Cytoskeleton

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Three Major Filamentous Systems of the Cytoskeleton

<table>
<thead>
<tr>
<th>Cytoskeletal Element</th>
<th>Description (diameter)</th>
<th>Protein Composition</th>
</tr>
</thead>
<tbody>
<tr>
<td>microfilaments</td>
<td>thin filaments (6-7 nm)</td>
<td>actin + associated proteins</td>
</tr>
<tr>
<td>microtubules</td>
<td>tubular structures (25 nm)</td>
<td>tubulin + associated proteins</td>
</tr>
<tr>
<td>intermediate filaments</td>
<td>rope-like fibers (~10 nm)</td>
<td>IF proteins</td>
</tr>
</tbody>
</table>

Actin and Microfilaments

- highly conserved globular protein (G-actin)
- polymerizes into thin filaments (F-actin)
- exhibits polarity (+ and – ends)
- (barbed and pointed ends)
- filaments stabilized by other proteins

Actin Polymerization

- requires nucleation (activation)
- elongation primarily at “+” end (= barbed end)
- ATP/ADP
  - G-actin has bound ATP
  - ADP-actin more likely to dissociate
- actin-binding proteins
  - monomer sequestration (eg., profilin)
  - capping proteins (eg., gelsolin)
- cellular regulation
  - rho family (ras-like G-proteins)
  - trimeric G-proteins
- drugs
  - cytochalasins (prevent assembly)
  - phalloidin (prevent disassembly)
Examples of Actin-Binding Proteins
- tight bundles
- loose bundles
- 3-D gels

Membrane Interactions
- submembrane cytoskeleton (e.g., RBC)
- focal adhesions (ECM interactions)
- adherens junctions (cell-cell interactions)

Focal Adhesions

Myosin and Force Generation
- large family of proteins (16)
- actin-activated ATPase
- converts chemical energy into mechanical energy

Ameboid Movement
- reorganization of cytoskeleton in pseudopodia
- force for protrusion?
- force for traction?
Microtubules

**Major Roles:**
1. Mechanical/Cell Shape
2. Mitotic Spindle
3. Cilia/Flagella

**Tubulin:**
- highly conserved
- heterodimer ($\alpha$, $\beta$)
- in vivo assembly from MTOC

Microtubule Organizing Centers

- centrioles
  - aka basal bodies of flagella
  - barrel-shaped (triplets of $\mu$T)
- centrosomes
  - amorphous matrix ($\pm$ centrioles)
  - organizes cytoplasmic $\mu$T array
  - forms spindle during cell division
- spindle pole bodies
  - located on nuclear membrane
  - forms mitotic spindle in many lower eukaryotes
Flagellar Movement

Motor Proteins

Kinesins

Kinesins and Mitosis

- large superfamily of proteins
  - defined by ‘motor’ domain
- tail regions highly divergent (function)
- chemomechanical cycle
  - similarities to myosin
  - two motors needed

- reorganization of μT during mitosis
  - disassembly of cytoplasmic μT
  - assembly of spindle apparatus
- duplication of centrosome to form spindle apparatus
- at least 4 kinesins implicated
  - separation of poles
  - migration of chromosomes
possible kinesin roles:
• stabilize metaphase plane
• sliding μT to drive poles apart
• pull asters toward membrane
• kinetochore: chromosome movement

Intermediate Filaments
• rope-like fibers extending from nucleus periphery
• extremely stable
  – resistant to detergent extraction
• only in metazoa
• subunits are part of large multigene family
  – related to nuclear laminas
  – tissue specific expression

Intermediate Filament Proteins

<table>
<thead>
<tr>
<th>Type</th>
<th>Examples</th>
<th>Cell Type</th>
<th>Size (kDa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>acidic keratins</td>
<td>epithelial</td>
<td>40-60</td>
</tr>
<tr>
<td>II</td>
<td>basic-neutral keratins</td>
<td>epithelial</td>
<td>50-70</td>
</tr>
<tr>
<td>III</td>
<td>vimentin</td>
<td>mesenchymal</td>
<td>53</td>
</tr>
<tr>
<td></td>
<td>desmin</td>
<td>muscle</td>
<td>52</td>
</tr>
<tr>
<td></td>
<td>GFAP</td>
<td>glial cells</td>
<td>51</td>
</tr>
<tr>
<td>IV</td>
<td>neurofilament</td>
<td>neurons</td>
<td>57-150</td>
</tr>
<tr>
<td>V</td>
<td>laminins (A, B, C)</td>
<td>nuclear lamina</td>
<td>60-70</td>
</tr>
</tbody>
</table>

head | heptad repeats | tail

IF Proposed Structure

IF Function
• mechanical support
  • abundant in cells and structures under mechanical stress (eg, epithelia, hair, nails, muscle, etc)
  • genetic defects linked to keratin: epidermolysis Bullosa simplex, epidermolytic hyperkeratosis
  • links to membranes and other cytoskeletal elements
• specialized function in differentiated cells
  • tissue specific expression
  • markers for tumor diagnosis