University of Baghdad
College of Medicine
2023-2024

Title: WBC Disorders – Part 1A

Grade: 4

Module: PATHOLOGY (Hematology)

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WBC Disorders (Part 1A)

Non-neoplastic WBC Disorders

Acute Leukemia

https://www.youtube.com/watch?v=aVd7OnG4c3c

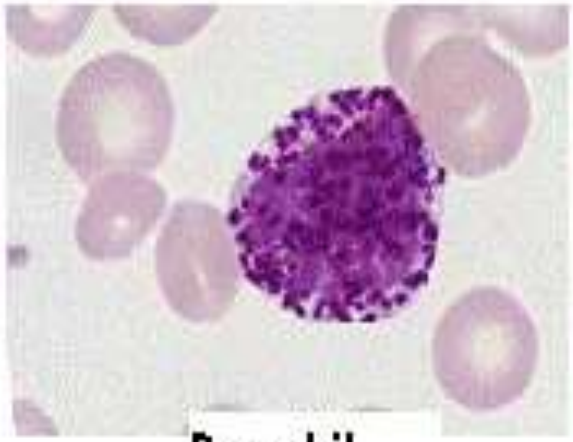
Objectives:

By the end of this session the student should be able to:

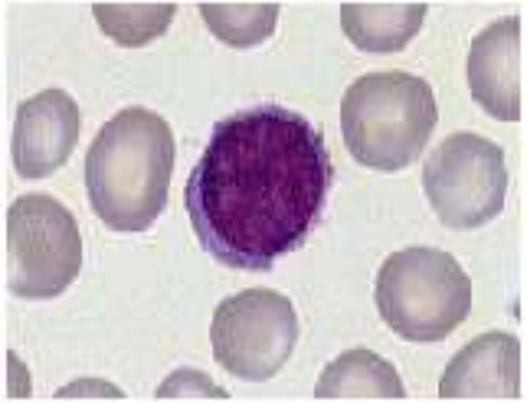
1. List the main causes of reactive leukocytosis.
2. Diagnose and classify acute leukemia (AL).
3. Differentiate the blasts' morphology between ALL and AML.

Peripheral blood smear, showing normal blood leukocytes morphology

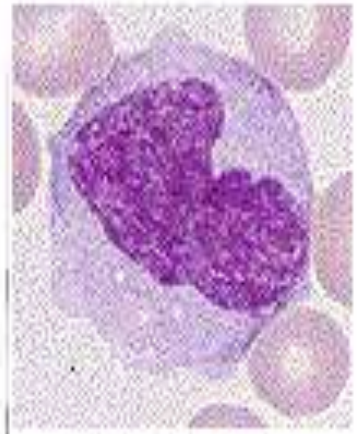
Eosinophil



Basophil



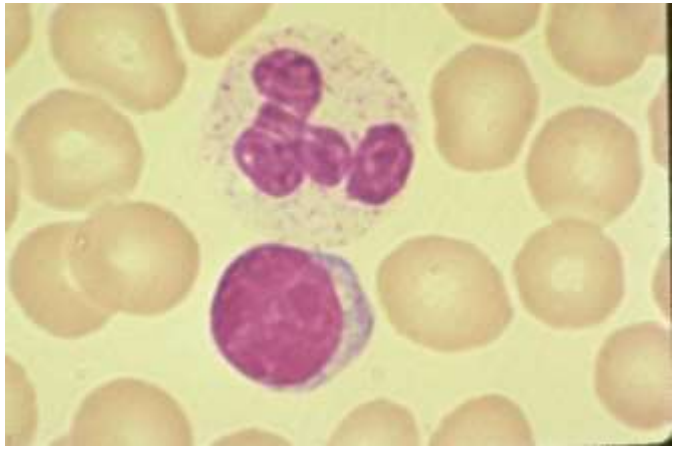
Lymphocyte



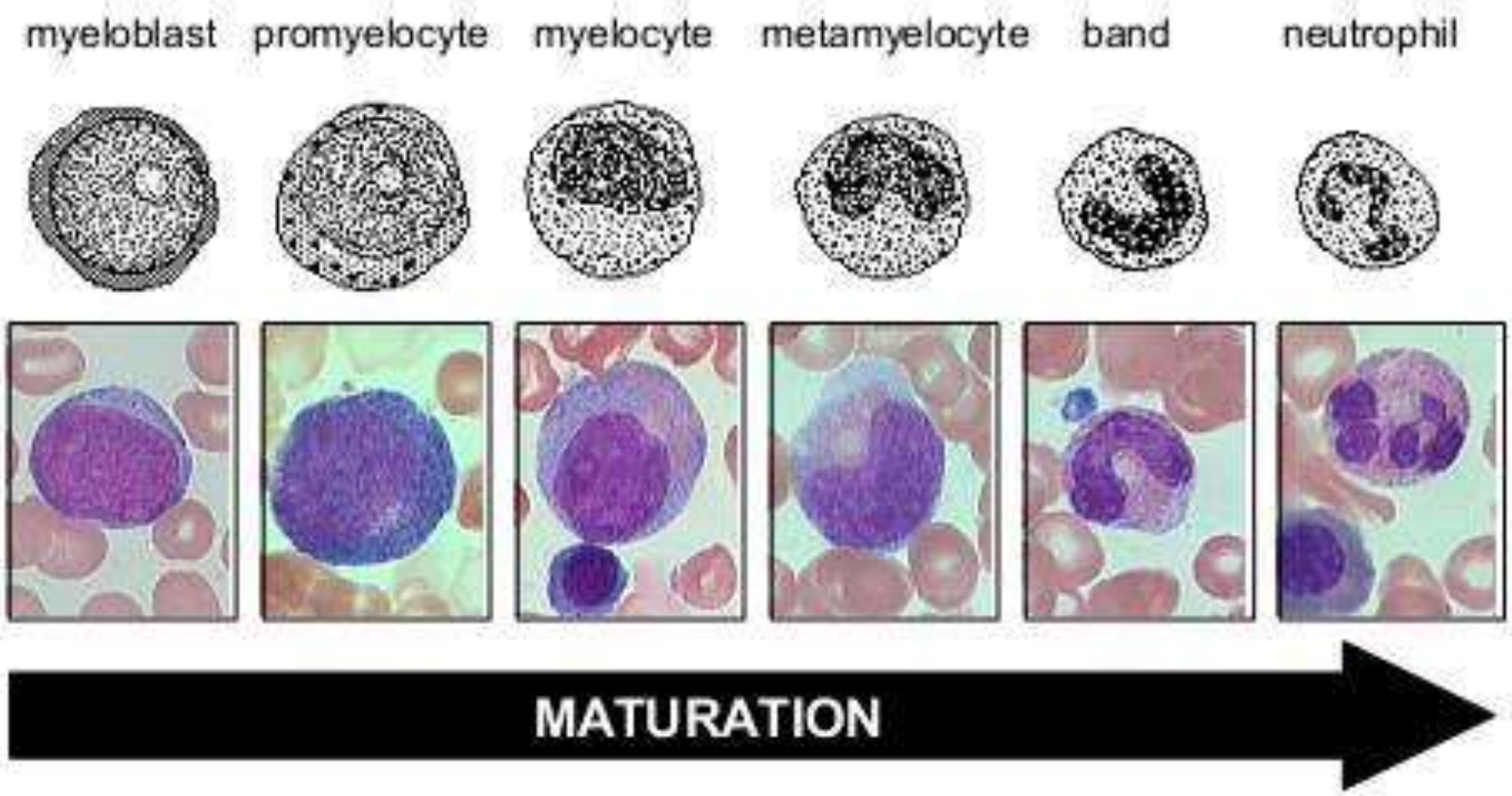
Monocyte

Neutrophil

Lymphocyte

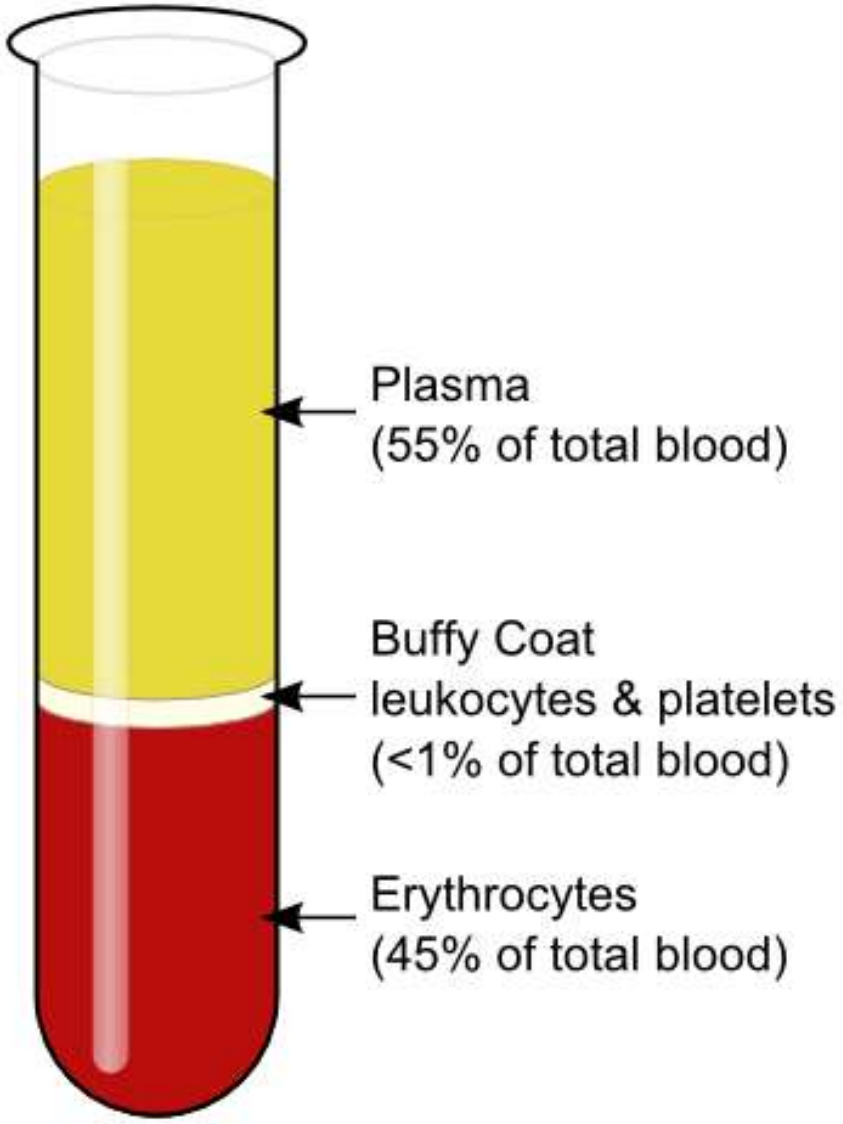


Bone marrow aspirate showing normal maturation sequence of granulocytic cells



Non-Neoplastic WBC Disorders

Whole blood after centrifugation



Terminology

Leukocytosis: is an increase in the total white blood cell (WBC) count above the upper limit of normal (> 11 × 10^9/L).

- Neutrophilia: Neutrophil count > 7.0 × 10^9/L
- Eosinophilia: Eosinophil count > 0.5 × 10^9/L
- Basophilia: (> 0.1 × 10^9/L)
- Lymphocytosis: (> 3.5 × 10^9/L)
- Monocytosis: (> 1.0 × 10^9/L)

Terminology

Leukopenia: is a decrease in the total WBC count below the lower limit of normal (< 4×10^9/L).

- Neutropenia: Neutrophil count < 2×10^9/L
- Lymphopenia: Lymphocyte count < 1.5×10^9/L

- ***Neutrophilia***
  - Acute bacterial infections
  - Myocardial infarction, burns
  - Uremia, eclampsia, acidosis, gout
  - Neoplasms of all types
  - Acute hemorrhage or hemolysis
  - Treatment with myeloid growth factors (G-CSF, GM-CSF):
    - Filgrastim (Neupogen)
    - Lenograstim (Granocyte)

- ***Eosinophilia***
  - Allergic disorders
  - Drug sensitivity
  - Parasitic infestations
  - Collagen vascular disorders
  - Certain malignancies
  - Myeloproliferative neoplasms
  - Treatment with GM-CSF

- ***Basophilia***

Neoplastic:

Often seen in chronic myeloid leukemia.

Reactive: seen in

- Ulcerative colitis,
- Myxoedema,
- Smallpox or chickenpox infections.

- ***Monocytosis***
  - Chronic bacterial infections (e.g., tuberculosis, bacterial endocarditis, syphilis)
  - Malaria
  - Collagen vascular diseases
  - Hodgkin disease, AML
  - Certain chronic myeloproliferative neoplasms
  - Inflammatory bowel diseases

- ***Lymphocytosis***
  - Infections:
    - Acute: infectious mononucleosis (EBV), rubella, mumps, infectious hepatitis, CMV, HIV, herpes, *B. pertussis*
    - Chronic: typhoid fever, syphilis, healing TB, toxoplasmosis
  - Neoplastic:
    - Chronic lymphoid leukemias
    - Acute lymphoblastic leukemia
    - Non Hodgkin Lymphoma (some)
  - Thyrotoxicosis

Neutropenia is a reduction below normal of the number of neutrophils in peripheral blood.

- ***Inadequate or ineffective granulopoiesis:***
  - Generalized marrow failure as in aplastic anemia
  - Isolated neutropenia: Congenital, Racial or familial, Cyclical neutropenia (3-4 weeks periodicity)
- ***Accelerated removal or destruction of neutrophils:***
  - Acquired: drug-induced, immune mediated
  - Infections: bacterial (typhoid, miliary tuberculosis), viral (hepatitis, influenza, HIV), fungal
  - Hypersplenism
- ***Altered distribution:*** as in stress and certain drugs

Lymphopenia is a reduction below normal of the number of lymphocytes in peripheral blood.

1. Congenital immunodeficiency diseases
2. Acquired:
   - Advanced HIV infection, miliary TB
   - Treatment with corticosteroids and other immunosuppressive therapy
   - Advanced Hodgkin disease

Which is more commonly encountered in routine work?

A. Reactive non-neoplastic WBC disorders.

B. Neoplastic WBC disorders.

Neoplastic WBC Disorders

Acute leukemia

- Definition
- Clinical features
- Diagnosis
- Classification

DEFINITION:

AL is usually an aggressive clonal malignant transformation involving the hematopoietic stem cells or early progenitors (blasts) and characterized by uncontrolled proliferation of blast cells in the BM with spillage into the peripheral blood and variable infiltration of other organs.

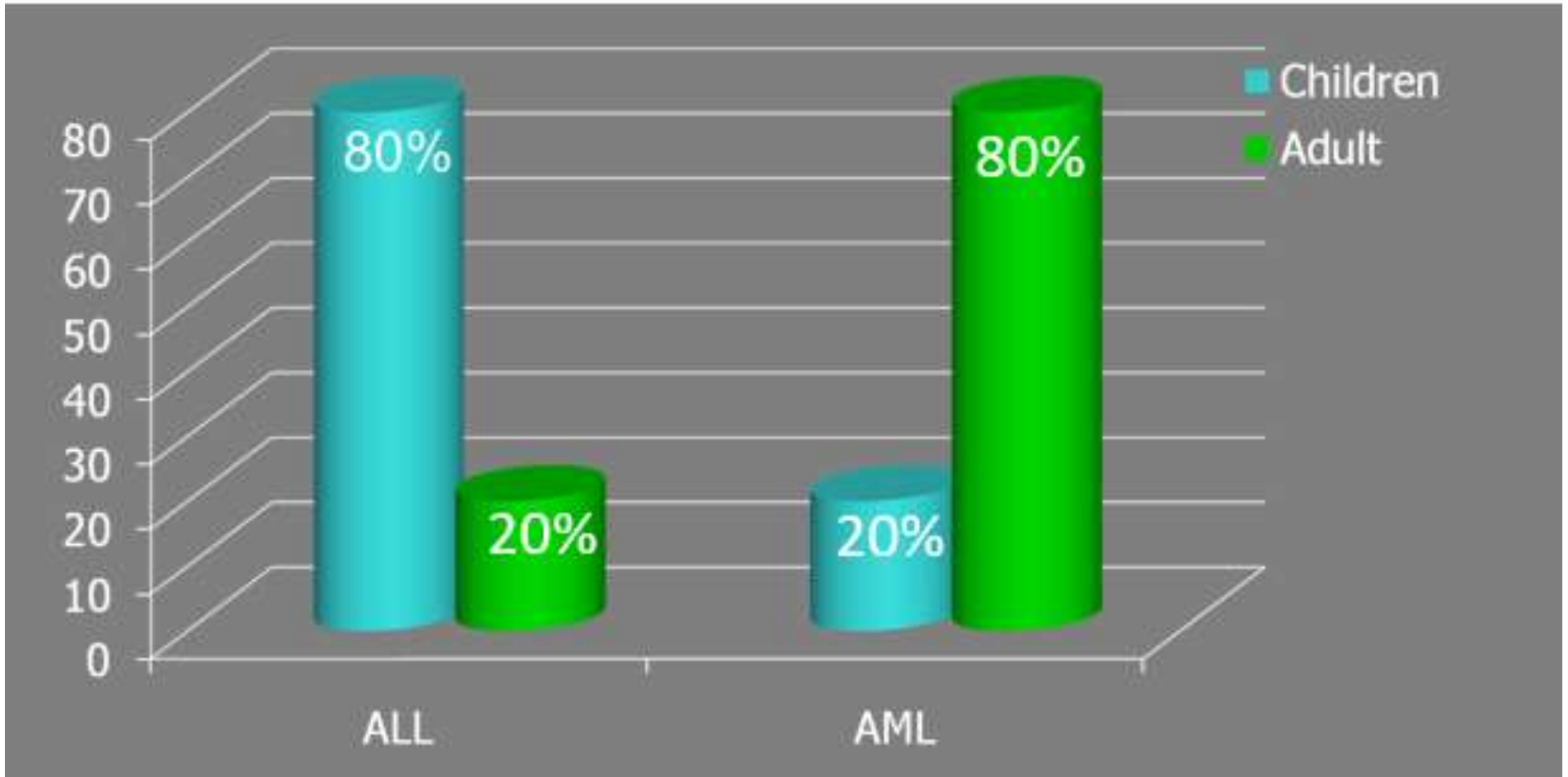
There are two major types of AL;

1. **Acute lymphoblastic leukemia (ALL)**
2. **Acute myeloblastic leukemia (AML).**

CLINICAL FEATURES:

AL occurs at any age, and could be classified accordingly into:

1. **Childhood AL which is usually lymphoblastic (ALL)**
2. **Adult AL which is usually myeloblastic (AML).**



The common symptoms & signs at presentation are mainly attributed to:

1. **Bone marrow failure.**
2. **Organ and Tissue Infiltration by the leukemic cell**

1. BONE MARROW FAILURE:

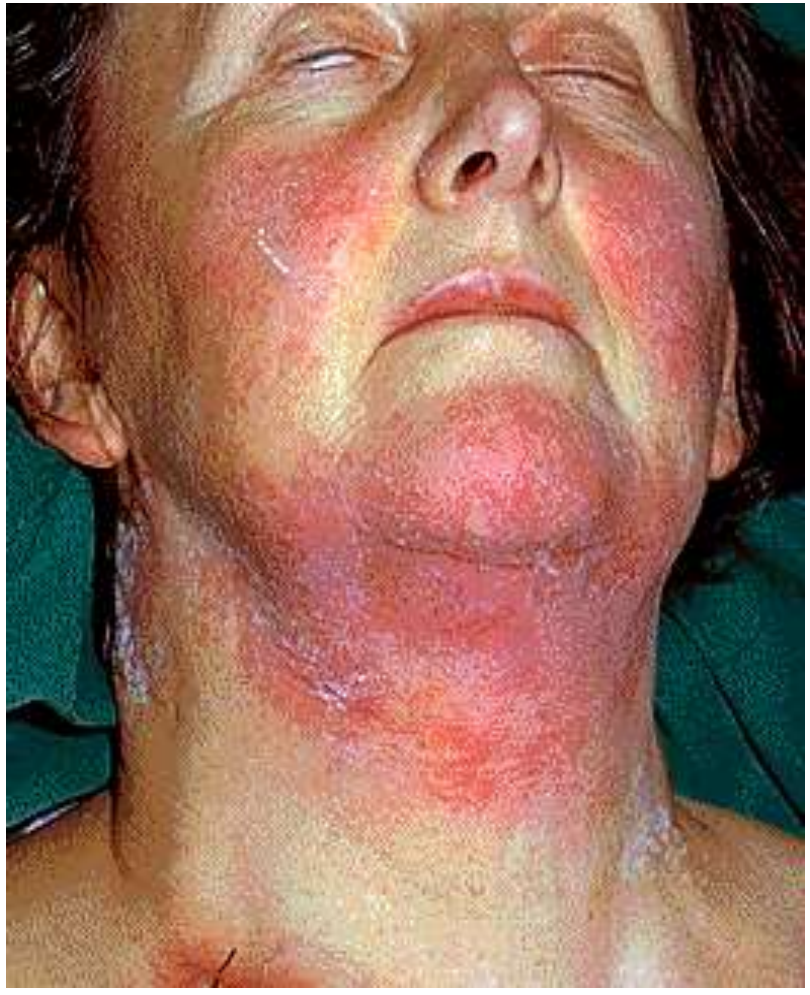
A. ANEMIA:

Pallor, weakness, fatigue, lethargy, dyspnea on exertion, angina and palpitation.

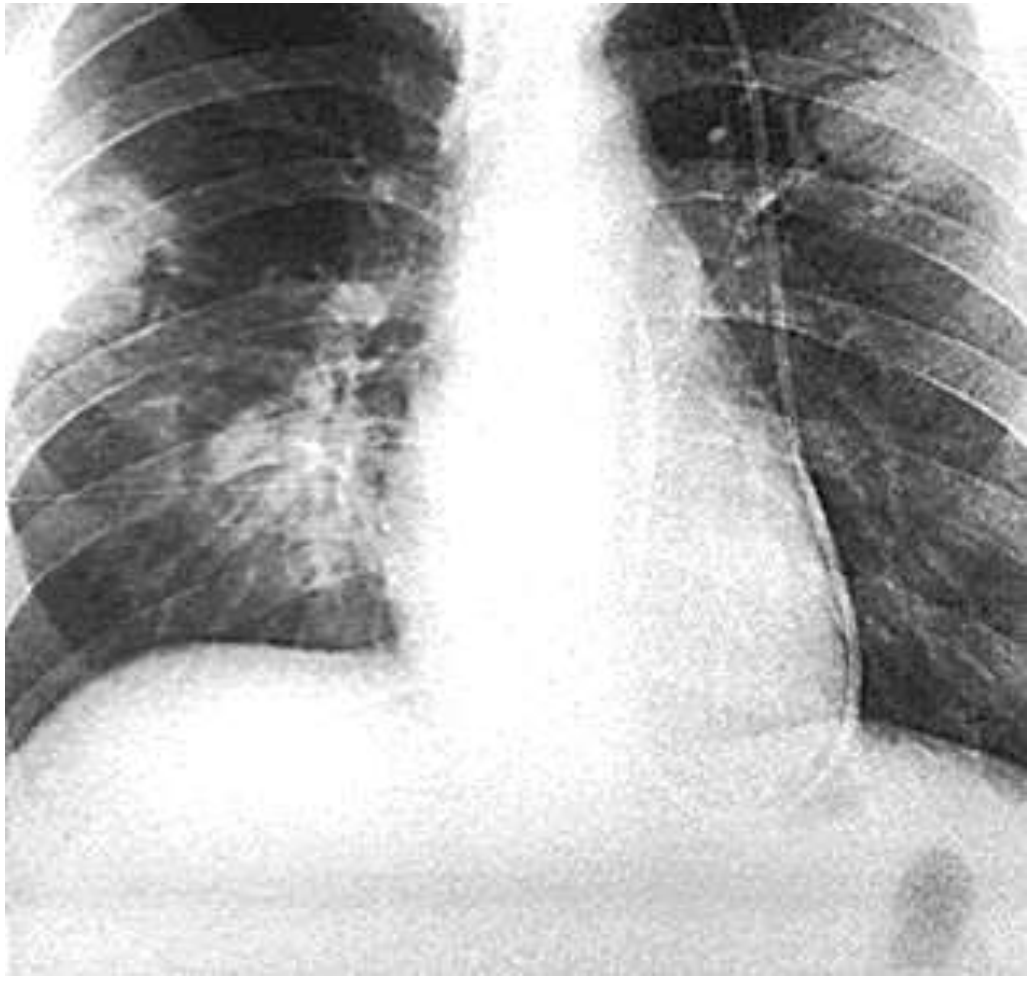
B. NEUTROPENIA:

Fever and infections due to reduced immunity, especially:

Skin infection



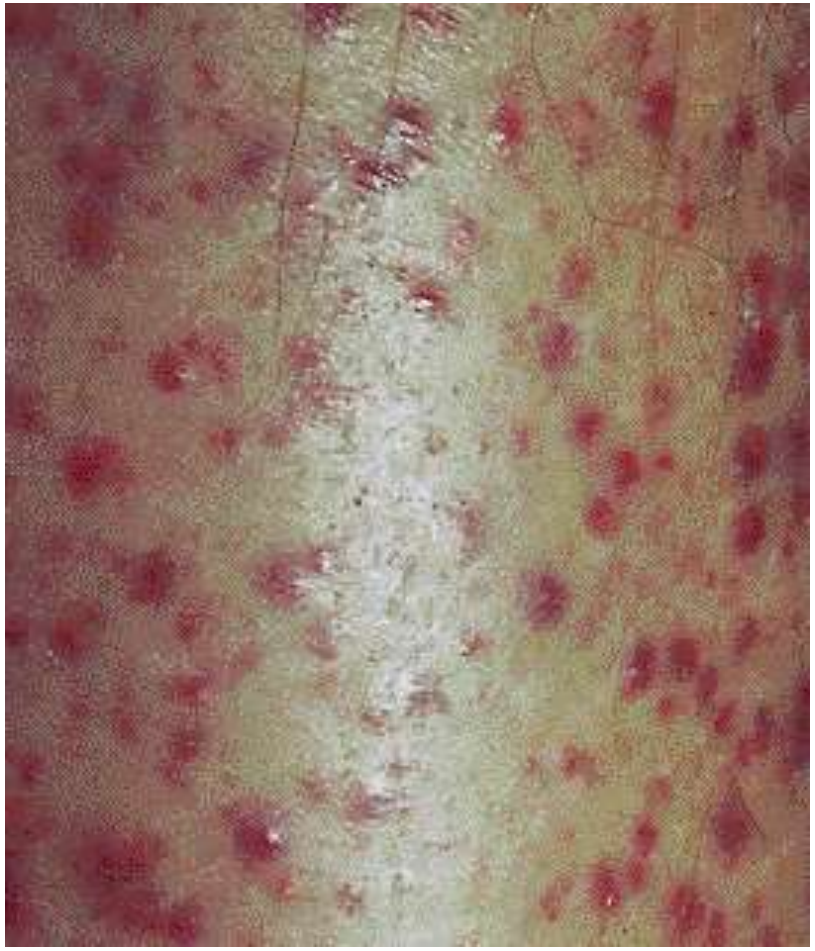
Respiratory infection



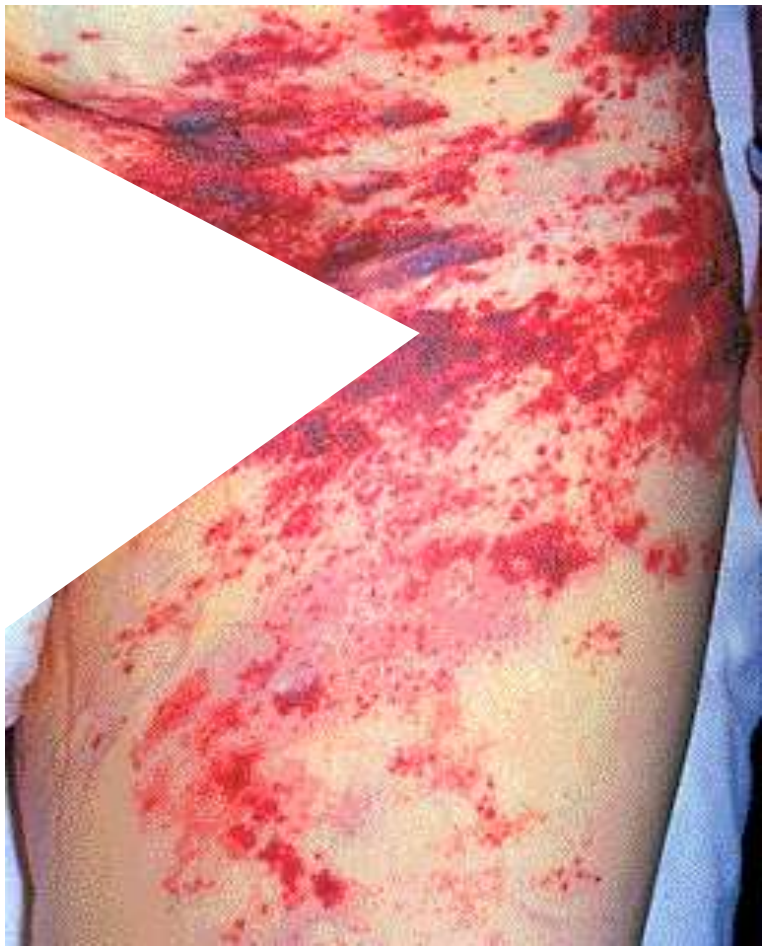
C. THROMBOCYTOPENIA:

Bleeding manifestations into the skin:

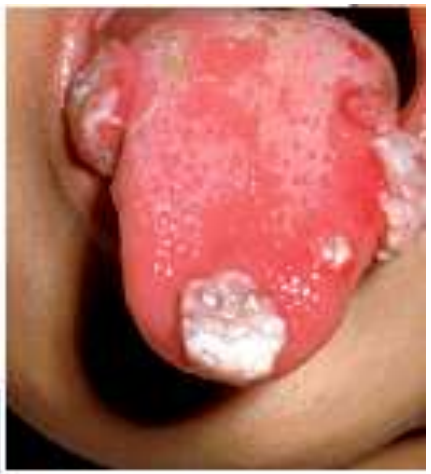
(Petechia)

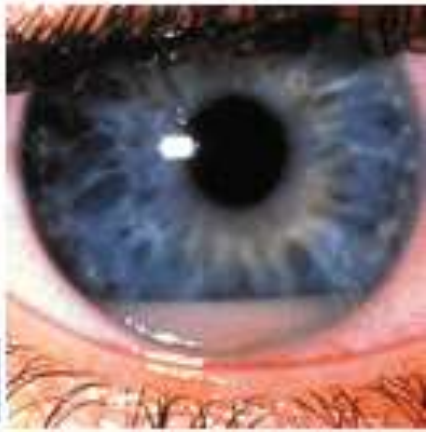


(Ecchymosis)



2. Organ and Tissue Infiltration by the leukemic cells: Splenomegaly..Hepatomegaly..Bone pain..Arthralgia..





A. Cervical lymphadenopathy; B. Facial palsy; C. Tongue infiltration;
D. Gum infiltration; E. Ocular infiltration; F. Ant. chamber infiltration

DIAGNOSIS OF AL:

Presence of blasts
in the BM and/or PB

≥ 20 %

< 20 %

Presence of specific recurrent (leukemia-associated) cytogenetic or molecular genetic abnormalities

Blood Film findings:

a. **RBCs:** anemia is almost always present.

b. **WBCs** total count may be:

- *Normal*
- *High count*
- *Low count*

Neutropenia is common

c. **Platelets:** decreased in most cases.

***Bone marrow aspirate findings*:**

BMA is necessary to confirm the diagnosis (especially when low counts).

Why?

- **Because the blast percentage in PB may not exceed the cutoff point of ≥20% required for the diagnosis of AL.**
- **The blast may exceed this percentage in the BM and,**
- **Doing genetic study on BM aspirate sample is essential to verify or exclude AL.**

CLASSIFICATION:

It is based on:

1. **Morphology of blasts.**
2. **Cytochemistry.**
3. **Immunophenotyping.**
4. **Genetic analysis.**

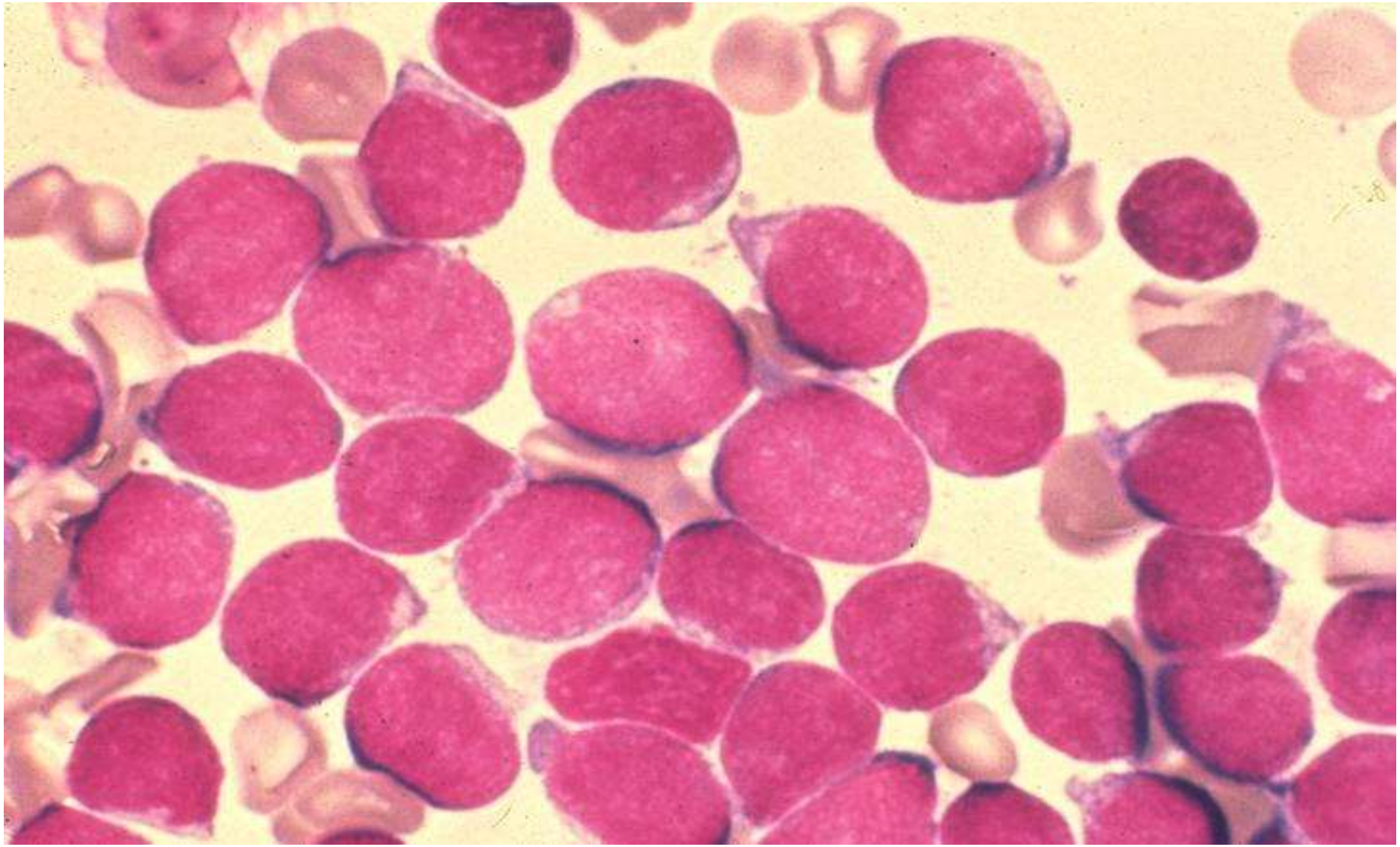
What are the two major types of AL?

- Acute Lymphoid Leukemia (ALL)
- Acute Myeloid Leukemia (AML)

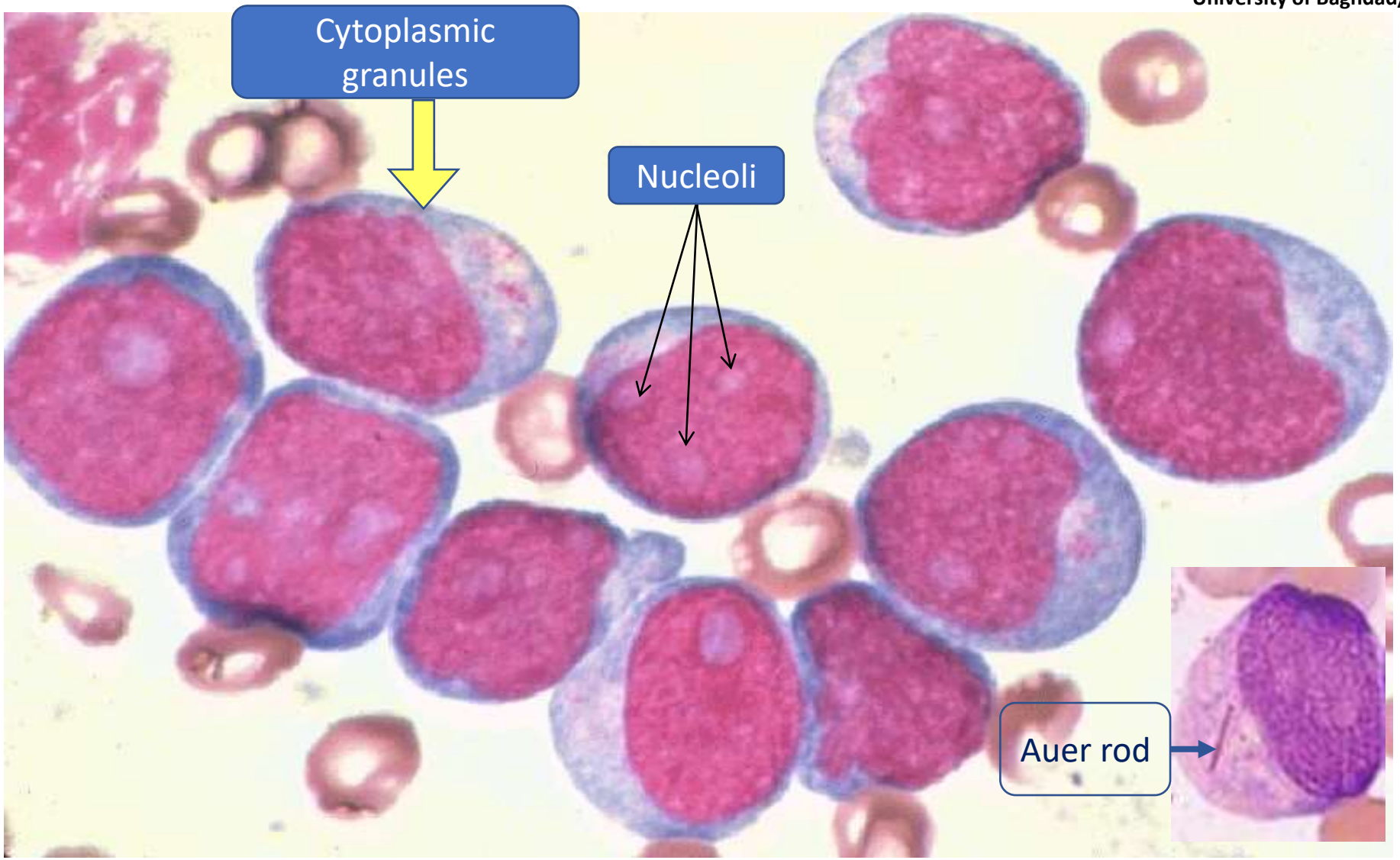
MORPHOLOGY IN AL

How can you morphologically differentiate between lymphoblasts (ALL) and myeloblasts (AML)?

The nuclei of **lymphoblasts** have somewhat coarse and rather clumped chromatin and one or two nucleoli; **myeloblasts** tend to have finer chromatin with multiple nucleoli and more cytoplasm, which may contain granules or Auer rods.



Acute lymphoblastic leukemia. The nuclei of lymphoblasts have somewhat coarse and rather clumped chromatin and one or two nucleoli.



Acute myeloblastic leukemia. Myeloblasts tend to have finer chromatin with multiple nucleoli and more cytoplasm, which contain granules or Auer rod.

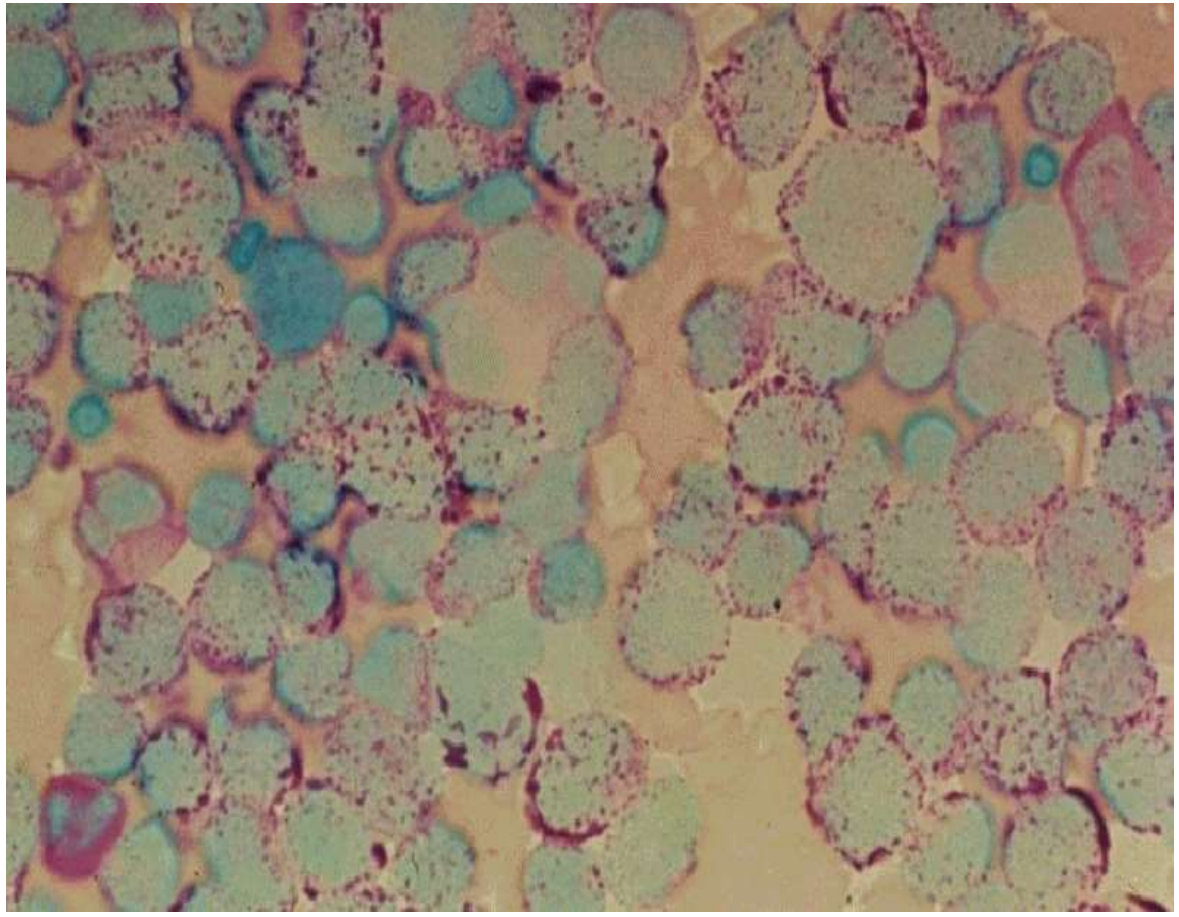
CYTOCHEMISTRY IN AL

Special stains	ALL	AML
Myeloperoxidase (MPO)	Neg.	Pos.(including Auer rods)
Sudan Black B (SBB)	Neg.	Pos.(including Auer rods)
Non-specific esterase	Neg.	Pos. in M4, M5
Periodic Acid-Schiff (PAS)	Positive in many cases	Pos. in M6 (fine blocks)
Acid Phosphatase	Positive in T-ALL	Pos. in M6 (diffuse)

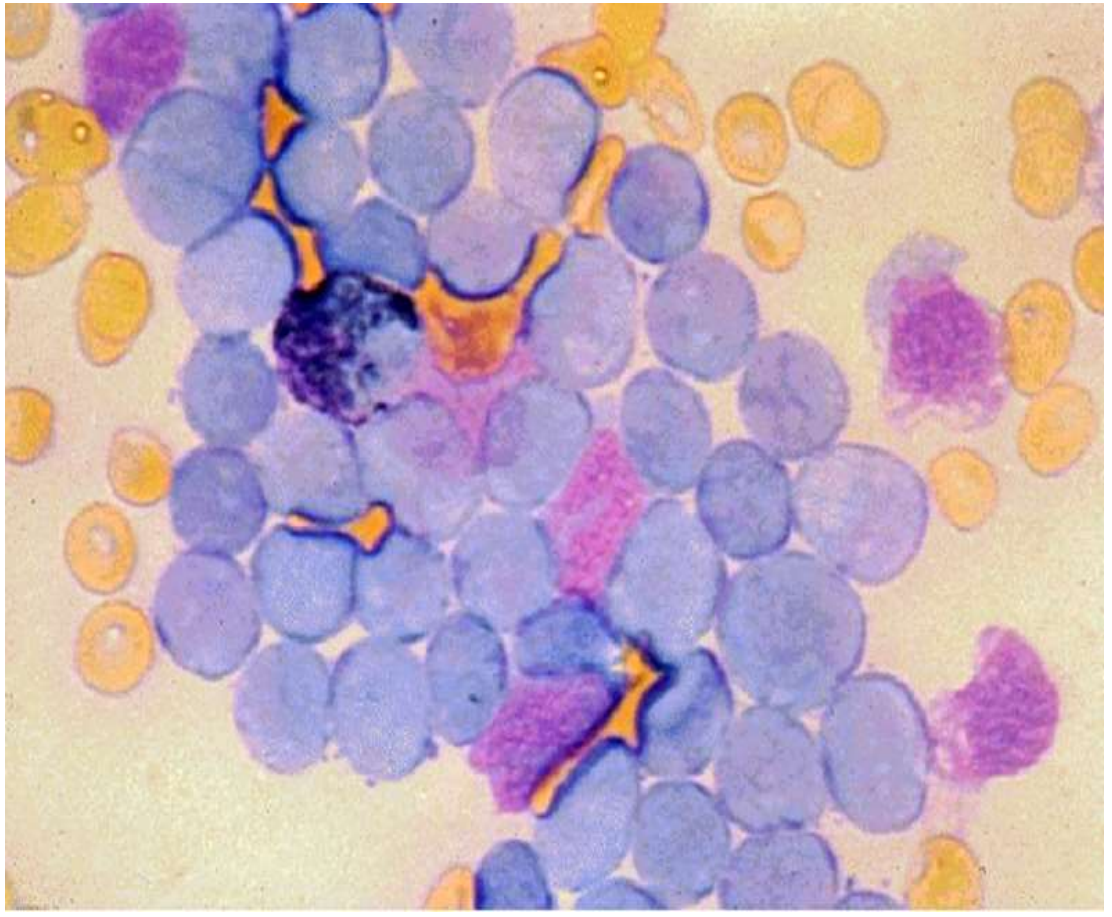
Cytochemistry in ALL

PAS; positive

(coarse granular or blocks in cytoplasm)

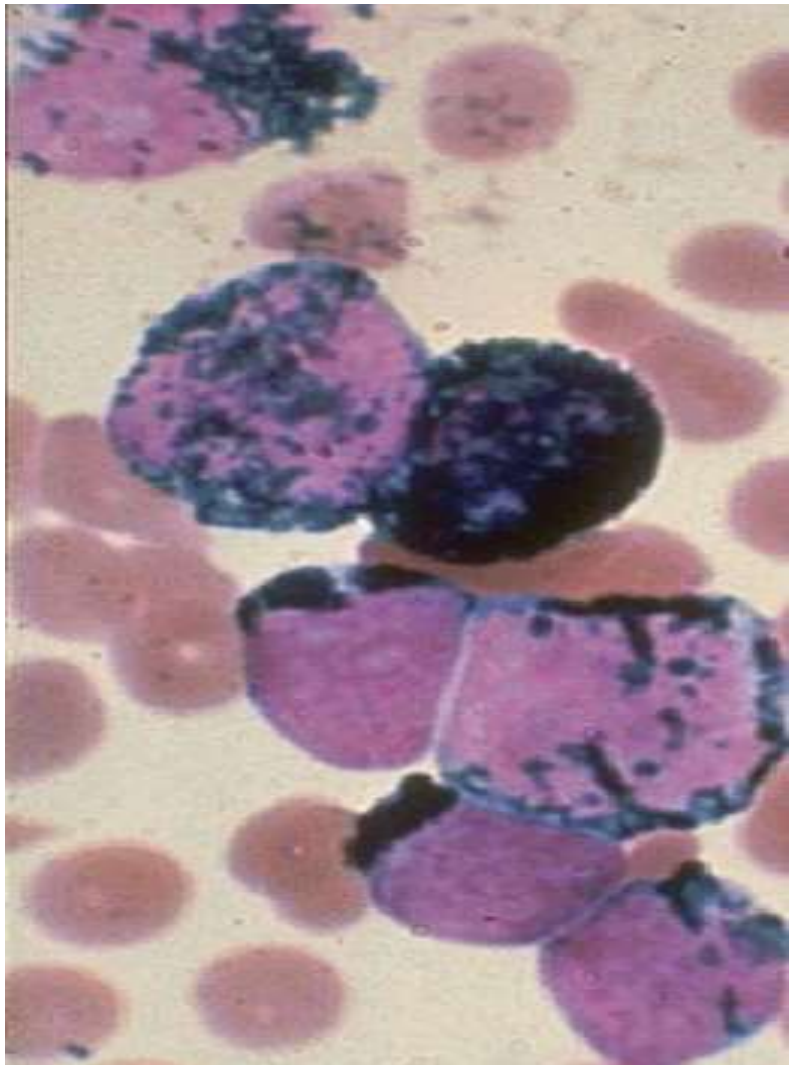


SBB; negative

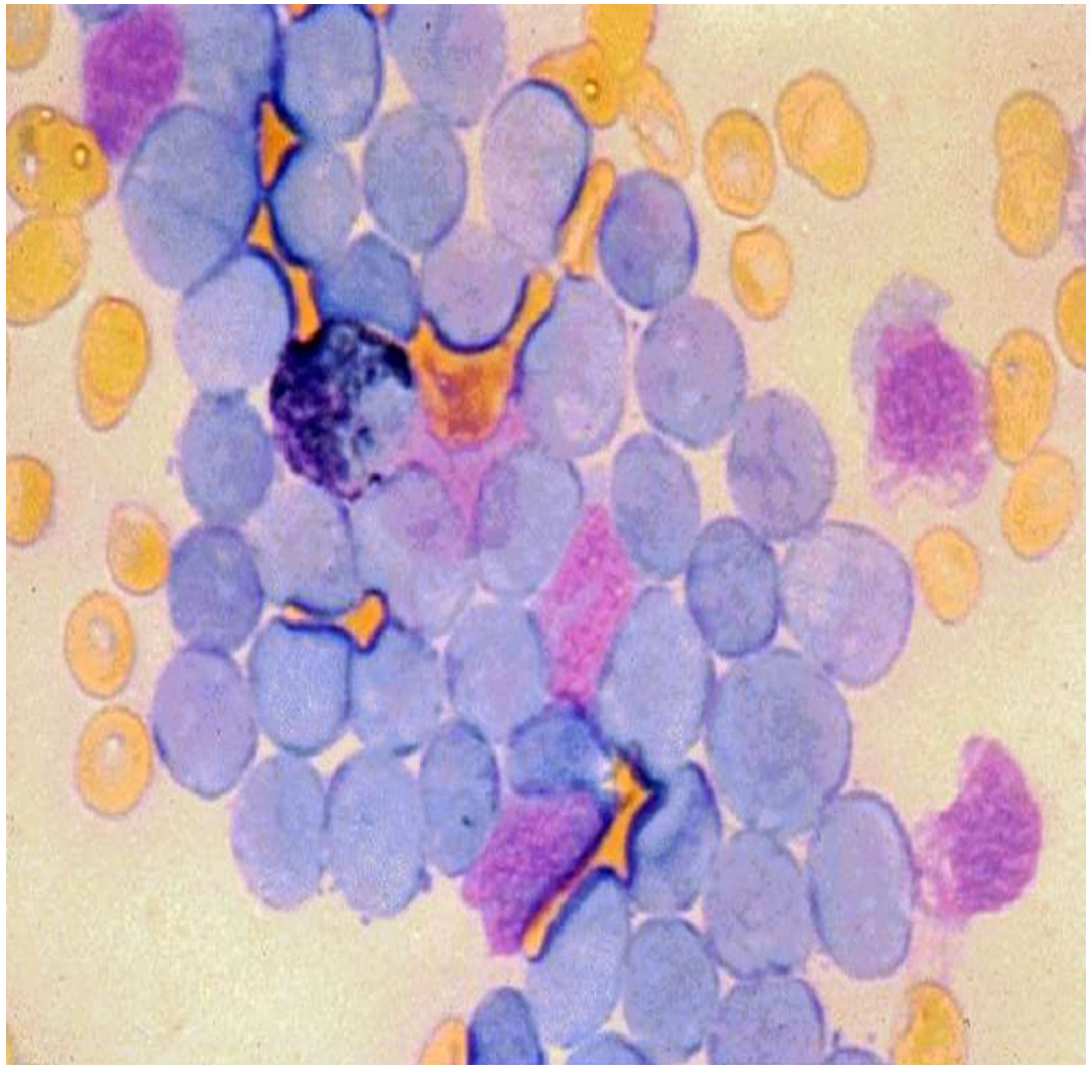


Cytochemistry: Sudan Black B (SBB)

Positive in AML



Negative in ALL



Which of the following stains is(are) positive for Lymphoblasts?

A. Sudan Black B.
B. Periodic Acid Schiff.
C. Myeloperoxidase.
D. Acid phosphatase.
E. Non-specific Esterase.

IMMUNOPHENOTYPING IN AL

Immunophenotyping (IPT) can be determined by **flowcytometry** or **immunohistochemistry.** It is very useful in typing and subtyping of AL.

- CD79a is the most **specific** marker for **B-cells**
- CD3 is the most **specific** marker for **T-cells**.
- Anti-myeloperoxidase (MPO) is the most **specific myeloid** marker.

Which of the following markers is the most specific for Myeloblasts?

A. CD3.

B. CD79a.

C. MPO.

GENETIC ANALYSIS IN AL

- ***ALL*: the most common karyotypic abnormalities in B-cell ALL *(with good prognosis)* is *hyperploidy* (>50 chromosomes/cell), which is associated with t(12:21).**

  Poor outcomes are observed with **11q23** and the Philadelphia (***Ph*$^{+}$**) chromosome.

- ***AML*: *good outcome*** correlates with **t(8:21)** & **t(15:17)**. Conversely, ***poor outcome*** correlates with ***Ph*$^{+}$**, and **t(6:9)**.

Case Scenario

A 40-year-old man presented with

- Weakness and Sore throat for one week duration.

Physical examination:

- Pale
- Petechial Rash on his legs.
- His Throat was red and inflamed
- Cervical and axillary lymph nodes enlargement.
- Splenomegaly

What Laboratory Investigations should be performed?

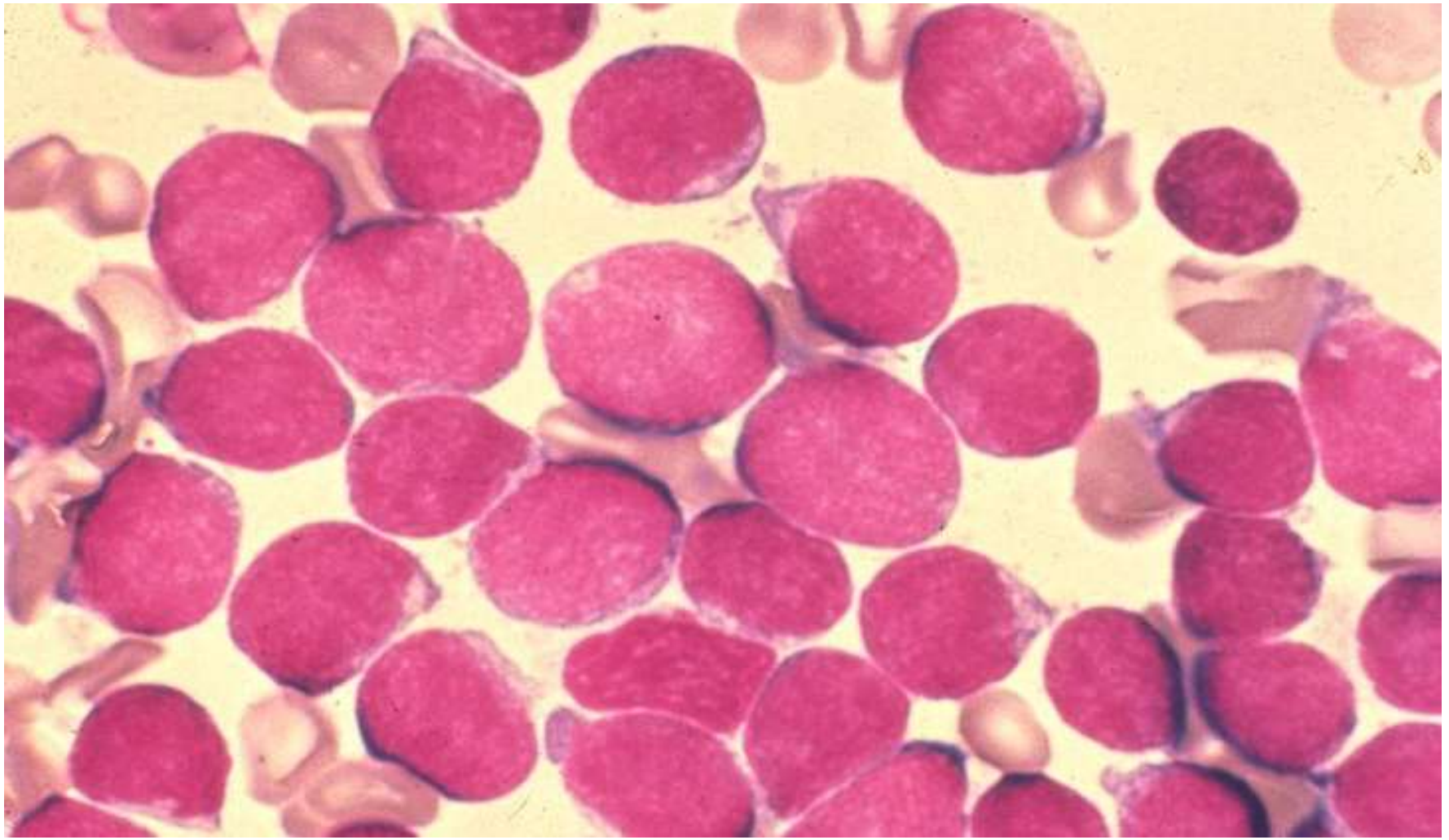
- Complete Blood Count (CBC)
- Blood Film

Investigations:

►CBC:

	Patient's Results		Normal Range
Hb	9.5 g/dL		13 – 17 g/dL
WBC	25 $\times 10^9$/L		4 – 11 $\times 10^9$/L
Neutrophils	1 $\times 10^9$/L	4%	2 – 7 $\times 10^9$/L
Lymphocytes	2 $\times 10^9$/L	8%	1.5 – 3.5 $\times 10^9$/L
Blasts	22 $\times 10^9$/L	88%	Abnormal
Platelets	20 $\times 10^9$/L		150 – 410 $\times 10^9$/L

►Blood Film and/or BM aspirate



Blood film; Leukemia Blasts showing no evidence of maturation.

What is your differential diagnosis?

A 4-year-old child presented with fever, pallor, and petechial bleeding for one week duration. Physical examination revealed cervical and axillary lymphadenopathy with splenomegaly.

The most likely diagnosis is:

A. Acute lymphoblastic leukemia.

B. Acute myeloblastic leukemia.

C. Chronic myeloid leukemia.

D. Chronic lymphocytic leukemia.

Summary

- Reactive leukocytosis is much more commonly encountered in routine work than neoplastic states.
- Absolute leukocytes counts are the target for interpretation not their percentages in the PB.
- Morphology, cytochemistry, IPT and genetic analysis. All these investigations may be required to reach the diagnosis and classify AL.
- The diagnosis of AL is based on a cutoff point of blast cells (20%) whether in the PB or BM.

End of WBC Part 1A