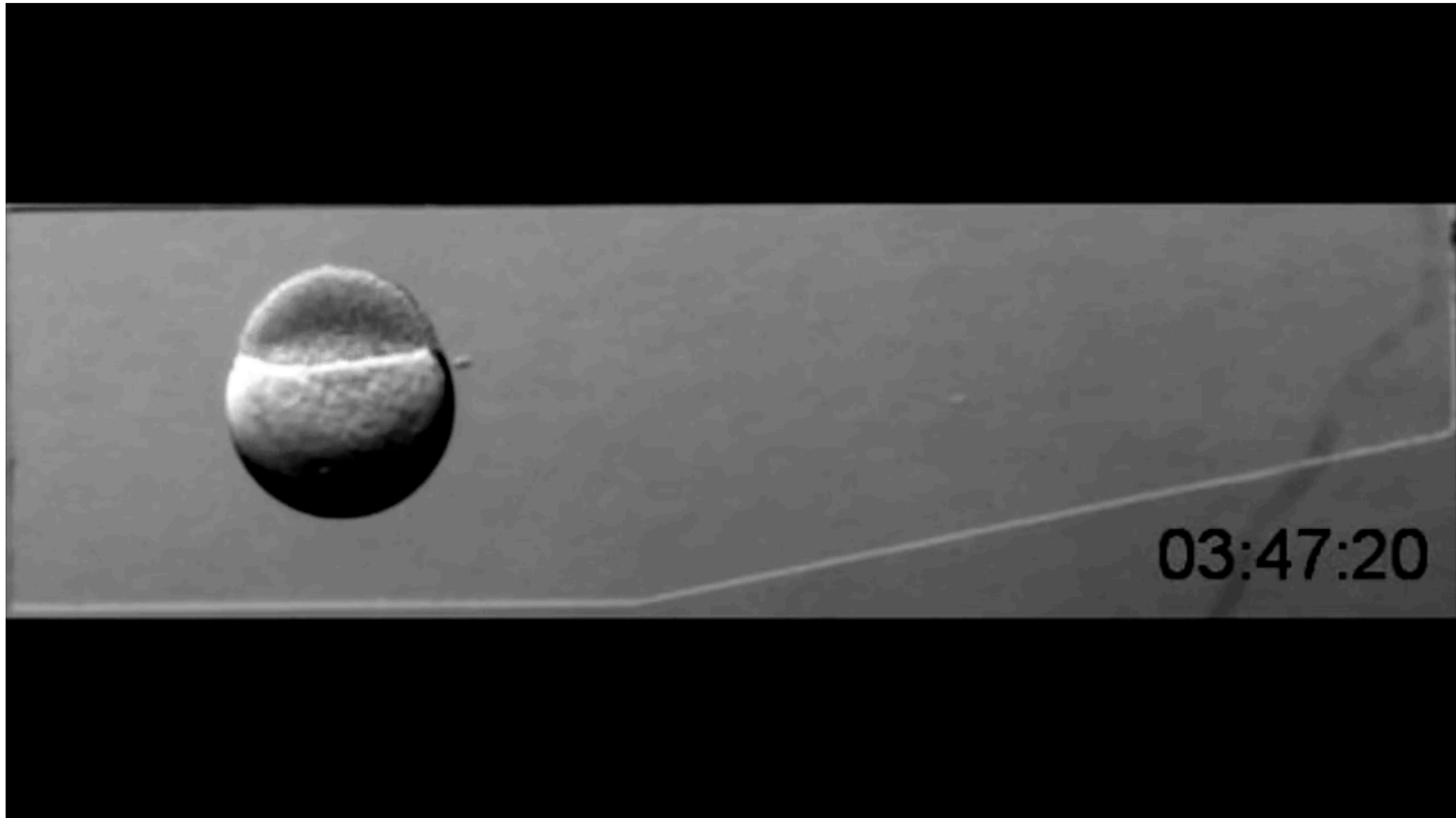


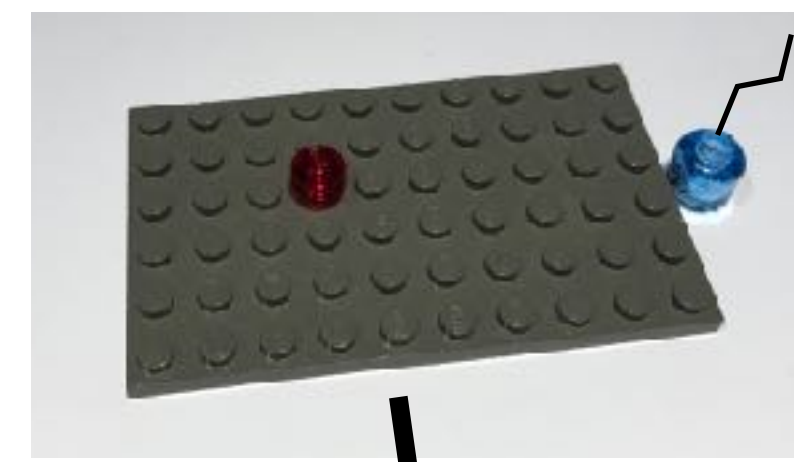


Big questions

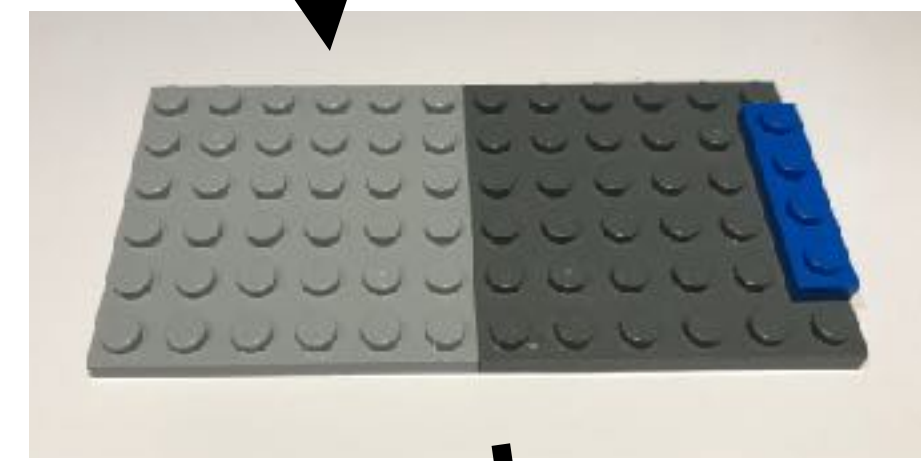


LEGO embryo - where are we?

Morphogenesis: Gastrulation and folding



Fertilisation

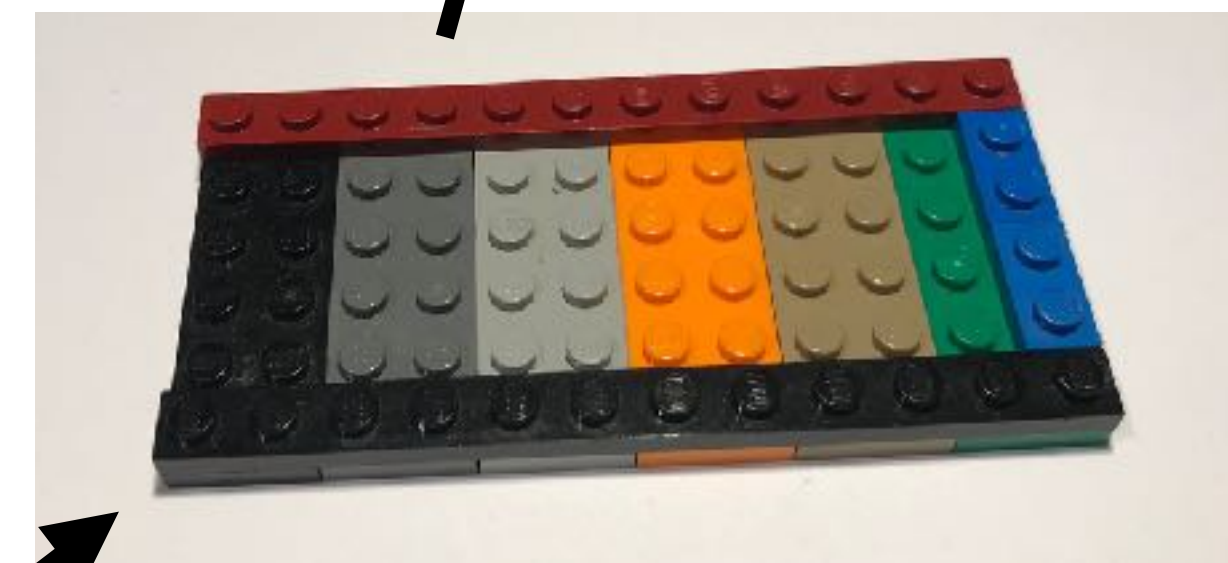
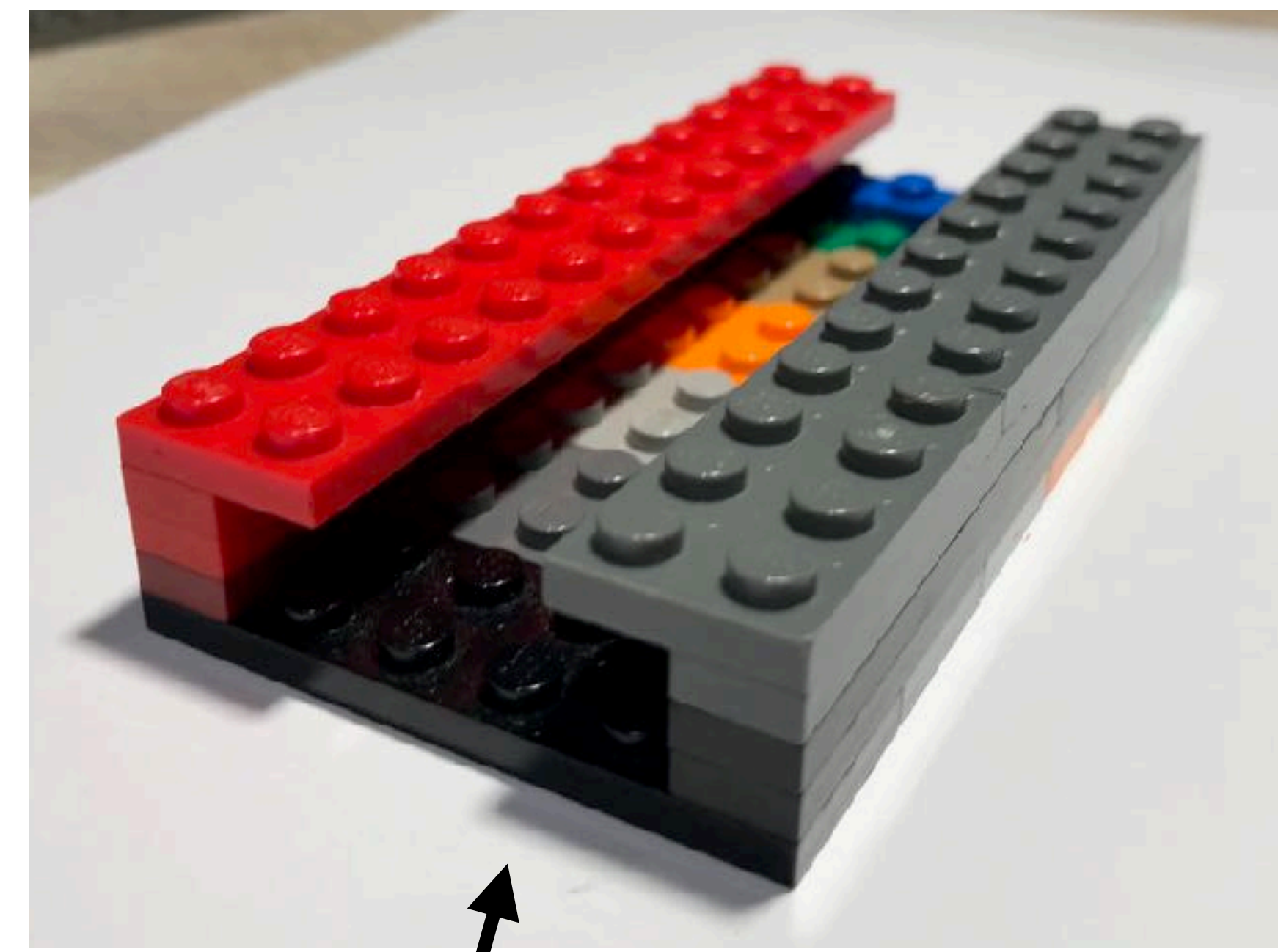


Symmetry breaking
Zygote



Maternal-to-Zygotic
transition

Early patterning
A-P axis



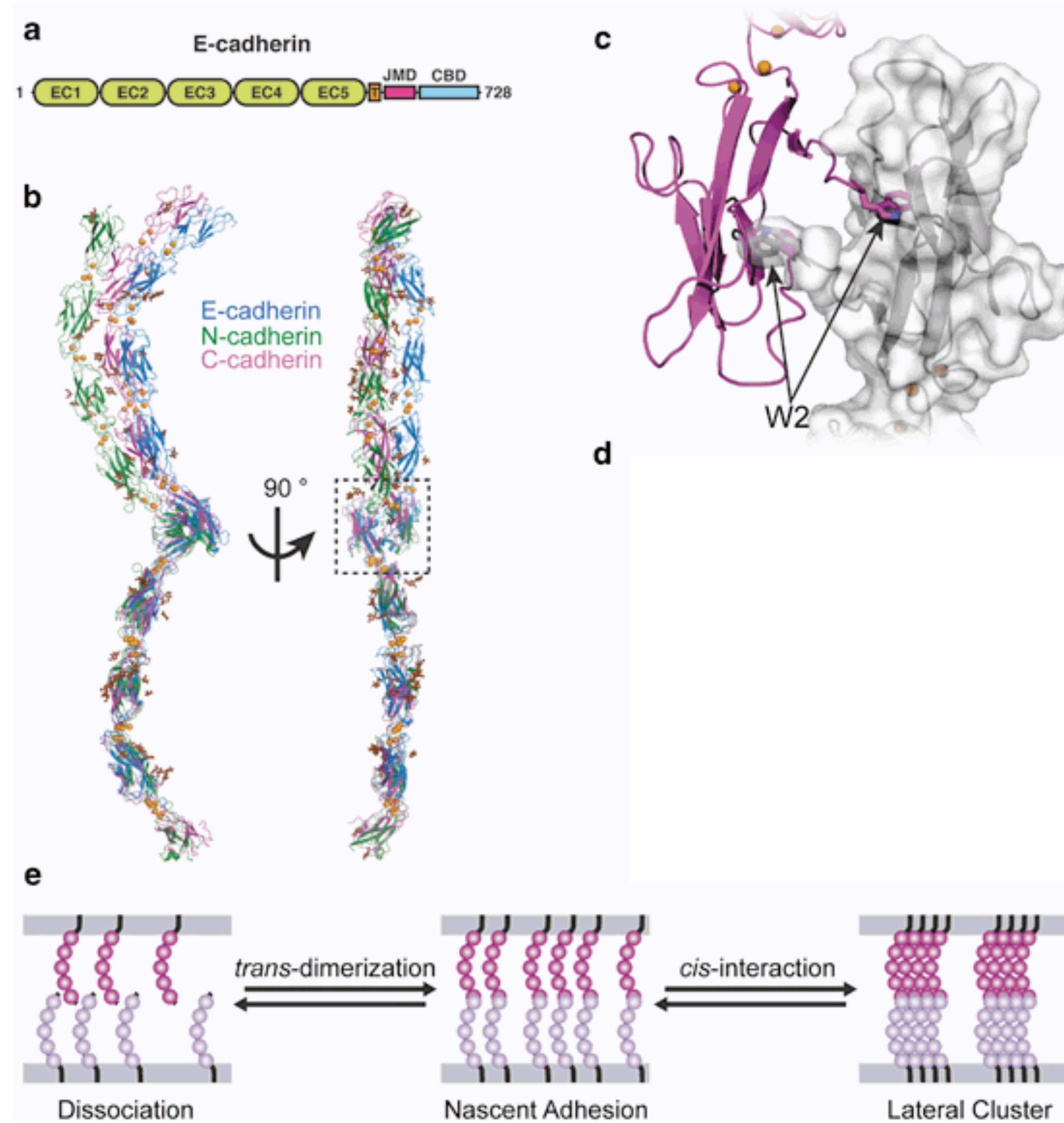
A-P, D-V and L-R axes in vertebrates

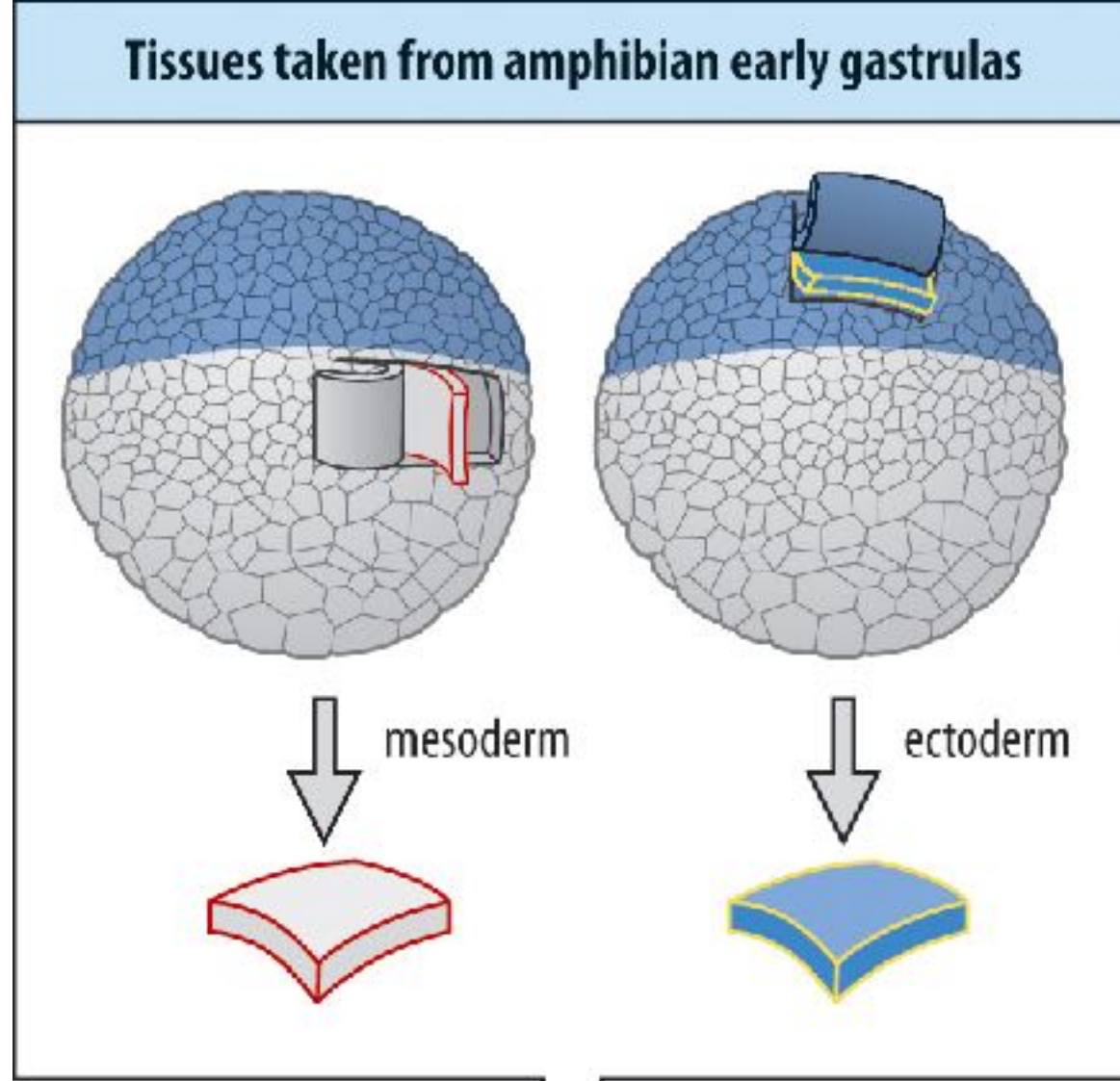
Today's menu

- **Morphogenesis**
- Basic mechanisms
- Cell-cell Adhesion
- Actin and myosin
- Oriented cleavage and blastocysts

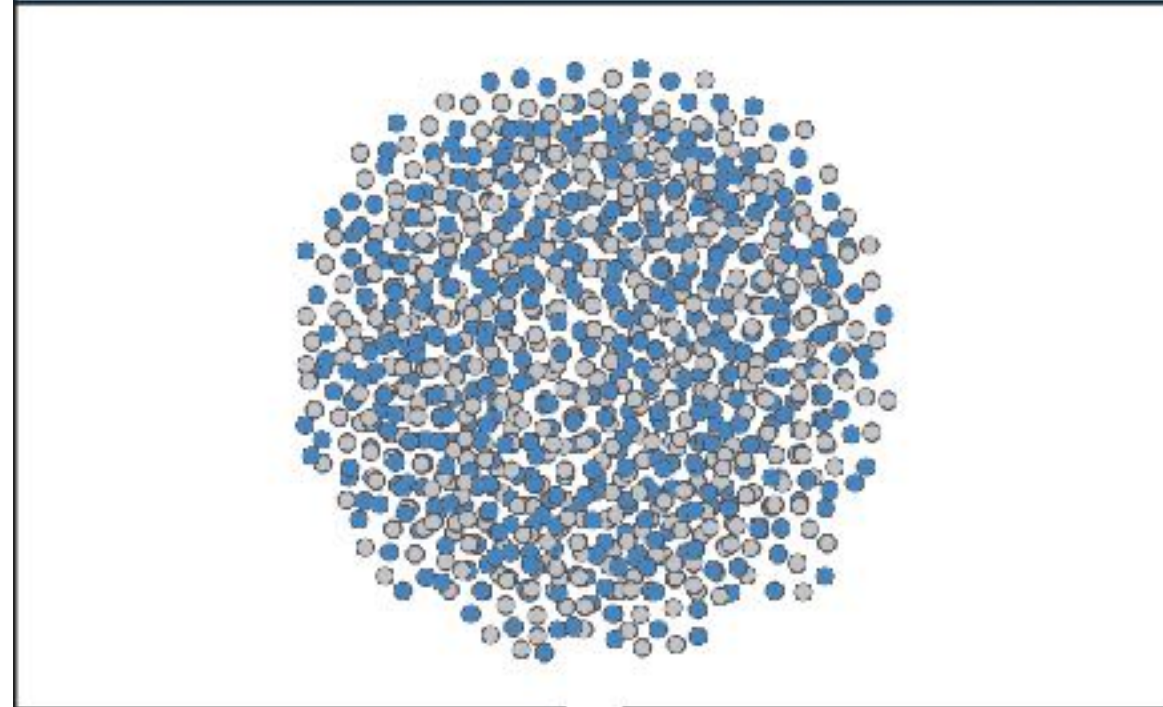
- **Gastrulation and folding**
- EMT, migration
- Bending the sheet
- Cell rearrangement in the sheet - convergent extension
- Other folding - brains and guts

Cadherin binding and clustering

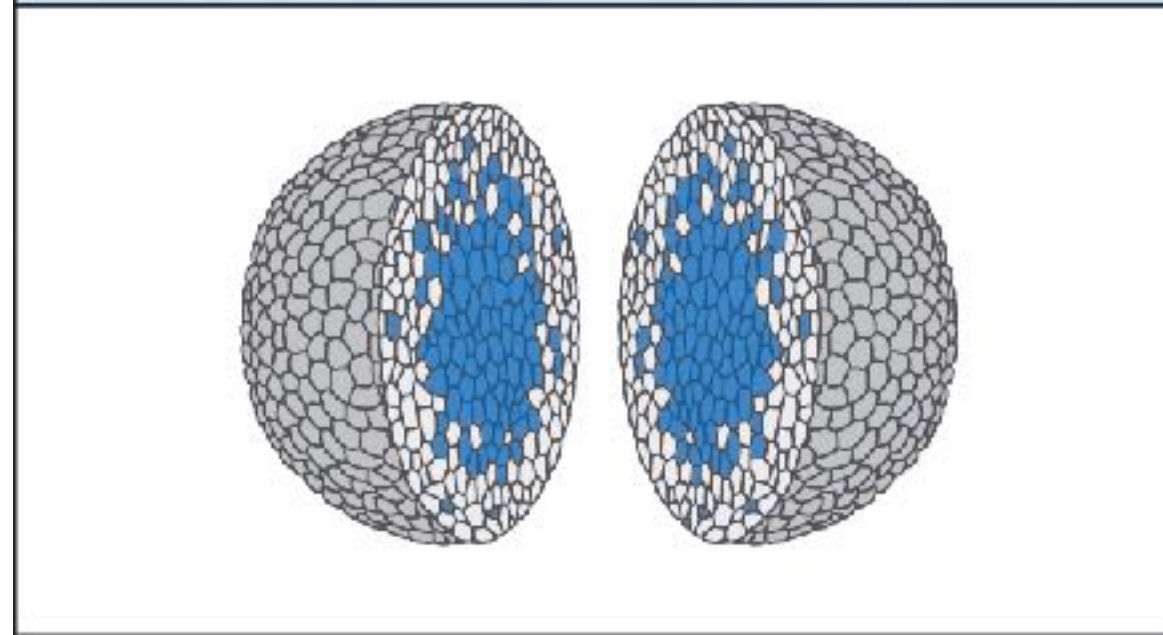




Tissues disaggregated into single cells



Spontaneous reaggregation. Cells sort out with mesodermal cells outermost



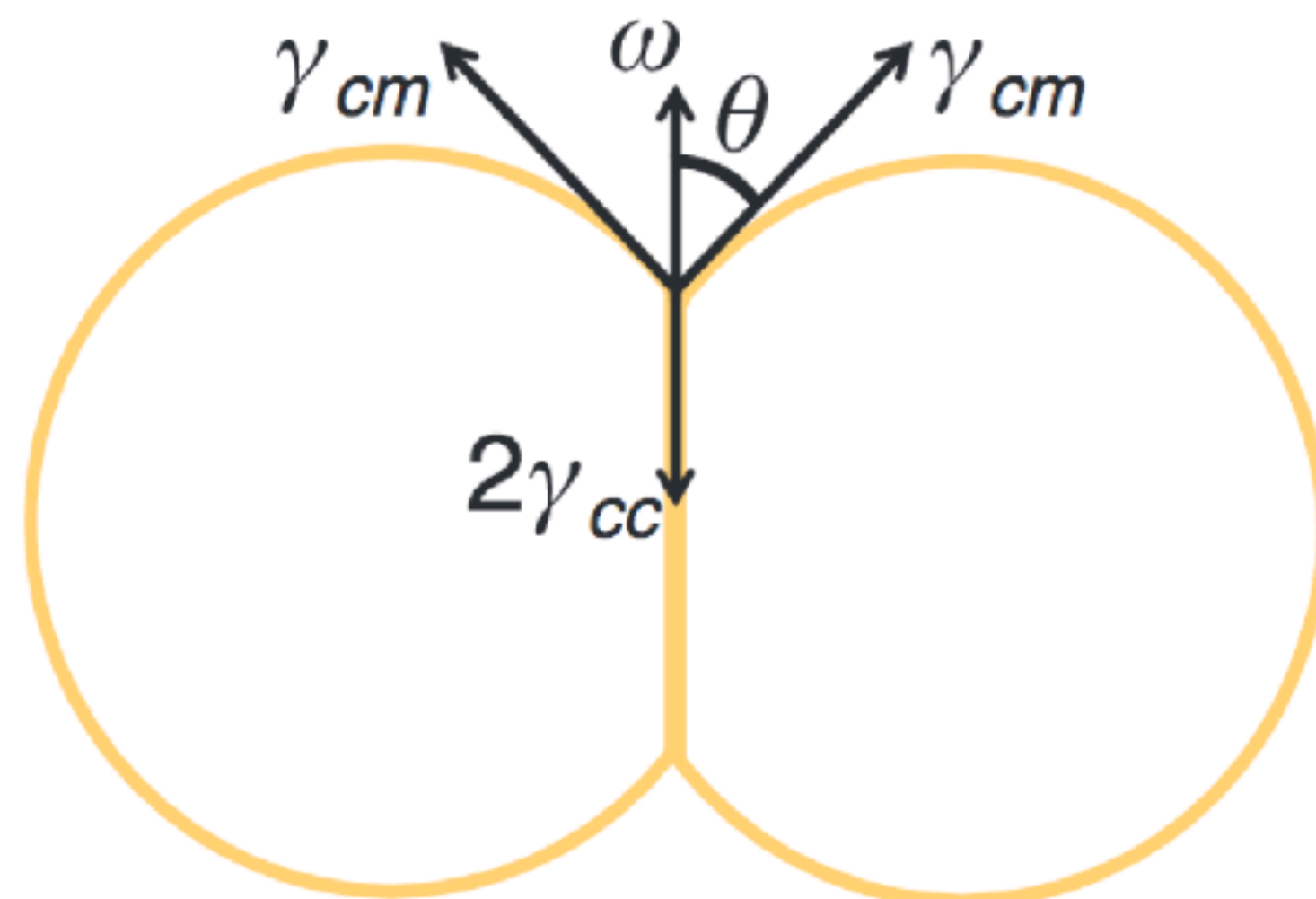
Cell sorting driven by differential surface tension and differential adhesion

- Surface tension: oil-water
- Surface tension modified by adhesion
- Cells with strongest interactions form inner layer

N-cadherin

E-cadherin

Balance of forces at contact point



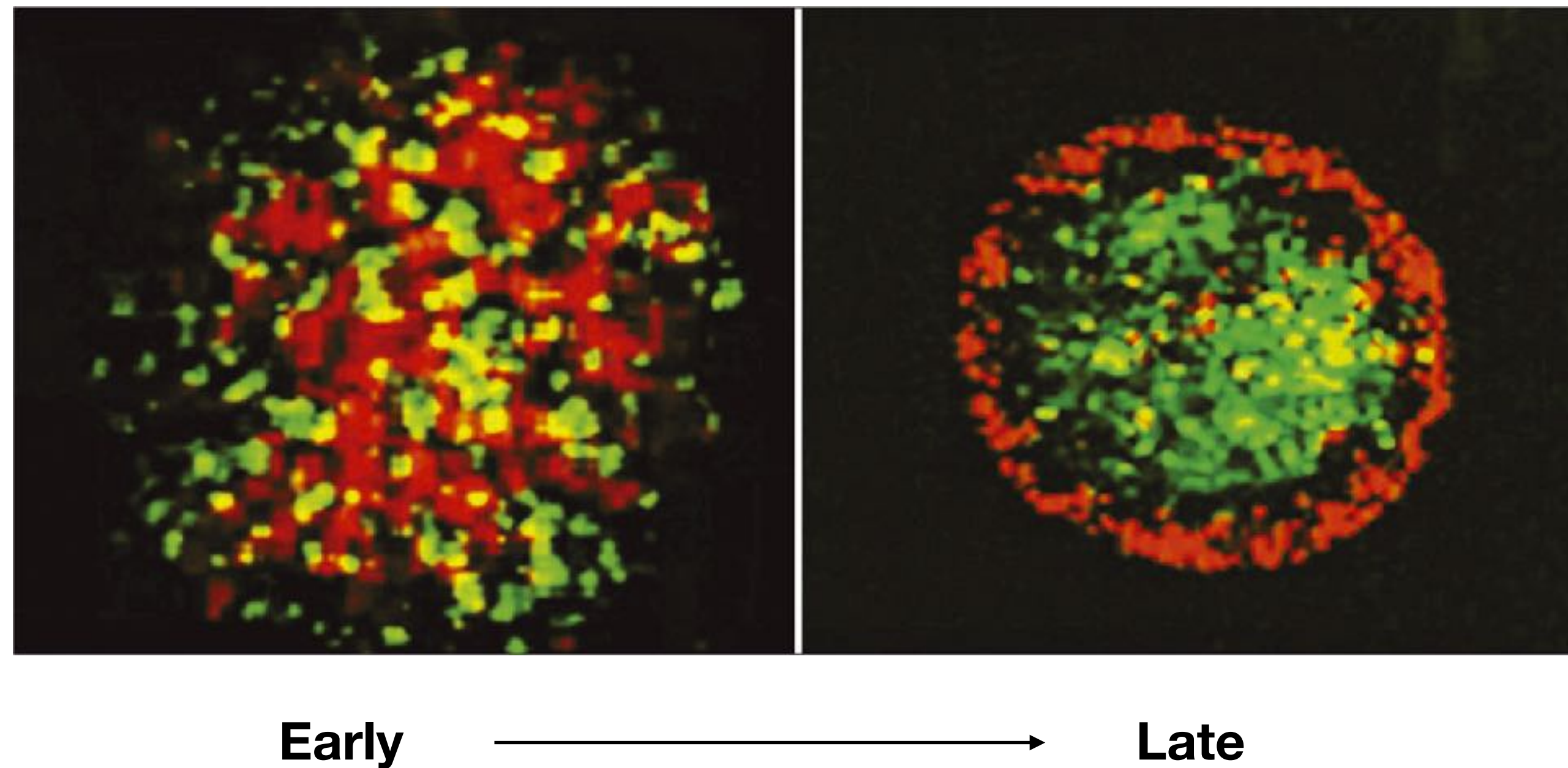
$$\cos \theta = \frac{2\gamma_{cc} - \omega}{2\gamma_{cm}}$$

- Tension from cortex contact with medium (γ_{cm})
- Tension from cortex contact with cell (γ_{cc})
- Tension from adhesion between cells (ω)

Individual puzzle

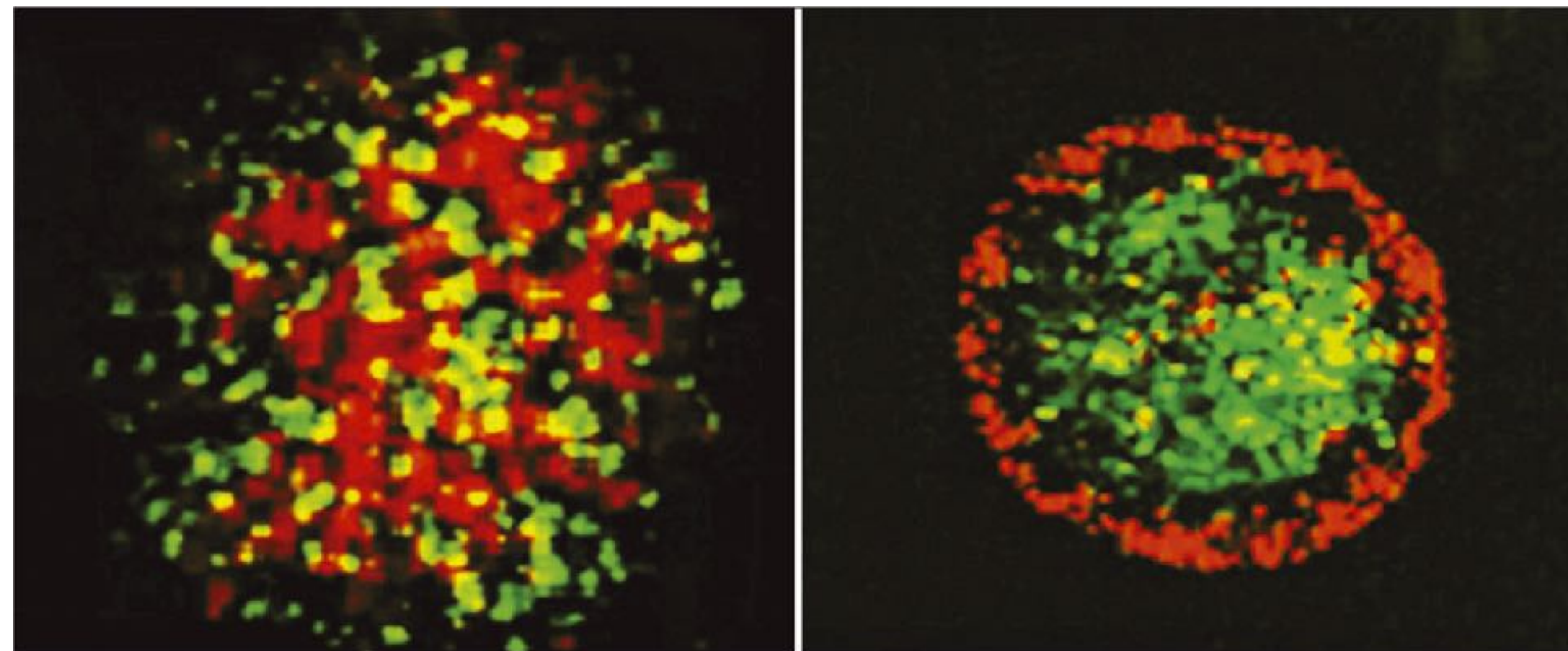


Cadherin *type* and *level* can drive cell sorting

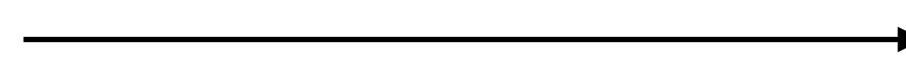


How could we explain this structure *only* with N-Cadherin?

Cadherin *type* and *level* can drive cell sorting



Early



Late

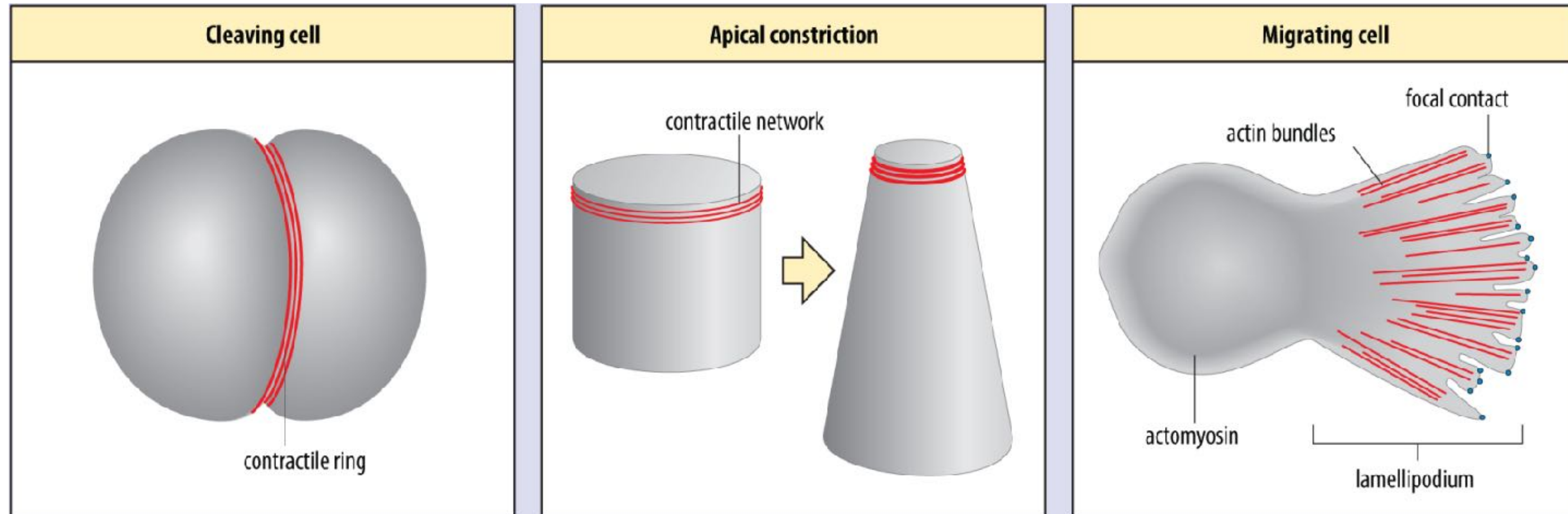
High N-cadherin

Low N-cadherin

Cellular “building blocks” of morphogenesis

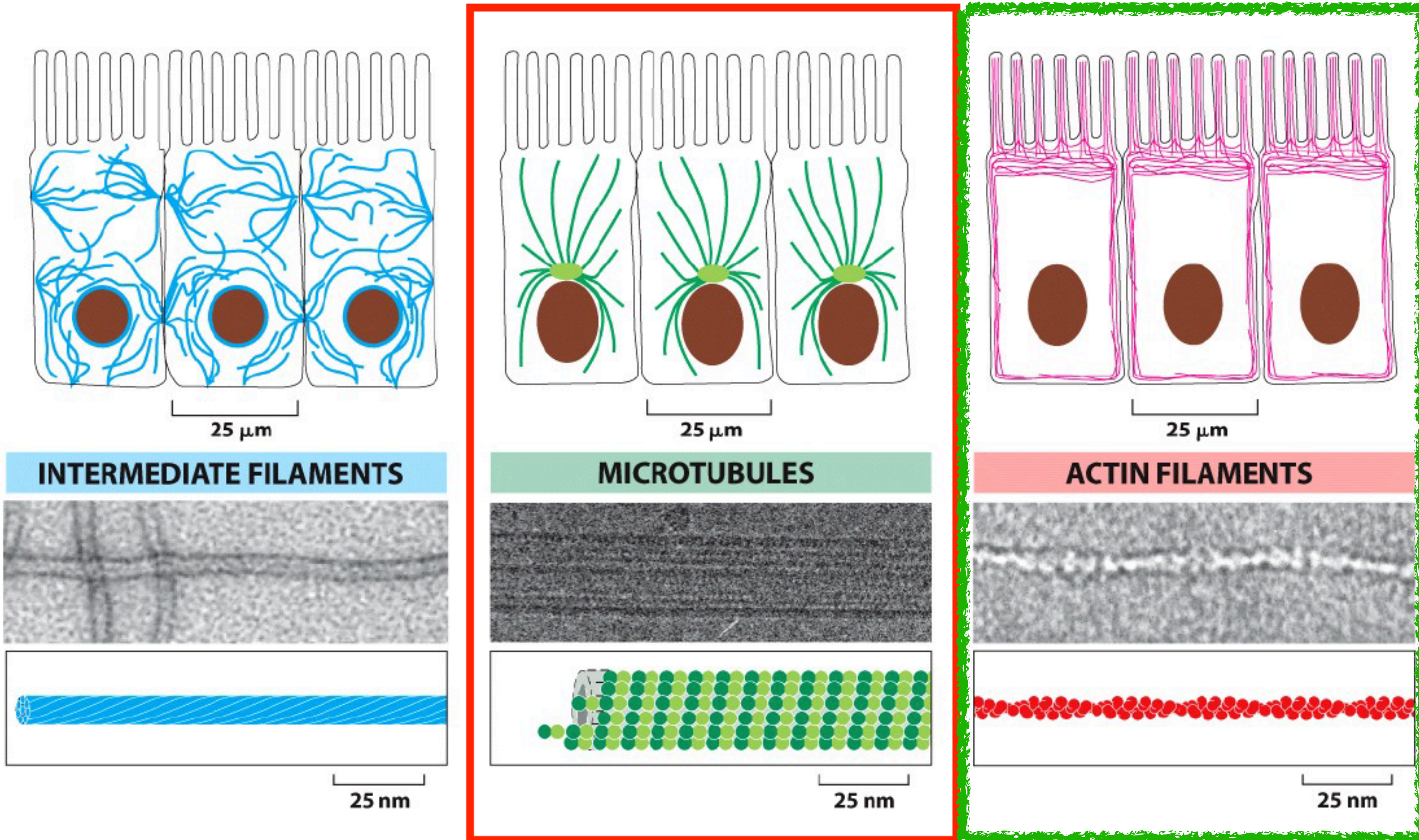
Oriented division

Death

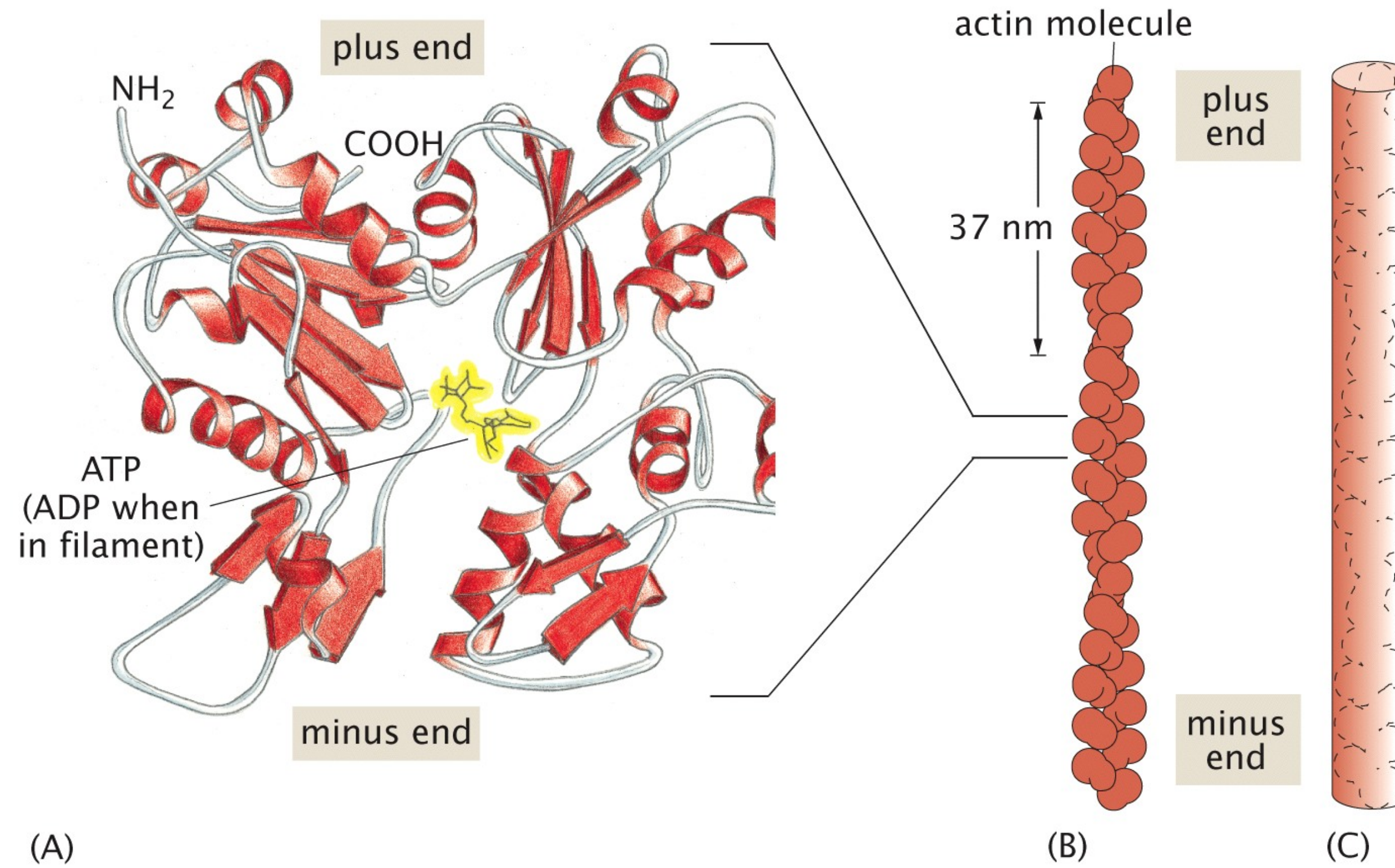




● Three principal cytoskeletal polymers



The actin polymer



(A) Figure 10.29 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

Actin filaments are floppier than microtubules

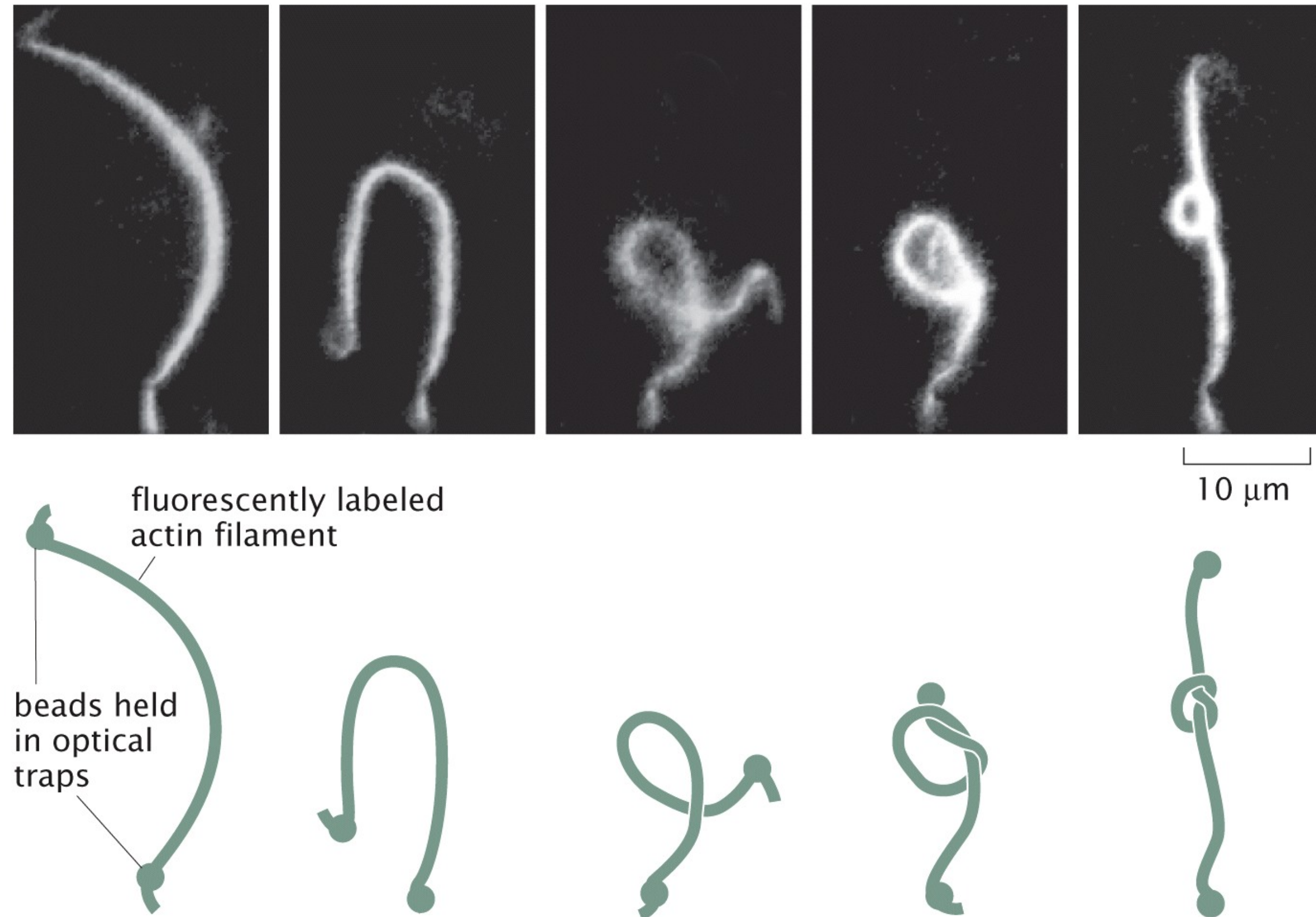


Figure 10.30 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

Myosin motors walk along actin filaments

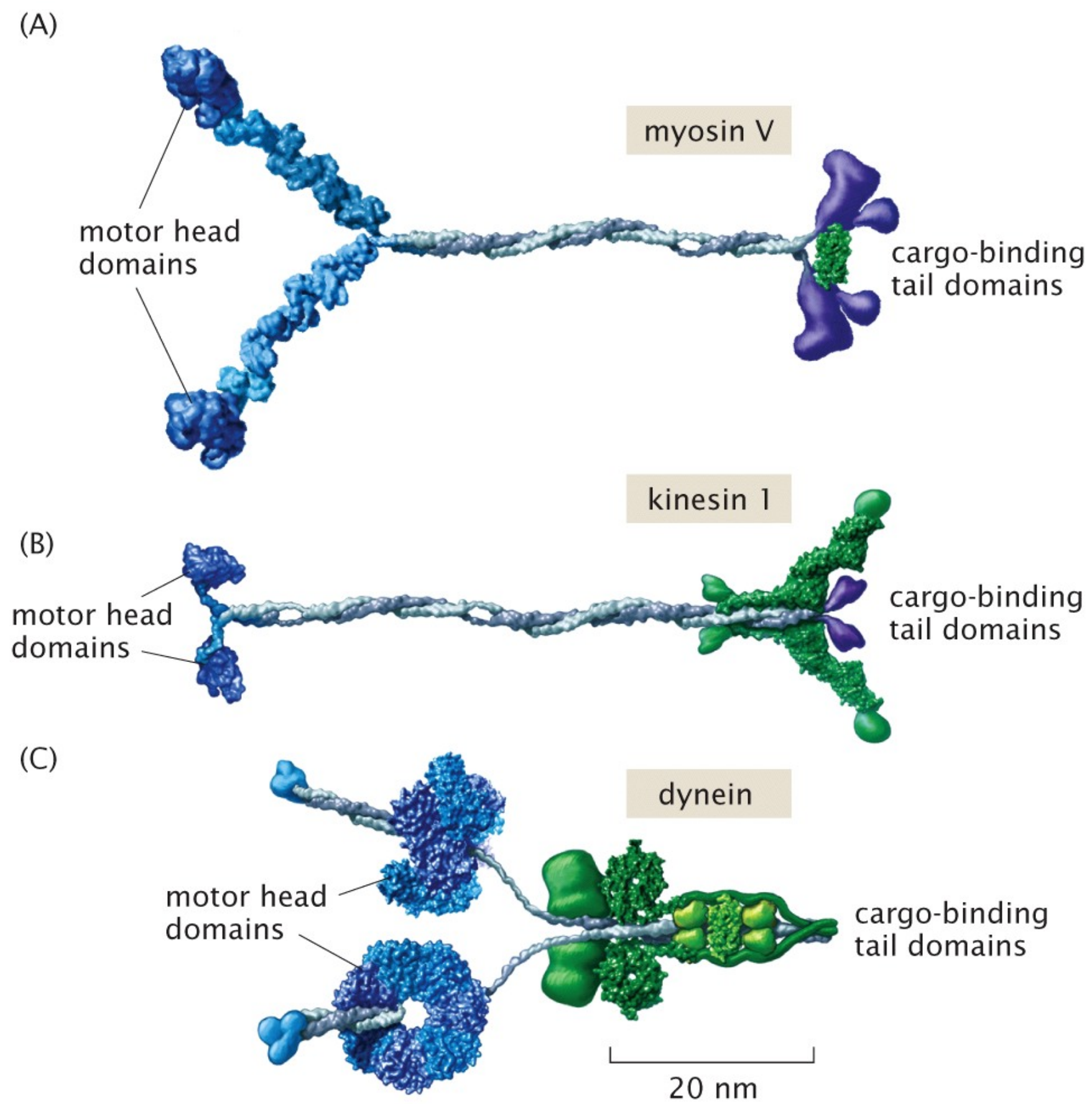


Figure 16.2 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

Myosin II

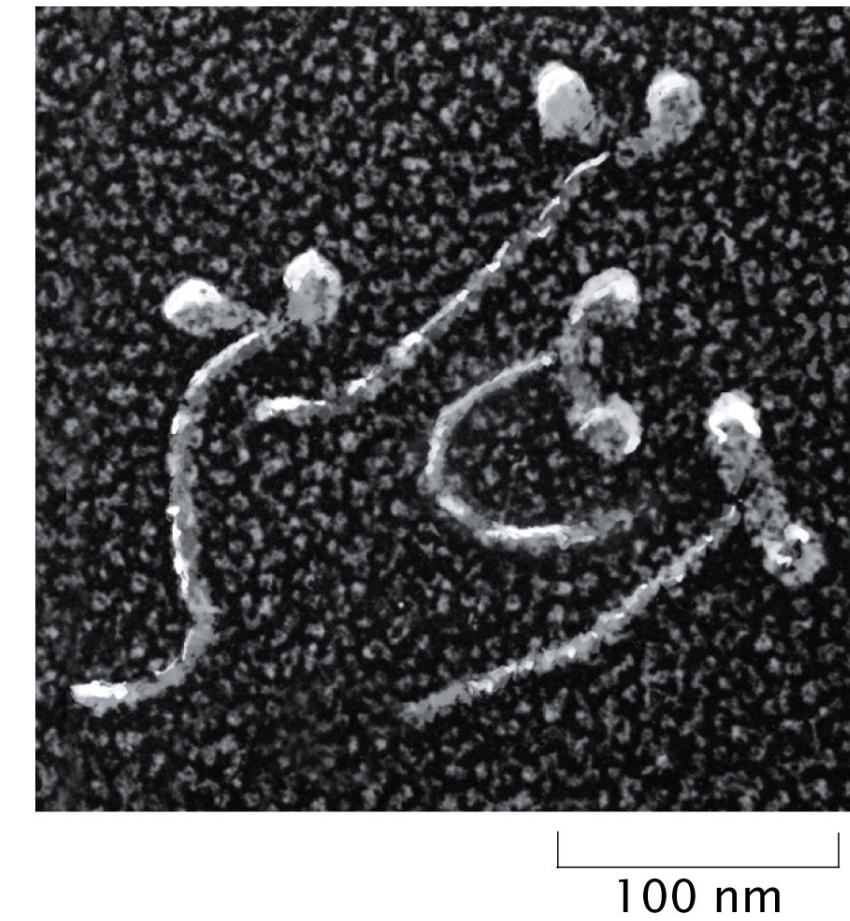
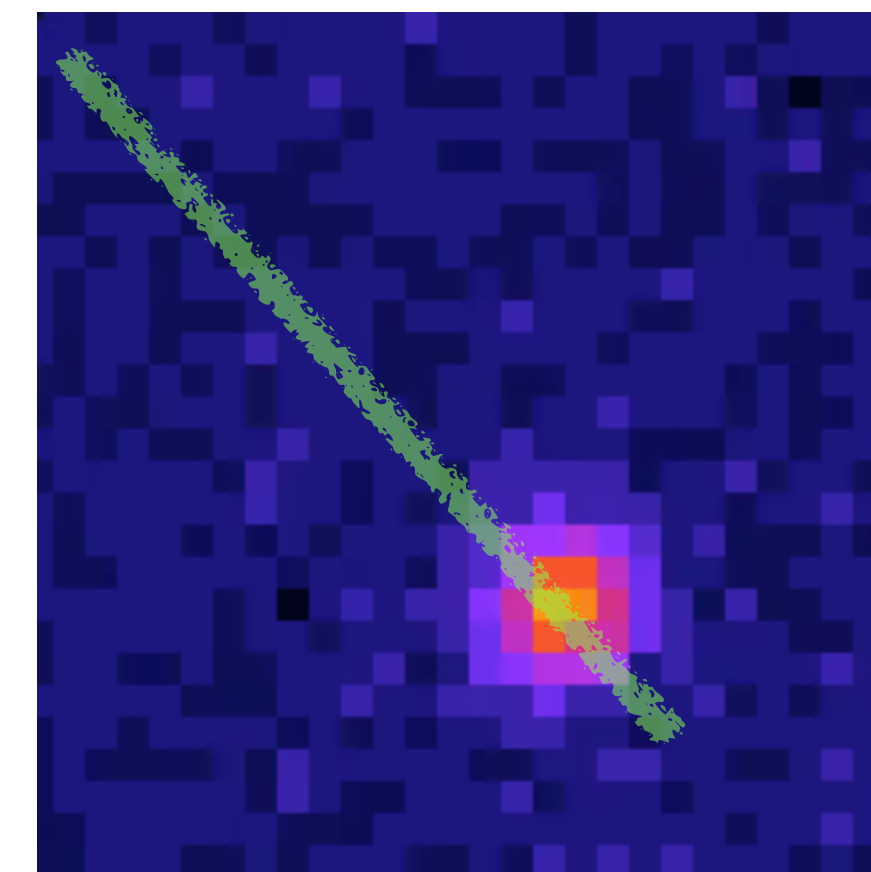


Figure 16.6b Physical Biology of the Cell, 2ed. (© Garland Science 2013)



Myosin II - Fluorescent bead

Myosin II *multimers* act to contract actin fibres

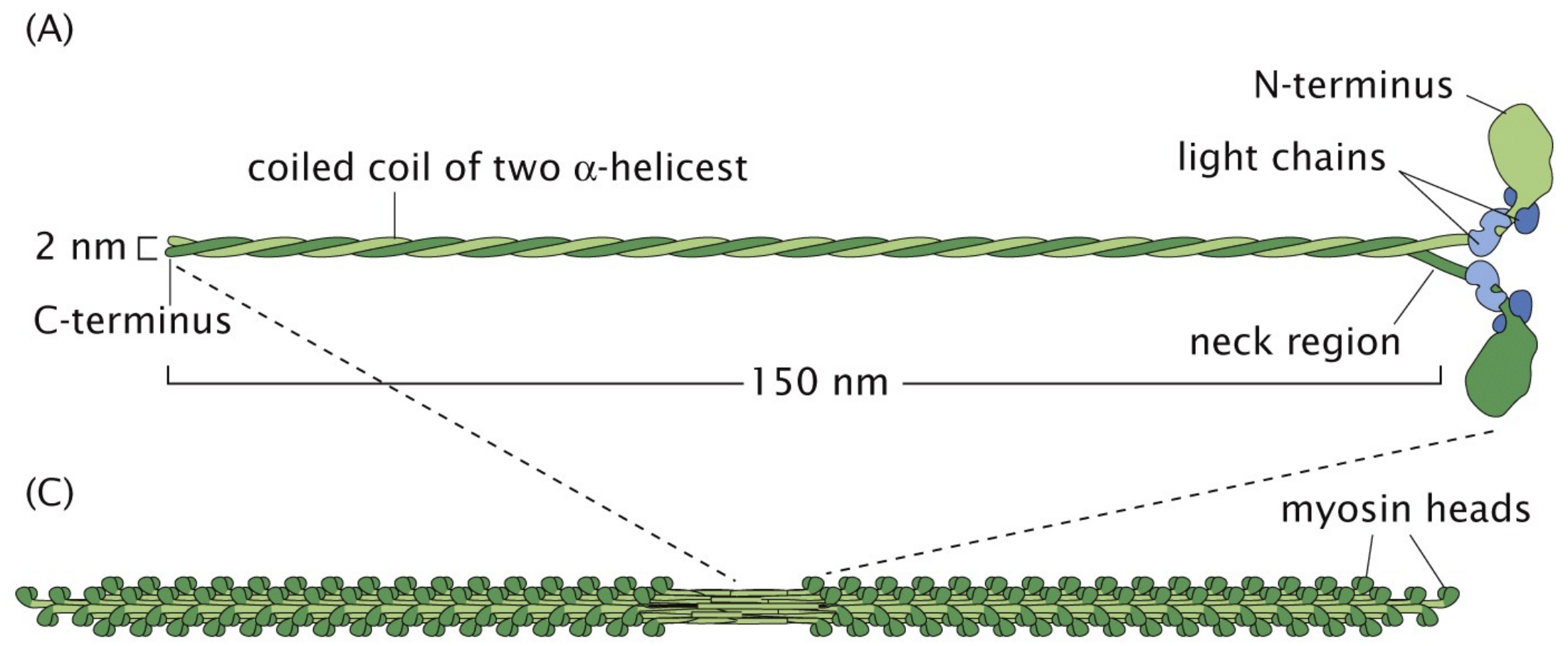


Figure 16.6ac Physical Biology of the Cell, 2ed. (© Garland Science 2013)

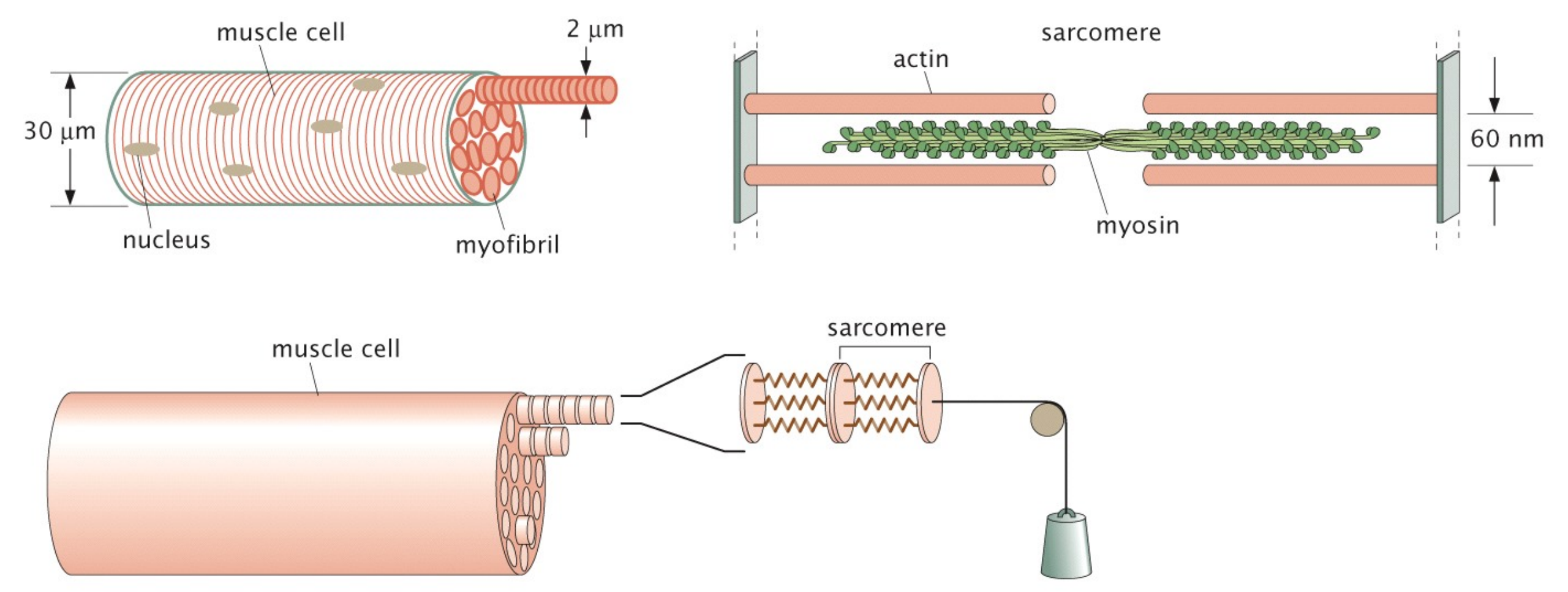


Figure 16.8 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

Myosin II *activity* regulated by phosphorylation cycle

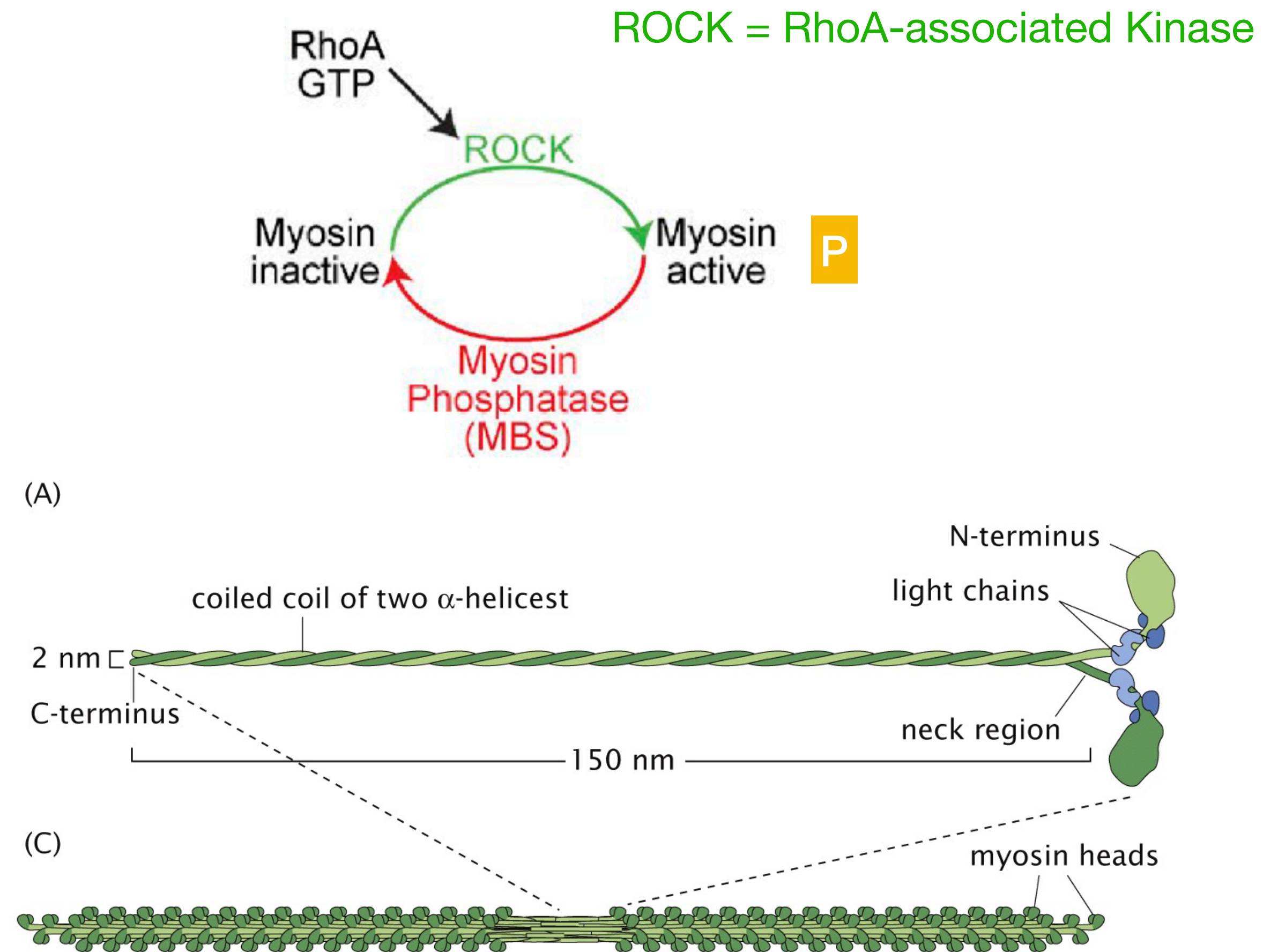
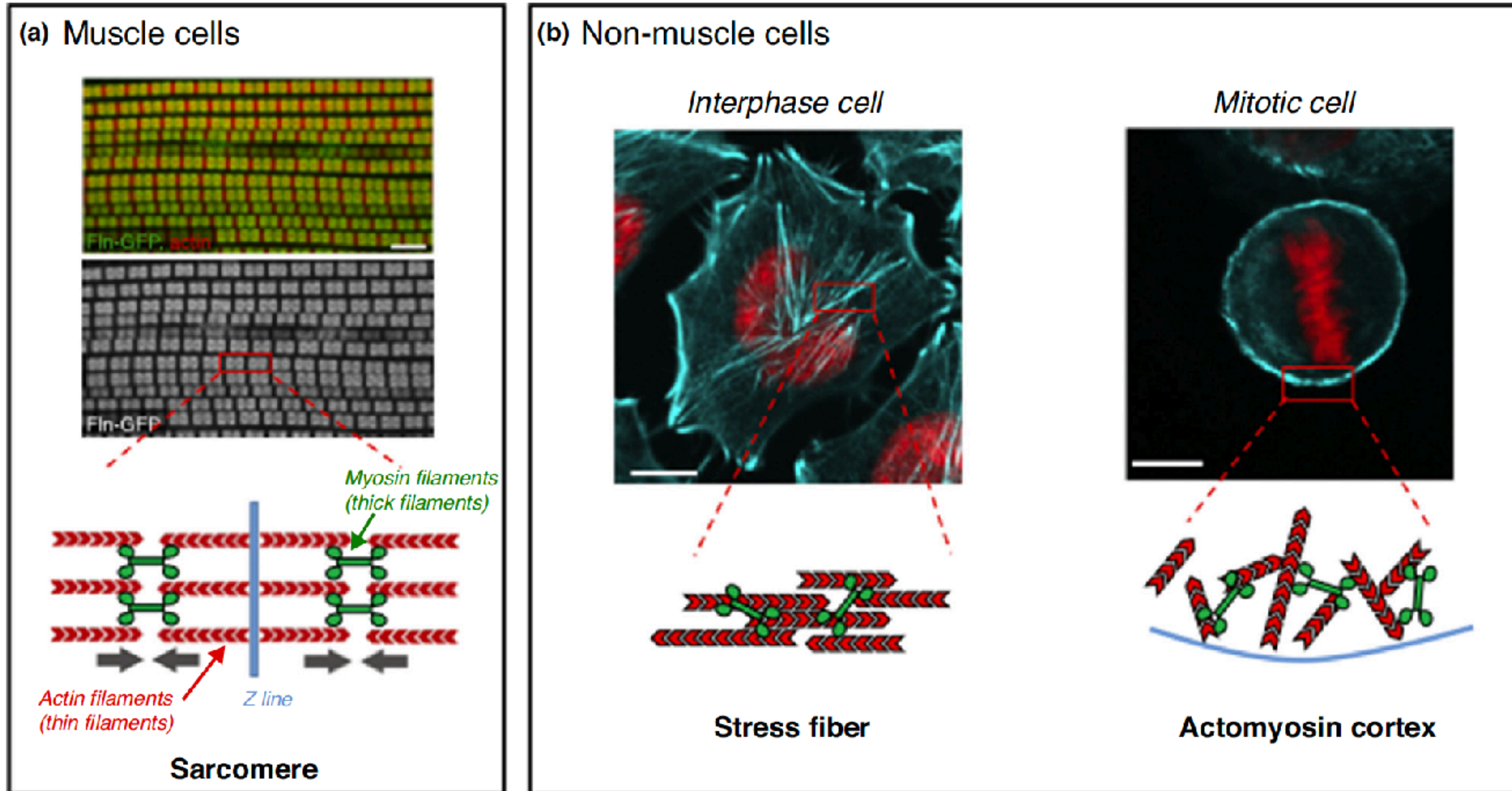


Figure 16.6ac Physical Biology of the Cell, 2ed. (© Garland Science 2013)

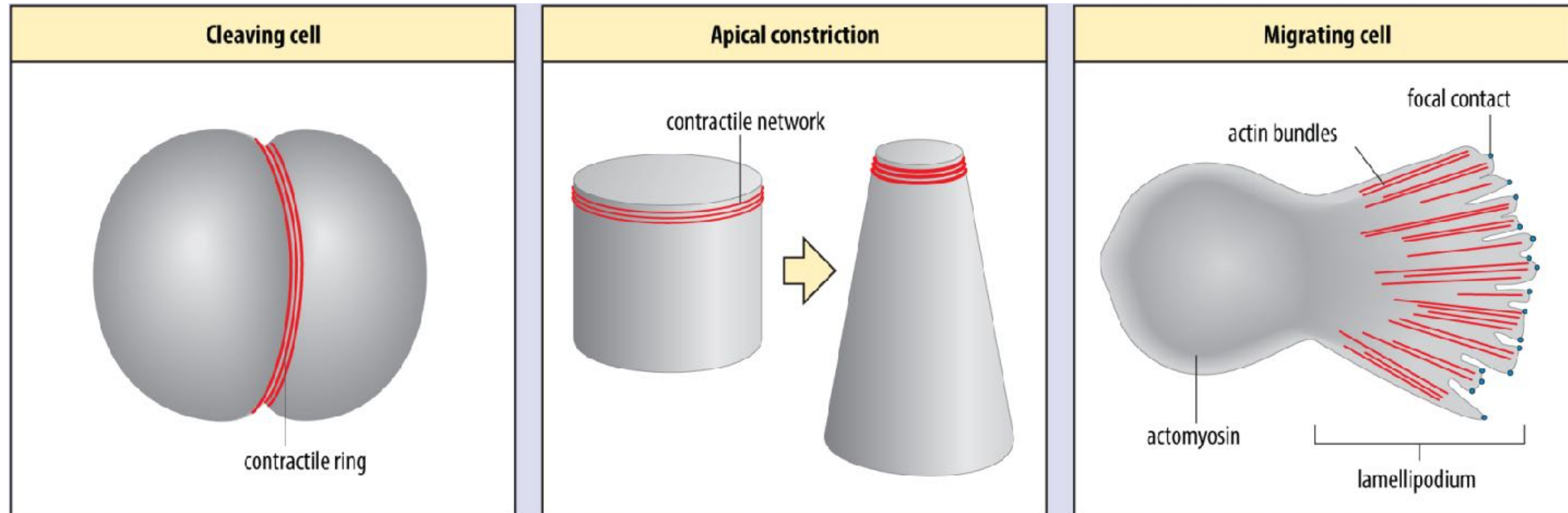
Architecture of actomyosin shapes contractility



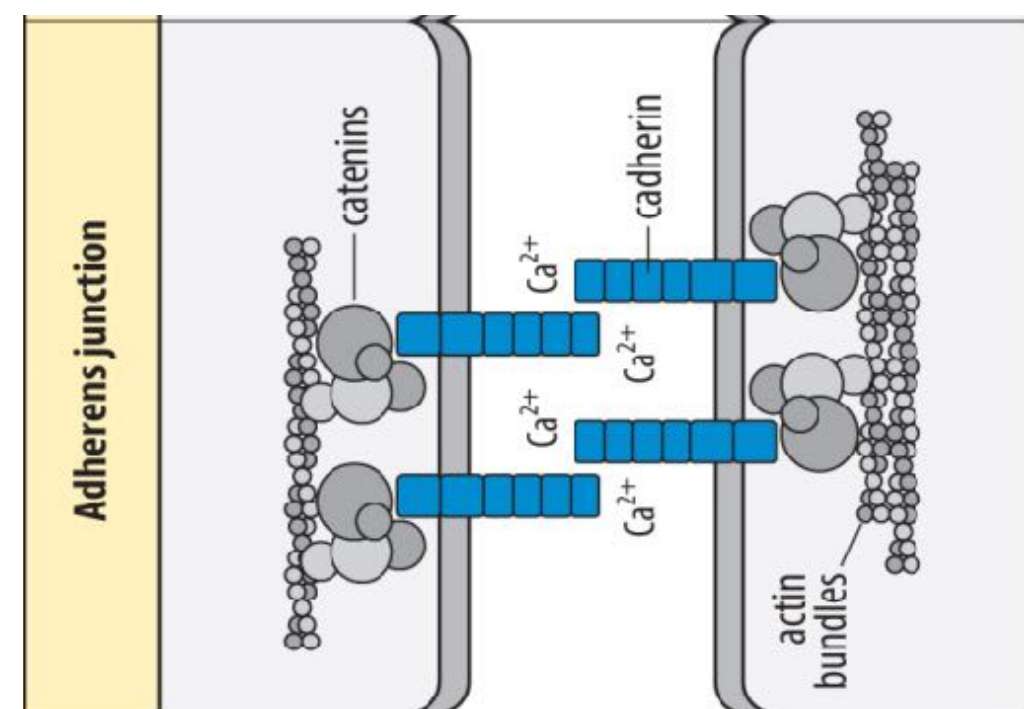
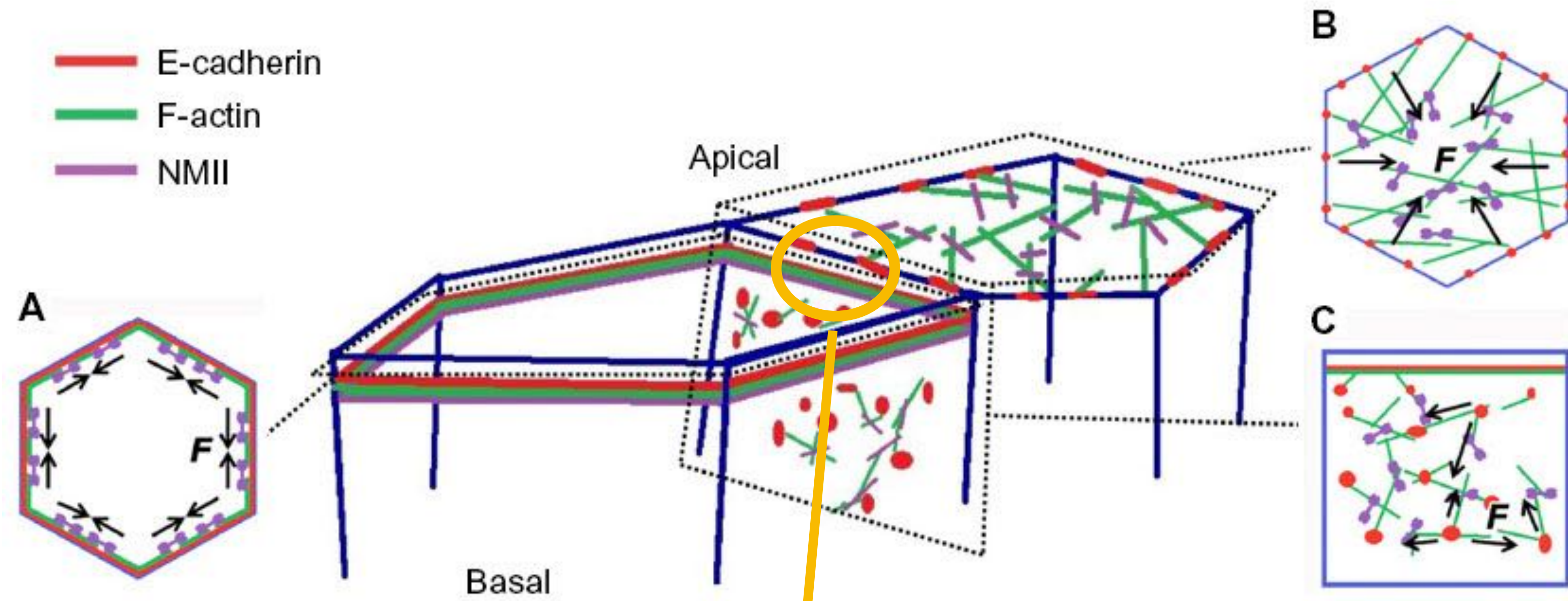
Cellular “building blocks” of morphogenesis

Oriented division

Death

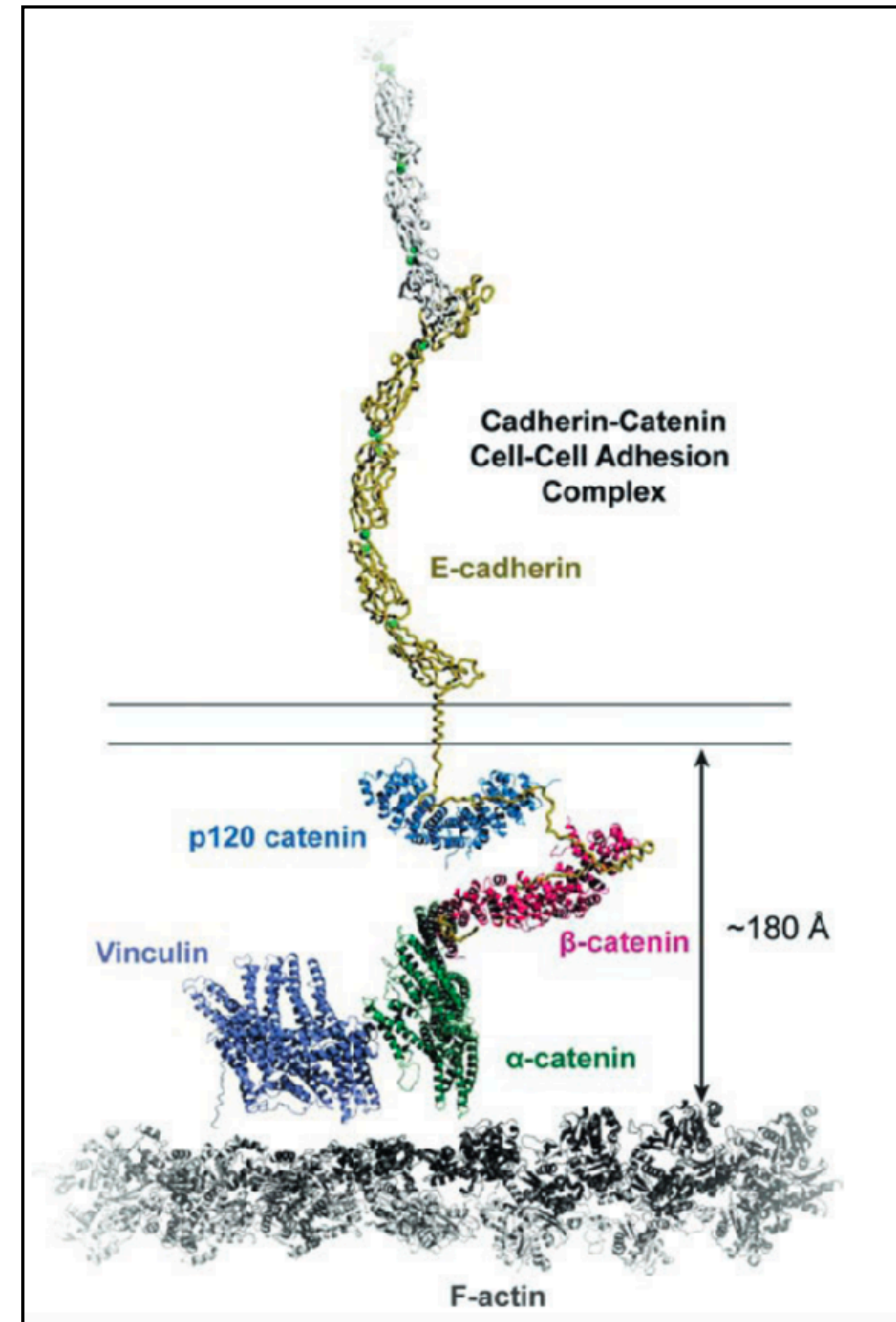
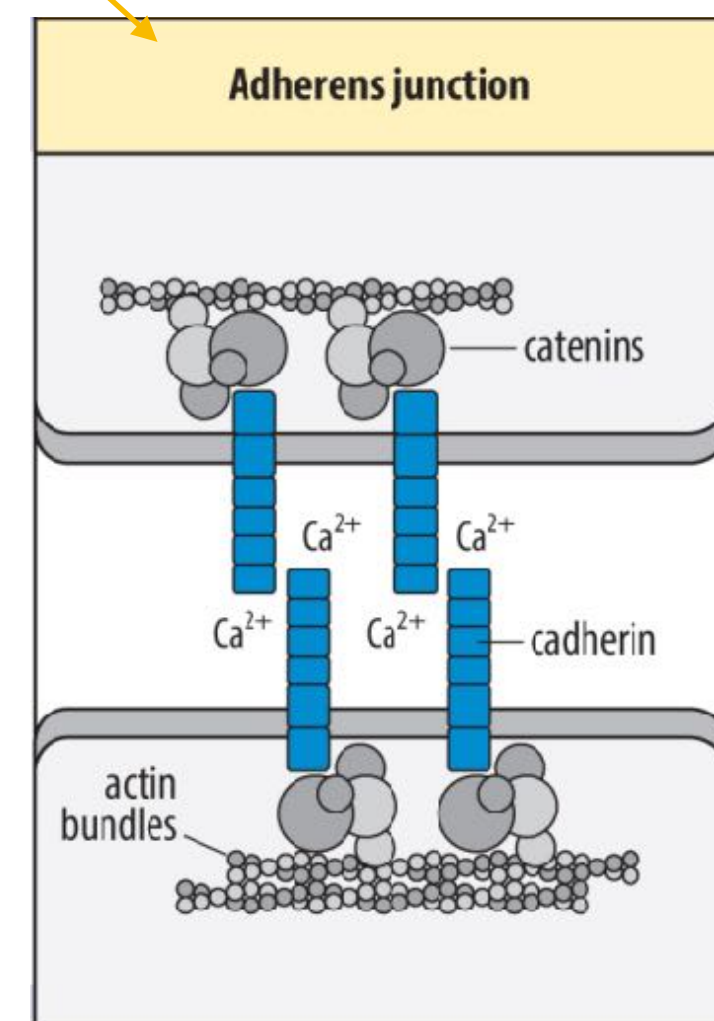
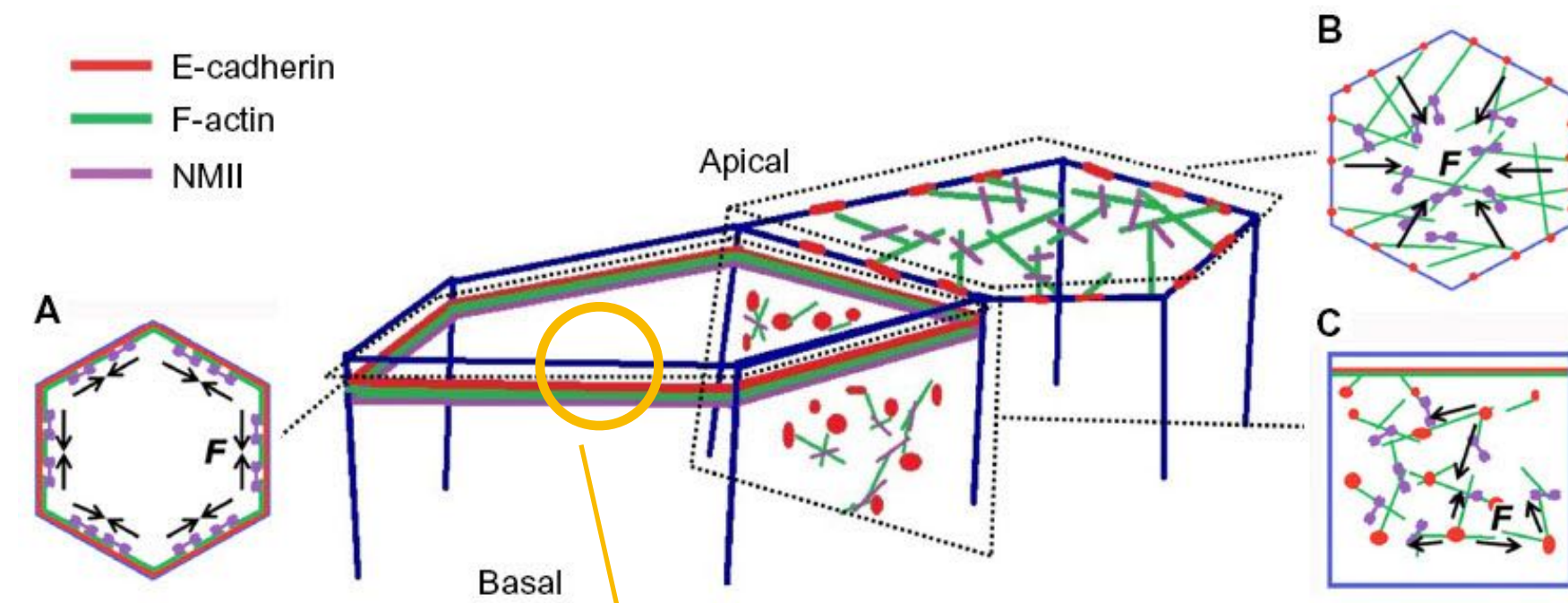


Epithelial structure and actomyosin activity

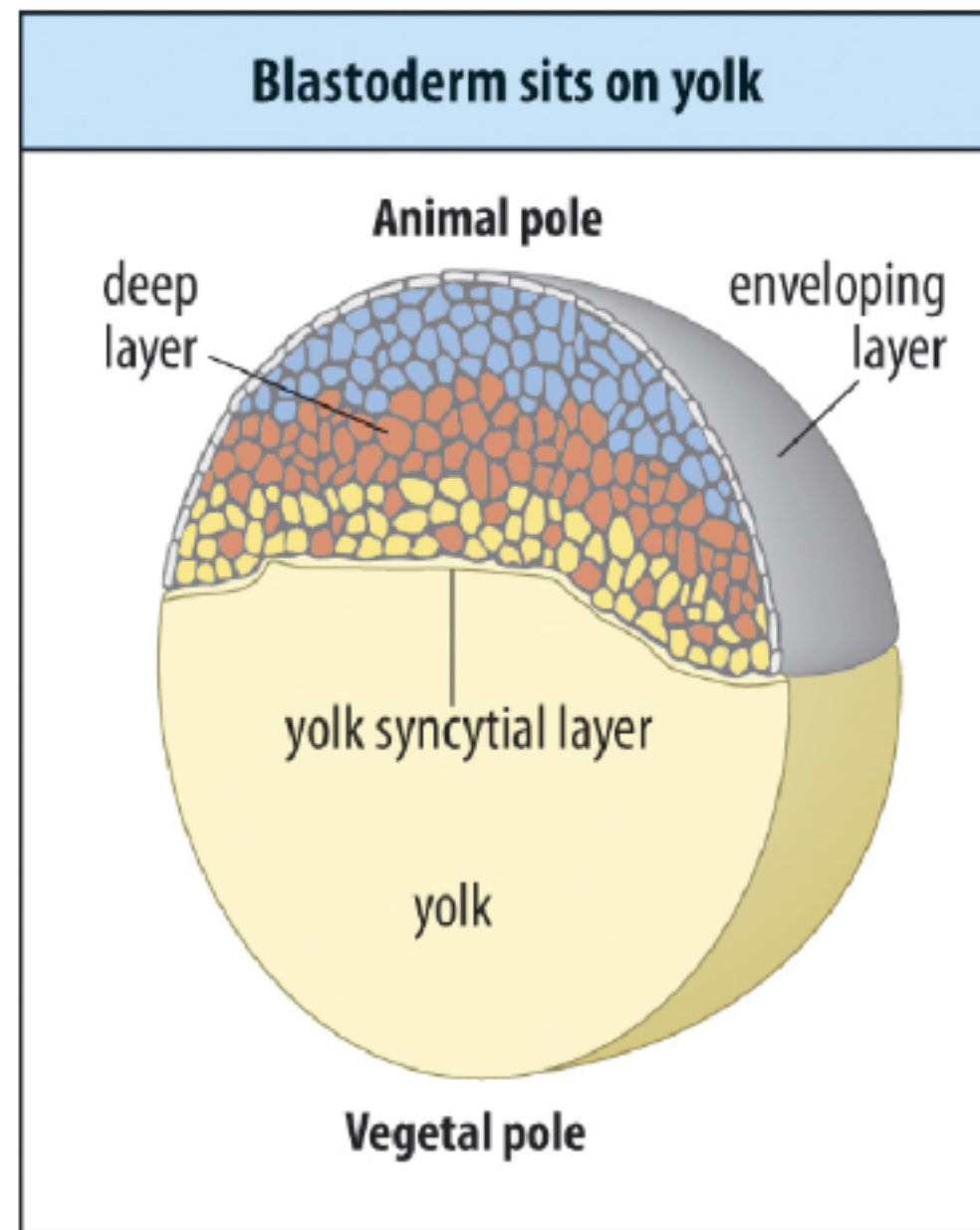


Epithelium often modelled as 2D network

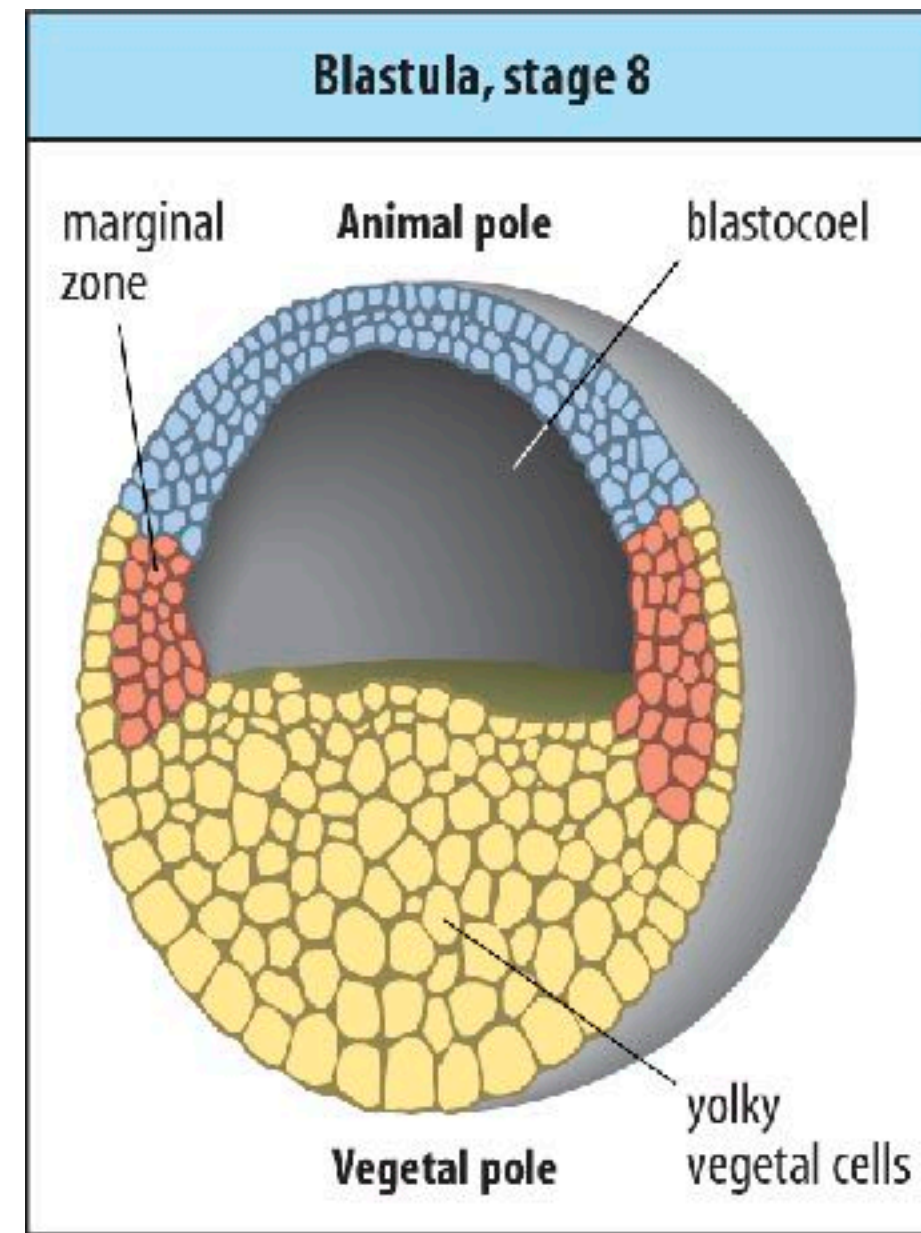
Epithelial structure and actomyosin activity



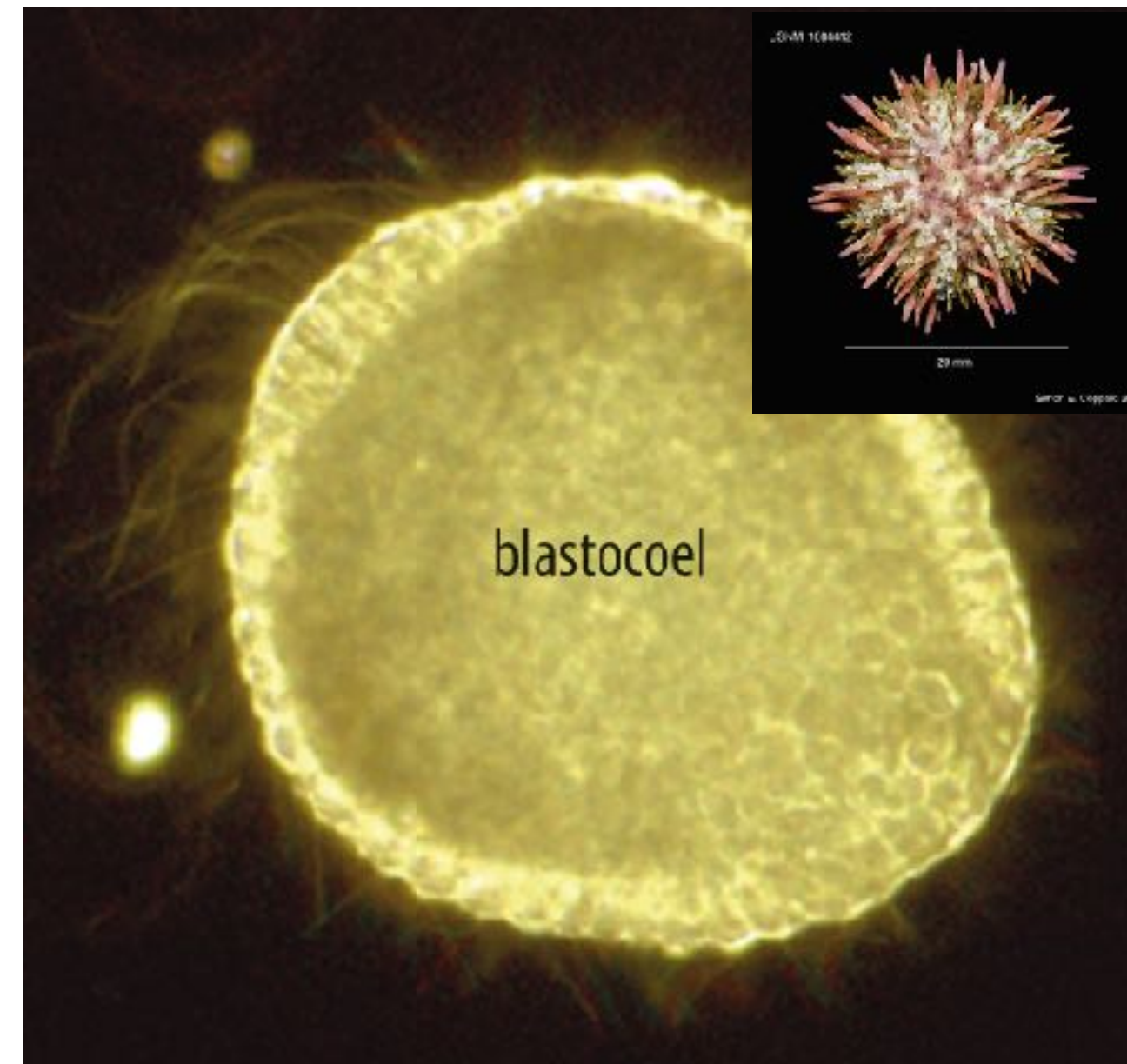
Oriented cleavage and formation of the blastula



zebrafish embryo

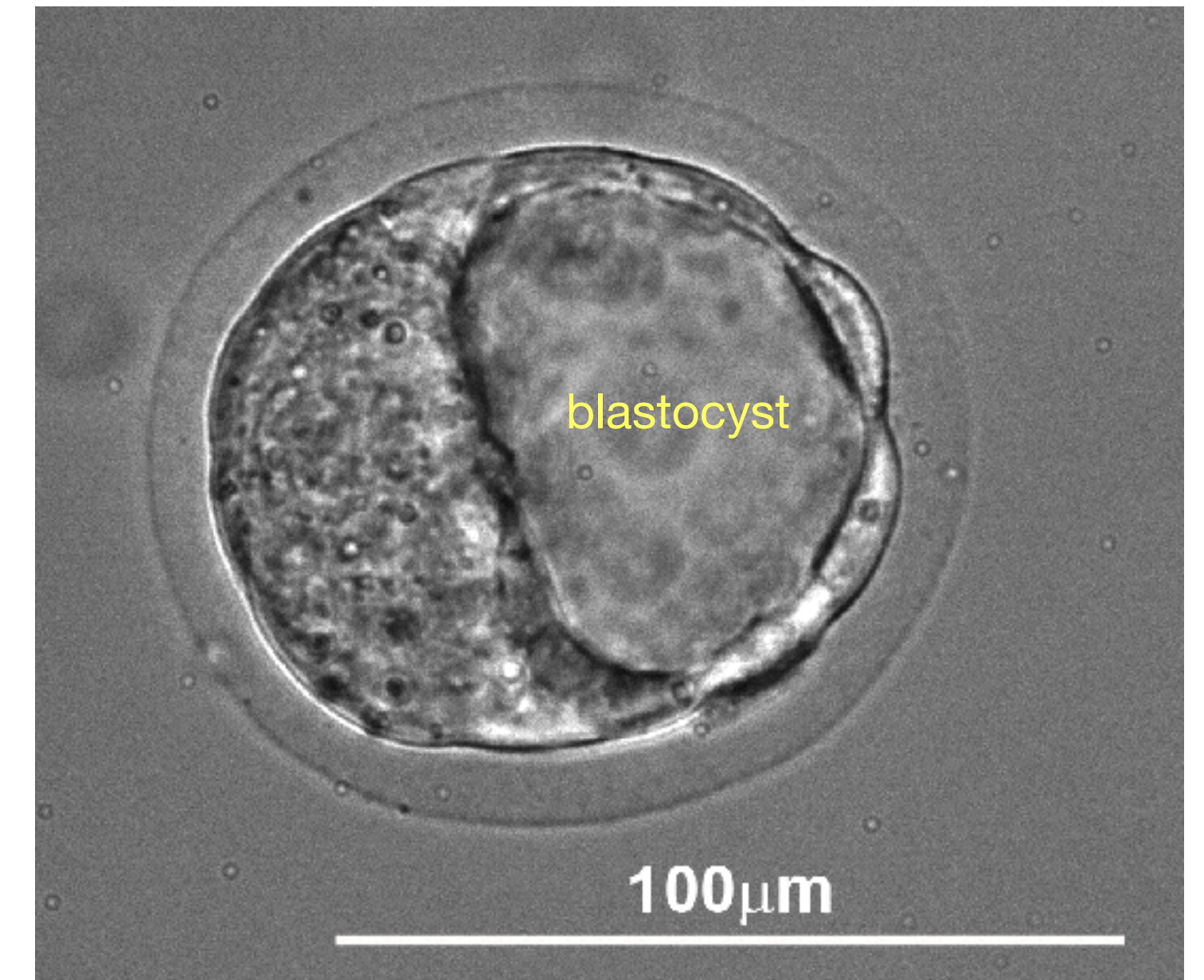


Frog embryo



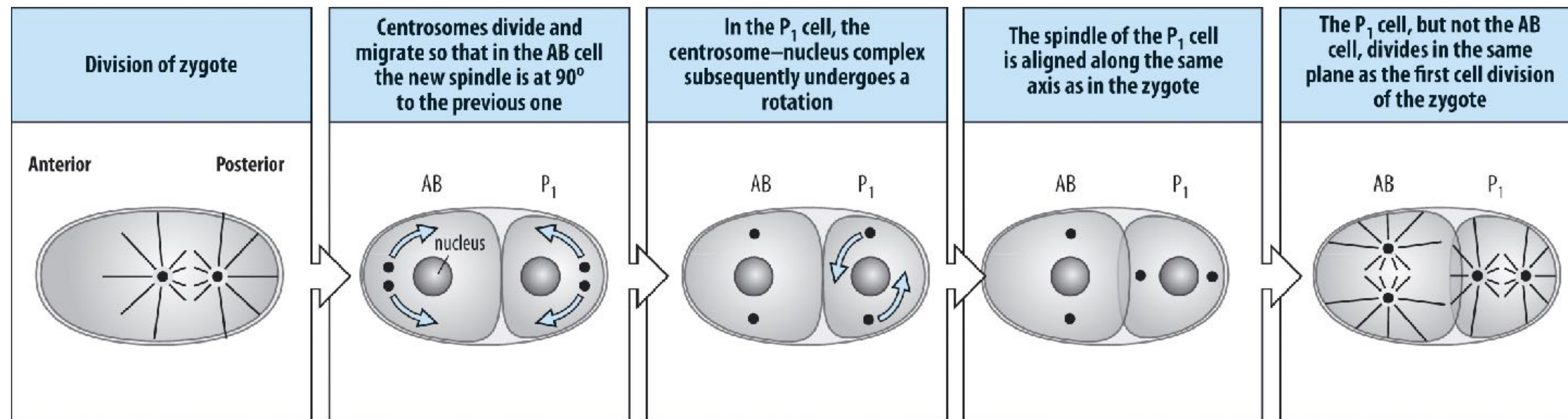
Sea-urchin embryo

50 microns



Mouse embryo

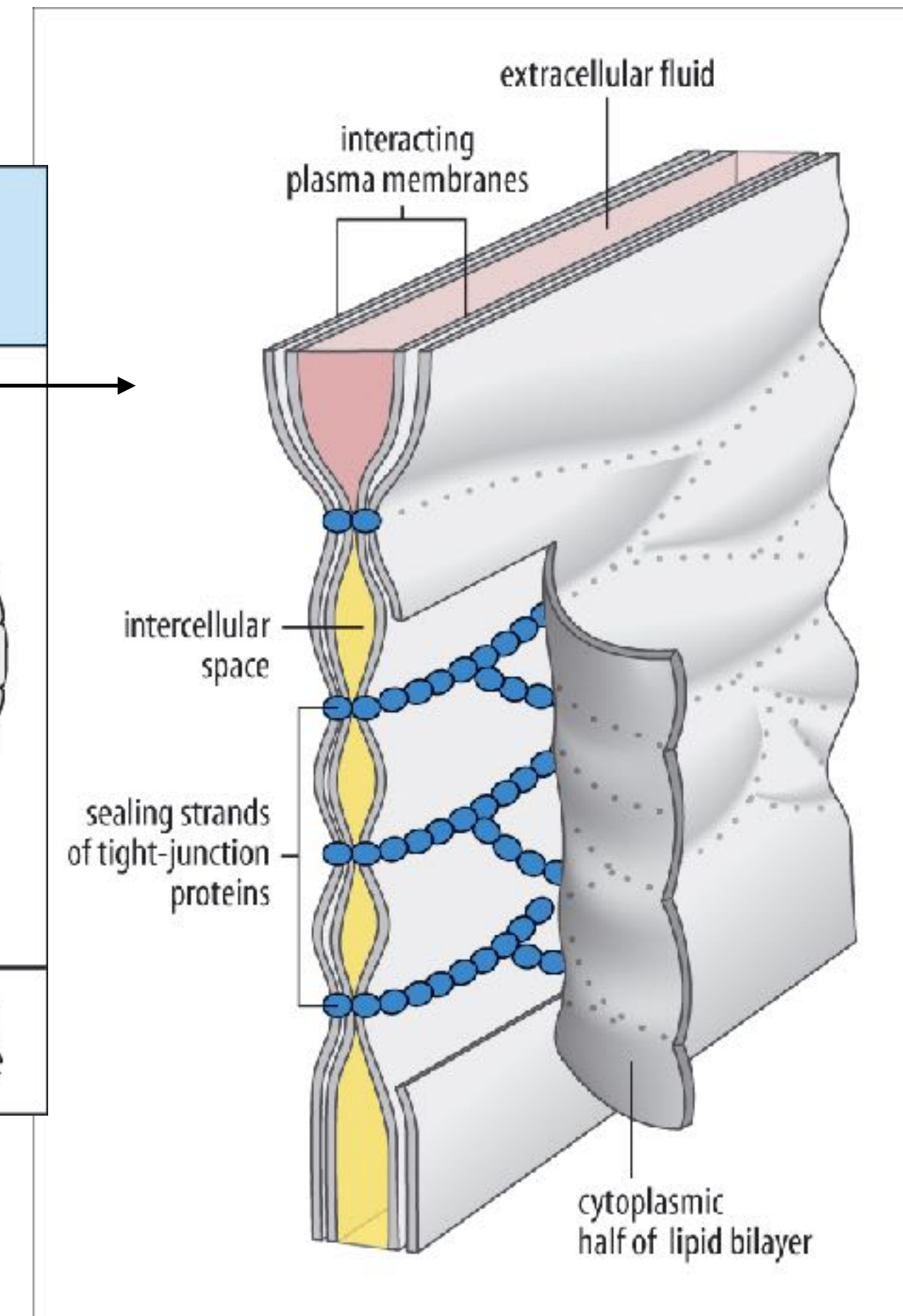
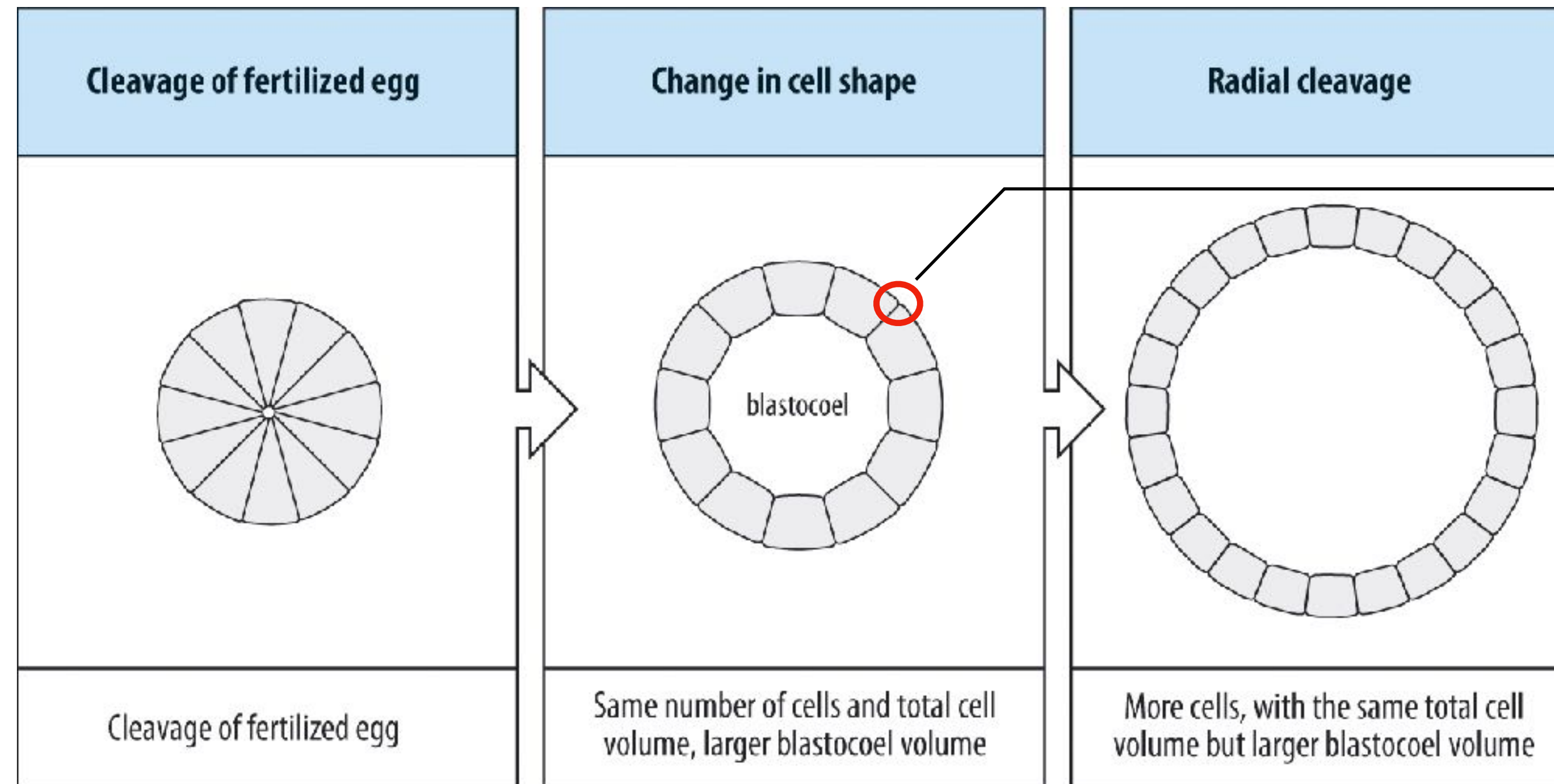
Reminder - Spindle position controls orientation of division



Signals for spindle positioning - interaction of cortex and spindle MT asters, +???

Version 1. Sea urchin

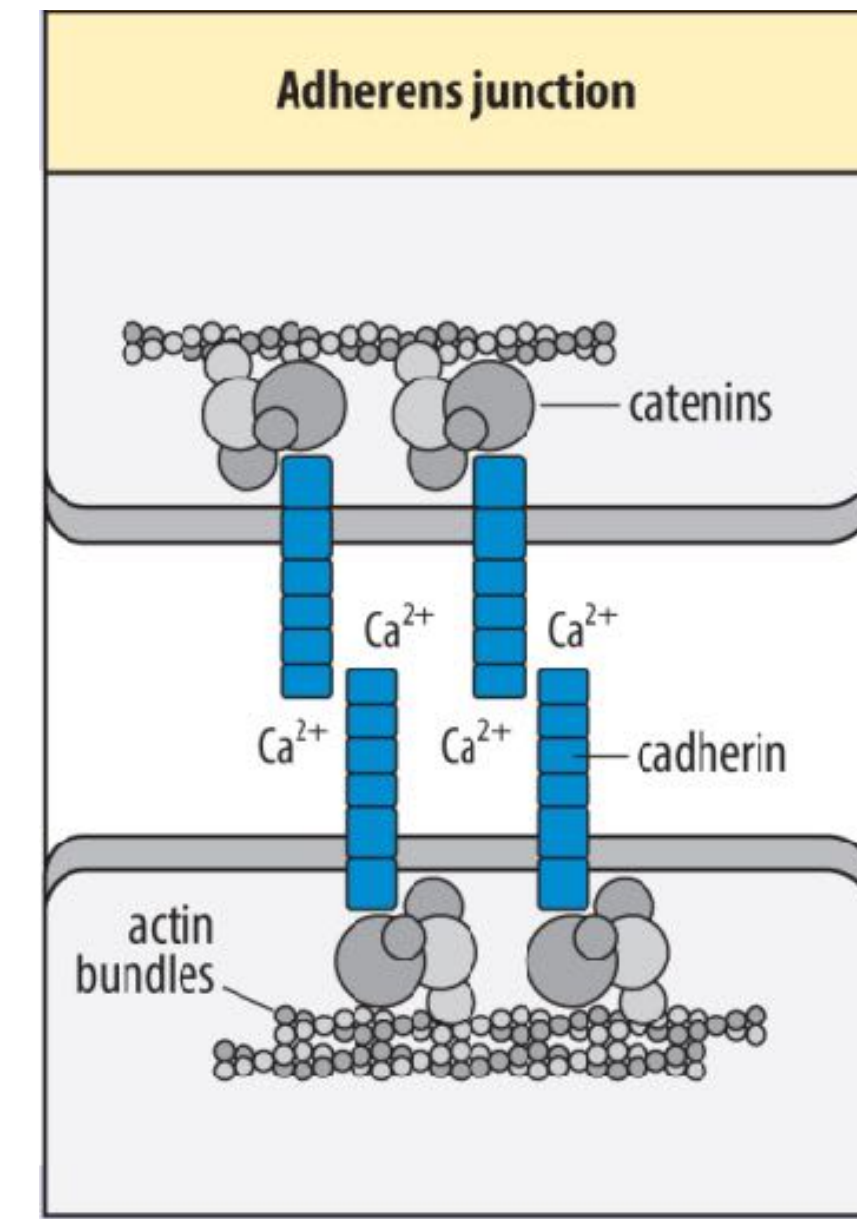
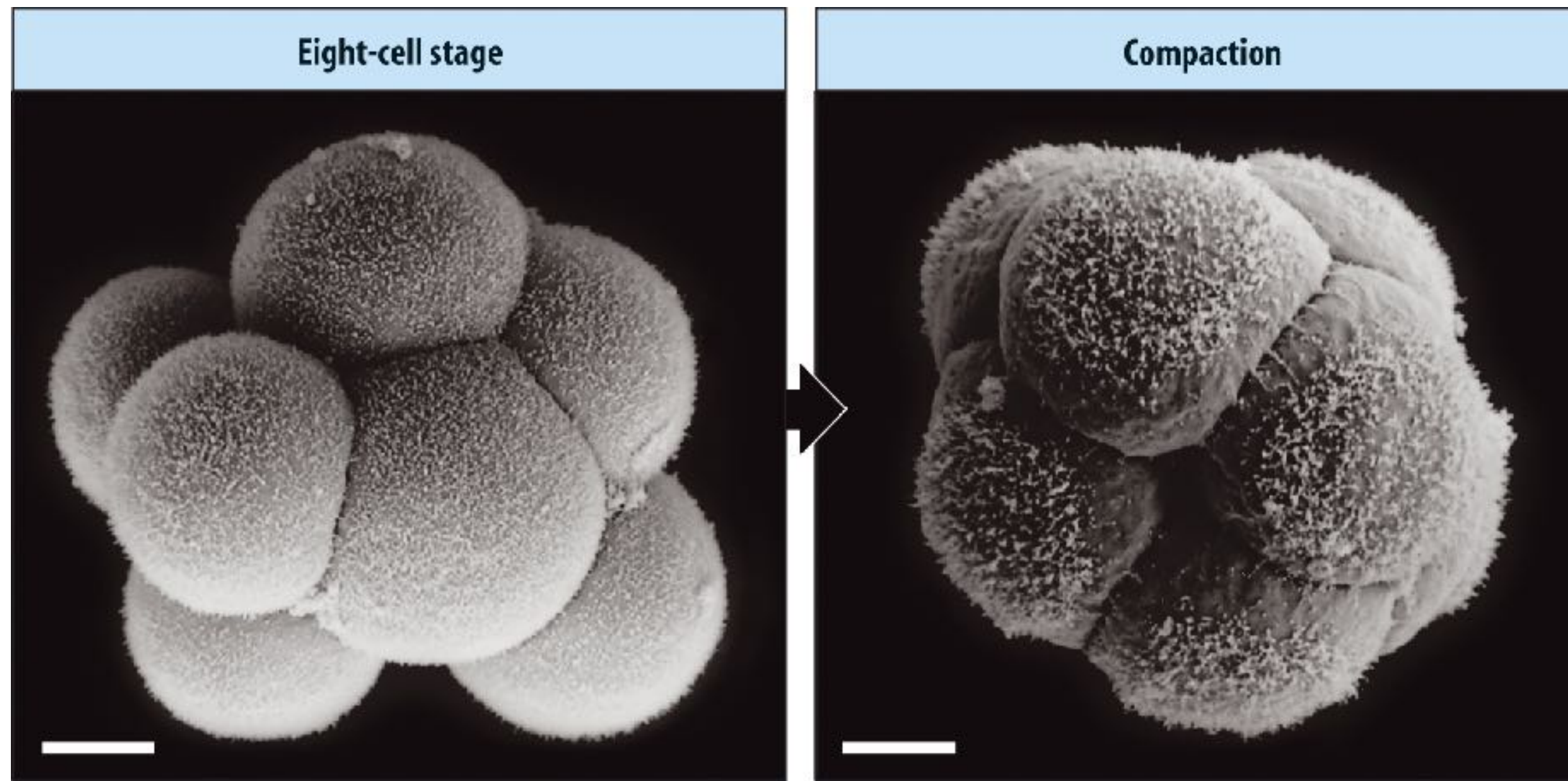
Septate junctions / Tight junctions



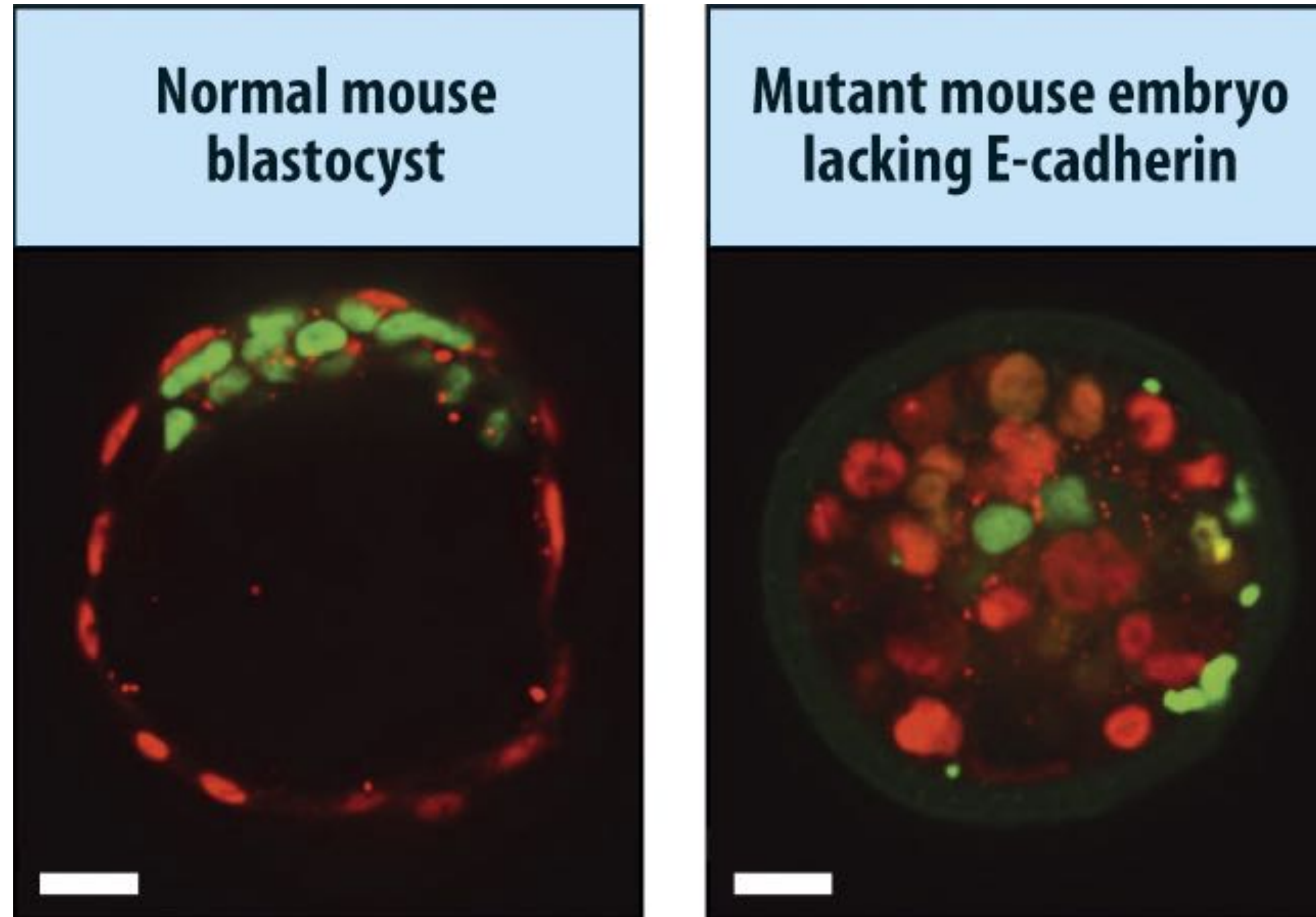
We will return to the sea urchin in part 2

membrane-spanning:
E-cadherin
+ Claudin, Occludin

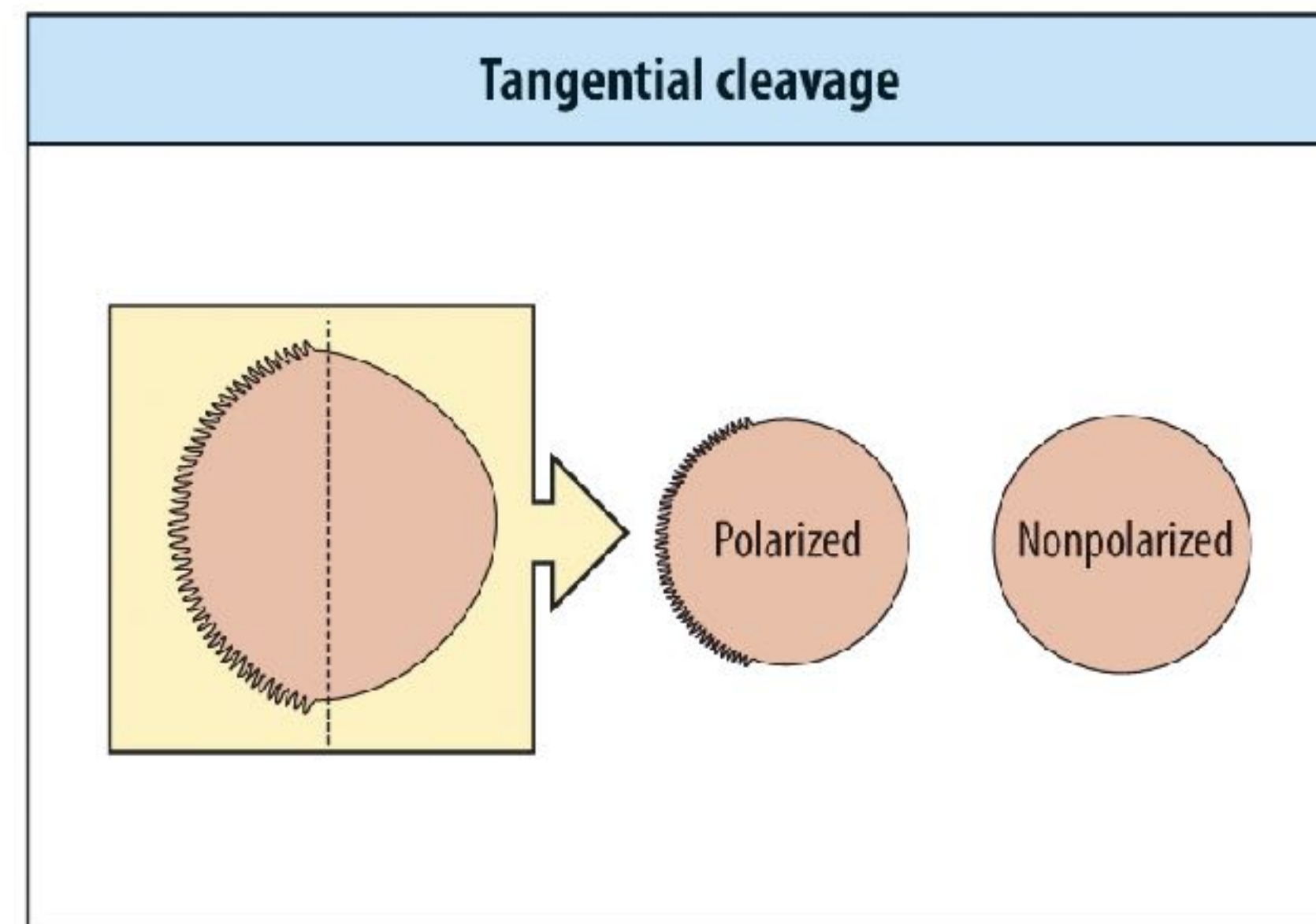
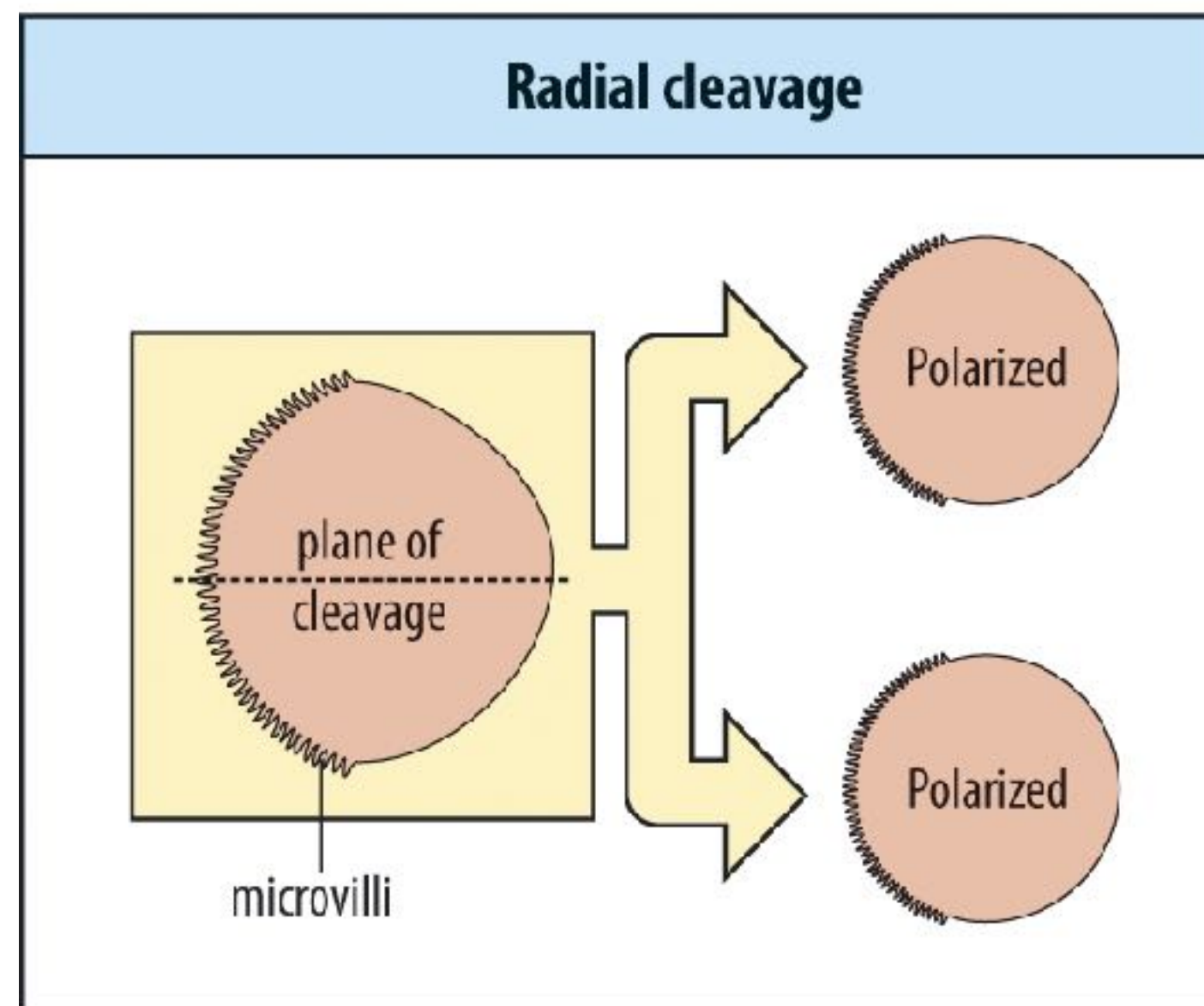
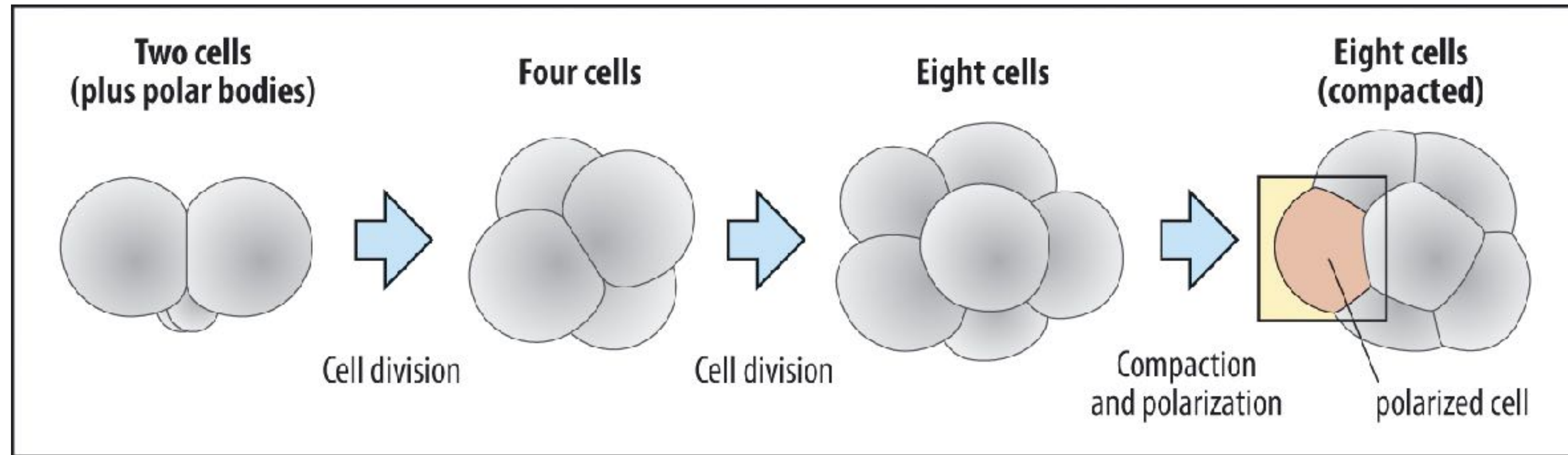
Version 2. Mouse



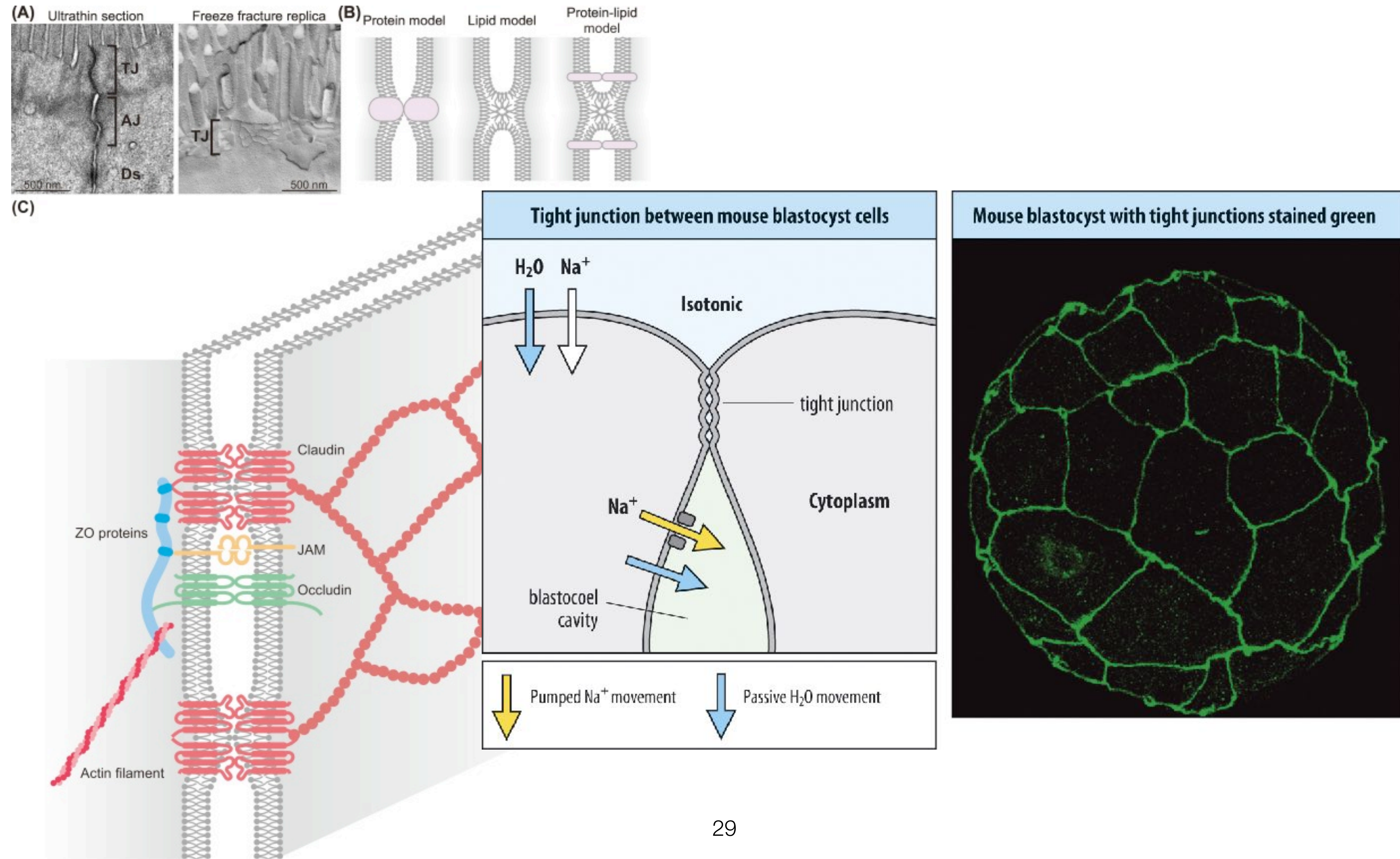
E-cadherin required for compaction and blastula formation



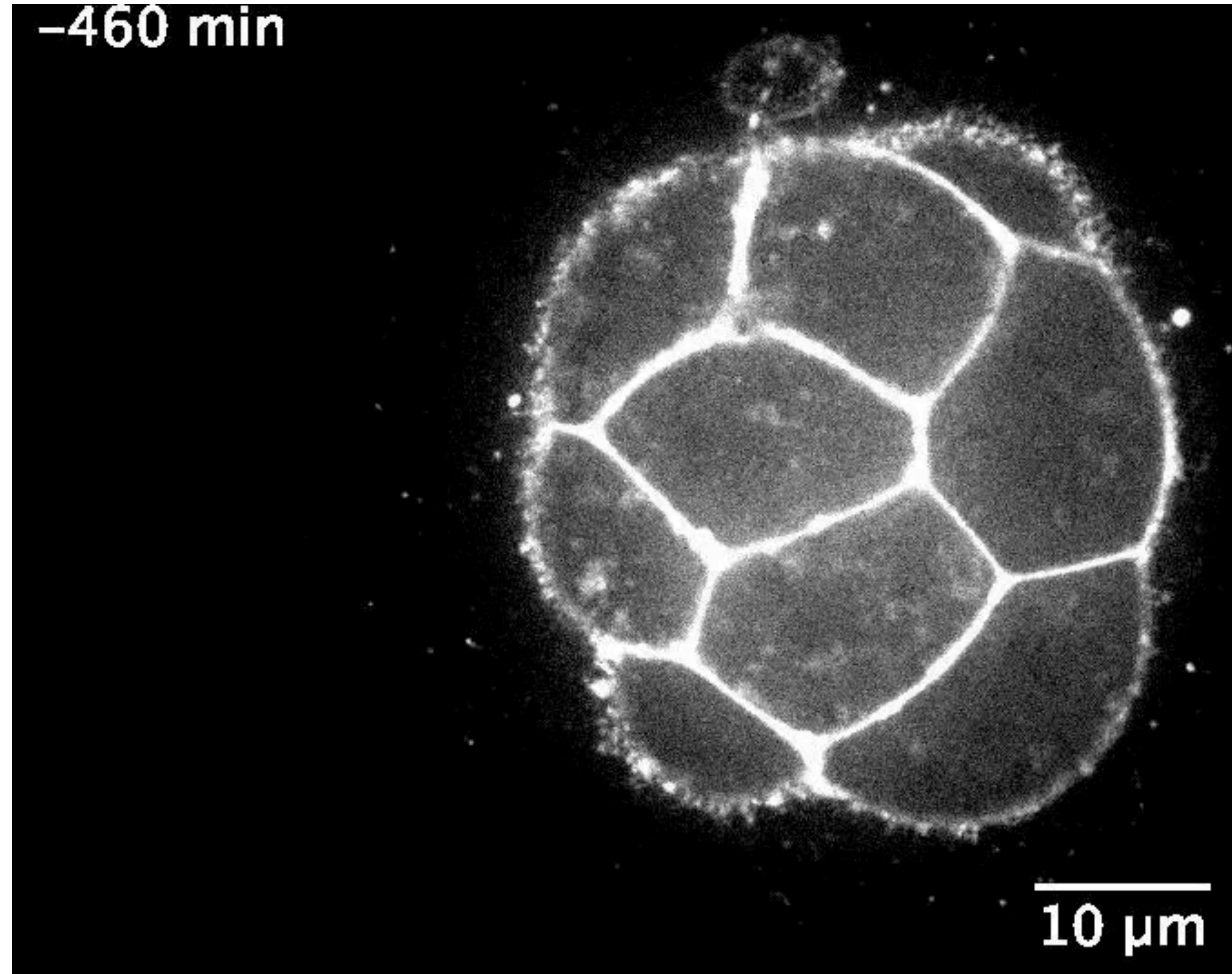
Consequence of changing cleavage orientation



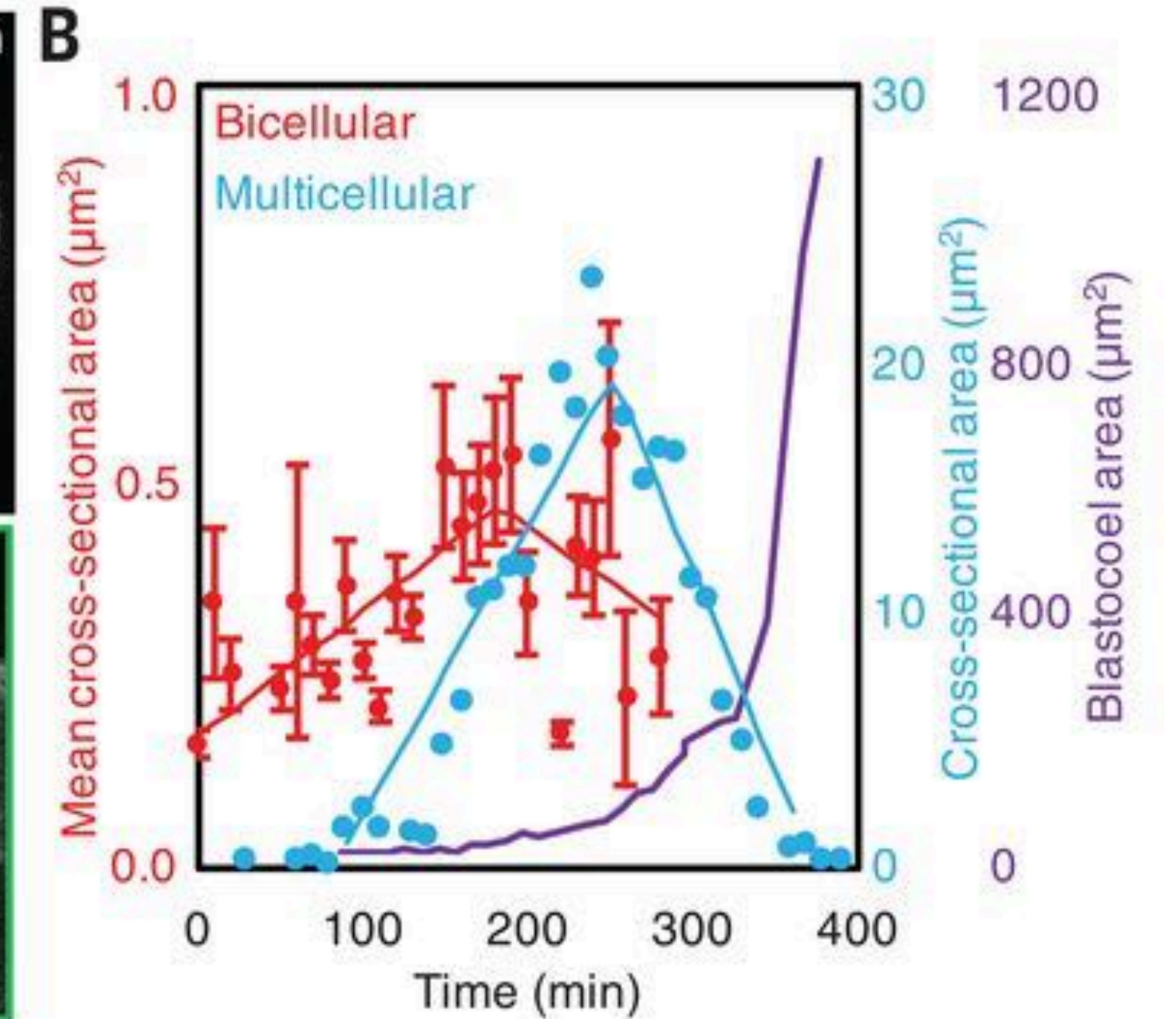
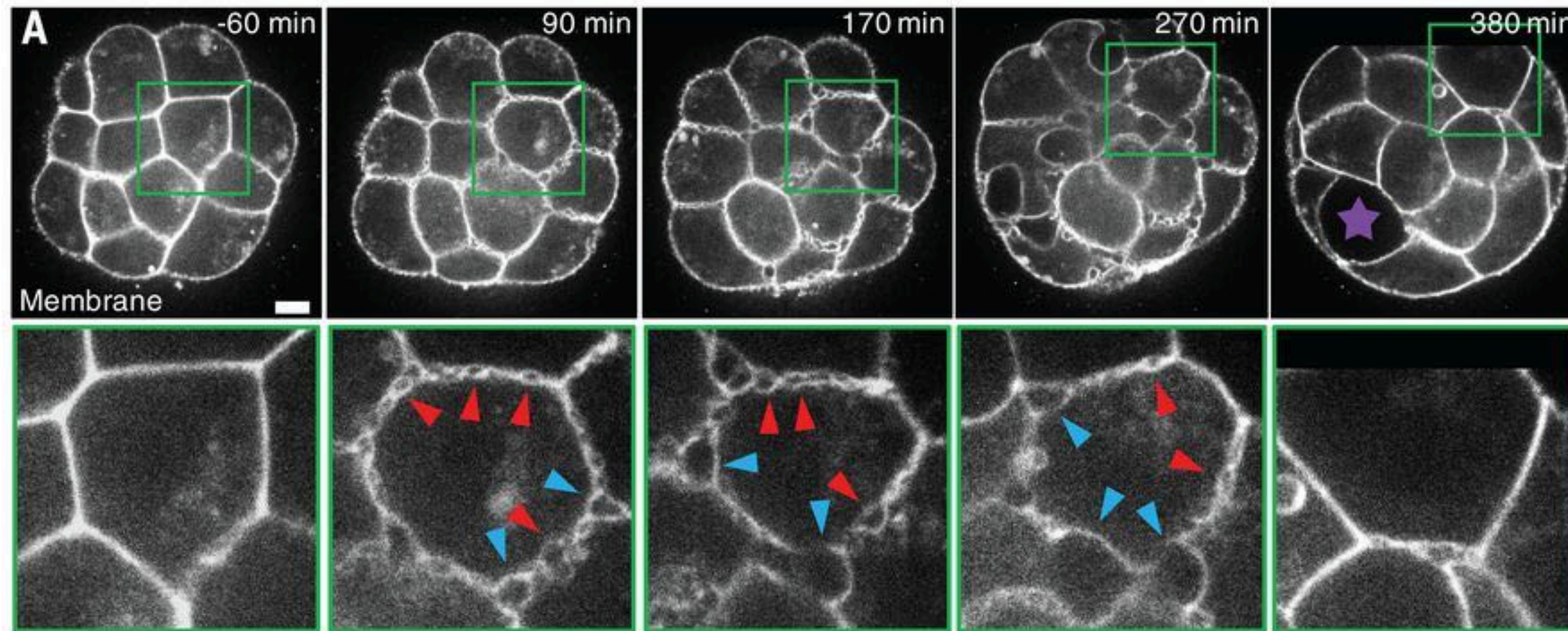
Tight junctions and Na⁺ pumps swell blastocoel



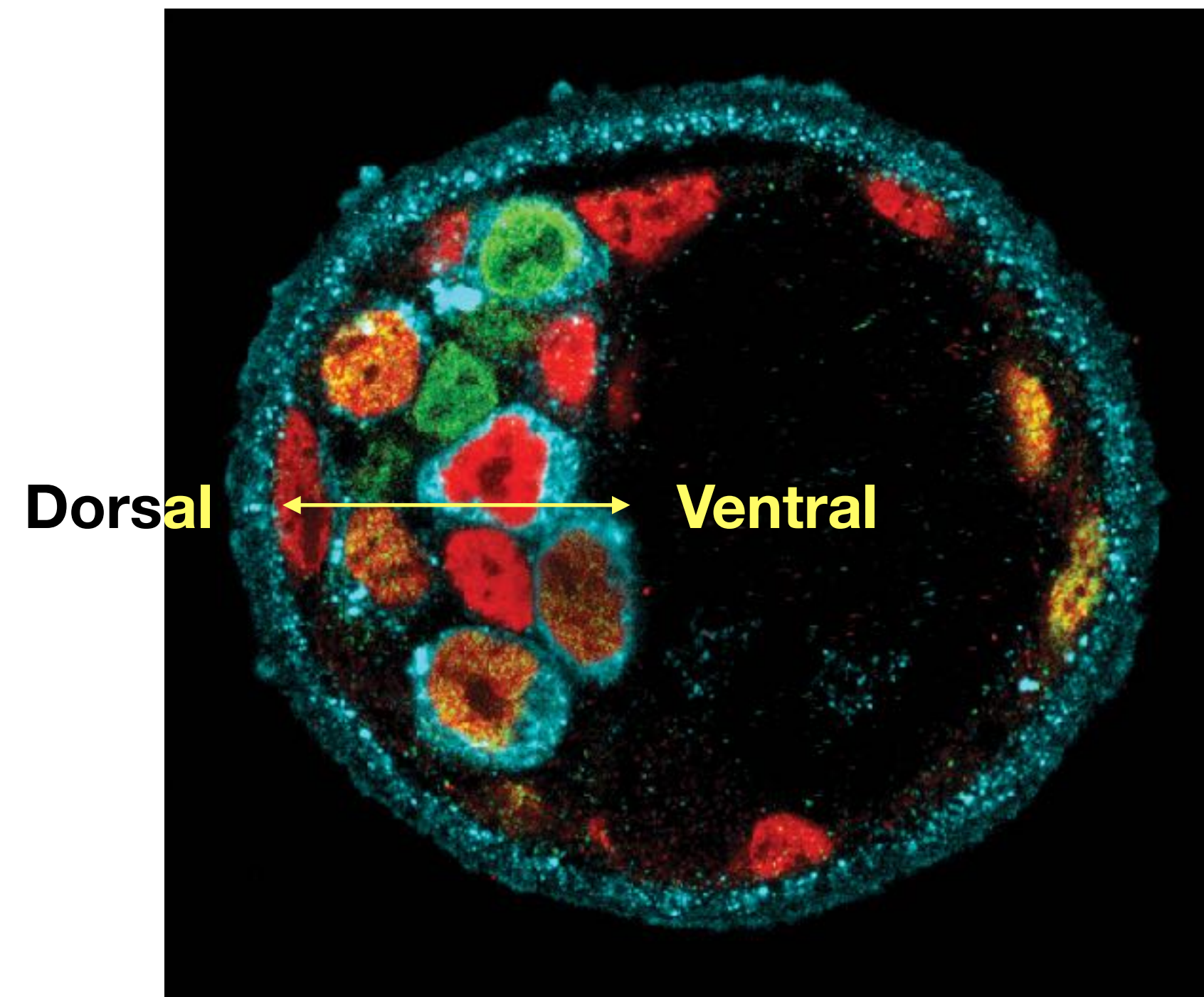
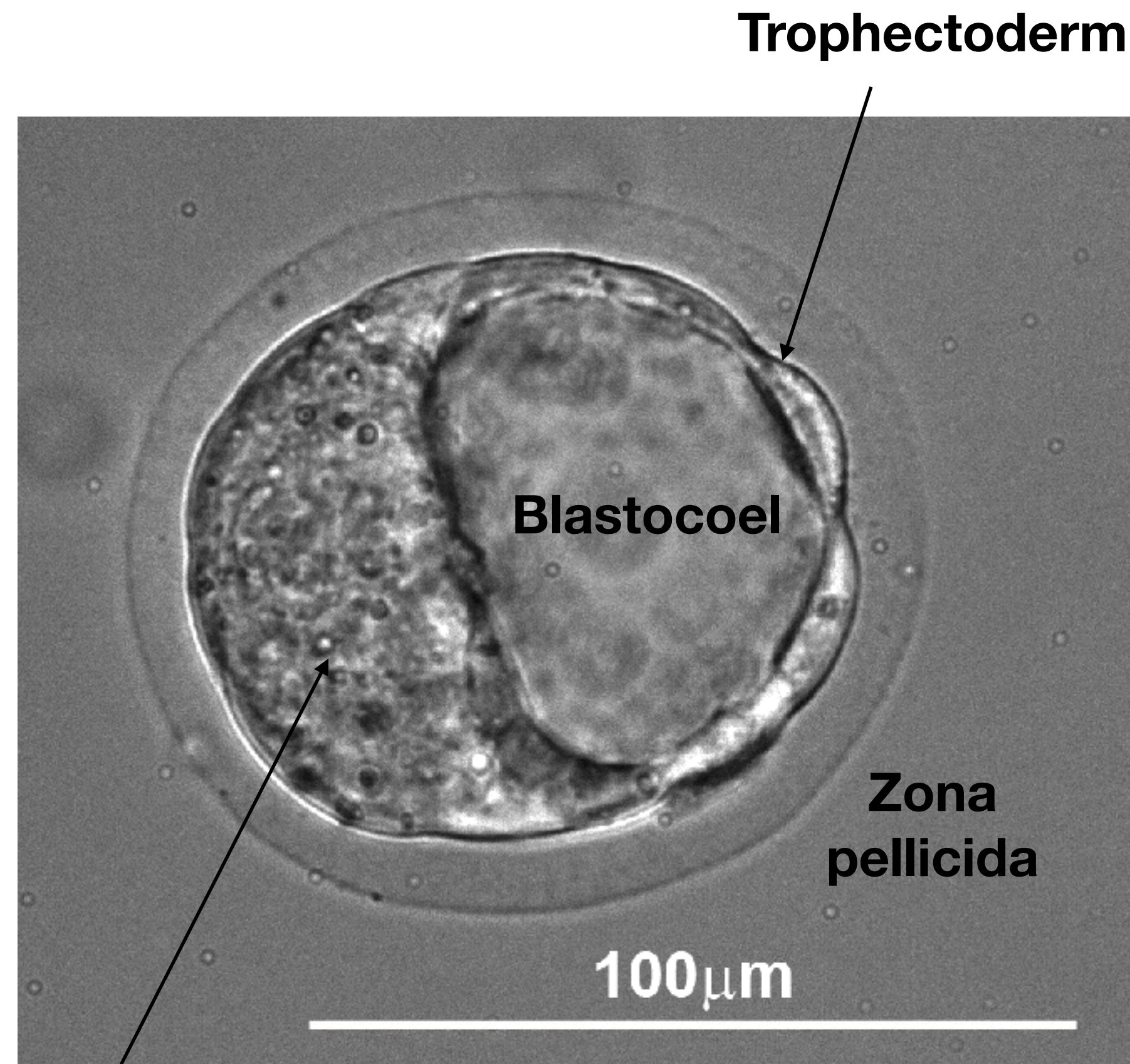
Blastocoel emerges by fracturing and ripening



Blastocoel emerges by fracturing and ripening



Generation of inner and outer cell layers (and an embryonic axis... D-V)

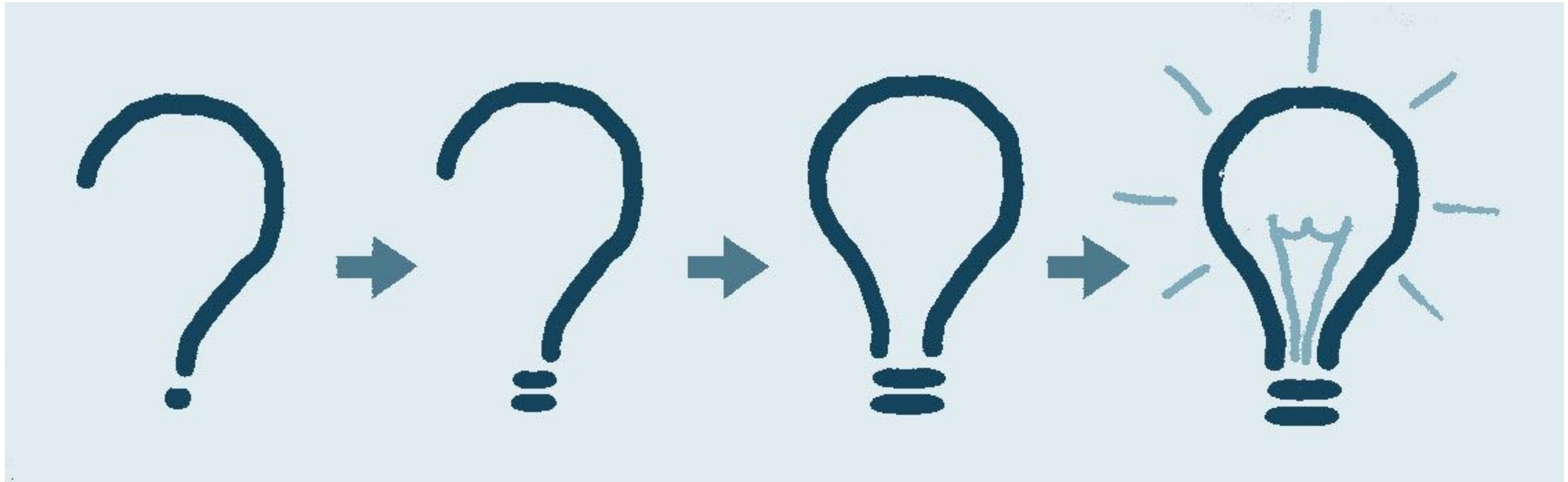


Inner cell mass (ICM)

Summary

- **Morphogenesis**
- Cell adhesion - Cadherins
- Cell division - orientation
- Actin and myosin provide tissue-level forces through surface complexes
- Oriented cleavage and blastula in sea urchin and mouse

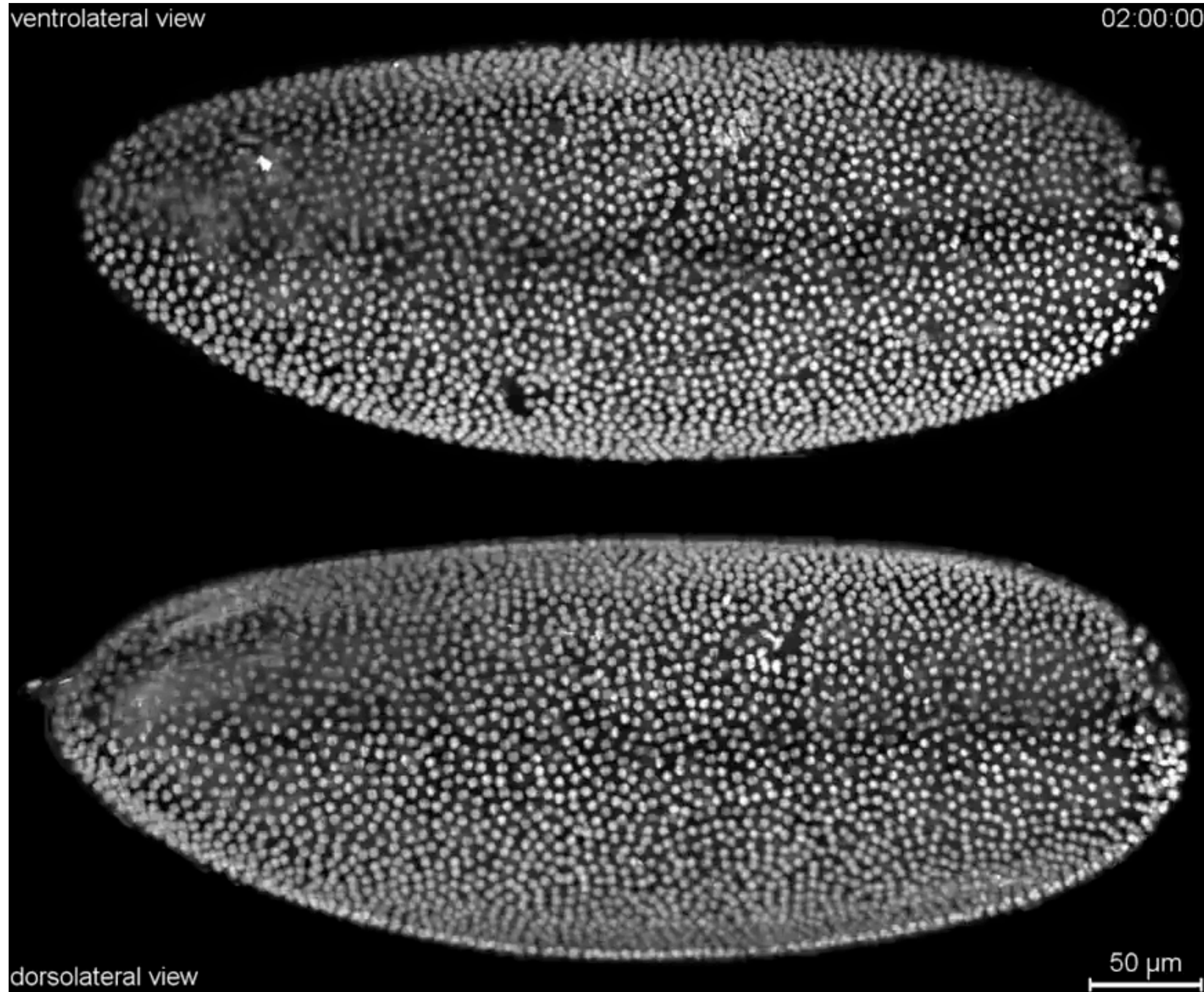
Questions?



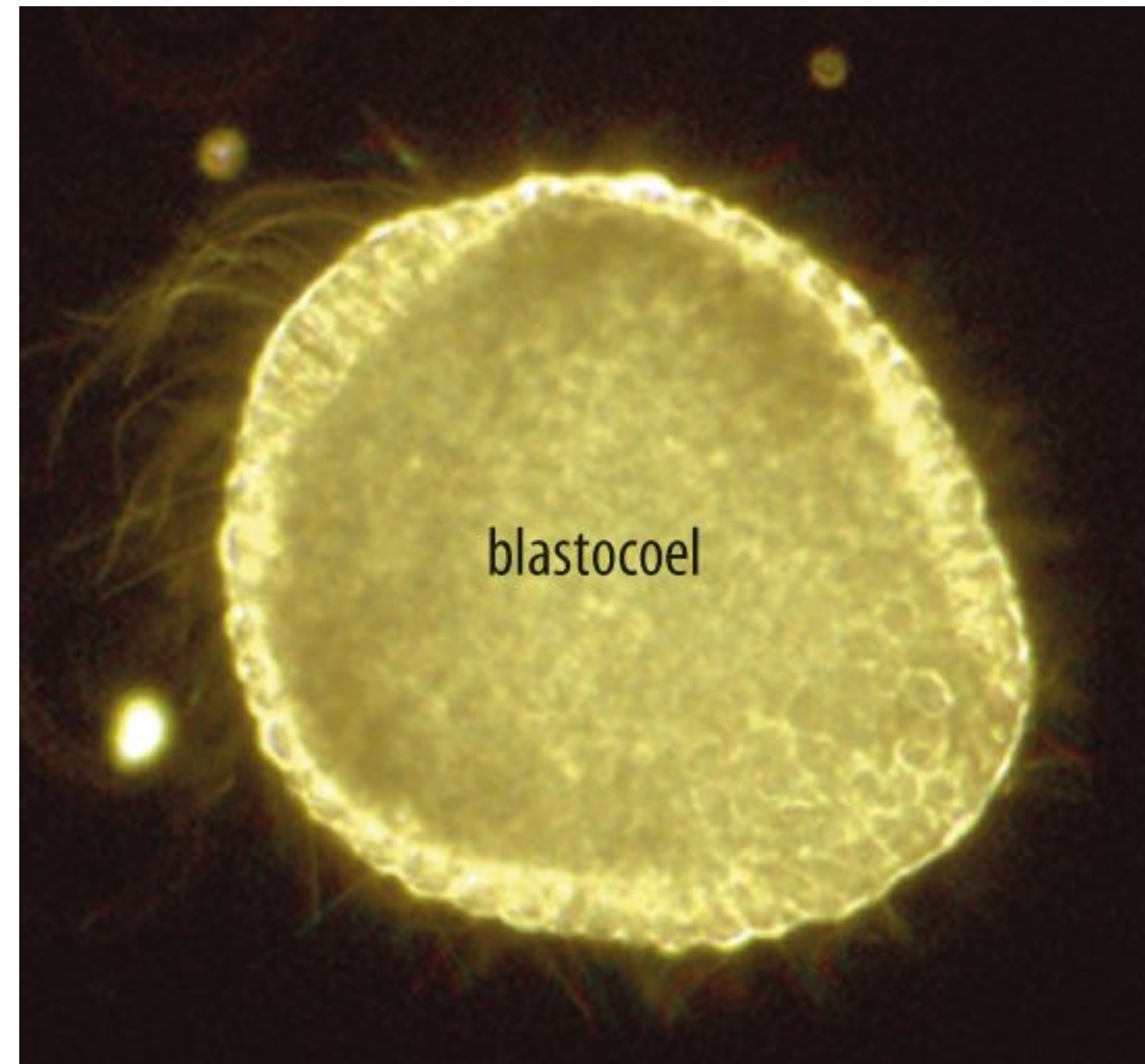
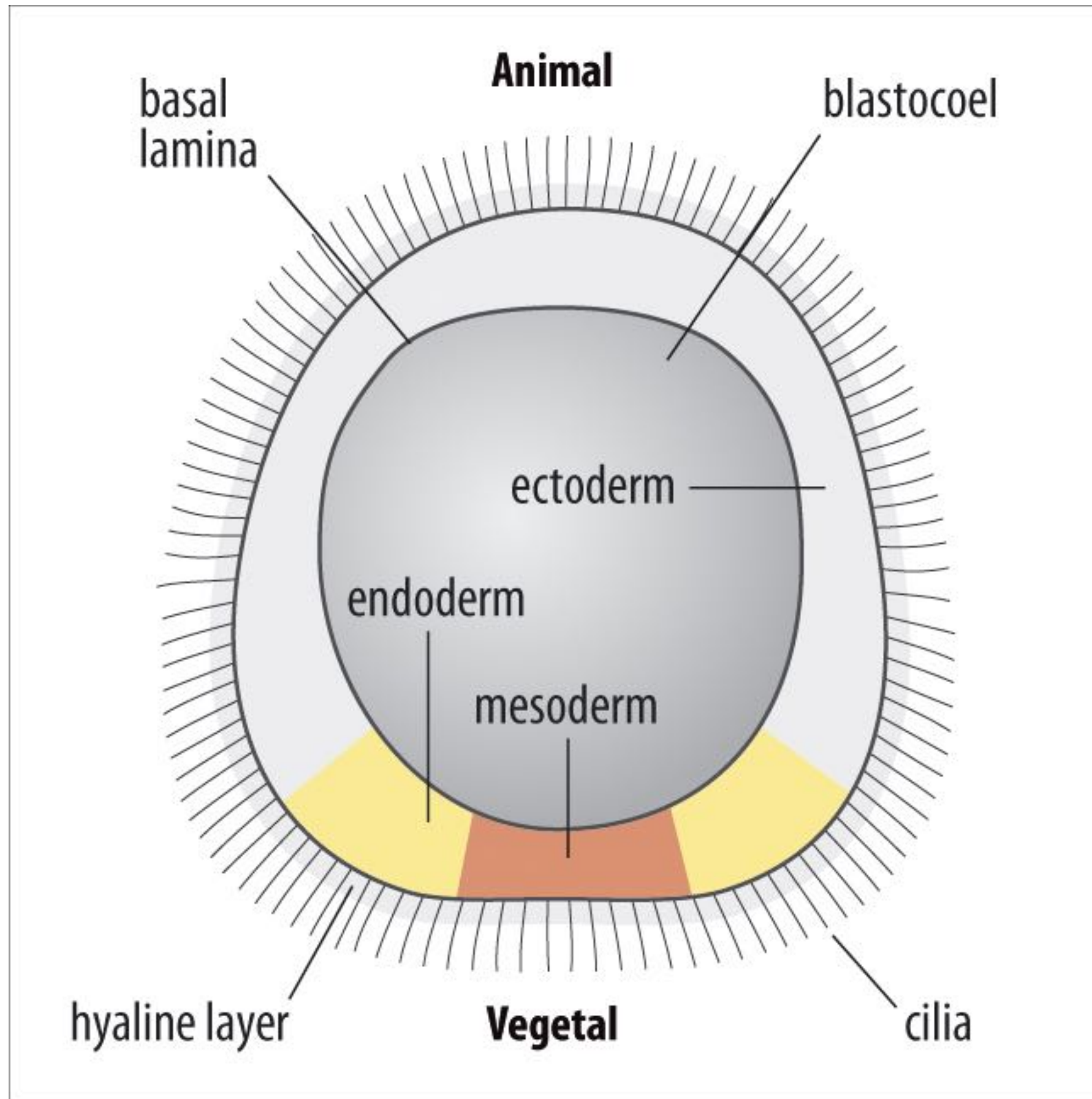
Take a break



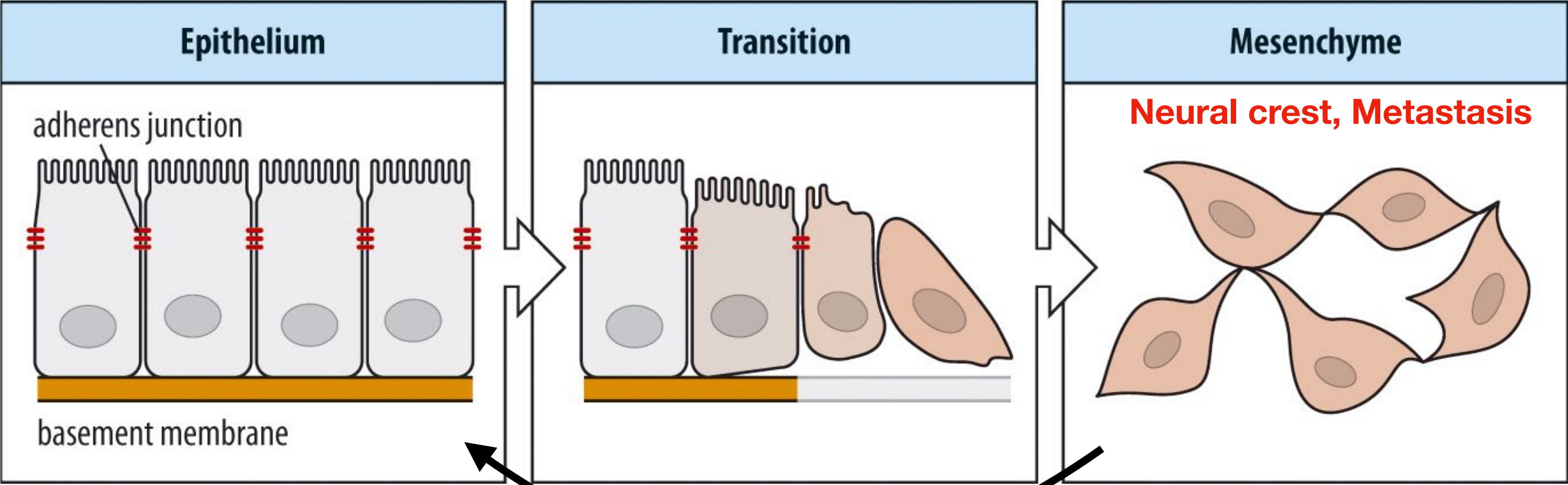
Big questions



Sea urchin blastula

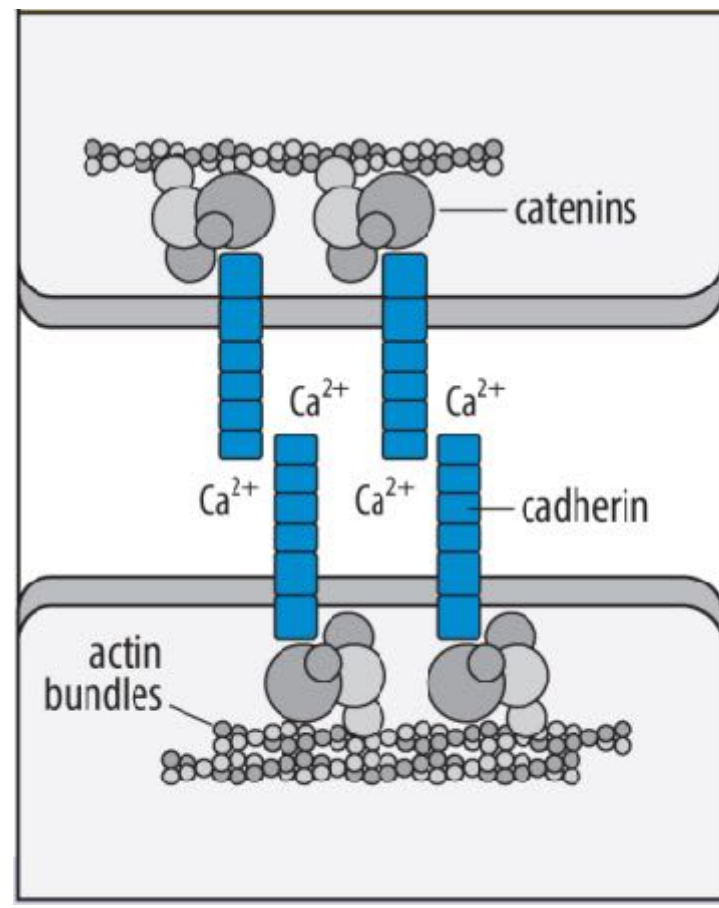


Epithelial - Mesenchymal Transition (EMT)

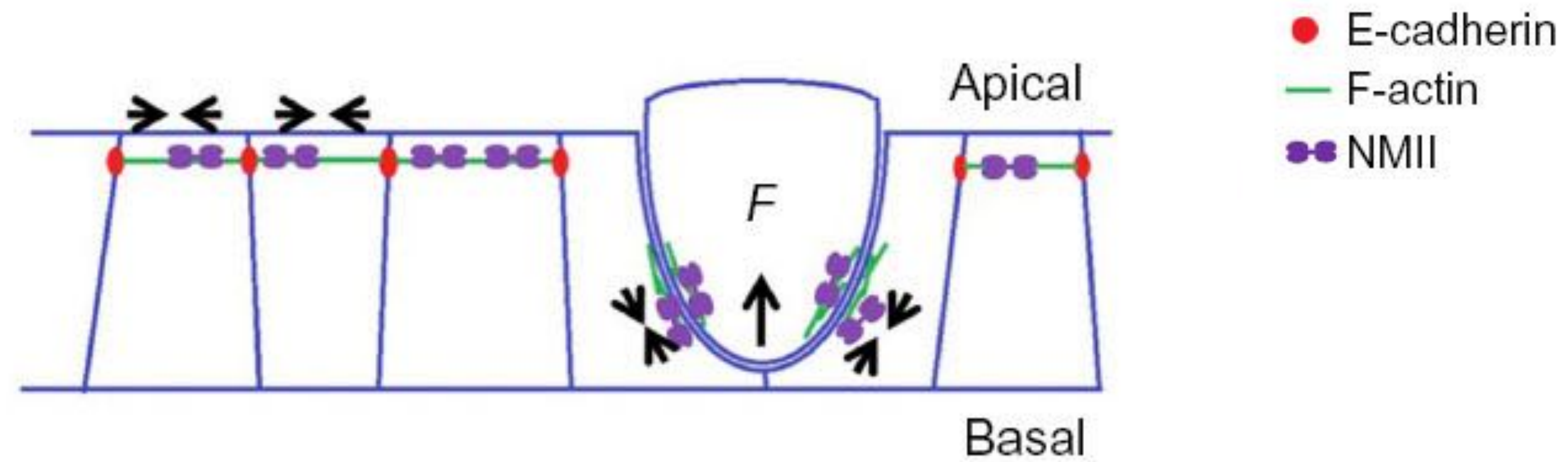


somites,
blood vessels,
kidney tubules

Snail ↑
Cadherins, catenins ↓



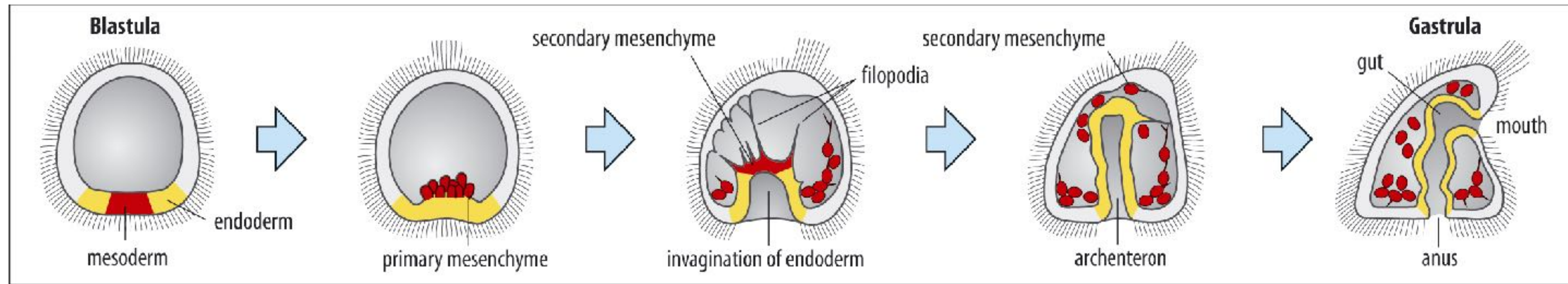
Delamination - cells leave an intact epithelium



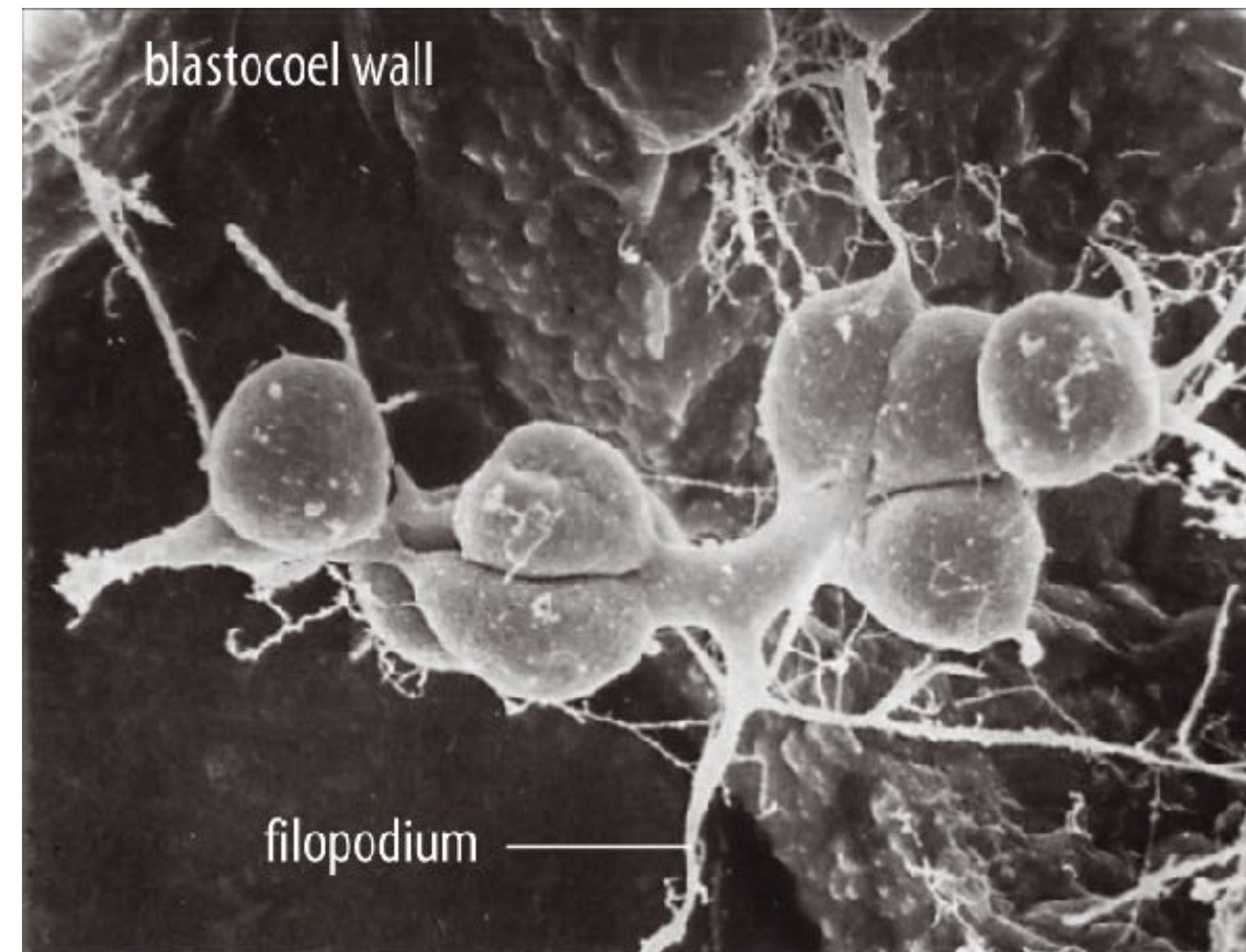
Seen also during neurogenesis (week 8 fine-grain patterns)

Sea-urchin gastrulation

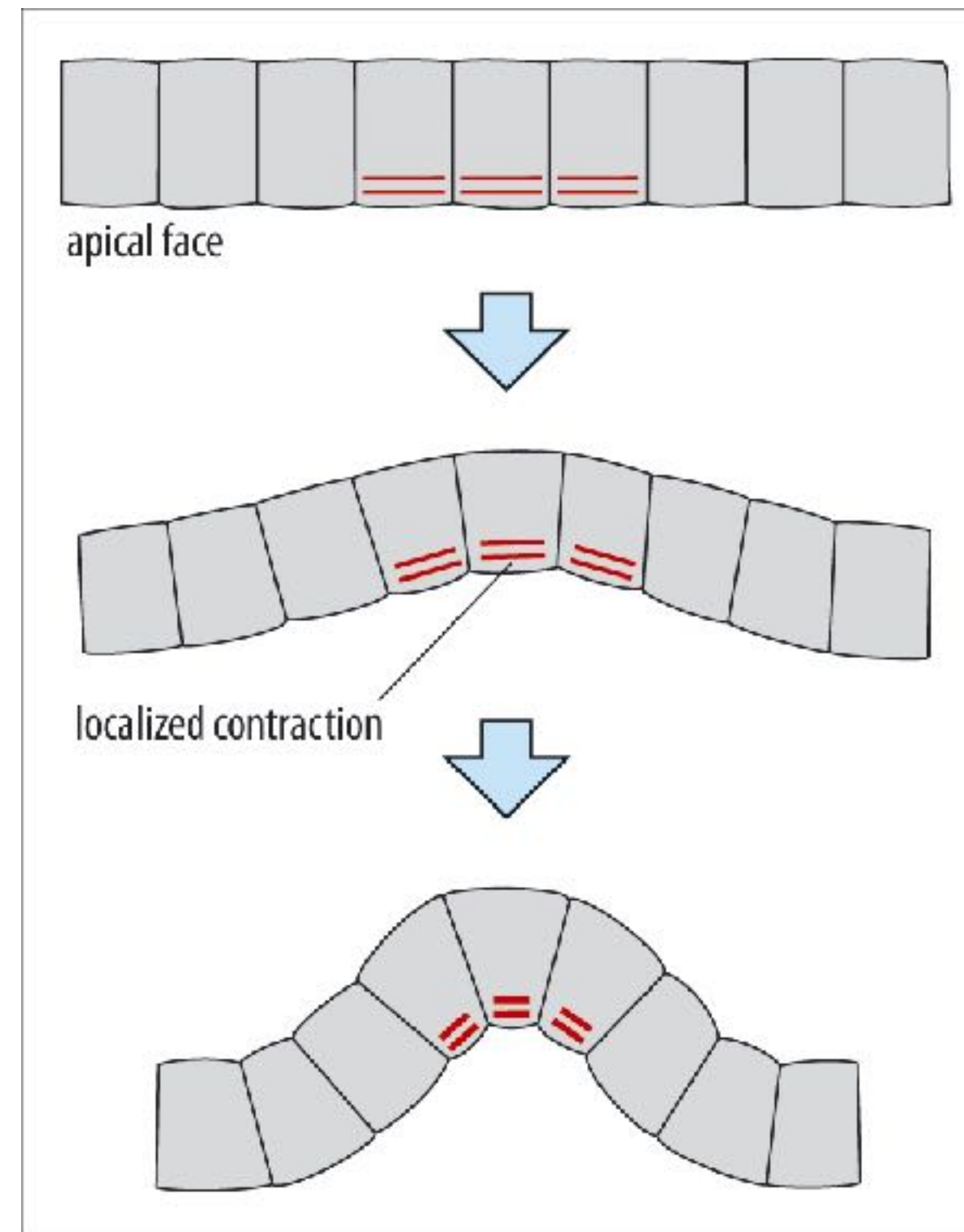
EMT / delamination, then migration and invagination



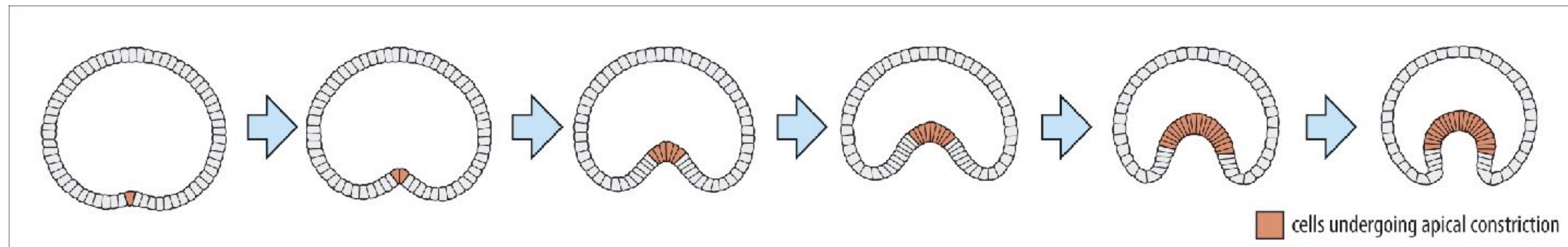
Apical constriction drives invagination Secondary mesenchyme EMT



Apical constriction drives invagination



Actomyosin contractility



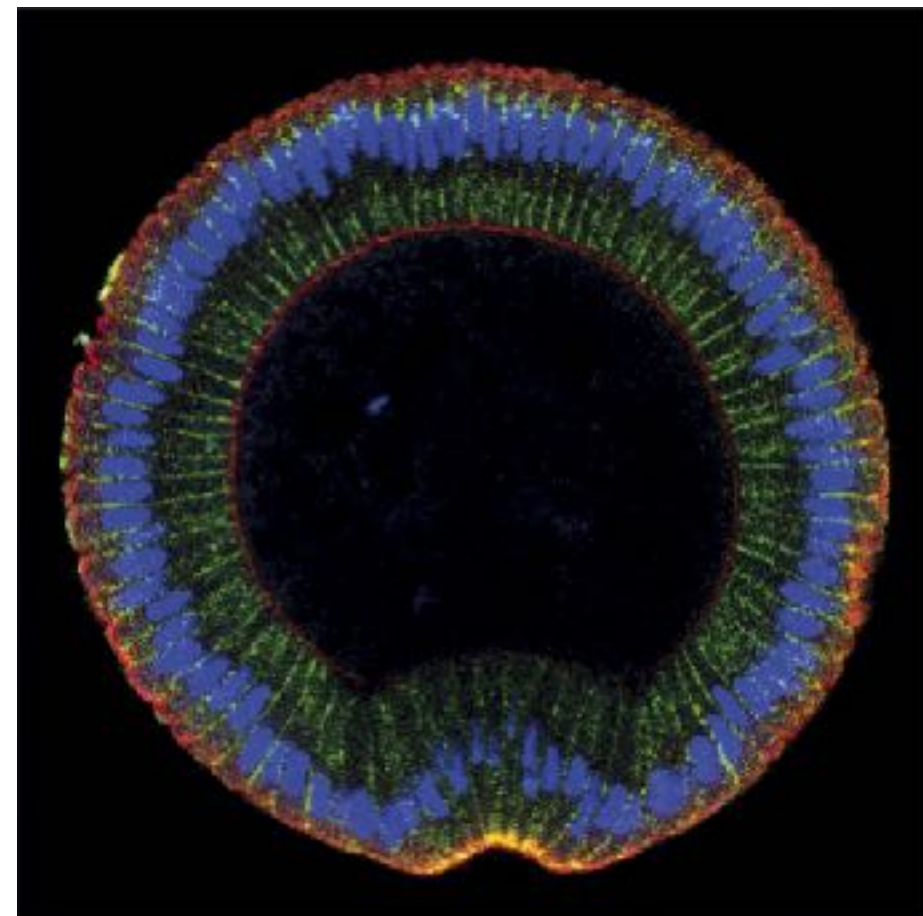
Drosophila gastrulation

Invagination, *then* EMT and migration

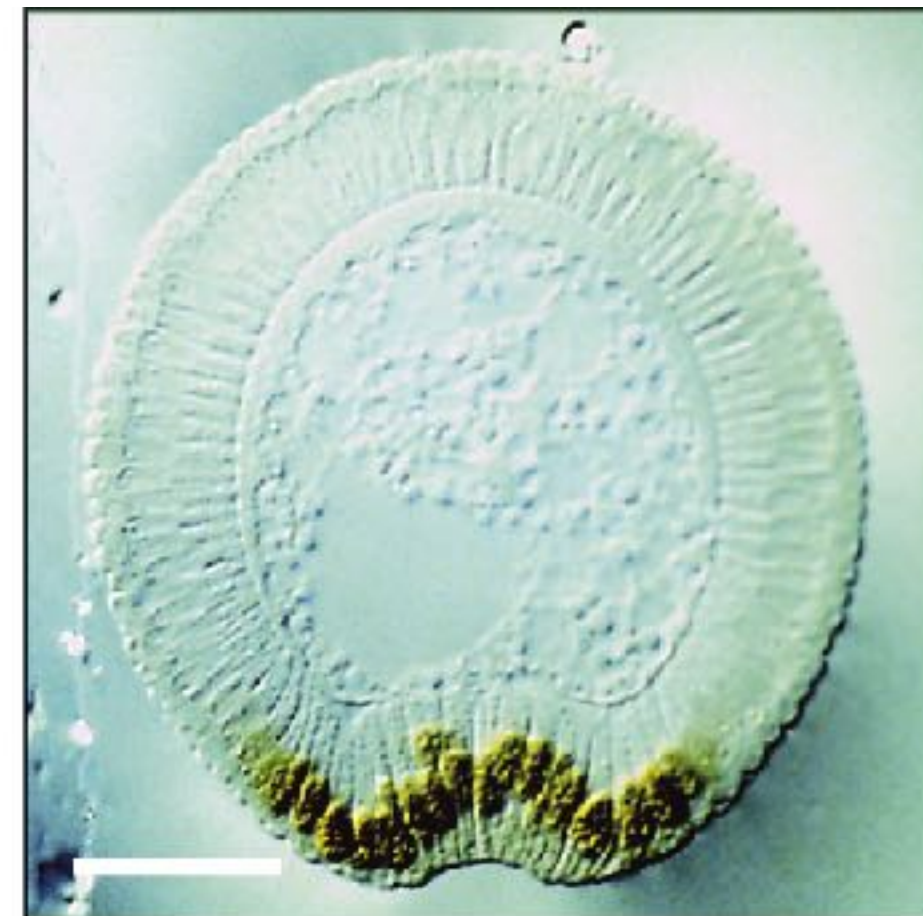
Anterior



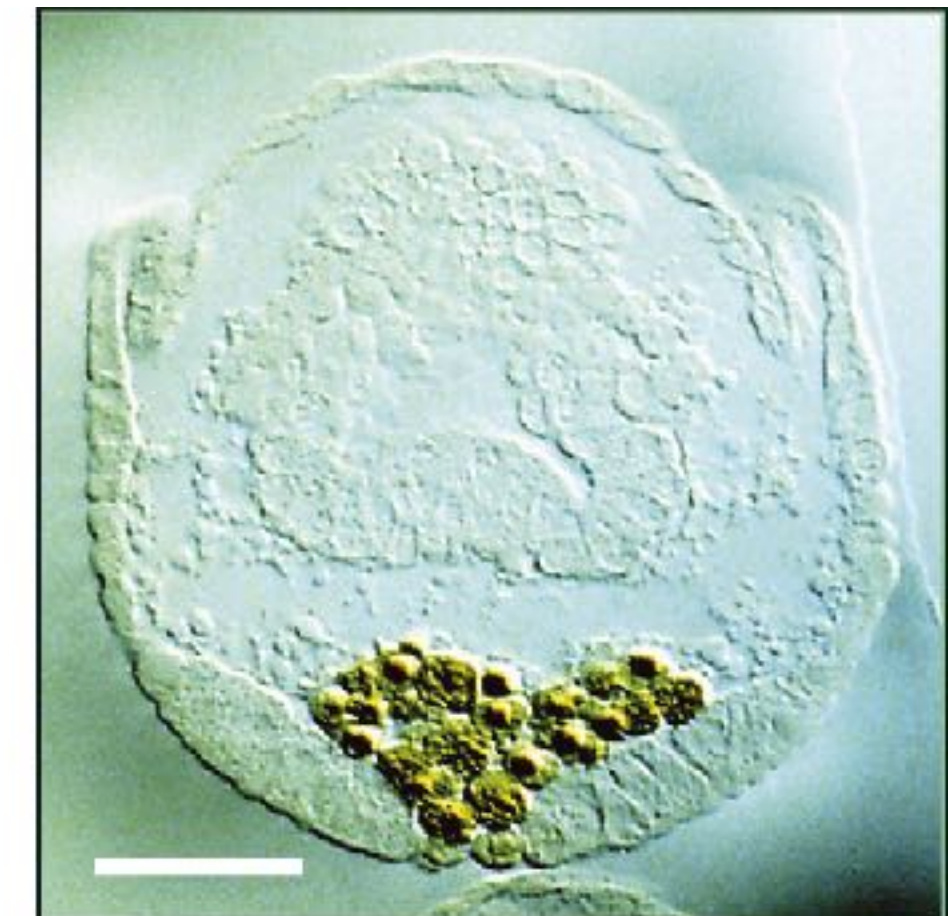
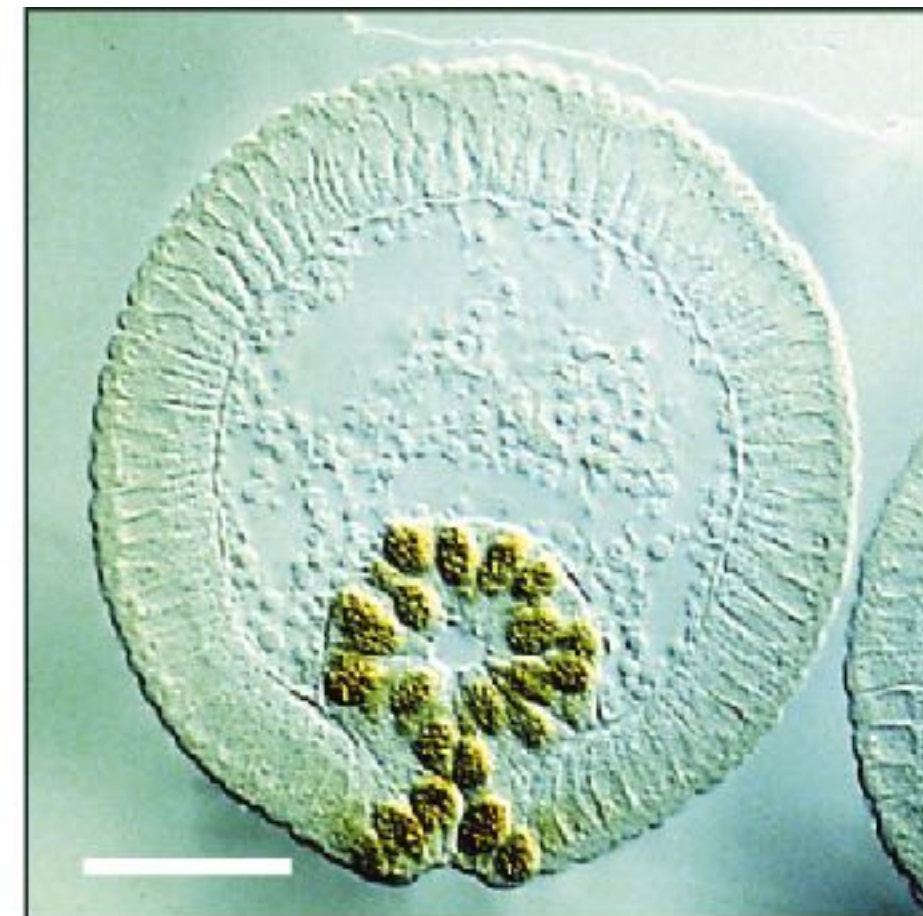
Posterior



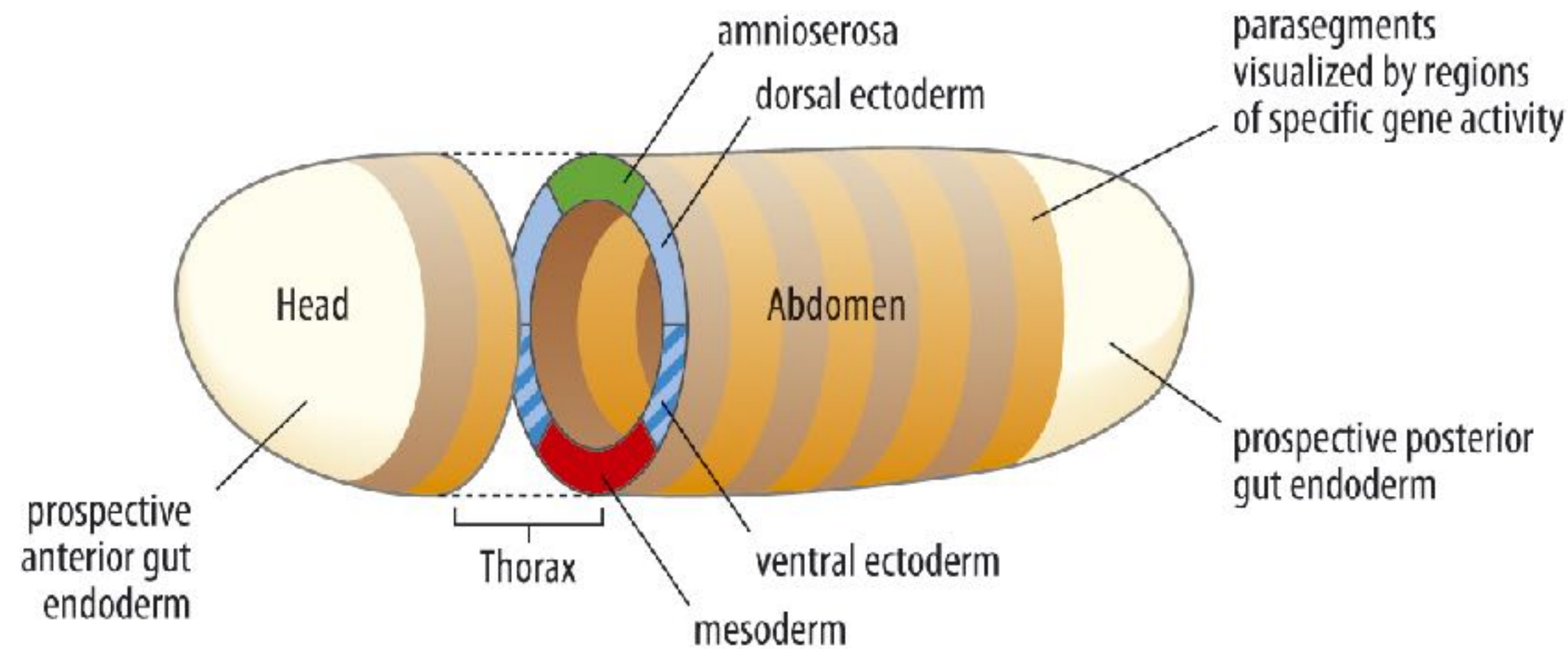
Myosin II



Twist



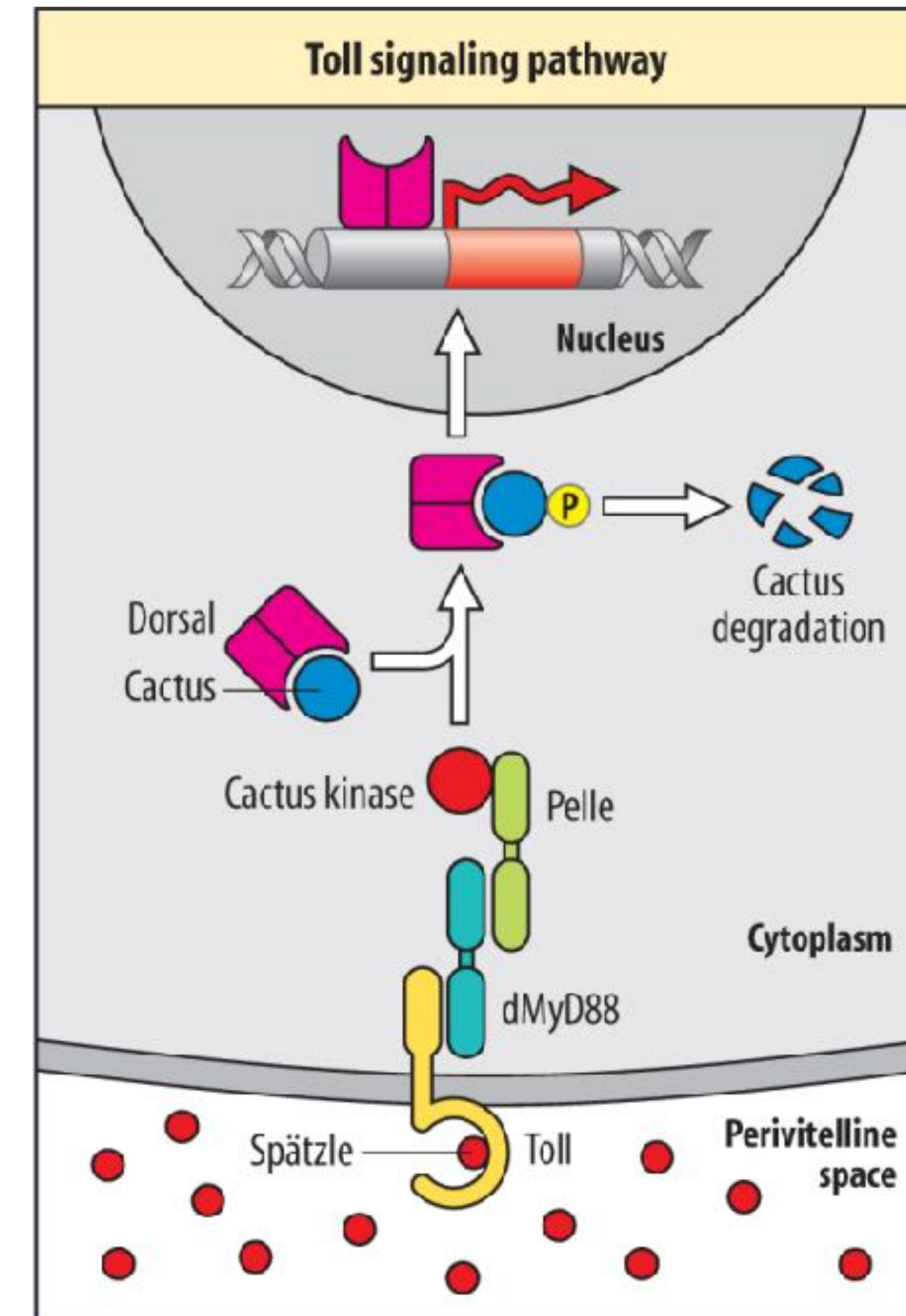
Genetic control of *Drosophila* gastrulation



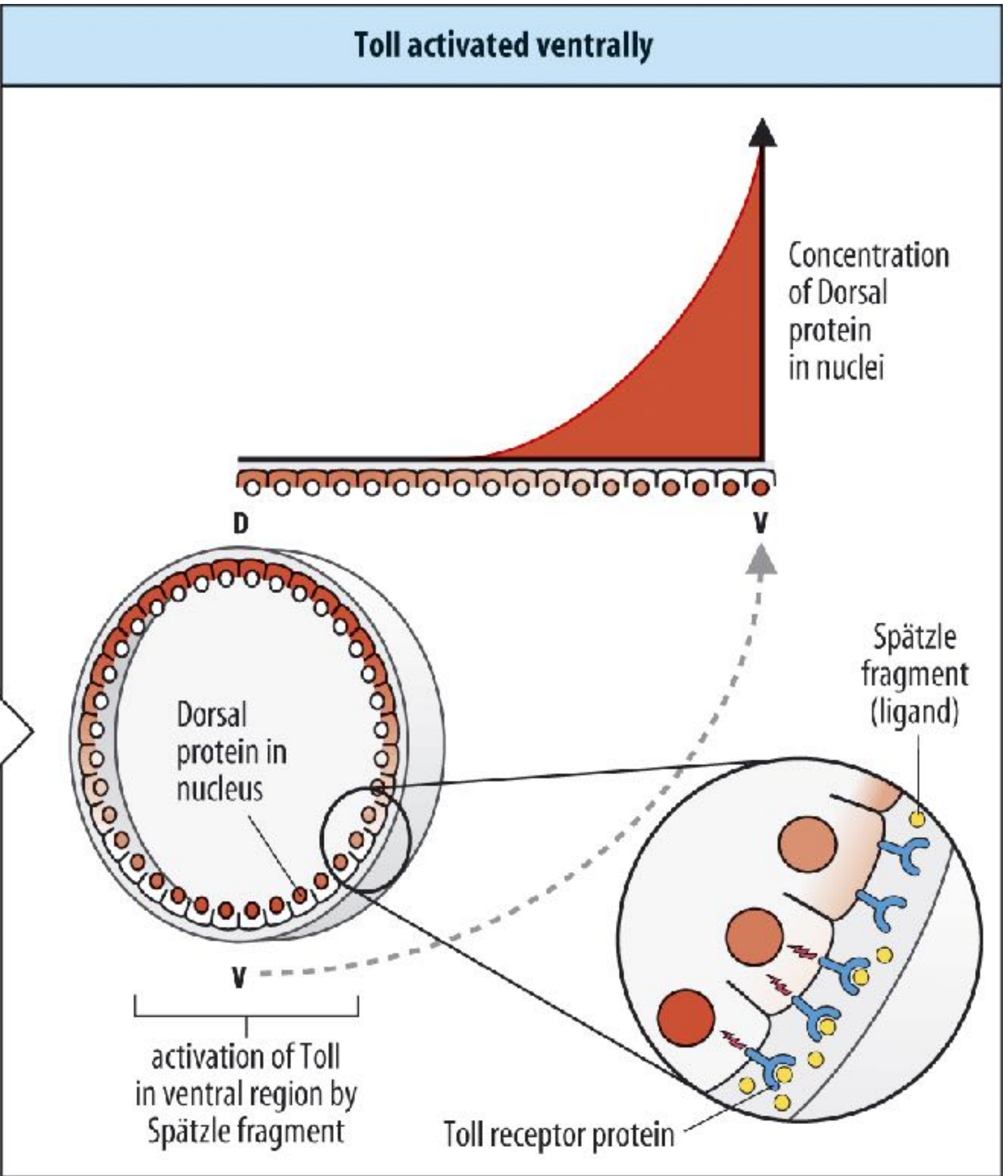
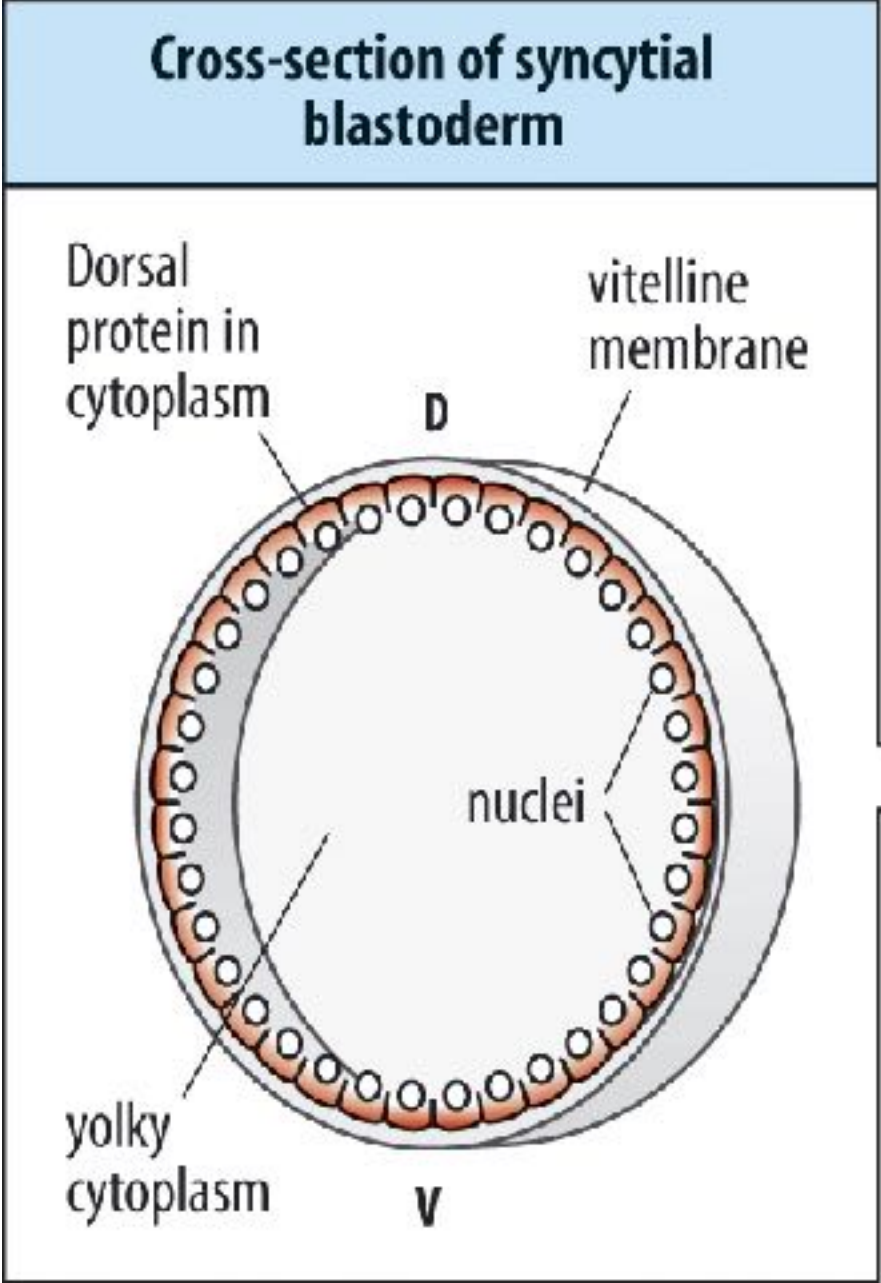
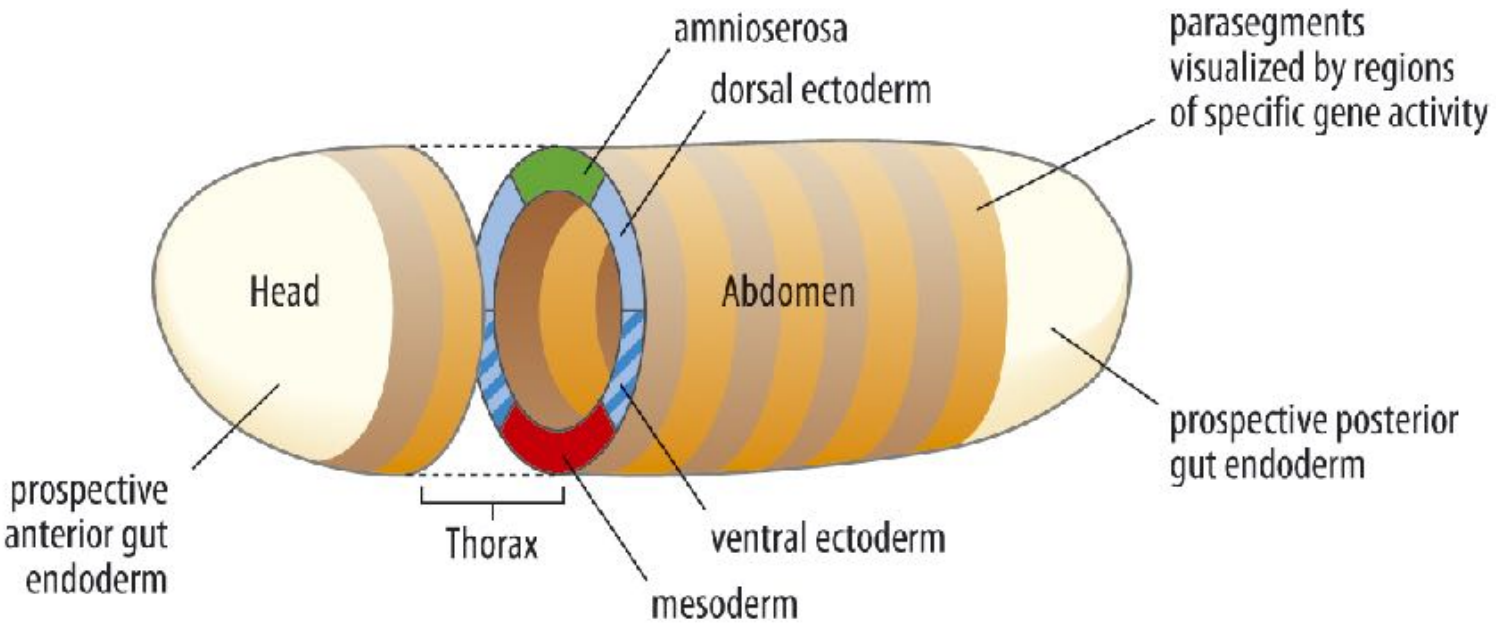
Toll receptor = human innate immunity receptor

Nobel prize in Physiology and Medicine 2011
Bruce A. Beutler & Jules A. Hoffmann

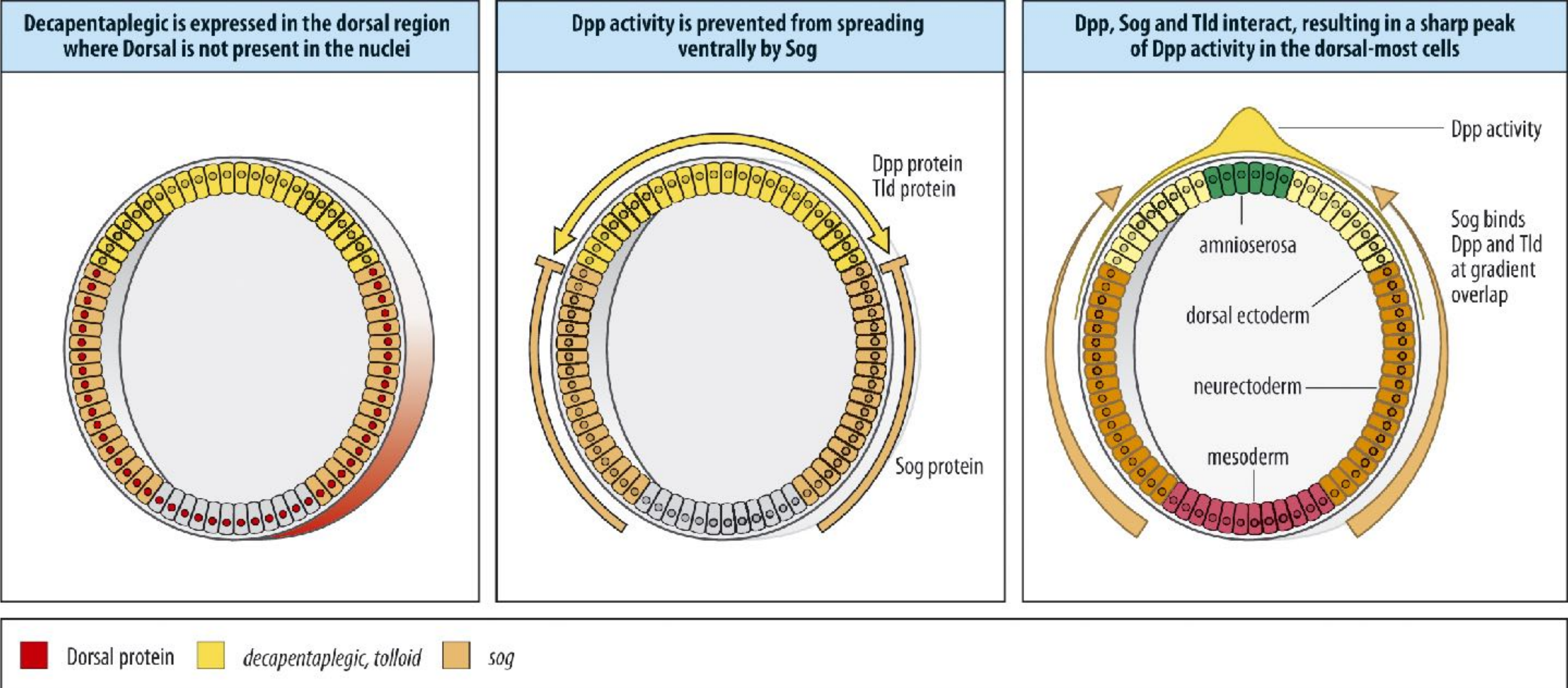
Lemaitre B, Nicolas E, Michaut L, Reichhart JM, **Hoffmann JA**.
The dorsoventral regulatory gene cassette *spätzle/Toll/cactus* controls
the potent antifungal response in *Drosophila* adults.
Cell 1996;86:973-983.



Genetic control of *Drosophila* gastrulation



Genetic control of *Drosophila* gastrulation

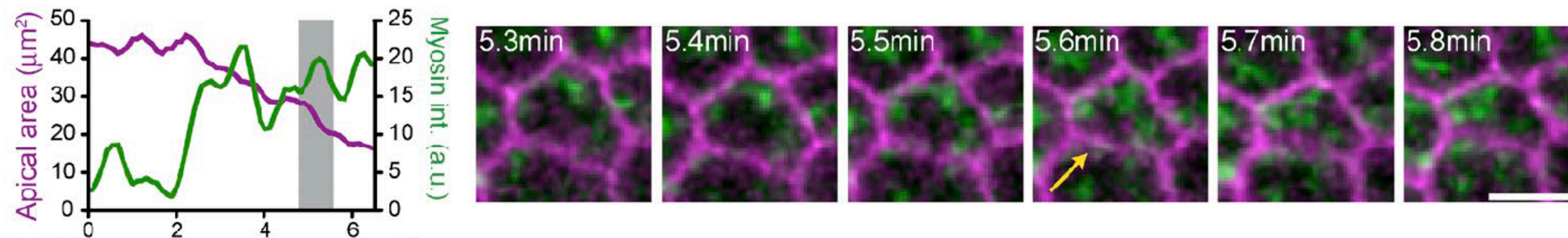
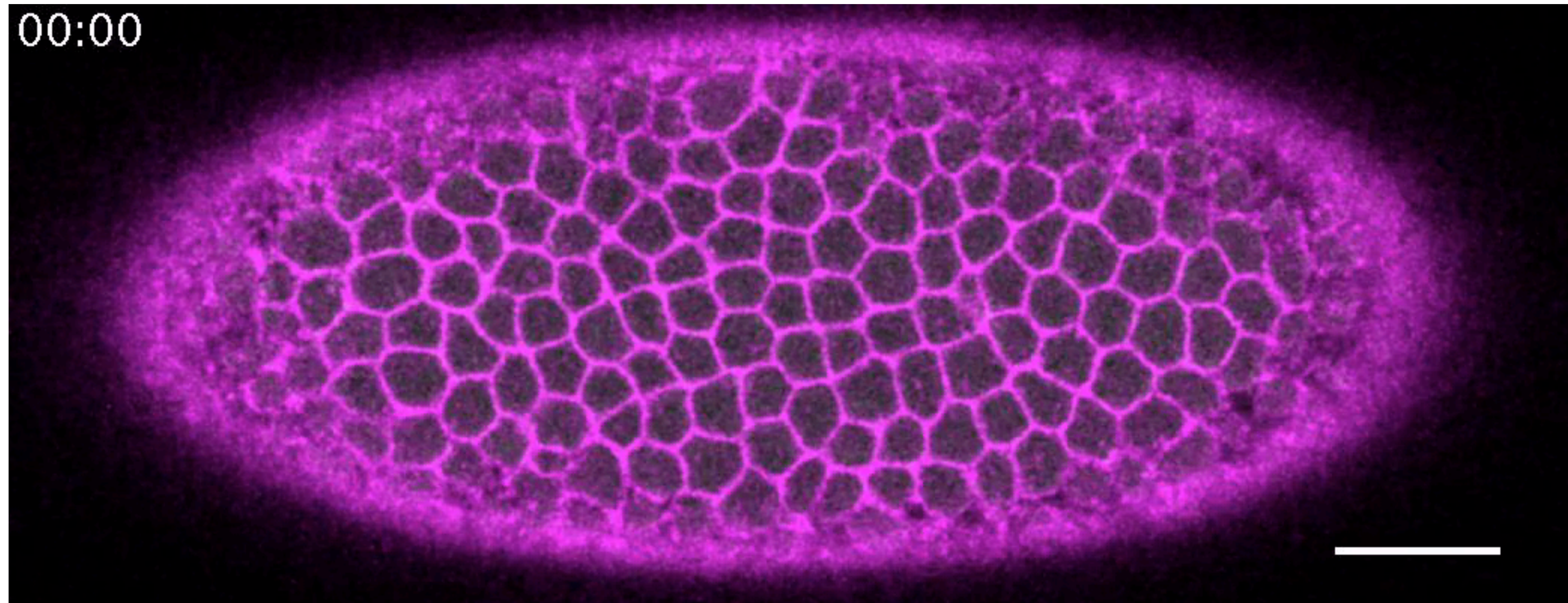


Decapentaplegic = BMP, Short gastrulation (sog) = Chordin

Actomyosin has pulsatile contractile cycles

Myosin II:GFP

Membrane:RFP

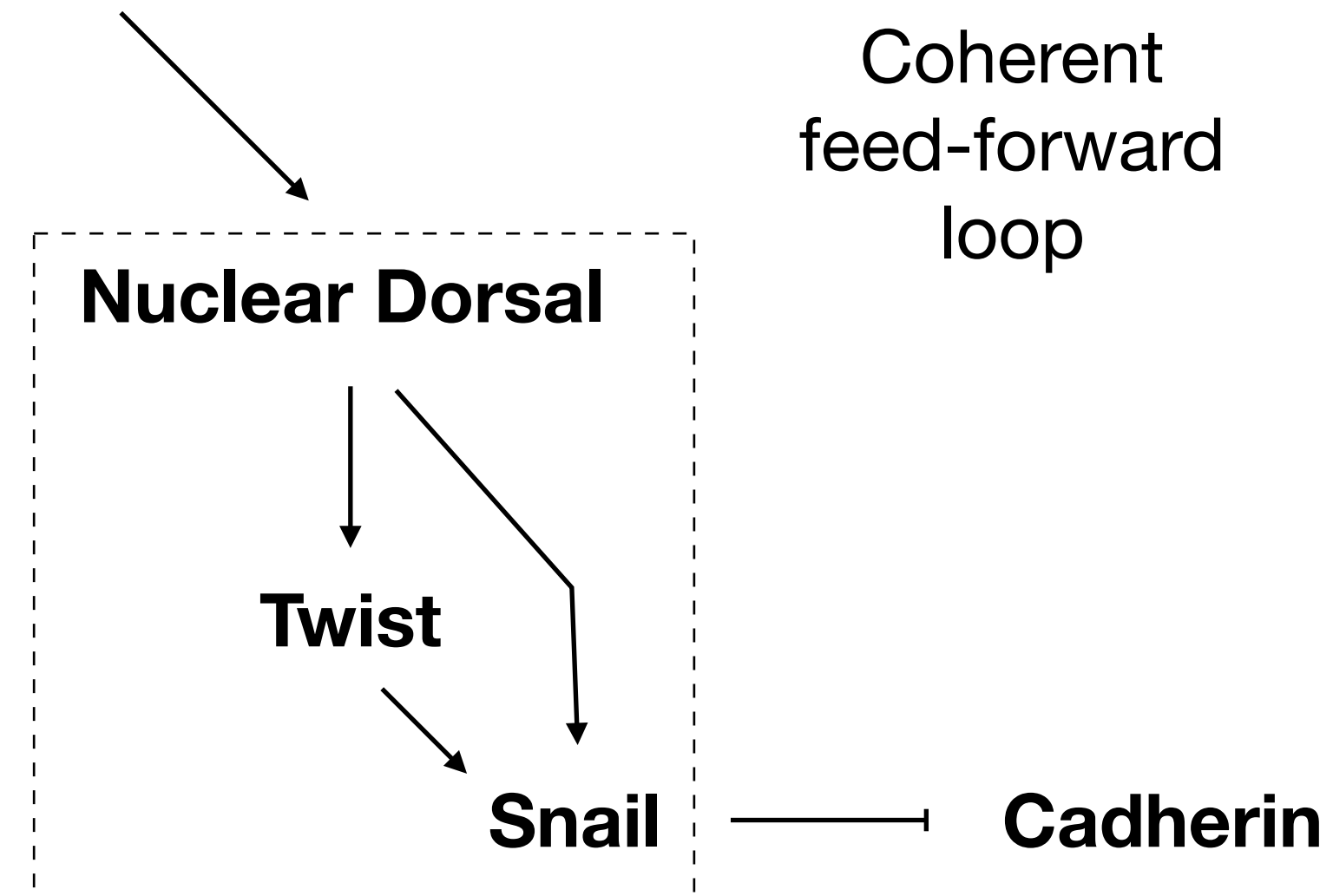


“**Ratcheting**” of apical area, gastrulation at 6 min

Timing and coordination of gastrulation?



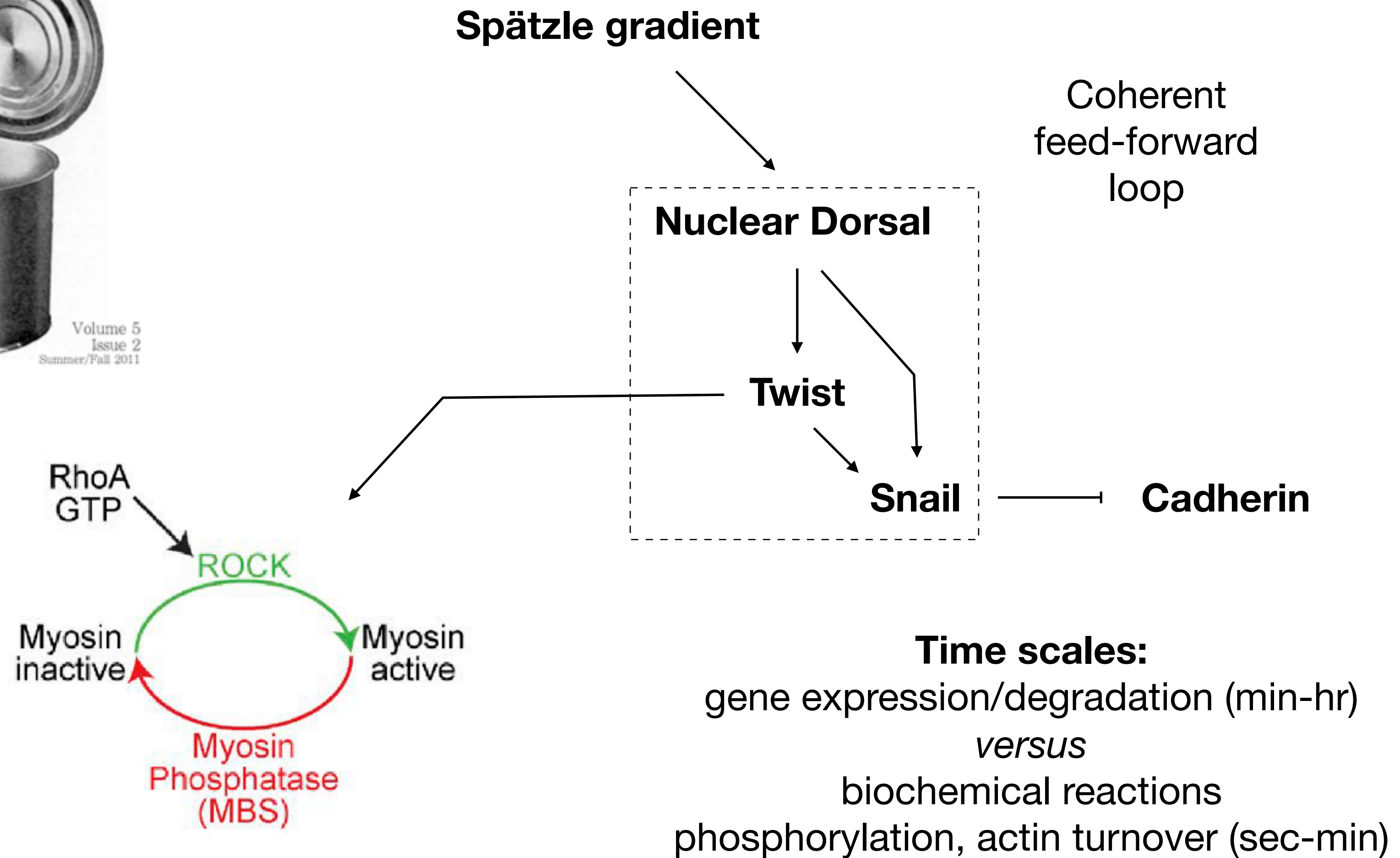
Spätzle gradient



How could this pathway control the observed *rapid* change in cell shape?



Timing of gastrulation regulated by myosin *activity*?

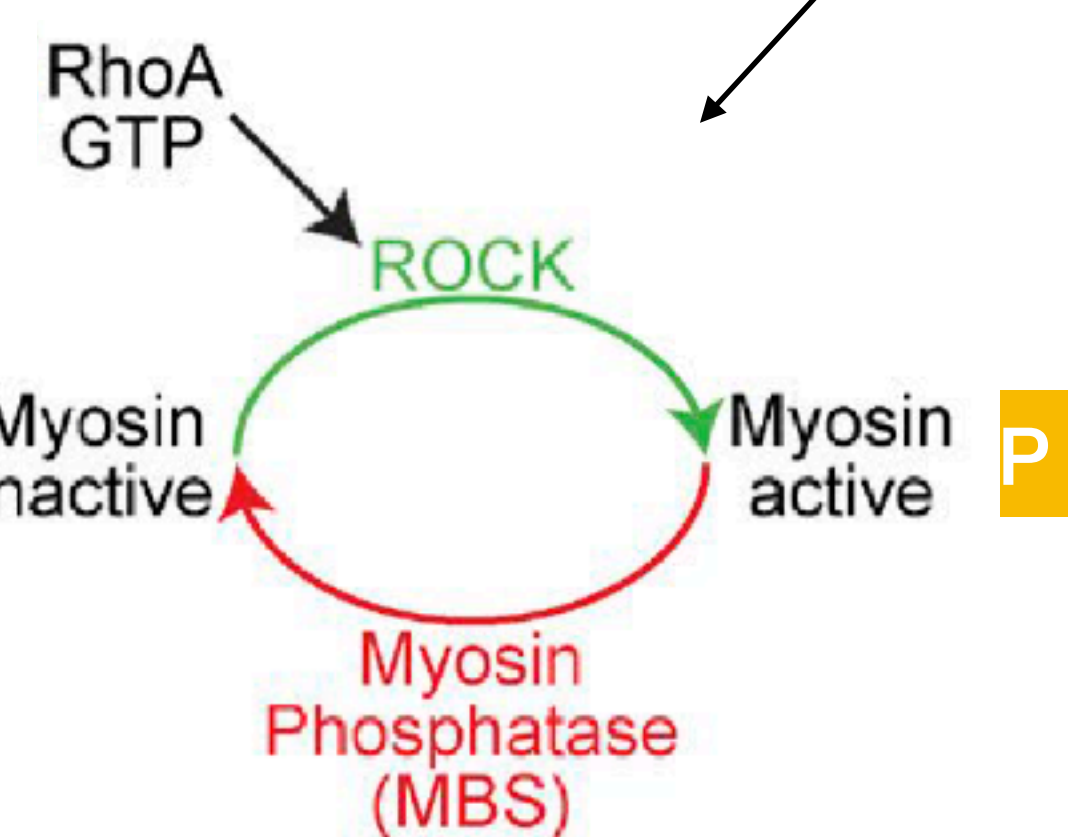
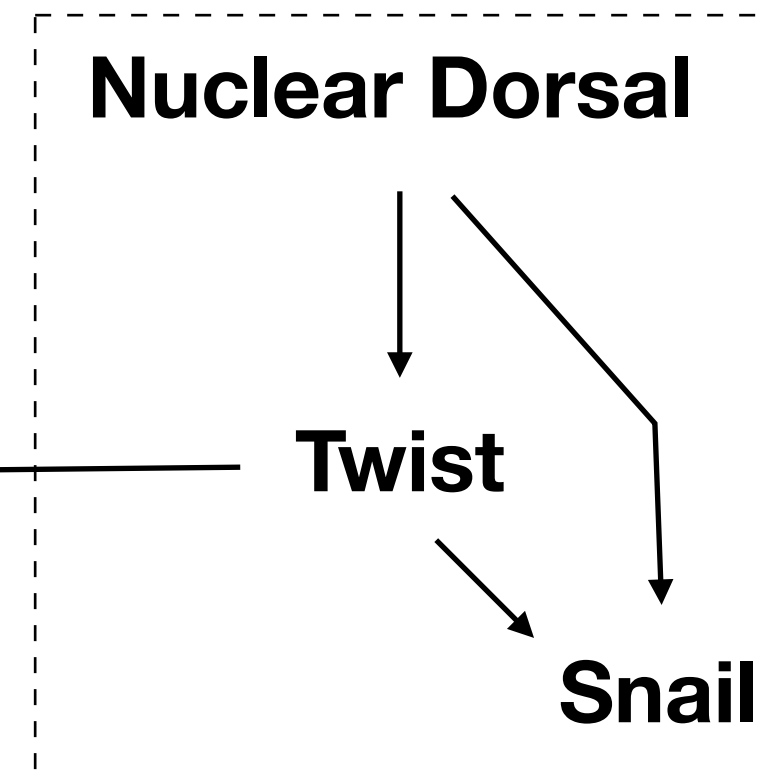


Timing of gastrulation regulated by myosin *activity*

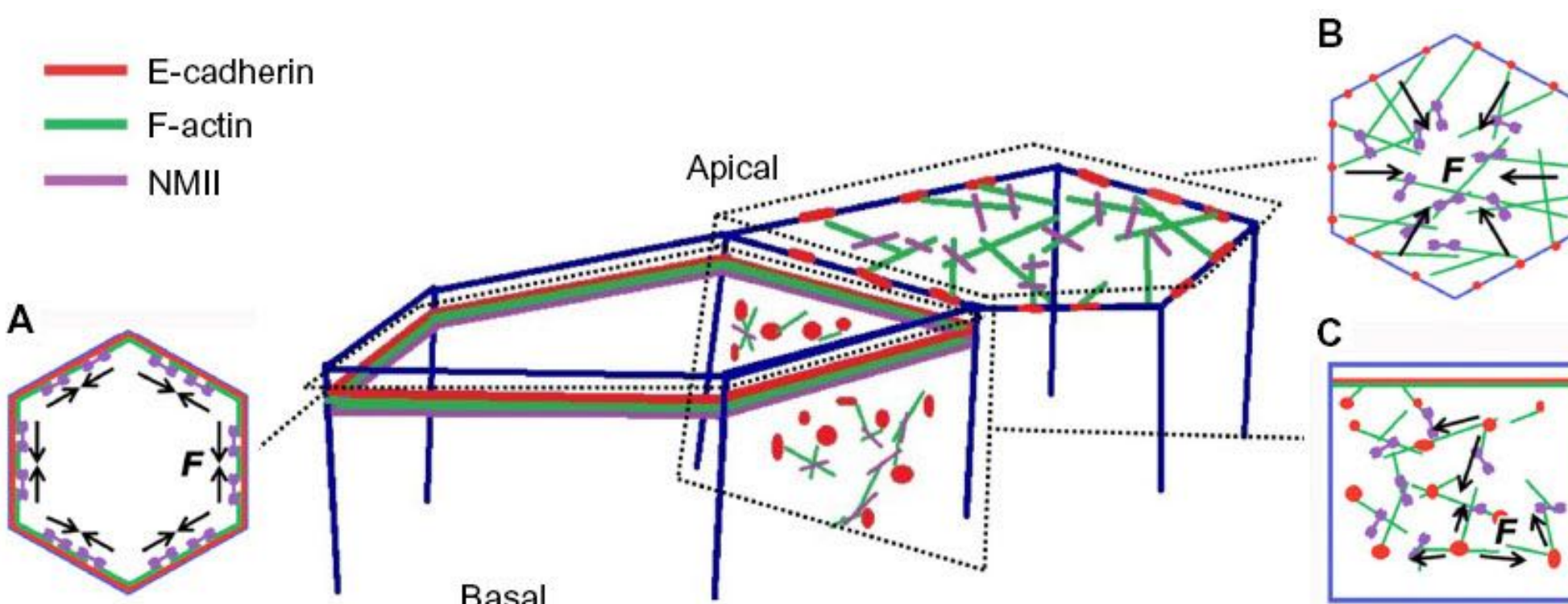


Spätzle gradient

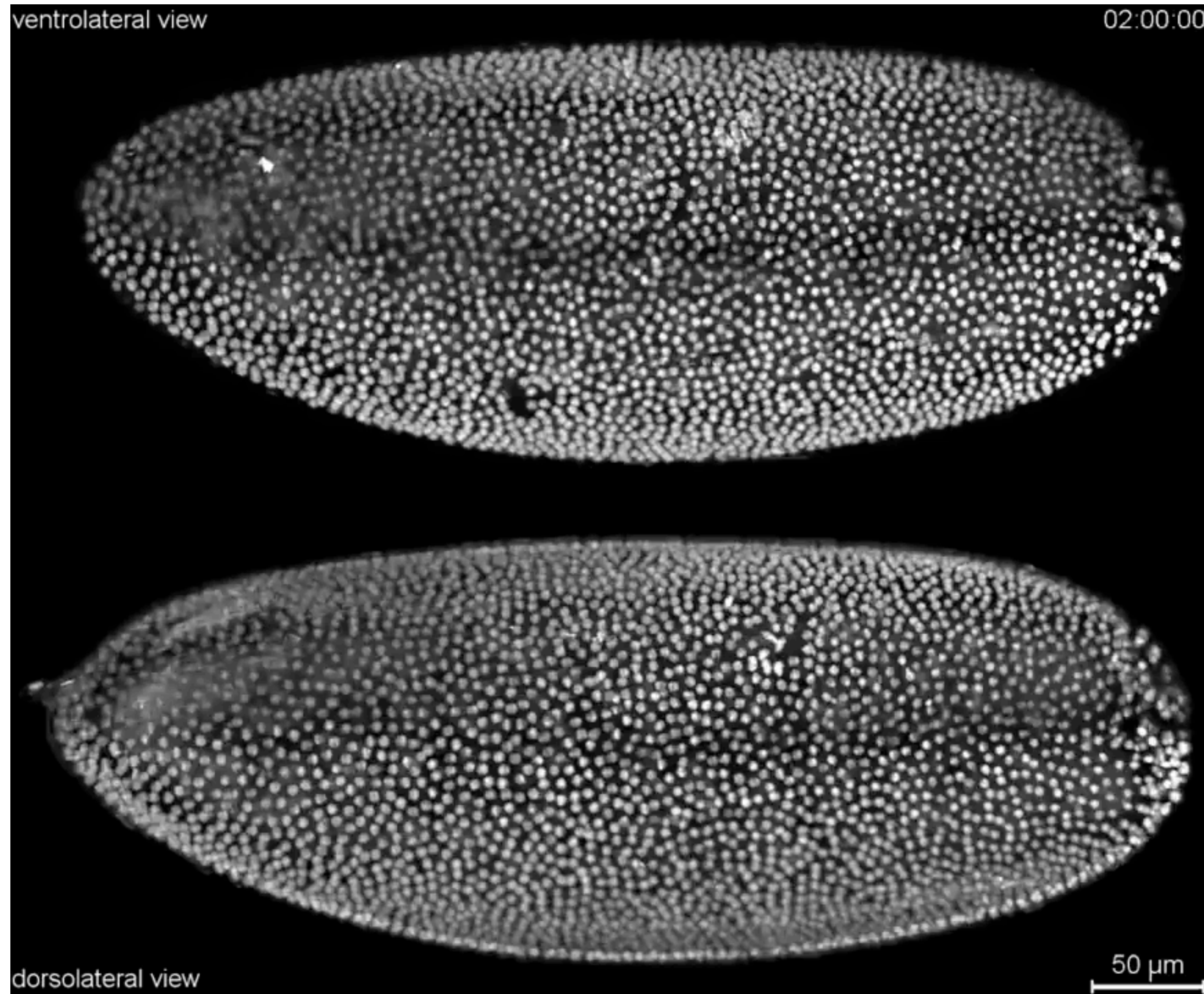
Coherent
feed-forward
loop



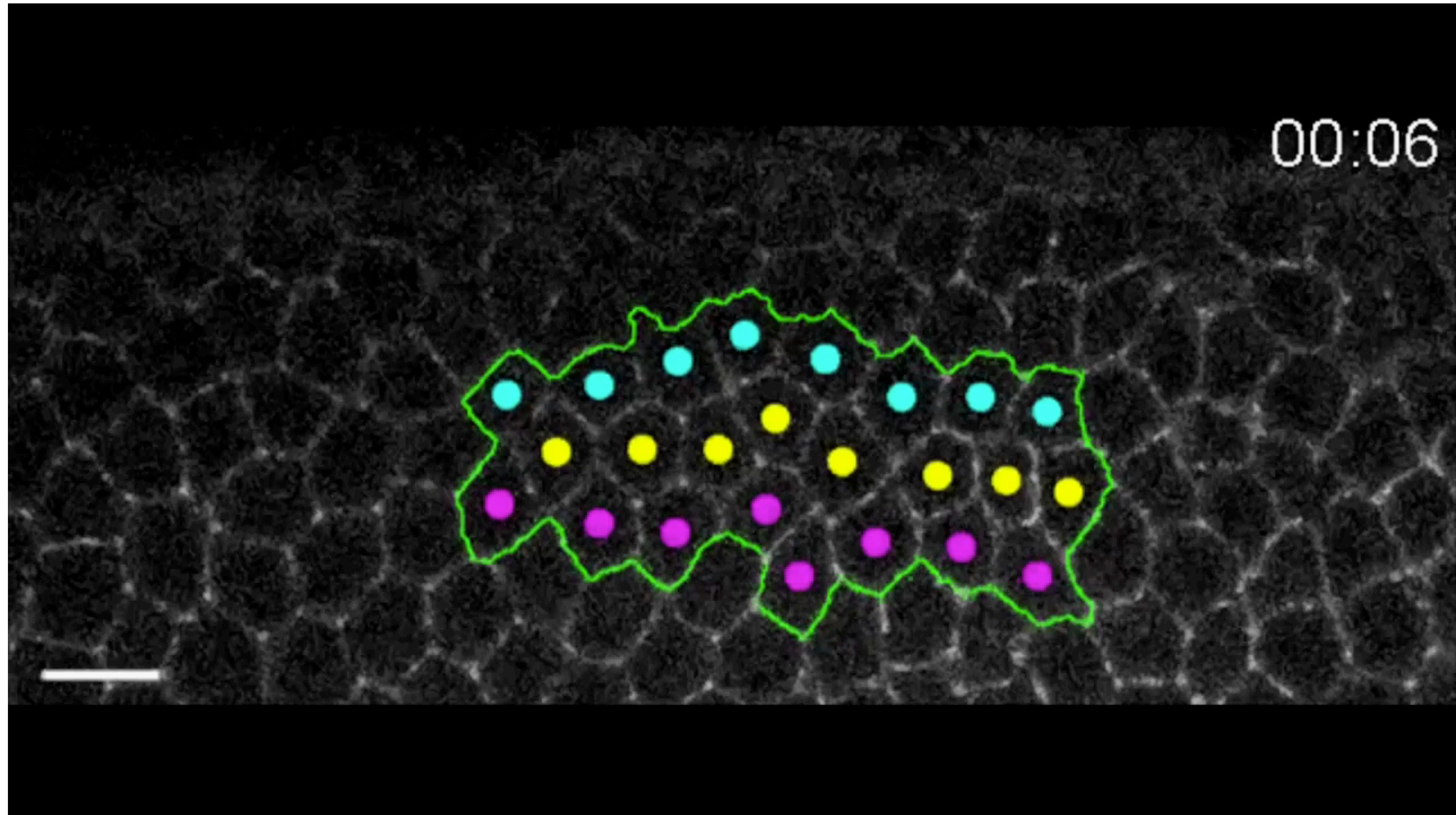
Bistability?



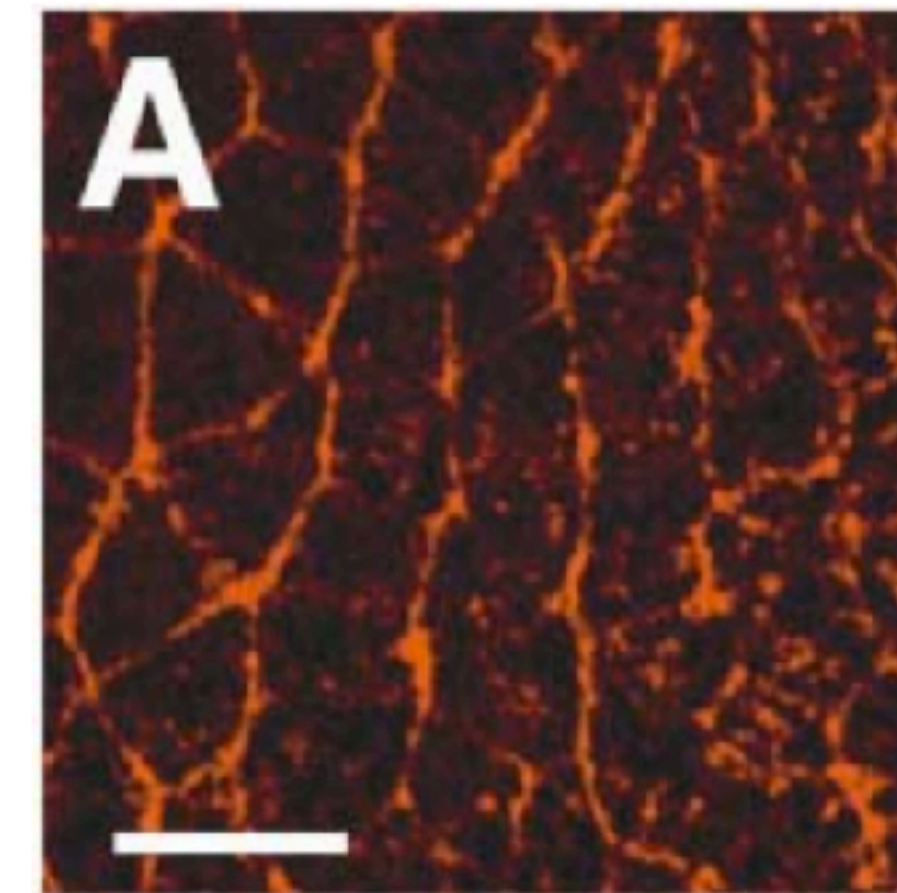
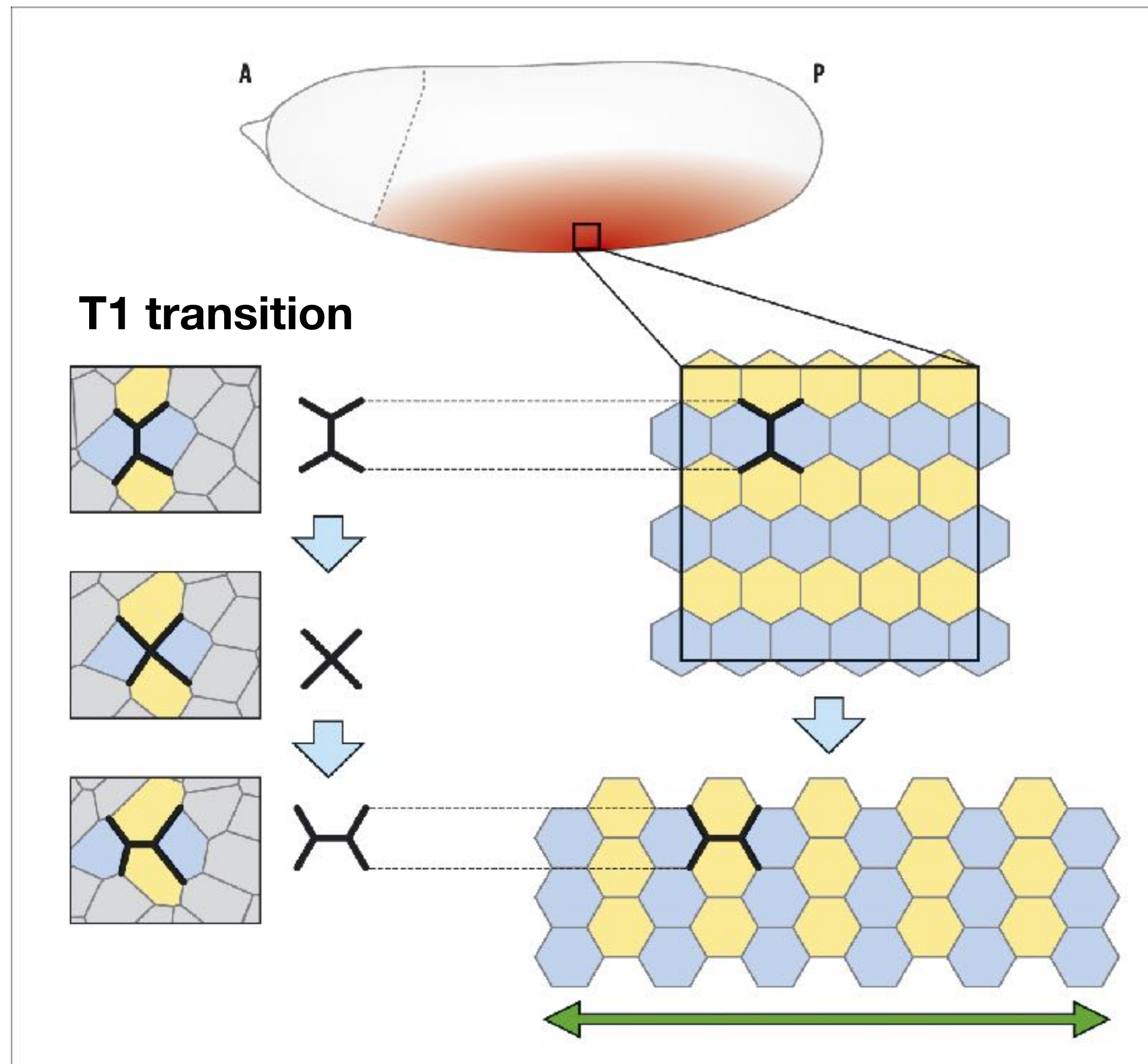
Extension of the body by “convergent extension”



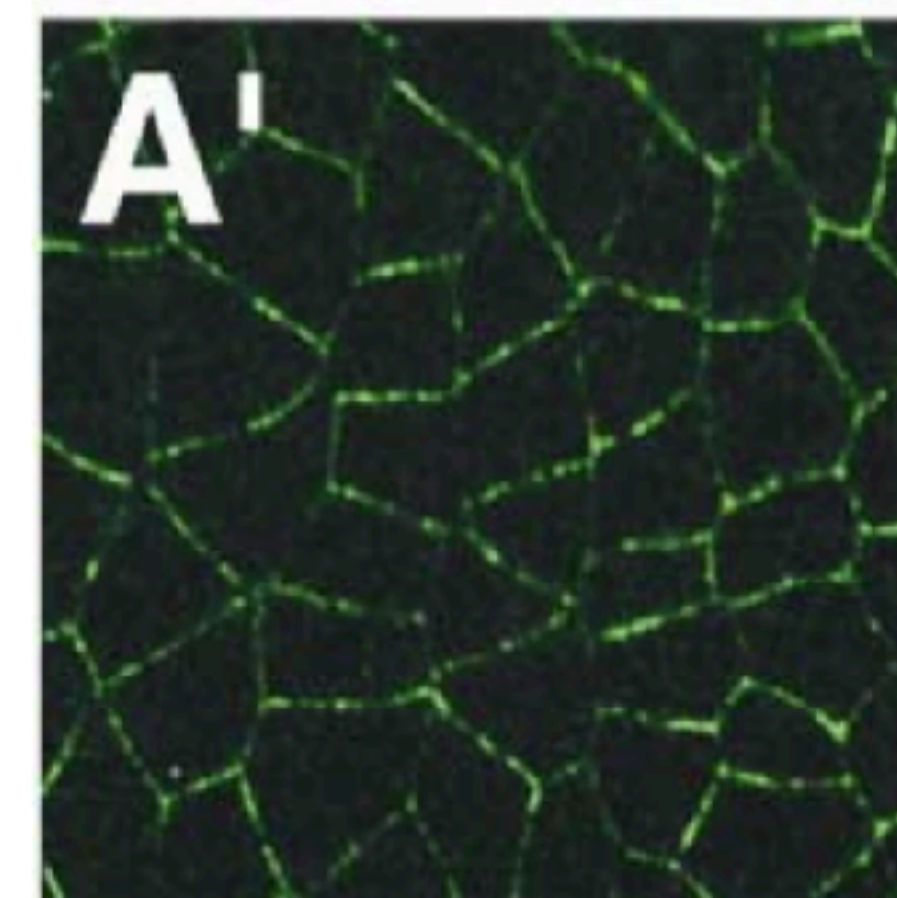
Extension of the body by “convergent extension”



Convergent extension regulated by planar cell polarity and actomyosion pulses

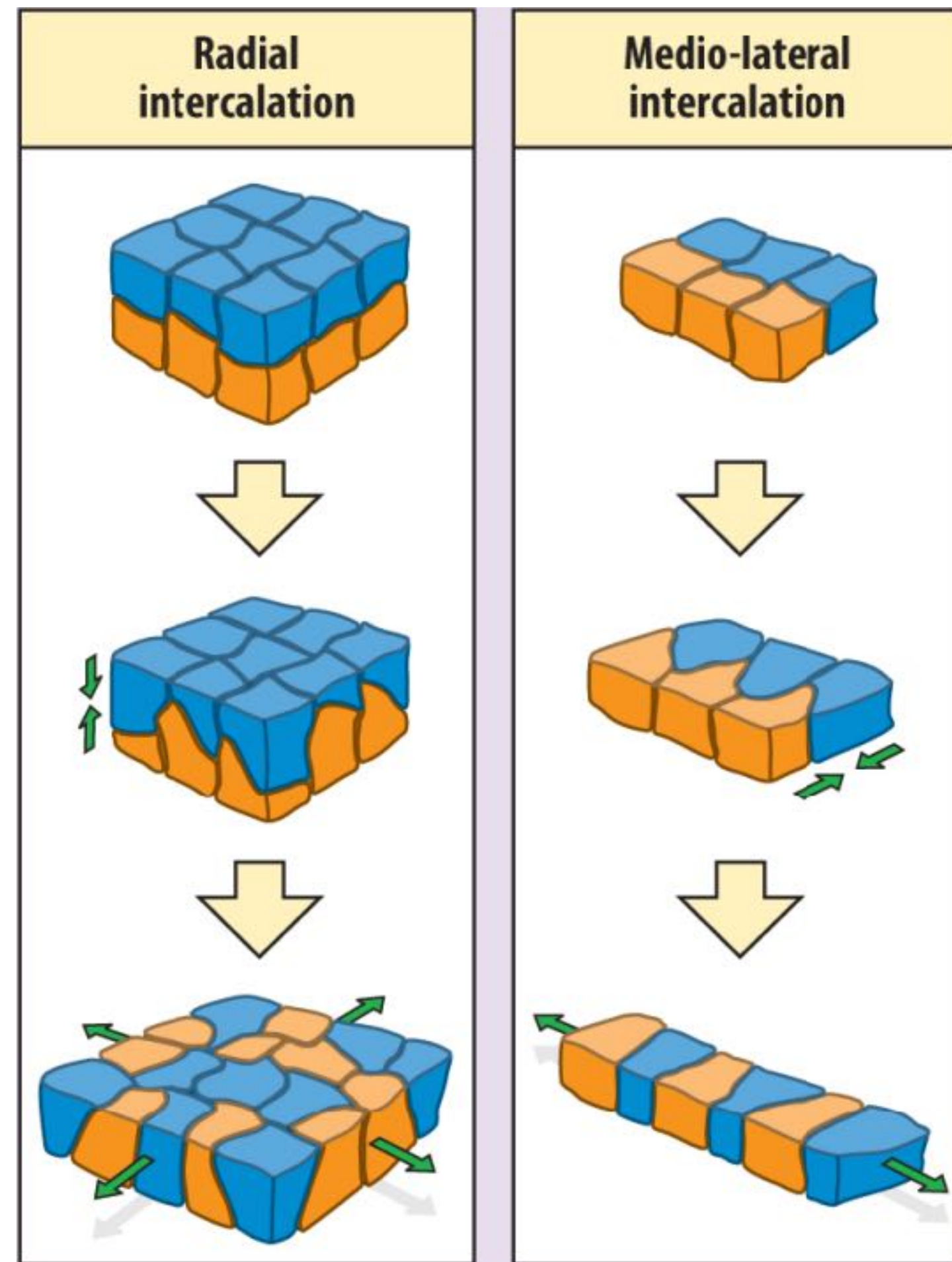


Myosin II



Par3

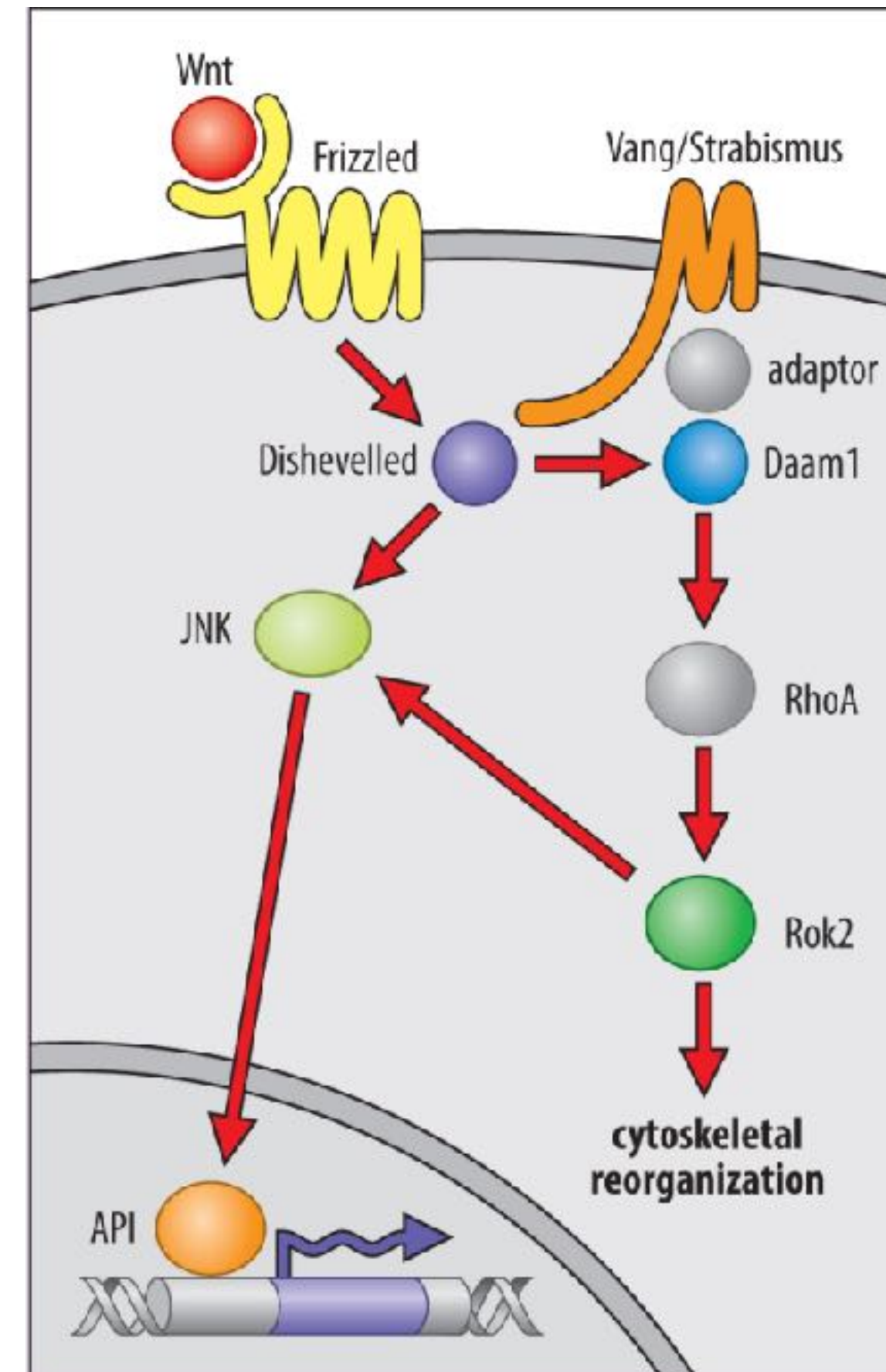
Convergent extension is regulated by planar cell polarity (PCP) pathway



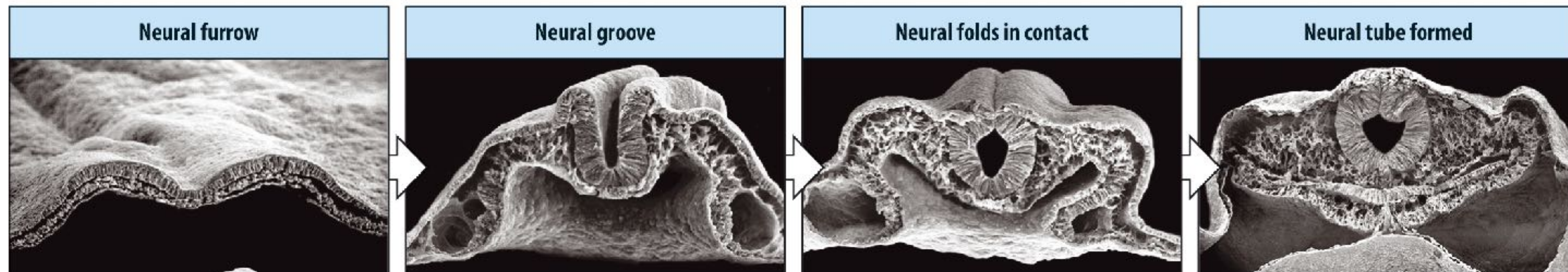
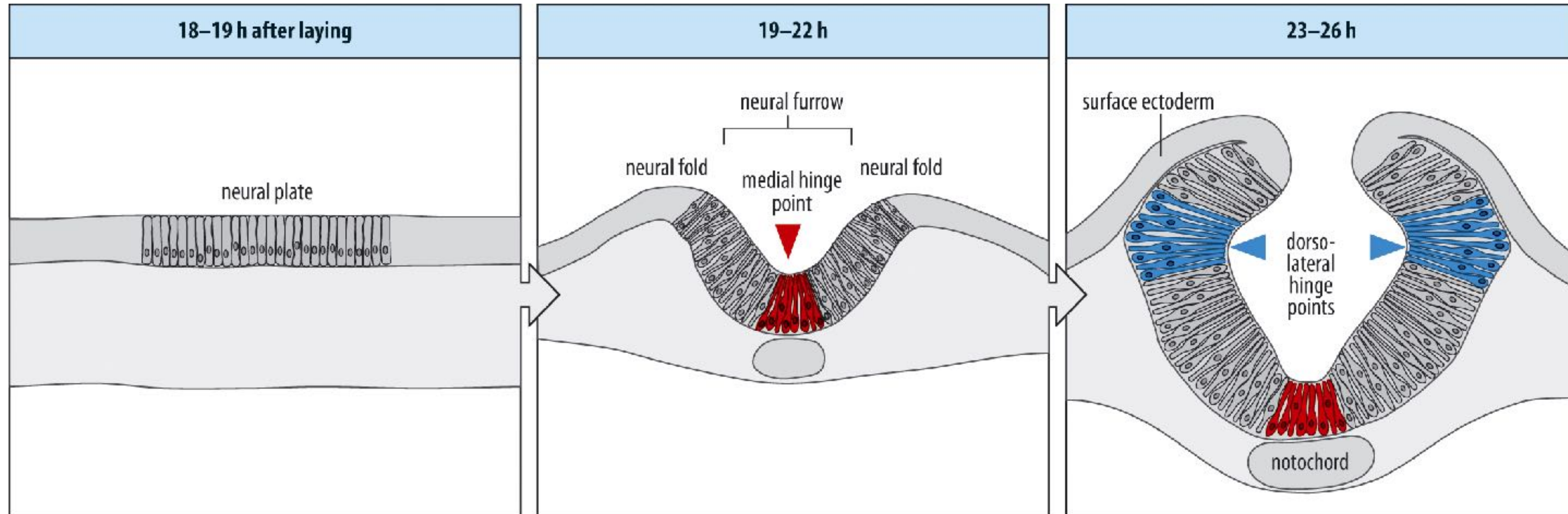
Animal cap

Notochord

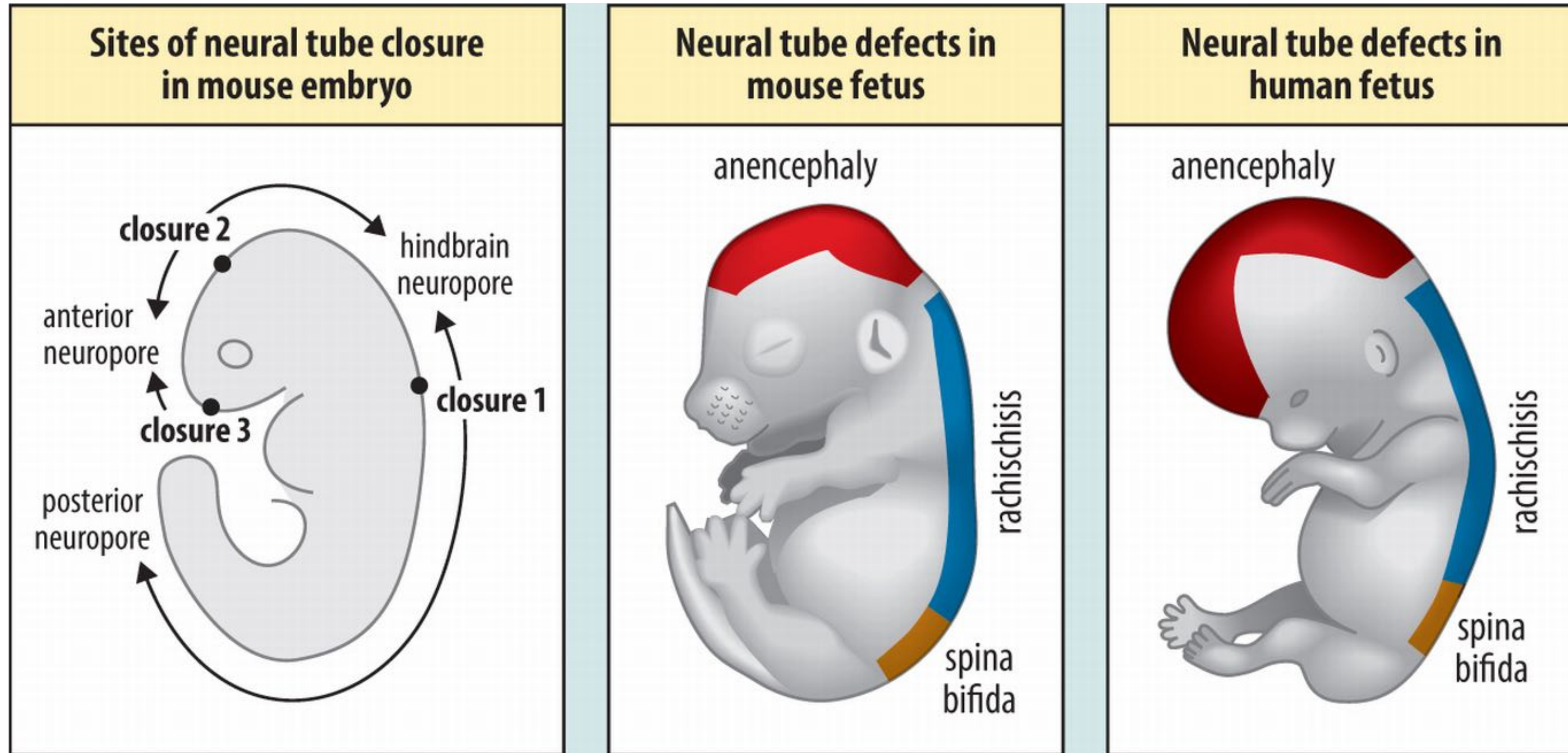
Xenopus and zebrafish



Neural tube formation

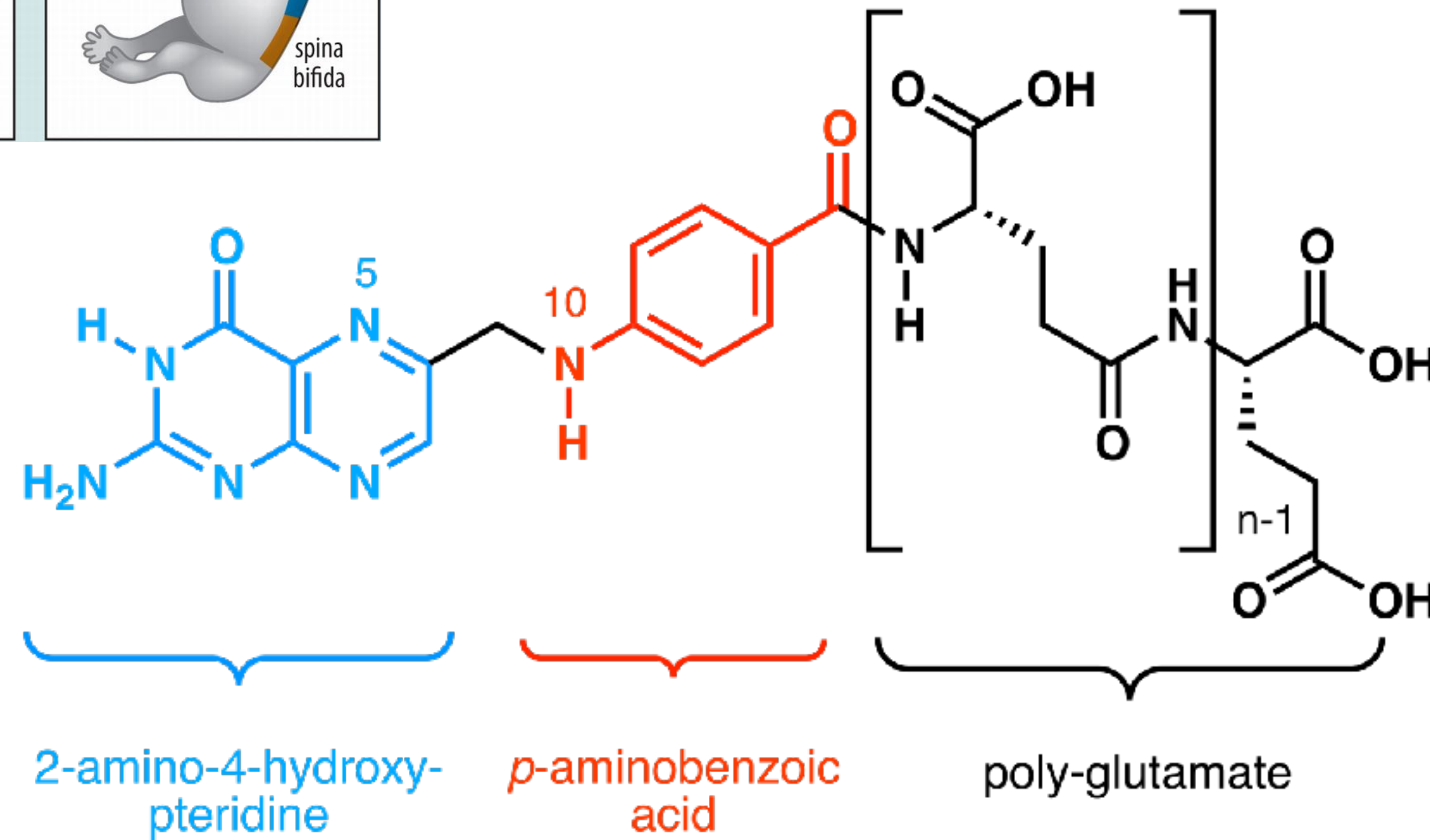
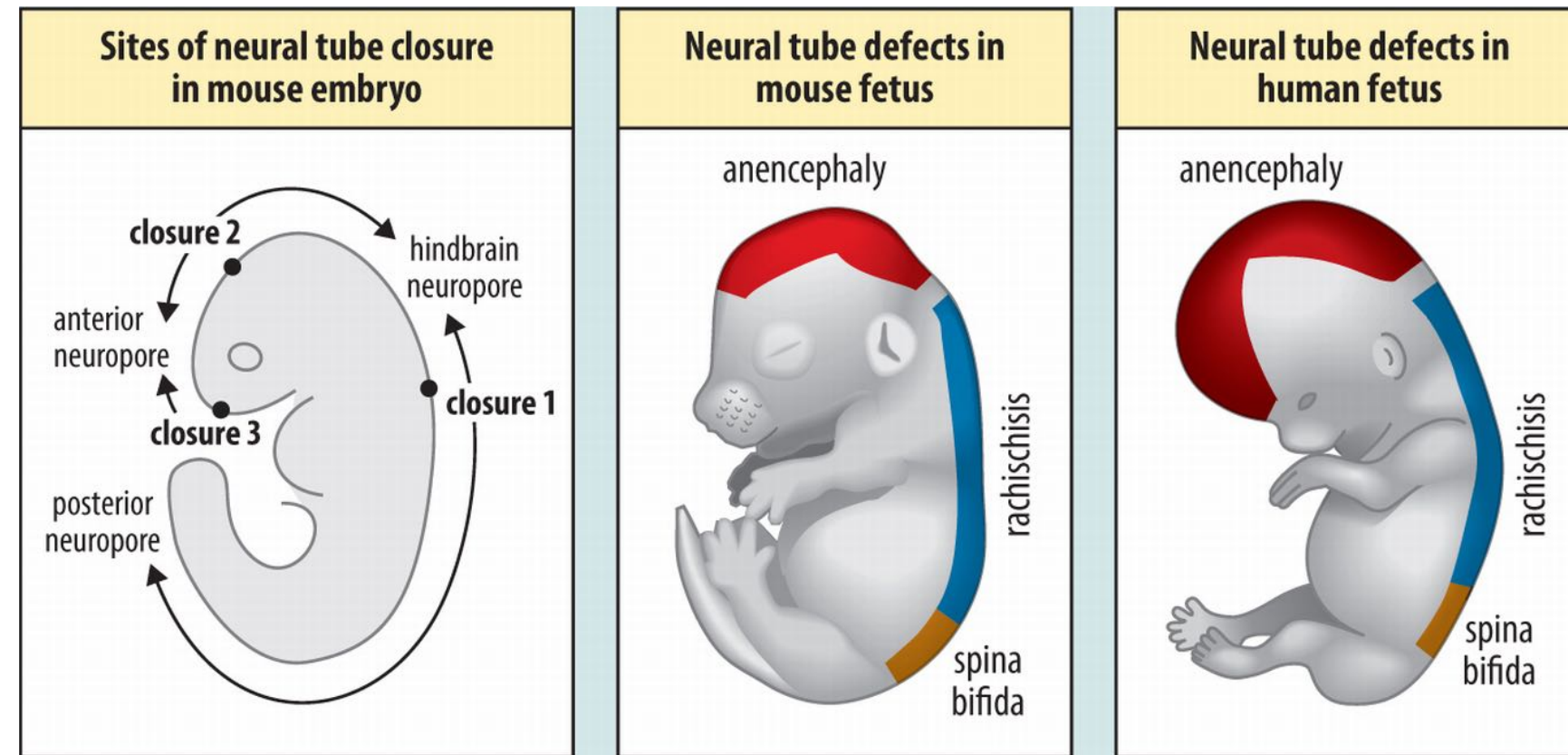


Neural tube defects



EphA7 splicing mutation

Neural tube defects



Vitamin B9, folic acid (Lucy Wills)

Vitamin B12 (Dorothy Hodgkin, Nobel Prize in Chemistry 1964)

Summary

- **Gastrulation: folding and shear**
- Gastrulation occurs in a variety of ways involving EMT, invagination and migration
- Epithelia are bent by localised contractility
- Convergent extension involves cell intercalations
- Genetic regulation of both adhesion and contractility
- Other folding - neural tube, brains and guts

Questions?

