Midterm II Review

What we will do today:

- Midterm format (brief).
- Review of selected topics from recent lectures.
- Questions.

Note, Midterm II covers material from Lectures 9-15 only

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Midterm Format

- Approximately 1 hour.
- There will be 2 parts to the exam:

  Part A: Multiple choice (20 x 1 marks)

  Part B: Long answer (2 x 10 marks)
  - About one page for each answer.
  - You may use point form.
  - Do not include excessive writing.
  - Diagrams may be included but they must be accompanied by text.

2

Things to Consider...

1. Are you familiar with the roles of each different type of MT during mitosis?
- *Astral, kinetochore, overlap.*

2. Motor proteins?
- *Cross-linking by motor proteins creates spindle elongation and foci at spindle poles.*
- *KRPs push and dyneins pull overlap MTs.*

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Components of the Mitotic Spindle

1. Microtubules
- Astral
- Kinetochore
- Overlap

2. Motor proteins
- Kinesin-related (+)
- Dynein (-)

3. Chromosomes (chromatids)

4. Centrosome
- Centrioles
- PCM

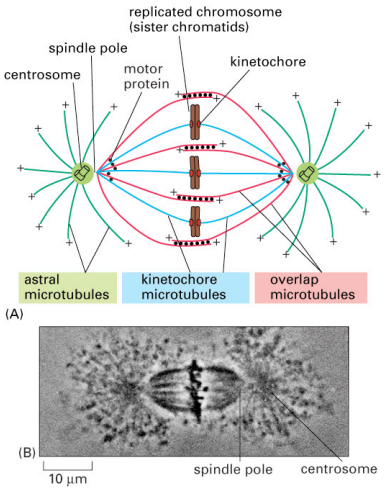

Figure 18–10. Molecular Biology of the Cell, 4th Edition.

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Anaphase A Separation

- Poleward (-) movement of chromatids by shortening of kinetochore MTs.

1. MT depolymerization at (+) ends by KRPs.
2. Continual loss of tubulin at (-) ends (i.e. as in poleward flux) without addition at (+) ends.

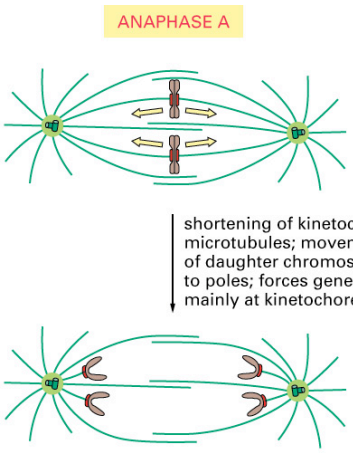

Figure 18–26 part 1 of 2. Molecular Biology of the Cell, 4th Edition.

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Anaphase B Separation

- Pulling by motor proteins at poles.
- Pushing by motor proteins at central spindle (i.e. at overlap MTs).

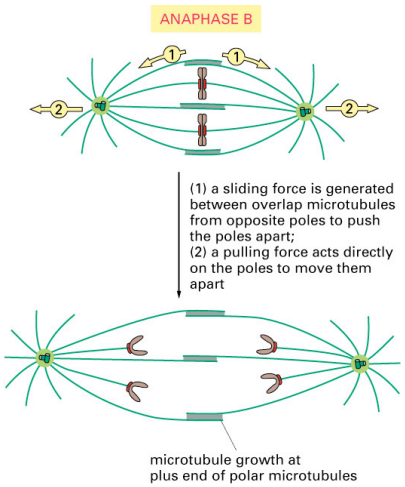

Figure 18–26 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

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Things to Consider...

1. How do signal sequences differ between import/export from cytosol to nucleus, and translocation across the mitochondrial membrane?

- *Mitochondrial signal sequence is modified as an α helix. (+) charges clustered on one side.*
- *A nuclear localization signals is a terminal signal sequence or a signal patch.*

2. How do nuclear pore complexes and translocator complexes differ?

- *Nuclear: large, proteins folded, receptors bind to nucleoporins, GTP hydrolysis.*
- *Mitochondria: small, proteins unfolded.*

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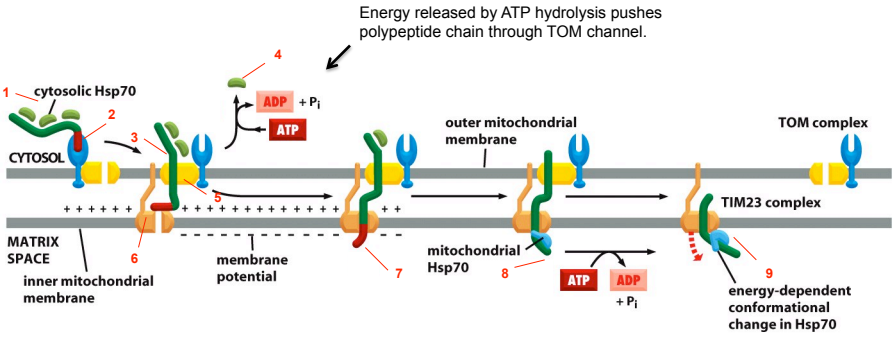

Note: The TOM complex is composed of multiple proteins, such as smaller receptor proteins (TOM 20 and TOM 5) and a pore-forming region (TOM 40).

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Figure 12-26 *Molecular Biology of the Cell* (© Garland Science 2008)

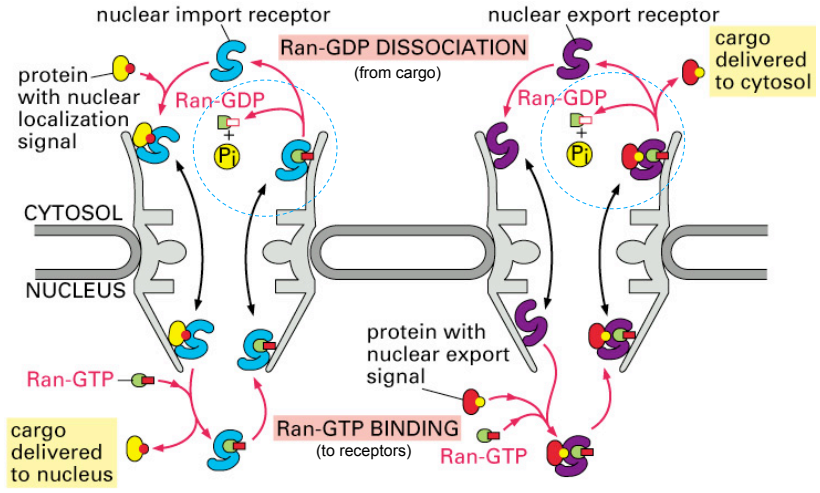

Figure 12–16. Molecular Biology of the Cell, 4th Edition.

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Things to Consider...

1. Why are molecular chaperones not needed for delivery of polypeptide chains to the ER?
   - *Because in most cases translocation to the ER lumen is cotranslational.*

2. What is the difference between posttranslational and cotranslational translocation?
   - *Translocation after or during translation.*
   - *Free or membrane-bound ribosomes.*
   - *Chaperones.*

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Double-Pass Membrane Proteins

- Internal signal sequence initiates translocation.
- Stop-transfer signal stops translocation.
- Both sequences are α helices and are released to the membrane.

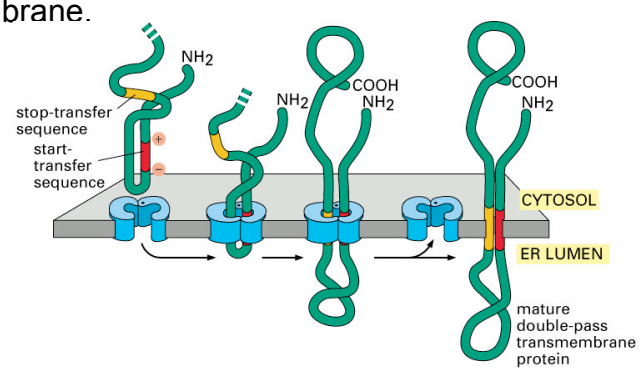

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Figure 12–49. Molecular Biology of the Cell, 4th Edition.

Things to Consider...

1. Can you describe the path of a protein from ribosome to plasma membrane?
   - *Translocation, budding, fusion by SNAREs, Golgi stack, secretory pathways etc.*

2. Why would cells have a regulated secretory pathway in addition to a constitutive one?
   - *These have very different functions, either maintain membrane or mediate secretion to cell exterior.*

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2 Secretory Pathways

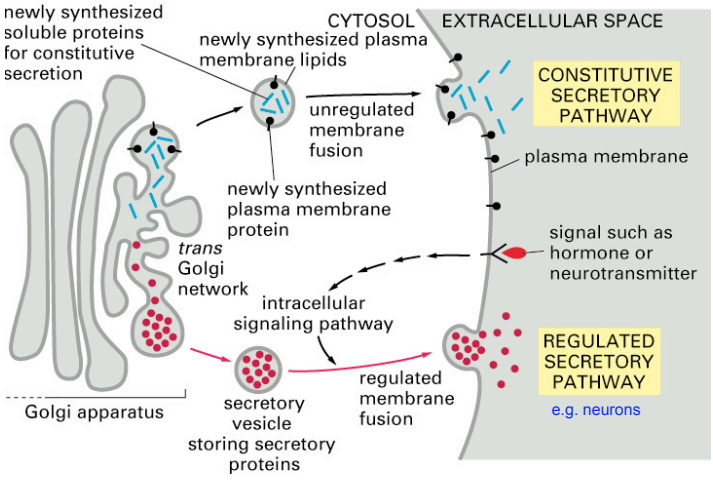

Figure 13–54. Molecular Biology of the Cell, 4th Edition.

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Things to Consider...

1. Though lipids primarily provide structure and fluidity to the membrane bilayer, they also play functional roles.
   - *Activated transmembrane receptors can induce phosphorylation (by PI-3 kinase) or cleavage (by PLC) of inositol phospholipids and initiate signalling pathways.*

2. Be familiar with the different classes of membrane proteins.
   - *Integral, peripheral, and lipid-anchored.*

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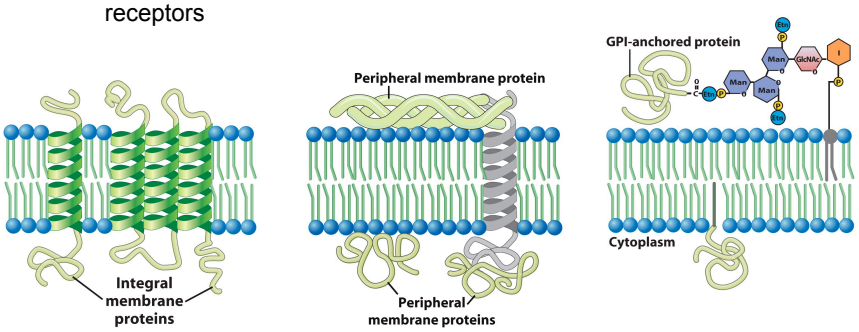
3 Types of Membrane Proteins

1. Integral Proteins
   e.g. ion channels, transporters, receptors

2. Peripheral Proteins
   e.g. spectrin, ankyrin

3. Lipid-anchored Proteins
   e.g. GPI, prenyl, or fatty acid-anchored



Karp 2008

Note: I prefer this more general classification, rather than that of Fig. 10-17 in Alberts.

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Things to Consider...

1. Be familiar with the physiological roles that each transporter can perform.
   - *Maintain Na^+ and K^+ gradients, keep cytosolic Ca^{2+} low, drive ATP synthesis.*

2. As well, you should know the energy source behind each transporter (e.g. ATP, phosphorylation, concentration gradient, electrochemical gradient).
   - *e.g. ABC transporters, P-type, antiporters etc.*

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Transport ATPases (Pumps)

Class of ATPase	Solute	Location	Function
P-type ATPase	Na^+ and K^+	Plasma membrane	Pumps Na^+ out and K^+ into cytosol
	Ca^{2+}	Plasma membrane	Pumps Ca^{2+} out of cell
	Ca^{2+}	Sarcoplasmic reticulum (SR)	Pumps Ca^{2+} into SR
V-type ATPase	H^+	Membrane of secretory vesicles	Pumps H^+ into vesicle lumen
F-type ATPase	H^+	Mitochondrial IM	H^+ gradient drives ATP synthesis
ABC transporters	Many (ions, sugars, amino acids, proteins)	Plasma membrane	Nutrient uptake, protein export.

Modified from Becker et al.

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Things to Consider...

1. What sets ion channels apart from transporters?
   - *No direct energy source required, ions move faster through channels, channels form aqueous pores...*

2. Explain how ion channels can maintain such high ion selectivity while conducting ions at nearly the same rate as ion diffusion in solution.
   - *e.g. loss of hydration shell, channel structure consumes potential binding energy, K^+ ions interact weakly with selectivity pore and most other ions are excluded, repulsive forces between K^+ ions reduce interaction with pore.*

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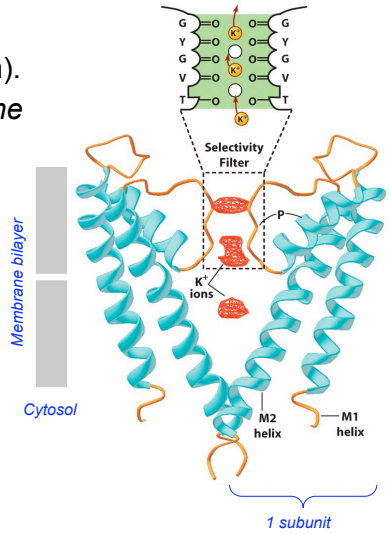
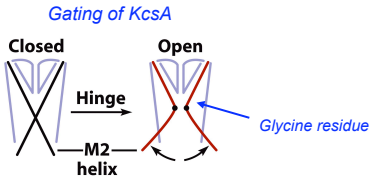
KcsA K^+ Channel

- Composed of *4 subunits* (2 shown).
- Each subunit has *2 transmembrane helices*. *M2* controls gating.
- And *1 pore-forming segment* composed of a *helix* and a *non-helical loop* that forms the *selectivity filter*.





Modified from Karp 2008

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Good luck!

Questions?

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