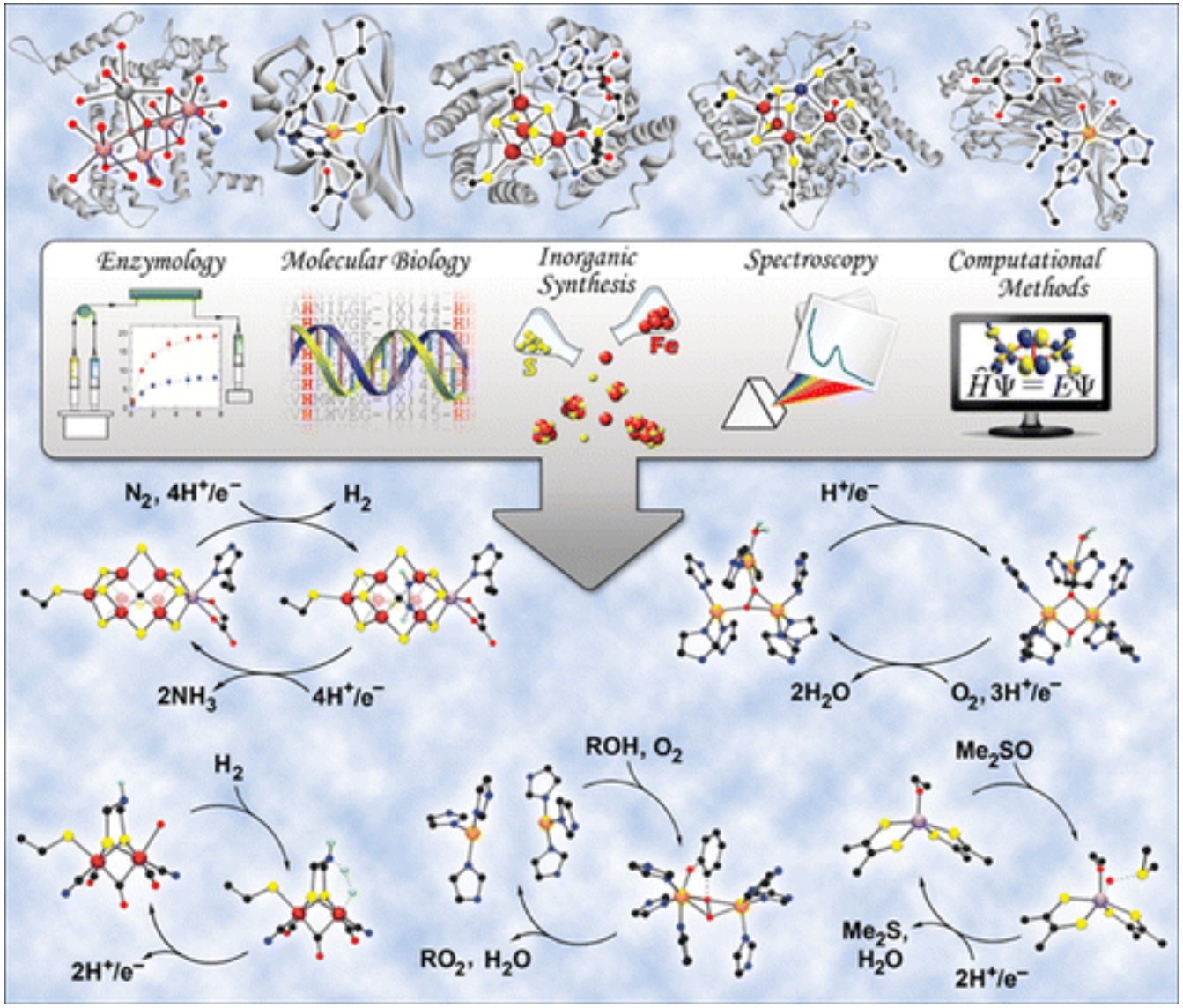
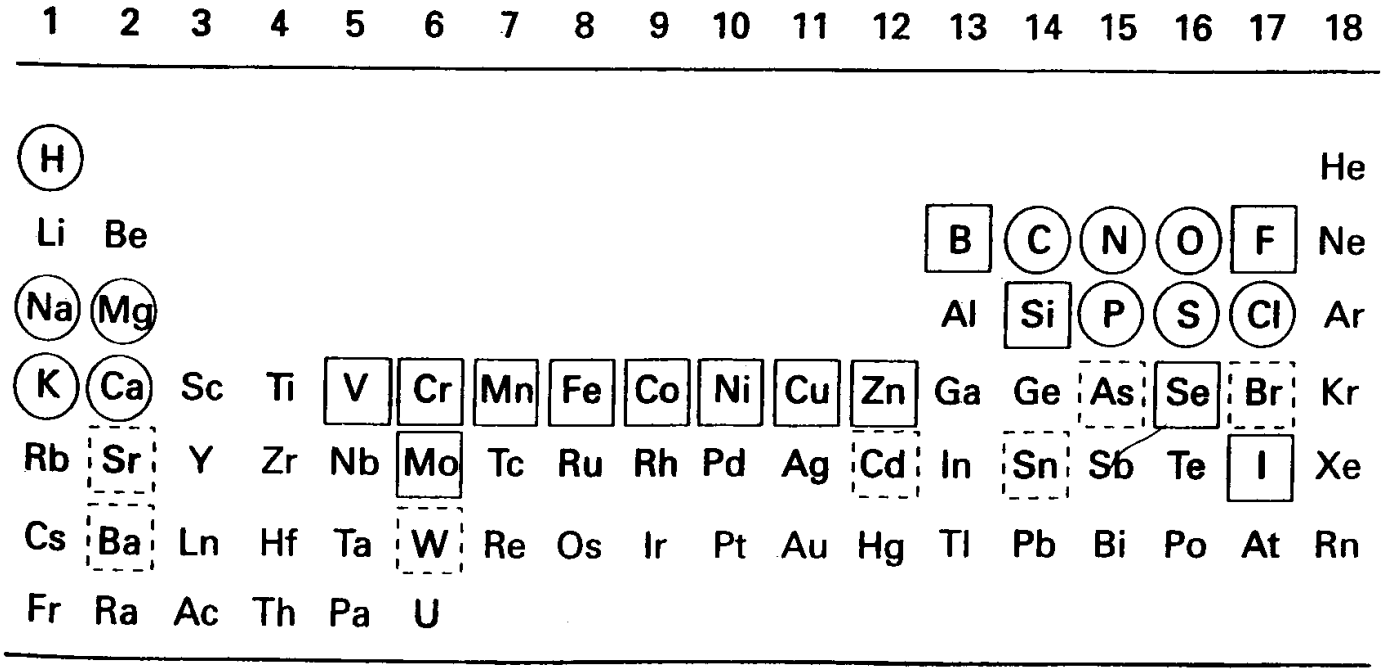

Enzymology
Molecular Biology
Inorganic Synthesis
Spectroscopy
Computational Methods
Fe
S
$\hat{H}\Psi = E\Psi$
N_2, $4H^+/e^-$
H_2
$2NH_3$
$4H^+/e^-$
H^+/e^-
$2H_2O$
O_2, $3H^+/e^-$
H_2
$2H^+/e^-$
ROH, O_2
RO_2, H_2O
Me_2SO
Me_2S, H_2O
$2H^+/e^-$



C + H + O + N = 99% del totale degli atomi

Na (100g), **K** (170g), **Mg** (25g), **Ca** (1100g) = *bulk metals*
Fe, **Cu**, **Zn**, **V**, **Cr**, **Mo**, **Mn**, **Co**, **Ni** = *trace e ultra-trace metals*

13 metalli essenziali

Metal	Mass / mg	Biological roles
V	0.11	Enzymes (nitrogenases, haloperoxidases)
Cr	14	Claimed (not yet proven) to be essential in glucose metabolism in higher mammals
Mn	12	Enzymes (phosphatase, mitochondrial superoxide dismutase, glycosyl transferase); photoredox activity in Photosystem II (see **equation 22.54** and discussion)
Fe	4200	Electron-transfer systems (Fe–S proteins, cytochromes); O_2 storage and transport (haemoglobin, myoglobin, haemerythrin); Fe storage (ferritin, transferritin); Fe transport proteins (siderophores); in enzymes (e.g. nitrogenases, hydrogenases, oxidases, reductases)
Co	3	Vitamin B_{12} coenzyme
Ni	15	Enzymes (urease, some hydrogenases)
Cu	72	Electron transfer systems (blue copper proteins); O_2 storage and transport (haemocyanin); Cu transport proteins (ceruloplasmin)
Zn	2300	Acts as a Lewis acid (e.g. in hydrolysis processes involving carboxypeptidase, carbonic anhydrase, alcohol dehydrogenase); structural roles
Mo	5	Enzymes (nitrogenases, reductases, hydroxylases)

Concentrazione intracellulare media dei *trace elements* nelle cellule di eucarioti:

$[Fe]_{totale}$ = 0.5 mM

$[Zn]_{totale}$ = 0.5 mM

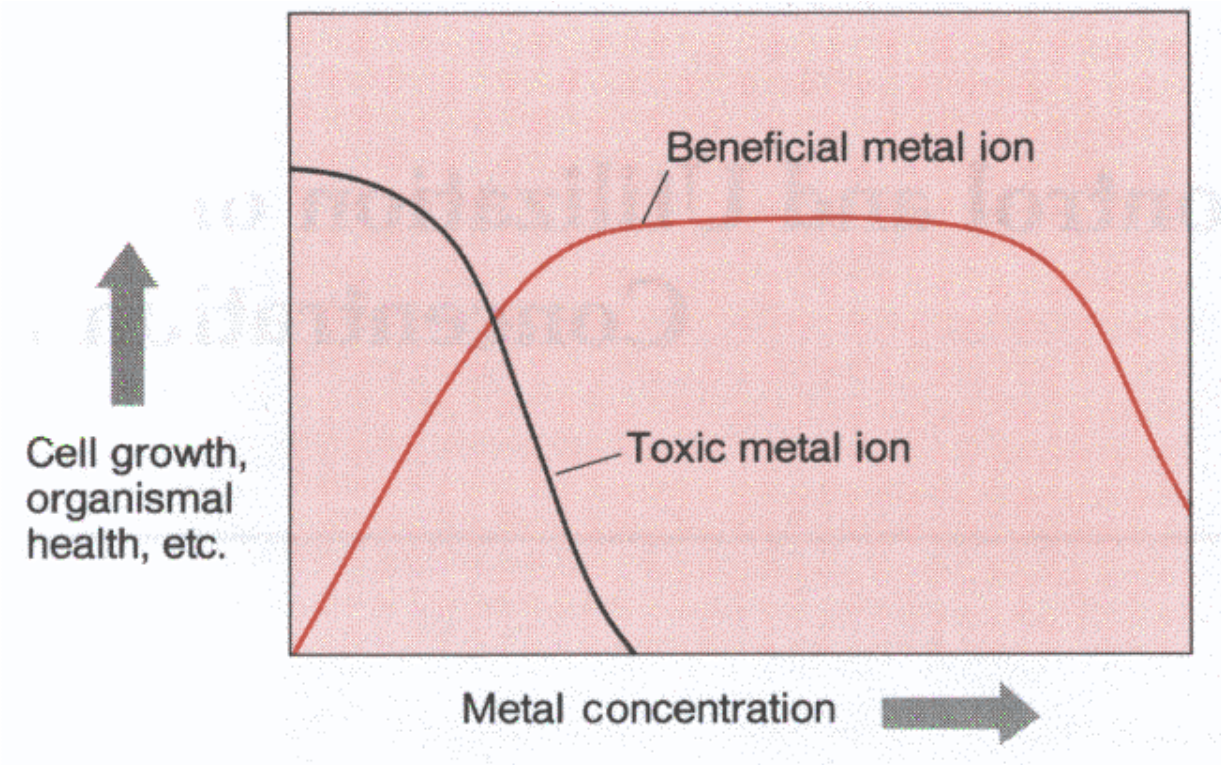
$[Cu]_{totale}$ = 50 μM

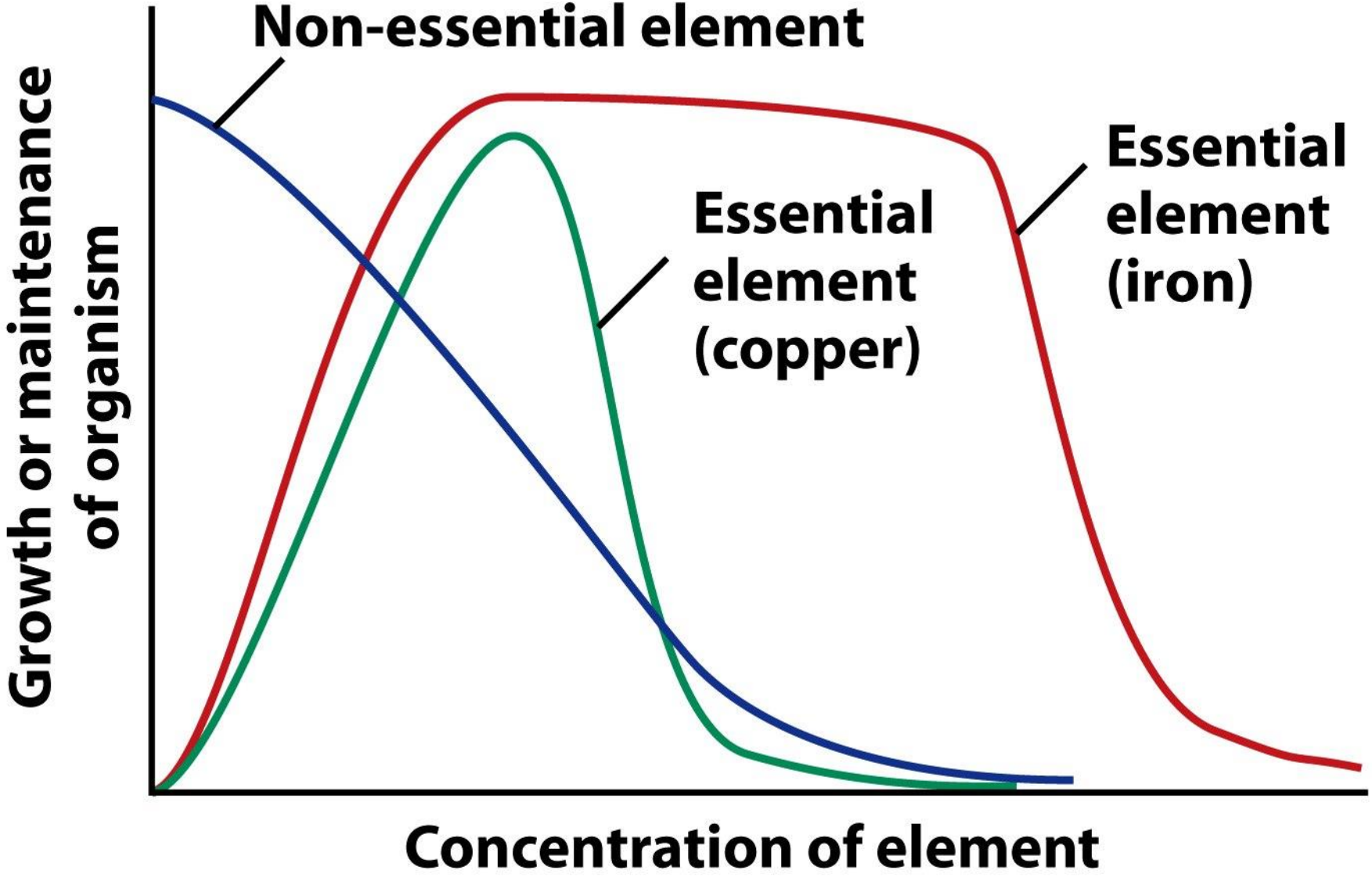
Metalloma

Ogni specie è caratterizzata da uno specifico
metalloma
definito come l'insieme di metalli contenuto in ogni tipo di cellula di quella specie, ognuno con la sua specifica **quantità**, **speciazione** e **localizzazione** all'interno di ogni cellula

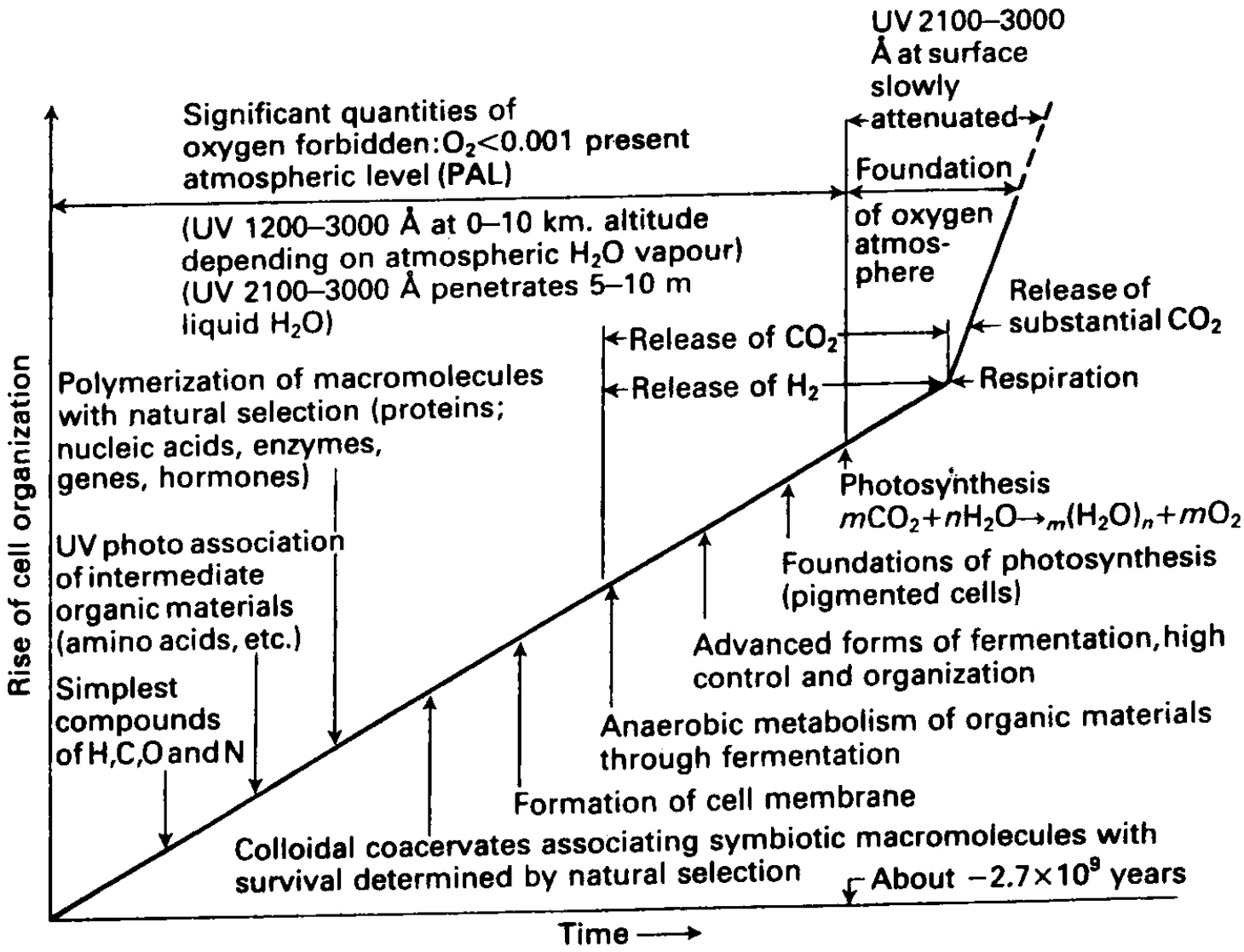
Come si stabilisce se un elemento è essenziale per una specie?

*Si definisce **essenziale** un elemento sistematicamente presente in una certa specie biologica e tale che la sua assenza (o carenza) nelle fonti nutritive di quella specie sia causa di malattie, disturbi metabolici o dello sviluppo*




Non-essential element
Essential element (copper)
Essential element (iron)
Growth or maintenance of organism
Concentration of element

Biodisponibilità degli elementi



Potenziali redox accessibili in acqua a pH 7: fra –0.4 V (H^+/H_2) e +0.8 V (O_2/OH^-)

Elemento	**Ambiente riducente**	**Ambiente ossidante**
Fe	Fe(II), (alta)	Fe(III), (bassa)
Cu	Come solfuro (bassa)	Cu(II), (moderata)
S	HS^- (alta)	SO_4^{2-} (alta)
Mo	MoS_2, $(MoO_nS_{4-n})^{2-}$ (bassa)	MoO_4^{2-} (moderata)
V	V^{3+}, solfuri di V(IV) (moderata)	VO_4^{3-} (moderata)

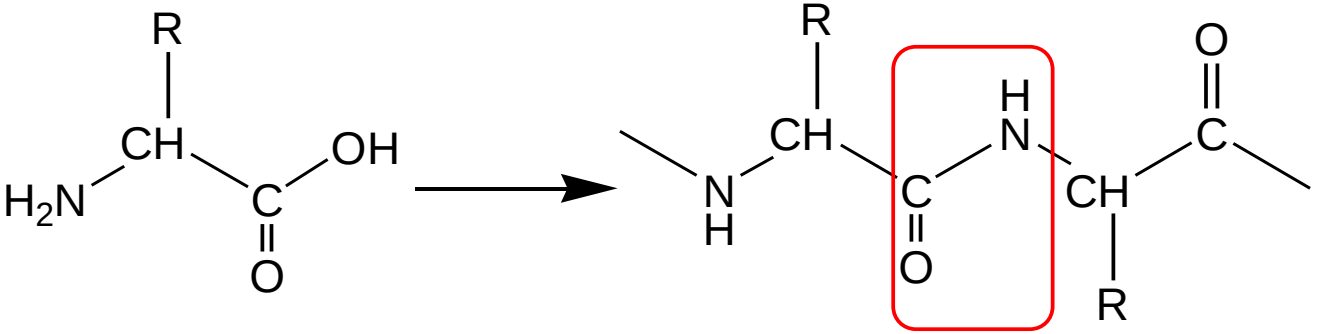
Ruoli dei metalli nei sistemi biologici

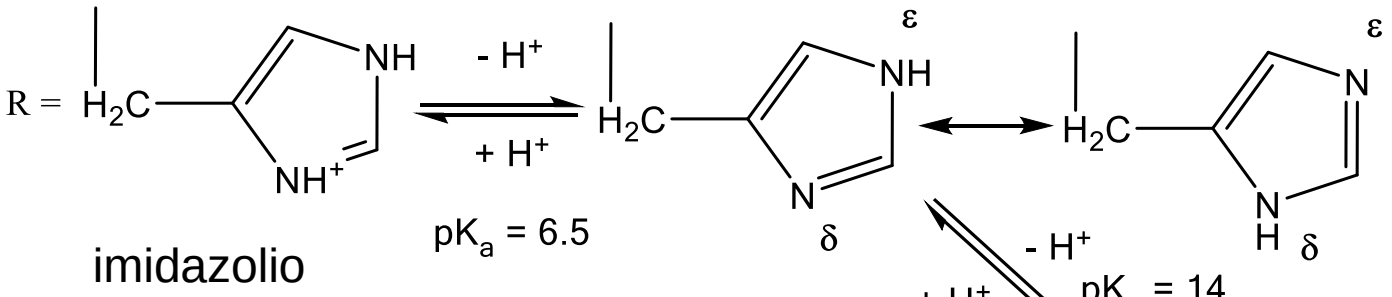
Ruolo strutturale

Endo- ed esoscheletri, stabilizzazione di DNA, RNA e proteine

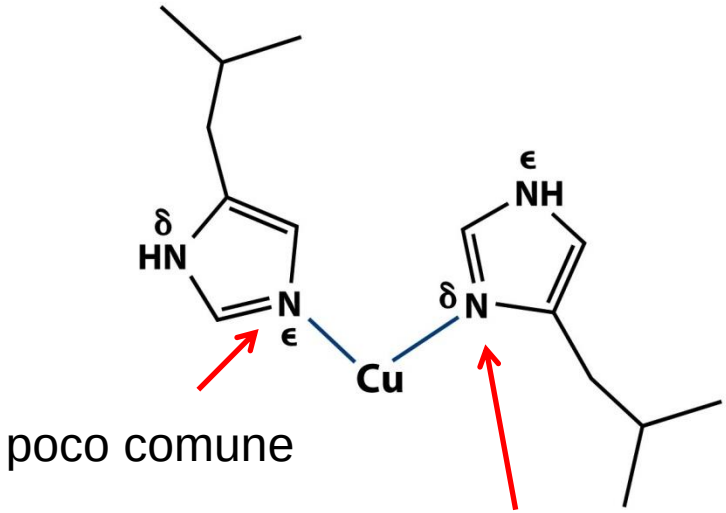
Ruolo funzionale

- Trasporto di carica
- Sintesi e metabolismo di molecole organiche
- Trasferimento di elettroni
- Attivazione di piccole molecole
- Reattività organometallica





imidazolato

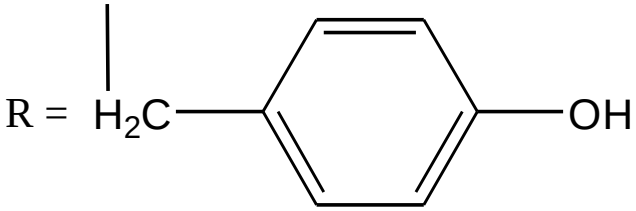


poco comune

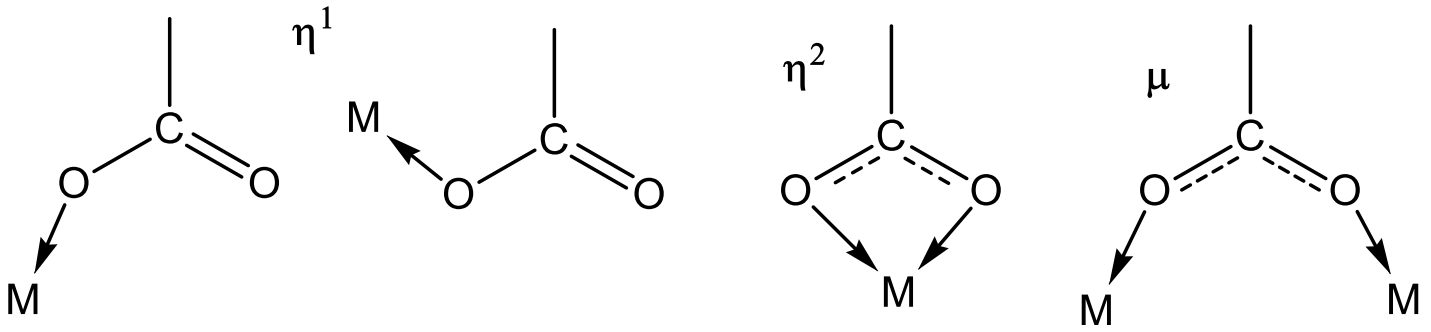
molto comune

R = CH_2SH
Cisteina (Cys), pK_a = 8.5

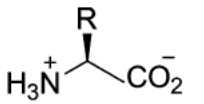
R = $–CH_2CH_2SCH_3$
Metionina (Met)



Tirosina, pK_a = 10



Glutammato (Glu): R = $–CH_2CH_2COO^–$ Aspartato (Asp): R = $–CH_2COO^–$
pK_a = 4.5



general amino acid structure

Amino acids with non-polar side chains:

Gly R = —H

Ala R = —

Val R =

Leu R =

Ile R =

Met R =

Phe R =

Trp R =

Pro

Amino acids with polar side chains:

Cys R =

Ser R =

Thr R =

His R =

Asn R =

Gln R =

Tyr R =

Amino acids with charged polar side chains:

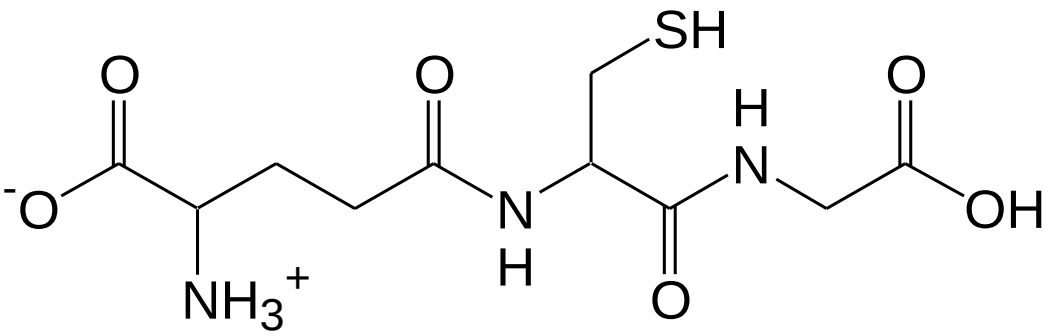
Asp R =

Glu R =

Arg R =

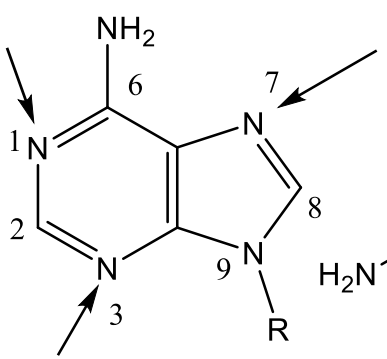
Lys R =

GLUTATIONE

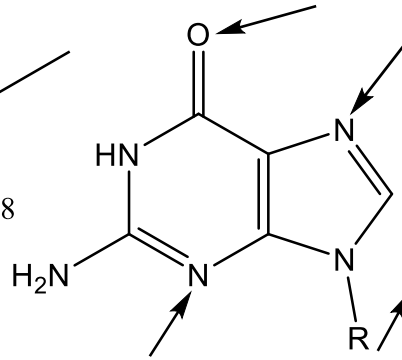


GSH

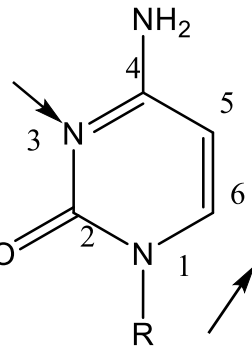
Glu-Cys-Gly
0.5 – 10 mM intracellulare
(riducente monoelettronico)



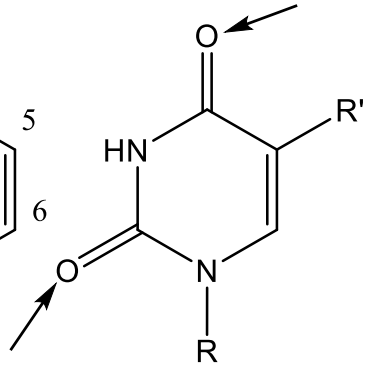
Adenina



Guanina



Citosina



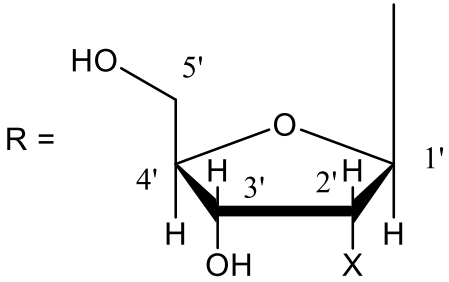
R' = H = Uracile

R' = CH_3 = Timina

Basi puriniche

Basi pirimidiniche

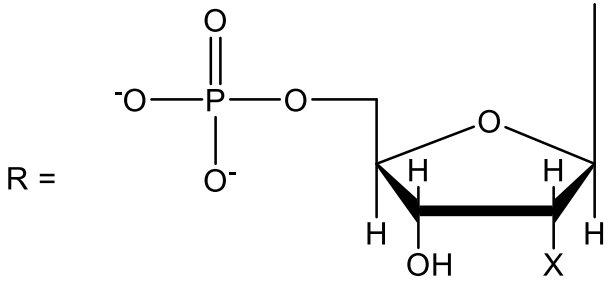
R = H **Nucleobase**



Nucleoside

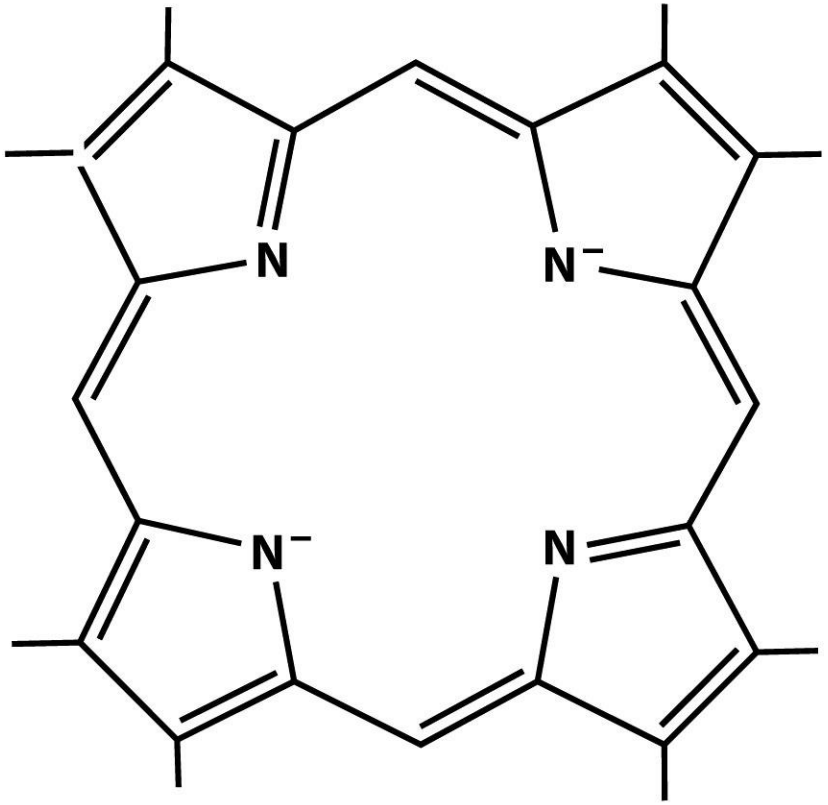
X = OH ribosio

X = H desossi-ribosio

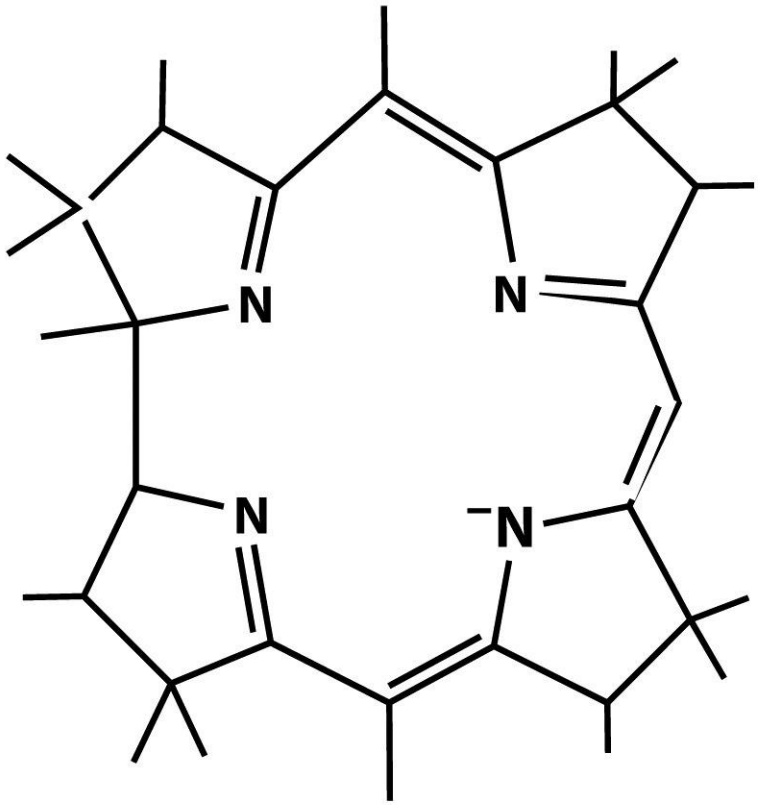


Nucleotide

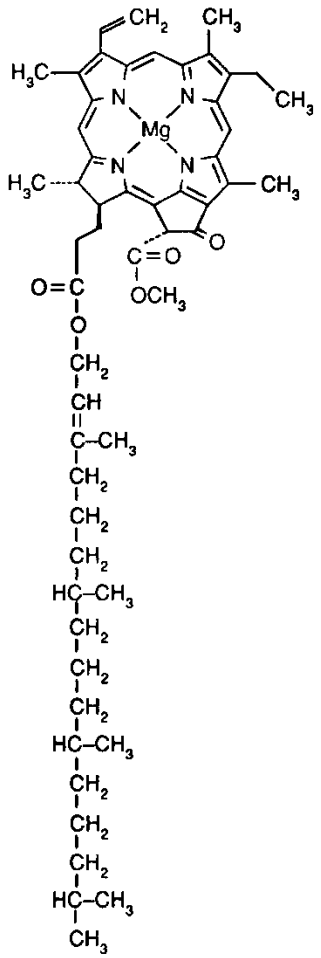
Leganti tetrapirrolici



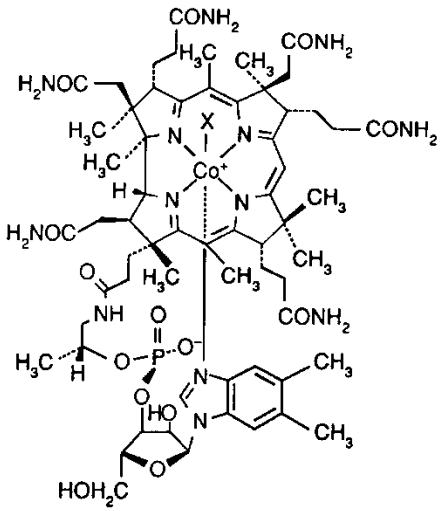
Porphyrin^{2-}



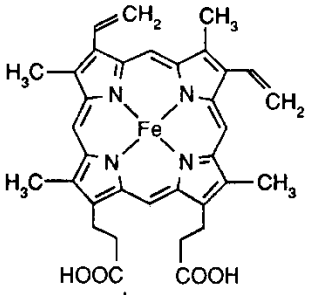
Corrin^{-}



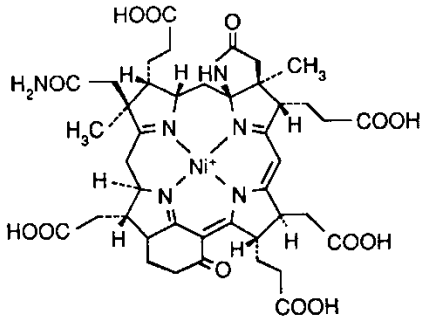
chlorophyll *a*



vitamin B_{12} (X = CN)



heme
(Fe-protoporphyrin IX)

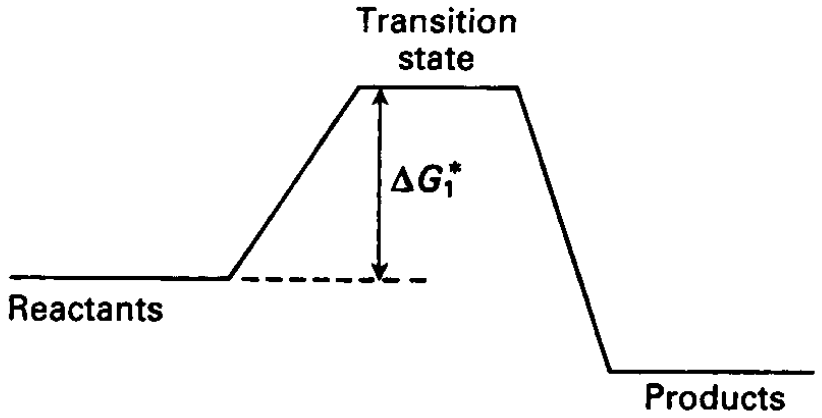


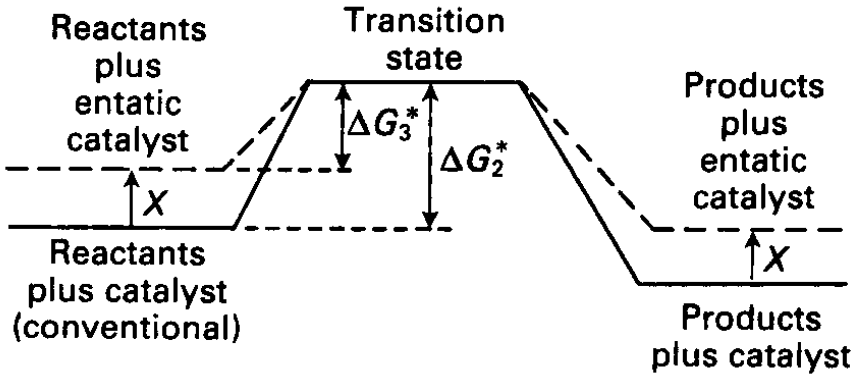
coenzyme F430

Table 2.6 Typical coordination environments of metal centers in proteins

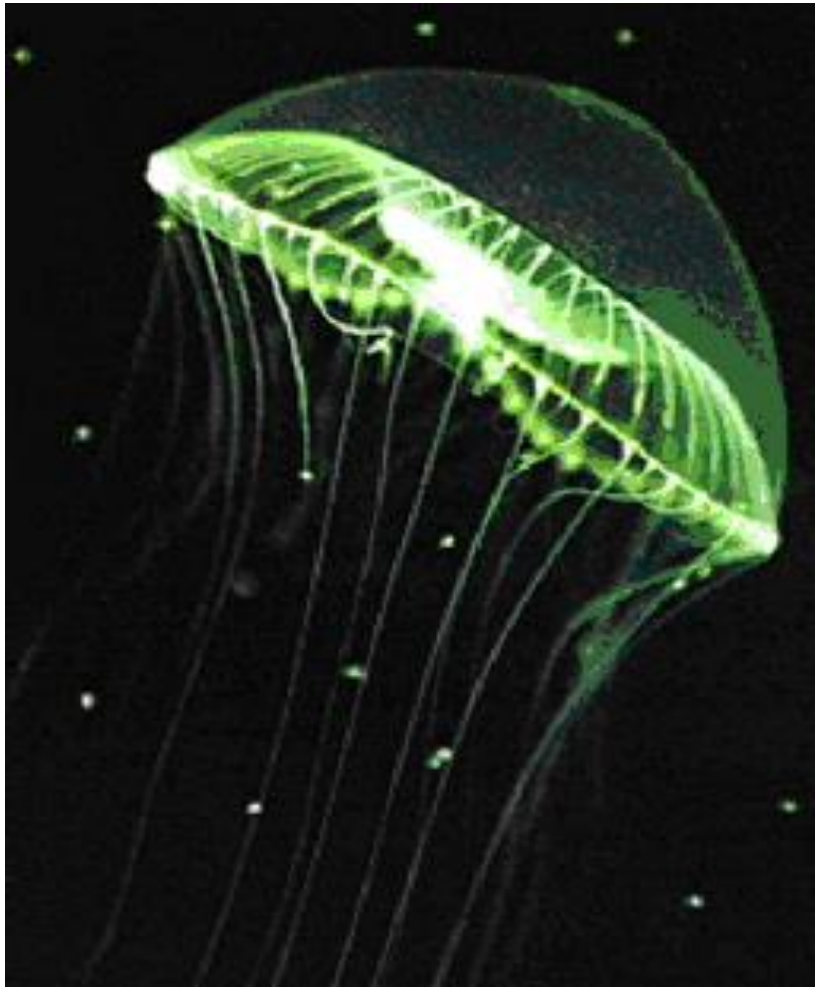
metal oxidation state	bond stability	typical number and type of side chain ligands	typical coordination geometry
Zn(II)	high	3: His, Cys^-, (Glu^-)	severely distorted tetrahedron
Cu(I)	high	3,4: His, Cys^-, Met	severely distorted tetrahedron
Cu(II)	high	3,4: His, (Cys^-)	distorted square planar arrangemer
Fe(II), Ni(II) Co(II), Mg(II)	low	4-6: His, Glu^-, Asp^-	distorted octahedron
Fe(III)	high	4-6: Glu^-, Asp^-, Tyr^-, Cys^-	distorted octahedron

Stato entatico

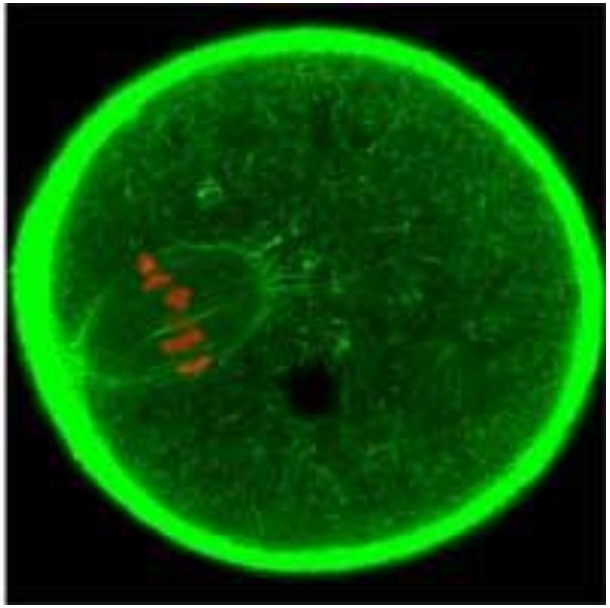




Ione	Raggio (Å)	Ione	Raggio (Å)
Na^+	1.02	Mg^{2+}	0.72
K^+	1.38	Ca^{2+}	1.00

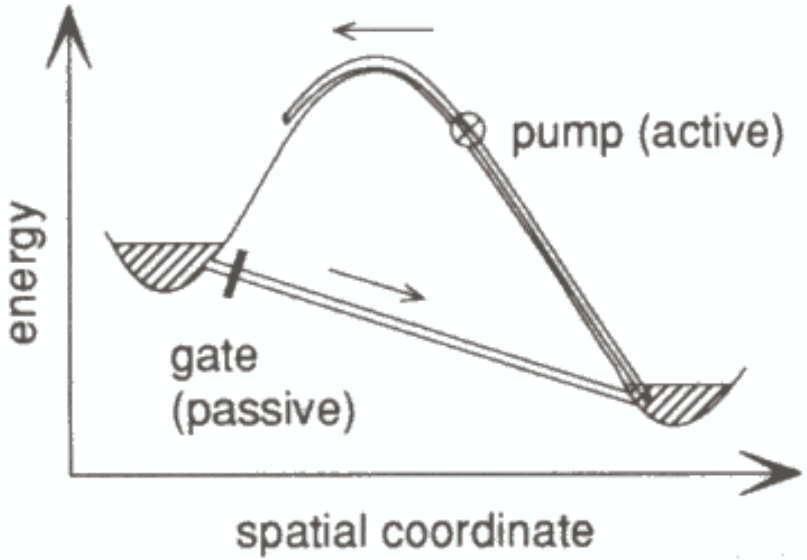


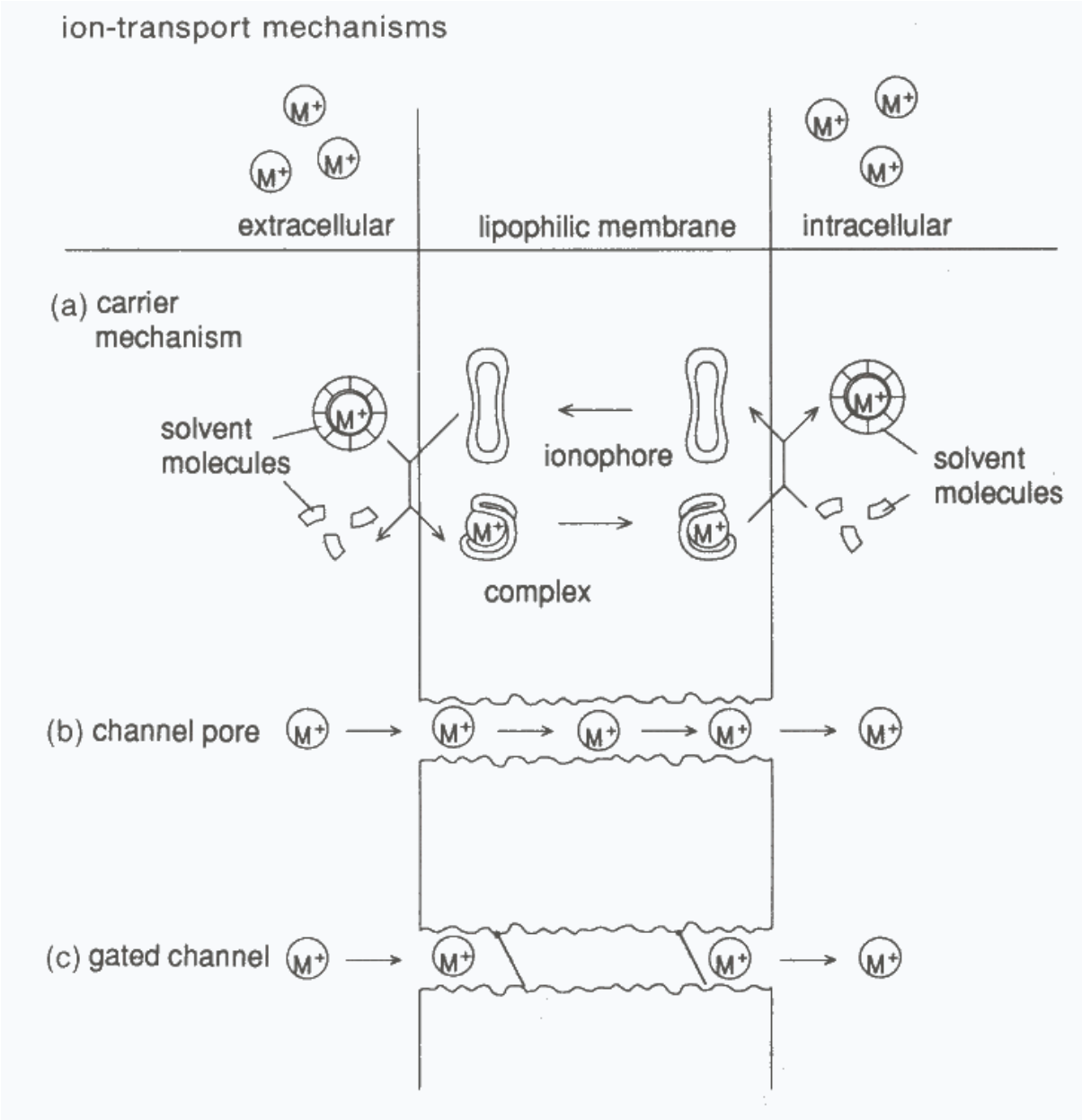
Green Fluoerescent Protein (GFP) = sensore fluorescente per Ca^{2+}



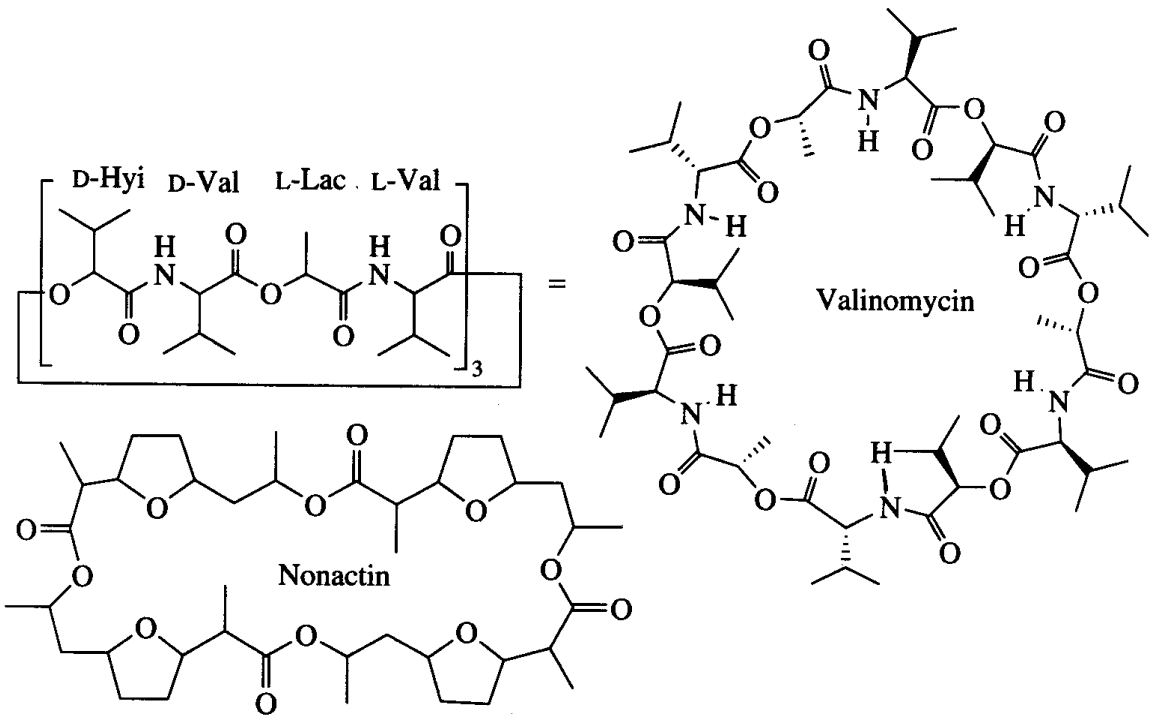
Ione	Intracellulare (mM)	Extracellulare (mM)
Na^+	10	150
K^+	100	5
Mg^{2+}	2.5	1.5
Ca^{2+}	0.1[a]	2.5
Cl^-	4	100

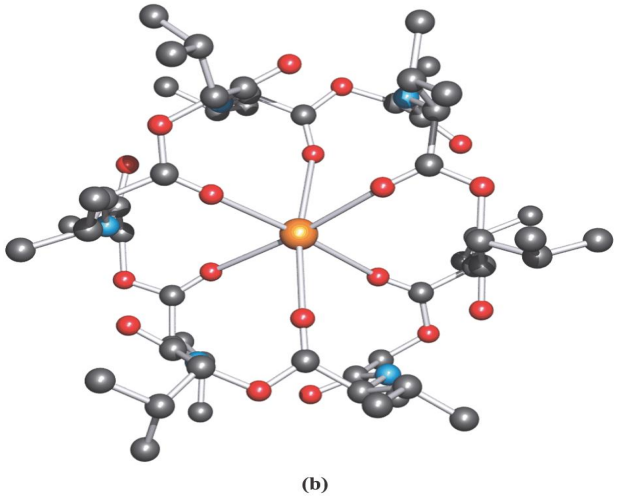
[a]0.1 μM nel citoplasma della cellula a riposo



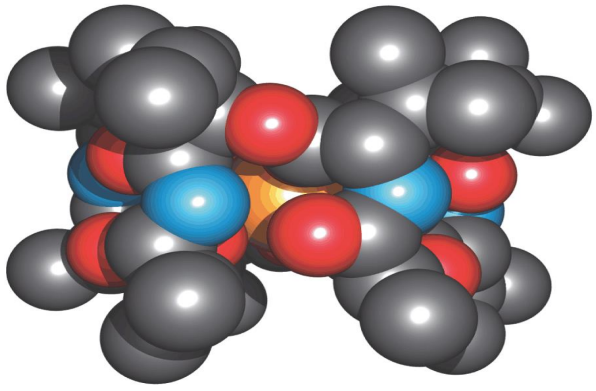

ion-transport mechanisms
M+
extracellular
lipophilic membrane
intracellular
(a) carrier mechanism
solvent molecules
ionophore
solvent molecules
complex
(b) channel pore
(c) gated channel

Ionofori

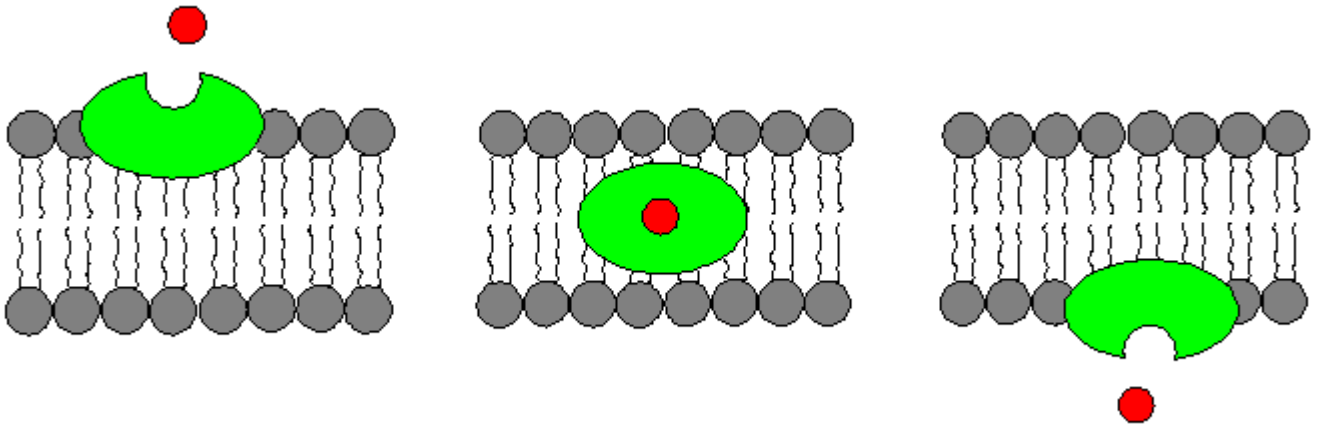




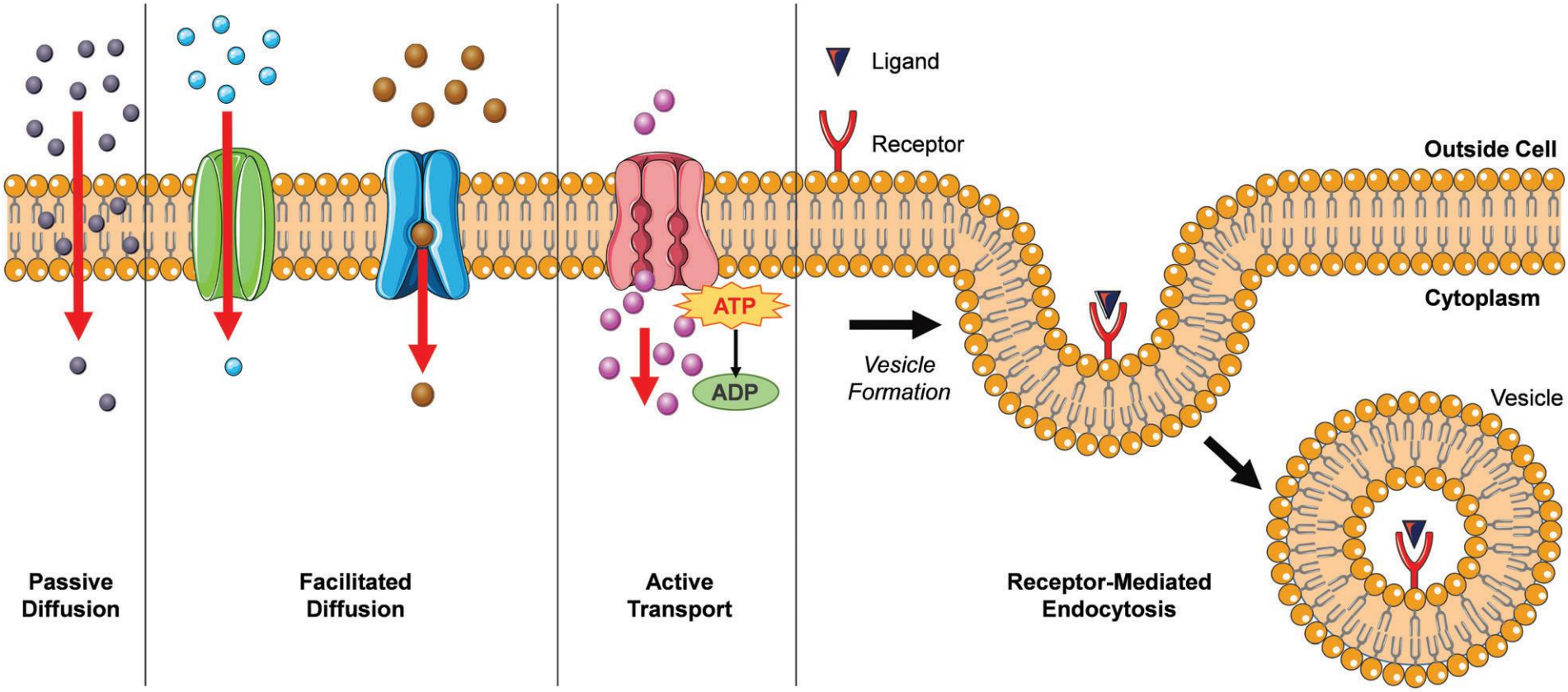
Addotto valinomicina – K^+



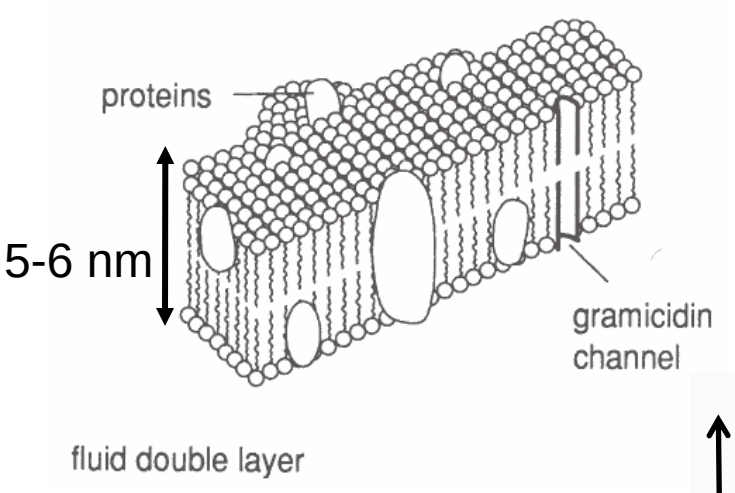
Ionofori

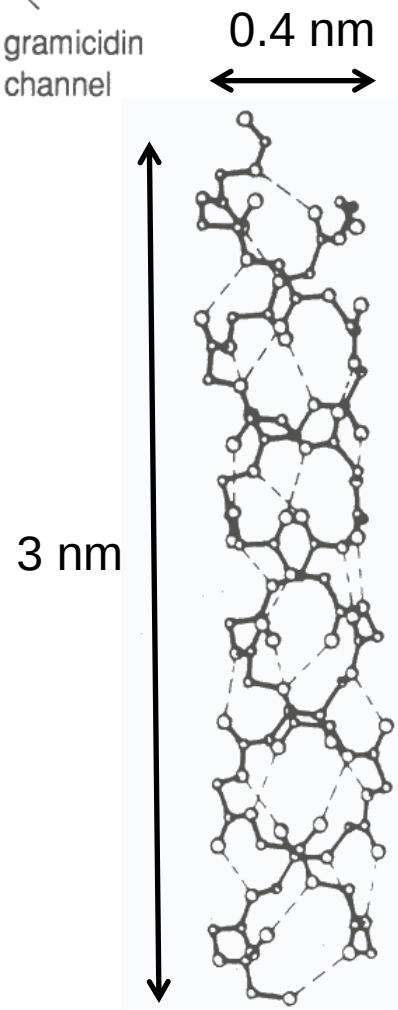


La valinomicina trasporta 10^3 – 10^4 ioni K^+ al secondo attraverso la membrana dei mitocondri.
La selettività K^+/Na^+ è dell'ordine di 10^4


Ligand
Receptor
Outside Cell
Cytoplasm
ATP
ADP
Vesicle Formation
Vesicle
Passive Diffusion
Facilitated Diffusion
Active Transport
Receptor-Mediated Endocytosis
Energy-Independent
Energy-Dependent

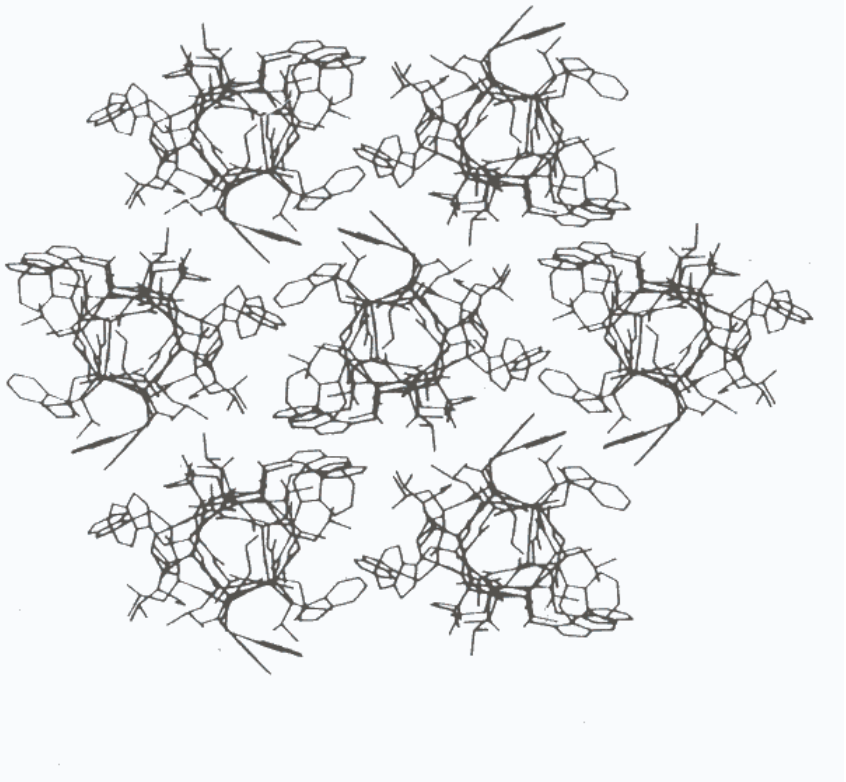
Ion channels



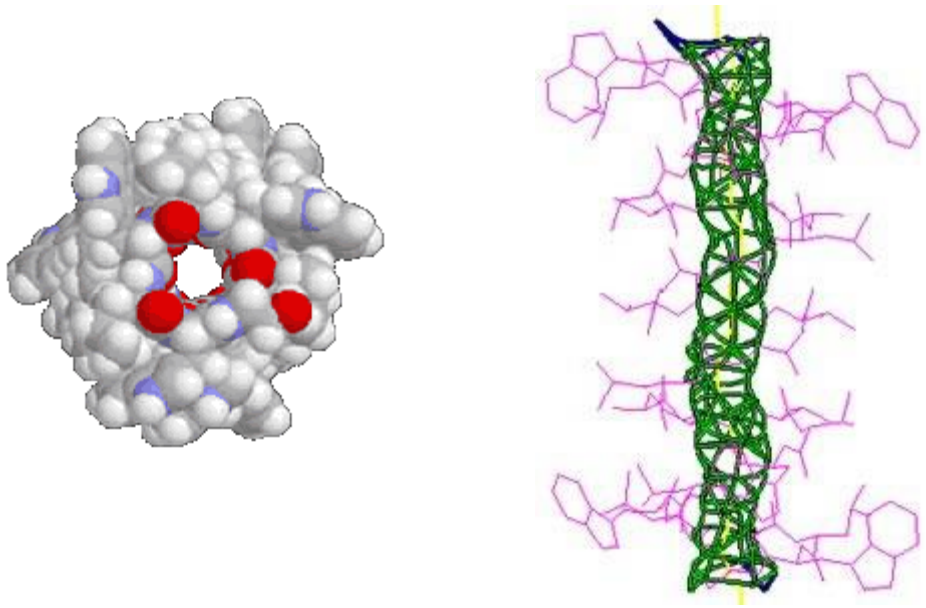


Gramicidina A

15 aminoacidi

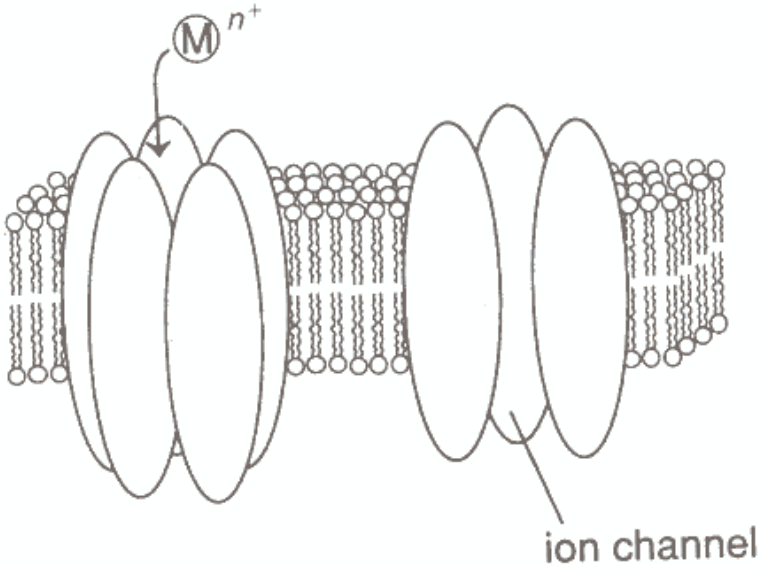


Gramicidina A

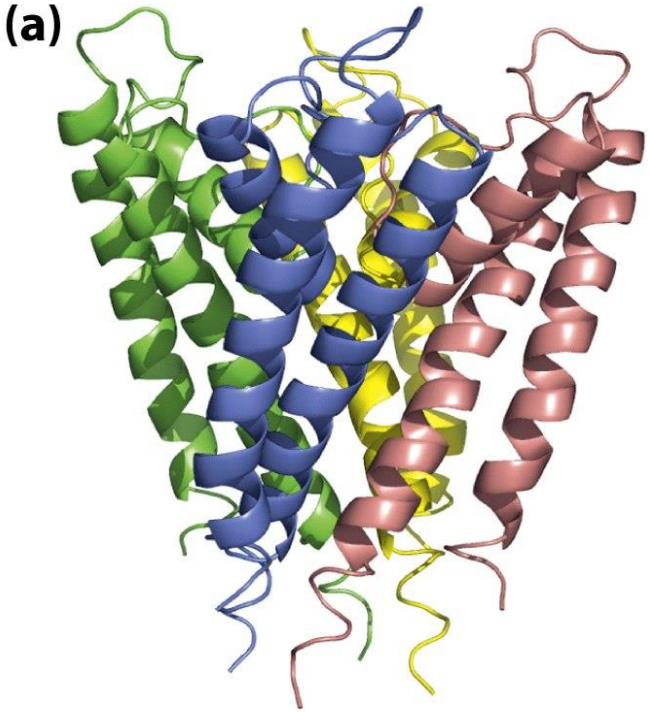


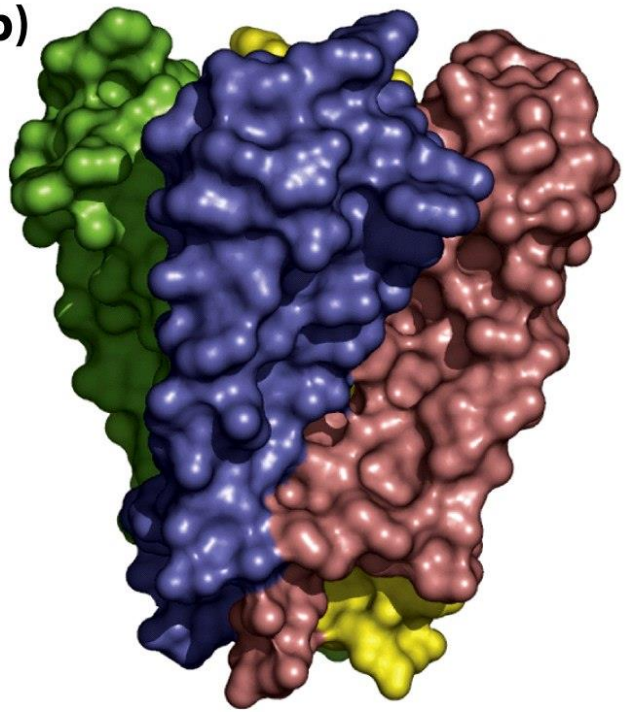
La gramicidina A trasporta 10^7 ioni K^+ o Na^+ al secondo attraverso la membrana. I cationi divalenti bloccano il poro.

Proteine trans-membrana



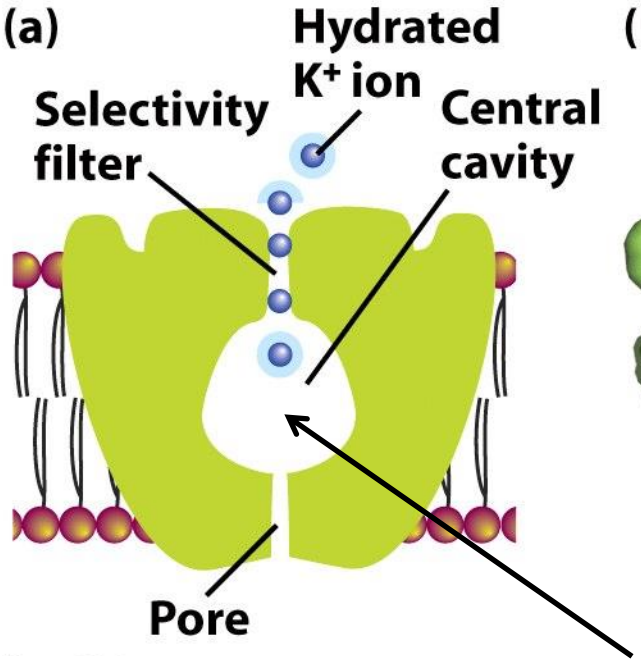
Canale del K^+



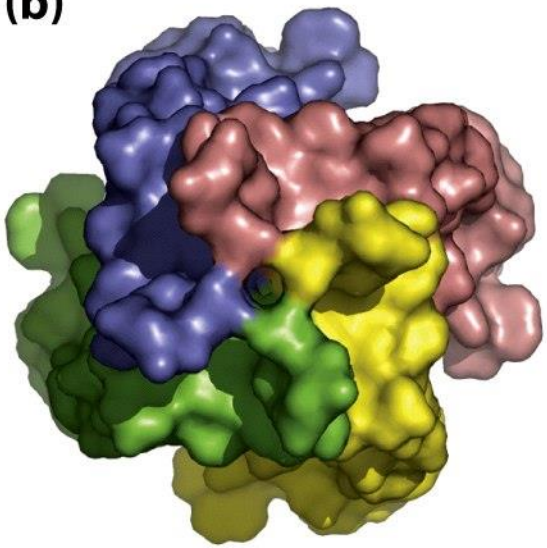


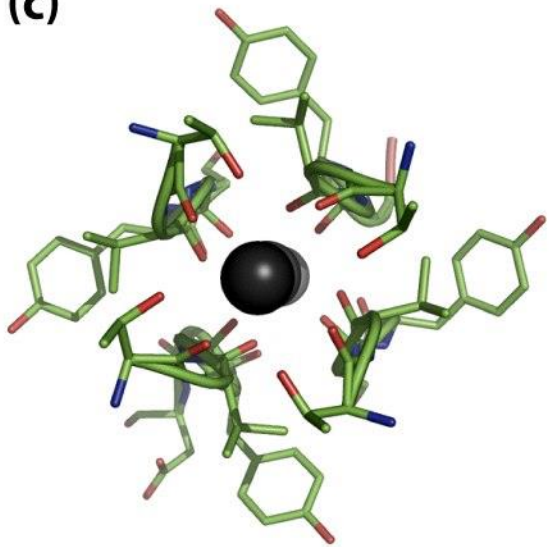
Canale del K^+ *potential gated*

Out

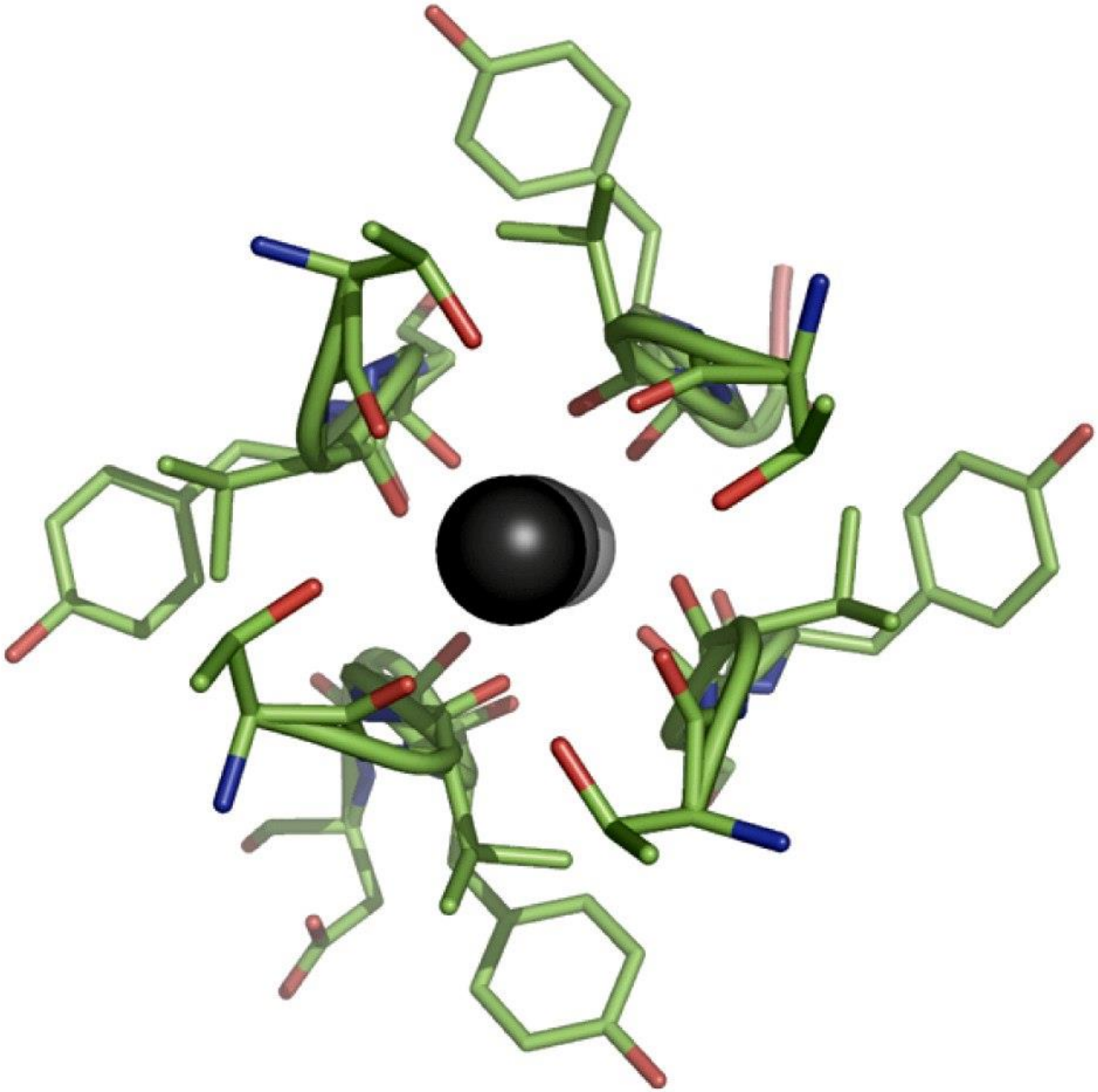


In

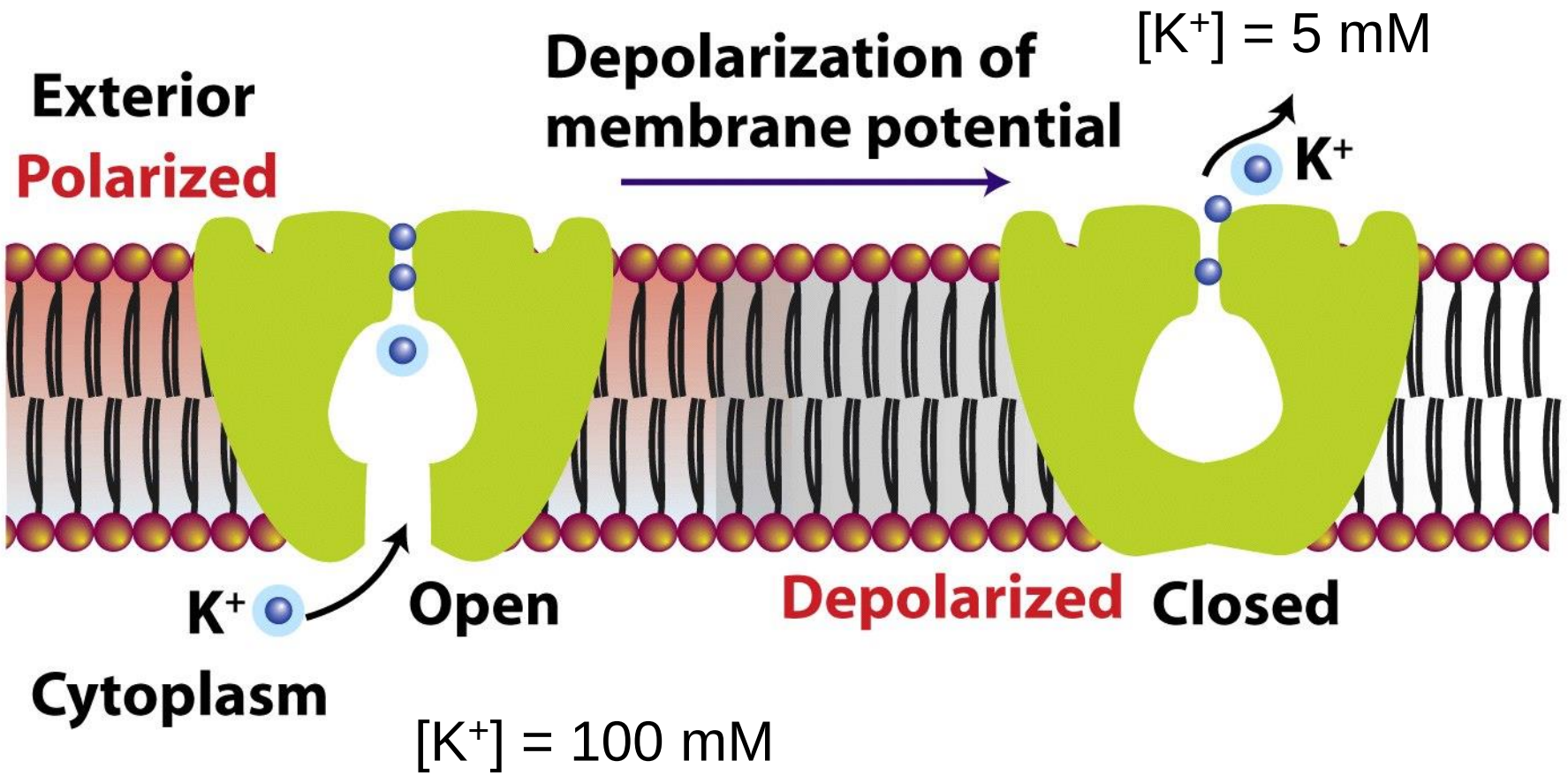




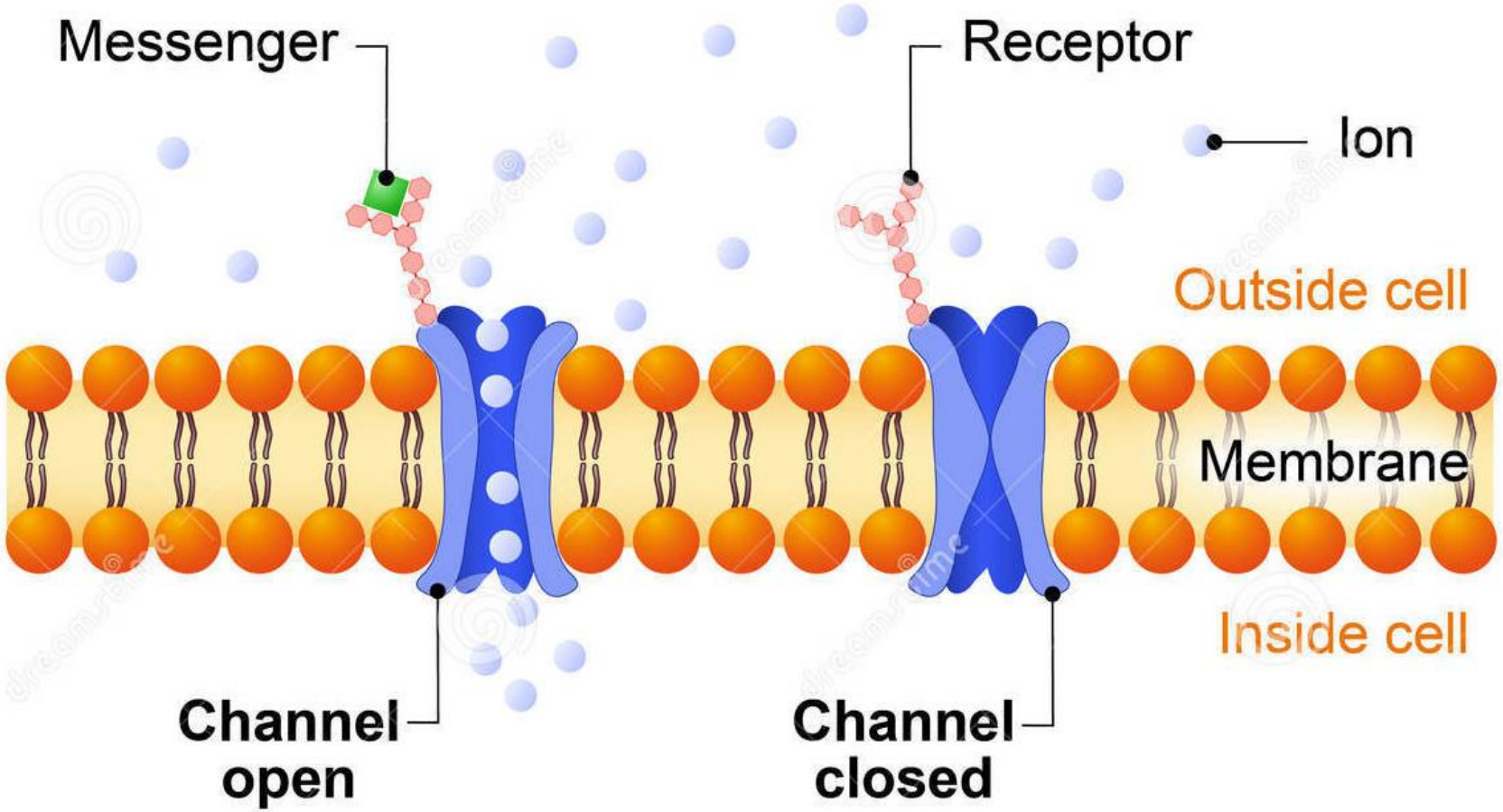
Ø = 1 nm
$[K^+]$ = 2 M



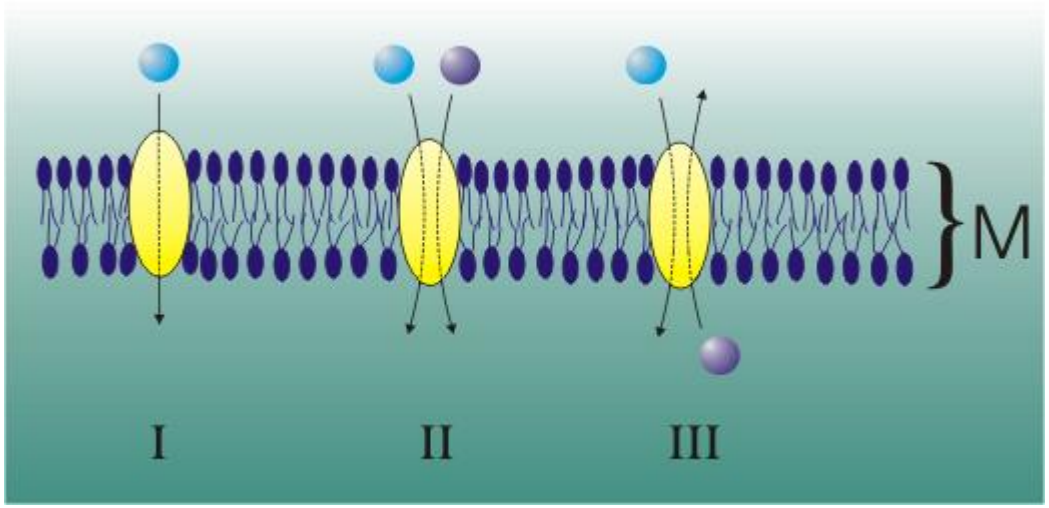
Canale del K^+ *potential gated*



Ligand-gated ion channel

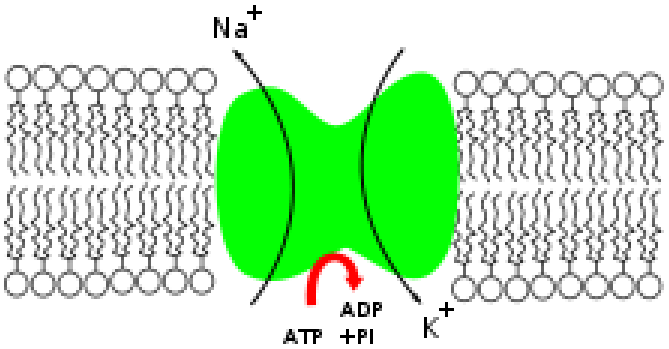


Pompe ioniche (ATPasi)



Uniporto *Simporto* *Antiporto*

Pompa ionica *antiporto* Na^+/K^+-ATPasi

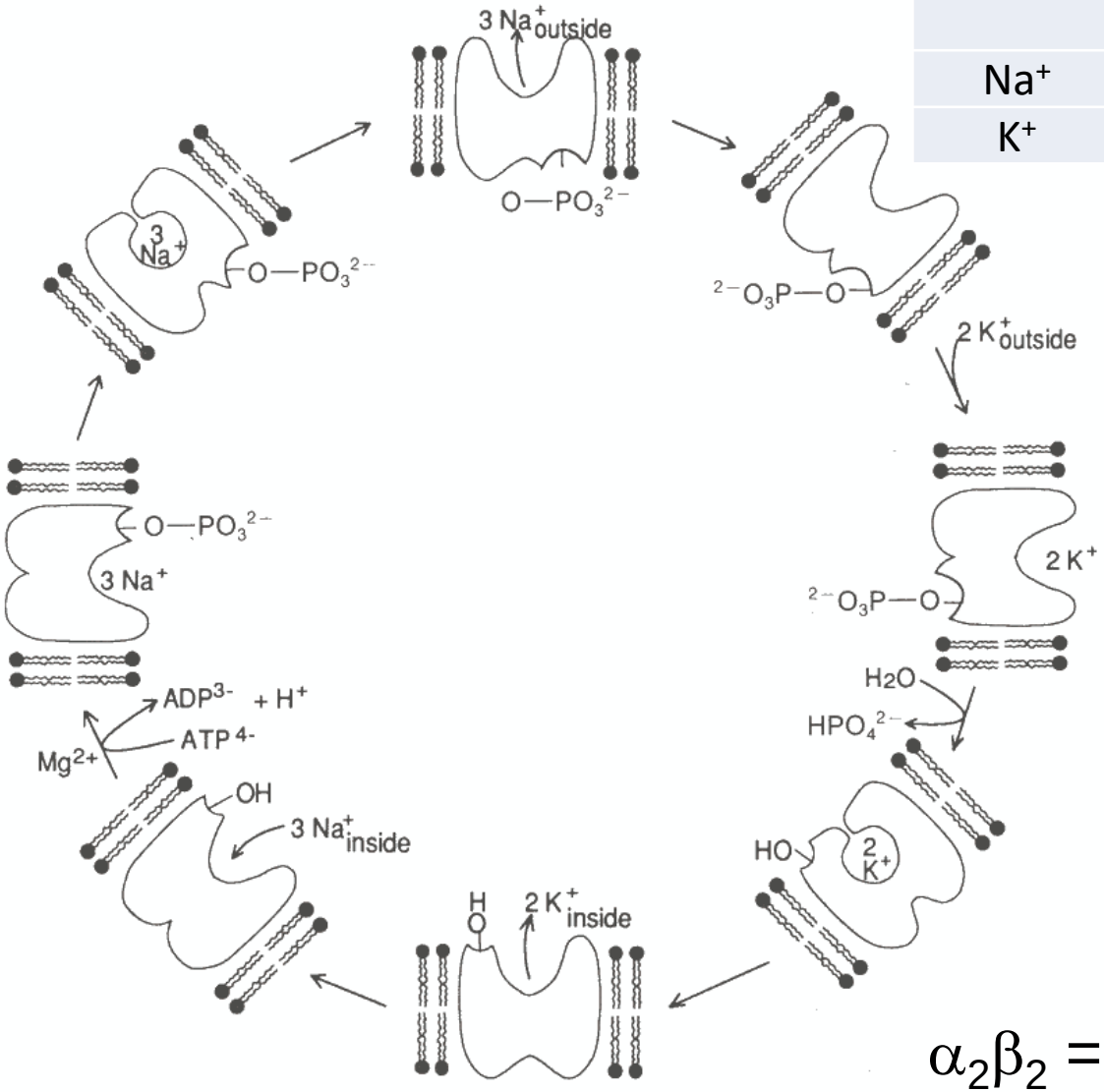


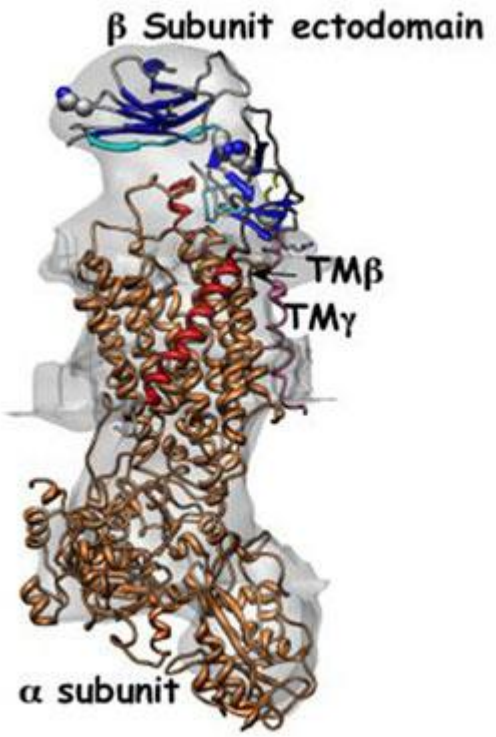
Ione	Intracellulare (mM)	Extracellulare (mM)
Na^+	10	150
K^+	100	5

Pompa ionica *antiporto* Na^+/K^+-ATPasi

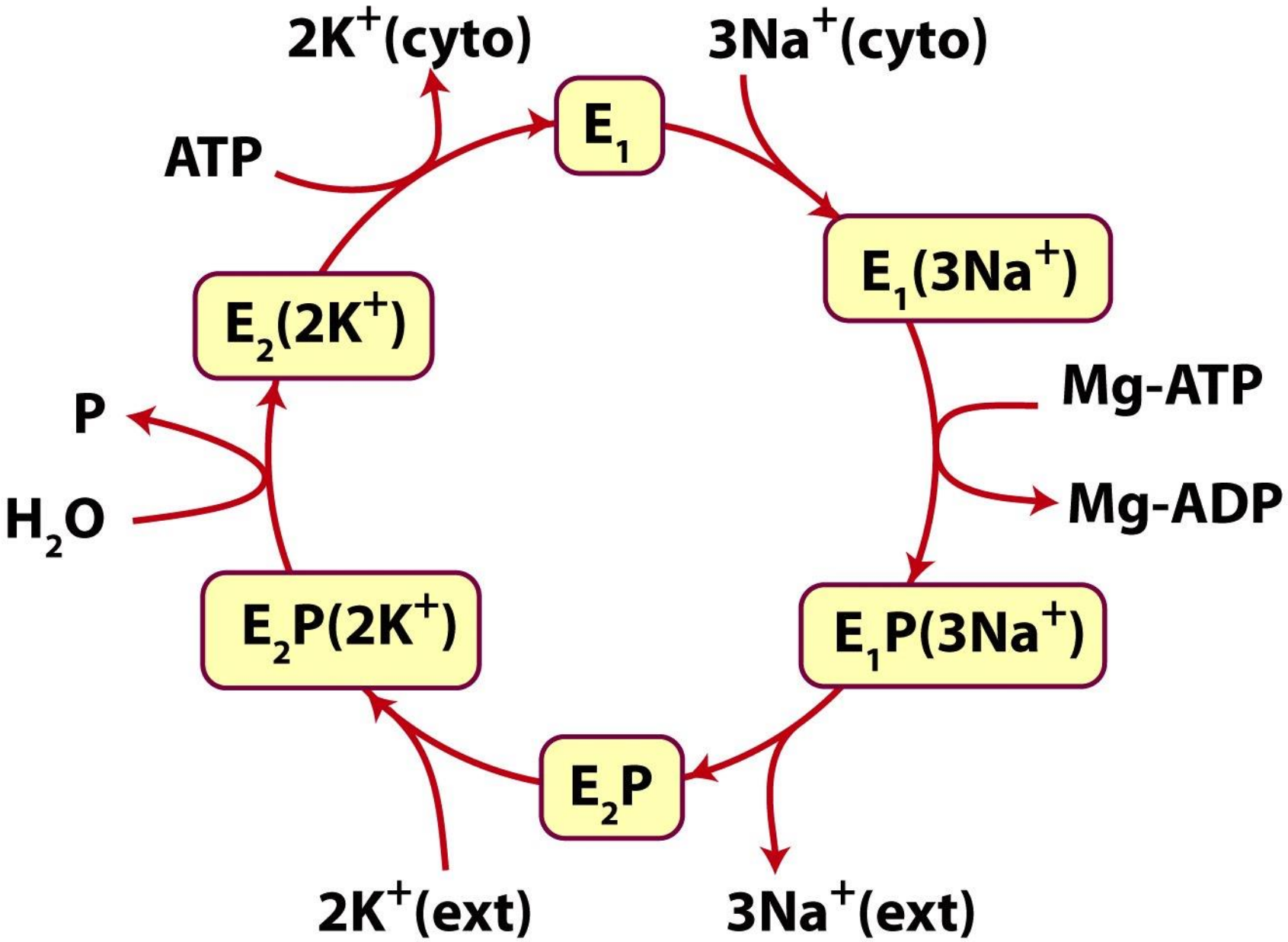
$3Na^+(in) + 2K^+(out) + Mg^{2+}ATP^{4-} + H_2O \rightarrow 3Na^+(out) + 2K^+(in) + Mg^{2+}ADP^{3-} + HPO_4^{2-} + H^+$

Ione	Intracellulare (mM)	Extracellulare (mM)
Na^+	10	150
K^+	100	5

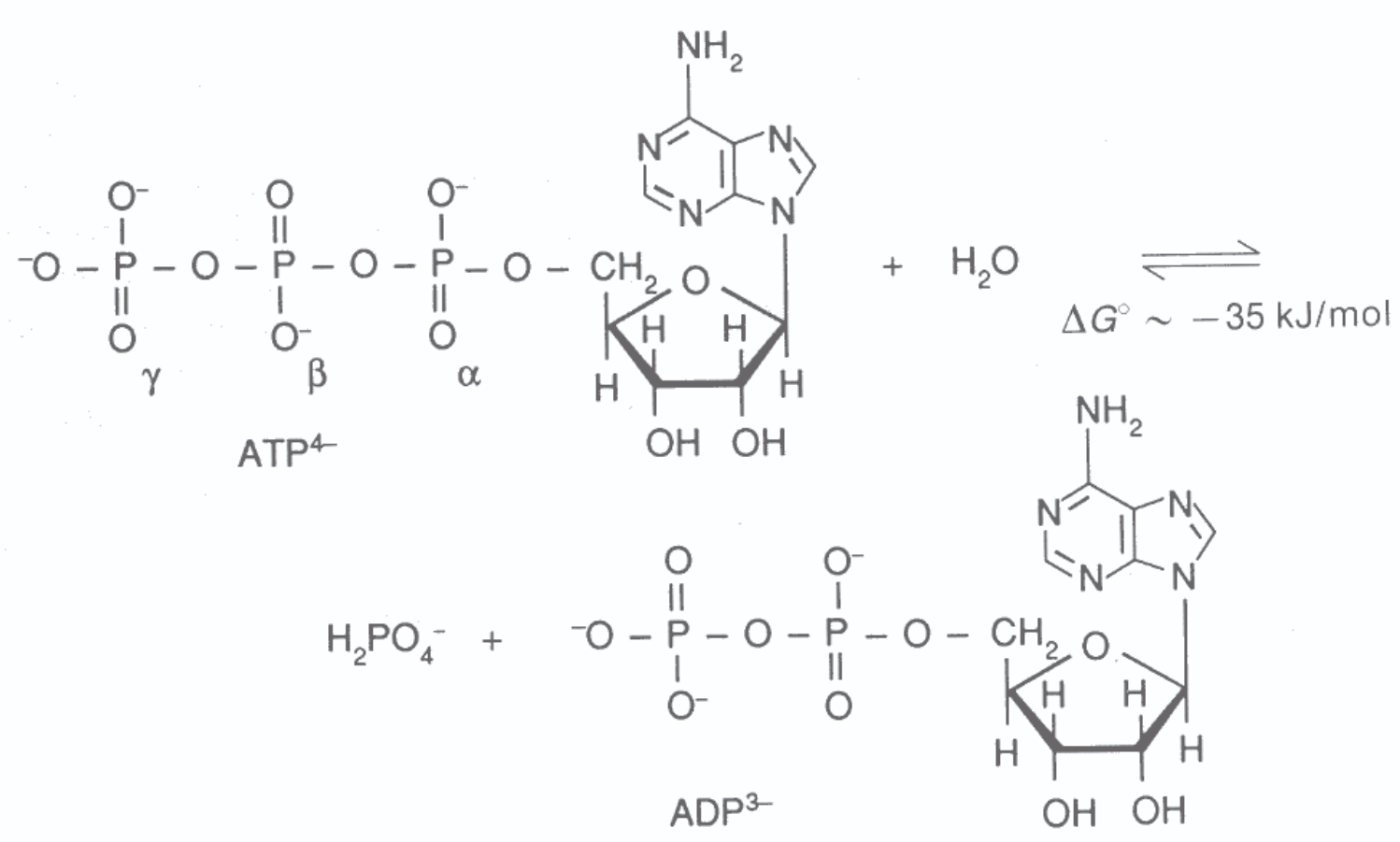




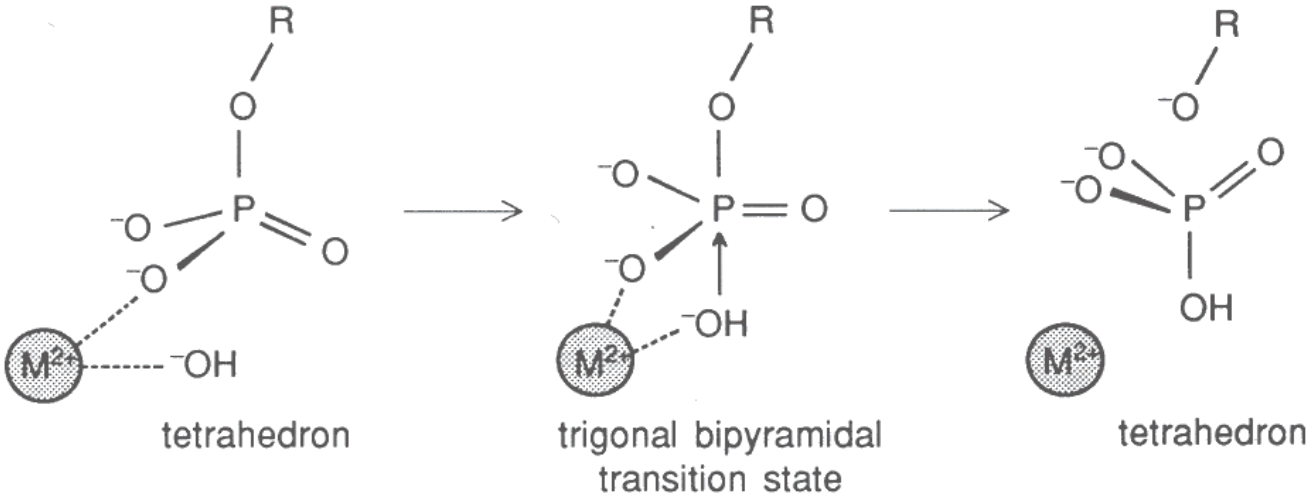
$\alpha_2\beta_2 = 2 \times 112(\alpha) + 2 \times 35(\beta)$

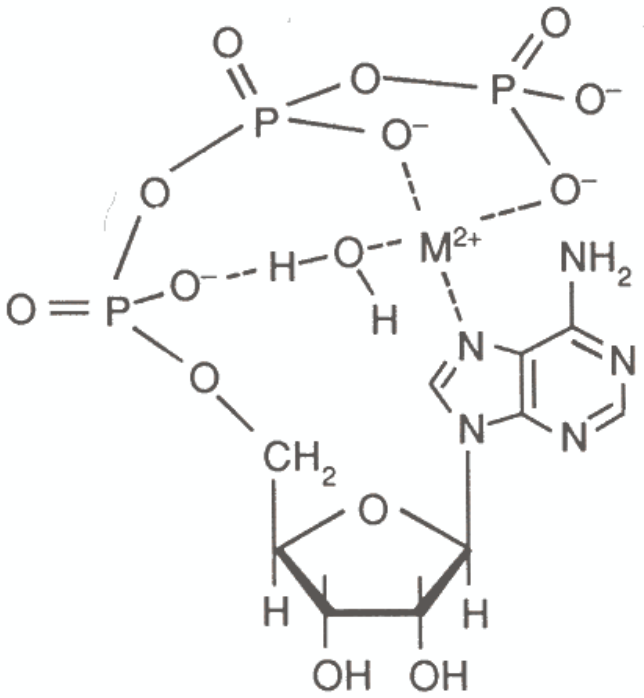

2K+(cyto)
3Na+(cyto)
ATP
E1
E1(3Na+)
E2(2K+)
Mg-ATP
P
H2O
Mg-ADP
E2P(2K+)
E1P(3Na+)
E2P
2K+(ext)
3Na+(ext)

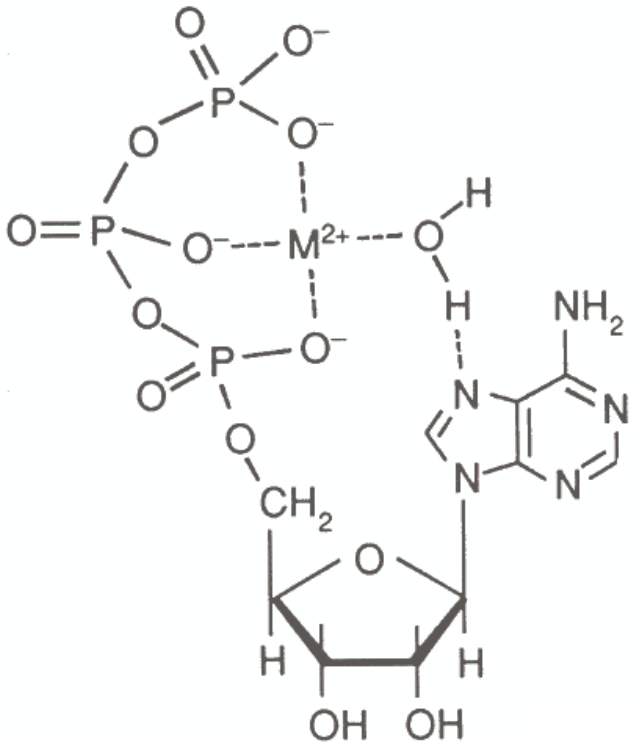
Magnesio


NH2
N
N
N
N
O⁻
O
O⁻
O⁻
P
P
P
O
O
O
O
O
O
O
γ
β
α
CH2
O
H
H
H
H
OH
OH
ATP4–
+
H2O
ΔG° ~ −35 kJ/mol
H2PO4⁻
+
ADP3–

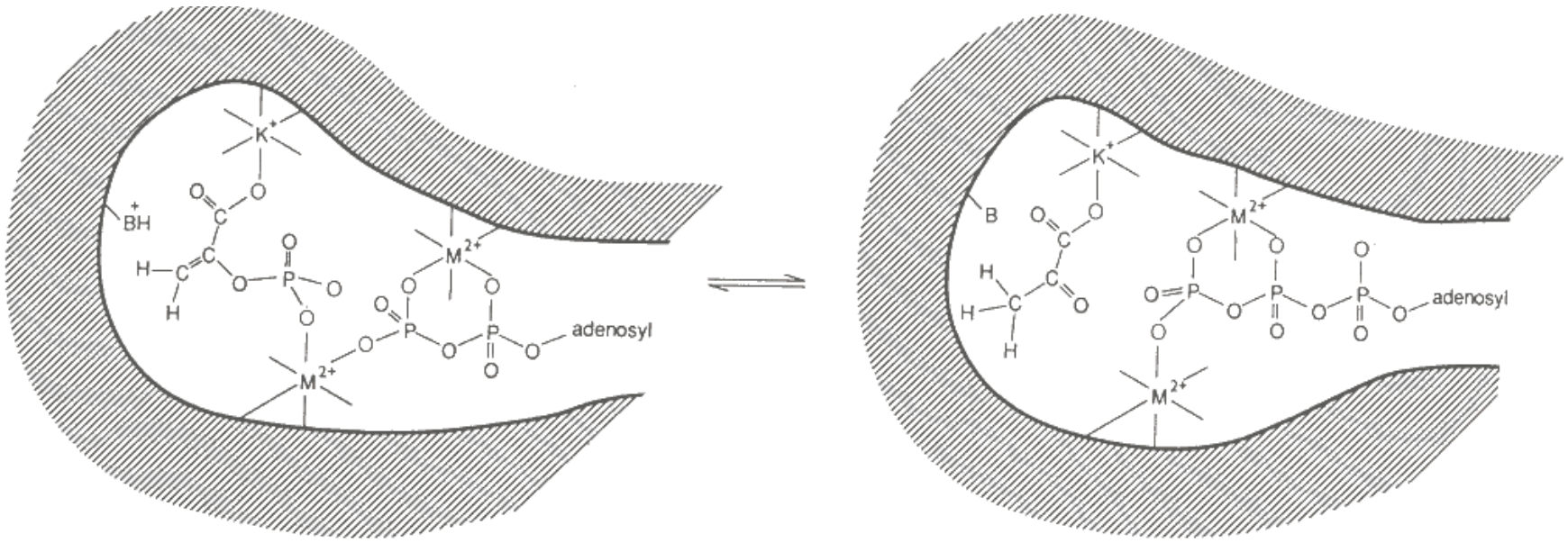
S_N2


R
O
P
O
$^-$O
$^-$O
M^{2+}
$^-$OH
tetrahedron
trigonal bipyramidal
transition state
tetrahedron


O
P
O$^-$
H
M^{2+}
NH_2
N
CH_2
H
OH


O
P
O$^-$
H
M^{2+}
NH_2
N
CH_2
H
OH

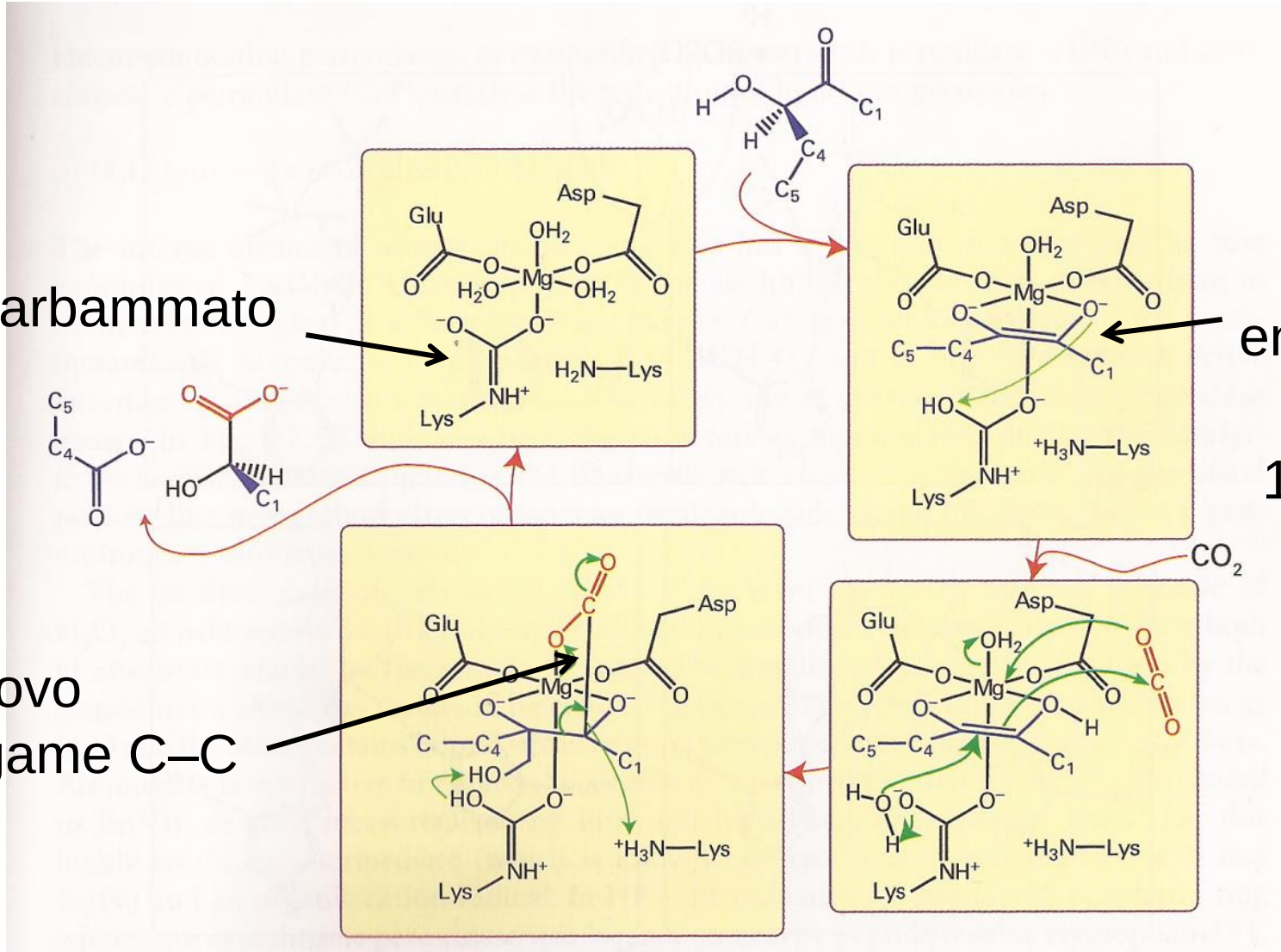
$$ATP^{4-} + X\text{–}H \xrightarrow[Mg^{2+}]{\text{kinasi}} ADP^{3-} + X\text{–}PO_3^{2-} + H^+$$



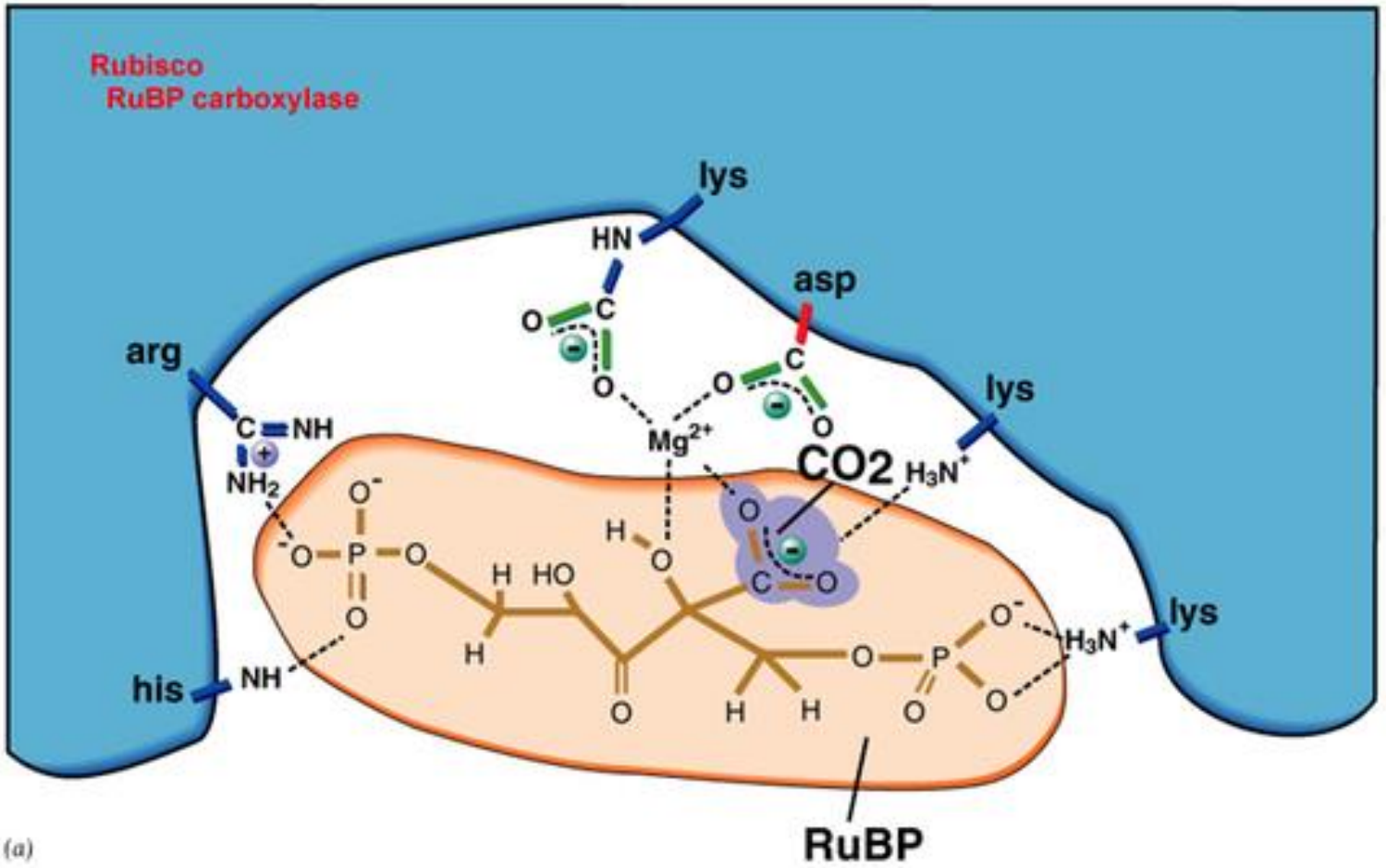
Piruvato-kinasi

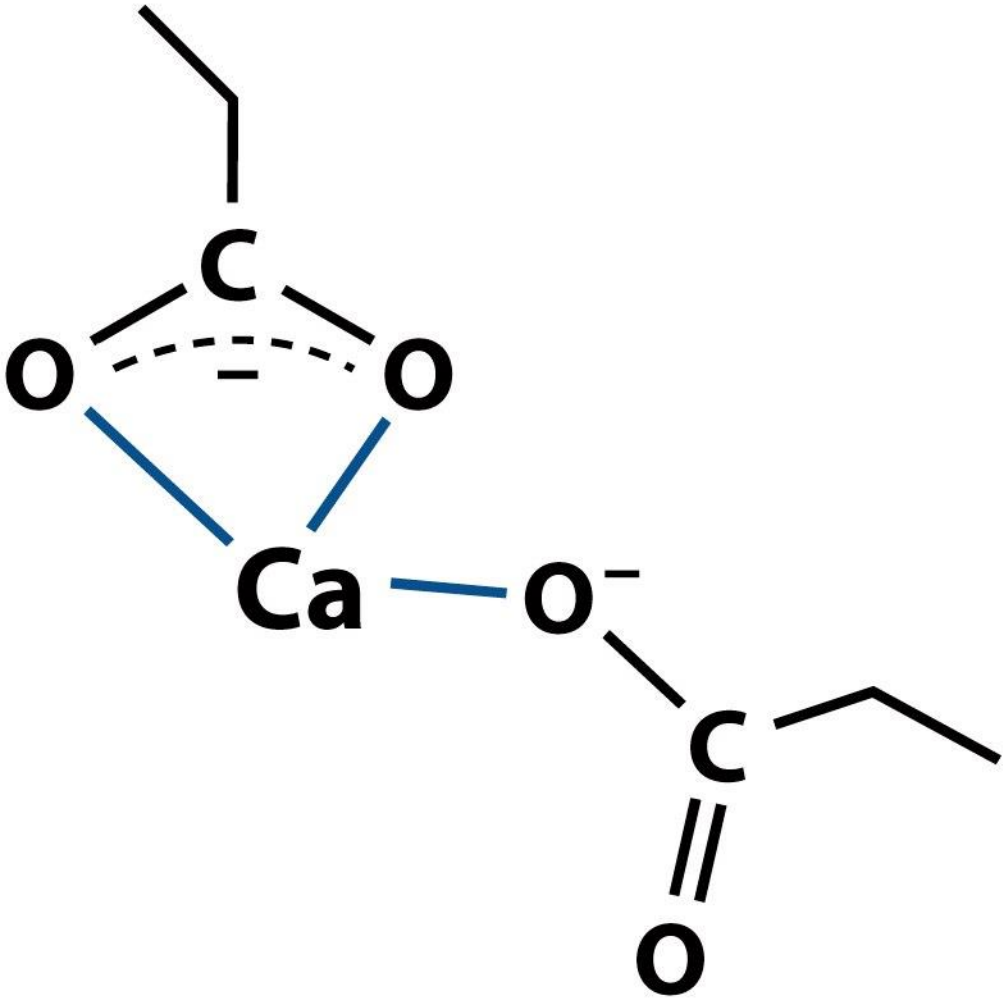
Ribulosio bisfosfato carbossilasi
'rubisco'

D-ribulosio 1,5-disfosfato + CO_2 + H_2O = 2 3-fosfo-D-glicerato + 2 H^+



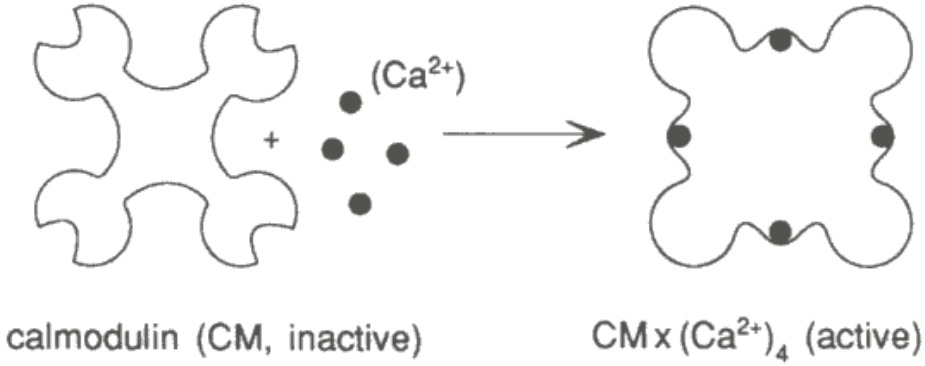
Rubisco

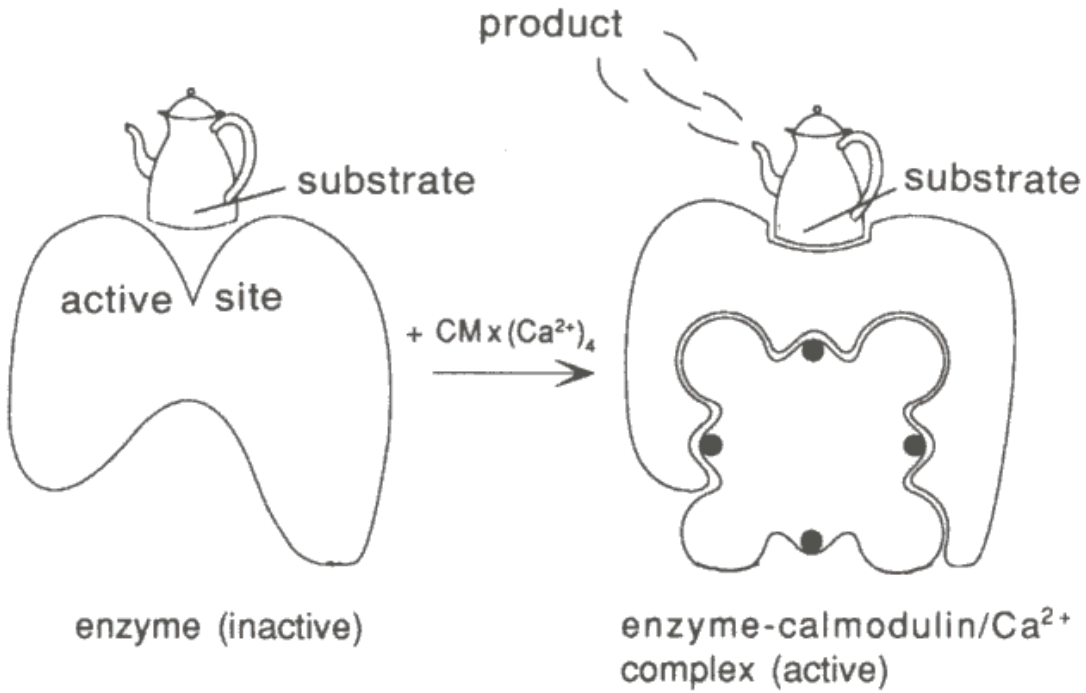

Rubisco
RuBP carboxylase
lys
HN
asp
arg
lys
C NH
NH2
Mg2+
CO2
H3N+
his
NH
lys
H3N+
RuBP
(a)



Ca^{2+} coordination

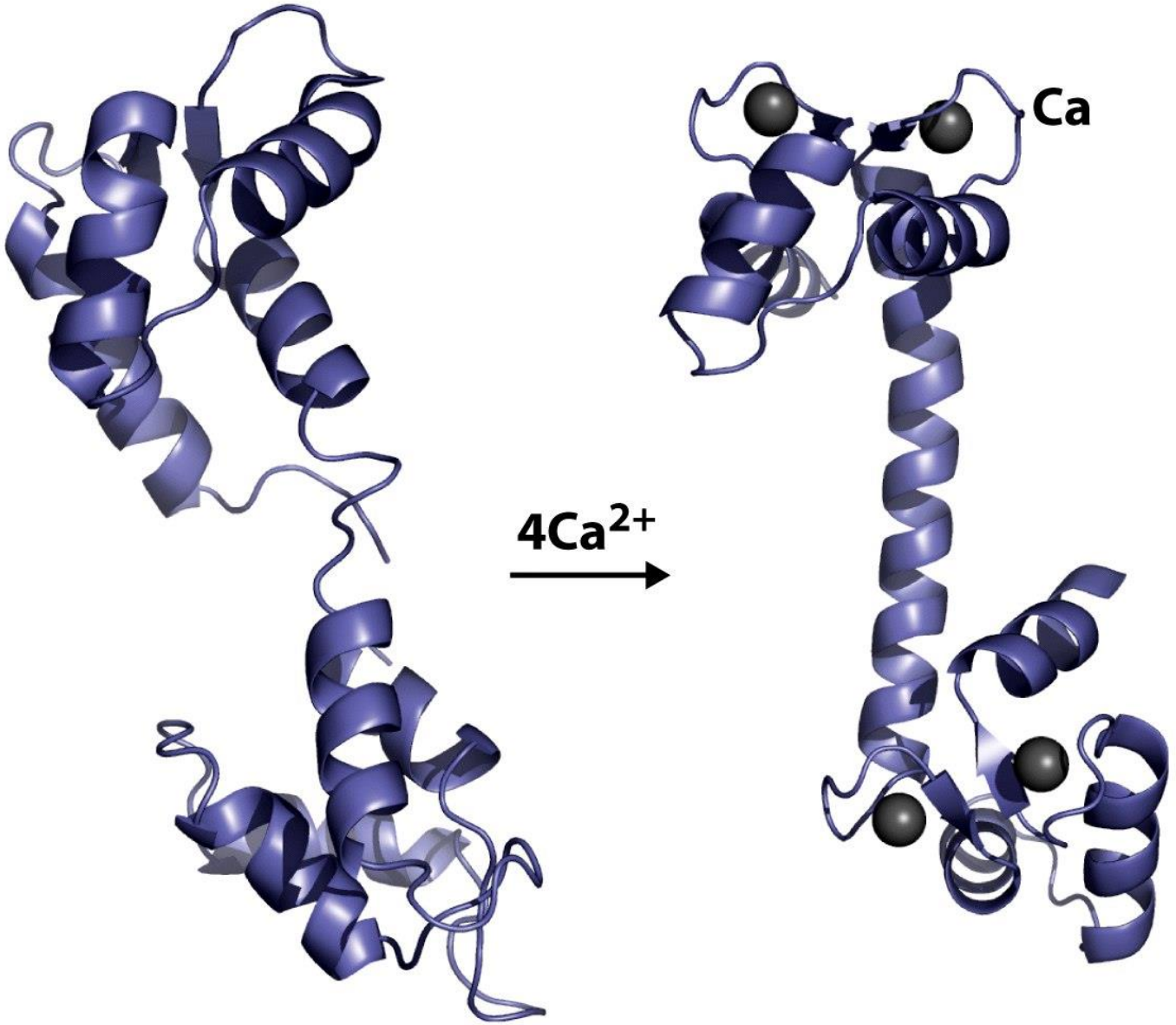
Ca^{2+}: messaggero secondario

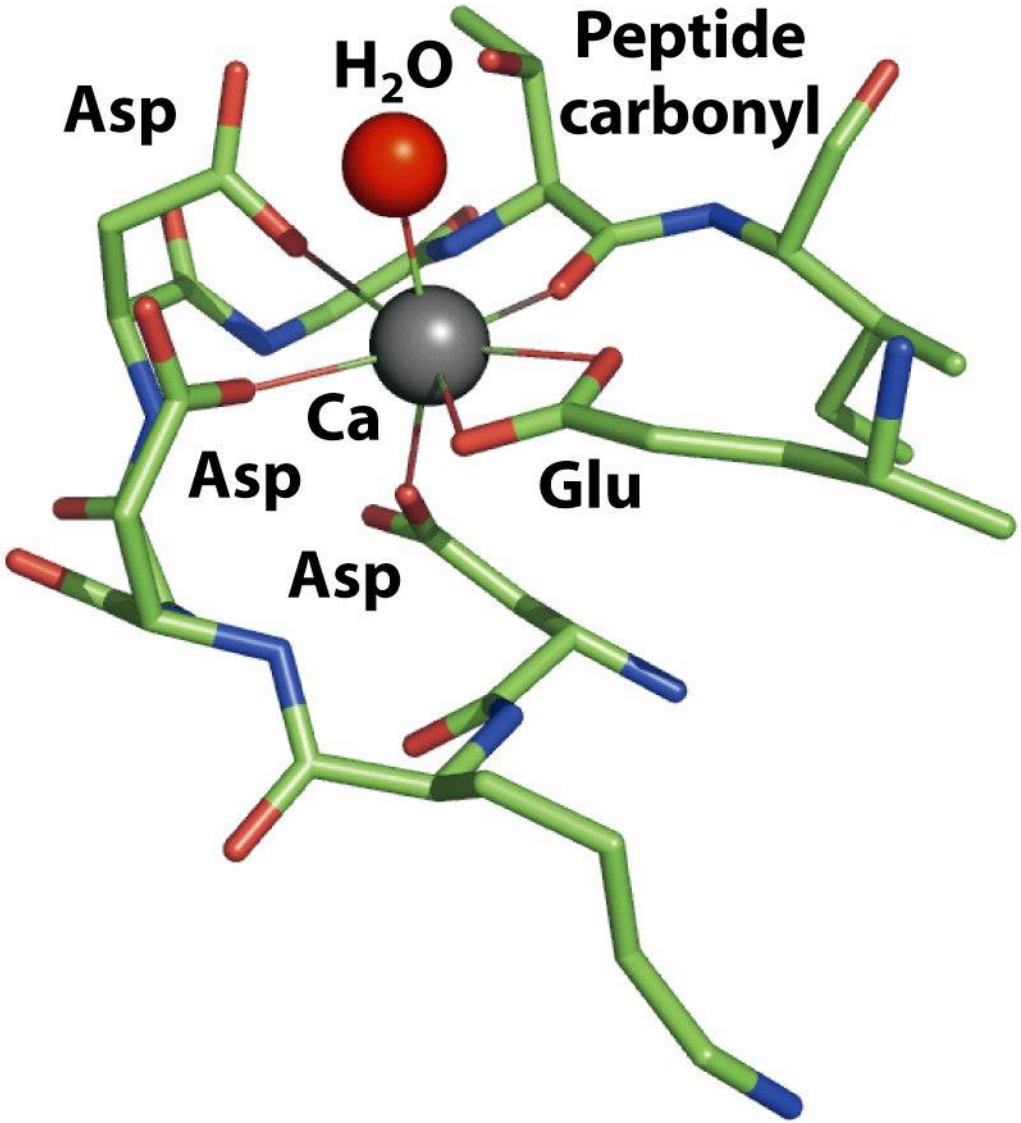




e.g.: NO sintasi (NO), adenilato e guanilato ciclasi (cAMP, cGMP), NAD kinasi (NADP)

CALMODULINA



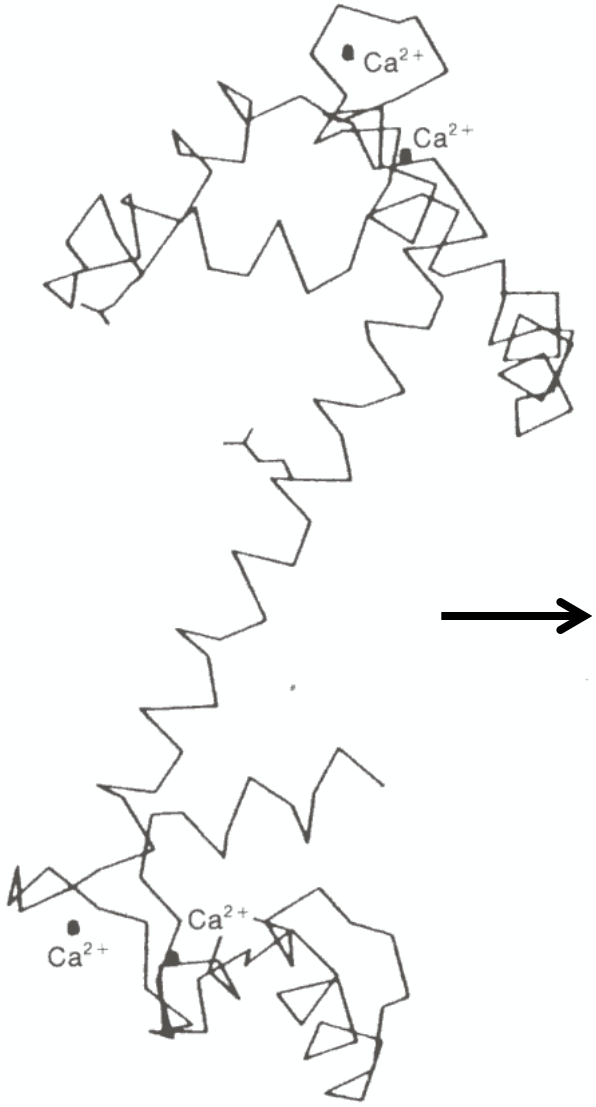


Uno dei siti di coordinazione del Ca^{2+} della apo-calmodulina

TROPONINA-C

Siti di binding ad affinità minore

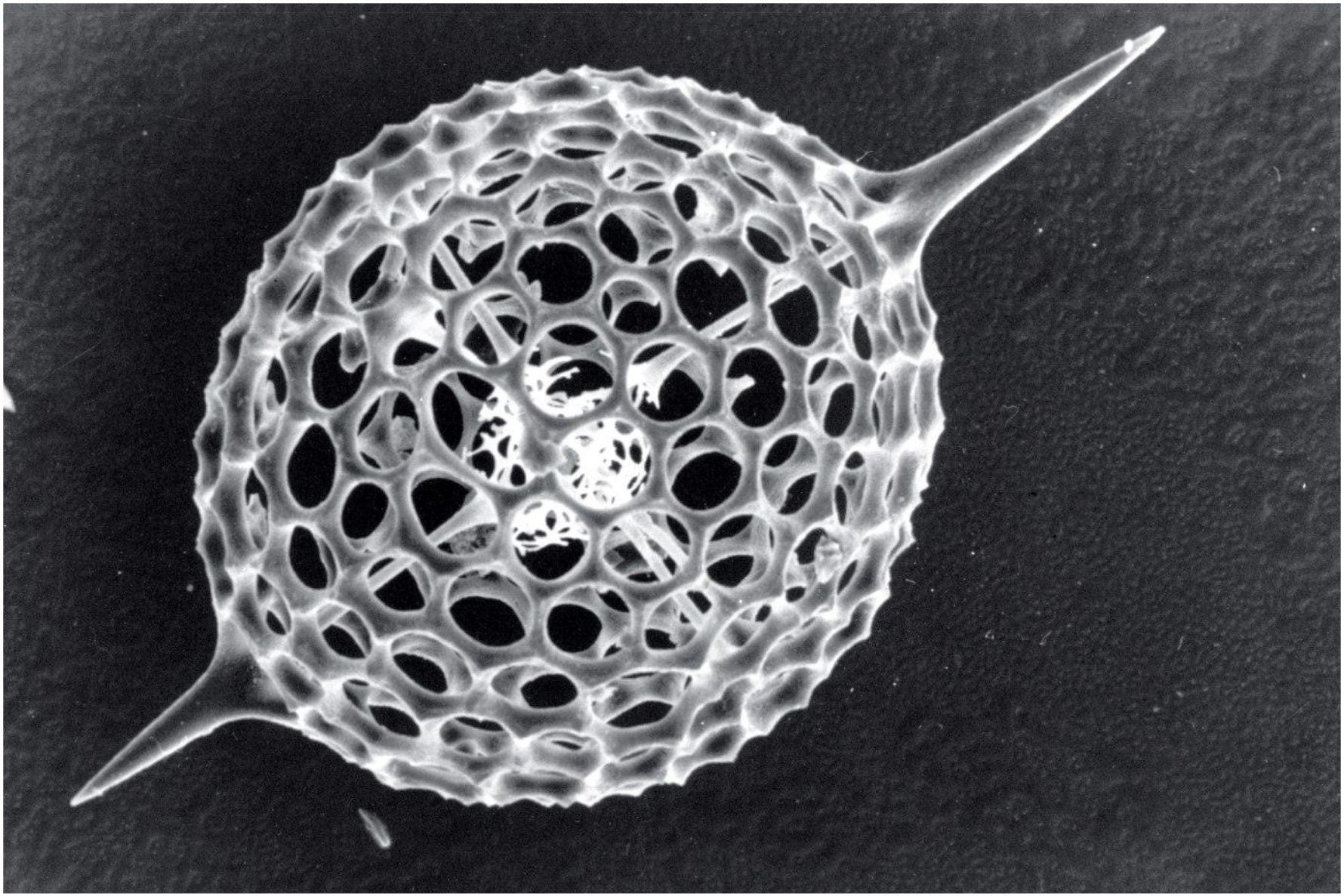
Siti di binding ad elevata affinità ($K > 10^6$)



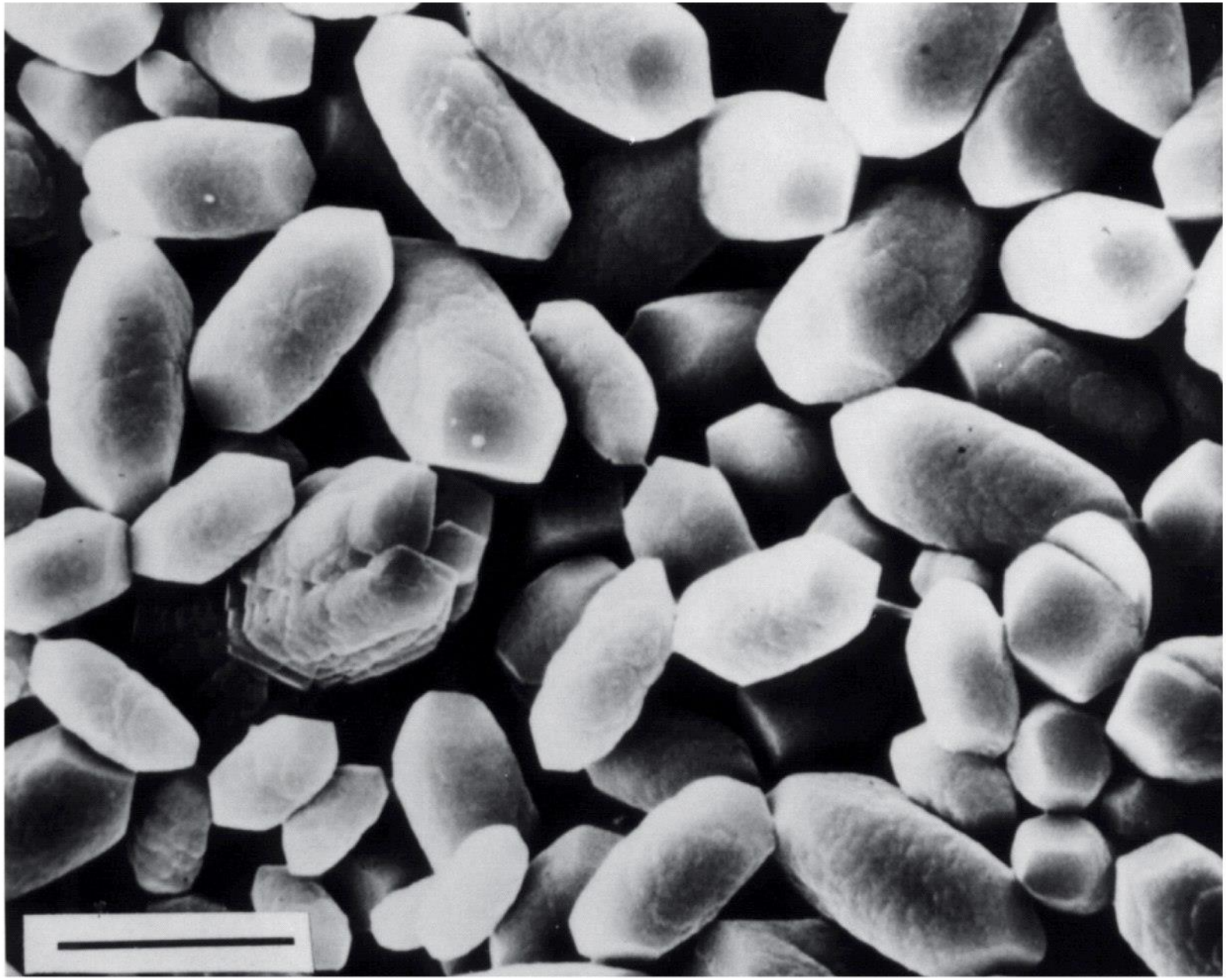
actina
miosina

I principali biominerali di elementi alcalino-terrosi

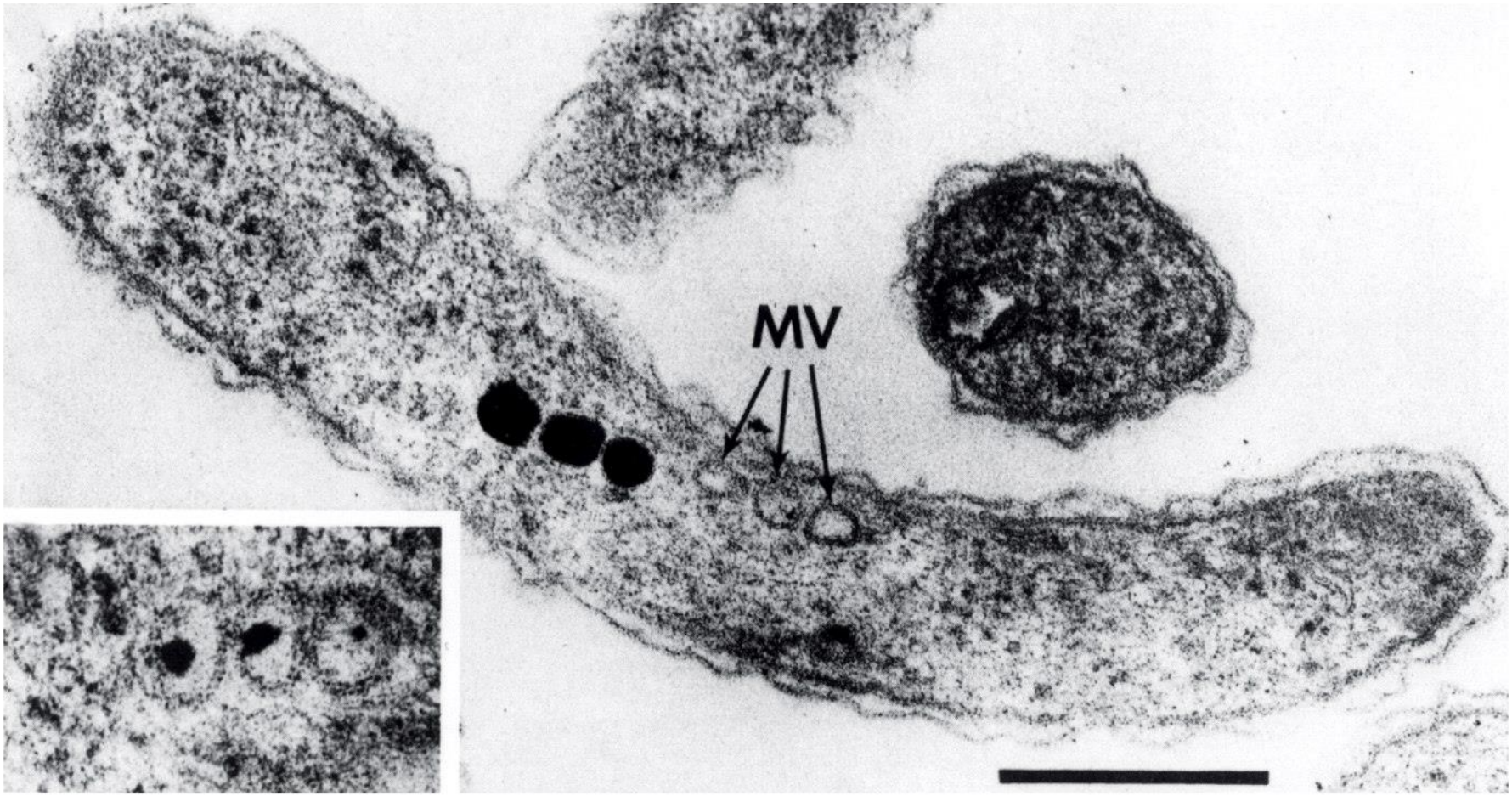
Composto	**Minerale**	**Presenza negli organismi viventi**
$MgCO_3$	Magnesite	Scheletro del corallo
$CaCO_3$	Aragonite	Conchiglie e perle
$CaCO_3$	Calcite	Uova di uccello, sistemi gravitazionali nell'orecchio interno
$CaCO_3 \cdot nH_2O$	Amorfo	Immagazzinamento di calcio nelle piante
$Ca(C_2O_4) \cdot nH_2O$	Whewellite (n = 1) Weddellite (n = 2)	Immagazzinamento di calcio nelle piante, calcoli renali
$3Ca_3(PO_4)_2 \cdot Ca(OH)_2$	Idrossiapatite	Ossa e denti nei vertebrati
$CaSO_4 \cdot 2H_2O$	Gesso	Sistema gravitazionale nelle meduse
$SrSO_4$	Celestite	Esoscheletro di certo plankton
$BaSO_4$	Barite	Sistema gravitazionale nelle alghe



Microscheletro siliceo di una radiolaria



Cristalli di calcite nell’orecchio interno: sensori gravitazionali



Cristalli di magnetite in archeobatteri magnetotattici

Table 26.1 The approximate concentrations (mol dm^{-3}), where known, of elements (apart from C, H, O, N, P, S, Se, Br, I, B, Si and W) in different biological zones

Element	External fluids (sea water)	Free ions in external fluids (blood plasma)	Cytoplasm (free ions)	Comments on status in cell
Na	$>10^{-1}$	10^{-1}	$<10^{-2}$	Not bound
K	10^{-2}	4×10^{-3}	$\leq 3\times10^{-1}$	Not bound
Mg	$>10^{-2}$	10^{-3}	$\approx10^{-3}$	Weakly bound as ATP complex
Ca	$>10^{-3}$	10^{-3}	$\approx10^{-7}$	Concentrated in some vesicles
Cl	10^{-1}	10^{-1}	10^{-2}	Not bound
Fe	10^{-17} (Fe(III))	10^{-16} (Fe(III))	$<10^{-7}$ (Fe(II))	Too much unbound Fe is toxic (Fenton chemistry) in and out of cells
Zn	$<10^{-8}$	10^{-9}	$<10^{-11}$	Totally bound, but may be exchangeable
Cu	$<10^{-10}$ (Cu(II))	10^{-12}	$<10^{-15}$ (Cu(I))	Totally bound, not mobile. Mostly outside cytoplasm
Mn	10^{-9}		$\approx10^{-6}$	Higher in chloroplasts and vesicles
Co	10^{-11}		$<10^{-9}$	Totally bound (cobalamin)
Ni	10^{-9}		$<10^{-10}$	Totally bound
Mo	10^{-7}		$<10^{-7}$	Mostly bound