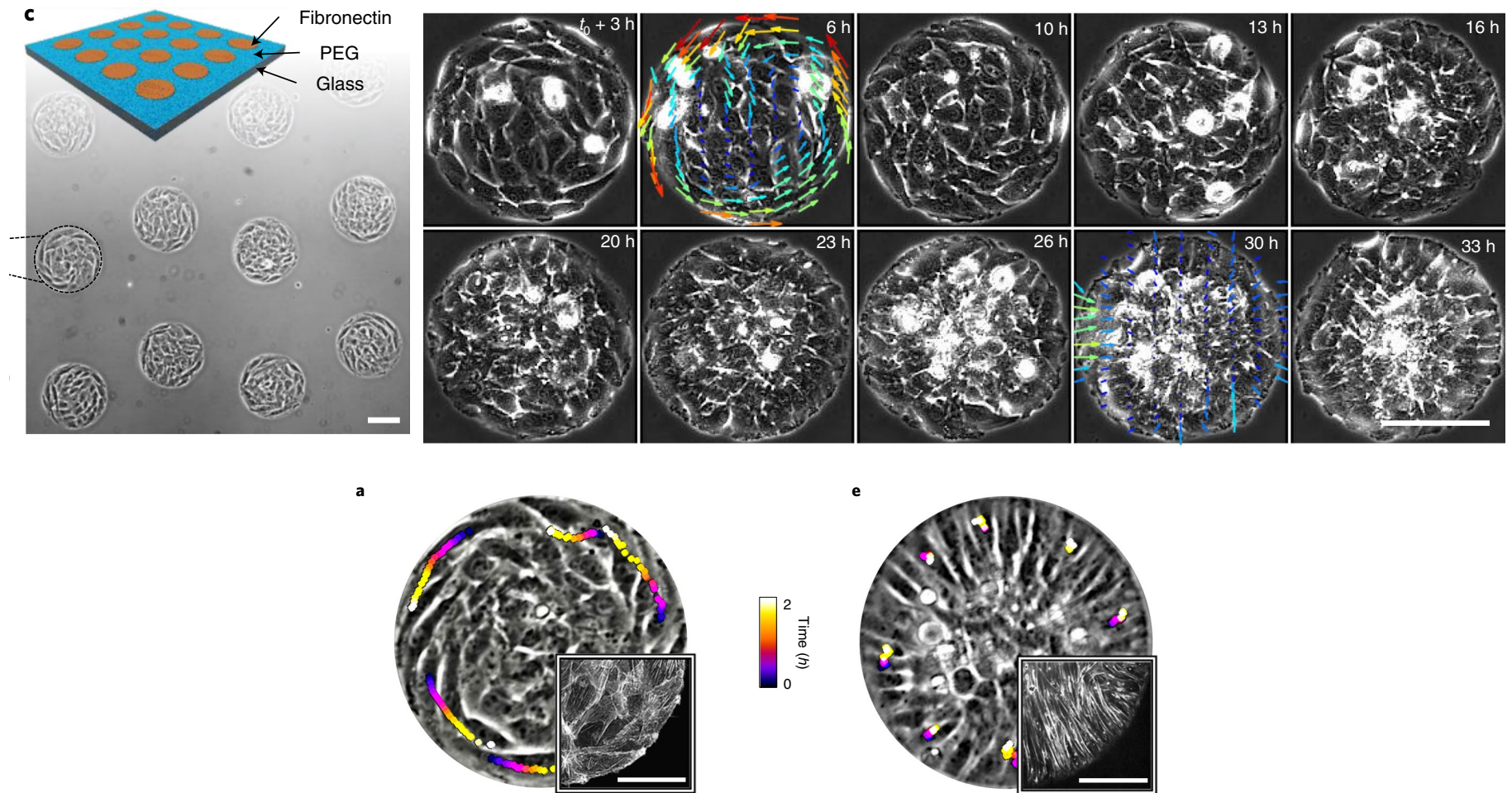
Cell clusters as active nematics



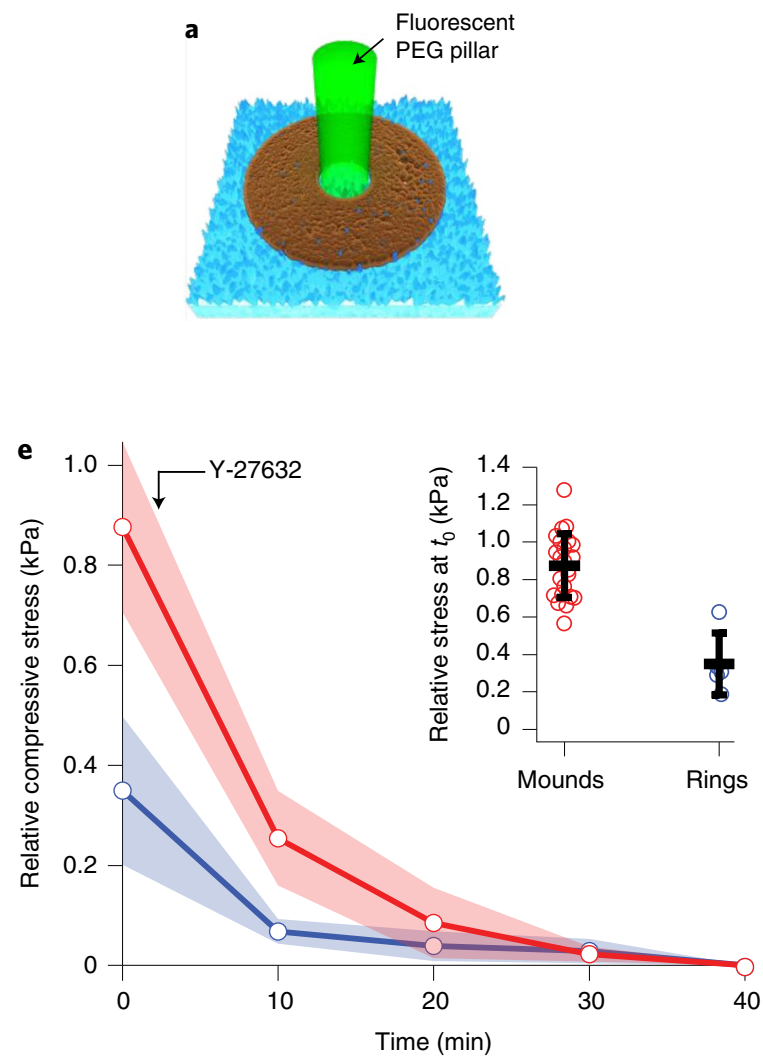
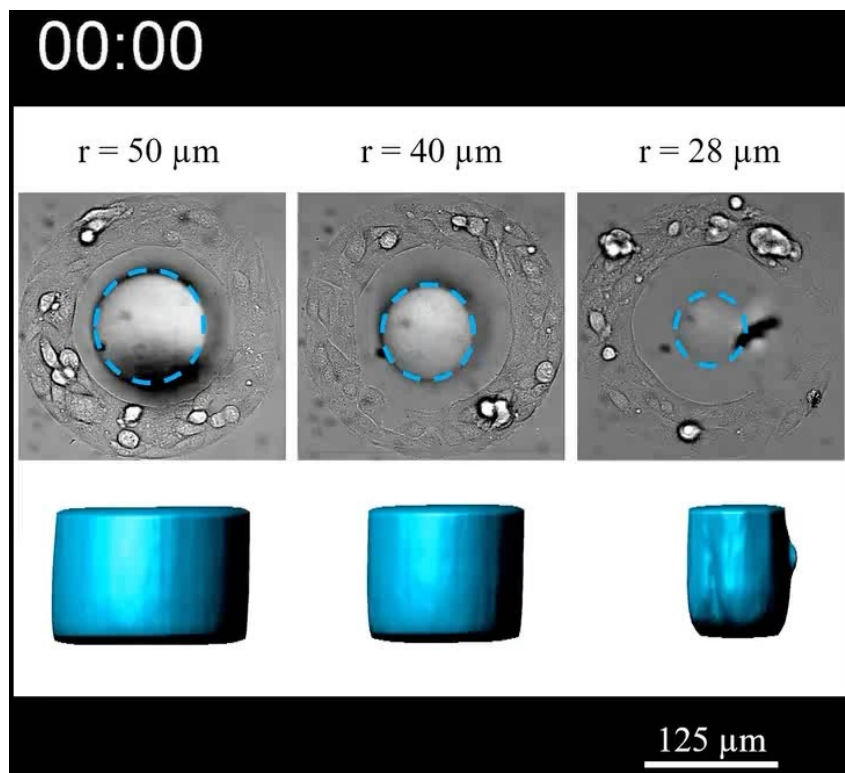
Cell clusters as active nematics



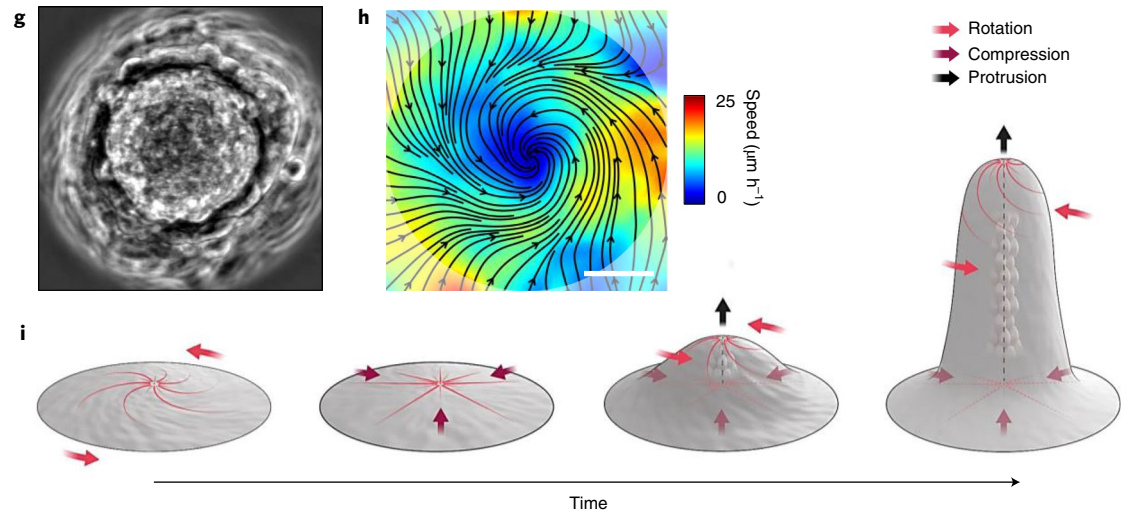
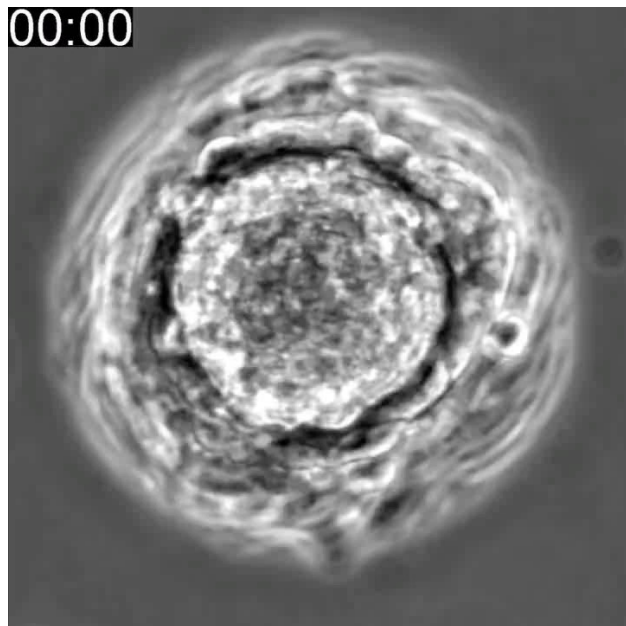
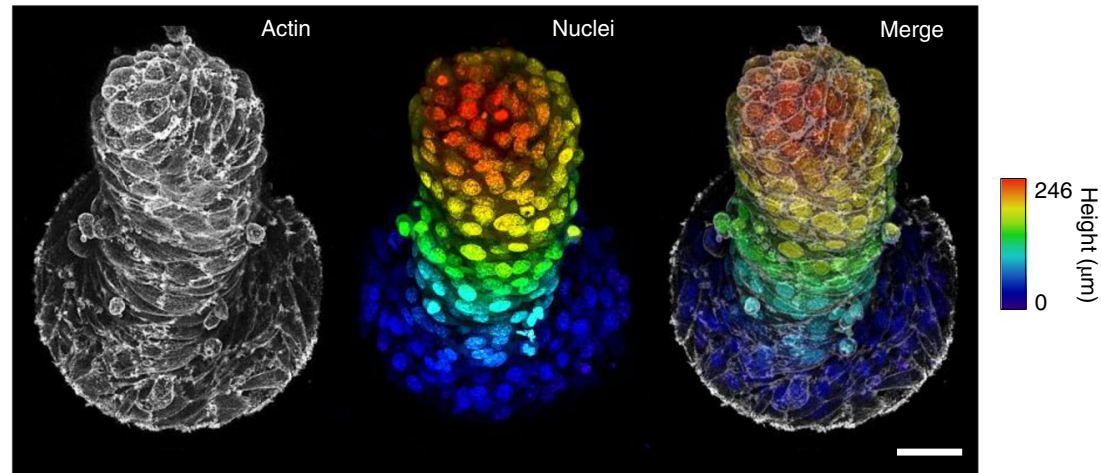
Topological defects organize stresses



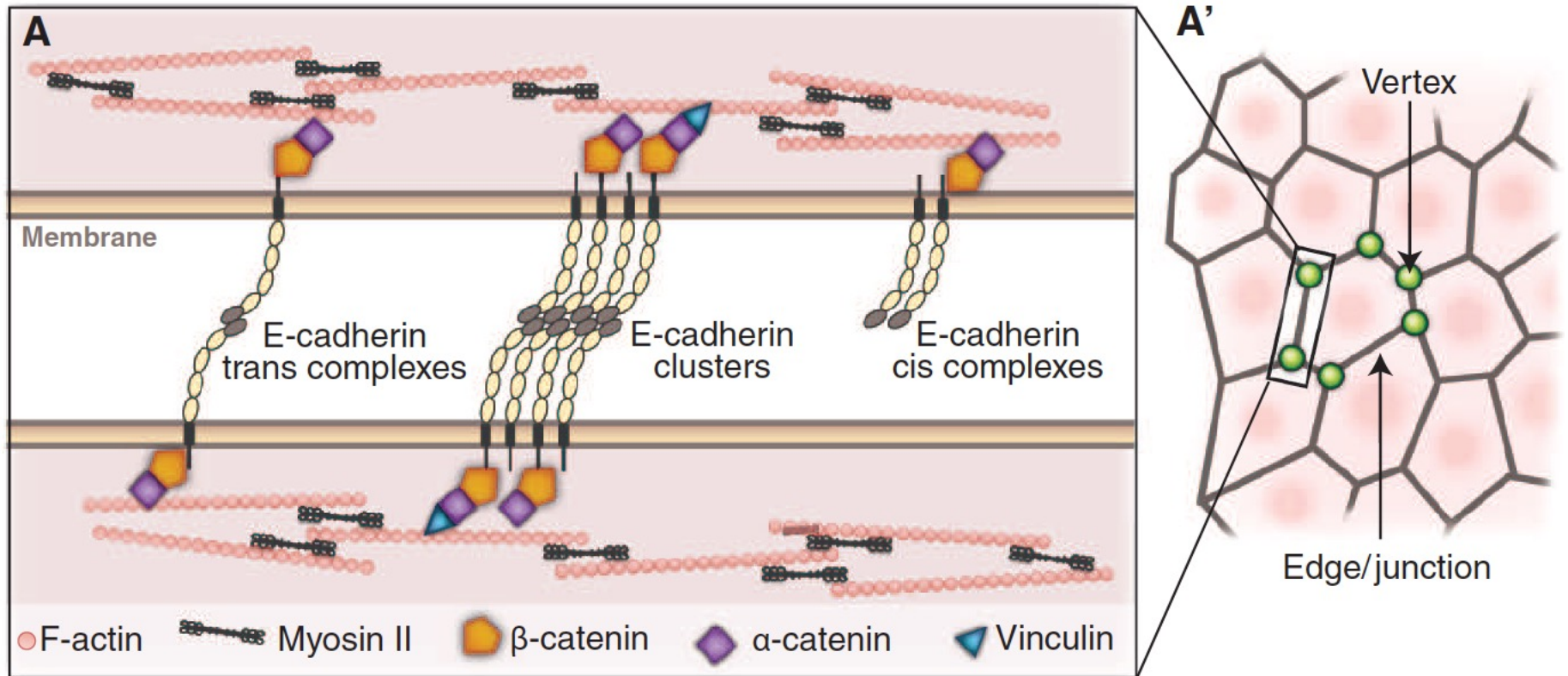
Topological defects organize stresses



Topological defects organize stresses

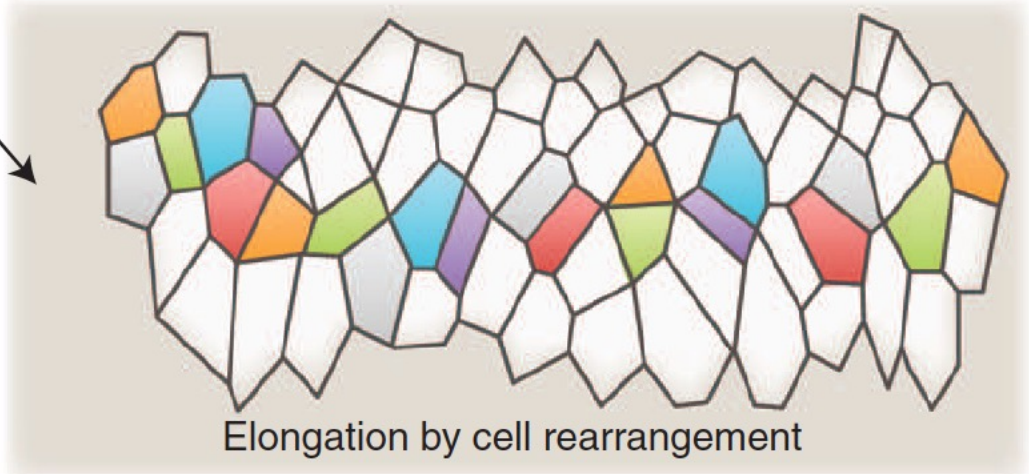
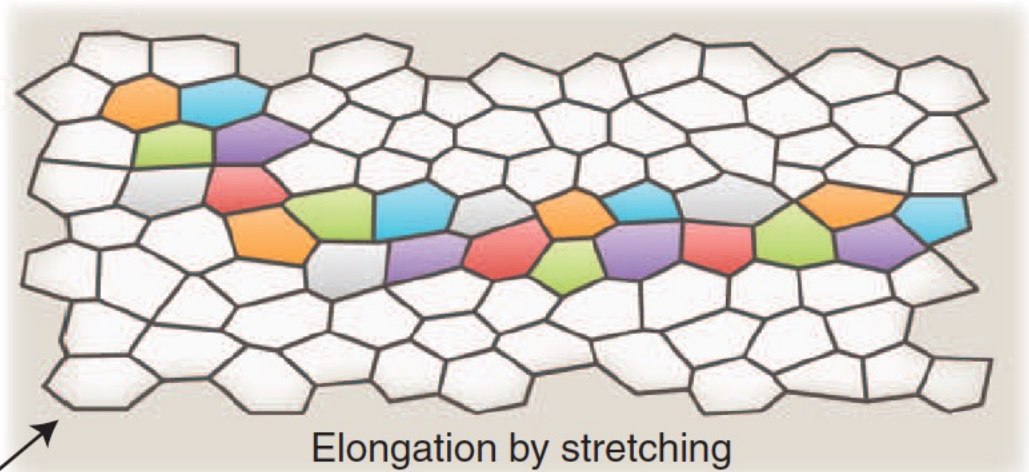
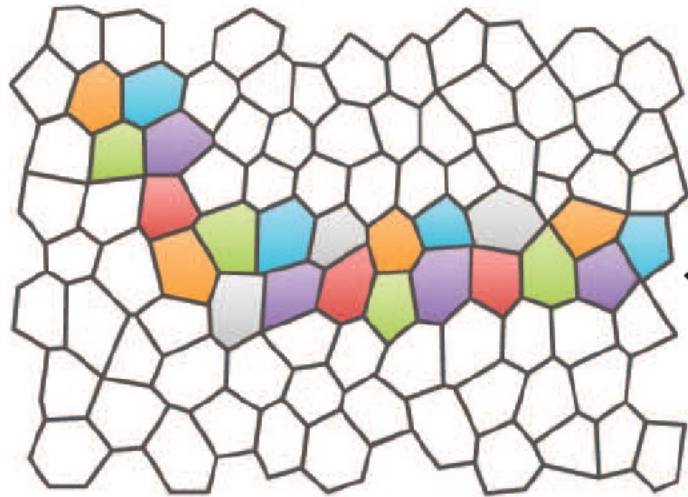


Epithelial tissue mechanics: basics

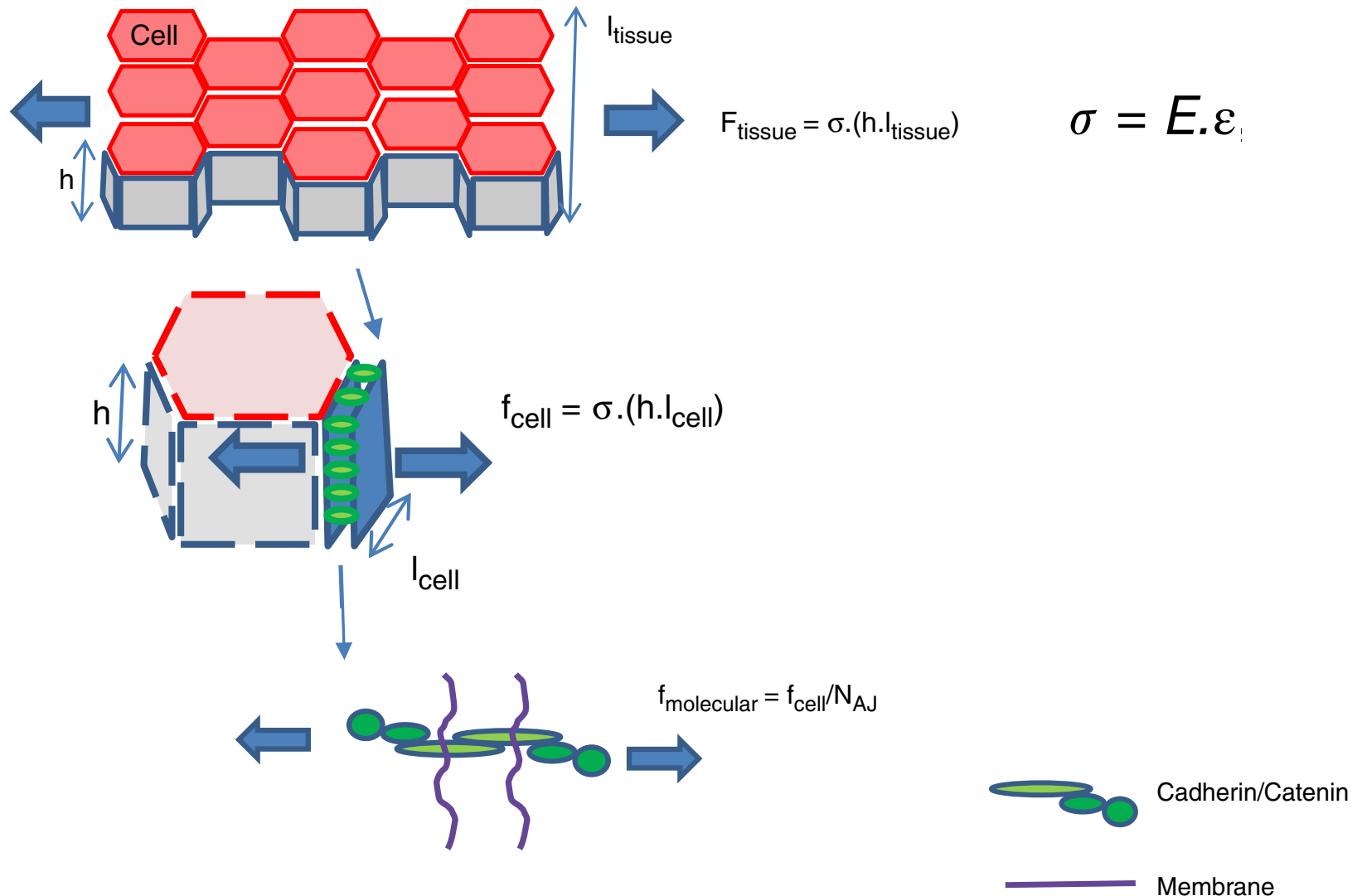


Epithelial tissue mechanics: basics

B

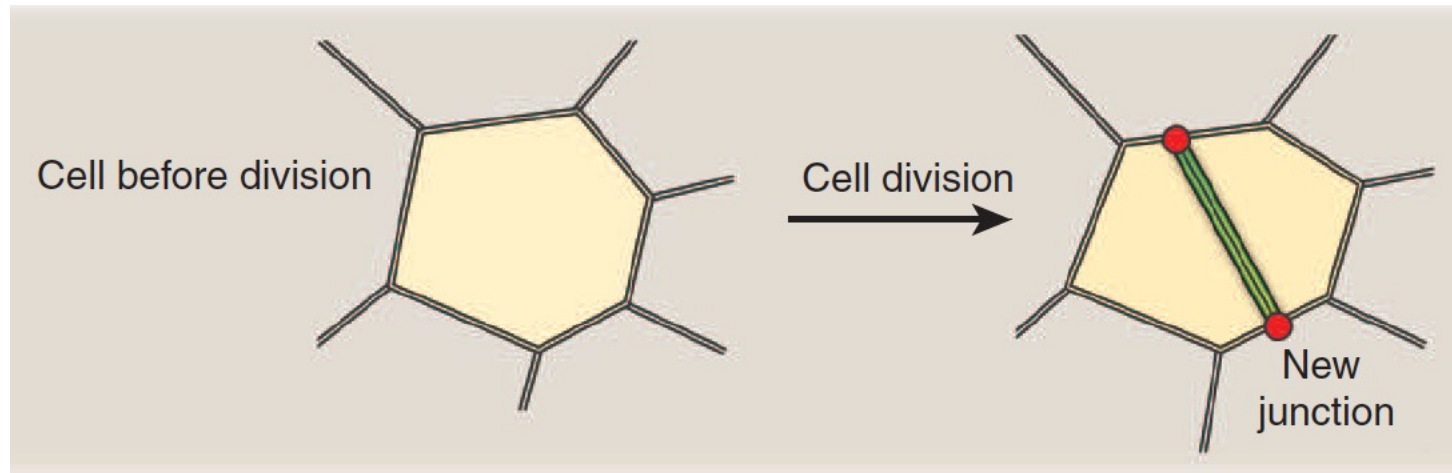


Hierarchical loading: from tissue to protein



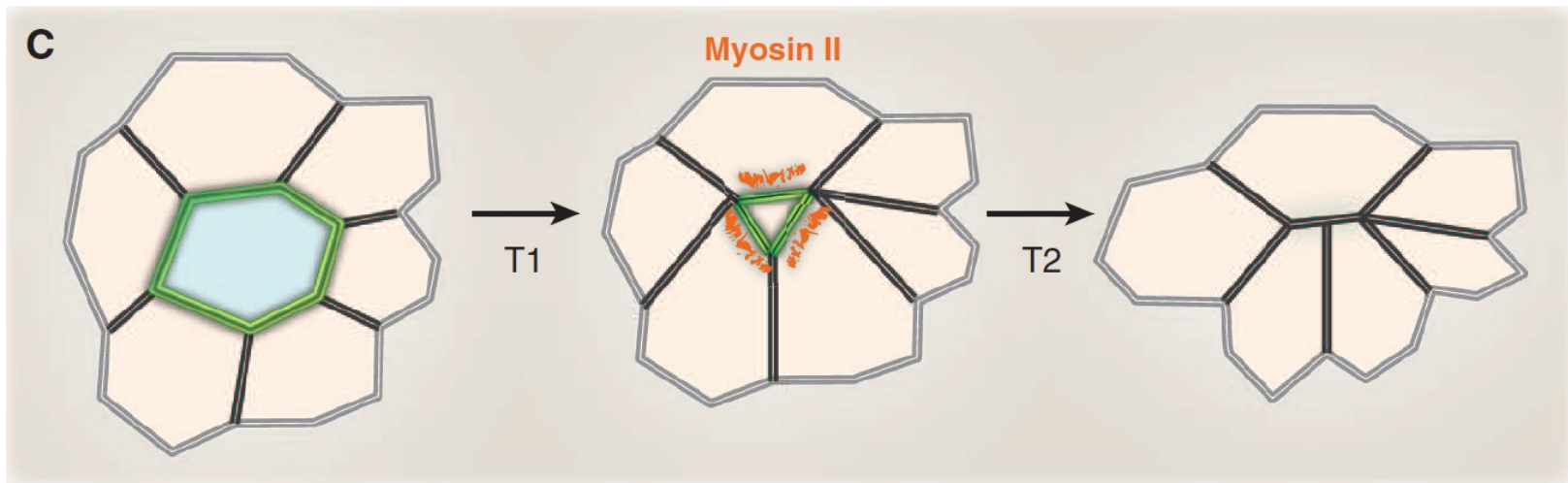
Epithelial tissue mechanics: basics

- Both solid-like and fluid-like behavior
- Cells continuously die or divide
 - Removal of junctions and cells by extrusion
 - Formation of new contacts during division
- Cells can change shape or intercalate
 - Neighbor exchange

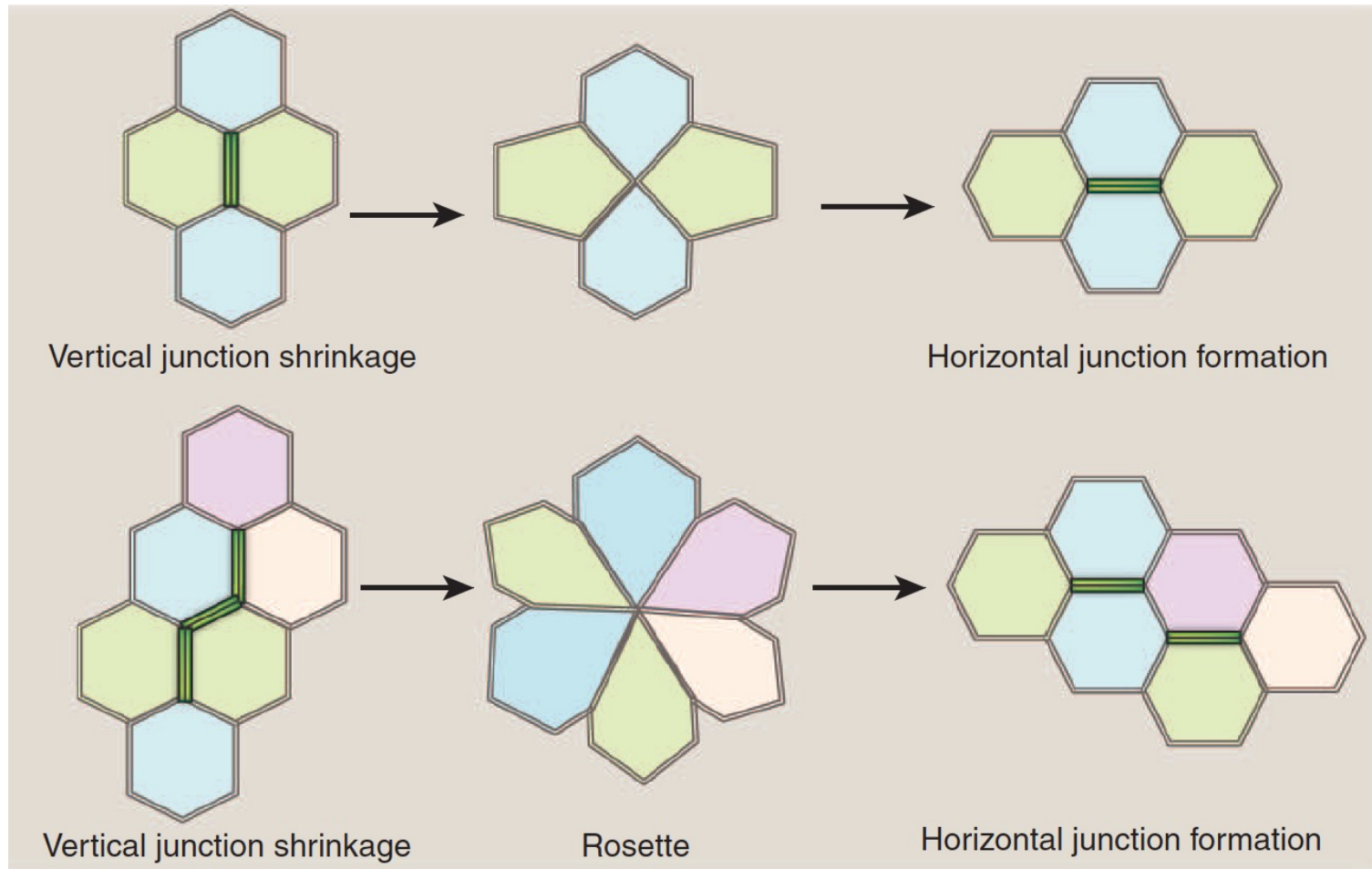


Epithelial tissue mechanics: basics

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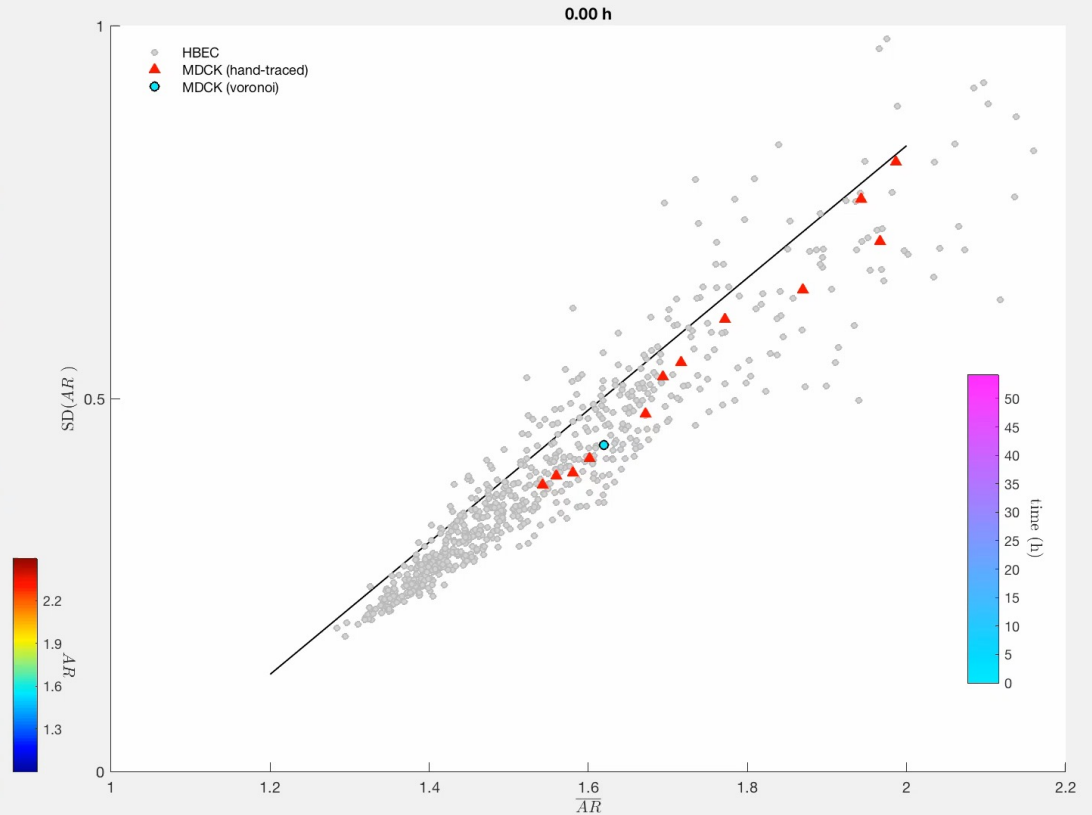
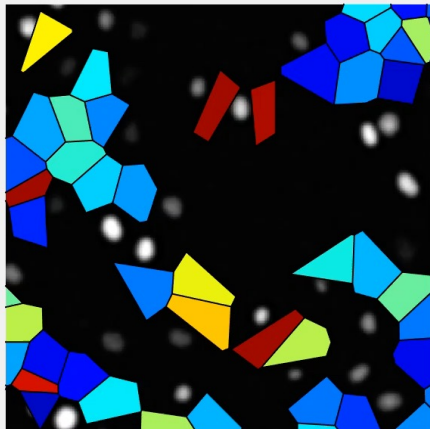
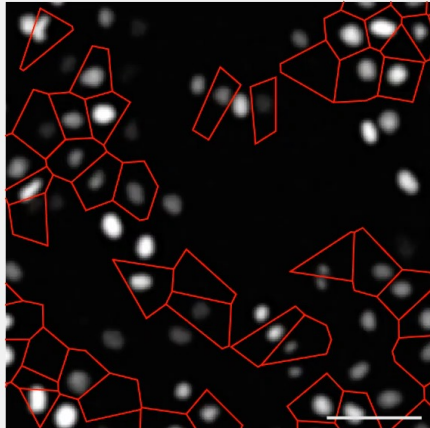
Epithelial tissue mechanics: basics



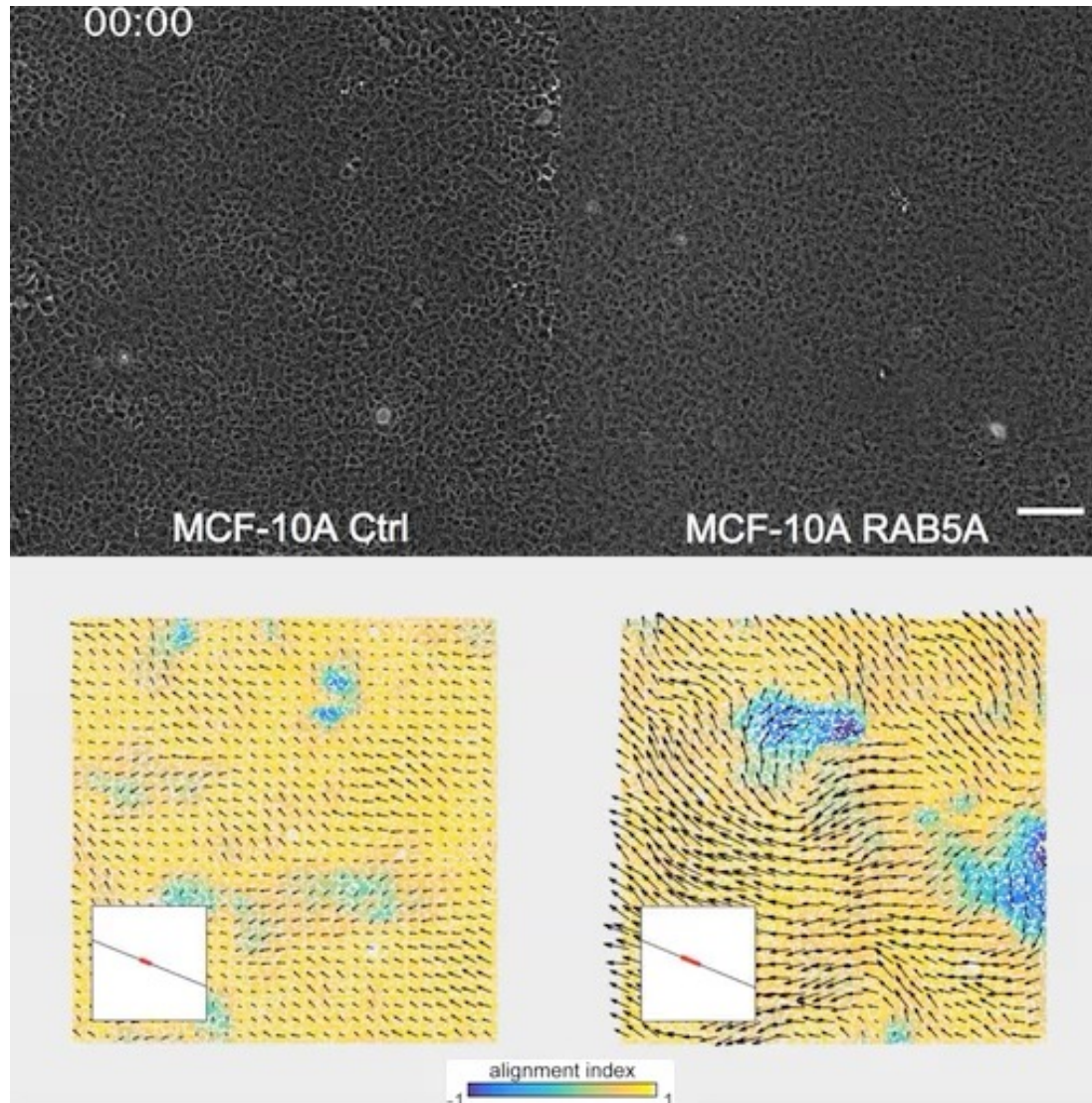
Jamming transitions

- During collective migration within confluent monolayers, cell sheets flow like a fluid yet remain fixed and solid-like at short timescales, with the motion of each cell constrained by the cell crowding due to its neighbours.
- As cell density rises, neighbouring cells restrict the motion of each cell, forcing them to move in groups
 - jamming or rigidity transition at large density
- Material parameters that encode cell properties such as cell-cell adhesion and cortical tension, rather than density alone, have been proposed to govern the rigidity transition in cell monolayers
 - Cell shape as an index

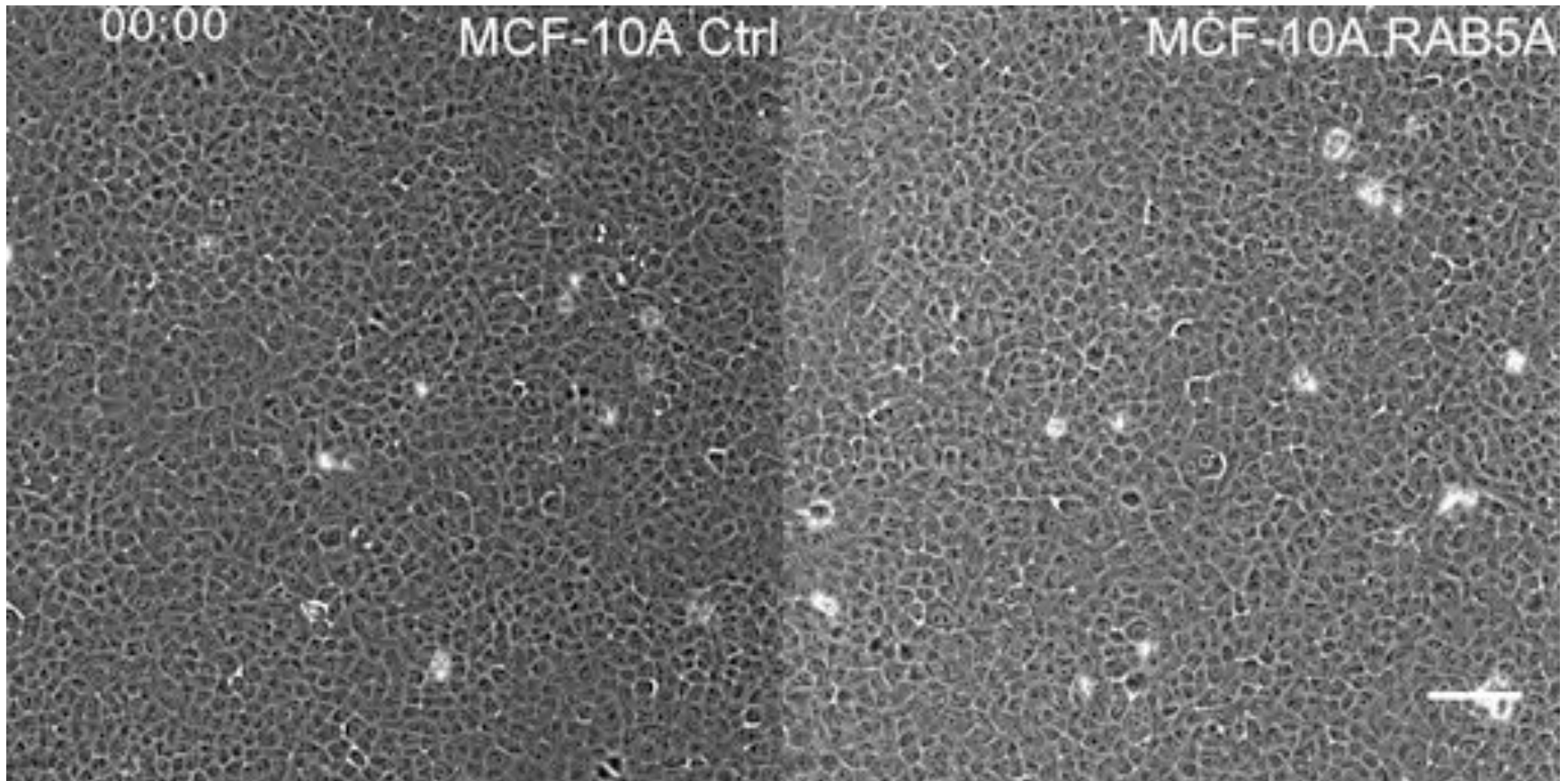
Jamming transitions



Jamming transitions

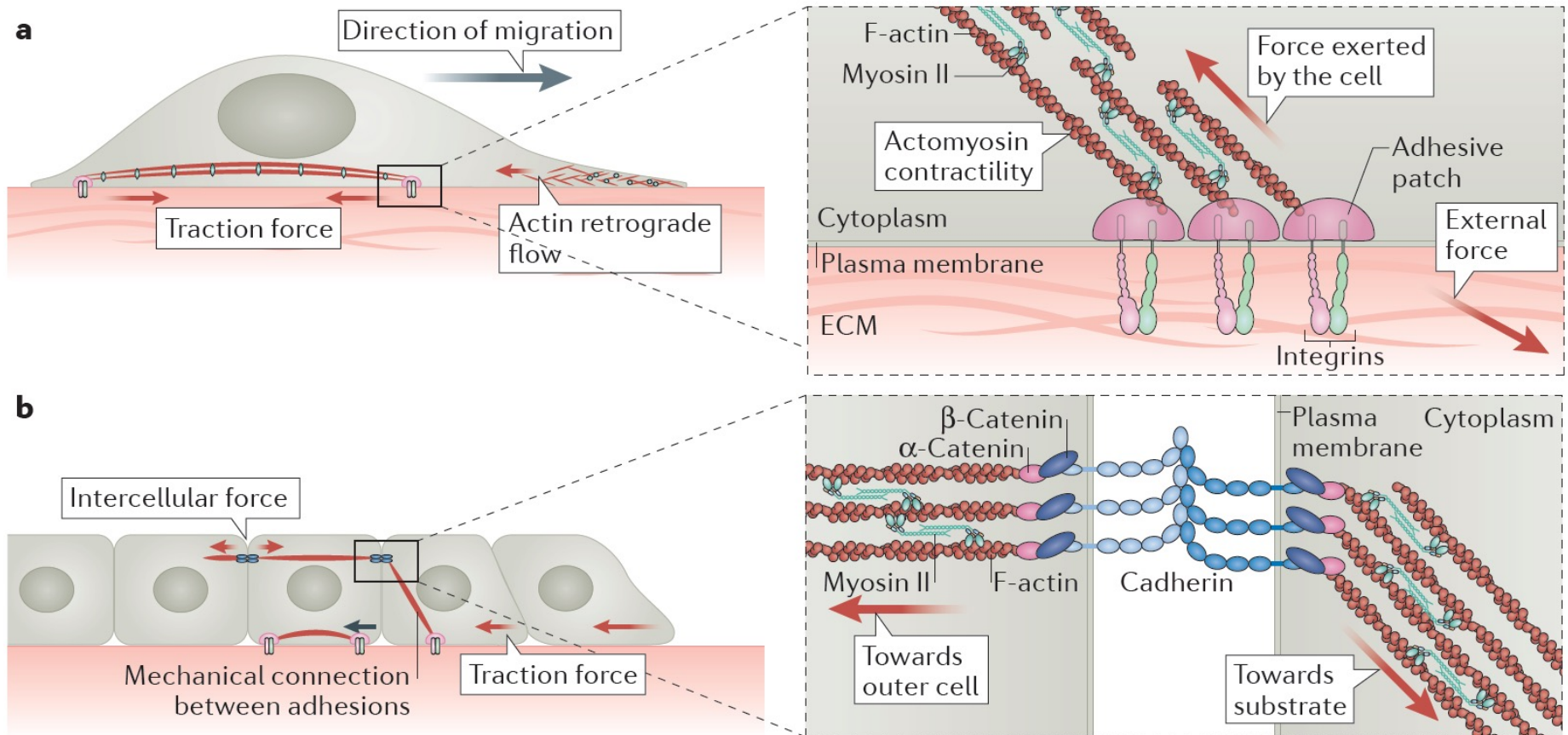


Jamming transitions



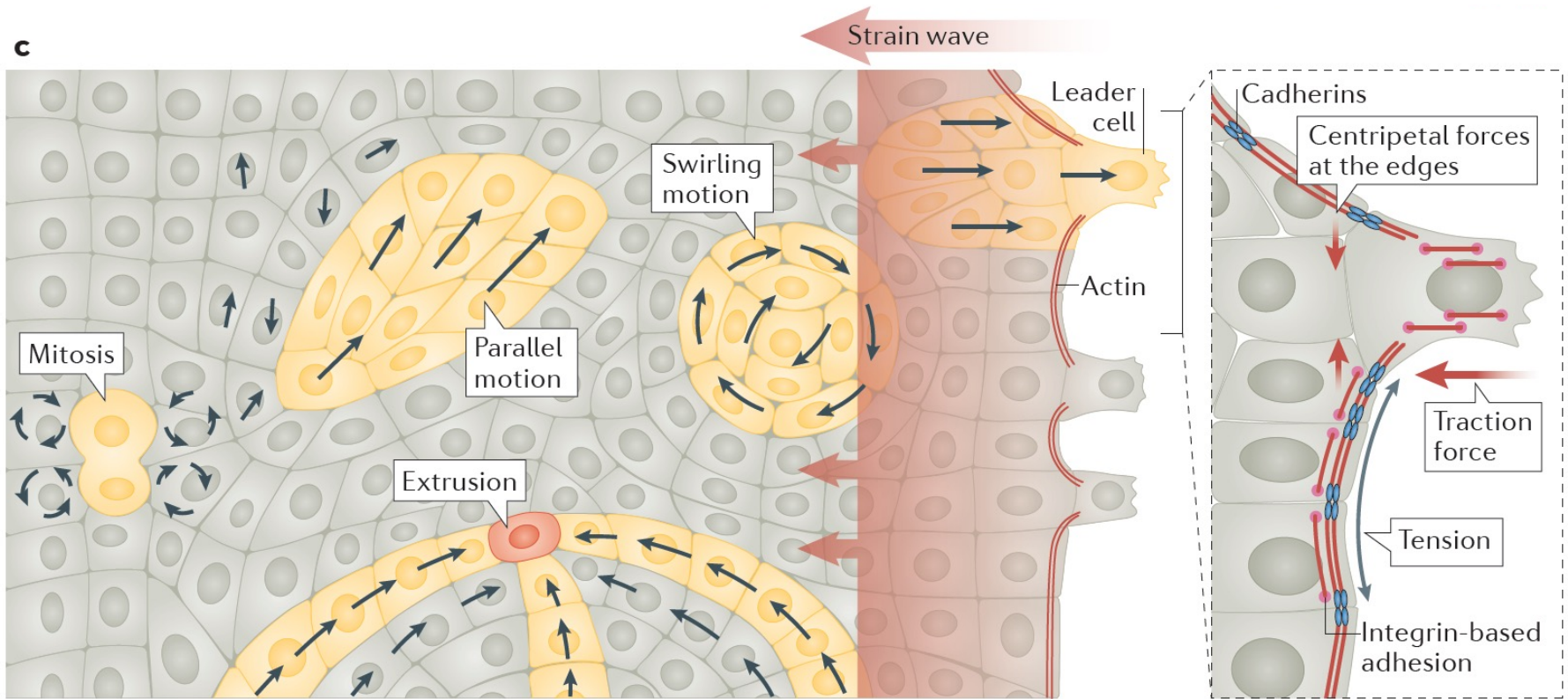
Cell movements from single cell to collective dynamics

- Cadherin and cell-cell coupling at adherens junctions
- Forces higher than 5pN drive a conformational switch on catenin



Cell movements from single cell to collective dynamics

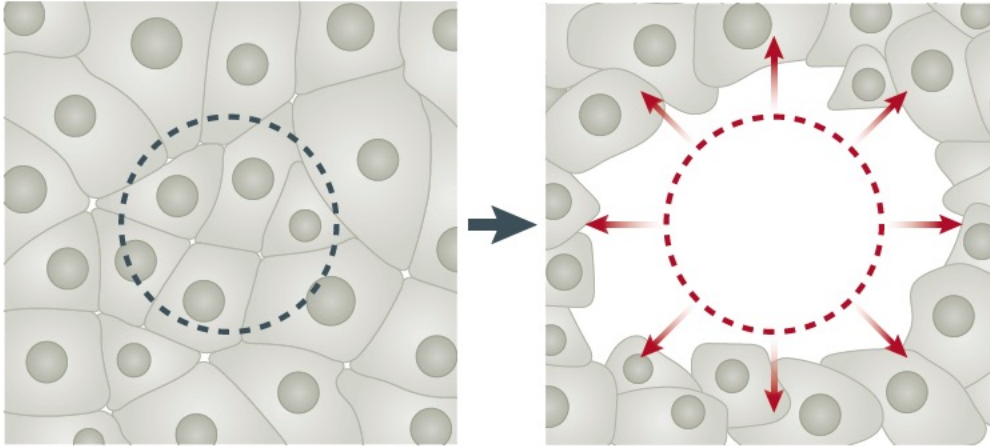
- Laminar flows, swirling motions, vortices
- Strain and velocity waves driven by mechanochemical signals



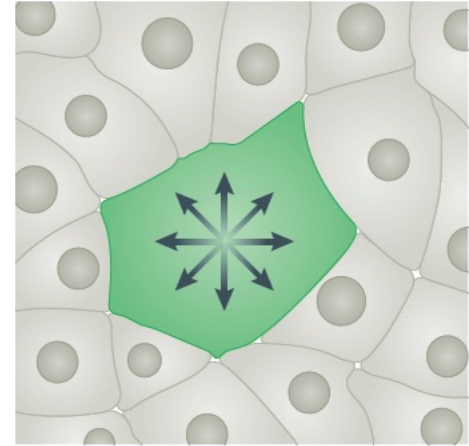
Experimental methods

Aa

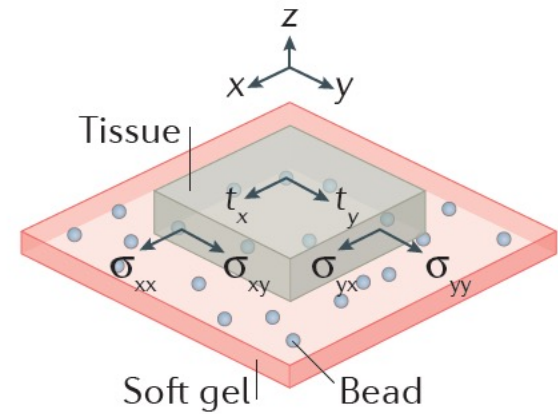
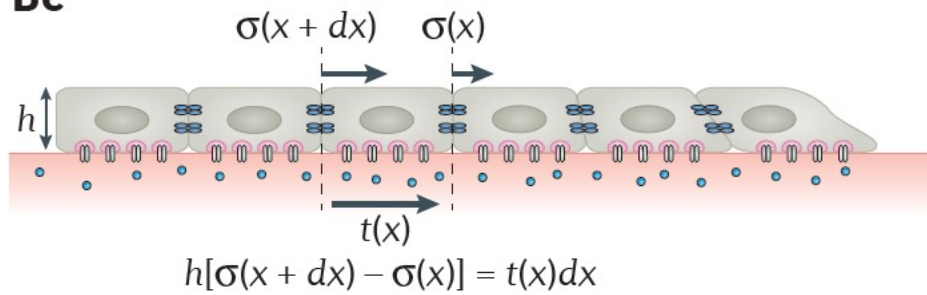
Laser ablation



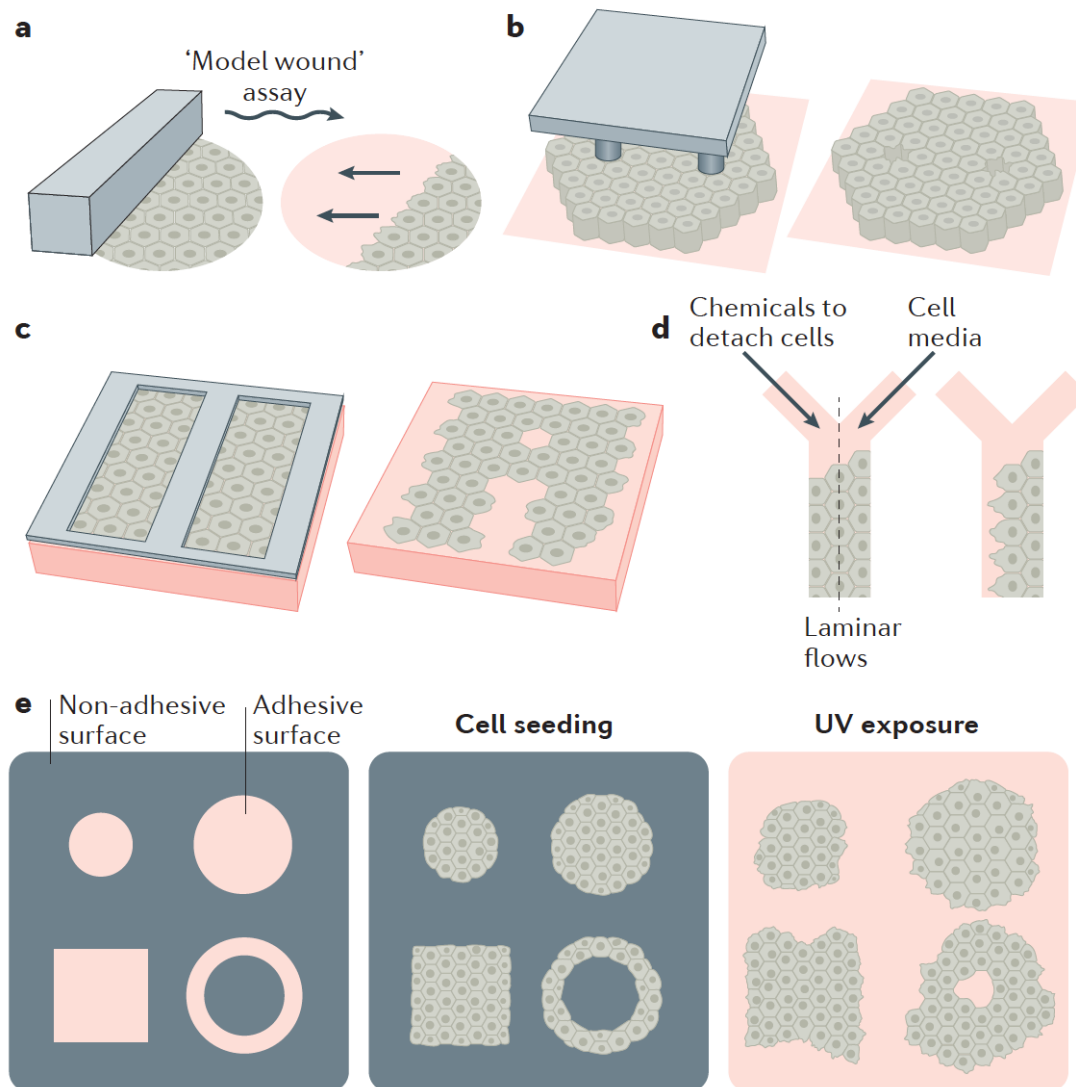
Ab Deformable droplet



Bc

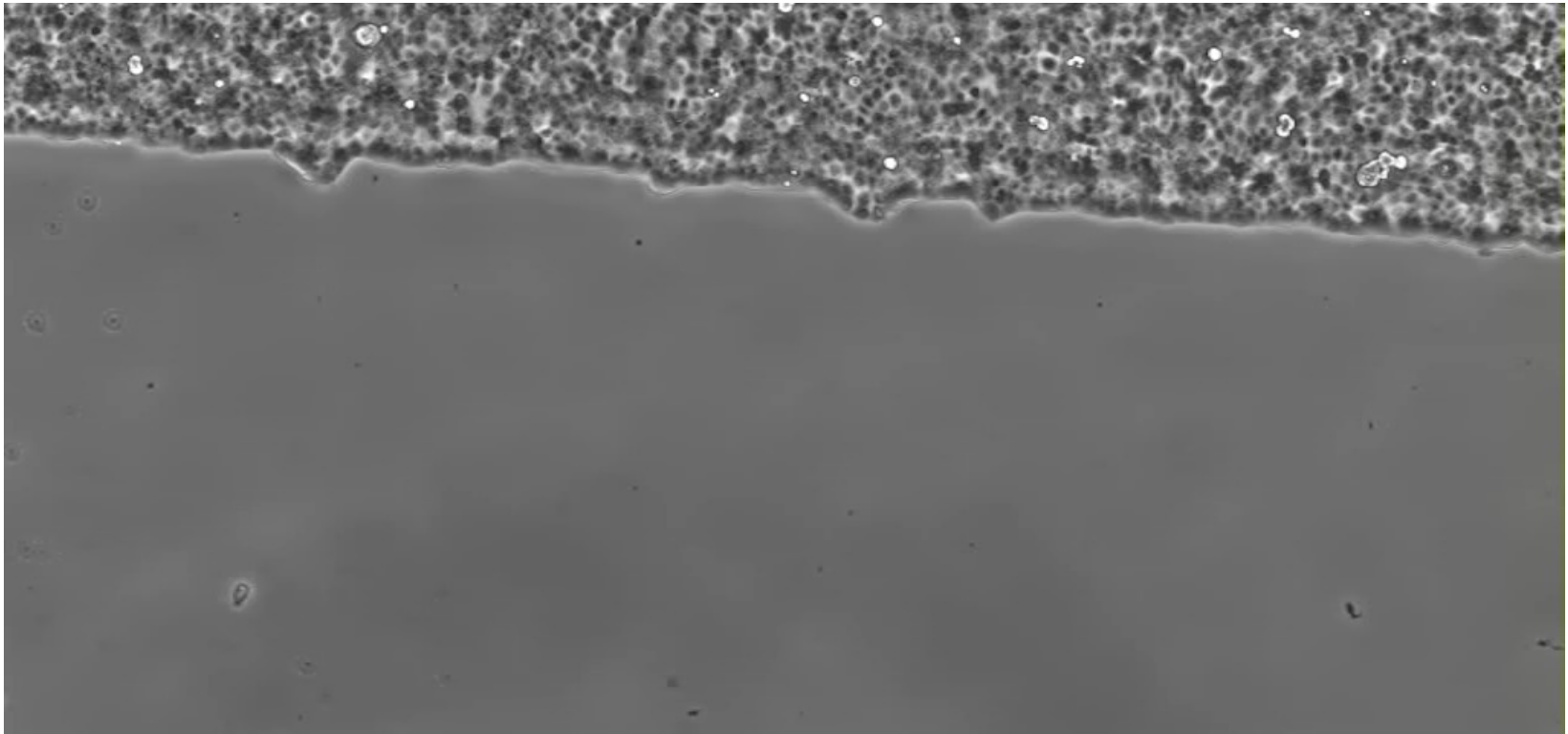


Experimental methods

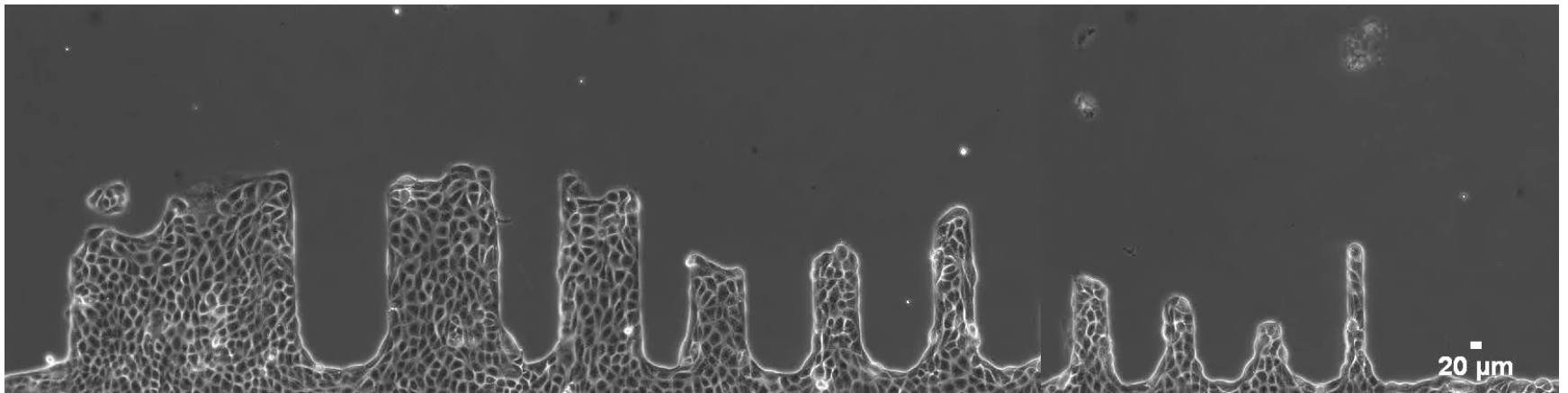
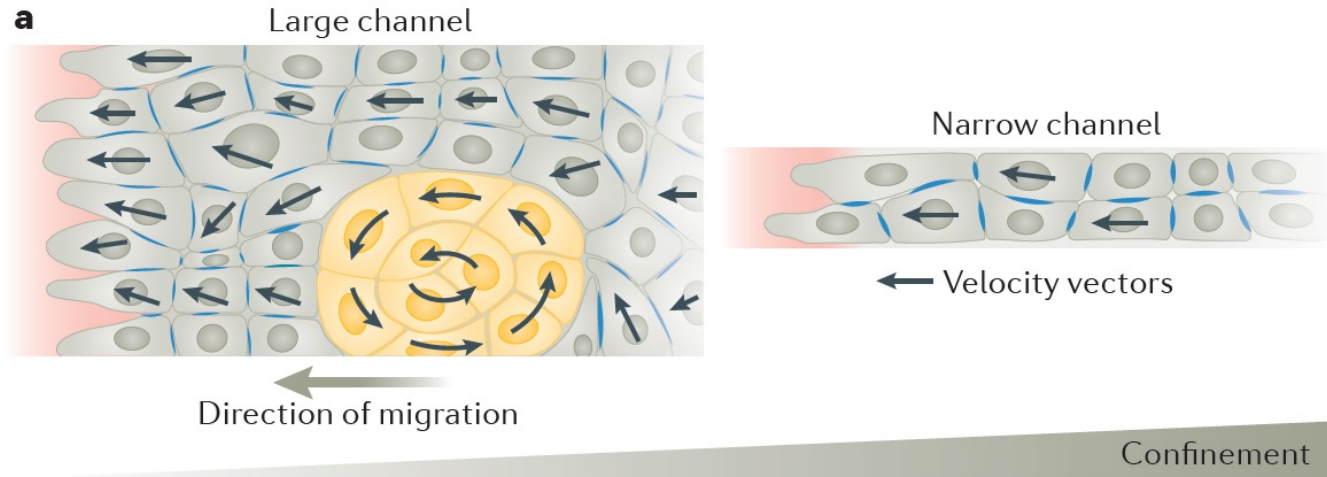


Cell movements from single cell to collective dynamics

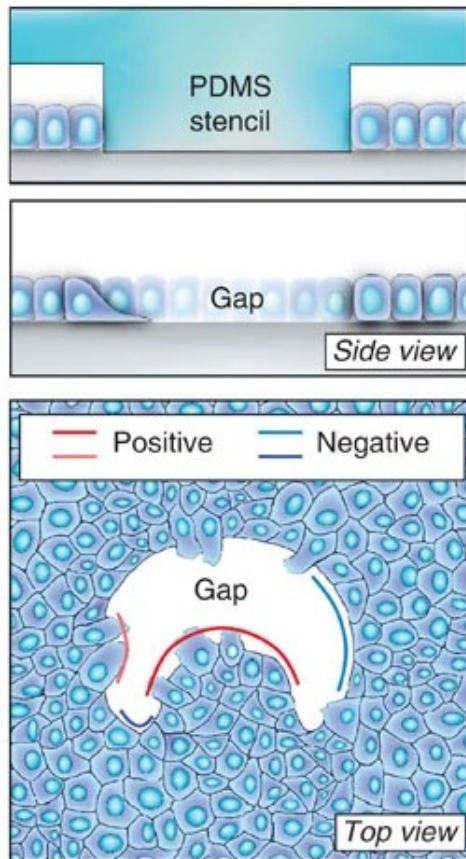
- Laminar flows, swirling motions, vortices
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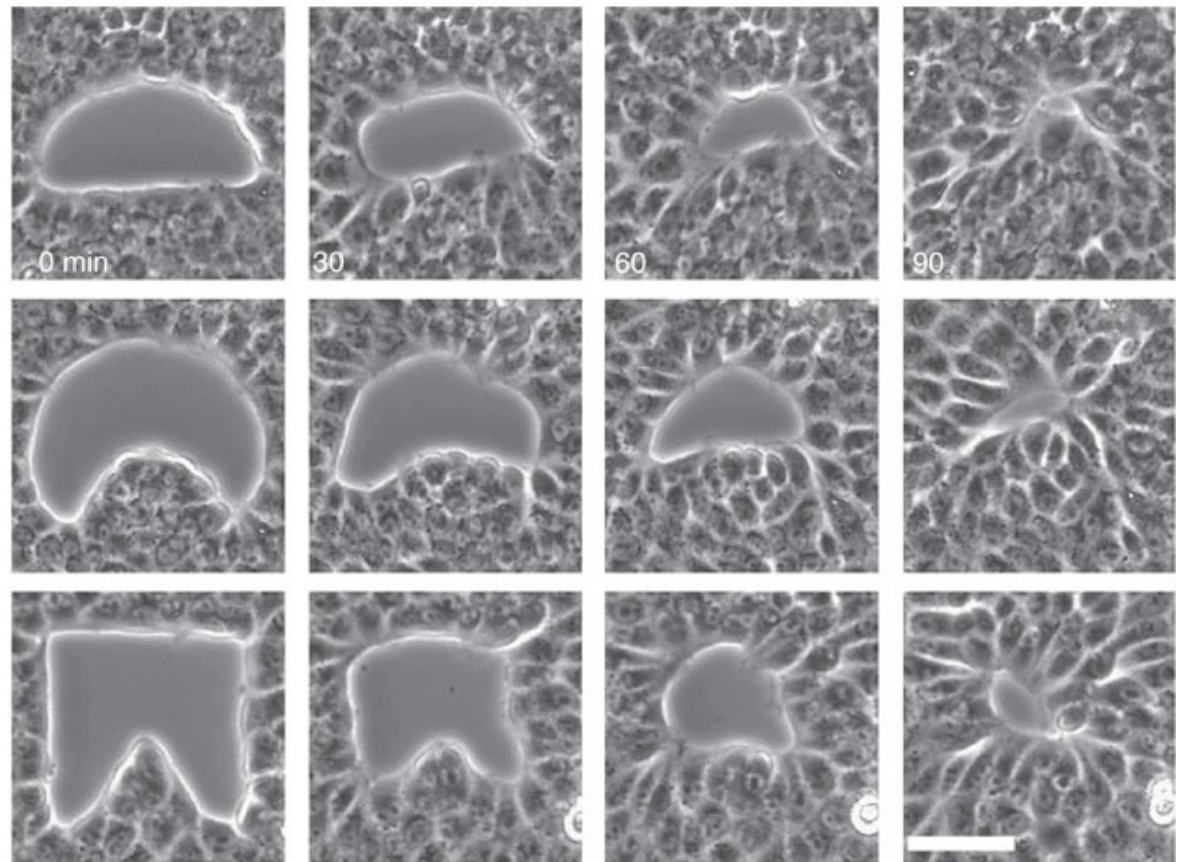
Tissue motion and geometric confinement



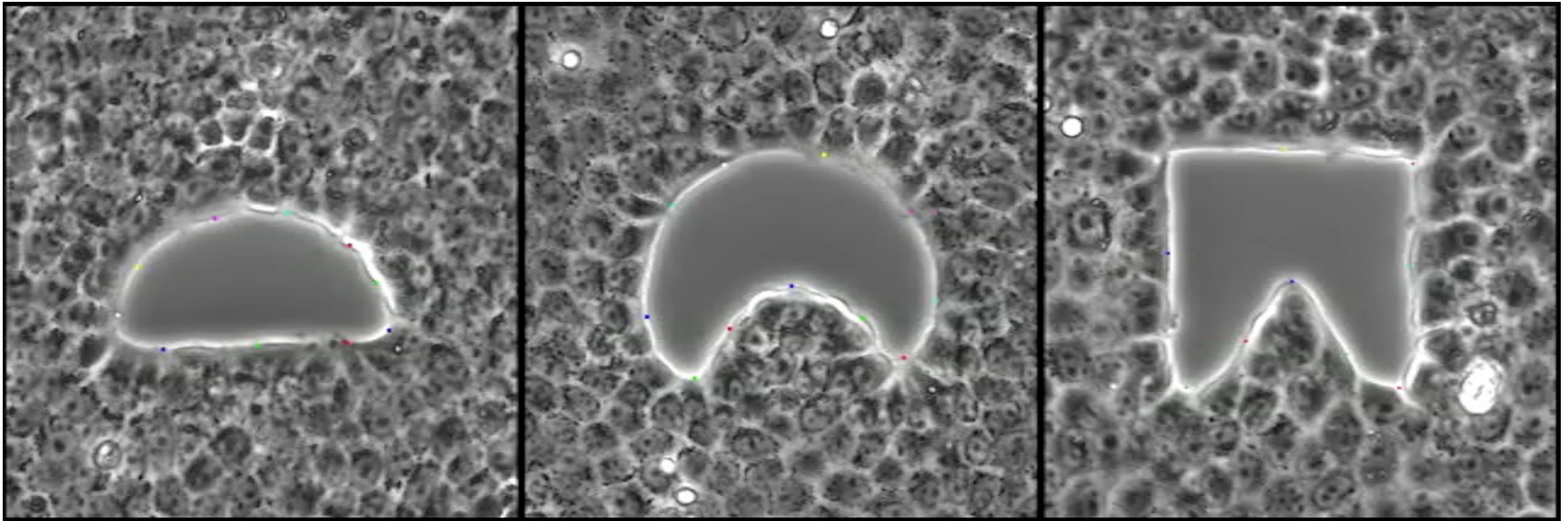
Curvature



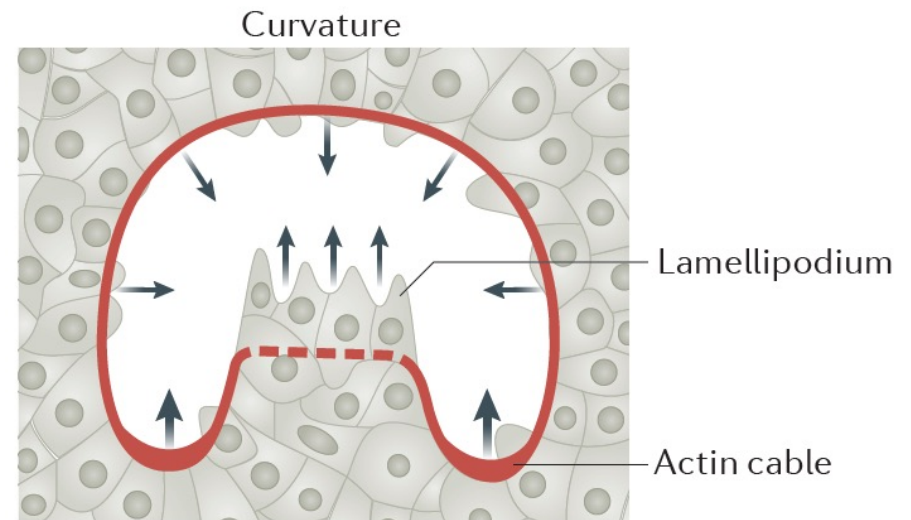
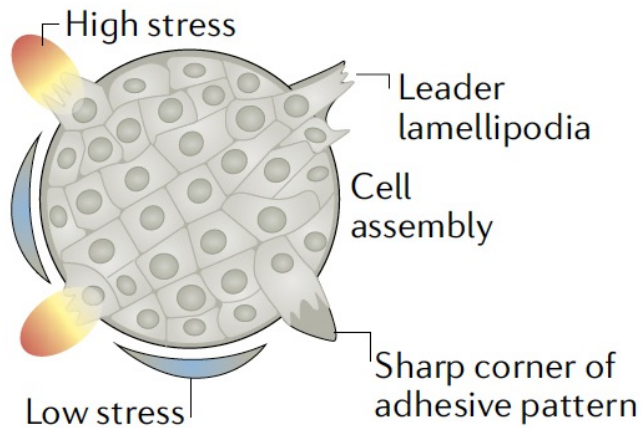
b



Tissue motion and geometric confinement

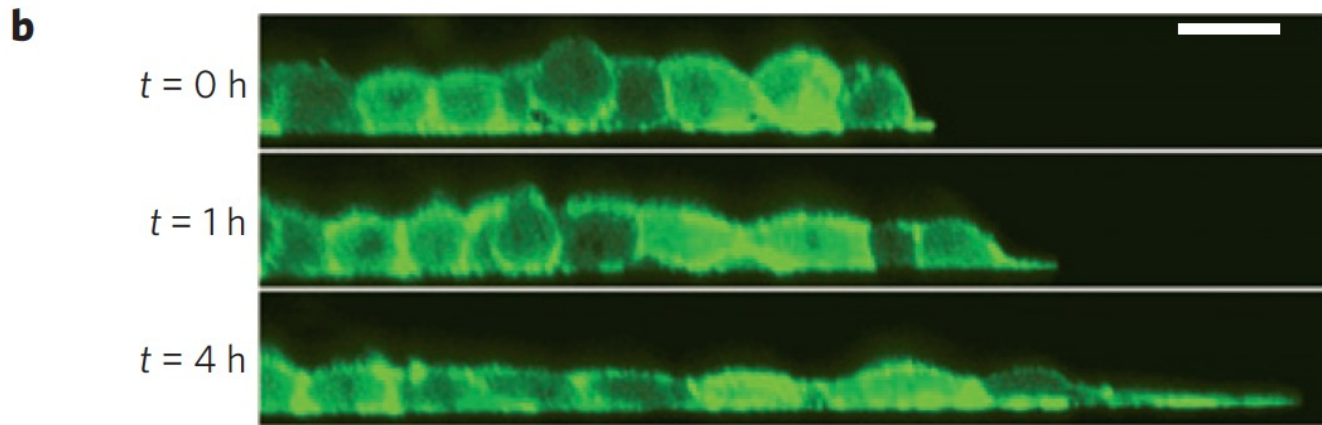
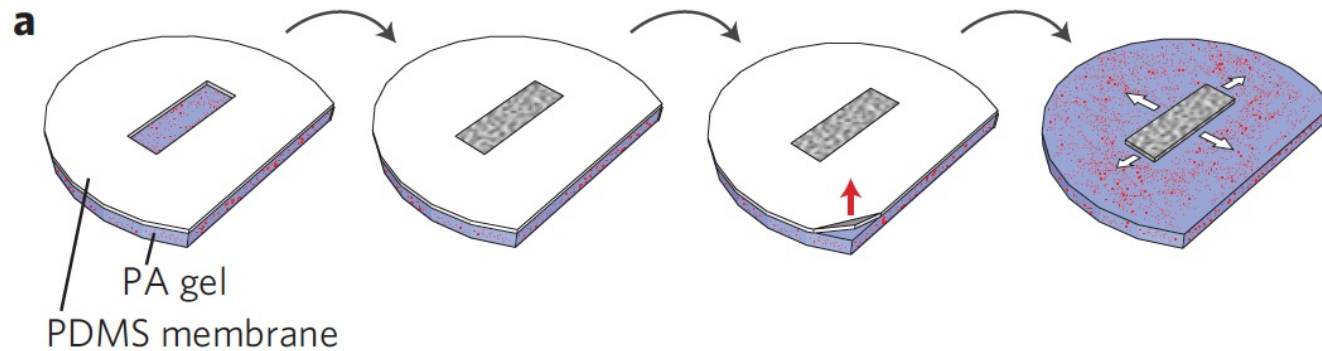


Geometric cues



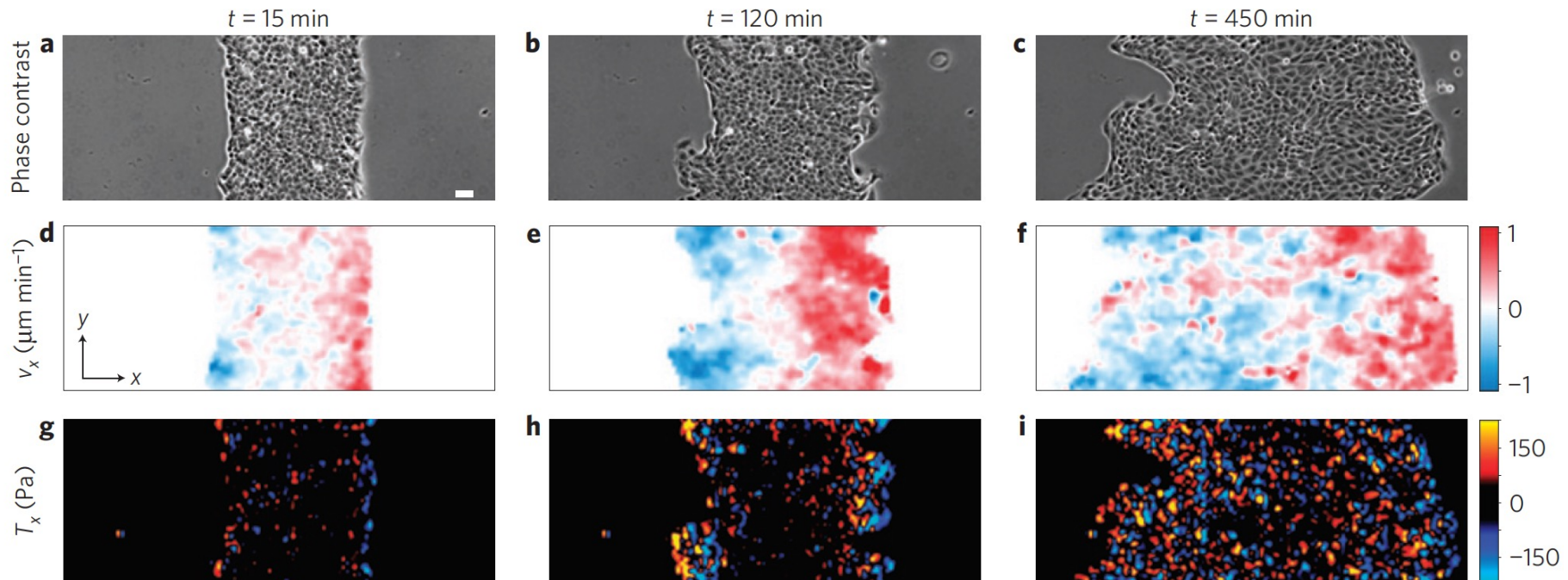
Mechanical waves

- Change in boundary conditions
- Tissue expansion and collective cell migration

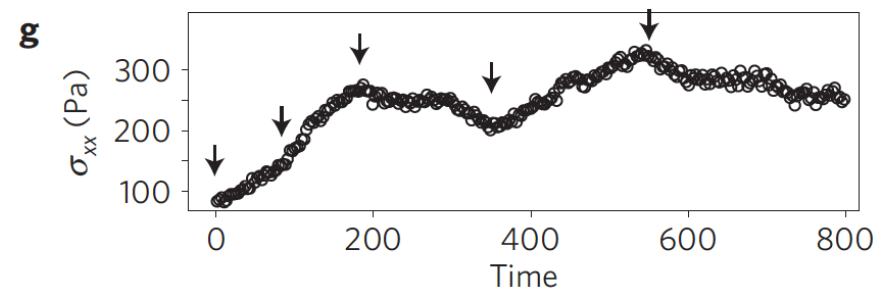
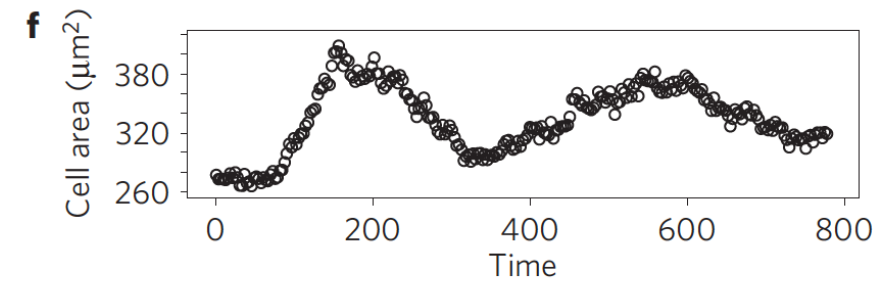
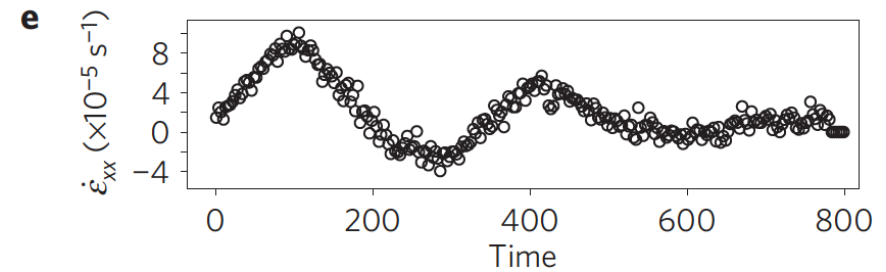
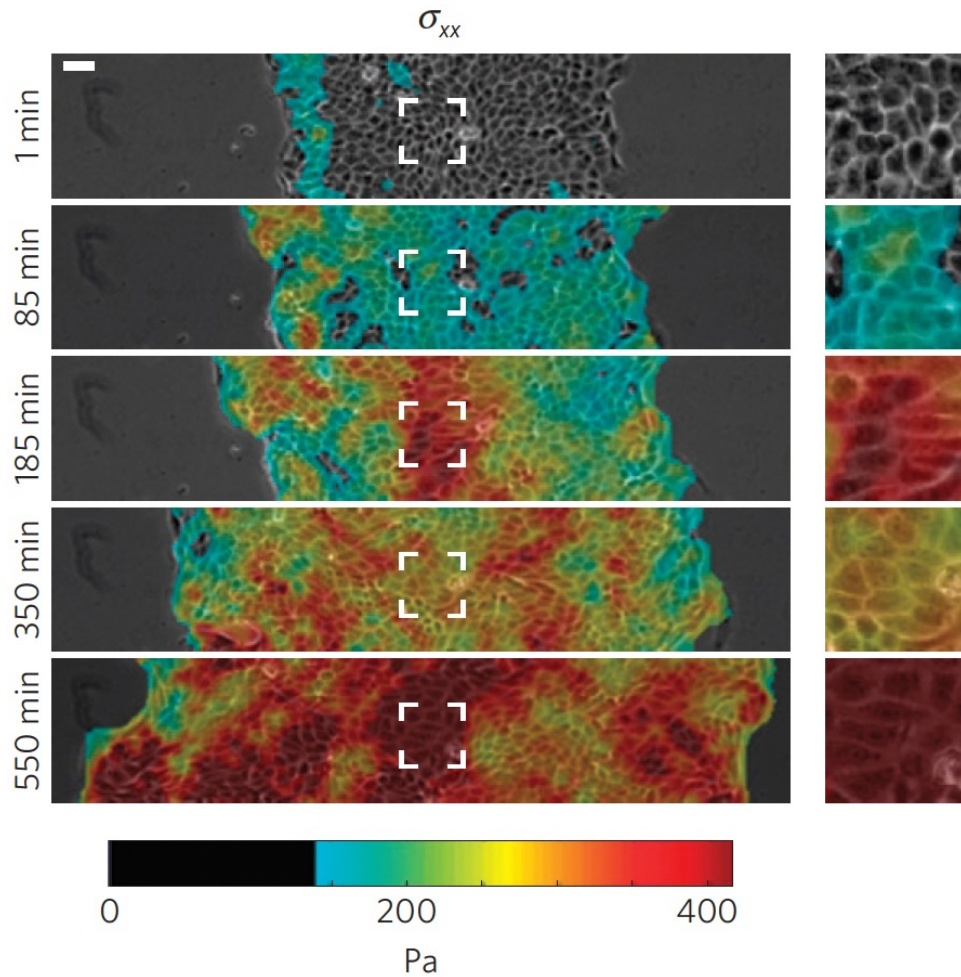


Mechanical waves

- Maps of cell velocity and cell substrate tractions (T_x)
- Initiated at the leading edge and progressively propagate towards the center



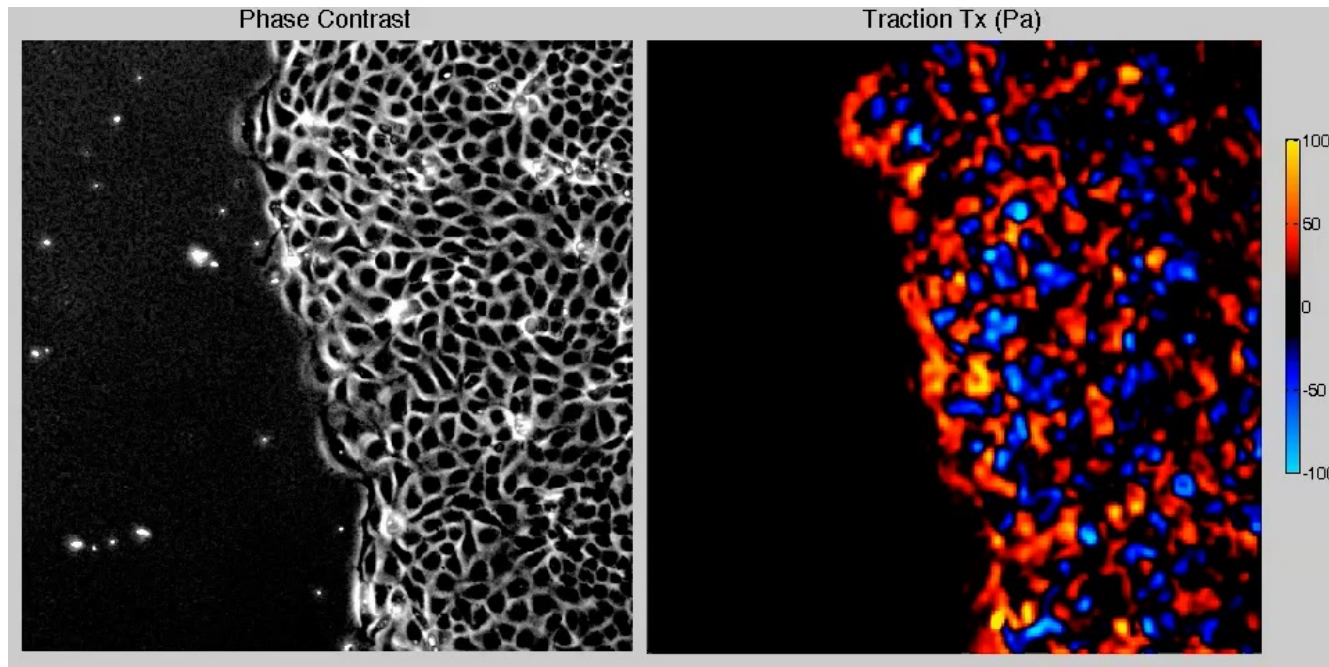
Mechanical waves (movie)



Forces during collective cell migration

- Large tractions are applied by cells many cell rows behind the edge
- Stresses within the cell sheet on a plane perpendicular to the substrate

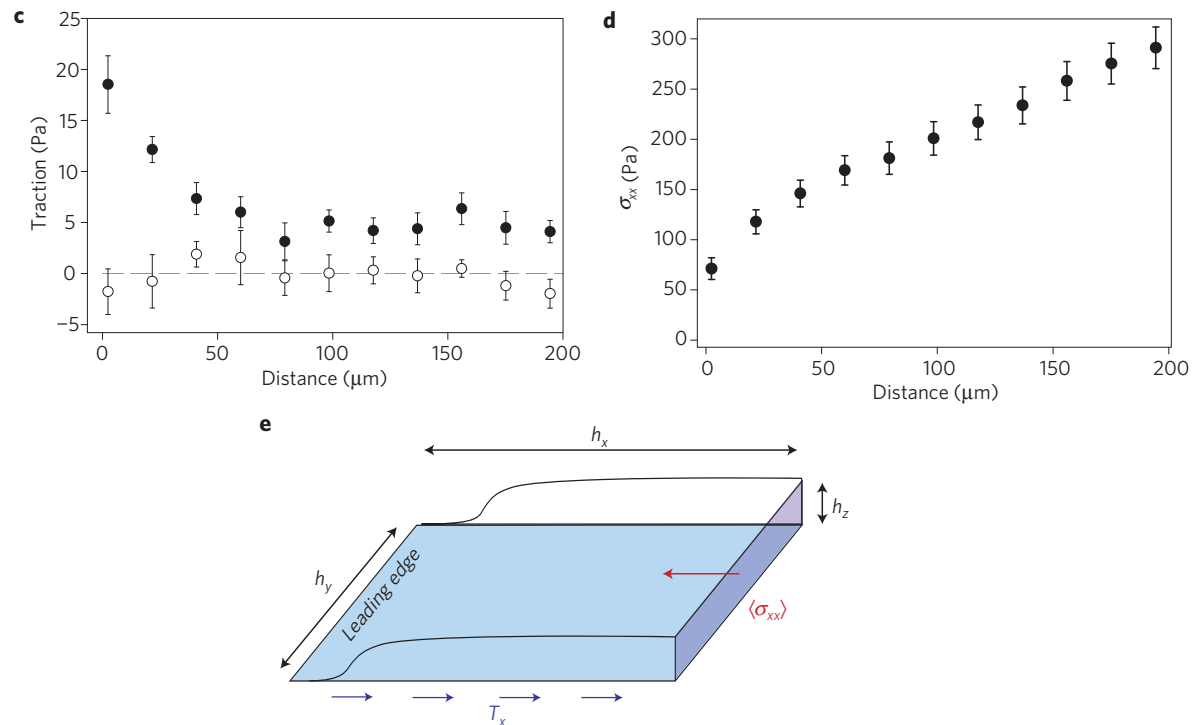
$$\langle \sigma_{xx}(x) \rangle = \frac{1}{h_z h_y} \int_0^x \int_0^{h_y} T_x(x', y') dx' dy'$$



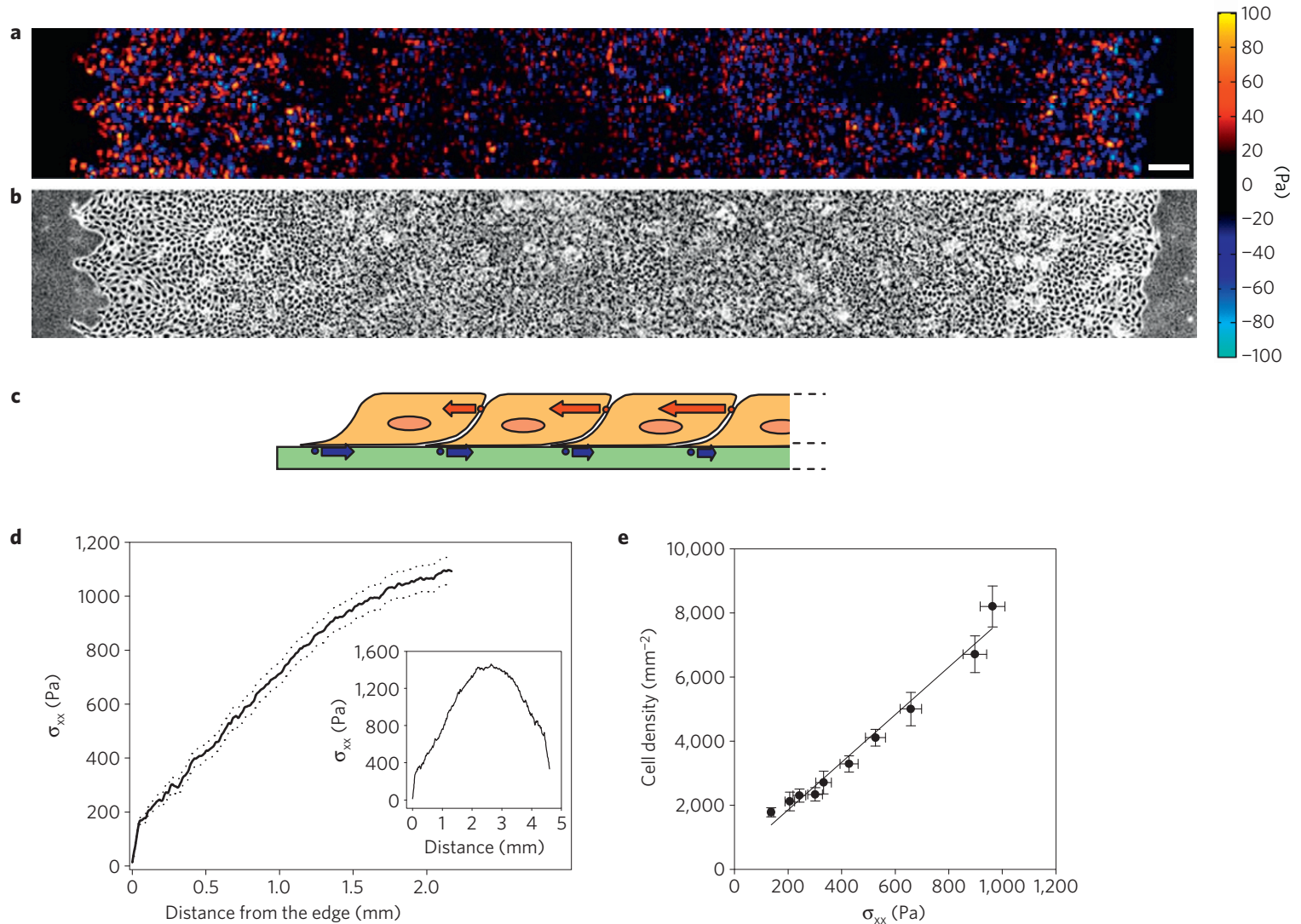
Forces during collective cell migration

- Large tractions are applied by cells many cell rows behind the edge
- Stresses within the cell sheet on a plane perpendicular to the substrate

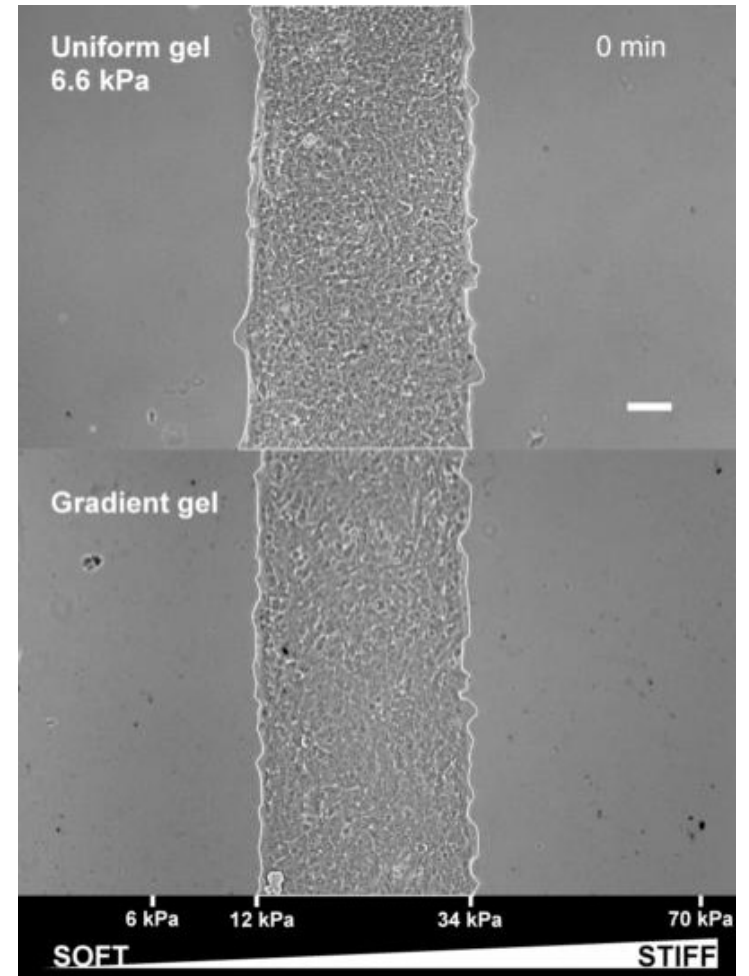
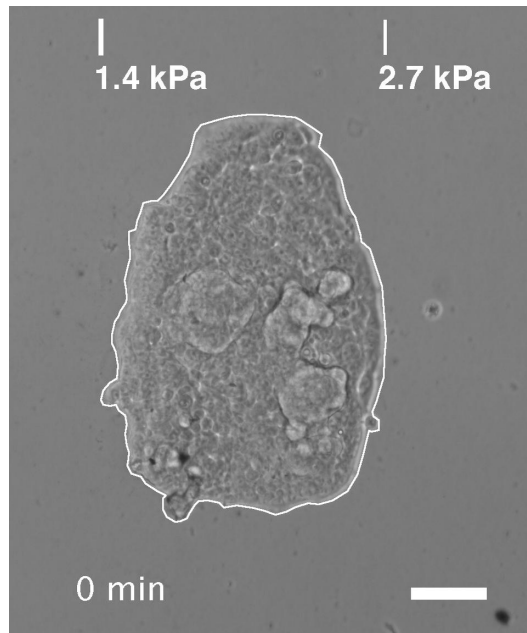
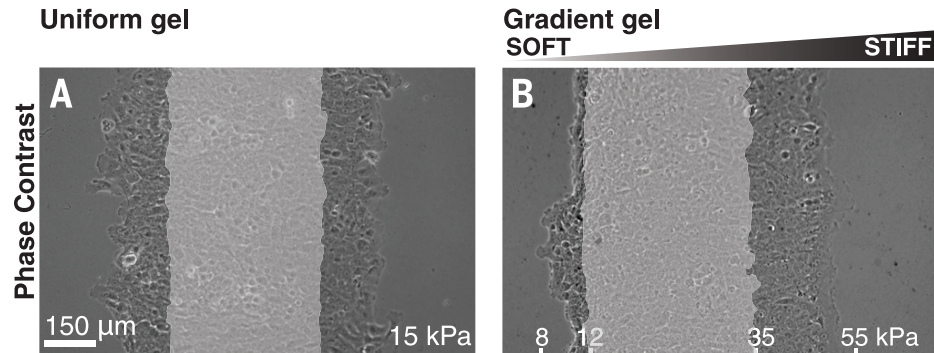
$$\langle \sigma_{xx}(x) \rangle = \frac{1}{h_z h_y} \int_0^x \int_0^{h_y} T_x(x', y') dx' dy'$$



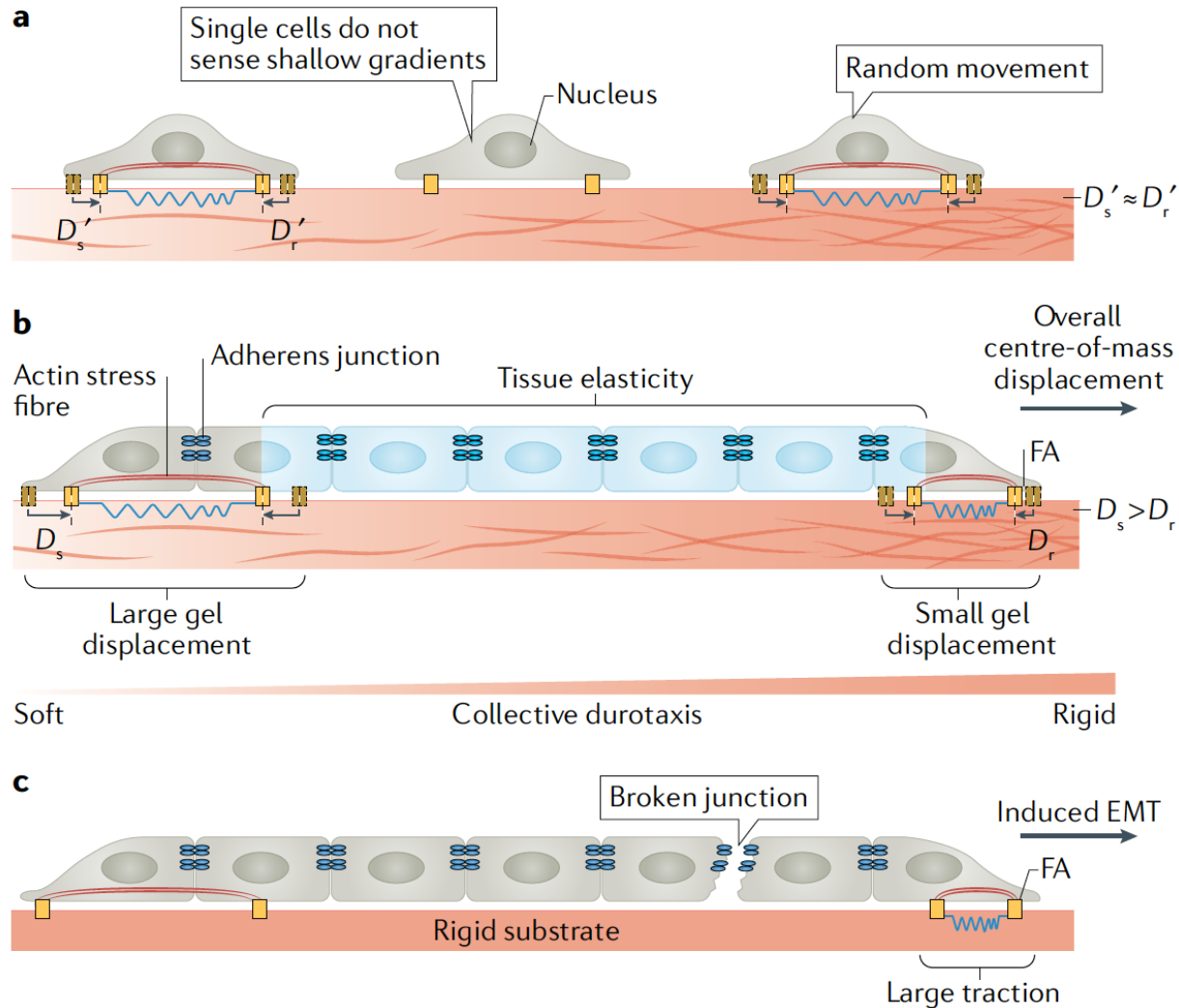
Forces during collective cell migration



Collective durotaxis

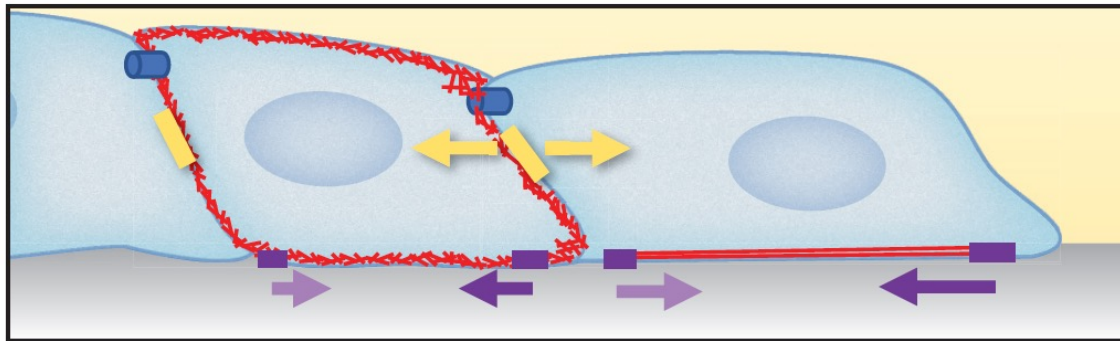


Collective durotaxis



Forces and interactions of migrating cells

a Side view



Biological structures

- Focal adhesions
- Gap junctions
- Adherens cell-cell junctions
- Actomyosin cortex
- Actomyosin stress fibers

Contact regulation of locomotion (CRL)

- Contact following of locomotion
- Contact inhibition of locomotion

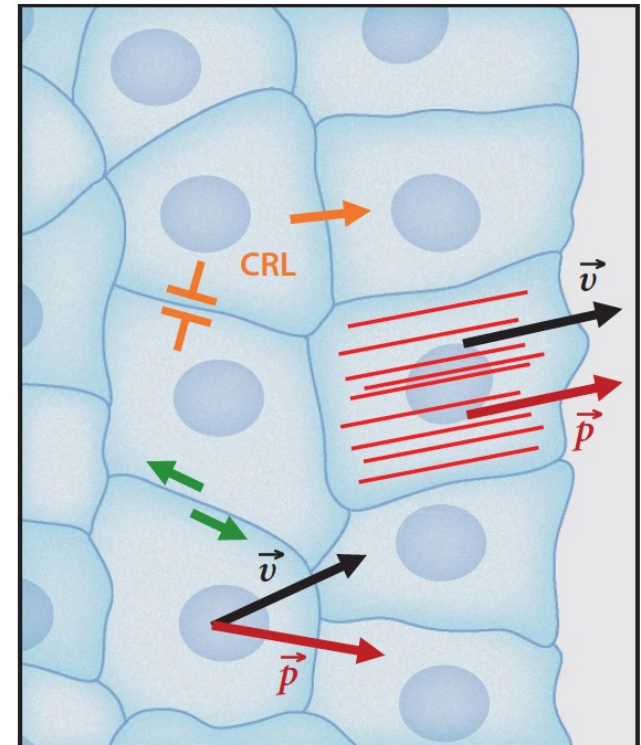
Variables

- Cell velocity $\vec{v}$
- Cell polarity $\vec{p}$

Forces

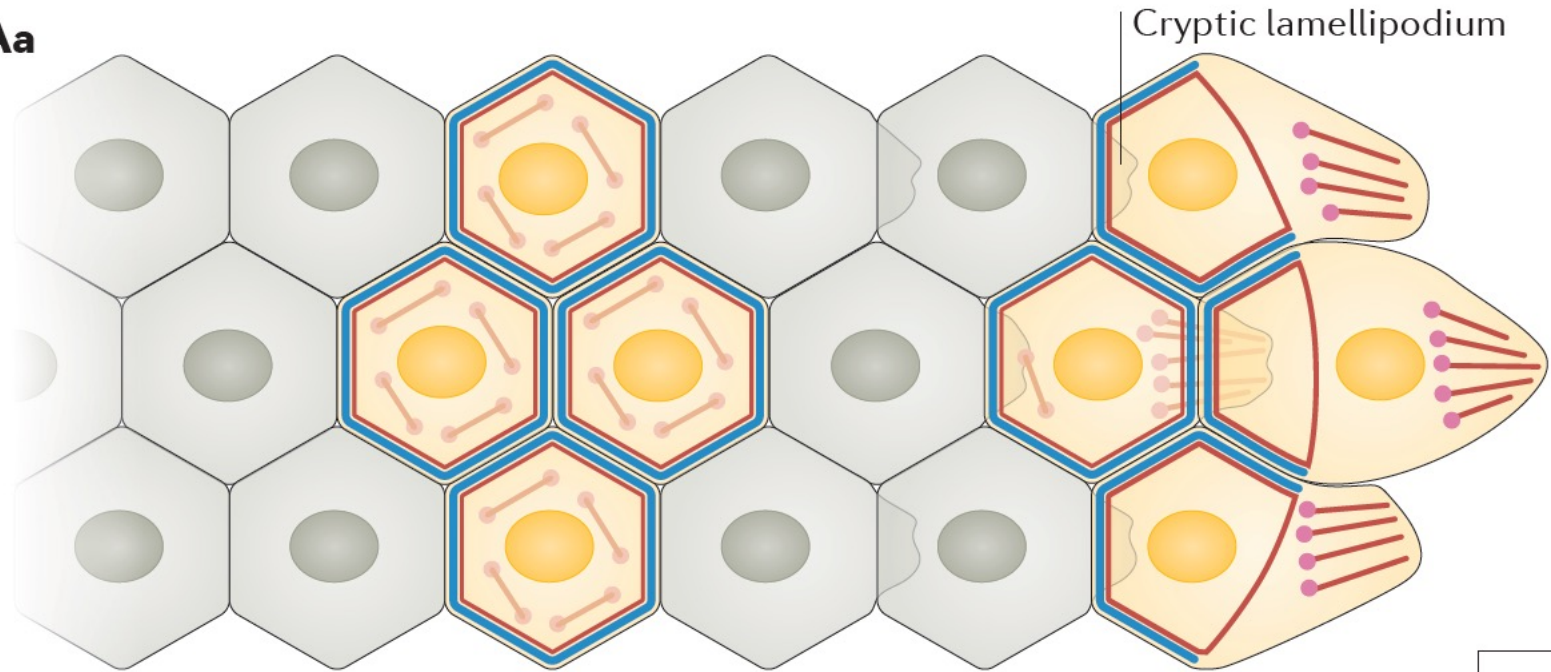
- Active traction
- Cell-substrate friction
- Cell-cell tension
- Cell-cell friction

b Top view



Forces and interactions of migrating cells

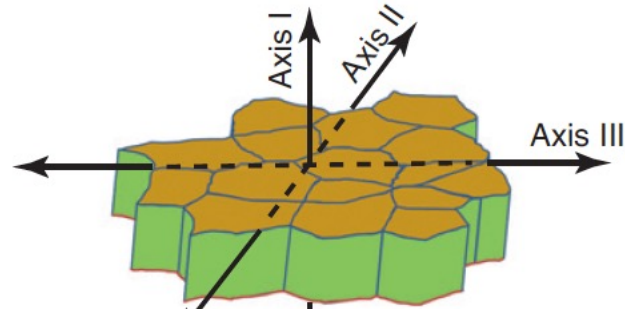
Aa



- Junctional components
- Actomyosin
- Focal adhesions

Epithelial tissue mechanics: basics

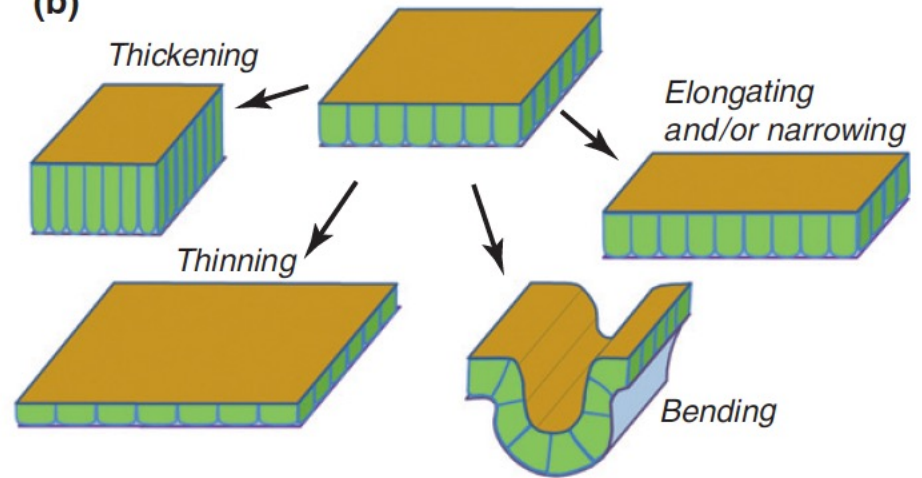
(a)



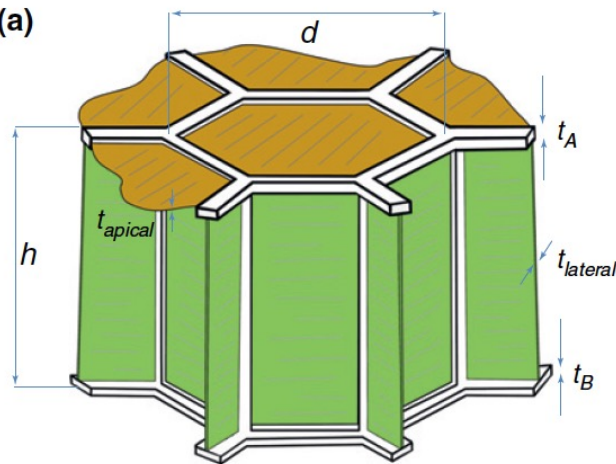
Axis II and III (planar axes):
e.g. left-right, dorsal-ventral,
anterior-posterior, oral-aboral
proximal-distal ...

Axis I: e.g.
apical-basal,
inside-outside,
interface-deep
ECM-free...

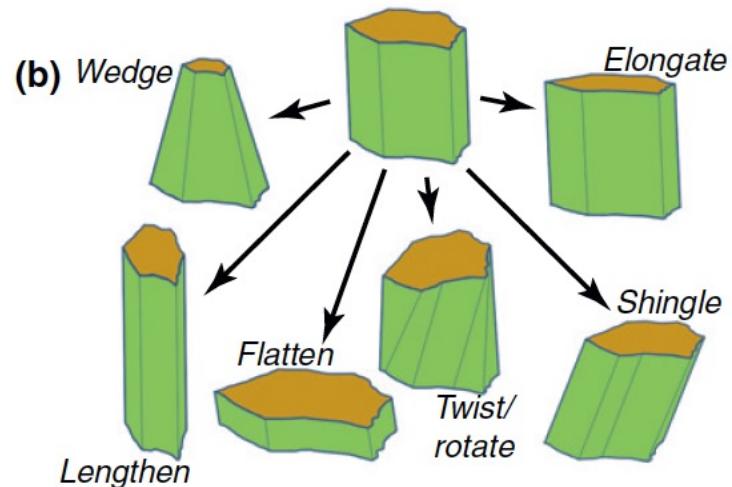
(b)



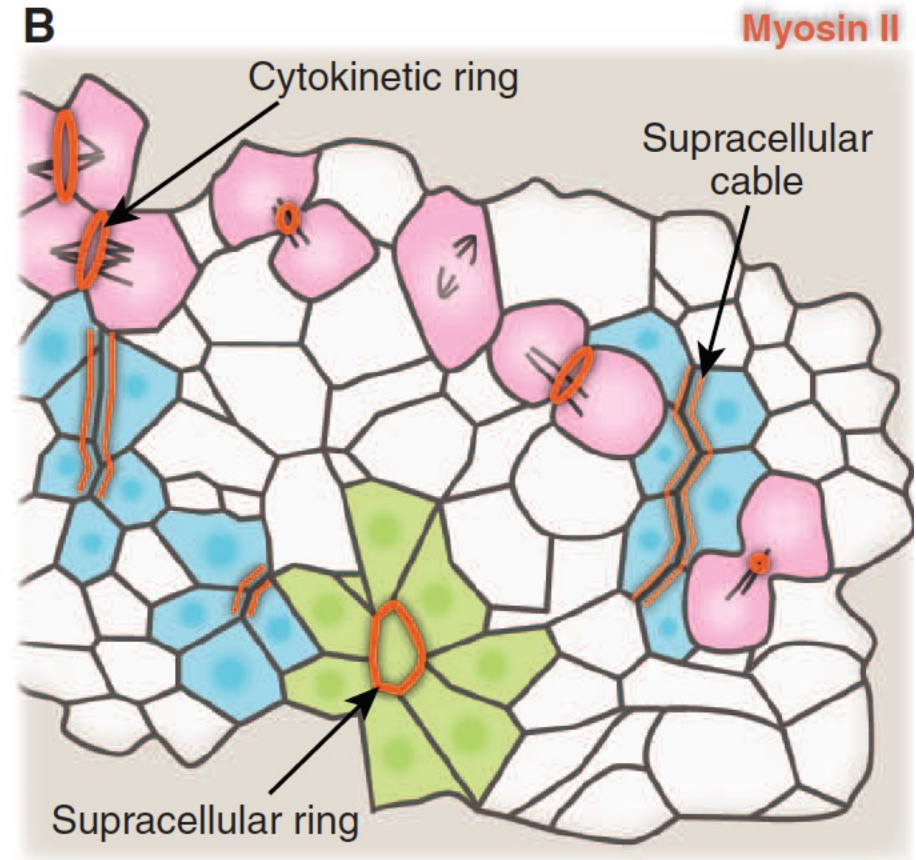
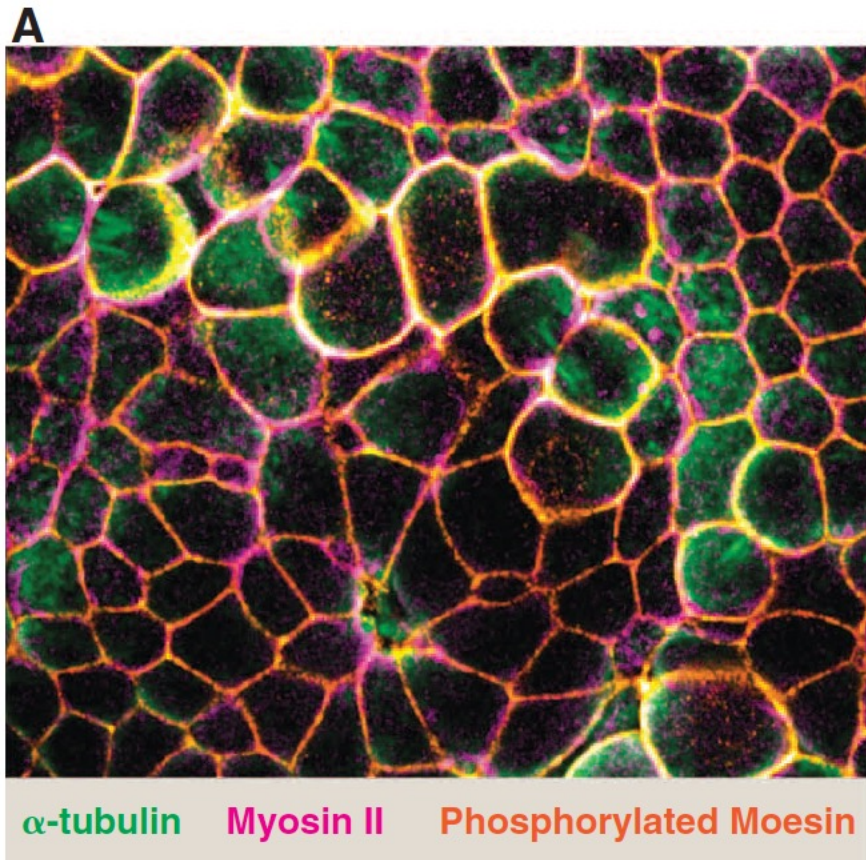
(a)



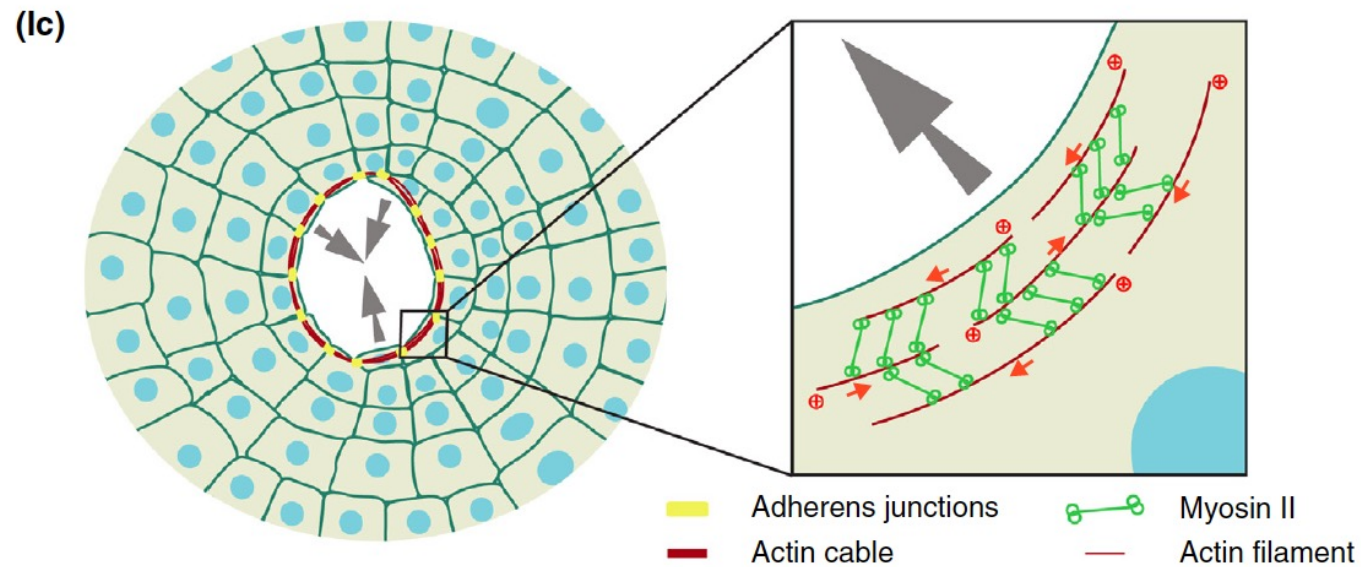
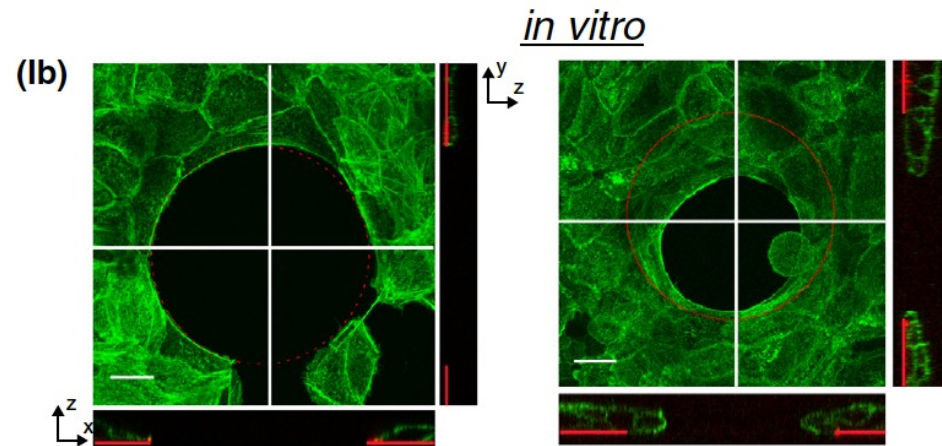
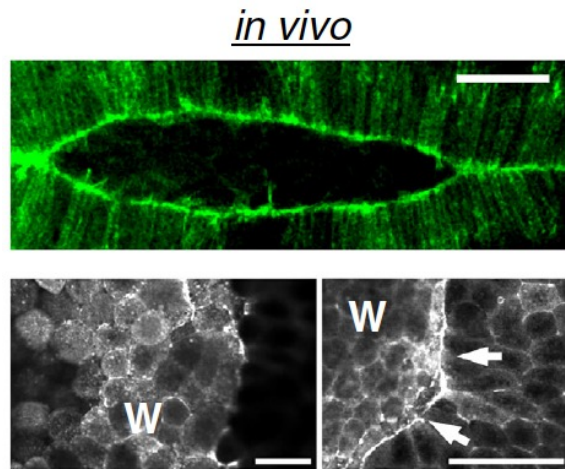
(b)



Epithelial tissue mechanics: actomyosin structures

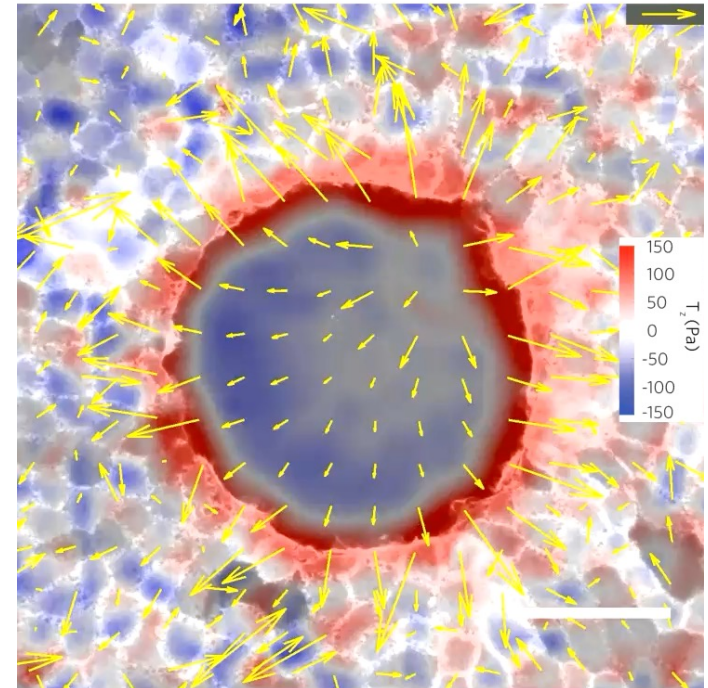
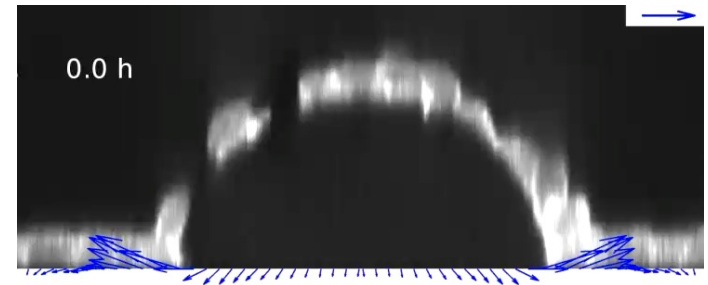
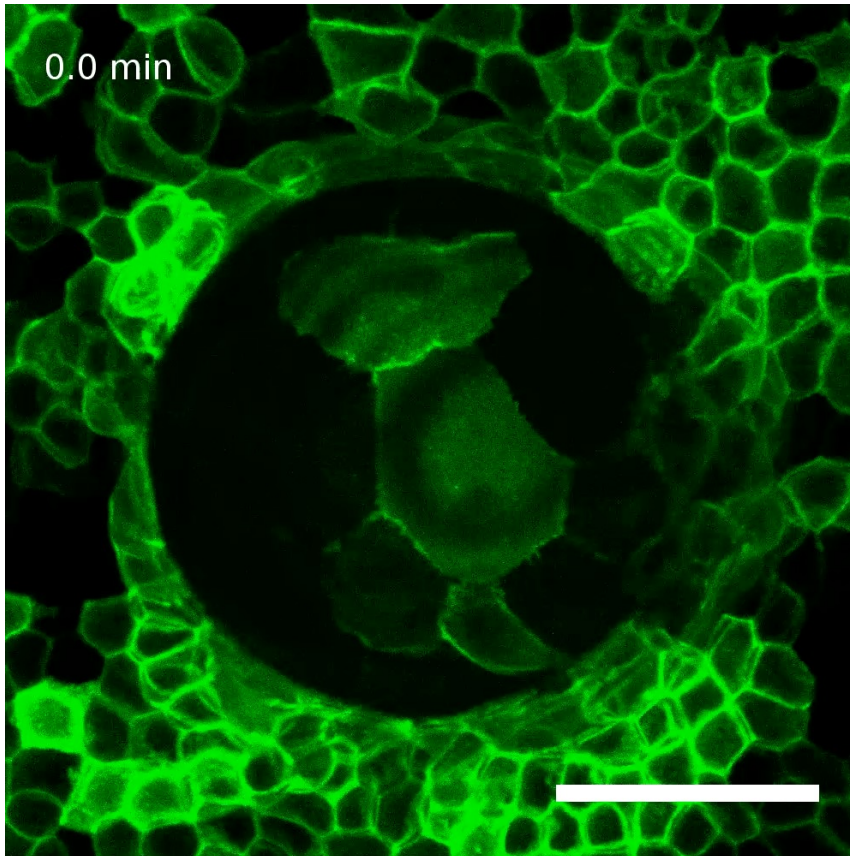


Supracellular cable

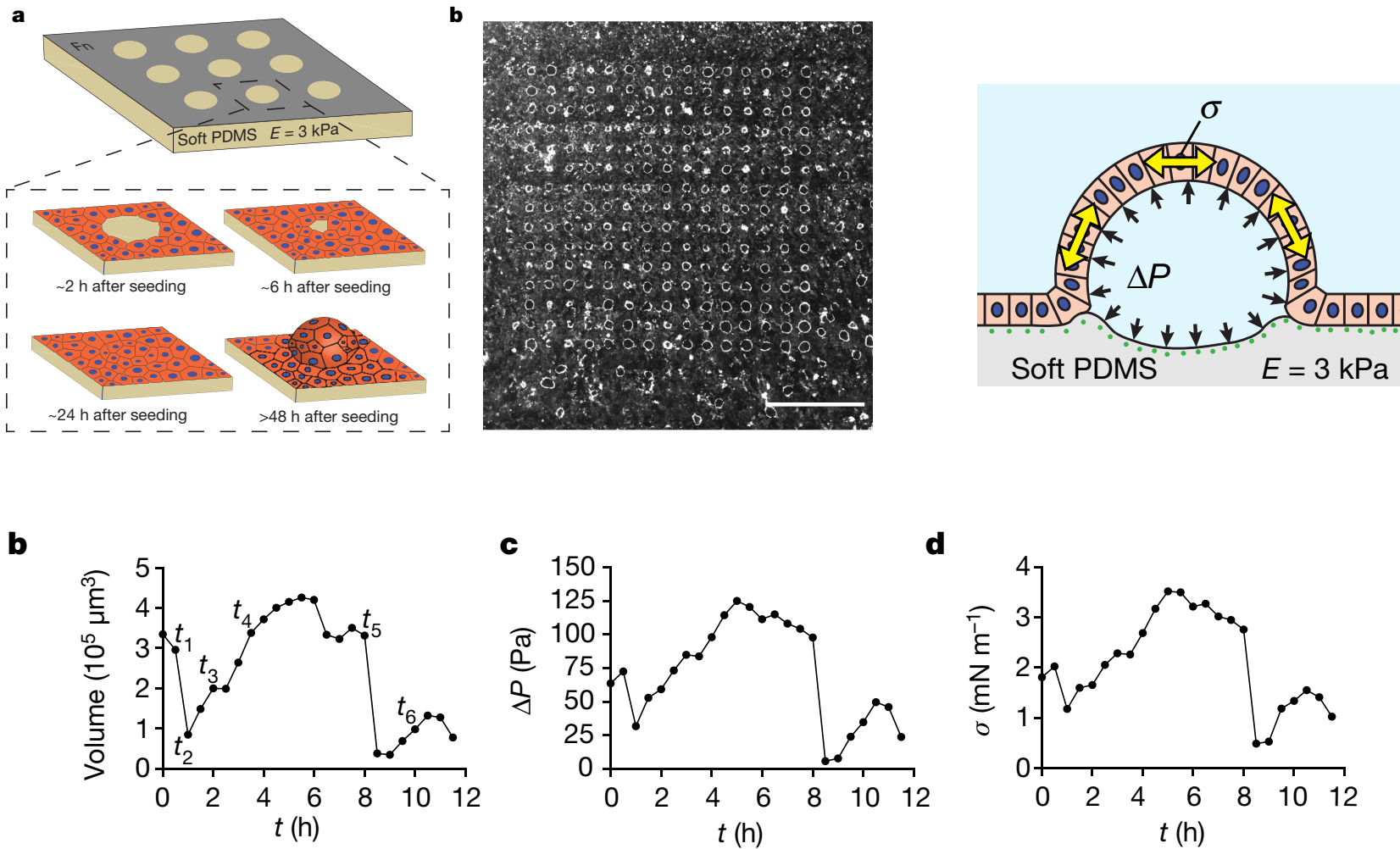


Active superelasticity

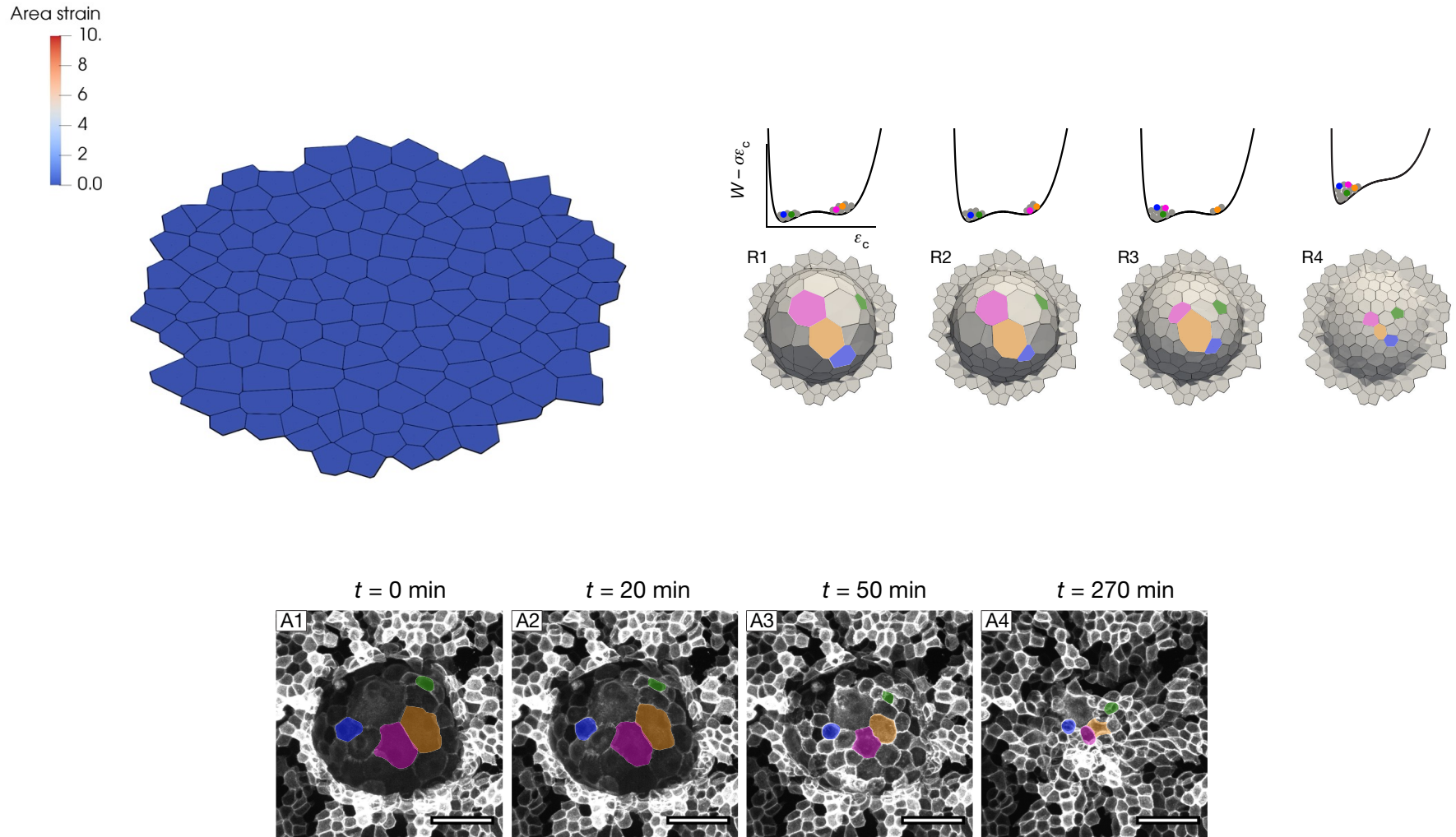
- Arrays of epithelial domes with controlled geometry
- Quantification of luminal pressure and epithelial tension



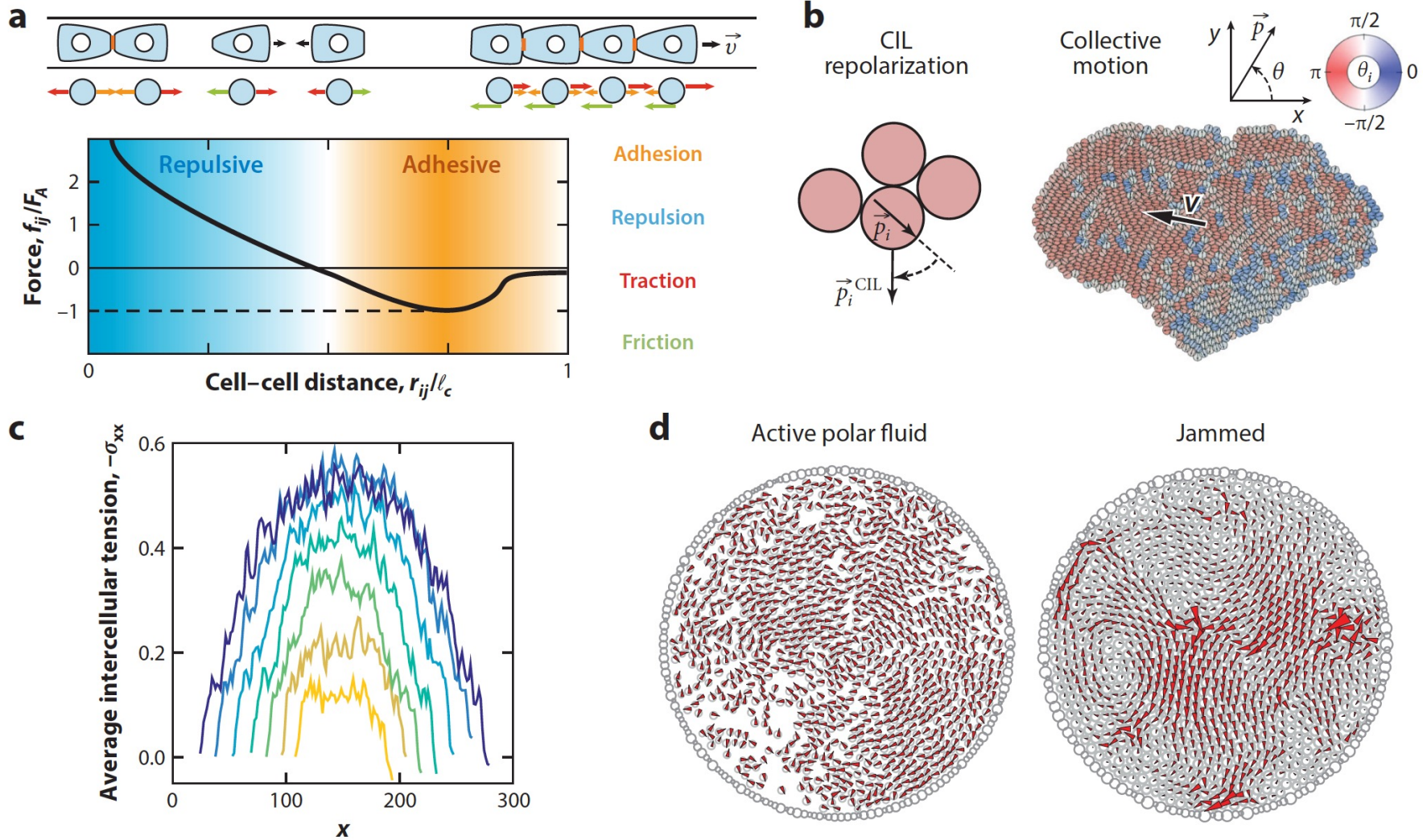
Active superelasticity



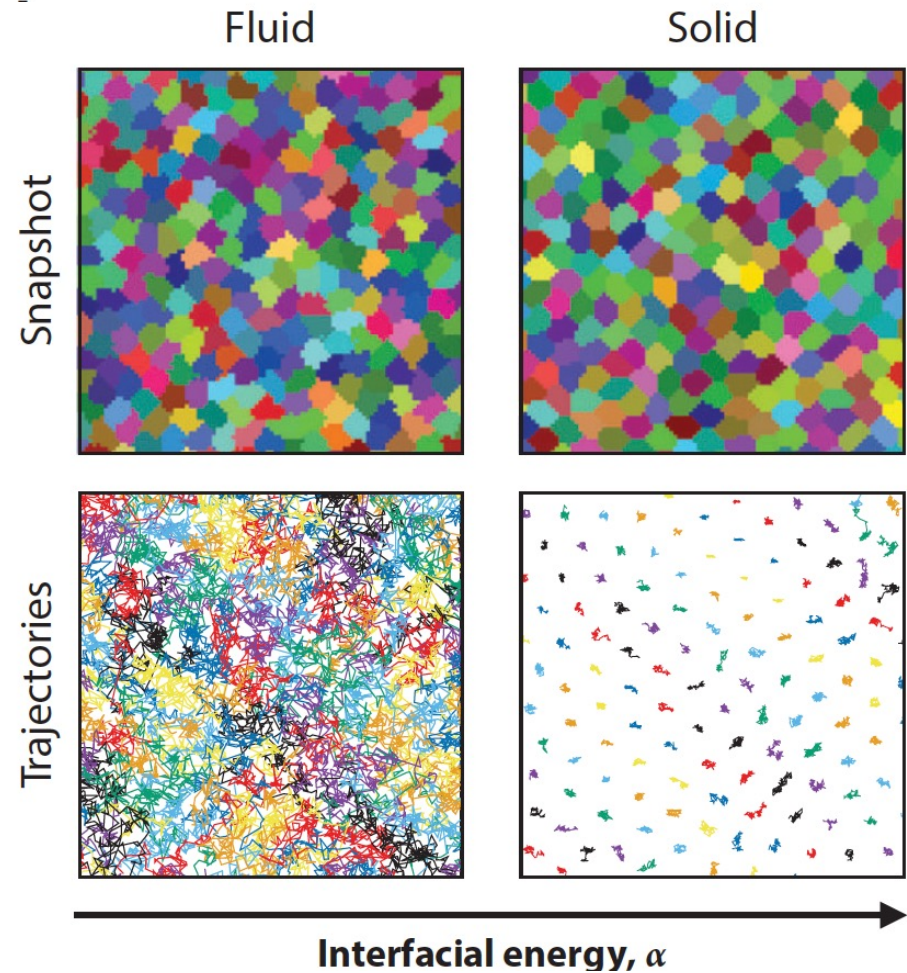
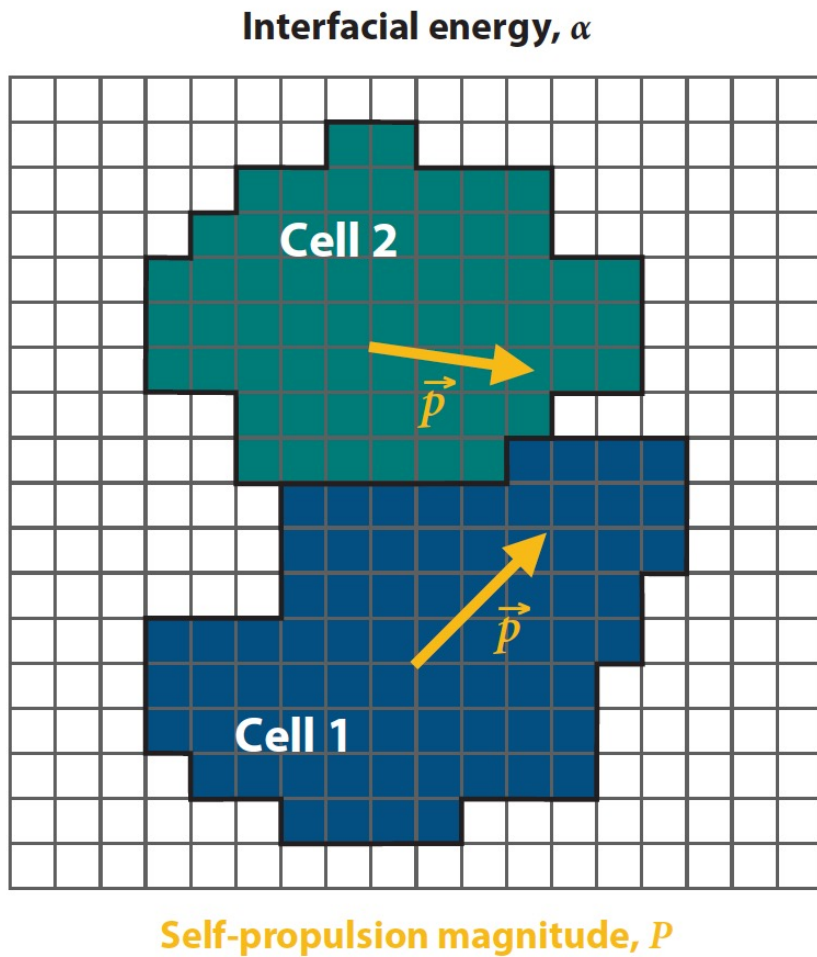
Active superelasticity



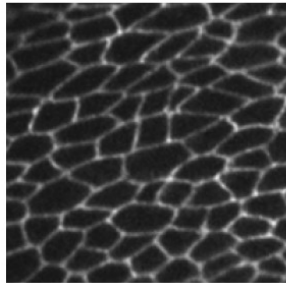
Particle Models



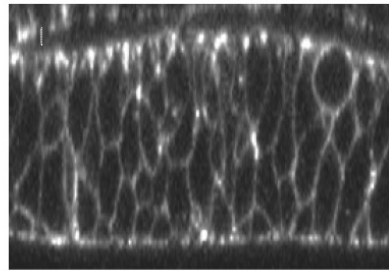
Lattice Models



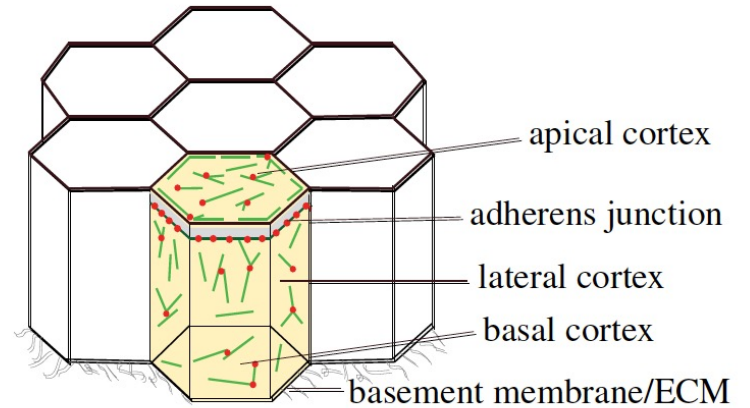
Vertex Models

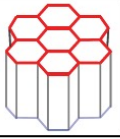
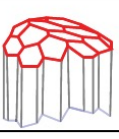
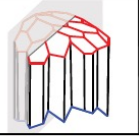
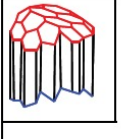


apical view

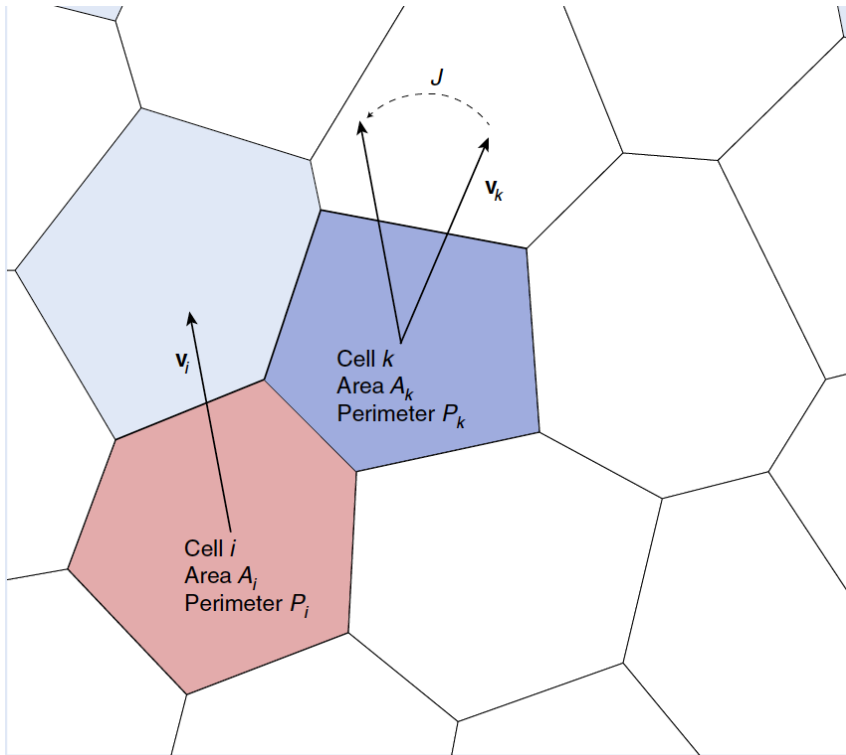


lateral section



2D apical vertex model		3D apical vertex model		2D lateral vertex model		3D vertex model	
							
(c)		(d)		(e)		(f)	
flat apical surface		apical surface		lateral cross section		apical and basal surface	

2D Vertex Models



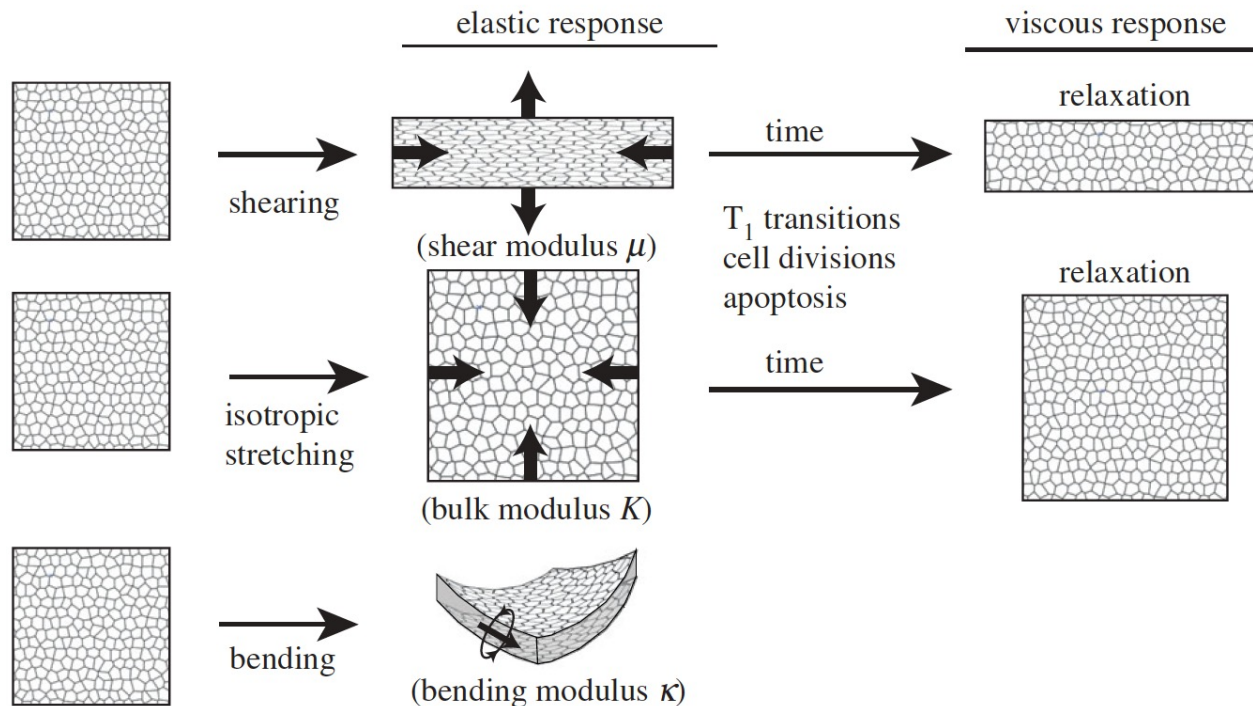
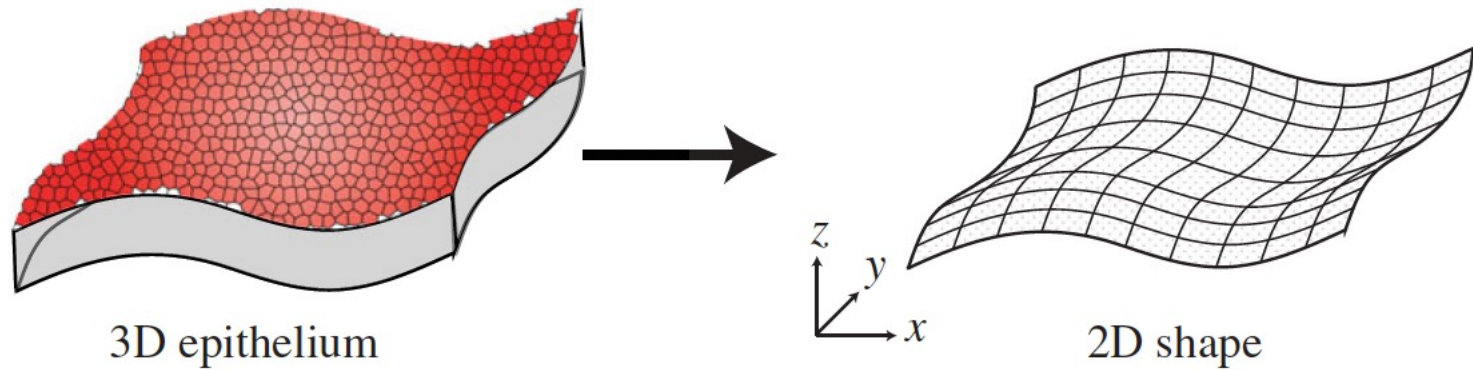
Equation of Motion

$$\frac{d\mathbf{x}_k}{dt} = \mu \mathbf{F}_k$$

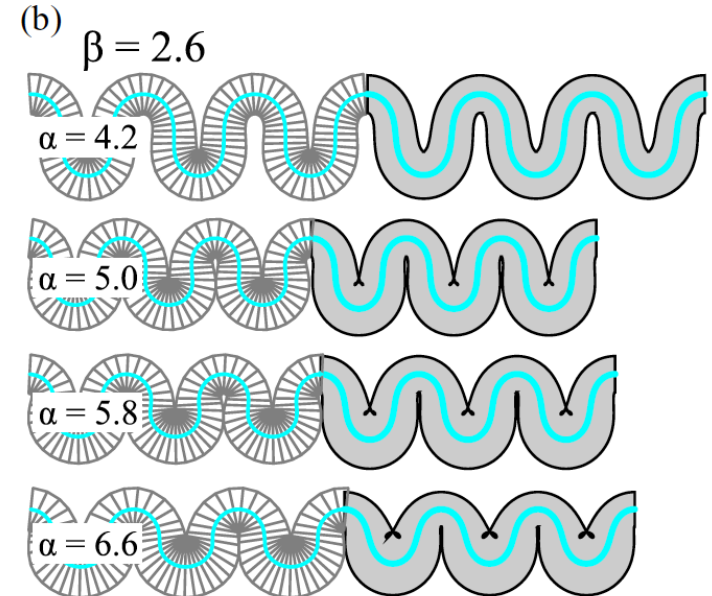
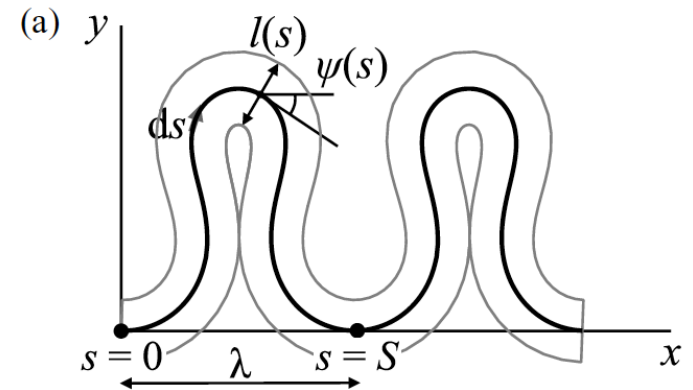
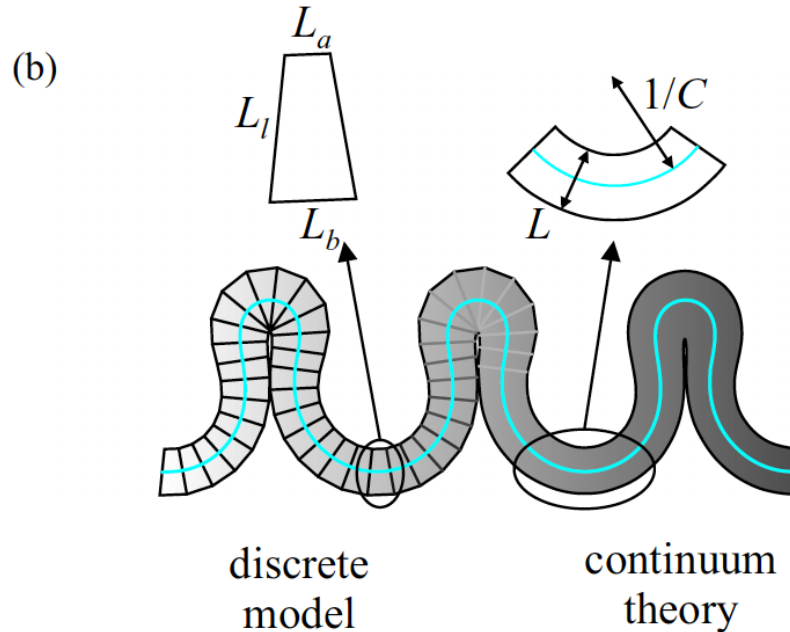
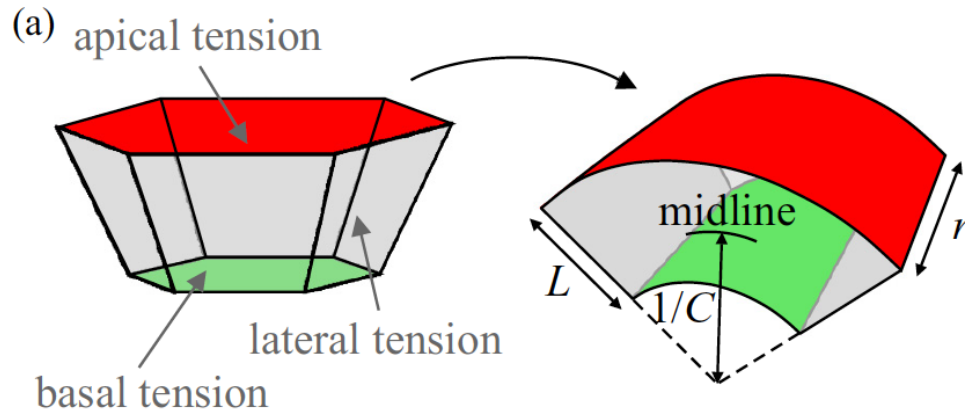
Energy Function

$$E_{\text{monolayer}} = \sum_i E_i = \sum_i K_{A_i} (A_i - A_{i0})^2 + K_{P_i} (P_i - P_{i0})^2$$

Continuum approach by coarse graining vertex model



Continuum approach by parametrization (elastica)



Models of cells and tissues at different scales

- Focal adhesions, actin, clutch model, surface tension, and continuum mechanics

