The CBC and Me:
Identifying and Evaluating Abnormalities

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Disclosure

• No conflicts of interest to disclose.
Learning Objectives

1. Describe the diagnostic testing and clinical approach to pediatric anemia.

2. Identify WBC abnormalities suspicious for hematologic disease.

3. Recognize common platelet disorders that result in thrombocytopenia or thrombocytosis.
Blood Basics

- Adult blood:
  - Blood volume: 5-6 L
  - 7% body weight
  - Circulates the entire body in 20-60 seconds

- Childhood blood:
  - Total blood volume: body weight
  - Neonate: 85 ml/kg
  - 1 month: 105 ml/kg
  - >2 months: 70-80 ml/kg

Components of Blood:
- Formed elements: blood cells
  - Erythrocytes
  - Leukocytes
  - Platelets
- Plasma
  - 90% water
  - 10% solutes

Serum = plasma without clotting factors
Blood Basics

• Hematopoiesis:
  • Continuous production of blood cell population.

• Bone marrow cavities and canals.

• Mediated:
  • Growth factors
  • Hematopoietic stem cells
# Blood Basics

<table>
<thead>
<tr>
<th>Cell Type</th>
<th>Illustration</th>
<th>Description*</th>
<th>Number of Cell per mm² (µl) of Blood</th>
<th>Duration of Development (D) and Life Span (LS)</th>
<th>Function</th>
</tr>
</thead>
<tbody>
<tr>
<td>ERYTHROCYTES</td>
<td></td>
<td>Biconcave, anucleate disc; salmon-colored; diameter 7–8 µm</td>
<td>4–6 million</td>
<td>D: 5–9 days LS: 100–120 days</td>
<td>Transport oxygen and carbon dioxide</td>
</tr>
<tr>
<td>(red blood cells; RBCs)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LEUKOCYTES</td>
<td></td>
<td>Spherical, nucleated cells</td>
<td>4800–11,000</td>
<td>D: 7–11 days LS: 6 hours to a few days</td>
<td>Destroy bacteria by phagocytosis</td>
</tr>
<tr>
<td>(white blood cells; WBCs)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Granulocytes</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Neutrophils</td>
<td></td>
<td>Nucleus multilobed; inconspicuous cytoplasmic granules; diameter 12–14 µm</td>
<td>3000–7000</td>
<td>D: 7–11 days LS: 6 hours to a few days</td>
<td>Destroy bacteria by phagocytosis</td>
</tr>
<tr>
<td>• Eosinophils</td>
<td></td>
<td>Nucleus bilobed; red cytoplasmic granules; diameter 12–15 µm</td>
<td>100–400</td>
<td>D: 7–11 days LS: about 5 days</td>
<td>Turn off allergic responses and kill parasites</td>
</tr>
<tr>
<td>• Basophils</td>
<td></td>
<td>Nucleus bilobed; large blue-purple cytoplasmic granules; diameter 10–14 µm</td>
<td>20–50</td>
<td>D: 3–7 days LS: a few hours to a few days</td>
<td>Release histamine and other mediators of inflammation</td>
</tr>
<tr>
<td>Agranulocytes</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Lymphocytes</td>
<td></td>
<td>Nucleus spherical or indented; pale blue cytoplasm; diameter 5–17 µm</td>
<td>1500–3000</td>
<td>D: days to weeks LS: hours to years</td>
<td>Mount immune response by direct cell attack (T cells) or via antibodies (B cells)</td>
</tr>
<tr>
<td>• Monocytes</td>
<td></td>
<td>Nucleus U- or kidney-shaped; gray-blue cytoplasm; diameter 14–24 µm</td>
<td>100–700</td>
<td>D: 2–3 days LS: months</td>
<td>Phagocytosis; develop into macrophages in tissues</td>
</tr>
<tr>
<td>PLATELETS</td>
<td></td>
<td>Discoid cytoplasmic fragments containing granules; stain deep purple; diameter 2–4 µm</td>
<td>150,000–500,000</td>
<td>D: 4–5 days LS: 5–10 days</td>
<td>Seal small tears in blood vessels; instrumental in blood clotting</td>
</tr>
</tbody>
</table>
Blood Basics: The CBC

- Red Blood Cell Indices:
  - MCV
  - RDW
  - MCH
  - MCHC

- Platelet Indices
  - MPV

WBC, White blood cell.
Blood Basics: The CBC

Manual

Automated
Blood Basics: The Red Blood Cell

• Biconcave disc
  • 7.8 μm diameter
  • Highly flexible membrane
  • 100-120 day lifespan
  • 3.9-6 million cells/μl
  • Produce 2.4 million RBCs/second

• Simple interior:
  • Lack of nucleus
  • Lack of organelles
  • Enzymes for glycolysis
  • Hemoglobin
Blood Basics: Hemoglobin

- **Structure**
  - **Heme Group:**
    - Protoporphyrin IX
    - Single atom of Iron
  - **Globin:**
    - Polypeptide chain

- Heme + Globin = Hemoglobin Chain:
  - 16,000 g/mol
  - Variety of different chains

- **Hemoglobin Protein:**
  - 4 loosely bound hemoglobin chains
Blood Basics: Hemoglobin

**Hemoglobin**

<table>
<thead>
<tr>
<th>Component</th>
<th>Name</th>
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<tbody>
<tr>
<td>Normal</td>
<td>Missense Mutation</td>
</tr>
<tr>
<td>Partial DNA Sequence of Beta Globin Gene:</td>
<td>CCT GAG GAG</td>
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<tr>
<td>Partial RNA Sequence:</td>
<td>GGA CTC CTC</td>
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<tr>
<td>Partial Amino Acid Sequence for Beta Globin:</td>
<td>CCU GAG GAG</td>
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<tr>
<td>Hemoglobin Molecule:</td>
<td>Pro Glu Glu</td>
</tr>
<tr>
<td>Red Blood Cell:</td>
<td>Pro Val Glu</td>
</tr>
<tr>
<td>Bart's</td>
<td>γ4</td>
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</tbody>
</table>

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# Blood Basics: The Newborn Screen

<table>
<thead>
<tr>
<th>NB Screen Result</th>
<th>Description</th>
<th>Genotype</th>
<th>Start Penicillin</th>
<th>Testing and Referral</th>
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<tbody>
<tr>
<td>FA</td>
<td>Normal</td>
<td>AA</td>
<td>No</td>
<td>None</td>
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<tr>
<td>FS</td>
<td>Sickle Cell Anemia OR Sickle-β Thalassemia OR Sickle with Hereditary Persistence of Fetal Hemoglobin</td>
<td>SS, S-β Thalassemia S-HPFH</td>
<td>Yes</td>
<td>Hematology referral &amp; Family testing and counseling</td>
</tr>
<tr>
<td>FSC</td>
<td>Sickle-C Disease</td>
<td>SC</td>
<td>Yes</td>
<td>Hematology referral &amp; Family testing and counseling</td>
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<tr>
<td>FSA</td>
<td>Sickle β Thalassemia</td>
<td>SA</td>
<td>Yes</td>
<td>Hematology referral &amp; Family testing and counseling</td>
</tr>
<tr>
<td>FSV</td>
<td>Sickle with Hemoglobin Variant</td>
<td>SV</td>
<td>Yes</td>
<td>Hematology referral &amp; Family testing and counseling</td>
</tr>
<tr>
<td>FSE/O or FBD/G</td>
<td>Sickle with indeterminate hemoglobin pattern; both indeterminate patterns indicate increased risk for sickling disorder</td>
<td>Multiple possibilities</td>
<td>Yes</td>
<td>Hematology referral &amp; Family testing and counseling</td>
</tr>
<tr>
<td>FC</td>
<td>Hemoglobin C Disease OR Hemoglobin C-B Thalassemia</td>
<td>CC, C-B Thalassemia</td>
<td>No</td>
<td>Hematology referral &amp; Family testing and counseling</td>
</tr>
<tr>
<td>FE/O</td>
<td>Multiple possibilities of E, O and B Thalassemia</td>
<td>Multiple possibilities</td>
<td>No</td>
<td>Family testing and counseling</td>
</tr>
<tr>
<td>F</td>
<td>Hereditary Persistence of Fetal Hemoglobin OR B Thalassemia Major (age dependent) Premature Infant</td>
<td>Multiple possibilities</td>
<td>No</td>
<td>Family testing and counseling</td>
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<tr>
<td>FAS</td>
<td>Sickle Cell Trait (carrier)</td>
<td>AS</td>
<td>No</td>
<td>Family testing and counseling</td>
</tr>
<tr>
<td>FAC</td>
<td>Hemoglobin C Trait (carrier)</td>
<td>AC</td>
<td>No</td>
<td>Family testing and counseling</td>
</tr>
<tr>
<td>FAV</td>
<td>Carrier of Hemoglobin Variant including E, O, G or G</td>
<td>AV A with either E, O, D or G trait</td>
<td>No</td>
<td>Confirmatory testing &amp; Contact hematology if needed</td>
</tr>
<tr>
<td>FAD/G</td>
<td>Presence of Hemoglobin Bart’s 10</td>
<td>AA</td>
<td>No</td>
<td>See Hb Bart’s flow sheet</td>
</tr>
<tr>
<td><strong>T</strong></td>
<td>Pattern suggests transfusion</td>
<td>Multiple possibilities</td>
<td>No</td>
<td>If not transfused repeat test &amp; If transfused repeat test 2 months after last transfusion</td>
</tr>
</tbody>
</table>

---

**Notes:**

1. AA: Normal
2. SS: Sickle Cell Anemia
3. S-β: Beta Thalassemia
4. S-HPFH: Sickle-Hemoglobin Persistence of Fetal Hemoglobin
5. Hematology referral & Family testing and counseling
6. Multiple possibilities of E, O and B Thalassemia
7. Contact hematology with results of family testing
8. Family testing and counseling
9. Family testing and counseling
10. AA: Normal
11. AV: A with either E, O, D or G trait
12. AA: Normal
13. If not transfused repeat test & If transfused repeat test 2 months after last transfusion

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## Pediatric Anemia

**Defining Anemia:**
- Reduction in RBC mass.
- Reduction in Hgb concentration.

**Varies substantially:**
- Age
- Race
- Gender

**Anemia:**
- Hct or Hgb below the 25th percentile

### Differential diagnosis: broad/variable

**Systematic approach to diagnosis**

---

### Normal values for hemoglobin, hematocrit, and mean corpuscular volume in children

<table>
<thead>
<tr>
<th>Age</th>
<th>Hemoglobin (g/dL)</th>
<th>Hematocrit (%)</th>
<th>MCV (fl)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>50th percentile</td>
<td>Lower limit*</td>
<td>50th percentile</td>
</tr>
<tr>
<td>1 year*</td>
<td>12.5</td>
<td>11</td>
<td>37</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>11</td>
<td>36</td>
</tr>
<tr>
<td>Caucasian</td>
<td>12.6</td>
<td>11</td>
<td>37</td>
</tr>
<tr>
<td>African American</td>
<td>12</td>
<td>11</td>
<td>36</td>
</tr>
<tr>
<td>2 to 3 years</td>
<td>12.9</td>
<td>11.7</td>
<td>38</td>
</tr>
<tr>
<td>Caucasian</td>
<td>12.3</td>
<td>11</td>
<td>37</td>
</tr>
<tr>
<td>African American</td>
<td>13.5</td>
<td>12</td>
<td>40</td>
</tr>
<tr>
<td>4 to 6 years</td>
<td>13.7</td>
<td>12</td>
<td>38</td>
</tr>
<tr>
<td>Caucasian</td>
<td>12.7</td>
<td>11.2</td>
<td>38</td>
</tr>
<tr>
<td>African American</td>
<td>13.5</td>
<td>12</td>
<td>40</td>
</tr>
<tr>
<td>7 to 10 years</td>
<td>13.6</td>
<td>11.3</td>
<td>38</td>
</tr>
<tr>
<td>Caucasian</td>
<td>12.9</td>
<td>10.6</td>
<td>38</td>
</tr>
<tr>
<td>African American</td>
<td>13.5</td>
<td>11.8</td>
<td>40</td>
</tr>
<tr>
<td>11 to 14 years</td>
<td>13.7</td>
<td>12.3</td>
<td>40</td>
</tr>
<tr>
<td>Caucasian</td>
<td>14.3</td>
<td>12.6</td>
<td>42</td>
</tr>
<tr>
<td>African American</td>
<td>12.9</td>
<td>10.6</td>
<td>38</td>
</tr>
<tr>
<td>Male</td>
<td>13.6</td>
<td>11.8</td>
<td>40</td>
</tr>
<tr>
<td>15 to 18 years</td>
<td>13.7</td>
<td>11.5</td>
<td>40</td>
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<tr>
<td>Caucasian</td>
<td>15.4</td>
<td>13.7</td>
<td>46</td>
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<tr>
<td>African American</td>
<td>12.8</td>
<td>10.7</td>
<td>38</td>
</tr>
<tr>
<td>Male</td>
<td>14.9</td>
<td>12.9</td>
<td>44</td>
</tr>
</tbody>
</table>
Classifying Pediatric Anemia: Size (MCV)

- **Microcytic (<70)**
  - Iron deficiency
  - Lead intoxication
  - Thalassemia
  - Sideroblastic anemia
  - Anemia of inflammation
  - Zinc deficiency

- **Normocytic (70-80)**
  - Infection
  - Acute blood loss
  - Anemia of inflammation
  - Drug-induced
  - Renal disease
  - TEC
  - Hemoglobinopathy
  - Hemolytic anemia:
    - Autoimmune
    - Membrane defect
    - Enzyme defect
    - MAHA

- **Macrocytic (>95)**
  - Vitamin B12/folate deficiency
  - Hypothyroidism
  - Drug-induced
  - Post-splenectomy
  - Diamond-Blackfan
  - Bone marrow failure
  - Bone marrow infiltration
  - Liver disease
  - Reticulocytosis
    - Sickle Cell Disease
Classifying Pediatric Anemia: Cause (Retic)

**Decreased Production**
- Iron deficiency
- Thalassemia
- Lead intoxication
- B12/folate/zinc
- Infection
- Drug-induced
- Anemia of Inflam.
- Bone marrow disease
- Hypothyroidism
- Renal disease
- Sideroblastic anemia
- DBA and TEC

**Increased Destruction**
- Hemoglobinopathy
- Hemolytic anemia:
  - Autoimmune
  - Membrane defect
  - Enzyme defect
  - MAHA

**RBC Loss**
- Hemorrhage
- Acute blood loss
- Chronic blood loss
- Liver disease
  - Blood loss
  - Hypersplenism
Classifying Pediatric Anemia: Age

• **Birth to 3 months**
  - Blood loss
  - Alloimmune hemolysis
    - Rh disease
    - ABO incompatibility
  - Congenital infection
  - Intrinsic hemolytic anemia
  - Twin-to-twin transfusion

• **3 to 6 months**
  - Hemoglobinopathy
  - Infection

• **9 months to teens**
  - Nutritional deficiency
  - All others....
Newborn Anemia

• Physiologic Anemia of Infancy
  • ↑ tissue oxygenation
  • ↓ erythropoietin

• Pathologic anemia in newborns:
  • Hgb <13.5: birth-4 weeks
  • Hgb <9: weeks 6-9
  • Signs of hemolysis
  • Signs of anemia
    • Irritability
    • Poor feeding
Approaching Anemia: Clinical

- **Age**
- **Sex**
  - X-linked diseases: G6PD deficiency
  - Postmenarchal female
- **Race/Ethnicity**
  - Thalassemia:
    - Mediterranean
    - Southeast Asian
  - Hemoglobin S and C
    - African descent
    - Hispanic populations
- **Symptoms:**
  - Severity
  - Acute: lethargy, tachycardia, pallor
  - Chronic: none or minimal sx’s
- **Hemolysis:**
  - jaundice
  - change in urine color
  - Scleral icterus
- **Bleeding:**
  - GI
  - Chronic epistaxis
  - menstrual
- **Dietary history:**
  - Formula or breastfed
  - Cow versus goat milk
    - Age of onset
    - Daily volume
  - Pica
  - Iron-rich food intake
- **PMHx**
  - Newborn jaundice
  - Newborn screen
  - Prior CBC’s
  - Underlying medical conditions
  - Drug/toxin exposure
- **Family History**
  - CCY or splenectomy
  - Gallstones
Approaching Anemia: Exam

• Assessing pallor

• Assessing hemolysis

• Clues
Approaching Anemia

Clinical Assessment Hemoglobin

MCV
- Macrocytic >85
- Normocytic 70-80
- Microcytic <70

Reticulocytes
- Reduced Production
- Increased Destruction
- RBC Loss

Absolute Reticulocyte Count
ARC = % Retic x RBC

ARC < 100 x10⁹
INADEQUATE RESPONSE

ARC > 100 x10⁹
APPROPRIATE RESPONSE
Microcytic Anemia

- Iron Deficiency Anemia
- Thalassemia
- Less Common
- Lead Intoxication
- Anemia of Inflammation (<20%)
- Congenital Sideroblastic Anemia
- Zinc Deficiency

IDA versus Thalassemia
- Red Cell Distribution Width (RDW)
  - Low RDW: Thal
  - High RDW: IDA

- RBC Count
  - >5 million: Thal
  - <5 million: IDA

- Mentzer Index = MCV/RBC
  - <11: Thal minor
  - >13: IDA

MCV <70
Reticulocyte <3%
ARC <100,000
Iron Deficiency Anemia

• Causes: Nutritional deficiency
  • Infants
    • Exclusive BF without iron supplementation
    • Formula with insufficient iron
    • Early transition to cows milk
  • Toddlers
    • Excessive cows milk intake
      • >24 ounces/day
  • Adolescents
    • Alternative diets: vegetarians, vegans
    • Endurance athletics
    • Obesity

• Other Causes:
  • Blood loss:
    • Menorrhagia
    • Chronic epistaxis
    • Occult GI bleeding (IBD)
  • Reduced iron absorption:
    • Celiac disease
    • Autoimmune gastritis
    • H. pylori gastritis
  • Rare genetic conditions:
    • IRIDA
    • SLC11A2 mutation

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Iron Metabolism

1. Duodenum Absorbed iron: 1-2 mg/day
2. Hepatocytes Hepatic iron stores, other tissues: ~ 500 mg
3. Ferroportin

- Erythrocytes: Recycled iron: 20-25 mg/day
- Bone marrow
- Ferroportin
- Plasma Transferrin 3^+ 3^+
- Plasma Iron: ~ 3 mg
- Hb-Iron: ~ 2500 mg
Iron Deficiency Anemia

- **Diagnosis:**
  - Age <3 yo, typical presentation:
    - CBC sufficient.
  - Age >3 yo or atypical presentation
    - CBC
    - Iron Studies

- **Iron Studies**
  - Serum iron: 1% measured iron
  - Ferritin: 99% measured iron
  - TIBC: transferrin.
  - Iron Saturation = serum iron/TIBC

```
<table>
<thead>
<tr>
<th></th>
<th>Normal</th>
<th>Negative iron balance</th>
<th>Poor erythropoiesis</th>
<th>Iron deficiency anaemia</th>
</tr>
</thead>
<tbody>
<tr>
<td>Iron stores</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Circulating iron</td>
<td></td>
<td></td>
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<tr>
<td>Stored in bone marrow</td>
<td>1-3+</td>
<td>0-1+</td>
<td>0</td>
<td>0</td>
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<tr>
<td>Serum ferritin mcg/dl</td>
<td>50-200</td>
<td>&lt;20</td>
<td>&lt;15</td>
<td>&lt;15</td>
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<tr>
<td>TIBC mcg/dl</td>
<td>300-360</td>
<td>&gt;360</td>
<td>&gt;380</td>
<td>&gt;400</td>
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<tr>
<td>Serum iron g/dl</td>
<td>50-150</td>
<td>NL</td>
<td>&lt;50</td>
<td>&lt;30</td>
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<tr>
<td>Saturation (%)</td>
<td>30-50</td>
<td>NL</td>
<td>&lt;20</td>
<td>&lt;10</td>
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<td>Protoporphyrin mcg/dl</td>
<td>30-50</td>
<td>NL</td>
<td>&gt;100</td>
<td>&gt;200</td>
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<td>Morphology</td>
<td>NL</td>
<td>NL</td>
<td>NL</td>
<td>Microcytic/hypochromic</td>
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</table>
```
Iron Deficiency Anemia

- Clinical Manifestations:
  - Neurocognitive
  - Exercise capacity
  - Febrile seizures
  - Pica
  - Cerebral vein thrombosis
  - Restless leg syndrome
  - Infection and immunity

- Nonanemic iron deficiency:
  - Easy fatigue in athletes
  - Cognitive function in adolescents

- Treatment
  - Oral supplementation
    - 3-6 mg/kg elemental iron
    - Once daily
    - Between meals without dairy
    - With water or juice (vitamin C)
  - Dietary changes
    - Limit cows milk: 6-20 oz/day
    - Discontinue the bottle!

- Treating Teens:
  - 65-130 mg once daily (1-2 tabs 1xD)
  - Combine with ascorbic acid
Hemolytic Anemia

**Intrinsic:**
- Membrane Defects
  - Hereditary Spherocytosis
  - Hereditary Elliptocytosis
- Enzyme Defects
  - G6PD deficiency
  - Pyruvate Kinase deficiency
- Hemoglobinopathies
  - Sickle cell disease
  - Hemoglobin E
  - Hemoglobin C
  - Thalassemia

**Extrinsic:**
- Autoimmune
  - Warm reactive
  - Cold agglutinin
  - Paroxysmal Cold Hemoglobinuria
  - SLE
  - Evan's syndrome
- Alloimmune
  - Rh disease of NB
  - ABO incompatibility
- Microangiopathic
  - HUS
  - TTP
  - DIC
- Mechanical
  - Congenital heart disease
  - Artificial heart valve

**Lab Studies:**
- Reticulocytosis
- Hyperbilirubinemia
- ↑ LDH
- ↓ Haptoglobin
- Plasma-free Hgb
- Urine: Hemoglobinuria, Bilirubinuria
- Peripheral smear: MCV 70-80, Reticulocyte >3%, ARC >100,000

**Common RBC Inclusions:**
- Howell Jolly Bodies (DNA) - Hyposplenism, Asplenism, Severe hemolytic anemia
- Heinz Bodies (Supravital stain) - G6PD deficiency, Oxidant drugs, Unstable hemoglobin
- Pappenheimer Bodies (Iron deposits) - Thalassemia, Sideroblastic anemia, Hemolytic anemia, Post-splenectomy
- Hemoglobin H Inclusion (Supravital stain) - Hemoglobin
- Basophilic Stippling (Ribosomes) - Lead poisoning, Thalassemia, Sickle cell anemia, MDS

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Pure Red Blood Cell Aplasia

Diamond Blackfan Anemia (DBA)
- Congenital
  - ribosomal protein mutations
- Presents <1 yo (90%)
  - No prior illness
  - Hgb 2-6
  - Retic <1%
  - MCV: macrocytic
  - Elevated Hgb F
- Congenital abnormalities (30-50%)
- Life-long condition
  - Transfusion support
  - Steroid therapy
  - Risk of malignancy

Transient Erythroblastopenia of Childhood (TEC)
- Acquired
  - Unknown etiology
- Presents 1-4 yo (80%)
  - Preceding viral illness
  - Hgb 3-9
  - Retic <1%
  - MCV normal
  - Normal Hgb F
- No anatomic abnormalities
- Spontaneous resolution
  - Weeks to months
  - Rare need for transfusion
White Blood Cell Abnormalities

• Leukocytosis
  • Lymphocytosis: ALC >4000

• Leukopenia
  • Lymphocytopenia: ALC <1500

• Abnormal WBC forms in periphery

Absolute Neutrophil Count (ANC)
WBC x %Neutrophils/100
Neutropenia

• Definitions:
  • Mild  ANC 1000-1500
  • Moderate  ANC 500-1000
  • Severe  ANC <500

• Infancy:
  • 0-7 days  ANC <5000
  • 14 days to 1 yr  ANC <1000

• Prevalence of neutropenia in U.S.
  • African-American: 4.5 to 10.5%
  • Haiti: 8.5%
  • Barbados/Trinidad: 6.4%
  • Jamaica: 2.7%
  • Mexican: 0.38%
  • Caucasian: 0 to 0.79%
Benign Familial Neutropenia

  - West Indians, Arab Jordanians, Yemenite Jews

- Traced to common West African allele:
  - SNP in \textit{DARC} gene
  - Receptor for inflammatory cytokines

- Normal bone marrow reserve of granulocytes
  - Defective granulocyte release from the marrow

- Neutropenia is benign
  - No propensity to infection
Neutropenia: Causes

• Acquired
  • Postinfectious
  • Drug-induced
  • Nutritional
    • Vit B12/folate deficiency
    • Copper deficiency
  • Immune
    • Alloimmune neonatal
    • Chronic autoimmune
    • Collagen vascular disease
    • Immunodeficiency
  • Hypersplenism
  • Bone marrow disorders
    • Leukemia
    • Aplastic Anemia
    • Chemotherapy
  • Chronic Idiopathic

• Congenital
  • Kostman Syndrome (SCN)
  • Shwachman-Diamond Syndrome
  • WHIM Syndrome
  • GATA2 Deficiency
  • Chediak-Higashi Syndrome
  • Glycogen Storage Disease Type 1b
  • GCSF receptor mutation

• Cyclic Neutropenia
Neutropenia: Approach

• Does neutropenia indicate a serious underlying disease?

• Is the patient at increased risk of infection because of neutropenia?
Neutropenia: Approach

Clinical Concern
- Recurrent oral ulcers and gingivitis
- Perirectal ulcers
- Recurrent Staph and Strep
  - Oropharyngeal and Otitis
  - Respiratory
  - Cellulitis
  - Bacteremia
- Unusual organisms
- Chronic diarrhea/FTT
- Chronic inflammation: ↑ ESR
- Recurrent fevers every 21 days

Incidental/Reassuring History
- No concerning infectious history
- Associated viral syndrome
- No oral/gingival issues
- Confirmed with repeat CBC:
  - Pseudoneutropenia: cell clumping
    - Sample left standing
    - Presence of anticoagulant

Urgent Hematology Referral:
- bone marrow biopsy
- genetic sequencing

Fever:
- Immediate ER evaluation
- Hematology consultation
- Inpatient: antimicrobials

Serial PE with CBC and ESR
- every 1-2 weeks
- gradually decreasing interval

Fever:
- CBC and blood culture
- ER eval if clinical concern
Neutropenia: Clinical Scenarios

Healthy Infant/Toddler
- Post-infectious
  - Transient, mild-moderate ANC
  - Viral etiology
    - RSV, Influenza, Parvo, EBV, HHV6
  - Onset: within 72hrs of illness start
  - Resolves: after 3-8 days
- Benign Neutropenia of Childhood
  - Chronic autoimmune neutropenia
  - Prolonged, moderate-severe ANC
  - Not associated with severe infections
  - Age 5-15 months
  - Resolves: after months-years

Older Child/Teenager
- Absent protracted/recurrent infection
  - ANC >800
  - Absent oral symptoms
  - Typically benign etiology
    - Followed with serial CBC
    - Lab eval with any febrile illnesses
- Recurrent infection
  - Diagnostic evaluation
    - ANA, complement
    - Anti-neutrophil antibodies
    - Ig levels and vaccine titers
    - HIV
    - Nutritional studies
Lymphocytosis

• Definitions:
  • Age >12 yo
  • Age <12 yo
  • ALC >4000
  • ALC >8000

• Lymphocyte subsets
  • T cells (CD3+)
  • B cells (CD20+)
  • NK cells (CD56+)
  • 60-80%
  • 10-20%
  • 5-10%

• Reactive versus Clonal
  • Clonal Lymphocytosis: Rare
    • Acute Leukemia
    • Hereditary Polyclonal B cell Lymphocytosis
    • CLL
    • Monoclonal B cell Lymphocytosis
    • Lymphoproliferative disease of LGL
  • Reactive
Reactive Lymphocytosis

- Mononucleosis
  - EBV, CMV
  - HHV6, Adenovirus
- Infectious Lymphocytosis
  - Coxsackie, poliovirus, enterovirus
  - WBC 20-100: 60-90% T-cells
  - Last 4-10 weeks
- Other viruses
  - Mumps, varicella, influenza, hepatitis, rubella, measles
- Pertussis
- Cat Scratch
- Toxoplasmosis
- Babesiosis
- Hypersensitivity reactions
  - Drug-induced
  - Serum sickness
- Stress-induced
  - Cardiac emergencies
  - Status epilepticus
  - Trauma
- Post-splenectomy
Lymphocytosis: Evaluation

- CBC
- Peripheral smear
  - Lymphocyte morphology
- EBV titers

- Ruling Out Clonality
  - Lymphocyte subsets
  - Clonal Ig rearrangements
  - Clonal TCR rearrangements
  - Kappa/lambda light chain expression

- Ruling Out Malignancy
  - Morphology
  - Flow cytometry
  - Bone marrow biopsy
    - Associated anemia, neutropenia, thrombocytopenia
Eosinophilia

• Definitions:
  • Eosinophilia: AEC >500
  • Hypereosinophilia: AEC >1500
  • Hypereosinophilia Syndrome
    • AEC >1500
    • End-organ damage (cardiac, neuro, pulm)

• Differential Diagnosis:
  • Neoplastic Eosinophilia
    • Leukemia/lymphoma with eosinophilia
    • Primary hypereosinophilia syndrome
  • Secondary Eosinophilias
Eosinophilia

• Childhood:
  • Neoplastic hypereosinophilia is VERY rare

• Secondary causes of eosinophilia are COMMON:
  • Asthma, atopic disease: mild-moderate
  • Food allergy, eosinophilic esophagitis
  • Infection: toxocariasis, filariasis
  • Medications
  • Primary immunodeficiencies:
    • ALPS, HyperIgE

• Severity of AEC
  • Does not predict etiology
  • Does not predict risk of end-organ damage

• Diagnostic evaluation HES is necessary:
  • Persistent AEC >1500
  • AEC 500-1500 with concern for organ dysfunction
Leukocytosis

• Generally driven by neutrophilia.

• “Left-Shift”
  • Increase in band forms
  • Metamyelocytes, myelocytes
  • Acute bacterial infection
    • WBC >25,000
    • Toxic granulations, Dohle bodies

• DDx broad:
  • Infection
  • Inflammation
  • Medications
  • Asplenia, cigarette exposure, stress, genetic

• Leukocytosis: malignancy

• Acute leukemia:
  • Circulating leukemic blasts driving increase in WBC’s

• Chronic leukemia: CML
  • Exceptionally rare in children
  • Distinct presentation:
    • Maturing granulocytes in periphery
    • Polycythemia
    • Thrombocytosis
    • Eosinophilia
    • Basophilia
    • Splenomegaly
Platelets

• Normal range: 150-450,000
• Lifespan: 8-10 days
• Function: primary hemostasis
Thrombocytopenia

• Definition:
  • General <150,000
  • ITP <100,000

• Symptoms:
  • Cutaneous:
    • Superficial ecchymoses
    • Petechiae
  • Mucosal
    • Epistaxis
    • Gingival bleeding
    • Wet purpura
    • GI/GU bleeding
  • Intracranial Hemorrhage

• Bleeding Risk
  • Inversely proportional to platelet count
  • Risk begins: Plts <100

• Surgical bleeding Plts <50
• Spontaneous bleeding Plts <20

• Younger platelets: ↑ hemostatic
  • More bleeding risk: disorders of platelet production
  • Less bleeding risk: disorders of platelet destruction
Thrombocytopenia: Causes

**Increased Platelet Destruction**
- Immune-mediated
  - Immune Thrombocytopenia (ITP)
  - Drug-induced
- Activation/Consumption
  - Microangiopathic HA
    - TTP, HUS, DIC
  - Major surgery, trauma
  - Kasabach-Merritt
- Mechanical Destruction
  - ECMO, bypass, dialysis, apheresis
- Sequestration/Trapping
  - Hypersplenism
  - Von Willebrand Disease: Type 2B, pseudo-vWF

**Decreased Platelet Production**
- Infection
  - EBV, CMV
  - Parvo, varicella, rickettsia
  - HIV
  - Bacterial sepsis
- Nutritional deficiency
  - B12/folate
  - Iron
- Bone marrow disease
  - Aplastic anemia
  - Malignancy
- Genetic
Congenital Thrombocytopenia: MPV

- **Small Plts (<7)**
  - Wiskott-Aldrich
  - X-linked Thrombocytopenia

- **Normal (7-11)**
  - Bone Marrow Failure
    - Fanconi Anemia
    - Dyskeratosis congenita
    - Shwachman-Diamond
    - Congenital Amegakaryocytic Thrombocytopenia (CAMT)
  - Thrombocytopenia-absent radius syndrome (TAR)
  - Amegakaryocytic thrombocytopenia with radioulnar synostosis

- **Giant Plts (>11)**
  - Bernard-Soulier Syndrome
  - MYH9-related disorders
  - Paris-Trousseau
  - Gray platelet
Spurious Thrombocytopenia

• Platelet clumping
  • Automated CBC: counted as leukocyte
  • False reading: thrombocytopenia

• Causes:
  • Improper blood collection
  • Delayed processing
  • Inadequate anticoagulation
  • Pseudothrombocytopenia
    • 0.1% population
    • EDTA-dependent antibodies

Verify Thrombocytopenia: Repeat CBC
Immune Thrombocytopenia (ITP)

- Most common cause of symptomatic thrombocytopenia
  - 1-6 cases/100,000 children
  - Peak incidence: 2-5 years

- Sudden onset of severe thrombocytopenia:
  - Defined: Plts <100,000
  - 80%: Plts <20,000
  - 45%: Plts <10,000

- Triggers:
  - Preceding viral illness: 60% cases
    - Within 4 weeks of onset
  - MMR vaccination

Figure 1. Proposed mechanism of immune dysregulation in ITP.
Immune Thrombocytopenia (ITP)

• Symptoms:
  • Mucocutaneous bleeding
  • Lack of systemic symptoms
  • Rare: serious hemorrhage (3%)
    • Prolonged epistaxis
    • Intracranial hemorrhage (0.5%)
    • GI/GU hemorrhage

• Diagnosis: Clinical/Exclusion
  • Typical presentation:
    • Age 1-10
    • acute onset, otherwise healthy
  • Plts <100,000
  • CBC otherwise normal
  • Peripheral smear: no hemolysis or blasts
  • Negative DAT (Coomb’s)
  • Response to treatment

• Indications for bone marrow biopsy:
  • Systemic symptoms, LAD, HSM
  • Atypical labs: cytopenias
  • Lack of response to treatment
  • Chronic ITP >12 months
Approach to Mild-Moderate Thrombocytopenia

- Repeat CBC:
  - Spurious thrombocytopenia

- Viral suppression: most common
  - Associated viral symptoms
  - Mild-moderation thrombocytopenia
  - Monitor serial CBC’s over 2-4 weeks

- Presumptive ITP
  - Otherwise healthy
  - Preceding illness
  - Monitor for spontaneous resolution

- Persistent thrombocytopenia:
  - Lasting >2-3 months
  - No clear etiology
  - Diagnostic dilemma: Hematology referral

- Diagnostic evaluation:
  - Anti-Platelet Antibodies and DAT
    - Chronic ITP
    - Evan’s syndrome
  - Rheumatology evaluation
    - Collagen vascular disorders
    - SLE
  - Bone marrow biopsy:
    - Malignancy/MDS
    - Bone marrow failure syndrome
  - Genetic sequencing
    - MYH9 disorders
    - Congenital thrombocytopenia
Thrombocytosis

**Reactive/Secondary**
- Stimulated megakaryopoiesis
- >600/million children
- Transient
- Plts <800
- MPV large
- Normal morphology
- ↑ CRP/ESR, vWF Ag, fibrinogen
- No bleeding/clotting
- No splenomegaly

**Essential/Primary**
- Myeloproliferative disease
- 1/million children
- Chronic
- Plts >1000
- MPV small or large
- Abnormal morphology
- ↑ PT/PTT, Anti-Phosphlipid Ab’s
- Associated bleeding/clotting
- Splenomegaly

**Mild** 450-700
**Moderate** 700-900
**Severe** 900-1000
**Extreme** >1000
Take Home Points

• Systematic approach to pediatric anemia: directs diagnosis.
  • Age
  • Size of RBC’s (MCV)
  • Bone marrow response (reticulocytes)

• Urgency of evaluating neutropenia depends on degree of clinical concern:
  • Infectious history
  • Oral health: gingivitis, ulcers

• Isolated thrombocytopenia in childhood is almost always benign in etiology:
  • Viral suppression
  • Immune thrombocytopenia
Question

• A 4 yo boy presents to the office as a sick visit for lethargy and dark urine. On exam he appears quite pale and has scleral icterus. He was seen at an urgent 3 days prior and started on an antibiotic for bronchitis. CBC reveals a Hgb of 7. Mom informs you that similar episodes happened to her grandfather. What are the most likely lab abnormalities.
  
a) MCV 61, retic 1.5%, LDH 150, serum iron 13, ferritin 2.
  b) MCV 80, retic 1%, LDH 275, ferritin 45, EBV titers positive.
  c) MCV 85, retic 0.1%, LDH 100, total bili 0.7, Hgb F <2%.
  d) MCV 89, retic 10%, LDH 1200, total bili 4.5, haptoglobin <10.
  e) MCV 107, retic 0.8%, LDH 3000, total bili 2.4, immature lymphocytes 28%.
A 9 month old full term girl presents to the office as a sick visit for fever to 103. Her PMHx is significant for 2 prior episodes of AOM, inpatient hospitalization for RSV with superimposed bacterial pneumonia, and recent cellulitis treated with an outpatient course of antibiotics. Her exam reveals lethargy and gingivitis. What is the appropriate course of action:

a) Trend CBC’s and ESR’s every 1-2 weeks.
b) Restart oral antibiotics as the cellulitis has likely recurred.
c) Refer to the ER for immediate evaluation.
d) Send to the lab for a CBC, blood culture, and CRP.
e) Administer IM ceftriaxone in the office and ask the parents to monitor her fever curve closely at home.
Question

• A 2 yo girl with no significant past medical history presents to the office with new onset diffuse bruising. She had a recent cold 3 weeks ago, but is otherwise well. Her exam reveals scattered petechiae, palatal petechiae, and mucosal purpura. CBC is obtained demonstrating a platelet count of 25. The other cell lines are normal. What is the most appropriate next step:
  a) Contact your local on-call hematologist to arrange for further evaluation later today.
  b) Repeat the CBC the following today to evaluate for spurious thrombocytopenia.
  c) Contact the state’s child services department given concern for possible nonaccidental trauma.
  d) Call EMS to transport the patient to the hospital emergently given concerns for acute leukemia.
  e) Refer to genetics for evaluation of congenital thrombocytopenia.