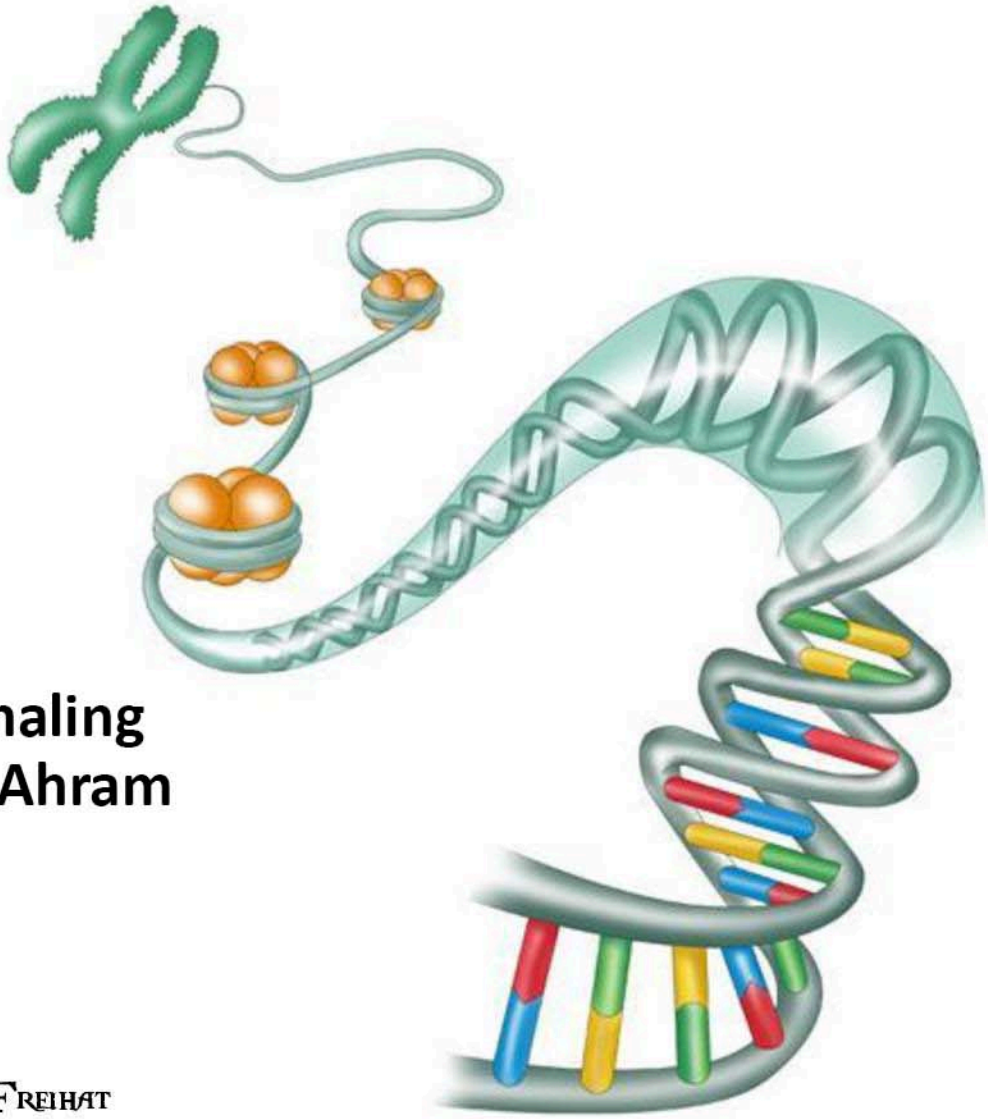
University of Jordan
Faculty of Medicine
Batch 2013-2019

Medical Committee
The University of Jordan

Genetics & Molecular Biology

● Slides ○ Sheet ○ Handout ○ other.....



Lecture # 9
Title: Cell Signaling
Dr. Mamoun Ahram
Done By:
Date:
Price:

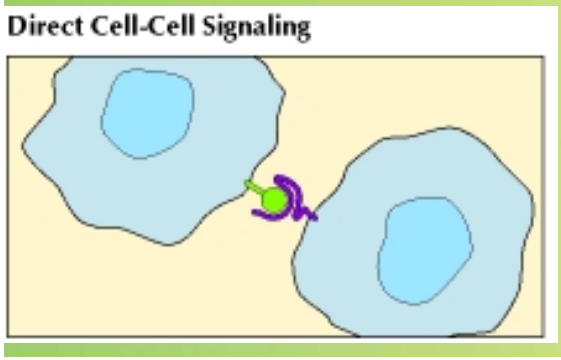
Designed by Nadeen Al-Freihat

Lecture 9:
Cell signaling

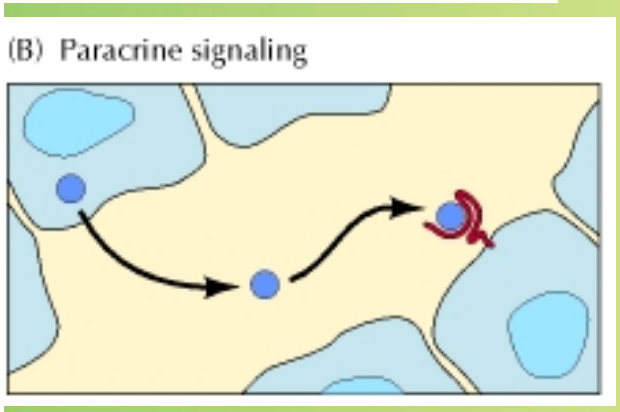
Dr. Mamoun Ahram
Faculty of Medicine
Second year, Second semester, 2014-2014

Principles of Genetics and Molecular Biology

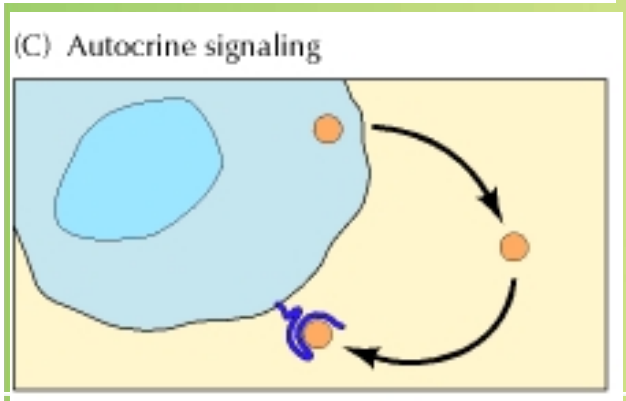
Modes of cell siganling



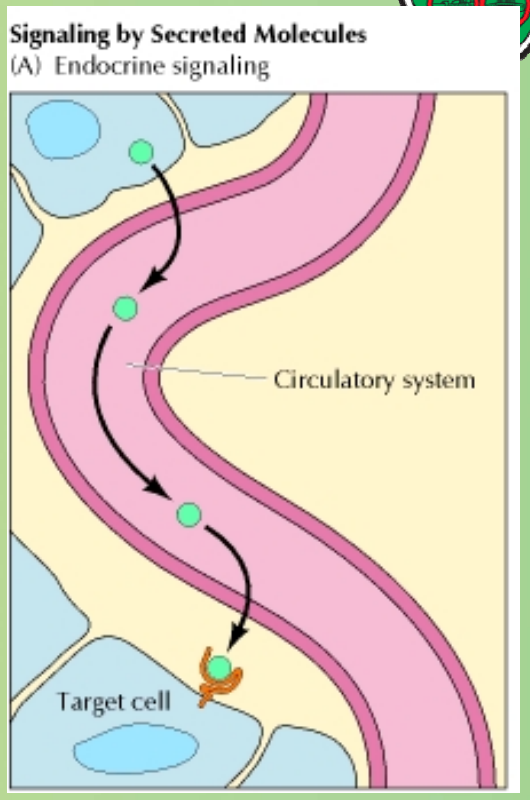
Direct interaction of a cell with its neighbor



A molecule released by one cell acts on neighboring target cells.



Cells respond to signaling molecules that they themselves produce

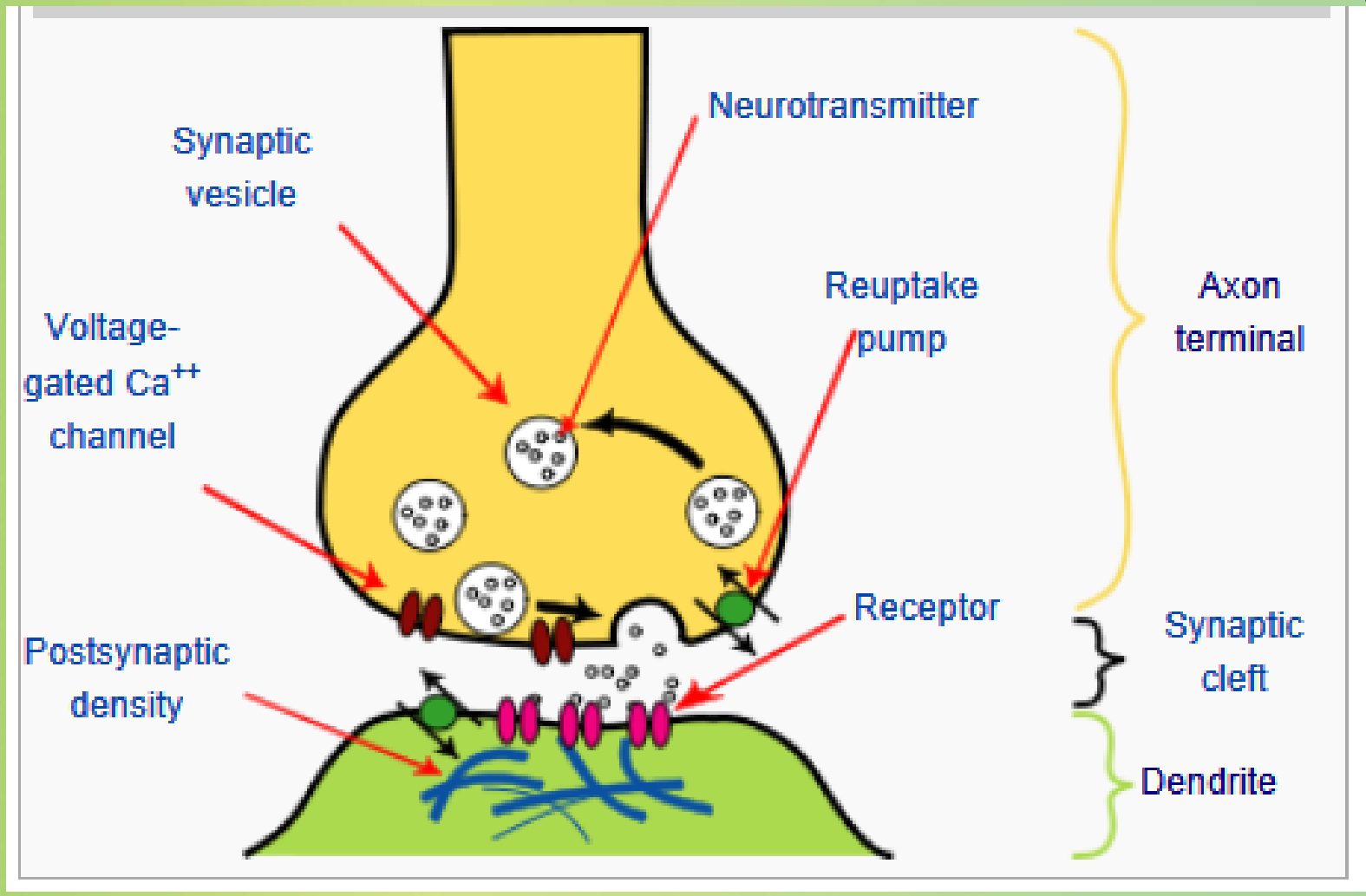


Signaling molecules are secreted by endocrine cells and carried through the circulation to act on target cells at distant body sites.

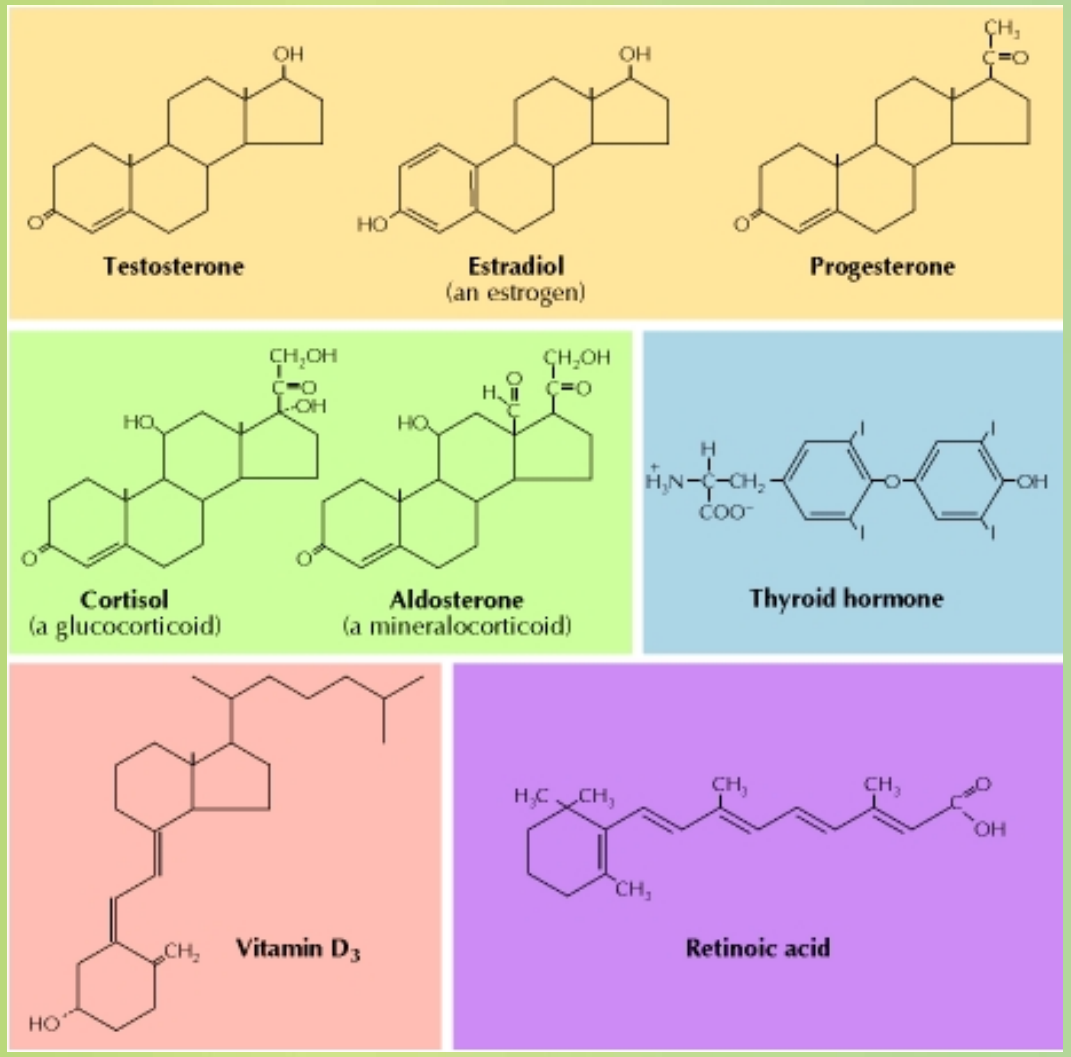
Classification of signaling molecules

- Peptides: growth factors (EGF), peptide hormones (insulin, glucagon), or neuropeptides (oxytocin, enkephalins)
- Small molecule neurotransmitters: derived from amino acids like Epinephrine and thyroid hormone (tyrosine), serotonin (tryptophan).
- Steroids: derived from cholesterol like estradiol, cortisol, calciferol (Vitamin D), and testosterone.
- Eicosinoids: derivatives of arachidonic acid including prostaglandins, leukotrienes, and thromboxanes B.
- Gasses: Nitric oxide (NO) and carbon monoxide (CO)

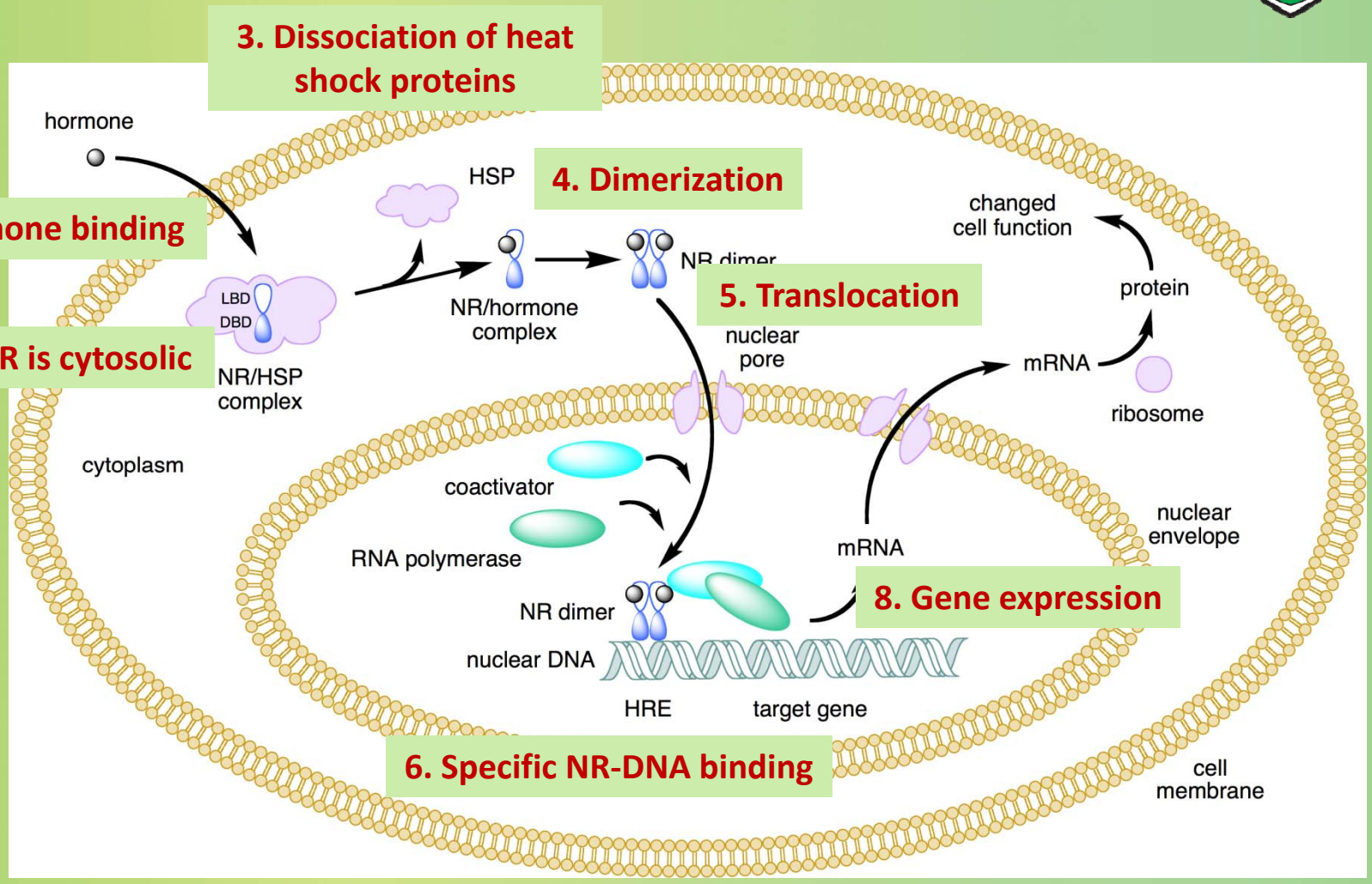
Mechanisms of action of neurotransmitter



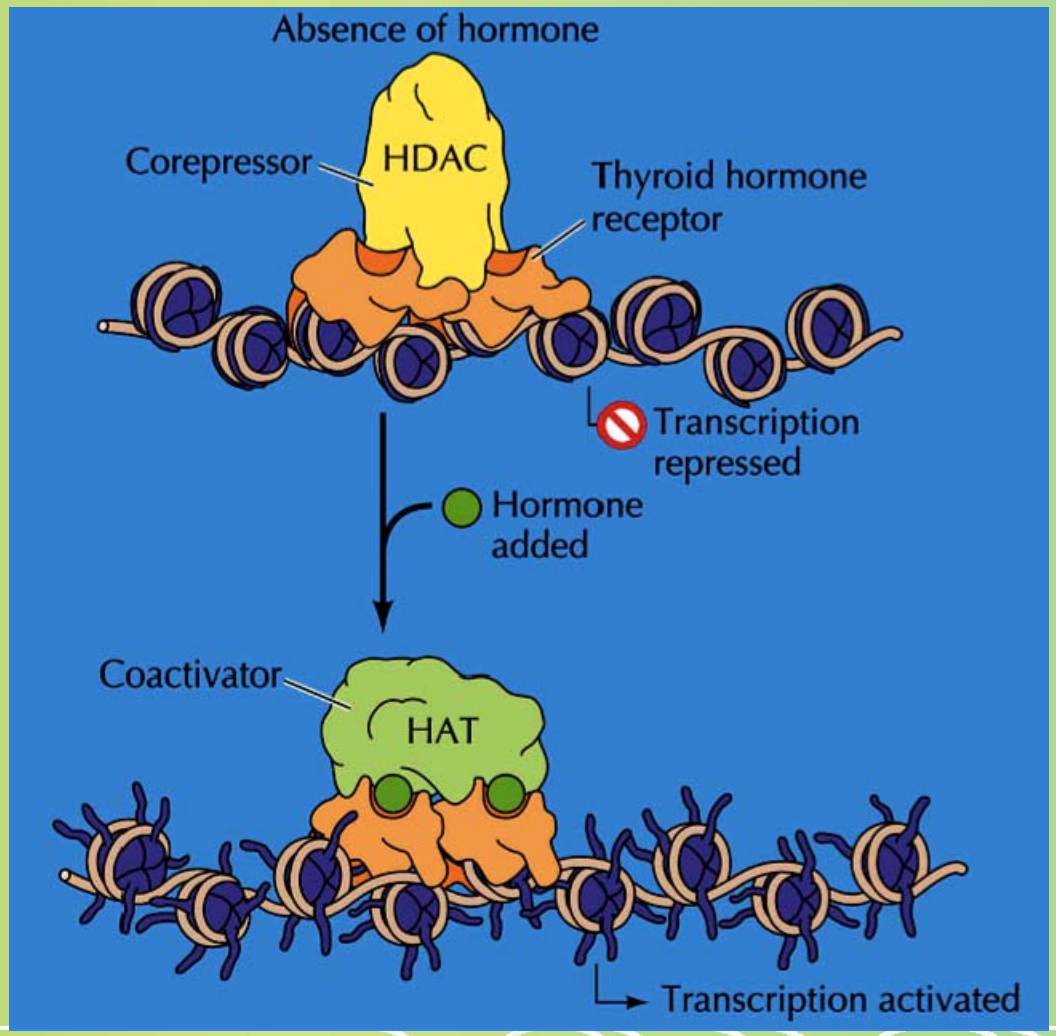
Lipophilic hormones



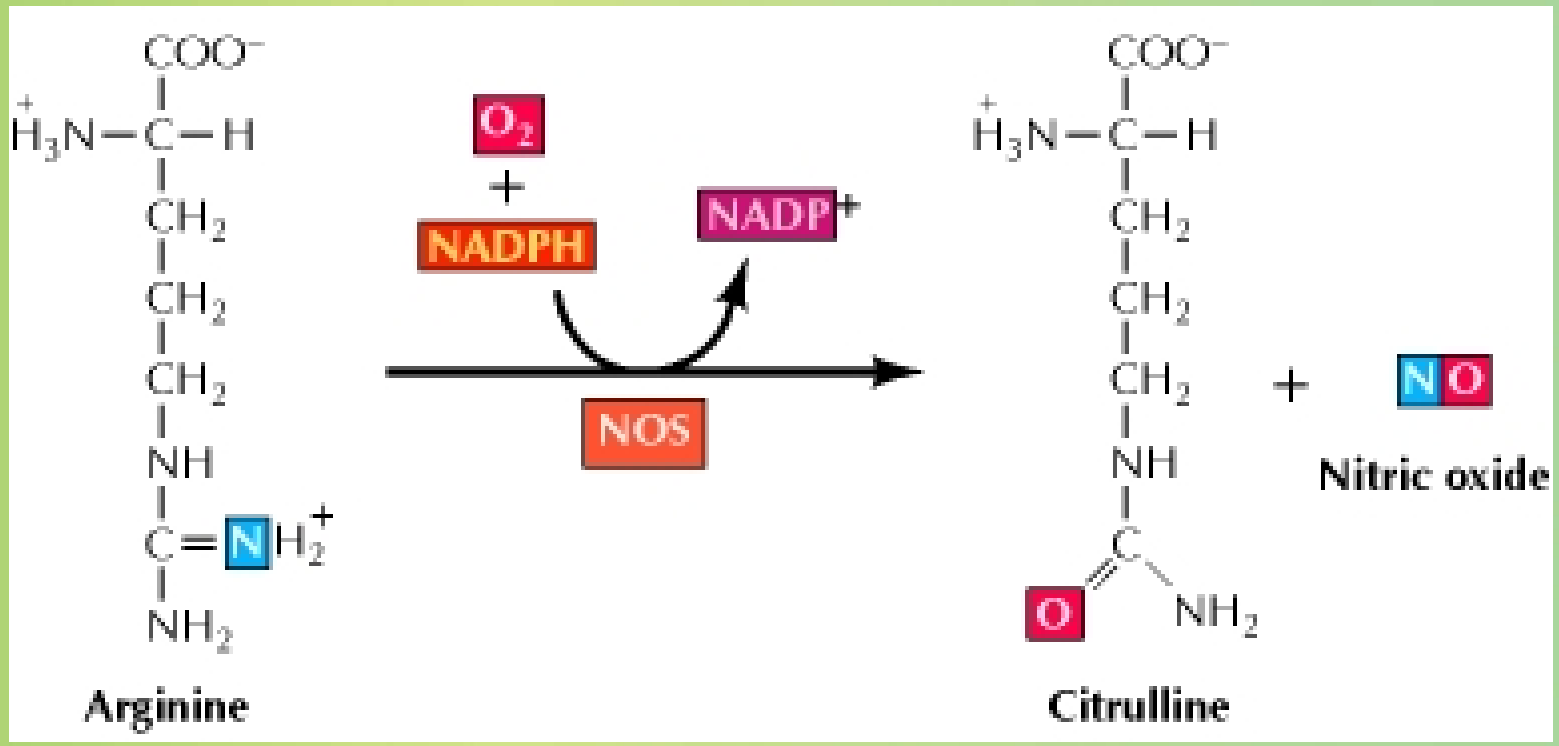
Mechanism of action of steroid receptors



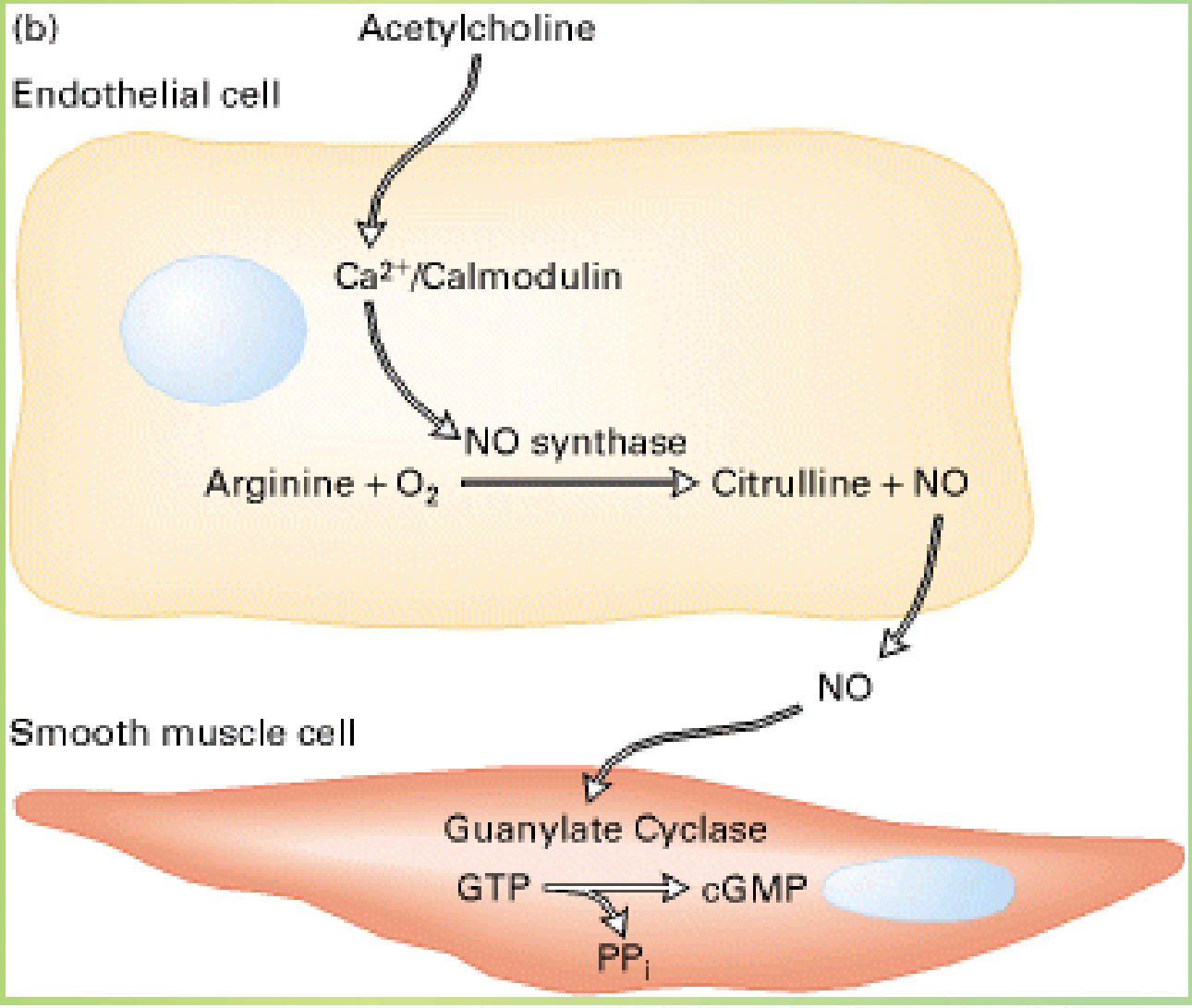
Gene regulation by the thyroid hormone receptor



Synthesis of nitric oxide (NO)



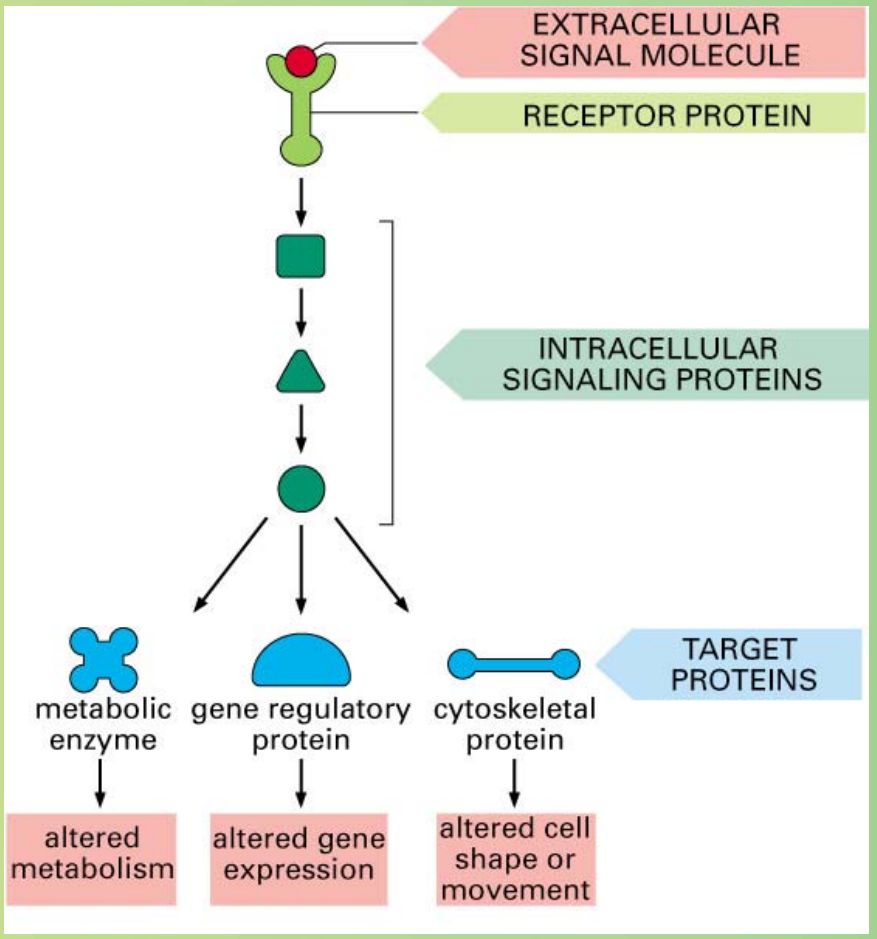
Mechanisms of action



Cell surface receptors

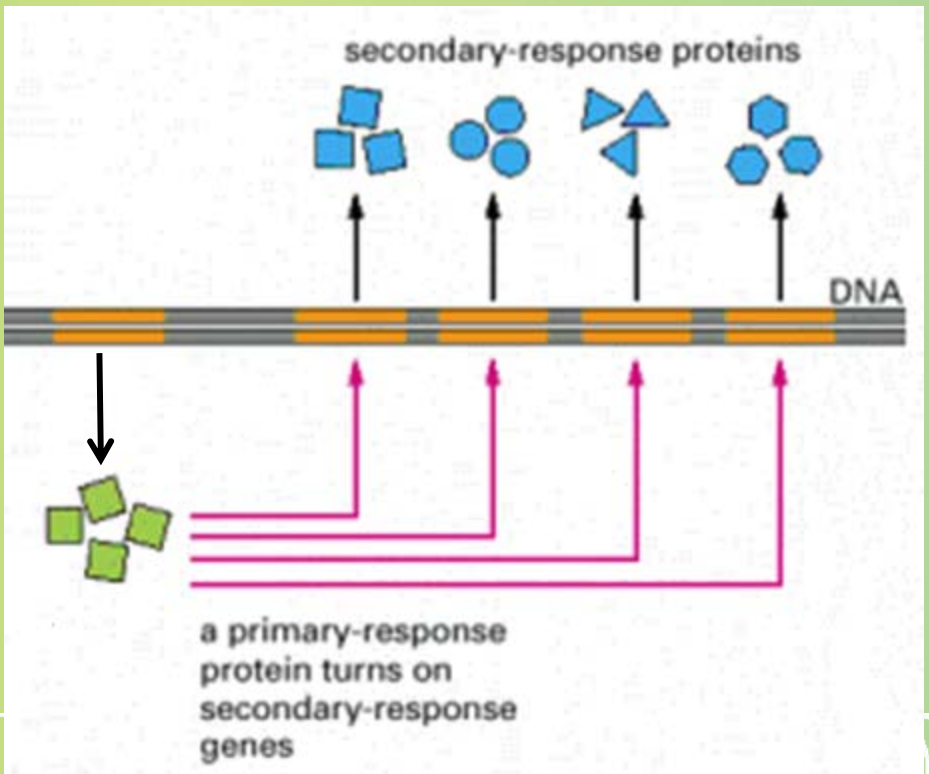
Players of signaling by cell surface receptors

- Ligand (hormone, growth factor)
- Receptor (GPCR, RTK)
- Transducers (G protein, Ras)
- Effector molecules (adenylate cyclase, MAPK, Ca^{2+})
- Second messengers
- Final target molecules (e.g., DNA, channel) → Response



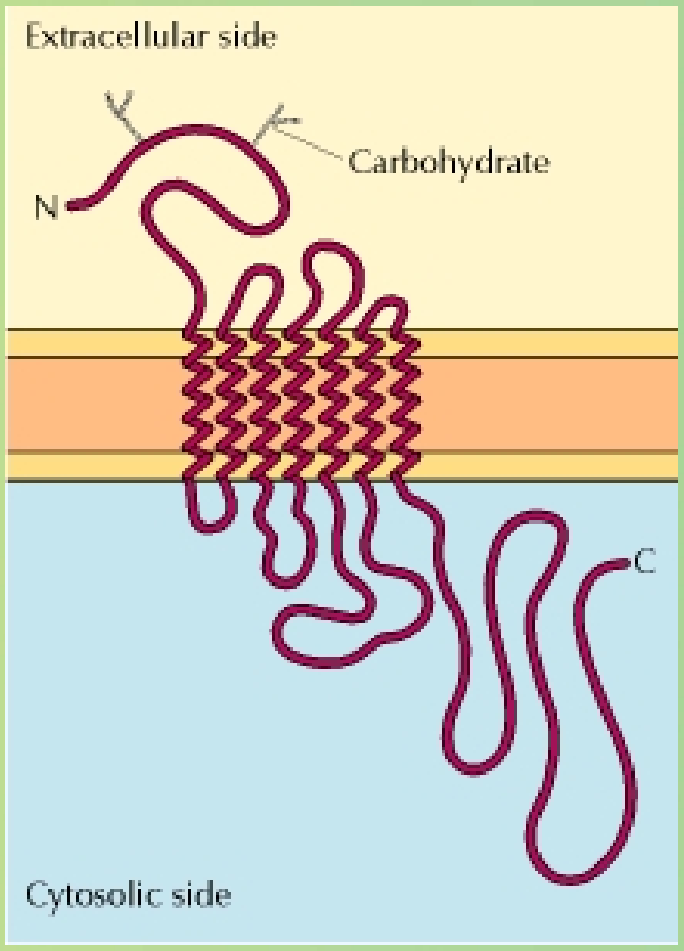
Types of response

- **Primary response direct activation of a small number of specific genes (30 minutes).**
- **Secondary response the protein products of the primary response activate other genes.**



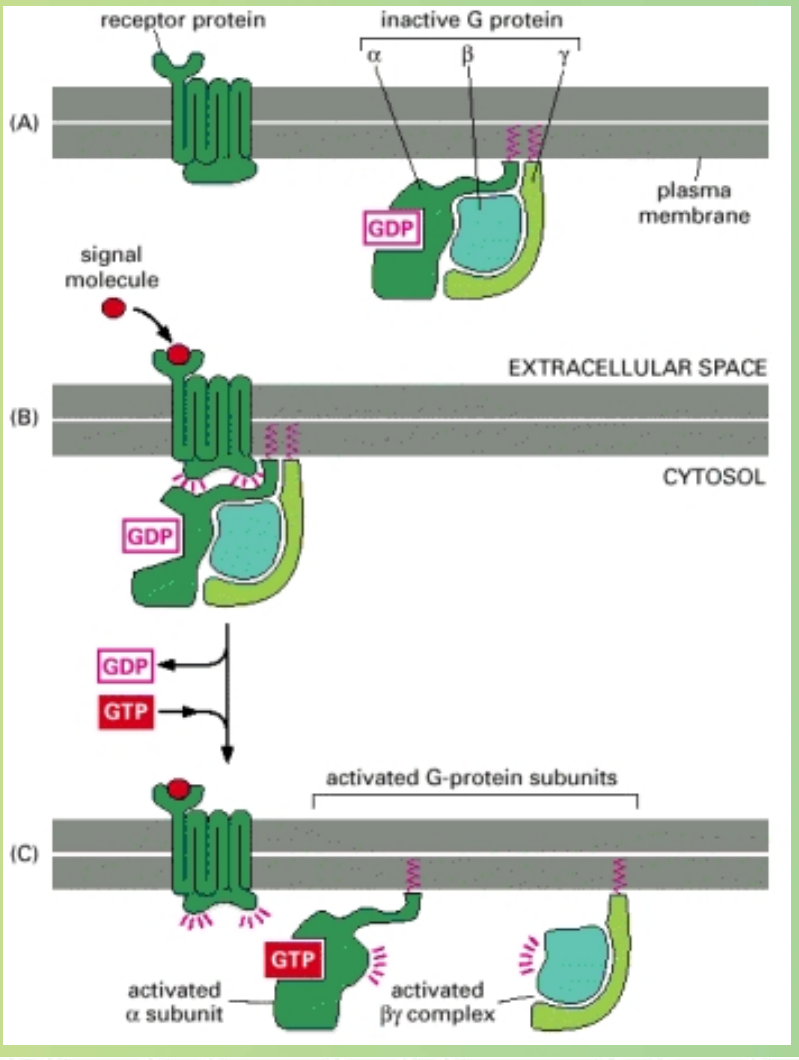
G protein-coupled receptors

- A family of receptors composed of seven membrane-spanning α helices.
- The binding of ligands to the extracellular domain of these receptors induces a conformational change that allows the cytosolic domain of the receptor to bind to a G protein.



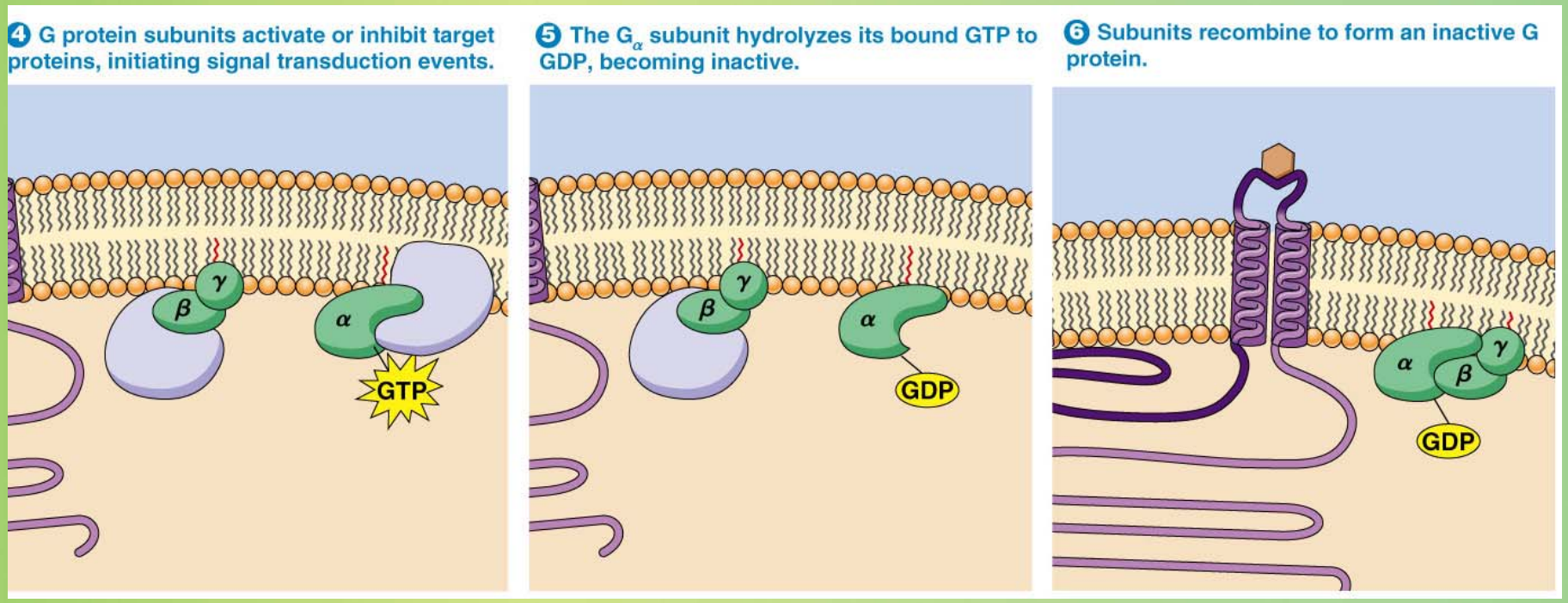
Heterotrimeric G proteins

- G proteins are composed of three protein subunits—α, β, and γ.
- In the unstimulated state, the α subunit has GDP bound and the G protein is inactive.
- When stimulated, the α subunit releases its bound GDP, allowing GTP to bind in its place.
- This exchange causes the trimer to dissociate into active components: α subunit and a βγ complex.



G protein inactivation

- The activity of the α subunit is terminated by hydrolysis of the bound GTP by an intrinsic GTPase activity, and the inactive α subunit (now with GDP bound) then reassociates with the βγ complex.

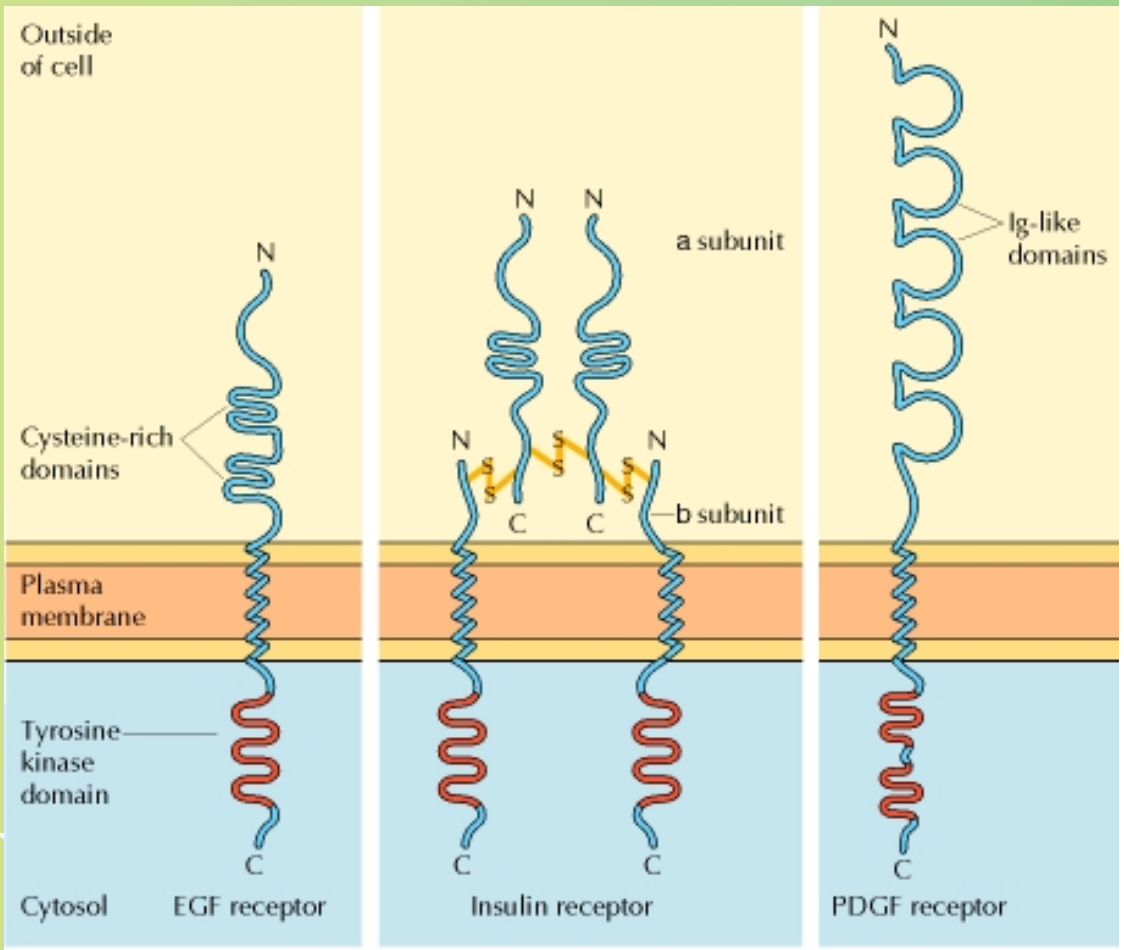


Types of G proteins

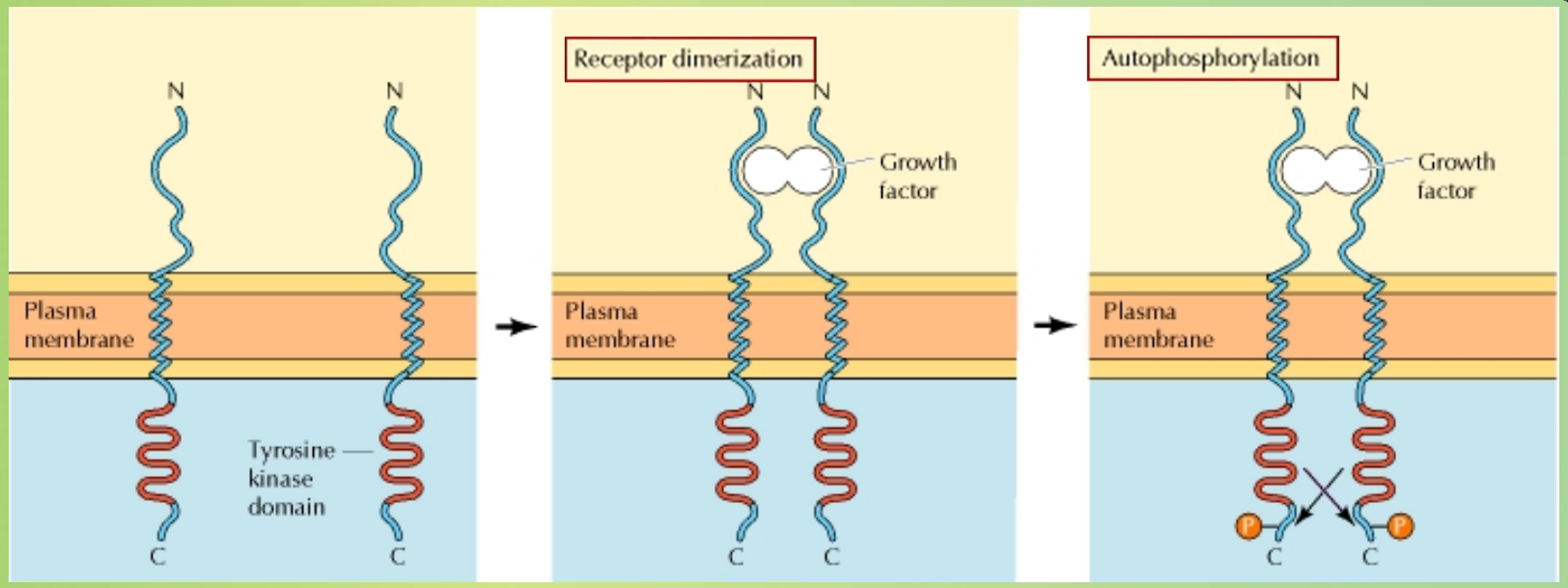
G_a class	Initiating signal	Downstream signal
G_{as}	β-Adrenergic receptor	Stimulates adenylate cyclase
G_{ai}	Acetylcholine, α-adrenergic	Inhibits adenylate cyclase
G_{aq}	Acetylcholine, α-adrenergic	Increases IP_3 and intracellular calcium
G_{at}	Photons	Stimulates cGMP phosphodiesterase
G_{a13}	Thrombin, other agonists	Stimulates Na^+ and H^+ exchange

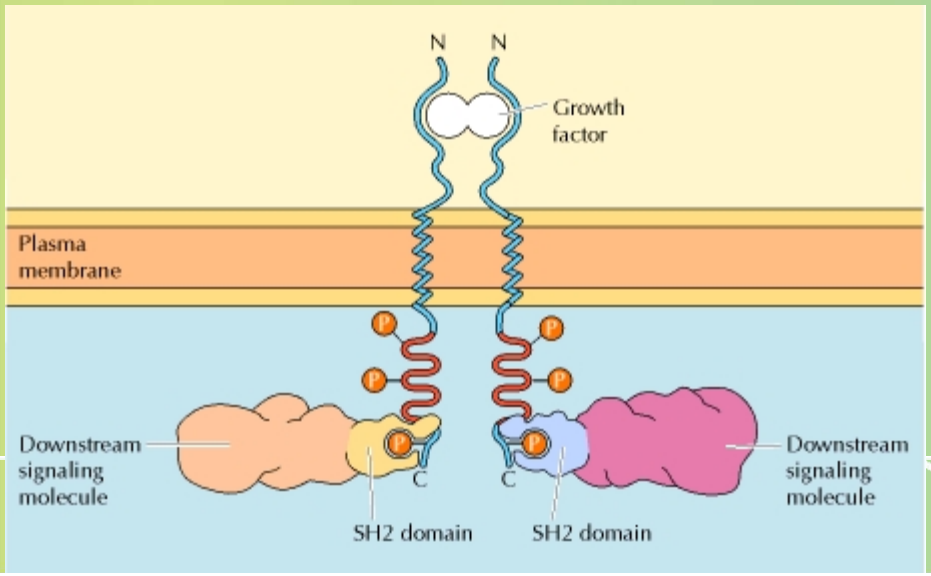
Receptor protein tyrosine kinase (RTK)

- Some receptors are directly linked to intracellular enzymes.
- RTKs have the enzymatic activity as part of the protein itself.
- Binding of ligands extraellularly activates the cytosolic kinase domains, resulting in phosphorylation of both the receptors themselves and intracellular target proteins.



Mechanism of activation of RTKs



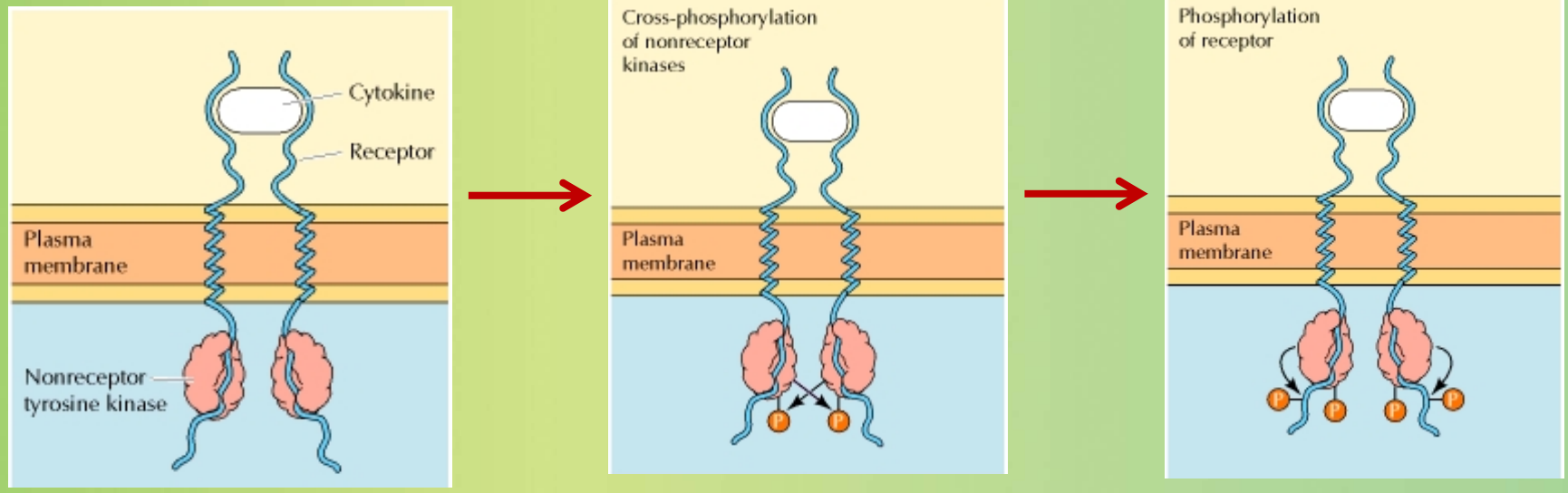


How does autophosphorylation activate signaling?

- **Autophosphorylation activates signaling by:**
  - **First, phosphorylation of tyrosines within the kinase domain increases the kinase activity**
  - **Second, phosphorylation of tyrosines outside the kinase domain creates high-affinity binding sites for the binding of other signaling proteins**

Nonreceptor protein tyrosine kinases

Cytokine receptor superfamily



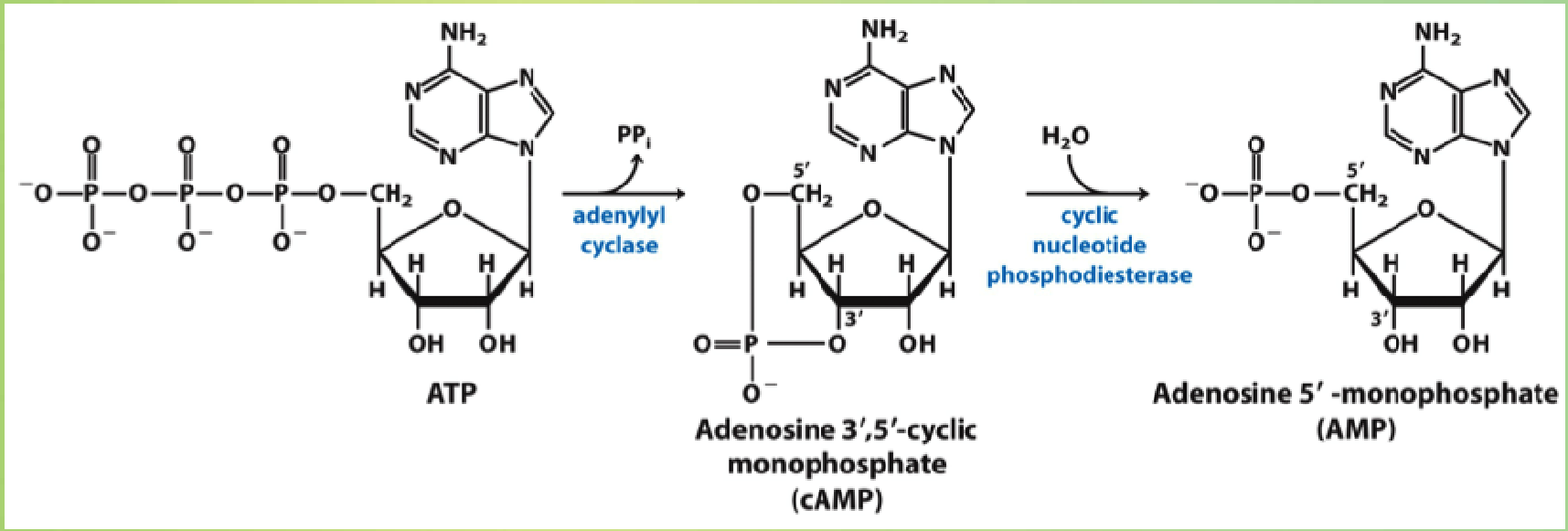
Examples: JAK and Src

Other examples

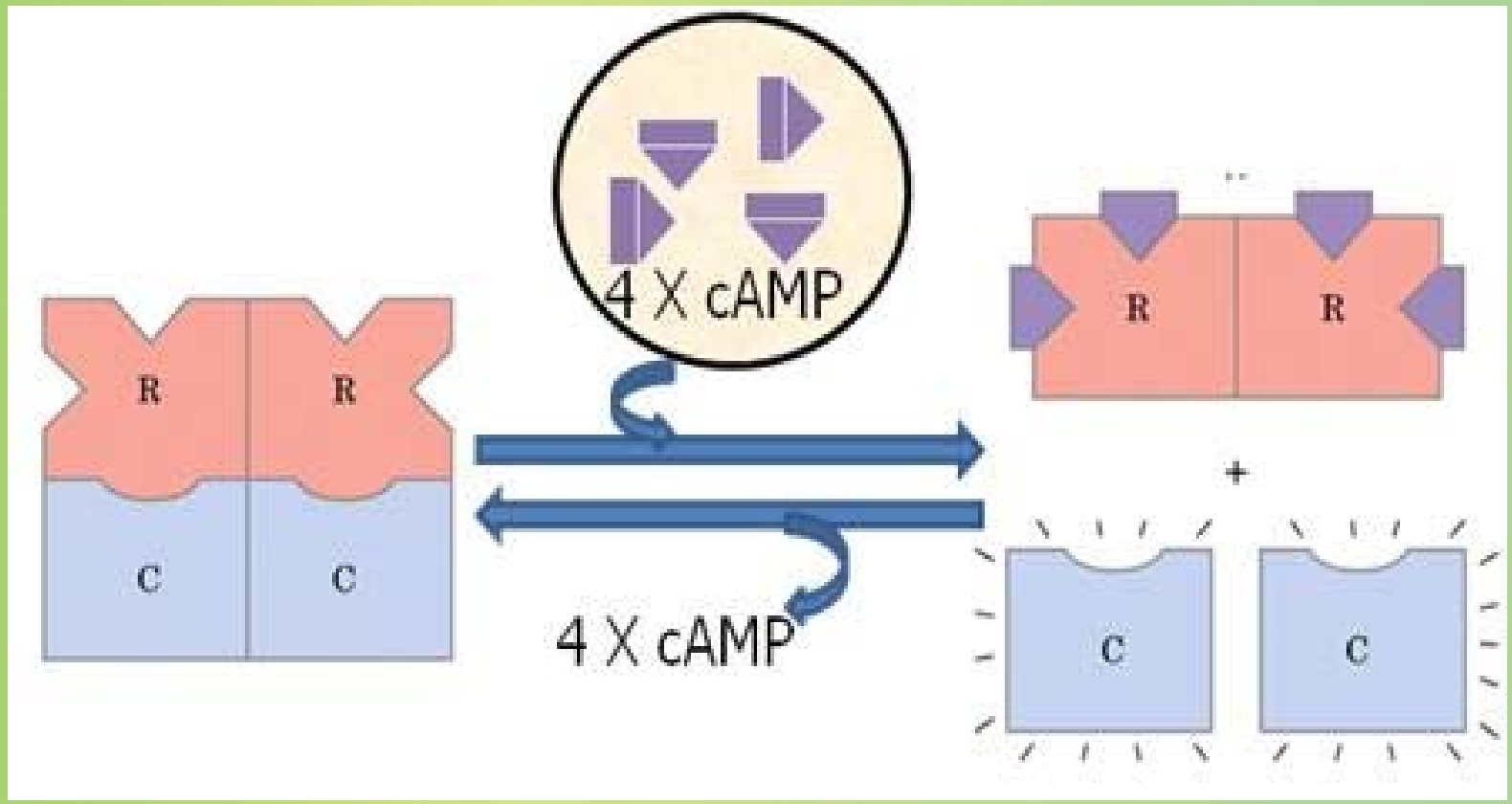
- Protein-tyrosine phosphatases: activation and inhibition roles
- Protein-serine/threonine kinase: transforming growth factor β (TGF-β)
- Receptor guanylyl cyclases
- Protease-associated receptor: tumor necrosis factor (TNF)

Second messengers

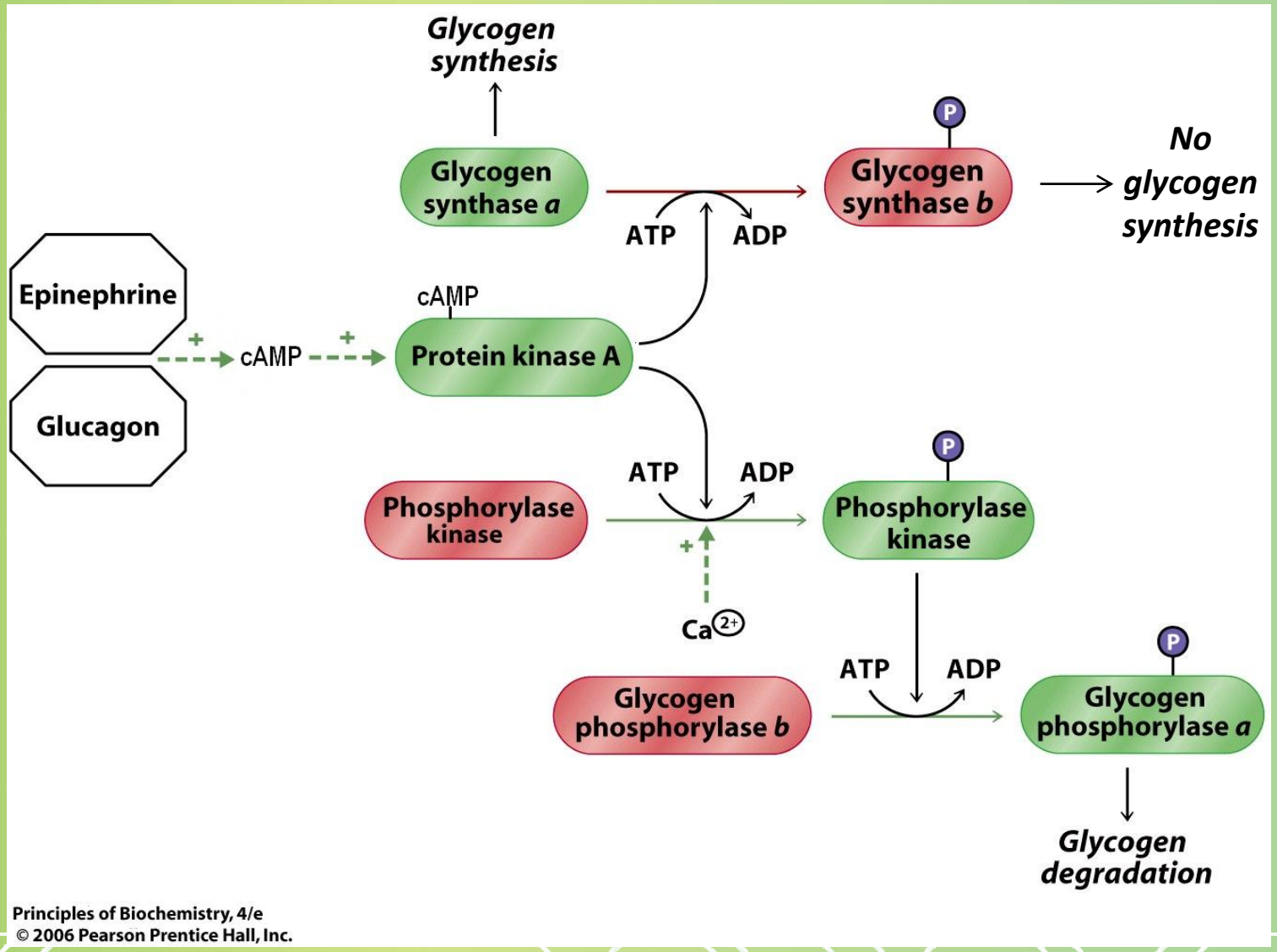
Synthesis and degradation of cAMP



Regulation of protein kinase A by cAMP

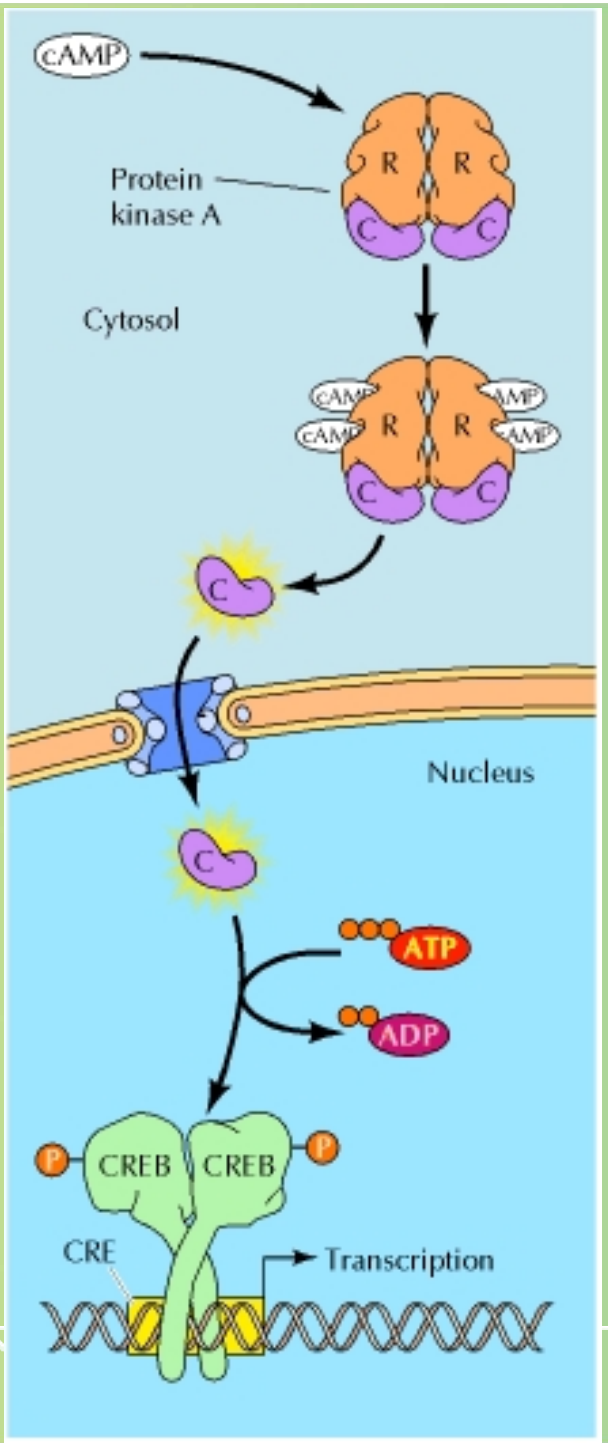


Regulation of glycogen metabolism



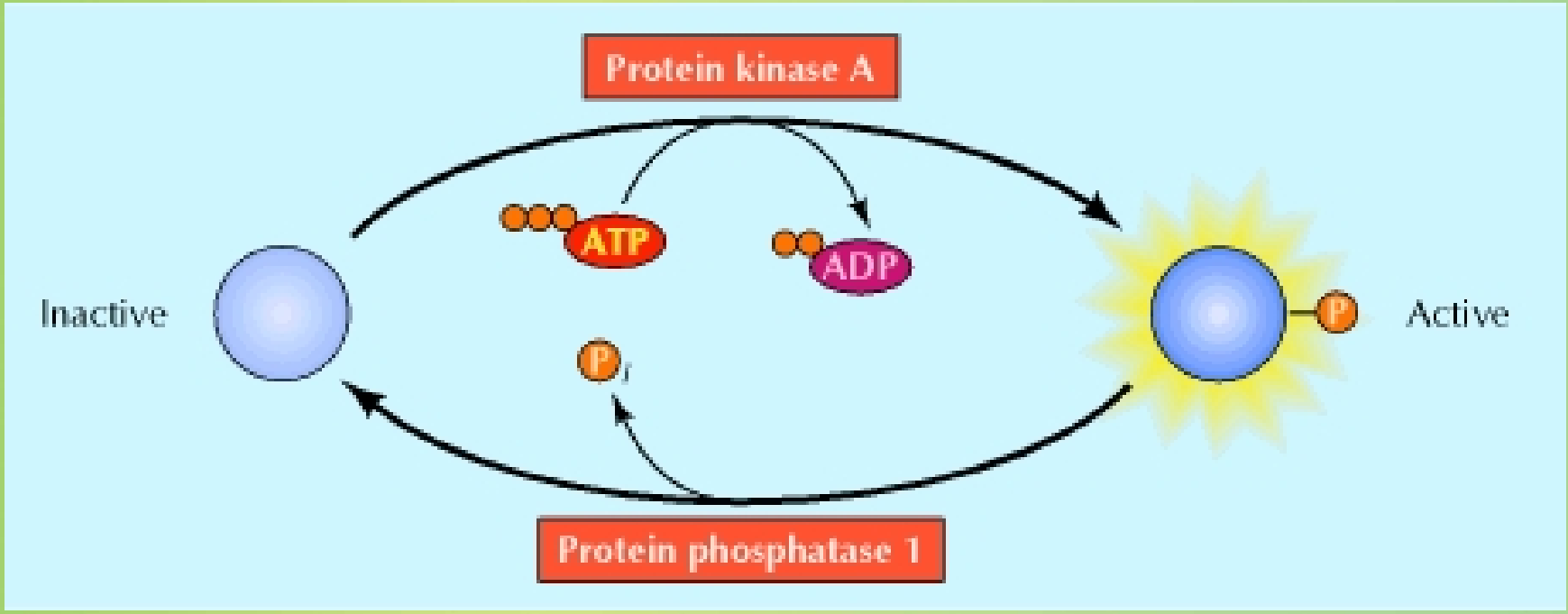
Cyclic AMP-inducible gene expression

- The free catalytic subunit of protein kinase A translocates into the nucleus and phosphorylates the transcription factor CREB (CRE-binding protein), leading to expression of cAMP-inducible genes.

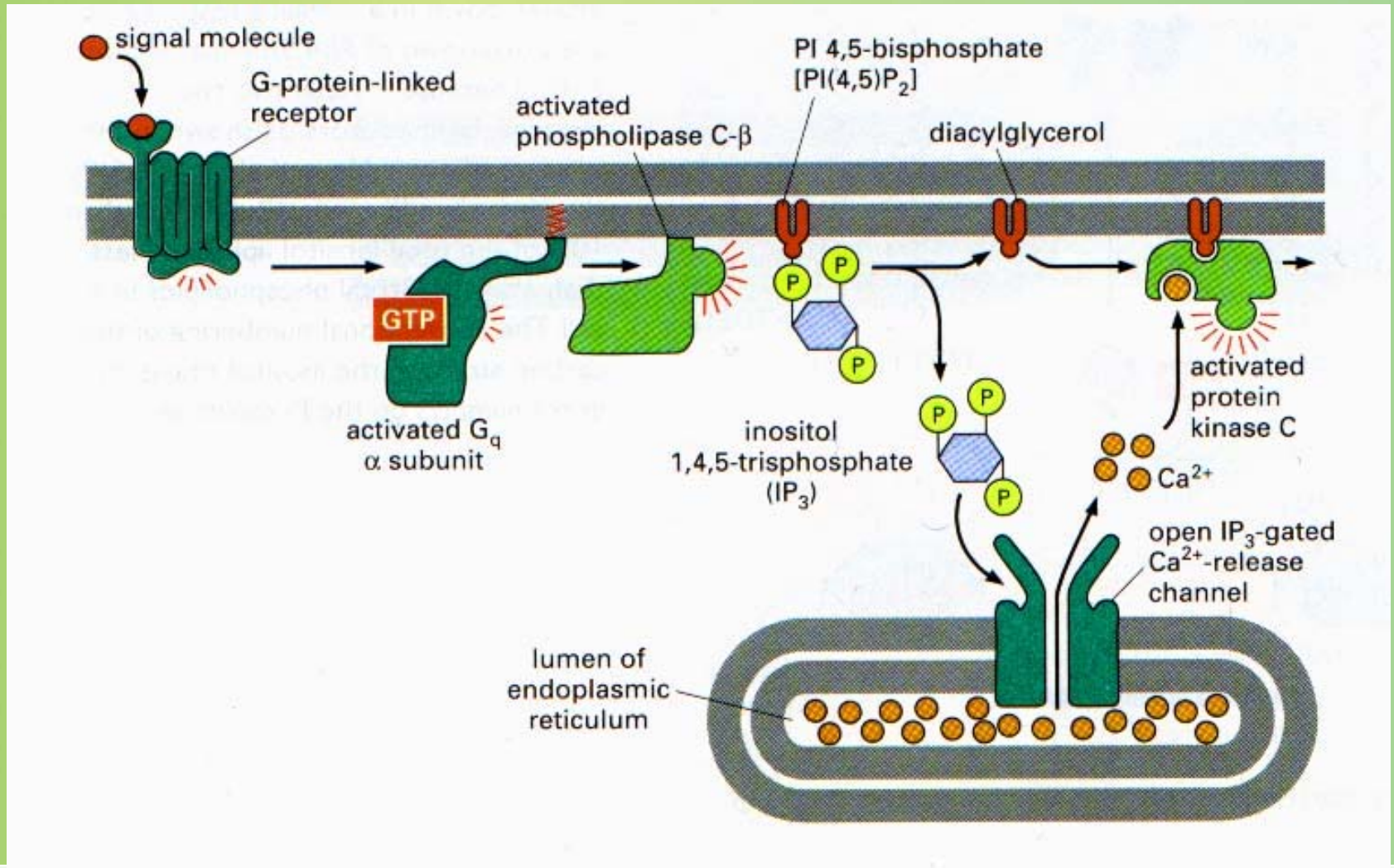


Regulation by dephosphorylation

- The phosphorylation of target proteins by protein kinase A is reversed by the action of protein phosphatase 1.

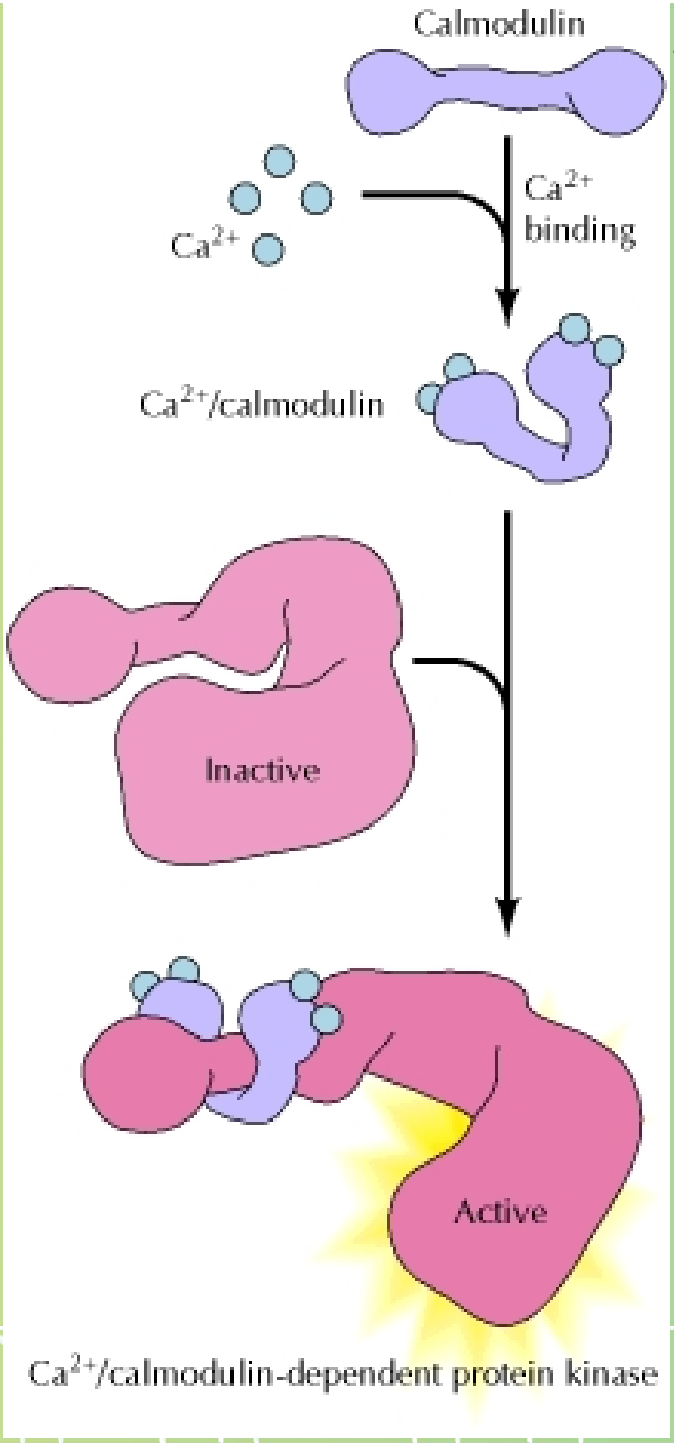


Phospholipids and Ca^{2+}



Ca^{2+}/calmodulin

- Ca^{2+} binds to calmodulin, which regulates many proteins such as:
- Ca^{2+}/calmodulin-dependent protein kinases signals actin-myosin contraction.
- CaM kinases regulates the synthesis and release of neurotransmitters.
  - CREB (at same site as PKA).

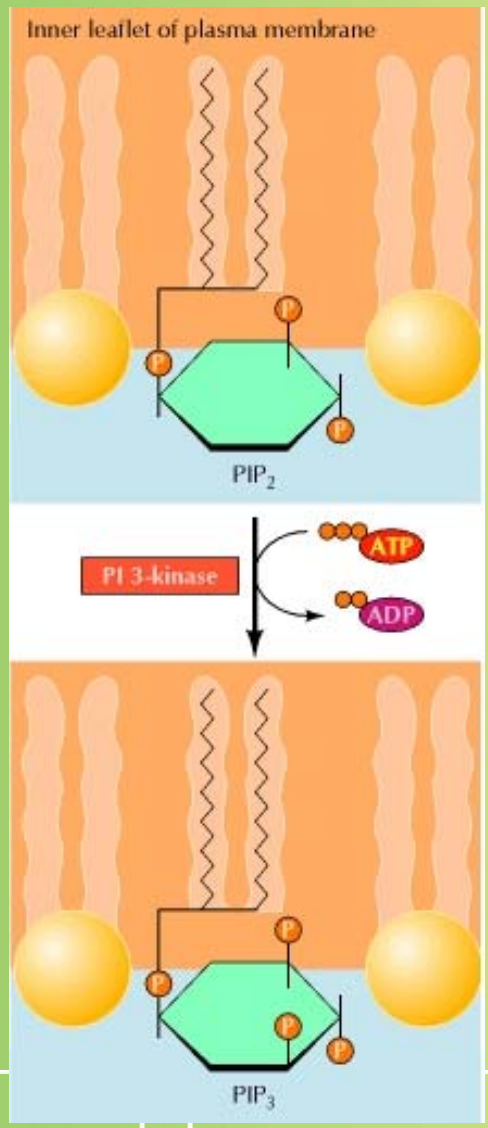


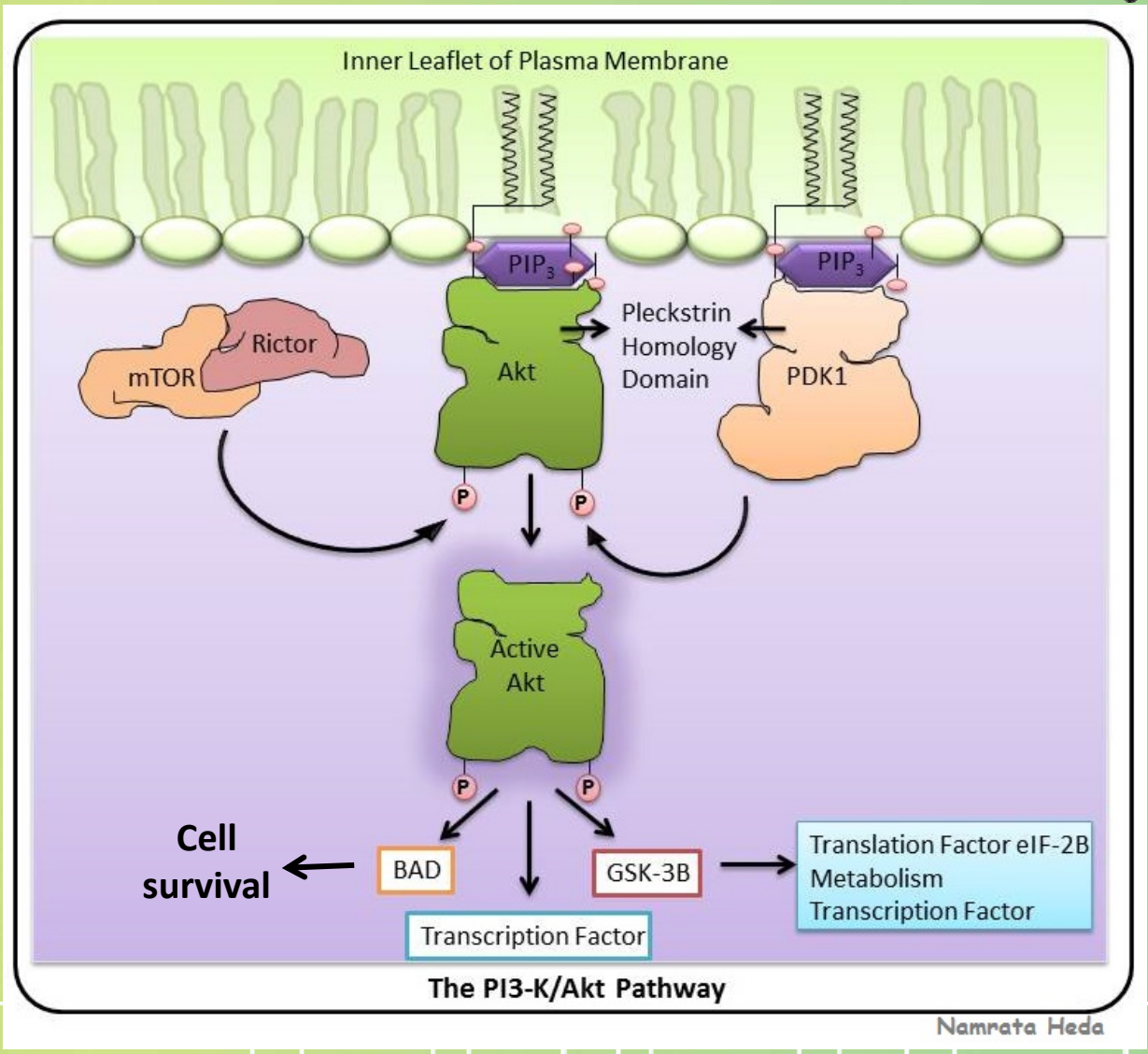
Why are second messengers good?

- Second messengers are often free to diffuse to other compartments of the cell
- The signal may be amplified significantly in the generation of second messengers
- The use of common second messengers in multiple signaling pathways often results in cross-talk between different signaling pathways

Signaling pathways

PI-3 kinase and AKT pathway





mTOR pathway and autophagy

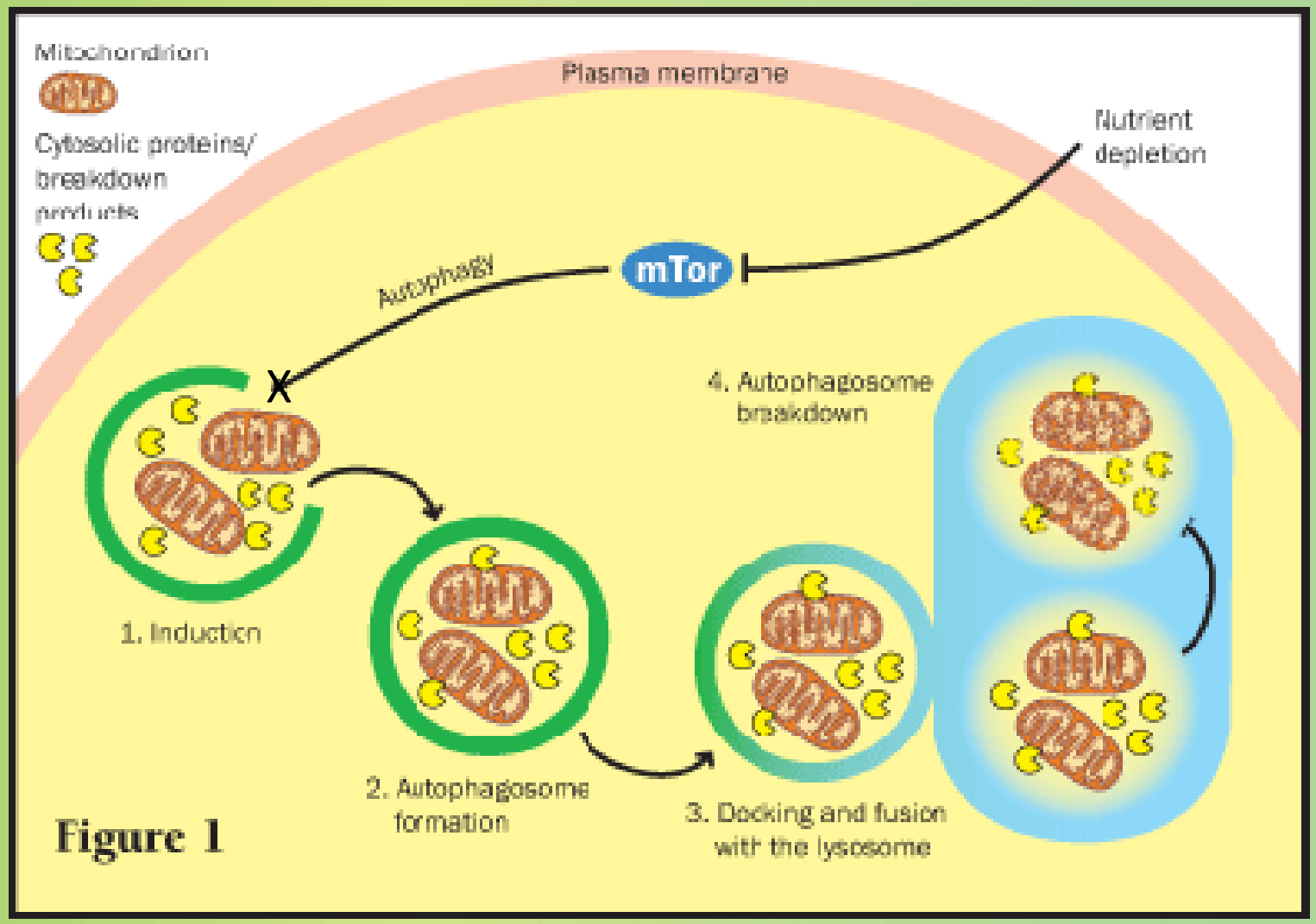
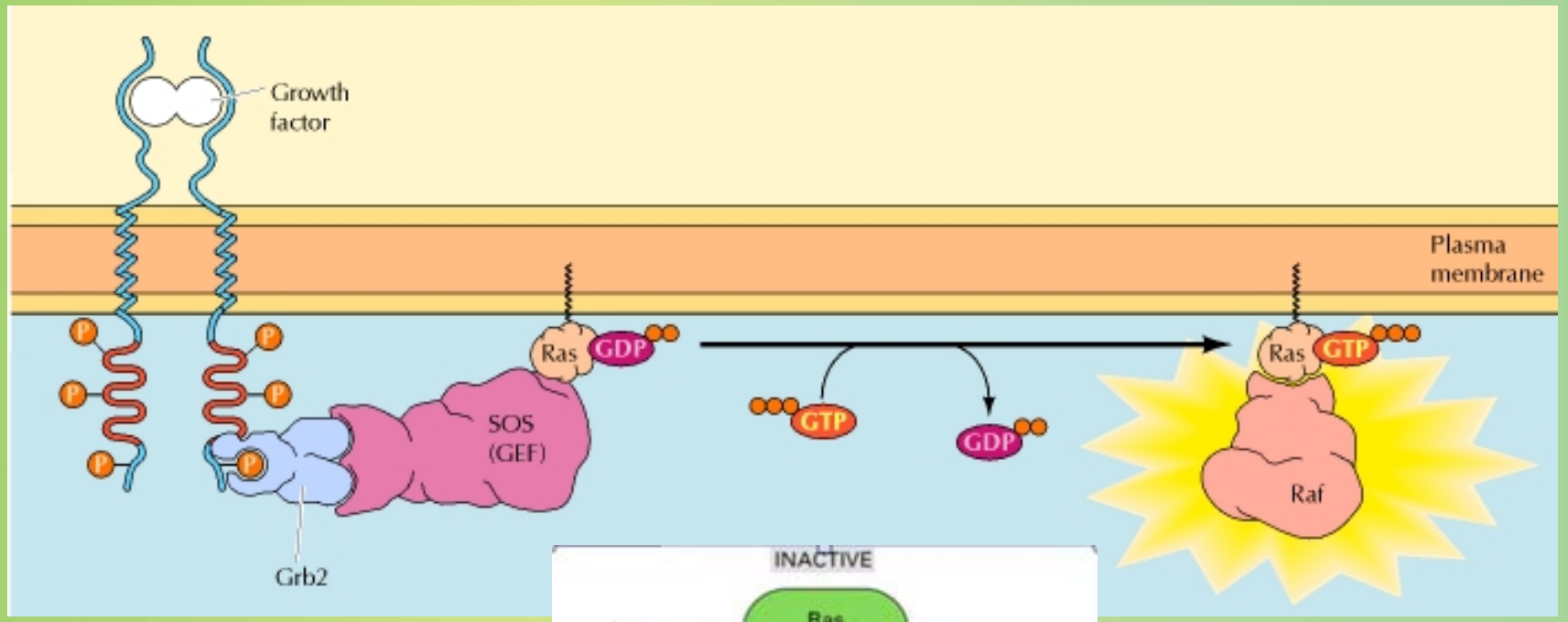
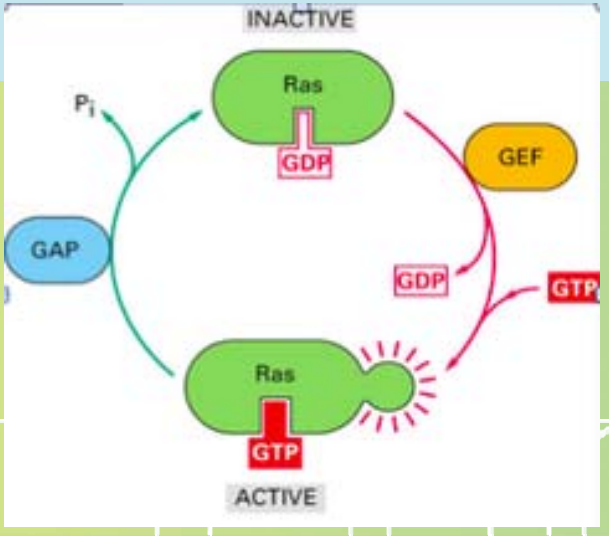


Figure 1

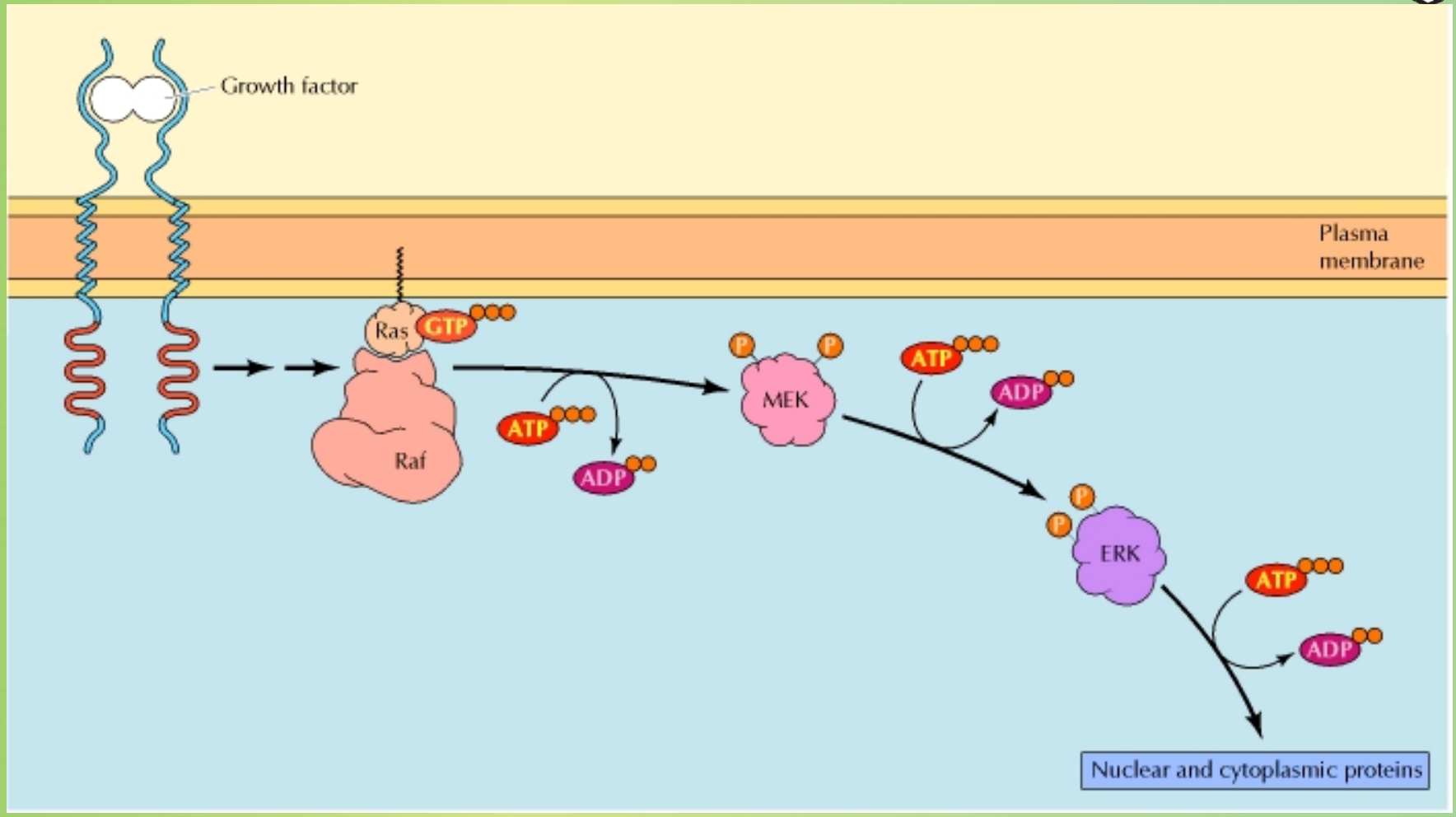
Ras activation by RTKs

Ras a member of the small GTP-binding protein



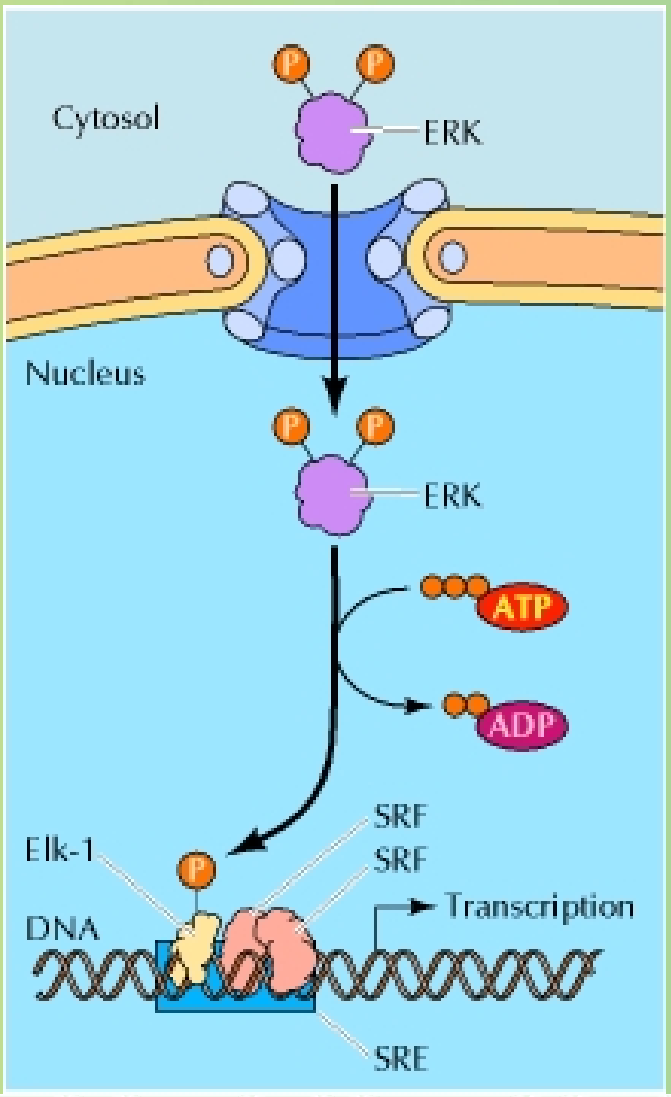


MAP kinase pathway



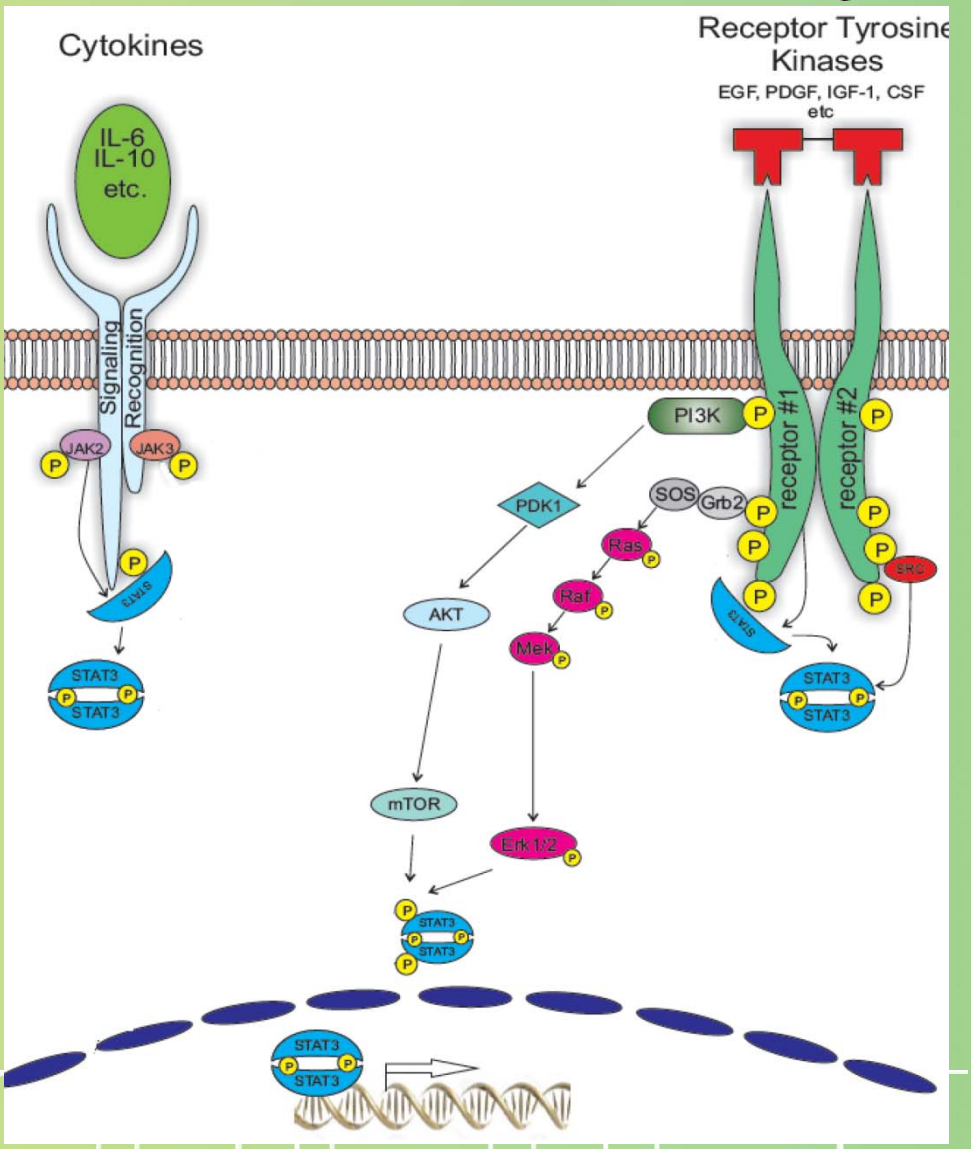
ERK induction of immediate-early genes

- ERK translocates to the nucleus and phosphorylates the transcription factor Elk-1.
- Elk-1 binds to the serum response element (SRE) in a complex with serum response factor (SRF).
- Phosphorylation stimulates Elk-1 and expression of immediate-early genes.
- These genes stimulate expression of secondary response genes.
- The ERK signaling leads to cell proliferation, survival, and differentiation.



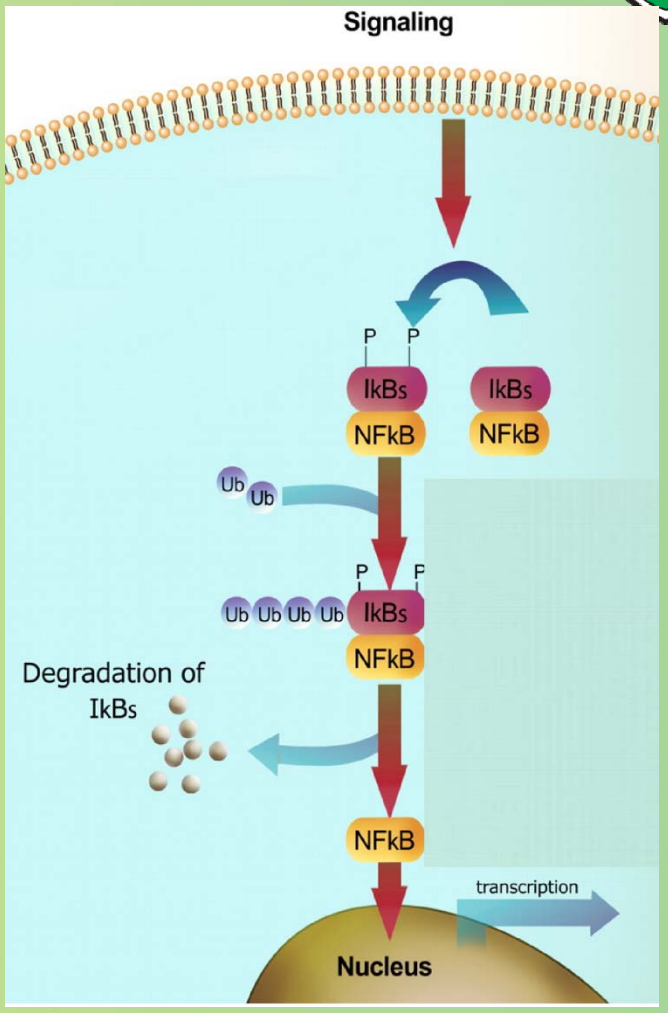
Regulation of gene expression by STATs

- STATs (transcription factors) link non-receptor tyrosine kinase pathways (like JAK pathway) to MAP kinase-regulated RTK pathways.
- Phosphorylation of STATs by the receptors themselves or receptor-associated kinases promotes their dimerization and translocation to the nucleus, where they stimulate transcription of their target genes.



NF-κB signaling

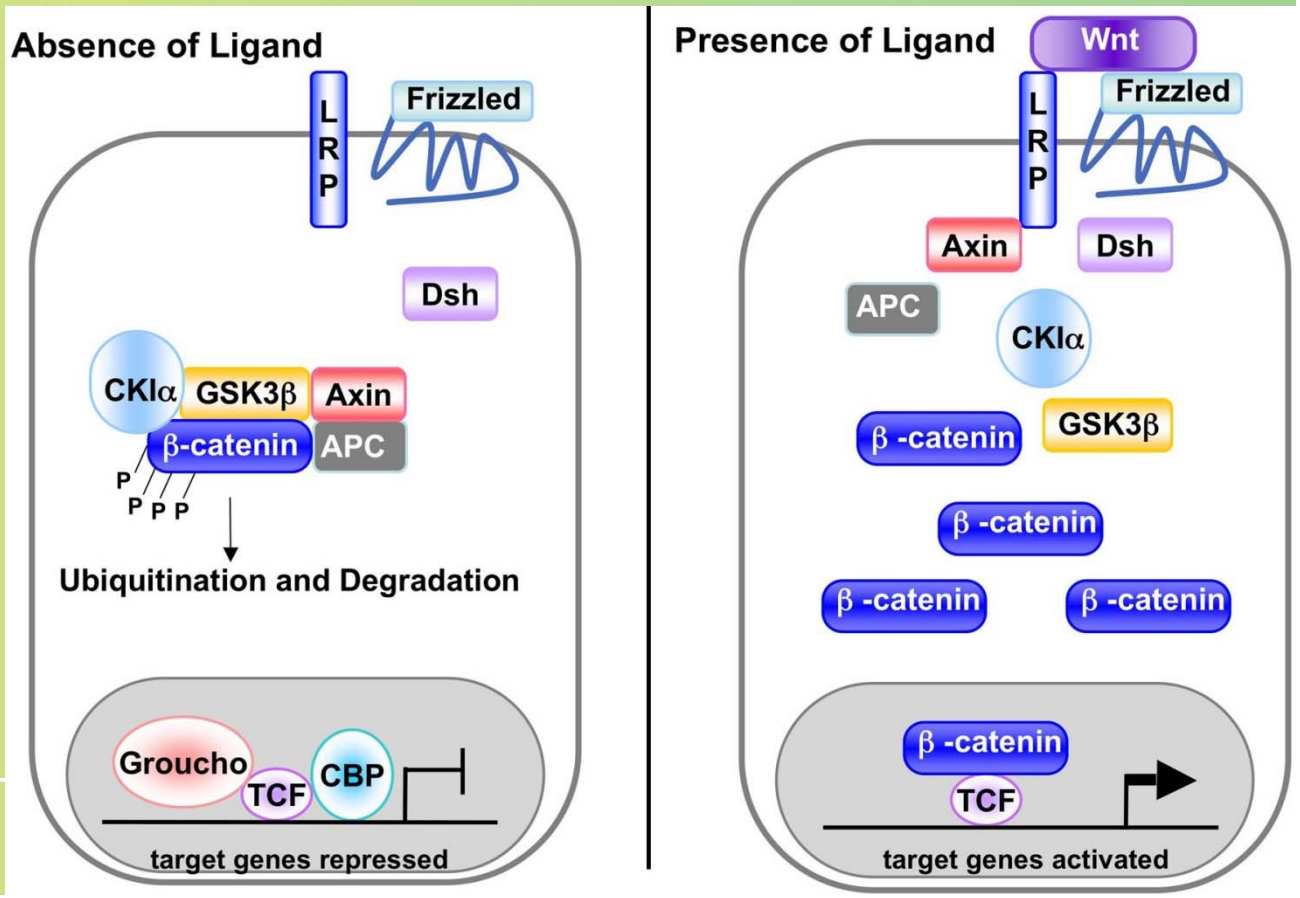
- Tumor necrosis factor activates its receptor TNF receptor and induces inflammation and cell death via activation of the transcription factor NF-κB by stimulating the phosphorylation and degradation of IκB.



Wnt signaling

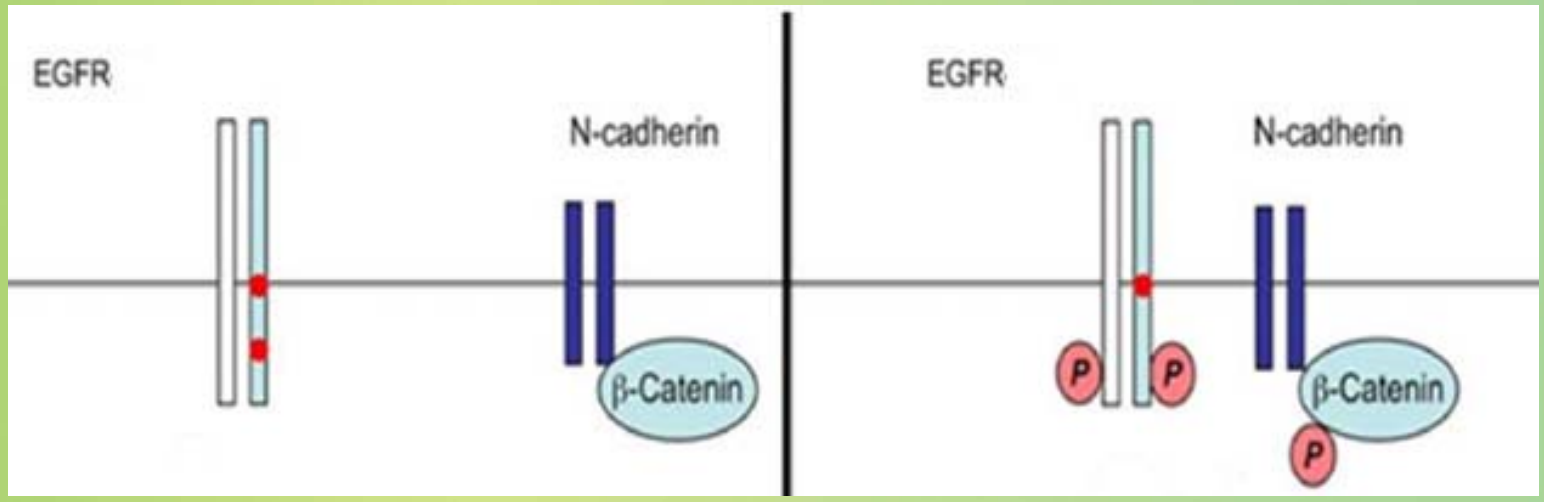
- Wnt proteins are growth factors that bind to the Frizzled receptors and block β-catenin degradation.
- β-catenin can then translocate into nucleus and activate gene expression by Tcf.

Remember:
β-catenin links cadherins to actin.



Role of adhesion molecules in signaling

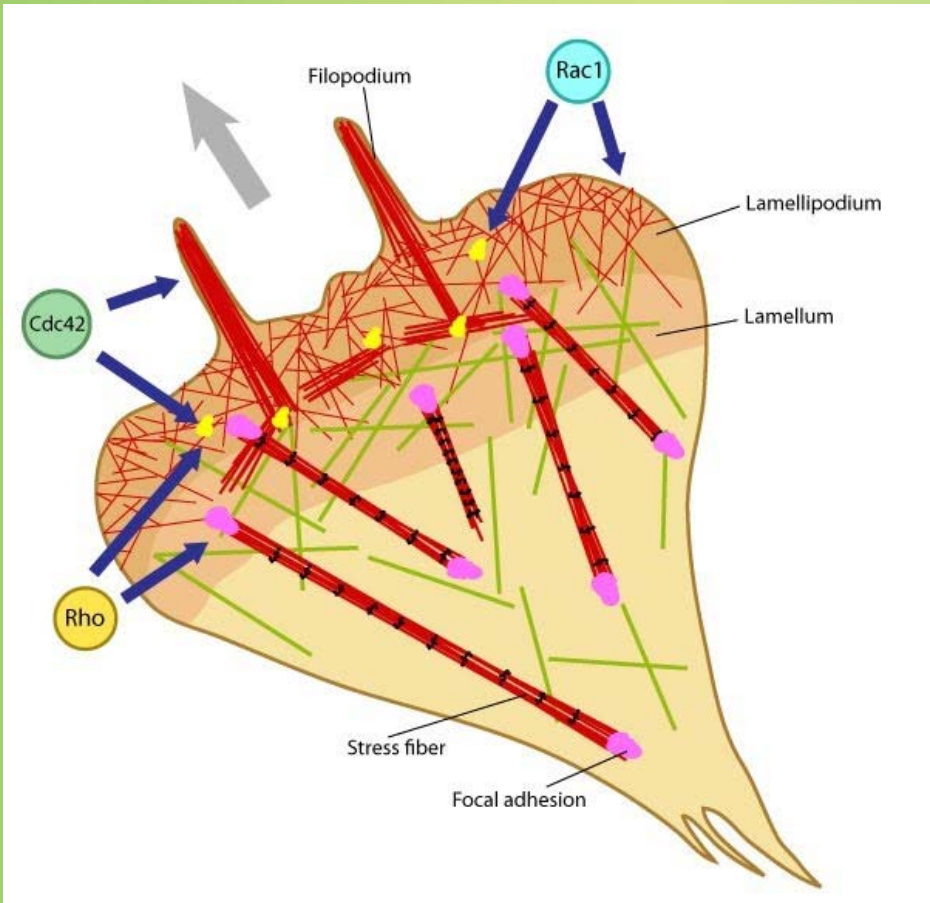
- **Interaction of cadherins with cell surface receptors result in dual regulation and signaling and promotion of cell survival.**

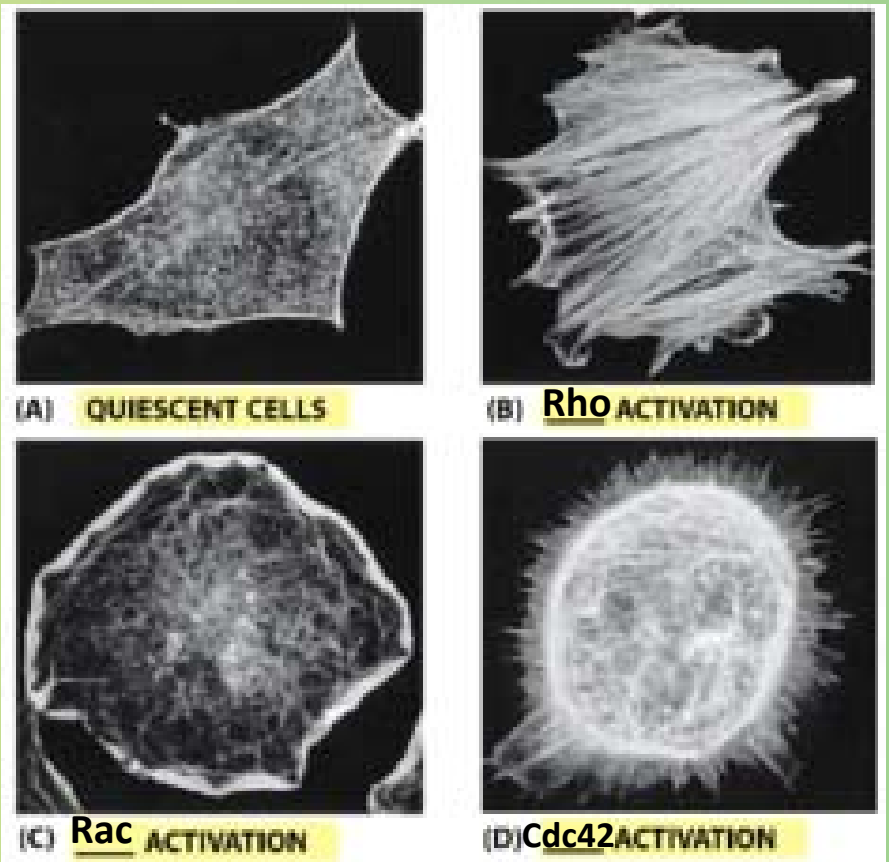


The Rho subfamily

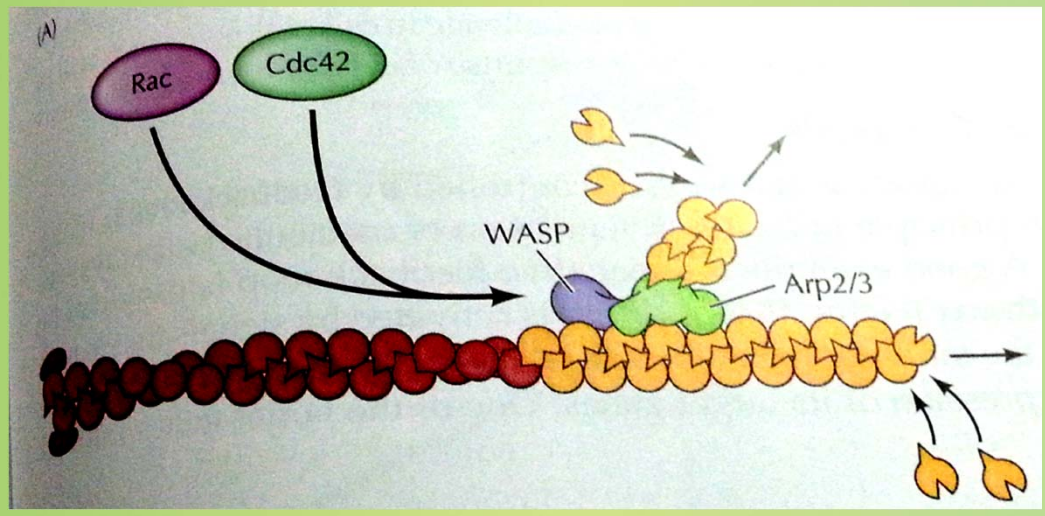
- Members of the Rho subfamily of small GTP-binding proteins (including Rho, Rac, and Cdc42) regulate the organization of the actin cytoskeleton (cell motility, cell adhesion, and cytokinesis).

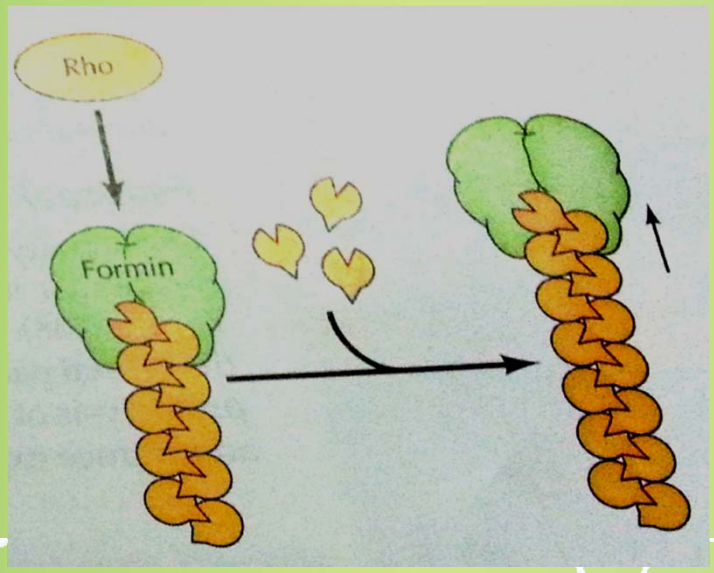
Biological effects

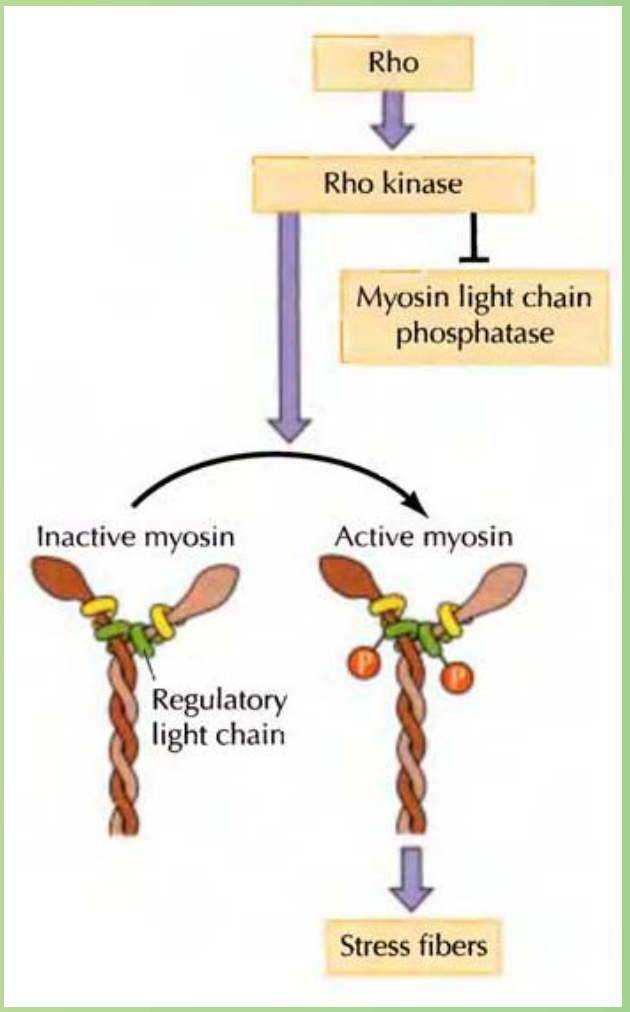




Mechanisms of action

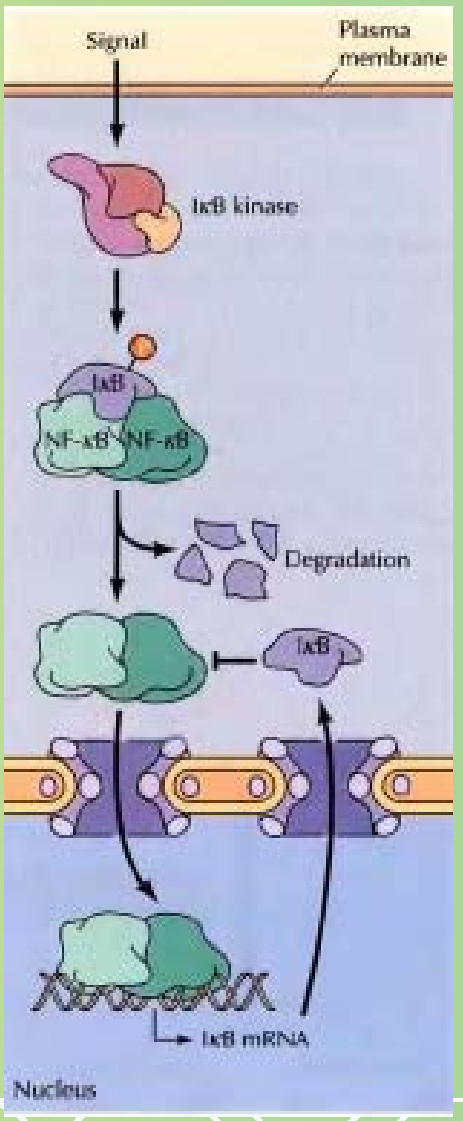






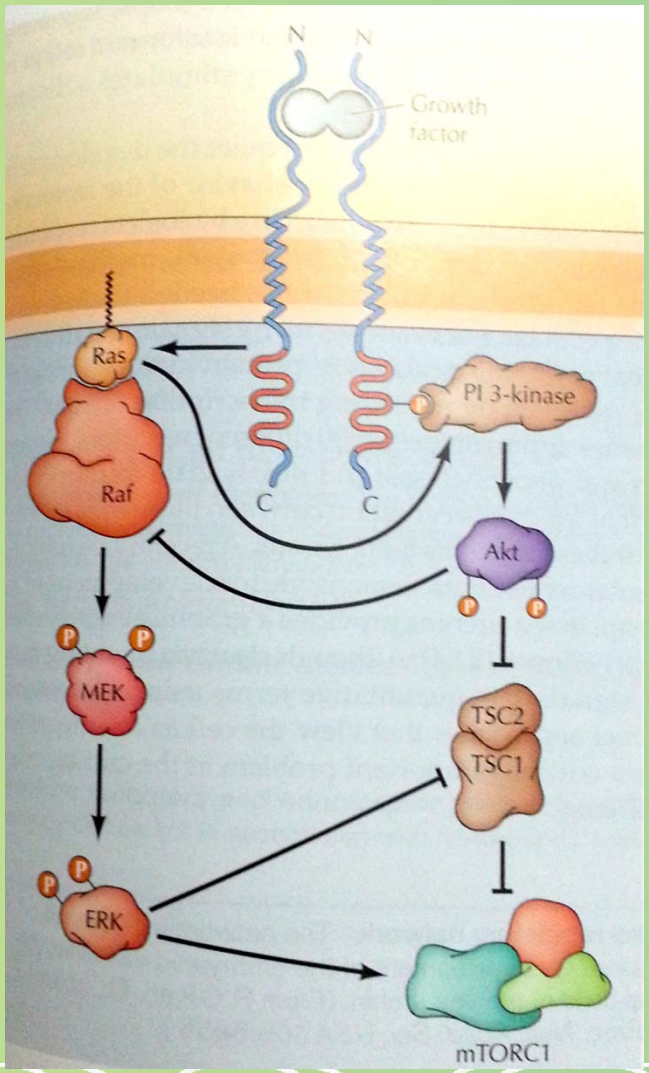
Signaling networks and regulation

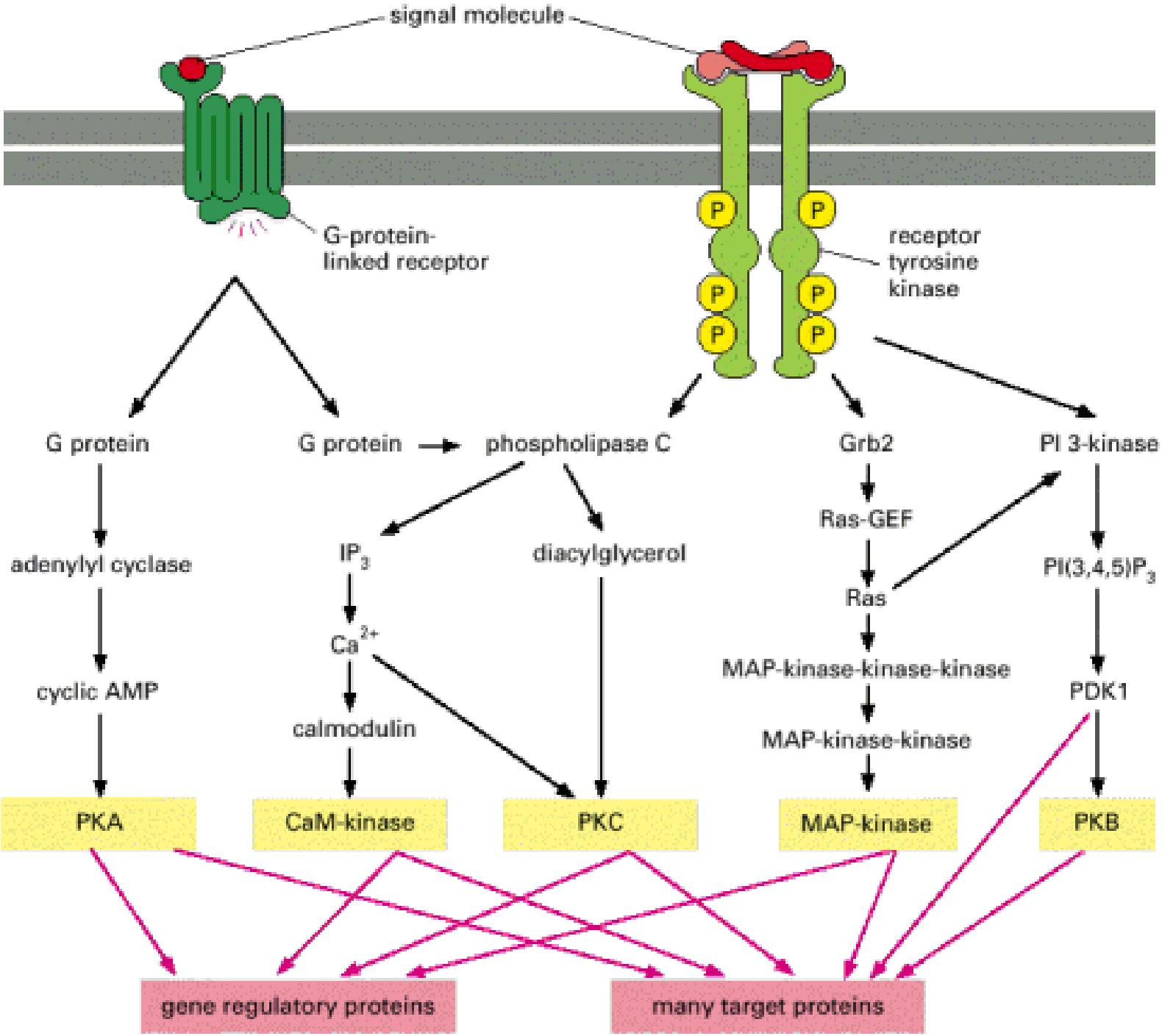
- An activation of one pathways leads to expression of its inhibitors.



Crosstalk

- The interaction of one signaling pathway with another.
- Examples:
  - cAMP and ERK
  - Cell adhesion molecules and receptor tyrosine kinases
  - ERK and PI-3 kinases




signal molecule
G-protein-
linked receptor
receptor
tyrosine
kinase
P
G protein
G protein
phospholipase C
Grb2
PI 3-kinase
adenylyl cyclase
IP3
diacylglycerol
Ras-GEF
Ras
PI(3,4,5)P3
cyclic AMP
Ca2+
MAP-kinase-kinase-kinase
PDK1
calmodulin
MAP-kinase-kinase
PKA
CaM-kinase
PKC
MAP-kinase
PKB
gene regulatory proteins
many target proteins

Cell-specific response. Why?

- Cells have distinct receptors.
- Cells contain a different combination of regulatory proteins that that influence cell behavior.
- The final effector (transcription factor) must have access to its DNA-binding site and if the chromatin is packaged tightly, the complex will not be able to bind to the DNA and, hence, activate transcription.