Cellular Communication

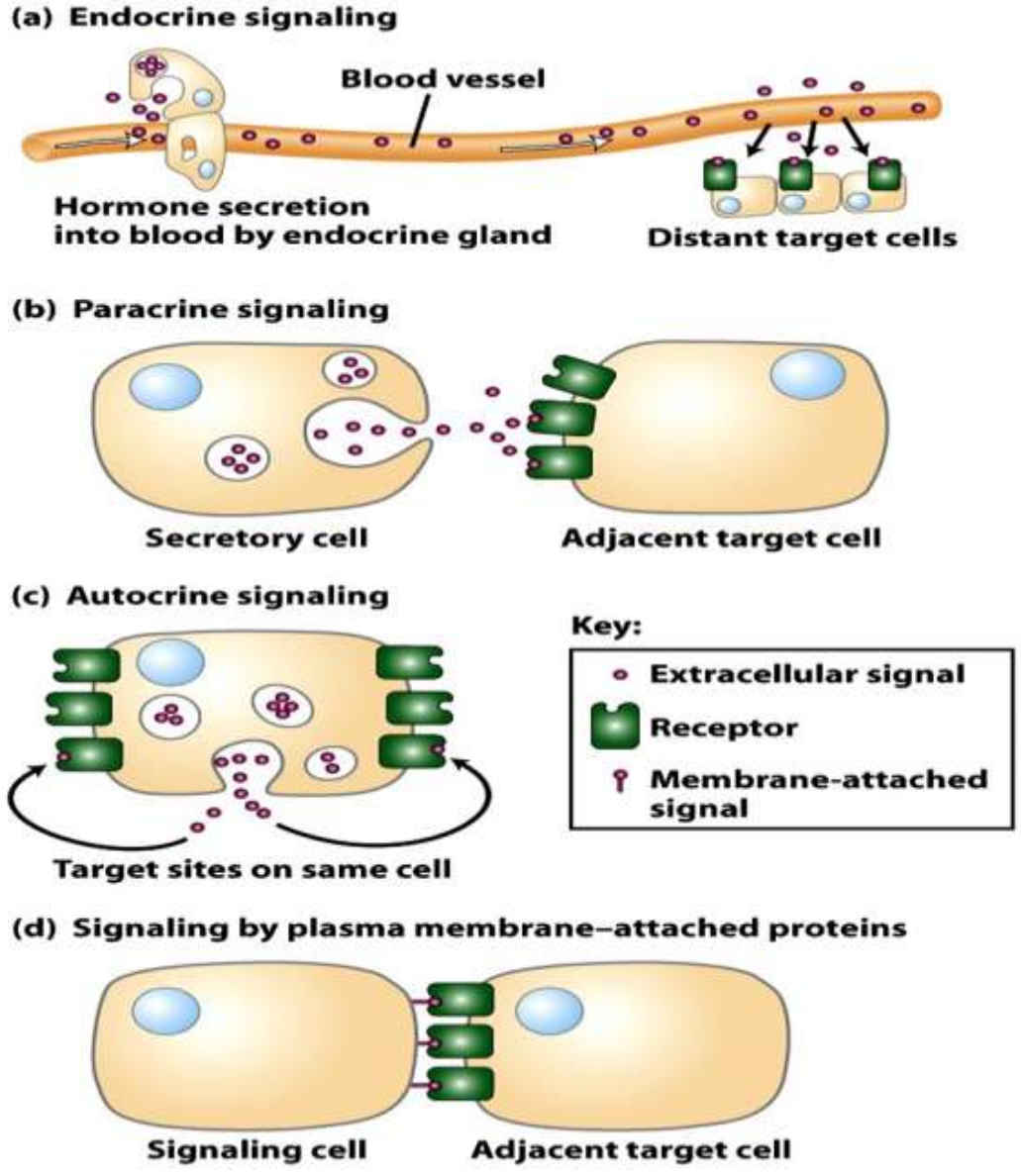
MODES OF COMMUNICATION AND SIGNALING

- Human body has several means of transmitting information between cells. These mechanisms include
- Direct communication between adjacent cells through gap junctions
- Autocrine and paracrine signaling, and
- The release of neurotransmitters and hormones produced by endocrine and nerve cells

Modes of Intercellular Communication

- Cells may communicate with each other directly via gap junctions or chemical messengers.
- With autocrine and paracrine signaling, a chemical messenger diffuses a short distance through ECF and binds to a receptor on the same cell or a nearby cell.
- Nervous signaling involves the rapid transmission of action potentials, often over long distances, and the release of a neurotransmitter at a synapse.
- Endocrine signaling involves the release of a hormone into the bloodstream and the binding of the hormone to specific target cell receptors

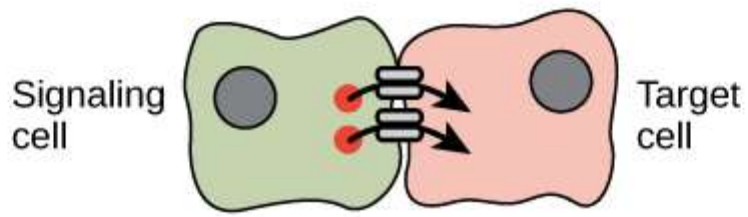
- Neuroendocrine signaling involves the release of a hormone from a nerve cell and thetransport of the hormone by the blood to a distant target cell.

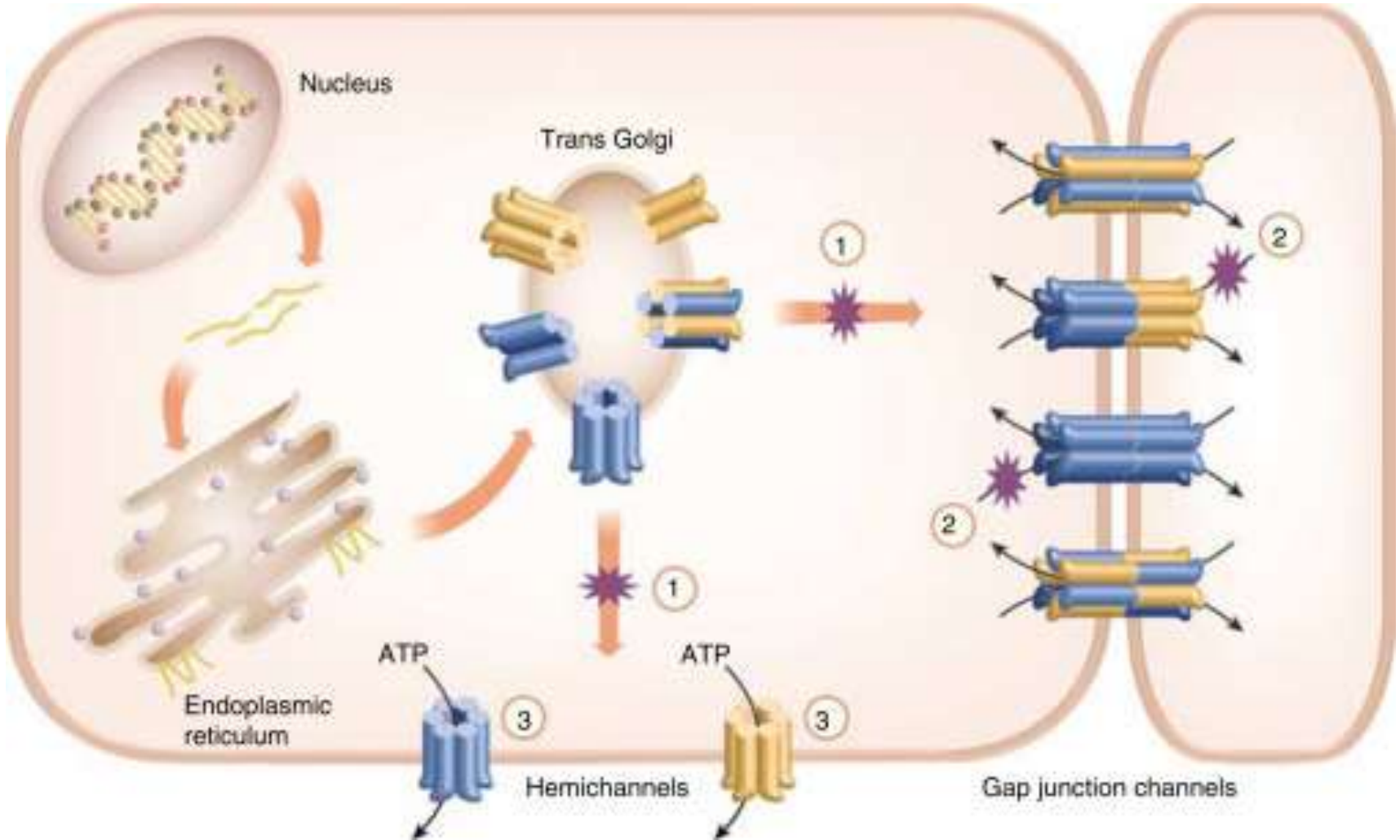

(a) Endocrine signaling
Blood vessel
Hormone secretion into blood by endocrine gland
Distant target cells
(b) Paracrine signaling
Secretory cell
Adjacent target cell
(c) Autocrine signaling
Target sites on same cell
Key:
Extracellular signal
Receptor
Membrane-attached signal
(d) Signaling by plasma membrane–attached proteins
Signaling cell
Adjacent target cell

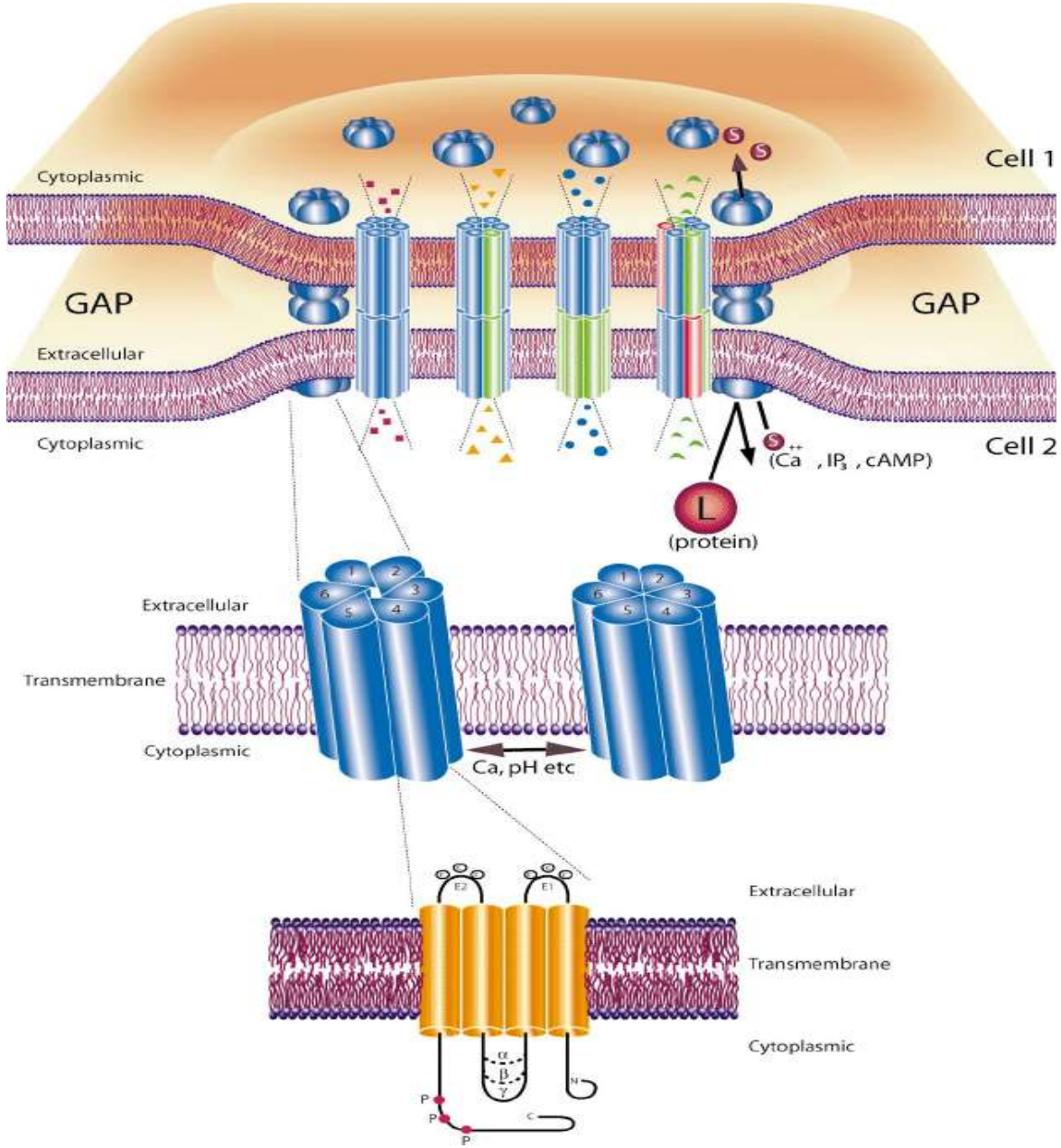
Gap Junctions

- They provide a pathway for direct communication between adjacent cells
- Gap junctions are specialized protein channels made of the protein connexin
- 6 connexins form a half-channel called a connexon.
- 2 connexons join end to end to form an intercellular channel between adjacent cells.
- Gap junctions allow the flow of ions (hence, electrical current) and small molecules between neighbouring cells

Signaling across gap junctions	A cell targets a cell connected by gap junctions.




Nucleus
Trans Golgi
1
2
2
1
ATP
ATP
3
3
Endoplasmic
reticulum
Hemichannels
Gap junction channels


Cell 1
Cytoplasmic
GAP
GAP
Extracellular
Cytoplasmic
Cell 2
(Ca++, IP3, cAMP)
L
(protein)
Extracellular
Transmembrane
Cytoplasmic
Ca, pH etc
Extracellular
Transmembrane
Cytoplasmic

- They are important in the transmission of electrical signals between neighboring cardiac muscle cells, smooth muscle cells, and some nerve cells.
- May also functionally couple adjacent epithelial cells.
- Are thought to play a role in the control of cell growth and differentiation by allowing adjacent cells to share a common intracellular environment.
- Often when a cell is injured, gap junctions close, isolating a damaged cell from its neighbors.
- This isolation process may result from a rise in calcium and a fall in pH in the cytosol of the damaged cell

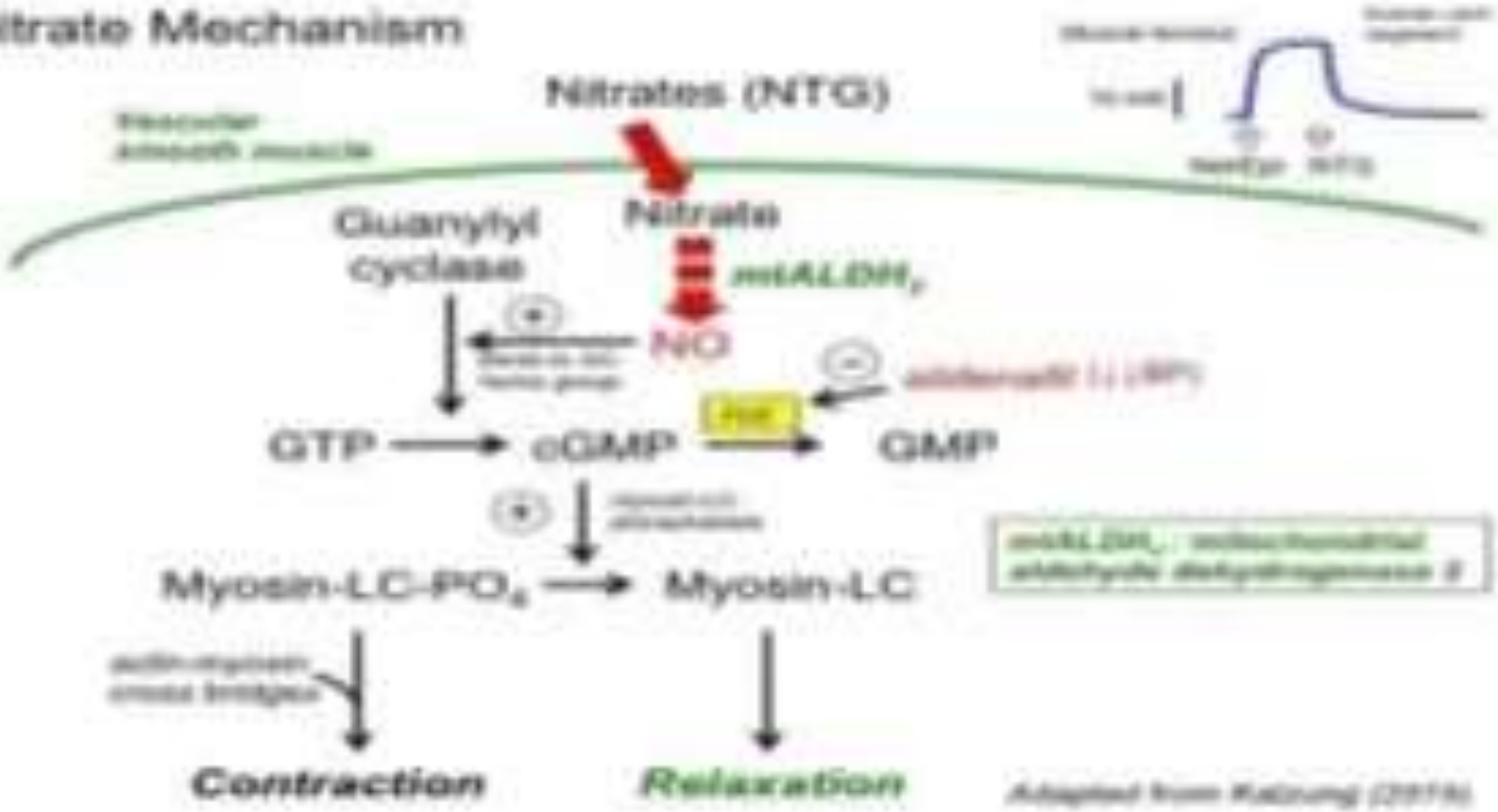
Paracrine and Autocrine Signalling

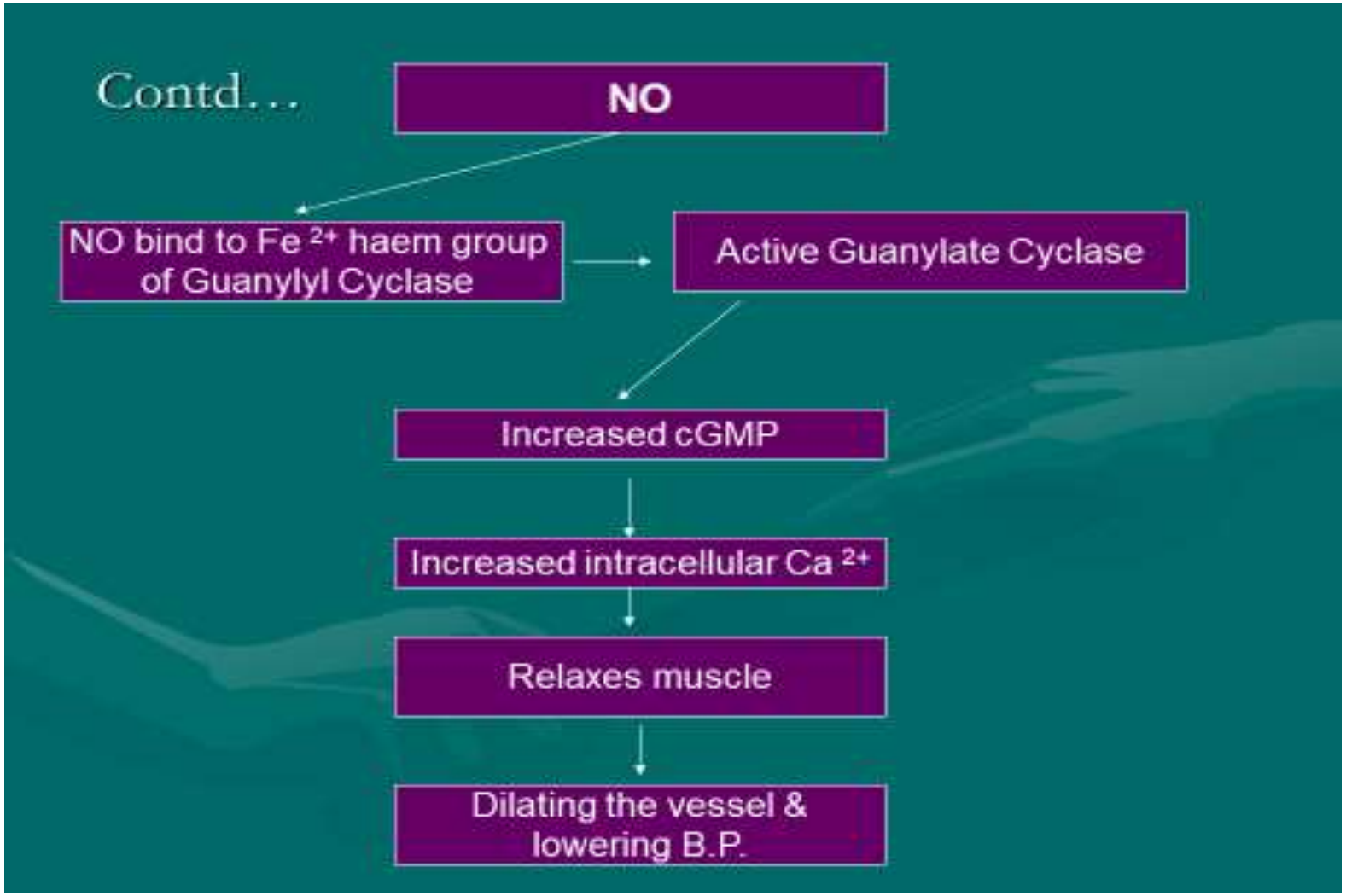
- Cells may signal to each other via the local release of chemical substances.
- This means of communication, present in primitive living forms, does not depend on a CVS
- In paracrine signaling, a chemical is liberated from a cell, diffuses a short distance through interstitial fluid, and acts on nearby cells.
- It affects only the immediate environment and bind with high specificity to cell receptors.
- They are also rapidly destroyed by extracellular enzymes or bound to extracellular matrix
- This prevents their widespread diffusion.

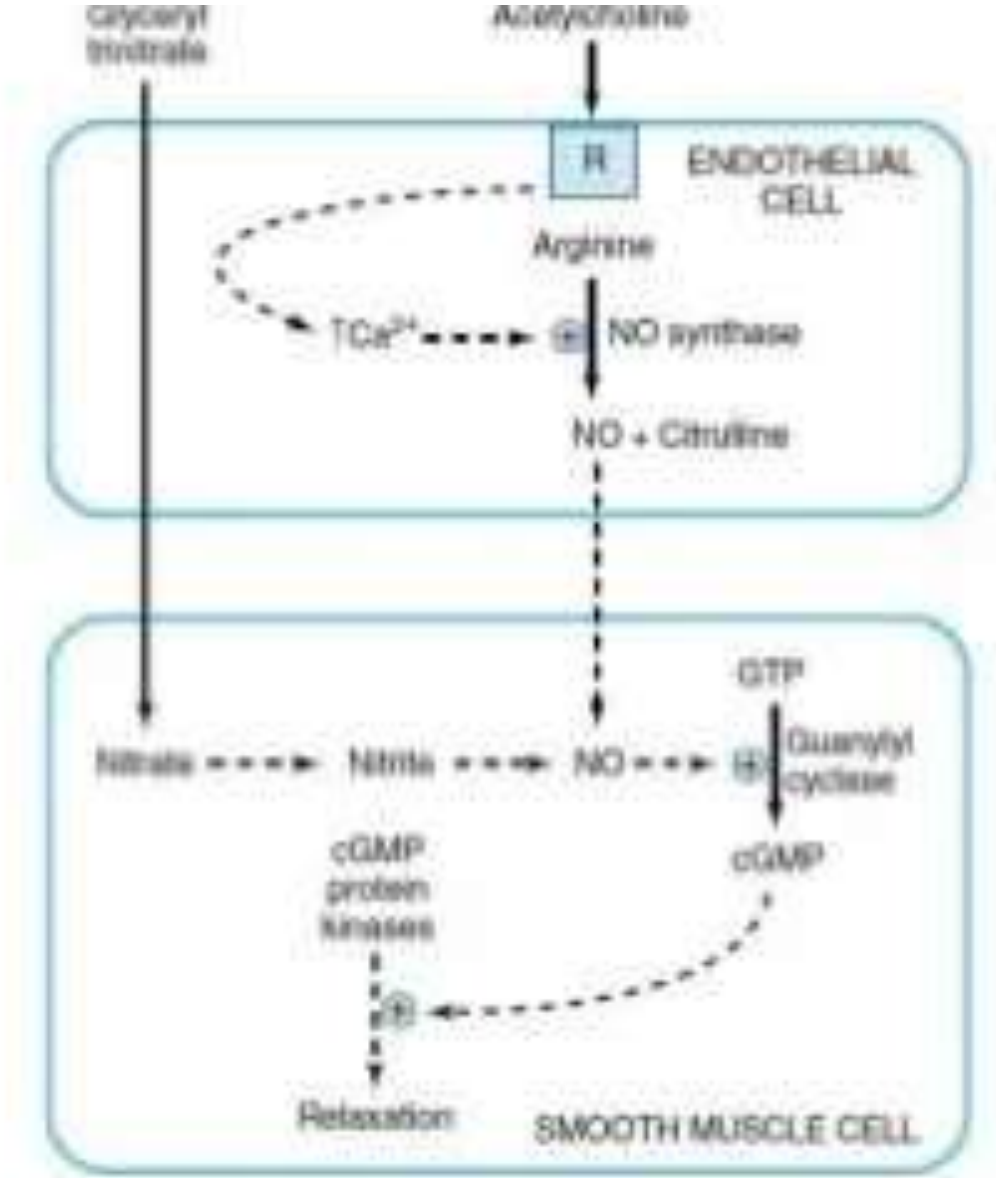
- An example of a paracrine signal is Nitric oxide (NO), also known as endothelium-derived relaxing factor (EDRF)
- NO has major roles in mediating vascular (blood vessel) smooth muscle tone, facilitating CNS neurotransmission activities, and modulating immune responses
- The production of NO results from the activation of nitric oxide synthase (NOS), which deaminates arginine to citrulline.
- NO is produced by endothelial cells (cells lining the inner surface of blood vessels)
- Once produced, it regulates vascular tone by diffusing from the endothelial cell to the underlying vascular smooth muscle cell

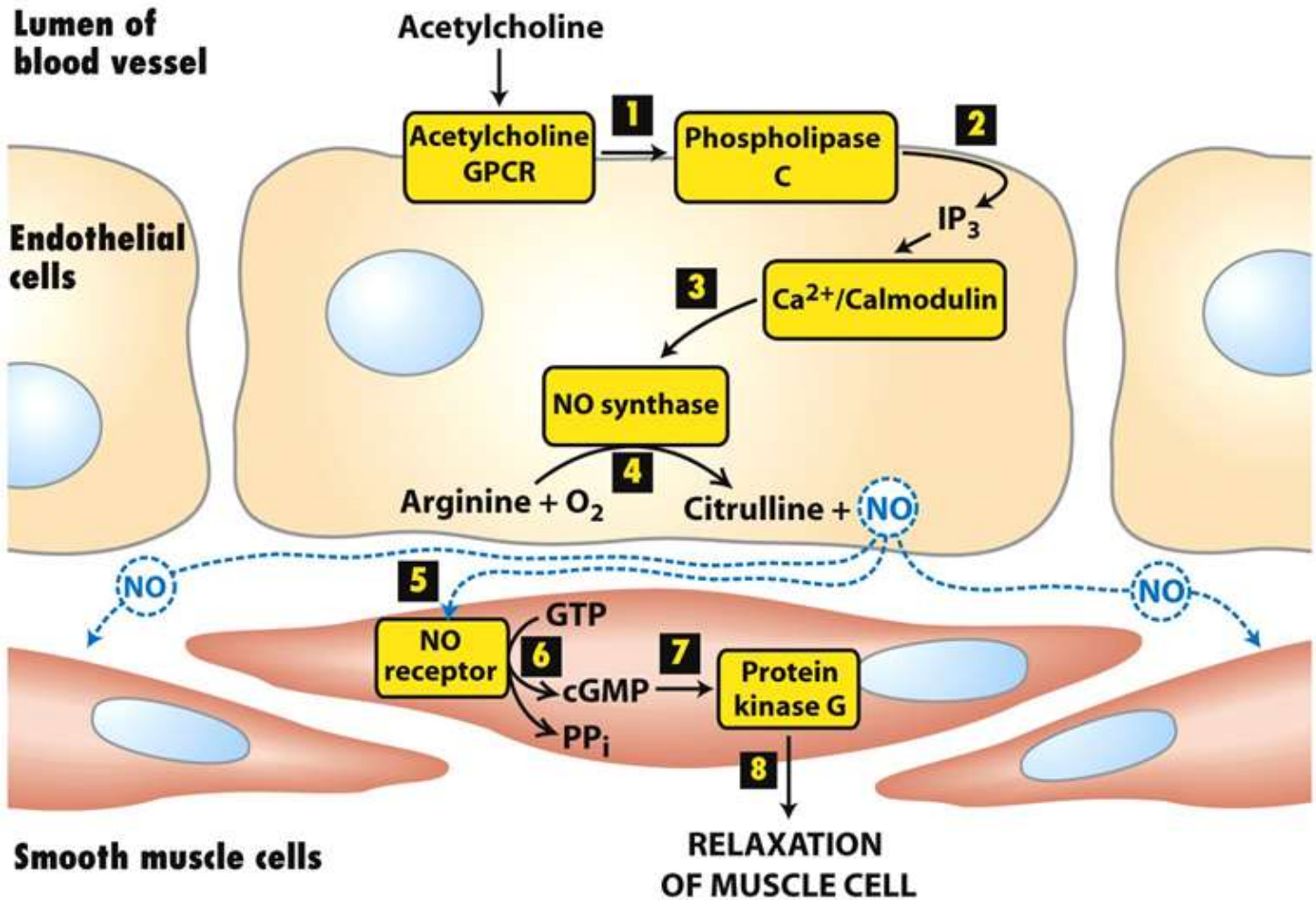
- Here, it activates its effector target, an enzyme in cytoplasm known as guanylyl cyclase.
- The activation of cytoplasmic guanylyl cyclase results in increased intracellular cyclic guanosine monophosphate (cGMP) levels
- cGMP causes activation of cGMP dependent protein kinase.
- This enzyme phosphorylates potential target substrates, such as calcium pumps in the sarcoplasmic reticulum leading to reduced cytoplasmic levels of calcium.
- In turn, this deactivates the contractile machinery in the vascular smooth muscle cell with resultant relaxation or a decrease of tone

Nitrate Mechanism




Contd…
NO
NO bind to Fe 2+ haem group of Guanylyl Cyclase
Active Guanylate Cyclase
Increased cGMP
Increased intracellular Ca 2+
Relaxes muscle
Dilating the vessel & lowering B.P.

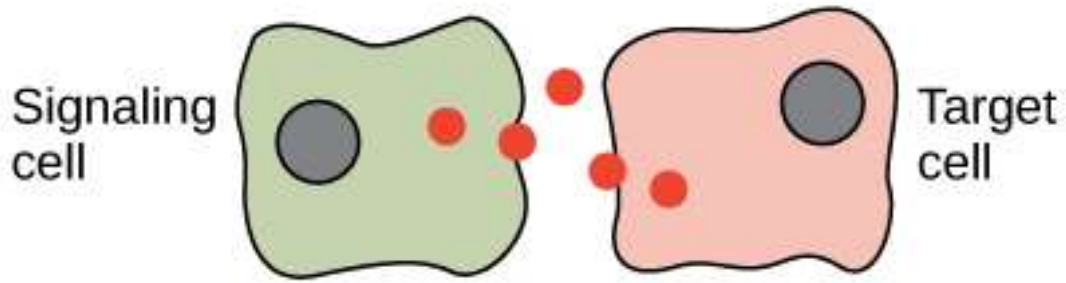

Glyceryl trinitrate
Acetylcholine
R
ENDOTHELIAL CELL
Arginine
↑Ca^{2+}
NO synthase
NO + Citrulline
GTP
Nitrate
Nitrite
NO
Guanylyl cyclase
cGMP
cGMP protein kinases
Relaxation
SMOOTH MUSCLE CELL


Lumen of blood vessel
Acetylcholine
Acetylcholine GPCR
1
Phospholipase C
2
IP3
Endothelial cells
3
Ca2+/Calmodulin
NO synthase
4
Arginine + O2
Citrulline + NO
NO
5
NO receptor
GTP
6
cGMP
7
Protein kinase G
PPi
8
RELAXATION OF MUSCLE CELL
Smooth muscle cells

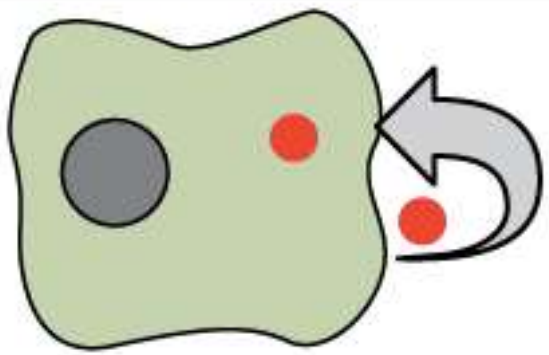
Autocrine signalling

- The cell releases a chemical into the interstitial fluid that affects its own activity by binding to a receptor on its own surface
- Eicosanoids (e.g., prostaglandins), are examples of signaling molecules that often act in an autocrine manner.
- These molecules act as local hormones to influence a variety of physiological processes, such as uterine smooth muscle contraction during pregnancy.

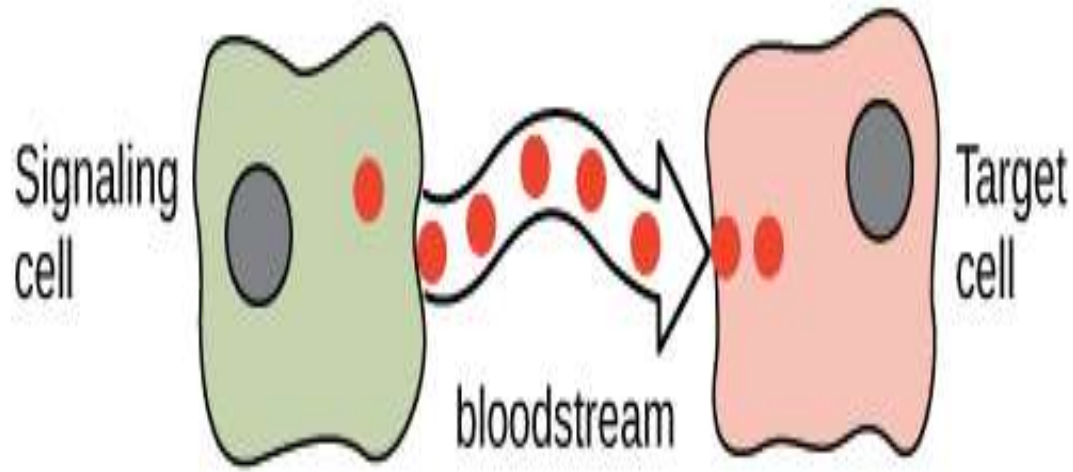
Paracrine	A cell targets a nearby cell.



Autocrine	A cell targets itself.

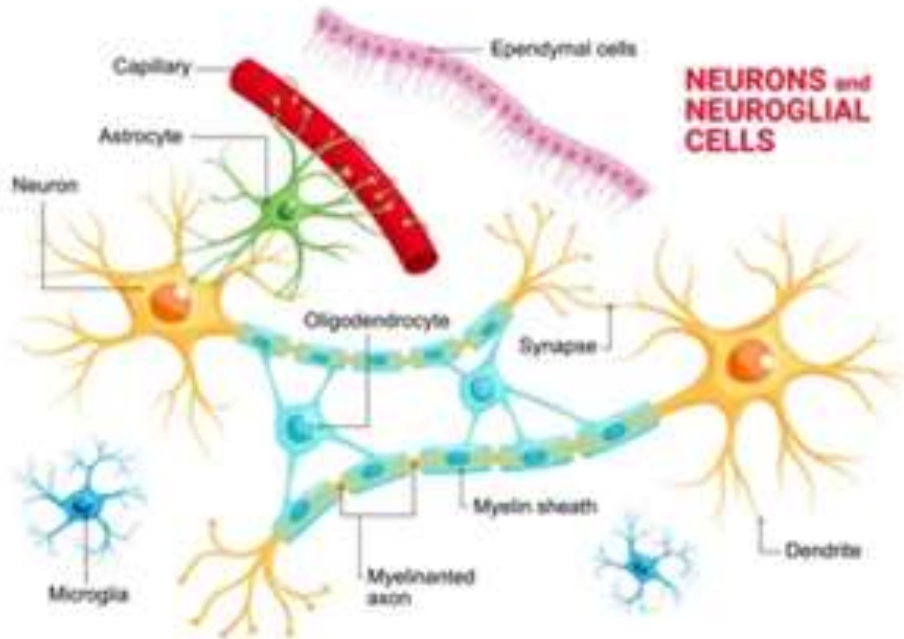


Endocrine	A cell targets a distant cell through the bloodstream.



The Nervous System

- For rapid communication between body parts, with conduction times measured in milliseconds.
- This system is also organized for discrete activities; it has an enormous number of “private lines” for sending messages from one distinct locus to another.
- Conduction of information along nerves occurs via action potentials, and signal transmission between nerves or between nerves and effector structures takes place at a synapse.
- Synaptic transmission is mediated via release of specific chemicals known as neurotransmitters from the nerve terminals
- Innervated cells have specialized protein molecules (receptors) in their cell membranes that selectively bind neurotransmitters.
- This will be discussed later under excitable tissue



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Endocrine signalling

- Endocrine system produces hormones in response to a variety of stimuli.
- Cf Nervous system, responses to hormones are much slower (seconds to hours) in onset, and the effects often last longer.
- Hormones are carried to all parts of the body by the bloodstream
- A particular cell can respond to a hormone only if it possesses the specific receptor (“receiver”) for the hormone.
- Hormone effects may be discrete. Eg: Vasopressin (ADH) increases the water permeability of kidney collecting duct cells but does not change the water permeability of other cells.

- Cells that are not traditional endocrine cells produce a special category of chemical messengers called tissue growth factors.
- Growth factors are proteins that influence cell division, differentiation, and cell survival.
- May exert effects in an autocrine, paracrine, or endocrine fashion.
- Many growth factors have been identified, and probably many more will be recognized in years to come.
- Nerve growth factor enhances nerve cell development and stimulates the growth of axons

- Epidermal growth factor stimulates the growth of epithelial cells in the skin and other organs.
- Platelet-derived growth factor stimulates the proliferation of vascular smooth muscle and endothelial cells.
- Insulin-like growth factors stimulate the proliferation of a wide variety of cells and mediate many of the effects of growth hormone.
- Growth factors appear to be important in the development of multicellular organisms and in the regeneration and repair of damaged tissues.

- Nervous and Endocrine Control Systems Overlap
- The distinction between them not always clear.
- 1st, nervous system exerts important controls over endocrine gland function.
- Eg: The hypothalamus controls the secretion of hormones from the pituitary gland.
- 2nd, specialized nerve cells, called neuroendocrine cells, secrete hormones.
- Eg: The hypothalamic neurons, secrete releasing factors that control secretion by the anterior pituitary gland, and the hypothalamic neurons, secrete ADH and oxytocin hormones into the circulation.

Molecular basis of Cell Signalling

- Cells communicate with one another by many complex mechanisms.
- Even unicellular organisms, such as yeast cells, utilize small peptides called pheromones to coordinate mating events that eventually result in haploid cells with new assortments of genes.
- The signaling pathways must be tightly regulated to maintain cellular homeostasis.
- Dysregulation of these signaling pathways can transform normal cellular growth into uncontrolled cellular proliferation or cancer

- Signaling systems consist of receptors that reside either in the plasma membrane or within cells
- Are activated by a variety of extracellular signals or 1st messengers, including peptides, protein hormones and growth factors, steroids, ions, metabolic products, gases, and various chemical or physical agents (e.g., light).
- Signaling systems also include transducers and effectors that are involved in conversion of the signal into a physiological response.
- The pathway may include additional intracelular messengers, called second messengers.

- Examples of 2nd messengers are
- Cyclic nucleotides such as cyclic adenosine monophosphate (cAMP) and cyclic guanosine monophosphate (cGMP),
- Inositol 1,4,5-trisphosphate (IP3) and diacylglycerol (DAG), and
- Calcium.

- A general outline for a signaling system is as follows:
- The signaling cascade is initiated by binding of a hormone (1st messenger) to its appropriate ligand-binding site on the outer surface domain of its cognate membrane receptor.
- This results in activation of the receptor; the receptor may adopt a new conformation (change in the shape of a macromolecule), form aggregates (multimerize), or become phosphorylated.
- This change usually results in an association of adapter signaling molecules that transduce and amplify the signal through the cell by activating specific effector molecules and generating a 2nd messenger
- The outcome of the signal transduction cascade is a physiological response, such as secretion, movement, growth, division, or death.

Signal transduction by the cell membrane

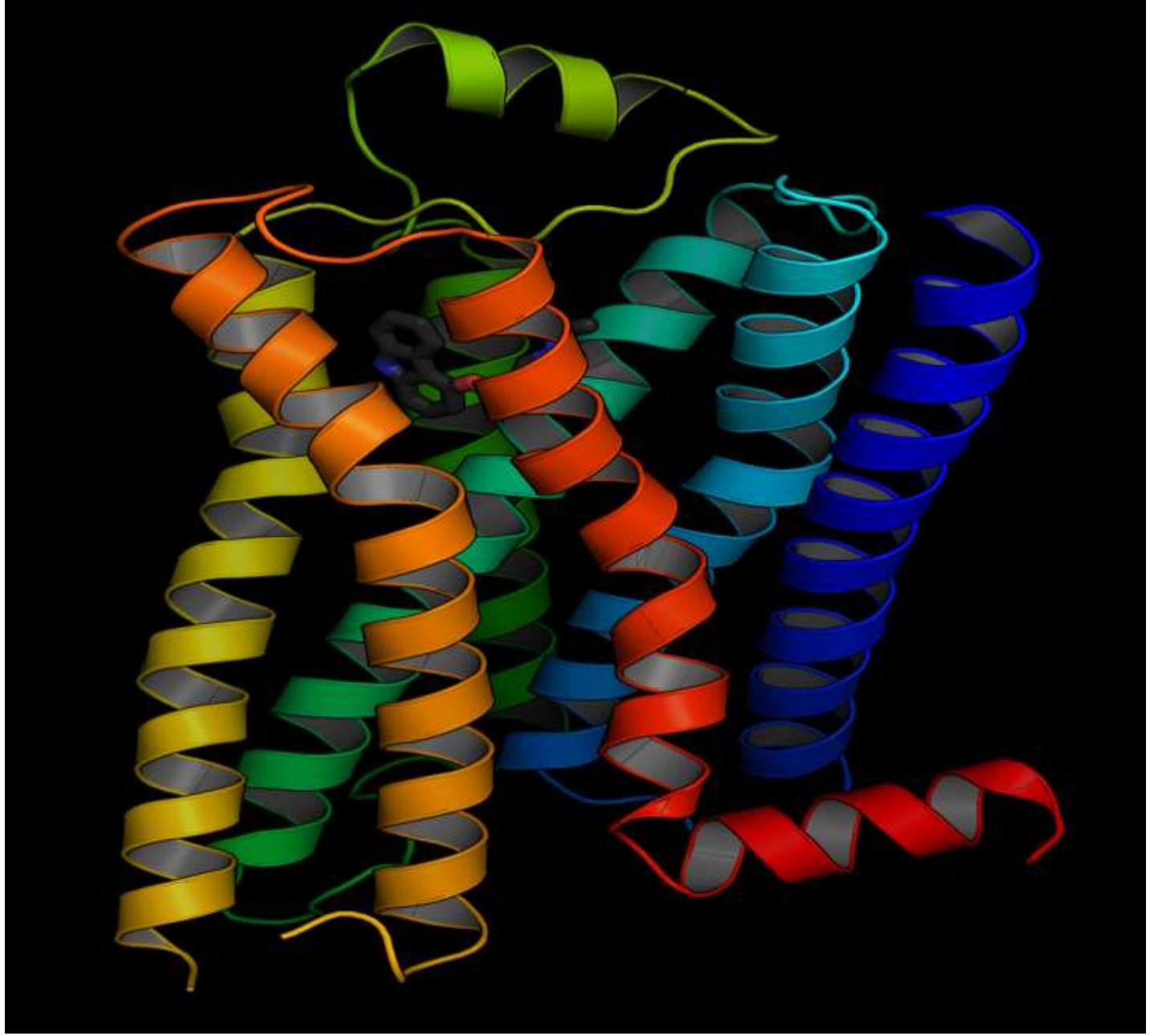
- The molecules involved in signaling are ligands or 1st messengers.
- Many bind directly to receptor proteins cell membrane while others cross the membrane and interact with intercellular receptors which are either in the cytoplasm or nucleus.
- Thus, cellular receptors divided into either cell-surface receptors or intracellular receptors
- 3 classes of cell-surface receptors:
- G-protein-coupled receptors,
- ion channel linked receptors, and
- enzyme-linked receptors.
- Intracellular receptors include steroid and thyroid hormone receptors

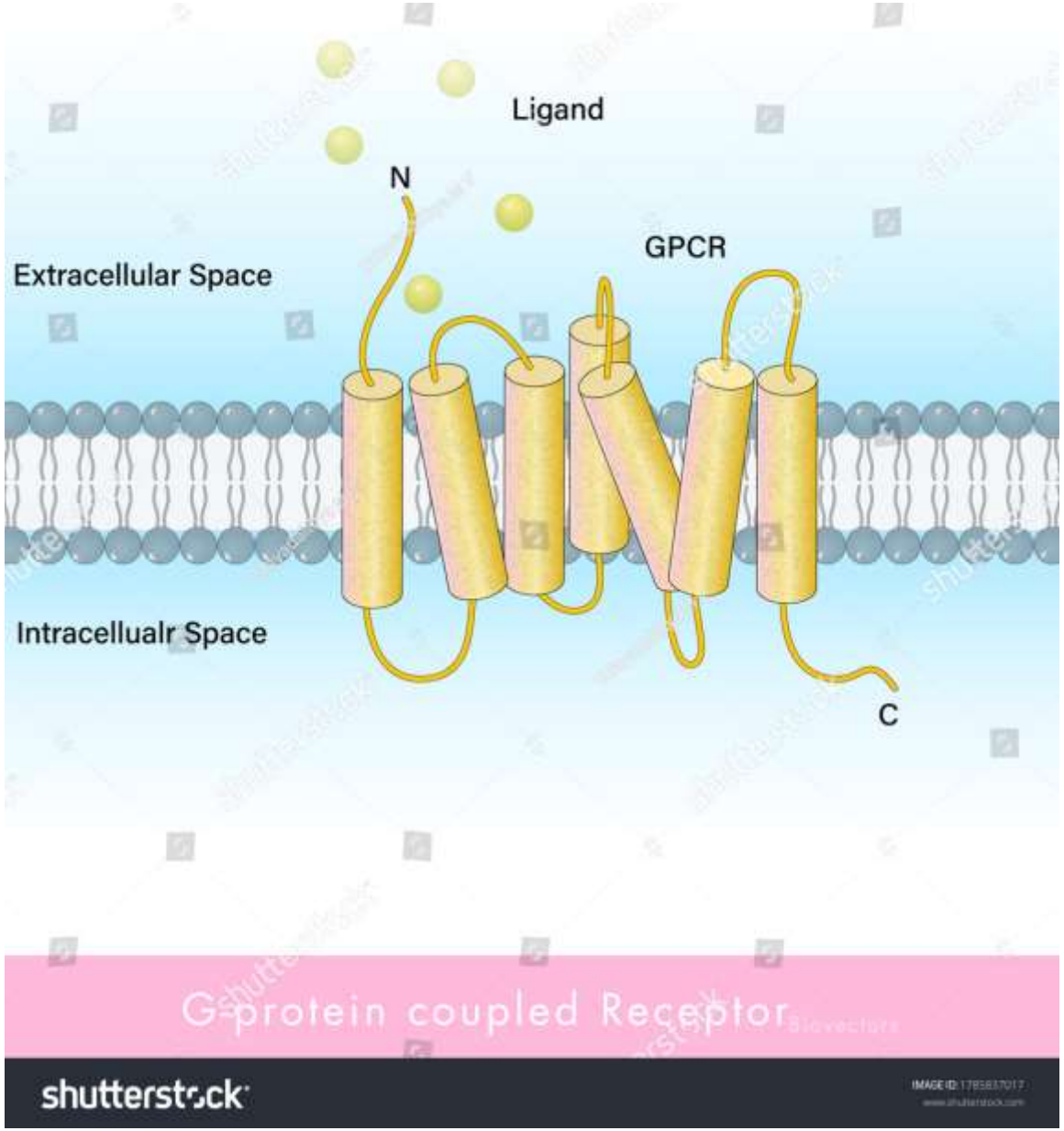
G-Protein-Coupled Receptors

- They transmit Signals through the Trimeric G Proteins
- G-protein-coupled receptors (GPCRs) are the largest family of cell-surface receptors, with more than 1,000 members.
- They indirectly regulate their effector targets, which can be ion channels or plasma membrane-bound effector enzymes, through the intermediary activity of a separate membrane-bound adapter protein complex called the trimeric GTP-binding regulatory protein or G protein
- GPCRs mediate cellular responses to numerous types of first messenger signaling molecules, including proteins, small peptides, amino acids, and fatty acid derivatives.
- Many 1st messenger ligands can activate several different GPCRs.

- GPCR’s have transcellular domain on one end of the molecule, separated by a seven-pass transmembrane-spanning region from the cytosolic regulatory domain at the other end, where the receptor interacts with the membrane-bound G protein.
- Binding of ligand or hormone to the extracellular domain results in a conformational change in the receptor that is transmitted to the cytosolic regulatory domain.
- This conformational change allows an association of the ligand-bound, activated receptor with a trimeric G protein associated with the inner leaflet of the plasma membrane

- There are more than 1000 known G-proteins – coupled receptors (GPCRS)
- All have seven transmembrane segments that loop in and out of the cell membrane.
- Some parts of the receptor that protrudes into cytoplasm are coupled to G-proteins that includes three (trimeric) parts – α, β and γ sub-units.
- The trimeric G-proteins are named for the ability to bind guanosine diphosphate (GDP) on the alpha sub-unit.




Ligand
N
GPCR
Extracellular Space
Intracellualr Space
C
G-protein coupled Receptor
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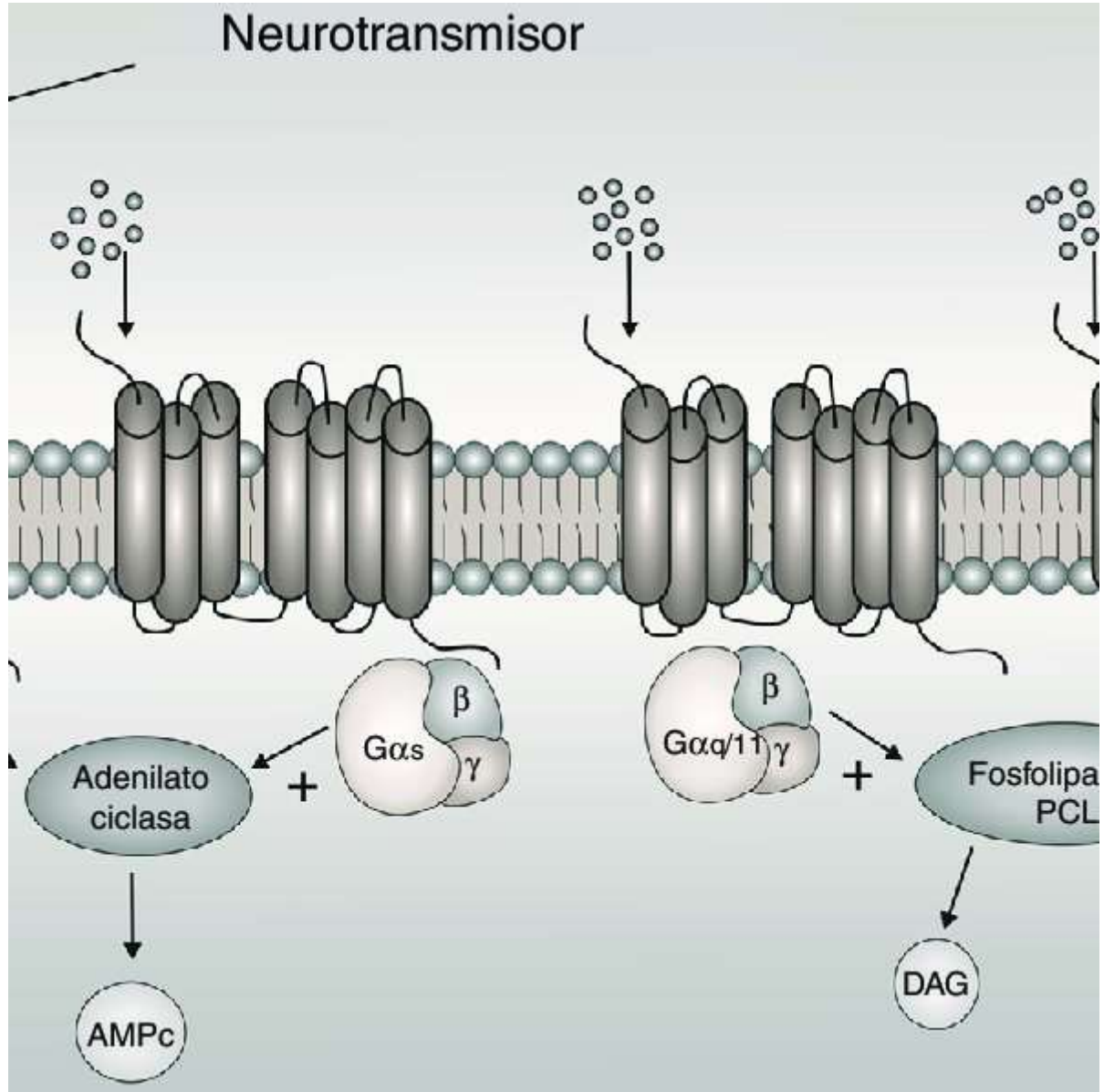

Neurotransmisor
β
Gαs
γ
Adenilato
ciclasa
+
AMPc
β
Gαq/11
γ
+
Fosfolipa
PCL
DAG

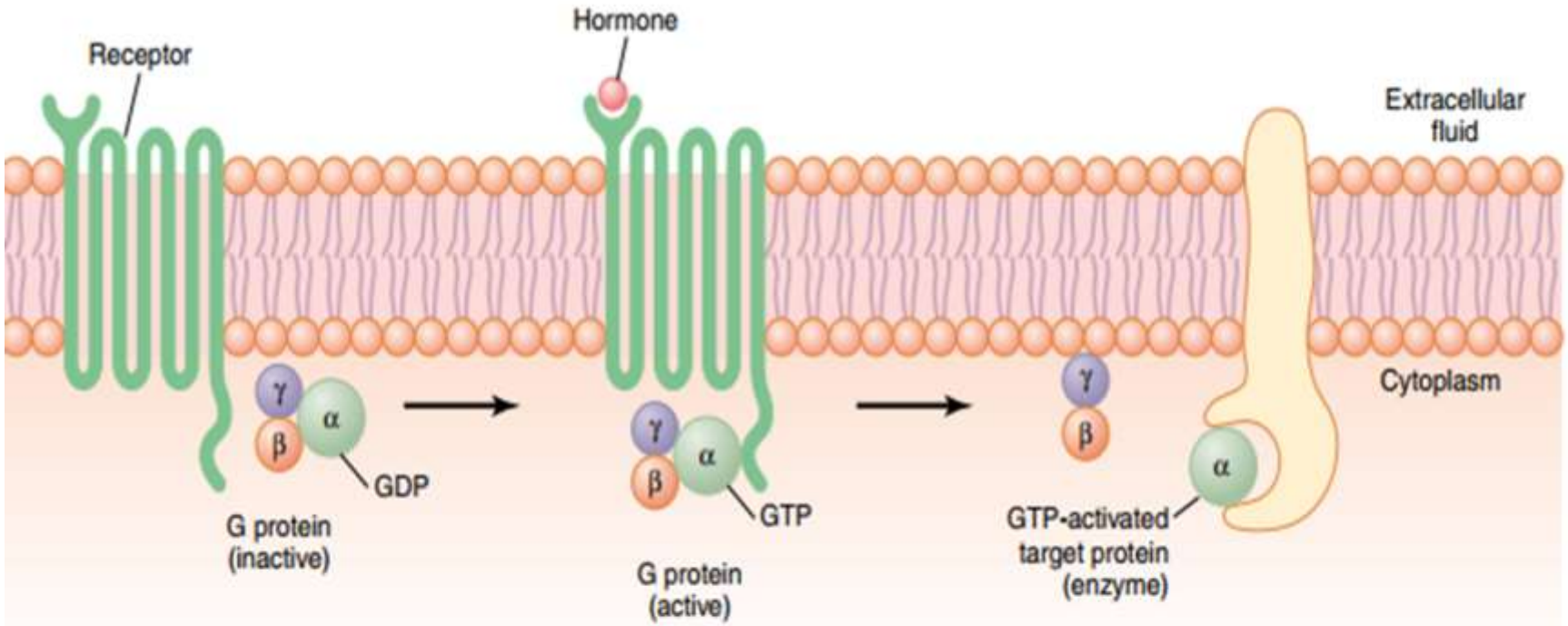


Figure 74-4 Mechanism of activation of a G protein–coupled receptor. When the hormone activates the receptor, the inactive α, β, and γ G protein complex associates with the receptor and is activated, with an exchange of guanosine triphosphate (GTP) for guanosine diphosphate (GDP). This causes the α subunit (to which the GTP is bound) to dissociate from the β and γ subunits of the G protein and to interact with membrane-bound target proteins (enzymes) that initiate intracellular signals.

- Interaction between activated receptor and the G protein, activates the G protein
- Activated G protein dissociates from the receptor and transmits the signal to its effector enzyme or ion channel
- The trimeric G proteins are named for their requirement for GTP binding and hydrolysis and have been shown to have a broad role in linking various seven-pass transmembrane receptors to membrane-bound effector systems that generate intracellular messengers.
- G proteins are tethered to the membrane through lipid linkage and are heterotrimeric, that is, composed of three distinct subunits.
- The subunits of a G protein are an α subunit, which binds and hydrolyzes GTP, and β and γ subunits, which form a stable, tight noncovalent-linked dimer.

- In resting state, the α sub-units binds GDP, and is associated with the β and γ subunits to form a trimeric complex that can interact with the cytoplasmic domain of the GPCR
- The conformational change that occurs upon ligand binding causes the GDP bound trimeric (α, β and γ complex) G protein to associate with the ligand-bound receptor.
- The association of the GDP-bound trimeric complex with the GPCR activates the exchange of GDP for GTP.

- Displacement of GDP by GTP causes the α subunit to dissociate from the receptor and from the β-γ subunits of the G protein.
- This exposes an effector binding site on the α subunit, which then associates with an effector molecule (e.g., adenylyl cyclase or phospholipase C) to result in the generation of second messengers (e.g., cAMP or IP3 and DAG).
- The hydrolysis of GTP to GDP by the α subunit results in the re-association of the α and β-γ subunits, which are then ready to repeat the cycle.

- The G proteins functionally couple receptors to several different effector molecules.
- Two major effector molecules regulated by G-protein subunits are adenylyl cyclase (AC) and phospholipase C (PLC).
- The association of an activated G-α subunit with AC can result in either the stimulation or the inhibition of the production of cAMP.
- This disparity is due to the two types of α subunit that can couple AC to cell-surface receptors.
- Association of an α s subunit (s for stimulatory) promotes the activation of AC and production of cAMP

- The association of an α i (I for inhibitory) subunit promotes the inhibition of AC and a decrease in cAMP.
- Thus, bidirectional regulation of adenylyl cyclase is achieved by coupling different classes of cell-surface receptors to the enzyme by either Gs or Gi
- In addition to α s and αi subunits, other isoforms of G-α protein subunits have been described.
- A q activates PLC, resulting in the production of the second messengers diacylglycerol (DAG) and inositol trisphosphate (IP3)

Sequence of events

i. Hormone binds with the receptor in the cell membrane and forms the hormone-receptor complex

ii. It activates the G protein

iii. G protein releases GDP from α-GDP unit

v. The α-subunit now binds with a new molecule of GTP, i.e. the GDP is exchanged for GTP

v. This exchange triggers the dissociation of α-GTP unit and β-γ dimmer from the receptor

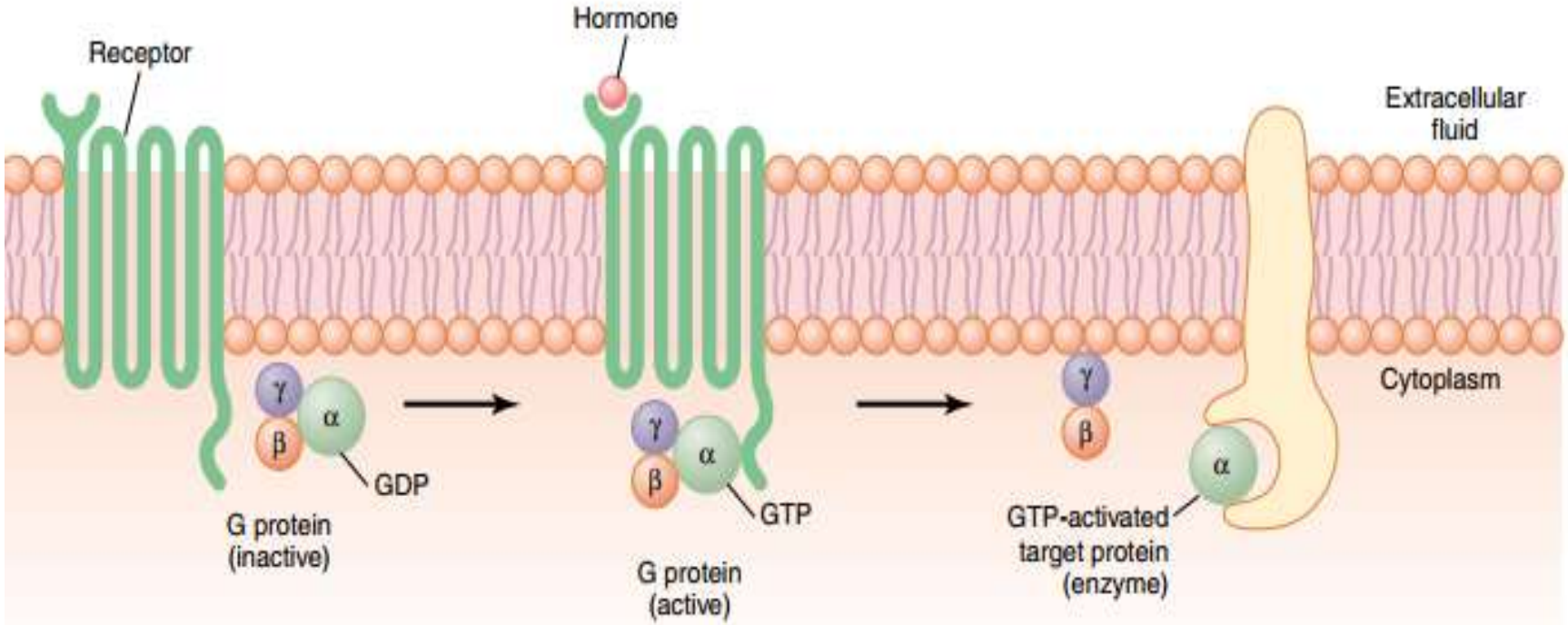
vi. Both α-GTP unit and β-γ dimmer now activate the second messenger pathways

vii. The α-GTP unit activates the enzyme adenylyl cyclase, which is also present in the cell membrane. Most of the adenyl cyclase protrudes into the cytoplasm of the cell from inner surface of the cell membrane

viii. Activated adenyl cyclase converts the ATP of the cytoplasm into cAMP

- When the action is over, α-subunit hydrolyzes the attached GTP to GDP by its GTPase activity.
- This allows the reunion of α-subunit with β-γ dimmer and commencing a new cycle

- When receptor activated (ie, this occurs when a hormone binds) conformational change causes GDP bound to G protein to be converted to GTP
- Replacement of GDP by GTP causes $\boldsymbol{\alpha}$ subunit to dissociate from trimeric complex
- It ($\boldsymbol{\alpha}$ subunit) then associates with intracellular signaling proteins; which in turn alter ion channel activity or intracellular enzymes such as adenylyl cyclase or phospholipase A which alter cell function



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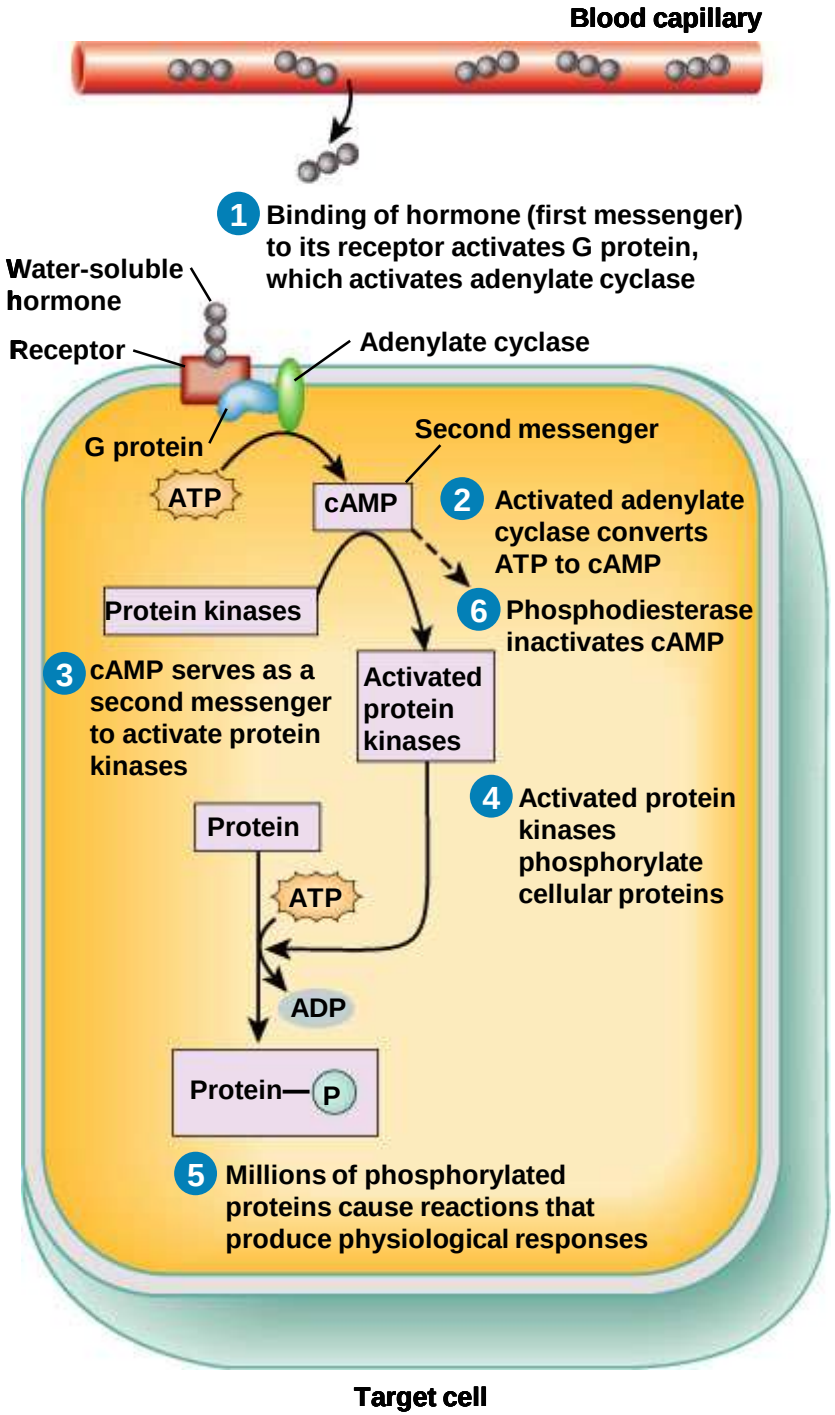
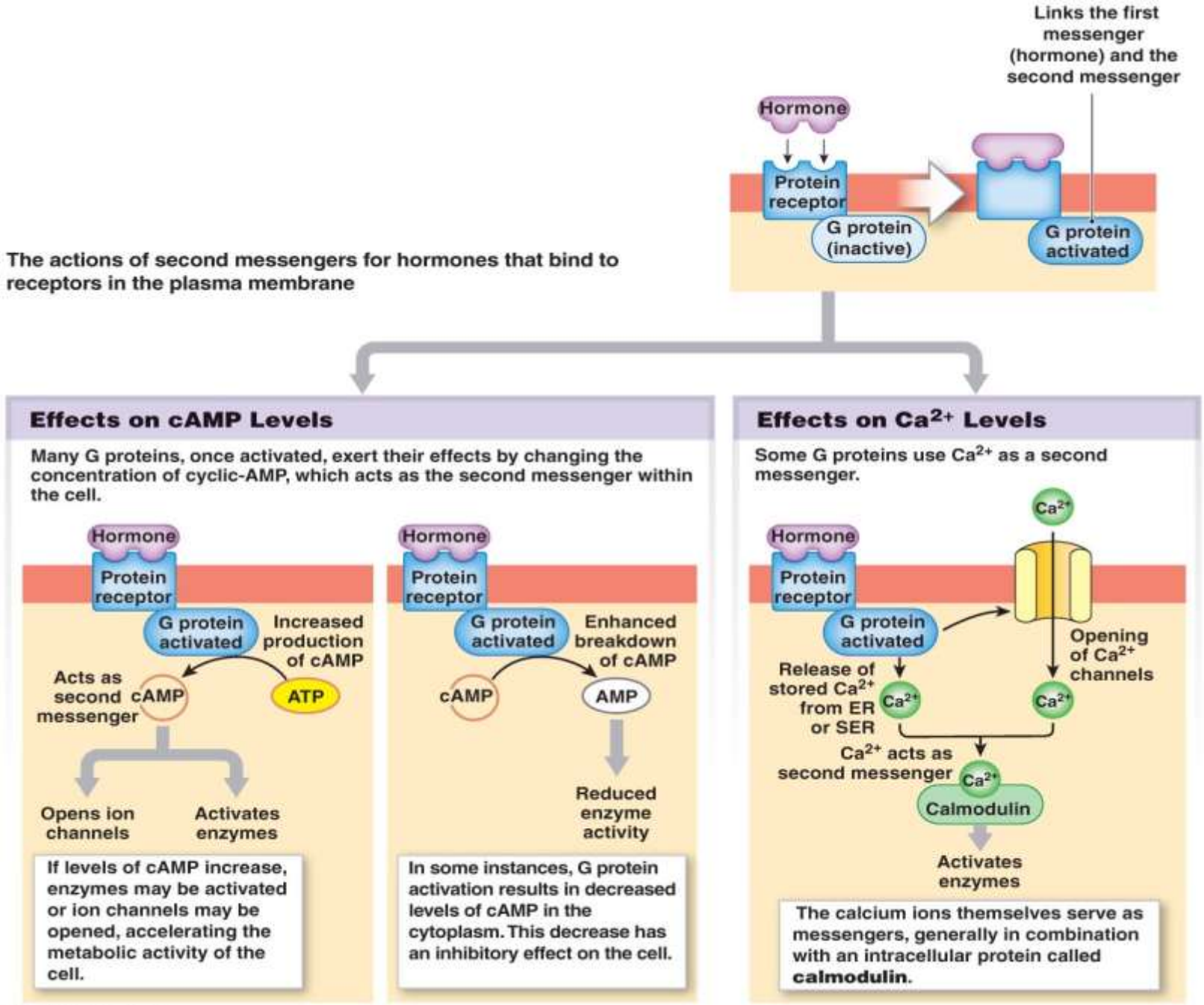
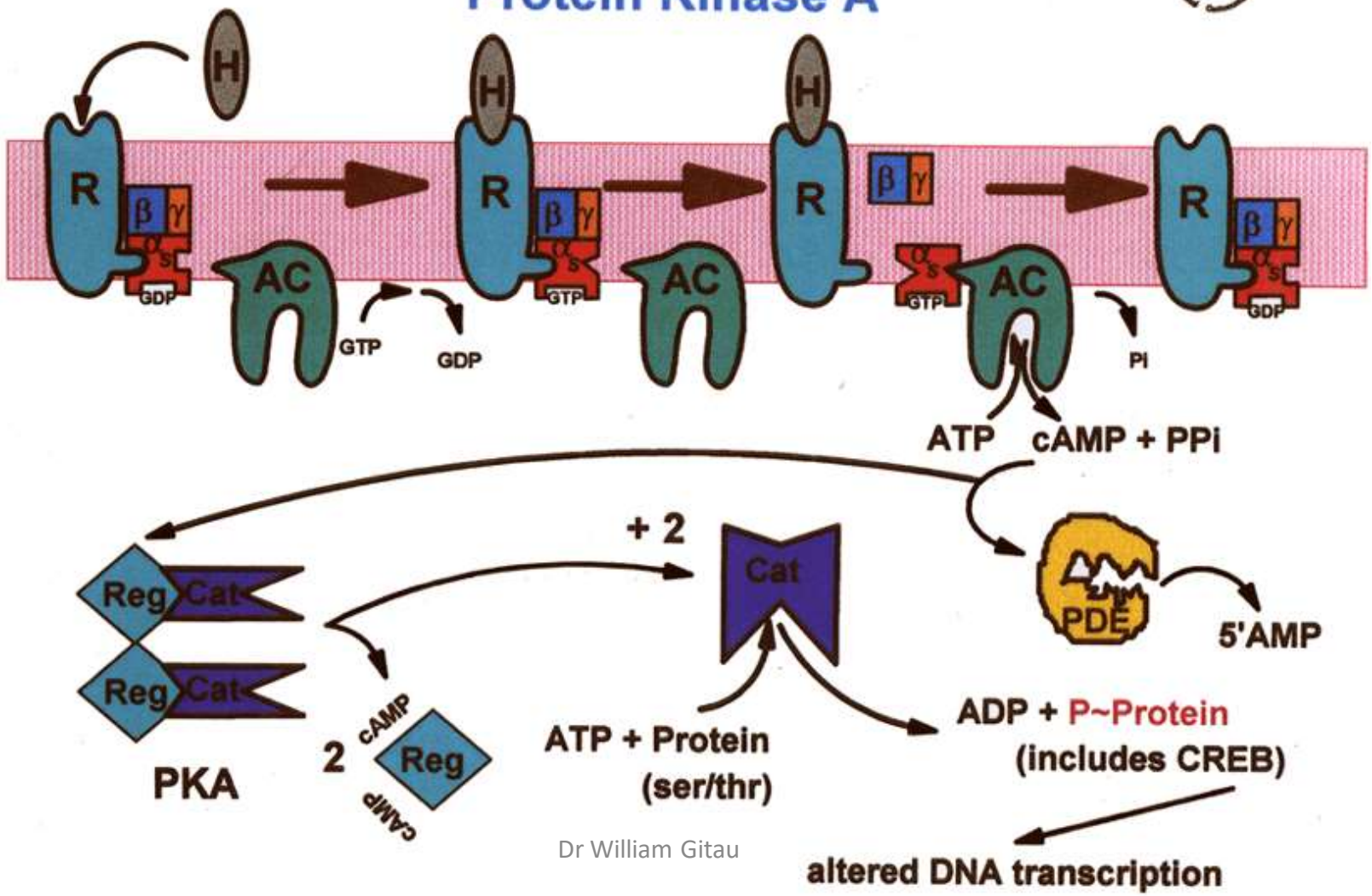
- Signalling is terminated when the hormone is removed
- This causes the ***α*** subunit to inactivates itself by converting GTP to GDP, then the ***α*** subunit recombines with the other subunits to form inactive, membrane bound trimeric G Protein


Blood capillary
1 Binding of hormone (first messenger) to its receptor activates G protein, which activates adenylate cyclase
Water-soluble hormone
Receptor
Adenylate cyclase
G protein
Second messenger
ATP
cAMP
2 Activated adenylate cyclase converts ATP to cAMP
Protein kinases
6 Phosphodiesterase inactivates cAMP
3 cAMP serves as a second messenger to activate protein kinases
Activated protein kinases
4 Activated protein kinases phosphorylate cellular proteins
Protein
ATP
ADP
Protein—P
5 Millions of phosphorylated proteins cause reactions that produce physiological responses
Target cell

The actions of second messengers for hormones that bind to receptors in the plasma membrane



Large G Proteins, Adenylyl Cyclase, Protein Kinase A



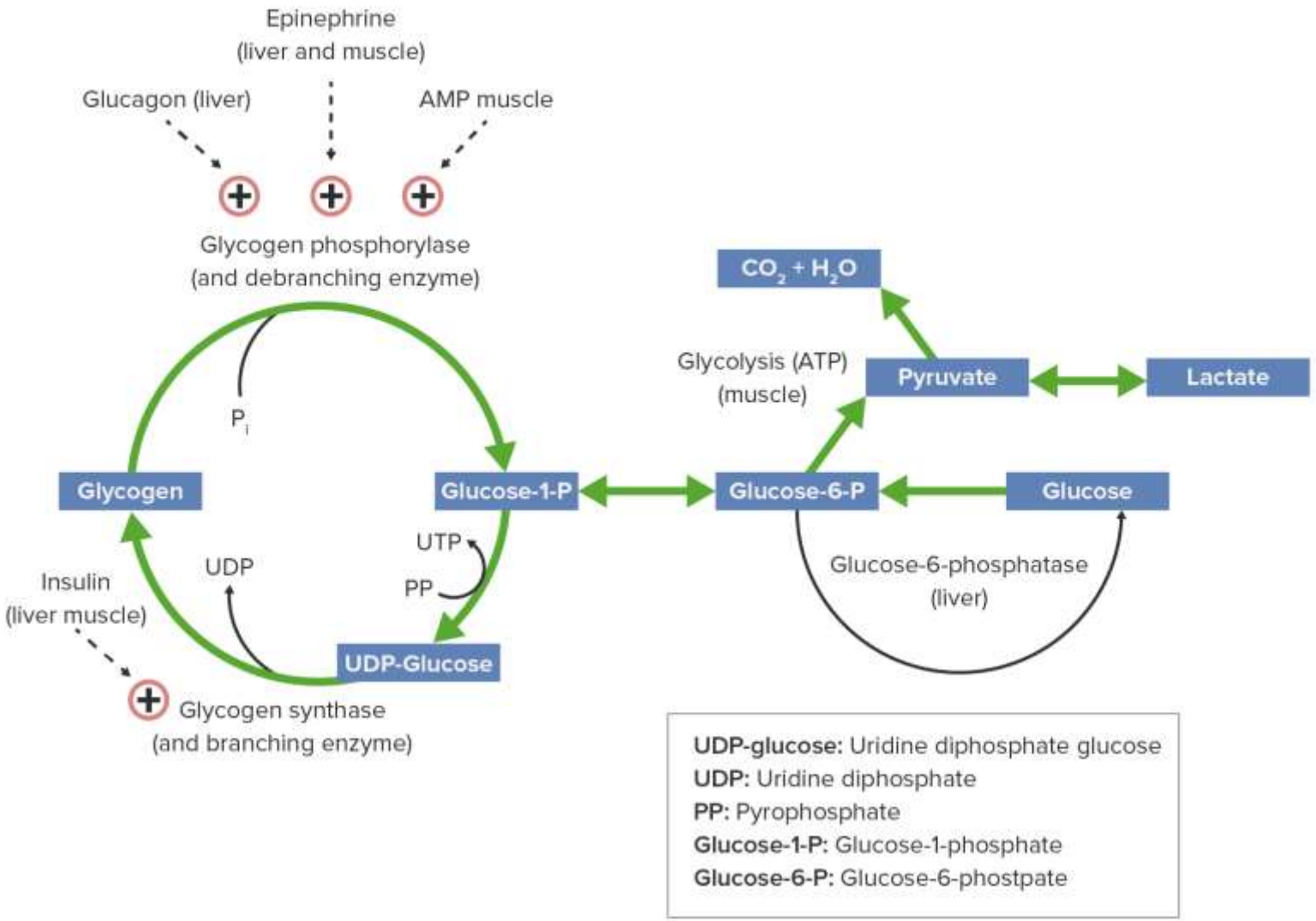
CYCLIC AMP PATHWAY(CYCLIC ADENOSINE MONOPHOSPHATE)

- In this 2nd messenger system, the Receptor/trimeric G protein is closely related to a membrane bound enzyme, called adenylyl cyclase
- Once the α subunit dissociates from the β, and γ subunits, it associates with the enzyme activating it
- The activated adenyl cyclase enzyme catalyzes the conversion of small amounts of adenosine triphosphate (ATP) into cyclic Adenosine mophosphate (cAMP).

- The cAMP plays fundamental roles in cellular responses to many hormone and neurotransmitters.
- The cAMP generated activates the cAMP dependent protein kinase (PKA).
- The PKA will then activate enzymes or inactivate enzymes for cellular activity

- The intracellular levels of cAMP are regulated by the activities of the Adenyl cyclase enzymes

- This system takes part in essential processes such as energy metabolism, muscle contraction, membrane transport and gene expression.
- It (PKA) phosphorylates numerous metabolic enzymes which include.
- Glycogen synthase
- Phosphorylate Kinase which inhibit glycogen synthesis and promotes glycogen breakdown to glucose.
- Acetyl COA carboxylase which inhibit lipid synthesis


Epinephrine
(liver and muscle)
Glucagon (liver)
AMP muscle
Glycogen phosphorylase
(and debranching enzyme)
$CO_2 + H_2O$
Glycolysis (ATP)
(muscle)
Pyruvate
Lactate
P_i
Glycogen
Glucose-1-P
Glucose-6-P
Glucose
UTP
UDP
PP
Insulin
(liver muscle)
Glucose-6-phosphatase
(liver)
UDP-Glucose
Glycogen synthase
(and branching enzyme)
UDP-glucose: Uridine diphosphate glucose
UDP: Uridine diphosphate
PP: Pyrophosphate
Glucose-1-P: Glucose-1-phosphate
Glucose-6-P: Glucose-6-phostpate

- It affects gene expression (protein synthesis)
- This is via presence of transcription factors referred to as cAMP-Response Element Binding Protein (CREB).
- Once activated, the CREB will interact with the transcriptional co-activators CREB-binding protein (CBP) in the nucleus to cause synthesis of proteins or inhibit synthesis or proteins

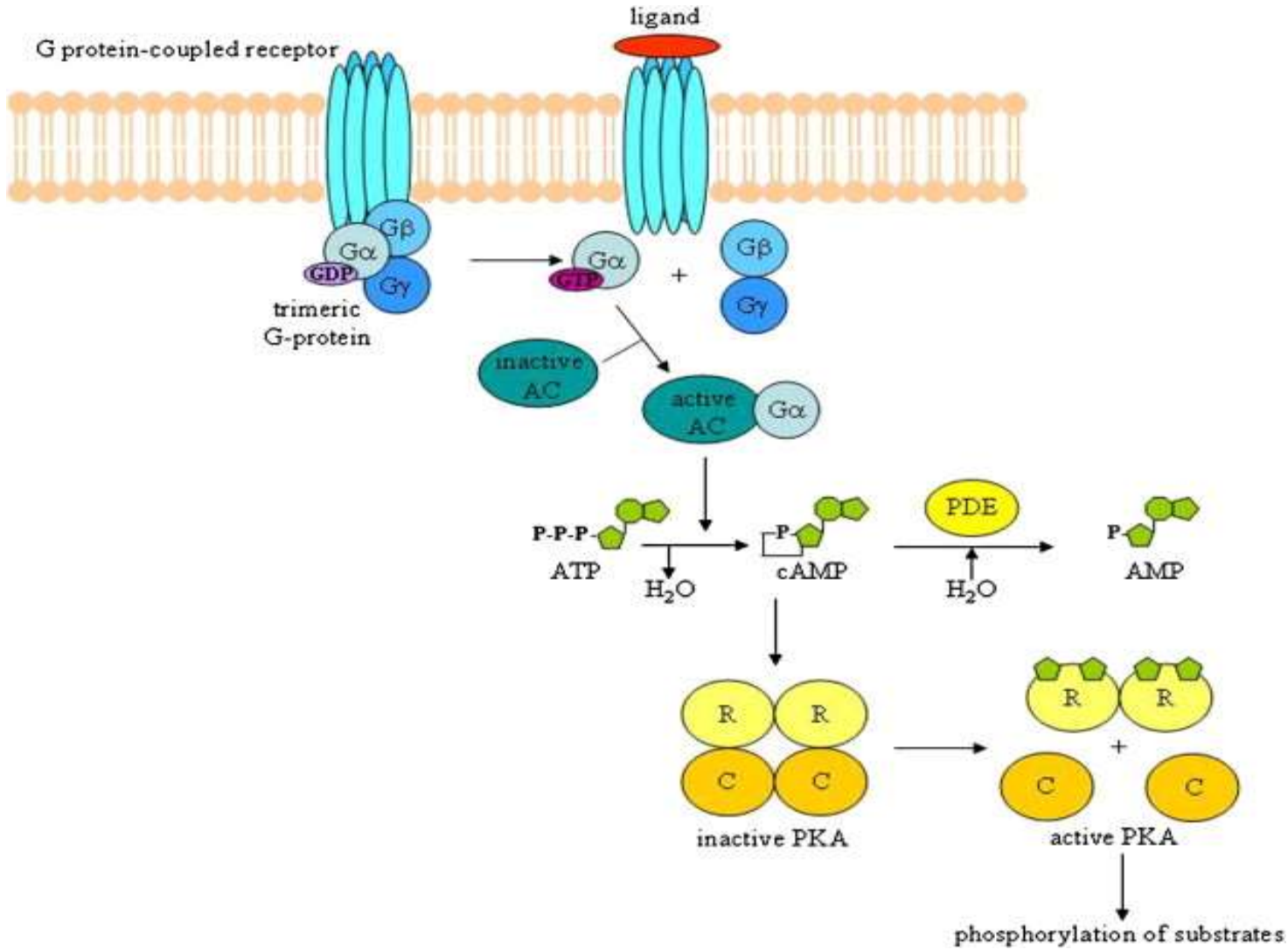
Importance of cAMP 2nd messenger system

- In the cell it activates a cascade of enzymes .i.e. one enzymes once activated activates the other and so on causing several effects in the cell and tissues.
- Few molecules of Adenyle cyclase enzymes inside the cell membrane cause activation of many various molecules.
- In this way even the slightest amount of hormone acting on the cell surface can initiate a powerful cascading activating force for the entire cells.

Protein Kinase A Is the Major Mediator of the Signaling Effects of cAMP

- cAMP activates protein kinase A (or cAMP-dependent protein kinase)
- PKA in turn catalyzes the phosphorylation of various cellular proteins, ion channels, and transcription factors.
- This phosphorylation alters the activity or function of the target proteins and ultimately leads to a desired cellular response.
- PKA may in some cell types, cAMP directly binds to and affects
- the activity of ion channels.
- Protein

- PKA consists of catalytic and regulatory subunits, with the kinase (phosphorylating) activity residing in the catalytic subunit.
- When cAMP conc’n in the cell are low, the 2 regulatory subunits bind to and inactivate 2 catalytic subunits, forming an inactive tetramer
- When cAMP is formed 2 molecules of cAMP bind to each of the regulatory subunits, causing them to dissociate from the catalytic subunits.
- This relieves the inhibition of catalytic subunits and allows them to catalyze the phosphorylation of target substrates and produce the resultant biological response

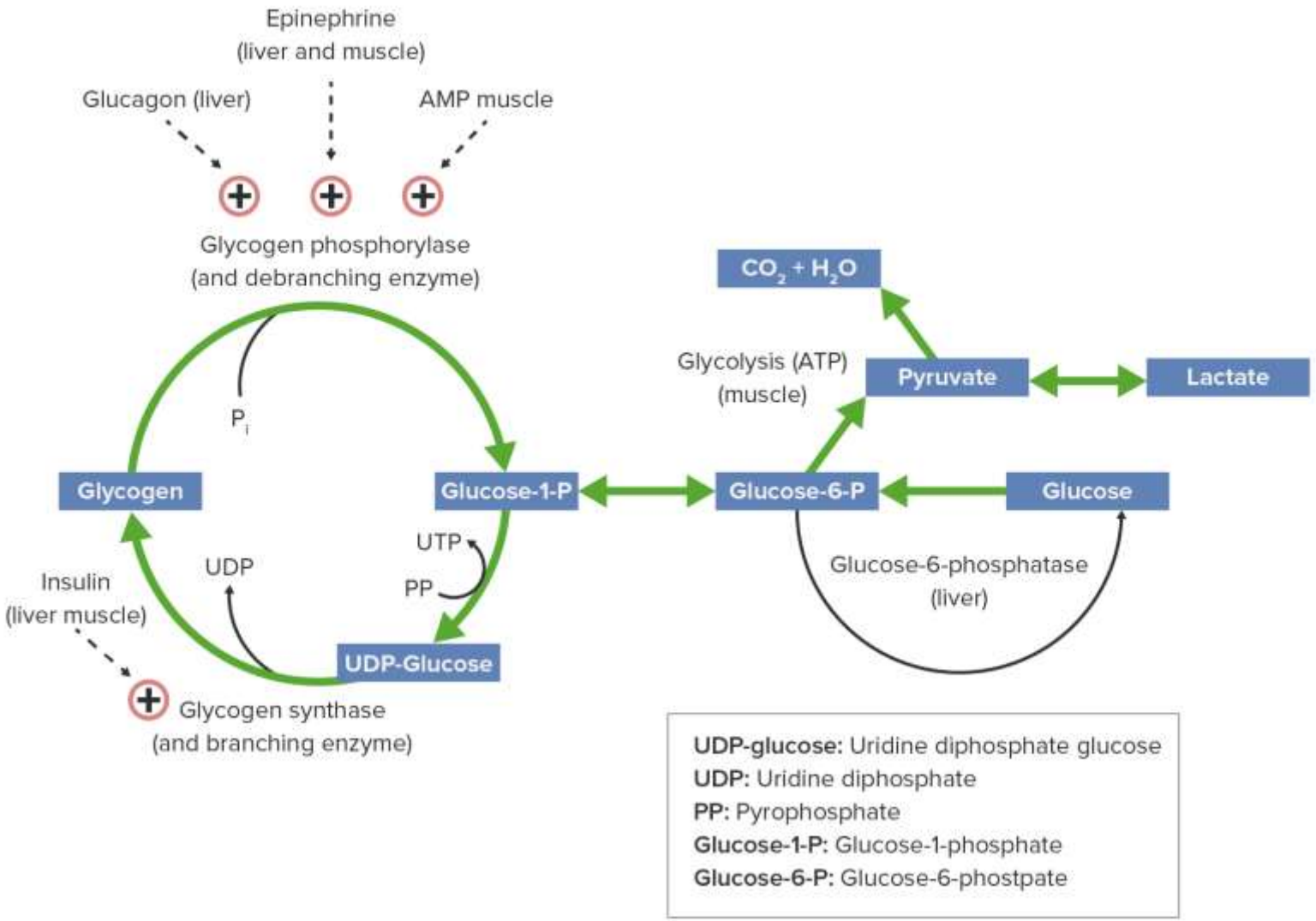

ligand
G protein-coupled receptor
Gβ
Gα
GDP
Gγ
trimeric
G-protein
Gα
GDP
+
Gβ
Gγ
inactive
AC
active
AC
Gα
P-P-P
ATP
H2O
P
cAMP
PDE
H2O
P
AMP
R
R
C
C
inactive PKA
R
R
+
C
C
active PKA
phosphorylation of substrates

Application in every day life of PKA is the "fight" or flight response.

- Example: If you were to cross a road and a car accelerates towards you as were doing so, the immediate response would be to run across the road in order to avoid being hit.
- Response is caused by the following reactions.
- The car acts as stimulus and the response is to run away which requires energy.
- This energy is in the form of ATP which is made by aerobic respiration which requires glucose.

- Glucose is generated by the breakdown of glycogen.
- The stimulus causes the adrenal glands to secrete the hormone epinephrine
- It will be released into blood
- Once it reaches the liver, it will bind to epinephrine receptors in the plasma membrane of liver cells
- This binding activates the trimeric G. proteins in the cytoplasm causing a conformational change which results in conversion of GDP to GTP

- This causes dissociation of the $\boldsymbol{\alpha}$ subunit from the $\boldsymbol{\beta}$, and $\boldsymbol{\gamma}$ subunits
- The $\boldsymbol{\alpha}$ subunit then binds to adenylyl cyclase activating it
- The activated adenylyl cyclase then causes conversion of ATP to cAMP.
- cAMP then binds to protein kinase A activating it


Epinephrine
(liver and muscle)
Glucagon (liver)
AMP muscle
Glycogen phosphorylase
(and debranching enzyme)
$CO_2 + H_2O$
Glycolysis (ATP)
(muscle)
Pyruvate
Lactate
P_i
Glycogen
Glucose-1-P
Glucose-6-P
Glucose
UTP
UDP
PP
Insulin
(liver muscle)
Glucose-6-phosphatase
(liver)
UDP-Glucose
Glycogen synthase
(and branching enzyme)
UDP-glucose: Uridine diphosphate glucose
UDP: Uridine diphosphate
PP: Pyrophosphate
Glucose-1-P: Glucose-1-phosphate
Glucose-6-P: Glucose-6-phostpate

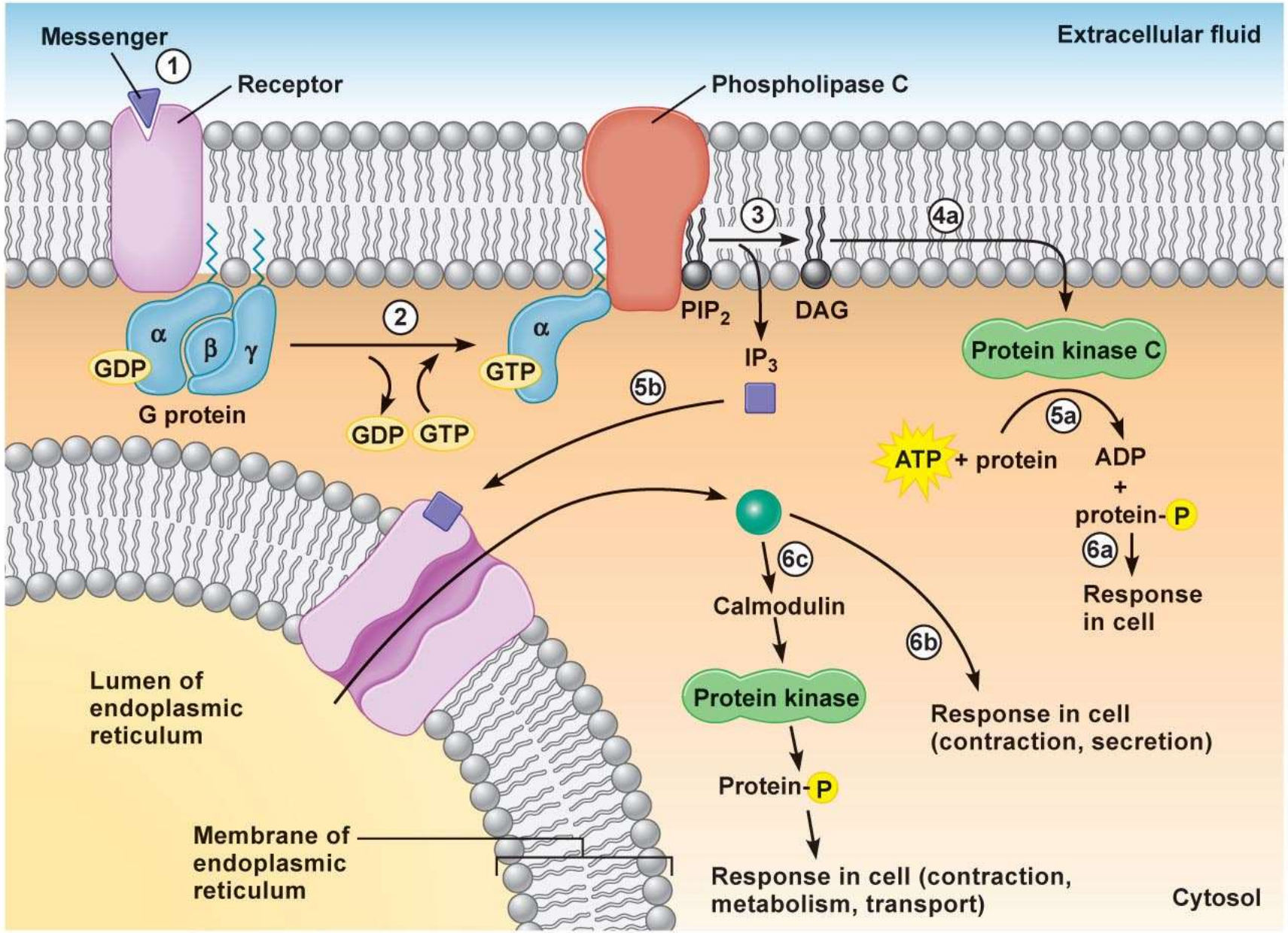
- PKA then goes on to phosphorylate **Glycogen Synthase A** into **Glycogen** S**ynthase B** which inactivates it (this is done to prevent glycogen synthesis whilst it is being broken down into glucose).
- PKA also phosphorylates **Phosphorylase kinase**
- Phosphorylase kinase then phosphorylates **Phosphorylase B** to **Phosphorylase A**
- Phosphorylase A catalyzes the conversion of glycogen to glucose which is then used in aerobic respiration to generate ATP
- This ATP provides the energy required to run away.

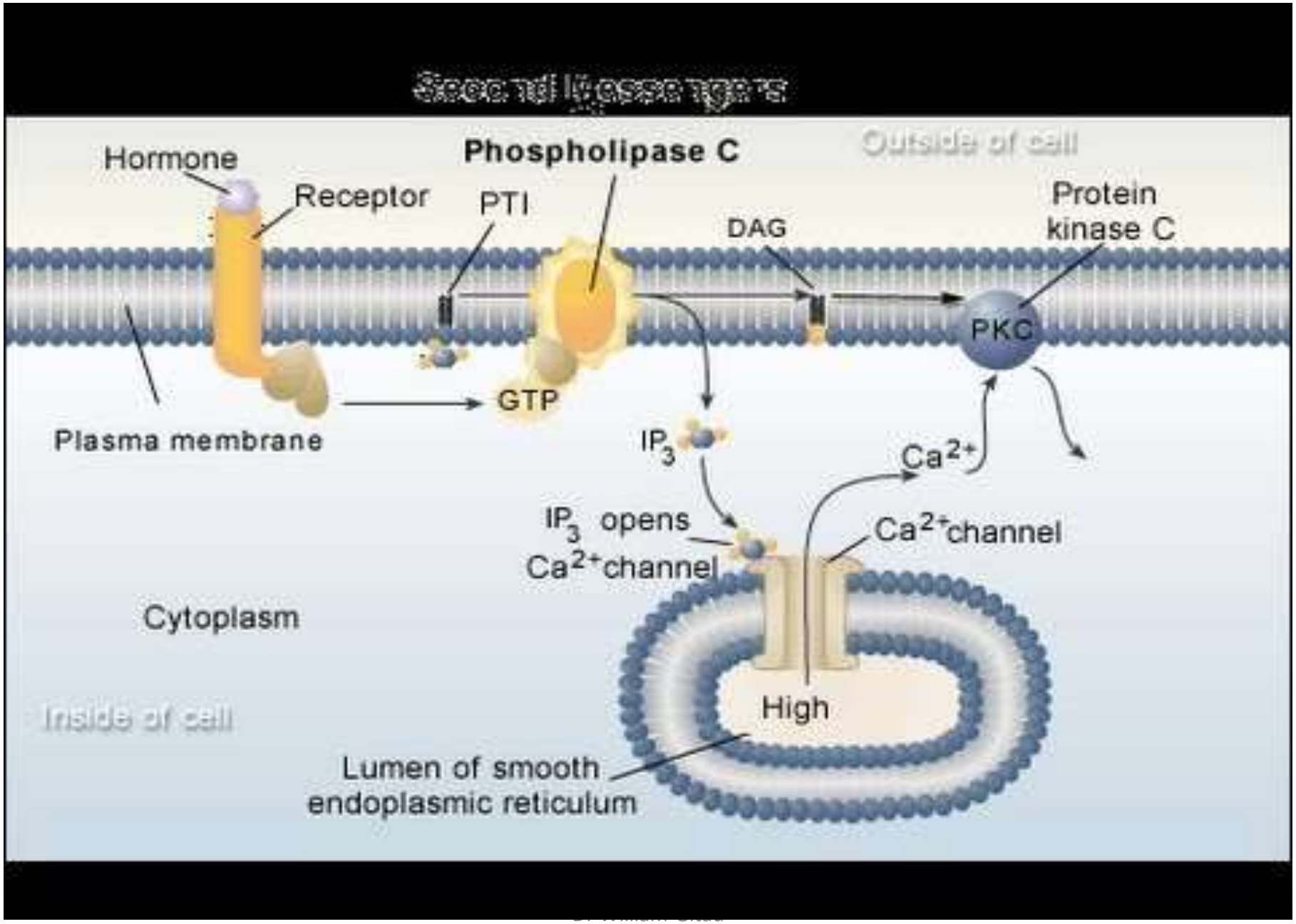
Phosphatidyl Inositol (PIP2)

- A membrane phospholipid
- Once ligand binds to receptor, activation of Phospholipase C (PLC)
- Causes splitting of phospholipid to Diacyl Glycerol (DAG) and Inositol triphosphate
- Via trimeric proteins
- DAG activates Protein Kinase C (PKC) which phosphorylates enzymes for activity
- Both DAG and IP3 serve as second messengers in the cell

- In its 2nd messenger role, DAG accumulates in the plasma membrane and activates the membrane-bound enzyme protein kinase C
- Activated PKC catalyzes the phosphorylation of specific proteins, including other enzymes and transcription factors, in the cell to produce appropriate physiological effects, such as cell proliferation.
- Several tumor-promoting phorbol esters that mimic the structure of DAG have been shown to activate PKC
- They can, therefore, bypass the receptor by passing through the plasma membrane and directly activating PKC causing the phosphorylation of downstream targets to result in cellular proliferation

- IP3 promotes the release of Ca++ ions into the cytoplasm by activation of endoplasmic or sarcoplasmic reticulum IP3-gated calcium release channels
- Increased Ca++ synergizes with the action of DAG in the activation of PKC
- It may also activate many other calcium-dependent processes
- IP3 is rapidly dephosphorylated to inositol & the DAG is converted to phosphatidic acid by the addition of a phosphate thus terminating the effects
- On removal of IP3, Ca++ rapidly pumped back to storage sites




Second messengers
Hormone
Receptor
Phospholipase C
PTI
Outside of cell
DAG
Protein kinase C
PKC
GTP
Plasma membrane
IP3
Ca2+
IP3 opens Ca2+ channel
Ca2+ channel
Cytoplasm
Inside of cell
High
Lumen of smooth endoplasmic reticulum

Cells Use Calcium as a Second Messenger

- Ca++ levels in cytoplasm are very low and kept so by powerful pumps into ECF and ER
- Several plasma membrane ion channels serve to increase Ca++ levels.
- Either are voltage-gated or may be controlled by phosphorylation via PKA or PKC on plasma membrane
- ER has 2 main Ca++ channels

1. IP3-gated calcium release channel
2. The ryanodine receptor, found in the sarcoplasmic reticulum of muscle cells

- Both channels regulated by +ve feedback where the released Ca++ can bind to the receptor to enhance further Ca++ release.
- This causes the Ca++ to be released suddenly in a spike, followed by a wave-like flow of the ion throughout the cytoplasm

- Ca++ activates many different signaling pathways and leads to numerous physiological events, such as Muscle contraction, neurotransmitter secretion among others
- It acts as a 2^{nd} messenger in two ways:
- • It binds directly to an effector molecule, such as PKC, to participate in its activation.
- • It binds to an intermediary Ca-binding protein, such as calmodulin
- Binding causes calmodulin to undergo a conformational change and increases the affinity of this intracellular calcium “receptor” for its effectors
- Ca-calmodulin complexes bind to and activate a variety of cellular proteins, including protein kinases that are important in many physiological processes, such as smooth muscle contraction (where activates Myosin light chain kinase enzyme)

Ion Channel-Linked Receptors

- Ion channels, found in all cells, are transmembrane proteins that cross the plasma membrane and are involved in regulating the passage of specific ions into and out of cells.
- They may be opened or closed by changing the membrane potential or by the binding of ligands, such as neurotransmitters or hormones, to membrane receptors.
- In some cases, the receptor and ion channel are one and the same molecule.
- Eg: At the neuromuscular junction, the neurotransmitter acetylcholine binds to a Ach receptor that is also an ion channel

- In other cases, the receptor and an ion channel are linked via a G protein, 2nd messengers, and other downstream effector molecules, as in the Ach cholinergic receptor.
- Another possibility is that the ion channel is directly activated by a cyclic nucleotide, such as cGMP or cAMP, produced as a consequence of receptor activation.
- This mode of ion channel control is predomnantly found in the sensory tissues for sight, smell, and hearing.
- The opening or closing of ion channels plays a key role in signaling between electrically excitable cells.

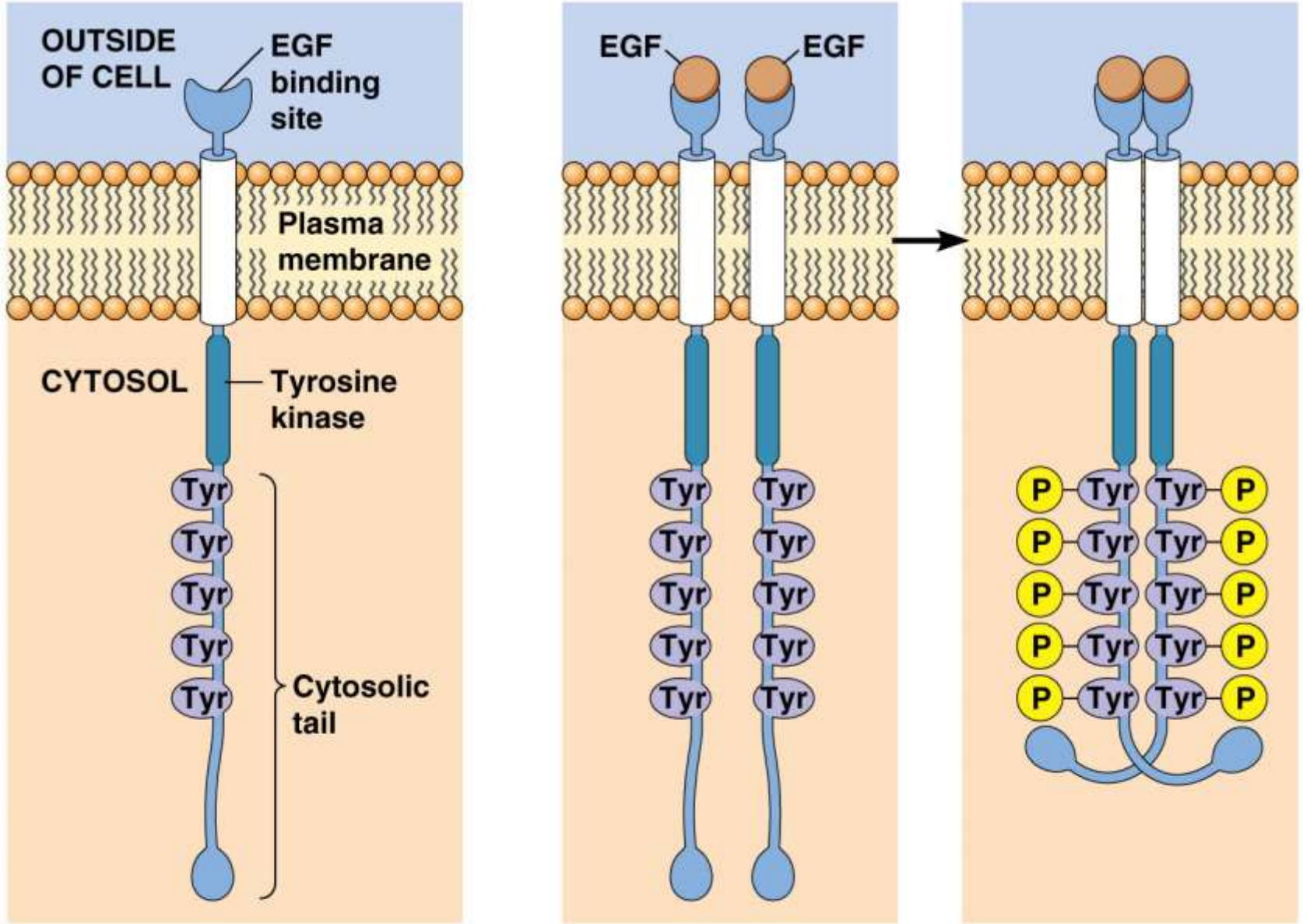
Tyrosine Kinase Receptors

- Many hormones, growth factors, and cytokines signal their target cells by binding to receptors that have tyrosine kinase activity and result in the phosphorylation of tyrosine residues in the receptor and other target proteins.
- Many of the receptors in this class of plasma membrane receptors have an intrinsic tyrosine kinase domain that is part of the cytoplasmic region of the receptor.
- Another group of related receptors lacks an intrinsic tyrosine kinase but, when activated, becomes associated with a cytoplasmic tyrosine kinase
- This family of tyrosine kinase receptors utilizes similar signal transduction pathway

- The receptors consist of a hormone-binding region that is exposed to the ECF. Examples include insulin, growth factors (e.g., epidermal, fibroblast, and platelet-derived growth factors), or cytokines.
- The signaling cascades results to gene activation with resultant protein synthesis, growth, cellular differentiation among others

Tyrosine Kinase receptors

- Are inactive monomers on plasma membrane
- 2 signal molecule binds to receptor sites causing conformational changes to structure causing them to become dimers
- Usually to regulate growth factors
- Stimulate cells to grow and repair tissue



(a) Structure of the epidermal growth factor (EGF) receptor

(b) Activation of the EGF receptor

- They undergo phosphorylation via multiple ATP’s to activate them
- This phosphorylated parts dangling inside the cytoplasm
- Once phosphorylated, active receptors now recognized by multiple relay proteins
- Each can trigger a separate cellular response
- NB: one tyrosine kinase can trigger many cellular responses

- One receptor tyrosine kinase can activate ten or more responses providing a way for cells to regulate growth
- Is major difference with G Proteins
- Many cancers caused by mutated tyrosine kinase receptors that activate without a signal molecule

- Examples include
- Epidermal growth factor receptors (EPGFR)
- Platelet Derived Growth Factor Receptor (PDGFR)
- Vascular Endothelial Growth Factor (VEGF)

SECOND MESSENGER SYSTEMS AND INTRACELLULAR SIGNALING PATHWAYS

- 2^{nd} messengers transmit and amplify the first messenger signal to signaling pathways inside the cell.
- Only a few 2^{nd} messengers are responsible for relaying these signals within target cells, and because each target cell has a different complement of intracellular signaling pathways, the physiological responses can vary.
- Thus, it is useful to keep in mind that every cell in our body is programmed to respond to specific combinations of messengers and that the same messenger can elicit a distinct physiological response in different cell types.
- Eg: The neurotransmitter acetylcholine can cause heart muscle to relax, skeletal muscle to contract, and secretory cells to secrete

INTRACELLULAR RECEPTORS AND HORMONE SIGNALING

- Located either in cytoplasm or nucleus
- Ligands must be lipid soluble to diffuse through cell membrane
- The mainresult of activation of the intracellular receptors is altered gene expression
- Steroid hormones receptors found in cytoplasm whereas thyroid hormone receptors found in nucleus

Steroid hormones

- When hormones bind, the hormone-receptor complex moves to the nucleus, where it binds to specific DNA sequences in the gene regulatory (promoter) region of specific hormone-responsive genes.
- The targeted DNA sequence in the promoter is called a hormone response element (HRE).
- Binding of the hormone-receptor complex to the HRE can either activate or repress transcription.
- The end result of stimulation by steroid hormones is a change in the readout or transcription of the genome.
- While most effects involve increased production of specific proteins, repressed production of certain proteins can also occur.
- These newly synthesized proteins and/or enzymes will affect cellular metabolism with responses attributable to that particular steroid hormone..