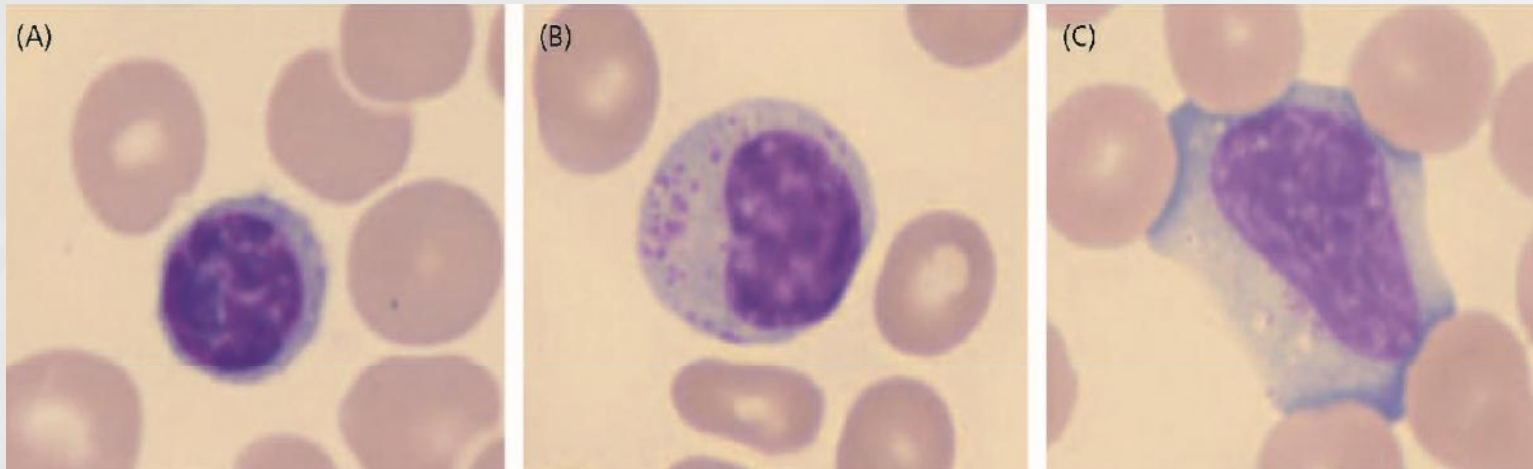
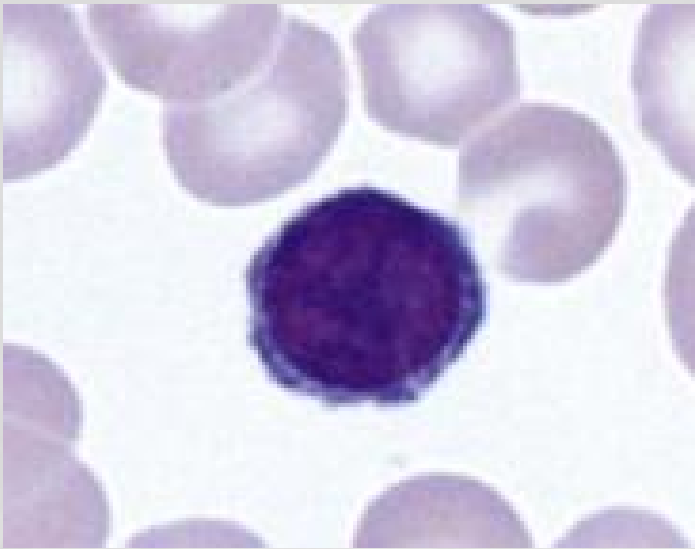


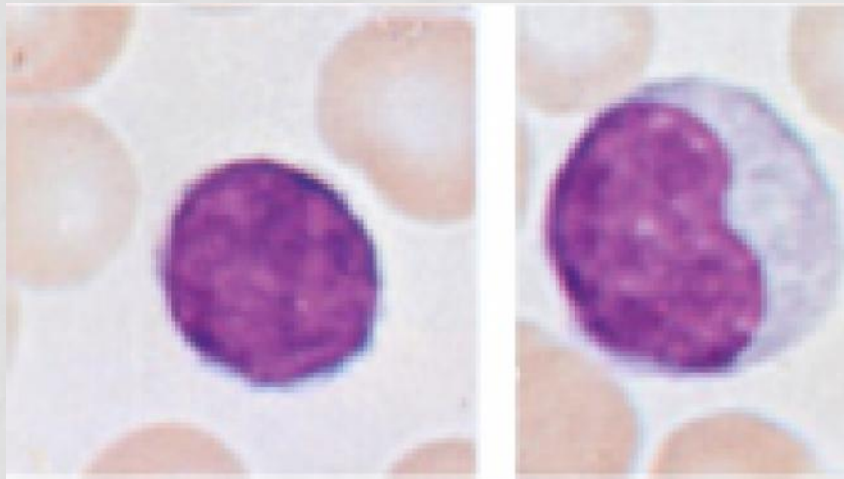
# Atlas of Atypical Lymphocytes



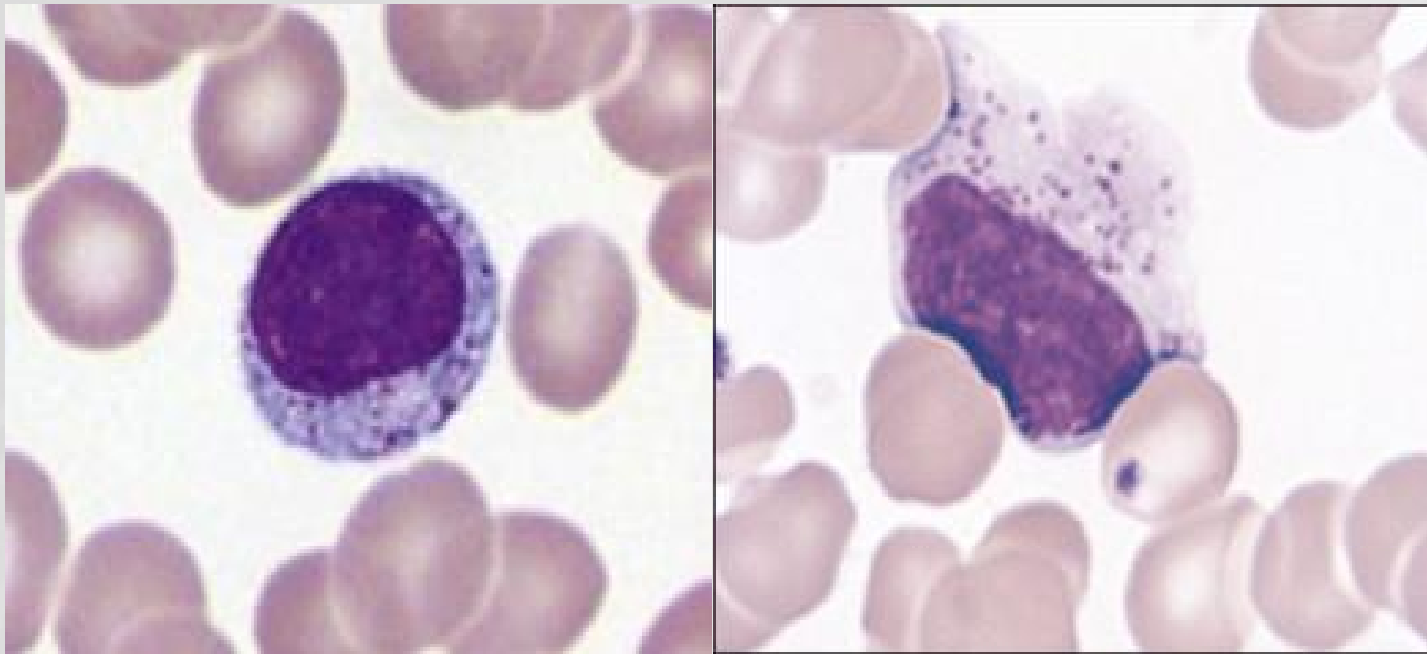
인제대학교 해운대백병원  
진단검사의학과 이정녀



Small lymphocyte with high N/C ratio, condensed chromatin, and scanty basophilic cytoplasm (W-G, x1000)



Small and large lymphocytes from the blood of normal subjects



Large granular lymphocyte with azurophilic cytoplasmic granules  
(W-G, x1000)

# Contents

## Nonmalignant Lymphocyte Disorders

Lymphocytosis

Lymphopenia

## Malignant Lymphocyte Disorders

# Nonmalignant Lymphocyte Disorders

## **Acquired, quantitative disorders**

is usually a self-limited reactive process to infection or inflammation

Both B and T cells are affected

Functionally normal, heterogeneous morphological process  
may have lymphadenopathy or splenomegaly

## **Congenital qualitative or quantitative disorders**

may affect both T and B cells or only one cell type.

Most will severely compromise the immune system

- Severe combined immunodeficiency system (SCIDS)
- Wiskott-Aldrich Syndrome, DiGeorge syndrome
- X linked agammaglobulinemia, ataxia telangiectasia

## **Acquired immune deficiency syndrome (AIDS)**

# Lymphocytosis

reference intervals :  $\approx 1.5\text{--}4.0 \times 10^9/\text{L}$  in the adult

$\approx 1.5\text{--}8.8 \times 10^9/\text{L}$  in the child

may be relative (secondary to neutropenia)

or absolute (usually seen in viral infections);

## Key Causes of Lymphocytosis

- Infectious - many viral, pertussis, tuberculosis, toxoplasmosis, rickettsial
- Chronic inflammation - ulcerative colitis, Crohn's disease
- Immune mediated - drug sensitivity, vasculitis, graft rejection, Graves' disease, Sjögren's syndrome
- Hematologic - ALL, CLL, lymphoma
- Stress - acute, transient

## Acute Infectious Lymphocytosis

Unknown etiology

: 주로 coxsackievirus A, B6, echoviruses, adenovirus type 12 등과 연관

Leukocytosis with absolute lymphocytosis

60-97% normal appearing lymphocytes (mainly T lymphs)

Atypical lymphocytes are uncommon.

## *Bordetella pertussis* infection

significant leukocytosis with an absolute lymphocytosis

Due to a redistribution of T lymphocytes from the tissues to the circulation

Lymphocytes are small, normal appearing lymphocytes

## Chronic Lymphocytosis/Persistent Polyclonal B Cell Lymphocytosis

Persistent reactive lymphocytosis : uncommon event in adults

- CLL 과 감별

persistent polyclonal lymphocytosis : rare condition

- predominantly in female smokers, and in the postsplenectomy state

## Retrovirus-Associated Diseases and Conditions

Human T lymphotropic virus type 1 (HTLV-1),

- retrovirus associated with adult T cell leukemia/lymphoma (ATL)

- a transient T cell lymphocytosis

- 대부분 HTLV-1 항체를 가지므로 무증상

- 일부에서 leukemia로 진행

## Infectious mononucleosis (IM)

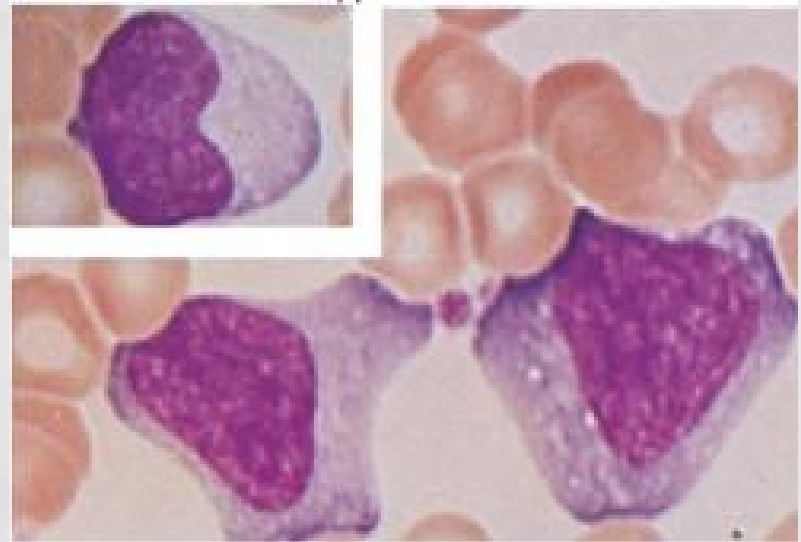
by Epstein-Barr virus infecting B lymphocytes.

- be killed by cytotoxic T cells,
- atypical (reactive) lymphocytes in the PBS - cytotoxic T cells
- nuclear alterations, abundant & basophilic cytoplasm
- monocytoïd lymphocytes
- plasmacytoïd lymphocytes

Cytologic alterations are not pathognomonic of IM

## Cytomegalovirus infection

Leukocytosis with absolute lymphocytosis  
similar to Infectious mononucleosis





## Reactive lymphocytes

- transformed, benign lymphocytes  
large as a result of antigen stimulation  
more than 30  $\mu\text{m}$  in diameter with varying size and shape
- round, elliptic, indented, cleft or folded nucleus,  
abundant, basophilic cytoplasm with/without vacuoles and/or  
azurophilic granules  
Most often the cytoplasm is gray, pale blue or deep blue in color.
- A few reactive lymphocytes is not abnormal.
- The word "atypical" is used to describe malignant-appearing cells.

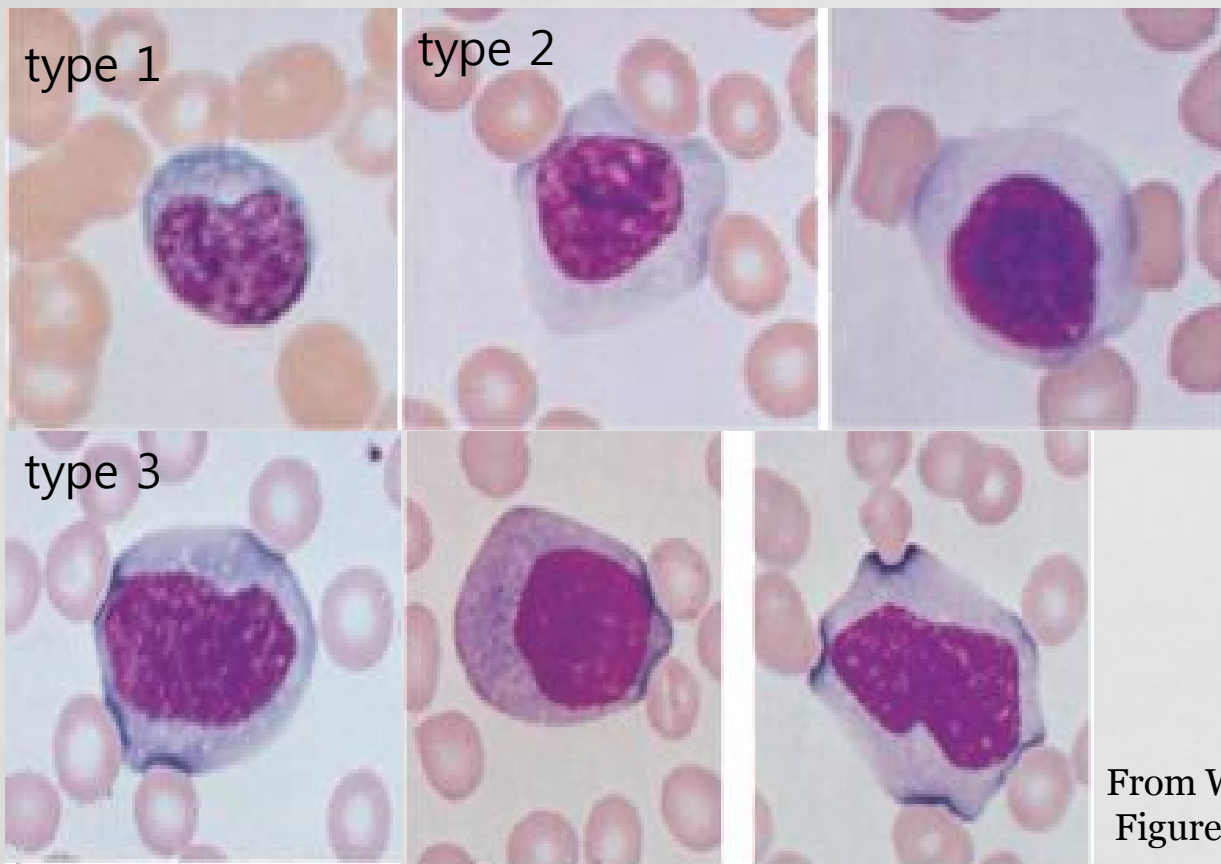
The distinctive cell associated with EBV or CMV is known as a "**Downey cell**", after **Hal Downey**, who contributed to the characterization of it in 1923

## Reactive lymphocytes -Downey cells

Downey type 1, foamy, basophilic cytoplasm with kidney-shaped nucleus

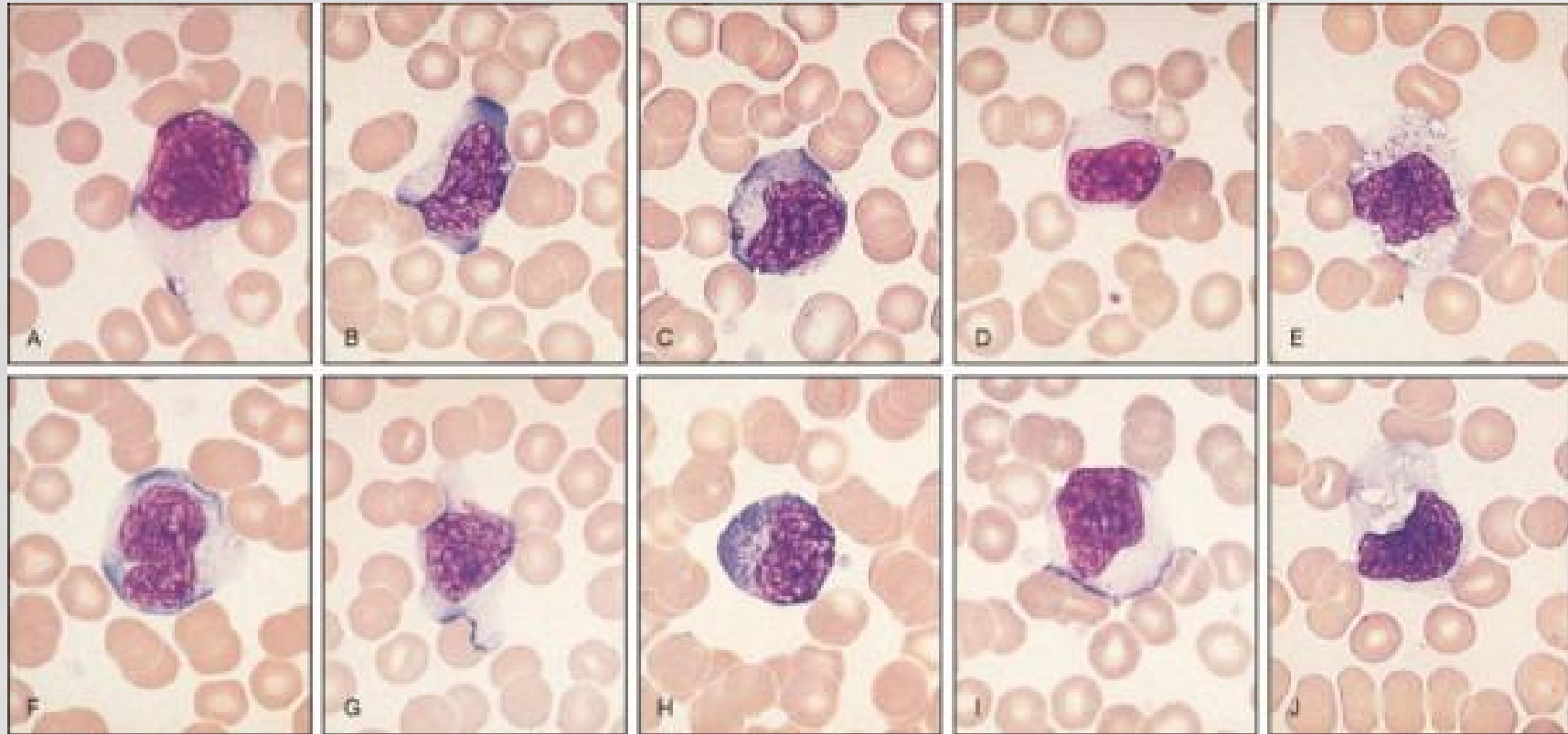
Downey type 2, less mature than type 1 cells with relatively less basophilic cytoplasm and plasmacytoid nucleus

Downey type 3, fine chromatin-type cytoplasm and 1–2 nucleoli,  
Immunoblasts or immunoblastic-like reactive lymphocytes



From Wintrobe's Clinical Hematology  
Figure 66-11.

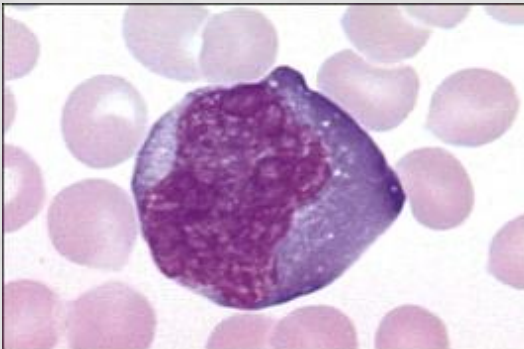
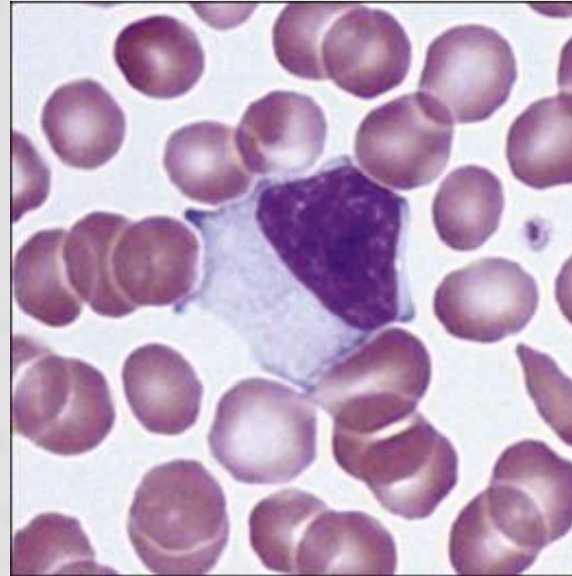
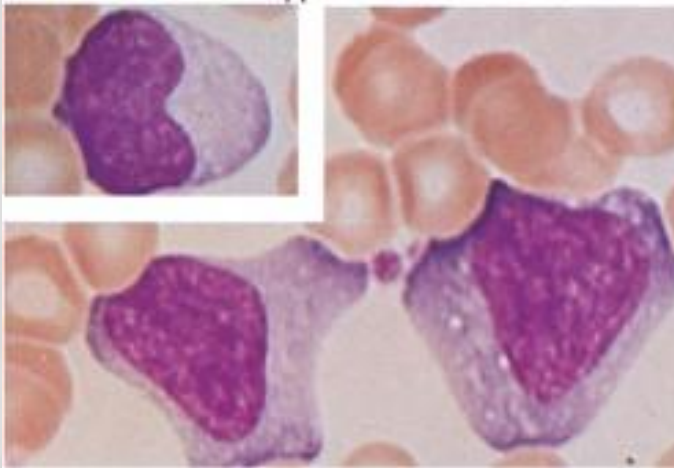
## Reactive lymphocytes



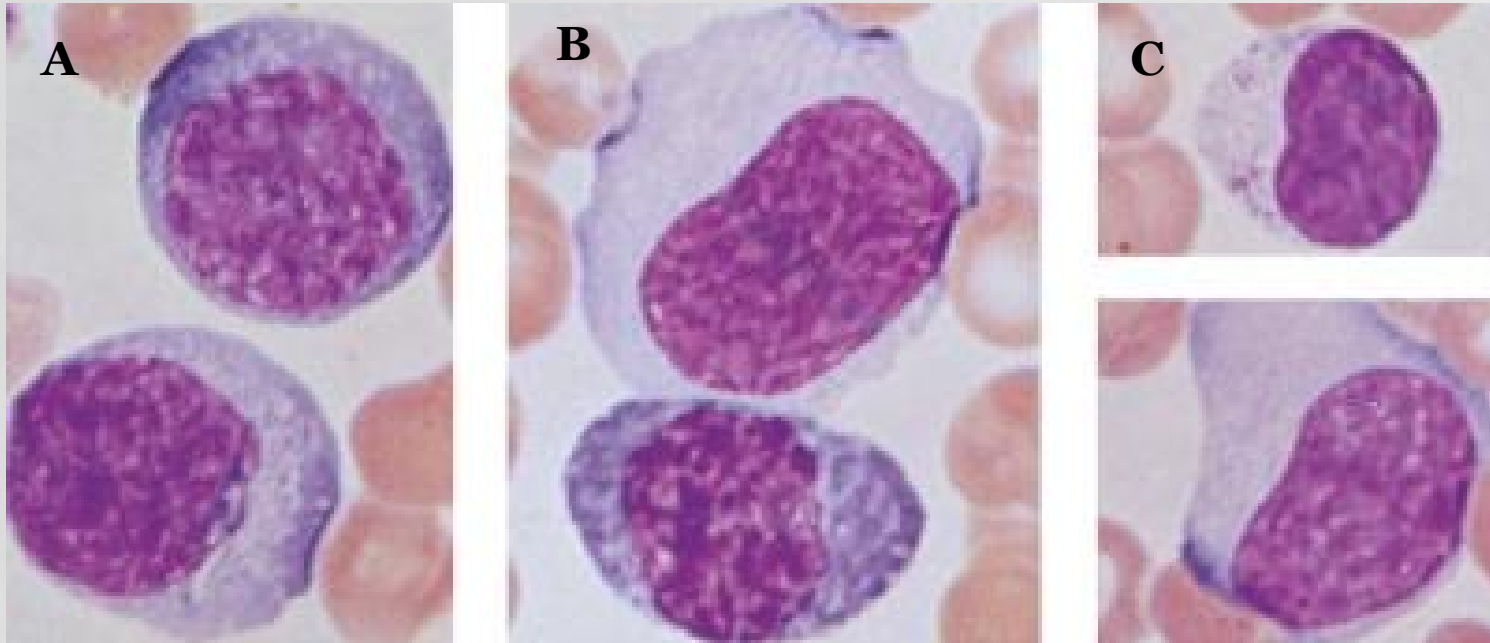
### From Color Atlas of Clinical Hematology

Infectious mononucleosis ; The cells are large with abundant vacuolated cytoplasm; the nuclei often show a fine blast-like chromatin pattern. The edges of the lymphocytes are often indented by adjacent red cells

## Reactive lymphocytes



## Reactive lymphocytes



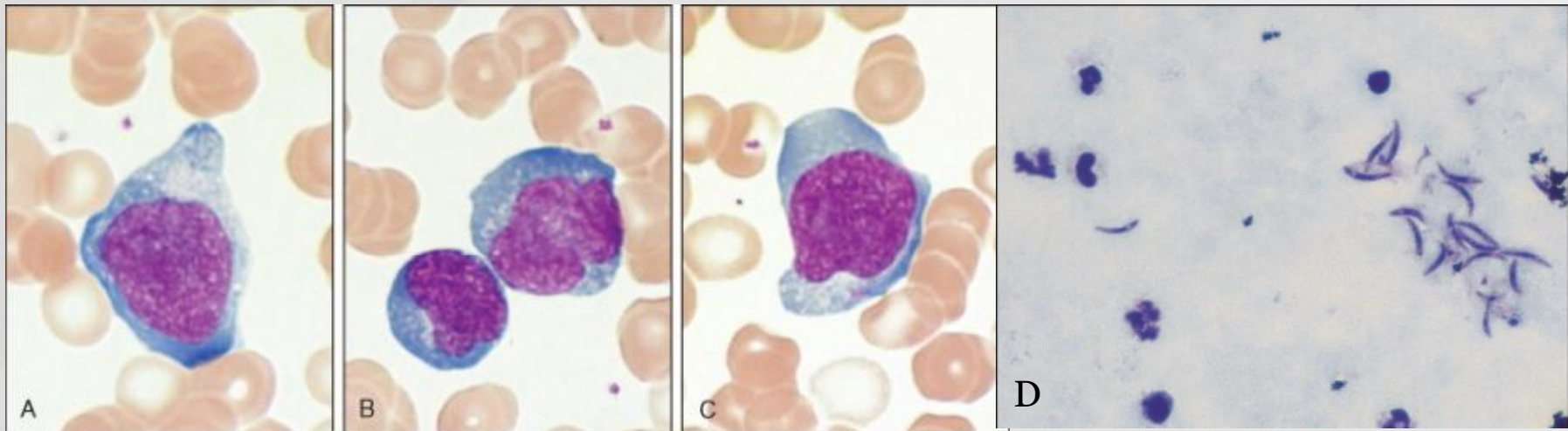
A, B : Lymphocytes resembling cells (plasmacytoid cells) in a patient with viral pneumonia

C : Lymphocytes with azurophilic granules

# Toxoplasmosis

Toxoplasmosis

- ; associated with lymphadenopathy, large number of atypical lymphocytes in the blood
- ; caused by the protozoan *Toxoplasma gondii*



Toxoplasmosis : (A~C) atypical lymphocytes in the blood

(D) trophozoite forms of *T. gondii* from a ruptured monocytes in thick PBS



# Lymphopenia

- caused by stress, drugs, irradiation, and some diseases
- below  $\approx 1.0 \times 10^9/L$  in adults  
below  $\approx 2.0 \times 10^9/L$  in children

## Key Causes of Lymphopenia

- Destructive – radiation, chemotherapy, corticosteroids
- Debilitative – starvation, aplastic anemia, terminal cancer, renal failure  
collagen vascular disease,
- Infectious – viral hepatitis, influenza, typhoid fever, TB
- AIDS associated – HIV cytopathic effect, nutritional imbalance, drug effect
- Congenital immunodeficiency – Wiskott-Aldrich syndrome
- Abnormal lymphatic circulation – intestinal lymphangiectasia, obstruction,  
thoracic duct drainage/rupture, CHF

## ACQUIRED IMMUNODEFICIENCY SYNDROME (AIDS)

AIDS is caused by infection with HIV, a retrovirus of the lentivirus subgroup

### ❖ HEMATOLOGIC MANIFESTATIONS

#### **Fall in CD4 lymphocyte count and lymphopenia**

Anemia, neutropenia, thrombocytopenia – either singly or combined

Bone marrow changes - variable cellularity, dysplastic change

Increased fibrosis, Granulomas (AFB, cryptococcal, uncertain)

Other infections [e.g., histoplasma, leishmania, pneumocystis]

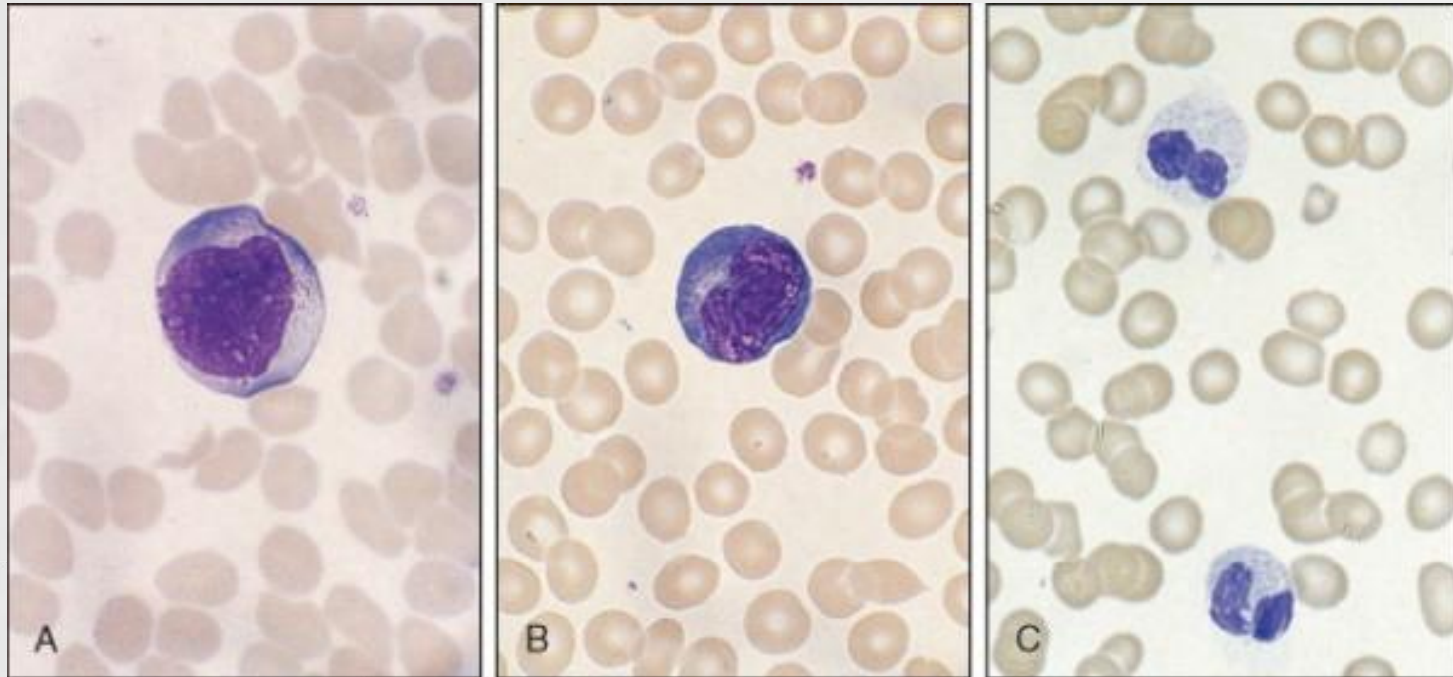
Benign nodules, Gelatinous degeneration,

Infiltration by lymphoma

Low serum vitamin B<sub>12</sub> and folate levels

Toxic change as a result of drugs





HIV infection: peripheral blood showing **(A, B)** immunoblasts and **(C)** pseudo-Pelger cells.

# **Malignant Lymphocyte Disorders**

Precursor lymphoid neoplasm

Mature B-cell neoplasms

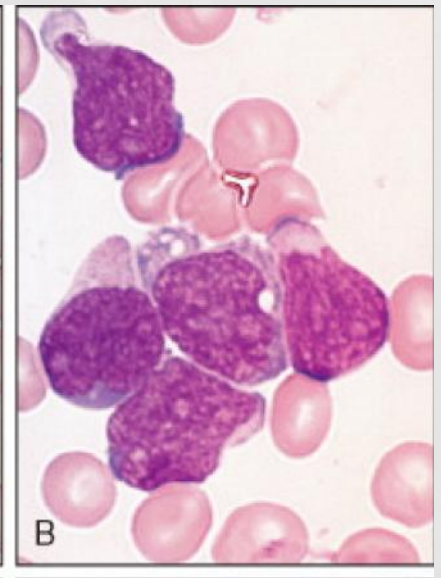
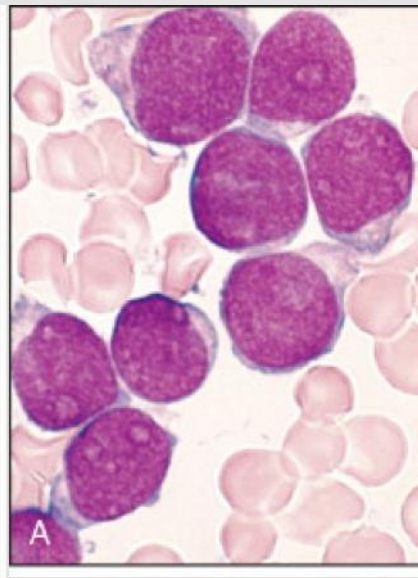
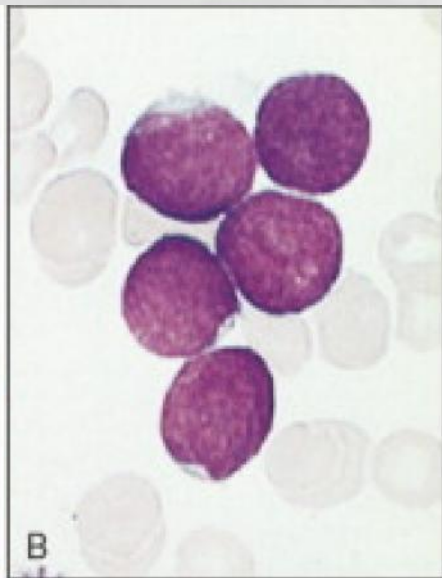
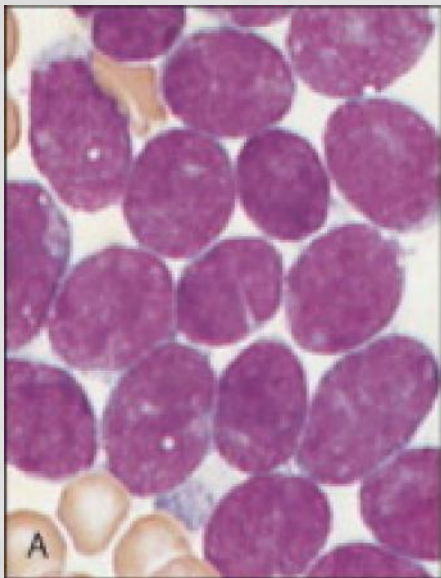
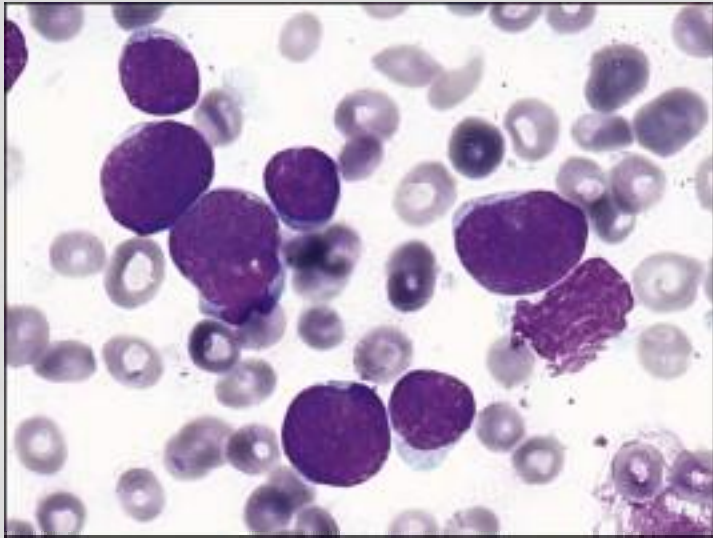
Mature T- and NK-cell neoplasms

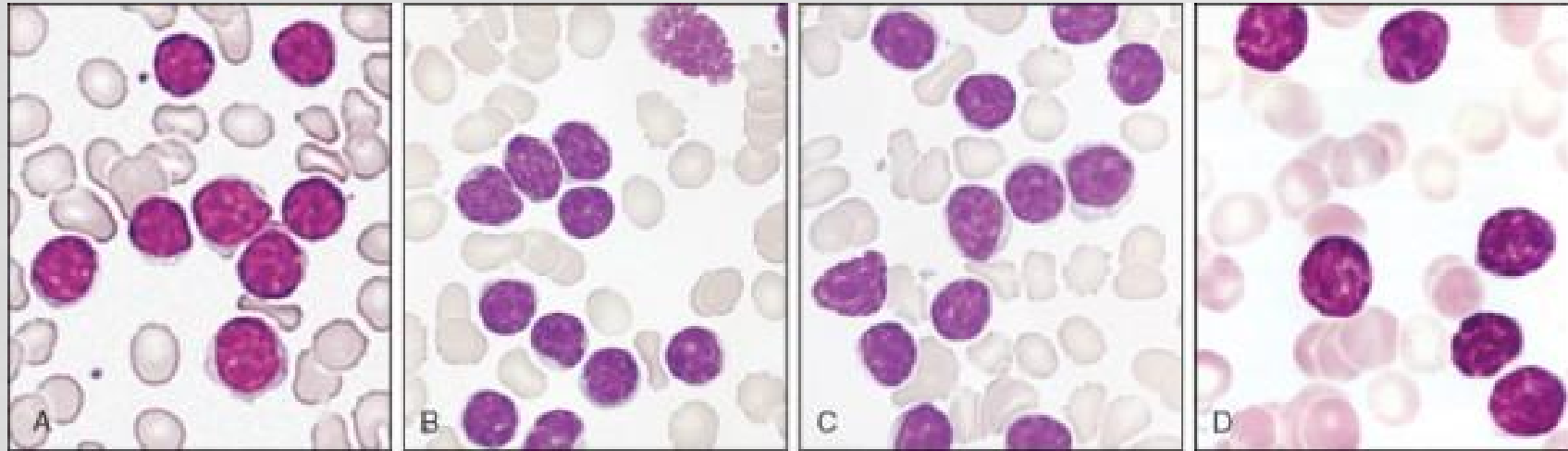
Hodgkin lymphoma

Immunodeficiency-associated lymphoproliferative disorders

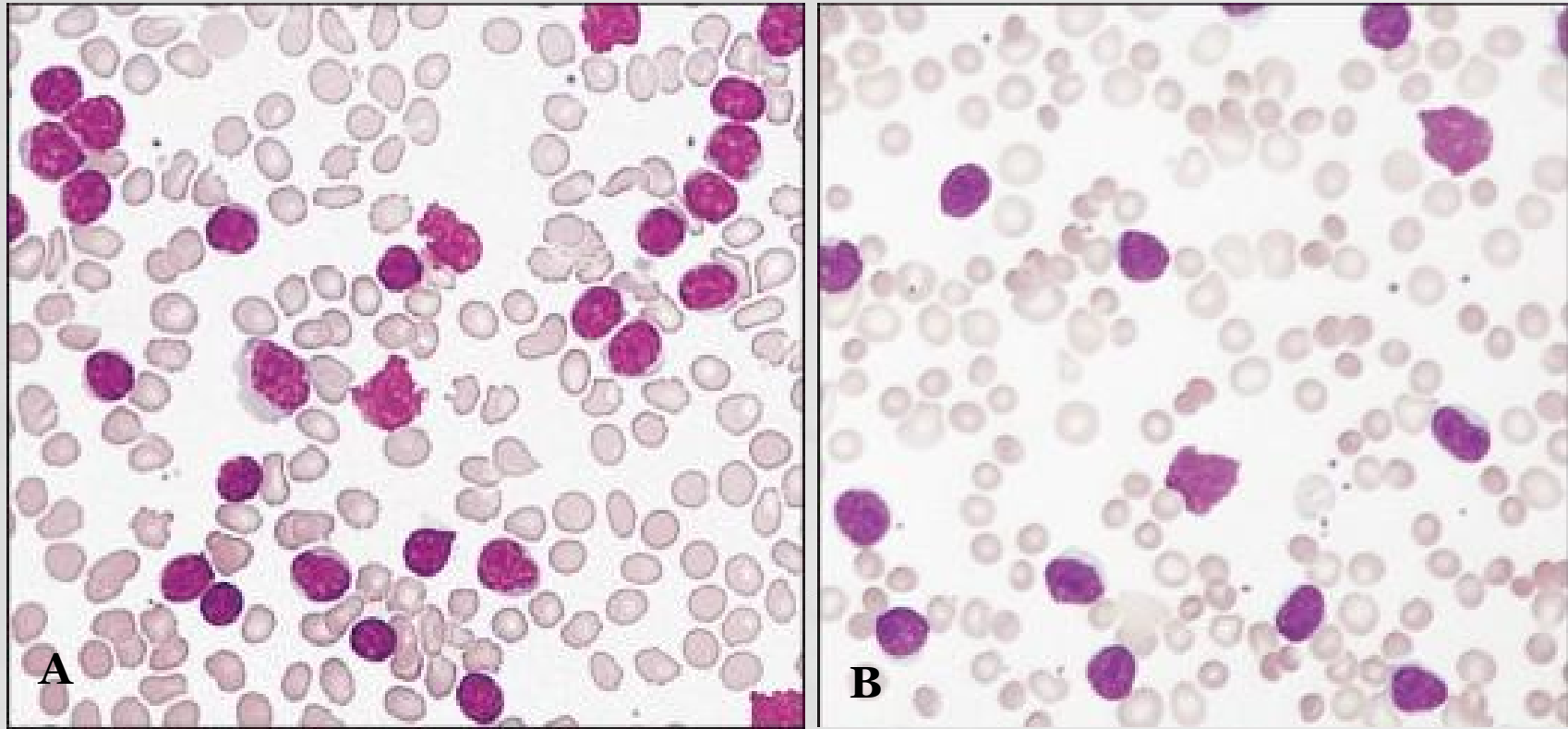
Histiocytic and dendritic cell neoplasms

## Precursor acute lymphoblastic leukemia



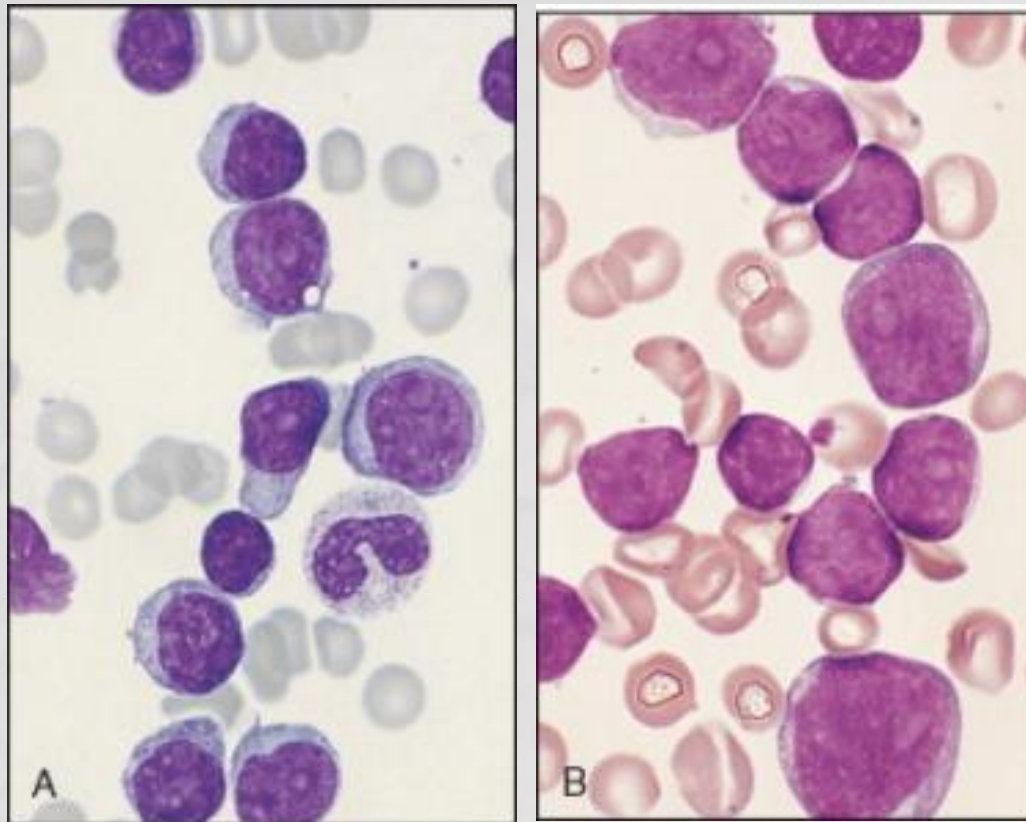


Chronic lymphocytic leukemia : Lymphocytes from the peripheral blood show thin rims of cytoplasm, condensed coarse chromatin, and only rare nucleoli.

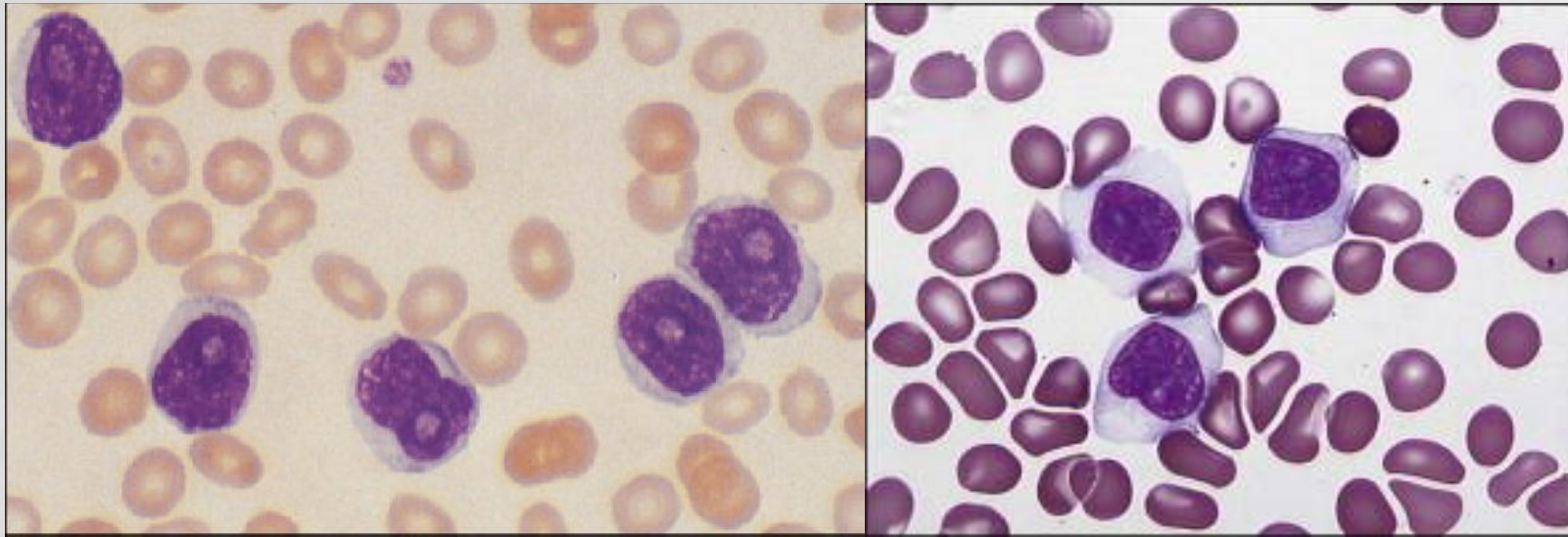


Chronic Lymphocytic Leukemia : (A)PBS showing the increased numbers of lymphocytes and occasional characteristic "smudge" cells. (B) CLL with autoimmune hemolytic Anemia; PBS shows increased numbers of lymphocytes, spherocytosis, and polychromasia.

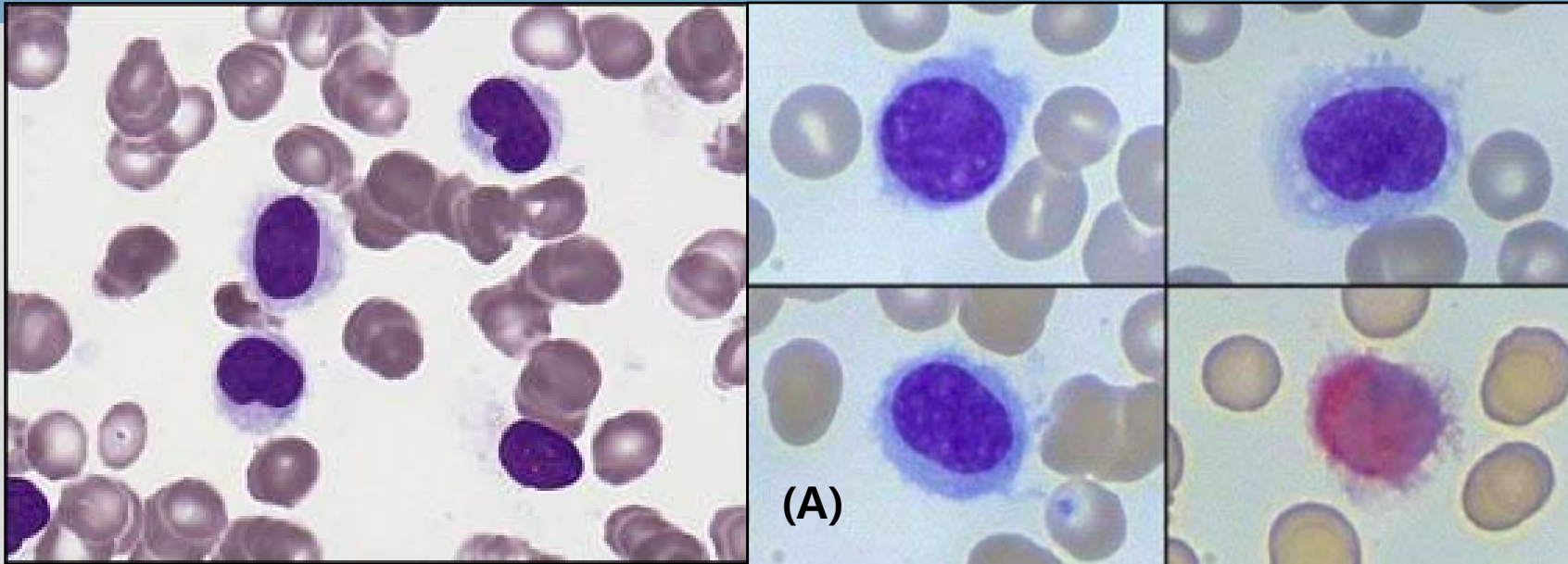




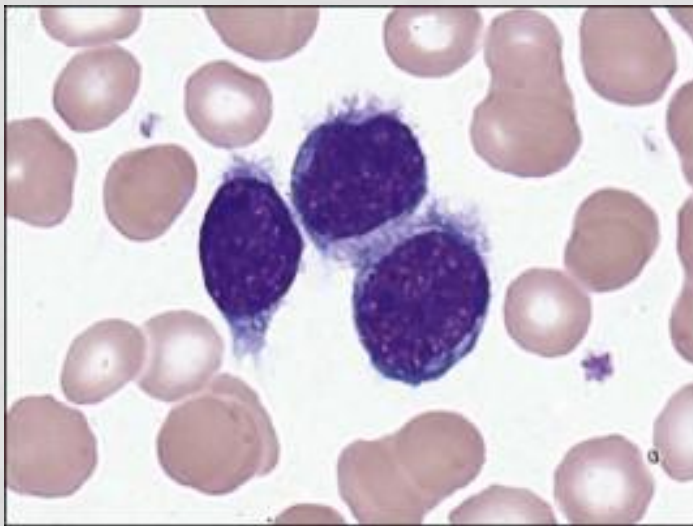
Chronic lymphocytic leukemia. **A, B**, Mixed cell type. The circulating lymphoid cells include greater than 10% but less than 55% prolymphocytes



B-cell prolymphocytic leukemia: PBS showing prolymphocytes that have prominent central nucleoli and an abundance of pale cytoplasm



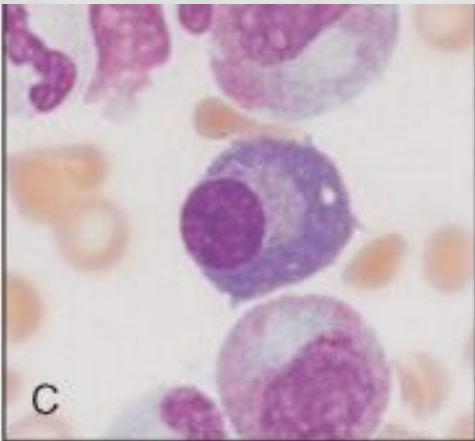
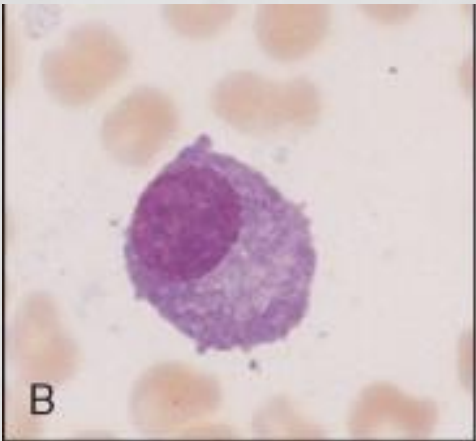
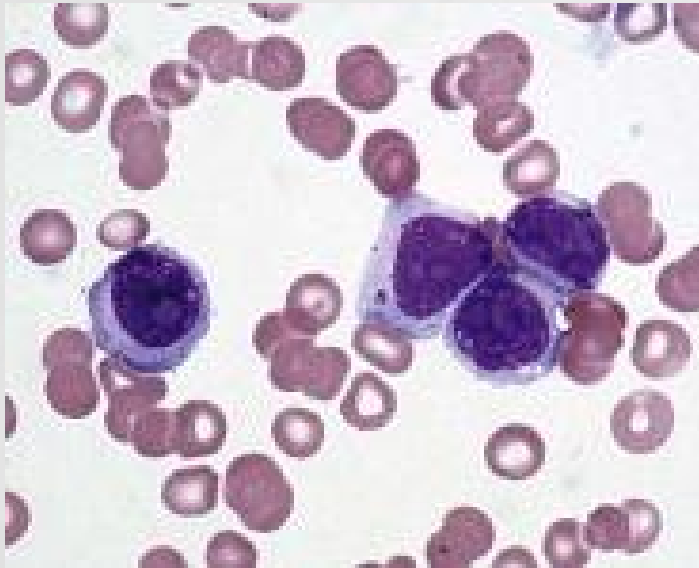
Hairy cell leukemia: typical cytochemical findings of hairy cells include  
**(A)** a strongly positive reaction to tartaric acid-resistant acid phosphatase (TRAP)

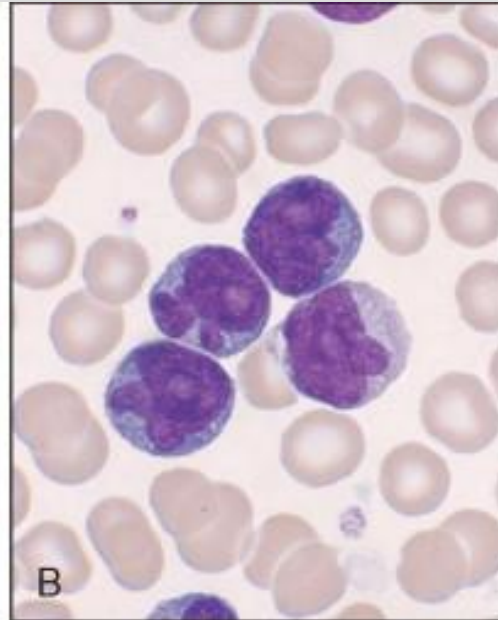
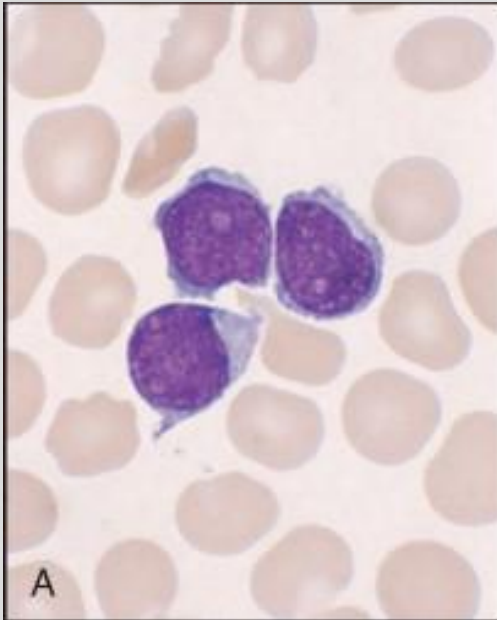


Villous lymphocytes (x1000)

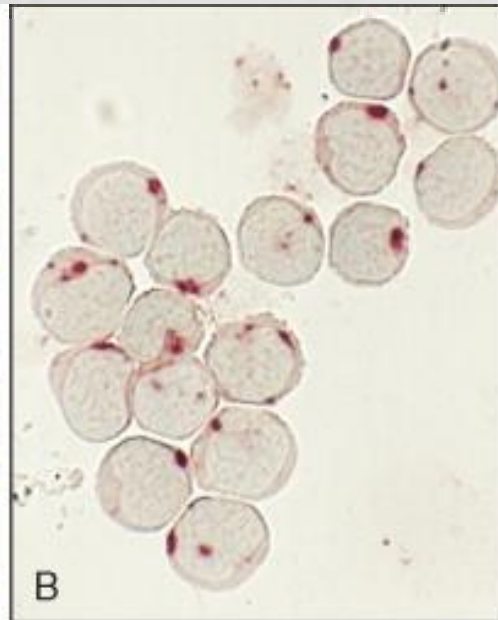
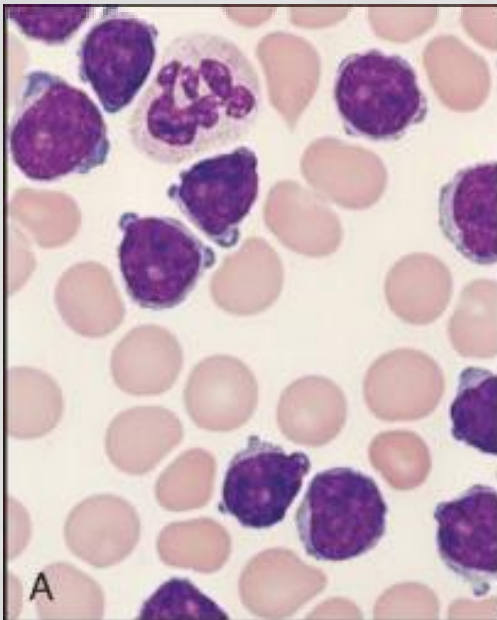


# Plasma cell in PBS or BM aspirates

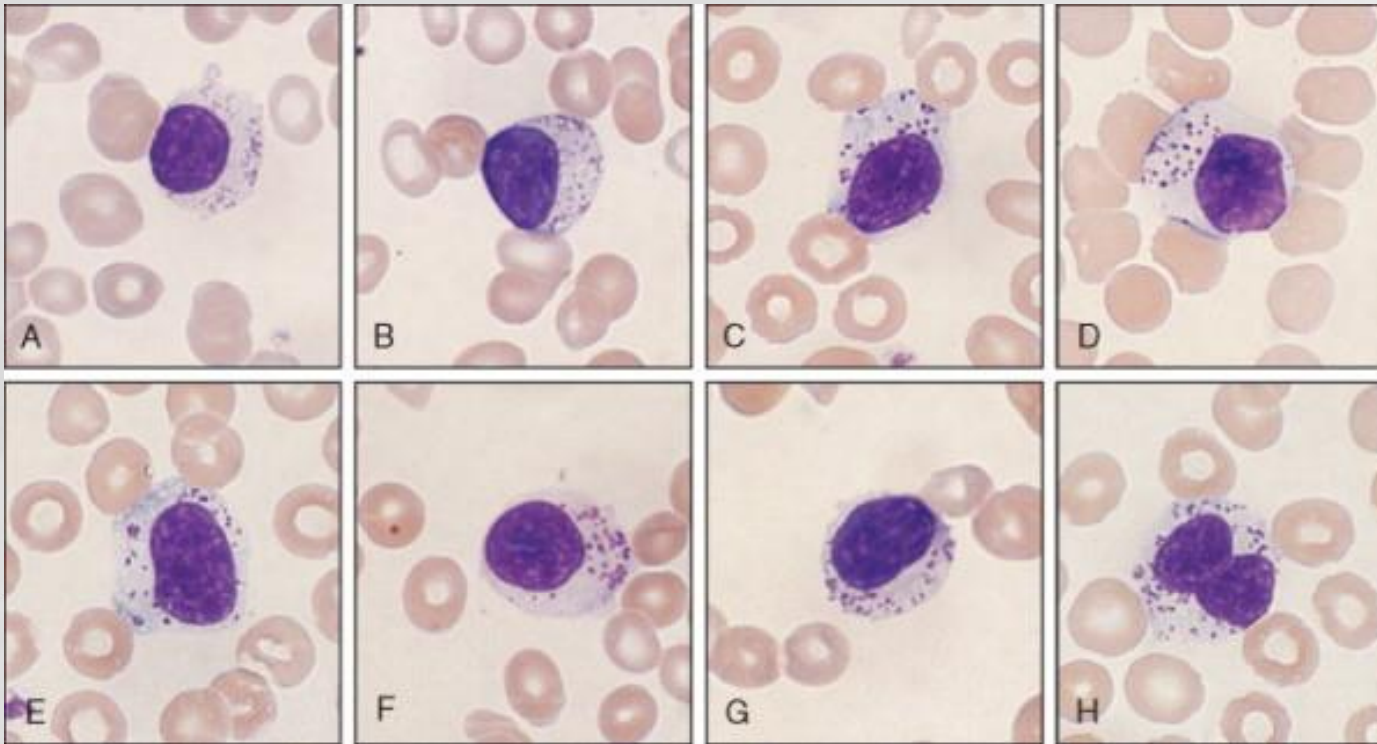




T-cell prolymphocytic leukemia.  
**A, B,** Small cell type with scant cytoplasm and irregular nuclear outline



T-cell prolymphocytic leukemia  
**(A)** Prolymphocytes with a prominent central nucleolus  
**(B)** "clump" positive in acid phosphatase stain

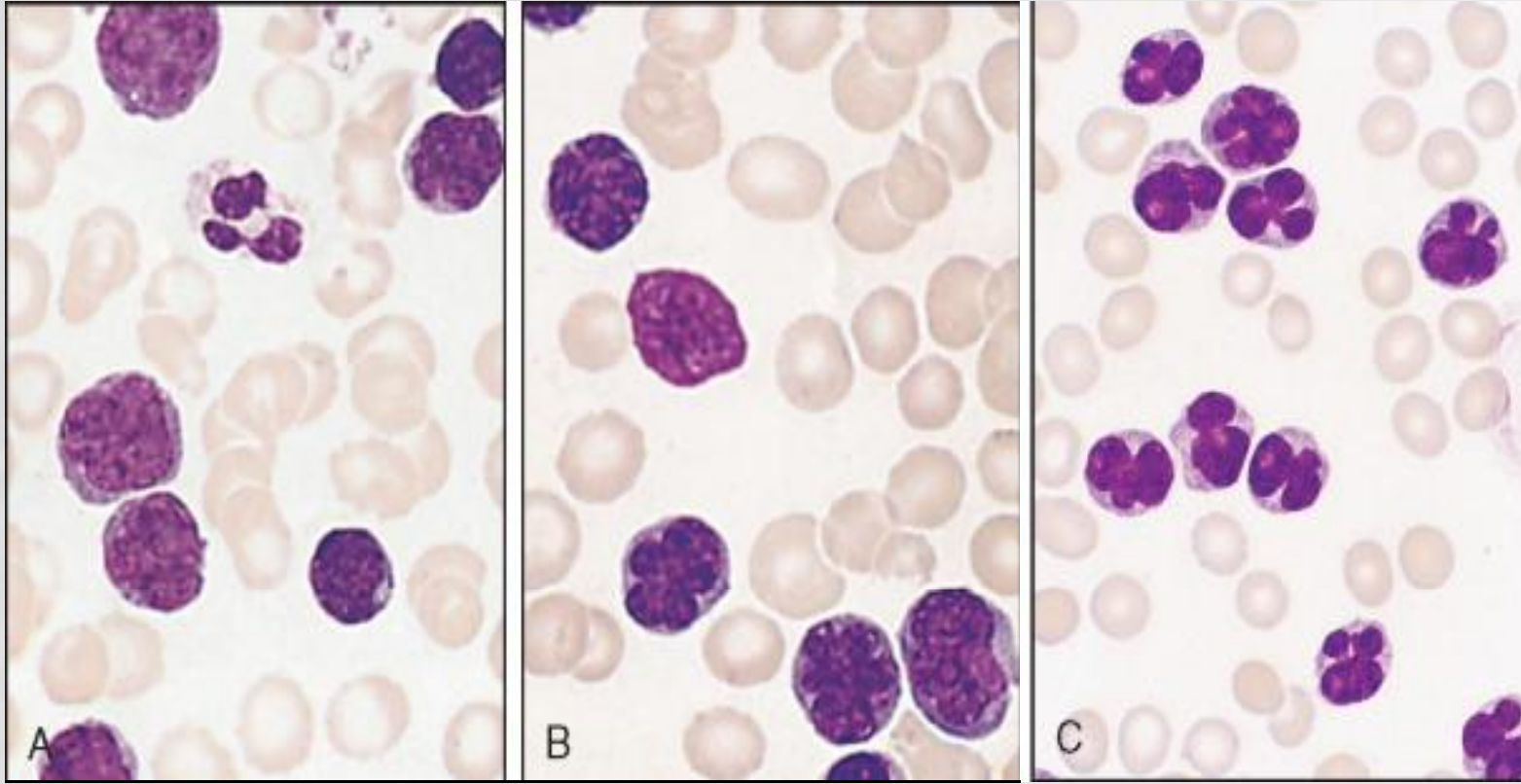


Large granular lymphocytic leukemia :

Large lymphocytes with multiple coarse, azurophilic, cytoplasmic granules.

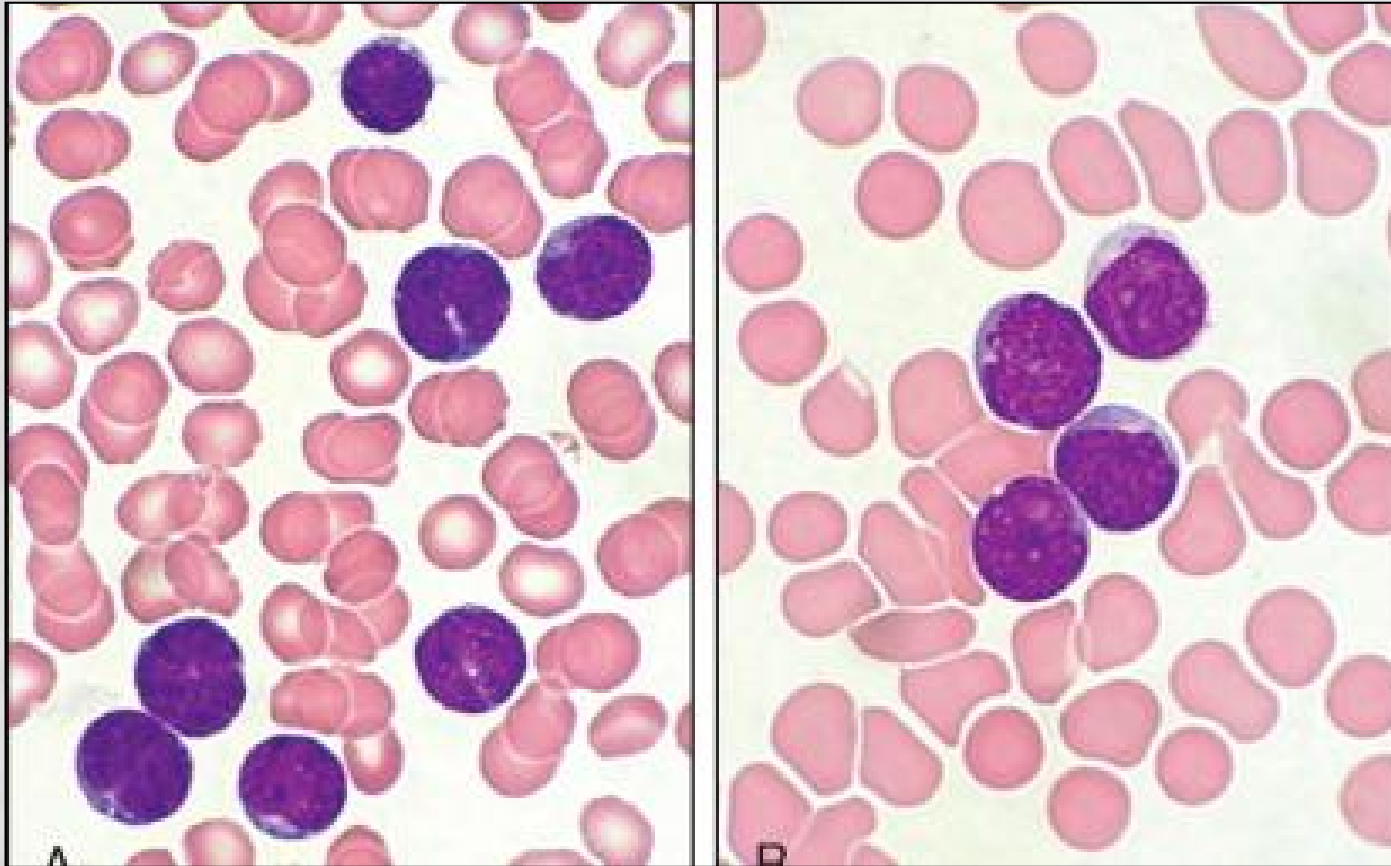
These cells are positive with CD8+, CD4-, CD3+, CD16+, and CD57+.

The patient had splenomegaly, chronic neutropenia, and lymphocytosis

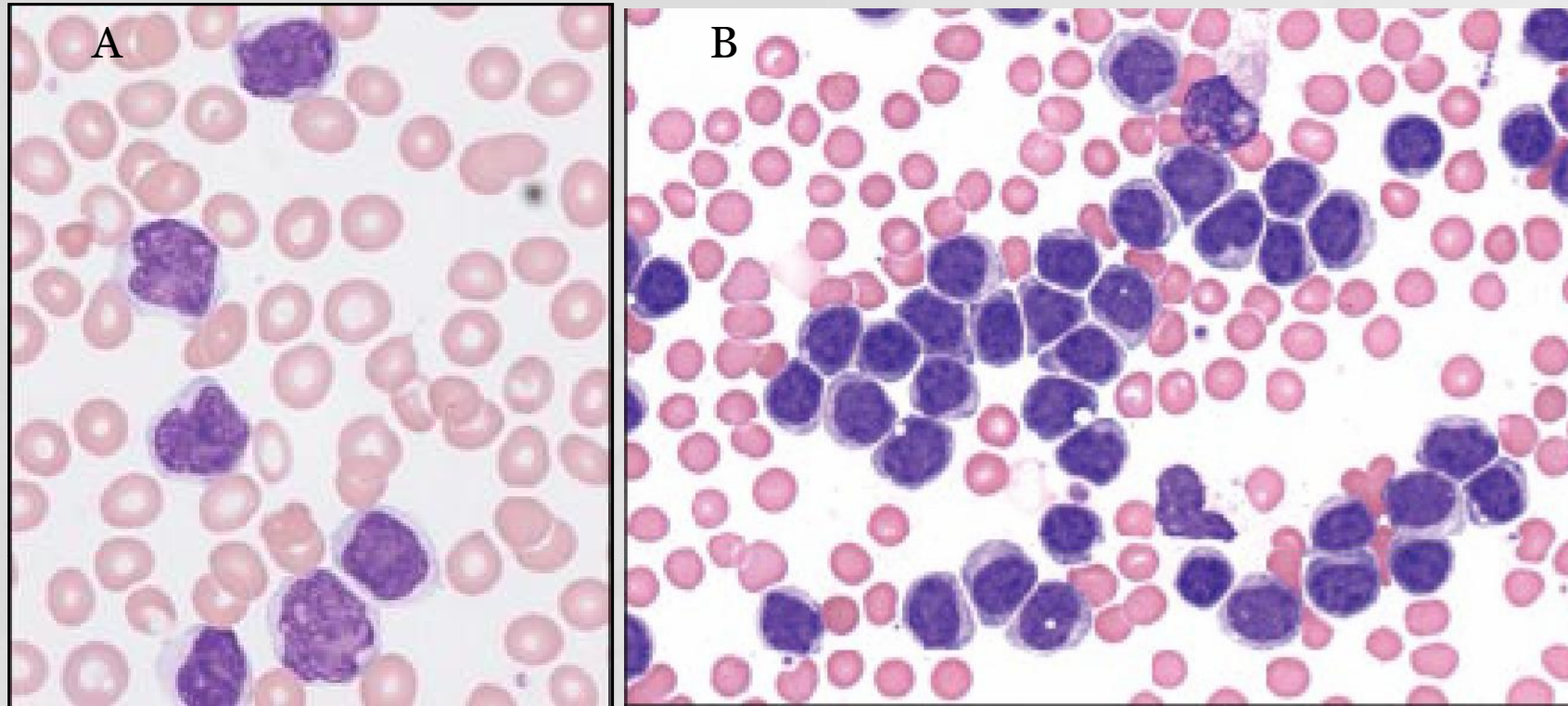


Adult T-cell leukemia/lymphoma syndrome.  
PBS showing the characteristic abnormal lymphocytes with convoluted nuclei.



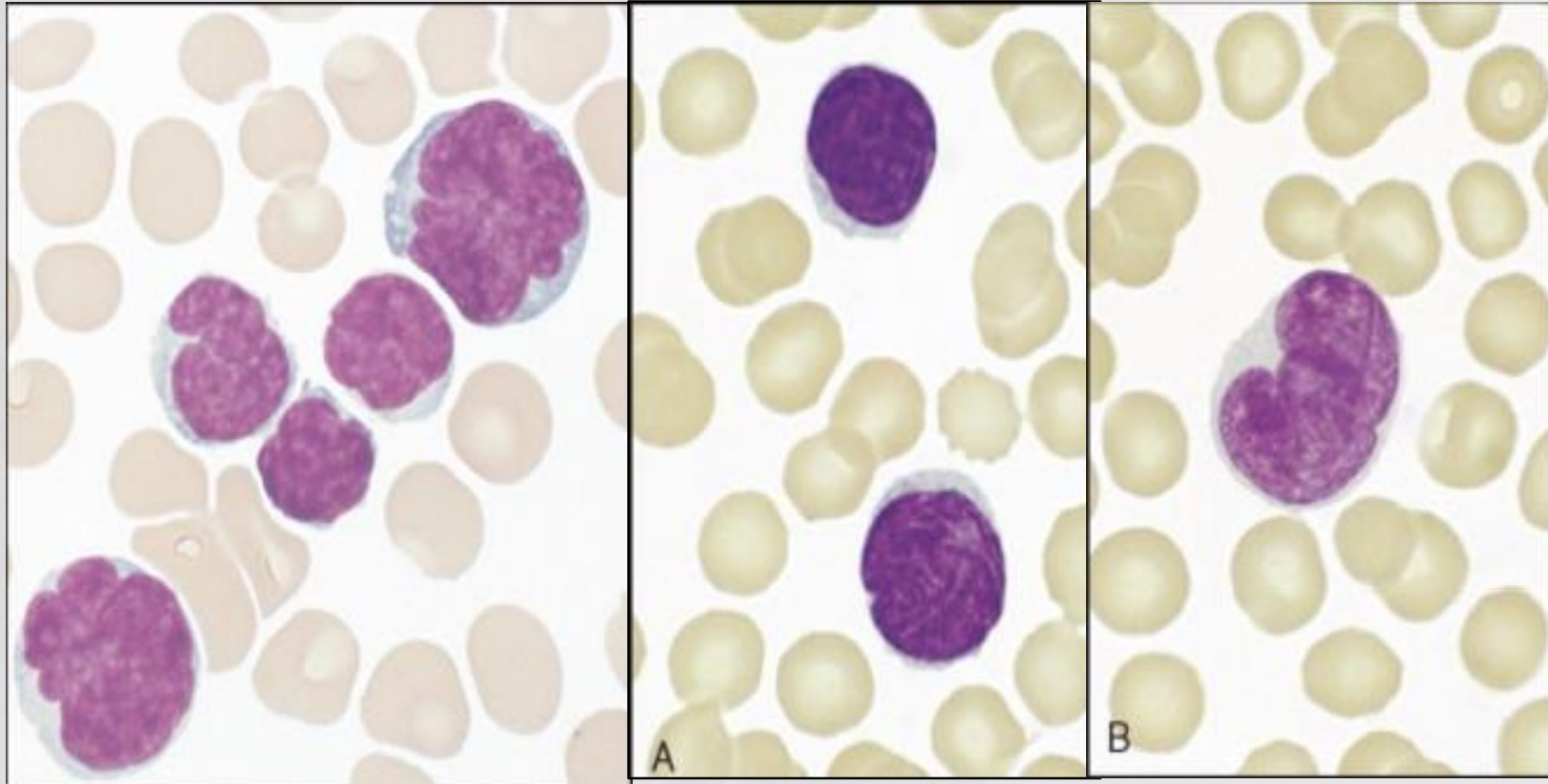


Follicular lymphoma: PBS shows presence of small lymphoid cells with nuclear clefts, diffuse nuclear chromatin, and scant, darkly staining cytoplasm.

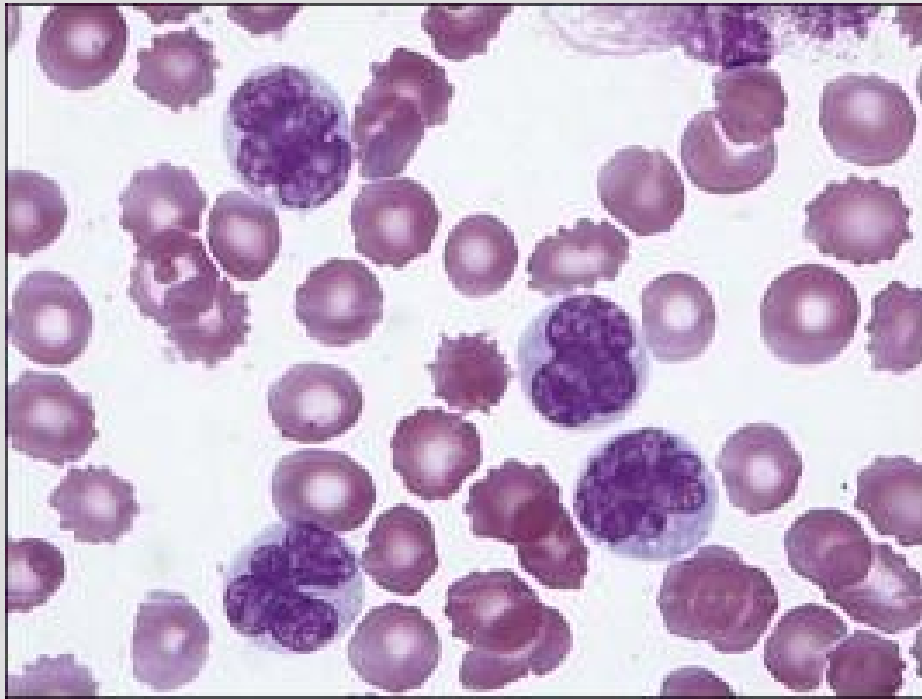


### Mantle cell lymphoma

(A) PBS showing medium-sized lymphocytes with irregular nuclear contours and scant pale cytoplasm. (B) BM aspirate showing diffuse infiltration with medium-sized lymphoid cells

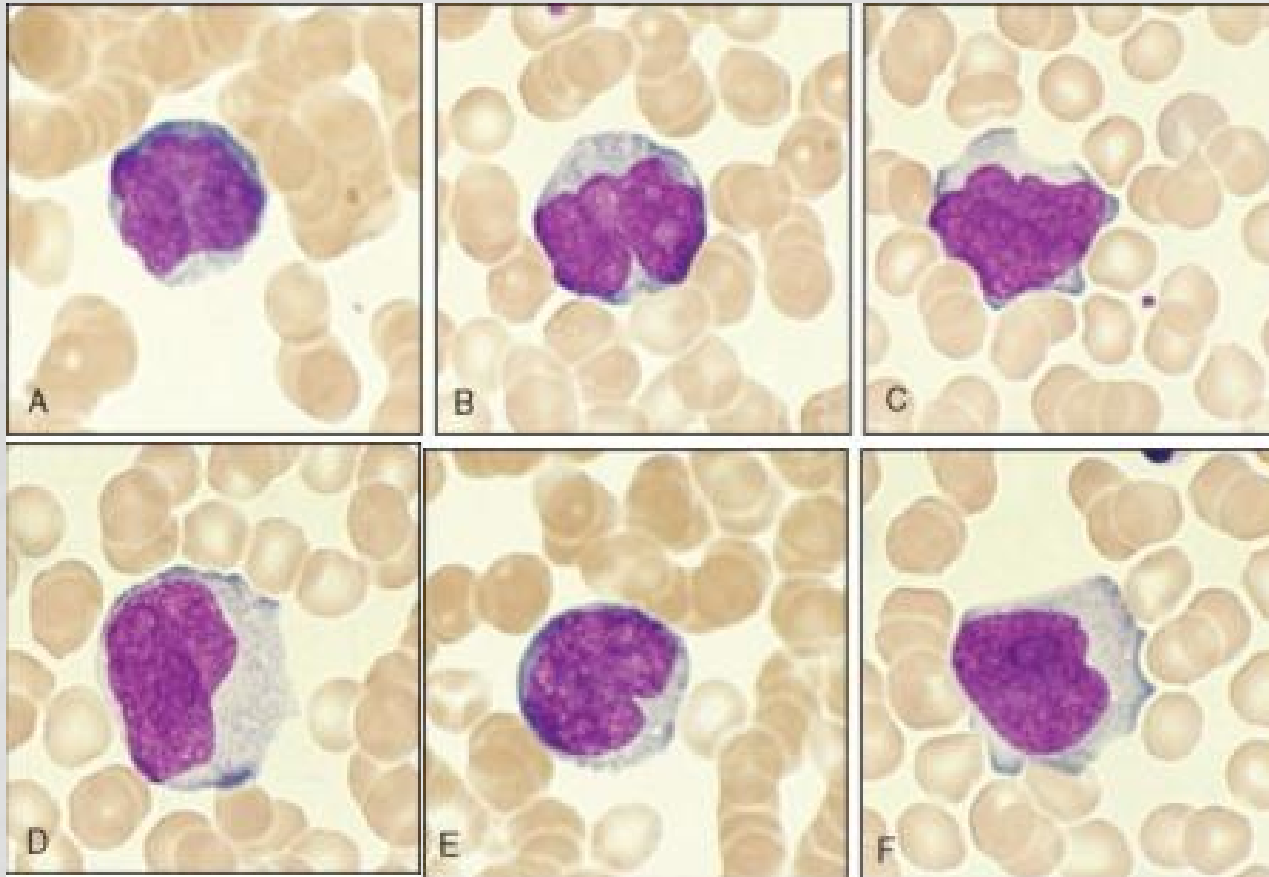


Sézary syndrome: abnormal cells in the peripheral blood have characteristic, cerebriform, large, and clefted nuclei with fine chromatin pattern and scanty cytoplasm.



Natural killer cell leukemia in a adult, with  
florete cells resembling adult T cell leukemia

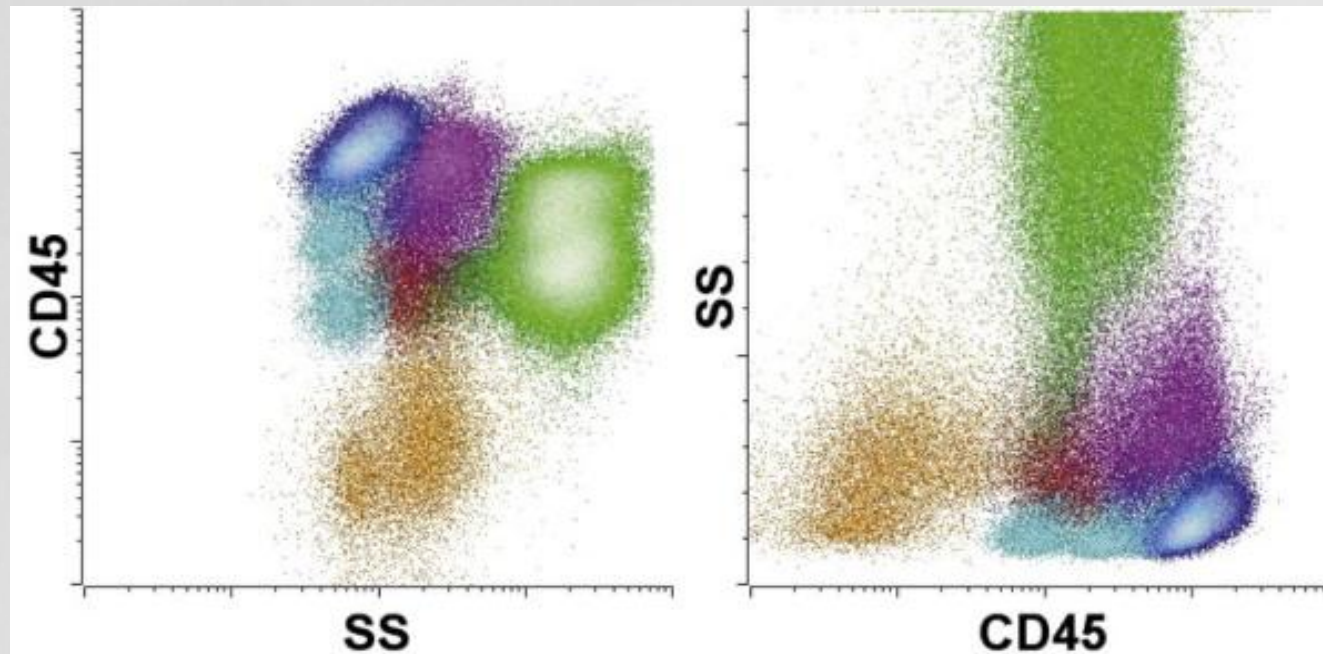




Peripheral T-cell lymphoma unspecified :  
PBS showing atypical lymphoma cells

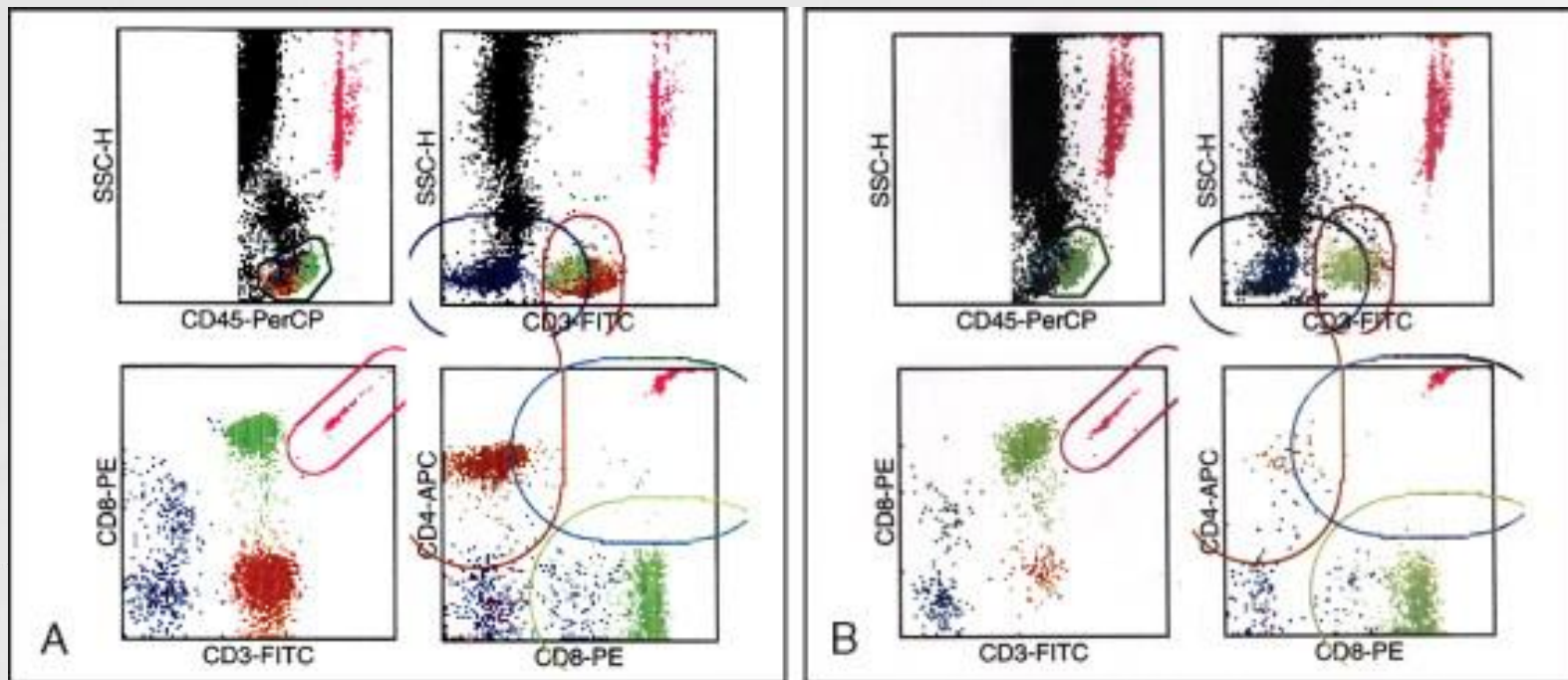
## Immunophenotyping by Flow cytometry

Flow cytometry ; a powerful, rapid, and cost effective technique for the identification and monitoring of hematopoietic cells(neoplasms)



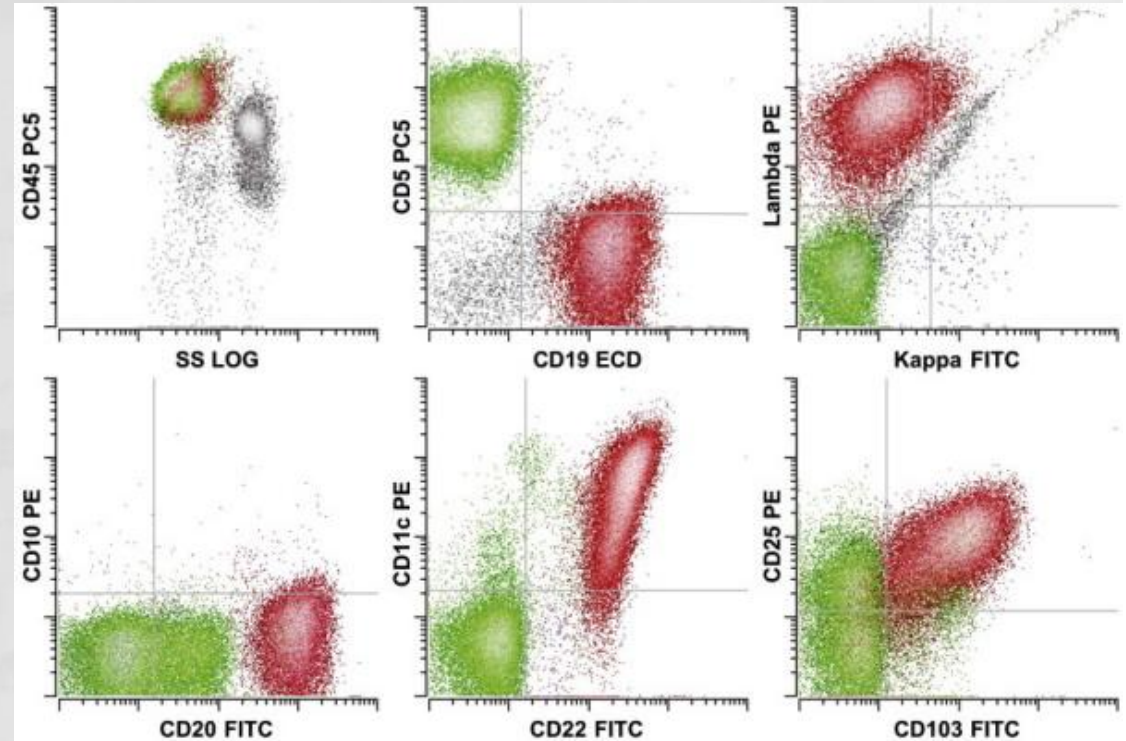
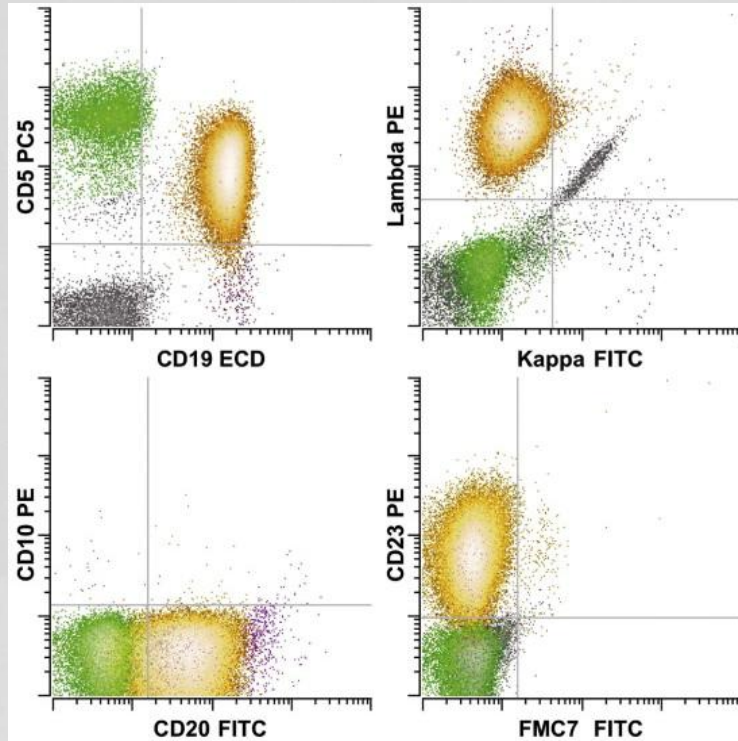
CD45 versus side scatter : used in the clinical laboratory as a starting point for the identification of white blood cell populations.

Major white cell populations colored are lymphocytes (*blue*), B cells (*light blue*), monocytes (*purple*), maturing neutrophils (*green*), blasts (*red*), basophils (*dark purple*), and maturing erythrocytes (*orange*).



HIV Infection: 4 color flow cytometry measurement of peripheral blood lymphocyte subpopulations. **A**, normal; **B**, HIV infection.

# Immunophenotyping by Flow cytometry



Chronic lymphocytic leukemia ; B cell

Hairy cell leukemia



## Summary

### Lymphocytes : reactive or malignant ?

- number of cells : absolute or relative
- morphology
- duration : transient or persistent
- clinical features : age, lymphadenopathy, symptoms 등등
- serologic test
- flow cytometry : clonality
- chromosome study
- bone marrow study

### Reference

Henry's Clinical Diagnosis and Management by Laboratory Methods (22<sup>nd</sup> eds)

Color Atlas of Clinical Hematology (4<sup>th</sup> eds)

Wintrobe's Clinical Hematology (12<sup>th</sup> eds)