Mechanics of tissue growth, homeostasis and repair
Part 1

Boulder Summer School 2024

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The Gang in London

UCL

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The Lister Institute of Preventive Medicine
CONACYT
LEVERHULME TRUST
PolarNet
EMBO Young Investigator Programme
UKRI Medical Research Council
wellcome
MARIE CURIE ACTIONS
European Commission
For Women in Science
NC3Rs
UCL IPLS
@YanlanMao

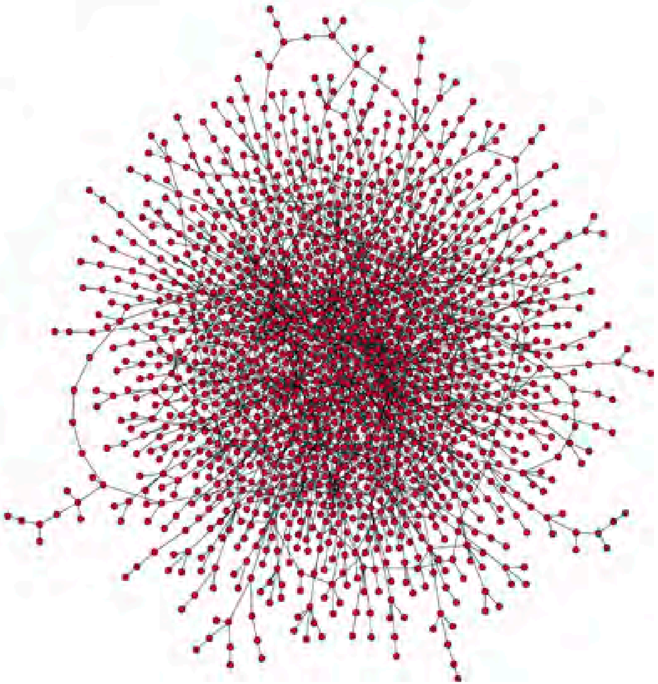
“..the genetic instructions and building blocks used by all organisms are the same”

Kelvingrove Art Gallery, Glasgow
Source: Yanlan’s iPhone

“ It’s all in our genes ”

Genes → ?? → Shape

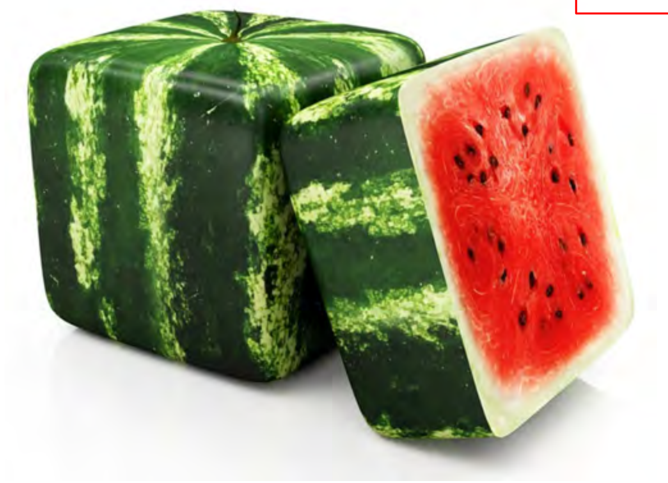
OMICs



The importance of mechanical forces

Mechanics

Cell Extrinsic Factors
- e.g. Basement membrane
- External forces



Genes

Shape

Role of mechanical forces

During development, homeostasis and repair, how do **<u>forces and molecules</u>** interact for tissues to:

Get into Shape

Stay in Shape

Get back into Shape

Structure of lectures

Day 1: Growth and morphogenesis

i) some biomechanics background

ii) case study on vertex modelling of tissue shape control

Day 2: Homeostasis and repair

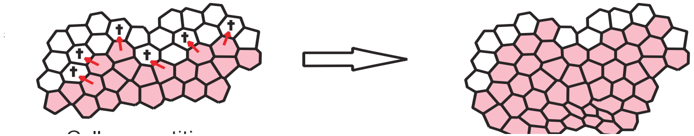
i) latest research – new tools and methods

ii) case study on vertex modelling of tissue repair

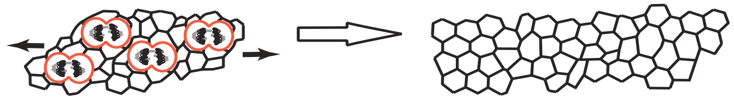
Exercise: modelling lymph node mechanics

Cellular mechanisms of tissue shape control

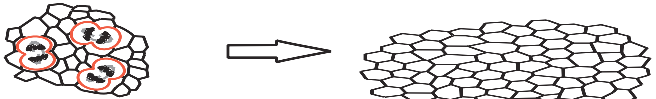
1. Growth rates



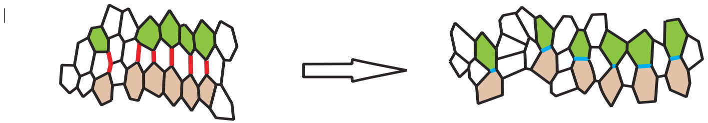
2. Division orientations



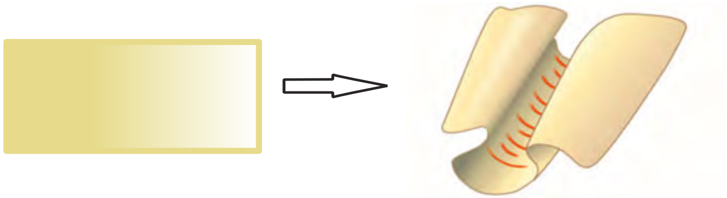
3. Cell shape changes



4. Planar cell rearrangements



5. 3D tissue sculpting



Mechanical control of cellular mechanisms

1. Growth rates

2. Division orientations

3. Cell shape changes

4. Planar cell rearrangements

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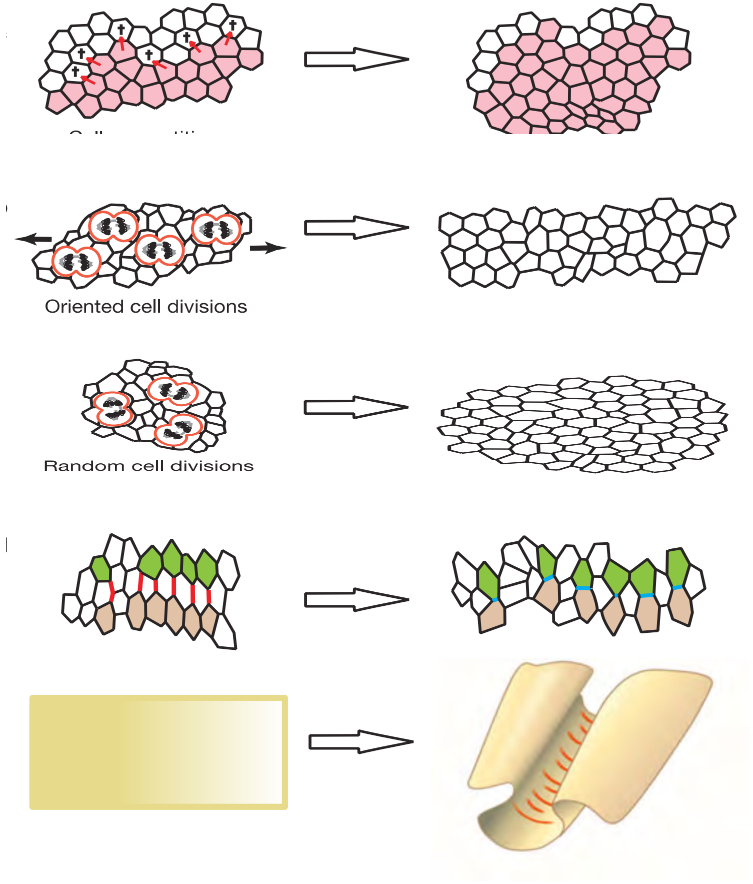
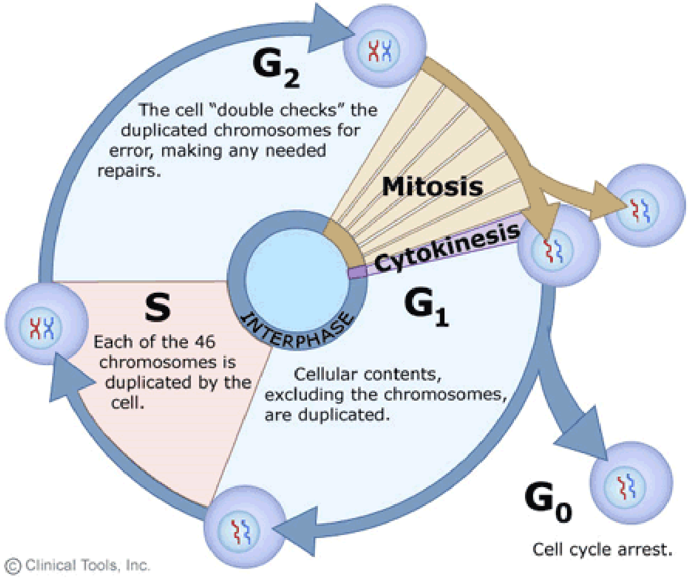
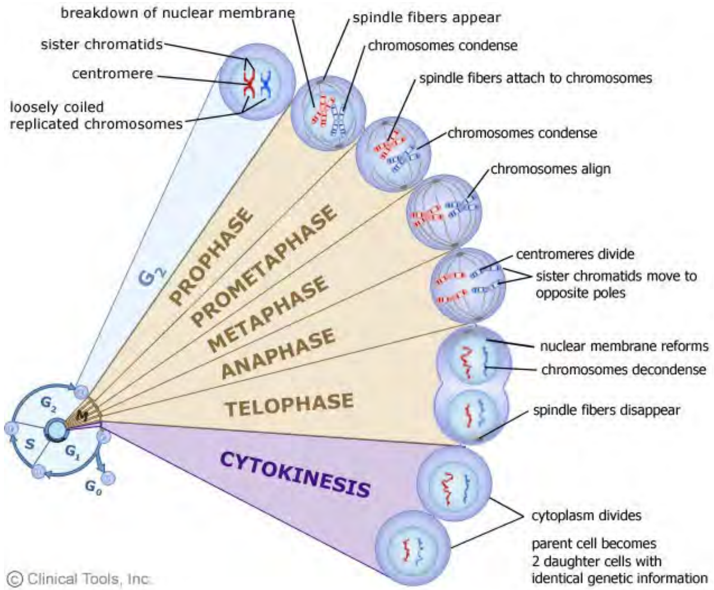


1. Mechanical control of growth rates

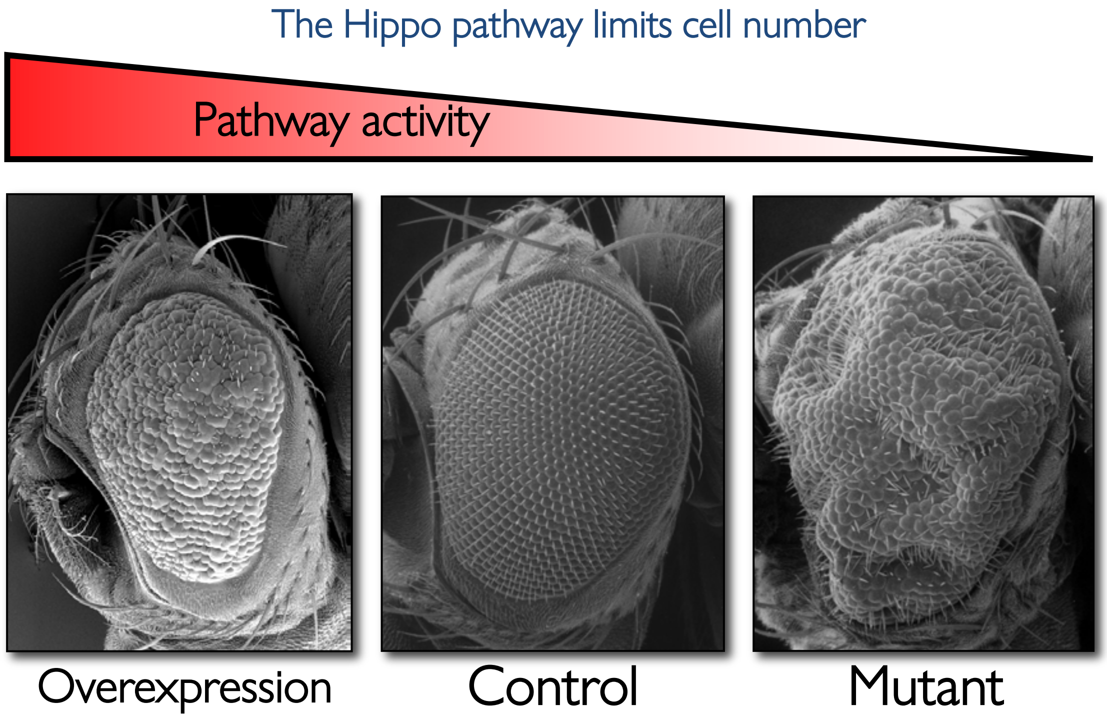
Many growth factors and signalling pathways regulate the cell cycle





1. Mechanical control of growth rates

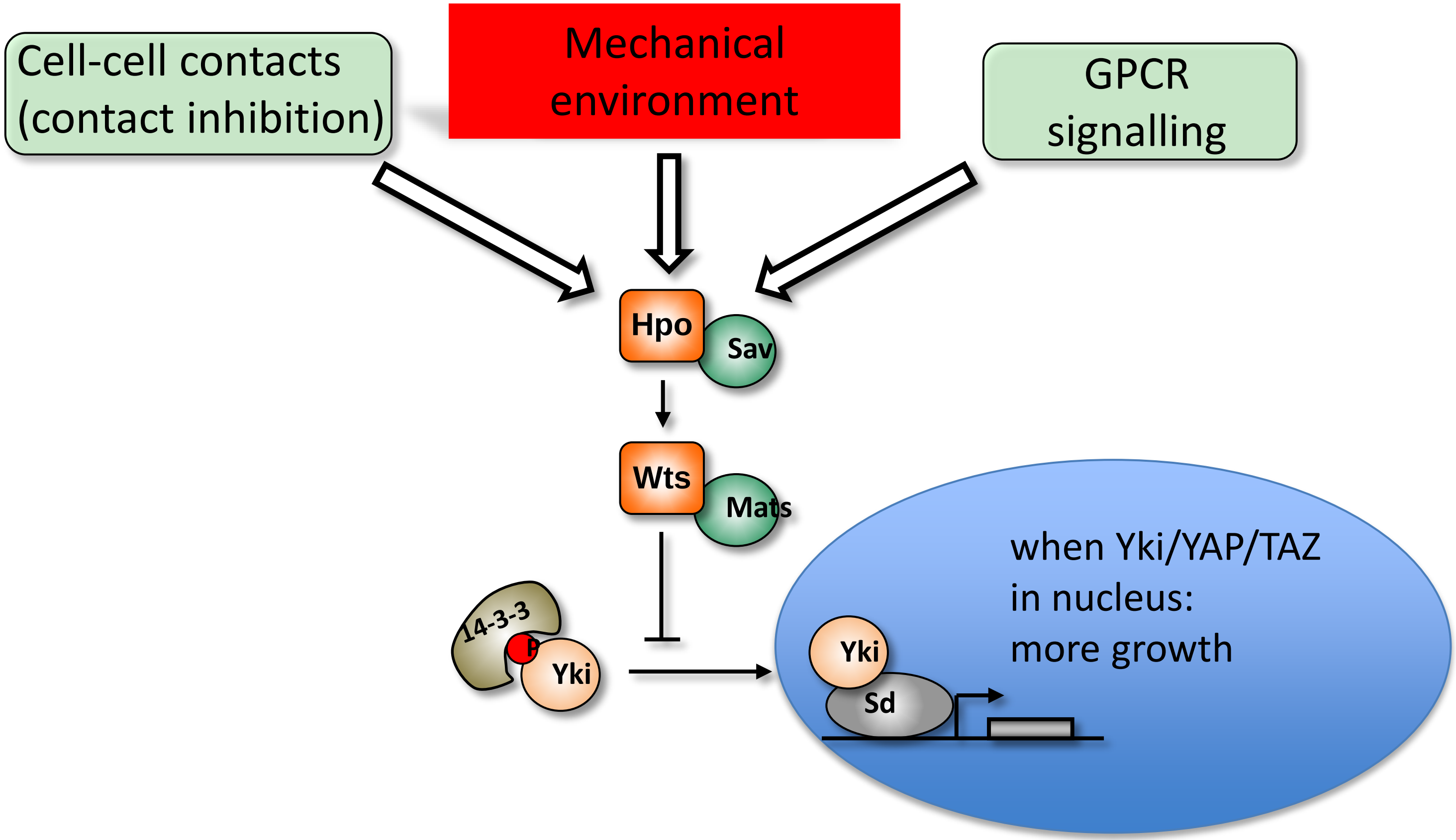
A. Some growth factors are also sensitive to mechanics, e.g. Yki/YAP/TAZ



Bryant, Xu, Hariharan, Pan, Halder, McNeill, Hafen, Harvey, Tapon, Jiang labs…

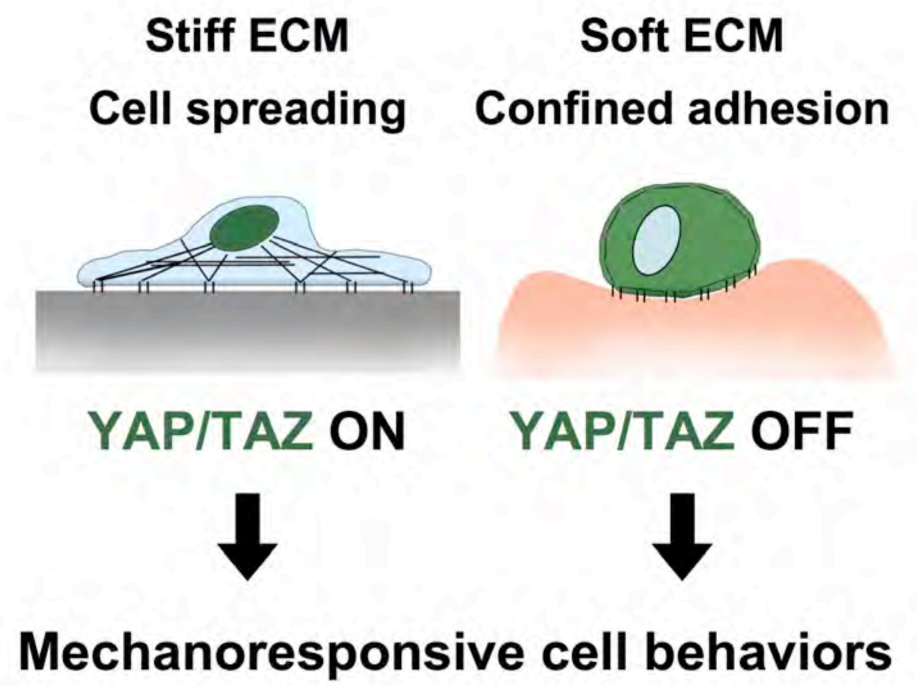
1. Mechanical control of growth rates

Hippo pathway upstream signals



1. Mechanical control of growth rates

A. Effect of mechanics on growth factors – transcriptional co-activators Yki/YAP/TAZ



Role of YAP/TAZ in mechanotransduction. Sirio Dupont,…,Stefano Piccolo. Nature 9 June 2011

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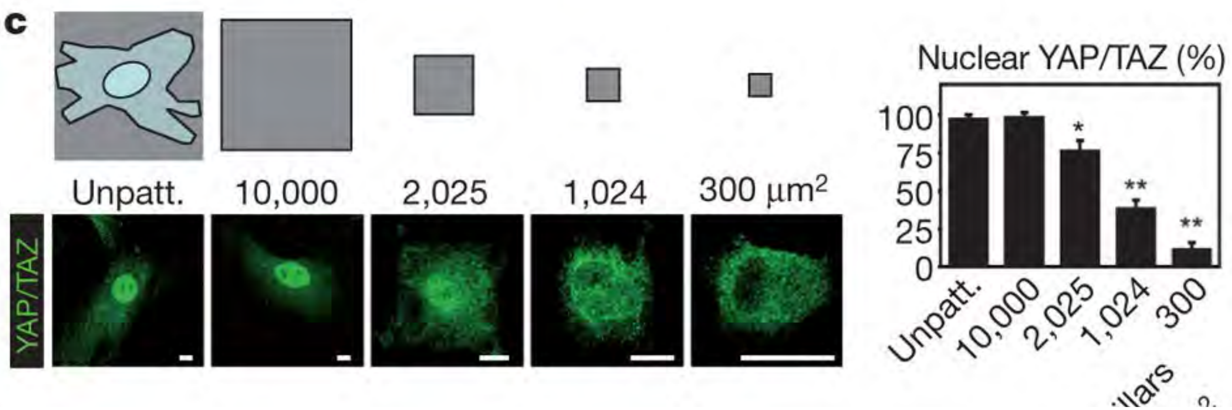

Micropatterning to constrain shape

Role of YAP/TAZ in mechanotransduction. Sirio Dupont,…,Stefano Piccolo. Nature 9 June 2011

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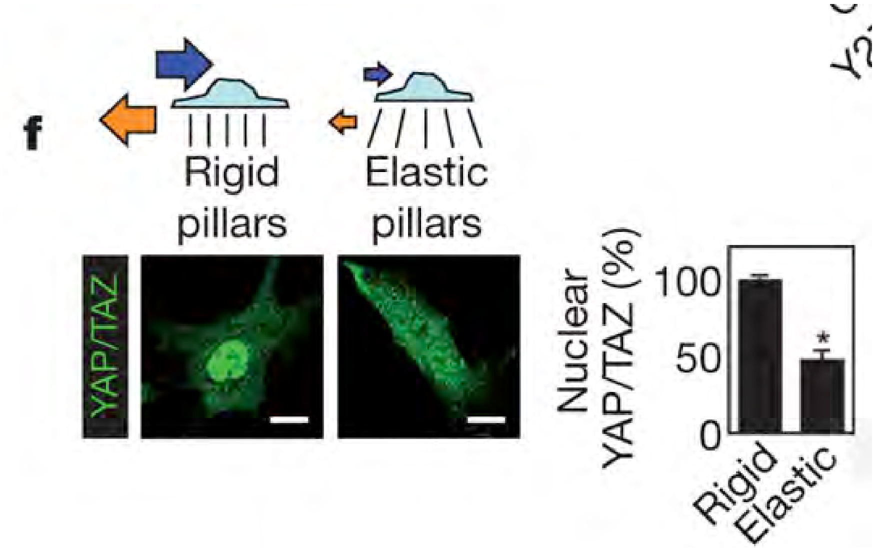
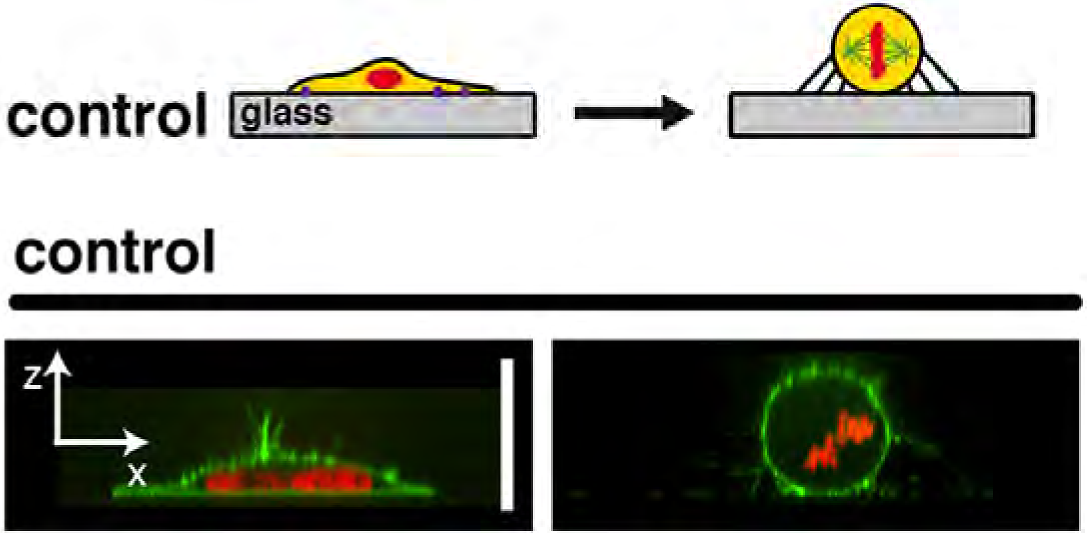


Role of YAP/TAZ in mechanotransduction. Sirio Dupont,…,Stefano Piccolo. Nature 9 June 2011

1. Mechanical control of growth rates

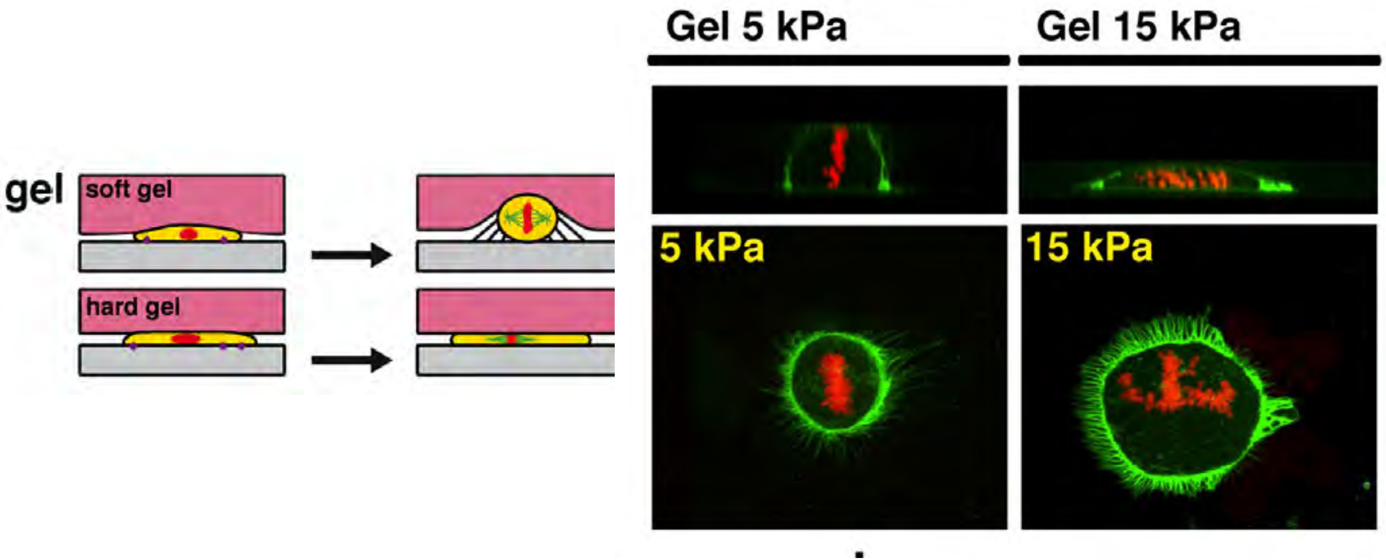
B. Effect of mechanics on mitosis



Mitotic Rounding Alters Cell Geometry to Ensure Efficient Bipolar Spindle Formation
Lancaster et al., Dev Cell 2013

1. Mechanical control of growth rates

B. Effect of mechanics on mitosis



Errors in mitosis!

Mitotic Rounding Alters Cell Geometry to Ensure Efficient Bipolar Spindle Formation
Lancaster et al., Dev Cell 2013

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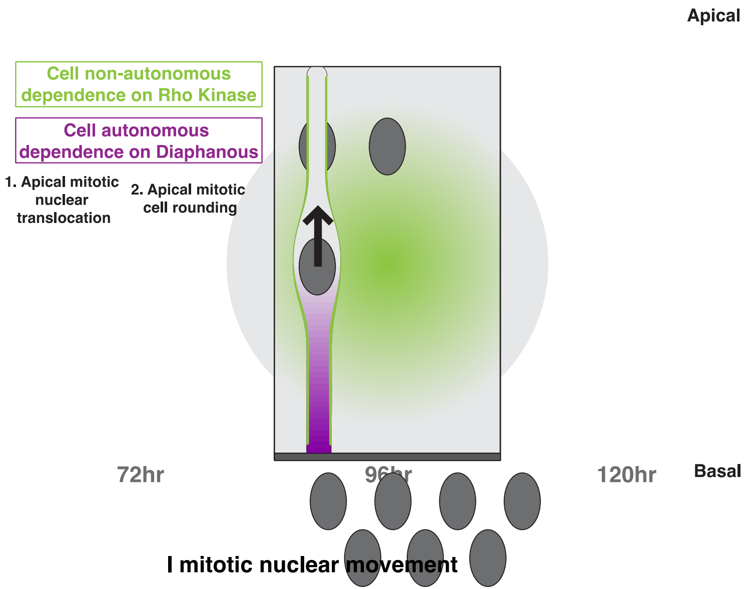
B. Effect of mechanics on mitosis

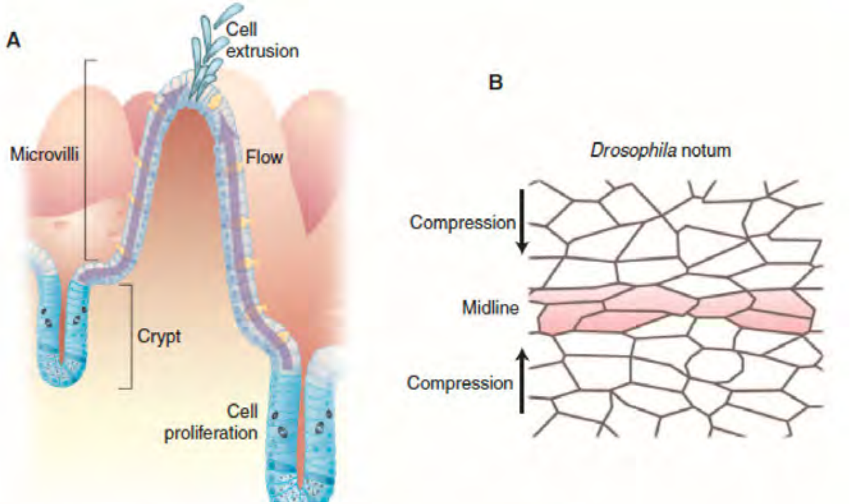


Tissue Mechanics Regulate Mitotic Nuclear Dynamics during Epithelial Development.
Kirkland et al., Current Biology 2020

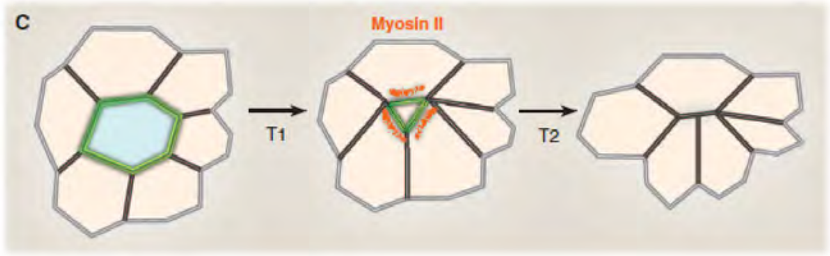
1. Mechanical control of growth rates

C. Effect of mechanics on **cell death** – non apoptotic extrusion

In the mammalian gut, cells divide in the crypt and move to the top of microvilli, where they die or extrude while alive.
Simons et al, Exp Cell Res 2011



In Drosophila, cell compression due to tissue growth changes cell shape (red) and causes live cell
Extrusion.
Marinari et al., Nature 2012



Extrusion occurs by cells destined to die signalling to surrounding epithelial cells to contract an actomyosin ring that squeezes the dying cell out.
Eisenhoffer et al., Nature 2012

Mechanical control of cellular mechanisms

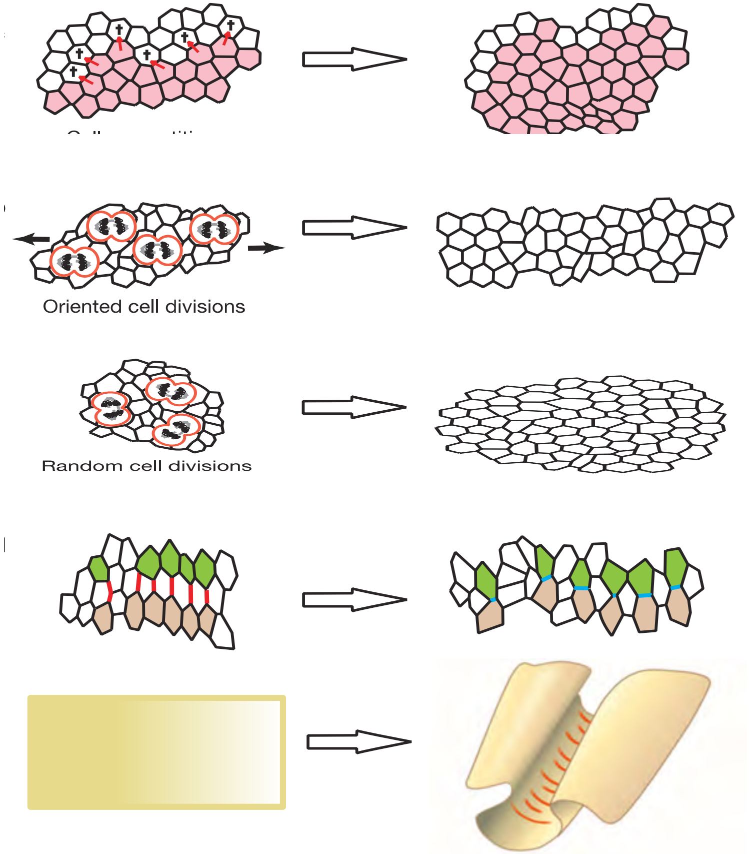
1. Growth rates

2. Division orientations

3. Cell shape changes

4. Planar cell rearrangements

5. 3D tissue sculpting



2. Mechanical control of division orientations

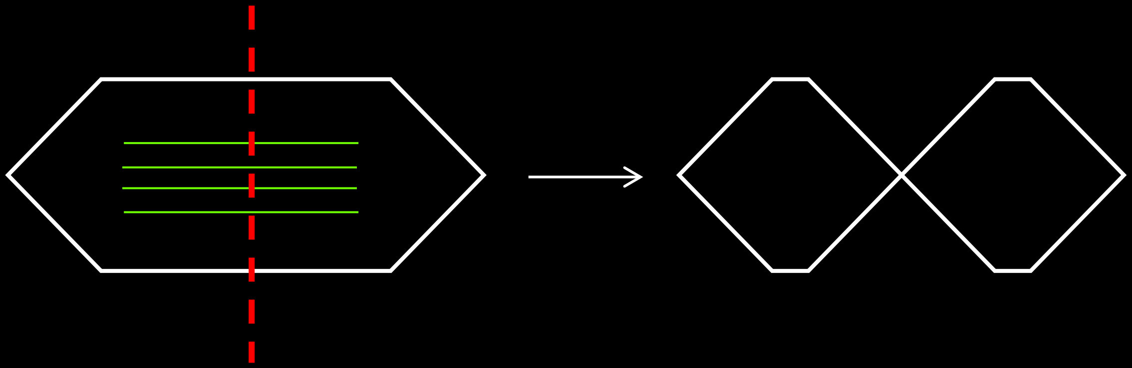
A. Cell shape and cell division orientation

1863: Hofmeister:
Cell geometry influences mitotic cleavage plans

1893: Hertwig's Rule:
Spindles align to longest axis of a cell

2. Mechanical control of division orientations

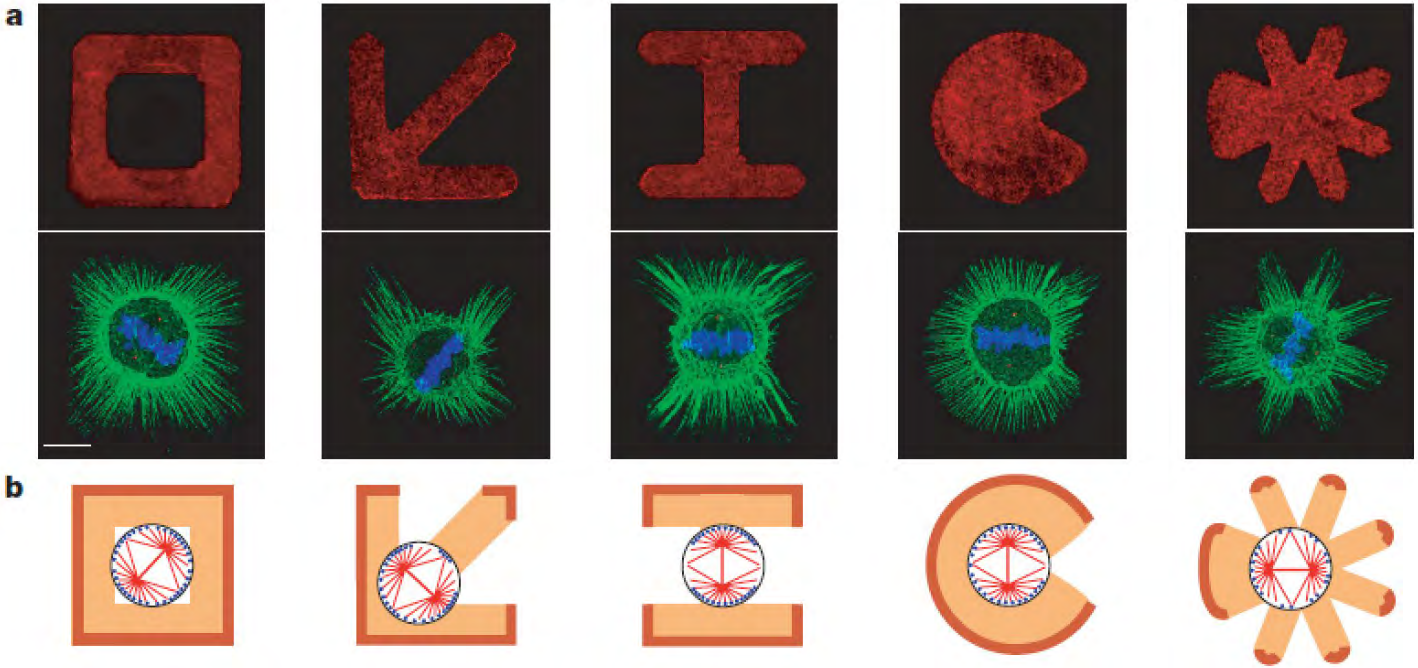
A. Cell shape and cell division orientation



Cell geometry dictates cell division orientation

2. Mechanical control of division orientations

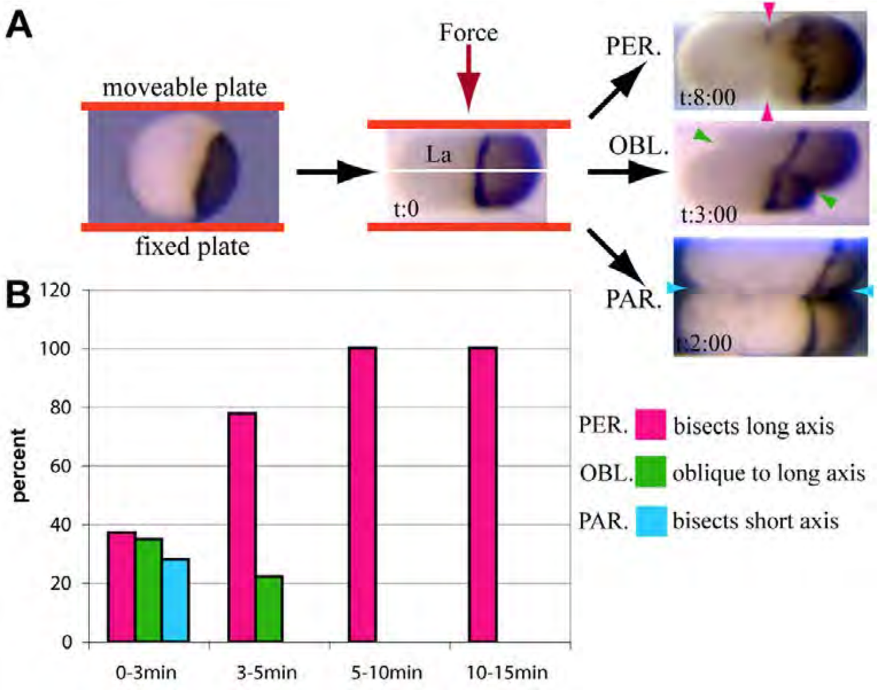
A. Cell shape and cell division orientation



Experimental and theoretical study of mitotic spindle orientation.
Thery et al., Nature 2007

2. Mechanical control of division orientations

A. Cell shape and cell division orientation



A default mechanism of spindle orientation based on cell shape is sufficient to generate cell fate diversity in polarised *Xenopus* blastomeres, Bernhard Strauss et al., Development 2006

Mechanical control of cellular mechanisms

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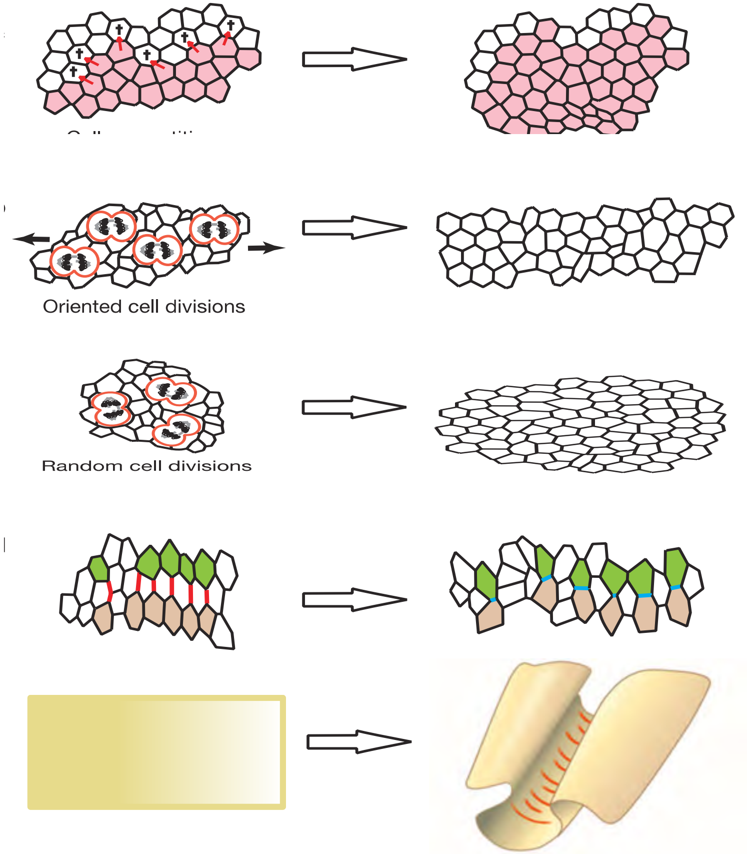


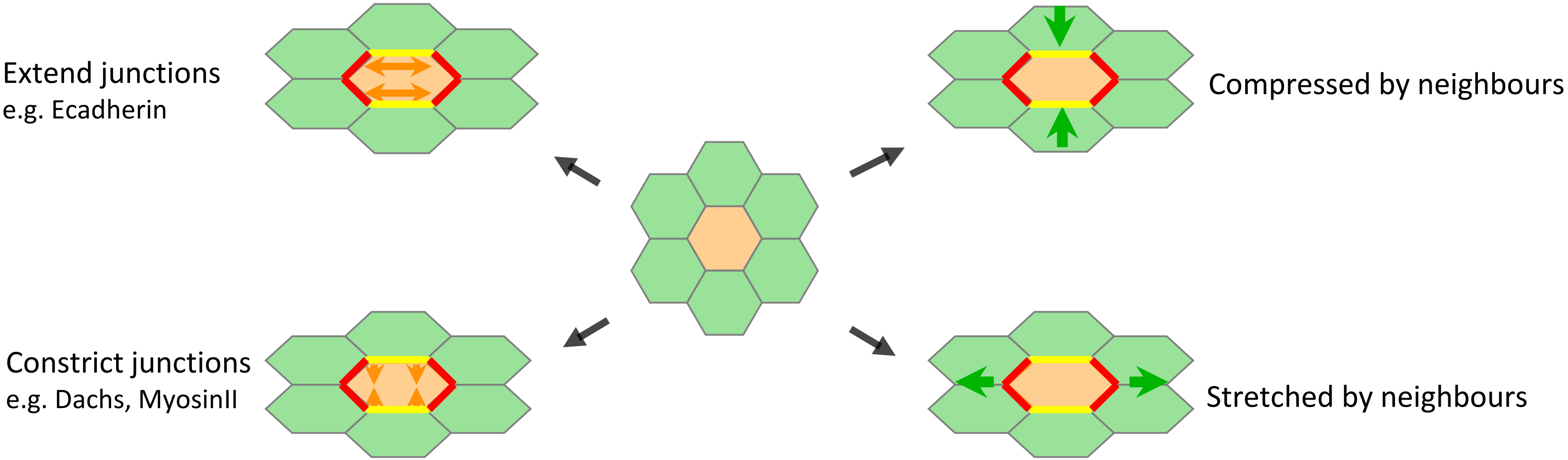
3. Mechanical control of cell shape

A. Different forces affect cell shape in a multicellular tissue

Planar view

Local
Cell autonomous

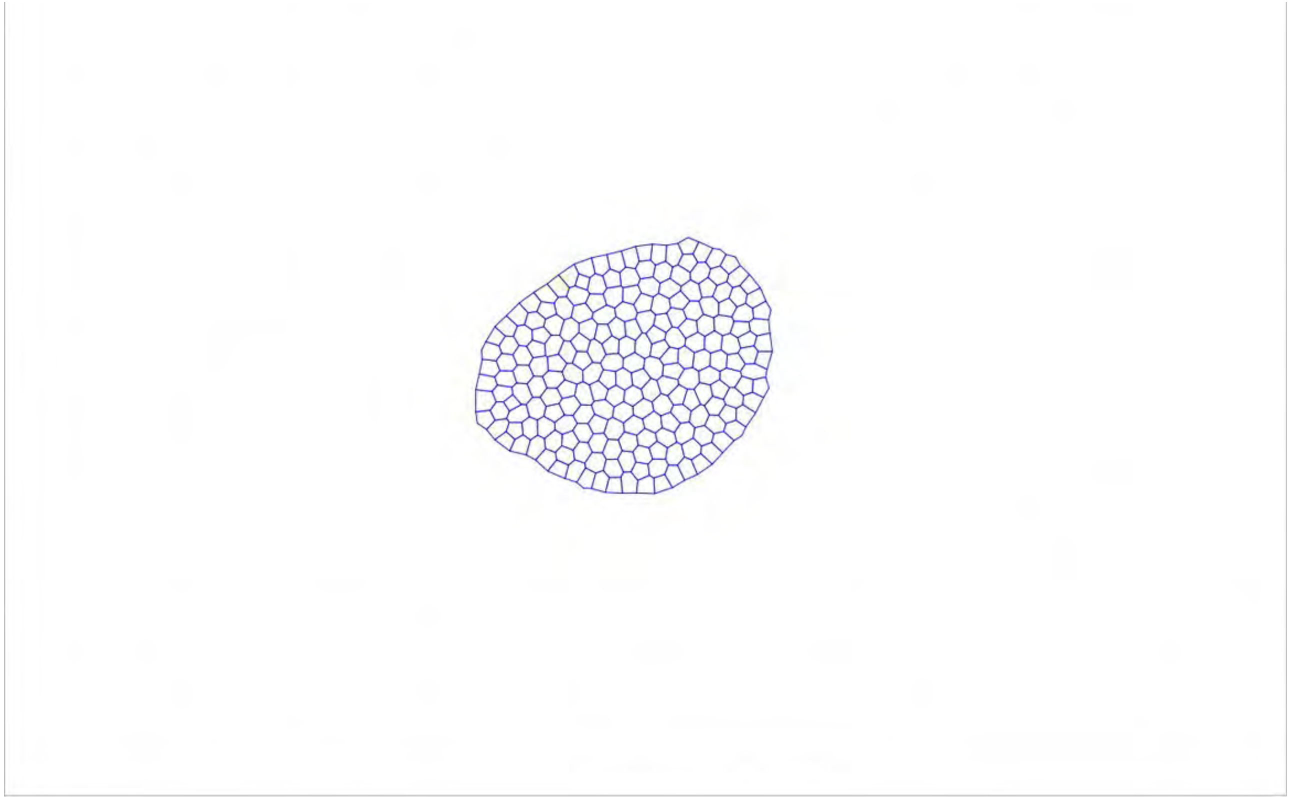
Global
Non cell autonomous



Mao et al., EMBO J, 2013

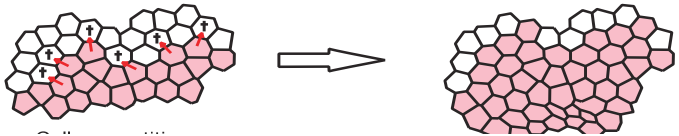
3. Mechanical control of cell shape

B. How forces are generated – differential tissue growth generates differential stress



Mechanical control of cellular mechanisms

1. Growth rates



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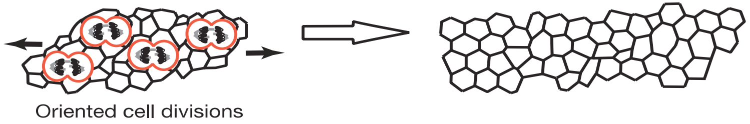

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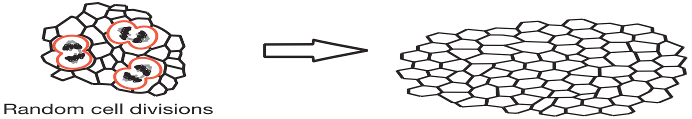

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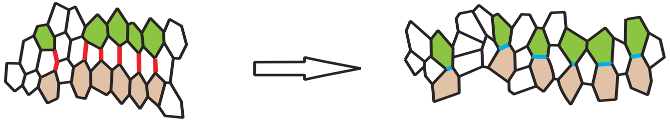
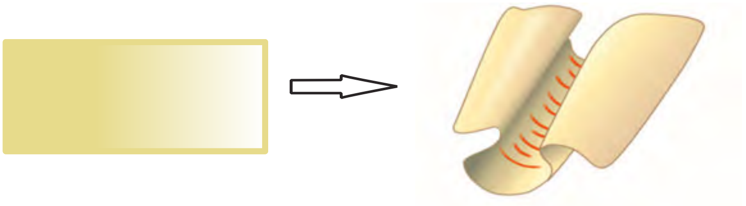
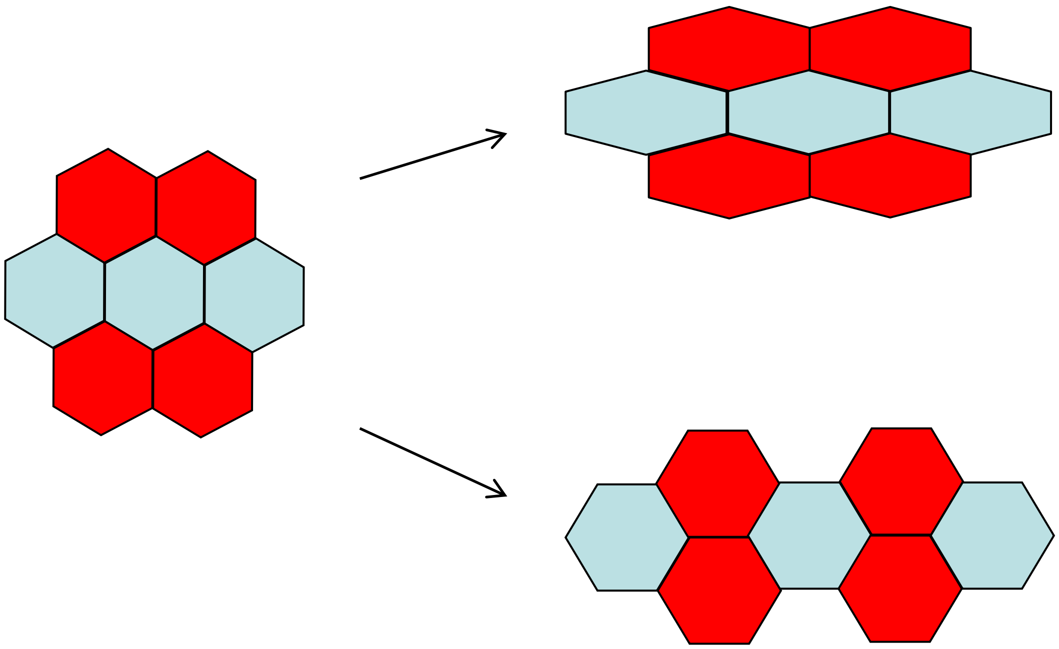


5. 3D tissue sculpting



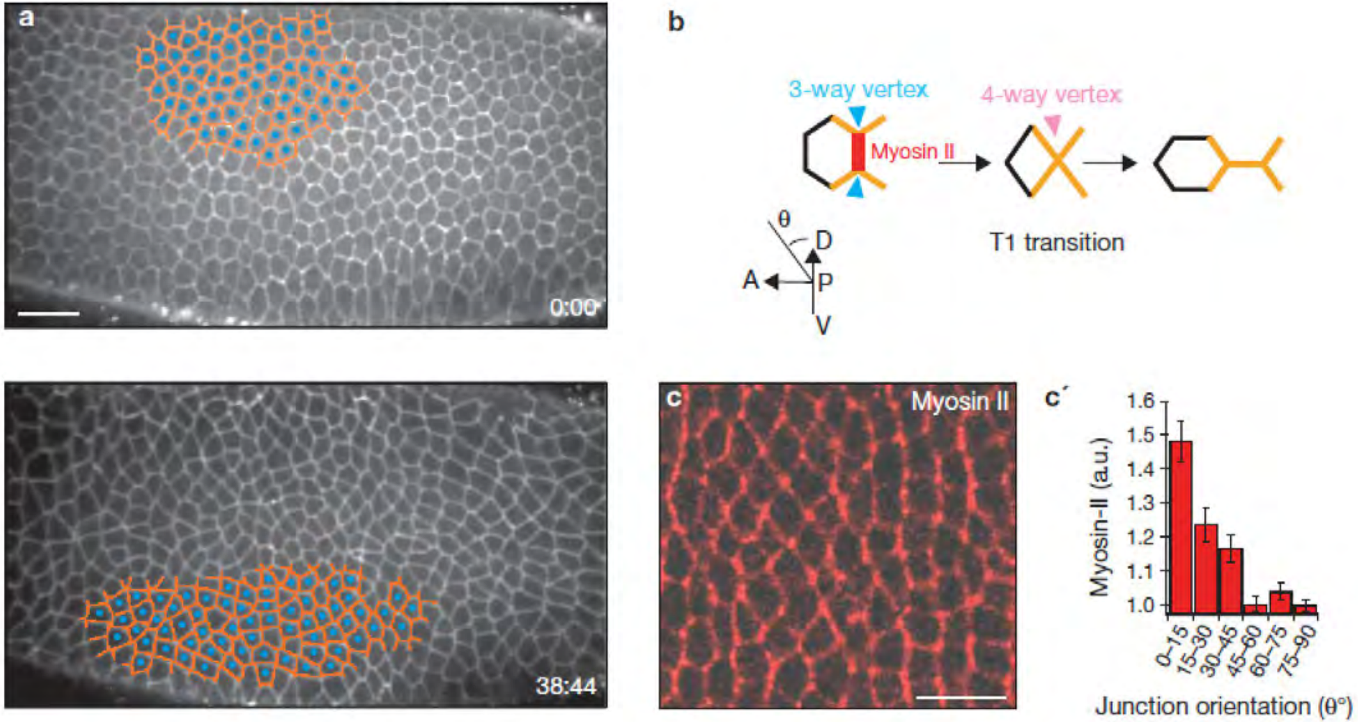
4. Mechanical control of cell rearrangements

A. Convergent / Extension - intercalation



4. Mechanical control of cell rearrangements

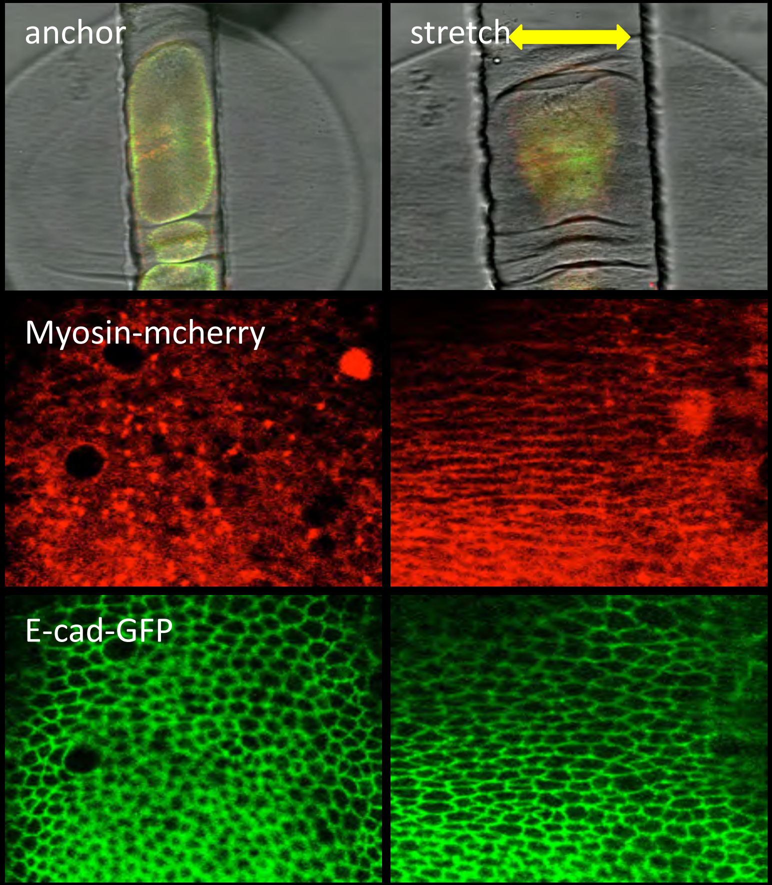
A. Convergent / Extension – intercalation via Myosin II polarisation



Lecuit and Zallen labs

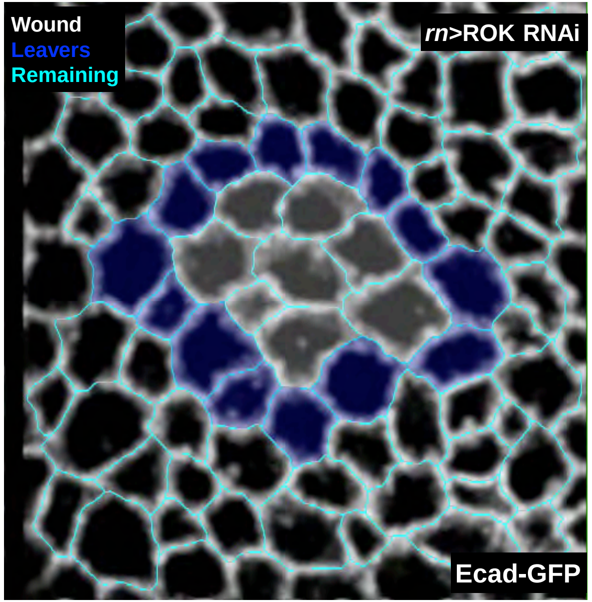
4. Mechanical control of cell rearrangements

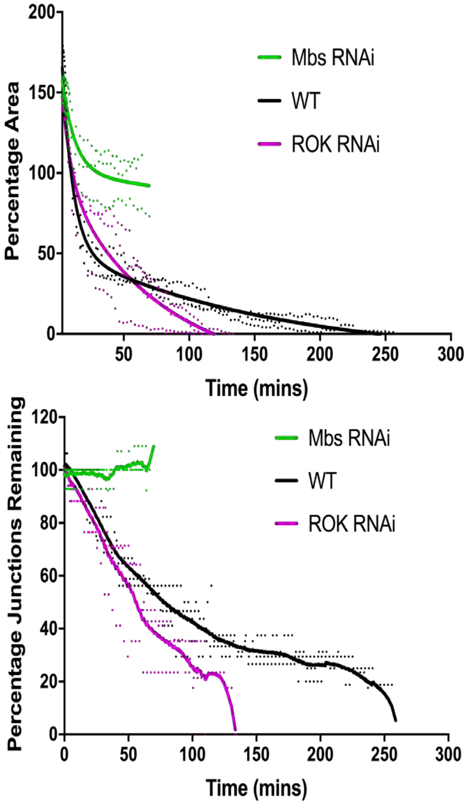
Myosin II polarisation is sensitive to mechanical forces



Duda et al, Dev Cell 2019

4. Mechanical control of cell rearrangements





Cell intercalation drive more efficient wound closure

Nature Physics: Tetley et al., 2019

Mechanical control of cellular mechanisms

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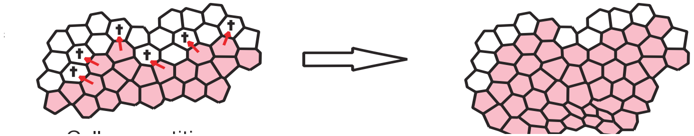

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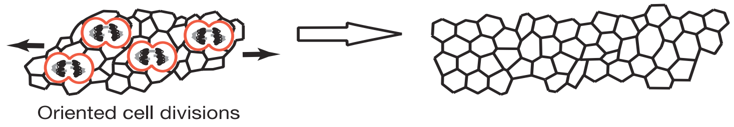

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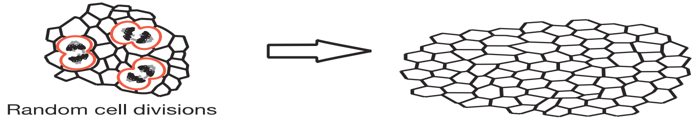

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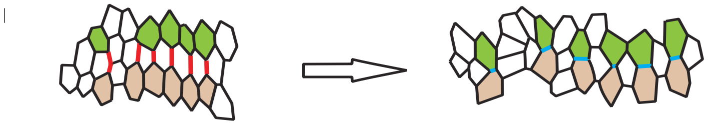
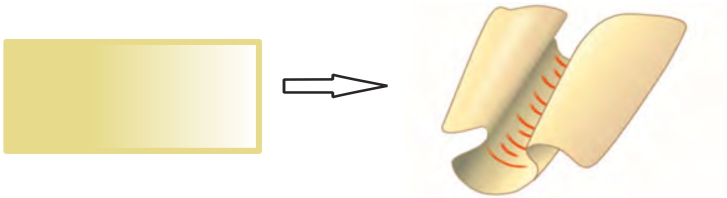
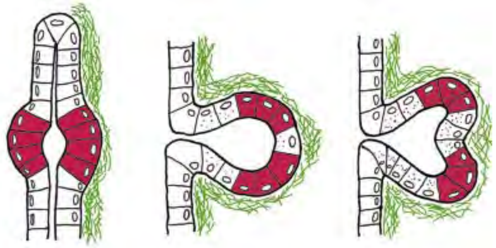


5. 3D tissue sculpting



5. Mechanical 3D sculpting of tissues

Organogenesis



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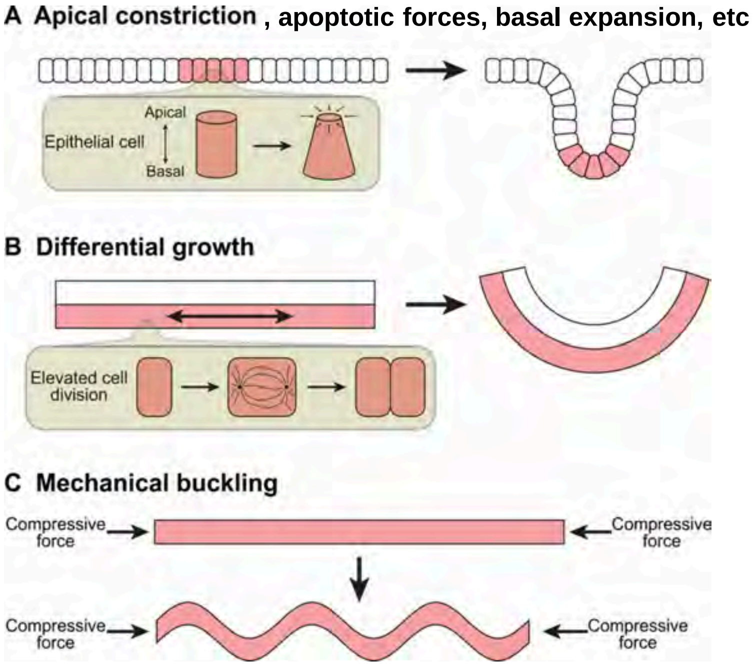
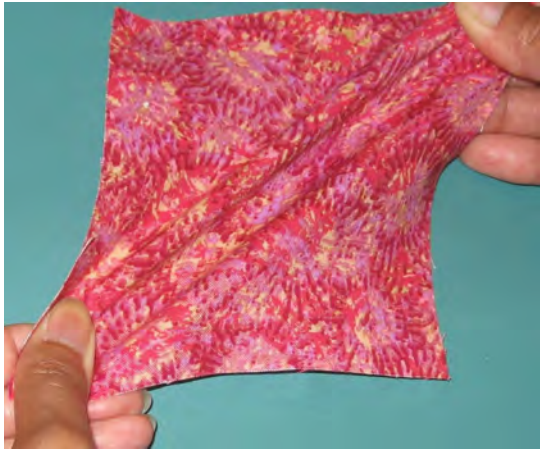


Works from labs of
Wieschaus,
Gartner,
Martin,
Leptin,
Suzanne,
Rauzi,
Dahmann,
Mahadevan, etc

Adapted from
Varner and Nelson 2014

5. Mechanical 3D sculpting of tissues

Differential planar growth rates induce precise patterns of folds

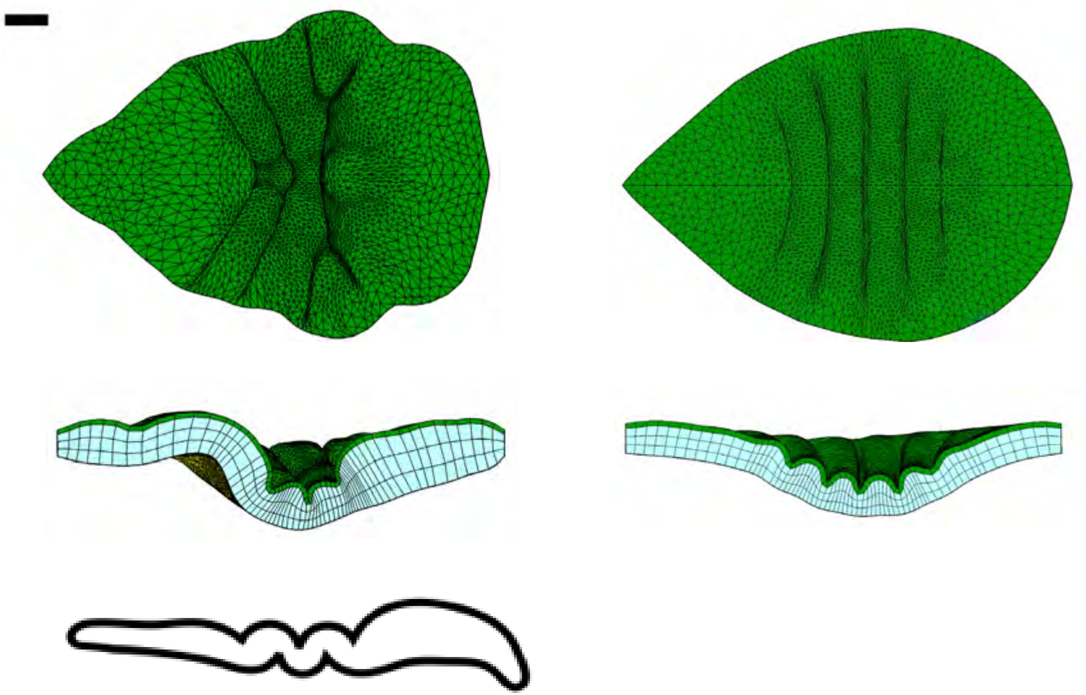


Finite Element Model of Tissue Folding

Tozluoglu et al., 2019

Differential planar growth rates induce patterns of folds

Experimental differential growth | Uniform growth



Tozluoglu et al., 2019

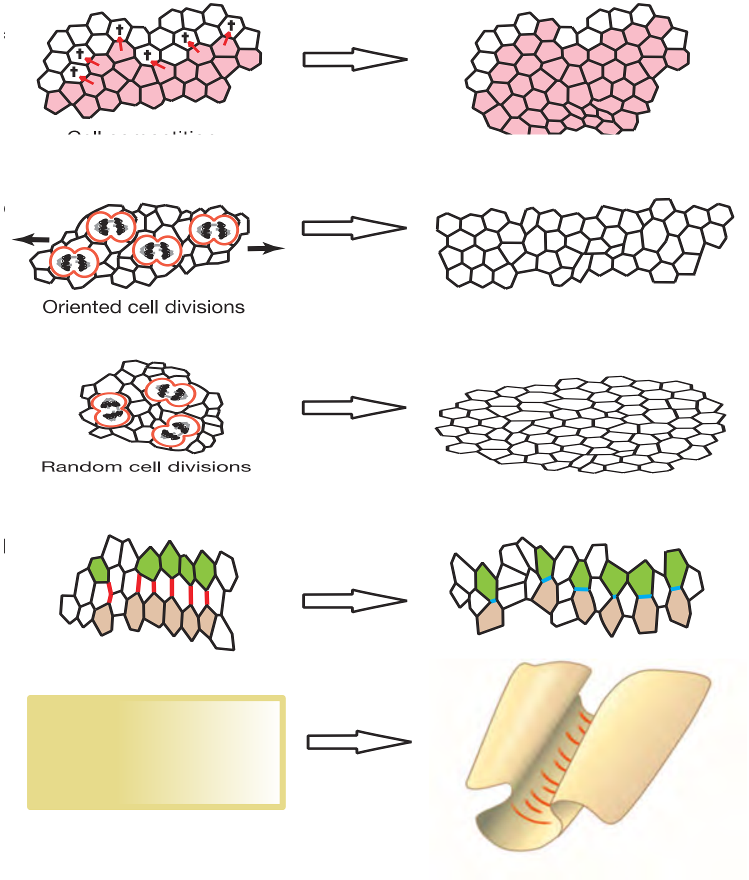
Cellular mechanisms of tissue morphogenesis

Interdependent processes through complex feedback mechanisms

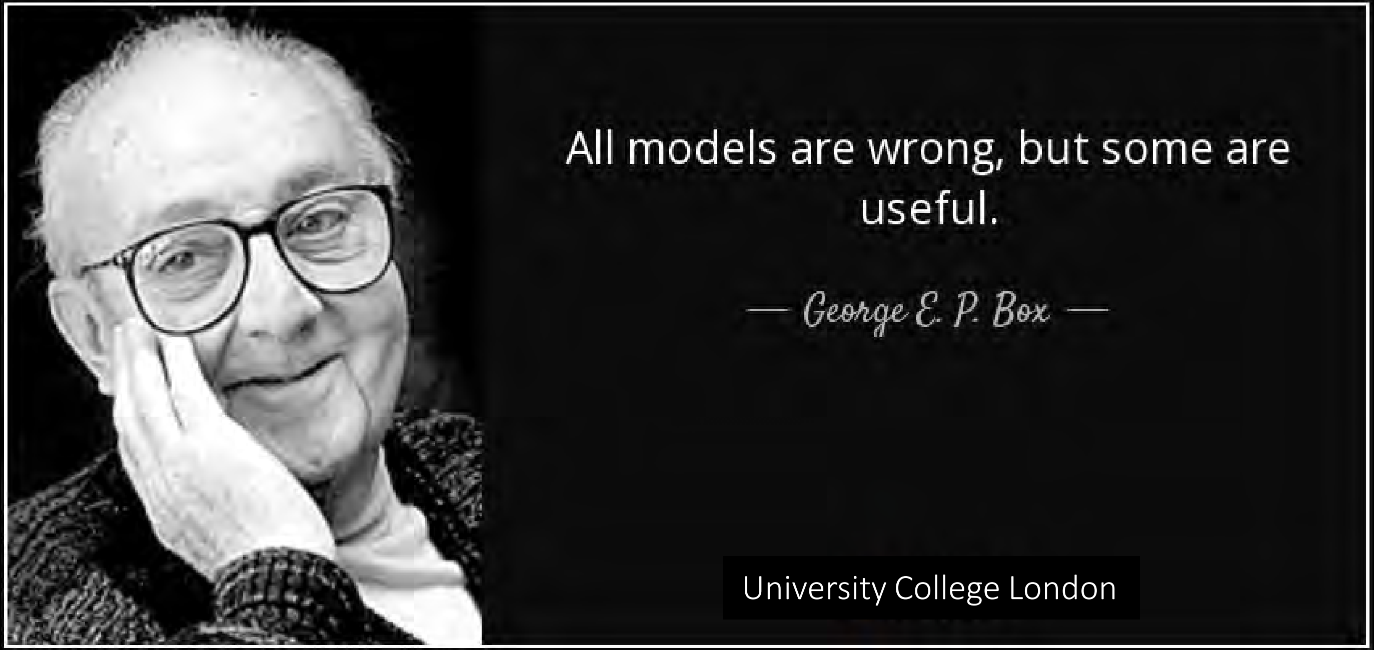
Biochemical

↓ ↑

Mechanical



Using mathematical models in biology



Using mathematical / physical models in biology
- working with biologists

What is your question? - Biology is often too complicated! (or biologists like to make it so, too many genes, too many variables…

Using mathematical / physical models in biology
- working with biologists

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Can you simplify the question? – can you frame it more precisely?

Using mathematical / physical models in biology - working with biologists

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What is your hypothesis? – again, force them to be as precise as possible

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What are the experiments / data to support hypothesis?

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- working with biologists

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Can you (I) create a simple mathematical model to test this hypothesis?

Using mathematical / physical models in biology - working with biologists

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What is your hypothesis? – again, force them to be as precise as possible

What are the experiments / data to support hypothesis?

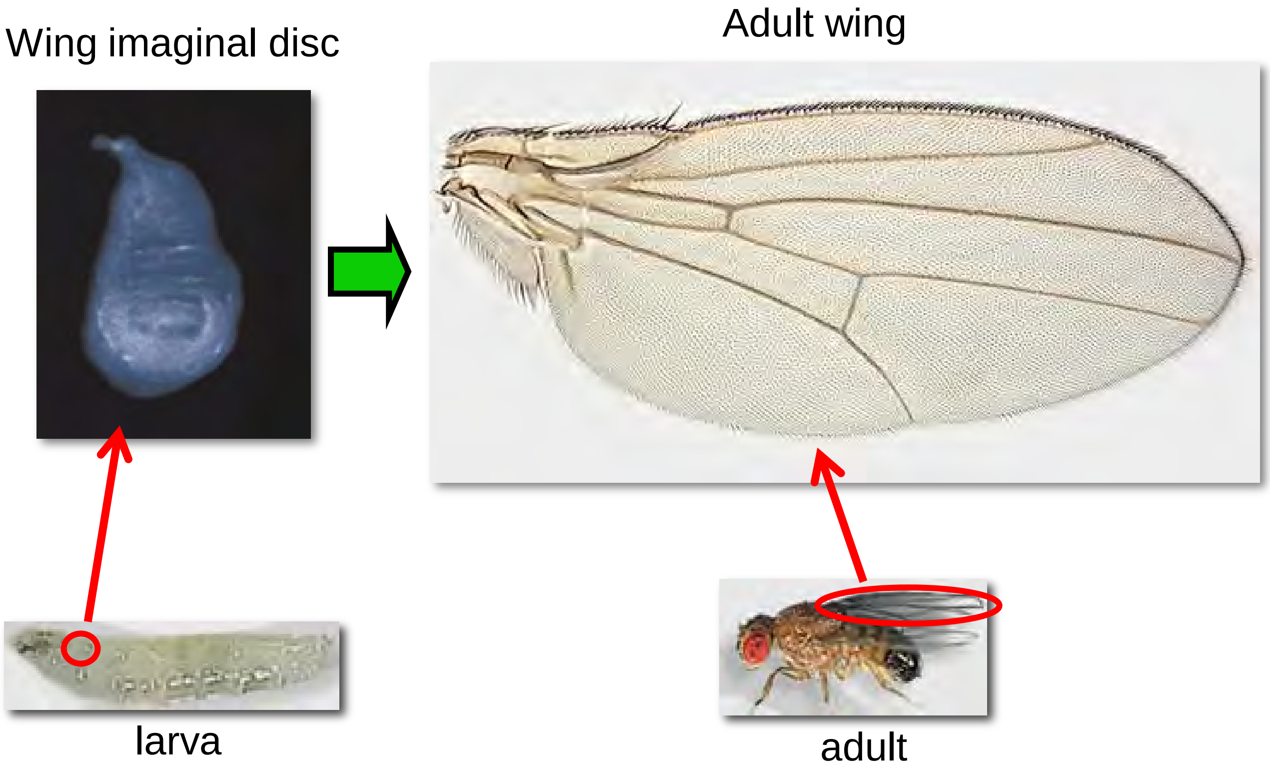
Can you (I) create a simple mathematical model to test this hypothesis?

What experiments can you (they) do to parameterise model and test model predictions?

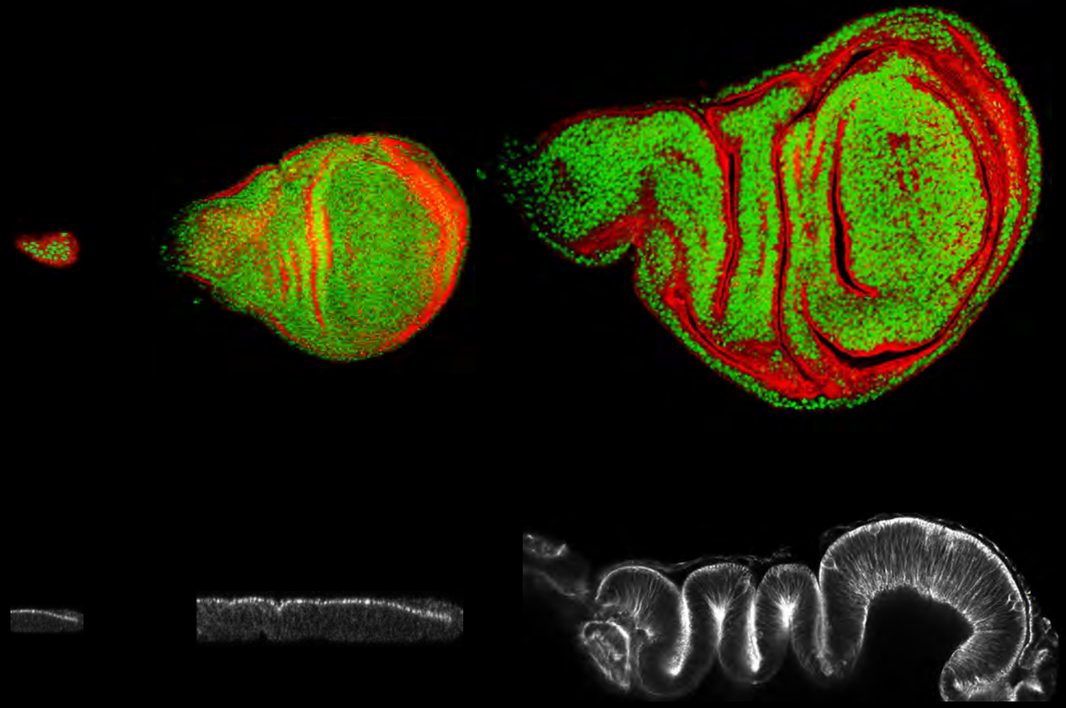
Case study 1:
Control of orientation of cell divisions in *Drosophila*

- Genetics
- Live imaging
- Computational 2D vertex model

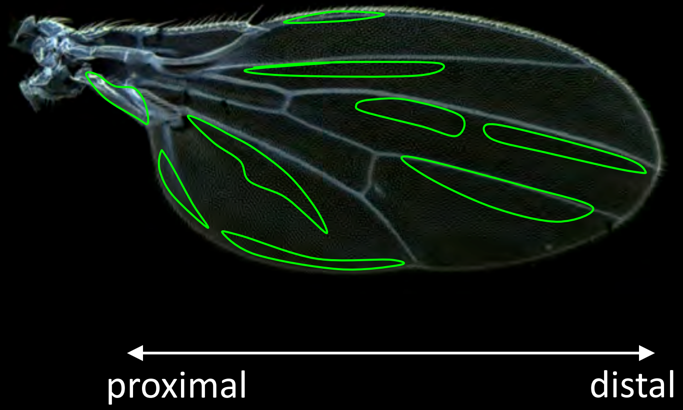
Drosophila imaginal discs: the organ precursors

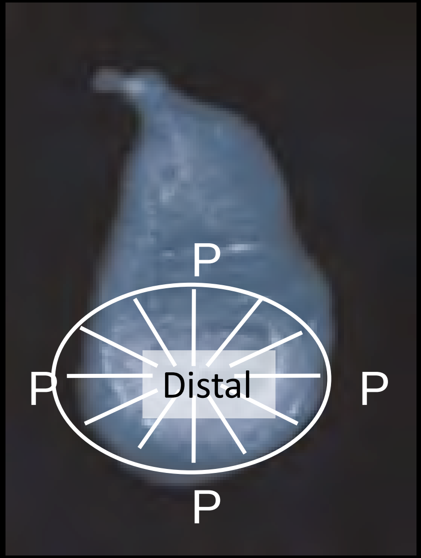


Drosophila imaginal discs: the organ precursors



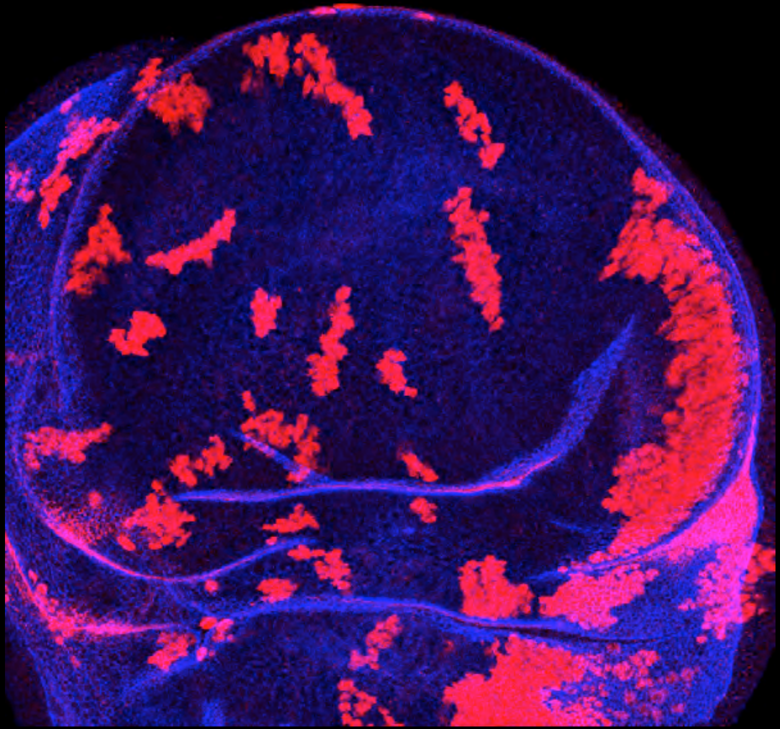
Drosophila wing growth in proximal - distal axis



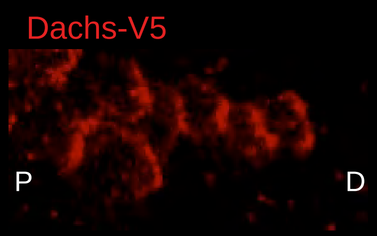


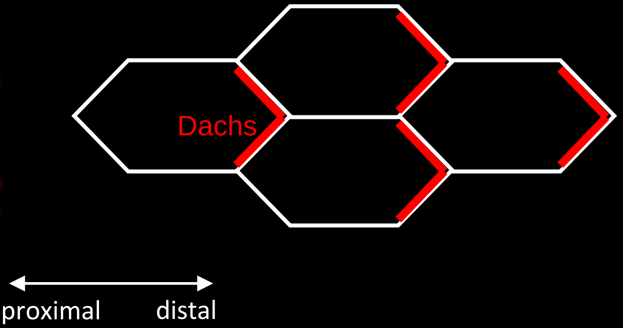
Baena-Lopez et al., 2005

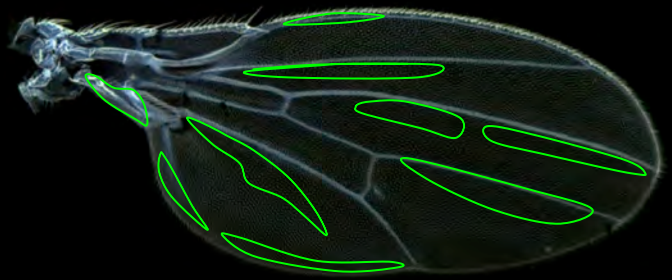
How are cell divisions oriented in the wing disc?



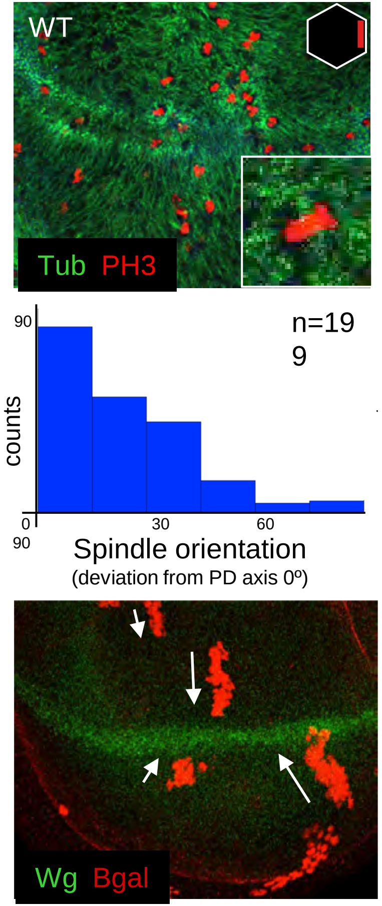
Planar polarisation of Dachs correlates with clone shape







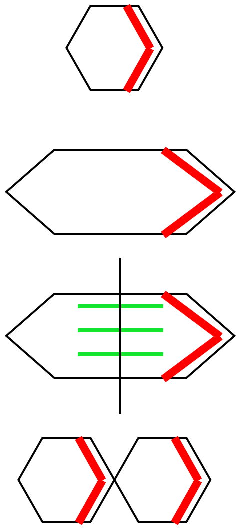
Polarisation of Dachs is required for spindle orientation and tissue growth



Mao et al., Genes & Dev 2011

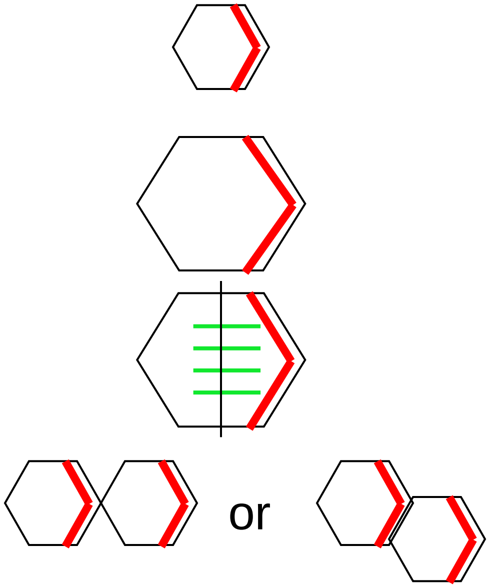
Models for how Dachs might orient cell division

Dachs (a myosin) affects cell shape, which orients spindle



Dachs orients spindle directly (somehow)

???



Thery et al., 2007

Using mathematical models in biology

What is your question?

Can you simplify the question?

What is your hypothesis?

Are there experiments / data to support hypothesis?

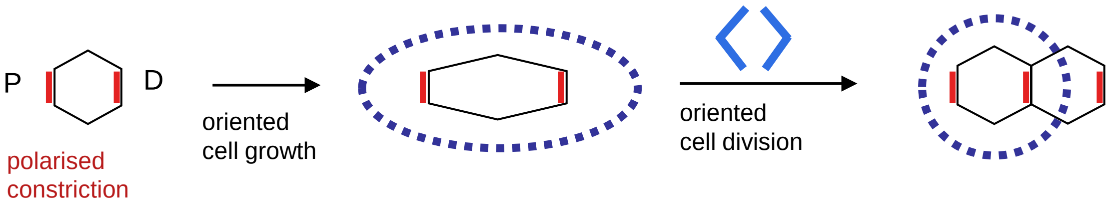
Can you create a simple mathematical model to test this hypothesis?

What experiments can you do to parameterise model and test model predictions?

Do cells show elongated growth and then divide in the same axis

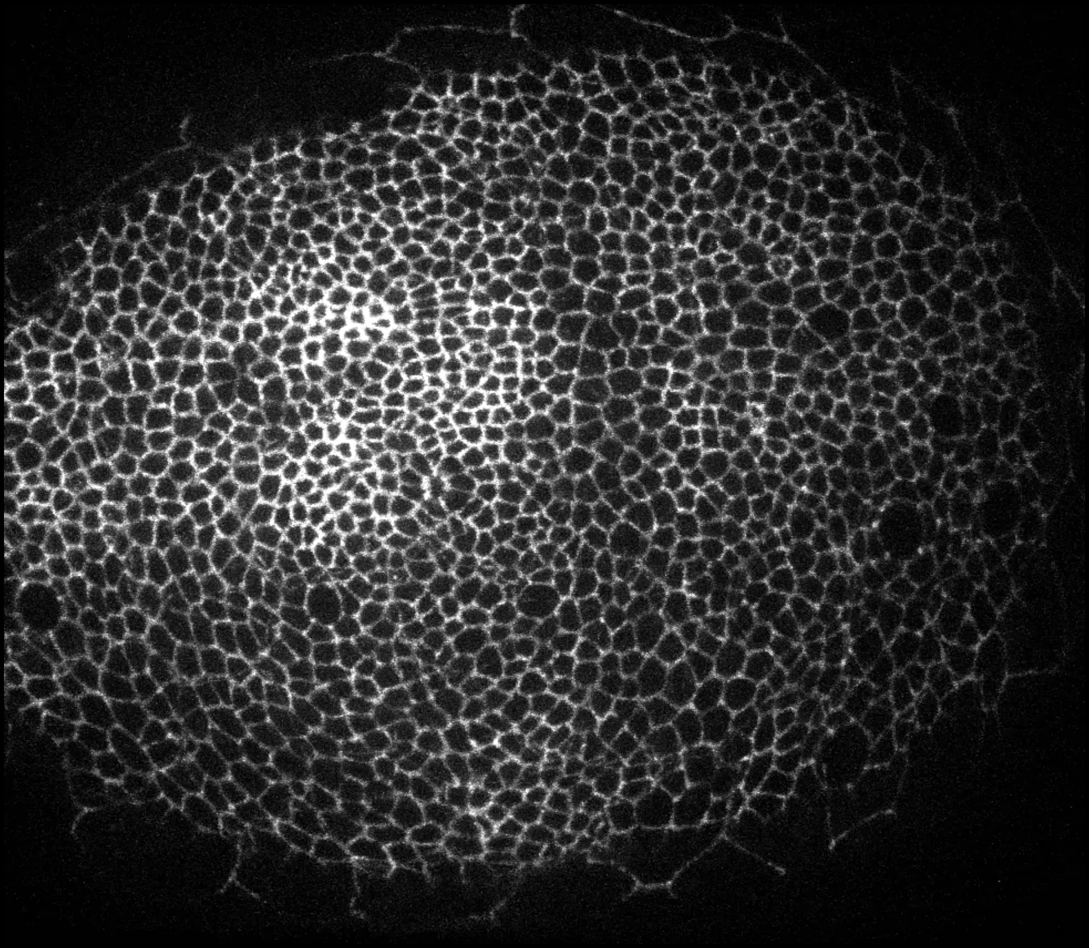
and

Does this correlate with the P-D axis?



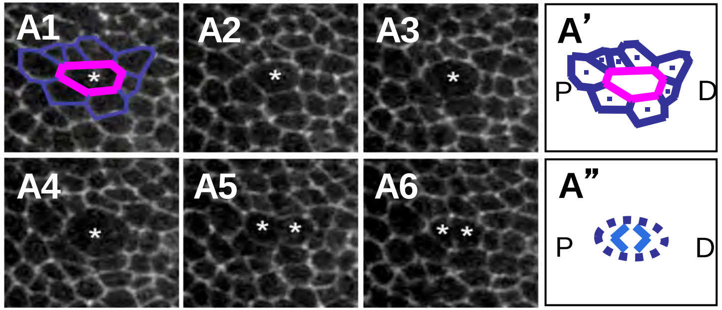
Proximal ⟷ Distal ????

Examine apical cell shape and orientation of cell divisions live



ArmGFP

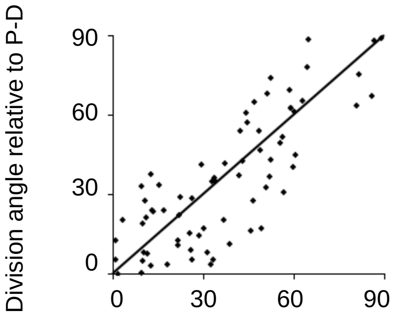
Cells do elongate, and divide perpendicular to long axis…



Dividing cell elongation ratio

=1.45 +/- 0.31

n=97

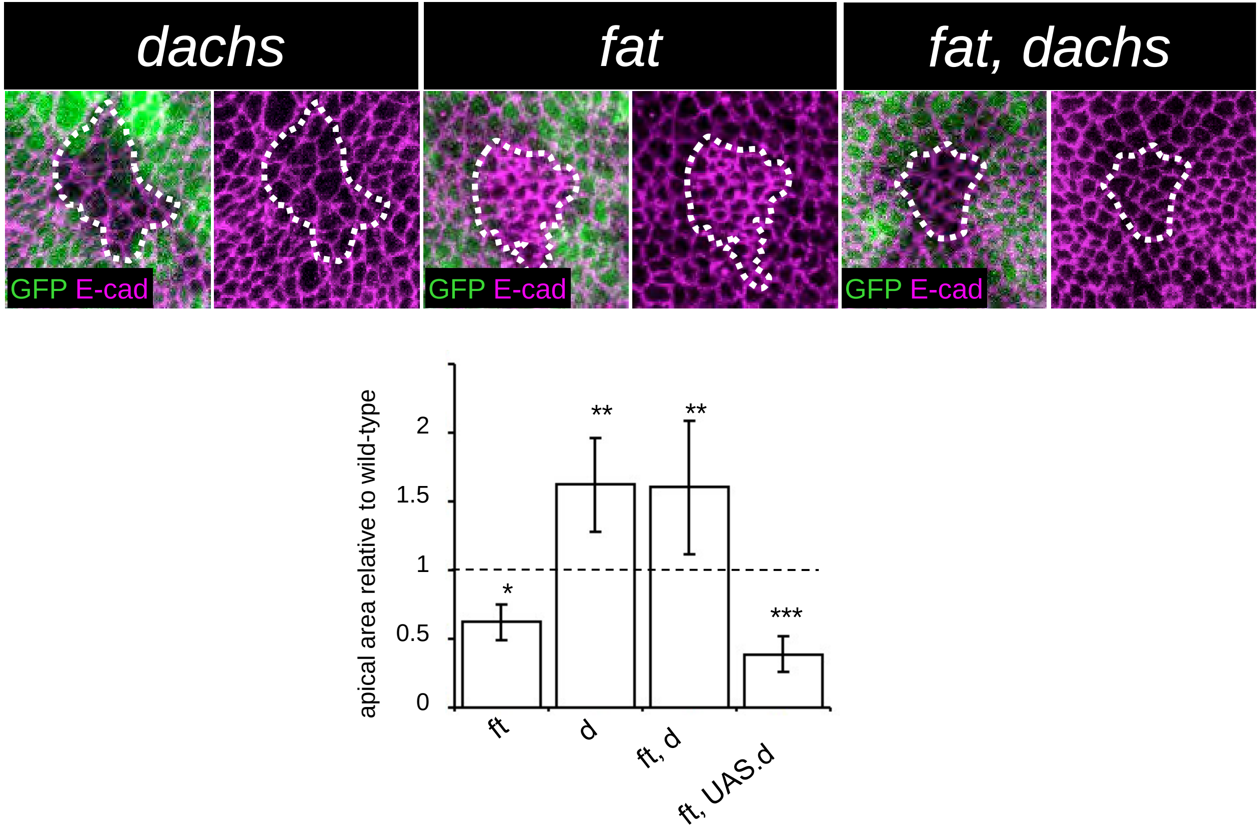


… even when it is not the P-D axis

Dividing cell elongation angle relative to P-D

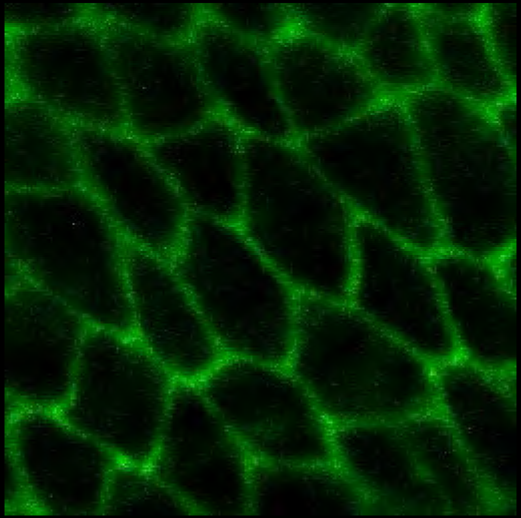
Geometry of apical cell-cell junctions determine orientation of cell division

Dachs can control apical cell geometry

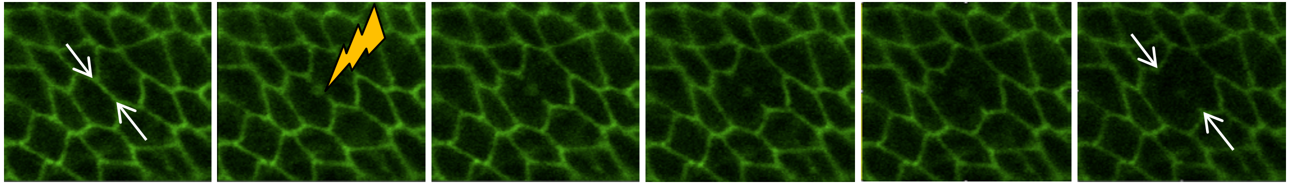


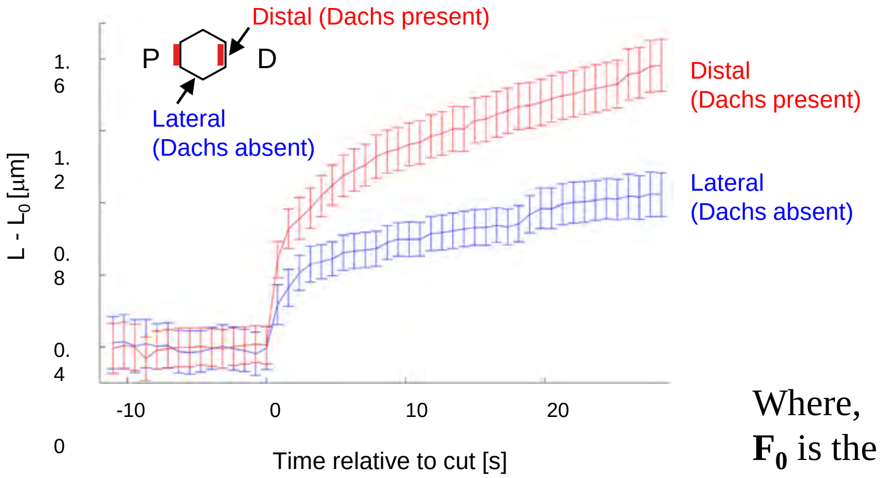
Mao et al., Genes & Dev 2011

Measuring junctional tension with laser ablation



Increased tension correlates with Dachs localisation





Distal rate = 0.69 µm/s, n=55 cuts
Lateral rate = 0.34 µm/s, n=39 cuts

Assume Kelvin-Voigt model:

$$\varepsilon(t) = L(t) - L(0) = \frac{F_0}{E}\left(1 - e^{-[\left(\frac{E}{\mu}\right)*t]}\right)$$

Where,
$\mathbf{F_0}$ is the tensile force before laser ablation = initial recoil velocity x $\boldsymbol{\mu}$
E is the elasticity,
$\boldsymbol{\mu}$ is the viscosity coefficient.

Using mathematical models in biology

What is your question?

Can you simplify the question?

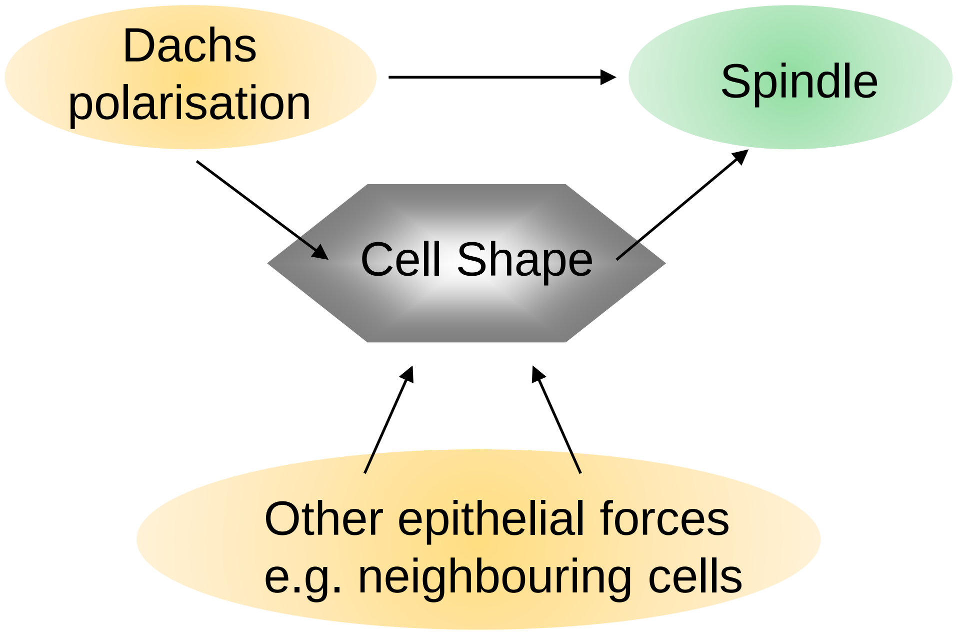
What is your hypothesis?

Are there experiments / data to support hypothesis?

Can you create a simple mathematical model to test this hypothesis?

What experiments can you do to parameterise model and test model predictions?

Supports Dachs influencing spindle orientation indirectly



How do forces contribute to epithelial packing??

Need a computational model of an epithelium

Using mathematical models in biology

What is your question?

Can you simplify the question?

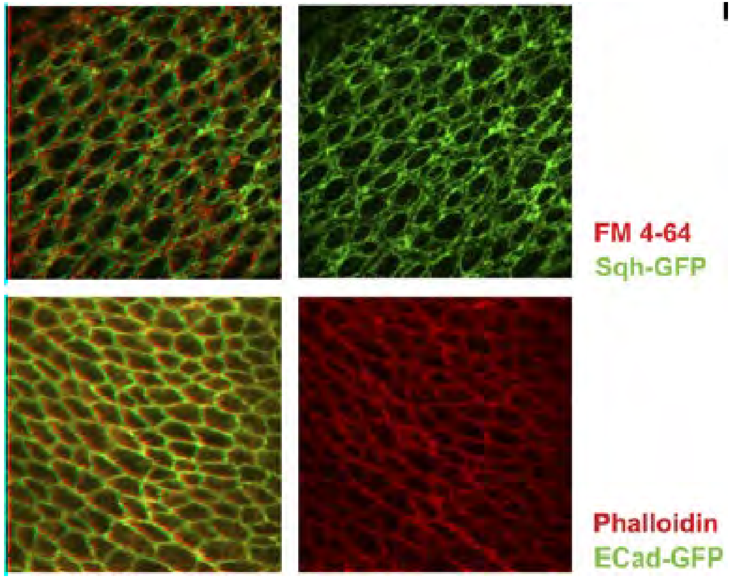
What is your hypothesis?

Are there experiments / data to support hypothesis?

Can you create a simple mathematical model to test this hypothesis?

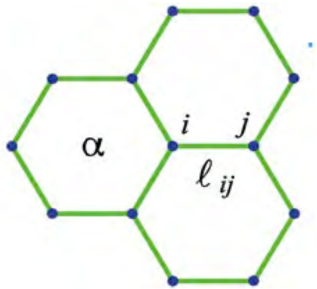
What experiments can you do to parameterise model and test model predictions?

Where do the forces come from?



Farhadifar et al., 2007

Apical:
E-cadherin
Actin
Myosin

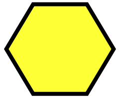


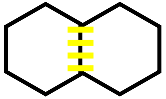
Adhesive and contractile molecules change force balances within the epithelium

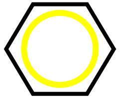
2D vertex model

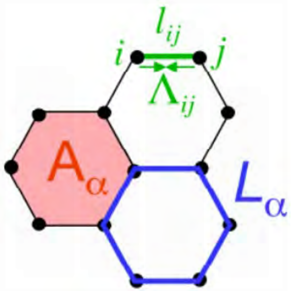
Quantitative description of forces at apical vertices

Potential Energy of Whole system = $\sum$ area elasticity + $\sum$ junctional tension + $\sum$ cortical contractility









$$E(\boldsymbol{R}_i) = \sum_{\alpha} \frac{K_\alpha}{2}\left(A_\alpha - A_\alpha^{(0)}\right)^2 + \sum_{\langle i,j \rangle} \Lambda_{ij}\ell_{ij} + \sum_{\alpha} \frac{\Gamma_\alpha}{2} L_\alpha^2$$

$$\boldsymbol{F}_i = -\frac{\partial E}{\partial \boldsymbol{R}_i}$$

$$\frac{\partial R_i}{\partial t} = \frac{F_i}{\zeta} + \mathrm{M}$$

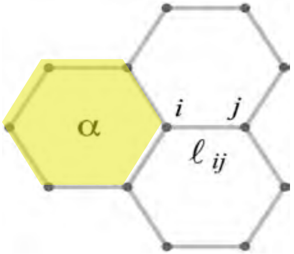
Nagai, Honda 2001, Farhadifar et al., 2007, etc

Area elasticity term

Pressure from volume and height of cell

$$E(\boldsymbol{R}_i) = \sum_{\alpha} \frac{K_\alpha}{2}\left(A_\alpha - A_\alpha^{(0)}\right)^2 + \sum_{<i,j>} \Lambda_{ij}\ell_{ij} + \sum_{\alpha} \frac{\Gamma_\alpha}{2} L_\alpha^2$$

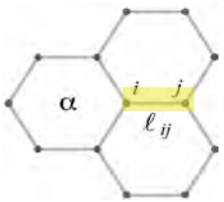
elastic coefficient — actual area — preferred area



Junctional line tension term

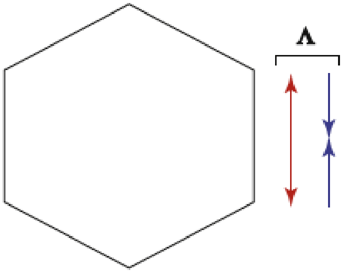
Force balance between adhesion molecules and contractile molecules

$$E(R_i) = \sum_{\alpha} \frac{K_\alpha}{2}\left(A_\alpha - A_\alpha^{(0)}\right)^2 + \sum_{<i,j>} \Lambda_{ij}\ell_{ij} + \sum_{\alpha} \frac{\Gamma_\alpha}{2} L_\alpha^2$$



Line tension coefficient
∝ contractility - adhesion

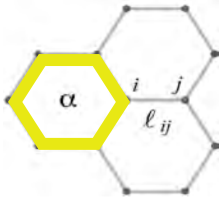
Length of junction



Perimeter contractility term

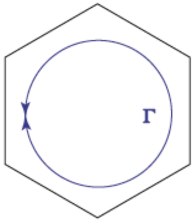
Contractile force of actin-myosin ring

$$E(\mathbf{R}_i) = \sum_{\alpha} \frac{K_\alpha}{2}\left(A_\alpha - A_\alpha^{(0)}\right)^2 + \sum_{<i,j>} \Lambda_{ij}\ell_{ij} + \sum_{\alpha} \frac{\Gamma_\alpha}{2} L_\alpha^2$$



Perimeter contractility coefficient

Length of cell perimeter



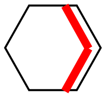
To include Dachs: How would you do it?

To include Dachs: modify line tension term

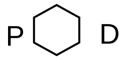
$$E(R_i) = \frac{K}{2}\sum_{\alpha}\left(A_\alpha - A_\alpha^{(0)}\right)^2 + \Lambda \sum_{<i,j>}(1 + \varphi_{ij}^{Dachs}) l_{ij} + \frac{\Gamma}{2}\sum_{\alpha} L_\alpha^2$$

φ_{ij}^{Dachs} = |sin (edge angle - PD angle)| φ_{MAX}^{Dachs}

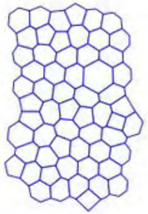
Dachs tension factor



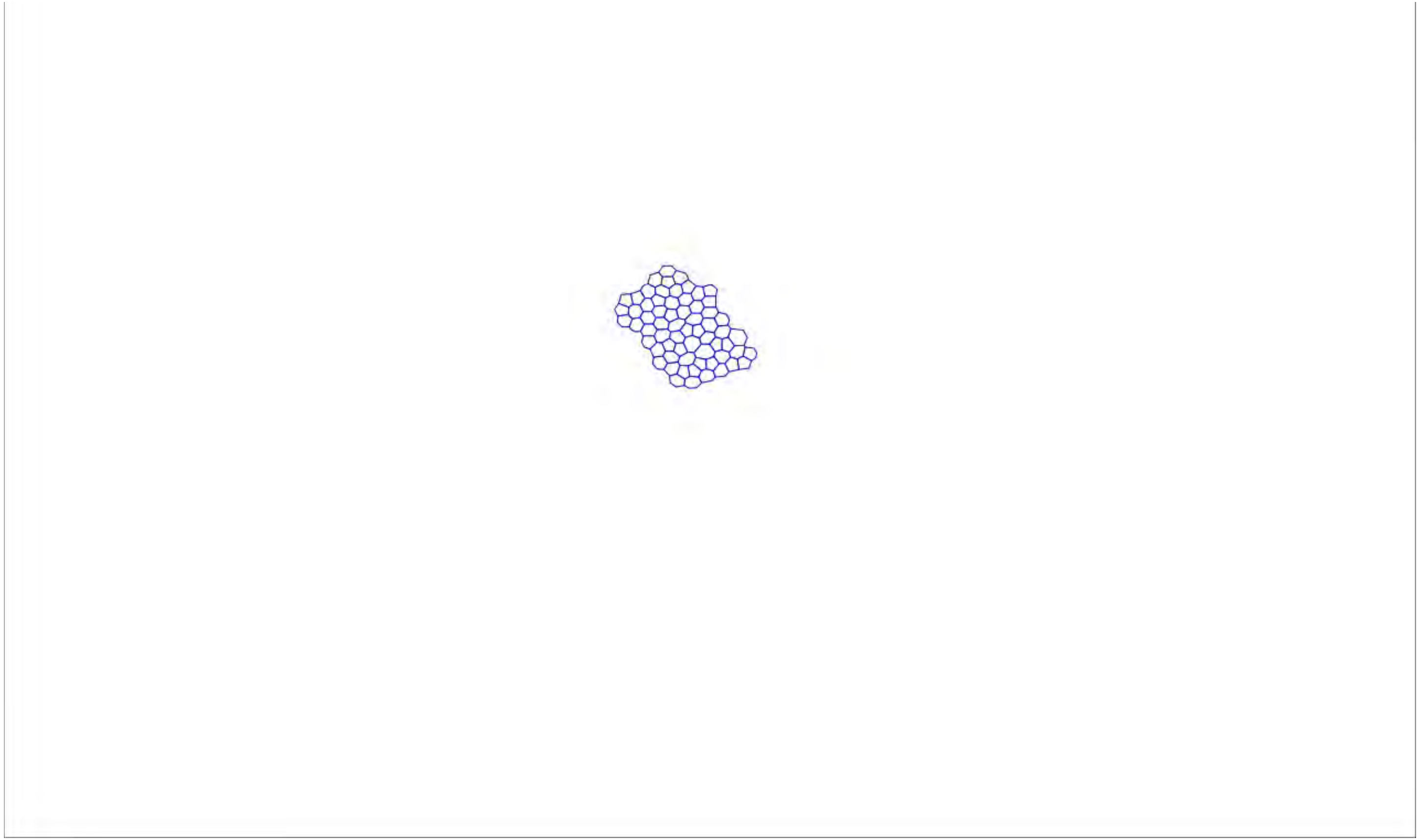
Isometric tension & division in long axis



48 hrs simulated growth = ~ 45 min run time

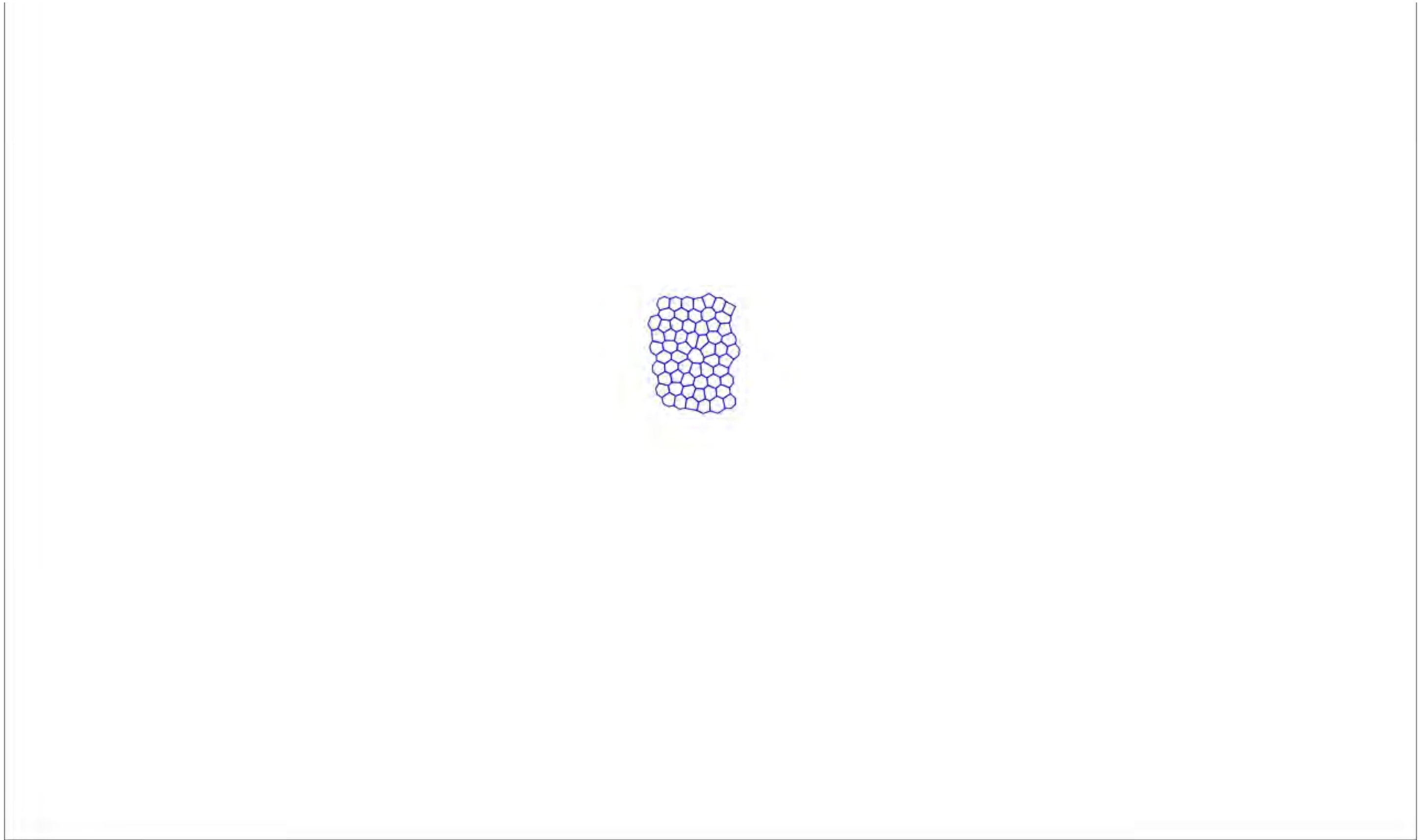


Polarized tension & division in long axis

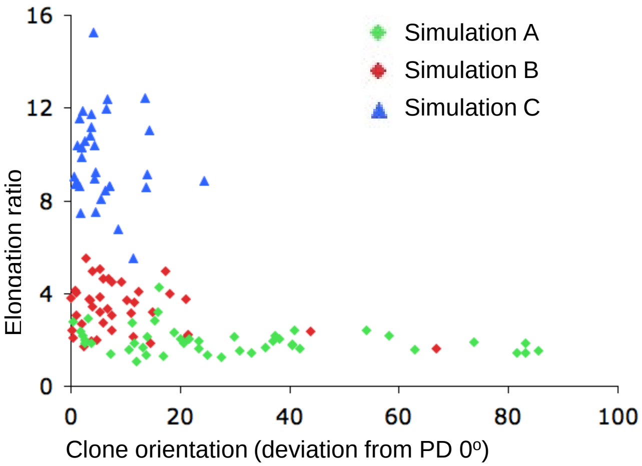


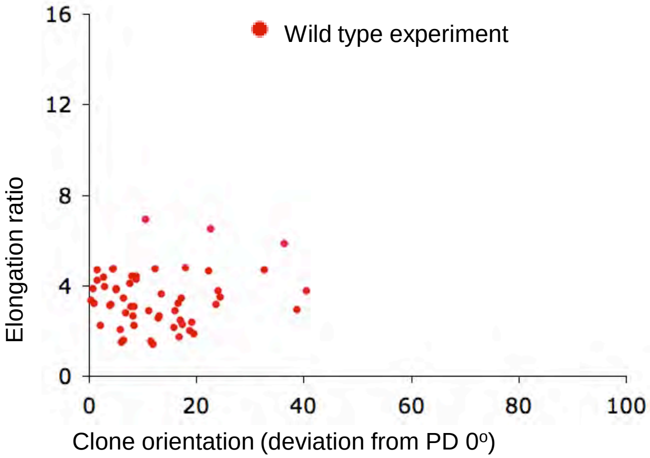
Isometric tension & polarized cell division

P D



Comparing simulations to *in vivo*





Maybe we parameterised (B) to get what we want? – partly true, we needed to parameterise Dachs-Max

Mao et al., Genes & Dev 2011

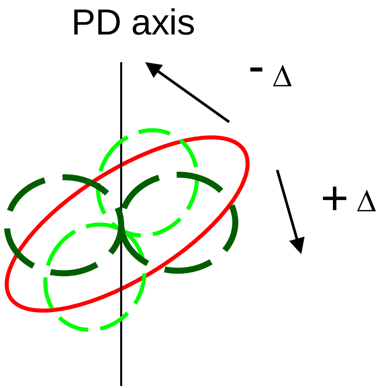
Cell shape vs spindle binding

Δ = angle between daughter cells and dividing cell

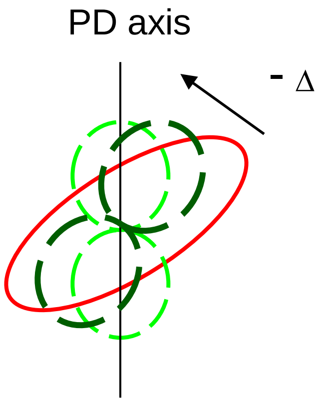
- Δ = towards P-D

+ Δ = away from P-D

cell shape



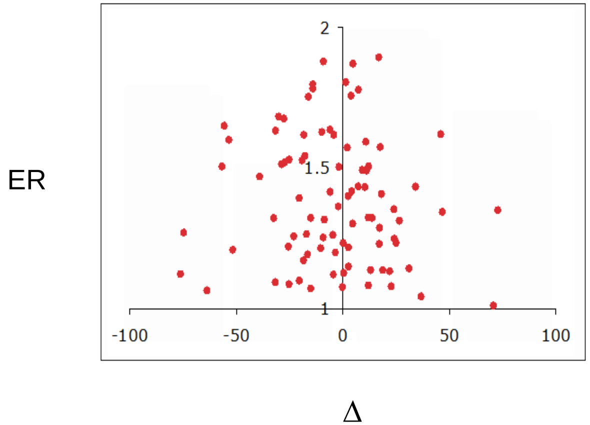
spindle



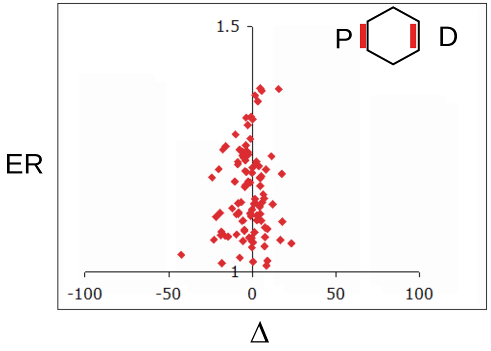
Mao et al., Genes & Dev 2011

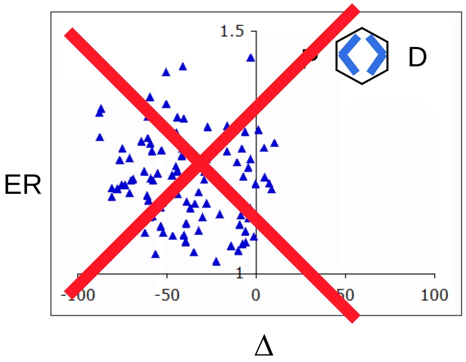
Δ Analysis

Experimentation



Simulations

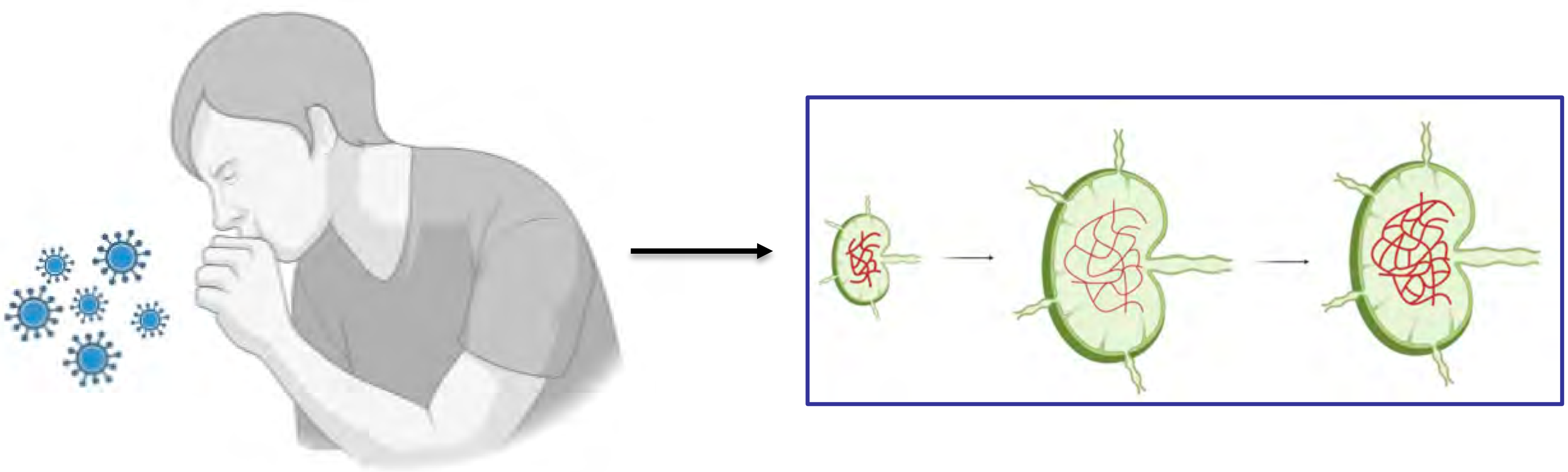




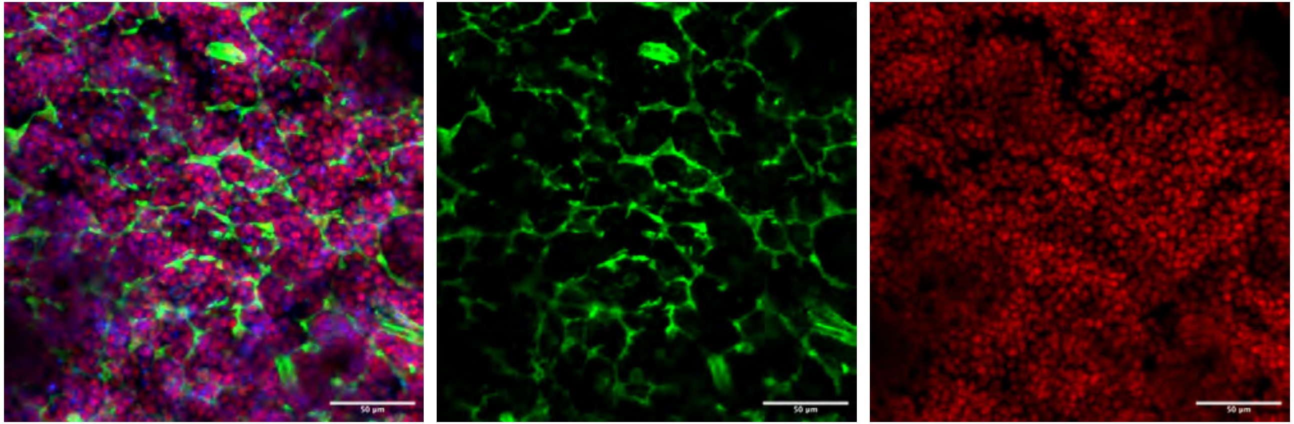
not via direct spindle binding

Exercise Questions

The Lymph Node: a dynamic homeostatic tissue

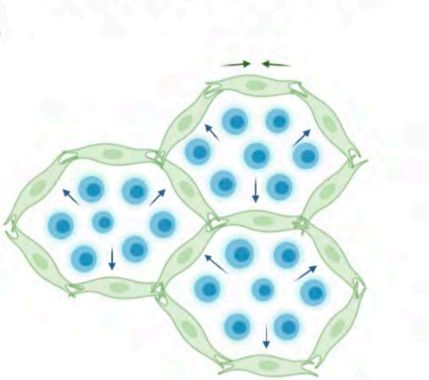


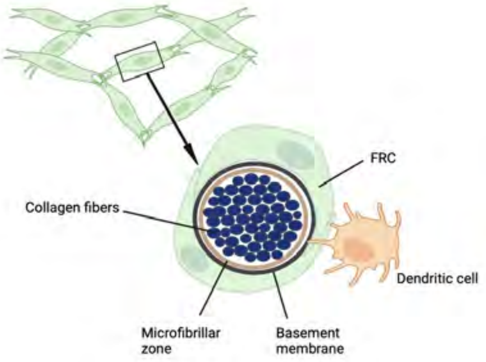
Geometry and topology of system:



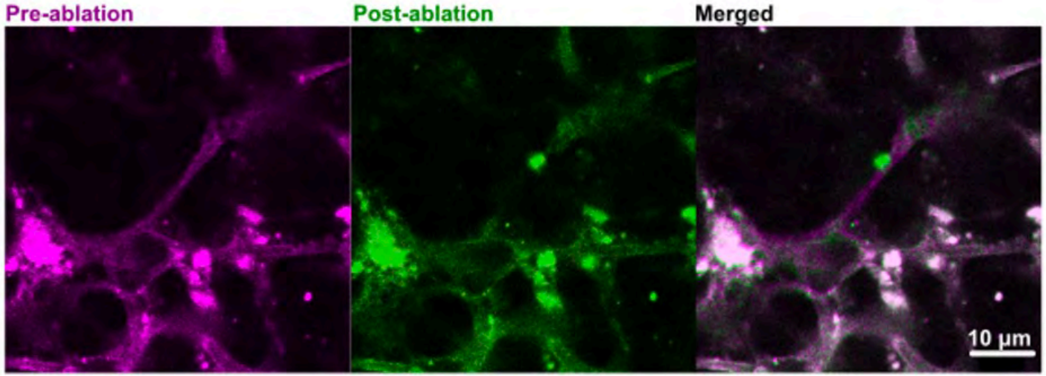
FRC Cells, T cells

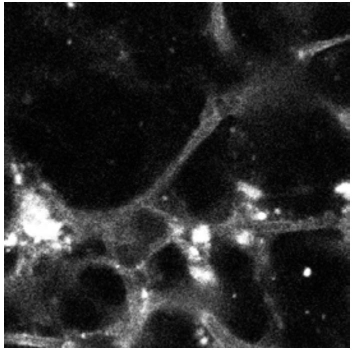
FRC network cartoon





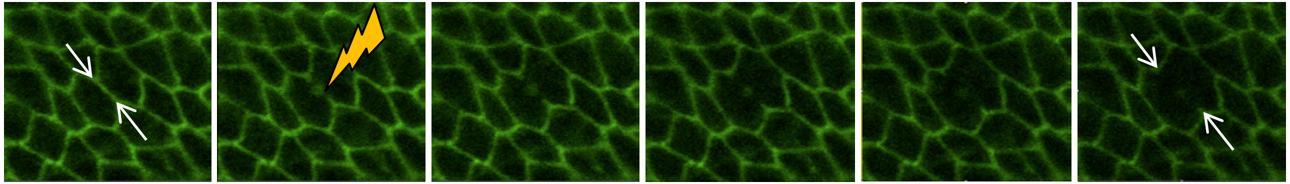
Mechanics of lymph node system:

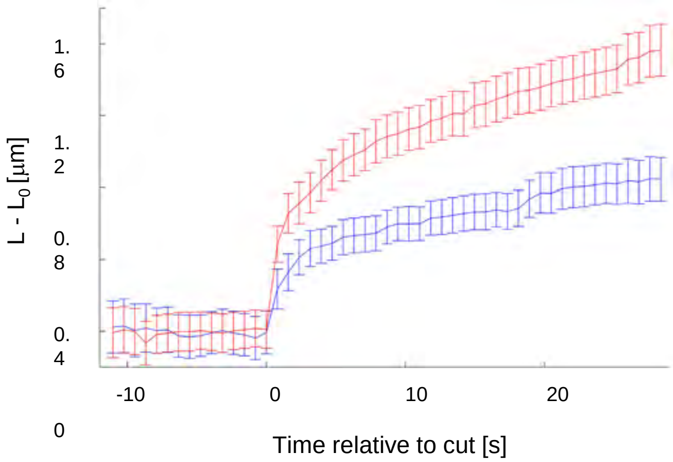




Questions:

1. Analyze the laser ablation data provided to infer the mechanical properties of the lymph node FRC network, with and without ECM (after collagenase treatment). Infer: initial tensile force, elasticity, viscosity.
2. Laser ablation measures relative changes in tensile force, how would you measure absolute forces?
3. What does the ECM do to the mechanical properties of the FRC network?
4. How would you model the lymph node FRC network?





Assume Kelvin-Voigt model:

$$\varepsilon(t) = L(t) - L(0) = \frac{F_0}{E}\left(1 - e^{-\left[\left(\frac{E}{\mu}\right)*t\right]}\right)$$

Fit data to calculate:

$\mathbf{F_0}$: the tensile force before laser ablation

E : the elasticity,

μ : the viscosity coefficient.

Further reading:

Mao et al., EMBO J 2013.
Differential proliferation rates generate patterns of mechanical tension that orient tissue growth

Farhadifar et al., Current Biology 2007
Influence of cell mechanics… on epithelial packing

Tetley et al., Nature Physics, 2019.
Tissue fluidity promotes epithelial wound healing

Liang et al., Bio Protoc. 2017,
Measurement of Mechanical Tension at Cell-cell Junctions Using Two-photon Laser Ablation

Mao and Wickstrom, Nature Reviews MCB 2024,
Mechanical state transitions in the regulation of tissue form and function