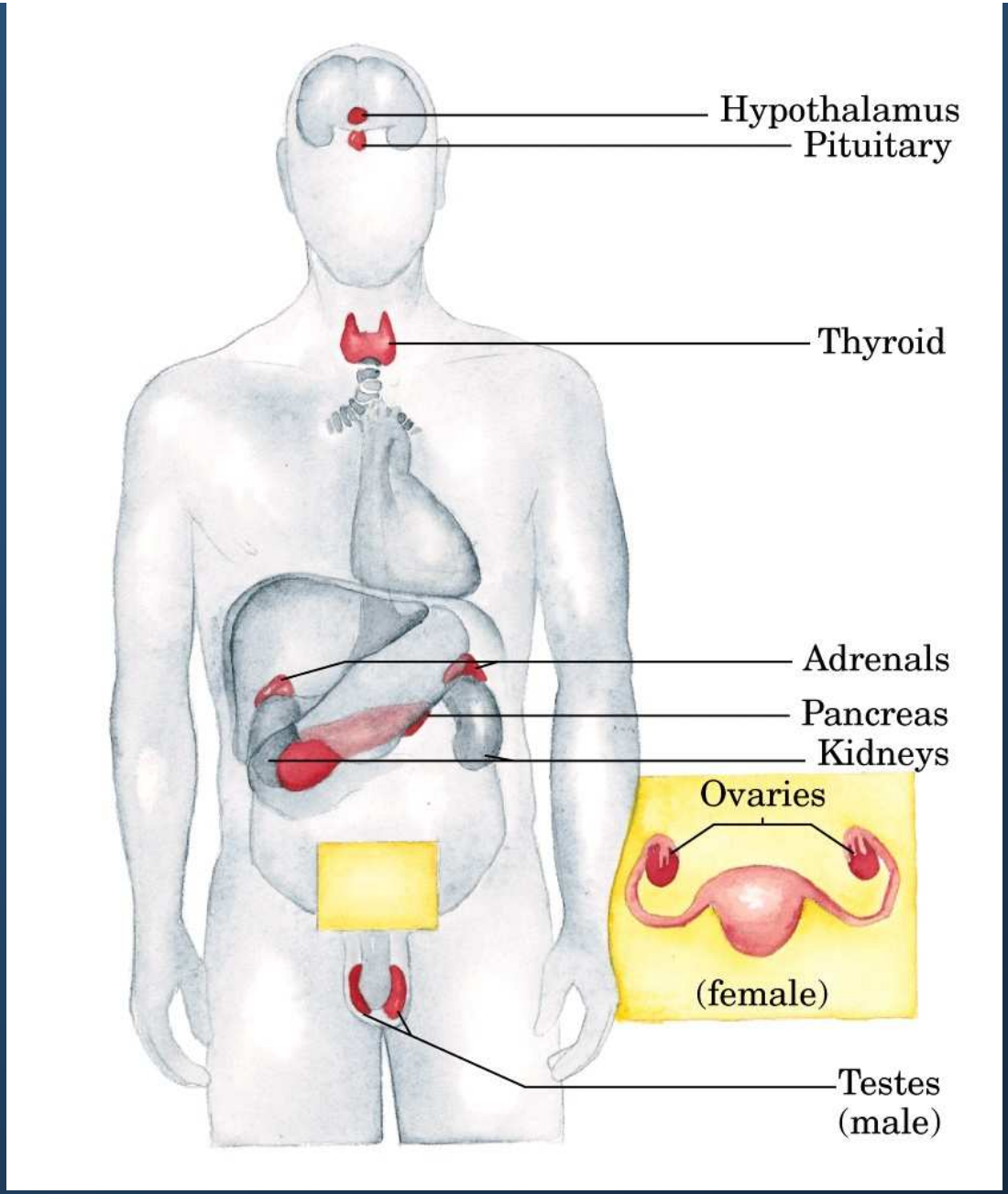
Sinalização celular:

Como as células se comunicam

Profa. Dra. Livia M. A. Tenuta



Sinais fisiológicos:

- **elétricos**
- **químicos**

75 trilhões de células

Tópicos a serem abordados

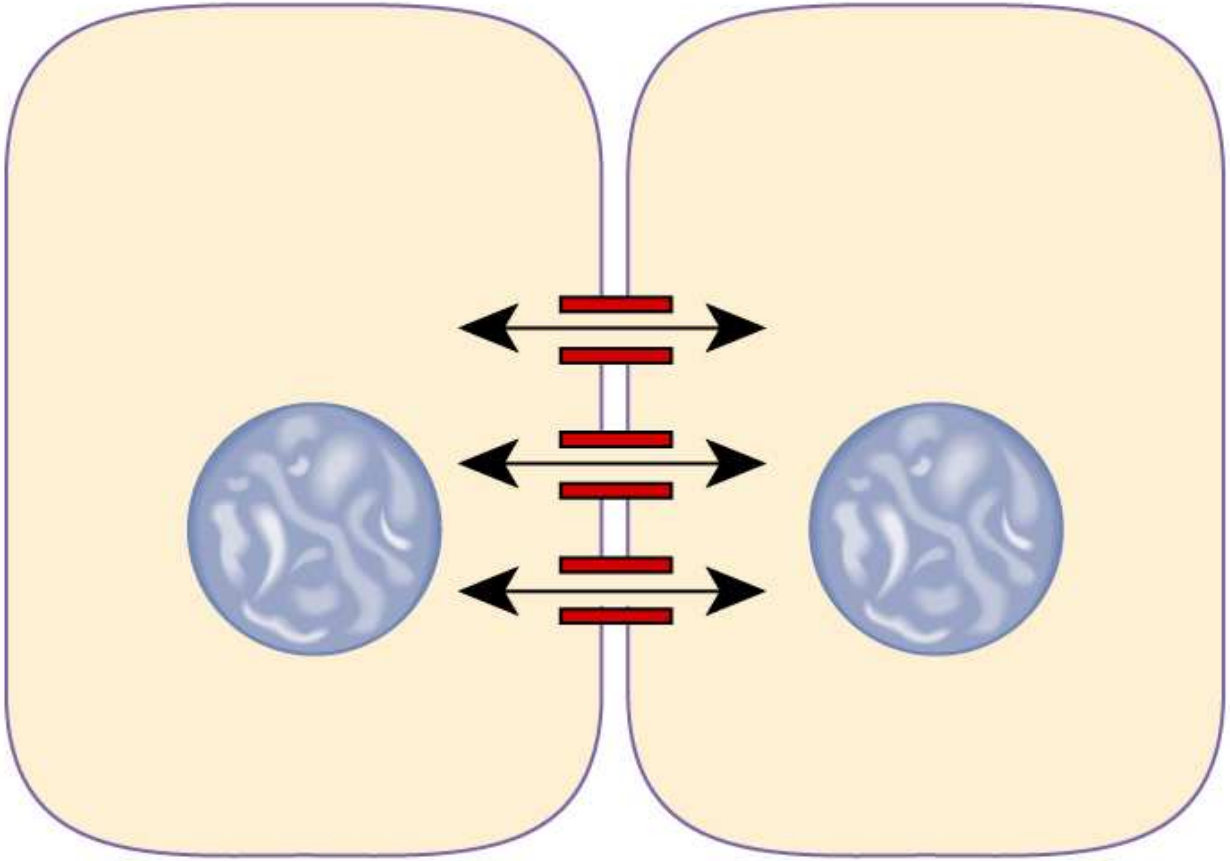
- Meios de comunicação celular
- Características e mecanismos moleculares da comunicação celular
- Tipos de transdutores de sinais e sua ação
  1) Canais iônicos
  2) Integrinas
  3) Receptores enzimáticos
  4) Receptores acoplados a proteínas G
- Regulação das vias de sinalização celular

Comunicação celular

1. Transferência direta de sinais elétricos e químicos através de junções comunicantes entre células adjacentes
2. Comunicação local por substâncias químicas que se difundem no meio extracelular
3. Comunicação à longa distância pela combinação de sinais elétricos transportados por células nervosas e sinais químicos transportados no sangue

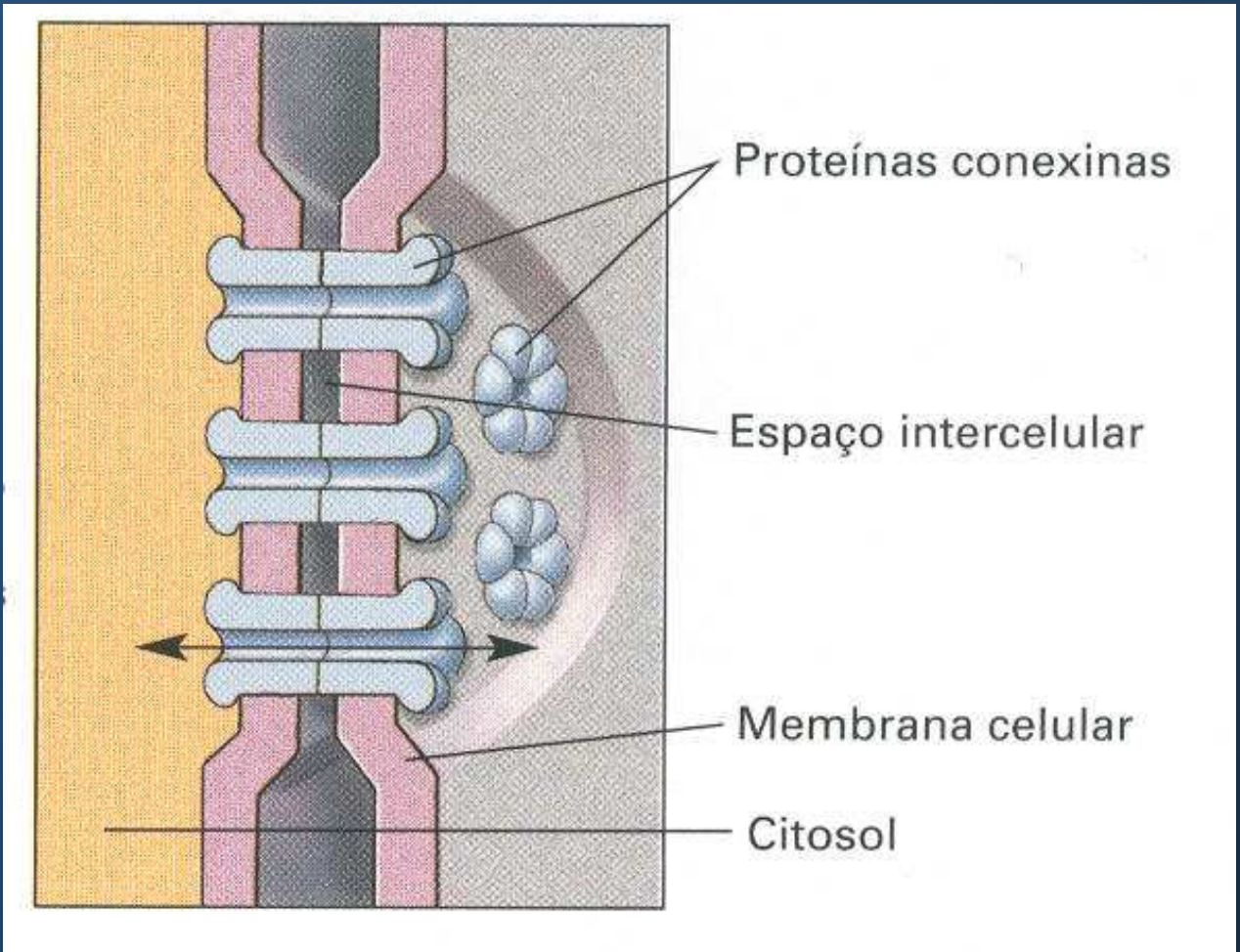
Meios de comunicação celular

Junções comunicantes



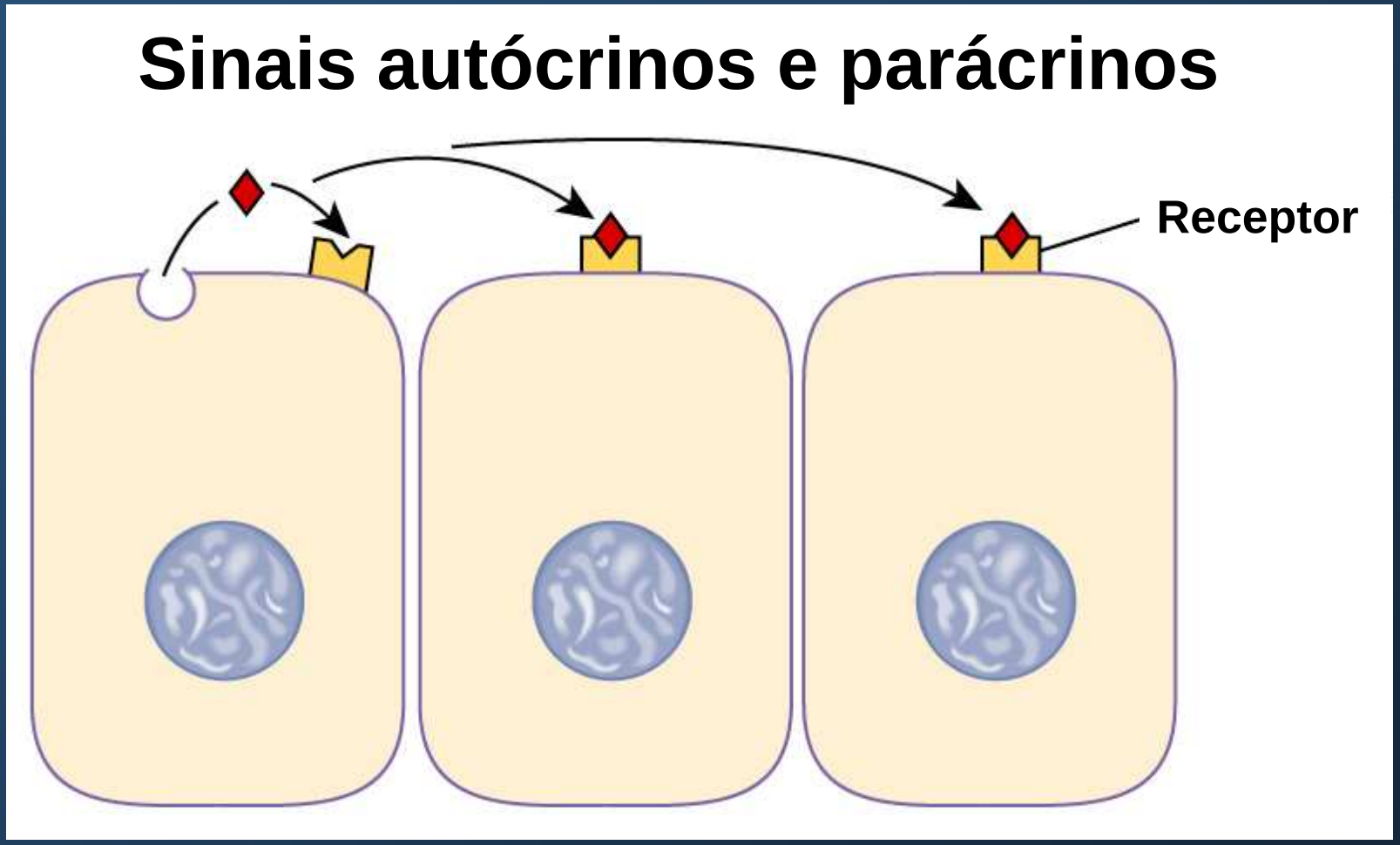
- Íons
- Pequenas moléculas
  - Aminoácidos, ATP, AMPc
- Sinais elétricos passam diretamente

Meios de comunicação celular



- **Músculo cardíaco, fígado, neurônios do cérebro, pâncreas, ovário, tireóide**

Meios de comunicação celular



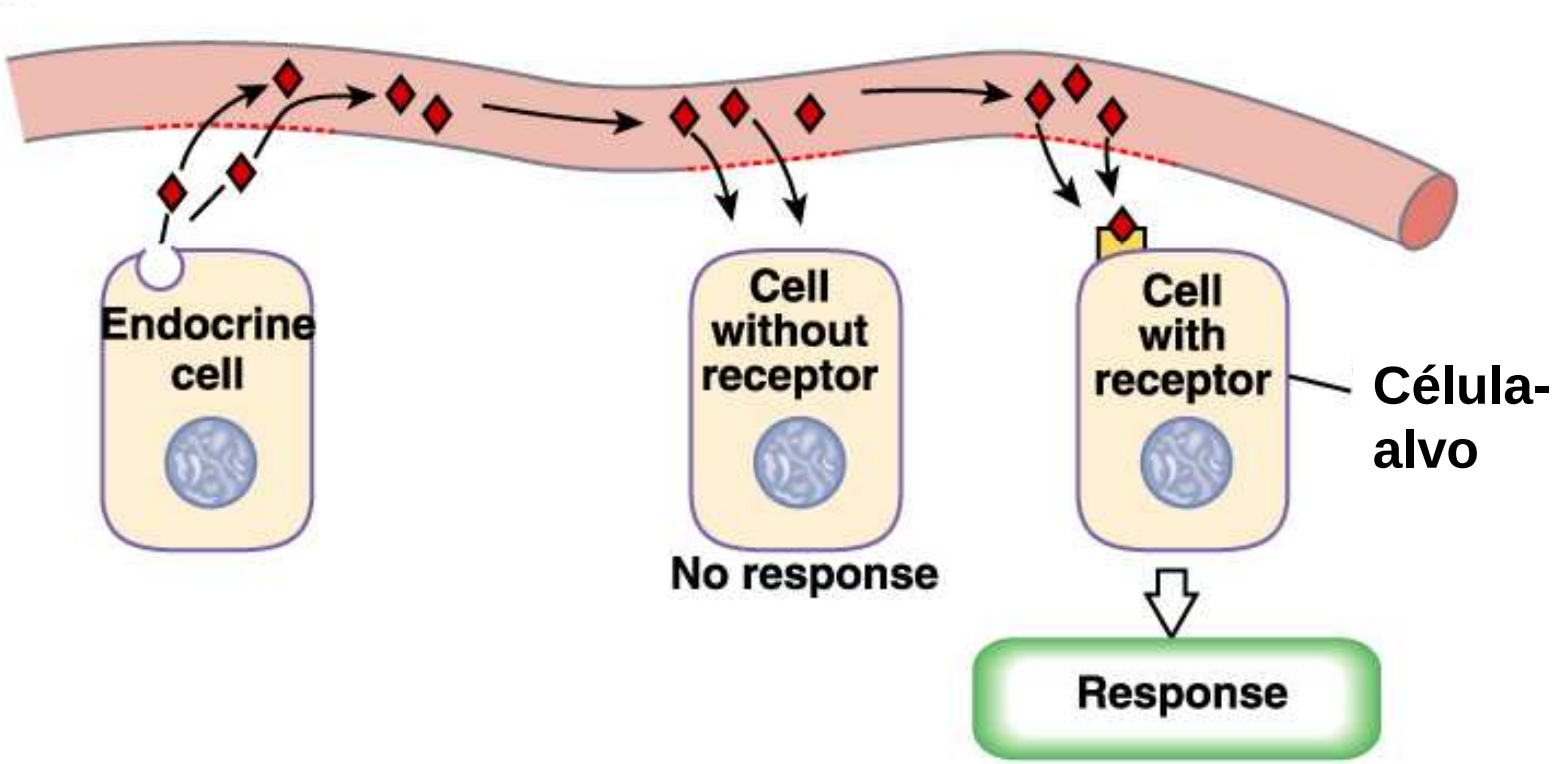

1

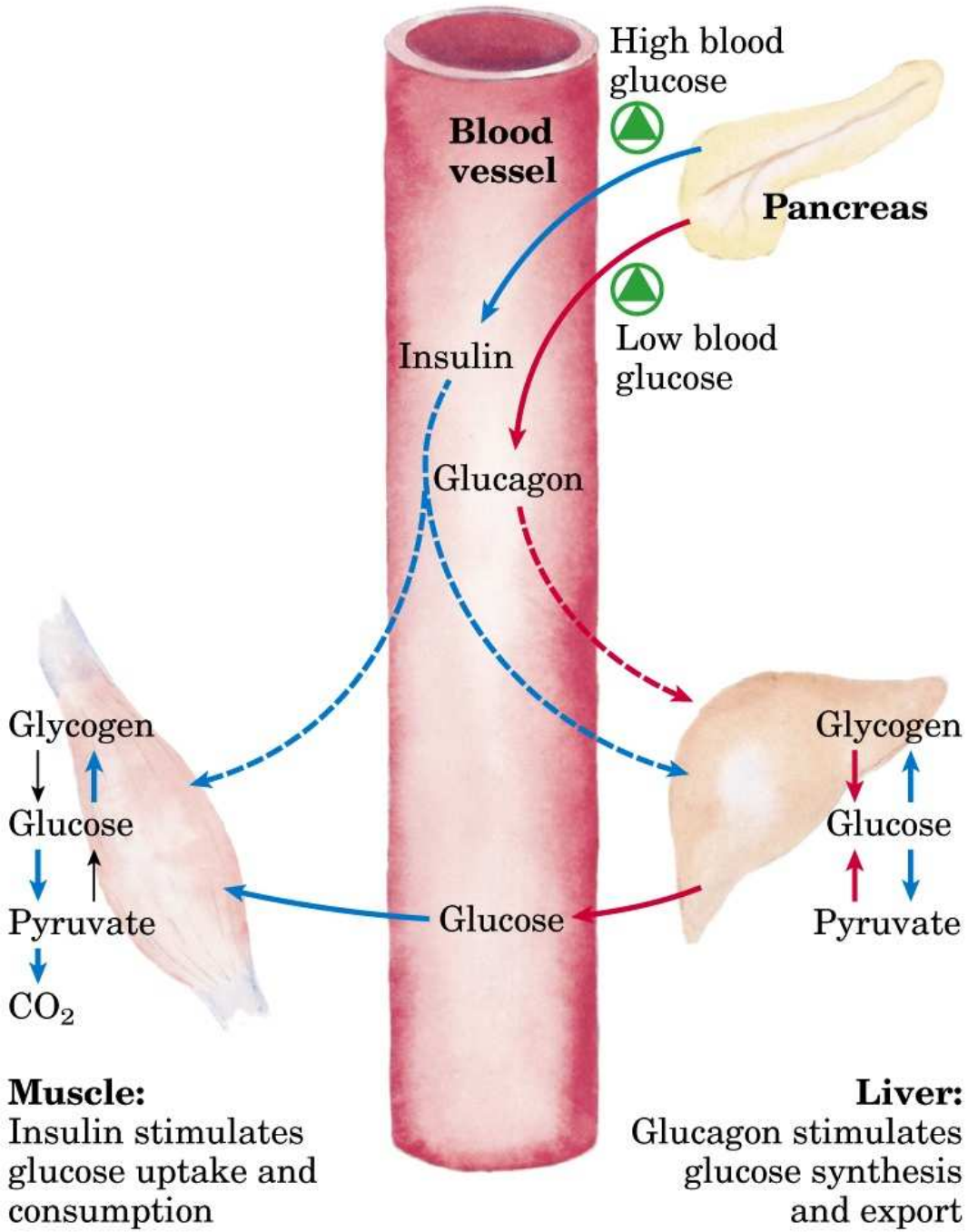
Substâncias autócrinas/parácrinas

- Neuromoduladores
- Citocinas
- Eicosanóides: prostaglandinas, tromboxanos e leucotrienos

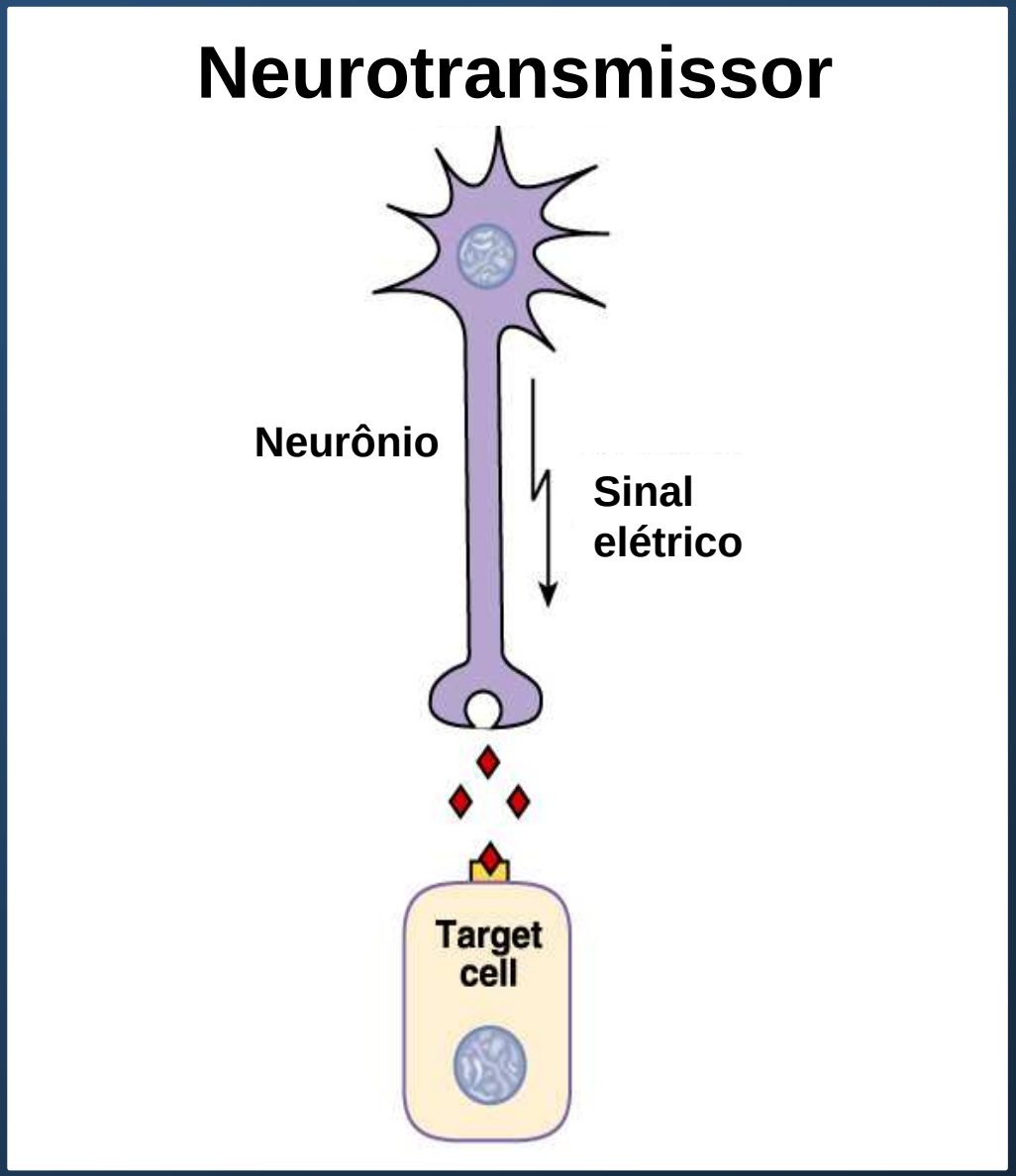
Meios de comunicação celular

Hormônios




High blood glucose
Blood vessel
Pancreas
Low blood glucose
Insulin
Glucagon
Glycogen
Glucose
Pyruvate
CO_2
Glycogen
Glucose
Pyruvate
Glucose
Muscle: Insulin stimulates glucose uptake and consumption
Liver: Glucagon stimulates glucose synthesis and export

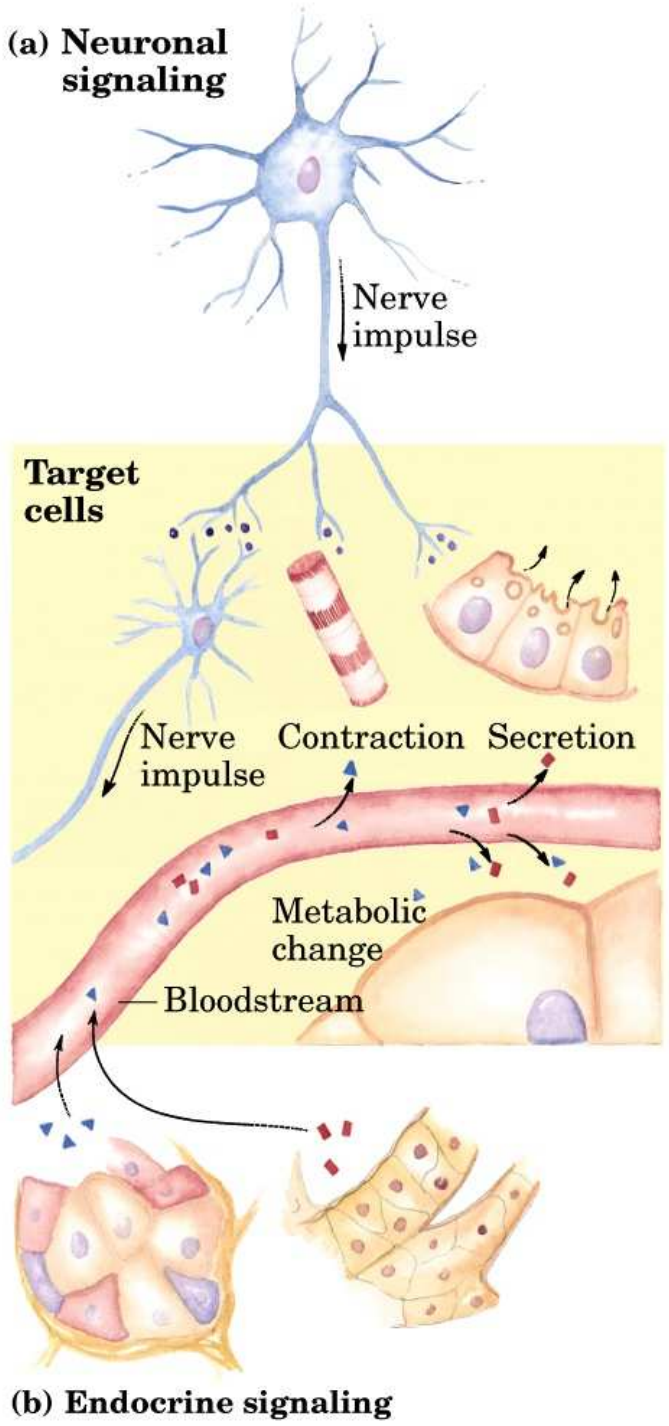
Meios de comunicação celular



Comunicação pelo sistema neuroendócrino

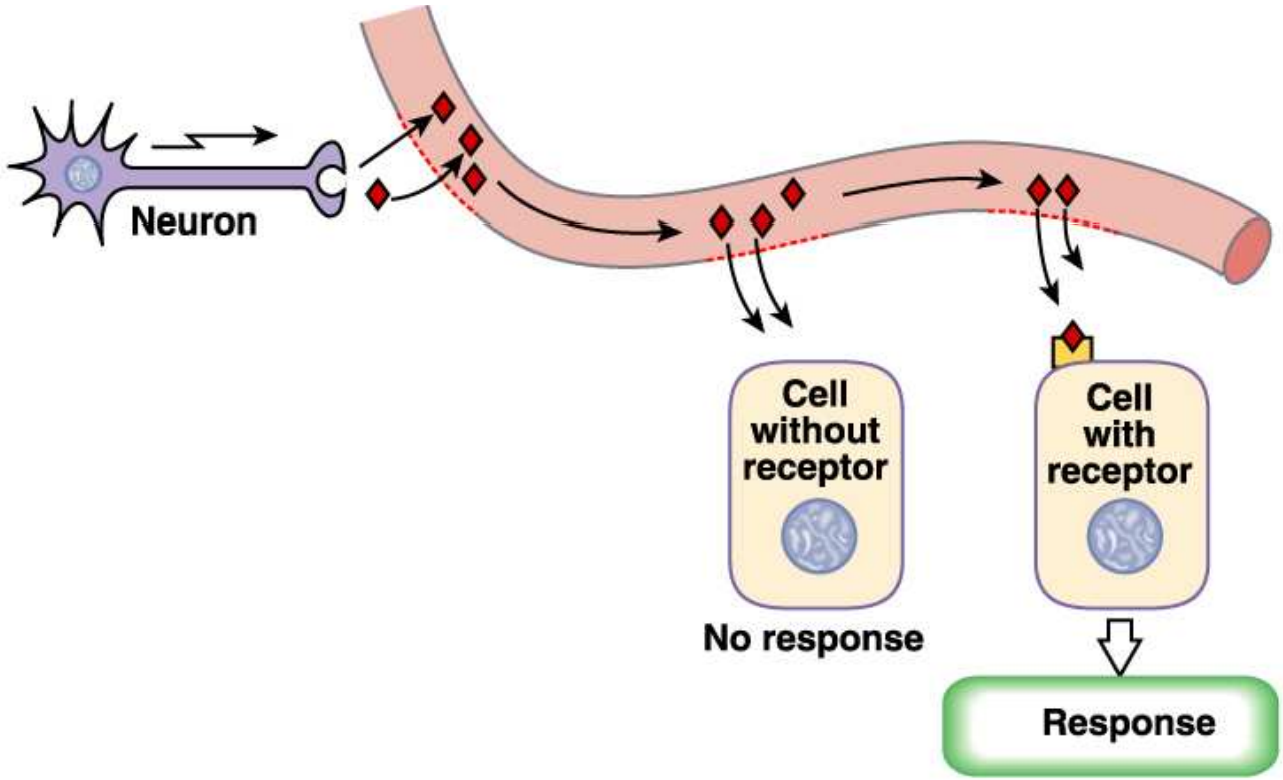
a) **Sinalização neuronal: sinais elétricos são gerados e conduzidos e então neurotransmissores são liberados.**

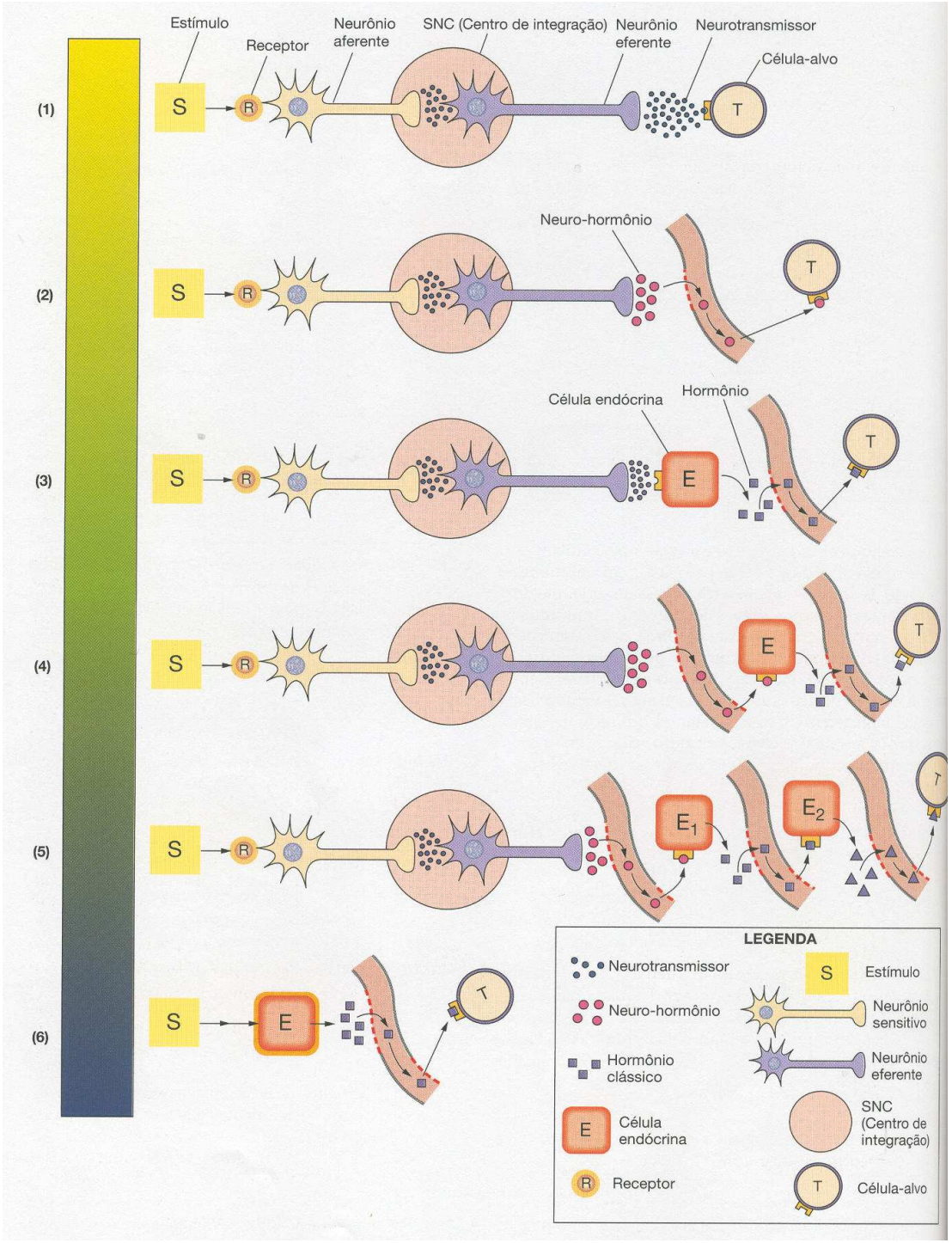
b) **Sistema endócrino: hormônios são secretados na corrente sanguínea e levados ao tecido alvo.**



Meios de comunicação celular

Neuro-hormônio




Estímulo
Receptor
Neurônio aferente
SNC (Centro de integração)
Neurônio eferente
Neurotransmissor
Célula-alvo
(1)
S
R
T
Neuro-hormônio
(2)
Célula endócrina
Hormônio
(3)
E
(4)
E1
E2
(5)
(6)
LEGENDA
Neurotransmissor
Neuro-hormônio
Hormônio clássico
Célula endócrina
Receptor
Estímulo
Neurônio sensitivo
Neurônio eferente
SNC (Centro de integração)
Célula-alvo

Citocinas

- Desenvolvimento celular
- Diferenciação celular
- Resposta imunológica

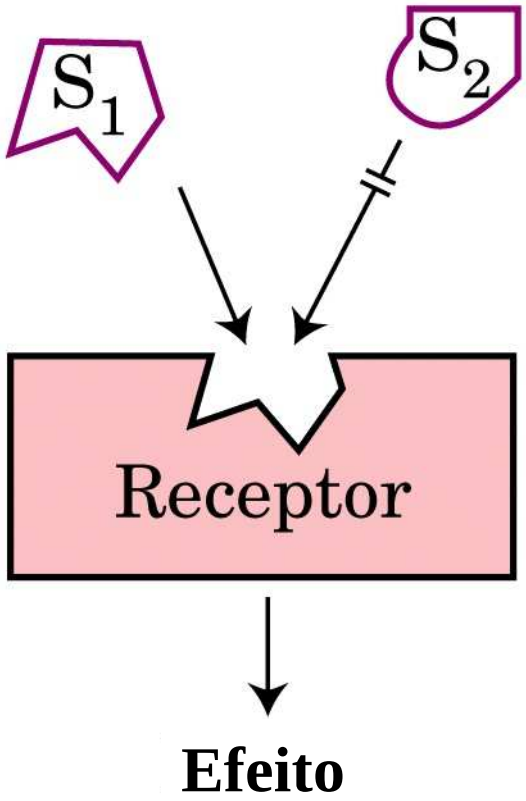
Citocinas x hormônios

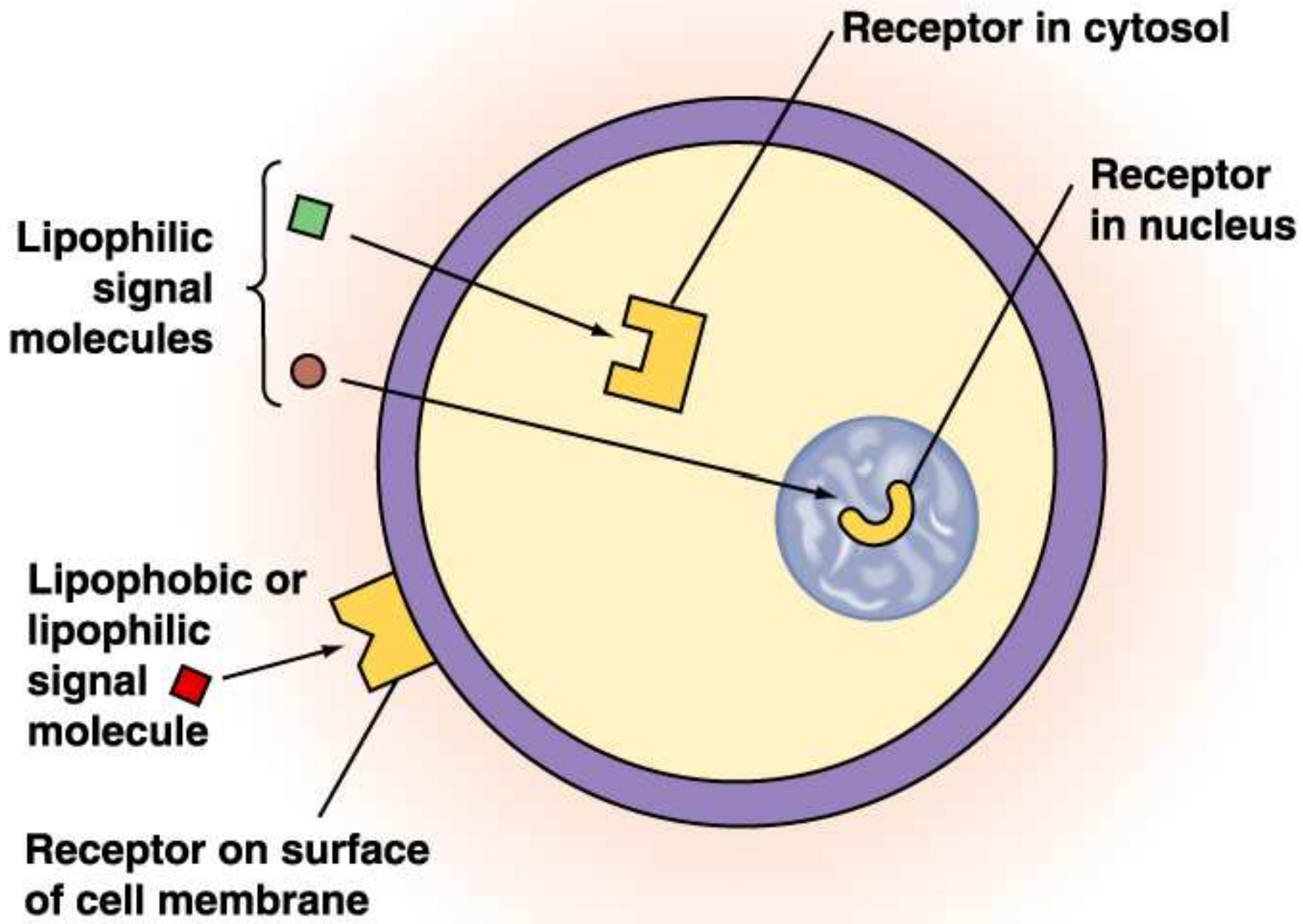
- **Hormônios**
  - produzidos por glândulas especializadas
  - armazenados e eventualmente liberados
- **Citocinas**
  - atuam em espectro mais amplo de células alvo
  - sintetizadas sob demanda

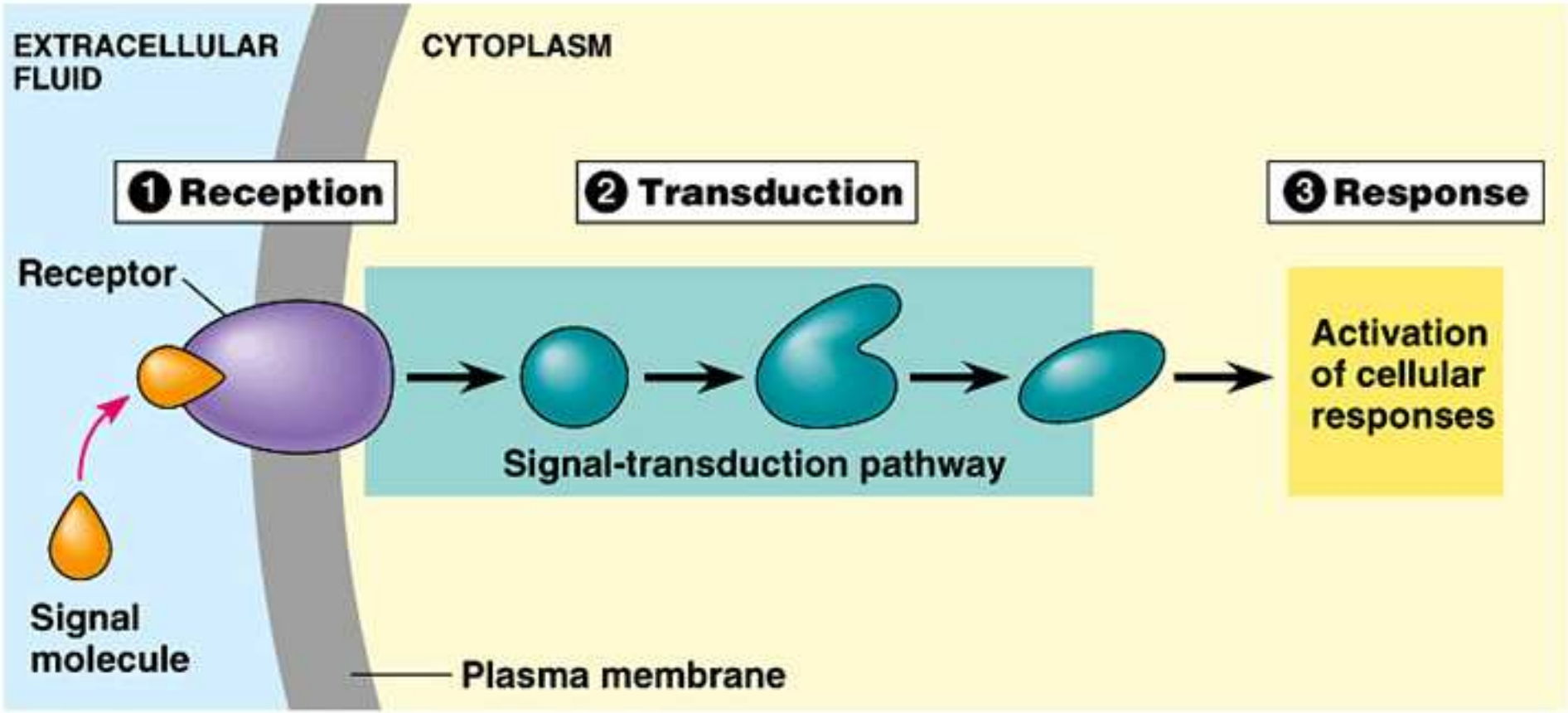
Características gerais da sinalização celular

1. o sinal (molécula sinalizadora) interage com um receptor
2. o receptor ativa mecanismos celulares, produzindo um segundo sinal ou uma mudança na atividade de uma proteína celular
3. a atividade metabólica da célula alvo se altera
4. o evento de transdução cessa e a célula retorna ao seu estado pré-estímulo

Especificidade




Receptor in cytosol
Receptor in nucleus
Lipophilic signal molecules
Lipophobic or lipophilic signal molecule
Receptor on surface of cell membrane



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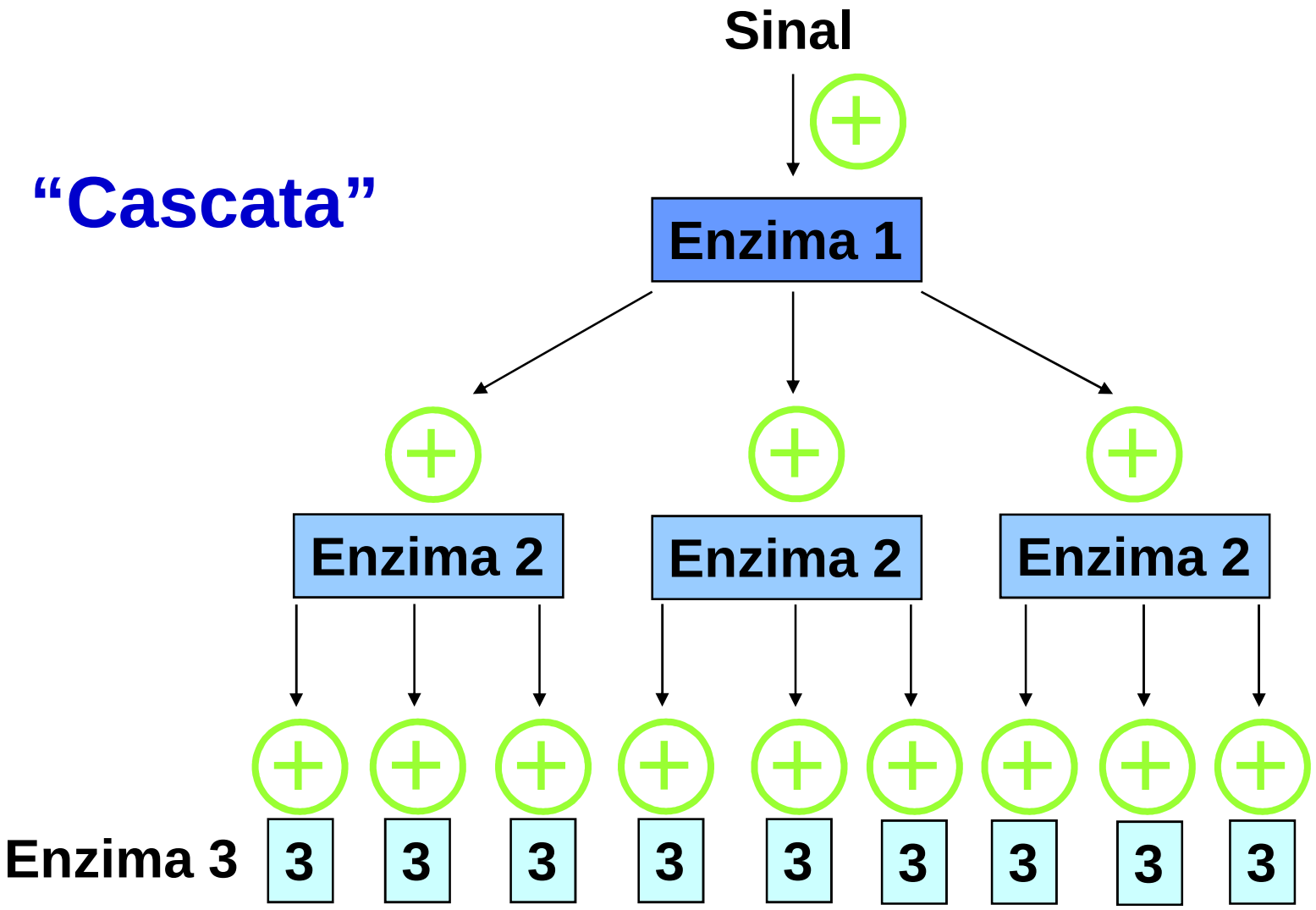
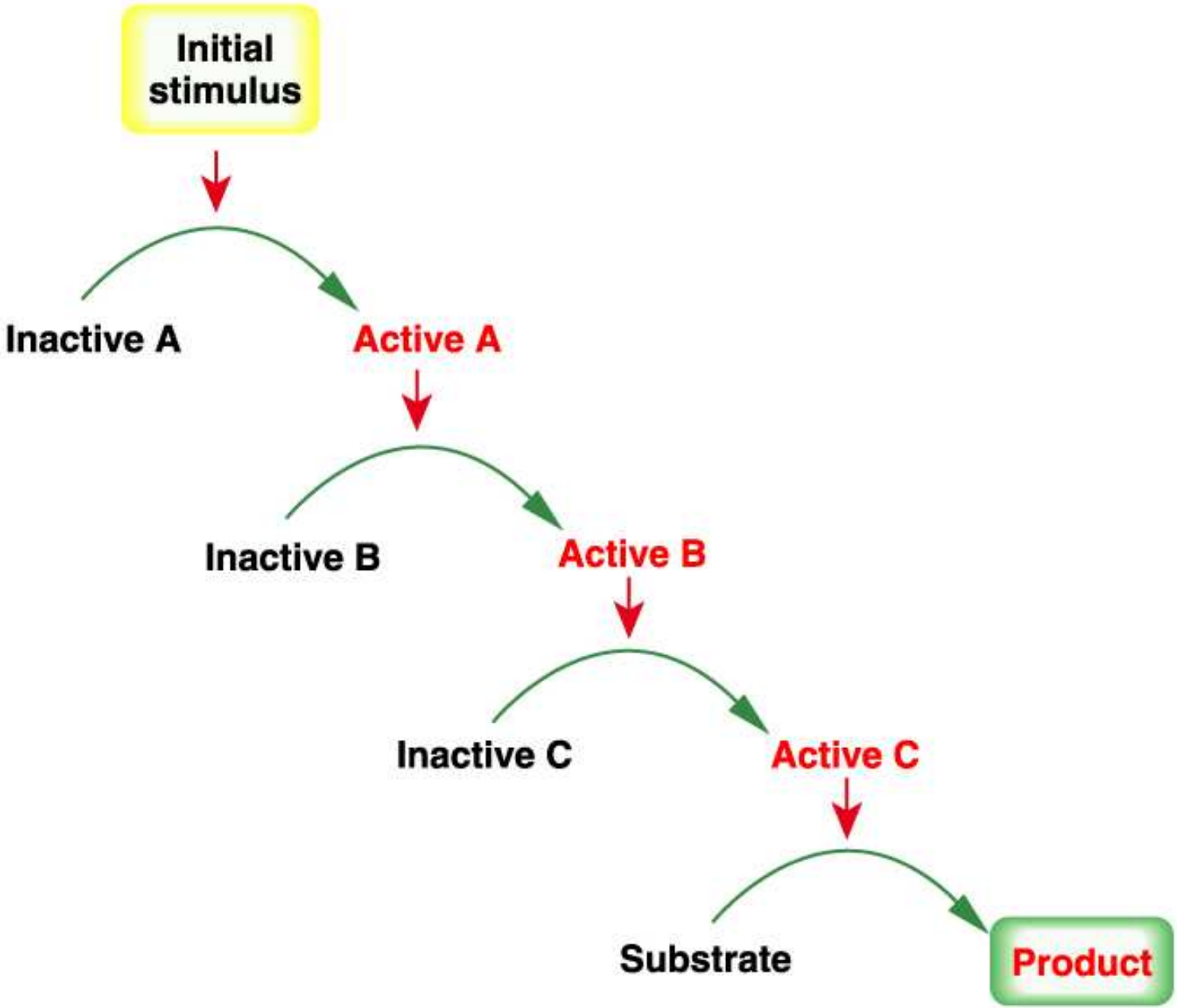
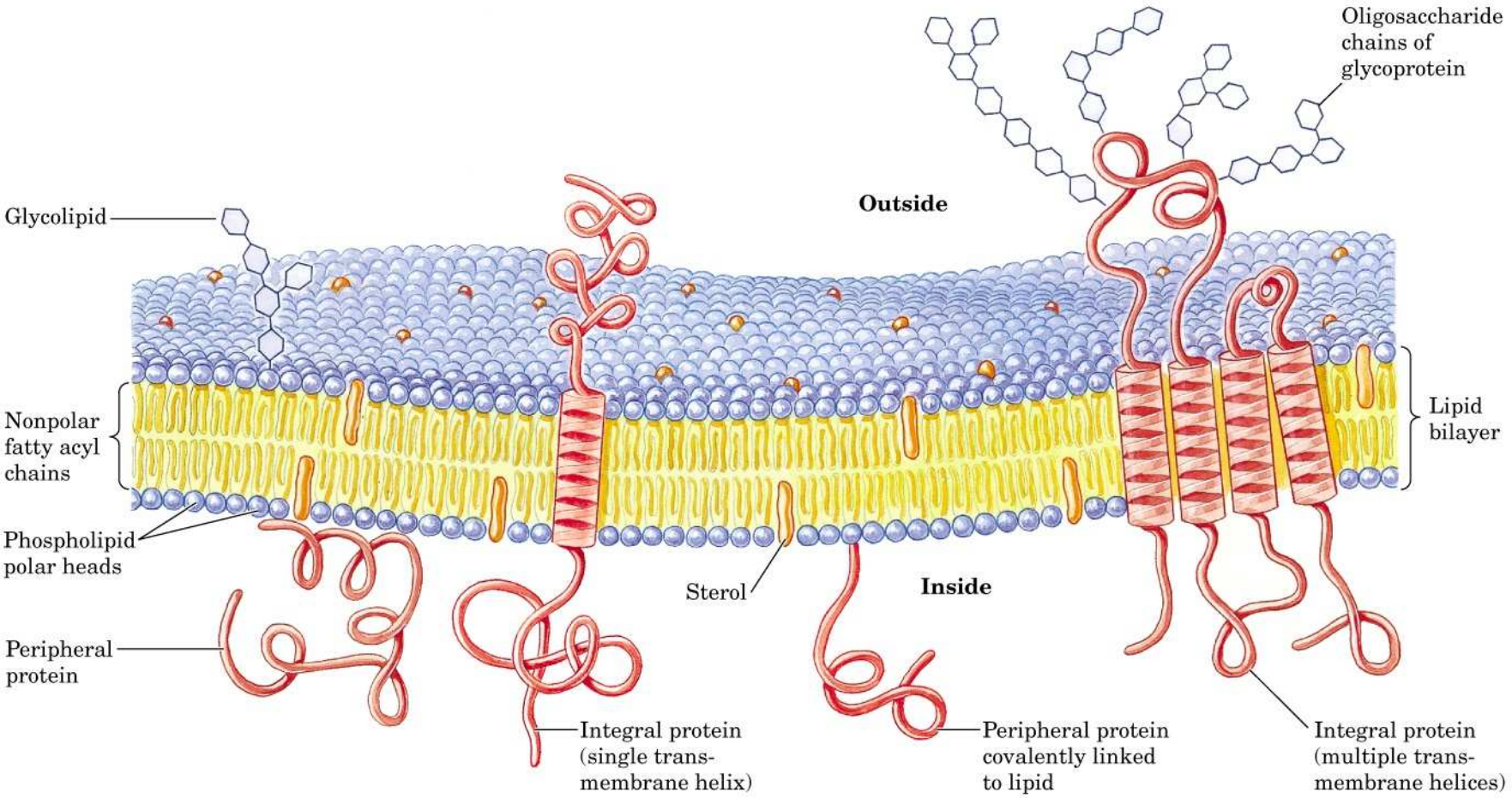
Amplificação

“Cascata”

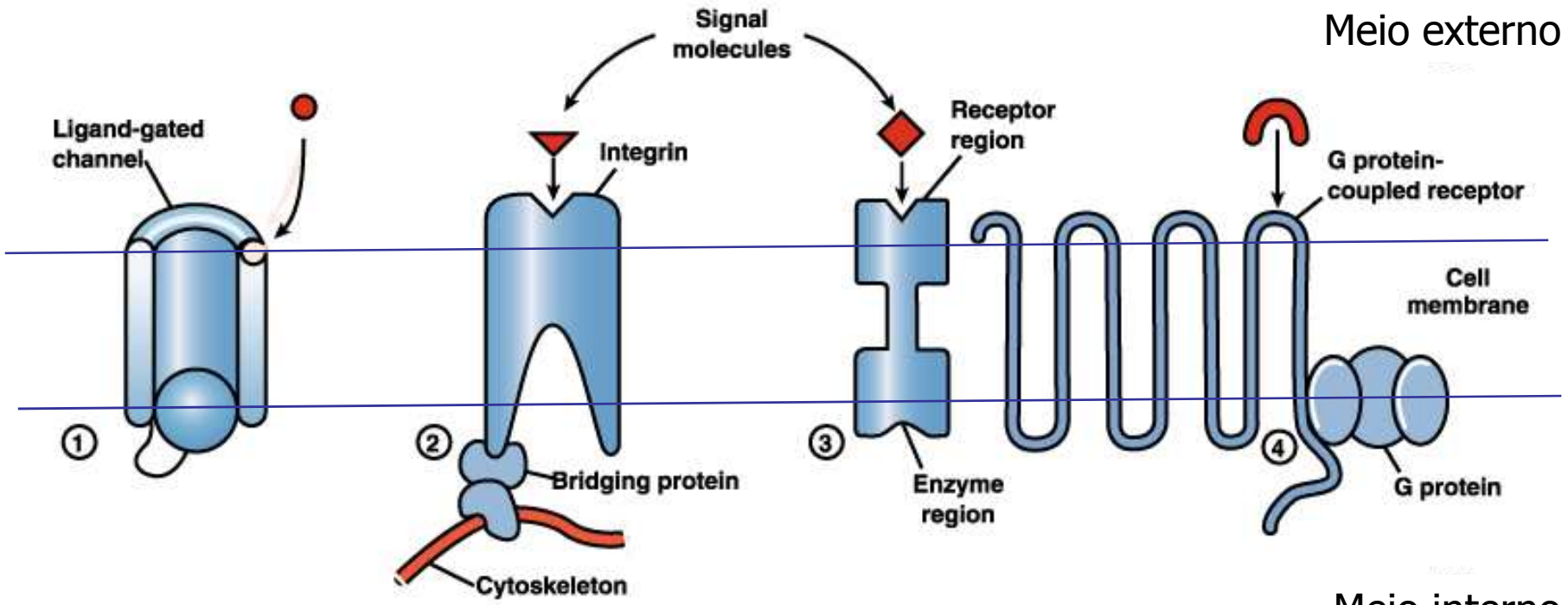



Initial stimulus
Inactive A
Active A
Inactive B
Active B
Inactive C
Active C
Substrate
Product



500 a 100.000 receptores na membrana de uma célula!

Quatro classes de receptores de membrana



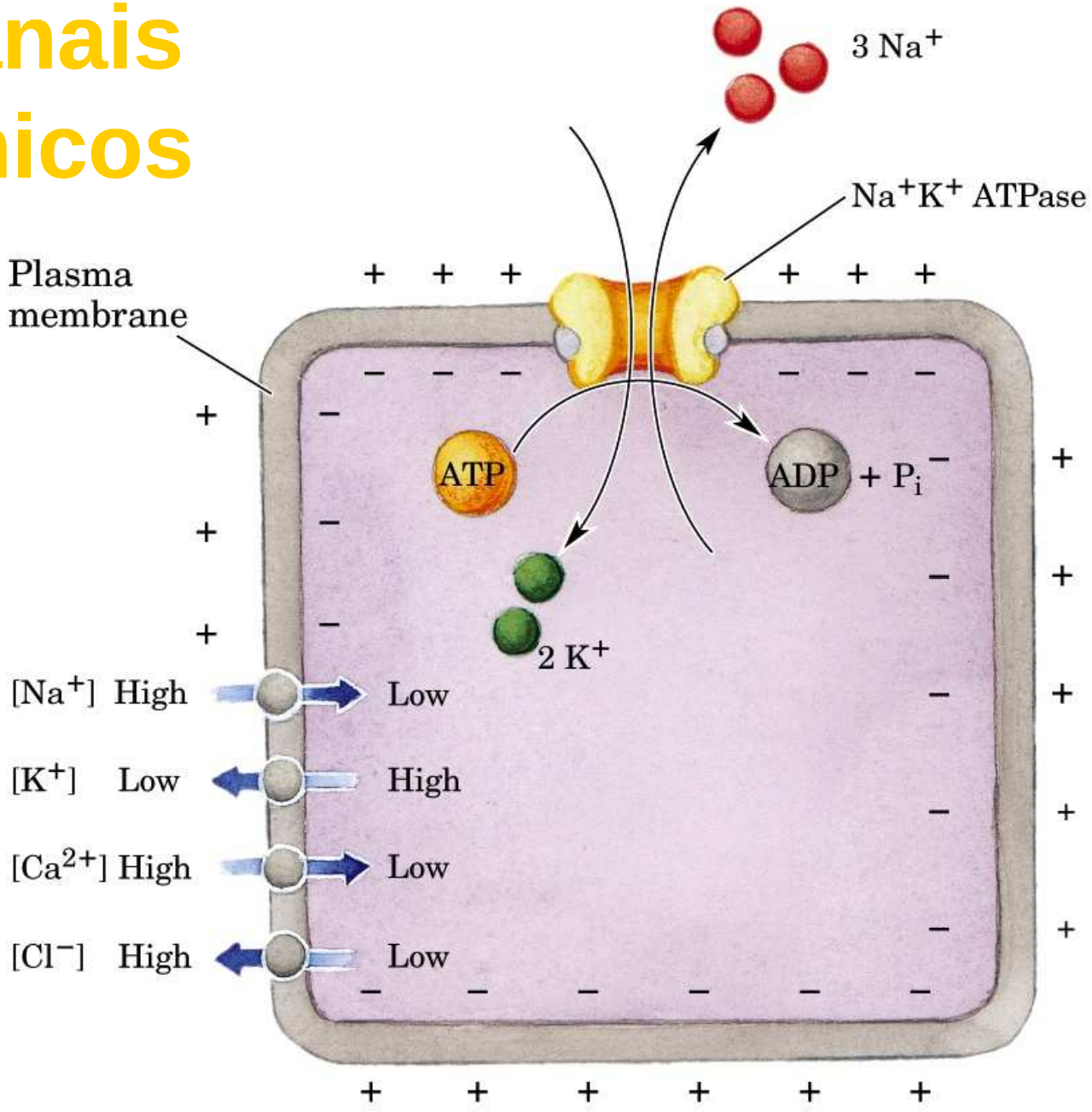
Canais iônicos

Integrinas

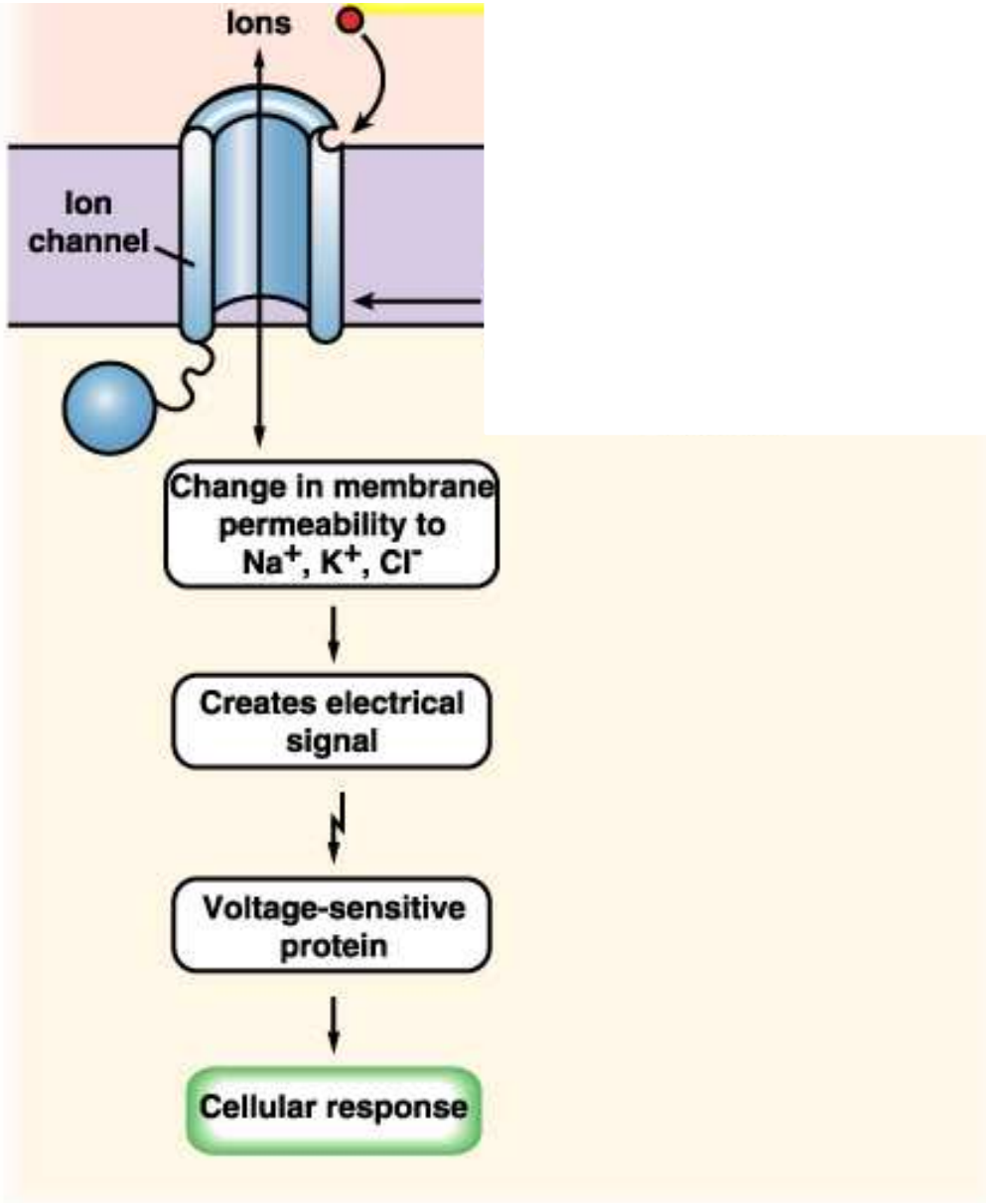
Receptores enzimáticos

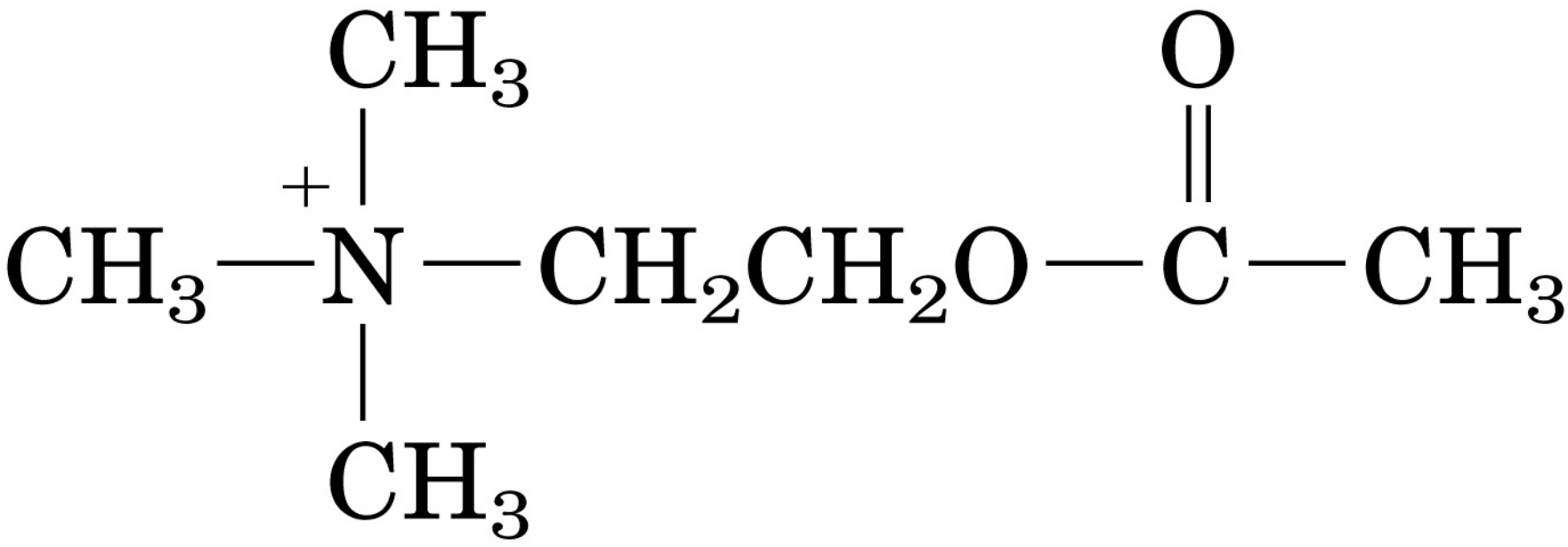
Receptores ligados a proteína G

1 Canais iônicos

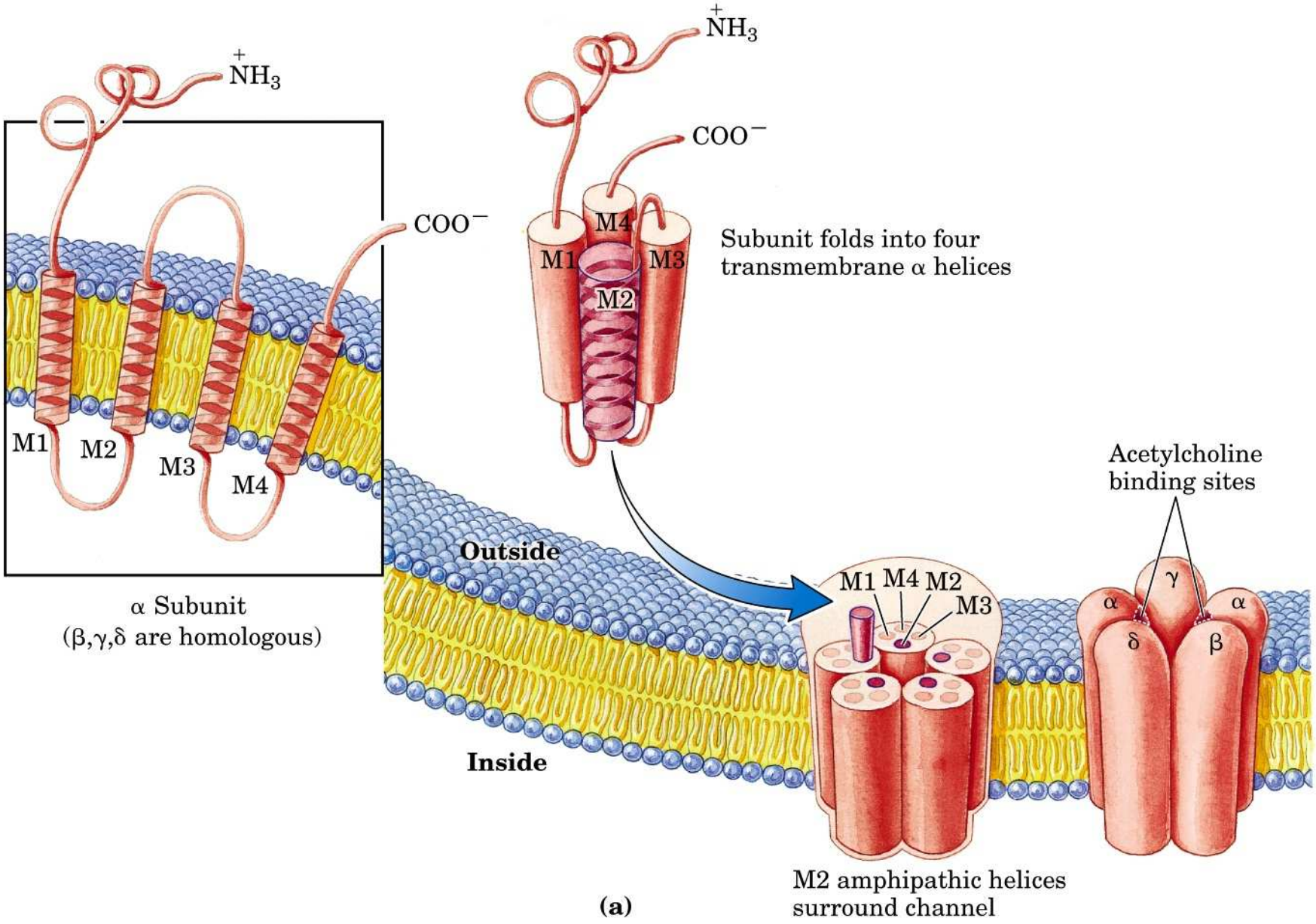


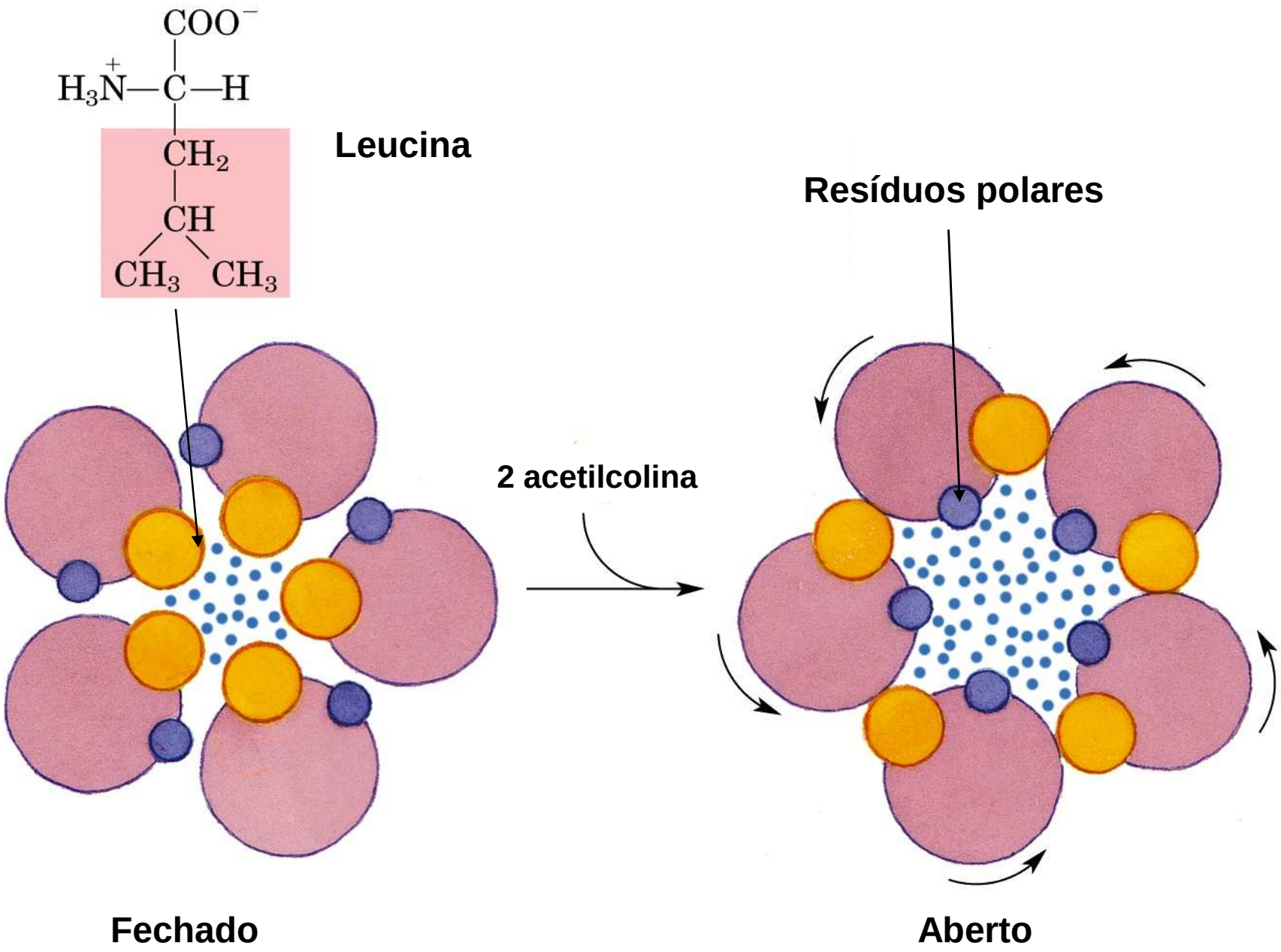
1 Canais iônicos

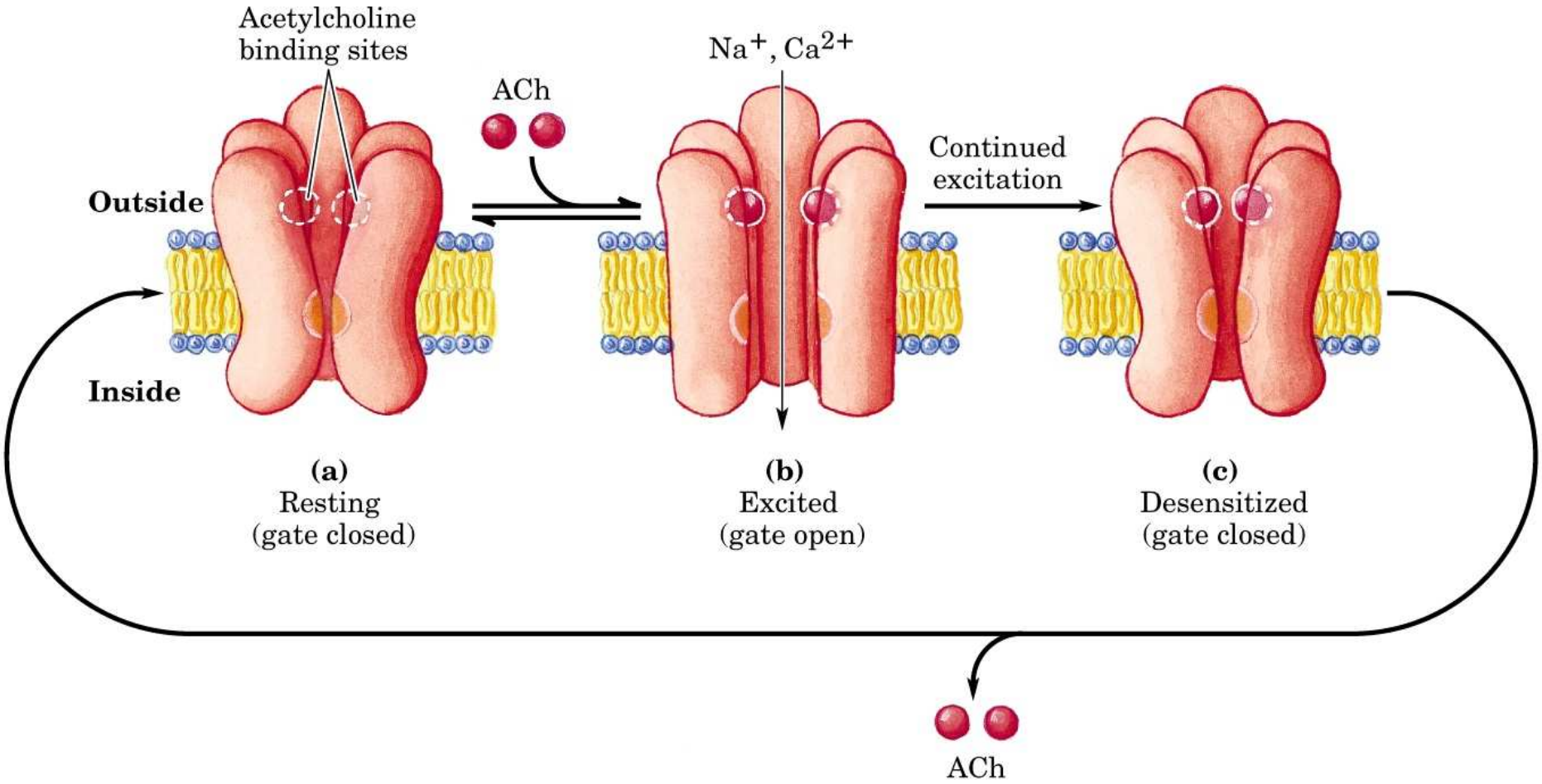


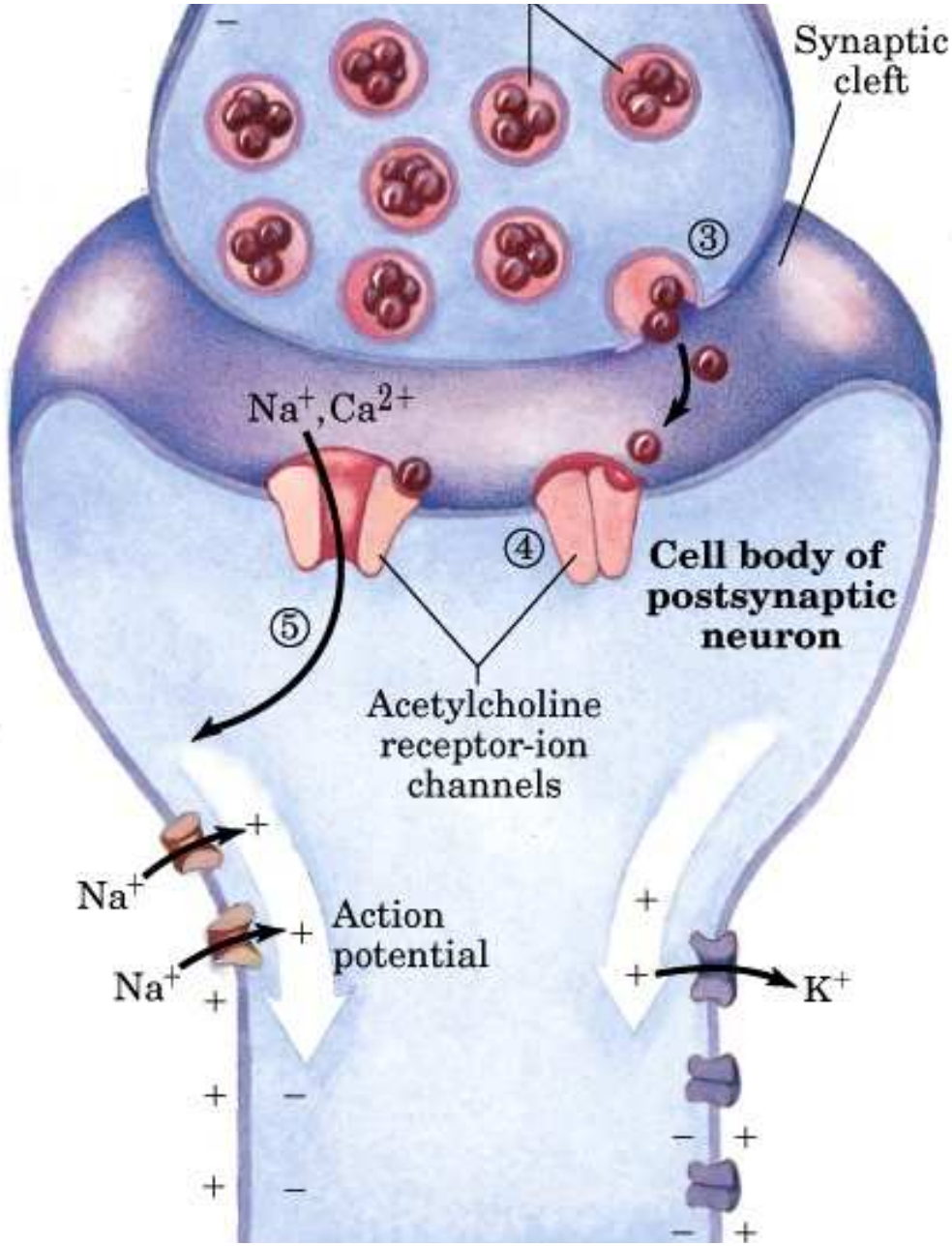


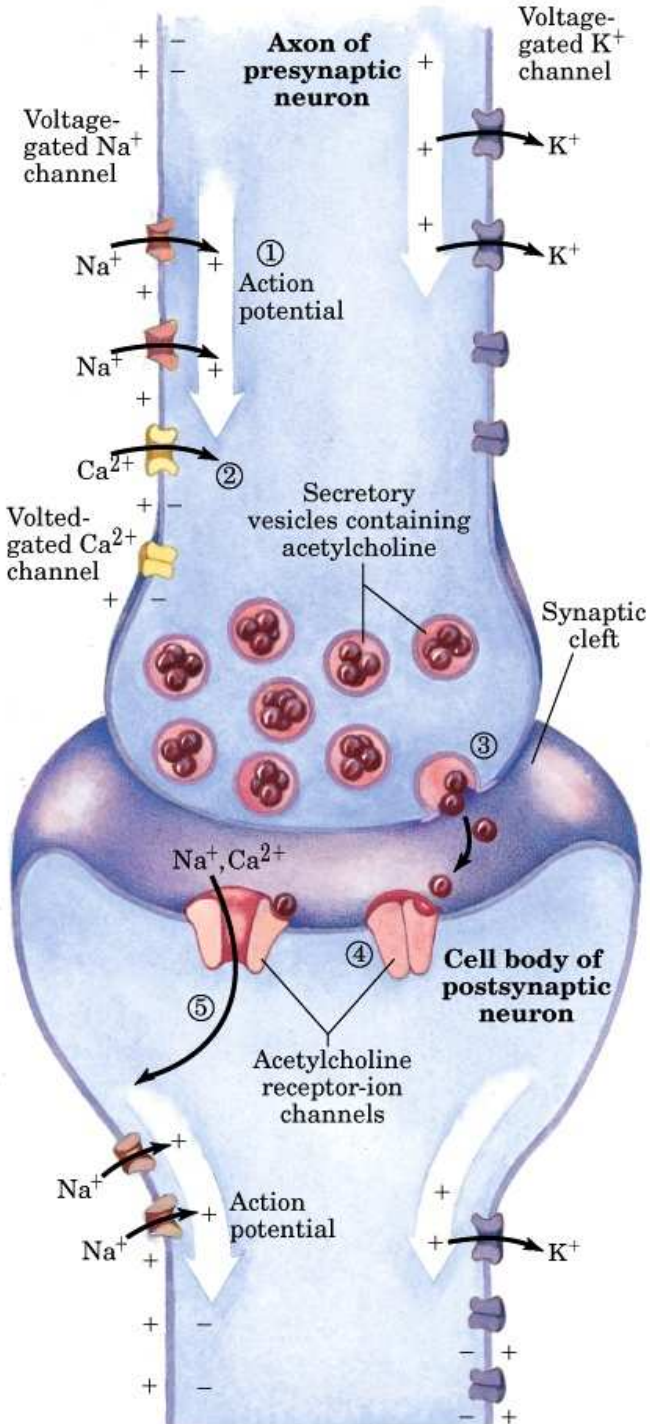
Acetilcolina


+NH3
COO−
M1
M2
M3
M4
α Subunit
(β,γ,δ are homologous)
+NH3
COO−
M4
M1
M3
M2
Subunit folds into four
transmembrane α helices
Outside
Inside
M1 M4 M2
M3
Acetylcholine
binding sites
γ
α
α
δ
β
M2 amphipathic helices
surround channel
(a)

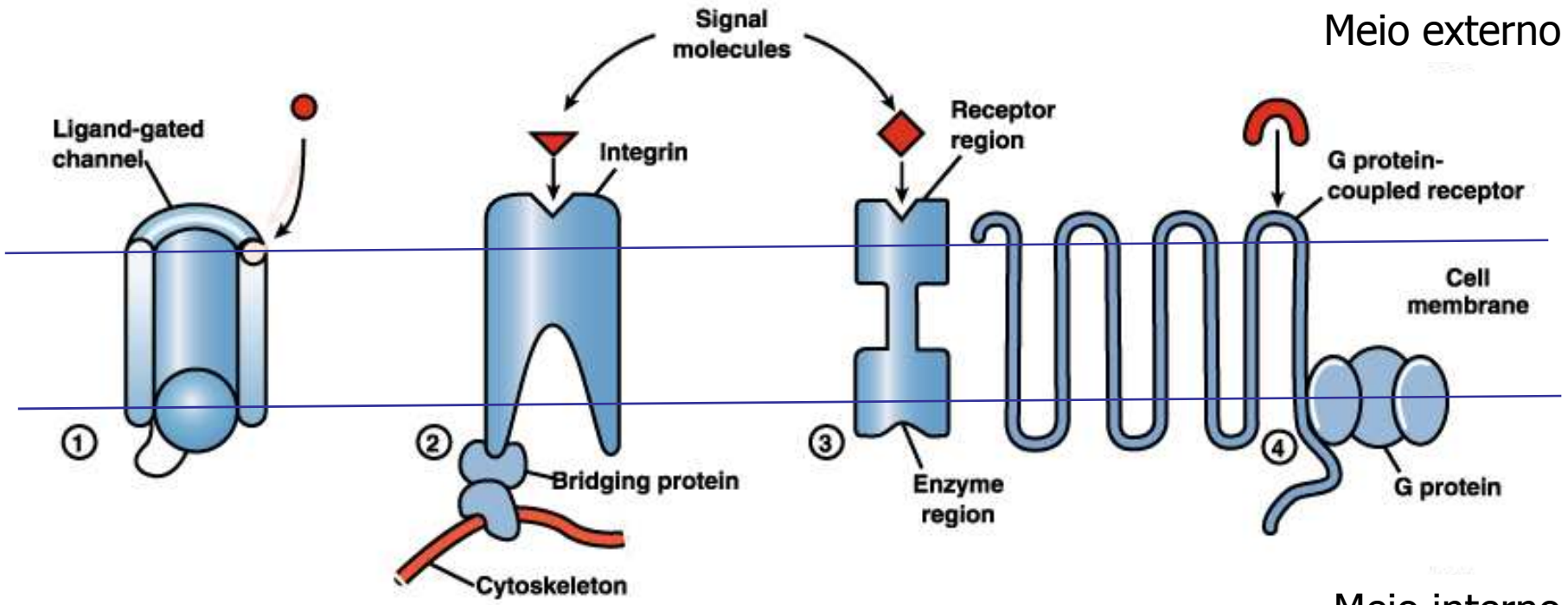

Leucina
Resíduos polares
2 acetilcolina
Fechado
Aberto


Acetylcholine binding sites
ACh
Na^+, Ca^{2+}
Continued excitation
Outside
Inside
(a) Resting (gate closed)
(b) Excited (gate open)
(c) Desensitized (gate closed)
ACh


Synaptic cleft
③
Na^+, Ca^{2+}
④
Cell body of postsynaptic neuron
⑤
Acetylcholine receptor-ion channels
Na^+
Na^+
Action potential
K^+


Axon of presynaptic neuron
Voltage-gated K+ channel
Voltage-gated Na+ channel
K+
K+
Na+
Na+
① Action potential
Ca2+
②
Volted-gated Ca2+ channel
Secretory vesicles containing acetylcholine
Synaptic cleft
③
Na+, Ca2+
④
Cell body of postsynaptic neuron
⑤
Acetylcholine receptor-ion channels
Na+
Na+
Action potential
K+

Quatro classes de receptores de membrana

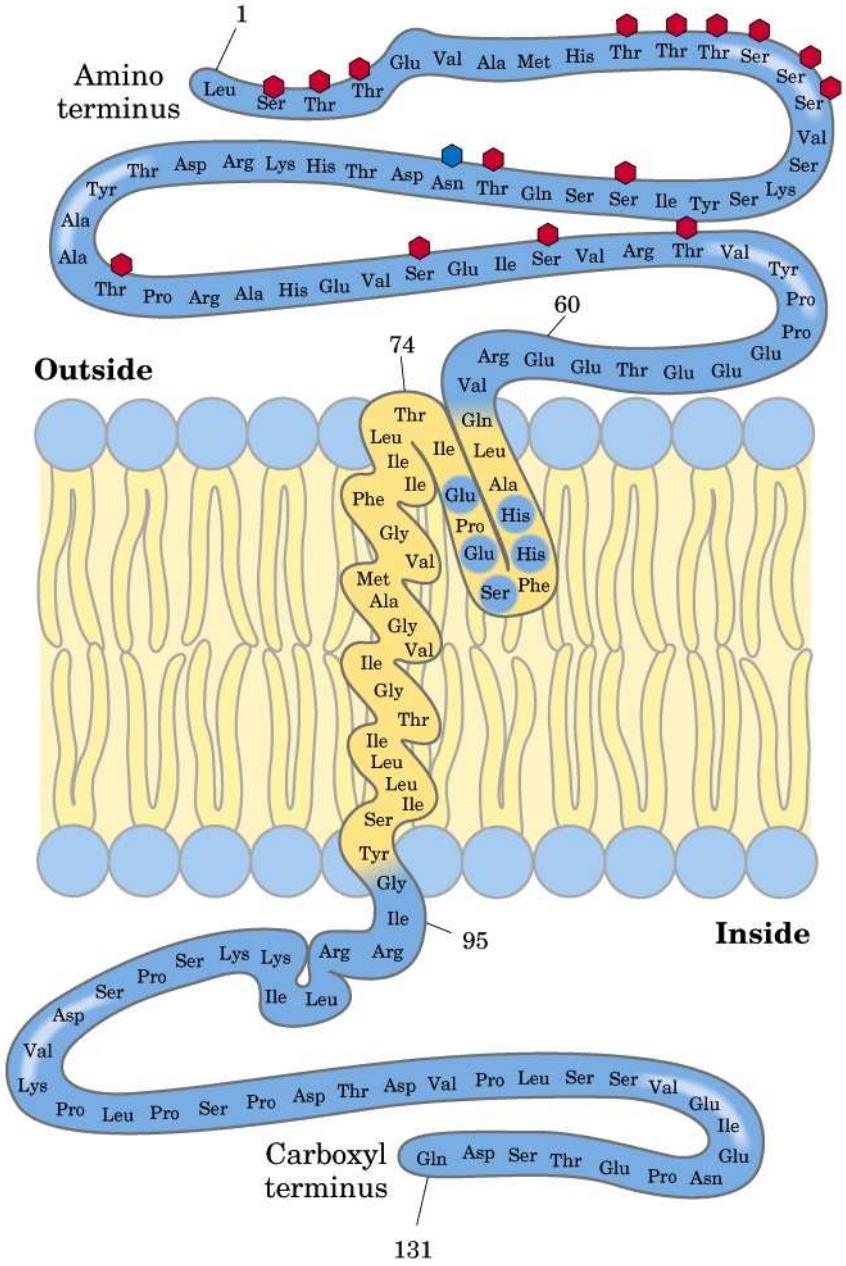


Canais iônicos

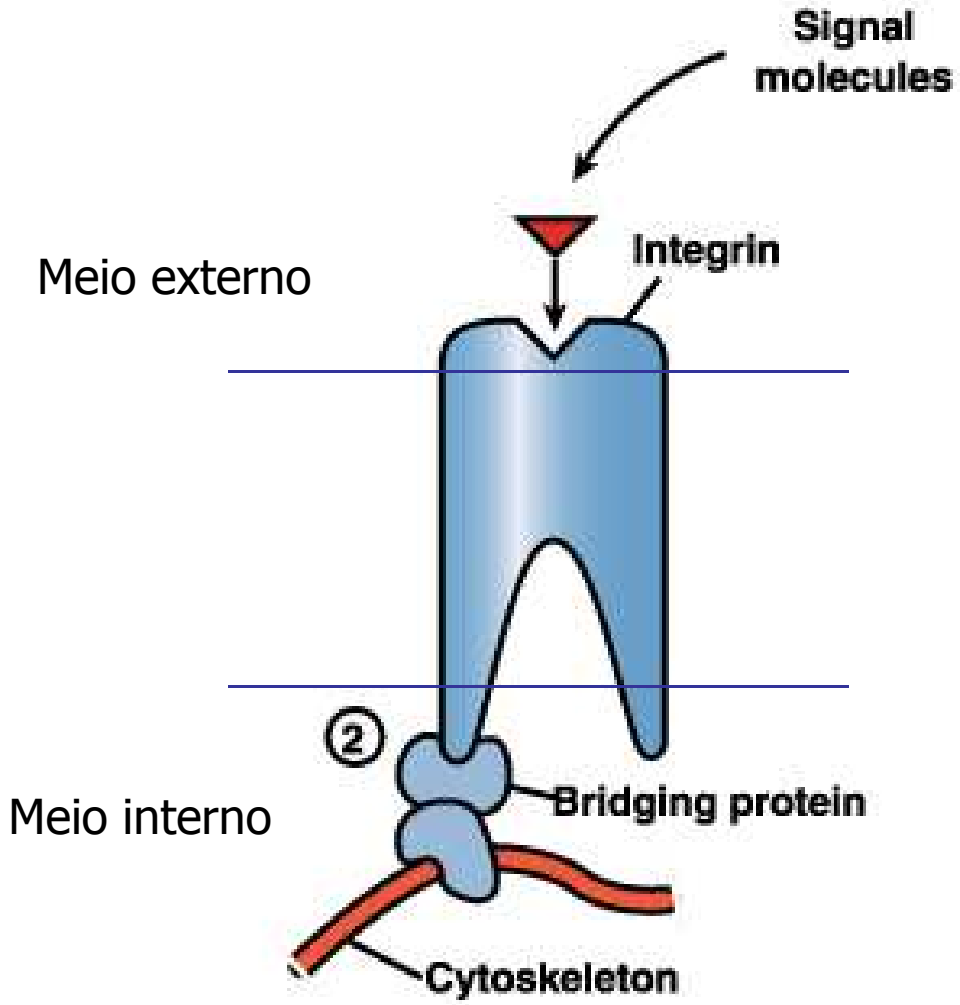
Integrinas

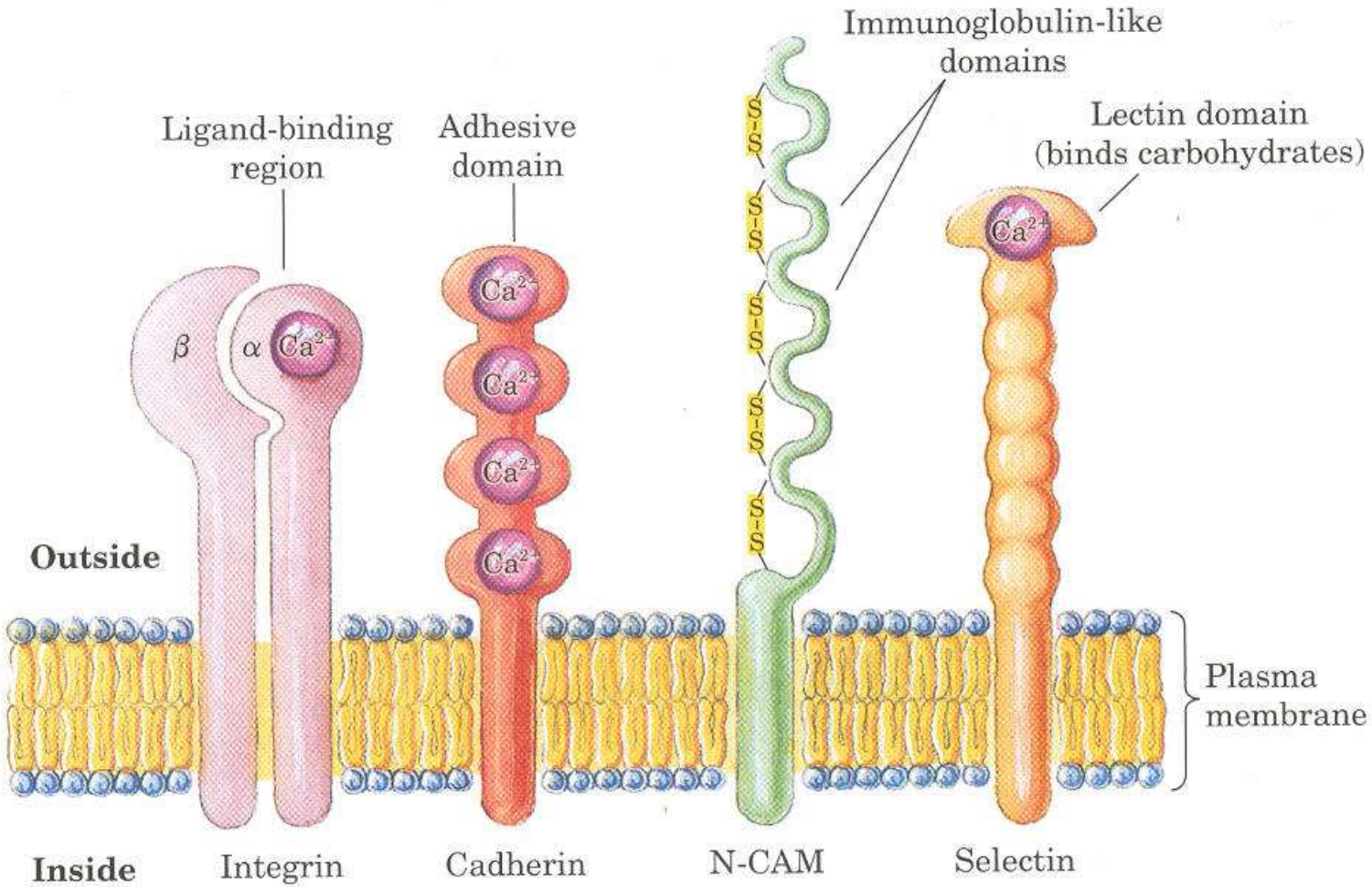
Receptores enzimáticos

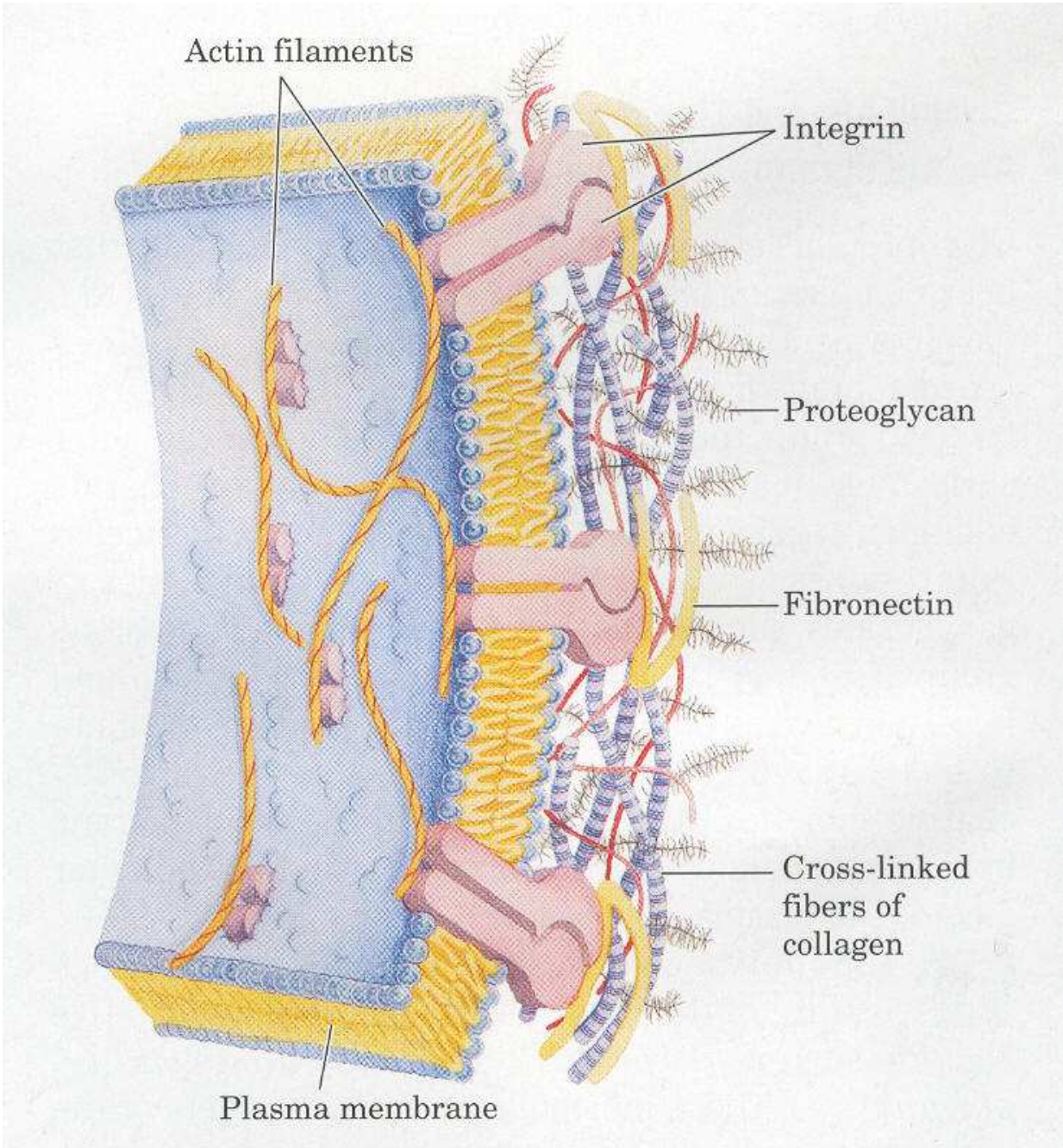
Receptores ligados a proteína G

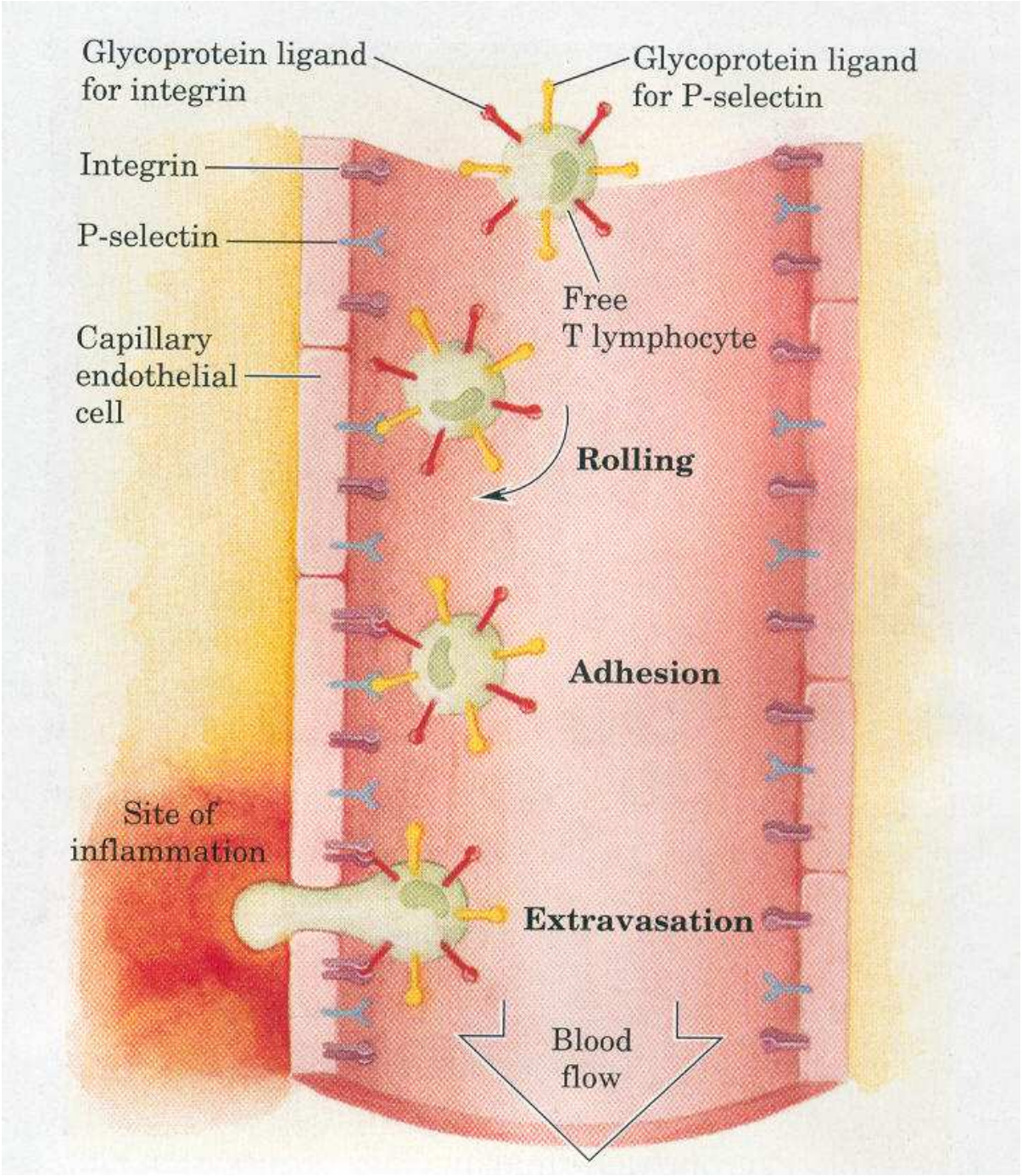

1
Amino terminus
Leu Ser Thr Thr Glu Val Ala Met His Thr Thr Thr Ser Ser Ser Ser Val Ser Lys Ser Ile Tyr Ser Ser Gln Thr Gly Asn Thr Asp His Lys Arg Asp Thr Tyr Ala Ala Thr Pro Arg Ala His Glu Val Ser Glu Ile Ser Val Arg Thr Val Tyr Pro Pro Glu Glu Thr Glu Glu Arg Val
60
74
Outside
Thr Leu Ile Ile Ile Phe Gly Val Met Ala Gly Val Ile Gly Thr Ile Leu Leu Ile Ser Tyr Gly Ile
Gln Leu Ala His His Phe Glu Pro Glu Ser
95
Inside
Ser Lys Lys Arg Arg Pro Ser Asp Ile Leu
Val Lys Pro Leu Pro Ser Pro Asp Thr Asp Val Pro Leu Ser Ser Val Glu Ile Glu
Carboxyl terminus
Gln Asp Ser Thr Glu Pro Asn
131

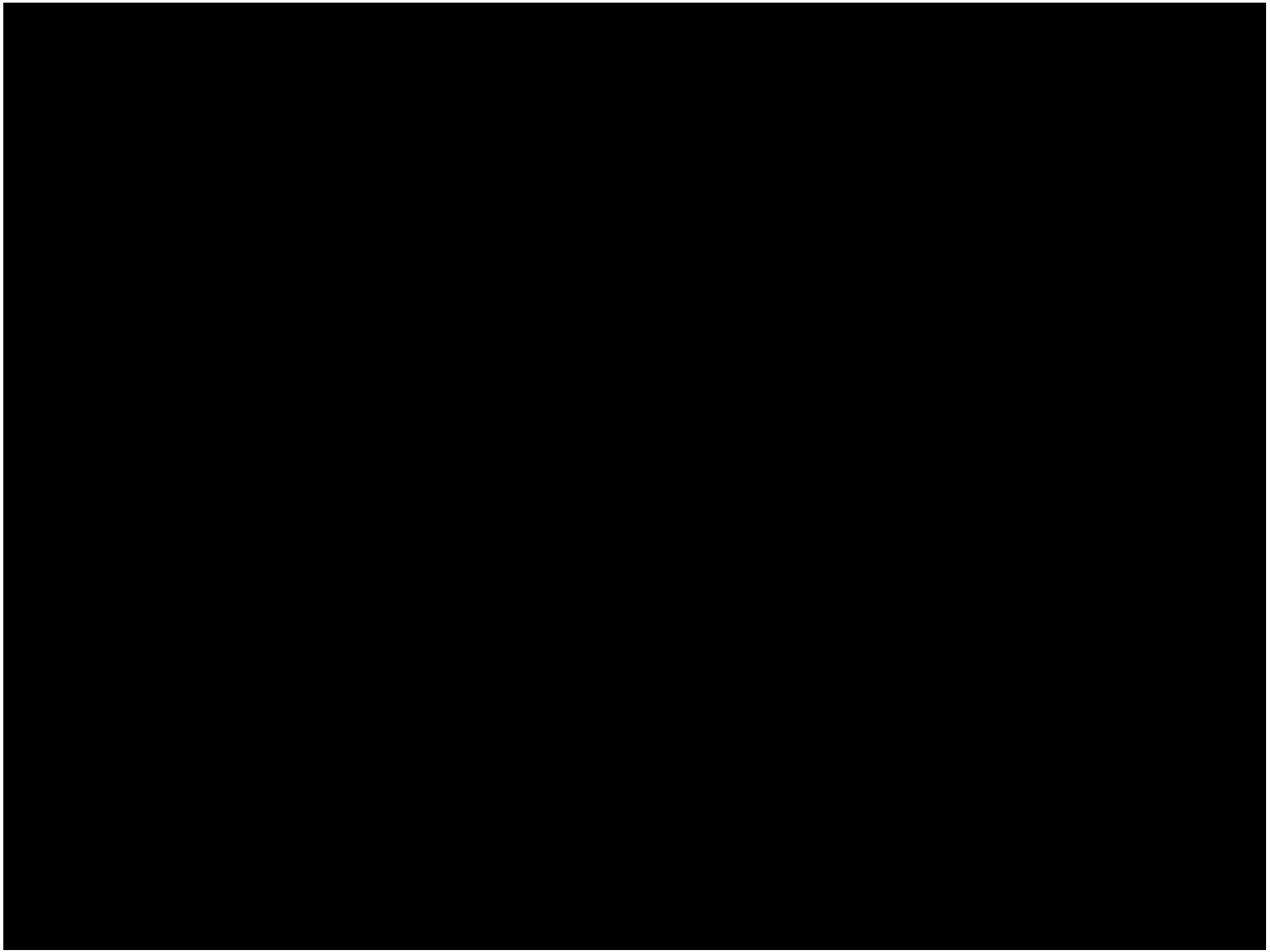
2 Integrinas

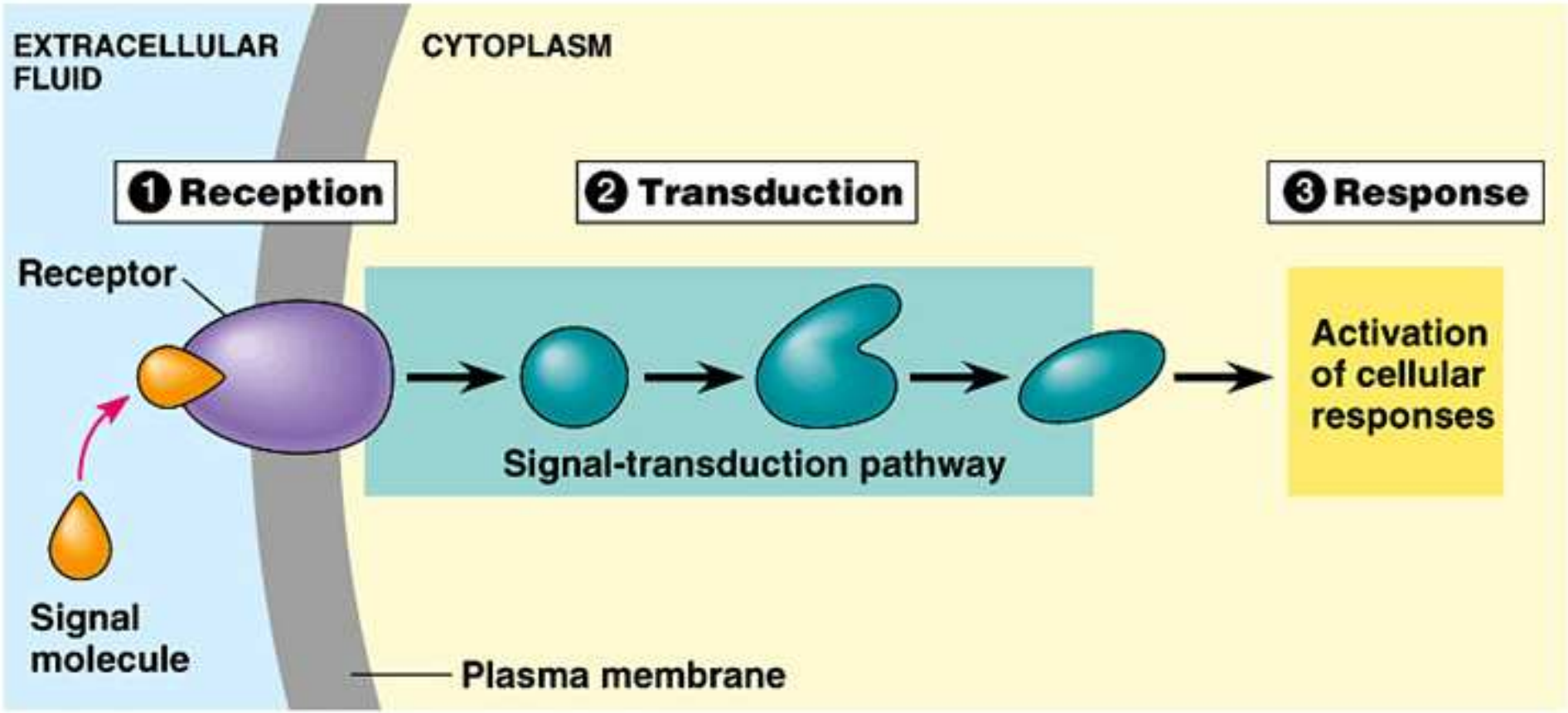



Ligand-binding region
Adhesive domain
Immunoglobulin-like domains
Lectin domain (binds carbohydrates)
β
α
Ca2+
S-S
Outside
Inside
Integrin
Cadherin
N-CAM
Selectin
Plasma membrane


Actin filaments
Integrin
Proteoglycan
Fibronectin
Cross-linked fibers of collagen
Plasma membrane


Glycoprotein ligand for integrin
Glycoprotein ligand for P-selectin
Integrin
P-selectin
Free T lymphocyte
Capillary endothelial cell
Rolling
Adhesion
Site of inflammation
Extravasation
Blood flow





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Amplificação

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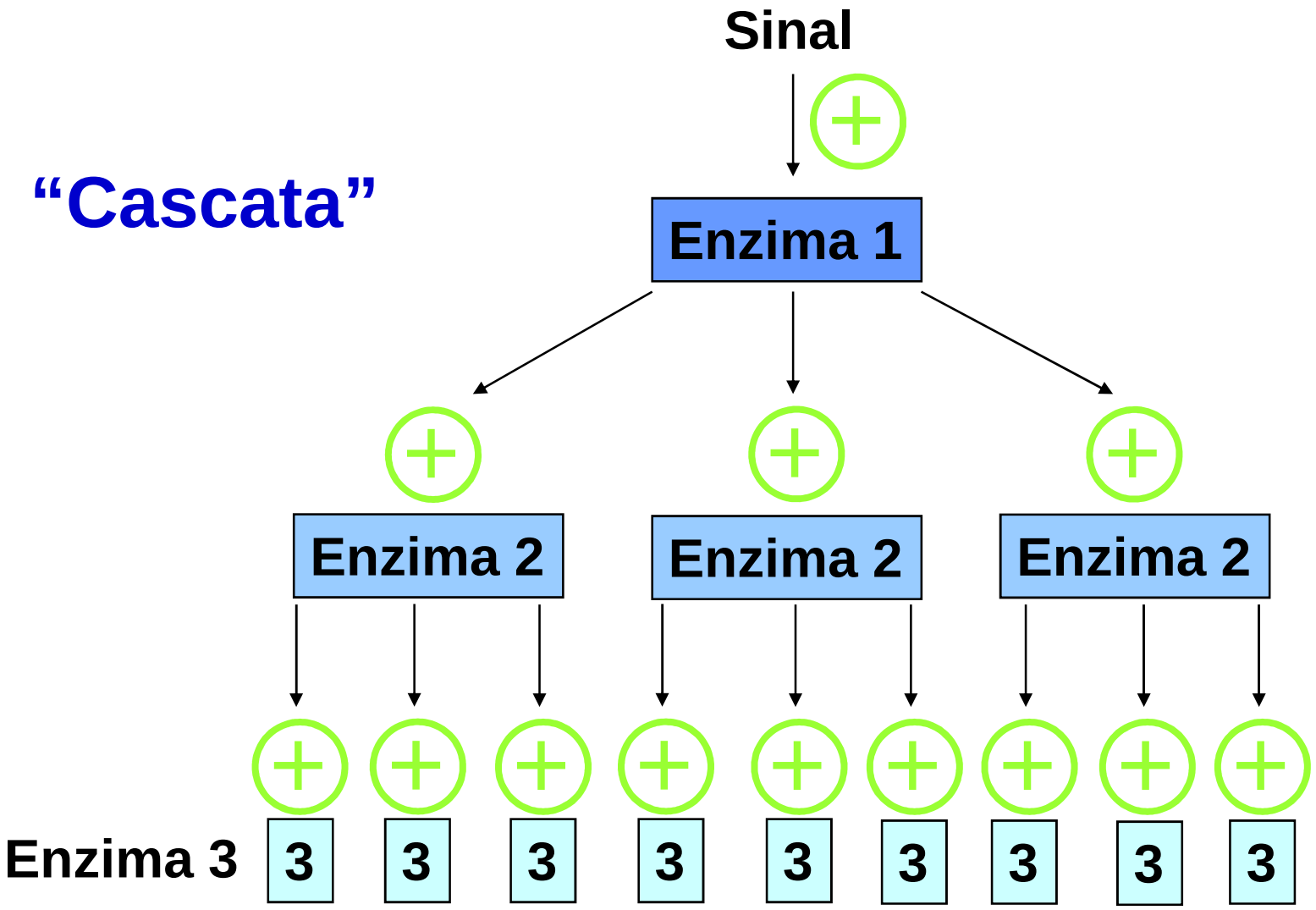
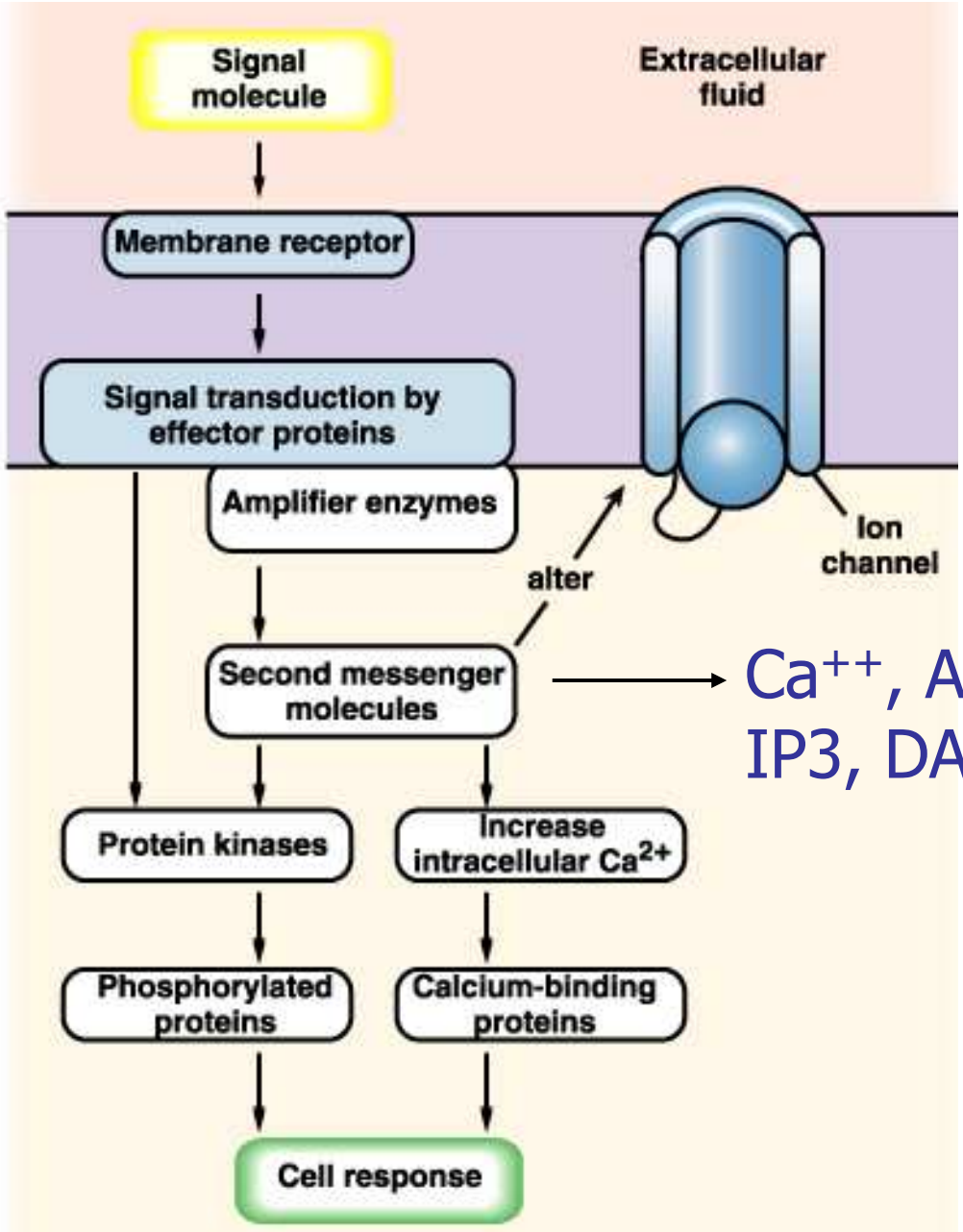
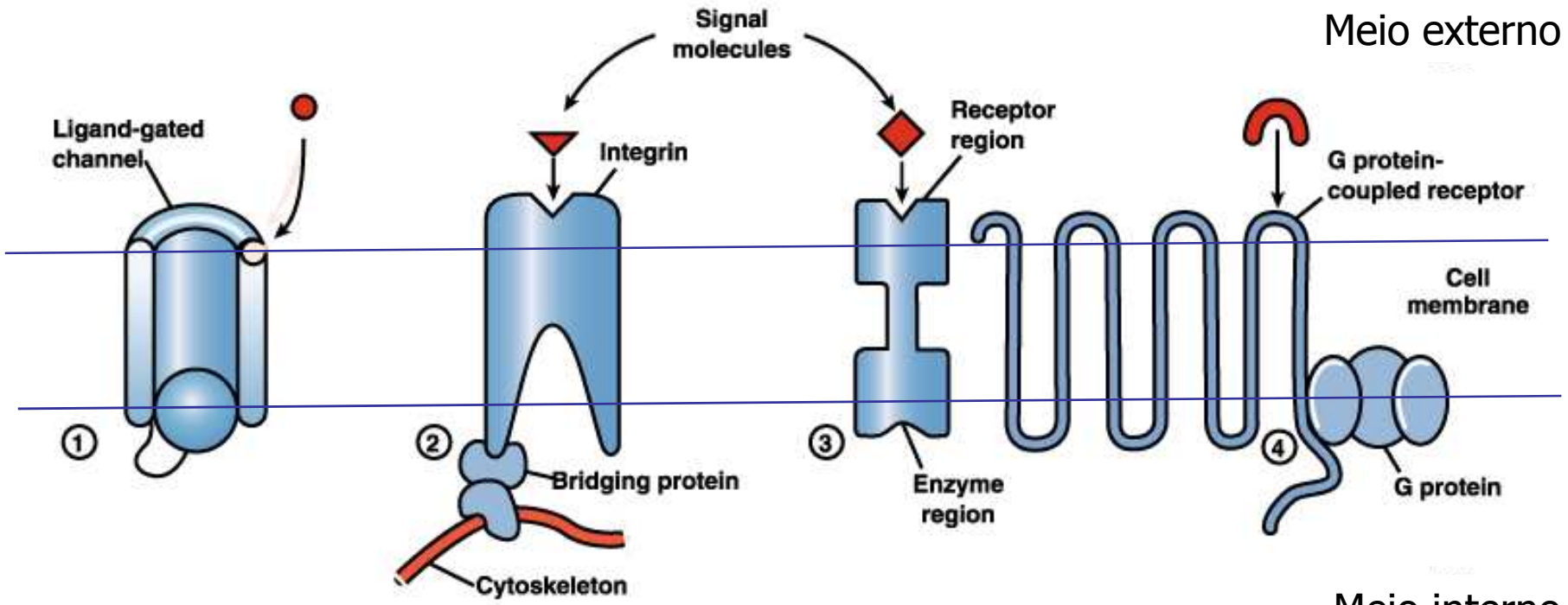




Ca^{++}, AMPc, GMPc, IP3, DAG

Quatro classes de receptores de membrana



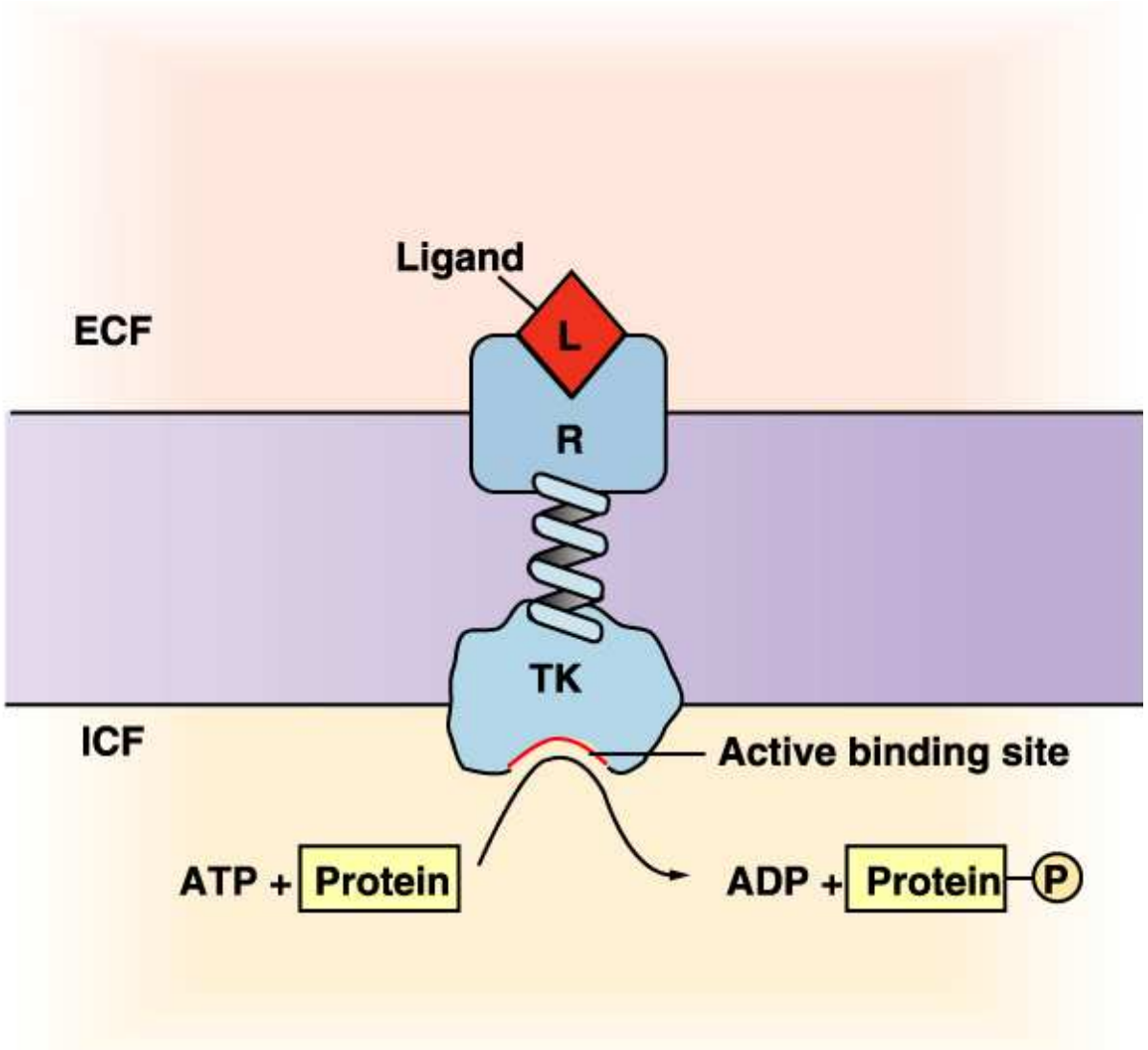
Canais iônicos

Integrinas

Receptores enzimáticos

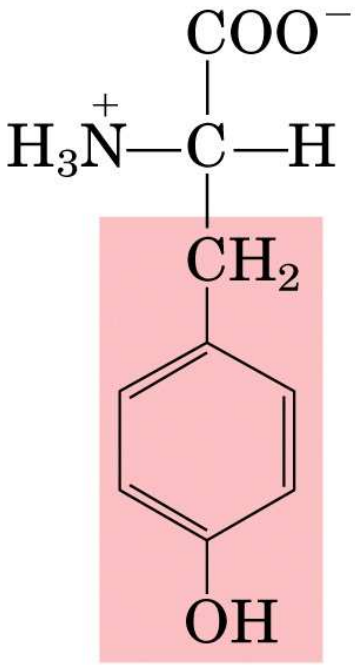
Receptores ligados a proteína G

3 Receptores enzimáticos

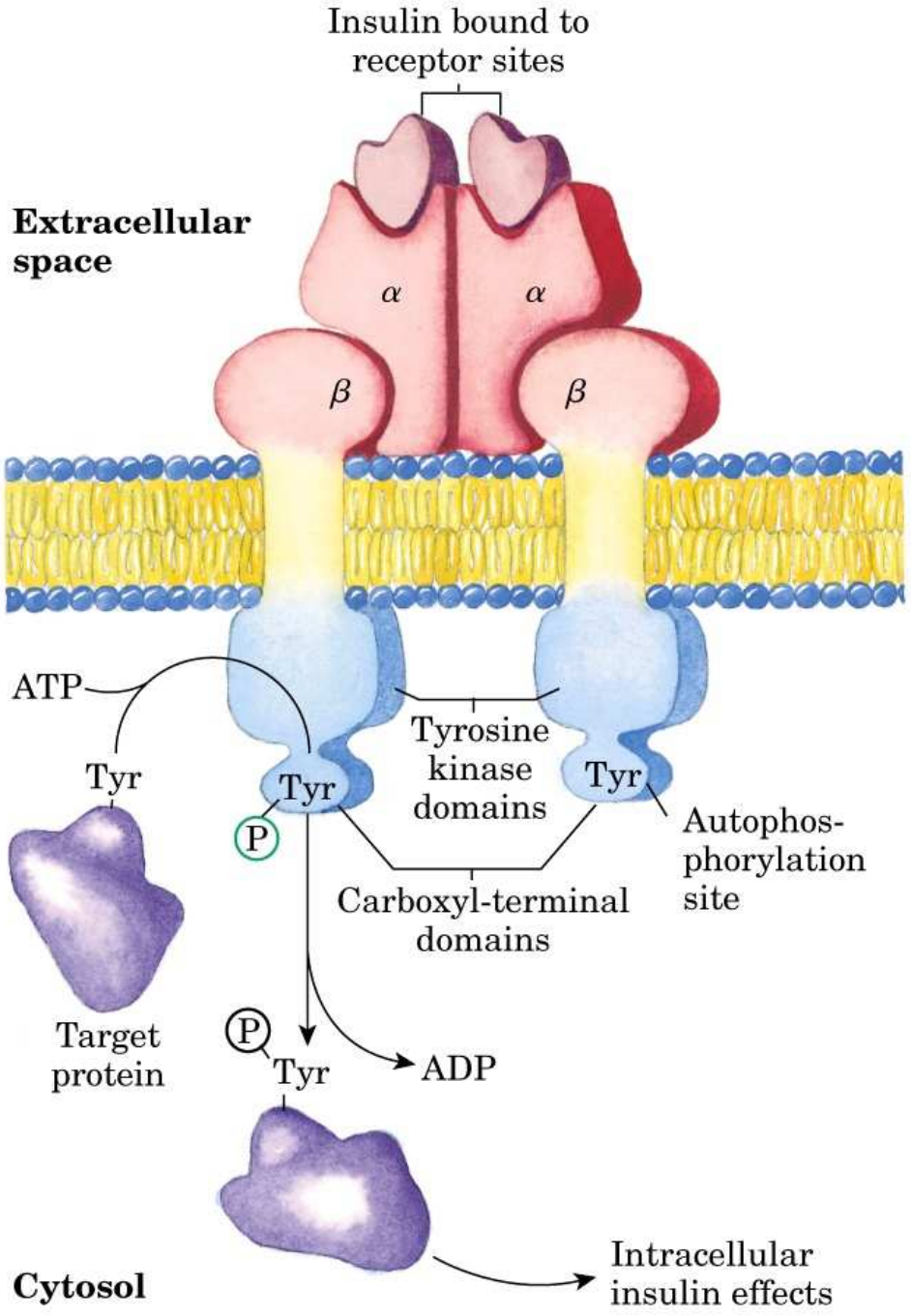


3 Receptores enzimáticos

Receptores com atividade de tirosina-quinase



Tirosina



Insulina

↓

Alvos:

MÚSCULO, FÍGADO, TEC. ADIPOSO

Efeitos metabólicos

↑ incorporação de glicose (músculo e fígado)

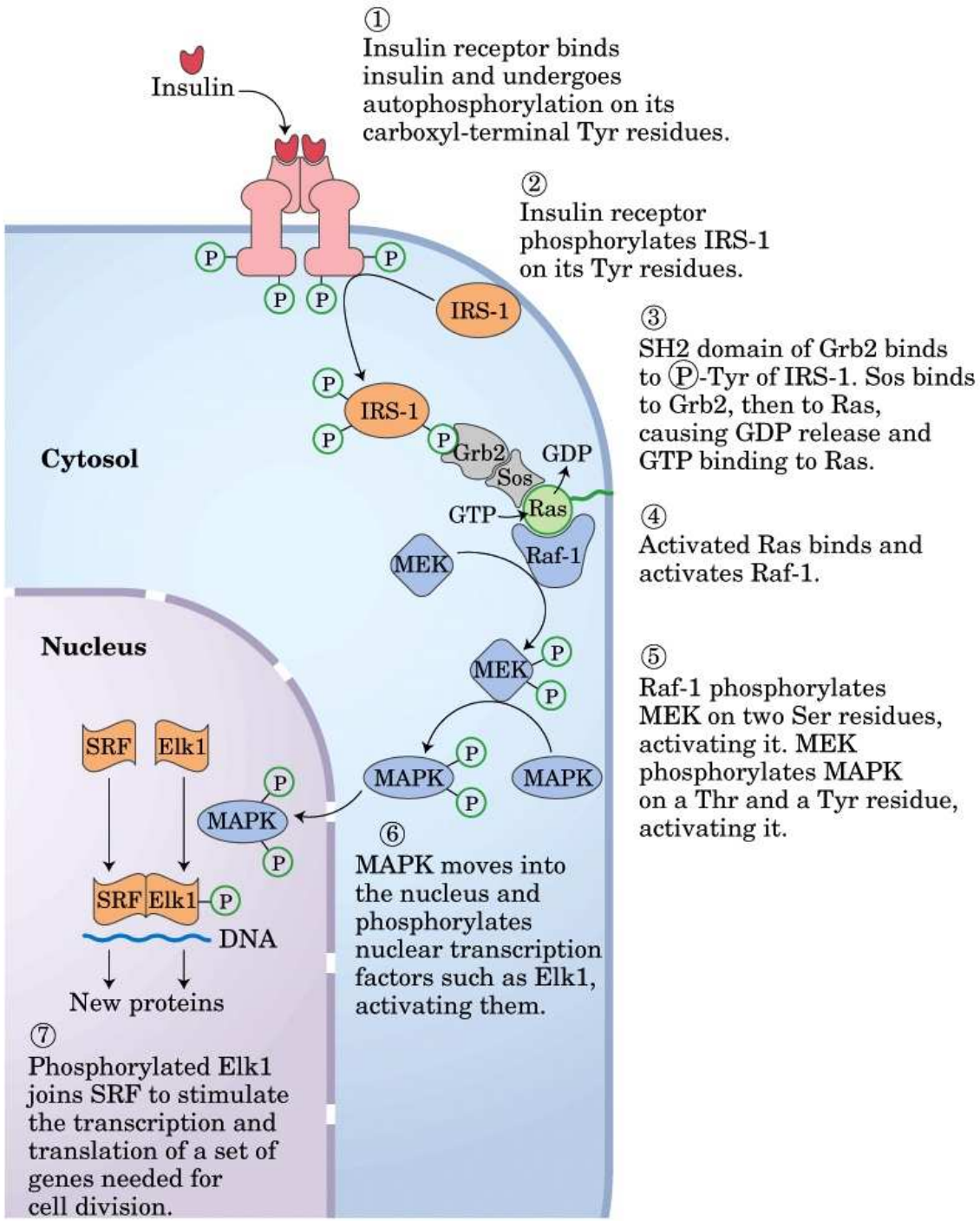
↑ síntese de glicogênio (fígado e músculo)

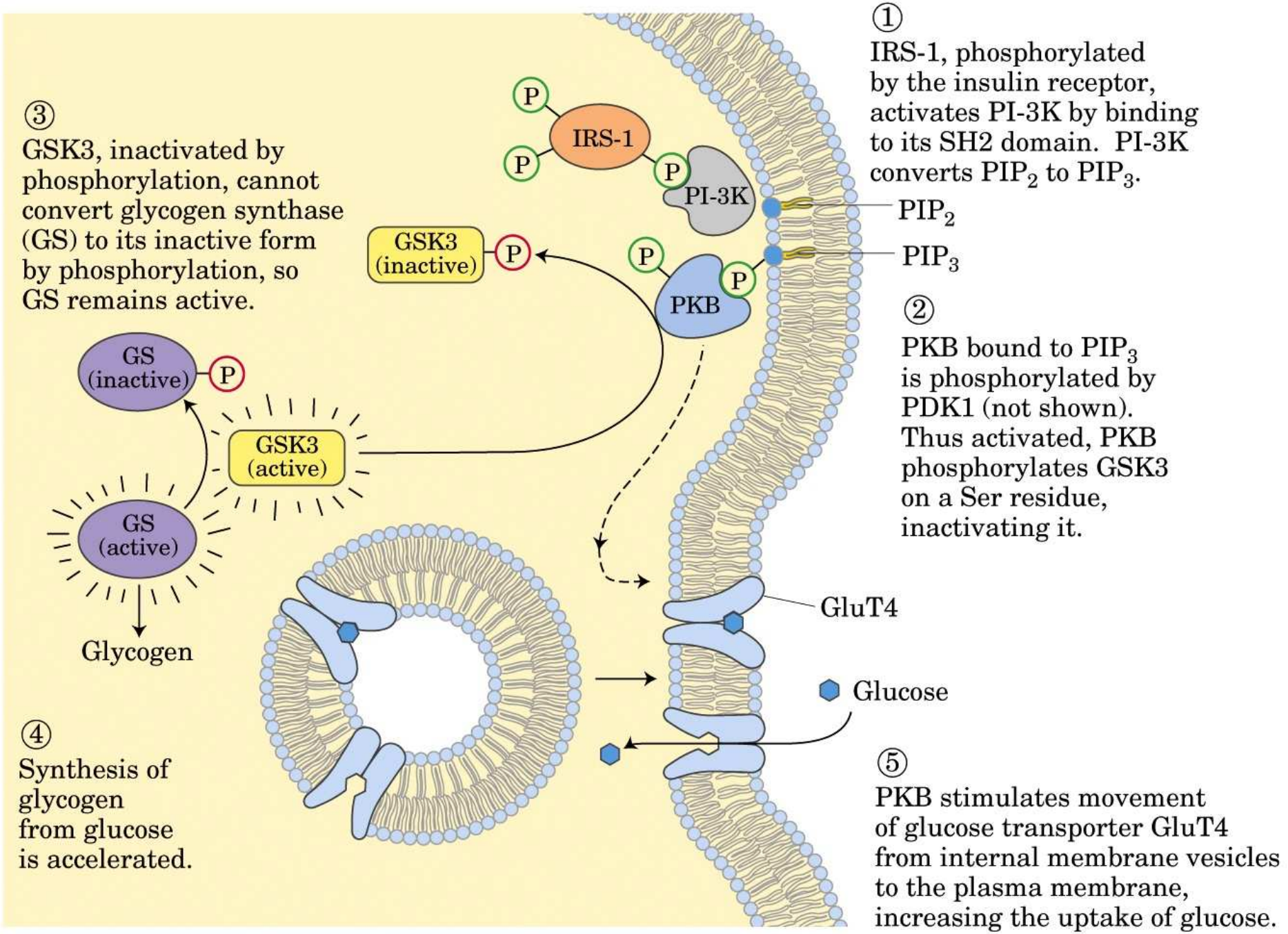
↓ degradação do glicogênio (fígado e músculo)

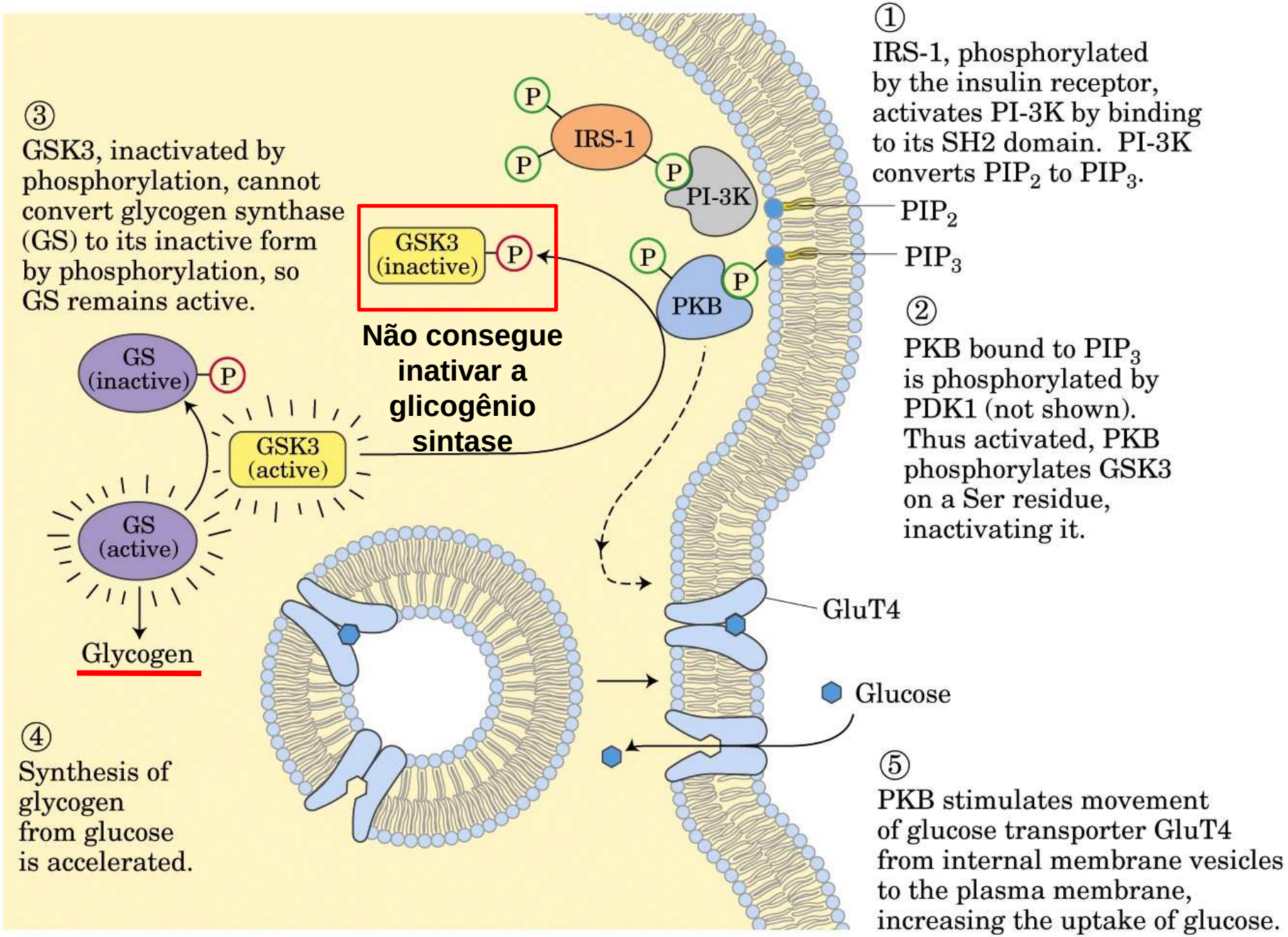
↑ glicólise, produção de acetil-CoA (fígado e músculo)

↑ síntese de ácidos graxos (fígado)

↑ síntese de triacilgliceróis (tecido adiposo)

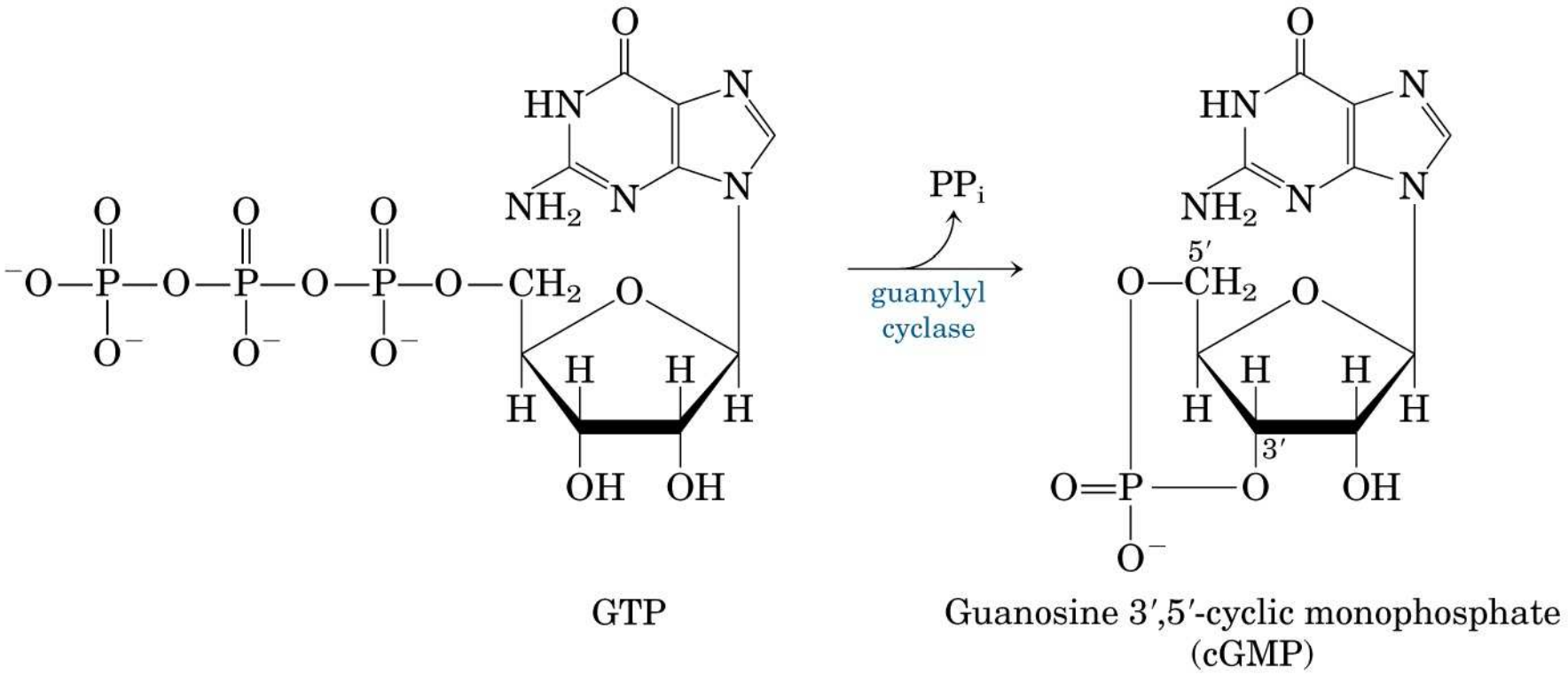

Insulin
① Insulin receptor binds insulin and undergoes autophosphorylation on its carboxyl-terminal Tyr residues.
② Insulin receptor phosphorylates IRS-1 on its Tyr residues.
③ SH2 domain of Grb2 binds to Ⓟ-Tyr of IRS-1. Sos binds to Grb2, then to Ras, causing GDP release and GTP binding to Ras.
④ Activated Ras binds and activates Raf-1.
⑤ Raf-1 phosphorylates MEK on two Ser residues, activating it. MEK phosphorylates MAPK on a Thr and a Tyr residue, activating it.
⑥ MAPK moves into the nucleus and phosphorylates nuclear transcription factors such as Elk1, activating them.
⑦ Phosphorylated Elk1 joins SRF to stimulate the transcription and translation of a set of genes needed for cell division.
Cytosol
Nucleus
IRS-1
Grb2
Sos
GDP
GTP
Ras
Raf-1
MEK
MAPK
SRF
Elk1
DNA
New proteins
P


①
IRS-1, phosphorylated by the insulin receptor, activates PI-3K by binding to its SH2 domain. PI-3K converts PIP_2 to PIP_3.
P
IRS-1
P
P
PI-3K
PIP_2
PIP_3
②
PKB bound to PIP_3 is phosphorylated by PDK1 (not shown). Thus activated, PKB phosphorylates GSK3 on a Ser residue, inactivating it.
P
P
PKB
③
GSK3, inactivated by phosphorylation, cannot convert glycogen synthase (GS) to its inactive form by phosphorylation, so GS remains active.
GSK3 (inactive)
P
GS (inactive)
P
GSK3 (active)
GS (active)
Glycogen
GluT4
Glucose
④
Synthesis of glycogen from glucose is accelerated.
⑤
PKB stimulates movement of glucose transporter GluT4 from internal membrane vesicles to the plasma membrane, increasing the uptake of glucose.


①
IRS-1, phosphorylated by the insulin receptor, activates PI-3K by binding to its SH2 domain. PI-3K converts PIP_2 to PIP_3.
P
IRS-1
P
P
PI-3K
PIP_2
PIP_3
②
PKB bound to PIP_3 is phosphorylated by PDK1 (not shown). Thus activated, PKB phosphorylates GSK3 on a Ser residue, inactivating it.
P
P
PKB
③
GSK3, inactivated by phosphorylation, cannot convert glycogen synthase (GS) to its inactive form by phosphorylation, so GS remains active.
GSK3 (inactive)
P
Não consegue inativar a glicogênio sintase
GS (inactive)
P
GSK3 (active)
GS (active)
Glycogen
GluT4
Glucose
④
Synthesis of glycogen from glucose is accelerated.
⑤
PKB stimulates movement of glucose transporter GluT4 from internal membrane vesicles to the plasma membrane, increasing the uptake of glucose.

3 Receptores enzimáticos

Receptores com atividade de guanilil ciclase



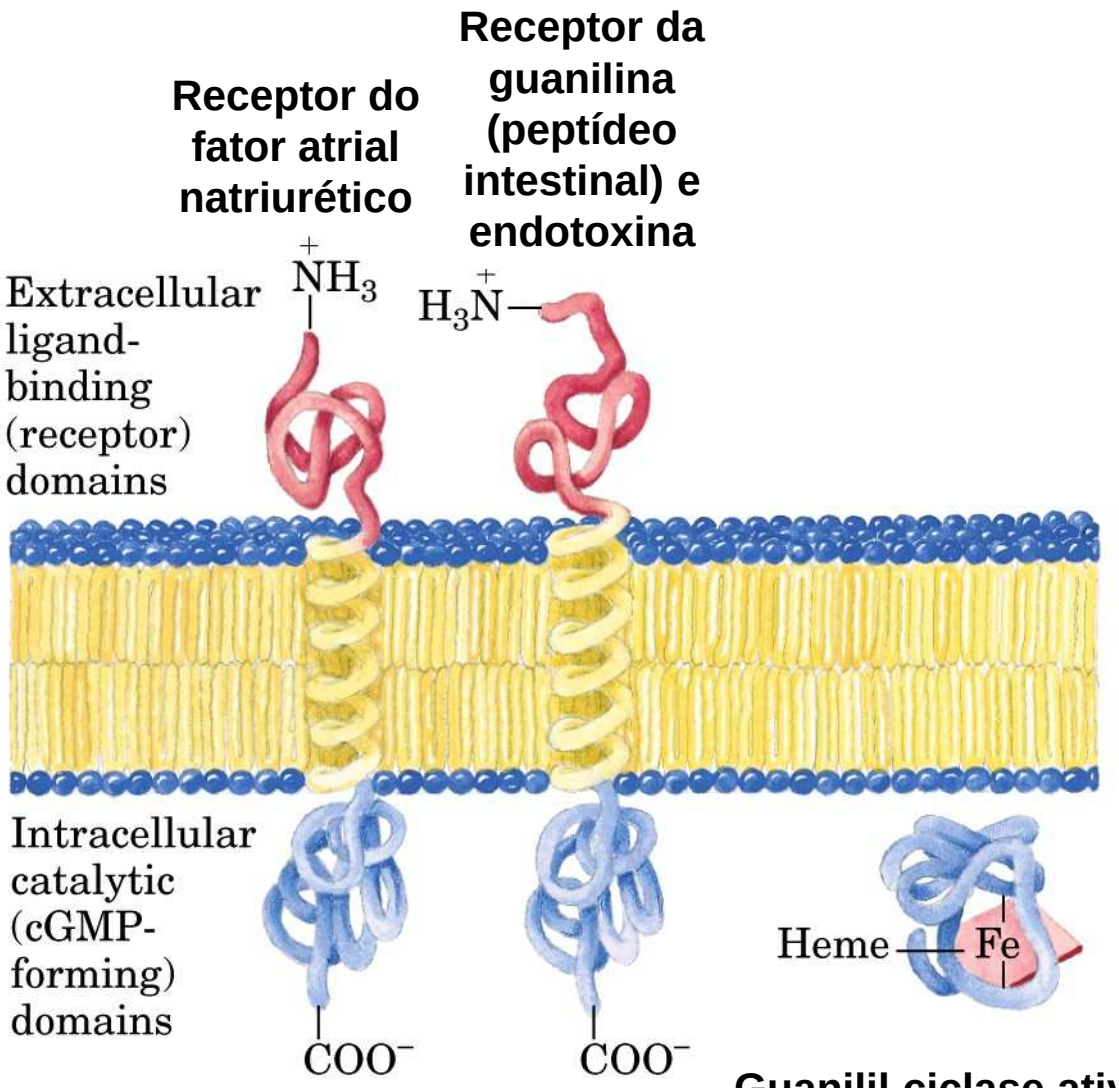
GTP

Guanosine 3′,5′-cyclic monophosphate (cGMP)

Ação do cGMP – segundo mensageiro

- **Rim e intestino:**
  - **Mudanças no transporte iônico e retenção de água**
- **Cérebro:**
  - **Desenvolvimento**
  - **Funcionamento no adulto**
- **Coração:**
  - **Relaxamento (reduz a força de contração do músculo cardíaco, pelo estímulo da bomba de extrusão de Ca^{++})**

Guanilil ciclase



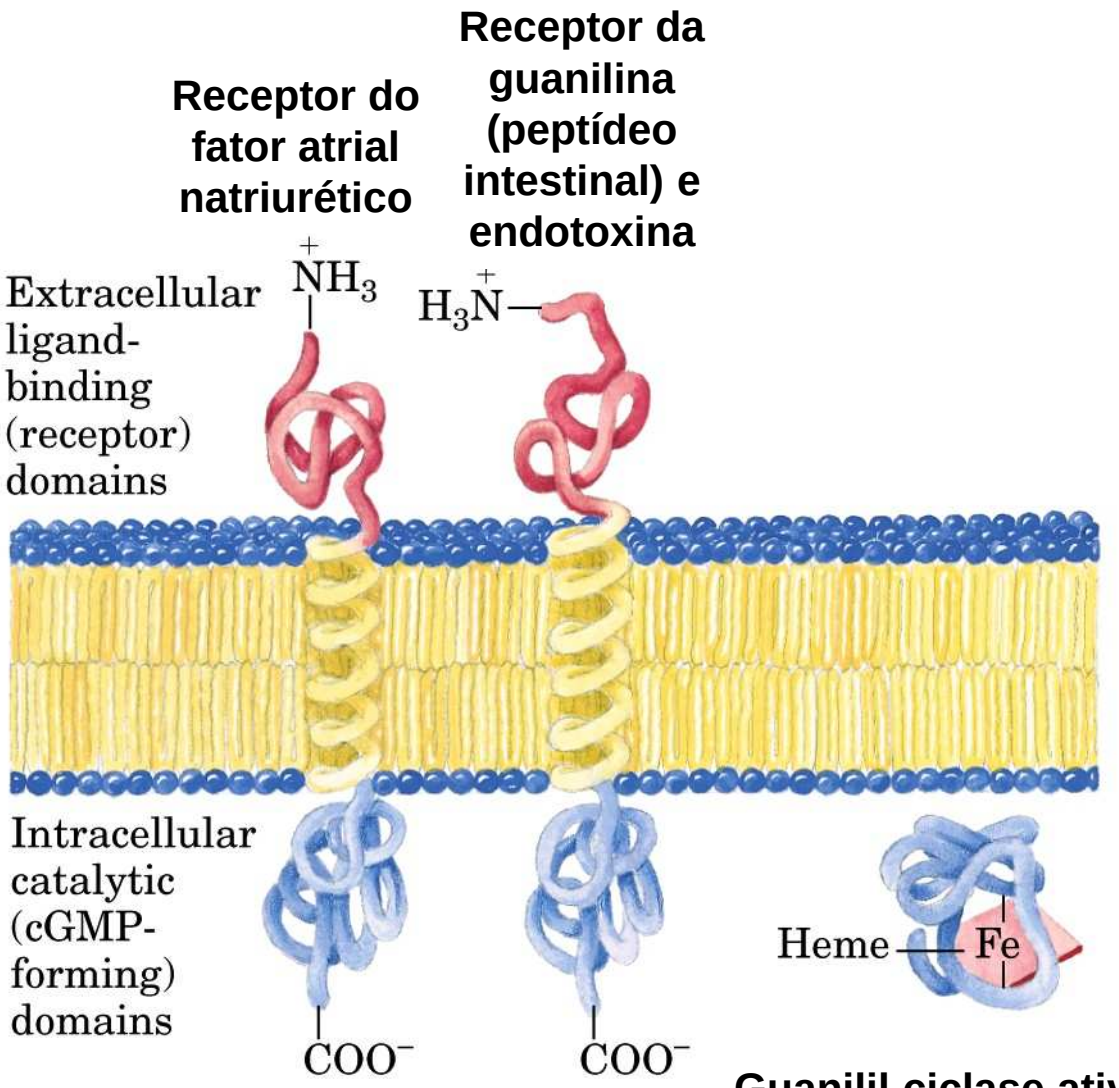
Guanilil ciclases de membrana

Guanilil ciclase ativada por NO solúvel

Fator atrial natriurético

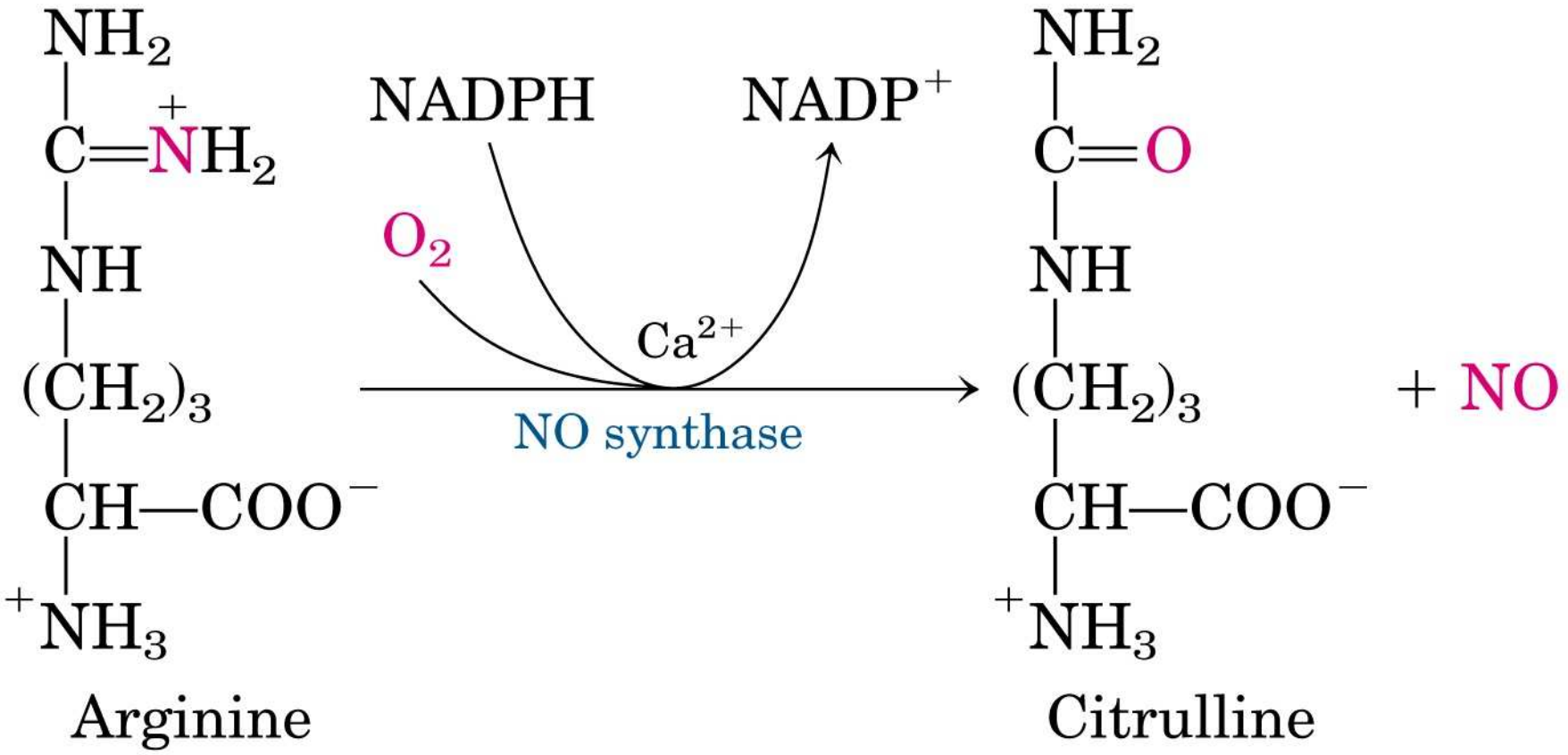
- Liberado pelas células do átrio do coração quando ele está distendido pelo volume de sangue aumentado
- Rim: ativa guanilil ciclase nas células dos ductos coletores
  - Aumento da excreção de Na^+ >>> aumento excreção de água
- Musculatura lisa dos vasos sanguíneos:
  - Relaxamento (vasodilatação) >>> aumento do fluxo sanguíneo >>> diminuição da pressão arterial

Guanilil ciclase



Guanilil ciclases de membrana

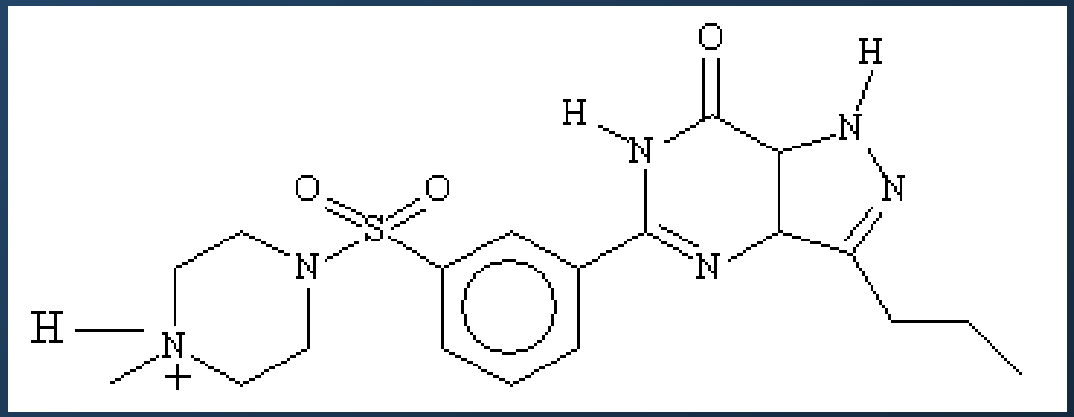
Guanilil ciclase ativada por NO solúvel



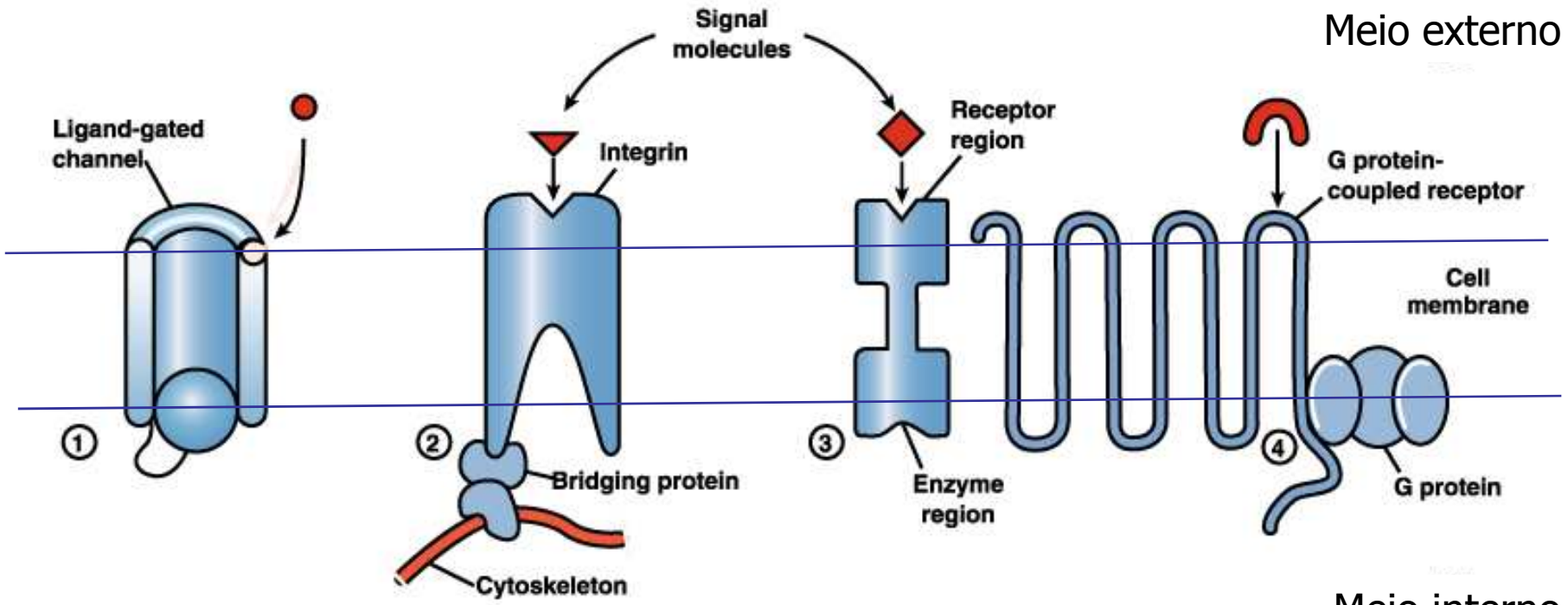
Tratamento de angina: nitrovasodilatadores

Degradação do cGMP

- **Fosfodiesterase**
  - **Converte cGMP a 5’-GMP**
  - **Várias isoformas**
- **Sildenafil (Viagra)**



Quatro classes de receptores de membrana



Canais iônicos

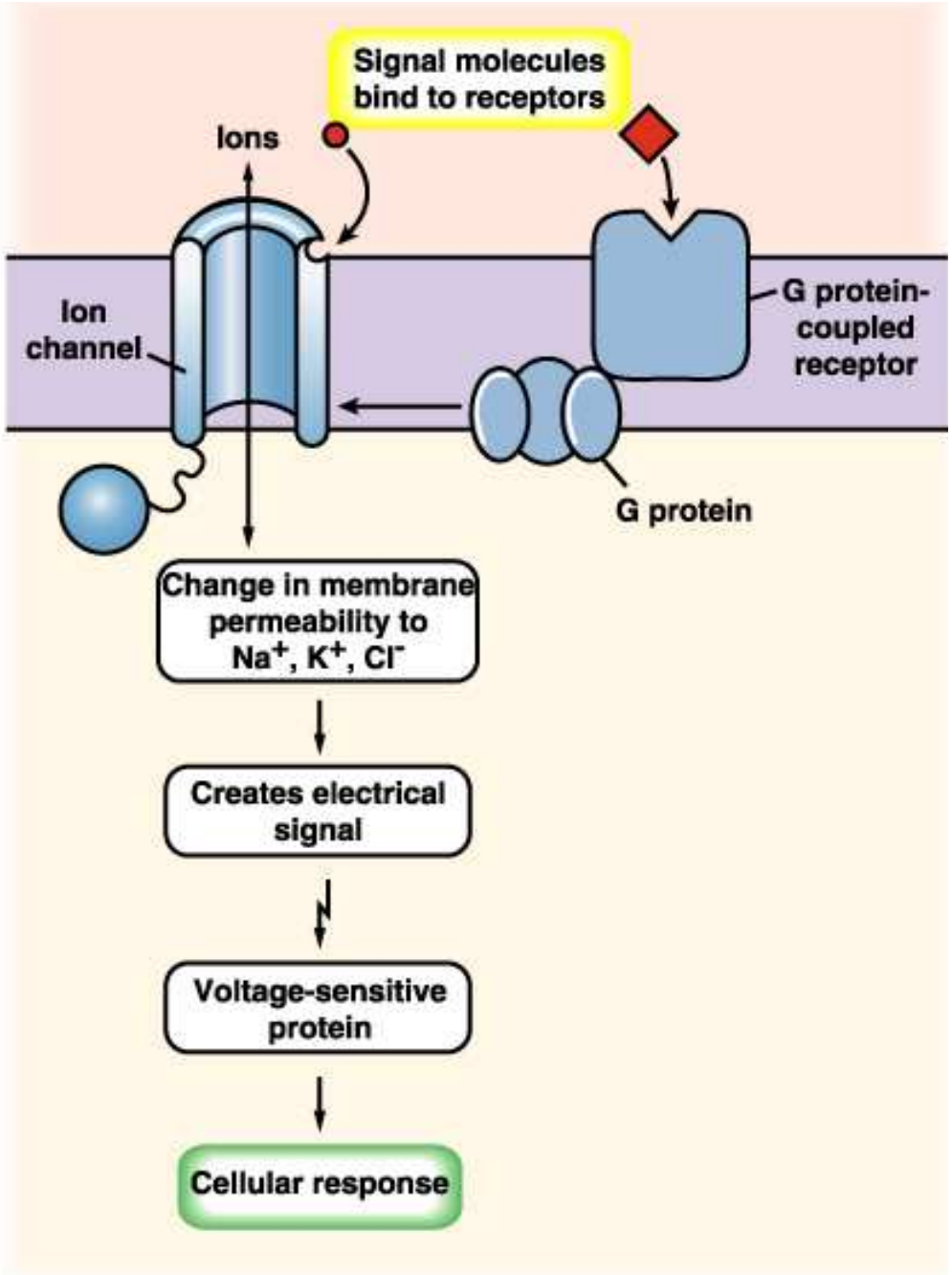
Integrinas

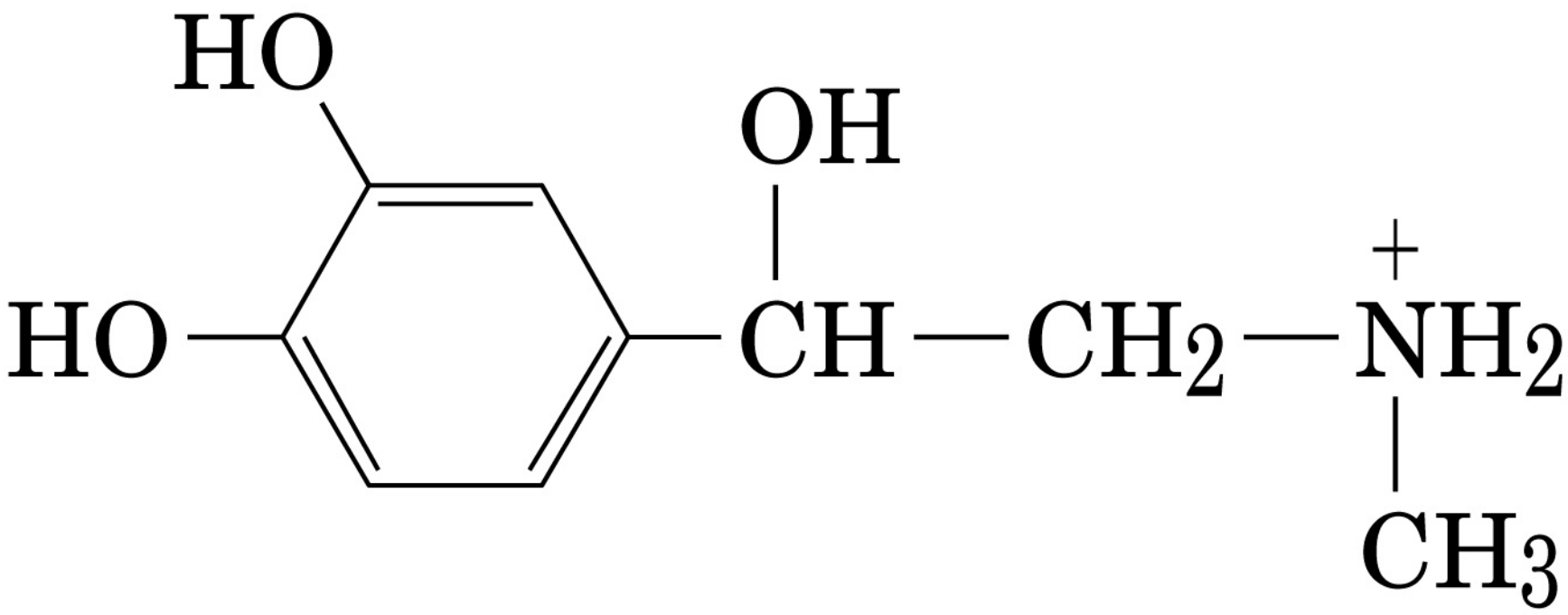
Receptores enzimáticos

Receptores ligados a proteína G

Receptores ligados a proteínas G

- **3 componentes essenciais:**
  - **Receptor de membrana com 7 segmentos transmembrana**
  - **Proteína G (proteína que se liga a nucleotídeo de guanosina)**
  - **Enzima na membrana que gera um segundo mensageiro intracelular**

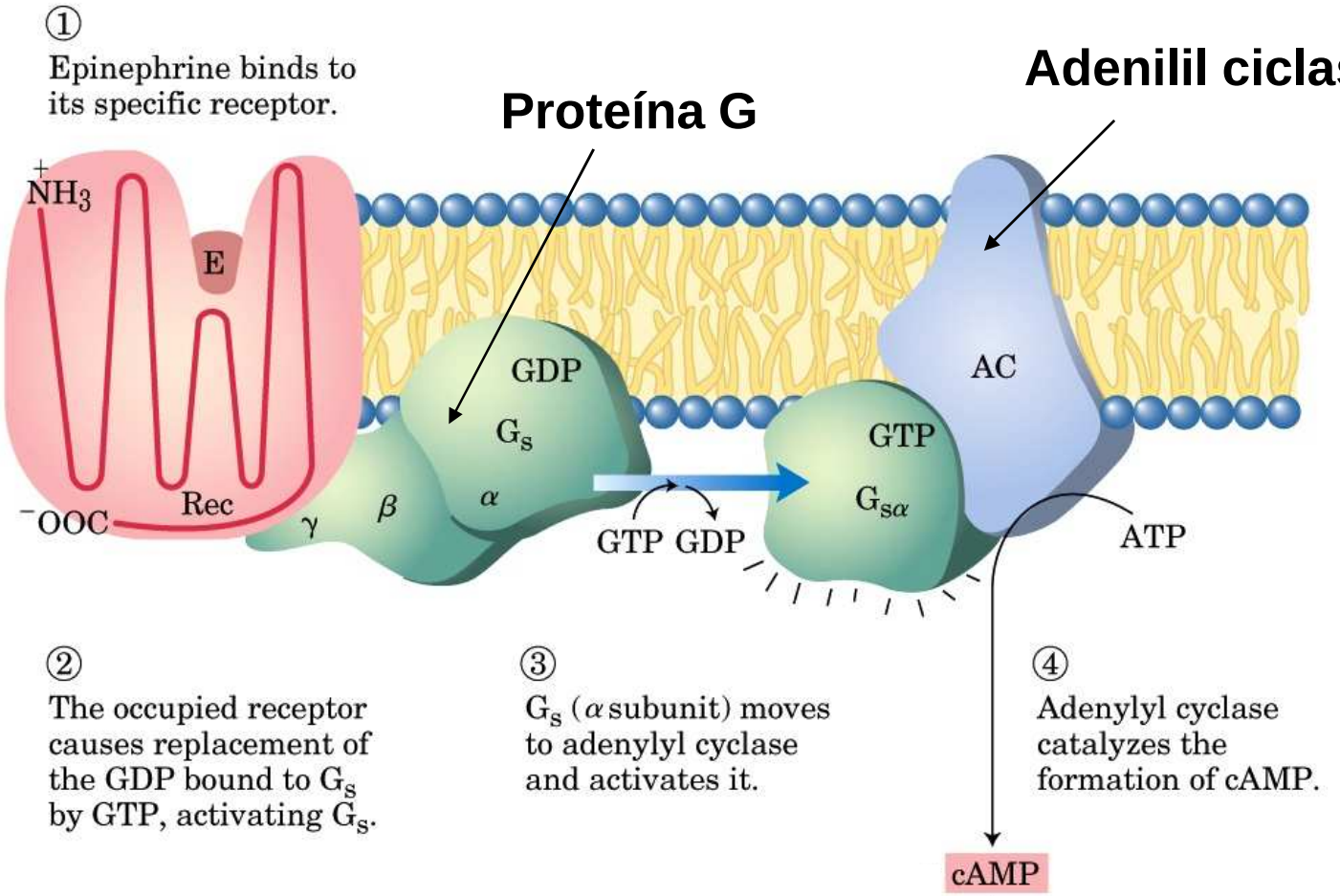

Signal molecules bind to receptors
Ions
Ion channel
G protein-coupled receptor
G protein
Change in membrane permeability to Na^+, K^+, Cl^-
Creates electrical signal
Voltage-sensitive protein
Cellular response

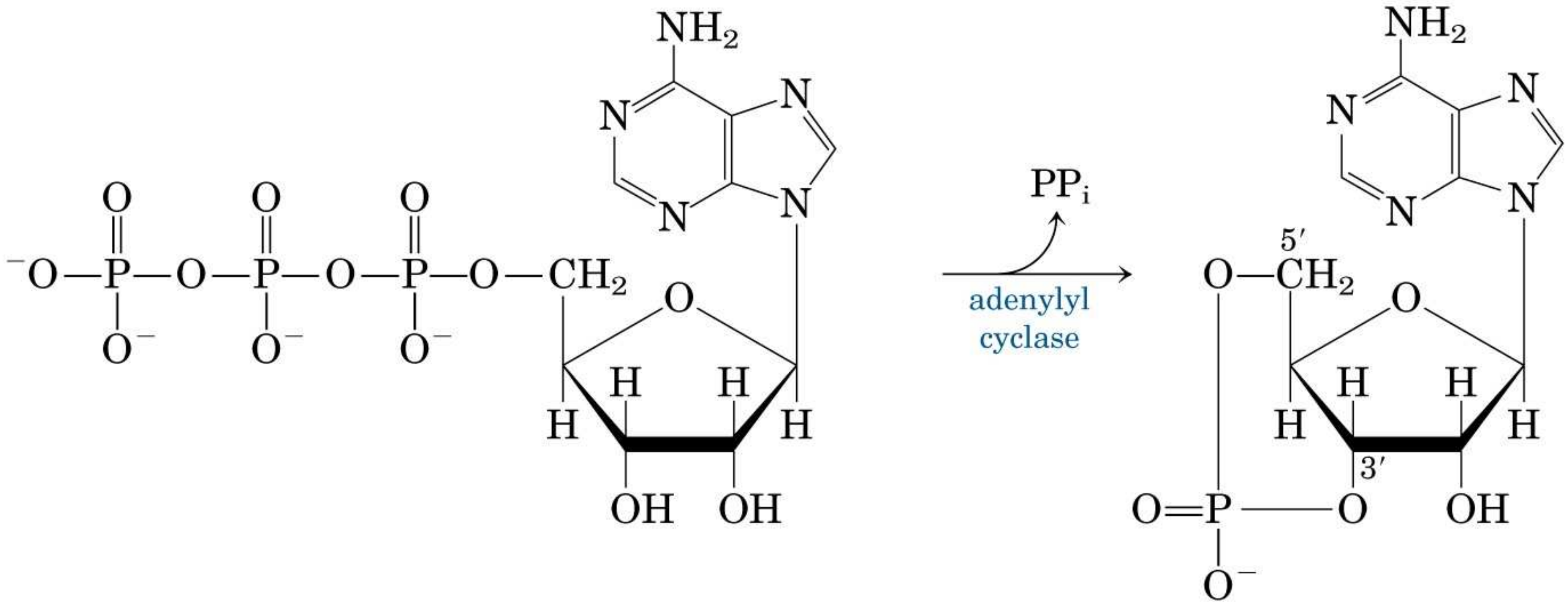


Epinefrina

(adrenalina)

Receptor serpentina





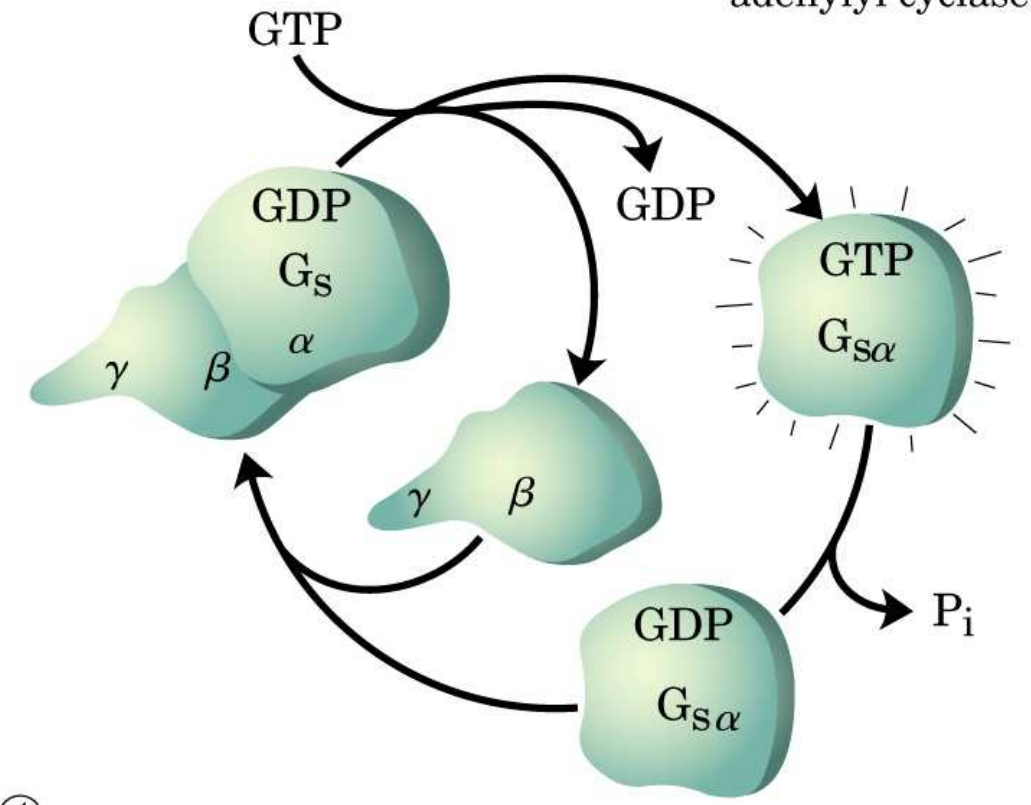
ATP

Adenosine 3′,5′-cyclic monophosphate (cAMP)

① G_S with GDP bound is turned off; it cannot activate adenylyl cyclase.

② Contact of G_S with hormone-receptor complex causes displacement of bound GDP by GTP.

③ G_S with GTP bound dissociates into α and βγ subunits. $G_{S\alpha}$-GTP is turned on; it can activate adenylyl cyclase.

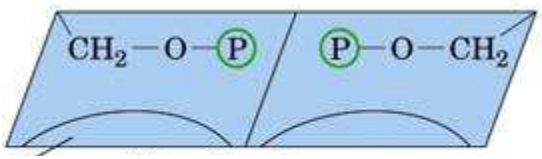


④ GTP bound to $G_{S\alpha}$ is hydrolyzed by the protein's intrinsic GTPase; $G_{S\alpha}$ thereby turns itself off. The inactive α subunit reassociates with the β, γ subunits.

Adrenalina ativa a glicogênio fosforilase

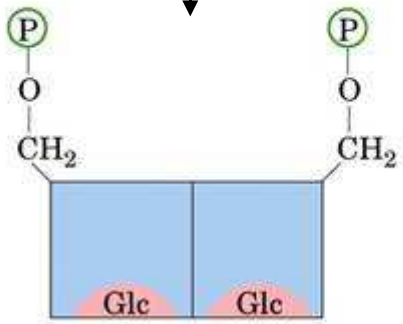
Forma ativa = “a”

Forma menos ativa = “b”



Fosforilase *a*

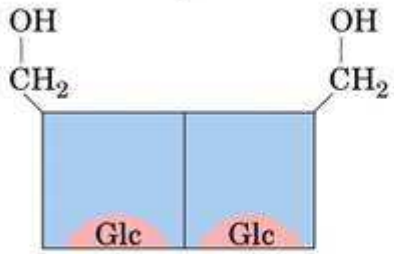
2 glicoses



[glicose]

Fosforilase *a* fosfatase

2 Pi

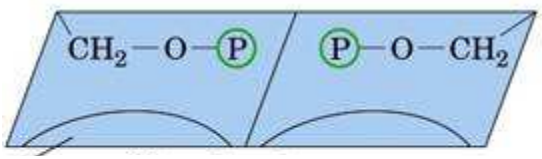


Fosforilase *b*

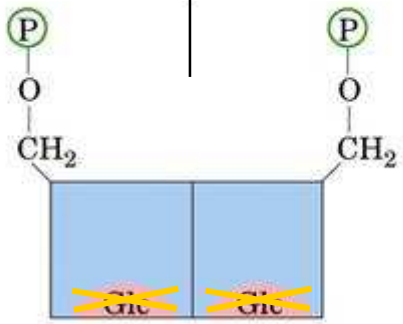
Adrenalina ativa a glicogênio fosforilase

Forma ativa = “a”

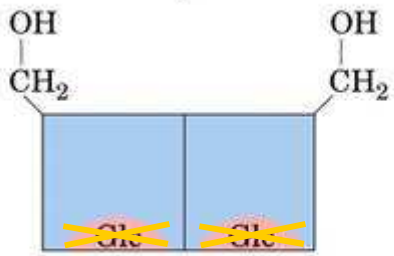
Forma menos ativa = “b”



Fosforilase *a*



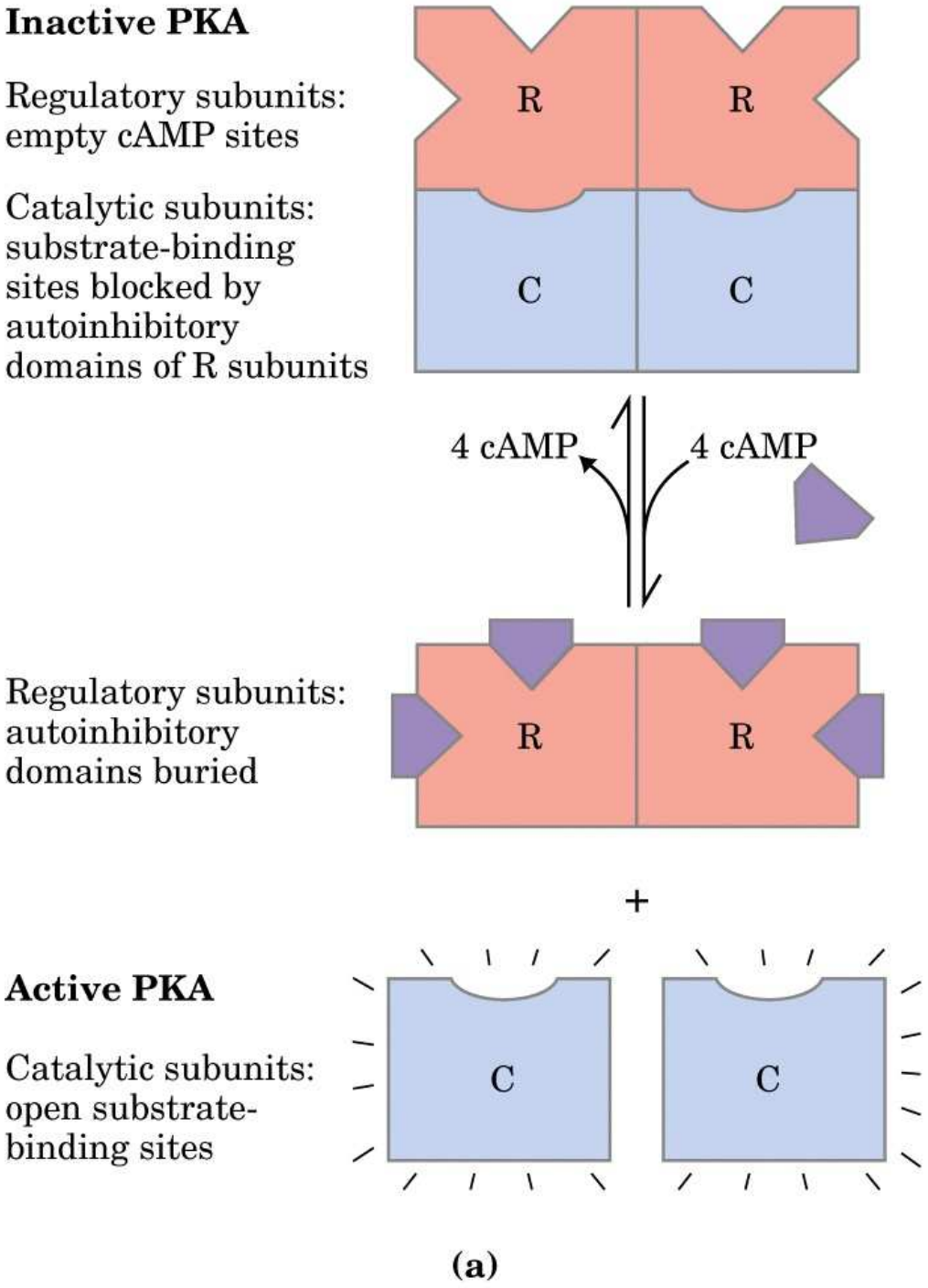
Fosforilase *b* quinase

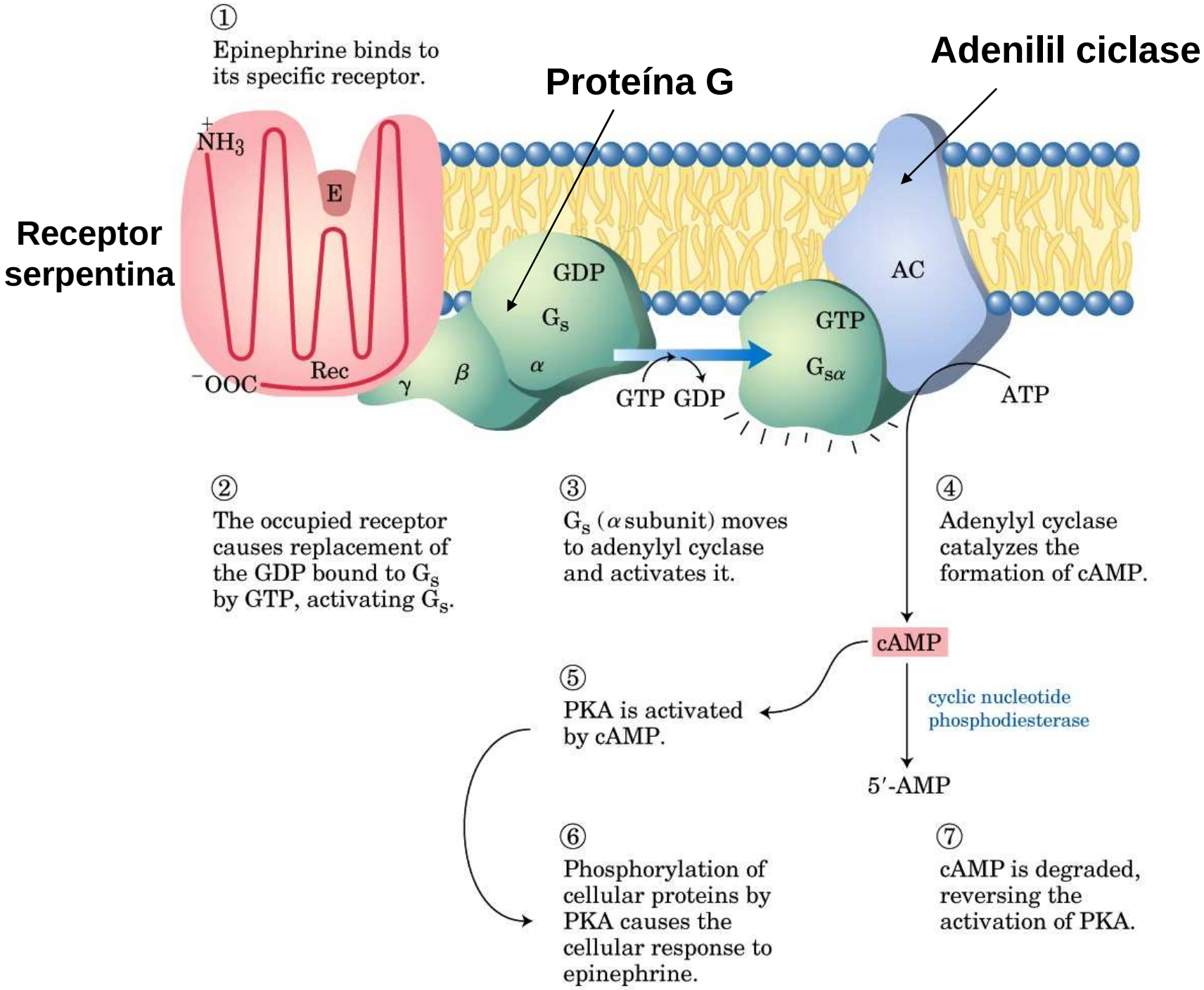


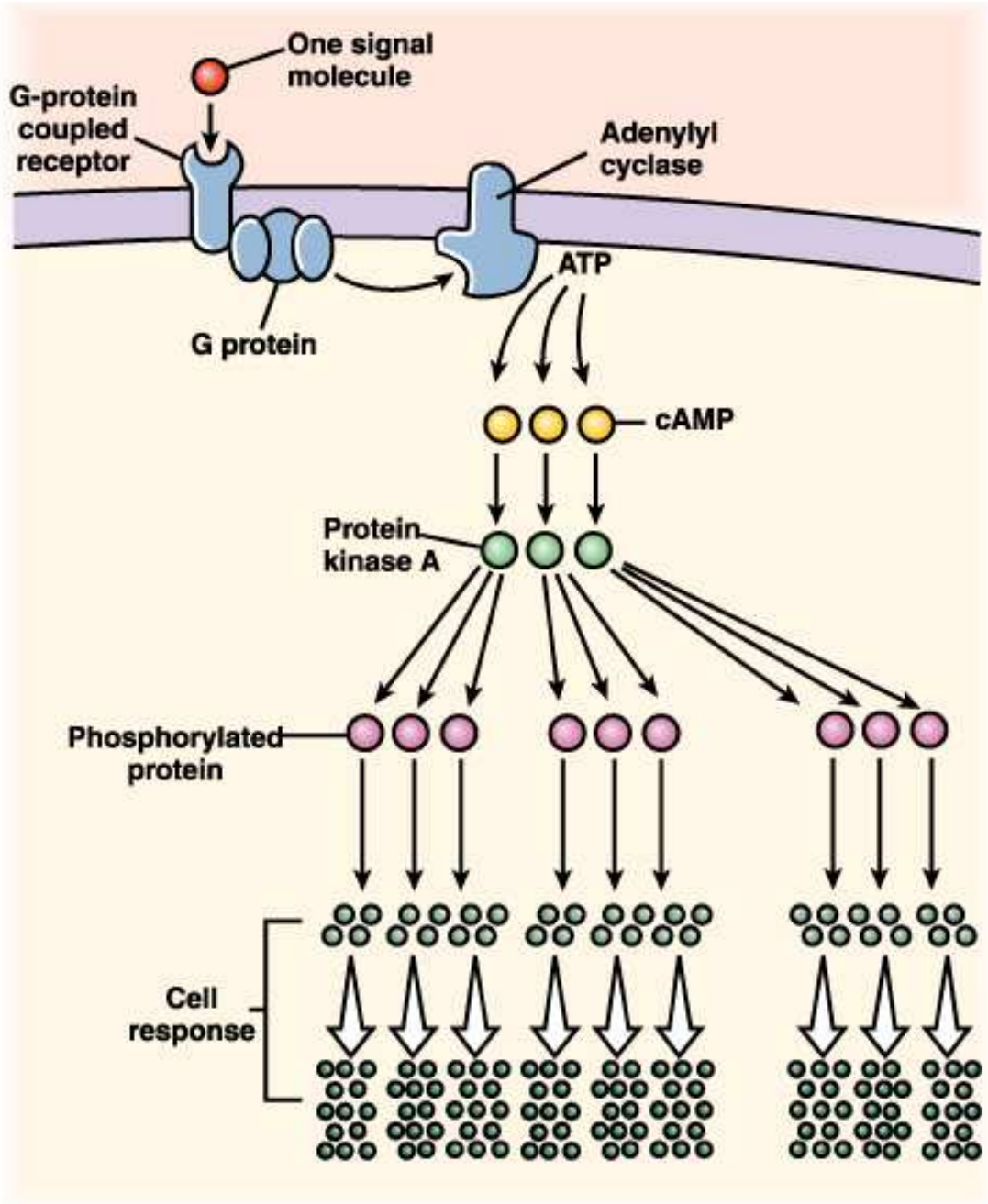
Fosforilase *b*

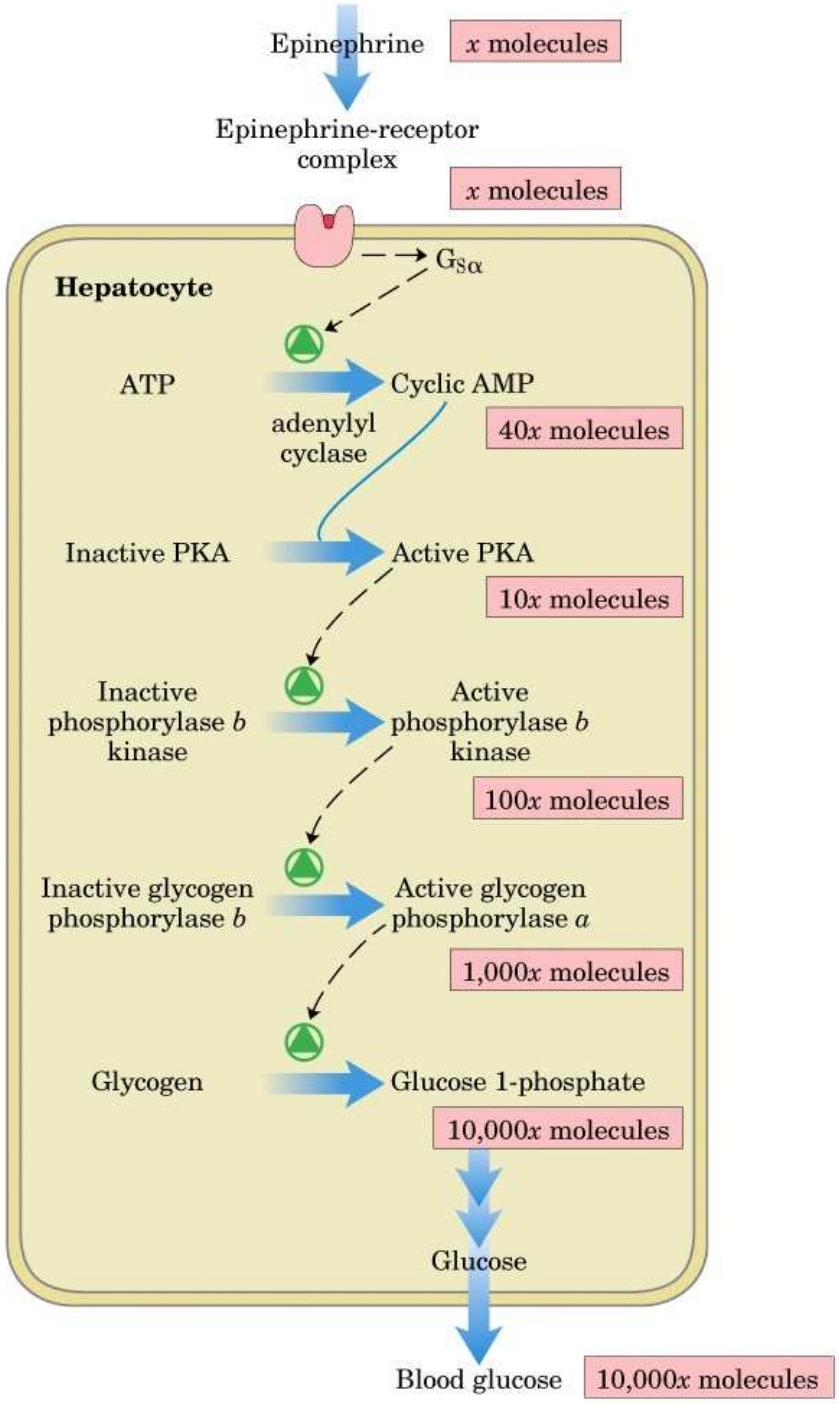
[glicose]

Fosforilase b quinase é ativada pela proteína quinase dependente de cAMP (PKA)




Receptor serpentina
Proteína G
Adenilil ciclase
① Epinephrine binds to its specific receptor.
+NH3
E
Rec
−OOC
GDP
Gs
α
β
γ
GTP GDP
GTP
Gsα
AC
ATP
② The occupied receptor causes replacement of the GDP bound to Gs by GTP, activating Gs.
③ Gs (α subunit) moves to adenylyl cyclase and activates it.
④ Adenylyl cyclase catalyzes the formation of cAMP.
cAMP
⑤ PKA is activated by cAMP.
cyclic nucleotide phosphodiesterase
5′-AMP
⑥ Phosphorylation of cellular proteins by PKA causes the cellular response to epinephrine.
⑦ cAMP is degraded, reversing the activation of PKA.

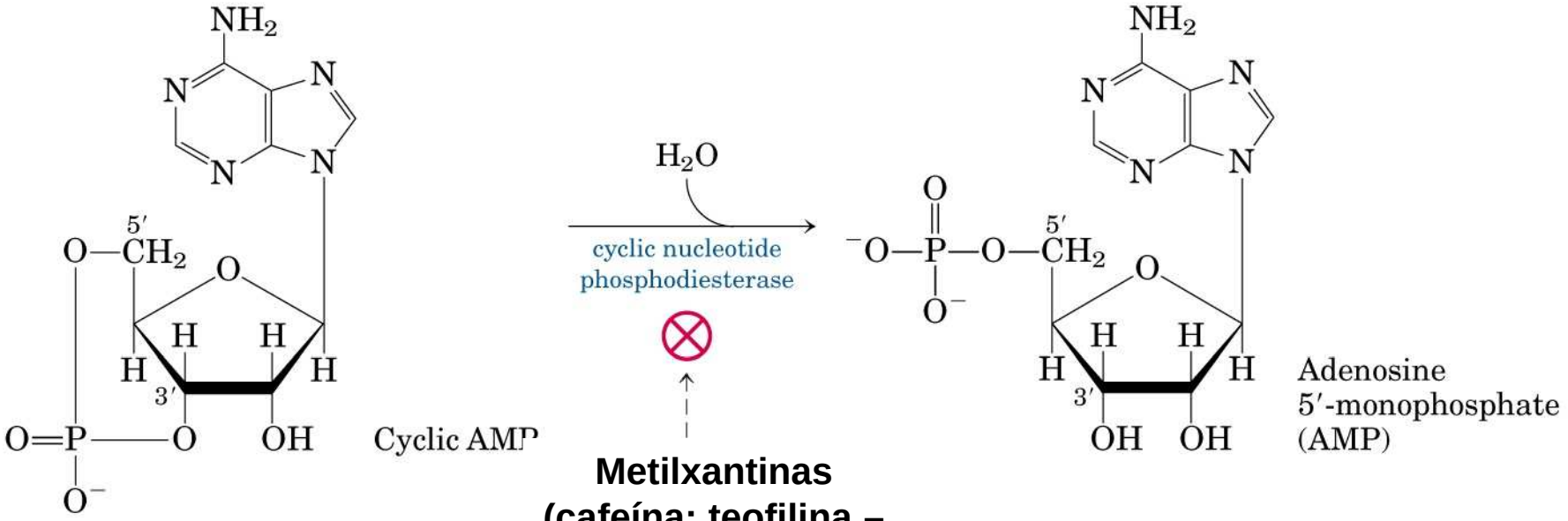

One signal molecule
G-protein coupled receptor
Adenylyl cyclase
ATP
G protein
cAMP
Protein kinase A
Phosphorylated protein
Cell response


Epinephrine
x molecules
Epinephrine-receptor complex
x molecules
Hepatocyte
$G_{s\alpha}$
ATP
Cyclic AMP
adenylyl cyclase
40x molecules
Inactive PKA
Active PKA
10x molecules
Inactive phosphorylase b kinase
Active phosphorylase b kinase
100x molecules
Inactive glycogen phosphorylase b
Active glycogen phosphorylase a
1,000x molecules
Glycogen
Glucose 1-phosphate
10,000x molecules
Glucose
Blood glucose
10,000x molecules

Algumas enzimas reguladas pela fosforilação dependente de cAMP (PKA)

Enzima	Via metabólica
Glicogênio sintase	Síntese de glicogênio
Fosforilase *b* quinase	Degradação do glicogênio
Piruvato quinase	Glicólise
Lipase sensível a hormônio	Mobilização de triacilgliceróis e oxidação de ácidos graxos
Histona H1	Condensação do DNA
Histona H2B	Condensação do DNA

Degradação do cAMP



Metilxantinas (cafeína; teofilina – chá)

Alguns sinais que utilizam cAMP como segundo mensageiro celular

ACTH

Dopamina

Adrenalina

Hormônio folículo-estimulante (FSH)

Glucagon

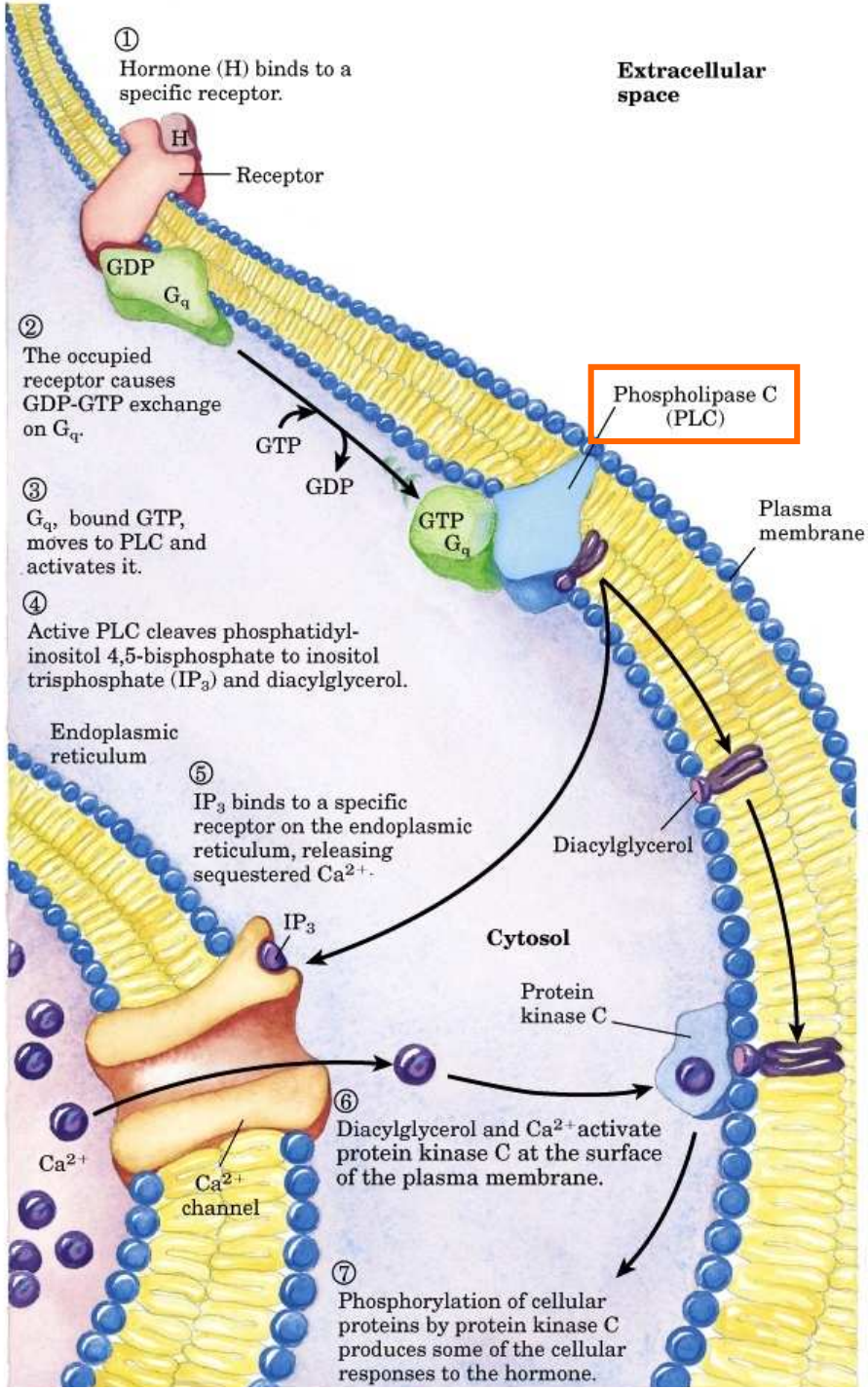
Paratormônio

Prostaglandina E1, E2

Somatostatina

Hormônio tireóideo estimulante (TSH)

Histamina (H2)



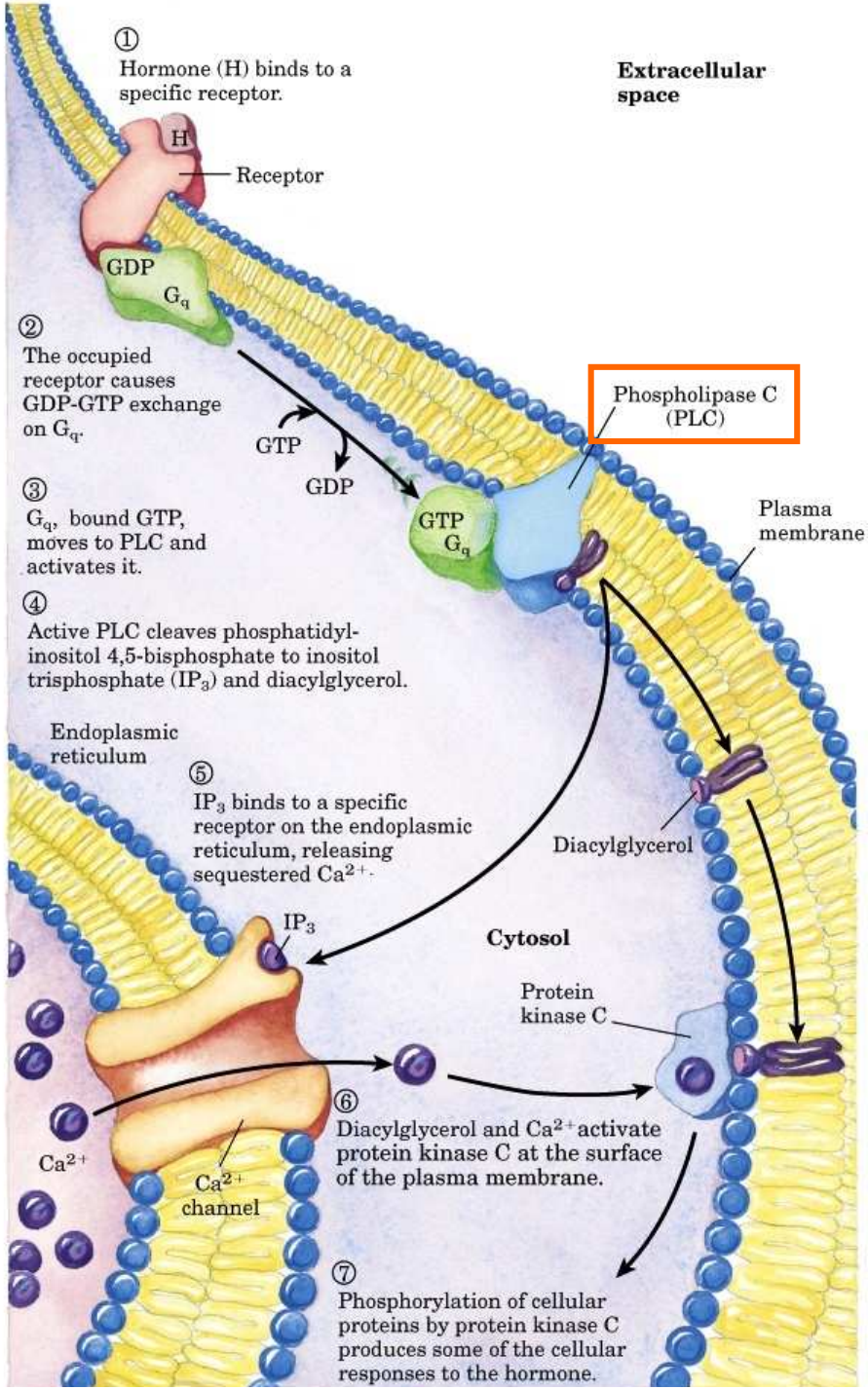
Fosfolipase C:

- atua num fosfolipídeo de membrana (fosfatidil-inositol 4,5 bifosfato)

. Diacilglicerol

. IP_3

} Segundos mensageiros



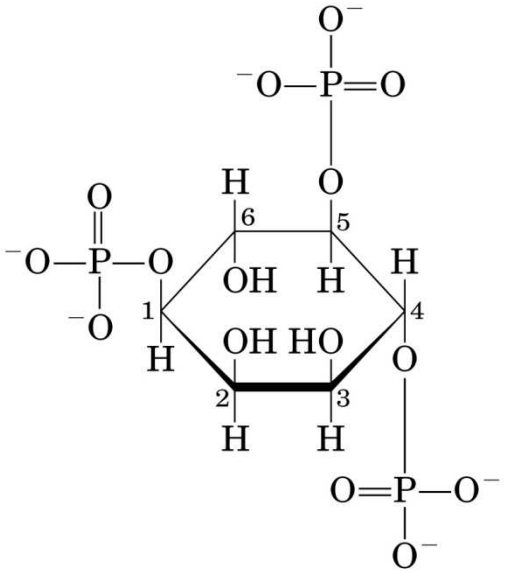
Fosfolipase C:

- atua num fosfolipídeo de membrana (fosfatidil-inositol 4,5 bifosfato)

  . Diacilglicerol

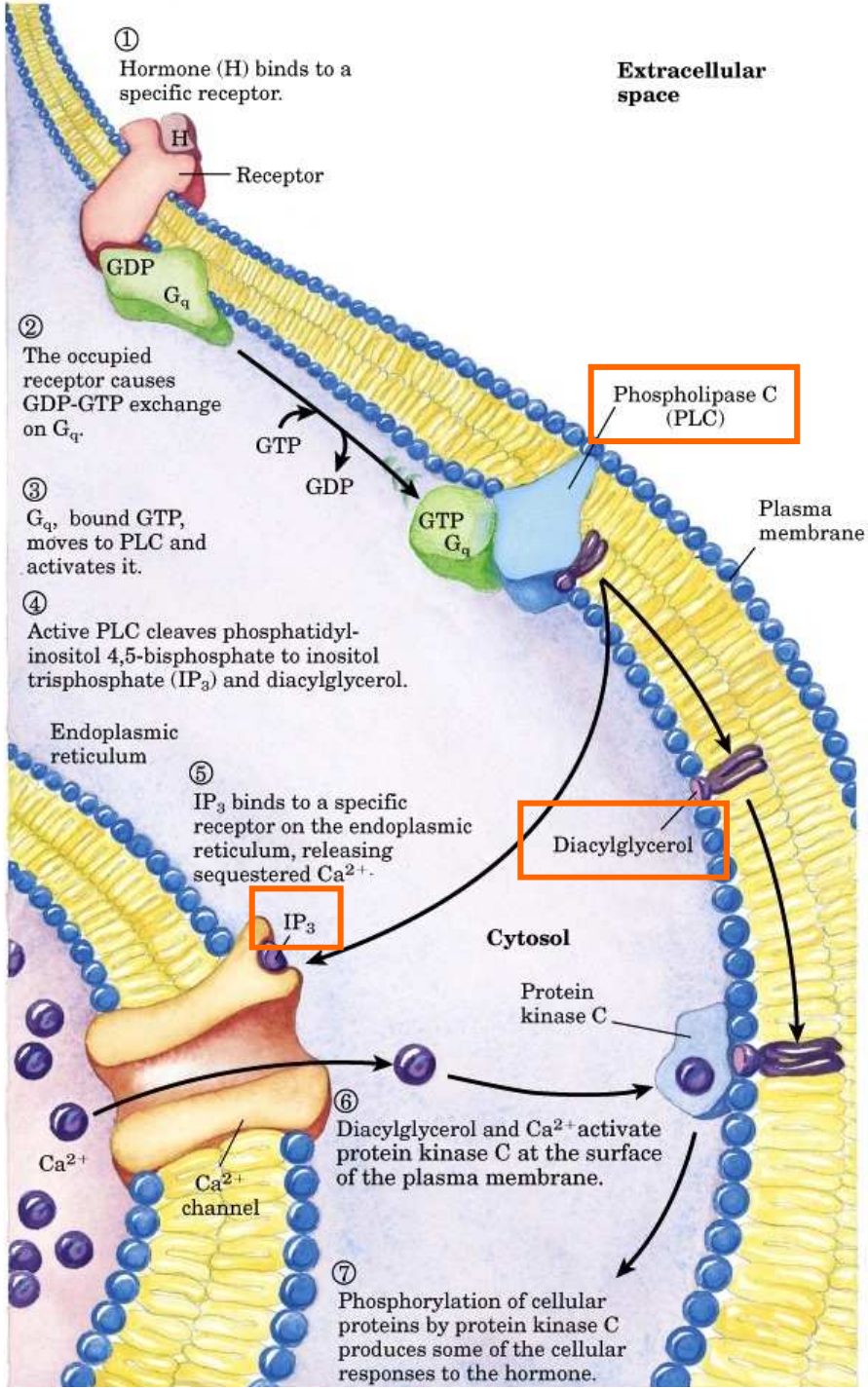
  . IP_3

  } Segundos mensageiros



Inositol 1,4,5-trisphosphate

IP_3



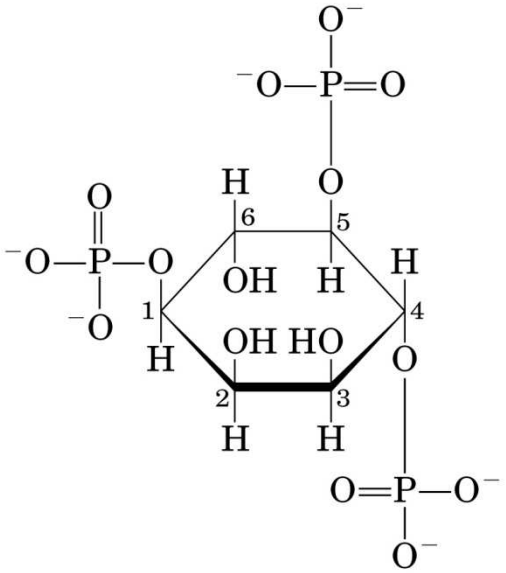
Fosfolipase C:

- atua num fosfolipídeo de membrana (fosfatidil-inositol 4,5 bifosfato)

  . Diacilglicerol

  . IP_3

  } Segundos mensageiros

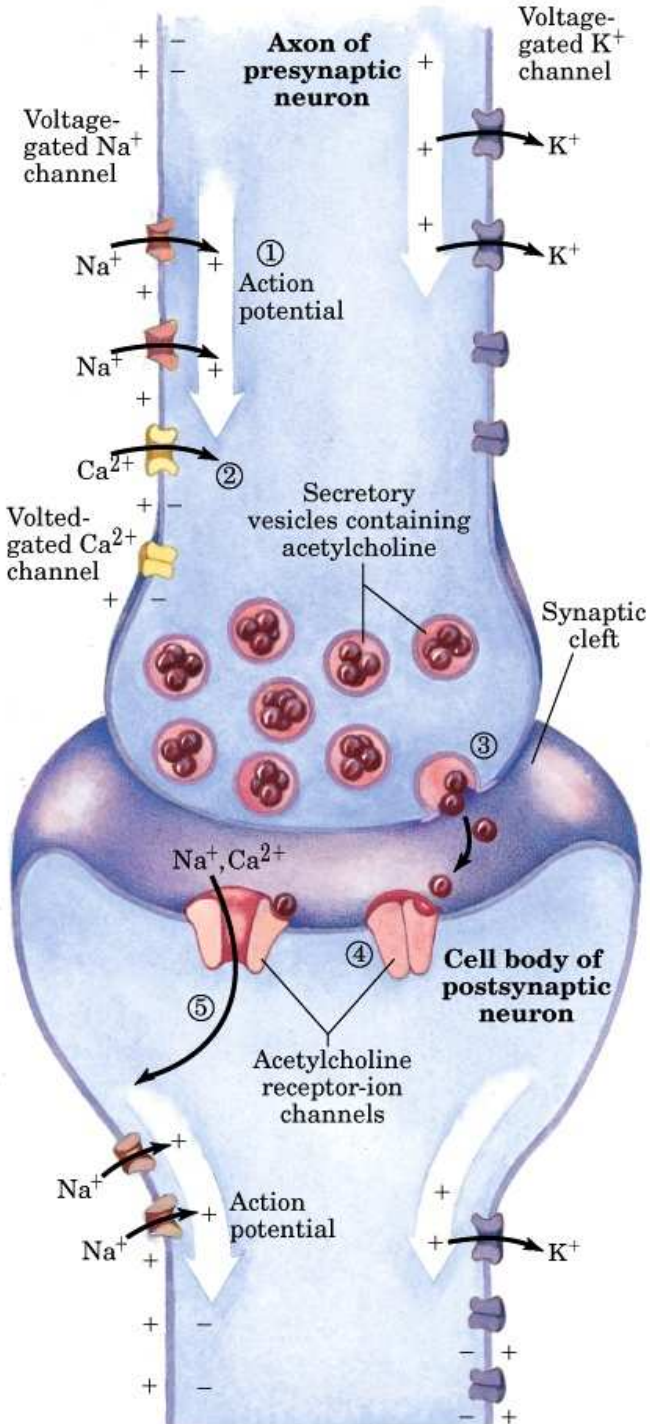


Inositol 1,4,5-trisphosphate

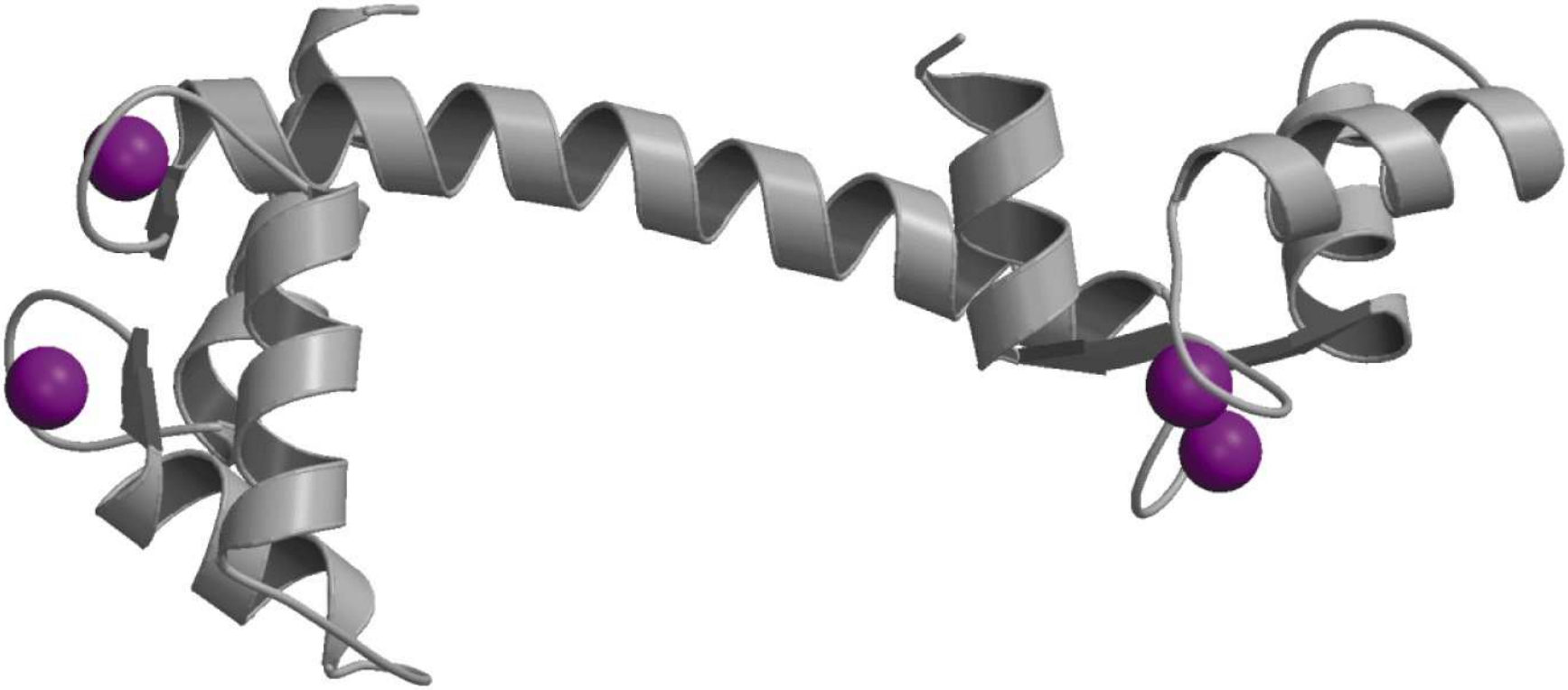
IP_3

Alguns sinais que atuam através de fosfolipase C e IP_3

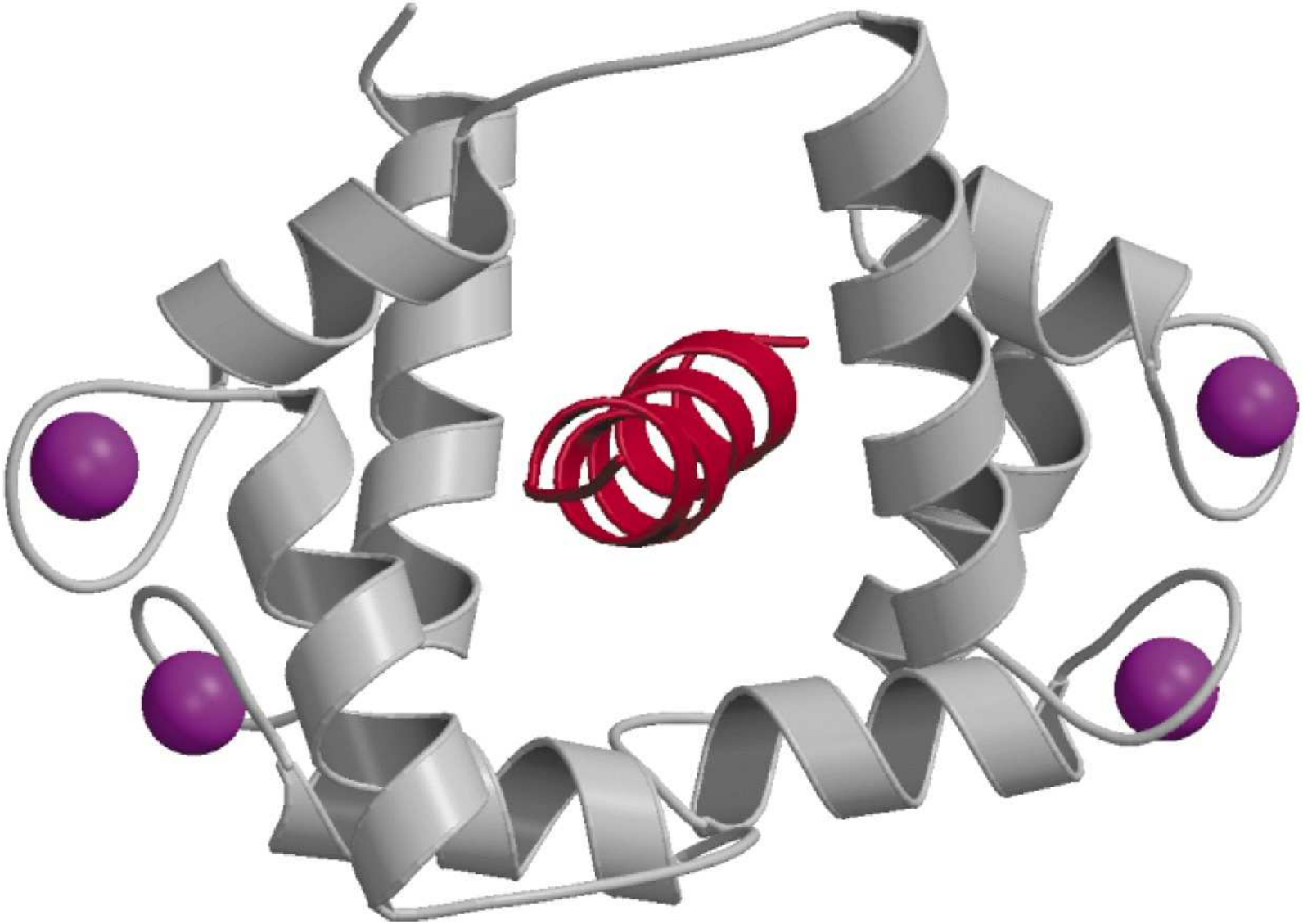
Acetilcolina (muscarínico M1)
Angiogenina
Angiotensina II
Auxina
Peptídeo liberador de gastrina
Hormônio liberador de gonadotrofina
Histamina (H1)
Luz
Oxitocina
Fator de crescimento derivado de plaquetas
Vasopressina


Axon of presynaptic neuron
Voltage-gated K+ channel
Voltage-gated Na+ channel
K+
K+
Na+
Na+
① Action potential
Ca2+
②
Volted-gated Ca2+ channel
Secretory vesicles containing acetylcholine
Synaptic cleft
③
Na+, Ca2+
④
Cell body of postsynaptic neuron
⑤
Acetylcholine receptor-ion channels
Na+
Na+
Action potential
K+

Calmodulina



(a)



(b)

Algumas proteínas reguladas por Ca^{++} e calmodulina

Adenilil ciclase (cérebro)

Proteínas quinase dependendes de Ca^{++}/ calmodulina

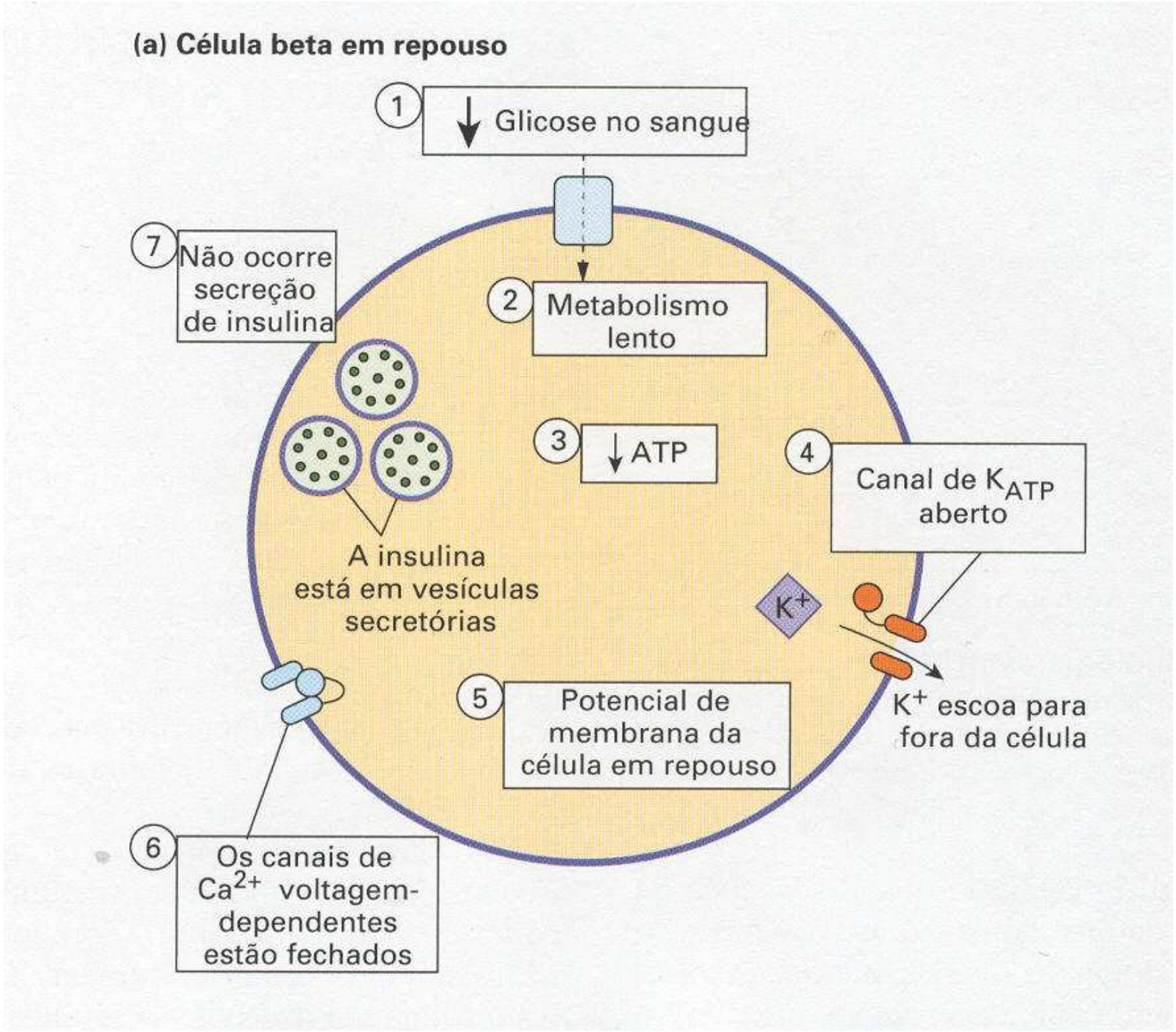
Fosfodiesterase do cAMP

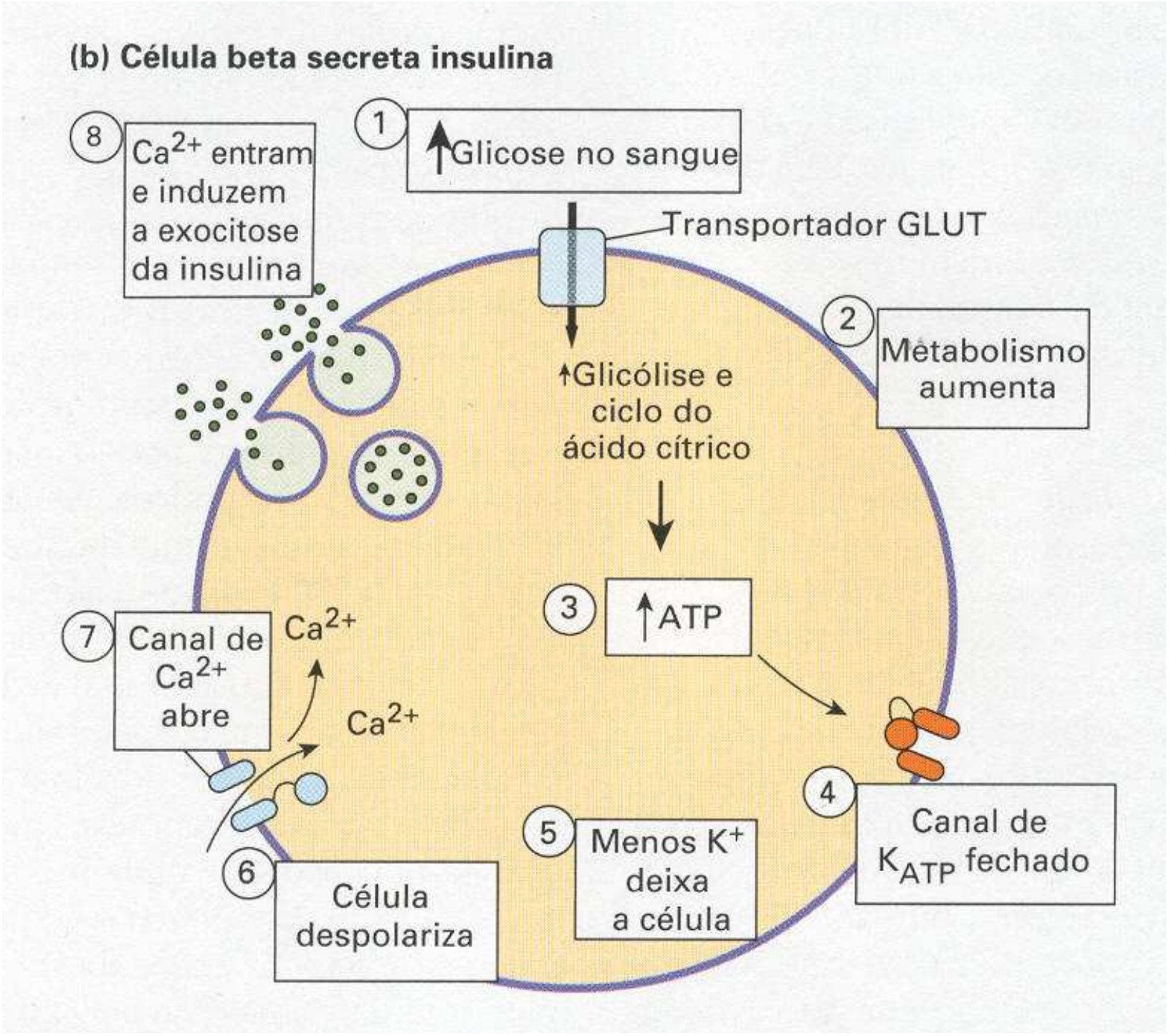
Quinases de cadeia leve de miosina

Quinase de NAD^{+}

Sintase de óxido nítrico

ATPase de Ca^{++} (bomba de Ca^{++})


(a) Célula beta em repouso
1
Glicose no sangue
7
Não ocorre secreção de insulina
2
Metabolismo lento
3
ATP
4
Canal de K_{ATP} aberto
A insulina está em vesículas secretórias
K^+
K^+ escoa para fora da célula
5
Potencial de membrana da célula em repouso
6
Os canais de Ca^{2+} voltagem-dependentes estão fechados


(b) Célula beta secreta insulina
1
Glicose no sangue
Transportador GLUT
2
Metabolismo aumenta
Glicólise e ciclo do ácido cítrico
3
ATP
4
Canal de K_{ATP} fechado
5
Menos K^+ deixa a célula
6
Célula despolariza
7
Canal de Ca^{2+} abre
Ca^{2+}
Ca^{2+}
8
Ca^{2+} entram e induzem a exocitose da insulina

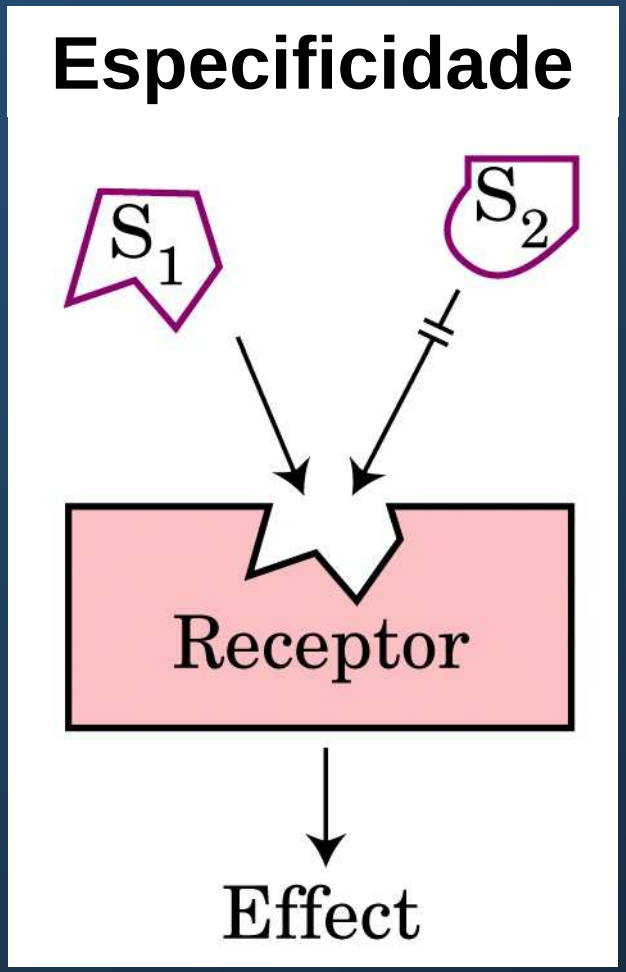
Enzimas amplificadoras

Enzima amplificadora	Localização celular	Ativada por	Converte	Em
Adenilil ciclase	Membrana	Receptor ligado a proteína G	ATP	cAMP
Guanilil ciclase	Membrana	Receptor enzimático	GTP	cGMP
	Citosol	Óxido nítrico (NO)		
Fosfolipase C	Membrana	Receptor ligado a proteína G	Fosfolipídeos de membrana	IP_3 e DAG

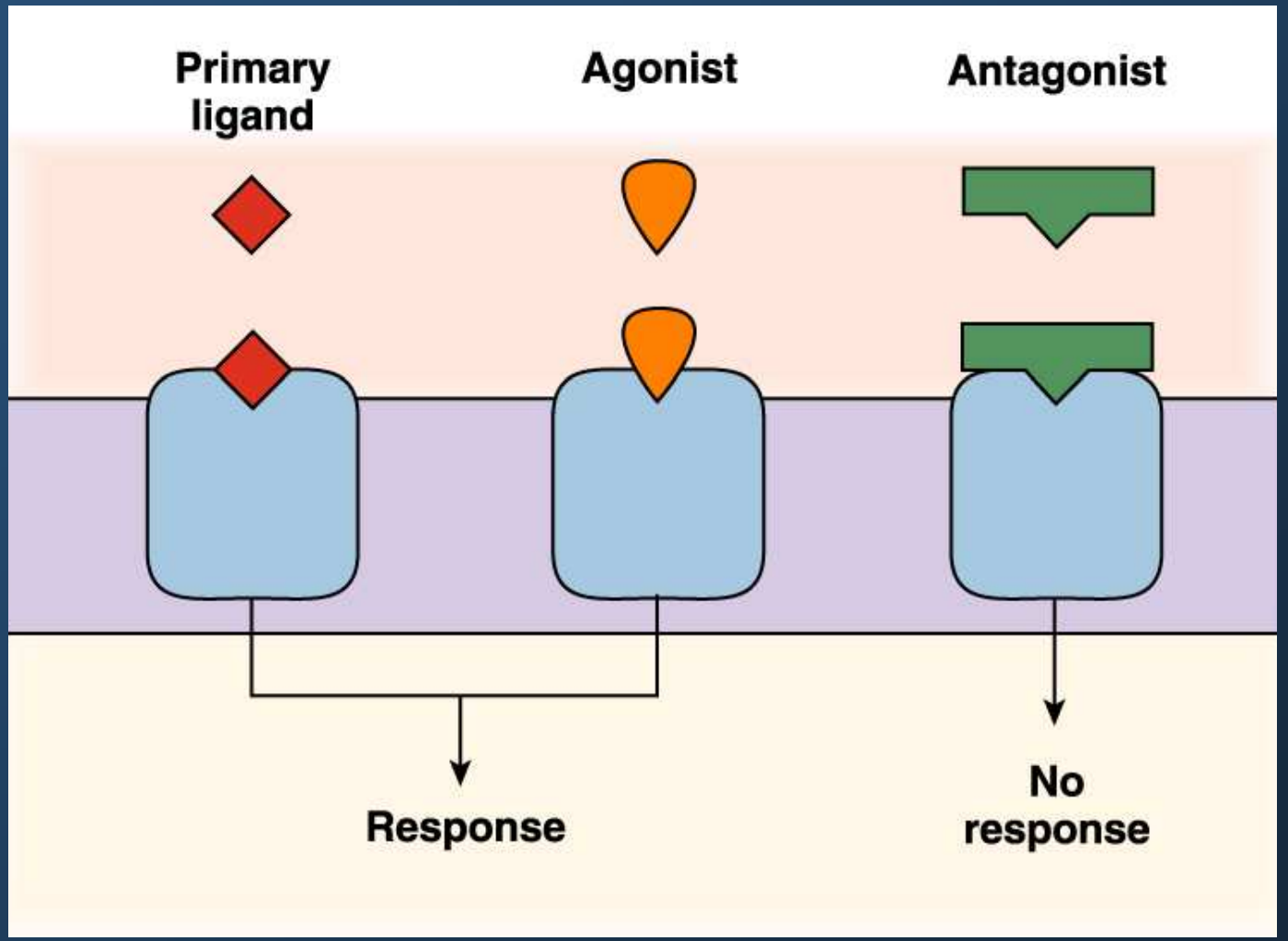
Segundo mensageiro

Tipo	Segundo mensageiro	Ação	Efeito
Íon	Ca^{2+}	Liga-se à calmodulina Liga-se a outras proteínas	Altera atividade enzimática Exocitose, contração muscular, movimento citoesquelético
Nucleotídeos	cAMP	Ativa proteínas quinases (PKA) Liga-se a canais iônicos	Fosforila proteínas Altera abertura de canais
	cGMP	Ativa proteínas quinases (PKC) Liga-se a canais iônicos	Fosforila proteínas Altera abertura de canais
Derivados de lipídeos	IP_3	Libera Ca^{2+} de reservas intracelulares	Efeitos do Ca^{2+}
	Diacilglicerol	Ativa PKC	Fosforila proteínas

Regulação das vias de sinalização



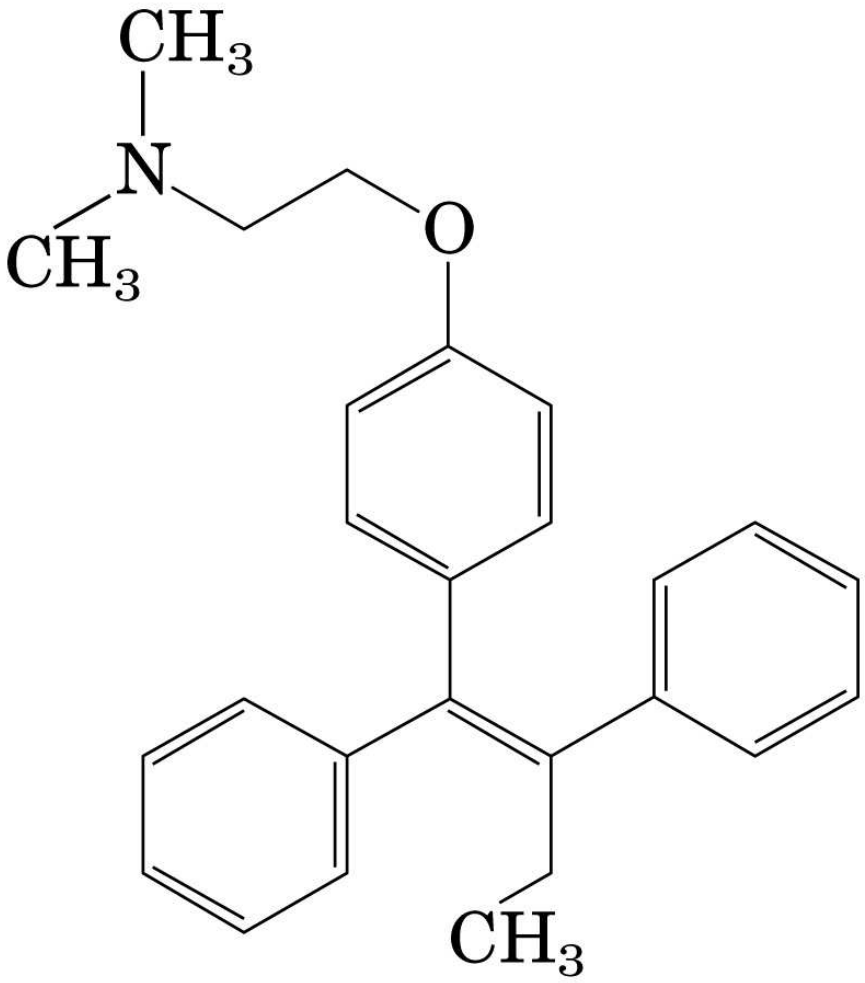
Agonistas x antagonistas



Agonistas x antagonistas

- Nicotina: agonista do receptor da acetilcolina
- Estrogênios dos anticoncepcionais: agonistas do estrógeno, com maior estabilidade
- Tamoxifeno: antagonista do receptor do estrógeno

Tratamento do câncer de mama - antagonista do receptor do estrógeno



Tamoxifeno

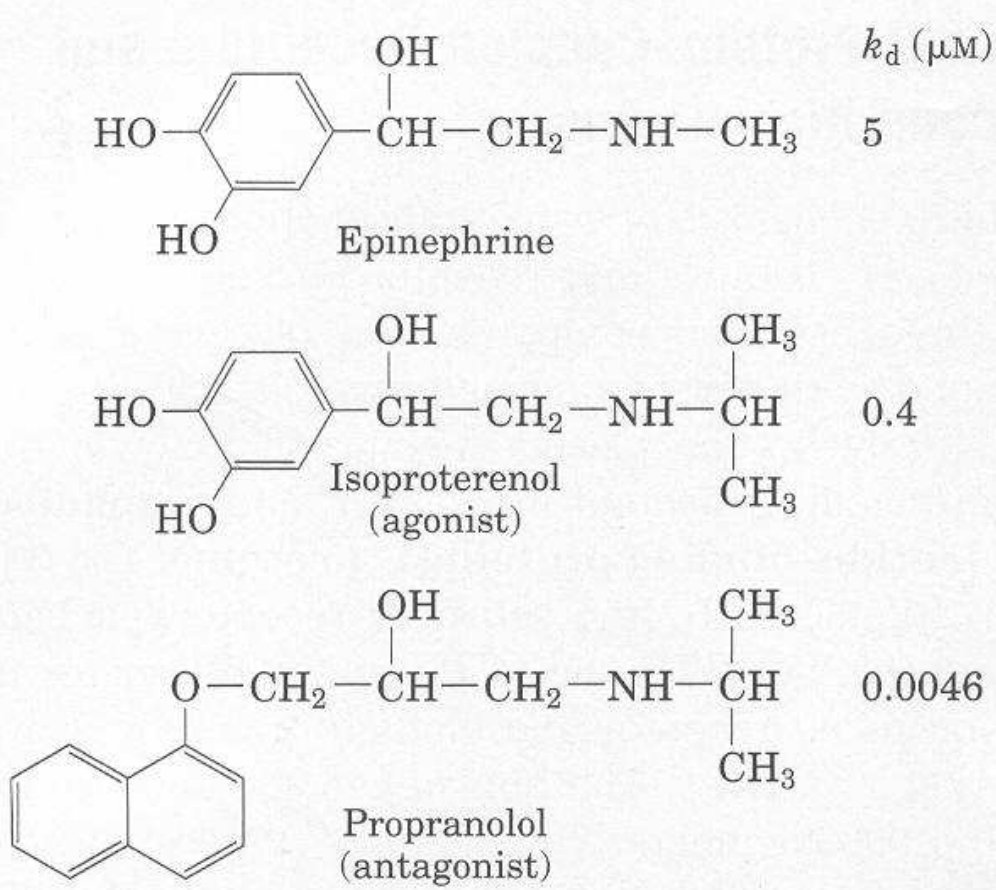
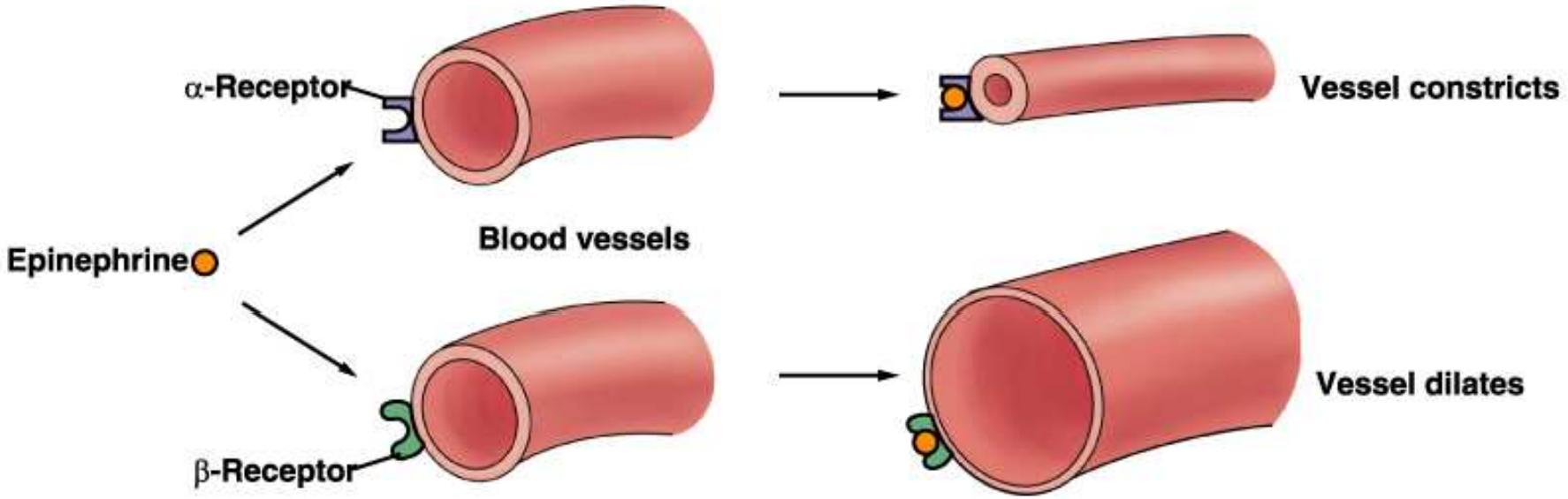


FIGURE 12–11 Epinephrine and its synthetic analogs. Epinephrine, also called adrenaline, is released from the adrenal gland and regulates energy-yielding metabolism in muscle, liver, and adipose tissue. It also serves as a neurotransmitter in adrenergic neurons. Its affinity for its receptor is expressed as a dissociation constant for the receptor-ligand complex. Isoproterenol and propranolol are synthetic analogs, one an agonist with an affinity for the receptor that is higher than that of epinephrine, and the other an antagonist with extremely high affinity.


α-Receptor
Vessel constricts
Epinephrine
Blood vessels
Vessel dilates
β-Receptor

Regulação positiva e negativa

- Saturação pode acontecer com enzimas, transportadores e receptores
- Regulação negativa:
  - Decréscimo no número de receptores
  - Decréscimo na afinidade de associação
- Regulação positiva:
  - Produção de novos receptores

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