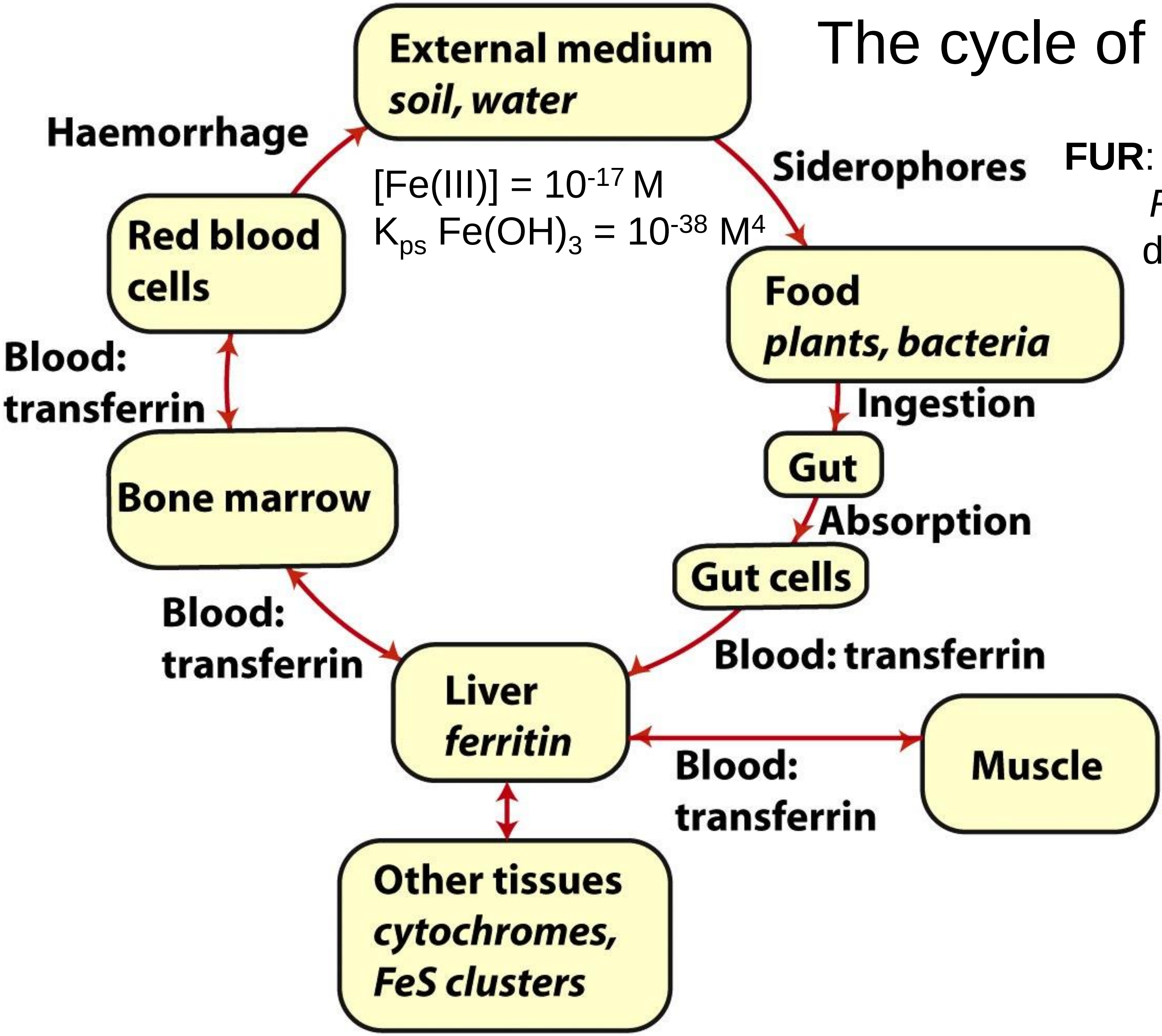
Homeostasis of trace metals: uptake, transport, delivery and processing, storage, excretion.

The cycle of iron



$[Fe(III)] = 10^{-17}$ M

K_{ps} $Fe(OH)_3 = 10^{-38}$ M^4

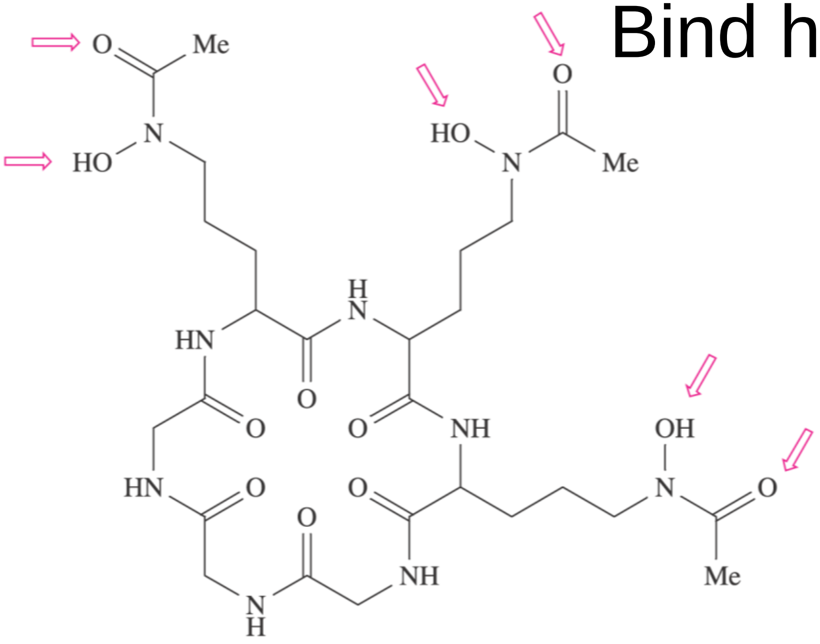
FUR: *Fe Uptake Regulation*, depends on [Fe(II)]

Hydroxamate siderophores

(fungi, yiest)

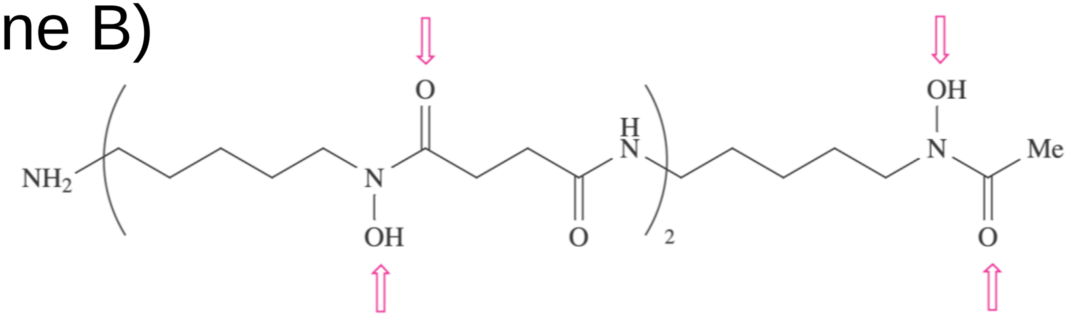
Bind high spin Fe(III)

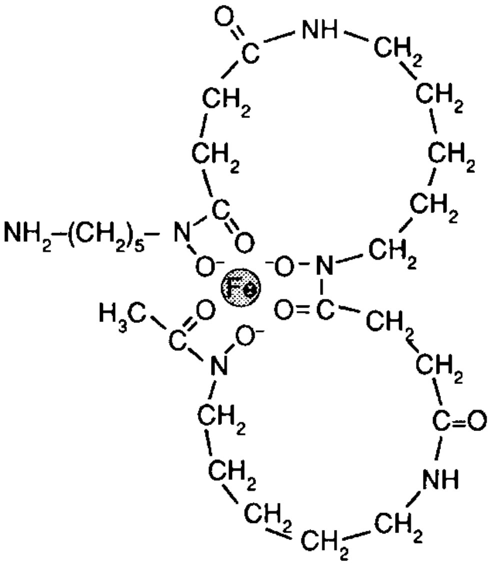
desferrichrome



(a)

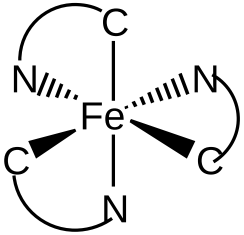
deferoxamine (or desferrioxamine B)



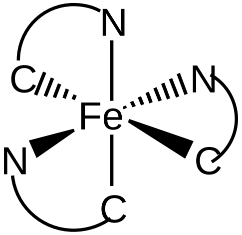


ferrioxamine B

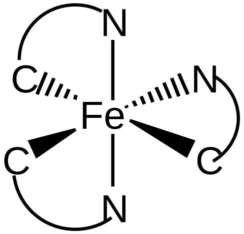
without Fe: deferrioxamine B



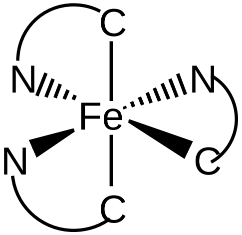
Λ-N-*cis,cis*



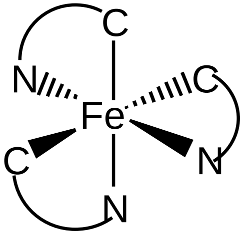
Λ-C-*trans,cis*



Λ-N-*trans,cis*



Λ-C-*cis,trans*

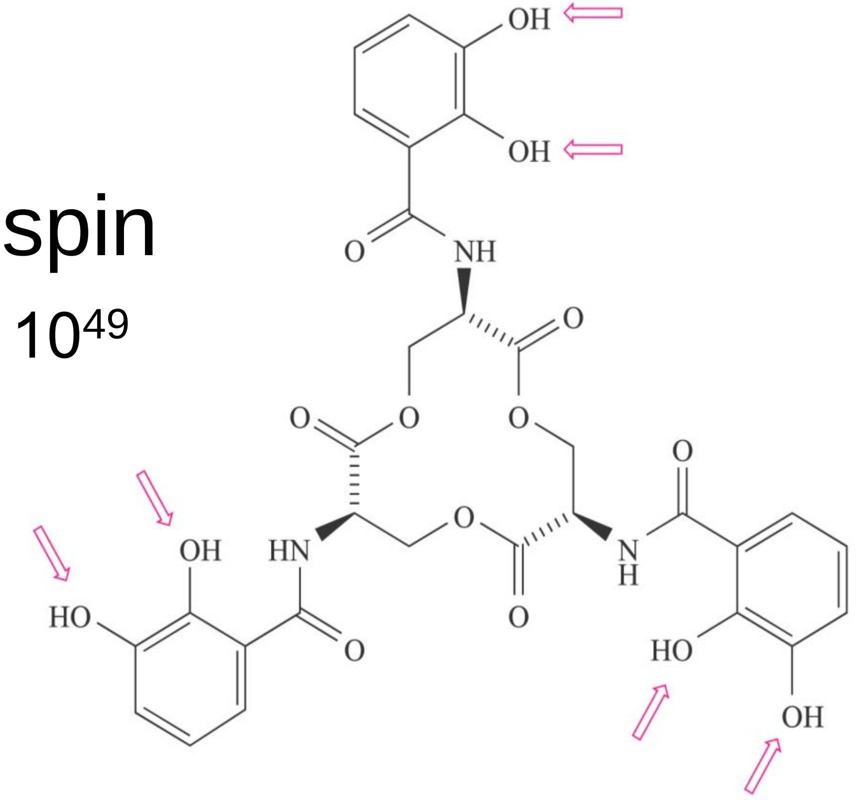


Λ-N-*trans,cis*

Catecholate siderophores

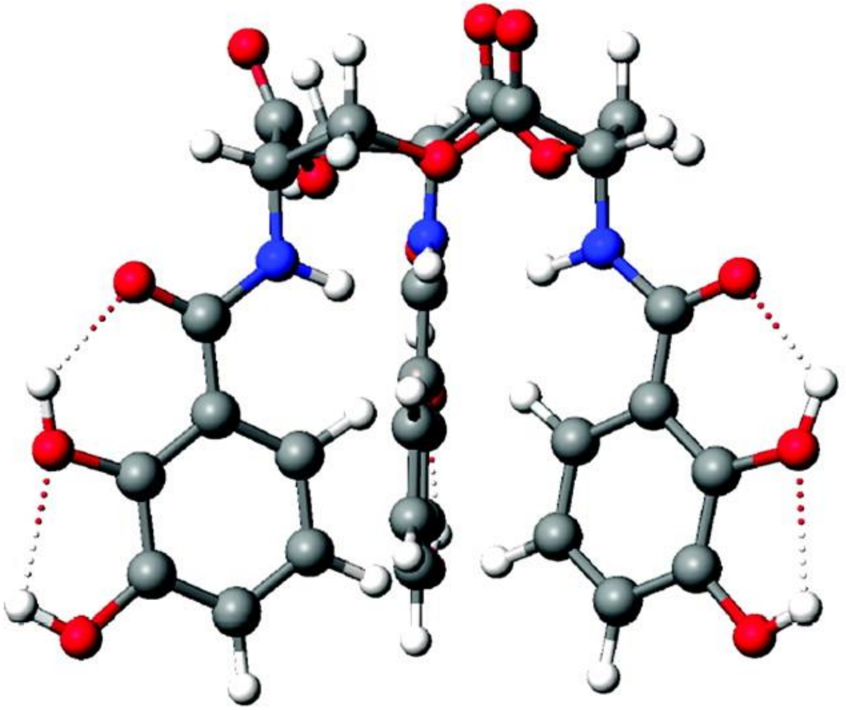
(bacteria)

Binds high spin Fe(III), $K_a = 10^{49}$

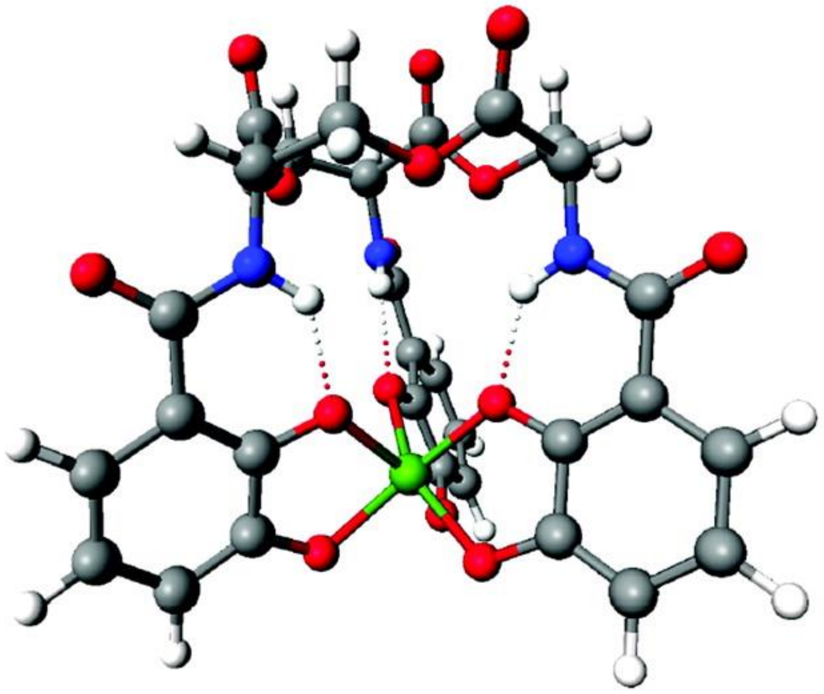


Enterobactin

the highest-affinity iron chelator yet identified in the natural world



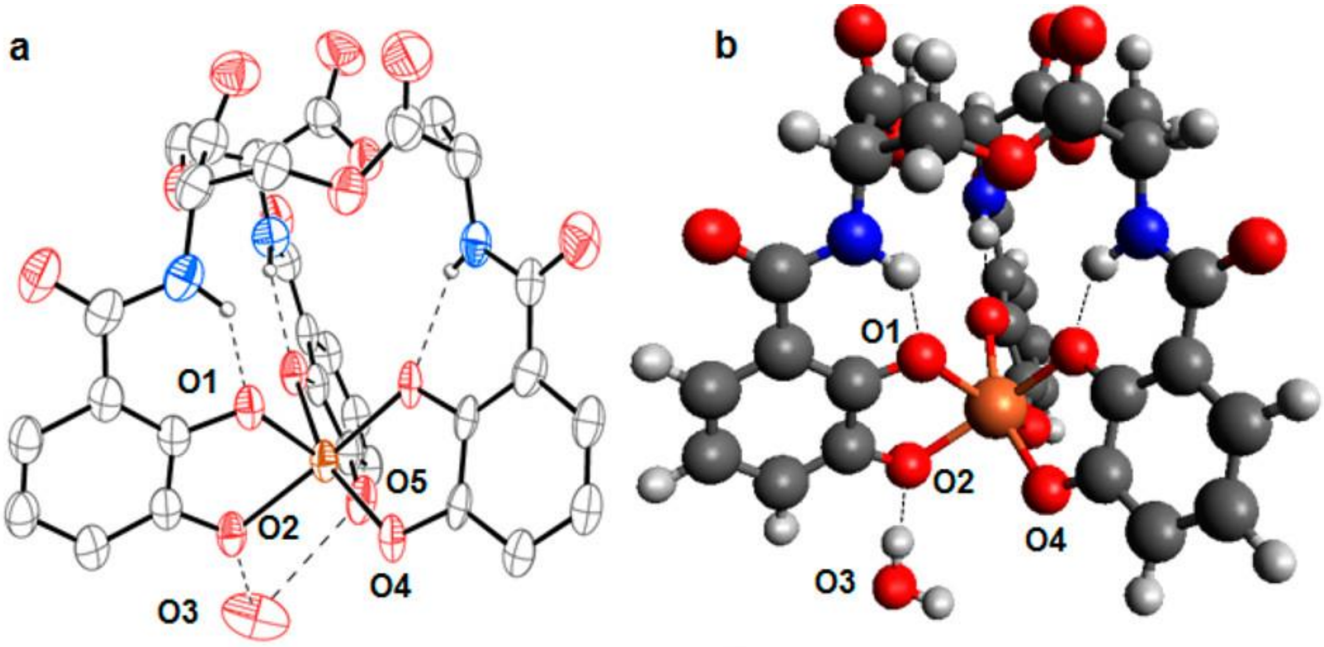
Model for uncoordinated enterobactin



Structure of V(IV)-enterobactin complex (1992)

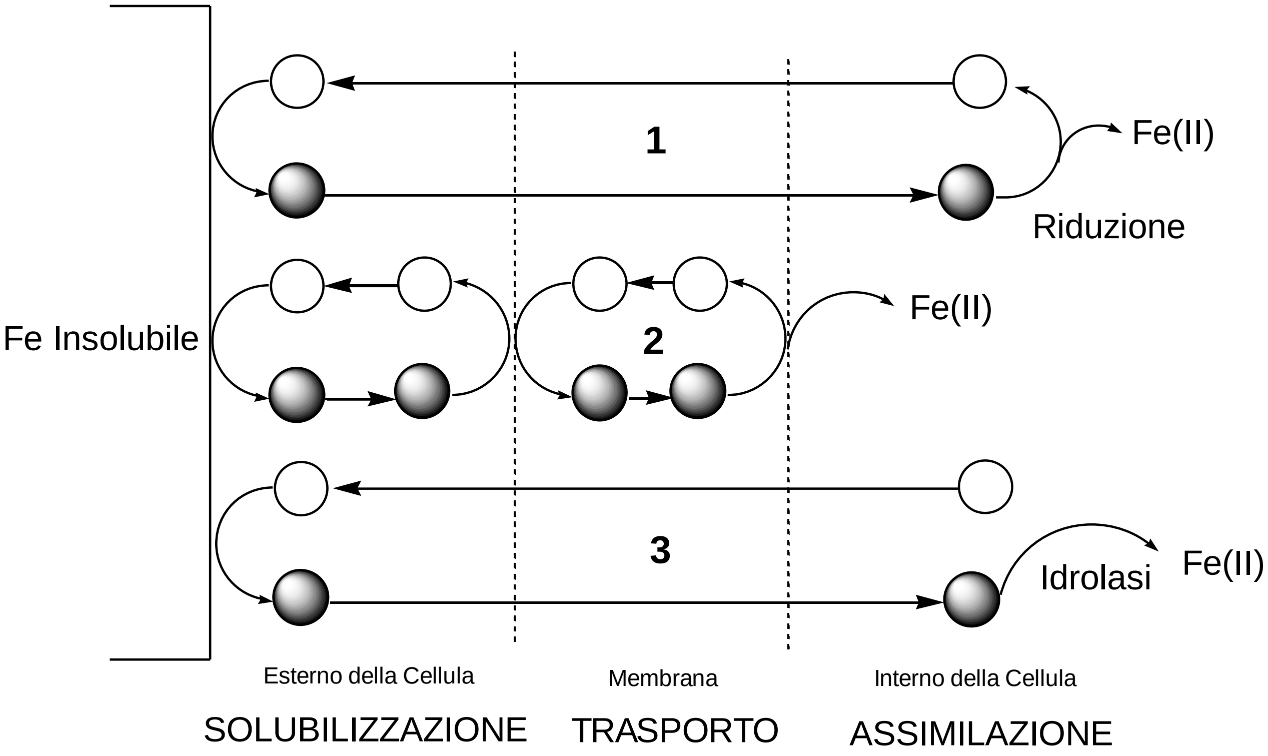
Δ

The recently determined crystal structure affords a definitive Δ assignment to $[Fe(ent)]^{3-}$



J. Am. Chem. Soc. **2017**, *139*, 15245-15250

trans-membrane transport mechanisms

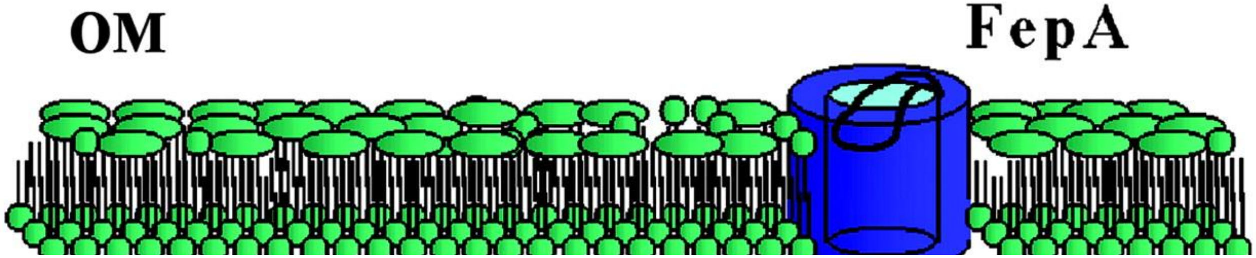


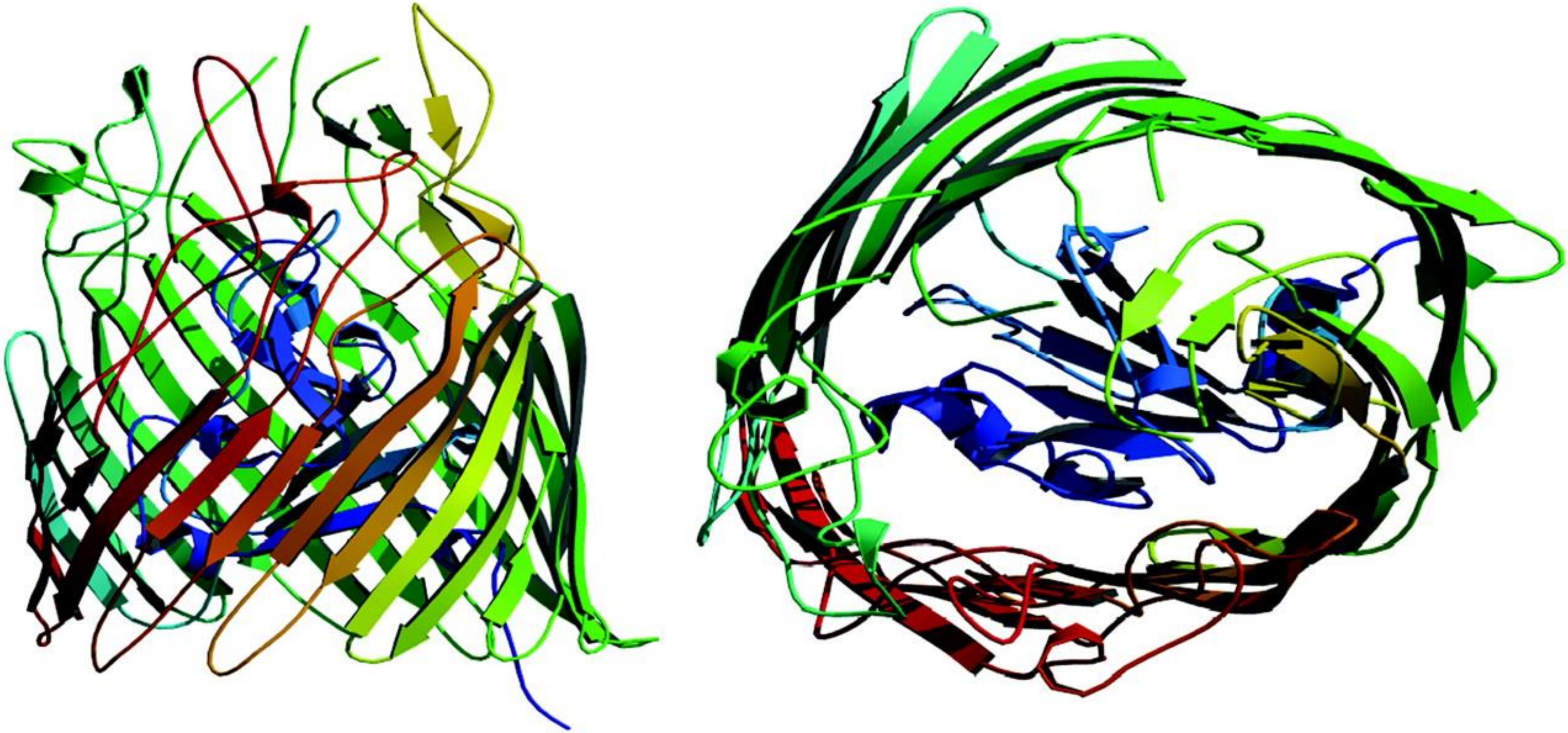
ferrichrome

desferrioxamine

enterobactin

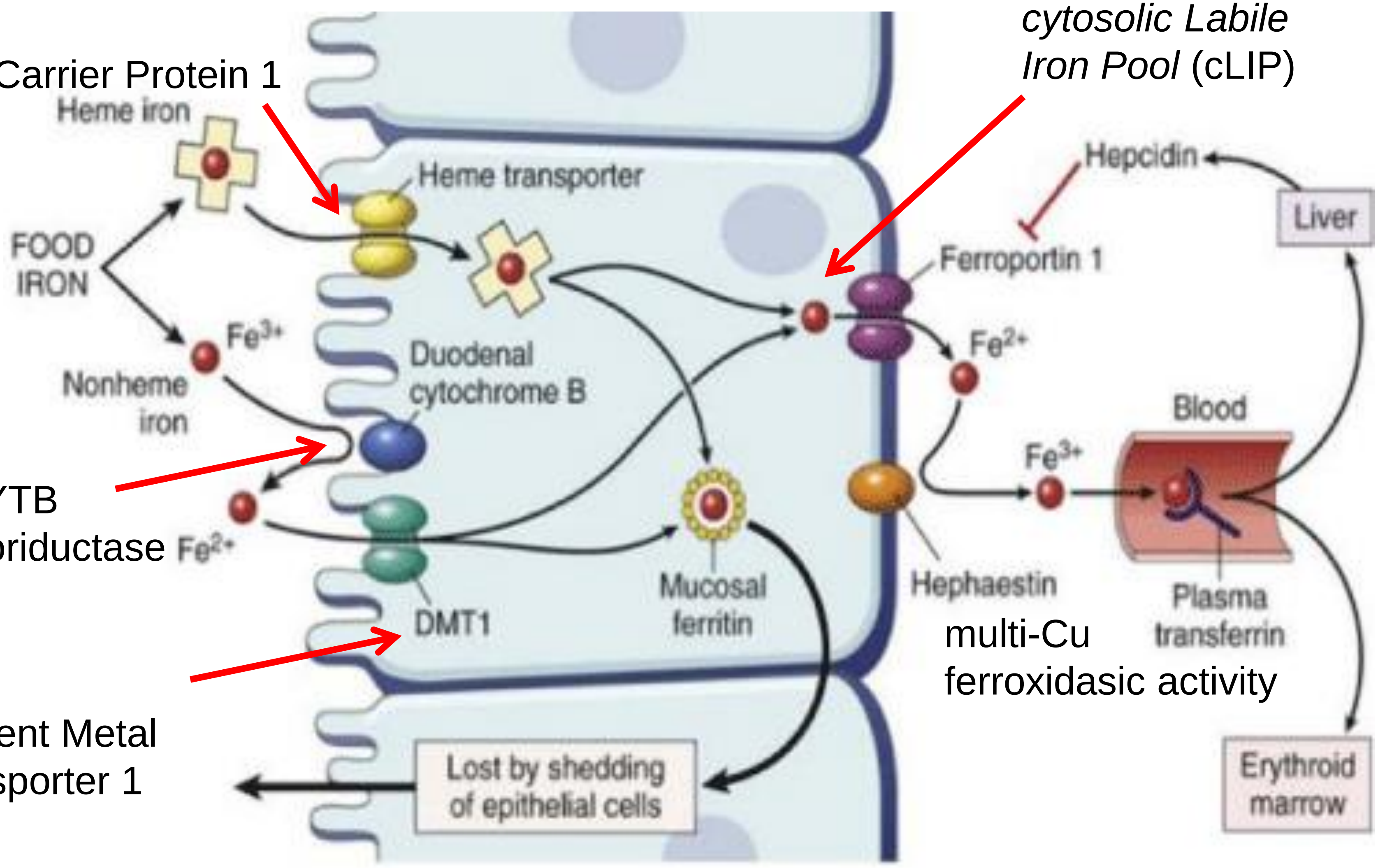
FepA: trans-membrane transport of Fe-enterobactin





The N-terminal «gate protein» is in blue

Iron absorption in the intestine and transport through enterocytes in the duodenum



HCP-1
Heme Carrier Protein 1

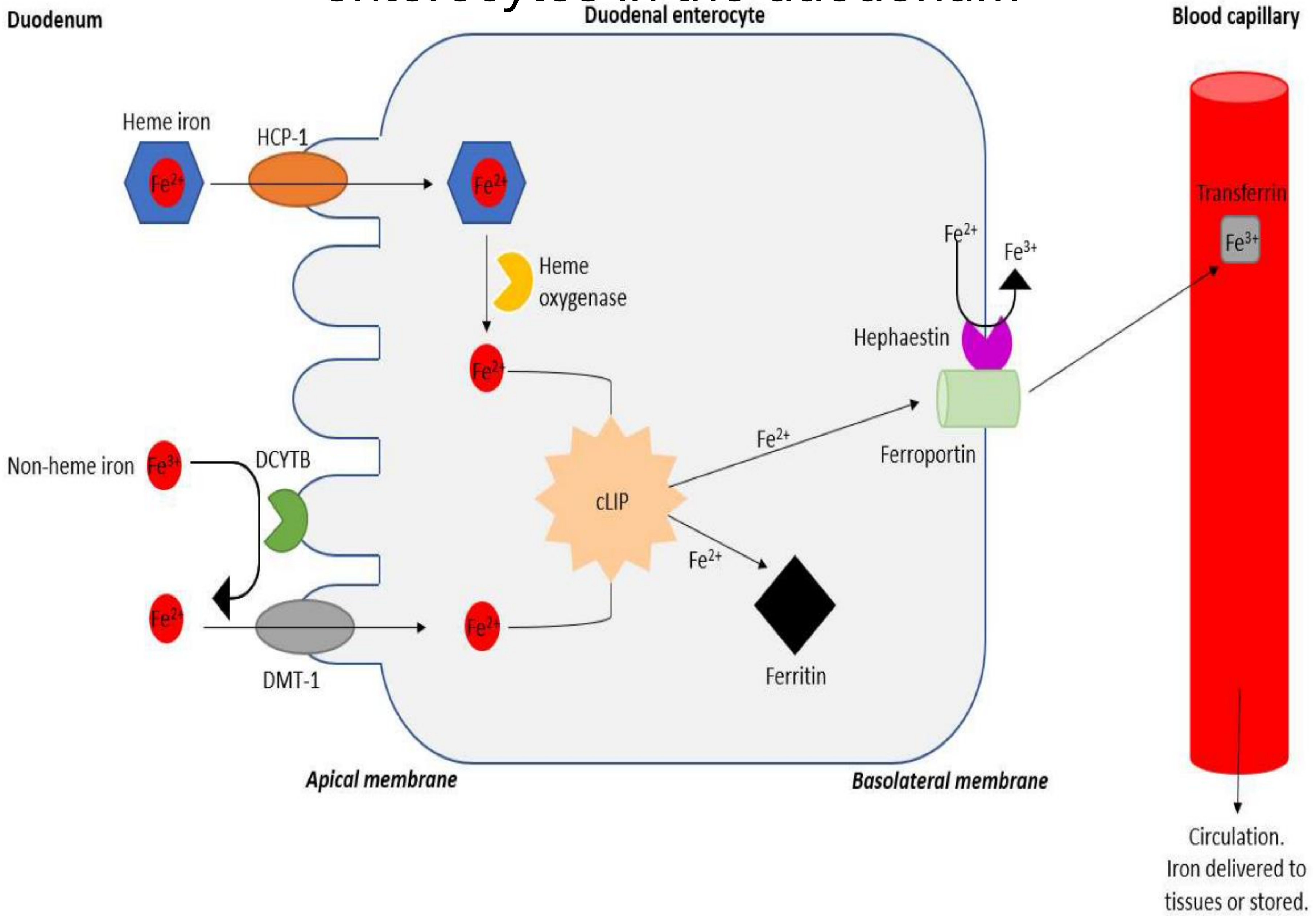
cytosolic Labile Iron Pool (cLIP)

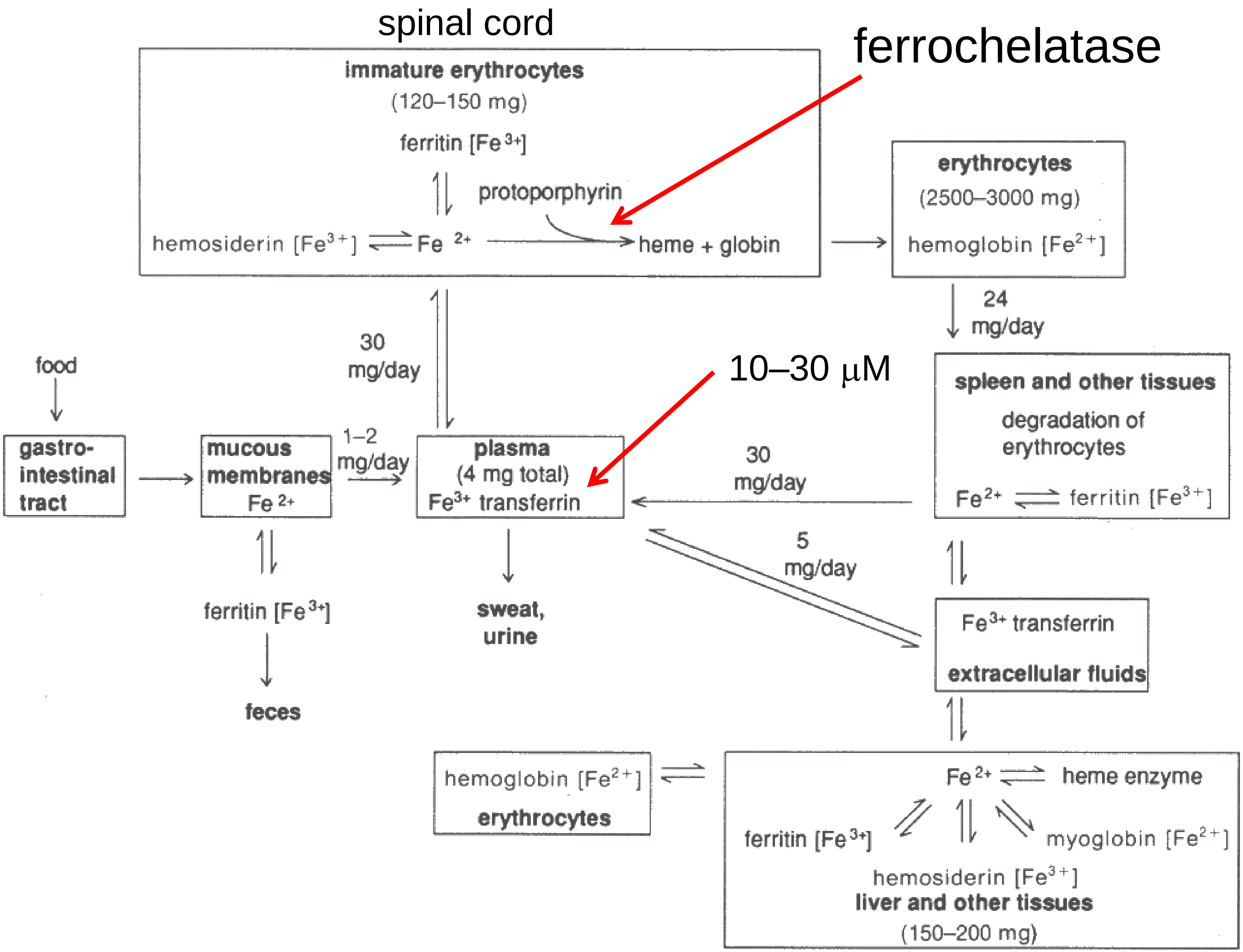
DCYTB
ferroriductase

Divalent Metal
Transporter 1

multi-Cu
ferroxidasic activity

Iron absorption in the intestine and transport through enterocytes in the duodenum





spinal cord

ferrochelatase

immature erythrocytes
(120–150 mg)
ferritin [Fe^{3+}]
protoporphyrin
hemosiderin [Fe^{3+}] ⇌ Fe^{2+} → heme + globin

erythrocytes
(2500–3000 mg)
hemoglobin [Fe^{2+}]

24 mg/day

30 mg/day

10–30 μM

food

gastro-intestinal tract

mucous membranes
Fe^{2+}

1–2 mg/day

plasma
(4 mg total)
Fe^{3+} transferrin

30 mg/day

spleen and other tissues
degradation of erythrocytes
Fe^{2+} ⇌ ferritin [Fe^{3+}]

5 mg/day

ferritin [Fe^{3+}]

feces

sweat, urine

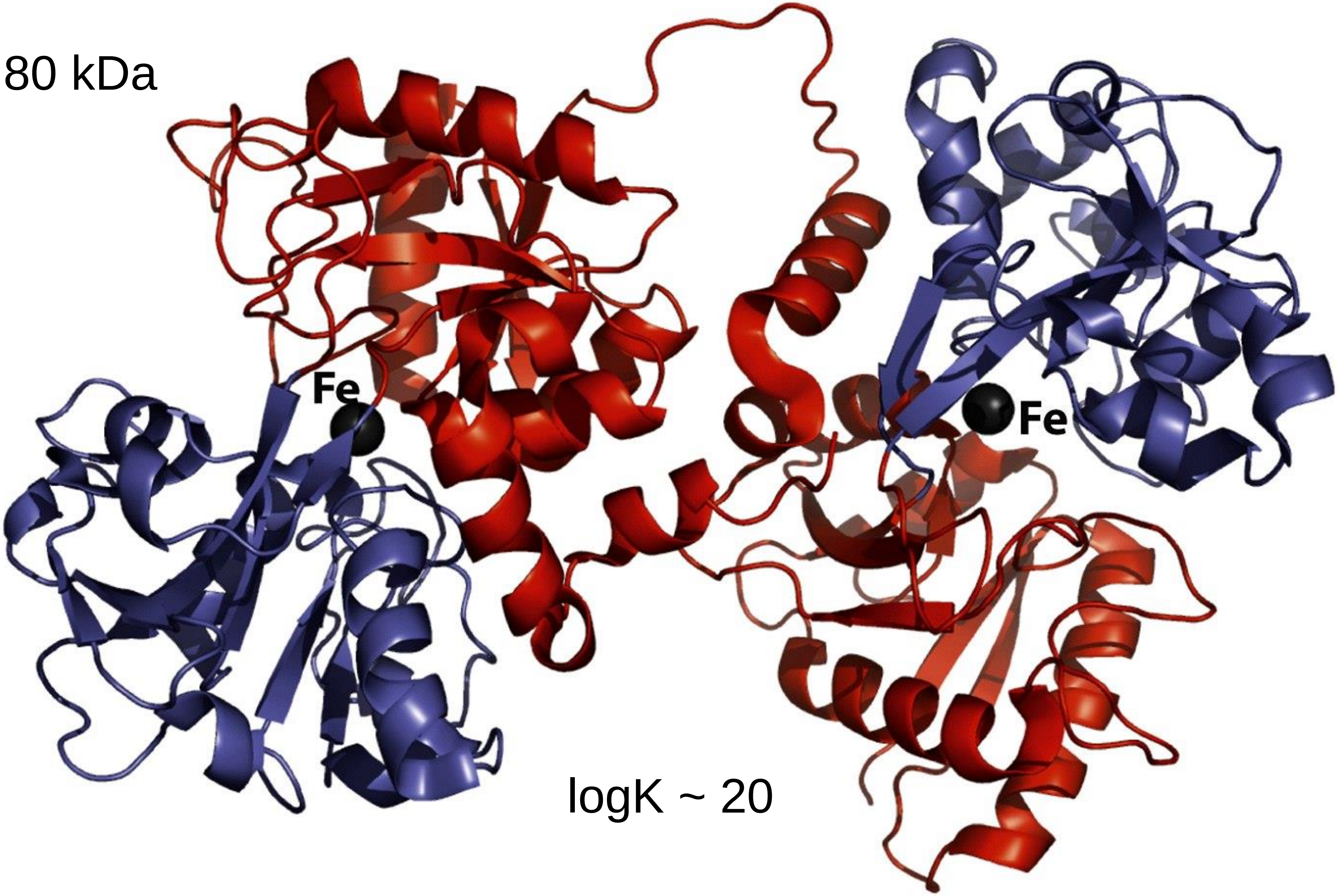
Fe^{3+} transferrin
extracellular fluids

hemoglobin [Fe^{2+}]
erythrocytes

Fe^{2+} ⇌ heme enzyme
ferritin [Fe^{3+}]
myoglobin [Fe^{2+}]
hemosiderin [Fe^{3+}]
liver and other tissues
(150–200 mg)

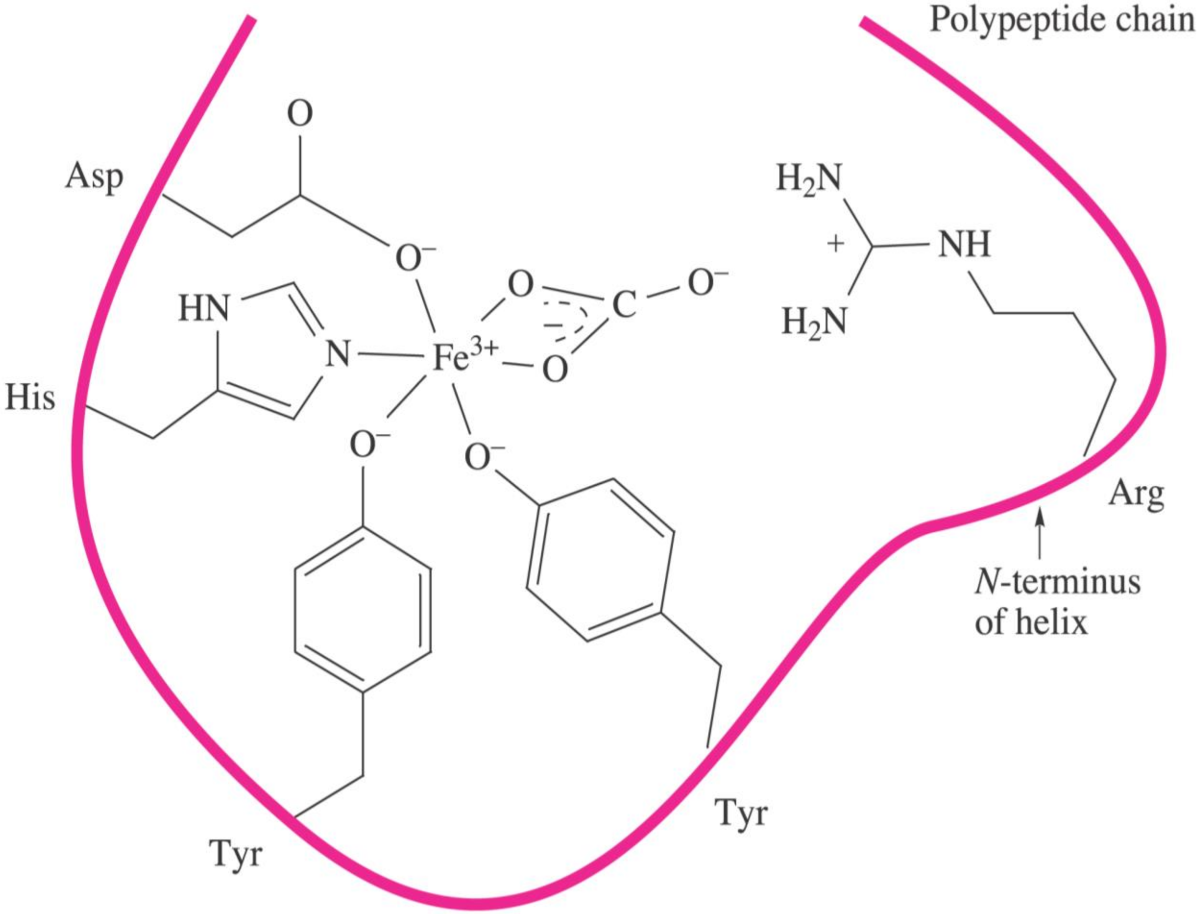
Transferrin
2 lobes, 2 Fe(III) coordination sites

80 kDa



logK ~ 20

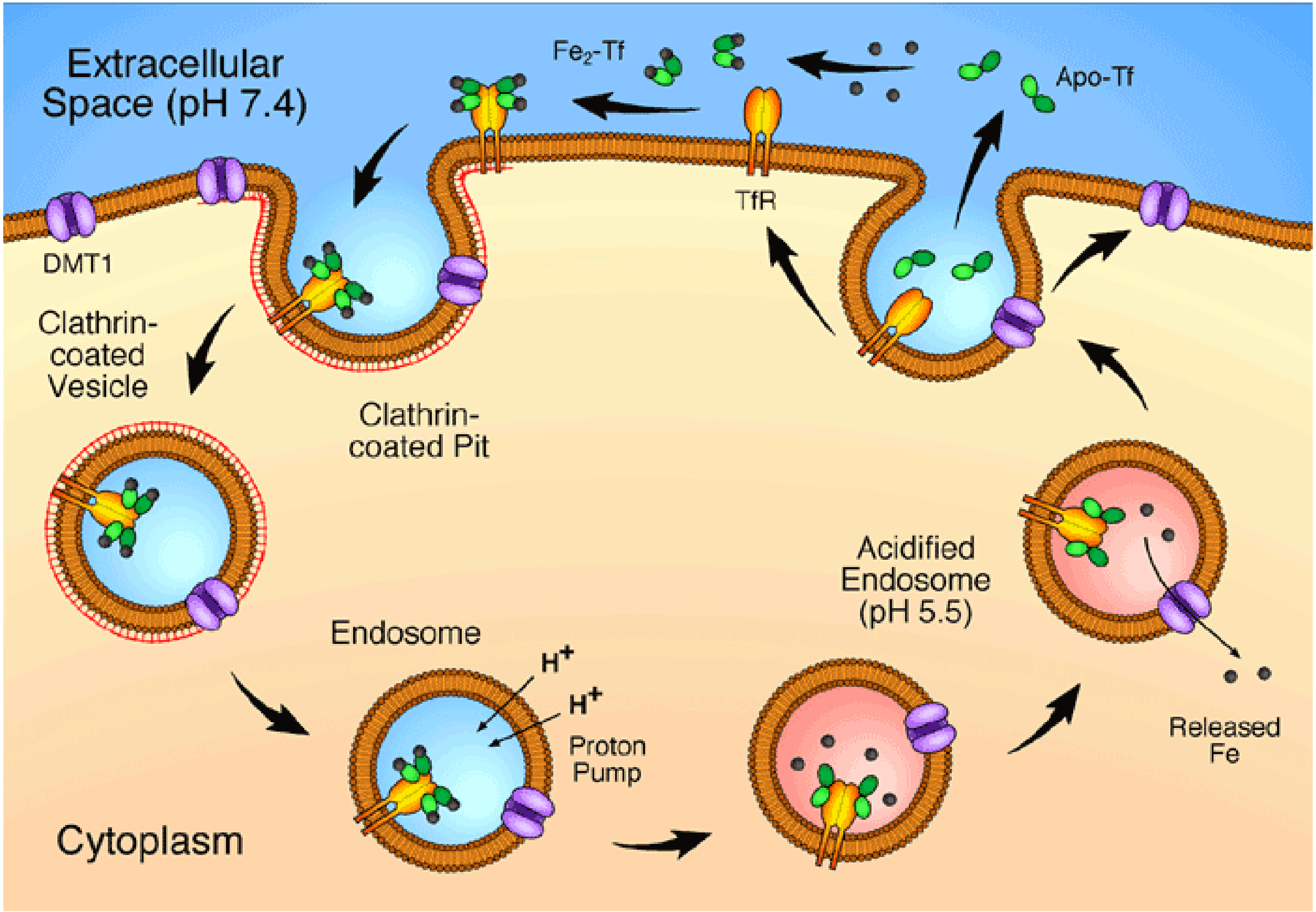
Fe(III) coordination site in transferrin



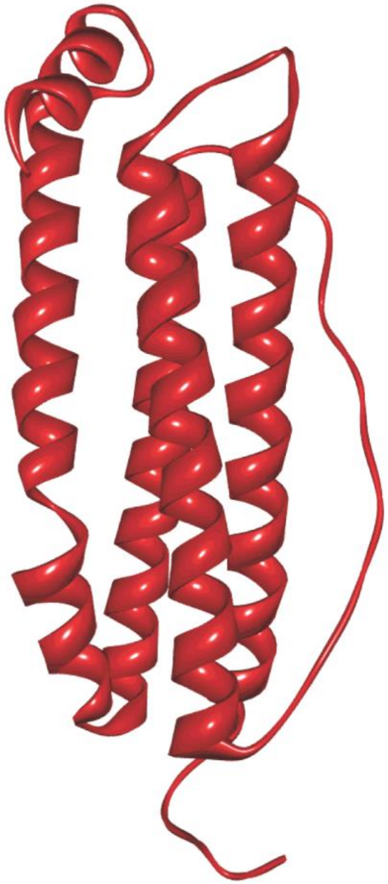
The coordination of Fe(III) involves the release of $3H^+$ and a remarkable **conformational change** of transferrin

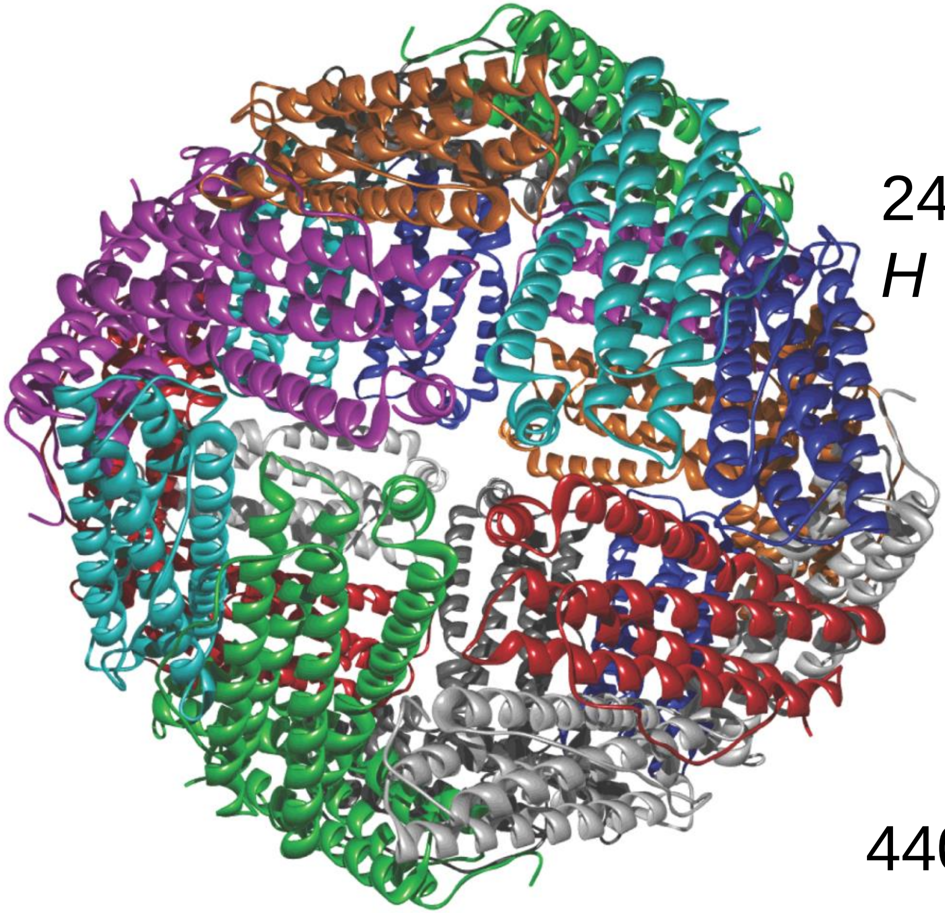
The transferrin cycle

endocytosis



Iron storage: Ferritin



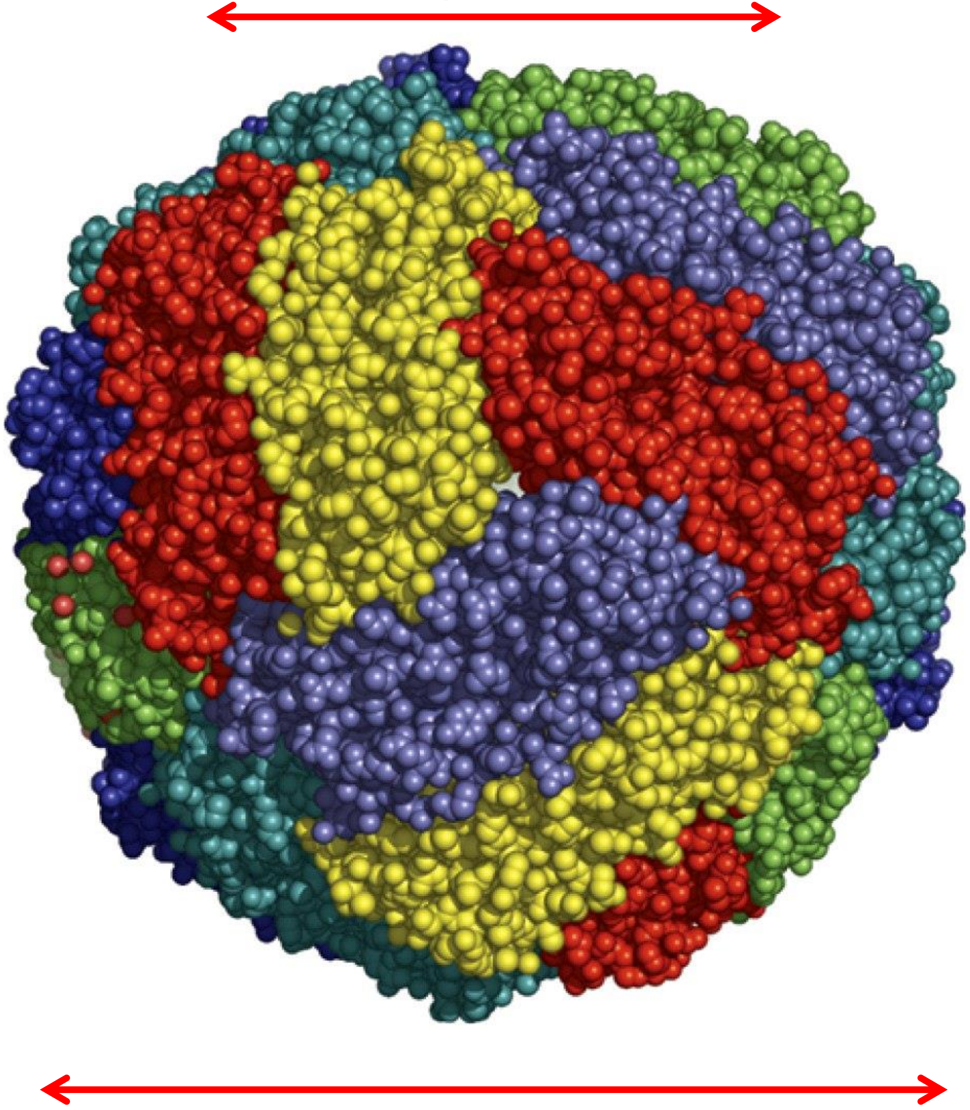


24 units,
H e *L*

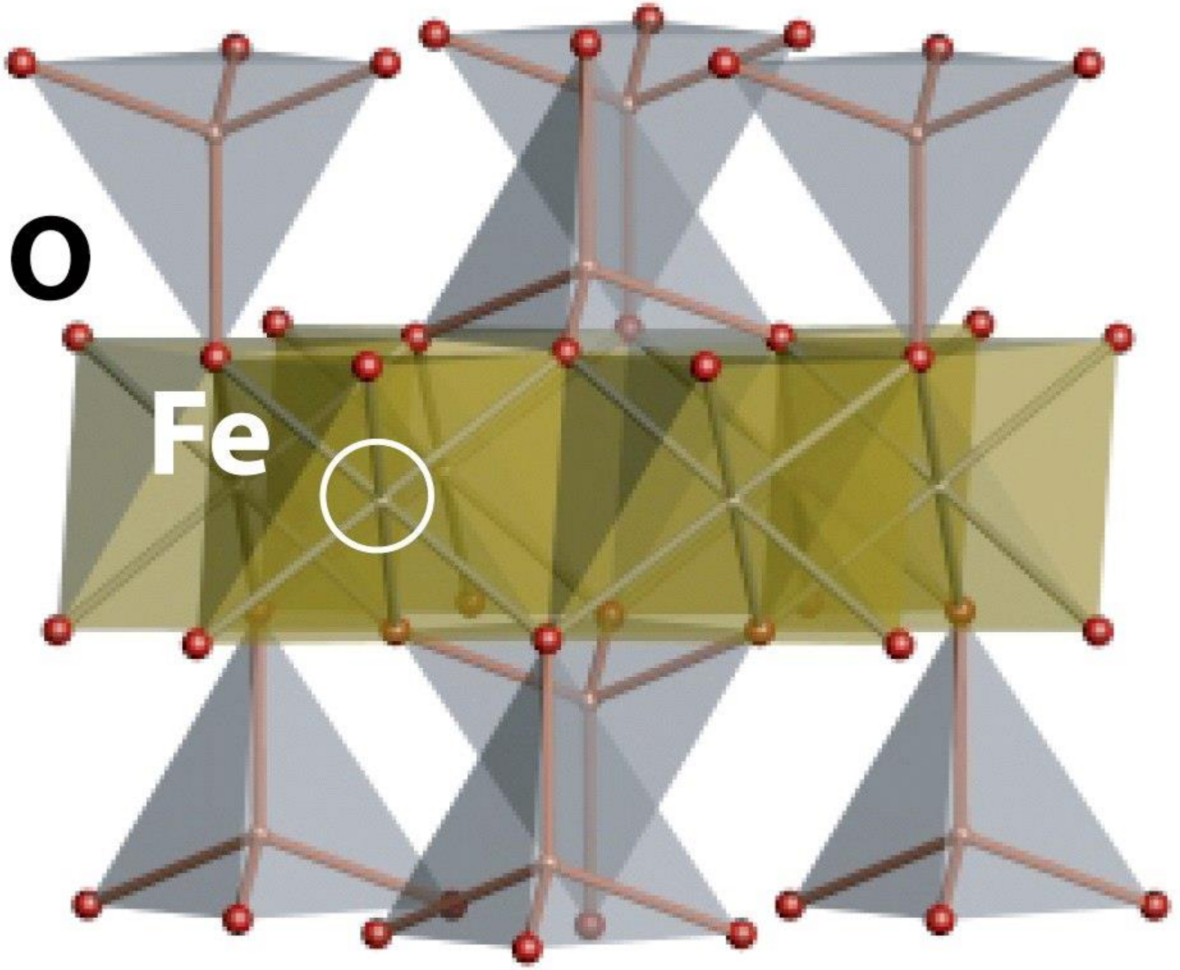
440 kDa

The *Heavy-chain ferritin* (*H*) has ferroxdasic activity
8 hydrophilic pores with ternary symmetry (Fe *in*)
6 hydrophobic pores with quaternary symmetry (Fe *out*)

In, 7.5nm



Out, 12 nm

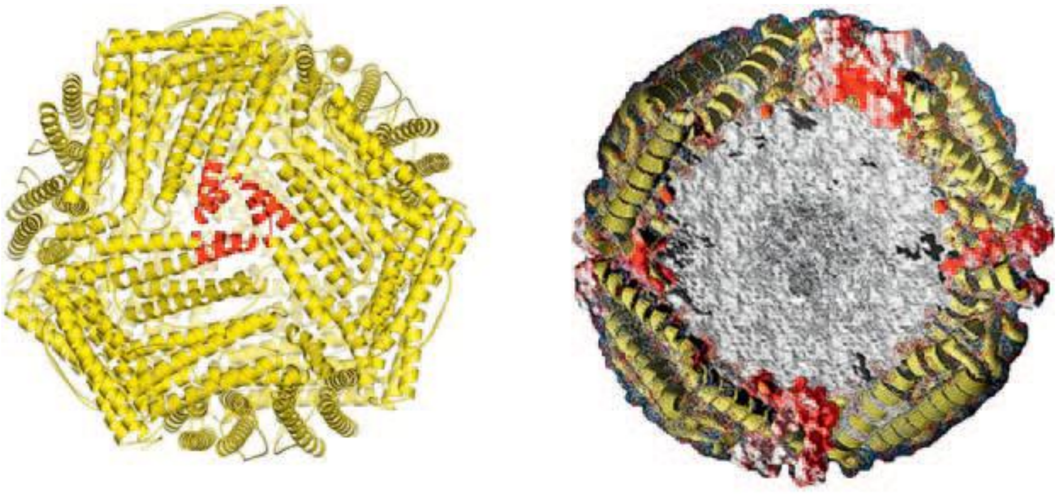


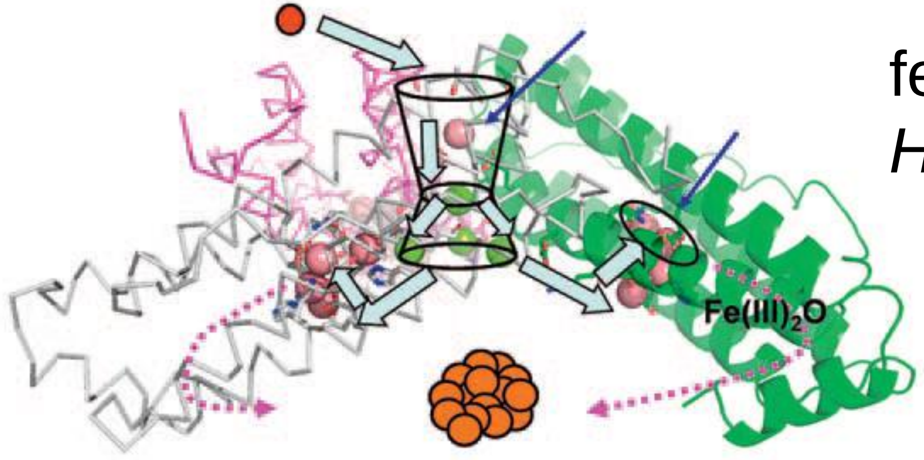
$Fe_9O_9(OH)_8(H_2PO_4)$

Ferrihydrite

$5Fe_2O_3{\cdot}9H_2O$

Up to 4500 Fe atoms Fe, 1200 on average



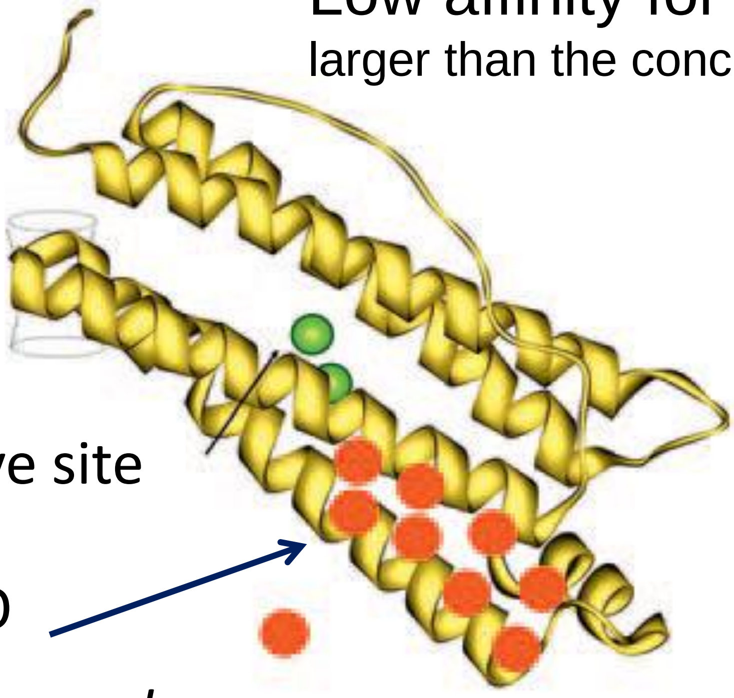


ferroxidasic site in
H-ferritin

ferroxidasic site

Low affinity for Fe(II), 15 μM

larger than the concentration of «free» Fe(II)



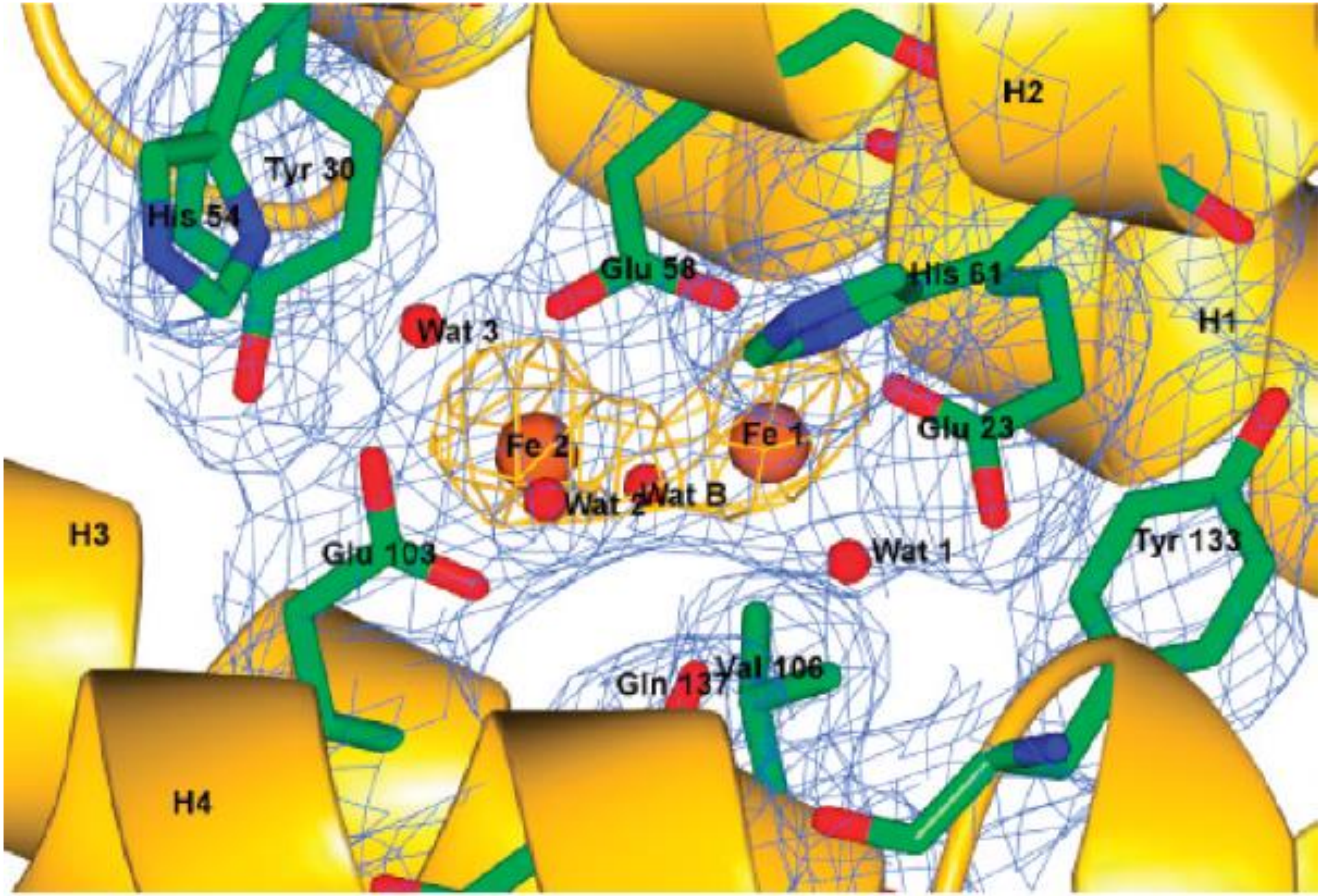
Active site

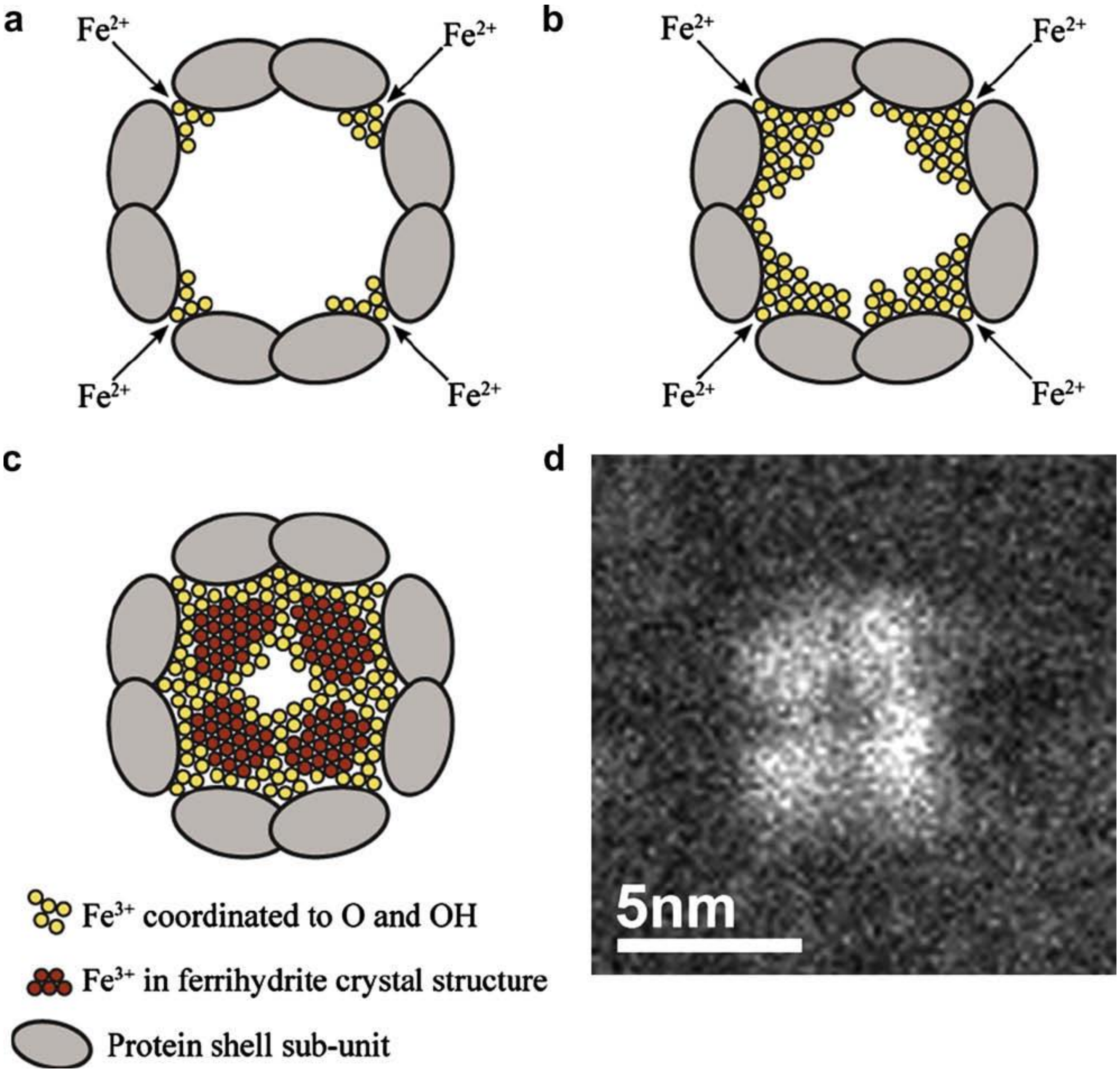
$Fe(III)_2O$

nucleation channel

Possibility for *chaperone* proteins for Fe(II) (e.g. PCPB1)

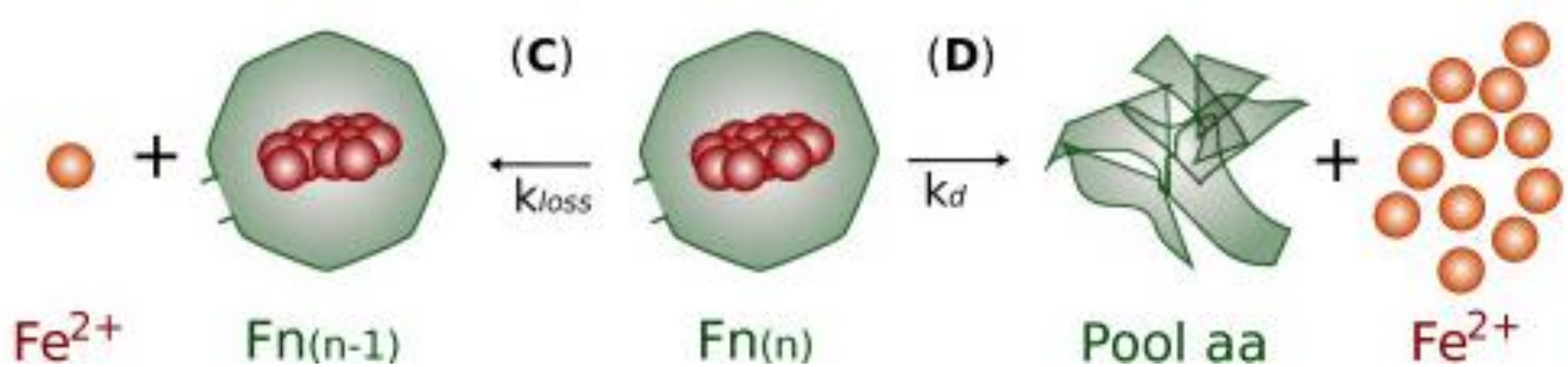
X ray structure of a ferroxidasic site



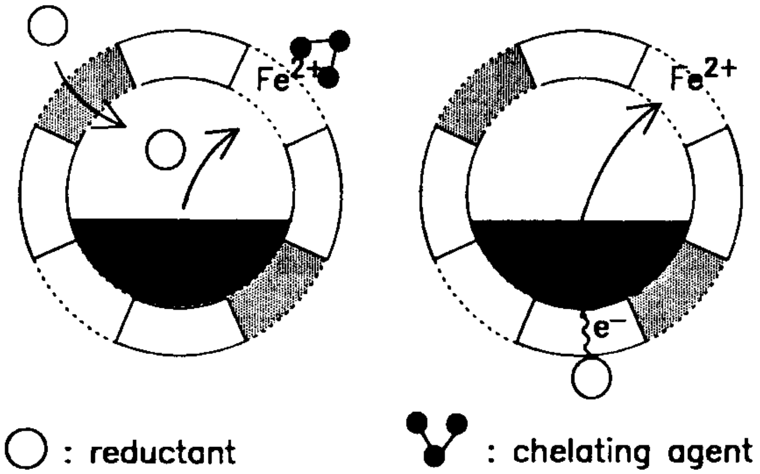

a
Fe2+
Fe2+
Fe2+
Fe2+
b
Fe2+
Fe2+
Fe2+
Fe2+
c
d
5nm
Fe3+ coordinated to O and OH
Fe3+ in ferrihydrite crystal structure
Protein shell sub-unit

TEM image

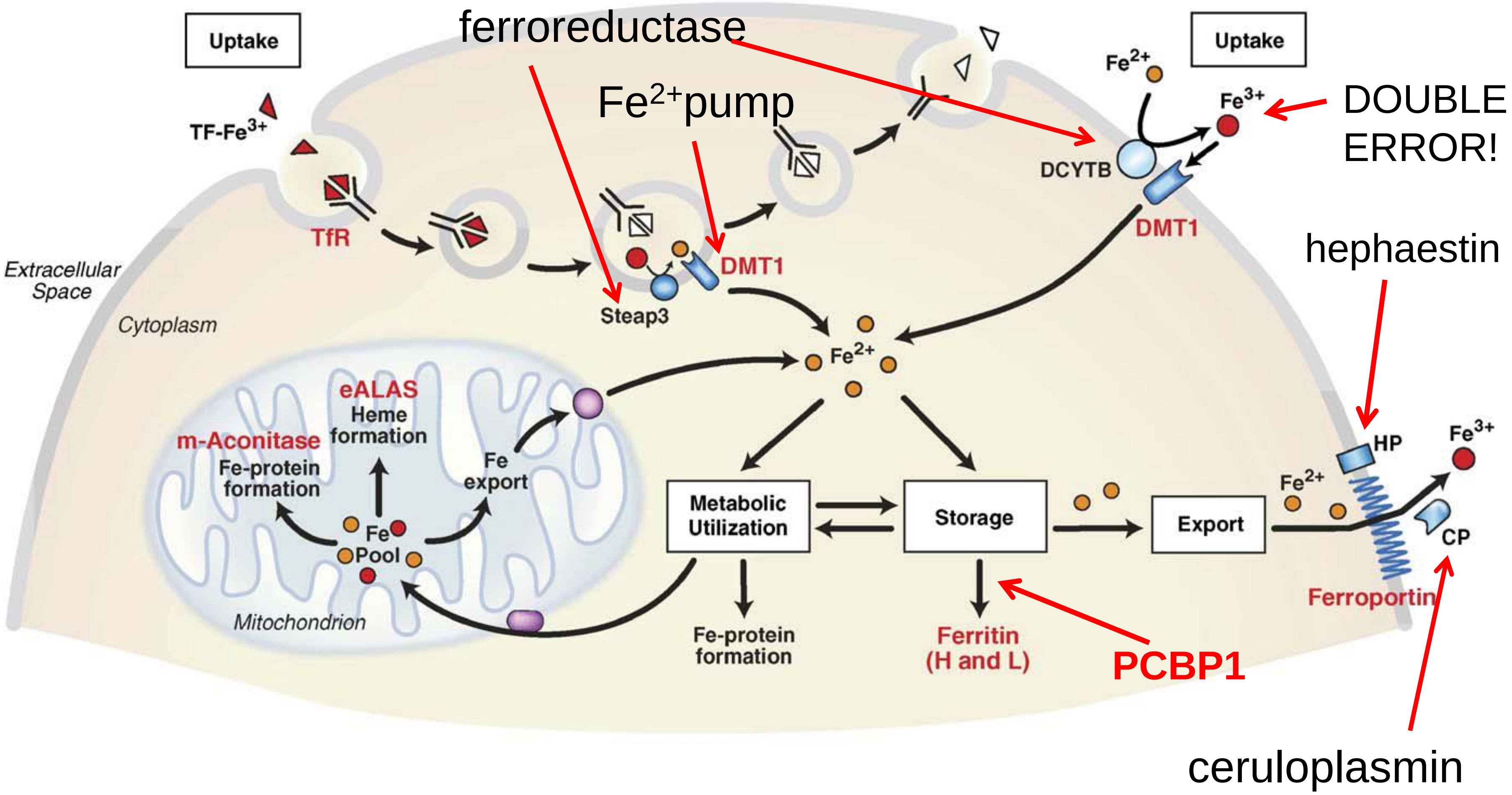
Iron release



protein degradation in lysosomes?



Iron homeostasis – IRP/IRE system

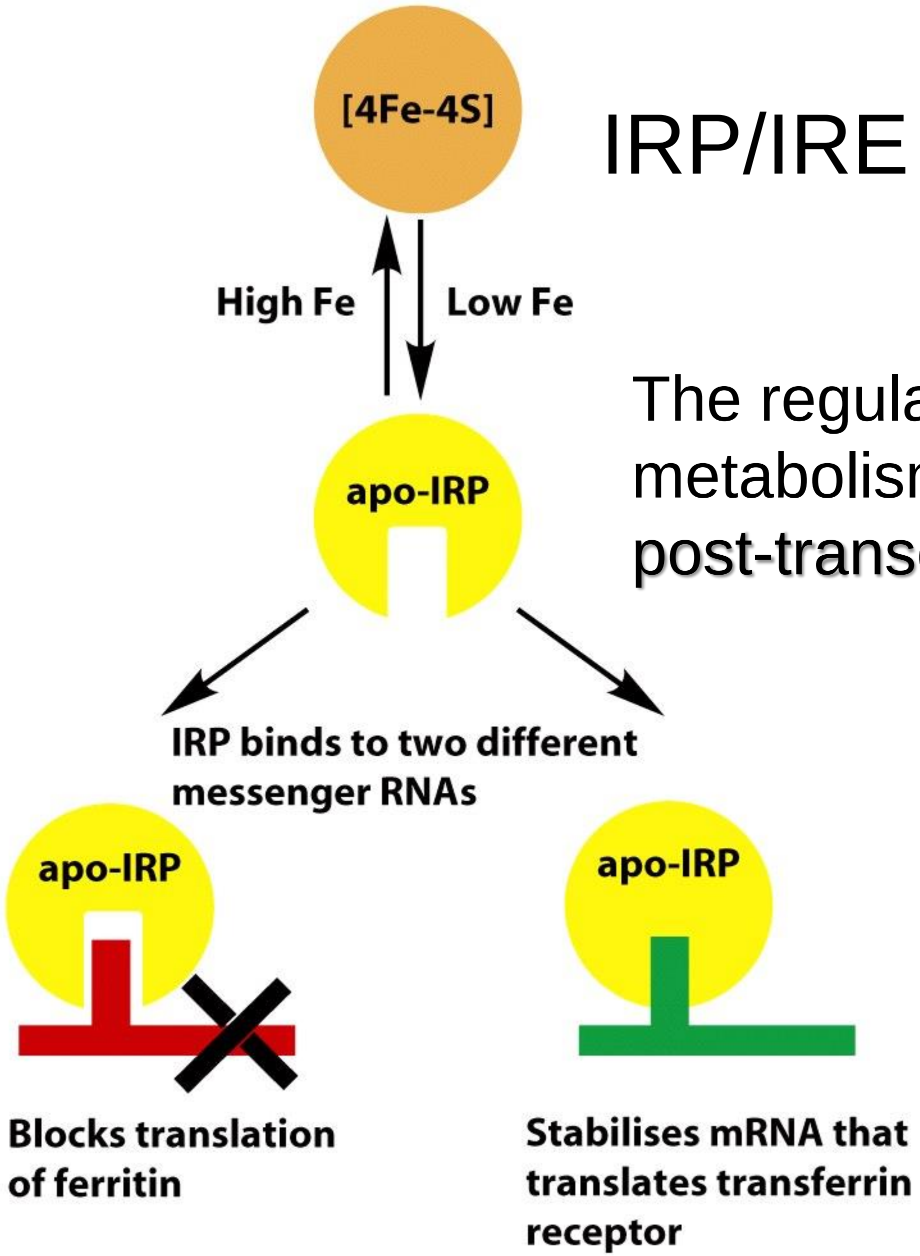


IRP: Iron Regulatory Proteins

IRE: Iron Responsive Elements (mRNA)

IRP/IRE system

The regulation of iron metabolism occurs at the post-transcriptional level



[4Fe-4S]

High Fe

Low Fe

apo-IRP

IRP binds to two different messenger RNAs

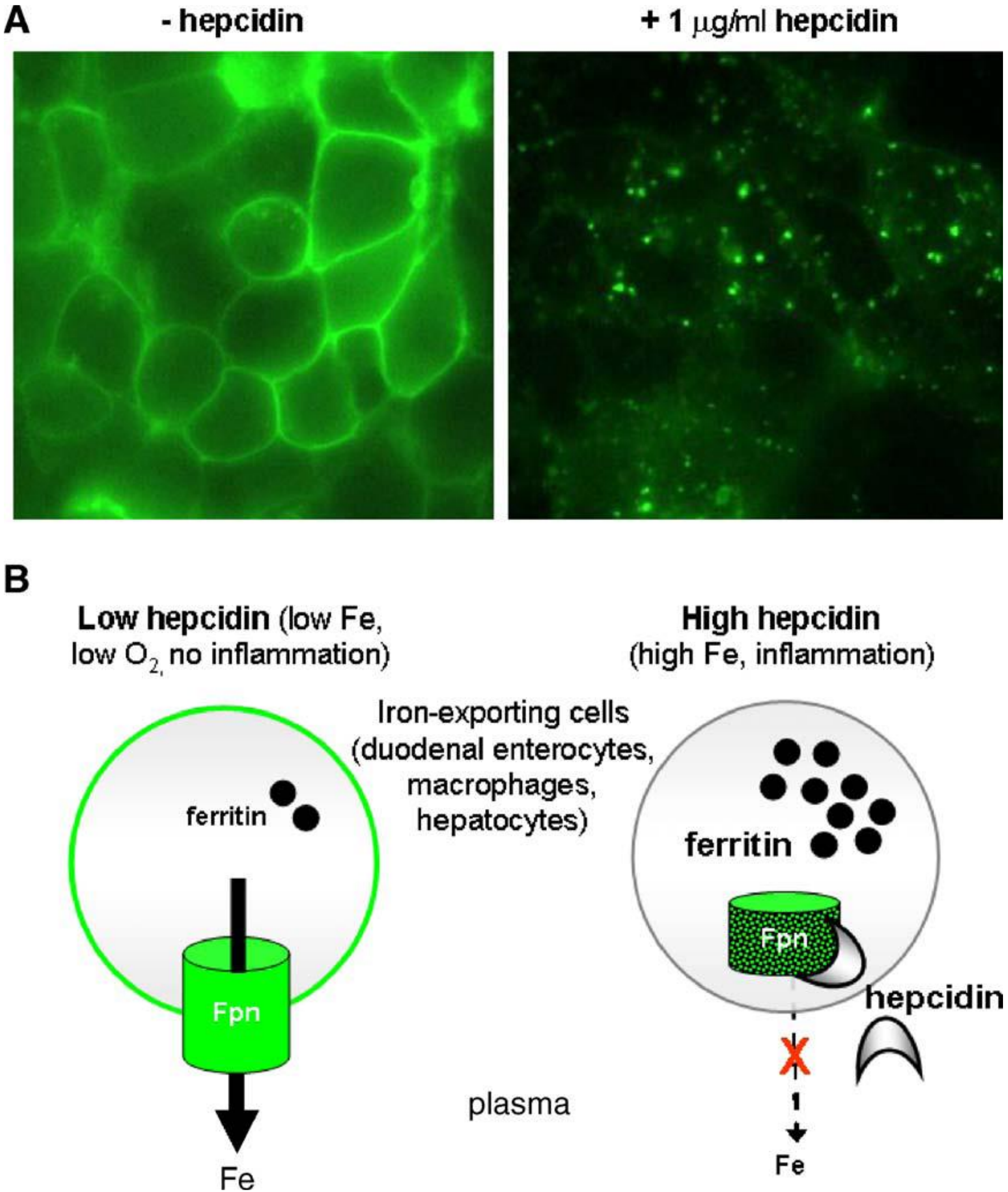
apo-IRP

apo-IRP

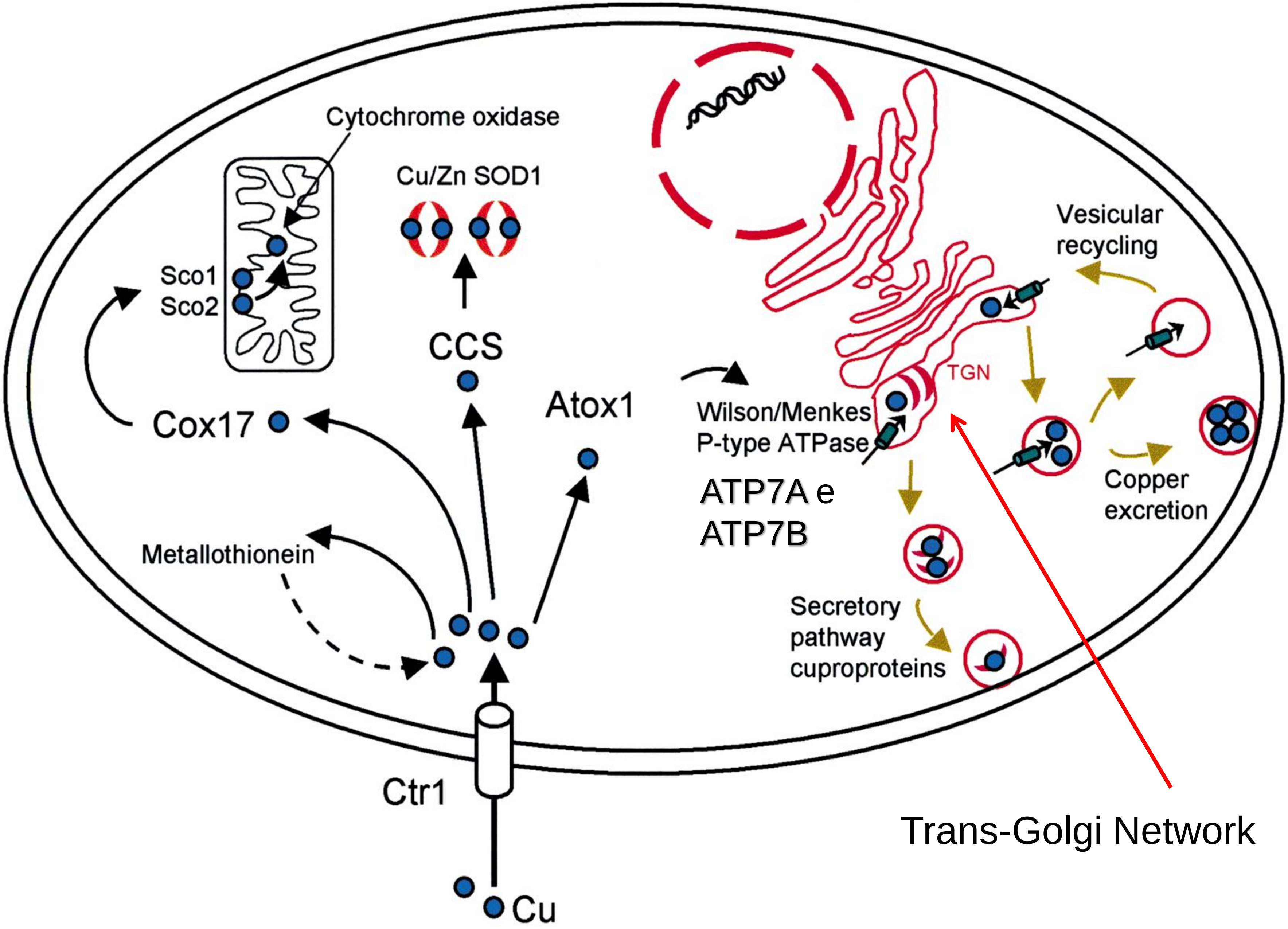
Blocks translation of ferritin

Stabilises mRNA that translates transferrin receptor

hepcidin-regulated iron export in plasma



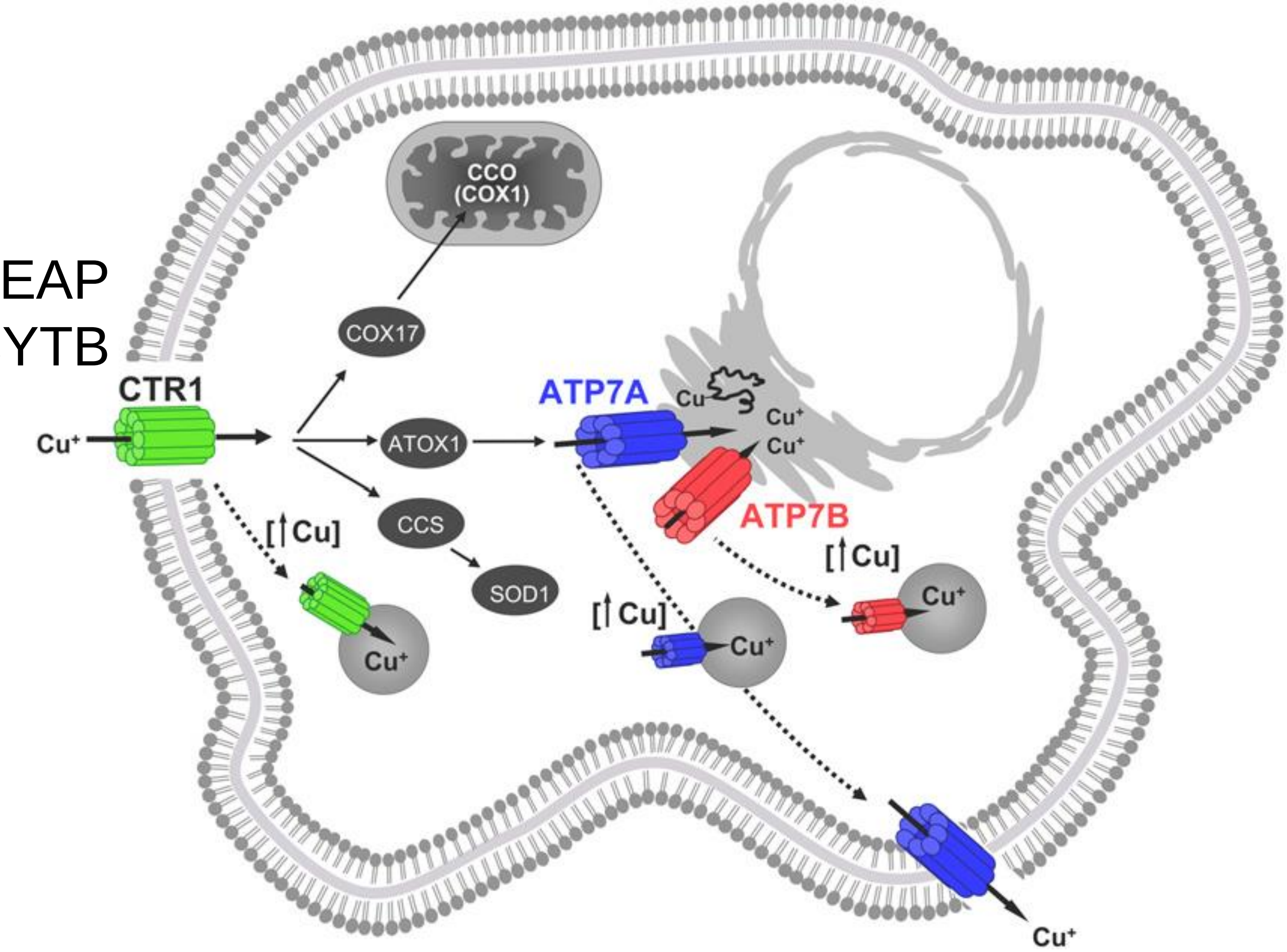
Copper homeostasis



ATP7A e
ATP7B

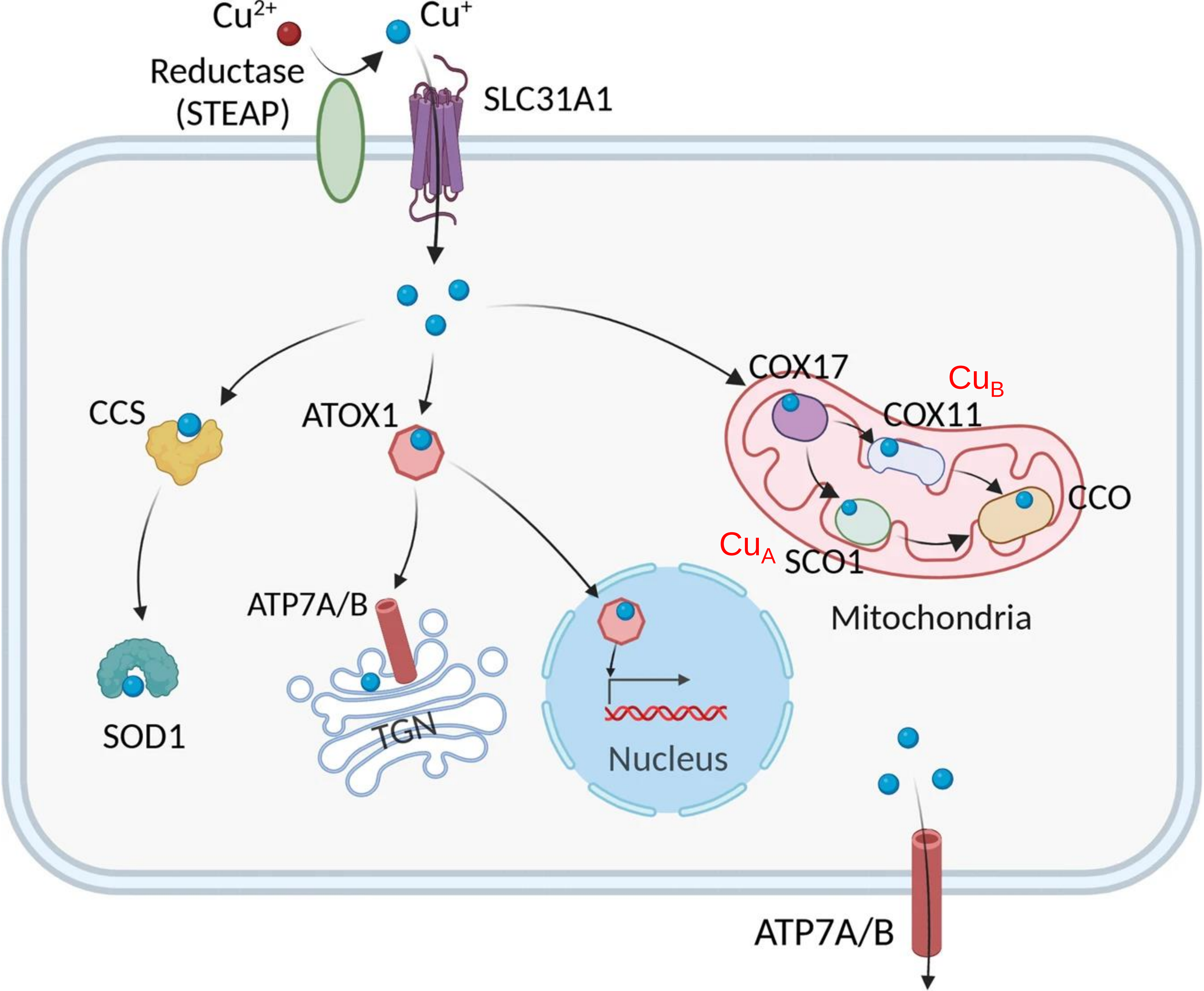
Trans-Golgi Network

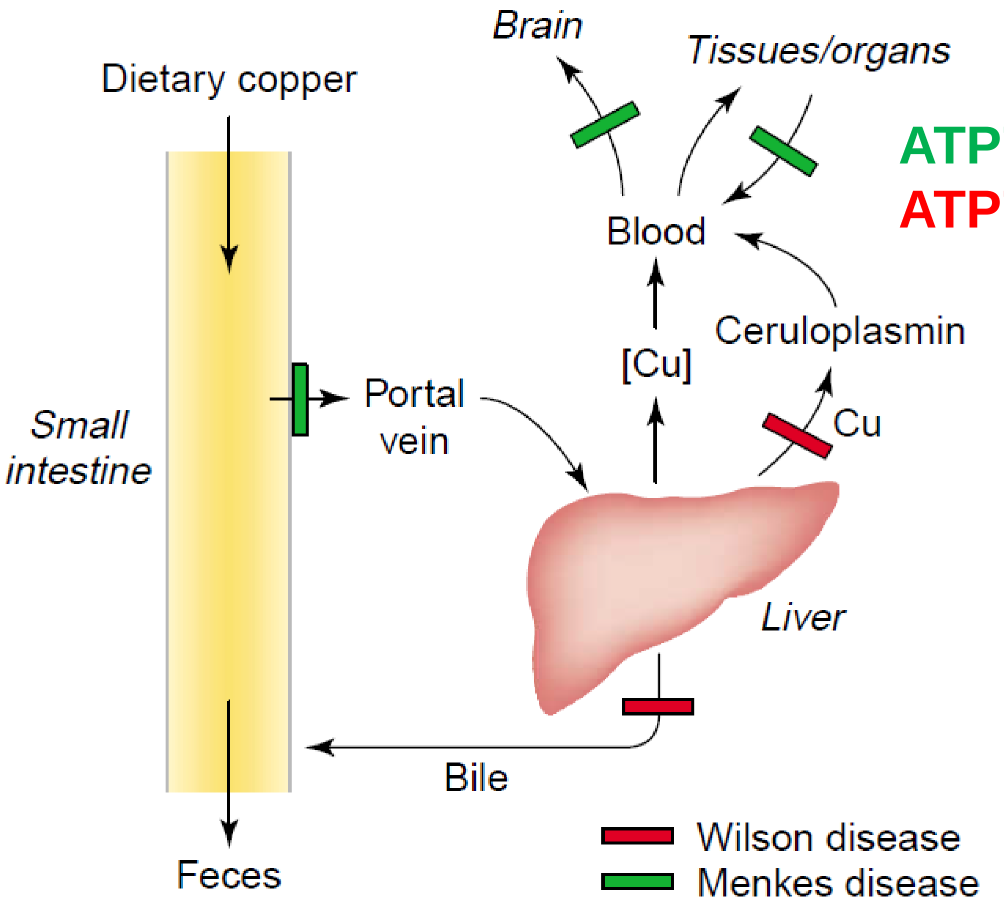
Copper homeostasis



STEAP
DCYTB

Cu^{2+}: albumin, ceruloplasmin, macroglobulins,…

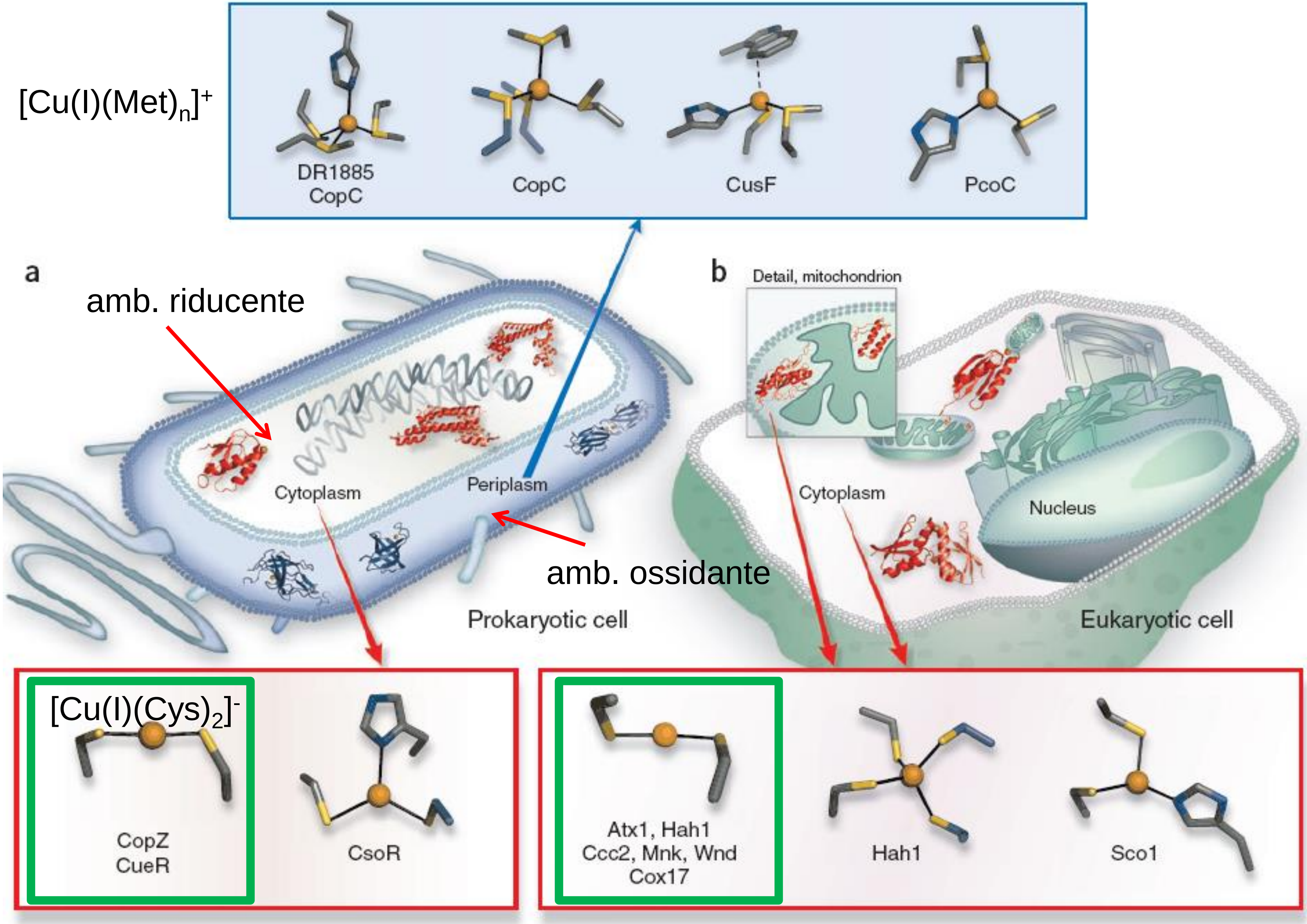

Cu^{2+}
Cu^{+}
Reductase (STEAP)
SLC31A1
CCS
ATOX1
COX17
Cu_B
COX11
CCO
Cu_A
SCO1
Mitochondria
SOD1
ATP7A/B
TGN
Nucleus
ATP7A/B



ATP7A: Menkes disease, Cu deficency
ATP7B: Wilson’s disease, Cu overload

ATP7A is expressed in most tissues/organs, with the exception of the liver.
ATP7B is found to be predominantly expressed in the liver

→ $Cu(II)(His)_2$



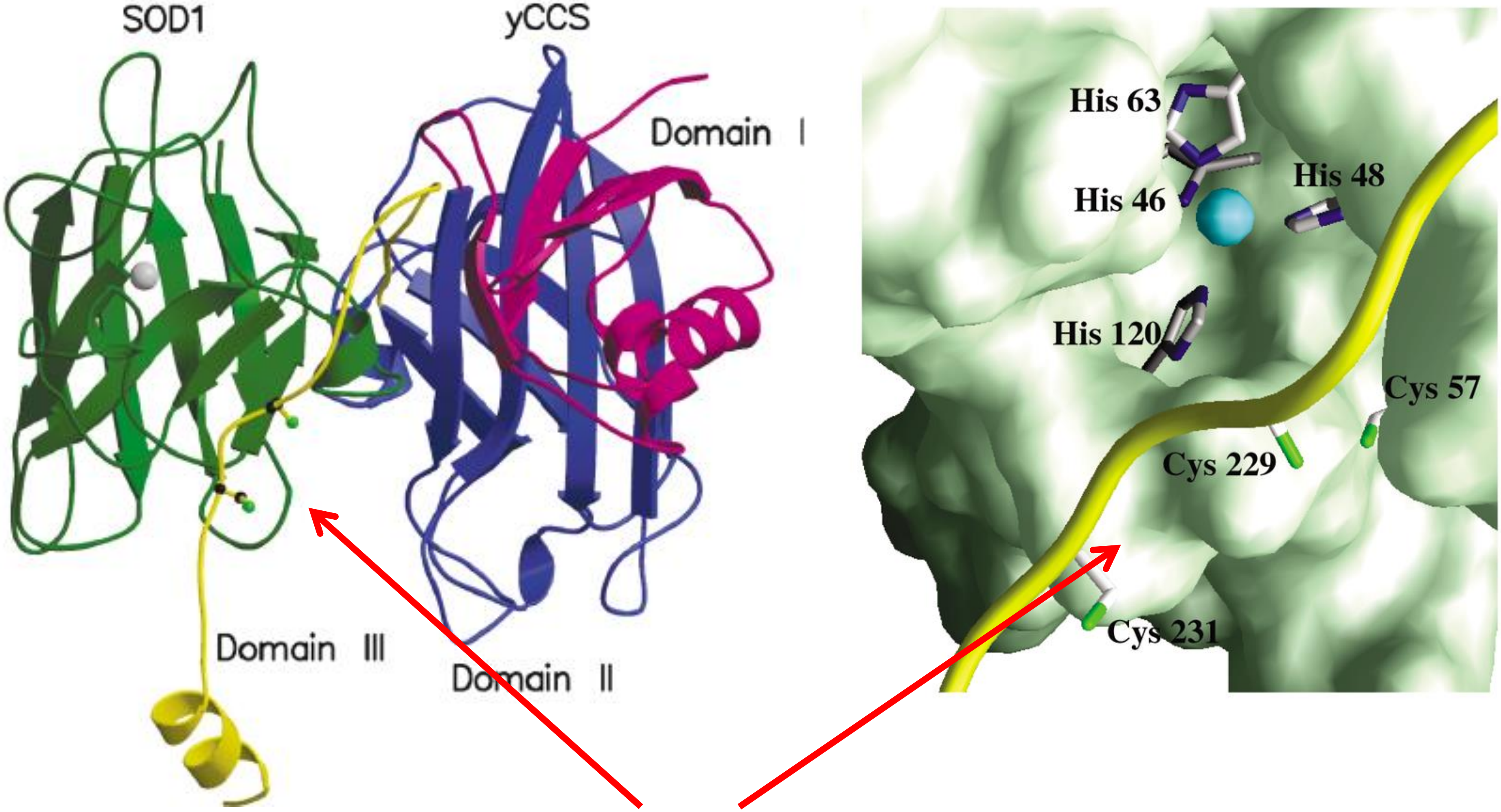
$[Cu(I)(Met)_n]^+$

amb. riducente

amb. ossidante

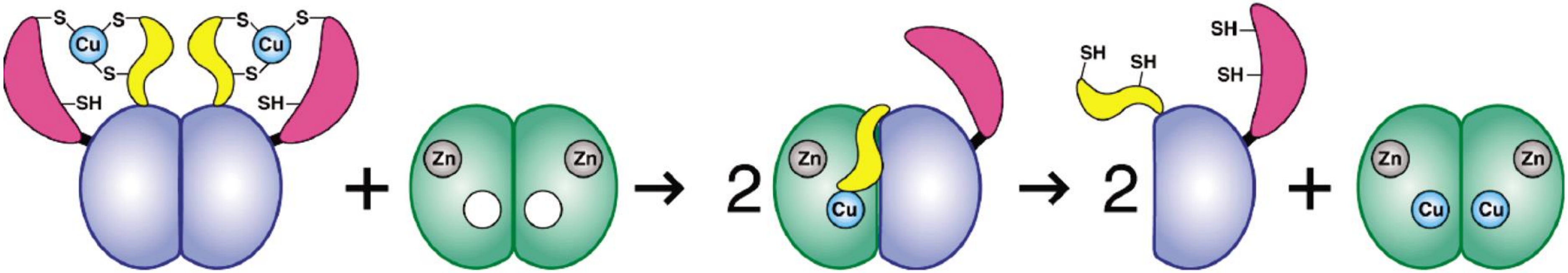
$[Cu(I)(Cys)_2]^-$

yCCS/Zn-SOD



Cu(I) binding site

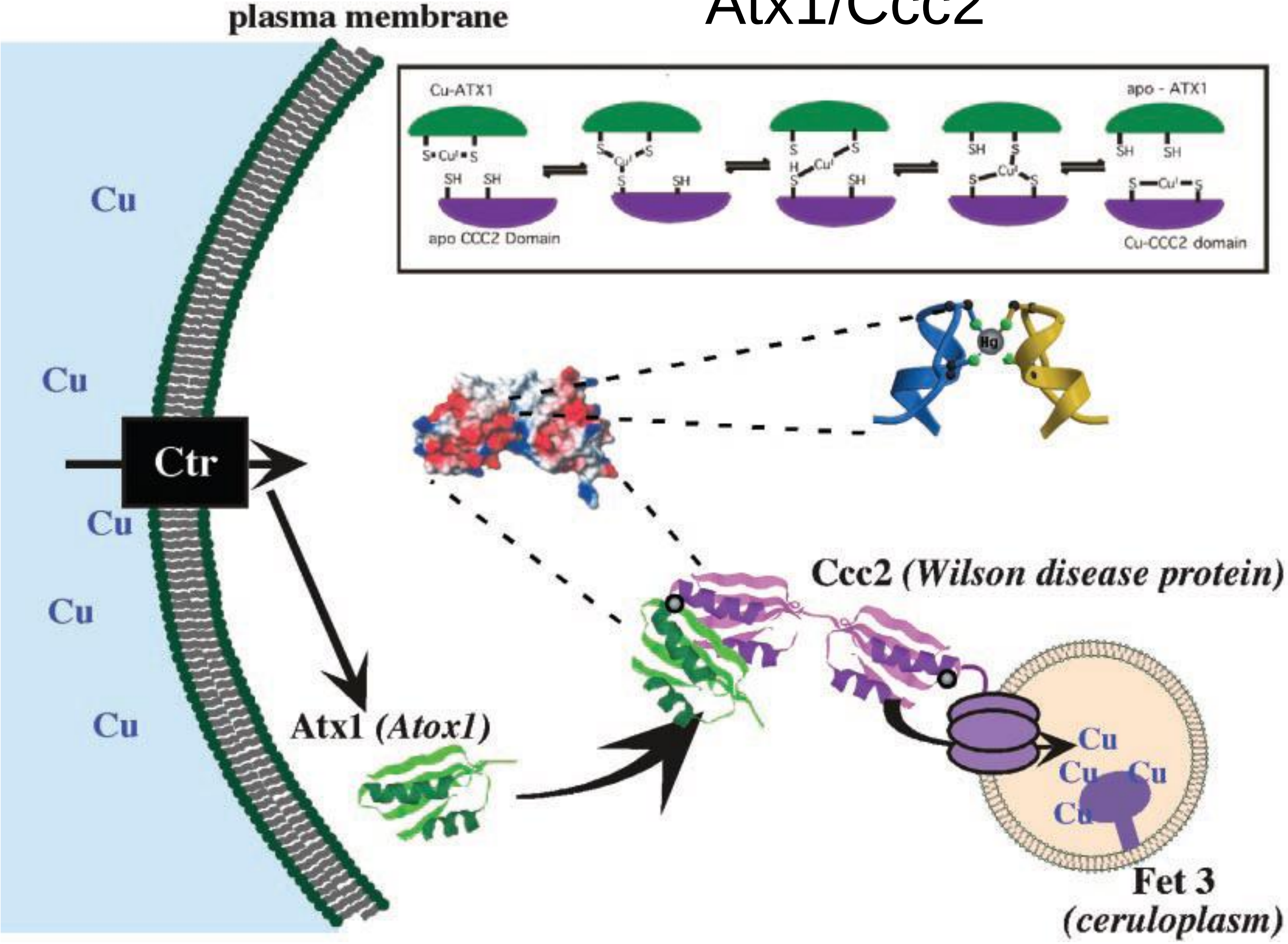
CCS/Zn-SOD



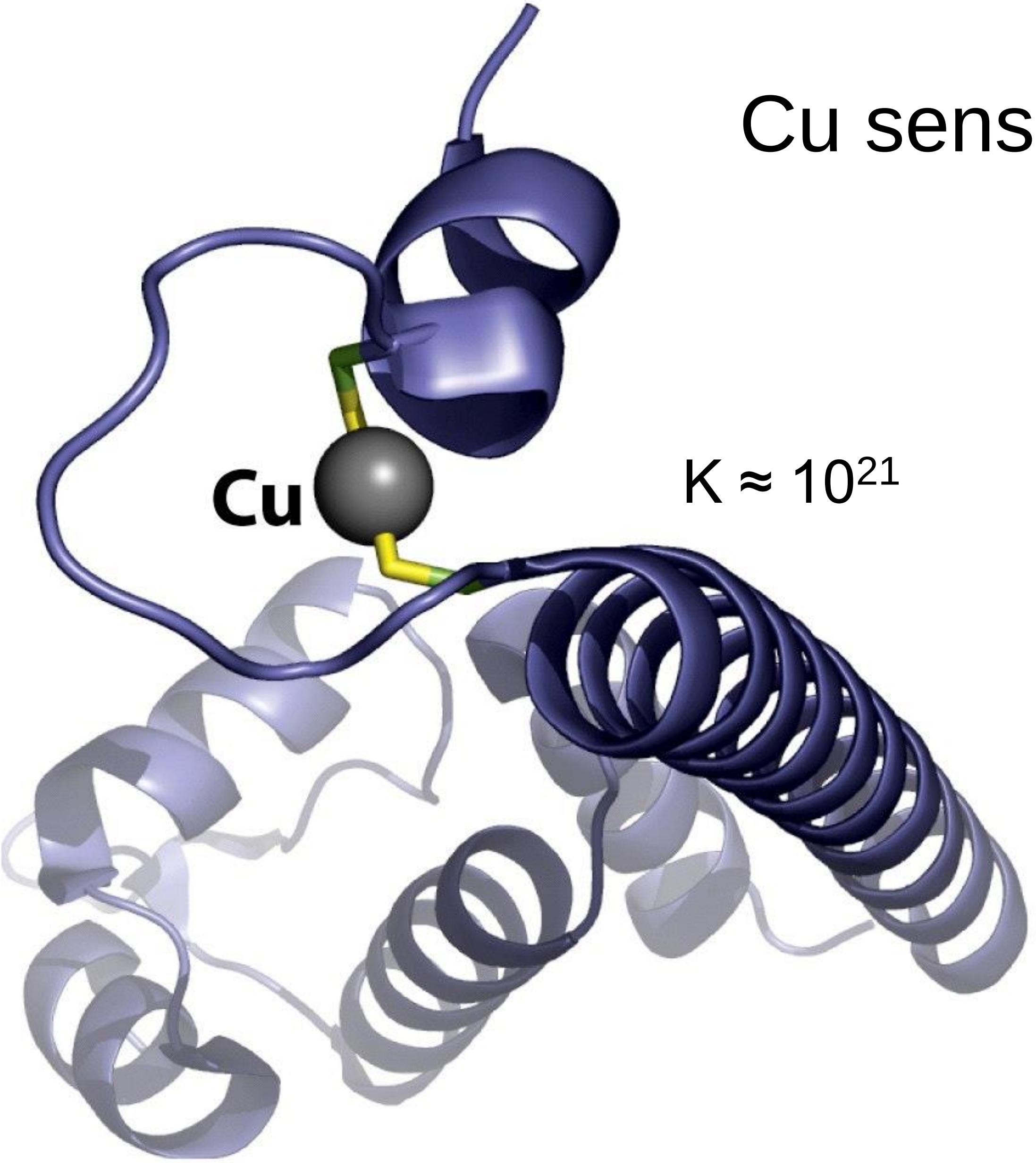
homodimers

heterodimer

Atx1/Ccc2


plasma membrane
Cu-ATX1
S Cu^I S
SH SH
apo CCC2 Domain
apo - ATX1
Cu-CCC2 domain
Hg
Cu
Cu
Ctr
Cu
Cu
Cu
Atx1 (Atox1)
Ccc2 (Wilson disease protein)
Cu
Cu
Cu
Cu
Fet 3
(ceruloplasm)

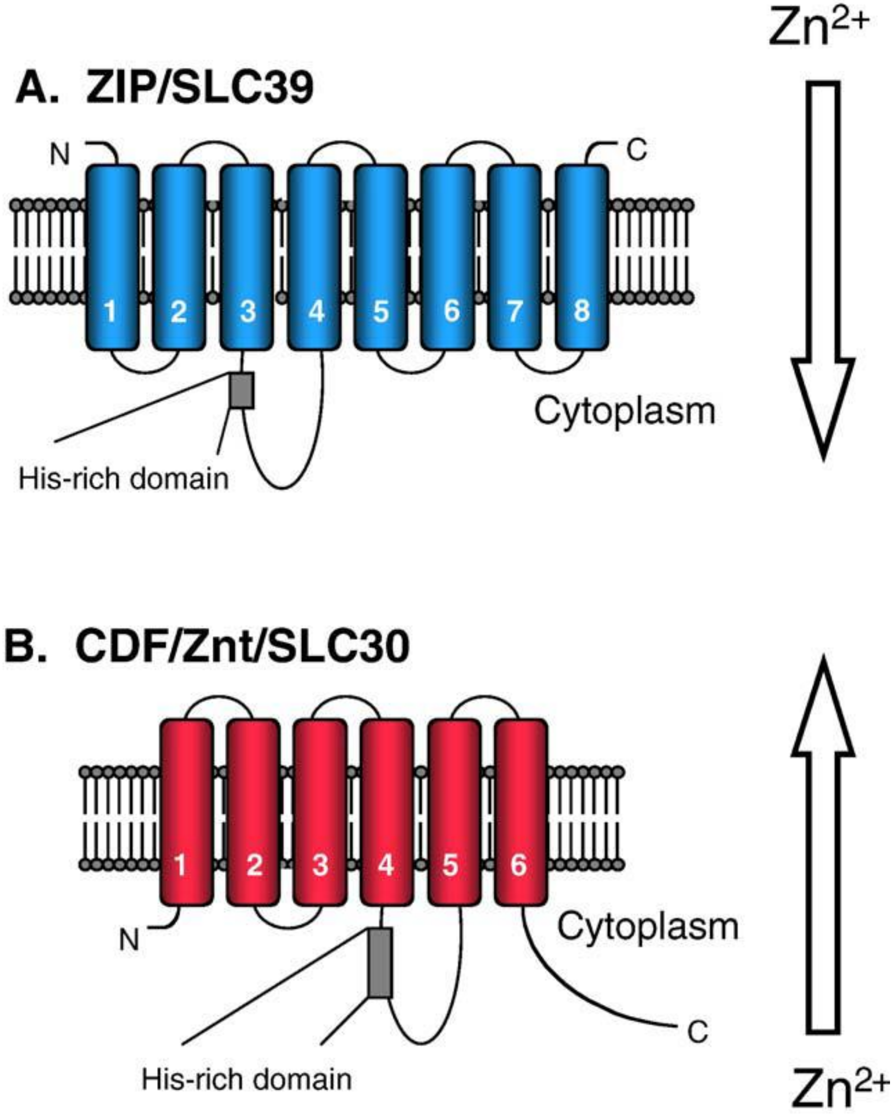
Cu sensors



K ≈ 10^{21}

CueR transcription factor (from *E. Coli*)

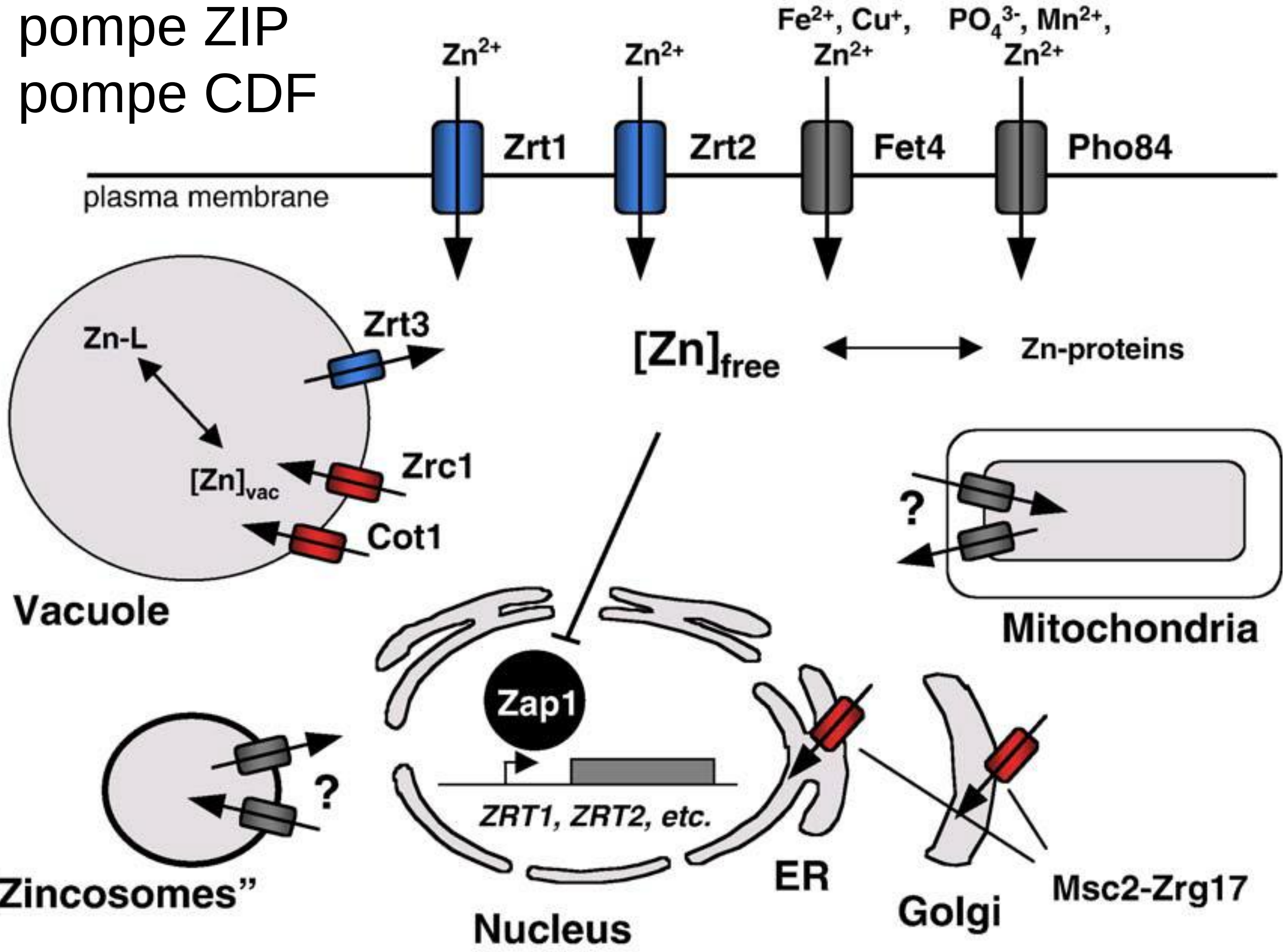
zinc homeostasis in eukariotic cells



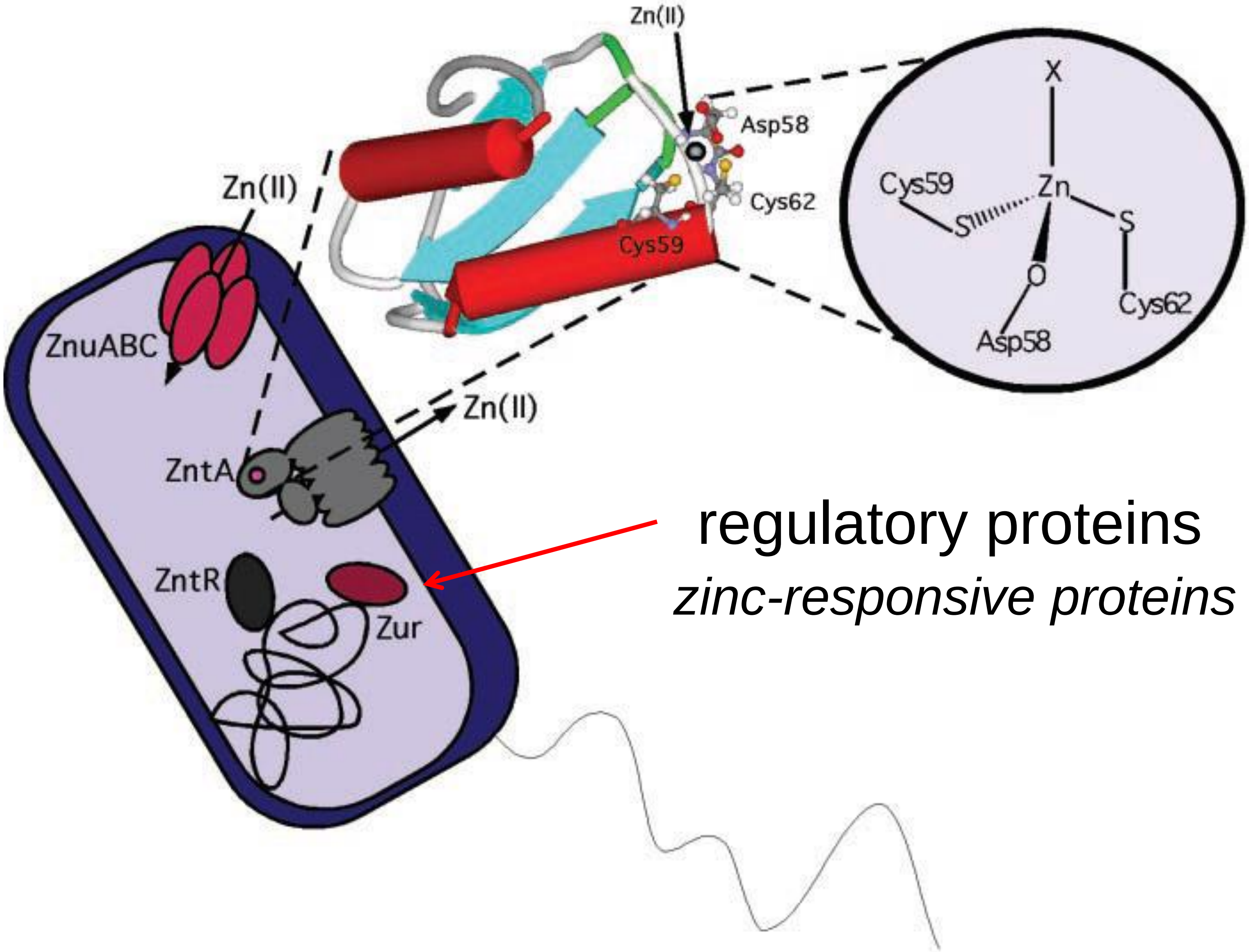
zinc homeostasis in eukariotic cells

🔵 = pompe ZIP

🔴 = pompe CDF



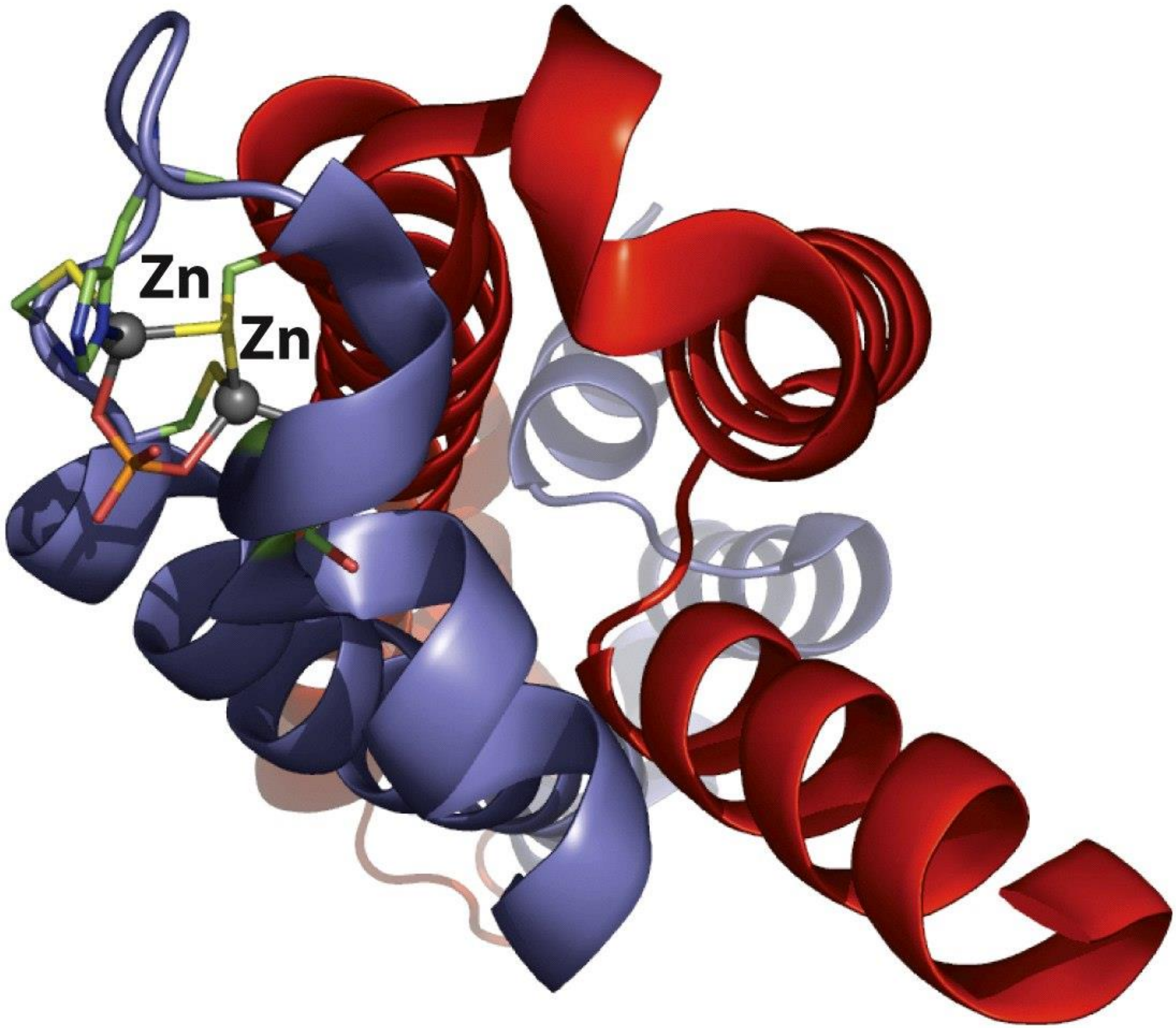
zinc homeostasis in bacteria



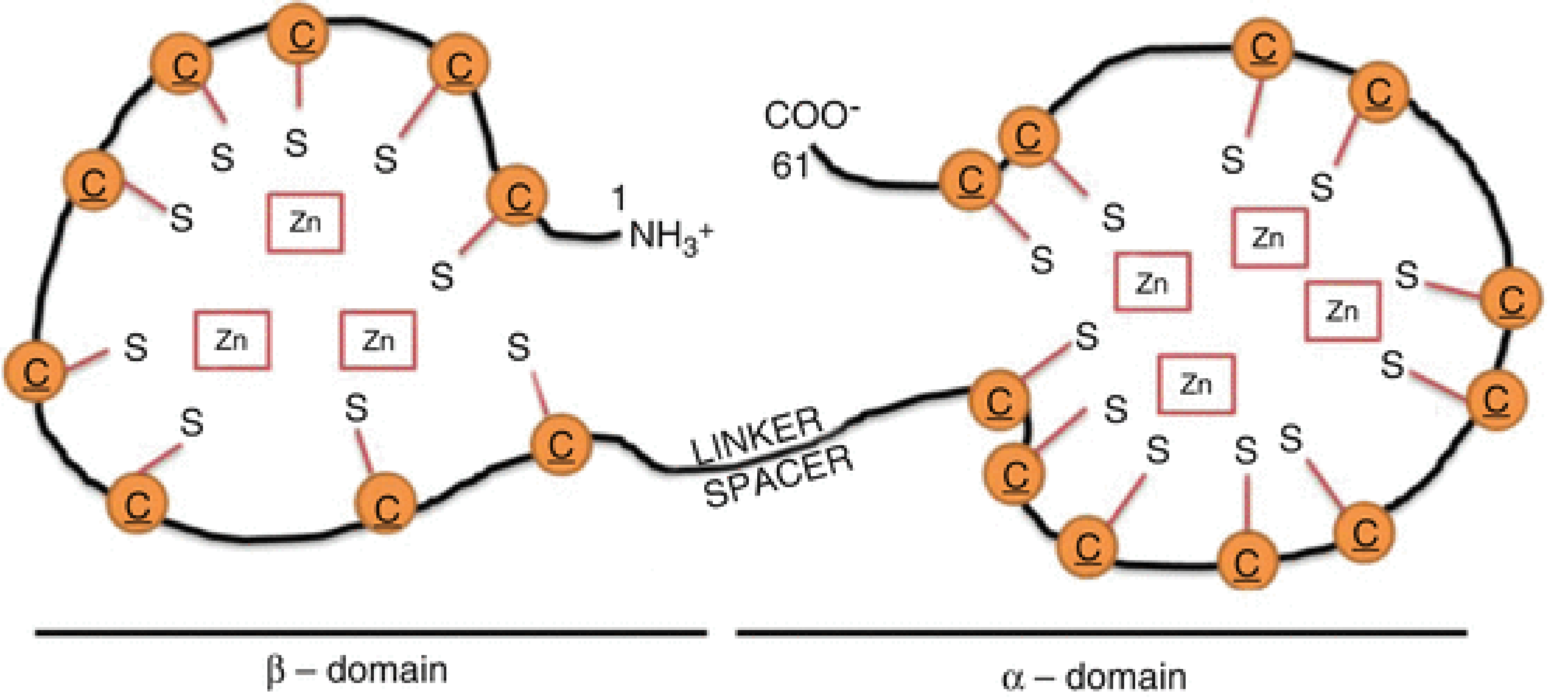
regulatory proteins

zinc-responsive proteins

ZntR transcription factor

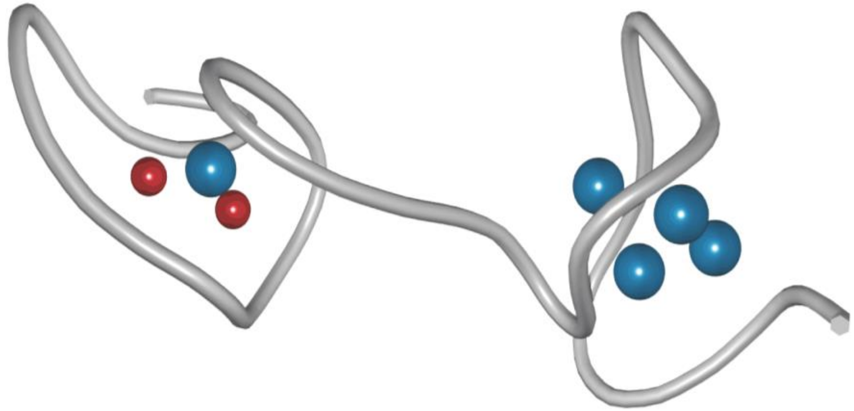


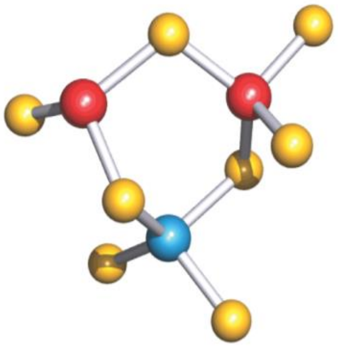
Metallothioneins



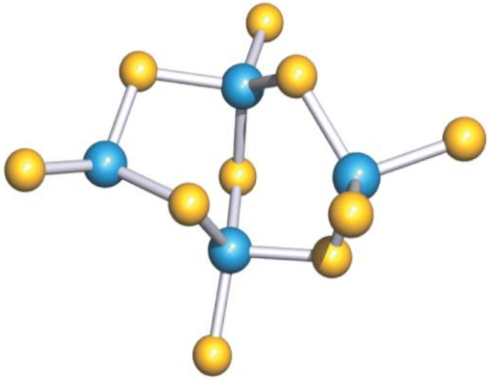
6 kDa,
ca. 60 a.a.
fino 30% cys

Metallothioneins





$CdZn_2S_8$



Cd_4S_{10}

multifunctional proteins (?):

- Detoxification/protection from xenobiotic metals (Cd, Hg)
- Temporary storage of Zn and Cu
- *Chaperones*
- Protective role towards ROS