Metabolism of iron

Prof. Mamoun Ahram
Hematopoietic-lymphatic system

Resources

- This lecture
- Yiannikourides and Latunde-Dada. A Short Review of Iron Metabolism and Pathophysiology of Iron Disorders. Medicines 2019, 6, 85. https://www.mdpi.com/2305-6320/6/3/85
- Lippincott's Biochemistry, 7th edition

- The Medical Biochemistry page, Iron and Copper Metabolism https://themedicalbiochemistrypage.org/iron-and-copper-homeostasis/
- Fleming and Ponka, Iron Overload in Human Disease, N Engl J Med 2012;366:348-59, https://www.nejm.org/doi/full/10.1056/nejmra1004967
- Brissot and Loréal, Iron metabolism and related genetic diseases: A cleared land, keeping mysteries, Journal of Hepatology 2016 vol. 64 j 505–515, https://www.sciencedirect.com/science/article/pii/S016882781500742 4?via%3Dihub

Importance of iron

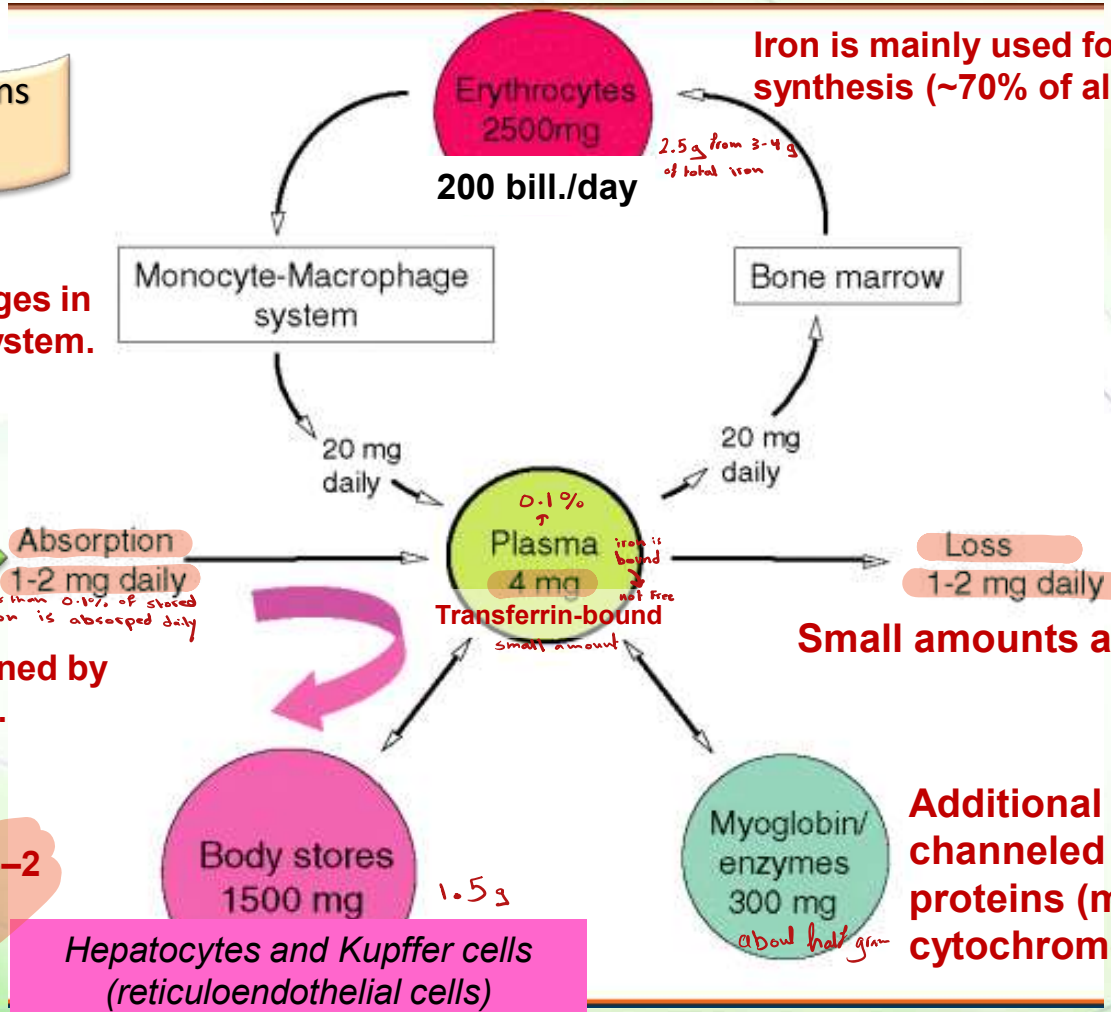
- Within the body, iron exists in two oxidation states: ferrous (Fe^{2+}) or, the highly insoluble, ferric (Fe^{3+}).
- It is also the prosthetic group of several enzymes such as redox cytochromes and the P450 class of detoxifying cytochromes.
  - and proteins → Hb
- Iron is important for metabolism and oxygen transport.
- Yet…
- Iron can be potentially toxic due its ability to form free radicals.
  - Solution: iron is not free.

* The body must balance the level of iron

What is life cycle of iron in the body?

Well-nourished persons
3-4g

Iron is mainly used for hemoglobin synthesis (~70% of all iron).



The released iron is scavenged by macrophages in the reticuloendothelial system.

Levels are maintained by dietary absorption.

Small amounts are lost

loss = absorption
level of iron is sort of constant
tight regulation of iron inside our system

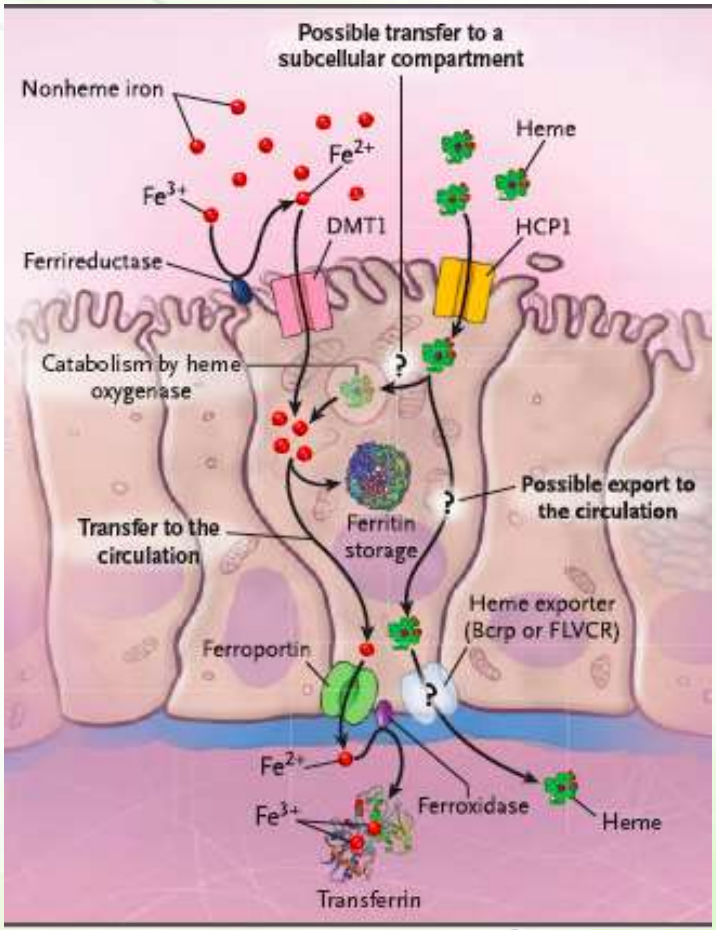
1 mg a day for men and 1.5–2 mg a day for women with regular menstrual periods

Additional iron (300 mg) is channeled to other cellular proteins (myoglobin and cytochromes).

Iron absorption

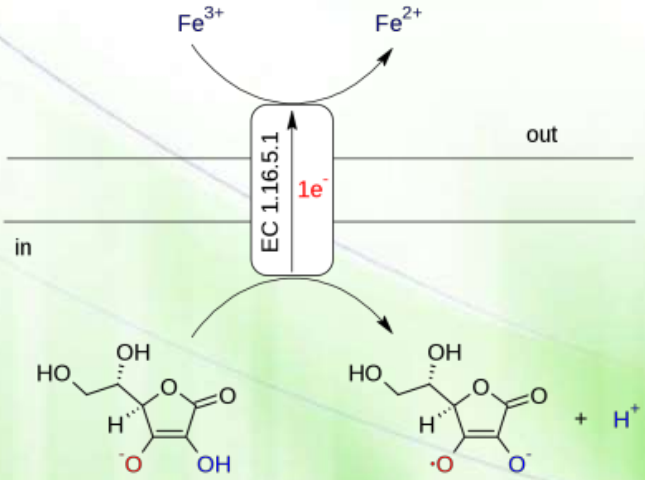
State of iron

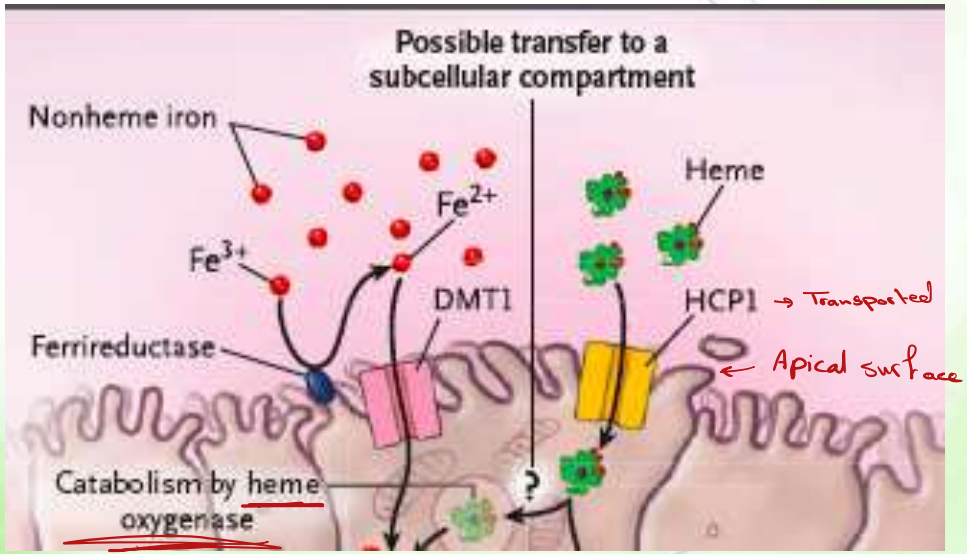
- Under conditions of neutral or alkaline pH, iron is found in the ferric Fe^{3+} state and, at acidic pH, in the ferrous Fe^{2+} state.
  - In the stomach, iron will be in the ferrous state. Fe^{+2} reduced
  - In the duodenum, iron is in the ferric state. Fe^{+3} oxidized (Alkaline conditions)
- However, to be absorbed, dietary iron must be in its ferrous Fe^{2+} form.



Site of absorption

- Ferrireductase enzyme on the enterocytes' brush border reduces Fe^{3+} to Fe^{2+} in a vitamin C-dependent reaction.
- Divalent metal transporter 1 (DMT1) transports iron into the cell.
  - DMT-1 can transport other metal ions such as zinc, copper, cobalt, manganese, cadmium, and lead.





Heme as a source of iron

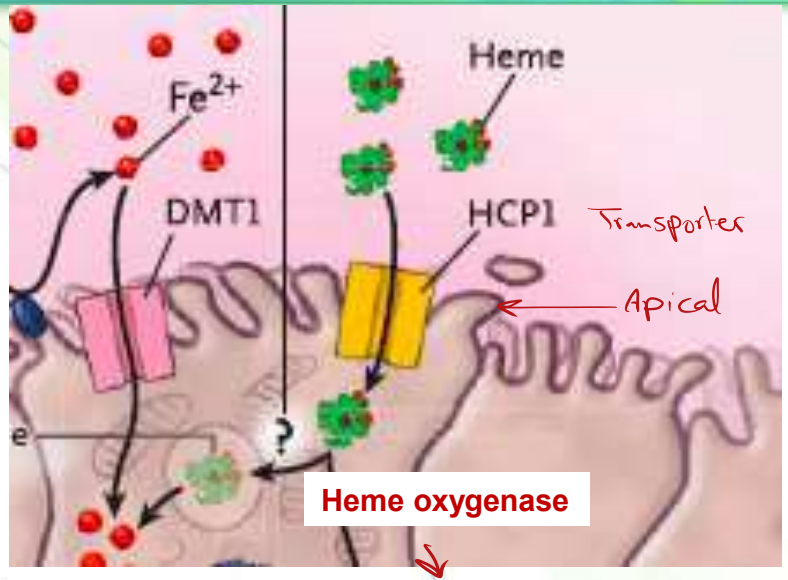
① Nutrition

- Iron can also be obtained from ② ingested heme.
- Heme is absorbed by a receptor called heme-carrier protein 1 (HCP-1) and iron is released by heme oxygenase-1 (HO-1).
- In other cells such as macrophages, heme oxygenase also extracts iron from heme.

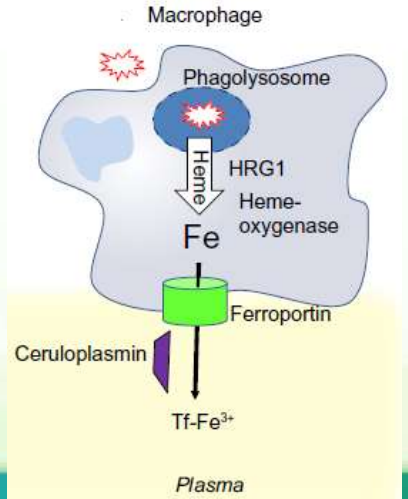
① phagocytosis of RBCs
② Release of heme
③ heme oxygenase extracts the iron
④ storing the heme

Proton-pump inhibiting drugs such as omeprazole greatly reduce iron absorption.

Reduce the stomach acidity then affects iron absorption
↓
changing the oxidation state of iron

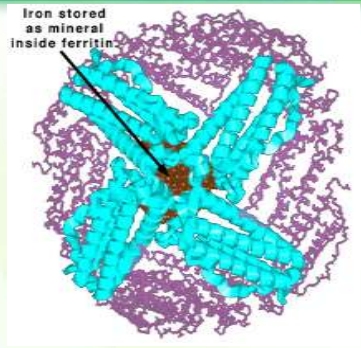


extract the iron from the heme



Fates of iron

After the absorption of iron by enterocytes

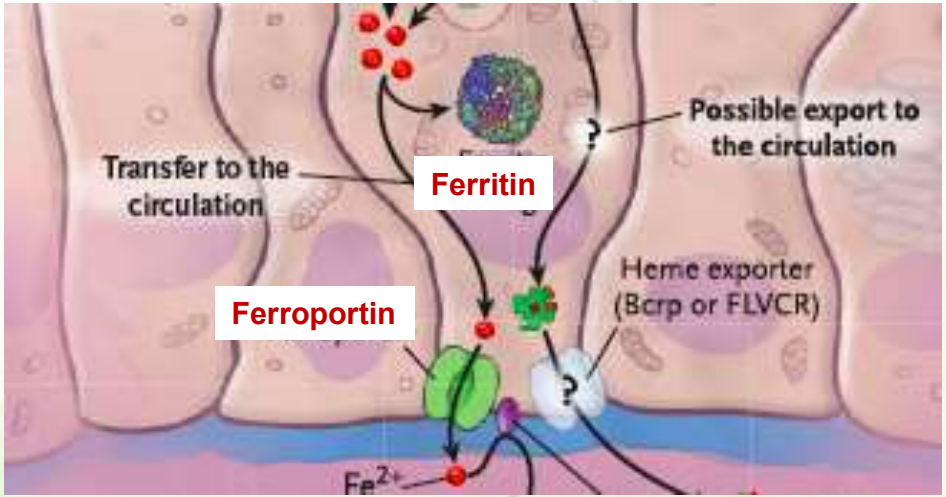


Fate 1: storage

- Cells can then store iron as ferritin. → the intestinal cells will conjugate it with inside the intestinal cells
  - Each Ferritin complex can store about 4500 iron (Fe^{3+}) ions.
- But, if cells are sloughed off from the tip of the villus into feces before absorption, iron is eliminated from the body. → intestinal cells die

Fate 2: Transport

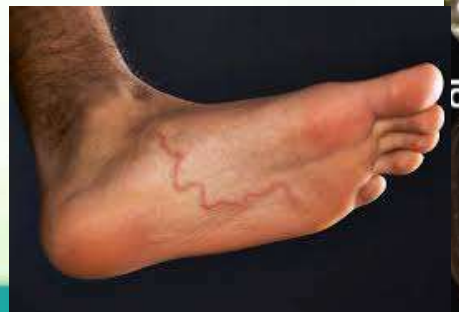
- Iron is transported out via a basolateral transporter known as ferroportin, which is distributed throughout the body on all cells. ↑ of the enterocytes

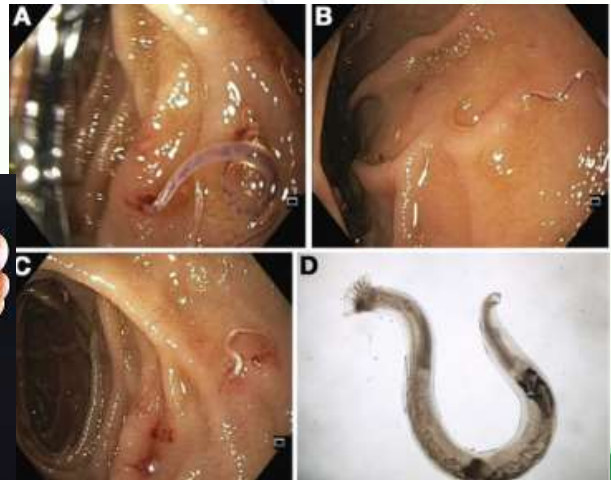


Intestine-related iron metabolism disorders

May Cause ← Anemia (IDA)

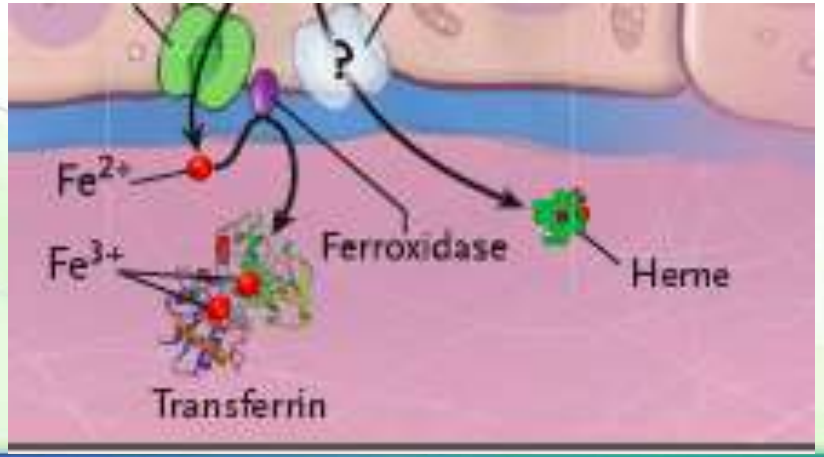
- Iron malabsorption — Inability of enterocytes to absorp iron
  - Gastrectomy (total or partial)
  - Celiac disease (villous atrophy)
  - Crohn's disease
  - Helicobacter pylori
- Intestinal hemorrhage (gastrointestinal-mediated iron loss)
  - Gastric cancer
  - Ulcers
  - Inflammatory bowl disease
  - Hookworm infection

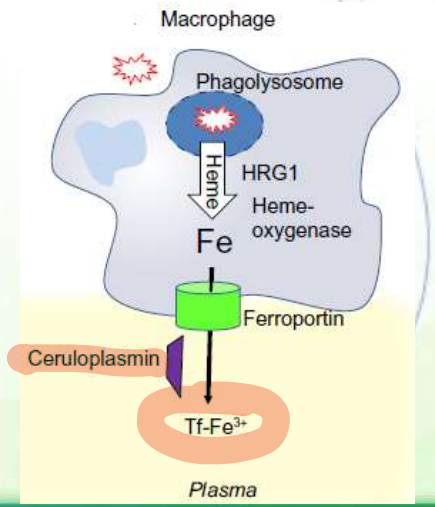




Ferroxidase and transferrin

- Once iron leaves the intestinal cells, an iron oxidase, known as hephaestin or ferroxidase, converts iron from the ferrous state to the ferric state.
  - Nonintestinal cells use the plasma protein ceruloplasmin to oxidize iron.
- Iron is rapidly bound to transferrin, an iron-binding protein of the blood that delivers iron to liver cells and from liver cells for storage to other tissues via receptor-mediated endocytosis.





Properties of transferrin

Transferrin + ferric = Holo transferrin

without ferric

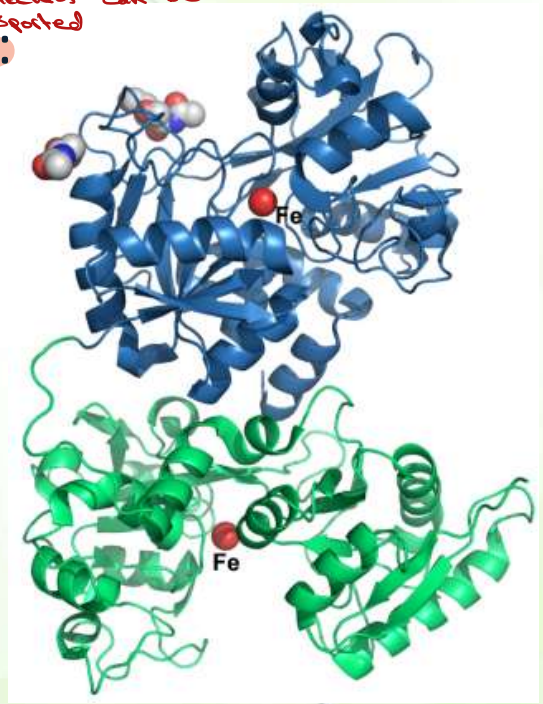
- Apotransferrin can bind several metals, but ferric, not ferrous, iron has highest affinity forming ferrotransferrin.
- Transferrin contains two sites that bind ferric irons:
  - 1/9 of the transferrin molecules have iron bound at both sites.
  - 4/9 of them have iron bound at one site
  - 4/9 have no iron bound.
    - *This means that iron-binding sites of transferrin are normally only about 1/3 saturated with iron.*
- When iron exceeds normal levels, non-transferrin-bound iron (NTBI) appears.

2 iron molecules can be transported

Saturated 2 atoms

one iron

free



Receptor-mediated endocytosis

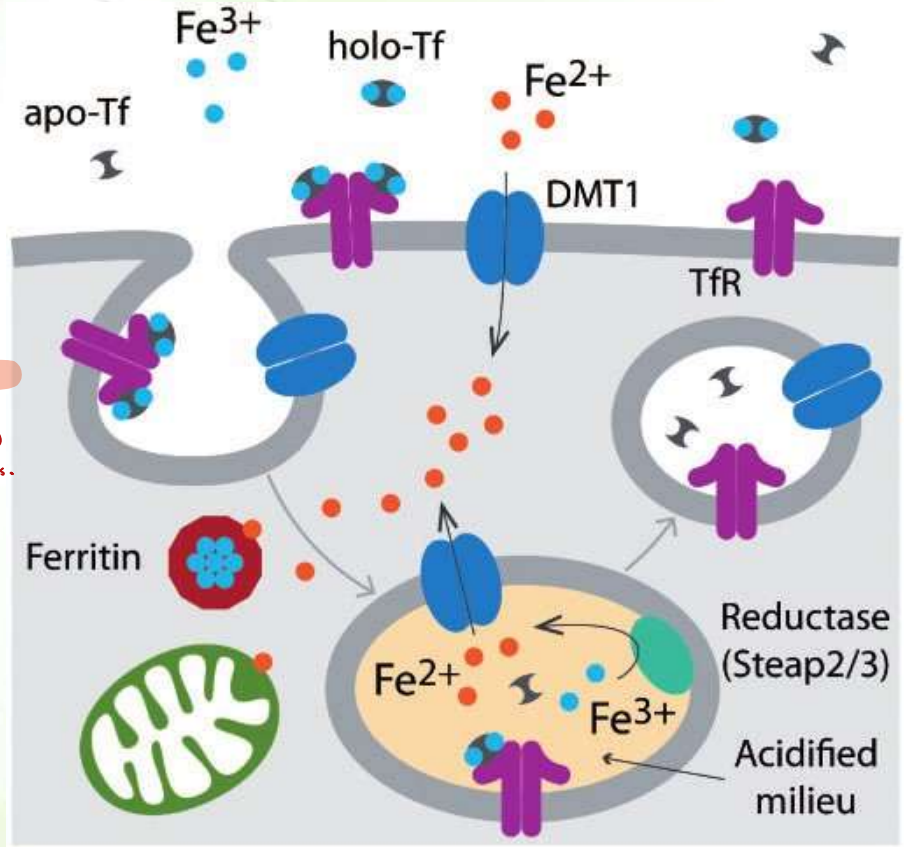
Holo transferrin passes the blood and the cells that need iron will induce transferrin receptors on their surfaces

Receptor mediated endocytosis

- Ferrotransferrin binds to a transferrin receptor (TfR) triggering endocytosis into early endosomes (pH of 6.0).
- Early endosomes are transformed into late endosomes (pH of 5.0) where Fe^{3+} atoms dissociate, get reduced into $Fe2^{+}$ by the ferrireductase STEAP3, and are transported into the cytosol via DMT1.
- The apotransferrin-transferrin receptor complex is recycled back to the surface, apotransferrin dissociates, and the receptor binds another transferrin.
- Affinity of TfR to iron: diferric Tf (Fe2Tf) >monoferric Tf (Fe1Tf) >apo-Tf

ferritin

In the late endosome
① ferric released from the transferrin and gets reduced (Fe^{+2})
② the transferrin gets released from the transferrin receptors.



Regulation of protein function

Hepcidin (iron sensor)

The main regulator of iron metabolism

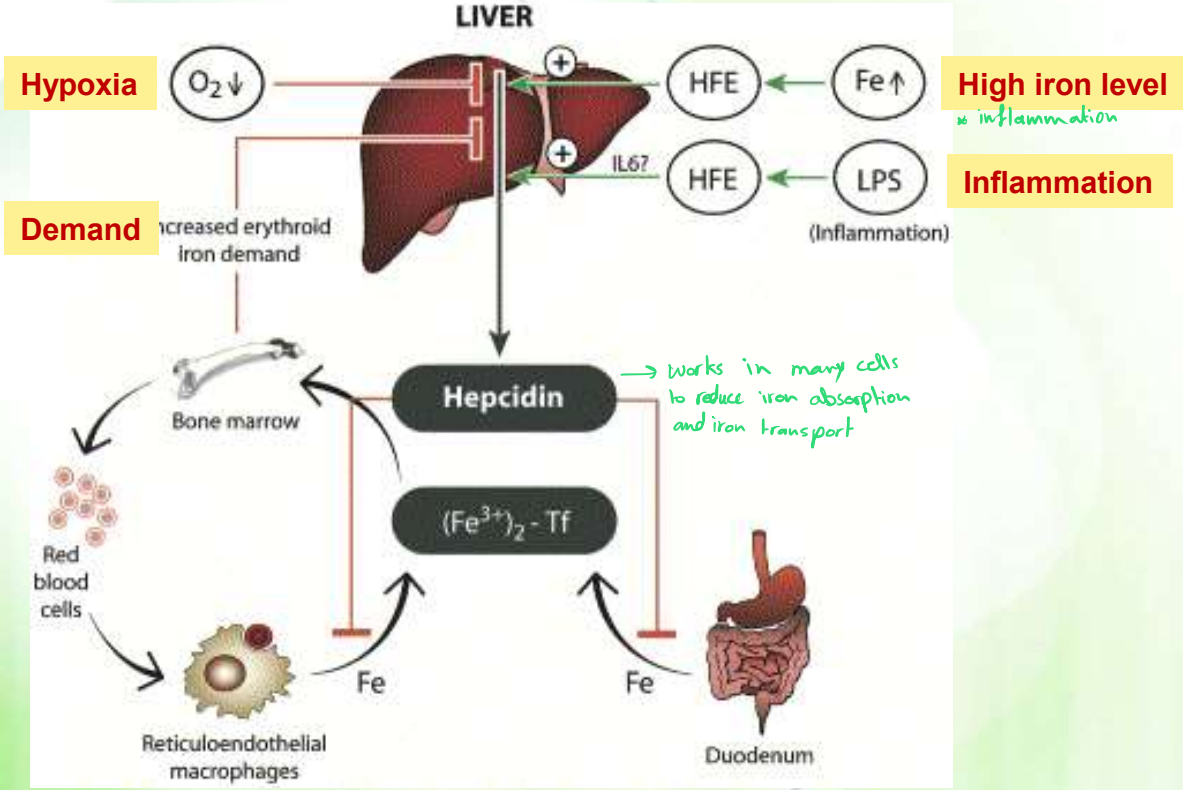
- Hepcidin is a peptide hormone (25 amino acids) secreted by the liver and **reduces** iron level.

A short peptide

Main function → (iron sensor)

- When iron level increases and in cases of inflammation, hepcidin secretion increases.
- When iron levels are low, there is high iron demand, or hypoxia, its release is suppressed.

↓ expression
↓ Release
↑ absorption (uptake)

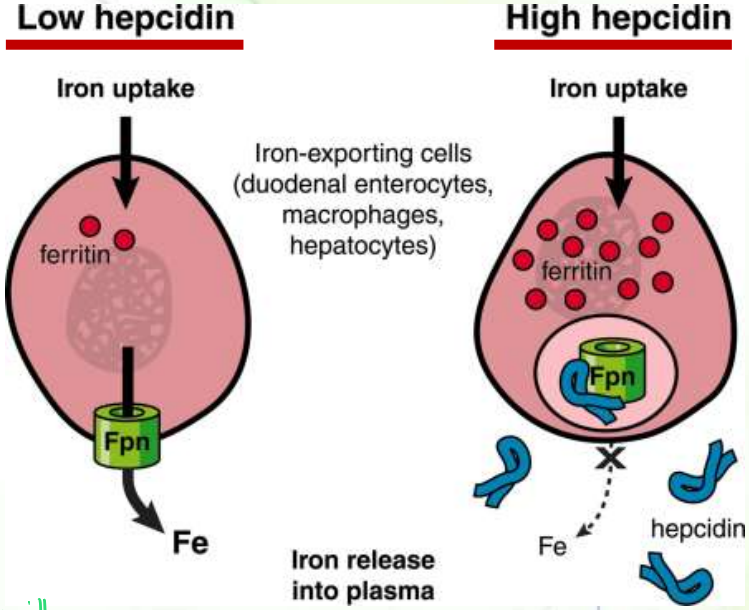


How does hepcidin reduce iron levels in the body?

ferroportin

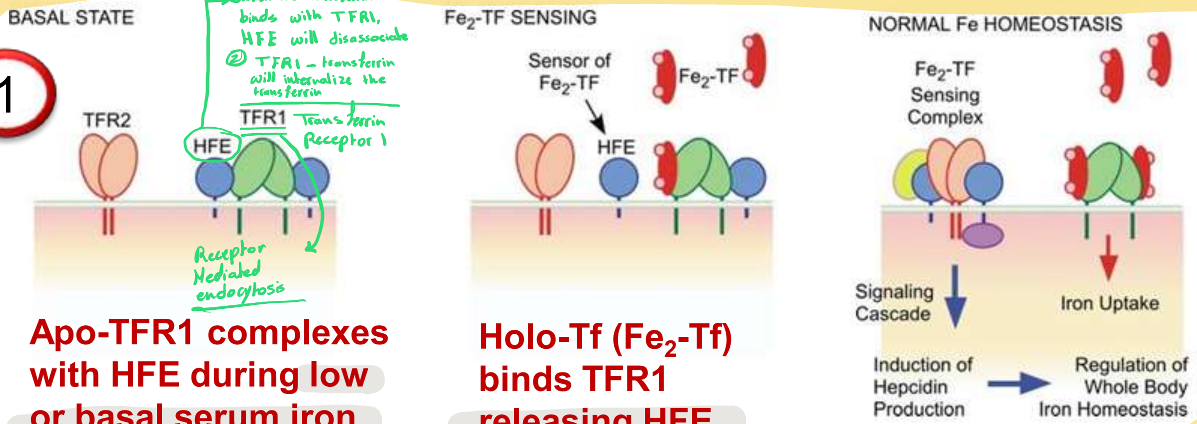
DMT1

- Hepcidin binds to the basolateral iron transporter ferroportin inducing ferroportin internalization and degradation.
  - This results in higher iron storage.
    - Iron is eliminated by sloughed off intestinal cells
    - Iron is not released from macrophages.
- Hepcidin also inhibits the presentation of the iron transporters (e.g. DMT1) in intestinal membranes decreasing iron absorption.



Regulation of hepcidin

More than one mechanism
More than one signaling pathway



1 **Apo-TFR1 complexes with HFE during low or basal serum iron conditions.**

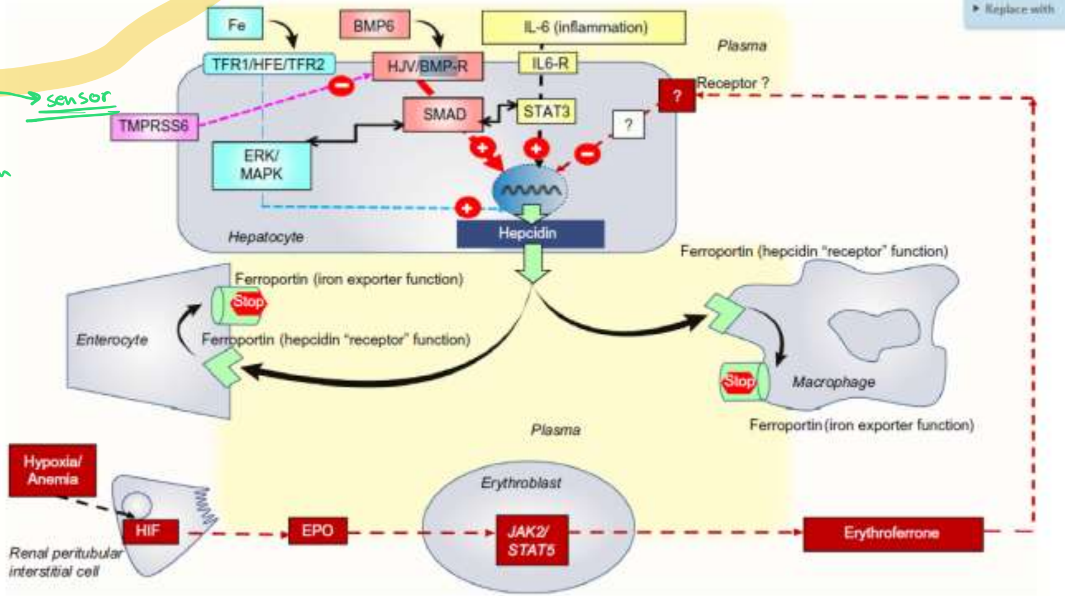
Holo-Tf (Fe_2-Tf) binds TFR1 releasing HFE.

HFE binds TFR2 and induces an intracellular signaling that stimulates hepcidin production.

2 **Inflammatory cytokine, IL6, induces the expression of hepcidin**

3 **Release of bone morphogenic protein 6 (BMP6) is induced by intracellular iron, which binds to its receptor (BMPR). BMPR is bound to hemojuvelin (HJV) protein stimulating the synthesis of hepcidin.**

4 **The expression of hepcidin is negatively regulated by anemia and hypoxia, which induce the synthesis of EPO (erythropoietin) by the kidney. EPO stimulates the synthesis of erythroferrone, which inhibits the synthesis of hepcicin.**



Post-transcriptionl regulation of expression → mRNA Stability

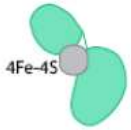
Iron-response element

DNA or RNA sequence

Iron regulatory protein

Bind with

The results of this binding depend on the location of this element.

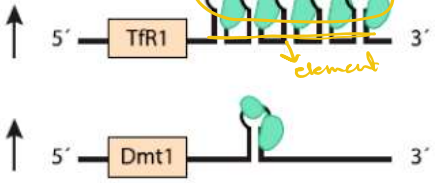


IRE in the 3′ UTR
mRNA stabilization

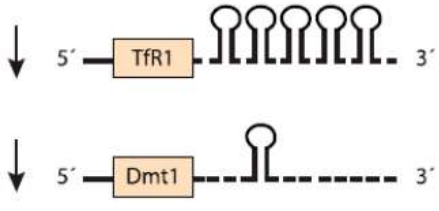
3′ untranslated region

iron regulatory protein

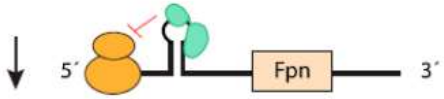
element



IRE in the 3′ UTR
mRNA degradation

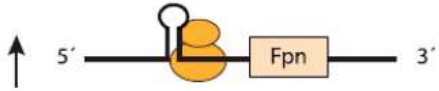


IRE in the 5′ UTR
Translational repression



Ferritin
ALAS

IRE in the 5′ UTR
Translation occurs

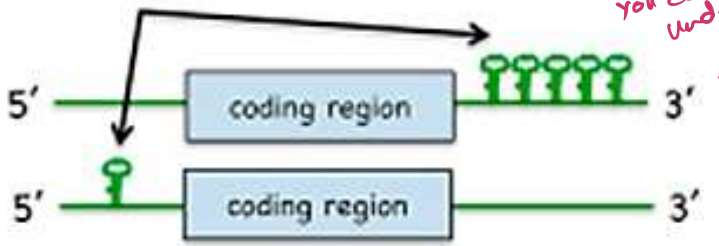


Ferritin
ALAS

IRE

11:00 - 18:30

you can understand better



Low Fe	High Fe
IRP1: active	IRP1: inactive
IRP2: active	IRP2: degraded
Fe uptake: ↑	Fe uptake: ↓
Fe storage: ↓	Fe storage: ↑
Fe export: ↓	Fe export: ↑
Heme synthesis: ↓	Heme synthesis: ↑
TCA cycle: ↓	TCA cycle: ↑

Iron Responce Element :-

1- 3′ untranslated region → Stabilizing of mRNA → long period work ↑ protein level

2- 5′ untranslated region → before the start codon → intruption of translation → destabilization
short period work ↓ protein level

Iron-related diseases

Hereditary hemochromatosis (HH)
Iron-deficiency anemia

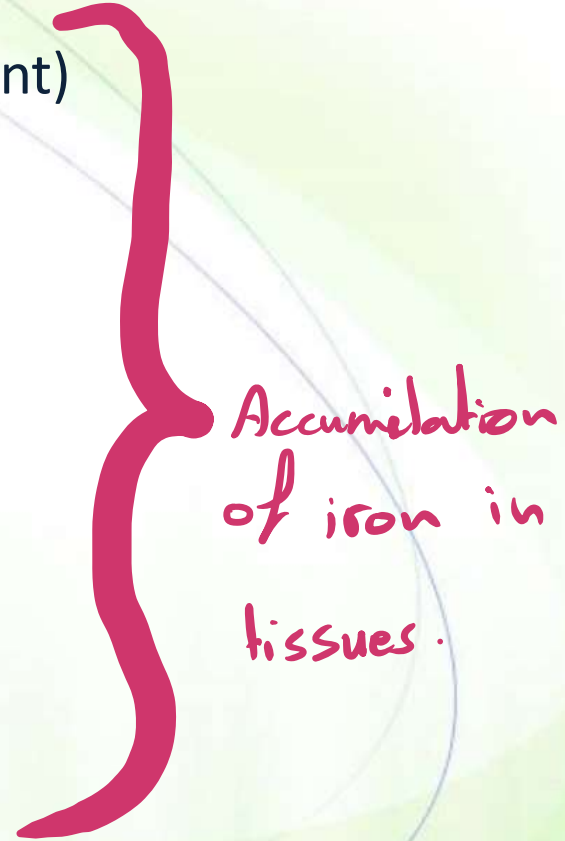
Hereditary hemochromatosis

excess iron in the system

- It is a group of disorders in iron metabolism that is characterized by excess iron absorption, saturation of iron-binding proteins and deposition of hemosiderin in the tissues.
  - more commonly in males than in females (why?) females have menestruation cycle
- The primary cause of hemochromatosis is the inheritance of an autosomal recessive allele designated as HFE (type I or primary HH) , but four other genes that regulate the hepcidin–ferroportin axis can also be involved.

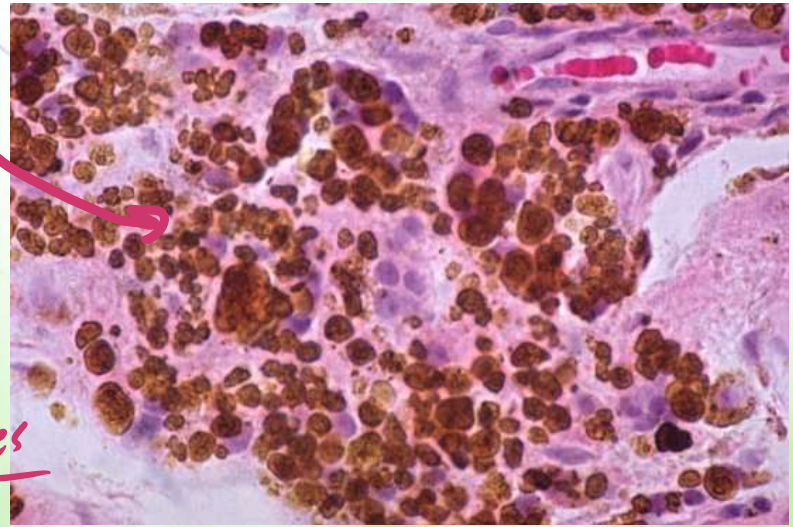
Groups/classes of hereditary hemochromatosis

- Type 1 (hemochromatosis protein, HFE-dependent)
  - Most common
- Type 2A (HJV-dependent)
- Type 2B (hepcidin-dependent)
- Type 3 (TfR2-dependent)
- Type 4 (ferroportin-dependent)
  - Autosomal dominant disorder



Hemosiderin → Iron is Toxic → Tissue Damage (liver, heart, skin — tissues that store iron)

- The normal total body iron stores may range from 2 to 6 gm, but persons with hemochromatosis have much greater stores exceeding 50 gm.
- If the capacity for storage of iron in ferritin is over-saturated, iron is stored as water-insoluble deposits known as hemosiderin, mainly in macrophages.
- Excess hemosiderin leads to cellular dysfunction and damage.



Dark brown spots in tissues

Affected organs and conditions

- Liver (hepatic fibrosis)
- Pancreas (diabetes mellitus)
- Joints (arthropathy)
- Skin (pigmentation)
- Heart (cardiomyopathy)
- Gonadotrophin-secreting cells (hypogonadotrophic hypogonadism)

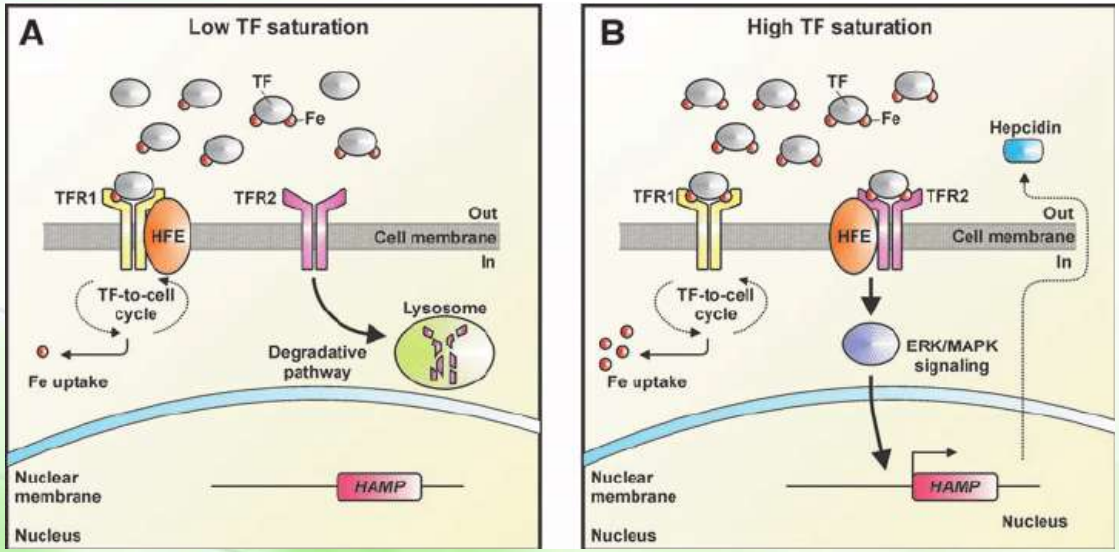
Regulation of transferrin receptor

- HFE is a major histocompatibility complex (MHC) class-1 gene.
- Normal HFE complexes with TfR1 reducing iron transfer into cells.
- Mutated HFE (e.g. C282Y) has reduced presence on membrane and/or lack of interaction with Tfr1, loss of inhibition of transferrin receptor, and, therefore, increased iron uptake and storage.

more prominant in adults

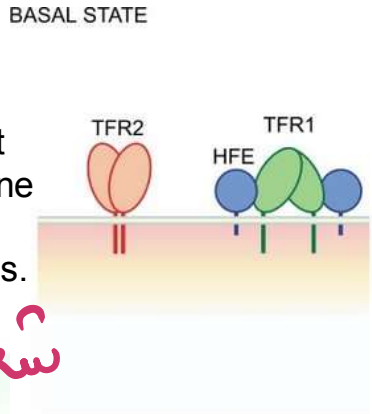
hepcidin expression

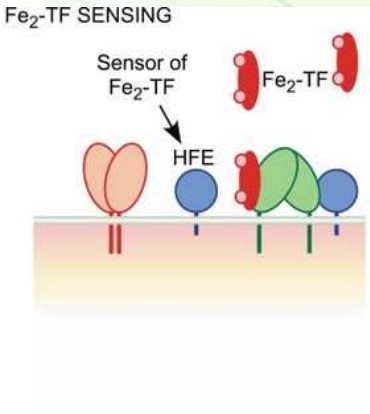
cystine → tyrosine



Mechanism of action

TFR1 exists as a complex with HFE at the plasma membrane during low or basal serum iron conditions.

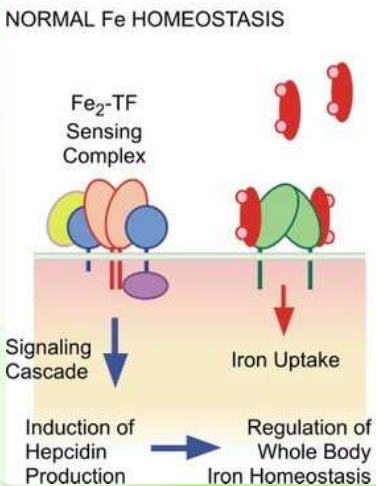


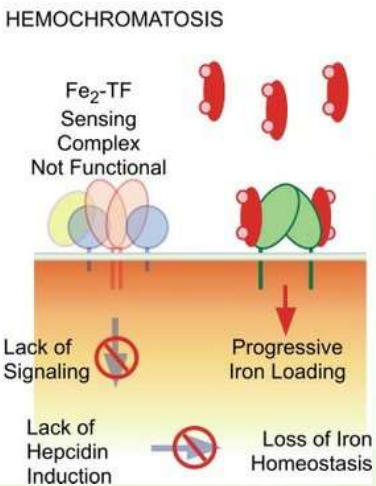


Serum Fe2 -TF competes with HFE for binding to TFR1. Increased serum transferrin saturation results in the dissociation of HFE from TFR1.

TFR1 شغال
TFR2 مش شغال

HFE binds TFR2 and induces a intracellular signaling that stimulates hepcidin production.





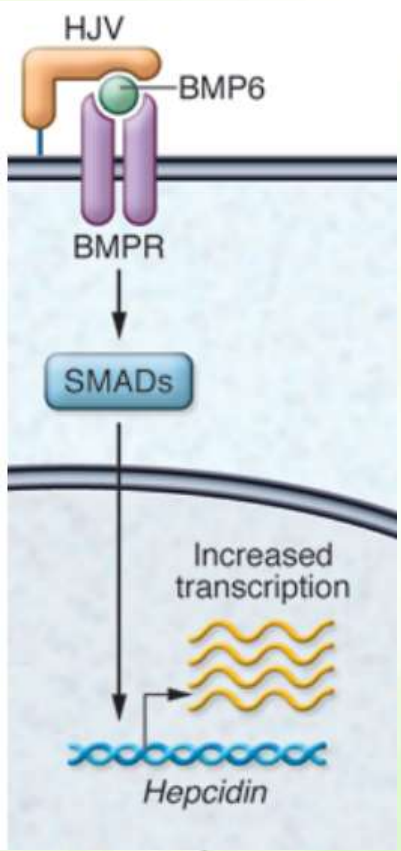
Mutation or absence of HFE or TFR2 prevents formation of a functional iron sensor and signal transduction effector complex leading to dysregulation of systemic iron homeostasis

Juvenile hemochromatosis → children

- Type 2A hereditary hemochromatosis
  - *AKA* HFE2 (HJV)-dependent hereditary hemochromatosis
- Mutations in HJV gene, which encodes the protein "hemojuvelin", account for the majority of JH.
- Normal HJV upregulates expression of hepcidin.

- Type 2B is also juvenile hemochromatosis but is caused by mutations in hepcidin gene. → children

abnormal hepcidin not functioning well.



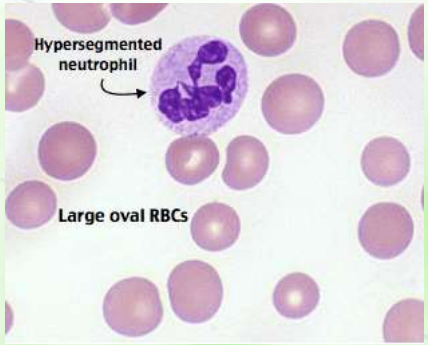
Iron-deficiency anemia

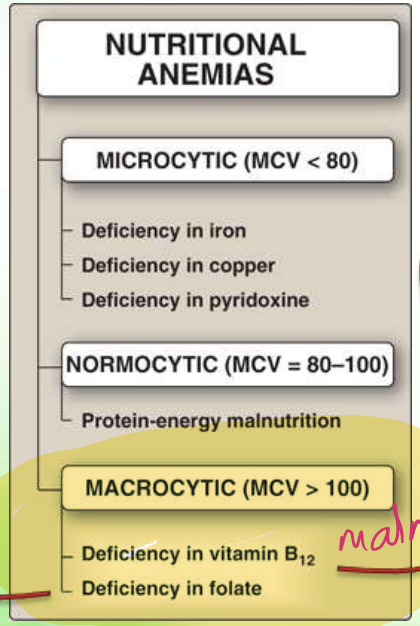
malnutrition

- Anemias are characterized by a deficiency in the number of mature erythrocytes in the circulation, lowering the oxygen-carrying capacity of the blood, causing tissue hypoxia, and clinical symptoms such as fatigue, weakness, increased cardiac output, as well as increased morbidity and mortality.

impaired nucleotide synthesis

Cells cannot synthesize DNA and, hence, cannot divide and megaloblasts accumulate.

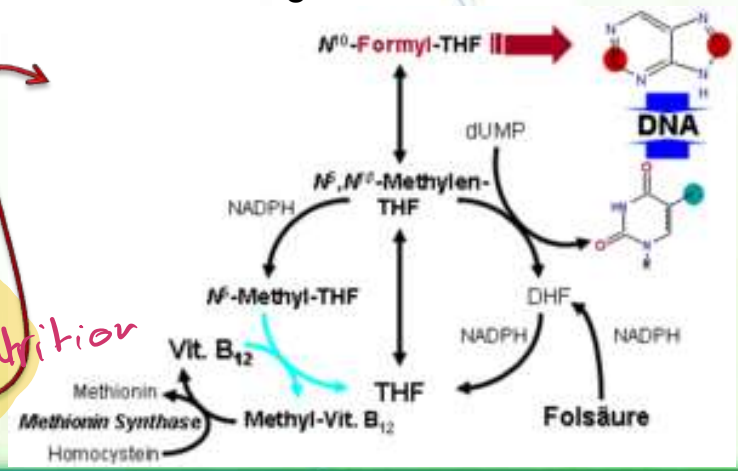




malnutrition

B_{12} is essential for folate regeneration

Folate is not regenerated

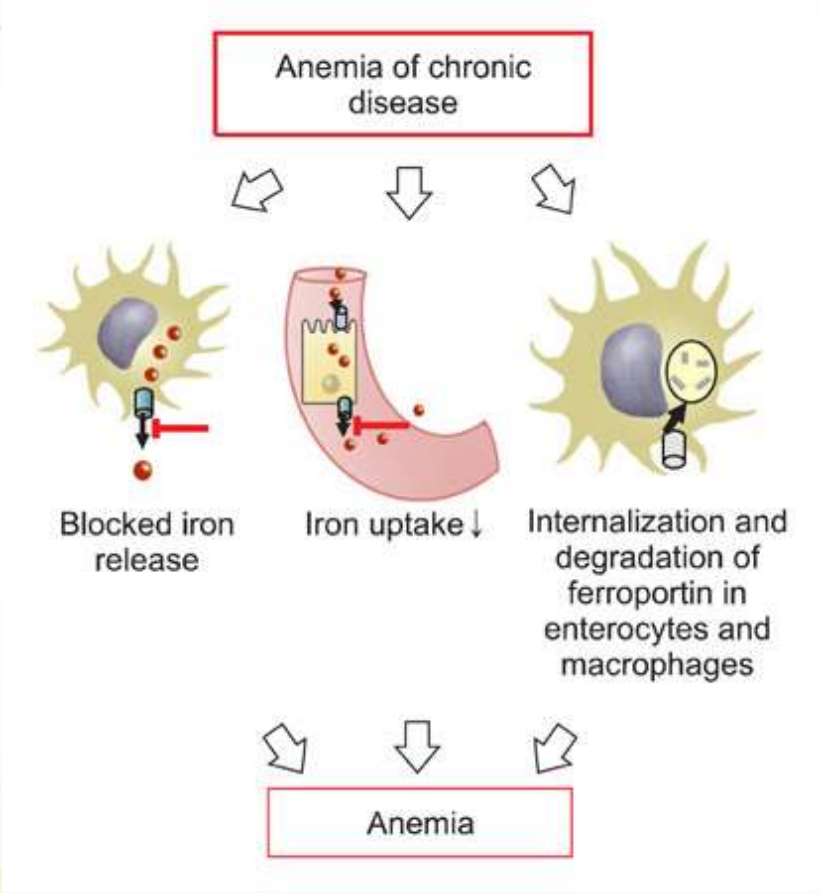


Anemia of chronic disease → advanced countries

inflammation ↑ stress ↑ immune ↓

- Causes: chronic kidney disease, chronic infections and chronic inflammatory diseases
- Inflammatory cytokines → increased hepcidin production by hepatocytes → downregulation of ferroportin expression in major iron-exporting cells such as macrophages, duodenal enterocytes, and hepatocytes → decreased enteric iron absorption and, perhaps more importantly, to increased iron retention within splenic macrophages and hepatocytes.

IL-6 / TNF



Additional molecular consequences of chronic inflammation