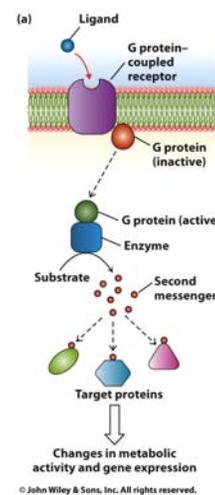


Signal Transduction Pathways

Pratt & Cornely, Chapter 10

Terms for Signal Transduction

- Ligand (first messenger)
- Receptor (transducer)
- Primary Effector
- Second messenger
- Second effector, etc.
- Target proteins/DNA



Ligands

- Hormones vs Local mediators
- Polar (insulin) vs nonpolar (steroidal hormone)
- Specific—high affinity
- Agonist vs antagonist

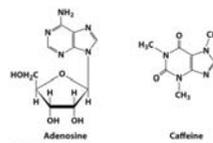


TABLE 10-1 Examples of Extracellular Signals

Hormone	Chemical Class	Source	Physiological Function
Auxin	Amino acid derivative	Most plant tissues	Promotes cell elongation and flowering in plants
Cortisol	Steroid	Adrenal gland	Suppresses inflammation
Epinephrine	Amino acid derivative	Adrenal gland	Prepares the body for action
Erythropoietin	Polypeptide (165 residues)	Kidneys	Stimulates red blood cell production
Growth hormone	Polypeptide (19 residues)	Pituitary gland	Stimulates growth and metabolism
Nitric oxide	Gas	Vascular endothelial cells	Triggers vasodilation
Thromboxane	Eicosanoid	Platelets	Activates platelets and triggers vasoconstriction

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Quantitative Ligand Binding

- K_D values
- Problem 7:
Derive an expression for the $[RL]/[R]_T$ ratio.

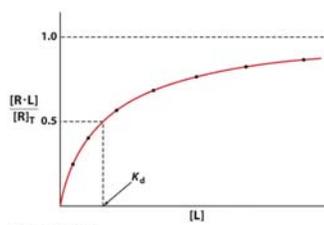
$$K = \frac{[R][L]}{[RL]} \text{ and } [RL] = \frac{[R][L]}{K}$$

$$\% \text{ bound} = \frac{[RL]}{[R] + [RL]}$$

Substitute:

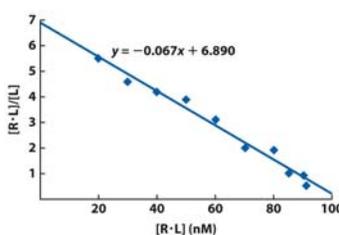
$$\% \text{ bound} = \frac{\frac{[R][L]}{K}}{[R] + \frac{[R][L]}{K}} = \frac{[R][L]}{K[R] + [R][L]} = \frac{[L]}{K + [L]}$$

Hyperbolic function!



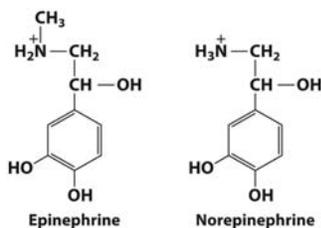
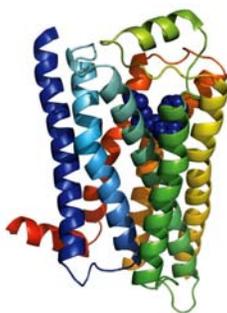
Scatchard Plot

- Problem 14: A Scatchard Plot is another method of representing ligand binding data. The slope is equal to $-1/K_D$. Use the chart to estimate K_D for calmodulin binding to calcium.



G-Protein Signaling Pathways

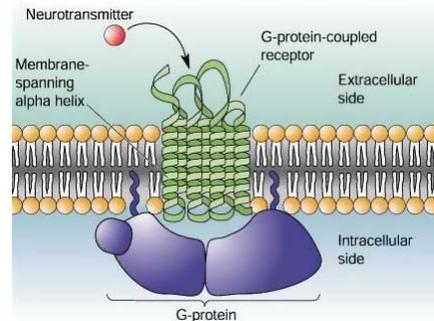
- Use β -adrenergic receptor as example of G-Protein Coupled Receptor (GPCR)
- 7-transmembrane (7-TM) receptor



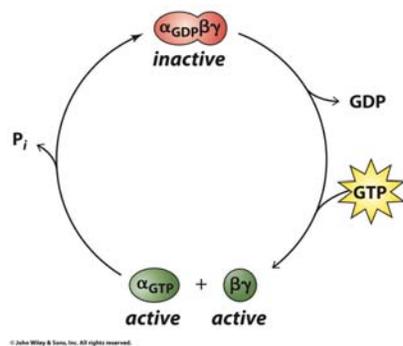
β -blockers: antagonist lowers BP

G-Protein Coupled

- Ligand binding causes trimeric G-protein to associate with receptor (figure not quite right)
- Three subunits, lipid anchored
 - α binds GDP
 - β, γ tightly associated
- Binding causes GDP release



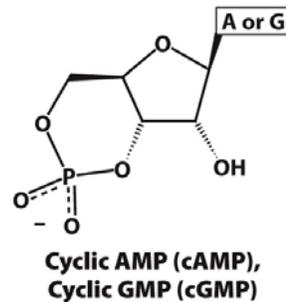
G-Protein Activation



- GTP binds
 - Destabilized trimer
 - Release each other and receptor as two active proteins
- Turn off: Slow GTP hydrolysis
 - Subunits reassemble to inactive form until they can bind receptor again

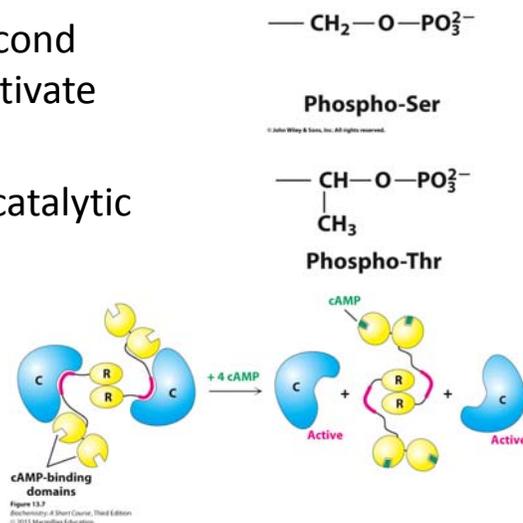
cAMP

- G-protein carries signal to another protein:
- Adenylate cyclase
- Catalyzes formation of cAMP
 - second messenger
- Amplification



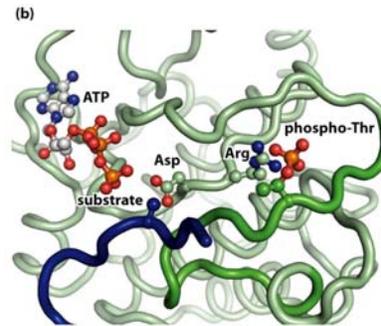
Protein Kinase A

- cAMP acts as second messenger to activate Protein Kinase A
- Regulatory and catalytic subunits
- Kinase!



Covalent Modification

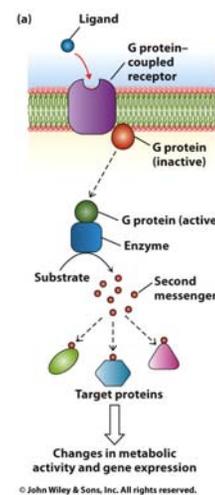
- Common activation/ deactivation strategy
- Changes protein conformation drastically
- Middle range time effect
- PKA activates the enzyme that releases glucose from storage



PKA is, itself, regulated by phosphorylation
 -Phosphorylation activates PKA
 -Positions Asp near substrates (ATP and blue peptide)

Protein Kinase A

- Exercise: use basic guide to explain mechanism of epinephrine affect on sugar release in muscle



Turning Off Pathway

- Can turn it off at any point
 - Receptor: ligand dissociates
 - G-protein: GDP formed
 - Second messenger: hydrolysis
 - Phosphorylated enzyme: phosphatase

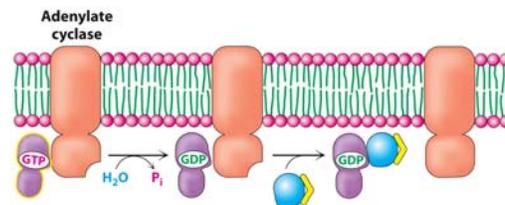
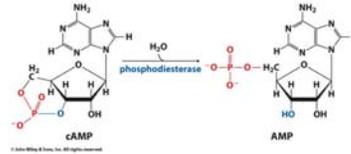
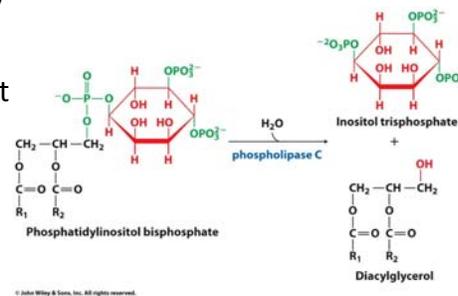


Figure 15.8
Biochemistry, 4th Edition
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Phosphoinositol Pathway

- Many G-Proteins for many pathways
 - Cross-talk—different paths can give same result
- Or same hormone can give different responses
 - α -adrenergic receptor (liver but not muscle)
 - Liver also has glucagon binding, so α -receptor allows for fine-tuning of signal
 - Target of this G-protein is phospholipase C



Two second Messengers

- $\text{PIP}_2 \rightarrow \text{IP}_3$
 - Opens Calcium gates
 - Activates Protein Kinase B (Akt) to make other second messengers
- $\text{PIP}_2 \rightarrow \text{DAG}$
 - Activates Protein Kinase C
 - Also requires Ca^{+2}
 - Especially important in cell division

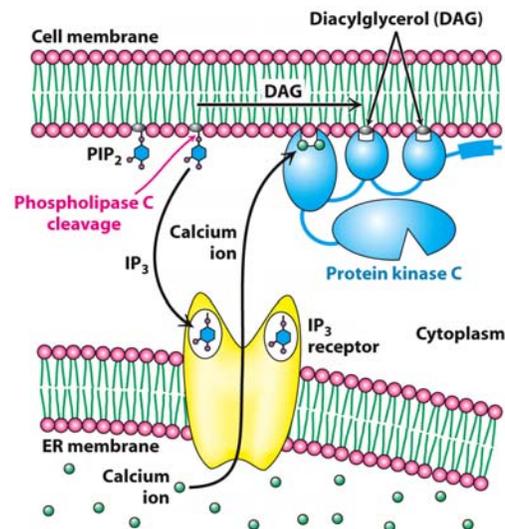
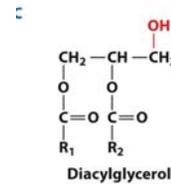
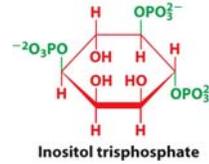
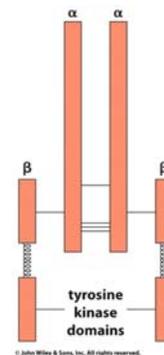


Figure 13.12
Biochemistry: A Short Course, Third Edition
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Receptor Tyrosine Kinases

- Second major class of receptors
 - Insulin binding as prototype
 - Mostly monomers that bind ligand and then dimerize
 - One subunit binds ligand
 - Second subunit become active kinases



Insulin Signaling

- Receptor Tyrosine Kinases
 - Dimerization and autophosphorylation
 - Adaptor proteins
 - Kinase cascade

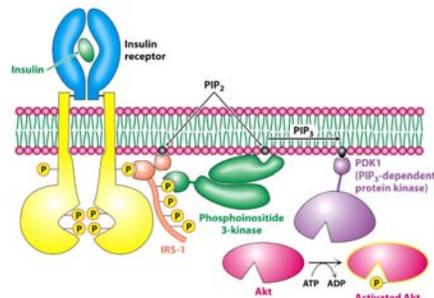
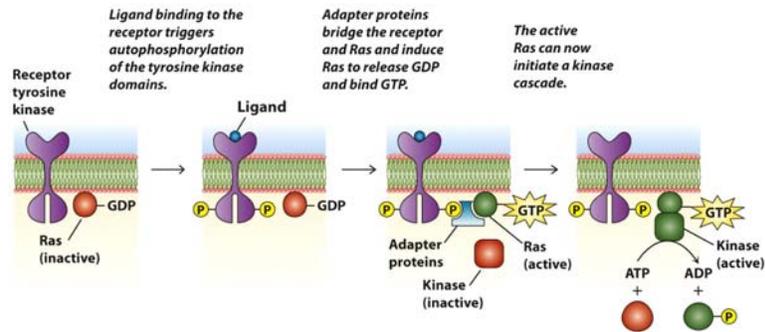


Figure 14.21
Biochemistry, Seventh Edition
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Epidermal Growth Factor

- Small G-protein: Ras



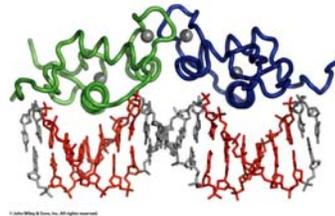
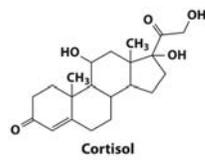
Oncogenes

- Ras targets nuclear proteins
- Key signal in cell growth
- **Problem 46:** Mutant Ras proteins have been found to be associated with various types of cancer. What is the effect on a cell if the mutant Ras is able to bind GTP but is unable to hydrolyze it?

Pathology

- Cholera
 - Covalent modification of a G-protein
 - Constitutively active
 - Opens chloride channel; leads to severe diarrhea
- Whooping cough
 - Toxin turns off an inhibitory G-protein
 - Adenylate cyclase remains active

Lipid Hormone Signaling



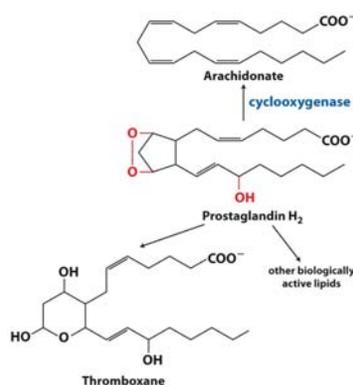
- Cortisol binds Zinc finger at ligand binding domain
 - dimerization
- DNA binding domain is zinc finger
- Zn finger dimer binds at the hormone response element
- Transcription factor—activate or inhibit
- Steroidal anti-inflammatory

Problem 53

- Steroid hormone receptors have different cellular locations. The progesterone receptor is located in the nucleus because it possesses a nuclear localization signal. This receptor only interacts with DNA once progesterone has bound. But the glucocorticoid receptor is located in the cytosol and does not move into the nucleus until its ligand is bound. Propose a role for glucocorticoid in this pathway that is distinct from progesterone.

Local Mediators

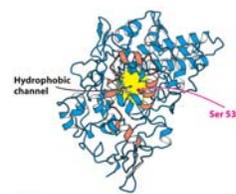
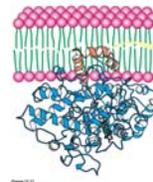
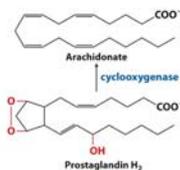
- Eicosanoids produced in response to cellular event
- Produce hormone-like responses in blood pressure, inflammation response, etc.



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Effect of Aspirin

- Arachidonate from membrane, travels through cavity in Prostaglandin H₂ synthase
- Aspirin covalently modifies Serine in cyclooxygenase active site
- COX-1 and COX-2 inhibitors



COX Targets

- NSAIDs (non-steroidal anti-inflammatory drugs) target cyclooxygenase
- Aspirin, ibuprofen: COX 1 and 2
- Vioxx: COX 2
- Acetaminophen: COX:3
 - Less side effects
 - Worse toxicity

