Signal Transduction Lec 3

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SECOND MESSENGER MECHANISMS FOR MEDIATING INTRACELLULAR HORMONAL FUNCTIONS

- **Most water-soluble extracellular chemical messengers activate second-messenger pathways.**
- Hormones can exert intracellular actions is to stimulate formation of the second messenger inside the cell membrane.
- The second messenger then causes subsequent intracellular effects of the hormone.

The only direct effect that the hormone has on the cell is to activate a single type of membrane receptor.

The second messenger does the rest.

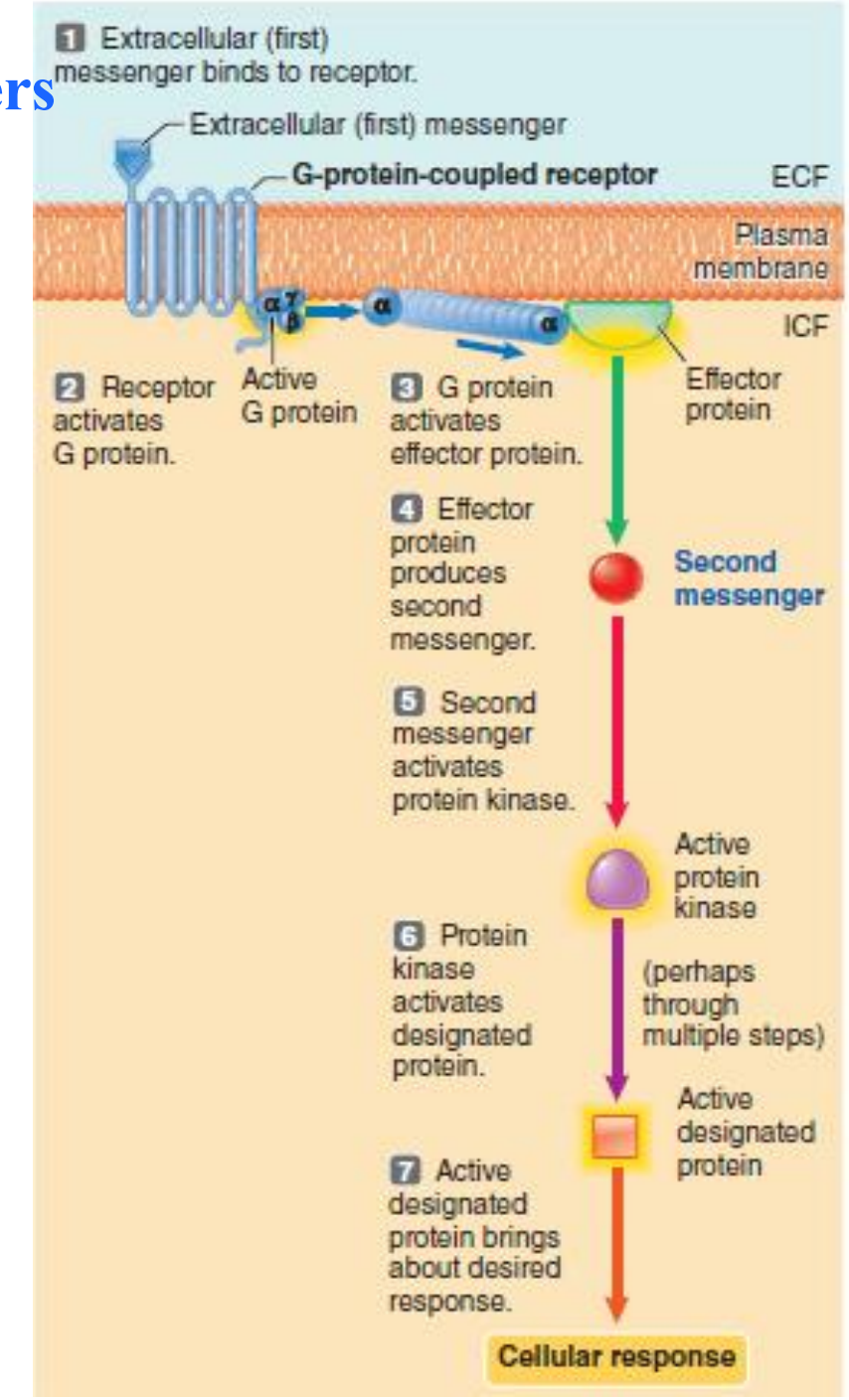
SECOND MESSENGER MECHANISMS FOR MEDIATING INTRACELLULAR HORMONAL FUNCTIONS

- Types of Second Messenger:
- 1. Calcium ions and associated *calmodulin*
- 2. Products of membrane phospholipid breakdown.
- 3. cAMP

- In some cases, a hormone may stimulate more than one second messenger system in the same target tissue

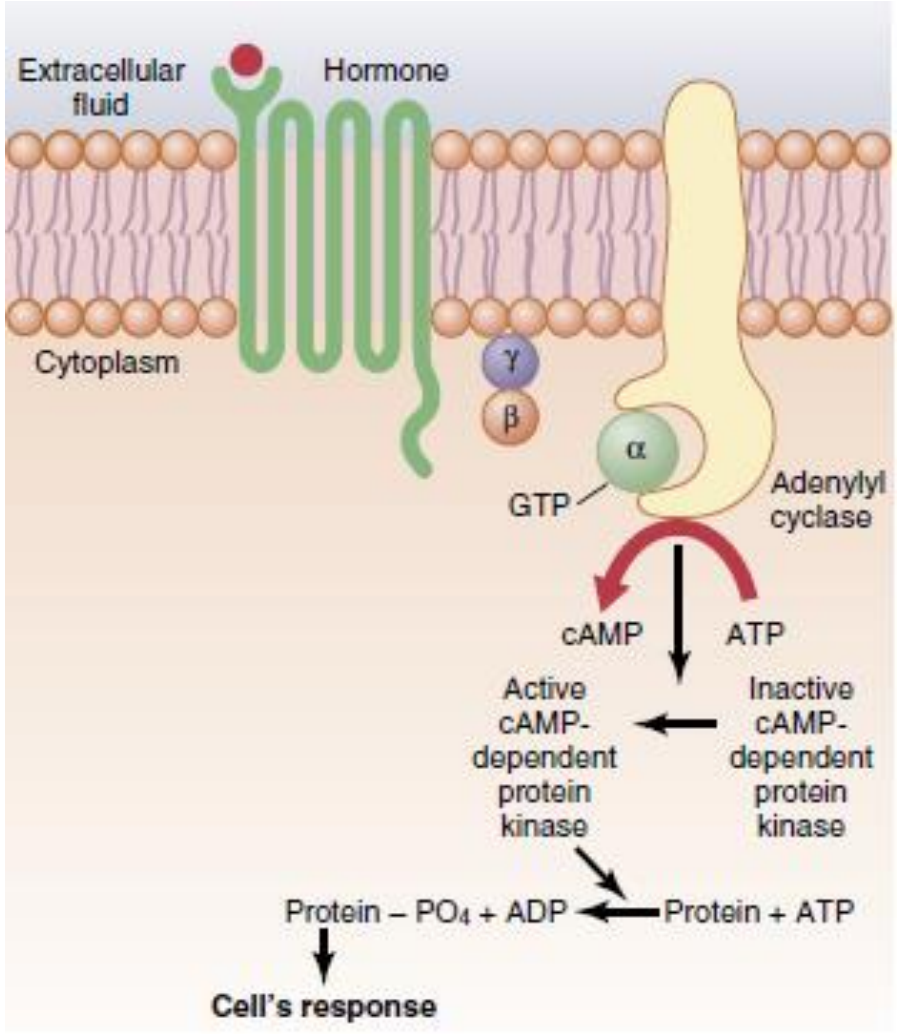
Most water-soluble extracellular chemical messengers activate second-messenger pathways via G-protein-coupled receptors

- Binding of the first messenger to the receptor activates the **G protein**,
- On activation, a portion of the G protein shuttles along the membrane to alter the activity of a nearby membrane protein called the **effector protein**.
- Once altered, the effector protein leads to an increased concentration of an intracellular messenger, known as the **second messenger**.
- The second messenger relays the orders through a cascade of chemical reactions inside the cell that cause a change in the shape and function of designated proteins.



Adenylyl Cyclase–cAMP Second Messenger System

- Stimulation of adenylyl cyclase, by the Gs protein
- Catalyzes the conversion of a small amount of cytoplasmic *Adenosine triphosphate ATP* into cAMP inside the cell.
- Then activates *cAMP-dependent protein kinase.*
- Phosphorylates specific cell proteins, triggering biochemical reactions that ultimately lead to the cell's response to the hormone.



A. cAMP:

❖ Regulation of adenylate cyclase:

Receptors that cause increase in cAMP do so by activating G_s, a stimulatory protein that activates adenylyl cylcase

Adenylyl cyclase is turned off by G_i, an inhibitory protein.

PKA enters the nucleus and phosphorylates CREB (CRE binding protein), which binds to the cAMP response element (CRE), a regulatory DNA sequence associated with specific genes. This results in activation of transcription of those genes.

B. cGMP:

1. produced from GTP by guanylyl cyclase;

2. activates cGMP-dependent kinases or other targets

3. example: G-prot. Coupled rhodopsin photoreceptor in rod cells of retina

Summary of how cAMP activates transcription:

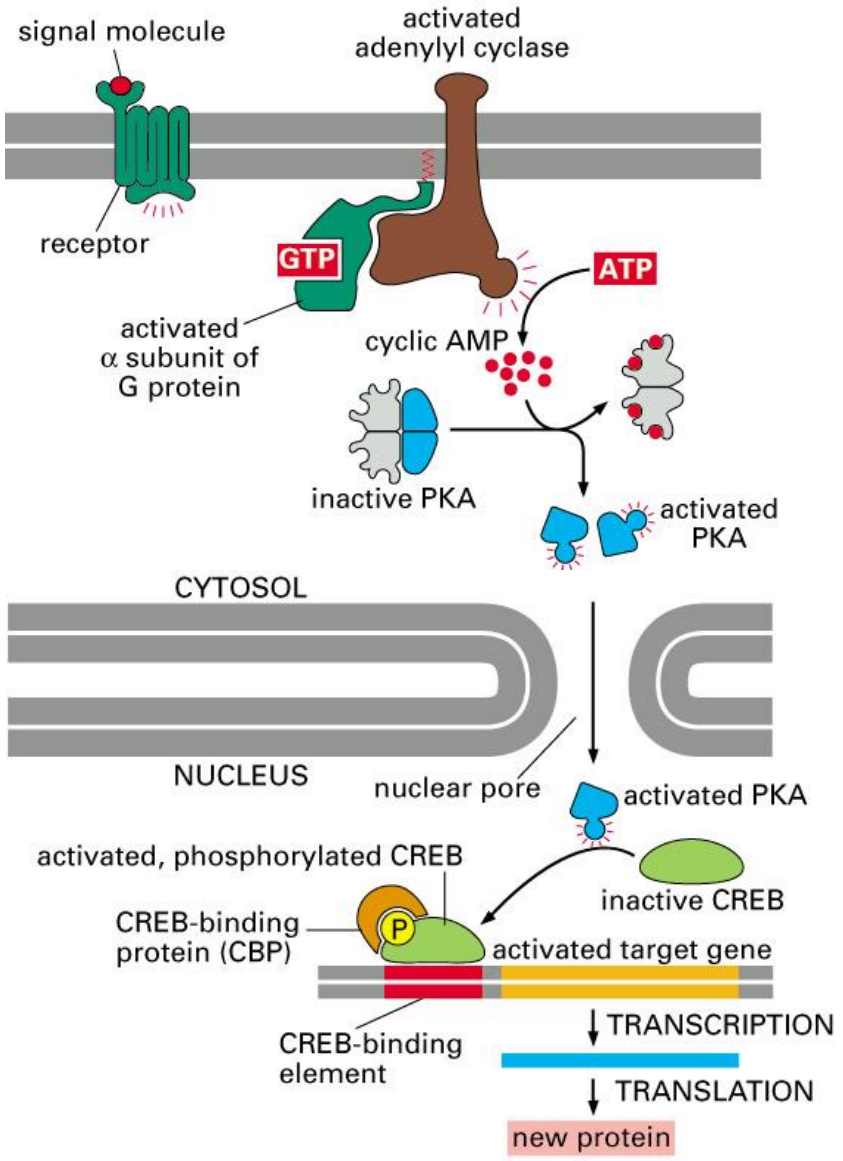


Figure 15–33 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

Table 75-3 Hormones That Use the Adenylyl Cyclase–cAMP Second Messenger System

Adrenocorticotropic hormone (ACTH)
Angiotensin II (epithelial cells)
Calcitonin
Catecholamines (beta receptors)
Corticotropin-releasing hormone (CRH)
Follicle-stimulating hormone (FSH)
Glucagon
Growth hormone–releasing hormone (GHRH)
Human chorionic gonadotropin (hCG)
Luteinizing hormone (LH)
Parathyroid hormone (PTH)
Secretin
Somatostatin
Thyroid-stimulating hormone (TSH)
Vasopressin (V_2 receptor, epithelial cells)

cAMP, Cyclic adenosine monophosphate.

Cell Membrane Phospholipid Second Messenger System

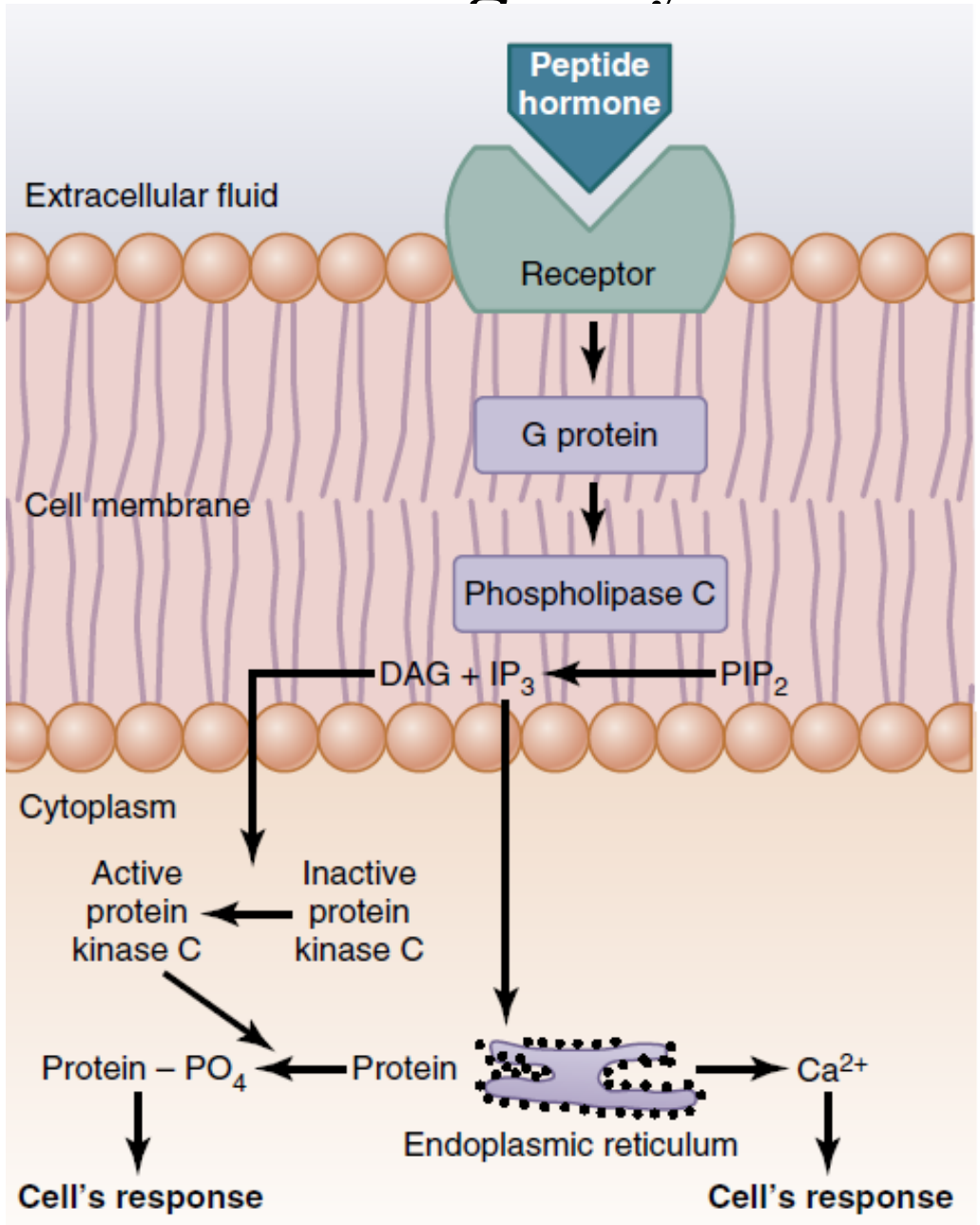
- Some hormones activate transmembrane receptors that activate the enzyme *phospholipase C*
- PLC catalyzes the breakdown of some phospholipids

in the cell membrane, especially *phosphatidylinositol bi-phosphate* (PIP2), into two different second messenger

- products:
- *Inositol triphosphate* (IP3)
- *Diacylglycerol* (DAG).

IP_3 mobilizes calcium ions from mitochondria and the endoplasmic reticulum, Ca ions can lead to different cellular responses

DAG, the other lipid second messenger, activates the enzyme *protein kinase C*, which then phosphorylates a large number of proteins, leading to the cell's response



Overview of PIP2

1. Phosphotidylinositol 4,5 bisphosphate (PIP2) triggers a 2-armed signaling pathway

   a. PIP2 is a minor PL in inner leaflet of PM bilayer that is produced by phosphorylation of phosphatidyl-inositol and is involved in signaling

   b. Ligand binding to certain receptors stimulates PIP2 hydrolysis by phospholipase C (PLC)

   c. This produces diacylglycerol (DAG) and inositol 1,4,5-phosphate (IP3), both of which are 2nd messengers

   d. PIP2 hydrolysis is activated by both GPRs and TKRs via different forms of PLC

   e. PLC-ß is stimulated by G_q proteins while

PLC-γ has SH2 domains that allow binding to activated tyrosine kinases

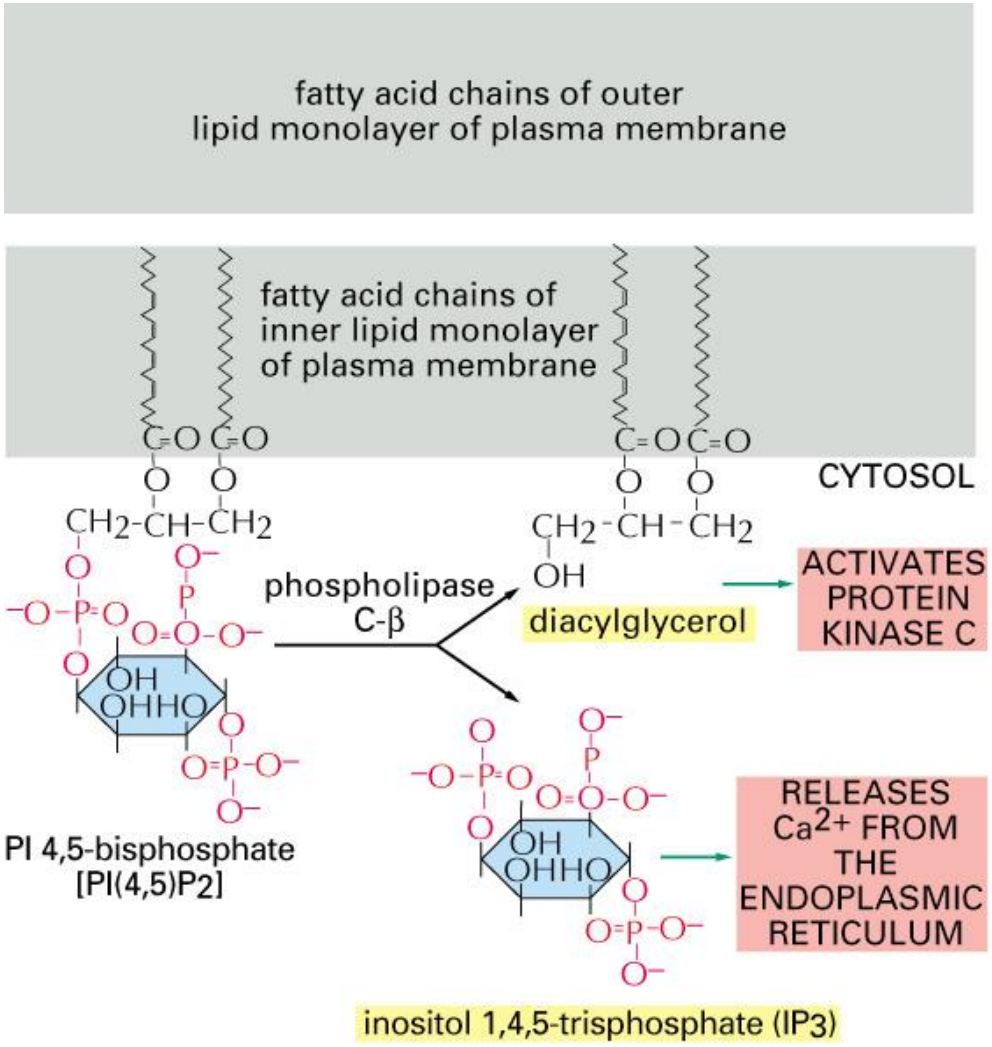


Figure 15–35. Molecular Biology of the Cell, 4th Edition.

PLC- signaling pathway

DAG: Remains associated with the PM
Stimulates the Ca^{+2}-dependent protein kinase C signaling pathway, which activates other targets including the MAP kinase cascade

IP3: Small polar molecule released into cytosol

a. Stimulates Ca^{+2} release from intracellular stores

b. Elevated Ca^{+2} alters activities of target proteins including kinases & phosphatases

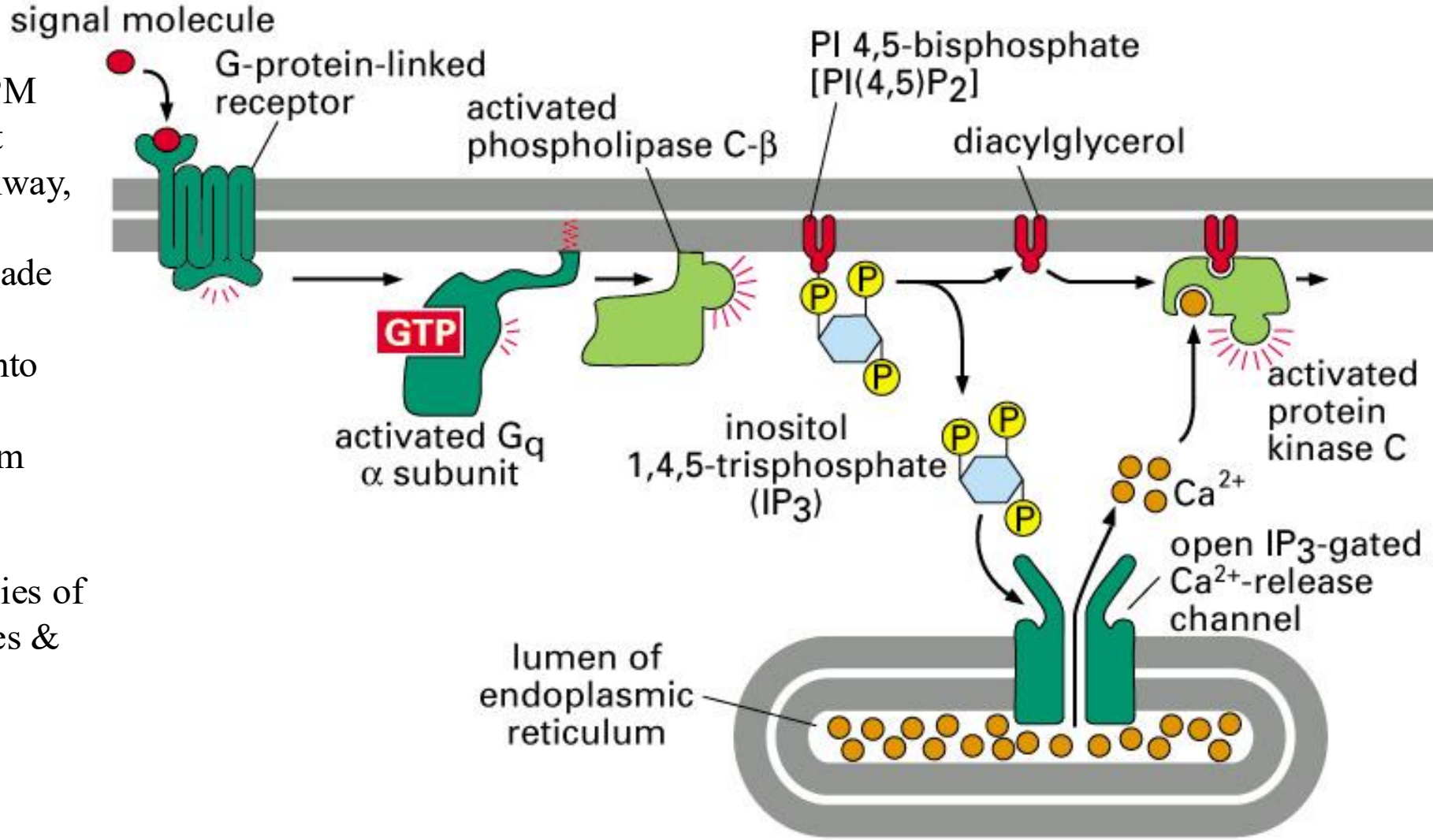


Table 75-4 Hormones That Use the Phospholipase C Second Messenger System

Angiotensin II (vascular smooth muscle)
Catecholamines (α receptors)
Gonadotropin-releasing hormone (GnRH)
Growth hormone–releasing hormone (GHRH)
Parathyroid hormone (PTH)
Oxytocin
Thyrotropin-releasing hormone (TRH)
Vasopressin (V_1 receptor, vascular smooth muscle)

Calcium-Calmodulin Second Messenger System

- Calcium entry may be initiated by:

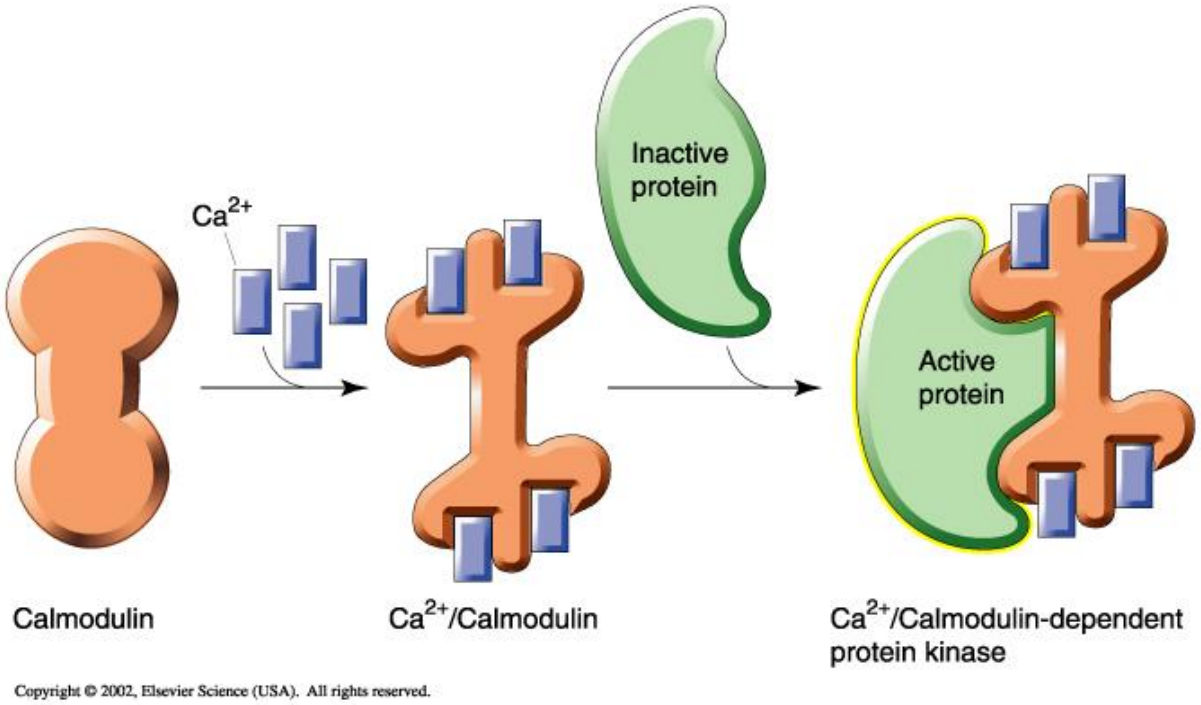
(1)Changes in membrane potential that open calcium channels

(2)Hormone interacting with membrane receptors that open calcium channels.

- Thus, another second messenger system operates in response to entry of calcium into the cells.

Calcium-Calmodulin Second Messenger System

- Calcium ions bind with the protein *calmodulin.*
- This protein has four calcium sites, and when three or four of these sites have become bound with calcium
- The calmodulin changes its shape
- Then initiates multiple effects inside the cell, including activation or inhibition of protein kinases



Calcium-Calmodulin Second Messenger System

Many of the Ca^{2+} -dependent cellular events are triggered by activation of **calmodulin**, an intracellular Ca^{2+} -binding protein

- The Ca^{2+} – calmodulin complex activates **Ca^{2+} –calmodulin dependent protein kinase (CaM kinase)** (or activates another kinase)
- Activation of CaM kinase by the Ca^{2+} – calmodulin complex is similar to activation of PKA by cAMP.

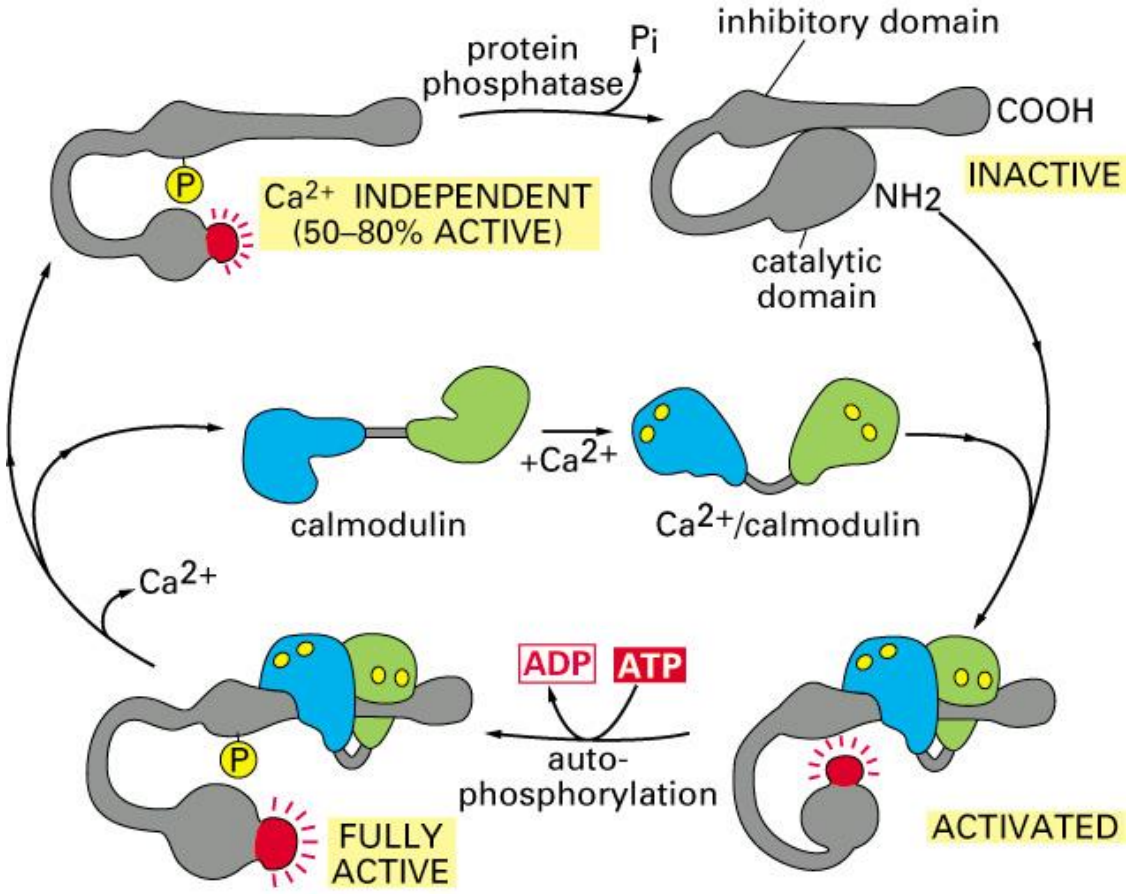
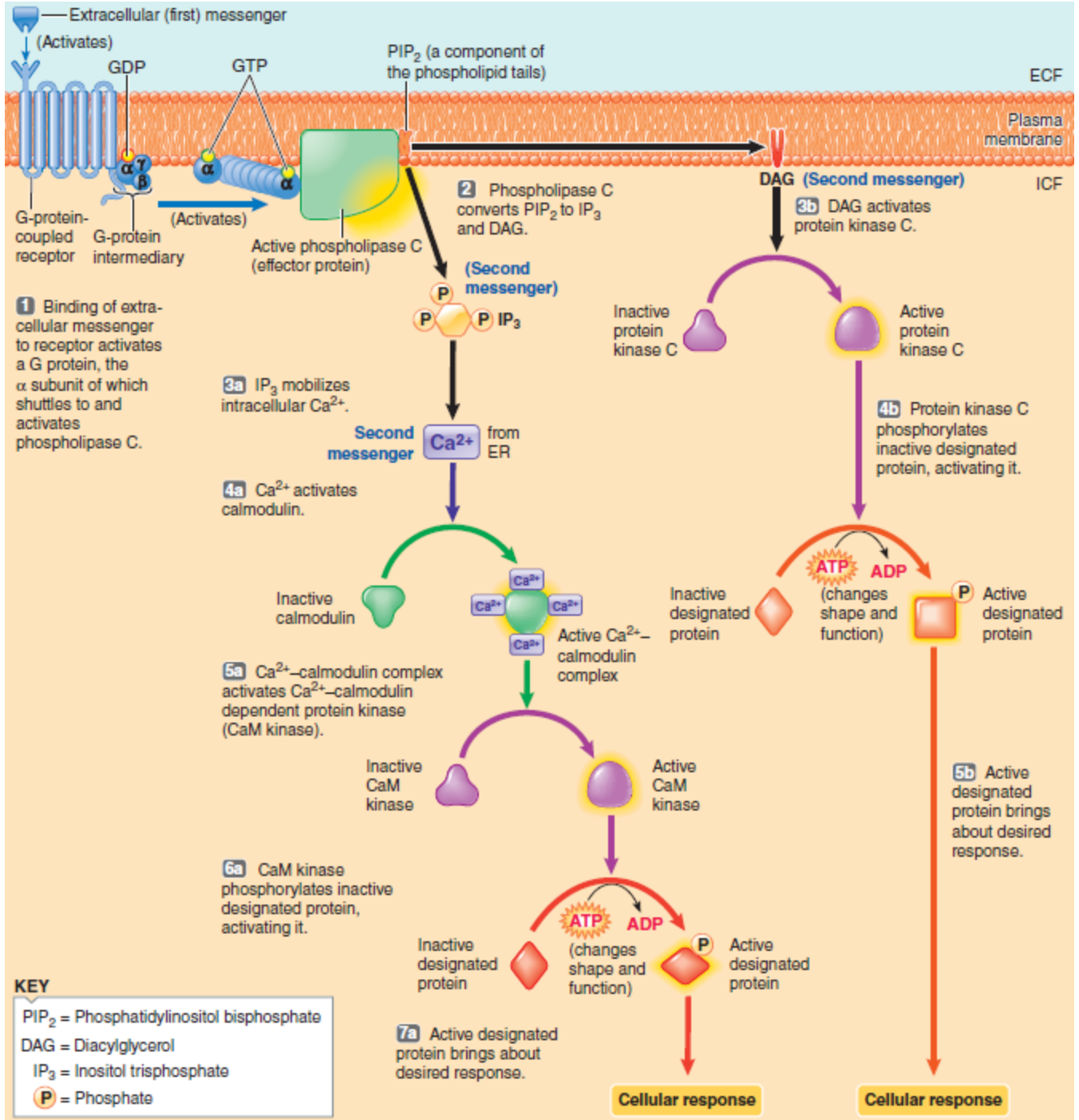
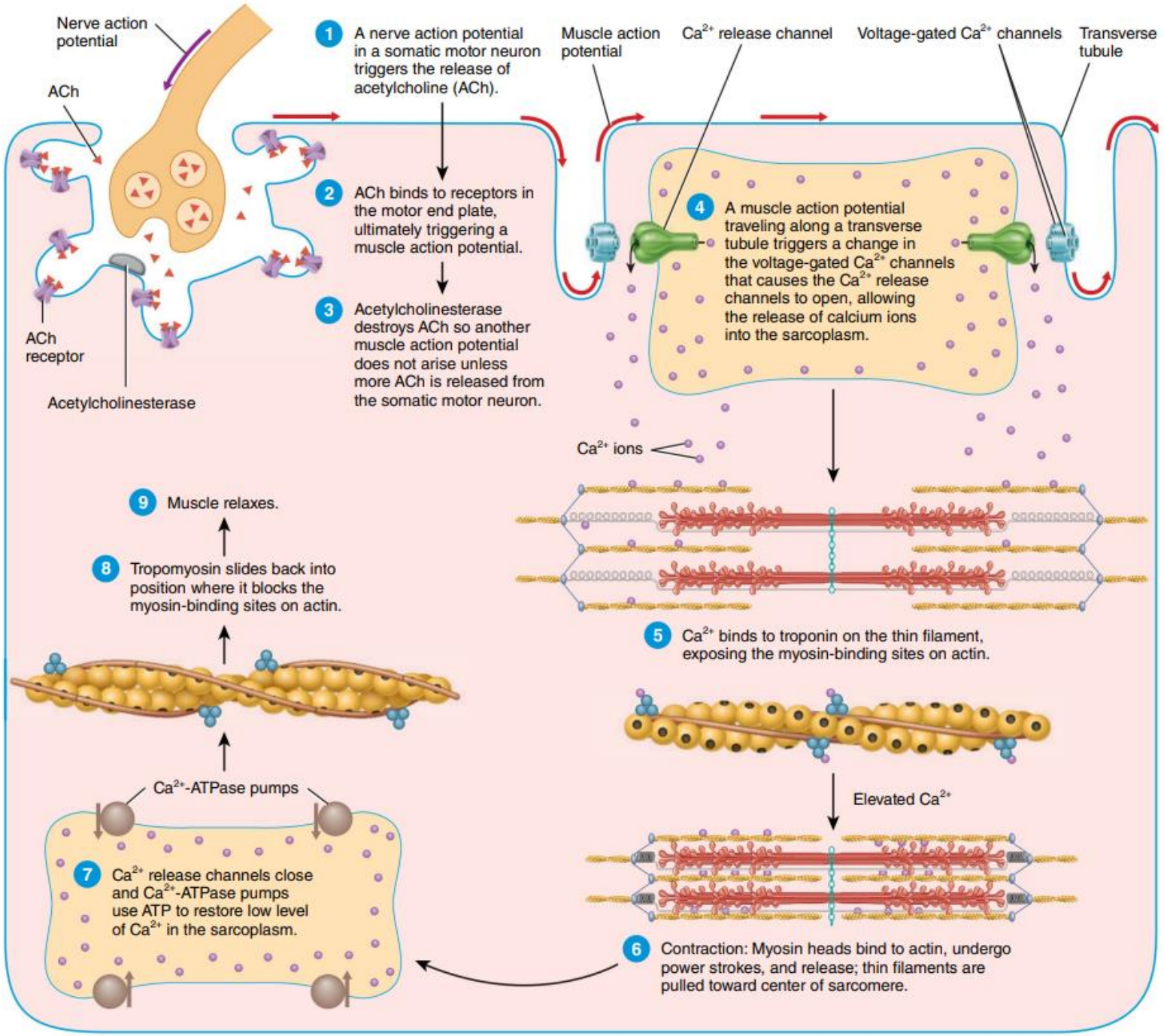


Figure 15–41. Molecular Biology of the Cell, 4th Edition.

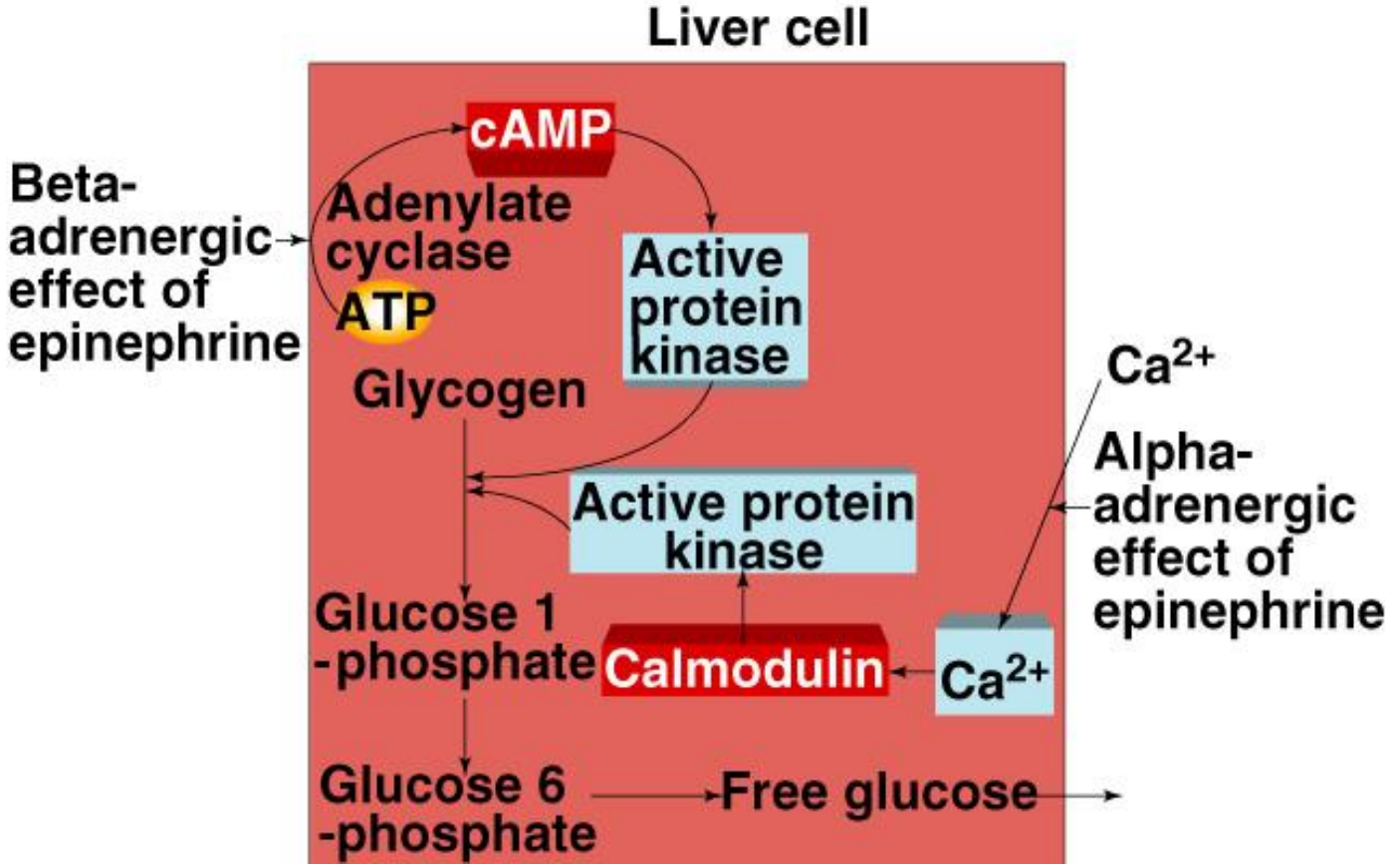

Extracellular (first) messenger
(Activates)
GDP
GTP
PIP2 (a component of the phospholipid tails)
ECF
Plasma membrane
ICF
G-protein-coupled receptor
G-protein intermediary
(Activates)
Active phospholipase C (effector protein)
2 Phospholipase C converts PIP2 to IP3 and DAG.
DAG (Second messenger)
3b DAG activates protein kinase C.
1 Binding of extracellular messenger to receptor activates a G protein, the α subunit of which shuttles to and activates phospholipase C.
(Second messenger)
IP3
Inactive protein kinase C
Active protein kinase C
3a IP3 mobilizes intracellular Ca2+.
Second messenger
Ca2+
from ER
4b Protein kinase C phosphorylates inactive designated protein, activating it.
4a Ca2+ activates calmodulin.
Inactive calmodulin
Active Ca2+–calmodulin complex
ATP
ADP
(changes shape and function)
Inactive designated protein
Active designated protein
5a Ca2+–calmodulin complex activates Ca2+–calmodulin dependent protein kinase (CaM kinase).
Inactive CaM kinase
Active CaM kinase
5b Active designated protein brings about desired response.
6a CaM kinase phosphorylates inactive designated protein, activating it.
ATP
ADP
(changes shape and function)
Inactive designated protein
Active designated protein
7a Active designated protein brings about desired response.
Cellular response
Cellular response
KEY
PIP2 = Phosphatidylinositol bisphosphate
DAG = Diacylglycerol
IP3 = Inositol trisphosphate
P = Phosphate

Other means by which Ca can enter the cells

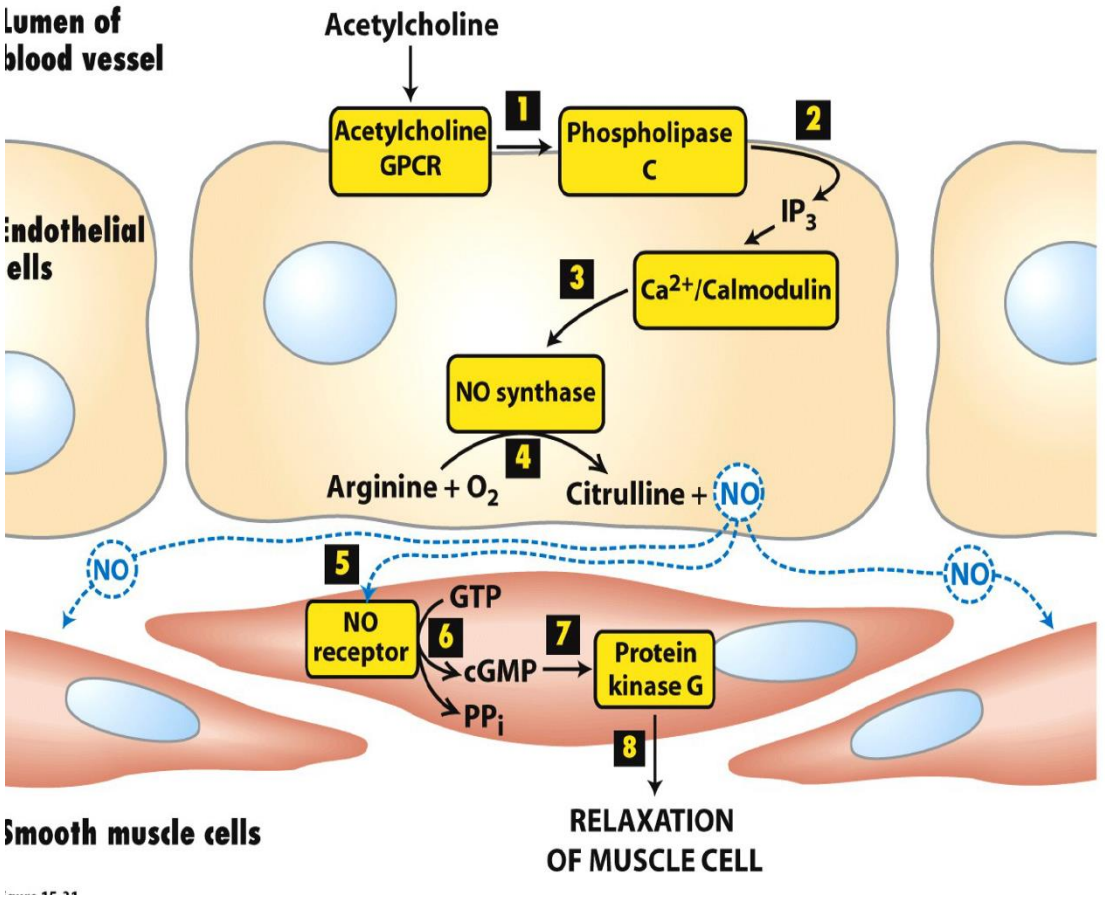


Examples of Second messenger pathways

Epinephrine effect using 2 second messenger systems



NO signaling through Ca-M second messenger system



Amplification of signal

❑In this way, even the slightest amount of hormone acting on the cell surface can initiate a powerful cascading activating force for the entire cell.

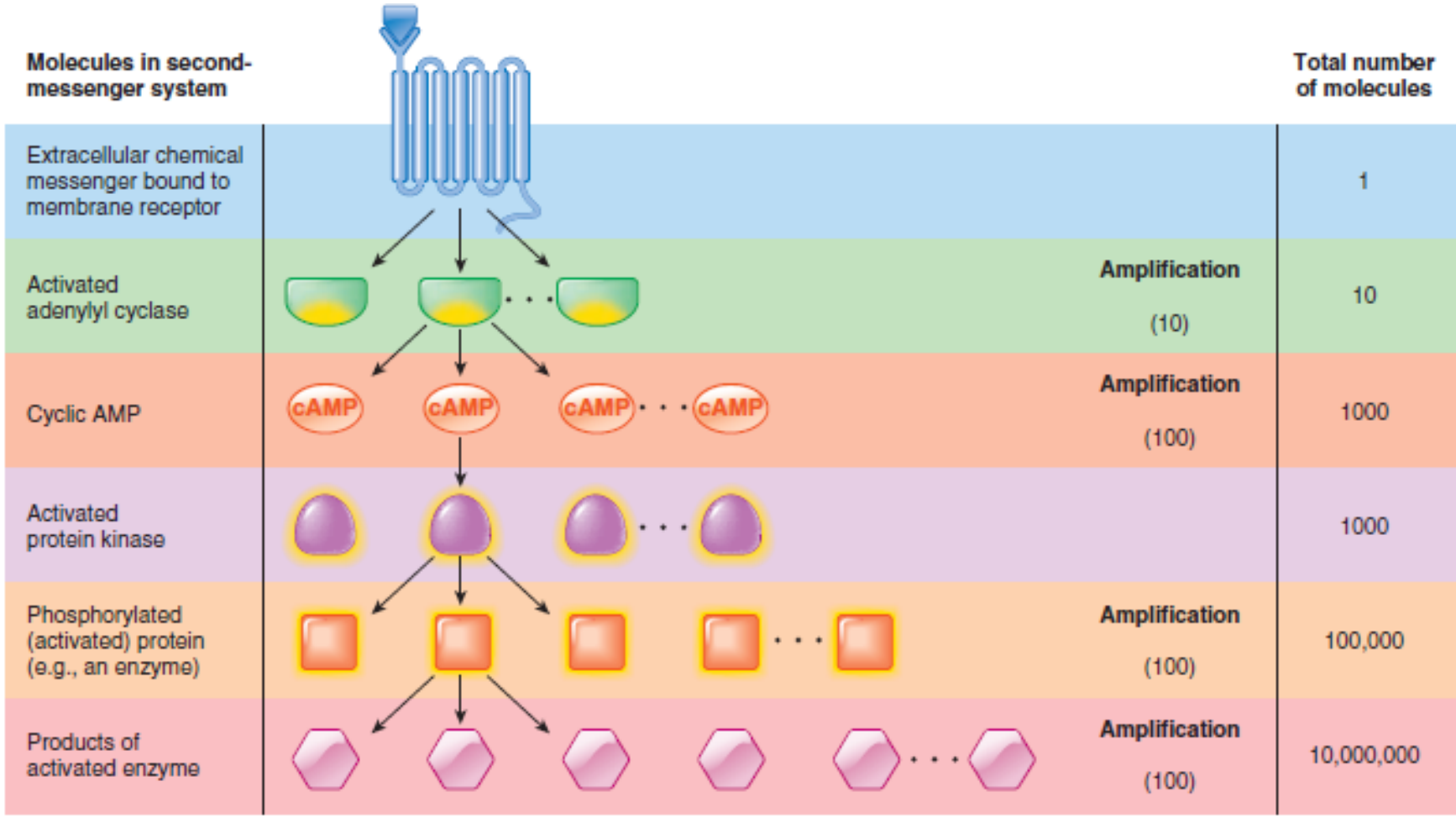


Figure 4-27 Amplification of the initial signal by a second-messenger pathway. Through amplification, very low concentrations of extracellular chemical messengers, such as hormones, can trigger pronounced cellular responses.

5 downstream kinases activated by different signaling cascades

Summary

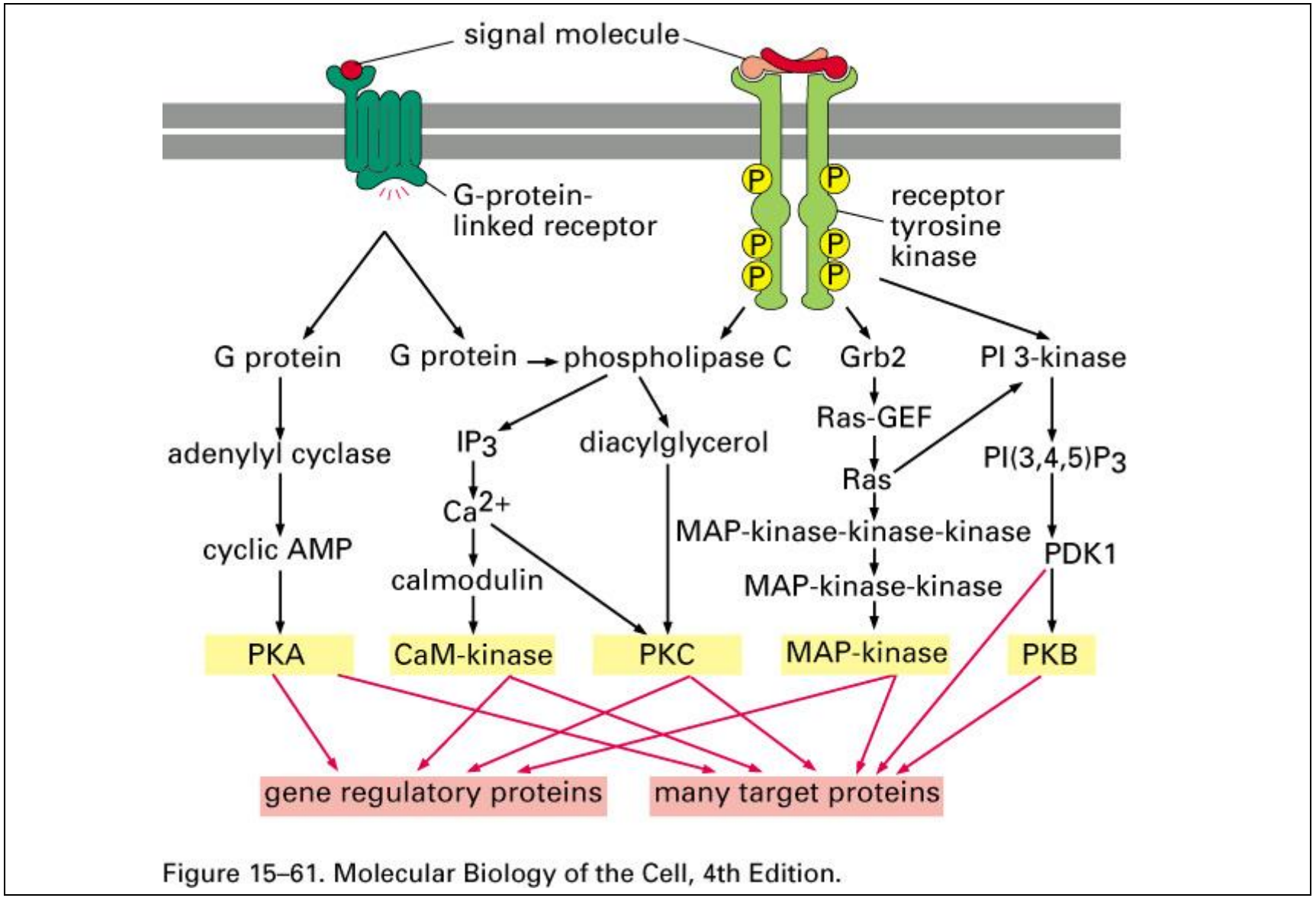


Figure 15–61. Molecular Biology of the Cell, 4th Edition.