Peripheral blood & Hematopoiesis
Part 1 Blood

- Dr. Larry Johnson
- Texas A&M University
Objectives of Peripheral blood & Hematopoiesis

Part 1
• Examine a blood smear and distinguish each of the formed elements.
• Describe the functions of the granulocytes, agranulocytes, platelets, and erythrocytes.
• Know the relative cell counts for each of the formed elements in the blood.

Part 2
• Describe the normal development of red and white blood cells.
BLOOD - DIAGNOSTIC VALUE - MOST EXAMINED

TYPES OF INFORMATION:

IDENTIFY NATURE OF DISEASE
  VIRAL – T LYMPHOCYTES
  BACTERIAL – NEUTROPHILS
  PARASITIC – EOSINOPHILS

FOLLOW THE COURSE OF DISEASE

ALLOWS METHOD TO EVALUATE THE EFFECTIVENESS OF TREATMENT
BLOOD COMPOSITION

PACKED RED BLOOD CELLS
(HEMATOCRIT)

BUFFY COAT
(WHITE BLOOD CELLS)

BLOOD PLASMA
BLOOD PLASMA
LIQUID PART OF BLOOD - 55% OF WHOLE BLOOD

COMPOSITION:
WATER
ALBUMIN - OSMOTIC PRESSURE and NUTRALIZES PROTEASES
GLOBULIN - ANTIBODIES (IgG, IgE, ETC.)
TRANSFERRIN - IRON, COPPER, ZINC
CHYLOMICRONS - LIPIDS FROM INTESTINE
LOW-DENSITY LIPOPROTEIN - CHOLESTEROL REMOVAL
BLOOD PLASMA

COMPOSITION con’t:

PLASMA CLOTTING FACTORS
- FIBRINOGEN
- PROTHROMBIN
- THROMBOPLASTIN
- THROMBIN

HORMONES - DIRECT ACTIVITY OF CELLS

CHEMOTAXIS FACTORS – ATTRACT IMMUNE CELLS
BLOOD SERUM

SERUM – YELLOW LIQUID FROM CLOTTED BLOOD (e.g., PLASMA MINUS CLOTTING FACTORS)
BLOOD (definition and function)

BLOOD - FLUID TISSUE COMPOSED OF ERYTHROCYTES (RBC), LEUKOCYTES (WBC), AND PLATELETS SUSPENDED IN BLOOD PLASMA

FUNCTION OF BLOOD IS TRANSPORTATION OF CELLS AND FLUID

RBC $O_2$ to and $CO_2$ from TISSUE
FUNCTION OF BLOOD IS TRANSPORTATION OF CELLS AND FLUID

WBC IMMUNE DEFENSE OF BODY

PLASMA NUTRIENTS TO TISSUE
WASTE FROM TISSUE
PROTEINS TO HOLD WATER IN PLASMA
HORMONES AND OTHER INFORMATIONAL MEDIATORS

PLATELETS PREVENT LOSS OF TRANSPORTATION
Blood summary: Composition and cells
Blood plasma is the liquid component of blood that holds blood cells in whole blood vs. blood serum is blood plasma without clotting factors.
GRANULAR LEUKOCYTES

NONGRANULAR LEUKOCYTES
Slide 21: Blood smear (Giemsa stain)

Lymphocyte
Eosinophil
Monocyte
Neutrophil
Basophil (Slide 22)
Platelets
<table>
<thead>
<tr>
<th>Cell Type</th>
<th>% Prevalence in normal blood</th>
<th>Distinguishing characteristics</th>
<th>Contents of specific granules</th>
<th>Life span</th>
</tr>
</thead>
</table>
| Neutrophils | 54-62%                       | 3-5 connected lobes, cytoplasm is granulated                        | 1. Azurophilic primary granules  
2. Myeloperoxidase  
3. Lysozyme  
3. Defensins  
2. Specific secondary granules | 1-4 days          |
| Lymphocytes | 25-33%                       | No granules in cytoplasm, dense-staining nucleus surrounded by narrow cytoplasmic rim | Agranular                                                                                      | Hours to many years |
| Monocytes   | 3-7%                         | Largest agranular leukocyte with horseshoe-shaped nucleus           | Agranular                                                                                     | Hours to years    |
| Eosinophils | 1-3%                         | Typically bilobed nucleus, cytoplasm has large eosinophilic granules | 1. Major basic proteins  
2. Others                                                                                   | 1-2 weeks         |
| Basophils   | 0-0.75%                      | Cytoplasm contains dark blue or brown granules, basophilic nucleus normally obscured by dense cytoplasmic granules | 1. Heparin  
2. Histamine  
3. Others                                                                                   | Several months    |
<table>
<thead>
<tr>
<th>CELL TYPE</th>
<th>MAIN FUNCTIONS</th>
</tr>
</thead>
<tbody>
<tr>
<td>ERYTHROCYTES</td>
<td>( \text{CO}_2 ) AND ( \text{O}_2 ) TRANSPORT</td>
</tr>
<tr>
<td>NEUTROPHILS</td>
<td>PHAGOCYTOSIS OF BACTERIA</td>
</tr>
<tr>
<td>EOSINOPHILS</td>
<td>PARASITIC INFECTIONS, INFLAMMATORY PROCESSES</td>
</tr>
<tr>
<td>BASOPHILS</td>
<td>RELEASE OF HISTAMINE AND OTHER INFLAMMATION MEDIATORS</td>
</tr>
<tr>
<td>MONOCYTES</td>
<td>MONONUCLEAR-PHAGOCYTE SYSTEM become macrophages</td>
</tr>
</tbody>
</table>
FORMED ELEMENTS (non-fluid, cellular) AND FUNCTIONS IN BLOOD con’t

<table>
<thead>
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<th>MAIN FUNCTIONS</th>
</tr>
</thead>
<tbody>
<tr>
<td>B LYMPHOCYTES</td>
<td>GENERATION OF ANTIBODY-PRODUCING PLASMA CELLS</td>
</tr>
<tr>
<td>T LYMPHOCYTES</td>
<td>KILLING OF VIRUS-INFECTED CELLS</td>
</tr>
<tr>
<td>NATURAL KILLER</td>
<td>KILLING OF SOME TUMOR AND VIRUS-INFECTED CELLS</td>
</tr>
<tr>
<td>(CYTOTOXIC T CELL)</td>
<td></td>
</tr>
<tr>
<td>PLATELETS</td>
<td>CLOTTING OF BLOOD</td>
</tr>
</tbody>
</table>
ERYTHROCYTES

MINUTE CORPUSCLES - RED COLOR OF BLOOD (iron)
BONE MARROW DERIVED
ANUCLEATED CELLS (MAMMALS)
  • NONFUNCTIONAL NUCLEI IN BIRDS, REPTILES, AMPHIBIA, AND FISH

4.8 TO 5.4 X 10^6/MM^3 = 1µL OF BLOOD, 45% OF BLOOD
25-30 X 10^{12}/PERSON (LARGER AT HIGH ELEVATION)
Peripheral blood smear (Leishman-Giemsa) basophil, eosinophil, and neutrophils

The basic functions of the leukocytes:

- Neutrophil: bacteria, fungi
- Eosinophil: parasites, allergic inflammatory responses
- Basophil: release histamine for inflammatory response
- Lymphocyte: B cells, T cells, Natural killer cells
- Monocyte: migrates from blood to tissue and becomes a macrophage
Peripheral blood smear (May-Grunwald-Giemsa)
Plasma cells
EM of white blood cells
Neutrophils
NEUROPHILS (GRANDULES)

AZUROPHILIC ACID PHOSPHATASE (HYDROLYTIC ENZYMES) PRIMARY LYOSOMES

SPECIFIC BASIC PROTEIN (PHAGOCYTINS, ANTI-BACTERIAL ACTION)
EOSINOPHILS

8-12 DAY LIFE SPAN
TRANSIENT OF BLOOD - 3 TO 4 HRS.

SPECIFIC GRANULES - RED/ORANGE, LARGE, CRYSTALLOIDS
  • MAJOR BASIC PROTEIN (KILLS PARASITIC WORMS)

COMMON IN LAMINA PROPRIA (CT UNDER LINING EPITHELIUM)
  • ALIMENTARY TRACT
  • RESPIRATORY TRACT

ATTRACTED BY CHEMOTACTIC FACTORS GIVEN OFF BY MAST CELLS AND BASOPHILS

ROLE
  • PARASITIC DISEASES
  • ALLERGY
<table>
<thead>
<tr>
<th>Cells</th>
<th>Specific Granules</th>
<th>Azurophilic Granules</th>
</tr>
</thead>
<tbody>
<tr>
<td>Neutrophils</td>
<td>Alkaline phosphatase&lt;br&gt;Collagenase&lt;br&gt;Lactoferrin&lt;br&gt;Lysozyme (2/3)</td>
<td>Acid phosphatase&lt;br&gt;α-Mannosidase&lt;br&gt;Arylsulfatase&lt;br&gt;β-Galactosidase&lt;br&gt;β-Glucuronidase&lt;br&gt;Cathepsin&lt;br&gt;5′Nucleotidase&lt;br&gt;Elastase&lt;br&gt;Collagenase&lt;br&gt;Myeloperoxidase&lt;br&gt;Lysozyme&lt;br&gt;Acidic mucosubstances&lt;br&gt;Cationic antibacterial proteins</td>
</tr>
<tr>
<td>Eosinophils</td>
<td>Acid phosphatase&lt;br&gt;Arylsulfatase&lt;br&gt;β-Glucuronidase&lt;br&gt;Cathepsin&lt;br&gt;Phospholipase&lt;br&gt;RNAse&lt;br&gt;Eosinophilic peroxidase&lt;br&gt;Eosinophilic chemo-tactic factor&lt;br&gt;Major basic protein</td>
<td></td>
</tr>
<tr>
<td>Basophils</td>
<td>Eosinophilic chemo-tactic factor&lt;br&gt;Heparin&lt;br&gt;Histamine&lt;br&gt;Peroxidase</td>
<td></td>
</tr>
</tbody>
</table>
BASOPHILS

LEAST NUMEROUS WBC

SPECIFIC GRANULES (BLUE, LARGE, HISTAMINE)
  • METACHROMASIA (HEPARIN in several varied colors)

MAY BE INDUCED TO DEGRANULATE (LIKE MAST CELLS)

SPECIFIC RECEPTORS FOR IgE IMMUNOGLOBULIN
  • IgE USUALLY DILUTES IN BLOOD
  • 20 FOLD IN PERSONS WITH HAY FEVER, ASTHMA, OR ALLERGIC DERMATITIS
BASOPHILS

HISTAMINE - ITCHING AND INCREASED VASCULAR PERMEABILITY – EDEMA

CUTANEOUS BASOPHIL HYPERSENSITIVITY - REBUCK WINDOW

- (BASOPHILS MIGRATE TO DERMIS - CELL MEDIATED IMMUNITY)

Note granules above nucleus
PLATELET FUNCTION - BLOOD CLOTTING

1. PRIMARY AGGREGATION OF PLATELET PLUG - ATTACHMENT TO DAMAGED TISSUE

2. SECONDARY AGGREGATION
   • ALPHA AND DELTA GRANULES – INDUCE FURTHER AGGREGATION

3. BLOOD COAGULATION - CASCADE OF PLASMA PROTEINS TO FORM BLOOD CLOT OR THROMBUS

4. CLOT RETRACTION - PLATELET ACTIN, MYOSIN AND ATP

5. CLOT REMOVAL - PLASMIN (PROTEOLYTIN ENZYME) AND PLATELET ENZYME CONTAINING GRANULES
PLATELET’S ROLE IN STOPPING BLEEDING

PRODUCTION OF SEROTONIN - VASOCONSTRICTION TO STOP BLOOD FLOW PRODUCE THROMBOPLASTIN

FIBRIN POLYMERIZES AND PRODUCES THE HEMOSTATIC PLUG
SUMMARY OF PLATELETS - THROMBOPLASTIDS

HAVE MANY METABOLIC FUNCTION OF WHOLE CELLS WHEN EXPOSED TO DAMAGED CELLS, STICKY - AGGREGATION

• STICK TO EACH OTHER
SUMMARY OF PLATELETS - THROMBOPLASTIDS

HAVE MANY METABOLIC FUNCTION OF WHOLE CELLS WHEN EXPOSED TO DAMAGED CELLS, STICKY - AGGREGATION

• STICK TO EACH OTHER

RELEASE ADP (FROM ATP INSIDE PLATELETS) WHICH ATTACHES TO OTHER PLATELETS
SUMMARY OF PLATELETS - THROMBOPLASTIDS

HAVE MANY METABOLIC FUNCTION OF WHOLE CELLS WHEN EXPOSED TO DAMAGED CELLS, STICKY - AGGREGATION

• STICK TO EACH OTHER

RELEASE ADP (FROM ATP INSIDE PLATELETS) WHICH ATTACHES TO OTHER PLATELETS

PRODUCE THROMBOPLASTIC WHICH CATALIZES PROTHROMBIN TO THROMBIN WHICH CATALIZES FIBRINOGEN TO FIBRIN,

FORMS NET TO CATCH RBC AND OTHER PLATELETS (BASIS OF THE BLOOD CLOT)
SUMMARY OF
PLATELETS
THROMBOPLASTIDS

ANUCLEATE (MAMMALS) ZONES

- HYDALOMERE – MICROTUBULES AND SMALL VESICLES
- GRANULOMERE - AZUROPHILIC GRANULES

GRANULES - SOME CONTAIN SEROTONIN - VASOCONSTRICTION

PLATELETS PRODUCED BY MEGAKARYOCYTES (BONE MARROW)
LAST 8 TO 11 DAYS
White blood cells in blood
White blood cells in blood
End of Part 1
Peripheral blood & Hematopoiesis
Part 2 Hematopoiesis

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Part 1

• Examine a blood smear and distinguish each of the formed elements.

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Part 2

• Describe the normal development of red and white blood cells.
Bone Marrow
The cell where hemoglobin appears in the developing erythrocytes is the basophilic erythroblasts (early erythroblast) and takes 4 days to form erythrocytes from stem cells in the bone marrow.
Bone marrow

Erythrocyte entering blood vessel
2. Describe the formation of **red blood cells**, neutrophils and lymphocytes in the bone marrow from their stem cells to their fully differentiated cells.

- **Erythropoiesis**
  - Takes about 1 week and stimulated by erythropoietin
  - Originate in bone marrow
  - Involves major cellular changes
    - Cell and nuclear volume decrease
    - Nucleoli diminishes and disappears
    - Chromatin density increases until nucleus extruded from cell
    - Decrease in polyribosomes (basophilia) and increase in hemoglobin (eosinophilic)
    - Mitochondria and other organelles disappear
  - Steps
    1. **Proerythroblast**
       - Large cell with loose, lacy chromatin, nucleoli, and basophilic cytoplasm
    2. **Basophilic erythroblast**
       - Strongly basophilic cytoplasm, condensed nucleus, and no visible nucleolus
    3. **Polychromatophilic erythroblast**
       - Cell volume is reduced, polysomes decrease, and some cytoplasmic areas fill with hemoglobin producing basophilic and acidophilic regions in cell
    4. **Orthochromatic erythroblast**
       - Cell and nuclear volumes continue to condense and no basophilia is evident
    5. **Reticulocyte**
       - Cell ejects nucleus to become a reticulocyte, has a small number of polyribosomes
    6. **Erythrocyte**
       - Enter blood circulation, lose polyribosomes, and mature as erythrocytes
Bone marrow
Platelets
Basophilic erythroblast

Platelets

Proerythroblast
Polychromatophilic erythroblast

Orthochromatophilic erythroblasts

Late polychromatophilic erythroblast

Basophilic erythroblast

Platelets

Polychromatophilic erythroblast

Proerythroblast
Polychromatophilic erythroblast
Orthochromatophilic erythroblasts
Late polychromatophilic erythroblast
Basophilic erythroblast
Proerythroblast
Erythrocytes
Platelets
Reticulocyte
# Red Blood Cell Series

<table>
<thead>
<tr>
<th>Cell Name</th>
<th>Size</th>
<th>Cytoplasm</th>
<th>Granules</th>
<th>Nucleus</th>
</tr>
</thead>
<tbody>
<tr>
<td>proerythroblast</td>
<td>16-20 µm</td>
<td>deep blue, no hemoglobin</td>
<td>none</td>
<td>large, round with delicate chromatin, 2-6 nucleoli</td>
</tr>
<tr>
<td>basophilic erythroblast (early erythroblast)</td>
<td>12-16 µm</td>
<td>tinge of hemoglobin</td>
<td>none</td>
<td>condensation of chromatin, no nucleoli</td>
</tr>
<tr>
<td>polychromatophilic erythroblast</td>
<td>10-16 µm</td>
<td>muddy color</td>
<td>none</td>
<td>checkerboard pattern</td>
</tr>
<tr>
<td>orthochromatophilic erythroblast (late erythroblast, normoblast)</td>
<td>8-10 µm</td>
<td>no basophilia</td>
<td>none</td>
<td>dense</td>
</tr>
</tbody>
</table>
Specific granules first appear in the myelocyte. Granulopoiesis typically takes 10 to 14 days.
## White Blood Cell Series

<table>
<thead>
<tr>
<th>Cell Name</th>
<th>Size</th>
<th>Cytoplasm</th>
<th>Granules</th>
<th>Nucleus</th>
</tr>
</thead>
<tbody>
<tr>
<td>myeloblast</td>
<td>15-20 µm</td>
<td>sky blue</td>
<td>no granules</td>
<td>red staining, round, delicate chromatin</td>
</tr>
<tr>
<td>promyelocyte</td>
<td>15-20 µm</td>
<td>sky blue</td>
<td>non-specific, 1-40 azurophilic granules</td>
<td>round to oval, delicate chromatin</td>
</tr>
<tr>
<td>neutrophilic myelocyte</td>
<td>17-25 µm</td>
<td>some basophilia, some pink</td>
<td>few fine neutrophilic granules</td>
<td>round to oval, delicate chromatin</td>
</tr>
<tr>
<td>neutrophilic metamyelocyte</td>
<td>12-16 µm</td>
<td>no basophilia, becoming pink</td>
<td>many fine neutrophilic granules</td>
<td>bean shaped, moderately dense chromatin</td>
</tr>
<tr>
<td>neutrophilic band or stab</td>
<td>12-16 µm</td>
<td>no basophilia, pink</td>
<td>many fine neutrophilic granules</td>
<td>band or S-shaped</td>
</tr>
<tr>
<td>eosinophilic myelocyte</td>
<td>13-17 µm</td>
<td>some basophilia, specific granules</td>
<td>few large eosinophilic granules</td>
<td>round to oval, delicate chromatin</td>
</tr>
<tr>
<td>eosinophilic metamyelocyte</td>
<td>12-16 µm</td>
<td>no basophilia, specific granules</td>
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Neutrophilic myelocyte

Reticulocytes
Neutrophilic myelocytes

Neutrophilic metamyelocytes

Platelets

Eosinophil or basophil

Neutrophilic myelocytes
Neutrophilic myelocytes

Neutrophilic metamyelocytes

Platelets

Eosinophil or basophil
Granulopoiesis

- Takes about 10 to 14 days
  - Majority of process is dominated by the synthesis of proteins for cytoplasmic granules
  - Steps
    1. **Myeloblast**
       - Most immature, finely dispersed chromatin, faint nuclei, and no cytoplasmic
    2. **Promyelocyte**
       - Basophilic cytoplasm and azurophilic granules being secreted in Golgi apparatus
    3. **Neutrophilic myelocyte**
       - Specific granules produced in Golgi, moderate number of azurophilic granules
    4. **Neutrophilic metamyelocyte**
       - Specific granules occupy most of cytoplasm, dispersed azurophilic granules, Golgi reduced
    5. **Neutrophilic band cell**
       - Nucleus is elongated but not yet polymorphic
    6. **Neutrophil**
       - 3-5 connected lobes, cytoplasm is granulated
Maturation of agranulocytes

- All lymphocyte progenitor cells originate in bone marrow and migrate to the thymus or other lymphoid organ (lymph node, spleen, etc.) to proliferate.
- Some lymphocytes migrate to thymus, and acquire properties to be T lymphocytes.
- Some lymphocytes differentiate into B lymphocytes in the bone marrow and then migrate to peripheral lymphoid organs.
- Steps
  - 1. Lymphoblast
    - Large cells capable of dividing 2-3X to form lymphocytes
  - 2. Lymphocyte
    - Maturation= nuclei becomes smaller, nucleoli become less visible, cells decrease in size
    - Bone marrow= B lymphocyte specialization
    - Thymus= T lymphocyte specialization
  - 3. Mature lymphocyte
    - Larger than newly formed lymphocytes
    - Acquire distinctive cell surface and other proteins
Cardiac stomach w/chronic infection
Cardiac stomach w/chronic infection
Lung with bronchus and lung – macrophage
• macrophage
Slide 17: in situ bone marrow

Megakaryocyte

Adipocyte
Slide 18: in situ bone marrow

Megakaryocytes

Adipocytes
Bone marrow

Megakaryocytes

Developing granulated cells
Lymphocytes

Unknown nuclei

Lymphocytes
Neutrophilic metamyelocytes

Neutrophilic band cells

Promyelocyte

Neutrophilic myelocytes

Neutrophilic band cell
Orthochromatophilic erythroblast
Reticulocytes
Basophilic erythroblast
Orthochromatophilic erythroblast
Neutrophilic metamyelocyte

Orthochromatophilic erythroblast

Proerythroblast

Orthochromatophilic erythroblast

Eosinophils or basophils

Unknown nuclei

Neutrophilic myelocyte

Neutrophilic band

Neutrophilic metamyelocyte

Orthochromatophilic erythroblast
White blood cell entering a blood vessel
Many illustrations in these VIBS Histology YouTube videos were modified from the following books and sources: Many thanks to original sources!

- Douglas P. Dohrman and TAMHSC Faculty 2012 Structure and Function of Human Organ Systems, Histology Laboratory Manual - Slide selections were largely based on this manual for first year medical students at TAMHSC
The End!