

IRON AND IRON BALANCE: An Overview

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What are some of the vital functions of Iron in the body?

- Some vital functions of Iron:
 - As component of Hb: facilitates O_2 transport in blood (OxyHb),
 - As component of Mb: facilitates O_2 storage in muscle,
 - As component of Cytochromes: facilitates movement of electrons within specific organelles (e.g., Oxidative Phosphorylation in Mitochondria),
 - As component of enzymes: Oxidases, Oxygenases,
 - Examples: Lysosomal enzyme Myelo-Peroxidase required for proper Phagocytosis and Killing of Bacteria by Neutrophils);

How is Iron distributed in the body?

- Iron in the body is distributed or classified as:
 - **Functional Iron** (About 80%):
 - Mainly located in RBC as Hb,
 - Lesser amount in Myoglobin, and Cytochromes
 - **Storage Iron** (About 20%):
 - Primarily as **Ferritin**, some as **Hemosiderin**,
 - **Transport Iron**:
 - Less than 1.0% of body Iron is in plasma, bound to **Transferrin**,
 - Each Transferrin binds 2Fe^{3+} (Ferric) ions;

- Concentration of Iron in Serum/Plasma vary with Age and Sex
 - Males: 18 – 45umol/L
 - Females: 14 – 32umol/L
- Marked **Circadian Rhythm (Diurnal variation)** is observed in Plasma Iron levels in both male and female, with higher values in the morning;

What are some of the dietary sources of Iron?

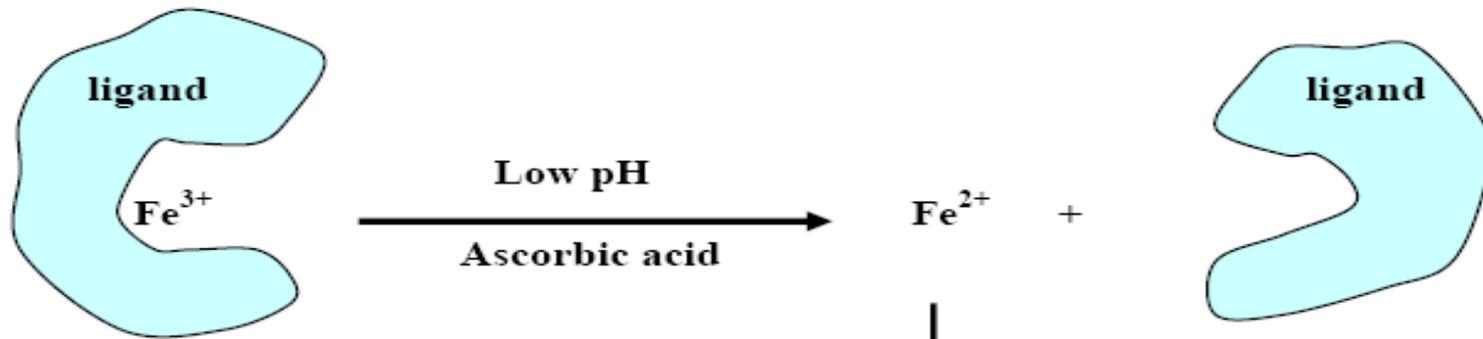
- Normal intake of Iron is about 0.2 – 0.4mmol/day (10 – 20mg/day);
- **Some good dietary sources of Iron:**
 - **Heme Iron is Ferrous Iron (Fe^{2+}):**
 - Liver, Fish, Meat, Egg yolk, Oysters,
 - **Non-Heme Iron is Ferric Iron (Fe^{3+}):**
 - Green leafy vegetables, Fruits, Dried beans

How is dietary Iron absorbed? (Fig. 1)

- Iron is absorbed mainly in Duodenum and Upper Jejunum;
- Dietary Iron is consumed usually bound to ligand;
- **In Stomach: High acidic environment (low pH):**
 - **Non-Heme Iron (Fe^{3+})** is converted to **Heme Iron (Fe^{2+})** releasing the Ligand;
 - Content of stomach with Fe^{2+} then enters Intestine

- In Duodenum: High alkaline environment (High HCO_3^-): Fig. 1:
 - Free Fe^{2+} ions are oxidized to Fe^{3+} ions,
 - Fe^{3+} ions are taken up by Intestinal Mucosal cells,
- **How is Heme absorbed?**
 - Heme is absorbed directly by Mucosal Cells,
 - Fe^{2+} ions then dissociate from Heme,
- **NB:** Absorption of Heme Iron is 2 to 3 times more effective than absorption of Non-Heme Iron;

In the stomach:



In Duodenum:

High pH (in Duodenum Fe^{2+} is converted to Fe^{3+} , which can also be absorbed)

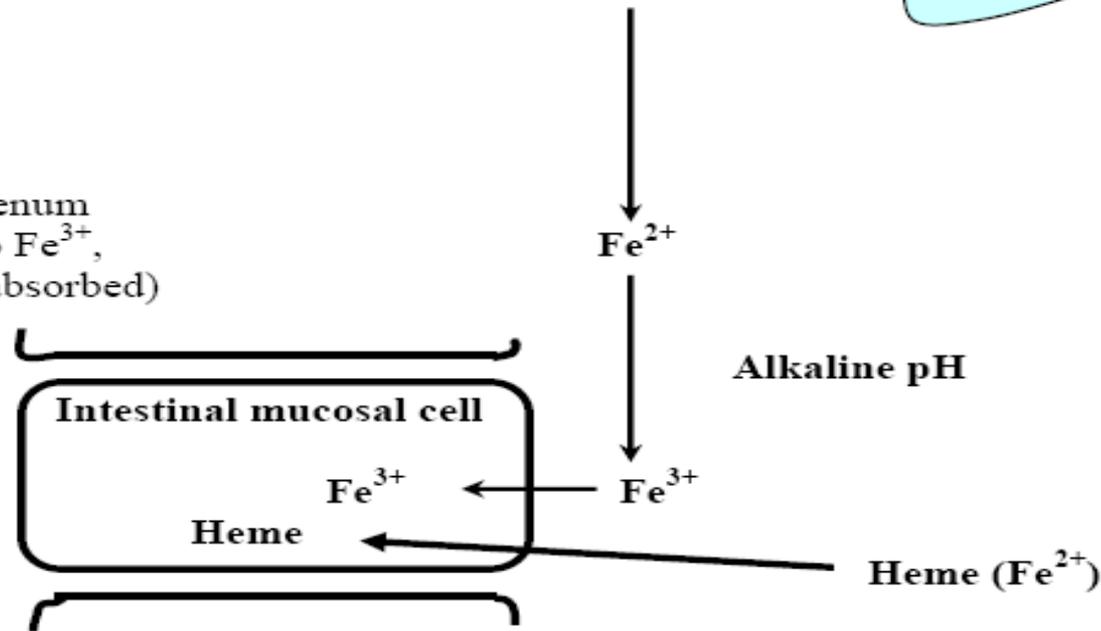


Fig. 1: Simplified Schematic diagram of Absorption of Iron:

Absorption of Fe^{2+} (Heme Iron) is about 2 to 3 times for effective than Fe^{3+} (Non-Heme Iron)

Does pH affect the rate of absorption of Iron?

- Acidic environment is required for efficient absorption of Iron;
- Conditions that interferes with Gastric Acid secretion reduces the rate of Iron absorption (e.g. Antacids)

How is Iron absorbed in the Enterocytes?

- Iron is absorbed via the Enterocytes in Proximal Duodenum;
 - **Ferric-Reductase** located in brush border of Duodenal Lumen **converts Fe^{3+} to Fe^{2+}** ;
 - Divalent Metal Transporter-1 (DMT-1) then transports Fe^{2+} and H^+ into the Enterocyte,
 - **NB:** DMT-1 is not a specific transport for Fe^{2+}

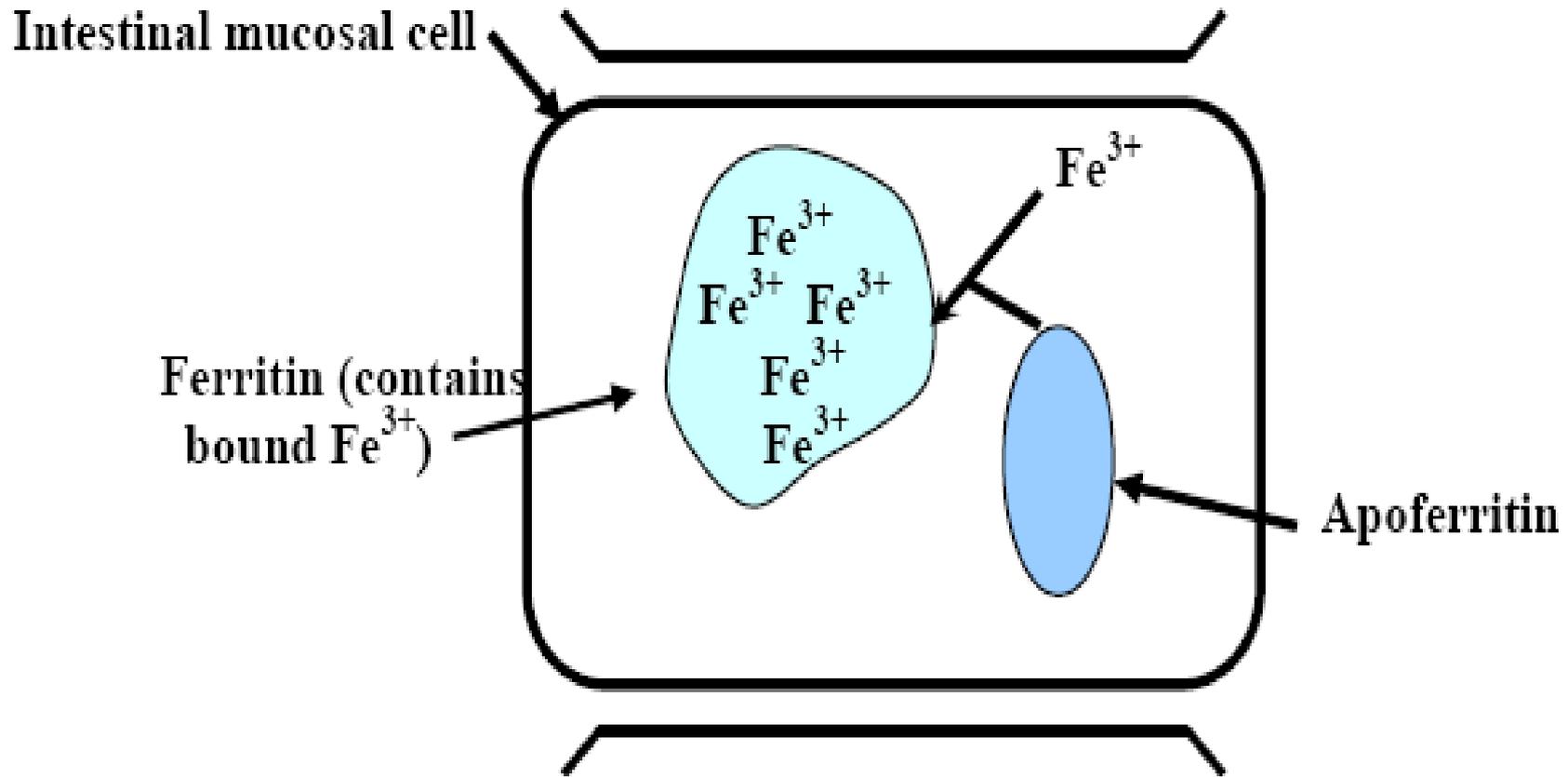
How is absorption of Iron regulated?

- **Apo ferritin** (Iron-binding protein) produced in Intestinal Mucosal cells regulates absorption of Iron



- **In Iron-depleted state:**
 - Apo ferritin level is low, allowing absorbed Fe^{3+} to move freely across the Intestinal Mucosal cell,
 - Fe^{3+} is converted to Fe^{2+} by Ferric Reductase and released into plasma,
 - **In plasma:** Ferroxidase converts Fe^{2+} back to Fe^{3+}
 - Transferrin binds and transports Fe^{3+} in plasma;

- **In Iron Replete state: (Fig. 2)**
 - Increased amount of **Apo ferritin** is produced to **trap Iron as Ferritin**,
 - Apo ferritin binds absorbed Fe^{3+} to form Ferritin, which is retained in the Intestinal Mucosal cells;
- Iron in Ferritin retained by mucosal cells is lost in the Intestinal lumen when the cells are sloughed,
- Mucosal cell retention is influenced by Iron status of the body:
 - It is reduced in Iron depletion,
 - It is Increased in state of Iron overload,



If the body does not need Iron,
The iron is stored as Ferritin in the mucosal cells.
The Iron is lost during when the cells die and slough

Fig. 2: Storage of Iron as Ferritin in Intestinal Mucosal Cells: Iron Replete state

What factors influence the capacity of the body to absorb Iron?

- Factors that can influence Iron absorption from diet:
- **State of Iron Stores:**
 - Amount of Iron in the body;
 - GIT increases absorption when iron store is low and decreases absorption when store is sufficient,
 - Percent of Iron absorbed (Iron Bioavailability) can vary from less than 1% to greater than 50%;
- **Rate of Erythropoiesis:**
 - Increase rate of RBC production can stimulate Iron uptake by several folds;

- **Chemical State of Iron or kind of Iron in the Diet:**
 - Heme Iron (Fe^{2+}) is 2 to 3 times more absorbable than Non-Heme Iron (Fe^{3+}) in Plant-based foods and Iron-fortified foods,
 - Gastric Acid increases uptake of Non-Heme Iron:
 - H^+ ion (low pH,) facilitates conversion of Fe^{3+} to Fe^{2+} before uptake,
 - Duodenal Microvilli contain Ferric Reductase that catalyzes conversion of Fe^{3+} to Fe^{2+} to promote uptake of Fe^{3+} ;

- **Content of diet: (presence or absent of Enhancers and Inhibitors):**
 - Bioavailability of Non-Heme Iron is affected by the kind of other foods Ingested at the same meal;
- **Some Enhancers of Non-Heme Iron absorption:**
 - Vitamin C (Ascorbic Acid and Dehydroascorbate),
 - Reducing agents (e.g., Fructose),
 - Citrate,

- **Some Inhibitors of Iron absorption:**
 - Oxalate,
 - Poly-phenols (in certain vegetables),
 - Tannins (in Tea),
 - Phytates (in Bran),
 - Calcium (in dairy products),
 - Fibers (Vegetarian diets are low in Heme Iron),
 - Antacids;
- **IMPORTANT TO NOTE:**
 - Iron bioavailability in vegetarian diet can be increased by include other sources of Iron and Enhancers of Iron absorption;

What three mechanisms regulate uptake of Iron?

- **Store Regulator:**
 - As body Iron stores fall, the Mucosa is signaled to moderately increase uptake of Iron;
- **Erythropoietic Regulator:**
 - In response to Anemia, Erythroid cells will signal Mucosa to increase Iron uptake more significantly
- **Dietary Regulator:**
 - A short-term increase in dietary Iron may not be absorbed, because the Mucosal cells may have accumulated Iron and “Block” additional uptake,

What is the estimated rate of Iron turnover?

- **Major turn-over of Iron:**
 - Synthesis and Destruction of RBC,
 - **Adult males:**
 - About 95% of Iron for RBC synthesis is recycled from breakdown of RBC, only 5% comes from dietary sources;
 - **Infants:**
 - Estimated to derive about 70% of Iron for RBC synthesis from breakdown of RBC and 30% from the diet;
 - **Adult females:** Estimates not well established

What are some of the sources of Iron loss?

- **Some sources of Iron Loss:**
 - **Adult males & females:**
 - Iron is loss in cells desquamated from Skin and Intestinal Mucosa,
 - Females additional losses via Menstruation, Pregnancy, Delivery and Postpartum,
- **NB:** Women of childbearing age require additional Iron to compensate for menstrual blood loss and for tissue growth during pregnancy;

- **Pathological GIT Iron loss:**

- GIT bleeding may occur in infants and children sensitive to Cow's milk,
- Adults with Peptic Ulcer Disease, Inflammatory Bowel Syndrome, or Bowel Cancer,
- Hookworm infections are associated with GIT blood loss and iron depletion;

IRON STORES

How is Iron stored in the body?

- Iron not needed for functional purposes is stored mainly as soluble protein complex **Ferritin**:
 - Ferritin binds and stores only Ferric (Fe^{3+}) ions,
 - Ferritin is present in:
 - Intestinal Mucosal cells,
 - Bone Marrow,
 - Liver,
 - Spleen, and
 - Skeletal Muscles;

- **When Iron store is adequate:**
 - Large amount of **Apo ferritin** is synthesized to Trap excess Iron within the Mucosal Cell preventing Transfer of Iron to capillary bed;
- **When Iron store is low:**
 - Virtually **no Apo ferritin** is synthesized so as not to compete against the transfer of Iron to Plasma,

- **Excess Iron absorption** (e.g., Hemochromatosis: disorder in Iron metabolism):
 - Body stores of Iron are greatly increased with very high deposit of Iron in many organs:
 - Liver,
 - Pancreas,
 - Skin;
- Much more Ferritin is present in Liver and Spleen

How significant is Transferrin?

- **Transferrin:** major transport protein for Iron in Blood
 - It binds and transport only Fe^{3+} ions,
 - 2Fe^{3+} per Transferrin,
 - Most of the Iron-Binding Capacity of blood plasma is due to Transferrin,
- Transferrin is usually one-third (33%) saturated with Iron,
- Transferrin saturation below 15% may indicate Iron deficiency with some degree of clinical effect expected;

- Higher than 33% saturation of Transferrin is a sensitive marker for Iron overload,
- Plasma level of Transferrin may reduce during:
 - Protein-energy malnutrition,
 - Acute-phase response,
 - Infections,
 - Neo-plastic disease,
 - Chronic Liver disease;

What is the role of Ceruloplasmin in Transport of Iron in Plasma?

- **Ceruloplasmin (Ferroxidase):** Copper-containing enzyme involved in Iron transport in blood plasma;
- Sequence of action is as follows:
 - Ferric Reductase converts Fe^{3+} stored by Ferritin to Fe^{2+} , which is then released from Mucosal Cell,
 - Fe^{2+} crosses the plasma membrane into the blood,
 - In the blood, **Ceruloplasmin** catalyzes conversion of **Fe^{2+} to Fe^{3+}** ;
 - **Transferrin** then binds and transports Fe^{3+} in blood
- **See Fig. 3:**

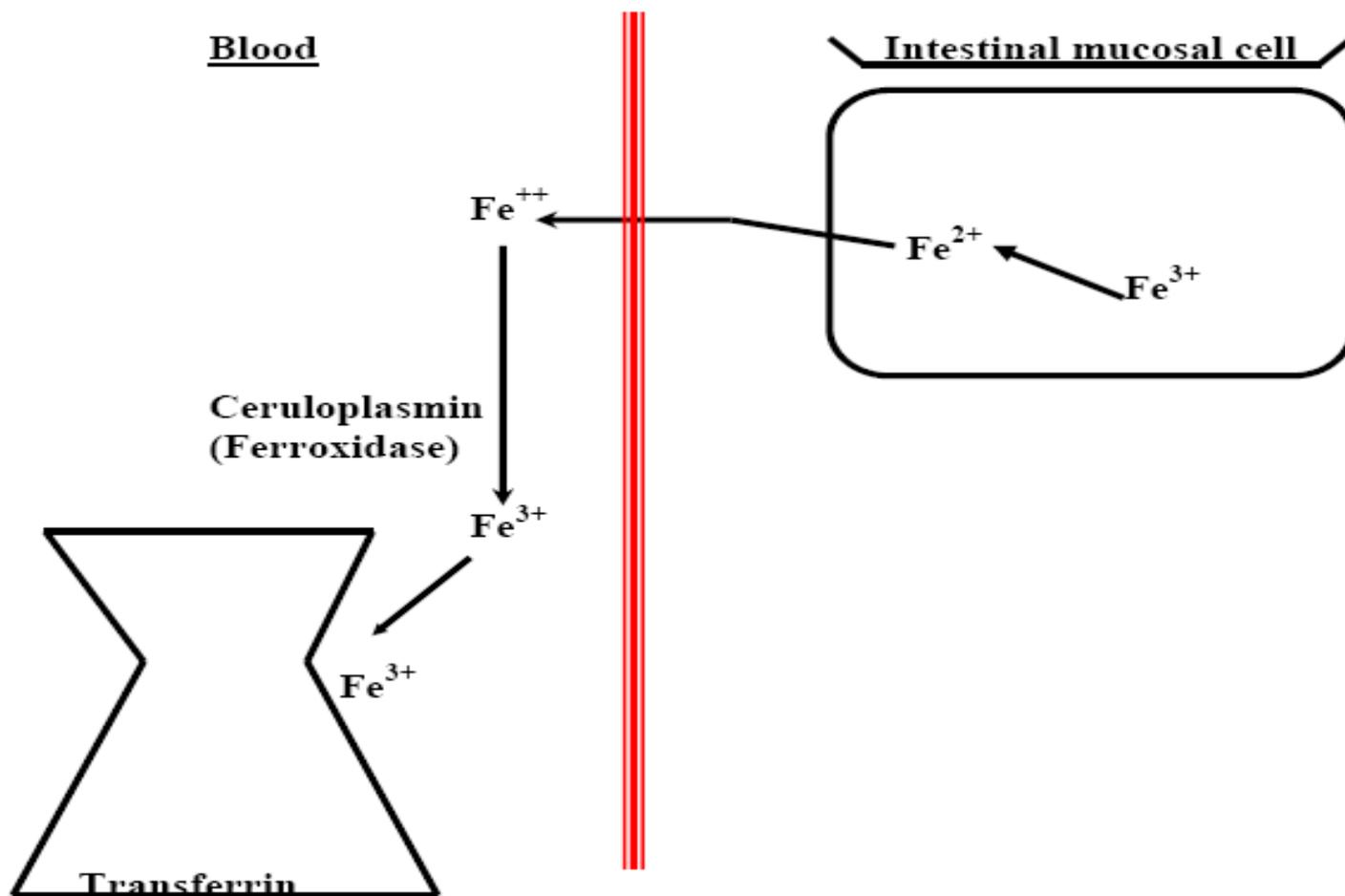


Fig. 3: Role of Ceruloplasmin in transport of Iron by Transferrin in plasma

Ferric Reductase converts Fe^{3+} stored by Ferritin to Fe^{2+} , which is then released from Mucosal Cell
 Fe^{2+} crosses the plasma membrane into the blood
 In the blood Ferroxidase (Ceruloplasmin) a copper-containing enzyme converts Fe^{2+} to Fe^{3+}
 Transferrin then binds and transports Fe^{3+} in blood

SUMMARY OF IRON METABOLISM

- **Schematic diagram in Fig 5 gives a brief overview of Iron metabolism**

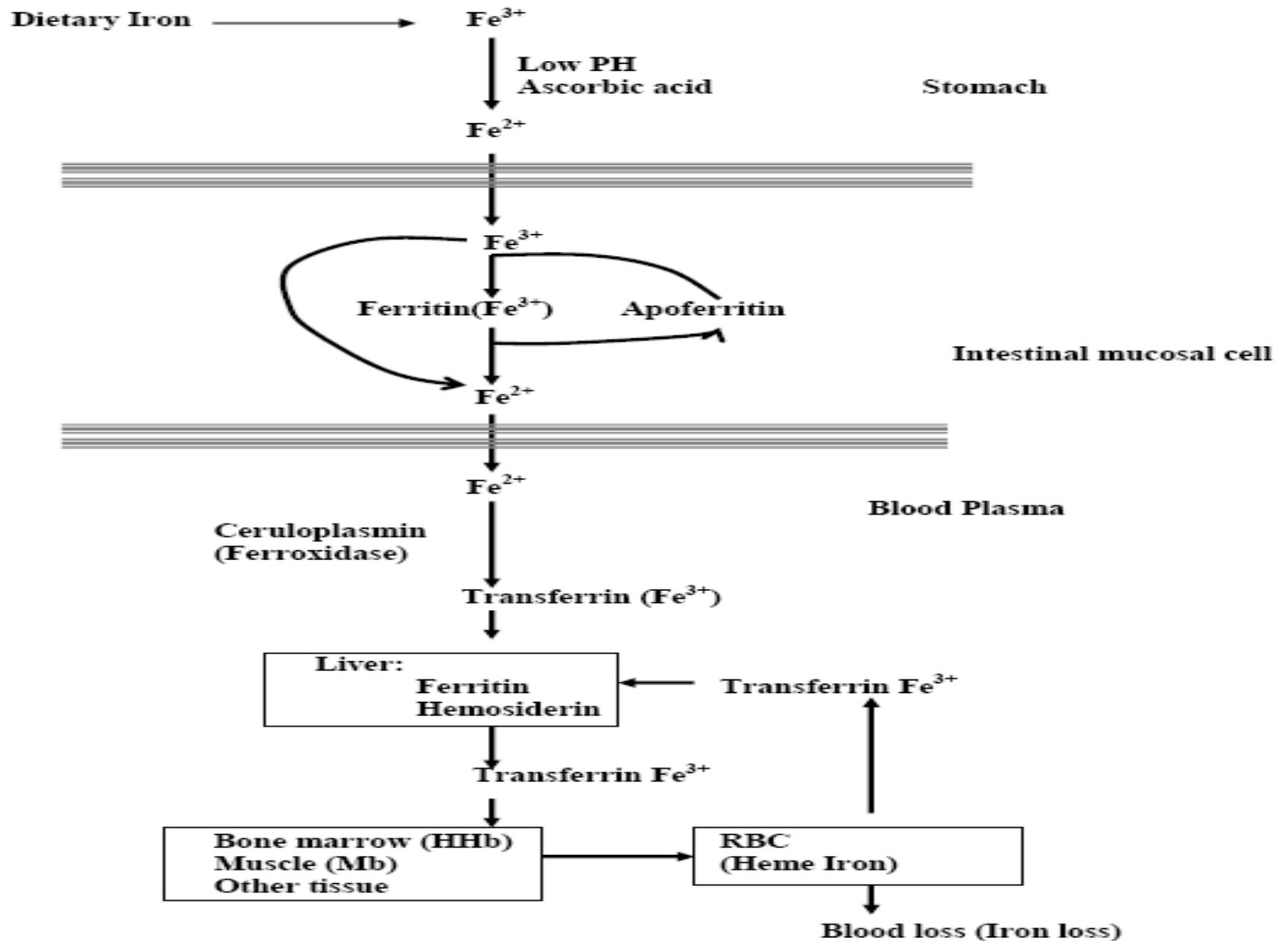


Fig. 5: Schematic representation of an Overview of Iron Metabolism

MANIFESTATIONS OF IRON DEFICIENCY

What is Anaemia?

- Anaemia is indicated when Hb level falls below recommended cutoff points;
- Hb cutoff points recommended by WHO:
 - Pregnant women: **Hb \geq 110.0g/L (11.0g/dL)**,
 - Children 6 to 59 months: **Hb \geq 110.0g/L (11.0g/dL)**
 - Non-pregnant women: **Hb \geq 120.0g/L (12.0g/dL)**
 - Men: **Hb \geq 130.0g/L (13.0g/dL)**

- **IMPORTANT TO NOTE:**
- Hb cutoffs can be affected by Altitude and Race,
- Anaemia can be diagnosed by measuring:
 - Hb concentration in blood or
 - Proportion of RBC in Whole blood (Hematocrit)
- **What Hb cutoff points are used to indicate Anaemia in PNG?**

What is Iron deficiency?

- Iron deficiency is a concept that is difficult to define,
- During adequate nutrition, Iron is stored in tissues and is used when dietary intake of Iron is inadequate or bioavailability of Iron is low,
- Iron stored mainly in the Liver is an Index of Iron Nutritional Status,
- **Depletion of Iron Store in the body constitutes Iron Deficiency;**

- Iron deficiency is a Spectrum ranging from:
 - **Iron depletion**, which causes no Physiological Impairments, to
 - **Iron-deficiency Anaemia**, which affects the functioning of several organ systems;
- **Iron deficiency usually occurs in three Sequentially developing stages;**

What are the Three Sequential stages of Iron deficiency?

- **First Stage: Depleted Iron Stores:**
 - Amount of Stored Iron is reduced, (**Serum Ferritin < 12ug/L**), but the amount of Functional Iron may not be affected;
 - No stored iron is available to mobilize if body requires more Iron,
 - **Hb level remains above Cutoff Levels, thus Anaemia is absent;**

- **Limitations in Diagnosis of the First Stage:**
 - Ferritin cannot be used to accurately assess depleted Iron stores in individuals with poor health, **Why?**
 - Because Ferritin is an **Acute-phase reactant**,
 - Serum Ferritin levels increase during Subclinical and Clinical Infectious and Inflammatory diseases,

Second Stage: Iron Deficient Erythropoiesis

- At this stage Iron stores are depleted, Transport Iron is significantly reduced,
- Hb level remains above Cutoff Levels, thus Anaemia is absent,
- Concentration of Transferrin Receptors are increased,
- Shortage of Iron results in increased free Erythrocyte Protoporphyrin concentration in young RBC,

Third Stage: Iron Deficiency Anaemia (IDA)

- IDA is the most severe form of Iron Deficiency,
 - Iron store is depleted,
 - There is significant reduction in Iron transport,
 - Iron supply is inadequate for Hb synthesis,
- Hb level falls below Cutoff Levels, thus Anaemia;
 - Shortage of Iron leads to underproduction of Iron-containing functional compounds, including Hb,
 - In Iron-deficiency Anemia, RBC are Microcytic and Hypochromic,

What are some consequences of Iron deficiency and IDA?

- **In Children IDA can cause:**
 - Developmental delays,
 - Behavioral disturbances,
 - Decreased motor activity,
 - Decreased attention to tasks,
- Developmental delays may persist past school age (5 years) if Iron deficiency is not fully reversed
- **IDA** contributes to **Lead poisoning** in children by increasing the ability of the GIT to absorb heavy metals, including lead;

- **Among pregnant women:**
 - IDA during first two trimesters of pregnancy is associated with:
 - Twofold-increased risk for preterm delivery
 - Threefold increased risk for delivering low-birth weight baby,
- Iron deficiency is associated with decreased Immuno-competence,

LABORATORY TEST FOR IRON STATUS

What are some of the lab tests used to assess Iron status?

- Iron status can be assessed by several lab tests, however, **no single biochemical test is accepted for diagnosing Iron deficiency;**
- Some Biochemical tests can be used to detect earlier changes in Iron Status;
- **Biochemical tests** include the following:
 - Erythrocyte Protoporphyrin Concentration,
 - Serum Ferritin Concentration,
 - Concentration of Transferrin Receptors,
 - Transferrin Saturation,

- Hematological tests based on characteristics of RBC (Hb level, Hematocrit, Mean Cell Volume or Mean Corpuscular Volume), are generally more available and less expensive than Biochemical tests;

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