

GCB Prelim Review

Cell 600

Theme III: Cell Motility and Adhesion

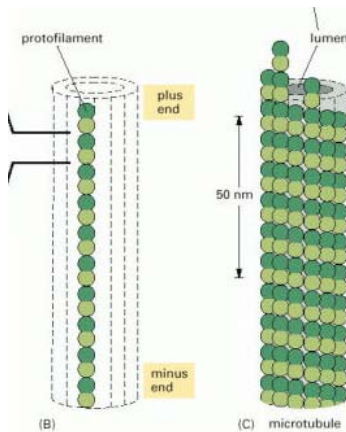
Based on Handouts from Fall 2003

Reference Chapter 16 in Alberts.

Microtubules and Dynamics

E. Holzbaur 10/29/03

Microtubules are made of alpha and beta tubulin, two related 50 kDa polypeptides. Both bind GTP, however, only Beta tubulin hydrolyzes it. Tubulin is an obligate dimer of alpha and beta subunits in the cell. Microtubules are formed when dimers arrange head to tail (thus microtubules are polar) into strands called protofilaments. Thirteen protofilaments form a hollow tube, which is the microtubule. Alpha tubulin is exposed at the minus end, Beta tubulin is exposed at the plus end.



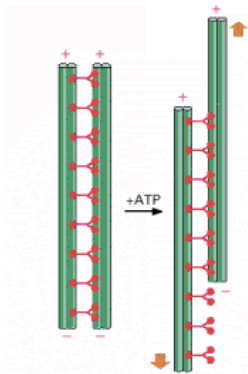
Microtubules are very similar to actin filaments. Both are polar, formed from subunits, have ends with different properties and neither requires an exogenous energy source for polymerization (GTP not required). However, microtubules are thicker and ~300 times stiffer. Also the cellular pool of unpolymerized tubulin is similar to the free subunit concentration in vitro at steady state.

Dynamic instability is the key property. GTP hydrolysis by the Beta tubulin destabilizes the end of the molecule. This causes it to depolymerize. This process happens rapidly. Thus microtubules can be observed growing and shrinking rapidly. It is thought that the microtubule grows as long as it has a GTP cap, loss causes rapid shrinkage.

Microtubules are heavily involved in cell transport, mitosis, cilia and flagella and neurons. Microtubule organizing centers are the origination point of microtubules. There are many proteins associated with microtubules, referred to as MAPs, these perform various functions including transport and enhancing stability. Taxol and nocodazole are two common drugs for manipulating microtubules.

Microtubule Motors

E. Holzbaur 10/31/03



These consist of kinesin (moves to + end) and dynein (moves to - end), these are common for force generation in cilia and flagella and both require ATP to operate. Dynein acts as a crossbridge between two microtubules and slides them to produce movement. Both act in the cytoplasm to transport proteins (vesicles, etc) along the microtubule network of the cell. This creates a very efficient delivery system. Both have similar structures, two heads each with long tails. The heads “walk” along the microtubule.

Actin and Myosin cytoskeleton

M. Ostap 11/3/03

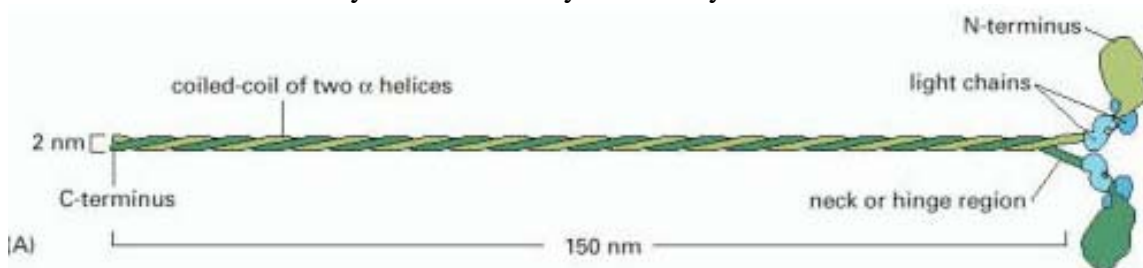
Intermediate filaments, these are less exciting and typically involved in maintaining cell integrity. They are not polarized.

Actin filament dynamics are involved in the locomotion of cells, they force protrusion, which results in forward movement. Actin is structurally polarized and has a barbed appearance. The pointed end is the minus end, and the barbed is the plus end. The barbed end is typically bound to ATP, while the pointed binds ATP. Actin is unique in that it “treadmills,” in that the barbed end is typically being extended while the pointed end is depolymerizing. The cell regulates both elongation and degradation in a effort to control the speed at which these processes take place.

Myosins

M. Ostap 11/5/03

Actin based motility relies on the myosin family of molecular motors.



Myosin is a dimer with a catalytic domain near the N-terminus. The light chains are responsible for the regulation of the catalytic activity of the dimer. ATP hydrolysis provides energy for myosin’s movement.

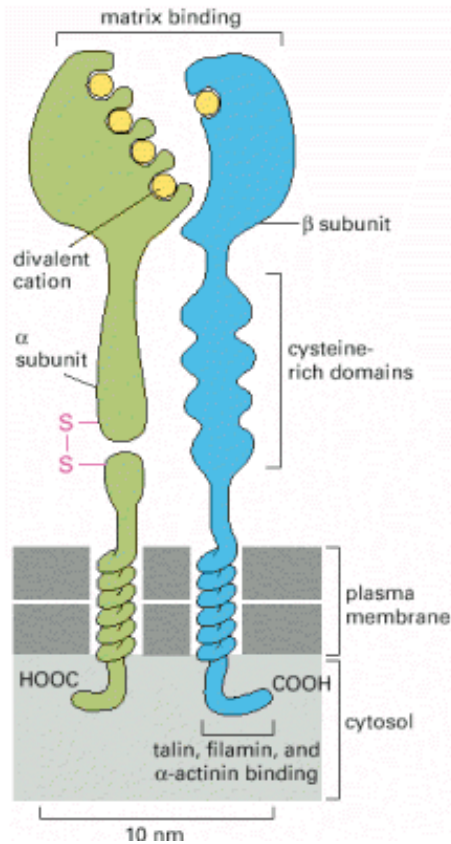
Myosin-II is critically important in muscles. It composes 60% of all muscle protein and is responsible for contraction. Many other myosins exist, with a diverse range of functions, some still unknown.

Cell Adhesion

D. Boettiger 11/7/03

There are many different cell adhesion molecules. The Ig super family (CAMs), Cadherins (Ca^{2+} dependent), selectins (bind sugars, immune system) and integrins. Integrins are primarily responsible for adhesion to the extracellular matrix (ECM). Adhesion molecules are involved in ligand recognition, both chemical and mechanical. They are also involved in regulated adhesion, otherwise known as inside-out signaling. This regulation has both a temporal and spatial component, and is particularly critical for migration. Adhesion molecules are also required for cytoskeletal connection. Adhesion requires these connections, as the plasma membrane provides little mechanical strength. Adhesion also controls cell function, this is known as outside-in signaling. Adhesion receptors participate in crosstalk, such that signals from one adhesion event can change other adhesion events. Finally, adhesion controls cell and tissue structure by providing anchor points for the cytoskeleton.

There are numerous different types of adhesion molecules and mechanisms. A good place to review would be Alberts chapter 19, pages 1065-90 for further info.



The key with ECM and interactions is that signaling based on integrin interaction with the ECM is critical to many biological processes. Genetic knockouts of specific parts of this signaling process can lead to developmental arrest. The ECM is composed of many different parts including: collagens (which are assembled into fibrils) and GlycosAminoGlycans (GAG's, form a hydrated gel). The basal lamina is constructed of ECM, it is a sheet which underlies epithelia, and surrounds muscle fibers and blood vessels.

Integrins are primarily responsible for adhesion to ECM. There are many different types each made of an alpha and a beta chain. There are currently about 17 known alpha chains and 8 known beta chains. Each combination has a different ligand, though some overlap. Integrin mediated adhesion is controlled by intracellular processes (inside-out signaling). As such integrins are connected to the cytoskeleton in order to provide increased mechanical strength and a means of transmitting the properties of the environment to the cell. The cytoplasmic domain is typically short. Integrin

mediated outside-in signaling is a popular topic! For a deeper review see Alberts chapter 19 pages 1090-1118.

Mitosis and Cytokinesis

This is best reviewed by going over the general process of mitosis with an emphasis on the cytoskeleton. This has been summarized to the extent we need elsewhere I believe.