HEMATOLOGY/HEMATOPOIESIS

Introduction
HEMATOLOGY

Introduction

- Study of blood & its components
- Window of rest of body
BLOOD
Raison d'etre

• Delivery of nutrients
  – Oxygen
  – Food
  – Vitamins

• Removal of wastes
  – Carbon dioxide
  – Nitrogenous wastes
  – Cellular toxins

• Repair of its conduit

• Protection versus invading microorganisms

• Multiple cellular & acellular elements
HEMATOLOGY

Divisions

- Red Blood Cells/Oxygen & CO$_2$ transport
- White Blood Cells/Protection *versus* microorganisms
- Coagulation/platelets/Maintenance of vascular integrity
HEMATOLOGY

Hematopoiesis

- In humans, occurs in bone marrow exclusively
- All cellular elements derived from pluripotent stem cell (PPSC)
- PPSC retains ability to both replicate itself and differentiate
- Types of differentiation determined by the influence of various cytokines
HEMATOPOIESIS

Pluripotent Stem Cell

- Committed Stem Cells
  - Differentiating Mitotic Component
    - Erythropoietin
    - Colony Stimulating Factors (GM-CSF, IL-3, G-CSF)
  - Myeloid Lineage
    - Granulocyte
    - Monocyte
    - Erythrocyte
    - Granulocyte/Monocyte Precursor
    - Monoblast
    - Thrombopoietin
    - Megakaryocyte
    - Platelets
    - T-lymphocyte
    - B-lymphocyte

Differentiating Post-Mitotic Compartment

Inflammatory Mediators

Macrophage

Lymphoid Lineage

Lymphocyte

Lymphoid Precursor
RED BLOOD CELLS

*Introduction*

- Normal - Anucleate, highly flexible biconcave discs, 80-100 femtoliters in volume
- Flexibility essential for passage through capillaries
- Major roles - Carriers of oxygen to & carbon dioxide away from cells
ERYTHROPOIETIN

- Cytokine - Produced in the kidney
- Necessary for erythroid proliferation and differentiation
- Absence results in apoptosis (programmed cell death) of erythroid committed cells
- Anemia of renal failure 2° to lack of EPO
ERYTHROPOIETIN

Mechanism of Action

PPSC → BFU-E → CFU-E

Stimulates proliferation

Accelerates Maturation

Reticulocyte → Mature RBC
ERYTHROPOIETIN

Mechanism of Action

- Binds specifically to Erythropoietin Receptor
- Transmembrane protein; cytokine receptor superfamily
- Binding leads to dimerization of receptor
- Dimerization activates tyrosine kinase activity
ERYTHROPOIETIN

Mechanism of Action

- Multiple cytoplasmic & nuclear proteins phosphorylated
- Nuclear signal sent to activate production of proteins leading to proliferation and differentiation
Erythropoietin
Response to Administration

rhuEPO 150 u/kg 3x/wk
RBC Precursors

- Pronormoblast
- Basophilic normoblast
- Polychromatophilic Normoblast
- Orthrochromatophilic Normoblast
- Reticulocyte
- Mature Red Blood Cell
- 5-7 days from Pronormoblast to Reticulocyte
RBC Assessment

- **Number** - Generally done by automated counters, using impedance measures.
- **Size** - Large, normal size, or small; all same size *versus* variable sizes (anisocytosis). Mean volume by automated counter.
- **Shape** - Normal biconcave disc, *versus* spherocytes, *versus* oddly shaped cells (poikilocytosis).
- **Color** - Generally an artifact of size of cell.
## Red Blood Cells

### Normal Values

<table>
<thead>
<tr>
<th>RBC Parameters</th>
<th>Normal Values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hematocrit</td>
<td></td>
</tr>
<tr>
<td>Females</td>
<td>35-47%</td>
</tr>
<tr>
<td>Males</td>
<td>40-52%</td>
</tr>
<tr>
<td>Hemoglobin</td>
<td></td>
</tr>
<tr>
<td>Females</td>
<td>12.0-16.0 gm/dl</td>
</tr>
<tr>
<td>Males</td>
<td>13.5-17.5 gm/dl</td>
</tr>
<tr>
<td>MCV</td>
<td></td>
</tr>
<tr>
<td></td>
<td>80-100 fl</td>
</tr>
<tr>
<td>Reticulocyte Count</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.2-2.0%</td>
</tr>
</tbody>
</table>
RETICULOCYTE

- Young red blood cell; still have small amounts of RNA present in them
- Tend to stain somewhat bluer than mature RBC’s on Wright stain (polychromatophilic)
- Slightly larger than mature RBC
- Undergo removal of RNA on passing through spleen, in 1st day of life
- Can be detected using supravital stain
- Important marker of RBC production
RETICUCULOCYTE COUNT

Absolute Value

- = Retic % x RBC Count
  - eg 0.01 x 5,000,000 = 50,000
- Normal up to 100,000
- More accurate way to assess body’s response to anemia
ANEMIA

Causes

• Blood loss
• Decreased production of red blood cells (Marrow failure)
• Increased destruction of red blood cells
  – Hemolysis
• Distinguished by reticulocyte count
  – Decreased in states of decreased production
  – Increased in destruction of red blood cells
HYPOPROLIFERATIVE ANEMIAS

Maturation Disorders

Hemolytic Anemias
RBC DESTRUCTION - EXTRAVASCULAR

*Markers*

- Heme metabolized to bilirubin in macrophage; globin metabolized intracellularly
- Unconjugated bilirubin excreted into plasma & carried to liver
- Bilirubin conjugated in liver & excreted into bile & then into upper GI tract
- Conjugated bilirubin passes to lower GI tract & metabolized to urobilinogen, which is excreted into stool & urine
RBC DESTRUCTION - INTRAVASCULAR

- Free Hemoglobin in circulation leads to:
  - Binding of hemoglobin to haptoglobin, yielding low plasma haptoglobin
  - Hemoglobin filtered by kidney & reabsorbed by tubules, leading to hemosiderinuria
  - Capacity of tubules to reabsorb protein exceeded, yielding hemoglobinuria
INTRAVASCULAR HEMOLYSIS
HEMOLYTIC ANEMIA
Commonly used Tests

<table>
<thead>
<tr>
<th>Test</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reticulocyte Count</td>
<td>Increased</td>
</tr>
<tr>
<td>Unconjugated Bilirubin</td>
<td>Increased</td>
</tr>
<tr>
<td>Lactate Dehydrogenase</td>
<td>Increased</td>
</tr>
<tr>
<td>Haptoglobin</td>
<td>Decreased</td>
</tr>
<tr>
<td>Urine Hemoglobin</td>
<td>Present</td>
</tr>
<tr>
<td>Urine Hemosiderin</td>
<td>Present</td>
</tr>
</tbody>
</table>

Problems with sensitivity & specificity