

Cytoskeleton II: Regulation

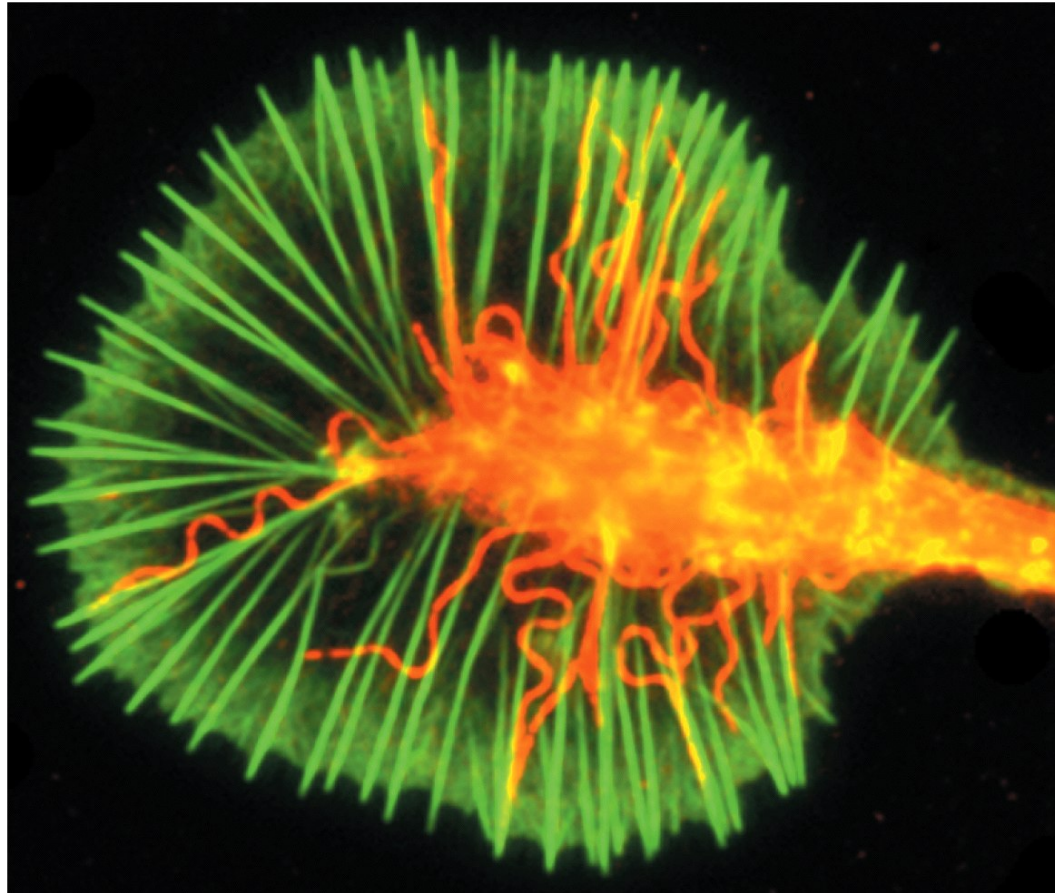


Figure 9-75b Cell and Molecular Biology, 5/e (© 2008 John Wiley & Sons)

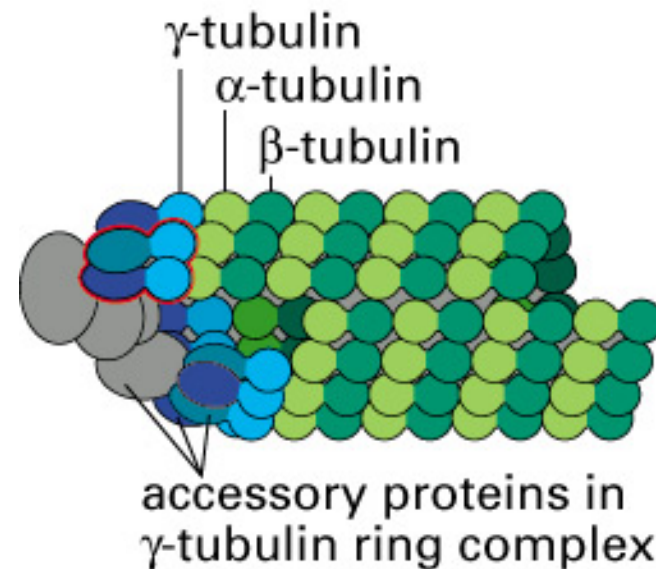
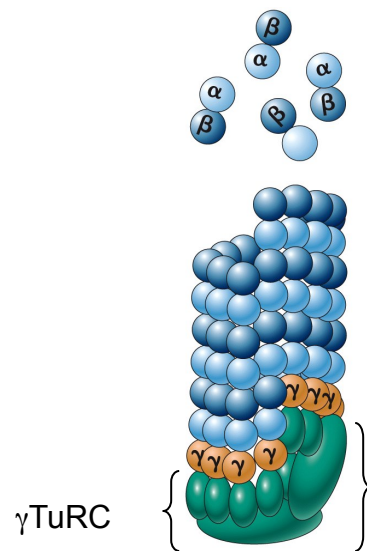
Accessory Proteins

- Filaments (AFs, MTs, IFs) are dynamic and under control of the cell.
- Form higher-order structures (e.g. mitotic spindle).
- Accessory proteins modify these cytoskeletal dynamics.



Nucleation by γ -tubulin Protein Complex

- MTOC (the centrosome) in animal cells.
- Spindle pole body in yeast.
- γ -tubulin is highly conserved.
- γ TuRC (ring complex) accelerates MT formation.
- Nucleation occurs at the (-) end.



Nucleation by γ -tubulin Complex

- MT initiation also occurs in cytosol
- e.g. plants.
- MTs nucleate daughter MTs.

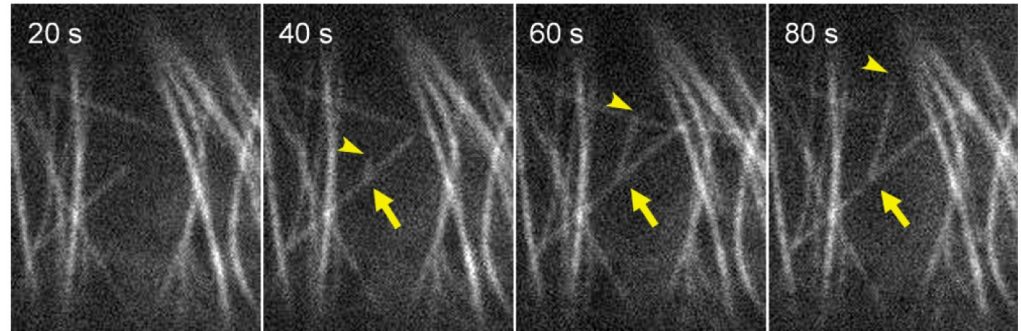
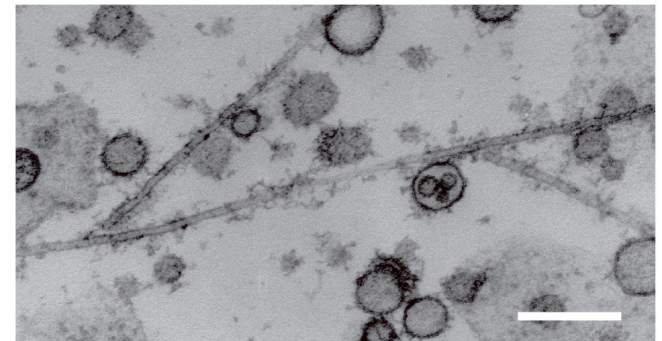
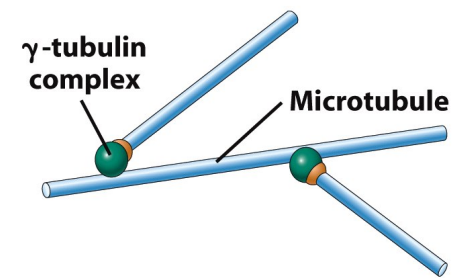


Figure 9-22a Cell and Molecular Biology, 5/e (© 2008 John Wiley & Sons)



(b)



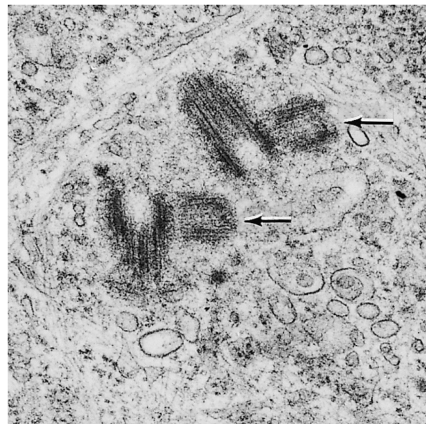
(c)

Figure 9-22bc Cell and Molecular Biology, 5/e (© 2008 John Wiley & Sons)

The Centrioles and Centrosome

- Centrosome contains 2 cylindrical centrioles.
- 9 triplets of MTs.
- Centrioles organize PCM (Pericentriolar Material).
- Centriole lumen and PCM contain γ -tubulin.
- PCM initiates MT assembly.

*2 pairs of centrioles
after duplication during
cell cycle.*



0.3 μm

Figure 9-18c Cell and Molecular Biology, 5/e (© 2008 John Wiley & Sons)

Karp. Cell and Molecular Biology

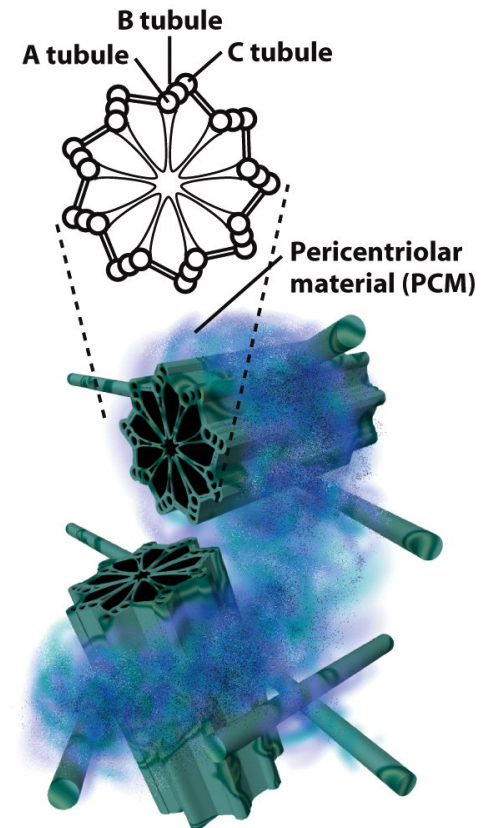
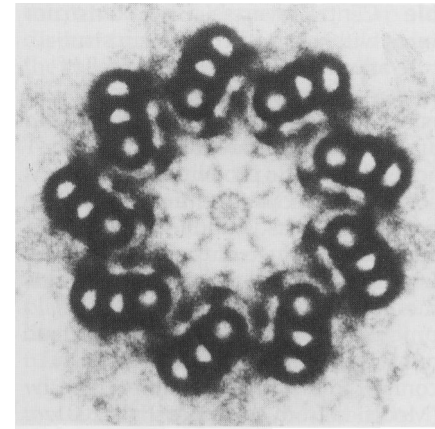


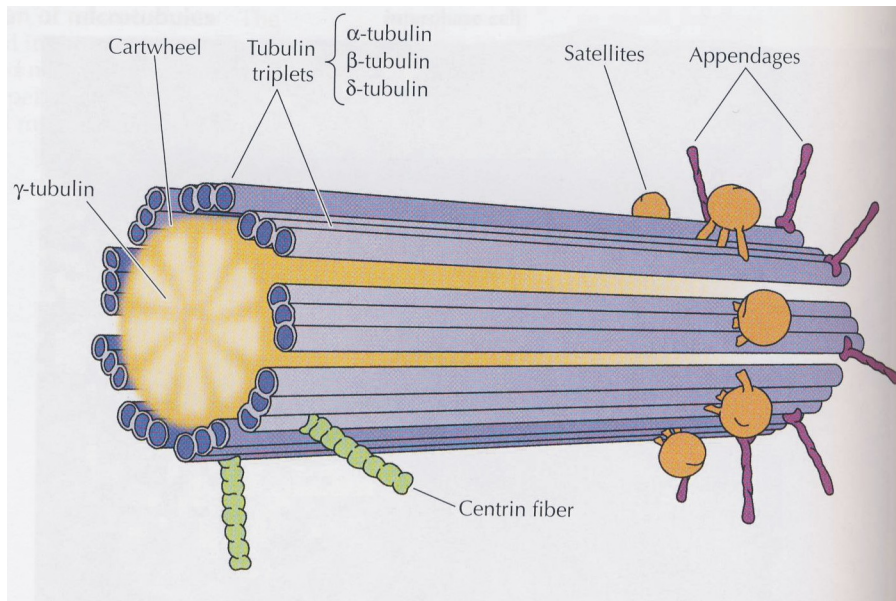
Figure 9-18a Cell and Molecular Biology, 5/e (© 2008 John Wiley & Sons)

The Centrioles and Centrosome

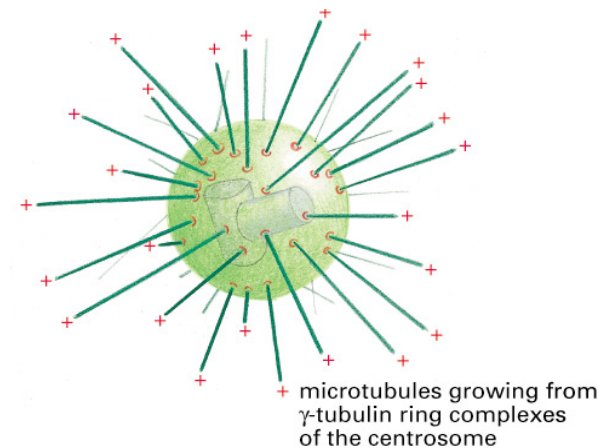
- Centrosome near nucleus.
- (-) ends of MTs are anchored.
- (+) ends of MTs emanate in astral configuration.



0.1 μm



Cooper and Hausman. The Cell: A Molecular Approach

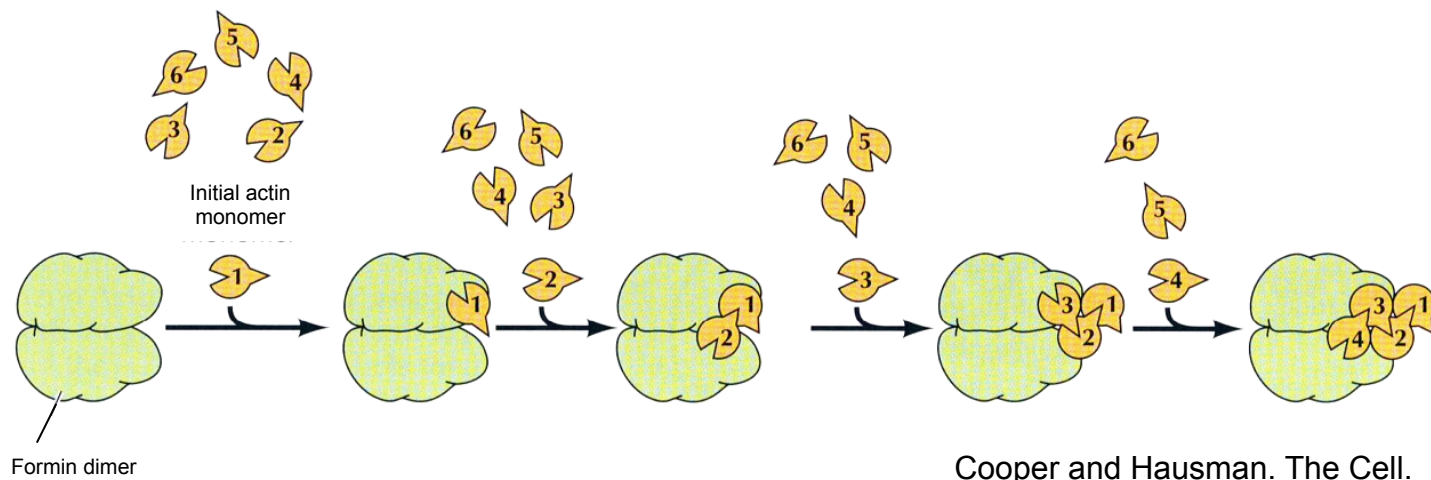


Nucleation of AFs

- Cell periphery (cortex), where density of AF proteins is highest.
- Location related to AF function (cell shape, movement).
- Facilitated by ABPs (actin binding proteins) including ARPs (actin related proteins).

Initiation of AFs (unbranched)

- Initiation by *formin*, an ABP
- Correct alignment for polymerization
- e.g. filaments of muscle cells.



Initiation of AF Branches



- Cell periphery
- Arp2/3 produces extensive branching.
- Complex contains 7 proteins.
- Binds at (-) end.
- 70° favourable angle.

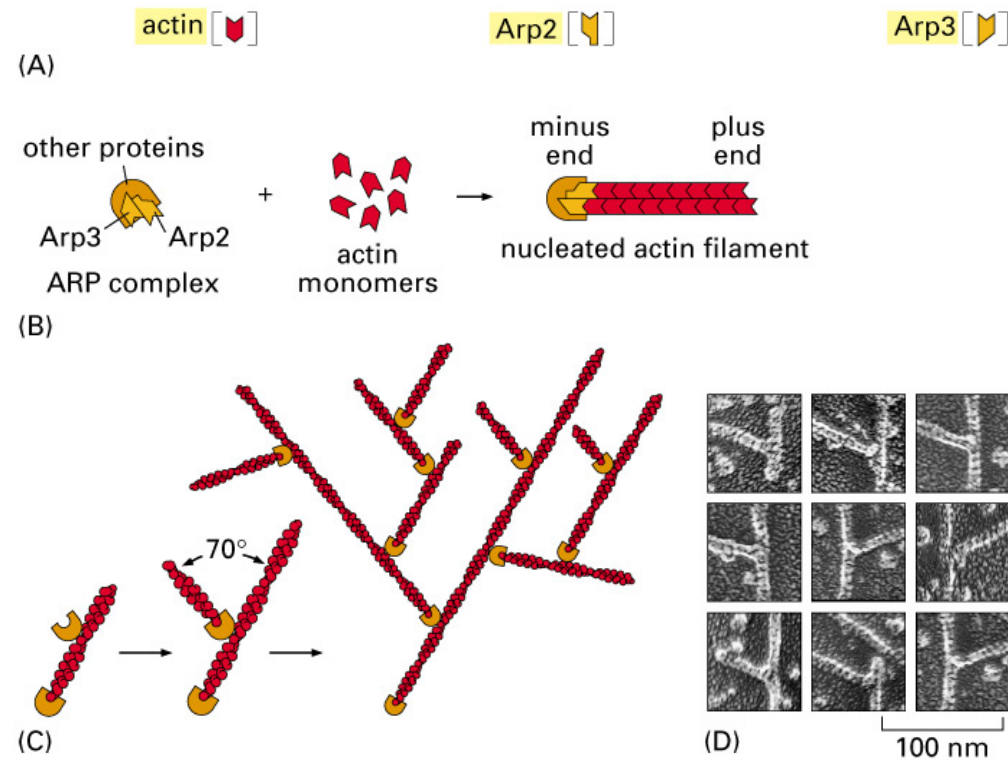


Figure 16–28 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

Control of Subunit Pools

- Both actin and tubulin are maintained in the cytosol at high concentrations.
- Concentration can exceed C_c .
- Accessory proteins may sequester unused subunits (*sequestering proteins*).
- Sequestered proteins are not hydrolyzed.
- Provide control or regulation of filament elongation.



Thymosin Sequesters Actin

- Thymosin sequesters, but profilin recruits monomers.
- Thymosin makes polymerization *less favourable*.
- Profilin *competes* with thymosin and promotes assembly.

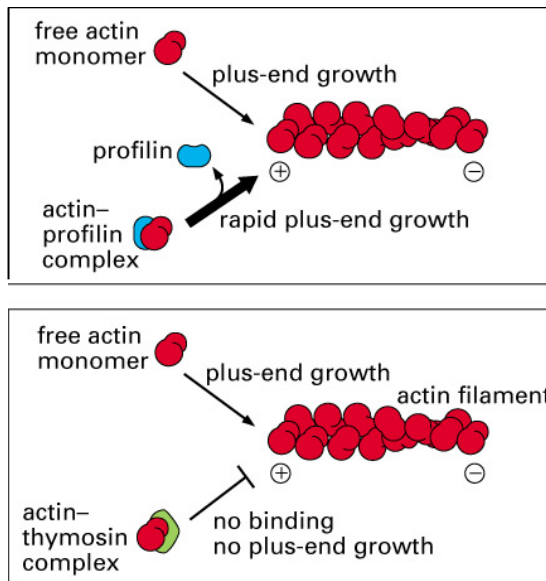


Figure 16-30 part 1 of 2. Molecular Biology of the Cell, 4th Edition.

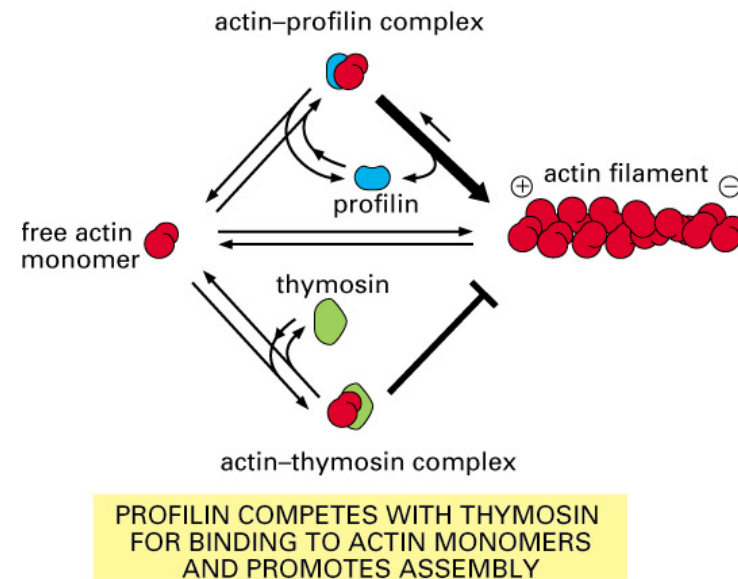


Figure 16-30 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

Stathmin Sequesters Tubulin

- Stathmin binding *prevents* polymerization.
- Decreases effective [tubulin].
- Promotes dynamic instability (catastrophe).

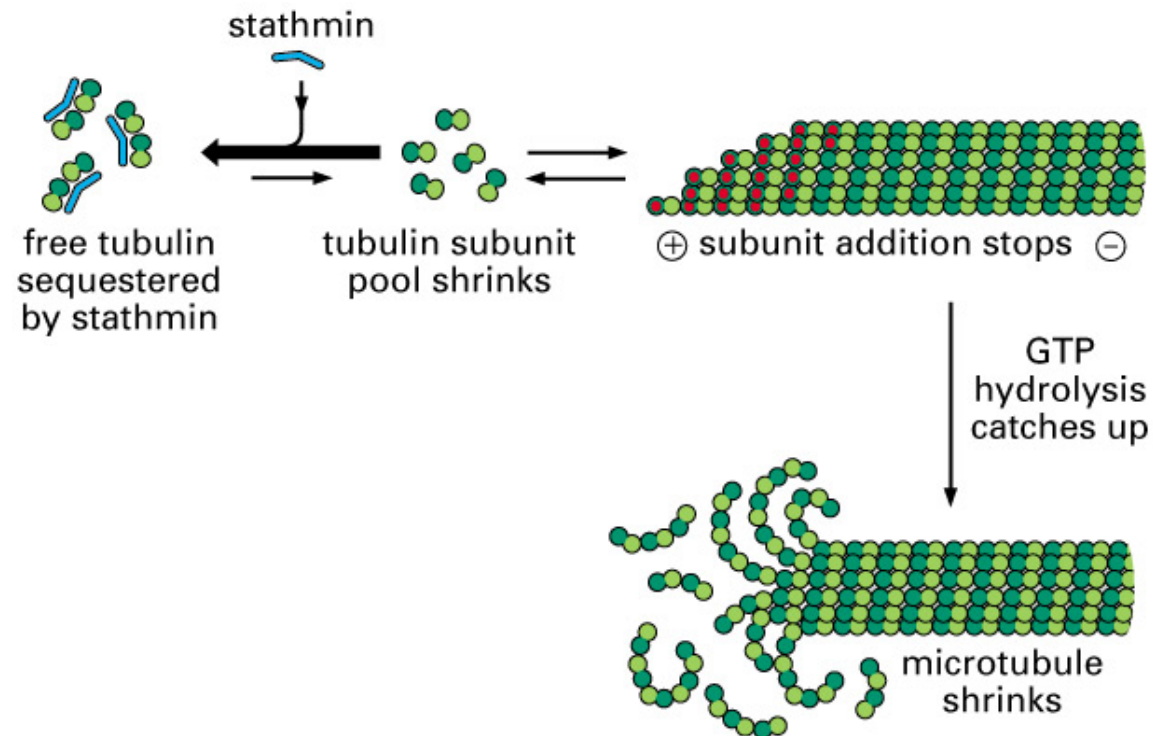


Figure 16-31. Molecular Biology of the Cell, 4th Edition.

MT-Associated Proteins (MAPs)

- Several binding domains.
- Length of projecting domain determines packing of MTs.
- MAP2 (neuronal cell bodies); *tau* (axons).
- *tau* and Alzheimer's Disease (*tau* unable to bind to MTs).
- Poorly soluble (hyperphosphorylated) *tau* may induce neurodegeneration.

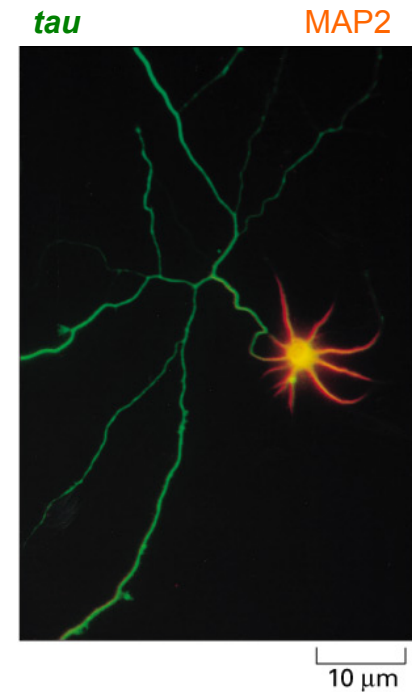


Figure 16–32. Molecular Biology of the

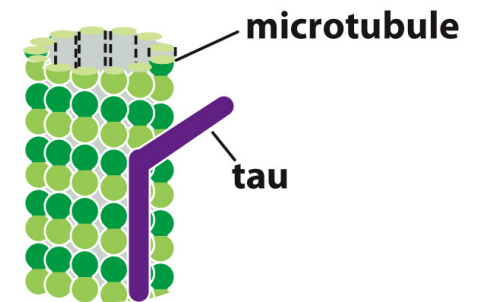
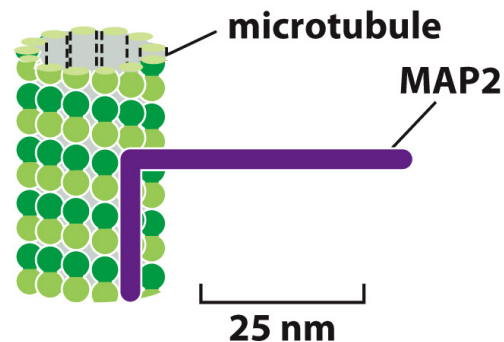
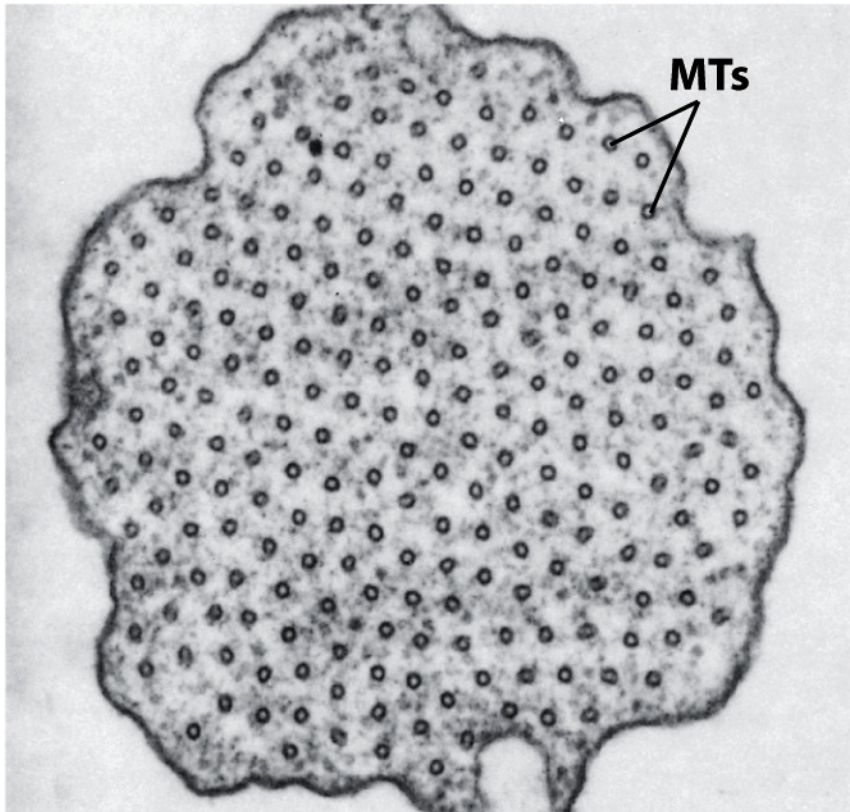


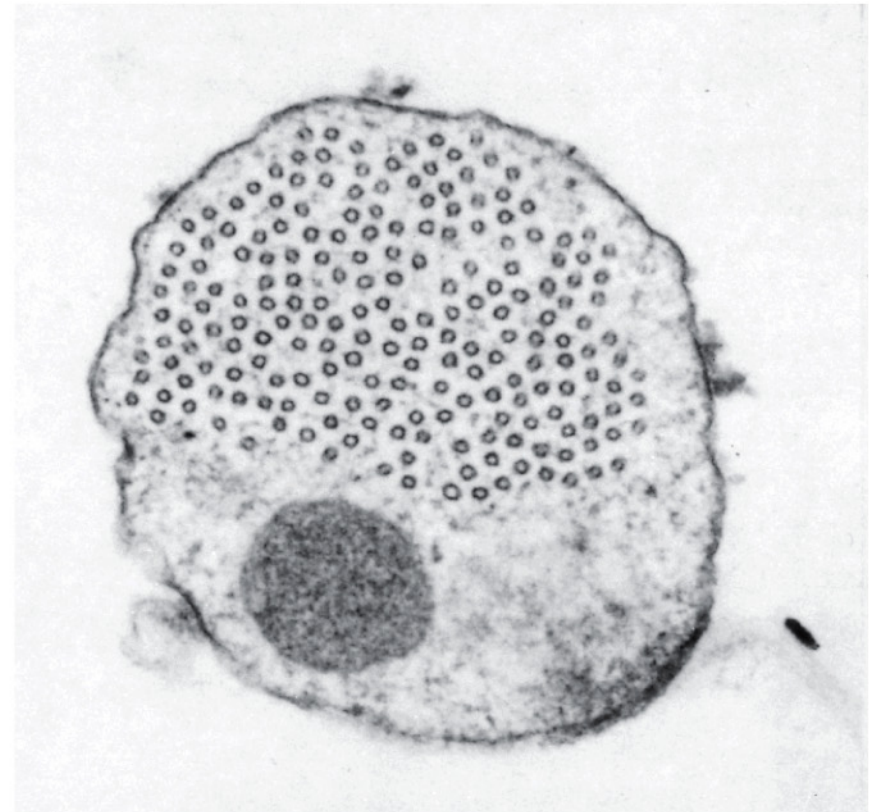
Figure 16-51ab Molecular Biology of the Cell 6e (© Garland Science 2015)



MAP2



tau



300 nm

Figure 16-51cd Molecular Biology of the Cell 6e (© Garland Science 2015)

AF Binding Proteins

Cofilin destabilizes AFs

- binds to side of proteins.
- binding induces mechanical stress.
- treadmilling, turnover.
- cell locomotion.

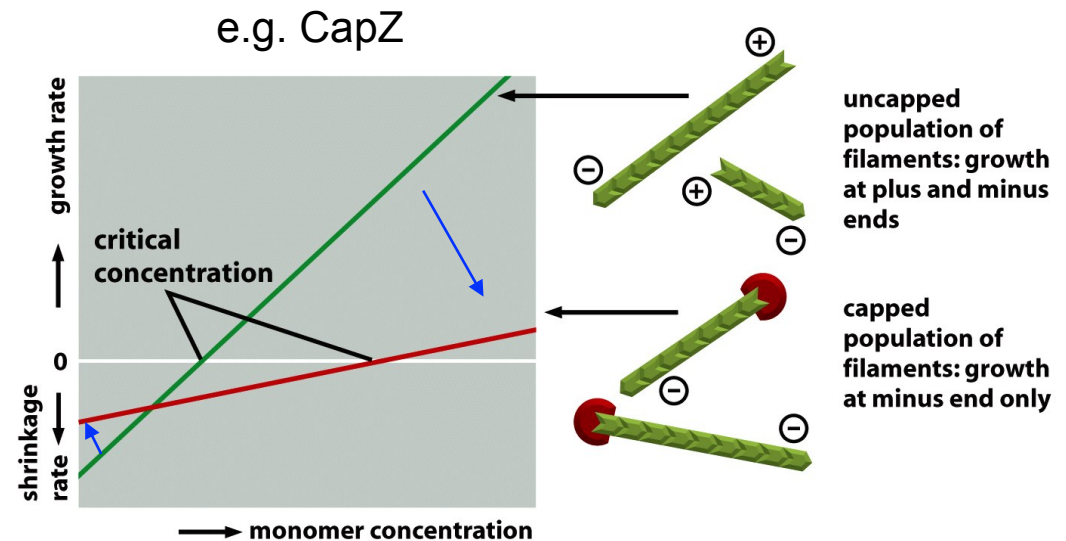
Tropomyosin stabilizes AFs

- binds to side of proteins.
- muscle contraction.

Modifications at AF Ends



- “Capping”
- Recall elongation slower at (-) end.
- e.g. CapZ (+), elongation occurs only at (-) end.
- e.g. tropomodulin (-) in muscle contraction.



Modifications at MT Ends

- Capping in MTs has dramatic effect on dynamic instability.
- Important during mitosis.

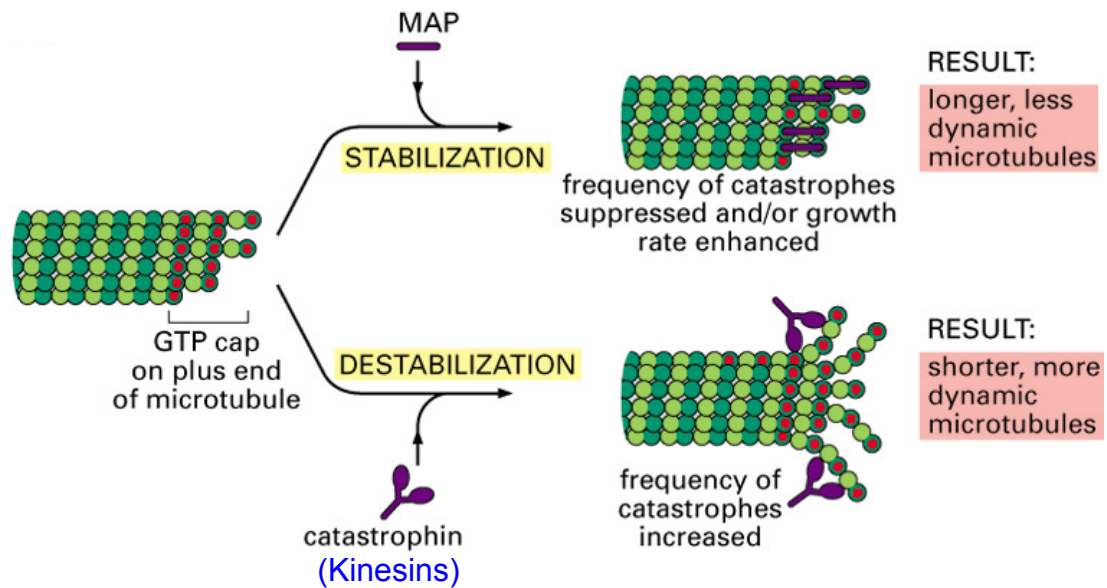


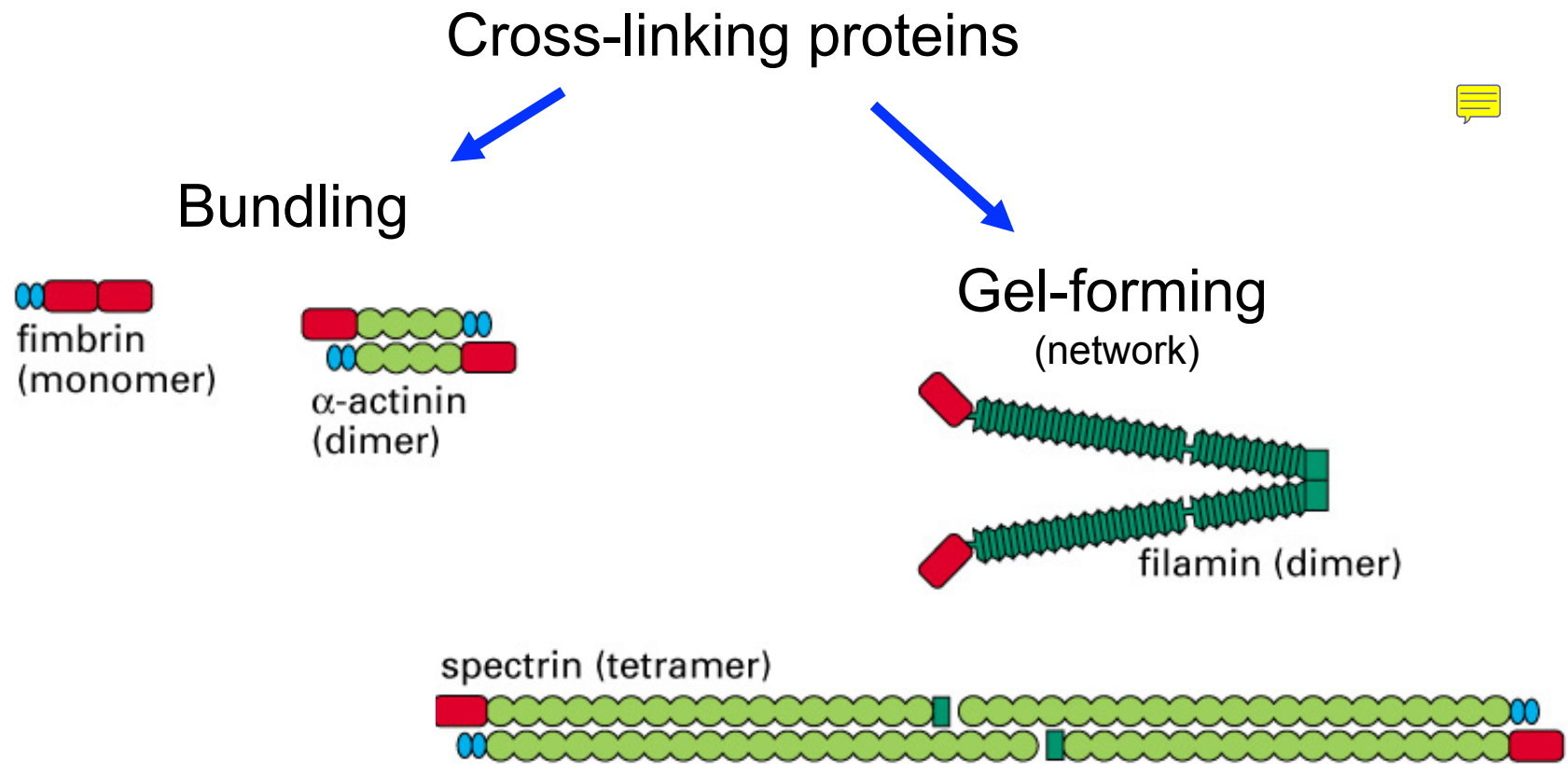
Figure 16-36 part 1 of 2. Molecular Biology of the Cell, 4th Edition.

Cross-Linking Proteins and AFs

- Formation of higher-order structures.
- Spacing of 2 actin binding domains of cross-linking protein determines type of structure.
- 2 major groups: bundling and gel-forming.

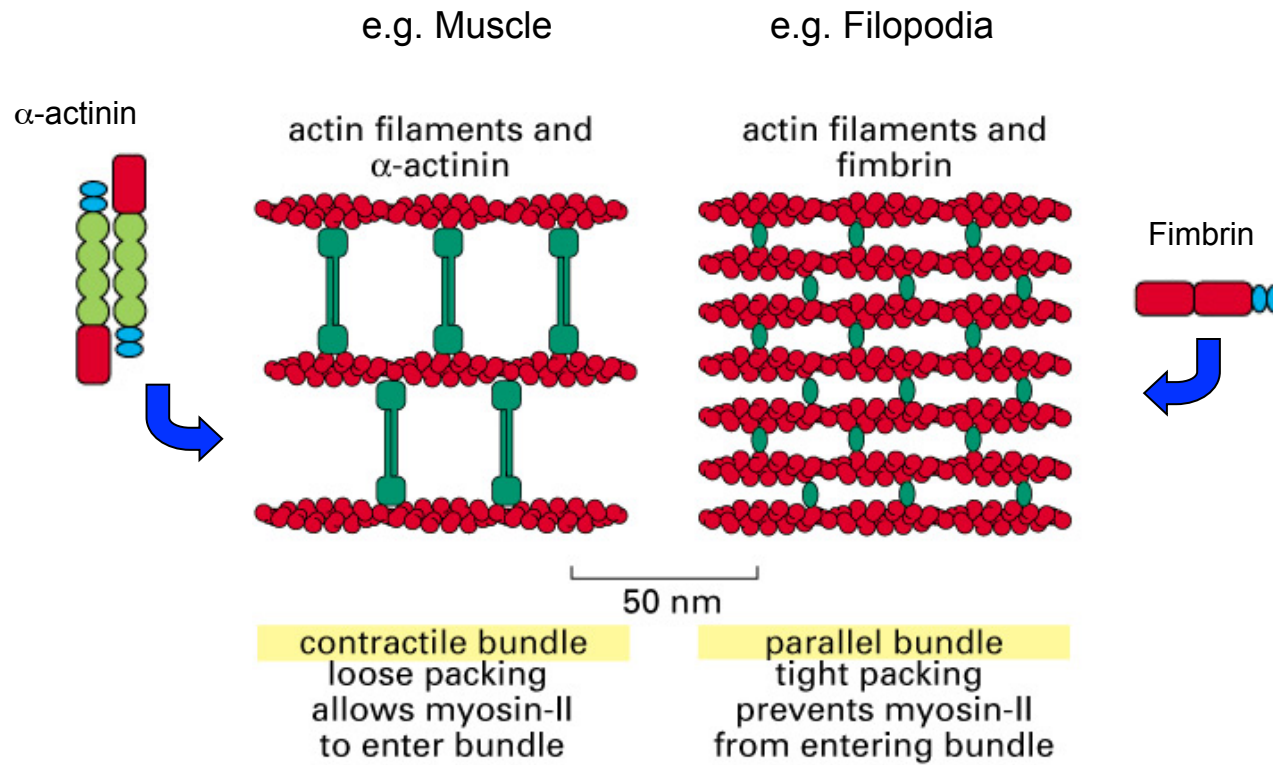


Cross-Linking Proteins and AFs



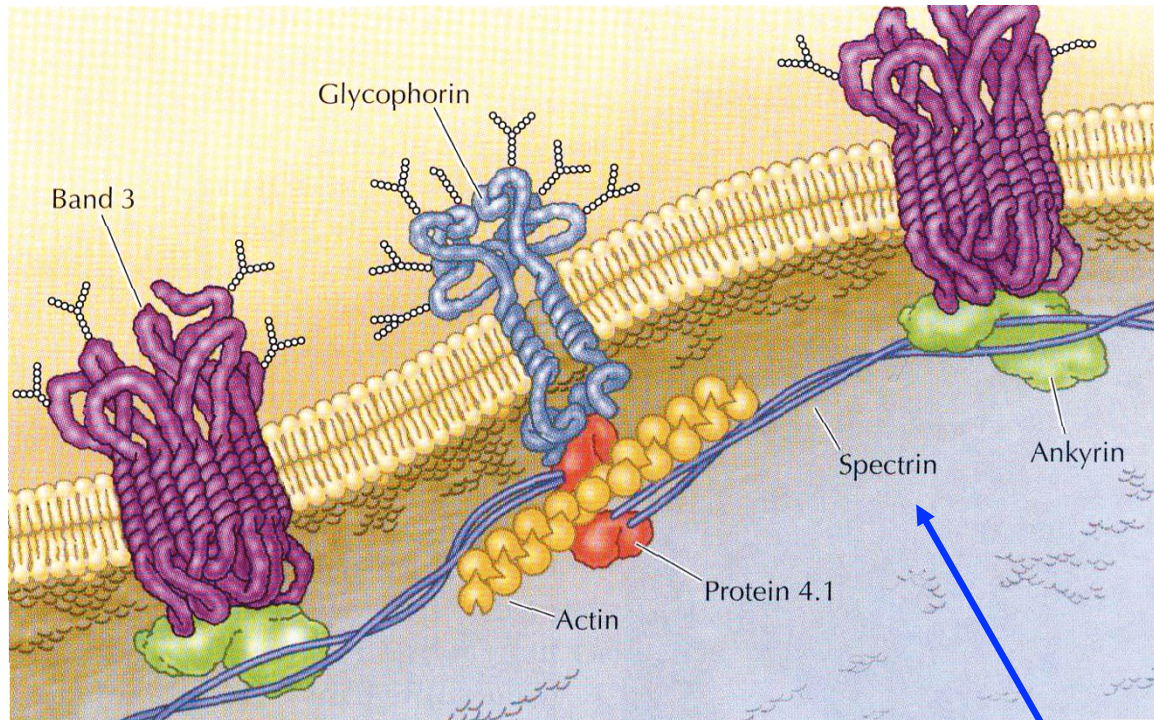
Red = actin binding domains

Actin Bundling Proteins



Gel-forming Proteins

Red blood cell membrane



Cooper and Hausman. The Cell.

- See also Fig. 10-41 in Alberts.
- Forms 2-D network.
- Gives RBC membrane flexibility.



spectrin (tetramer)



Gel-forming Proteins

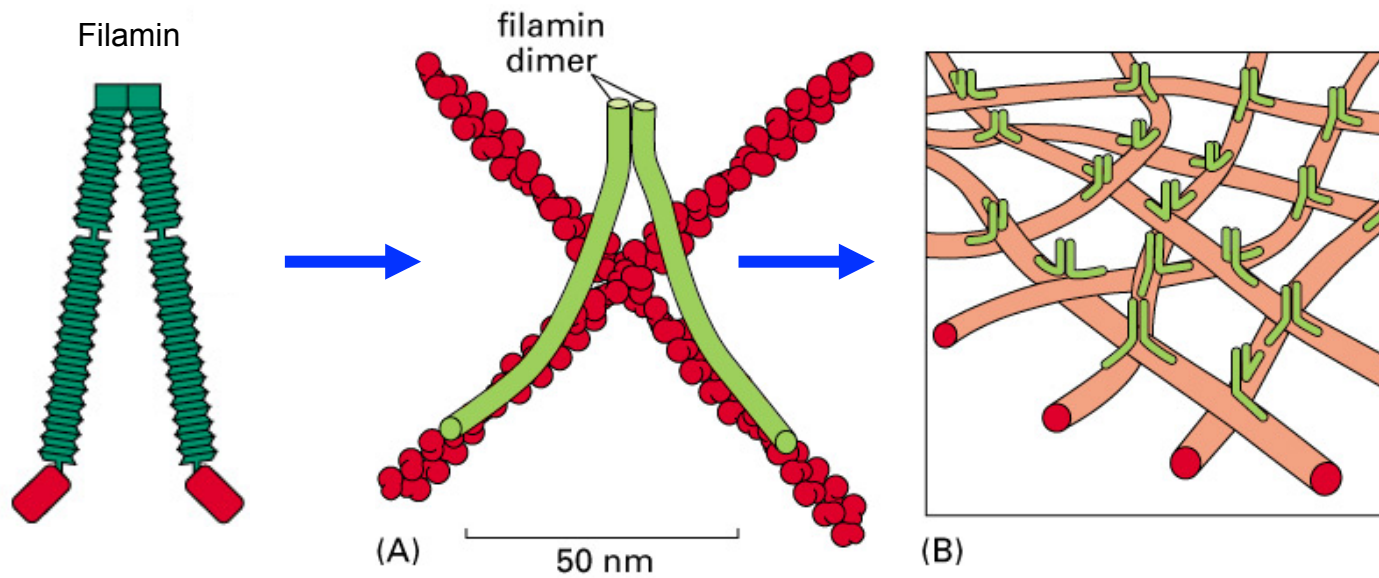


Figure 16-42. Molecular Biology of the Cell, 4th Edition.

e.g. lamellipodia in locomotion

Changes in Cell Shape During Embryonic Development

- Vertebrate embryo.
- Folding of neural tube during nervous system development.
- Cell height – MTs
- “Rolling” tube – AFs.

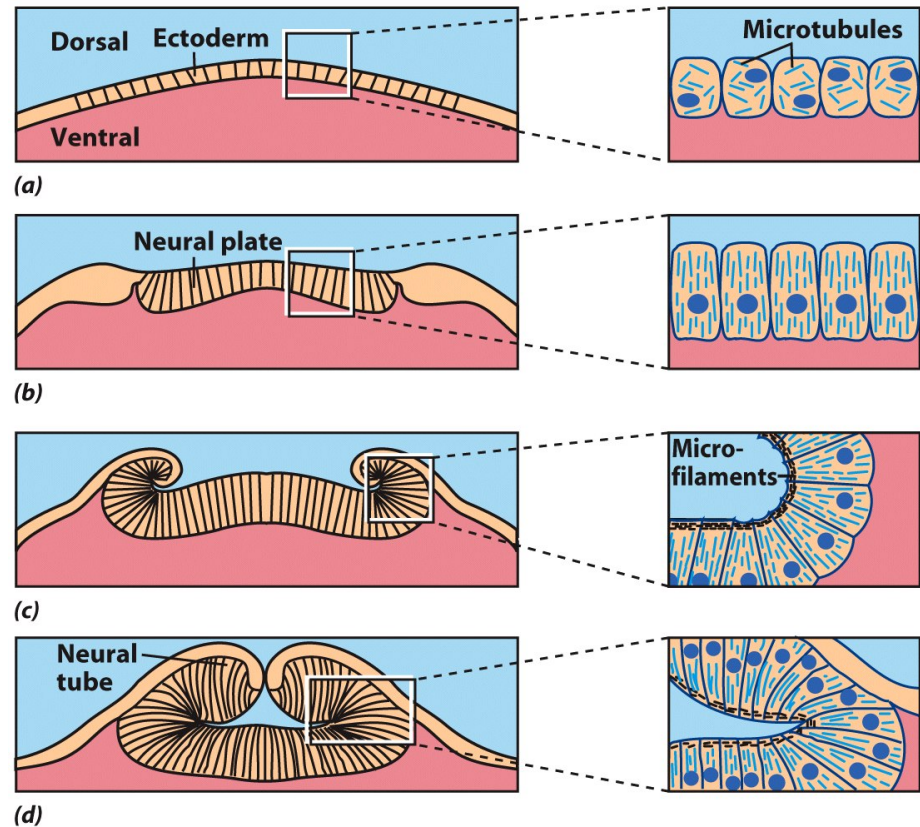


Figure 9-77a-d Cell and Molecular Biology, 5/e (© 2008 John Wiley & Sons)

Changes in Cell Shape During Embryonic Development

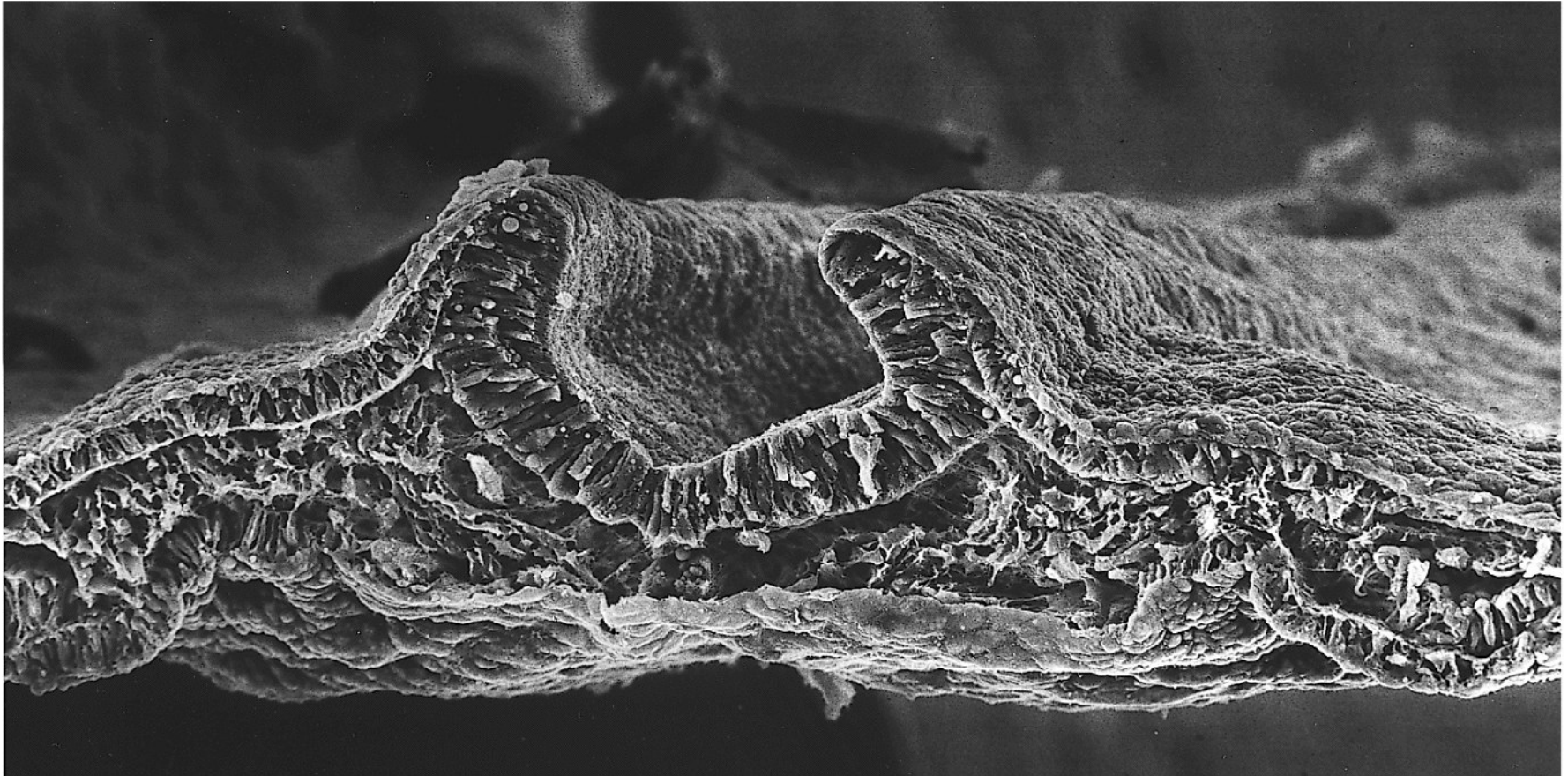


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Induction by Extracellular Signals

- Example, crawling neutrophil.
- Chemical activates WASP...
- Polymerizing filaments push membrane.

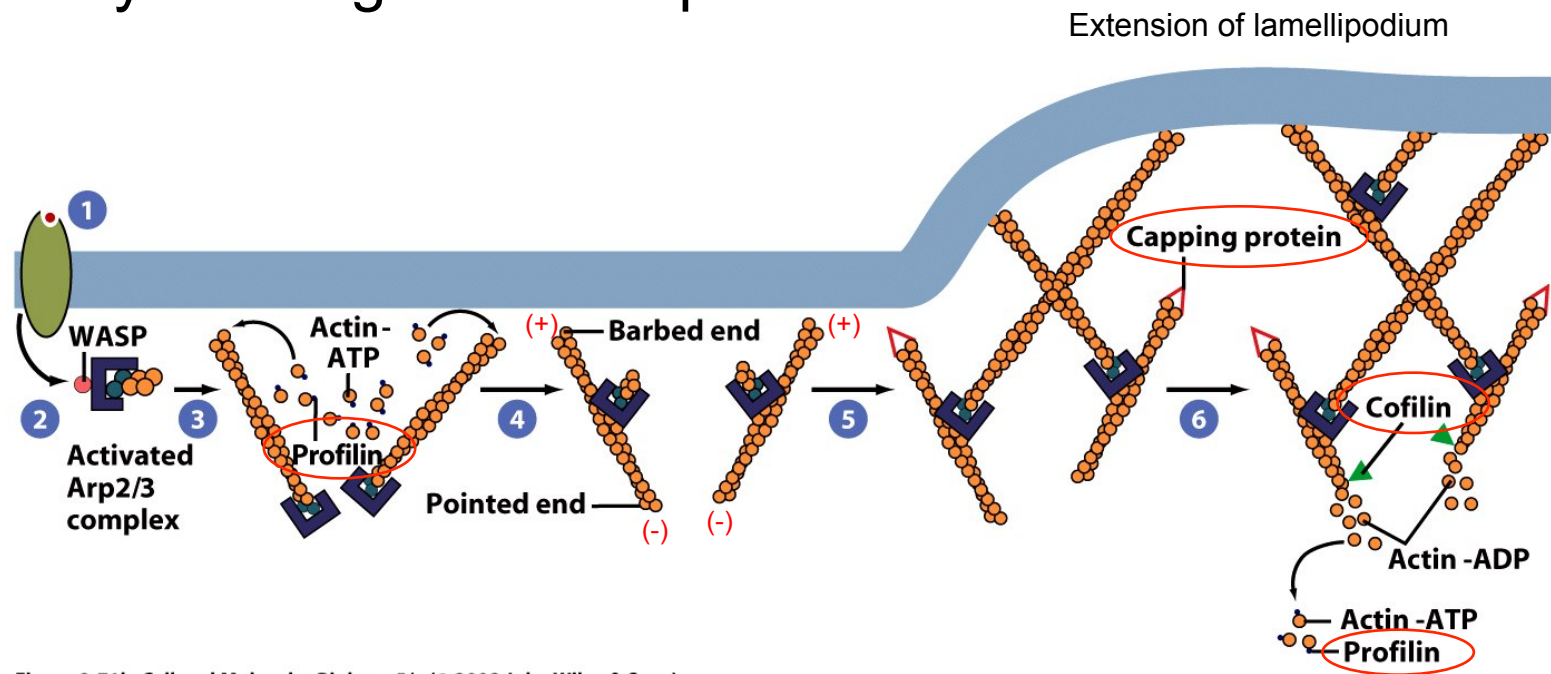


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WASP = Wiskott-Aldrich Syndrome Protein

Sequence of steps

1. Extracellular signal
2. Activation of WASP
3. Nucleation and branching by Arp2/3
4. Promotion of assembly by profilin
5. Elongation reduced by capping proteins
6. Destabilized by cofilin and return of subunits to pool

Things to Consider...

1. Think about how each accessory protein affects the stability of AFs and MTs?
2. Many of the dynamic mechanisms that we discussed today do not apply to IFs. Why?