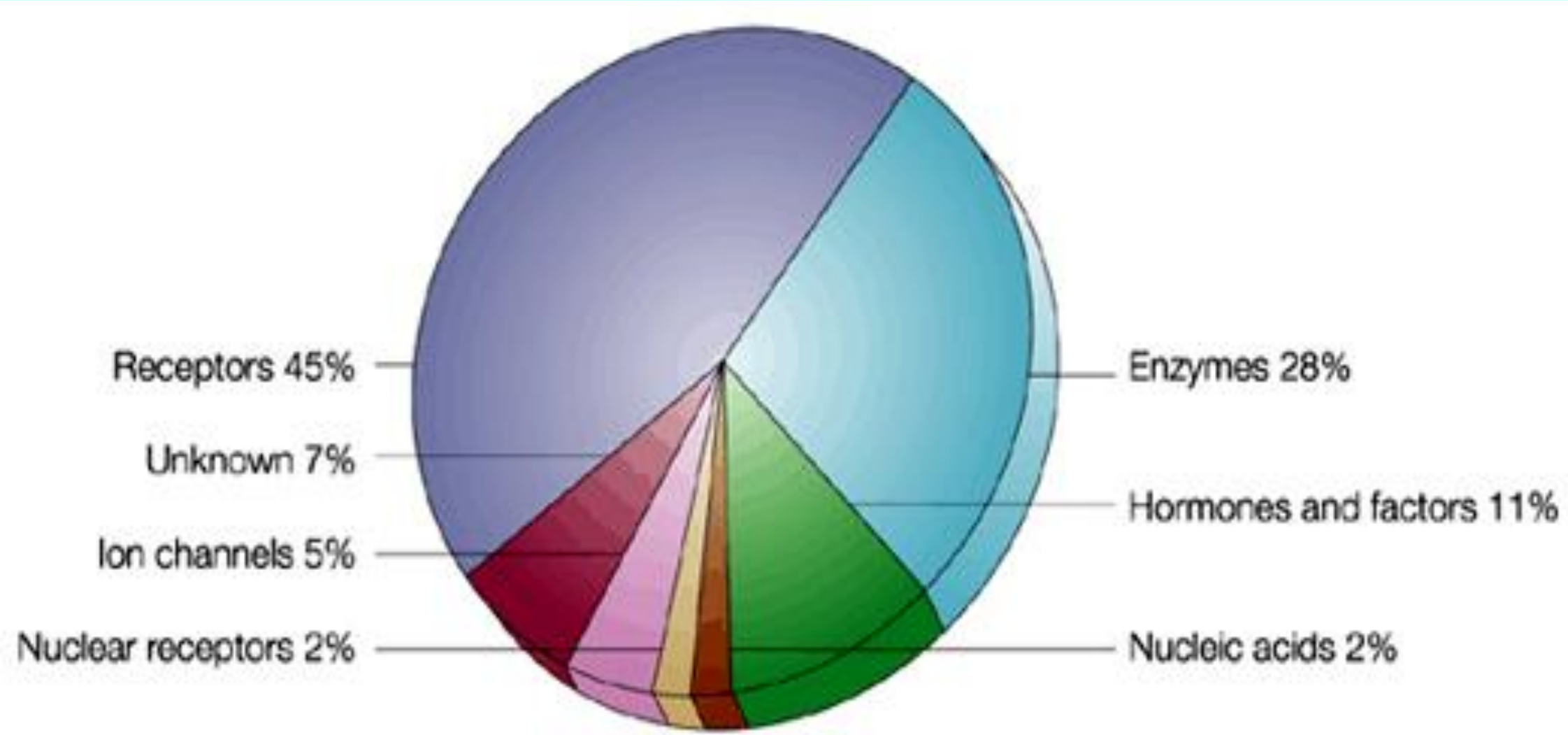
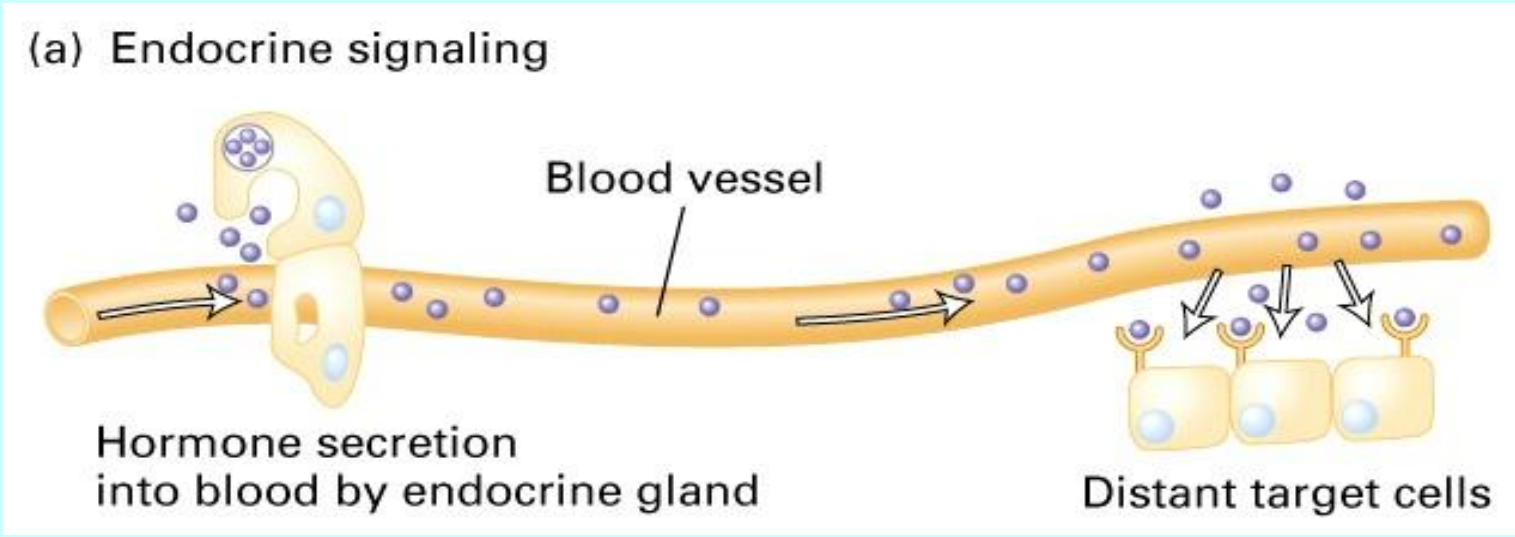
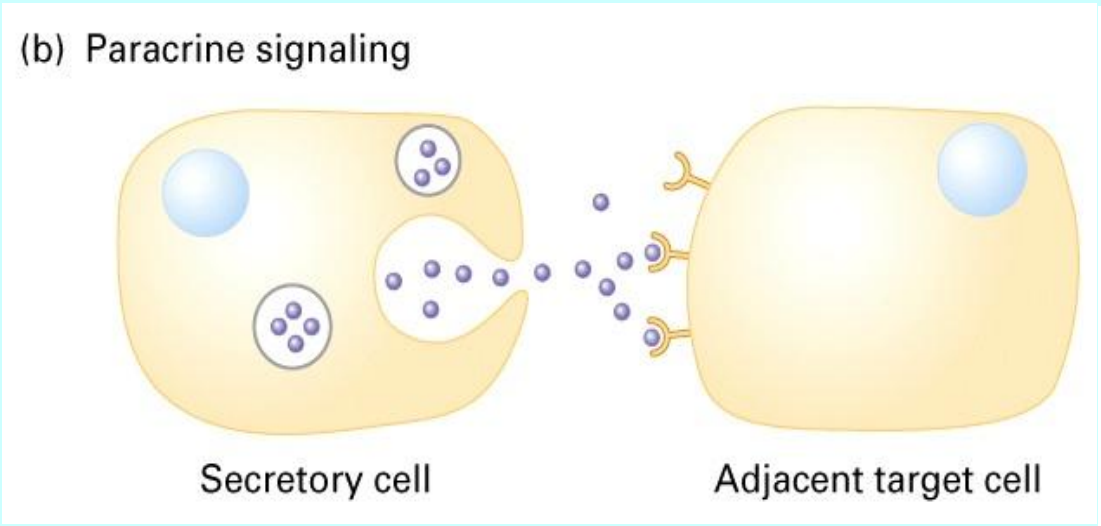
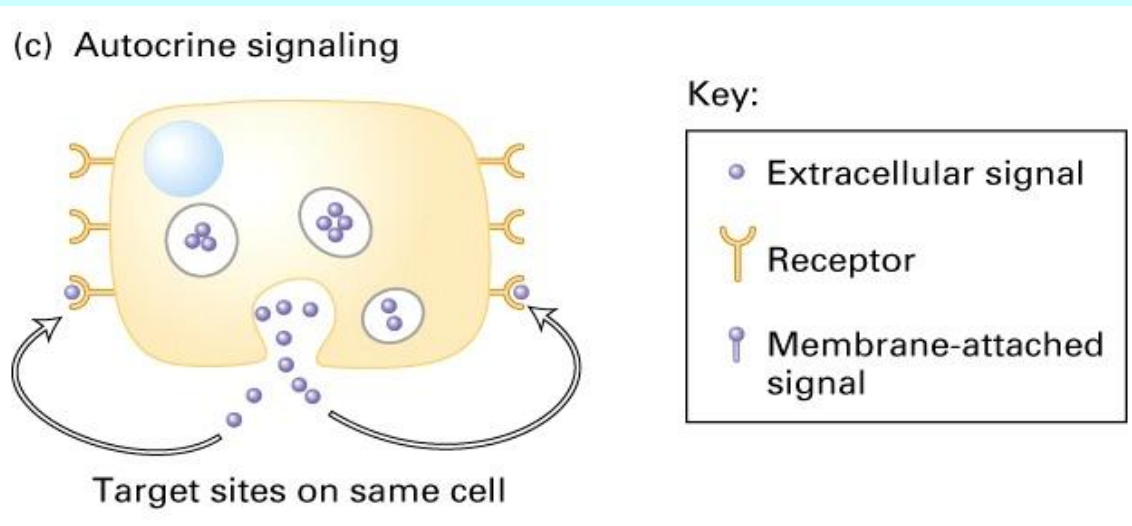
TARGETS DEI FARMACI ATTUALMENTE IN USO



FORME DI SIGNALING CELLULARE

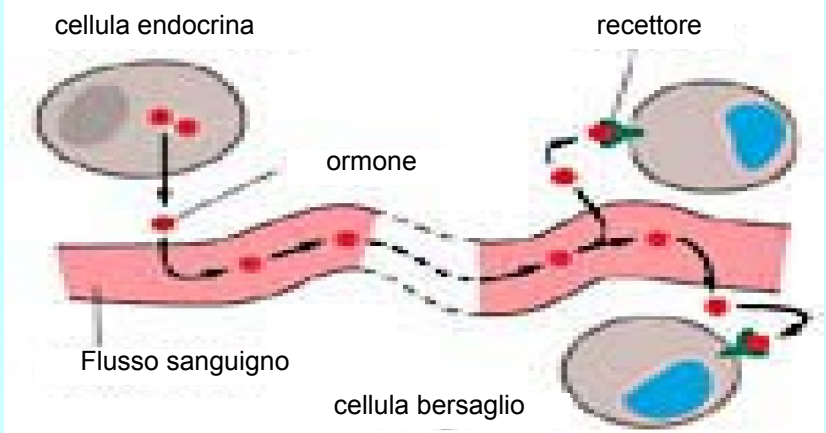




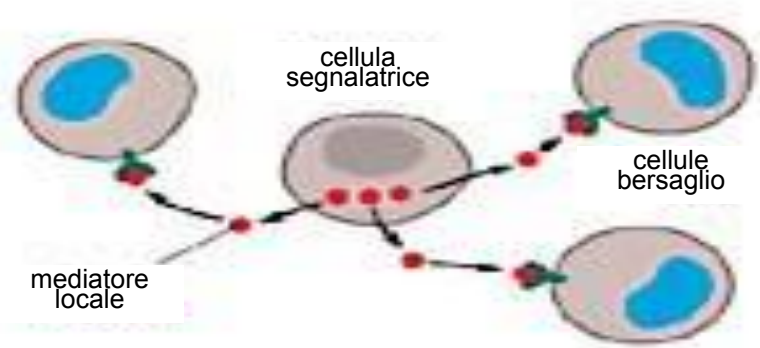


FORME DI SIGNALING CELLULARE

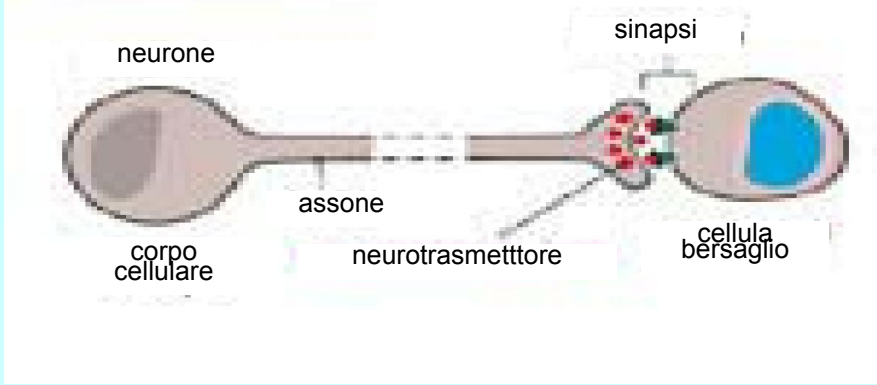
(A) ENDOCRINO



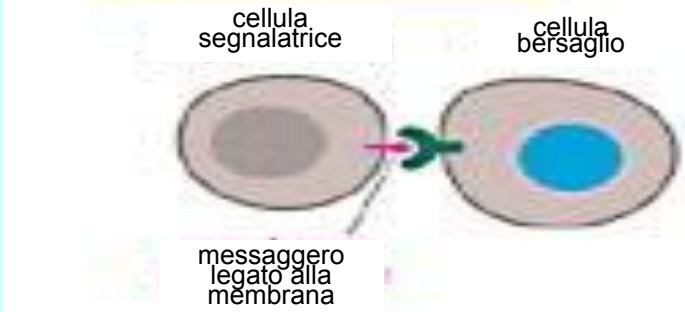
(B) PARACRINO



(C) NEUROCRINO



(D) CONTATTO-DIPENDENTE

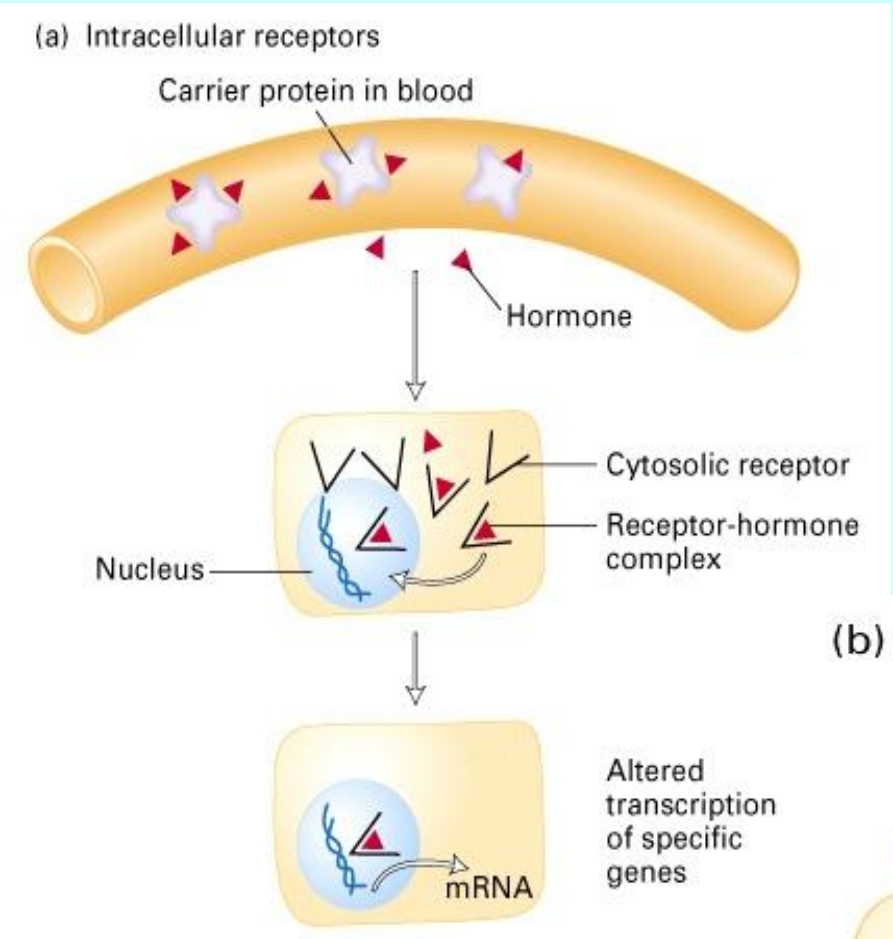


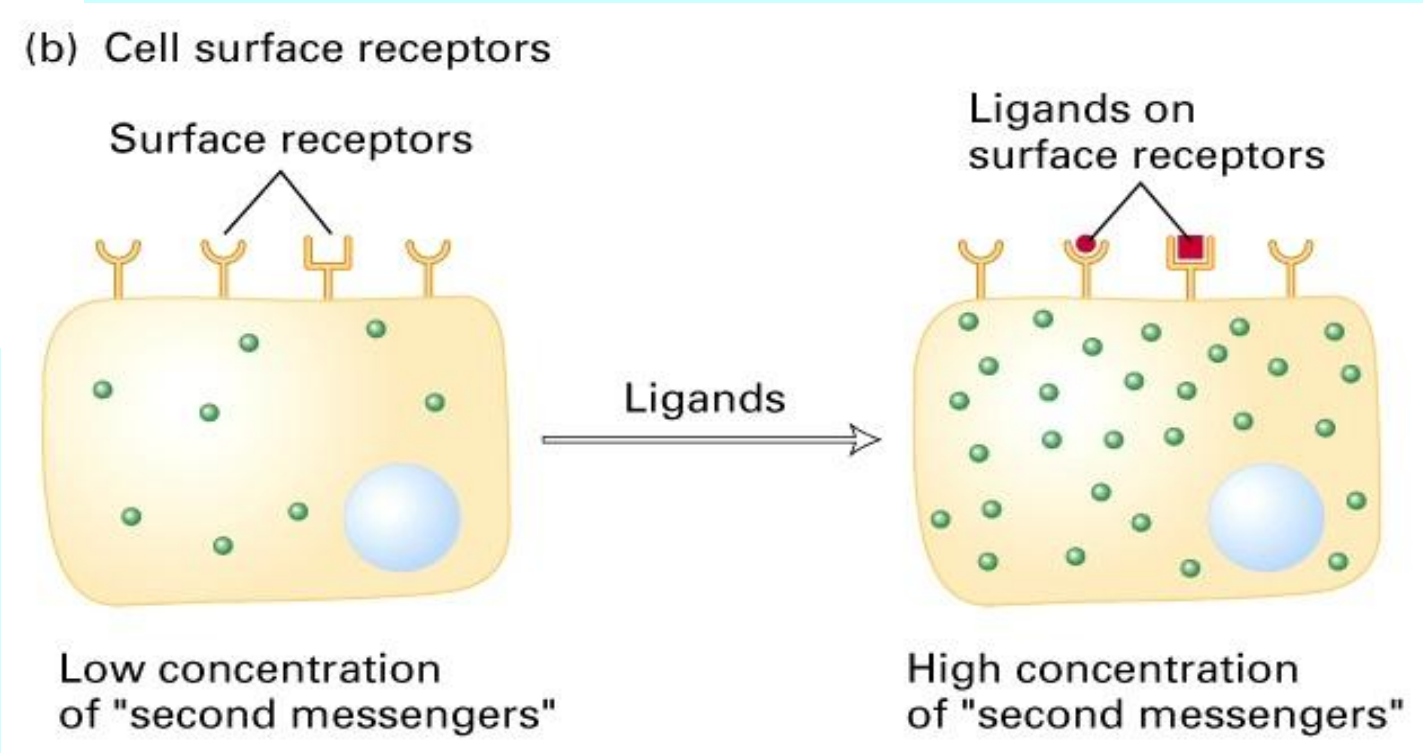
(A) Gli ormoni prodotti dalle ghiandole endocrine sono secreti nel flusso sanguigno e possono essere distribuiti all'intero organismo. (B) I messaggeri paracrini sono rilasciati dalle cellule nel mezzo extracellulare e possono diffondere e agire localmente. (C) I segnali nervosi sono trasmessi lungo gli assoni a cellule bersaglio remote. (D) Il signaling contatto-dipendente (*juxtacrine*) richiede che le cellule siano a contatto l'una con l'altra membrana contro membrana. Le differenze cruciali tra le varie modalità riguardano la velocità e la selettività con cui i segnali sono trasmessi alle cellule bersaglio.

RECETTORI

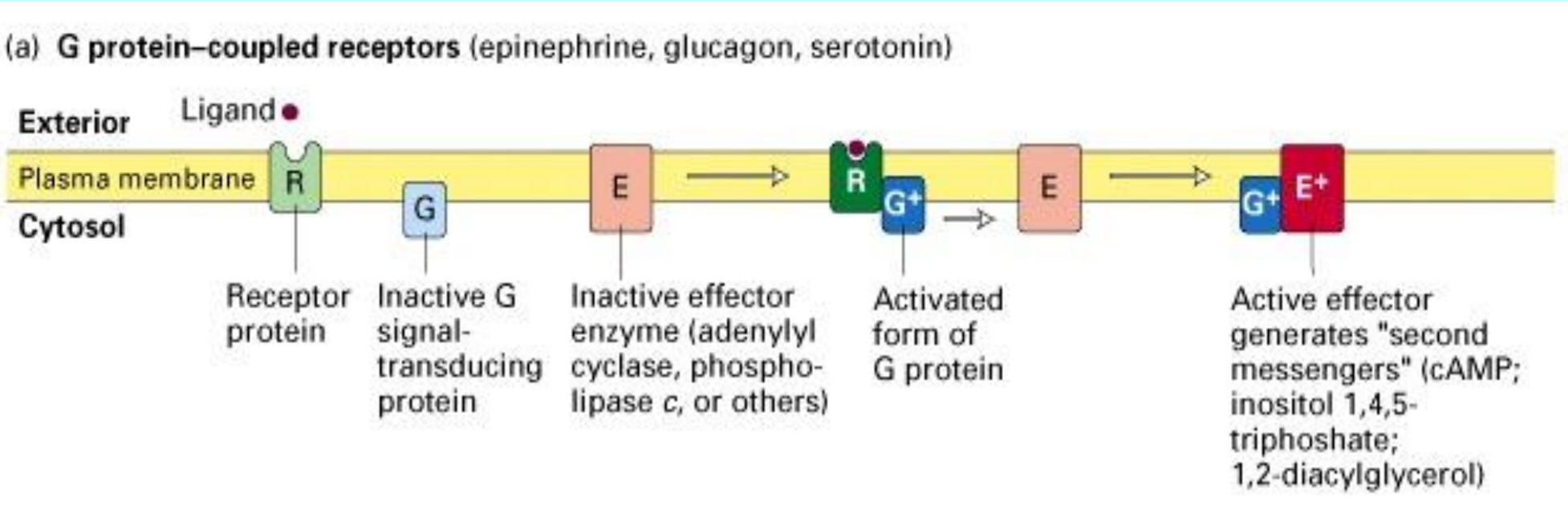
RECETTORI INTRACELLULARI

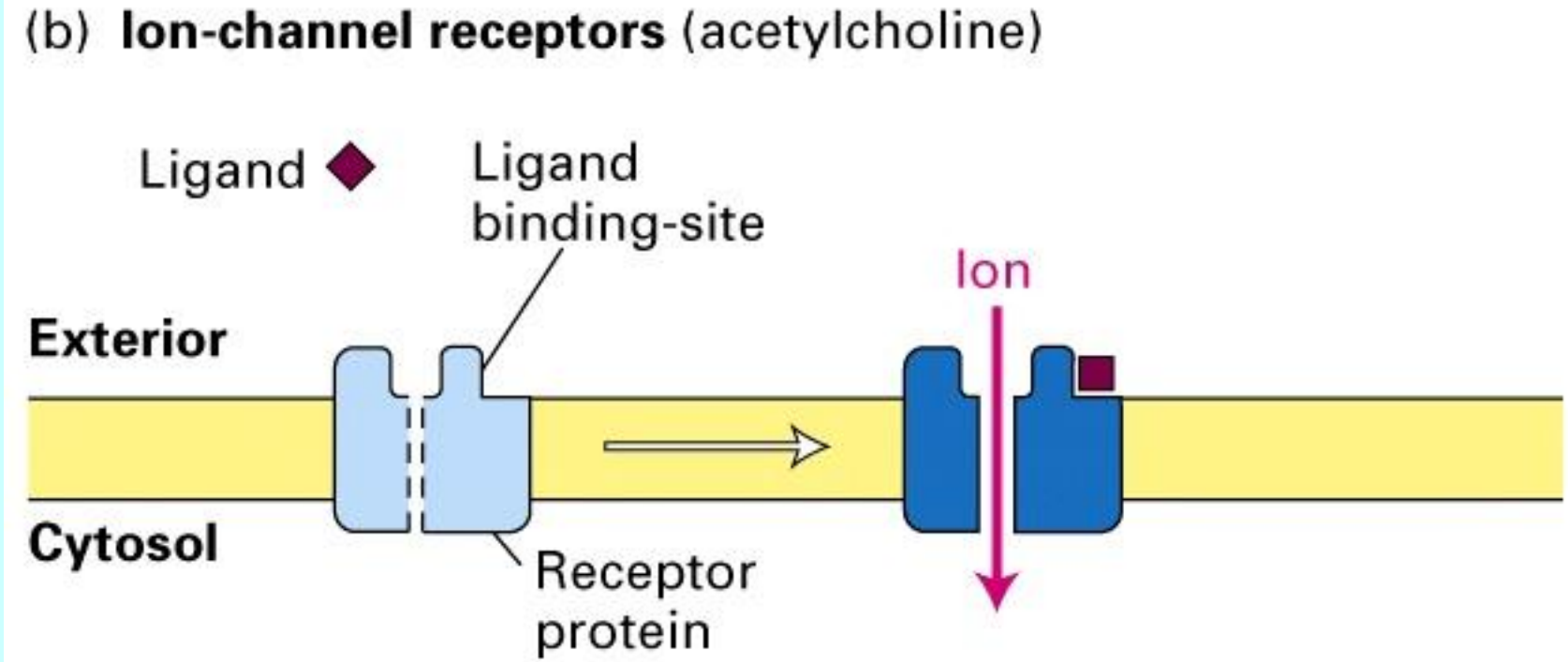
RECETTORI DI MEMBRANA



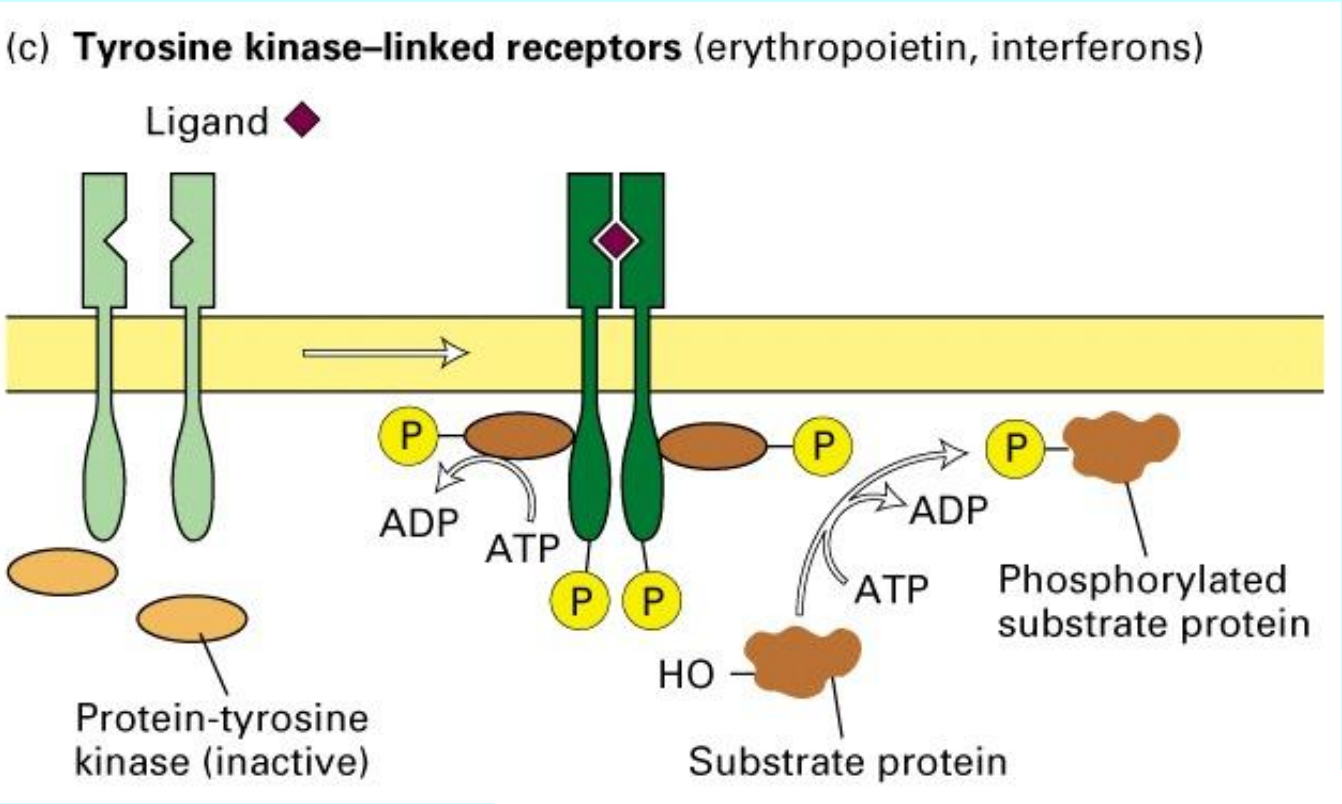


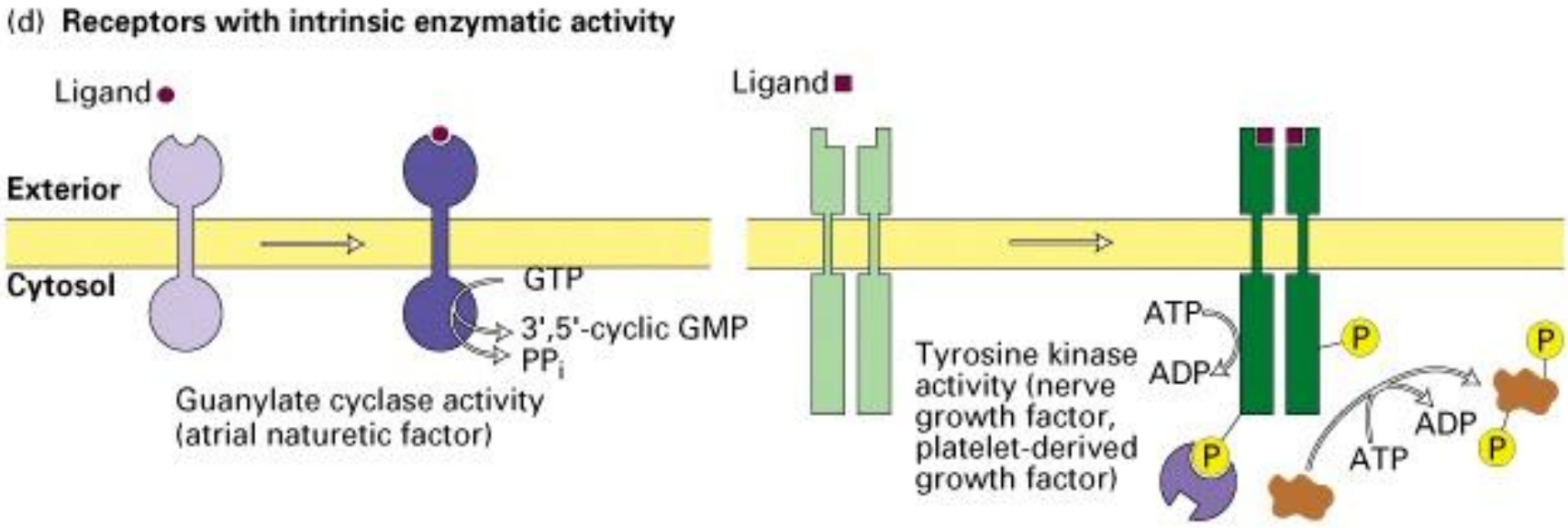
RECETTORI


(a) G protein–coupled receptors (epinephrine, glucagon, serotonin)
Exterior
Ligand
Plasma membrane
Cytosol
R
G
E
R
G+
E
G+
E+
Receptor protein
Inactive G signal-transducing protein
Inactive effector enzyme (adenylyl cyclase, phospho-lipase c, or others)
Activated form of G protein
Active effector generates "second messengers" (cAMP; inositol 1,4,5-triphoshate; 1,2-diacylglycerol)

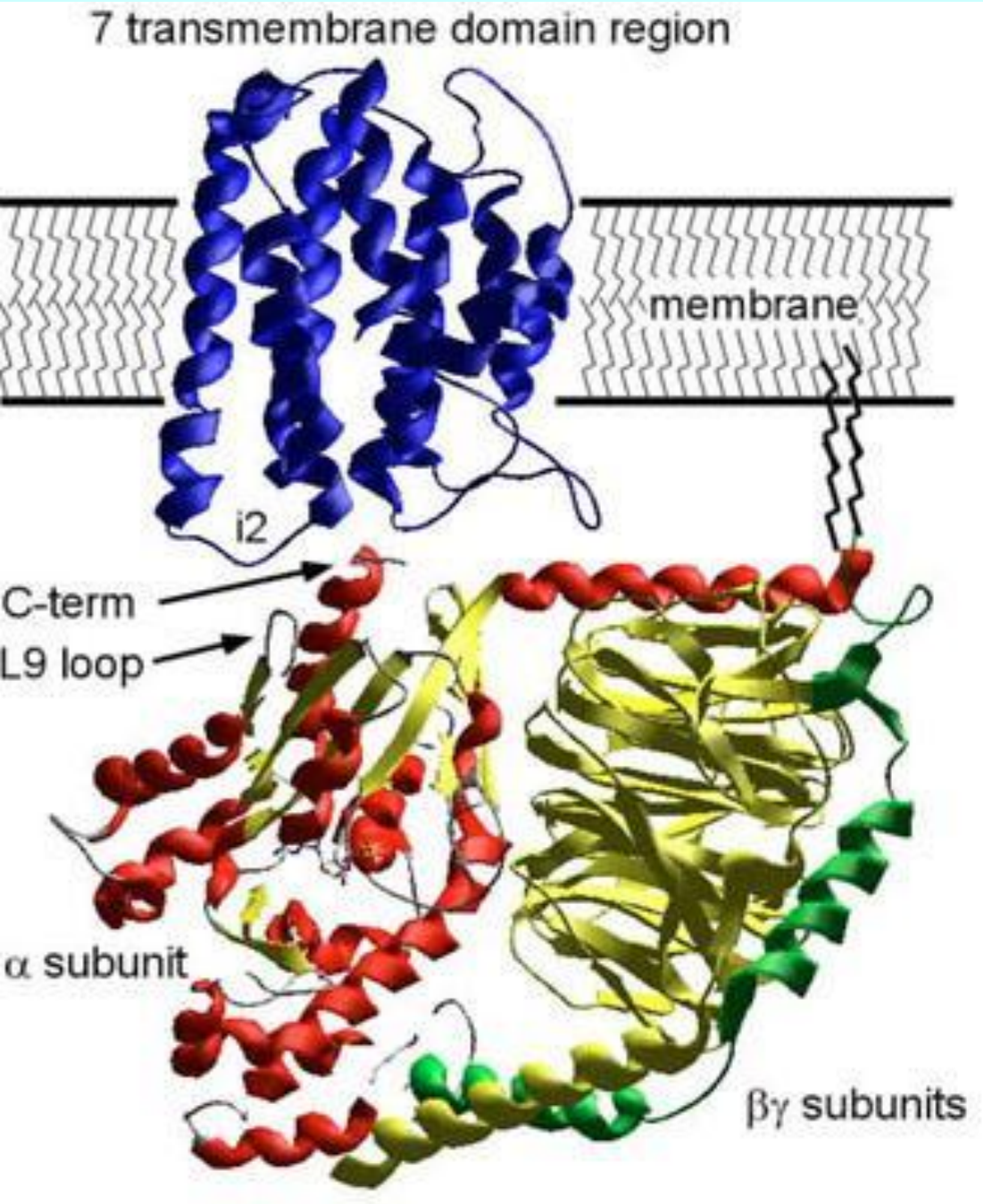

(b) Ion-channel receptors (acetylcholine)
Ligand
Ligand binding-site
Ion
Exterior
Cytosol
Receptor protein

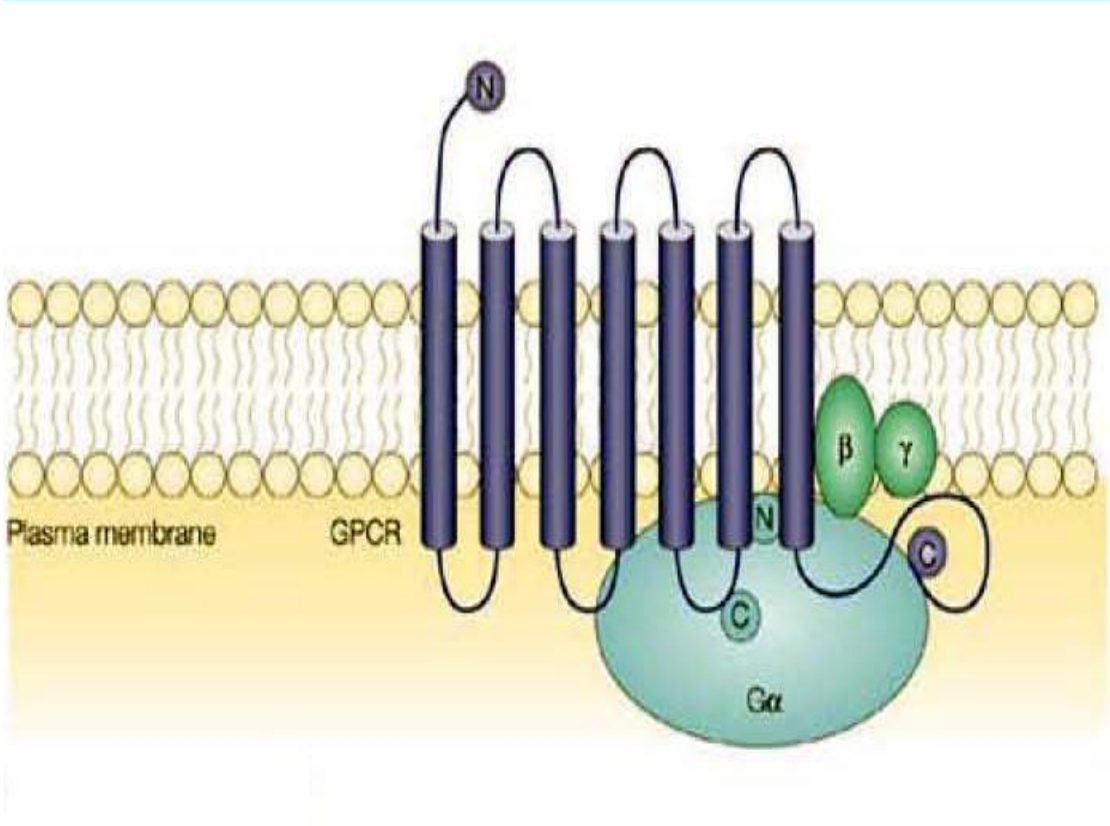
RECETTORI


(c) Tyrosine kinase–linked receptors (erythropoietin, interferons)
Ligand
P
ADP
ATP
P
P
P
ADP
ATP
P
Phosphorylated substrate protein
HO
Protein-tyrosine kinase (inactive)
Substrate protein

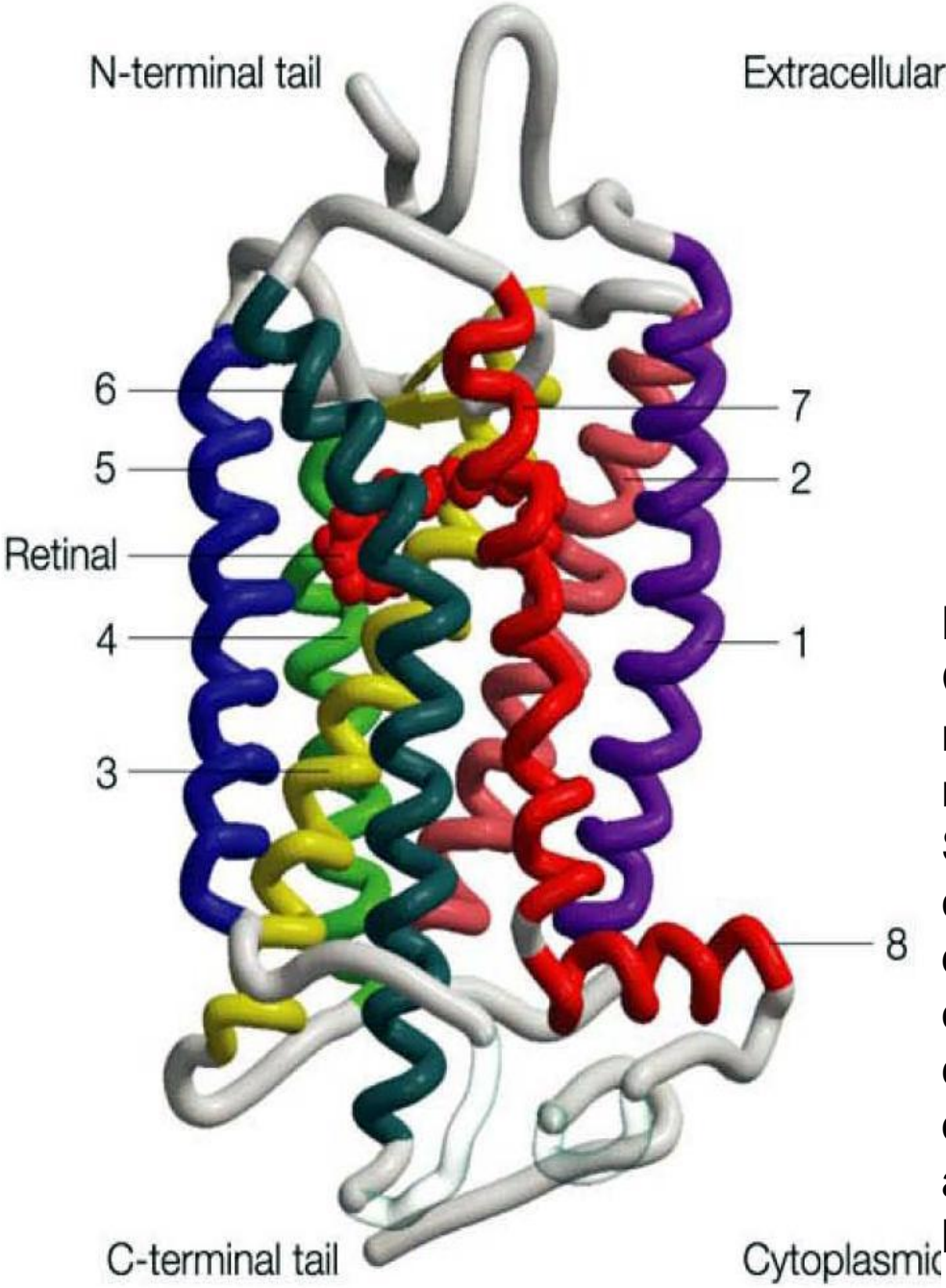

(d) Receptors with intrinsic enzymatic activity
Ligand
Exterior
Cytosol
GTP
3',5'-cyclic GMP
PP_i
Guanylate cyclase activity (atrial naturetic factor)
Ligand
ATP
ADP
P
P
Tyrosine kinase activity (nerve growth factor, platelet-derived growth factor)
ADP
ATP
P
P

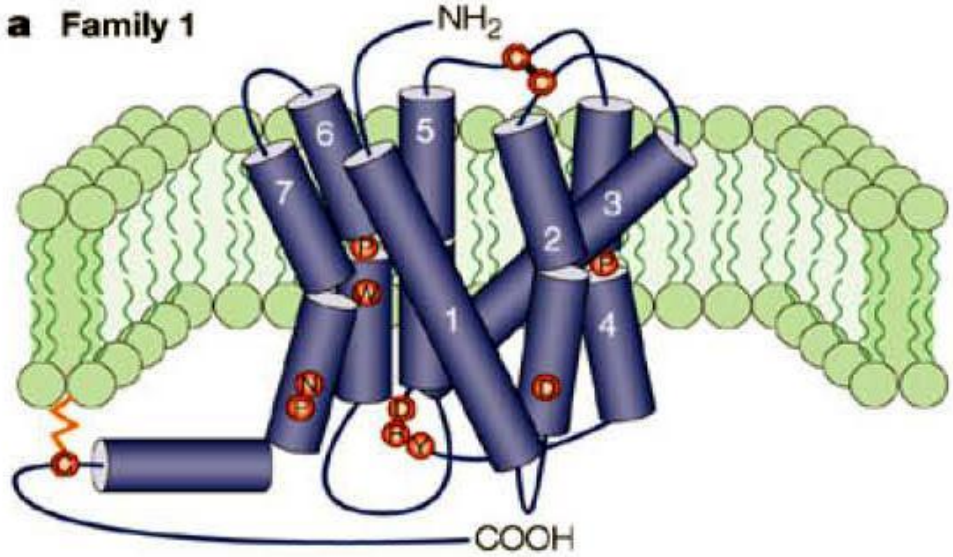
RECETTORI GPCR





RECETTORI 7TM

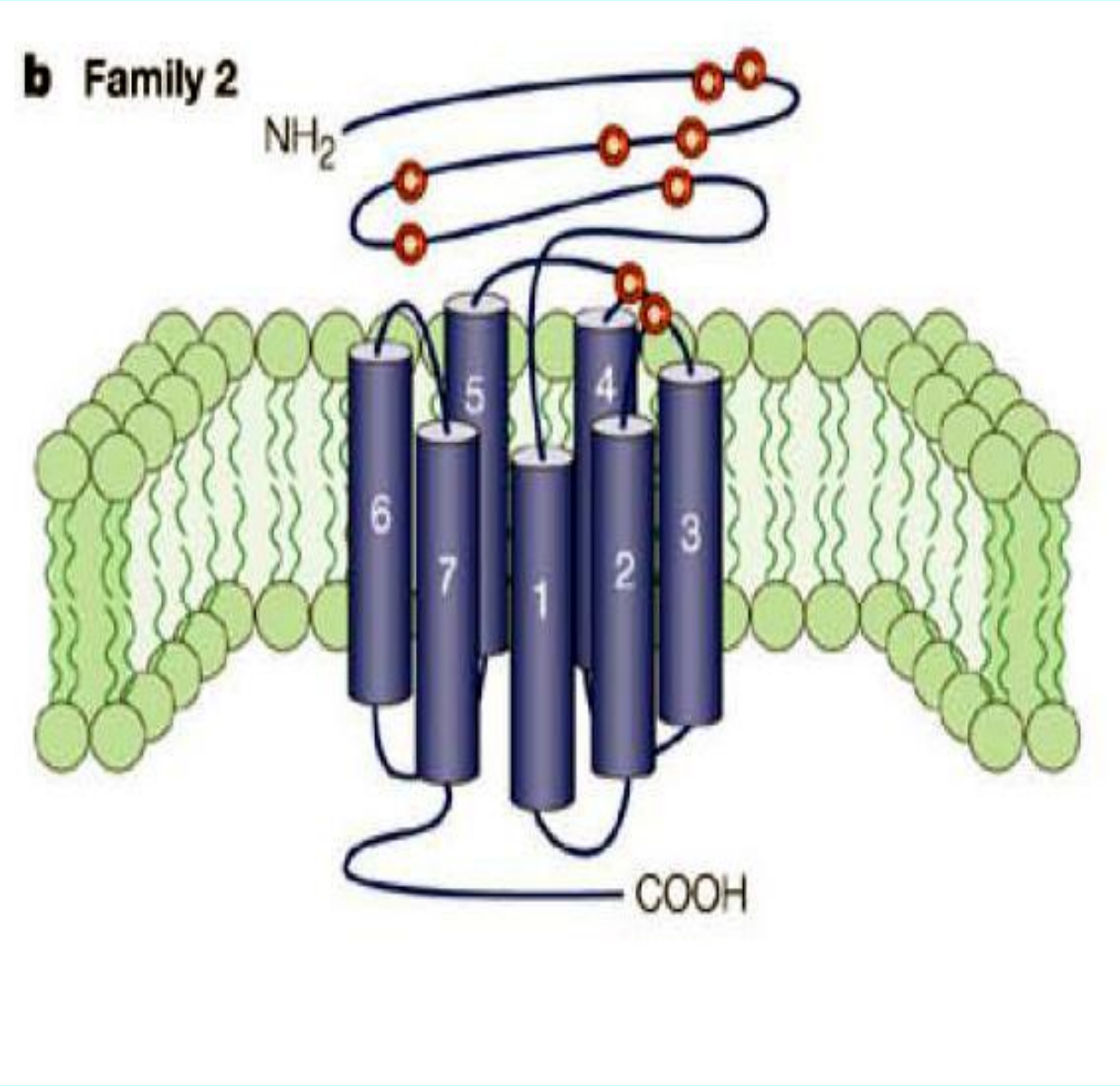




La Famiglia 1 (Rhodopsin-like) è il sottogruppo maggiore. Comprende recettori per odori, importanti neurotrasmettitori (dopamina e serotonina), e anche neuropeptidi e ormoni.

Sono caratterizzati da molti aminoacidi altamente conservati (indicati in figura con cerchi rossi) e un ponte disolfuro che connette il primo e il secondo loop extracellulari. Molti di questi recettori hanno anche una cisteina palmitoilata sulla coda carbossi-terminale, la quale serve per ancorarsi alla membrana. La presenza di amminoacidi come la prolina nelle α−eliche distorce la loro struttura.

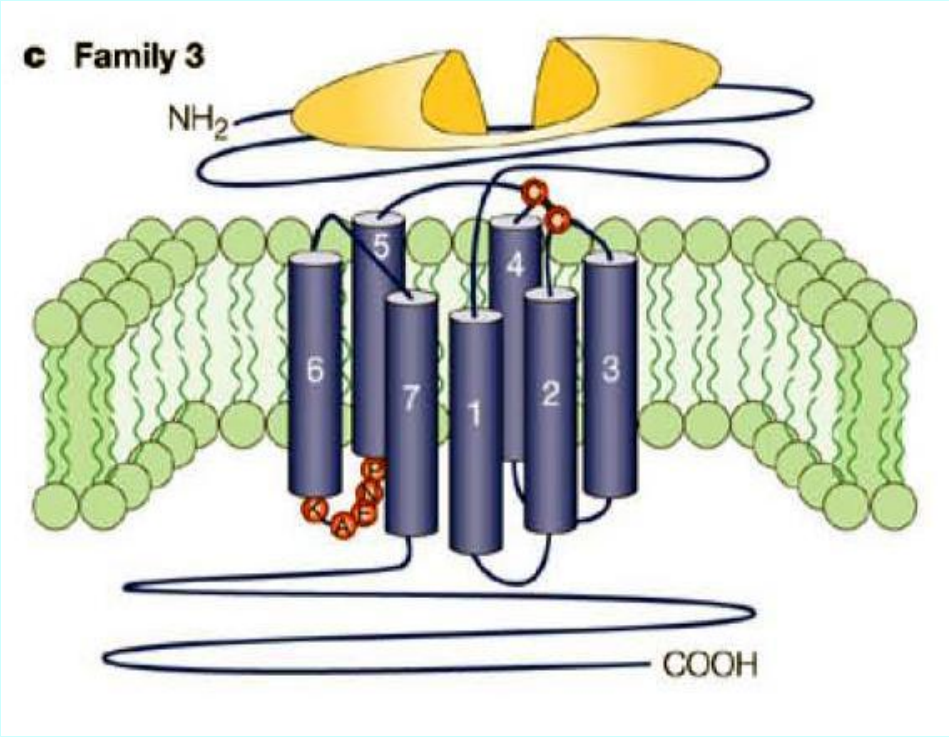
RECETTORI 7TM



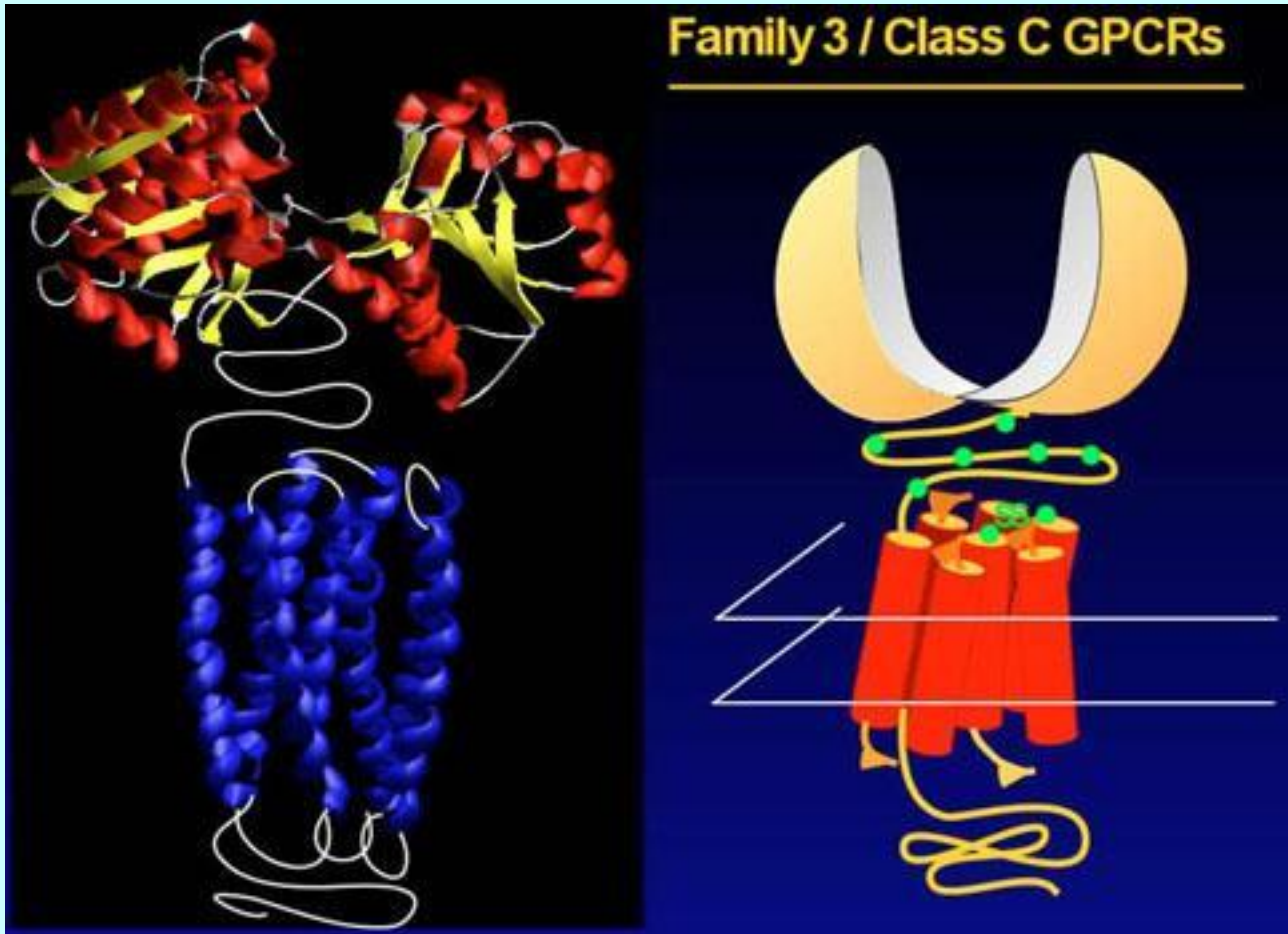
I GPCRs della famiglia 2 o famiglia B (Secretin receptor family) sono caratterizzati da un amino terminale relativamente lungo, che contiene molte cisteine che formano un network di ponti disolfuro. La loro morfologia è simile ad alcuni recettori della famiglia 1, ma il sito di palmitoilazione manca e residui e motivi conservati sono differenti da quelli della famiglia 1.

Si sa poco sull'orientazione dei domini TM, che è probabilmente differente da quella dei recettori della famiglia 1. I ligandi per i GPCRs della famiglia 2 includono ormoni, come glucagone, secretina e ormone paratiroide.

RECETTORI 7TM

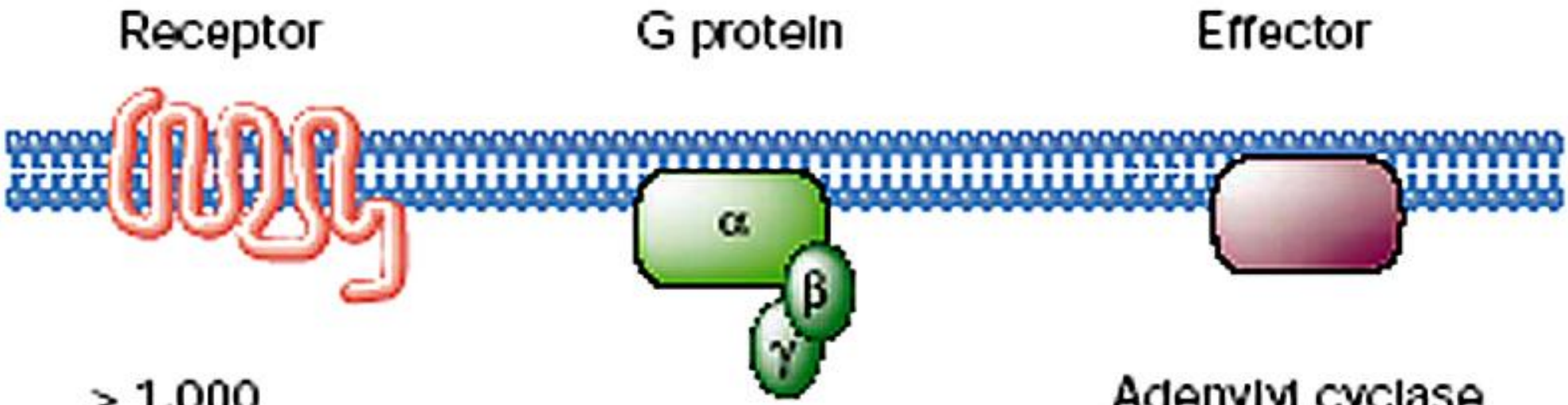


La famiglia 3 (Metabotropic glutamate/pheromone) comprende i recettori metabotropici del glutammato, i recettori Ca^{2+}-sensibili e i $GABA_B$ (acido γ-amminobutirrico, tipo B). Questi recettori sono caratterizzati da lunghi tratti ammino terminali e lunghe code carbossiliche. Il dominio di legame è collocato sul tratto ammino terminale e forma un dimero collegato con un ponte disolfuro.



Eccetto che per due cisteine nei loop extracellulari 1 e 2 i recettori della famiglia 3 non hanno alcuna caratteristica delle famiglie 1 e 2.
Una caratteristica unica di questi recettori è che il terzo loop intracellulare è corto e altamente conservato. Ad oggi, si sa poco sull'orientazione dei domini TM.

RECETTORI GPCR



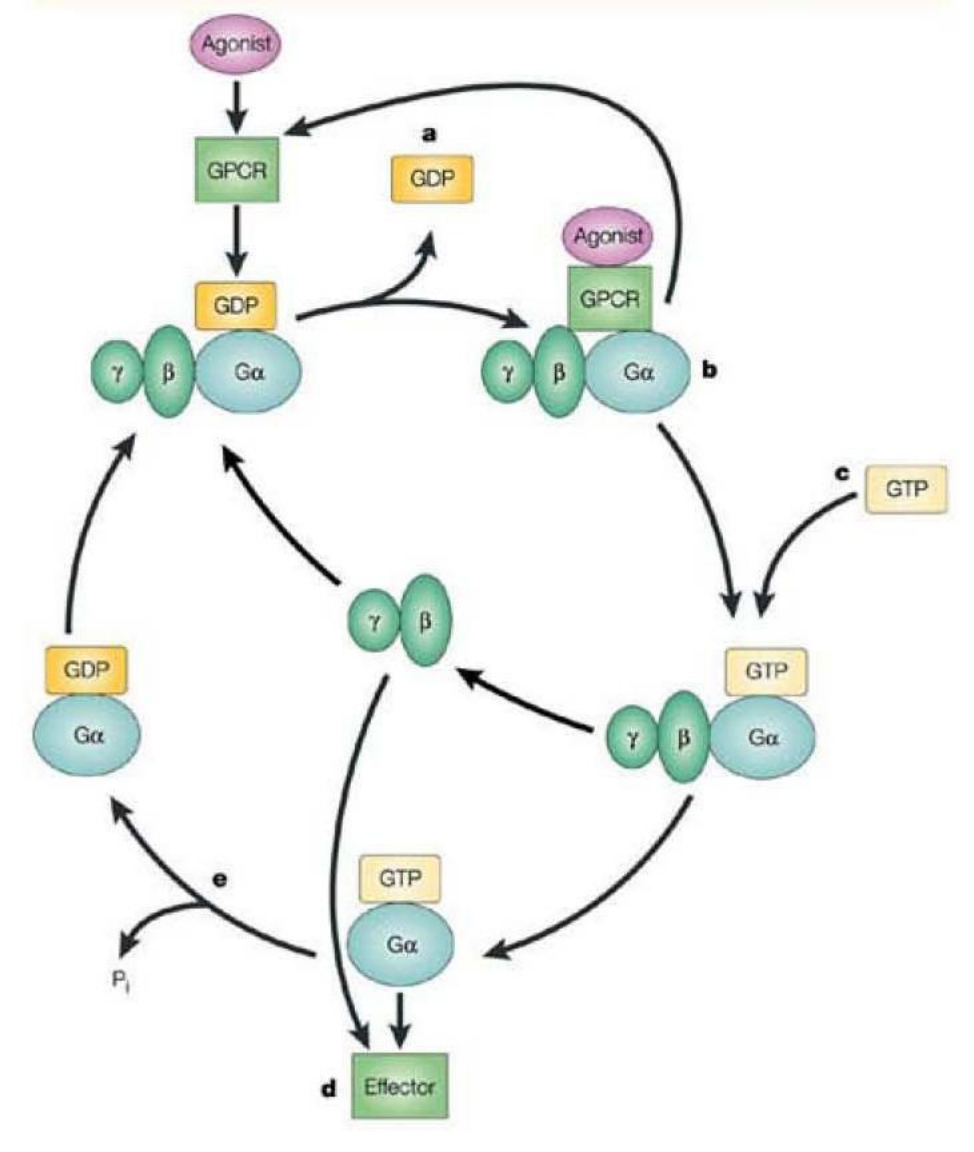
> 1,000
Adrenergic
Dopaminergic
Opioid
Sensory – rhodopsin, olfactory, taste
Glycoprotein hormones
Lipids and other small molecules

15 α-subunits
6 β-subunits
12 γ-subunits

Adenylyl cyclase
Phospholipases C & A_2
cGMP phosphodiesterase
Potassium channels
Calcium channels

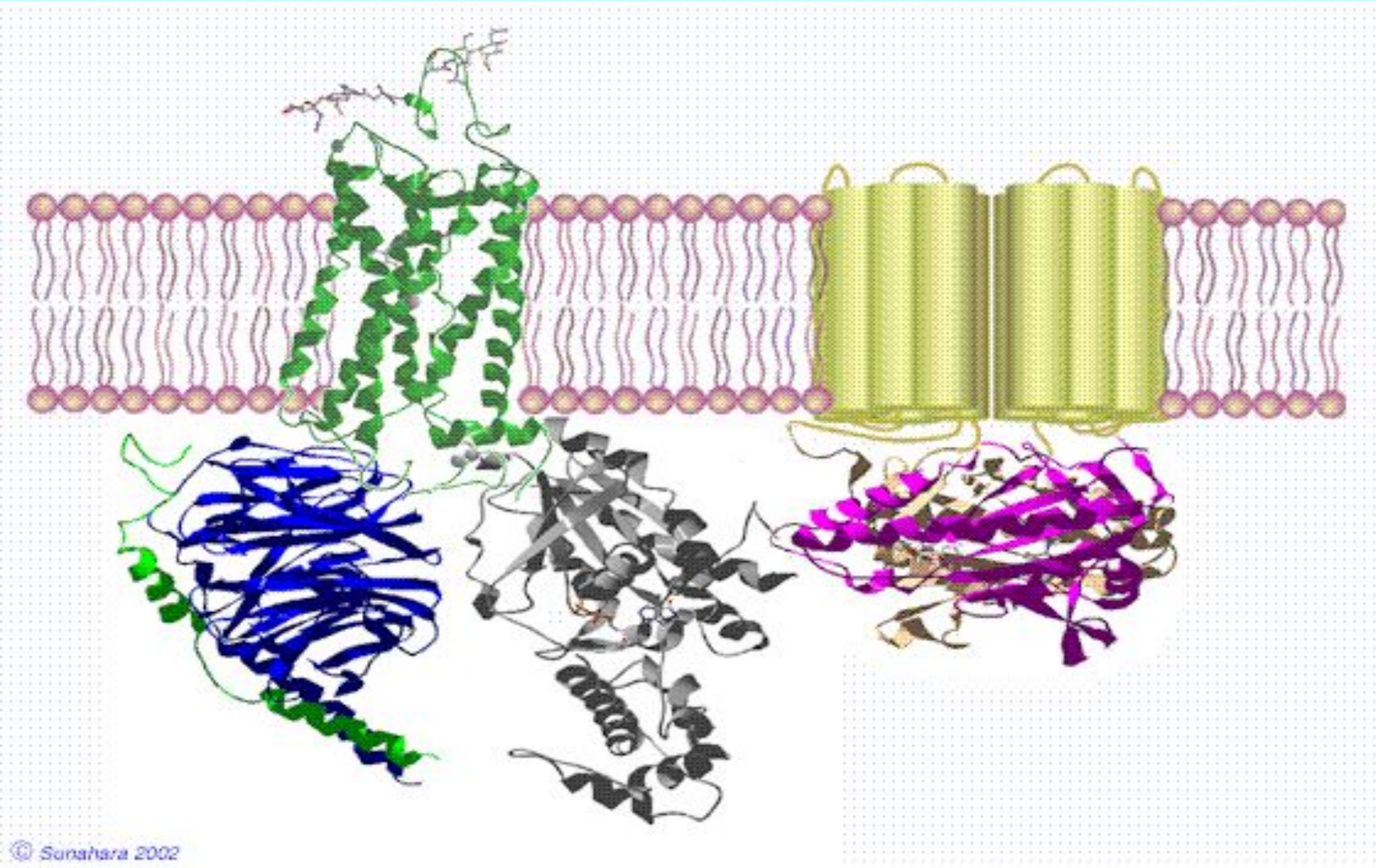
Nature 415, 206-211 (2002)

RECETTORI GPCR

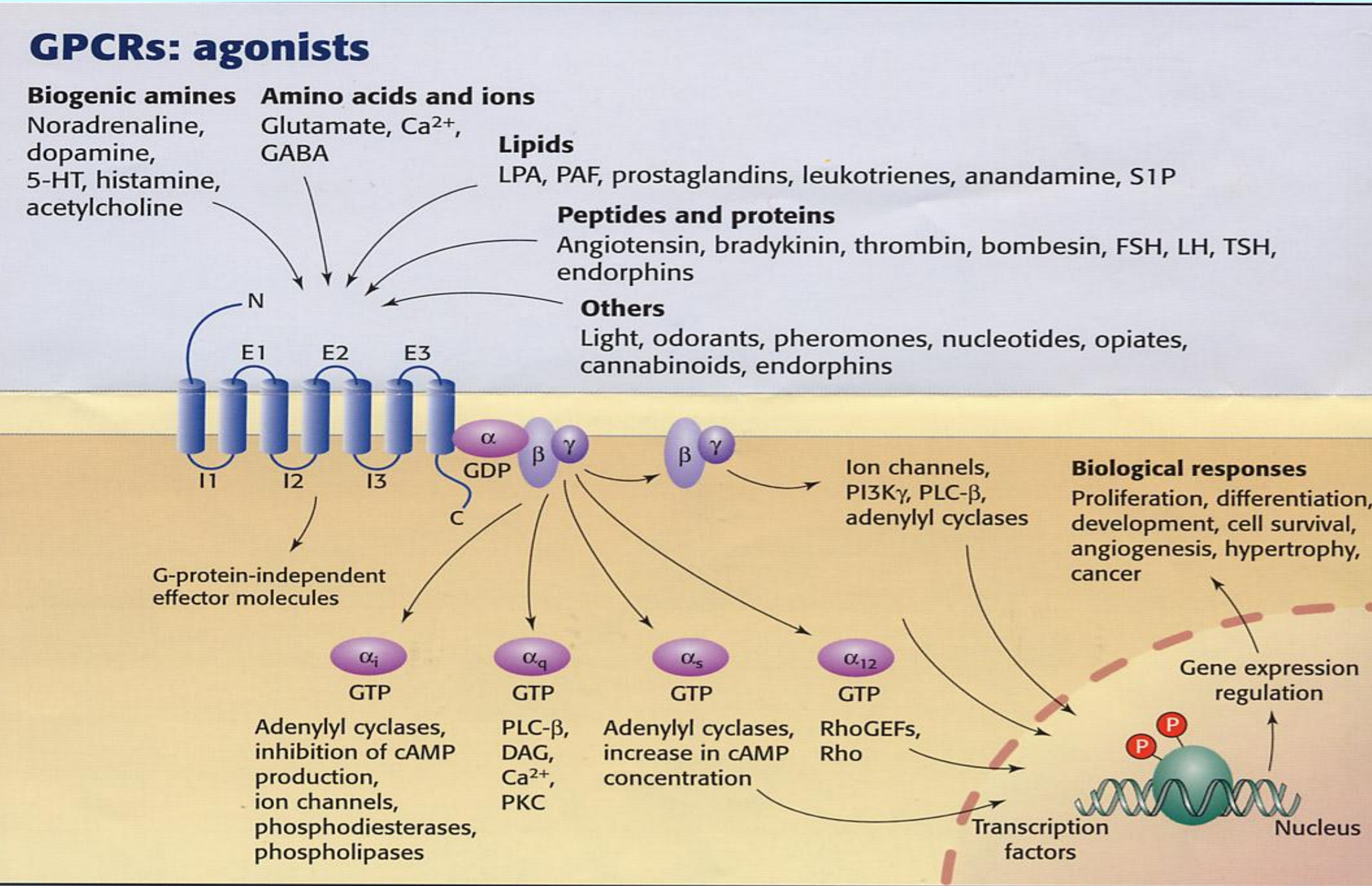


1) Attivazione del Recettore
2) Dissociazione del GDP dalla subunità α e complessazione con GTP
3) Dissociazione delle subunità βγ dalla α
4) Attivazione degli effettori
5) Idrolisi del GTP
6) Riassociazione delle subunità α e βγ

RECETTORI GPCR

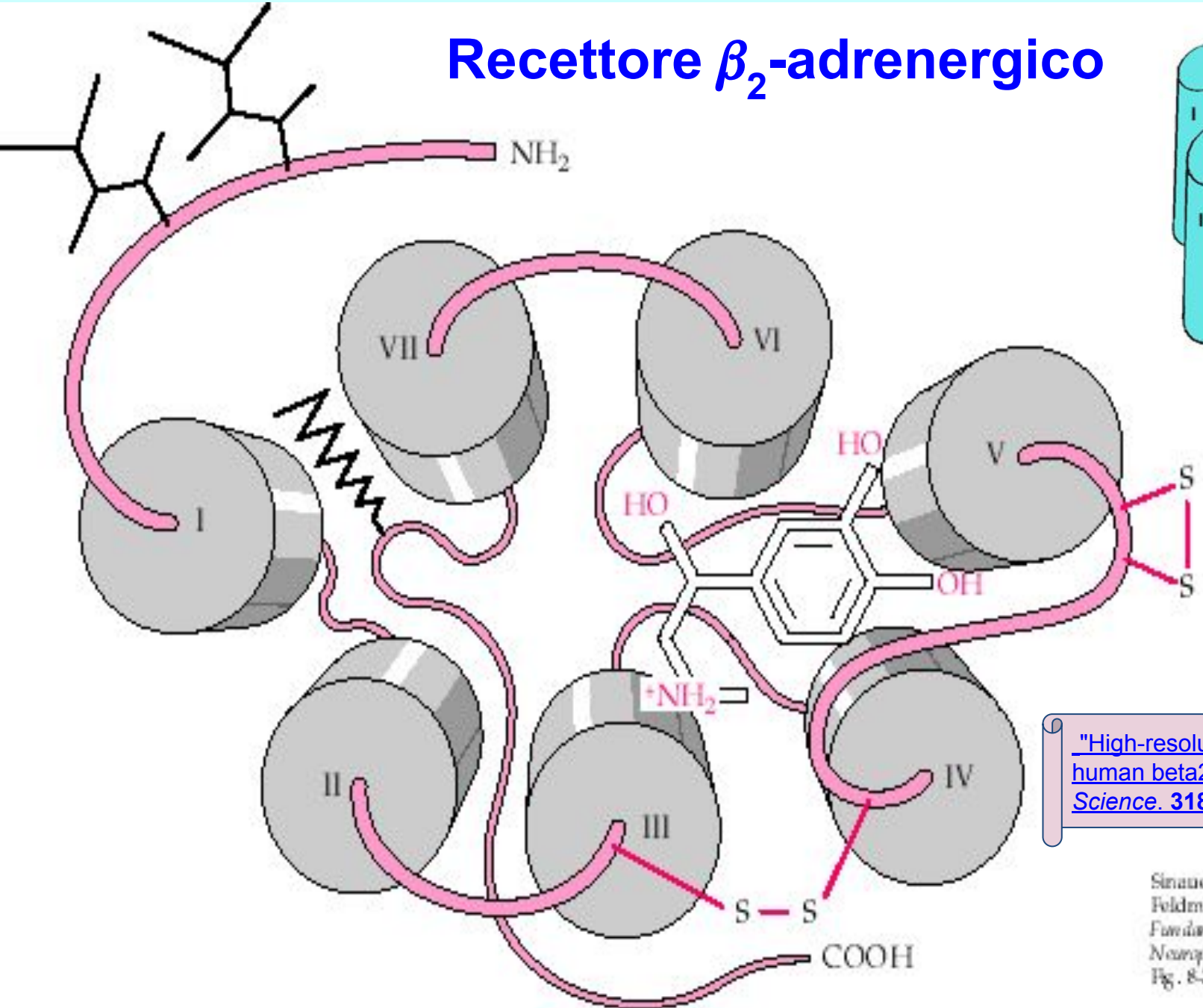


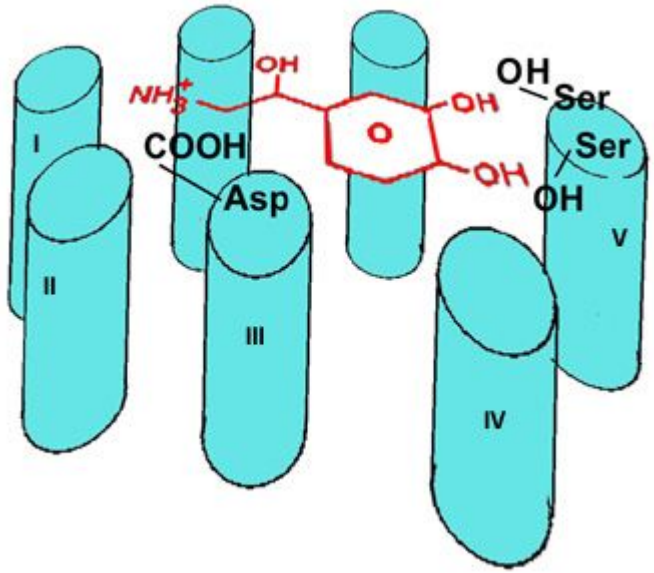
RECETTORI GPCR



RECETTORI GPCR

Recettore β_2-adrenergico





"High-resolution crystal structure of an engineered human beta2-adrenergic G protein-coupled receptor". *Science*. **318** (5854): 1258–65.

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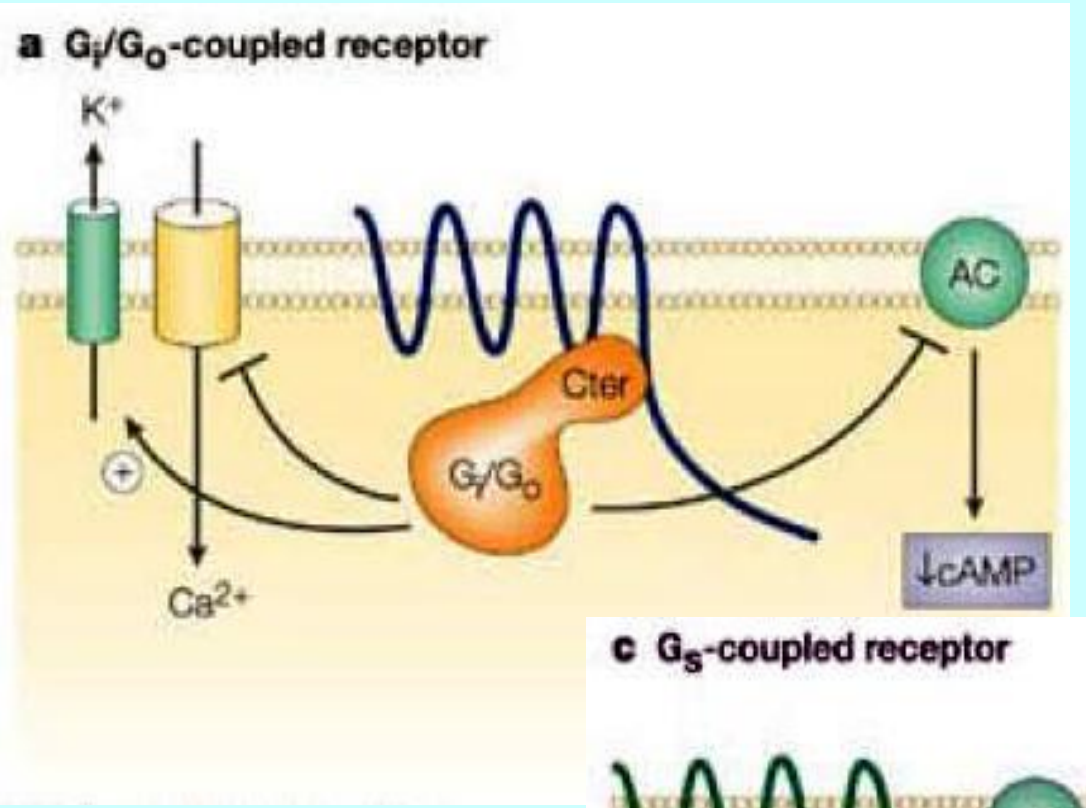
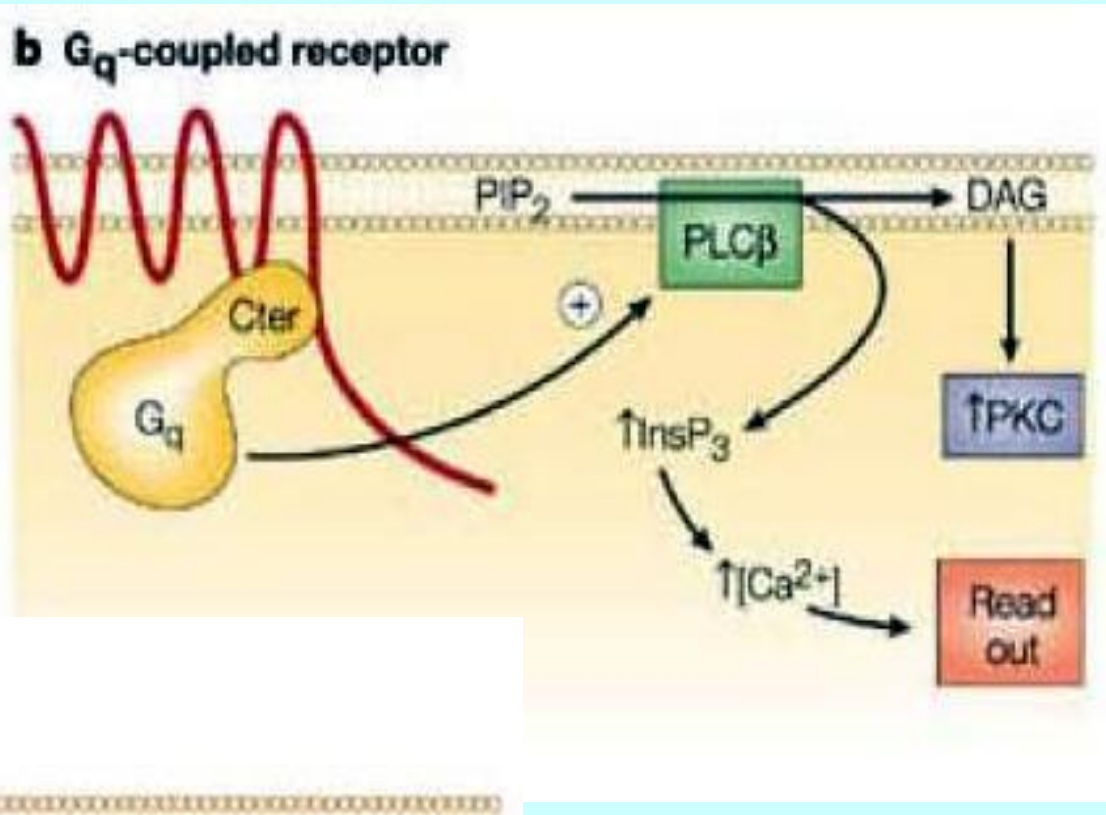
Feldman
Fundamentals of
Neuropsychopharmacology
Fig. 8-33

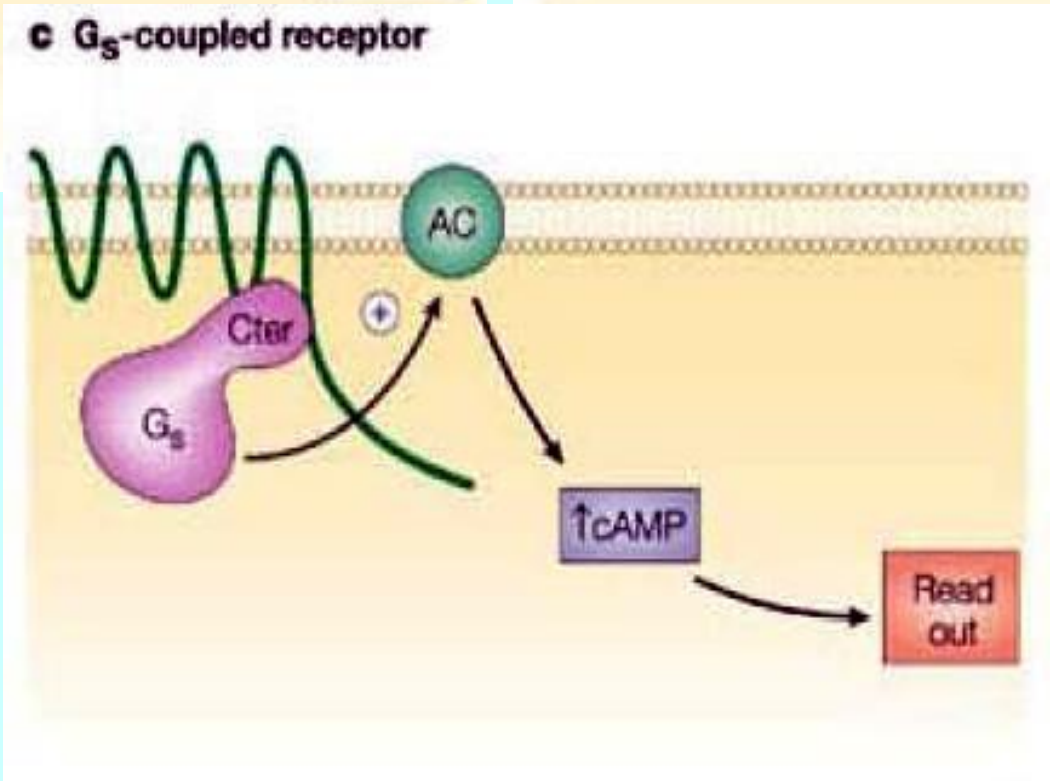
Classe G-protein	Subunità G-protein	Effettori	Regolazione dell'effettore
G_s	$G\alpha_s$	AC1-9	Stimolazione
		c-Src	Stimolazione
		RGS-PX1 (GAP)	Stimolazione
		Tubulina	Stimolazione di attività GTPase
	G_{olf}	ACS	Stimolazione
G_i	$G\alpha_{tr}$	PDE6	Stimolazione
	$G\alpha_{tc}$	PDE6	Stimolazione
	$G\alpha_g$	PDE	Stimolazione
	$G\alpha_{si1-3}$	AC5, AC6	Inibizione
	$G\alpha_o$	c-Src	Stimolazione
		RAP1GAPII	Sequestro di RAP1GAP
		Canali del Ca^{2+}	Inibizione ($G\alpha_o$)
		Canali del K^+ (GIRK)	Stimolazione ($G\alpha_i$)
		Tubulina	Stimolazione di attività GTPase
		GRIN1 e GRIN2	Sconosciuta
	Gβγ	PI3Kγ, PI3Kβ	Stimolazione
		AC2, AC4, AC7	Stimolazione
		AC1	Inibizione
		PLCβ1−3	Stimolazione
		GRK2, GRK3	Reclutamento alla membrana plasmatica
		GIRK1, GIRK2 e GIRK4	Stimolazione
		Canali K^+ ATP-sensitive	Stimolazione
		Canali del Ca^{2+}	Inibizione
		Small GTP binding proteins	Reclutamento alla membrana plasmatica

Classe G-protein	Subunità G-protein	Effettori	Reg.dell'effettore
	Gβγ	(Rho e Rac) Tirosina kinase di Bruton	Stimolazione
		Tsk tirosina kinase	Stimolazione
		Proteina kinase D	Stimolazione
		Calmodulina	Inibizione calmodulina kinase
		Tubulina	Aumentata attività GTPase
		Dinamina I	Aumentata attività GTPase
		Shc	Stimolazione (attivazione indiretta di MAPK)
		Raf1 proteina kinase	Sequestro di Gβγ
		Ras guanina nucleotide releasing factor/CDC25Mm	Stimolazione (attivazione indiretta di Ras)
		KSR-1	Sequestro di Gβγ
G_q	$G\alpha_{q, 11, 14, 15/16}$	PLCβ1−4	Stimolazione
		LARG-RhoGEF	Stimolazione (attivazione indiretta di Rho)
		GRK2, GRK3	Stimolazione
		Tirosina kinase di Bruton	Stimolazione (stimolazione indiretta di p38 MAPK)
G_h (TGII) Small G-proteins	$G\alpha_{12}$ $G\alpha_{13}$	P115-RhoGEF, PDZ-RhoGEF, LARG-RhoGEF	Stimolazione (attivazione indiretta di Rho)
		GAP1m	Stimolazione (attivazione indiretta di Ras)
		E-caderina	Stimolazione (stimolazione del rilascio di β catenina)
	$G\alpha_h$ Arf RhoA	PLCδ1	Stimolazione
		PLD1 e/o PI4P 5 kinase	Stimolazione

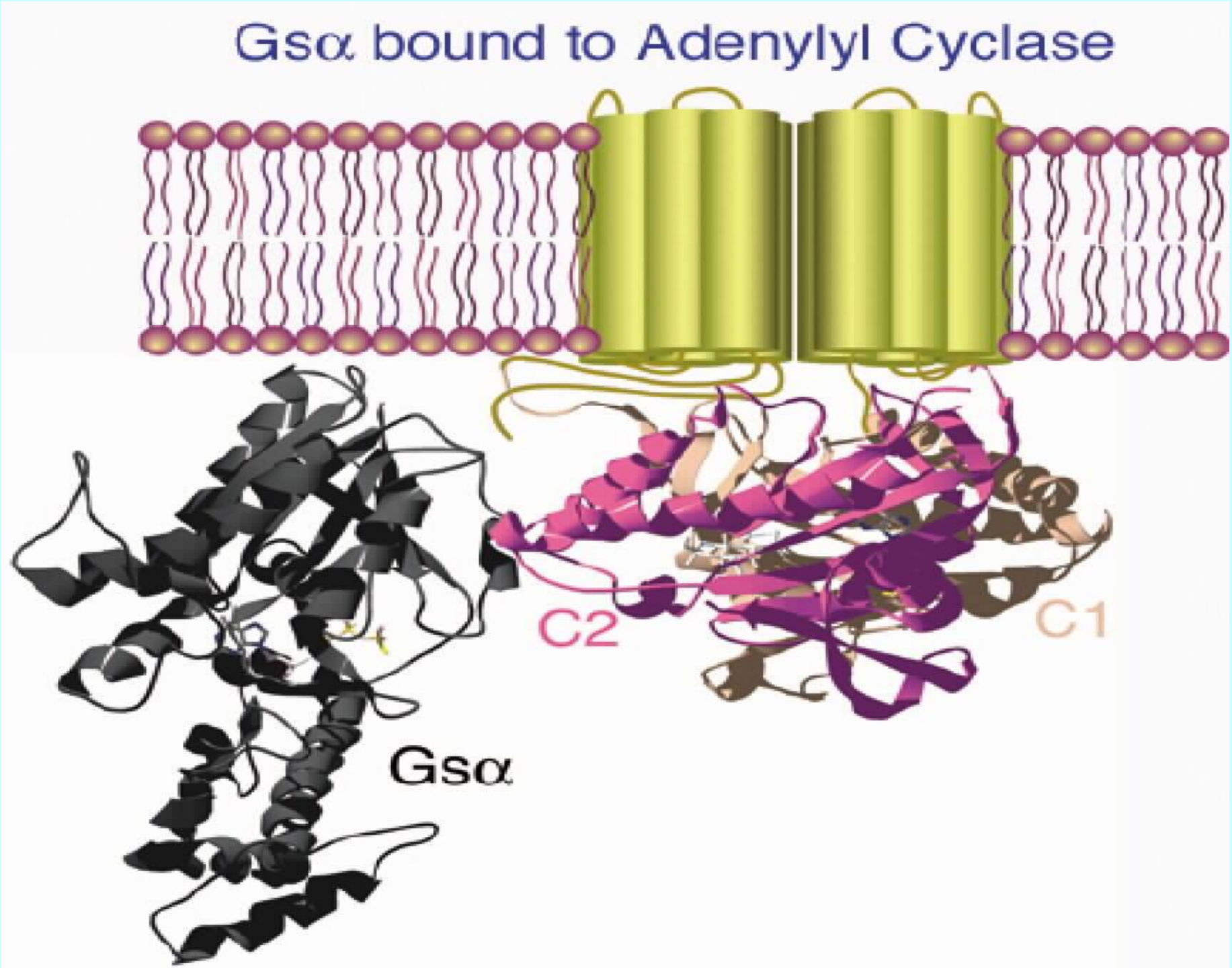
SUBUNITÀ α DI GPCR



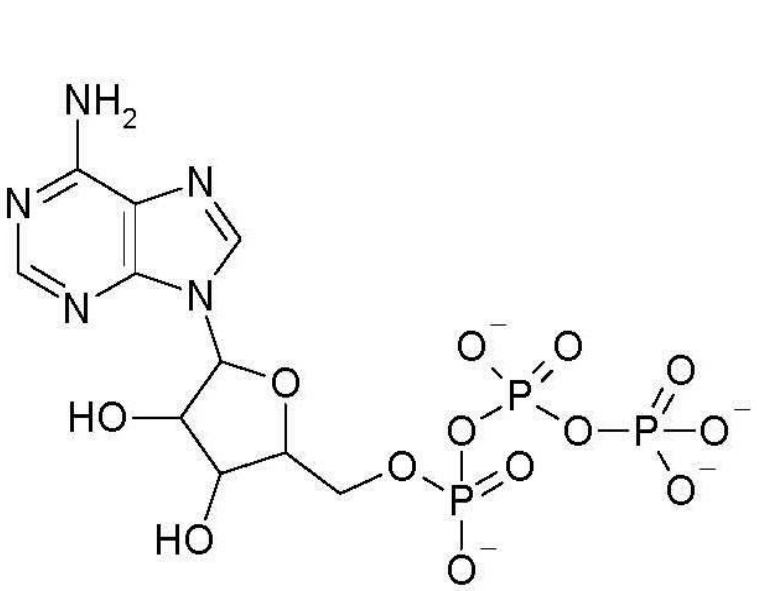




EFFETTORI – ADENILATO CICLASE

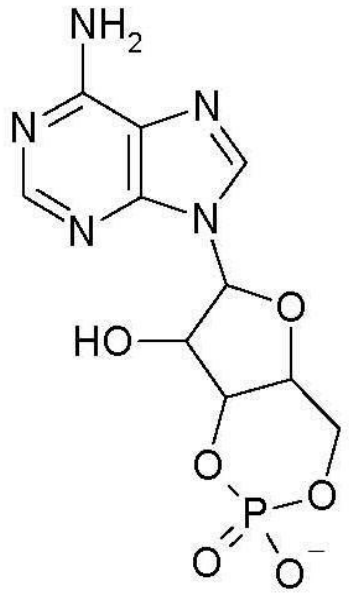


EFFETTORI – ADENILATO CICLASE

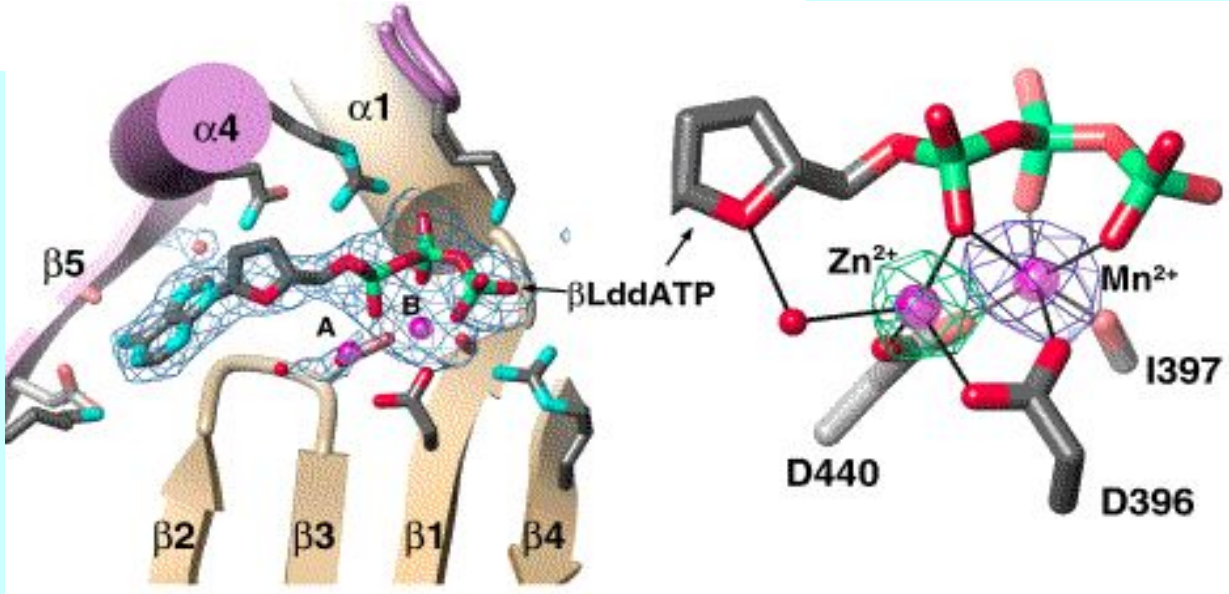


Adenosina trifosfato (ATP)

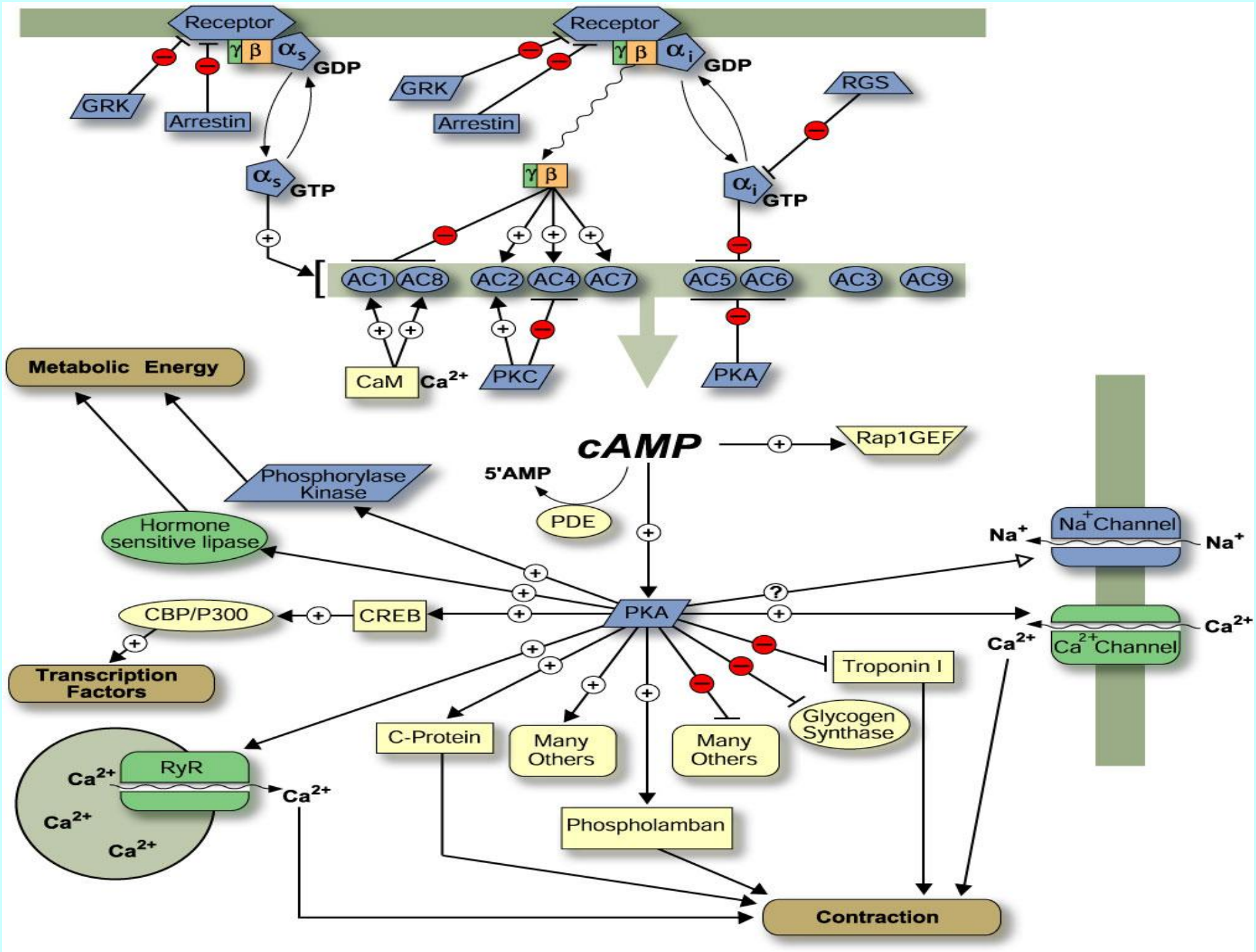
PP



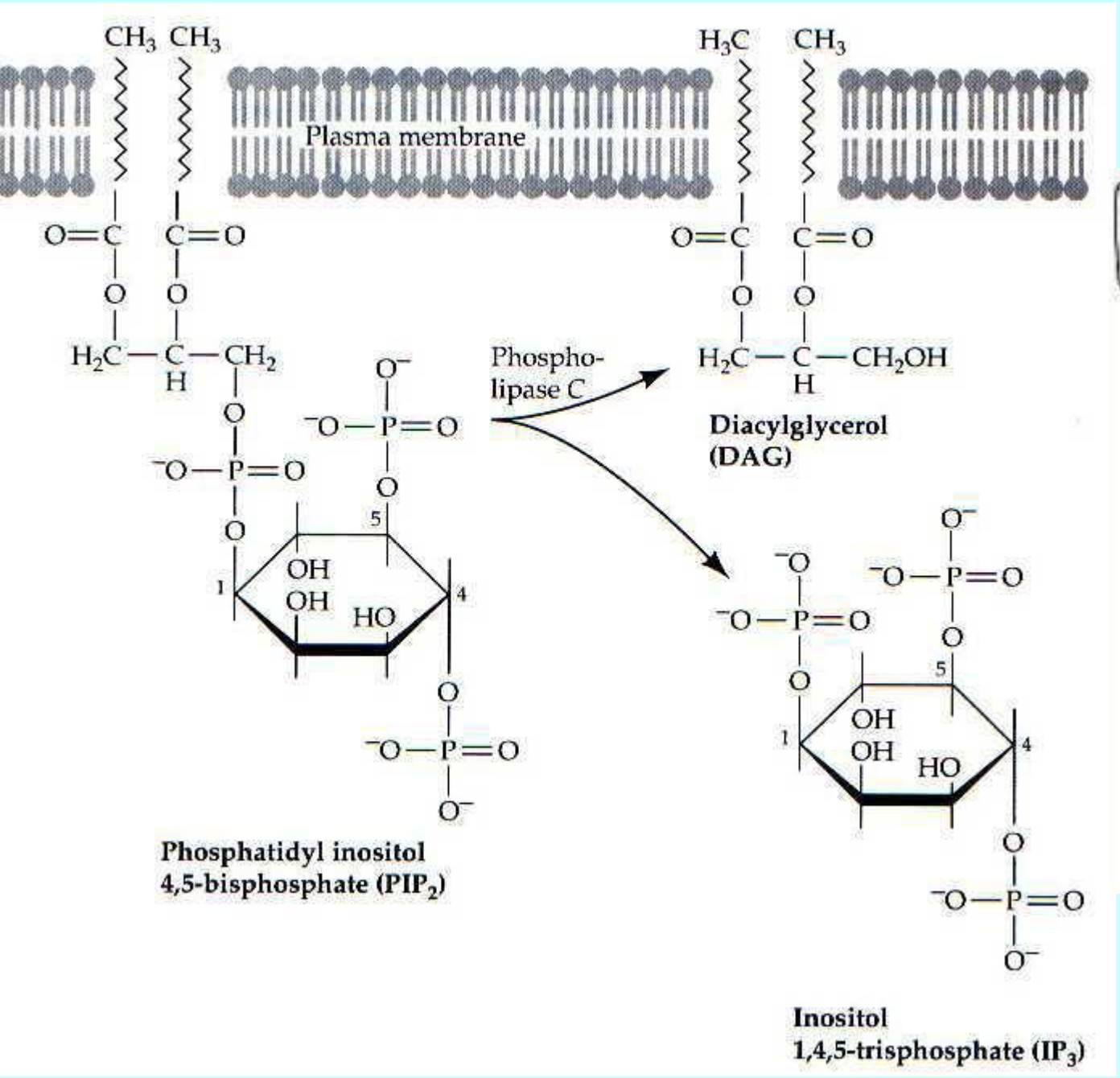
c-AMP

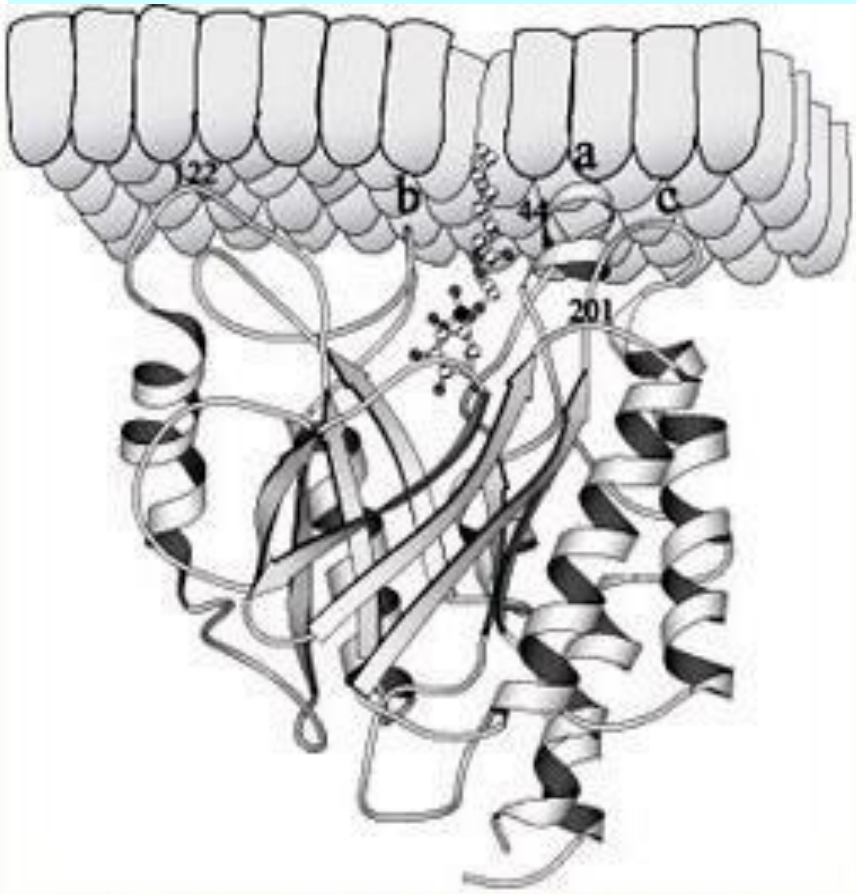


ADENILATO CICLASE

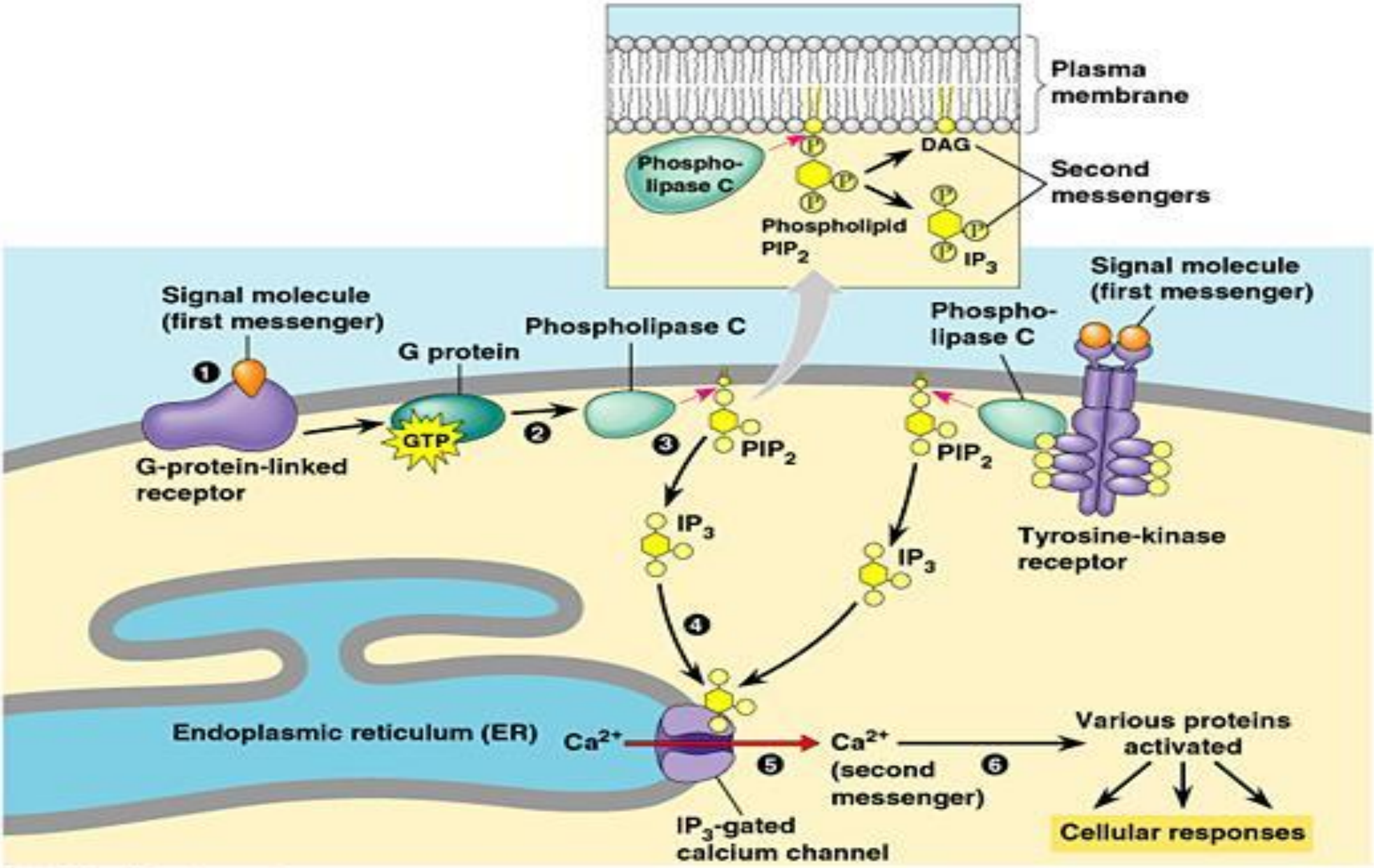


EFFETTORI – PLC

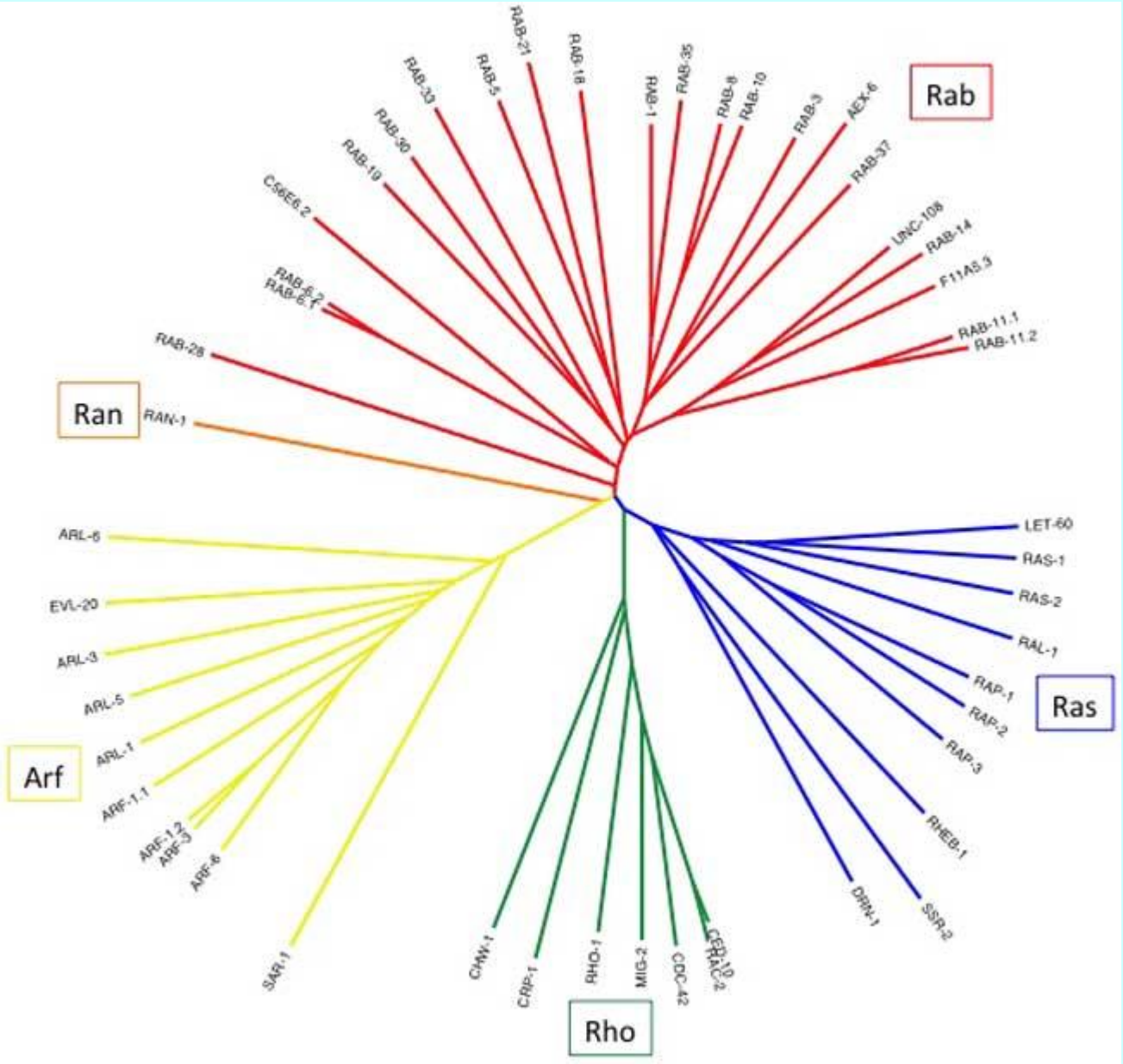




FOSFOLIPASE C



SMALL GTPase



Superfamiglia delle **Ras** (Rat sarcoma). Rispondono a molti stimoli extracellulari, possono interagire con molteplici effettori che regolano le reti di segnalazione citoplasmatica, controllano l'espressione genica e regolano proliferazione cellulare, differenziazione e sopravvivenza. Hanno un ruolo cruciale nell'oncogenesi.

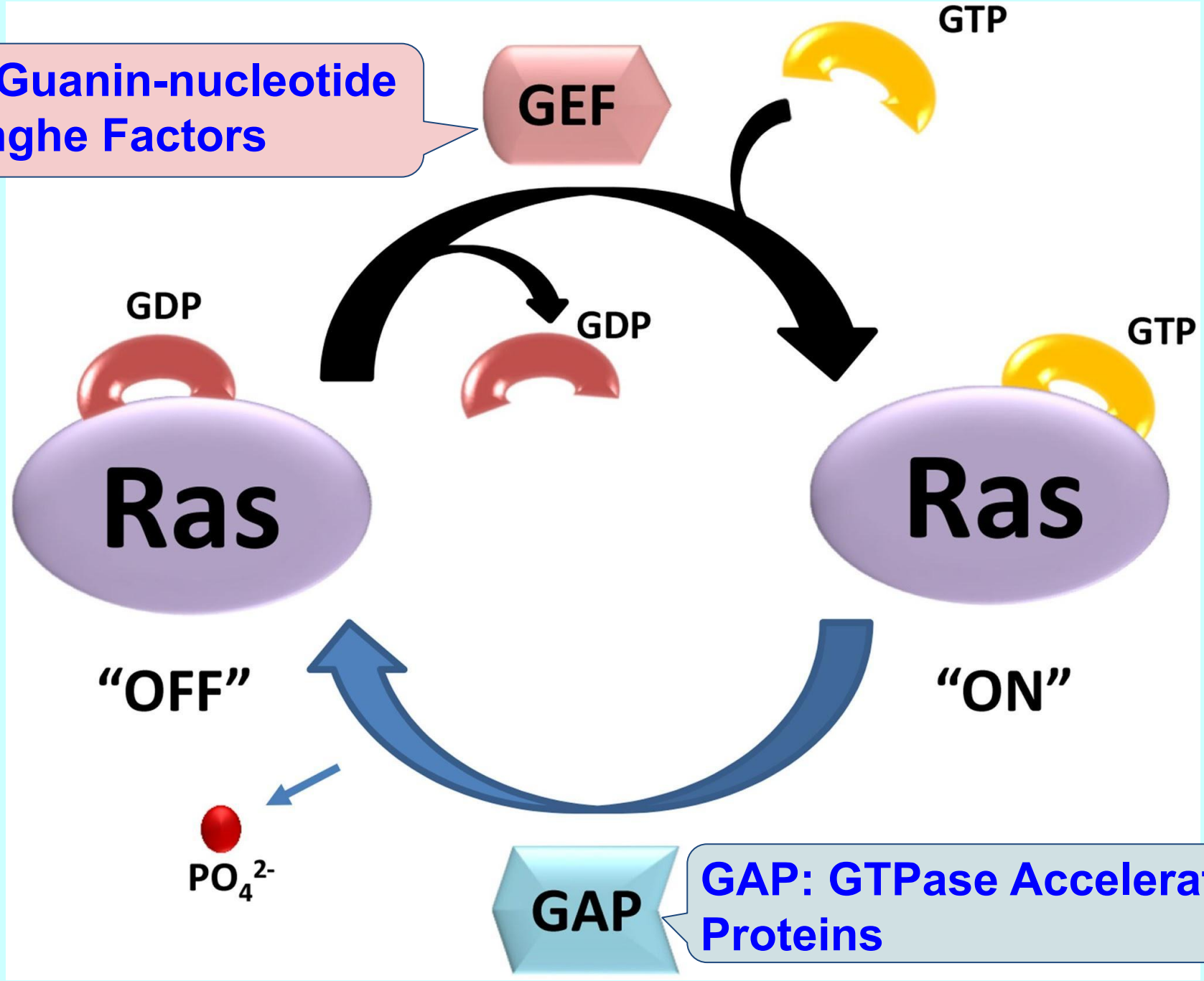
Le **Rho** (Ras homologous) fungono da regolatori chiave degli stimoli extracellulari, mediano reti di segnalazione che regolano l'organizzazione dell'actina, la progressione del ciclo cellulare e l'espressione genica.

Le **Rab** (Ras-like proteins in brain) sono regolatori del trasporto vescicolare intracellulare e dello spostamento di proteine tra diversi organelli endocitici e secretori.

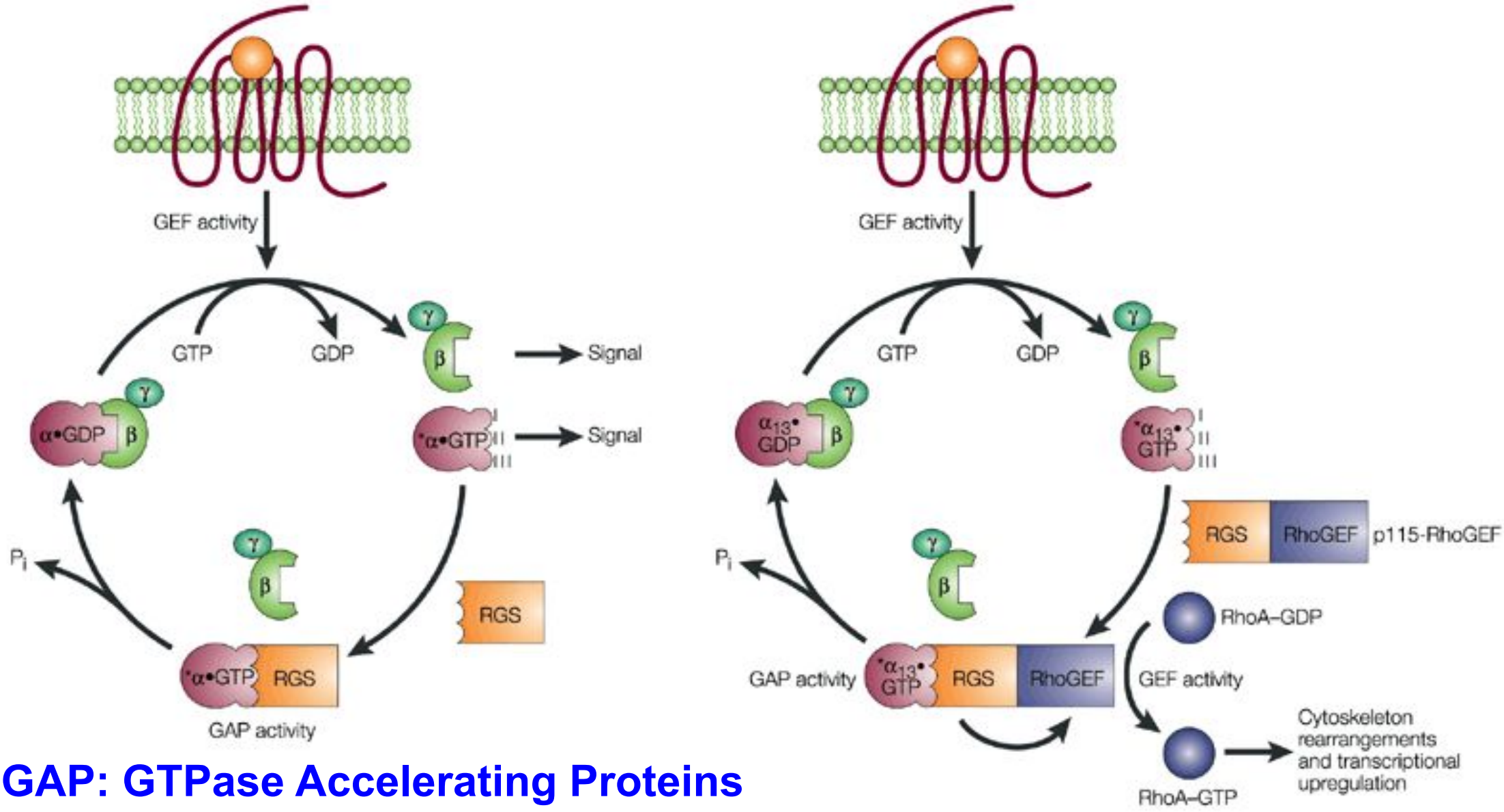
Le **Ran** (Ras-like nuclear) è la small GTPase più abbondante nella cellula ed è meglio conosciuta per la sua funzione nel trasporto nucleocitoplasmatico di RNA e proteine.

Le **Arf** (ADPribosylation factor) sono coinvolte nella regolazione del trasporto vescicolare.

SMALL GTPase



PROTEINE RGS



GAP: GTPase Accelerating Proteins

GEF: Guanin-nucleotide Excanghe Factors

RGS: Regulators of G-Protein Signalling

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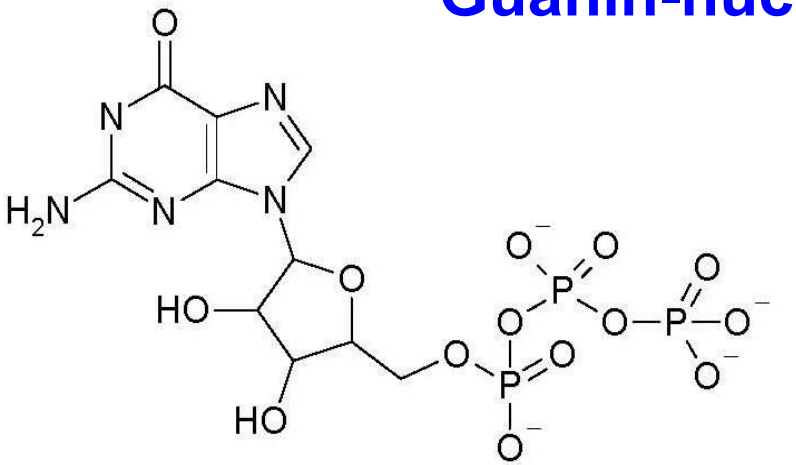
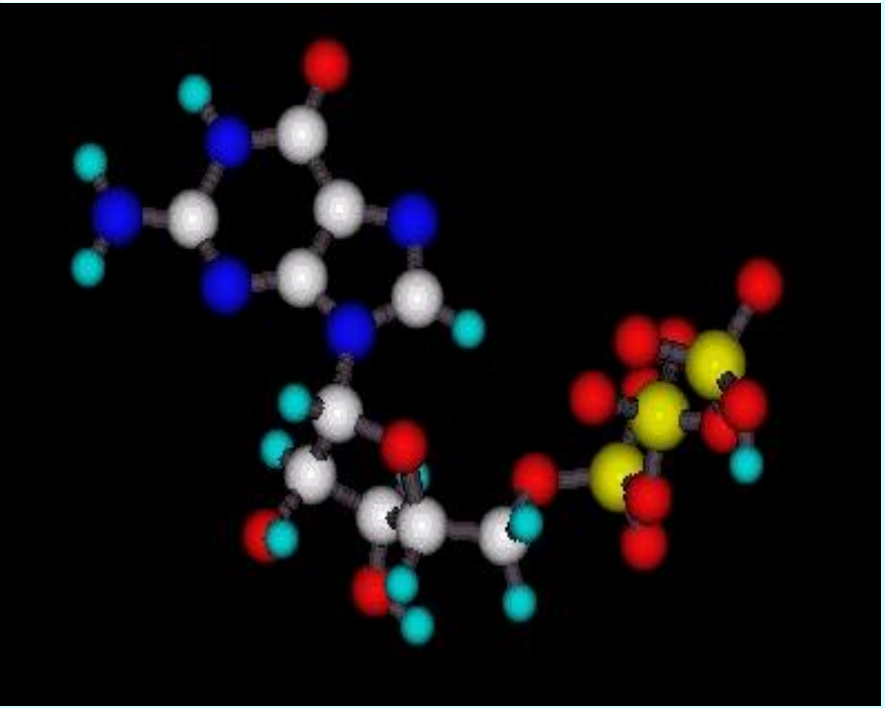
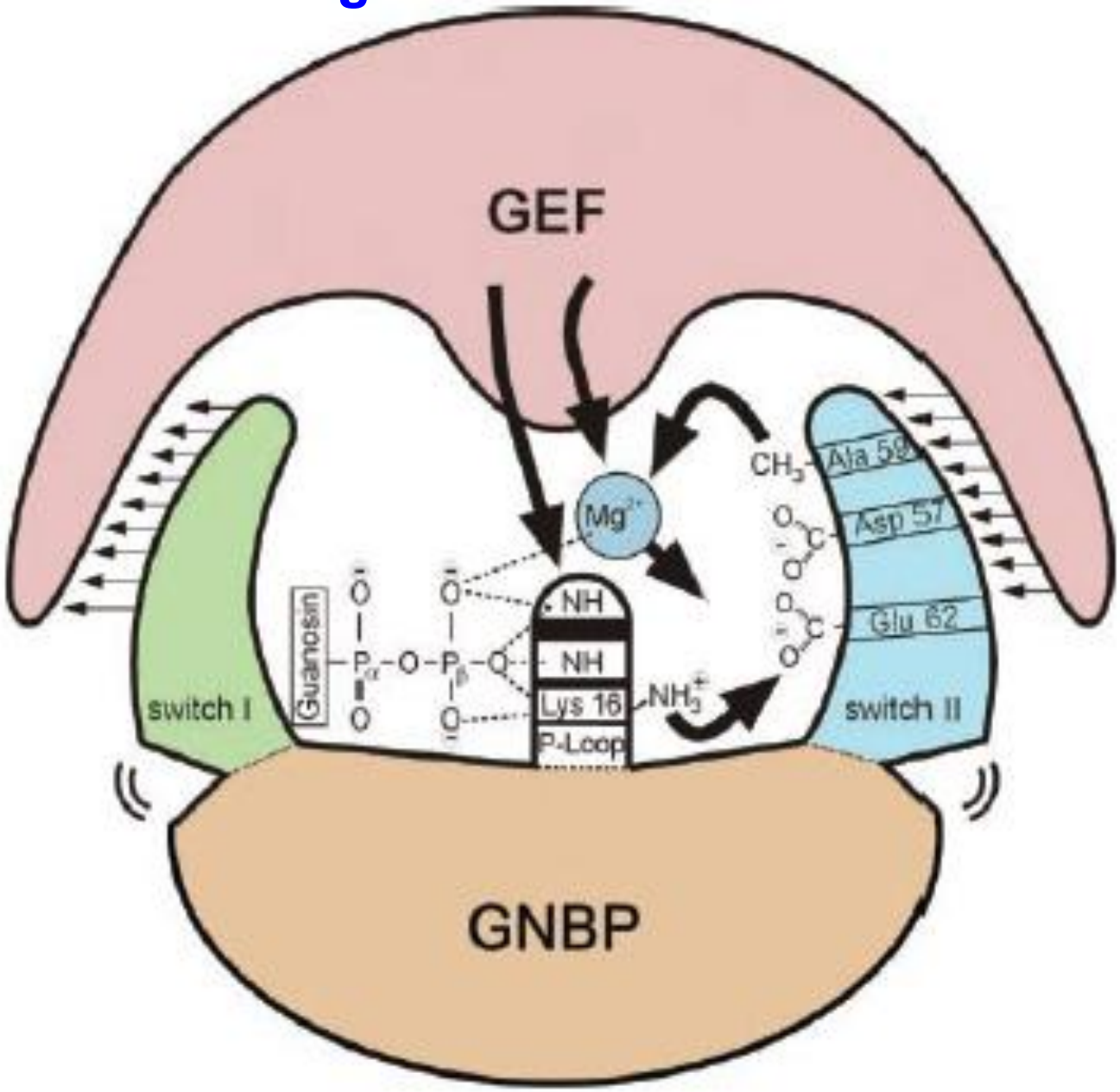
PROTEINE GEF

Guanin-nucleotide Exchange Factors



Guanosina trifosfato (GTP)





PROTEINE GAP

GTPase Accelerating Proteins

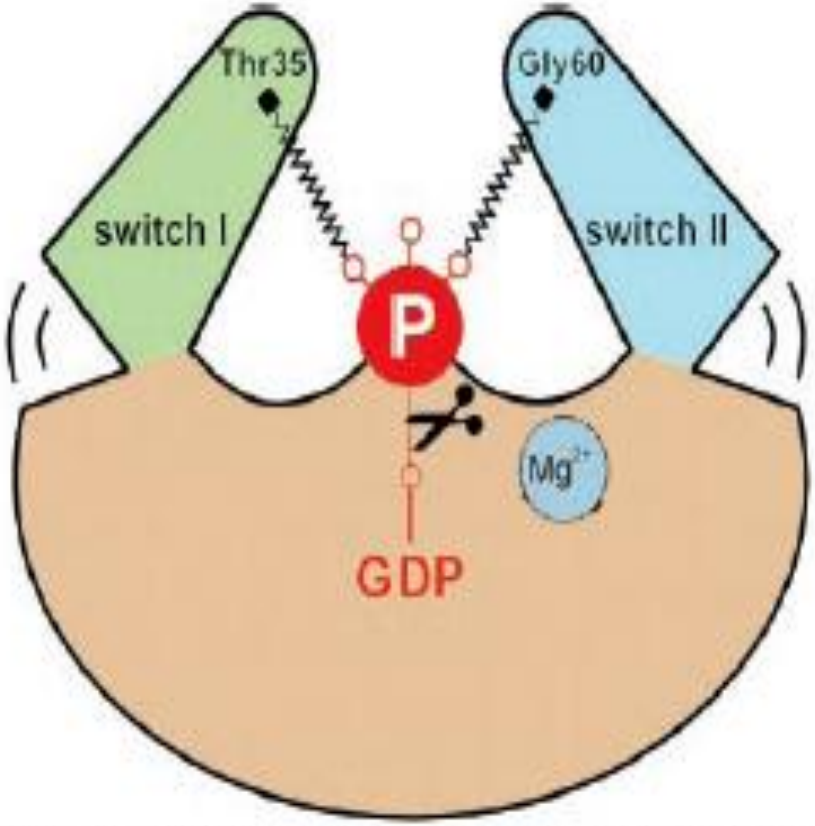
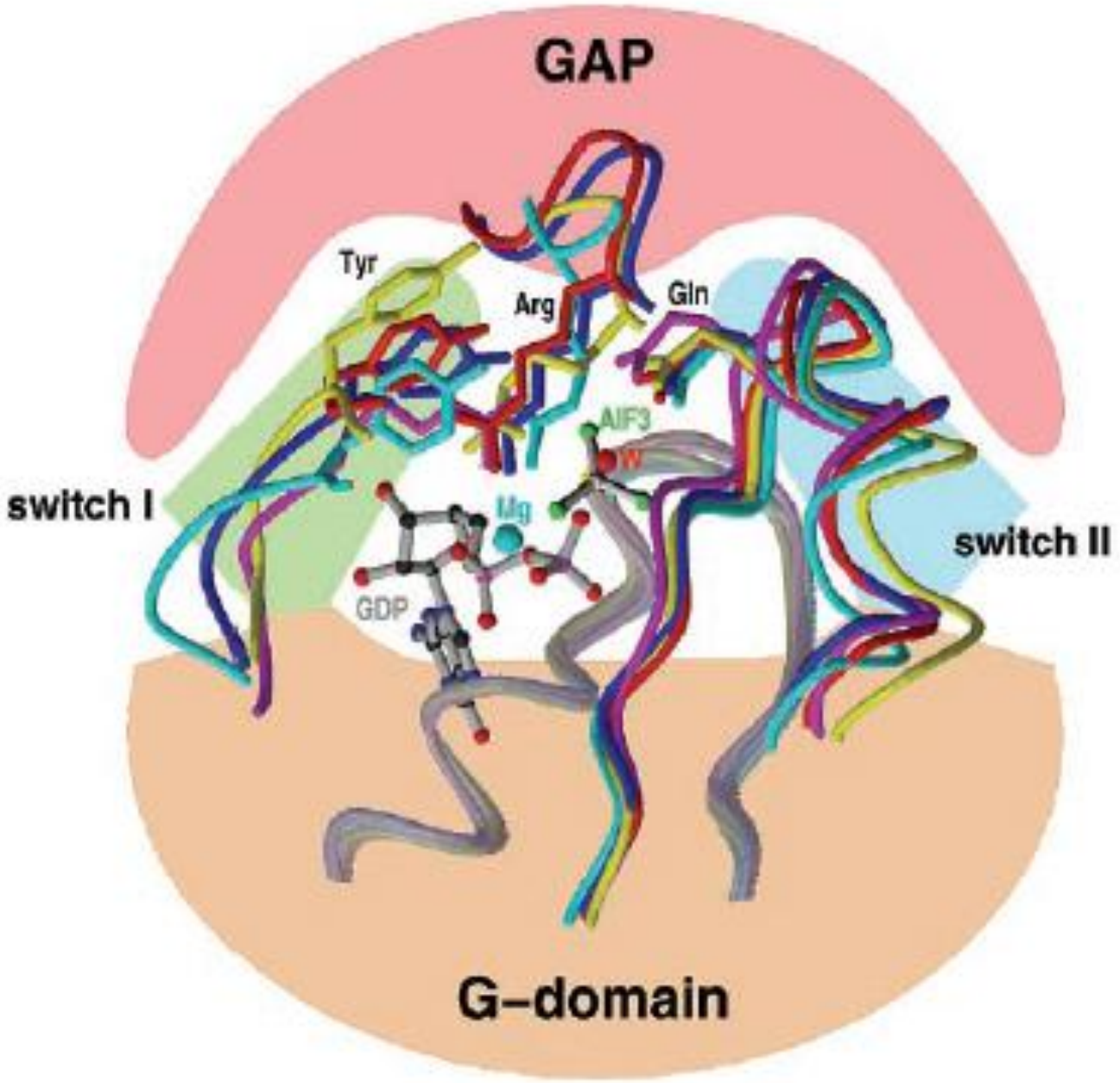
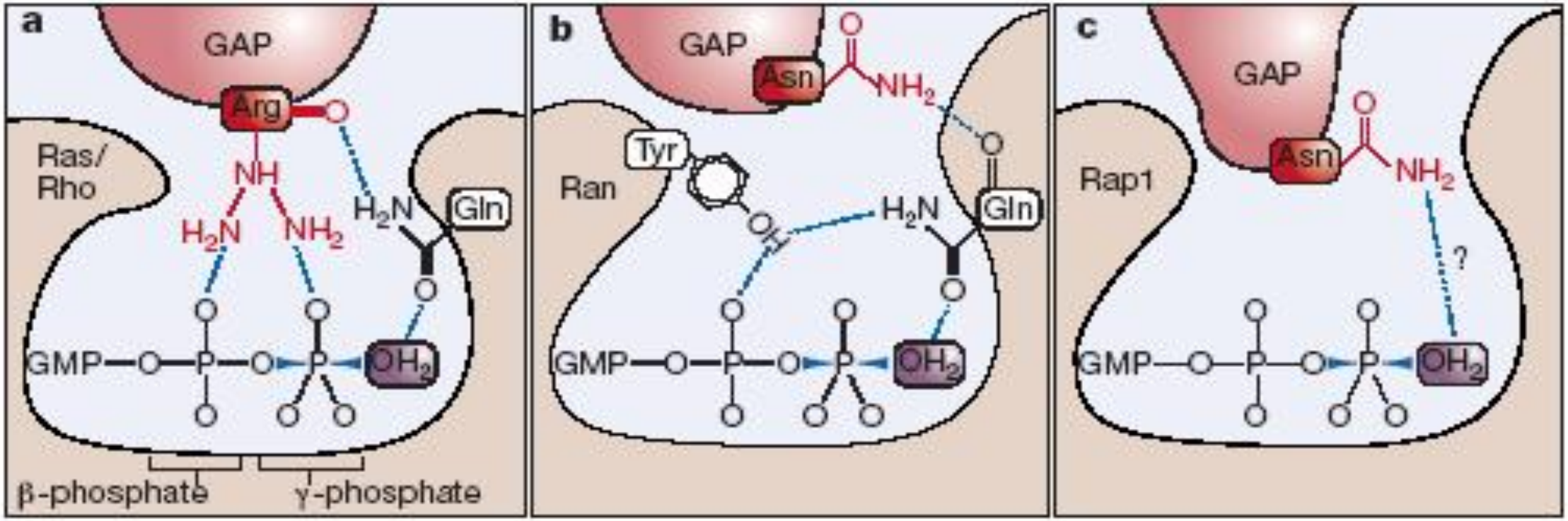


Diagramma schematico del meccanismo che coinvolge i dominii Switch I e Switch II che interagiscono con il gruppo γ-fosfato tramite i gruppi NH della catena degli amminoacidi invarianti Thr e Gly (meccanismo molla caricata). l'idrolisi del gruppo γ-fosfato permette il rilassamento dei due domini in differenti conformazioni.



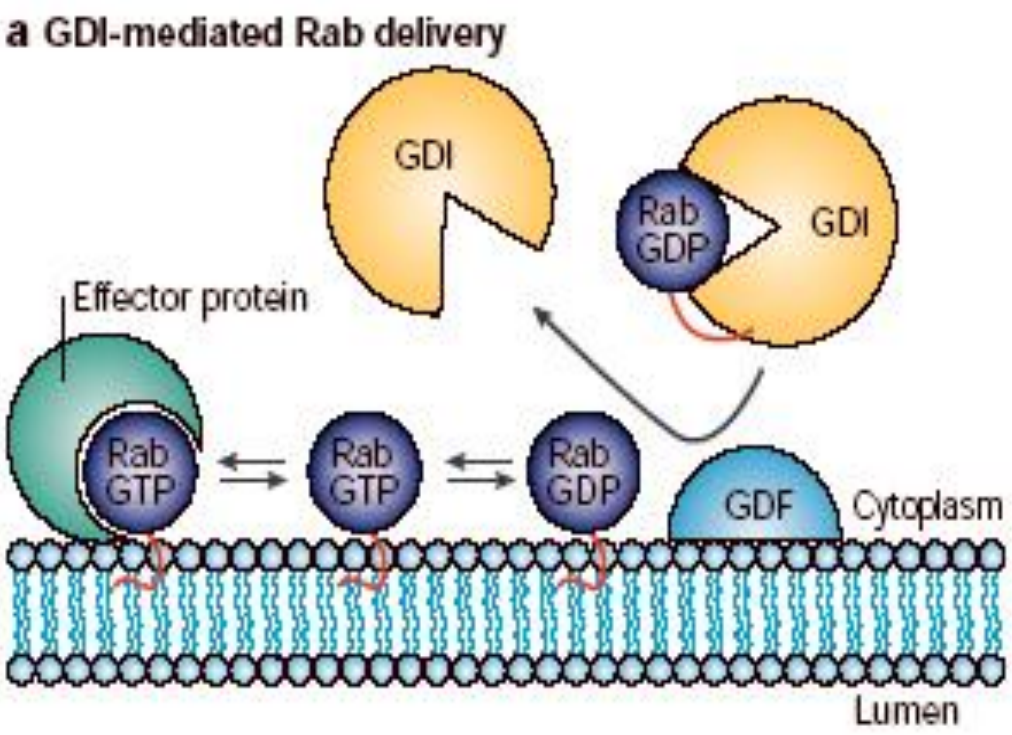
PROTEINE GAP

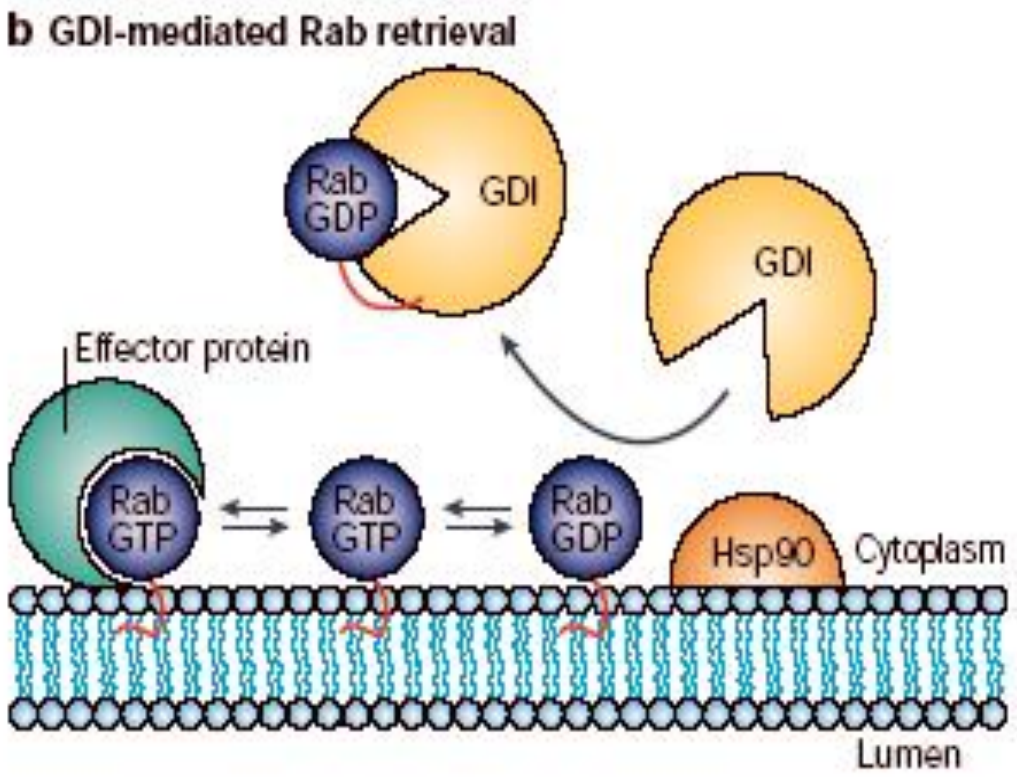


The active sites of some G-protein–GAP complexes. The complexes are shown in the transition state through which they pass during GTP hydrolysis. A common task for all GAPs is to position and fix a water molecule (purple) that attacks the γ-phosphate of GTP. a, Ras in complex with RasGAP[2], or Rho in complex with RhoGAP[5]. Here, the 'arginine finger' (Arg) in the GAP helps to stabilize the glutamine (Gln) in the G protein, which in turn positions and fixes the H_2O. The arginine finger also neutralizes the negative charge that develops at the γ-phosphate, by contacting this and the β-phosphate. b, Ran in complex with RanGAP[3]. RanGAP does not provide an arginine finger, instead using an asparagine (Asn) to stabilize the glutamine. The negative charge is neutralized by a tyrosine (Tyr) in Ran itself. c, Rap1 in complex with Rap1GAP. Wittinghofer and colleagues[1] propose that here, an asparagine (Asn) in the GAP positions and fixes the H_2O directly. GMP, guanosine monophosphate.

PROTEINE GDI

GDP Dissociation Inhibitor

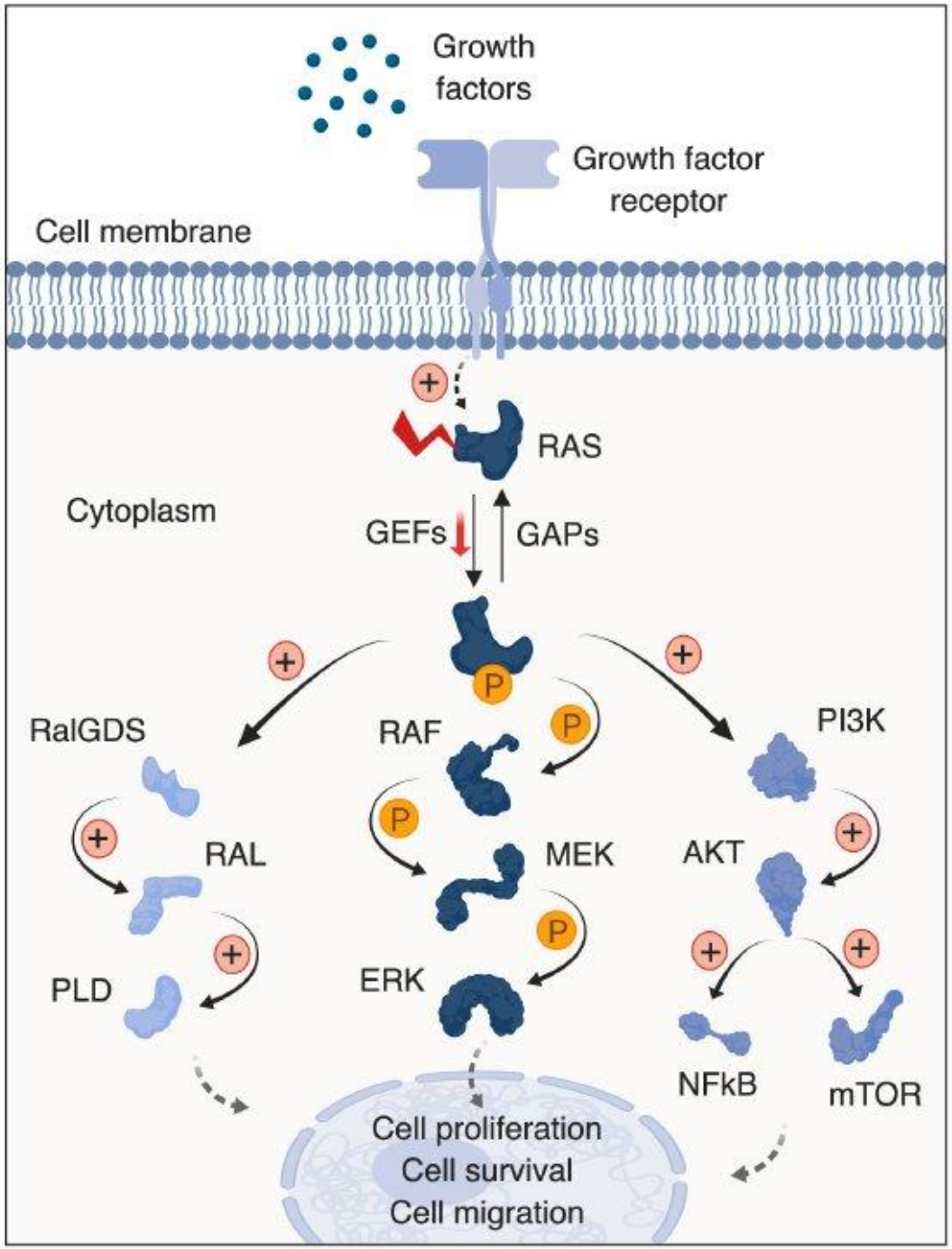




GDI Displacement Factors

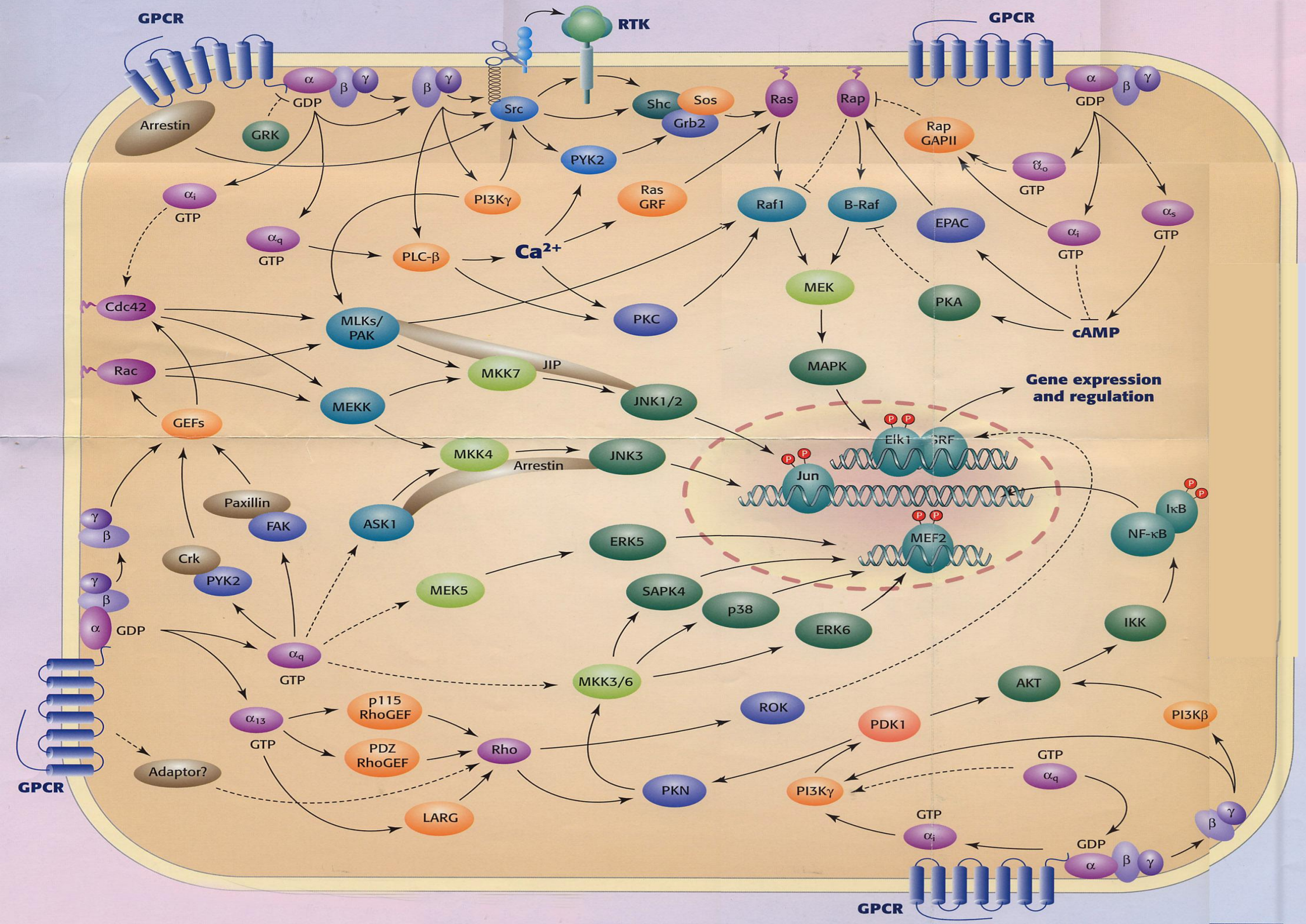
Heat Shock Protein 90

SMALL GTPase



Signaling pathways mediate dalle RAS proteins. Mutazioni delle RAS inibiscono idrolisi di GTP GAP-indotta, che porta ad accumulo di RAS attiva.

GAPs: GTPase-activating proteins
GEFs: guanine nucleotide exchange factors
RalGDS: Ral guanine nucleotide dissociation stimulator
RAL: RAS-related protein
PLD: phospholipase D
RAF: rapidly accelerated fibrosarcoma
MEK: Mitogen-activated protein kinase kinase
ERK: extracellular signal-regulated kinase
PI3K: phosphoinositide 3-kinase
NF-κB: nuclear factor kappa-light-chain-enhancer of activated B cells
mTOR: mammalian target of rapamycin.


GPCR
RTK
GPCR
GPCR
Arrestin
GRK
GDP
Src
Shc
Sos
Grb2
Ras
Rap
Rap GAPII
GDP
PYK2
PI3Kγ
Ras GRF
Raf1
B-Raf
EPAC
GTP
GTP
GTP
GTP
PLC-β
Ca2+
MEK
PKA
cAMP
Cdc42
MLKs/ PAK
PKC
Rac
MKK7
JIP
MAPK
Gene expression and regulation
MEKK
JNK1/2
GEFs
MKK4
Arrestin
JNK3
Elk1
SRF
Jun
Paxillin
FAK
ASK1
IκB
NF-κB
ERK5
MEF2
Crk
PYK2
MEK5
SAPK4
p38
ERK6
IKK
GDP
GTP
MKK3/6
AKT
p115 RhoGEF
ROK
PDK1
PI3Kβ
PDZ RhoGEF
Rho
Adaptor?
PKN
PI3Kγ
GTP
LARG
GTP
GDP
GPCR
GPCR

DIMERIZZAZIONE RECETTORI 7TM

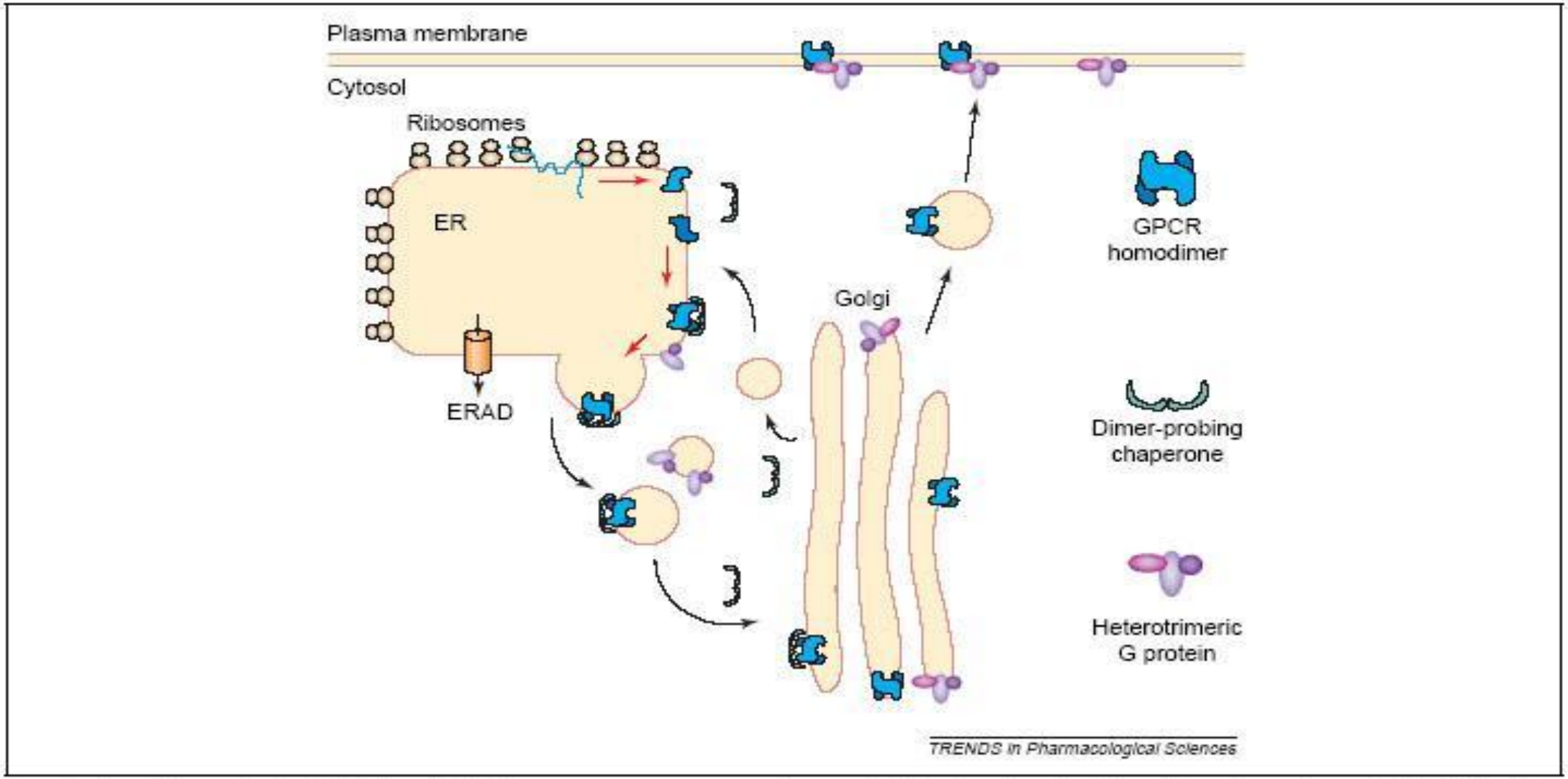
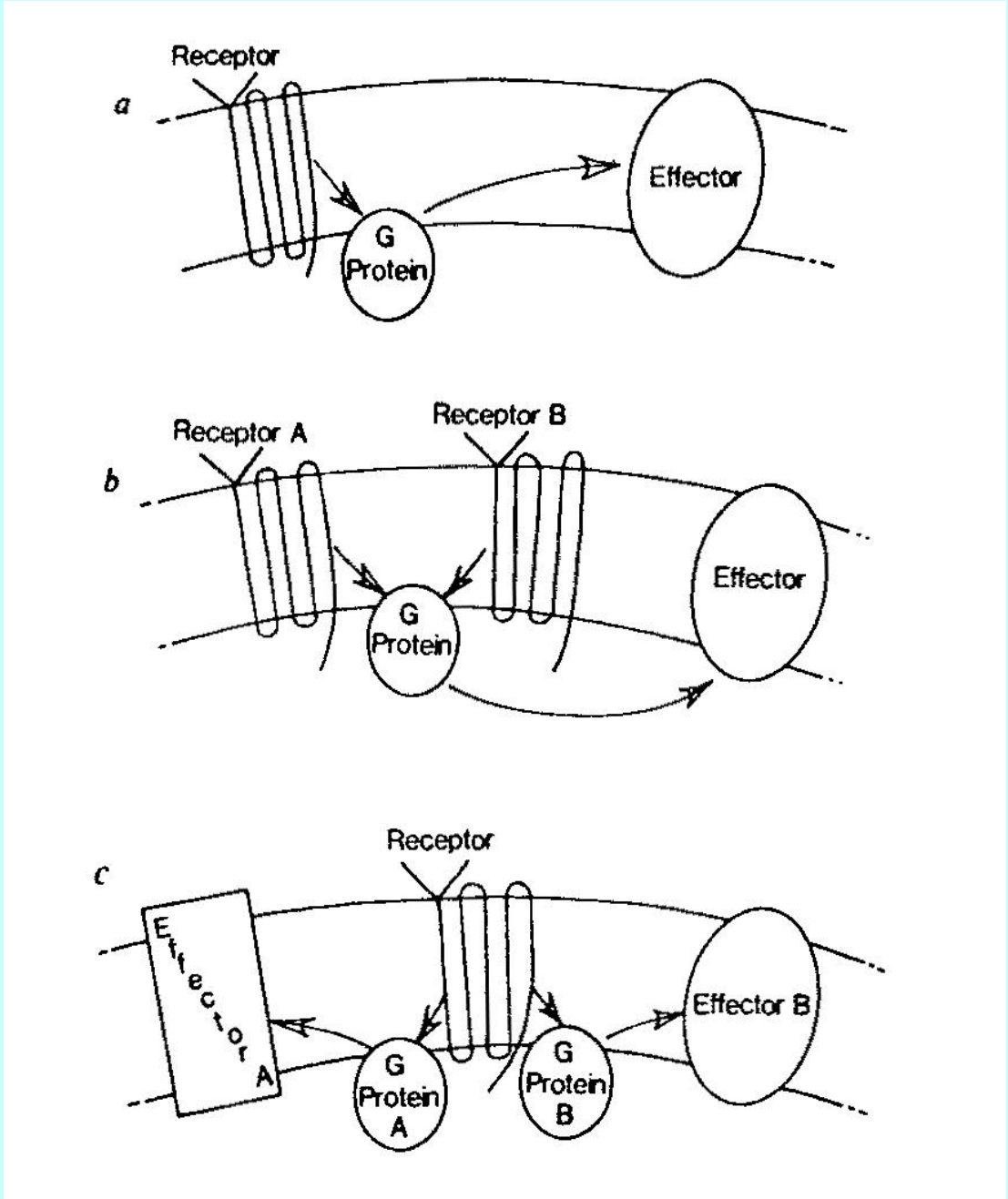
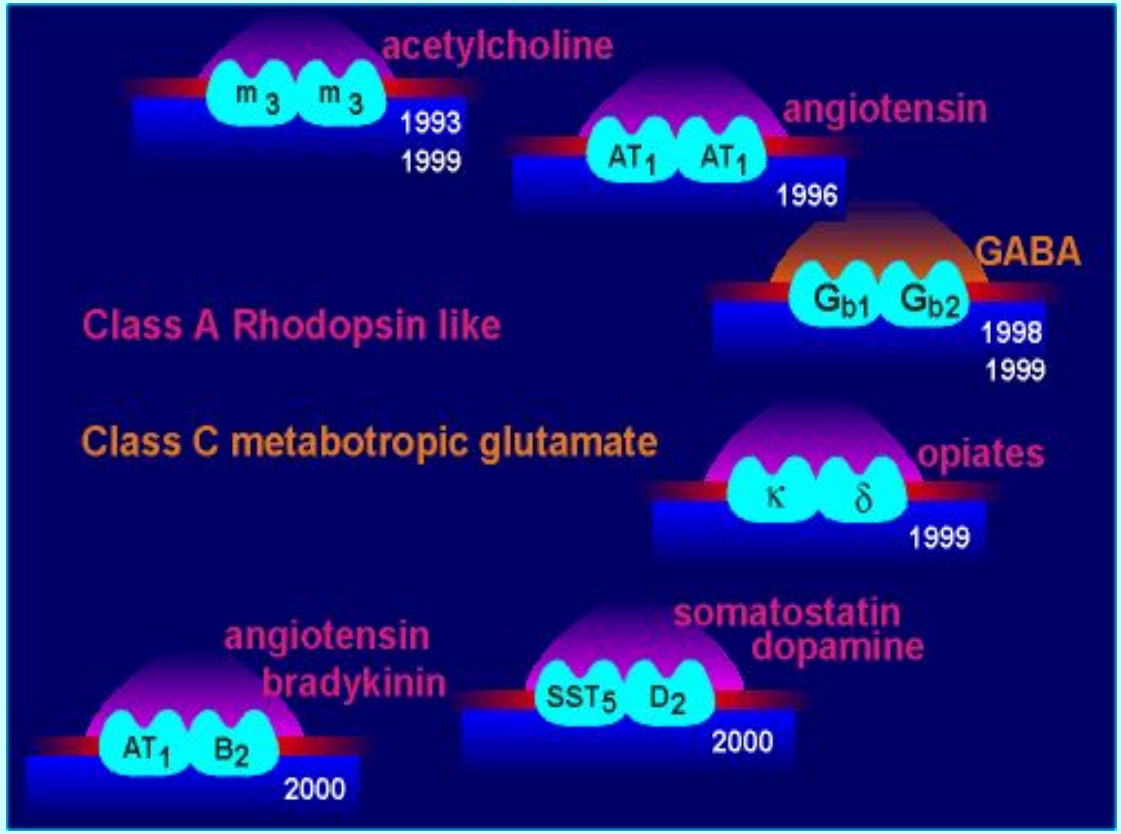


Figure 1. A model proposing that GPCR dimerization is a necessary step to pass quality-control checkpoints along the biosynthetic pathway. Nascent GPCR folding in the endoplasmic reticulum (ER) dimerize and interact with dimer-probing cytosolic chaperone proteins. Unfolded and monomeric GPCRs are degraded rapidly by the ER-associated degradation (ERAD) pathway. After maturation through the Golgi apparatus, GPCR homodimers are exported to the plasma membrane where they interact with a single heterotrimeric G protein. It is noteworthy that previous studies have suggested that the α- and βγ-subunits of G proteins also form a heterotrimer at an endomembrane location before reaching the plasma membrane [88,89]. Indeed, β- and γ-subunits are reported to combine in the ER, whereas heterotrimer assembly and α-subunit palmitoylation are reported to occur at the Golgi apparatus, thus generating a signal that enables the heterotrimer to leave the Golgi apparatus for the plasma membrane [88]. An alternative model (not shown in the figure) that has been proposed recently postulates that the assembly of the G-protein heterotrimer takes place at the ER, with its transport to the plasma membrane occurring independently of the Golgi apparatus [90].

ACCOPPIAMENTI RECETTORI 7TM





RECETTORI 7TM

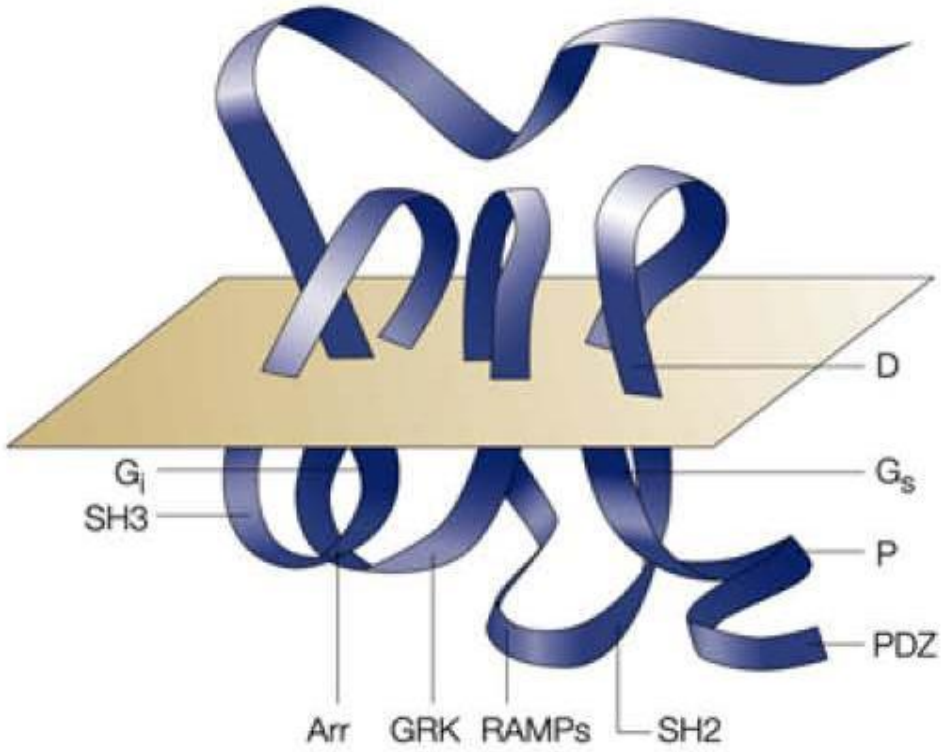


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Figure 1 | **Schematic diagram of a hypothetical G-protein-coupled receptor.** Labels denote general regions of interaction of the receptor with other cellular proteins, including different G proteins (G_i, G_s), PDZ-, SH2- and SH3-DOMAIN proteins, receptor-activity-modifying proteins (RAMPs), arrestin (Arr), G-protein-coupled receptor kinase (GRK), sites for dimerization with other GPCRs (D), and phosphorylation sites that lead to uncoupling and internalization (P). Any one of these active processes could be considered a form of expression of efficacy. The figure is a general description of various loci for protein interactions, but does not represent accurate locations, as, in most cases, these are not well characterized at present.

DESENSIBILIZZAZIONE GPCR

DOWNREGULATION - Eliminazione del recettore dalla cellula dovuta a:

- Diminuzione della biosintesi della proteina
- Destabilizzazione del m-RNA del recettore
- Aumento della degradazione del recettore

DISACCOPPIAMENTO del recettore dalla Proteina G attraverso un rapido processo di fosforilazione da parte di proteine kinasi:

- Desensibilizzazione omologa (non-agonista-specifica). I secondi messaggeri cAMP e DAG attivano rispettivamente la proteina kinasi A (PKA) e la proteina kinasi C (PKC) che fosforilano i GPCRs.
- Desensibilizzazione eterologa (agonista specifica). Per gli adrenoreccettori β è mediata da membri della famiglia delle GPCR kinasi (GRK). Queste kinasi, attivate dal dimero βγ delle Proteine G, fosforilano esclusivamente GPCRs complessati con agonisti o comunque in forma attiva.

La fosforilazione di GPCRs mediata dalle GRK aumenta l'affinità del recettore per proteine citoplasmatiche note come β-arrestine che si legano al recettore ed impediscono l'accoppiamento con le Proteine G. Le β-arrestine sono anche coinvolte nel processo di internalizzazione del recettore insieme alla proteina clatrina e la Proteina Adattatrice 2 (AP2).

RECETTORI 7TM

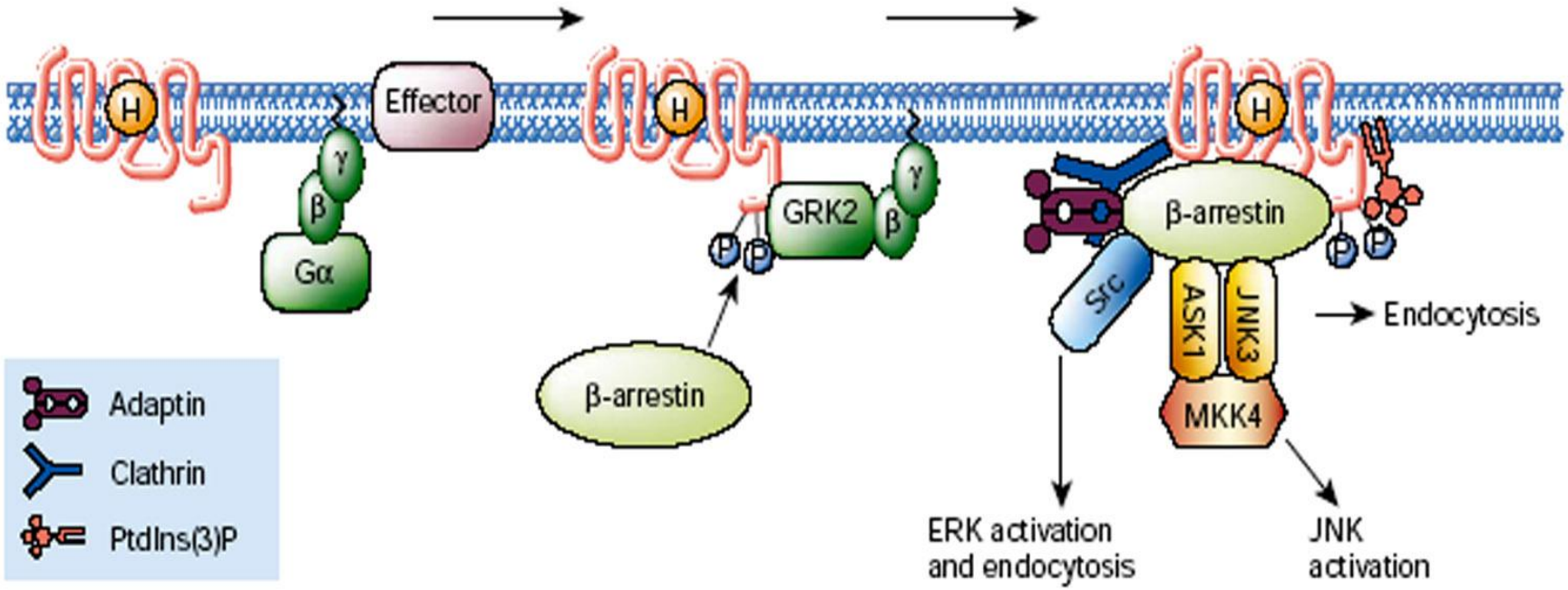
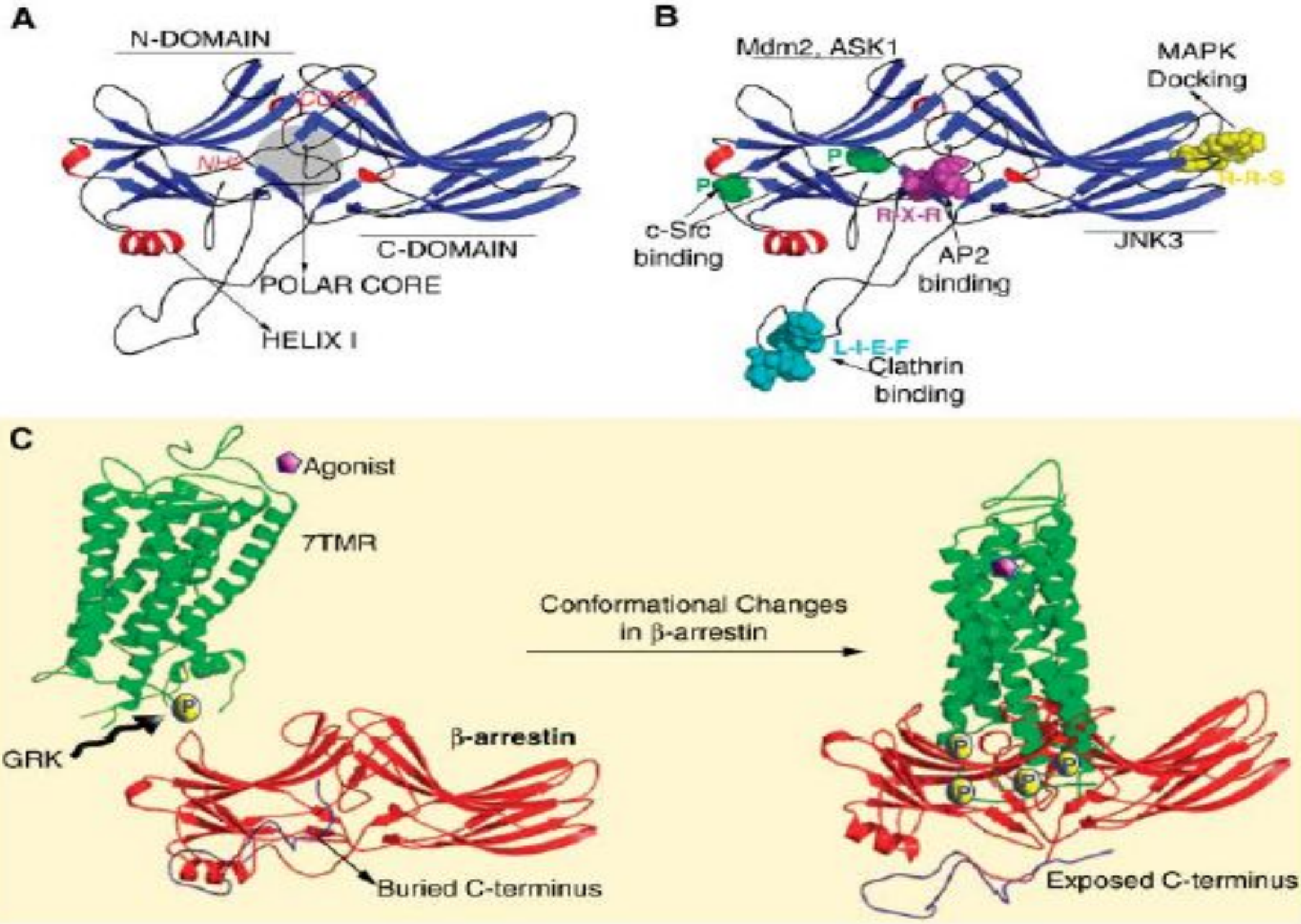
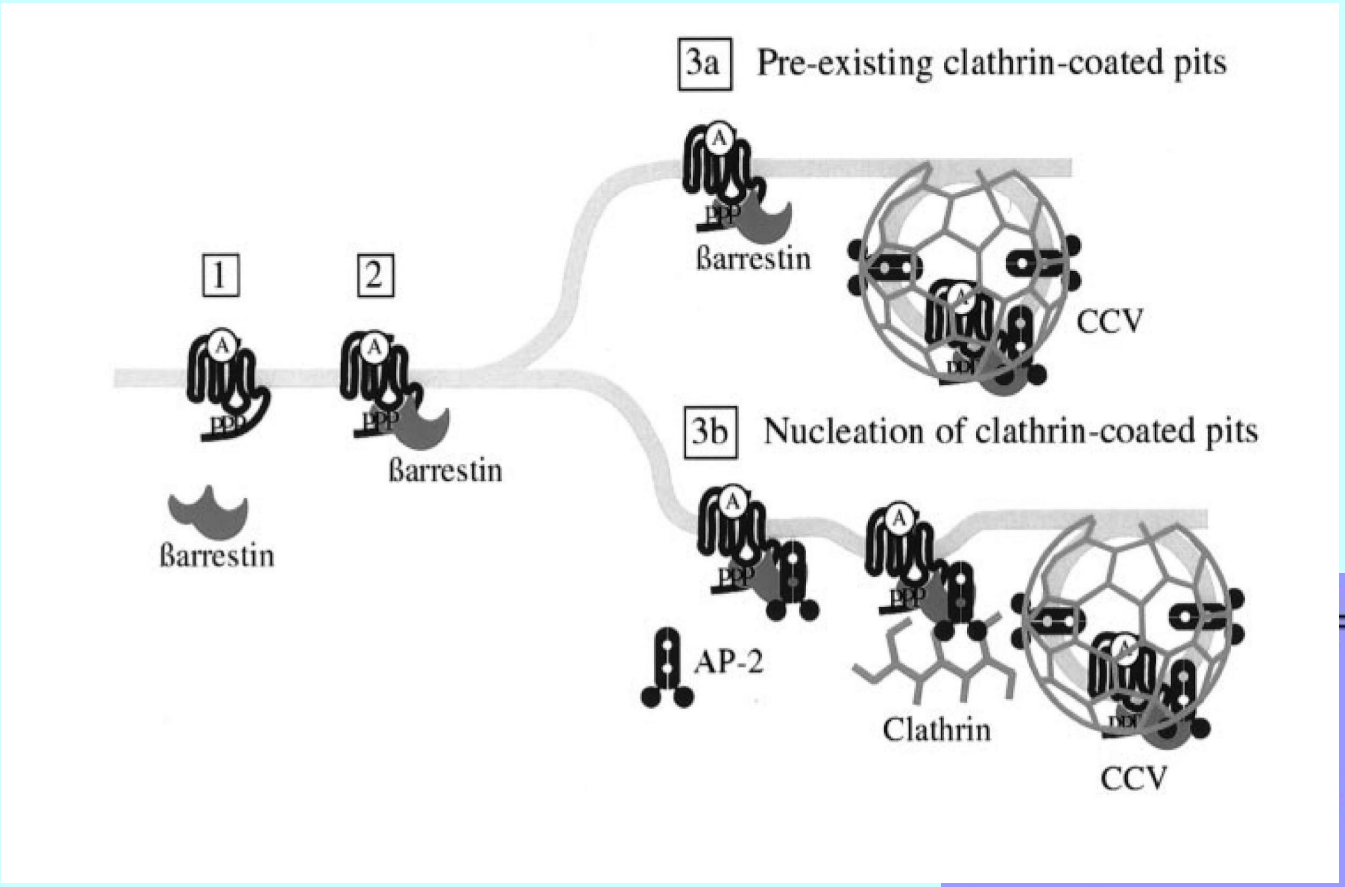
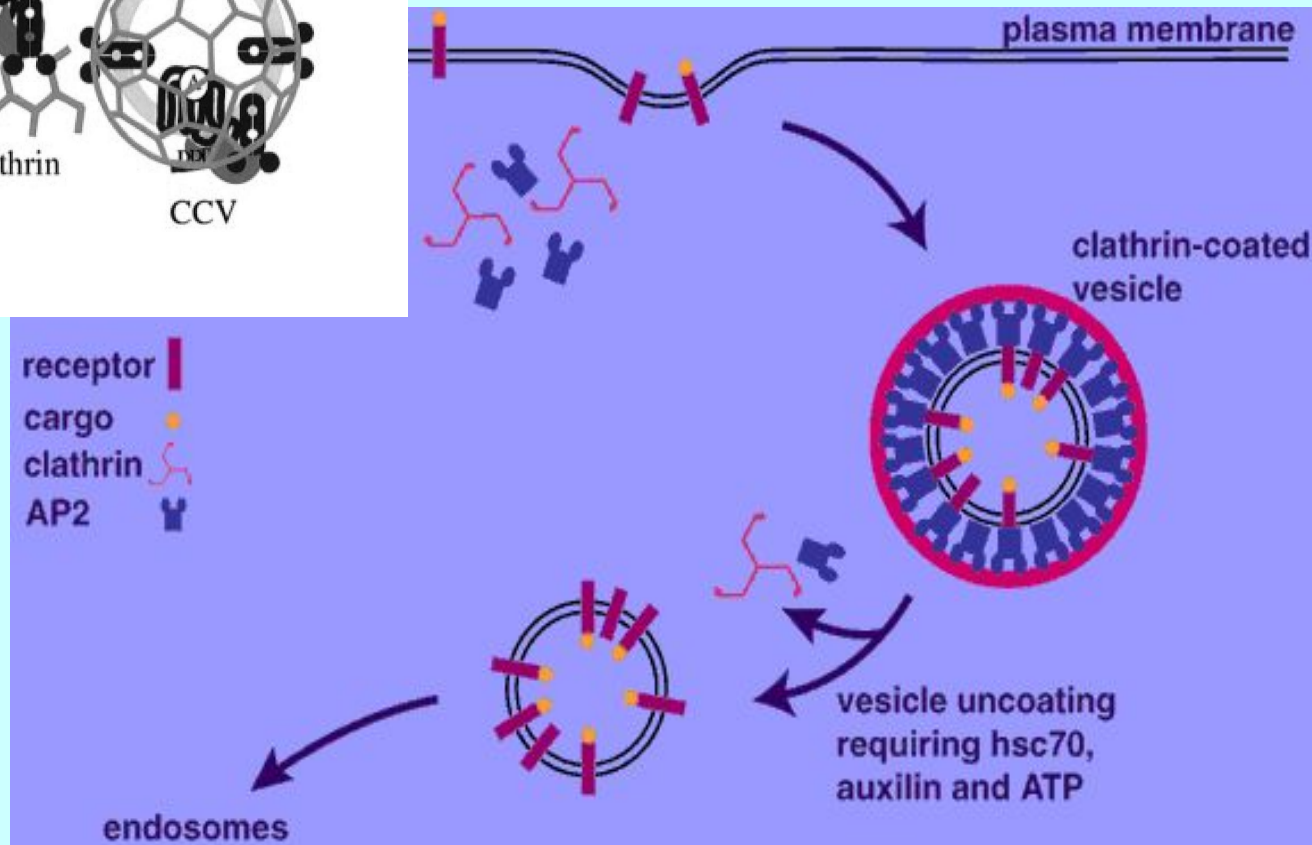


Figure 2 The many signalling roles of seven-transmembrane-spanning receptors. Agonist binding to the receptor results in the dissociation of heterotrimeric G proteins into Gα-GTP and Gβγ subunits, which activate G-protein effectors. One consequence of Gβγ subunit release is enhanced GRK-mediated phosphorylation of the agonist-occupied receptor. β-Arrestin binds to the GRK-phosphorylated receptor and not only uncouples the receptor from its G protein, but also acts as a scaffold to recruit clathrin and the clathrin adaptor AP2, which target the activated receptor for endocytosis. β-Arrestin-mediated interaction with Src can promote activation of extracellular-regulated kinase (ERK) and endocytosis, by phosphorylating members of the internalization complex such as dynamin. The interaction of β-arrestin with a MAPK kinase kinase (for example, ASK1) and c-Jun N-terminal kinase 3 (JNK3) can lead to the assembly of the JNK module with a MAPK kinase (for example, MKK4), which leads to activation of the MAPK JNK3. Thus the β-arrestins seem to act as adaptors that link GPCRs to both clathrin and the clathrin adaptor AP2, and as scaffolds that bring together other elements of a receptor complex to facilitate the activation of signalling pathways.


A
N-DOMAIN
COOH
NH2
C-DOMAIN
POLAR CORE
HELIX I
B
Mdm2, ASK1
MAPK
Docking
P
P
R-X-R
R-R-S
c-Src
binding
AP2
binding
JNK3
L-I-E-F
Clathrin
binding
C
Agonist
7TMR
Conformational Changes
in β-arrestin
GRK
β-arrestin
Buried C-terminus
Exposed C-terminus

DESENSIBILIZZAZIONE GPCR





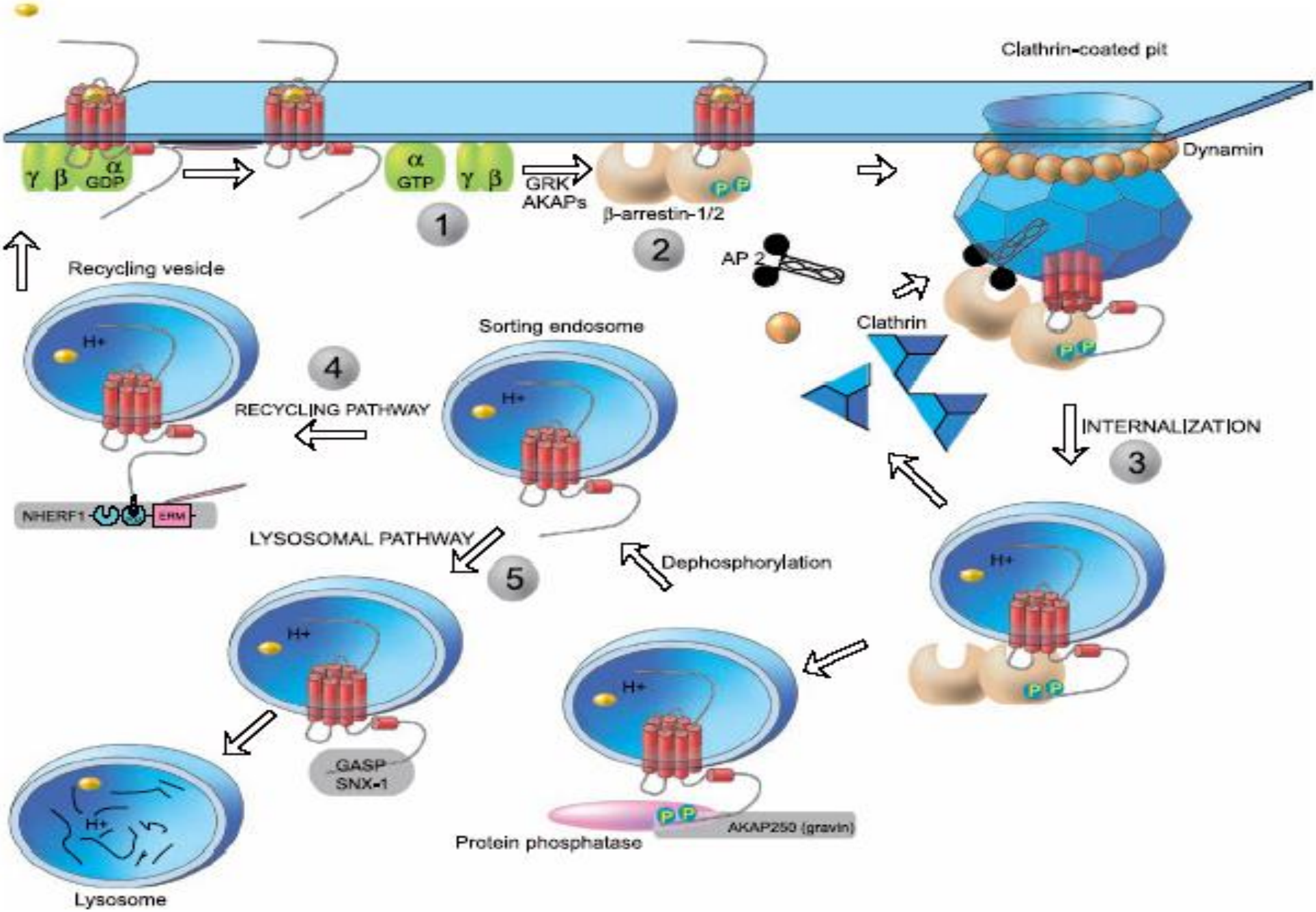


Fig. 8. Pathways involved in desensitization and resensitization of GPCR signaling. Typically activation of a GPCR leads to (1) activation and inhibition of specific signaling pathways in the cell, (2) short-term desensitization mediated by phosphorylation of GPCRs by GRKs followed by β-arrestin binding to GPCRs that uncouple the receptor at the plasma membrane from the G-protein, (3) endocytosis of the receptor, followed by postendocytic sorting of the receptor either (4) back to the plasma membrane or (5) to lysosomes for degradation.

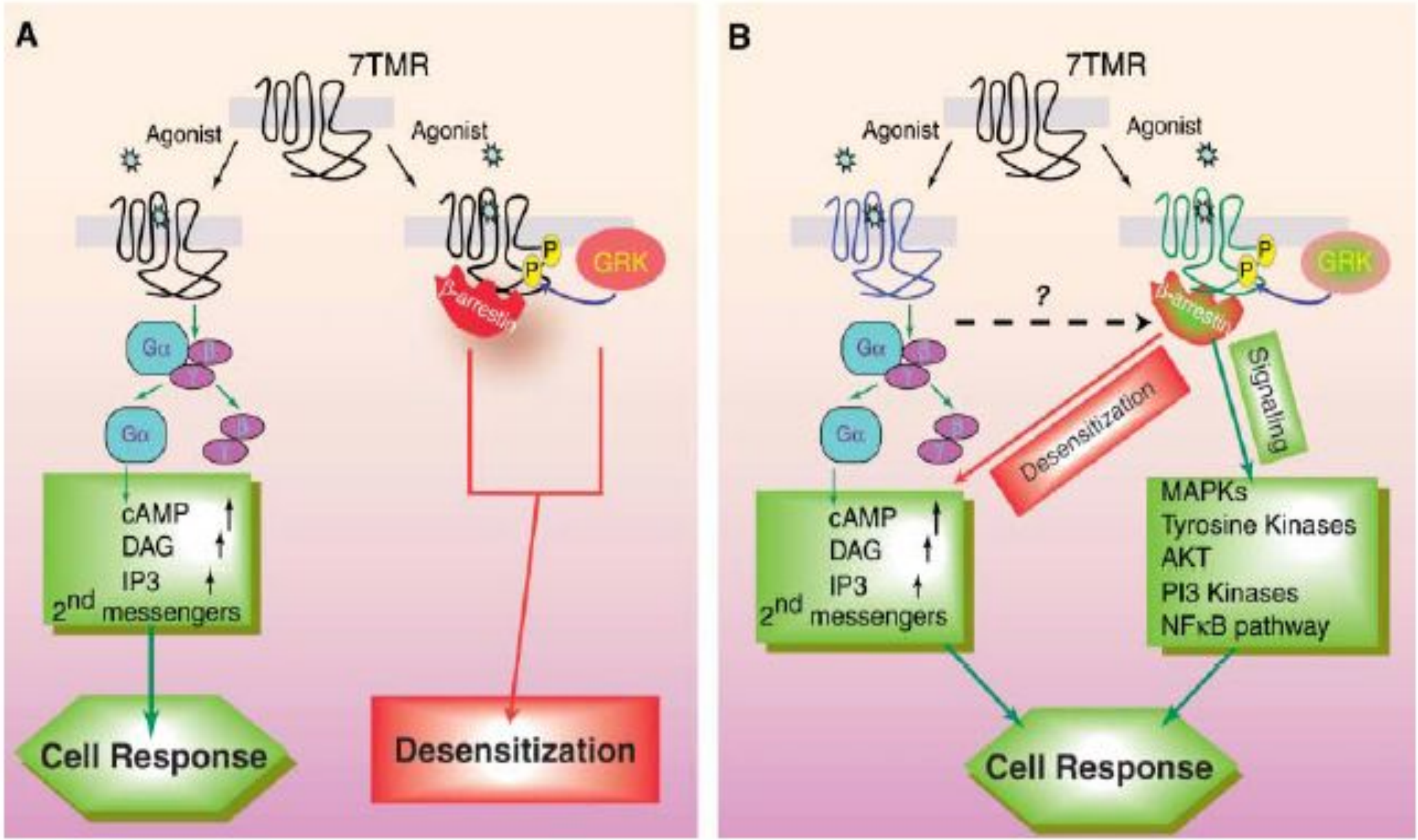
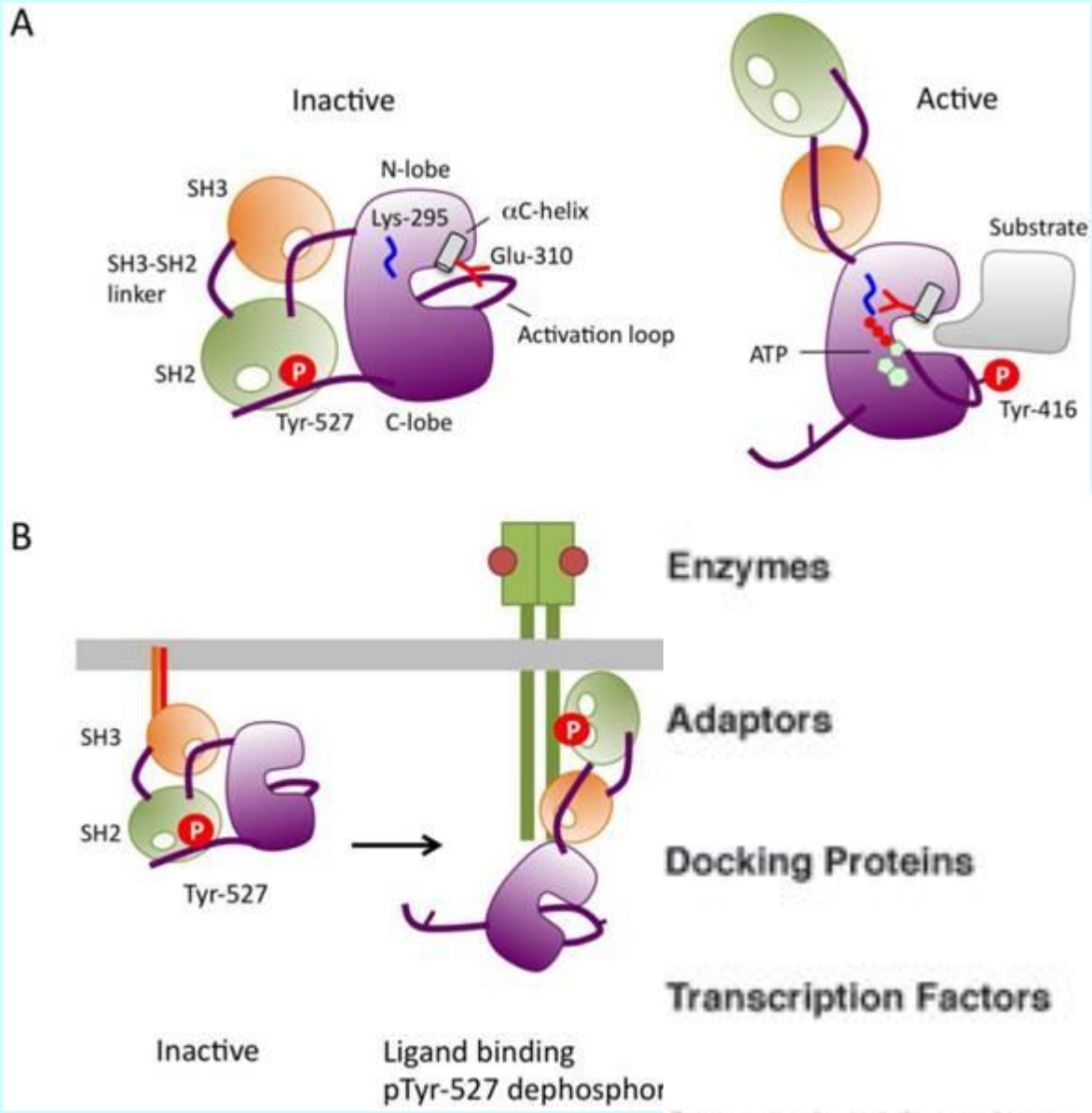
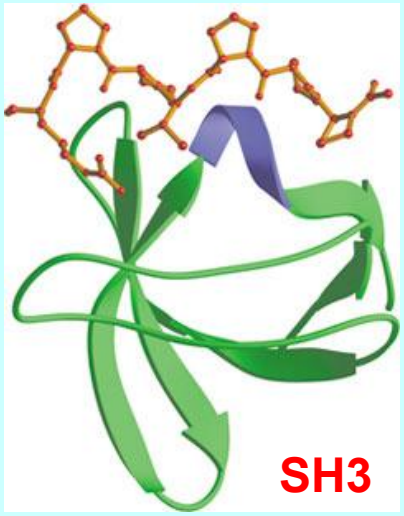
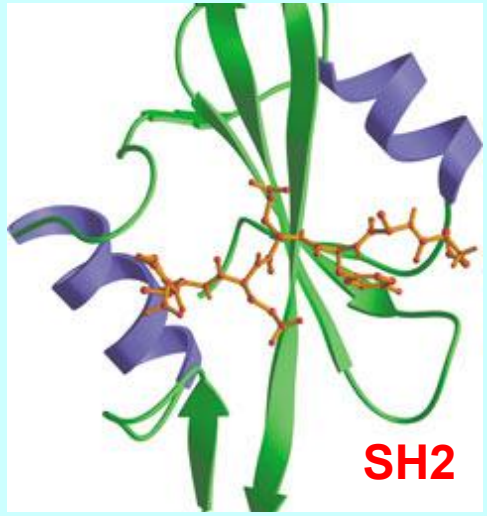


Fig. 1. Signal transduction by seven transmembrane receptors. (A) Classical paradigm. The active form of the receptor (R*) stimulates heterotrimeric G proteins and is rapidly phosphorylated by G protein–coupled receptor kinases (GRKs), which leads to β-arrestin recruitment. The receptor is thereby desensitized, and the signaling is stalled. (B) New paradigm. β-Arrestins not only mediate desensitization of G protein–signaling but also act as signal transducers themselves.

Kinasi c-SRC e domini SH2 - SH3







FUNZIONI INDIPENDENTI DALLE PROTEINE G

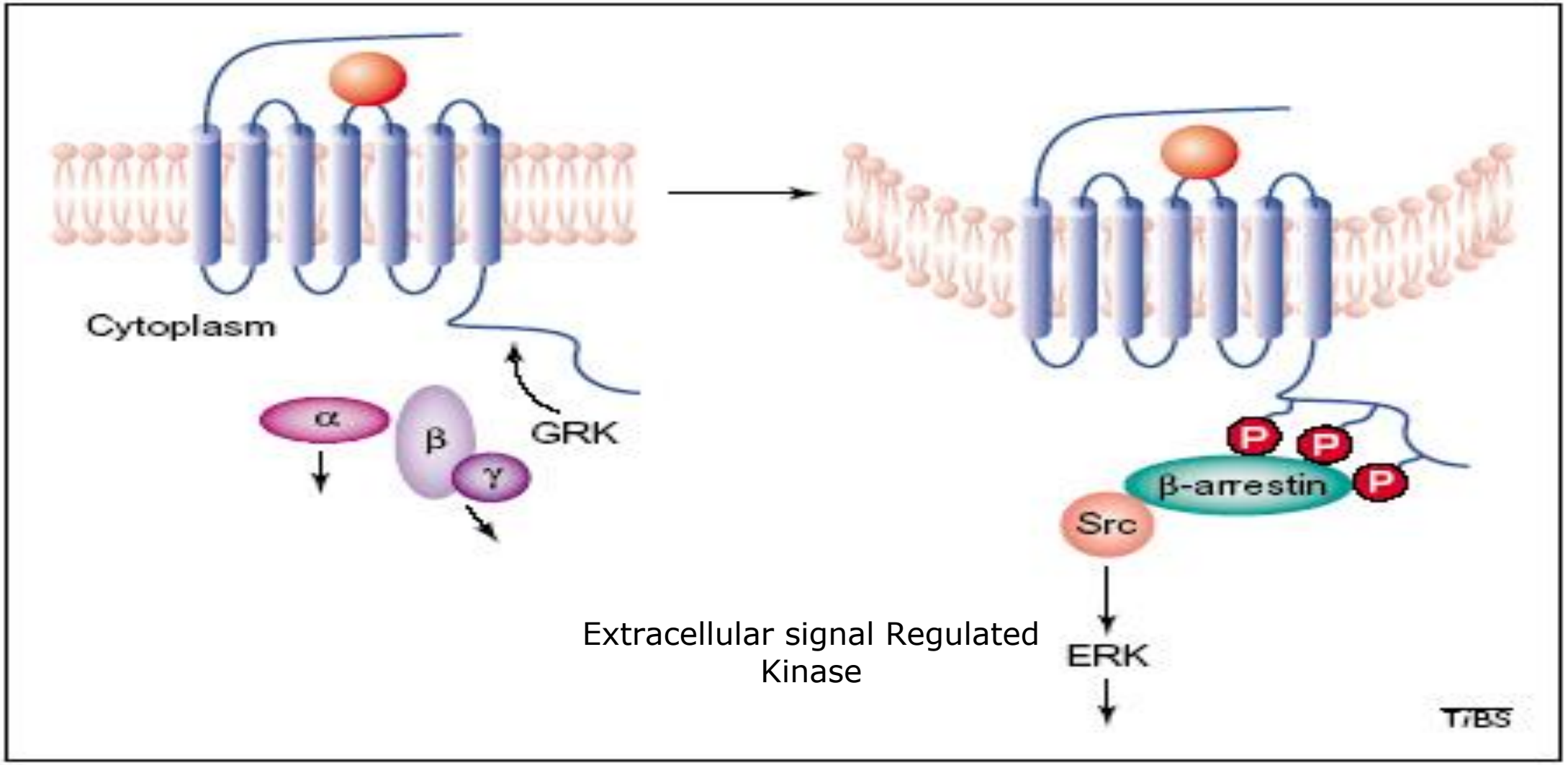


Fig. 2. β_2 Adrenoceptor can mediate the Src kinase pathway in a G-protein-independent manner. Ligand (red circle) binding to β_2 adrenoceptor (blue) promotes the release of Gα and Gβγsubunits, activating downstream effector molecules. A specific GRK (G-protein-coupled receptor kinase) phosphorylates the carboxy tail of the receptor, promoting the association of β-arrestin with, and the uncoupling of G proteins from, the receptor. β-Arrestin also binds Src, thereby recruiting Src to the membrane. β-Arrestin, interacting with clathrin, targets the complex to clathrin-coated pits (membrane indentation). Src-dependent activation of the ERK pathway requires both β-arrestin-mediated recruitment, and the targeting, of the receptor to clathrin-coated pits.

FUNZIONI INDIPENDENTI DALLE PROTEINE G

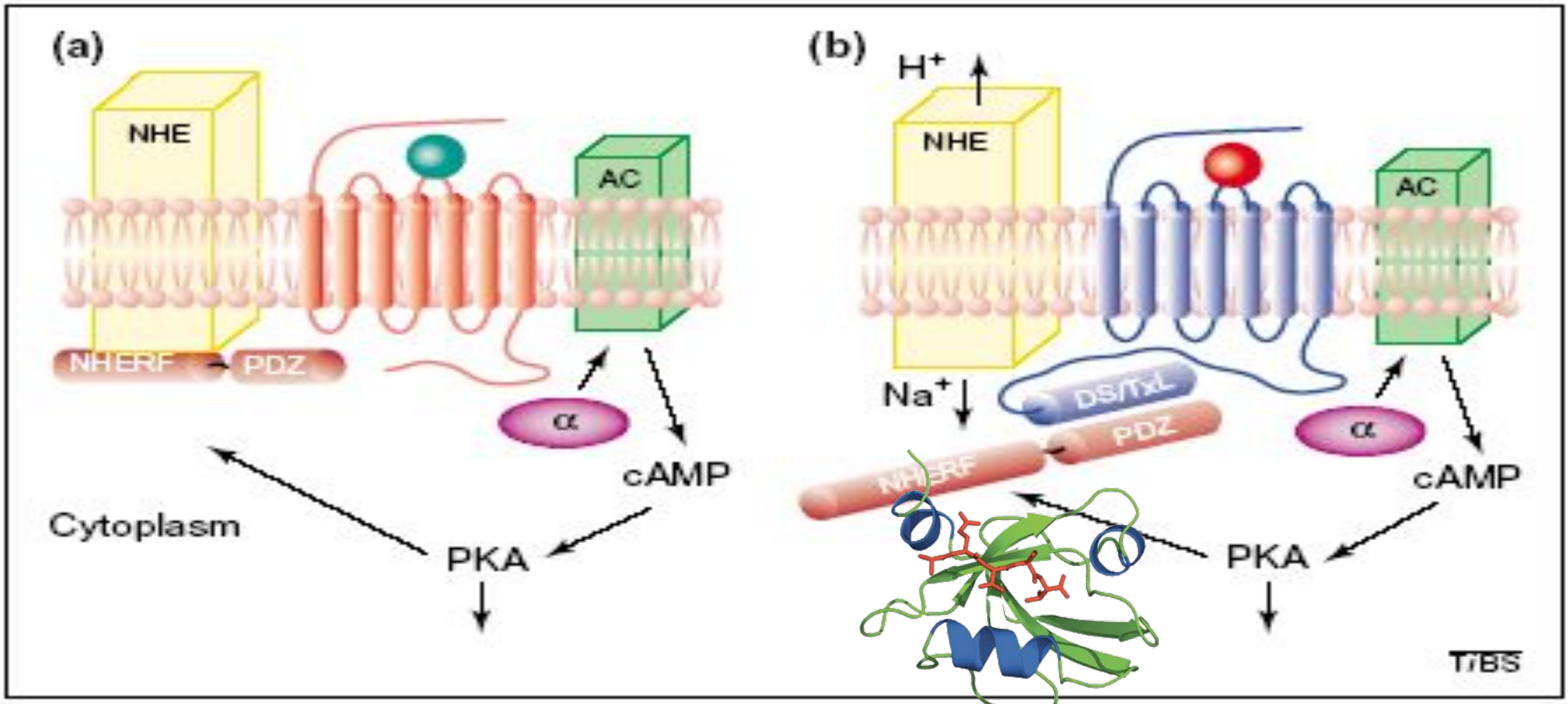


Fig. 4. β_2 adrenoceptor regulates the Na^+–H^+ exchanger (NHE) in a G-protein-independent manner. (a) Activation (green circle) of the parathyroid hormone receptor (red) attenuates NHE activity in a protein kinase A (PKA)-dependent manner via Na^+–H^+ exchanger regulatory factor (NHERF) binding. PKA activity is stimulated via a G-protein-dependent pathway, promoting the association of the amino terminus of NHERF with NHE, attenuating the Na^+–H^+ exchange. (b) Activation (red circle) of β_2 adrenoceptor (blue), which also activates PKA, positively regulates NHE. A NHERF-binding site (DS/TxL) in the carboxy tail of β_2 adrenoceptor becomes available after ligand binding. The carboxy terminal PDZ domain on NHERF directs its association with the receptor, thereby releasing NHE from the negative regulatory effect of NHERF. Abbreviation: AC, adenylyl cyclase.

RECETTORI INFEDELI

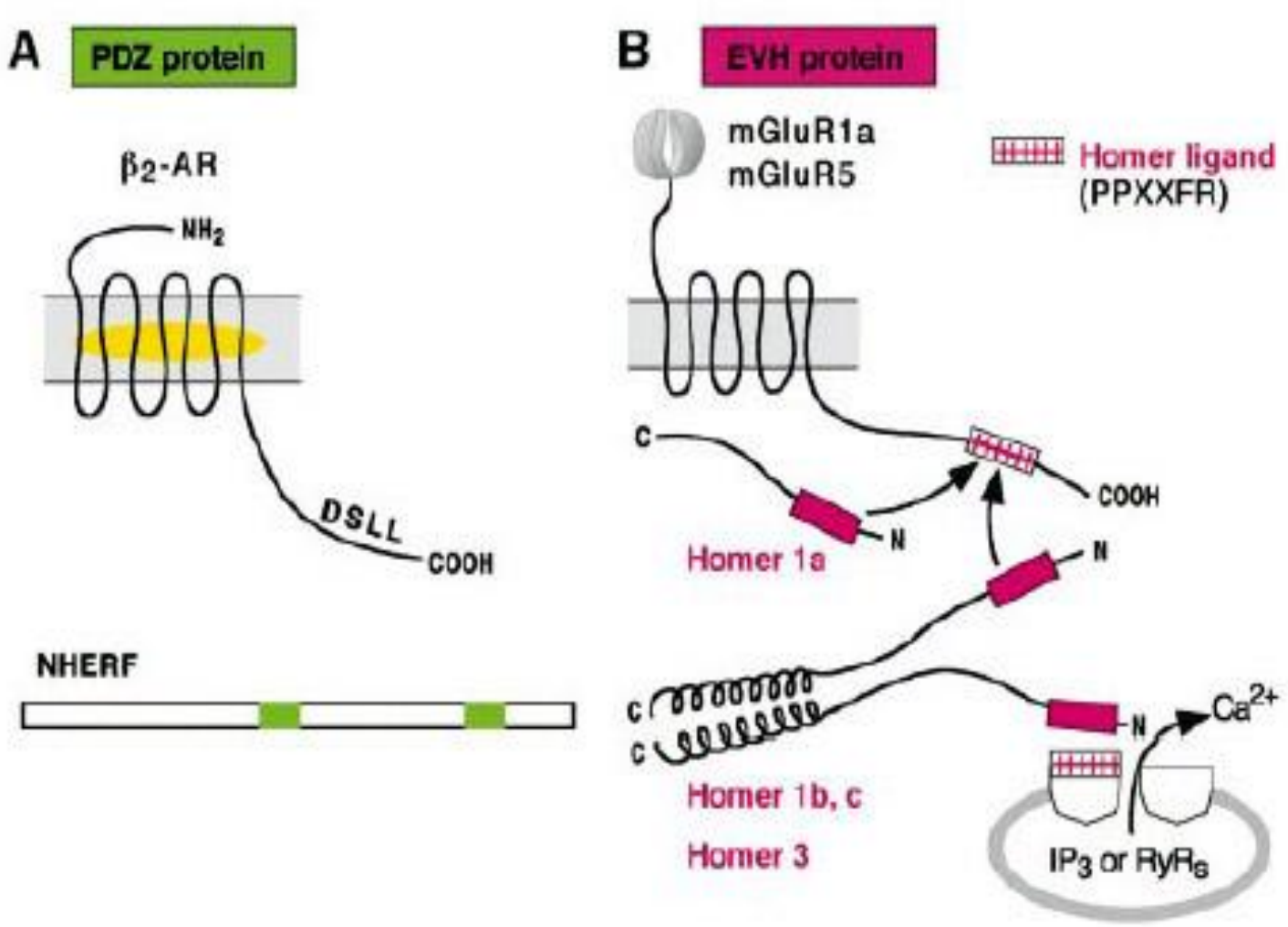
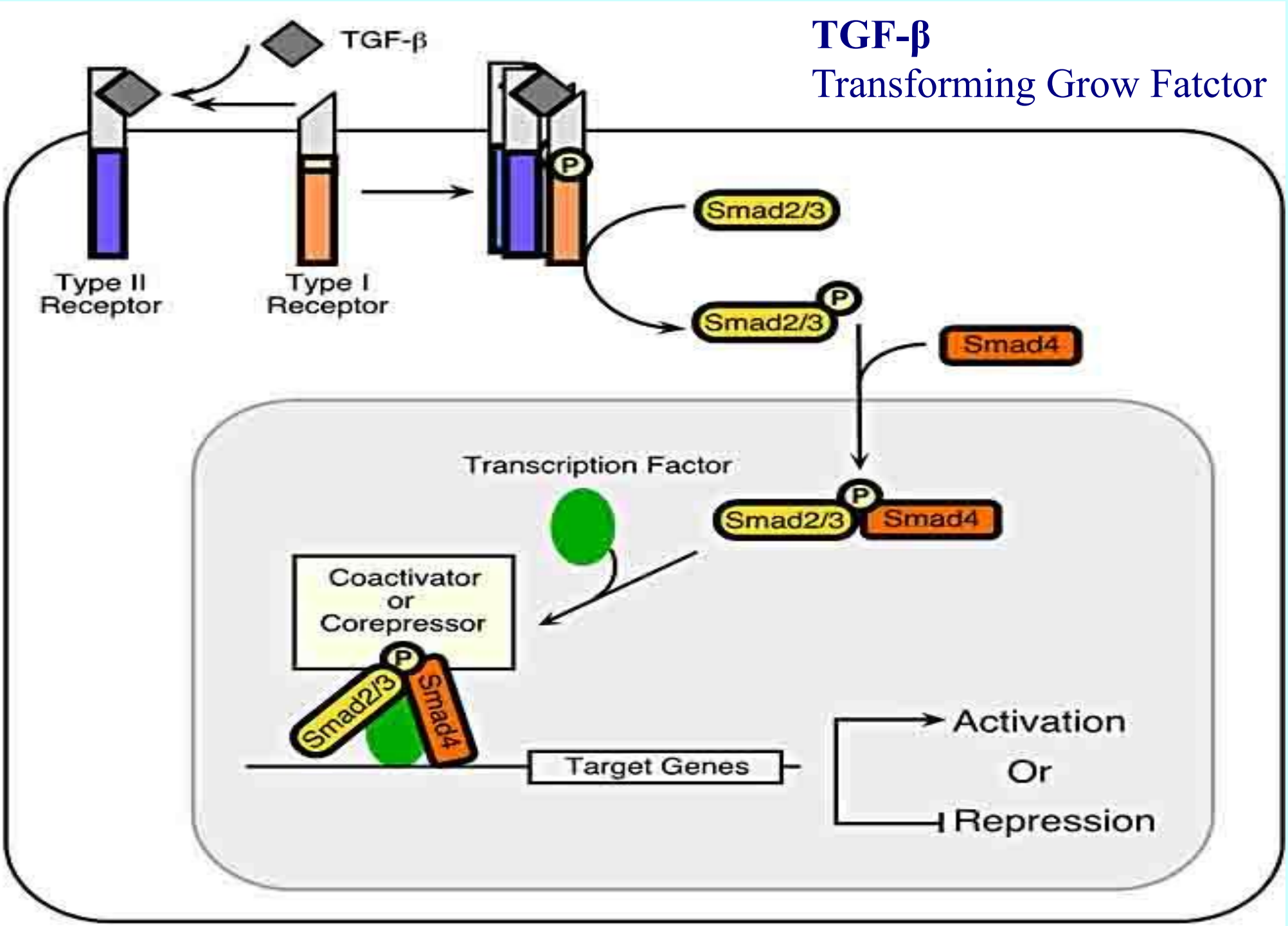


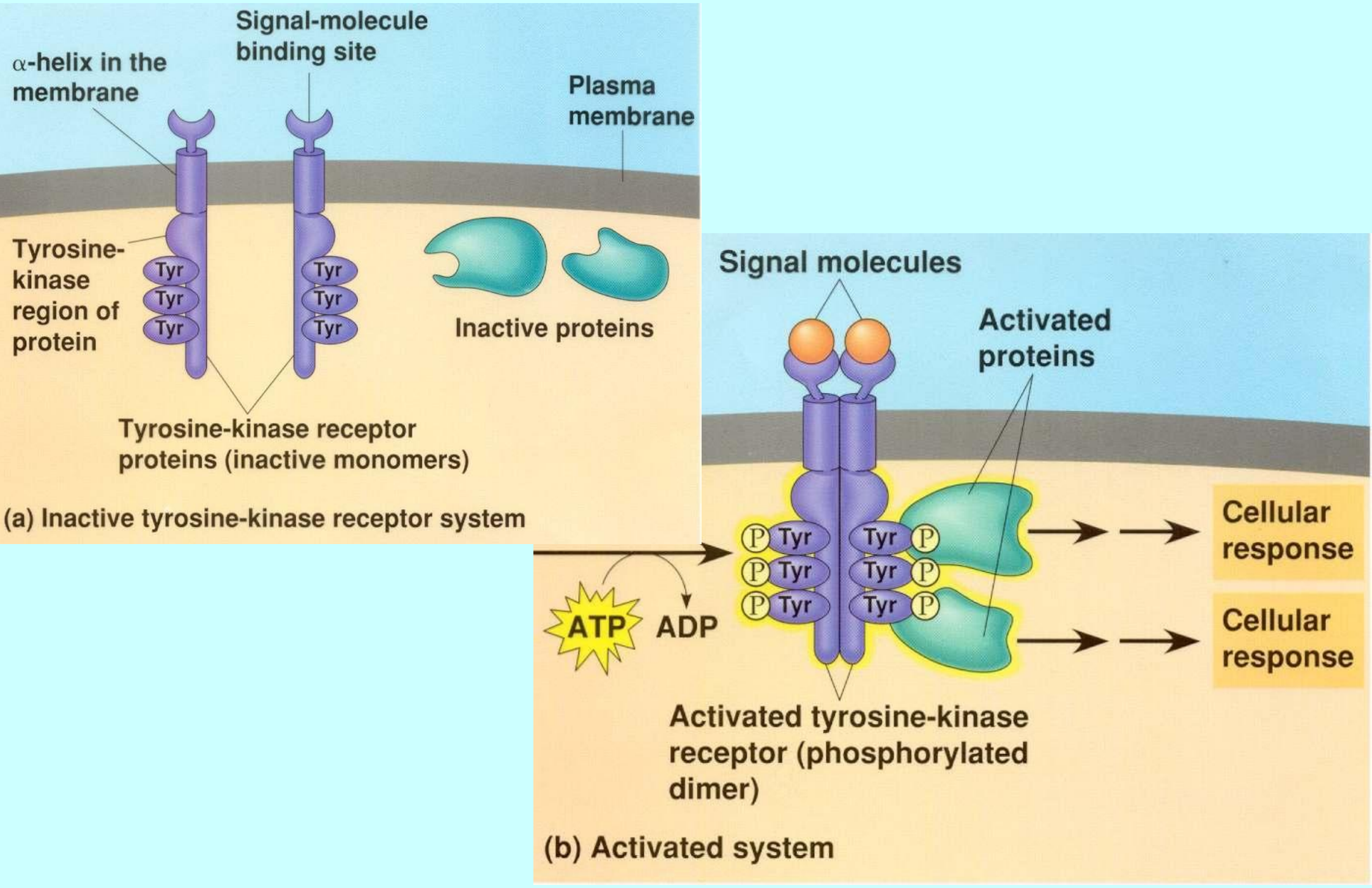
Fig. 3. GPCRs are unfaithful to G proteins. Two examples of transduction triggered via a direct interaction of GPCRs with proteins containing PDZ and EVH-like domains. (**A**) β_2-AR is unfaithful to G proteins in establishing a direct interaction between their C-terminal domain (DSSL) and a PDZ domain of the Na^+/H^+ exchanger regulatory factor (NHERF) (Hall *et al.*, 1998). (**B**) The mGluR1a and mGluR5 are unfaithful to G proteins in establishing a direct interaction between their Homer ligand sequence (PPXXP) and the EVH-like domain of Homer proteins. Homer 1b, 1c, 2 and 3, but not Homer 1a, encode a C-terminal CC domain that confers the possibility of self-multimerization. The EVH domain of Homer proteins also interact with the Homer ligand of IP3 and ryanodine receptors (RyRs). Homer 1a blocks the association of mGluRs with the CC-Homer complexes (Tu *et al.*, 1998; Xiao *et al.*, 1998).

RECETTORI SER/THR KINASE

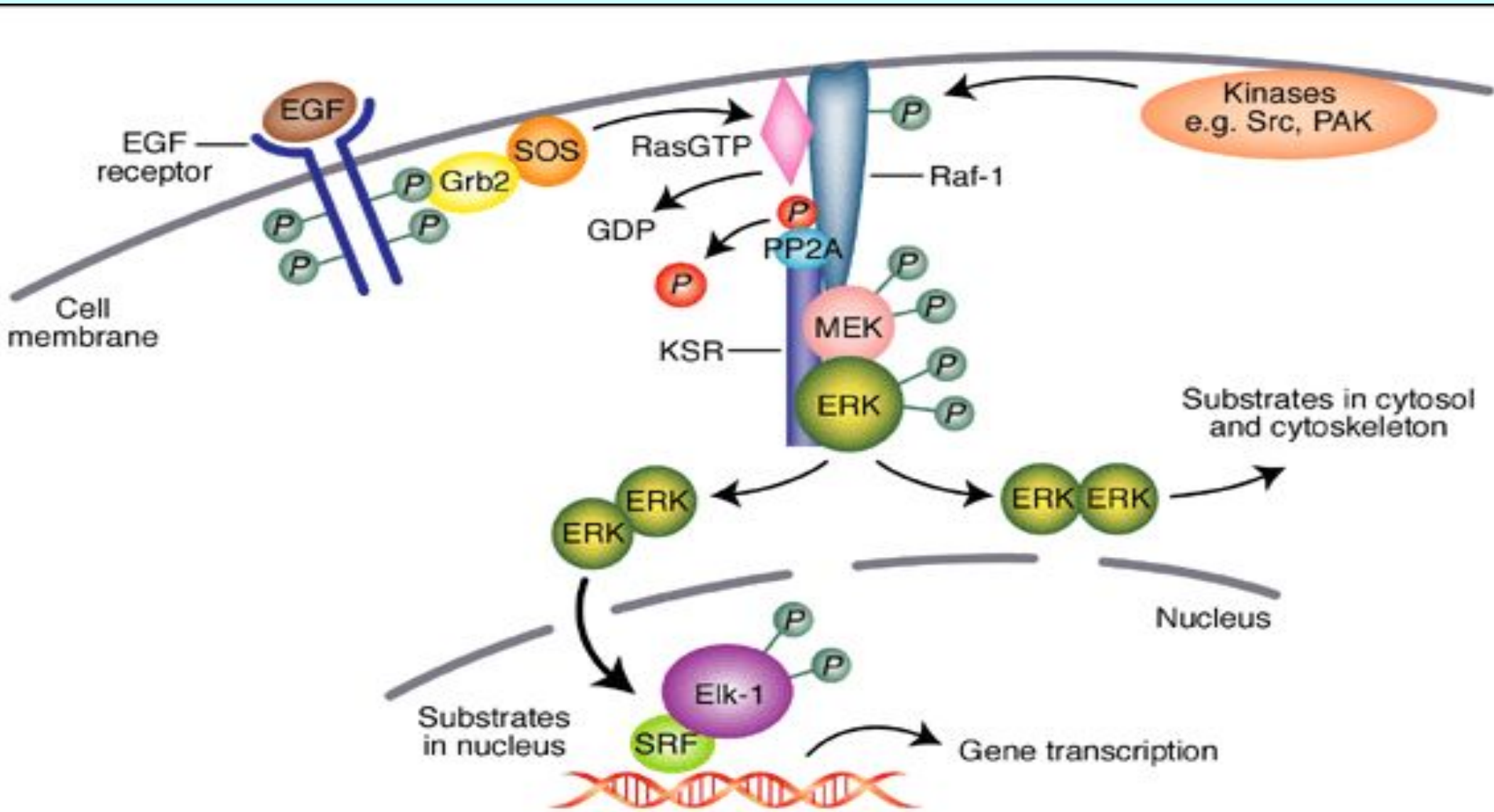
TGF-β
Transforming Grow Fatctor



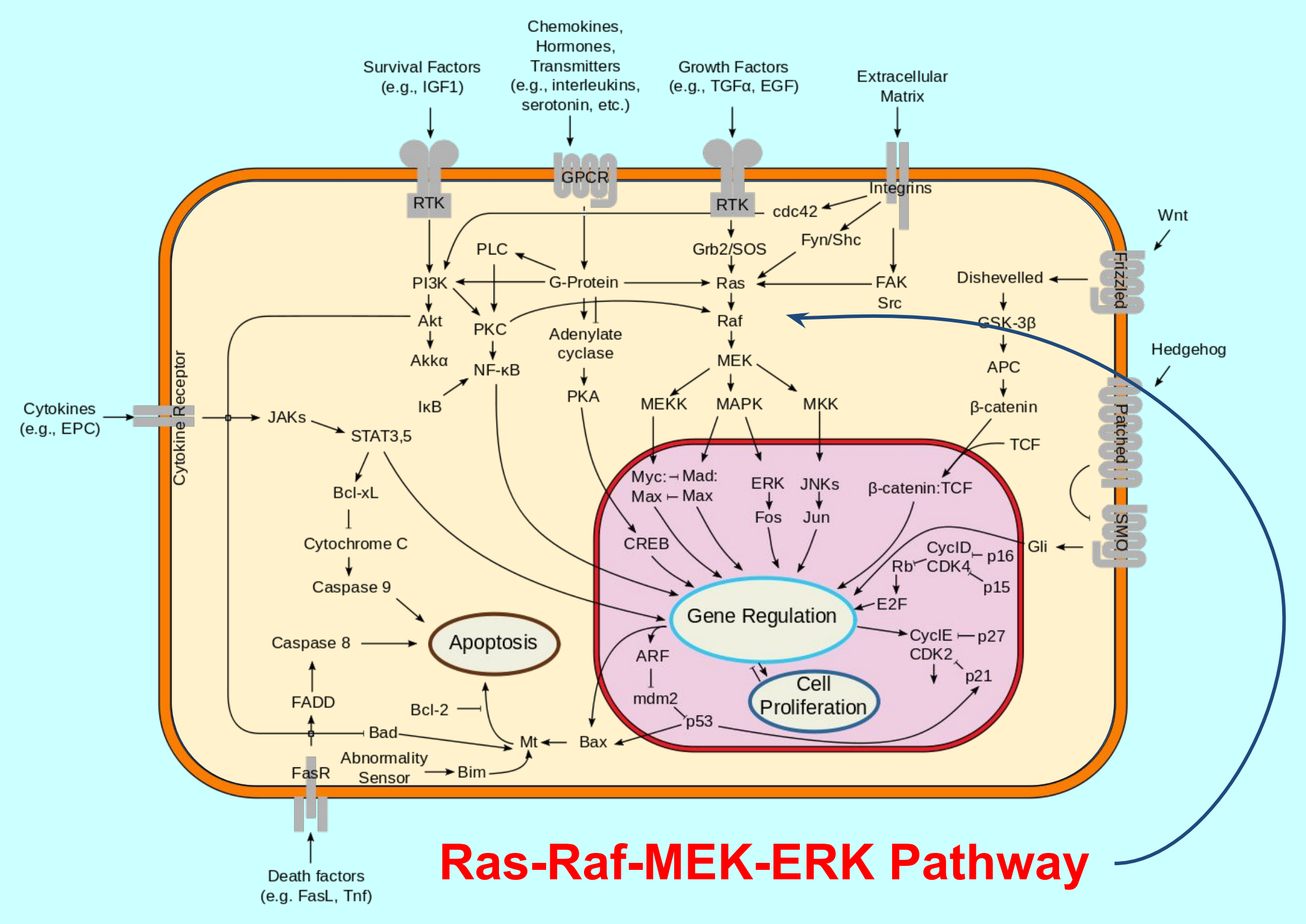
RECETTORI TYR KINASE



RECETTORI TYR KINASE



The organisation and function of the Ras-Raf-MEK-ERK pathway



Ras-Raf-MEK-ERK Pathway

TRANSATTIVAZIONE

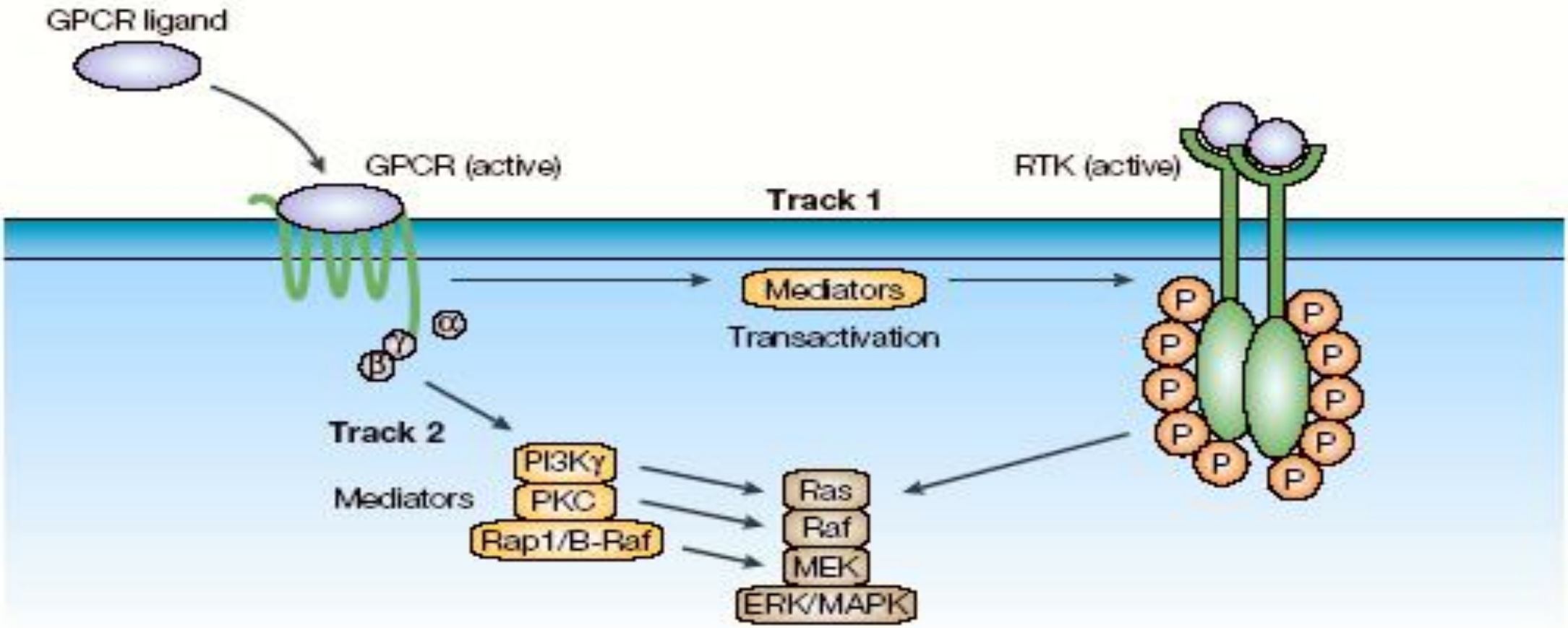


Figure 4 | **Activation of ERK/MAPK through parallel tracks.** G-protein-coupled receptors (GPCRs) can also activate the extracellular signal-regulated kinase (ERK)/mitogen-activated protein kinase (MAPK) pathway in the absence of receptor tyrosine kinase (RTK) activation. Mediators in this pathway can, for example, be phosphatidylinositide 3-kinase (PI3Kγ), which activates the cascade by an unknown mechanism involving Ras. Protein kinase C (PKC) isoforms can activate Raf by direct phosphorylation. Through Rap1 or B-Raf, GPCRs can also activate MAPK and ERK kinase (MEK). In many cell types, parallel pathways from GPCRs to ERK/MAPK activation exist, which include the RTK transactivation process. We designate this phenomenon, which is highly cell-type dependent, 'multi-track signalling'.

RECETTORI PAR

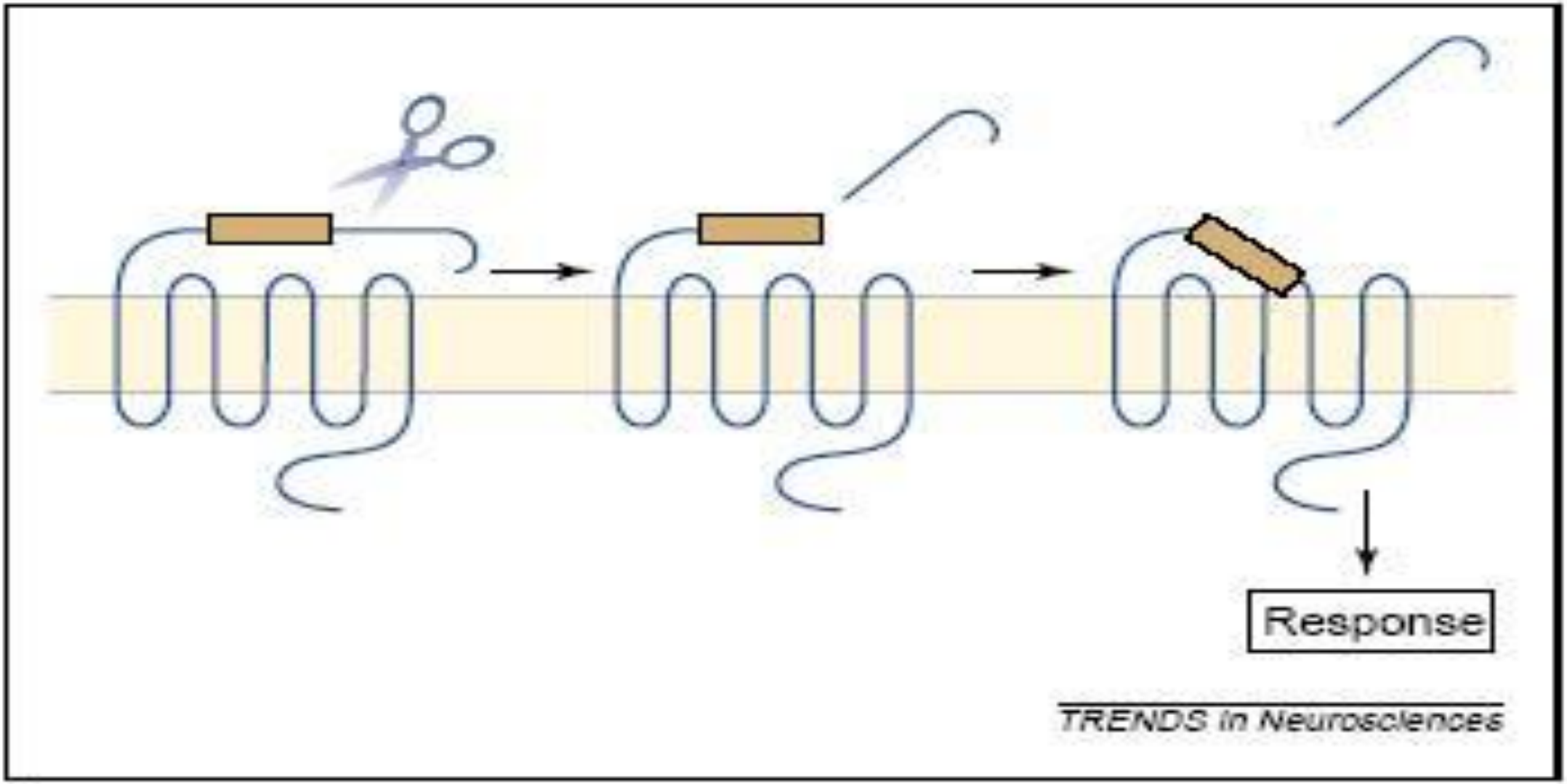
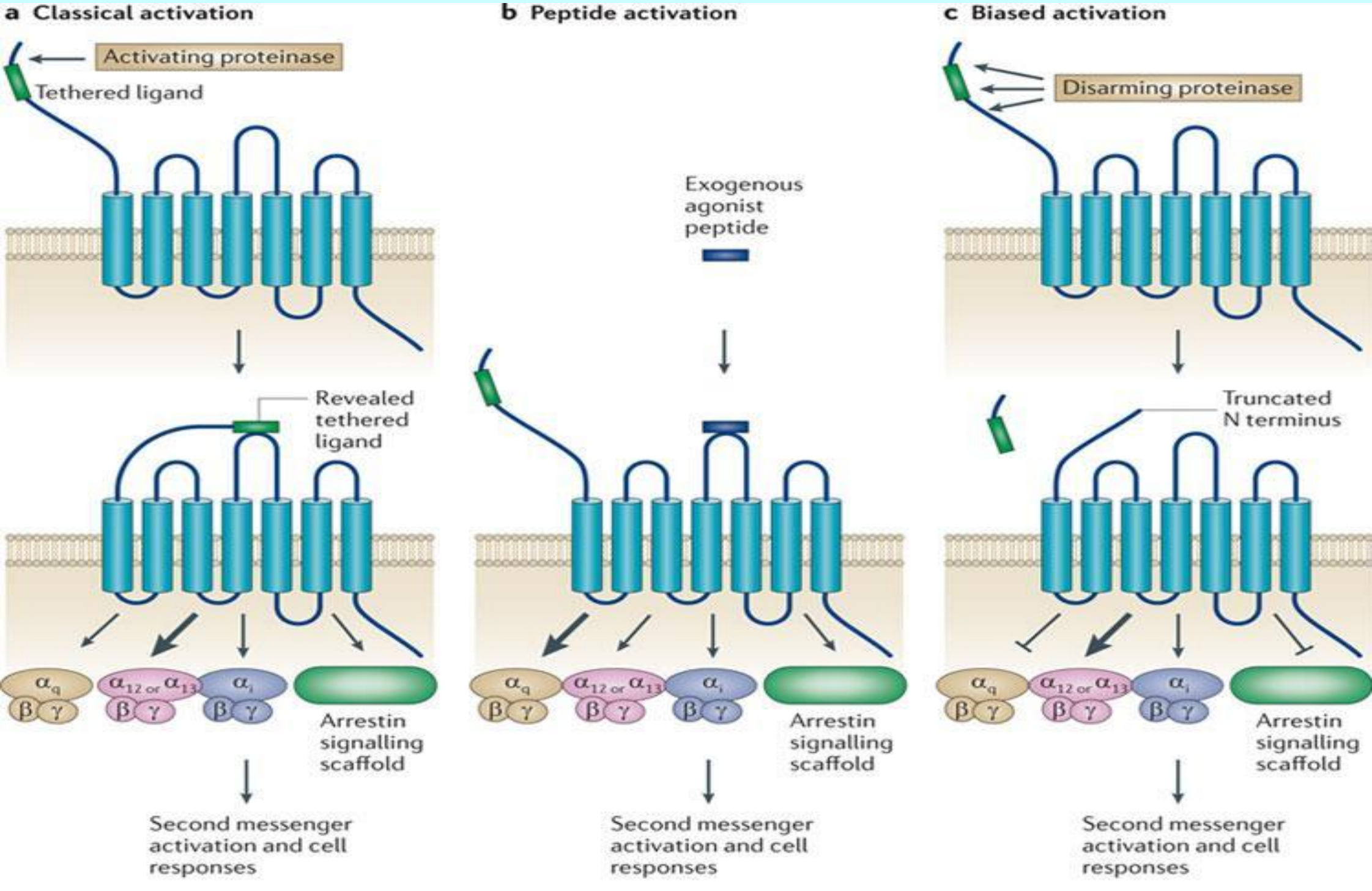
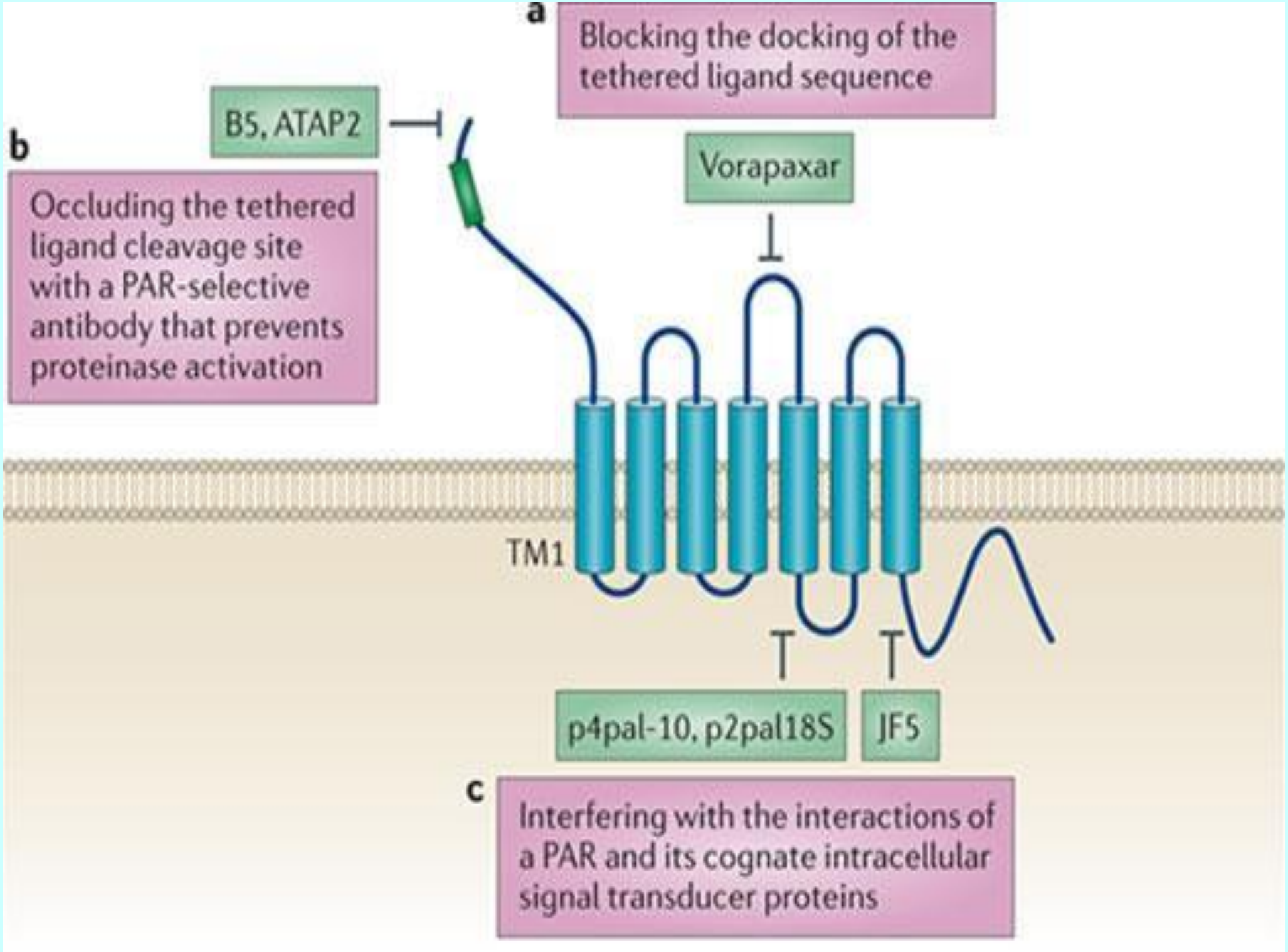


Fig. 1. Mechanism of activation of proteinase-activated receptors (PARs). Proteinases (represented by scissors) at the cell surface cleave the N-terminal domain of the receptor, thus exposing a new N-terminal domain, which binds and activates the receptor, inducing an intracellular signal.

RECETTORI PAR



RECETTORI PAR



TRATTAZIONE QUANTITATIVA DELL'AZIONE DI LIGANDI SU RECETTORI

La maggior parte delle interazioni farmaco-recettore può essere descritta con un'equazione analoga all'equazione di Michaelis-Menten usata per descrivere le interazioni enzima-substrato

$$D + R \underset{k_2}{\overset{k_1}{\rightleftharpoons}} DR \xrightarrow{k_3} \text{Effect}$$

TRATTAZIONE QUANTITATIVA DELL'AZIONE DI LIGANDI SU RECETTORI

Un farmaco (D) forma un complesso con un recettore (R)

$$R + D \xrightarrow{K_D} RD$$

All'equilibrio

$$K_D = \frac{(R)\ (D)}{(RD)}$$

dove K_D è la costante di dissociazione del complesso farmaco-recettore.
Se R_T è la quantità totale del recettore:

$$R = R_T - RD \text{ e } \frac{(R_T - RD)\ (D)}{(RD)} = K_D$$

Riarrangiando algebricamente:

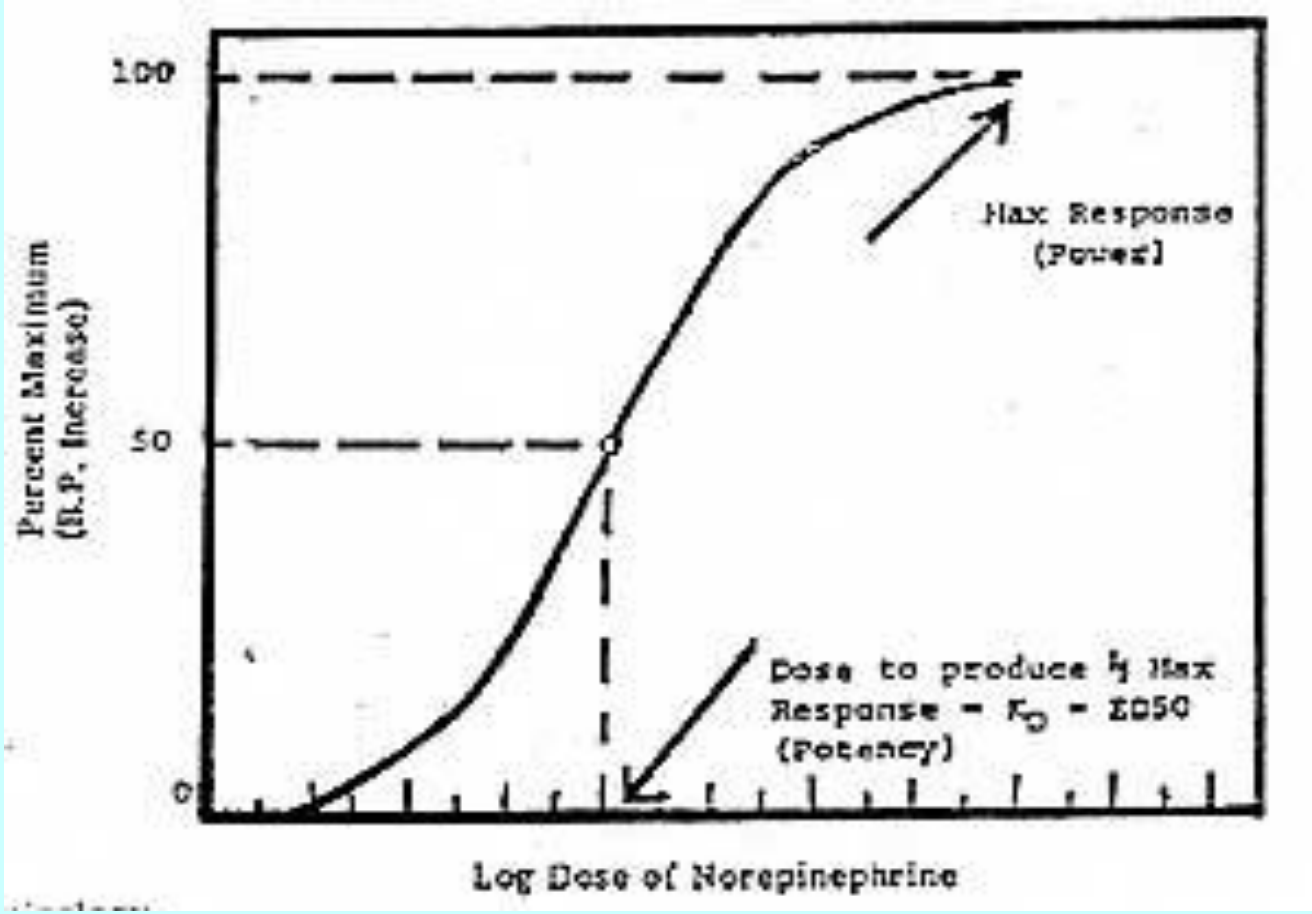
$$\frac{(RD)}{(R_T)} = \frac{(D)}{K_D + (D)}$$

Secondo il modello di occupazione dei recettori, l'entità della risposta è direttamente proporzionale alla frazione di farmaco legato (RD/R_T) e la risposta massima (E_{max}) è proporzionale a R_T. Perciò la frazione di risposta massima prodotta:

$$\frac{e}{E_{max}} = \frac{(D)}{K_D + (D)}$$

TRATTAZIONE QUANTITATIVA DELL'AZIONE DI LIGANDI SU RECETTORI

Tipica curva Log Dose-Risposta



Le curve log dose-risposta vengono usate durante lo sviluppo preclinico per comparare la potenza (in termini di ED_{50}) di analoghi differenti rispetto al lead compound.

TRATTAZIONE QUANTITATIVA DELL'AZIONE DI LIGANDI SU RECETTORI

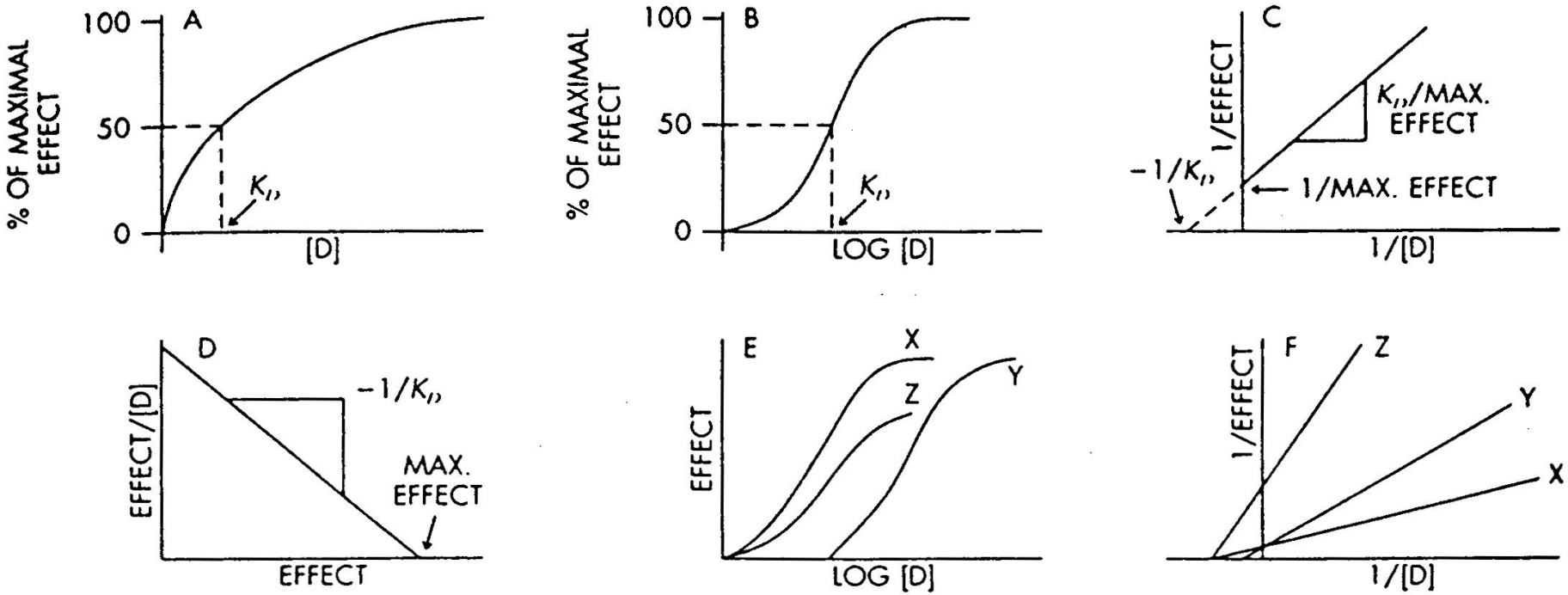


Figure 2–6. *Different representations of dose–effect curves.*

A. The ideal relationship between the concentration of a drug [D] and the magnitude of response to it. When plotted on a linear scale, a typical hyperbolic curve results.

B. A sigmoidal dose–effect curve results when the magnitude of effect observed is plotted versus the logarithm of the drug concentration.

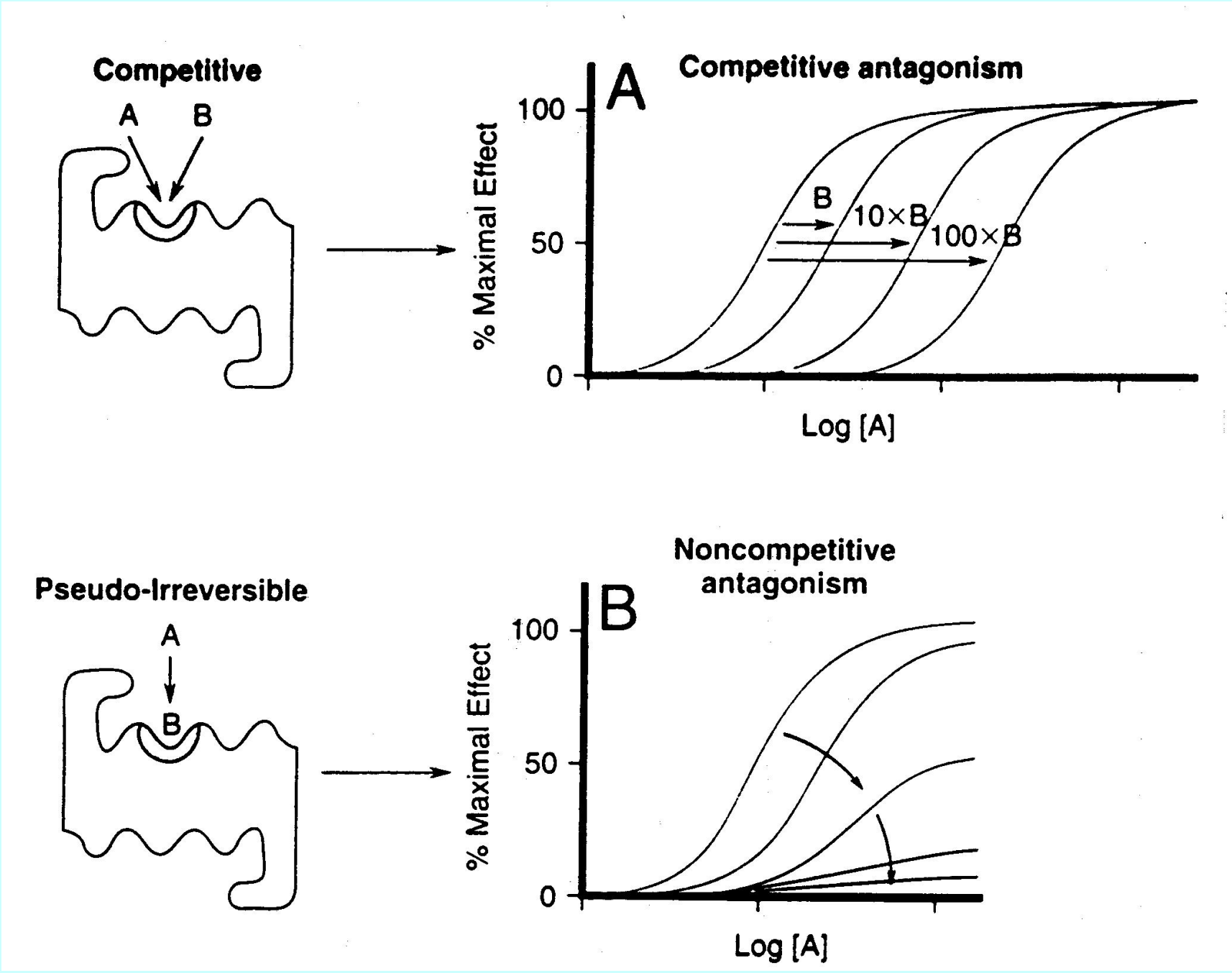
C and *D*. A double-reciprocal plot (*C*) and a Scatchard or Eadie–Hofstee type of plot (*D*) of the concentration dependence of drug effect

E and *F*. Representative log dose–effect curves (*E*) and double-reciprocal plots (*F*).

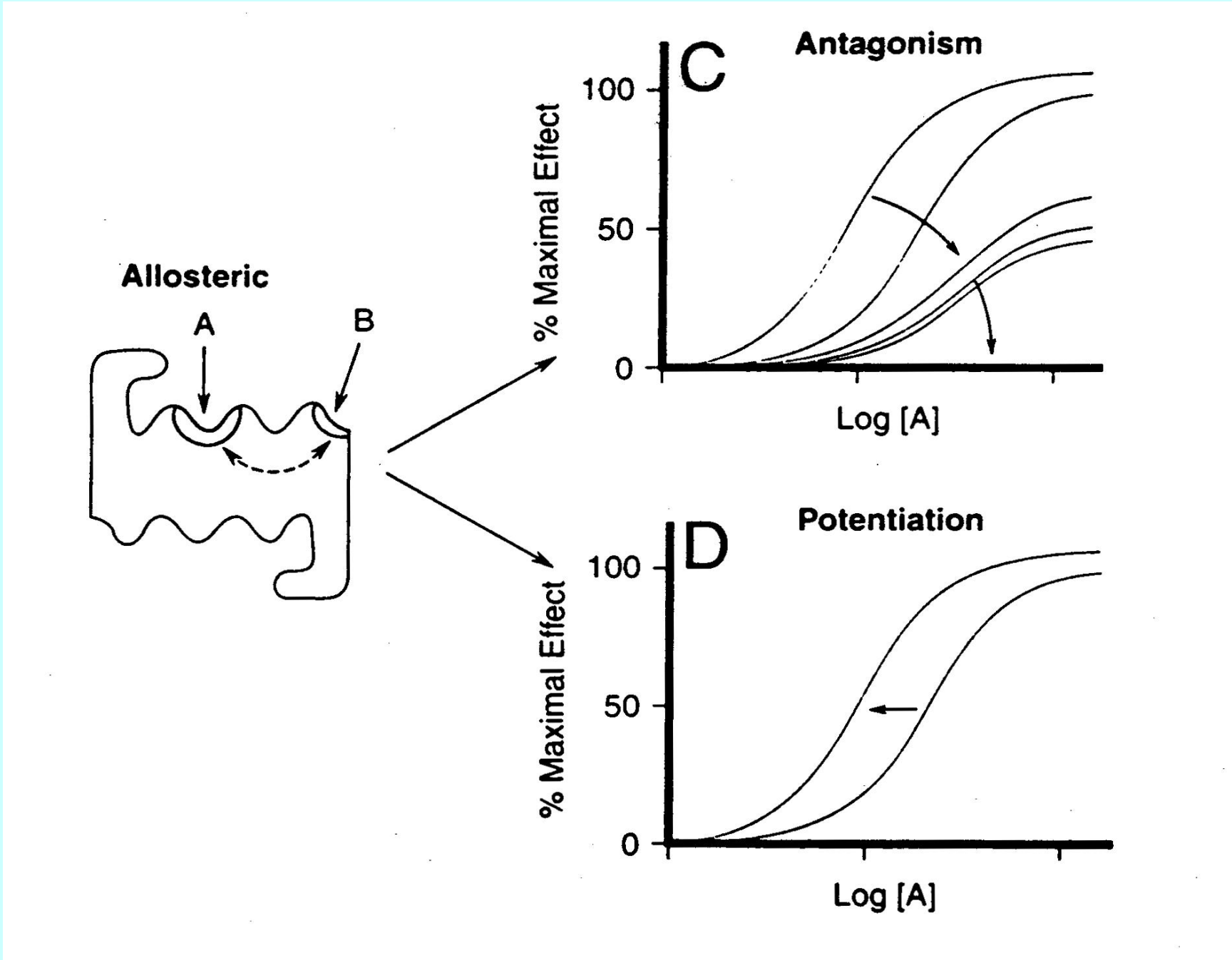
Curves X and Y. Two agonists with similar efficacy but differing in potency (X more potent than Y) *or* an agonist in the absence (X) and presence (Y) of a competitive antagonist.

Curves X and Z. Two agonists with similar potency but differing in efficacy (full agonist, X, and partial agonist, Z) *or* an agonist in the absence (X) and presence (Z) of a noncompetitive antagonist.

TRATTAZIONE QUANTITATIVA DELL'AZIONE DI LIGANDI SU RECETTORI



TRATTAZIONE QUANTITATIVA DELL'AZIONE DI LIGANDI SU RECETTORI



TRATTAZIONE QUANTITATIVA DELL'AZIONE DI LIGANDI SU RECETTORI

Termini che indicano l'abilità di un farmaco a legare il recettore:

- Potenza
- Affinità
- K_D o ED_{50} (corrispondente alla K_m in analogia con Michaelis-Menten)

TRATTAZIONE QUANTITATIVA DELL'AZIONE DI LIGANDI SU RECETTORI

Termini che indicano l'abilità di un farmaco a produrre una risposta:

- Efficacia
- Potenza
- Attività intrinseca (α) (Ariens, 1950's):

$$\alpha = \frac{E_{max} \text{ di un farmaco di riferimento}}{E_{max} \text{ dell'agonista più attivo nella stessa serie strutturale}}$$

α = 1; ligandi del recettore con uguale affinità
α = 0; il ligando è un antagonista puro
0<a<1; parziali agonisti

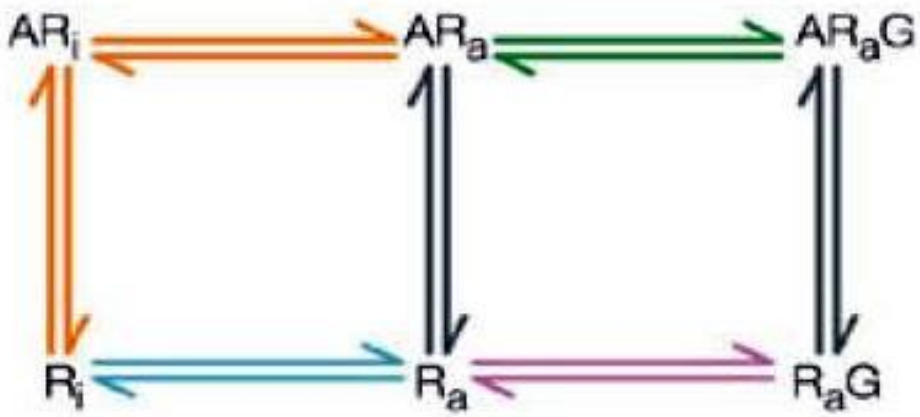
a Simple binding and activation

$A + R_i \rightleftharpoons AR_i \rightleftharpoons AR_a$

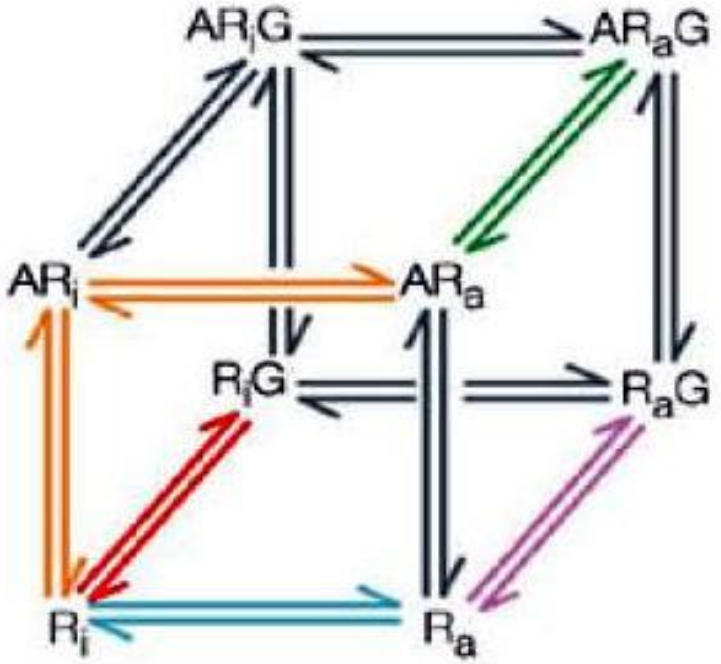
b Simple ternary complex model

$A + R_i \rightleftharpoons AR_i \rightleftharpoons AR_a + G \rightleftharpoons AR_aG$

c Extended ternary complex model



d Cubic ternary complex model



Ligand-mediated receptor activation

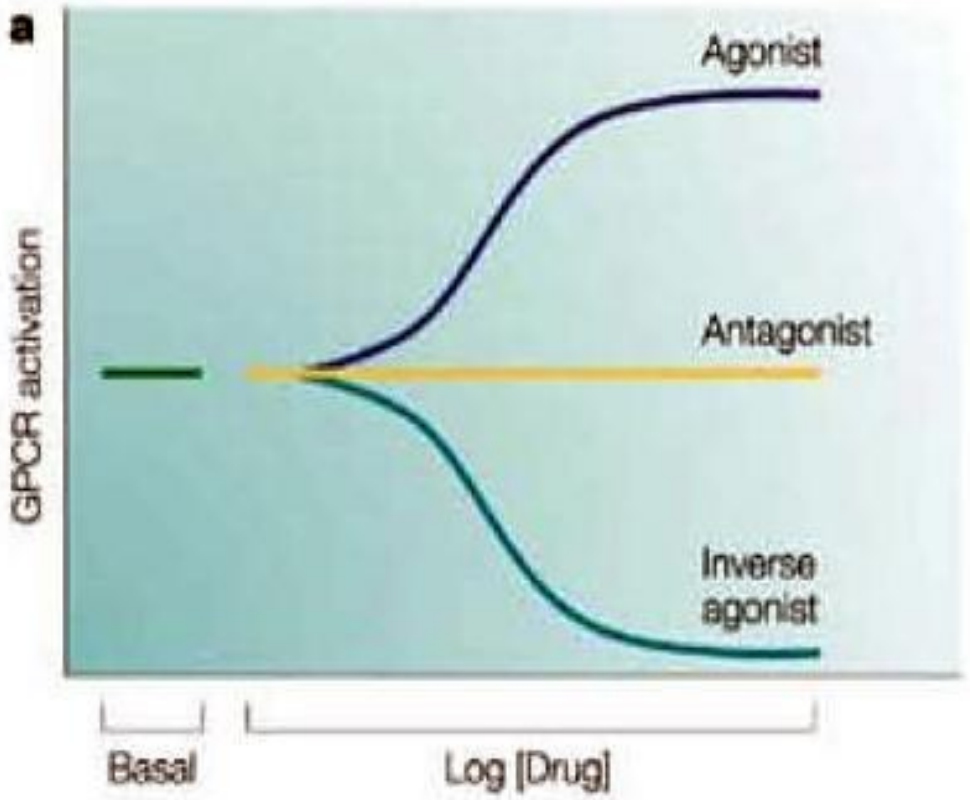
Receptor activation by allosteric modulation

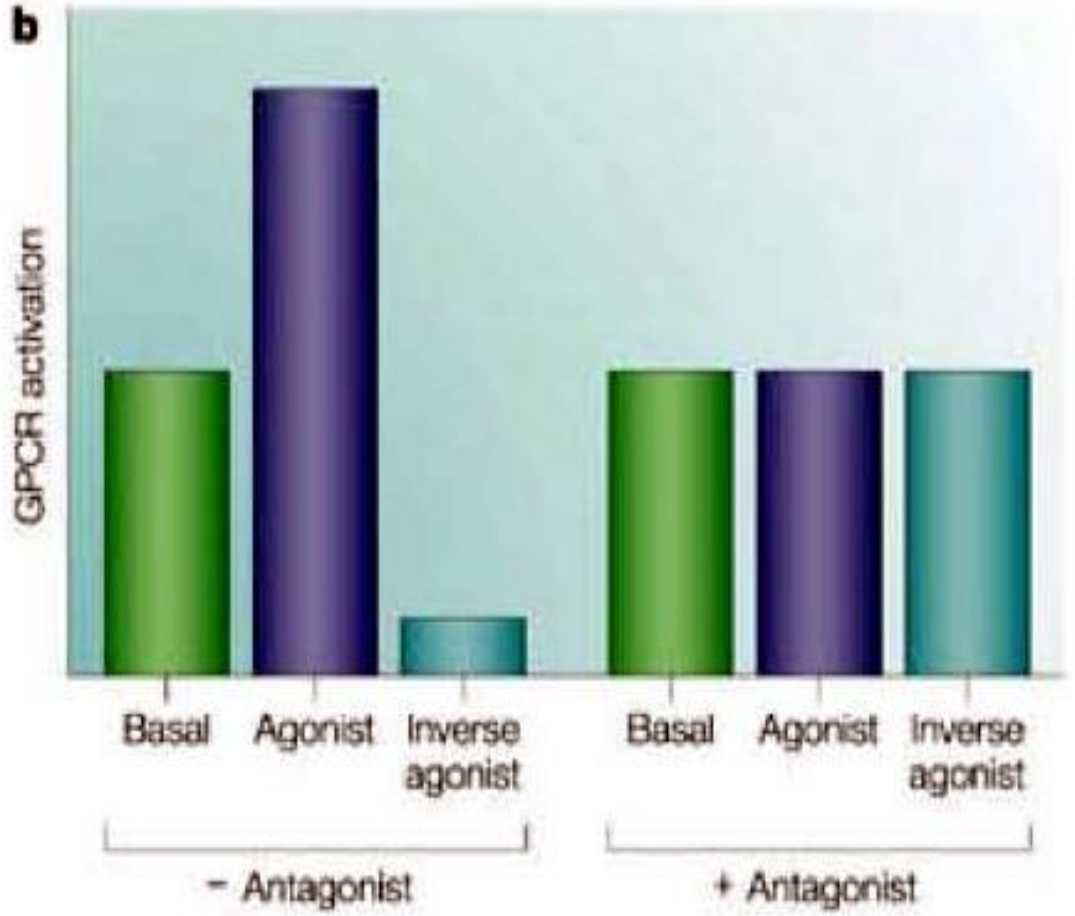
Activation of G protein in absence of ligand

Activation of G protein in presence of ligand

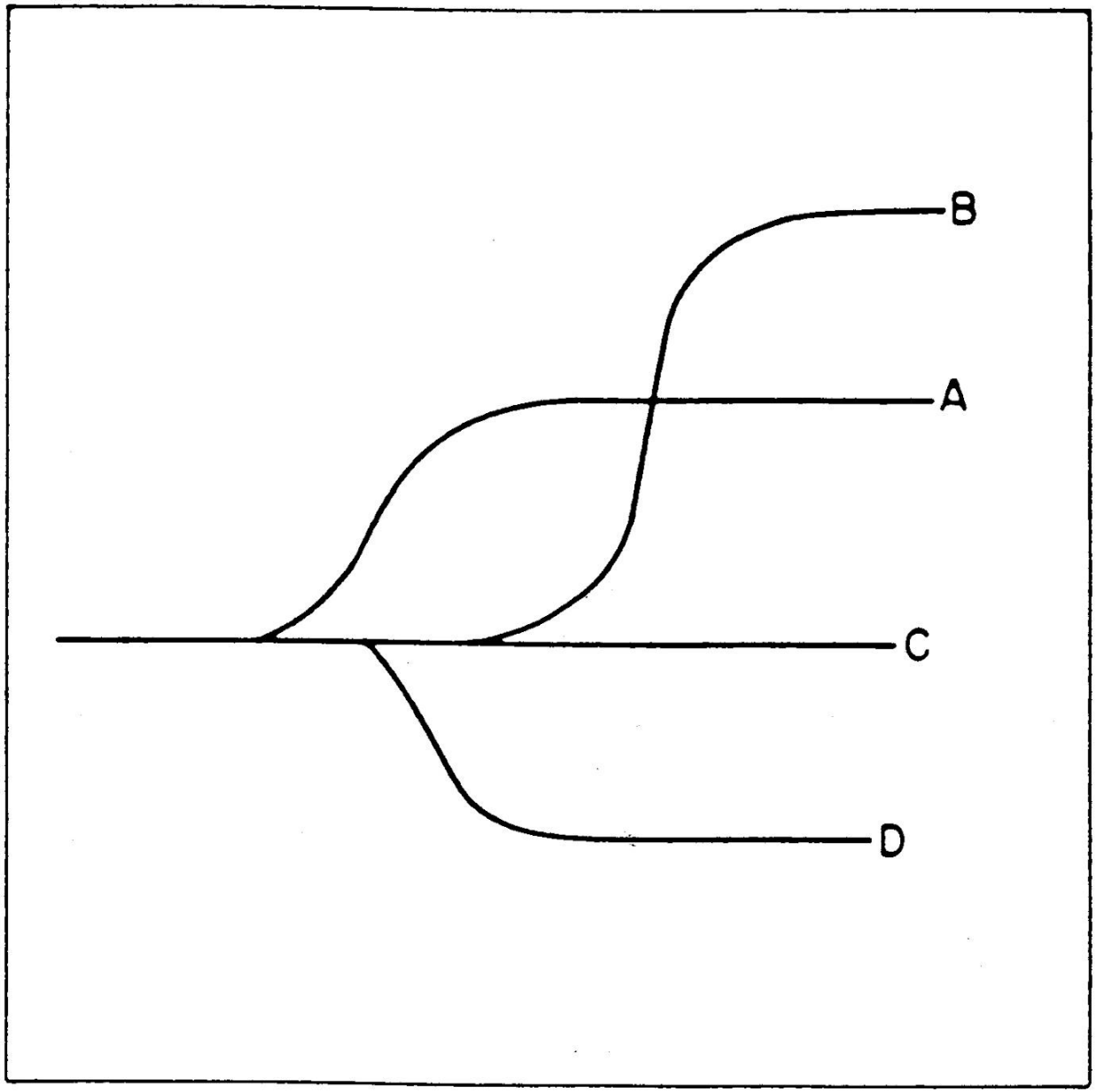
Inactive receptor coupled with G protein

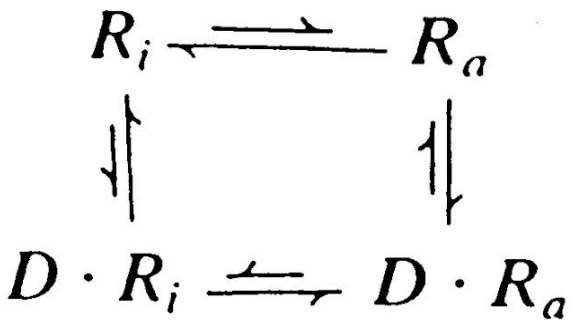
AGONISMO, ANTAGONISMO E AGONISMO INVERSO





TRATTAZIONE QUANTITATIVA DELL'AZIONE DI LIGANDI SU RECETTORI





A – Potenza (K) per $R_a > R_i$ e alta affinità

B – K per $R_a >> R_i$ e minore affinità

C – K per $R_a = R_i$

D – K per $R_a < R_i$