HEMATOLOGY/HEMATOPOIESIS

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 & CO₂ transport
- Coagulation/platelets/Maintenance of vascular integrity
- White Blood Cells/Protection *versus* pathogens/microorganisms
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

PLURIPOTENT STEM CELLS
HEMATOPOIESIS

HEMATOPOIESIS – GROWTH FACTORS
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 of erythroid committed cells
- Anemia of renal failure 2° to lack of EPO

**ERYTHROPOIETIN**

*Mechanism of Action*

- Stimulates Proliferation
- Accelerates Maturation
- EPO
- CFU-E
- BFU-E
- 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

GROWTH FACTORS – Mechanisms of Action
ERYTHROPOIETIN
Mechanism of Action

- Multiple cytoplasmic & nuclear proteins phosphorylated via JAK-STAT pathways
- Nuclear signal sent to activate production of proteins leading to proliferation and differentiation
- Signal also sent to block apoptosis
Erythropoietin

Response to Administration

Erythropoietin (aerosol units)

Hemoglobin (g/dL)

Hematocrit

Time

rhuEPO 150 u/kg 3x/wk
RBC Precursors

- Pronormoblast
- Basophilic normoblast
- Polychromatophilic Normoblast
- Orthochromatophilic Normoblast
- Reticulocyte
- Mature Red Blood Cell
- 5-7 days from Pronormoblast to Reticulocyte
RETICULOCYTE

- Important marker of RBC production
- 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

RETICULOCYTE COUNT

Absolute Value

- = Retic % x RBC Count
  - eg 0.01 x 5,000,000 = 50,000
- Normal up to 100,000/μl
- More accurate way to assess body’s response to anemia
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>80-100 fl</td>
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<tr>
<td>Reticulocyte Count</td>
<td>0.2-2.0%</td>
</tr>
</tbody>
</table>
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
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

Acute Hemolytic Event

Serum Haptoglobin

Hemoglobin

Urine Hemosiderin

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