((Anemia))

- Anemia: ↓ in circulating RBCs thus patients will result with hypoxia (↓ O_2 delivery to tissues)
- Presentation:
  - → Weakness, fatigue, dyspnea and headache
  - → Pale conjuctiva and skin
  - → Angina (due to ↓ delivery to the heart especially with pre-existing CAD)
- How to confirm anemia?
  - → Hb, hematocrit & RBC count
- Definition of anemia (according to gender):
  - → Males: < 13.5 g/dl
  - → Females: < 12.5 g/dl
- Classification of anemia (based on MCV):
  - → Microcytic (cells are small) = MCV < 80
  - → Normocytic (normal size) = MCV 80-100
  - → Macrocytic (cells are large) = MCV > 100

* RBCs: carrying O_2 within hemoglobin
* MCV: Mean corpuscular Volume → estimating size of RBC
* Normal MCV: 80-100

((Microcytic Anemias))

- Microcytosis: occurs due to an extra-division of erythroblasts (why?) → because there is ↓ Hb formation & RBCs will divide further to keep a normal concentration of Hb within them
- Hb = heme + (globin) → thalassemia results in ↓ globin chain synth.
  - ↓
  - (iron) + (protoporphyrin) → sideroblastic anemia → ↓ protoporphyrin
    - ↓
    - * iron-deficiency results in ↓ heme → ↓ Hb → microcytic anemia
    - * Anemia of chronic diseases also lead to ↓ Fe

* Erythroblast: it is a large cell from which RBCs will be produced after certain number of divisions
* iron from:
  - → meat → easily absorbed
  - → vegetable

absorption in duodenum by enterocytes → transported to blood by the carrier ferroportin → then, in the blood, iron will bind to transferrin → live & bone marrow macrophages (for storage bound with ferritin)

[1] Iron-deficiency anemia:

* Cause: lack of iron in the body

((Microcytic Anemias))

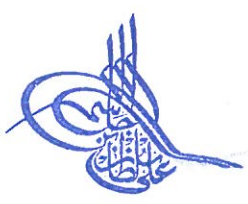
[1] Iron-deficiency Anemia (continued):

* Lab measurments of iron status:
  - serum iron
  - TIBC: indicating how much transferrin present in plasma
  - % saturation: how transferrin bound to iron
  - serum ferritin: how much iron is stored
* Iron deficiency is caused by dietry lack or blood loss:
  - Infants: breast milk contains ↓ iron
  - Children: poor diet (malnutrition)
  - Adults:
    - males: peptic ulcer disease
    - females: menorrhagia or pregnancy
  - Elderly:
    - Developed countries: colon polyps / cancer
    - Developing countries: hookworms
      - Necator americanus
      - Ancylostoma duodonale
* Stages of iron deficiency:
  - Storage iron is depleted (↓ ferritin / ↑ TIBC) ⟹ why? ⟹ liver recognizes that storage is depleted thus pumping more transferrin to find more iron.
  - Serum iron is depleted
    - serum iron ↓
    - % saturation ↓
  - Normocytic anemia
  - Microcytic, hypochromic anemia: smaller cells with less color (expanded central area of pallor in RBCs).
* Clinical features:
  - Anemia (with its features)
  - Koilonychia (spoon-shaped nails)
  - Pica
* Lab findings:
  - Microcytic hypochromic anemia
  - ↓ ferritin, ↑ TIBC
  - ↓ serum iron, ↓ % saturation
  - ↑ FEP

* Fe^{2+} is more easily absorbed into the body ⟶ this form of iron is maintained by acidity. If there is gastrectomy ⟶ ↓ acid ⟶ ↓ Fe^{2+}

* Pica: chewing abnormal things (ice, dirt...etc)

* FEP: Free erythrocyte protoporphyrin. It will be increased in iron-deficiency anemia because there is no enough iron to bind to protoporphyrin and generate heme.



((Microcytic Anemias))

[1] Iron-deficiency anemia (continued):

* Treatment: iron sulfate (supplement)
* Plummer-vinson syndrome:
  - → Iron-deficiency anemia
  - → Esophageal web (dysphagia)
  - → Red-beefy tongue (glossitis)

[2] Anemia of chronic disease:

* Associated with:
  - → chronic inflammation: in which there will production of Hepcidin which locks iron in its storage sites (so it cannot be used)
  - → Cancers
* Lab findings:
  - → ↑ ferritin (because stored iron is not used), ↓TIBC
  - → ↓ serum iron, ↓ % saturation
  - → ↑FEP
* Treatment: of underlying condition

[3] Sideroblastic anemia:

* Cause: There is a defect in protoporphyrin synthesis
* When there is failure of production of protoporphyrin → iron will get trapped in mitochondria → accumulation → formation of a ring around the nucleus of the cells
* Sideroblastic anemia can be:
  - → Congenital: defect in ALA synthase (key-rate limiting enzyme).
  - → Acquired:
    - → Alcoholism: poison destroying mitochondria
    - → Lead poisoning: which can lead to:
      - → ALA dehydrogenase denaturation
      - → Ferrochelatase denaturation
    - → Vitamin B6 deficiency: a cofactor for ALA synthase.
* Laboratory findings:
  - → ↑ ferritin, ↓TIBC
  - → ↑ serum iron, ↑ % saturation

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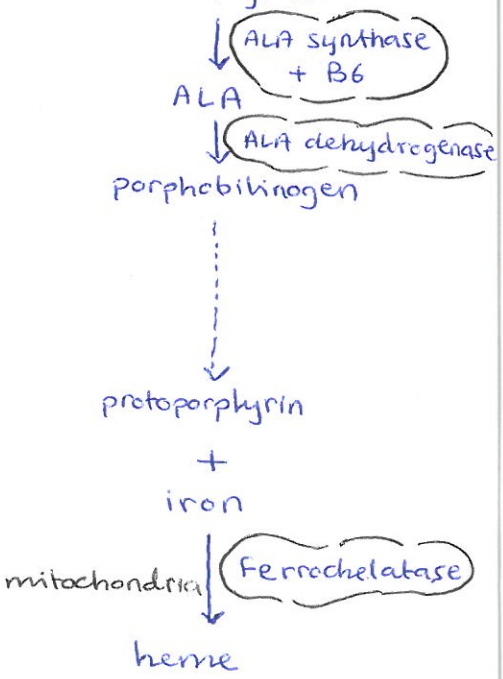
* Bacteria requires iron for their survival thus when there is chronic inf. the body produces hepcidin to hide iron from bacteria (which is not even present! ;)

* Production of protoporphyrin



* Vitamin B6 deficiency: in isoniazid treatment.

((Microcytic Anemias))

4 Thalassemia (↓ production of globin chains)
- divided into:
  - α: ↓ production of α chains
  - β: ↓ production of β chains

* α-thalassemia:
  - Cause: gene deletion
    - 1 gene deleted: asymptomatic
    - 2 genes deleted: mild anemia with slightly ↑ RBC count. Notice that cis deletion (2 copies on same chromosome) is worst than trans deletion (1 copy from each chromosome) due to ↑ risk of sever thalassemia in offspring
    - 3 genes deleted: severe anemia with formation of tetramers of β-chain (β_4) = HbH → damaging RBCs
    - 4 genes deleted: formation of γ-chain tetramers (γ_4) = Hb Bart → hydrops fetalis

* β-thalassemia:
  - Cause: gene mutation
    - β^0: absent β-chain production
    - β^+: ↓ β-chain production
  - Types:
    - β-thalassemia minor (β/β^+):
      - Mildest form of disease (asymptomatic)
      - Microcytic hypochromic anemia
      - Target cells on blood smear
      - ↑ HBA_2
    - β-thalassemia major (β^0/β^0)
      - Most sever form of disease
      - No problem in fetus (because HbF doesn't contain β-chains) but will present with severe anemia few months after birth
      - (α_4) is formed Leading to ineffective erythropoiesis & extravascular hemolysis (within splenic macrophages)

→ Clinical features:
1. Skull X-ray: hair on end appearance
2. Chipmunk face
3. Hepatosplenomegaly

* Abnormality of globin chain: example → sickle cell anemia
* Carriers of thalassemia are protected against plasmodium falciparum malaria
* Normal types of Hb:
  - Fetal: $\alpha_2\gamma_2$
  - HbA: $\alpha_2\beta_2$
  - HbA_2: $\alpha_2\delta_2$
* There are 4 copies of α-chain present on chromosome (16)
* cis deletion → Asia
  trans deletions → Africa

* There are 2 copies of β-chain present on chromosome (11)
* Normal RBC:

* Target cell:

((Microcytic Anemias))

4 Thalassemia (continued)

β-thalassemia major (β^0/β^0) continued—

- Chronic blood transfusions are needed & this can lead to secondary hemochromatosis
- Microcytic hypochromic anemia with target cells
- No HbA, ↑ HbA_2 and ↑ HbF

((Macrocytic Anemia))

- Cause: Folate / vitamin B12 deficiency (megaloblastic anemia) → Therefore, production of DNA precursor molecules will be reduced resulting in:
  - Megaloblastic anemia: large RBCs
  - Hypersegmented neutrophils: > 5 lobes
- Other causes of macrocytic anemia include:
  - Alcoholism
  - Liver disease
  - Drugs (5-FU)

  } hypesegmented neutrophils will not be seen here

- Folate:
  - Obtained from: green vegetables and fruits
  - Absorbed in: jejunum
  - Developing within months due to minimal storage
  - Cause of folate deficiency:
    - Malnutrition (alcoholics and elderly)
    - ↑ demand (pregnancy, cancer & hemolytic anemia)
    - Folate antagonists (e.g. methotrexate)
  - Clinical & laboratory findings:
    - Macrocytic RBCs with hypersegmented neutrophils
    - Glossitis
    - ↓ serum folate
    - ↑ serum homocysteine (because there is no passage of methyl group to homocysteine and conversion into methionine)
    - Normal methylmalonic acid

* Macrocytic: because one less division of erythroblast occurs resulting in cells with bigger size
* Folate:
  It enters body as tetrahydropholate → methylated → mTHF

  ⇓

  vitamin B12 will take the methyl group allowing THF to participate in DNA production

  ⇓

  vitamin B12 will pass methyl group to homocysteine to be converted into methionine
* Methotrexate: it inhibits DHF reductase
* methylmalonic acid is normally converted into Succinyl CoA through vitamin B12

((Macrocytic Anemias))

- Vitamin B12 (less common than folate deficiency):
  - * It takes years to develop vitamin B12 deficiency due to large hepatic stores
  - * Causes of vitamin B12 deficiency:
    - → Pernicious anemia: in which there is autoimmune destruction of gastric parietal cells → ↓ Intrinsic factor → ↓ absorption of vit B12
    - → Pancreatic insufficiency: vitamin B12 will not be cleaved from R-bineder
    - → Damage to terminal ileum (Crohn's disease)
    - → Dietary deficiency (very rare except in vegans)
  - * Clinical and lab findings:
    - → Macrocytic anemia with hypersegmented nuclei
    - → Glossitis
    - ** → Subacute combined degeneration of spinal cord: due to accumulation of methyl-malonic acid (cannot be converted to succinyl CoA)
    - → ↓ serum vitamin B12
    - → ↑ serum homocysteine
    - → ↑ methylmalonic acid

* Vitamin B12 from liver
↓
entering body
↓
vitamin B12 – R binder
↓
in small intestine it will be cleaved by pancreatic proteases into vitamin B12
↓
vitamin B12 + (IF)
↓
produced by parietal cells in stomach
↓
absorption in terminal ileum

((Normocytic Anemia))

- Cause:
  - → ↑ peripheral destruction
  - → Underproduction
  - * You can distinguish between these two by reticulocyte count
    - → if corrected reticulocyte count is > 3%. → there is no problem with production of RBCs
    - → if corrected reticulocyte count is < 3%. → there is underproduction.
  - * Peripheral destruction of RBCs (hemolysis):
    - → Extravascular: by reticulo endothelial system outside the blood vessel
    - → Intravascular: RBCs destroyed within blood vessels

* Reticulocytes: young RBCs, large, with blue cytoplasm (due to presence of RNA).
* Normal reticulocyte count 1-2%.
* When there is anemia reticulocyte count must be > 3%.
* A decrease in total RBCs falsely elevates percentage of reticulocyte
⇓
* reticulocyte count must be corrected by multiplying it by $\left(\frac{Hct}{45}\right)$

((Normocytic Anemia))

- Extravascular hemolysis:
  - → destruction by reticuloendothelial system (macrophages of spleen, liver & lymph node)
    - ↓
    - Hemoglobin released
      - → globin → broken down to amino acids
      - → heme
        - → iron
        - → protoporphyrin
          - ↓
          - unconjugated bilirubin
          - ↓
          - bound to serum albumin
          - ↓
          - conjugated in liver
          - ↓
          - excreted in bile
  - → Clinical and laboratory findings:
    - → Anemia with splenomegaly
    - → Jaundice due to accumulation of unconjugated bilirubin resulting from excessive hemolysis
    - → ↑ risk of bilirubin gallstones
    - → Corrected reticulocyte count > 3%.

- Intravascular hemolysis:
  - → Destruction of RBCs within blood vessels
    - ↓
    - hemoglobin released directly into the blood
    - ↓
    - it will bind to haptoglobin ⟹ Therefore, Free haptoglobin in serum will be decreased. Eventually all haptoglobin will be used → hemoglobinemia results → leaking to urine → hemoglobinuria
    - ↓
    - go to spleen to be reprocessed
  - → Clinical and laboratory findings:
    - → Hemoglobinuria
    - → Hemosideriuria
    - → Hemoglobinemia
    - → ↓ serum haptoglobin

((Normocytic Anemia: Extravascular Hmolysis))

1 Hereditery spherocytosis:

- Intrerited defect of RBC cytoskeleton - membrane tethering proteins:
  - Spectrin
  - Ankyrin
  - Band 3.1
- RBCs are spherocytic (instead of being biconcave) with loss of central pallor
- Spherocytes will be destroyed by splenic macrophages
- Clinical and laboratory findings
  - Spherocytes with central pallor
  - ↑ RDW, ↑ MCHC
  - Splenomegaly, jaundice with unconjugated bilirubin and increased risk of bilirubin gallstones
- Diagnosis:
  - Osmatic fragility test: spherocytes are easily burst in hypotonic solution
- Treatment: splenectomy
  - Howell-Jolly bodies (fragments of RBCs nuclei) will emerge on blood smear ⟹ Normally, these fragments are removed by the spleen

2 Sickle-cell disease:

- Autosomal recessive; mutation in β-chain of Hb
  - At the 6th amino acid position, glutamic acid (hydrophilic) will be replaced with valine (hydrophobic) ⟹ **HbS**
    - ⇓
    - it polymerizes when deoxygenated
    - ↓
    - resulting in sickle cells
    - 
- ↑ risk of sickling with:
  - Hypoxemia
  - Dehydration
  - Acidosis

* Carriers of sickle cell disease are protected from falciparum malaria.

[2] Sickle cell disease (continued):

- Presence of HbF protects against sickling ⟹ This explains why the disease will appear after 6 months of birth when HbF is no more the predominant hemoglobin
  Patients are treated with hydroxyuria which ↑ HbF
- Patients will have: anemia, jaundice with unconjugated hyperbilirubinemia and increased risk for bilirubin gallstones
  - Some of RBCs might break down within blood vessels with features of intravascular hemolysis such as ↓ haptoglobin and target cells on blood smear
- Irreversible sickling leads to vaso-occlusion
  - Dactylitis: swollen hands and feet due to vaso-occlusive infarcts of bones. It is a common presenting sign in infants
  - Autosplenectomy: infarcted, shrunken, fibrotic spleen ⟶ leading to complications such as:
    - ↑ risk of infection with encapsulated organisms (children).
    - ↑ risk of Salmonella paratyphi osteomyelitis
    - Howell-Jolly bodies on blood smear
  - Acute chest syndrome
    - dyspnea, chest pain and lung infiltrates ⟶ precipitated by pneumonia
  - Pain crisis
  - Renal papillary necrosis
- Sickle cell trait (carrier): HbA / HbS ⟹ percentage of HbS is < 50%. Thus no sickling will occur

[3] Hemoglobin C (Less common than sickle cell disease):

- Autosomal recessive; mutation of β-chain of Hb
  - glutamic acid is replaced by lysine
  - HbC crystals are detected on blood smears

* Sickle cell disease:
  90% HbS, 8% HbF, 2% HbA_2 (with no HbA)
* Sickle cell trait:
  55% HbA, 43% HbS, 2% HbA_2

((Normocytic Anemia: Intravascular Hemolysis))

1. G6PD deficiency:
   - → X-linked recessive disorder with reduced half-life of G6PD. Therfore, cells will not be protected against oxaditive stress
   - → There are two major variants of the disease:
     - → African: mild
     - → Mediterranean: severe
   - → Triggers of oxaditive stress:
     - → Infections
     - → Drugs (primaquine)
     - → Fawa beans
     - oxidative stress will result in percipitation of Hb as Heiz bodies → which will be removed by splenic macrophages resulting in bite cells

   - → Presentation: hemoglobinuria & back pain

2. Immune hemolytic anemia:
   - → Antibody-mediated destruction of RBCs by IgG or IgM
     - → IgG-mediated disease: is an extravascular hemolysis
       - * IgG will bind to surface of RBCs in warm temperature → IgG bound to membrane of RBCs will be eaten by splenic macrophages resulting in spherocytes
       - * This is associated with SLE, CLL and certain drugs
       - * Treatment:
         - → Splenectomy
         - → Cessation of drug if it is caused by one
         - → Steroids
         - → IV immunoglobulin
     - → IgM-mediated disease: is an intravascular hemolysis → Associated with: ① Mycoplasma pneumonia ② Infectious mononucleosis
       - * IgM will bind to surface of RBCs in cold temperature → fixing complement → and leading to intravascular hemolysis

* Reduced glutathione is important in protecting RBCs from damage by ROS
  ↓
  NADPH is needed to be produced by (G6PD) so converting oxidized glutathione into reduced glutathione
* Patients with G6PD deficiency are protected against falciparum malaria
* Heinz preparation is used to screen for the the disease
* Enzymatic studies confirm the deficiency but they must be done after resolution from the hemolytic episode.

((Normocytic Anemia: Intravascular hemolysis))

[2] Immune hemolytic anemia (continued):
- Coomb's test is used to dianoge this disease. There are two types of this test:
  - Direct: you add anti-IgG1 to patient's RBCs → if IgG present on surface of RBCs → agglutination will occur
  - Indirect: you add anti-IgG1 to patient's RBCs + serum → if IgG present in serum → agglutination will occur

[3] Microangiopathic hemolytic anemia:
- There are small blood vessels within which a thrombus will develop → blood vessel becomes narrower → when RBC tries to pass → it will be sheared resulting in schistocytes

- Conditions in which you see this disease:
  - Thrombotic thrombocytopenic purpura (TTP)
  - Hemolytic uremic Syndrome (HUS)
  - DIS

[4] Malaria:
- Infection of RBCs and Liver by: Plasmodium
- Transmission: Female Anopheles mosquito
- RBCs will rupture as part of plasmodium life cycle → resulting in intravascular hemolysis and cyclical fever:
  - P. falciparum: daily fever
  - P. vivax & ovale: fever every other day

((Anemia due to underproduction))

- Decreased production of RBCs from bone marrow with ↓ corrected reticulocyte count
- Causes:
  - Microcytic anemia (ex. iron-deficiency anemia)
  - Macrocytic anemia (ex. folate/vit B12 anemia)
  - Renal failure (↓ erythropoietin)
  - Damage to RBC precursor cells in bone marrow (parvovirus B19 infection)