

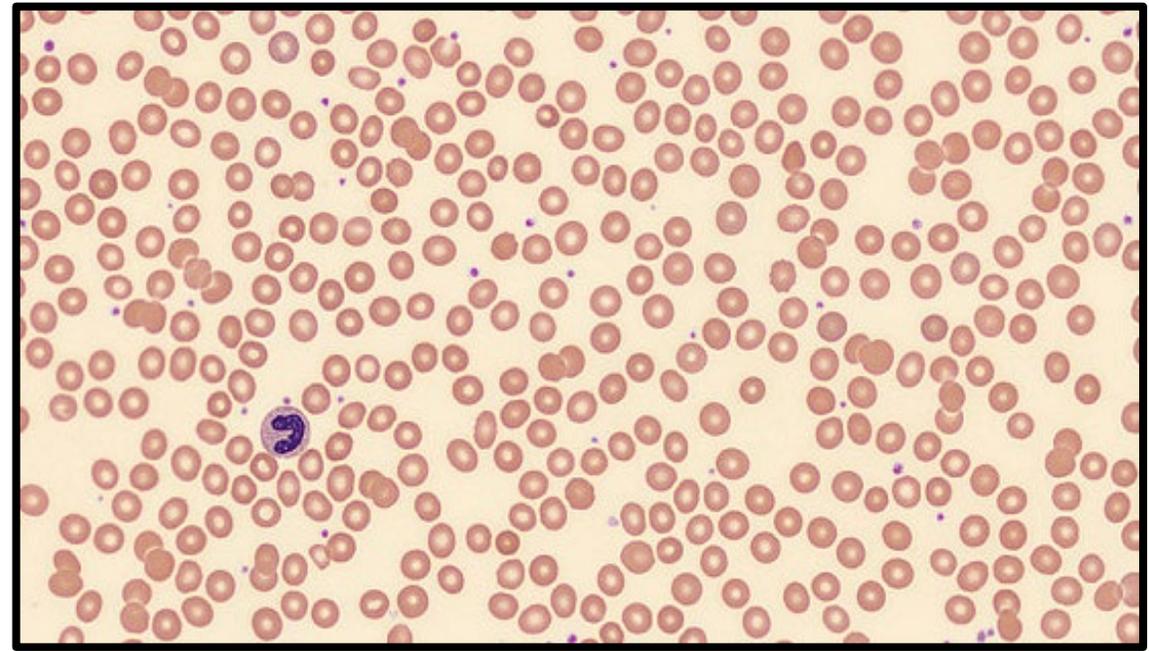
Peripheral Blood Smear Morphology

Afshan Idrees, MD, MBA, MPH

Assistant Professor Pathology

University of Miami-Miller School of Medicine

Medical Director of Hematology & Coagulation
Laboratories at UHealth



Conflict of Interest Disclosure

None

Learning Objectives

- Identify normal blood cells
- Recognize variations in morphology in normal as well as non-neoplastic disease states

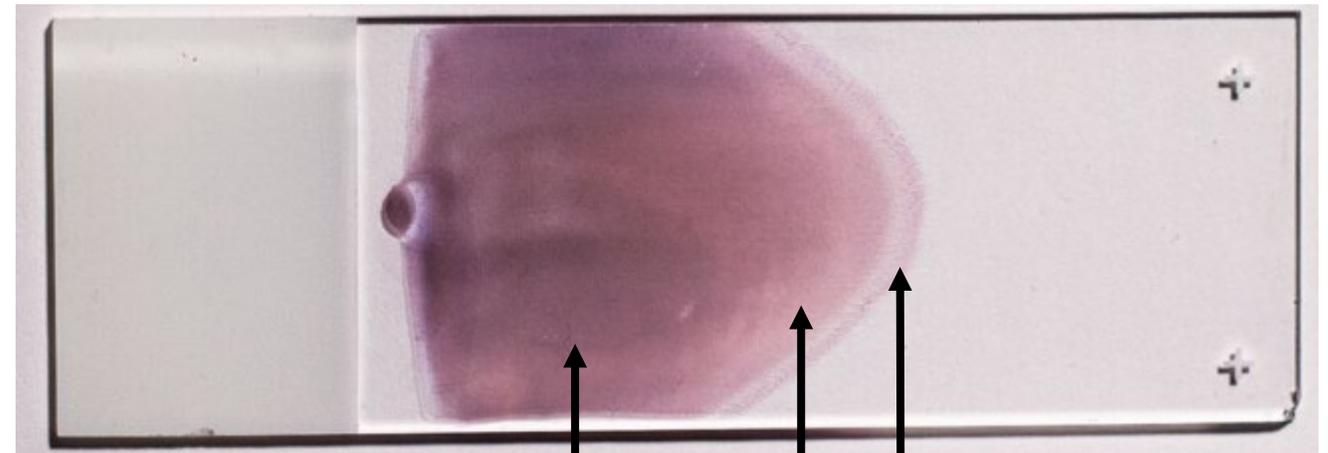
Peripheral Blood Smear

- CBC with differential, a common and routinely ordered lab test
- Minimally invasive
- Sample collected in a lavender top (EDTA) tube
- High yield data
- Used for screening and basic health assessment
- Peripheral smear examined via two common pathways:
 - a. hematology analyzer flag
 - b. physician request
- Catalyst for a bone marrow examination



Review of the Blood smear

- Scan at low magnification
- Assess the quality and quantity of WBCs, platelets and RBCs
- Don't forget to focus on the feather edge and sides
- Large cells, blasts and platelet clumps are often dragged to these areas

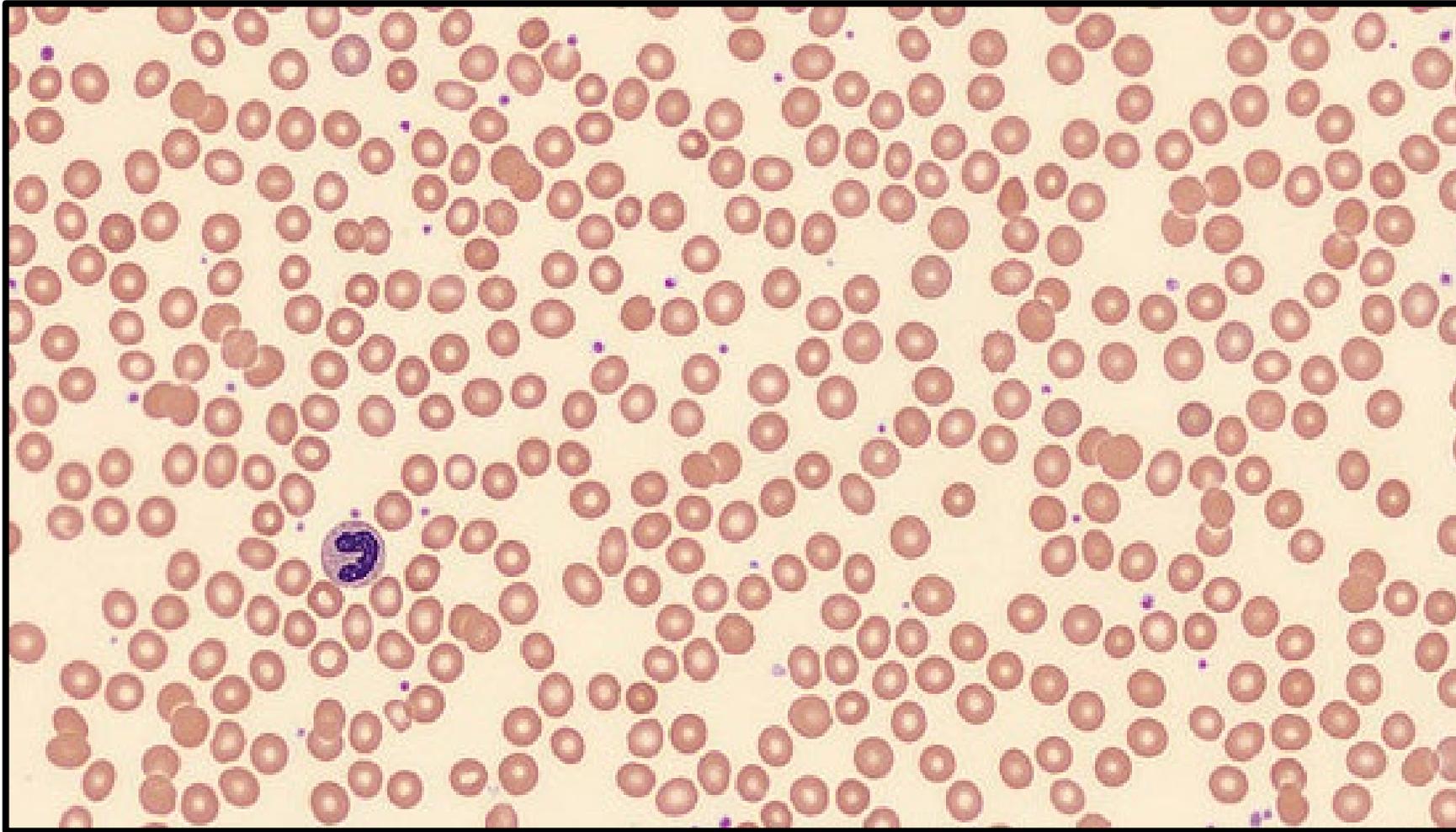


Thick layer

Mono-layer

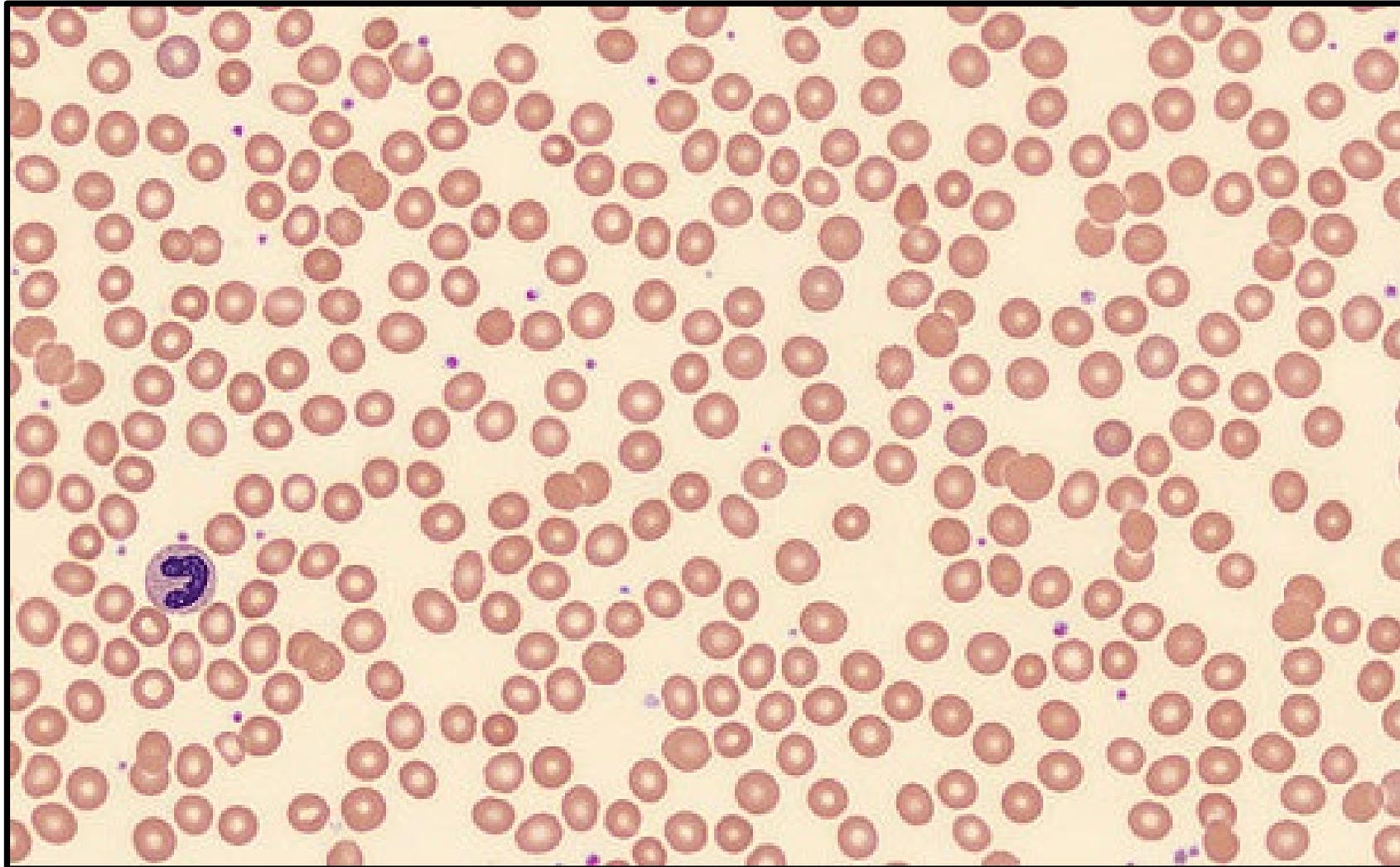
Feather edge

Normal Blood Smear



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

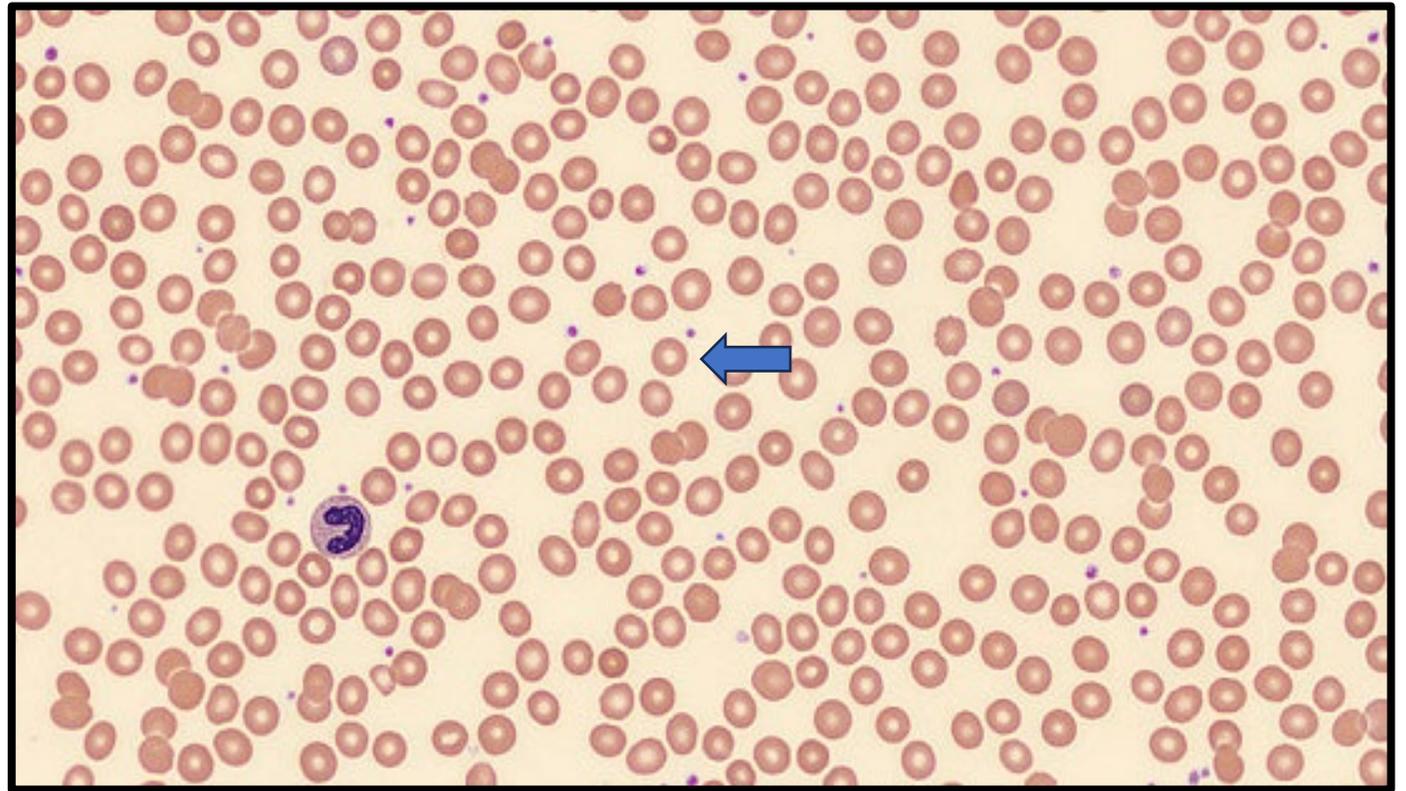
Red Blood Cells



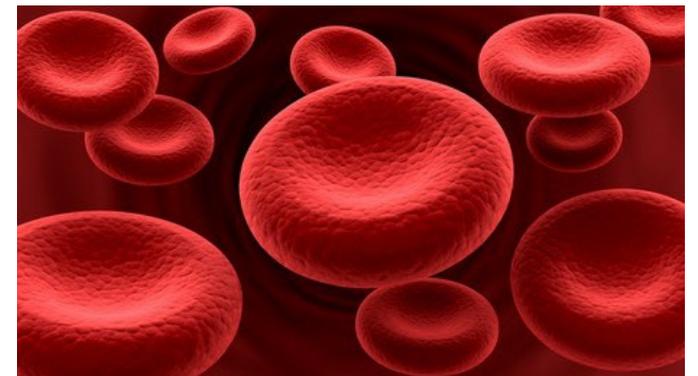
Source: Dr. Afshan Idrees, University of Miami Department of
Pathology & Laboratory Medicine

Red Blood Cells

- Circular disks, 6-8 um diameter
- Non-nucleated
- Appear pinkish with a central pale area
- Depth of staining provides a rough guide to hemoglobin content
- Terms, “Normochromic, hypochromic and hyperchromic” used to describe this feature
- Shape, size and hemoglobin content varies with disease states
- Diameter is not the same as volume
- Terms, “macrocytic, microcytic and normocytic” refer to MCV not the diameter
- Anisocytosis and poikilocytosis



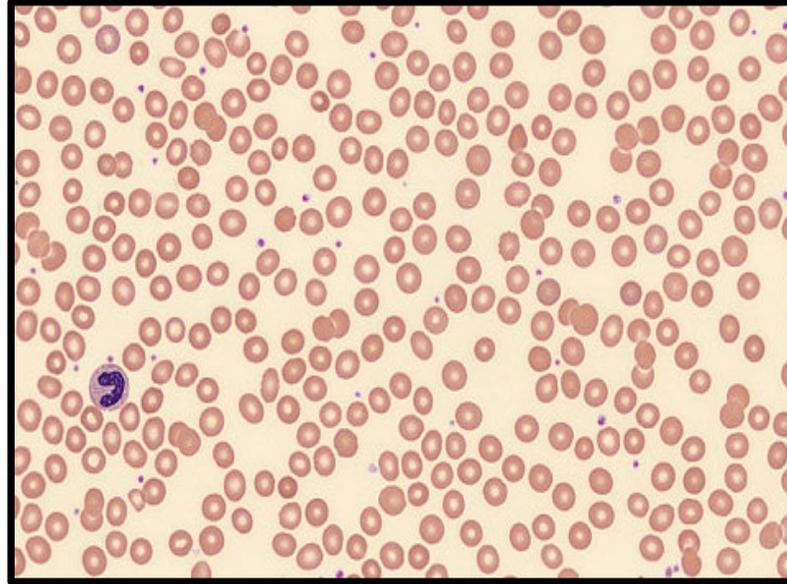
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine



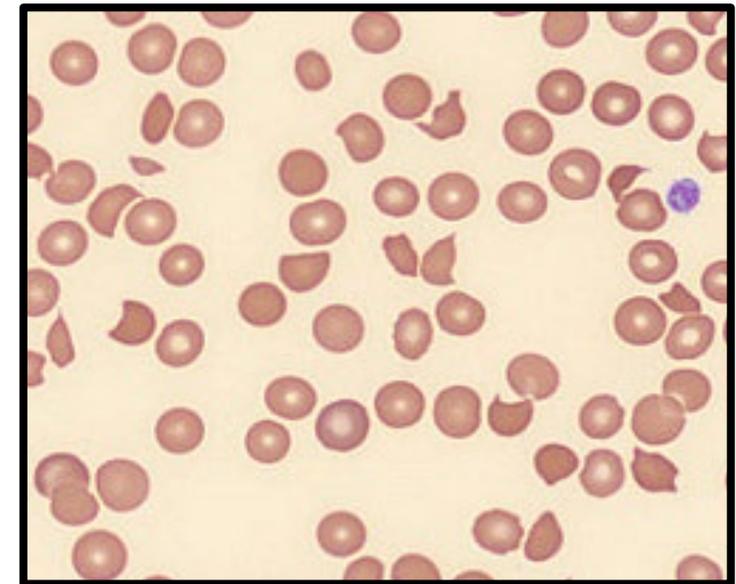
Red cell quantity



Erythrocytosis



Normal



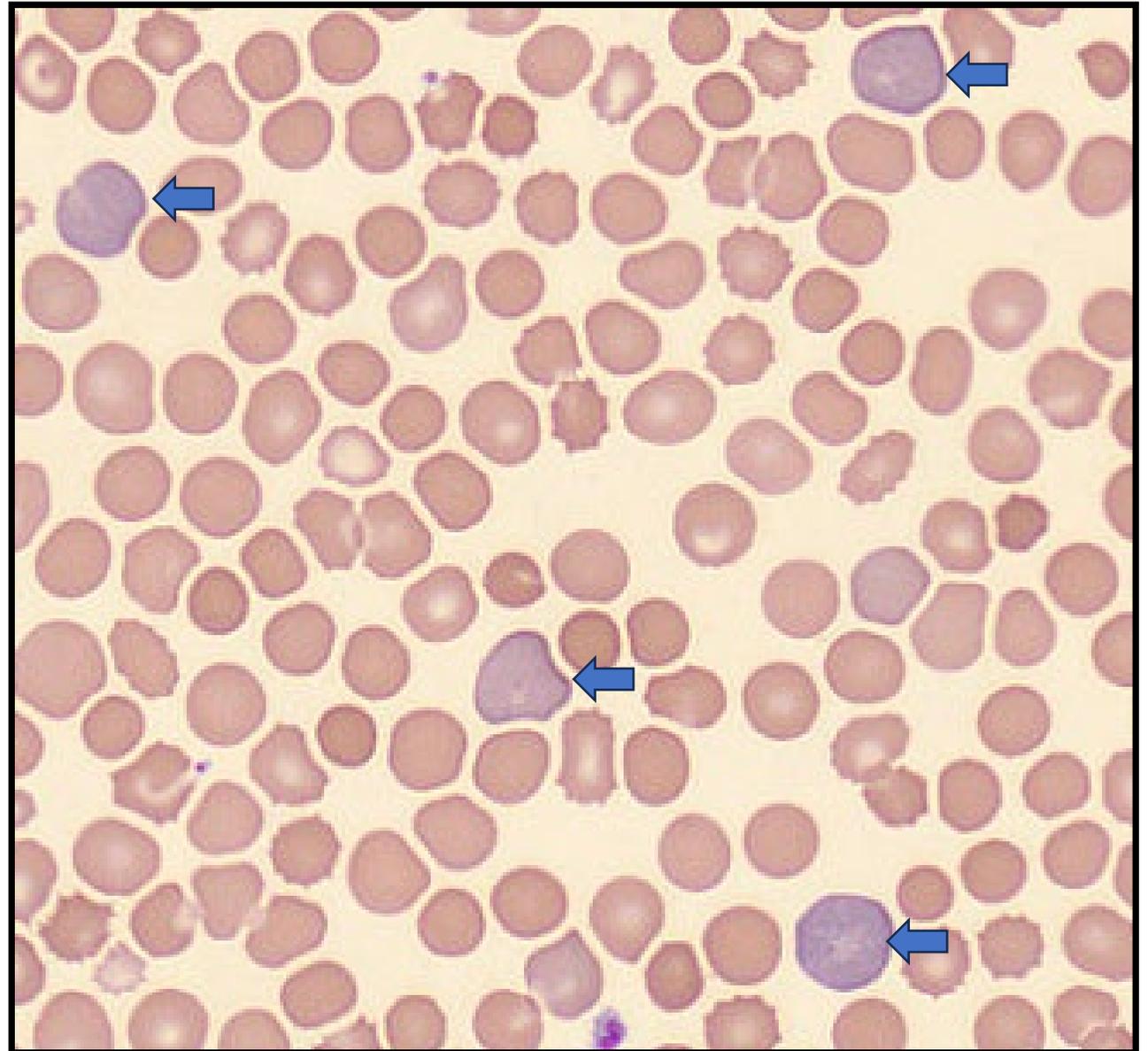
Anemia

Red cell quality

- Variation in size & shape
- Red cell inclusions
- Red cell distribution
- Artifact

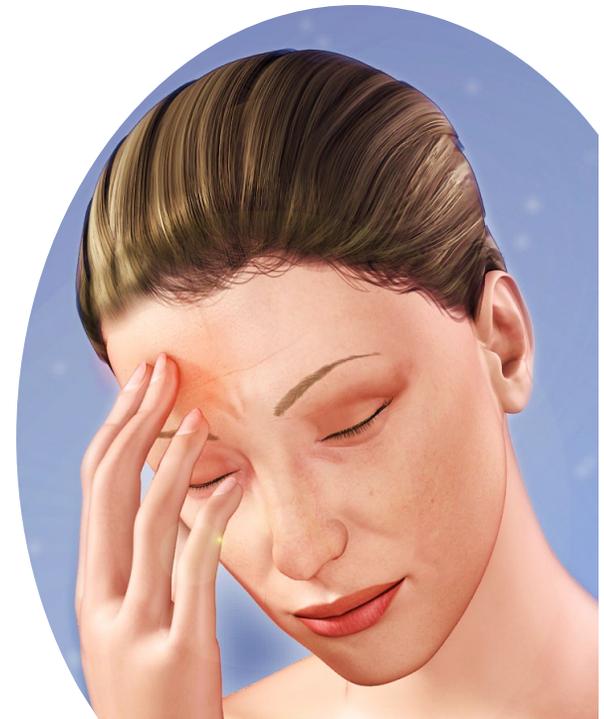
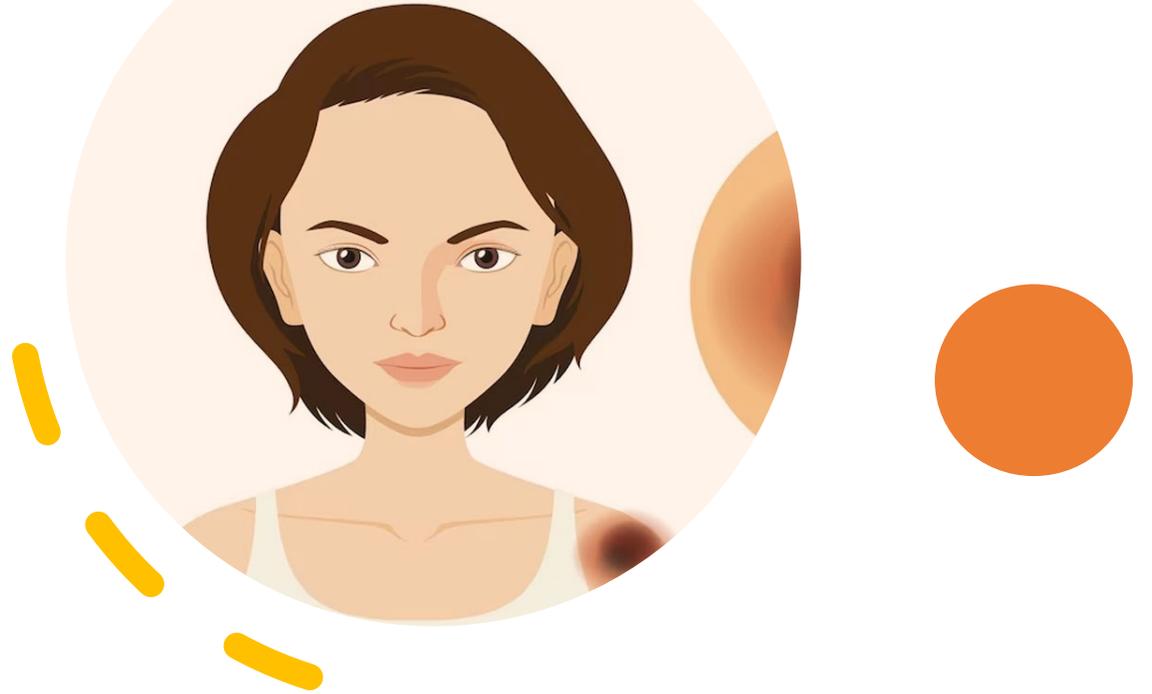
Polychromasia

- Polychromatophilic cells (Wright's stain) are young non-nucleated red cells
- Combination of the affinity of Hb for acid stains and RNA for basic stains
- They lack central pallor, appear larger and slightly basophilic due to RNA
- They lose their RNA in a day or so after reaching the blood from the marrow
- They provide an assessment of the rate of red cell production
- Also called reticulocytes (Cresyl blue stain)
- Seen in hemolysis



Case

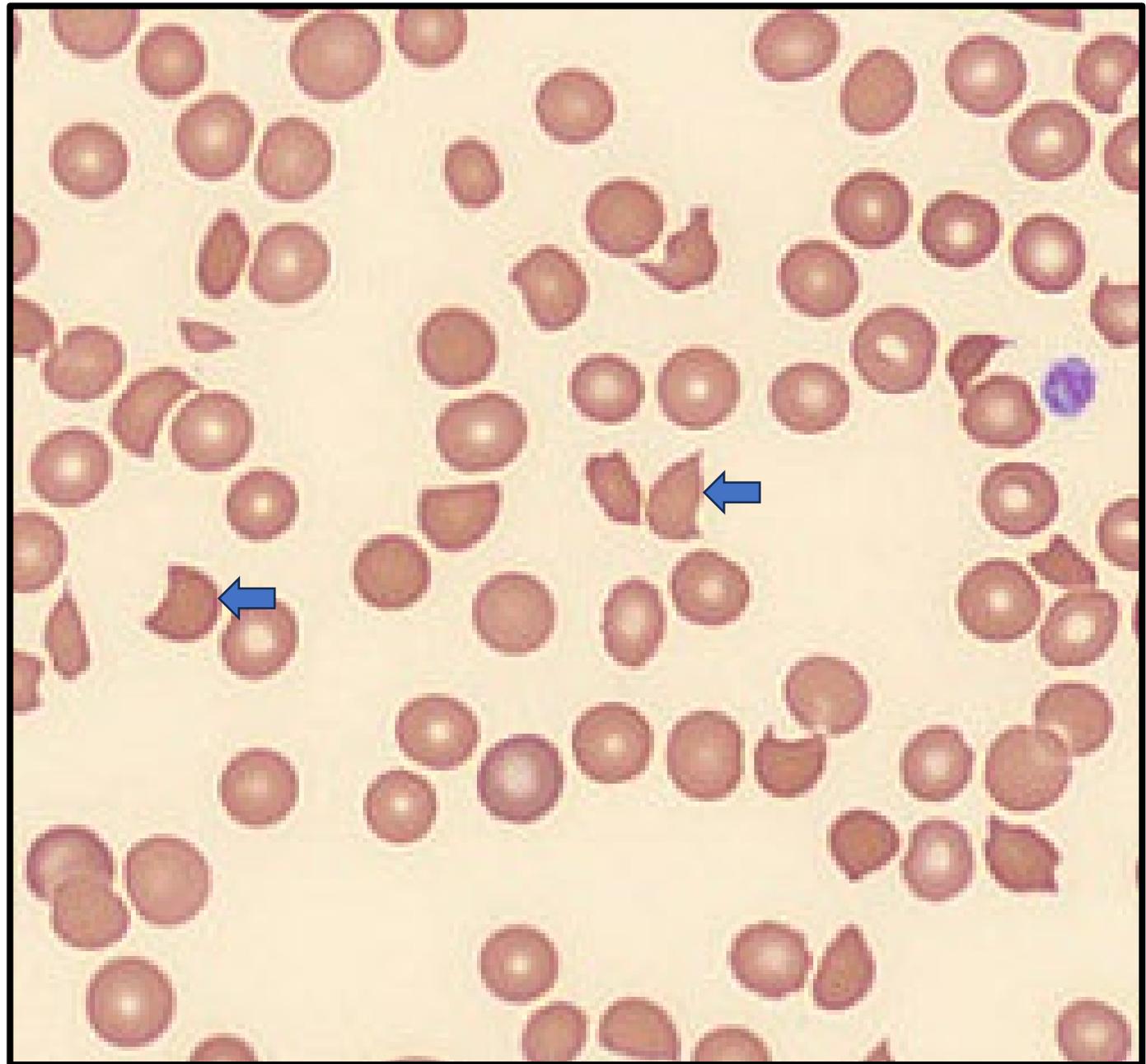
- 53yr/F with diabetes mellitus presented to the urgent care with an acute onset easy bruising and fatigue for the last 3 days
- No prior history of bleeding
- Physical exam: purpura and bruises on all extremities



Lab results

	Result	Reference range
• Hb	8 ↓	12.5-15 g/dL
• RBC	4	3.5-5.5 x10 ⁶ /uL
• MCV	88	80-95 fL
• Retic	10 ↑	0.5-2 %
• WBC	8.4	4-11 x 10 ³ /uL
• Platelets	18 ↓	150-450 x 10 ³ /uL
• LDH	2,000 ↑	135-225 units/L
• T bilirubin	3 ↑	0-1 mg/dL
• Haptoglobin	<10 ↓	30-200 mg/dL
• DAT	Negative	Negative
• ADAMTS13 activity	6 ↓	56-243%

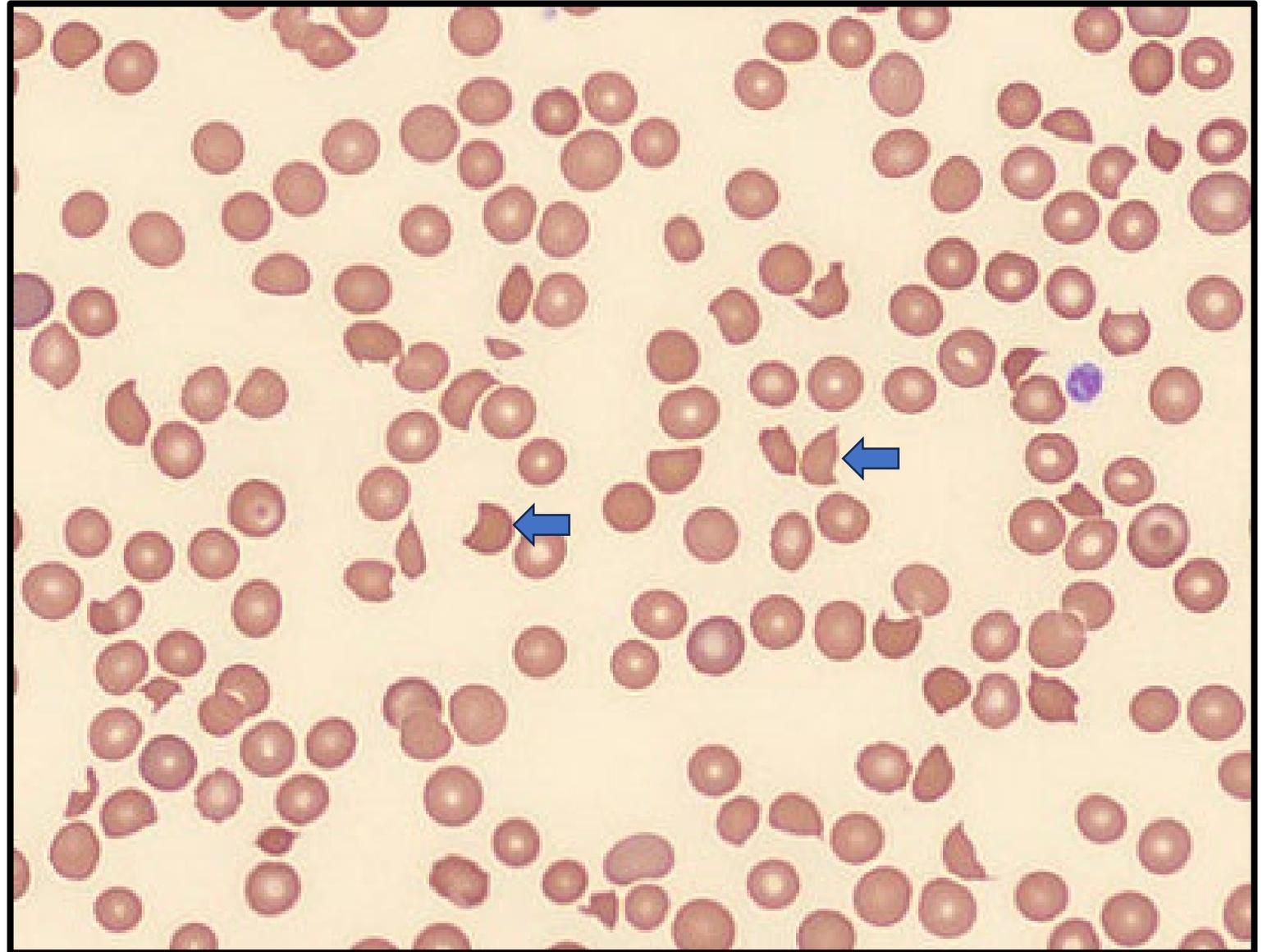
Peripheral Blood Smear



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Schistocytes

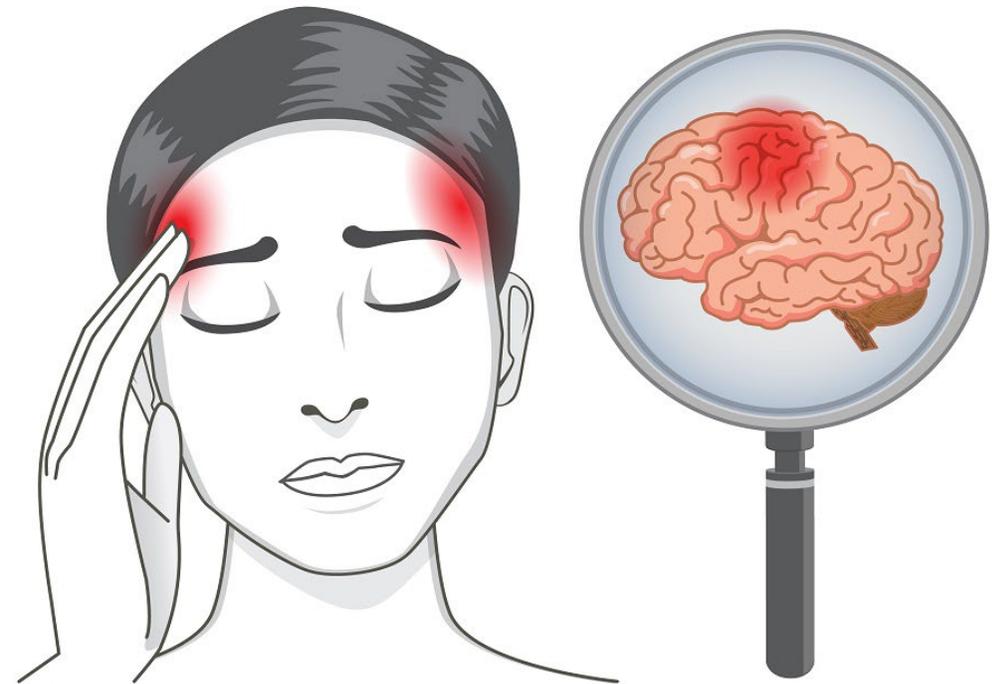
- Fragmented RBCs, helmet cells
- Indicate hemolysis
- Seen in small blood vessel disease or presence of fibrin, e.g., microangiopathic hemolytic anemia
- Other causes: burns, DIC and sepsis



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Case

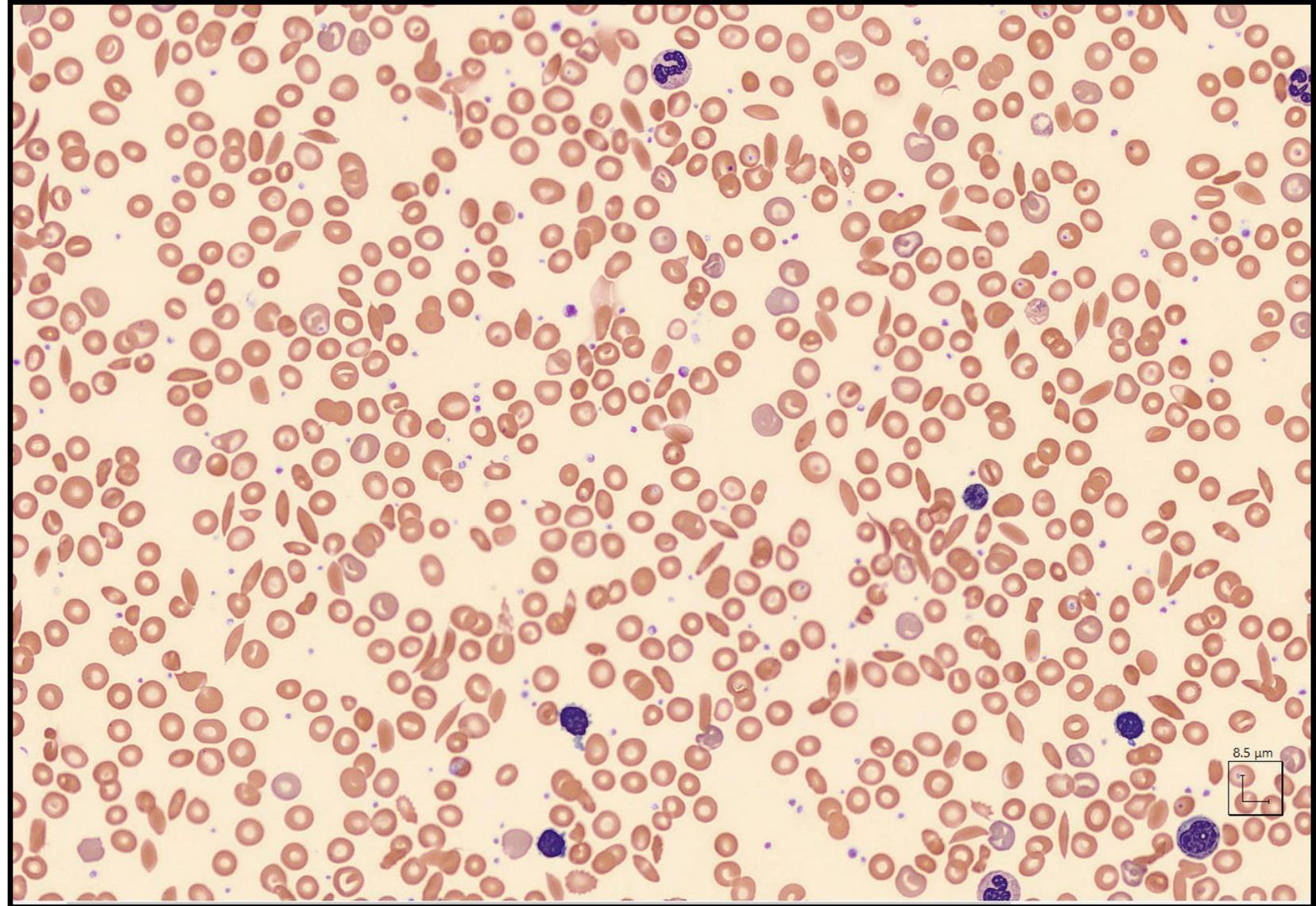
- 6 yr/F with history of hemoglobinopathy
- Transient episode of headache and hemiparesis
- CT scan unremarkable



Lab results

	Result	Reference Range
• Hb	7.9 ↓	12.5-15 g/dL
• RBC	2.5 ↓	3.5-5.5 x10 ⁶ /uL
• MCV	96	80-98 fL
• Retics	19 ↑	0.5-2 %
• WBC	21.5 ↑	4-11 x 10 ³ /uL
• Platelets	420	150-450 x 10 ³ /uL

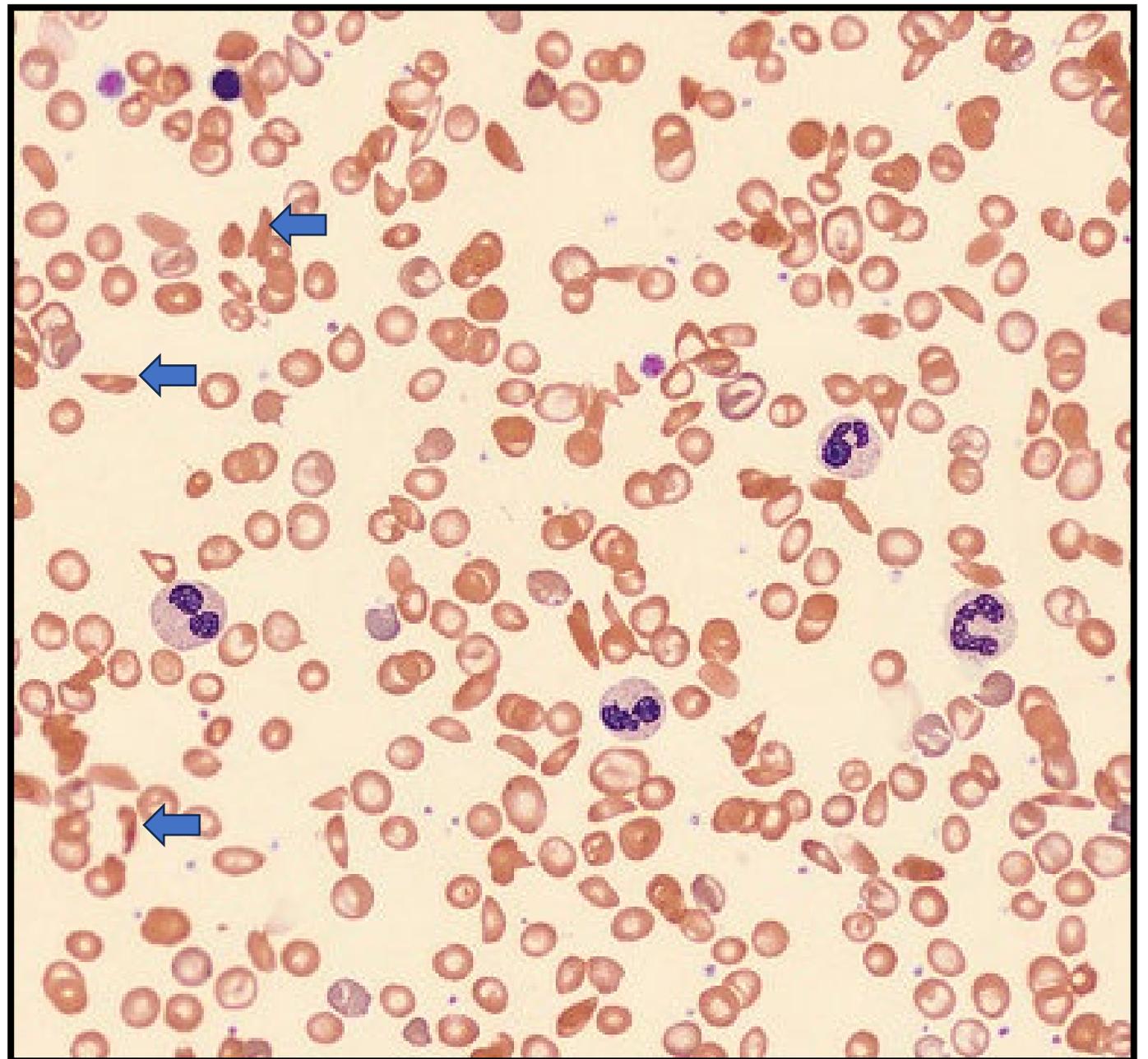
Peripheral Blood Smear



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Sickle cells

- Curved or sickle shaped RBCs
- Seen in sickling disorders
- Sickle cell disease
- Hb SC disease
- Sickle cell with beta thalassemia



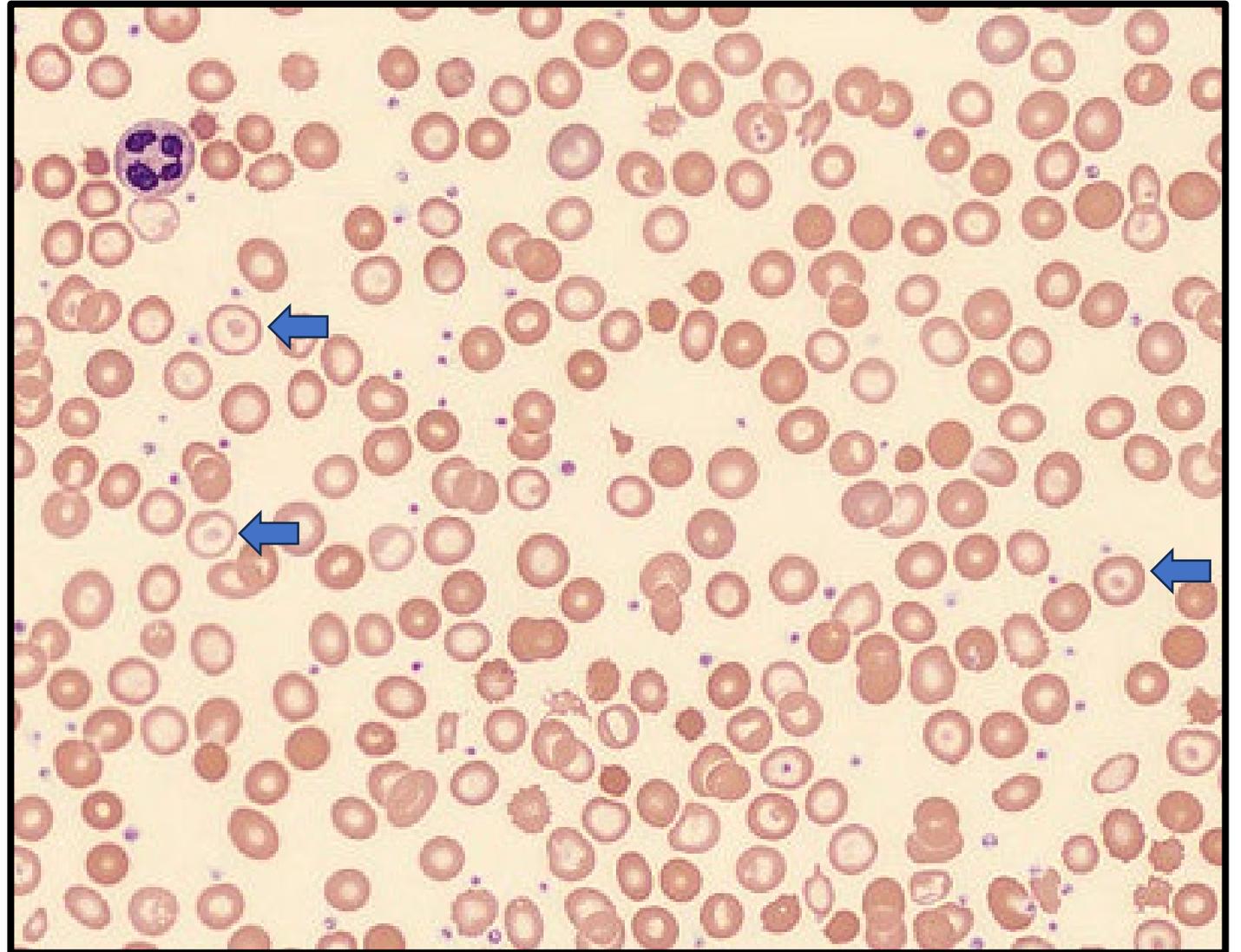
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Target cells

- Target cells are RBCs with a thin excess membrane, peripheral rim of hemoglobin, and dark central hemoglobin containing area
- Frequently seen in thalassemia, HbC disease, liver disease and sickle cell anemia, post-splenectomy
- Lack of normal reduction of surface membrane with aging

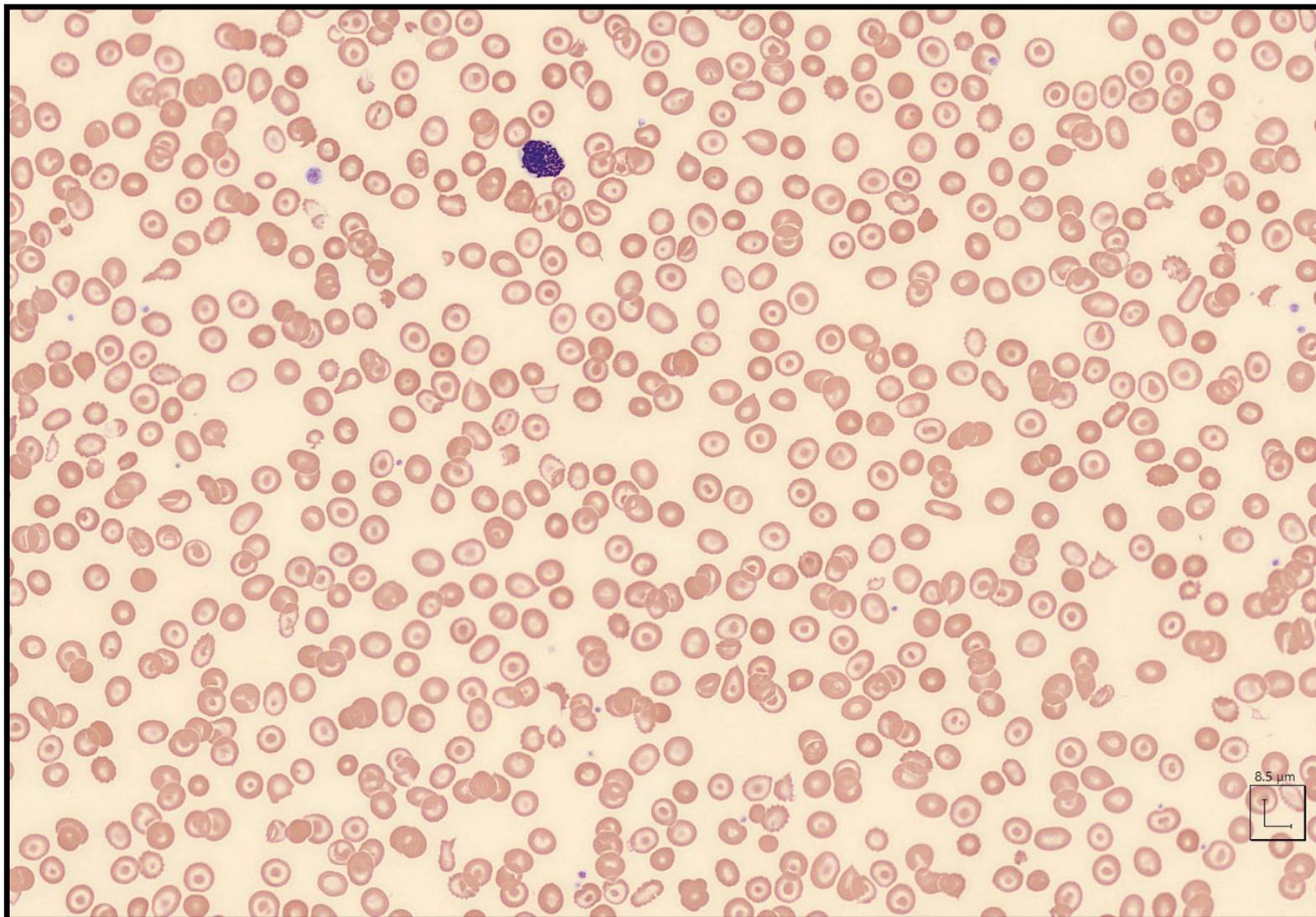


[Fine Art America.com](https://www.fineartamerica.com)



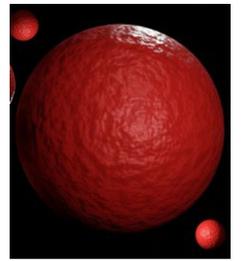
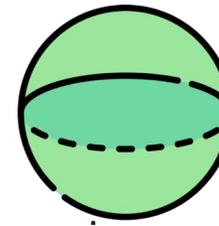
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Target cells post-transplant

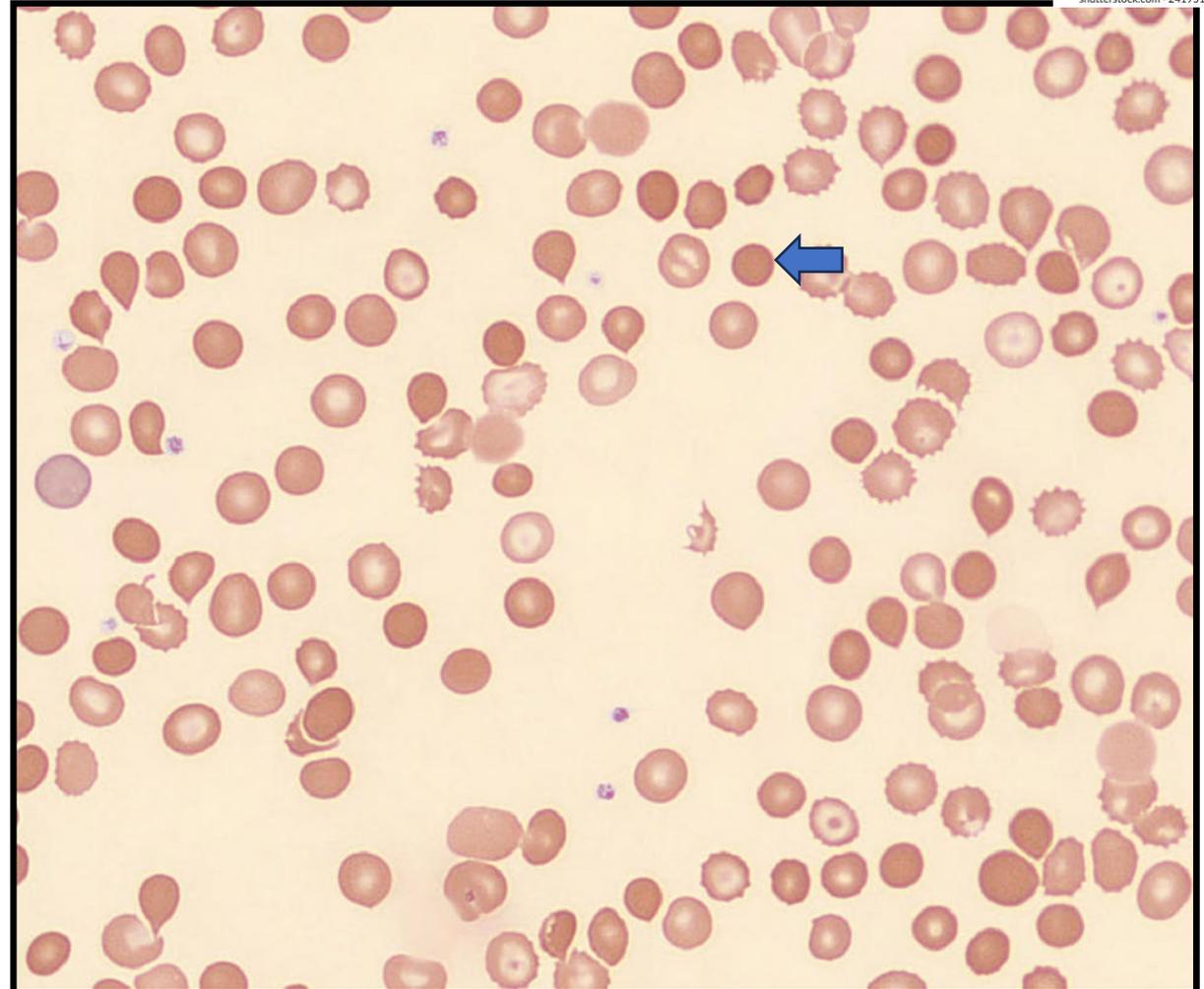


Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

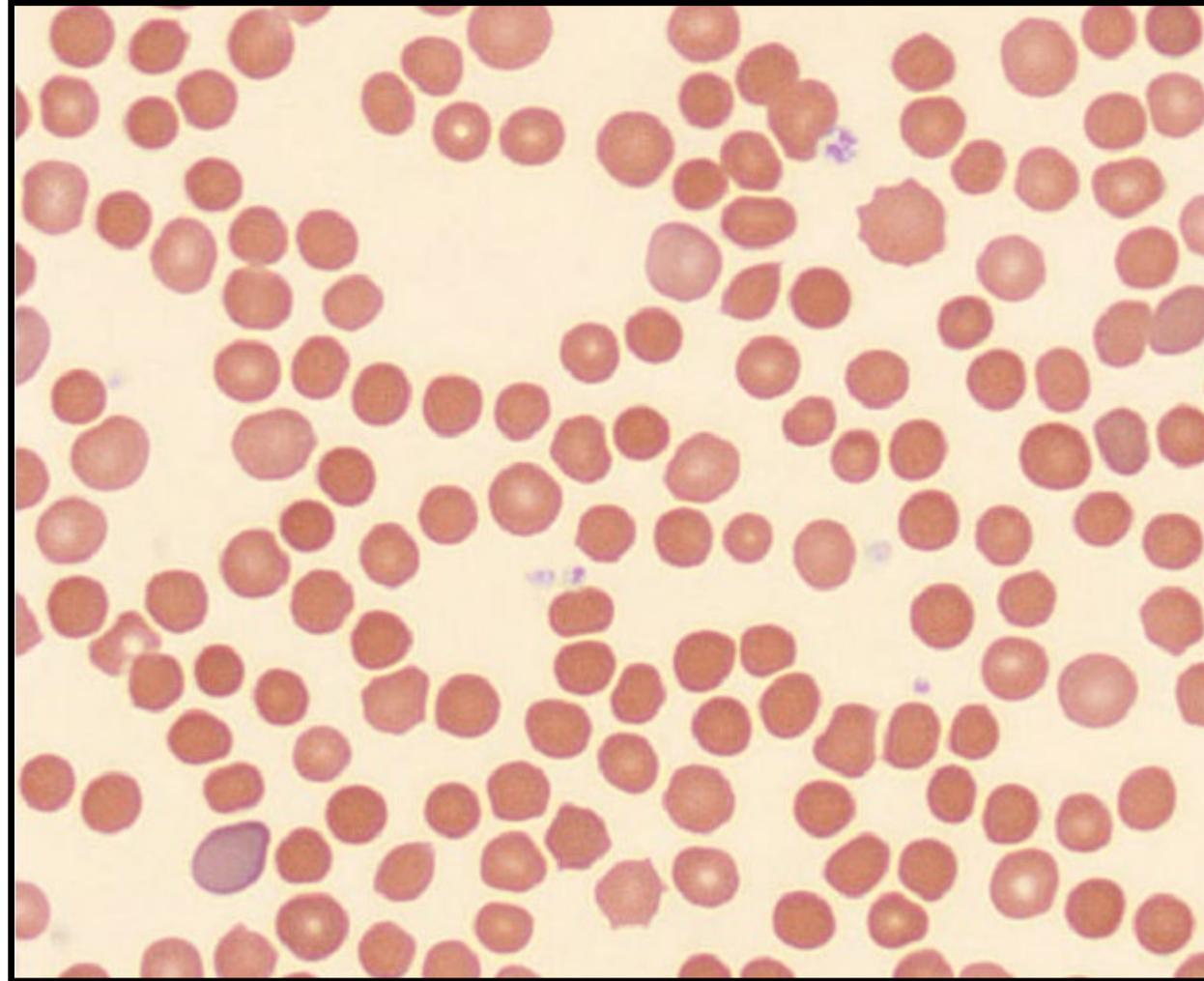
Spherocytes



- Acquired cases seen in autoimmune hemolytic anemia
- Red cells coated with antibody go through spleen
- Fc portion of the antibody interacts with the Fc receptor on splenic macrophages
- A portion of RBC is phagocytosed, leading to the formation of spherocytes
- Spherocytes are more intensely stained, lack central pallor and have a smaller diameter
- MCV is normal
- MCHC is increased

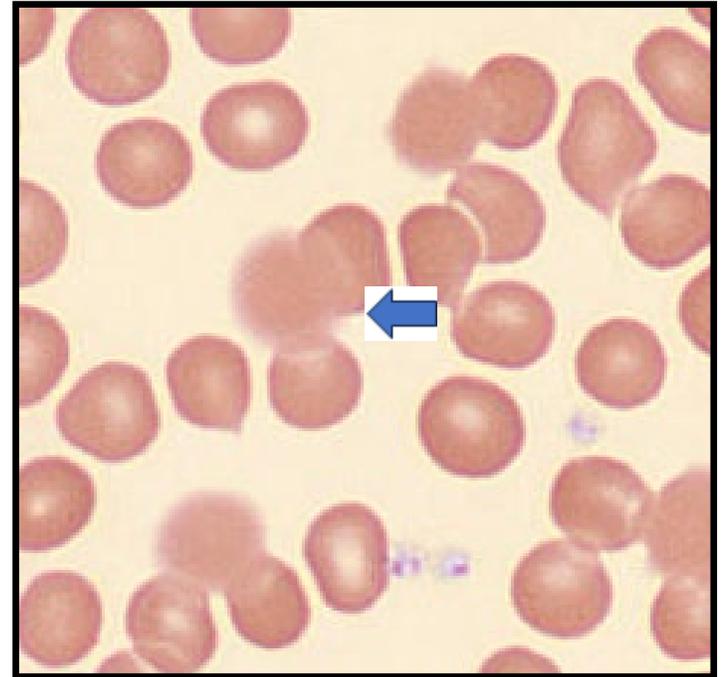
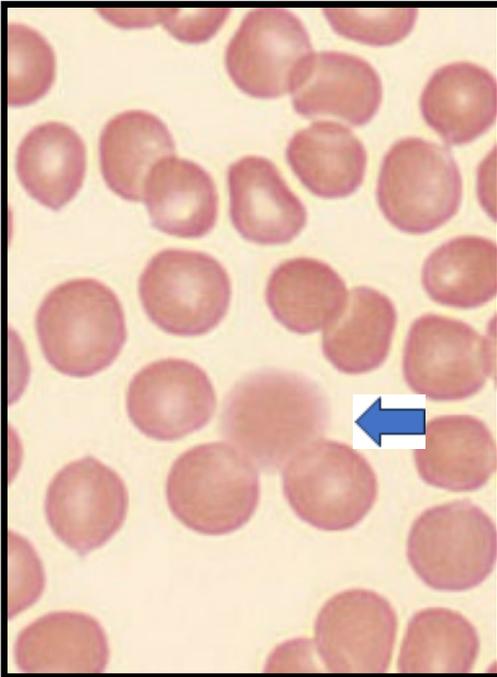


Spherocytosis



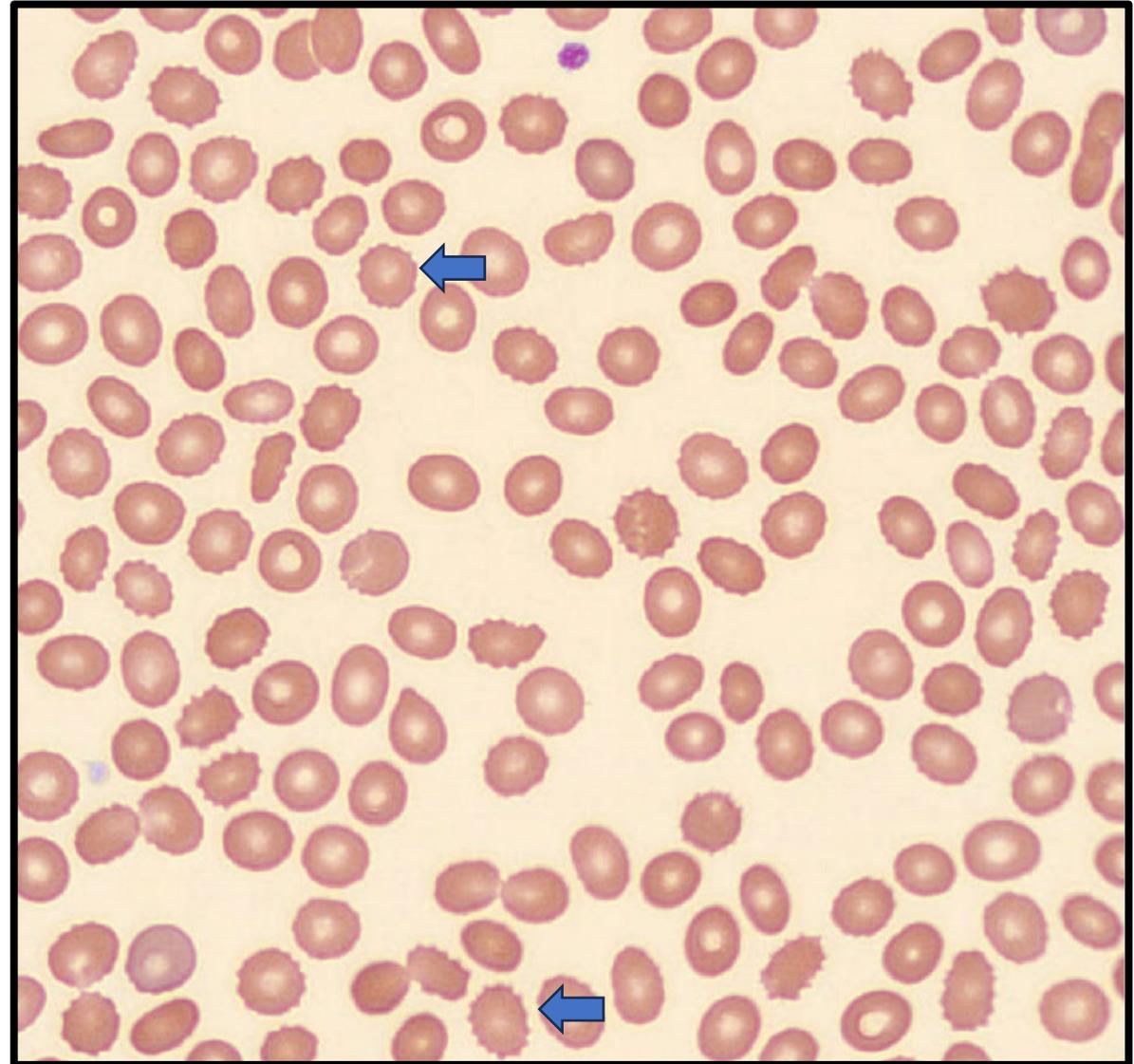
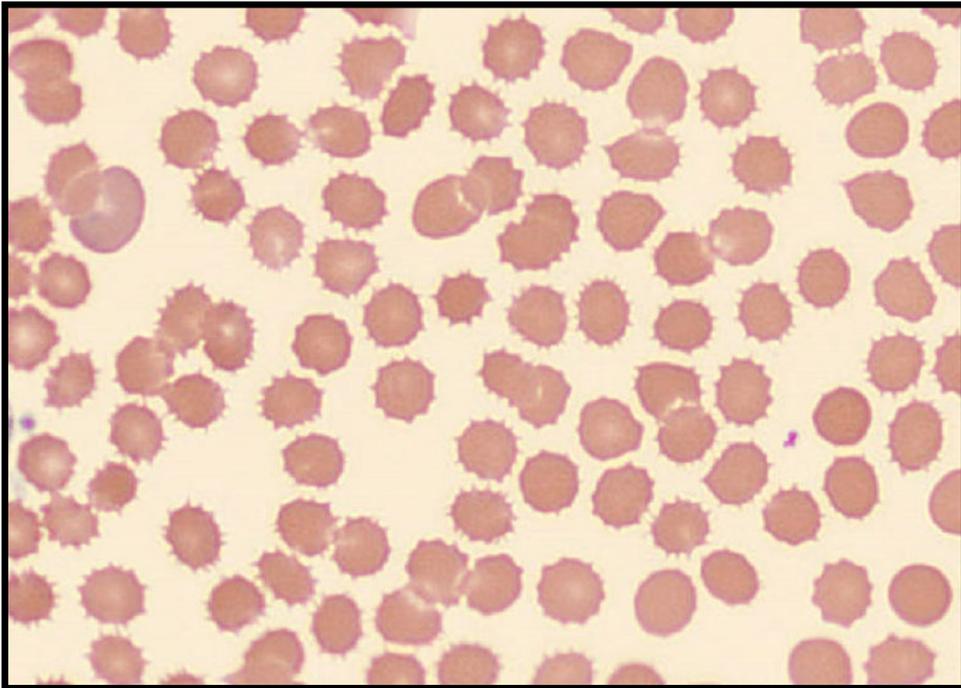
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Ghost RBCs



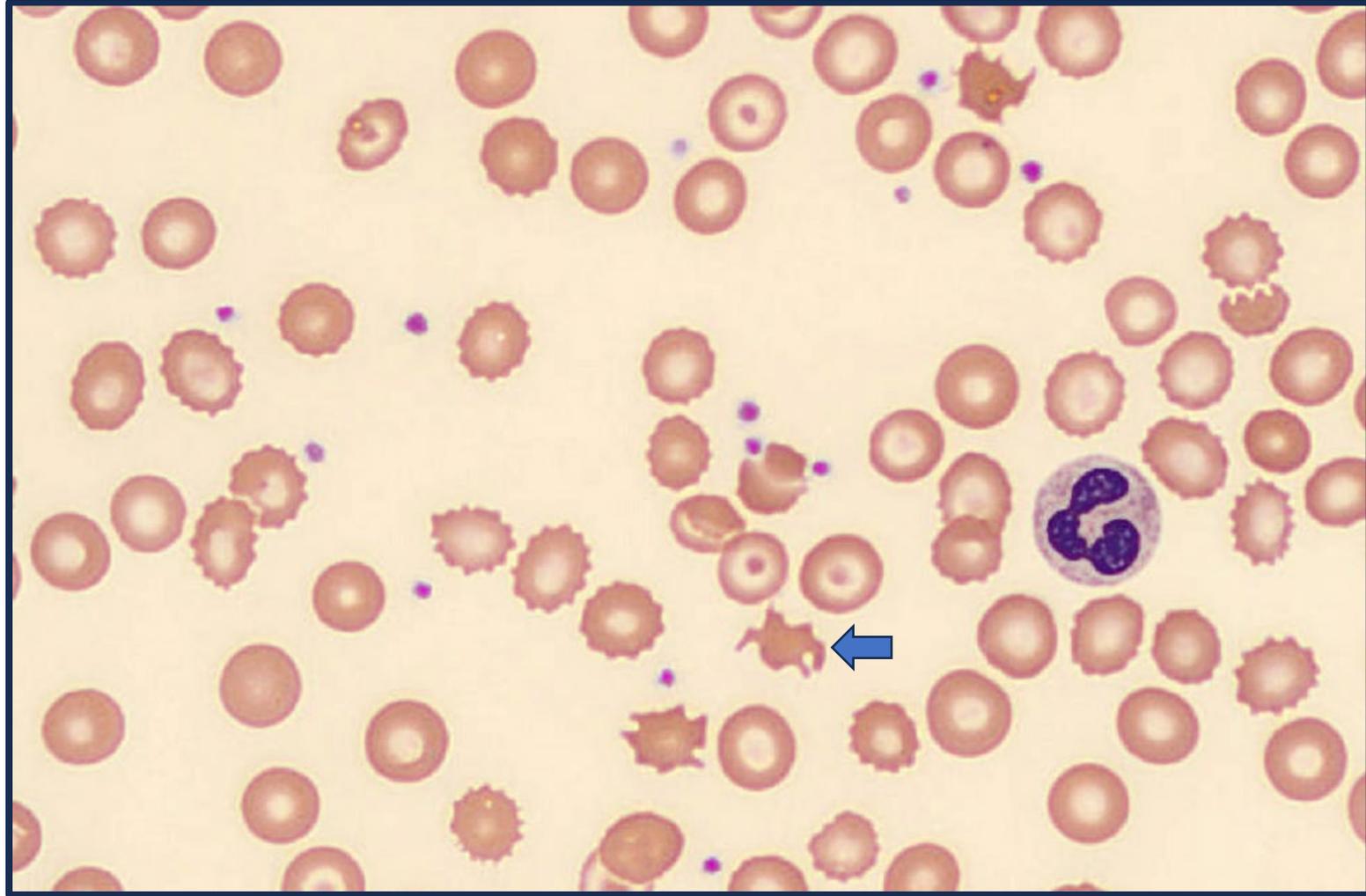
Echinocytes

- Regularly spiculated RBCs
- Seen as an artifact
- Or because of hyperosmolarity or decreased ATP



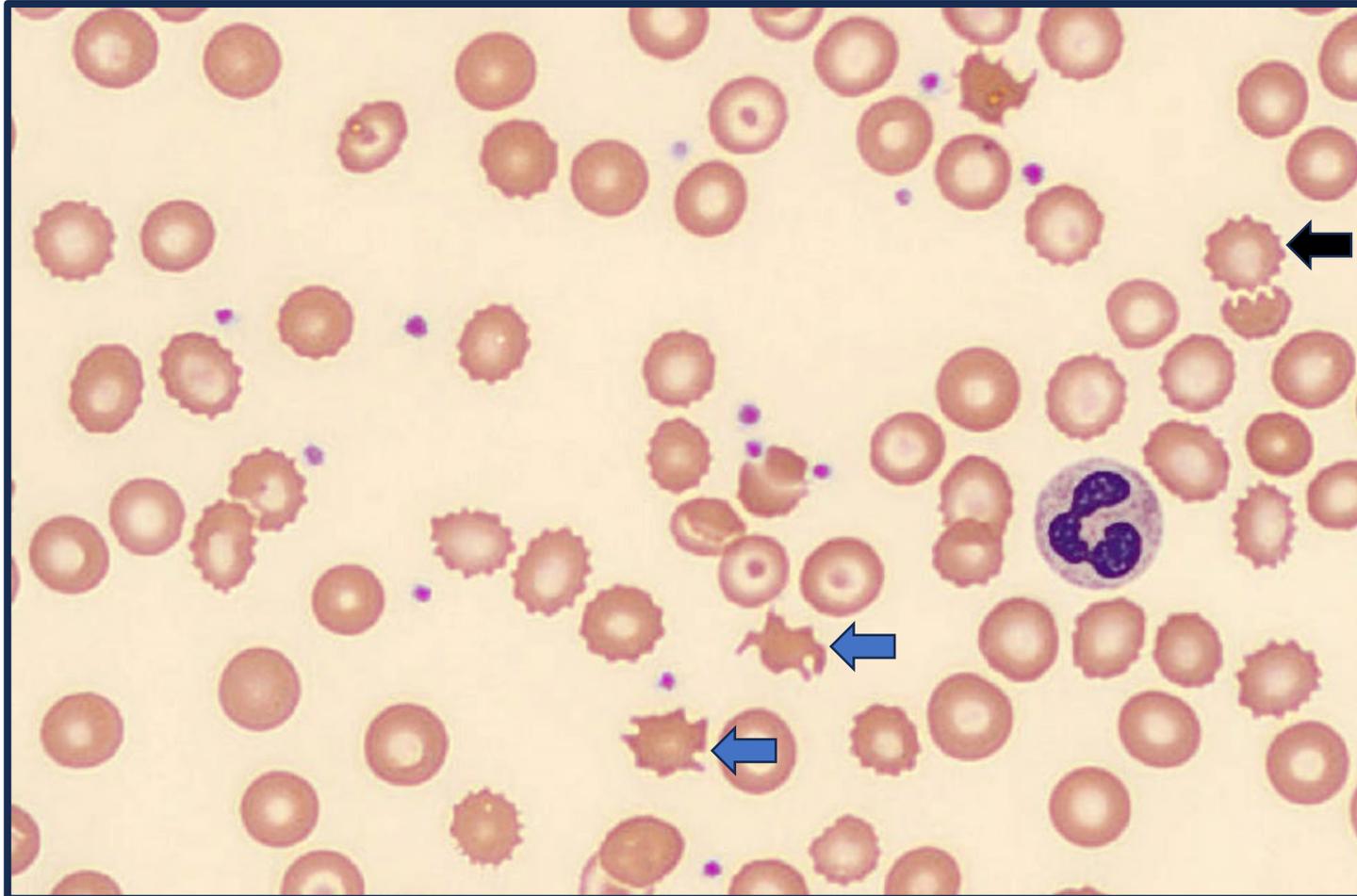
Acanthocytes

- Irregularly spiculated cells
- Ends of the spicules are bulbous
- Seen in conditions with an imbalance of cholesterol and phospholipids
- Abetalipoproteinemia, liver disease.



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Echinocytes and Acanthocytes



Black arrow: Echinocyte
Blue: Acanthocyte

Stomatocytes

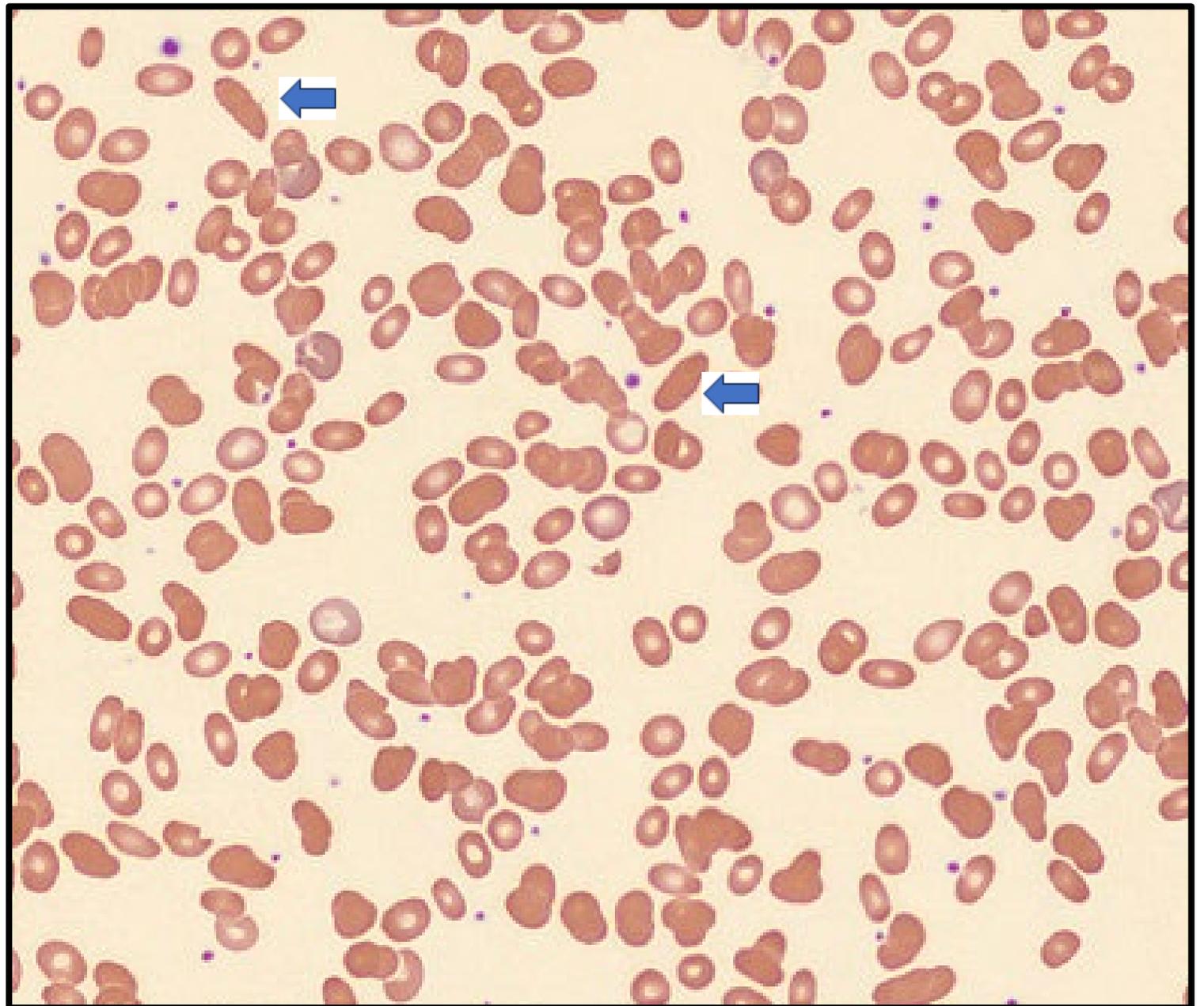
- Instead of a normal central zone of pallor, a slit-like pattern is seen
- Abnormality of red cell membrane permeability
- Seen in hereditary or acquired disorders
- Alcoholism
- Hepatobiliary disease
- Hydroxyurea, propranolol can cause an increase in stomatocytes
- can be seen in normal individuals



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

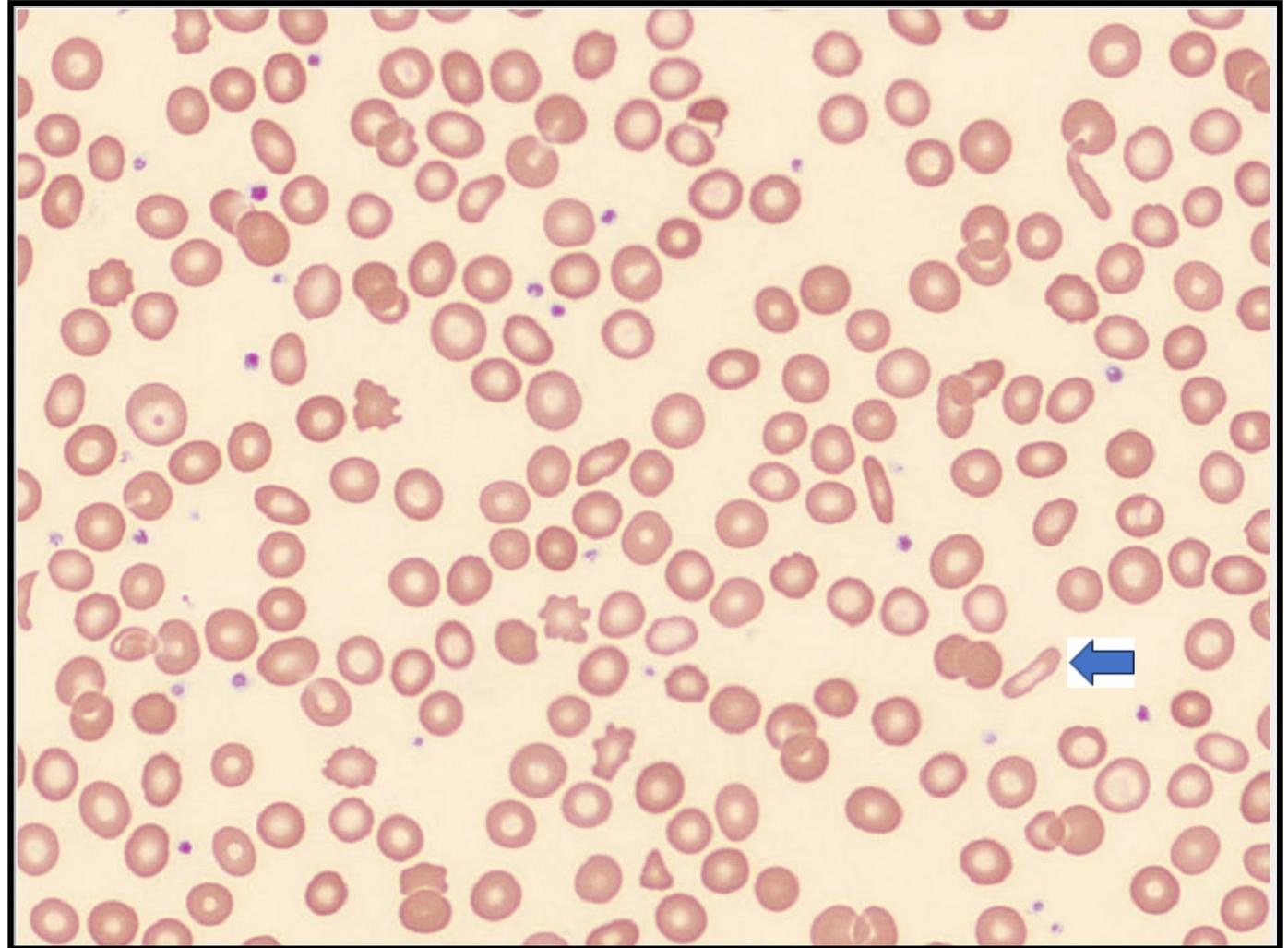
Elliptocytes

- Seen in normal individuals, comprise <10% of cells
- Also seen in iron deficiency anemia, myelofibrosis with extramedullary hematopoiesis and sickle cell anemia
- Most abundant in hereditary elliptocytosis



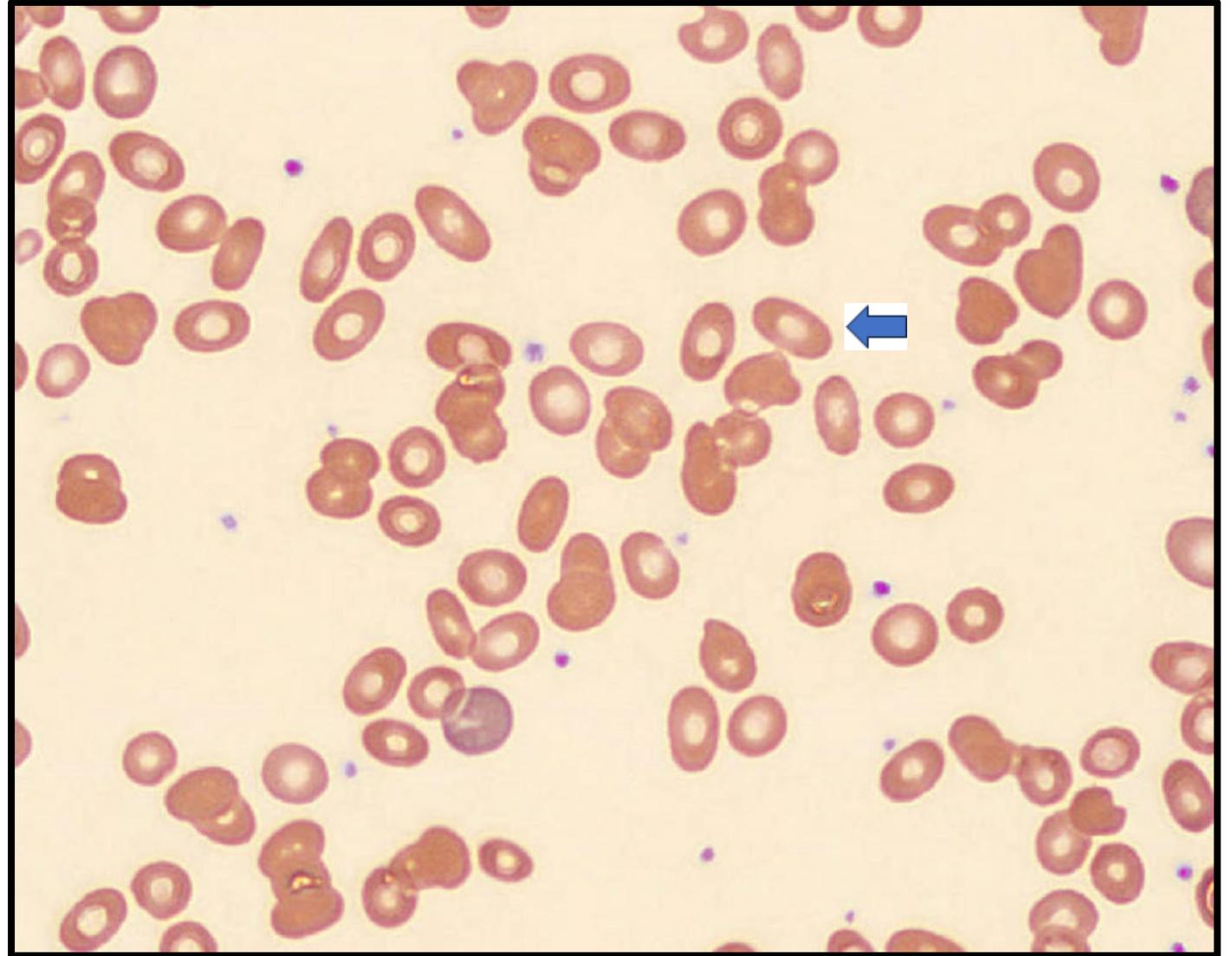
Pencil cells

- In early iron deficiency anemia, red cells are normocytic, normochromic
- Later they develop microcytosis with anisopoikilocytosis
- Cell membranes of iron deficient RBCs are very stiff
- This leads to elongated, hypochromic elliptocytes and pencil cells



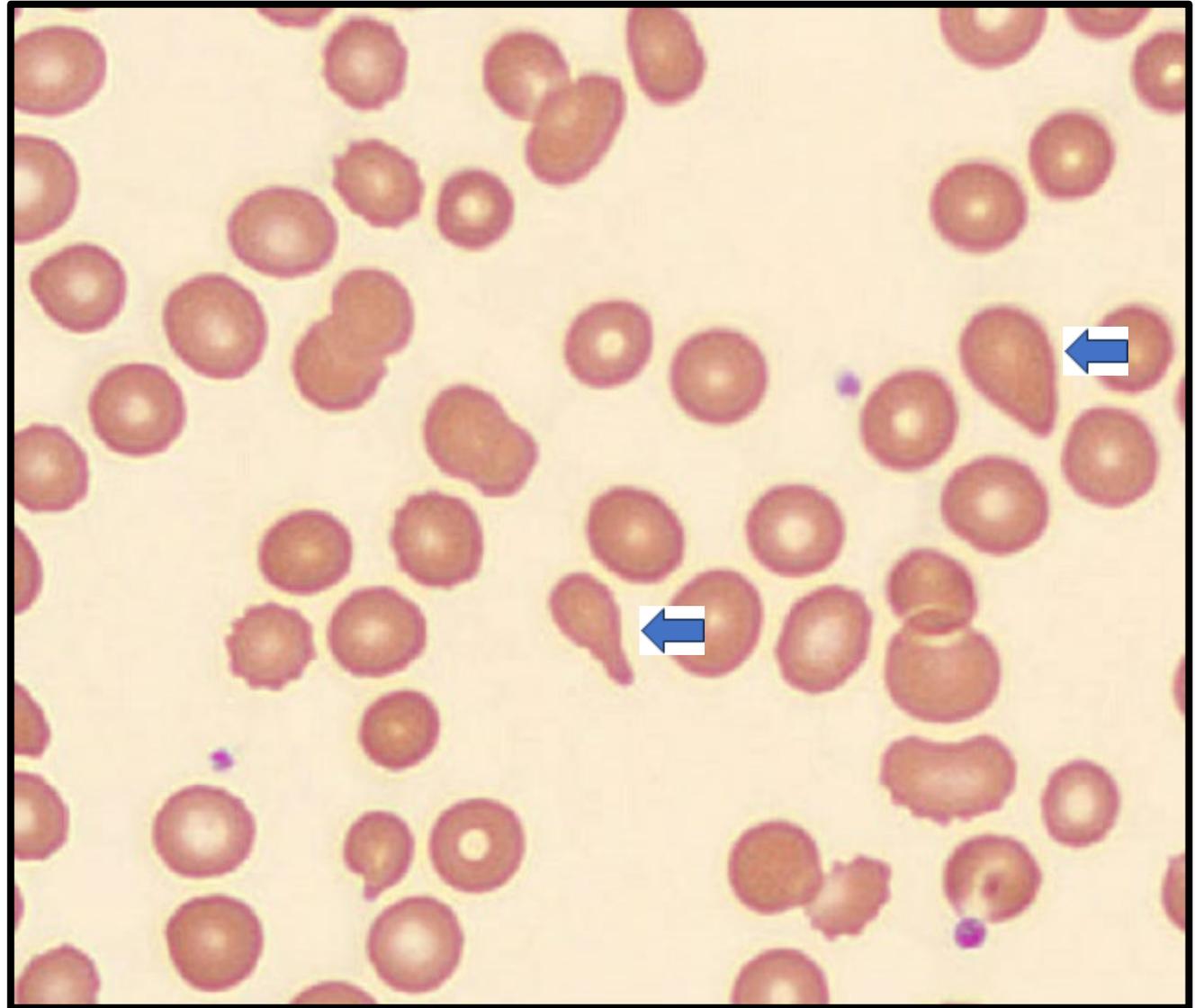
Ovalocytes

- Seen in megaloblastic anemia
- Morphologically similar to elliptocytes

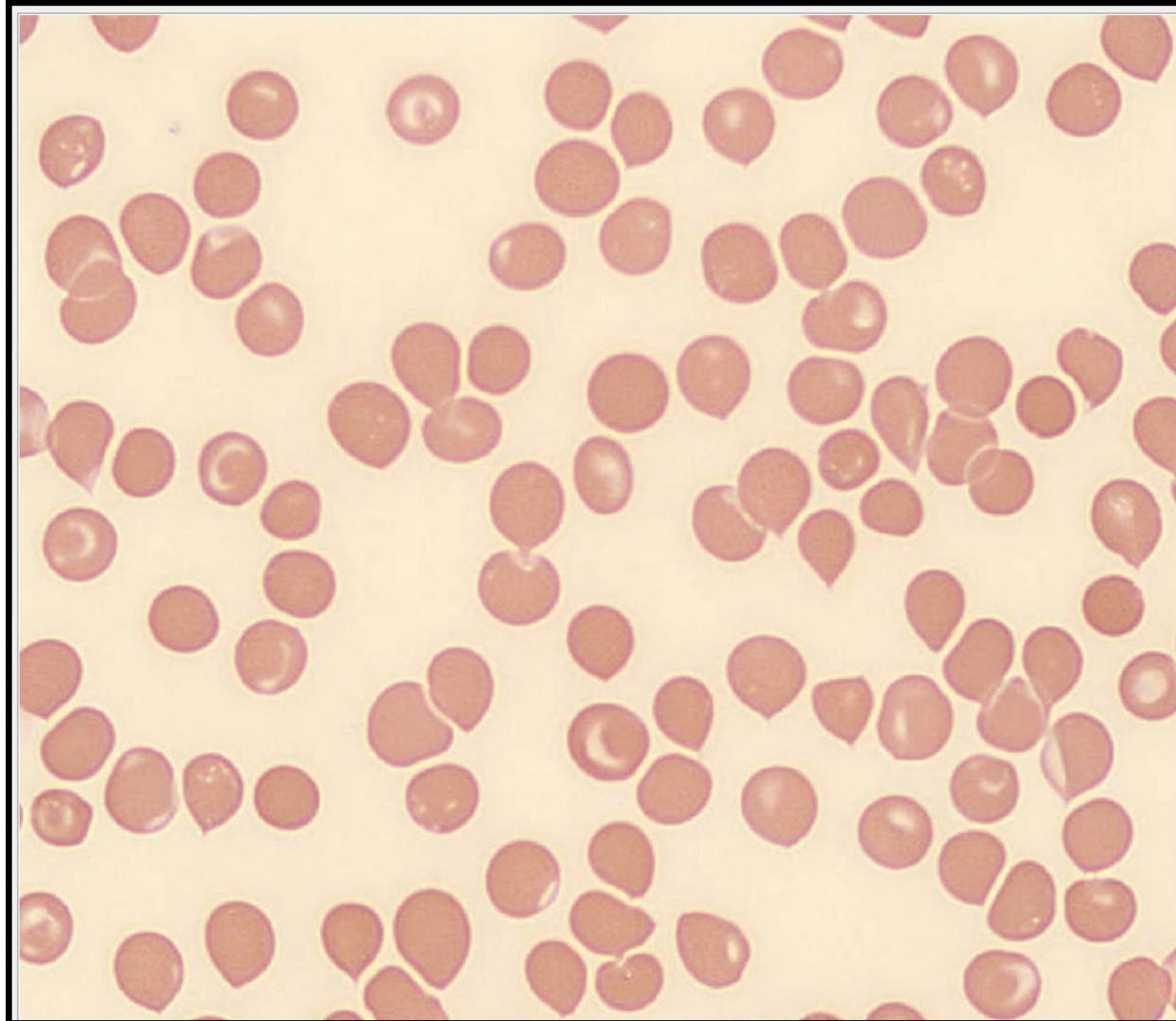


Tear Drop cells

- Associated with bone marrow infiltration by fibrosis, granulomatous inflammation or malignancy
- True tear drop cells tips point in different directions



Tear drop cells artifact



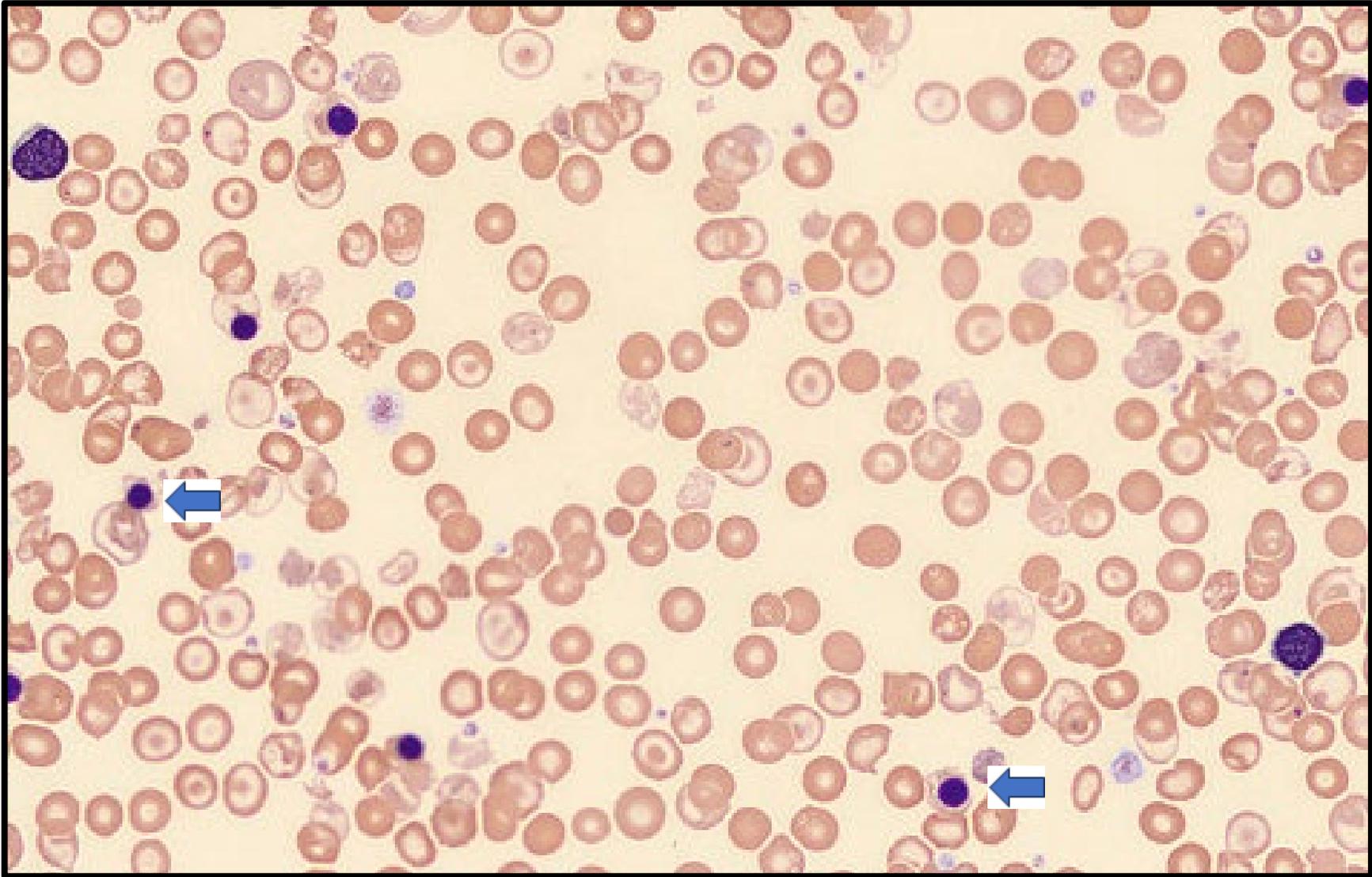
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Case

- 36 yr/F with sickle cell disease/beta thalassemia
- Presented with pain crisis
- Lab results:

	Result	Reference range
• Hb	9.1 ↓	12.5-15 g/dL
• RBC	3.8	3.5-5.5 x10 ⁶ /uL
• MCV	65 ↓	80-95 fL
• Retics	10% ↑	0.5-2%
• WBC	23 ↑	4-11 x 10 ³ /uL
• Platelet	236	150-450 x 10 ³ /uL

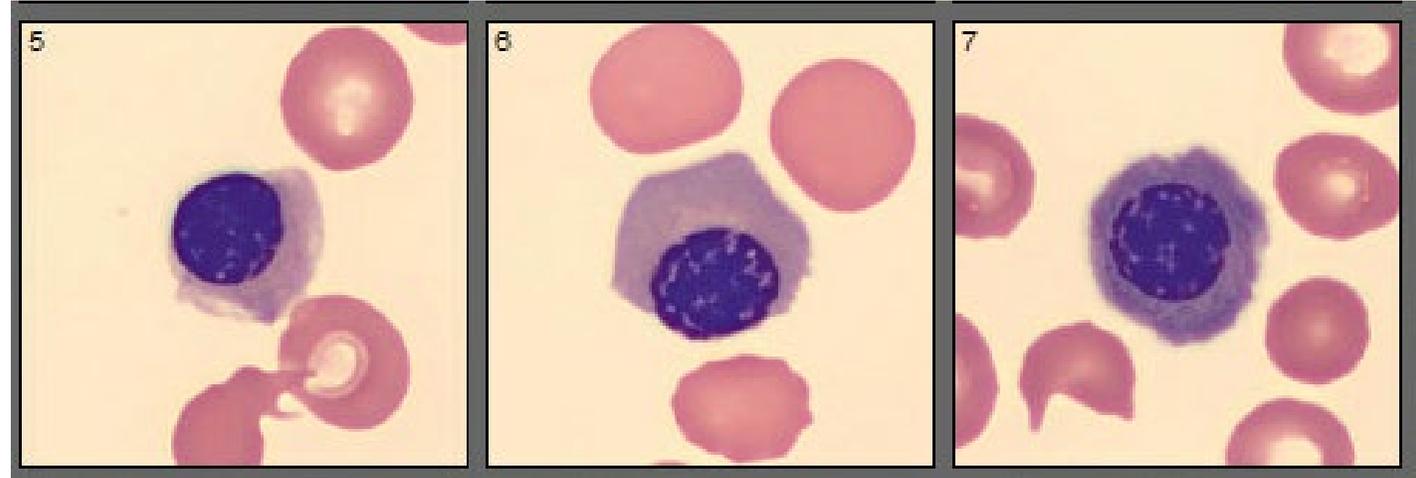
Smear



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

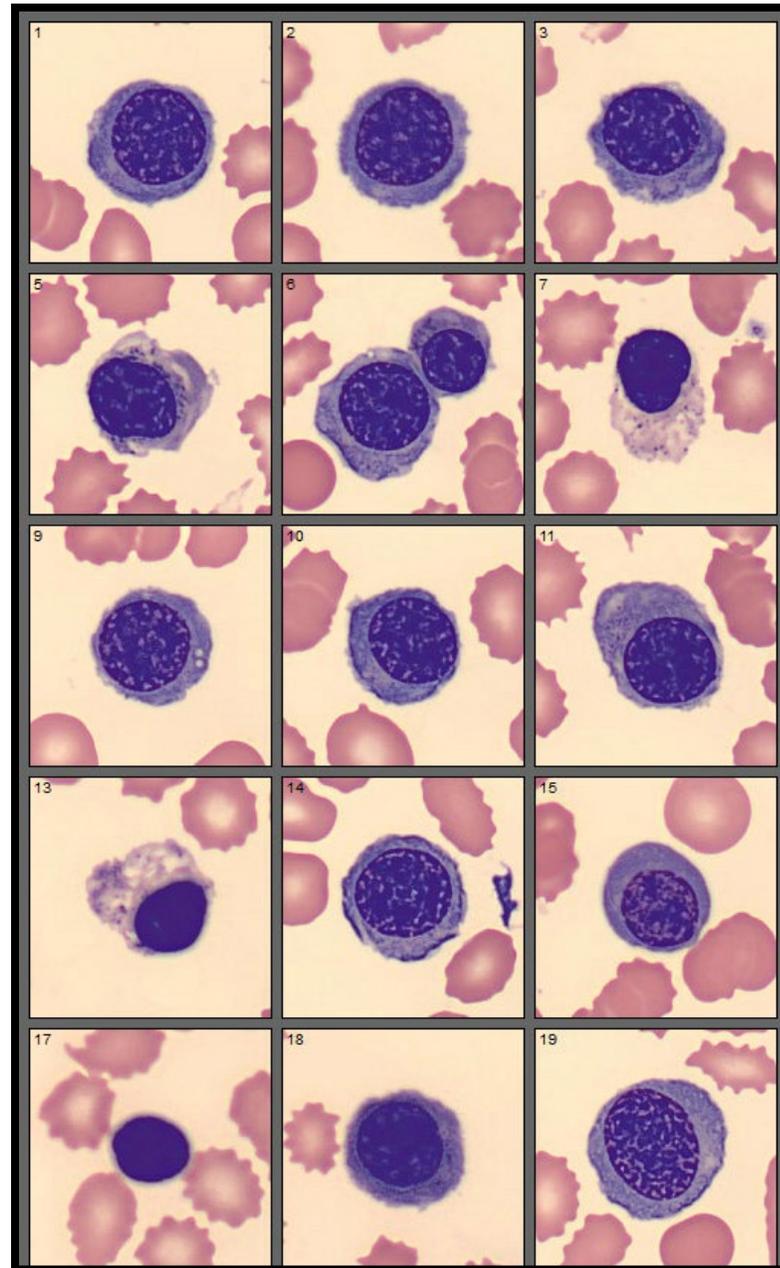
Nucleated Red Cells (nRBCs)

- Mature RBCs do not contain a nucleus
- In humans, nRBCs are only present in the bone marrow normally
- Their presence in peripheral blood indicates increased production due to excessive demand, extramedullary hematopoiesis or bone marrow infiltrating disease
- Large number of these cells are seen in thalassemia, sickle cell disease



Source: Dr. Afshan Idrees, University of Miami Department of
Pathology & Laboratory Medicine

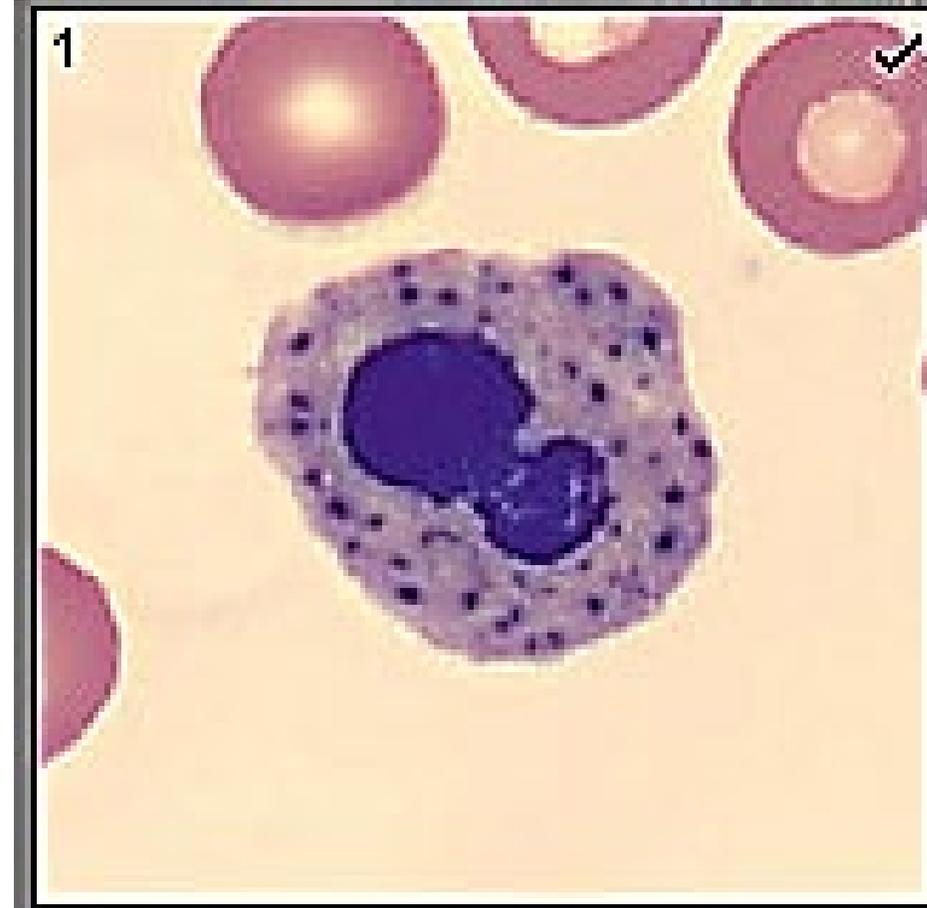
Nucleated Red Cells (nRBCs)



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

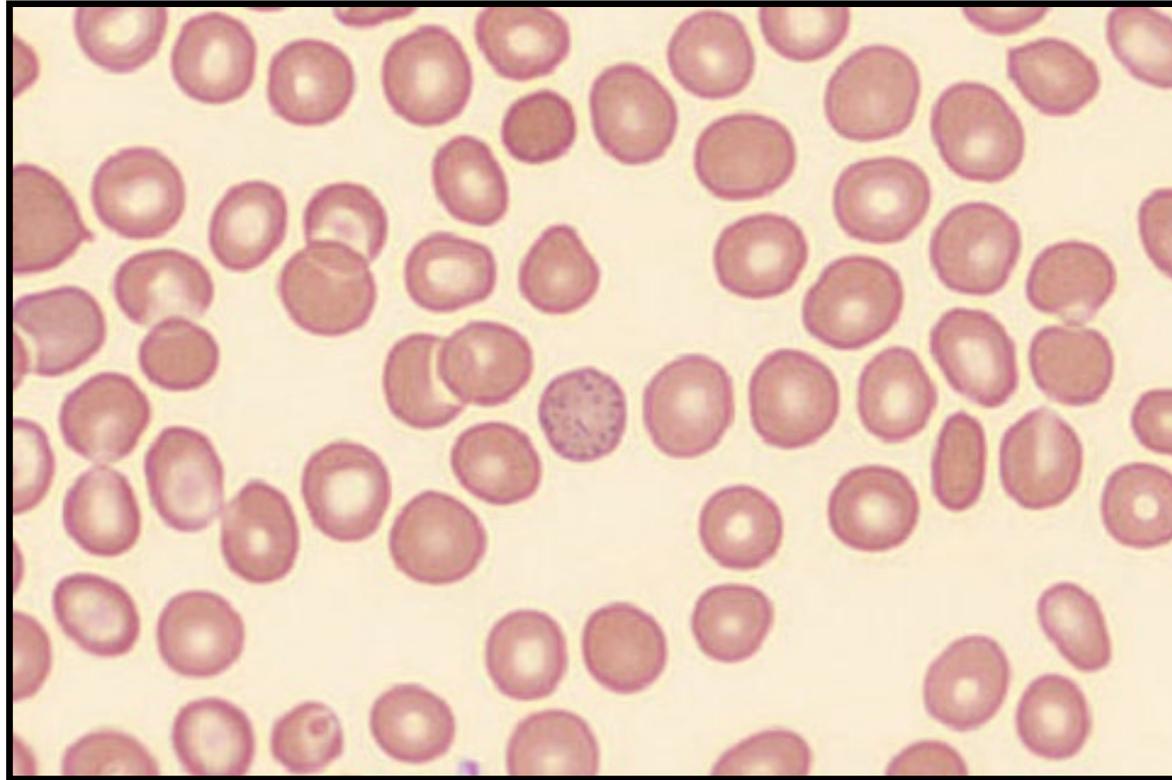
Basophilic stippling

- Fine or coarse
- Deep blue granules representing RNA
- Commonly seen in increased red cell production
- Fine stippling seen in reticulocytes
- Coarse stippling seen in hemoglobinopathies and MDS



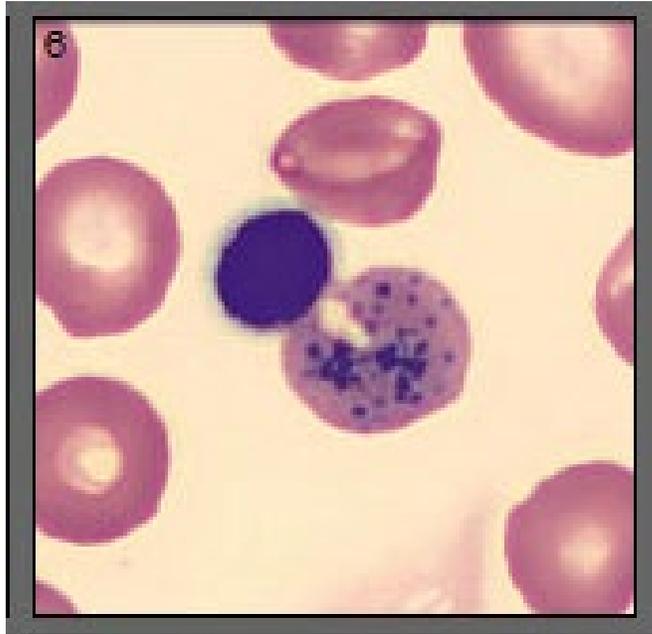
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Fine Basophilic stippling in alpha thalassemia



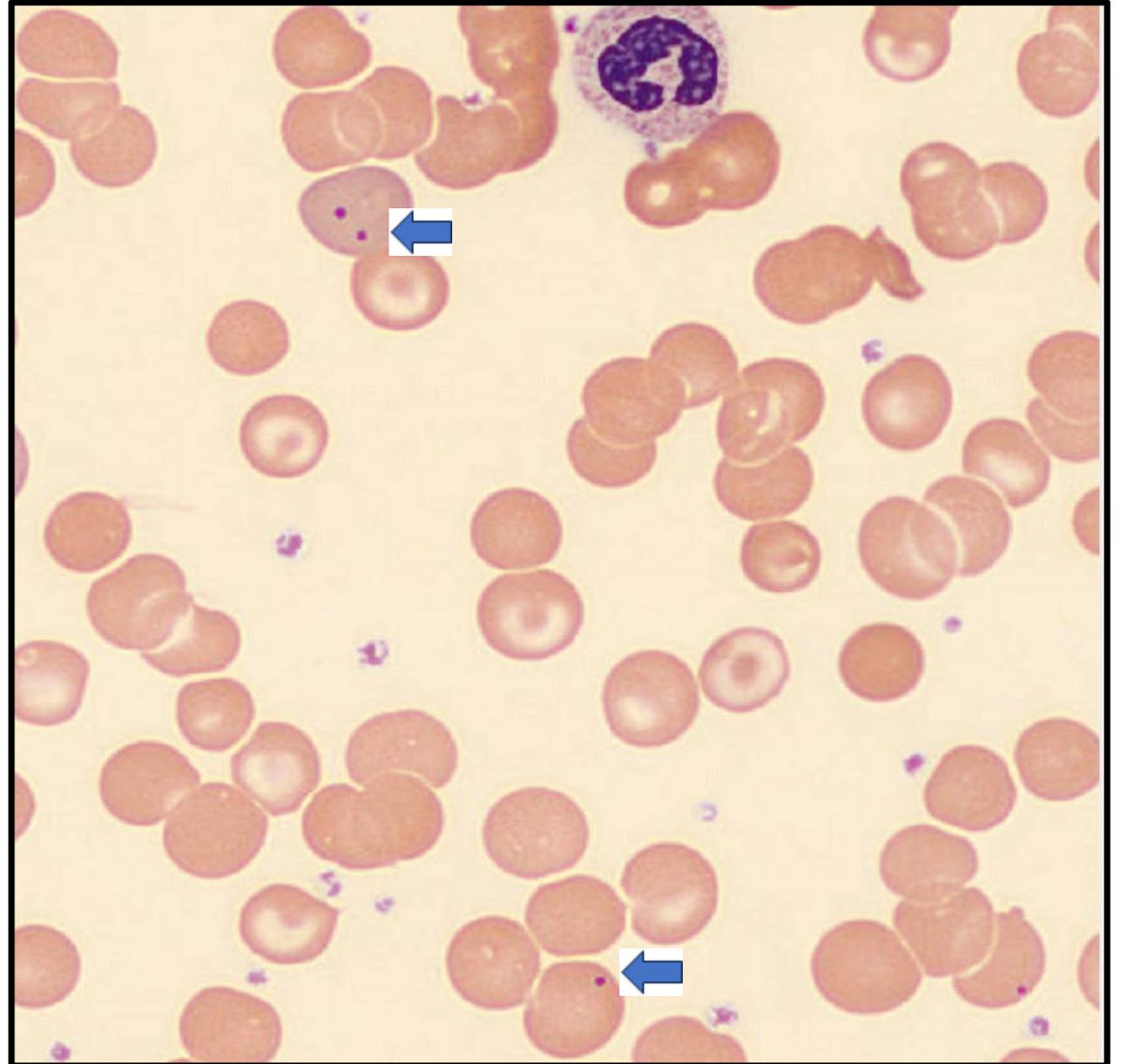
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Basophilic stippling vs ring sideroblast in MDS



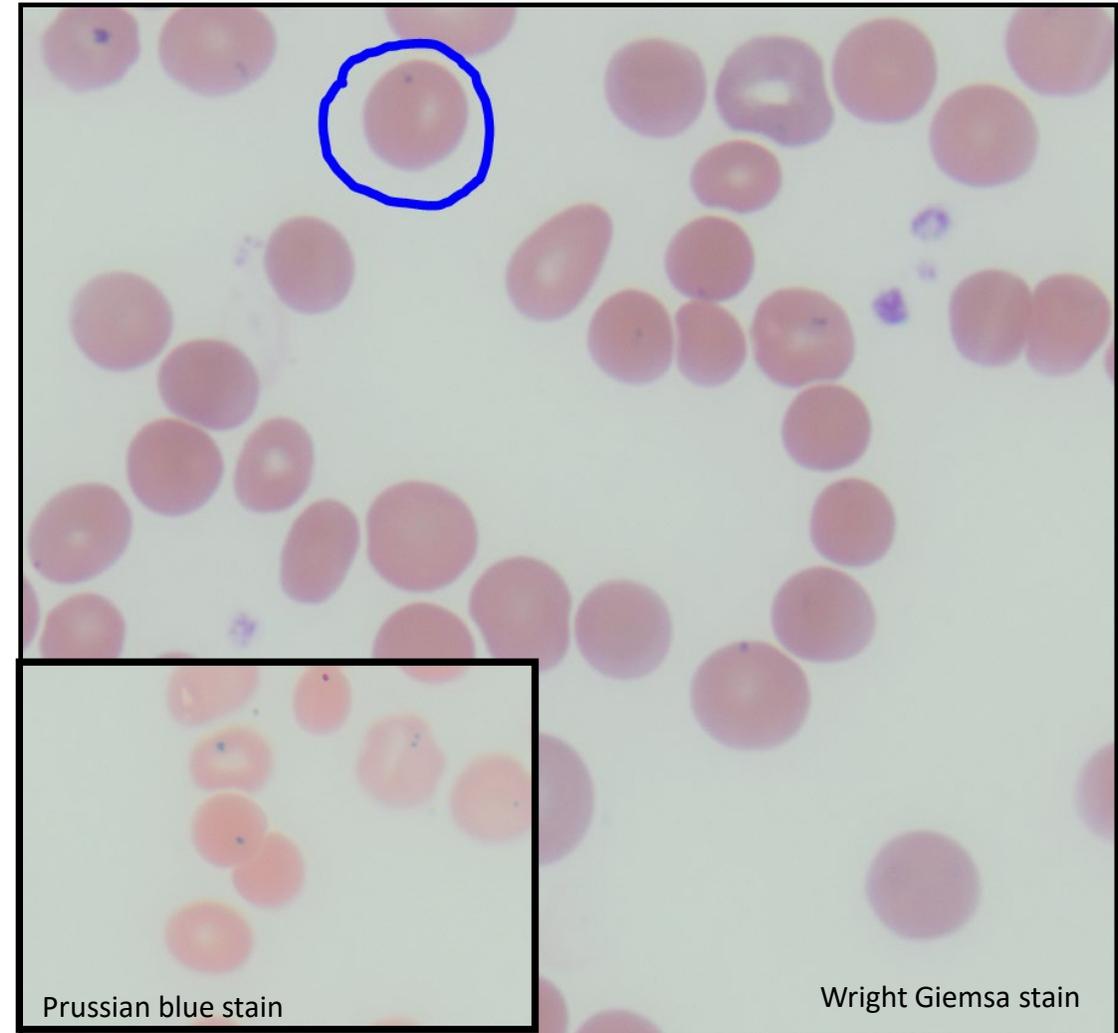
Howell-Jolly Bodies

- Smooth, round remnants of nuclear chromatin
- In physiological conditions, these are removed or groomed by the spleen.
- Can be single or multiple
- They can be seen in hemolytic anemia, post-splenectomy
- Multiple Howell-Jolly bodies can be seen in megaloblastic anemia or abnormal erythropoiesis



Pappenheimer bodies

- Appear as single or paired or multiple dot like inclusions
- Iron-containing
- Purple on Giemsa stain
- Blue on Prussian blue stain
- Seen in ineffective erythropoiesis, post-splenectomy, lead poisoning



Red cell distribution

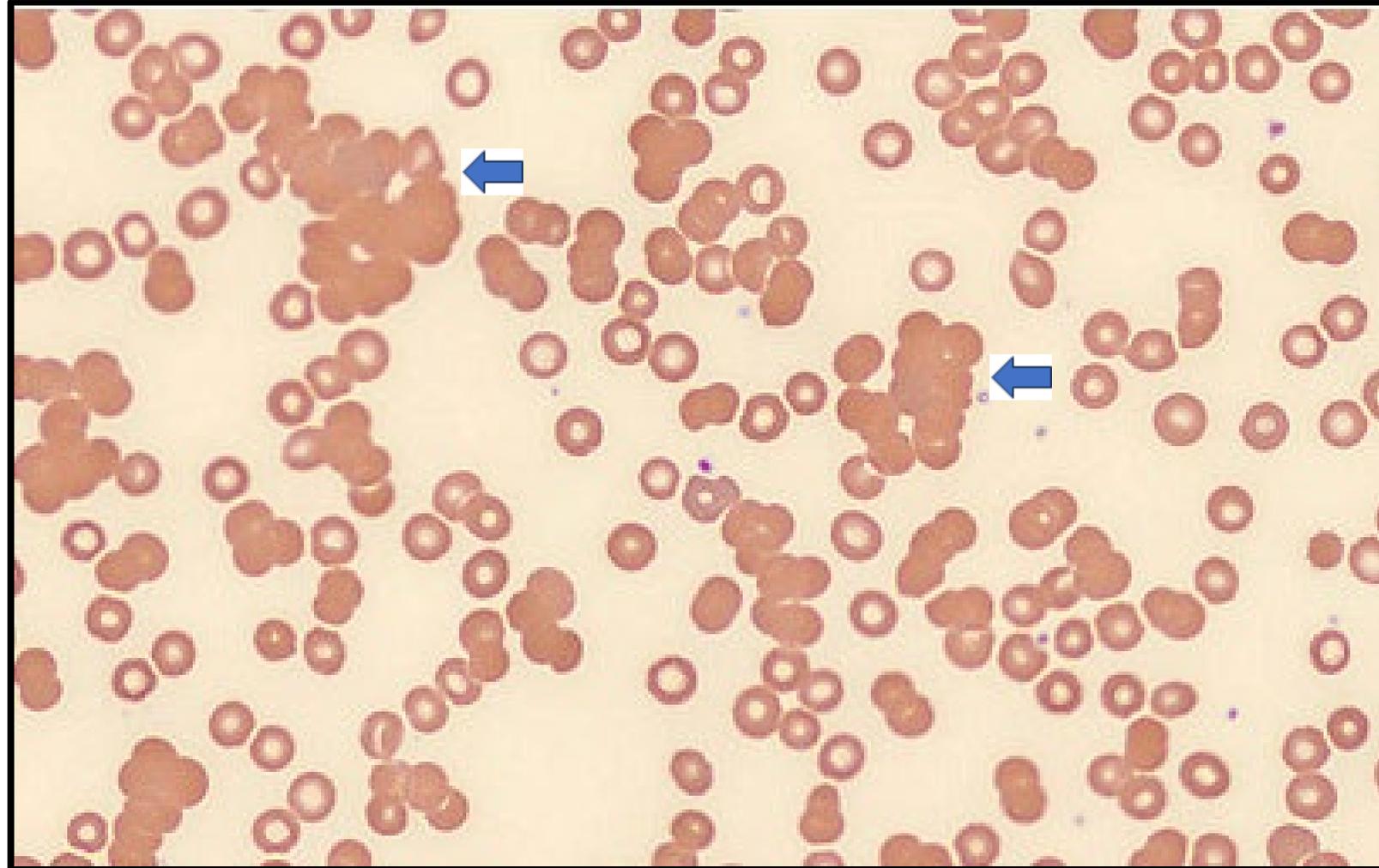
Case

- 74/M with monoclonal gammopathy of unknown significance and M-spike of IgM
- Lab results:

	Result	Reference Range
• Hb	7.4 ↓	12.5-15 g/dL
• RBC	1.8 ↓	3.5-5.5 x10 ⁶ /uL
• MCV	100 ↑	80-98 fL
• MCHC	43.8 ↑	32-35.2 g/dL
• WBC	6.5	4-11 x 10 ³ /uL
• Platelet	130 ↓	150-450 x 10 ³ /uL

RBC Agglutination

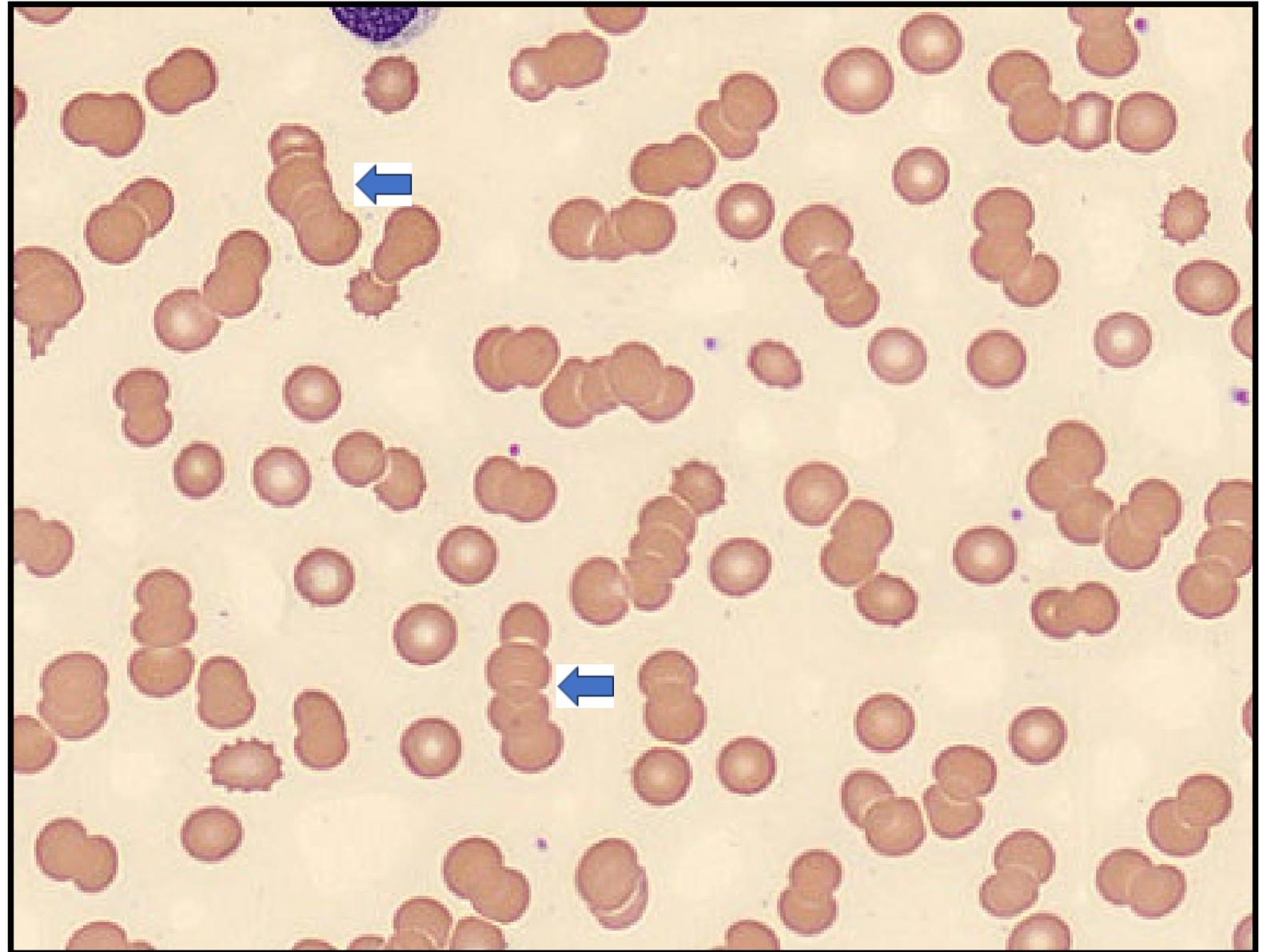
- Agglutination or clumping of RBCs differs from Rouleaux
- It tends to show more irregular clumps compared to linear rouleaux
- Mycoplasma pneumoniae or infectious mononucleosis
- Lymphoplasmacytic lymphoma



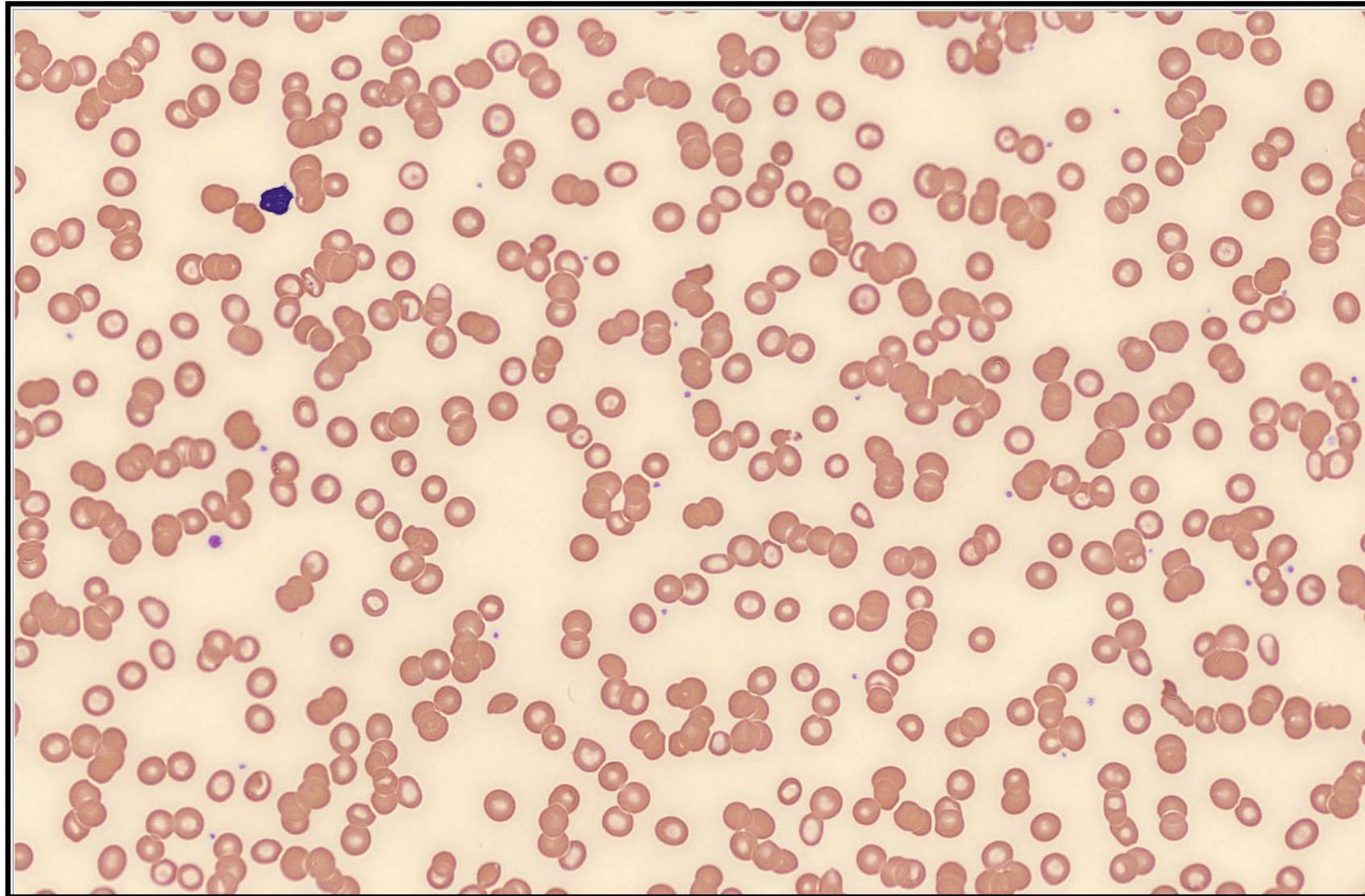
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Rouleaux

- Alignment of red cells one upon another so that they resemble stack of coins
- Elevated plasma fibrinogen or globulins can cause rouleaux as well as an increased ESR
- Marked rouleaux is seen in paraproteinemia, plasma cell myeloma



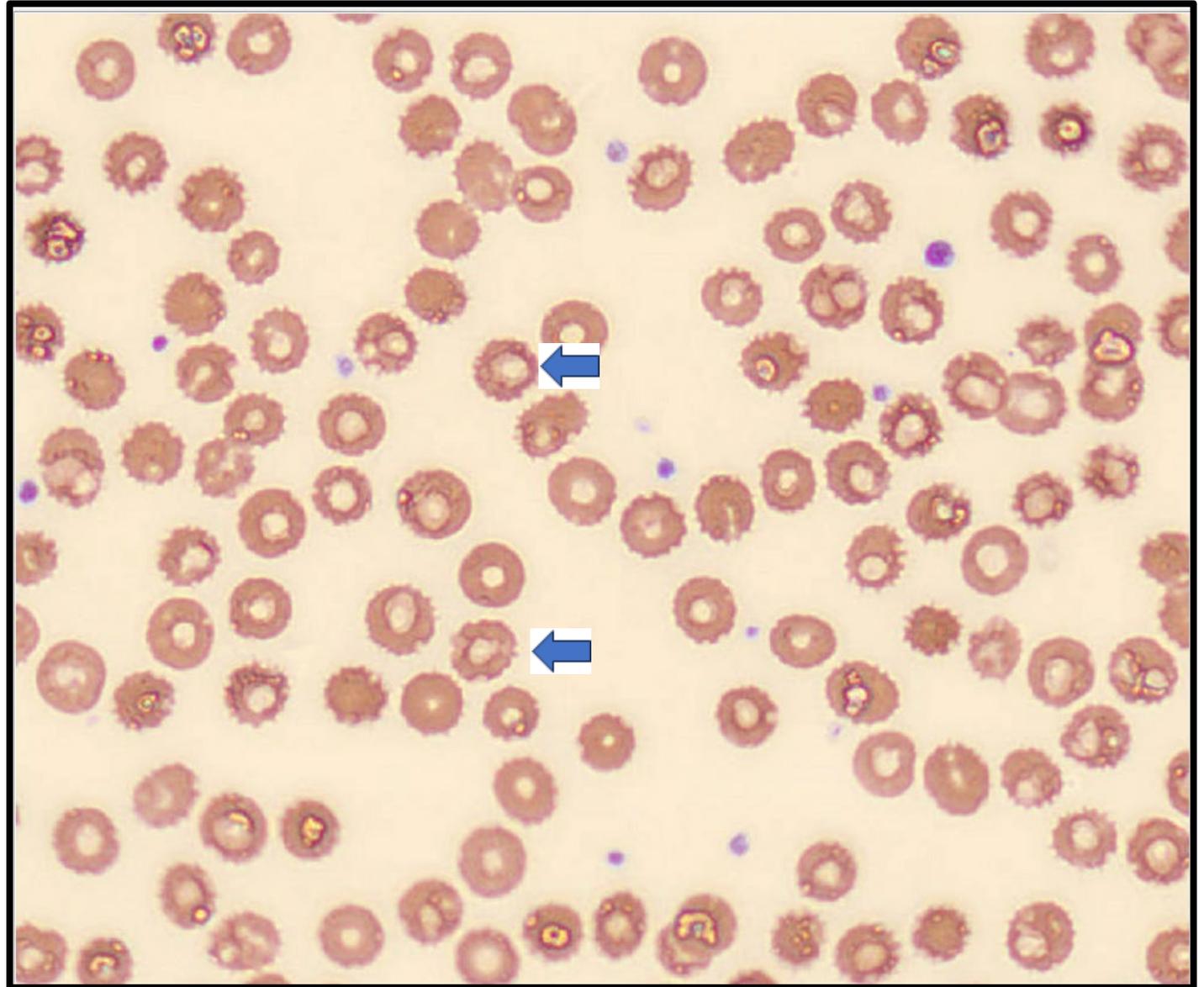
Rouleaux



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

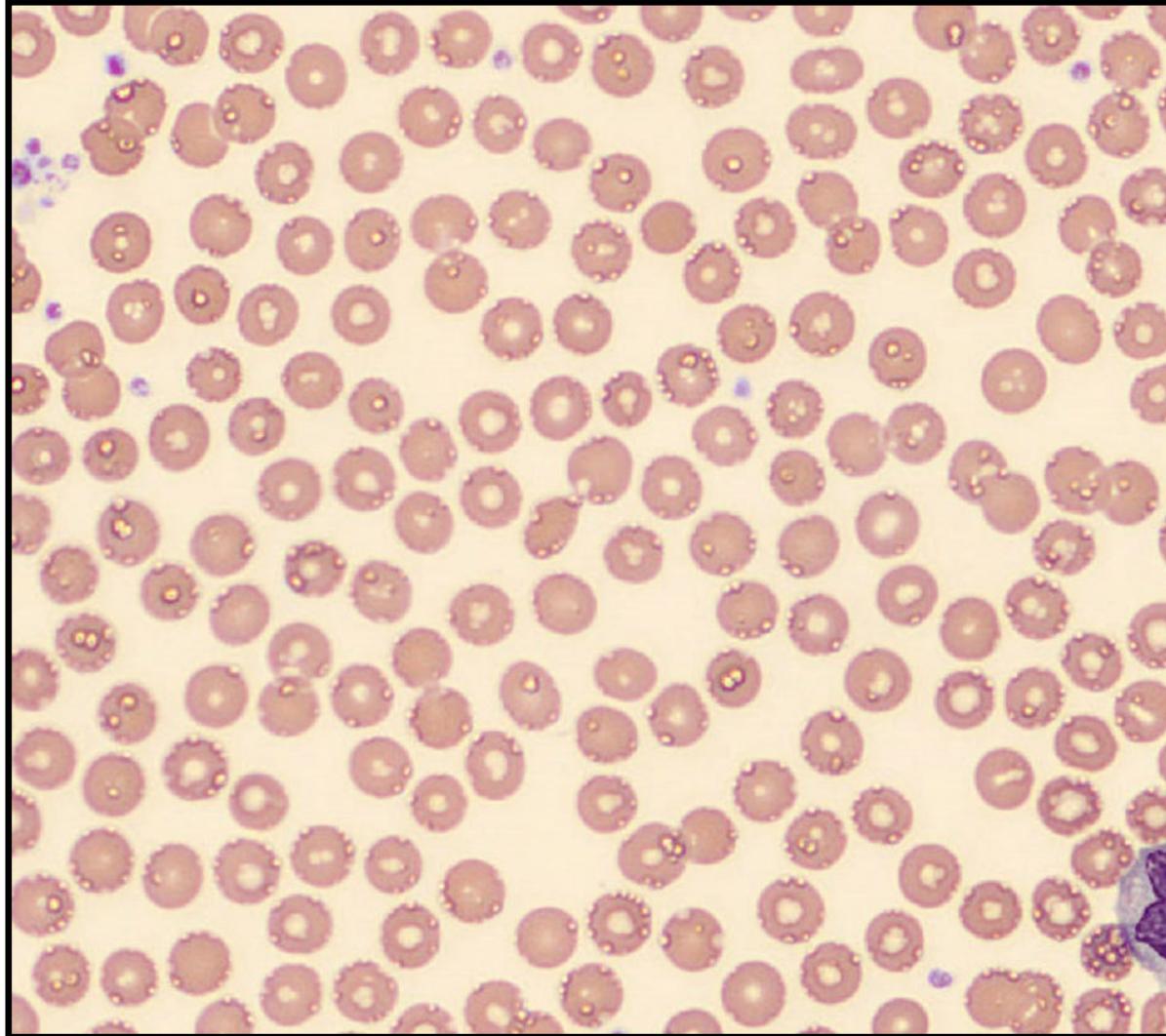
Artifact

- Tiny pits or bubbles in the red cells can be caused by a small amount of water contaminating the Wright stain or
- Due to insufficient slide drying



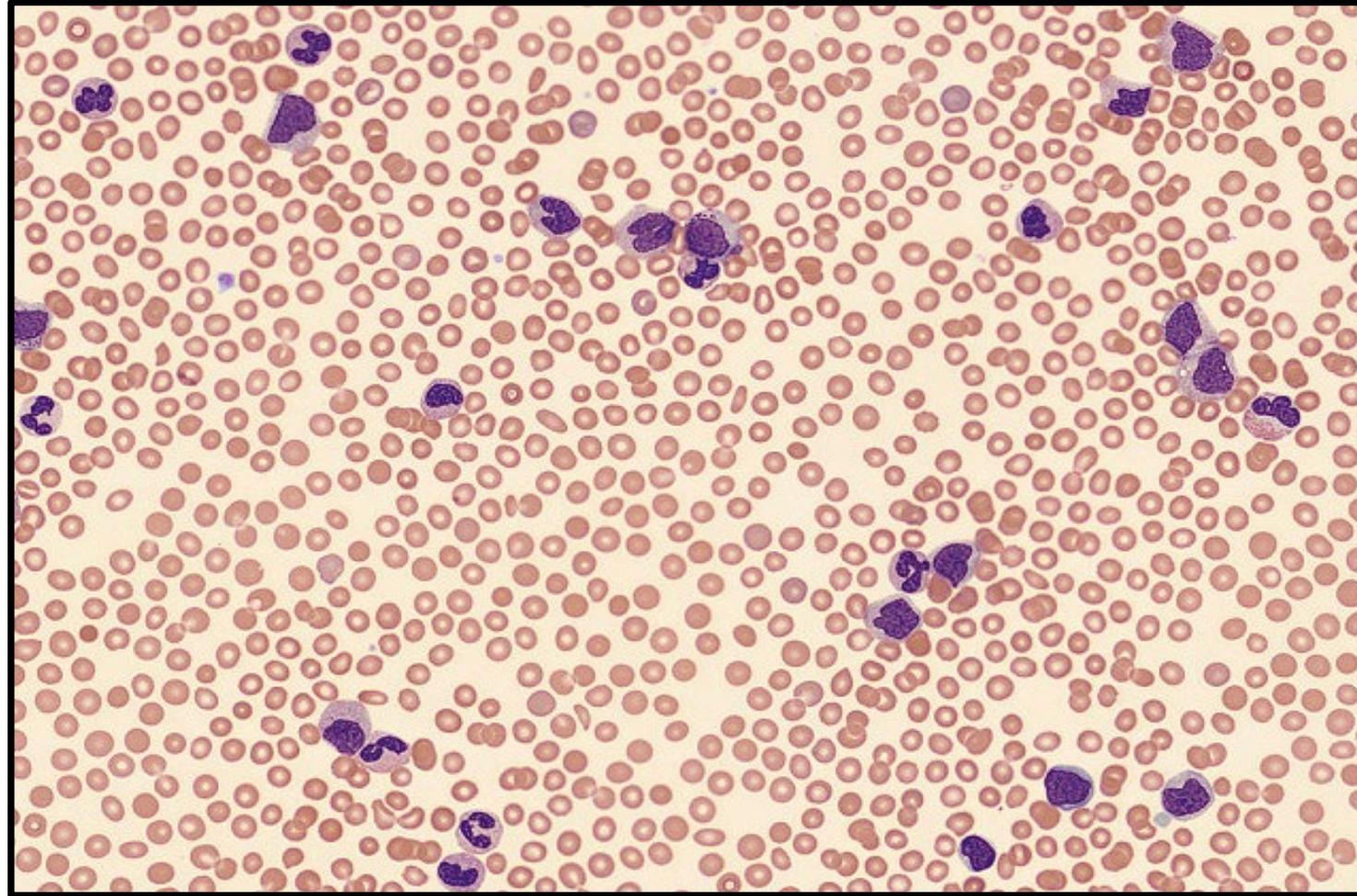
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Water Artifact



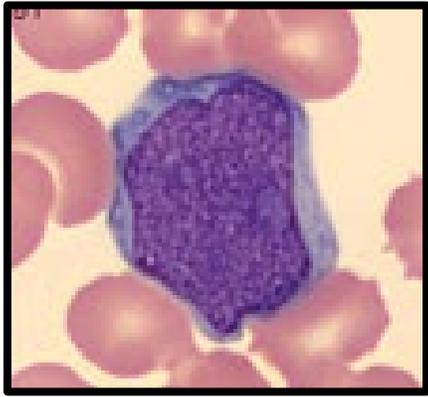
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

White Blood cells

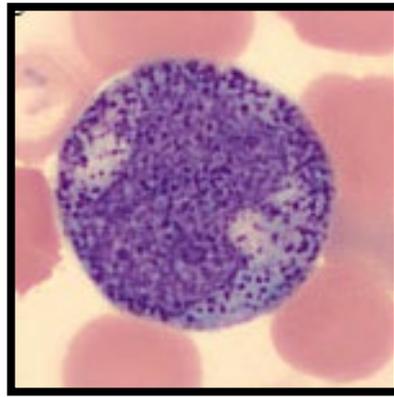


Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

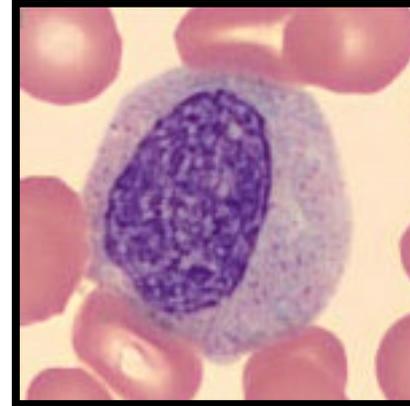
Myeloid maturation



Blast



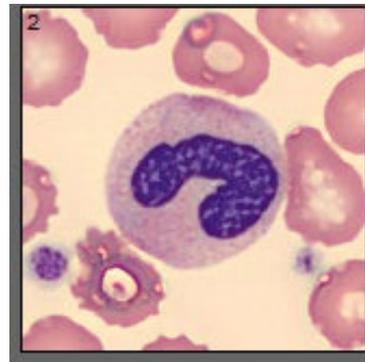
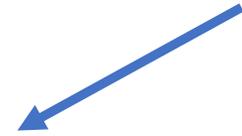
Promyelocyte



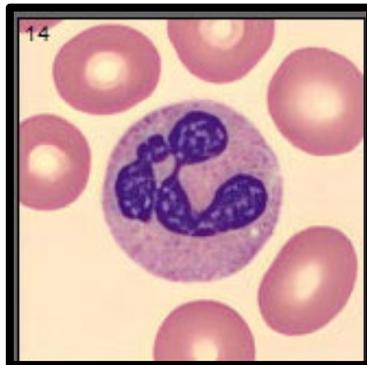
Myelocyte



Metamyelocyte



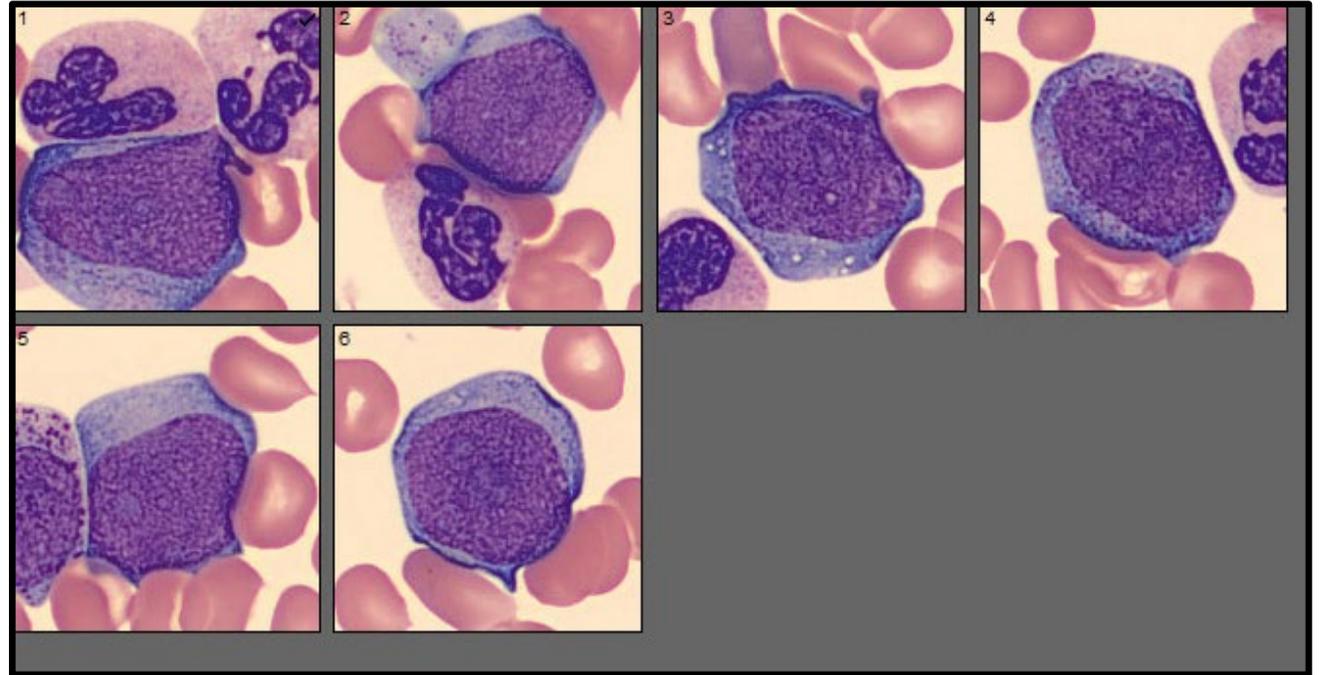
Band



Neutrophil

Blast

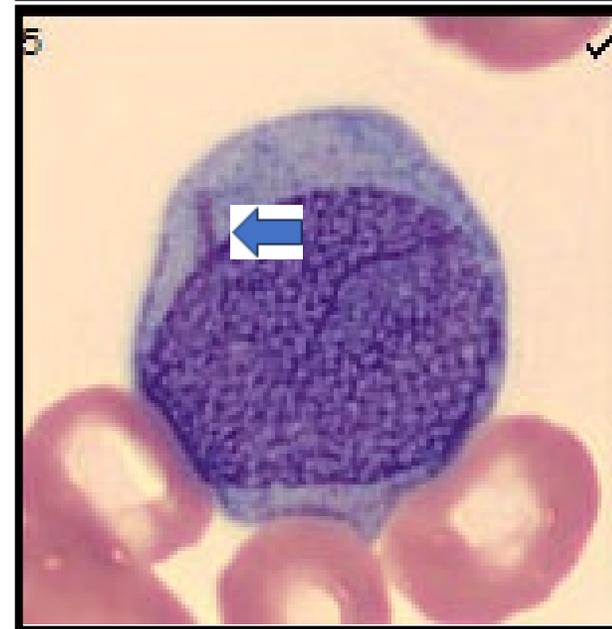
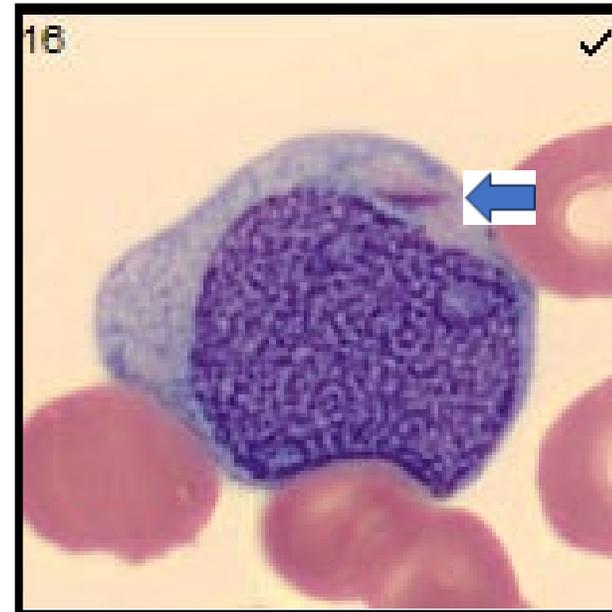
- Not seen in peripheral blood under normal conditions
- Larger cell, 15 μm , with large nucleus
- Fine chromatin, nucleolus
- Cytoplasm pale blue
- High N/C ratio
- GCSF therapy
- Large numbers seen in acute leukemia, high risk MDS, myeloid neoplasms



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

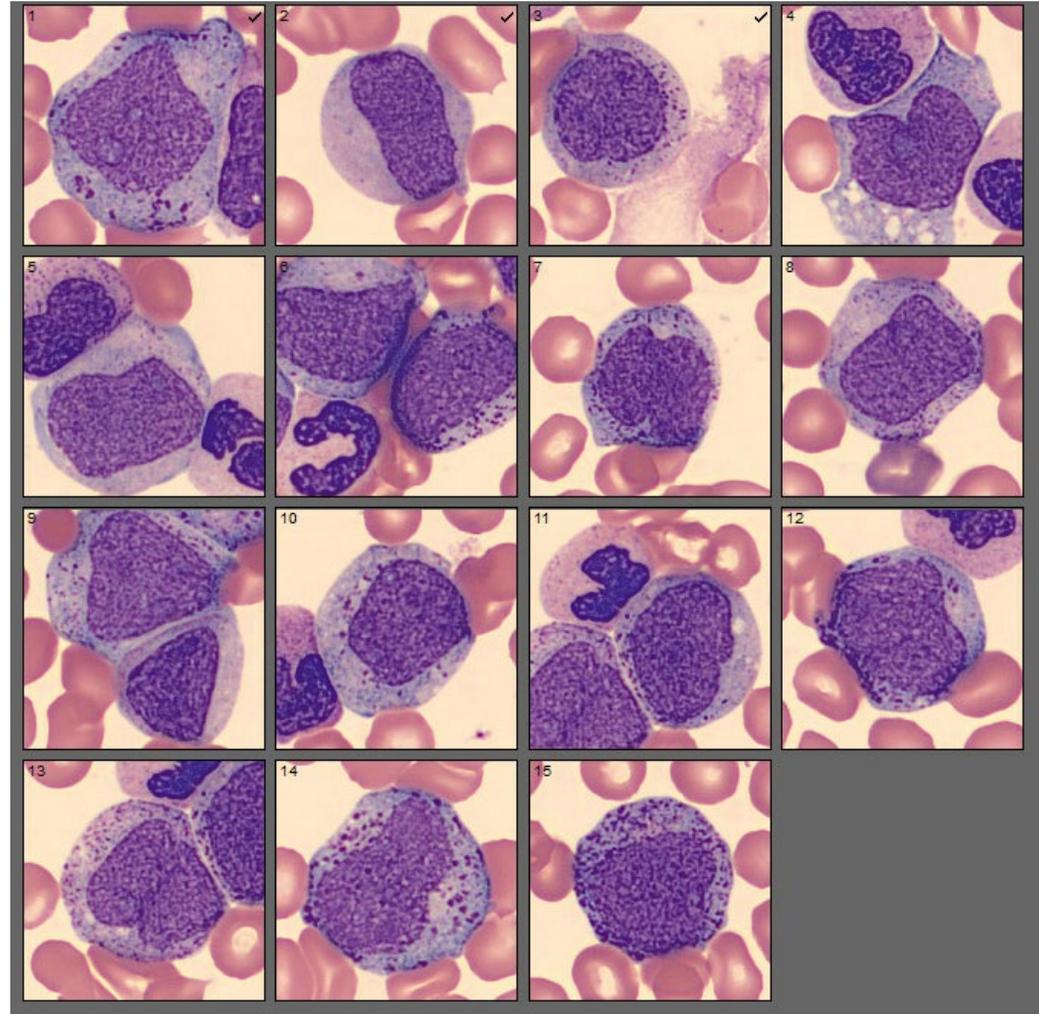
Blast with Auer rods

- Auer rods are rod like inclusions in the cytoplasm of myeloid cells
- Represent abnormal azurophilic granules
- Seen in acute myeloid leukemia, myelodysplastic syndrome



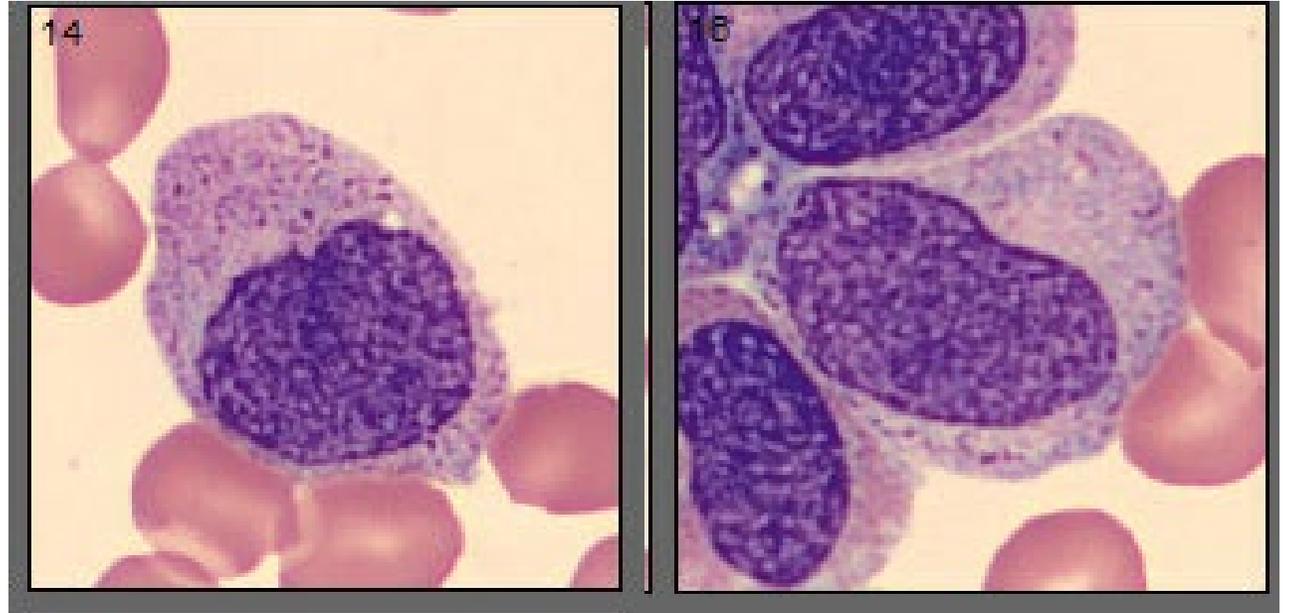
Promyelocytes

- Larger than the blast
- Nucleus slightly eccentric
- Nucleolus may be visible
- Chromatin still immature
- Increased amount of basophilic granular cytoplasm
- Numerous primary granules



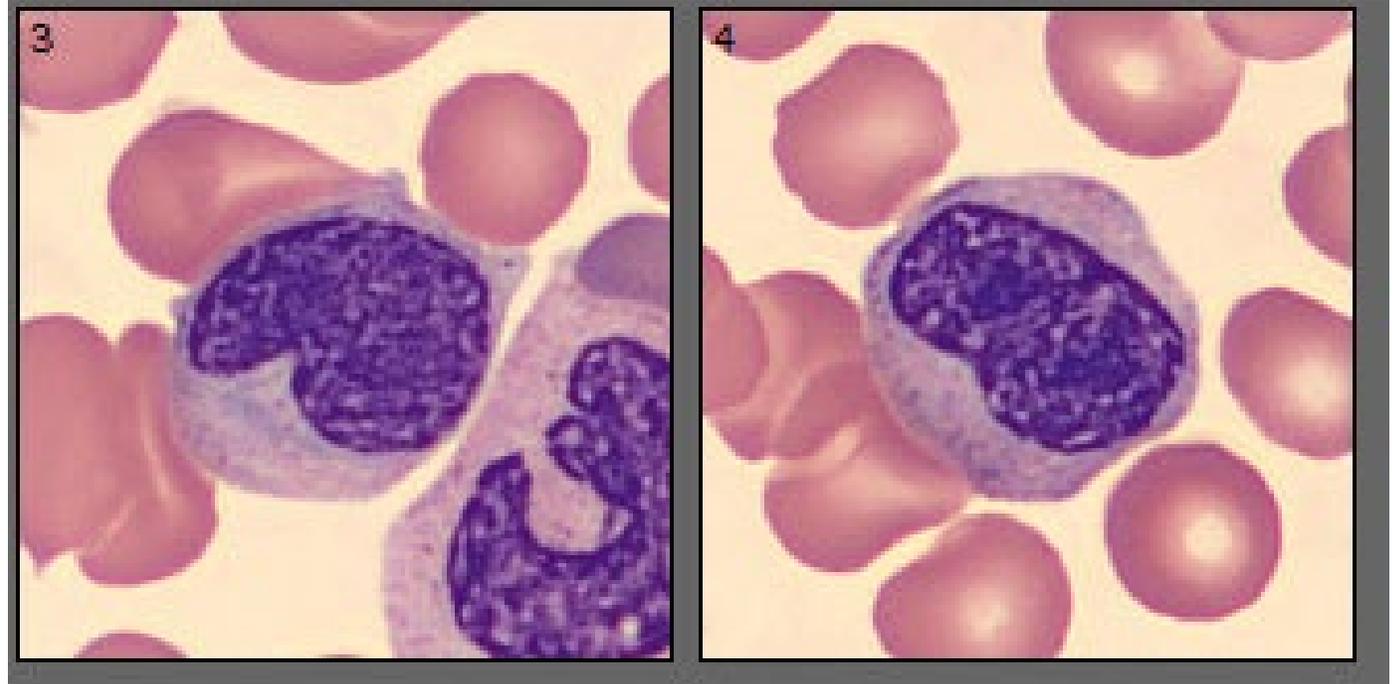
Myelocytes

- Late stage myelocyte has condensed chromatin
- Cytoplasm more pinkish
- Myelocyte is the last stage capable of cell division



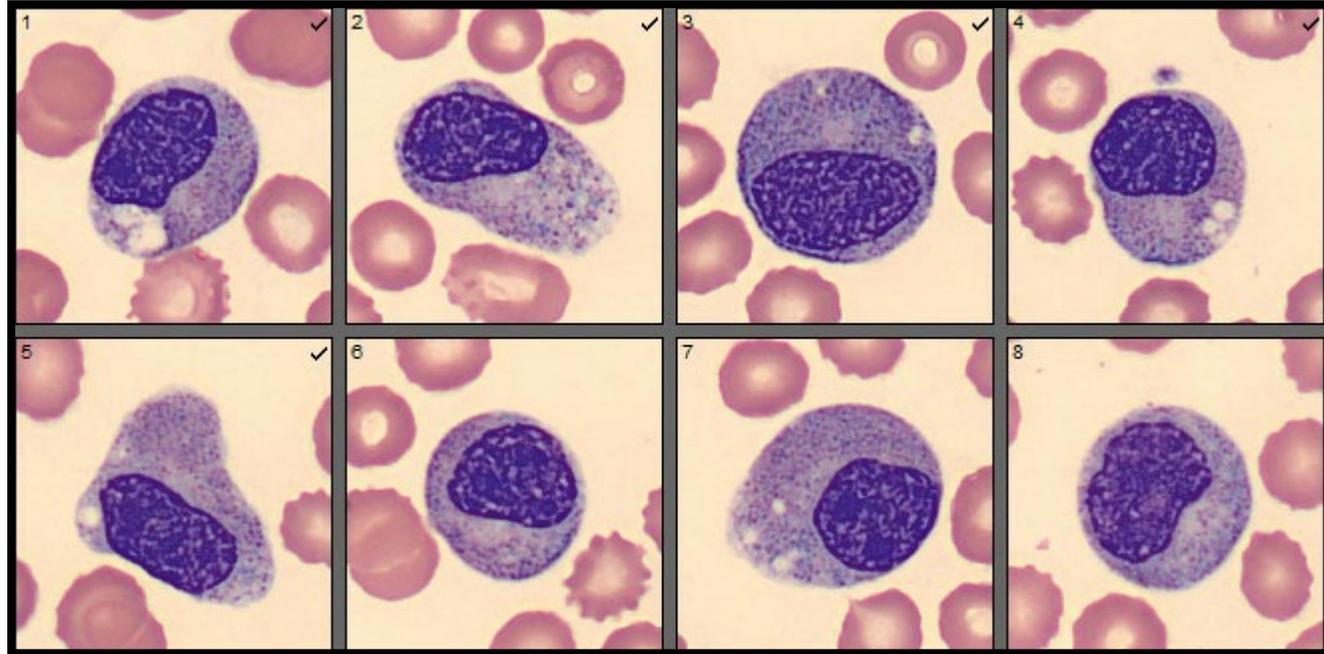
Metamyelocyte

- Indented kidney-shaped nucleus
- Condensed chromatin
- Cytoplasm very similar to myelocyte/neutrophil



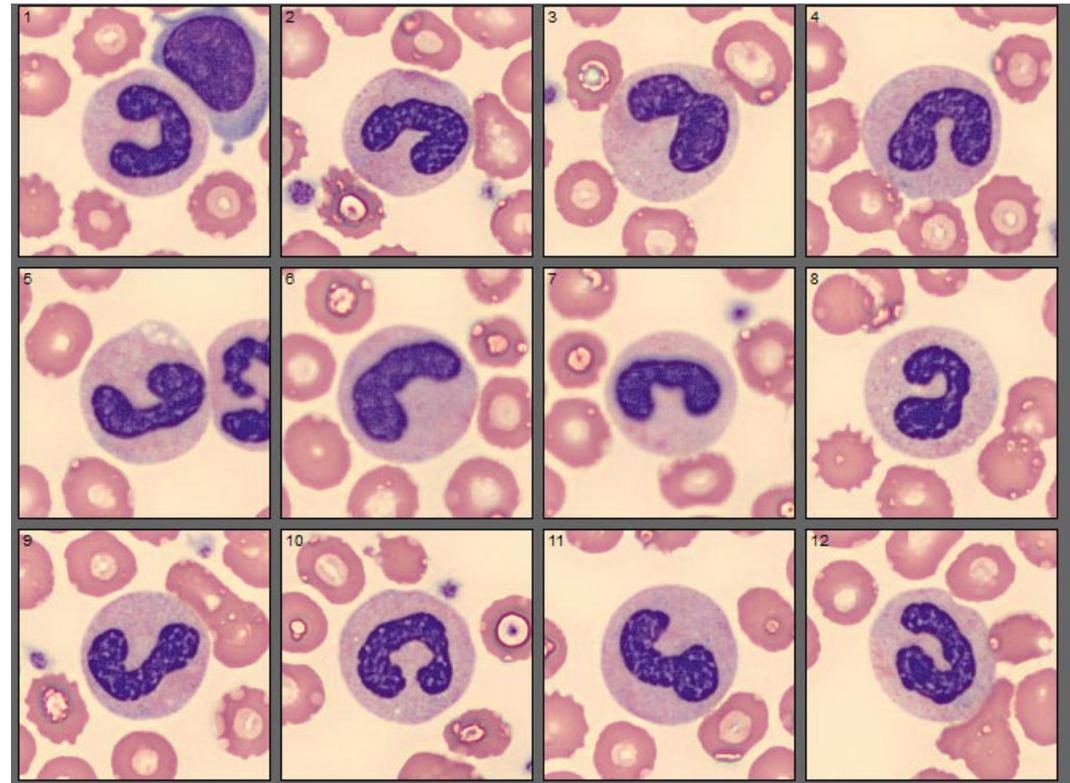
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Myeloid left shift



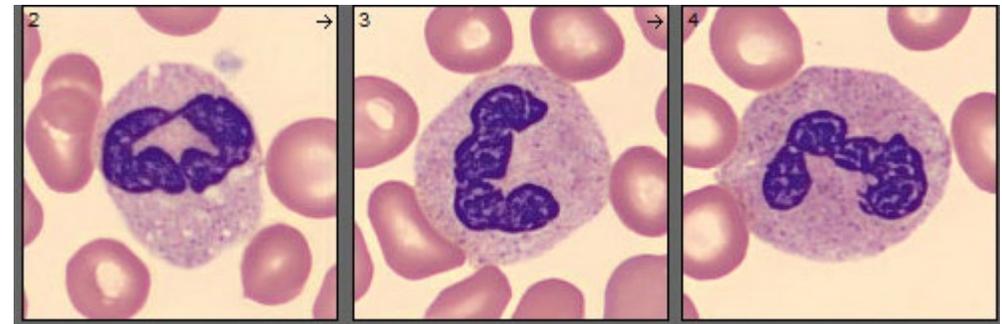
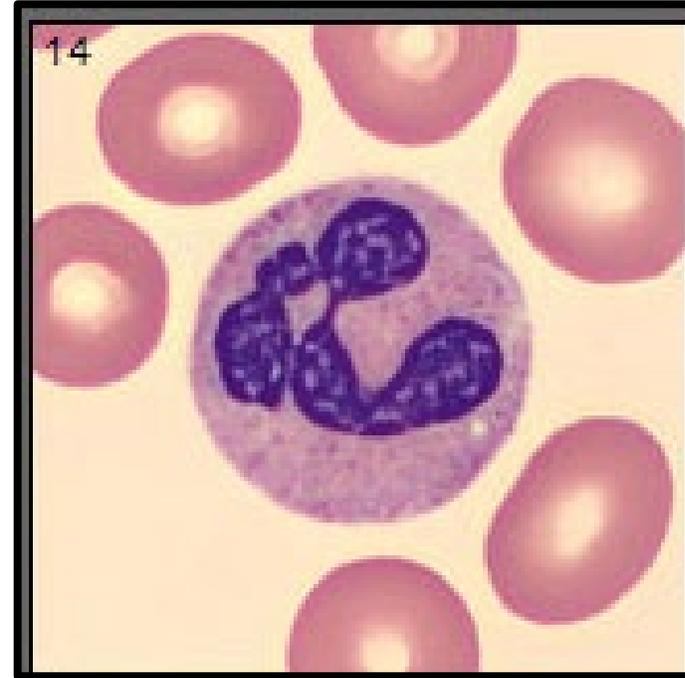
Bands

- Approx. 3% of WBC
- Curved nuclei, U-shaped
- Nucleus starts to develop indentations
- True filaments are not present

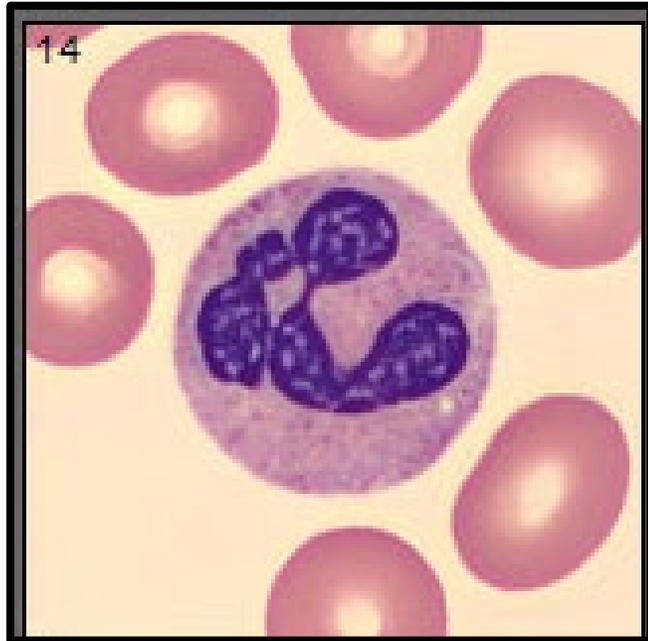


Segmented neutrophils

- Mature neutrophils, avg. 12 μm diameter
- Irregular shaped nucleus, looks like letters E, Z or S
- Segmented nuclear lobes, 2-5 lobes, median of 3
- At least 2 lobes are separated by a filament
- Coarse chromatin
- Abundant cytoplasm with tan pink granules



Segmented neutrophil



Bands

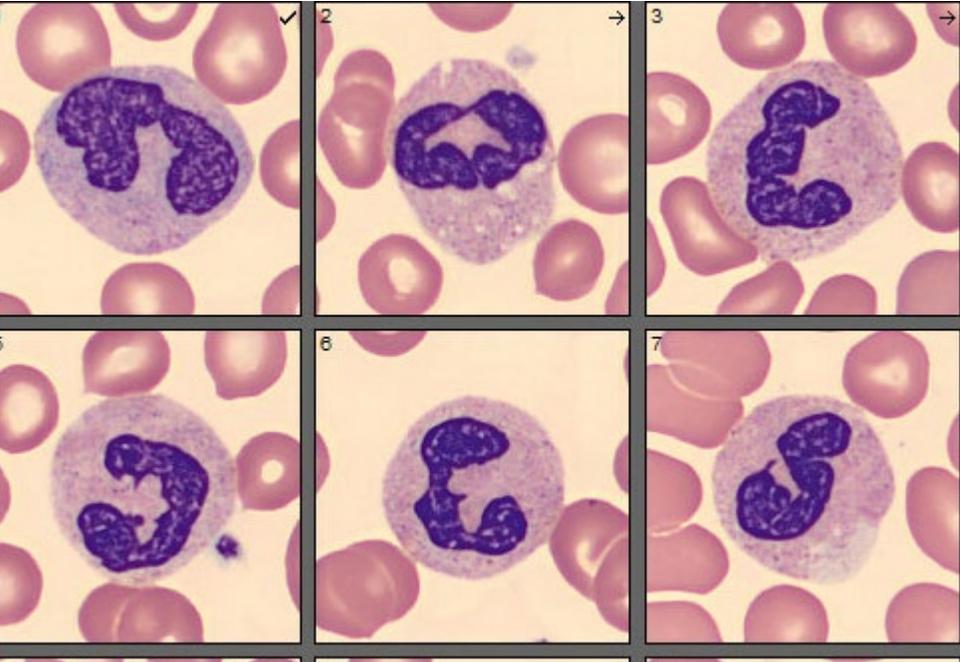


Toxic granulation

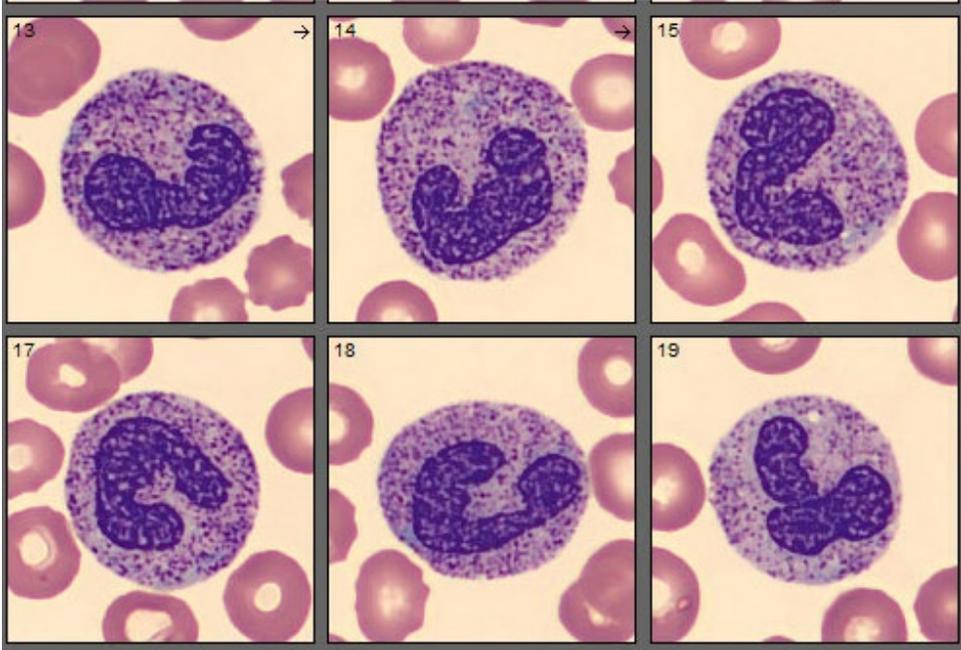
- Seen in reactive and proliferative states
- Toxic granulation seen in infection, inflammation, GCSF therapy
- More basophilic and larger granules



Normal neutrophils

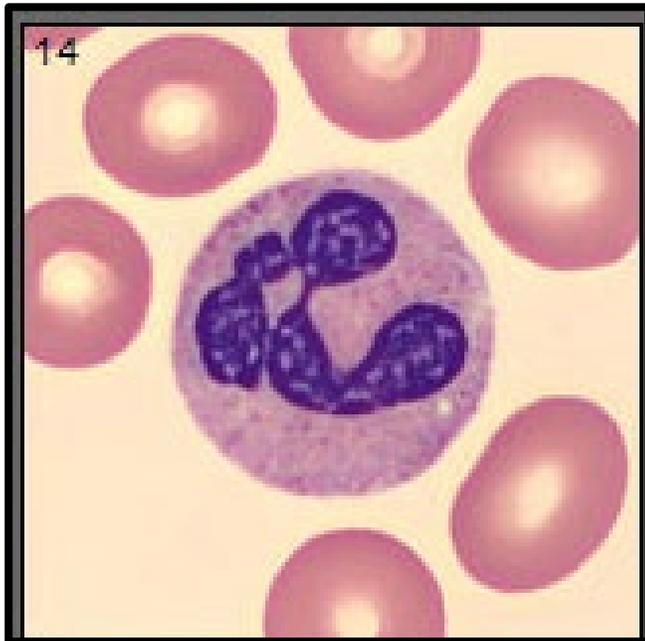


Toxic granulation

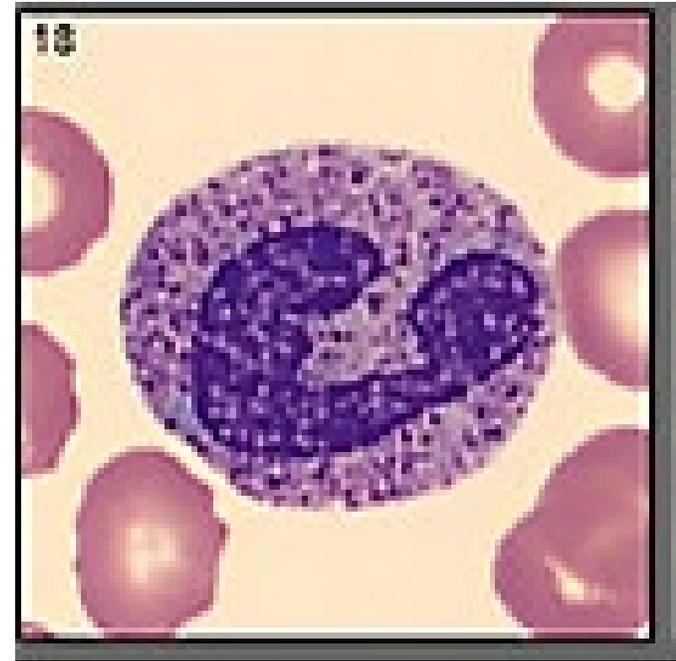


Toxic granulation

Normal segmented neutrophil



Neutrophil with toxic granulation



Dohle Bodies

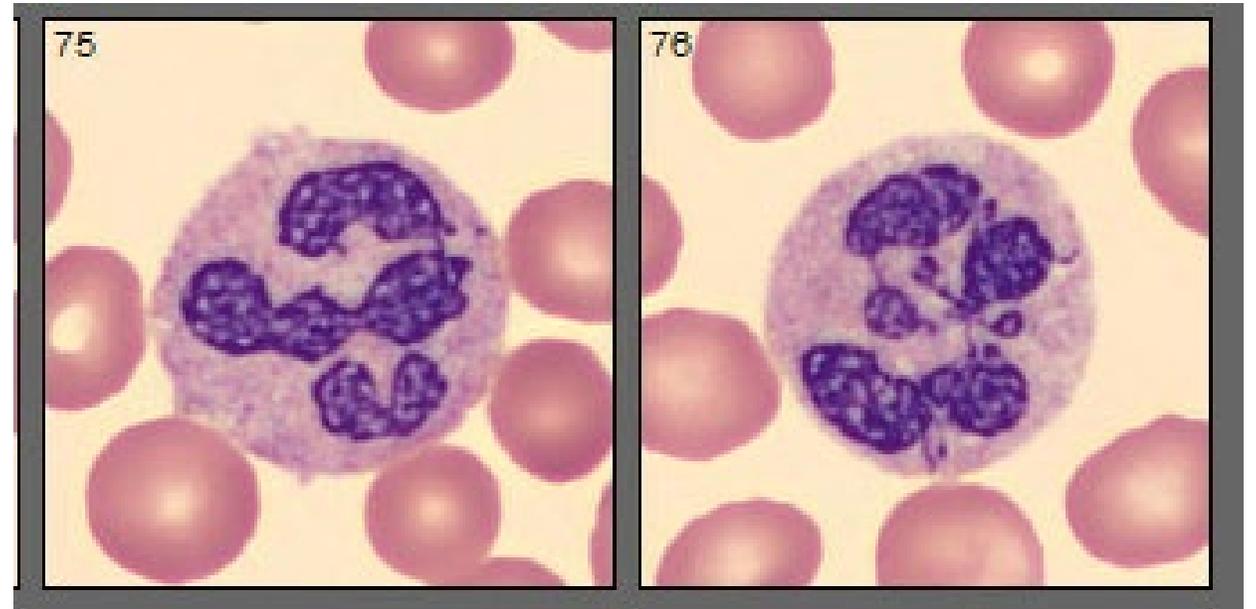
Dohle bodies represent endoplasmic reticulum and ribosomes

Faint, small pale blue inclusions at the periphery of the cell

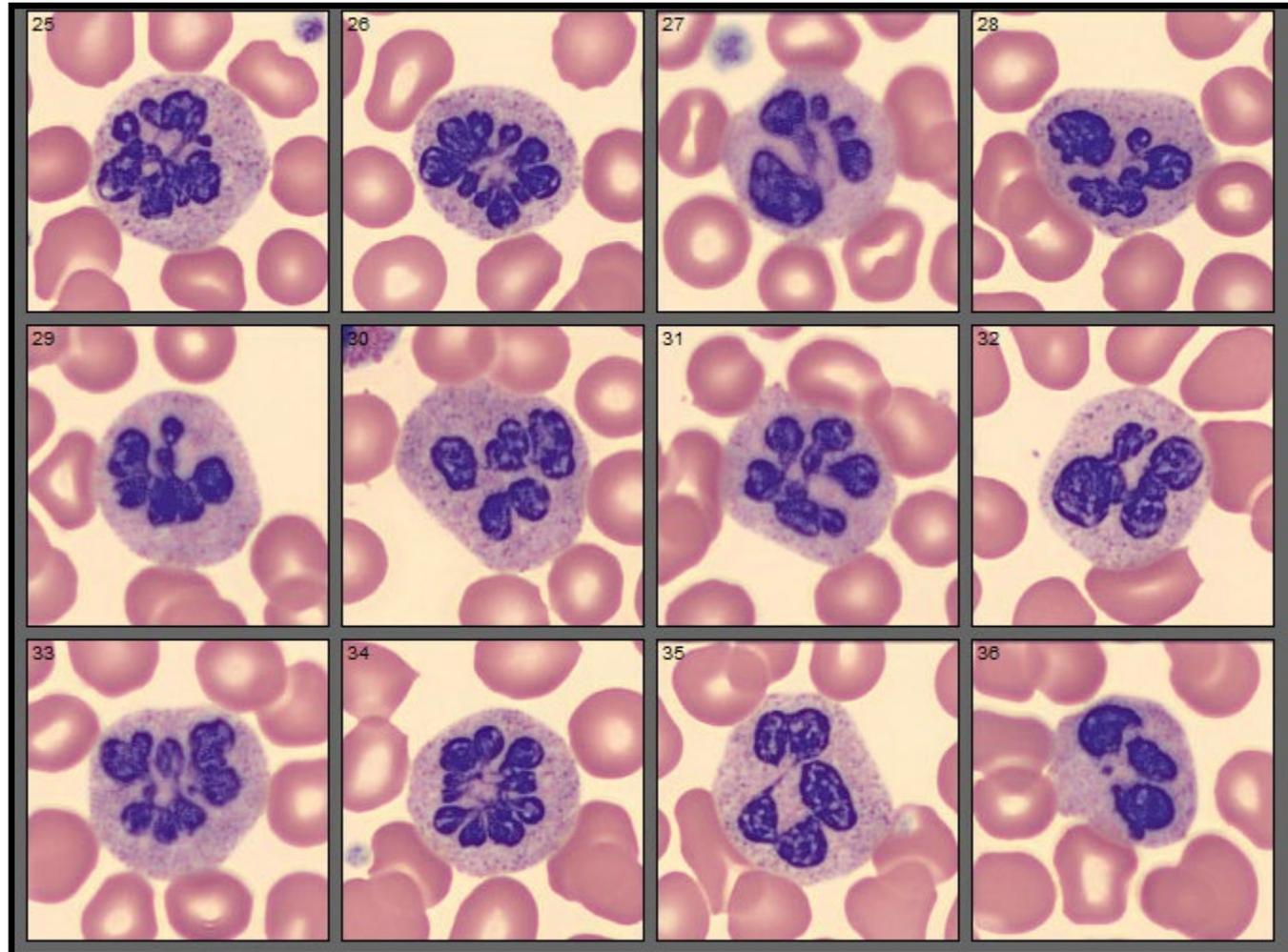


Hyper-segmented neutrophils

- Normally no more than 5% of neutrophils have 5 lobes
- Six or more nuclear lobes
- Seen in Vitamin B12 deficiency



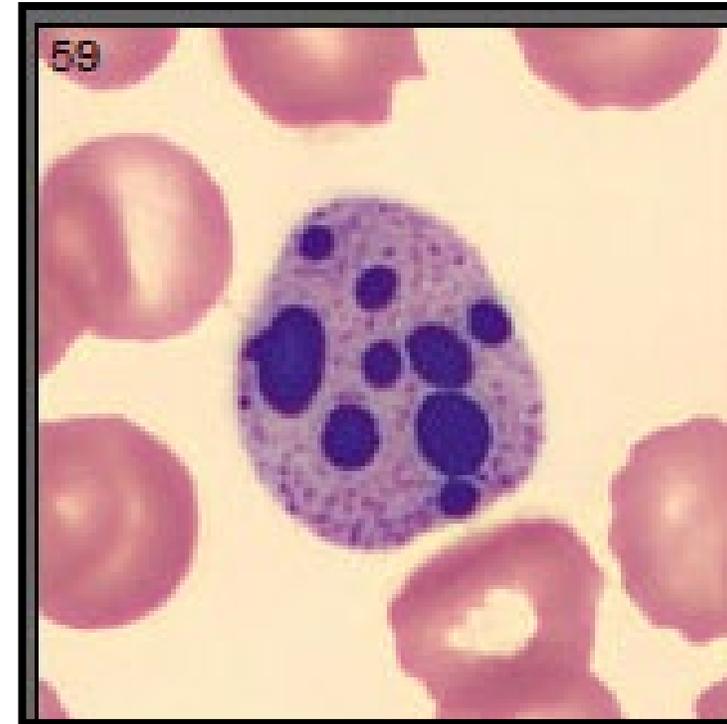
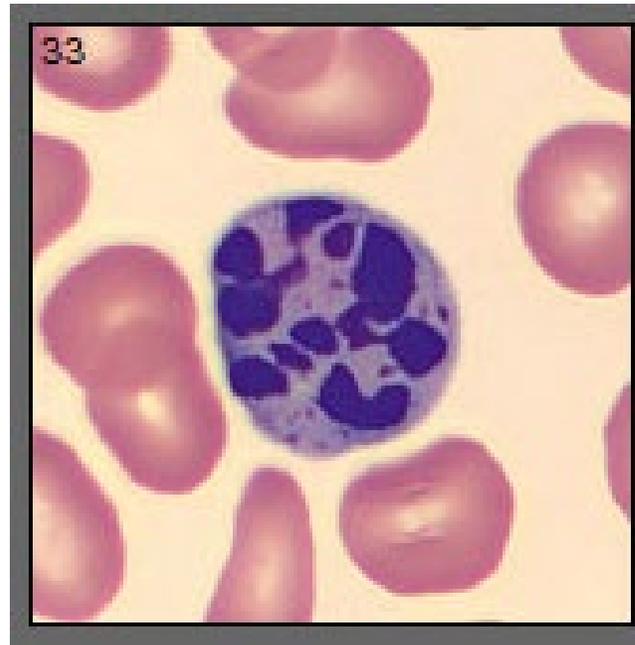
Hyper-segmented neutrophils



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

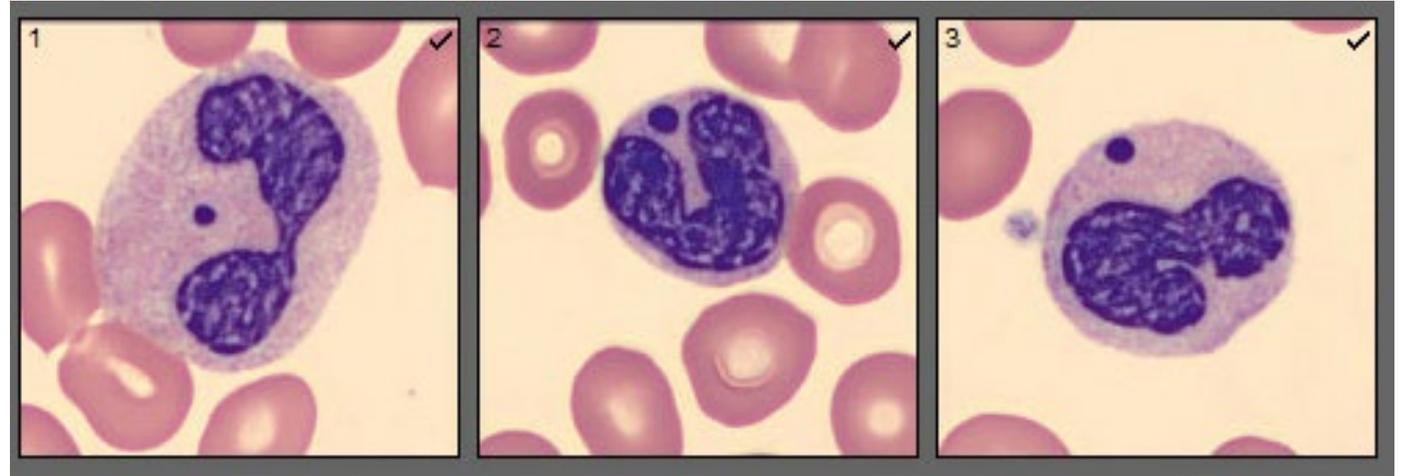
Karyorrhexis

- Fragmented nuclear material
- Dying cell



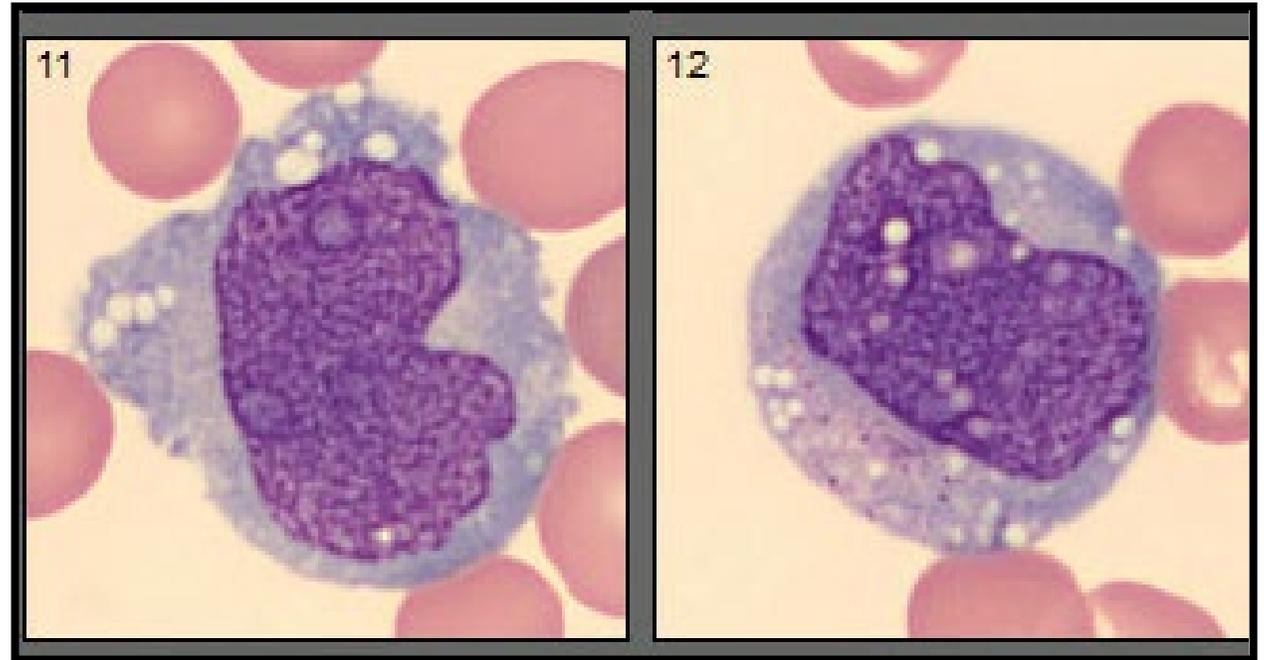
Howell-Jolly like inclusions

- Sharp dot-like dark purple inclusions in the cytoplasm
- Post-kidney transplant
- Represent nuclear material



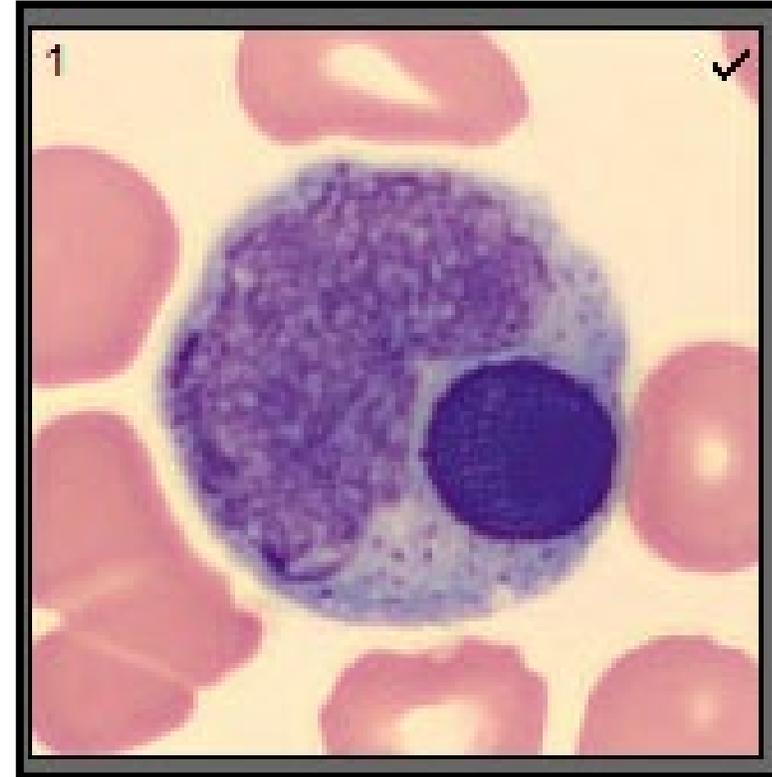
Monocytes

- Largest cell of normal blood, 14-20 um
- 2-3 times the diameter of an RBC
- Avg. 4% of WBC
- Single nucleus with indentation, may be round or oval
- Fine chromatin
- Abundant gray-blue cytoplasm, ground glass appearance with red-purple granules and vacuolation
- Vacuoles are phagocytic and may contain debris, pigment, bacteria or ingested cells



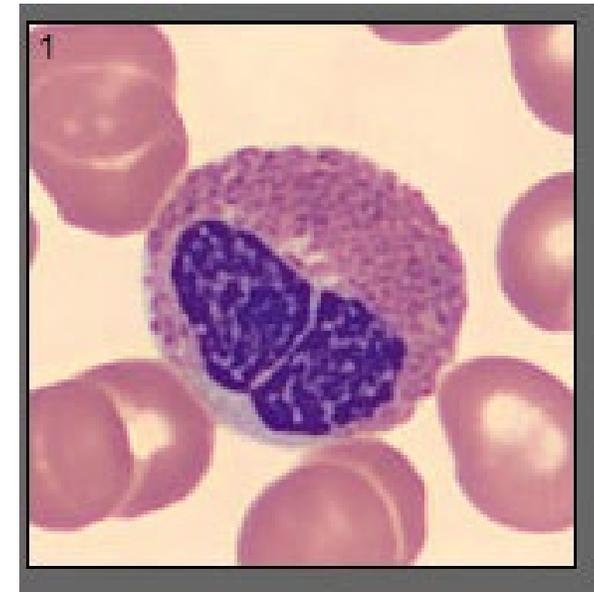
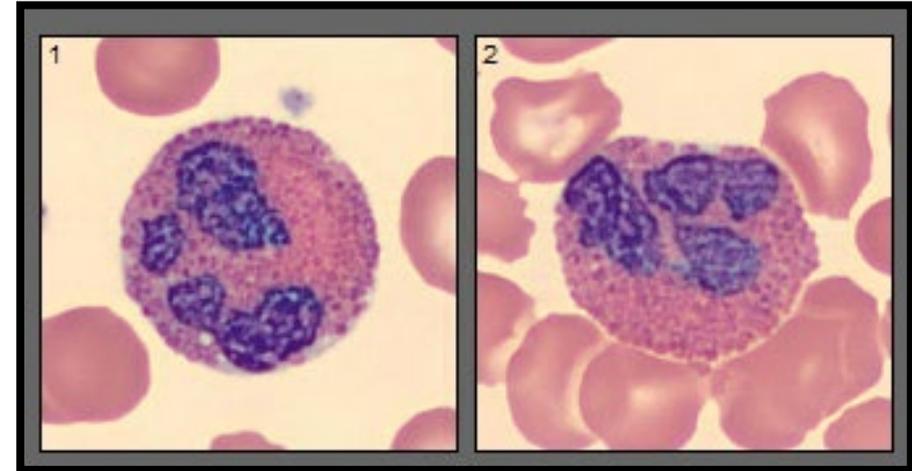
Hemophagocytosis

- Evidence of phagocytosis in monocytes in blood films is pathologic
- Indicates the presence of active infection



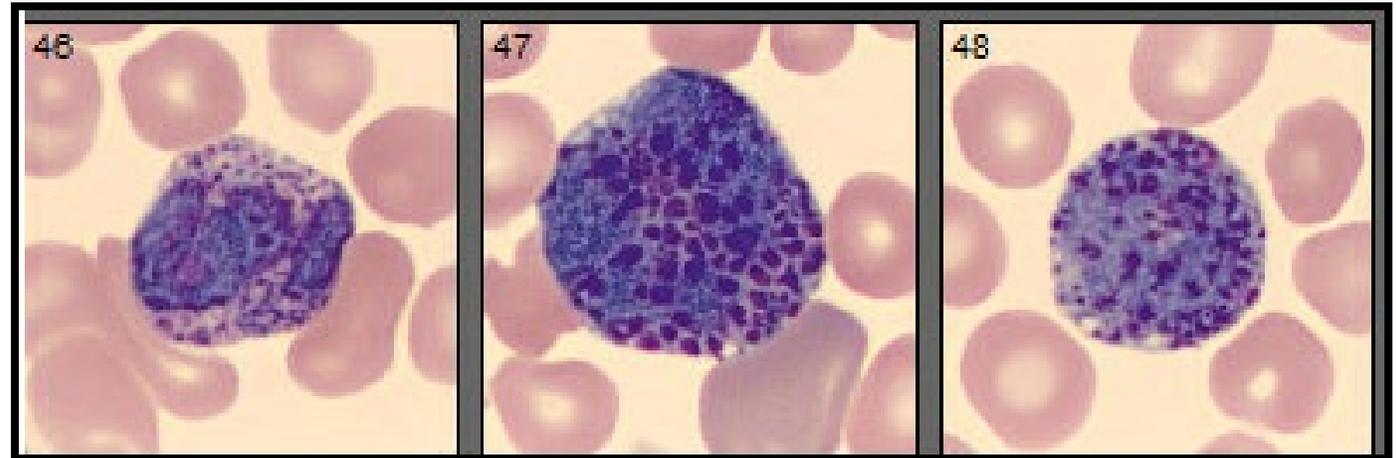
Eosinophils

- Average diameter 13 μm
- Approximately 3% of WBC count
- Structure is similar to neutrophils
- Cytoplasm contains larger red-coral granules
- Nucleus usually has two lobes, can be more



Basophils

- Approx. 0.5% of WBC
- Basophils have large, dark purple-black granules
- Granules typically overlay the nucleus and obscure it
- Reactive basophilia can be seen in diabetes, hypersensitivity reactions, myxedema and ulcerative colitis
- Neoplastic condition with basophilia, CML



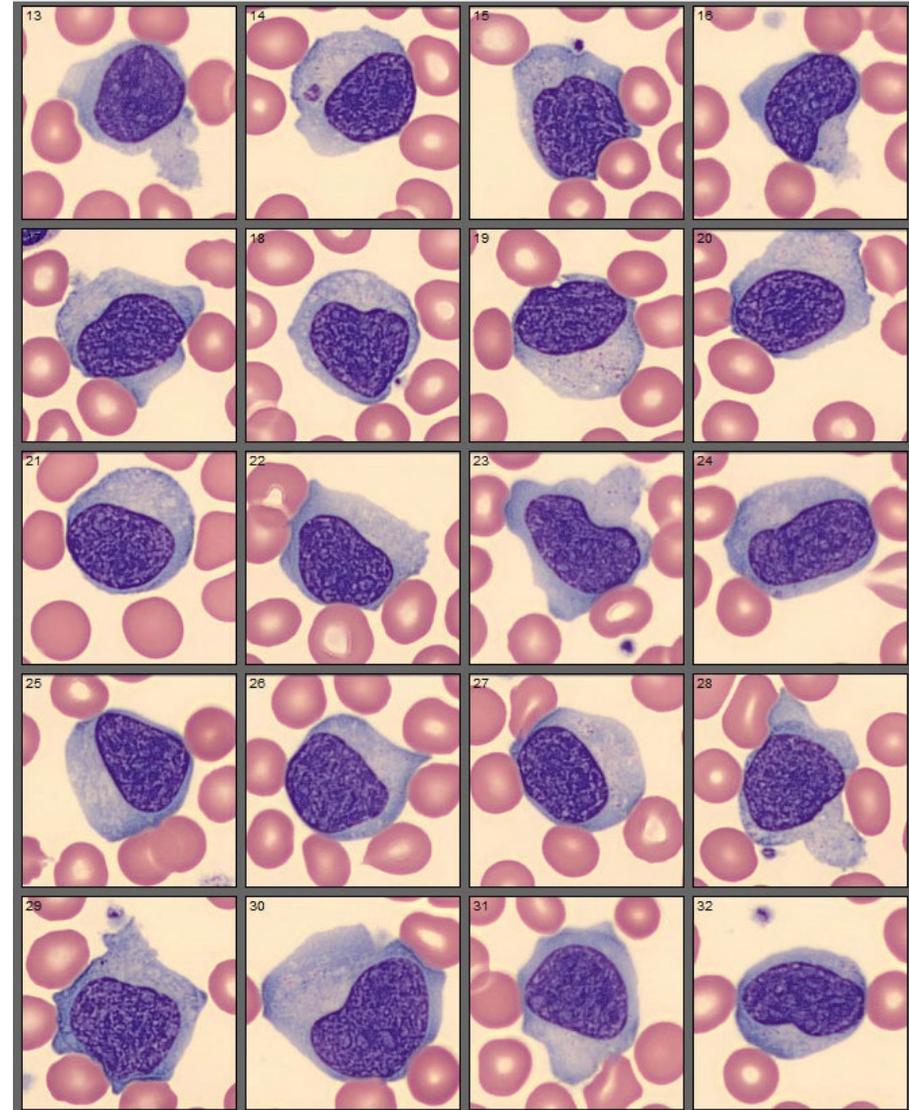
Lymphocyte

- Small lymphocytes approximately the same size as RBCs, 6-10 μm
- Single nucleus with clumped chromatin
- Scant blue cytoplasm

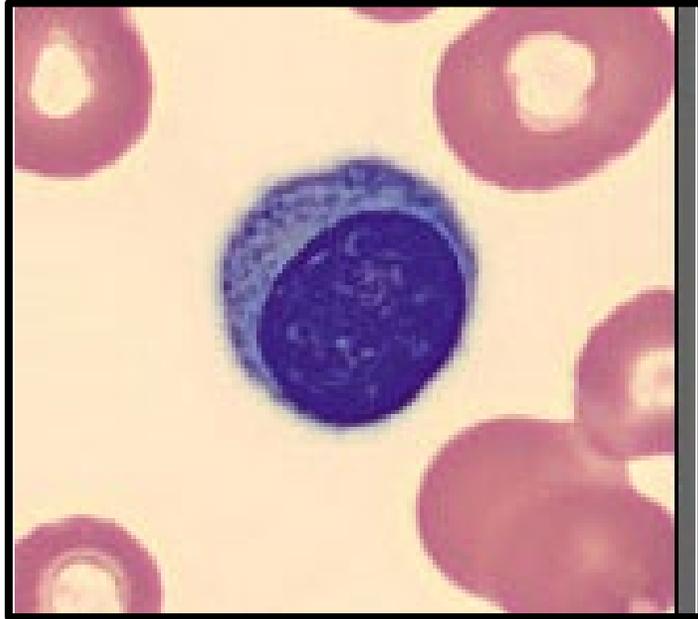


Reactive lymphocytes

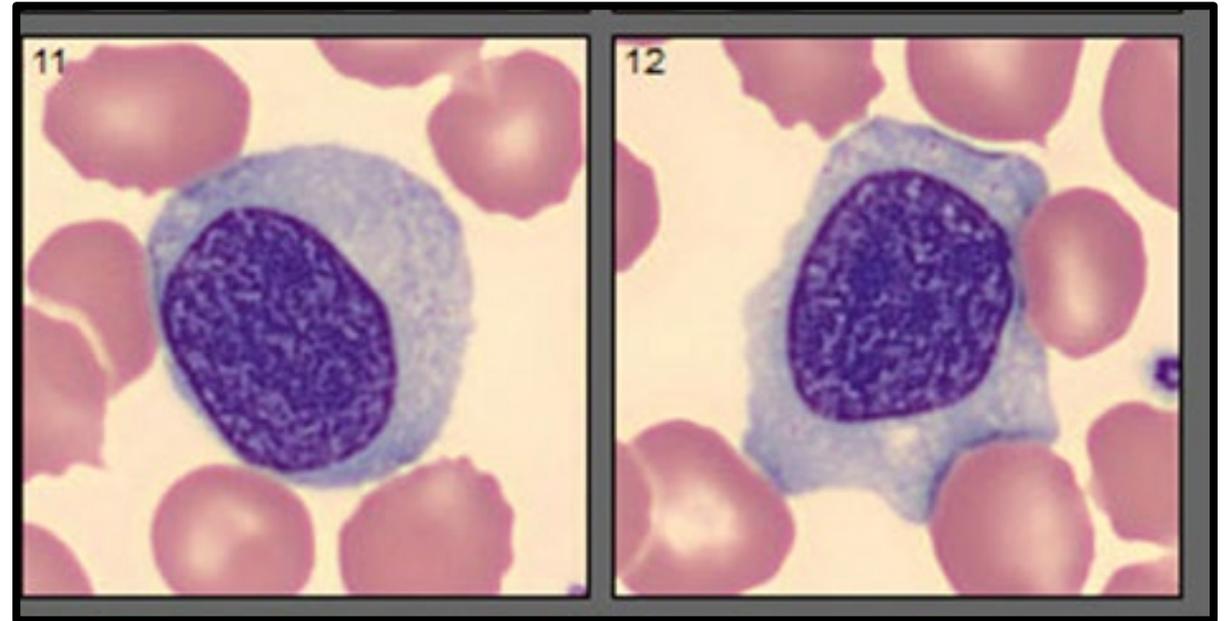
- Relatively larger cells, 12-15 um
- Less dense nuclei
- Abundant cytoplasm
- May or may not contain cytoplasmic granules
- Seen in viral infections



Lymphocyte

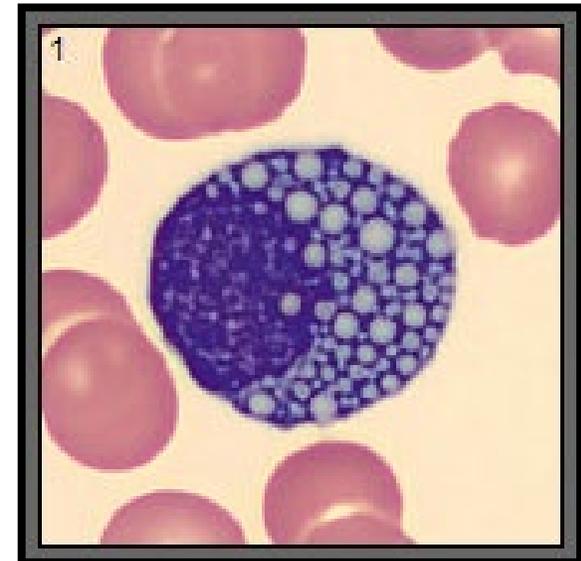
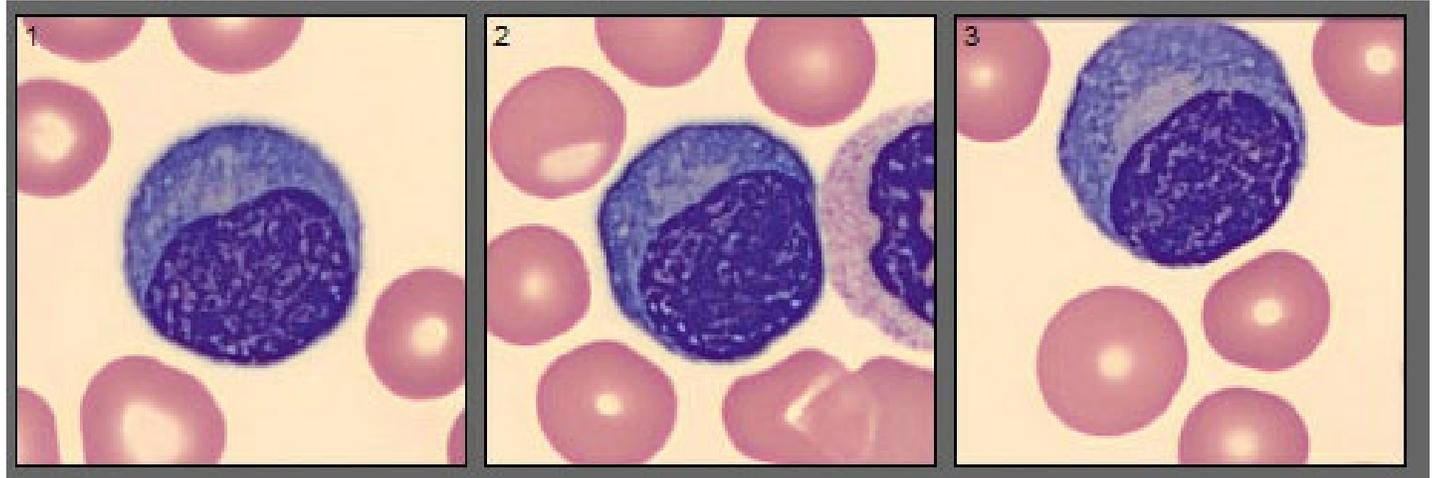


Reactive lymphocytes

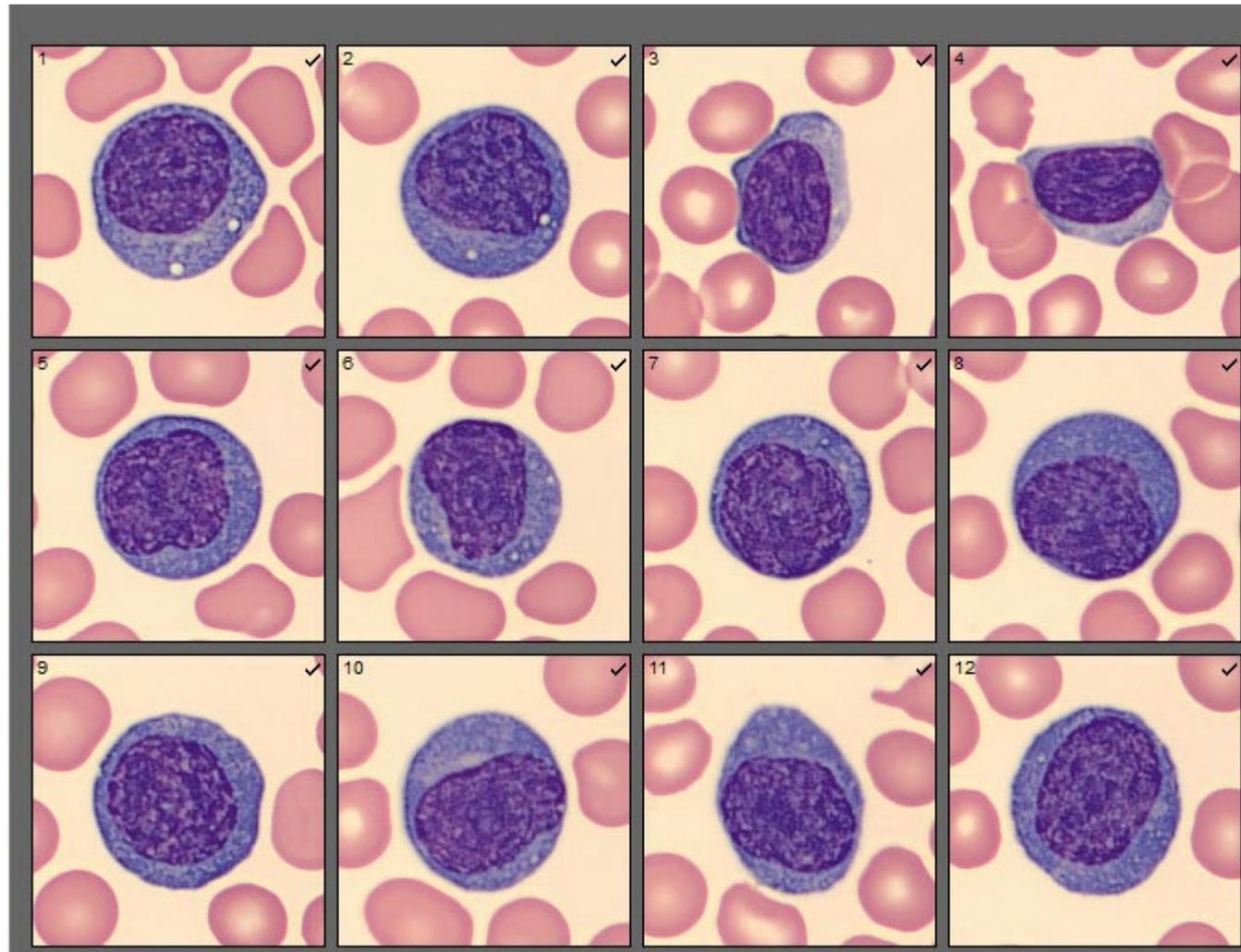


Plasma cell

- Plasma cells can be seen rarely in normal individuals
- Single, round eccentric nucleus with clumped chromatin
- Abundant blue cytoplasm
- Peri-nuclear clear zone
- Plasma cell myeloma
- Mott cell

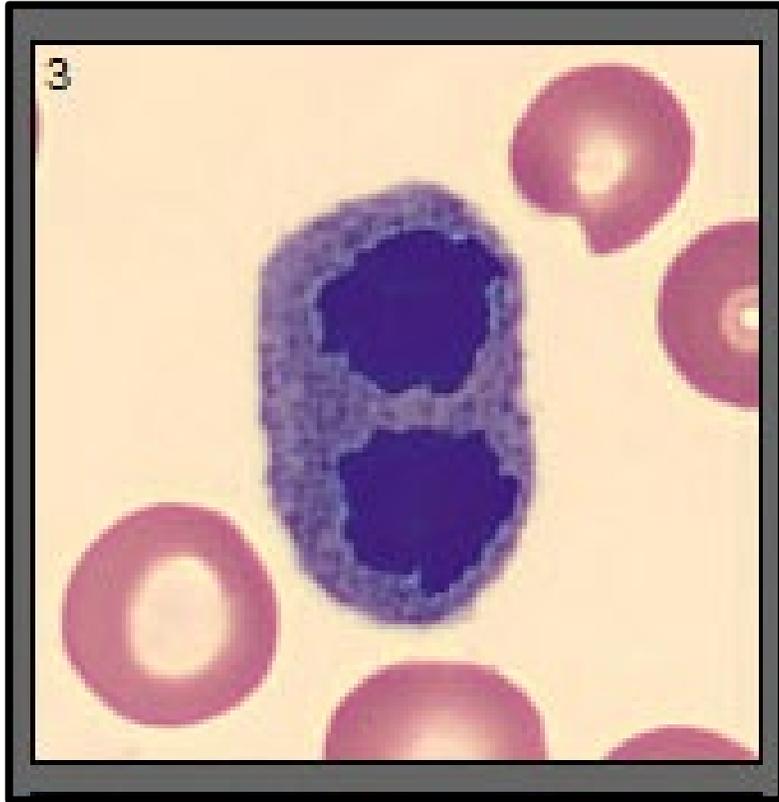


Dengue fever



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Mitotic activity



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine



Alamy.com

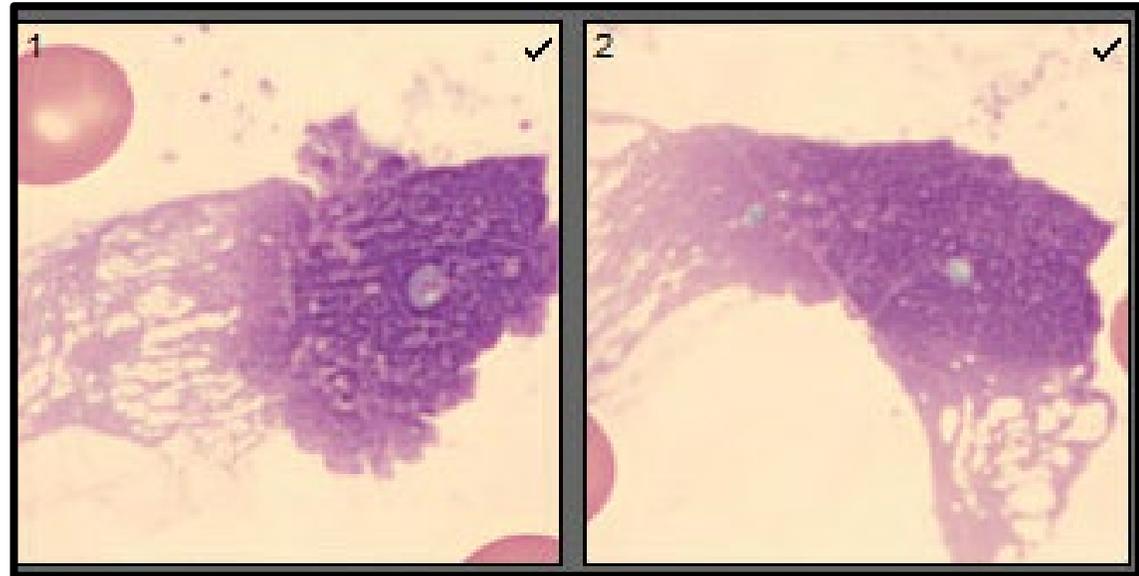
Ruptured cell

- Damaged or broken leukocytes
- can be seen in normal blood films

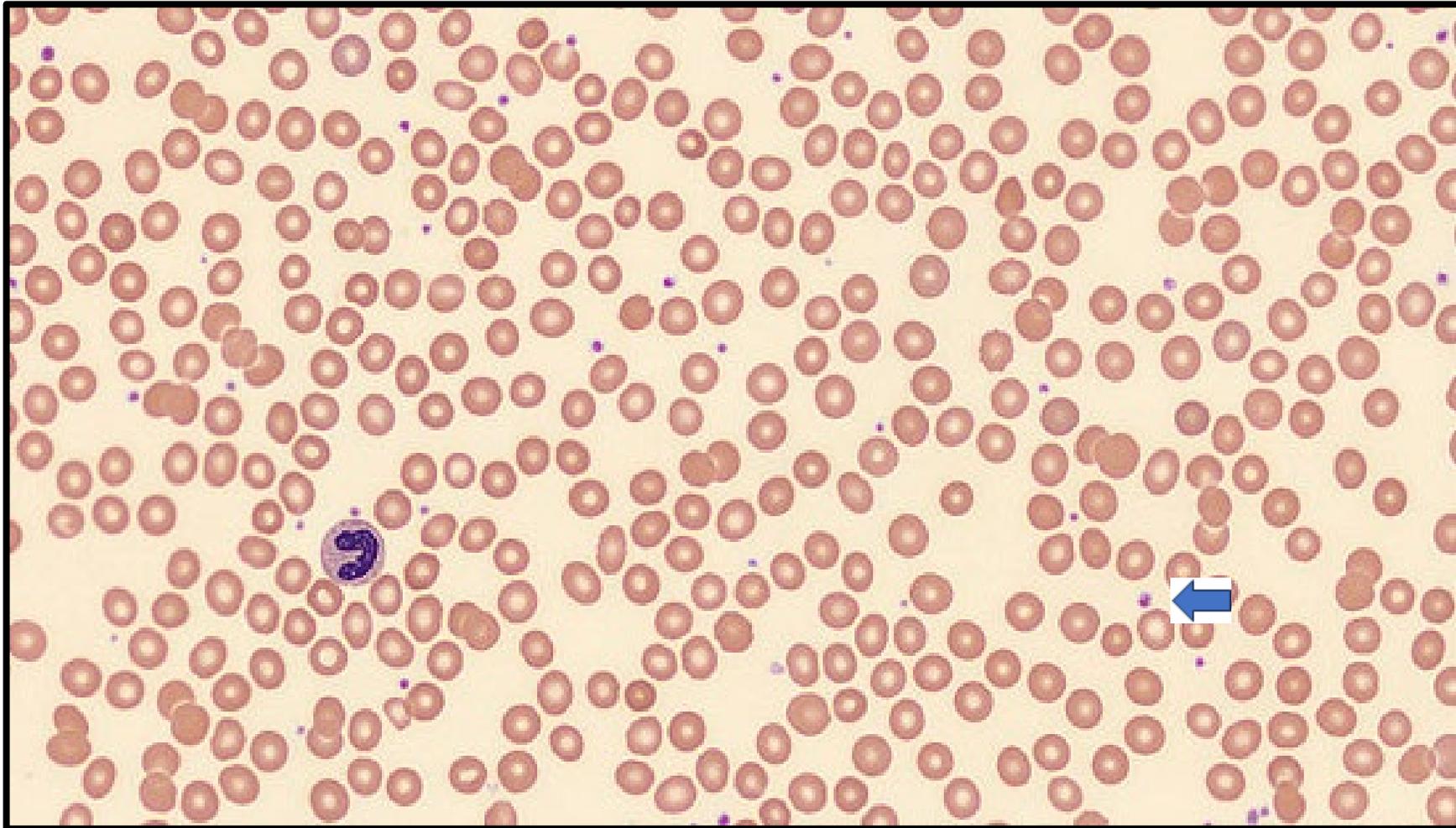


Smudge

- Nuclear remnant from a broken white blood cell
- Abundant smudge seen in chronic lymphocytic leukemia



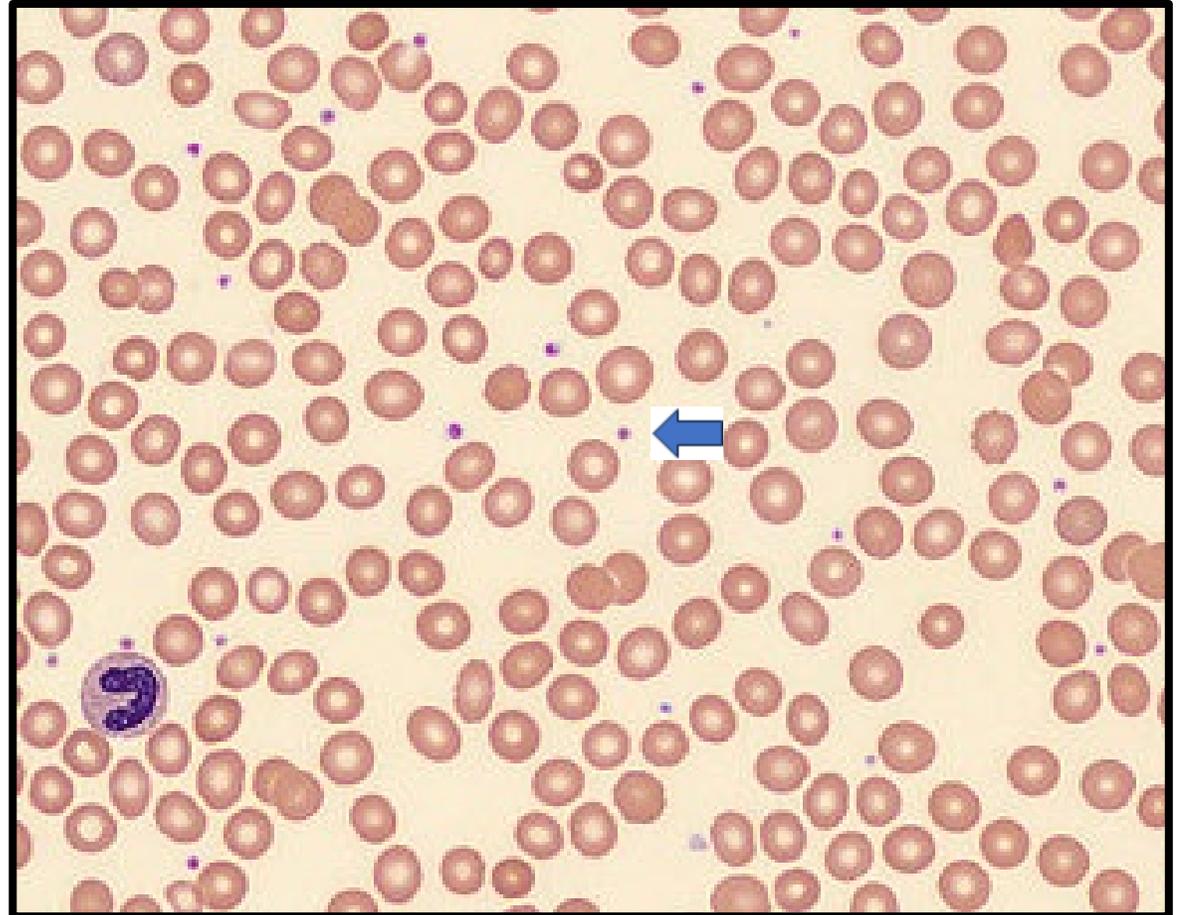
Platelets



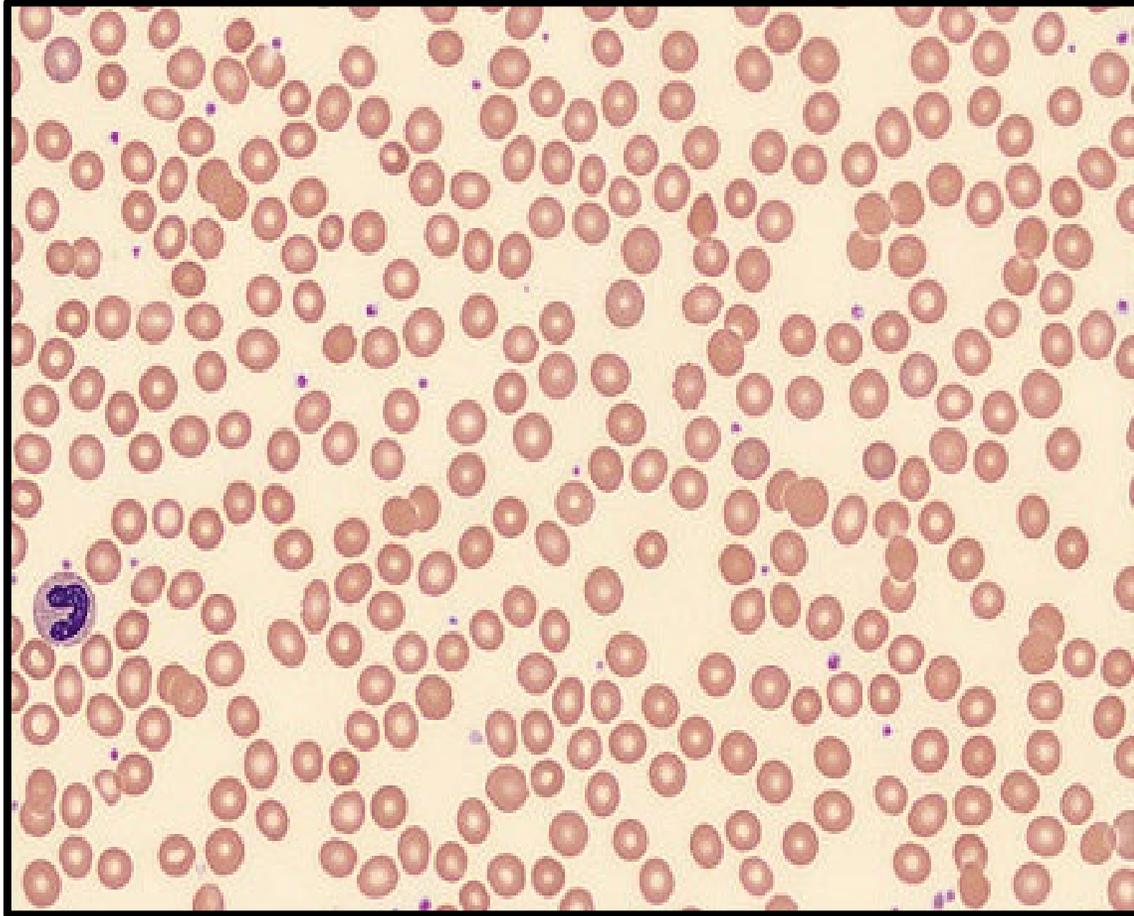
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Platelets

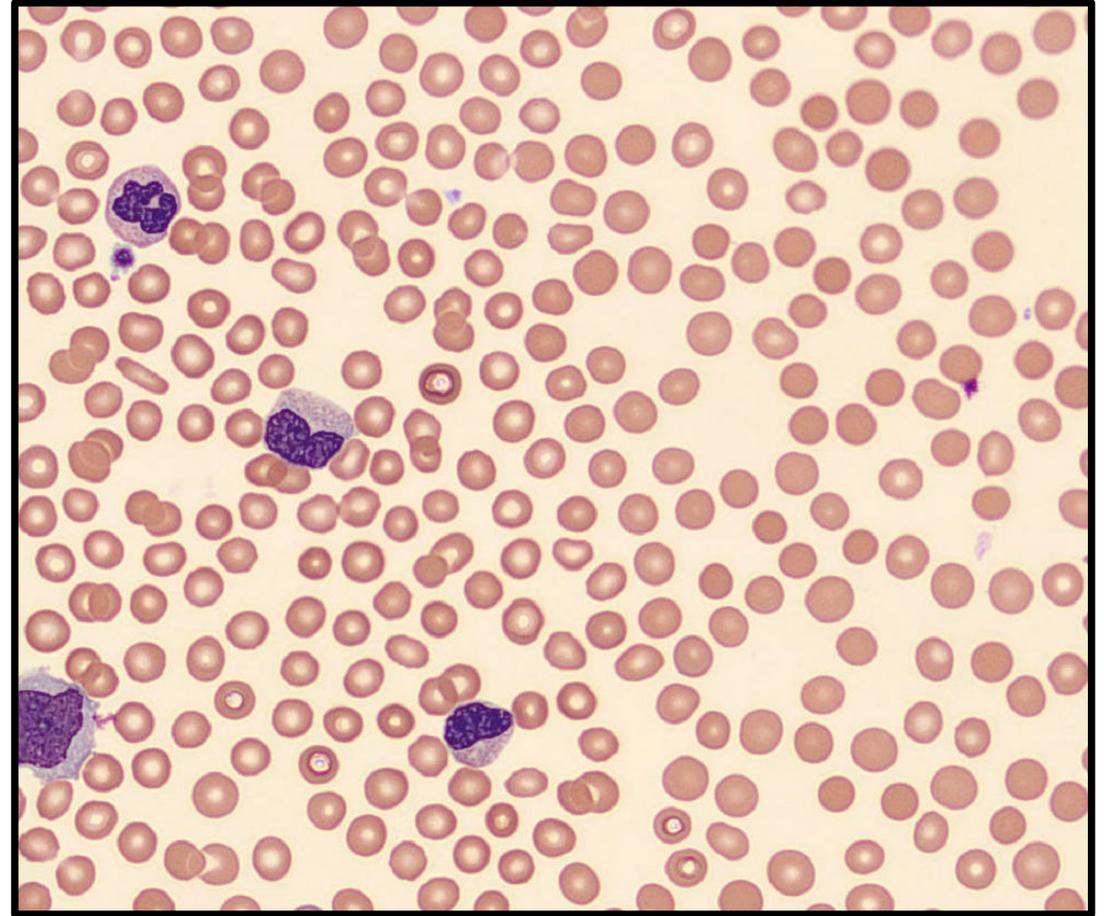
- Anucleated, thin disks, 2 to 4 μm in diameter
- Involved in primary hemostasis, maintenance of vascular integrity and coagulation



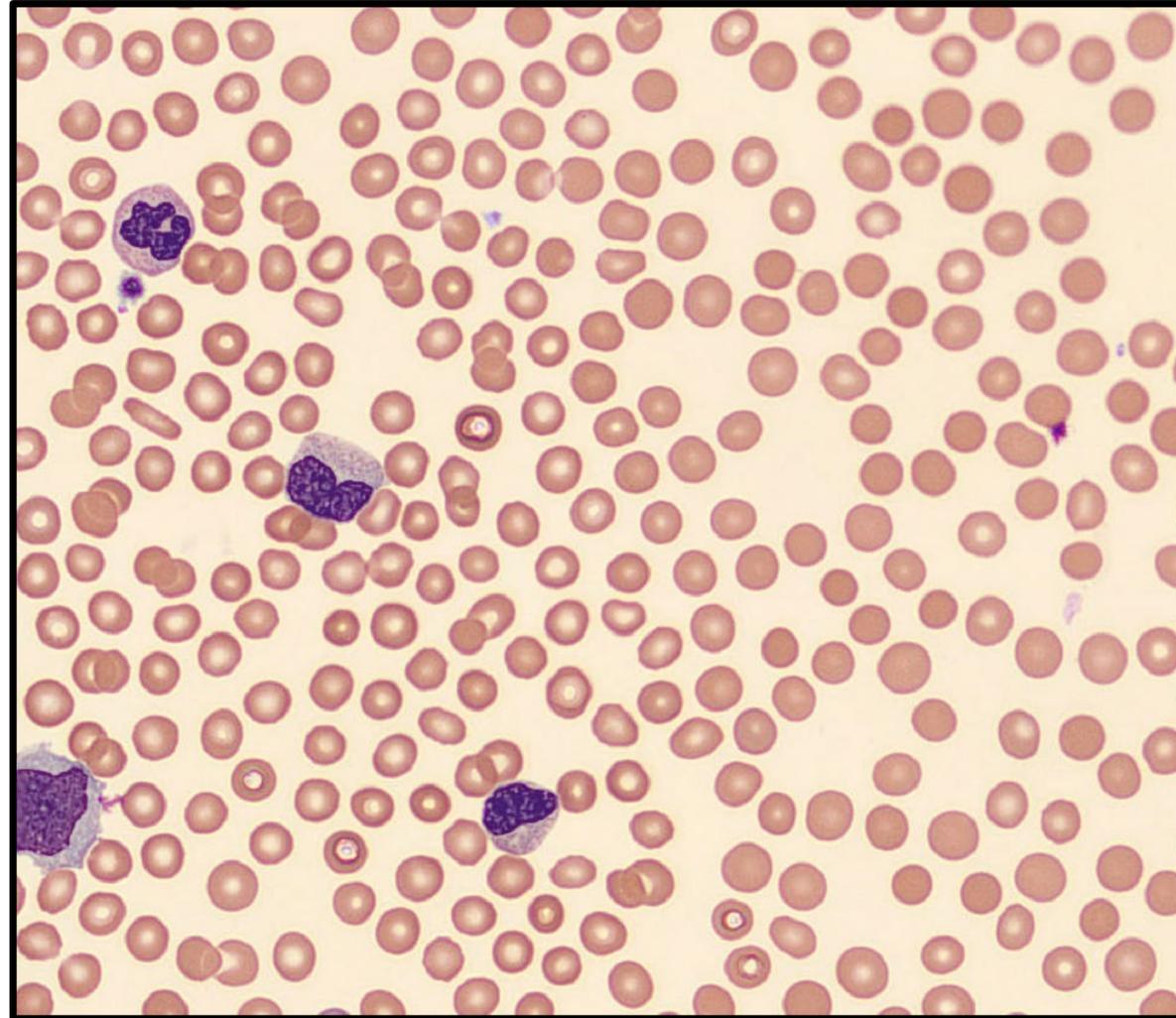
Normal control



Thrombocytopenia

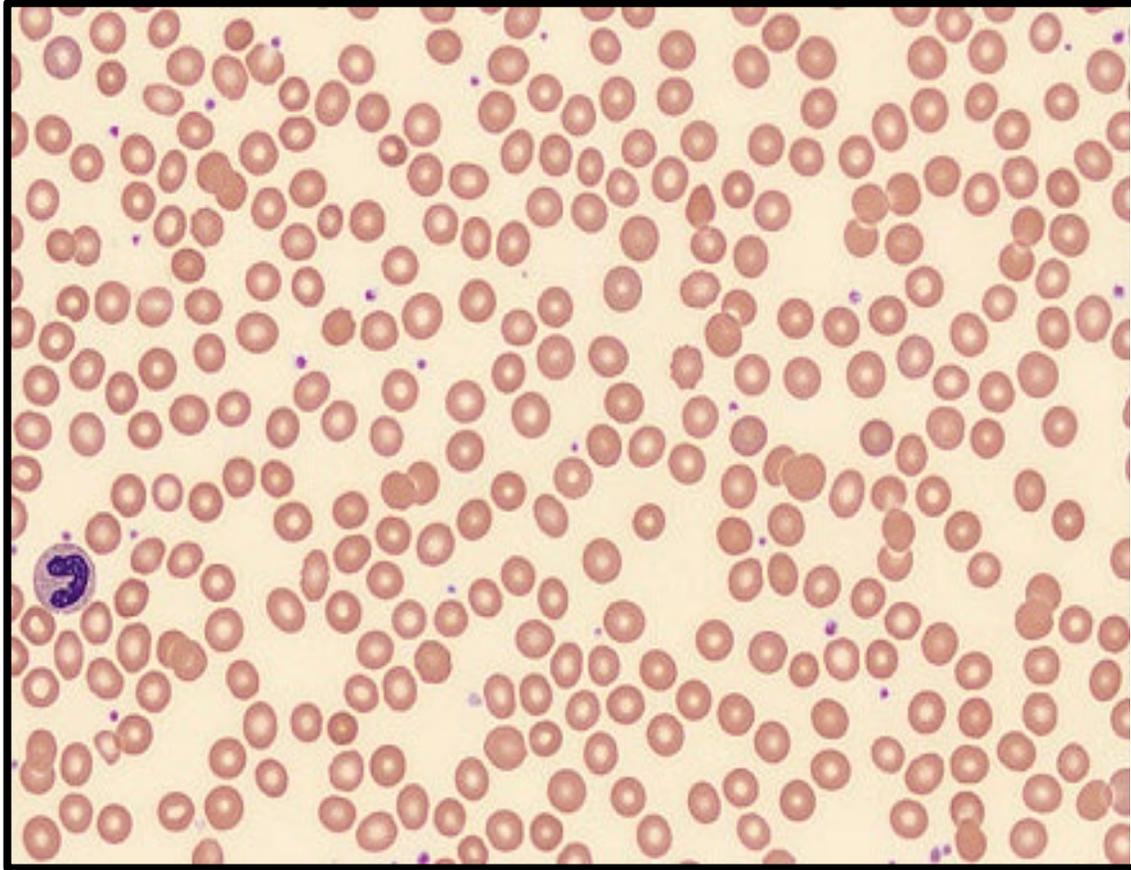


Thrombocytopenia



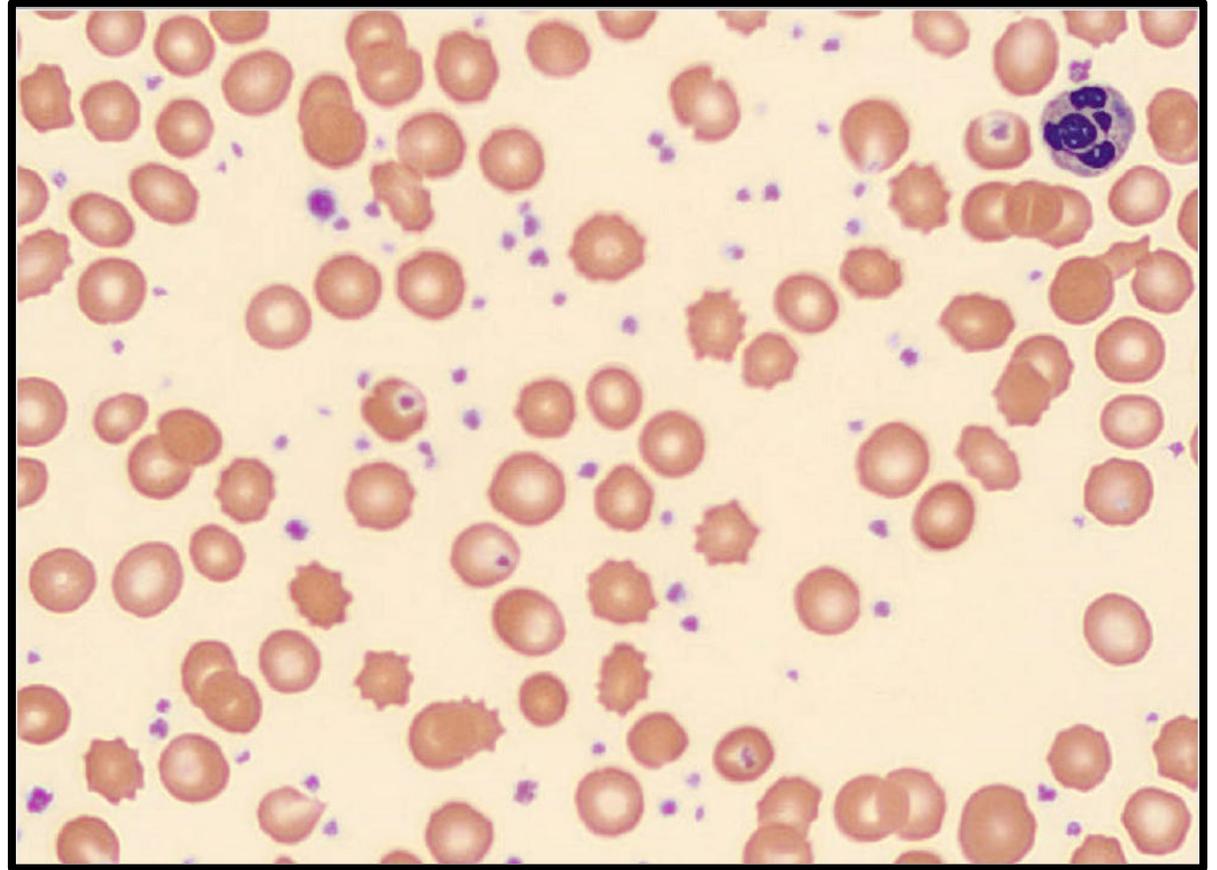
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Normal control



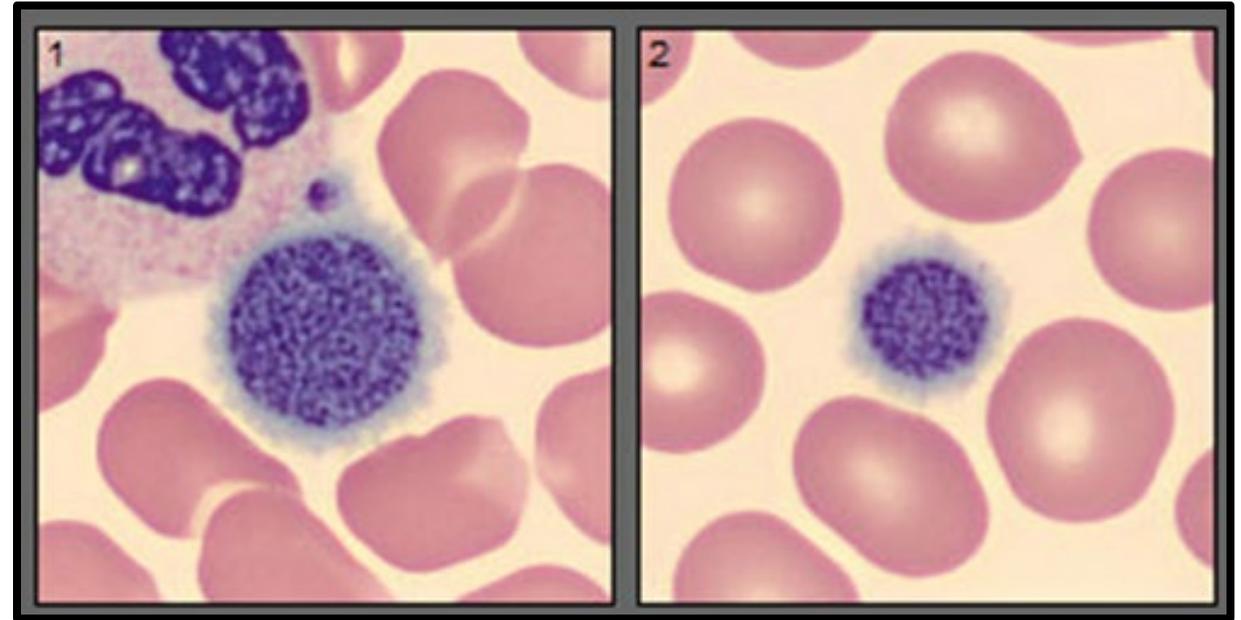
Normal

Thrombocytosis



Large and Giant platelets

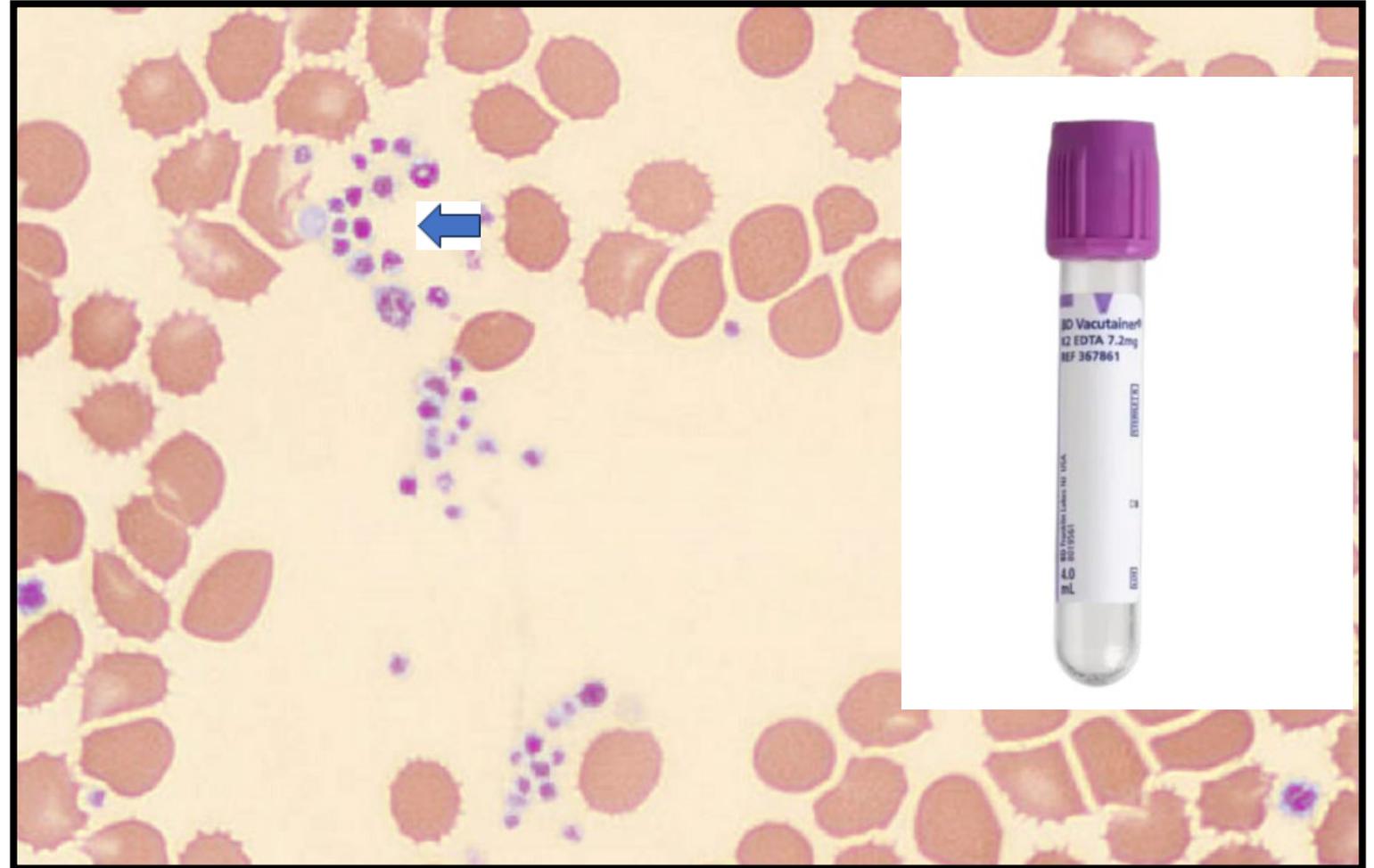
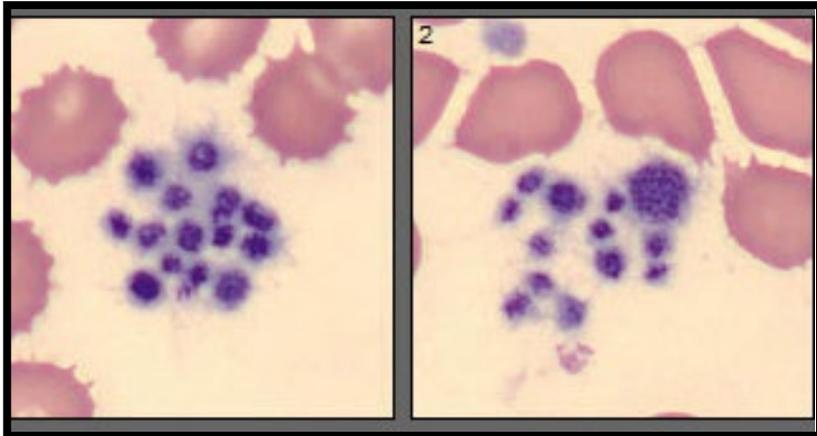
- Size variation can be seen in inherited and acquired disorders
- Large and Giant platelets seen in Bernard-Soulier syndrome, Gray platelet syndrome, May-Hegglin anomaly



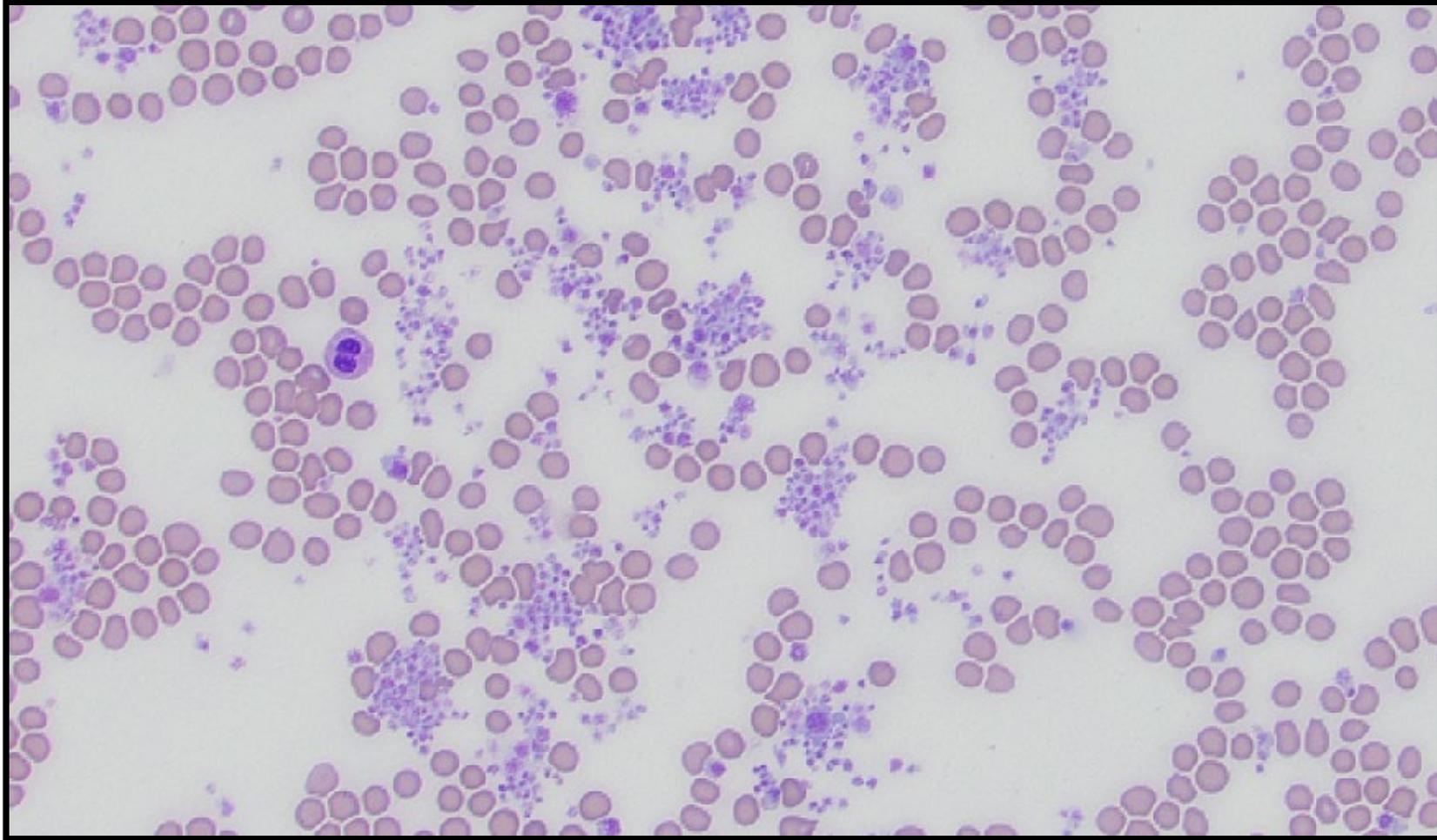
Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Platelet clumps: an in-vitro phenomenon

- Lab artifact
- Autoantibodies causing platelet clumps
- EDTA induced exposure of the epitopes on platelets

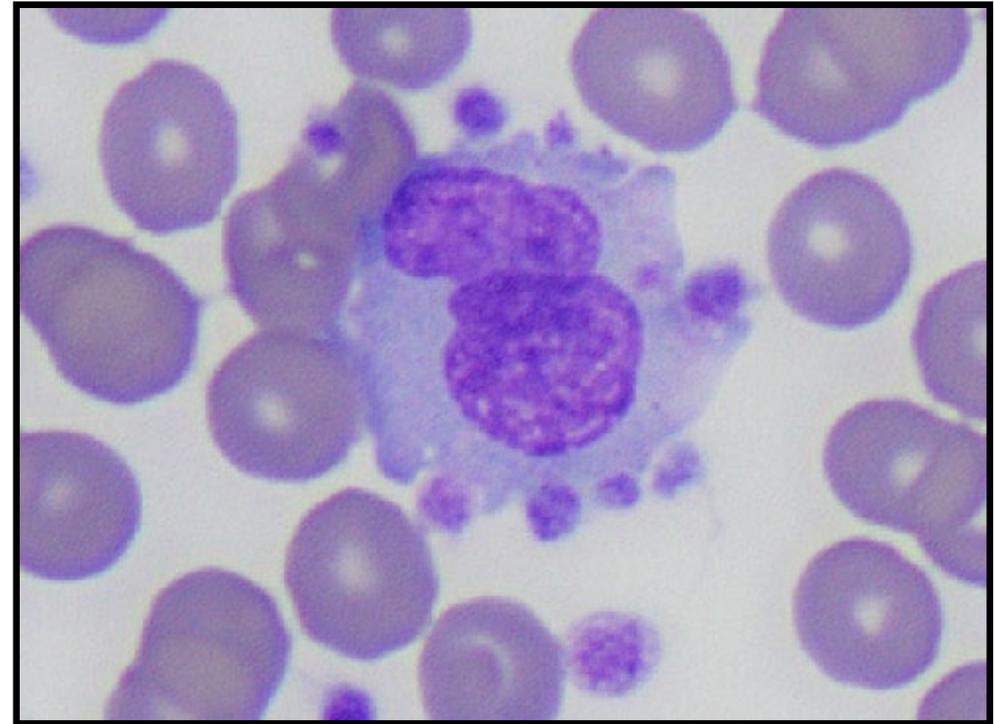


EDTA tube



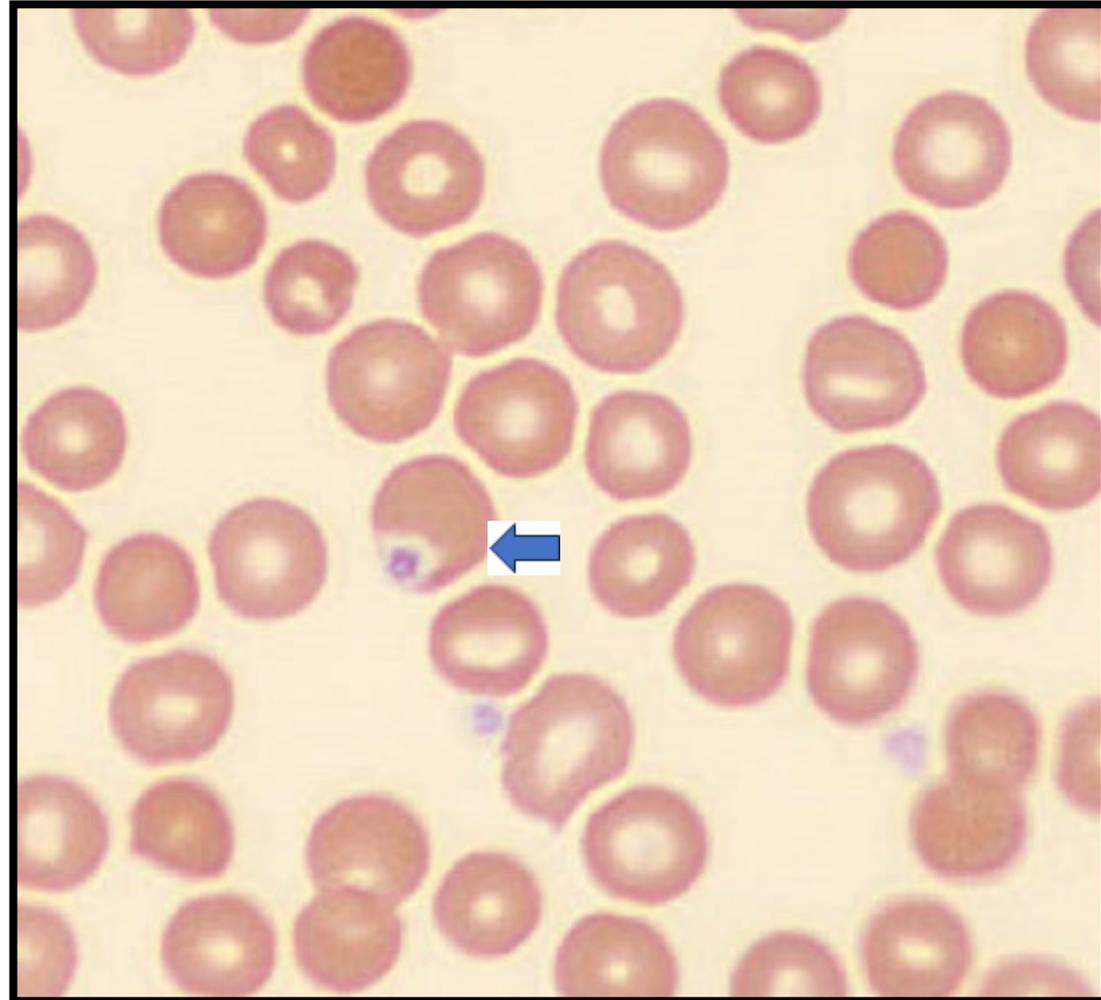
Source: Dr. Afshan Idrees, University of Miami Department of
Pathology & Laboratory Medicine

Platelet satellitism



Platelet overlying an RBC

- Look alike of an RBC inclusion
- Compare with the surrounding platelets



Source: Dr. Afshan Idrees, University of Miami Department of Pathology & Laboratory Medicine

Thank you