

Signal Transduction

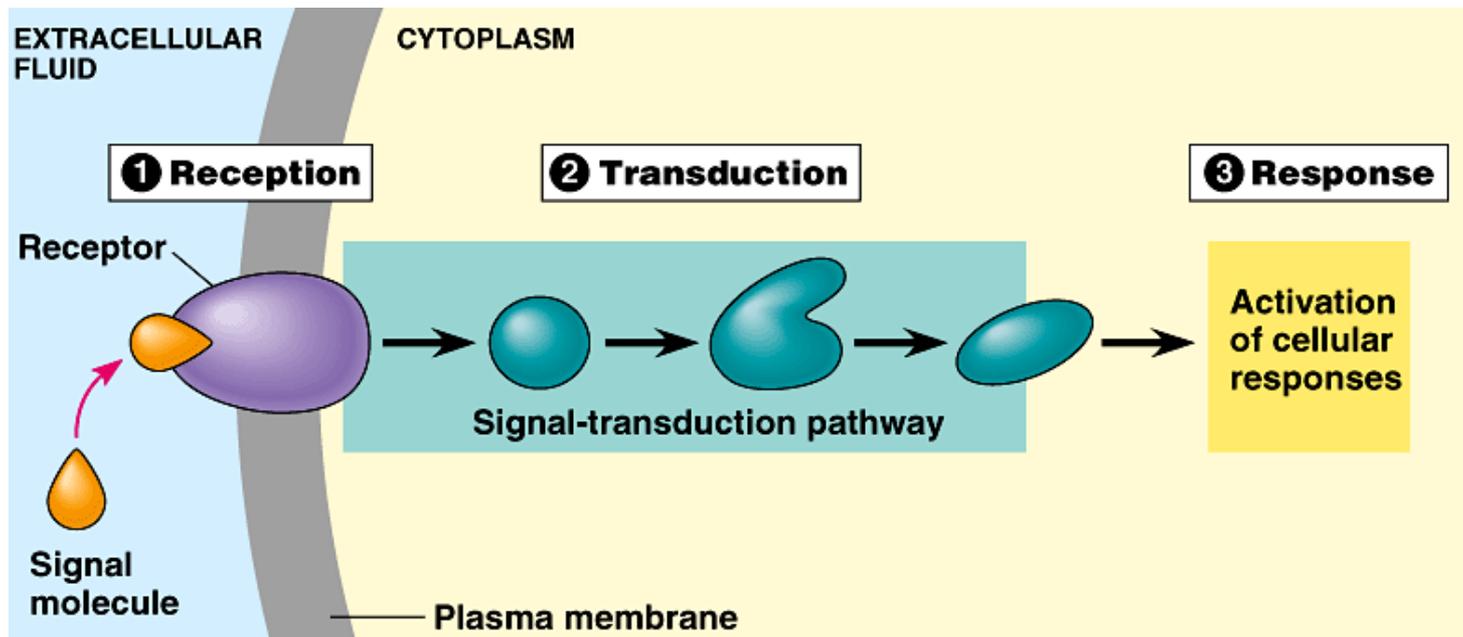
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Cell to Cell Communication

Can occur in both eukaryotes and prokaryotes, mostly through chemical signals to trigger specific reactions in the cell

May cause growth, the production of specific molecules, and other actions (like muscle contraction)

Signal transduction occurs when a signaling molecule activates a receptor. In turn, this receptor alters intracellular molecules creating a response.



Signal transduction - general steps

- 1) Synthesis and release of signaling molecules
- 2) Transport of signal to target cell
- 3) Detection of signal by a specific receptor protein
- 4) Intracellular events, often mediated by second messengers, that change metabolism or gene expression
- 5) Termination of the signal and response

Reception: Chemical signal binds to the receptor molecule (on cell surface or inside cell)

Transduction: Reaction or series of reactions (cascade) leading to the final response

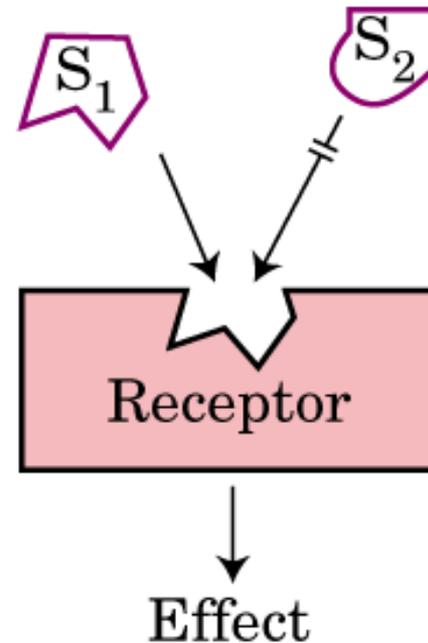
Response: The specific cellular response

Requirement of Biosignaling

- requires a receptor to detect signals;
- the receptor must link to or generate an intracellular response;
- Such linking molecules are known as “second messengers”;
- This transduction system must meet four specific criteria.

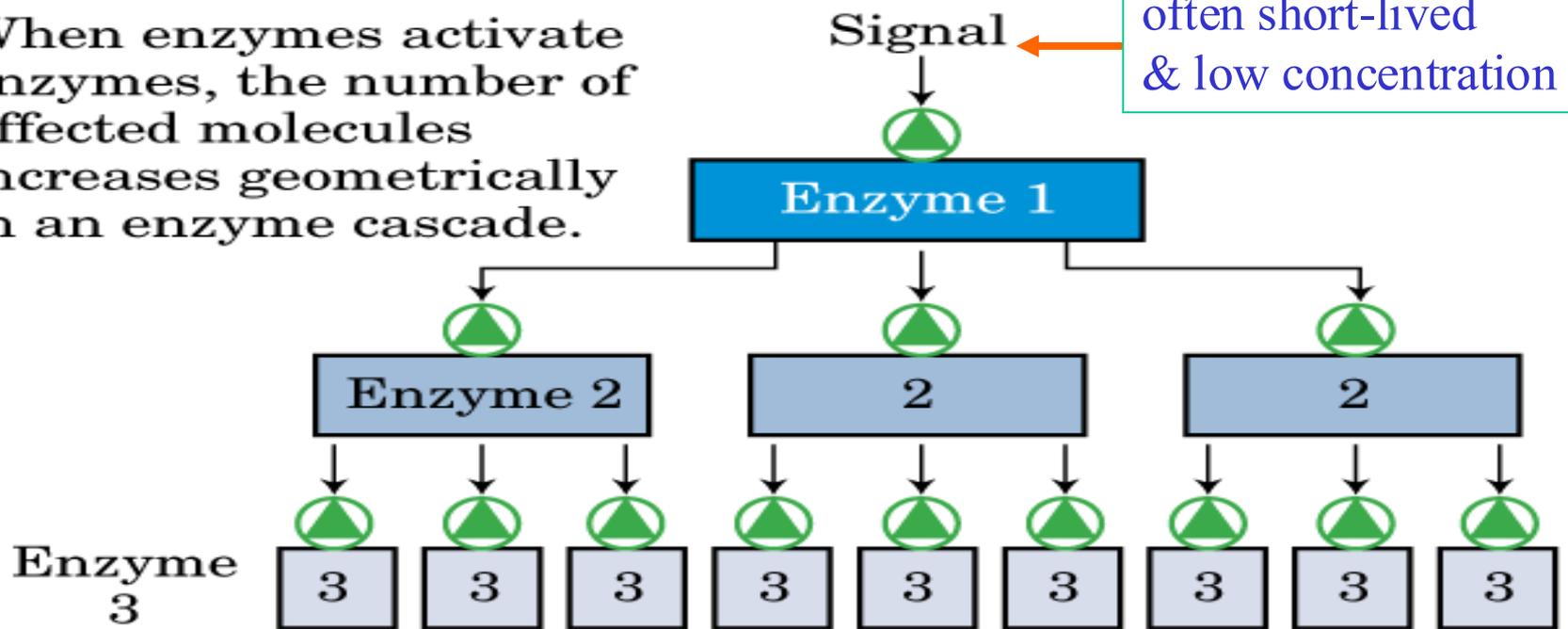
(a) Specificity

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



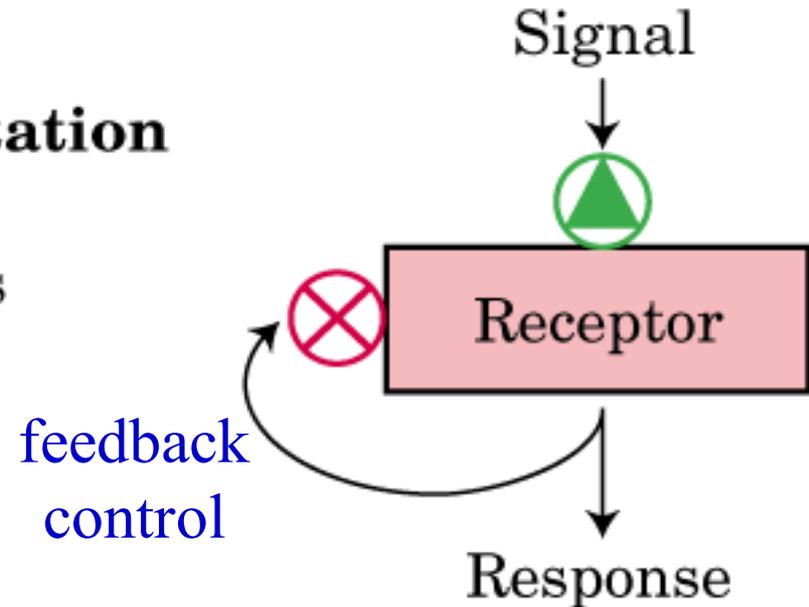
(b) Amplification

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



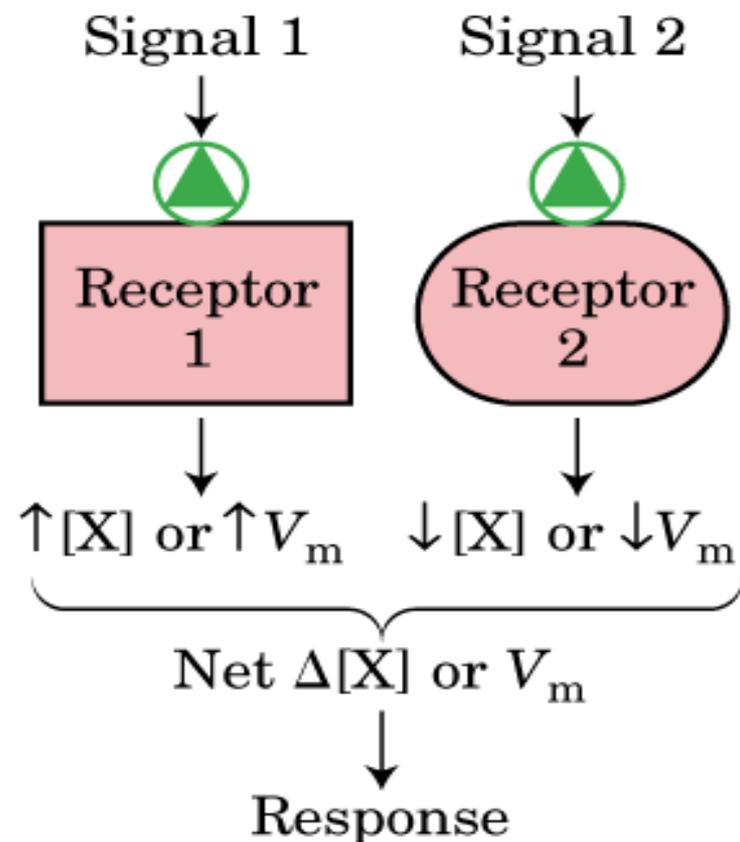
(c) Desensitization/Adaptation

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



(d) Integration

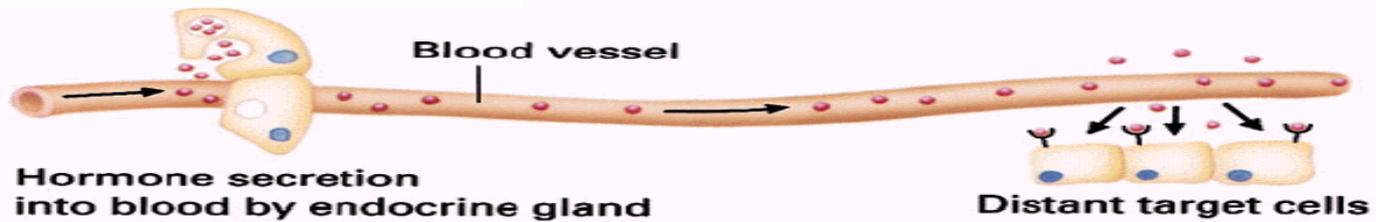
When two signals have opposite effects on a metabolic characteristic such as the concentration of a second messenger X , or the membrane potential V_m , the regulatory outcome results from the integrated input from both receptors.



TYPES OF SIGNALING

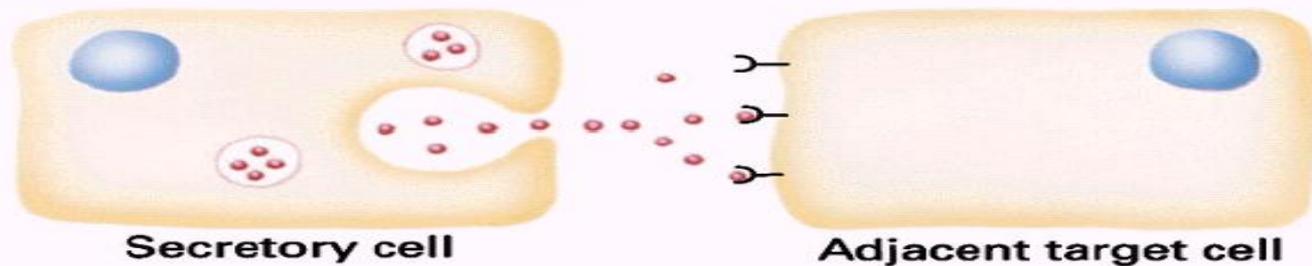
- Endocrine signaling – signaling molecules act on target cells distant from their site of synthesis by cells of endocrine organs
- Paracrine signaling – signaling molecules released by a cell only affect target cells in close proximity
- Autocrine signaling – cells respond to substances that they themselves release
- Receptor-counter receptor signaling – target cells respond to plasma membrane-bound molecules on signaling cells

ENDOCRINE SIGNALING



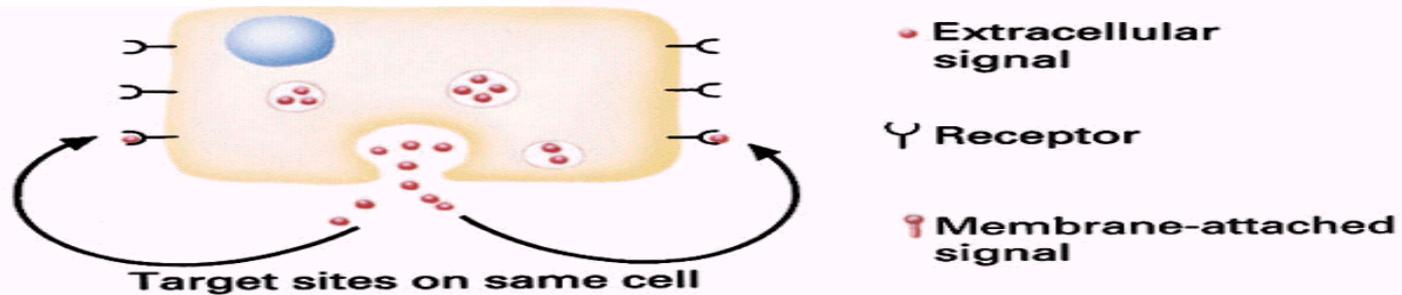
Examples: peptide and steroid hormones

PARACRINE SIGNALING



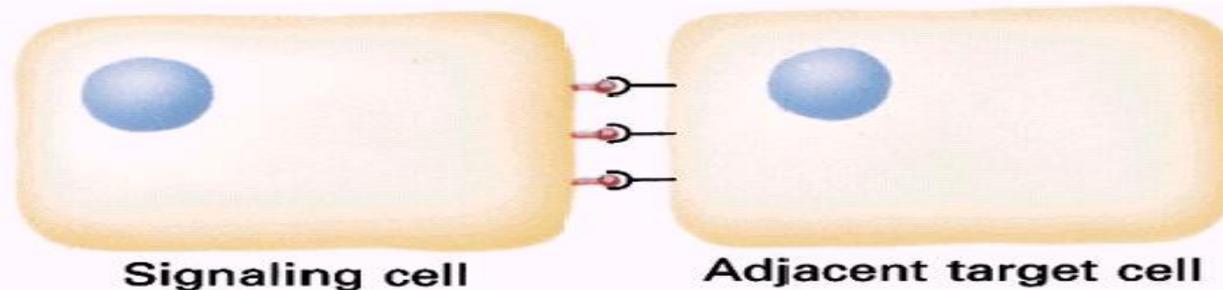
Examples: nerve-nerve, nerve-muscle cells, cytokines

AUTOCRINE SIGNALING



Examples: cytokines/immune cells, growth factors

SIGNALING BY PLASMA MEMBRANE-ATTACHED PROTEINS



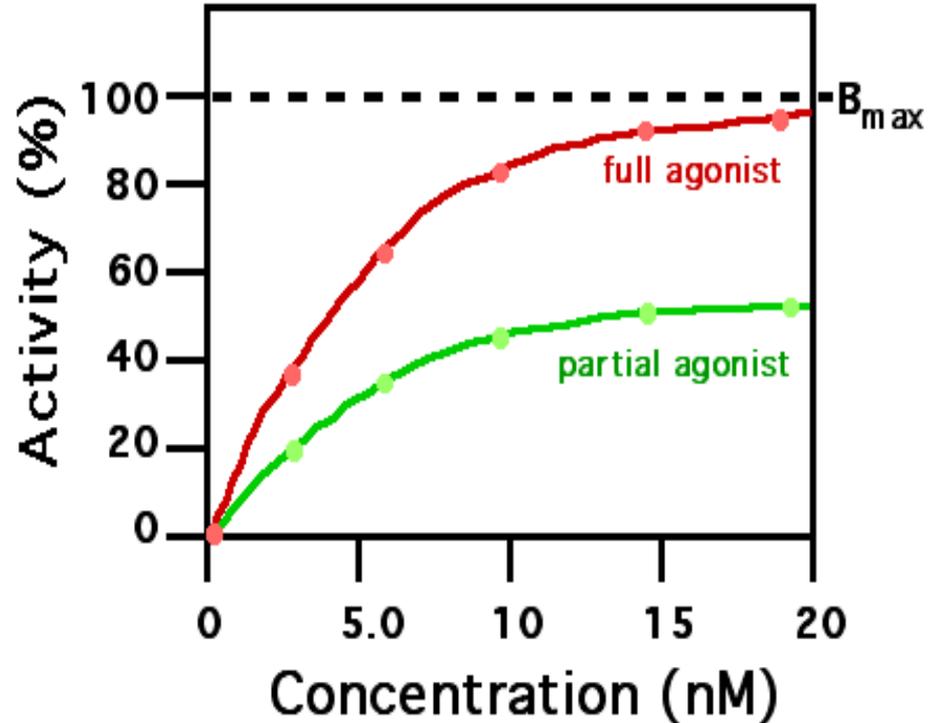
Example: T lymphocytes and antigen presenting cells

Ligands (signalling molecules)

Ligand: it is a substance (usually a small molecule), that forms a complex with a biomolecule (receptor) by intermolecular forces (such as ionic bonds, hydrogen bonds and van der Waals forces) to serve a biological purpose.

The tendency or strength of binding is called affinity and the association is usually reversible.

A ligand that can bind to a receptor, alter the function of the receptor and trigger a physiological response is called an agonist for that receptor.



Ligands that bind to a receptor but fail to activate the physiological response are receptor antagonists.

Receptors

Receptors are protein molecules, embedded in either the plasma membrane or the cytoplasm or nucleus of a cell, to which one or more specific kinds of signalling molecules may attach.

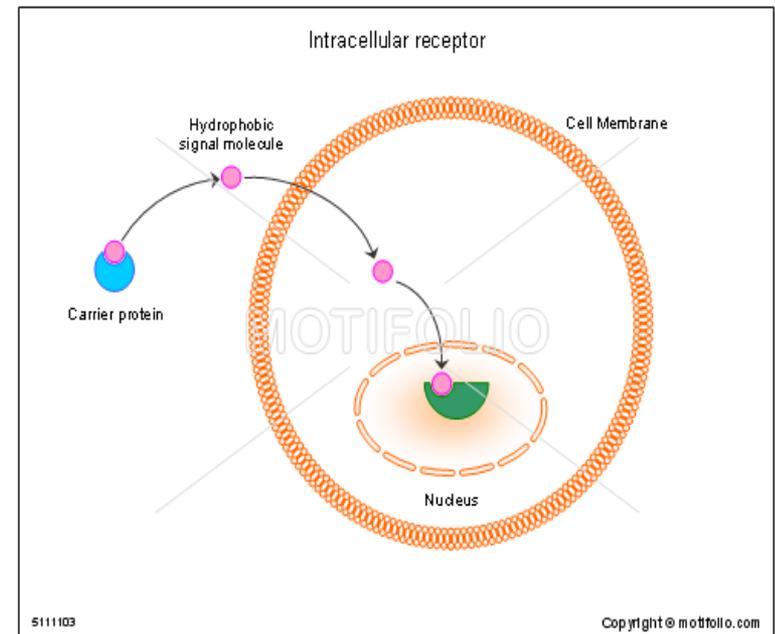
Each type of receptor recognizes and binds only certain ligand shapes (in analogy to a lock and key where the lock represents the receptor and the key, its ligand).

Classification of receptors based on their location

1- Intracellular receptors:

found in the cytoplasm or inside nucleus and function in the nucleus as transcription factors to alter the rate of transcription of particular genes.

The steroid hormones and thyroid hormones are hydrophobic and thus diffuse freely across the plasma membrane and bind to intracellular receptors.

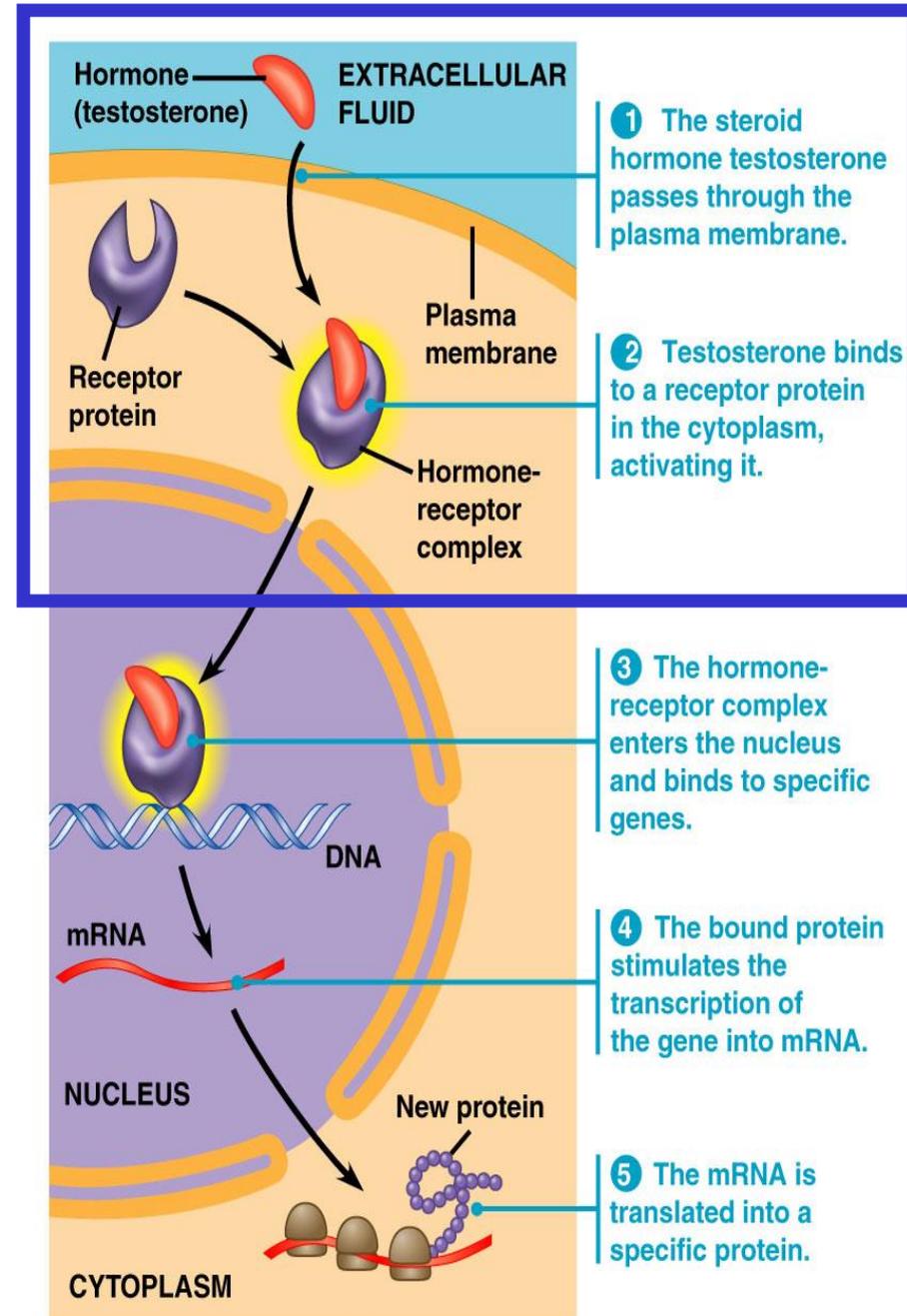


Intracellular receptors

They are receptors located inside the cell rather than on its cell membrane.

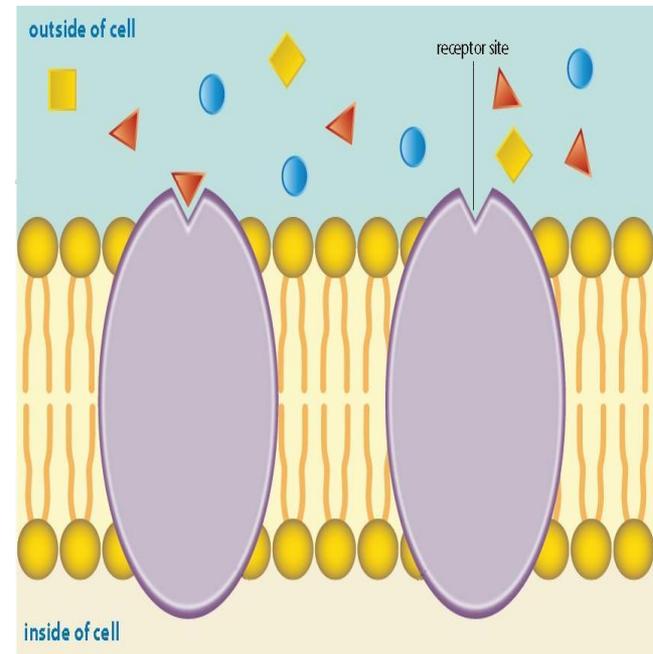
The typical ligands for nuclear receptors are usually intracellular second messengers like inositol trisphosphate (IP₃), and lipophilic hormones like the steroid hormones testosterone and progesterone and derivatives of vitamins A and D

To initiate signal transduction, the ligand must pass through the plasma membrane. On binding with the receptor, the ligands pass through the nuclear membrane into the nucleus, enabling gene transcription and protein production.



2- Plasma-membrane receptors:

Cell surface receptors are specialized integral membrane proteins that take part in communication between the cell and the outside world. Extracellular signaling molecules attach to the receptor, triggering changes in the function of the cell. The binding initiates a chemical change on the intracellular side of the membrane.



The peptide, protein hormones and the catecholamine hormones are hydrophilic and thus interact with cell surface receptors and transmit their signals through second messenger that are generated intracellularly

Four main types of extracellular receptors

- 1-Ion-channel receptors
- 2-G protein-linked receptors
- 3-Receptors associated with tyrosine kinases
- 4-Receptors with intrinsic enzymatic activity

Structure of cell surface receptors:

- 1- Extracellular domain (N-terminal) which binds the hormone.
- 2- Transmembrane domain: One or more than one membrane spanning region that are α -helices, this varies in structure from a simple linear hydrophobic region to a more complex (eg G-receptors).
- 3- Intracellular domain (C-terminal) which initiate the intracellular signaling cascade (Signal transduction).

- **Mechanisms of signal transduction that follow the binding of signaling molecules to plasma membrane receptors include:**

- 1- Phosphorylation of receptors at tyrosine residues (receptor tyrosine kinase activity),
- 2- Conformational changes in signal transducer proteins (e.g., proteins with SH2 domains, heterotrimeric G proteins)
- 3- Increases in the levels of intracellular second messengers
Examples: cAMP, inositol trisphosphate (IP3), and diacylglycerol (DAG).

G-protein Coupled Receptors (GPCRs)

- G proteins bind GTP (guanosine triphosphate), they control and amplify intracellular signaling pathways

Exist in two states

- 1) bound GTP: active
- 2) bound GDP: inactive

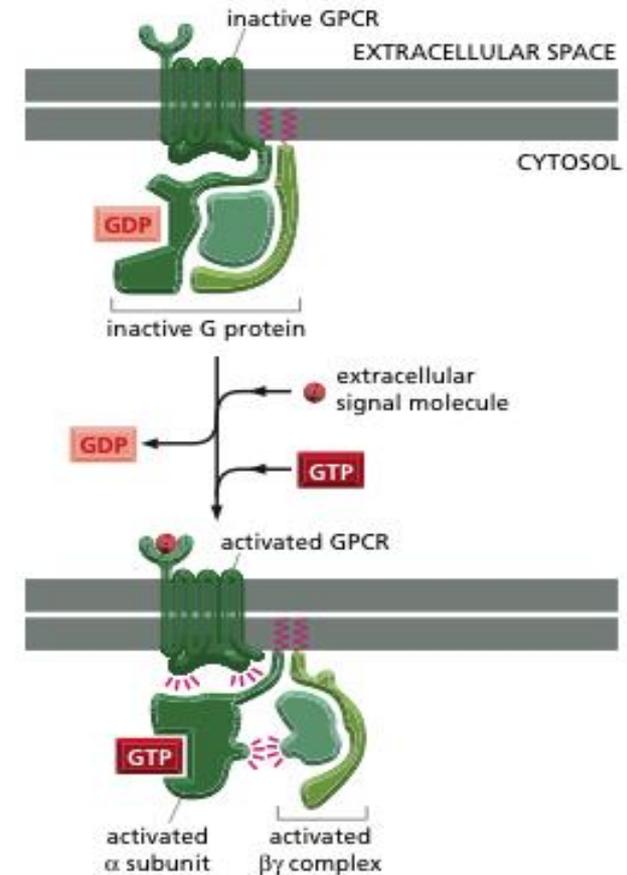
- The extracellular domain connects to the intracellular domain through seven transmembrane spans

- The intracellular domain is coupled to a heterotrimeric G-protein

- The heterotrimeric G-protein is composed of 3 subunits: $G\alpha$, $G\beta$, and $G\gamma$

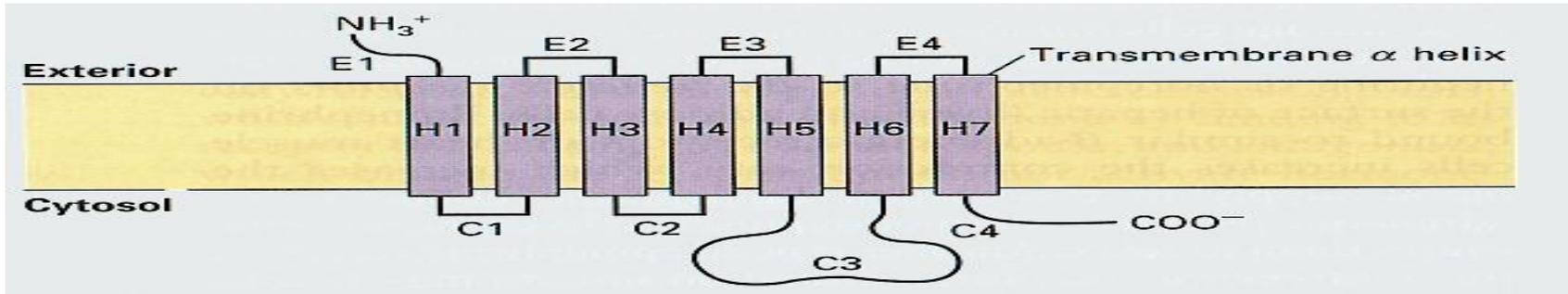
- When the $G\alpha$ subunit is bound to GDP it is “OFF”; and when it is bound to GTP it is “ON”

- The conformational change relayed to the intracellular domain causes the $G\alpha$ subunit to release GDP and bind to GTP thereby activating both the $G\alpha$ and $G\beta/G\gamma$ subunits



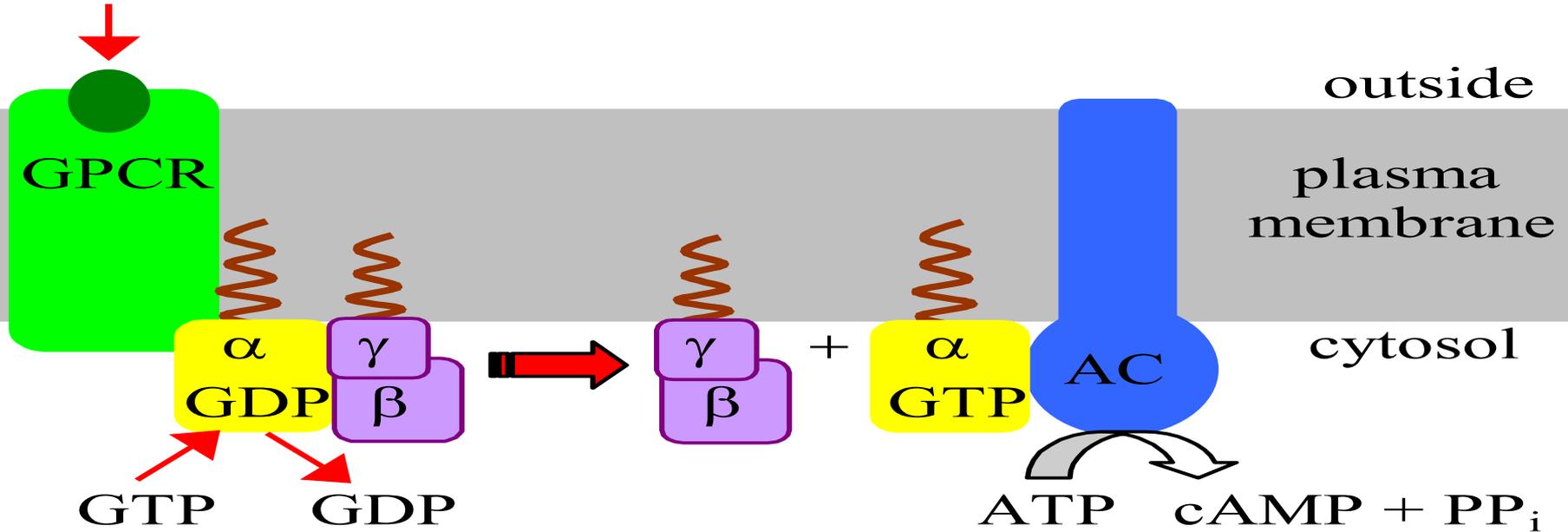
General structure of G protein-linked receptors

- In the transmembrane domain, there are seven alpha helices
- Loop between helices 5&6 involved in interactions with G protein



- When all subunits ($G\alpha$, $G\beta$, and $G\gamma$) are associated together, the G protein is in an **inactivate state**
- Once epinephrine or other cAMP-linked hormones bind to the receptor, the hormone-receptor complex interacts with the G protein to bring about its activation;
- Following interaction of the hormone-receptor complex with the G protein, GTP displaces GDP;
- Binding of GTP produces a conformational change in the G protein that causes the α subunit to dissociate. The α subunit then interacts with adenylate cyclase;
- The α subunit is inactivated when the bound GTP is hydrolyzed to become GDP, which is catalyzed by a GTPase activity that is part of the α subunit.

hormone
signal



The α subunit of a G-protein (G_{α}) binds **GTP**, & can hydrolyze it to **GDP + P_i**.

Adenylate Cyclase (AC) is a transmembrane protein, with cytosolic domains forming the catalytic site.

The sequence of events by which a hormone activates cAMP signaling:

1. Initially G_{α} has bound **GDP**, and $\alpha, \beta,$ and γ subunits are complexed together.

$G_{\beta\gamma}$, the complex of β & γ subunits, **inhibits** G_{α} .

2. **Hormone binding**, usually to an extracellular domain (GPCR), causes a **conformational change** in the receptor that is transmitted to a **G-protein** on the cytosolic side of the membrane.

The nucleotide-binding site on G_{α} becomes more accessible to the cytosol, where $[GTP] > [GDP]$.

G_{α} releases GDP & binds GTP (**GDP-GTP exchange**).

3. Substitution of **GTP** for GDP causes another conformational change in G_{α} .

G_{α} -GTP dissociates from the inhibitory $\beta\gamma$ complex & can now bind to and activate Adenylate Cyclase.

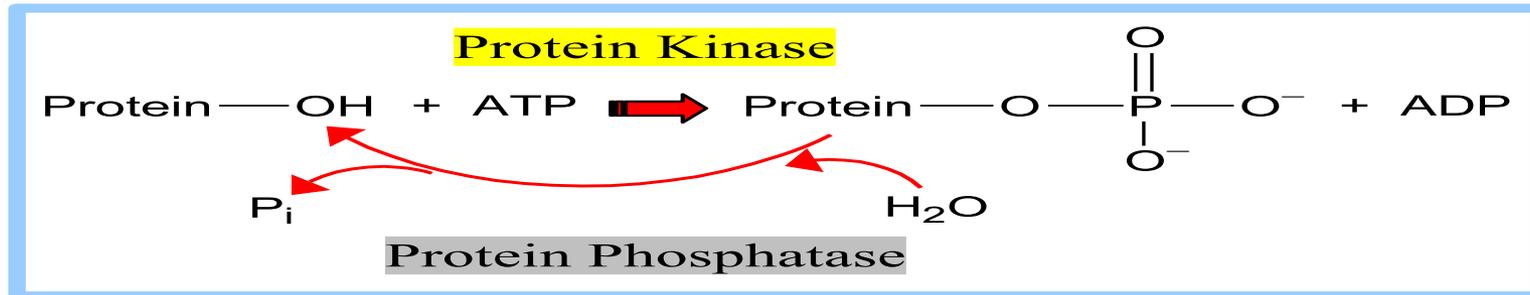
4. **Adenylate Cyclase**, activated by the stimulatory G_{α} -GTP, catalyzes synthesis of **cAMP**.

5. **Protein Kinase A** (cAMP Dependent Protein Kinase) catalyzes transfer of phosphate from ATP to serine or threonine residues of various cellular proteins, altering their activity.

Receptor Tyrosine Kinases (RTKs)

• Protein kinases are enzymes that add a phosphate group from ATP onto a substrate protein; this reaction is called phosphorylation.

RTKs are the high-affinity cell surface receptors for many polypeptide growth factors, and hormones



This reversibility contributes to the dynamic nature of cells

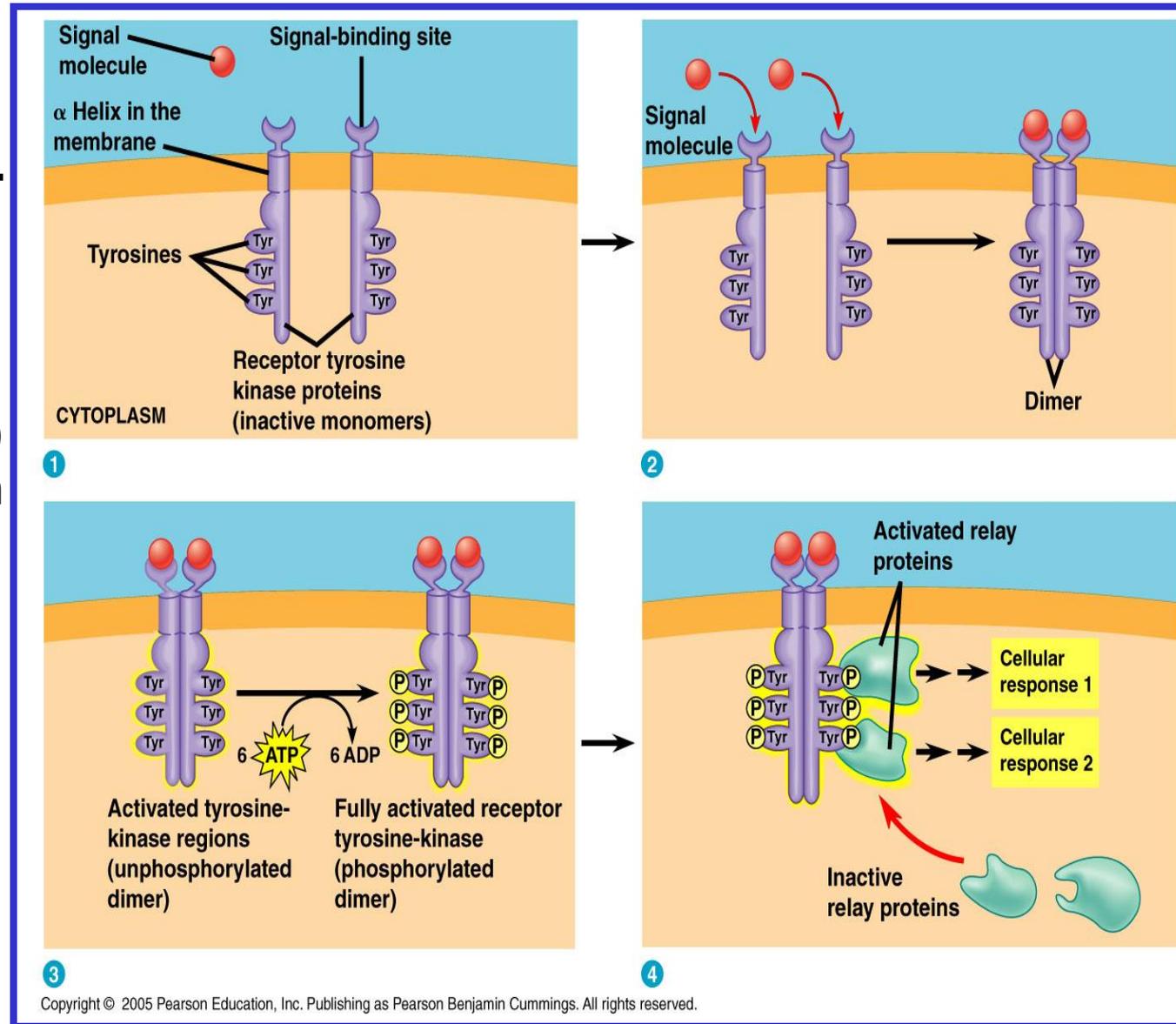
RTKs need to form dimers in the plasma membrane; the dimer is stabilized by ligands binding to the receptor. The interaction between the cytoplasmic domains stimulates the autophosphorylation of tyrosines within the domains of the RTKs, causing conformational changes.

The receptors' kinase domains are subsequently activated, initiating phosphorylation signaling cascades of downstream cytoplasmic molecules that facilitate various cellular processes such as cell differentiation and metabolism

Receptor Tyrosine Kinase

Steps involved:

- 1-Ligand Reception
- 2-Receptor Dimerization
- 3-Catalysis (Phosphorylation)
- 4-Subsequent Protein Activation
- 5-Further Transduction
- 6-Response

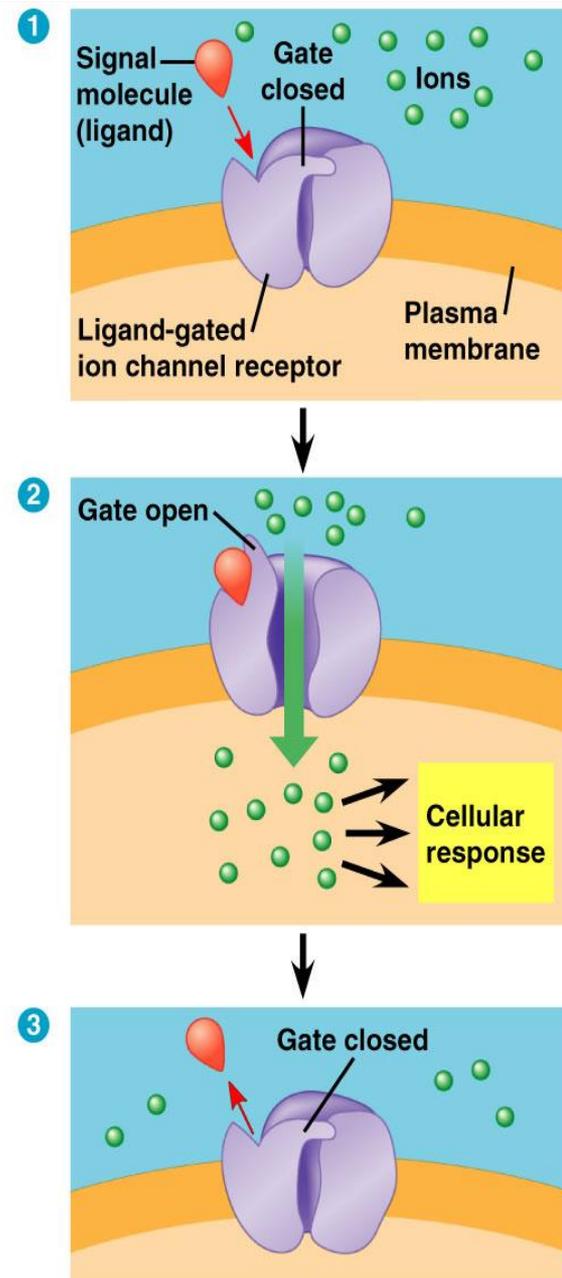


Ion-Channel Receptor

Ion-Channel Receptor are a group of transmembrane ion channels that are opened or closed in response to the binding of a ligand

A ligand-gated ion channel, upon binding with a ligand, changes conformation to open a channel in the cell membrane through which ions relaying signals can pass

An example of an ion allowed into the cell during a ligand-gated ion channel opening is Ca^{2+} ; it acts as a second messenger initiating signal transduction cascades and altering the physiology of the responding cell.



Reversibility is assured by pumping ions back out again (using separate protein)

Second messengers

Small molecules or ions that occur in the cytoplasm of a cell, and they are generated in response to a hormone binding to a cell-surface receptor, and activates various kinases that regulate the activities of other enzymes

But in addition to their job as relay molecules, second messengers serve to greatly amplify the strength of the signal.

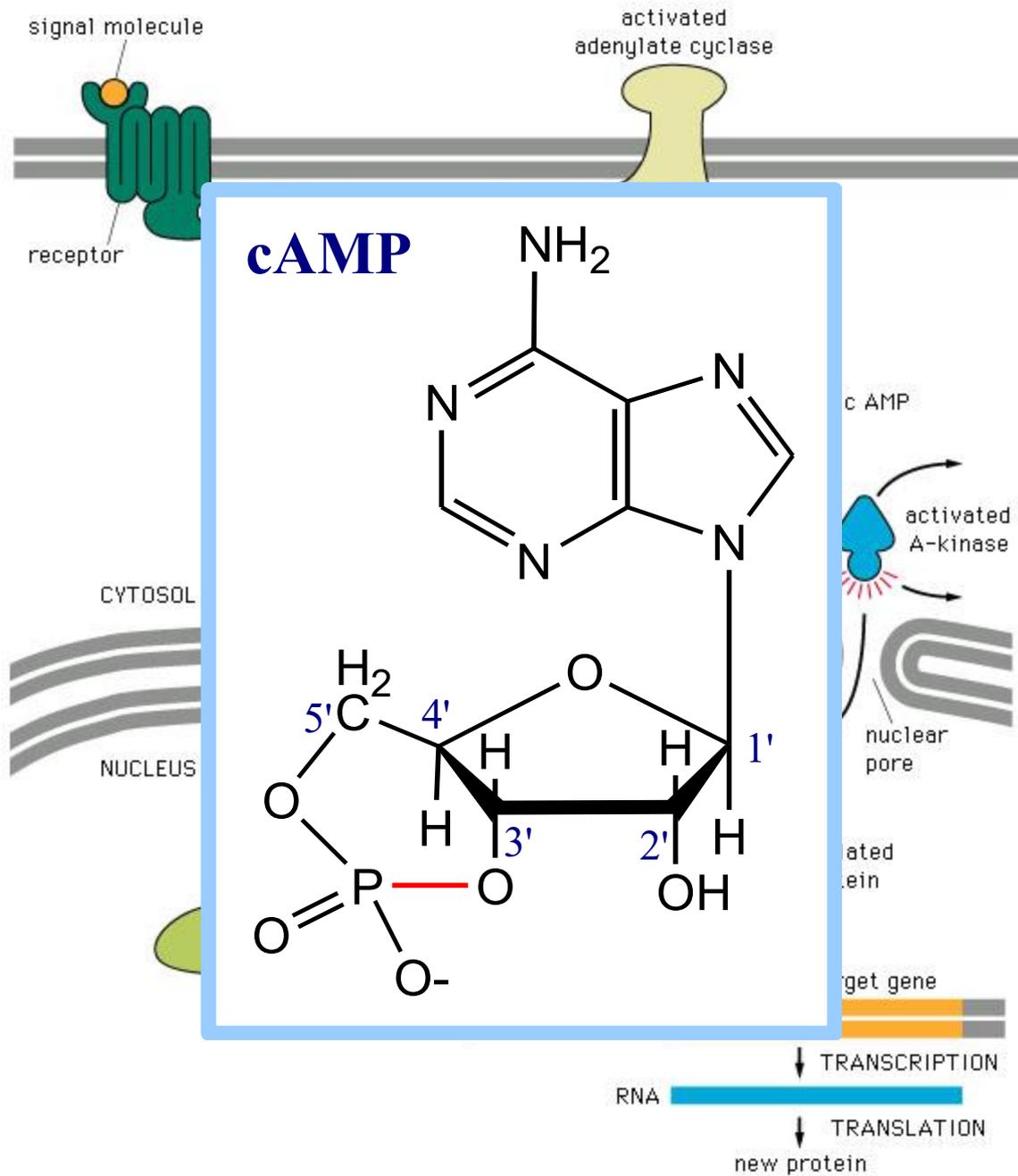
There are three major classes of second messengers:

- 1.cyclic nucleotides (e.g. cAMP)
- 2.inositol trisphosphate (IP₃) and diacylglycerol (DAG)
- 3.calcium ions (Ca²⁺)

The effect of cAMP on cellular metabolism

- Increased [cAMP] increases glycogen breakdown
- Increased [cAMP] increases phosphorylase activity (via a complex cascade)
- Phosphorylase (or glycogen phosphorylase) is a key enzyme involved in glycogen breakdown

Cyclic AMP (cAMP) 2nd Messenger



Inositol trisphosphate (IP₃) and diacylglycerol (DAG)

IP₃ opens channels to release calcium ions from intracellular stores

- IP₃ is able to increase [Ca²⁺] by associating with the IP₃ channel or IP₃ receptor;
- At least three molecules of IP₃ must bind to sites on the cytosolic side of the membrane protein to open the channel and release Ca²⁺.
- Increase [Ca²⁺] → activates protein kinase C (PKC)
- IP₃ is a short-lived messenger (less than a few seconds)

Diacylglycerol activates protein kinase C (PKC)

- PKC phosphorylates Ser or Thr residues of specific target proteins, changing their catalytic activities;

PKC is a family of protein kinase enzymes that are involved in controlling the function of other proteins through the phosphorylation of hydroxyl groups of serine and threonine amino acid residues on these proteins.

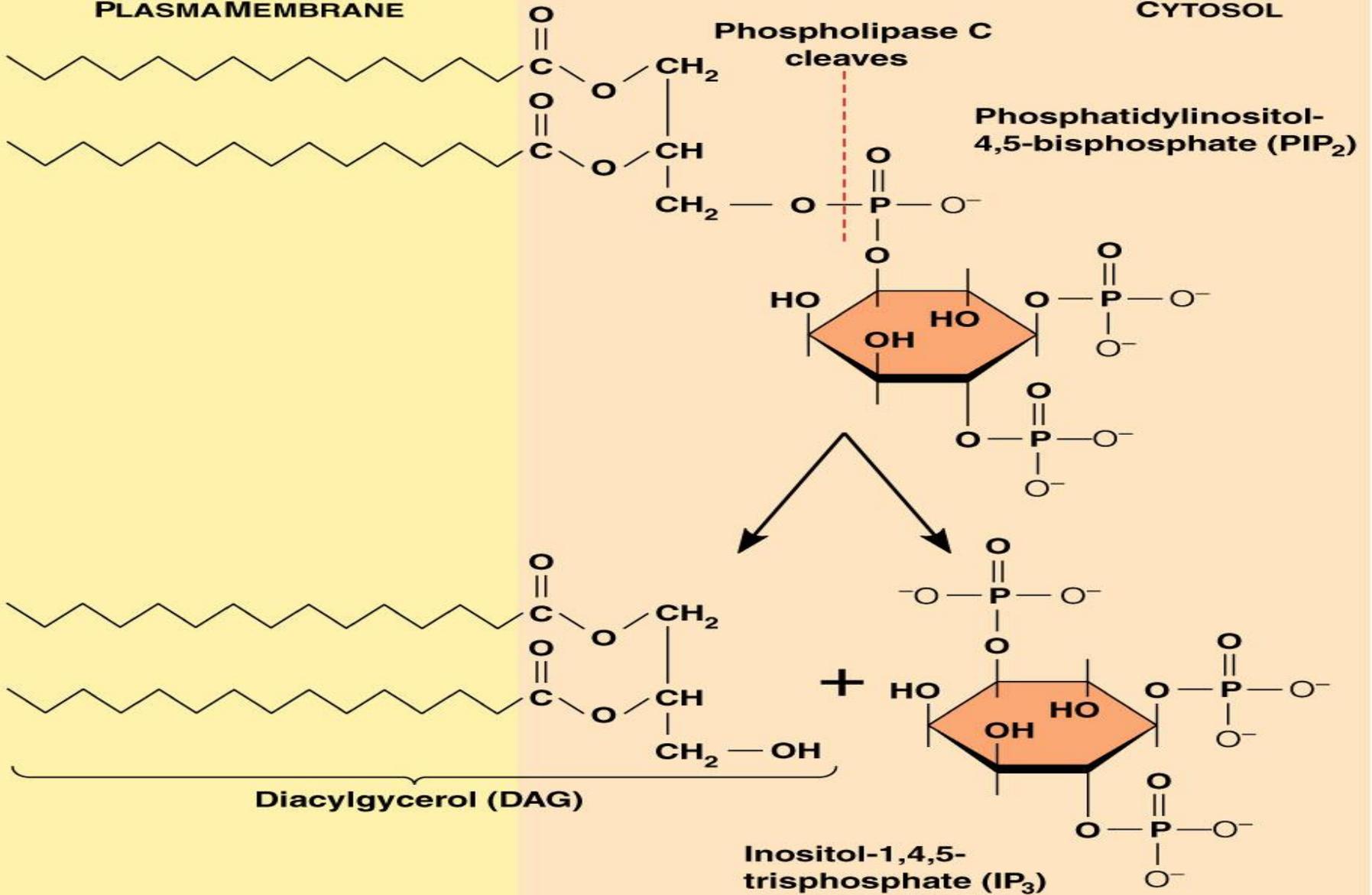
PKC enzymes in turn are activated by signals such as increases in the concentration of diacylglycerol (DAG) or calcium ions (Ca²⁺).

PLASMA MEMBRANE

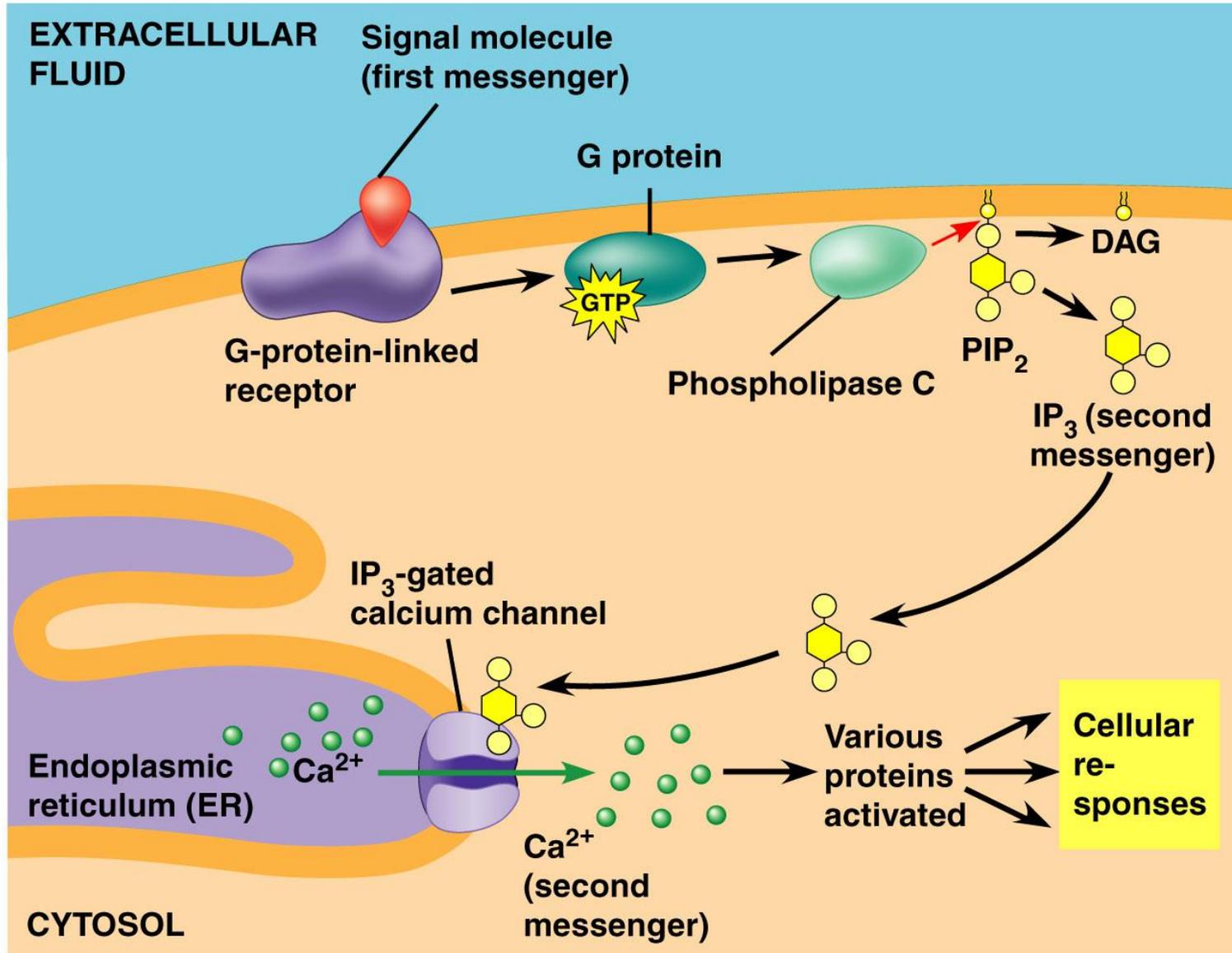
CYTOSOL

Phospholipase C
cleaves

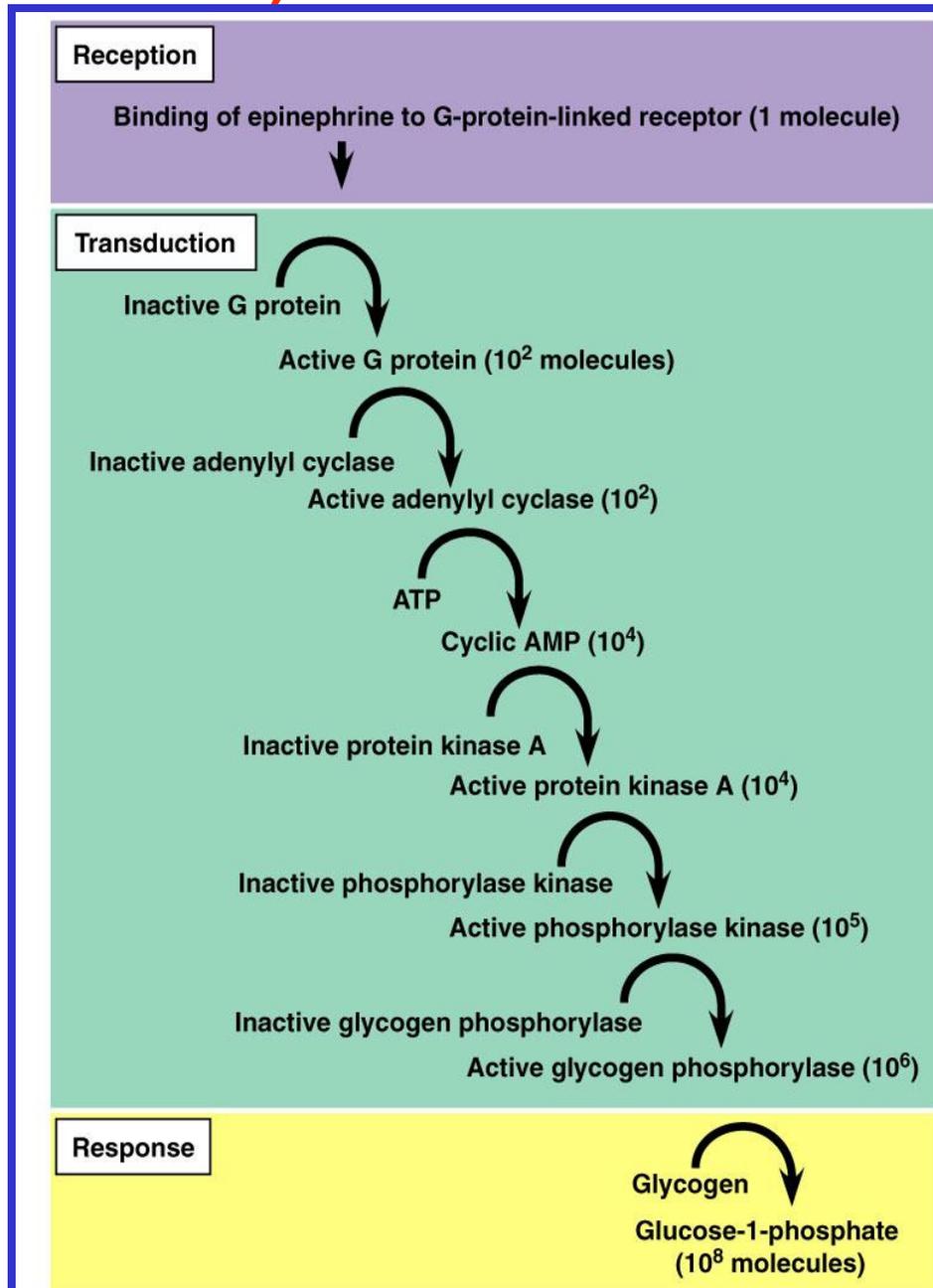
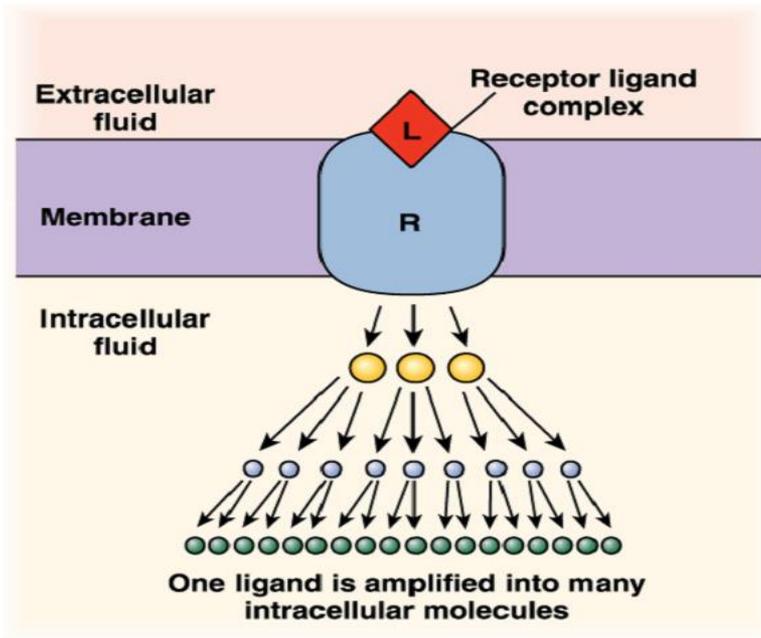
Phosphatidylinositol-
4,5-bisphosphate (PIP₂)



Ca²⁺ as a second messenger



Signal Amplification (Cascade)



Responding to the Signal: Effector Proteins

- The final step in cell signaling is activation of the effector proteins
- The effector proteins carry out the cellular response to the signal
- Often the cellular response involves expression of previously inactive genes which requires effector proteins called transcriptional activators or transcription factors
- Transcription factors are proteins that bind to specific DNA sequences called promoters
- Effector proteins can also directly act on proteins that regulate cell shape to induce changes in morphology by rearranging the cytoskeleton
- Other types of effector proteins directly regulate cell growth by arresting the cell cycle or altering cellular metabolism

Various Responses

