# INTRODUCTION TO ANEMIA

Ahmad T. Mansour
American Board of Pathology (AP/CP)
American Board of Hematopathology
Jordanian Board of Pathology

- Introduction to anemia including classification
- Anemia of diminished production
- Anemia of RBC loss (hemolytic and nonhemolytic) 1
- Anemia of RBC loss (hemolytic and nonhemolytic) 2
- Polycythemia

Ah.mansour@ju.edu.jo

# Introduction

1. Which one of the following parameters is used to assess the volume of the RBC?
   - A. MCV
   - B. MCHC
   - C. RDW
   - D. Hematocrit
   - E. MCH

2. Which of the following is most helpful in the workup for immune hemolytic anemia?

A. Iron indices

B. Coombs test

C. Hemoglobin electrophoresis

D. Bone marrow examination

3. All of the following are examples of microcytic anemia, except:

A. Iron deficiency anemia
B. Thalassemia
C. Megaloblastic anemia
D. Lead poisoning
E. Sideroblastic anemia

4. All the following are associated with increased reticulocyte count, except:

A. Immune hemolytic anemia

B. Spherocytosis

C. Microangiopathic hemolytic anemia

D. Aplastic anemia

E. G6PD deficiency

5. All the following are clinical manifestations of anemia of diminished production, except:

A. Skin pallor
B. Shortness of breath
C. Gallbaldder stones
D. Muscle weakness
E. Confusion

# Introduction

-Definition of anemia.

-Pathophysiology of anemia.

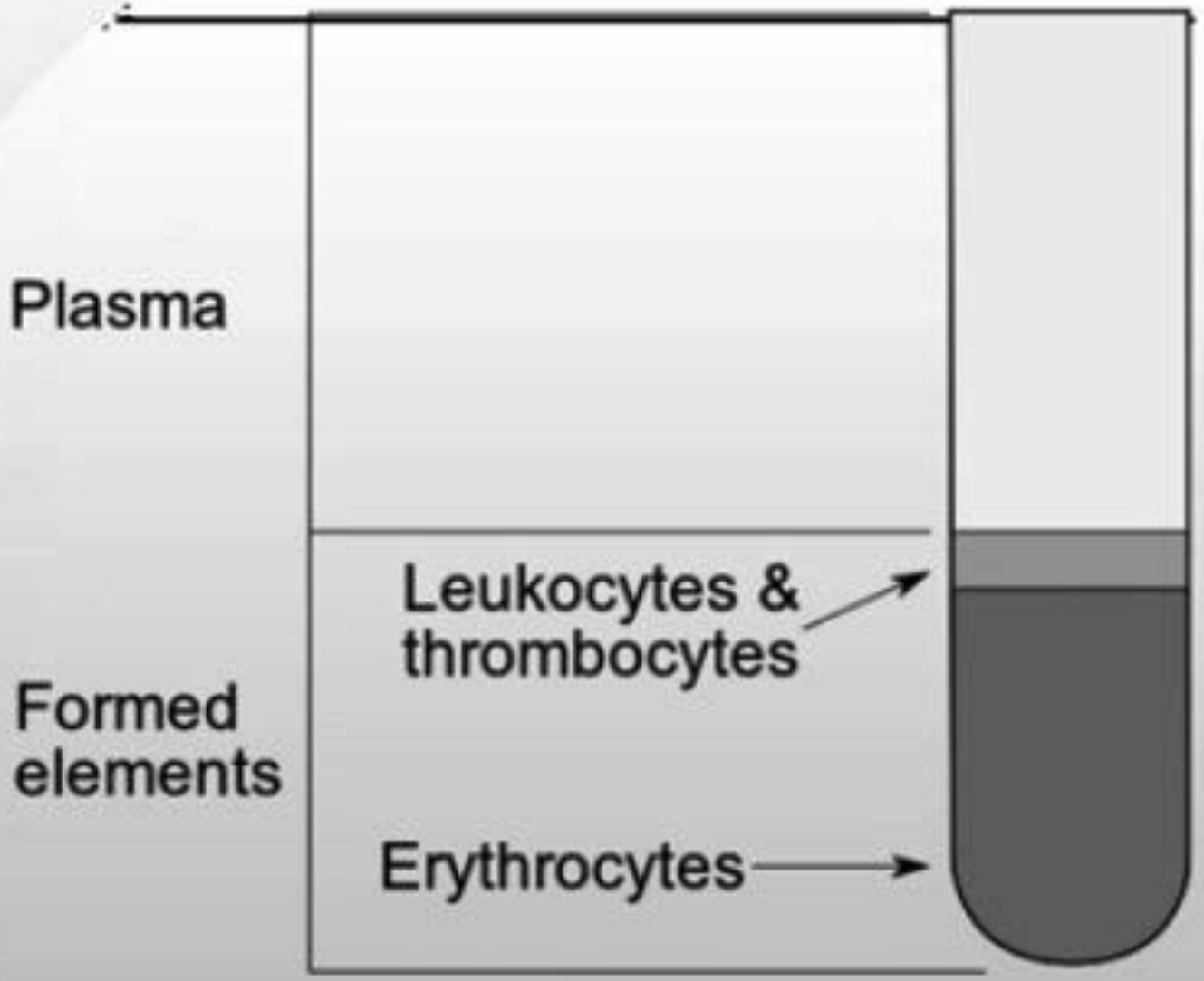
-Classification of anemia.

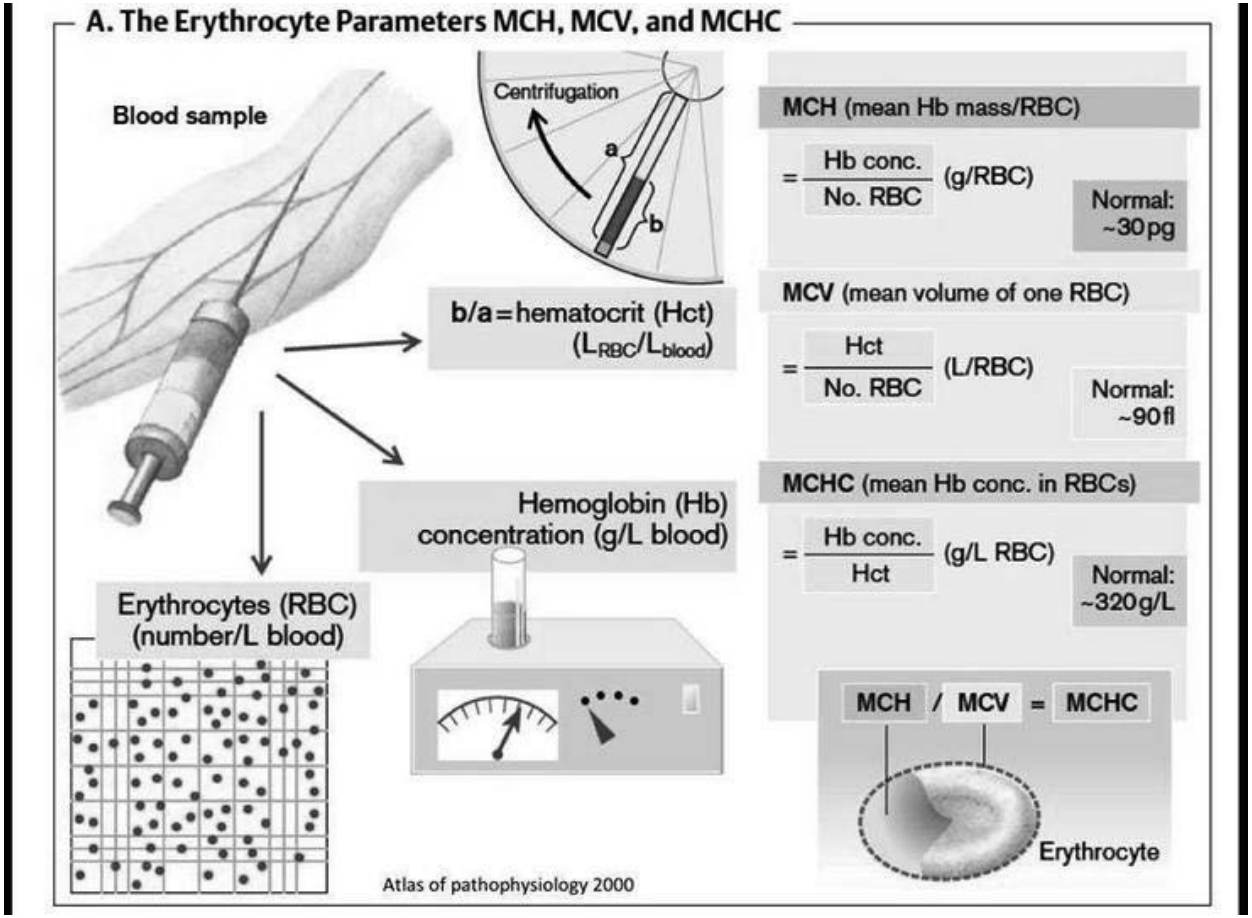
# What is anemia?!

*Reduction in the oxygen-transporting capacity of blood, which usually results from a **decrease in the red cell mass** to subnormal levels.*

- Reflected in the decrease in hematocrit or hemoglobin concentration.

# Hematocrit


Plasma
Leukocytes &
thrombocytes
Formed
elements
Erythrocytes


A. The Erythrocyte Parameters MCH, MCV, and MCHC
Blood sample
Centrifugation
a
b
b/a = hematocrit (Hct)
(L_RBC/L_blood)
Hemoglobin (Hb)
concentration (g/L blood)
Erythrocytes (RBC)
(number/L blood)
MCH (mean Hb mass/RBC)
= Hb conc. / No. RBC (g/RBC)
Normal: ~30 pg
MCV (mean volume of one RBC)
= Hct / No. RBC (L/RBC)
Normal: ~90 fl
MCHC (mean Hb conc. in RBCs)
= Hb conc. / Hct (g/L RBC)
Normal: ~320 g/L
MCH / MCV = MCHC
Erythrocyte
Atlas of pathophysiology 2000

- Mean cell volume (MCV): the average volume of a red cell expressed in femtoliters (fL)
- Mean cell hemoglobin (MCH): the average content (mass) of hemoglobin per red cell, expressed in pictograms.
- Mean cell hemoglobin concentration (MCHC): the average concentration of hemoglobin in a given volume of packed red cells, expressed in grams per deciliter.
- Red cell distribution width (RDW): the coefficient of variation of red cell volume.
- Hematocrit: the ratio of packed red cells to total blood volume.
- RBC count: the number of RBCs per unit volume, usually expressed in number (usually in millions) /microliter, for example $5x10^6$/microliter.

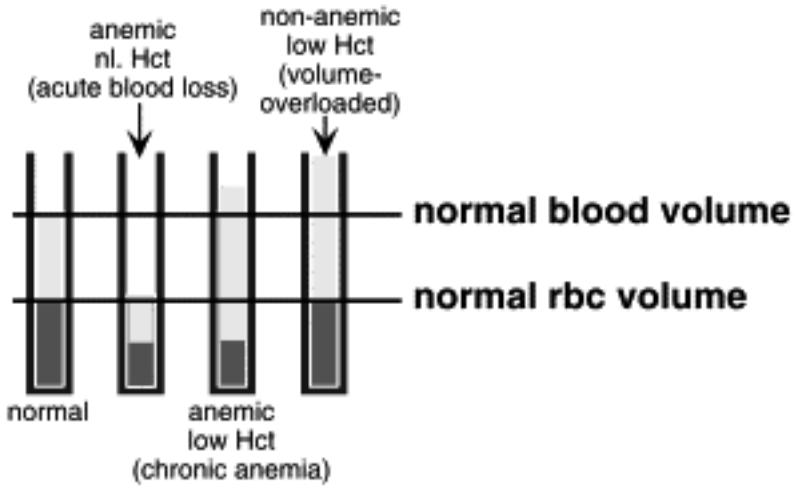
| Measurement (units) | Men | Women |
|---|---|---|
| Hemoglobin ( gm/dL) | 13.6-17.2 | 12.0-15.0 |
| Hematocrit (%) | 39-49 | 33-43 |
| Red cell count ($\times 10^6/\mu L$) | 4.3-5.9 | 3.5-5.0 |
| Reticulocyte count (%) | 0.5-1.5 | |
| Mean cell volume (fL) | 82-96 | |
| Mean cell hemoglobin (pg) | 27-33 | |
| Mean cell hemoglobin concentration (gm/dL) | 33-37 | |
| Red cell distribution width | 11.5-14.5 | |

# Question time!!!

- 45 year old male, was injured in a car accident, he bled profusely. Upon presentation he was obtunded, pale and distressed
- Vital signs were as follows:
  - heart rate 140 beat/minute.
  - Respiratory rate 25/minute
  - Blood pressure 80/30
- His hemoglobin and hematocrit were **within reference range.**

A 29 year old female, 8months pregnant, in a routine prenatal visit, she was found to have a hematocrit that is slightly below normal limits

# ARE THE PATIENTS ANEMIC???!!!


anemic
nl. Hct
(acute blood loss)
non-anemic
low Hct
(volume-
overloaded)
normal blood volume
normal rbc volume
normal
anemic
low Hct
(chronic anemia)

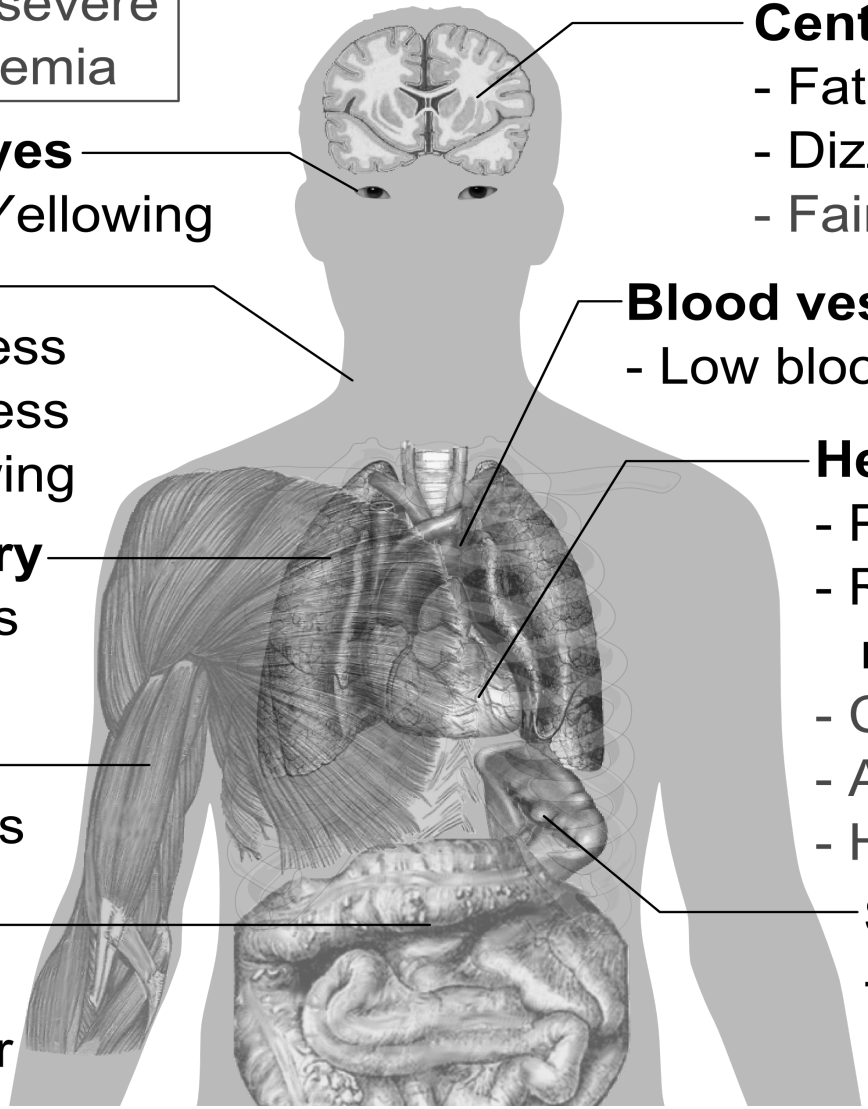
# Clinical manifestations of anemia

- Result from decreased tissue oxygenation as well as from the underlying disease.

# Pathophysiology of anemia

## Anemia

Red = In severe anemia



**Central**
- Fatigue
- Dizziness
- Fainting

**Eyes**
- Yellowing

**Skin**
- Paleness
- Coldness
- Yellowing

**Blood vessels**
- Low blood pressure

**Heart**
- Palpitations
- Rapid heart rate
- Chest pain
- Angina
- Heart attack

**Respiratory**
- Shortness of breath

**Muscular**
- Weakness

**Intestinal**
- Changed stool color

**Spleen**
- Enlarge-ment


FREE
2B
ME

# Work up

Depending on the differential diagnosis, a number of other blood tests also may be performed to evaluate anemia, including

(1) iron indices (serum iron, serum iron-binding capacity, transferrin saturation, and serum ferritin concentrations), which help distinguish among anemias caused by iron deficiency, chronic disease, and thalassemia.

(2) plasma unconjugated bilirubin, haptoglobin, and lactate dehydrogenase levels, which are abnormal in hemolytic anemias.

(3) serum and red cell folate and vitamin B12 concentrations, which are low in megaloblastic anemias.

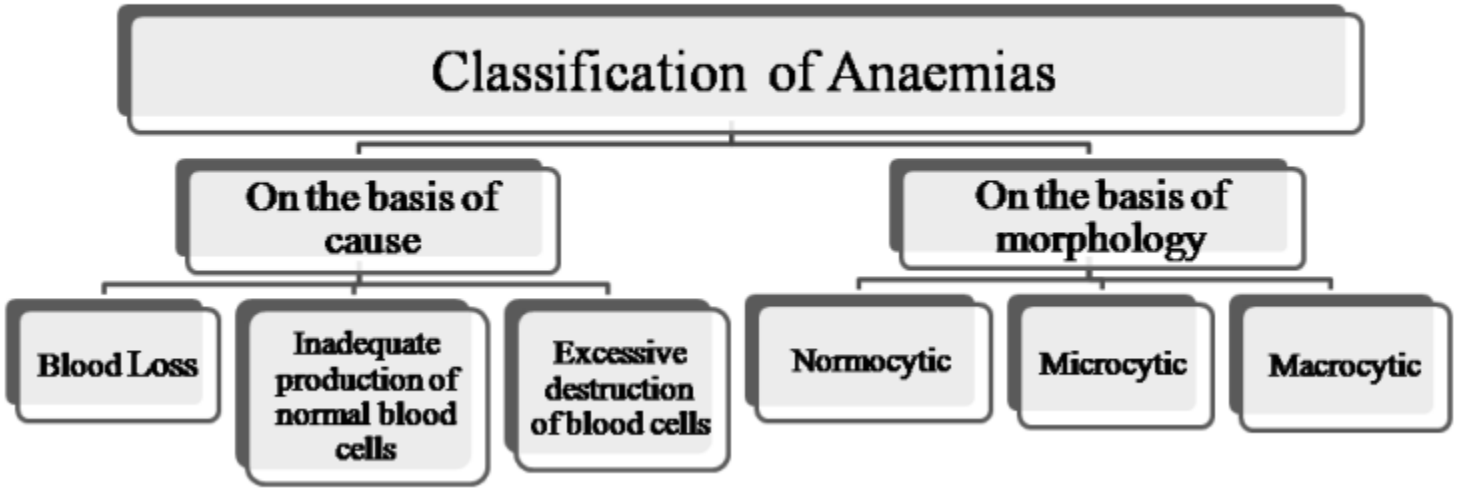
(4) hemoglobin electrophoresis, which is used to detect abnormal hemoglobins.

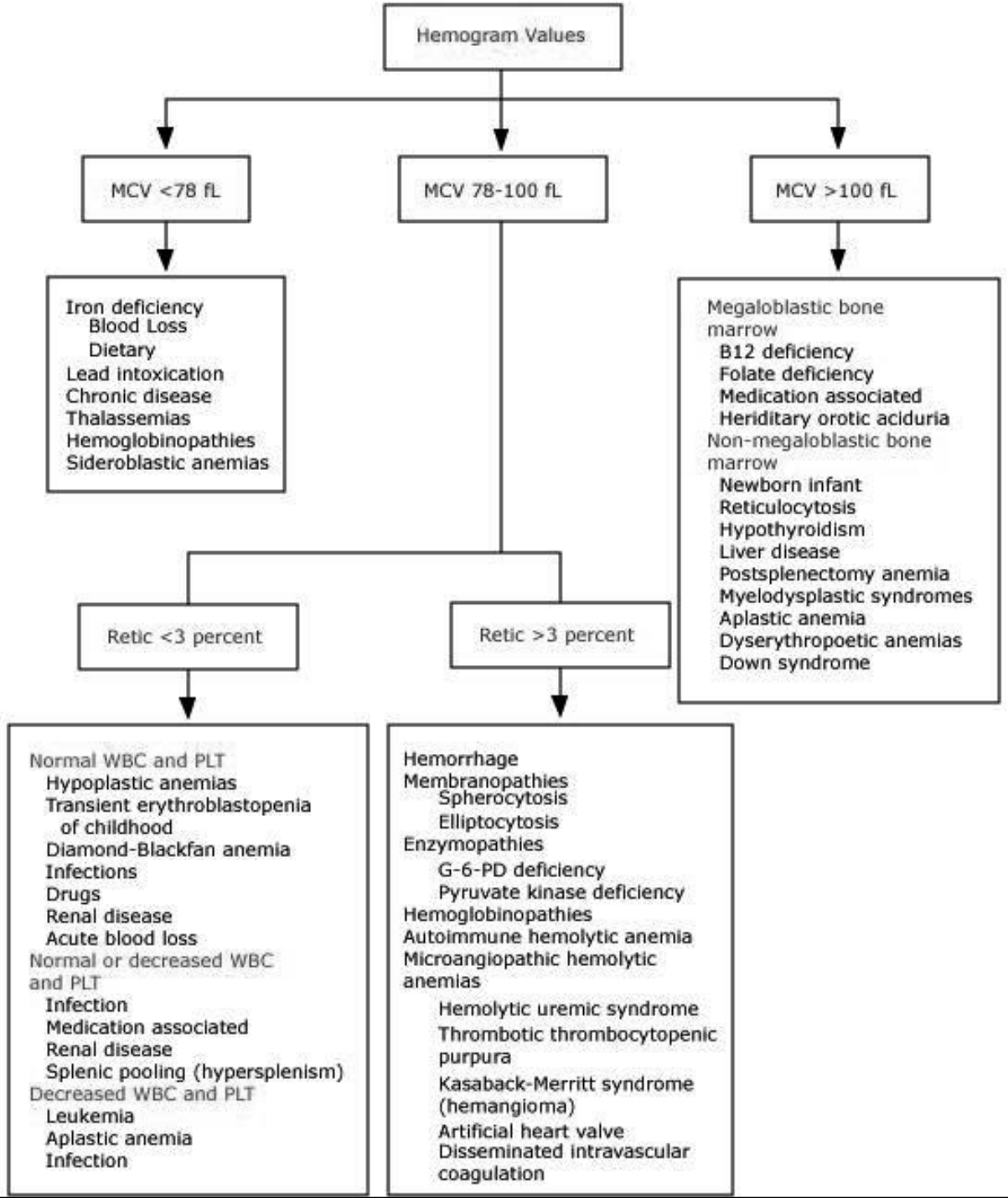
(5) the Coombs test, which is used to detect antibodies or complement on red cells in suspected cases of immunohemolytic anemia

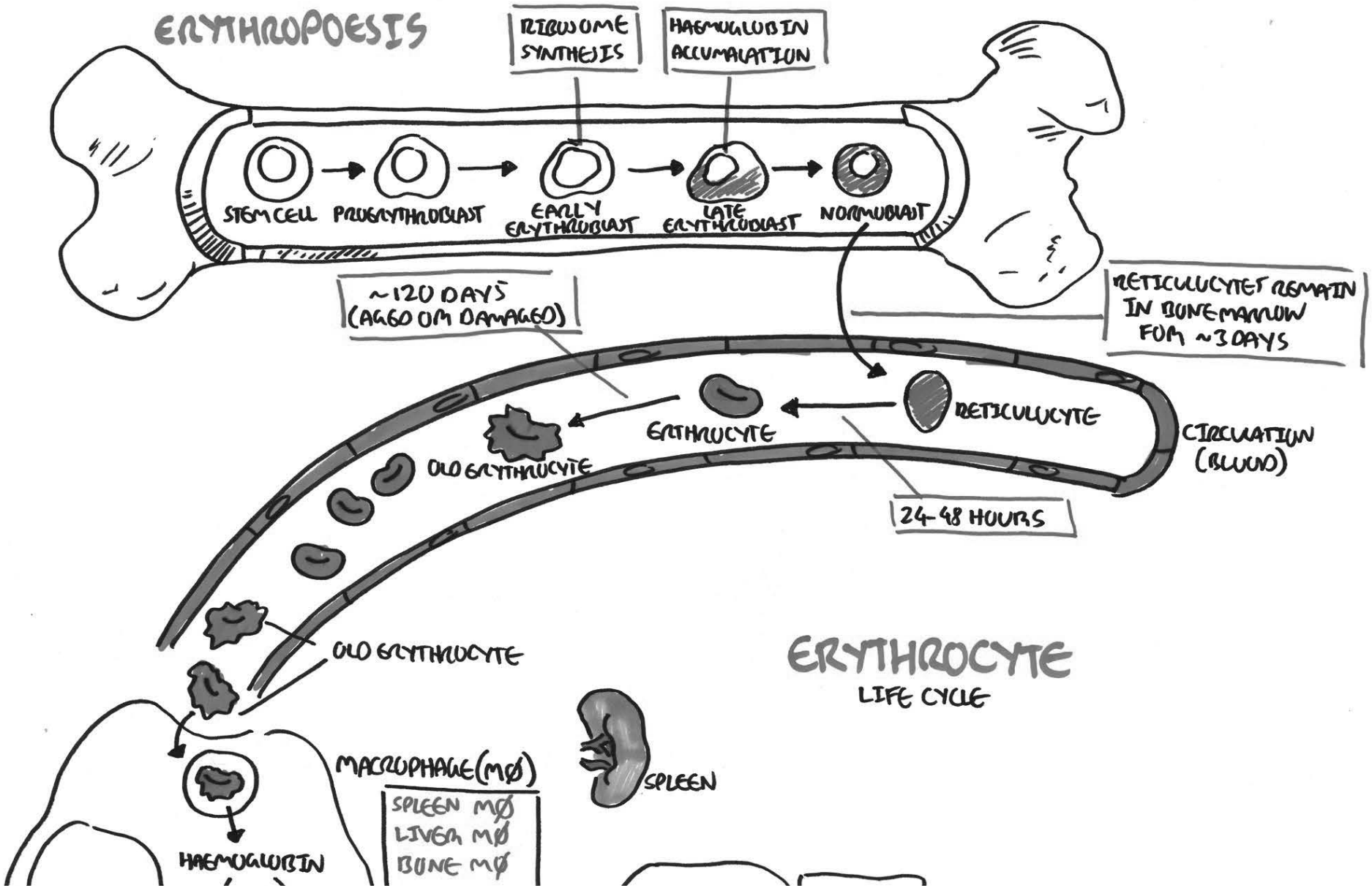
In isolated anemia, tests performed on the peripheral blood usually suffice to establish the cause.

However, if the anemia is associated with other cytopenias, then a more serious etiology should be sought and a bone marrow examination is warranted.

# Classification of Anemia




Hemogram Values
MCV <78 fL
MCV 78-100 fL
MCV >100 fL
Iron deficiency
Blood Loss
Dietary
Lead intoxication
Chronic disease
Thalassemias
Hemoglobinopathies
Sideroblastic anemias
Megaloblastic bone marrow
B12 deficiency
Folate deficiency
Medication associated
Heriditary orotic aciduria
Non-megaloblastic bone marrow
Newborn infant
Reticulocytosis
Hypothyroidism
Liver disease
Postsplenectomy anemia
Myelodysplastic syndromes
Aplastic anemia
Dyserythropoetic anemias
Down syndrome
Retic <3 percent
Retic >3 percent
Normal WBC and PLT
Hypoplastic anemias
Transient erythroblastopenia of childhood
Diamond-Blackfan anemia
Infections
Drugs
Renal disease
Acute blood loss
Normal or decreased WBC and PLT
Infection
Medication associated
Renal disease
Splenic pooling (hypersplenism)
Decreased WBC and PLT
Leukemia
Aplastic anemia
Infection
Hemorrhage
Membranopathies
Spherocytosis
Elliptocytosis
Enzymopathies
G-6-PD deficiency
Pyruvate kinase deficiency
Hemoglobinopathies
Autoimmune hemolytic anemia
Microangiopathic hemolytic anemias
Hemolytic uremic syndrome
Thrombotic thrombocytopenic purpura
Kasaback-Merritt syndrome (hemangioma)
Artificial heart valve
Disseminated intravascular coagulation


ERYTHROPOESIS
RIBOSOME SYNTHESIS
HAEMOGLOBIN ACCUMULATION
STEM CELL
PROERYTHROBLAST
EARLY ERYTHROBLAST
LATE ERYTHROBLAST
NORMOBLAST
~120 DAYS (AGED OR DAMAGED)
RETICULOCYTES REMAIN IN BONE MARROW FOR ~3 DAYS
ERYTHROCYTE
RETICULOCYTE
CIRCULATION (BLOOD)
OLD ERYTHROCYTE
24-48 HOURS
OLD ERYTHROCYTE
ERYTHROCYTE
LIFE CYCLE
MACROPHAGE (MØ)
SPLEEN
SPLEEN MØ
LIVER MØ
BONE MØ
HAEMOGLOBIN

# Introduction

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C. Microangiopathic hemolytic anemia

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E. G6PD deficiency

5. All the following are clinical manifestations of anemia of diminished production, except:

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B. Shortness of breath

**C.Gallbaldder stones**

D.Muscle weakness

E. Confusion

Thank You

# RBC disorders 2
# Anemia of diminished production

Ahmad T. Mansour, MD

- Iron deficiency anemia
- Anemia of chronic disease
- Megaloblastic anemia
- Others
  - anemia  in liver disease
  - anemia in renal disease
  - aplastic anemia
  - myelophthisic anemia

1. All of the following can be found in iron deficiency anemia, except:
   A. Low ferritin
   B. Low serum iron
   C. Low TIBC
   D. Low transferrin saturation
   E. Low MCV

2. Anemia of chronic disease is caused by elevated levels of:

A. Hepcidin

B. Iron

C. Ferritin

D. B12

E. neutrophils

3. All of the following are true regarding megaloblastic anemia, except:

A. defective DNA synthesis, resulting in nuclear immaturity

B. macrocytic anemia

C. can be seen in the setting of pernicious anemia

D. most common cause is nutritional deficiency of B12

E. can be associated with neurological symptoms in the case of B12 deficiency.

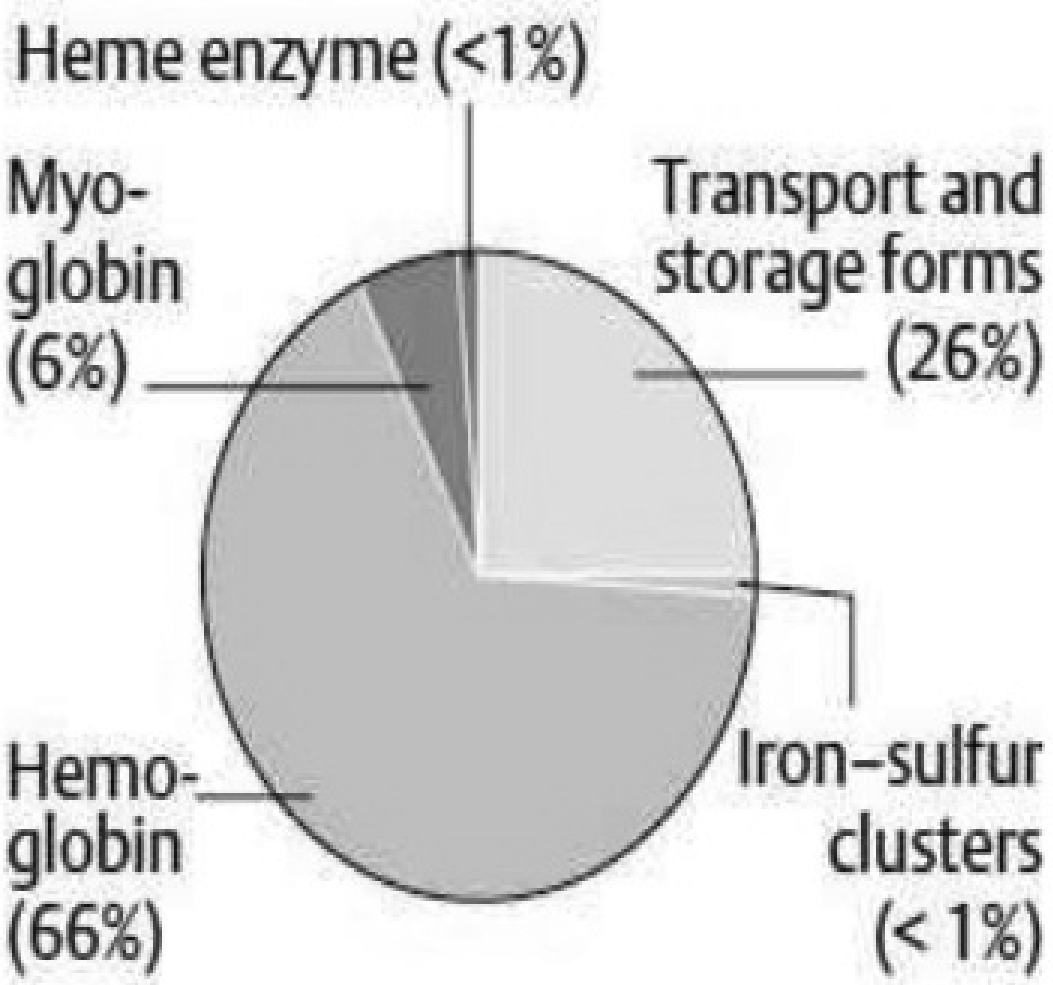
4. The most common cause of anemia in patients with liver disease is:

A. Iron deficiency
B. Hypersplenism
C. Therapy related hemolytic anemia
D. Therapy related suppression of EPO receptor
E. Alcoholic-cirrhosis-induced folate deficiency

5. One of the following can cause myelophthisic anemia:

A. Tuberculosis
B. B12 deficiency
C. Folate deficiency
D. Iron deficiency
E. Anemia of chronic disease.

# Iron: Distribution

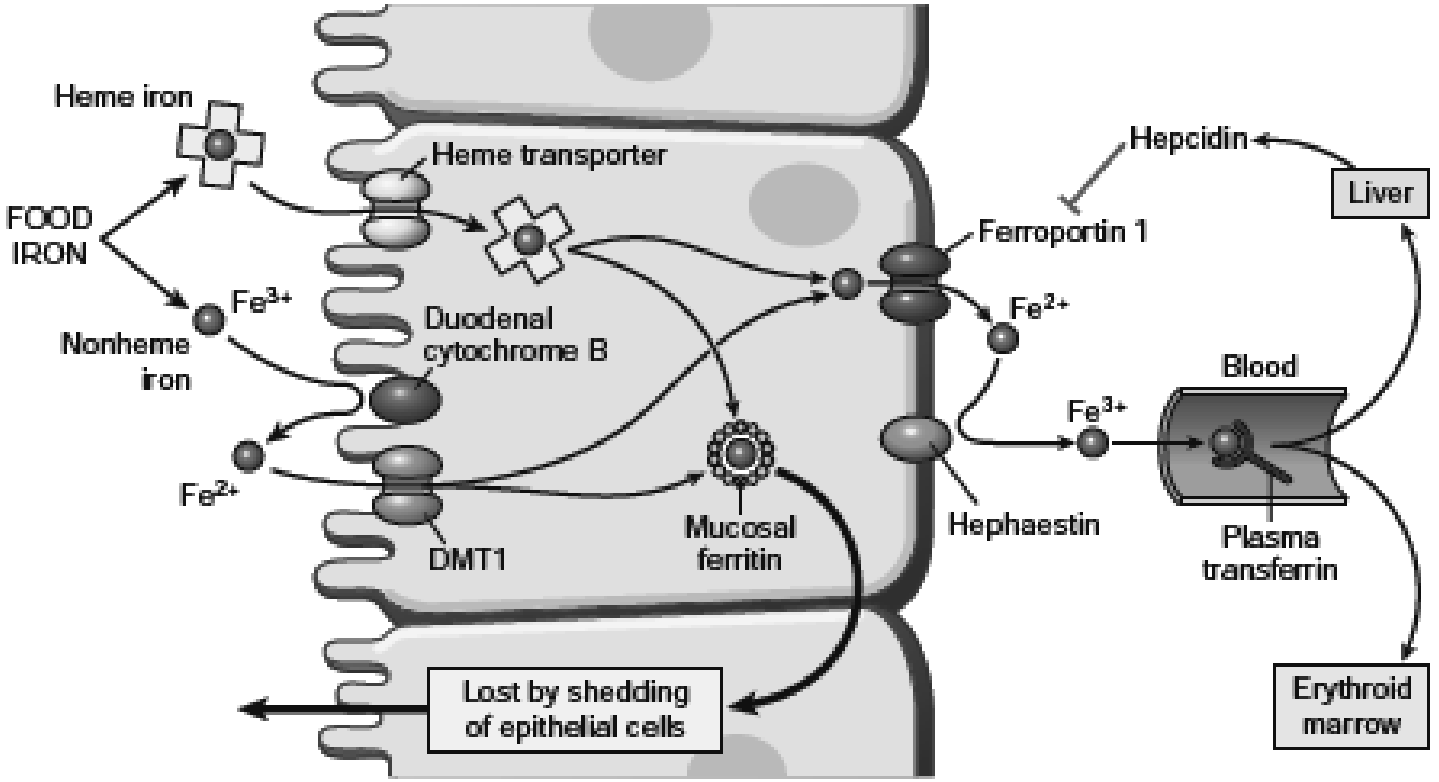



- human body: 4–5 g iron (protein-bound)
- **heme proteins (~72%)**
  - hemoglobin (2.5 g)
  - myoglobin (0.15 g)
- **transport and storage proteins (~26%)**
  - transferrin (1.0 g)
  - serum ferritin (0.0001 g)
- **iron–sulfur clusters (<1%)**
  - cofactors in the respiratory chain, other redox chains

- Transferrin is the major transport protein in plasma and is normally one third occupied.

- Plasma ferritin is derived largely from the storage pool of body iron; its levels correlate well with body iron stores, if total iron is decreased ferritin will be low, and vice versa.

- Iron balance is maintained through absorption, excretion is limited to 1-2mg/day through shedding of mucosal cells.


Heme iron
FOOD IRON
Nonheme iron
Fe3+
Fe2+
Heme transporter
Duodenal cytochrome B
DMT1
Mucosal ferritin
Lost by shedding of epithelial cells
Hepcidin
Liver
Ferroportin 1
Fe2+
Fe3+
Hephaestin
Blood
Plasma transferrin
Erythroid marrow

# Iron deficiency anemia

- Deficiency of iron is the most common nutritional disorder in the world and results in clinical signs and symptoms that are **mostly related to inadequate hemoglobin synthesis**.

- Iron deficiency can result from
  - (1) dietary
  - (2) impaired absorption,
  - (3) increased requirement,
  - (4) chronic blood loss.

# Question time!!!

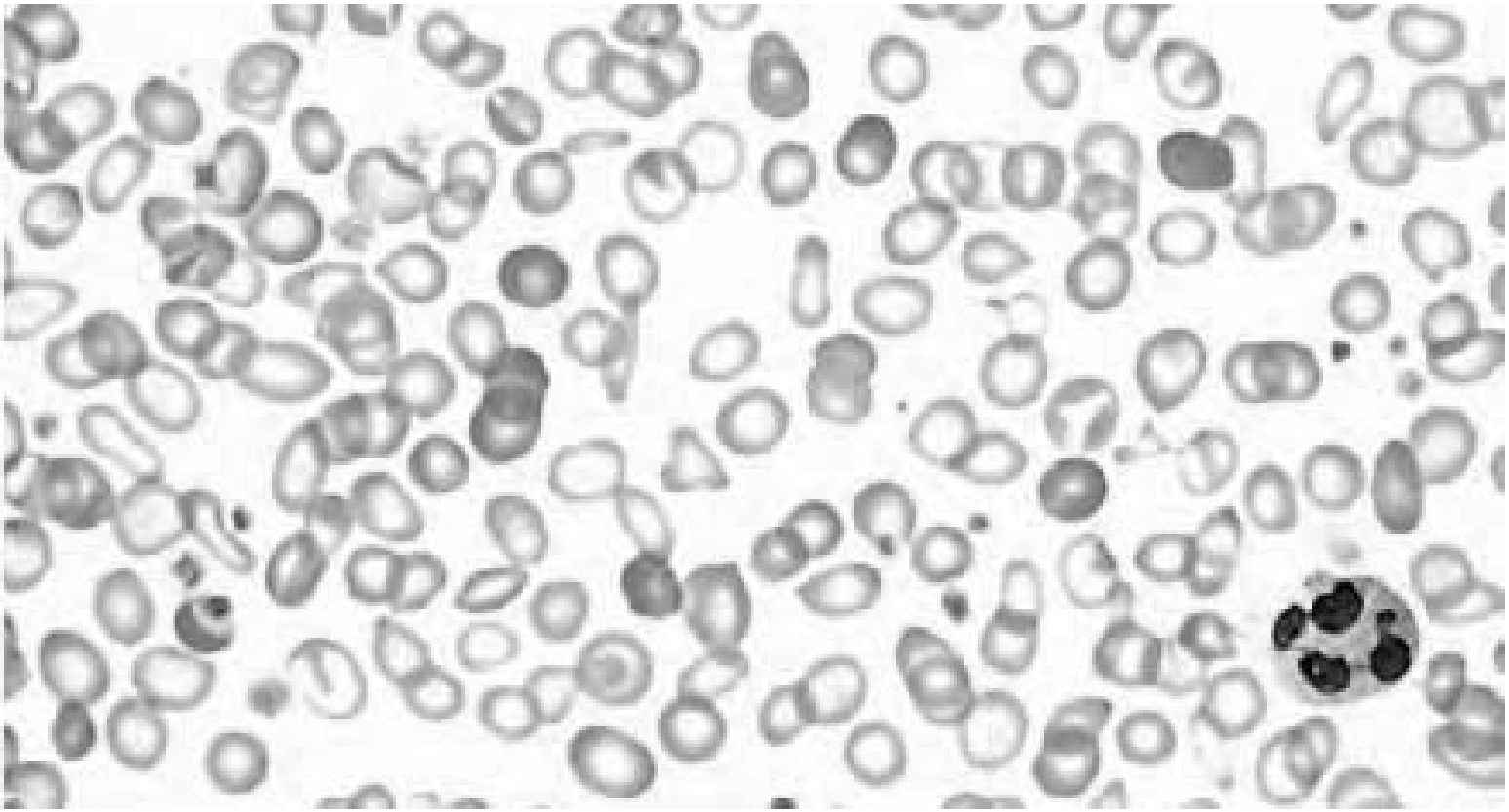
- Which clinical scenario is **more serious**
  - A 19 year old lady, with sever menorrhagia presenting with shortness of breath on exertion, fatigue, pallor and a hemoglobin of 7.6g/dl, low MCV
  - A 79 year old asymptomatic gentleman who, on routine check up, was found to have a hemoglobin of 11g/dl and low MCV

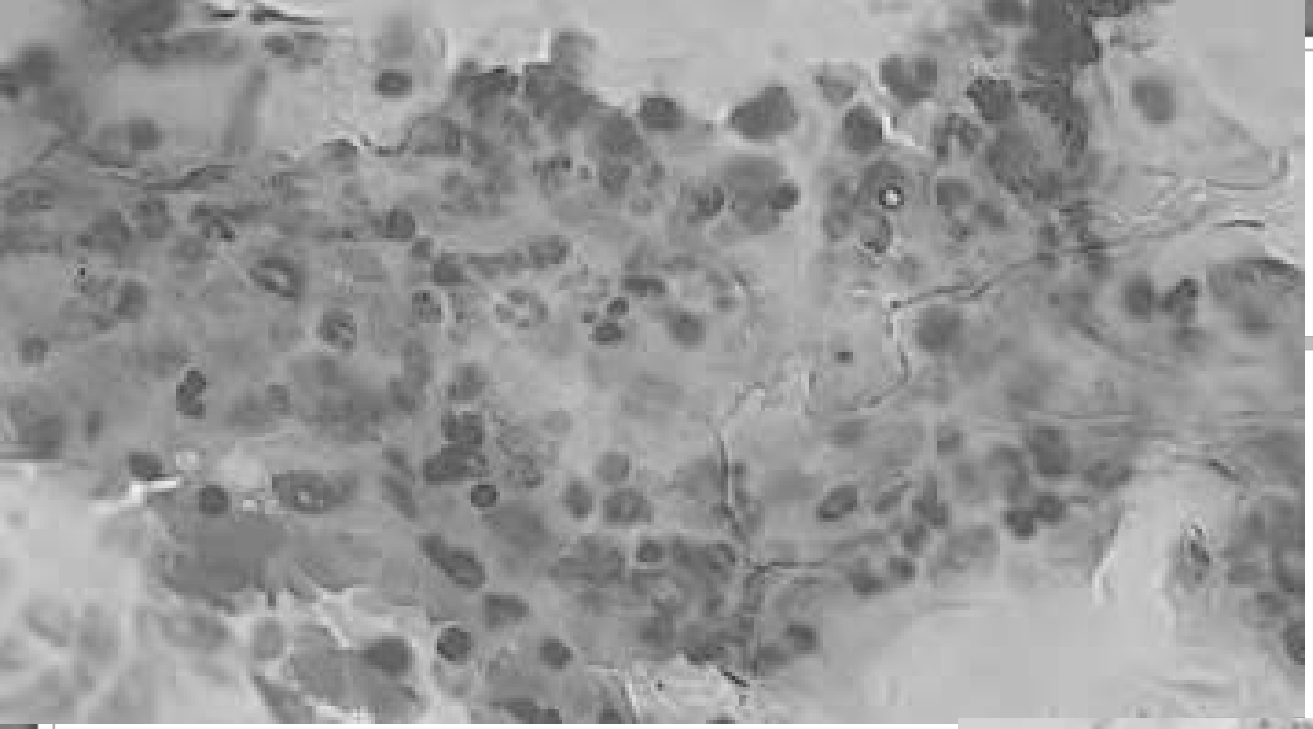
**Think of anemia as you think of fever!!!**

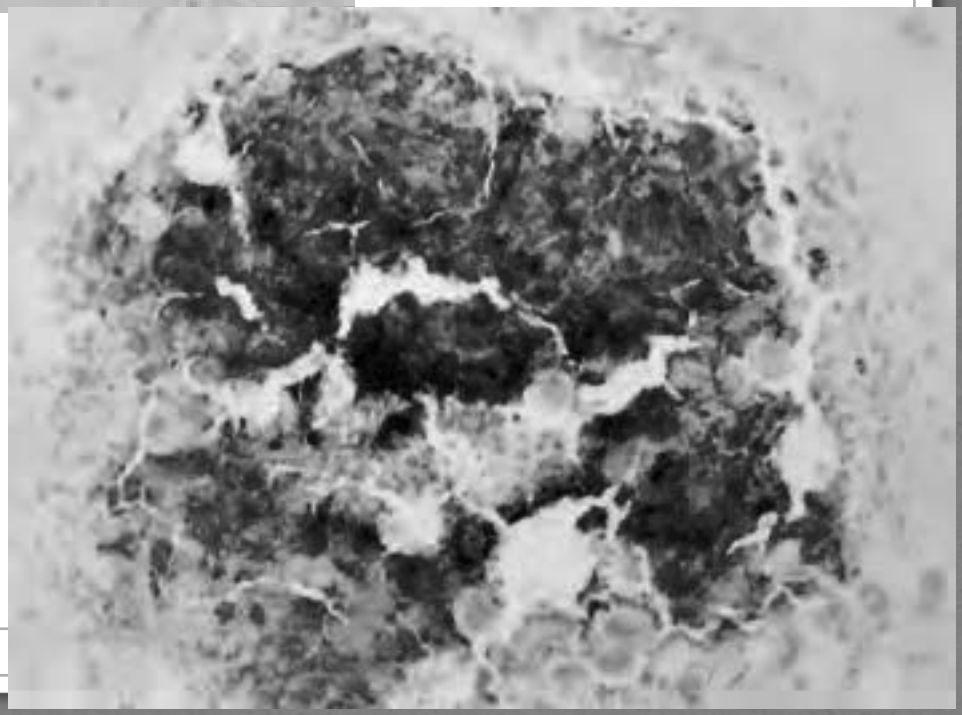
# pathogenesis

- Negative iron balance due to any reason
- Compensation by storage iron
- Progressive deficiency until complete depletion
- Anemia develops accompanied by low ferritin and low transferrin saturation

# morphology

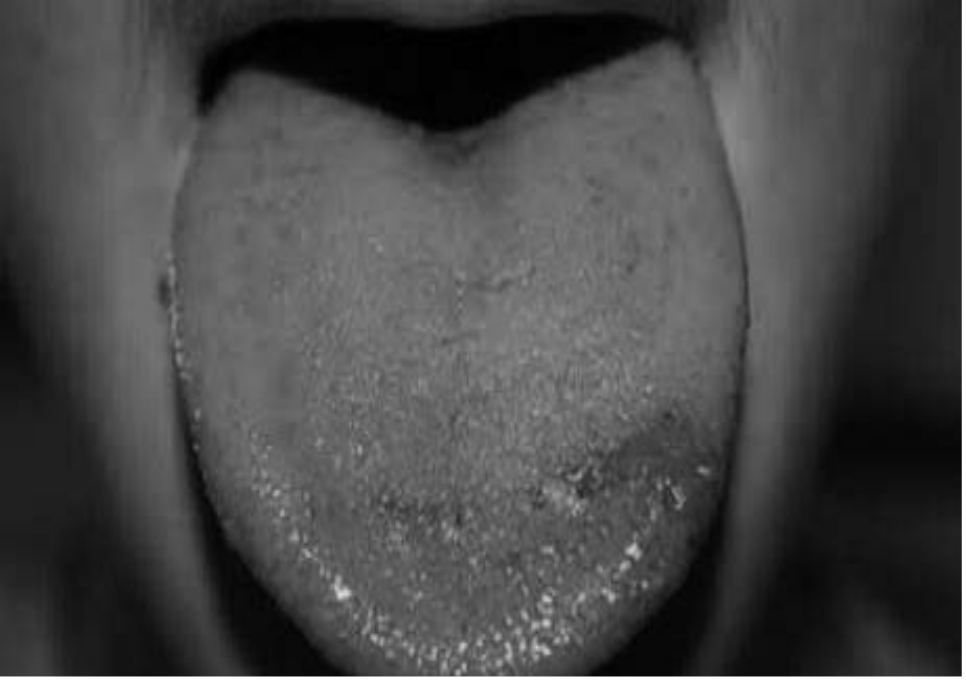


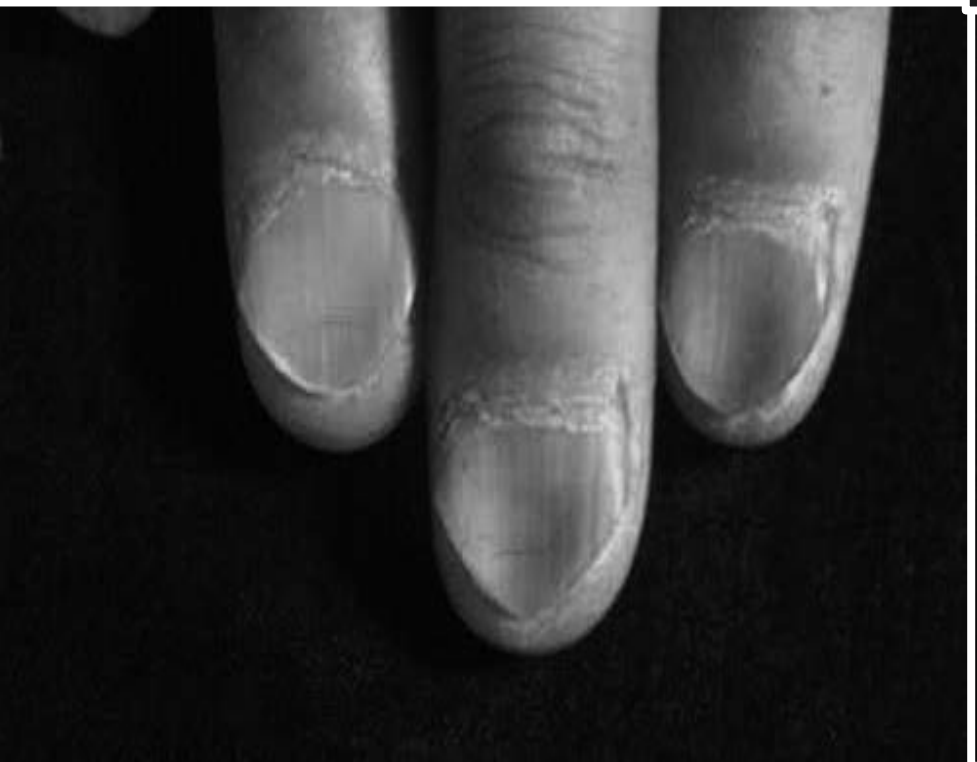


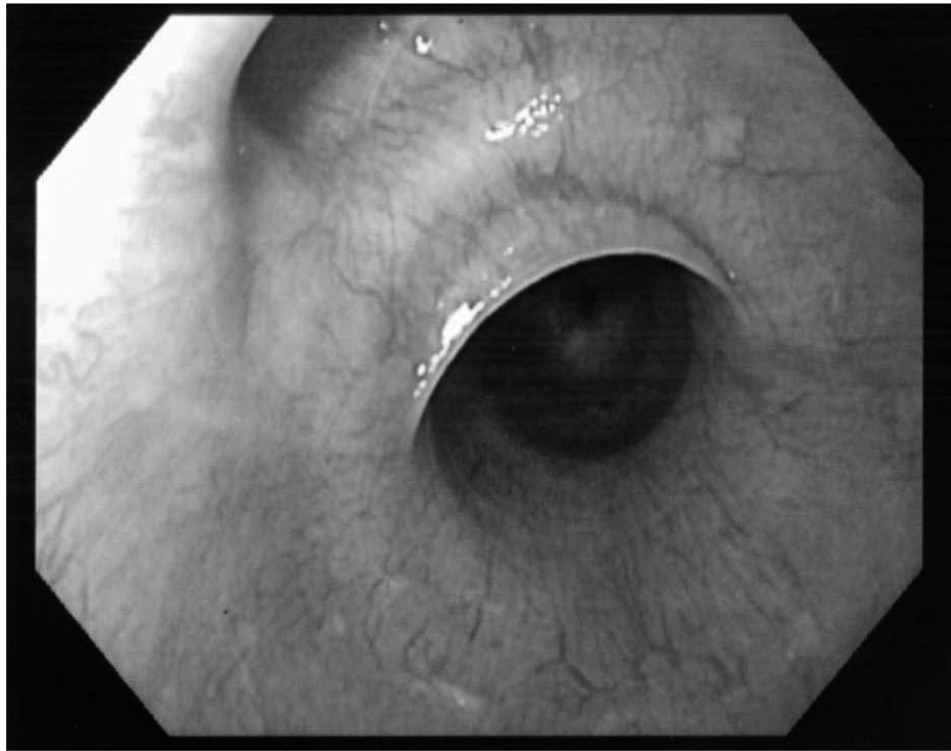


# Clinical presentation

Symptoms and signs of anemia







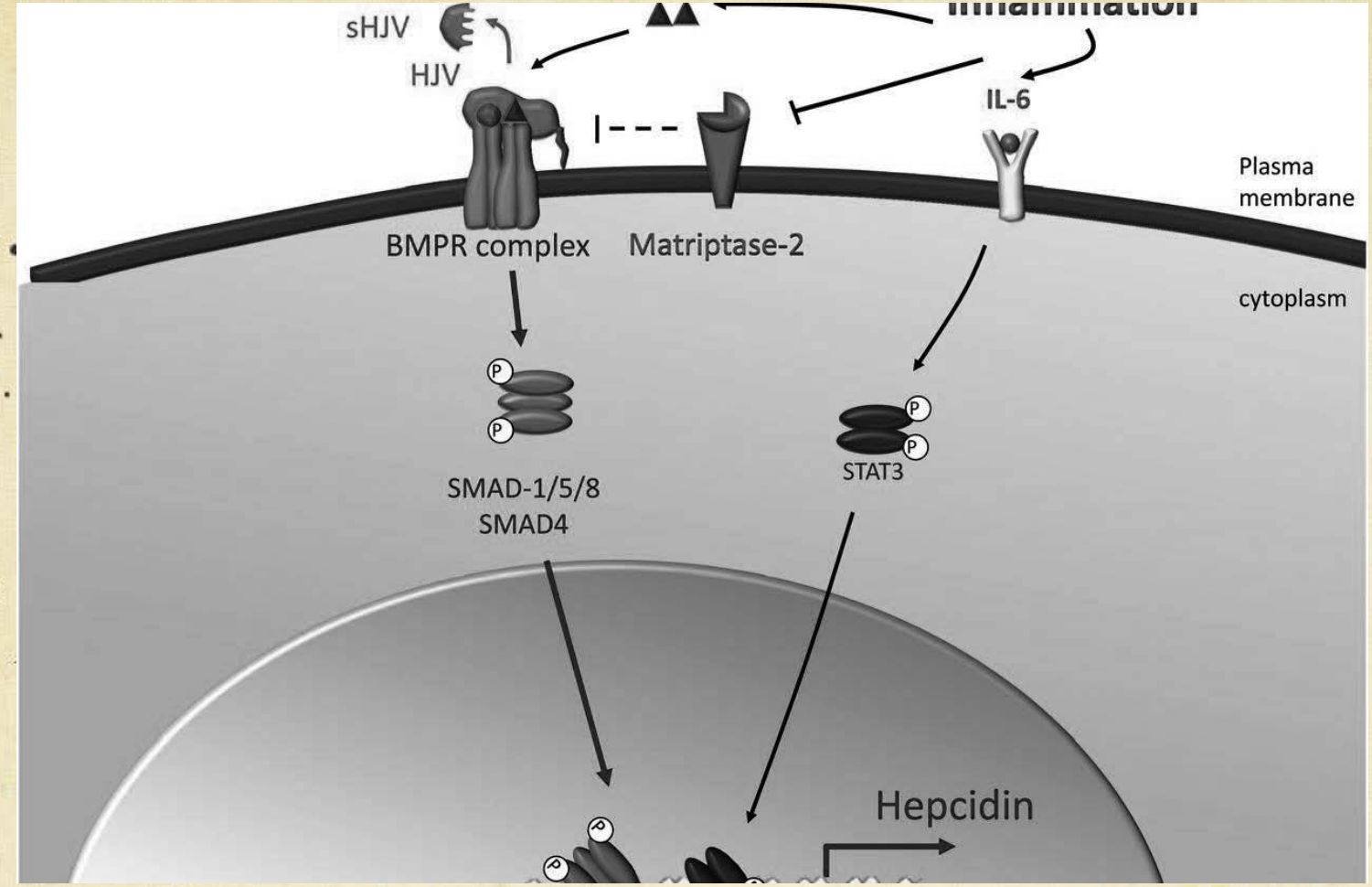
# Lab findings

- Low hemoglobin and hematocrit
- Low MCV
- Low MCH
- Low iron levels
- Low ferritin
- **High TIBC**
- **High RDW**
- Low hepcidin
- Low transferrin saturation

- Treat by iron supplementation and treating the underlying cause.

# Anemia of chronic disease

- Anemia of chronic disease is impaired red cell production associated with **chronic diseases that produce systemic inflammation**
- The most common cause of **anemia among hospitalized** patients
- Examples include chronic microbial inflammation, autoimmune inflammation, and malignancy.




# IL6 results in increased hepcidin

- Starves the EP cells of iron.
- Inhibits erythropoietin and subsequently decreases EP proliferation
    - Possible immunologic role for hepcidin

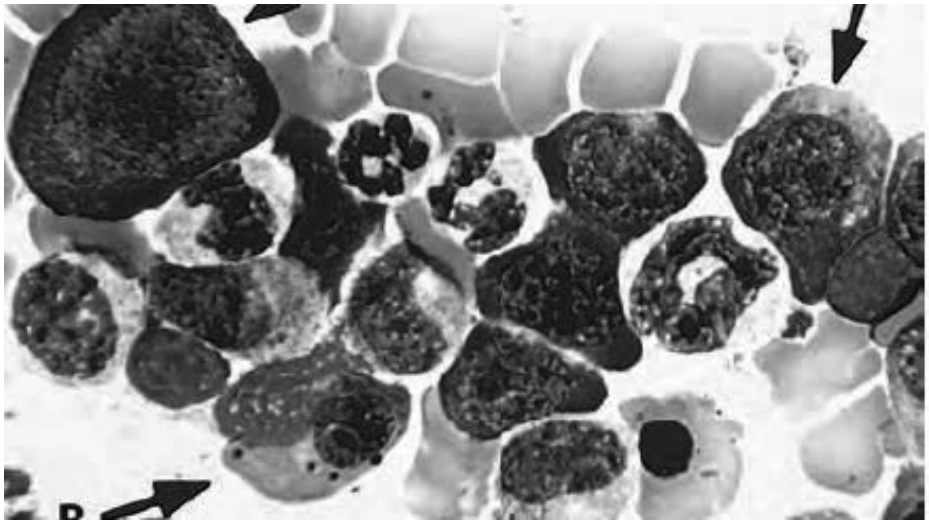
# Clinical presentation

- Mild anemia
- Signs and symptoms of underlying disease

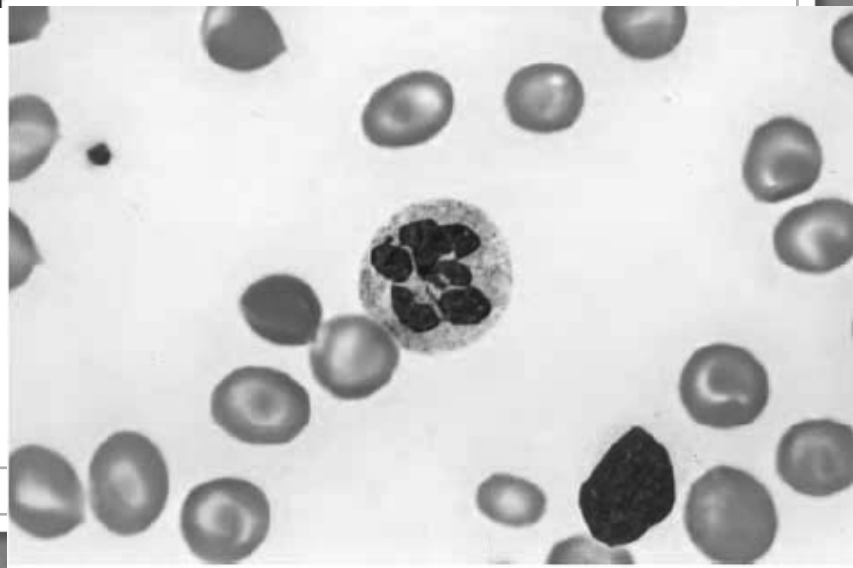
# Lab findings

- Low HB and Hct
- Can be hypochromic microcytic or normochromic normocytic.
- High ferritin and low TIBC (exactly opposite to iron deficiency anemia)

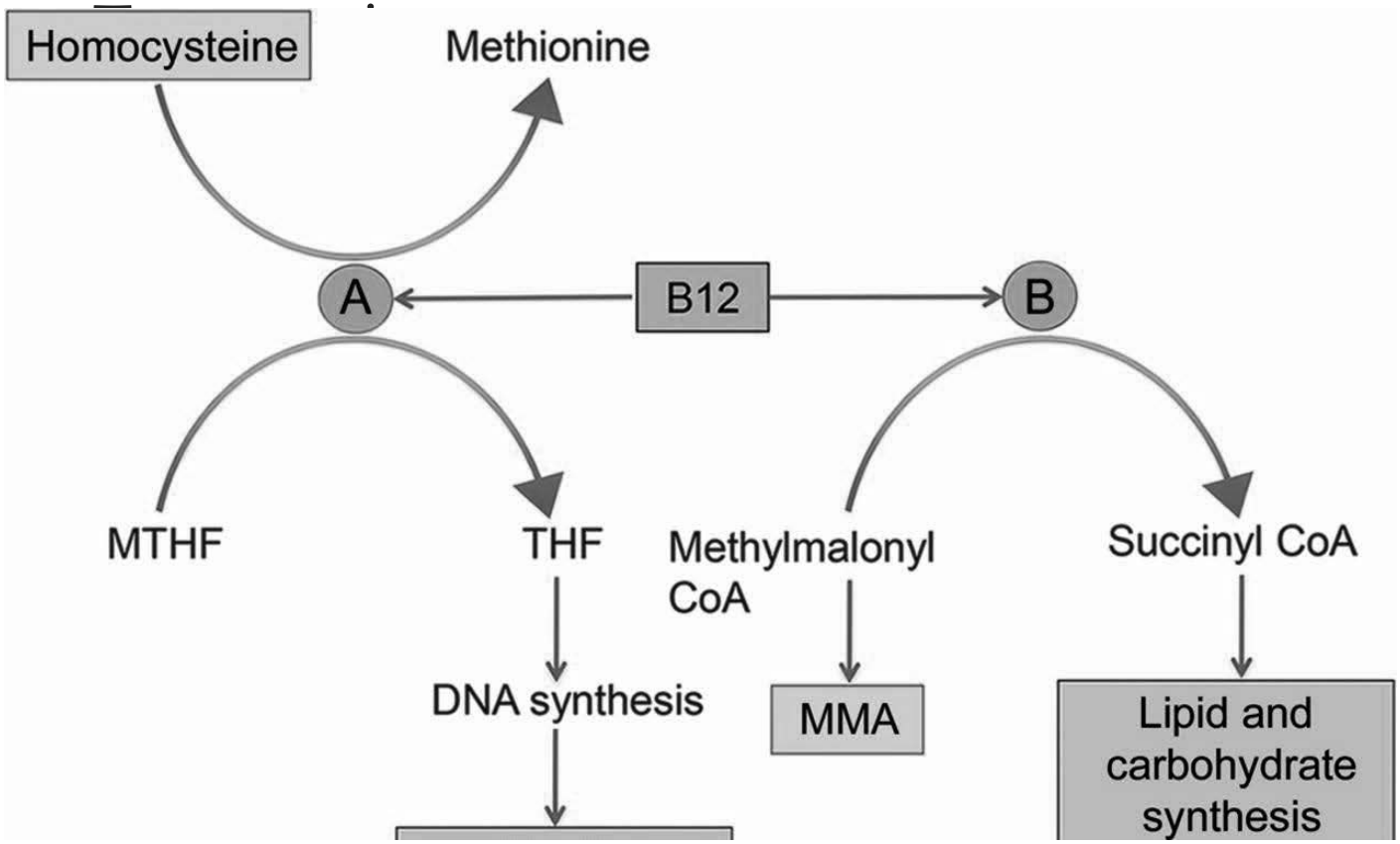
# Megaloblastic anemia

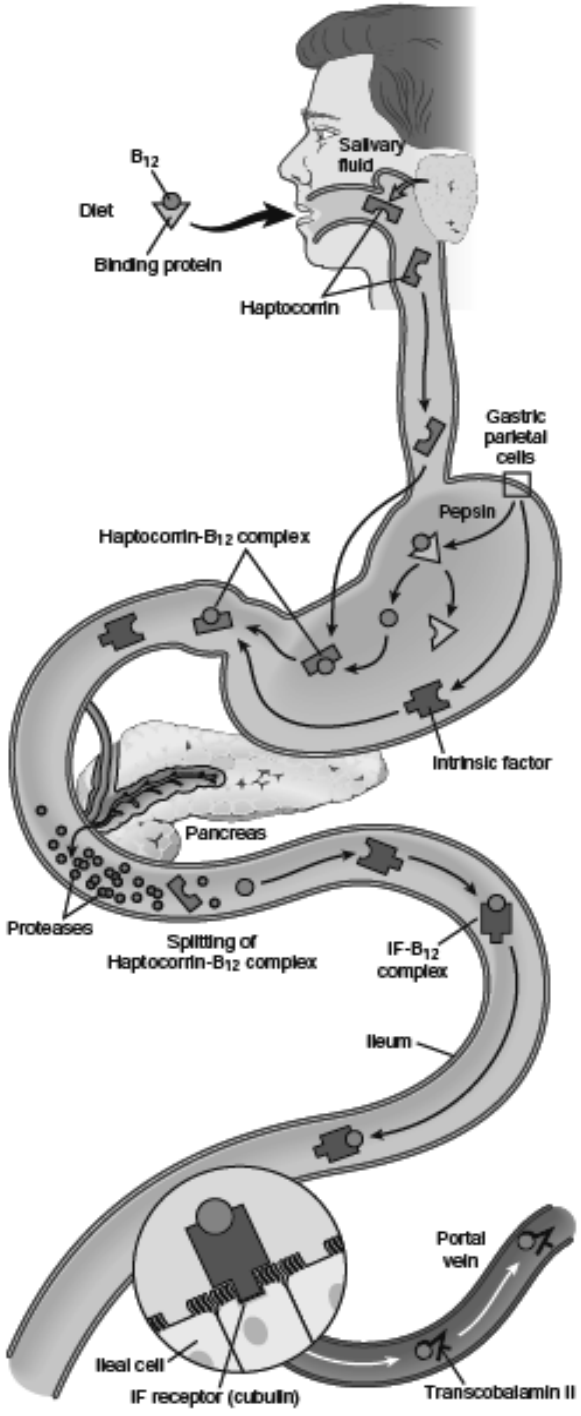


Vitamin B12 or folate deficiency



# Biochemical role of B12




$B_{12}$
Diet
Binding protein
Salivary fluid
Haptocorrin
Gastric parietal cells
Pepsin
Haptocorrin-$B_{12}$ complex
Intrinsic factor
Pancreas
Proteases
Splitting of Haptocorrin-$B_{12}$ complex
IF-$B_{12}$ complex
Ileum
Portal vein
Ileal cell
IF receptor (cubulin)
Transcobalamin II

## Vitamin $B_{12}$ Deficiency

### Decreased Intake

Inadequate diet, vegetarianism

### Impaired Absorption

Intrinsic factor deficiency
- Pernicious anemia
- Gastrectomy

Malabsorption states

Diffuse intestinal disease (e.g., lymphoma, systemic sclerosis)

Ileal resection, ileitis
- Competitive parasitic uptake
- Fish tapeworm infestation

Bacterial overgrowth in blind loops and diverticula of bowel

# Pernicious anemia

Autoimmune attack on gastric mucosa.

three types of antibodies

1-parietal canalicular antibodies

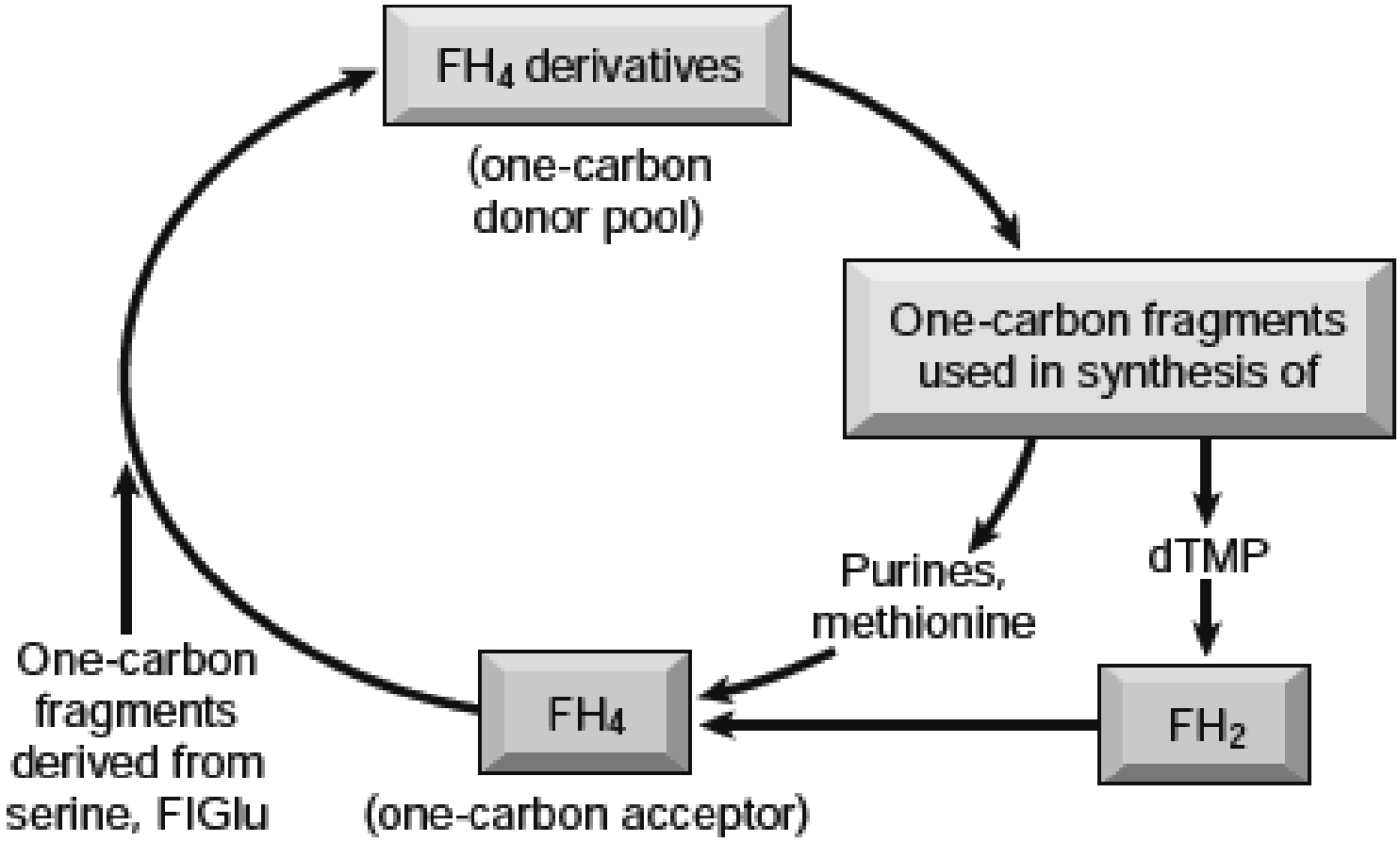
2-blocking antibodies

3-intrinsic factor–B12 complex antibodies

# Clinical manifestations

- Related to anemia similar to those found in folate deficiency
- Additionally, leukopenia with hypersemented neutrophils can be seen
- Neurological symptoms:
  - Numbness
  - Unsteady gate
  - Loss of position sense
- Increase risk of malignancy in patient with pernicious anemia

# Folate


$FH_4$ derivatives
(one-carbon donor pool)
One-carbon fragments used in synthesis of
Purines, methionine
dTMP
$FH_2$
$FH_4$
(one-carbon acceptor)
One-carbon fragments derived from serine, FIGlu

| Folic Acid Deficiency |
| --- |
| **Decreased Intake** |
| Inadequate diet, alcoholism, infancy |
| Impaired Absorption |
| Malabsorption states |
| Intrinsic intestinal disease |
| Anticonvulsants, oral contraceptives |
| Increased Loss |
| Hemodialysis |
| **Increased Requirement** |
| Pregnancy, infancy, disseminated cancer, markedly increased hematopoiesis |
| **Impaired Utilization** |
| Folic acid antagonists |
| **Unresponsive to Vitamin $B_{12}$ or Folic Acid Therapy** |
| Metabolic inhibitors of DNA Synthesis and/or Folate Metabolism (e.g., Methotrexate) |

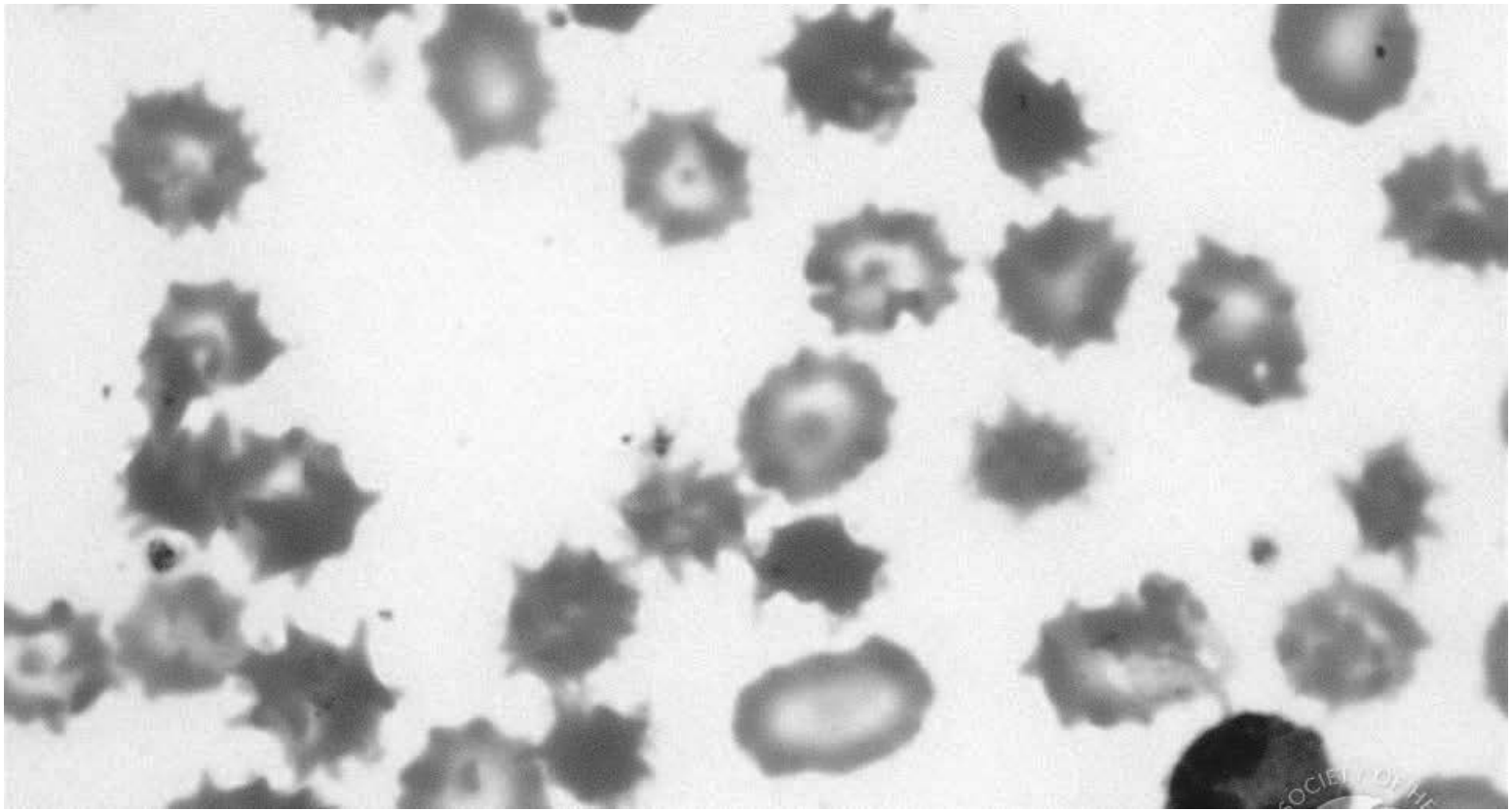
# Clinical manifestations

- Nonspecific symptoms of anemia, weakness, fatigue…etc
- GI symptoms due to the effect on GI epithelial lining cells.
- NO neurological symptoms
- Diagnose by serum and RBC folate levels.

# Anemia in liver disease

- Multiple etiologies:
  - Iron deficiency is the most common
  - Hypersplenism
  - Therapy related hemolytic anemia and suppression of EPO receptor
  - Alcoholic-cirrhosis-induced folate deficiency

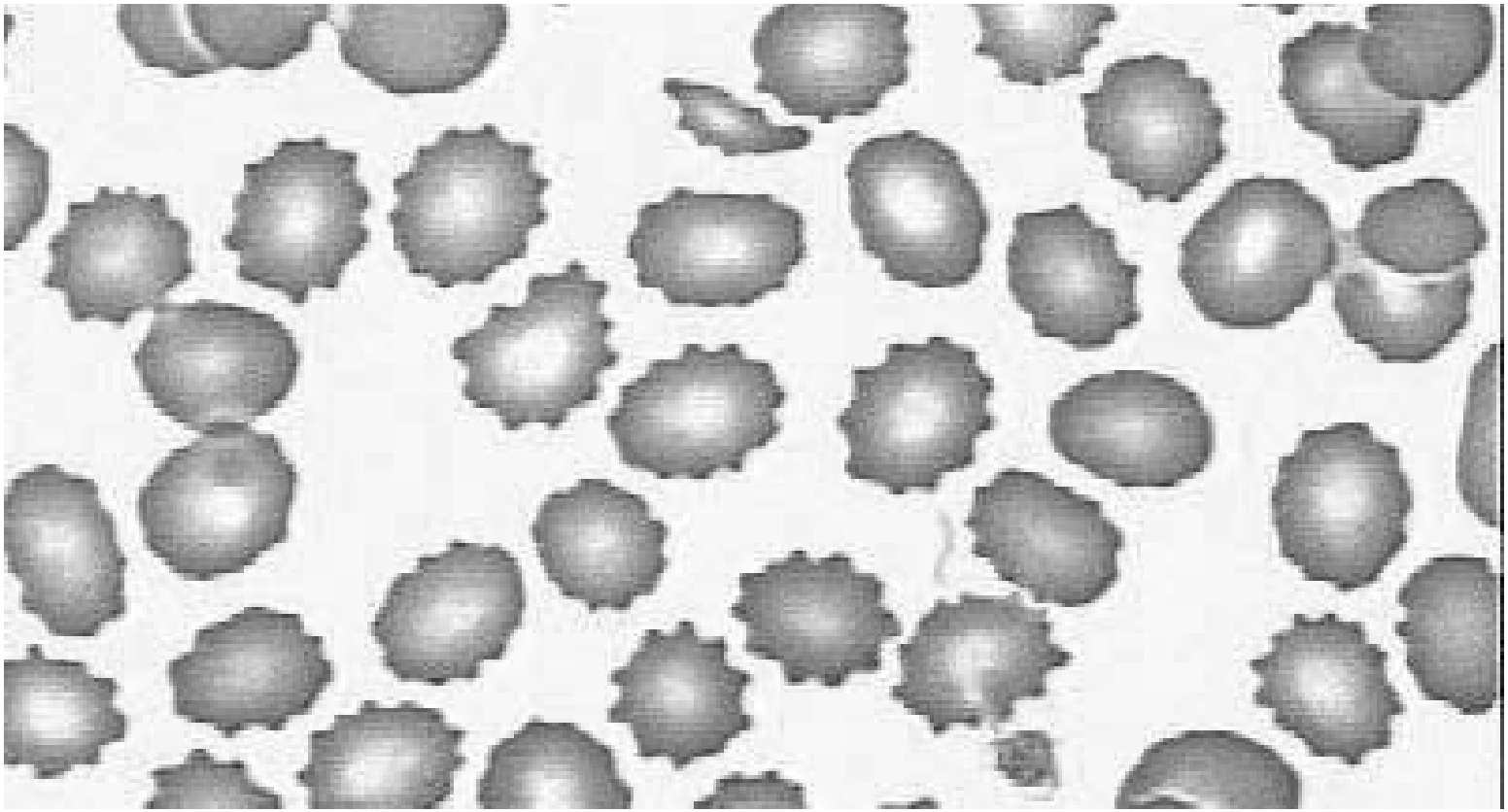
# Spur cells



# Anemia of renal disease

- Most commonly a normochromic normocytic anemia
- Related to decrease EPO production by the damaged kidney.
- High levels of inflammatory cytokines
- Hemolysis
- Chronic bleeding
- Folate deficiency in patients on dialysis.

# ecchinocytes



# Aplastic anemia

*Aplastic anemia refers to a syndrome of chronic primary* ***hematopoietic failure*** *and attendant* ***pancytopenia*** *(anemia, neutropenia, and thrombocytopenia)*

## Acquired

Idiopathic
Acquired stem cell defects
Immune mediated

## Chemical Agents

Dose related
- Alkylating agents
- Antimetabolites
- Benzene
- Chloramphenicol
- Inorganic arsenicals

Idiosyncratic
- Chloramphenicol
- Phenylbutazone
- Organic arsenicals
- Methylphenylethylhydantoin
- Carbamazepine
- Penicillamine
- Gold salts

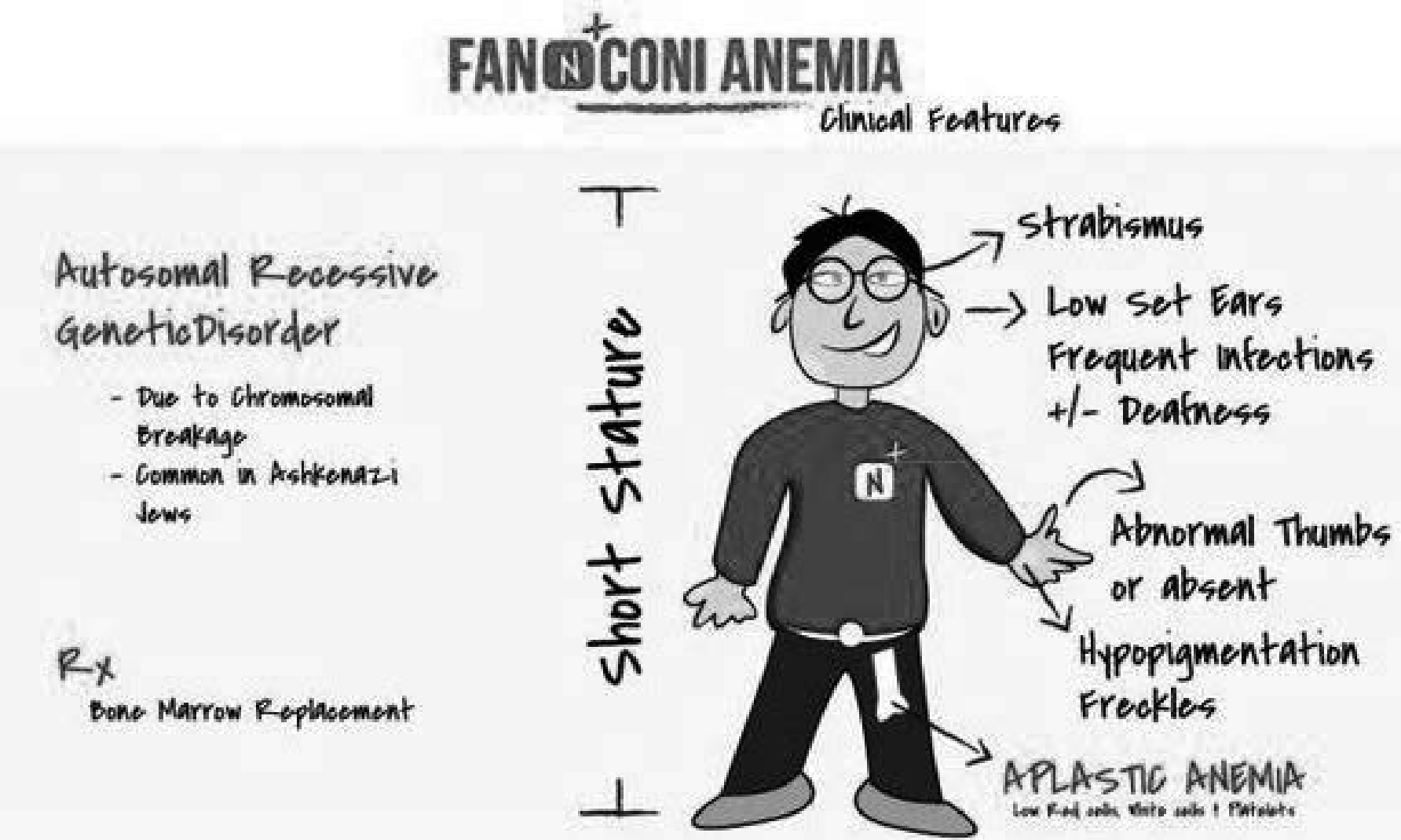
## Physical Agents

Whole-body irradiation
Viral Infections
Hepatitis (unknown virus)
Cytomegalovirus infections
Epstein-Barr virus infections
Herpes zoster (varicella zoster)

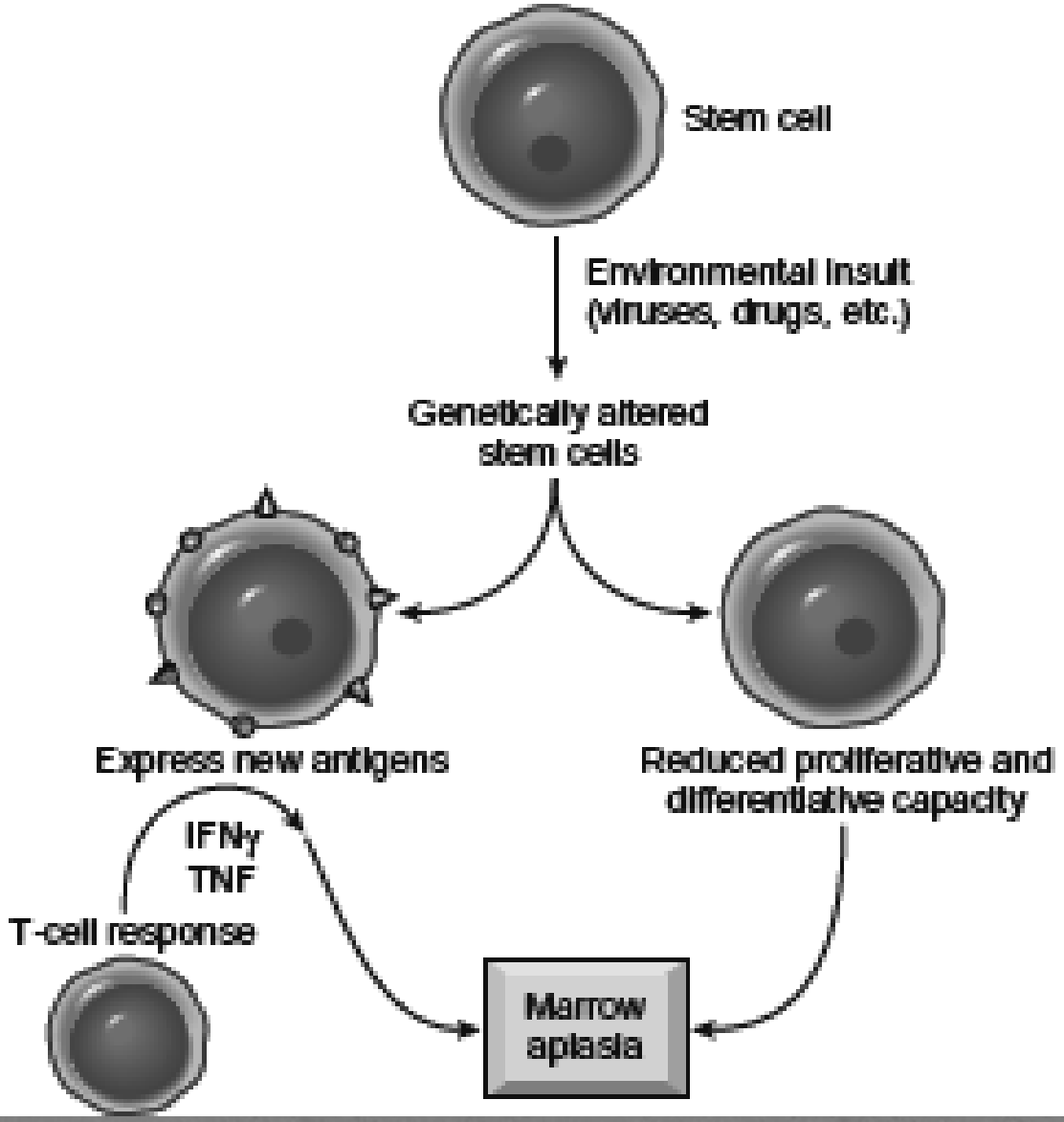
## Inherited

Fanconi anemia
Telomerase defects

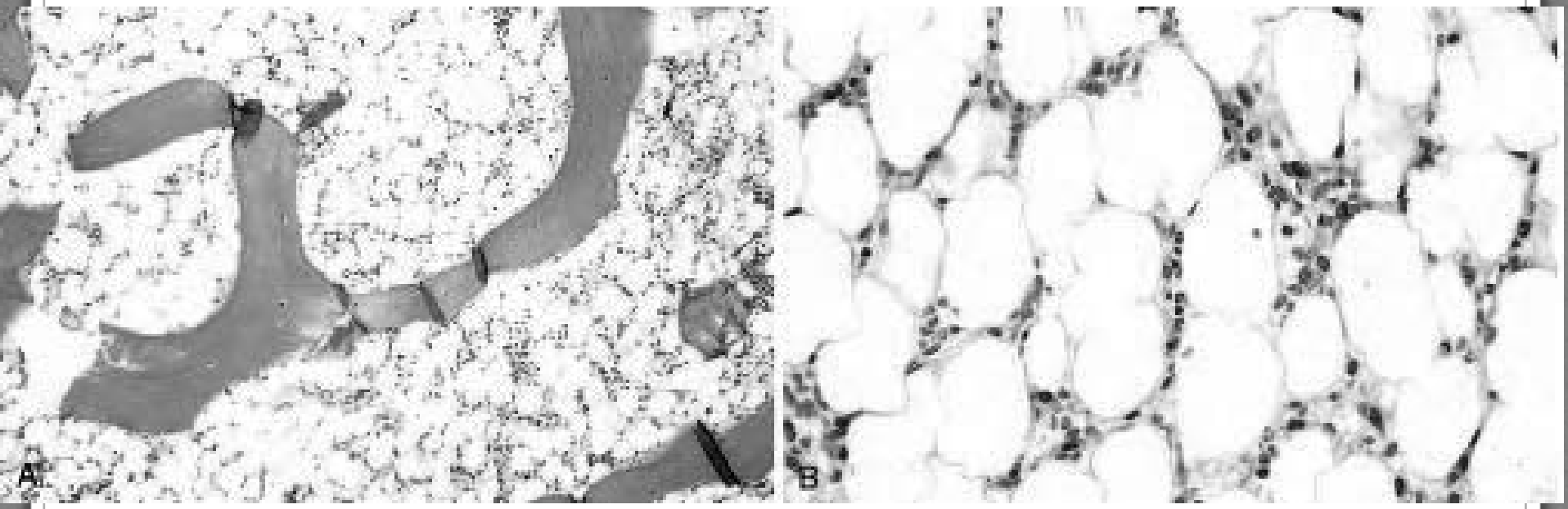
- Fanconi anemia is an autosomal recessive disease characterized by a defect in the mutliprotein complex that is required for DNA repair.



# Pathogenesis of aplastic anemia



# Morphology



# Clinical features

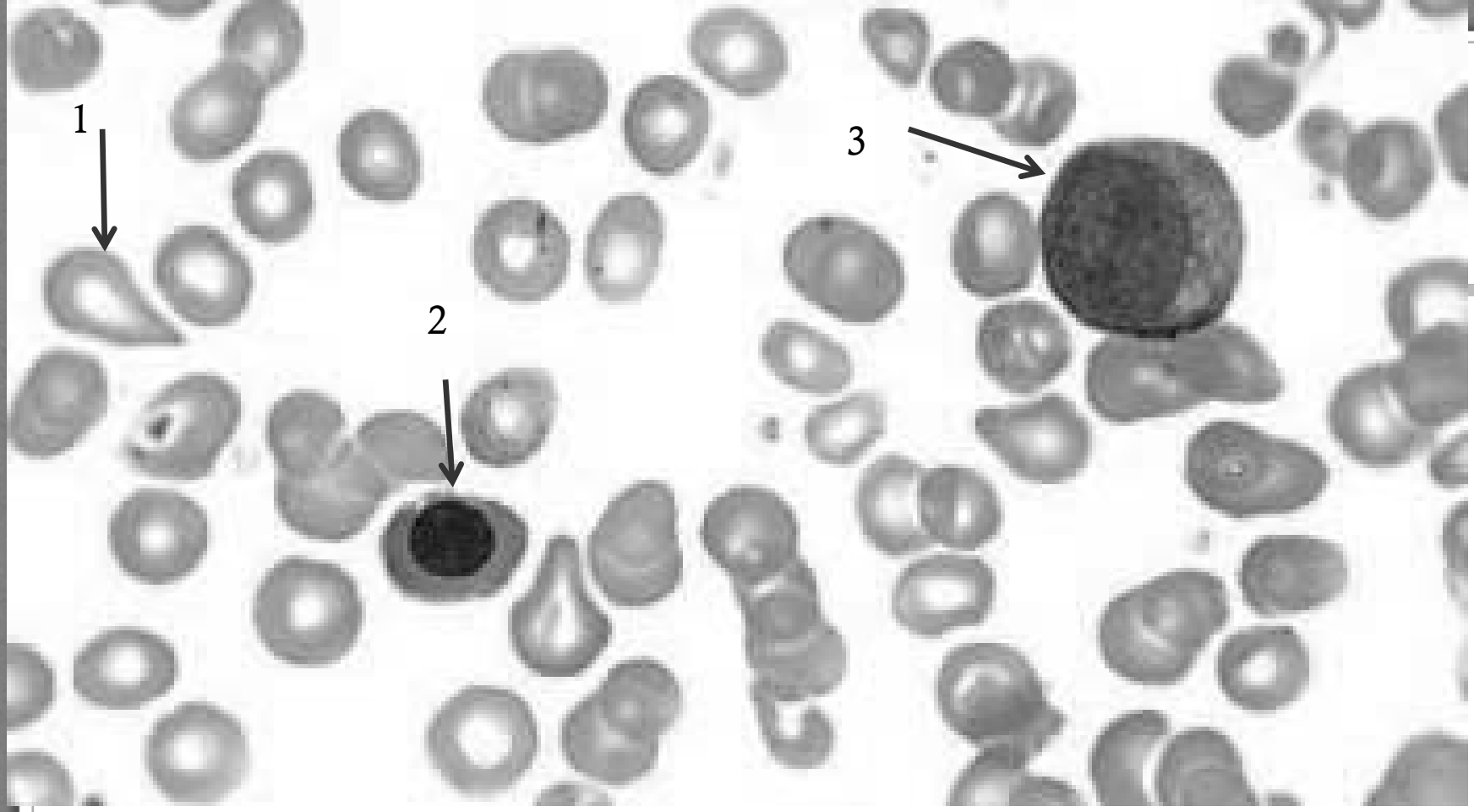
- Any age with no gender predilection
- Stigmata of pancytopenia
- Normocytic and occasionally macrocytic anemia.
- No splenomegaly
- No increased reticulocyte count
- Bone marrow exam is a must for diagnosis
- Respond well to immunosuppressive therapy, BM transplantation is the treatment of choice with 5 year survival of more than 75%.

# Myelophthisic anemia

Extensive infiltration of the marrow by tumors or other lesions.

- Metastatic cancer (lung, breast, prostate)
- Tuberculosis
- Lipid storage disorders
- Osteoscelrosis

- Leukoerythroblastic reaction on peripheral blood.




1-tear drop RBC
2-immature erythroid precursor cell
3-immture myeloid cell

- Patients present with anemia and thrombocytopenia
- WBC are usually less affected.

- **Treatment is directed at the underlying etiology.**

1. All of the following can be found in iron deficiency anemia, except:
   A. Low ferritin
   B. Low serum iron
   C. Low TIBC
   D. Low transferrin saturation
   E. Low MCV

2. Anemia of chronic disease is caused by elevated levels of:

A. Hepcidin

B. Iron

C. Ferritin

D. B12

E. Neutrophils

3. All of the following are true regarding megaloblastic anemia, except:

A. defective DNA synthesis, resulting in nuclear immaturity

B. macrocytic anemia

C. can be seen in the setting of pernicious anemia

D. most common cause is nutritional deficiency of B12

E. can be associated with neurological symptoms in the case of B12 deficiency.

4. The most common cause of anemia in patients with liver disease is:

A. Iron deficiency
B. Hypersplenism
C. Therapy related hemolytic anemia
D. Therapy related suppression of EPO receptor
E. Alcoholic-cirrhosis-induced folate deficiency

5. One of the following can cause myelophthisic anemia:

A. Tuberculosis
B. B12 deficiency
C. Folate deficiency
D. Iron deficiency
E. Anemia of chronic disease.

# THANK YOU

# RBC disorders 3

Anemia of blood loss (1)

- **Robin's Basic pathology 9th edition; Pages 408-425.**

**AND**

- **Lectures.**

- Anemia of blood loss, hemorrhage
- Hemolysis
  - extrinsic
    - Immune hemolytic anemia
    - Hemolytic anemia resulting from mechanical trauma to the red cells
    - Infection

1- Other than anemia, one of the following can be seen in the setting of anemia of hemorrhage:

A. Leukocytopenia
B. Neutropenia
C. Leukocytosis
D. Lymphocytopenia
E. Thrombocytopenia

2- Which one of the following is most helpful to differentiate between intravacular and extravscular hemolysis:

A. LDH
B. Haptoglobin
C. Bilirubin
D. Hemoglobiuria

3- Warm antibody immune hemolytic anemia is most commonly caused by:

A. IgM
B. IgA
C. IgG
D. IgD
E. IgE

4. Which one of the following is a characteristic finding in microangiopathic hemolytic anemia:

A. Target cells
B. Sickle cells
C. Spur cells
D. Ecchinocytes
E. Schistocytes

5. Which one of the following can cause cerebral malaria:

A. P.vivax
B. P. ovale
C. P. falciparum
D. P. malarie

- Anemia of blood loss, hemorrhage
- Hemolysis
  - extrinsic
    - Immune hemolytic anemia
    - Hemolytic anemia resulting from mechanical trauma to the red cells
    - Infection

- intrinsic
  - Hereditary
    - Membranopathies-spherocytosis
    - Hemoglobinopathies-thalassemia and sickle cell disease
    - Enzymopathies-G6PD deficiency
  - Acquired
    - Paroxysmal nocturnal hemoglobinuria.

- **Anemia of blood loss, hemorrhage**
- Hemolysis
  - extrinsic
    - Immune hemolytic anemia
    - Hemolytic anemia resulting from mechanical trauma to the red cells
    - Infection

# Anemia of blood loss

- It has two types
  - Acute
  - Chronic

- Acute: resulting from external (wound) or internal (rupture aortic aneurysm) hemorrhage.
- If the blood loss is less than 20% of the total volume, healthy patients can tolerate that with a few symptoms
- If it exceeds 20% the immediate threat is from hypovolemia not anemia.

- The full effect of anemia starts to appear after 2-3 days when the fluid shifts into the intravascular space.
- Erythropoietin will be released recruiting more stem cells to proliferate and differentiate into red cells.

- Normochromic normocytic anemia
- Leukocytosis
- As Red cell production increases, it turns into slightly macrocytic anemia
- Thrombocytosis upon recovery.

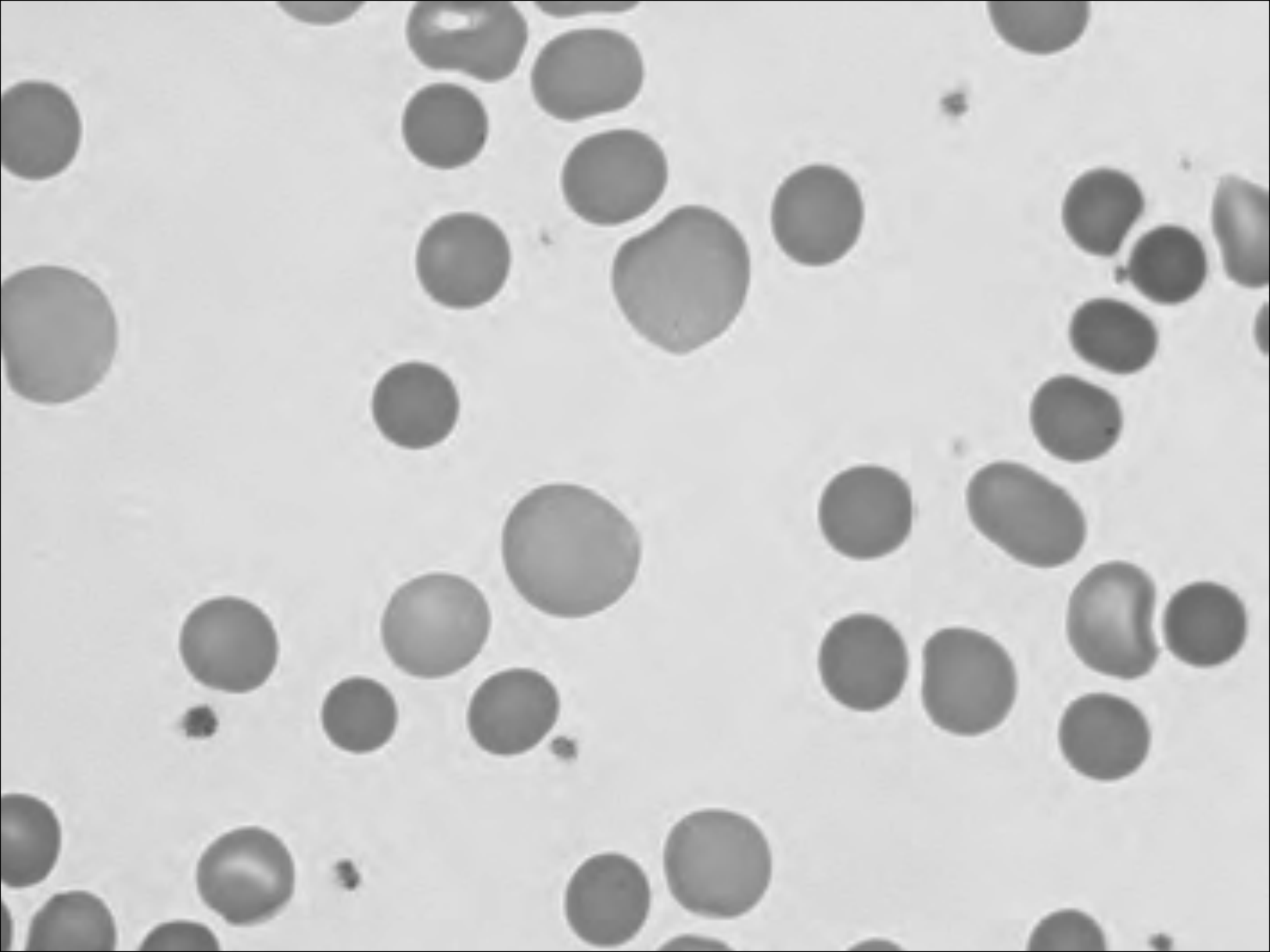
- Anemia of chronic blood loss is iron deficiency anemia, discussed earlier.

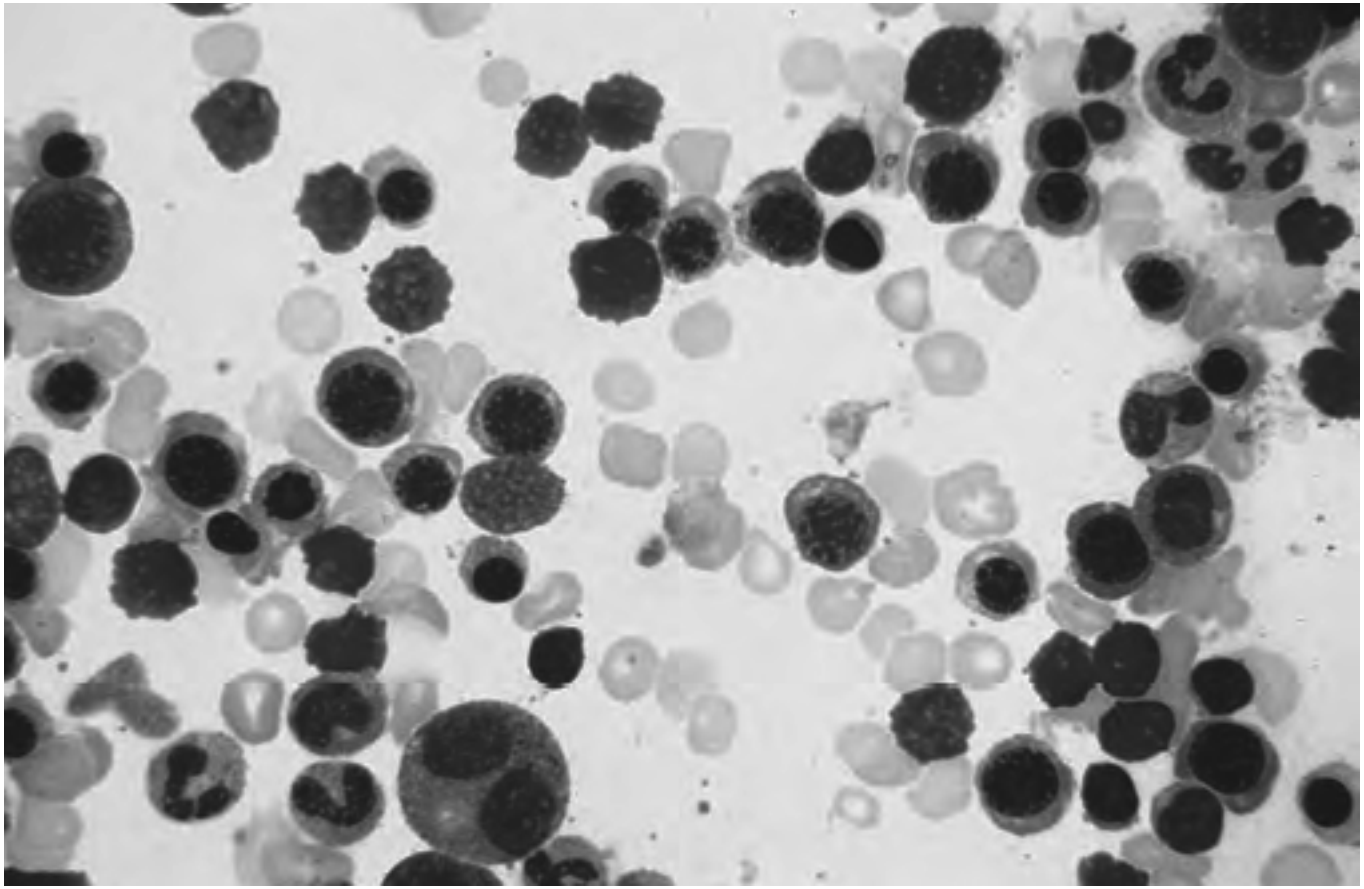
# Hemolysis-general

- Increase destruction of red cells resulting in decreased RBC survival.
- Elevated erythropoietin levels
- Accumulation of hemoglobin degradation products, bilirubin and iron.
- Erythroid precursor hyperplasia in the bone marrow and reticulocyte counts in the blood.

- Extravascular or intravascular.
  - Extravascular: spleen or liver
    - No hemoglobinuria or hemoglobinemia
    - Low haptoglobin
    - High LDH
    - Splenomegaly.
    - Jaundice; high bilirubin and possibly gallbladder stones.

- Intravascular
  - Hemoglobinemia and hemoglobiuria
  - Low haptoglobin
  - High LDH
  - Splenomegaly.
  - Jaundice; high bilirubin and possibly gallbladder stones.





- Anemia of blood loss, hemorrhage
- **Hemolysis**
  - **extrinsic**
    - **Immune hemolytic anemia**
    - Hemolytic anemia resulting from mechanical trauma to the red cells
    - Infection

# Immune hemolytic anemia

*Hemolytic anemias in this category are caused* ***by antibodies that bind to red cells****, leading to their premature destruction.*

## Warm Antibody Type

*Primary* (idiopathic)

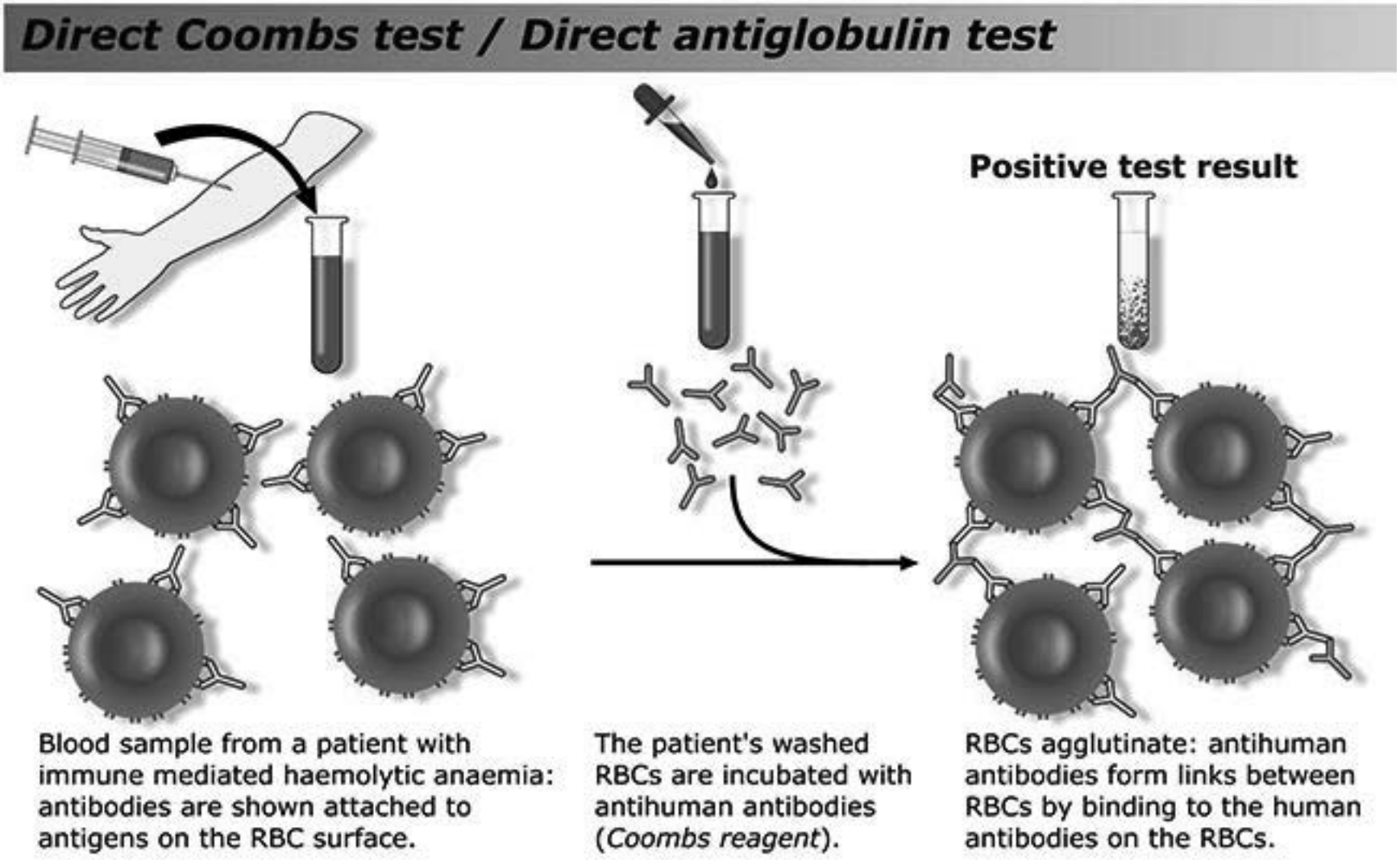
*Secondary*: B cell neoplasms (e.g., chronic lymphocytic leukemia), autoimmune disorders (e.g., systemic lupus erythematosus), drugs (e.g., α-methyldopa, penicillin, quinidine)

## Cold Antibody Type

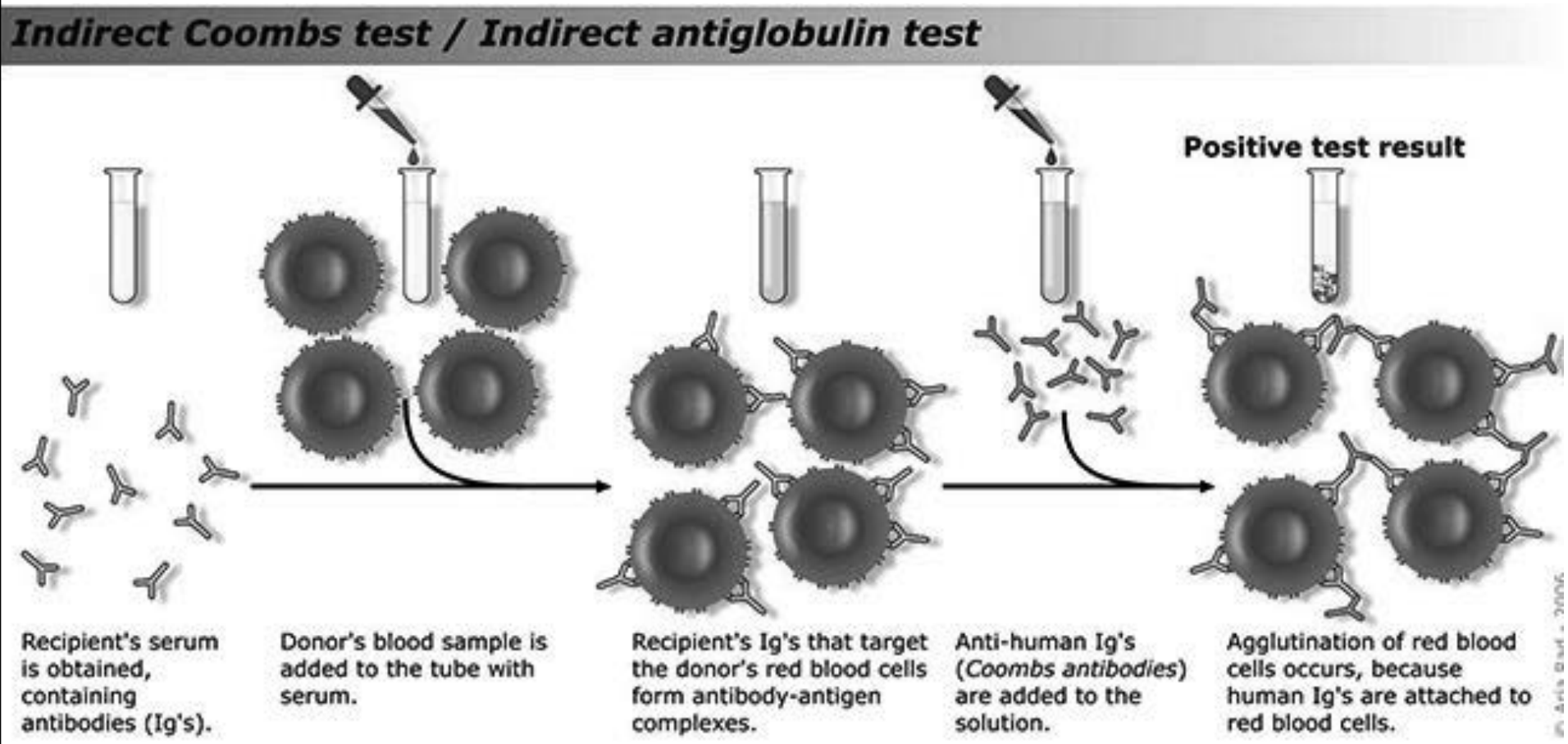
*Acute: Mycoplasma* infection, infectious mononucleosis

*Chronic*: idiopathic, B cell lymphoid neoplasms (e.g., lymphoplasmacytic lymphoma)

# Coombs test-direct



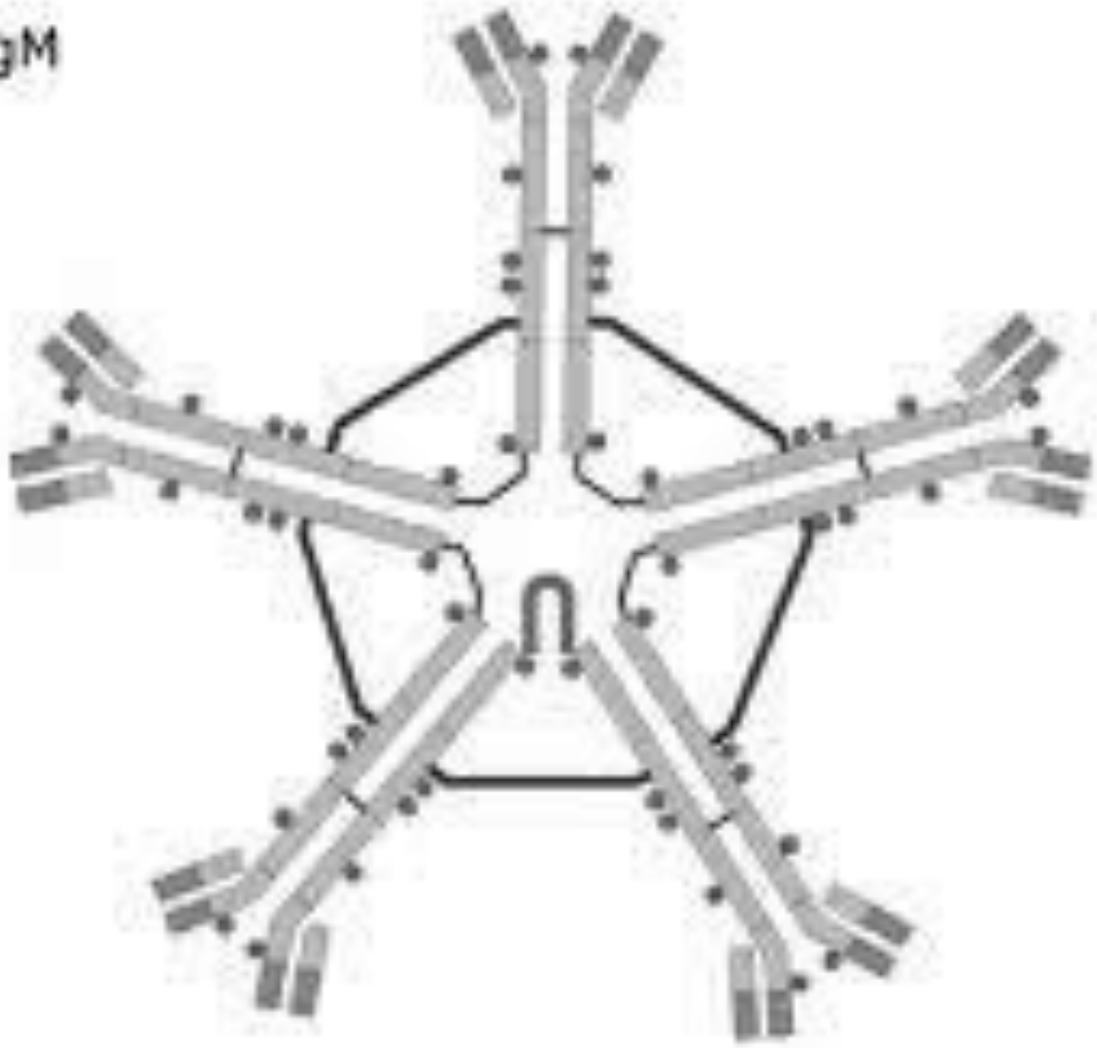
# Coombs test-indirect

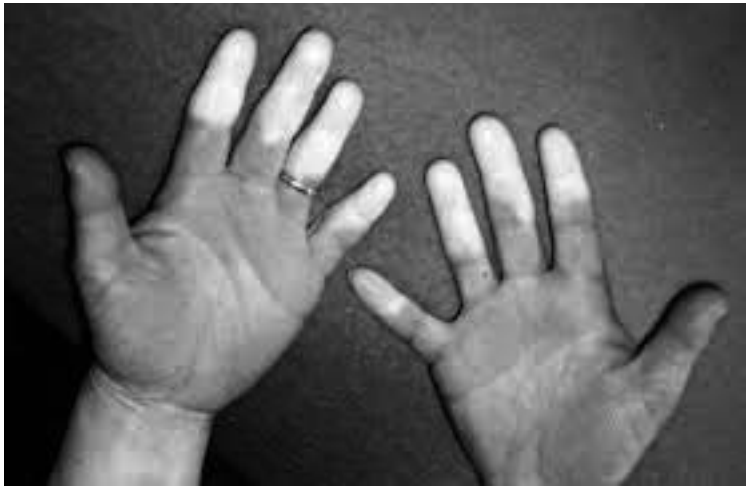


- Warm immunohemolytic anemia:
  - IgG and rarely IgA
  - 37 C°
  - Over 60% idiopathic.
  - Remaining minority is caused by autoimmune diseases (SLE), B cell neoplasms (CLL), or drugs (methyl-dopa, penicillin).
  - Most patients have mild anemia with splenomegaly and require no treatment.

- Cold immunohemolytic anemia:
  - IgM
  - Low temperatures in cold weather
  - Extravascular hemolysis.
  - Could be idiopathic
  - Secondary to mycoplasma, infectious mononucleosis, B cell neoplams (lymphoplamsacytic lymphoma)
  - Usually mild without clinical significance.

IgM

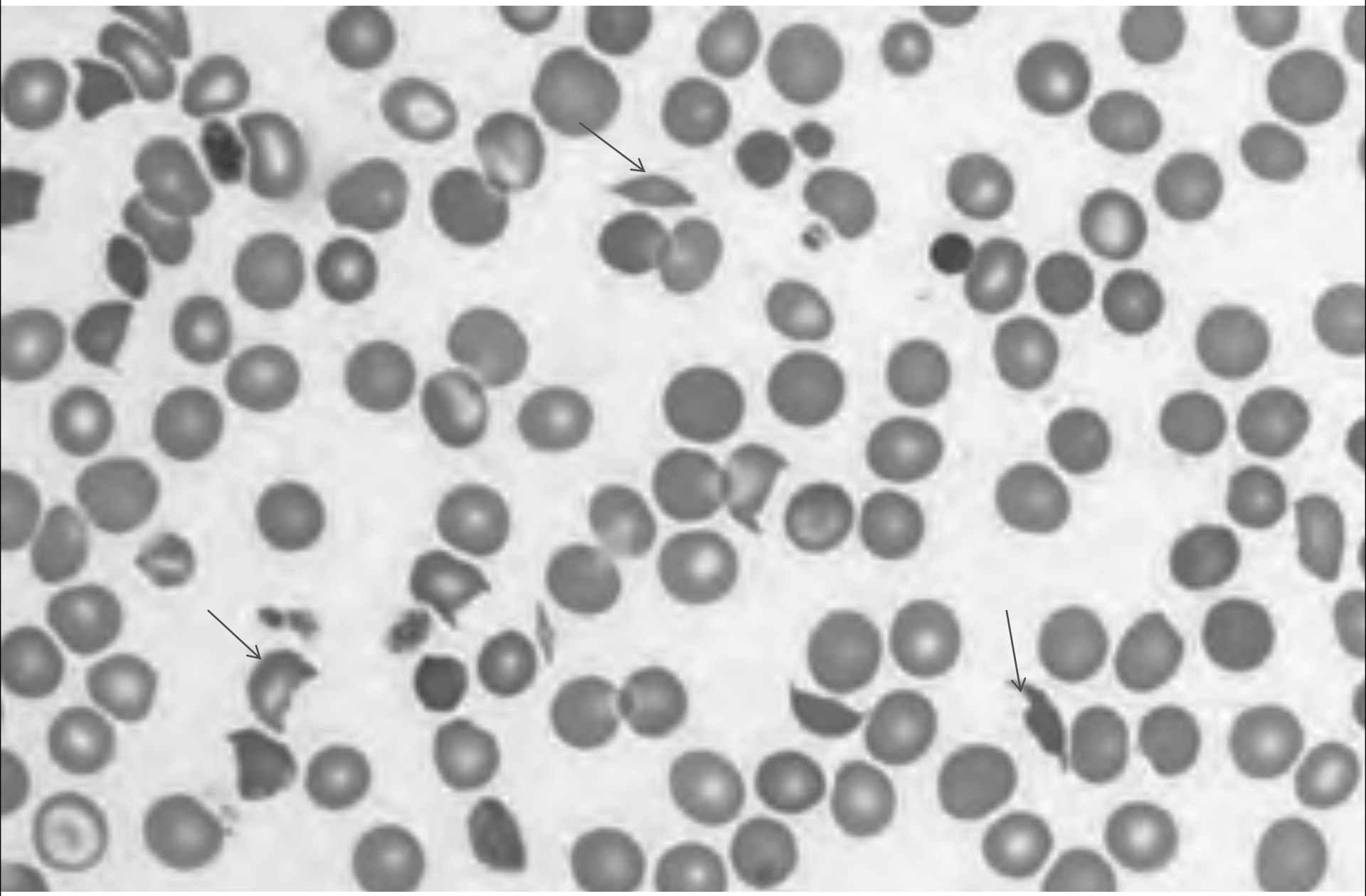




- Anemia of blood loss, hemorrhage
- Hemolysis
  - extrinsic
    - Immune hemolytic anemia
    - **Hemolytic anemia resulting from mechanical trauma to the red cells**
    - Infection

# Hemolytic anemia resulting from mechanical trauma to the red cells

- Repeated physical activity
- Cardiac valves
- Microangiopathic hemolytic anemia
  - DIC, most commonly
  - Malignant hypertension
  - SLE
  - Thrombotic thrombocytopenic purpura
  - Hemolytic uremic syndrome
  - Disseminated cancer



schistocytes

- Microangiopathic hemolytic anemia is not, by itself, a serious disease, but it points to a serious underlying disorders.

- Anemia of blood loss, hemorrhage
- Hemolysis
  - extrinsic
    - Immune hemolytic anemia
    - Hemolytic anemia resulting from mechanical trauma to the red cells
    - **Infection**

# infection

- Malaria:
  - Parasites within the RBCs, rupture resulting in hemolysis and episodic symptoms
  - Hematin released from the RBCs results in brown pigmentation of the spleen, liver and bone marrow
  - Massive splenomegaly and occasional hepatomegaly.
  - Falciparum can cause cerebral malaria which can be fatal.

1- Other than anemia, one of the following can be seen in the setting of anemia of hemorrhage:

A. Leukocytopenia
B. Neutropenia
C. Leukocytosis
D. Lymphocytopenia
E. Thrombocytopenia

2- Which one of the following is most helpful to differentiate between intravacular and extravscular hemolysis:

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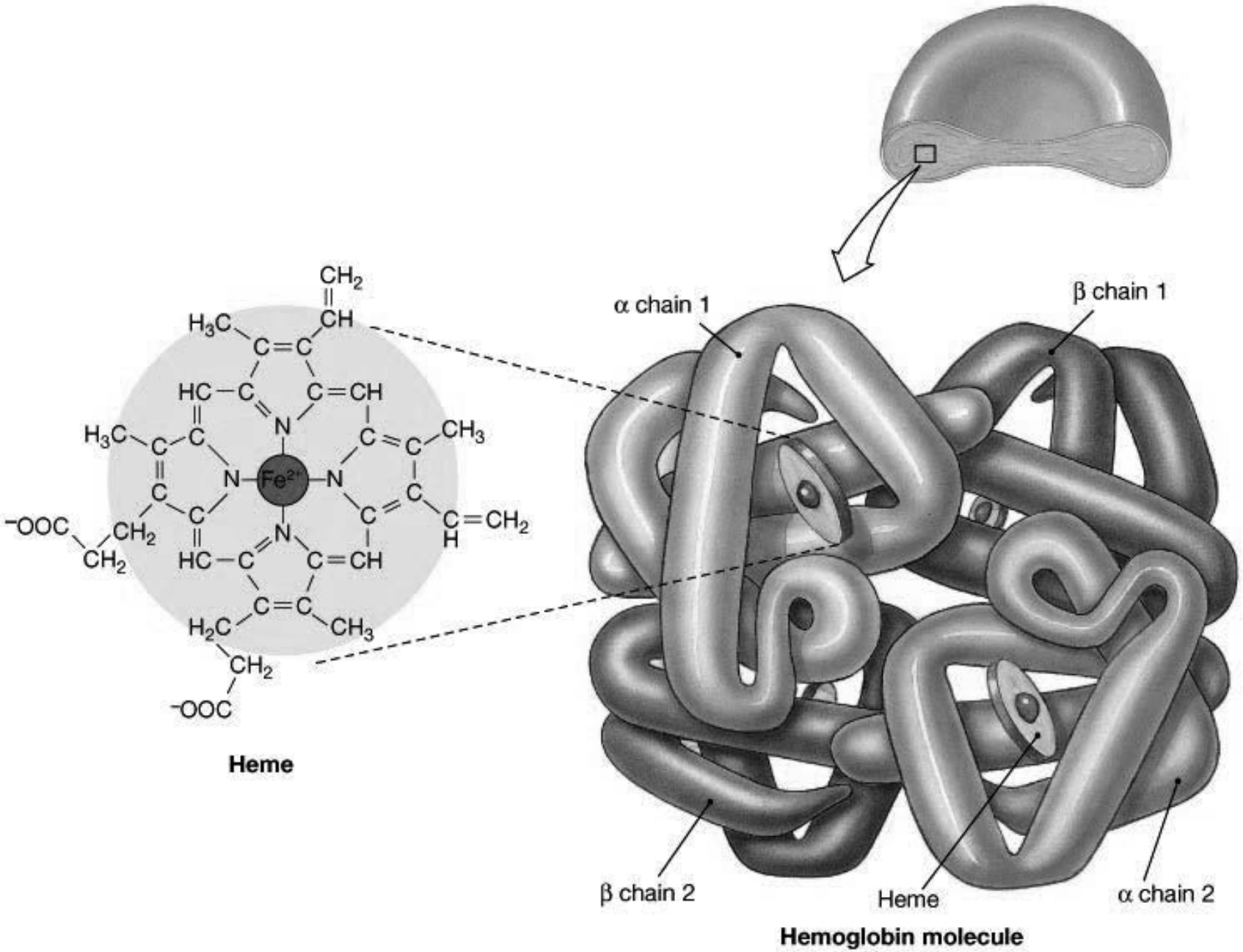
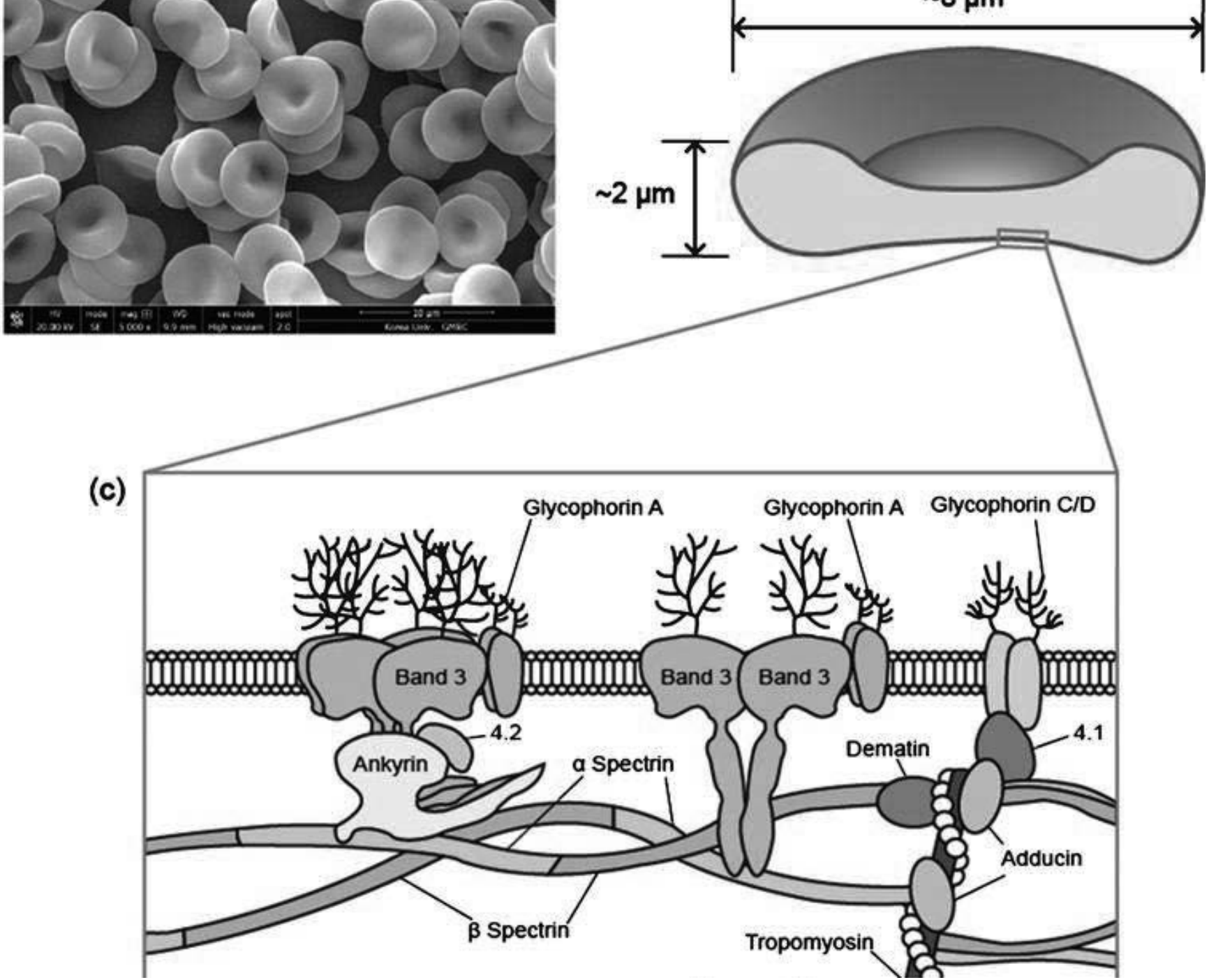
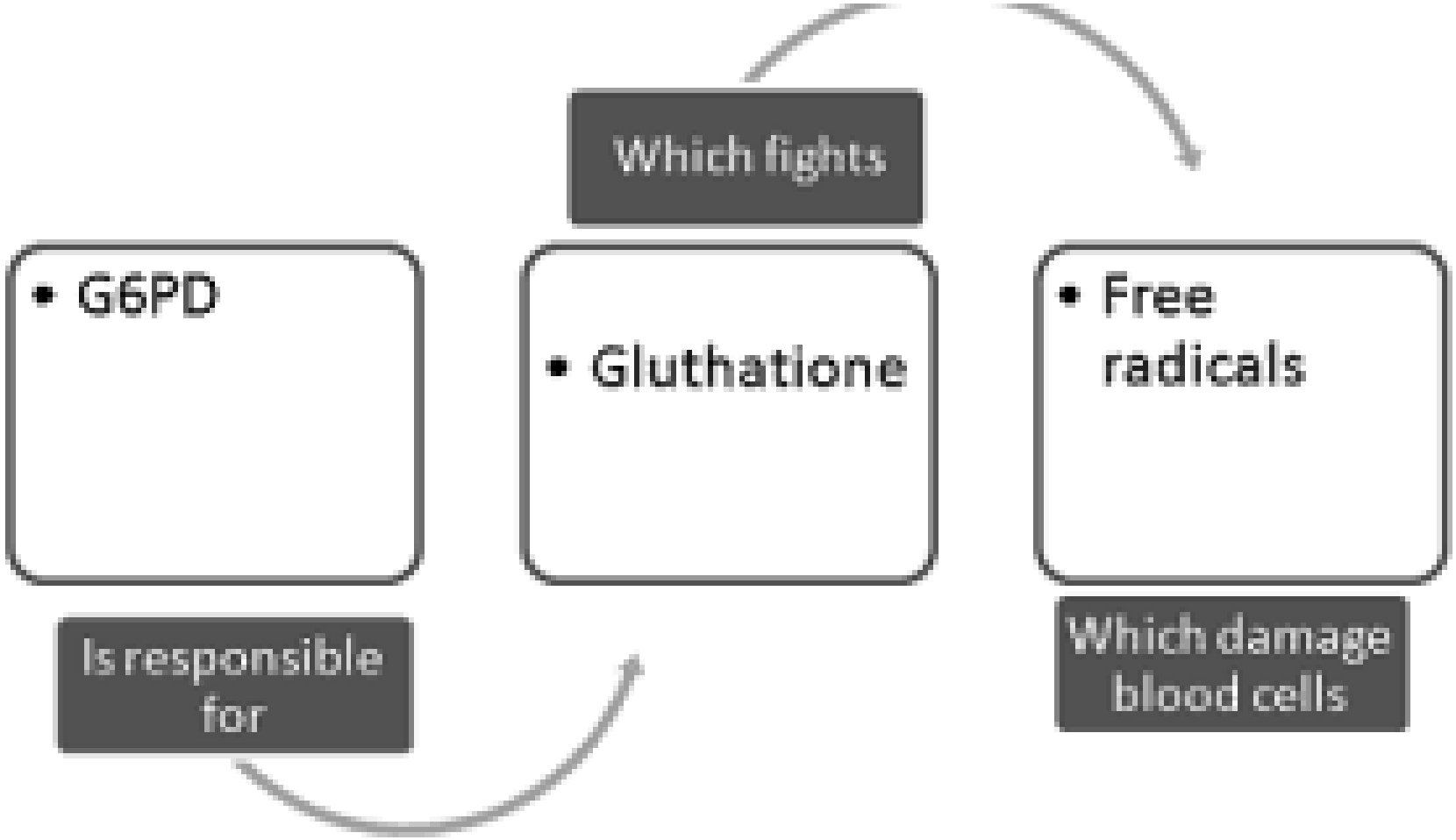
# THANK YOU

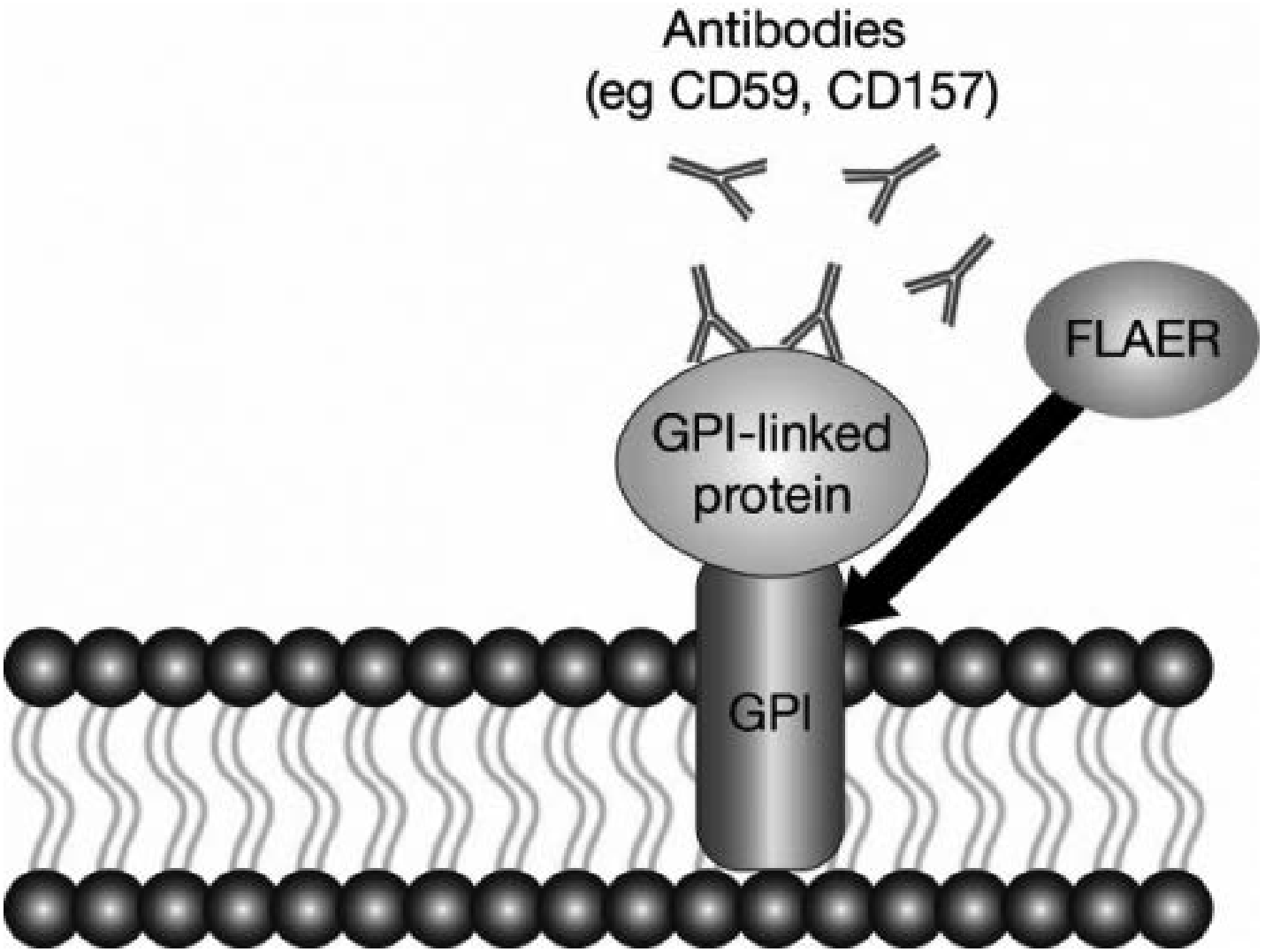
# RBC disorders 4

Ahmad Mansour, MD


α chain 1
β chain 1
β chain 2
Heme
α chain 2
Heme
Hemoglobin molecule


~2 μm
(c)
Glycophorin A
Glycophorin A
Glycophorin C/D
Band 3
Band 3
Band 3
4.2
Ankyrin
α Spectrin
Dematin
4.1
Adducin
β Spectrin
Tropomyosin


• G6PD
Is responsible for
• Gluthatione
Which fights
• Free radicals
Which damage blood cells


Antibodies
(eg CD59, CD157)
FLAER
GPI-linked
protein
GPI

1- what is the mode of inheritance in the vast majority f spherocytosis cases?

A. Autosomal dominant
B. Autosomal recessive
C. X-linked dominant
D. X linked recessive

2- The amino acid present at the sixth position of the normal alpha-globin chain is replaced by which one of the following amino acids in sickle cell disease?

A. Lysine
B. Valine
C. Serine
D. Alanine
E. None of the above

3- In thalassemia disorders, when only one alpha gene is affected, what do we call that?

A. Normal
B. Silent carrier
C. Thalassemia trait-*cis*
D. Thalassemia trait-*trans*
E. HbH disease

4- gallbladder stones are a frequent complication of G6PD deficiency?

TRUE FALSE

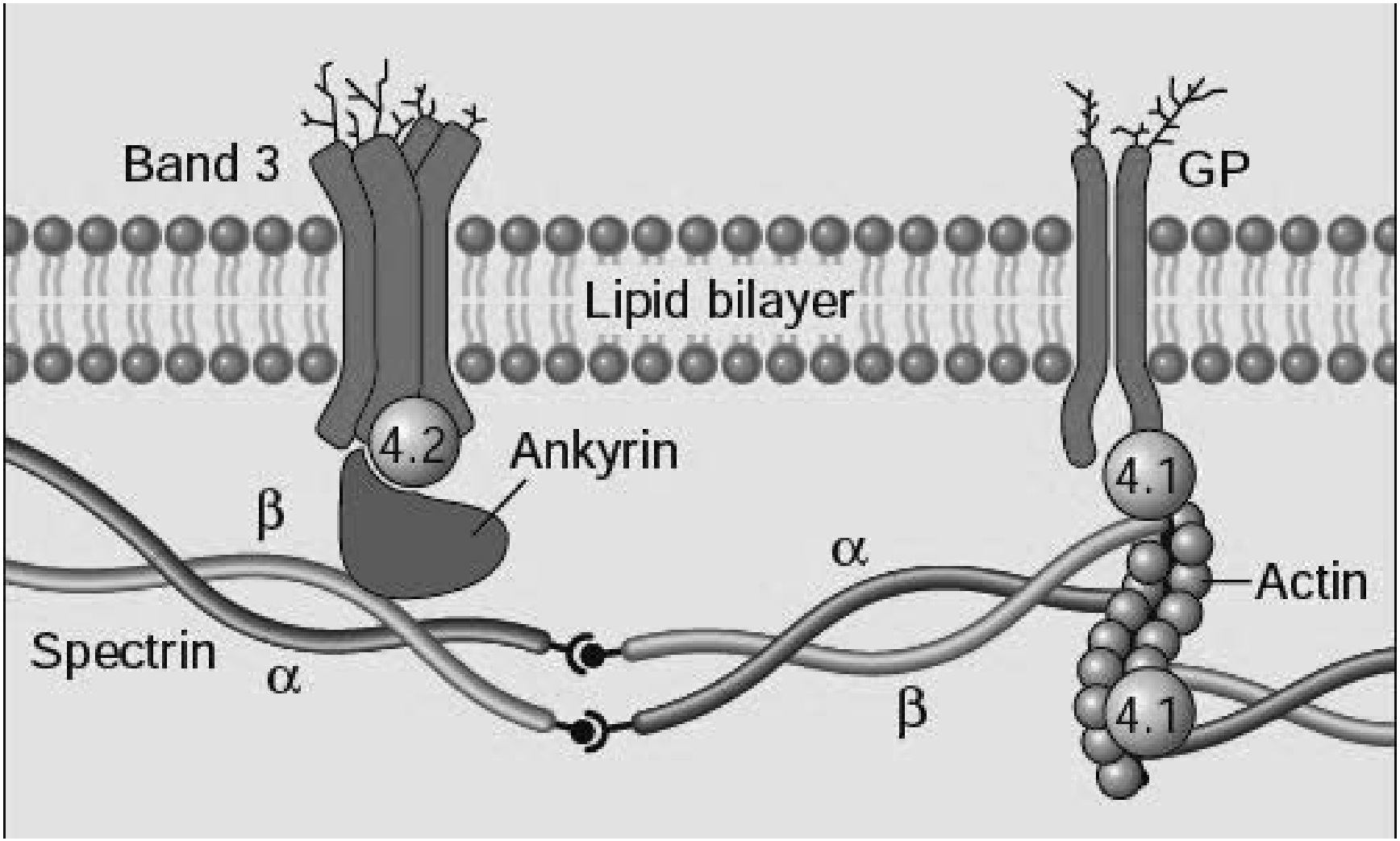
5- Paroxysmal nocturnal hemoglobinuria results from an acquired mutation in which of the following genes:

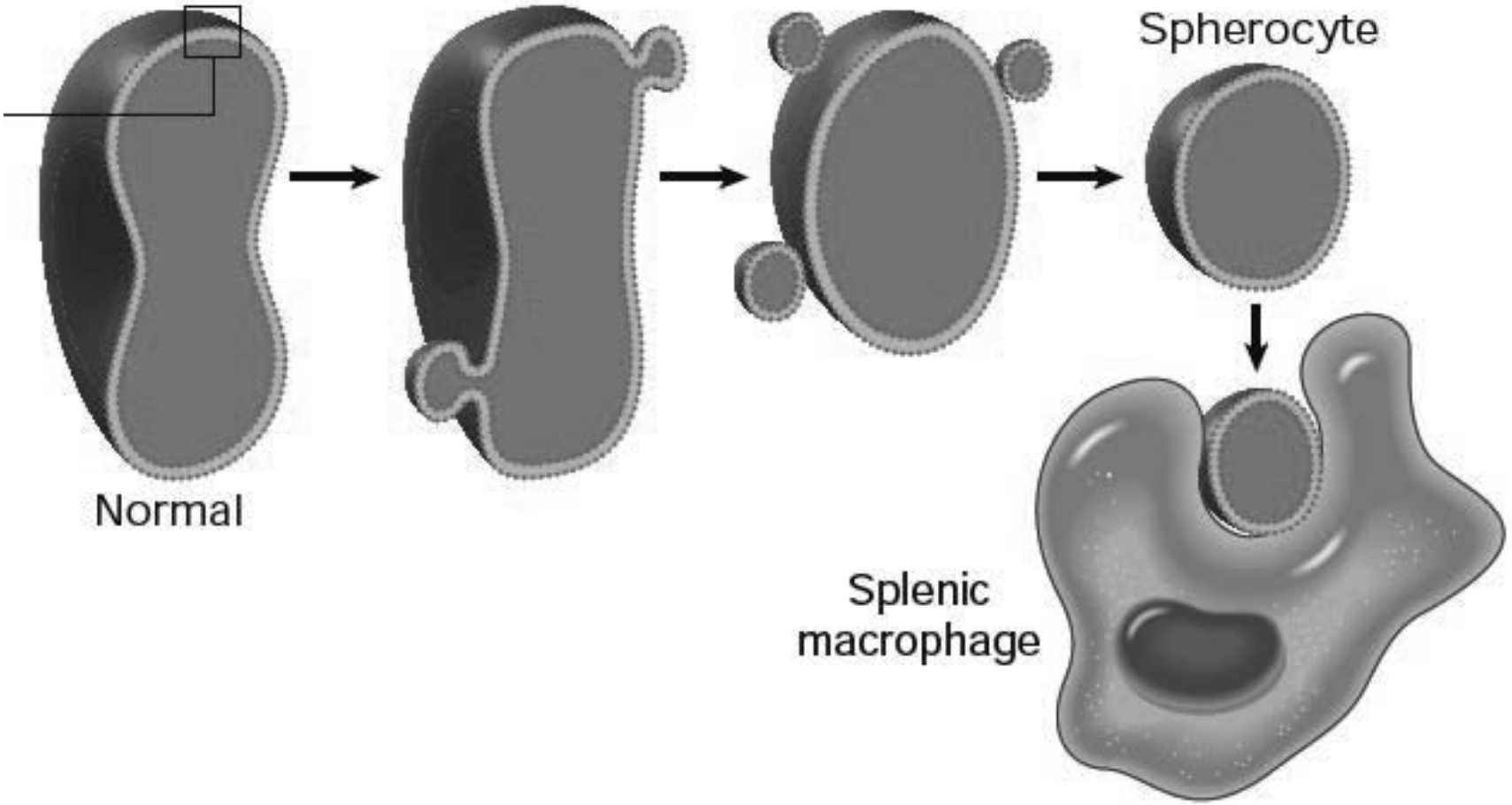
A. Alpha hemoglobin
B. Beta hemoglobin
C. Erythropoietin
D. PIGA
E. G6PD

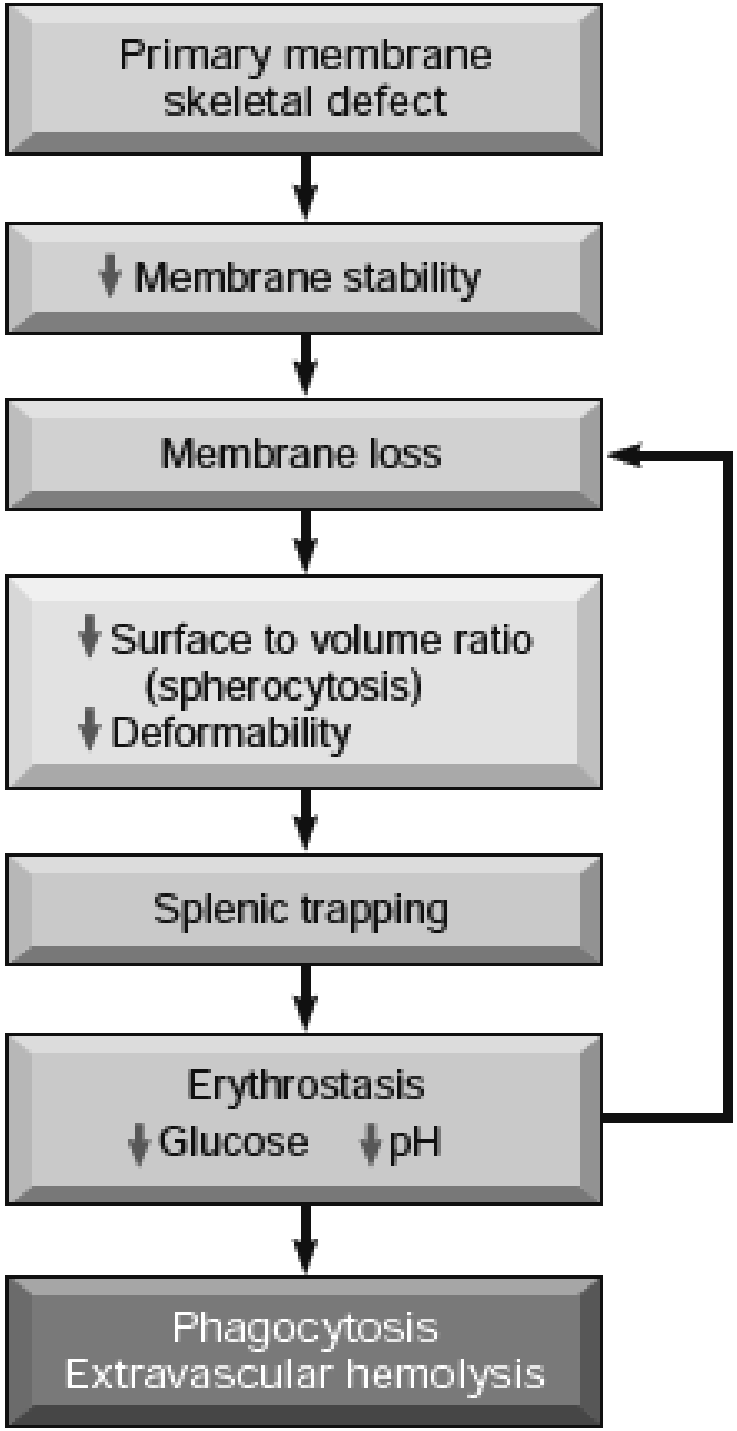
- Anemia of blood loss, hemorrhage
- Hemolysis
  - extrinsic
    - Immune hemolytic anemia
    - Hemolytic anemia resulting from mechanical trauma to the red cells
    - Infection

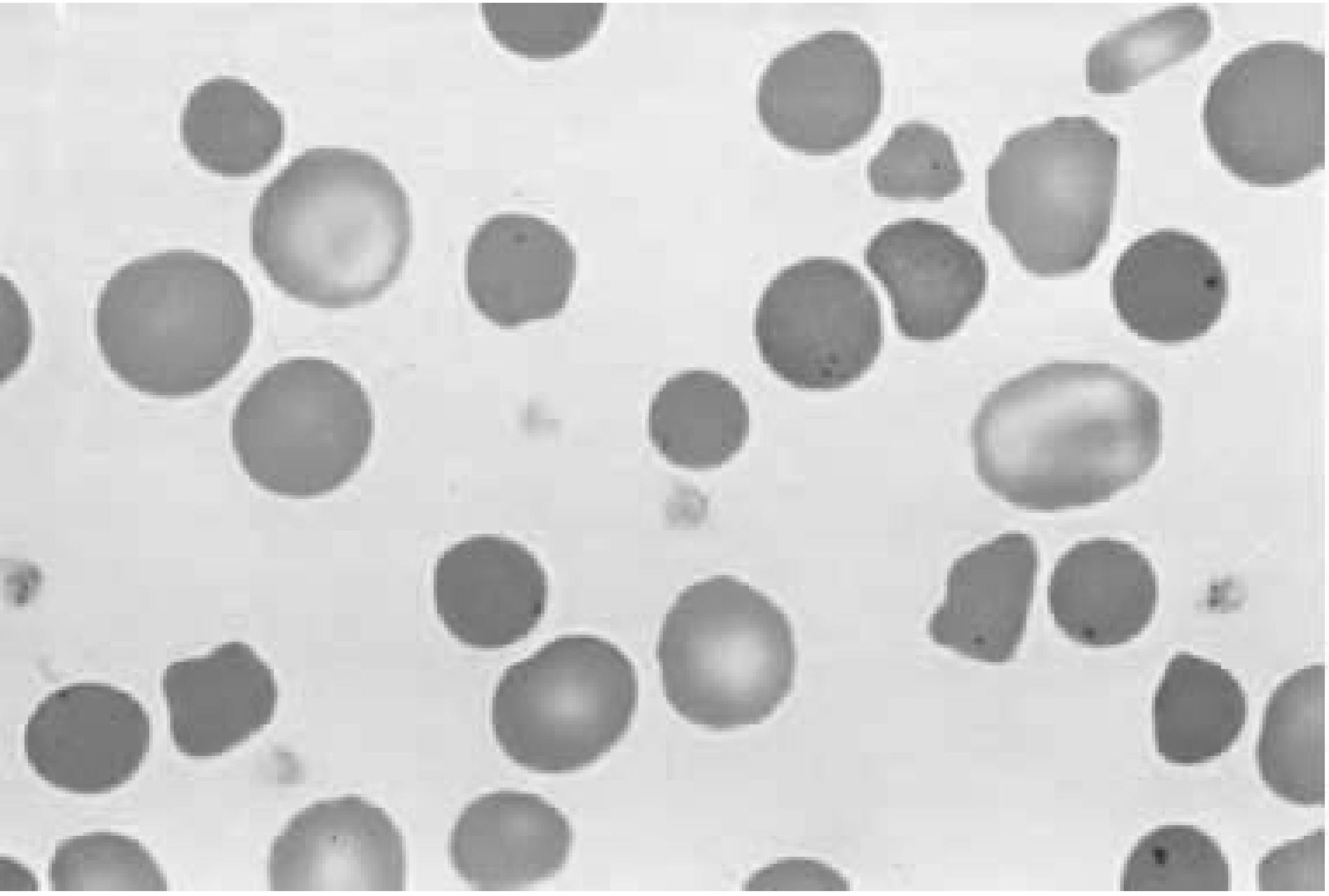
- intrinsic
  - Hereditary
    - Membranopathies-spherocytosis
    - Hemoglobinopathies-thalassemia and sickle cell disease
    - Enzymopathies-G6PD deficiency
  - Acquired
    - Paroxysmal nocturnal hemoglobinuria.

# Hereditary spherocytosis




Spherocyte
Normal
Splenic
macrophage

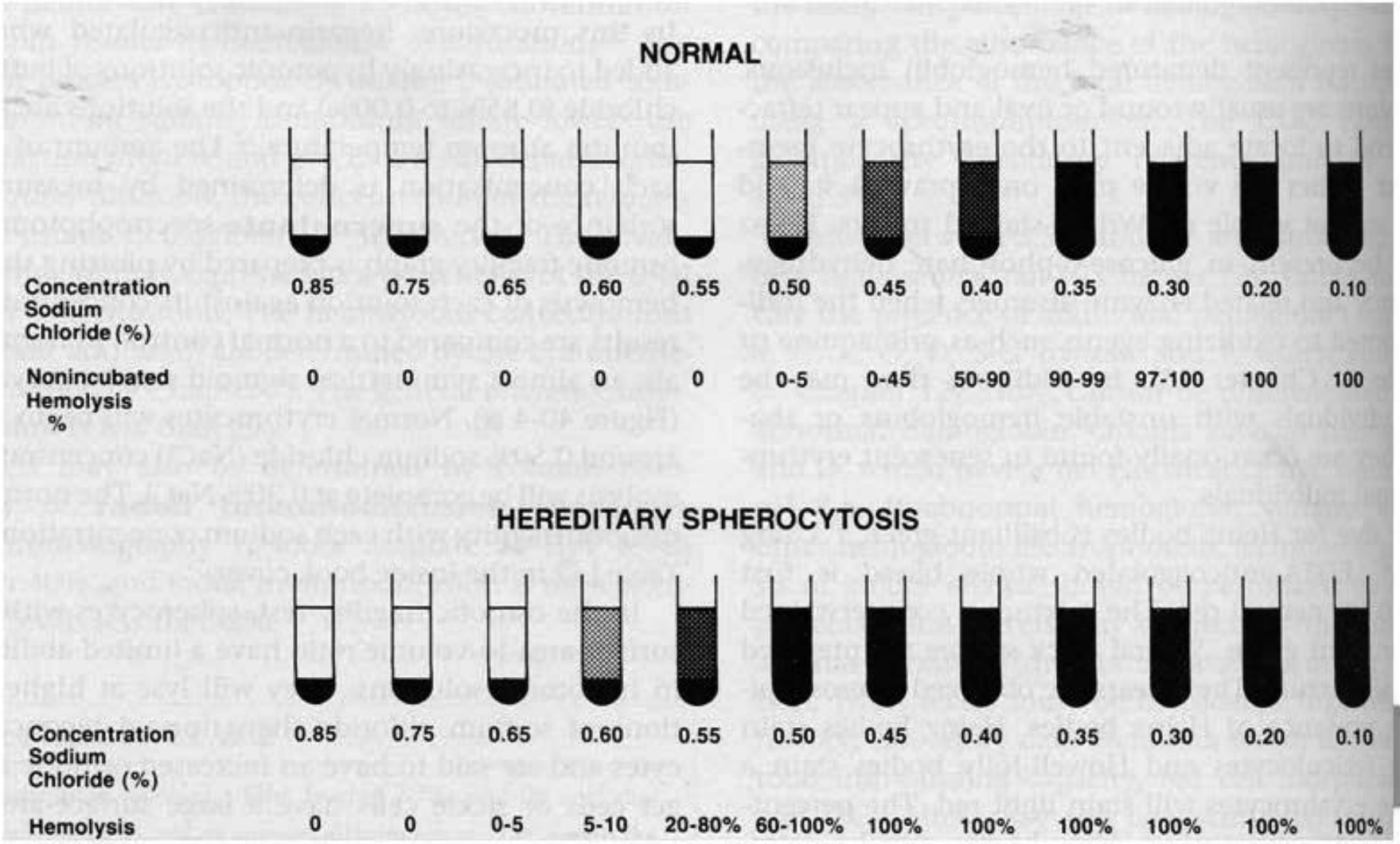

Primary membrane skeletal defect
↓ Membrane stability
Membrane loss
↓ Surface to volume ratio (spherocytosis)
↓ Deformability
Splenic trapping
Erythrostasis
↓ Glucose ↓ pH
Phagocytosis
Extravascular hemolysis



- Mostly autosomal dominant
- Prevalent in north Europe
- Mutation in ankyrin, band 3, and spectrin.

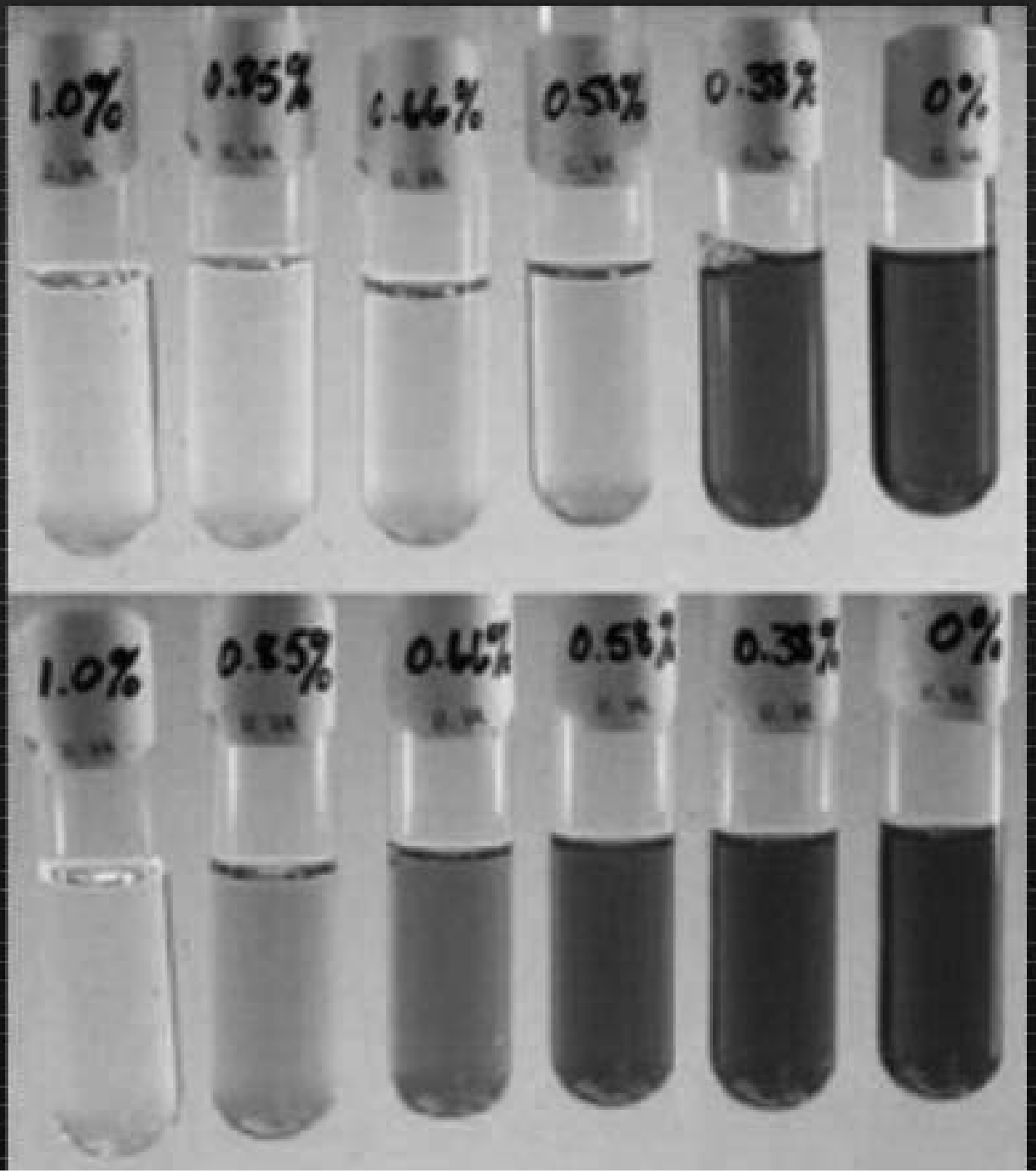
- Moderate clinical course, mostly.
- Can be complicated by aplastic  crisis (parvo B19).
- Anemia, jaundice, gallbladder stones, splenomegaly.
- MCHC is high.
- Diagnosis involves osmotic fragility test
- No definitive treatment
  - Symptomatic treatment with splenectomy.

# OSMOTIC FRAGILITY TEST



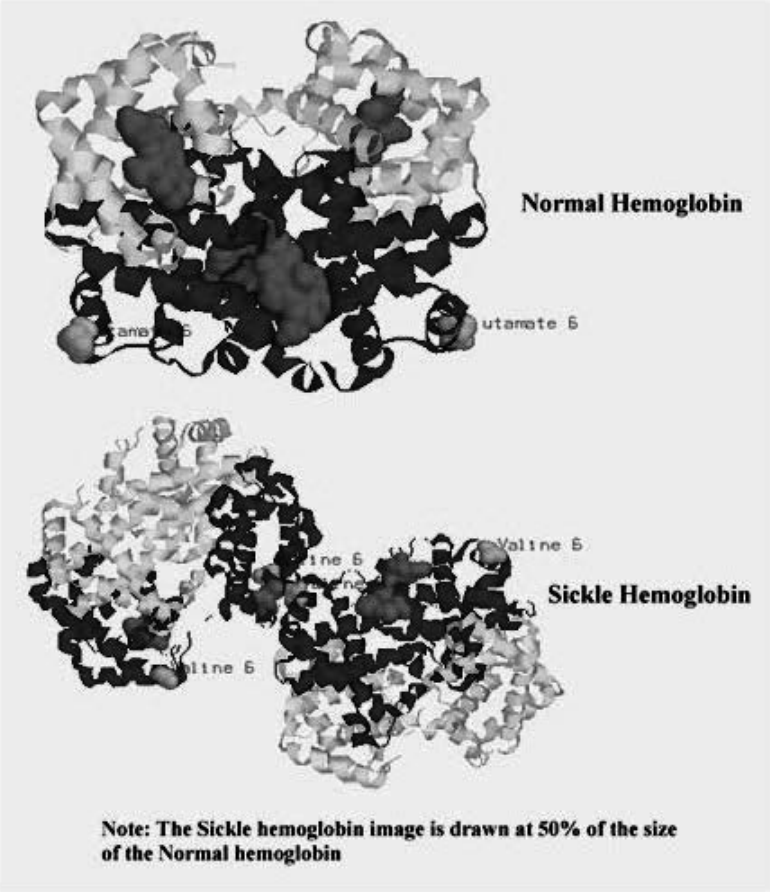
Normal

**Abnormal-**
HS cells lyse more readily at low ionic strength

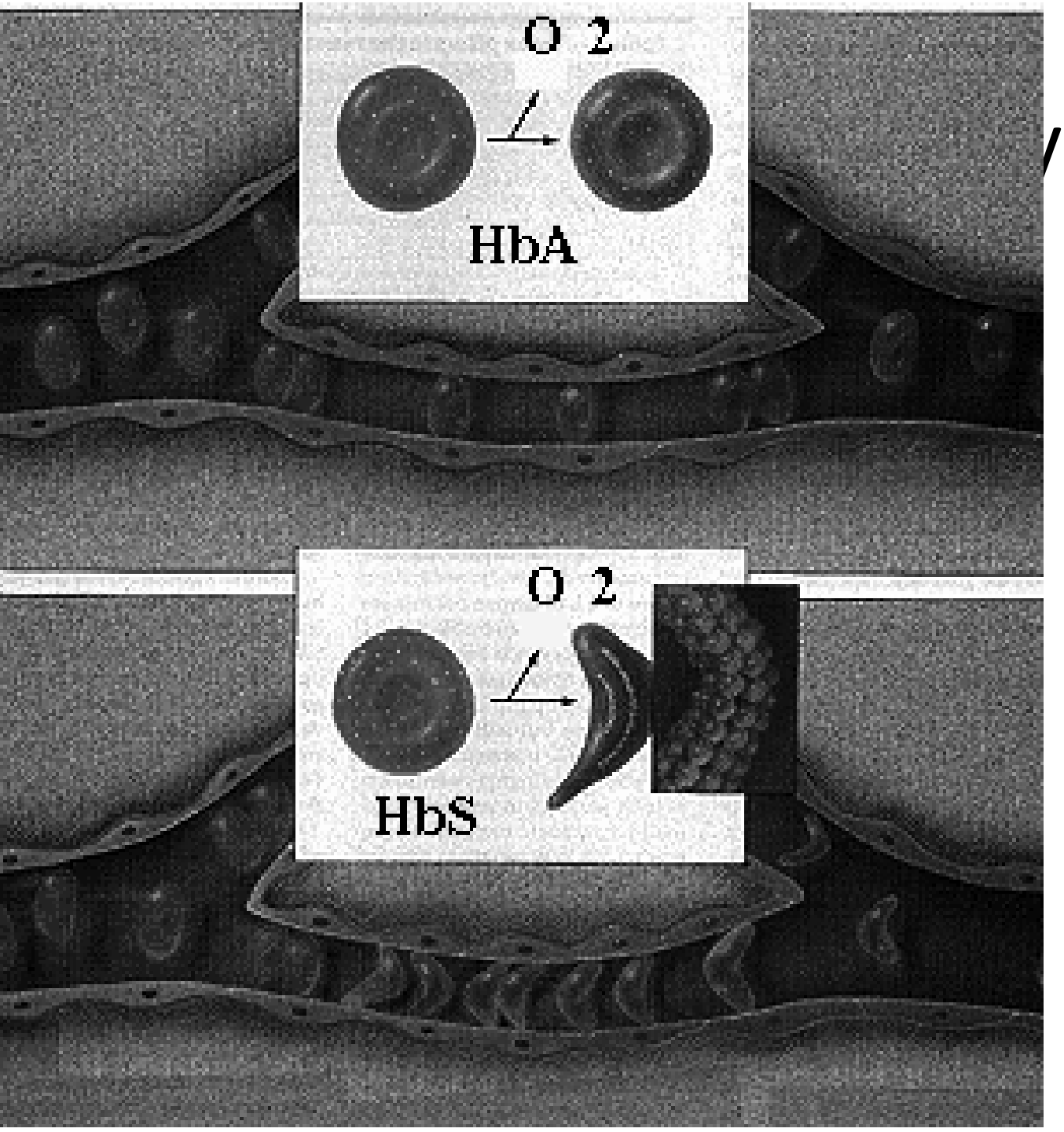


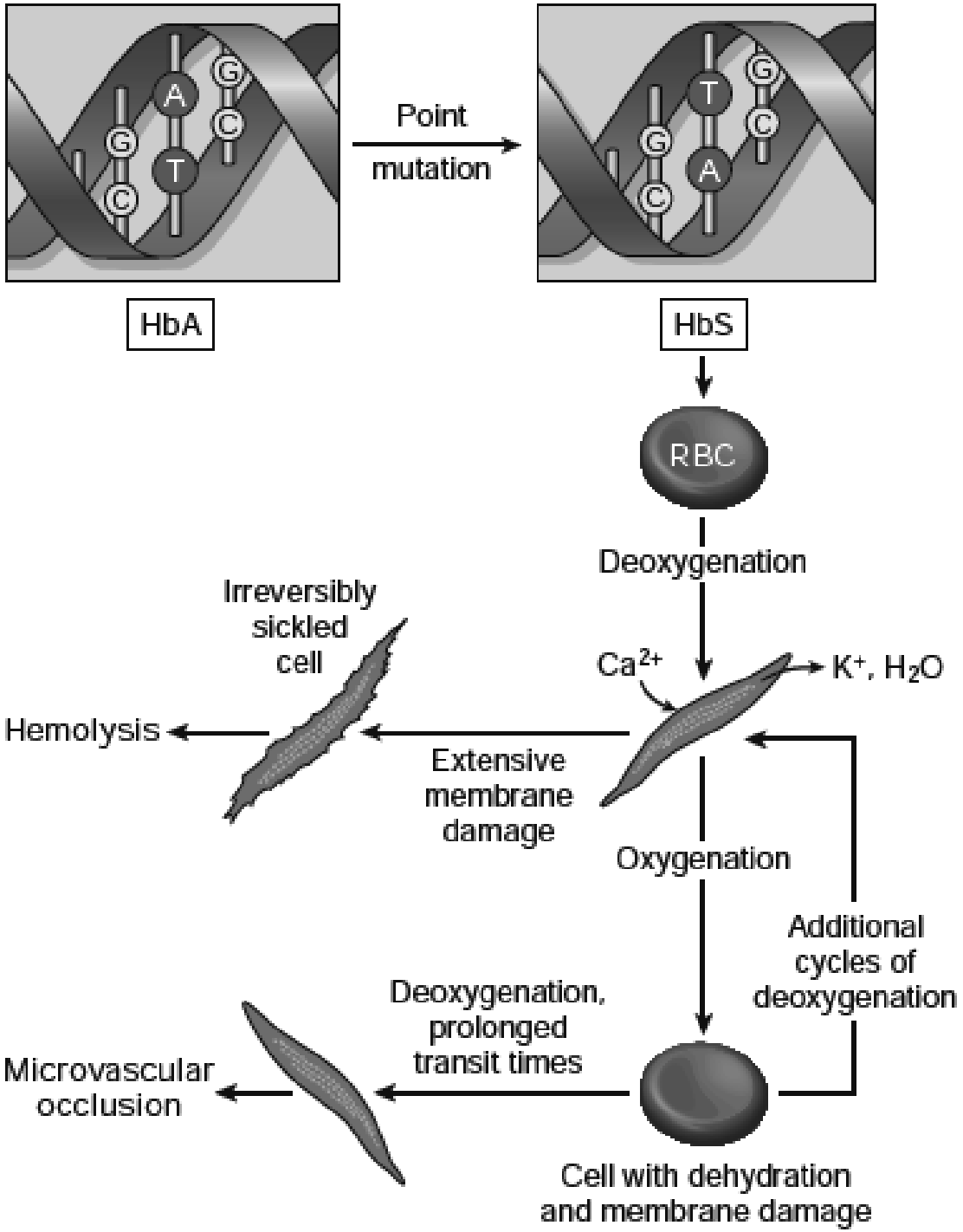
- intrinsic
  - Hereditary
    - Membranopathies-spherocytosis
    - Hemoglobinopathies-thalassemia and sickle cell disease
    - Enzymopathies-G6PD deficiency
  - Acquired
    - Paroxysmal nocturnal hemoglobinuria.

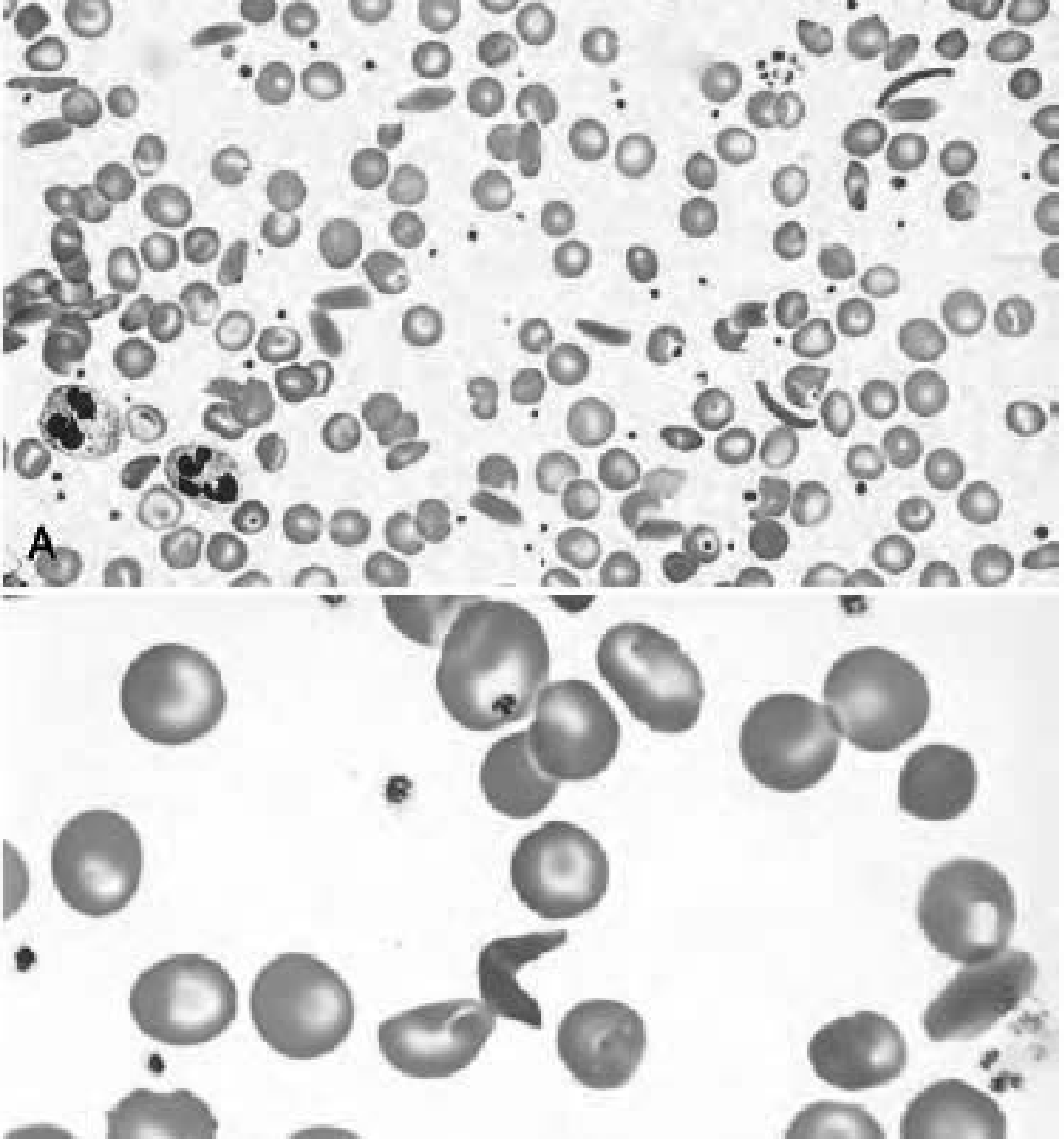
# Sickle cell anemia



- The most common hemoglobinpathy
- In homozygotes all HB is replaced by HbS
- In heterozygotes half is replaced.
- Gene frequency is ~30%
- 8% in black Americans.


O 2
HbA
O 2
HbS


A
G
C
T
G
C
Point
mutation
T
G
C
A
G
C
HbA
HbS
RBC
Deoxygenation
Irreversibly
sickled
cell
Ca2+
K+, H2O
Hemolysis
Extensive
membrane
damage
Oxygenation
Additional
cycles of
deoxygenation
Deoxygenation,
prolonged
transit times
Microvascular
occlusion
Cell with dehydration
and membrane damage


A

- Three important factors influence sicling in the body
  - Presence of hemoglobins other than HbS
  - Intracellular concentration of hemoglobin
  - Transit time for RBCs within the vasculature

Presence of hemoglobins other than HbS

- HBA (α2β2)…weak
- HbF(α2γ2)…weak
- HbC…strong

## Intracellular concentration of hemoglobin

- Dehydration…..high conc.
- Alpha thalassemia….low conc.

Transit time for RBCs within the vasculature

- Short time….no sickling
- Long time….sickling

- Chronic hemolytic anemia
- Fatty change in the heart, liver and renal tubules
- Reticulocytosis and erythroid hyperplasia in bone marrow
- Bone changes, prominent cheekbones and crew-cut skull
- Extramedullary hematopoiesis in liver and spleen.

- Mild splenic congestion, autosplenectomy in adults.
- Increased risk of infections, salmonella osteomyelitis.
- Vessel occlusion, bone pain, acute chest syndrome, stroke.
- Aplastic crisis

- Diagnosis with electrophoresis to demonstrate HbS and fetal DNA via amniocentesis or chorionic villi biopsy.

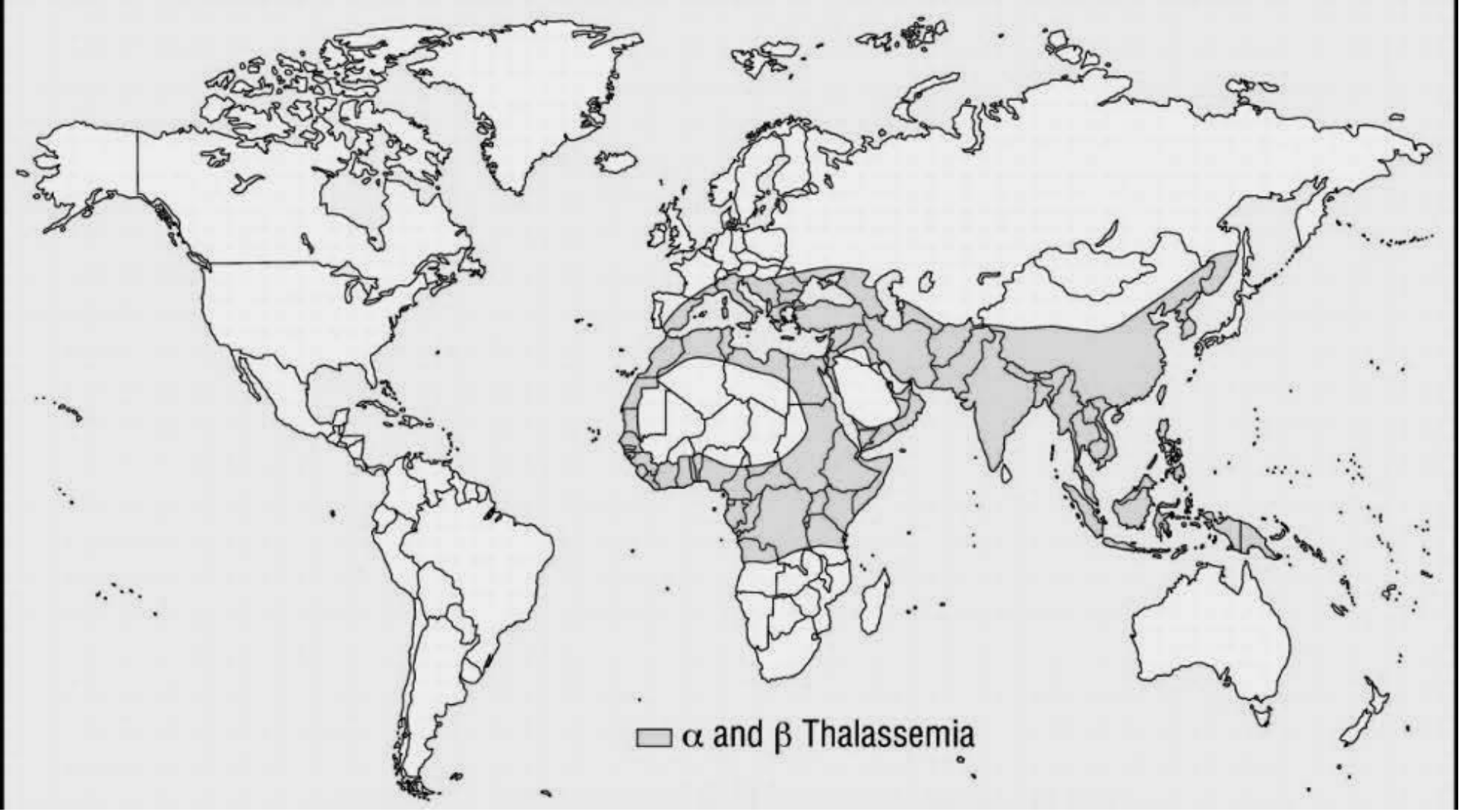
- Variable clinical course
  - SICKLE CELL TRAIT IS MOSTLY ASYMPTOMATIC.

- HYDROXYUREA
  - Increase HbF
  - Anti inflammatory due to decrease WBC production
  - Increase MCV
  - Production of NO
- BONE MARROW TRANSPLANT

# Thalassemia

*The thalassemia syndromes are a heterogeneous group of disorders caused by inherited mutations that* ***decrease the synthesis*** *of either the α-globin or β-globin chains that compose adult hemoglobin, HbA (α2β2), leading to anemia, tissue hypoxia, and* ***red cell hemolysis*** *related to the imbalance in globin chain synthesis*

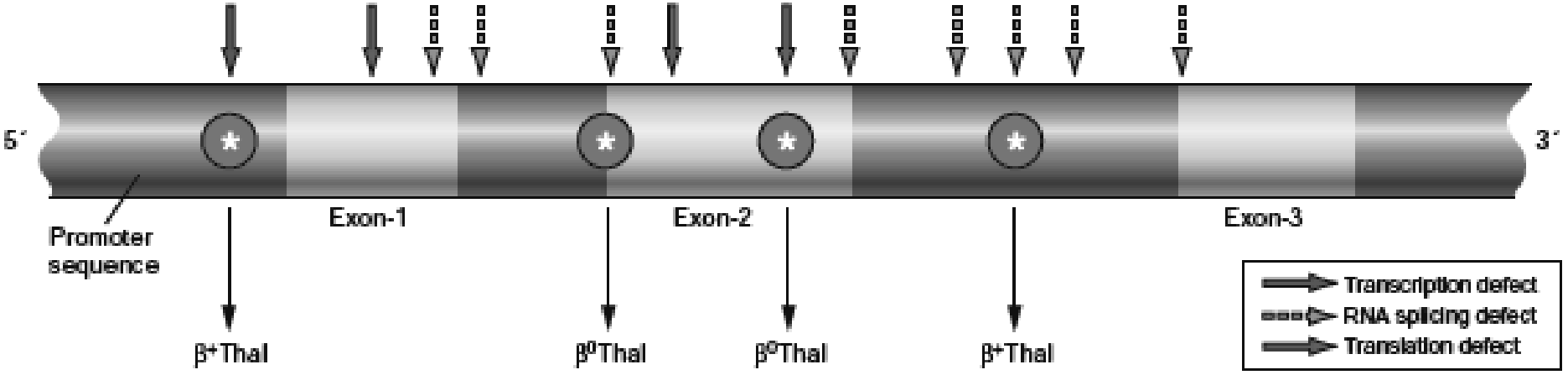
- 4 alpha genes, chromosome 16
- 2 beta genes, chromosome 11


α and β Thalassemia

# B thalassemia

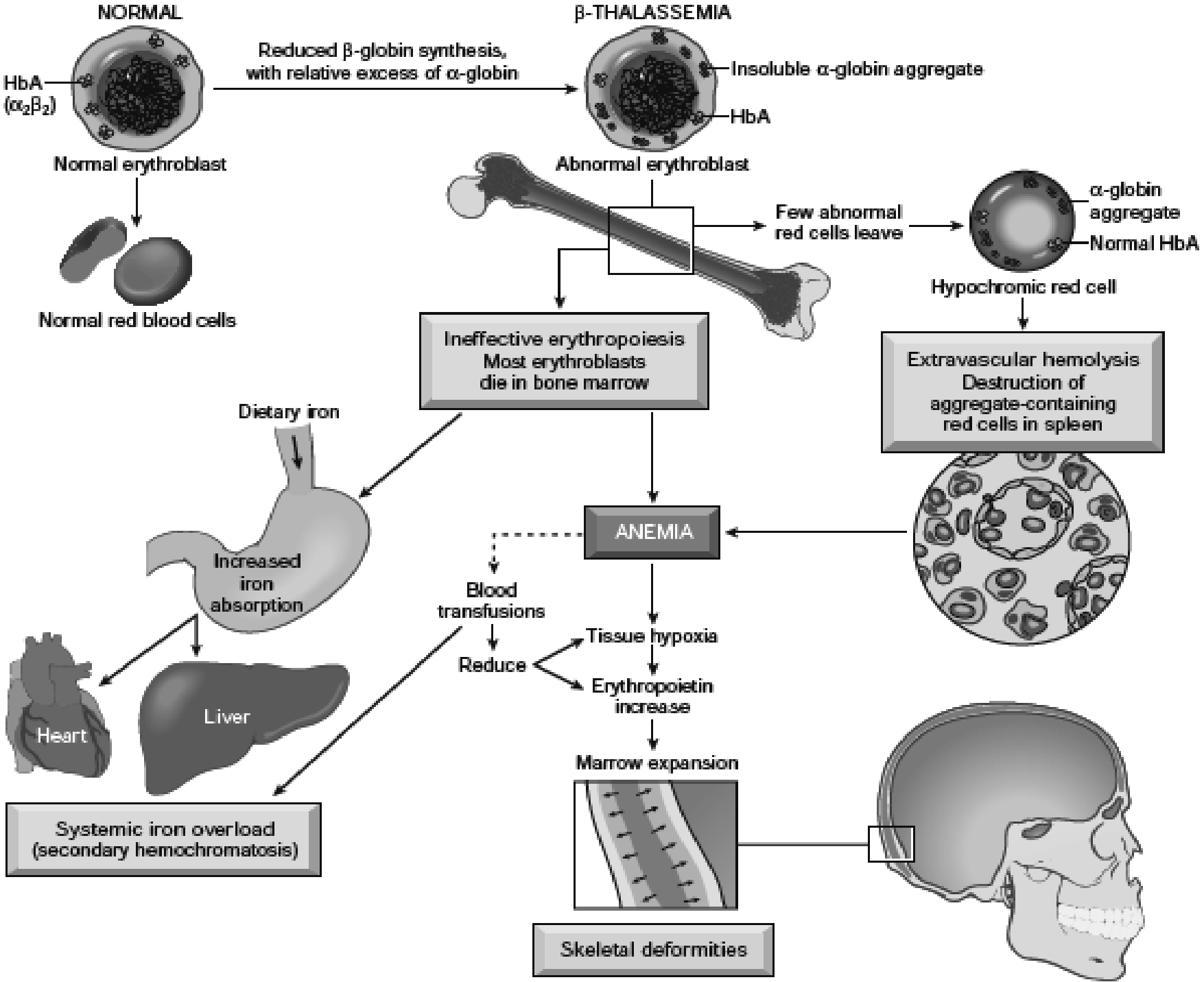
- The β-thalassemias are caused by mutations that diminish the synthesis of β-globin chains.
- Two categories of causative mutations
  - (1) $\beta^0$mutations, associated with absent β-globin synthesis
  - (2) $\beta^+$mutations, characterized by reduced (but detectable) β-globin synthesis.

**unlike sickle cell disease, the amino acid sequence is **INTACT!**




-Promoter region mutation
-Splicing mutations
-Chain termination mutations

- Two mechanisms of anemia
  - Underhemoglobinization
  - Decreased red cell survival due to chain imbalance.


NORMAL
β-THALASSEMIA
HbA
($\alpha_2\beta_2$)
Reduced β-globin synthesis, with relative excess of α-globin
Insoluble α-globin aggregate
HbA
Normal erythroblast
Abnormal erythroblast
Few abnormal red cells leave
α-globin aggregate
Normal HbA
Hypochromic red cell
Normal red blood cells
Ineffective erythropoiesis
Most erythroblasts die in bone marrow
Extravascular hemolysis
Destruction of aggregate-containing red cells in spleen
Dietary iron
ANEMIA
Increased iron absorption
Blood transfusions
Tissue hypoxia
Reduce
Erythropoietin increase
Heart
Liver
Marrow expansion
Systemic iron overload (secondary hemochromatosis)
Skeletal deformities

- *B* thalassemia major (Homozygousβ-thalassemia) (β0/β0, β+/β+, β0/β+)
- *B* thalassemia minor (Heterozygousβ-thalassemia) (β0/β, β+/β)
- *B* thalassemia intermedia (Variable)

  (β0/β+, β+/β+, β0/β, β+/β)

# B-Thalassemia Major.

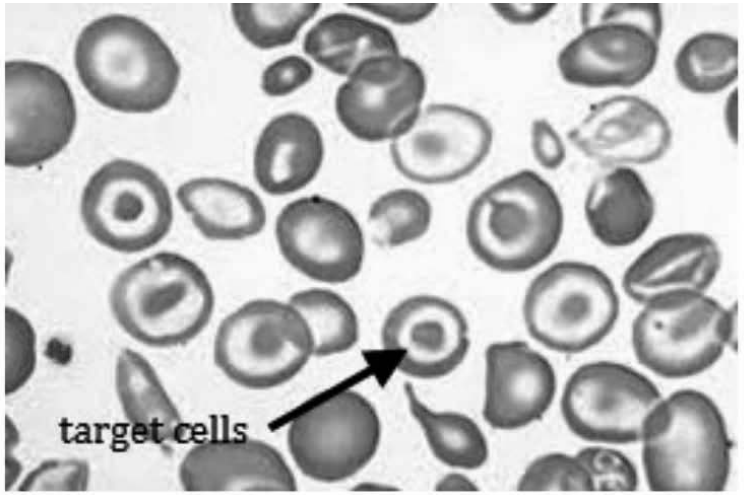
-common in the Mediterranean areas and the Middle East

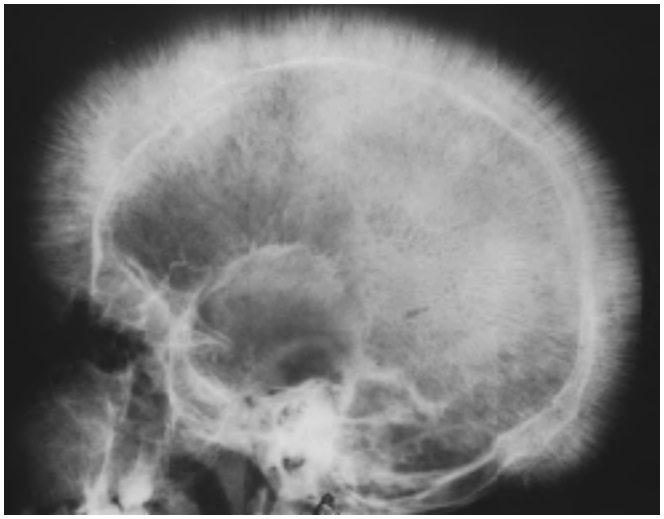
-anemia manifests 6-9months of life after as hemoglobin synthesis switches from HbF (α2γ2) to hemoglobin A (α2β2)

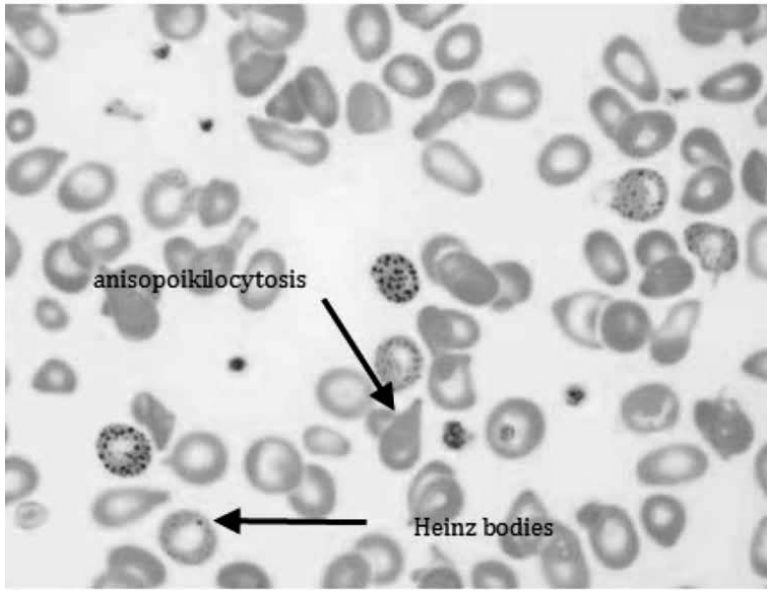
Low hemoglobin 3-6g/dL

Elevated HbF and HbA2(α2δ2)

# Morphology





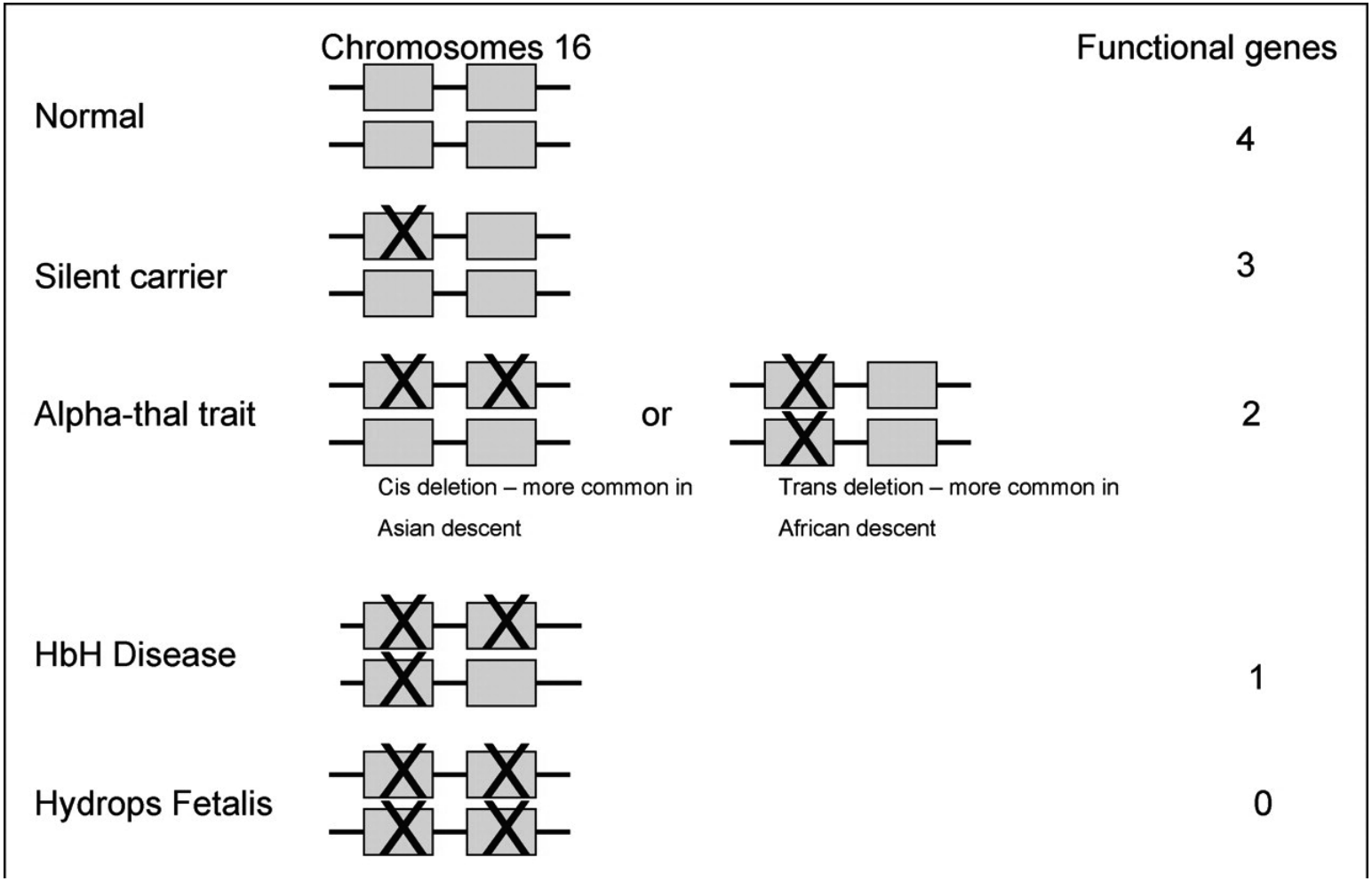
- Hepatosplenomegaly
- Cardiac disease
- Transfusion dependent, role of chelation therapy
- Guarded prognosis
- Stem cell transplantation is the only hope for cure.

# B-thalassemia minor

- Same ethnic groups as B major
- Usually asymptomatic
- Mild PB smear findings
- Bone marrow EP hyperplasia
- Elevated HbA2

- Important to recognize due to
  - Differentiate from IDA
  - Genetic counseling

# Alpha thalassemia

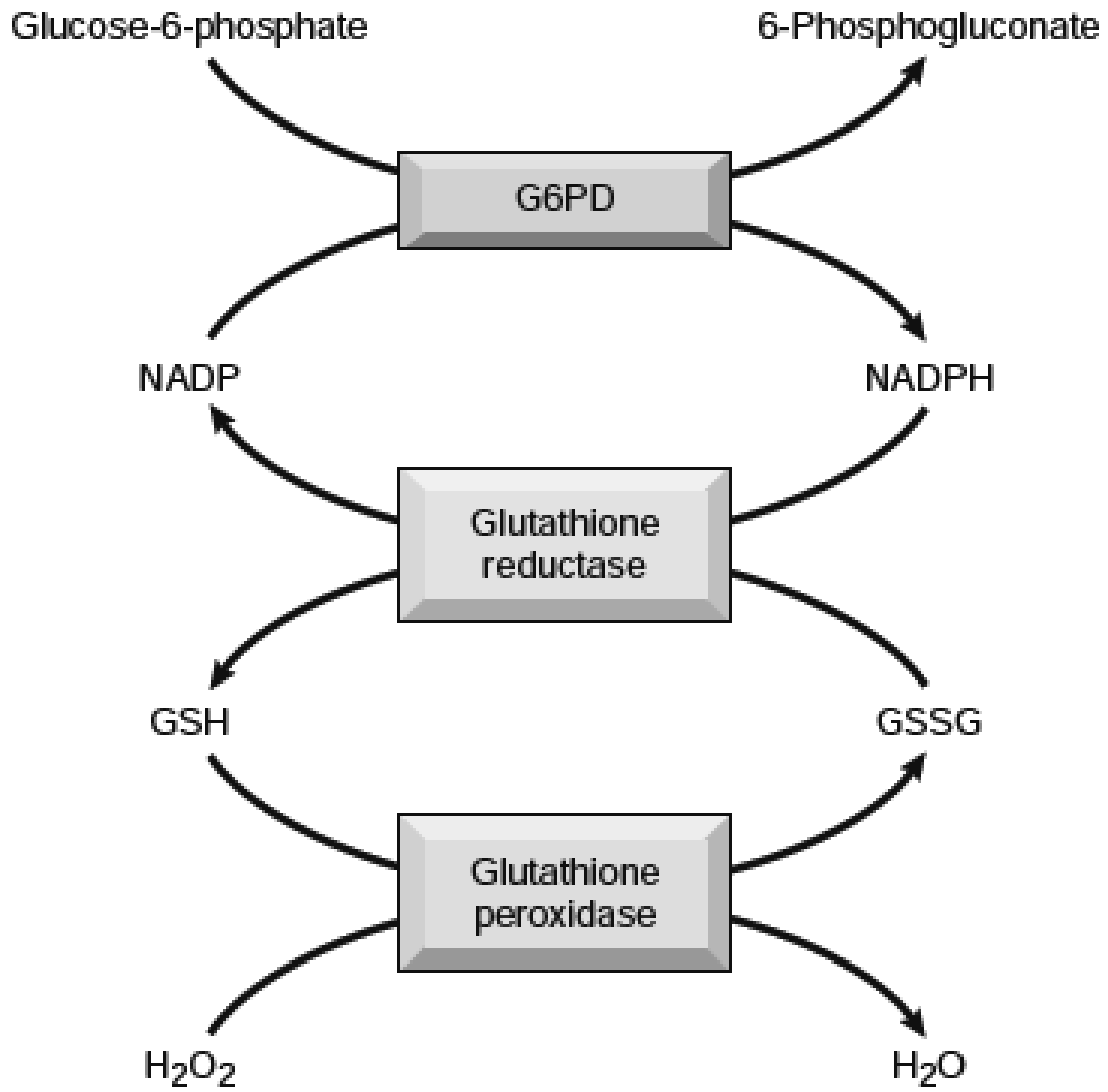


- Silent Carrier State: asymptomatic, microcytosis.
- Alpha thalassemia trait: microcytosis and mild to no anemia
- HbH: moderately severe anemia similar to B-thalassemia intermedia
- Hydrops fetalis: lethal without in utero transfusion.

- The mutations in B thalassemia are **point mutations** or small deletions while in alpha thalassemia they are **large deletions.**

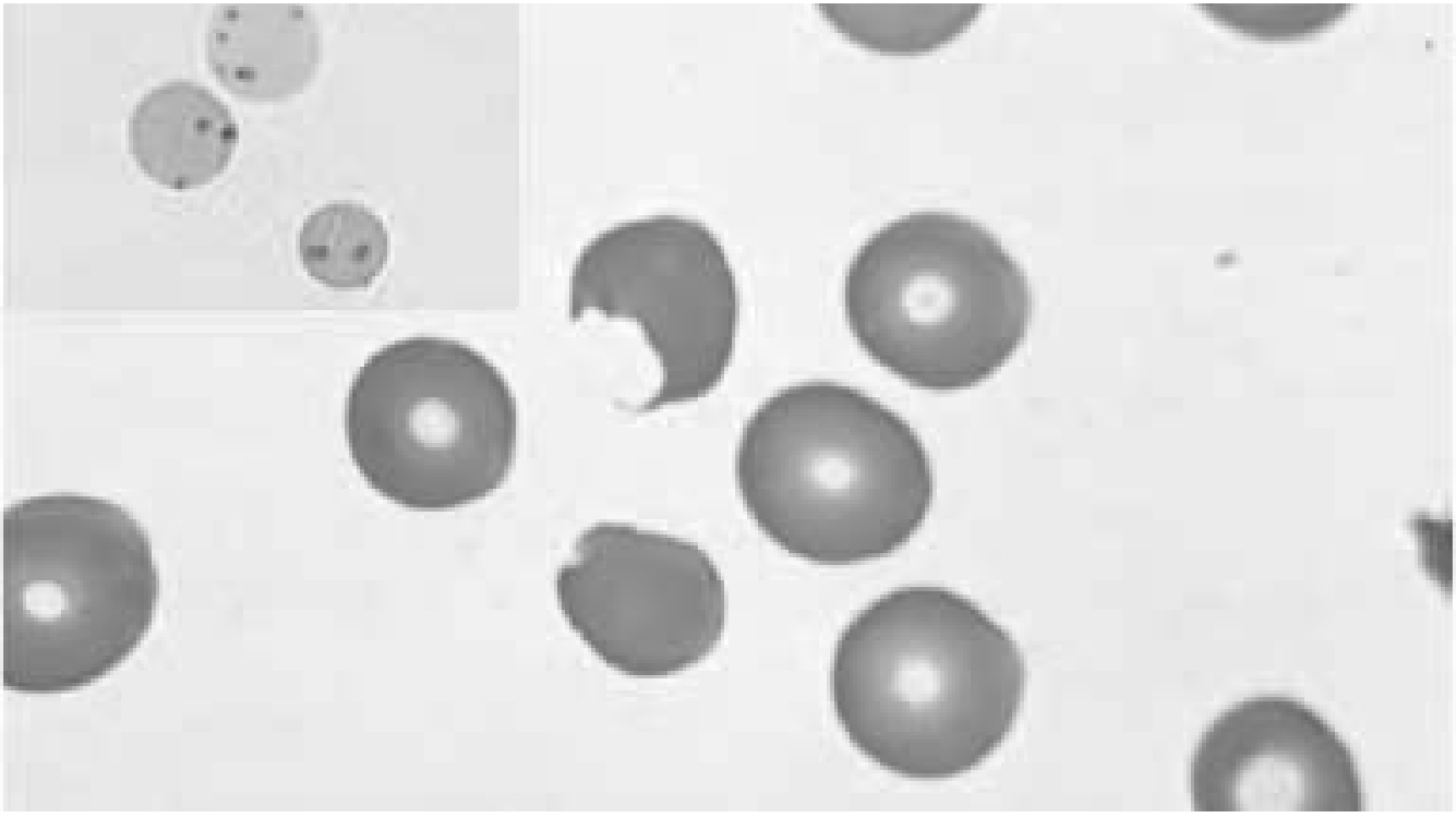
- intrinsic
  - Hereditary
    - Membranopathies-spherocytosis
    - Hemoglobinopathies-thalassemia and sickle cell disease
    - Enzymopathies-G6PD deficiency
  - Acquired
    - Paroxysmal nocturnal hemoglobinuria.

# Glucose 6-phosphate dehydrogenase deficiency



- X-linked disorder
- More common in males
- Numerous mutations
- G6PD A- and G6PD Mediterranean

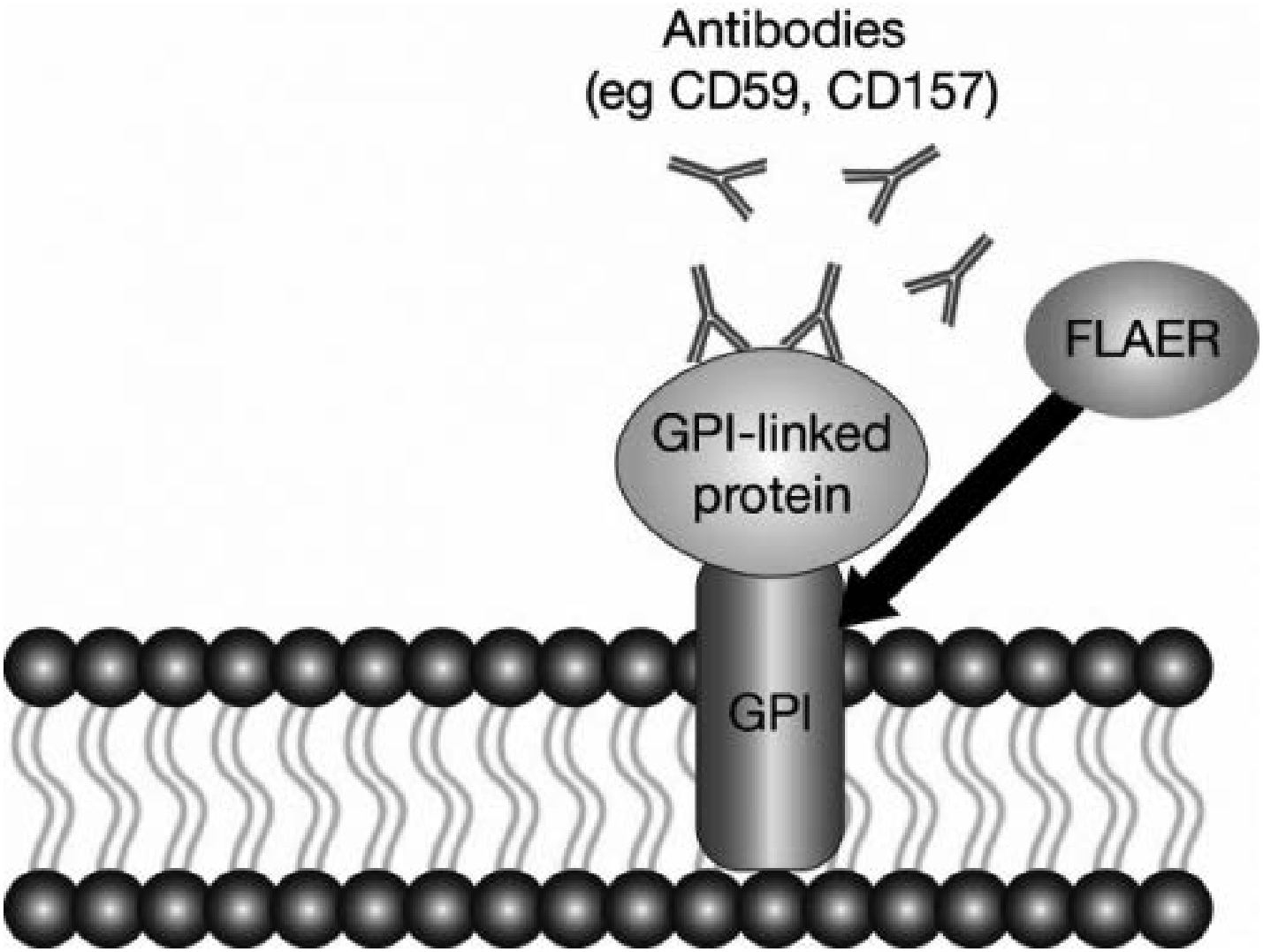
- Presents most commonly as episodic hemolysis
  - Infections: most common cause.
  - Drugs: antimalarials, nitofurantoin
  - Certain foods: fava beans



- Hemolysis can be either intra- or extravascular hemolysis
- Hemolysis stops after old RBC hemolyze even if the offending agent is still effective.
- Since it's **episodic acute (rather than chronic)** hemolysis, features related to chronic hemolysis (splenomegaly and gallbladder stones) are typically **absent.**

- intrinsic
  - Hereditary
    - Membranopathies-spherocytosis
    - Hemoglobinopathies-thalassemia and sickle cell disease
    - Enzymopathies-G6PD deficiency
  - Acquired
    - Paroxysmal nocturnal hemoglobinuria.

# Paroxysmal nocturnal hemoglobinuria (PNH)



- Paroxysmal nocturnal hemoglobinuria (PNH) is a disease that results from **acquired mutations** in the phosphatidylinositol glycan complementation group A gene (PIGA), an enzyme that is essential for the synthesis of certain membrane-associated **complement regulatory proteins**

- It is the only hemolytic anemia resulting from an acquired genetic defect.
- Mutation in the *PIGA* gene, present on the X chromosome.

# Clinical manifestations

- Low level chronic hemolytic anemia
- NOCTURNAL!!!
- Increased risk of thrombosis
- Association with aplastic anemia
- Treatment may place the patient at risk of Niesseria infections.

1- what is the mode of inheritance in the vast majority f spherocytosis cases?

A. Autosomal dominant
B. Autosomal recessive
C. X-linked dominant
D. X linked recessive

2- The amino acid present at the sixth position of the normal alpha-globin chain is replaced by which one of the following amino acids in sickle cell disease?

A. Lysine
B. Valine
C. Serine
D. Alanine
E. None of the above

3- In thalassemia disorders, when only one alpha gene is affected, what do we call that?

A. Normal
B. Silent carrier
C. Thalassemia trait-cis
D. Thalassemia trait-trans
E. HbH disease

4- gallbladder stones are a frequent complication of G6PD deficiency?

TRUE FALSE

5- Paroxysmal nocturnal hemoglobinuria results from an acquired mutation in which of the following genes:

A. Alpha hemoglobin
B. Beta hemoglobin
C. Erythropoietin
D. PIGA
E. G6PD

*Thank you*

# RBC disorders 5

Ahmad Mansour, MD

1- Relative polycythemia occurs in the setting of

A. Wilms tumor
B. Dehydration
C. Renal cell carcinoma
D. Polycythemia vera
E. hypoxia

2- All of the following are examples of secondary absolute polycythemia, except:

A. Smokers
B. Renal artery stenosis
C. Polycystic kidney
D. Polycythemia vera
E. High altitude

3- The risk acute myeloid leukemia in polycythemia vera is

A. 2%
B. 10%
C. 30%
D. 50%
E. 70%

4- One of the following is a major criterion for PV

A. High hemoglobin
B. Hypercellular bone marrow
C. Low erythropoietin level
D. High erythropoietin level
E. Endogenous erythroid colony formation in vitro

# Polycythemia

Polycythemia denotes an abnormally **high red cell count**, usually with a corresponding increase in the **hemoglobin level.**

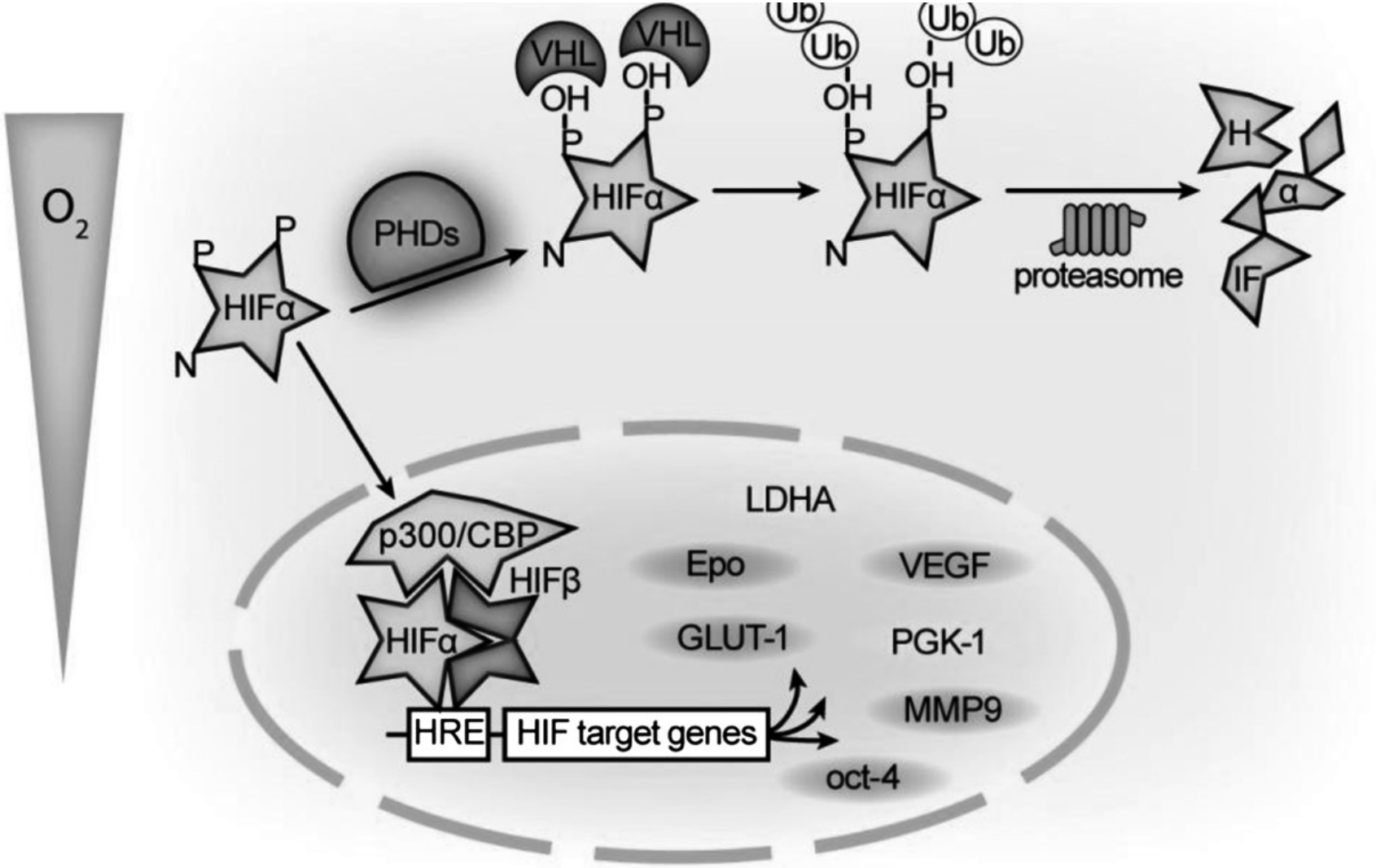
- Relative
- Absolute
  - Primary
  - Secondary

# Relative

- Decrease plasma volume with intaact total RBC mass
  - Dehydration
  - Diarrhea
  - Diuretic therapy

# Absolute

- Increase RBC mass
  - With high EPO (secondary)
  - Low EPO (primary)


O2
P
P
HIFα
N
PHDs
VHL
VHL
OH
OH
P
P
HIFα
N
Ub
Ub
Ub
Ub
OH
OH
P
P
HIFα
N
proteasome
H
α
IF
LDHA
p300/CBP
HIFβ
HIFα
Epo
VEGF
GLUT-1
PGK-1
MMP9
HRE
HIF target genes
oct-4

- Hypoxia
  - Generalized
    - Smoking
    - High altitude
    - High affinity hemoglobins
  - Localized
    - Renal artery stenosis
    - Polycystic kidney disease
- Certain neoplasms
  - Wilms tumor
  - Renal cell carcinoma
  - Celebellar hemangioma
  - Hepatocellular carcinoma

# Polycythemia vera

- Low EPO

Polycythemia vera (PCV) is characterized by increased marrow production of red cells, granulocytes, and platelets **(panmyelosis)**,

but it is the increase in red cells (polycythemia) that is responsible for most of the clinical symptoms

- Strongly associated with **JAK2 mutation**
- Valine-to-phenylalanine substitution at residue 617.
- Patients are prone to both thrombosis and bleeding.
- Splenomegaly mild at first severe in spent phase
- Hypercellular bone marrow.

- Late in the disease course, bone marrow fibrosis and significant organomegaly is present.

# Clinical features

- Pruritis
- Headache dizziness
- Hyperuricemia  and gout
- increased risk of both major bleeding and thrombotic episodes.
  - Deep venous thrombosis
  - Stroke
  - Myocardial infarction
  - Bowel infarction
  - Budd-chiari syndrome

- Epistaxis and bleeding gums
- Major hemorrhage can occur in ~10% of the patients

- Phlebotomy and JAK2 inhibitors
- Spent phase; fibrosis and splenomegaly
- 2% might transform to acute myeloid leukemia

# Criteria for diagnosis

(2major and one minor or the first major with two minor)

**Major criteria**

1. Haemoglobin >18.5 g/dL in men, 16.5 g/dL in women or other evidence of increased red cell volume*

2. Presence of JAK2 V617F or other functionally similar mutation such as JAK2 exon 12 mutation

**Minor criteria**

1. Bone marrow biopsy showing hypercellularity for age with trilineage growth (panmyelosis) with prominent

erythroid, granulocytic and megakaryocytic proliferation

2. Serum erythropoietin level below the reference range for normal

3. Endogenous erythroid colony formation in vitro

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THANK YOU