细胞信号转导（Cell Signaling）

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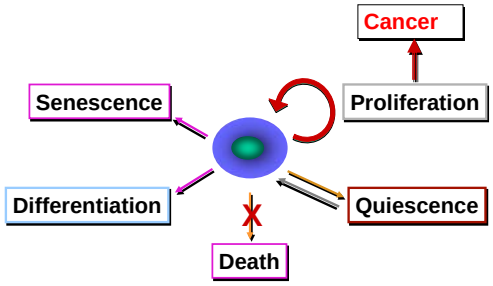
Fates of a cell



General introduction
- Common features of signal transduction
- Cell surface signal transducers, receptors

Ion channels
Secondary messengers
- cAMP
- cGMP
- Lipids
- Calcium

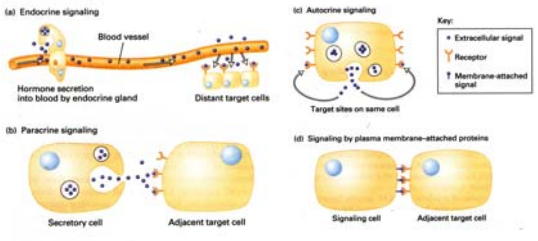
G proteins
- Trimeric G proteins
- Monomeric G proteins, Tyr kinase/MAP kinase

Protein modules

What is “Signal Transduction”?

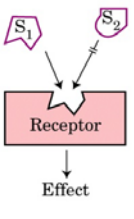
This expression first made its mark in the biological literature around 1974. Physical scientists and electronic engineers had earlier used the term to describe the conversion of energy or information from one form into another. Signal transduction at the cellular level refers to the movement of signals from outside the cell to inside; cascade of information from the plasma membrane to the nucleus in response to an extracellular stimulus in living organisms.

General schemes of intercellular signaling



Four basic features of signal-transducing systems

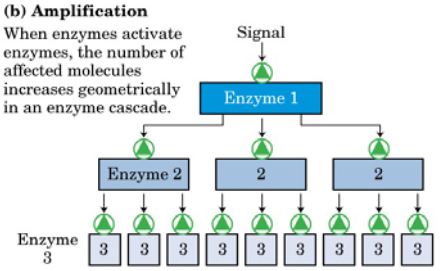
(a) Specificity
Signal molecule fits binding site on its complementary receptor; other signals do not fit.



Four basic features of signal-transducing systems

(b) Amplification

When enzymes activate enzymes, the number of affected molecules increases geometrically in an enzyme cascade.



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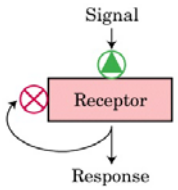
Four basic features of signal-transducing systems

(c) Desensitization/Adaptation

Receptor activation triggers a feedback circuit that shuts off the receptor or removes it from the cell surface.



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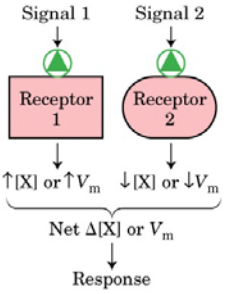
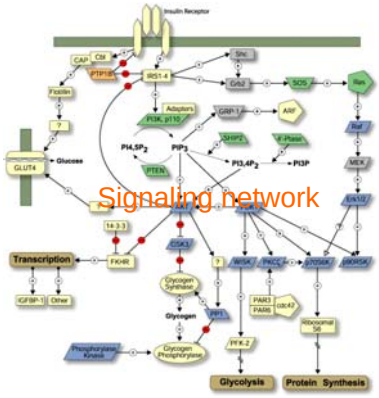
Four basic features of signal-transducing systems

(d) Integration

When two signals have opposite effects on a metabolic characteristic such as the concentration of a second messenger X, or the membrane potential V_m, the regulatory outcome results from the integrated input from both receptors.





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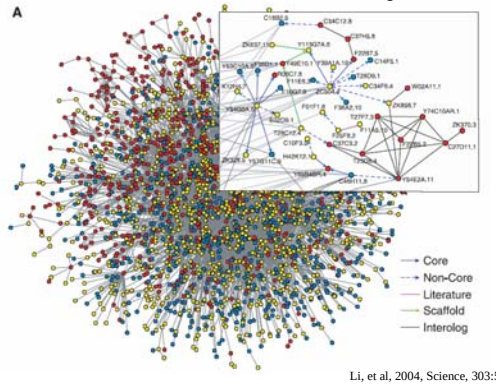
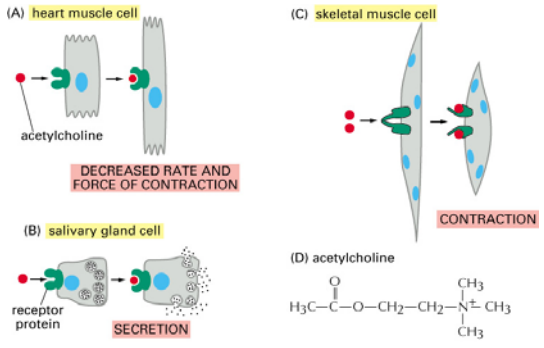
Protein interactome network in C. elegans

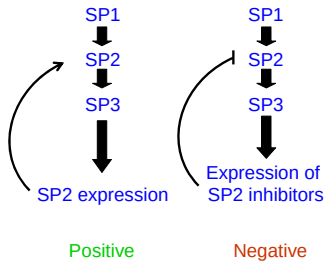


Li, et al, 2004, Science, 303:540

Context-specificity of cell signaling: Different cells respond differently to the same extracellular signal



Positive and negative feedback mechanisms



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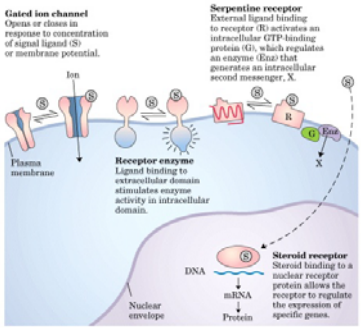
Four general types of signal transducers



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Ligand-activated cell-surface receptors

- Ion-channel receptors: acetylcholine
- G-protein-coupled receptors: epinephrine, glucagon, serotonin
- Tyrosine kinase-linked receptors: interferons
- Tyrosine kinase receptors: EGF, PDGF, insulin
- Tyrosine phosphatase receptors: CD45
- Serine/therinine kinase receptors: TGFβ, BMP
- Guanylate cyclase receptor: atrial naturetic factor

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Protein modules

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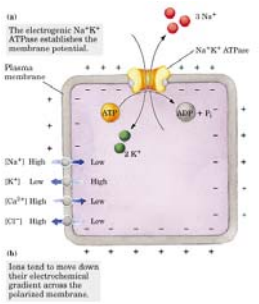
Transmembrane electrical potential



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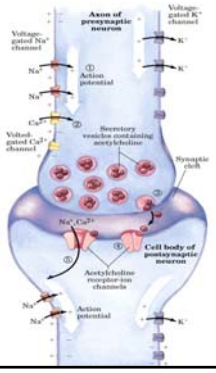
Role of voltage-gated and ligand-gated ion channels in neural transmission



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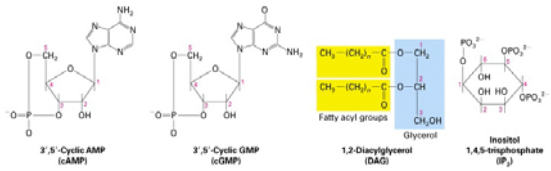
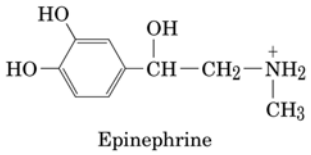
Second messengers



Secondary messengers:

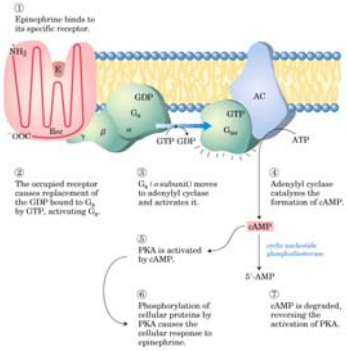
cAMP
cGMP
Lipids
Calcium



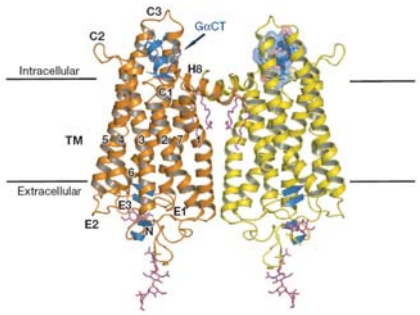
Epinephrine

Produced by Chromaffin cells in adrenal medulla
Functions:
- Increase heart rate, blood pressure, sweating, rate of respiration
- Stimulate conversion of glycogen to glucose

Transduction of the epinephrine signal: the β-adrenergic pathway

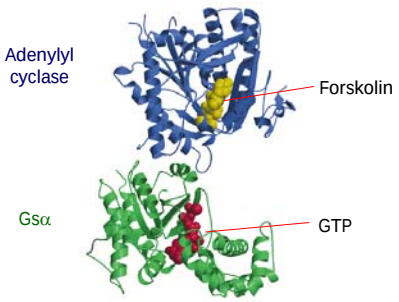


Structure of Opsin associated with the C-terminal Gsα



Scheerer et al, Nature, 2008

Interaction of $G_{s\alpha}$ with adenylyl cyclase



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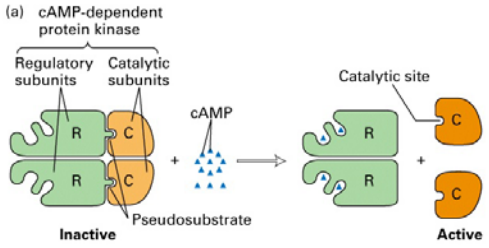
Activation of cAMP-dependent protein kinase (PKA)



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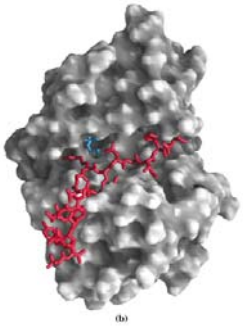
Activation of cAMP-dependent protein kinase (PKA)



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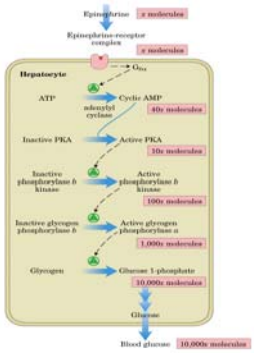
Epinephrine cascade: A example of signal amplification



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cAMP as a secondary messenger

TABLE 20-3 Metabolic Responses to Hormone-Induced Rise in cAMP in Various Tissues

Tissue	Hormone Inducing Rise in cAMP	Metabolic Response
Adipose	Epinephrine; ACTH; glucagon	Increase in hydrolysis of triglyceride; decrease in amino acid uptake
Liver	Epinephrine; norepinephrine; glucagon	Increase in conversion of glycogen to glucose; inhibition of synthesis of glycogen; increase in amino acid uptake; increase in gluconeogenesis (synthesis of glucose from amino acids)
Ovarian follicle	FSH; LH	Increase in synthesis of estrogen, progesterone
Adrenal cortex	ACTH	Increase in synthesis of aldosterone, cortisol
Cardiac muscle cells	Epinephrine	Increase in contraction rate
Thyroid	TSH	Secretion of thyroxine
Bone cells	Parathyroid hormone	Increase in resorption of calcium from bone
Skeletal muscle	Epinephrine	Conversion of glycogen to glucose
Intestine	Epinephrine	Fluid secretion
Kidney	Vasopressin	Resorption of water
Blood platelets	Prostaglandin I	Inhibition of aggregation and secretion

SOURCE: E. W. Sutherland, 1972, *Science* **177**:401.

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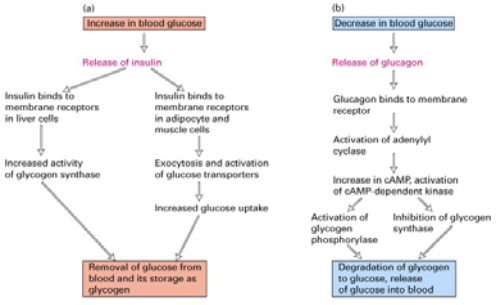

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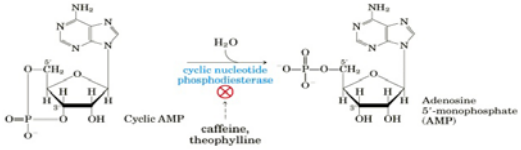
Inactivation of cAMP



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Medicine: β-adrenergic receptor

Cardiac muscle cells possess β1 receptor, whose activation increases heart rate. Practolol（心得灵）, an β1-selective antagonist, can slow heart contraction and is used to treat cardiac arrhythmia and angina.

Smooth muscle cells have β2 receptors, whose activation promotes relaxation. Terbutaline（特布他林）, an agonist selective for β2, is used in the treatment of asthma.

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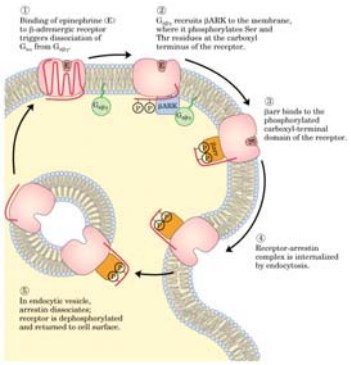
Desensitization of the β-adrenergic receptor



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Secondary messengers:

cAMP
cGMP
Lipids
Calcium

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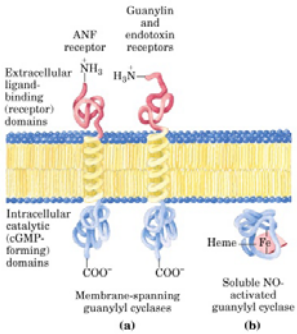
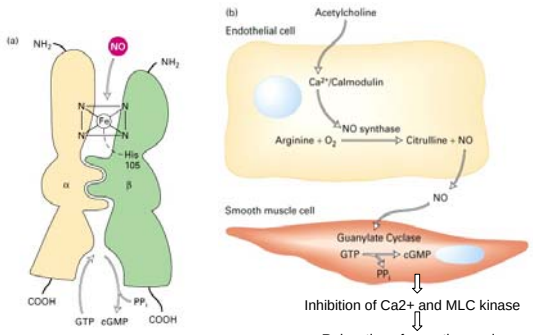
Guanylyl cyclases



Endotoxin, produced by E. coli and other gram-negative bacteria in intestine, stimulates cGMP production, increases Cl- secretion and consequently decreases reabsorption of water by the intestinal epithelium, producing diarrhea

35

NO (Nitric Oxide) signaling



Inhibition of Ca2+ and MLC kinase
⇩
Relaxation of smooth muscle
Vasodilation

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Secondary messengers:

cAMP
cGMP
Lipids
Calcium

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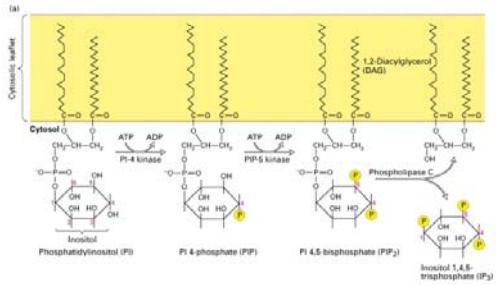
Lipids as messengers



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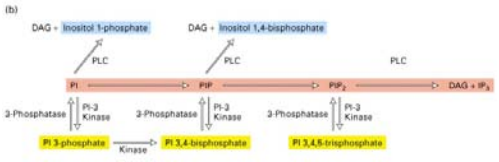
Lipid phosphorylation



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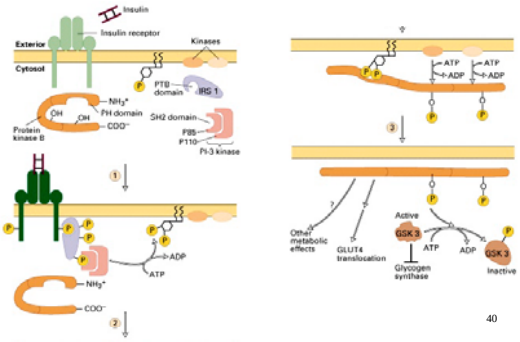
Insulin regulates glucose metabolism via PI3K



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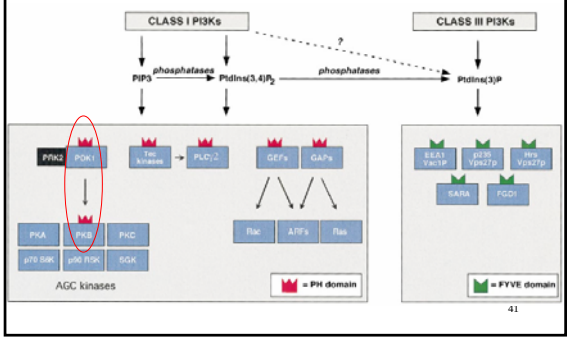
PH domain-containing proteins



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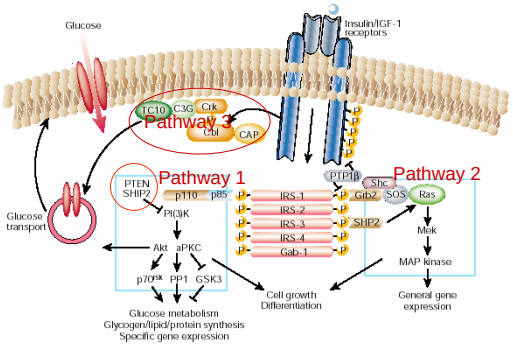
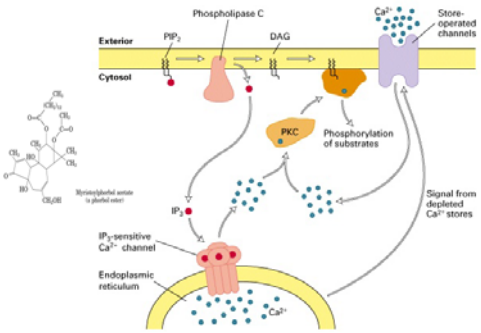
Glucose metabolism controlled by Insulin signaling



Lipids and calcium as secondary messengers



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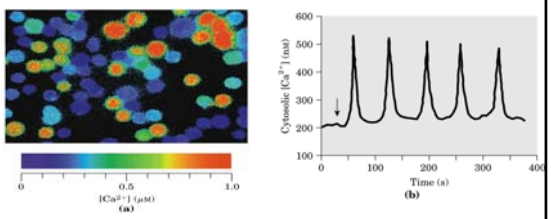
Triggering of oscillations in intracellular [Ca^{2+}] by extracelluar signals



Norepinephrine treatment of hepatocytes

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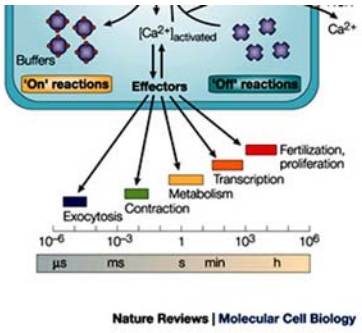
Calcium activated proteins



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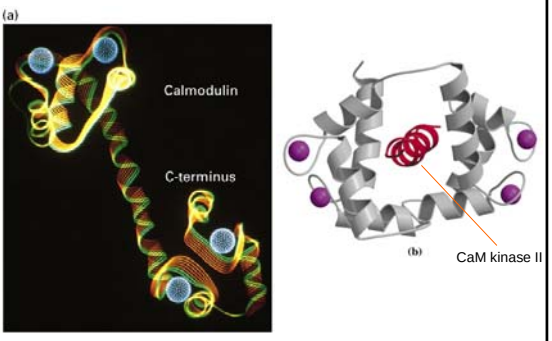
Calmodulin



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Calcium activated proteins

- Calmodulin: CaM kinase; phosphorylase b kinase (glycogen breakdown)
- Protein kinase C
- Calcineurin: protein phosphatase 1B
- Tropinin - muscle contraction
- Synaptotagmin (neurotransmitter release)
- Guanylyl cyclase

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Calcium activated proteins

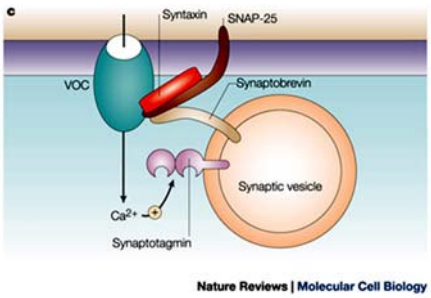

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TABLE 20-4 Cellular Responses to Hormone-Induced Rise in Inositol 1,4,5-Trisphosphate (IP_3) and Subsequent Rise in Cytosolic Ca^{2+} in Various Tissues

Tissue	Hormone Inducing a Rise in IP_3	Cellular Response
Pancreas (acinar cells)	Acetylcholine	Secretion of digestive enzymes, such as amylase and trypsinogen
Parotid (salivary gland)	Acetylcholine	Secretion of amylase
Pancreas (β cells of islets)	Acetylcholine	Secretion of insulin
Vascular or stomach smooth muscle	Acetylcholine	Contraction
Liver	Vasopressin	Conversion of glycogen to glucose
Blood platelets	Thrombin	Aggregation, shape change, secretion of hormones
Mast cells	Antigen	Histamine secretion
Fibroblasts	Peptide growth factors, such as bombesin and PDGF	DNA synthesis, cell division
Sea urchin eggs	Spermatozoa	Rise of fertilization membrane

source: M. J. Berridge, 1987, *Ann. Rev. Biochem.* **56**:159; M. J. Berridge and R. F. Irvine, 1984, *Nature* **312**:315.

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Protein modules

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GTP-binding proteins: A superfamily

- Trimeric G proteins: G_s, G_i, G_q, G_t
- Small GTP-binding proteins
  - ✓ Ras: cell growth
  - ✓ Rac, Rho, Cdc42: cell migration
  - ✓ Rab:membrane trafficking
  - ✓ ARF: membrane trafficking
  - ✓ Ran: Nuclear transport
- Other GTP-binding proteins: Dynamin, EF-Tu

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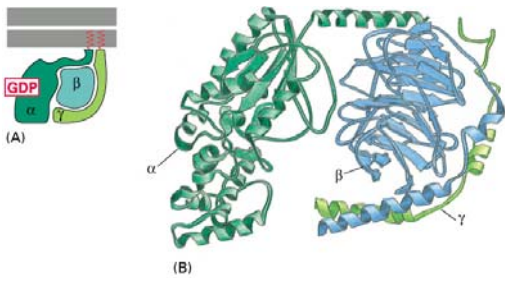
Trimeric G protein



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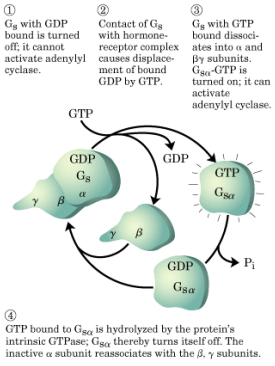
Self-inactivation of G_s



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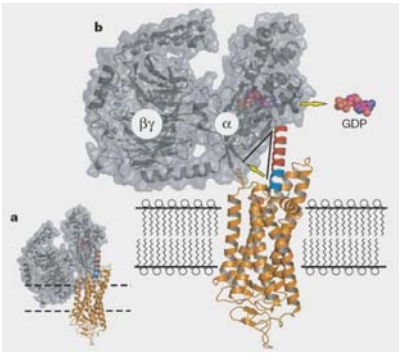
Structure of Opsin associated with the C-terminal Gsα



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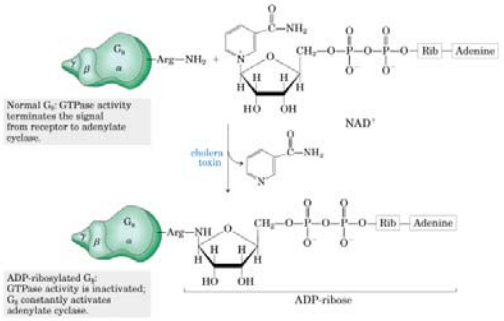
Scheerer et al, Nature, 2008

Toxins produced by bacteria that cause cholera (affecting Gsα) and whooping cough (pertussis) (affecting Giα)



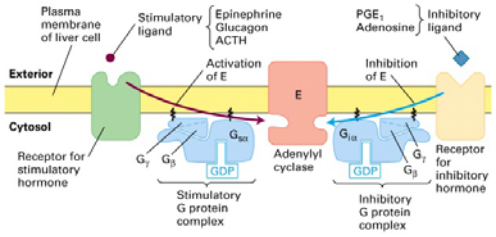


TABLE 20-5 Properties of Mammalian G Proteins Linked to GPCRs

G_α Subclass*	Effect	Associated Effector Protein	2nd Messenger
G_s	↑	adenylyl cyclase	cAMP
	↑	Ca^{2+} channel	Ca^{2+}
	↓	Na^+ channel	Change in membrane potential
G_i	↓	adenylyl cyclase	cAMP
	↑	K^+ channel	Change in membrane potential
	↓	Ca^{2+} channel	Ca^{2+}
G_q	↑	Phospholipase C	IP_3, DAG
G_o	↑	Phospholipase C	IP_3, DAG
	↓	Ca^{2+} channel	Ca^{2+}
G_t	↑	cGMP phosphodiesterase	cGMP
G_{olf}	↑	Phospholipase C	IP_3, DAG
	↓	Adenylyl cyclase	cAMP

*A given G_α may be associated with more than one effector protein. To date, only one major $G_{\beta\gamma}$ has been identified, but multiple $G_{\beta\gamma}$ and $G_{\alpha\beta}$ proteins have been described. In some cases (not indicated in this table) effector proteins are regulated by coincident binding to G_α and $G_{\beta\gamma}$.

key: ↑ = stimulation; ↓ = inhibition. IP_3 = inositol 1,4,5-trisphosphate; DAG 5 1,2-diacylglycerol.

source: See A. C. Dolphin, 1987, *Trends Neurosci.* **10**:53; L. Birnbaumer, 1992, *Cell* **71**:1069.

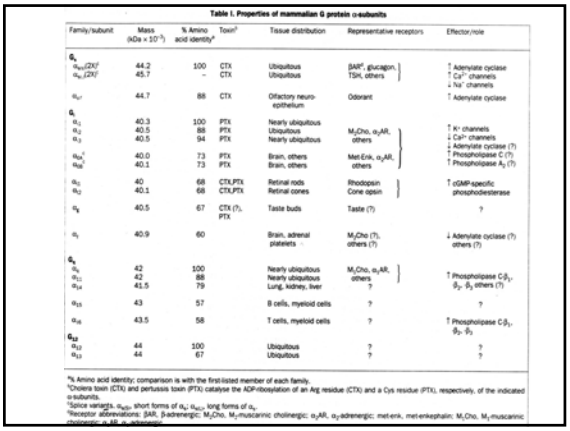
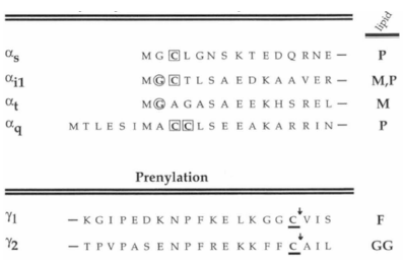


Table II. Properties of mammalian G protein β- and γ-subunits

Subunit	Mass (kDa × 10^{-3})	% Amino acid identity[a]	Tissue distribution	Effector/role
β				
β_1	37.3	100	Ubiquitous	Required for G_α-receptor interaction
β_2	37.3	90	Nearly ubiquitous	Inhibition of G_α activation
β_3	37.2	83	Nearly ubiquitous	
β_4	37.2	89	Nearly ubiquitous	Modulate activation of certain adenylate cyclases by $G_{\alpha s}$ or calmodulin
				Support of agonist-induced receptor phosphorylation and desensitization
γ				
γ_1	8.4	100	Retina, other (?)	↑ Phospholipase C
γ_2	7.9	38	Brain, adrenal, other (?)	↑ K^+ channels (?)
γ_3	8.5	36	Brain, testis	
γ_4	(?partial)	(34)	[Kidney, retina (?)]	↑ Phospholipase A_2 (?)
γ_5	7.3	25	Liver, other (?)	
γ_6	7.5	35	Brain, other (?)	

[a]% Amino acid identity: comparison is with the first-listed member of each family.

Lipid modification of trimeric G proteins



P: palmitoylation; M: myristoylation; F: farnesylation; GG: geranylgeranylation

Small GTP-binding proteins

- Ras: cell growth
- Rac, Rho, Cdc42: cell migration
- Rab: membrane trafficking
- ARF: membrane trafficking
- Ran: Nuclear transport

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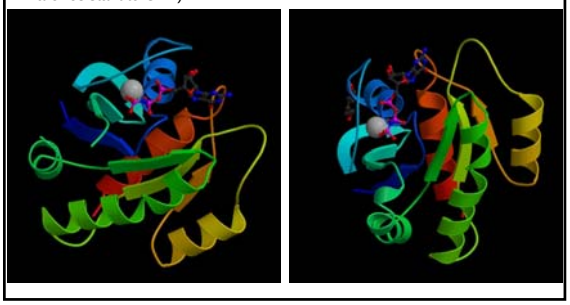
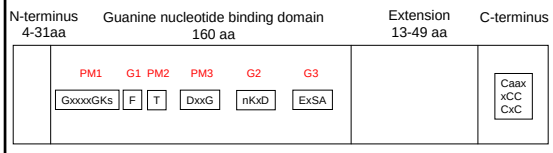
Ras

&; Oncogenes discovered from murine sarcoma viruses (Harvey virus - H-ras, Kirsten virus - K-ras) and N-ras;
&; Activated in 10-50% of human tumors (G12V, Q61L, both of which are resistant to GAP).



Conserved motifs in Ras-like proteins



PM: phosphate/Mg^{2+} binding regions; G: guanine base bidning regions;
Caax: a=aliphatic, x= any residue

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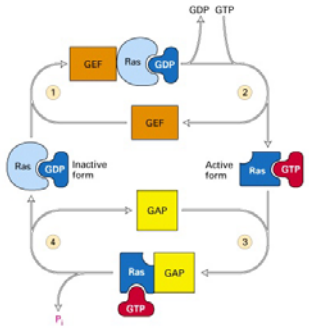
The GTP/GDP cycle of small GTP-binding proteins



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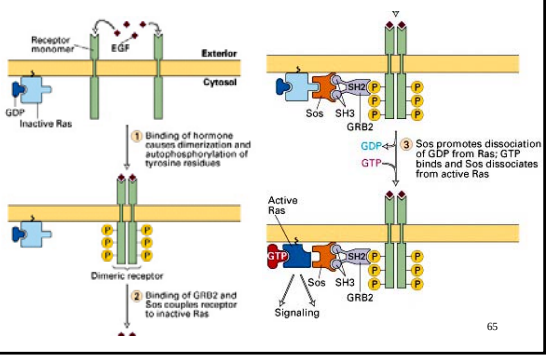
Ras mediates signaling from Receptor Tyrosine Kinase (RTK)



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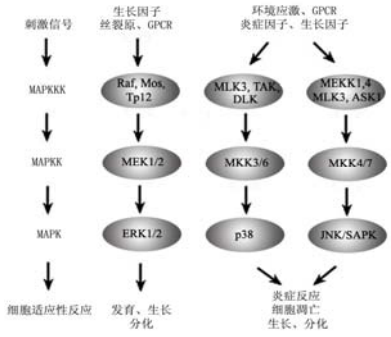
MAP kinase pathways



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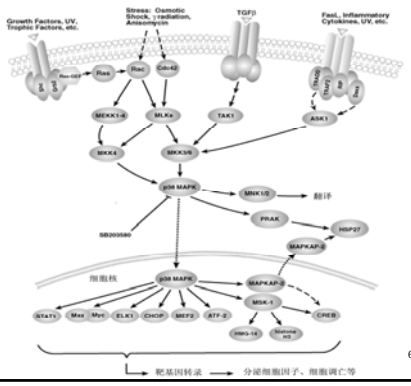
The MAP kinase pathways: p38 as an example



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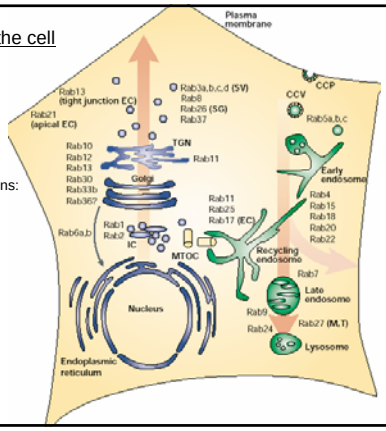
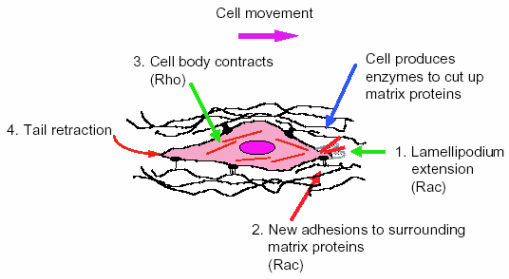
Rab proteins in the cell

Functions of Rab proteins:
Tethering and guiding vesicle fusion;
Vesicle formation.



Steps of cell migration



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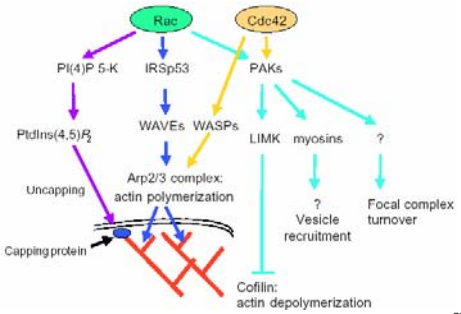
Ridley, 2001

Rac-induced lamellipodium extension



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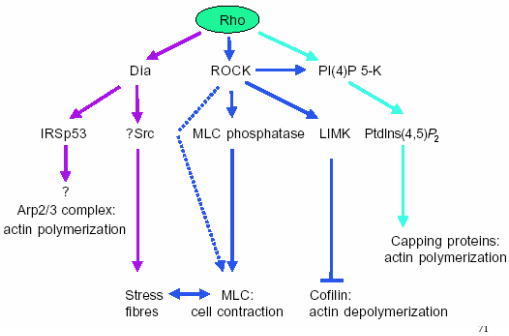
Ridley, 2001

Rho-induced actin reorganization



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Ridley, 2001

General introduction
- Common features of signal transduction
- Cell surface signal transducers, receptors

Ion channels

Secondary messengers
- cAMP
- cGMP
- Lipids
- Calcium

G proteins
- Trimeric G proteins
- Monomeric G proteins, Tyr kinase/MAP kinase

Protein modules

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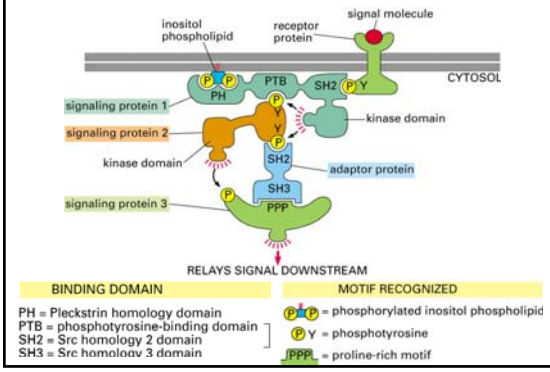
Protein Modules: protein building blocks

Interactions between intracellular signaling proteins are mediated by modular binding domains



Protein modules

PTB domains: ~ 100-150 aa, bind to in NPXY motifs: Shc, IRS-1

PDZ domains: ~80-90 aa, recognize short peptide motifs (4-5 residues) at the C-terminus of membrane proteins, usually containing a hydrophobic residue at the very end; protein-protein interaction: Dishevelled, FAP

SH2 (src homology): ~100 aa, binds to phosphotyrosine residues: Src, Grb2, Shc, STAT

SH3: binds to proline-rich sequences (PXXP): Src, Nck

WW domains: bind to Pro-rich sequences (XPPXY): Nedd4 (E3 ubiquitin ligase), Smurf, Dystrophin

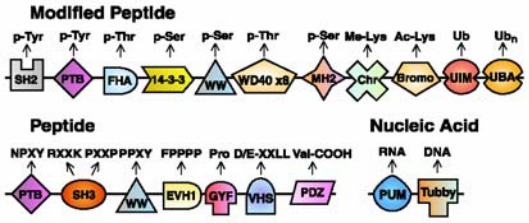
Death domains: Fas

LIM domains: recognize turn-based motifs

PH (Pleckstrin-homology) domains: associate with phosphinositides ($PI3,4P_2$; $PI4,5P_2$; $PI3,4,5P_3$), target proteins to the plasma membrane: Akt, SOS

FYVE domains: associate with phosphinositides (PI3P), target proteins to endosomes: EEA1, SARA

Building blocks - modular interaction domains

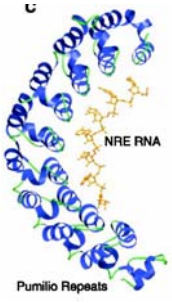


There are about 115 SH2 and 253 SH3 domains encoded by the human genome.

Pawson and Nash, 2003

Repeated motifs

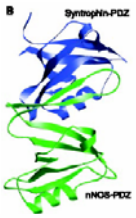
Some interaction domains are assembled from repeated motifs (up to 50 copies): HEAT, TPR, Arm, ankyrin, leucine-rich, Pumilio repeat.

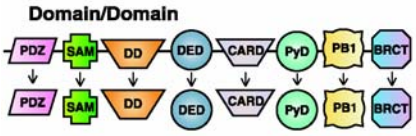


Pawson and Nash, 2003

Protein interaction domains

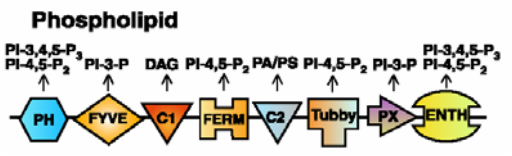
Domains bind to short peptide motifs, whereas a number of modular domains undergo homo- or heterotypic domain-domain interactions.





Pawson and Nash, 2003

Protein-lipid interaction domains



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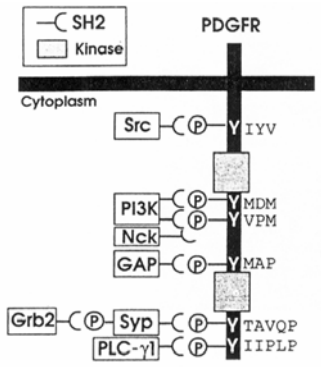
Pawson and Nash, 2003

Specificity of SH2 binding to phosphotyrosine



80
Pawson, 1995

谢谢!

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