



Metabolism of iron

Prof. Mamoun Ahram
Hematopoietic-lymphatic system



- 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,
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- Brissot and Loréal, Iron metabolism and related genetic diseases: A cleared land, keeping mysteries, Journal of Hepatology 2016 vol. 64 j 505–515,
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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 a number of enzymes such as redox cytochromes and the P450 class of detoxifying cytochromes.
- 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.**

What is life cycle of iron in the body?



Well-nourished people: 3-4g

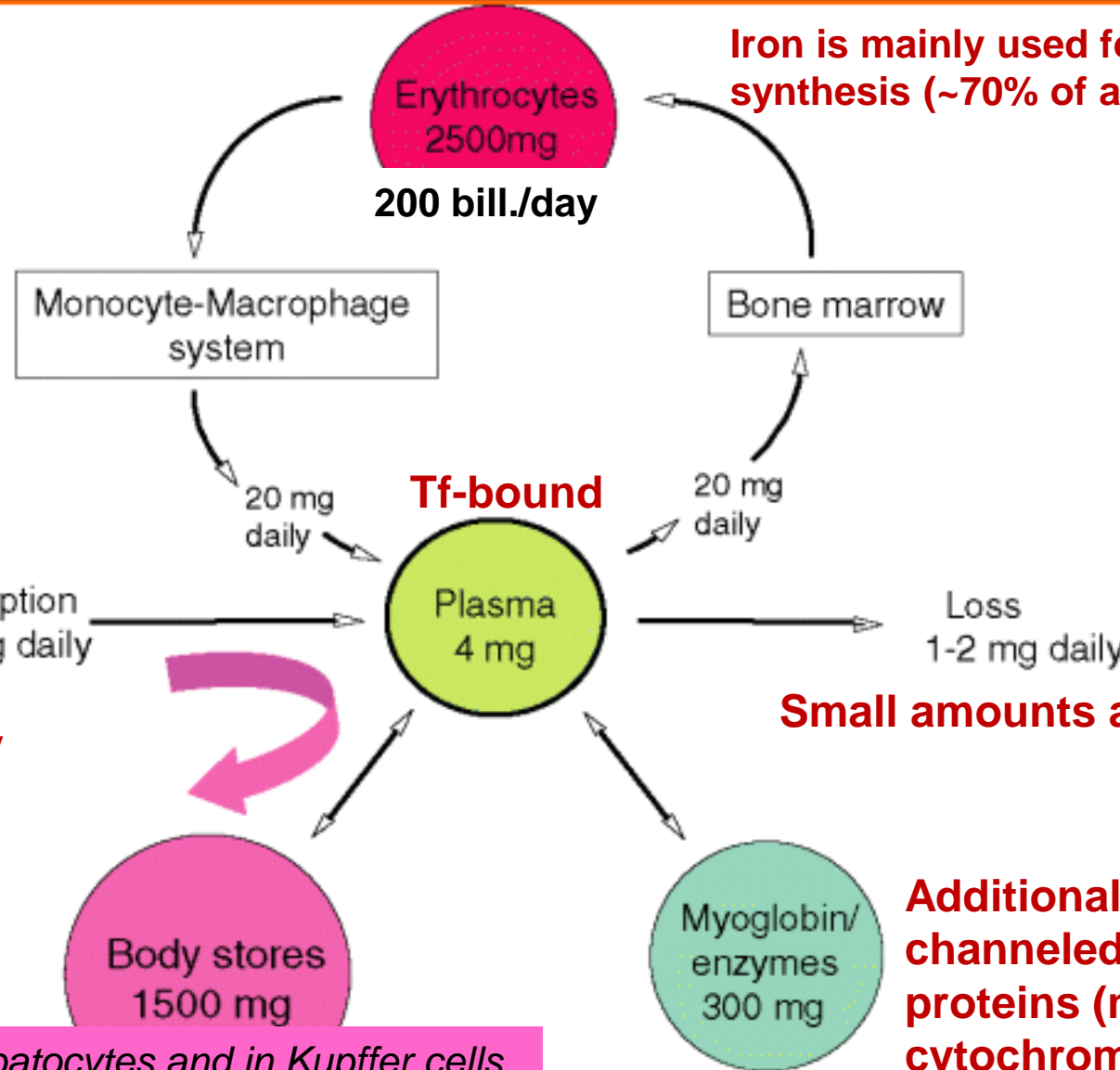
The released iron is scavenged by macrophages in the reticuloendothelial system.



Levels are maintained by dietary absorption.

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

Hepatocytes and in Kupffer cells (reticuloendothelial cells)



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

Small amounts are lost

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

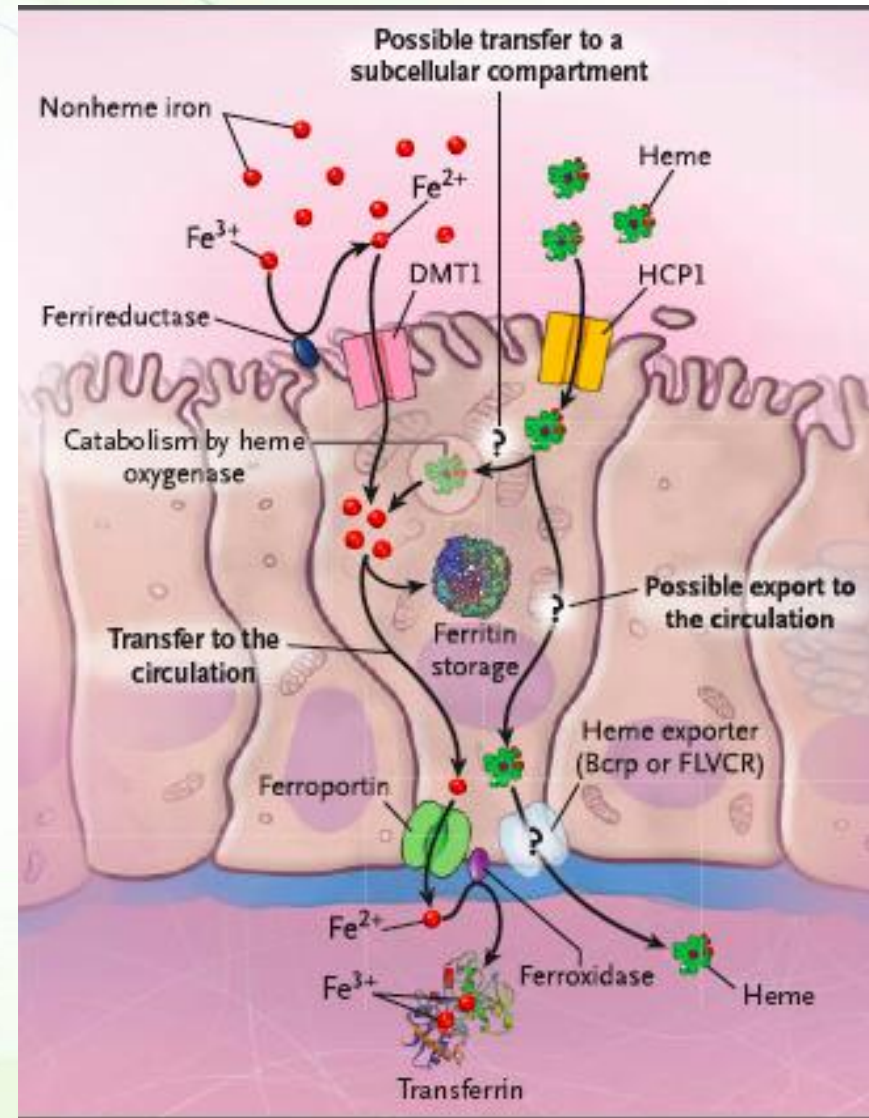


Iron absorption

State of iron



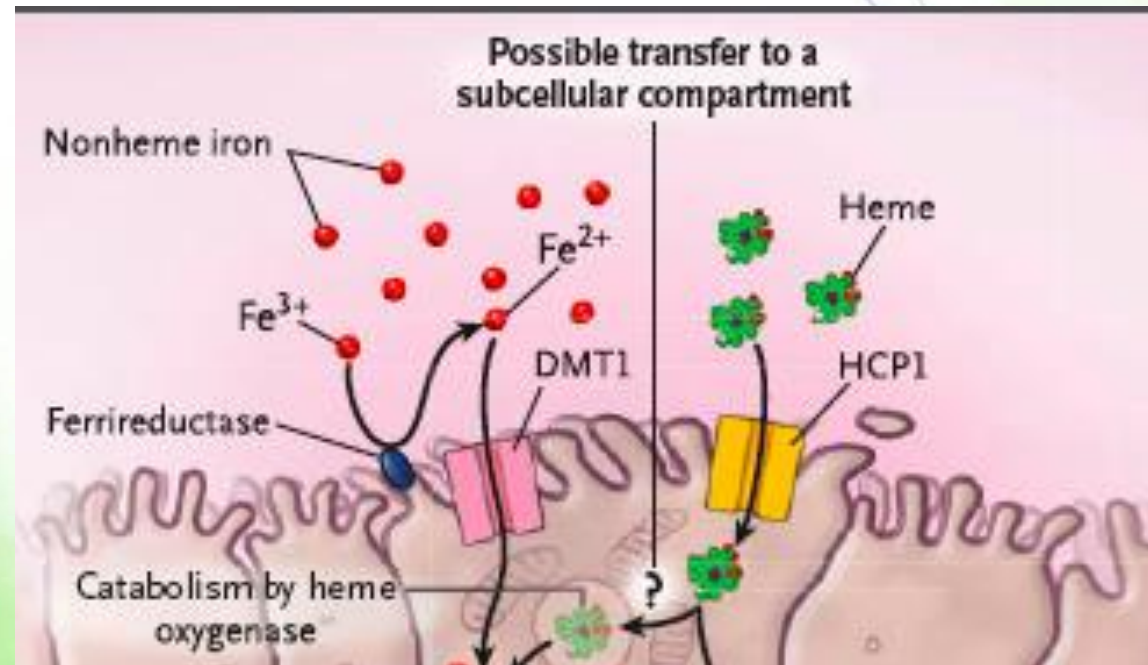
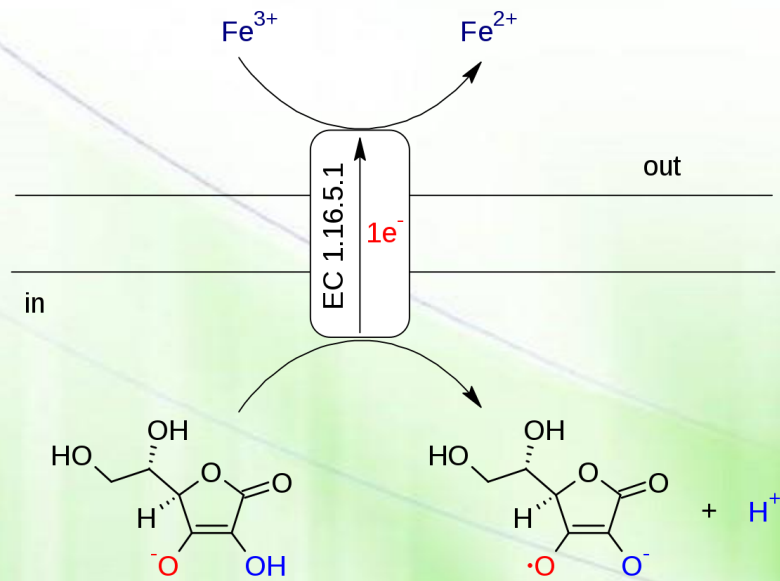
- Under conditions of neutral or alkaline pH, iron is found in the Fe^{3+} state and in the Fe^{2+} state at acidic pH.
- In the stomach, iron will be in the ferrous state.
- In the duodenum, iron is in the ferric state.
- However, to be absorbed, dietary iron must be in its ferrous Fe^{2+} form.



Site of absorption



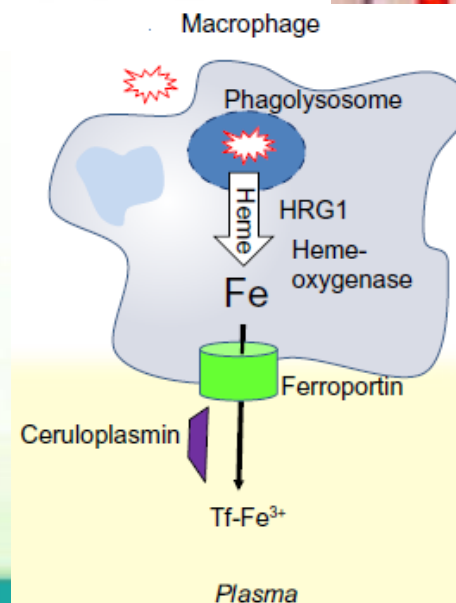
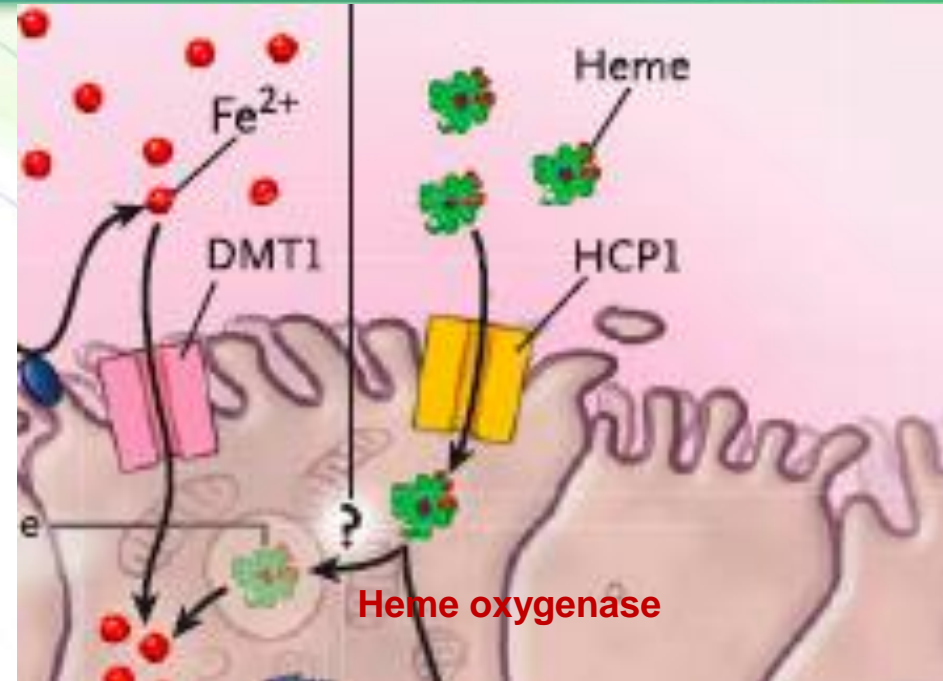
- A ferric reductase enzyme on the enterocytes' brush border, Dcytb (duodenal cytochrome b), reduces Fe^{3+} to Fe^{2+} .
- Divalent metal transporter 1 (DMT1) transports iron into the cell.
 - DMT-1 can transport other metal ions such as zinc, copper, cobalt, manganese, cadmium, and lead.



Heme as a source of iron



- Iron can also be obtained from 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.

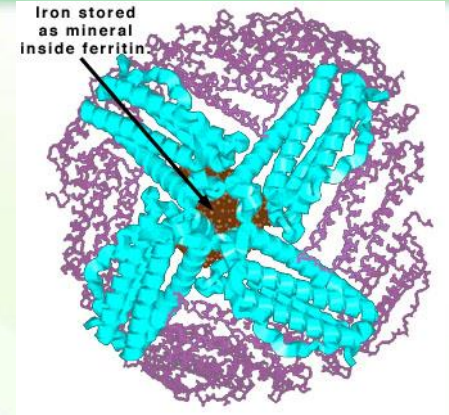


Fates of iron



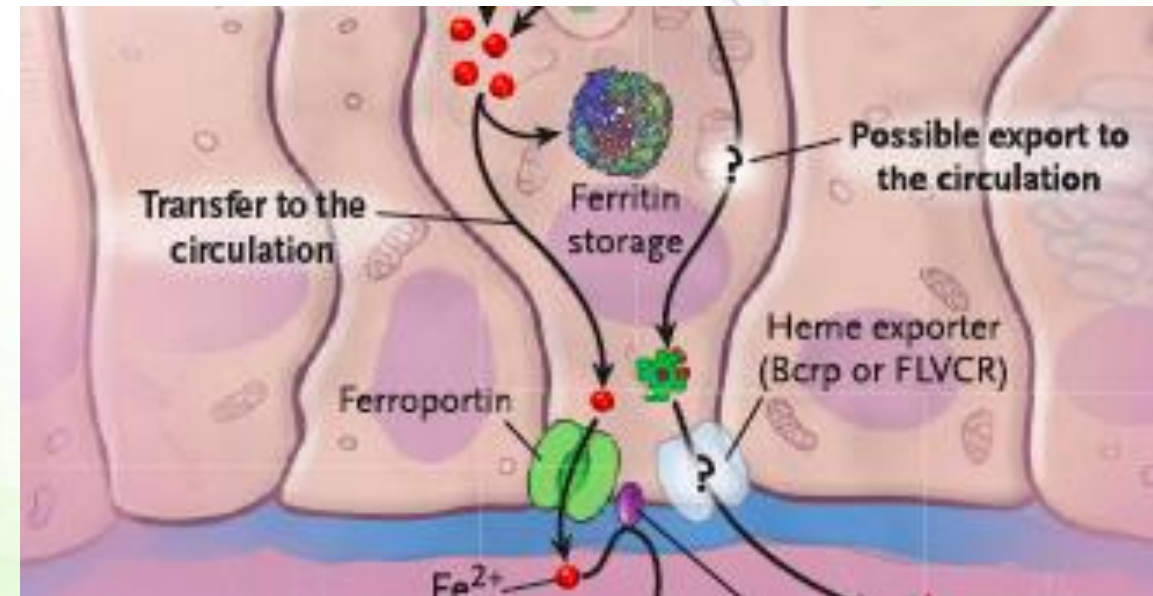
Fate 1: storage

- Cells can then store iron as ferritin.
 - 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, iron is eliminated from the body.



Fate 2: Transport

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



Intestine-related iron metabolism disorders



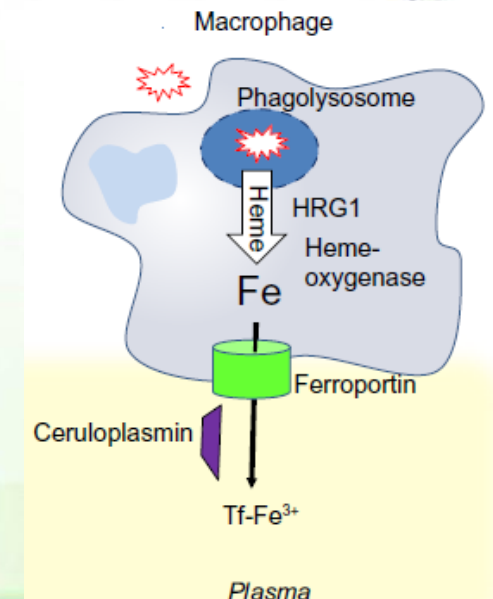
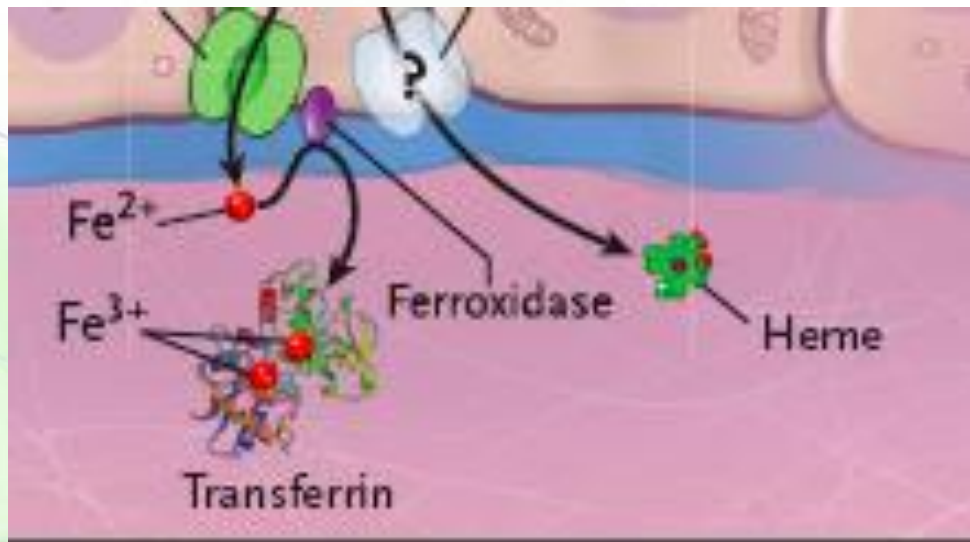
- Iron malabsorption
 - Gastrectomy (total or partial)
 - Celiac disease (villous atrophy)
 - Crohn's disease
 - Helicobacter pylori
- Intestinal hemorrhage (gastrointestinal-mediated iron loss)
 - Gastric cancer
 - Ulcers
 - Inflammatory bowel disease
 - Hookworm infection



Ferroxidase and transferrin



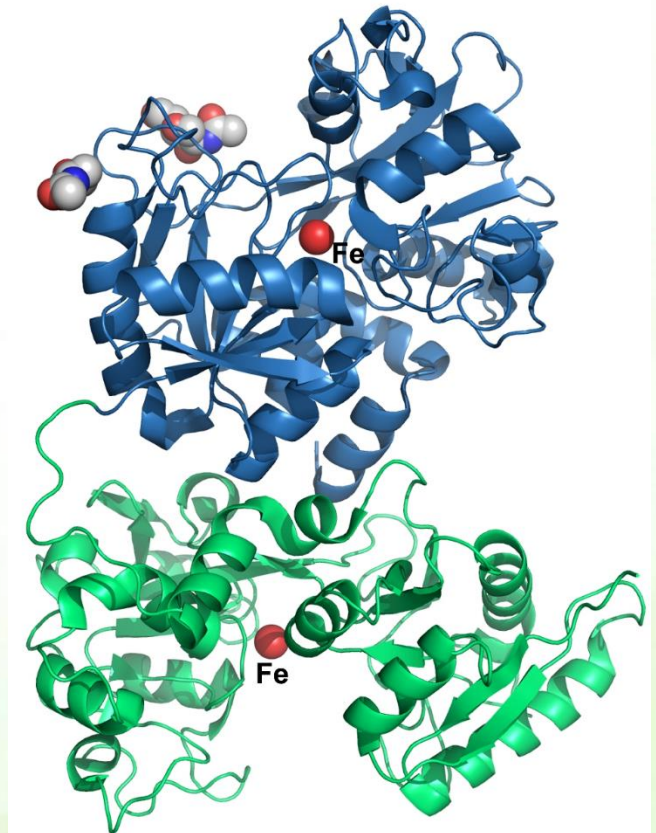
- Once iron leaves intestinal cells, an iron oxidase, known as hephaestin or ferroxidase, converts iron from the ferrous state to the ferric state.
 - Nonintestinal cells use the plasma protein ceruloplasmin to oxidize iron.
- Iron is rapidly bound to transferrin, an iron-binding protein of the blood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



Properties of transferrin



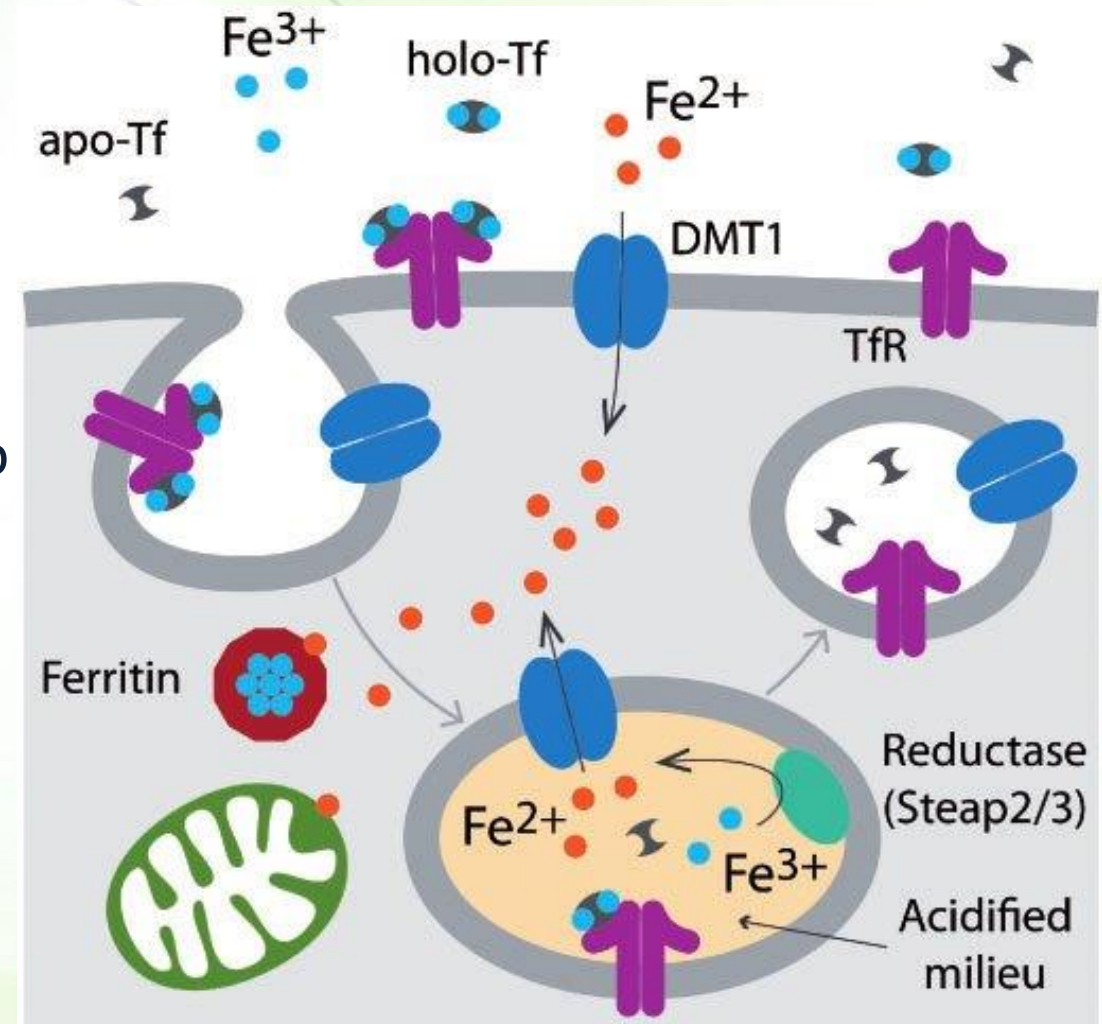
- Apotransferrin can bind several metals, but ferric, not ferrous, iron has highest affinity forming ferrotransferrin.
- Transferrin contains two sites that bind ferric iron:
 - 1/9 of the transferrin molecules have iron bound at both sites
 - 4/9 of them have iron bound at one site
 - 4/9 have no iron 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.



Receptor-mediated endocytosis



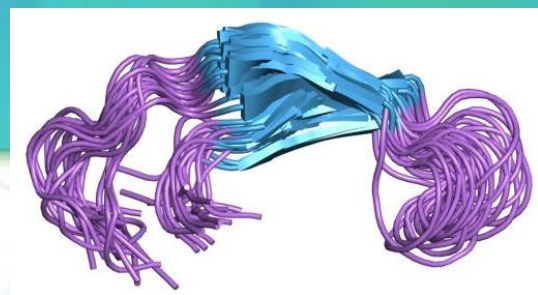
- Ferrotransferrin binds to a transferrin receptor (TfR) triggering endocytosis into early endosomes (pH of 6.0).
- Early endosomes are transformed into late endosomes (pH of 5.0) where Fe^{3+} atoms dissociate, get reduced into Fe^{2+} by the ferrireductase STEAP3, and are transported into the cytosol 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 (Fe_2Tf) > monoferric Tf (Fe_1Tf) > apo-Tf



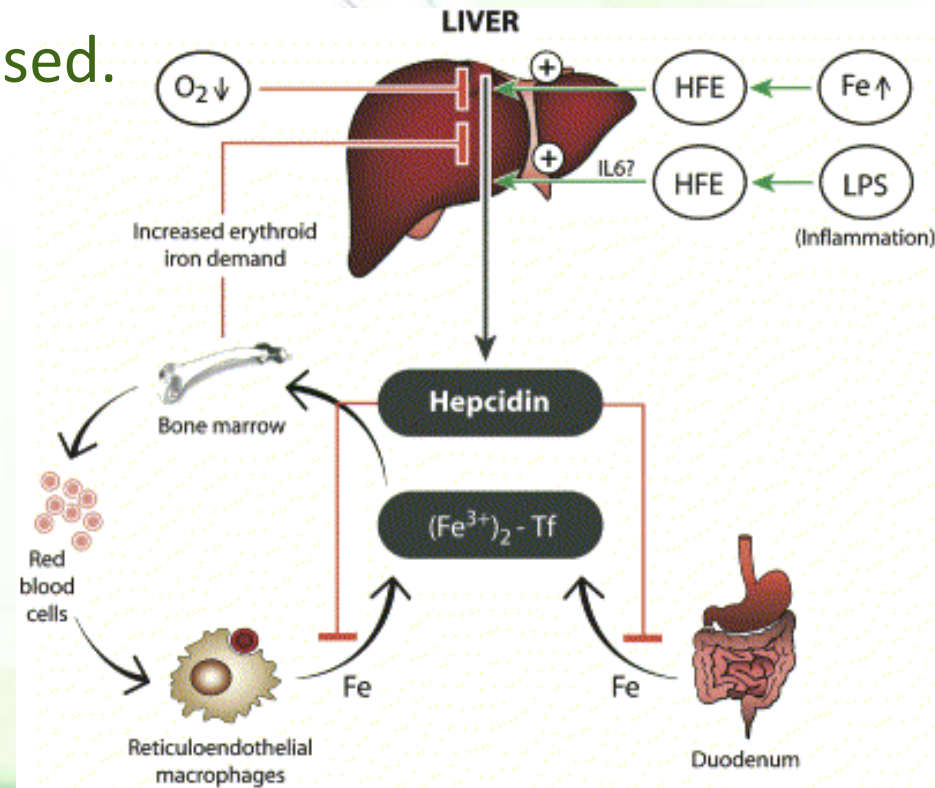
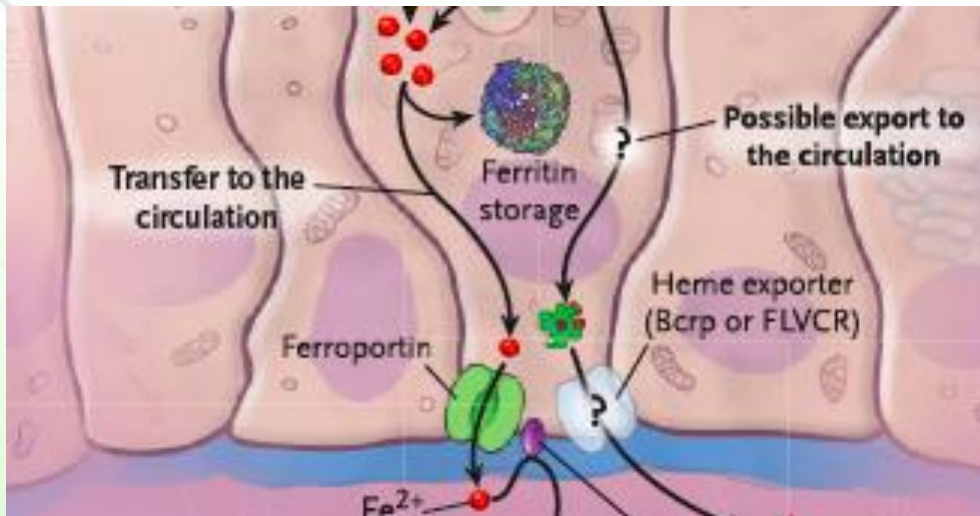


Regulation of protein function

Hepcidin (iron sensor)



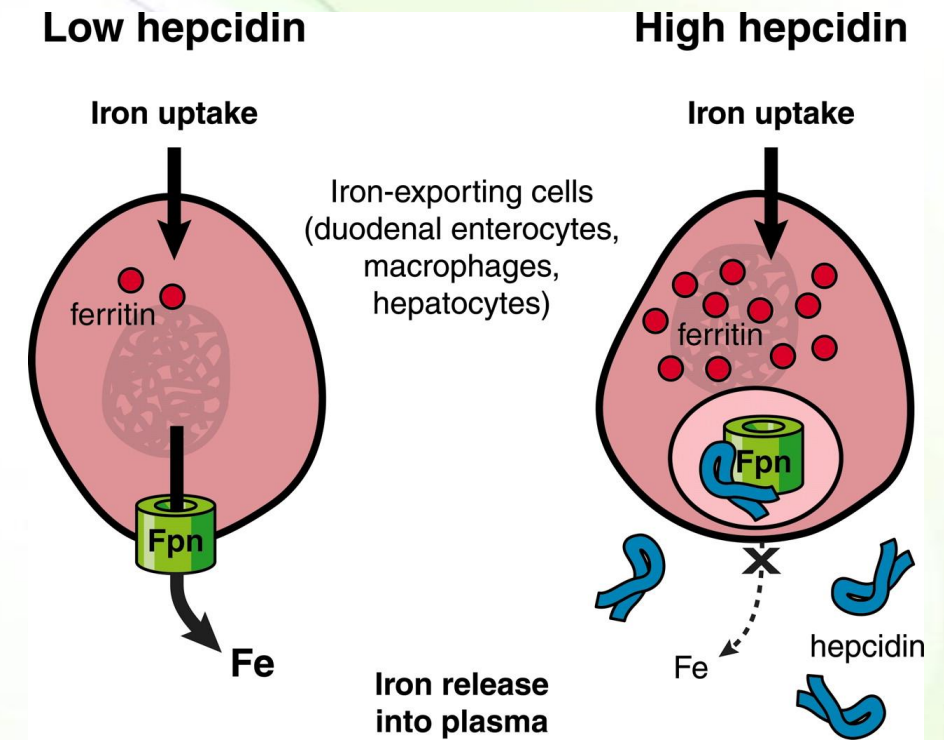
- Hepcidin is a peptide hormone (25 amino acids) secreted by the liver and regulates iron level
 - When iron level increases and in cases of inflammation, hepcidin secretion increases.
 - When iron levels are low, its release is suppressed.



How does hepcidin reduce iron levels in the body?



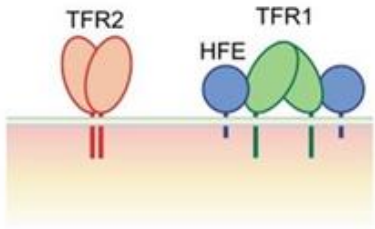
- Hepcidin binds to the basolateral iron transporter ferroportin inducing ferroportin internalization and degradation.
 - This results in higher iron storage.
- Hepcidin also inhibits the presentation of the iron transporters (e.g. DMT1) in intestinal membranes decreasing iron absorption



Regulation of hepcidin

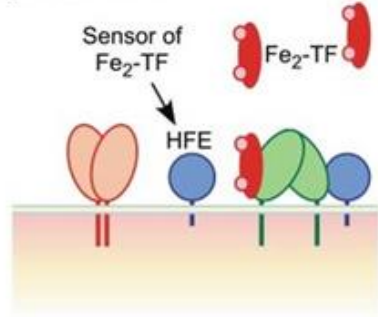


BASAL STATE



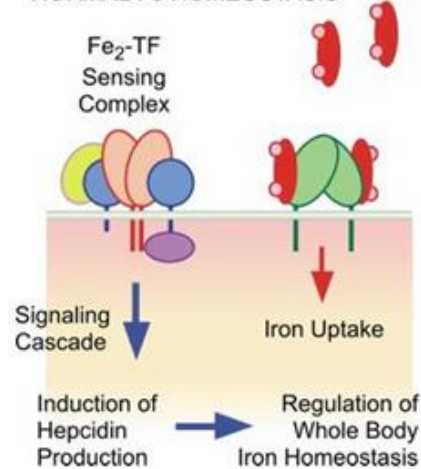
TFR1 complexes with HFE during low or basal serum iron conditions.

Fe₂-TF SENSING



Fe₂-Tf binds TFR1 releasing HFE.

NORMAL Fe HOMEOSTASIS

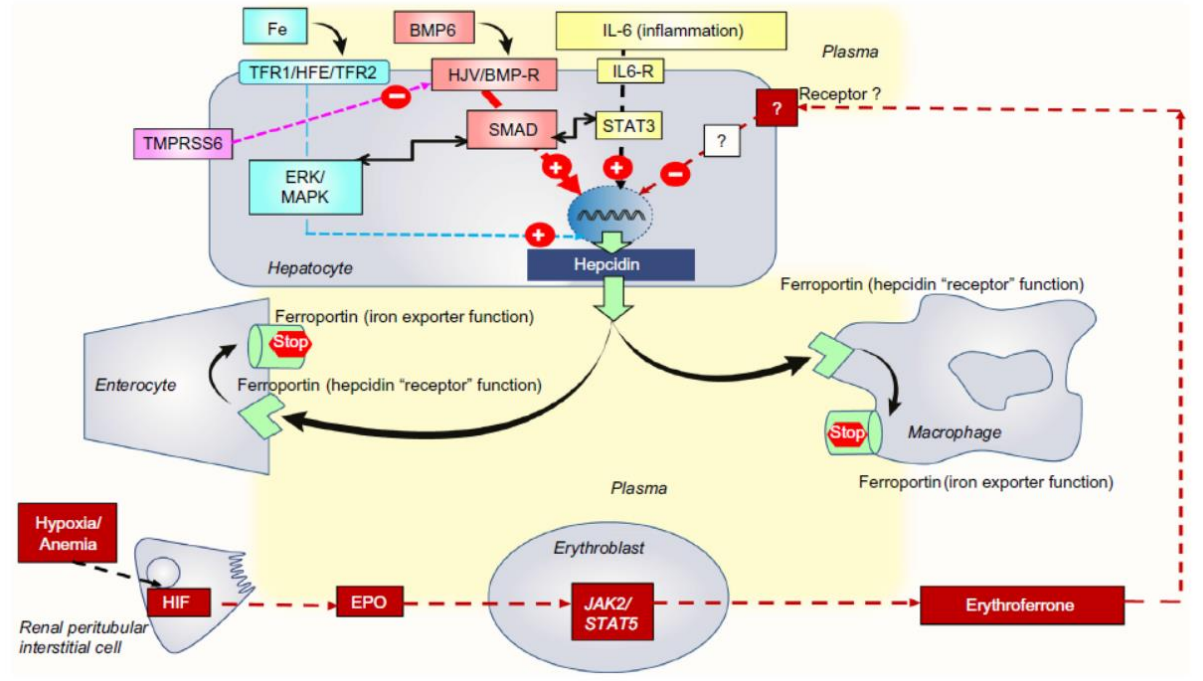


HFE binds TFR2 and induces an intracellular signaling that stimulates hepcidin production.

Inflammatory cytokine, IL6, induces expression of hepcidin

Bone morphogenic protein 6 (BMP6) binds to its receptor (BMPR) and activates expression of hepcidin. BMPR is bound to hemojuvelin (HJV) protein and is degraded by TMPRSS6.

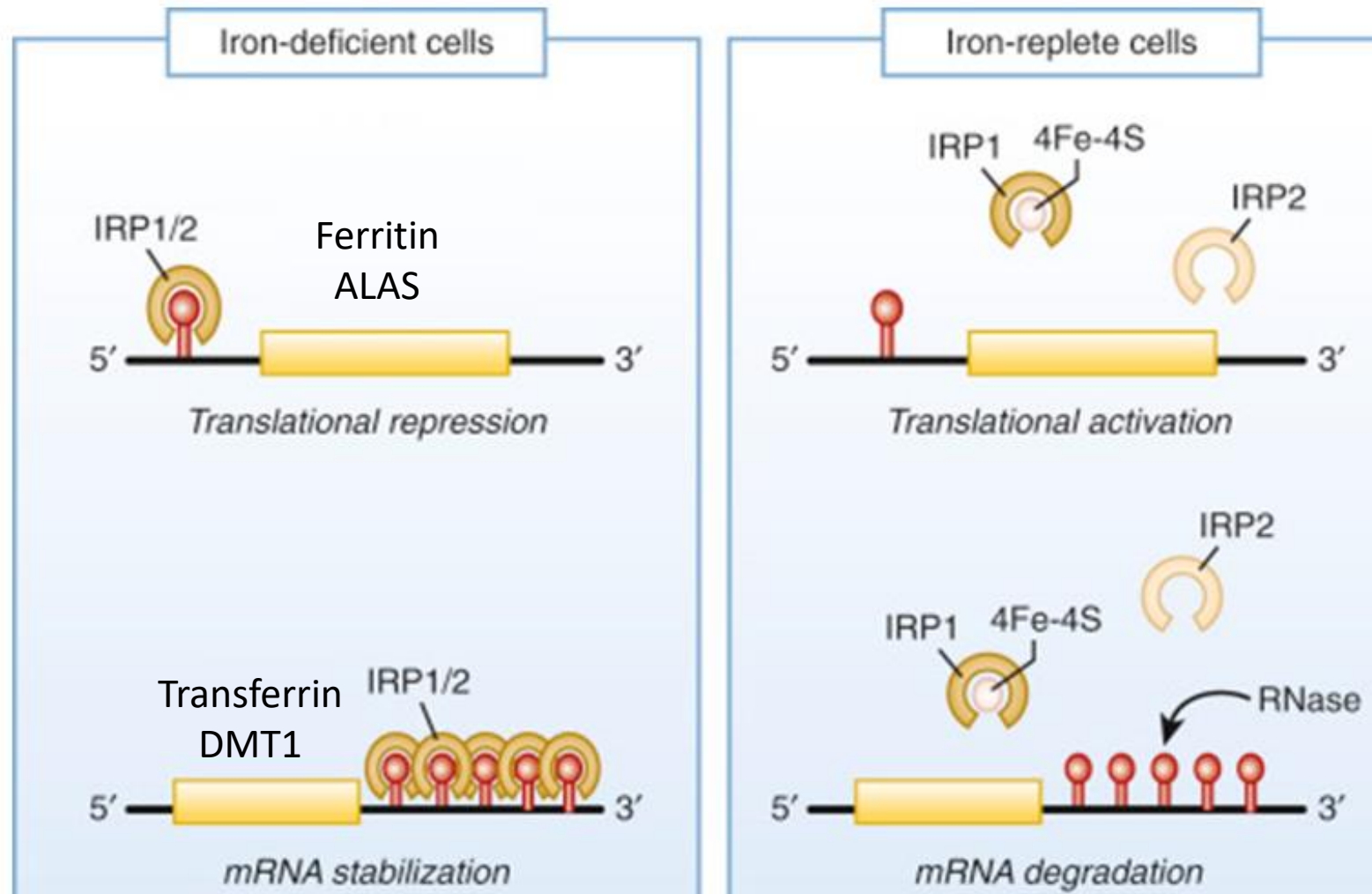
The expression of hepcidin is negatively regulated by anemia and hypoxia through erythroferrone, which is produced by the erythroblasts in response to EPO (erythropoietin) synthesis by the kidney.





Post-transcriptional regulation of expression

Iron-response element and its binding protein





Iron-related diseases

Hereditary hemochromatosis (HH)
Iron-deficiency anemia

Hereditary hemochromatosis



- 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?)
- 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

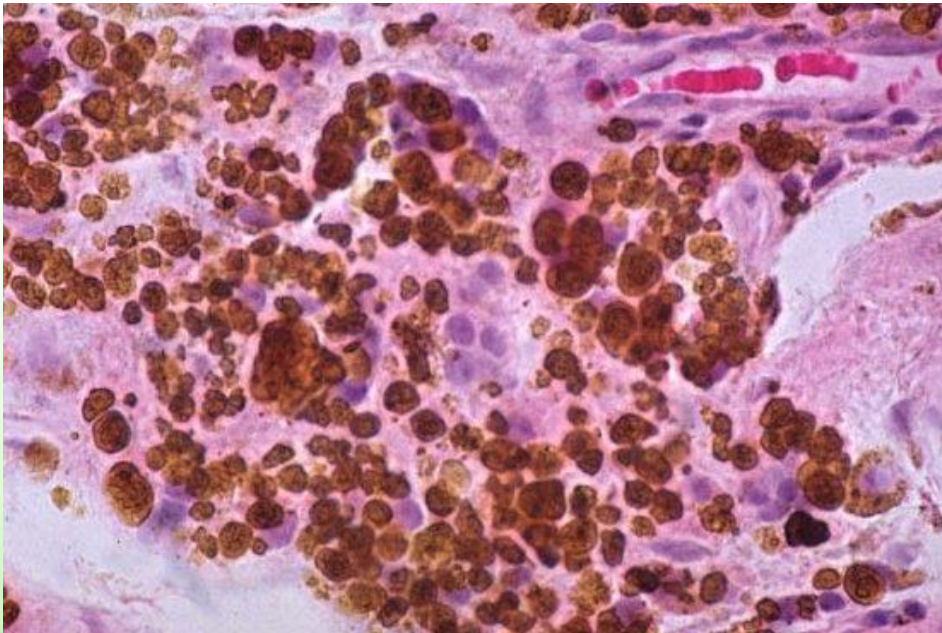


- Type 1 (hemochromatosis protein, HFE-dependent)
 - Most common
- Type 2A [HFE2 (HJV) dependent]
- Type 2B (hepcidin, HAMP-dependent)
- Type 3 (TfR2-, TfR2-dependent)
- Type 4 (ferroportin dependent)
 - Autosomal dominant disorder

Hemosiderin



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



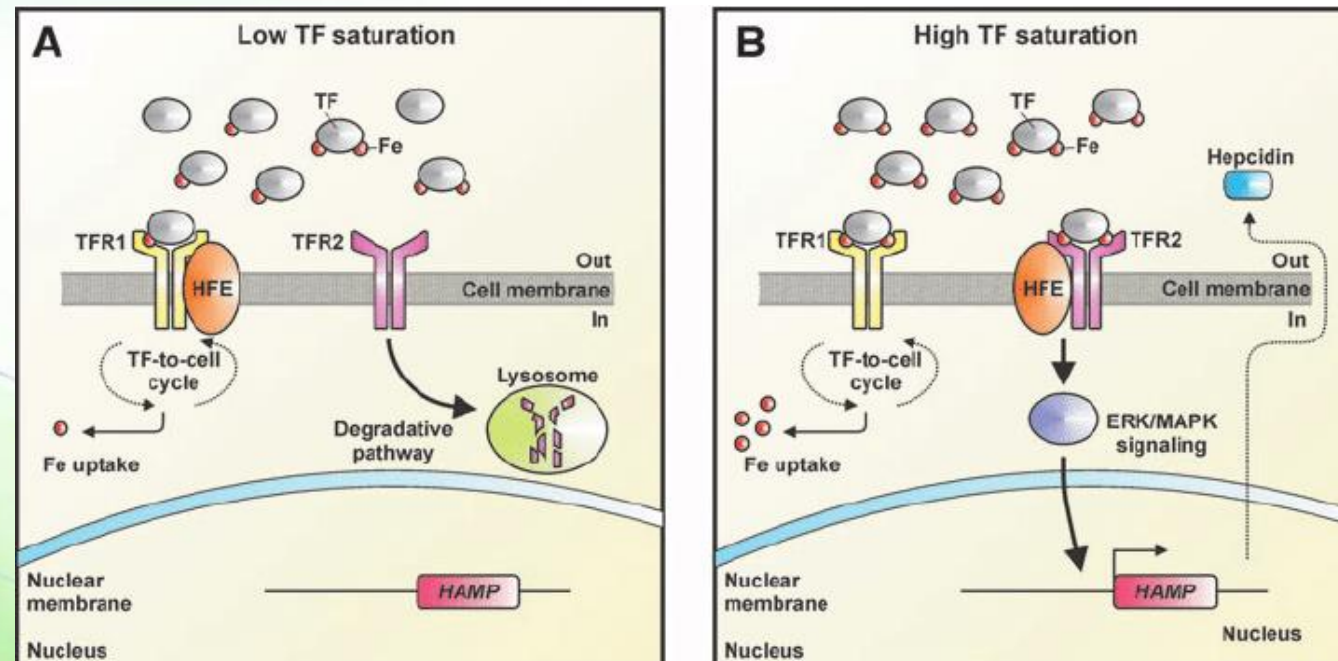
Affected organs and conditions

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

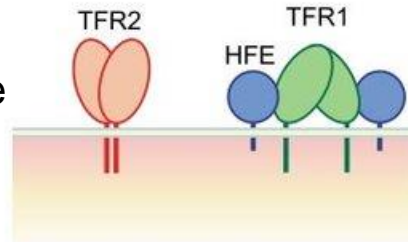


Mechanism of action

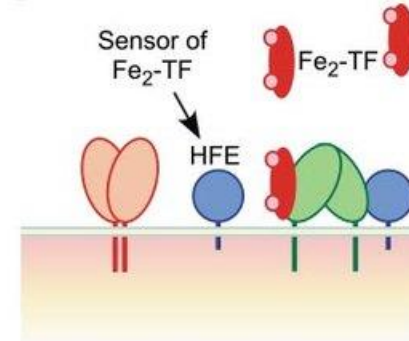


TFR1 exists as a complex with HFE at the plasma membrane during low or basal serum iron conditions.

BASAL STATE



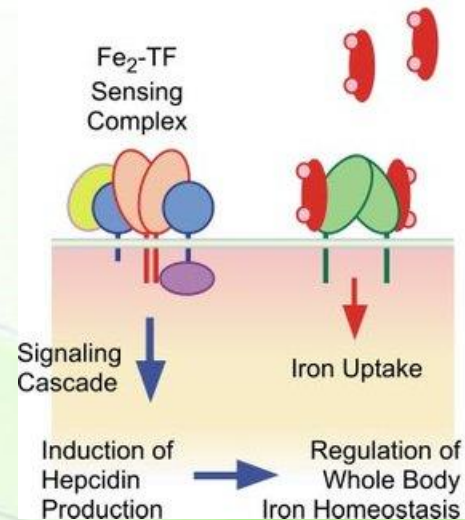
Fe₂-TF SENSING



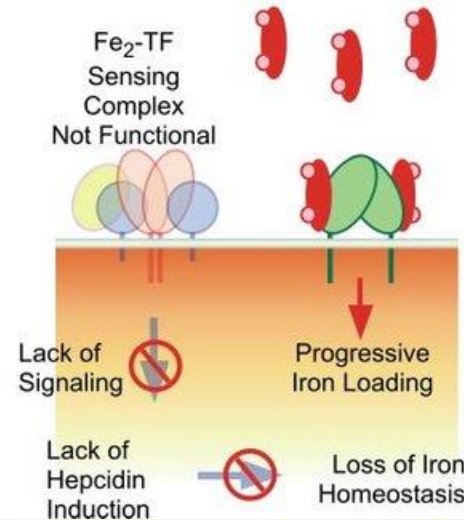
Serum Fe²⁺-TF competes with HFE for binding to TFR1. Increased serum transferrin saturation results in the dissociation of HFE from TFR1.

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

NORMAL Fe HOMEOSTASIS



HEMOCHROMATOSIS



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

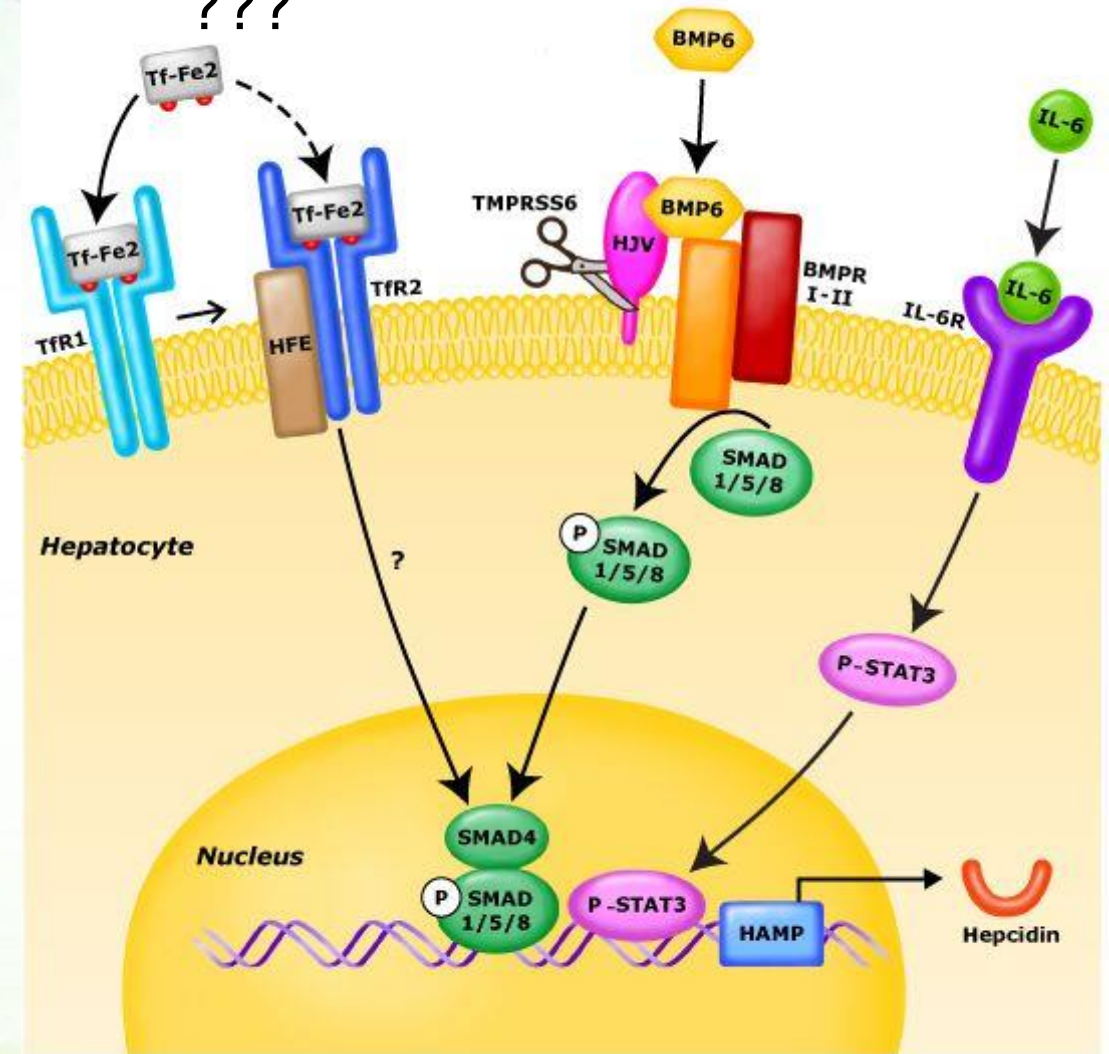
Juvenile hemochromatosis



- 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.

Re-record

No production of hepcidin
???

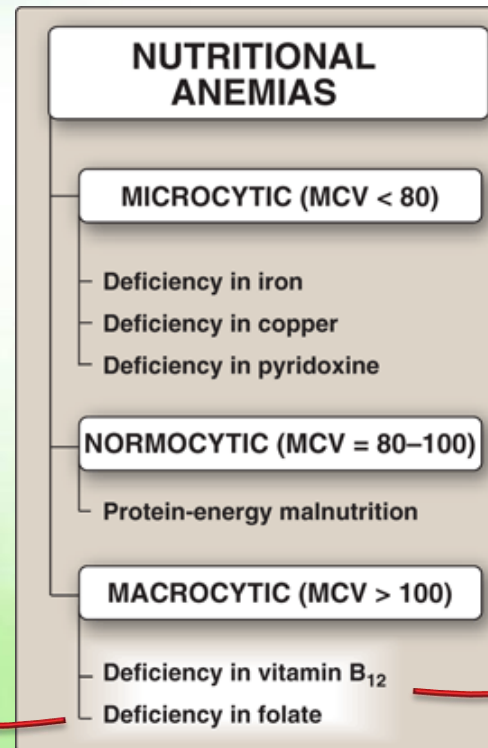
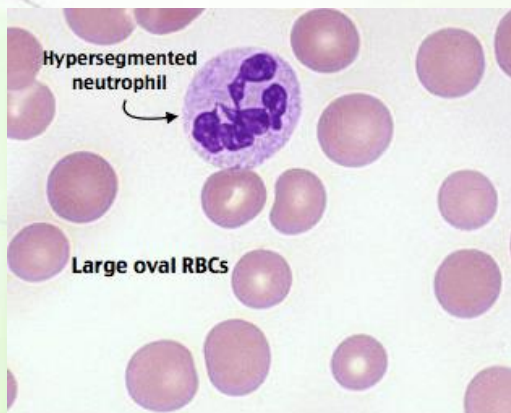


Iron-deficiency anemia

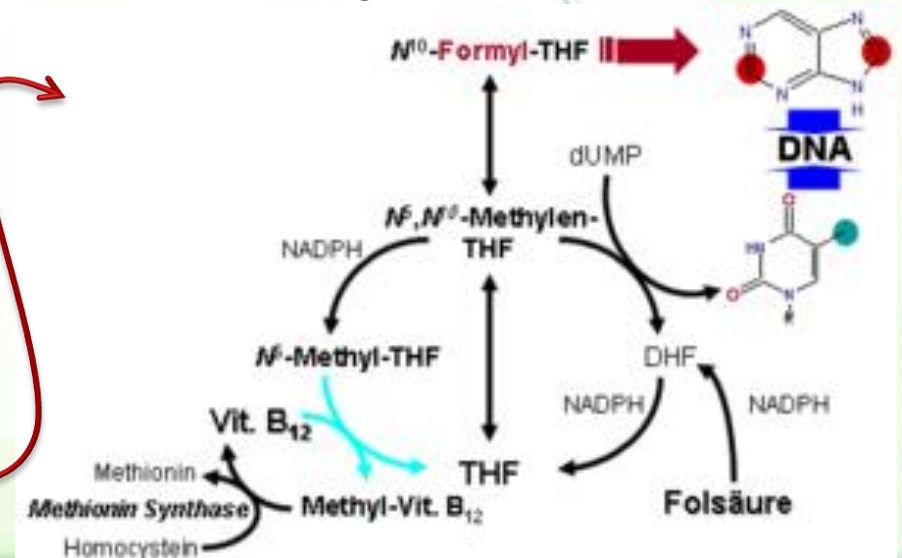


- Anemias are characterized by a deficiency in the number of mature erythrocytes in the circulation, lowering the oxygen-carrying capacity of the blood, causing tissue hypoxia, and clinical symptoms such as fatigue, weakness, increased cardiac output, as well as increased morbidity and mortality.

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



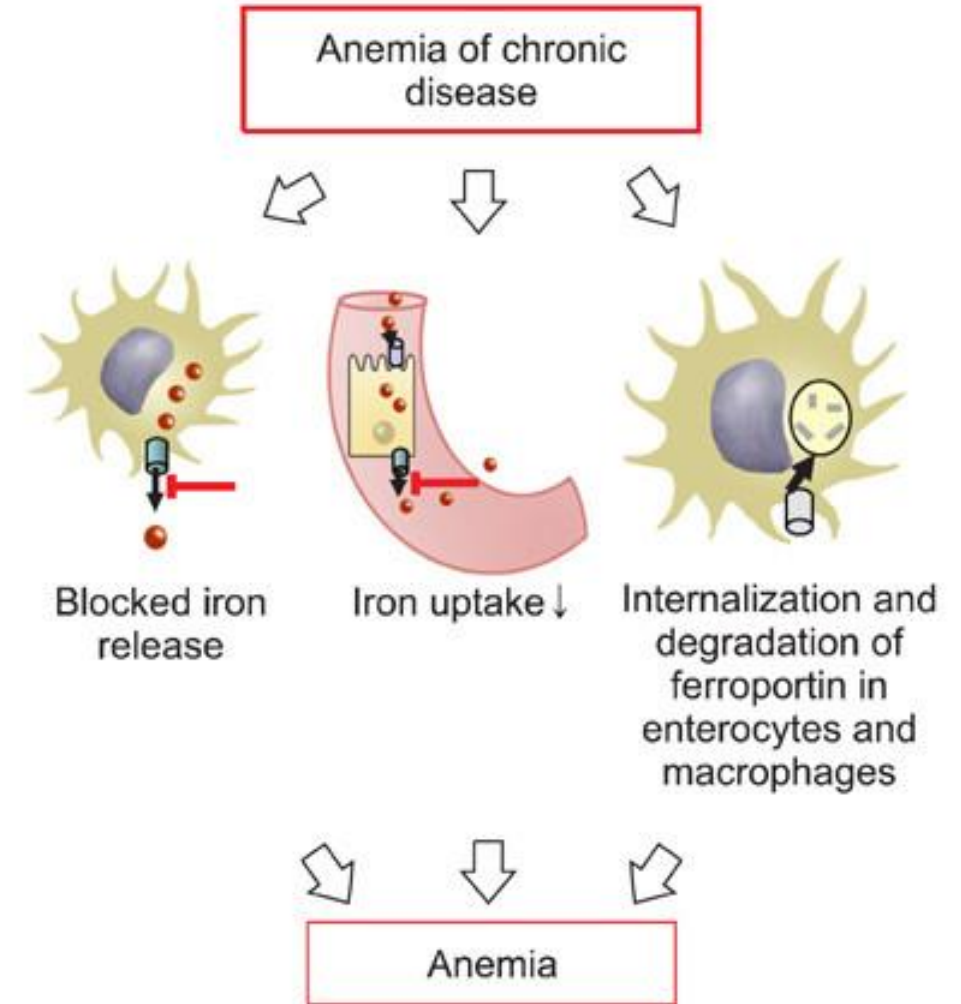
Folate is not regenerated



Anemia of chronic disease



- 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.



Additional molecular consequences of chronic inflammation

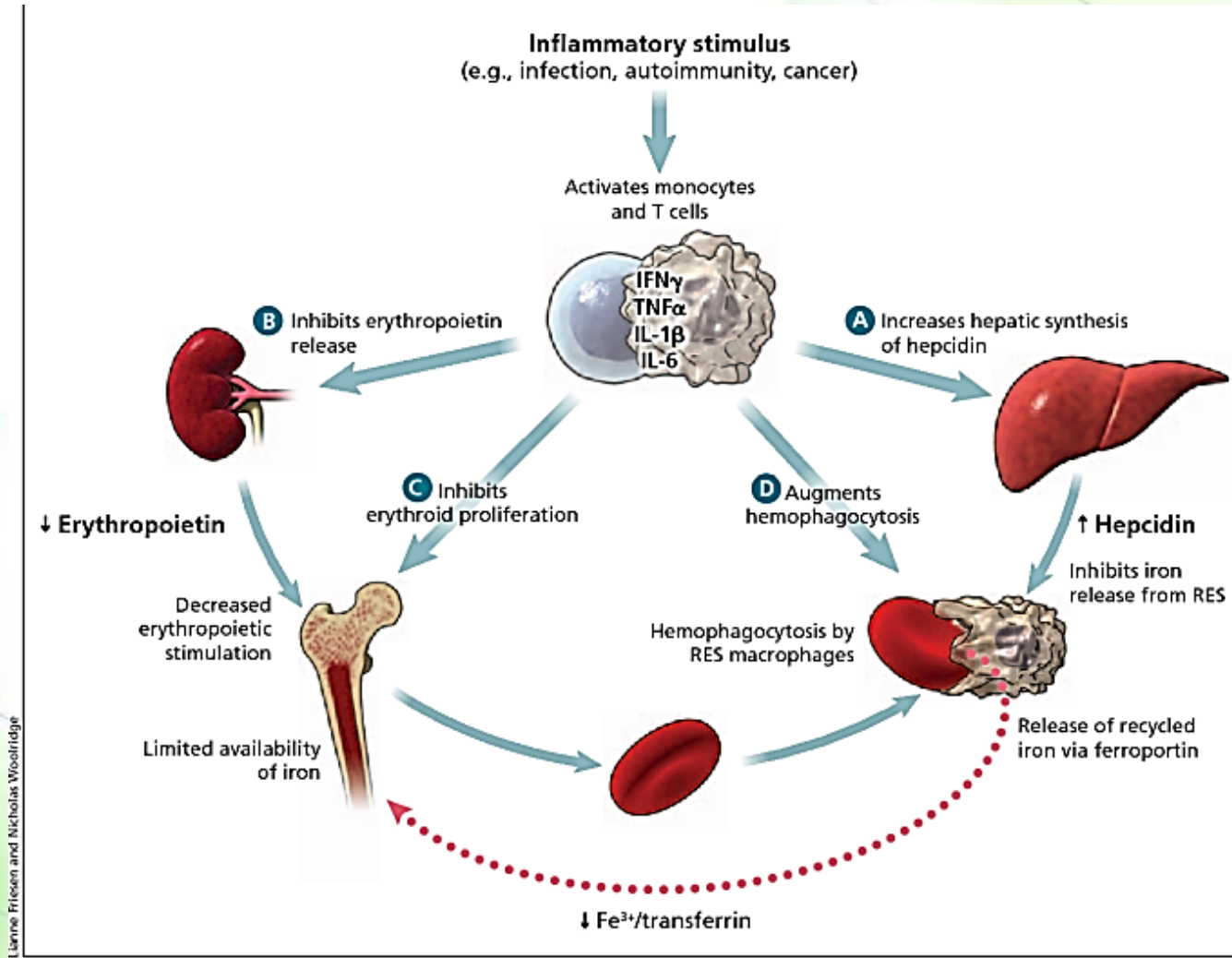


Figure 1: In inflammatory diseases, cytokines released by activated leukocytes and other cells exert multiple effects that contribute to the reduction in hemoglobin levels: (A) Induction of hepcidin synthesis in the liver (especially by interleukin-6, IL-6, along with endotoxin). Hep-