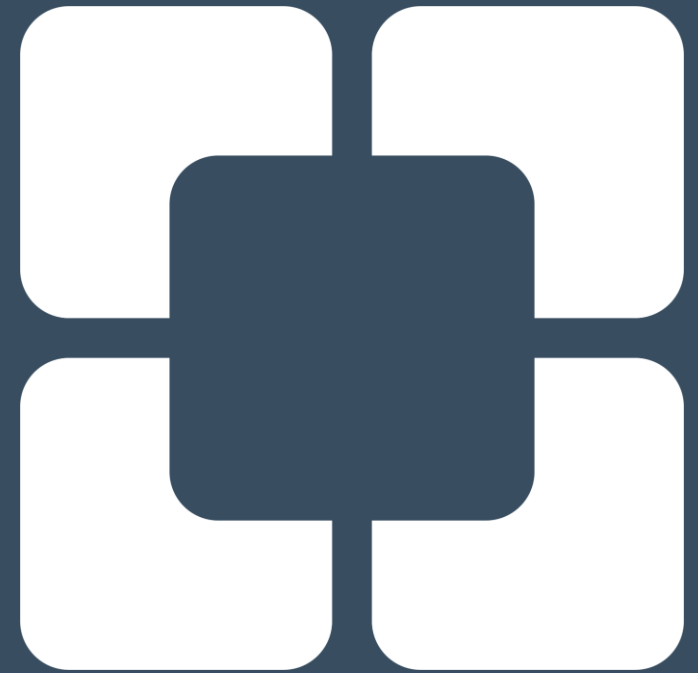


# White Blood Cell Morphology

Pinal Patel  
Laboratory Education Specialist



# Leukopoiesis

- Hematopoietic stem cells in the bone marrow



# Three Pools of Development

- Stem cell pool
  - hematopoietic stem cells (HSCs) capable of self-renewal and differentiation
- Proliferation (mitotic) pool
  - consists of CMP( granulocyte, erythrocyte, monocyte and megakaryocyte), GMP (myeloblasts, promyelocytes and myelocytes)

# Three Pools of Development

- Maturation (storage) pool
  - cells undergoes nuclear maturation and are available for release (metamyelocytes, bands and segmented neutrophils)



## Maturation Sequence

Myeloblast

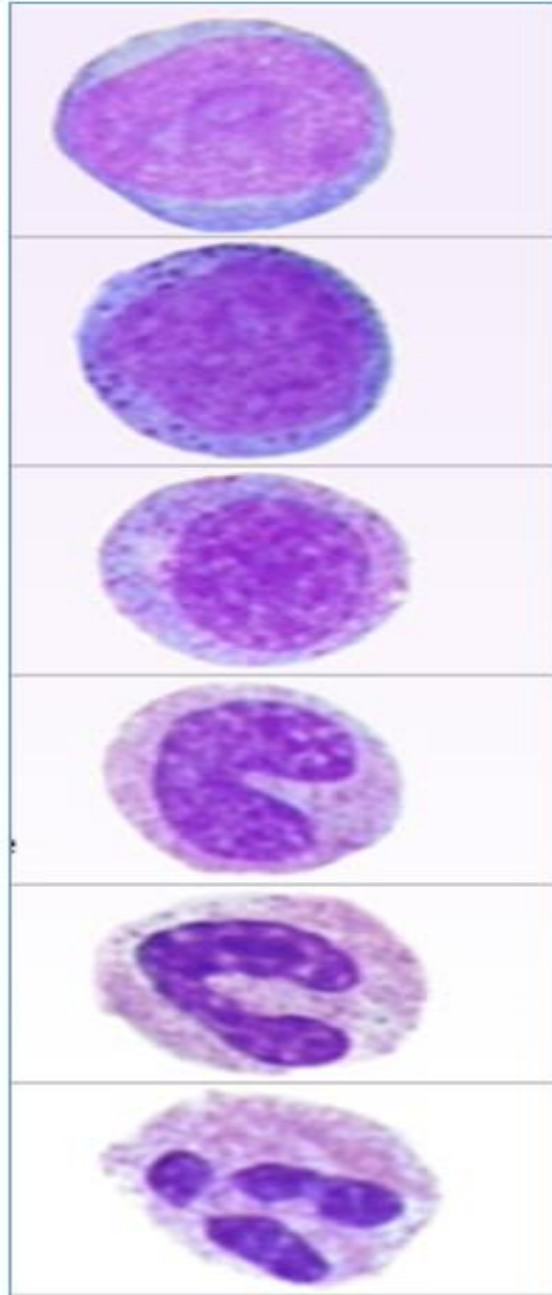
Promyelocyte

Myelocyte

Metamyelocyte

Band

Segmented  
Neutrophil



Seen in Bone  
Marrow

Peripheral  
Blood

# Myeloblast

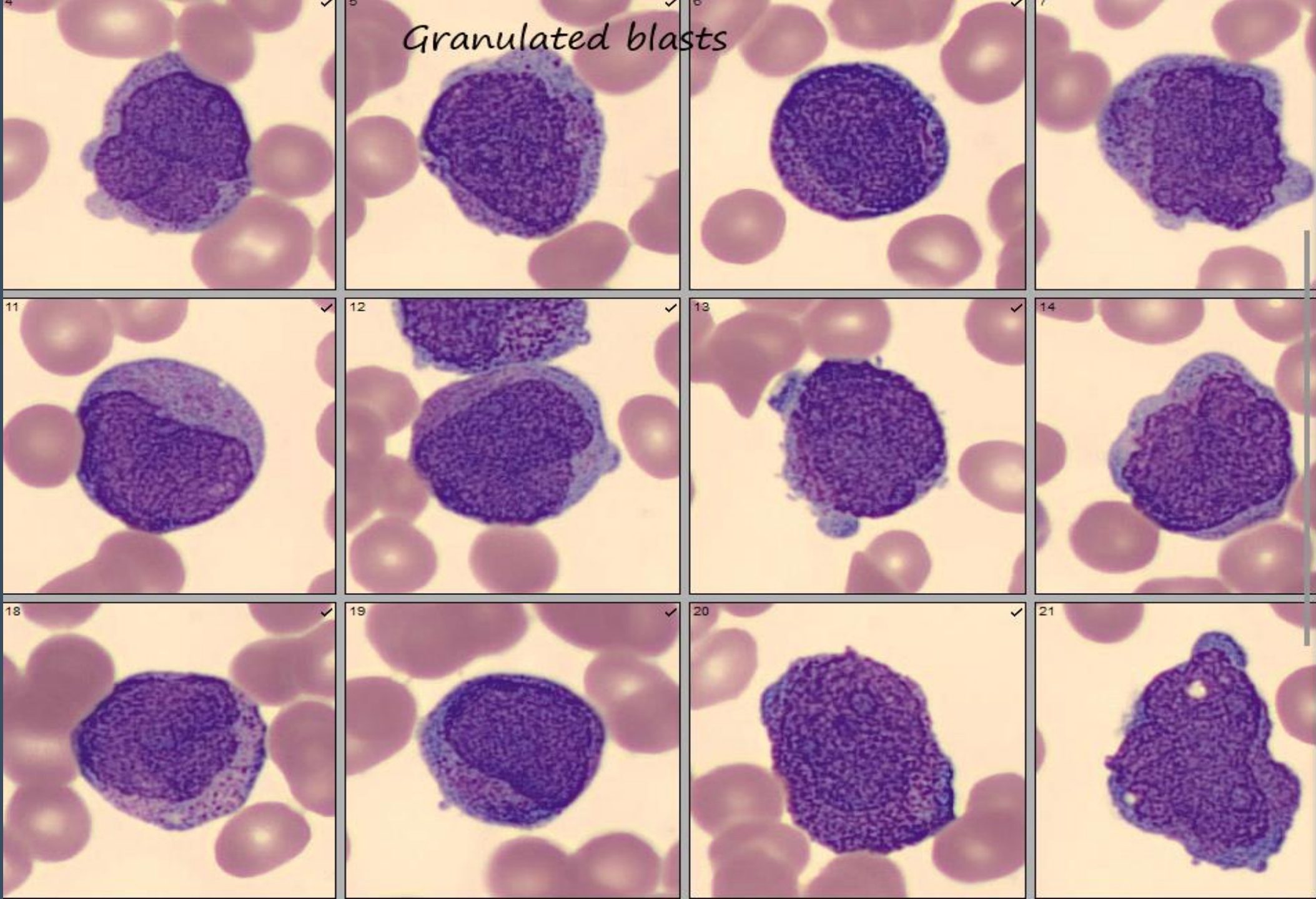
- High nucleus to cytoplasmic ratio (the nucleus occupies most of the cell with very little cytoplasm)
- Basophilic cytoplasm, fine nuclear chromatin and presence of nucleoli
- Some blasts may appear as granular blasts (certain acute myeloid leukemia)
- Auer rods can be present in the cytoplasm of blast







Granulated blasts

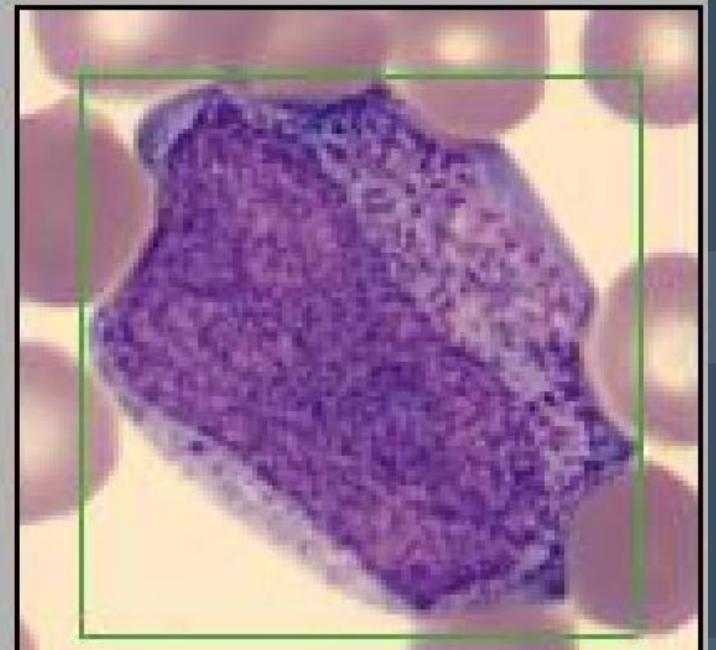
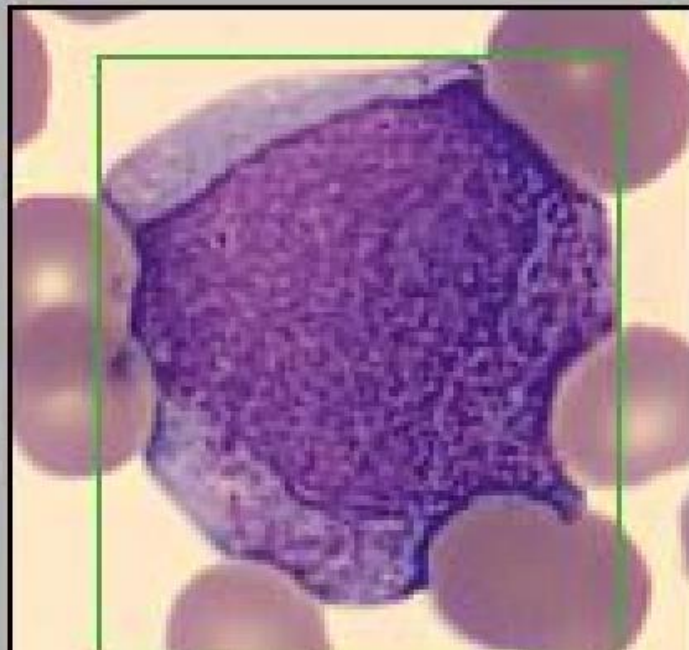
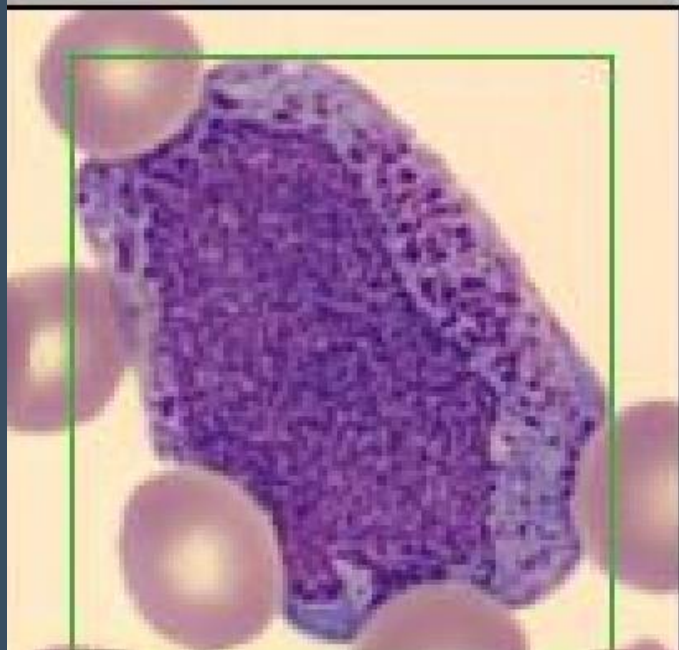
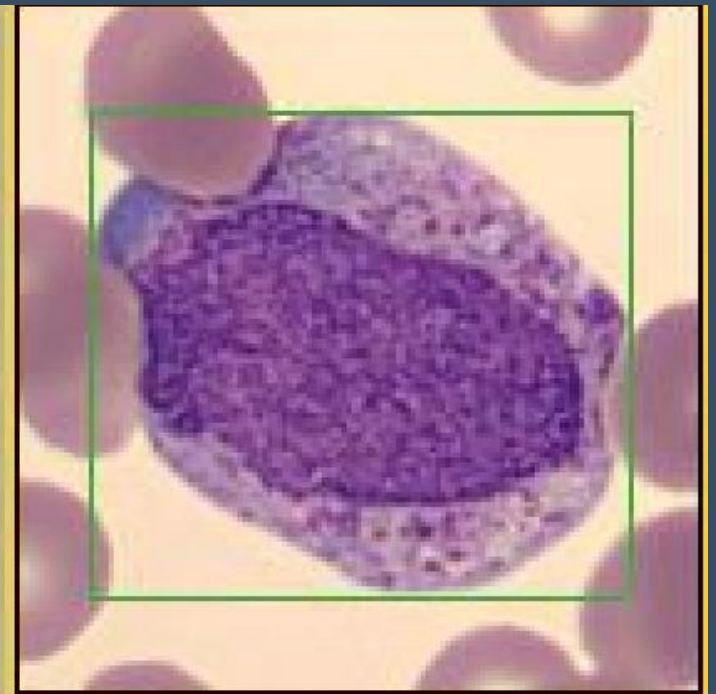
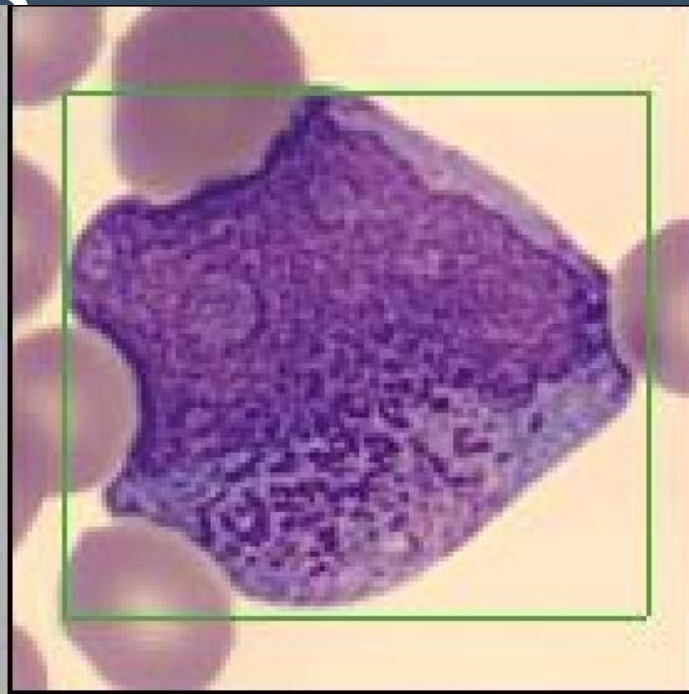
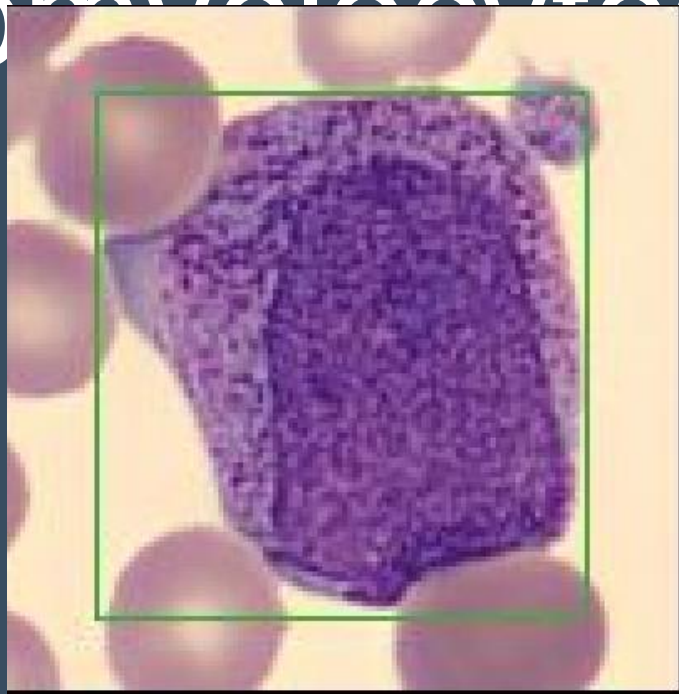




# Promyelocyte

- The cytoplasm is basophilic with primary (azurophilic) granules
- Round or oval shape nucleus
- Chromatin clumping in the nucleus may be visible
- Nucleoli can be seen but may be obscured by the granules

# Promyelocytes



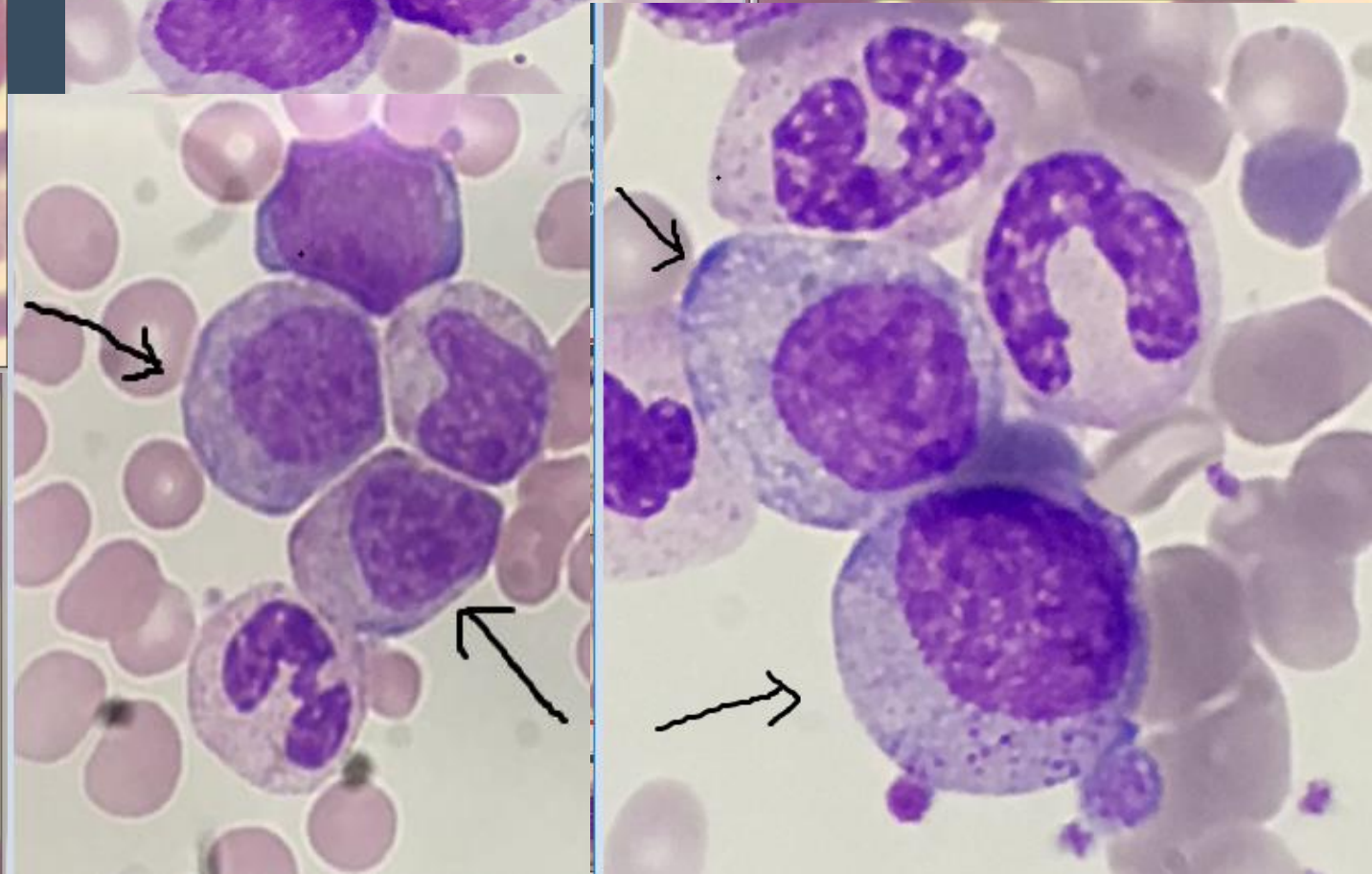
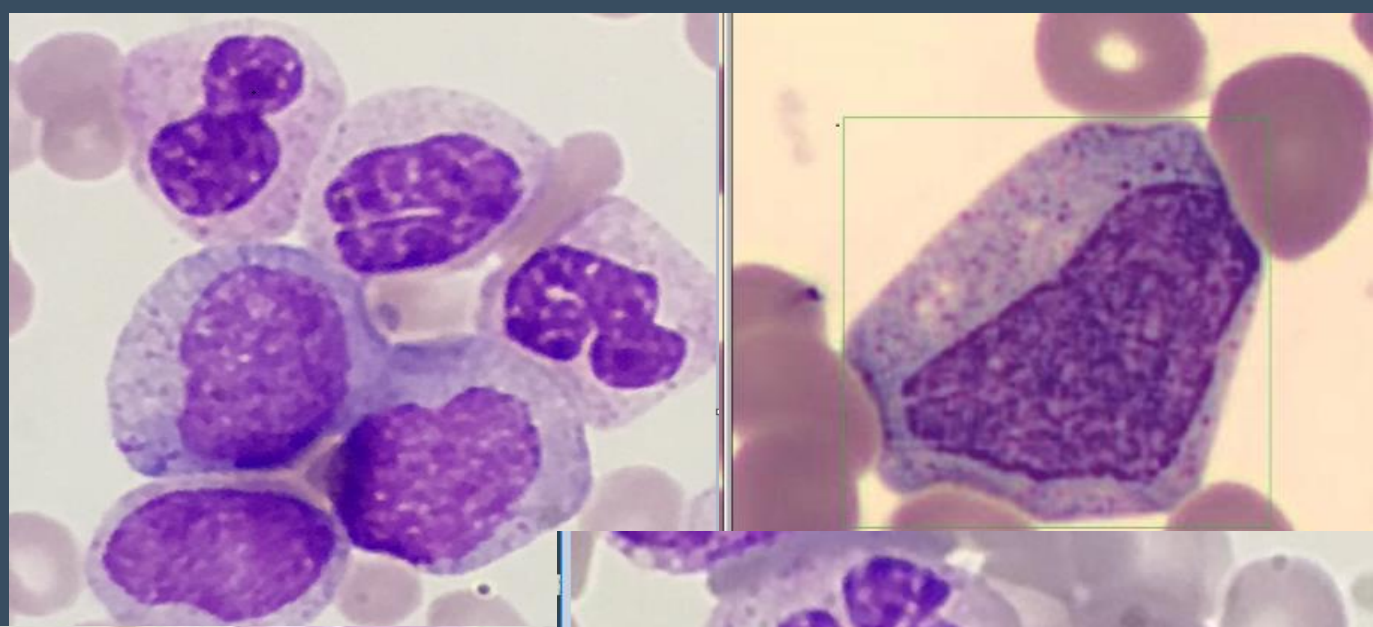
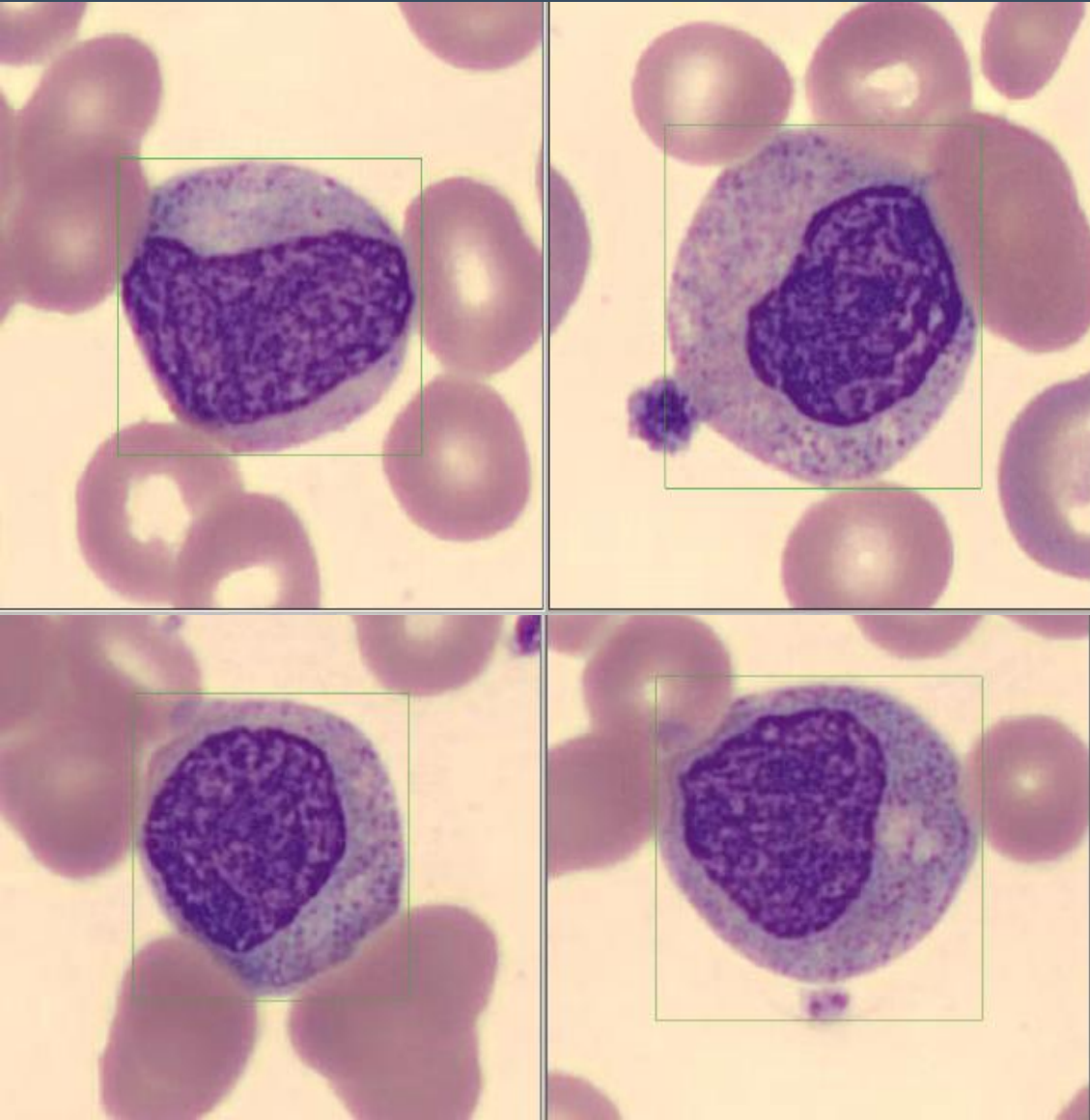
# Myelocyte

- Final stage of cell division
- Secondary granules begin to produce
- Lavender pink to blue cytoplasm
- Overall size of the cell is smaller than promyelocyte
- Nucleus heterochromatin
- Difficult to see nucleoli





# Myelocytes

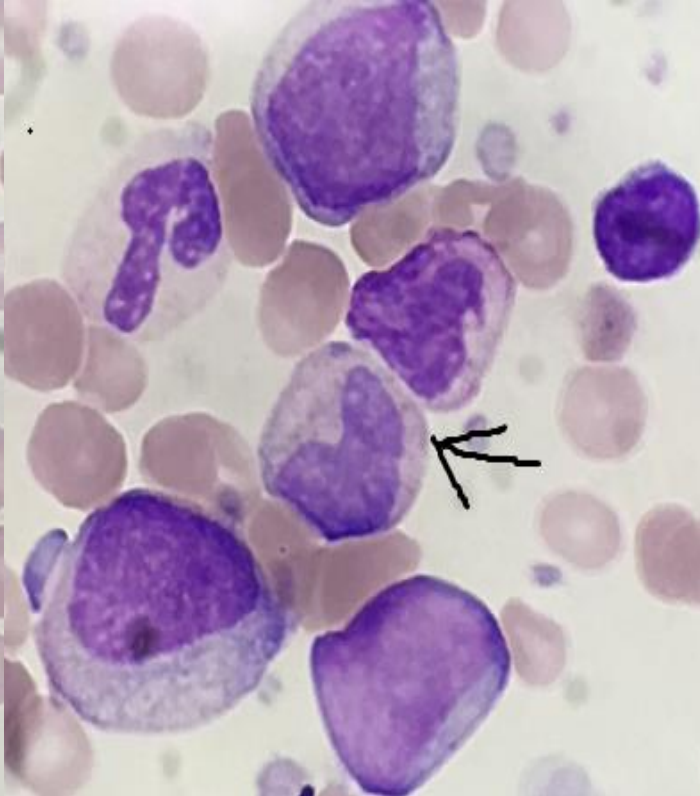
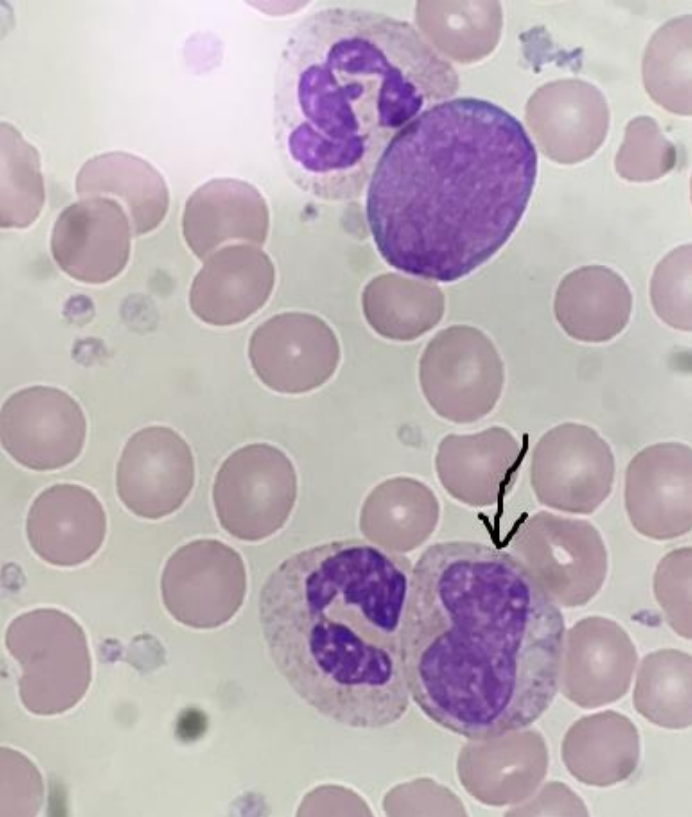
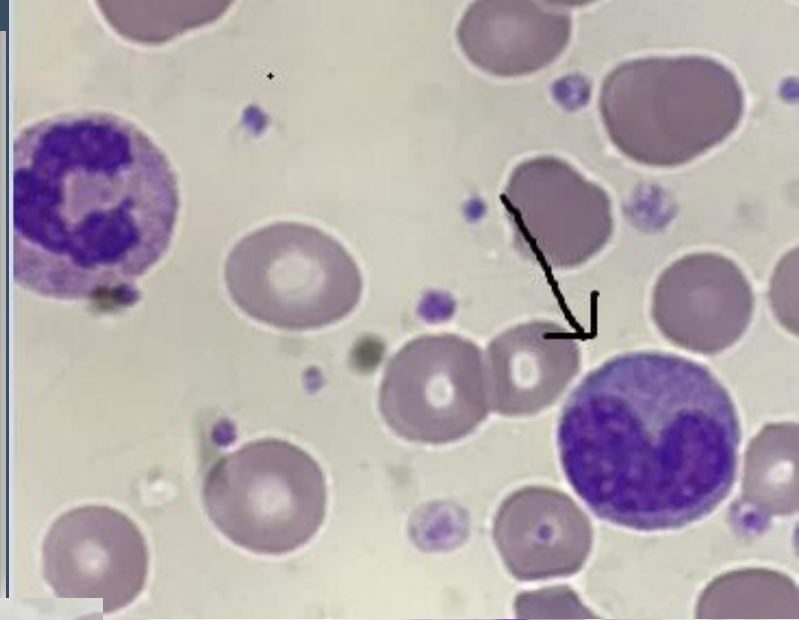
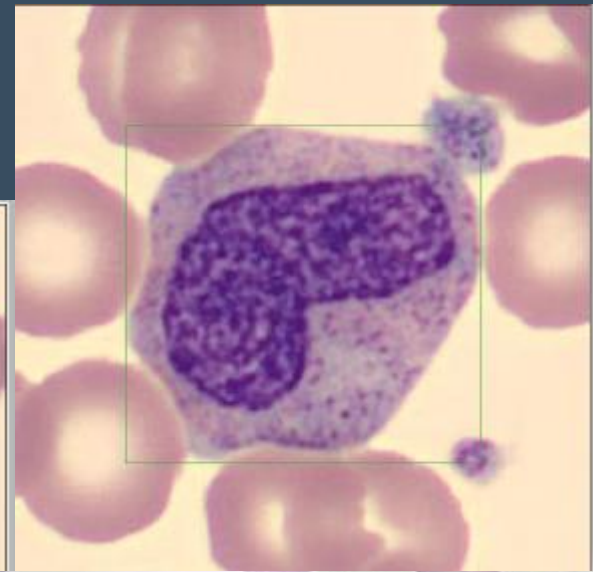
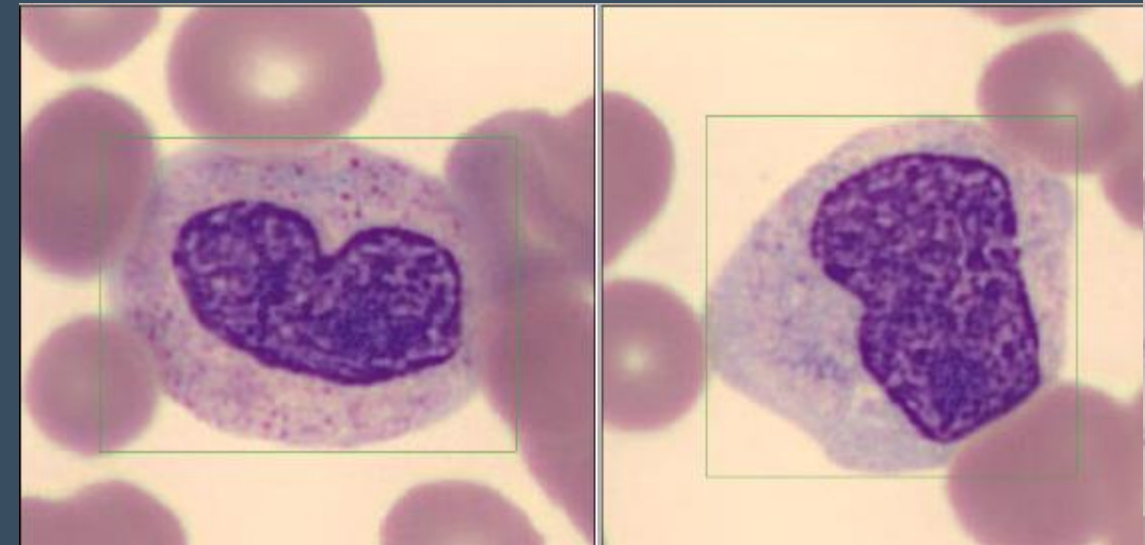
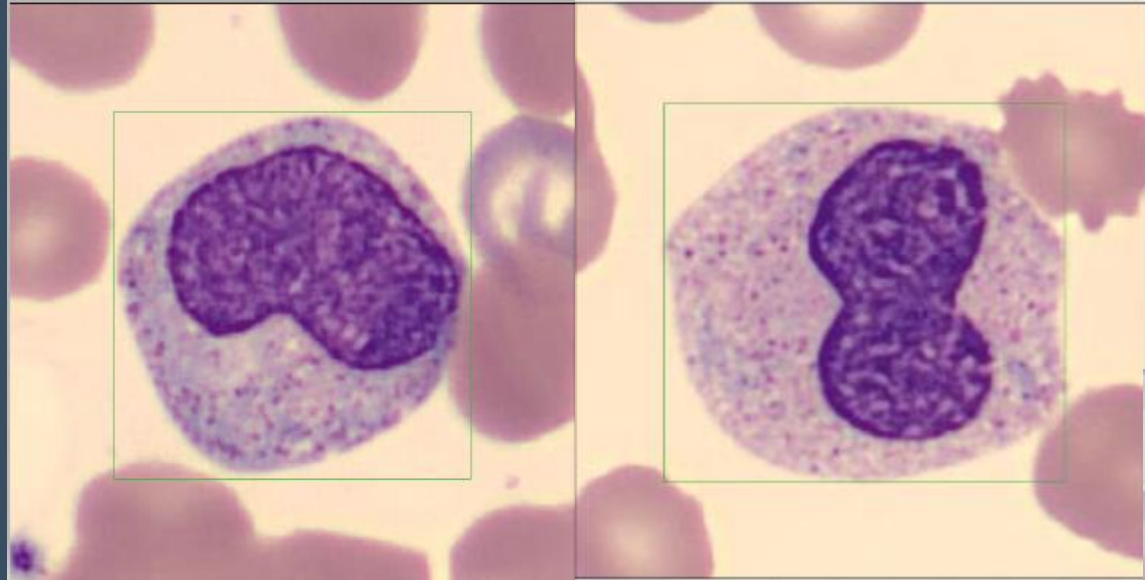


# Metamyelocyte

- The cells are no longer capable of cell division
- The nucleus is indented (Kidney bean shaped)
- Progressively clumped chromatin
- Little to no basophilia in the cytoplasm
- Nucleoli are absent



# Metamyelocytes

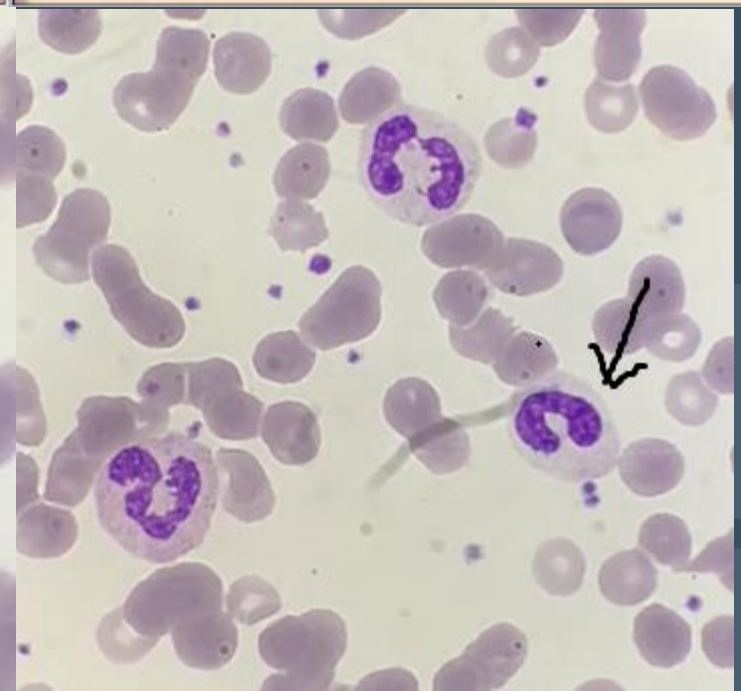
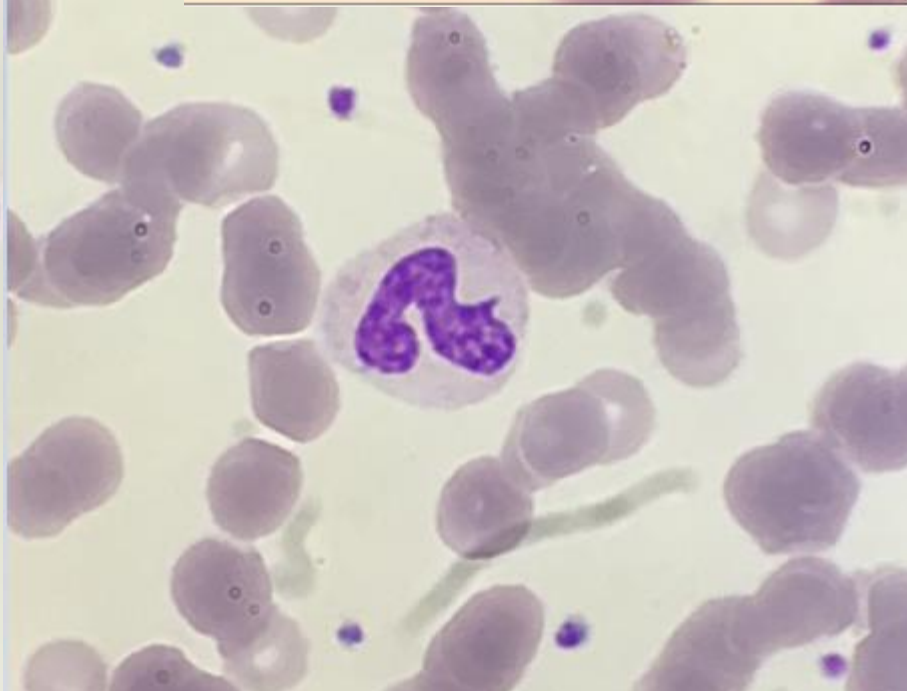
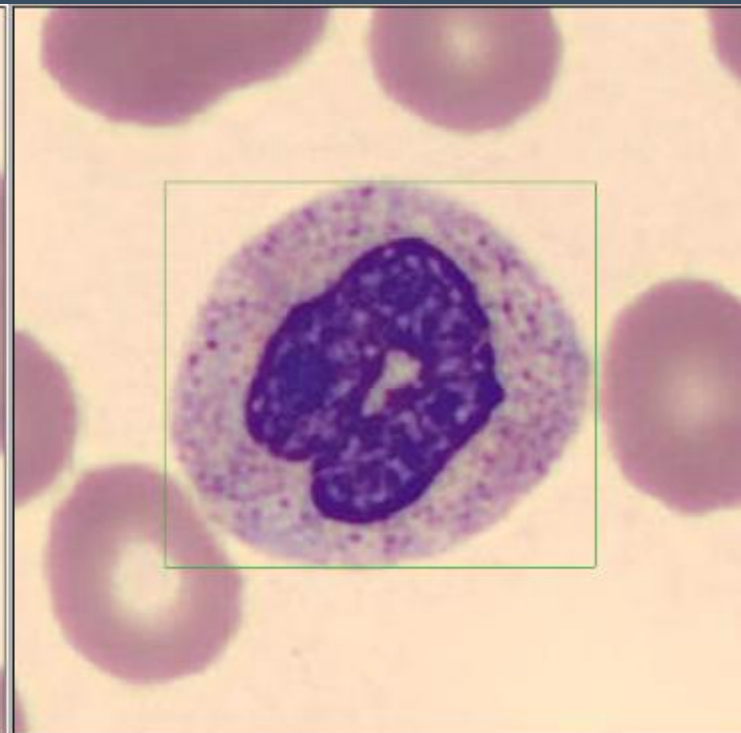
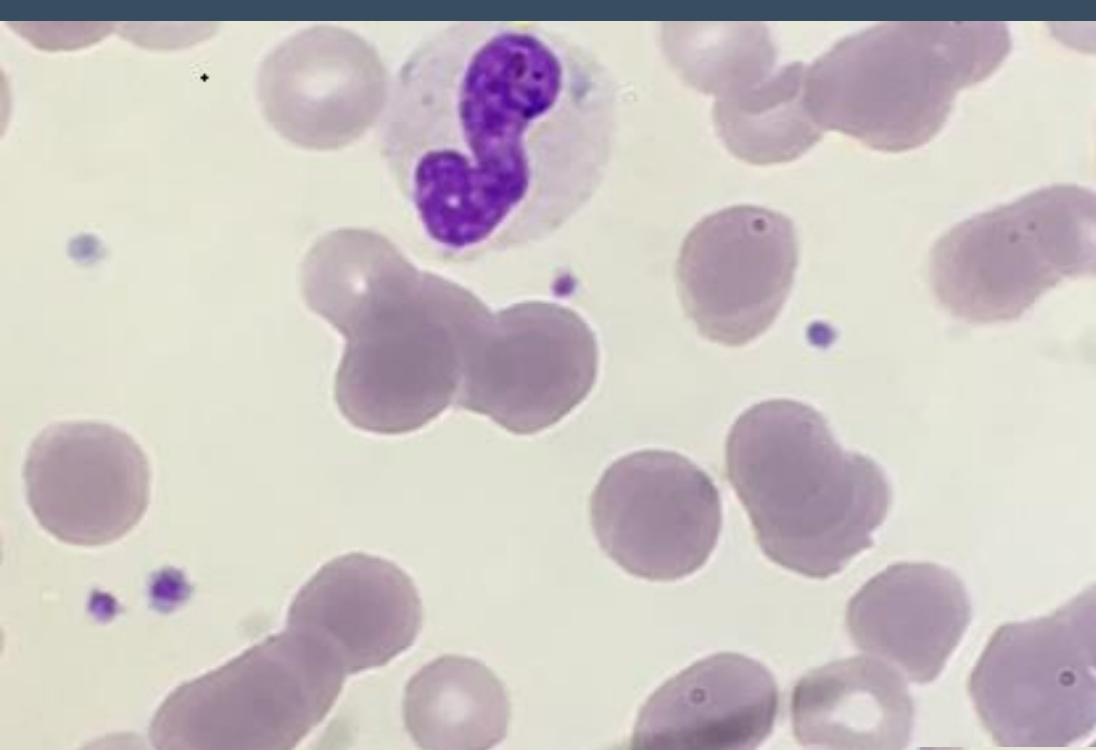


# Band Neutrophil

- Cytoplasmic basophilia is absent
- Nucleus is highly clumped
- Nucleus indentation exceeds one half the diameter of the nucleus and have same diameter throughout its length
- An elevated band count is useful in diagnosis of patients with infection







# Segmented Neutrophil

- Deep purplish blue nucleus with two to five lobes connected by filaments
- Pink cytoplasm with purple-pink granules
- The most predominant cell type in adult peripheral blood (50% -70 %)
- Normal values
  - Relative or percentage- 50-70 %
  - Absolute – 1.7-7.5 k/uL

# Segmented Neutrophil

- Neutrophilia – absolute count  $> 7.5/\mu\text{L}$  of blood
  - Physiological and pathological causes
- Neutropenia – absolute count  $< 1.7/\mu\text{L}$  of blood
- Each relative number multiplied by the total WBC count= absolute number
  - e.g. neutrophils % (67%) X WBC count( 10.5 K/ $\mu\text{L}$ )  
= ANC of 7.0



# Neutrophil Kinetics

- Transit time is ~12 days from myeloblast to neutrophil
- The half-life is ~ 7 hours in the blood
- In the peripheral blood, divided into circulating (CNP) and marginating (MNP) pools
  - Cells in MNP are loosely localized to the walls of capillaries in tissues (liver, spleen and lungs)
  - Marginated neutrophils make up a considerably large portion of peripheral neutrophils

# Neutrophil Kinetics

- Integrins and selectins are important in diapedesis
- Those who do not migrate into the tissues undergo apoptosis and removed by splenic macrophages
- Variable life span in the tissues depending on whether or not responding to the infectious or inflammatory agents.



# Neutrophil Functions

- Phagocytosis and destruction of foreign materials and microorganisms
  - Involves seeking (chemotaxis, motility and diapedesis) and destruction (phagocytosis and digestion)
    - Chemotactic agents ( produced by microorganisms, damaged cells, lymphocytes or phagocytes) bind to neutrophils
    - Neutrophils roll along endothelial cells using stronger adhesive molecules(selectins and integrins)



# Neutrophil Functions

- Diapedesis or transmigration of neutrophils either between or through endothelial cells
- Phagocytosis process begin once at the site of infection or inflammation
- Recognize the pathogens then attach and engulf by forming phagosome within the neutrophil cytoplasm
- The phagosome allows the reduced NADPH complex which generates reactive oxygen species such as hydrogen peroxide

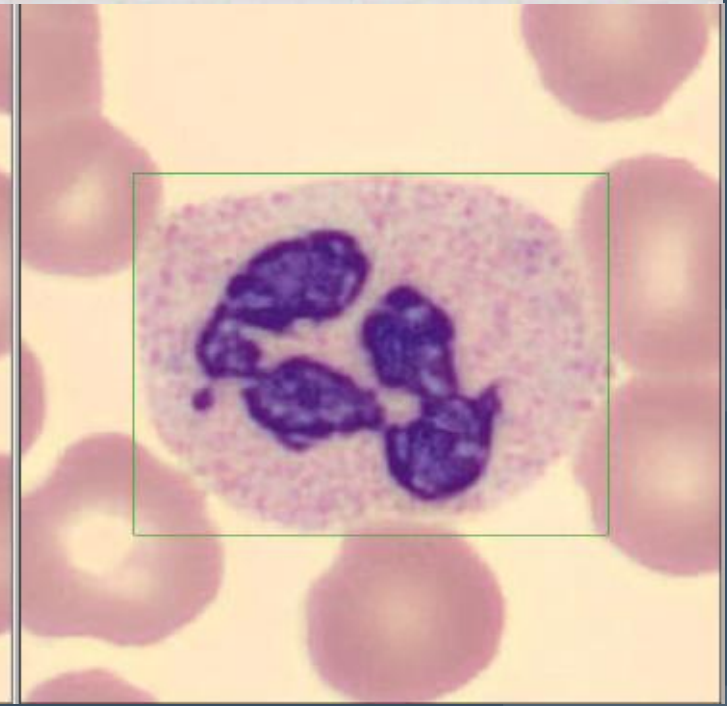
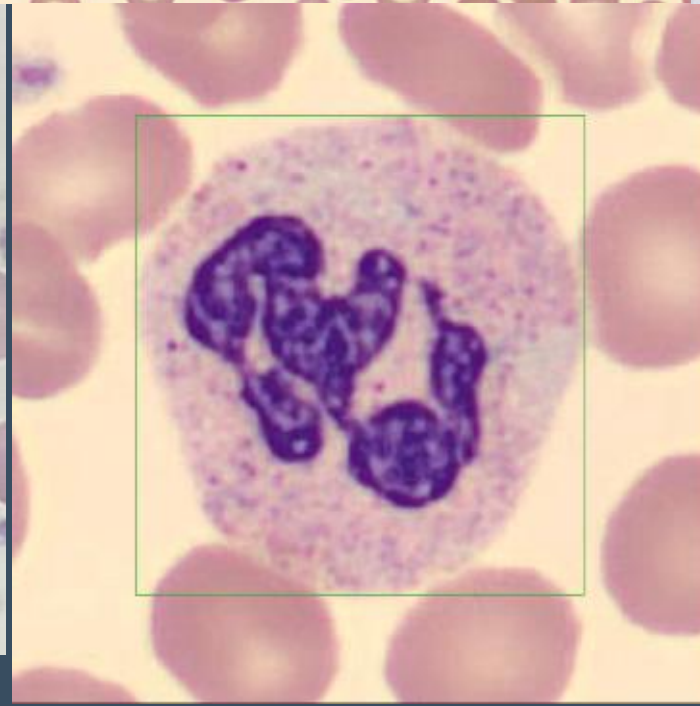
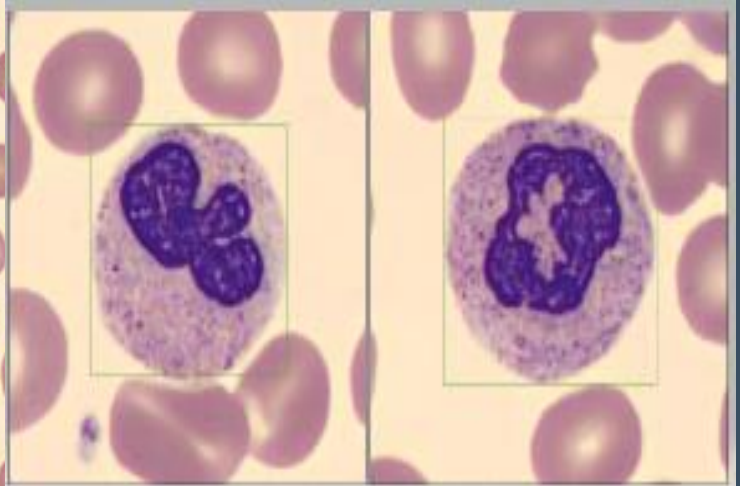
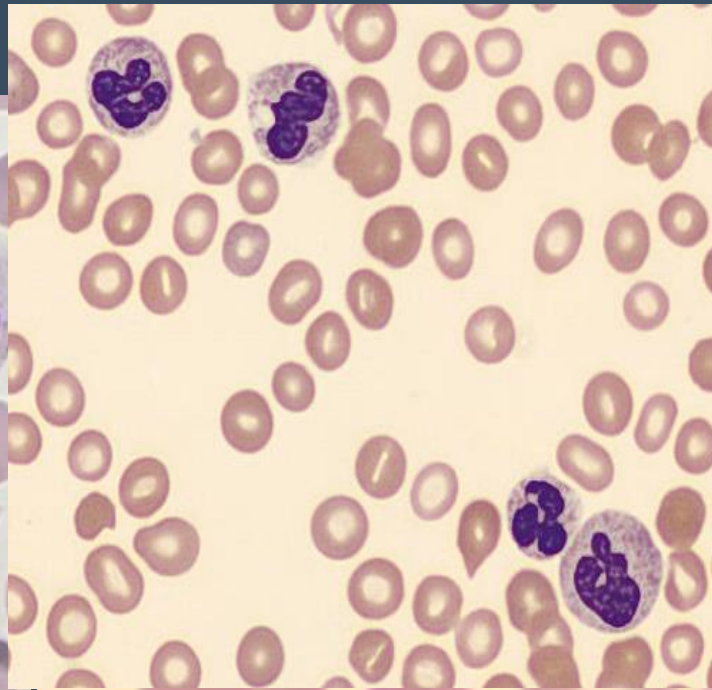
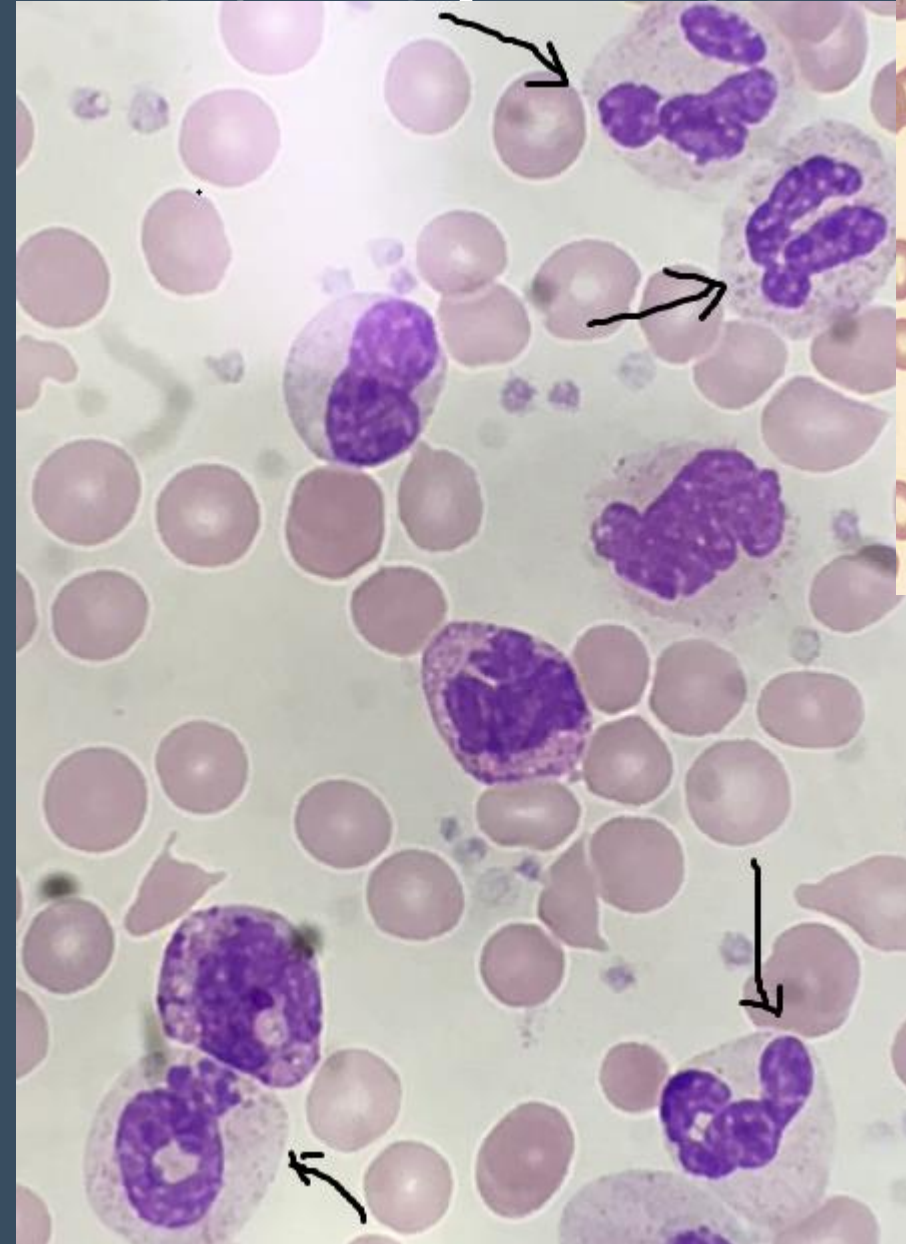
# Neutrophil Functions

- Generate neutrophil extracellular traps or NETs
  - Trap and kill gram positive and gram negative bacteria and fungi
- Source of transcobalamin I
  - Proper absorption of B12

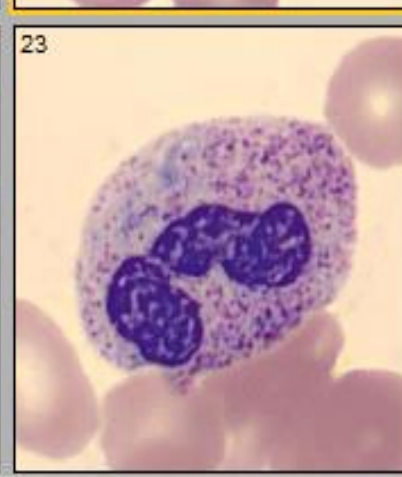
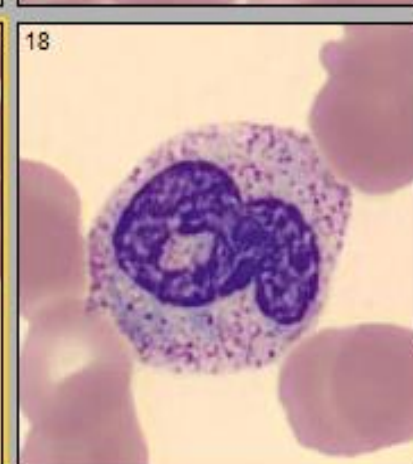
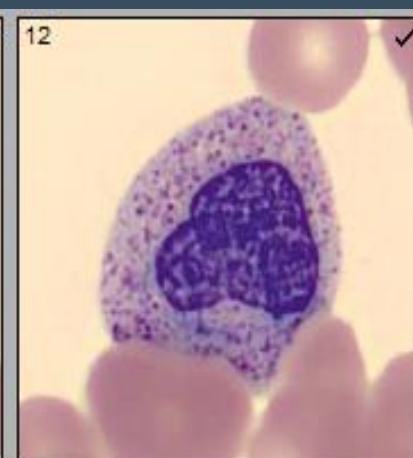
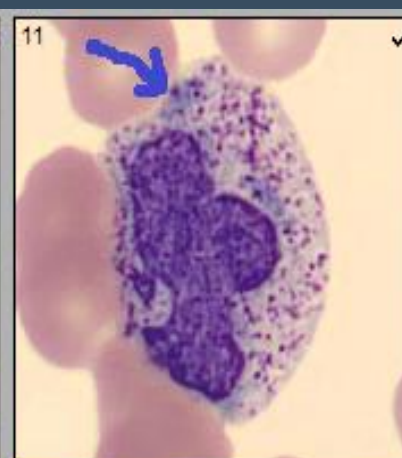
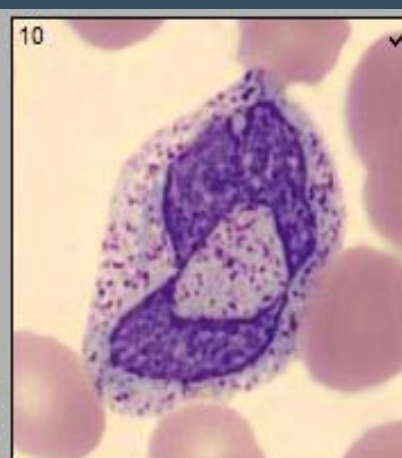
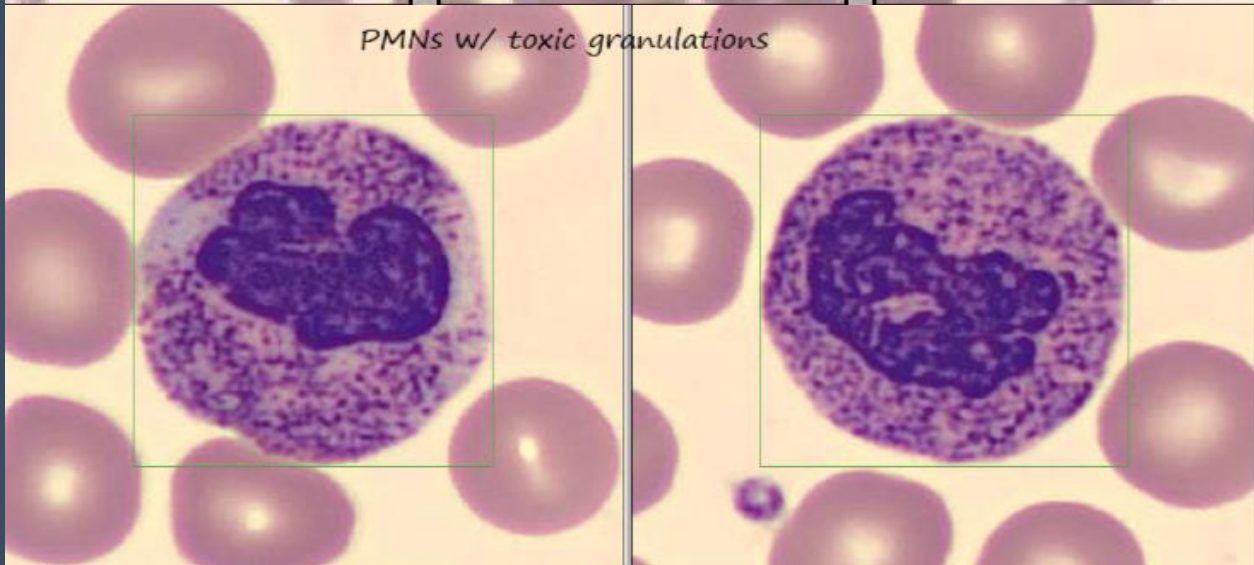
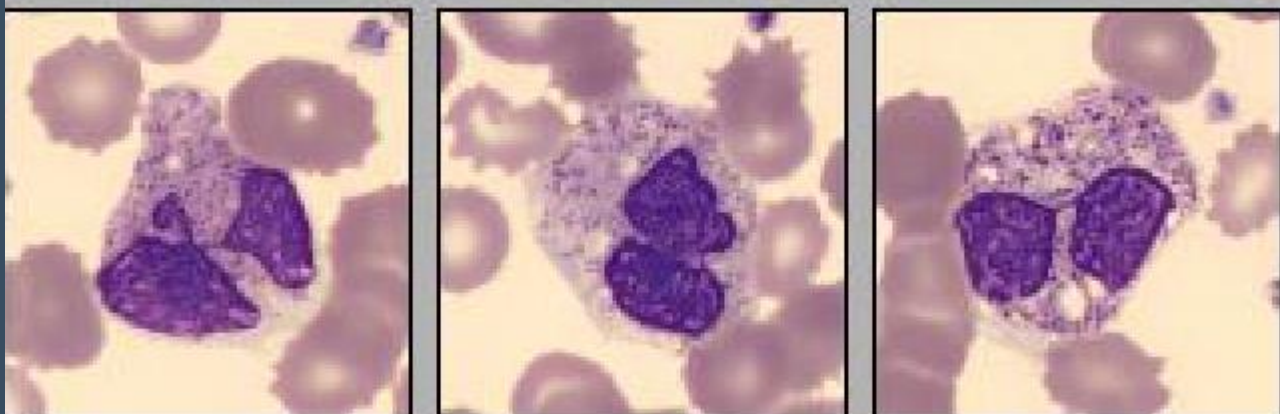




# Neutrophils

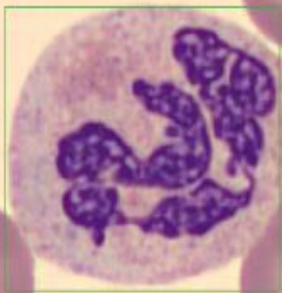
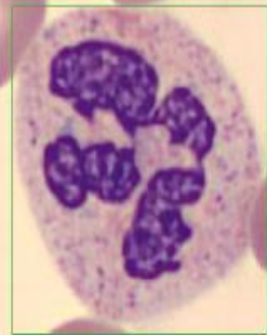




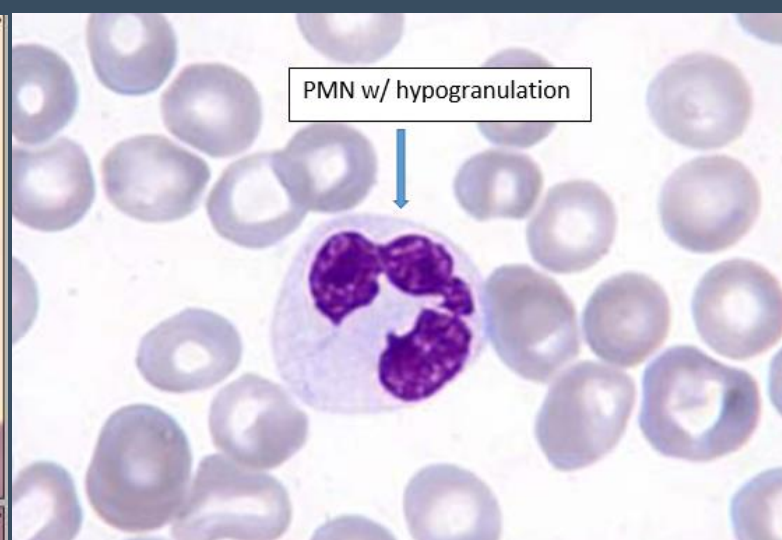




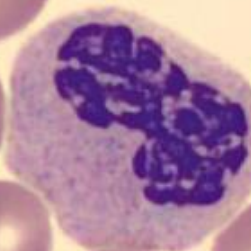
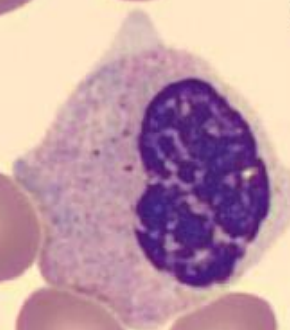
Hypersegmented PMNs



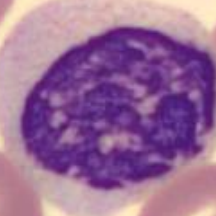
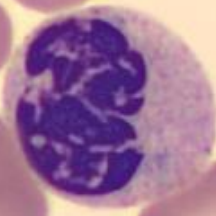
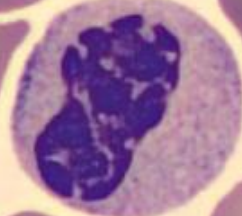
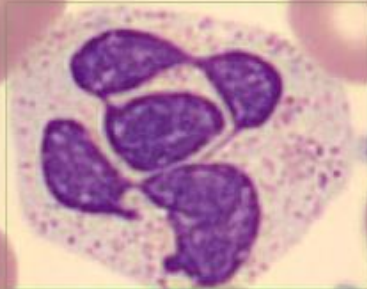
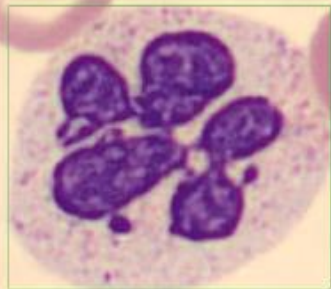
PMN w/ hypogranulation



Dysplastic PMNs



Hypersegmented PMNs

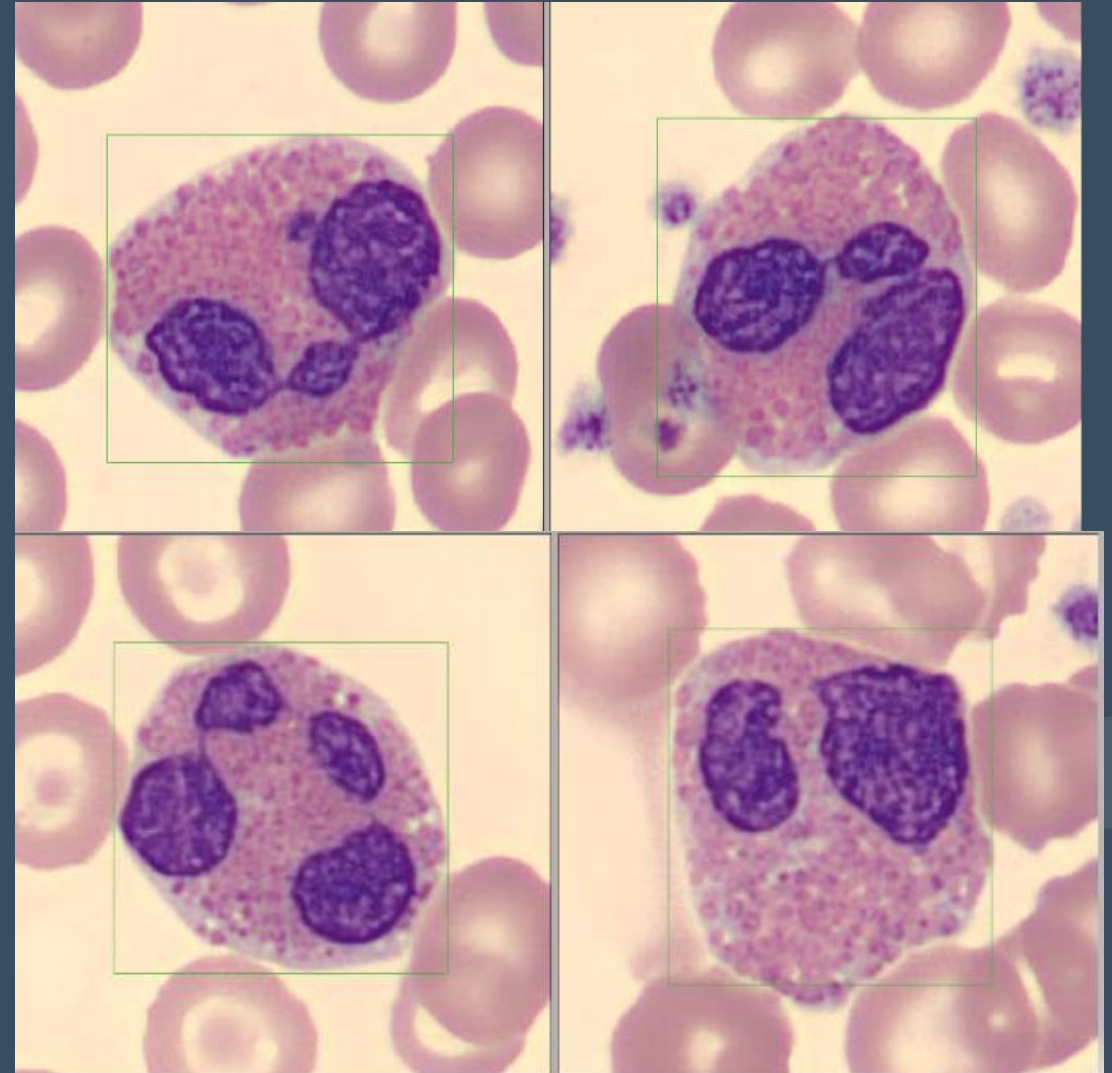


# Eosinophils

- Arise from common myeloid progenitor (CMP)
- Charcot leyden crystal protein in the primary granules
- Eosinophil myelocyte is the first maturation phase identified by light microscopy
- Large, reddish -orange granules in the cytoplasm which are more intensely colored
- Mature eosinophils display a bi-lobed nucleus
- An increased production in infection by parasitic helminths and allergic disorders such as asthma

# Eosinophils

- Normal values
  - Relative or percentage- 1-3%
  - Absolute – 0-0.3 k/uL
- Eosinophilia – absolute count  $> 0.3/\mu\text{L}$  of blood
- Eosinopenia – decrease in eosinophil count



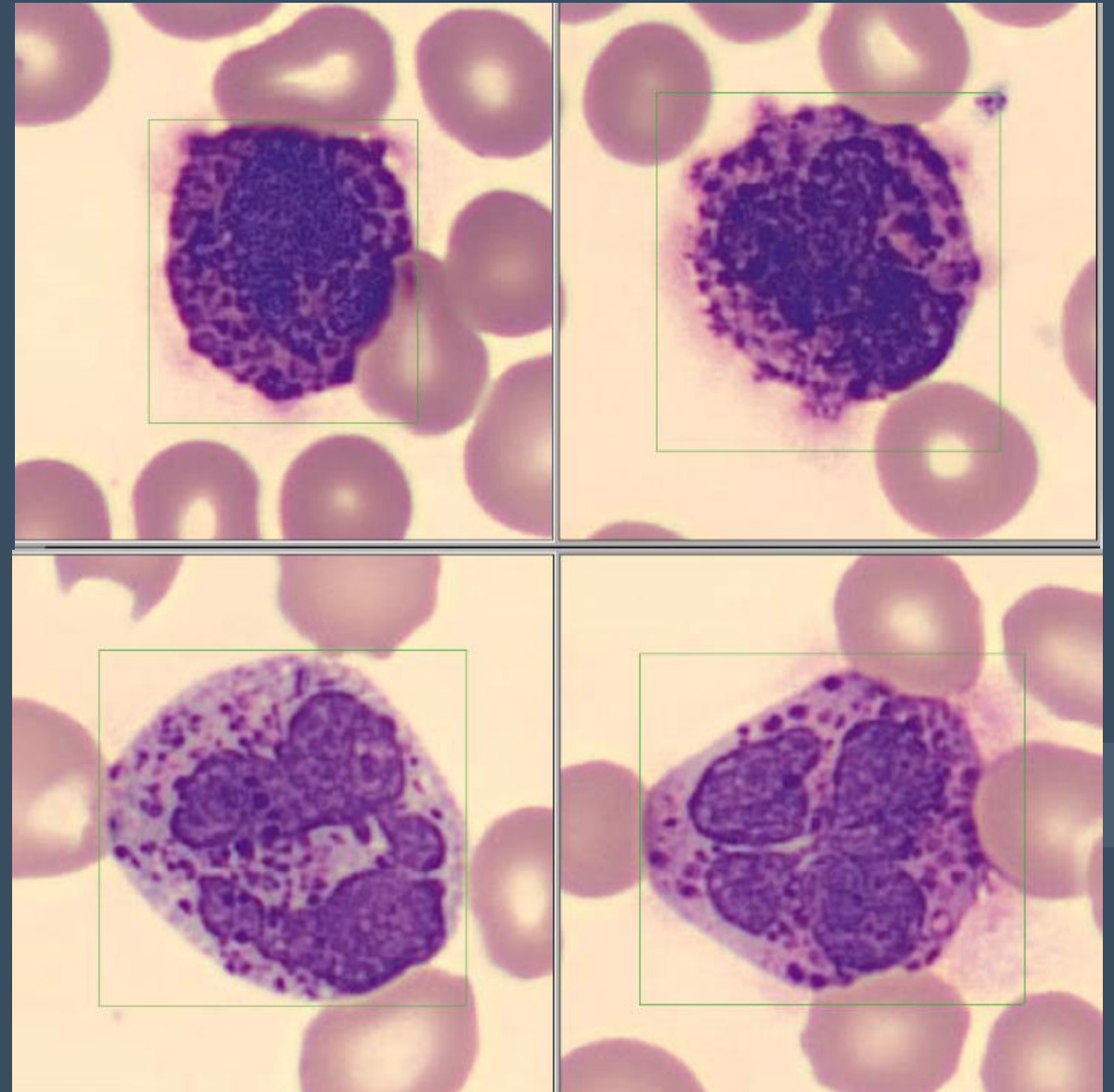


# Basophils

- Mature in the marrow and circulate in the blood as mature cells with granules
- Mast cell precursors leave the bone marrow and mature in the tissues
- Immature or mature basophils
- The chromatin is clumped and nucleus is often lobulated and obscured by its granules
- Cytoplasm contain large blue-black granules
- Granules are water soluble and may be dissolved during staining process

# Basophils

- Histamine and heparin granules
- Normal values
  - Relative or percentage- 0-2 %
  - Absolute – 0-0.2 k/uL
- Basophilia – absolute count  $> 0.2$  /ul of blood
- Basopenia – decrease in basophil count



# Monoblast

- Derived from granulocyte monocyte progenitor (GMP)
- Macrophage colony stimulating factor (M-CSF) is the major cytokine responsible for growth and differentiation
- The development stages  
Monoblast>Promonocyte>Monocyte
- Differentiation of a Monoblast from myeloblast is almost impossible on morphology alone



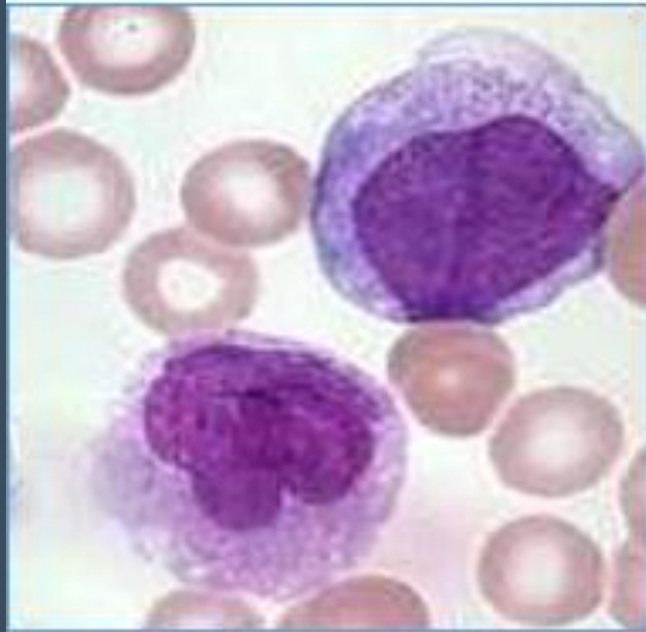
# Promonocyte

- Nucleus is slightly indented or folded
- Delicate chromatin pattern and nucleolus may be apparent
- The cytoplasm is blue and contains scattered azure granules which give the grainy appearance
  - Lysosomal enzymes, peroxidase, non-specific esterases and lysozyme
  - Electron microscope and cytochemical stain can be helpful identifying azure granules
- Identification of monoblasts and promonocytes are achievable when marked proliferation of monocytic cells present

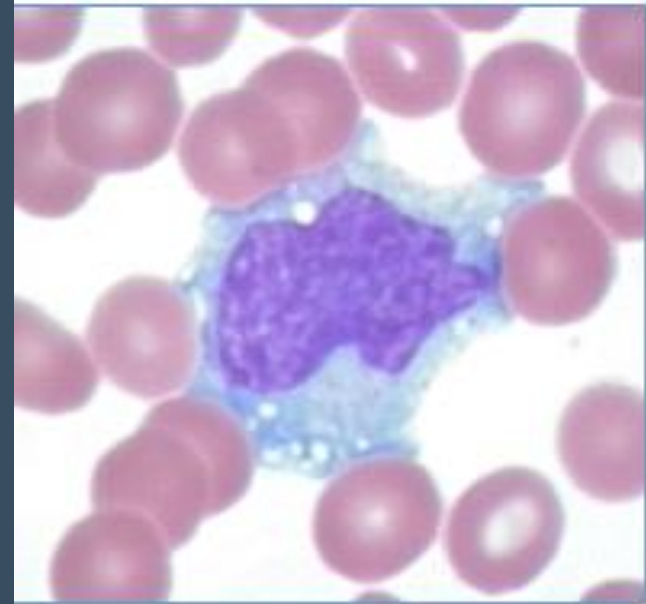
# Monocytes



Monoblast



Promonocyte



Monocyte

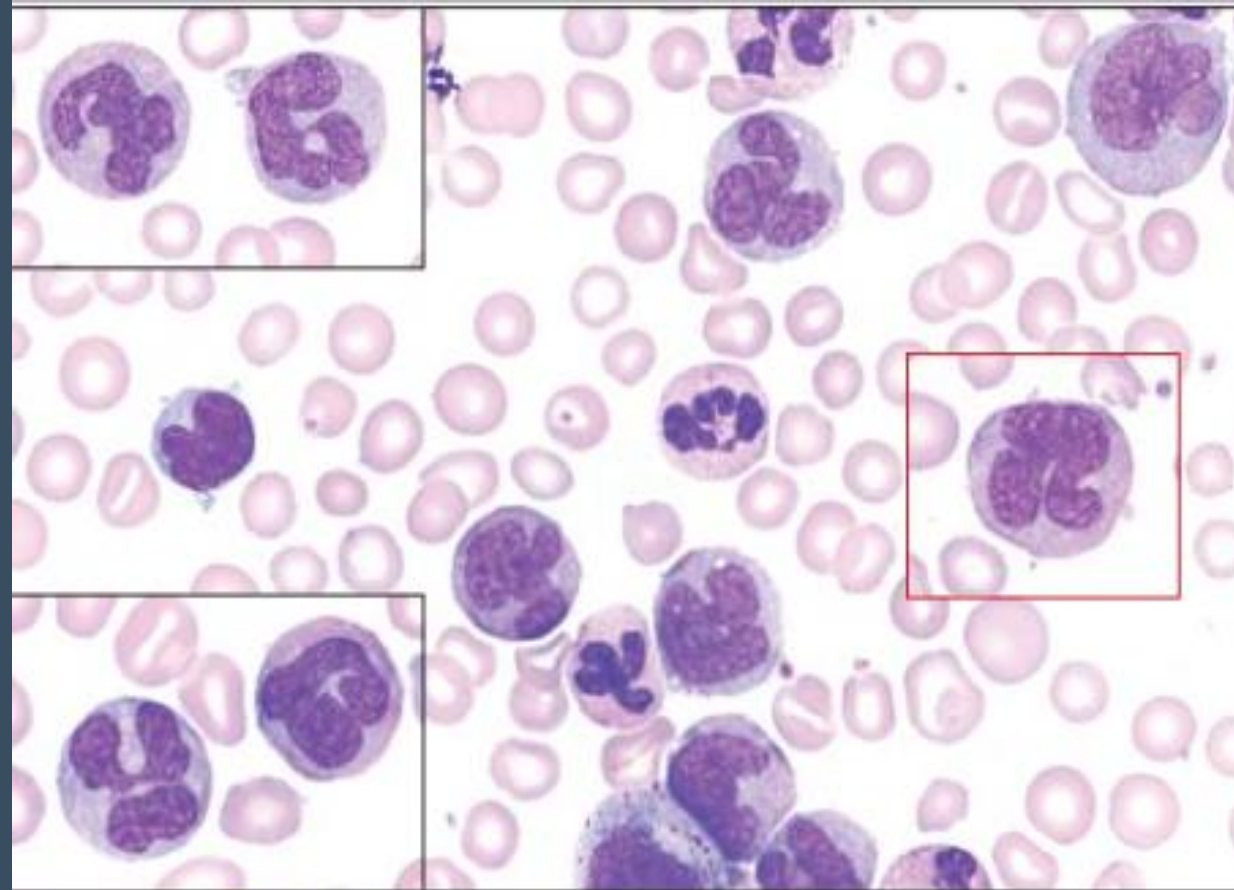


# Monocyte

- Slightly immature cells ultimately mature into macrophages, osteoclasts, or dendritic cells
- Nucleus is kidney shaped or deeply indented horseshoe shaped
- The chromatin pattern is looser in compare to other WBCs
- The cytoplasm is blue-gray with fine azure granules
- Cytoplasmic and nuclear vacuoles may be present
- No storage pool of mature monocytes in bone marrow

# Monocyte

- Relative monocytosis after bone marrow recover from marrow failure
- Normal values
  - Relative or percentage- 2-11 %
  - Absolute – 0.1-1.3 k/uL
- Monocytosis
  - absolute count  $> 1.3/\mu\text{L}$  of blood
- Monopenia
  - decrease in monocyte count



Courtesy of Olga Pozdnyakova, MD, PhD.

# Lymphocytes

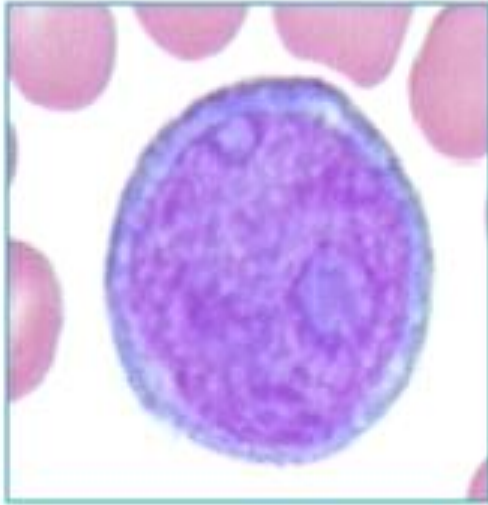
- Three major groups
  - T cells, B cells and natural killer (NK) cells
- Humoral immunity by producing antibodies
  - B cells
  - ~ 3% to 21% of circulating lymphocytes
- Hematogones have homogeneous nuclear chromatin pattern and extremely scanty cytoplasm
- Hematogones are seen in newborn peripheral blood, bone marrow and regenerative bone marrows

# Lymphocytes- T cells and NK cells

- Cellular Immunity
- T cells
  - Comprise 51% to 88% of circulating lymphocytes
- Medium and large lymphocytes referred to as reactive or variant lymphocytes
- NK cells are large because of increased amount of cytoplasm and contains azurophilic granules
  - ~ 4%- 29% of circulating lymphocytes

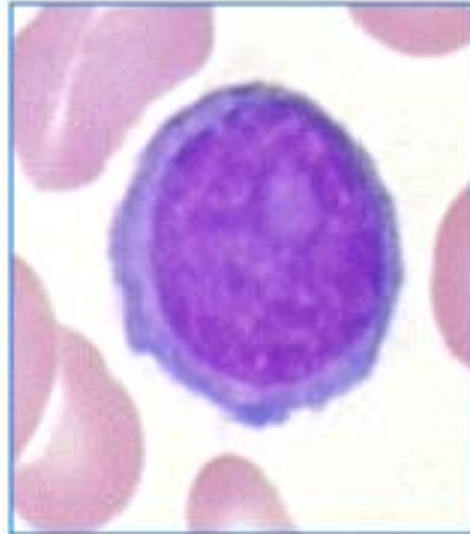


# Lymphocytes

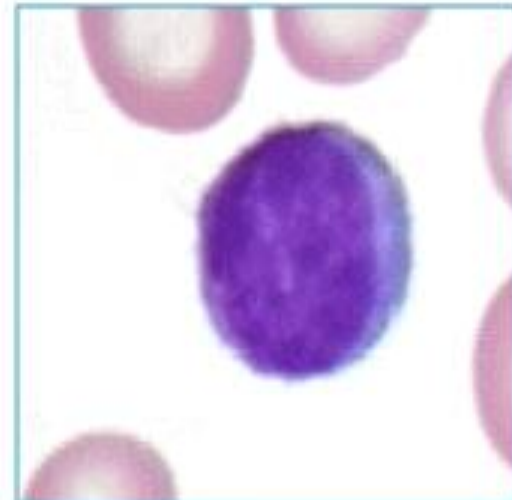


Lymphoblast

Prolymphocyte



Lymphocyte





# Lymphoblast

- Large round nucleus with one distinct nucleolus or two nucleoli
- Cytoplasm is blue non granular, may be moderate or scanty in amount
- Differentiation of cells in the lymphocytic maturation sequence based on nuclear structure



# Prolymphocyte

- Either T cell or B cell origin
- The chromatin is intermediate between lymphoblasts and mature lymphocytes
- Single prominent centrally located distinct nucleoli
- Moderately abundant pale blue cytoplasm lacks granules and vacuoles
- An increased prolymphocytes in prolymphocytic leukemia

# Lymphocyte

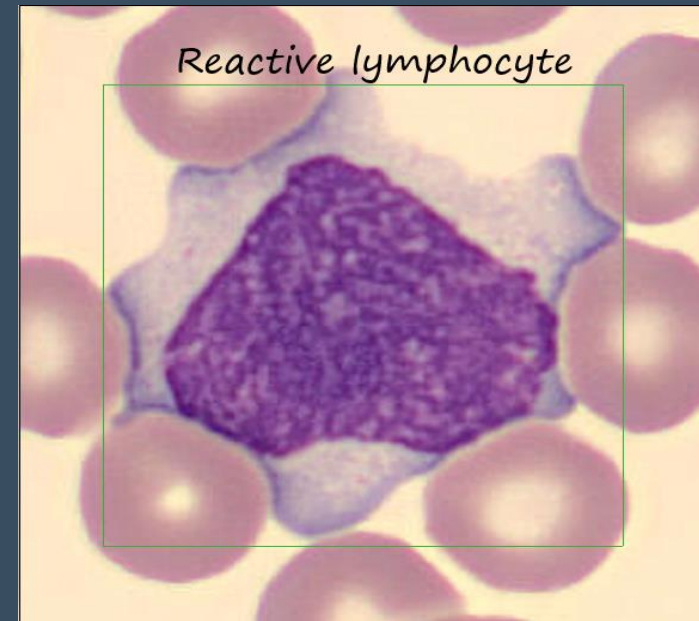
- Lymphocytes are variable in size, condensed and clumped chromatin
- Large nucleus in relation to the cytoplasm occupies ~ 90% of the cell area
- The size of the nucleus of small lymphocytes is comparable to the size of normal red blood cells
- Normal values
  - Relative or percentage- 18-42 %
  - Absolute – 1.0-3.2 k/uL
- Lymphocytosis – absolute count  $> 3.2/\text{ul}$  of blood
- Lymphopenia – absolute count  $< 1.0/\text{ul}$  of blood

# Reactive Lymphocyte

- Morphologic characteristics different from normal lymphocyte appearance
- Demonstrate a wide variety of morphologic features (nucleus and amount and color of cytoplasm)
- Often large with moderate to abundant dark blue cytoplasm
- An increased amount of RNA, which is reflected by associated increase in cytoplasmic basophilia.

# Reactive Lymphocyte

- Nuclear chromatin is mature clumped and no nucleoli
- Most often associated with viral illnesses
- An increased number is a morphologic hallmark of infectious mononucleosis.
- Indented cytoplasmic margins by surrounding red blood cells and appear a darker blue than the rest of the cytoplasm.

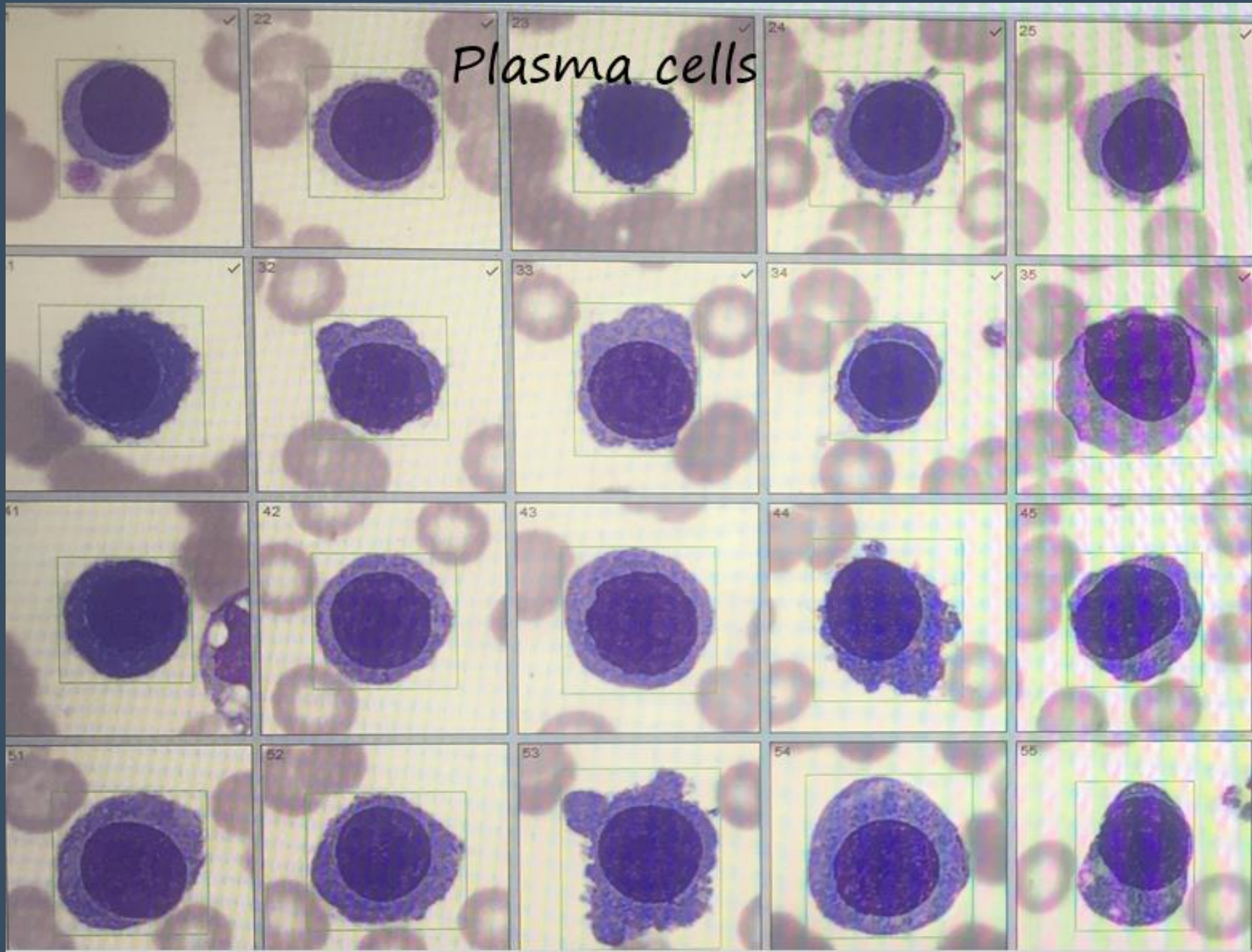




# Plasma cell

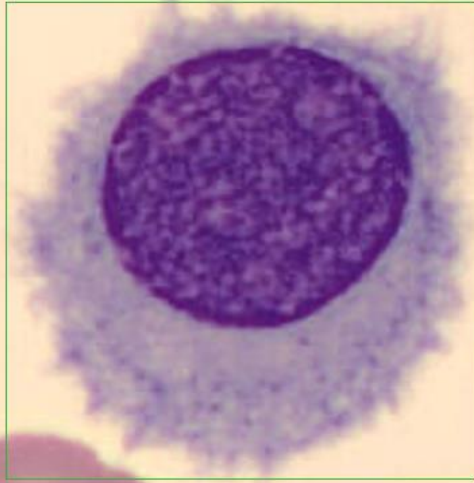
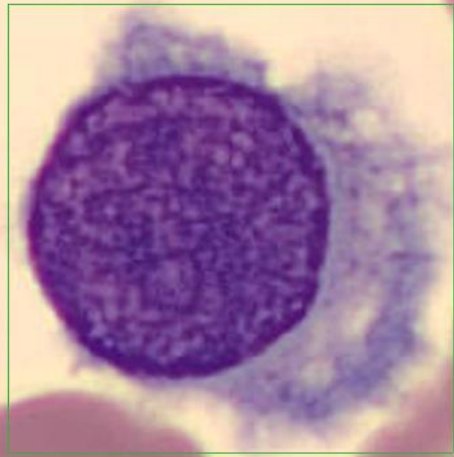
- The descendant of B lymphocytes
- Nucleus is relatively small, round or oval and eccentrically placed with dense chromatin
- Moderate amount of dark blue cytoplasm
- A prominent hof or paranuclear clear zone containing the Golgi apparatus
- Plasma cells produce immunoglobulin
- Russell bodies, flame cells and crystalize materials

Plasma cells

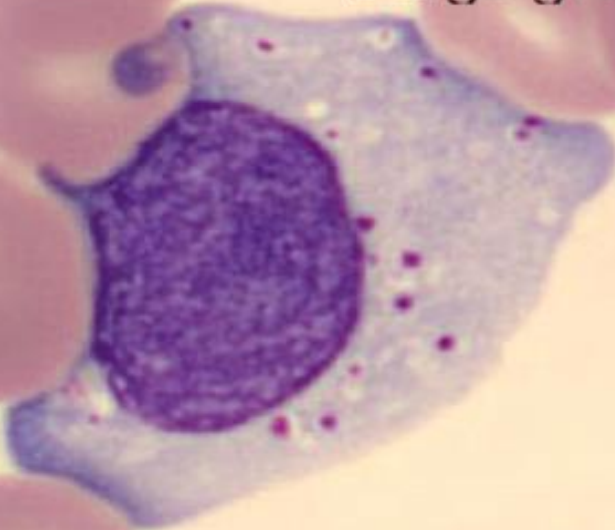




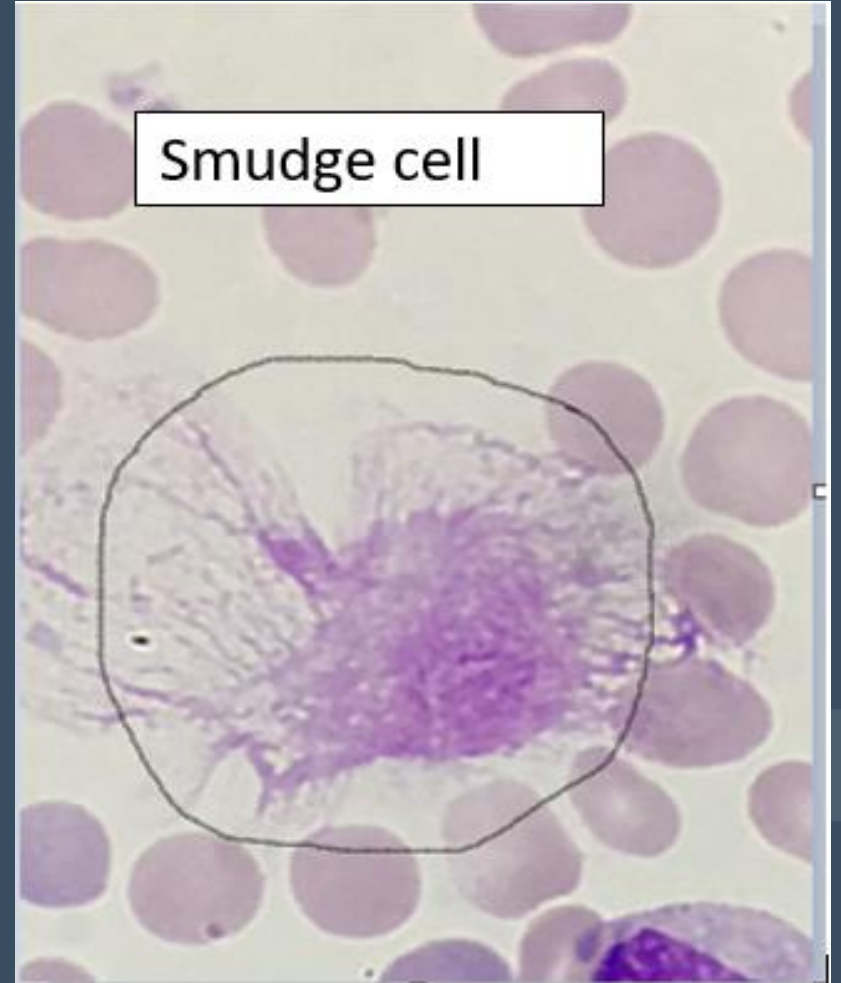
Hairy cells



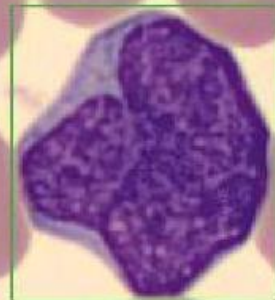
Large granular lymphocytes



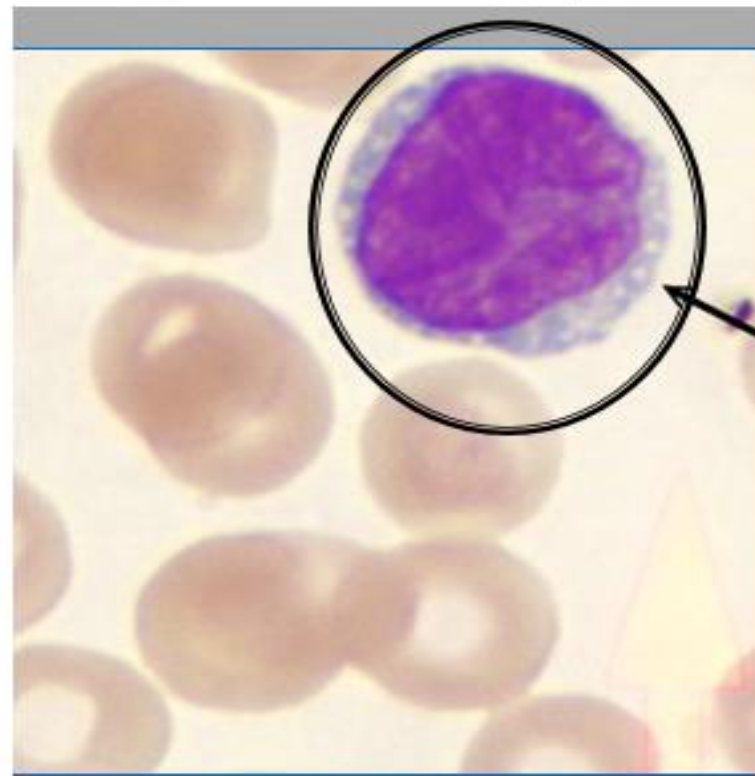
Smudge cell



Lymphoma cells



Sezary cell



Thank You!

