

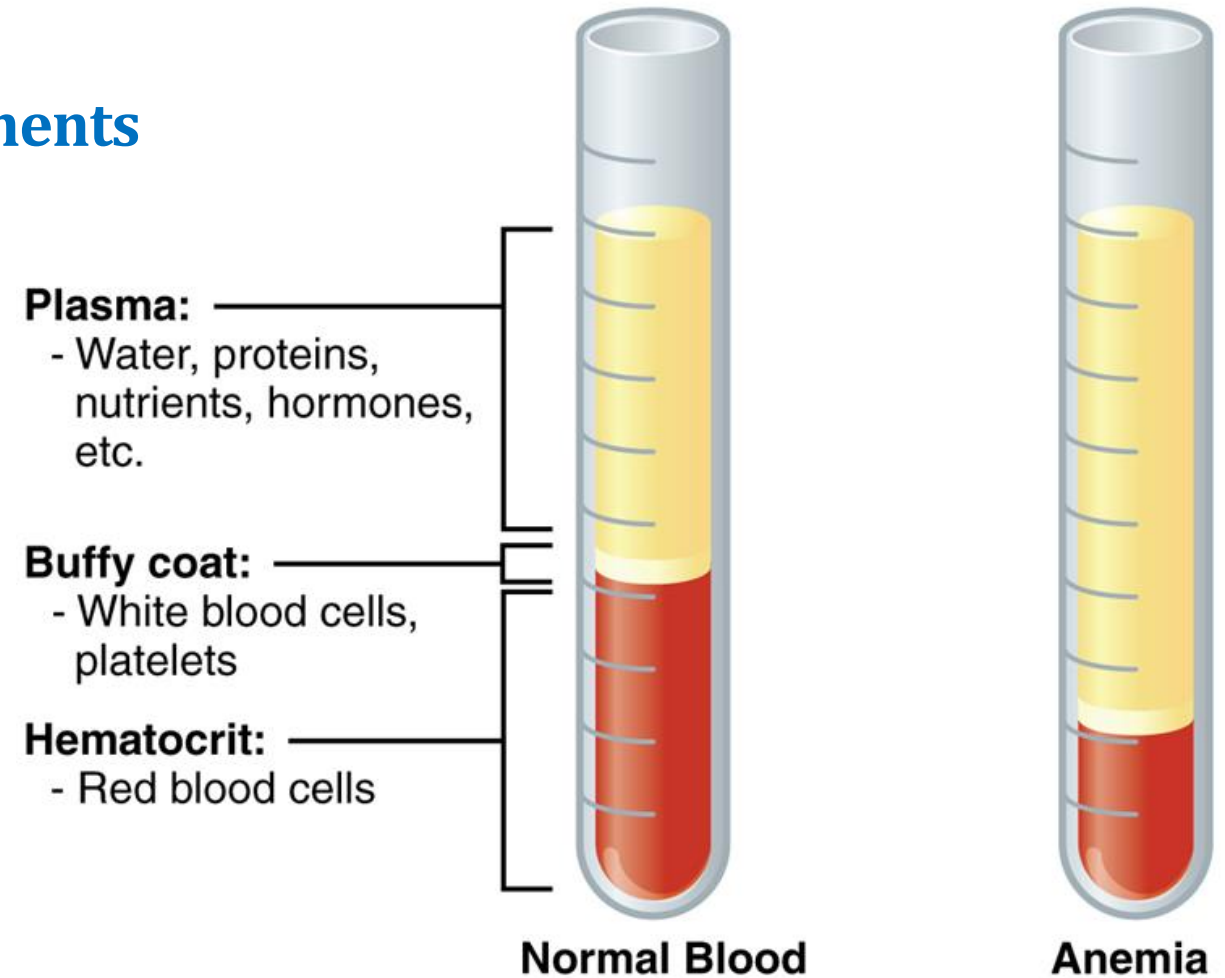
Normocytic Anemias

Jason Ryan, MD, MPH



Anemia

- Decrease in **red blood cell measurements**
- RBC count: part of CBC
- Hemoglobin: concentration in g/dL
- Hematocrit: volume % of red cells



Anemia

RBC Measurements

- **“Rule of 3”**
- $\text{Hgb} = 3 \times \text{RBC count}$
- $\text{Hct} = 3 \times \text{Hgb}$

Normal Ranges

Measurement	Men	Women
Hemoglobin (g/dL)	> 13.5 (13.6 to 16.9)	>12 (11.9 to 14.8)
Hematocrit (%)	~45 (40 to 50)	~40 35 to 43
RBC count ($\times 10^6$ /microL)	~5 (4.2 to 5.7)	~4.5 (3.8 to 5.0)

Anemia

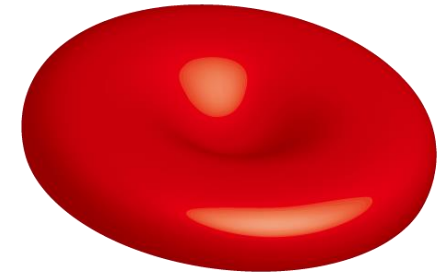
Clinical Features

- Vary based on degree of anemia and onset (rapid/slow)
- Can be asymptomatic
- Weakness
- Fatigue
- Dyspnea
- Pallor - unhealthy pale appearance
 - Skin of palms and face
 - Also nails and conjunctivae

Pallor versus Normal Palm



Red Blood Cell Indices



- Measured by automated blood counters
- Describe **mean** characteristics of RBCs
- Used in evaluation of anemias

Index	Units	Normal
Mean corpuscular volume (MCV)	Femtoliters (fL)	80 to 100
Mean corpuscular hemoglobin (MCH)	Picograms/cell (pg/cell)	27.5 to 33.2
Mean corpuscular Hgb concentration (MCHC)	Grams per deciliter (g/dL)	34 grams

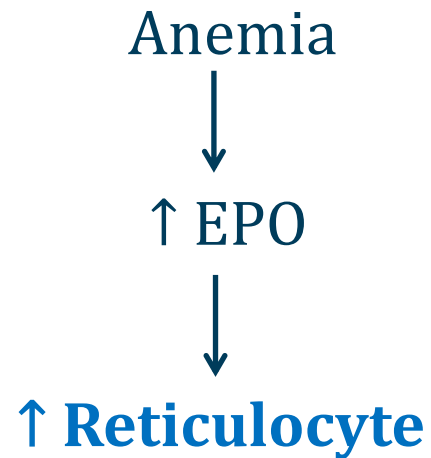
Anemia Classification

- Anemia = decreased red cell measurements (RBC, Hgb, Hct)
- Further classified by MCV
- Normal range 80 to 100 fL

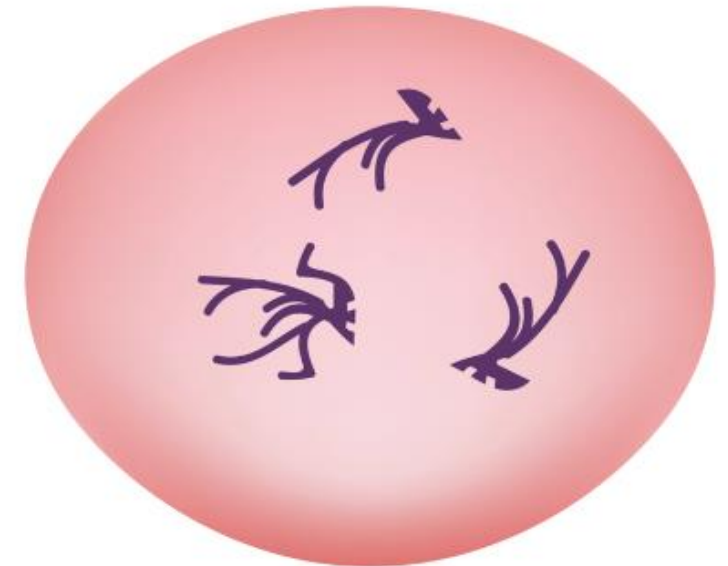
Microcytic MCV<80	Normocytic MCV 80-100	Macrocytic MCV>100
Iron deficiency Thalassemia Anemia chronic disease Sideroblastic Anemia Lead poisoning	Anemia chronic disease Hemolysis Aplastic anemia Chronic kidney disease	Folate/B12 deficiency Liver disease Alcohol use Reticulocytosis

Reticulocytes

- Immature red blood cells
- Usually about 1 to 2% of RBCs in peripheral blood
- Should increase with anemia if RBC production is normal



Reticulocyte



Reticulocyte Count

- Used in anemia to determine if bone marrow response is adequate
- Reticulocyte count should rise if bone marrow responding normally
- Must be **corrected** for degree of anemia
- If $< 2\%$ → **inadequate bone marrow response**

Hct 45 (normal)	→	Hct 11
Retic 1% (normal)		Retic 8%

$$\text{Corrected RC} = 8\% * (11/45) = 2\%$$

Reticulocyte Production Index

- Normal reticulocytes circulate ~1 day
- In anemia → premature release of reticulocytes
- **Can circulate longer**
- RPI corrects for longer life of reticulocytes in anemia
- RPI > 3% is a normal marrow response to anemia
- RPI < 2% seen with bone marrow failure

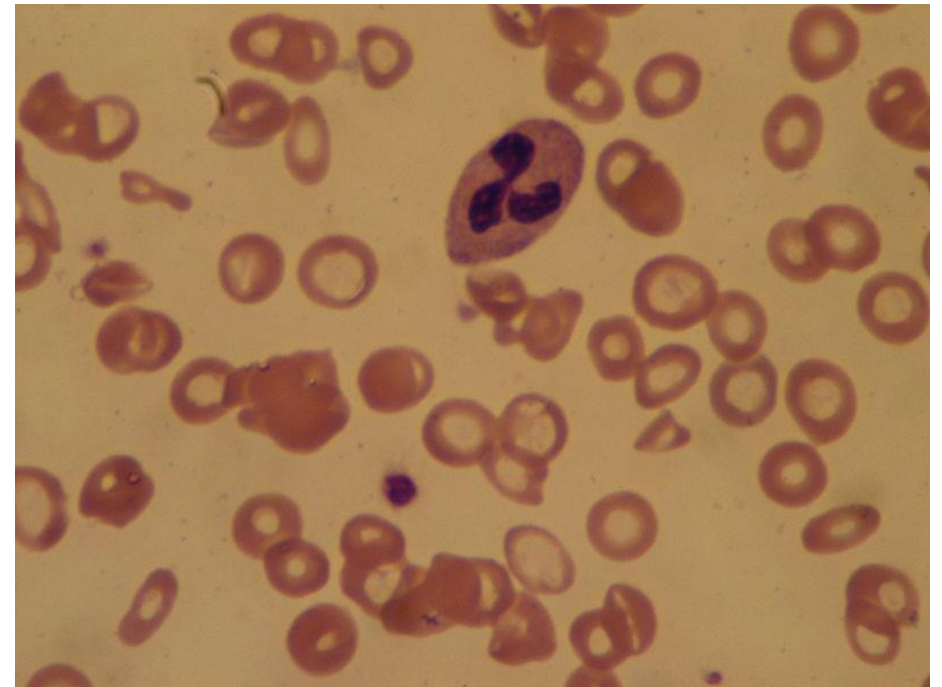
$$\text{RPI} = \frac{\text{Corrected Retic \%}}{\text{Maturation Time}}$$

Hgb	MT
15	1
12	1.5
8	2
5	2.5

Blood Smear

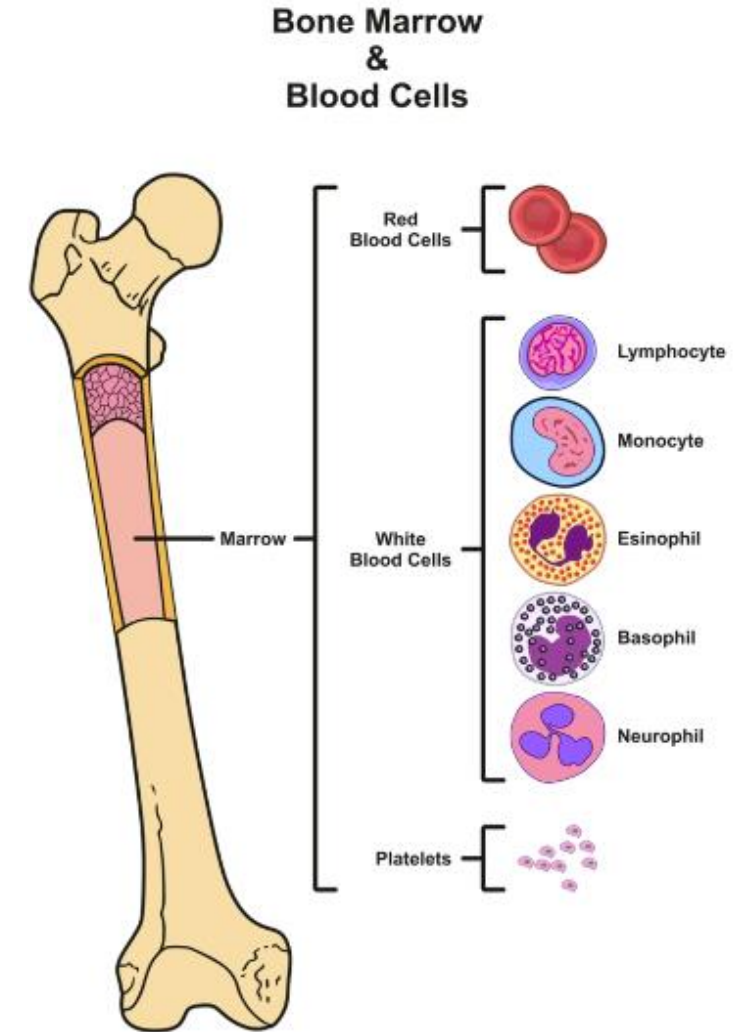
- Microscopic examination of peripheral blood
- Classic findings in some disorders
- Hypochromia: iron deficiency
- Spherocytes: spherocytosis
- Target cells: thalassemia
- Dacrocytes (tear drop cells): marrow fibrosis
- Acanthocytes: liver disease (spiked RBCs)
- Schistocytes: hemolysis

Hypochromia



Normocytic Anemias

- **Decreased red cell production**
 - Low reticulocyte count
 - Anemia or chronic disease
 - Chronic kidney disease (\downarrow EPO)
 - Hypothyroidism
 - Bone marrow failure
 - Aplastic anemia
- **Increased red cell destruction/loss**
 - High reticulocyte count
 - Acute blood loss
 - Hemolysis

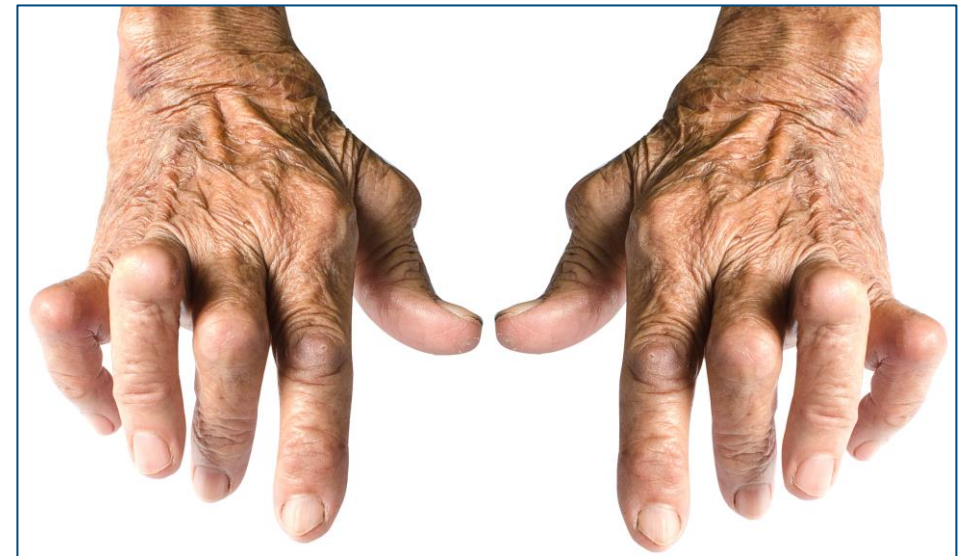


Normocytic Anemia

Anemia of chronic disease

- **Most common normocytic anemia**
- Anemia in association with **inflammation**
- Common in rheumatoid arthritis, lymphoma, other chronic conditions
- Usually a **mild anemia (Hgb > 10g/dL)**
- Symptoms from anemia rare
- Often incidental finding on blood testing
- Microcytic in about 25% cases

Rheumatoid Arthritis

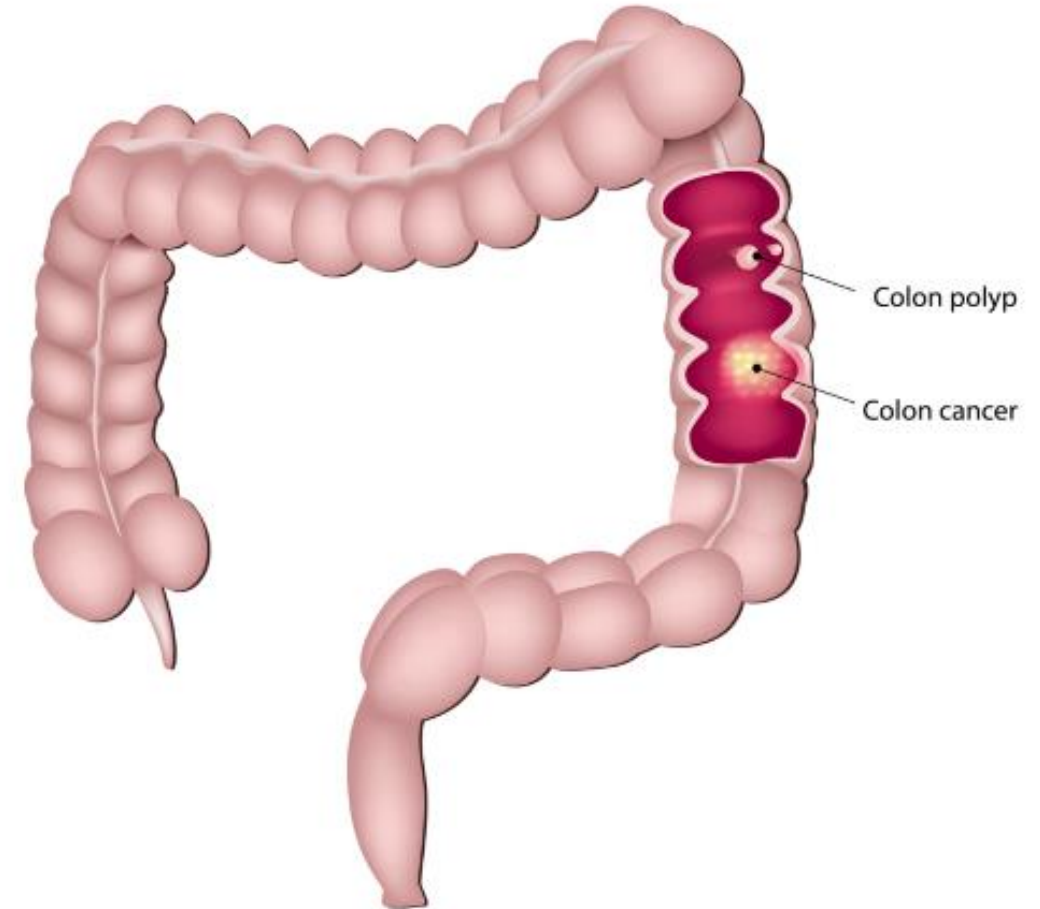


Normocytic Anemias

Blood loss

- **Acute blood loss** → normocytic anemia
 - Loss of red cells → fewer remaining red cells
 - Remaining red cells have normal MCV
- **Chronic blood loss** → microcytic anemia
 - Loss of iron in hemoglobin
 - Leads to iron deficiency
 - Common with occult GI bleeding

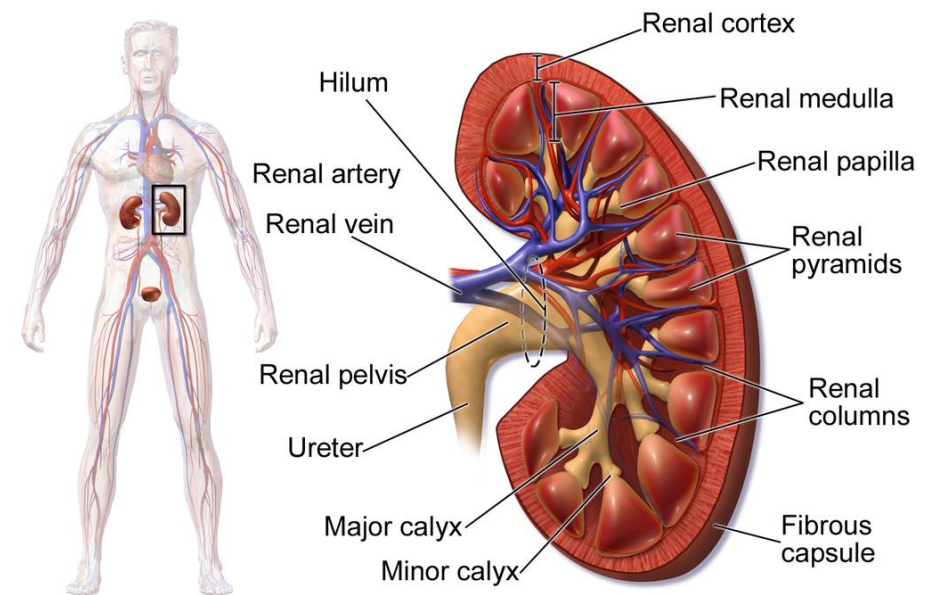
COLON CANCER AND POLYP



Normocytic Anemia

Chronic Kidney Disease

- Decreased production of erythropoietin
- **Erythropoiesis-stimulating agents**
 - Recombinant human EPO
 - Epoetin or darbepoetin (longer-acting)
 - Used when Hgb < 10 g/dL
 - Goal Hgb: 10 to 11.5 g/dL
 - Higher Hgb: ↑ mortality and myocardial infarction
 - May cause or worsen hypertension
 - Good response requires iron
 - Iron deficiency → poor response

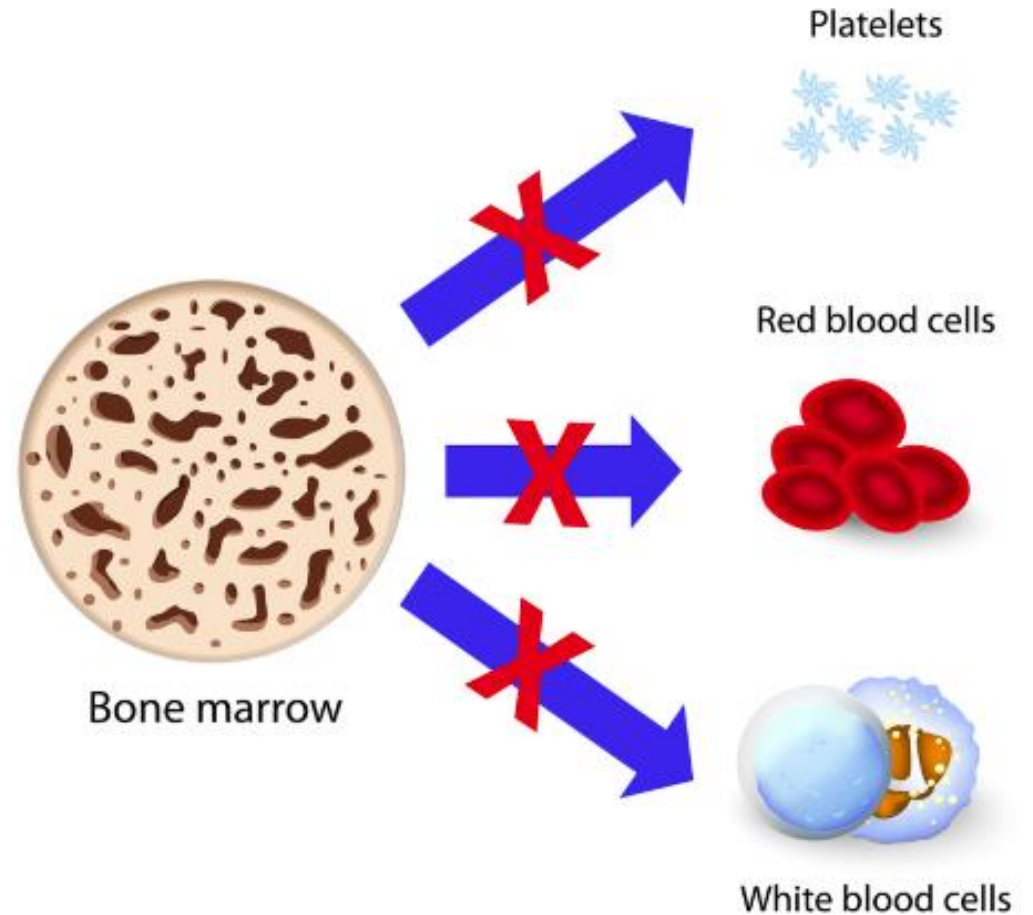


Kidney Anatomy

Aplastic Anemia

- Pancytopenia
- ↓ WBC, ↓ Platelets, ↓ RBC
- Low reticulocyte count
- Hypocellular bone marrow
- Absence of infiltrate or fibrosis

APLASTIC ANEMIA



Terminology

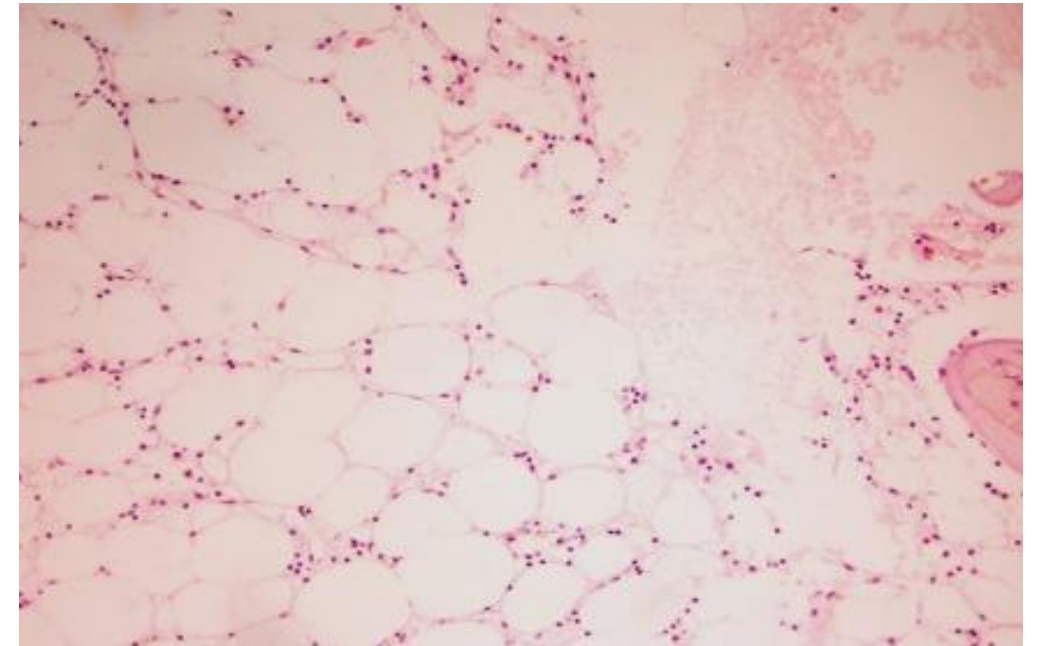
- Bone marrow failure
 - Bone marrow cannot produce cells
 - Results in pancytopenia
 - Many causes: fibrosis, tumors
 - Low reticulocyte count
- Aplastic anemia:
 - Specific type of bone marrow failure
 - Defective stem cells → acellular/hypocellular bone marrow



Aplastic Anemia

Diagnosis

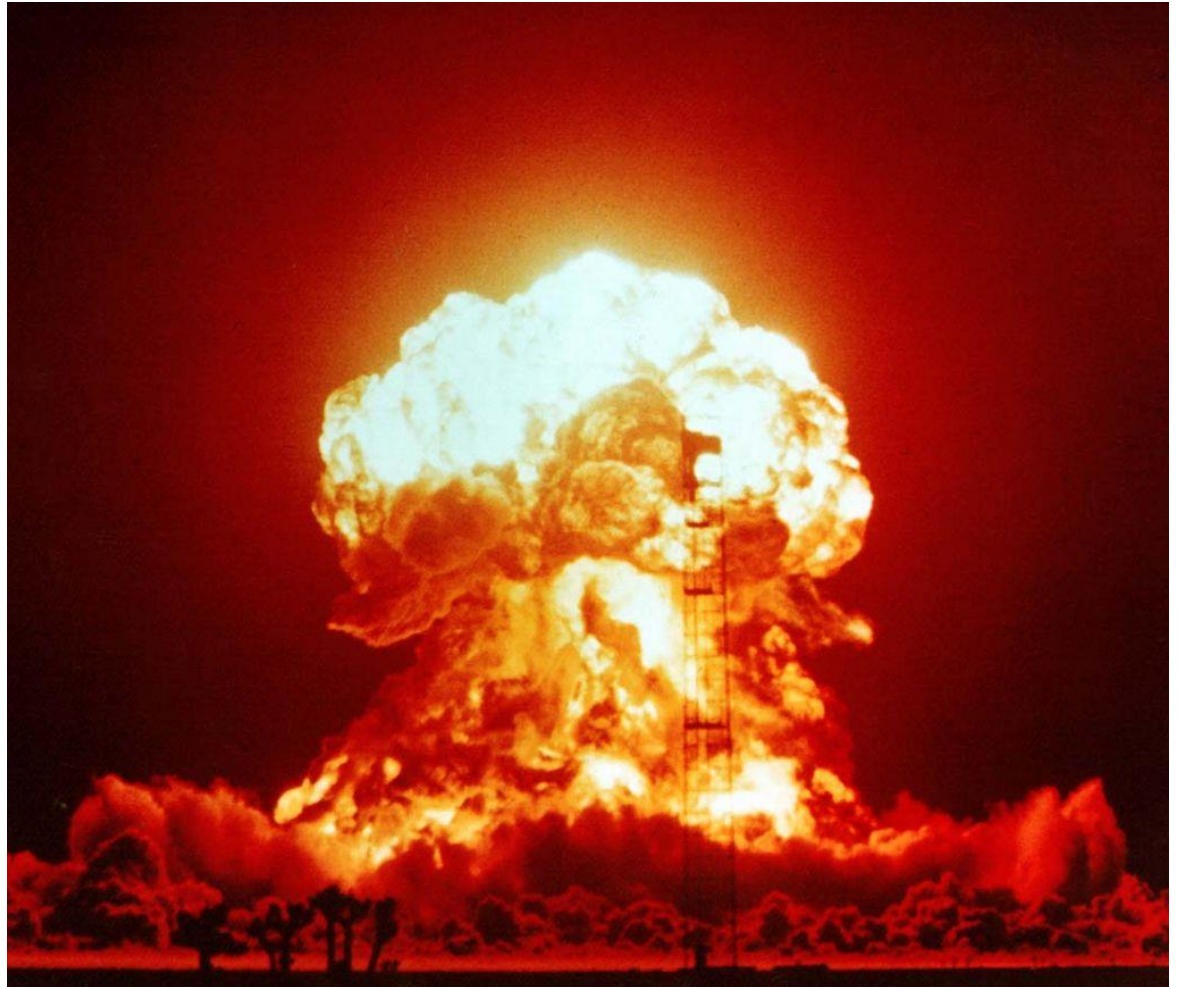
- Pancytopenia
- **Bone marrow biopsy**
- Acellular or hypocellular bone marrow
- Absence of cells/replacement with fat



Aplastic Anemia

Causes

- Most commonly **idiopathic**
- Radiation exposure
- Drugs
- Toxins (benzene)
- Viruses
- Inherited (Fanconi anemia)



Aplastic Anemia

Mechanism

- Most cases involve **autoimmune damage** to stem cells
- Even when an underlying cause is present
- Drugs, chemicals, or viruses alter stem cells
- Leads to autoimmune destruction
- Key clinical point: **immunosuppression improves AA**

Aplastic Anemia

Drugs

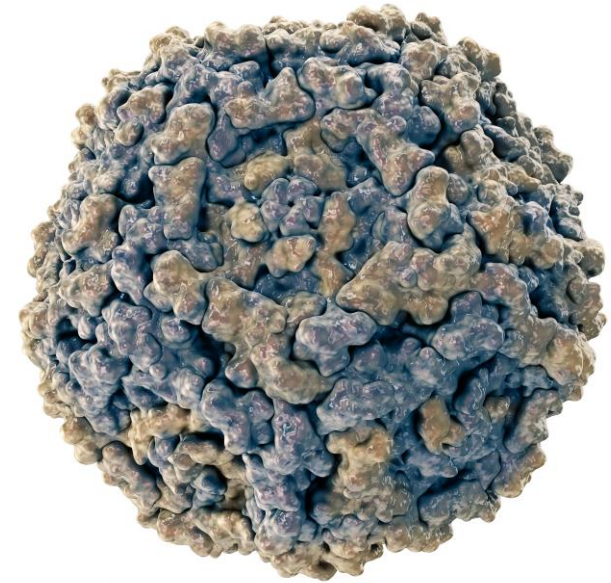
- Chemotherapy
 - Anticipated effect
- Antibiotics
 - Chloramphenicol and sulfonamides
- NSAIDs
 - Phenylbutazone and indomethacin
- Anti-seizure drugs
 - Carbamazepine and phenytoin
- Anti-thyroid drugs
 - Methimazole and propylthiouracil

Aplastic Anemia

Viruses

- **Parvovirus B19**
 - Infects proerythroblasts
 - Usually causes ↓ RBCs
 - Pancytopenia can occur
 - ↑ risk: immunocompromised
- **Acute viral hepatitis**
 - Develops weeks to months after acute hepatitis
- Others: HIV, EBV, CMV

Parvovirus



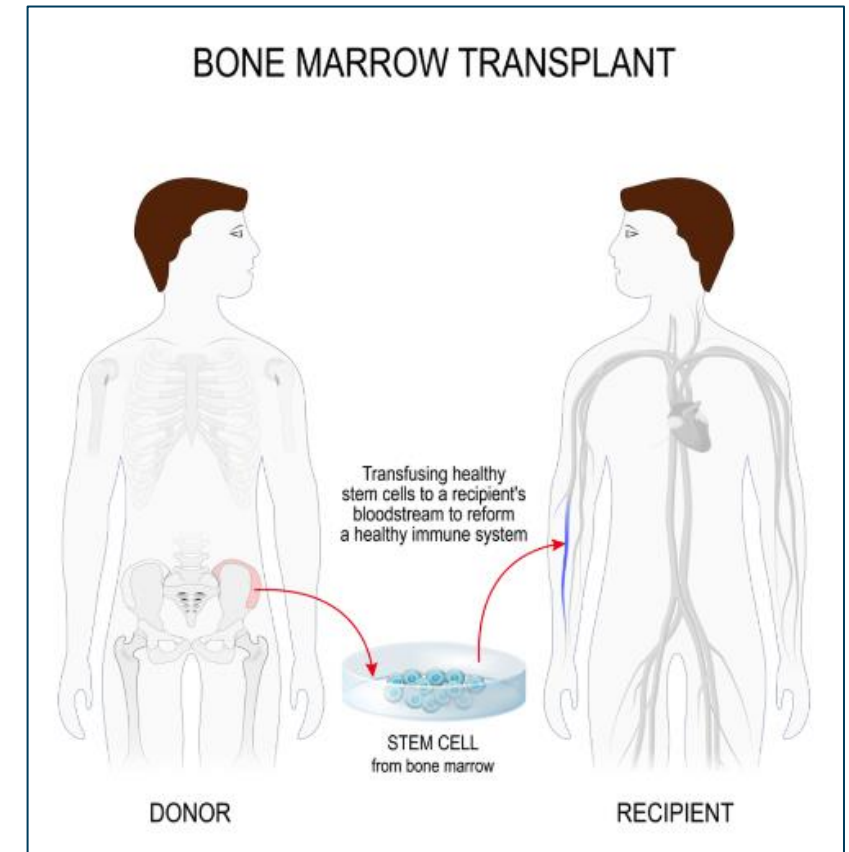
Fanconi Anemia

- Inherited aplastic anemia
- Autosomal recessive or X-linked
- Usually presents in children < 16 years old
- More than half of patients have **physical deformities**
 - Short stature, malformed thumbs
 - Skin: hypo or hyperpigmentation, café-au-lait spots
- More than 13 genetic abnormalities identified
- Many involve **DNA repair enzymes**

Aplastic Anemia

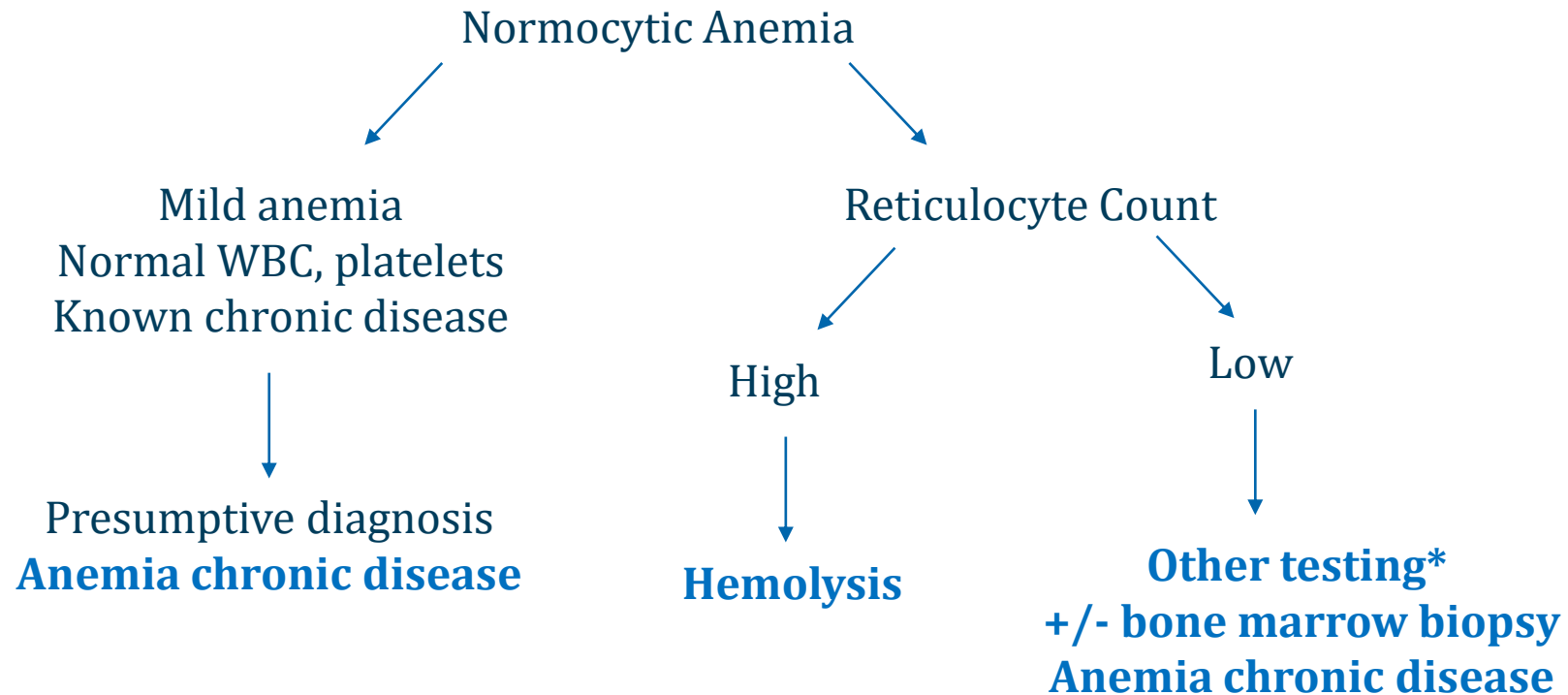
Treatment

- Stop offending agent
- Blood and platelet transfusions
- Bone marrow stimulation
 - GM-CSF: granulocyte/macrophage-colony stimulating factor
- Immunosuppression
 - Antithymocyte globulin (T-cell antibodies)
 - Cyclosporine
- Bone marrow transplant



Normocytic Anemia

Workup



Microcytic Anemias

Jason Ryan, MD, MPH



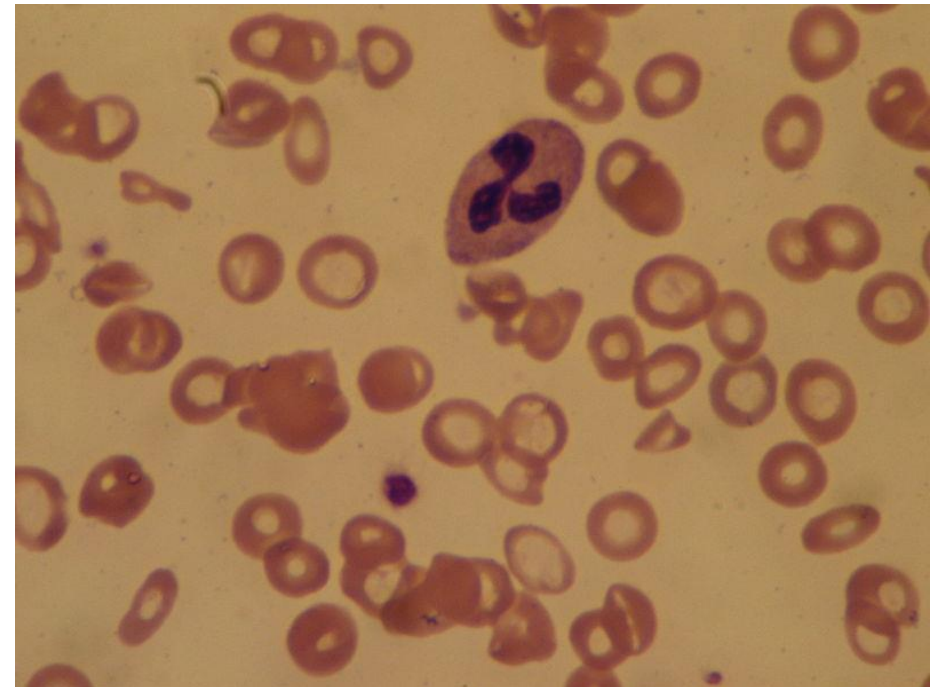
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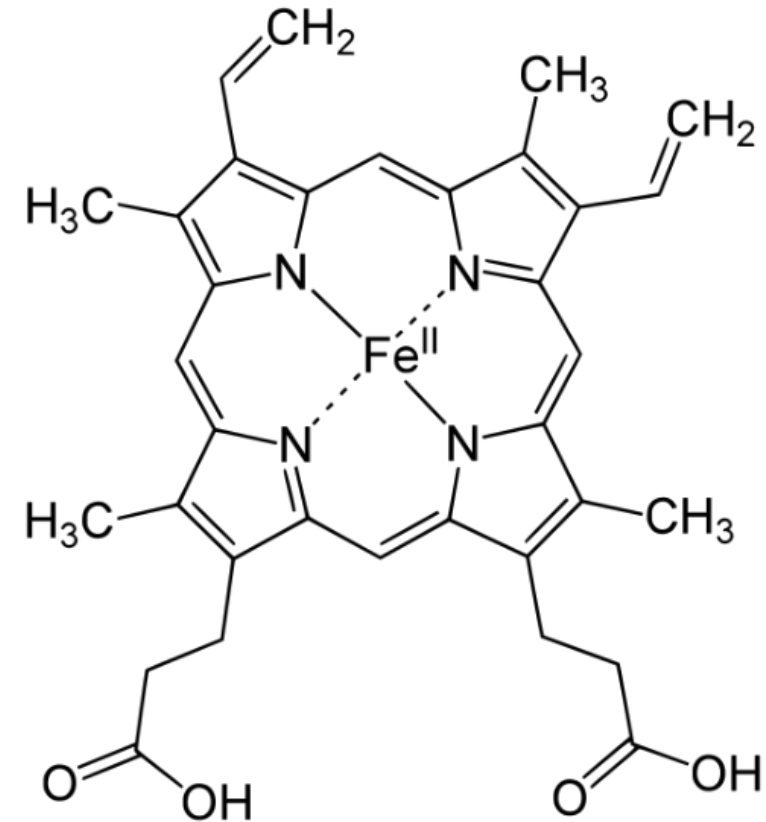
Microcytic Anemias

- Usually due to ↓ **hemoglobin** in red cells
- Usually associated with ↓ MCH and MCHC
- Low hemoglobin → hypochromic RBCs on smear
- Low hemoglobin → ↓ reticulocytes



Hemoglobin

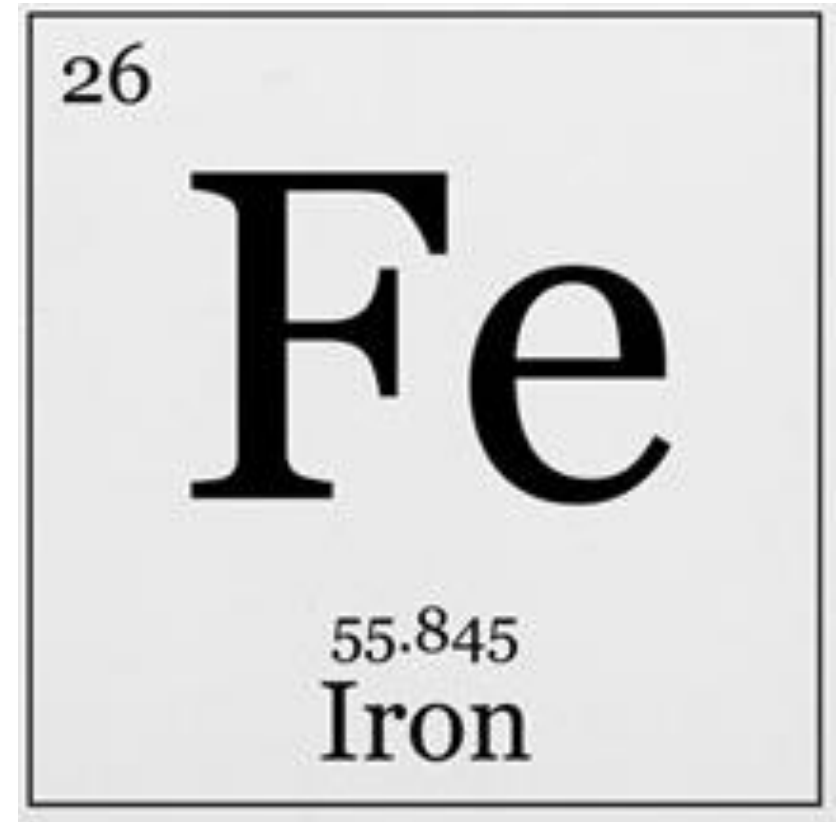
- **Iron**
- **Heme molecules**
- **Globin chains**
- Microcytic anemia from lack of hemoglobin
 - Loss of iron
 - Loss of heme (lead, sideroblastic)
 - Loss of globin chains (thalassemia)



Heme

Iron Metabolism

- Iron always bound to a protein
- Transport protein: **transferrin**
 - Transported in blood via transferrin
 - ↑ transferrin when iron stores are low
- Storage protein: **ferritin**
 - Stored intracellularly as ferritin
 - Stored in **macrophages** of **liver and bone**

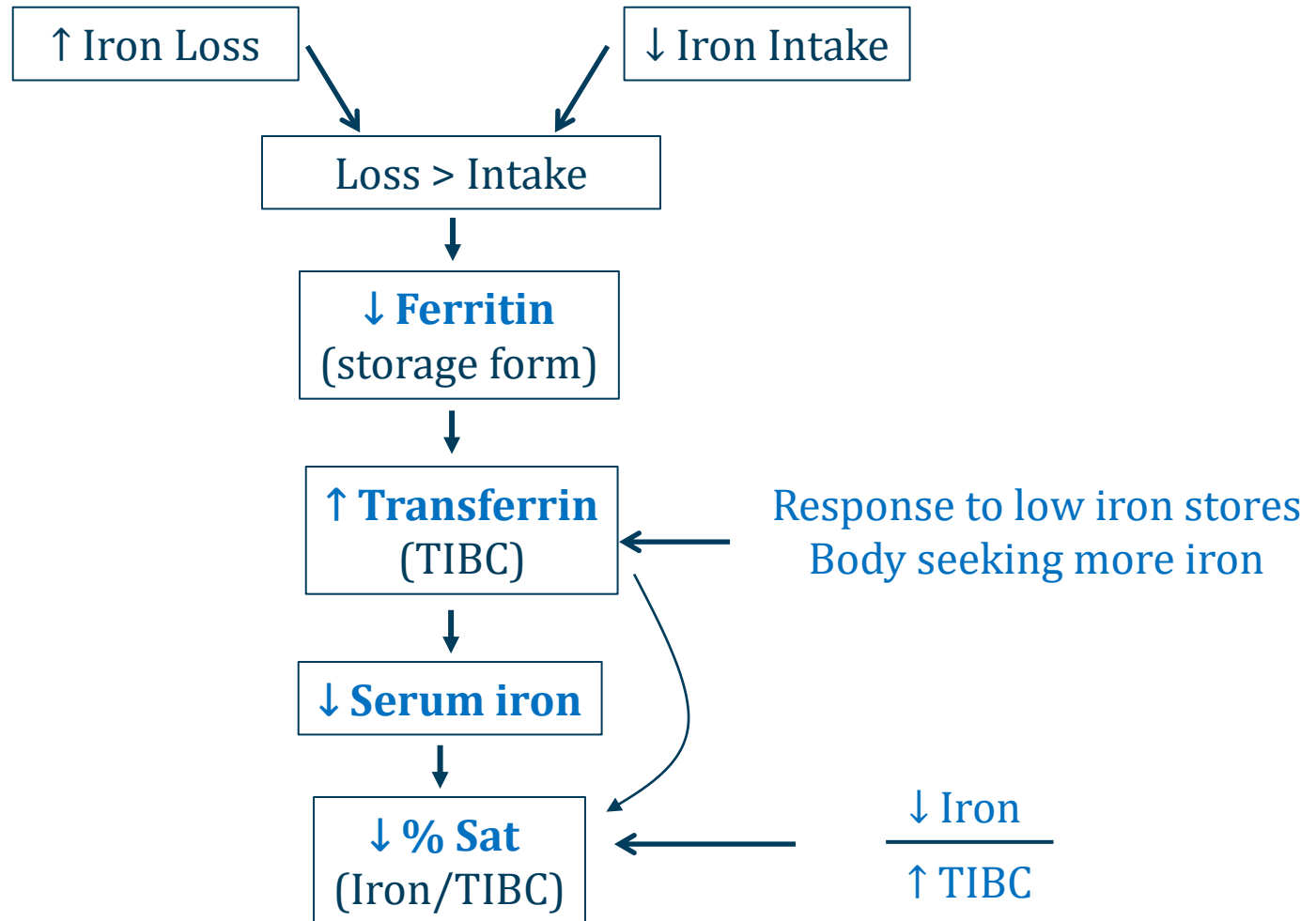


Iron Measurements

Test	Interpretation
Serum iron	Iron level
Total Iron Binding Capacity	Amount of transferrin in serum
Serum ferritin	Amount of storage iron
% saturation	Amount of transferrin bound to Fe

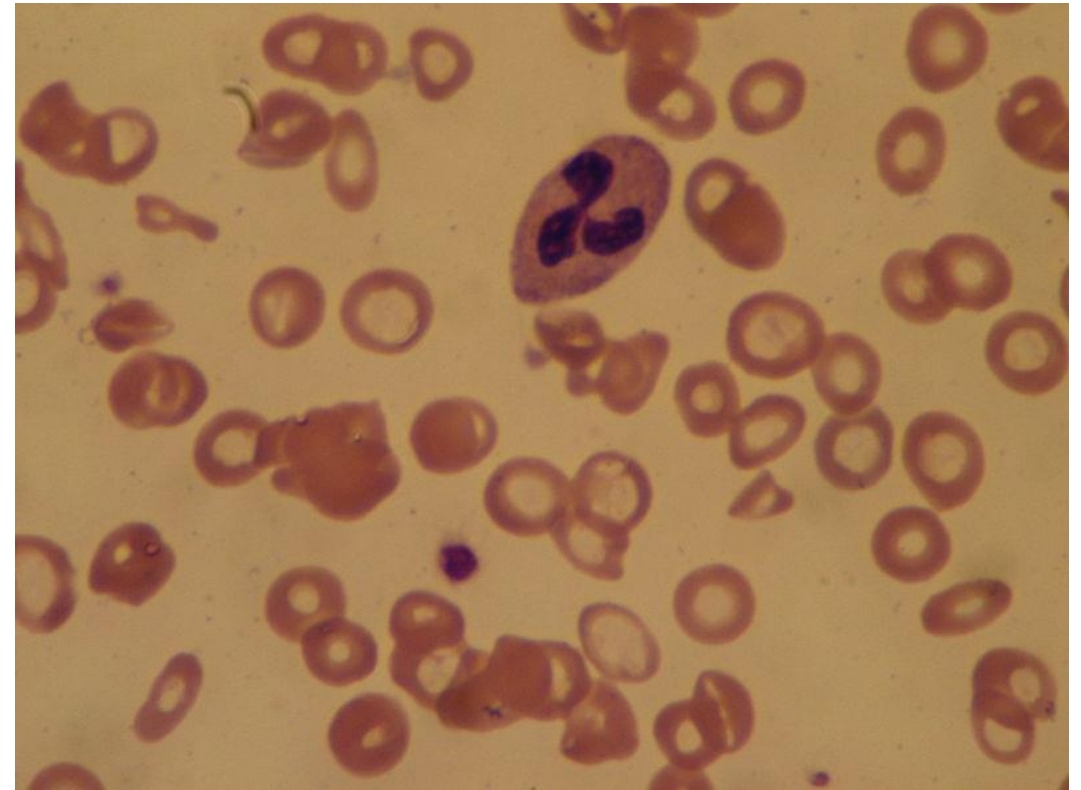
Iron Deficiency

Test	Fe Deficiency
Serum iron	↓
TIBC	↑
Serum ferritin	↓
% saturation	↓↓



Iron Deficiency Anemia

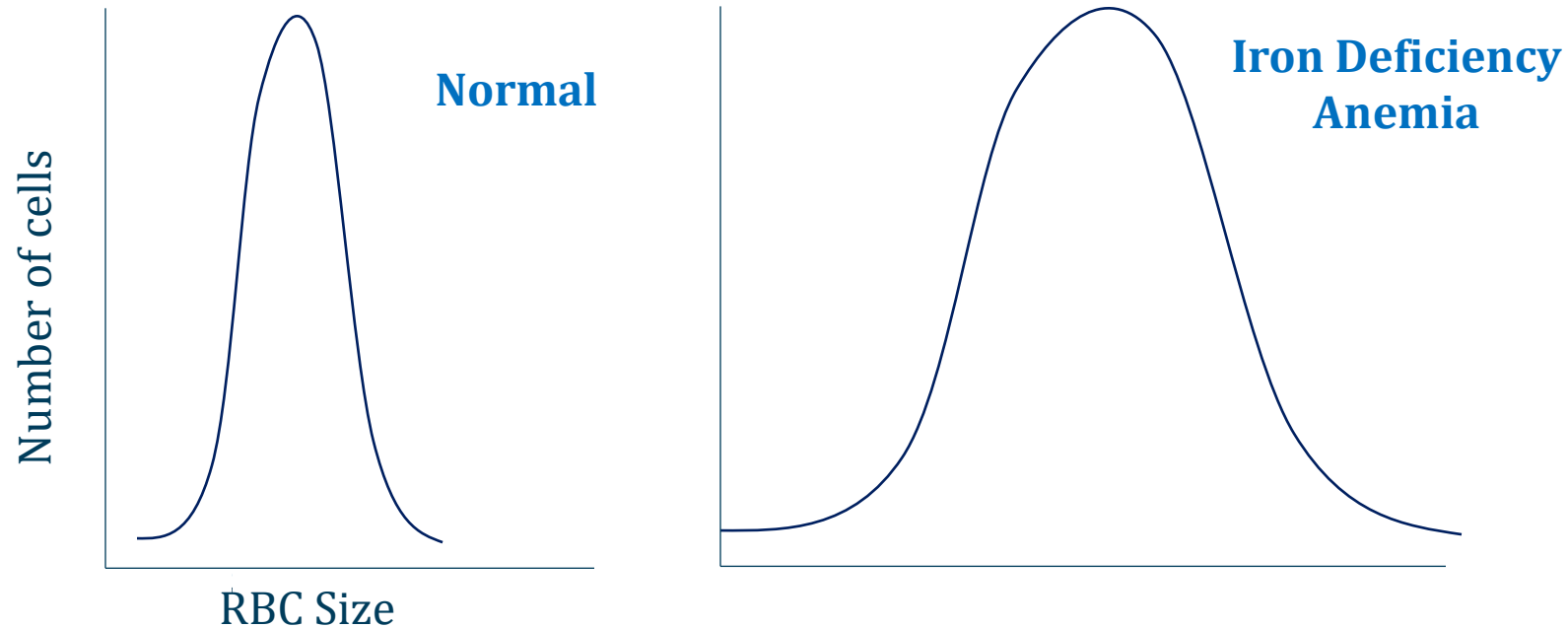
- **Microcytic, hypochromic anemia**
 - ↓ RBC/Hgb/Hct
 - Low MCV (small cells)
 - Hypochromic (low hemoglobin)
 - ↓ MCV/MCH/MCHC
- Initially may be normocytic
- Low reticulocyte count (↓ Hgb)



Red Cell Distribution Width

RDW

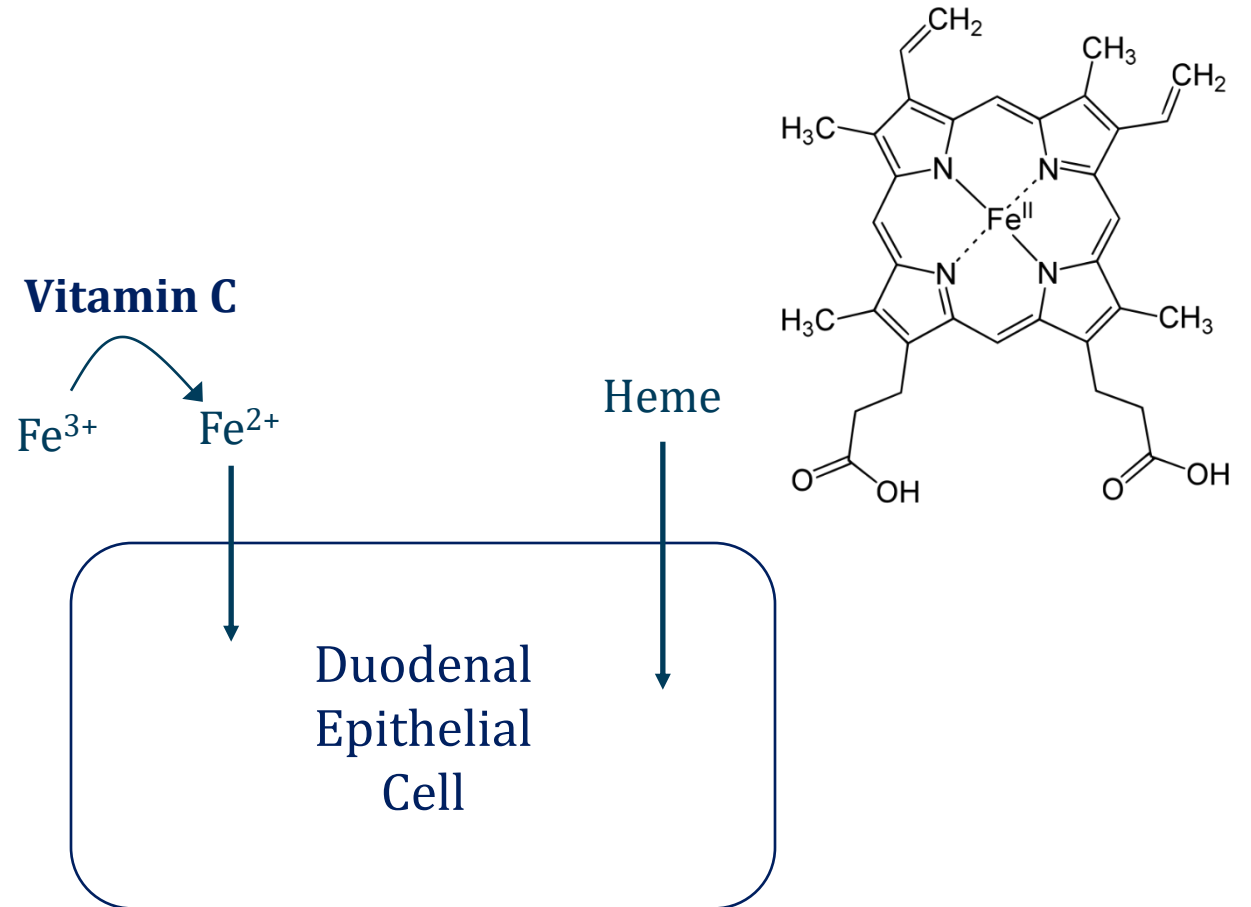
- Spectrum of RBC size
- Often increased in **iron deficiency**
- Normal RDW makes iron deficiency unlikely → consider thalassemia



Iron Deficiency

Underlying Mechanisms

- **Lack of iron uptake from gut**
 - Heme iron easily absorbed
 - Fe^{2+} iron easily absorbed
 - Fe^{3+} + iron poorly absorbed
- **Loss of iron**
 - Usually due to chronic blood loss



Iron Deficiency

Inadequate GI uptake

- **Babies**
 - Iron stores depleted ~ 6 months
 - Recommendation: add iron-containing foods
 - Exclusive breast feeding → iron deficiency
- **Malabsorption**
 - Any disease affecting duodenum or acid production
 - Loss of acid → **more Fe³⁺**
 - Status post gastrectomy
 - Proton pump inhibitors
 - Celiac disease
- Rarely malnutrition

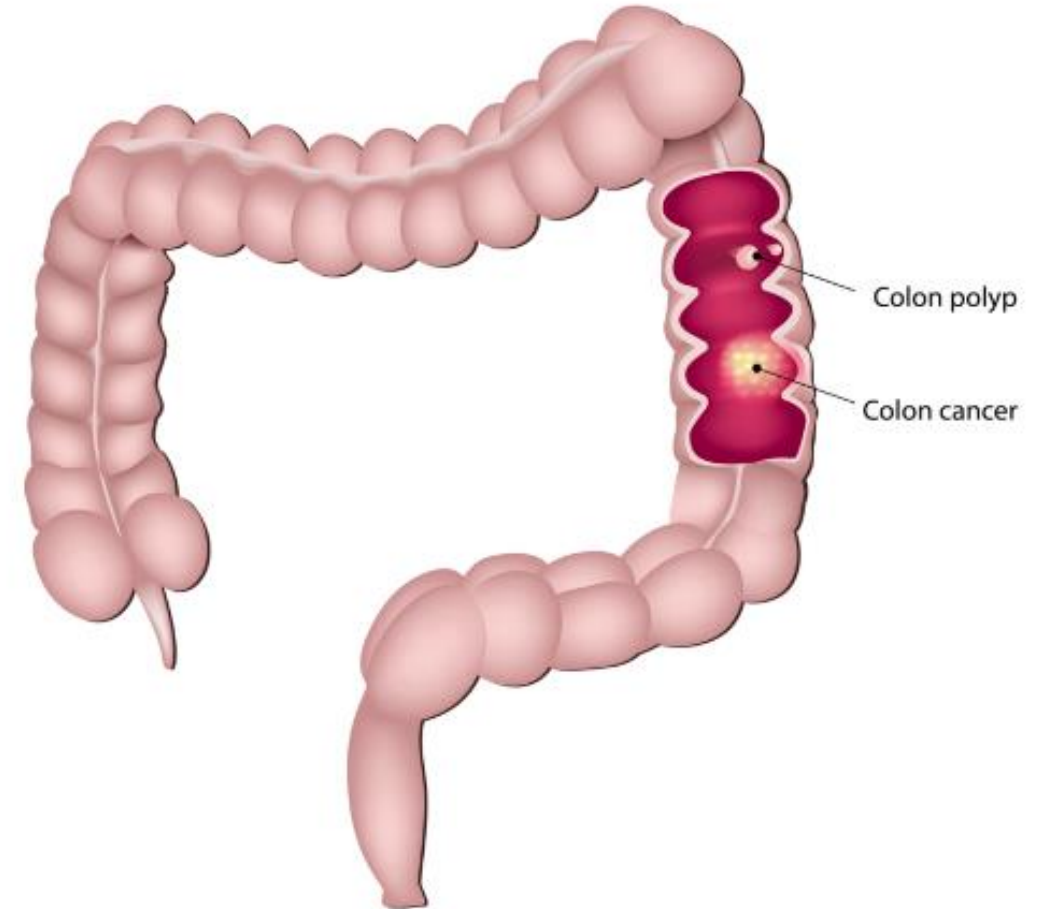


Iron Deficiency

Loss of iron

- **Chronic or recurrent blood loss**
 - Menstruation
 - Peptic ulcers
 - Colon cancer
- Iron deficiency: must consider **GI blood loss**
- Colon cancer can present as iron deficiency

COLON CANCER AND POLYP



Iron Deficiency

Other causes

- **Pregnancy**
 - “Negative iron balance” in pregnancy
 - Expansion in mothers Hgb mass
 - ↑ demand of fetal growth
 - Prenatal vitamins often contain **iron and folate**



Øyvind Holmstad/Wikipedia

Iron Deficiency Anemia

Clinical features

- Fatigue, pallor, dyspnea
- **Pica**
 - Craving for non-food substances
 - Clay or dirt (geophagia)
 - Ice (pagophagia)
- **Restless legs syndrome**

Restless Leg Syndrome



Iron Deficiency Anemia

Treatment

- Severe and highly symptomatic: RBC transfusion
 - Severely reduced hematocrit or myocardial ischemia
- **Oral iron**
 - Gastrointestinal side effects very common
 - Constipation, diarrhea, epigastric pain, nausea/vomiting
 - Black, green or tarry stools
- **Intravenous iron**
 - Used when GI side effects prohibit oral replacement
 - Or patients with malabsorption
 - Pregnancy (more rapid repletion of iron)



Anemia of Chronic Disease

- Anemia in association with **inflammation**
- Common in rheumatoid arthritis, lymphoma, other chronic conditions
- Usually a **mild anemia (Hgb > 10g/dL)**
- Symptoms from anemia rare
- Often incidental finding on blood testing
- Normocytic anemia ~75% of cases
- Microcytic anemia ~ 25% of cases

Rheumatoid Arthritis



Anemia of Chronic Disease

Mechanisms

- Triggered by cytokines
- **Increased hepcidin**
 - Acute phase reactant
 - Produced by liver
 - Inhibits iron transport
- Iron trapped in cells as **ferritin**
 - Ferritin usually increased
 - Contrast with iron deficiency

Normal Liver



Anemia of Chronic Disease

Diagnosis

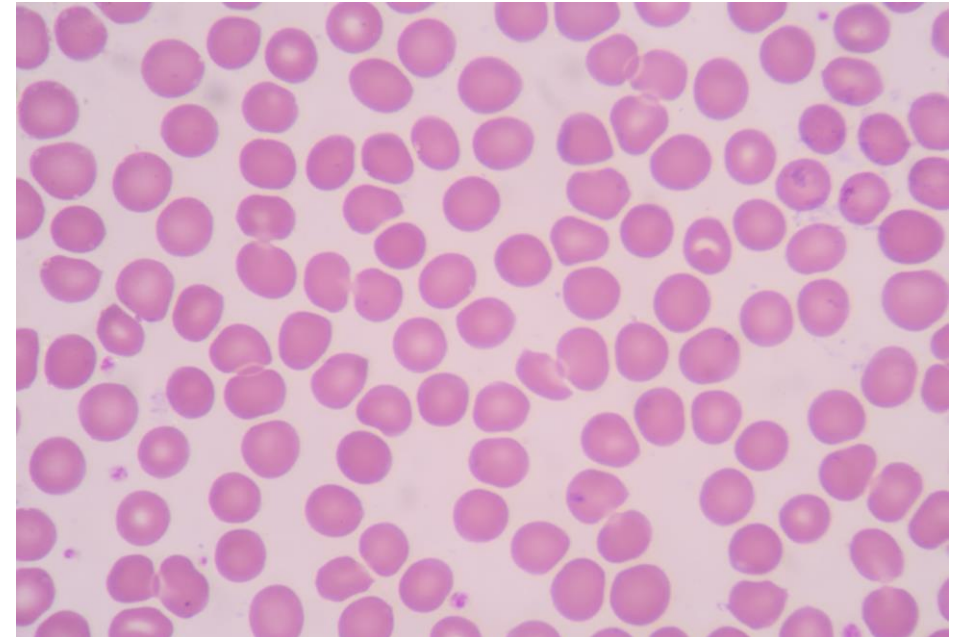
- **Serum iron low**
 - Thought to be protective
 - Bacteria may use iron for growth/metabolism
- **Ferritin not low – normal or increased**
 - Iron trapped in storage form
 - Ferritin is acute phase reactant
- Transferrin (TIBC) is usually low
- % saturation usually low
- Low reticulocyte count (↓ Hgb)

Test	Fe Deficiency	Chronic Disease
Serum iron	↓	↓
Serum ferritin	↓	NI or ↑
TIBC	↑	↓
% saturation	↓	↓
Reticulocytes	↓	↓

Anemia of Chronic Disease

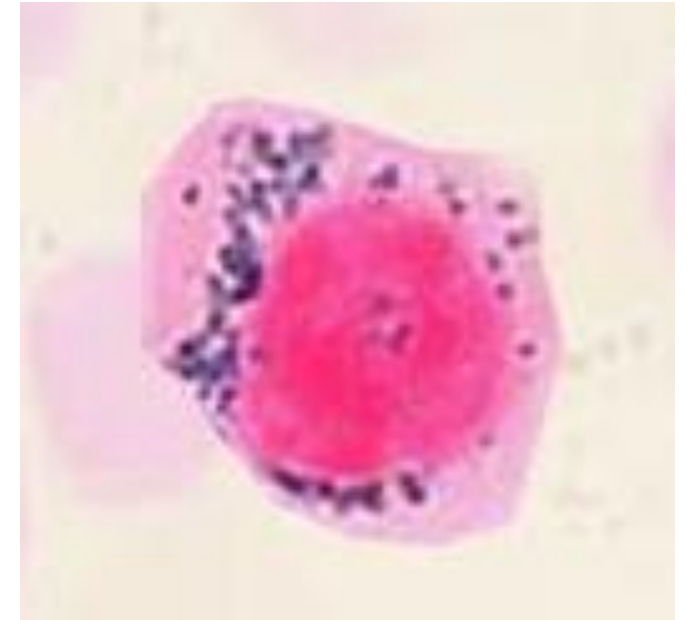
- Usually **normocytic and normochromic**
- Microcytic/hypochromic in about 25% cases
 - Low iron availability may lead to small red cells
 - MCV usually mildly decreased (70-80)
- Important to distinguish from iron deficiency
 - Does not respond to iron
- First line therapy: **treat underlying disease**
 - NSAIDs, glucocorticoids, chemotherapy

Normocytic Normochromic Red Cells



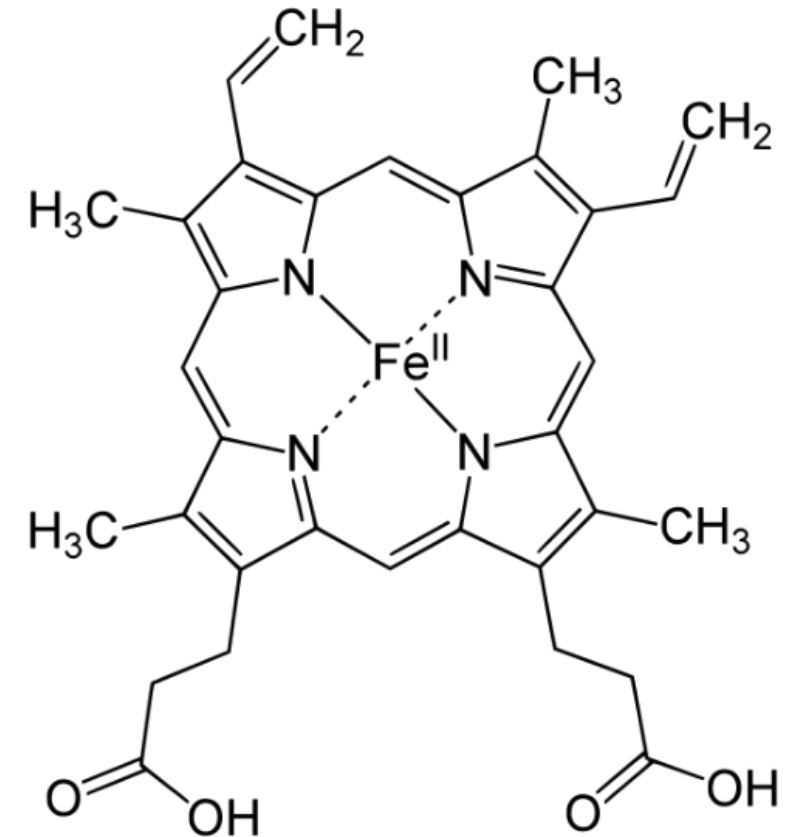
Sideroblastic Anemia

- **Ring sideroblasts**
 - Nucleated red cell precursors
 - Iron-loaded mitochondria seen with Prussian blue stain
 - Perinuclear ring of blue granules
- **Sideroblastic anemia**
 - Rare cause of anemia
 - Usually microcytic anemia
 - Ring sideroblasts in marrow



Sideroblastic Anemia

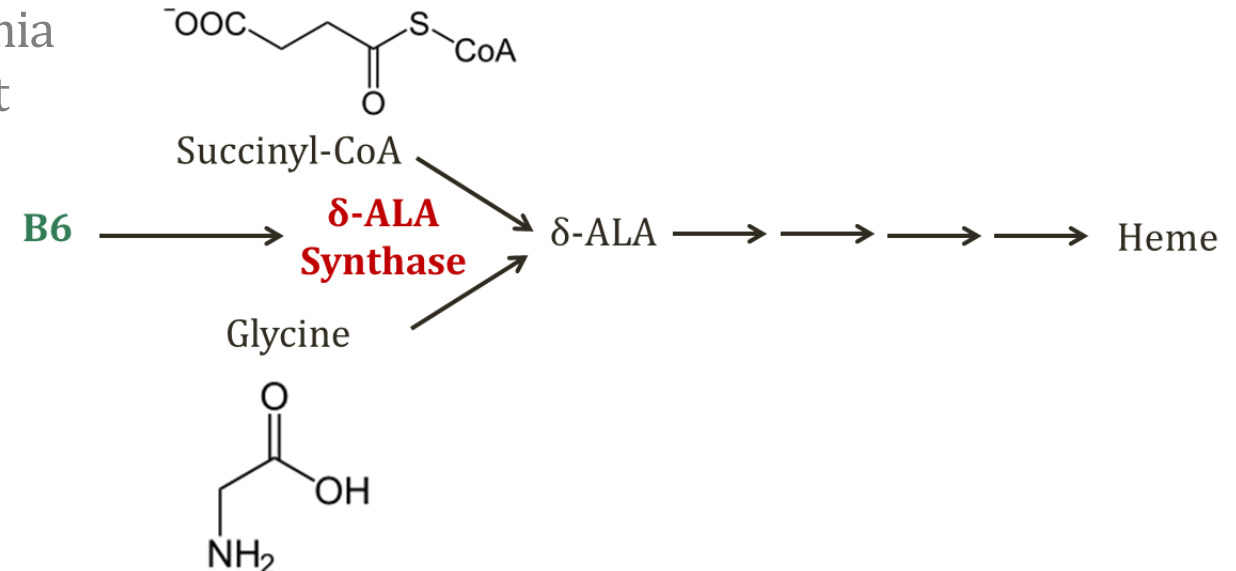
- Failure to make **protoporphyrin**
- Iron cannot bind protoporphyrin to form heme
- Iron accumulation in mitochondria



Heme

Sideroblastic Anemia

- In adults often **acquired** secondary to a **toxin**
 - Alcohol (mitochondrial poison)
 - Vitamin B6 deficiency (isoniazid)
- X-linked sideroblastic anemia
 - Rare, inherited deficiency of **ALA synthase**
 - Most common hereditary sideroblastic anemia
 - B6 supplementation often used as treatment



Sideroblastic Anemia

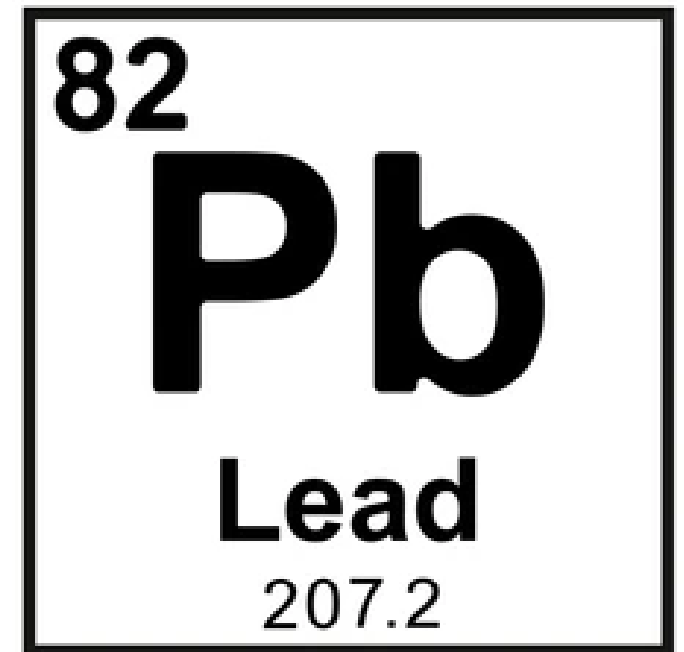
Diagnosis

- Microcytic, hypochromic anemia
- Iron studies often show **iron overload**
 - ↑ serum iron
 - ↑ ferritin
- Diagnosis: **bone marrow biopsy**
 - Ringed sideroblasts
- Often clues from history
 - Isoniazid treatment

Test	Fe Deficiency	Chronic Disease	Sideroblastic
Serum iron	↓	↓	↑
TIBC	↑	↓	↓/--
Serum ferritin	↓	↑	↑
% saturation	↓	Normal	↑/--
Reticulocytes	↓	↓	↓

Lead Poisoning

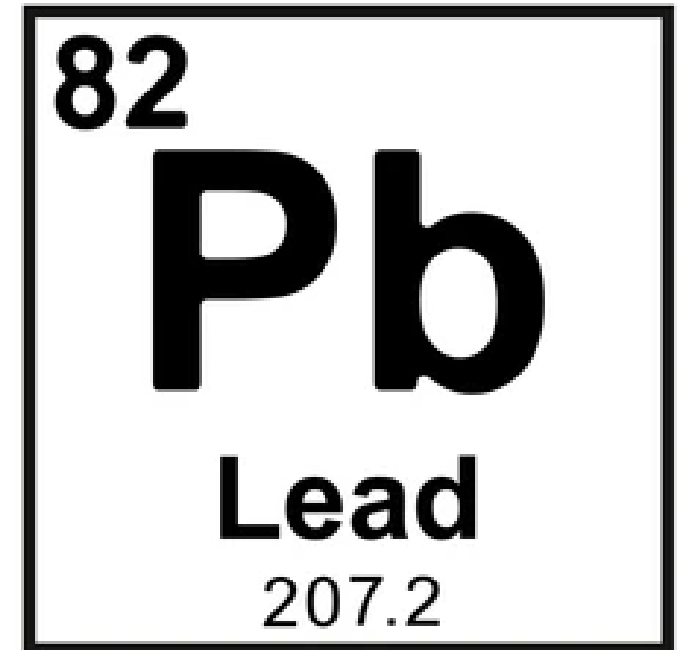
- Inhibits heme synthesis via two enzymes
 - Delta-aminolevulinic acid (δ -ALA) dehydratase
 - Ferrochelatase
- \downarrow heme synthesis \rightarrow **microcytic, hypochromic anemia**
 - Requires chronic exposure (usually months)
- Adults: **inhalation** from industrial work (battery factory)
- Children: eating lead-paint or contaminated water



Lead Poisoning

Clinical Features

- Affects many organ systems
- GI: abdominal pain, constipation, anorexia
- Neurologic: behavioral changes, poor concentration
- Diagnosis: serum lead level
 - ≥ 5 mcg/dL = elevated blood lead level
- Treatment: remove exposure +/- **chelation**
 - Calcium disodium EDTA (ethylenediaminetetraacetate)
 - DMSA (2,3-dimercaptosuccinic acid; succimer)



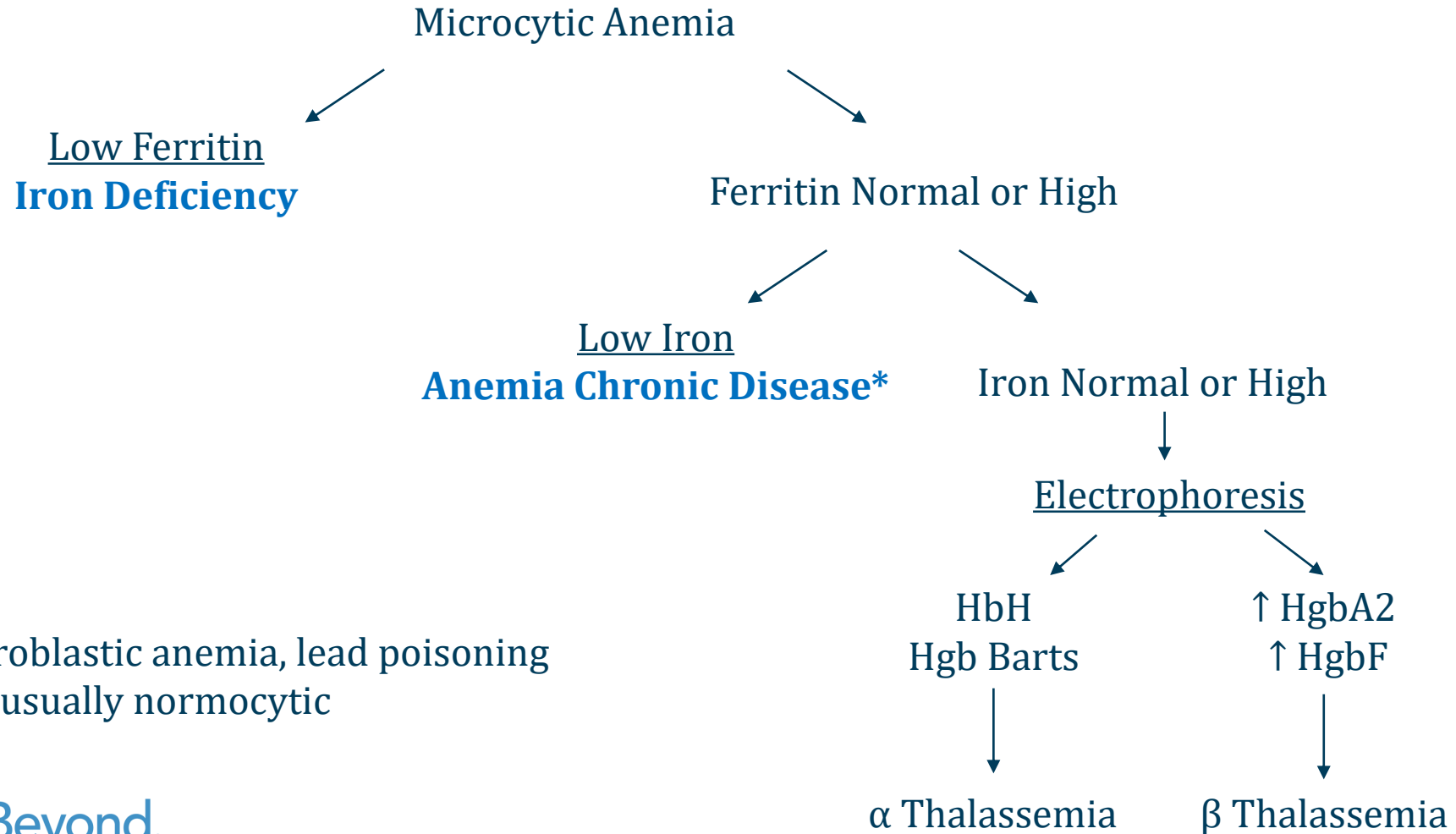
Microcytosis

Evaluation

- MCV less than 80 fL
- Most common causes: **iron deficiency** and **thalassemia**
- Key diagnostic test: **serum ferritin**
 - Low = iron deficiency
 - Normal or high = other causes
- Most common cause iron deficiency in adults: **blood loss**
- Most common source blood loss: **GI tract**
 - Consider colonoscopy for colon cancer

Microcytic Anemias

Workup



Rare causes: sideroblastic anemia, lead poisoning

*ACD: usually normocytic

Thalassemias

Jason Ryan, MD, MPH



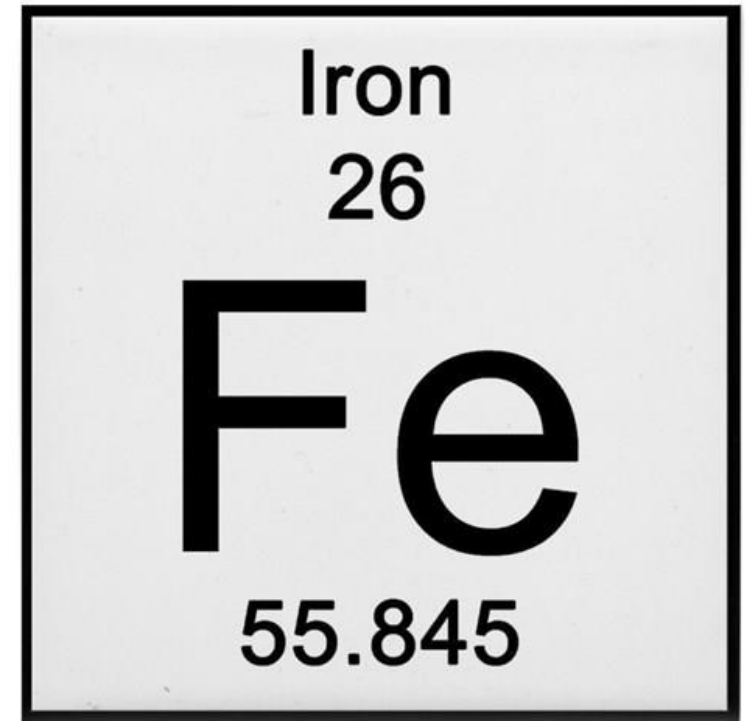
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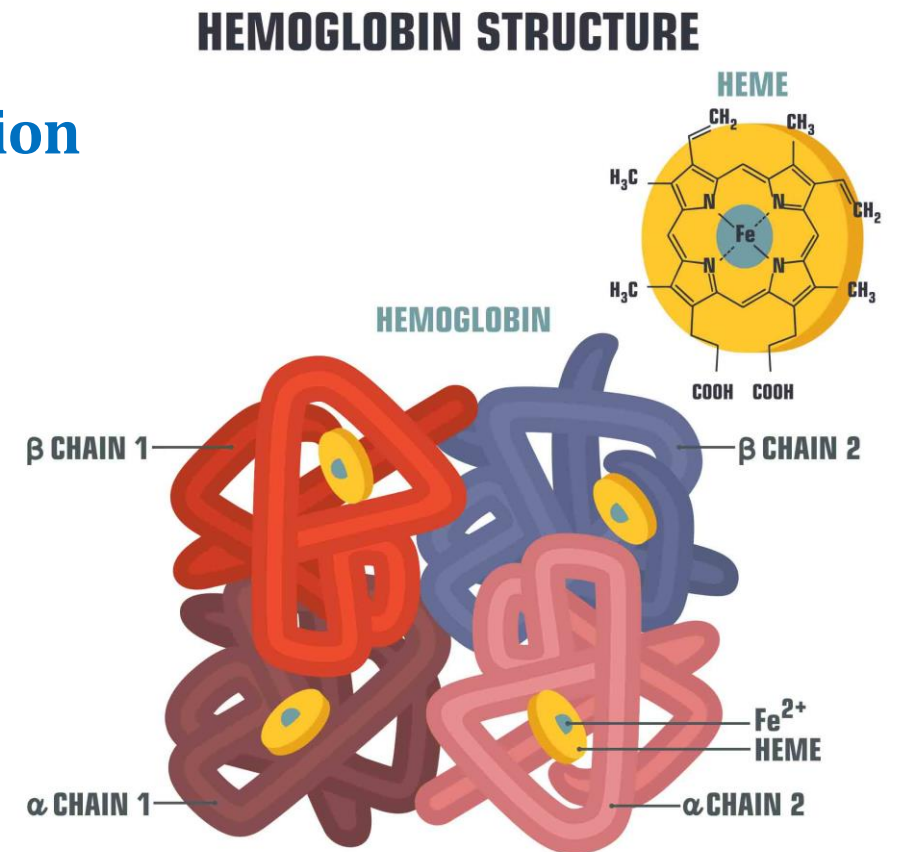
Microcytic Anemias

- Usually due to ↓ **hemoglobin** in red cells
- Very common cause: **iron deficiency**
 - Check iron studies before considering other causes
 - Consider thalassemia if iron studies show adequate iron stores



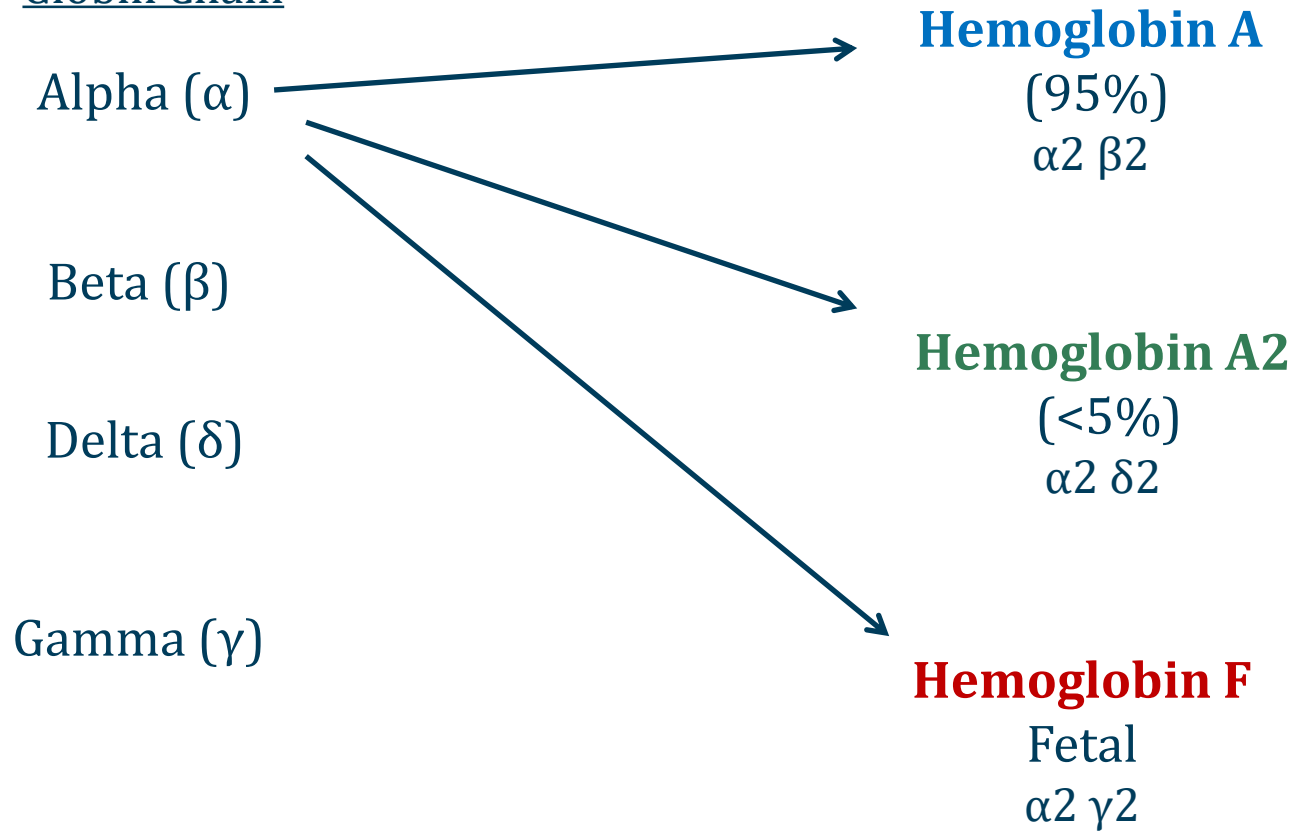
Hemoglobinopathies

- Abnormal hemoglobin
- Structural disorders: sickle cell disease
- Thalassemias: **decreased globin chain production**
- Alpha thalassemia: alpha globin
- Beta thalassemia: beta globin



Globin Chains and Hemoglobin

Globin Chain



**All Hgb has two alpha globins
Other chain determines type**

Globin Chains and Hemoglobin

Globin Chain

Alpha (α)

Beta (β)

Delta (δ)

Gamma (γ)

Hemoglobin A

(95%)
 $\alpha_2 \beta_2$

Hemoglobin A2

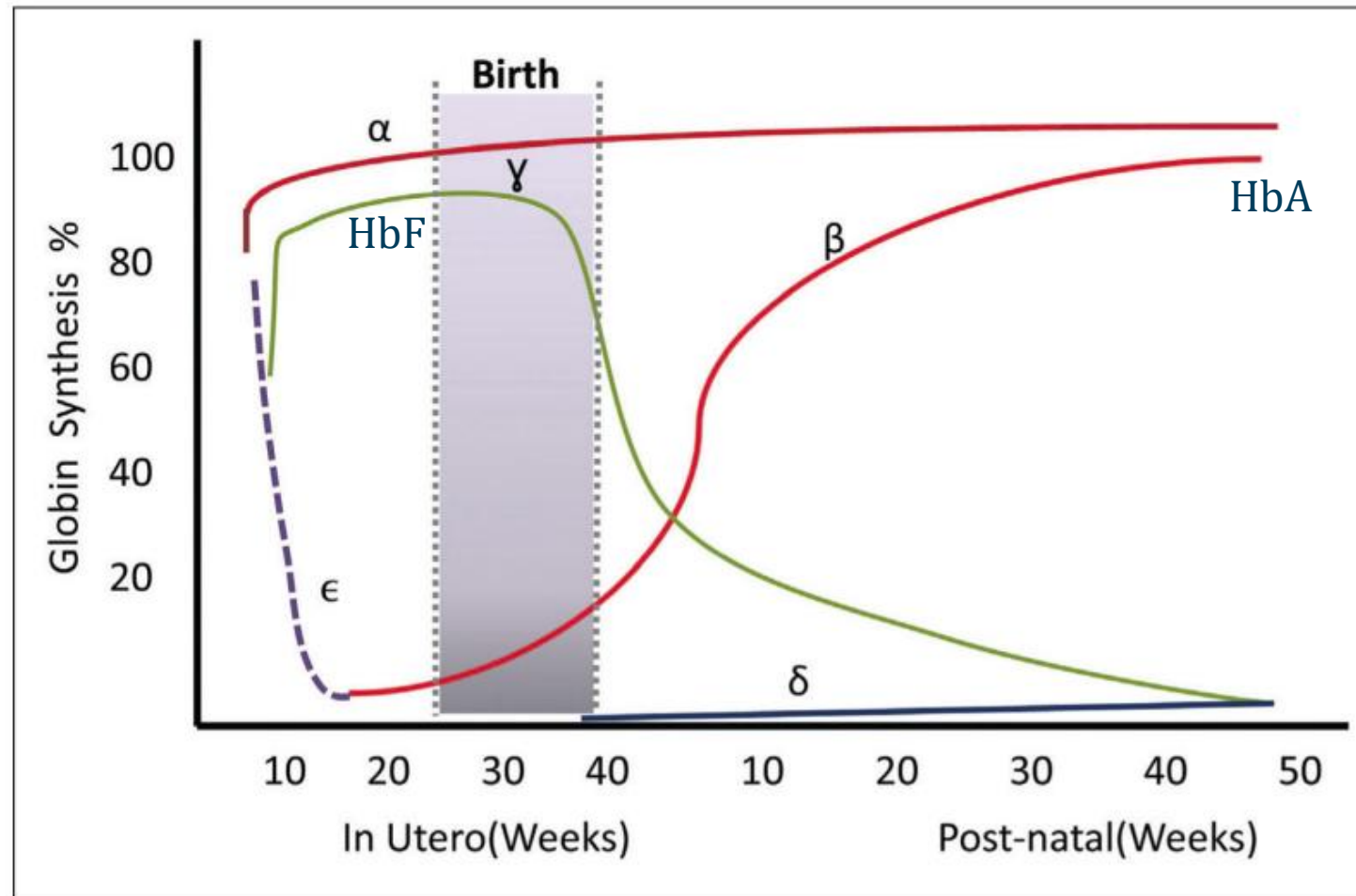
(<5%)
 $\alpha_2 \delta_2$

Hemoglobin F

Fetal
 $\alpha_2 \gamma_2$

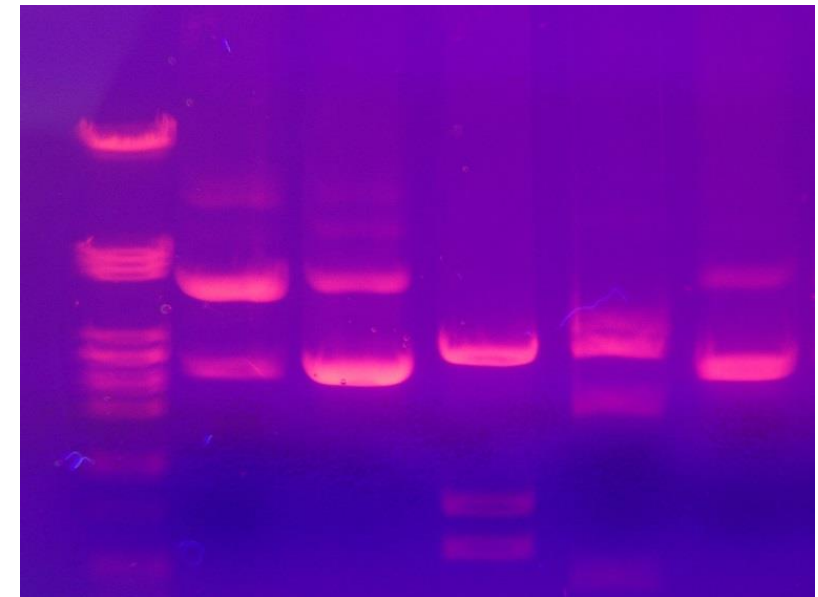
All Hgb has two alpha globins
Other chain determines type

Globin Chains and Hemoglobin



Hgb Electrophoresis

- Used to diagnose **hemoglobinopathies**
 - Thalassemia
 - Sickle cell disease
- Electrical charge applied to sample on gel
- Different hemoglobins → different distances moved
- Determines HgbA, HgbA2, HgbF
- Often done for microcytic anemia + no iron deficiency



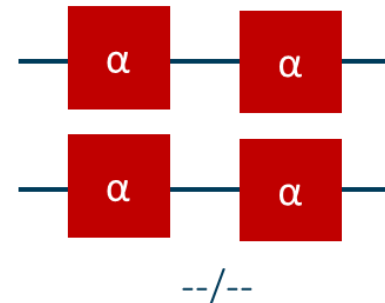
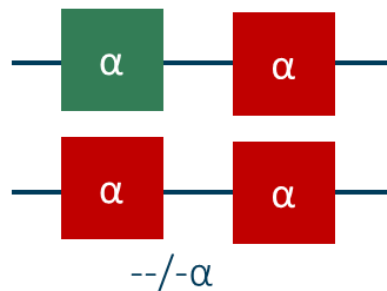
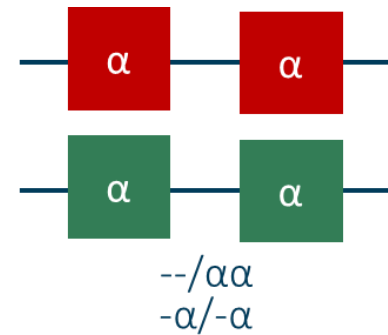
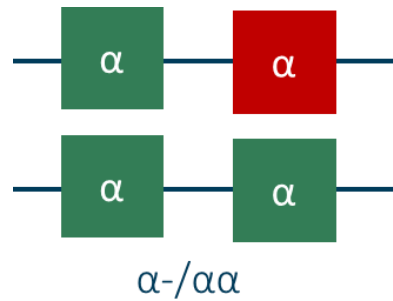
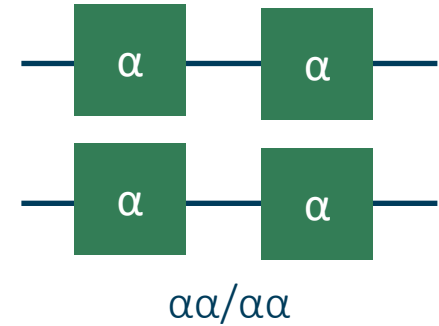
Thalassemias

- Spectrum of severity
- Thalassemia **minor**
 - Often asymptomatic
 - Identified on routine blood testing or blood smear
 - May have **microcytosis with ↑ red cell count**
 - Contrast with iron deficiency: microcytosis with ↓ RBC
- Thalassemia **major**
 - Severe loss of globin production
 - Lifelong transfusions or death



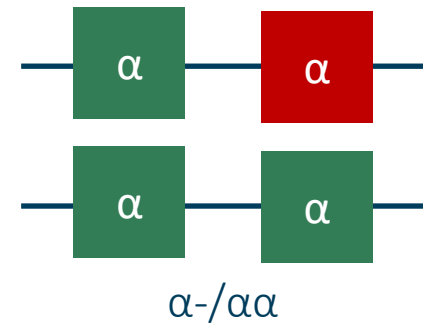
Alpha Thalassemia

- **Four genes** code for alpha chains
- Two on each copy of chromosome 16
- Often caused by **gene deletions** → ↓ α chains → alpha thalassemia



Alpha Thalassemia Minima

- Normal red cells
- No symptoms
- Carrier state



Alpha Thalassemia Minor

Alpha Thalassemia Trait

- No symptoms
- ↓ MCV/MCH/MCHC
- Electrophoresis: usually normal
- Diagnosis: genetic testing

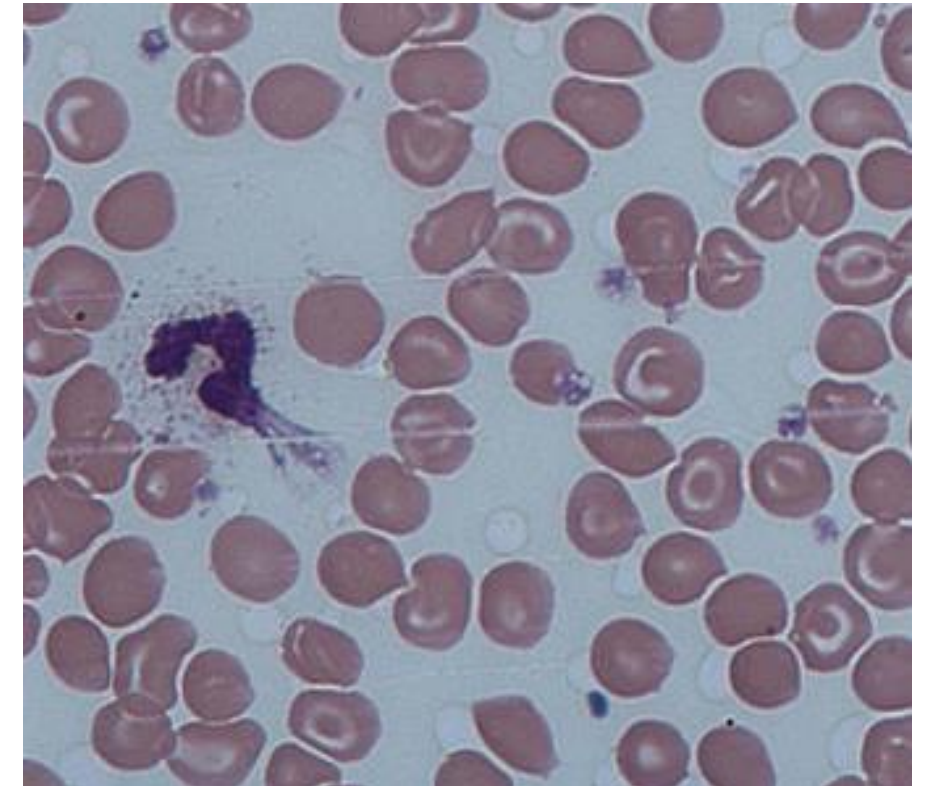


Thalassemia Minor

Differential diagnosis

- Alpha and beta minor can mimic **iron deficiency**
- Both may lead to low MCV and MCH
- Key distinguishing factors for thalassemia:
 - **Red cell count normal or increased**
 - RDW normal
 - Iron and ferritin normal or increased (↑ turnover)
- **Target cells**
 - Classic finding in thalassemia
 - Caused by increased surface to volume ratio
 - Rarely seen in iron deficiency

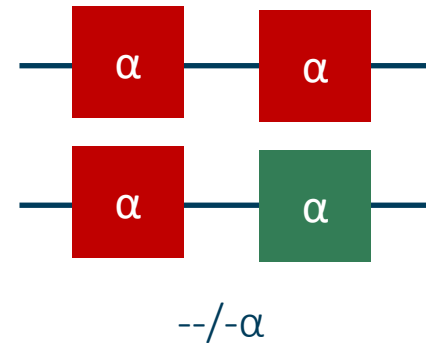
Target Cells



Hemoglobin H Disease

Alpha thalassemia intermedia

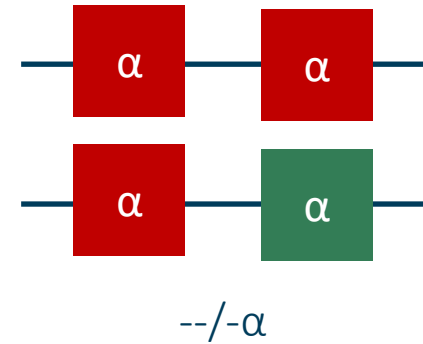
- Very little alpha globin production
- Excess beta globin
- **HbH forms: four beta chains**
 - Easily damaged
 - Affinity for oxygen 10x HbA
 - Poor oxygen delivery



Hemoglobin H Disease

Clinical features

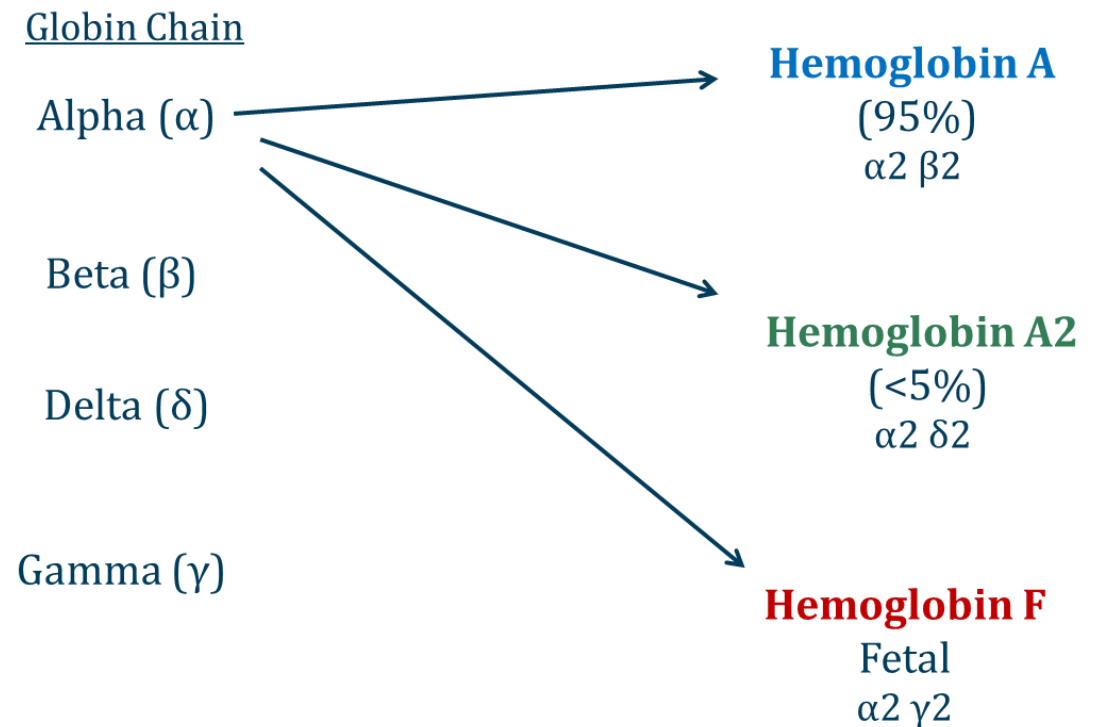
- Highly variable presentation
- Often symptomatic in infancy
- Hypochromic, microcytic anemia
- Low MCV, MCH, MCHC
- **Extravascular hemolysis**
 - Abnormal RBC deformability
 - Splenomegaly
 - Indirect hyperbilirubinemia
 - May cause neonatal jaundice
- Bizarre red cell morphologies



Hemoglobin H Disease

Diagnosis

- **Electrophoresis**
 - HBH
 - Decreased HbA, HbA₂, HbF
- **DNA testing**



Hemoglobin H Disease

Treatment

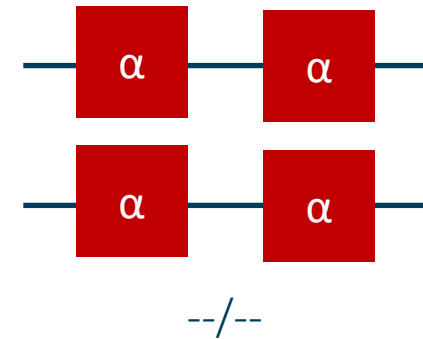
- **Blood transfusions**
 - Usually not transfusion dependent
 - May require **episodic transfusions** with stress
 - Especially during infection or pregnancy
 - Or with aplastic crisis from B19 infection
- Splenectomy
- Bone marrow transplantation
- Long term risk: **iron overload**
 - Ineffective erythropoiesis → ↑ intestinal iron uptake
 - Also from transfusions



Hemoglobin Barts

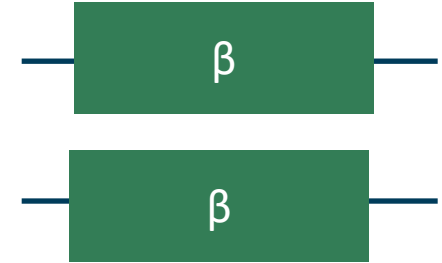
Alpha thalassemia major

- **No α globin**
- Cannot form HbF
- **Hgb Barts**
 - Forms in utero
 - Four gamma globin chains
 - Cannot release oxygen to tissues
- **Hydrops fetalis**
 - Massive total body edema
 - High output heart failure
- In utero death or death hours after birth



Beta Thalassemia

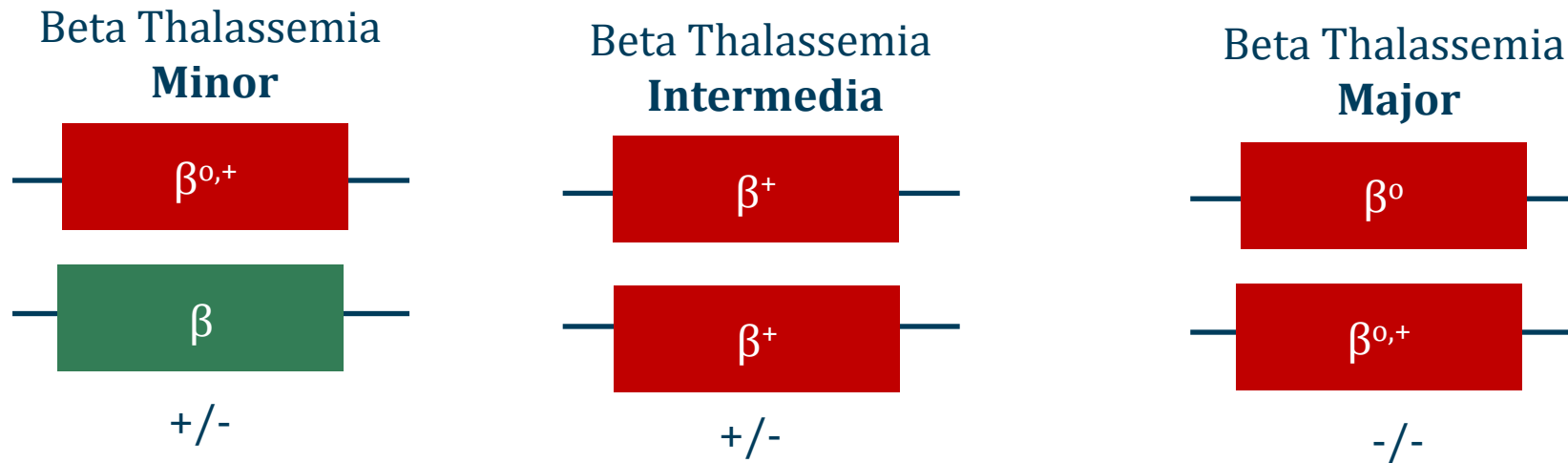
- \downarrow β globin chain synthesis
- Two genes code for beta chains
- One on each copy of chromosome 11
- Often caused by mutations (not deletions)
- Wide spectrum of disease depending on mutation
 - β^0 = no function; β^+ = some function



β

Beta Thalassemia

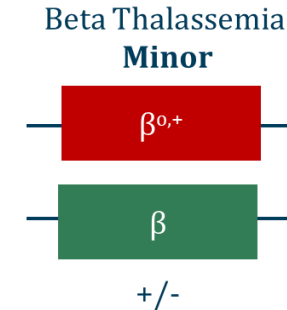
- Previously classified as minor, intermedia or major
- Now classified as **transfusion-dependent** or **transfusion-independent**



Beta Thalassemia Minor

Beta Thalassemia Trait

- Reduced β globin synthesis
- Asymptomatic
- Possible microcytosis with \uparrow RBC
- Diagnosis by electrophoresis
- **\uparrow HgbA2**
 - HgbA2: $\alpha_2\delta_2$ – no beta chains
 - Normal $\sim 3\%$



Globin Chain

Alpha (α)

Beta (β)

Delta (δ)

Gamma (γ)

Hemoglobin A

(95%)
 $\alpha_2\beta_2$

Hemoglobin A2

(<5%)
 $\alpha_2\delta_2$

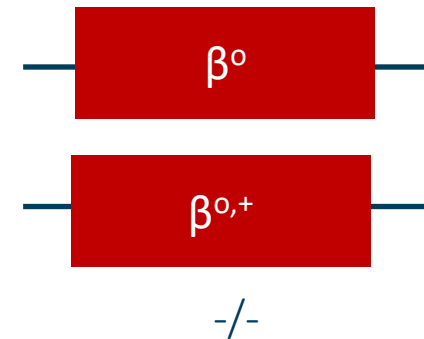
Hemoglobin F

Fetal
 $\alpha_2\gamma_2$

Transfusion-Dependent Beta Thalassemia

Beta Thalassemia Major

- No or severely limited β globin production
- Anemia beginning **1st year of life**
 - Usually begins 6 to 12 months
 - Occurs when HgbF ($\alpha 2\gamma 2$) production wanes
- Ineffective erythropoiesis
 - Alpha chains form tetramers
 - Precipitate \rightarrow RBC damage
 - Failure to produce RBCs
- **Life-long transfusion-dependent anemia**

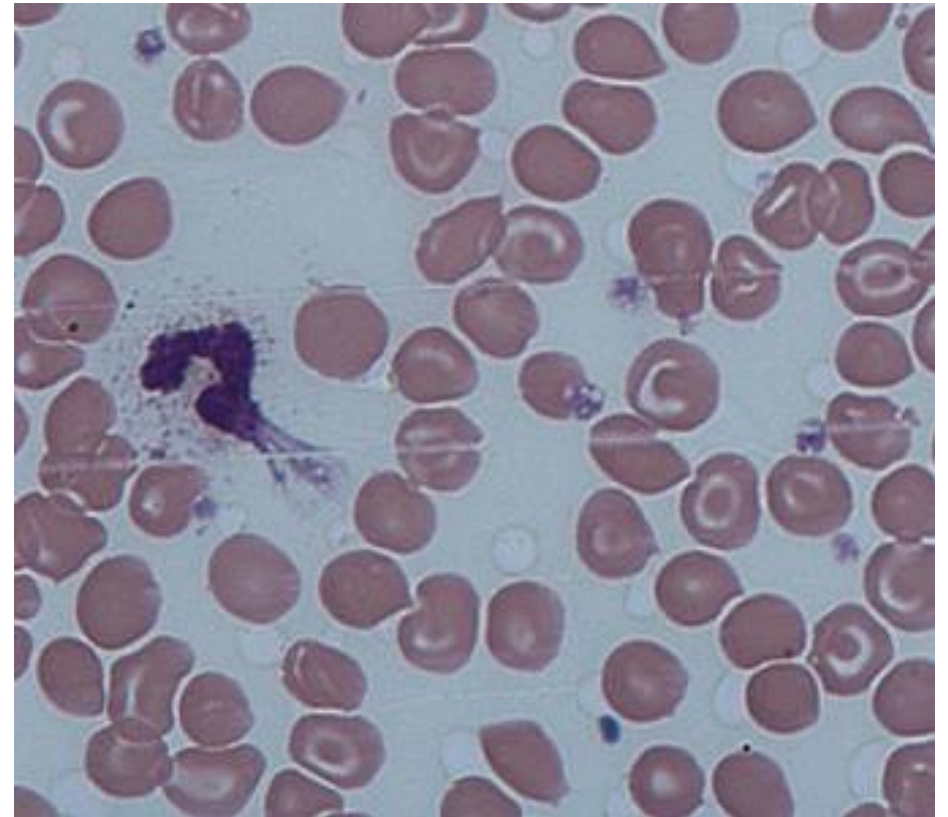


Transfusion-Dependent Beta Thalassemia

Clinical Features

- Hypochromic, microcytic anemia
- Bone changes
- Iron overload
 - Ineffective erythropoiesis → ↑ intestinal iron uptake
 - Also from transfusions
- Splenomegaly
 - Spleen clears abnormal RBCs
- Bizarre red cell morphology
 - Abnormal size and shapes
 - Many abnormalities possible

Target Cells



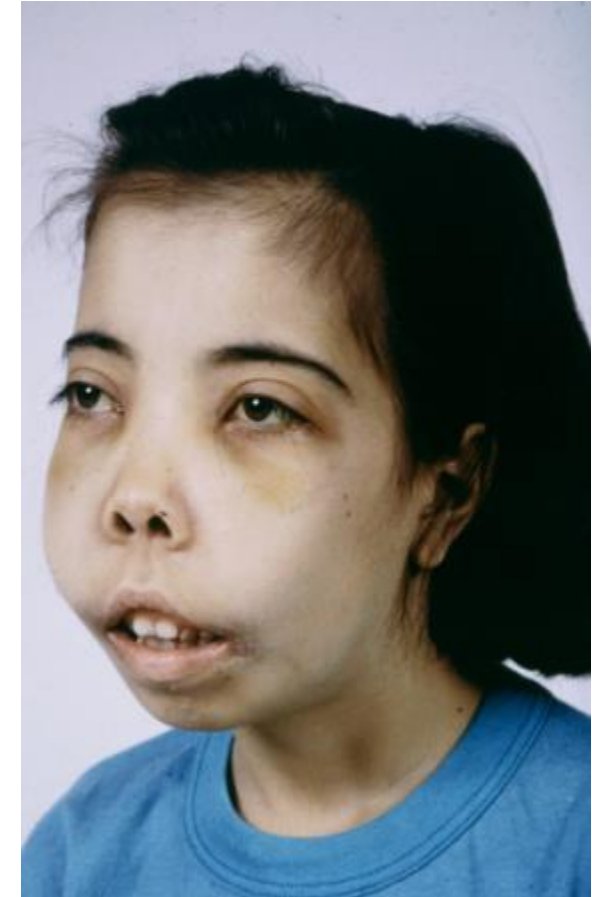
Dr Graham Beards

Transfusion-Dependent Beta Thalassemia

Bone Changes

- ↑↑ EPO without normal response
- Erythroid hyperplasia
- Massive expansion of bone marrow
- Abnormalities of **skull and facial bones**
- “Chipmunk facies”
- Delayed skeletal maturation
- Widening of marrow spaces → osteoporosis

Chipmunk Facies



Transfusion-Dependent Beta Thalassemia

Diagnosis

- **Electrophoresis**
 - ↓ or absent HbA ($\alpha_2\beta_2$)
 - ↑ HbA2 ($\alpha_2\delta_2$)
 - ↑ HbF ($\alpha_2\gamma_2$)
- **Genetic testing**

Globin Chain

Alpha (α)

Beta (β)

Delta (δ)

Gamma (γ)

Hemoglobin A
(95%)
 $\alpha_2\beta_2$

Hemoglobin A2
(<5%)
 $\alpha_2\delta_2$

Hemoglobin F
Fetal
 $\alpha_2\gamma_2$

Transfusion-Dependent Beta Thalassemia

Treatment

- **Blood transfusions**
- Splenectomy
- Bone marrow transplantation
- Long term risk: **iron overload**
- **Iron chelation therapy** usually required
 - Deferasirox
 - Deferoxamine
 - Deferiprone



Folate Supplementation

- All patients with **thalassemia major**



Needpix.com/Public Domain

Red Cell Distribution Width

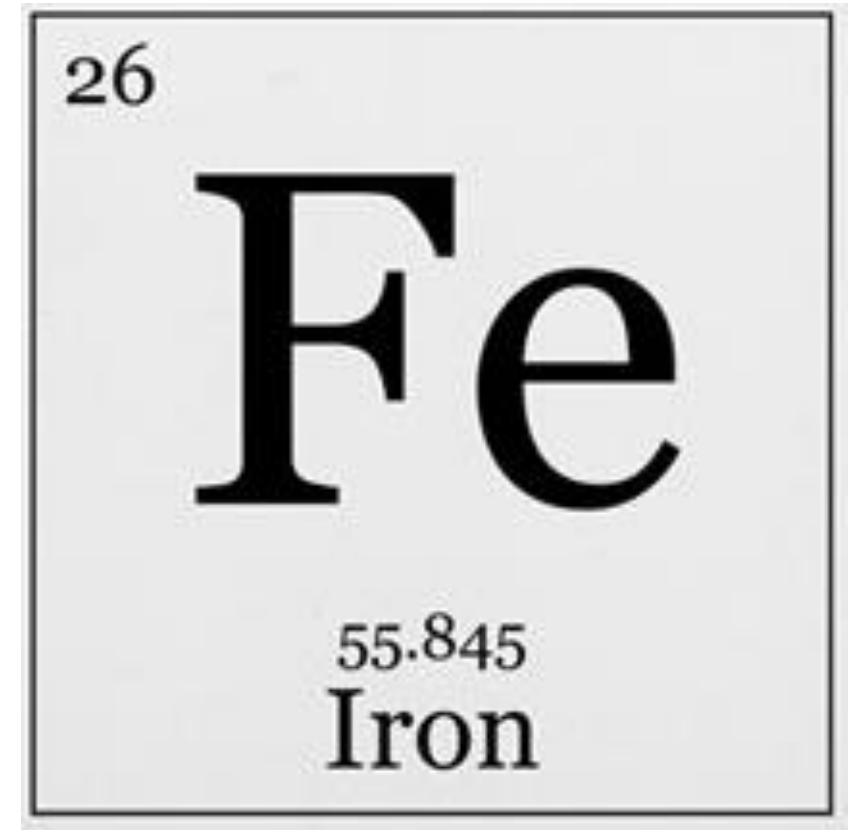
RDW

- Spectrum of RBC size
- Normal RDW: 11.5 to 14.5%
- Often normal in thalassemia
- Wider in iron deficiency (often $> 17\%$)
- Normal RDW makes iron deficiency unlikely
- Microcytic anemia + normal RDW: **almost always thalassemia**

Thalassemia

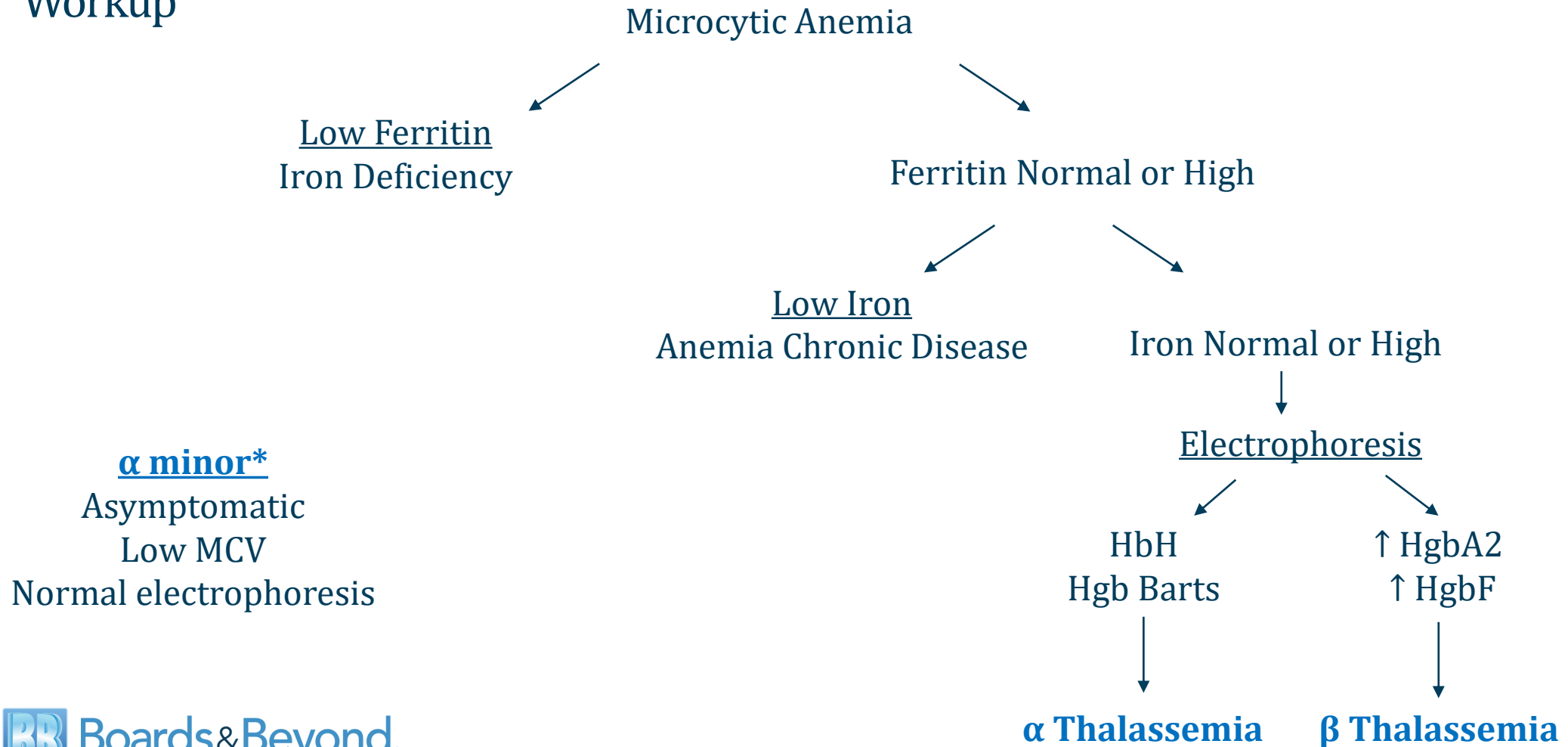
Iron Studies

- High red cell turnover
- **Serum iron level elevated** in severe forms
 - HbH, Beta major
 - Ineffective erythropoiesis → ↑ intestinal iron uptake
- Also may see **elevated ferritin**
- Low serum ferritin not seen in isolated thalassemia
 - Unless complicated by iron deficiency



Microcytic Anemias

Workup



Macrocytic Anemias

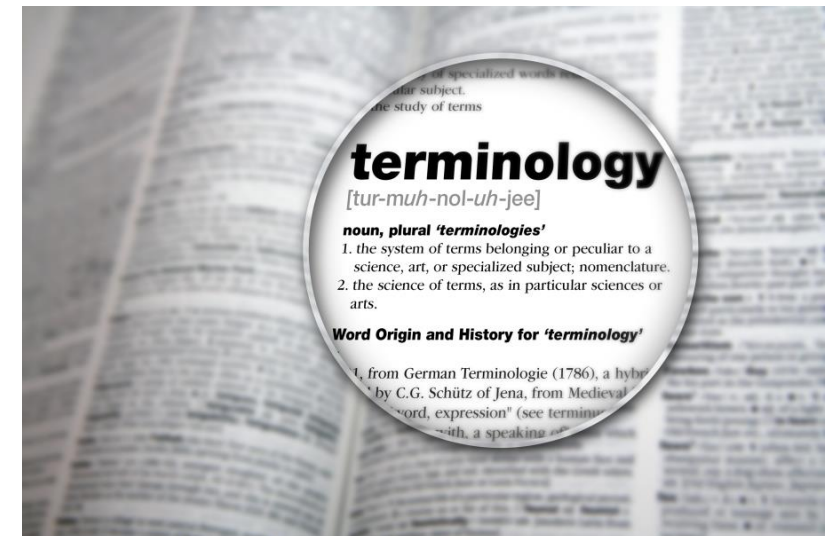
Jason Ryan, MD, MPH



Macrocytic Anemias

Terminology

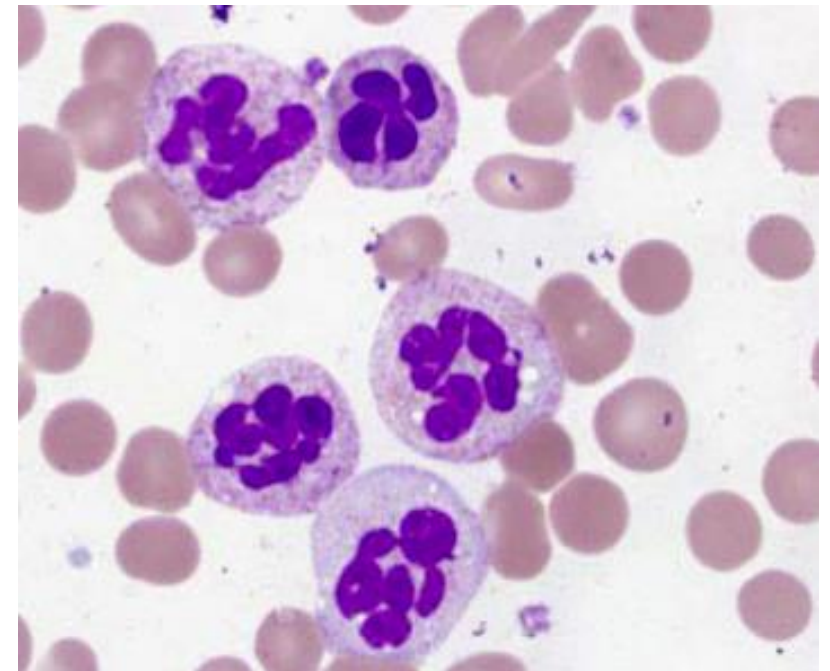
- Normal MCV: 80 to 100 fL
- Macrocytosis = \uparrow MCV
- Macrocytosis may occur with or without anemia
- Macrocytic anemia = \downarrow RBCs + \uparrow MCV
- Megaloblastic anemia = subtype of macrocytic anemia



Megaloblastic Anemia

- Impaired cell division in bone marrow
- Caused by arrested DNA synthesis
- Macrocytic anemia (\downarrow RBCs + \uparrow MCV)
- Large red cell precursors in marrow (megaloblasts)
- **Hypersegmented neutrophils**
 - Normal lobes = two to five
 - Hypersegmented = six or more lobes

Hypersegmented Neutrophils



Wikipedia/Public Domain

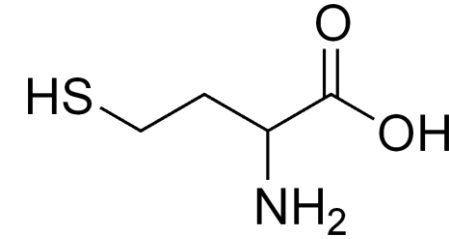
Megaloblastic Anemias

Causes

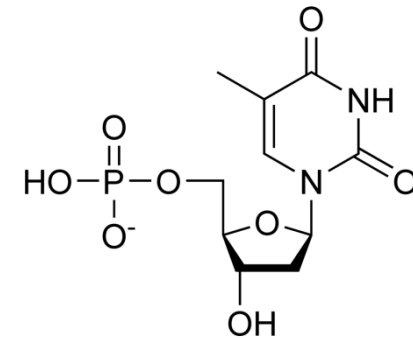
- **Vitamin B12 deficiency**
- **Folate deficiency**
- Drugs
- Inborn errors of metabolism (orotic aciduria)

Folate and Vitamin B12

- Both required for **DNA synthesis**
- Need for synthesis of **thymidine**
- Both required for metabolism of **homocysteine**
- Deficiency of either vitamin:
 - ↓ DNA synthesis → megaloblastic anemia
 - ↑ homocysteine



Homocysteine

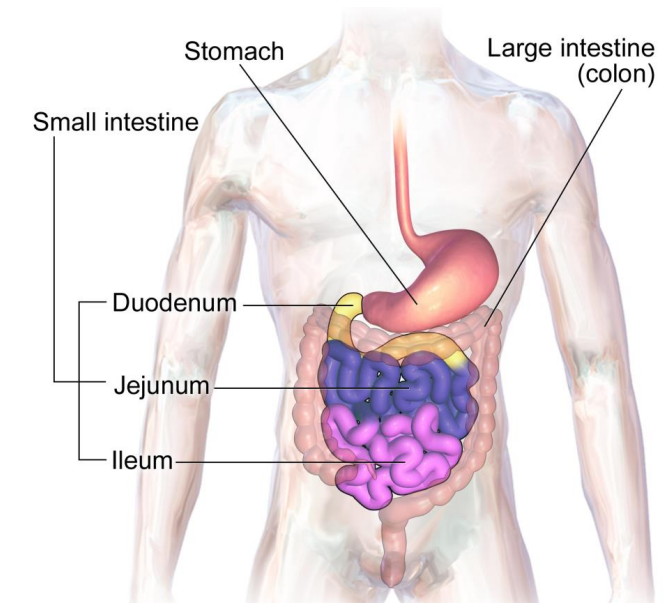
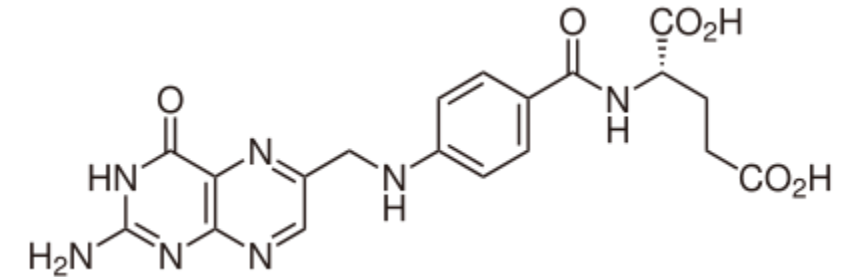


Thymidine
monophosphate

Folate

- Water soluble vitamin
- Cereals and grains fortified with folic acid
- Also found in green leafy vegetables
- Dietary deficiency very rare in the US
- Absorbed in the **jejunum**
- Dietary folate converted to **active form** in steps
 - Dihydrofolate
 - Tetrahydrofolate
 - **5,10-methylene THF** (used in DNA synthesis)

Folate



Folate Deficiency

Causes

- **Malnutrition**
 - Requires severely restricted diet
 - Heavy alcohol consumption
 - Anorexia
- **Increased use**
 - Pregnancy
 - Chronic hemolysis
- **Malabsorption**
 - Gastric bypass surgery
 - Celiac disease



Drugs

-
- The diagram illustrates the metabolic pathway of folate and the sites of action for Trimethoprim and Methotrexate. At the top, a vertical double line separates the 'Plasma' compartment on the left from the 'GI Tract' compartment on the right. In the GI Tract, 'Folate' is shown with a horizontal arrow pointing left towards 'Folate' in the Plasma. In the Plasma, another horizontal arrow points left from 'Folate' to 'DHF'. From 'DHF', a curved arrow points down and to the left towards 'THF'. The enzyme 'Dihydrofolate Reductase' is labeled in purple text, with a red arrow pointing to it from the text 'Trimethoprim' and 'Methotrexate' at the bottom right.

Folate Deficiency

Clinical findings and treatment

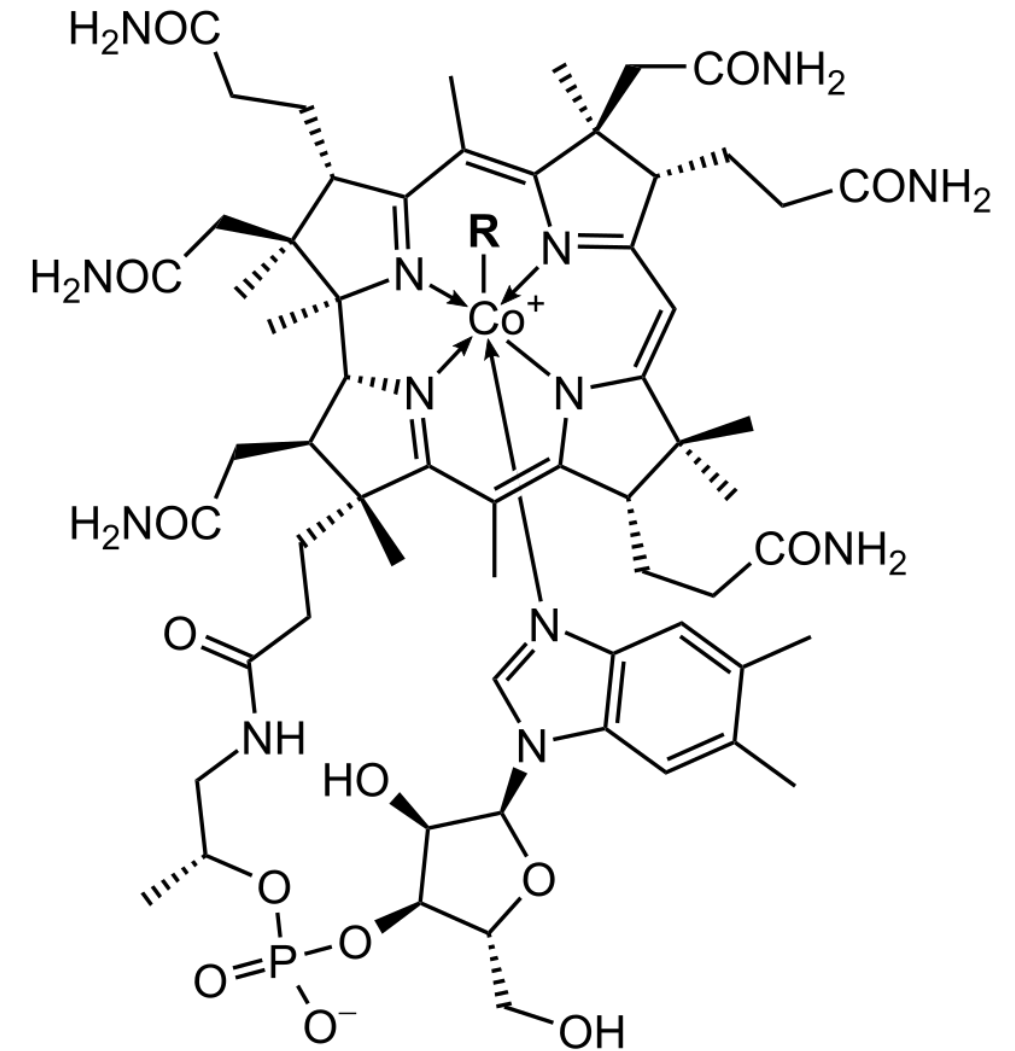
- Megaloblastic anemia
- Low reticulocyte count
- **Low serum folate level**
 - Normal: greater than 4 ng/mL
 - Borderline: 2 to 4 ng/mL
 - Low: less than 2 ng/mL
- Treatment: **folate supplementation**



Vitamin B12

Cobalamin

- Large, complex structure (corrin ring)
- Contains element cobalt
- Only synthesized by bacteria
- Found in meats, eggs, dairy

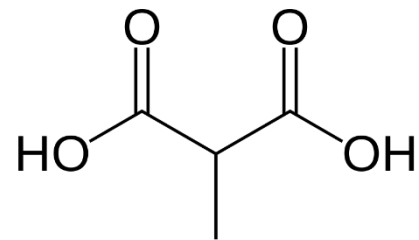


Vitamin B12

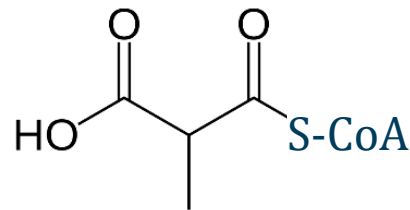
Metabolic roles

- Required for DNA synthesis and homocysteine metabolism
- Also required for metabolism of **methylmalonic acid**
- B12 deficiency: elevated methylmalonic acid

Methylmalonic Acid



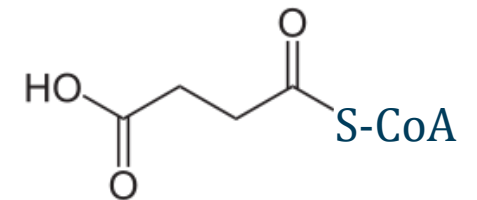
Methylmalonyl-CoA



Methylmalonyl-CoA
mutase

B12

Succinyl-CoA



Vitamin B12

Metabolic roles

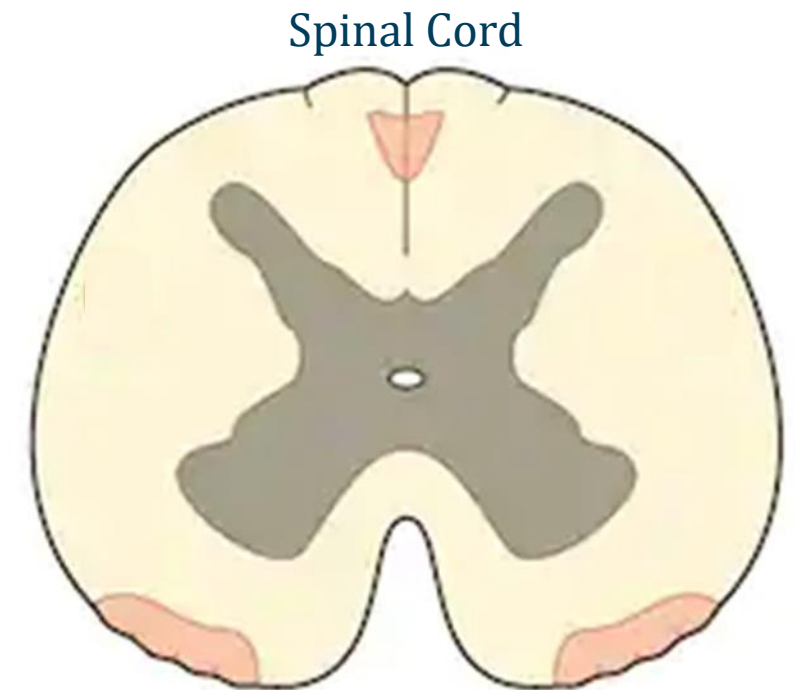
- Required for myelination of nerve fibers
- Deficiency: **slowly-progressive neuropathy**
- **Neuropsychiatric changes**
 - Depression
 - Irritability
 - Cognitive decline



Vitamin B12

Subacute combined degeneration

- Neuropathy associated with B12 deficiency
- Degeneration of posterior and lateral columns of spinal cord
- Slowly progressive symptoms
- **Legs** affected more than arms
- Weakness, ataxia, numbness and paresthesias
- Severe forms: spasticity, paraplegia and incontinence
- **Unsteady gait**
- Positive **Romberg sign**



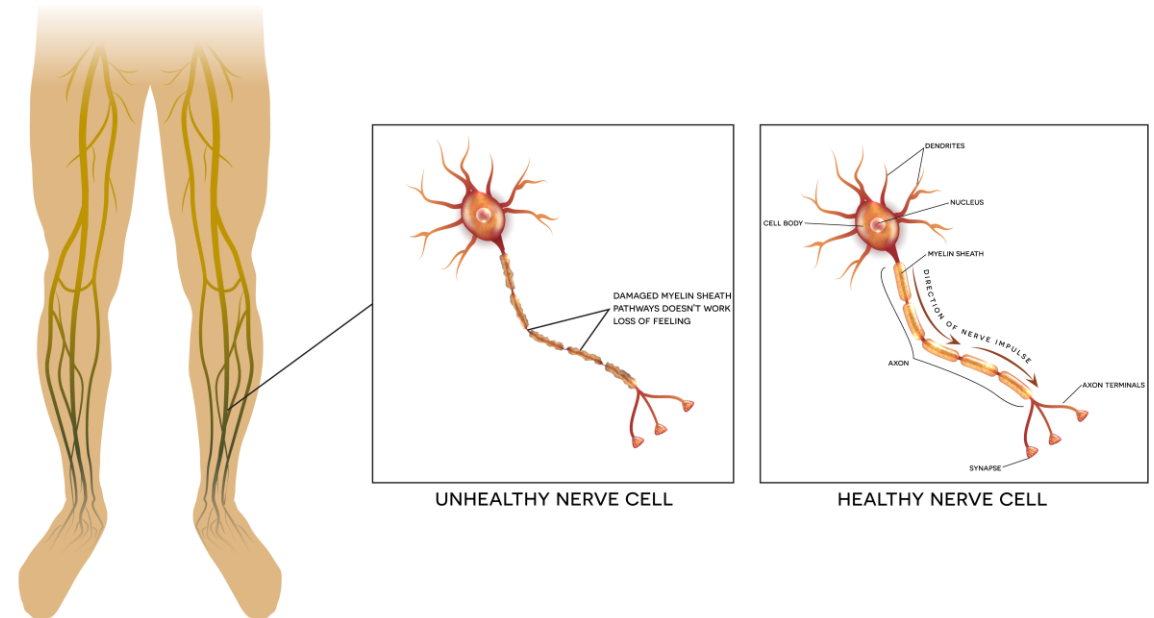
Vitamin B12 Deficiency

Clinical Features

- Megaloblastic anemia
- Low reticulocyte count
- **Symmetrical neuropathy**
- **Behavioral changes**
- Glossitis

PERIPHERAL NEUROPATHY

NERVE DAMAGE



Vitamin B12

Diagnosis and treatment

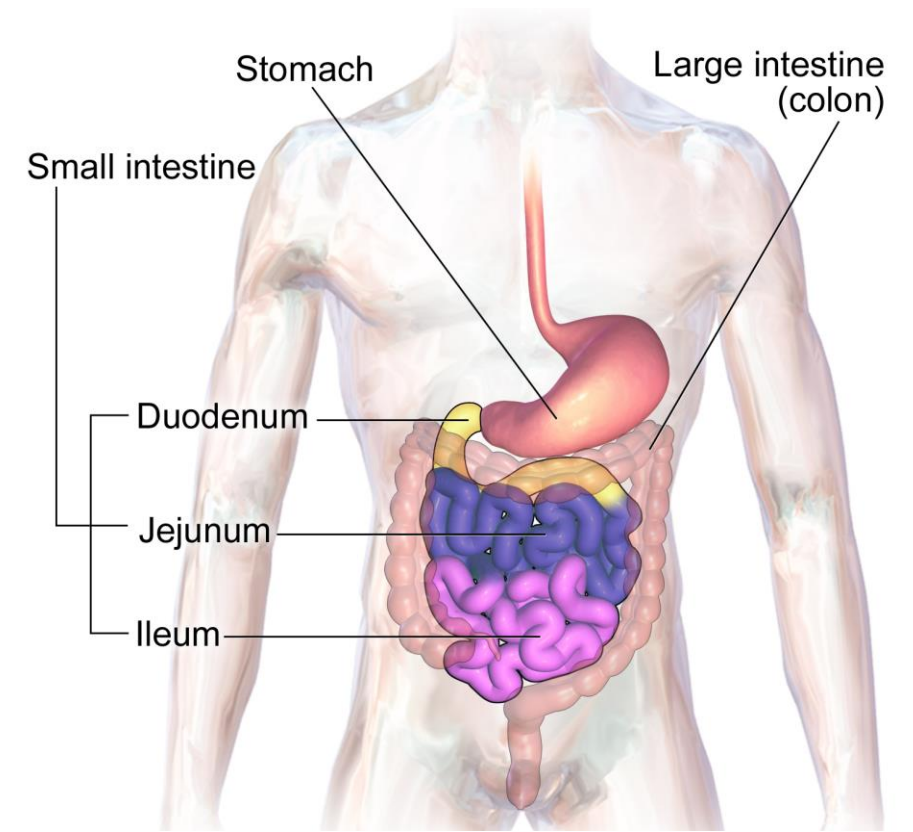
- **Serum vitamin B12 level**
 - Normal: above 300 pg/mL
 - Borderline: 200 to 300 pg/mL
 - Low: less than 200 pg/mL
- Elevated **homocysteine** and **methylmalonic acid**
 - Useful when B12 borderline or low normal
- Treatment: **B12 supplementation**
 - Oral if normal absorption and no/mild symptoms
 - IM for impaired absorption or highly symptomatic
 - Folate may improve anemia but not neuropathy*



Vitamin B12

Absorption

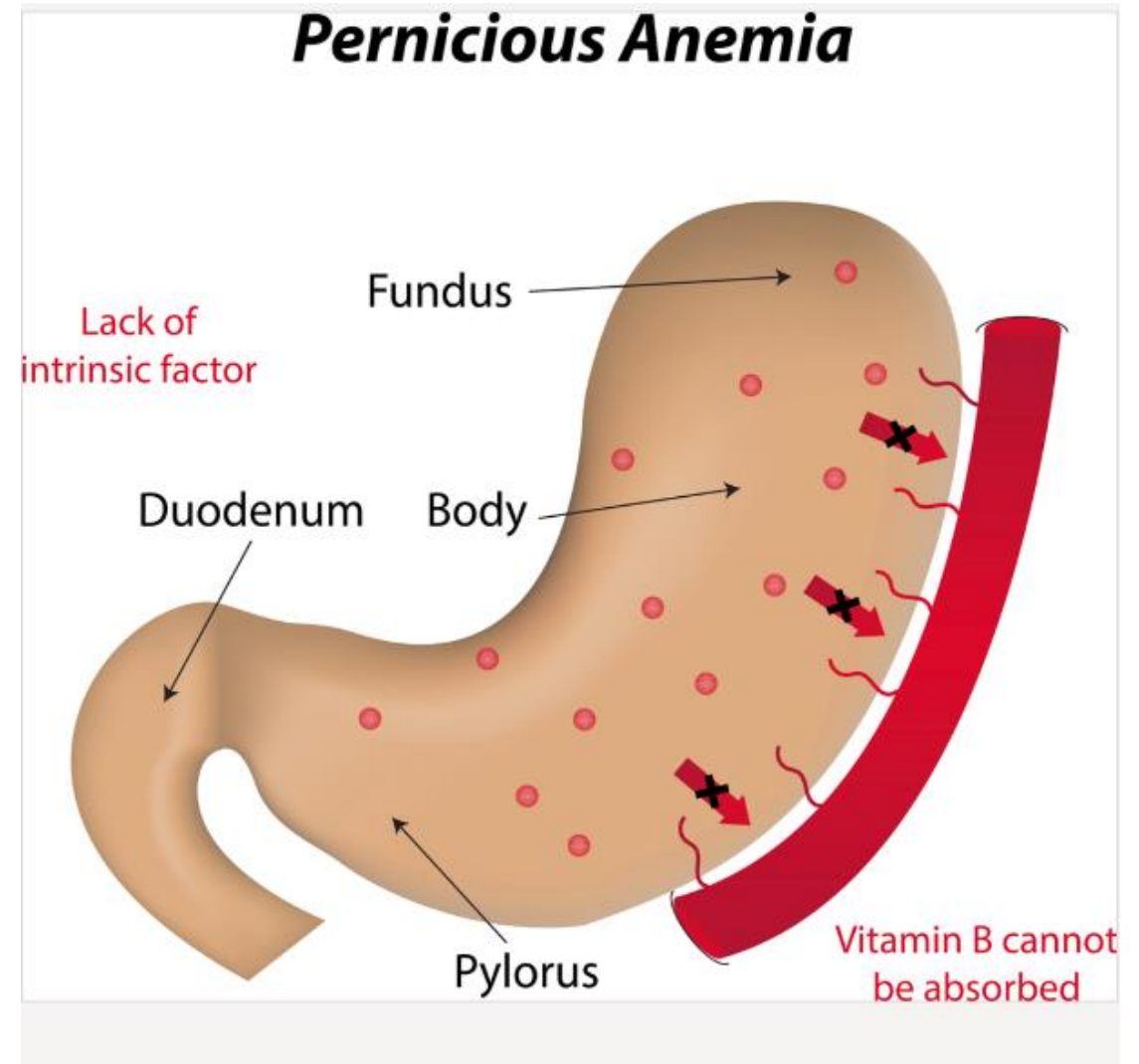
- Protein-bound in foods
- Dissociated by gastric acid and pepsin
- Haptocorrin in saliva bind B12 in stomach
- Gastric parietal cells → intrinsic factor (IF)
- IF binds B12 → absorbed in ileum



Vitamin B12

Deficiency causes

- **Pernicious anemia**
 - Autoimmune condition
 - Antibodies against IF or parietal cells
 - Anti-IF antibodies measured clinically
 - Supports diagnosis but low sensitivity
- **Low dietary intake**
 - B12 stored in liver
 - Requires years to become deficient
 - Occurs with **strict absence of meat**
 - Also lack of supplementation

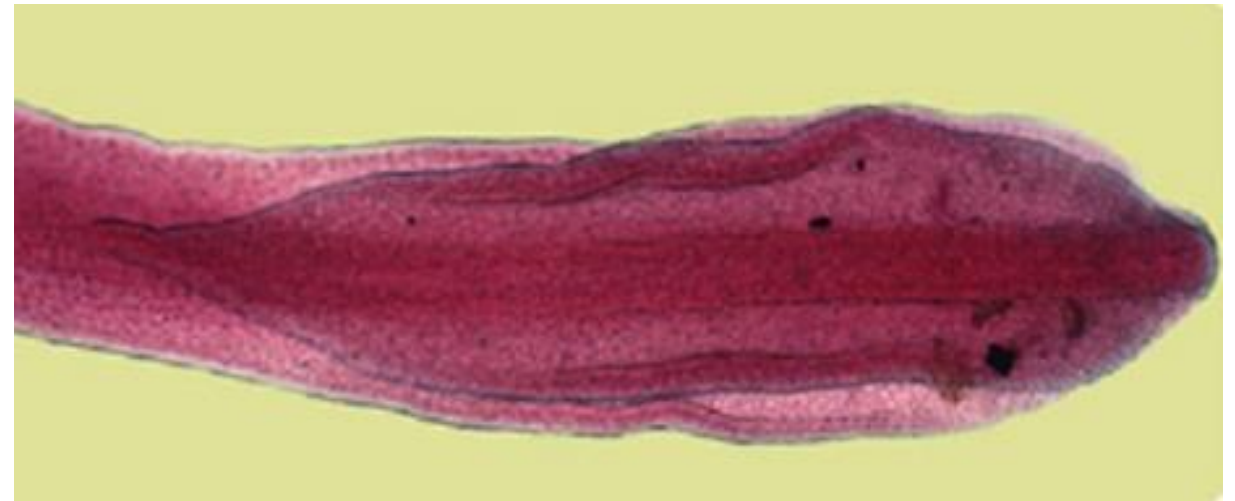


Vitamin B12

Deficiency causes

- Ileum resection/dysfunction
 - Crohn's disease
- Loss of intrinsic factor from stomach
 - Gastric bypass or gastrectomy
- **Diphyllobothrium latum**
 - Helminth (tapeworm)
 - Transmission from eating infected fish
 - Consumes B12
 - Diagnosis: eggs or proglottids in stool

Diphyllobothrium latum

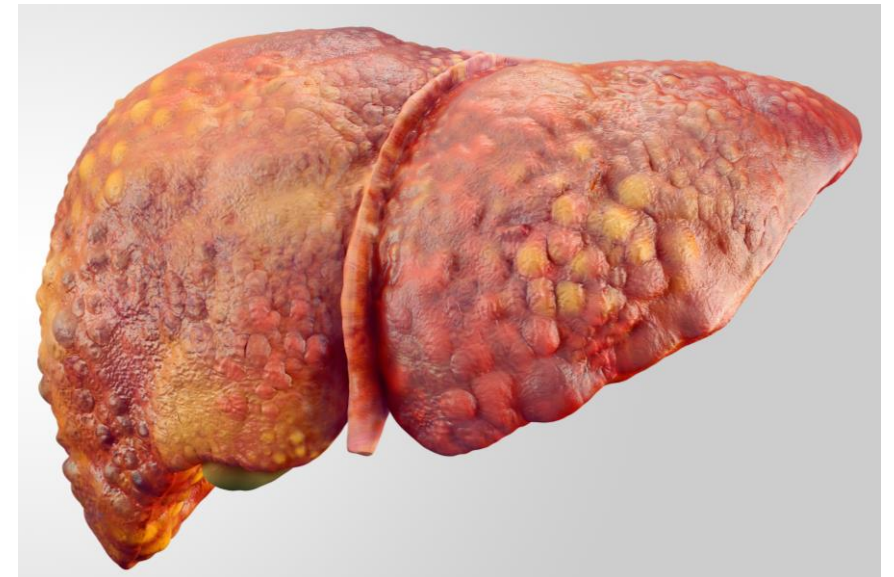


Macrocytosis

Other Causes

- **Liver disease**
 - Exact mechanism not known
- **Heavy alcohol consumption**
 - Common cause of macrocytosis
 - Acetaldehyde may induce membrane changes in RBCs
 - Also causes resistant hypertension
- Hypothyroidism
- Drug-induced (many causes)

Liver Cirrhosis

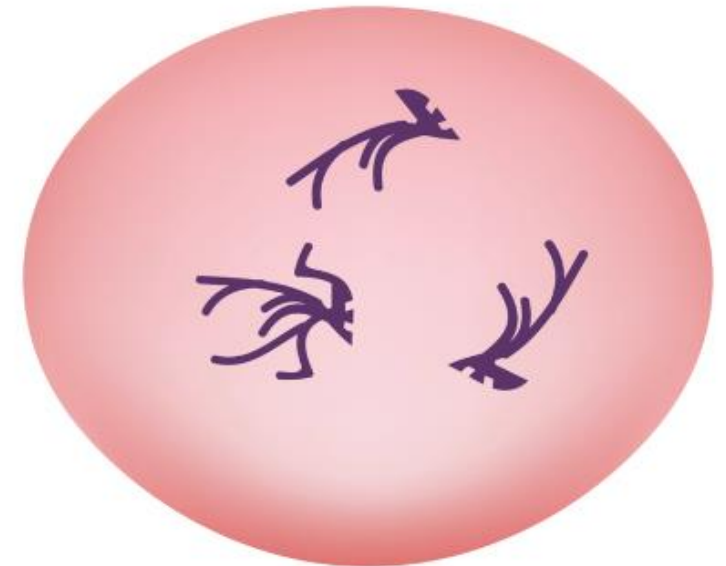


Macrocytosis

Reticulocytosis

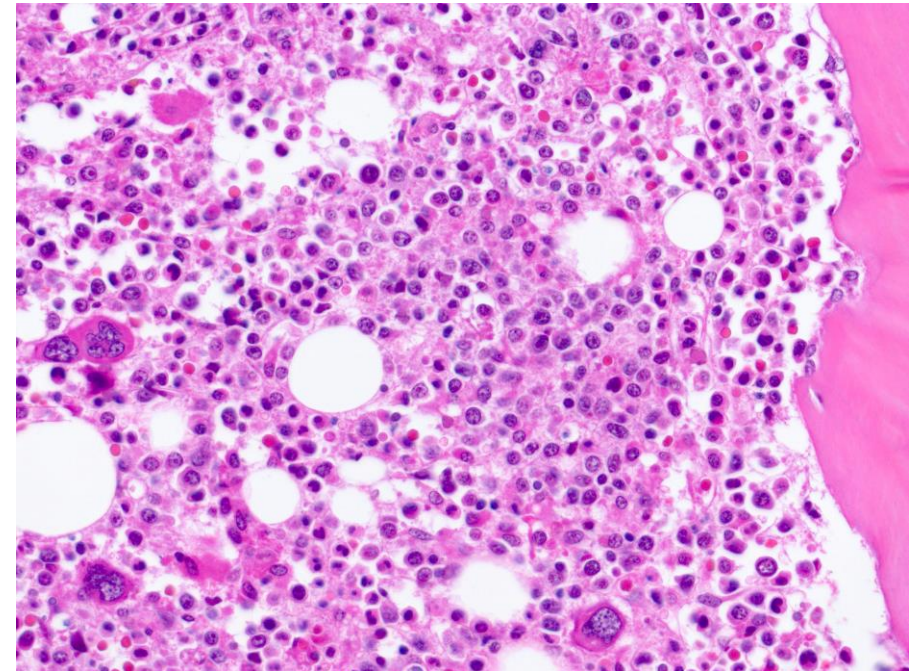
- Reticulocytes have MCV of 103 to 126 fL
- Normal RBCs: 80 to 96 fL
- Need LOTS of reticulocytes to raise average MCV > 100
- Usually raise average MCV but do not reach > 100
- Any MCV > 100 must consider **hemolysis**

Reticulocyte



Myelodysplastic Syndromes

- Abnormal stem cell proliferation
- Causes cytopenias: anemia, neutropenia, thrombocytopenia
- Often progresses to acute myelogenous leukemia
- Can present as **isolated macrocytic anemia**
- MCV may be normal or increased
- Blood testing often normal (B12/folate)
- Diagnosis: bone marrow biopsy



Macrocytosis

Workup

- B12 and folate measurements
- Reticulocyte count
- Peripheral blood smear
 - Abnormal white cells
 - Evidence of hemolysis
 - Hypersegmented neutrophils



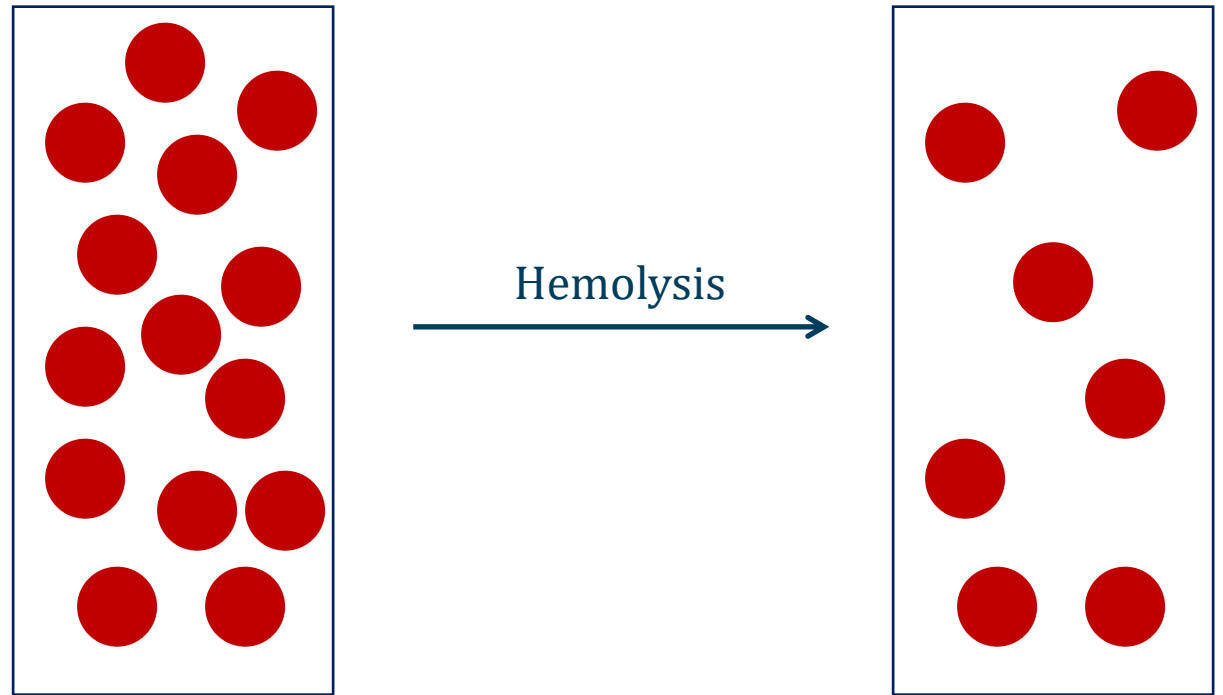
Extrinsic Hemolysis

Jason Ryan, MD, MPH

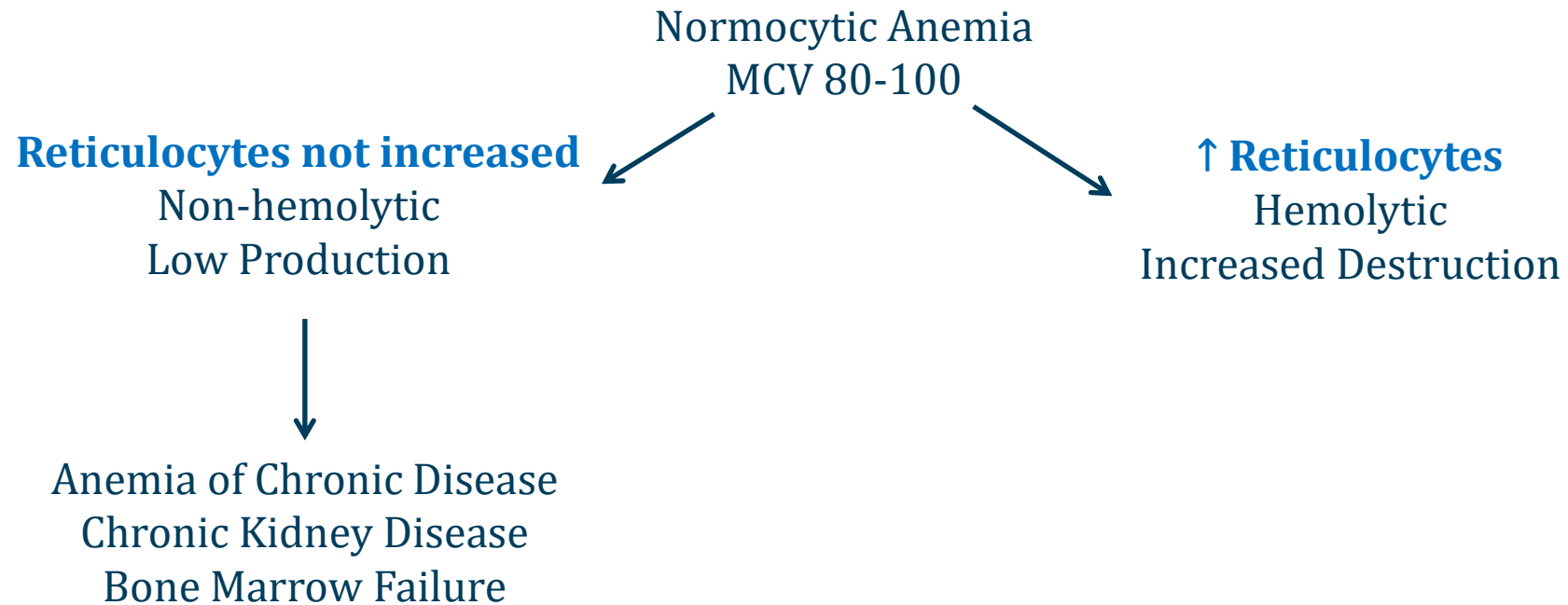


Hemolysis

- Destruction of red blood cells
- Causes a normocytic anemia
- May also cause macrocytic anemia
 - Marked reticulocytosis \rightarrow \uparrow MCV



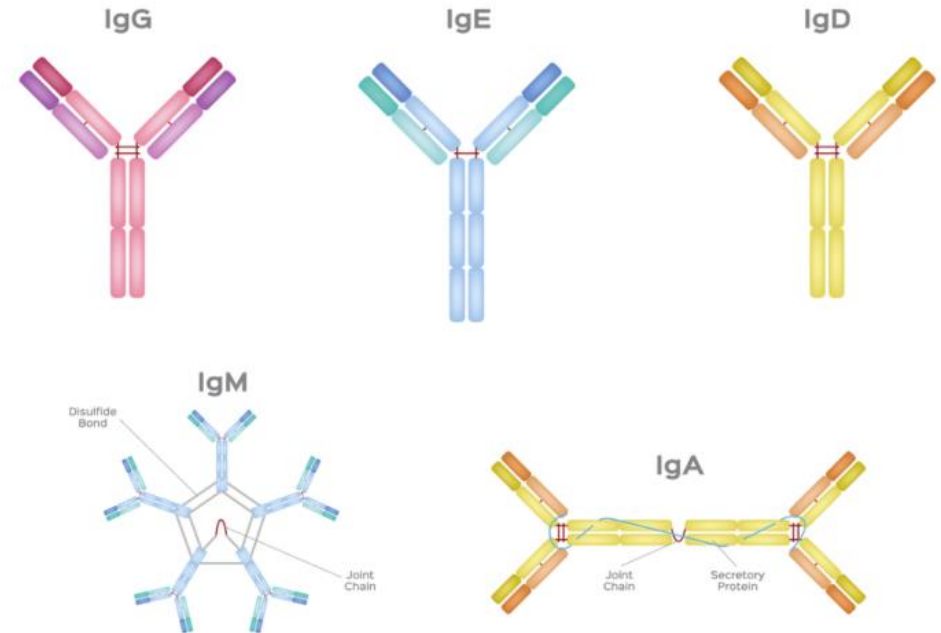
Normocytic Anemias



Hemolysis

Extrinsic versus Intrinsic

- **Extrinsic hemolysis**
 - Cause is extrinsic to the red cell
 - Antibodies
 - Mechanical trauma (narrow vessels)
 - RBC infection
- **Intrinsic hemolysis**
 - Cause is intrinsic to red blood cells
 - Failure of membrane, hemoglobin, or enzymes

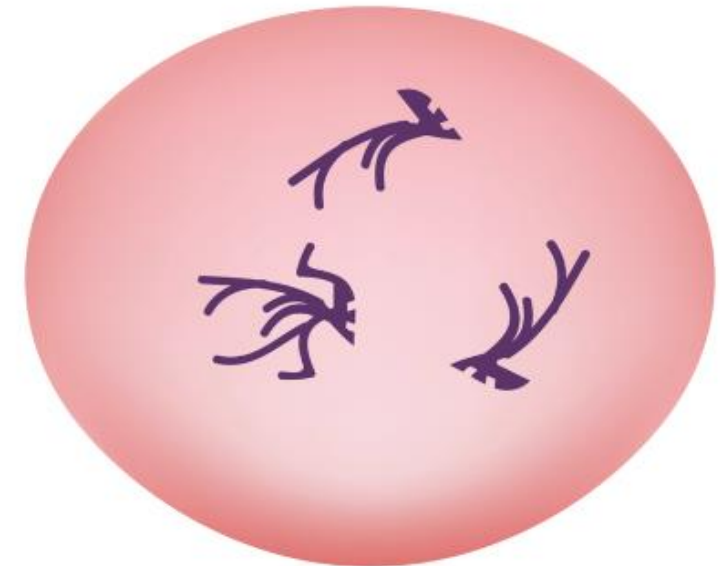


Hemolysis

Clinical features

- Usually normocytic anemia
- **Elevated lactate dehydrogenase (LDH)**
 - Glycolysis enzyme - spills out of RBCs
 - Also elevated in many malignancies (lymphoma, leukemia)
- **Increased reticulocytes**
 - Hallmark of hemolysis
 - Usually about 1-2% of RBCs in peripheral blood

Reticulocyte

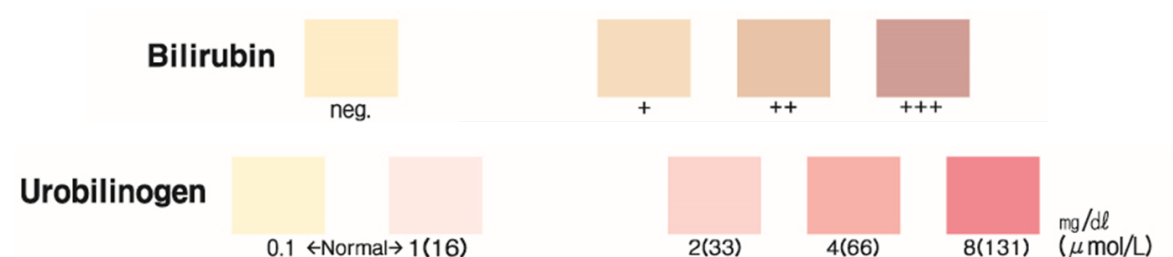


Hemolysis

Clinical features

- **Elevated unconjugated (indirect) bilirubin**
 - Not water soluble
 - Bound to albumin in plasma
 - May cause **jaundice**
 - May cause pigment gallstones
- No bilirubin in urine
 - Unconjugated bilirubin not water soluble
- May see increased urinary urobilinogen
 - Produced by GI bacteria
 - Common in hyperbilirubinemia
 - \uparrow bilirubin excretion \rightarrow \uparrow urobilinogen

Urine Dipstick Testing

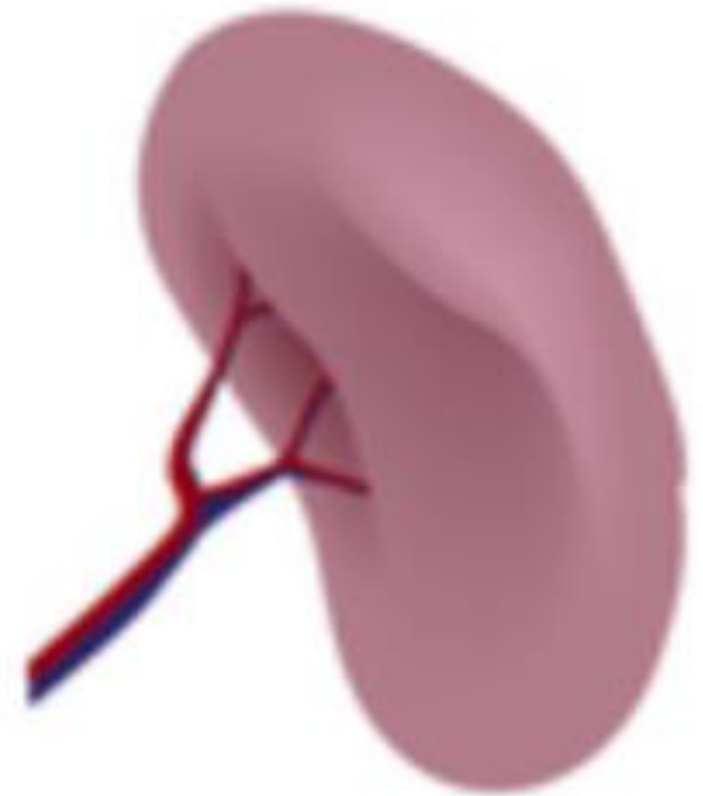


Hemolysis

Intravascular versus Extravascular

- **Intravascular:** occurs inside blood vessels
 - Destruction of RBCs inside blood vessels and outside of spleen
- **Extravascular:** occurs in liver and spleen
 - Liver can remove severely damaged RBCs
 - Spleen destroys poorly deformable RBCs
- Both cause normocytic anemia and \uparrow retic count

Spleen



Intravascular Hemolysis

Unique features

- **Decreased haptoglobin**
- Plasma protein that binds free hemoglobin
- Intravascular: **very low or undetectable**
- Extravascular: low or normal
- Classically taught as low in intravascular only
- Studies show can be low in both types



Intravascular Hemolysis

Unique features - urinary findings

- Haptoglobin saturation → free excess hemoglobin
- “Hemoglobinemia”
- Filtered in kidneys → **hemoglobinuria (dark urine)**
- Some reabsorbed in proximal tubules
- Iron converted into ferritin → **hemosiderin** in tubular cells
- Hemosiderin in urine as cells slough off



Intravascular Hemolysis

Unique features - urinary findings

- **No red cells plus + Hgb**
- Also occurs in rhabdomyolysis
- Myoglobin from muscle damage



Urine test strip

Leukocytes

Nitrite

Urobilinogen

Protein

pH

Hemoglobin

Specific gravity

Ketone

Bilirubin

Glucose

Hemolysis

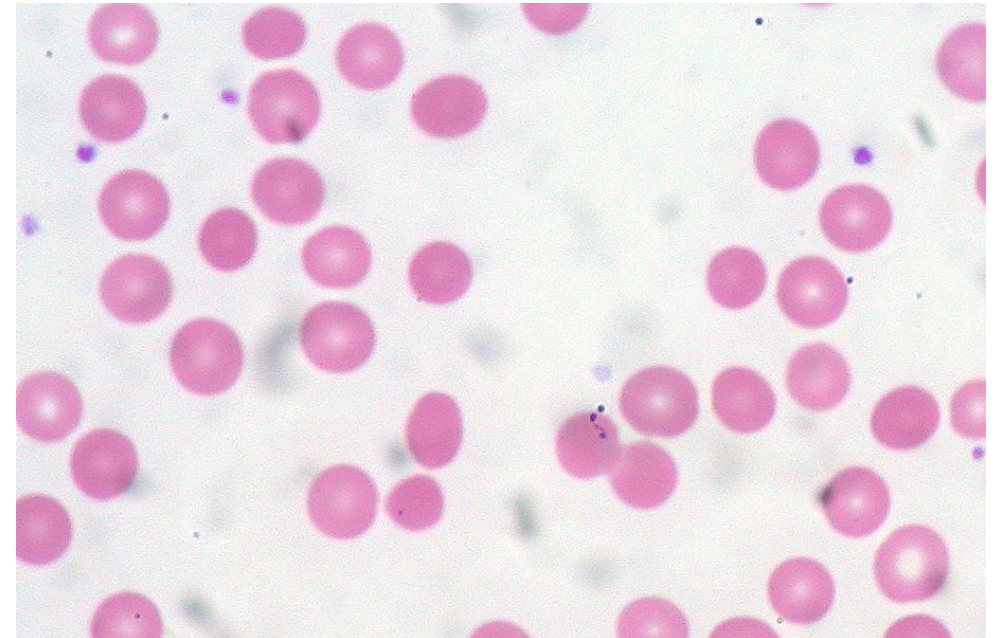
Classic Findings

- Normocytic anemia
- ↑ LDH
- ↑ indirect bilirubin
- **↑ reticulocyte count**
- No urinary bilirubin
- ↑ urinary urobilinogen
- ↓ haptoglobin (lower in intravascular)
- Urine hemoglobin and hemosiderin (intravascular)
- Back, abdominal or flank pain (acute hemolysis)

AIHA

Autoimmune Hemolytic Anemia

- Red cell destruction from **autoantibodies**
- Results in extravascular hemolysis
- Red cell membrane removed in pieces by spleen
- Peripheral smear: **spherocytes**
 - Smaller than normal RBCs
 - Spherical
 - Partial removal RBC membrane
- Can be “warm” or “cold”



Warm AIHA

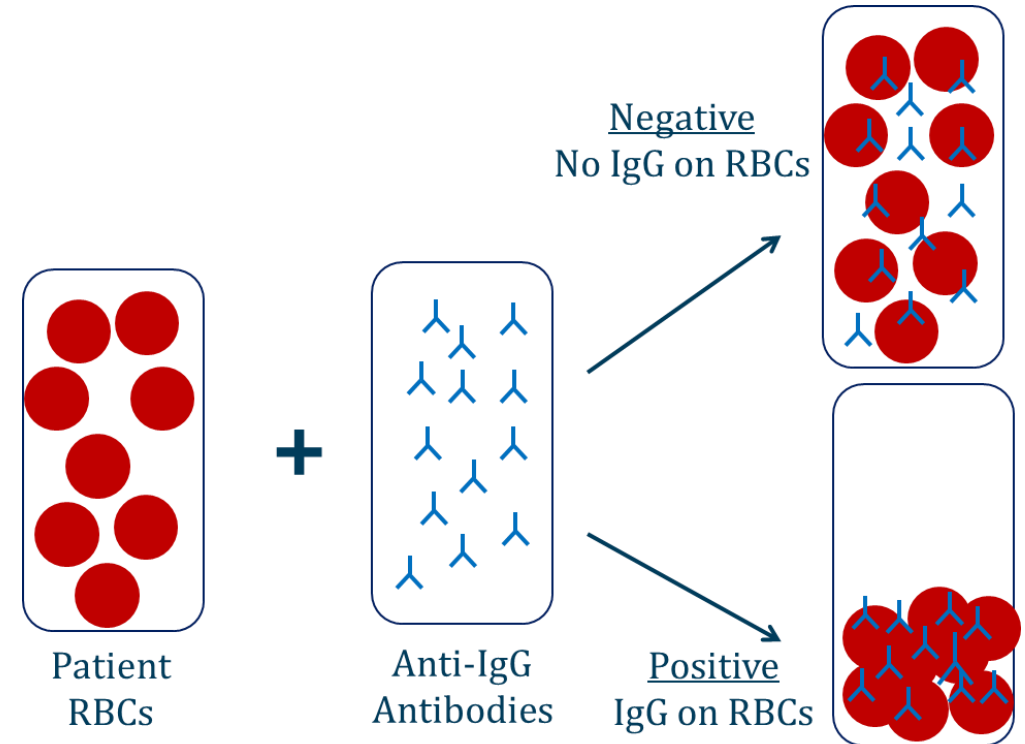
- Most common type of AIHA
- Antibodies bind at **body temp 37°C (“warm”)**
- IgG antibodies against RBC surface antigens
- Clinical features of hemolytic anemia
 - Jaundice
 - Splenomegaly
- Spherocytes



Direct Antiglobulin Test

DAT or Coombs Test

- Identifies red blood cells coated with antibodies or complement
- Warm AIHA: patient RBCs plus **anti IgG antiserum**
- Positive if agglutination occurs
- Indicates patient's **RBCs covered with IgG**



Warm AIHA

Associated Conditions

- Most cases idiopathic
- Associated with many disorders
- Lupus
- Non-Hodgkin lymphoma
- Chronic lymphocytic leukemia (CLL)



SYSTEMIC
LUPUS
ERYTHEMATOSUS

Warm AIHA

Treatment

- First line therapy: **glucocorticoids**
- Second line: rituximab
 - Antibody against B cells
 - Decreases antibody production
- Splenectomy for refractory disease



Cold AIHA

Cold Agglutinin Disease

- Less common type of AIHA
- **IgM antibodies**
- Antibodies bind at $< 30^{\circ}\text{C}$ (“cold”)
 - Occurs in limbs
 - Also fingertips, toes, nose, ears
- Extravascular +/- intravascular hemolysis
 - IgM activates complement
 - Complement-coated red cells cleared by spleen
 - Complement-mediated red cell intravascular lysis
- Spherocytes less common



Cold AIHA

Clinical features

- Symptoms after cold exposure
- Hemolytic anemia
- **Acrocyanosis**
 - Dark purple discoloration
 - Fingertips, toes, nose, and ears

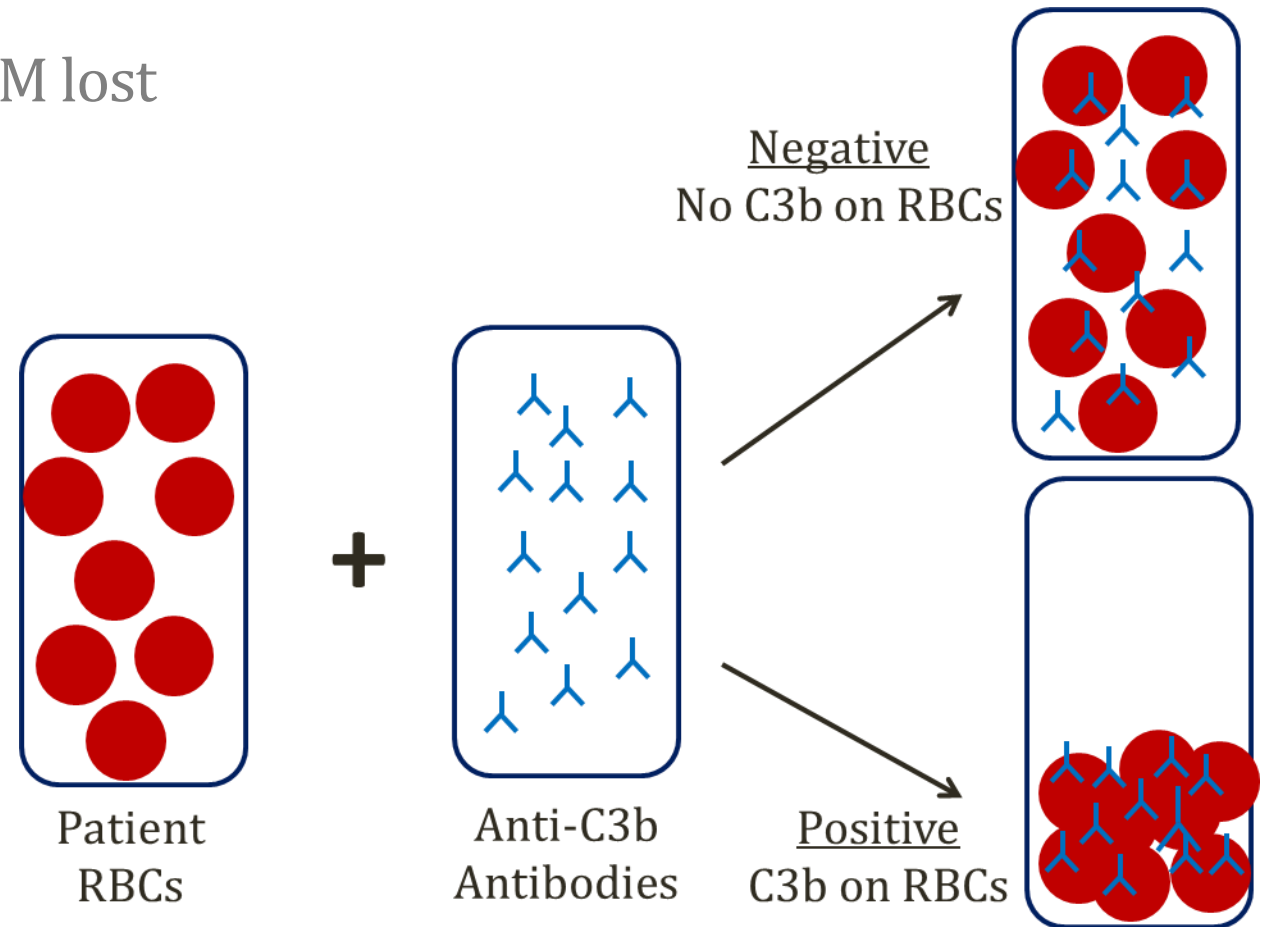
Acrocyanosis



Cold AIHA

Diagnostic testing

- Direct antiglobulin test
- RBCs warmed in central organs → IgM lost
- Leaves C3b bound to red cells
- **DAT positive for C3b**
- **↑ cold agglutinin titer**
 - Autoantibodies to RBCs



Cold AIHA

Associated conditions

- **Lymphoid disorders**
 - Lymphoma
 - Chronic lymphocytic leukemia (CLL)
- **Secondary to infection**
 - Mycoplasma pneumonia
 - Epstein–Barr virus (infectious mononucleosis)

Cold AIHA

Treatment

- Avoid cold (stay warm!)
- Rituximab



Drug-Induced Hemolytic Anemia

- Subtype of immune hemolytic anemia
- Occurs when drug binds to RBC surface
- Induces immune response (hapten)
- Commonly-associated drugs
 - Diclofenac (anti-inflammatory)
 - Piperacillin
 - Ceftriaxone
- Labs indicate hemolysis
- DAT positive
- Treatment: stop offending drug

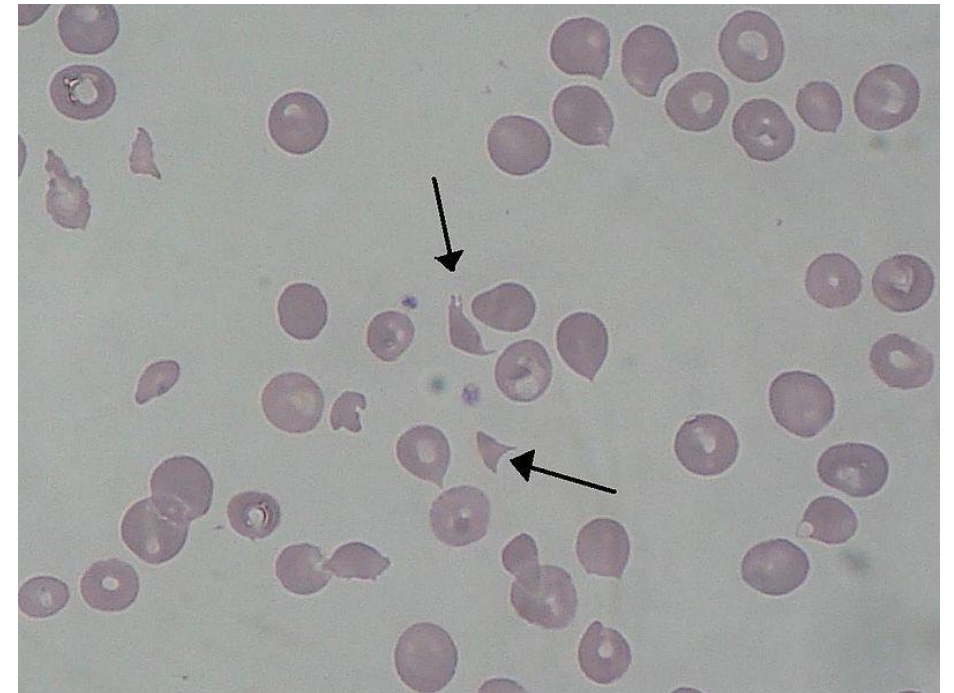


MAHA

Microangiopathic hemolytic anemia

- Non-immune hemolysis
- Shearing of RBCs in small blood vessels
- Thrombi in microvasculature → narrowing
- Blood smear: **schistocytes**
- Thrombotic thrombocytopenic purpura (TTP)
- Hemolytic uremic syndrome (HUS)
- Disseminated intravascular coagulation (DIC)
- Severe hypertension
- Preeclampsia/HELLP

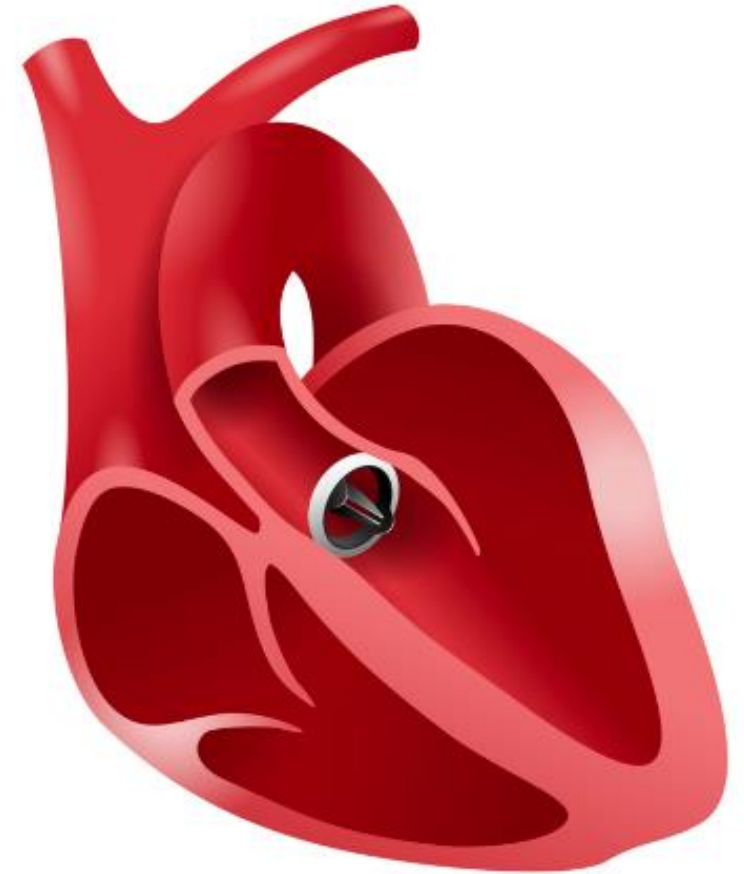
Schistocytes



Mechanical Hemolysis

- Shear forces destroy RBCs in large blood vessels
- Aortic stenosis
- Mechanical heart valves
- Left ventricular assist devices
- Hemolytic anemia may occur
- **Schistocytes** on blood smear
- Similar features to MAHA

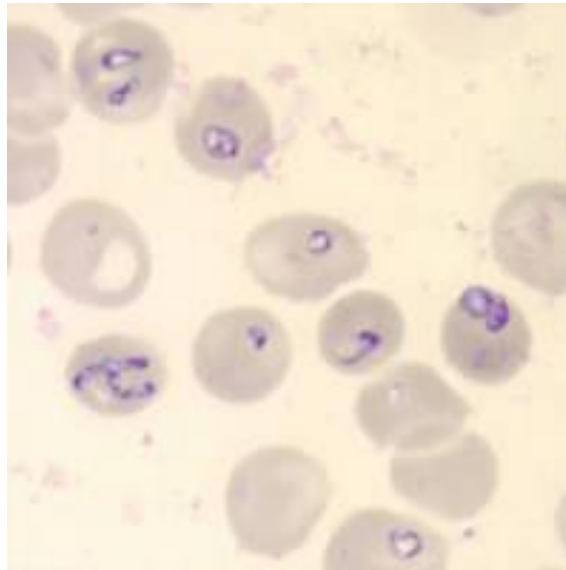
Mechanical Aortic Valve



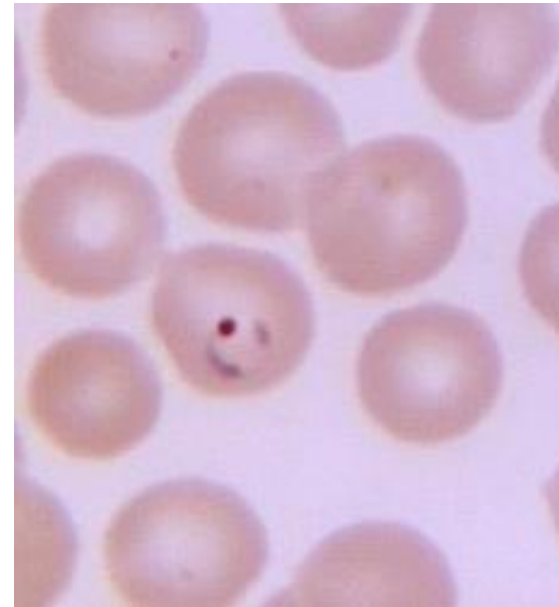
Red Blood Cell Infections

- May cause hemolytic anemia
- Classic infectious agents: malaria and babesia

Babesia
Ring Forms

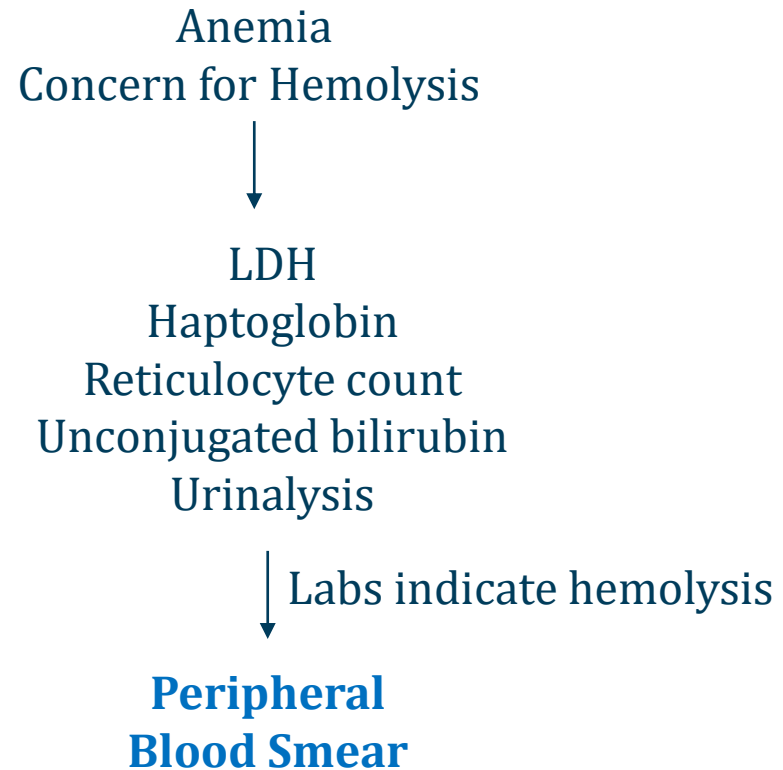


Malaria
Trophozoite Ring



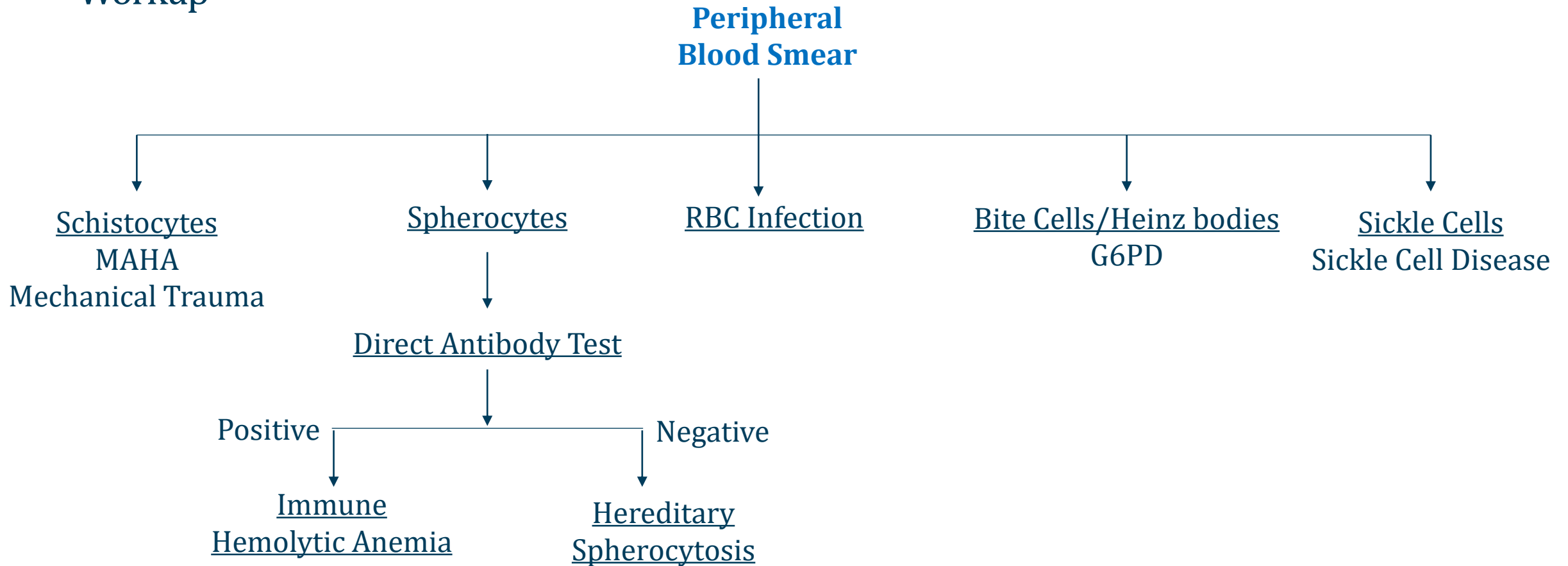
Hemolysis

Workup



Hemolysis

Workup



Intrinsic Hemolysis

Jason Ryan, MD, MPH



Hemolysis

Extrinsic versus Intrinsic

- **Extrinsic hemolysis**
 - Cause is extrinsic to the red cell
 - Antibodies
 - Mechanical trauma
- **Intrinsic hemolysis**
 - Cause is intrinsic to red blood cells
 - Failure of membrane, hemoglobin, or enzymes
 - Membrane: hereditary spherocytosis
 - Enzyme: G6PD deficiency
 - Hemoglobin: sickle cell disease



Paroxysmal Nocturnal Hemoglobinuria

PNH

- Complement-mediated hemolysis
- Loss of protective proteins in RBC membrane
 - Decay Accelerating Factor (DAF/CD55)
 - MAC inhibitory protein (CD59)
- **Acquired** genetic mutation in stem cell
 - Loss of glycosylphosphatidylinositol (GPI) anchor
 - Attaches proteins to cell surface
 - Lead to loss of DAF/CD59 on RBC cell membranes
- Platelets and WBCs may also have lysis



Paroxysmal Nocturnal Hemoglobinuria

Clinical Features

- Classically causes sudden **hemolysis at night**
 - Slowing of respiratory rate with sleep
 - Also shallow breathing
 - Mild \uparrow CO₂ \rightarrow mild respiratory acidosis \rightarrow \uparrow complement activity
 - Red urine on awakening
- **Anemia from hemolysis**
 - Fatigue, dyspnea

Hemoglobinuria



Paroxysmal Nocturnal Hemoglobinuria

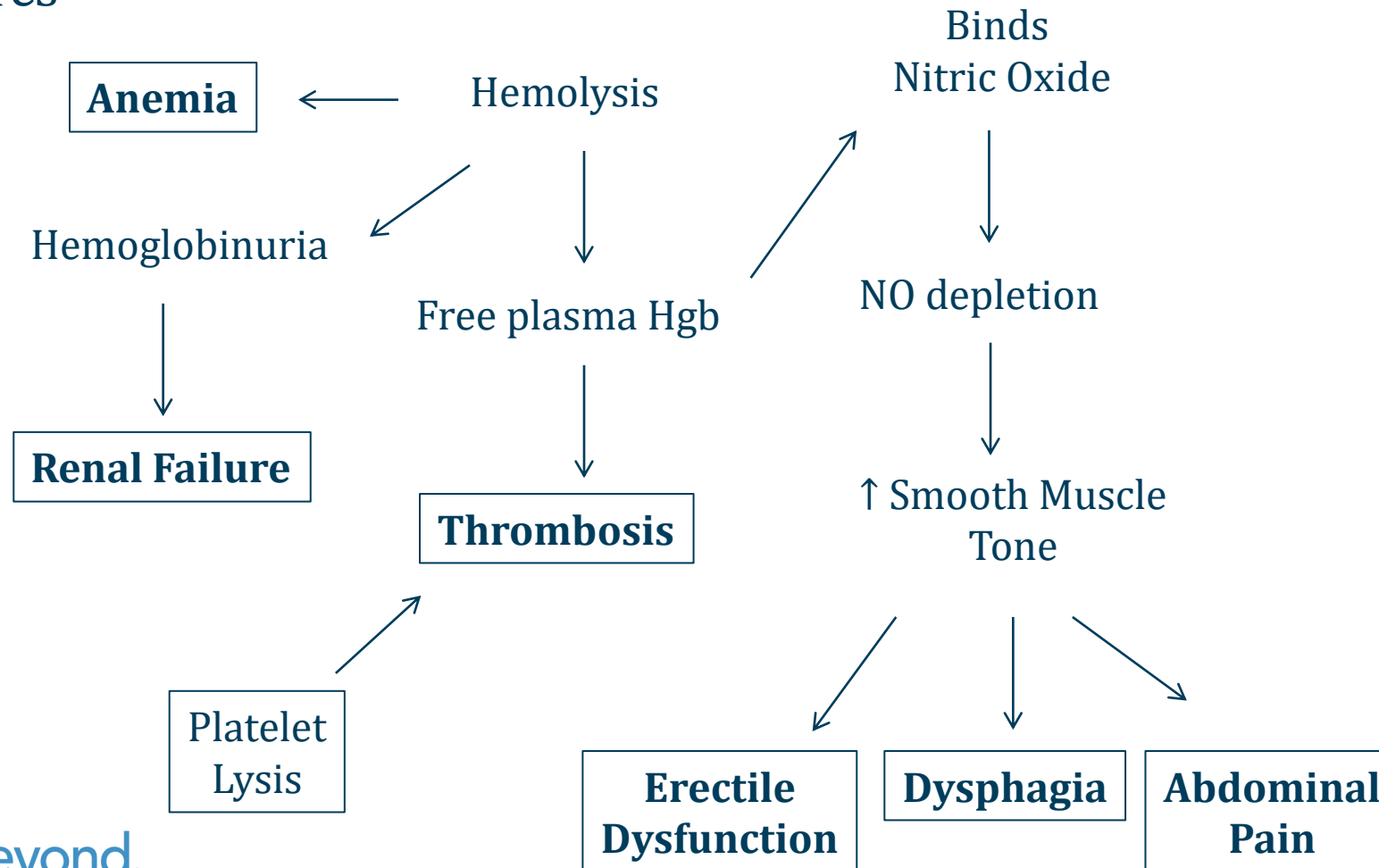
Clinical Features

- **Abdominal pain**
 - Free Hgb binds nitric oxide
 - Smooth muscle tension
- **Thrombosis**
 - Leading cause of death
 - Usually venous clots
 - Unusual locations: portal, mesenteric, cerebral veins
- Hemoglobinuria may lead to **renal failure**



Paroxysmal Nocturnal Hemoglobinuria

Clinical Features



Paroxysmal Nocturnal Hemoglobinuria

Diagnosis

- Suspected with hemolysis and unexplained thrombosis
- Labs may show evidence of hemolysis
- Direct antibody testing will be negative
- **Flow cytometry** confirms diagnosis
 - Red cells will be deficient in GPI-anchored proteins
 - Reduced or absent CD59 and CD55

Paroxysmal Nocturnal Hemoglobinuria

Treatment

- **Ravulizumab and eculizumab**
 - Anti-complement antibody therapies
 - Binds to complement component C5
 - Protects against intravascular hemolysis
 - Results in stable Hgb levels, fewer transfusions
- Hematopoietic stem cell transplantation (HCT)



G6PD Deficiency

Glucose-6-Phosphate Dehydrogenase Deficiency

- Red cell enzyme
- Necessary for generation of NADPH
- NADPH protects RBCs from **oxidative damage**
- **Hemolysis** occurs with **oxidative stressors**
 - Infections
 - Fava beans
 - Drugs
 - Antibiotics (sulfa drugs, dapsone)
 - Anti-malarials (primaquine, quinidine)

G6PD Deficiency

Glucose-6-Phosphate Dehydrogenase Deficiency

- X-linked recessive disorder (males)
- **Recurrent hemolysis** after **exposure to trigger**
 - Red cells become rigid
 - Consumed by splenic macrophages (extravascular)
 - Some lysis in blood vessels (intravascular)

**Exposure
to
Trigger**



Hemolysis



G6PD Deficiency

Classic presentation

- Acute episode of hemolysis
- Jaundice, dark urine, anemia
- May have back pain
- Onset after exposure to trigger



G6PD Deficiency

Peripheral Blood Smear

- **Heinz bodies** and **bite cells**
- Heinz bodies: oxidized Hgb precipitated in RBCs
- Bite cells: membrane removal by splenic macrophages

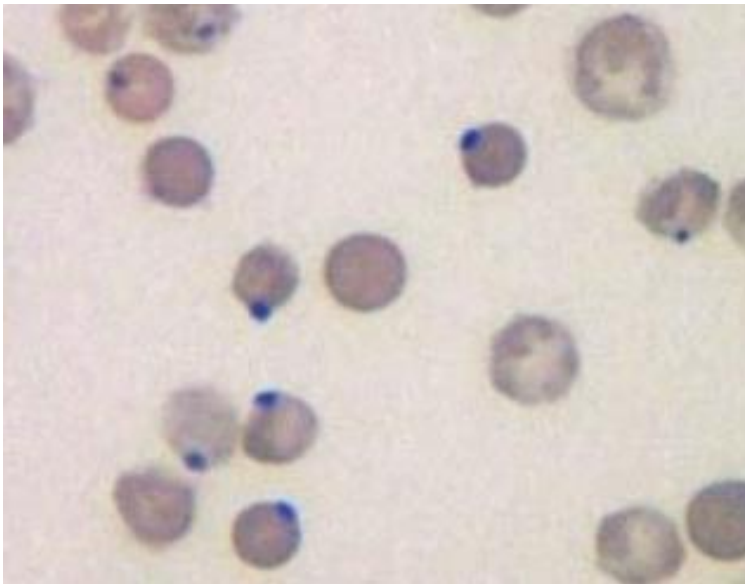


Image courtesy S Bhimji MD



@PathologyDiscussionForum

G6PD Deficiency

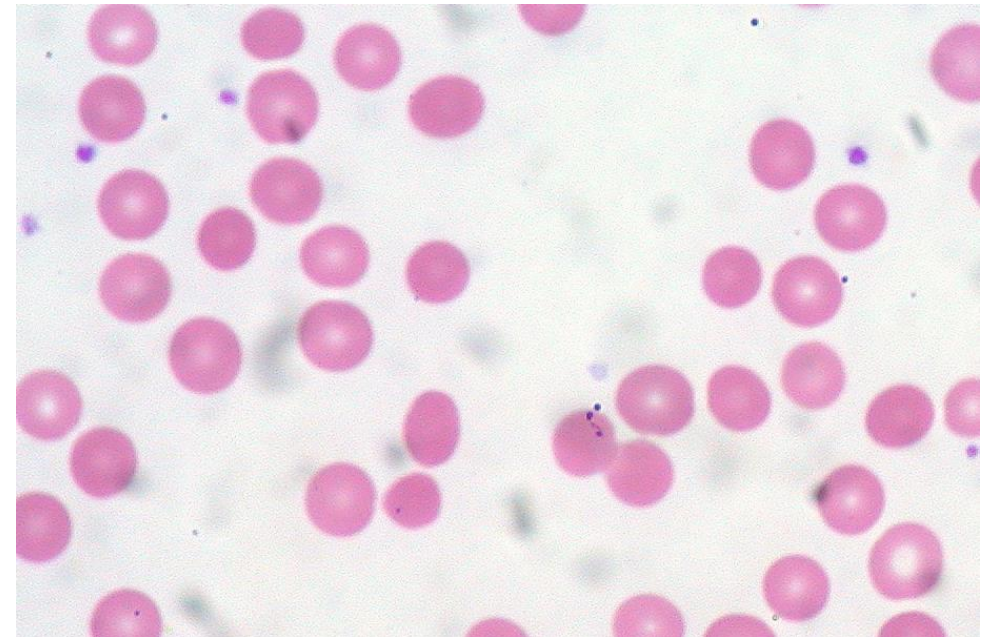
Diagnosis and Treatment

- Screening test: **fluorescent spot test**
 - Detects reduction of NADP → NADPH
 - Add glucose-6-phosphate and NADP to red cells
 - Positive test for G6PD deficiency if blood spot fails to fluoresce under UV light
- Confirmatory test: quantitative rate of **NADPH generation**
- Must test **outside of acute attack**
 - Triggers → destruction of enzyme-poor cells
 - Remaining cells may have normal enzyme levels
- Treatment: avoidance of triggers



Hereditary Spherocytosis

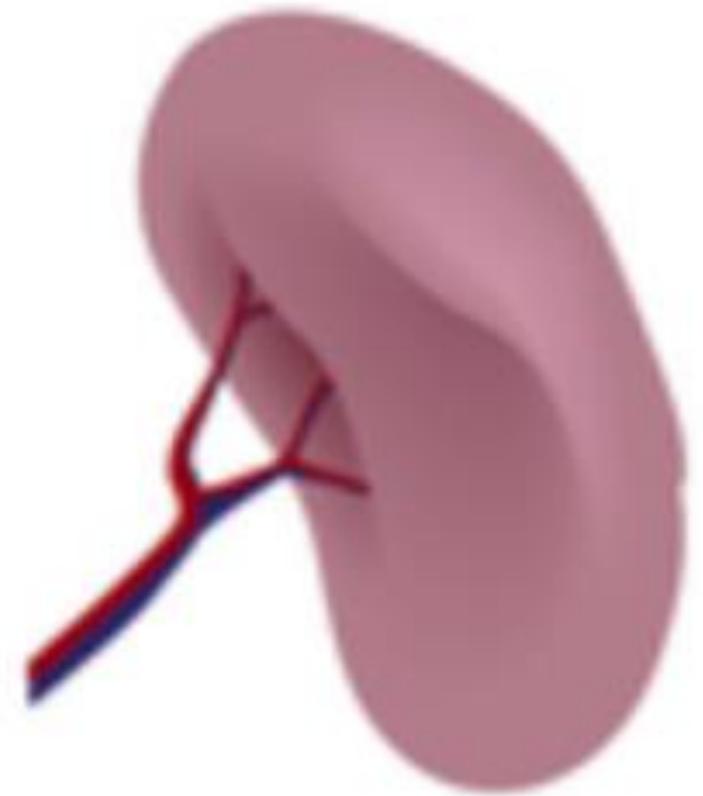
- Autosomal dominant or recessive disorder
- Results in **spherocytes**
 - Slightly smaller than normal RBCs
 - Spherical shape
 - Lacks central pallor
- **Abnormal cytoskeleton membrane proteins**
 - Common involves spectrin
 - Other proteins: ankyrin, band 3, band 4.2



Hereditary Spherocytosis

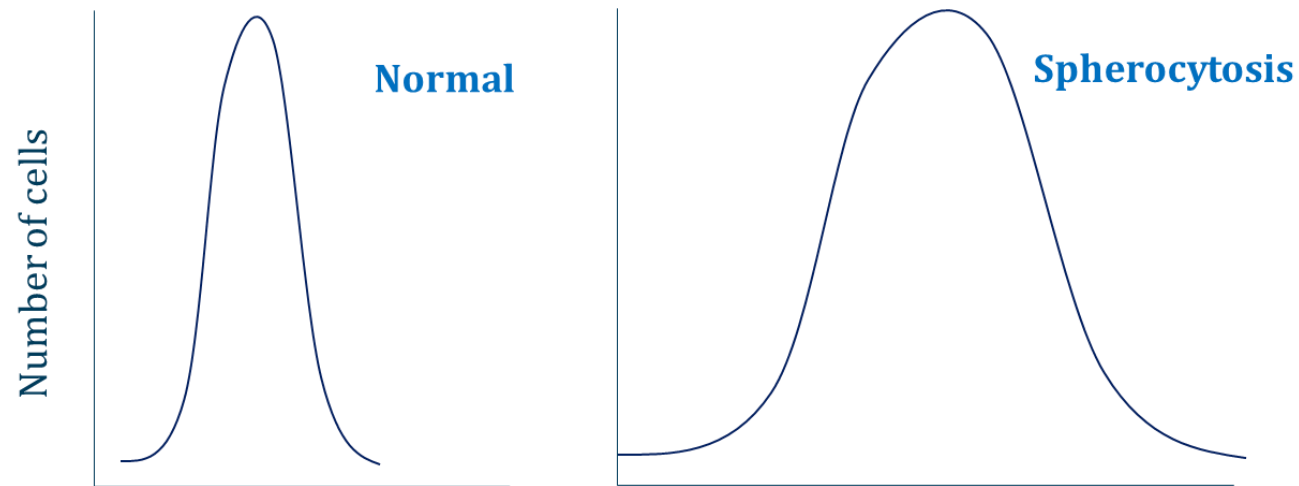
- Oxygen carrying function of spherocytes normal
- Disease from **chronic destruction in spleen**
- Splenomegaly
- Increased bilirubin
- Jaundice
- Bilirubin gallstones
- Can present at any age including newborn period

Spleen



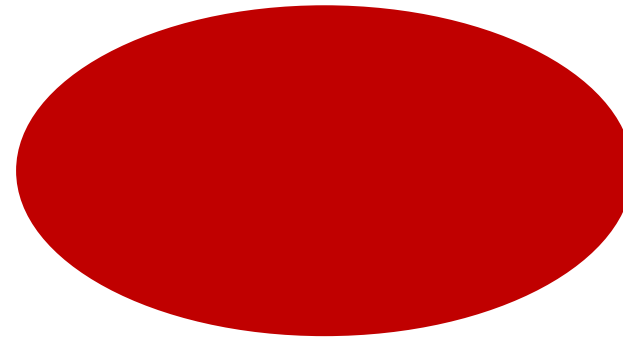
Hereditary Spherocytosis

- Progressive loss of cell membrane
- Over time, more and more membrane lost
- Results in a high RDW
- High RDW also seen in nutritional deficiency (iron, B12, folate)

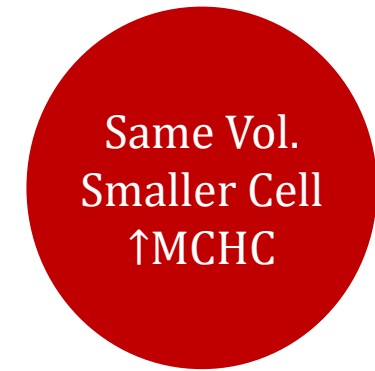


Hereditary Spherocytosis

- Decreased red cell volume
- Results in a **high MCHC**
 - Normal 31 to 36%
 - Levels > 36% common in HS
- MCV usually normal or low
 - Spherocytes: low MCV
 - Reticulocytes: high MCV



Normal



Spherocyte

Hereditary Spherocytosis

- Risk of aplastic crisis with **parvovirus B19 infection**
 - Patients dependent on marrow to replace hemolyzed cells
- Initial presentation may be a **child with infection**
 - Hemolysis compensated until B19 exposure
 - Spherocytosis seen on blood smear



Hereditary Spherocytosis

Diagnosis

- **EMA (eosin-5 maleimide) test**
 - Preferred test
 - Fluorescent dye that binds to membrane proteins
 - Low RBC fluorescence in HS patients
- **Osmotic fragility test**
 - Spherocytes susceptible to osmotic lysis
 - Poor ability to swell like normal RBCs
 - Will lyse in hypotonic solution
 - Measure Hgb release in hypotonic solution
 - Osmotic fragility will be \uparrow if spherocytosis present



Hereditary Spherocytosis

Treatment

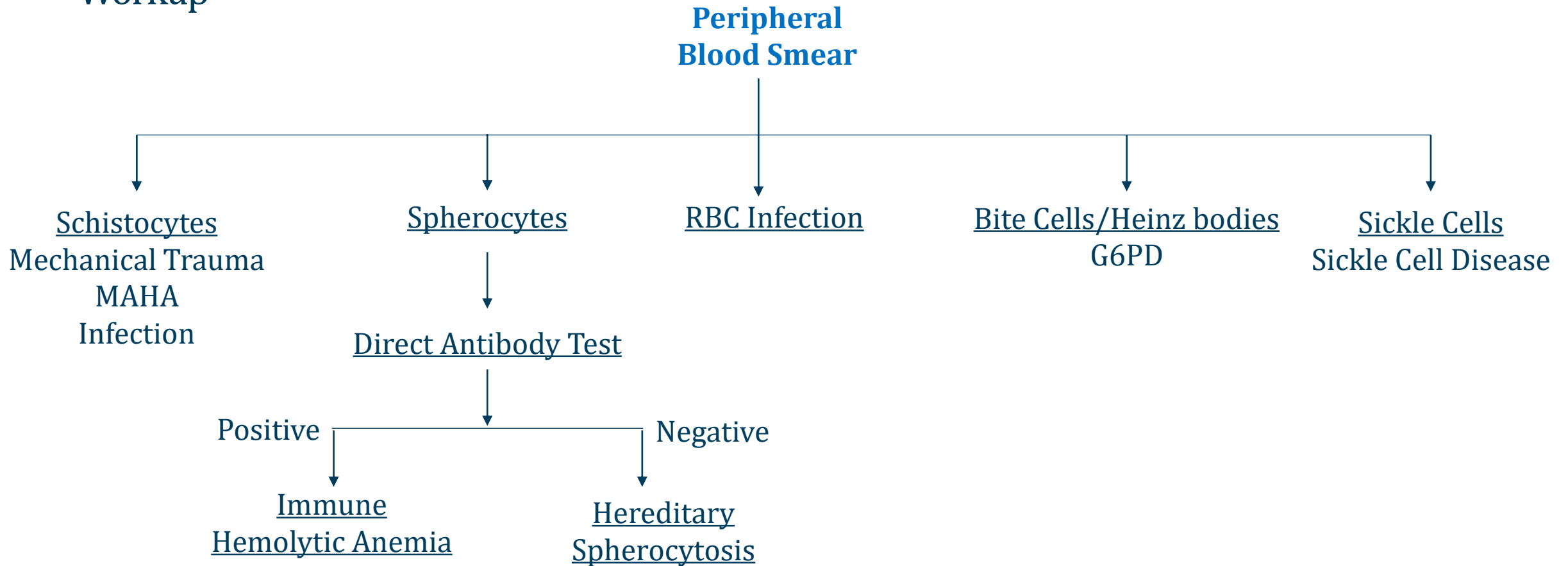
- Transfusions as needed
- Folate supplementation
- Definitive treatment: **splenectomy**
- Spherocytes will persist but hemolysis resolves
- **Howell-Jolly bodies appear**
 - Some RBCs leave marrow with nuclear remnants
 - Normally cleared by spleen
 - Presence in peripheral blood indicates splenic dysfunction

Howell-Jolly Body



Hemolysis

Workup



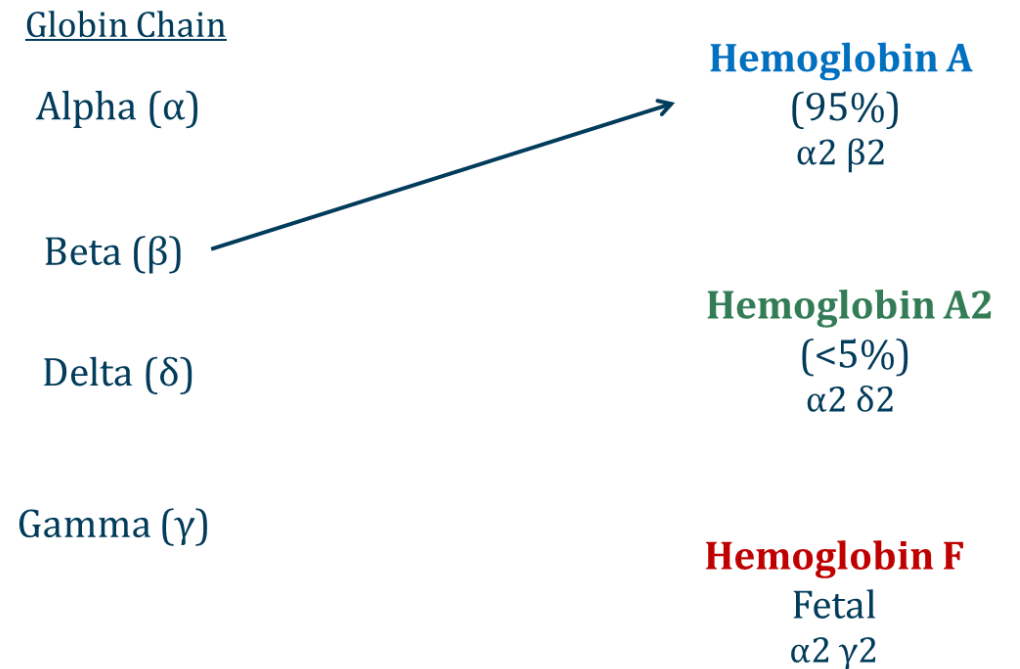
Sickle Cell Disease

Jason Ryan, MD, MPH



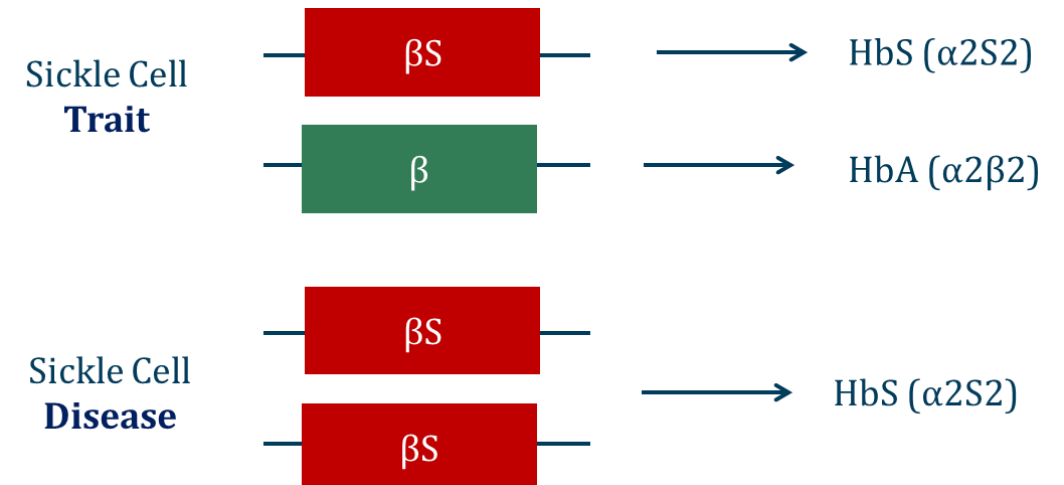
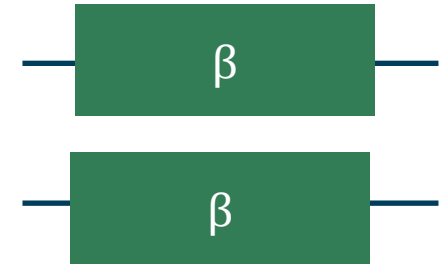
Sickle Cell Disease

- Autosomal recessive disorder
- **Abnormal β hemoglobin chains**
- Beta chains found in hemoglobin A ($\alpha_2 \beta_2$)
- Makes up 95% of adult Hgb



Sickle Cell Disease

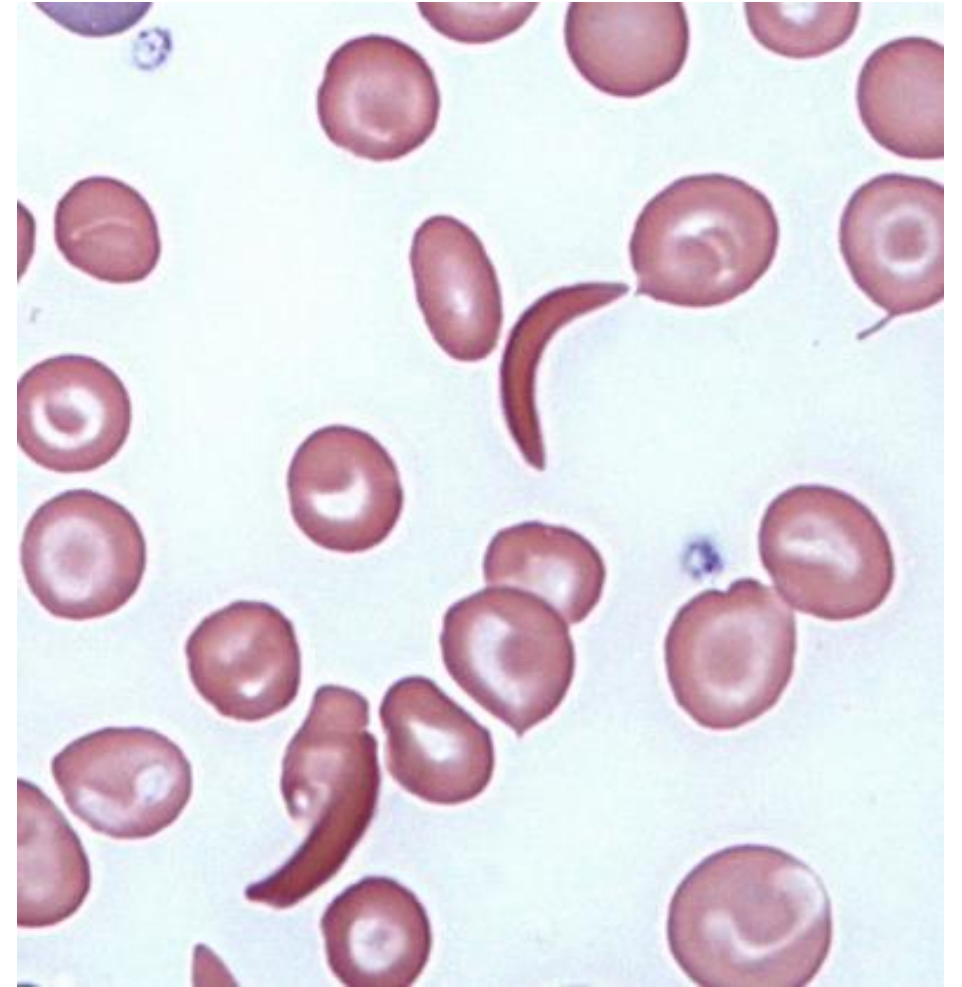
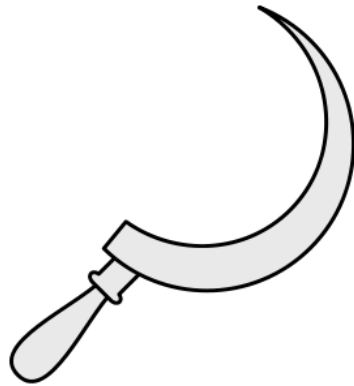
- Single base substitution in β globin gene
 - Adenine changed to thymine
- Produces **HbS** instead of HbA
 - HbA: glutamate
 - HbS: valine
- Altered shape of beta chains in HbS
- One abnormal gene: sickle cell trait (SA)
- Two abnormal genes: sickle cells disease (SS)



Sickle Cell Disease

Pathophysiology

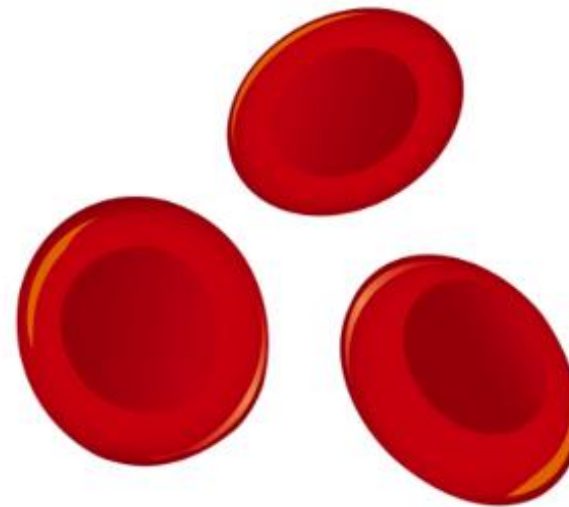
- Deoxygenated HbS is poorly soluble
- **Polymerization** when oxygen low
 - Also in volume depletion, acidosis, infection
- Red blood cells form crescents
- Appearance of a sickle



Sickle Cell Disease

Major clinical manifestations

- **Chronic hemolytic anemia**
- **Vaso-occlusion of small blood vessels**



Normal
Red Blood Cell



Sickled
Red Blood Cell

Sickle Cell Disease

Hemolysis

- Sickling is reversible
- Cells continuously sickle/de-sickle in circulation
- Leads to RBC membrane damage
- **Chronic hemolytic anemia**
 - Anemia, elevated unconjugated bilirubin, pigment gallstones
 - Extravascular and intravascular hemolysis
- Usually no/limited anemia symptoms
 - Increased EPO and adaptation to lower Hgb
- All patients: **folate supplementation**



Sickle Cell Disease

Aplastic crisis

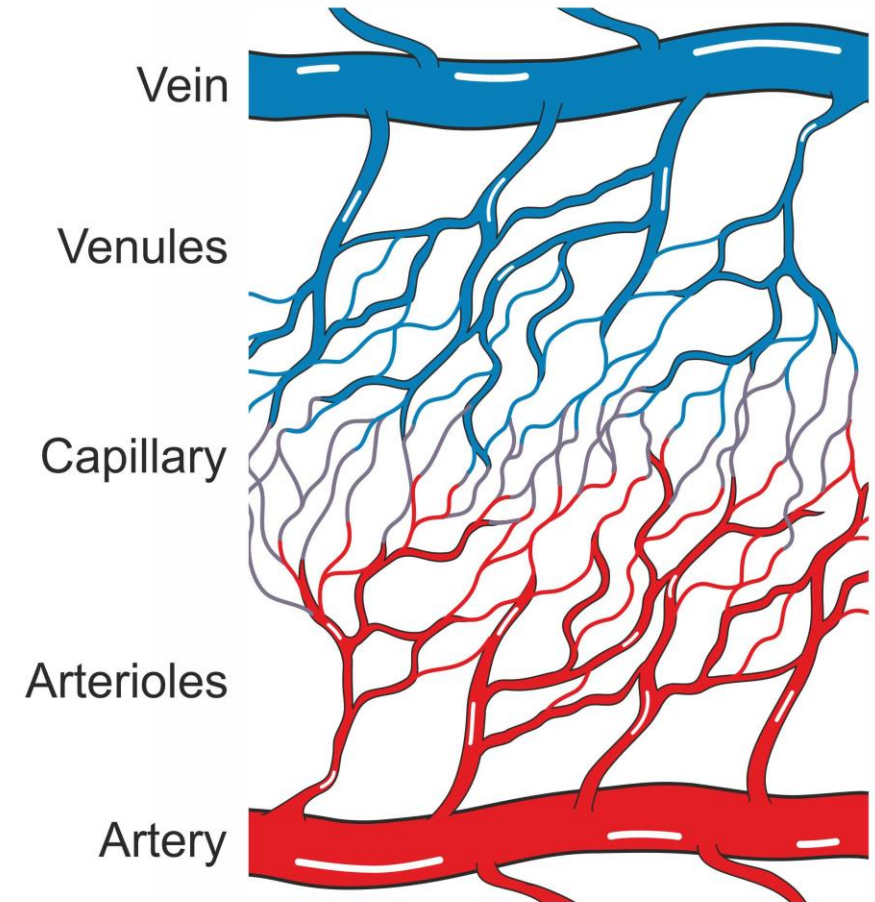
- High reticulocyte count required to maintain Hgb
- Infection may suppress bone marrow function
- Classic cause: parvovirus B19
- Acute hemoglobin decrease
- Fatigue, pallor and dyspnea
- Treatment: **blood transfusion**
- Usually improves after infection



Sickle Cell Disease

Vaso-occlusion

- Sick cells may **occlude microvasculature**
- May affect any organ
- Classic clinical manifestations:
 - Acute pain crises
 - Splenic sequestration
 - Acute chest syndrome
 - Swollen hands (“dactylitis”)
 - Spleen failure → infections
 - Renal dysfunction



Sickle Cell Disease

Pain Crises

- Sudden onset of pain
- Most common type of vaso-occlusive event
- May affect any part of body
- Abdomen, bones, joints, soft tissue, fingers, toes
- Swollen hands and/or feet especially in children
- Can be preceded by a trigger
 - Cold, volume depletion, stress, alcohol, menses
 - Patients can develop strategies to avoid triggers



Sickle Cell Disease

Pain Crises

- Clinical diagnosis
- Often managed at home
- Rarely requires hospital care
- Treatment: **hydration and opioids**
- Oral or intravenous hydration
- Hydroxyurea decreases episodes



Sickle Cell Disease

Dactylitis

- Vaso-occlusion of small bones in hands and feet
- Pain and swelling in hands or feet
- Fingers may look like “sausage” digits
- Common in infants and young children

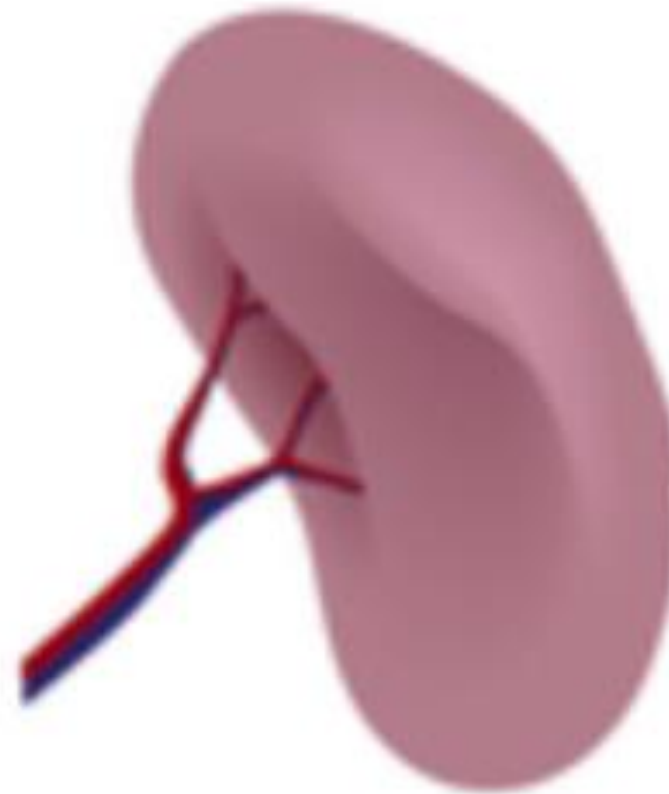


Sickle Cell Disease

Splenic sequestration crisis

- Vaso-occlusion in spleen
- Pooling of red cells
- Marked fall in **hemoglobin level**
- **Rapidly enlarging spleen**
- Left lower quadrant pain
- Risk of **hypovolemic shock**

Spleen

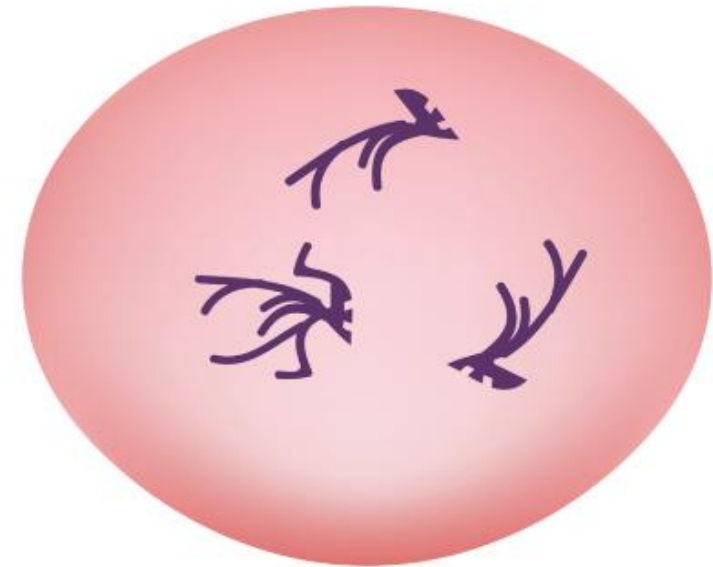


Sickle Cell Disease

Splenic sequestration crisis

- **Common in children**
 - Occurs in spleens yet to develop fibrosis
 - May occur before sickle cell disease is diagnosed
- **High reticulocyte count**
 - Contrast with aplastic crisis
- Acute treatment: **fluids and blood transfusion**
 - Treatment for shock and severe anemia
- Definitive treatment: **splenectomy**
 - May be done after first episode
 - Prevents recurrent episodes

Reticulocyte



Sickle Cell Disease

Splenic failure

- Splenic vaso-occlusion → splenic fibrosis
- Functional hyposplenism in early childhood
- Often before one year of age
- Patients functionally asplenic by four years old
- Increased risk of **infection with encapsulated organisms**
- Especially *S. pneumoniae* and *H. influenzae*

Sickle Cell Disease

Vaccinations

- **Pneumococcal vaccine**
 - Two types: PCV13 and PPSV23
 - Children usually receive PCV13
 - Patient with sickle cell receive both
- **Meningococcal vaccination**
- All other standard vaccines
 - Influenza
 - HIB



Sickle Cell Disease

Chest syndrome

- Leading cause of death in adults with SCD
- Vaso-occlusion of pulmonary microvasculature
- **Clinical syndrome**
 - Fever and chest pain
 - Wheezing and cough
 - Hypoxemia and respiratory distress
 - New pulmonary infiltrate
- Often triggered by infection (pneumonia)
 - Increased sickling in lungs
 - Once begun → inflammation/acidosis → more sickling



Sickle Cell Disease

Chest syndrome

- Chest pain and shortness of breath
- Infiltrate on chest x-ray
- Looks like pneumonia
- Treatment:
 - Fluids and pain medication (similar to pain crisis)
 - Antibiotics, oxygen, bronchodilators
 - Transfusions as needed

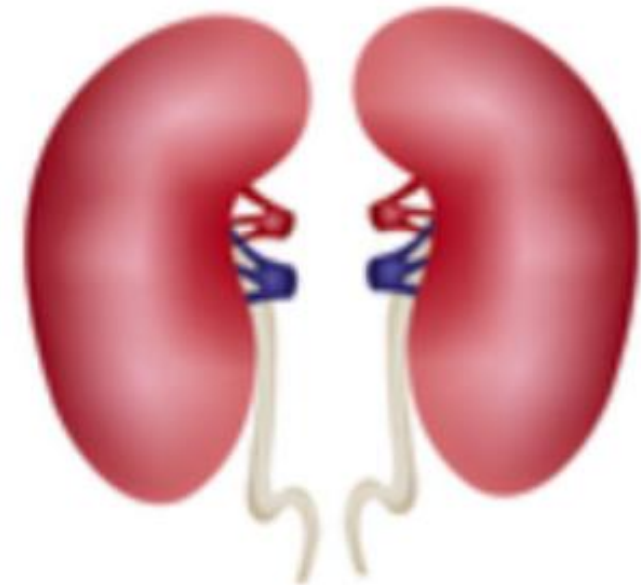
Chest Syndrome



Sickle Cell Disease

Renal dysfunction

- Vaso-occlusion of **vasa recta in renal medulla**
 - Medulla has low oxygen and high osmolality
 - Promotes sickling
- May impair concentrating ability
 - “Hyposthenuria:” dilute urine (lots of water)
 - Cannot raise urine osmolality
 - Causes nocturia and polyuria
- Hematuria
- Increased risk of chronic kidney disease



KIDNEYS

Sickle Cell Disease

Musculoskeletal complications

- **Avascular necrosis**
 - Bone collapse
 - Most commonly femoral or humeral head
- **Osteomyelitis**
 - Infection of infarcted bone
 - Most common Salmonella
 - *S. aureus* most common non-SCD
 - *S. aureus* less common than salmonella

Femoral Avascular Necrosis



Sickle Cell Disease

Stroke

- Common cause of **pediatric stroke**
- Stroke in SCD treated with **exchange transfusions**
 - Remove patient's blood
 - Replace with donor blood (non-sickle cell)
 - Improves blood flow

Ischemic Stroke



Sickle Cell Disease

Other manifestations

- Bilirubin gallstones (hemolysis)
- Priapism
- Retinopathy

Pigment Gallstones



Sickle Cell Disease

Diagnosis

- Prenatal: fetal DNA from chorionic villus sampling
- Newborns: **hemoglobin electrophoresis, HPLC or isoelectric focusing**
- Predominantly HbF (no beta chains)
- Sickle cell disease: small amount HbS and no HbA (FS)
- Sickle cell trait: HbA and HbS (FAS)

Globin Chain

Alpha (α)

Beta (β)

Delta (δ)

Gamma (γ)

Hemoglobin A

(95%)
 $\alpha_2 \beta_2$

Hemoglobin A2

(<5%)
 $\alpha_2 \delta_2$

Hemoglobin F

Fetal
 $\alpha_2 \gamma_2$

Sickle Cell Disease

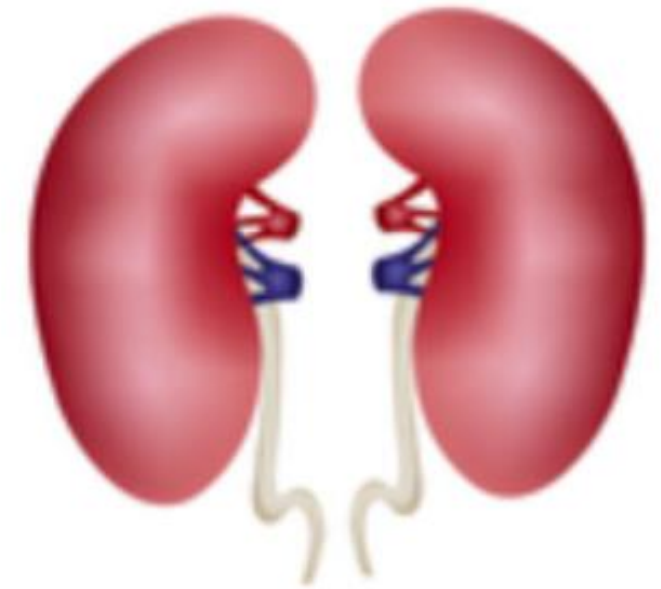
Diagnosis in older children and adults

	Normal (AA)	Sickle Cell Disease (SS)	Sickle Cell Trait (AS)
HbA	97%	--	>50%
HbA2	3%	3%	3%
HbF	<2%	5-15%	<2%
HbS	--	>85%	30-40%

Sickle Cell Trait

Clinical manifestations

- One mutated beta globin gene
- Special complications may occur
- **Kidney dysfunction**
- Renal sickling may cause **hematuria**
- May see loss of concentrating ability



KIDNEYS

Sickle Cell Trait

Clinical manifestations

- **↑ risk of renal medullary carcinoma**
 - Rare, aggressive renal cancer
 - Almost exclusively occurs in sickle cell trait
 - Presents with hematuria and flank pain
- **Splenic infarct at high altitude**
 - Left upper quadrant pain



Sickle Cell Disease

Treatment

- Immunizations
- Folate supplementation
- **Hydroxyurea**
 - Raises amount of HbF
 - Mechanism unclear
 - Limiting side effect: **myelosuppression**
- Transfusions
 - Iron overload may develop
- Bone marrow transplant is curative

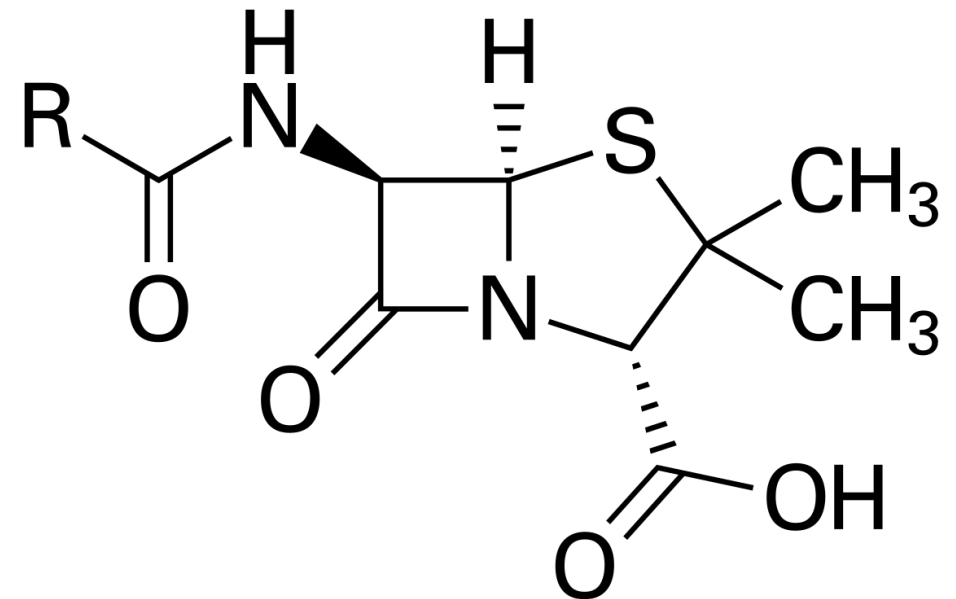


Sickle Cell Disease

Antibiotic prophylaxis

- **Oral penicillin**
- Given to all children at least until age 5
 - Allergy: erythromycin
- Decreases risk of pneumococcal infection
- Decreased risk of death in studies

Penicillin



Sickle Cell-Beta Thalassemia

- Variant sickle cell disorder
 - Sickle mutation combined with another beta globin gene mutation
- One beta gene: sickle cell
- One beta gene: beta thalassemia
- Clinical manifestations similar to sickle cell disease
- Vary depending on beta thalassemia gene function

Hemoglobin C

- Rare mutation of beta gene different from sickle cell disease
- Glutamate replaced by lysine (not valine)
- Hemoglobin C disease (HbC): mild anemia from hemolysis, splenomegaly
- Peripheral smear: **HbC crystals**



Hemoglobin SC Disease

- One HbS gene plus one HbC gene
- More common than homozygous HbC disease
- At risk for same complications as sickle cell disease
- Lower frequency of complications



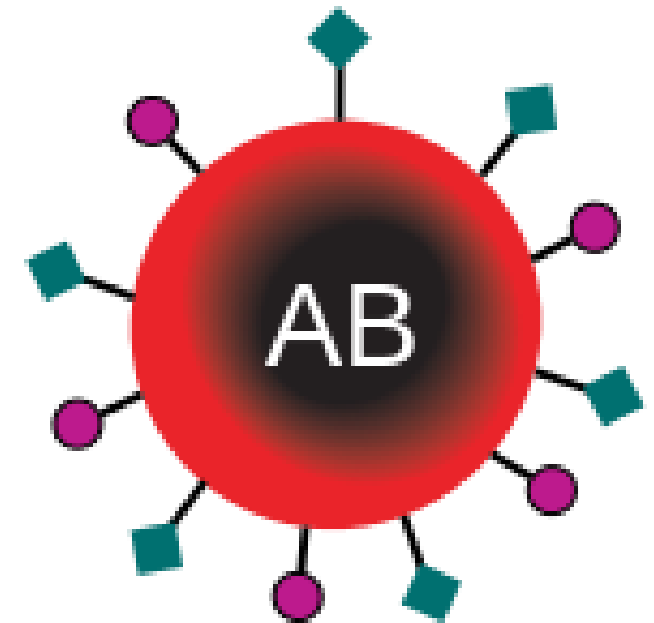
Blood Products

Jason Ryan, MD, MPH



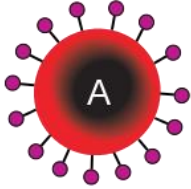
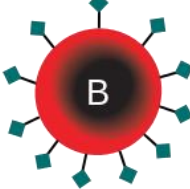
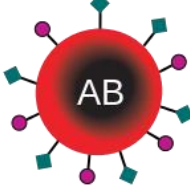



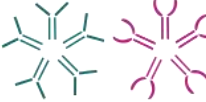



Blood Groups

- Red cells contain surface antigens (proteins)
- Antibodies may form against red cell antigens
- “Blood type” defined by RBC antigens and associated antibodies
- Important for safely administering blood transfusions
- Must match transfusion to blood type
- Two major blood groupings: **ABO system** and **Rh system**



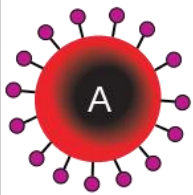
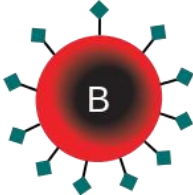
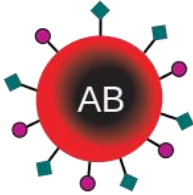
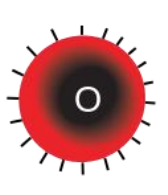


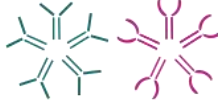



ABO System

- A and B antigens found on RBCs
- Patients who lack A or B generate antibodies
- Must match ABO type to avoid antibody attack of transfused cells

	Group A	Group B	Group AB	Group O
Red blood cell type				
Antibodies in Plasma	 Anti-B	 Anti-A	None	 Anti-A and Anti-B
Antigens in Red Blood Cell	 A antigen	 B antigen	 A and B antigens	None

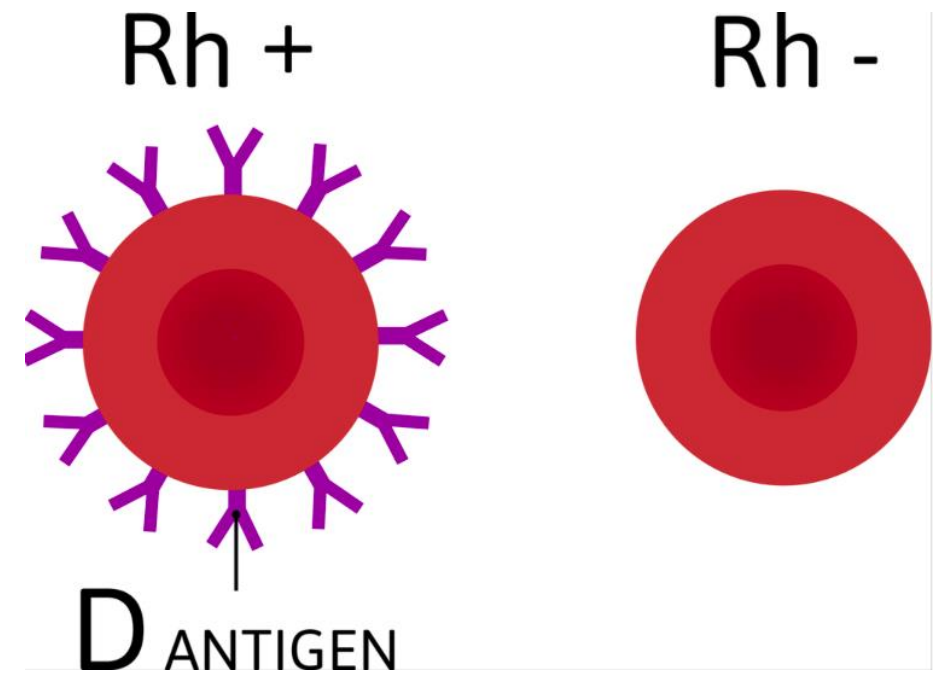
ABO System

- A and B antibodies appear in blood by 4 to 6 months of age
 - Exposure to bacterial antigens with similar structure
 - Occurs as the gut becomes colonized
- Antibodies: mostly IgM with some IgG
 - IgM does not cross placenta
 - Usually do not lead to newborn hemolysis

	Group A	Group B	Group AB	Group O
Red blood cell type				
Antibodies in Plasma	 Anti-B	 Anti-A	None	 Anti-A and Anti-B
Antigens in Red Blood Cell	 A antigen	 B antigen	 A and B antigens	None

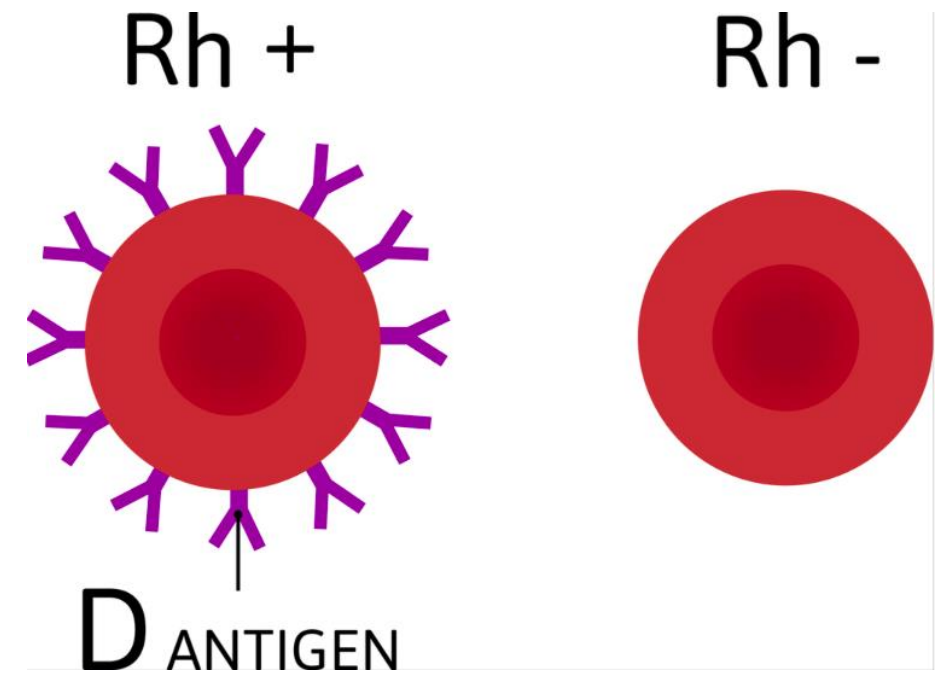
Rh System

- Most important blood group system after ABO
- More than 50 antigens are part of Rh system
 - Named for letters following AB: C, D, E
- Presence/absence of D antigen is critical
 - D antigen highly immunogenic
 - “Rh positive:” has the D antigen (of the Rh system)
 - “Rh negative:” lacks the D antigen (of the Rh system)



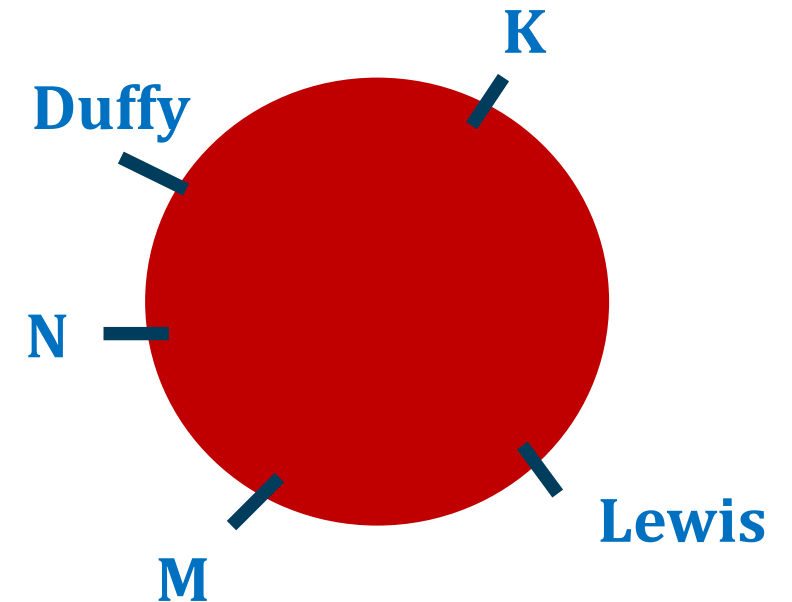
Rh System

- Anti-D antibodies occur with exposure to Rh⁺ cells in Rh⁻ patient
 - Rh⁻ mother with Rh⁺ baby
 - Rh⁻ patient given Rh⁺ blood
- D antigen matched for transfusions
 - Rh⁻ receive Rh⁻ products
- Other Rh antigens not routinely tested
- Anti-D antibodies: IgG
- May cross placenta → newborn hemolytic disease



Other Antigens

- Only ABO and Rh routinely tested
- Only tested when patient has abnormal screening test
- Antibodies from **pregnancy** or **transfusion**



Transfusion Medicine

Common Tests

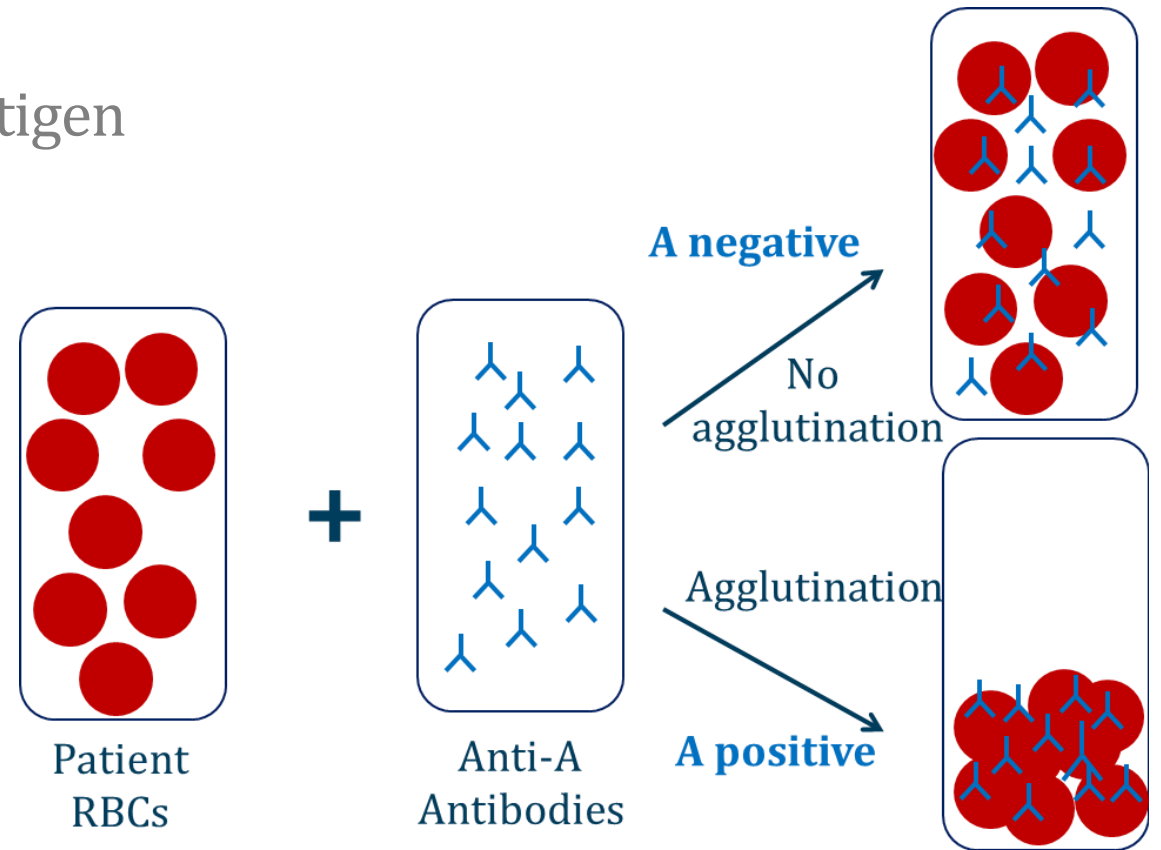
- **Blood type**
 - Determines ABO and Rh status
- **Type and screen**
 - Screening test for minor antibodies
 - Further testing if positive
- **Type and crossmatch (“type and cross”)**
 - Matching of donor blood to patient



Blood Type Testing

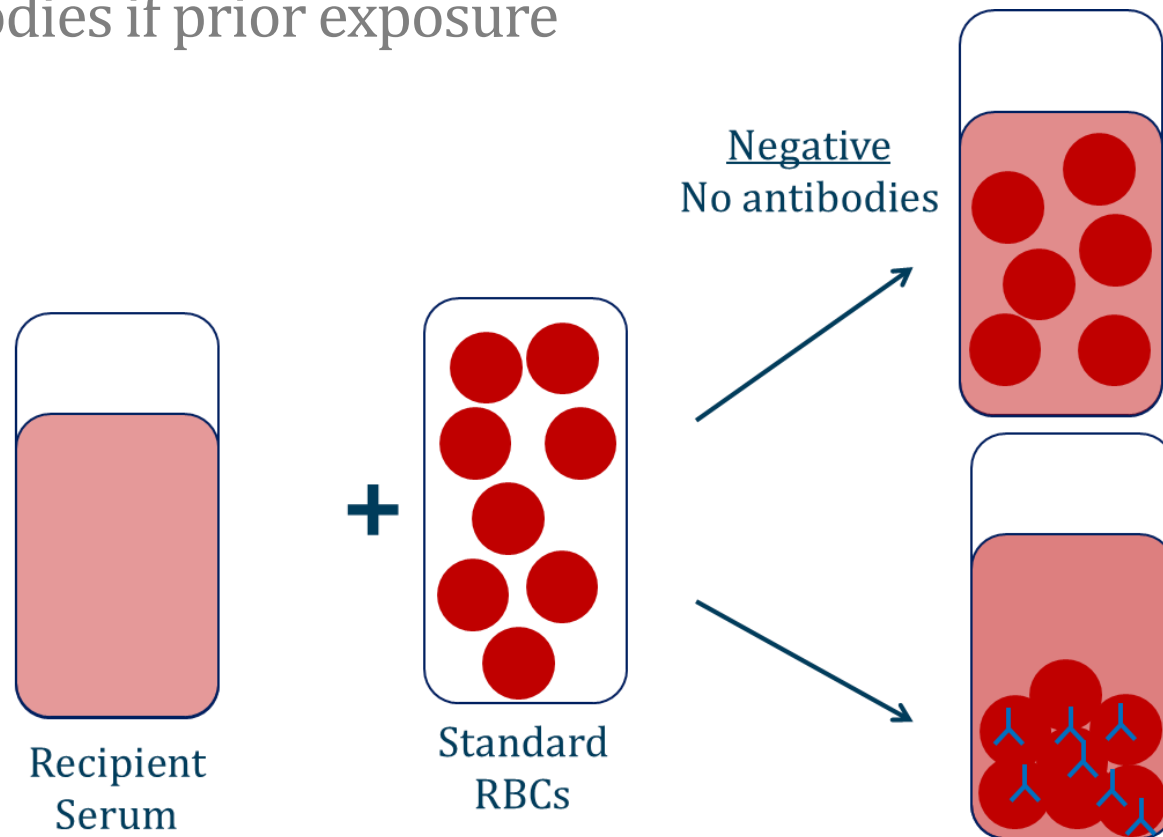
Indirect Antiglobulin (Coombs) Test

- Patient red cells plus antibodies
 - Anti-A; Anti-B; Anti-D
- Agglutination indicates presence of antigen
- Determine ABO type: A, B, AB, O
- Determine Rh type: positive/negative



Type and Screen

- Screen for antibodies to minor antigens (Duffy, Lewis, etc.)
- Will only have antibodies if prior exposure

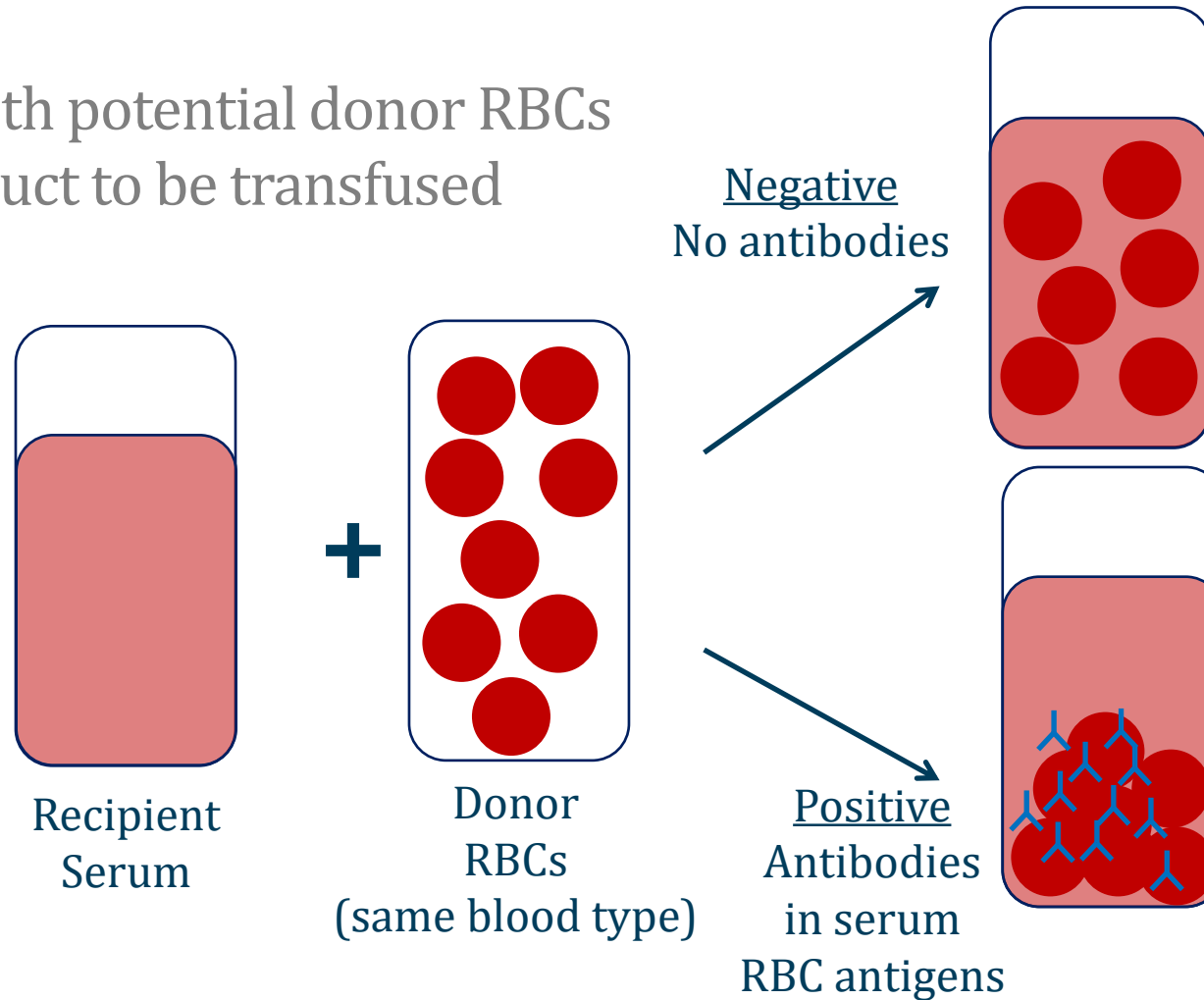


Abnormal Screen

- Determine which antibody is present
- Test patient's serum against large panel of antigens
- Subsequent transfusions: test donor blood for antigen
- Challenging in patients with long transfusion history
 - Sickle cell disease
 - Beta thalassemia major

Type and Cross

- Patient serum with potential donor RBCs
- Final test of product to be transfused



Blood Products

- **Packed RBCs**
 - RBCs with plasma removed
 - Usually administered instead of “whole blood”
 - Minimizes volume given to patient
- **Platelets**
 - Express ABO and some HLA antigens
 - Do not express Rh antigens
 - Reactions from mismatch less common than with RBCs
 - ABO mismatched platelets can be transfused if needed



Red Blood Cells

Other Products

Fresh Frozen Plasma

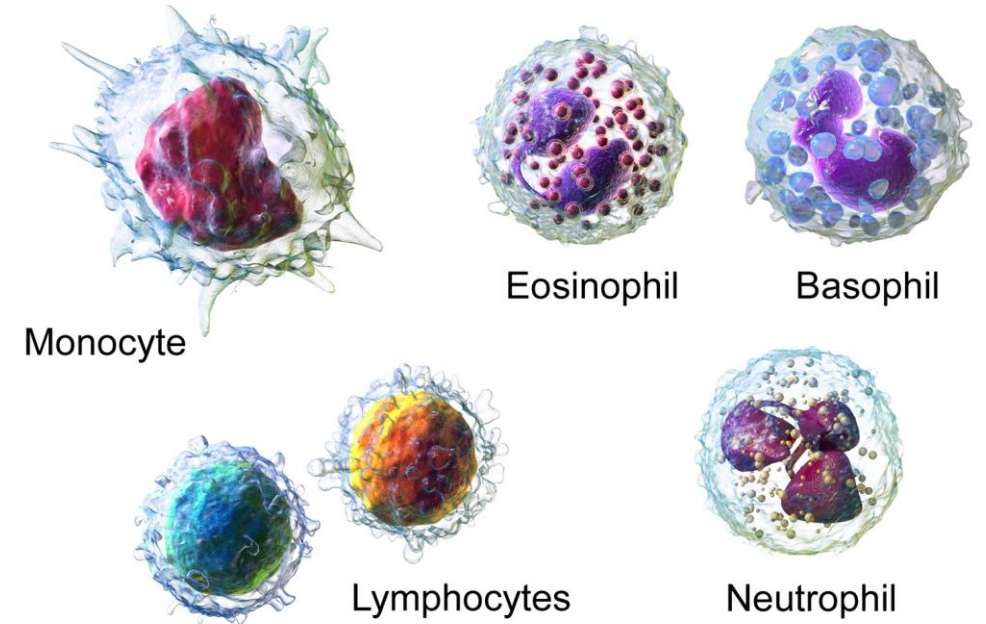
- **Fresh frozen plasma (FFP)**
 - Plasma after removal of RBC, WBC, and platelets
 - Corrects deficiencies of any clotting factor
- **Prothrombin complex concentrate (PCC)**
 - Mixture of vitamin K-dependent factors (II, VII, IX, and X)
 - Lower risk of adverse events compared to FFP
- **Cryoprecipitate**
 - Precipitate that forms when FFP is thawed
 - Contains vWF, factor VIII, factor XIII and **fibrinogen**
 - Massive bleeding or rare ↓ fibrinogen disorders



Blood Product Modifications

Leukoreduction

- Leukocyte removal from red cell products
- Standard practice at most hospitals
- Lowers risk of many transfusion complications
- Febrile non-hemolytic transfusion reactions
- Limits GVHD in transplant patients
 - Decreased production of HLA antibodies
- Virus transmission
 - CMV, HTLV and EBV

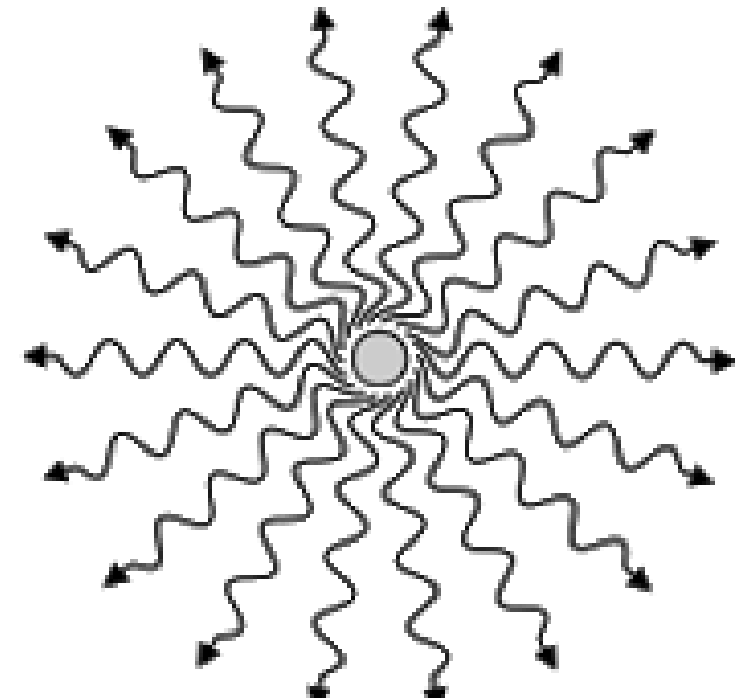


White Blood Cells

Blood Product Modifications

Irradiation

- Inactivates T-lymphocytes in blood products
- Prevents **transfusion-associated graft versus host disease**
 - Donor T-cells not destroyed by host immune system
 - T-cells engraft → attack recipient cells
 - Skin, bone marrow, and gastrointestinal tract
 - May cause pancytopenia
 - Often fatal
- Irradiation used in **immunocompromised patients**
 - Stem cell transplant
 - Immunosuppressive therapies



Blood Product Modifications

Washing of red cells

- Eliminates residual plasma in blood products
- Used in patients with **prior allergic reaction** to transfusion
- Also **severe IgA deficiency**
 - Do not make IgA antibodies
 - Exposure to IgA → anti-IgA antibodies
 - May react to IgA in donor plasma
 - Can lead to anaphylaxis



Transfusion Reactions

- Acute hemolytic reaction
- Delayed hemolytic reaction
- Anaphylaxis/urticaria
- Febrile reaction
- TRALI
- TACO
- Other reactions possible (e.g., sepsis)



AHTR

Acute hemolytic transfusion reaction

- Life-threatening complication of blood transfusion
- Pre-formed antibodies → donor RBCs
- Intravascular hemolysis of transfused RBCs
- Usually from transfusion of **incorrect blood product**
- Fever, chills, flank pain, dark urine
- Jaundice with elevated bilirubin
- Can lead to DIC → oozing from intravenous sites
- **Direct antiglobulin test will be positive**
 - Indicates antibodies attached to red cell surface
 - Key test in evaluating transfusion reactions



AHTR

Acute hemolytic transfusion reaction

- Occurs during transfusion or within 24 hours
- Usual cause is **system or clerical error**
- Numerous safety measures used to prevent:
 - Blood type, antibody screen, cross match
 - Careful patient identification
- Treatment: **stop transfusion** + supportive care
 - **IV fluids** to prevent kidney injury
 - Supplemental oxygen
 - Vasopressors if shock occurs



Delayed Hemolytic Reactions

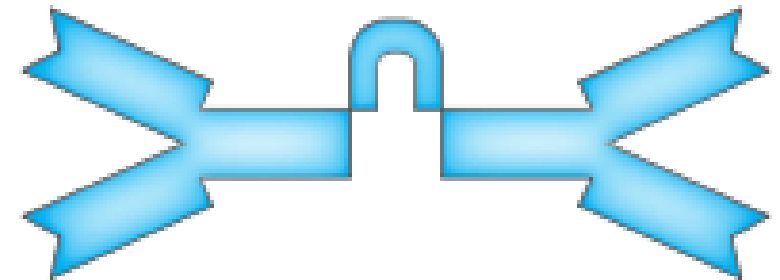
- Occur beyond 24 hours after transfusion
- Caused by antibodies to minor red cell antigens
- Prior exposure → immune memory
- Re-exposure → delayed antibody response
- Leads to fever and low-grade hemolysis
- **Positive direct antiglobulin test**
- Treatment: supportive care



Anaphylaxis

- Occurs seconds to minutes after transfusion begins
- Hives, angioedema, wheezing
- **Hypotension and shock may occur**
- May occur in **severely IgA-deficient individuals**
 - Produce anti-IgA antibodies
 - React with IgA in transfused product
- Also occurs due to plasma proteins in transfused product
- Stop transfusion
- Epinephrine and anti-histamines

IgA Antibody



Urticarial Allergic Reactions

- **Hives only**
- No other features of anaphylaxis
- No wheezing, hypotension or angioedema
- Caused by plasma substances in blood product
- Treatment: stop transfusion + diphenhydramine
 - Assess patient for evidence of anaphylaxis
- **Can continue transfusion after assessment**
 - Provided urticaria resolves and patient stable

Urticaria (hives)



FNHTR

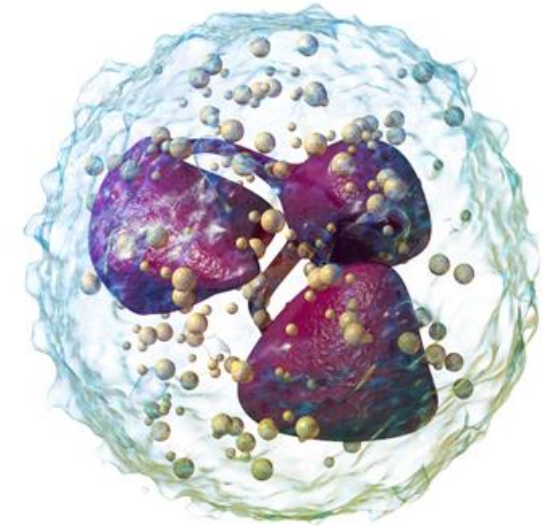
Febrile non-hemolytic transfusion reaction

- Benign reaction
- Fever and chills with no other systemic symptoms
- Normal exam and no evidence of hemolysis on labs
- Caused by **cytokines in blood products**
 - Generated by WBCs during storage
 - Accumulate in stored blood components
 - Usually occur with **non-leukoreduced products**
- Stop transfusion
- Antipyretics (acetaminophen)

TRALI

Transfusion-related acute lung injury

- Respiratory distress due to blood transfusion
- Results from **neutrophil activation** by blood products
 - Some patients predisposed with neutrophils in lungs
 - Neutrophils release cytokines and enzymes
 - Damage the pulmonary capillary endothelium
 - Exudative fluid loss → pulmonary edema



Neutrophil

TRALI

Transfusion-related acute lung injury

- Acute hypoxemic respiratory failure
- Occurs minutes to hours after transfusion start
- Chest x-ray shows pulmonary edema (non-cardiogenic)
- Patients often hypertensive
 - Contrast with anaphylaxis
- Stop transfusion
- Oxygen and respiratory support
 - Resolves with time
 - Does not require antibiotics, diuretics, bronchodilators

Pulmonary Edema



TACO

Transfusion-associated circulatory overload

- Volume overload from blood transfusion
- Acute hypoxemic respiratory failure after transfusion
- Chest x-ray shows pulmonary edema
- Evidence of **heart failure**: ↑ JVP, S3, ↑ BNP
- Stop transfusion
- Oxygen and ventilator support
- **Diuresis**

TRALI	TACO
Fever Normal JVP Low BNP	Elevated JVP Third heart sound High BNP

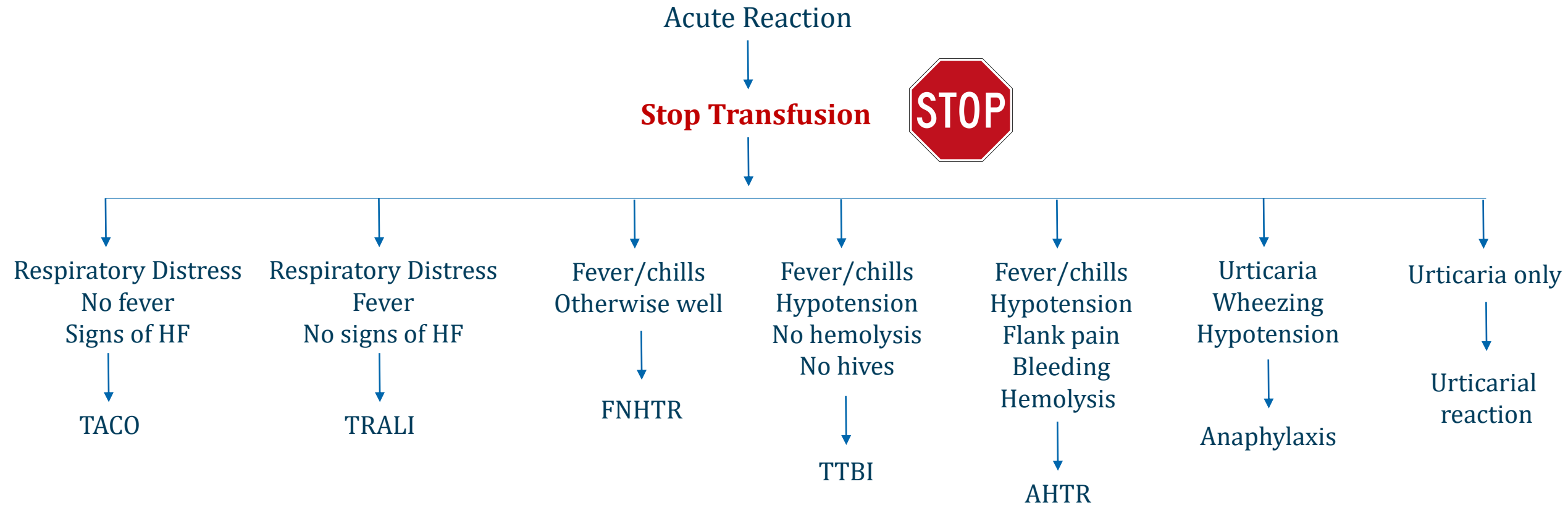
TTBI

Transfusion-transmitted blood infection

- Bacterial infection caused by transfusion
- Fever, chills
- Hypotension from septic shock
- Diagnosis: blood culture
- Treatment: antibiotics



Transfusion Reactions



Transfusion Reactions

Onset

- Seconds to minutes: **anaphylaxis**
- Within an hour: **AHTR**
- Minutes to hours: many causes
 - FNHTR
 - TRALI
 - TACO
 - TTBI
 - Urticaria
- Days to weeks: **delayed hemolytic reaction**



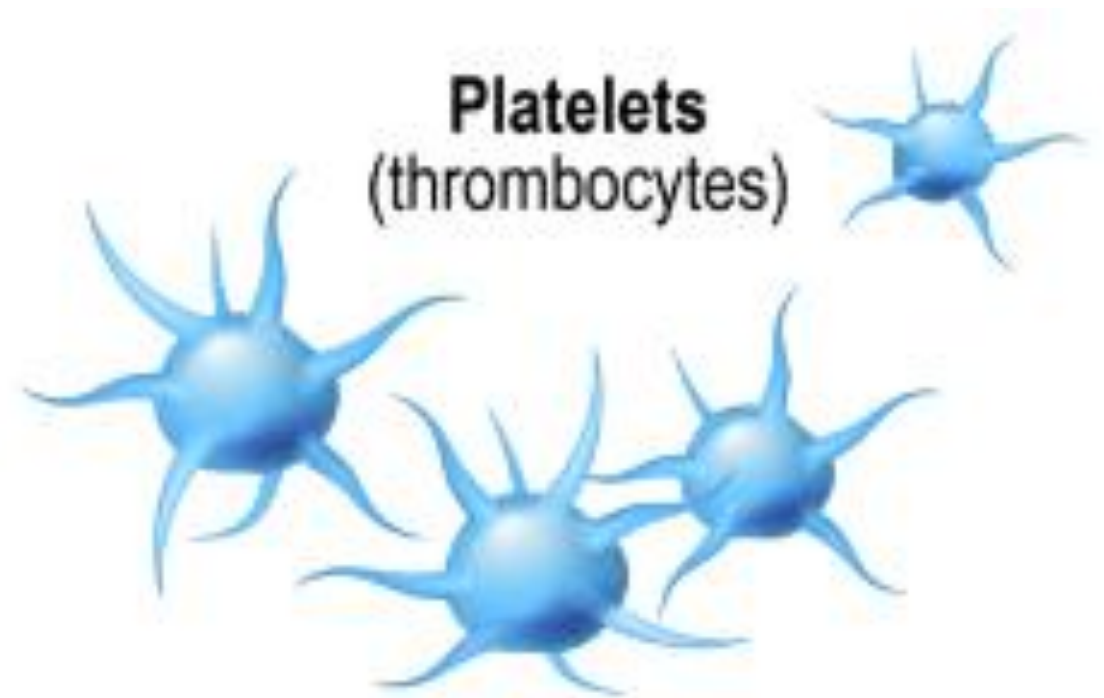
Thrombocytopenia

Jason Ryan, MD, MPH



Platelet Function

- Platelet activation → thrombus formation
- **Von Willebrand Factor**
 - Binds platelets together and to damaged endothelium
- **GPIb**
 - Platelet surface molecule
 - Binds vWF
- **GPIIb/IIIa**
 - Platelet surface molecule
 - Binds fibrinogen or vWF



Types of Bleeding

- **Abnormal platelets**
 - Skin bleeding
 - Mucosal bleeding
 - Petechiae: pinpoint, round spots on skin
 - Purpura: coalesced petechiae
- **Abnormal coagulation factors**
 - Joint bleeding, deep tissue bleeding

Petechiae



Thrombocytopenia

- Platelet count **below 150,000/microL**
 - Normal: 150,000 to 400,000/microL
- Risk of bleeding only when severe: $< 50,000/\text{microL}$
- Long list of causes
- Asymptomatic, isolated thrombocytopenia: most commonly ITP
- Hospitalized patients with thrombocytopenia: many causes
 - Platelet consumption
 - Bone marrow suppression
 - Drugs

Thrombocytopenia

Etiology:

Autoimmune	- ITP - SLE, RA
Consumption	- HUS/TTP, DIC, HIT
Production	- Bone marrow failure (presents as pancytopenia) - Myelodysplasia - B12/Folate deficiency
Drug Induced	- NSAIDs - Sulfa drugs - IIb/IIIa inhibitors - Heparin - Quinine - EtOH
Infection	- HIV, Hep C, EBV
Genetic	- Bernard Soulier (AR Defect in GPIb-IX) - Glanzmann Thrombasthenia (AR Defect in GPIIb-IIIa)
Sequestration	- Seen with hypersplenism, portal hypertension
Pseudo	- Pseudothrombocytopenia (platelet clumping, resulting in abnormal test)
Special Scenarios: - Malignancy (bone marrow invasion or DIC) - Pregnancy (gestational thrombocytopenia, preeclampsia, HELLP) - Liver failure (↓ TPO, splenic sequestration)	

Thrombocytopenia

Treatment

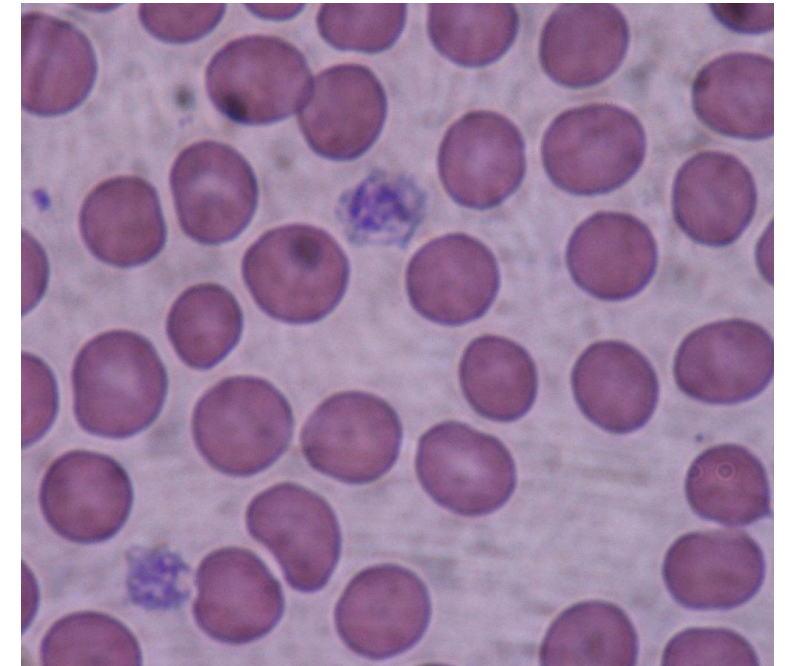
- Platelet transfusions
- Various cutoffs used based on specific patient scenarios
- Common thresholds
 - Bleeding: $< 50,000/\text{microL}$
 - Planned surgery: $< 50,000/\text{microL}$
 - Bleeding prevention: $< 10,000/\text{microL}$
- Platelets express ABO antigens on their surface
 - Matched transfusions result in greater increase in platelets
 - Mismatched platelets used when inventory low
 - Transfusion reactions uncommon (but may occur)



Congenital Platelet Disorders

- **Bernard-Soulier syndrome**
 - Autosomal recessive disorder
 - Deficiency of GPIb platelet receptors
 - Platelets cannot bind vWF
 - Thrombocytopenia with **giant platelets**
- **Glanzmann's Thrombasthenia**
 - Autosomal recessive disorder
 - Functional deficiency of GPIIb/IIIa receptors
 - Bleeding but platelet count usually normal
 - Blood smear: isolated platelets (no clumping)
 - Absent platelet aggregation in response to stimuli

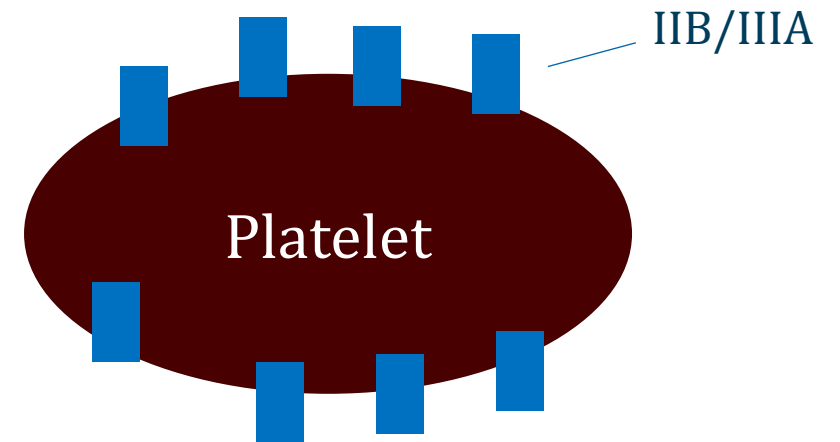
Giant Platelets



Immune Thrombocytopenia

Idiopathic thrombocytopenic purpura or ITP

- Disorder of decreased platelet survival
 - Platelet consumption by splenic macrophages
 - Commonly caused by anti-GPIIB/IIIA antibodies
- Presents with **isolated thrombocytopenia** +/- bleeding
 - Normal WBC and RBC
 - Normal coagulation



Immune Thrombocytopenia

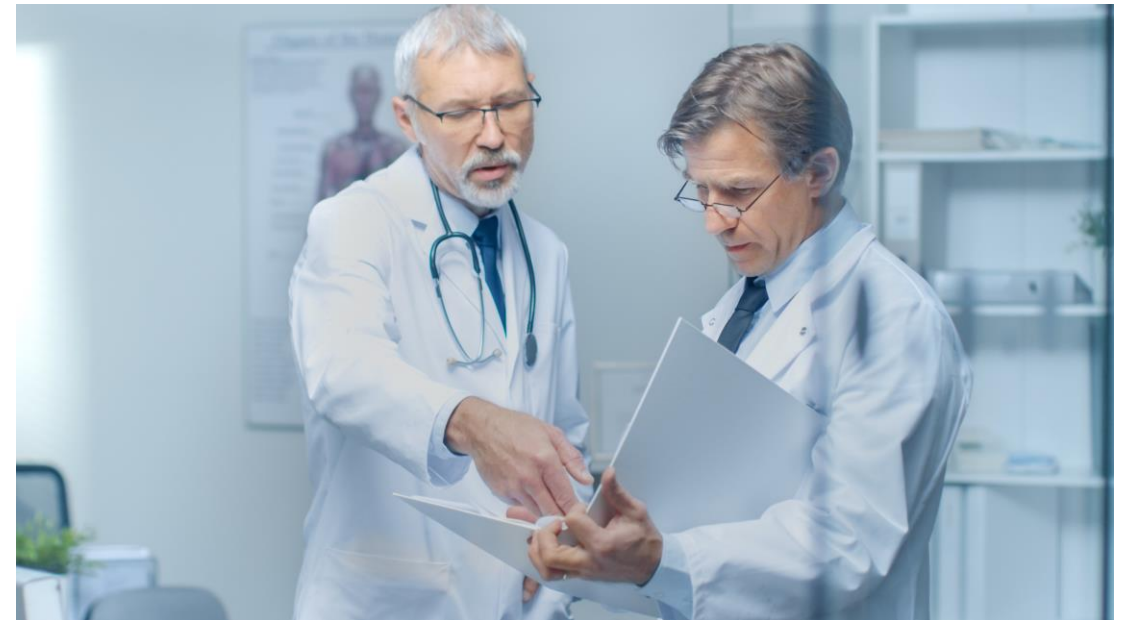
Causes

- **Primary**: no associated condition
- May occur **secondary** to another condition
 - Chronic lymphocytic leukemia
 - Systemic lupus erythematosus
 - HIV
 - Hepatitis C virus
 - Cytomegalovirus
 - Varicella zoster virus
- Testing at diagnosis: HIV and hep C
- Other tests if clinical features suggestive (e.g., ANA for lupus)

Immune Thrombocytopenia

Diagnosis

- Diagnosis of exclusion
- No reliable antibody test
- Antibodies not identified in many patients
- Isolated thrombocytopenia
- Normal WBC and RBC
- No other cause of thrombocytopenia



Immune Thrombocytopenia

Treatment

- Children: **often resolves spontaneously**
 - Often mild thrombocytopenia with lower bleeding risk
 - Only observation and monitoring usually required
 - Usually treated only if major bleeding or planned surgery
- Adults: often chronic with persistent thrombocytopenia

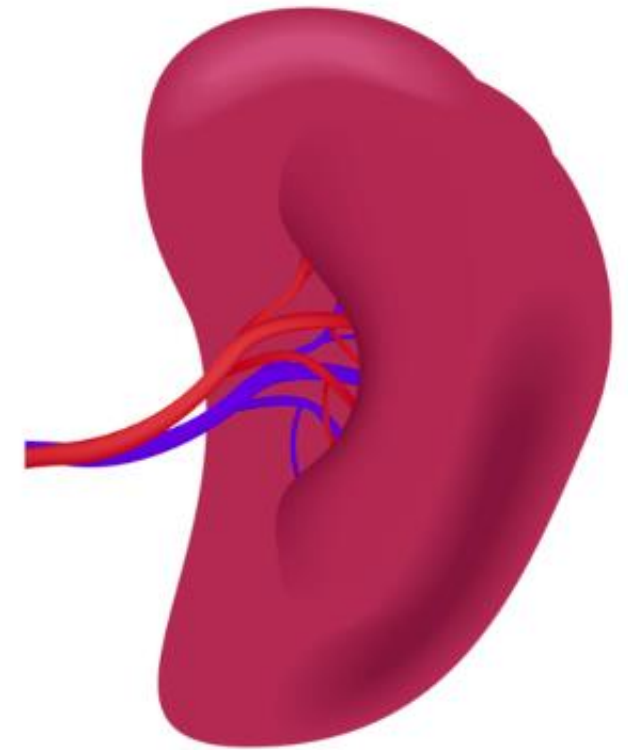


Immune Thrombocytopenia

Treatment

- Platelet count $> 30,000/\text{microL}$ and no bleeding: observe
- Treatment usually restricted to platelets $< 30,000$
- First line therapy: **corticosteroids**
 - Will raise platelet count
- Alternative: **intravenous immune globulin (IVIG)**
 - Pooled antibodies that blocks Fc receptors in macrophages
- Refractory thrombocytopenia: **splenectomy**
- Platelet transfusions used if severe bleeding

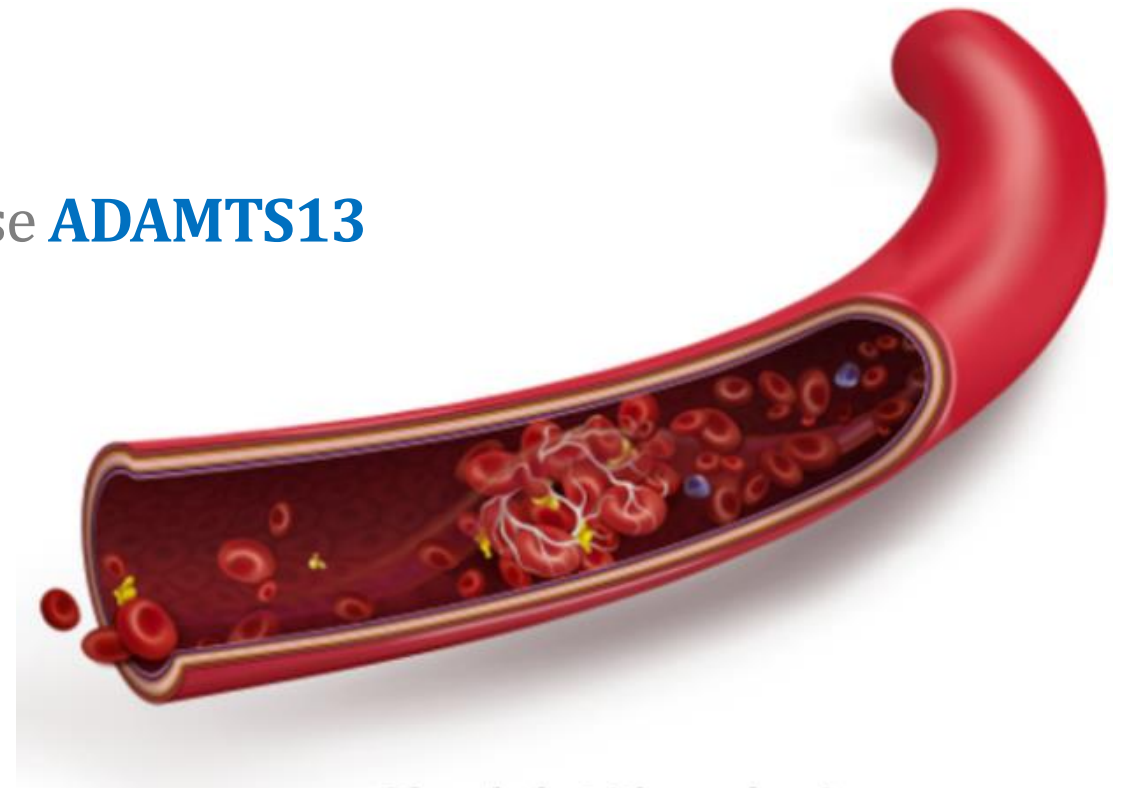
Spleen



Thrombotic Thrombocytopenic Purpura

TTP

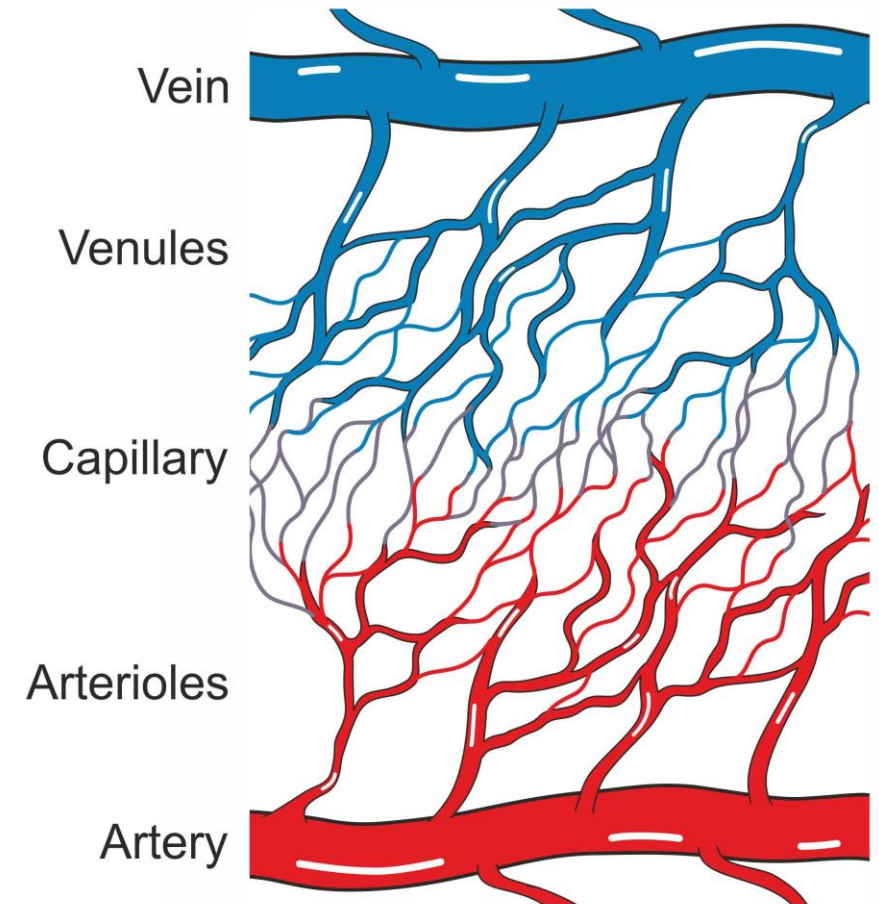
- “Thrombotic Microangiopathy”
 - Disorder of **small vessel thrombus** formation
- “Platelet-rich thrombi”
 - Consumption of platelets → thrombocytopenia
- Decreased activity of vWF cleaving protease **ADAMTS13**



Blood clot (thrombus)

ADAMTS13

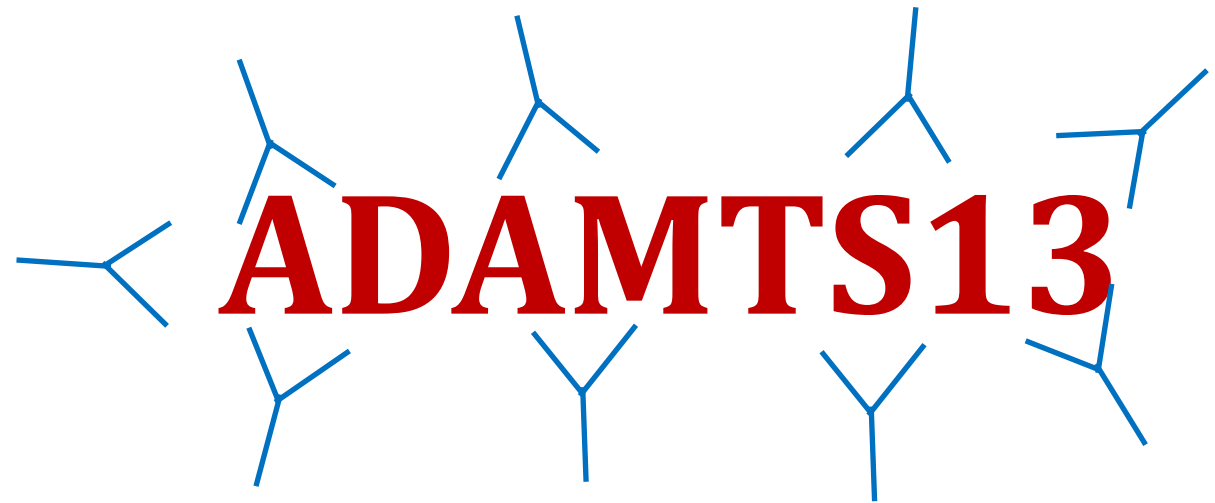
- vWF stored in cells as **large multimers**
- Secreted into plasma by endothelial cells
- Remains attached to the endothelial surface
- **ADAMTS13** cleaves large multimers
- Prevents large multimers from accumulating
- Especially in small arterioles and capillaries
 - High shear stress promotes platelet aggregation
 - Multimers + shear stress → thrombus formation
- Deficient ADAMTS13 → **small vessel thrombus**



Thrombotic Thrombocytopenic Purpura

Cause

- Severe ADAMTS13 deficiency
- Usually < 10% normal activity
- Usual cause: **acquired autoantibody to ADAMTS13**
- Results in large vWF multimers in areas of high shear stress
- Obstruction small vessels

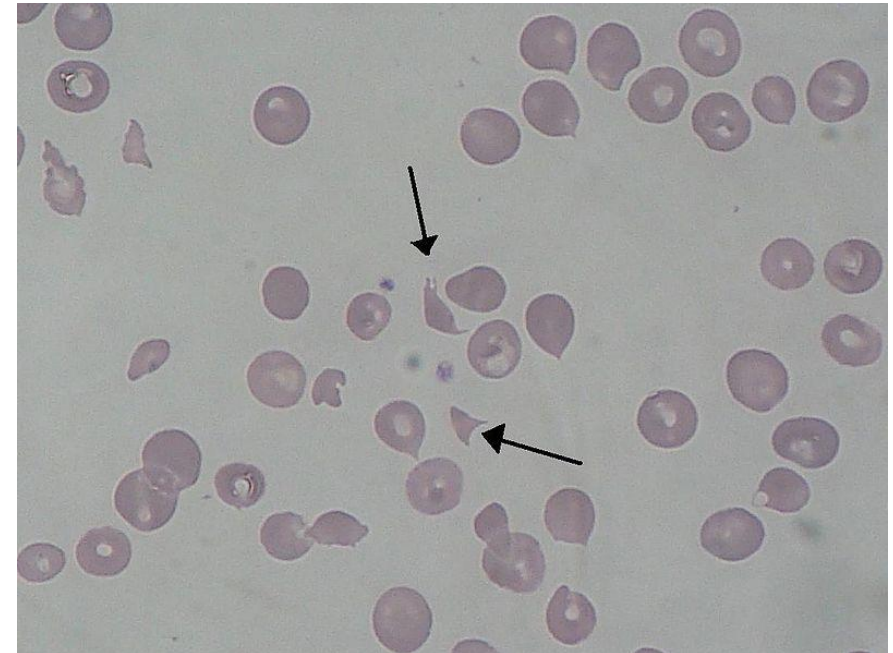


MAHA

Microangiopathic hemolytic anemia

- Shearing of RBCs as they pass through small vessels with thrombi
- **Non-immune hemolytic anemia**
 - Anemia
 - ↑ LDH, ↑ bilirubin, ↓ haptoglobin
 - ↑ reticulocytes
 - Negative Coombs testing
- Blood smear: **schistocytes**
- Seen in several disorders including TTP
 - Also HUS and DIC

Schistocytes



Thrombotic Thrombocytopenic Purpura

Clinical features

- Petechiae/purpura and bleeding (thrombocytopenia)
- Fatigue and dyspnea (anemia)
- Neurological symptoms (thrombus)
- Headache, confusion, seizures, stroke
- Fever in some cases
- Usually normal renal function

Purpura



Thrombotic Thrombocytopenic Purpura

Diagnosis

- Symptoms often lead to CBC + peripheral smear
- Suspected with MAHA and thrombocytopenia
 - Increased reticulocytes and bilirubin + schistocytes on smear
- **ADAMTS13 activity testing**
 - Usually <10% normal activity
 - Not required to initiate therapy
 - Early therapy can be life saving
- PT/PTT should be normal (contrast with DIC)
- May see elevated d-dimer



Thrombotic Thrombocytopenic Purpura

Treatment

- **Plasma exchange (PEX)**
 - Plasmapheresis
 - Removes antibodies
 - Very high mortality without PEX
 - **Treatment should not be delayed**
 - Classic clinical presentation → start PEX
- Glucocorticoids
- Platelet count monitored to determine efficacy
- Platelet transfusion not often used

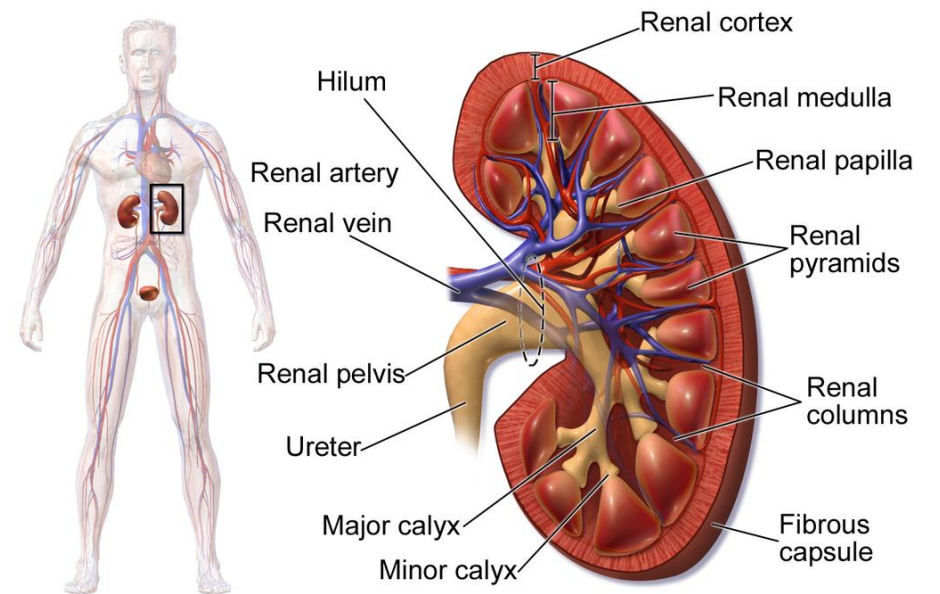
Plasmapheresis



Hemolytic Uremic Syndrome

HUS

- Also a “thrombotic microangiopathy” like TTP
- Also caused by platelet-rich thrombi in small vessels
- MAHA and thrombocytopenia
- **Acute kidney injury common**
- Fever or CNS symptoms uncommon

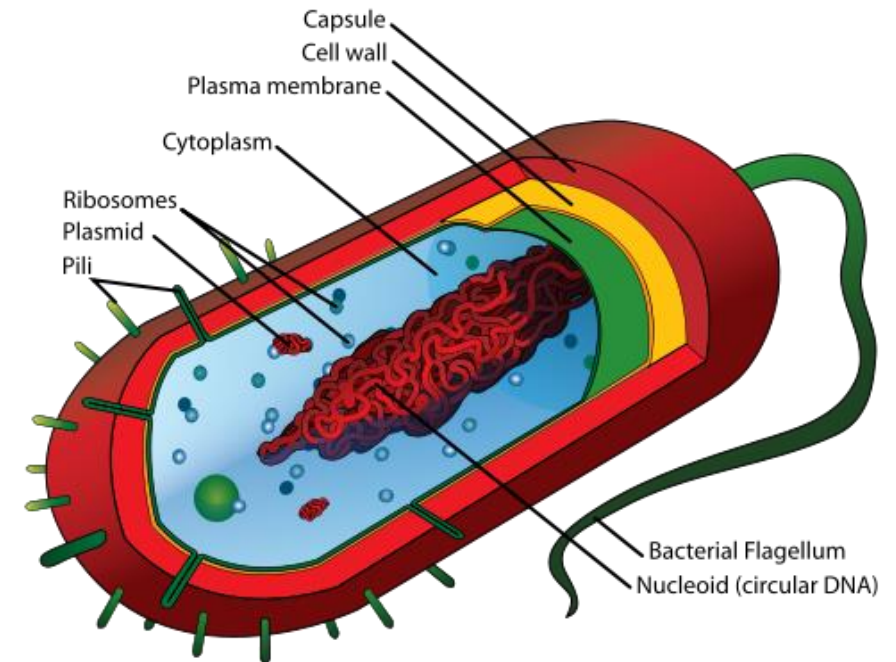


Kidney Anatomy

Hemolytic Uremic Syndrome

HUS

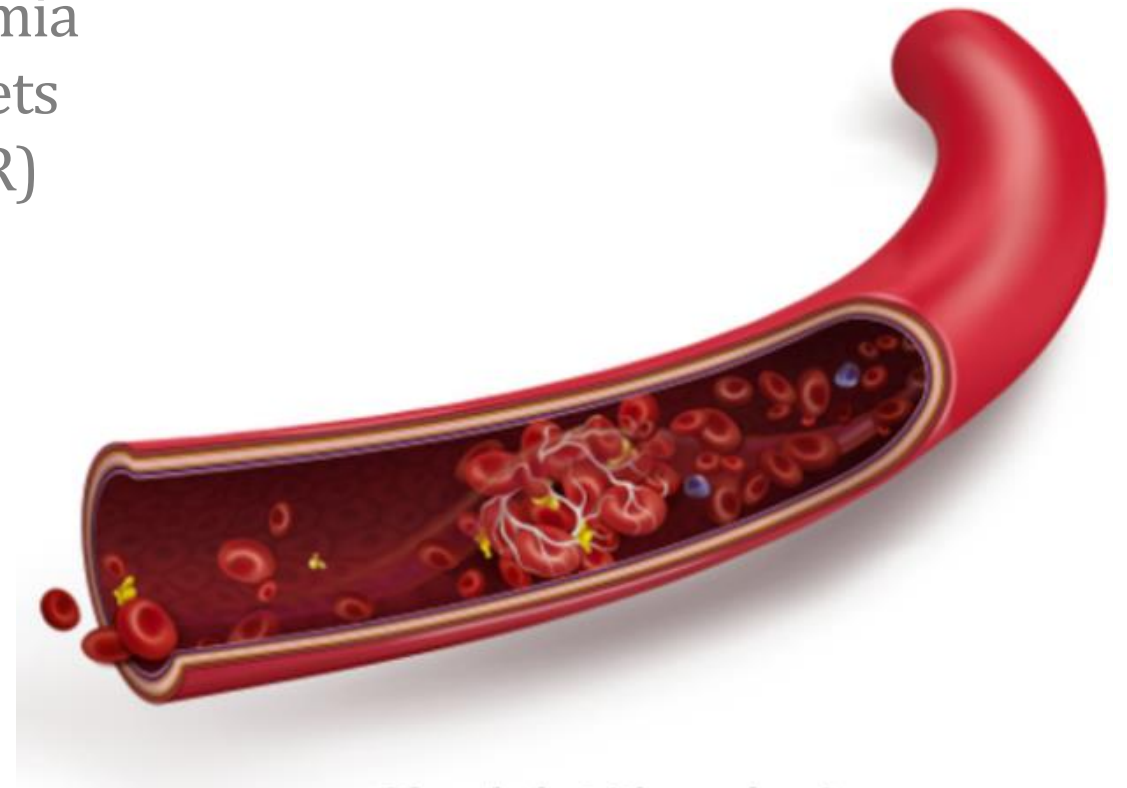
- Often occurs in children
- Commonly follow GI infection **E. Coli O157:H7**
- **Shiga-like toxin** causes microthrombi (not ↓ ADAMTS13)
- Treatment: **supportive care**
 - Transfusions
 - Dialysis for severe renal failure
- Plasma exchange not effective



Disseminated Intravascular Coagulation

DIC

- Widespread activation of clotting cascade
- Diffuse thrombi (platelets/fibrin) → ischemia
- Consumption of clotting factors and platelets
- Prolonged coagulation times (PT/PTT/INR)
- Thrombocytopenia
- Microangiopathic hemolytic anemia



Blood clot (thrombus)

Disseminated Intravascular Coagulation

Causes

- Occurs secondary to another process
- **Sepsis**
 - Endotoxin from bacteria
 - Cytokines
- **Obstetrical emergencies**
 - Amniotic fluid contains tissue factor
 - DIC seen in conjunction with amniotic fluid embolism
- Malignancy

Disseminated Intravascular Coagulation

Diagnosis

- Clinical and laboratory diagnosis
- Requires appropriate clinical setting (e.g., sepsis)
- Microangiopathic hemolytic anemia
- **Elevated PT/PTT/INR**
- Low platelets
- Low fibrinogen (consumption)
- Elevated D-dimer



Disseminated Intravascular Coagulation

Treatment

- **Treat underlying disorder**
- Supportive care
- Blood and platelet transfusions
- Fresh frozen plasma: replace clotting factors



Thrombocytopenia

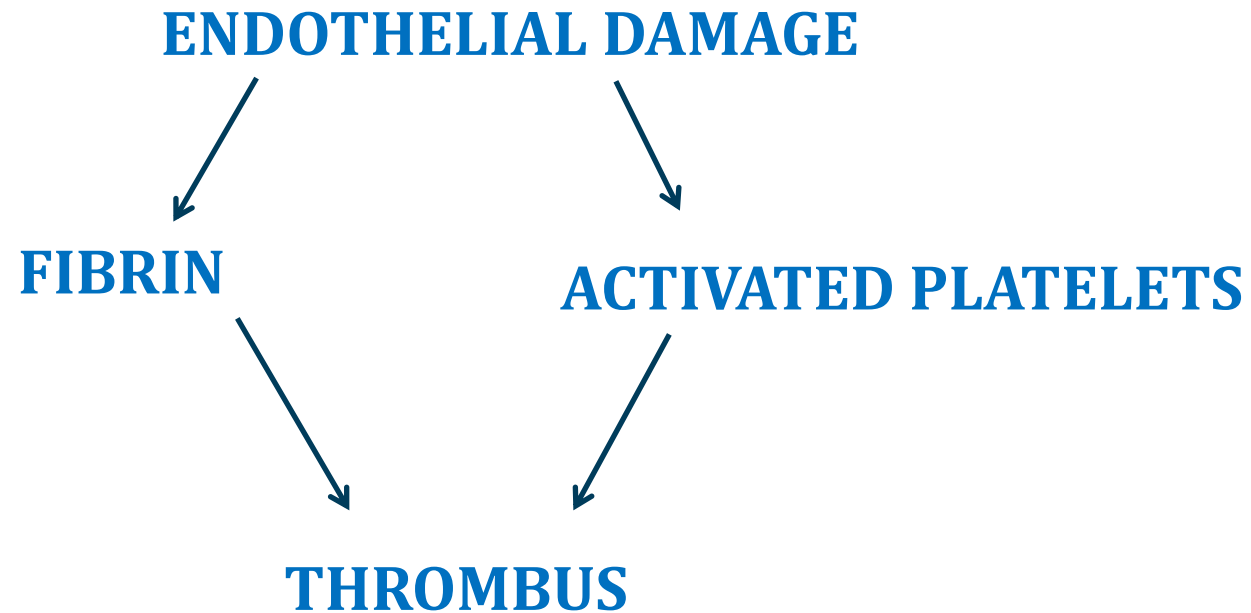
	Platelet Thrombi		Platelet/Fibrin Thrombi	
	ITP	TTP	HUS	DIC
↓ Platelets	+	+	+	+
MAHA	--	+	+	+
↑ PT/PTT	--	--	--	+
	CNS symptoms Fever		Child GI Illness	

Coagulopathy

Jason Ryan, MD, MPH



Thrombus Formation



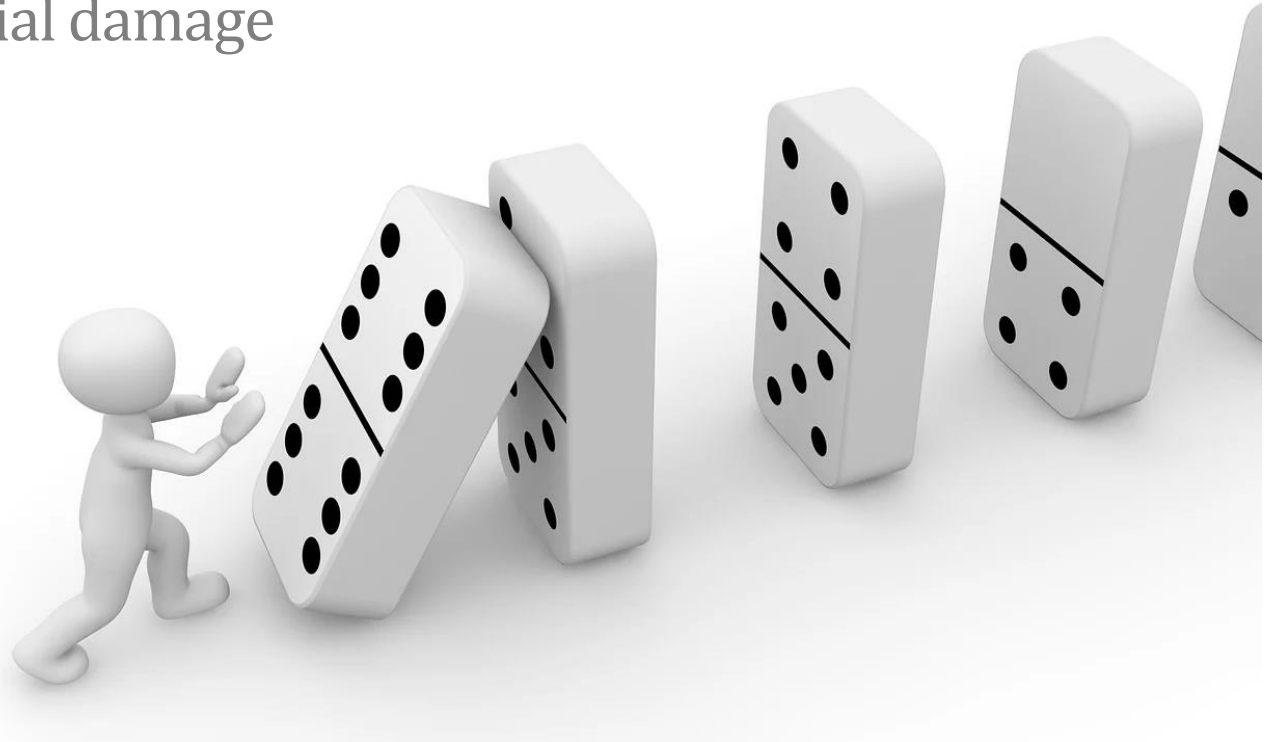
Coagulation Factors

- Proteins synthesized in liver
- Activate when triggered by **endothelial damage**
- Form an insoluble protein: **fibrin**
- Fibrin mesh prevents blood loss
- Named by Roman numerals: I, V, X
- Circulate as inactive enzymes (zymogens)
- Activated forms: Ia, Va, Xa

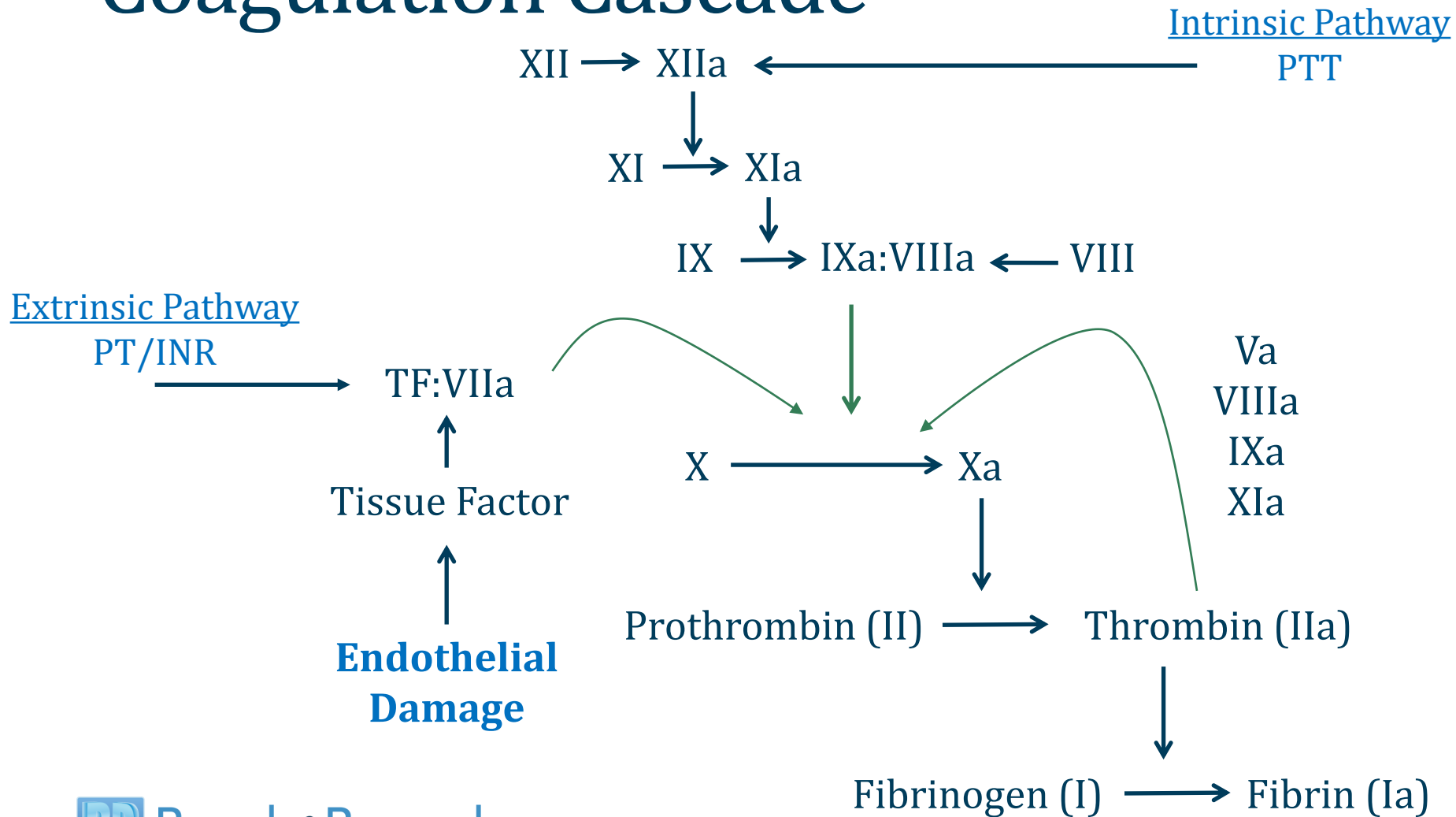


Coagulation Cascade

- Sequential activation of clotting factor zymogens
- Constant low level of activation in blood
- Amplification occurs with endothelial damage
- Leads to fibrin generation



Coagulation Cascade



Coagulation Testing

- **Prothrombin time (PT)**
 - Extrinsic pathway
 - Add plasma to tissue factor
 - Measure time to form clot: normal ~ 10s
- **International normalized ratio (INR)**
 - Patient PT/Control PT
 - Normal = 1
- **Partial thromboplastin time (PTT)**
 - Intrinsic pathway
 - Add plasma to (-) charge substance (silica)
 - Measure time to form clot: normal ~ 30s



Hemophilias

- X-linked recessive diseases
 - Mostly affects males
 - Female carriers usually asymptomatic
- **Hemophilia A**: deficiency of **factor VIII**
- **Hemophilia B**: deficiency of **factor IX**

Alexei Nikolaevich

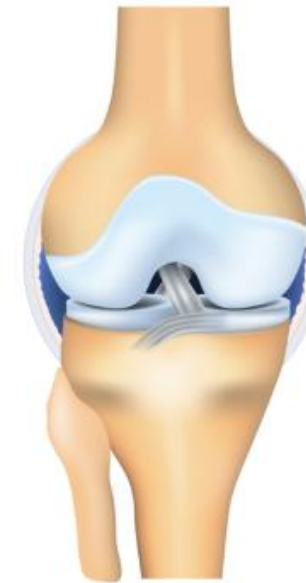


Hemophilias

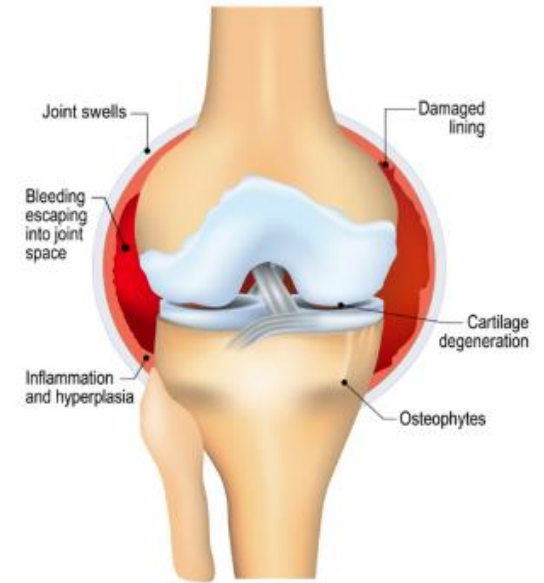
Clinical features

- Spontaneous or easy bleeding
- **Hemarthrosis**
 - Recurrent joint bleeds
 - Common initial presentation
- **PTT usually prolonged**
 - In moderate to severe forms
 - May be normal in mild disease
 - Factors VIII, IX both part of intrinsic pathway
- PT, platelet count: normal

HEMOPHILIA



Healthy joint

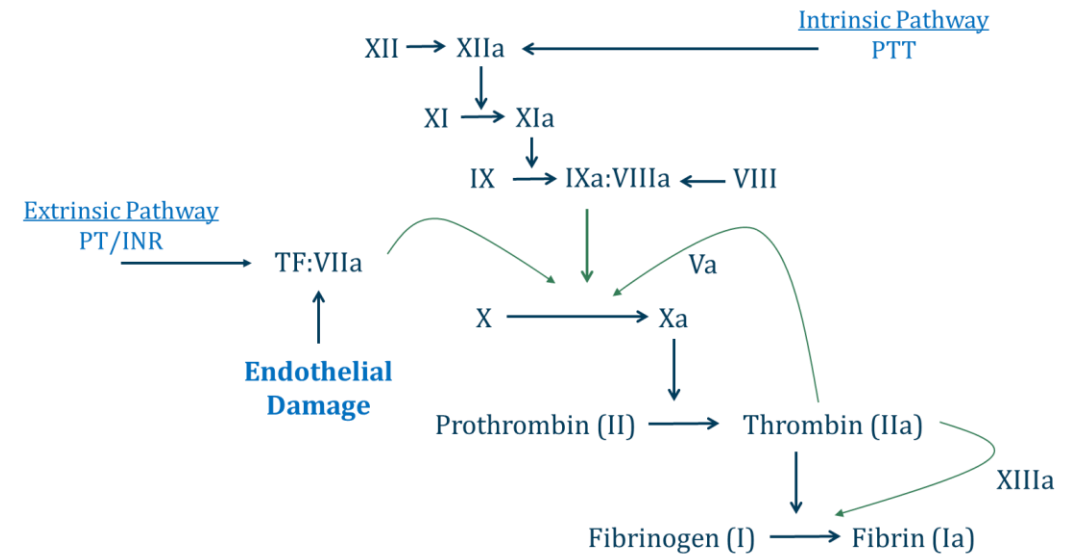


Hemophilia

Hemophilias

Diagnosis

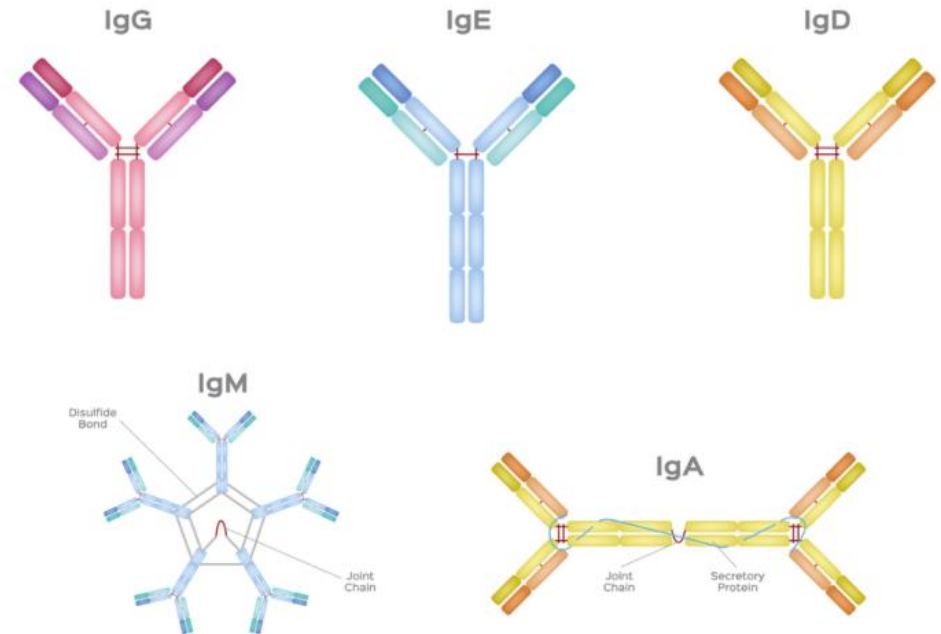
- **Factor VIII or IX activity assay**
- Normal: 55 to 150% normal value
- Hemophilia A: factor VIII activity level < 40% normal
- Hemophilia B: factor IX activity level < 40% of normal
- **Negative VWF antigen test**
 - Von Willebrand disease may also ↑ PTT



Hemophilias

Treatment

- **Replacement factor VIII and IX**
- Target factor activity level: 80 to 100%
- Acute, severe bleeding
- Before urgent or emergent surgery
- Some elective surgeries
- May lead to **antibody inhibitors**
 - Inadequate response to replacement factors
 - Regular screening performed

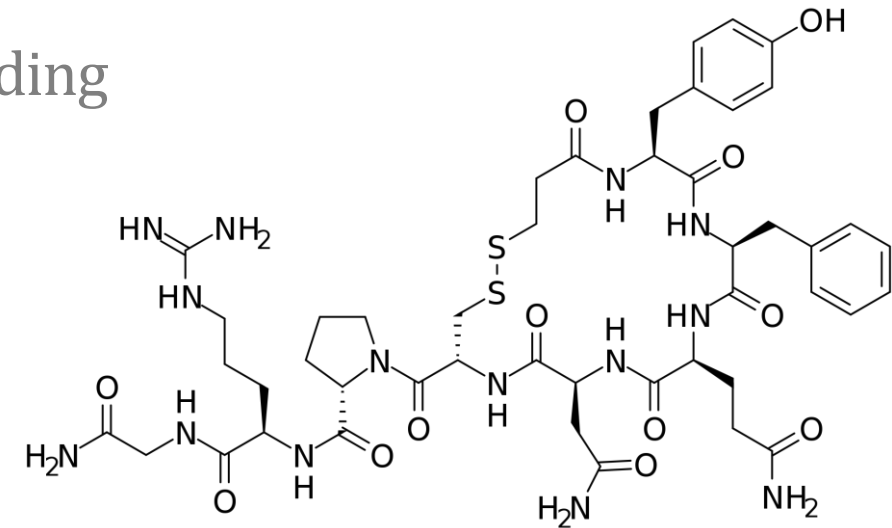


Hemophilias

Treatment - Desmopressin

- 1-deamino-8-D-arginine vasopressin (DDAVP)
- Synthetic analogue of antidiuretic hormone
- Triggers release of vWF from endothelial cells
- Increases **vWF and factor VIII levels**
- Only used for brief periods (24 - 48 hours)
- Can be used before minor surgery or with minor bleeding
- Prolonged use: diminishing response (tachyphalxis)

Desmopressin



Treatment - Desmopressin

- May cause hyponatremia
- Also has vasodilating properties
- May cause flushing, headache
- Other uses:
 - von Willebrand disease
 - Central diabetes insipidus (mimics ADH)
 - Bedwetting (decreases urine volume)

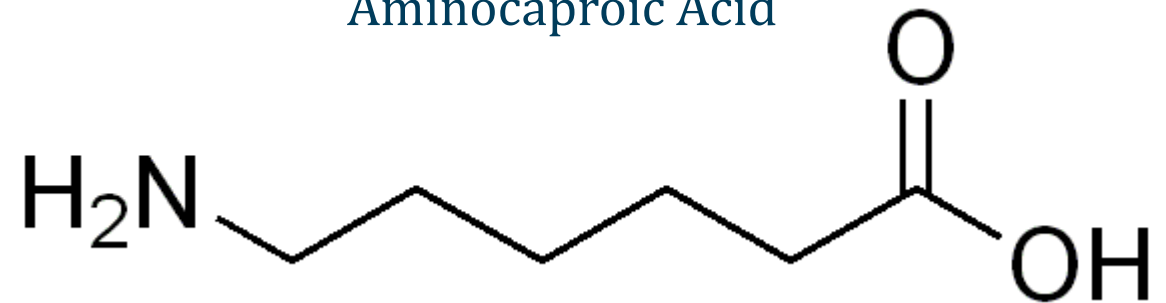
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Hemophilia

Treatment – Aminocaproic acid and Tranexamic acid

- Antifibrinolytic drugs
 - Inhibit formation of plasmin
 - Less breakdown of formed clots
- Useful against bleeding in mouth or nose
- Mucosal areas of increased fibrinolysis
- Dental bleeding
- Epistaxis

Aminocaproic Acid



Acquired Coagulation Inhibitors

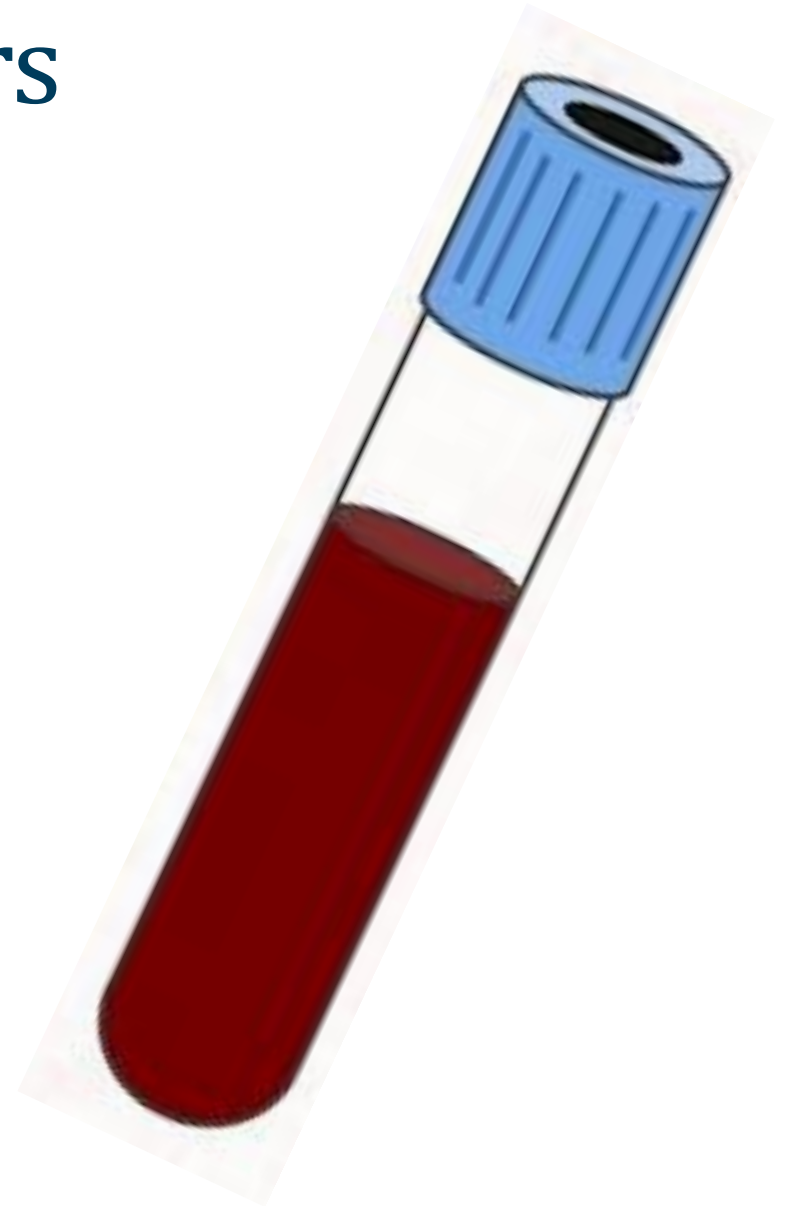
- Antibodies
- Inhibit activity or increase clearance of clotting factor
- Inhibitors of **factor VIII** most common
- Often occur in association with:
 - Malignancy
 - Post-partum
 - Autoimmune disorders
- May respond to treatment with immunosuppression
 - Most commonly prednisone



IgG

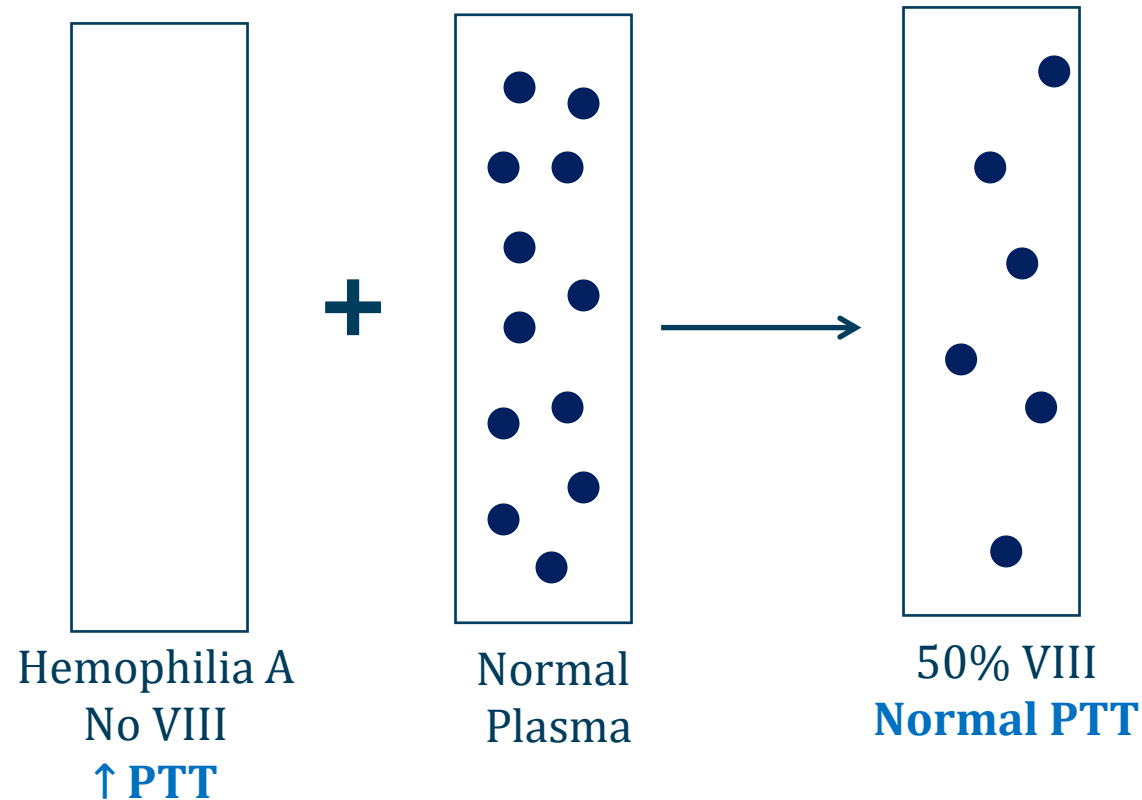
Acquired Coagulation Inhibitors

- Can present similar to hemophilia
 - Deficient activity of VIII → bleeding
 - Prolonged PTT
 - “Acquired hemophilia”
- Differentiate from hemophilia A: **mixing study**
- PTT does not correct on mixing with normal plasma



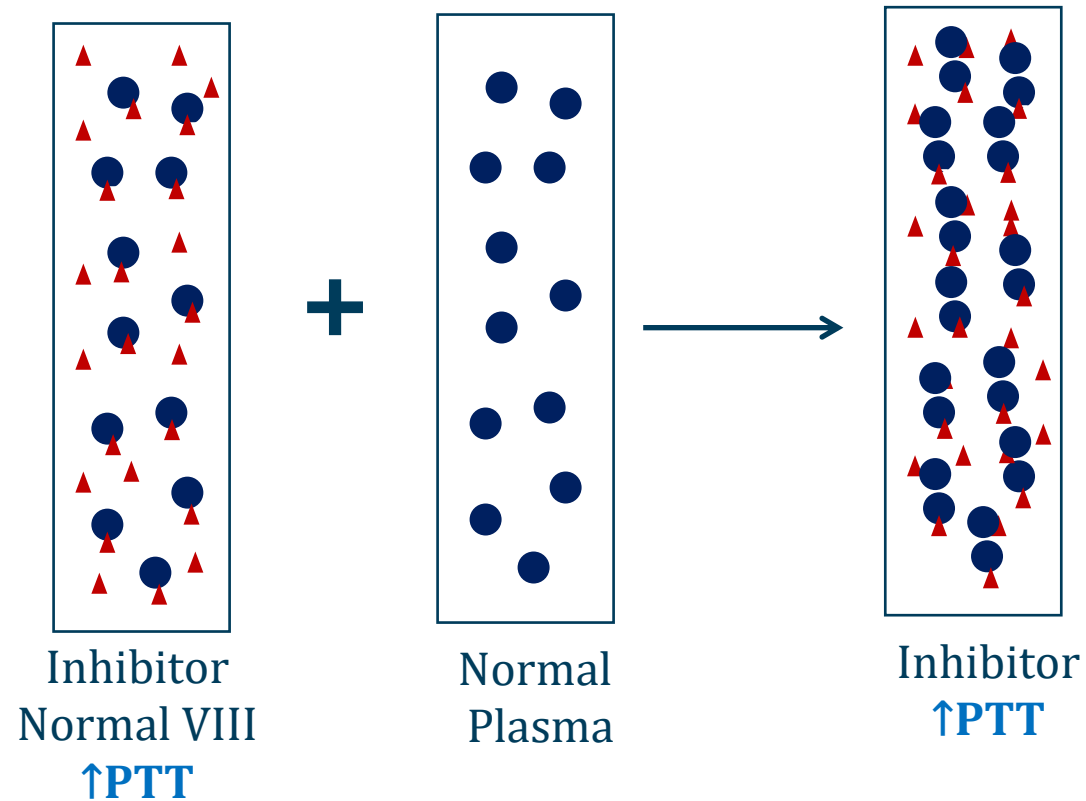
Mixing Study

- Used to evaluate **unexplained prolongation of a clotting test**
- Clotting factors ~ 50% normal → normal PT/PTT



Mixing Study

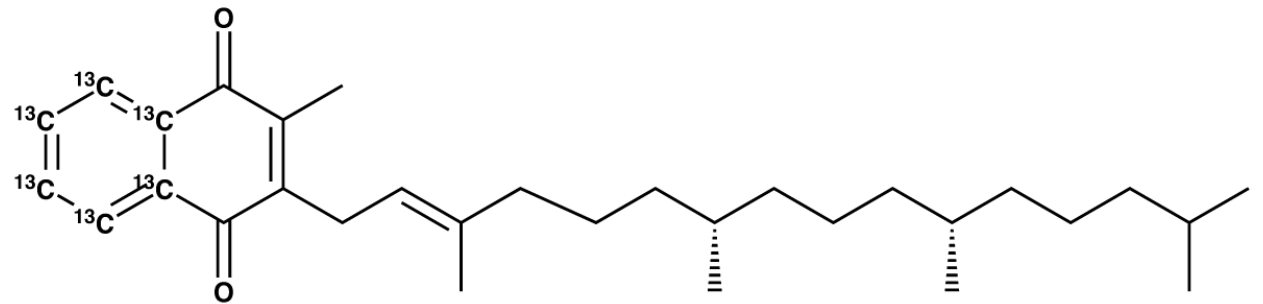
- Clotting factors \sim 50% normal \rightarrow normal PT/PTT



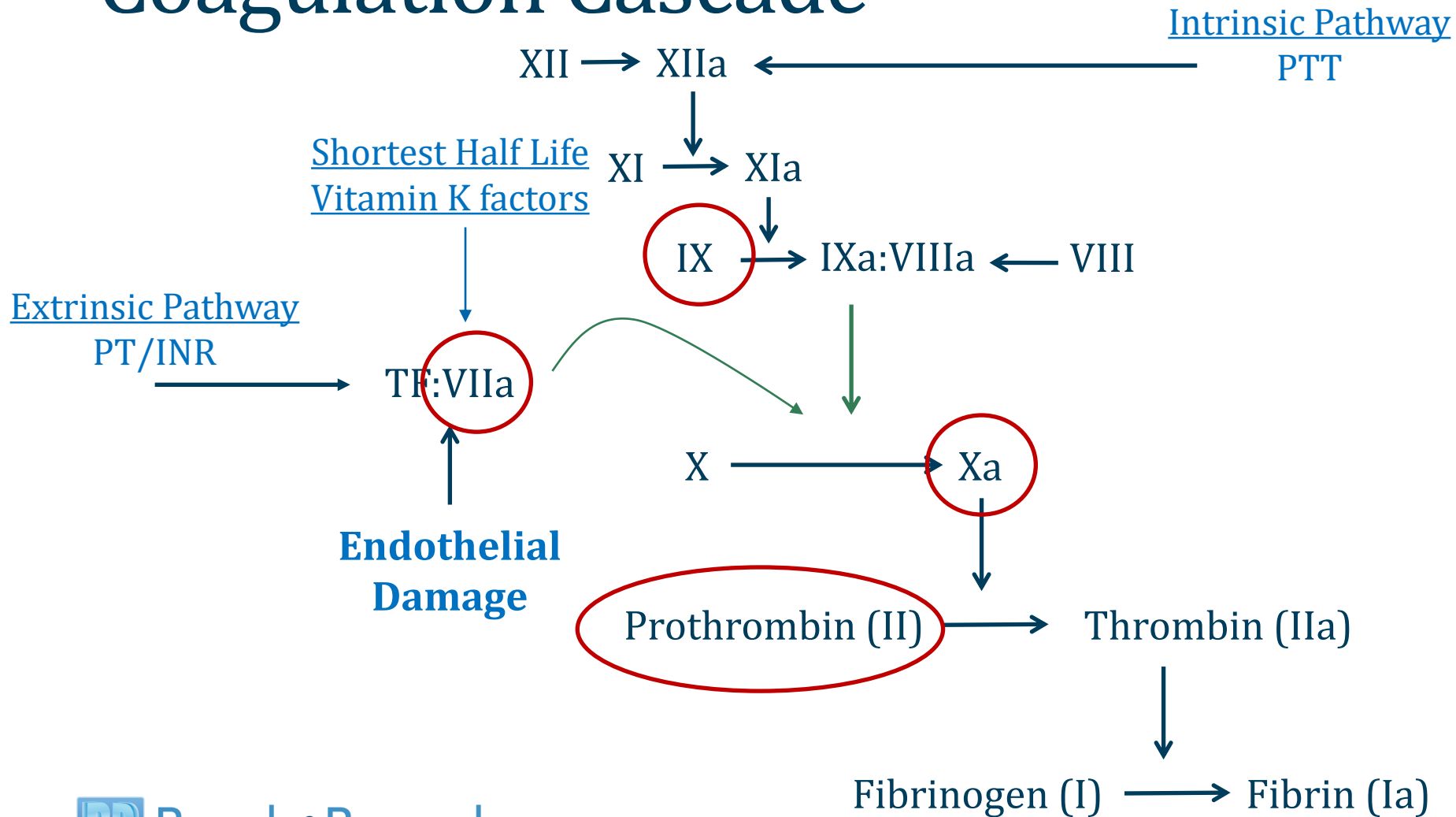
Vitamin K Deficiency

- Deficiency of vitamin K-dependent factors
 - Factors II, VII, IX, X
- **Bleeding**
- **Prolonged PT/INR**
 - Factor VII has shortest half-life
- Can see prolonged PTT (less sensitive)

Vitamin K1



Coagulation Cascade



Vitamin K Deficiency

Causes and treatment

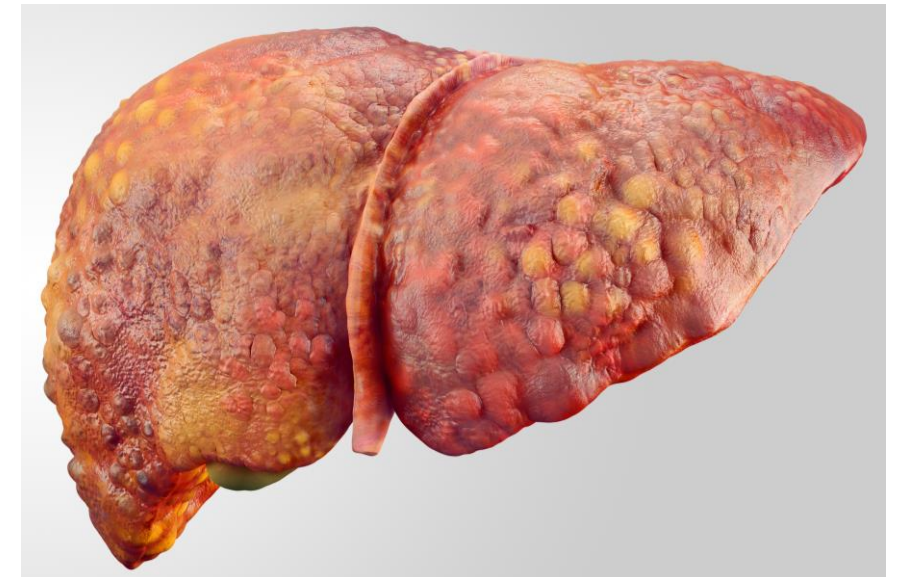
- Dietary deficiency rare
- GI bacteria produce sufficient quantities
- Warfarin – inhibits vitamin K activation in liver
- **Antibiotics** - deplete GI bacteria
- Newborns - sterile GI tract
- Malabsorption - vitamin K is fat soluble
 - Cystic fibrosis or pancreatic insufficiency
- Treat with vitamin K
- Can use PCC and FFP if bleeding



Liver Disease

- **Loss of clotting factors**
 - Advanced liver disease → ↓ clotting factor synthesis
- **Thrombocytopenia** also common
 - Decreased hepatic synthesis of thrombopoietin
 - Platelet sequestration in spleen from portal hypertension

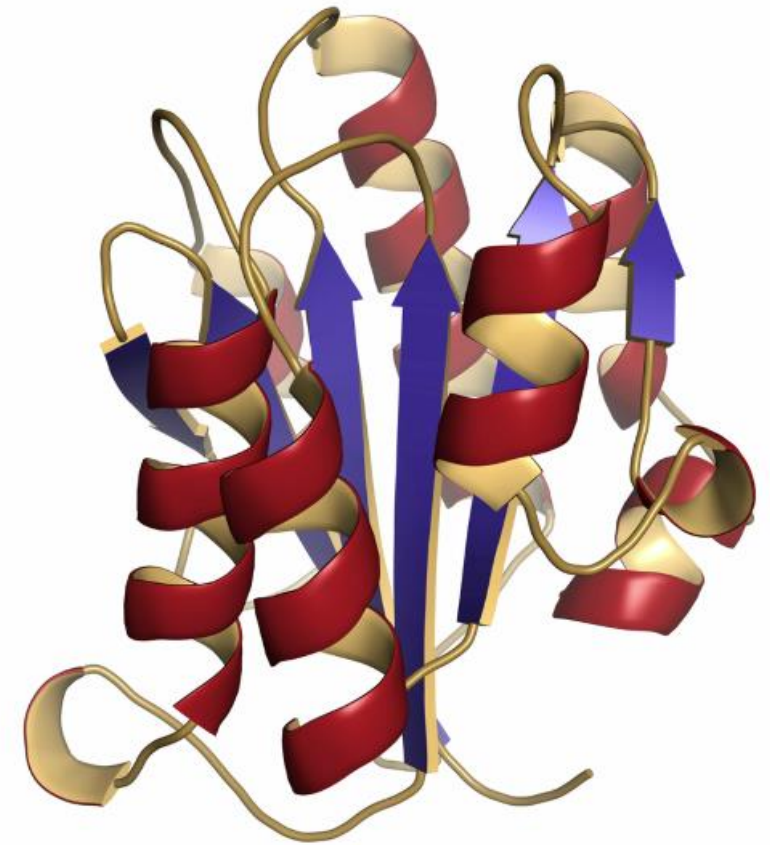
Liver Cirrhosis



Von Willebrand Disease

- Deficient function of **von Willebrand Factor**
 - Large glycoprotein
 - Found in endothelial cells and platelets
 - Factor VIII circulates bound to vWF
- Binds platelets to endothelium and other platelets
- Most common inherited bleeding disorder
 - Affects up to 1 percent of population
- Gene mutations → ↓ level or function of vWF
 - Most cases autosomal dominant (males=females)

Von Willebrand Factor



Von Willebrand Disease

- Usually mild, non-life-threatening bleeding
- Easy bruising
- Skin bleeding
- Prolonged bleeding from mucosal surfaces
- Severe nosebleeds
- Heavy menstrual bleeding



Von Willebrand Disease

Diagnosis

- Normal CBC and PT/INR
- **Increased PTT**
 - Depending on severity of disease
 - Factor VIII circulates bound to vWF
 - Usually no joint/deep tissue bleeding like hemophilia
- **VWD screening tests**
 - VWF antigen test: measurement of VWF protein level
 - VWF activity test: test of VWF binding to platelets
 - Factor VIII activity
 - Low levels suggest VWD



Hypercoagulable States

Jason Ryan, MD, MPH



Hypercoagulable States

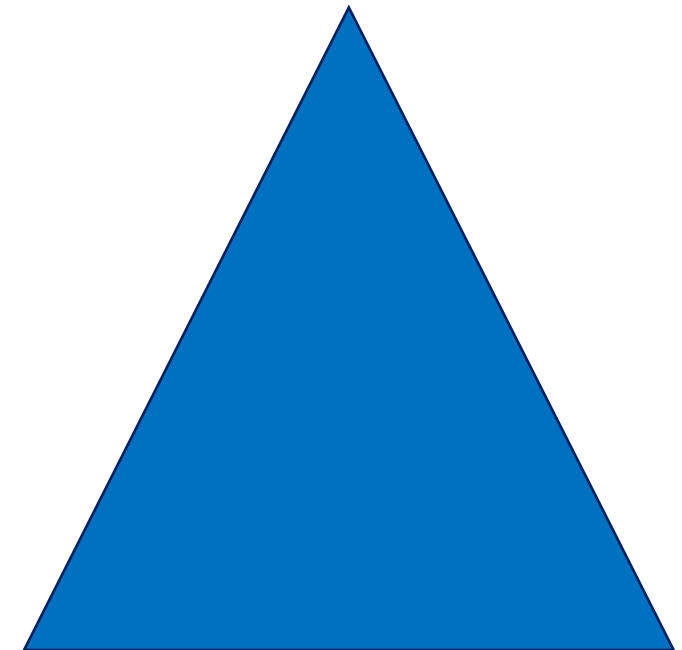
- Predisposition to venous or arterial thrombi
- Often **“venous thromboembolism”**
 - Deep venous thrombosis
 - Pulmonary embolism
- Sometimes arterial thrombosis
 - Stroke
 - Myocardial infarction
 - Ischemic limb



Blood clot (thrombus)

Virchow's Triad

- **Endothelial damage**
 - Endothelium makes natural anticoagulants
- **Stasis of blood**
 - Normal blood flow prevents pooling of clotting factors
- **Hypercoagulability**
 - Conditions that increase clot formation



Hypercoagulable States

- **Post surgery (“post-op state”)**
 - Hypercoagulable (inflammation from surgery)
 - Stasis (immobile)
 - Endothelial damage (surgery)
- Fall, fracture or trauma
 - Hypercoagulable (inflammation from trauma)
 - Stasis (immobility)
 - Endothelial damage (trauma)
- Long plane flights
 - Stasis (immobility)



Hypercoagulable States

- **Malignancy**
 - Some tumors produce pro-coagulants (i.e., tissue factor)
 - Inflammation associated with tumor growth
 - Decreased activity, surgery, bed rest
- Consider occult malignancy with **unprovoked venous thrombosis**
 - Absence of surgery, trauma, immobilization or known cancer
 - Limited workup for occult malignancy often done
 - History, physical exam and **age-appropriate cancer screening**



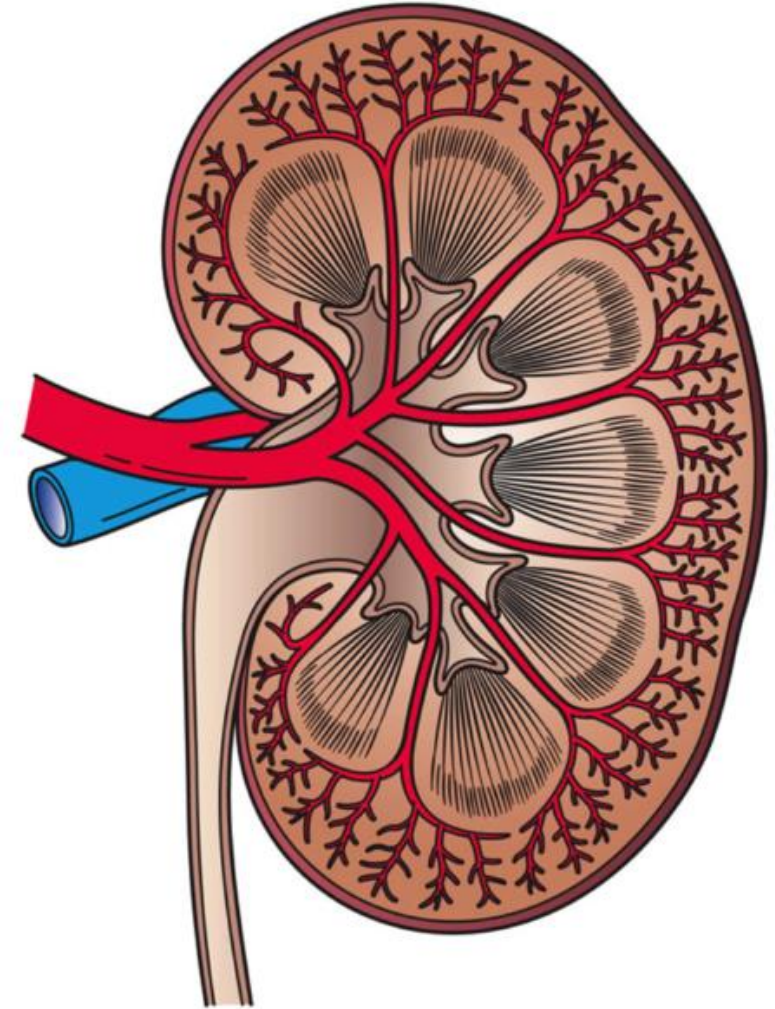
Hypercoagulable States

- **Pregnancy**
 - Probably evolved to protect against blood loss at delivery
 - Many clotting factor levels change
 - Fetus also obstructs venous return → DVTs common
- Estrogen-containing oral contraceptives
 - Estrogen increases production coagulation factors



Hypercoagulable States

- **Nephrotic syndrome**
 - Multiple mechanisms
 - Loss of anti-clotting factors in urine (ATIII)
- **Smoking**
 - Associated with atherosclerosis and MI/Stroke
 - Some data linking smoking to DVT/PE
 - Evidence that smoking increases fibrinogen levels



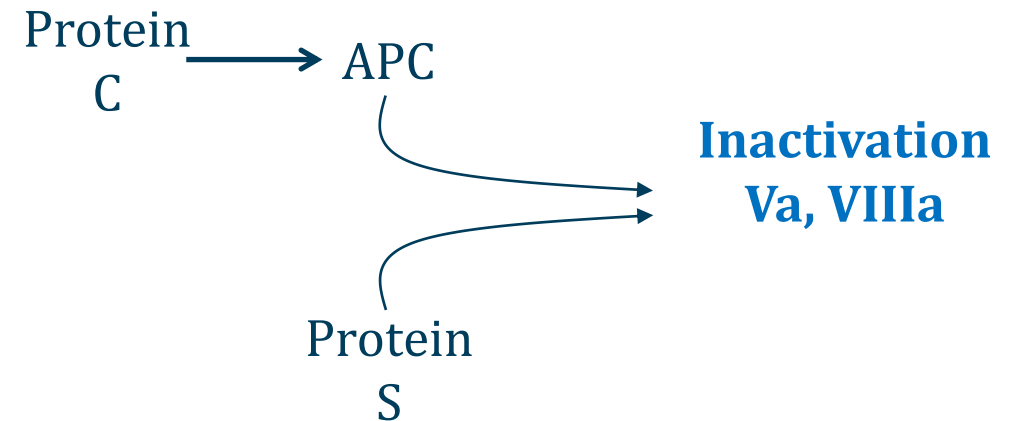
Inherited Thrombophilia

- Inherited hypercoagulable states
- Genetic tendency to **venous thromboembolism**
- Most involve coagulation pathway defects
- All associated with venous clots (DVT/PE)
- Involve anticoagulation factors
 - Counteract coagulation cascade
 - Protein C
 - Protein S
 - Antithrombin III

Hypercoagulable Condition
Factor V Leiden Mutation
Prothrombin gene mutation
Antithrombin deficiency
Protein C/S deficiency

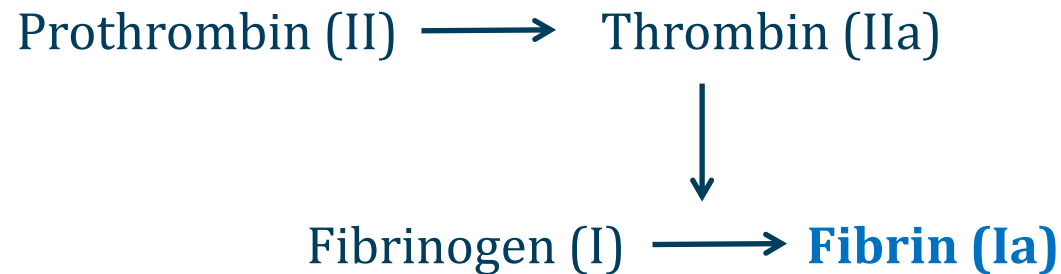
Factor V Leiden Mutation

- Most common inherited thrombophilia
- Named for Leiden, Netherlands
- Abnormal **factor V**
- Mutation in factor V gene
- Not inactivated by **activated protein C (APC)**
- Factor V remains active longer → hypercoagulability
- Diagnosis: genetic testing



Prothrombin Gene Mutation

- **Prothrombin 20210** gene mutation
 - Guanine to adenine change in prothrombin gene
 - Occurs at nucleotide 20210
- Heterozygous carriers: 30% ↑ prothrombin levels
- Diagnosis: genetic testing



Antithrombin III Deficiency

- Inherited deficiencies due to gene mutations
- Acquired deficiencies:
 - Impaired production (liver disease)
 - Protein losses (nephrotic syndrome)
- Classically presents as **heparin resistance**
 - Heparin activates ATIII
 - Deficiency state → little effect of heparin
 - Escalating dose of heparin → no/little change in PTT



Protein C or S Deficiency

- Deficiency associated with thrombosis
- Protein C deficiency associated with **warfarin skin necrosis**
- Initial warfarin therapy → ↓ protein C (short half life)
- If protein C deficient → marked ↓ protein C
- Result: **thrombosis of skin tissue**
- Large dark, purple skin lesions
- Diagnosis: low protein C/S levels



Bakoyiannis C, Karaolani G, Patelis N, Maskanakis A, Tsaples G, Klonaris C, Georgopoulos S, Liakakos T

Inherited Thrombophilia

Workup

- **First episode of VTE**
 - Testing not routinely done for unselected patients
- Testing performed only in special circumstances
 - Strong family history
 - Usually first-degree relative with VTE before age 45
 - Recurrent, multiple thromboses
 - Thromboses in unusual anatomic locations
 - Portal, hepatic, mesenteric or cerebral veins

Antiphospholipid Syndrome

- Caused by **antiphospholipid antibodies**
- Occur in association with **lupus** or as primary disease
- Increased risk of **venous and arterial thrombosis**
 - Most commonly deep vein thrombosis
 - Also arterial thrombosis → stroke
- Risk of **recurrent fetal loss**
- Interference with **laboratory testing**
 - Increased PTT
 - False positive syphilis (RPR/VDRL)



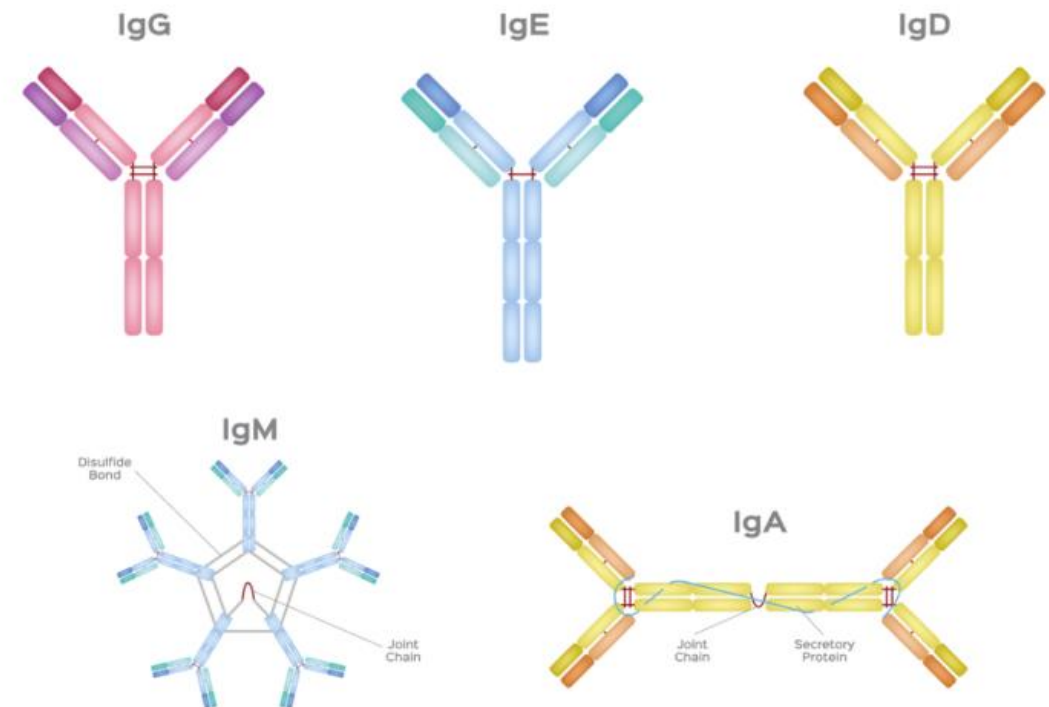
SYSTEMIC
LUPUS
ERYTHEMATOSUS

Antiphospholipid Syndrome

Specific antibodies

- **Anti-cardiolipin**
 - False positive RPR/VDRL
 - Syphilis also produces similar antibodies
- **“Lupus anticoagulant”**
 - Interferes with PTT test
 - False elevation of PTT
- **Anti- β 2 glycoprotein**

Antibodies



Antiphospholipid Syndrome

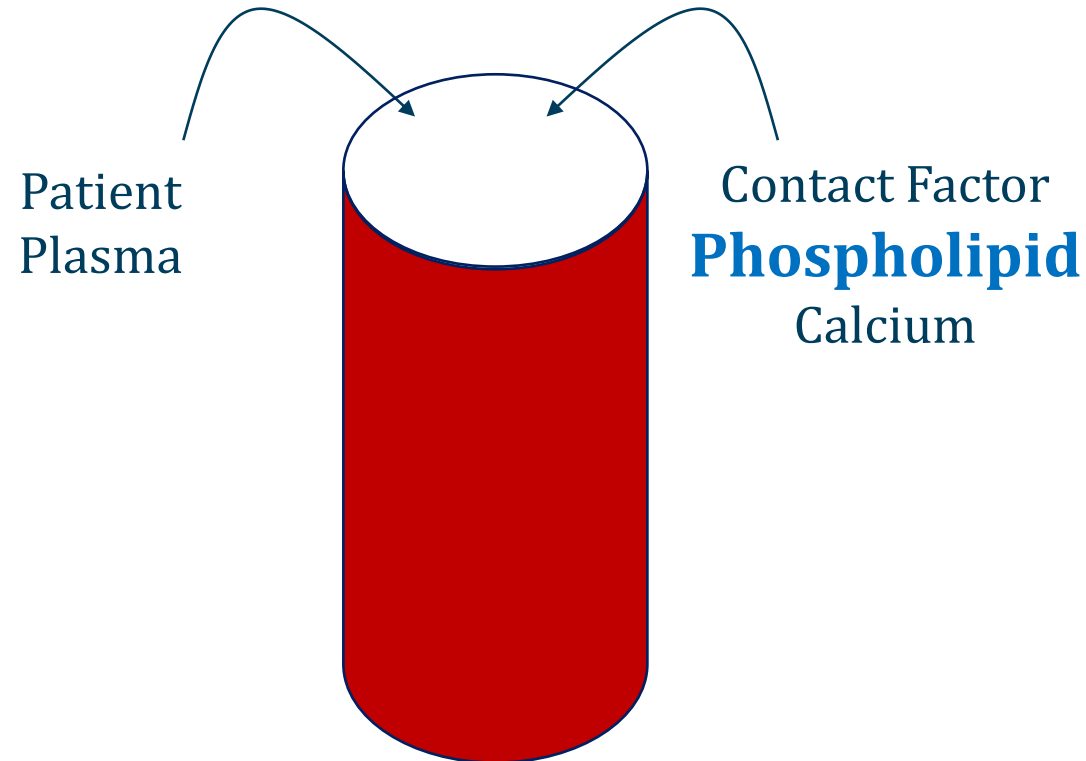
Antibody detection

- **Anti-cardiolipin** and **anti- β 2 glycoprotein**
 - Enzyme-linked immunosorbent assay (ELISA) testing
- **“Lupus anticoagulant”**
 - Detected *indirectly* through coagulation assays
 - Antibodies interfere with coagulation testing

Lupus Anticoagulant

PTT Testing

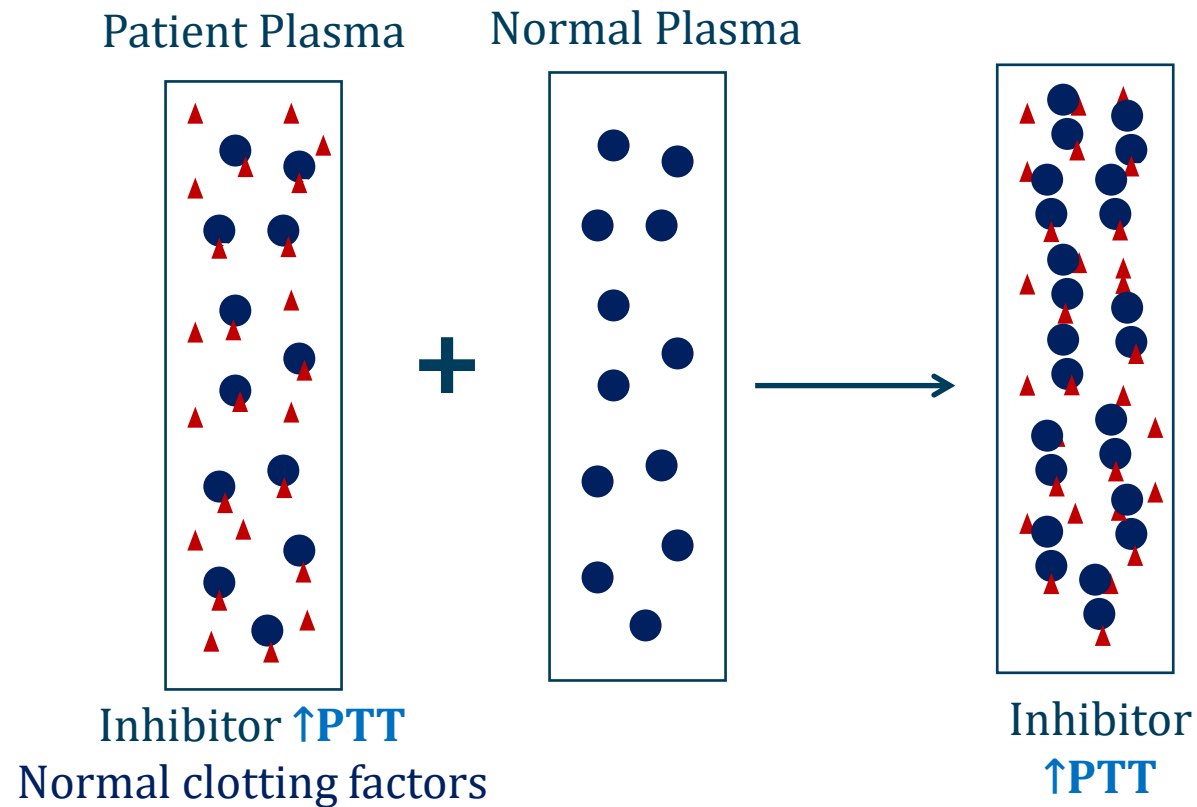
- Lupus anticoagulant binds phospholipid \rightarrow \uparrow PTT
- Addition of phospholipid corrects prolonged PTT back to normal



Lupus Anticoagulant

Mixing Study

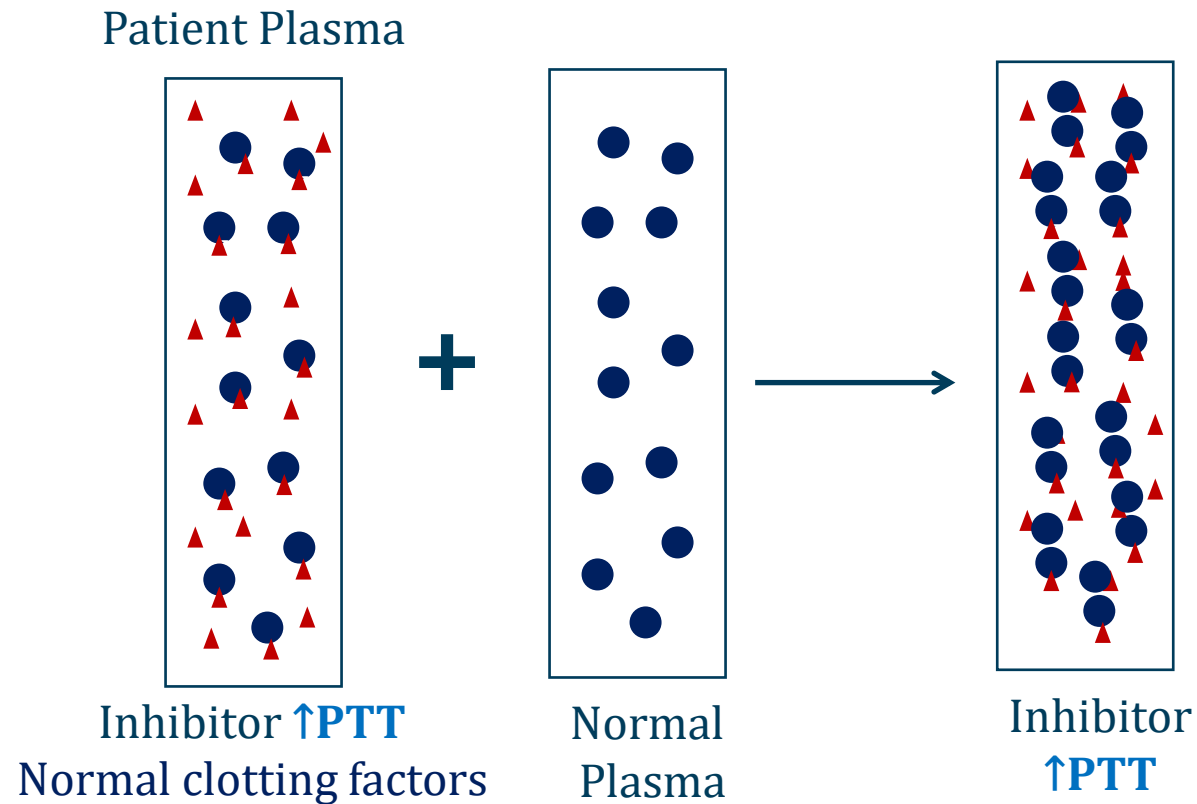
- Abnormal PTT will not correct with mixing study



Lupus Anticoagulant

Mixing Study

- Abnormal PTT will not correct with mixing study



Lupus Anticoagulant

Other Tests

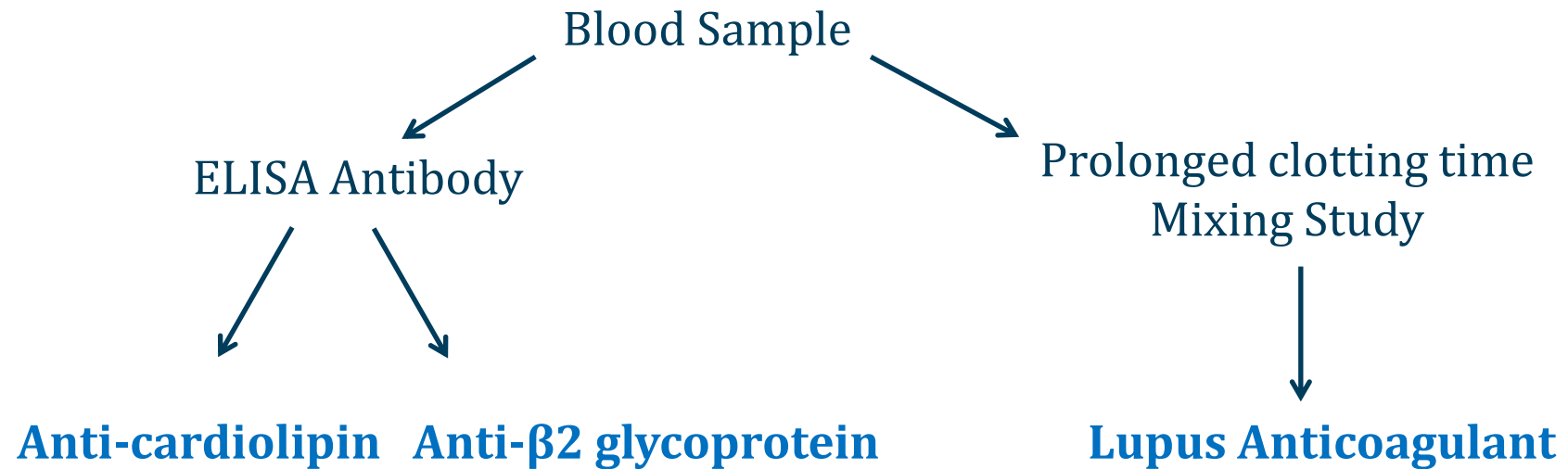
- Only ~ 50% patients with lupus anticoagulant have \uparrow PTT
- Dilute Russell viper venom time (DRVVT) may be used
- Time to clot in presence of Russell's viper venom (activates factor X)
- Time to clot will be prolonged if lupus anticoagulant present
- Time to clot will not correct with mixing study

Russell's Viper



Antiphospholipid Antibodies

Testing



Antiphospholipid Syndrome

Diagnosis

- Antibodies alone do not meet diagnostic criteria
- Syndrome = one laboratory plus one clinical criteria
- **Lab criteria (2 positive results > 12 weeks apart)**
 - Lupus anticoagulant
 - Anti-cardiolipin
 - Anti- β 2-glycoprotein
- **Clinical criteria**
 - Arterial or venous thrombosis
 - Unexplained fetal death after 10 weeks
 - ≥ 3 consecutive fetal losses before 10 weeks



Antiphospholipid Syndrome

Treatment

- Patients with no thrombotic event: usually no treatment
- Acute thromboembolism: standard therapy
- Long-term after acute thromboembolism
 - VTE: warfarin
 - Arterial embolism: warfarin +/- aspirin
- Avoid estrogen-containing contraceptives

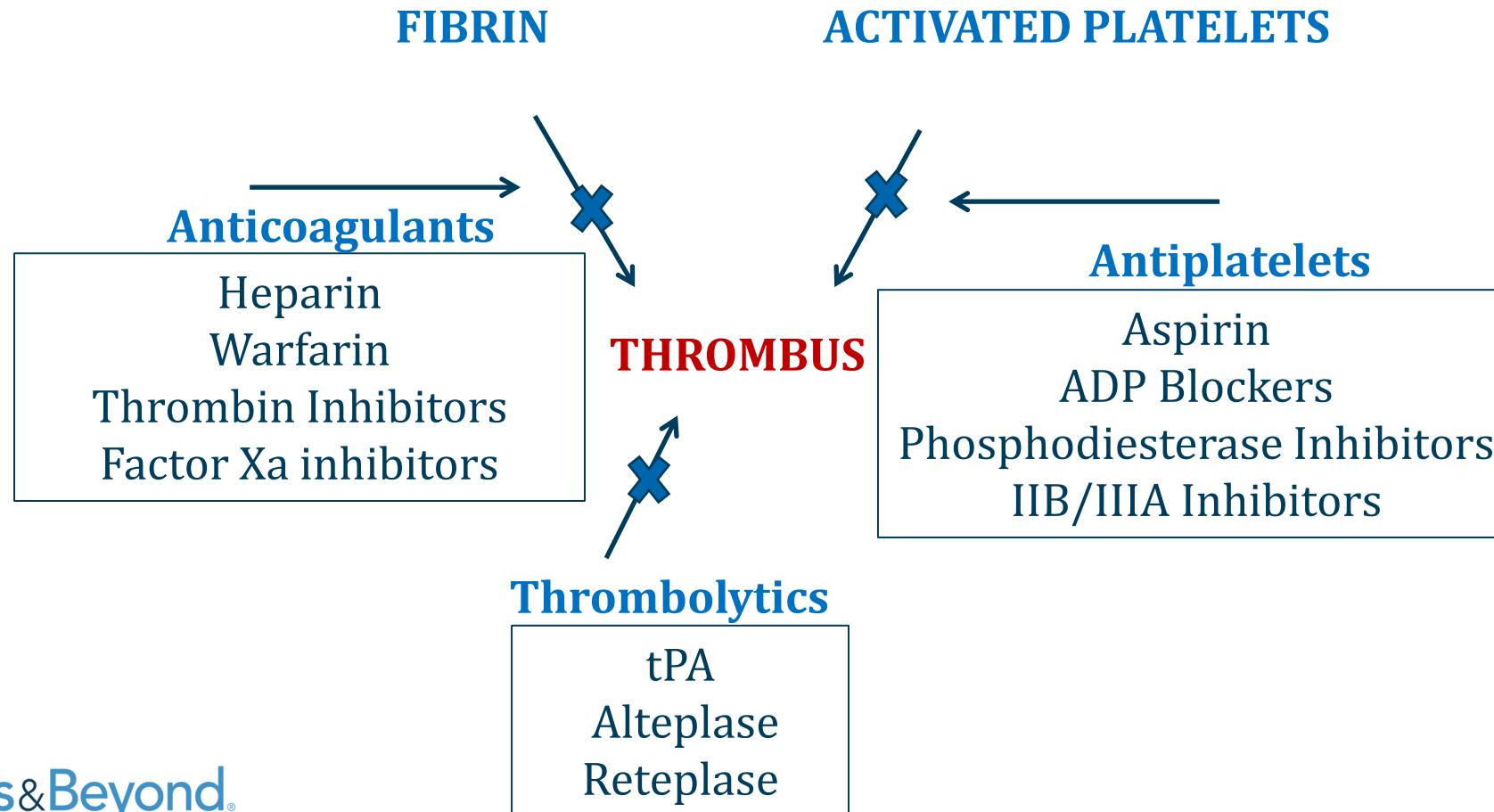


Antiplatelet Drugs

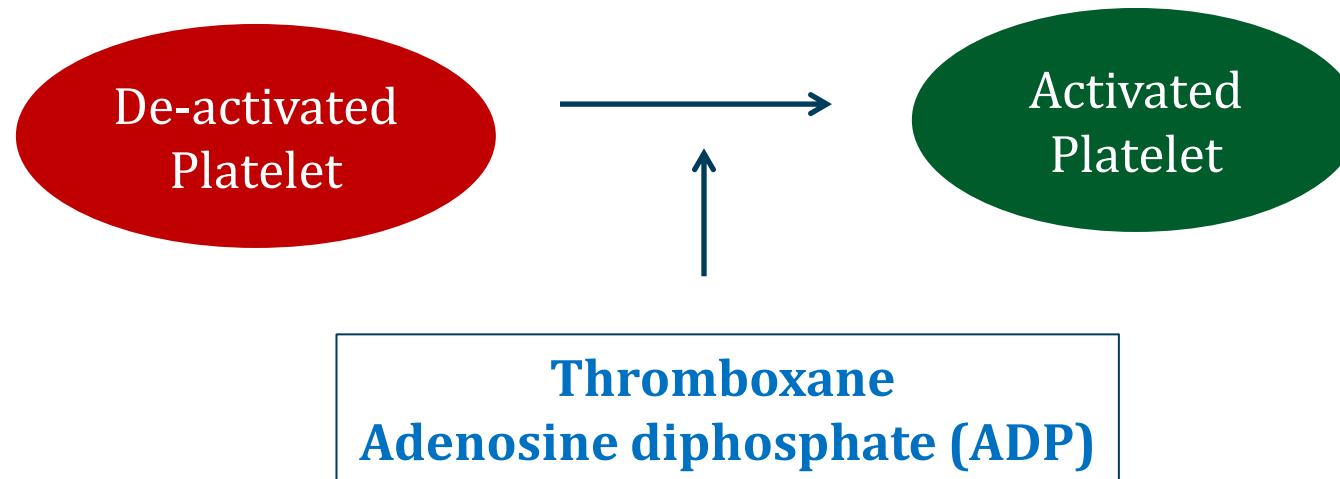
Jason Ryan, MD, MPH



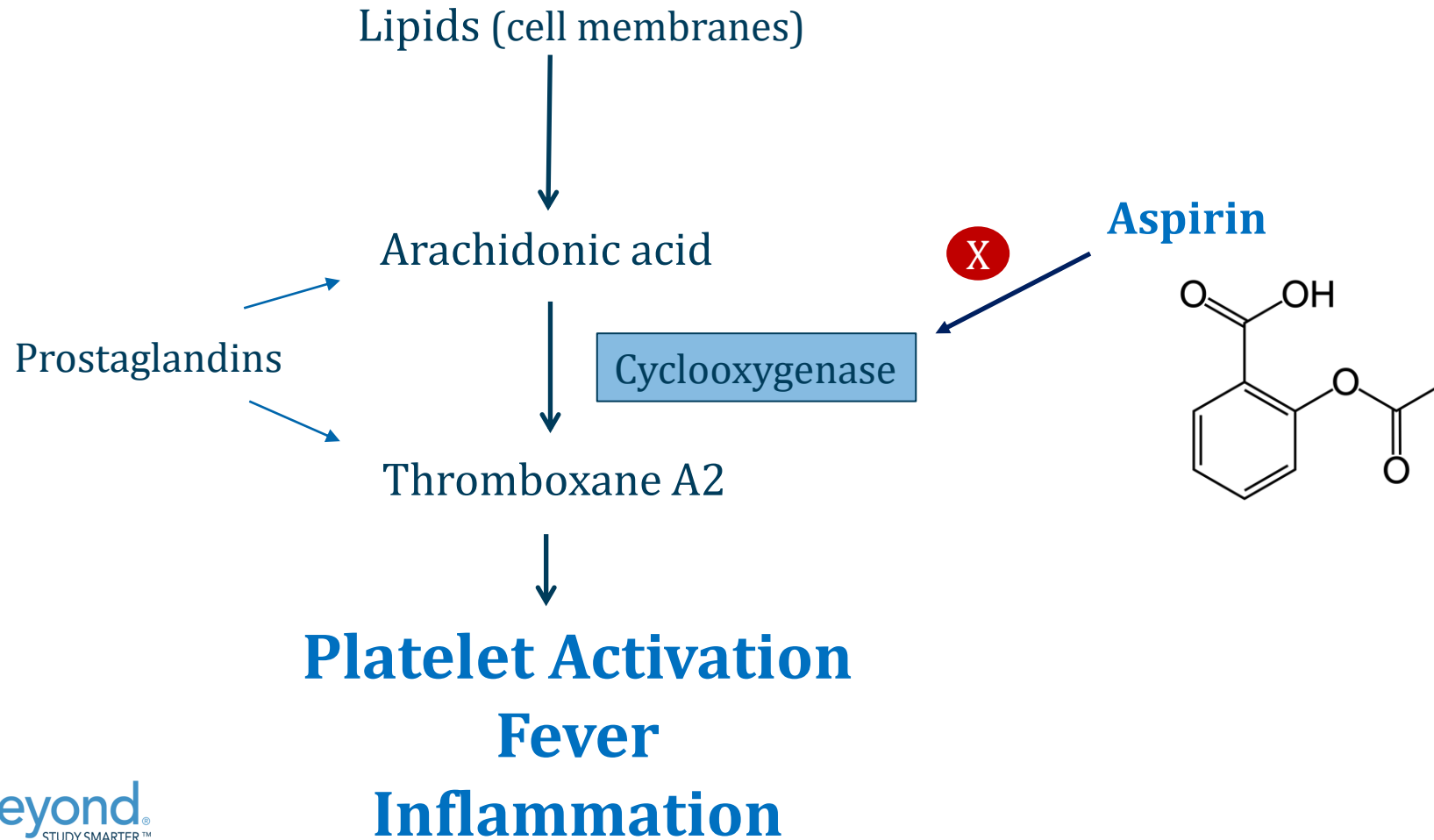
Thrombus Formation



Antiplatelets



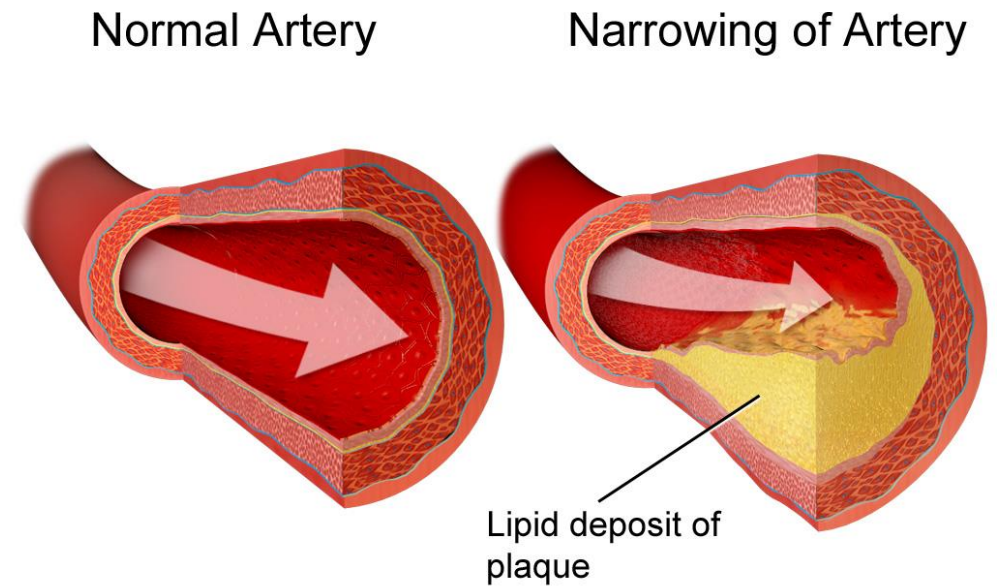
Aspirin



Aspirin

Clinical uses

- **Platelet inhibitor**
 - Coronary artery disease, stroke
 - Low dosages effective (75 to 81mg/day)
- Antipyretic and analgesic
 - Moderate doses (650 mg to 4 g/day)
- Anti-inflammatory
 - High doses (4 to 8 g/day)



Coronary Artery Disease

Aspirin

Adverse Effects

- **Bleeding**
- Gastritis/Ulcers
 - COX important for maintenance of GI mucosa
- **Reye's syndrome**
 - Liver failure and encephalopathy
 - Associated with aspirin use in children
 - Aspirin not generally used in children
 - Exception: Kawasaki disease



NSAIDs

Ibuprofen, naproxen, indomethacin, ketorolac, diclofenac

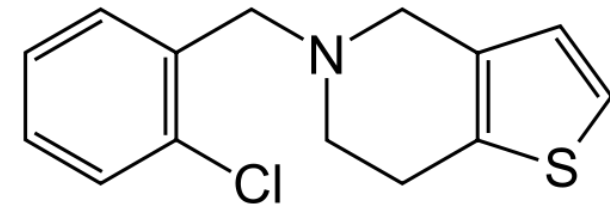
- Nonsteroidal anti-inflammatory drugs
- NSAIDs *reversibly* inhibit COX-1 and COX-2
- Aspirin irreversibly inhibits COX-1 and COX-2
- Decreases activity for lifetime of platelet (7-10 days)
- All NSAIDs may cause bleeding and peptic ulcers
- All NSAIDs reduce pain and inflammation via ↓ PGs



Thienopyridines

Ticlopidine, clopidogrel, prasugrel

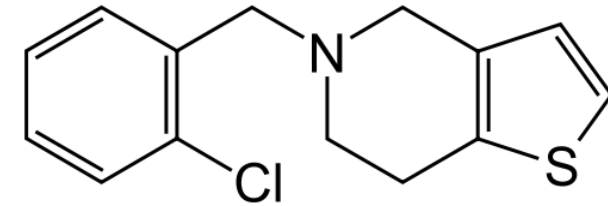
- Irreversible blockers of **P2Y₁₂ receptor**
 - Binds ADP on platelets
- Block effects of ADP on platelets
- Used in aspirin allergy
- Added to aspirin for prevention of MI and stroke
- Used after coronary stent implantation
- Major adverse effect is **bleeding**



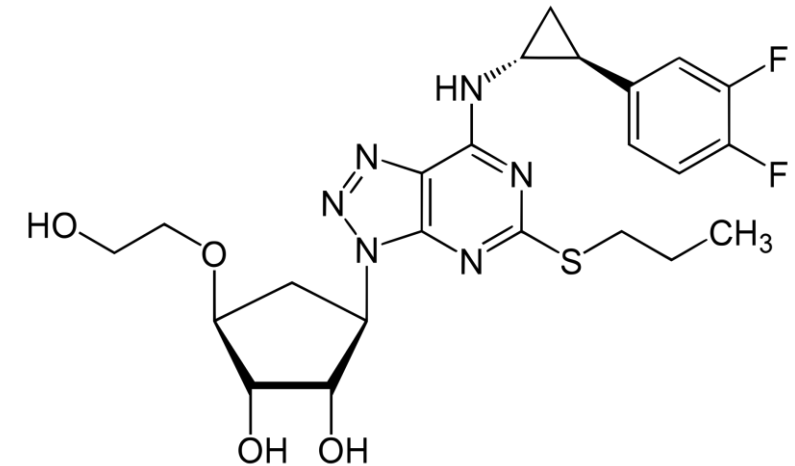
Ticlopidine

Ticagrelor

- Similar action to thienopyridines
- Different structure - NOT a thienopyridine
- Reversible antagonist to P2Y₁₂ receptor
- Unique side effect: **dyspnea**
 - Occurs in more than 10% of patients
 - No hypoxemia
 - Normal lung exam and pulmonary testing
 - Likely sensory - mechanism unclear
 - Can lead to discontinuation of therapy



Ticlopidine

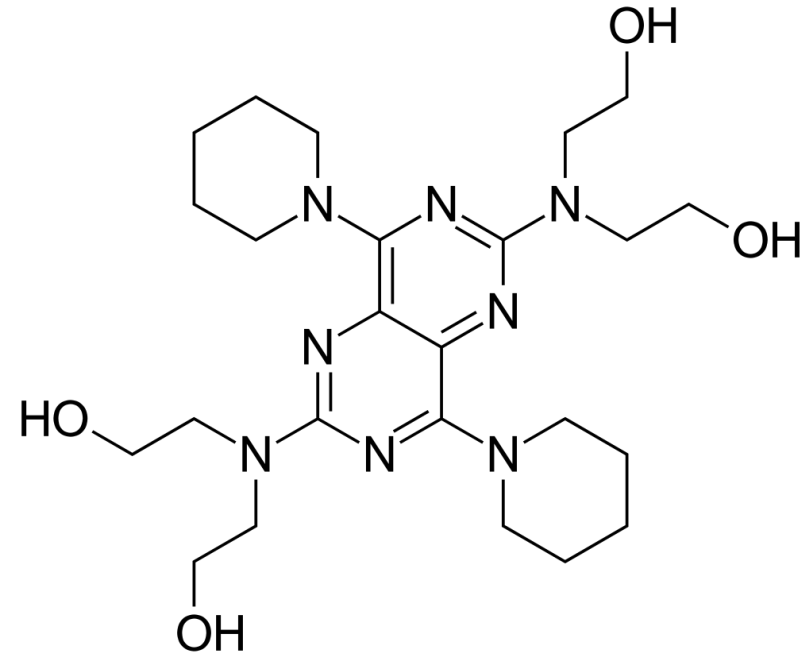


Ticagrelor

Phosphodiesterase Inhibitors

Dipyridamole, cilostazol

- Inhibit phosphodiesterase III in platelets
 - Breaks down cAMP
 - \uparrow cAMP \rightarrow \downarrow platelet activation
- Also vasodilators
 - \uparrow cAMP in vascular smooth muscle
- Rarely used as antiplatelet drugs
- Often used for vasodilation effects

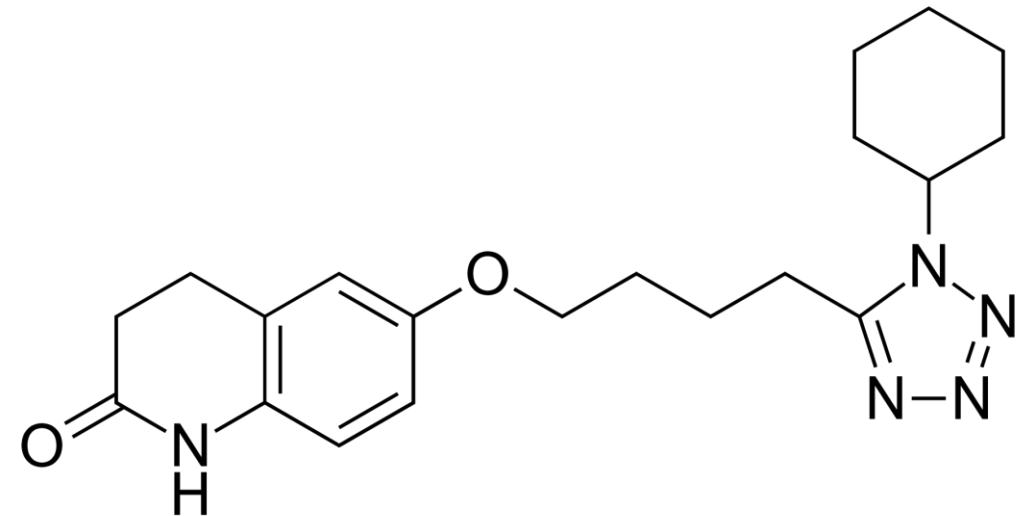


Dipyrimadole

Phosphodiesterase Inhibitors

Dipyridamole, cilostazol

- **Dipyrimadole**
 - Used with aspirin for stroke prevention (antiplatelet)
 - Used in chemical cardiac stress testing (vasodilator)
- **Cilostazol**
 - Vasodilator
 - Used in peripheral arterial disease



Cilostazol

Phosphodiesterase Inhibitors

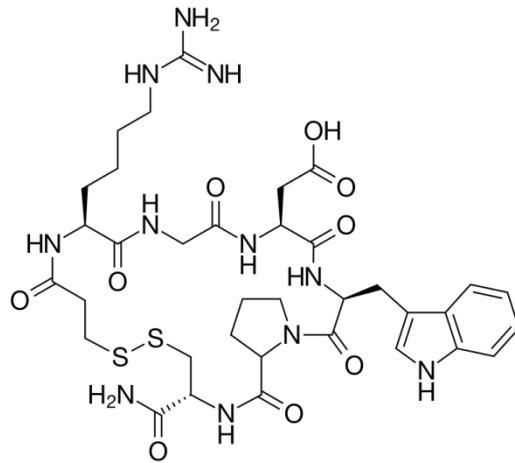
Adverse effects

- Related to vasodilation
- Flushing
- Dizziness
- Headache

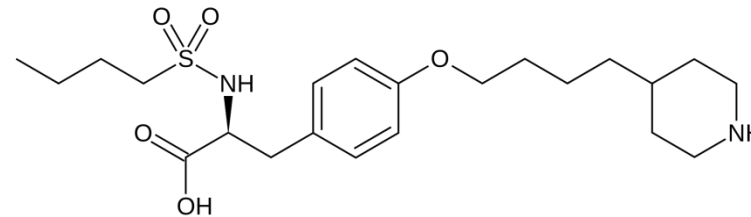


IIB/IIIA Receptor Blockers

- Abciximab, eptifibatide, tirofiban
- Bind and block IIB/IIIA receptors
- IV drugs used in acute coronary syndromes/stenting



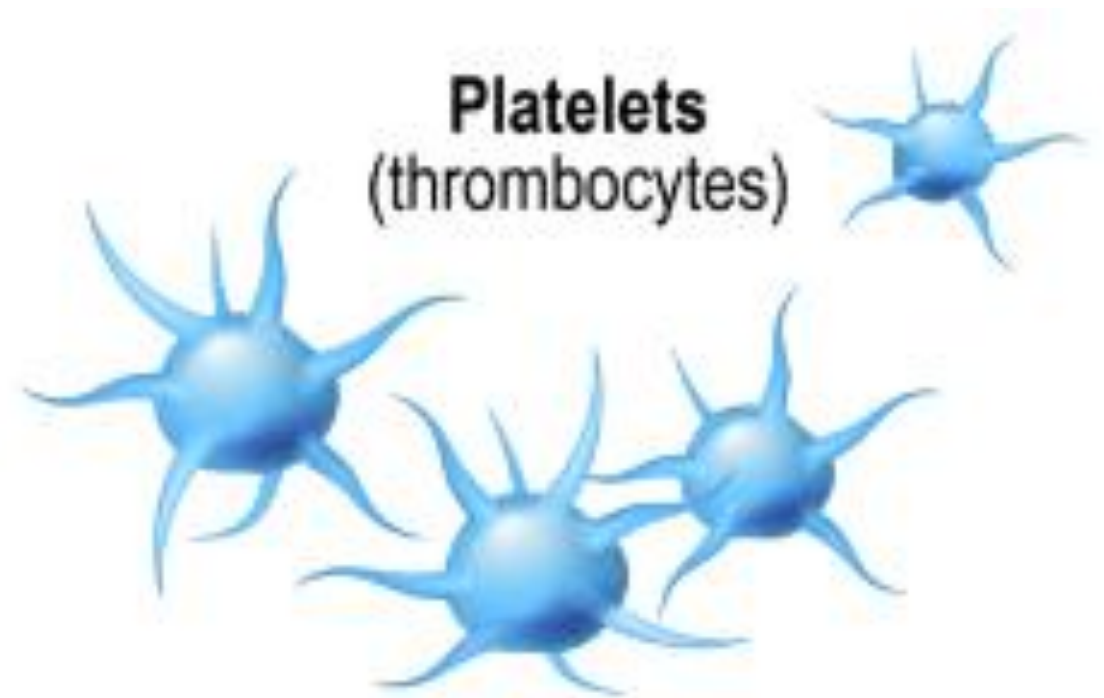
Eptifibatide



Tirofiban

IIB/IIIA Receptor Blockers

- Main adverse effect is bleeding
- Can cause **thrombocytopenia**
 - May occur within hours of administration
 - Mechanism poorly understood
 - Platelet count monitored after administration

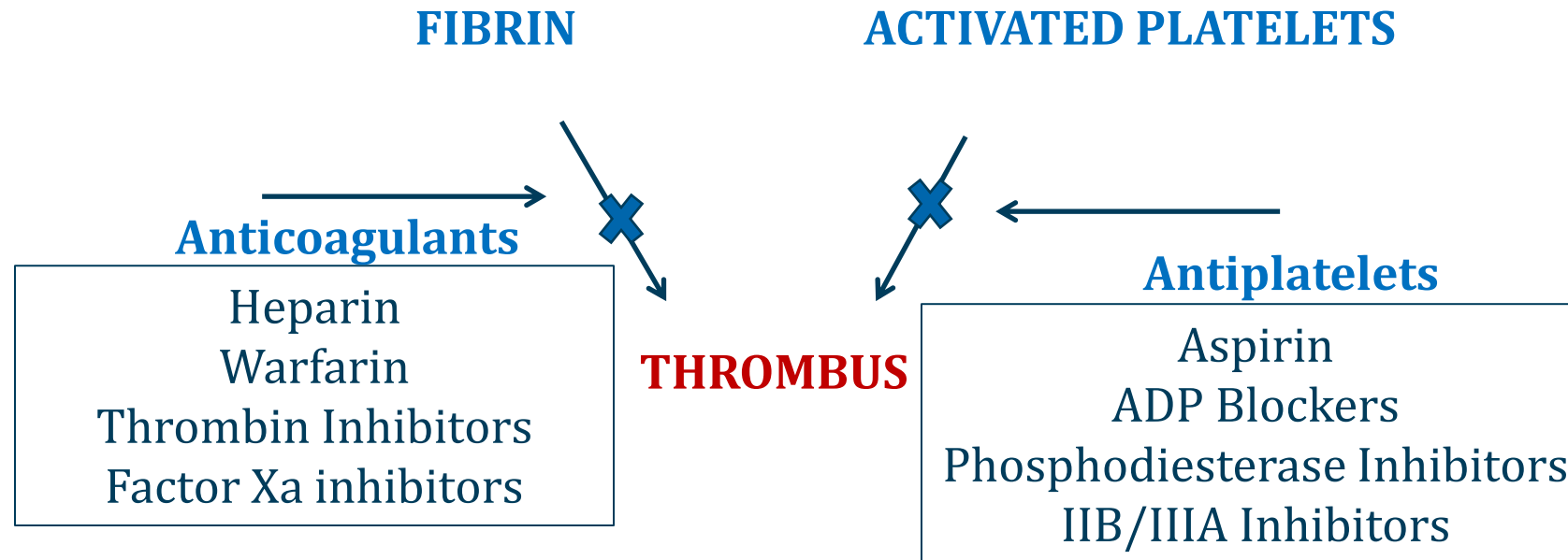


Anticoagulants

Jason Ryan, MD, MPH

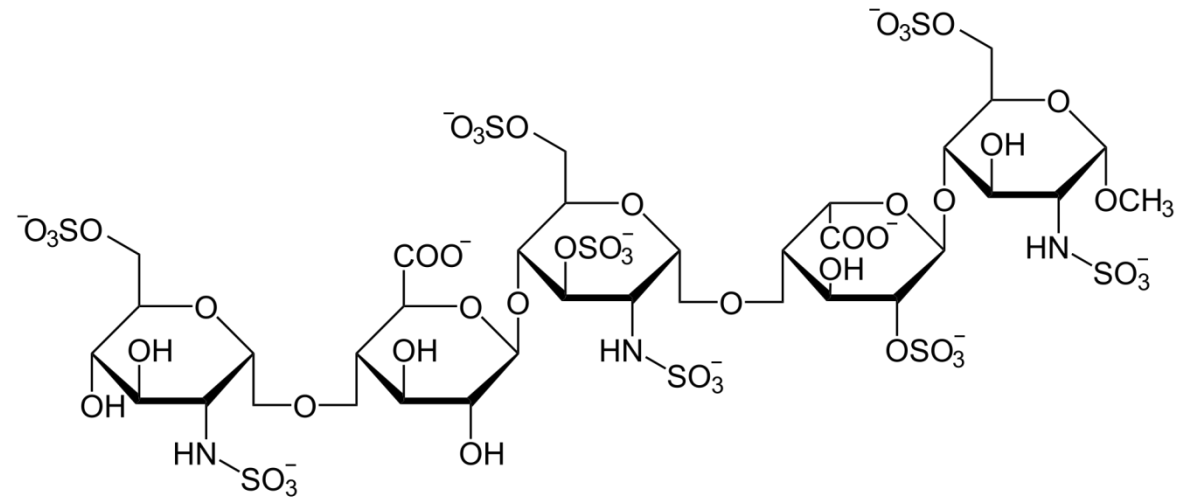


Thrombus Formation

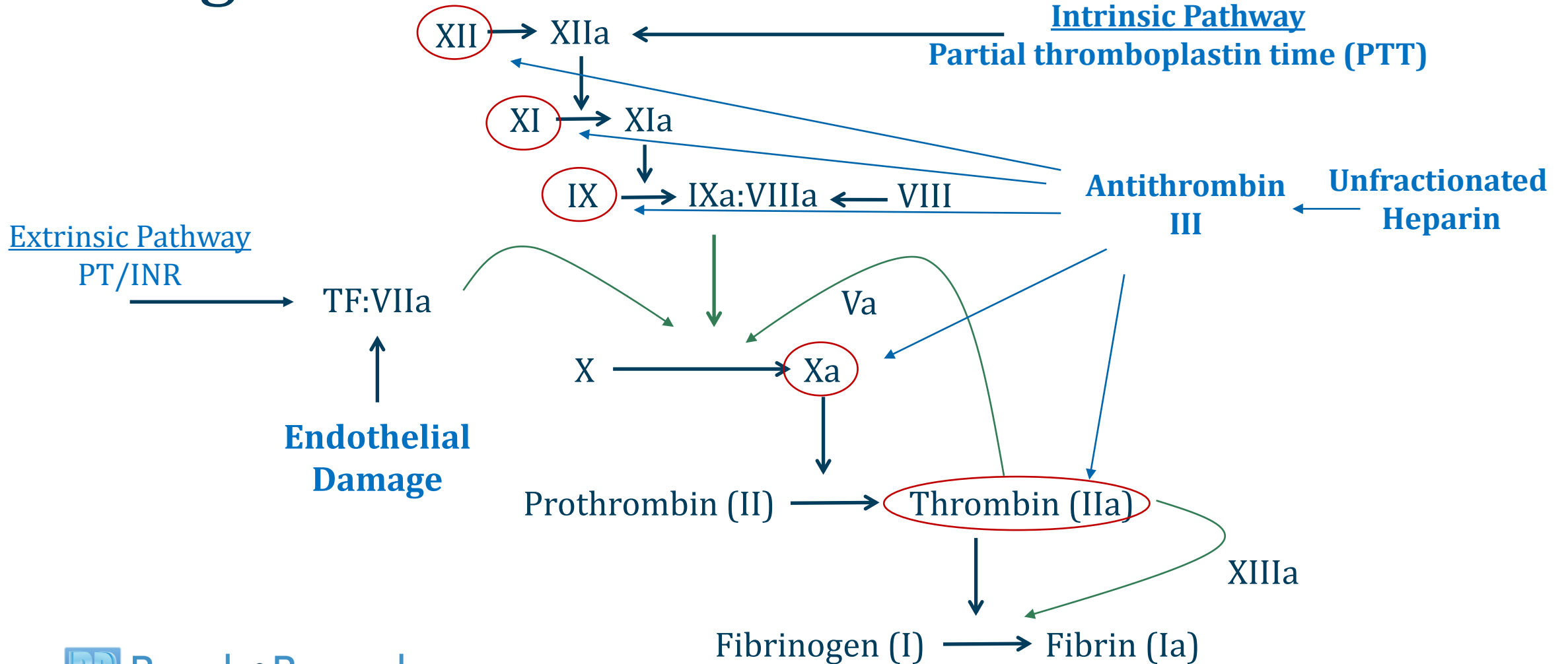


Heparin

- Polymer
- Occurs naturally in mast cells
- Molecules with varying chain lengths
- Unfractionated: widely varying polymer chain lengths
- Low molecular weight: smaller polymers only



Coagulation Cascade



Unfractionated Heparin

- Given IV or SQ → rapid onset
- **Increases partial thromboplastin time (PTT)**
- May increase prothrombin time (PT) at high dosages
- Lots of binding to plasma proteins
- Variable response from patient to patient
- Dose must be adjusted to reach goal PTT



Protamine

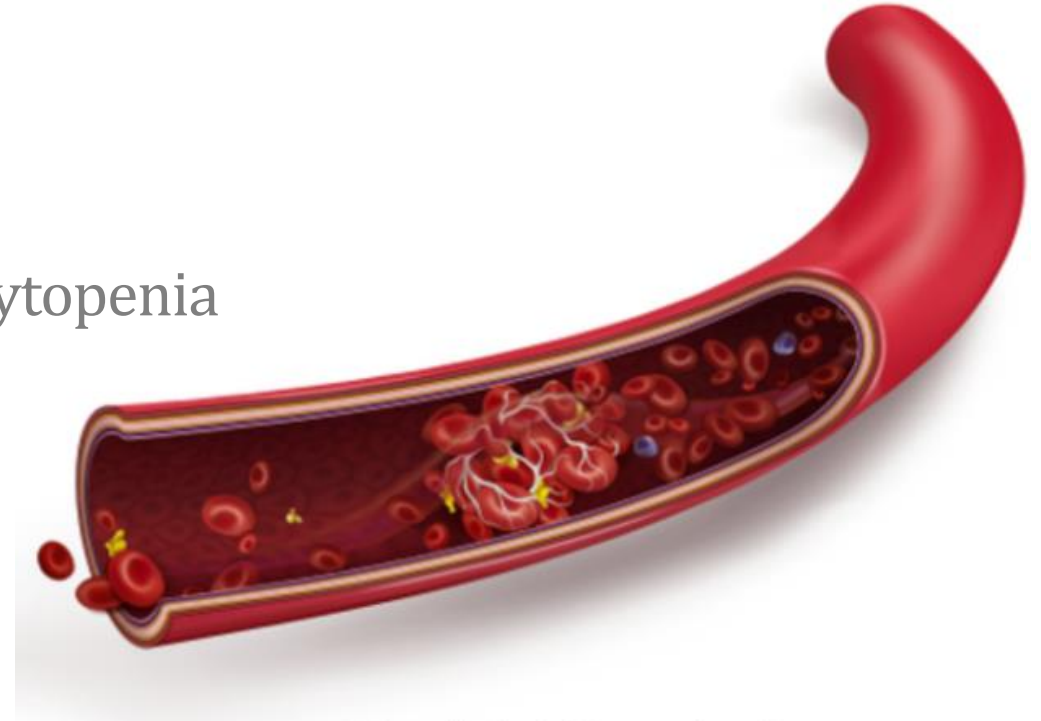
- Reversal agent for unfractionated heparin
- Binds heparin → neutralizes drug
- Used in heparin overdose
- Used in cardiac surgery
 - High-dose heparin administered for heart-lung bypass
 - Quick reversal at completion of case
- Less effective with LMWH



Unfractionated Heparin

Clinical uses and adverse effects

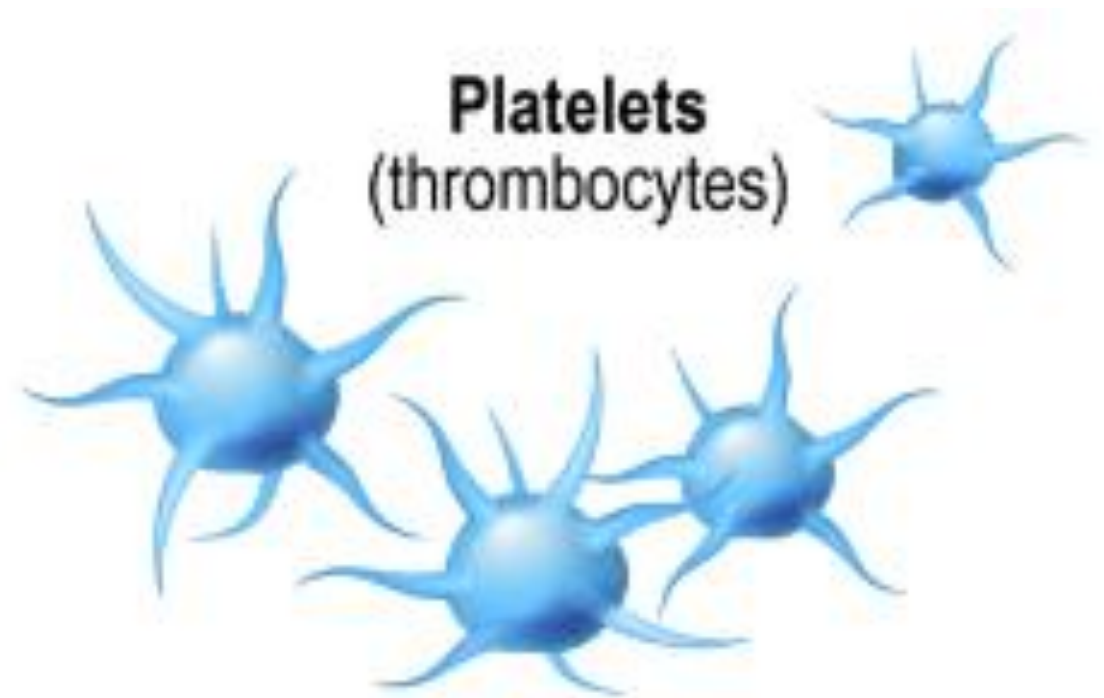
- Acute management of thrombotic disorders
 - DVT/PE
 - Acute coronary syndromes
 - Stroke
- Prophylaxis for DVT in hospitalized patients
- Major adverse effect: **bleeding**
- Rarely may cause heparin-induced thrombocytopenia



Blood clot (thrombus)

Heparin and Thrombocytopenia

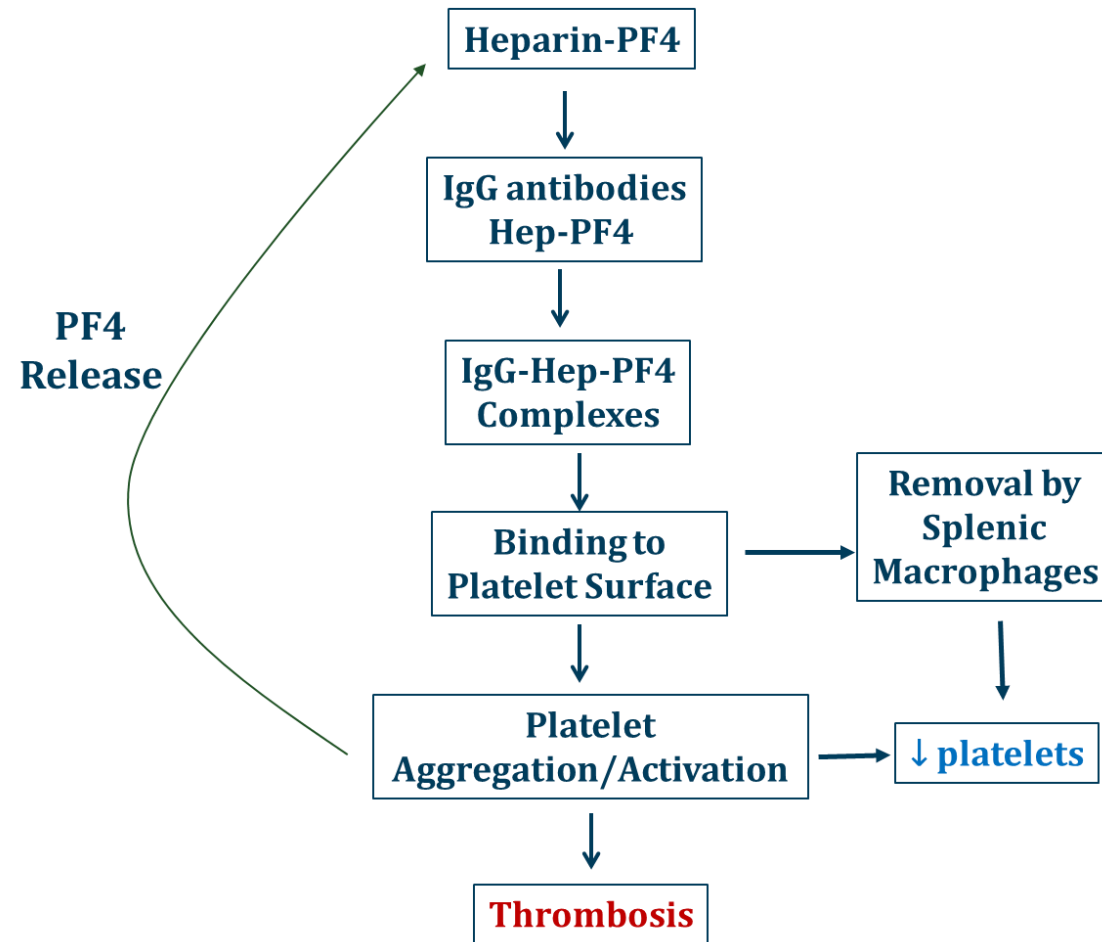
- “Non-immune” thrombocytopenia
 - Heparin-induced thrombocytopenia type I
 - Mild (10-20%) ↓ platelets
 - Direct suppressive effect on platelet production
 - May continue heparin



Heparin and Thrombocytopenia

- **Heparin-induced thrombocytopenia (HIT) type II**
 - Immune-mediated reaction
 - Antibodies bind platelet factor 4-heparin
 - Splenic destruction of platelets
 - Causes thrombocytopenia and thrombosis
 - Rare: 0.2 – 5% heparin patients
- Presents as **drop in platelets plus thrombosis**

Heparin-Induced Thrombocytopenia



Heparin-Induced Thrombocytopenia

Diagnostic criteria - 4 T's

- **Timing**: occurs 5-10 days after first exposure to heparin
 - Earlier if prior exposure
- **Thrombocytopenia**: fall in platelets **more than 50% from baseline**
- **Thrombosis**: arterial or vein thrombosis
 - Often causes **skin necrosis**
 - Existing clots may extend on heparin
- **O**ther causes thrombocytopenia must not be present
- More criteria = HIT more likely
- Can calculate a 4T score

CRITERIA

Heparin-Induced Thrombocytopenia

Diagnosis and management

- Presumptive diagnosis made clinically based on 4 T's
- **Stop heparin if high suspicion**
- Patients with HIT must use alternative drugs
- Usually **argatroban**: direct thrombin inhibitor
- Alternatives: bivalirudin or fondaparinux



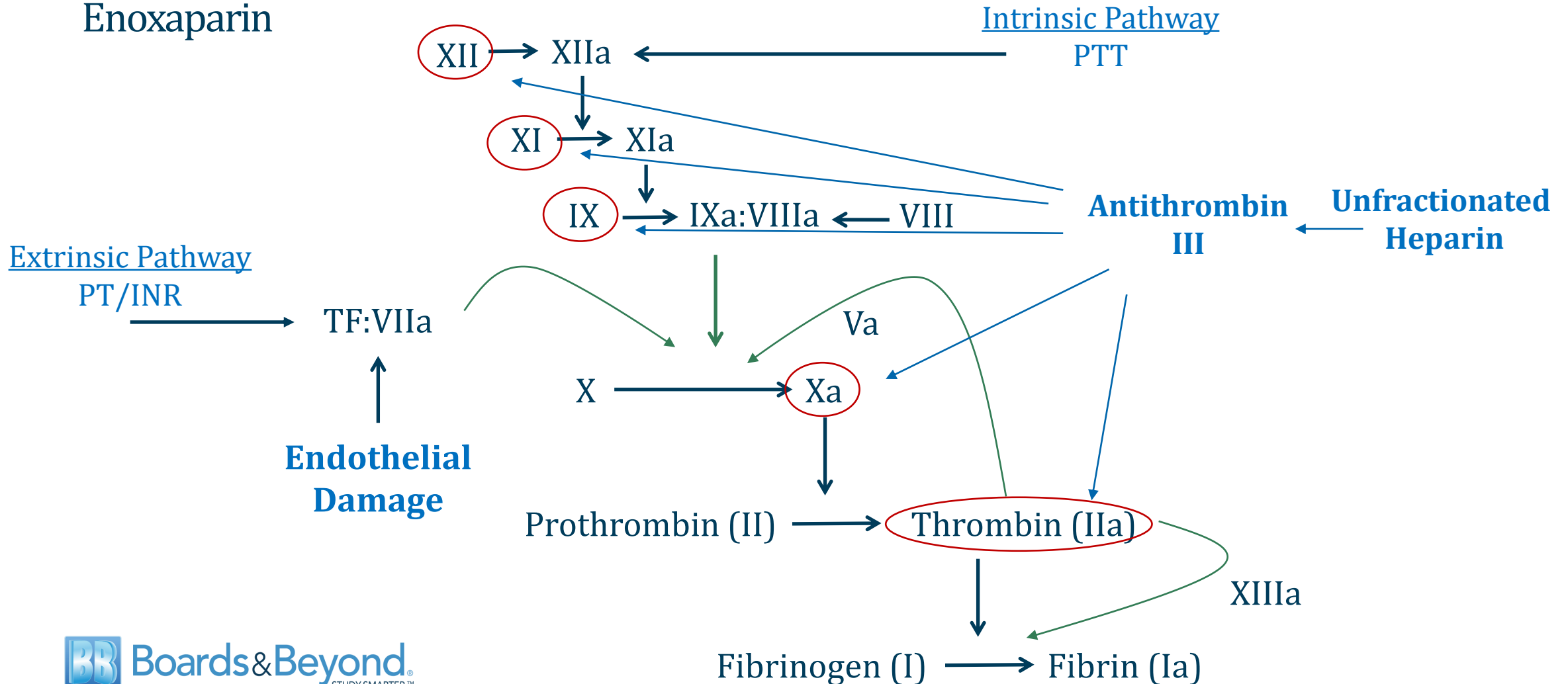
Heparin-Induced Thrombocytopenia

Diagnosis and management

- Definitive diagnosis: antibody testing or functional testing
- **HIT antibody testing**
 - Autoantibodies to platelet factor 4 complexed with heparin
 - Widely available but can have false positives
- Functional assays
 - Serotonin release assay – gold standard
 - Tests ability of PF4-heparin antibody to activate platelets
 - More difficult to obtain but more specific for diagnosis
- Usually diagnosis made if clinical suspicion high + positive HIT ab

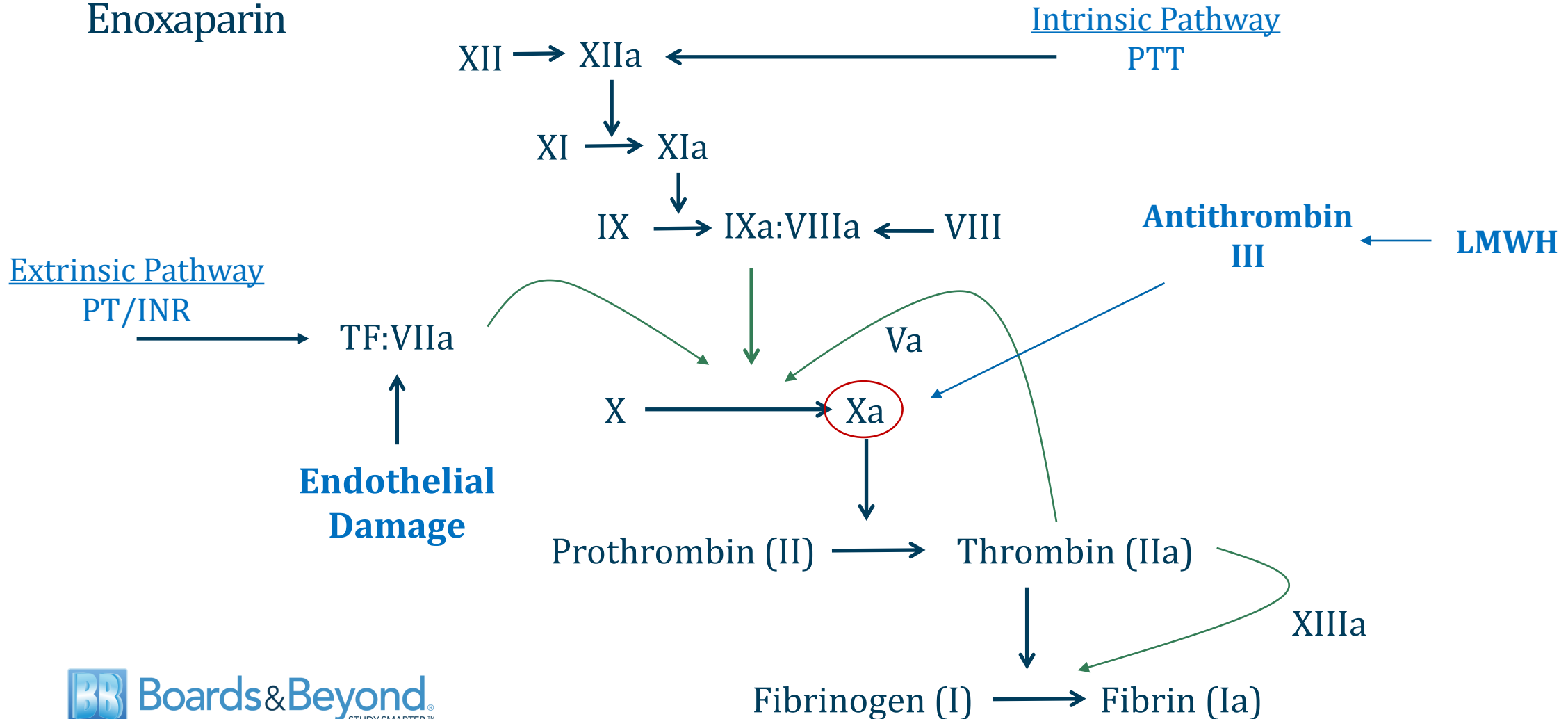
Low Molecular Weight Heparin

Enoxaparin



Low Molecular Weight Heparin

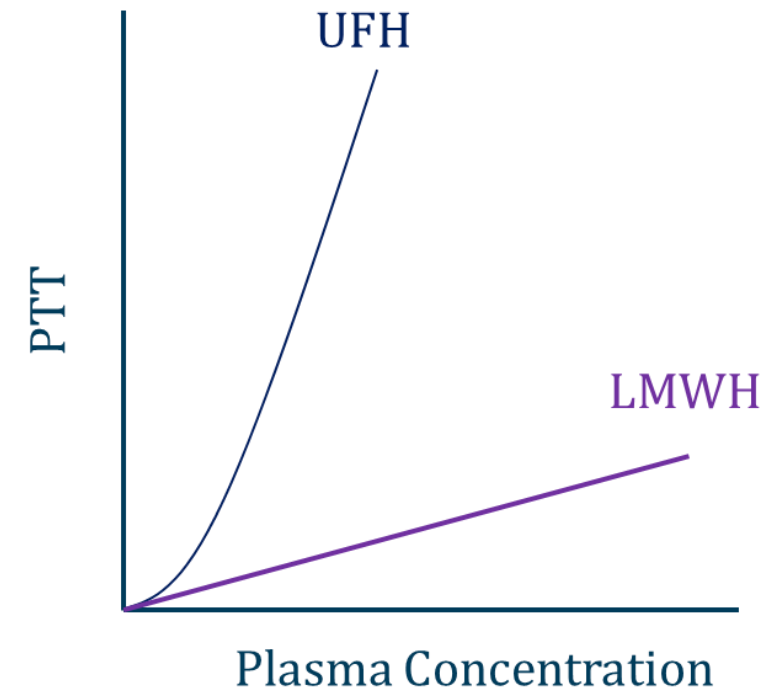
Enoxaparin



Low Molecular Weight Heparin

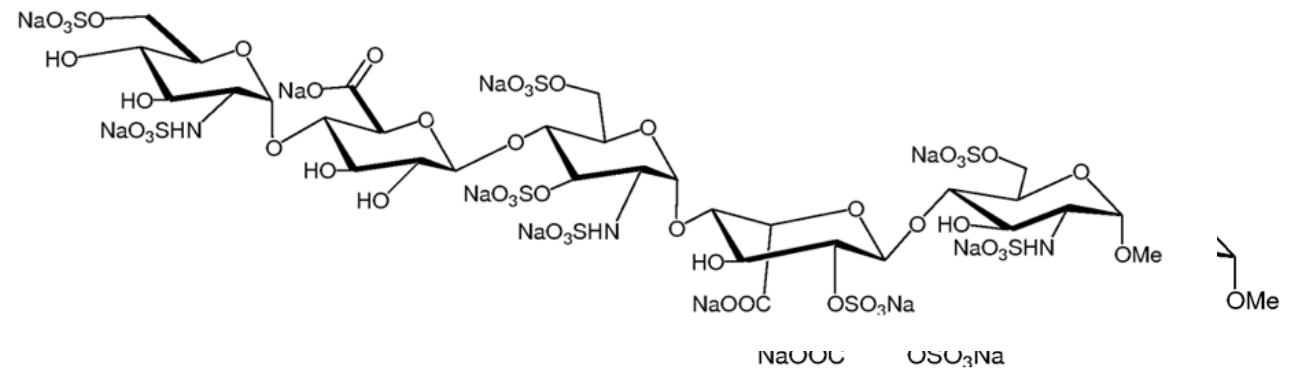
Enoxaparin

- Dose based on patient weight – usually no titrating
 - Reduced binding to plasma proteins and cells
- Lower incidence of HIT (but may still cause)
- If monitoring required, must check **anti Xa levels**
 - Level of anti-Xa substances in blood
 - Cannot use PTT
- Monitoring indications:
 - **Extreme obesity**
 - **Renal insufficiency**
 - Unpredictable effects of LMWH



Fondaparinux

- Inhibits factor Xa via antithrombin III
- Alternative to low molecular weight heparin
- Used in special circumstances
- Prevention of DVT after surgery or during hospitalization
- Some acute coronary syndromes
- Can be used in HIT



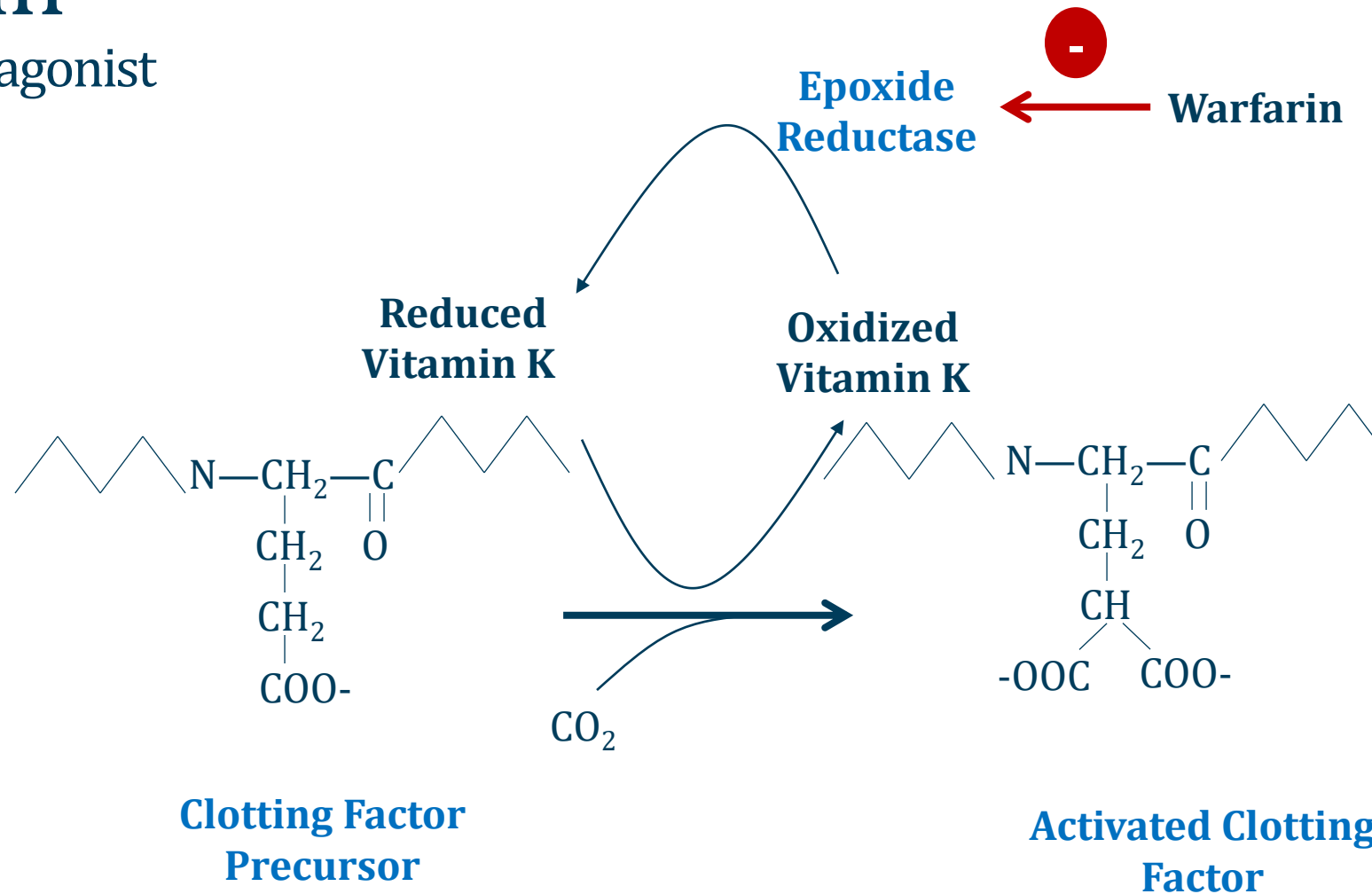
Warfarin

- Antagonist to **vitamin K function in liver**
 - Inhibits activation of clotting factors by gamma carboxylation
 - Loss of ability to activate clotting factors
- Vitamin K dependent factors: II, VII, IX, and X
 - Also anti-clotting factors protein C and S
- Causes ↓ levels of all vitamin K dependent factors



Warfarin

Vitamin K Antagonist



Warfarin

Clinical effects

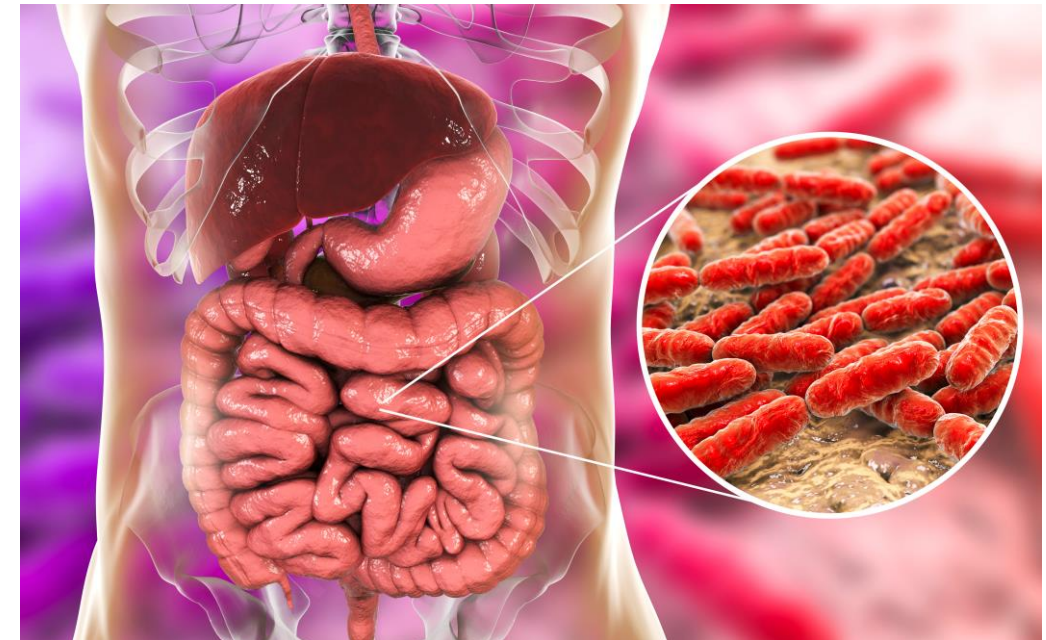
- Takes days to achieve its effects
- Time required for clotting factor levels to fall
- Dose adjusted to reach target **PT/INR**
- Factor VII levels alter prothrombin time
- PT/INR most sensitive to warfarin



Warfarin

Clinical effects

- Vitamin K comes from **diet** and **GI bacteria**
- Warfarin effect varies with diet
- Green vegetables → ↑ vitamin K
- Antibiotics may ↓ GI bacteria → ↓ vitamin K



Warfarin

Prothrombotic effect

- Protein C: anti-clotting factor
 - Also vitamin K dependent
 - Short half-life
- **Brief pro-thrombotic effect**
 - Protein C deficiency
 - Eventually other factors fall → antithrombotic
- Use heparin or another drug when starting warfarin
 - Especially if clot present (DVT/PE)
 - Warfarin alone may extend clot

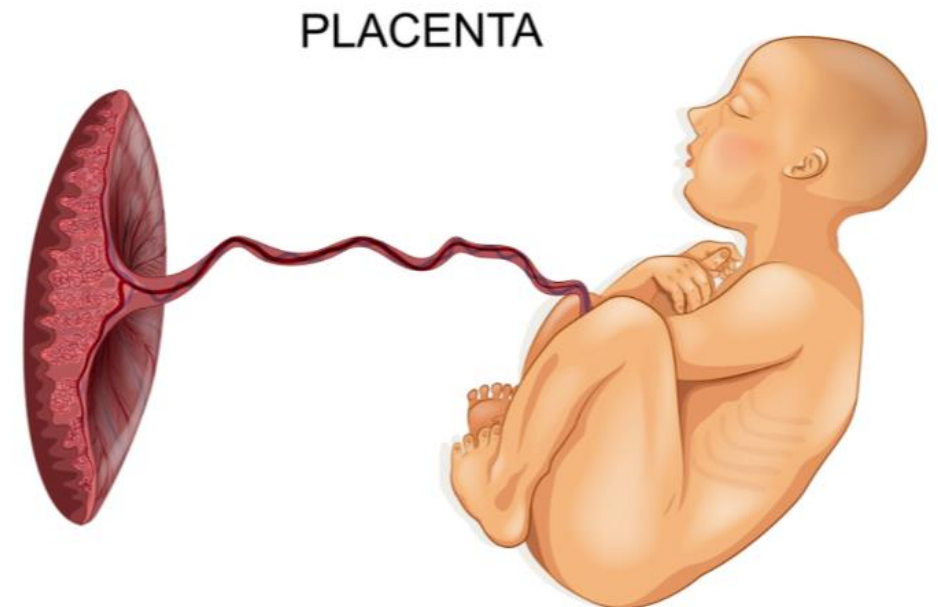


Blood clot (thrombus)

Warfarin

Adverse effects

- **Crosses placenta**
 - Avoided in pregnancy
 - Fetal warfarin syndrome: abnormal fetal development
 - Heparin often used (does not cross placenta)
- Mainly adverse effect is **bleeding**
- Rarely causes skin necrosis



Warfarin

Skin necrosis

- Rare complication of warfarin therapy
- Can also occur with very high dosages
- Occurs in patients with **protein C deficiency**
- Initial exposure to warfarin → ↓ protein C
- Result: thrombosis of skin tissue
- Large dark, purple skin lesions

Warfarin Skin Necrosis



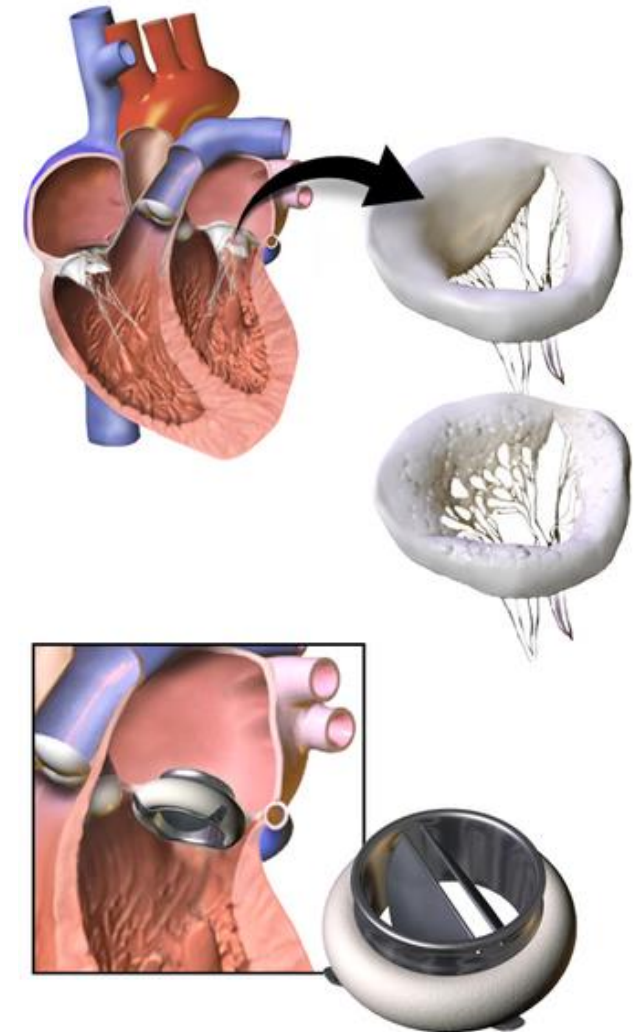
Bakoyiannis C, Karaolani G, Patelis N, Maskanakis A, Tsaples G, Klonaris C, Georgopoulos S, Liakakos T

Warfarin

Clinical uses

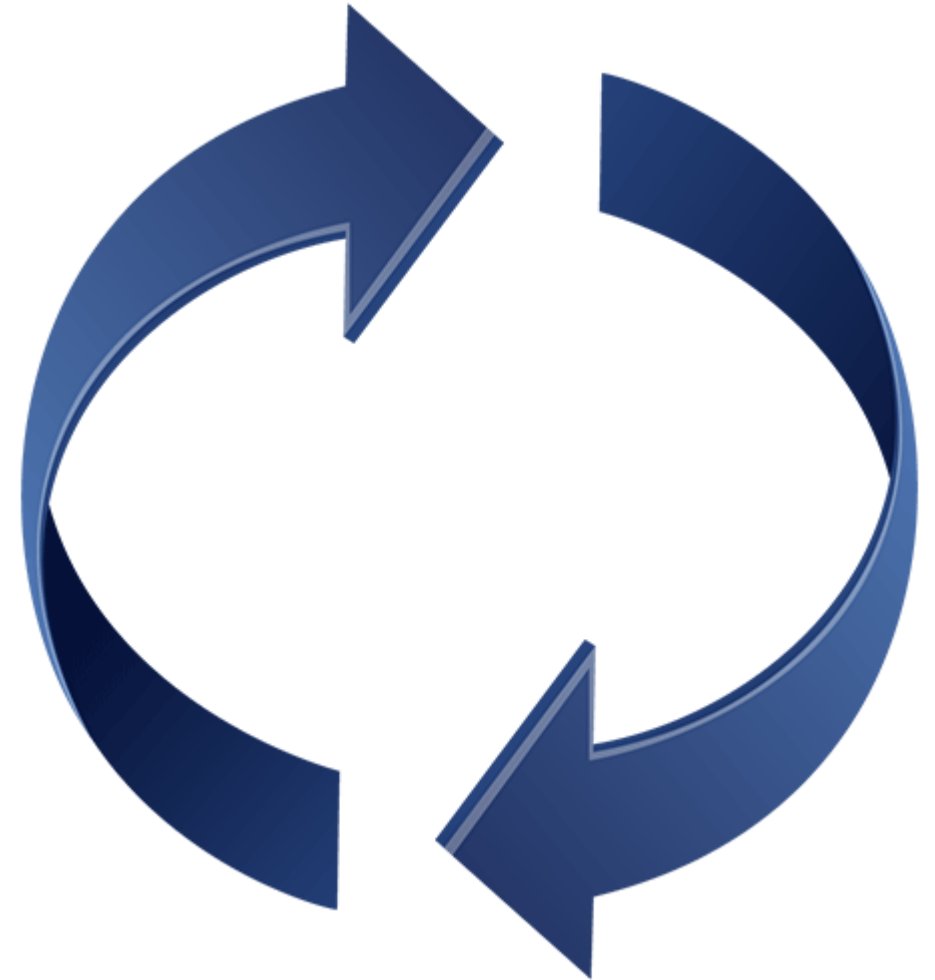
- Stroke prevention atrial fibrillation
- Mechanical heart valves
- DVT/PE
- Preferred oral drug in advanced kidney disease
- Dose titrated to achieve goal INR
- Usually 2 to 3 depending on indication

Mechanical Mitral Valve



Warfarin Reversal

- Indicated if INR too high or bleeding
- **Vitamin K (oral or IV)**
 - Usually oral unless major bleeding
 - IV: small risk of anaphylaxis
- **Fresh Frozen Plasma (FFP)**
 - Frozen plasma after removal of RBC, WBC, and Plt
 - Replaces clotting factors
- **Prothrombin complex concentrates (PCC)**
 - Mixture of vitamin K-dependent factors
 - Lower risk of adverse events compared to FFP



Warfarin Reversal

- Normal INR: 1.0
- Goal INR usually 2.0 to 3.0
- Mild elevations < 4.5 without bleeding: hold one dose or adjust dose

Indication	Treatment
Active bleeding + INR > 2.0	Hold warfarin + PCC or FFP
Asymptomatic INR > 10	Hold warfarin + oral vitamin K
Asymptomatic INR > 4.5	Hold warfarin +/- oral vitamin K

Chronic Oral Anticoagulation

- Several indications
- Atrial fibrillation
- Mechanical heart valves
- Prior DVT or PE
- Prior standard: warfarin
 - Downside: requires INR checks
- Newer alternatives: **DOACs**
 - Do not require INR checks

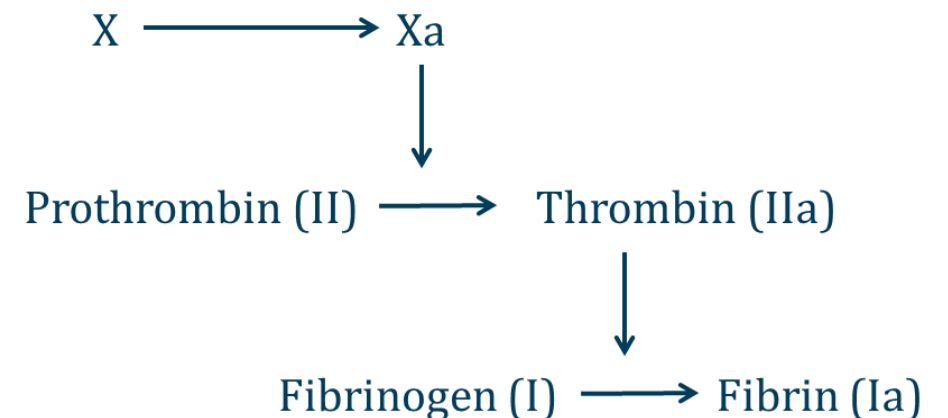
Atrial Fibrillation



Direct Oral Anticoagulants

DOACs

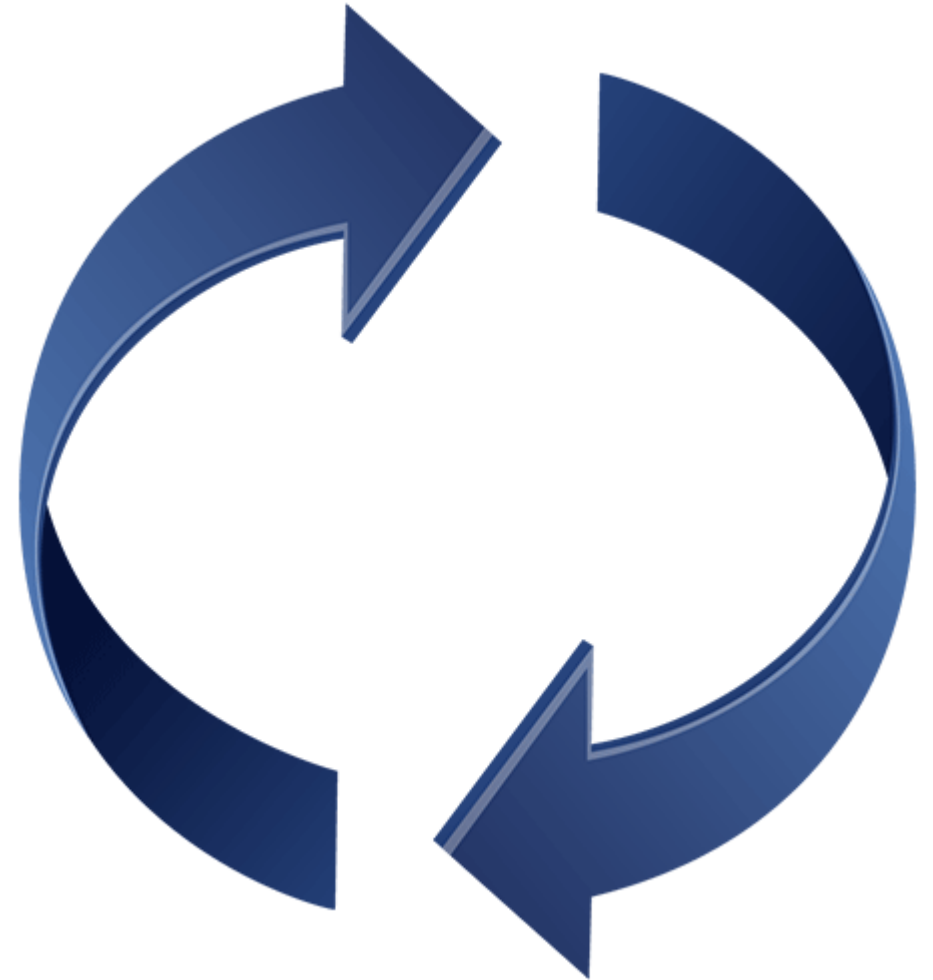
- Oral alternatives to warfarin
- Factor Xa inhibitors: **rivaroxaban** and **apixaban**
- Thrombin inhibitor: **dabigatran**
- Standard dosing - **no INR monitoring**
- Can increase PT or PTT
- Major adverse effect: **bleeding**
- Cannot be used in advanced renal failure
 - Warfarin preferred in advanced kidney disease



Direct Oral Anticoagulants

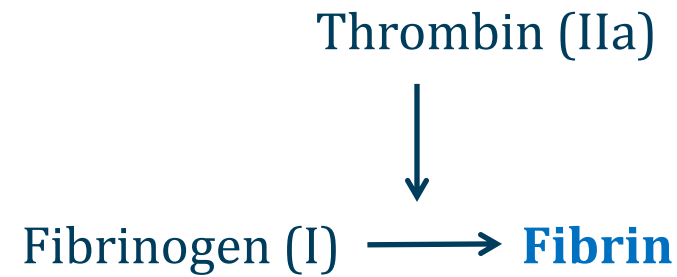
Reversal agents

- Dabigatran: **idarucizumab**
 - Anti-dabigatran monoclonal antibody
- Apixaban and rivaroxaban: **andexanet alfa**
 - Inactive “decoy” of Xa that binds drug
 - Can also use PCC



Direct Thrombin Inhibitors

- Oral: dabigatran (DOAC)
- Parenteral: argatroban, bivalirudin, desirudin

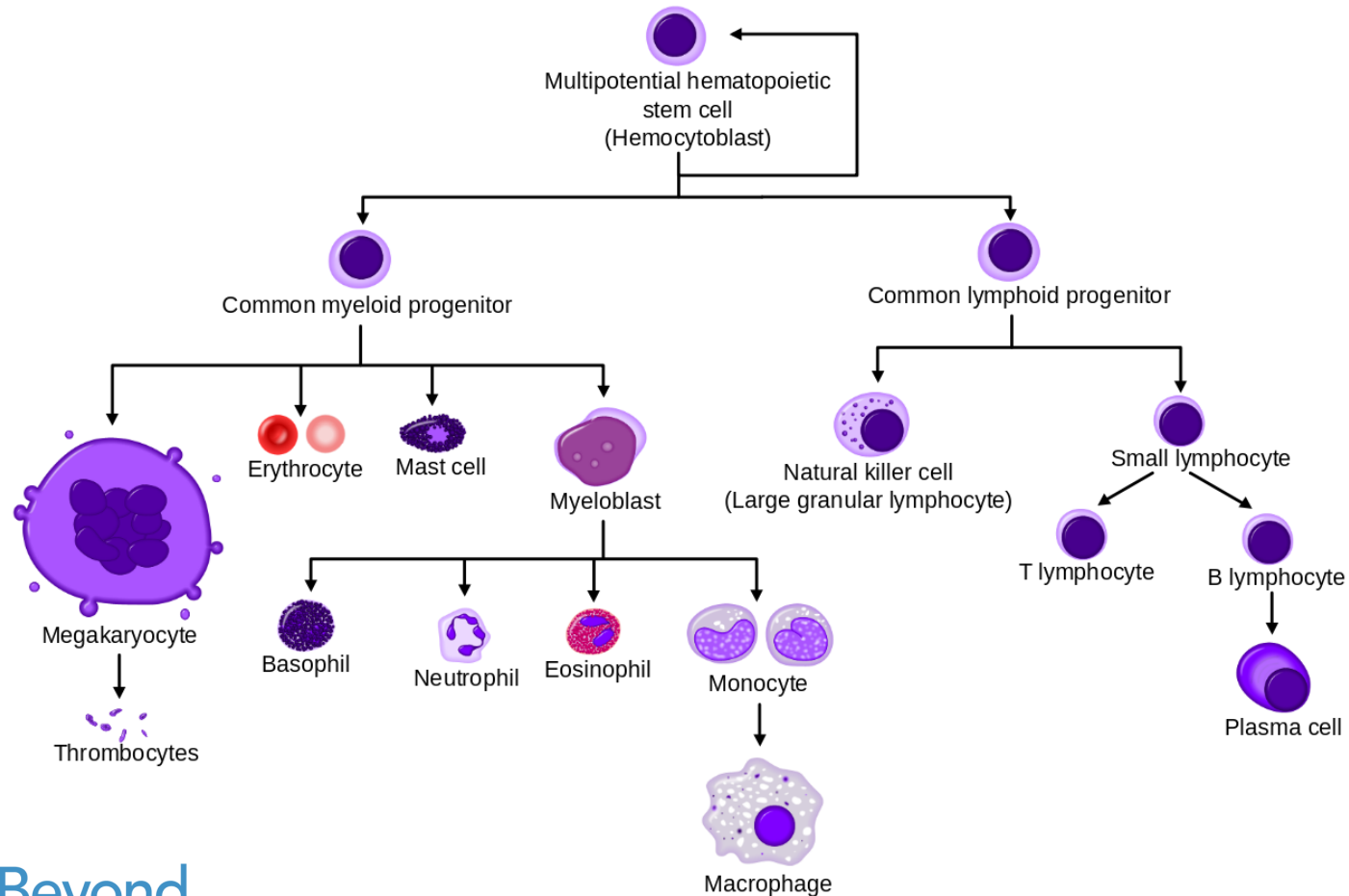


Plasma Cell Disorders

Jason Ryan, MD, MPH

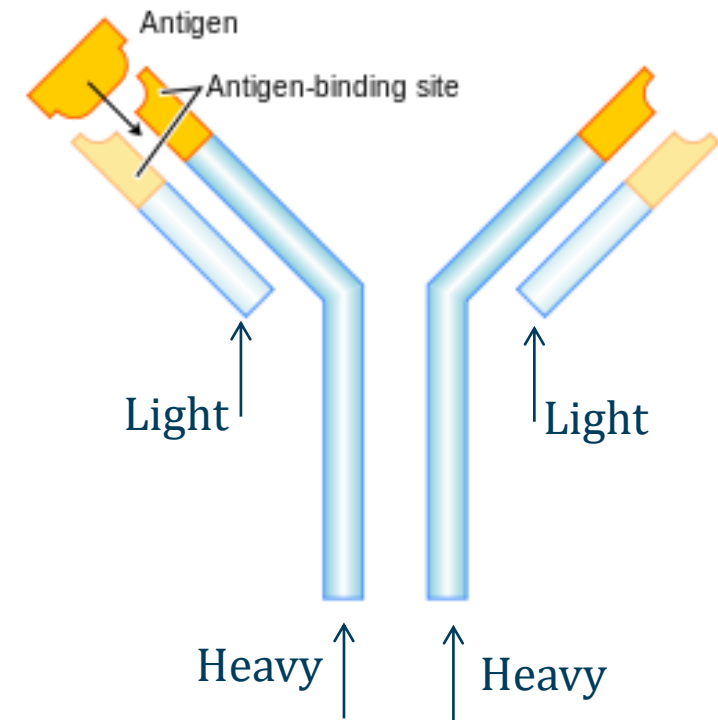


Hematopoiesis



Multiple Myeloma

- Malignancy of **plasma cells**
- Plasma cells produce **monoclonal proteins**
 - Non-functional immunoglobulins
 - Also called “paraproteins”
 - “Monoclonal:” proteins are identical
 - IgG (~50%)
 - IgA (~20%)
 - Light chains only (~15%)
- Median age at diagnosis: 65 to 74 years



Multiple Myeloma

Clinical Features

- **Bone destruction** – major pathologic feature
- **CRAB features**
- Calcium elevation
- Renal failure
- Anemia
- Bone lesions
- Also increased **infection risk**


CRAB Feature	Criteria
Calcium	> 11mg/dL
Renal insufficiency	CrCl < 40mL/min or Cr > 2 mg/dL
Anemia	Hgb < 10 g/dL
Bone disease	Lytic lesions

Multiple Myeloma

Hypercalcemia

- Present in ~ 28% of cases at diagnosis¹
- **Tetany**
- Seizures
- Prolonged Qt interval
- Impairs renal ability to concentrate urine
- Polyuria → **volume contraction**
 - Decreased GFR
 - Contributes to renal failure

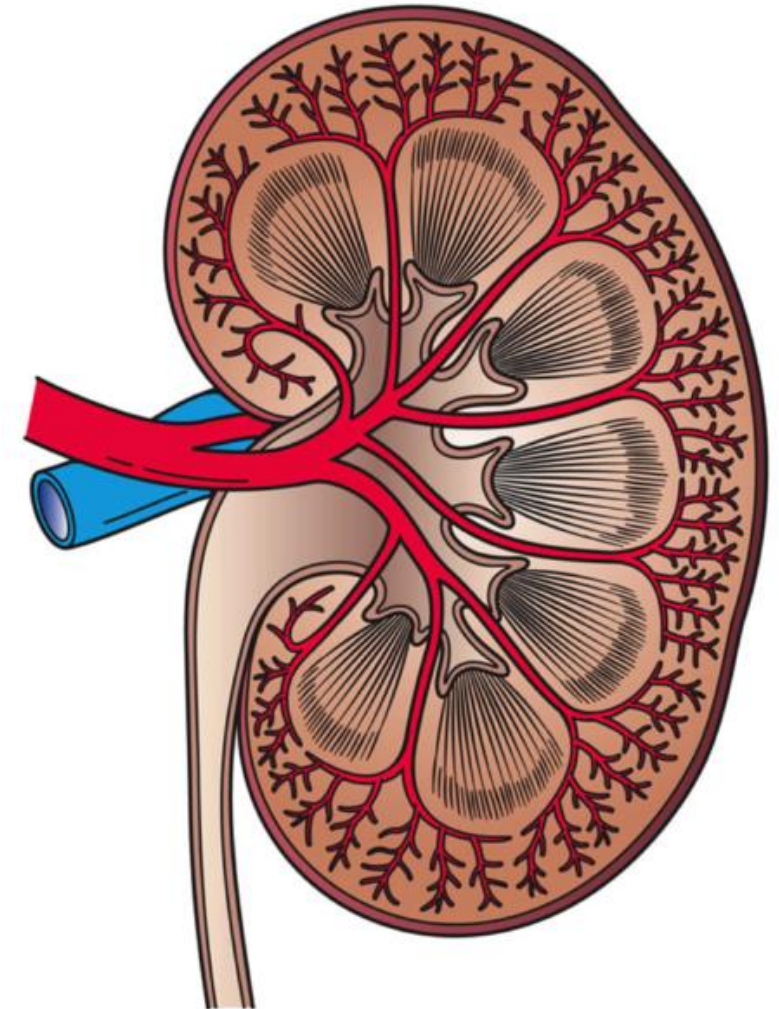
1 H	
3 Li	4 Be
11 Na	12 Mg
19 K	20 Ca



Multiple Myeloma

Renal Failure

- Increased creatinine ~50% patients at diagnosis
- Hypercalcemia
- Light chains cause **“myeloma kidney”**
- Light chains in urine = “Bence Jones” proteins
- Combine with Tamm–Horsfall protein (THP)
- Form obstructing casts

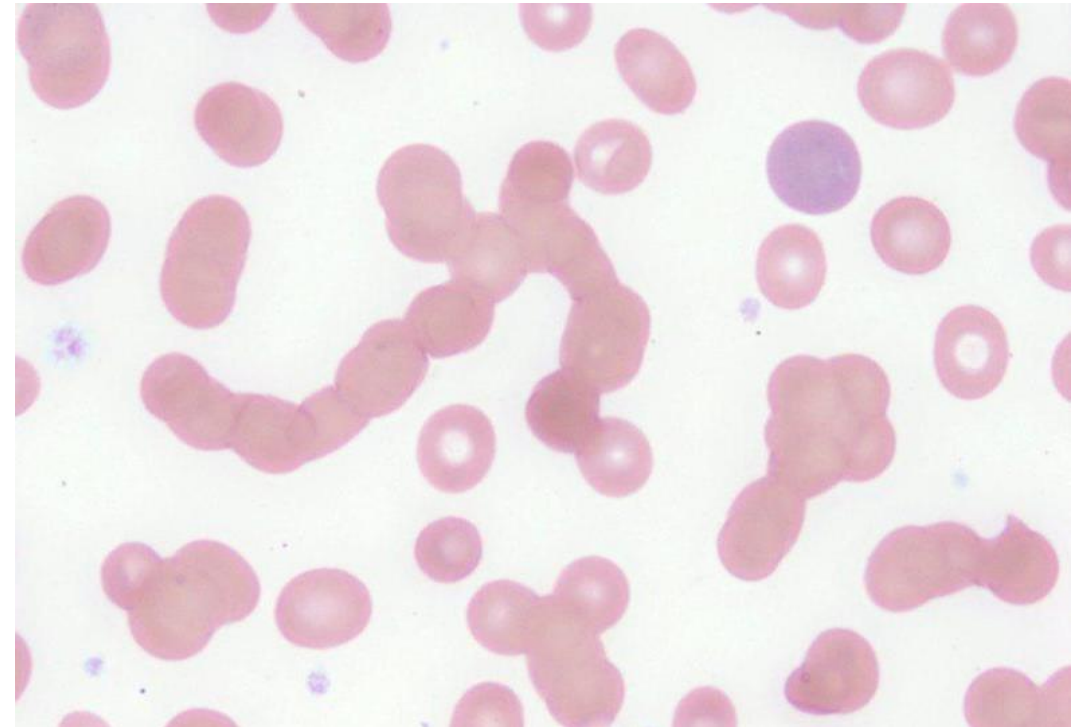


Multiple Myeloma

Anemia

- Present in about 75% of cases at diagnosis
- Usually normocytic and normochromic
- Multifactorial
 - Bone marrow replacement by plasma cells
 - Renal failure (low EPO)
- Weakness, pallor
- Peripheral smear: **rouleaux**
 - RBCs form a stack of coins
 - Caused by elevated protein levels in plasma

Rouleaux



Multiple Myeloma

Bone lesions

- Osteoclast-mediated bone resorption
- Caused by cytokines from myeloma cells
- “Lytic lesions” on x-ray (“punched out”)
- Pathologic fractures
- Often vertebral column



Multiple Myeloma

Infections

- **Leading cause of death**
- Decreased normal immunoglobulins
- Recurrent **bacterial infections**
 - Strep Pneumoniae
 - Staph Aureus
 - E. Coli

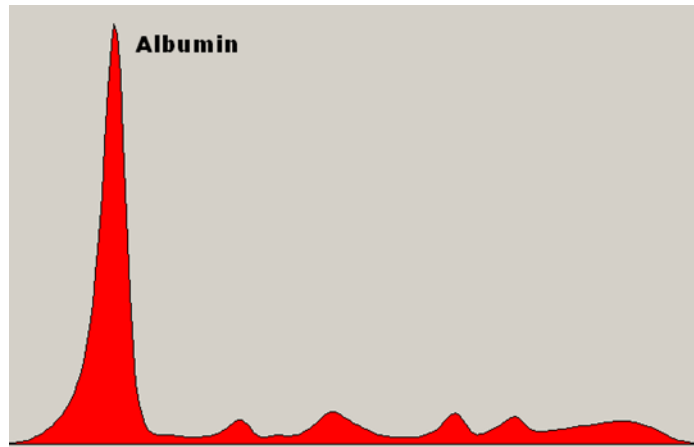
RUL Pneumonia



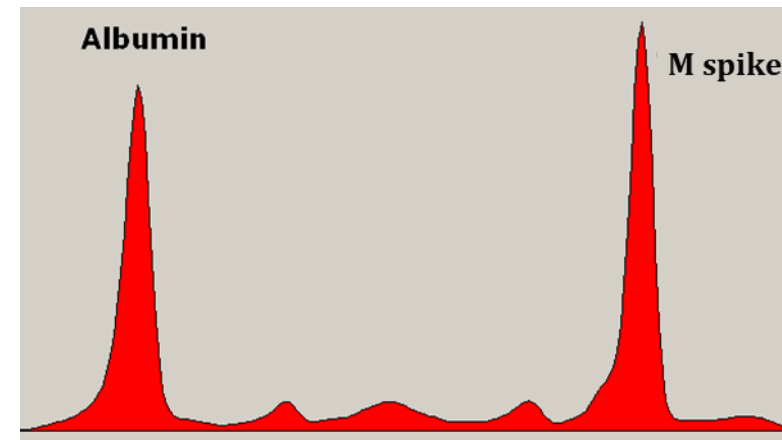
Multiple Myeloma

Monoclonal protein detection

- **Serum protein electrophoresis (SPEP)**
 - Electrical current separates serum proteins based on size and charge
 - Multiple myeloma: “M spike”
 - M protein ≥ 3 g/dL in most cases
- **Urine protein electrophoresis (UPEP)**
 - Light chain only cases have negative SPEP but positive UPEP



Normal SPEP

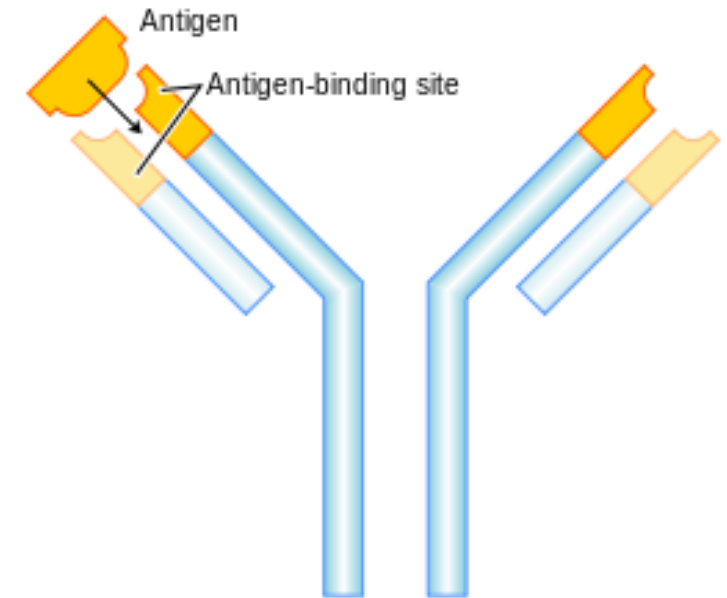


Multiple Myeloma

Multiple Myeloma

Monoclonal protein detection

- **Free light chains**
- Test for serum kappa and lambda light chains
- Most myelomas produce excess light chains – either kappa or lambda
- Ratio of kappa to lambda becomes markedly abnormal
- Normal kappa/lambda ratio: 0.26 to 1.65
- “Involved light chain:” produced by myeloma cells
- “Uninvolved light chain:” not produced by myeloma cells
- Involved/uninvolved FLC ratio followed in patients



Multiple Myeloma

Diagnosis

- **Bone marrow biopsy:** clonal plasma cells ≥ 10 percent
 - Key diagnostic test in workup
- Plus at least one **CRAB features** of end-organ damage
- Or presence of at least one **SLIM marker** of myeloma
 - Indicate inevitable clinical features of myeloma
 - **S**ixty: $\geq 60\%$ plasma cells in the bone marrow
 - **L**ight chains: involved/uninvolved FLC ratio ≥ 100
 - **M**RI: more than one bone lesion



CRAB Feature	Criteria
Calcium	$> 11\text{mg/dL}$
Renal insufficiency	$\text{CrCl} < 40\text{mL/min}$ or $\text{Cr} > 2 \text{ mg/dL}$
Anemia	$\text{Hgb} < 10 \text{ g/dL}$
Bone disease	Lytic lesions

Multiple Myeloma

Diagnosis

- Urine dipstick negative for protein
 - Mostly detects albumin
 - Poor detection of light chains
 - Urine protein electrophoresis (UPEP) required
- May cause **elevated total protein**
 - Not always present
 - Light chain only cases may have normal protein levels
- Can see an **increased gamma gap**
 - Elevated total protein with normal albumin



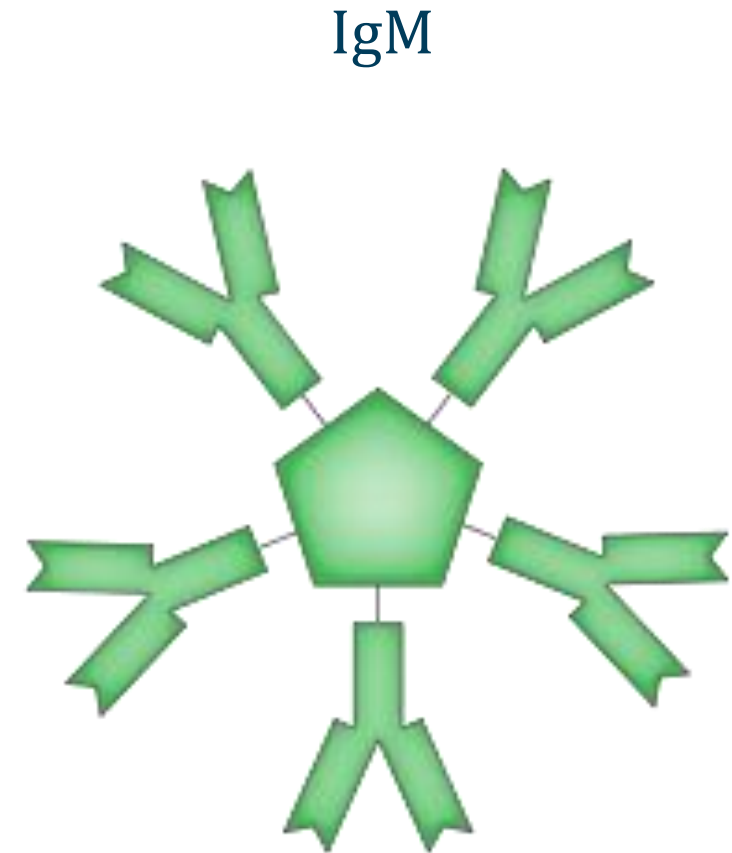
Multiple Myeloma

Treatment

- **Autologous hematopoietic cell transplantation (HCT)**
 - Patient stem cells harvested
 - Chemotherapy
 - Stem cells used to reestablish bone marrow
- **Bisphosphonates**
 - Inhibit osteoclasts
 - Improve skeletal outcomes
 - Bone pain and fractures

Waldenstrom Macroglobulinemia

- Form of **B-cell lymphoma**
- Also called lymphoplasmacytic lymphoma
- Tumor cells differentiate into plasma cells
- Produce **IgM antibodies**
- Leads to **hyperviscosity symptoms**
- Median age at diagnosis is 70 years



Waldenstrom Macroglobulinemia

Clinical features

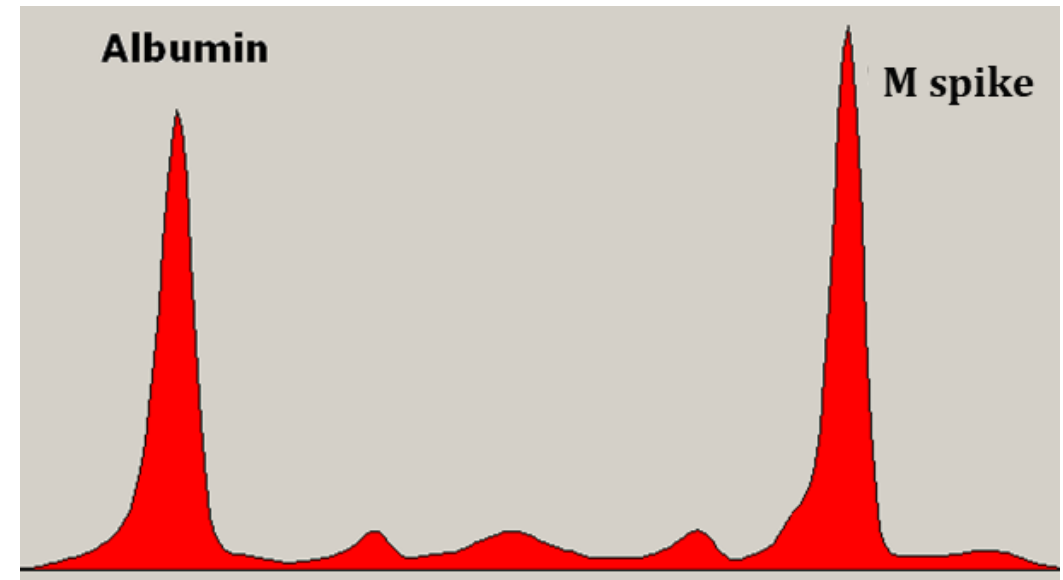
- Weakness, fatigue, weight loss
- Lymphadenopathy, hepatomegaly or splenomegaly
- **Neurologic symptoms** from hyperviscosity
 - Headache
 - Blurred vision
 - Neuropathy
 - Stroke
- Anemia



Waldenstrom Macroglobulinemia

Diagnosis

- SPEP: M spike from IgM > 3g/dL
- Bone marrow biopsy: $\geq 10\%$ lymphoplasmacytic cells
- Serum viscosity
 - Normal 1.5 centipoise (CP)
 - Hyperviscosity: usually above 4 CP



Waldenstrom Macroglobulinemia

Treatment

- **Plasmapheresis**
 - Emergency management of hyperviscosity
 - Removes IgM antibodies
 - Indicated with severe neurologic impairment
- Chemotherapy

Plasmapheresis



Mr Vacchi /Wikipedia

MGUS

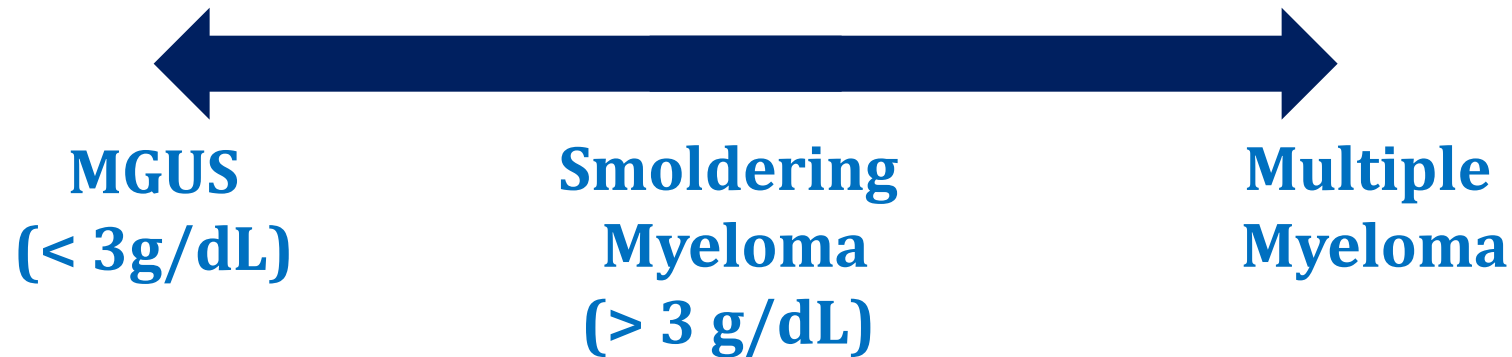
Monoclonal gammopathy of undetermined significance

- Premalignant disorder
- Non-IgM MGUS (most common): may progress to multiple myeloma
- IgM MGUS: may progress to Waldenstrom Macroglobulinemia
- Generally asymptomatic
- Usually identified when working up another condition
- Anemia, neuropathy, hypercalcemia, etc.

MGUS

Monoclonal gammopathy of undetermined significance

- SPEP: monoclonal protein < 3 g/dL
- Bone marrow findings that do not meet criteria (< 10%)
- Managed with close monitoring for disease progression



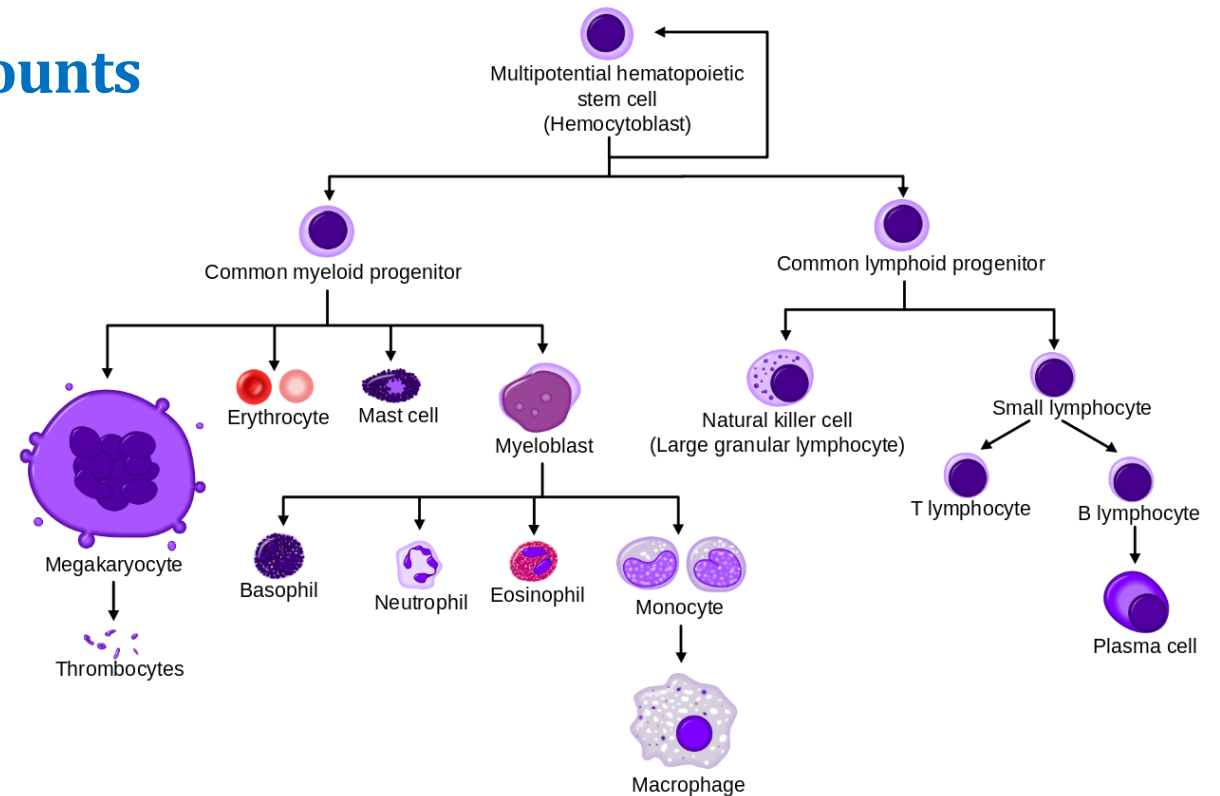
Myeloproliferative Disorders

Jason Ryan, MD, MPH



Myeloproliferative Disorders

- Overproduction of **myeloid cells**
 - Granulocytes, red cells, platelets
- Leads to **increased peripheral cell counts**



Myeloproliferative Disorders

Major Types

- Chronic myeloid leukemia (granulocytes)
- Polycythemia vera (red blood cells, platelets and granulocytes)
- Essential thrombocytosis (platelets)
- Myelofibrosis

Polycythemia Vera

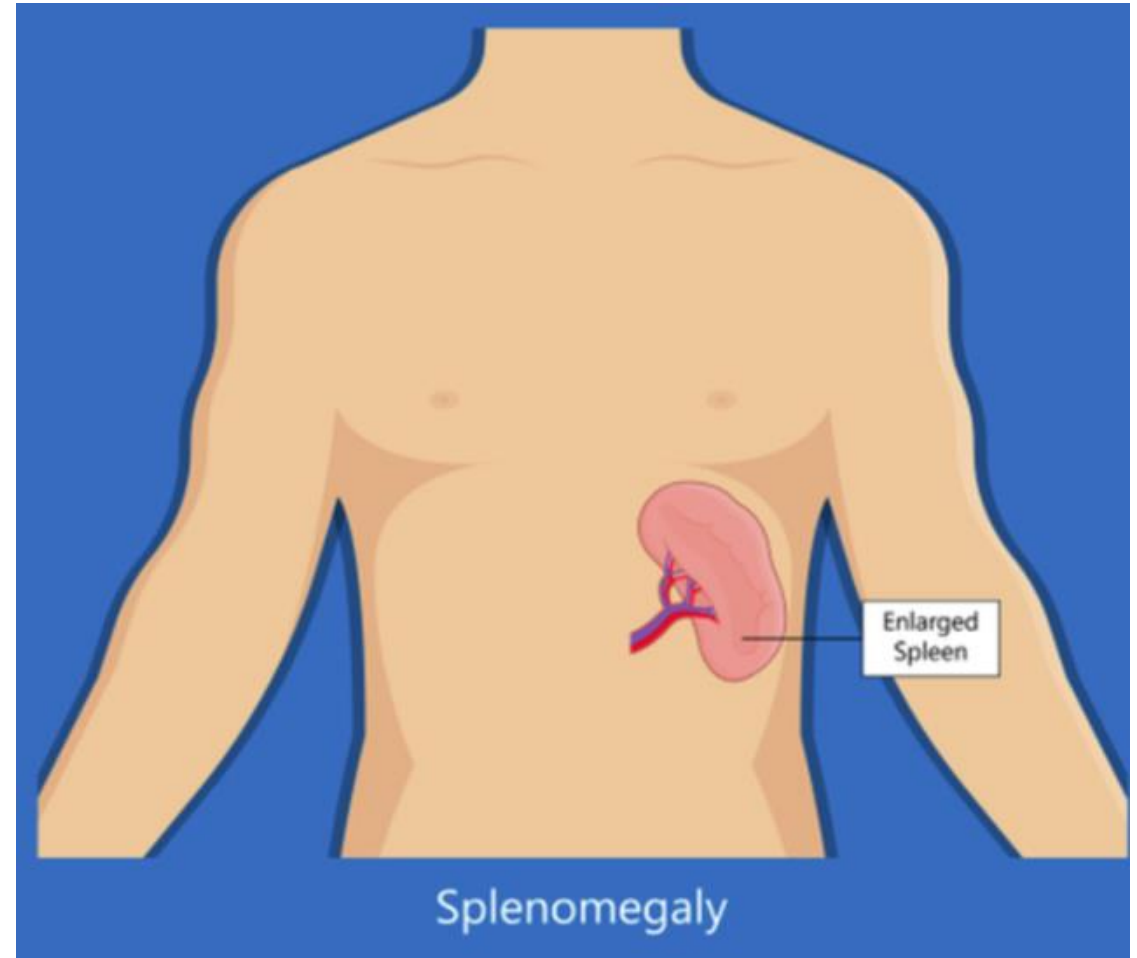
- **Elevated red blood cell mass**
 - Increased hemoglobin and hematocrit
- May also have **leukocytosis** or **thrombocytosis**
 - Increased WBC
 - Increased platelets



Polycythemia Vera

Clinical features

- Many patients asymptomatic
 - Detected by routine CBC
- **Hypertension and flushing**
 - Increased RBC mass → ↑ blood volume
- **Thrombosis**
 - Increased viscosity of blood
 - Also increased platelets
 - Deep vein thrombosis
 - Often Budd Chiari syndrome (hepatic vein)
- **Splenomegaly**



Polycythemia Vera

Clinical features

- Red, puffy skin (“facial plethora”)
- **Aquagenic pruritus**
 - “Unbearable” pruritus after warm bath or shower
 - Often presenting complaint
- **Erythromelalgia**
 - Burning sensation in hands and feet



Polycythemia Vera

Differential Diagnosis

- Must determine **erythropoietin (EPO) level**
- Must exclude \uparrow RBC due to \uparrow EPO
- EPO level is low in polycythemia vera
- EPO level is high in other conditions
 - Hypoxia (lung disease)
 - EPO secreting tumors
 - Renal cell carcinoma
 - Hepatocellular carcinoma

Measurement	Hypoxia	RCC/HCC	P. Vera
PaO ₂	↓	Normal	Normal
EPO	↑	↑	↓

Polycythemia Vera

JAK2 mutation

- Gene for enzyme **Janus kinase 2**
- Mutation → abnormal cell function/growth
- Progenitor cells: **hypersensitivity to cytokines**
- More growth and longer survival

Disorder	Genetics
Polycythemia Vera	JAK2 (~95%)
Essential Thrombocytosis	JAK2 (~60%)
Myelofibrosis	JAK2 (~60%)



Polycythemia Vera

Diagnostic criteria

- **Elevated hemoglobin or hematocrit**
 - > 16.5 g/dL or 49% in men
 - > 16.0 g/dL or 48% in women
- **Bone marrow biopsy with hypercellularity**
 - “Trilineage growth” or panmyelosis
 - Erythroid, granulocytic, and megakaryocytic proliferation
- **Presence of *JAK2* mutation**
- Minor criteria: reduced serum erythropoietin
- All 3 major or first two plus minor criteria



Polycythemia Vera

Treatment

- **Phlebotomy**
 - Goal Hct < 45%
- **Low-dose aspirin**
 - Thrombosis prevention
- **Hydroxyurea**
 - Inhibits ribonucleotide reductase → ↓ DNA → ↓ cell division
- **Ruxolitinib**
 - JAK inhibitor
 - Used in refractory cases only

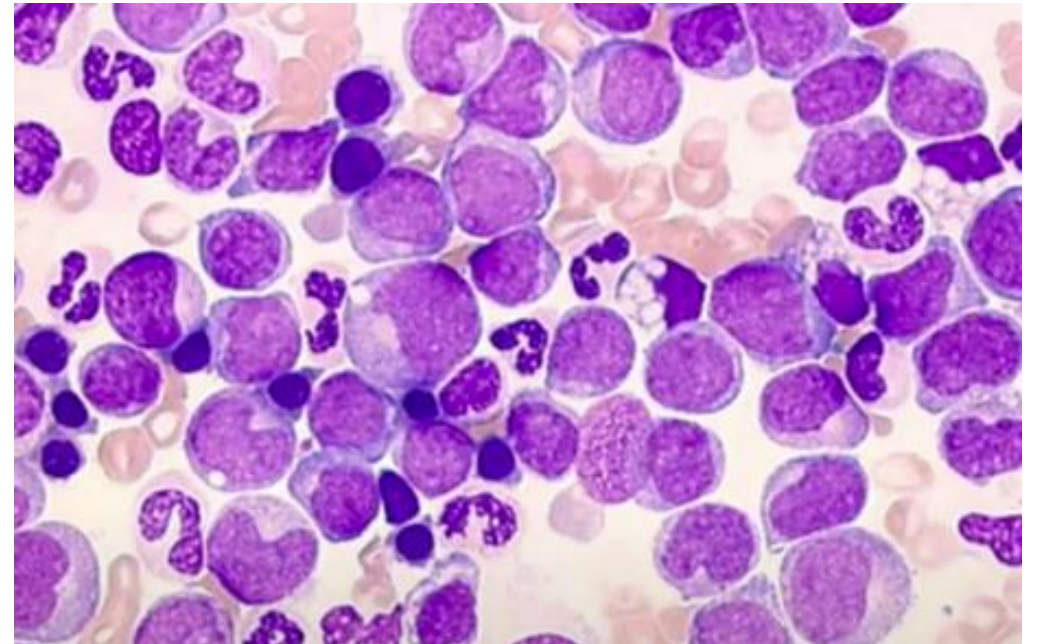


Polycythemia Vera

Complications

- Spent phase
 - Occurs in ~15% of patients
 - Progression to myelofibrosis
- Leukemia
 - Usually acute myeloid leukemia (AML)
 - Rarely chronic myeloid leukemia (CML)
- Gout
 - Excess DNA turnover from ↑ RBC production
 - ↑ uric acid

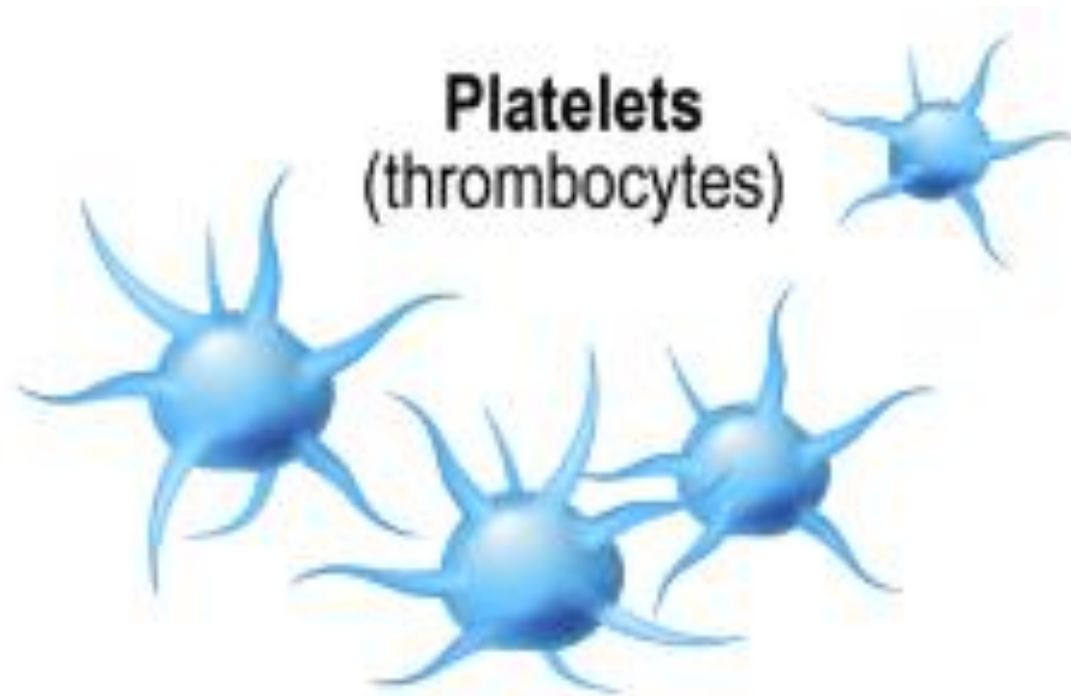
Acute Myeloid Leukemia



Essential thrombocytosis

Essential thrombocythemia

- Proliferation of myeloid cells
- Predominantly affects **megakaryocytes/platelets**



Essential thrombocytosis

Clinical features

- Often incidental finding of increased platelets
- Vasomotor symptoms – microvascular dysfunction
 - Headache
 - Dizziness or syncope
 - Erythromelalgia
 - Visual disturbances
- **Thrombosis**
- **Bleeding** - abnormal platelet function
- Pruritus rare (contrast with P. vera)



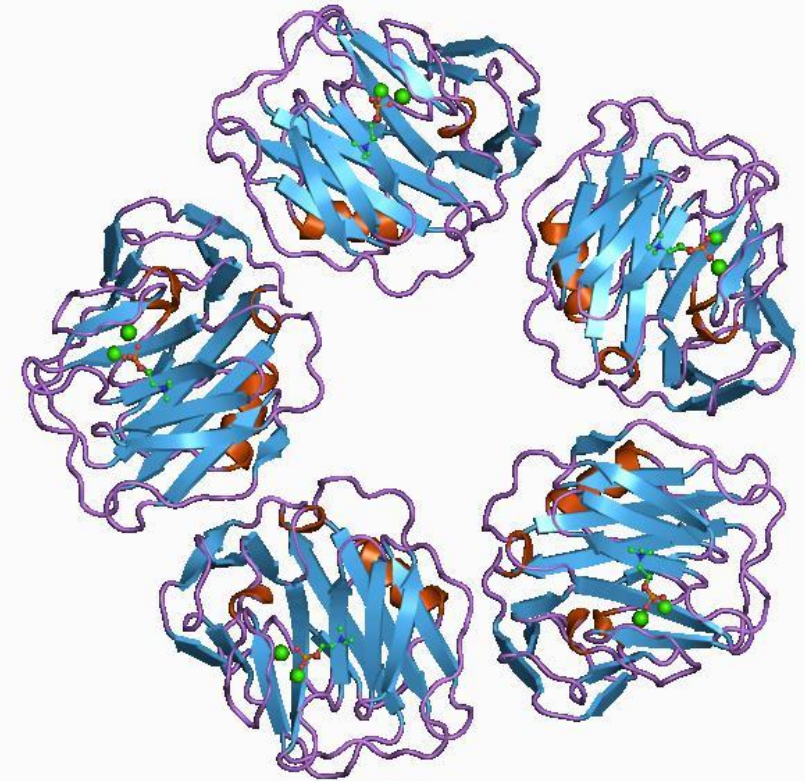
Blood clot (thrombus)

Essential thrombocytosis

Diagnosis

- Must exclude a **reactive thrombocytosis**
 - Iron deficiency anemia – iron studies
 - Acute bleeding or hemolysis – CBC and LDH
 - Infections/inflammation
 - Metastatic cancer
- Key blood test: **acute phase reactants**
 - C-reactive protein, fibrinogen, ESR, ferritin
 - Increased levels suggest occult inflammation

C-reactive Protein



Essential thrombocytosis

Diagnostic criteria

- **Platelet count > 450,000**
 - Normal: 150k to 450k
- **Bone marrow biopsy:** proliferation of megakaryocytes
- Does not meet criteria for P. vera, CML, or other syndromes
- Presence of JAK2 or other ET mutation
- Minor criteria: no identifiable cause of thrombocytosis
 - Iron deficiency, infection, inflammation

CRITERIA

Essential thrombocythosis

Treatment

- Vasomotor symptoms often respond to low dose aspirin
- Treatment based on **thrombosis risk**
 - Major factors: age and presence of JAK 2 mutation
- High risk patients treated with hydroxyurea and aspirin

High Risk	Low Risk
Age > 60 + JAK2 mutation	Age ≤ 60 + no JAK2 mutation

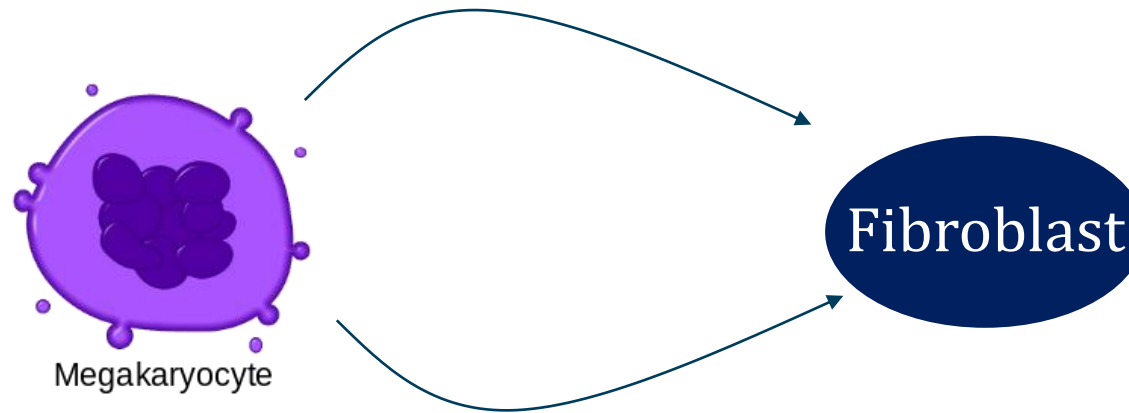
Myelofibrosis

- Bone marrow replaced by fibrous scar tissue
- Primary myelofibrosis - myeloproliferative disorder
- Secondary myelofibrosis – due to another process
 - Polycythemia vera
 - Chronic leukemia
 - Other causes



Primary Myelofibrosis

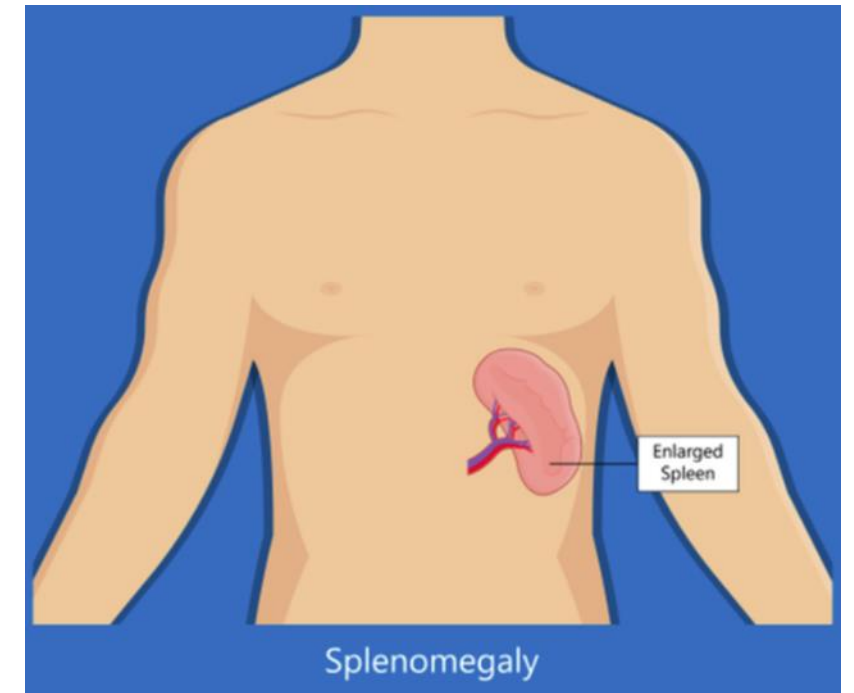
- Excess collagen from fibroblasts → marrow fibrosis
- Fibroblasts stimulated by growth factors from megakaryocytes
- Median age at presentation is 67 years



Primary Myelofibrosis

Clinical features

- Most patients have fatigue
- Bone marrow failure → **extramedullary hematopoiesis**
 - Spleen, liver, lymph nodes
 - Can be seen in CNS, lungs, bladder, even in skin!
- **Massive splenomegaly**
 - Spleen is principle site of extramedullary hematopoiesis
 - Left upper abdominal pain
 - Early satiety (compression of stomach)
- May also see enlarged liver and lymph nodes

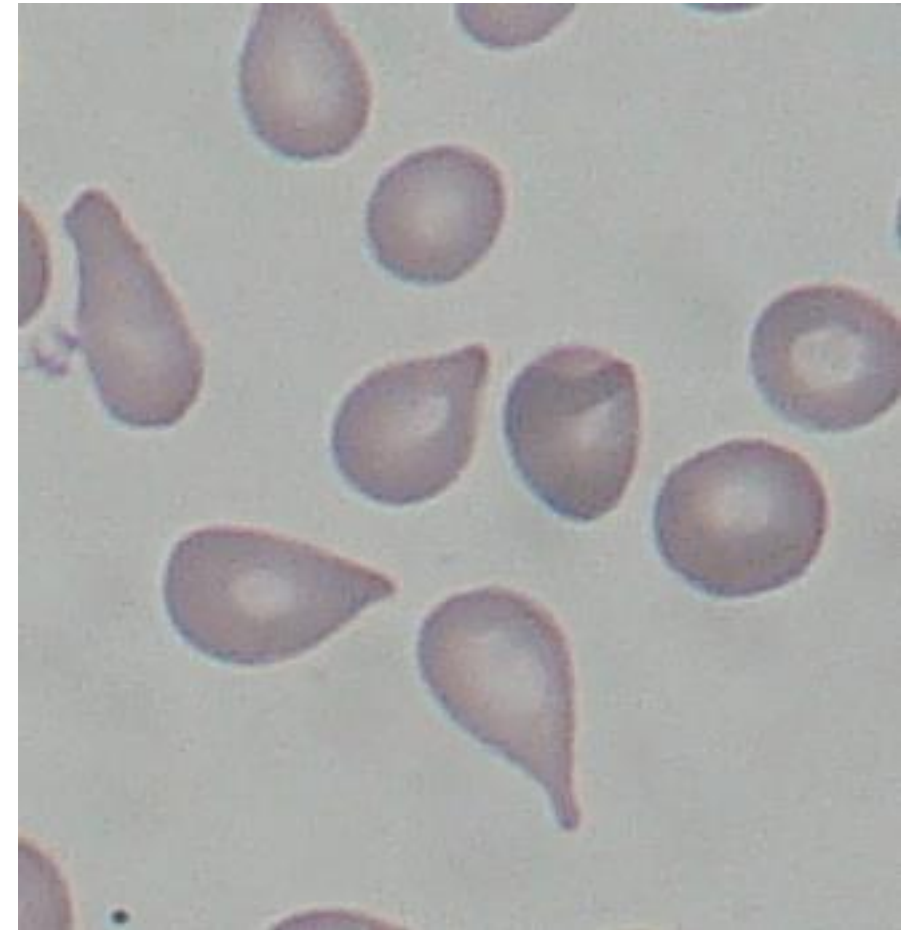


Primary Myelofibrosis

Peripheral smear and blood counts

- **Tear drop cells (dacrocytes)**
 - Classic finding of myelofibrosis
 - Red blood cells deformed leaving fibrotic marrow
- Leukoerythroblastosis
 - Inappropriate release of cells from marrow
 - Immature erythroid and granulocyte precursors in blood
- Anemia
- Platelet and WBC variable
 - May be low or high

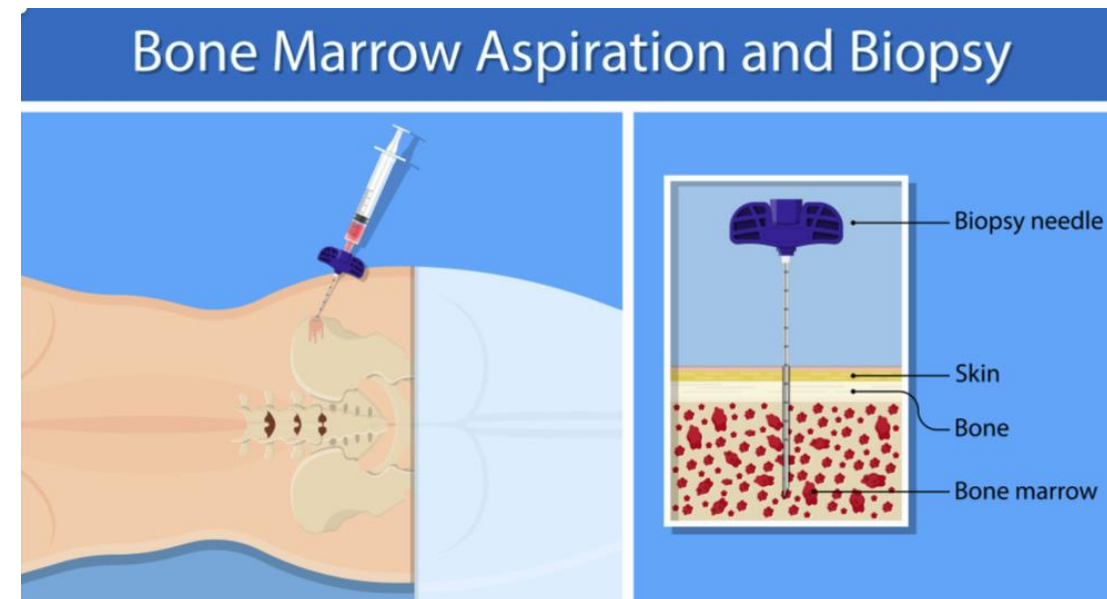
Dacrocyte



Primary Myelofibrosis

Diagnostic workup

- Fatigue and splenomegaly → CBC
- Anemia +/- abnormal platelets and white cells
- Peripheral smear shows **dacrocytes** and other findings
- Bone marrow aspiration: “dry” tap
- Bone marrow biopsy: fibrosis



Primary Myelofibrosis

Treatment

- Median overall survival about six years
- Least favorable prognosis of myeloproliferative disorder
- Death from progression, leukemic transformation, thrombosis or infections
- Eligible patients: allogeneic hematopoietic cell transplantation (HCT)
- Other treatments: hydroxyurea and ruxolitinib



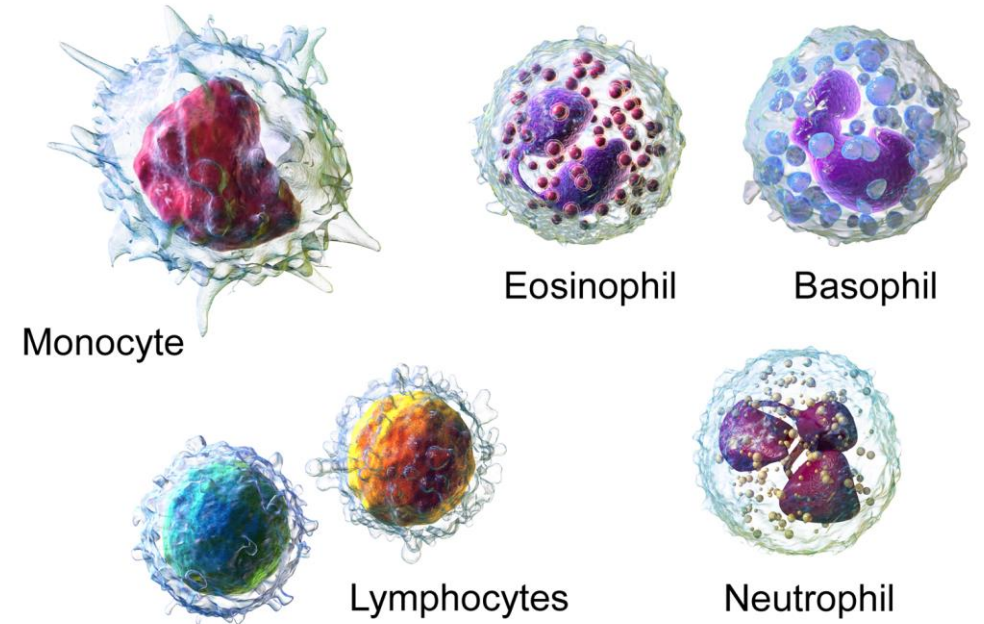
Acute Leukemias

Jason Ryan, MD, MPH



Leukemia

- Malignant proliferation of white blood cells
- Cells appear in blood (contrast with lymphoma)
- May see increased white blood cell count

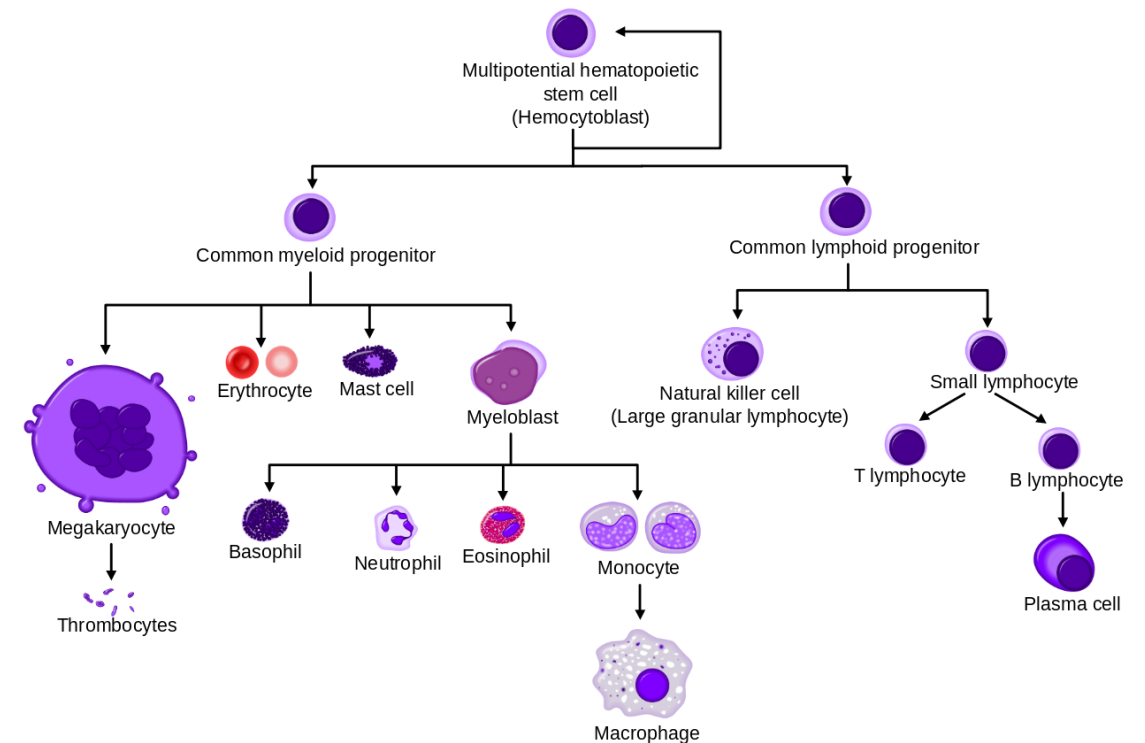


White Blood Cells

Leukemias

Classification

- Myeloid versus lymphoid
- Acute versus chronic
- Acute
 - Rapid onset of symptoms
 - Involves **blasts** in bone marrow
- Chronic
 - Slower onset of symptoms (or no symptoms)
 - Malignant cells are not blasts (more mature)



Acute Lymphoblastic Leukemia

ALL

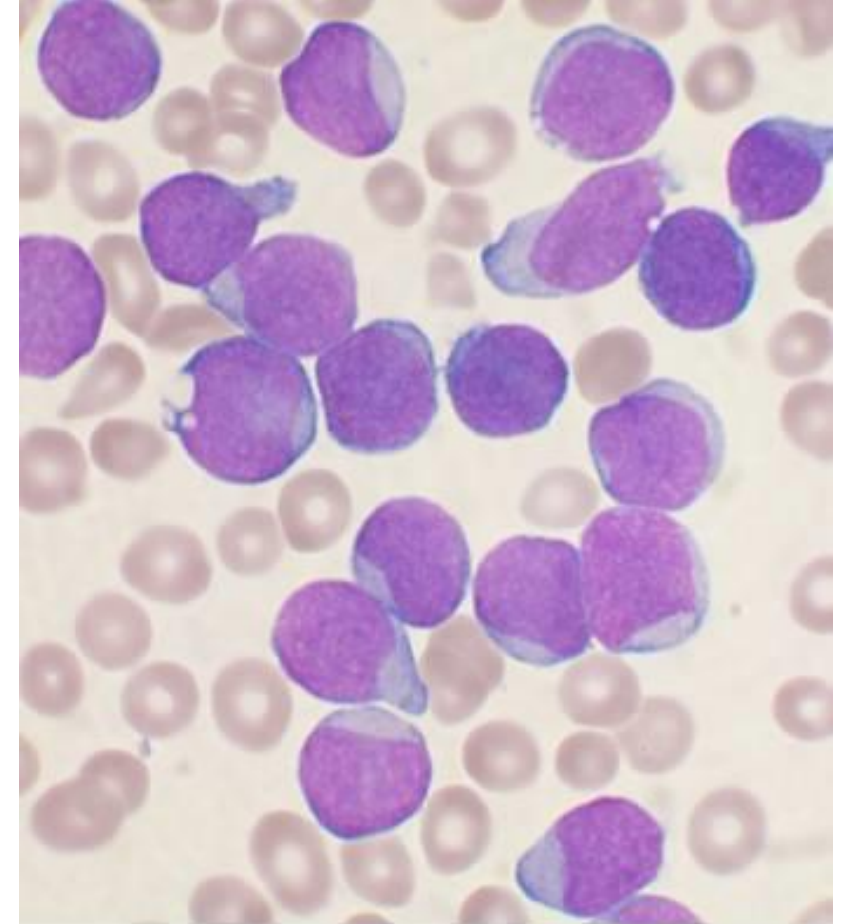
- Mostly a disease of children
- Peak incidence ~ **4 years old**
- Fever and bone pain from marrow expansion
- Lymphadenopathy, **splenomegaly**, hepatomegaly
 - Infiltration by malignant cells
- Headache, vomiting
 - Meningeal spread
- May cause bone marrow depression
 - Anemia, thrombocytopenia, neutropenia



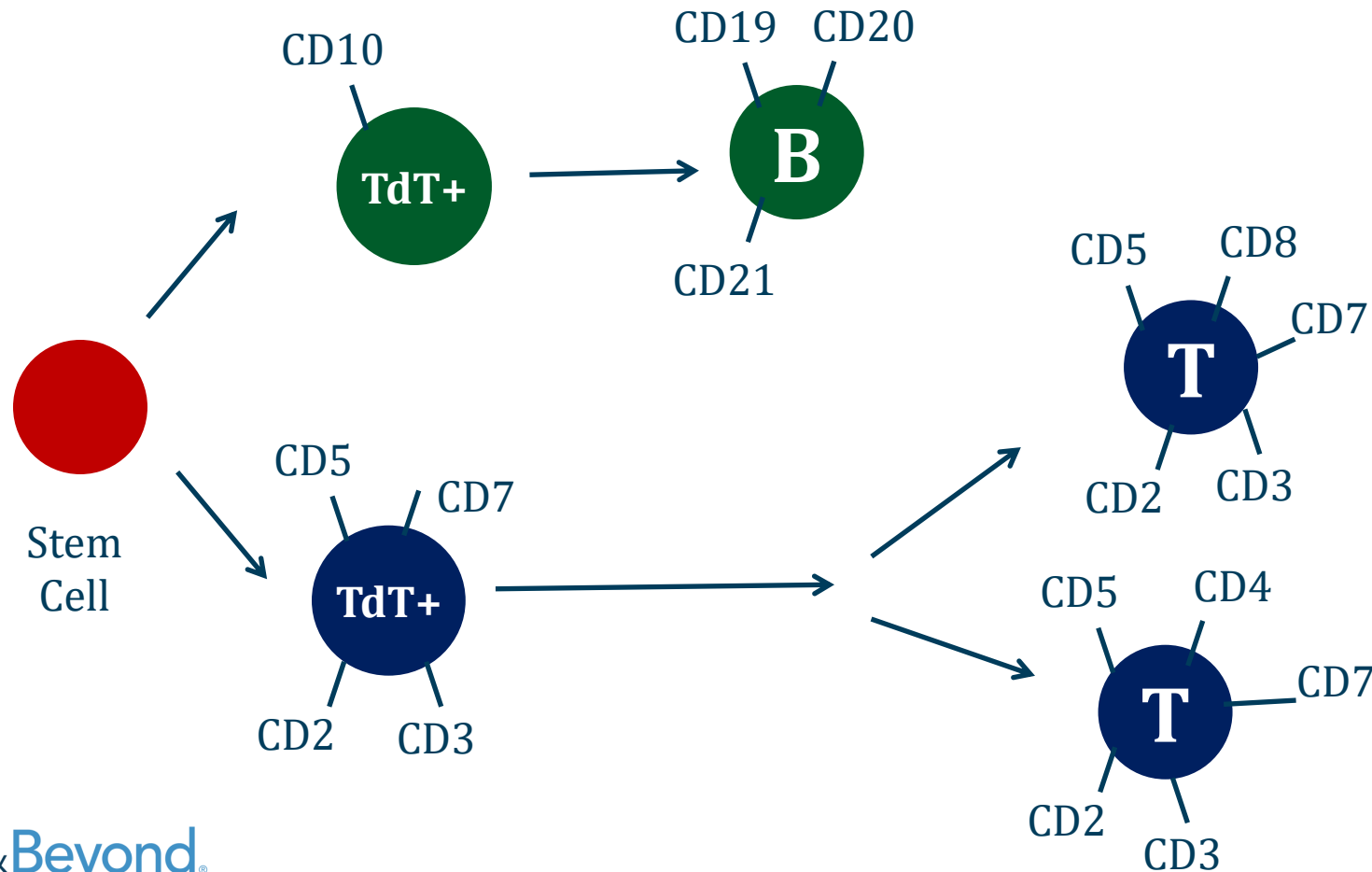
Acute Lymphoblastic Leukemia

Clinical findings

- Key test: CBC
- **Anemia**
- **Thrombocytopenia**
- Lymphoblasts
- Total WBC variable
 - 50% show WBC count $< 10,000/\mu\text{L}$
 - 20% show WBC count $> 50,000/\mu\text{L}$
- **Diagnosis: bone marrow biopsy**
 - Hypercellular with lymphoblasts



Lymphocyte Development



Lymphocyte Antigens

Primarily T-Cell Associated	Primarily B-cell Associated
CD1	CD10
CD2	CD19
CD3	CD20
CD4	CD21
CD5	CD22
CD7	CD23
CD8	

Acute Lymphoblastic Leukemia

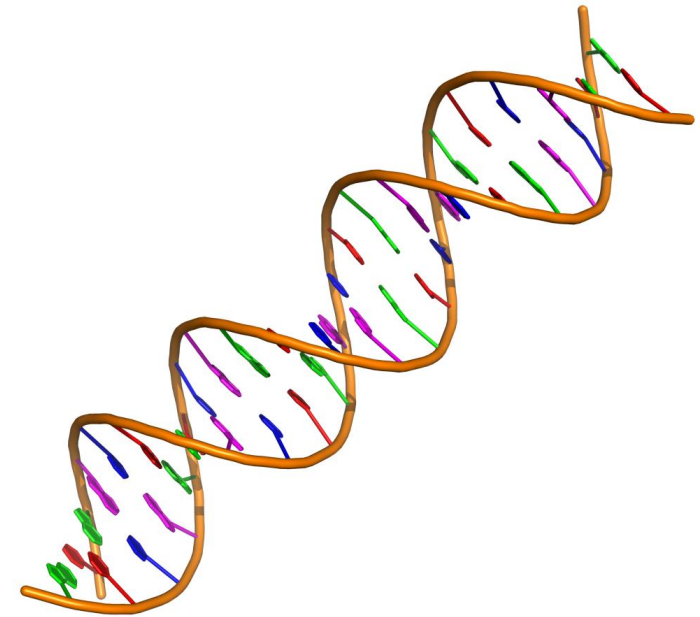
Malignant cell characteristics

- Usually **pre-B cell malignancy** ~ 70 to 80% cases
- **CD10+**
 - “Common acute lymphoblastic leukemia antigen” or “CALLA”
- **Terminal deoxynucleotidyl transferase (TdT)**
 - DNA enzyme
 - Found only in pre-B and pre-T blasts
 - NOT seen in myeloblasts
- Also CD19+, sometimes CD22+

Acute Lymphoblastic Leukemia

Genetics

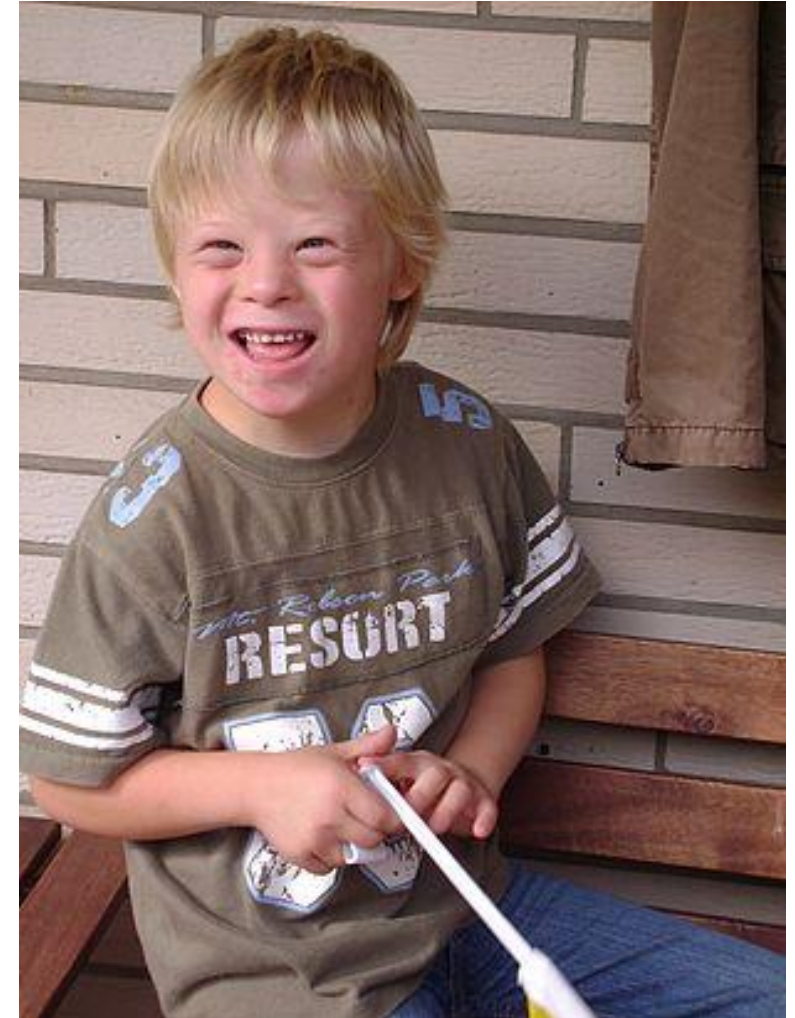
- Many different abnormalities reported in B-ALL
- **Philadelphia chromosome t(9;22)**
 - 20 to 30% ALL in adults
 - 2 to 3% ALL in children
 - Associated with a poor prognosis
- **t(12;21)**
 - Fusion product of two genes: TEL-AML1
 - TEL-AML1 impairs differentiation of blasts
 - Good prognosis
 - Most common rearrangement in children



Acute Lymphoblastic Leukemia

Associated conditions

- **Down Syndrome**
- Risk of ALL ↑↑ 10-20x
- 1-3% ALL cases have Down syndrome



Acute Lymphoblastic Leukemia

Diagnosis

- Suspected in a child with fatigue, fever, splenomegaly
- CBC with peripheral smear shows anemia, low platelets, lymphoblasts
- Bone marrow biopsy: hypercellular with lymphoblasts
- Immunophenotyping
 - Identification of cell markers
- Cytogenetics
 - Identification of gene rearrangements



Acute Lymphoblastic Leukemia

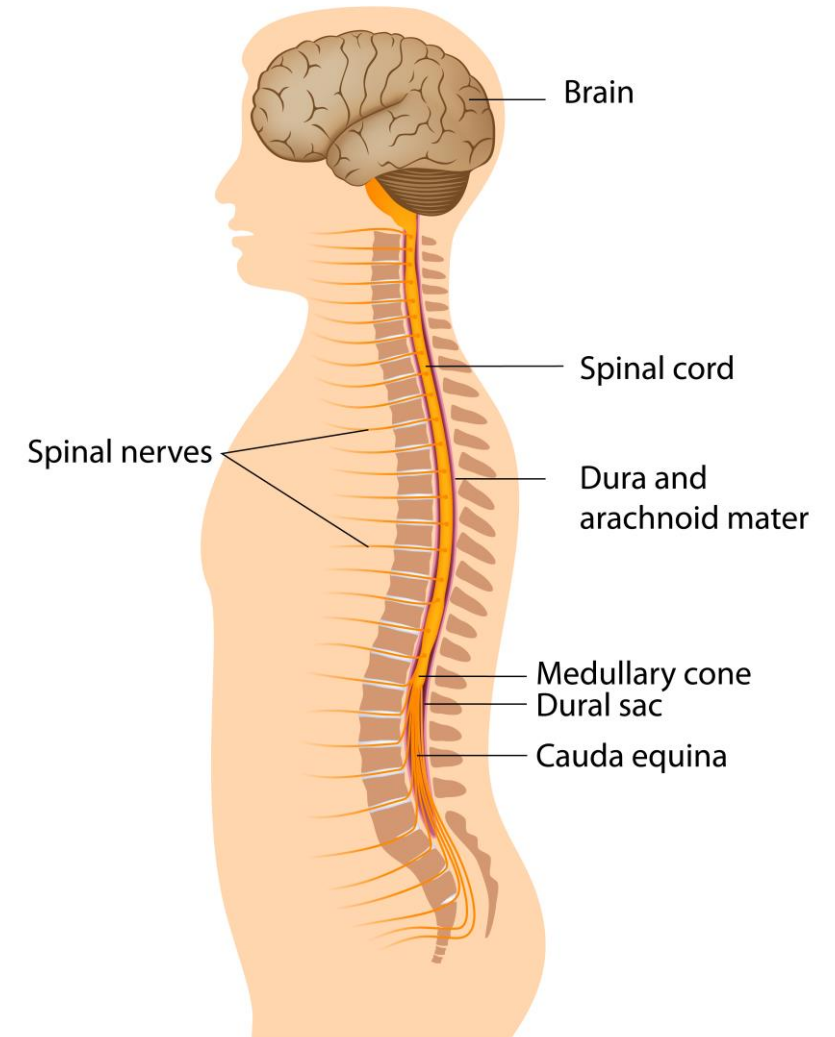
Treatment

- Cure rates are over 85%
- **Induction therapy**
 - Goal: complete remission of disease
 - Eradication of leukemia cells from marrow
 - Restoration of normal hematopoiesis
 - Multi-drug regimens
 - Often vincristine, corticosteroids and asparaginase
- Followed by **consolidation** and **maintenance** therapy

Acute Lymphoblastic Leukemia

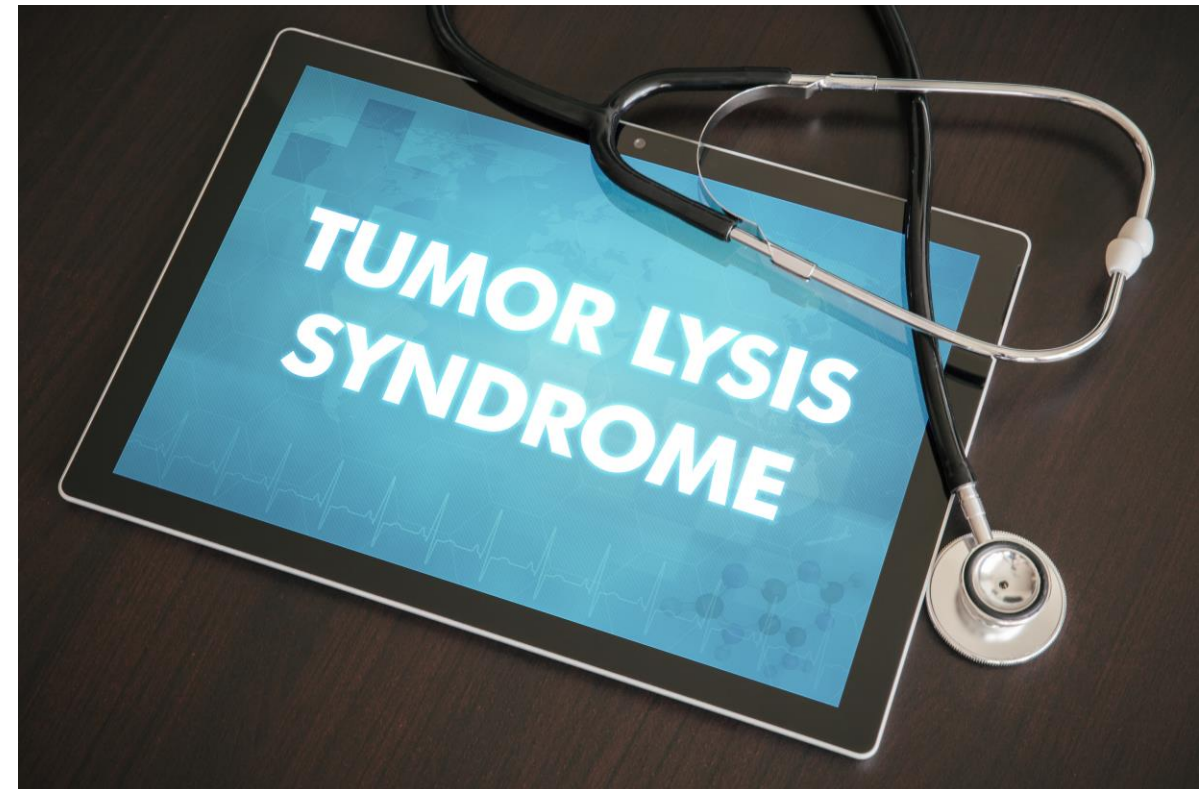
Treatment

- **Central nervous system**
 - “Sanctuary site”
 - Poor penetration by chemotherapy drugs
 - Relapse may occur without targeted treatment
- Localized treatments used
 - Radiation
 - Intrathecal chemotherapy



Tumor Lysis Syndrome

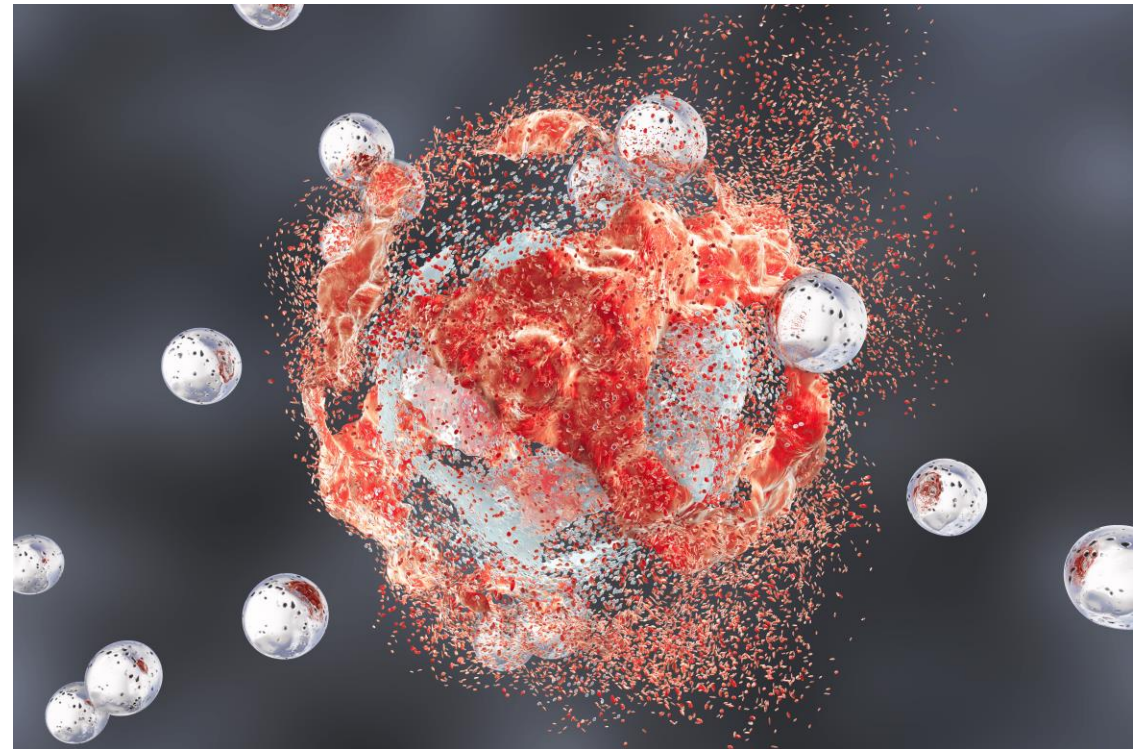
- Occurs with treatment of high-grade malignancies
 - Especially high grade Burkitt lymphoma and ALL
- Massive tumor cell lysis
- Release of intracellular contents
- Potassium, phosphate, and uric acid
- Hyperkalemia (arrhythmias)
- Hyperphosphatemia → hypocalcemia
- Hyperuricemia
- Acute kidney injury



Tumor Lysis Syndrome

Prevention

- Intravenous fluids
- **Hypouricemic agents**
 - Allopurinol: ↓ uric acid formation
 - Rasburicase: ↑ uric acid breakdown
 - Limit development of hyperuricemia
- Monitoring
 - Electrolytes and uric acid
 - Urine output



T-Cell ALL

T-Cell Acute Lymphoblastic Leukemia/Lymphoma

- Less common form of ALL
- Common in adolescent males
 - Teens to 20s
- Often presents as a **mass**
 - Lymphadenopathy
 - Mediastinal mass
 - Anterior with pleural effusions
- Tumor compression may occur
 - Superior vena cava syndrome
 - Tracheal obstruction

Mediastinal Mass

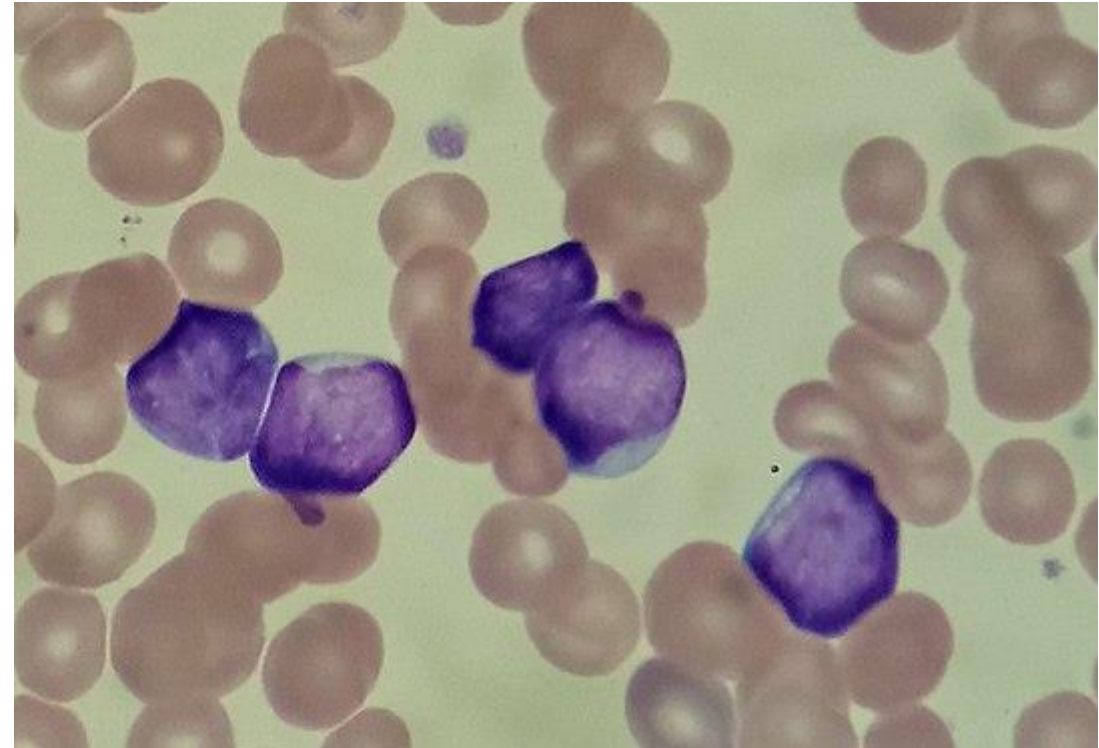


T-Cell ALL

T-Cell Acute Lymphoblastic Leukemia/Lymphoma

- Diagnosis: bone marrow or other biopsy
- Pathology: **lymphoblasts**
 - Different markers from B-cell ALL
 - Usually CD7+
 - Can see CD2, CD3, CD5, CD4, CD8
 - Not CD10+
- Treatment similar to B-cell ALL

Lymphoblasts



Acute Myeloid Leukemia

AML

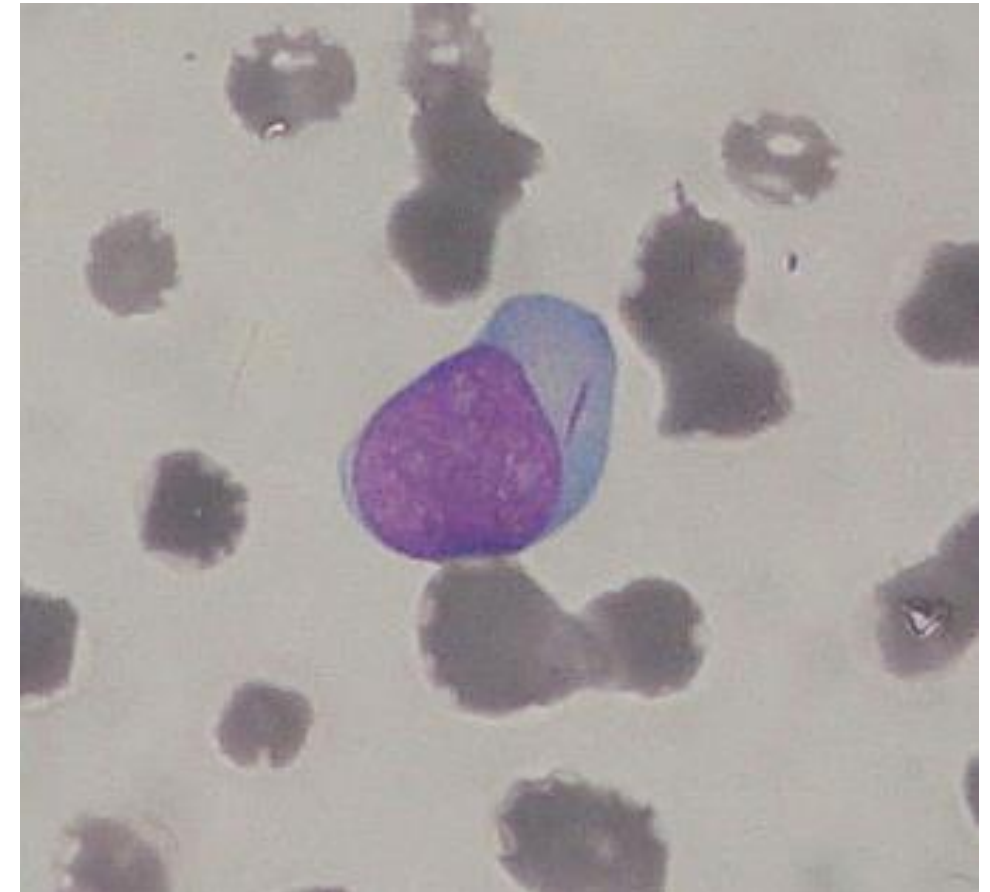
- Malignancy of myeloblasts
- Common in **adults**
 - Median age at diagnosis: 65
- Symptoms from **bone marrow suppression**
 - Myeloblasts suppress cell growth
 - Anemia: fatigue, weakness, pallor
 - Thrombocytopenia: bleeding (especially gums)
 - Neutropenia: infections
- Enlarged lymph nodes, spleen, liver less common than ALL
- LDH often elevated (nonspecific)

Acute Myeloid Leukemia

Blood counts and peripheral blood findings

- **Anemia and thrombocytopenia**
- WBC variable
 - 50% have WBC > 10,000
 - 20% have WBC > 100,000
- **Myeloblasts**
 - Myeloperoxidase (MPO) positive
- **Auer rods**
 - Pathognomonic AML
 - Accumulation of MPO
- In bone marrow biopsy > 20% myeloblasts

Myeloblast with Auer Rod



Acute Myeloid Leukemia

Subtypes

- Classified into numerous subtypes (WHO system)
- Based on morphology, surface markers, genetics
- Key subtype: **Acute Promyelocytic Leukemia (APML)**
 - Defined by translocation t(15;17)
 - Creates a fusion gene: PML-RARA
 - Promyelocytic leukemia gene (chromosome 15)
 - Retinoic acid receptor alpha (chromosome 17)
- Diagnosis via translocation detection

APML

Acute Promyelocytic Leukemia

- Abnormal retinoic acid receptor (RAR)
- Treatment: **all trans retinoic acid (ATRA)**
 - Form of vitamin A
- ATRA started as soon as diagnosis is suspected
 - Yields short term remission (~ 4 months)
 - Must be followed or combined with other therapy
- **Disseminated intravascular coagulation**
 - Promyelocytes contains high levels of MPO (Auer rods common)
 - Release → DIC (common initial presentation)

Acute Myeloid Leukemia

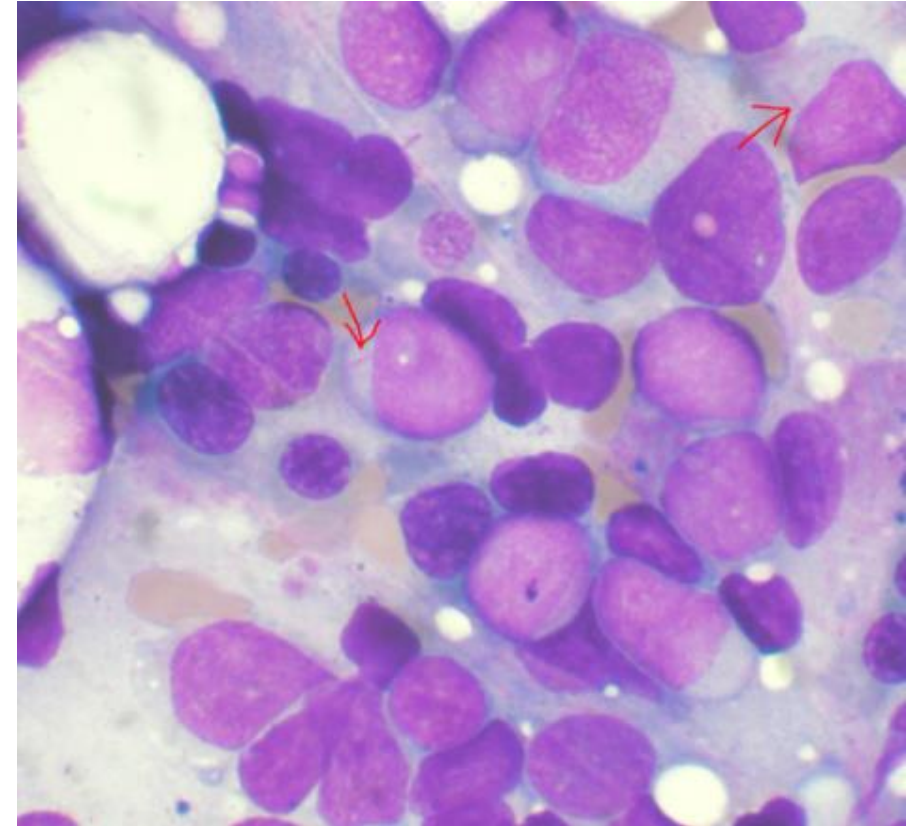
Treatment

- Various treatment regimens
- Induction therapy
 - Often with cytarabine and daunorubicin
- Followed by consolidation +/- maintenance therapy
- Hematopoietic stem cell transplantation

Myelodysplasia

Myelodysplastic Syndromes (MDS)

- Abnormal **myeloid** progenitor cells
- Leads to **ineffective hematopoiesis**
 - Almost all have anemia
 - May present with **macrocytic anemia**
 - Some have thrombocytopenia and neutropenia
- Diagnosis: bone marrow biopsy
 - Blasts < 20% cells
- Can progress to AML (> 20% blasts)



MDS
< 20% cells
blasts



AML
> 20% cells
blasts

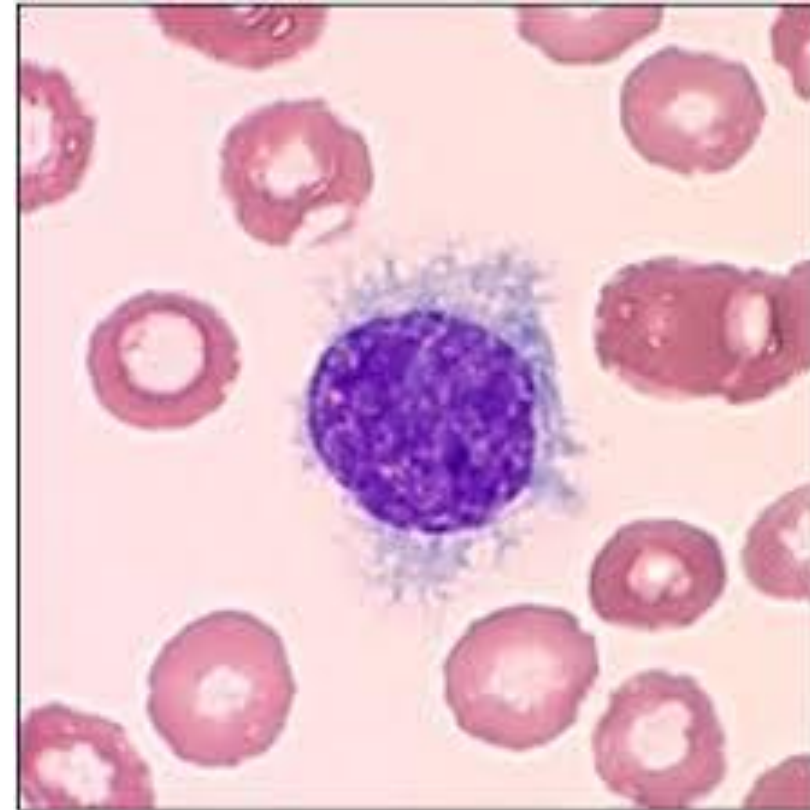
Myelodysplasia

Myelodysplastic Syndromes (MDS)

- Associated with **environmental factors**
 - Prior radiation
 - Chemotherapy
 - Usually years after exposure
- Treatment: transfusions for cytopenias
- Monitoring versus chemotherapy
 - Based on patient factors and disease risk

Hairy Cell Leukemia

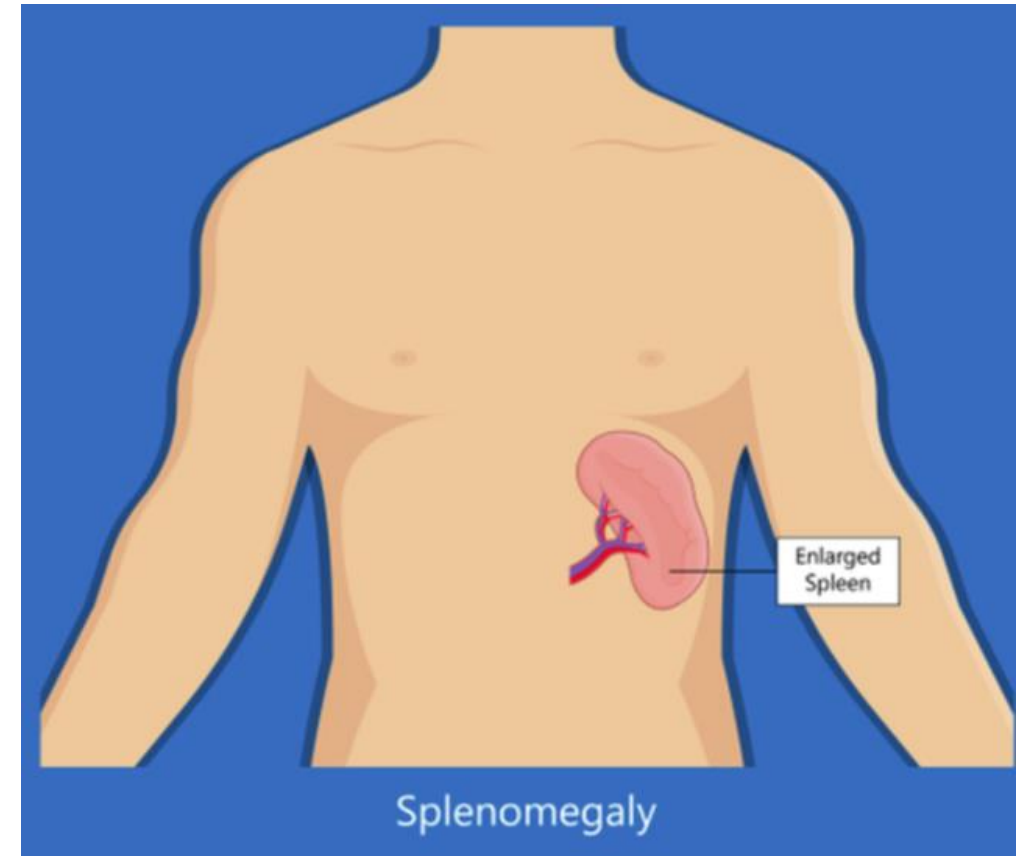
- Rare chronic **B-cell malignancy**
 - Express CD19, CD20, CD22
- Peripheral smear: **hairy cells**
 - Lymphocytes
 - Hair-like cytoplasm projections



Hairy Cell Leukemia

Unique Features

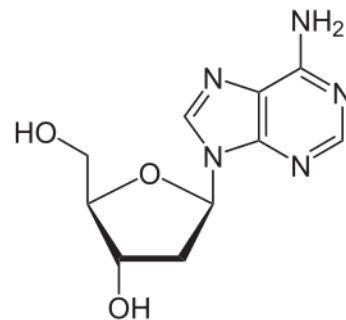
- **Massive splenomegaly**
 - Invasion by malignant cells
 - May cause abdominal pain
- Hairy cells induce **marrow fibrosis**
 - “Dry tap” on bone marrow aspiration
 - Pancytopenia occurs
- **BRAF mutation**
 - Present in nearly all cases
 - Leads to malignant cell growth



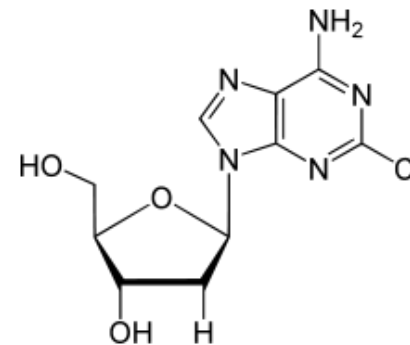
Hairy Cell Leukemia

Clinical features and treatment

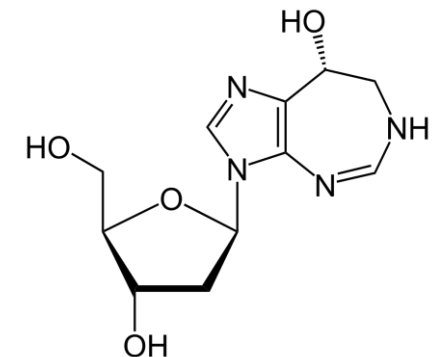
- Median age at diagnosis: 52 years
- Often **abdominal pain**
- Fatigue, weakness
- Peripheral smear with hairy cells
- Immunophenotyping shows B cell markers
- Treatment: purine analogs
 - Mimic structure of purines
 - Cladribine
 - Pentostatin



Deoxyadenosine
(found in DNA)



Cladribine



Pentostatin

Chronic Leukemias

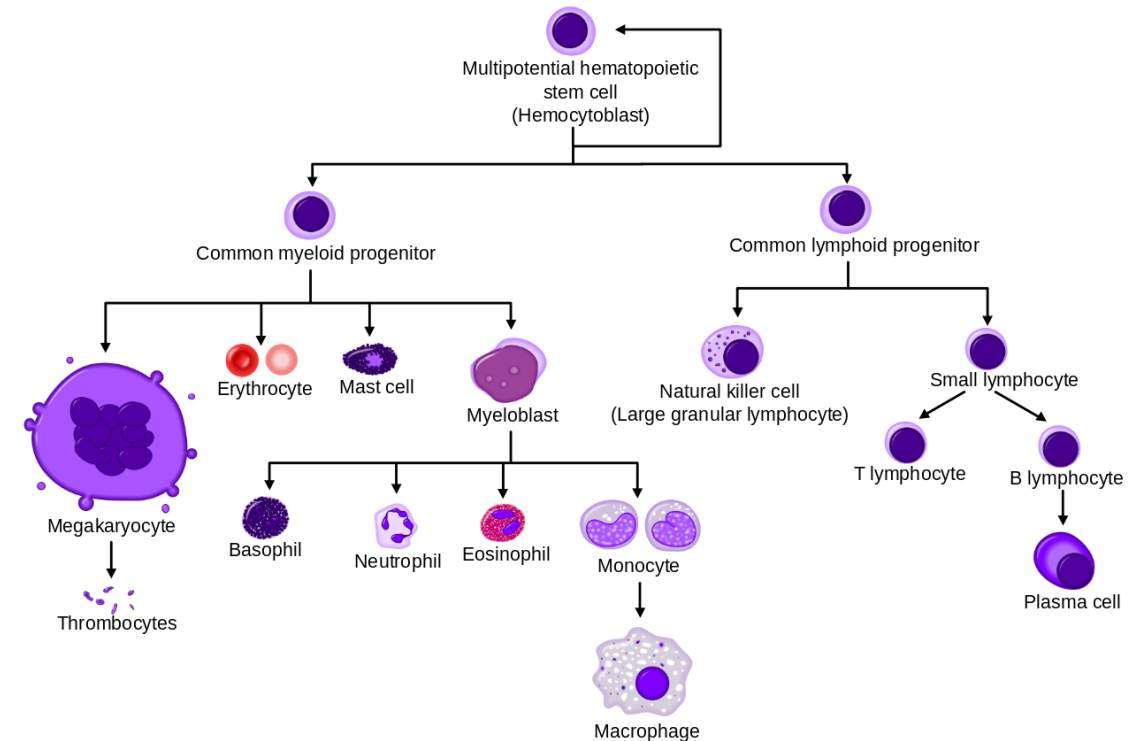
Jason Ryan, MD, MPH



Leukemias

Classification

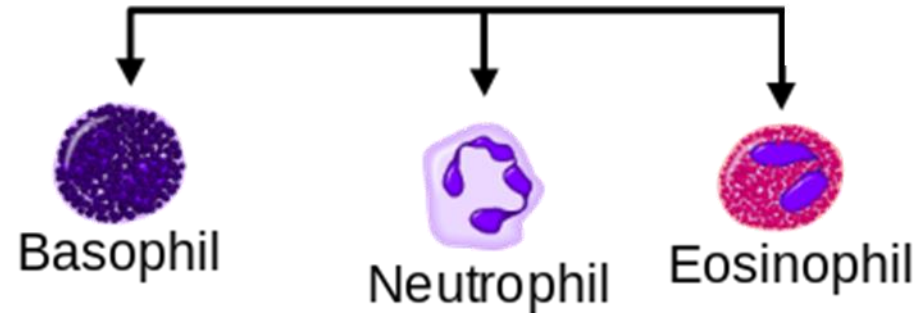
- Myeloid versus lymphoid
- Acute versus chronic
- Acute
 - Rapid onset of symptoms
 - Involves blasts in bone marrow
- **Chronic**
 - Slower onset of symptoms (or no symptoms)
 - Malignant cells are not blasts (more mature)



Chronic Myeloid Leukemia

CML

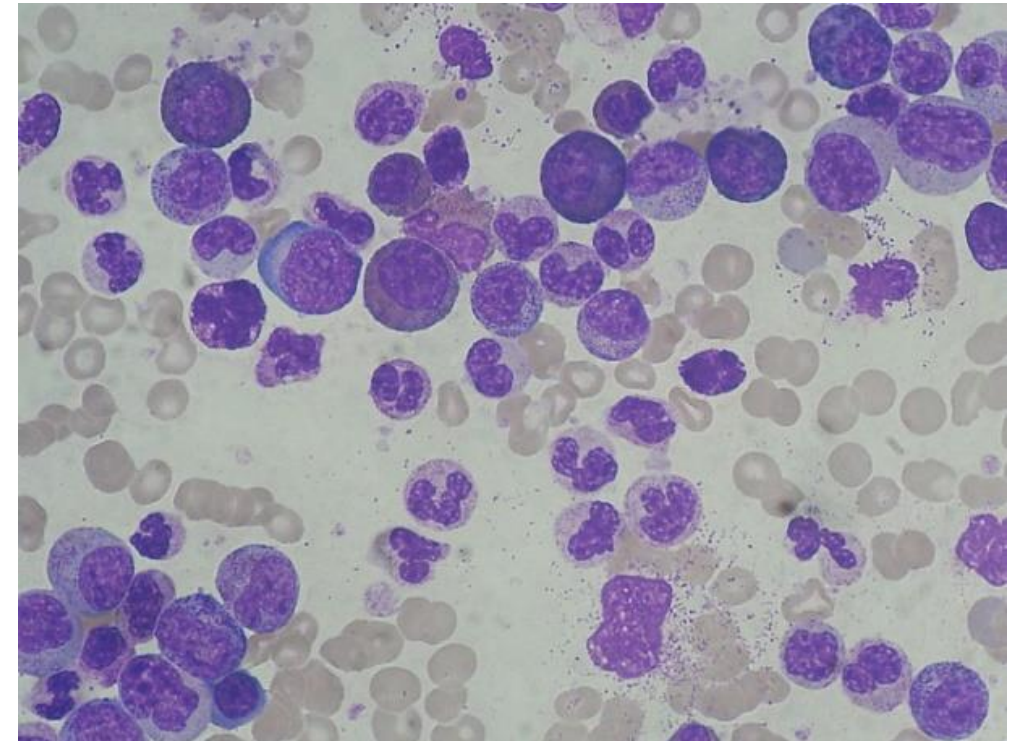
- Malignant disorder of myeloid progenitor cells
- Excess production of **granulocytes**
 - Neutrophils, basophils, eosinophils
- Classified as a myeloproliferative disorder



Chronic Myeloid Leukemia

Clinical findings

- Peripheral blood (chronic phase):
 - Leukocytosis (median WBC 100,000/microL)
 - ↑ neutrophils
 - ↑ **basophils** (rare finding!) and eosinophils
 - ↑ myeloblasts, promyelocytes, myelocytes, bands
- Classic finding: **myelocyte bulge**
 - Number myelocytes > metamyelocytes
 - Usually more metamyelocytes (more mature)
- Mild anemia
- Normal or increased platelets



Leukemoid Reaction

Left shift

- Normal response to infection
- More bands and neutrophils
- Must be distinguished from CML
- Bone marrow biopsy not usually helpful
- Leukemoid reaction
 - **Toxic granulation neutrophils** (dark, coarse granules)
 - Metamyelocytes > myelocytes (no myelocyte bulge)
- Key features CML:
 - Myelocyte bulge
 - No evidence of infection



	Normal	Infection
WBC	10,000/ μ L	17,000 / μ L
Neutrophils	55%	80%
Bands	5%	12%

Chronic Myeloid Leukemia

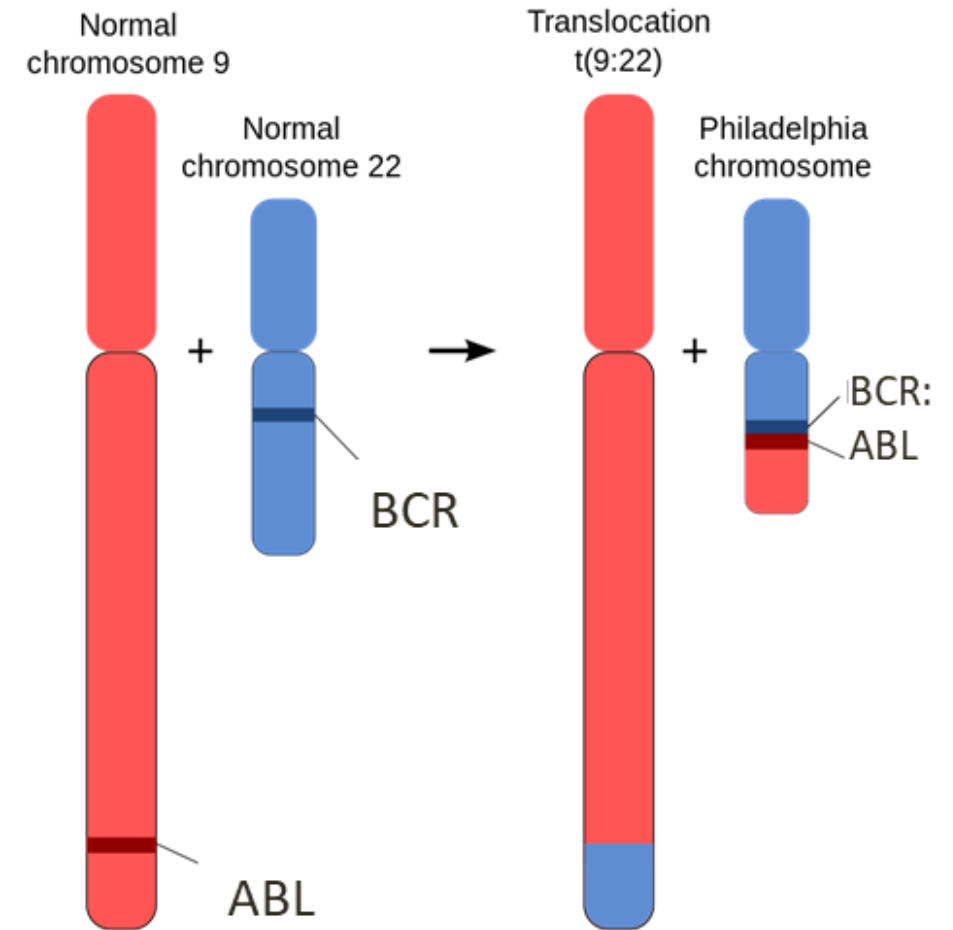
Leukocyte Alkaline Phosphatase Score

- Enzyme found in normal neutrophils
- Absent in neutrophils of CML
- Enzyme level assessed with **LAP score**
 - Low in CML
 - High in leukemoid reaction
- Largely replaced by genetic testing for CML chromosomal abnormalities

	CML	Leukemoid
LAP Score	Low	High
Toxic Granules	Absent	Present
Myelocyte bulge	Present	Absent
Evidence of infection	Absent	Present

Philadelphia Chromosome

- Genetic hallmark of CML
- 9;22 translocation
- **BCR-ABL fusion gene**
- Tyrosine kinase
- Long cell life → accumulation



Chronic Myeloid Leukemia

Clinical features

- Affects adults: median age ~ 50 years
- **Chronic phase (usually years)**
 - Can be asymptomatic (↑ WBC on blood testing)
 - Fatigue, malaise, weight loss, splenomegaly
 - Few peripheral blasts (usually < 2%)
- **Accelerated phase (usually months)**
 - Treatment failure (rising WBC)
- **Blast crisis**
 - Acute leukemia (> 20% blasts in periphery or marrow)
 - Usually myeloblasts (AML)
 - Less commonly lymphoblasts (ALL)

Chronic Myeloid Leukemia

Diagnosis

- CBC with CML findings (neutrophils, bands)
- No evidence of infection or leukemoid reaction
- Bone marrow biopsy: granulocyte hyperplasia
- Cytogenetics to identify t(9;22) or BCR-ABL gene



Chronic Myeloid Leukemia

Tyrosine Kinase Inhibitors

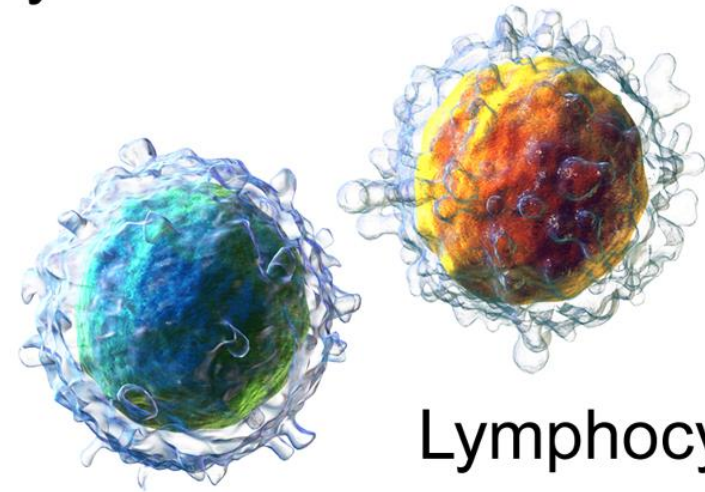
- Imatinib, dasatinib, nilotinib
- Used for treatment in CML in chronic phase
- Long-term control of disease (not curative)
- Bone marrow transplant may be used after failure



Chronic Lymphocytic Leukemia

CLL

- Malignant proliferation of **B lymphocytes**
- Excess circulating lymphocytes
- **Characteristic immunophenotype**
 - CD5+
 - CD20+ (“Co-express CD20 and CD5”)
 - CD19 or CD23



Lymphocytes

Small Lymphocytic Lymphoma

SLL

- Same malignant cells as CLL
- Differentiated by degree of lymphocytosis (\uparrow WBC)
- CLL: increased WBC
- SLL: normal or mild lymphocytosis
- SLL definition: lymphocyte count of < 5000
- CLL definition: lymphocyte count of > 5000

Chronic Lymphocytic Leukemia

Clinical features

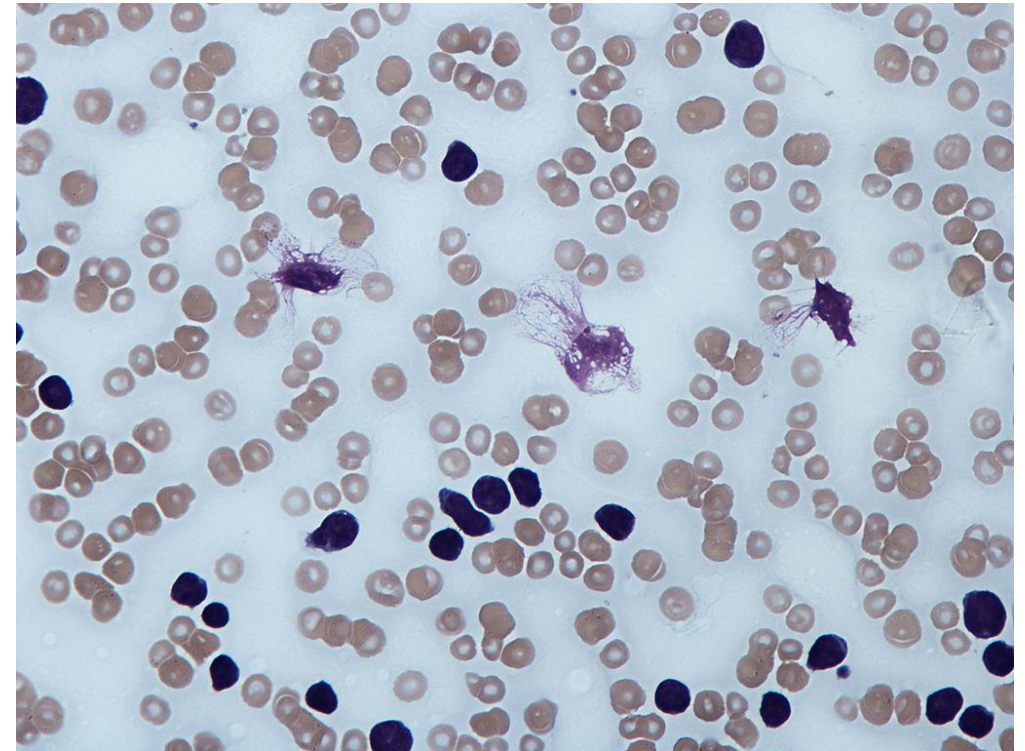
- Usually affects older adults (median age ~ 70)
- Patients often **asymptomatic**
 - Routine CBC: increased lymphocytes
- B symptoms in 5-10% of patients
 - Fevers, sweats
- Possible signs on exam
 - Lymphadenopathy
 - Hepatosplenomegaly



Chronic Lymphocytic Leukemia

Peripheral blood smear

- Classic finding: **smudge cells**
- Peripheral lymphocytes are fragile
- Disrupted during preparation of blood smear



Chronic Lymphocytic Leukemia

Diagnosis

- Suspected when CBC shows lymphocytosis
- Absolute B lymphocyte count ≥ 5000 for at least three months
- **Flow cytometry** of peripheral lymphocytes
 - Lymphocytes express CD5 plus B-cell antigens (CD19, CD20, CD23)



Chronic Lymphocytic Leukemia

Complications

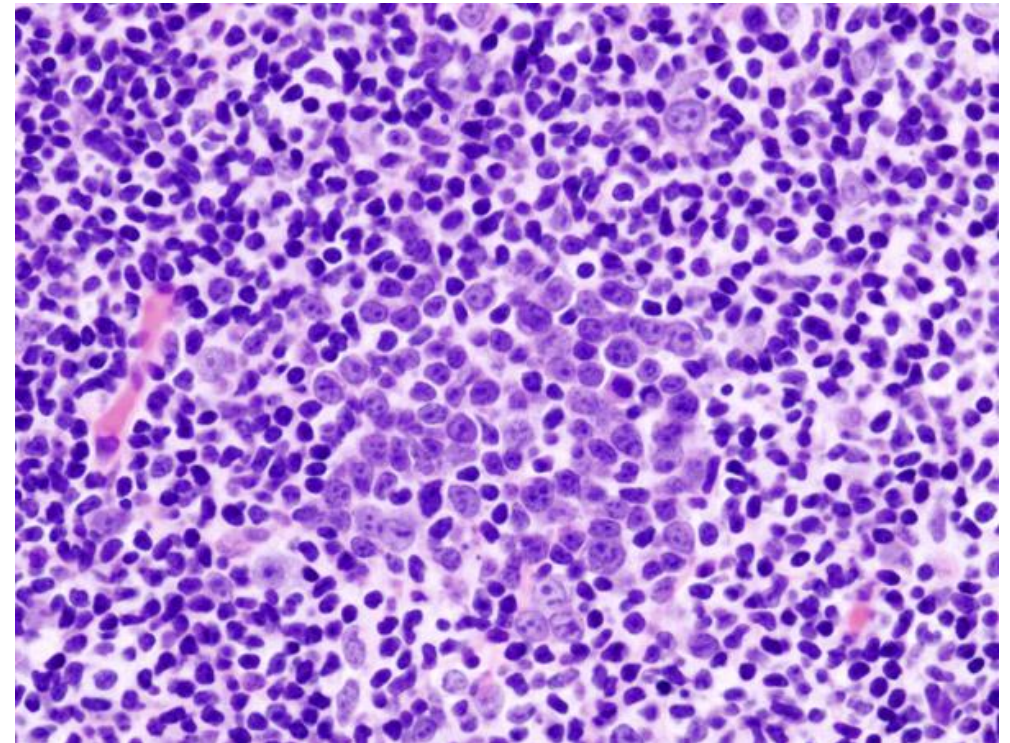
- **Cytopenias**
 - Neutropenia, anemia, thrombocytopenia
 - May be present at diagnosis
 - Usually not severe
- **Hypogammaglobulinemia**
 - ↓ IgG, IgA, IgM (one or more)
 - Some patients also have increased but dysfunctional gamma globulins
 - Increased susceptibility to **bacterial infections**
- **Autoimmune hemolytic anemia**
 - Autoantibodies may develop

Chronic Lymphocytic Leukemia

Complications

- May transform into **diffuse large B cell lymphoma**
- Classic presentation
 - Patient with known CLL
 - Rapid growth of single lymph node
 - Biopsy: diffuse large B cell lymphoma

Diffuse Large B cell Lymphoma



Chronic Lymphocytic Leukemia

Treatment

- Asymptomatic: **observation**
 - Even if lymphocytosis is extreme
- Treatment indications:
 - Bone marrow failure
 - Massive splenomegaly
 - Severe constitutional symptoms
- Treatment: chemotherapy or immunotherapy



Hodgkin Lymphoma

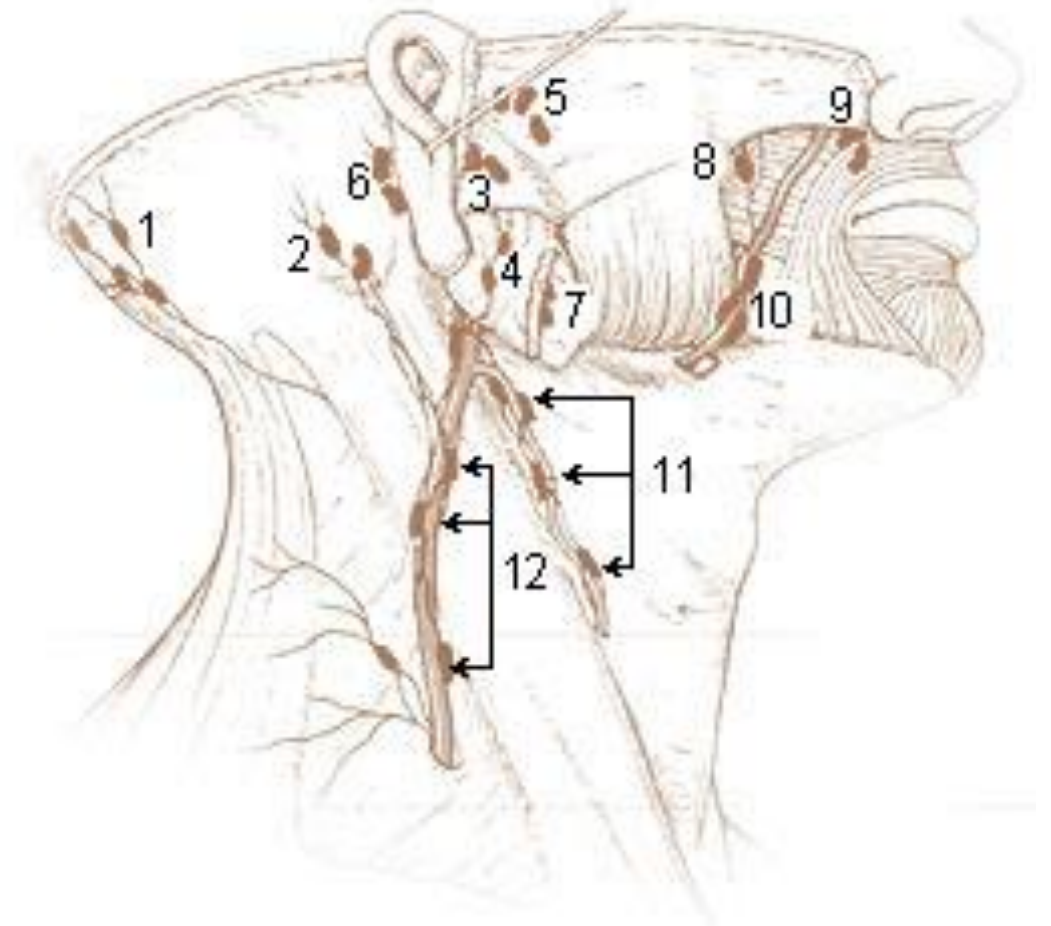
Jason Ryan, MD, MPH



Lymphomas

- Malignancies of lymphocytes (B cells, T cells)
- Often involve **lymph nodes**
- Also “extranodal” (skin, GI tract)

Lymph Nodes



Lymphomas

Signs and symptoms

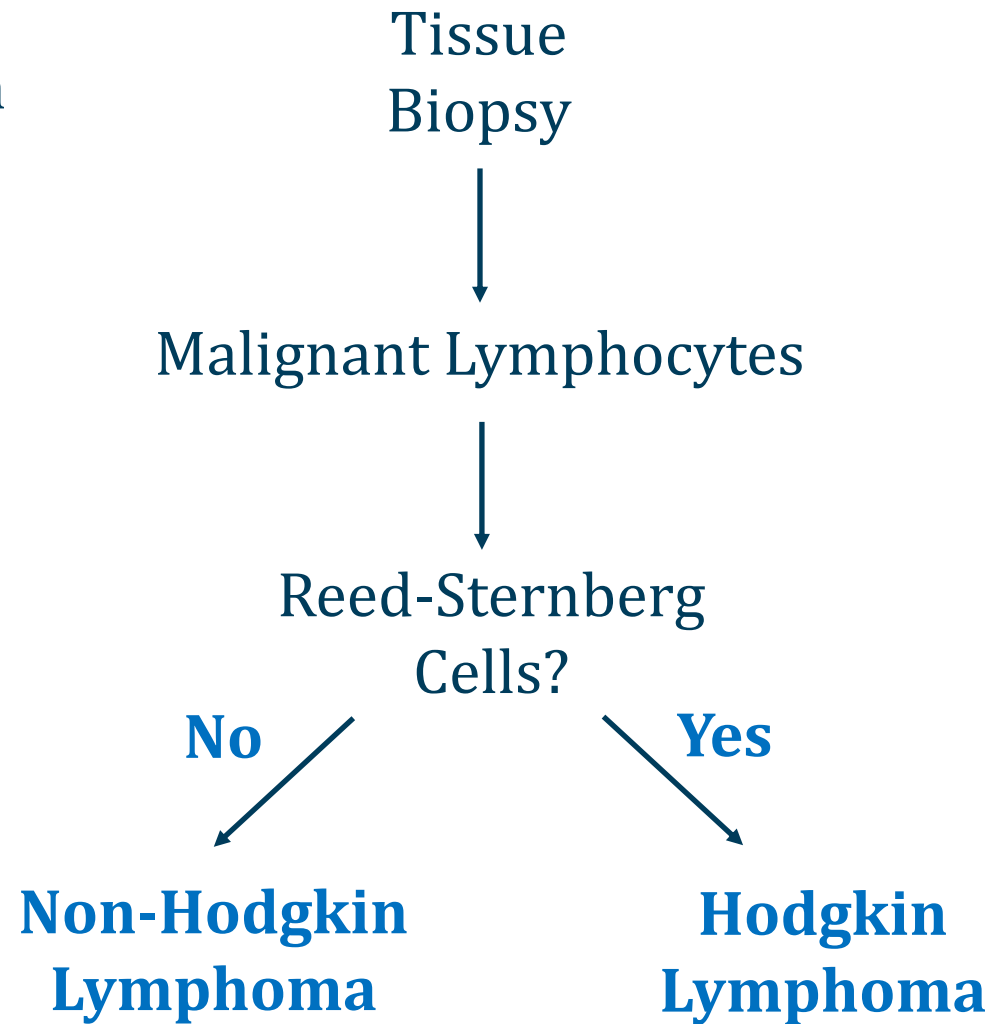
- Enlarged, **painless** lymph nodes
- Can present as **mediastinal mass**
- “B symptoms”
 - Systemic symptoms
 - Fever, chills, night sweats
- Pruritus
- Cell counts often normal
- LDH and ESR may be elevated

Lymphadenopathy



Lymphomas

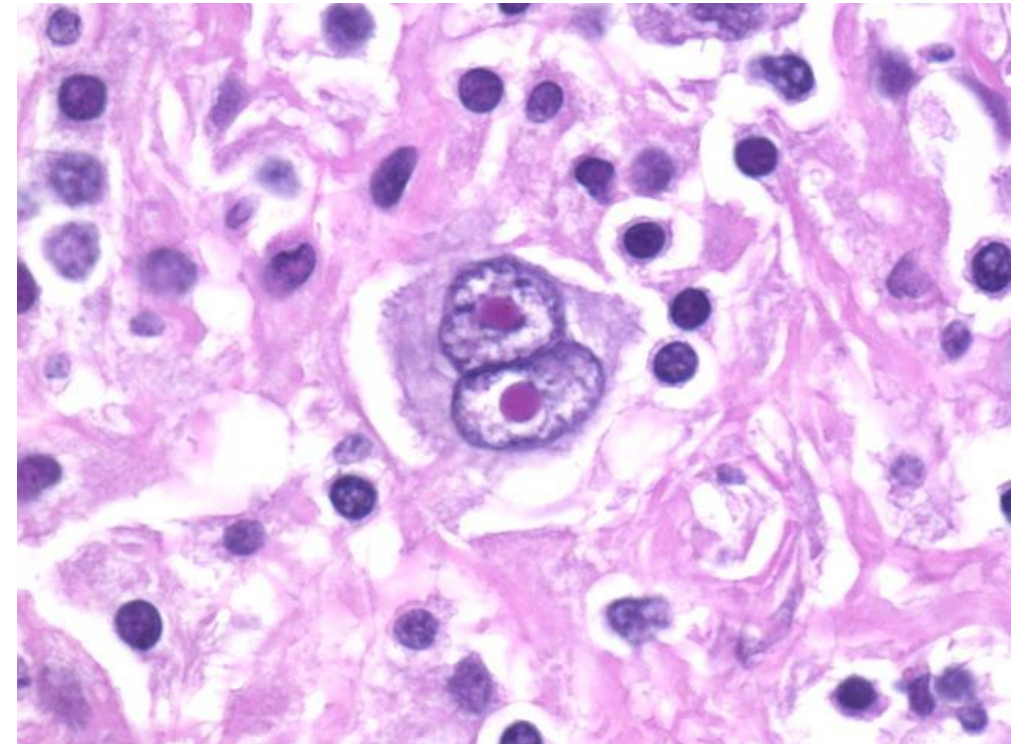
Hodgkin and Non-Hodgkin



Reed-Sternberg Cells

- Large cell with multi-lobed nucleus
 - Two halves; often mirror images (“owl-eyed”)
- Usually derive from B cells
- Usually **CD15+ and CD30+**
- Usually NOT positive for B cell markers
 - CD19, CD20, CD21, CD22

Reed-Sternberg Cell

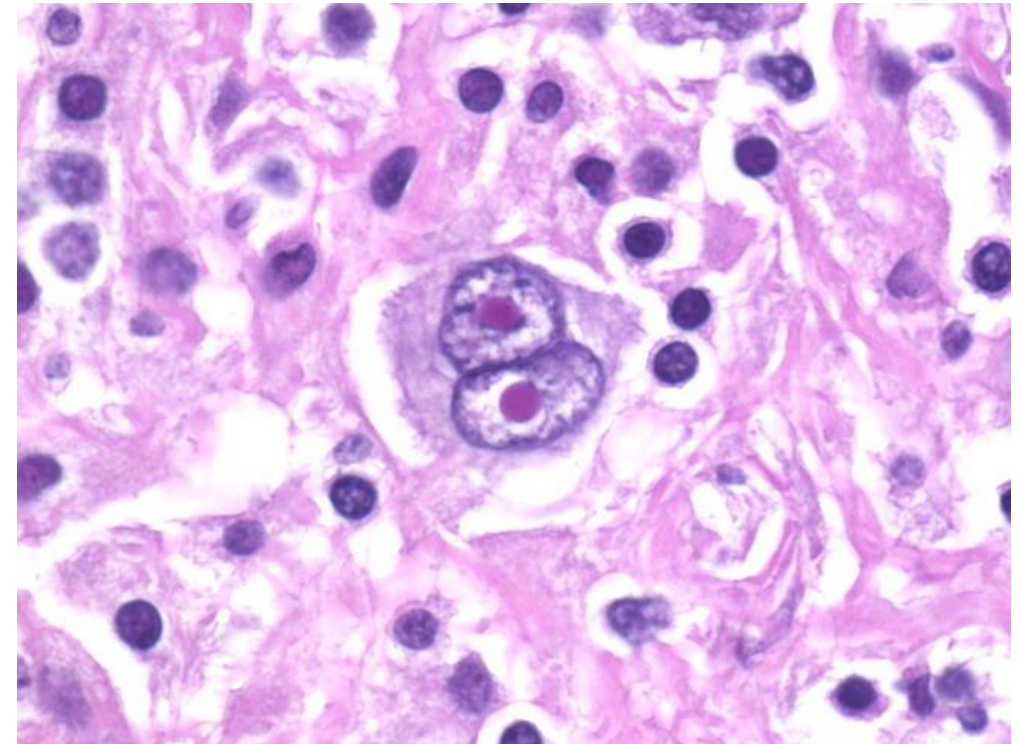


Hodgkin Lymphoma

Lymph node characteristics

- Malignant cell: **Reed-Sternberg cell**
 - A *minority* of cells in enlarged nodes (~1 to 5%)
- Release cytokines → generate **reactive cells**
 - Majority of cells in node are reactive
- **B symptoms common**
 - More than non-Hodgkin lymphoma

Reed-Sternberg Cell

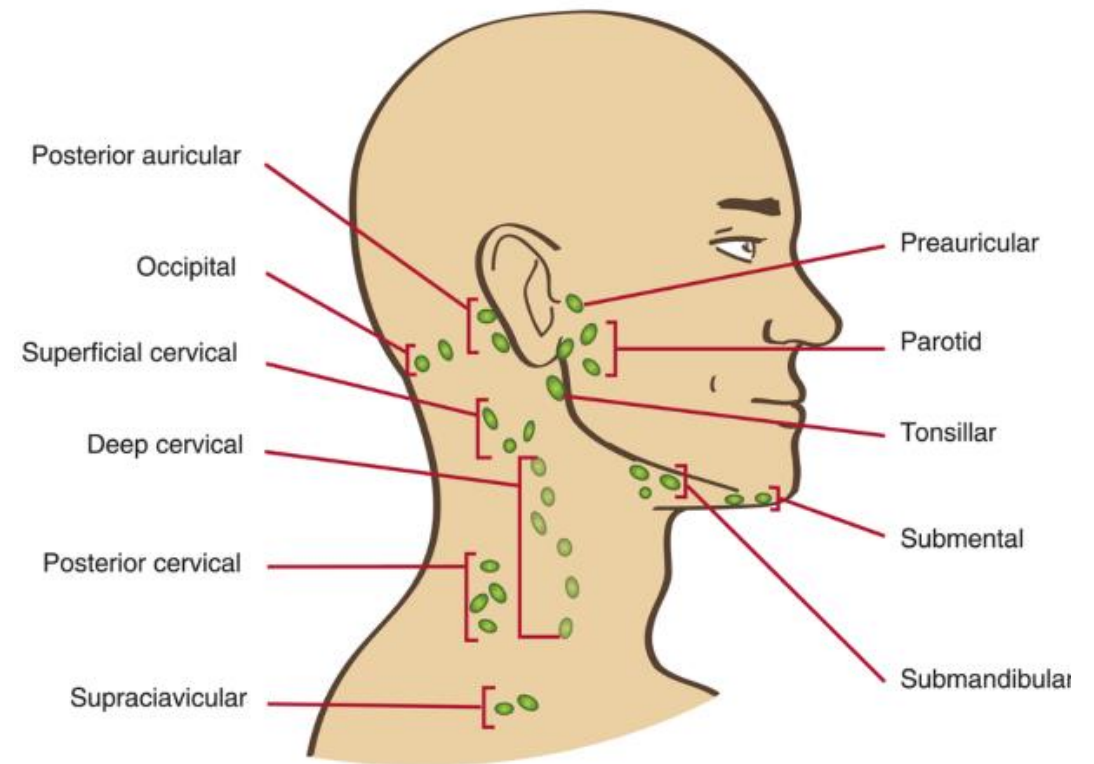


Hodgkin Lymphoma

Clinical features

- Commonly presents with **lymphadenopathy**
 - Neck is most common site
 - Enlarged cervical and/or supraclavicular nodes
 - Non-tender, firm, rubbery nodes
- Can present as **mediastinal mass**
 - Usually anterior mediastinum

Lymph nodes of the neck and head



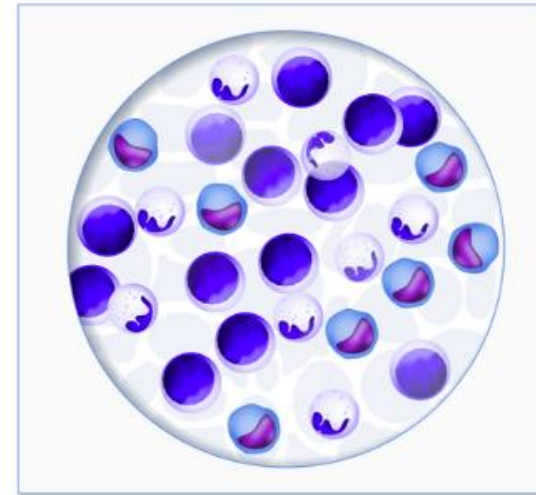
Hodgkin Lymphoma

Clinical features

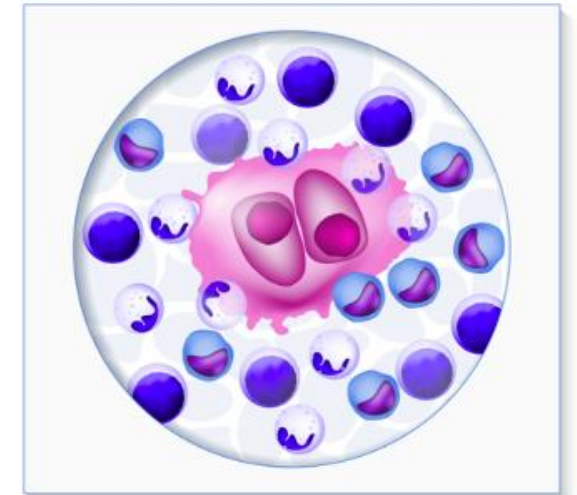
- B symptoms in ~40% of patients
- Spreads in a predictable manner
- Limited disease highly curable
- Stage is strongest predictor of prognosis

Hodgkin's lymphoma

Normal



Reed–Sternberg cells



Hodgkin Lymphoma

Differential diagnosis

- **Reactive lymphadenopathy**
 - Response to infection or inflammation
 - Other evidence of infection or inflammation often present
 - Sore throat, rash, joint pain, etc.
- Lymphoma: persistent, isolated lymphadenopathy
 - B symptoms may be present
 - No other evidence of infection or inflammation
- Key test: lymph node biopsy
 - RS cells indicate Hodgkin lymphoma



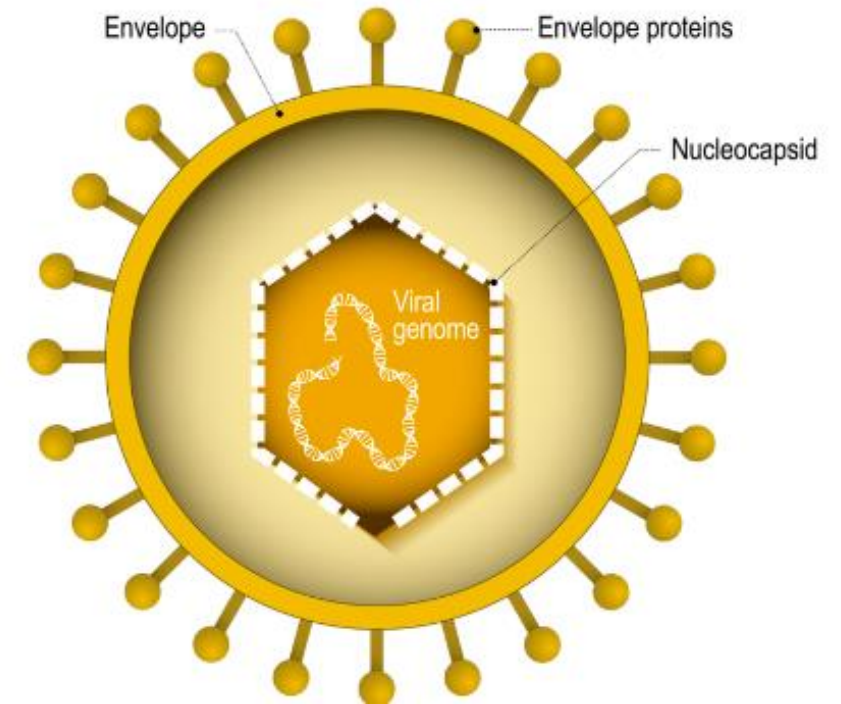
Hodgkin Lymphoma

Epidemiology and risk factors

- **Bimodal age distribution**
 - Peaks at age 20 and 65
 - May occur in young patients
- Prior EBV infection (virus infects B cells)
- Immunosuppression (HIV, transplant)
- Autoimmune disease: rheumatoid arthritis, SLE

EBV

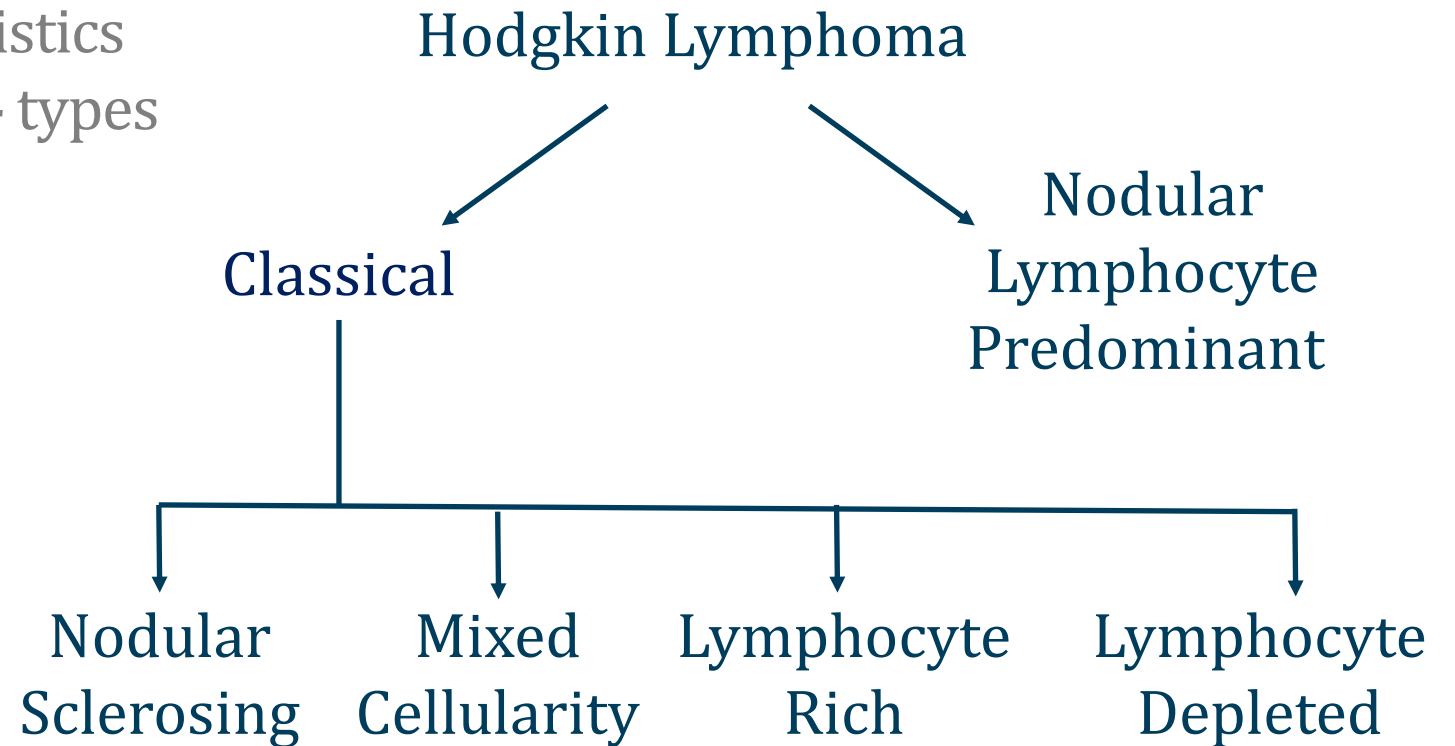
Epstein-Barr virus



Hodgkin Lymphoma

WHO classification

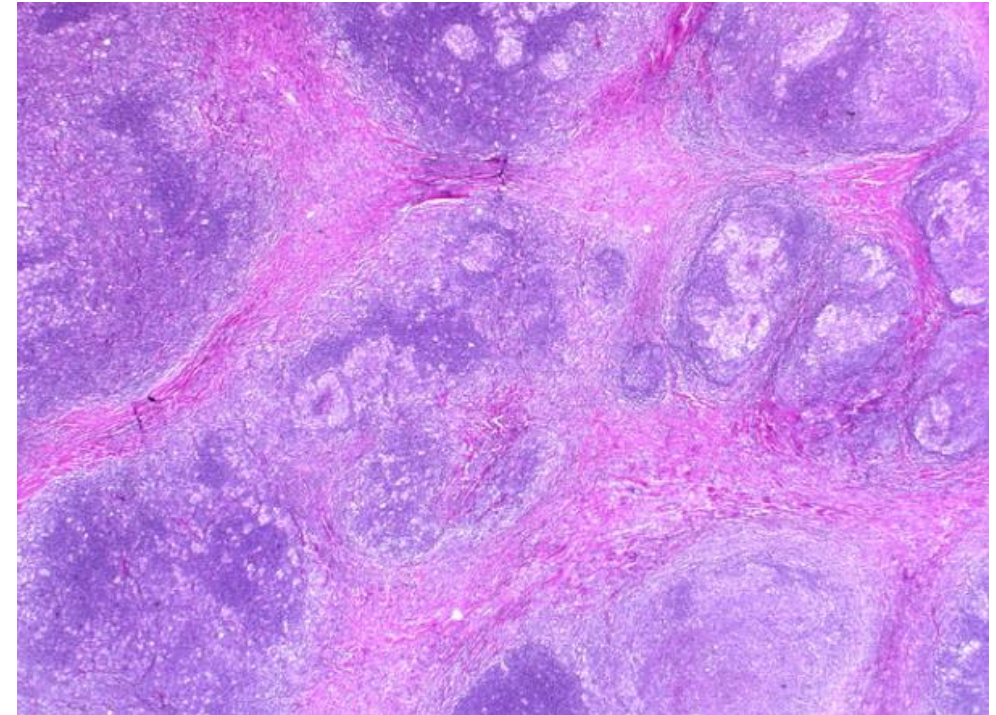
- Determined from **tissue biopsy**
- Most cases: **classical HL**
- NLP: unique RS cell characteristics
- Classical HL subdivided into 4 types



Hodgkin Lymphoma

Nodular sclerosing

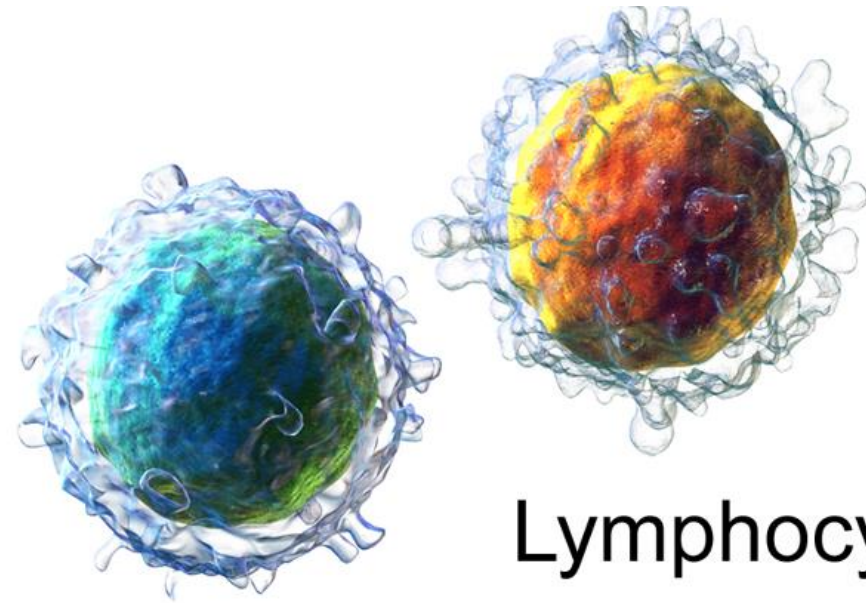
- Most common type Hodgkin lymphoma
- 60% to 80% of all cases
- Nodules of growth surrounded by fibrous bands
- Slow-growing (“indolent”)
- Symptom onset over weeks to months
- Good long-term survival



Hodgkin Lymphoma

Other classical subtypes

- Mixed cellularity
 - Eosinophils, neutrophils, macrophages, plasma cells
- Lymphocyte rich
 - Excellent prognosis
- Lymphocyte depleted
 - Poor prognosis



Lymphocytes

Hodgkin Lymphoma

Staging

- Usually done via **PET/CT scan**
 - CT scan: identifies nodes/masses
 - PET: Fluorodeoxyglucose (FGD-18) identifies metabolic activity
- Staging often done via **Lugano classification**

Stage	Features
I	One node or group of nodes
II	Two or more nodal groups on same side of diaphragm
III	Lymph structures on both sides of diaphragm
IV	Disseminated disease; one or more extranodal organs

Hodgkin Lymphoma

Treatment

- Many different regimens
- Chemotherapy plus radiation
- Common regimen: **ABVD**
 - Adriamycin (doxorubicin)
 - Bleomycin
 - Vinblastine
 - Dacarbazine

abvd

Non-Hodgkin Lymphoma

Jason Ryan, MD, MPH



Non-Hodgkin Lymphomas

- Diverse group of lymphocyte malignancies
- Many subtypes with different clinical patterns
- Often presents as lymphadenopathy
- Commonly extranodal
- Skin, GI tract, salivary glands, etc.

Lymphadenopathy



Lymphocyte Antigens

Primarily T-Cell Associated	Primarily B-cell Associated
CD1	CD10
CD2	CD19
CD3	CD20
CD4	CD21
CD5	CD22
CD7	CD23
CD8	

B cells → surface immunoglobulin

Non-Hodgkin Lymphoma

Classification

- **B and T cell malignancies**
 - Most are B cell disorders
 - Malignant cells may obliterate lymph node architecture
- More than two dozen subtypes per WHO
- Classified by:
 - B versus T cell
 - Cell size (small versus large)
 - Histologic appearance
 - Expression of markers (“immunophenotype”)
 - Genetics



Non-Hodgkin Lymphoma

Major Subtypes

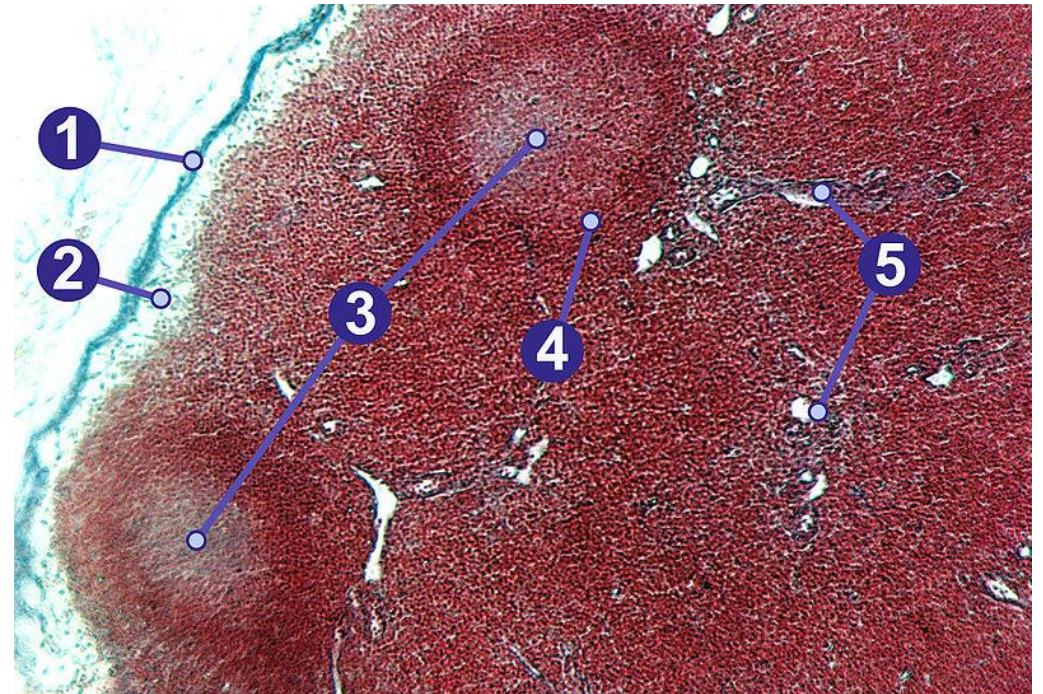
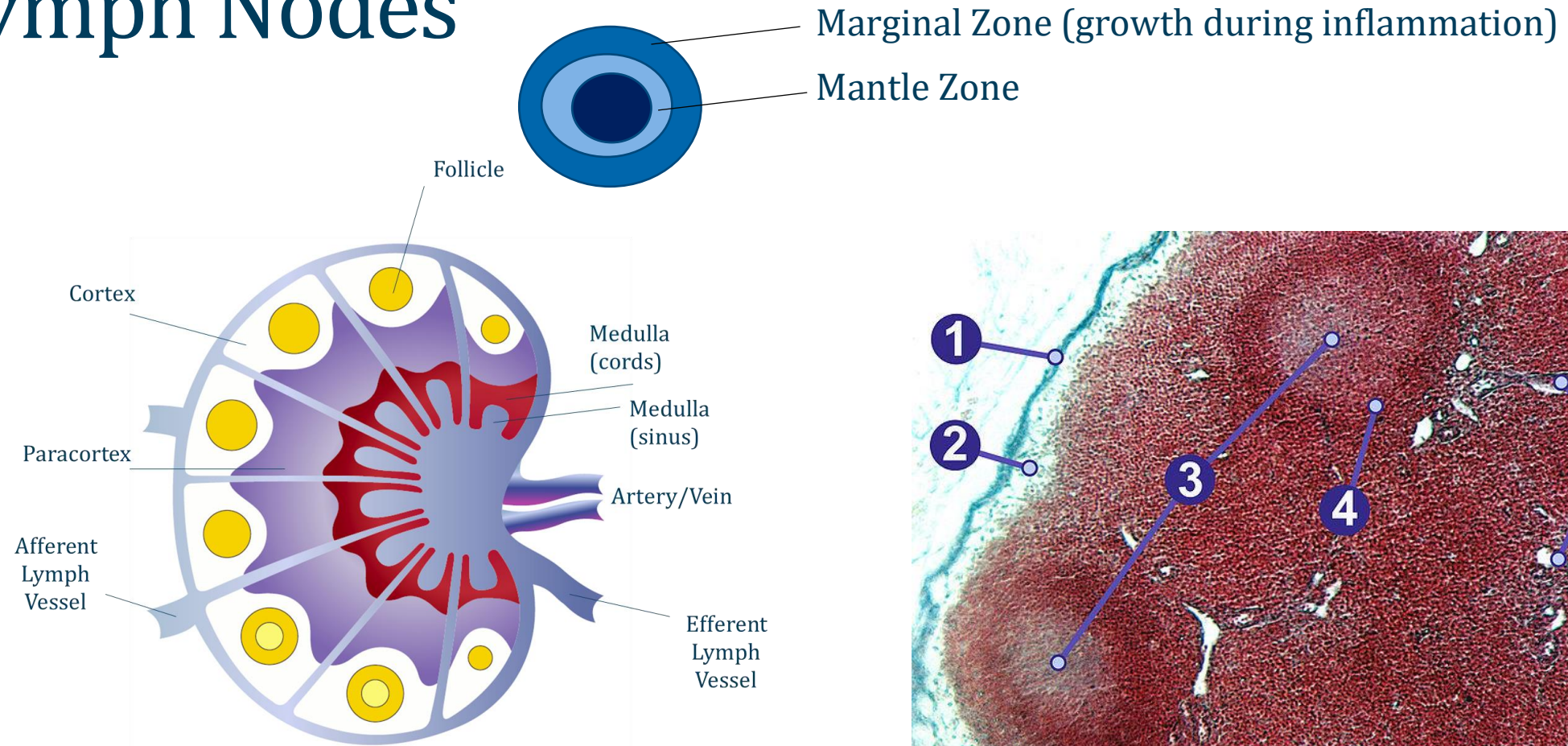
B-cell NH Lymphomas

Type	Aggressive	Indolent
Characteristics	Rapid growth B-symptoms common	Slower growth
Examples	Diffuse Large B cell Burkitt lymphoma Mantle cell lymphoma	Follicular Lymphoma Marginal zone lymphoma Small lymphocytic

T-cell NH Lymphomas

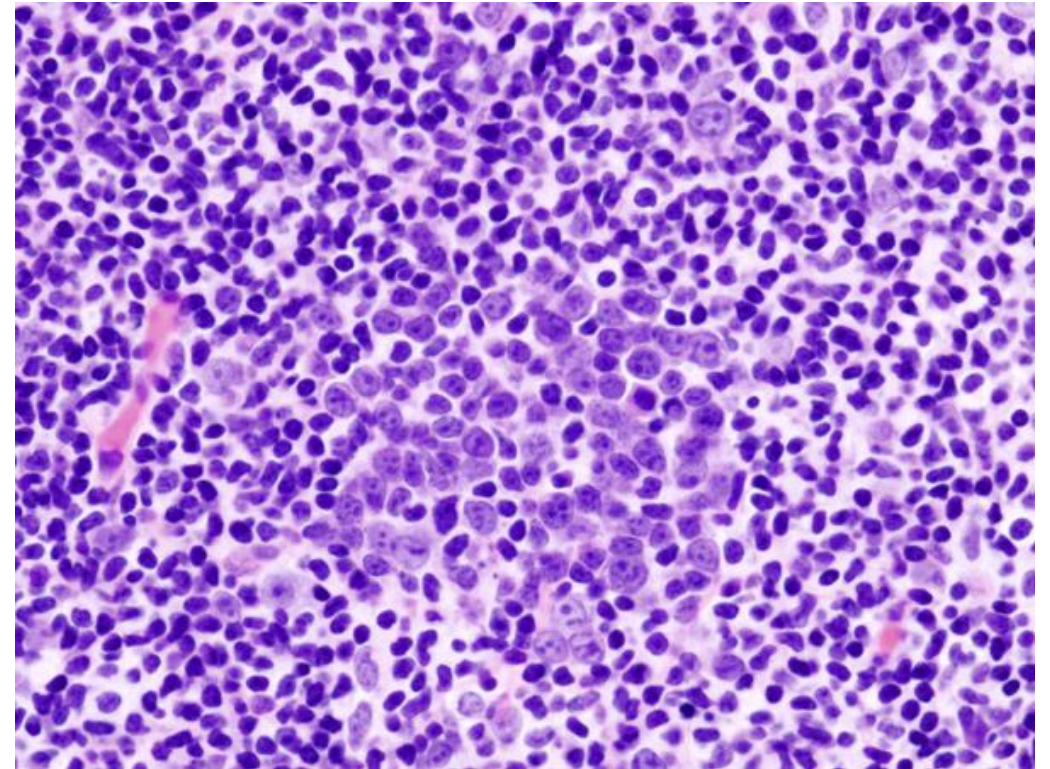
Adult T-cell Leukemia/Lymphoma
Cutaneous T-cell Lymphomas

Lymph Nodes



Diffuse Large B-cell Lymphoma

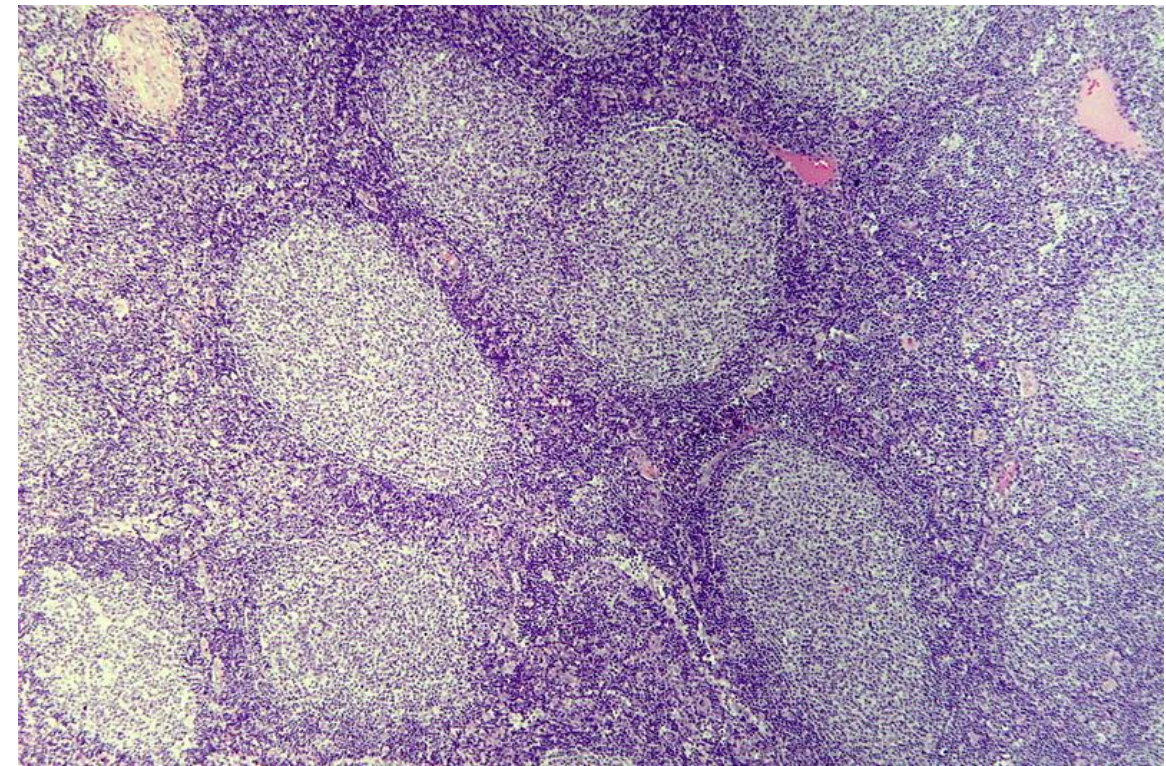
- **Most common non-Hodgkin lymphoma**
- B cell malignancy
 - Express CD19, CD20
 - Most cells express surface immunoglobulin
- Malignant cells obliterate follicles
- Median age at presentation is 64 years
- Occurs in HIV - AIDS defining malignancy
- Often treated with rituximab
 - Anti-CD20 antibody



Follicular Lymphoma

- **B cell malignancy**
 - Usually express CD19, CD20
 - Most cells express surface immunoglobulin
- Growth of **follicles**

Follicular Lymphoma

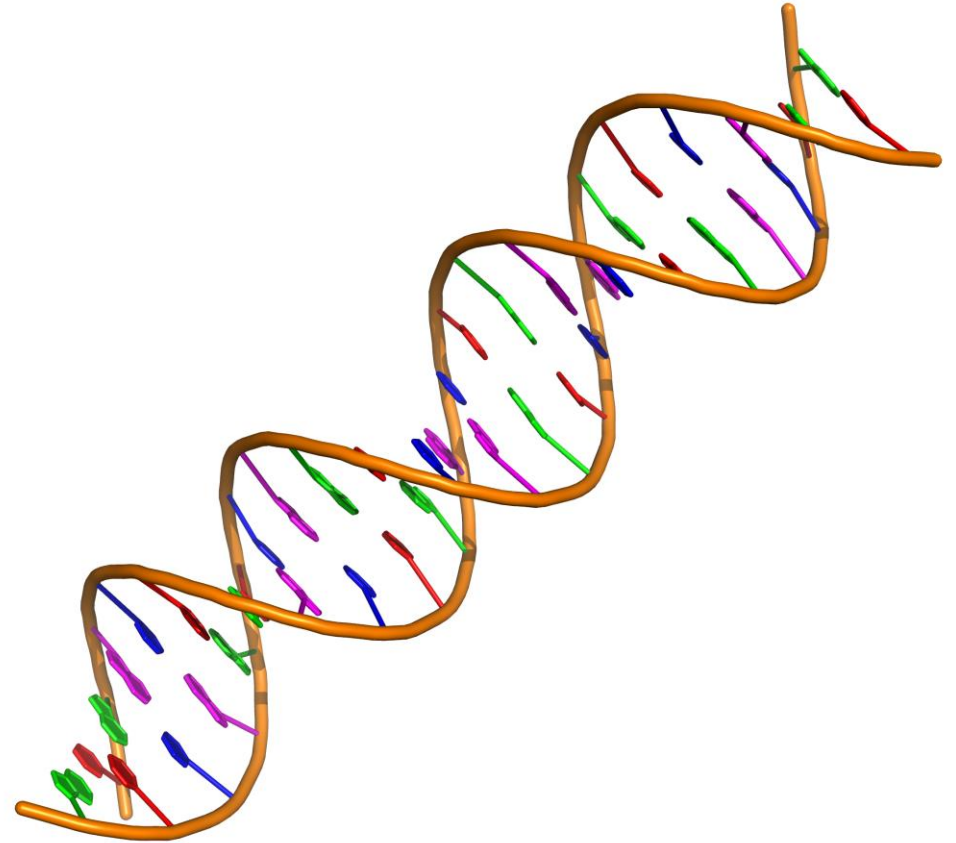


Patho/Wikipedia

Follicular Lymphoma

Genetics

- Genetic hallmark: **14;18 translocation**
- Overexpression of **BCL2**
 - Blocks apoptosis (“antagonist” of apoptosis)
 - Uncontrolled cell growth
- BCL2 expression used for diagnosis



Follicular Lymphoma

Clinical features

- Median age at diagnosis: **65 years**
- Indolent course: waxes/wanes for years
 - May have long-standing lymphadenopathy
 - Not all patients require treatment
 - Difficult to cure
- May transform into diffuse large B cell lymphoma
 - Poor prognosis

Follicular Lymphoma

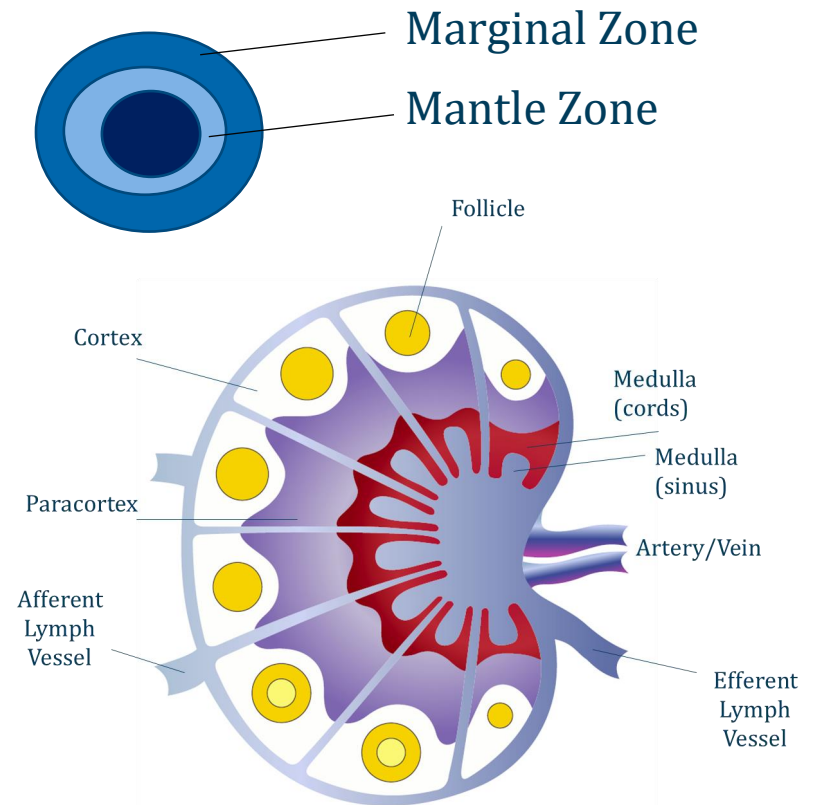
Differential diagnosis

- Must distinguish from **reactive lymphadenopathy**
 - Caused by infection → ↑ follicle growth
- Reactive lymphadenopathy (LAD)
 - Lack of BCL2 overexpression
 - B cell death → debris → macrophages

Lymphoma	Reactive LAD
Similar size/shape follicles	Varying size/shape follicles
Relative absence of macrophages	Tingible body (debris-laden) macrophages
++ BCL2 Staining	-- BCL2 Staining

Mantle Cell Lymphoma

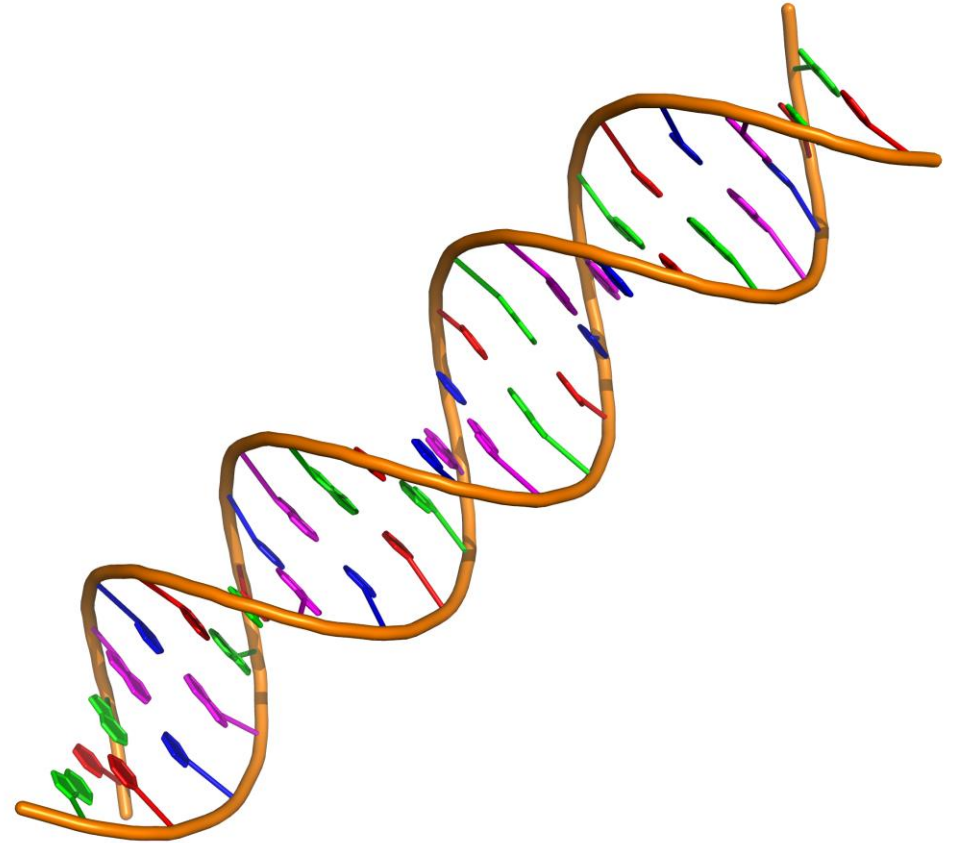
- Mantle zone: ring of lymphocytes surrounding germinal center of follicle
- **B cell malignancy**
 - Usually express CD19, CD20
 - Most cells express surface immunoglobulin
 - **Express CD5** ("Co-express CD20 and CD5")
- Median age at diagnosis: 68 years
- Median overall survival: 3 to 4 years (poor prognosis)



Mantle Cell Lymphoma

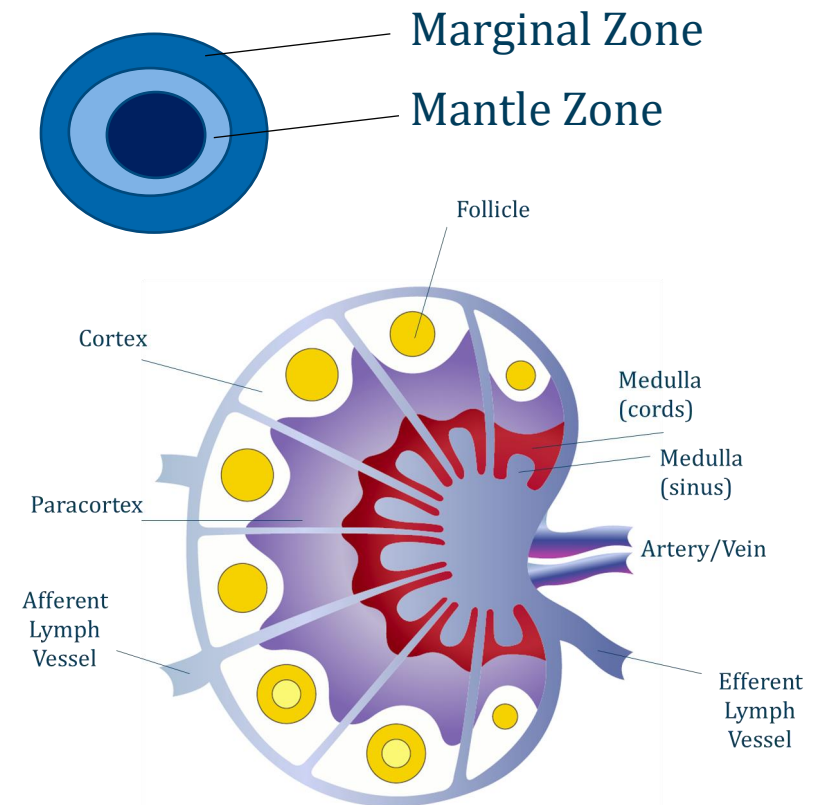
Genetics

- 50 to 65%: **11;14 translocation**
- Overexpression of **cyclin D1**
- Promotes cell growth and division
- Leads to uncontrolled cell growth
- Diagnosis: positive staining for cyclin D1



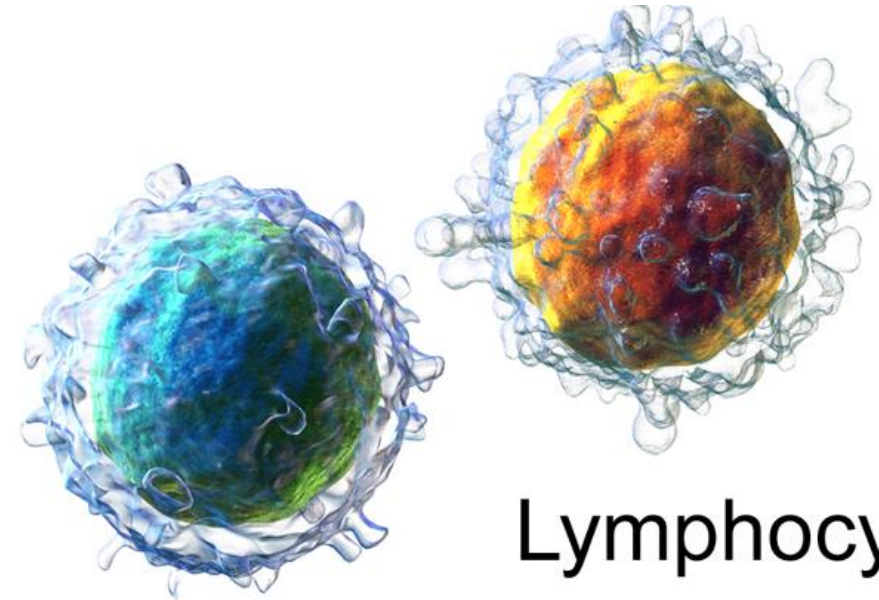
Marginal Zone Lymphoma

- Group of B cell malignancies
- Marginal zone forms from inflammation
- Often extranodal – GI tract, spleen
- Lymphoma in **chronic inflammatory disorders**
 - Salivary glands in Sjogren's
 - Thyroid gland in Hashimoto's thyroiditis
 - Stomach in chronic H. Pylori infection (MALToma)



Small Lymphocytic Lymphoma

- B-cells co-express **CD20 and CD5**
 - Similar markers to mantle cell lymphoma
 - Typically **negative for cyclin D1**
- Same malignant cells as chronic lymphocytic leukemia
 - Only difference is degree of lymphocytosis (↑WBC)
 - Peripheral blood: normal or mild lymphocytosis
- SLL definition: lymphocyte count of <5000
 - If >5000 → CLL

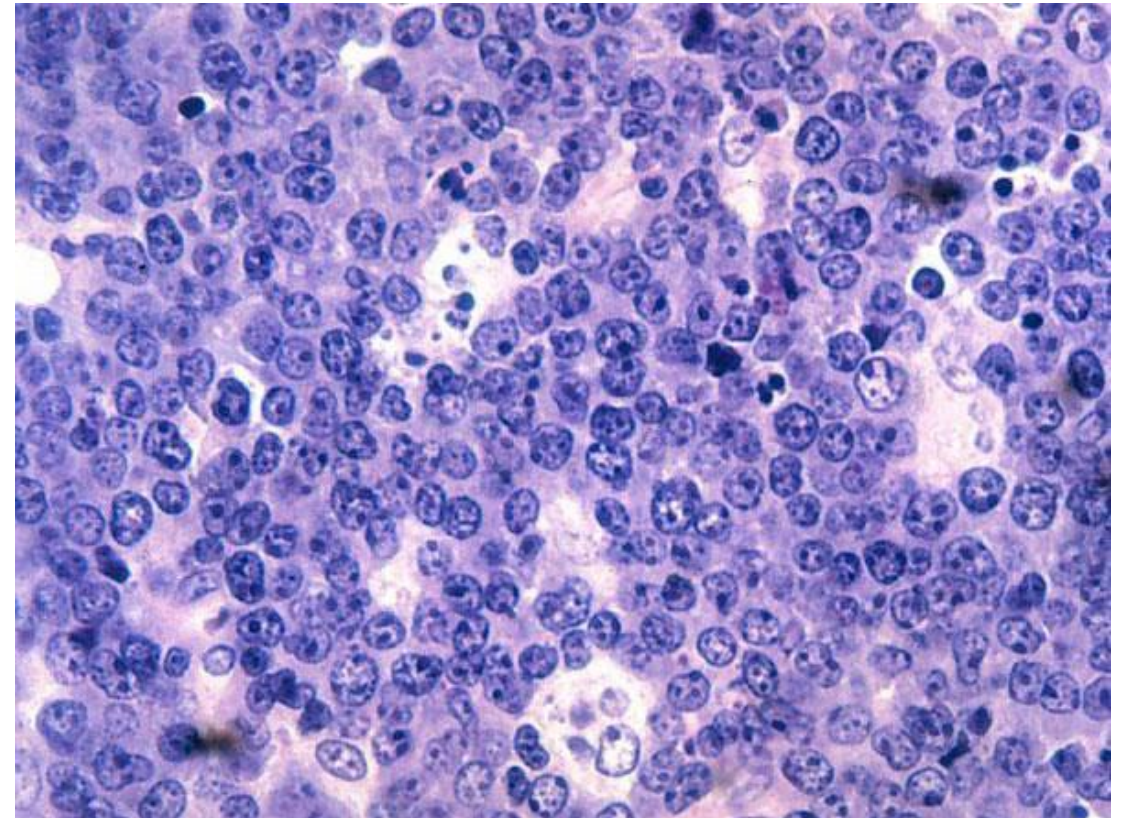


Lymphocytes

Burkitt Lymphoma

- **Highly aggressive B cell malignancy**
 - Usually express CD19, CD20
 - Most cells express surface immunoglobulin
- Key distinctions:
 - “Starry sky” morphology
 - Epstein-Barr virus (EBV)
 - C-myc translocation

Burkitt Lymphoma
Starry Sky Pattern



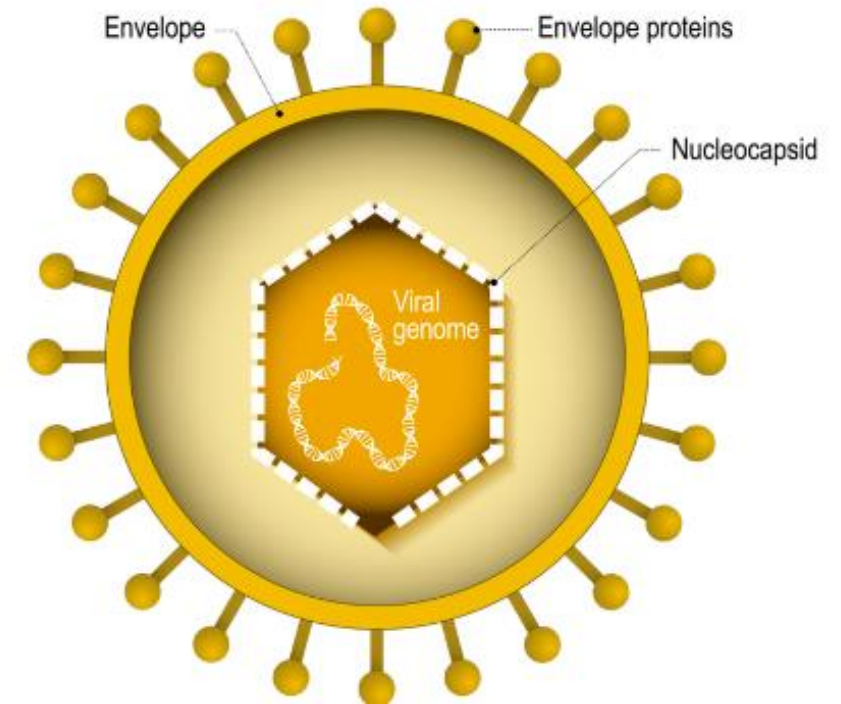
Burkitt Lymphoma

Associations

- **Epstein Barr virus (EBV) infection**
 - Nearly all endemic tumors associated with latent infection
 - Endemic tumors express CD21 (EBV receptor)
- **C-myc translocation**
 - Cell growth promoter
 - Gene translocated close to immunoglobulin genes
 - Overexpression → uncontrolled cell growth

EBV

Epstein-Barr virus



Burkitt Lymphoma

Clinical forms

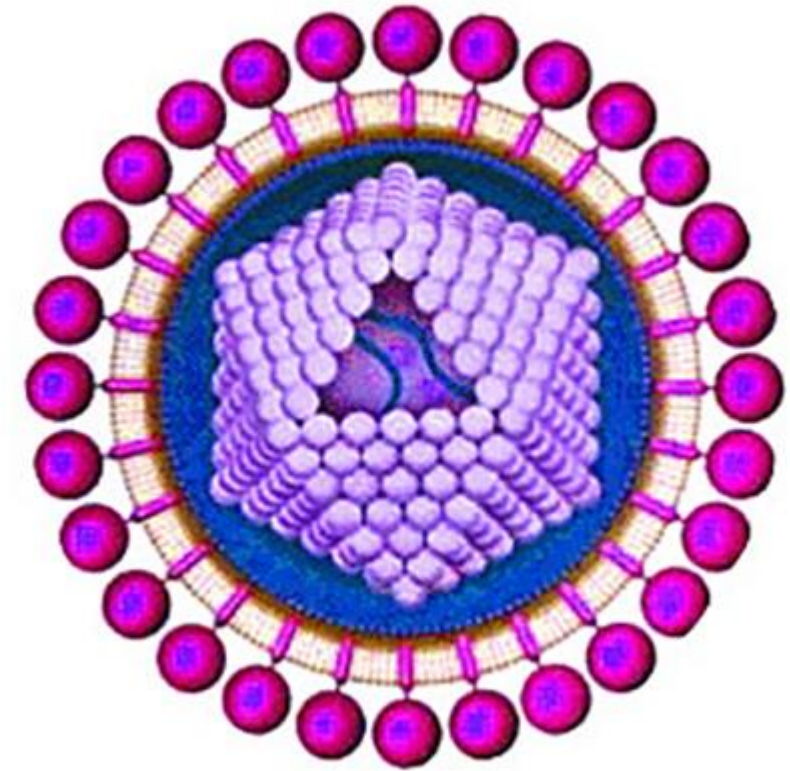
- **Endemic form**
 - Found in Africa and New Guinea
 - 30 to 50% of childhood cancer in some regions
 - Children four to seven years old
 - Male to female ratio ~ 2:1
 - Commonly presents as facial/mandible mass
- **Sporadic form**
 - Also occurs in children
 - Abdominal mass: ileocecum or peritoneum
- Immunodeficiency-related (variable features)



T-Cell Leukemia/Lymphoma

- **CD4+ T cell malignancy**
- Occurs with **HTLV-1 infection**
 - RNA Virus
 - Infects CD4+ T cells
- Key diagnostic test: **anti-HTLV1 antibodies**

HTLV-1 Virus



T-Cell Leukemia/Lymphoma

Clinical presentation

- Patient often from endemic HTLV-1 regions
 - Japan, Caribbean, West Africa
 - Northeast Iran, southeastern United States
- Lymphocytosis
- Lymphadenopathy
- **Skin lesions** (ulcers, nodules, papular rash)
- **Lytic bone lesions** with \uparrow calcium
 - Don't confuse with multiple myeloma
- Rapidly progressive symptoms \rightarrow usually fatal in months

Cutaneous T-cell Lymphomas

CTCL

- Skin disorders of **malignant CD4+ T-cells**
- Presents with **skin lesions**
- Various forms caused by similar malignant cells
- Localized disease: mycosis fungoides
- Diffuse systemic disease: Sezary syndrome

T cell



Cutaneous T-cell Lymphoma

Mycosis Fungoides

- Usually occurs 50 to 60 years of age
- **Skin patches, plaques, or growths**
- Varying size and shape
- Lesions progress slowly
- May mimic eczema or contact dermatitis
- Diagnosis: **skin biopsy**
 - Neoplastic T cells in epidermis and upper dermis
 - “Cerebriform nuclei”

Mycosis Fungoides

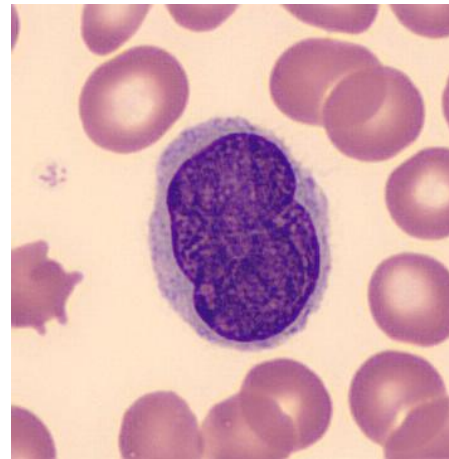


Cutaneous T-cell Lymphoma

Sezary syndrome

- T-cell lymphoma affecting skin of entire body
- **Widespread erythema**
 - Skin may be bright red
- Lymphadenopathy
- Associated leukemia
- **Sezary cells** in blood
 - Malignant T-cells
 - Multi-lobed “cerebriform” nucleus

Sezary Cell



El*Falaf

Sezary Syndrome



James Heilman/Wikipedia

Amyloidosis

Jason Ryan, MD, MPH



Amyloid

- Pathologic aggregate of **amyloid proteins**
- “Pathologic”: damages tissues
- Accumulate in extracellular space of tissues
- More than 20 proteins form amyloid
- Different proteins = different diseases



Amyloid

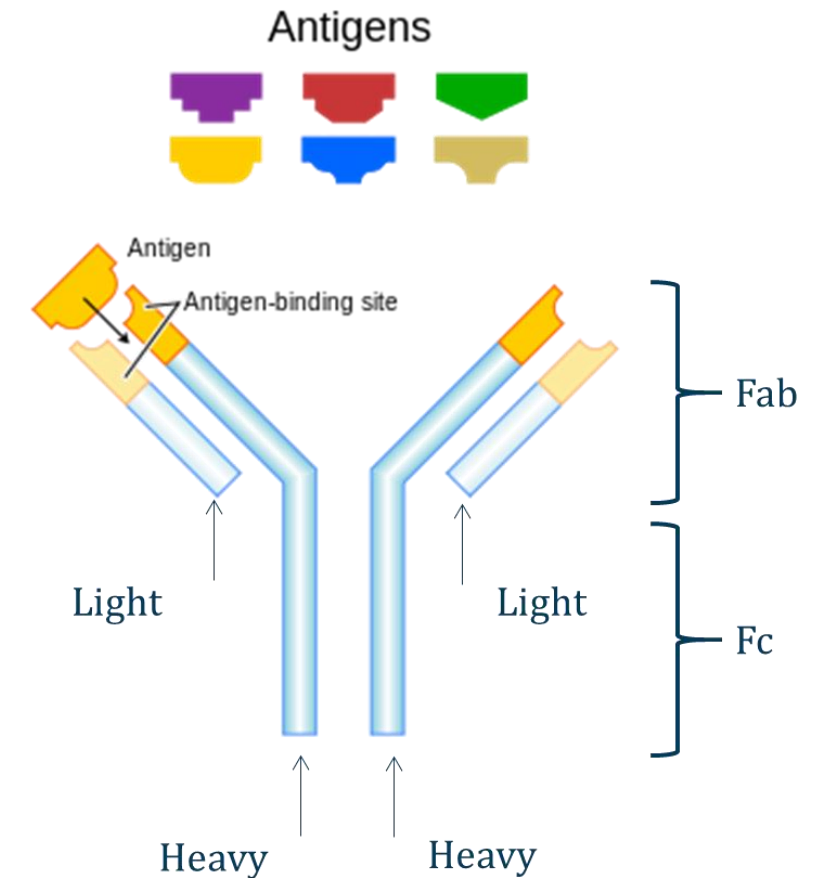
- Diffuse amyloid deposition = **amyloidosis**
- Localized amyloid deposition occurs in many disorders
 - Alzheimer's: beta amyloid
 - Cerebral amyloid angiopathy: beta amyloid
 - Type II diabetes: amylin deposits in pancreas



Systemic Amyloidosis

Subtypes

- Primary (AL) amyloidosis: light chains
- Secondary (AA) amyloidosis: serum amyloid A
- Dialysis-related amyloidosis: beta-2 microglobulin
- Age-related systemic amyloidosis: transthyretin
- Familial amyloidosis: many types
 - Most common: abnormal transthyretin gene



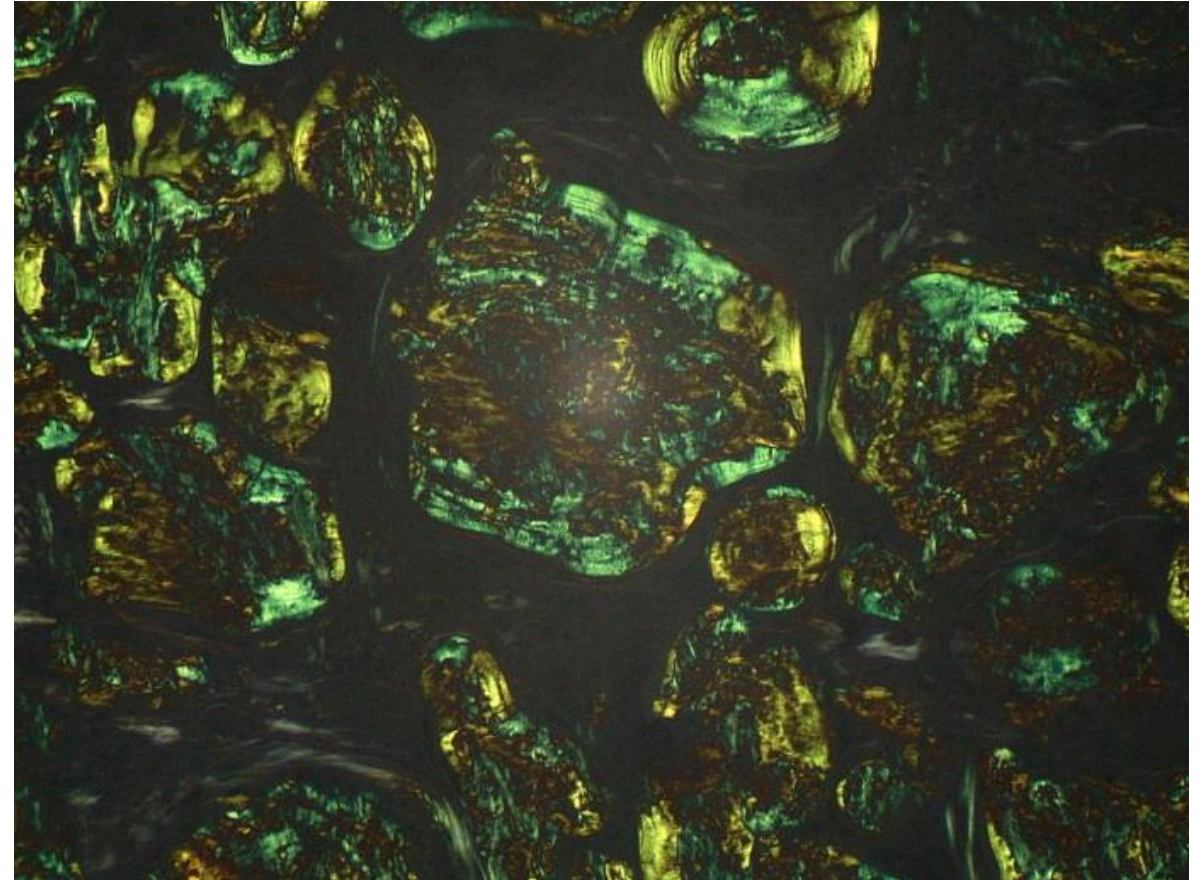
Transthyretin

- Formerly called prealbumin
- Transports thyroid hormone and retinol (vitamin A)
- Amyloidosis: amyloid transthyretin (ATTR)
- Mutant form seen in hereditary amyloidosis
- Normal transthyretin seen in **age-related amyloidosis**

Amyloid Proteins

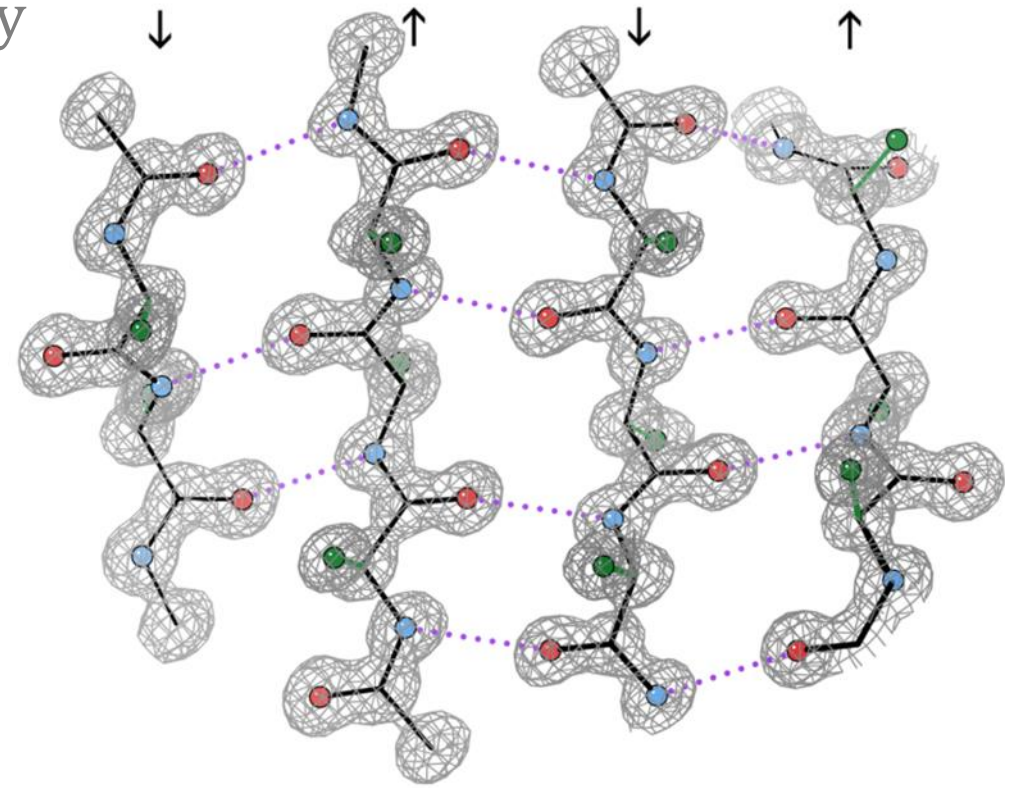
- Pink on standard microscopy
 - Similar to collagen, fibrin, other proteins
- **Apple-green birefringence**
 - Specialized stain for detection
 - Congo red
 - Pink under ordinary light
 - Apple-green under polarized light

Apple-Green Birefringence



Amyloid Proteins

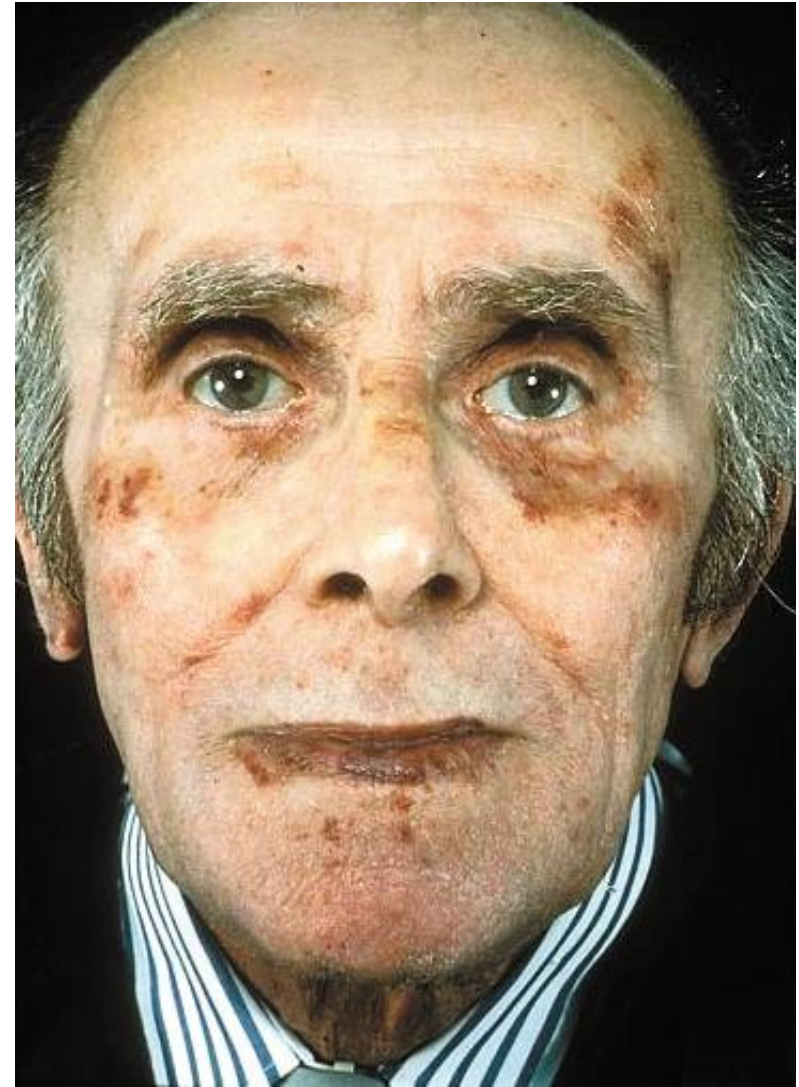
- Forms **beta-pleated sheets**
- Detected by crystallography and spectroscopy
- Responsible for Congo Red staining



Amyloidosis

Clinical features

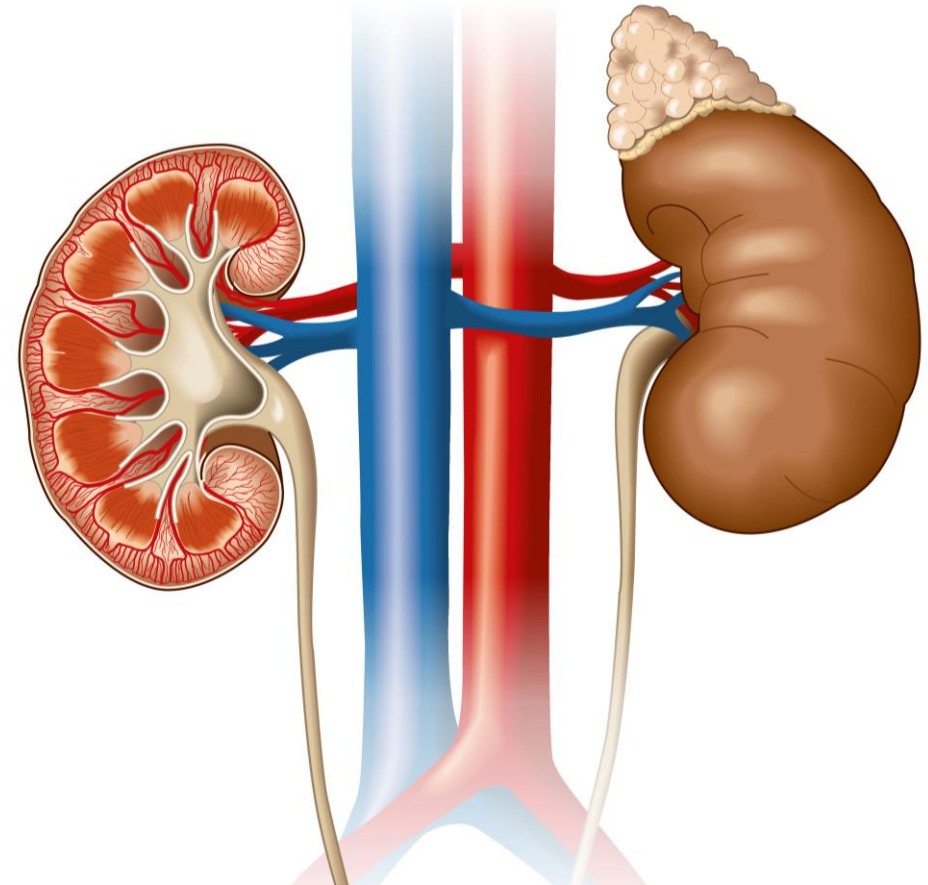
- May involve almost any tissue/organ
- Skin: **facial or neck purpura**
 - Blood vessels: bleeding
 - Classically periorbital purpura (raccoon eyes)
- Muscles: **enlarged tongue**
- Peripheral neuropathy
- **Hepatosplenomegaly**
- **Carpal tunnel syndrome**



Amyloidosis

Clinical features: kidney

- Most commonly involved organ
- May lead to renal insufficiency
- May cause proteinuria and nephrotic syndrome
- Common exam scenario
 - Proteinuria and renal insufficiency
 - Facial purpura
 - Enlarged tongue



Amyloidosis

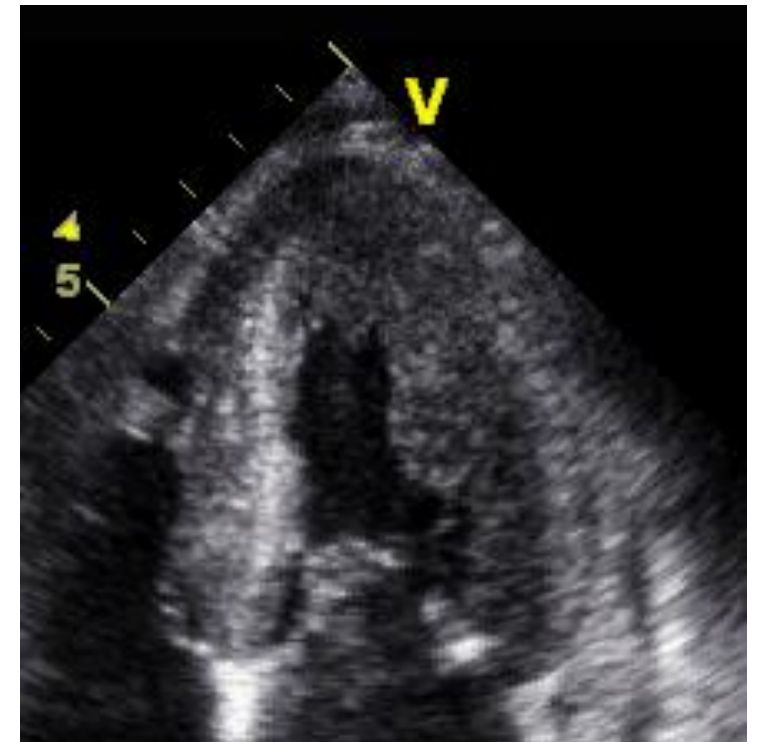
Clinical features: heart

- Common with primary and age-related amyloidosis
- Arrhythmias, sudden death
- Increased wall thickness with low voltage
- Causes a **restrictive** cardiomyopathy
- Heart failure preserved ejection fraction (HFpEF)

Low EKG voltage



Increased LV Wall Thickness



Amyloidosis

Diagnosis with biopsy

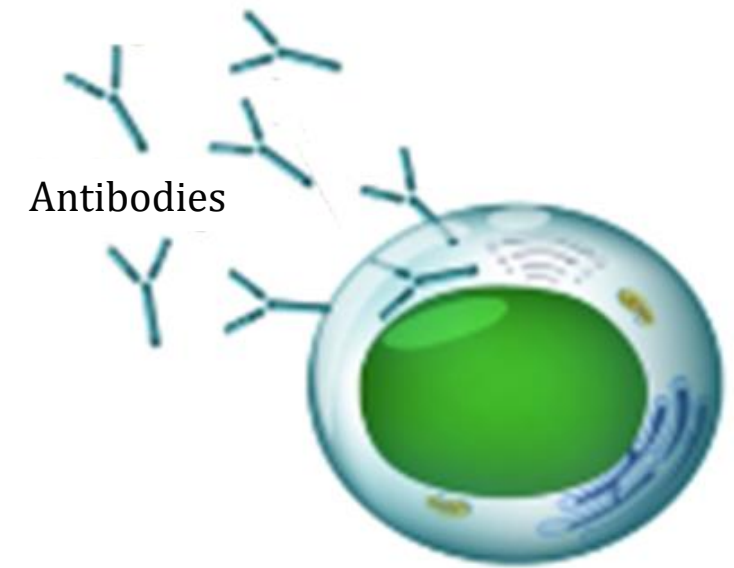
- Can be done on any involved organ
- **Abdominal fat pad**
 - Easy to access
 - Low risk procedure
 - Good sensitivity



Primary Amyloidosis

AL Amyloidosis

- **Plasma cell malignancy**
- Amyloid formed from **light chains**
- Can occur alone or with a plasma cell disorder
 - Multiple myeloma
 - Waldenstrom's
 - Some forms of lymphoma
- Presents with multi-system amyloidosis findings
- Bone marrow biopsy: monoclonal plasma cells
- Can be treated with stem cell transplantation



Primary Amyloidosis

AL Amyloidosis

- Amyloid deposition in tissues on biopsy
- Must identify that amyloid is **light-chain-related**
 - Several techniques
 - Mass spectrometry
 - Immunoelectronmicroscopy
- Must identify **monoclonal plasma cell activity**
 - Serum or urine monoclonal protein
 - Abnormal free light-chain ratio
 - Bone marrow biopsy: clonal plasma cells

Suggestive Clinical Features



Tissue Biopsy



Amyloid Proteins
Special testing: light chains



Evidence of
Monoclonal plasma cells

Secondary Amyloidosis

AA Amyloidosis

- Occurs in chronic inflammatory conditions
- Rheumatoid arthritis, ankylosing spondylitis, inflammatory bowel disease
- Chronic osteomyelitis and tuberculosis
- Amyloid: **serum amyloid A (SAA) proteins**
 - Acute phase reactants
 - Many roles in inflammatory response

Rheumatoid Arthritis



Secondary Amyloidosis

AA Amyloidosis

- Suspect in patients with **chronic inflammatory conditions**
- Most commonly involves the kidney
- Often presents as nephrotic syndrome
- Treatment: control underlying inflammatory disorder

Suggestive Clinical Features



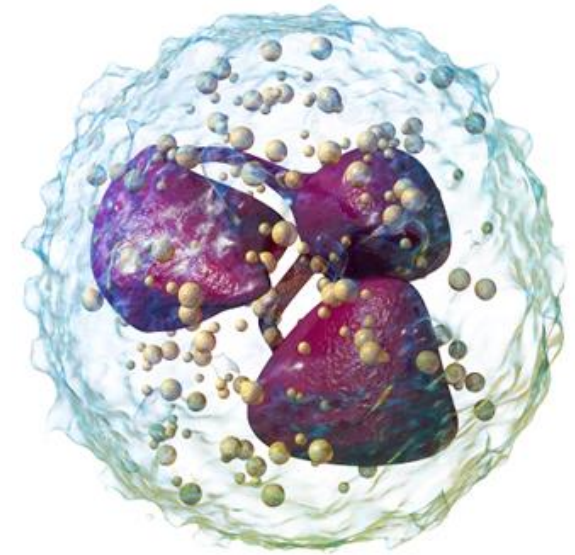
Tissue Biopsy



Amyloid Proteins
Special staining: SAA proteins

Familial Mediterranean Fever

- Rare hereditary disorder
- Inflammatory disease of **neutrophils**
- Recurrent episodes of **fever** and **inflammatory pain**
- “Serosal” inflammation
 - Abdominal pain, pleuritis, pericarditis
- Major cause of death: **secondary (AA) amyloidosis**
- Treatment: **colchicine**
 - Good evidence for efficacy
 - Protects from development of amyloidosis



Neutrophil

Dialysis-related Amyloidosis

- Complication of **chronic kidney disease**
- Caused by **β 2 microglobulin**
 - Found on cell surfaces
 - Component of MHC class I molecules
 - Dialysis does not effectively remove β 2 microglobulin
- Bones, joints, tendons
- Shoulder pain
- Carpal tunnel syndrome
- Bone cysts

Amyloid Deposits in Wrist



Dialysis-related Amyloidosis

Diagnosis and treatment

- **Usually diagnosed clinically**
 - Patient with CKD on dialysis
 - Shoulder pain or carpal tunnel syndrome
 - Bone cysts on imaging
- Fat pad biopsy unreliable
- May respond to hemodialysis modifications
 - Use of specific membranes
 - Removes beta2-microglobulin
- Definitive treatment: kidney transplant

Hemodialysis

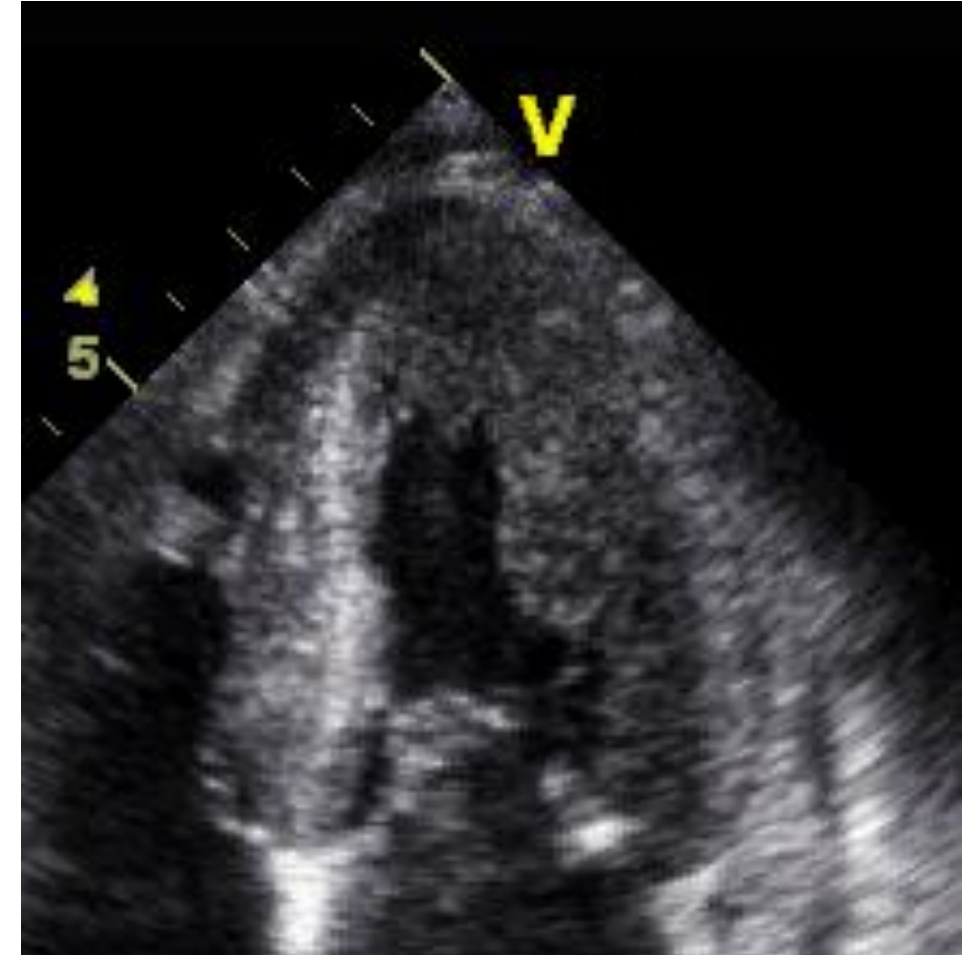


Age-related Amyloidosis

Senile Amyloidosis

- Wild-type (normal) transthyretin
- Form of **ATTR amyloidosis**
 - Wild-type ATTR amyloidosis
- Usually develops > 70 years old
- Predominantly occurs in the **heart**
- Rarely other significant organ involvement
- Major clinical effect: **HFpEF**

Increased LV Wall Thickness

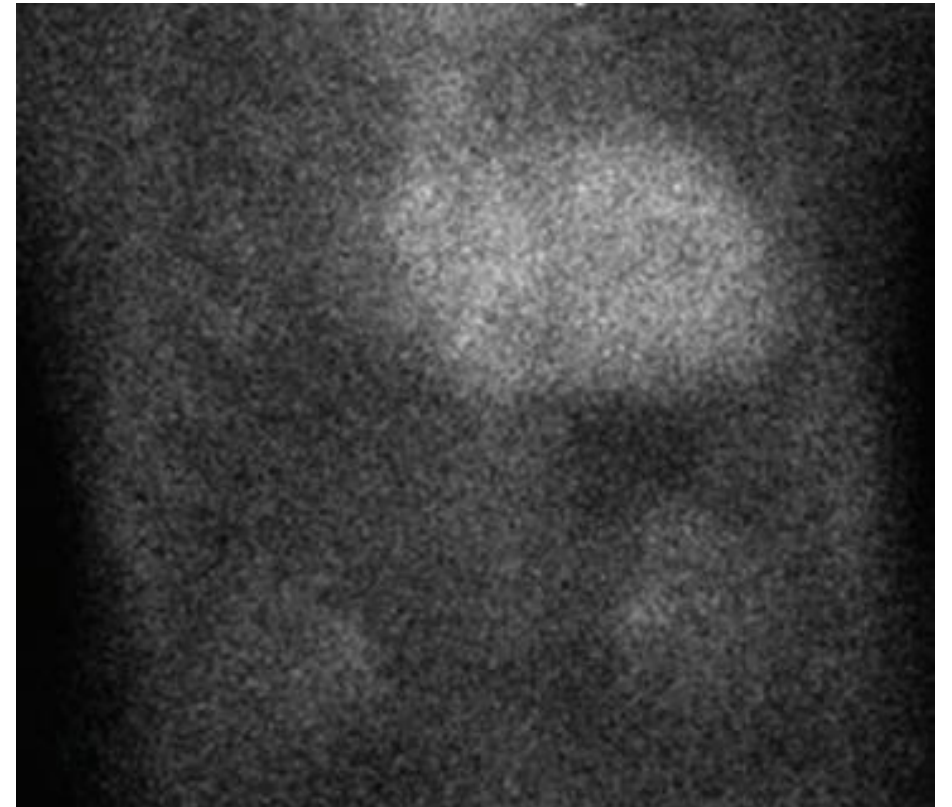


Age-related Amyloidosis

Diagnosis and treatment

- Specialized diagnostic testing for ATTR amyloid
 - Myocardial biopsy rarely performed
 - Cardiac MRI
 - Technetium Tc-99m pyrophosphate nuclear scan
- **Tafamidis**
 - Stabilizes transthyretin
 - Reduces formation of amyloid

99mTc-PYP Uptake Scan



Familial Amyloidosis

- Autosomal dominant
- Several gene mutations that code for amyloid proteins
- Many forms are related to **transthyretin**
 - Hereditary ATTR amyloidosis
- Mutant proteins produced by liver
- Can be treated with **liver transplant**
- Symptoms in adulthood

