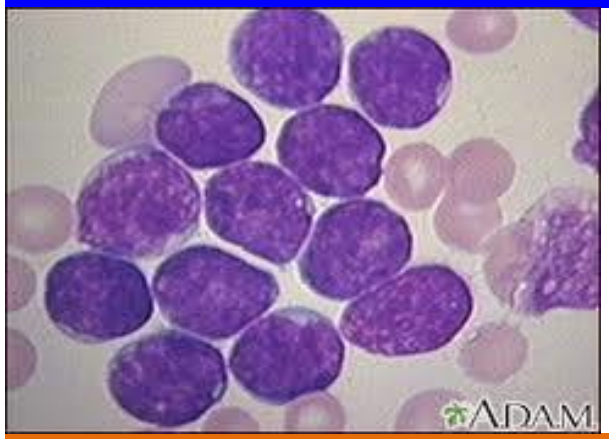
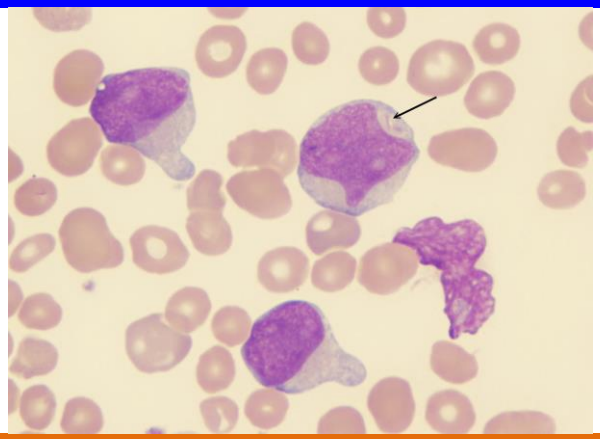
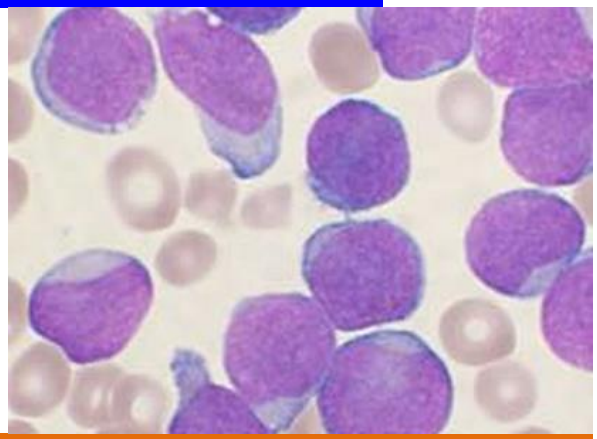
Queen Sirikit National Institute of Child Health

Acute Leukemia



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Department of Pediatrics
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Outlines

- Overview of human hematopoiesis
- Acute leukemia of childhood
  - **Leukemias**
    - Acute lymphoblastic leukemia (ALL)
    - Acute myeloid leukemia (AML)
  - Signs and symptoms
  - Investigations
  - Treatment

Most Common Pediatric Cancers

Age 0-14	
Leukemia	**32%**
CNS	20
Lymphoma	11
Neuroblastoma	8
Rhabdo/STS	7
Kidney	6
Bone	6
Germ cell	4
Retinoblastoma	3
Liver	1

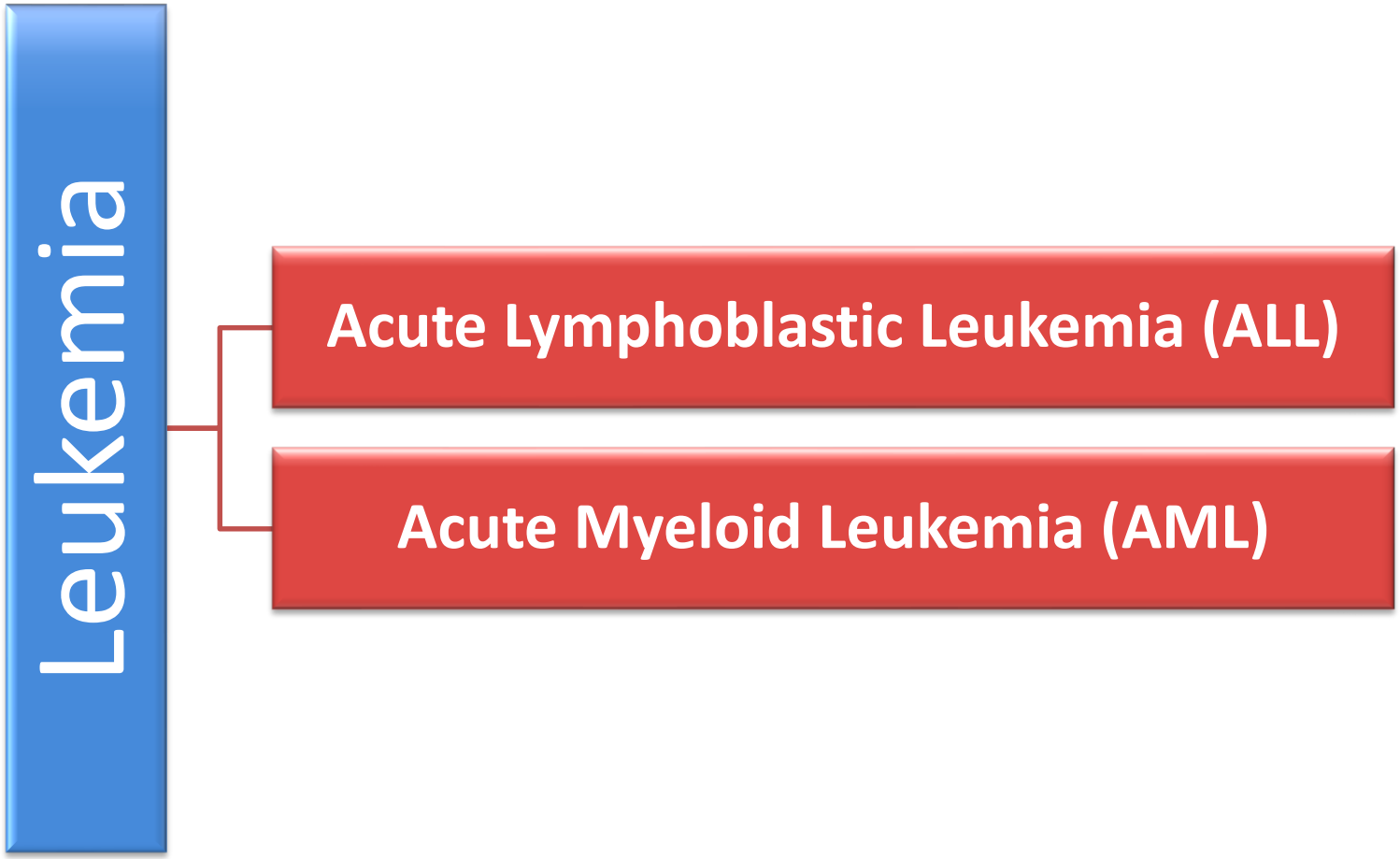
Age 15-19	
Lymphoma	25%
Germ cell	14
Leukemia	**12**
CNS	10
STS	8
Bone	8
Thyroid cancer	7
Melanoma	7

Leukemia

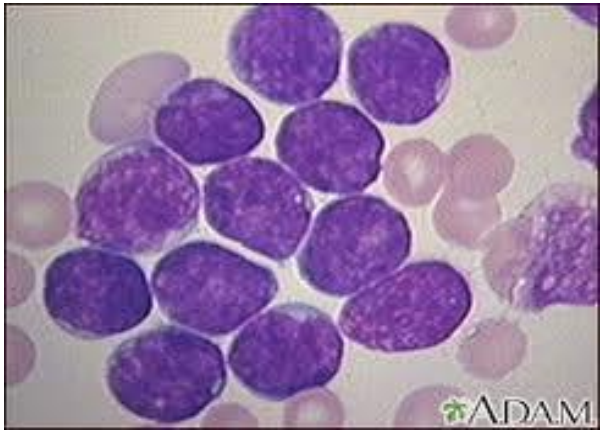
Leukemia = Leuk + emia

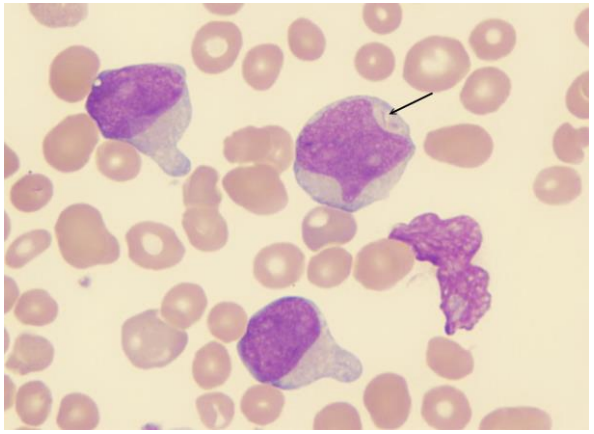
(white) (blood)

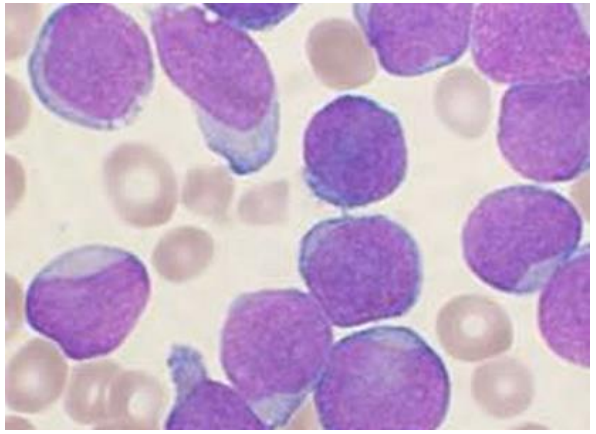
Pediatric leukemia



Pediatric leukemia

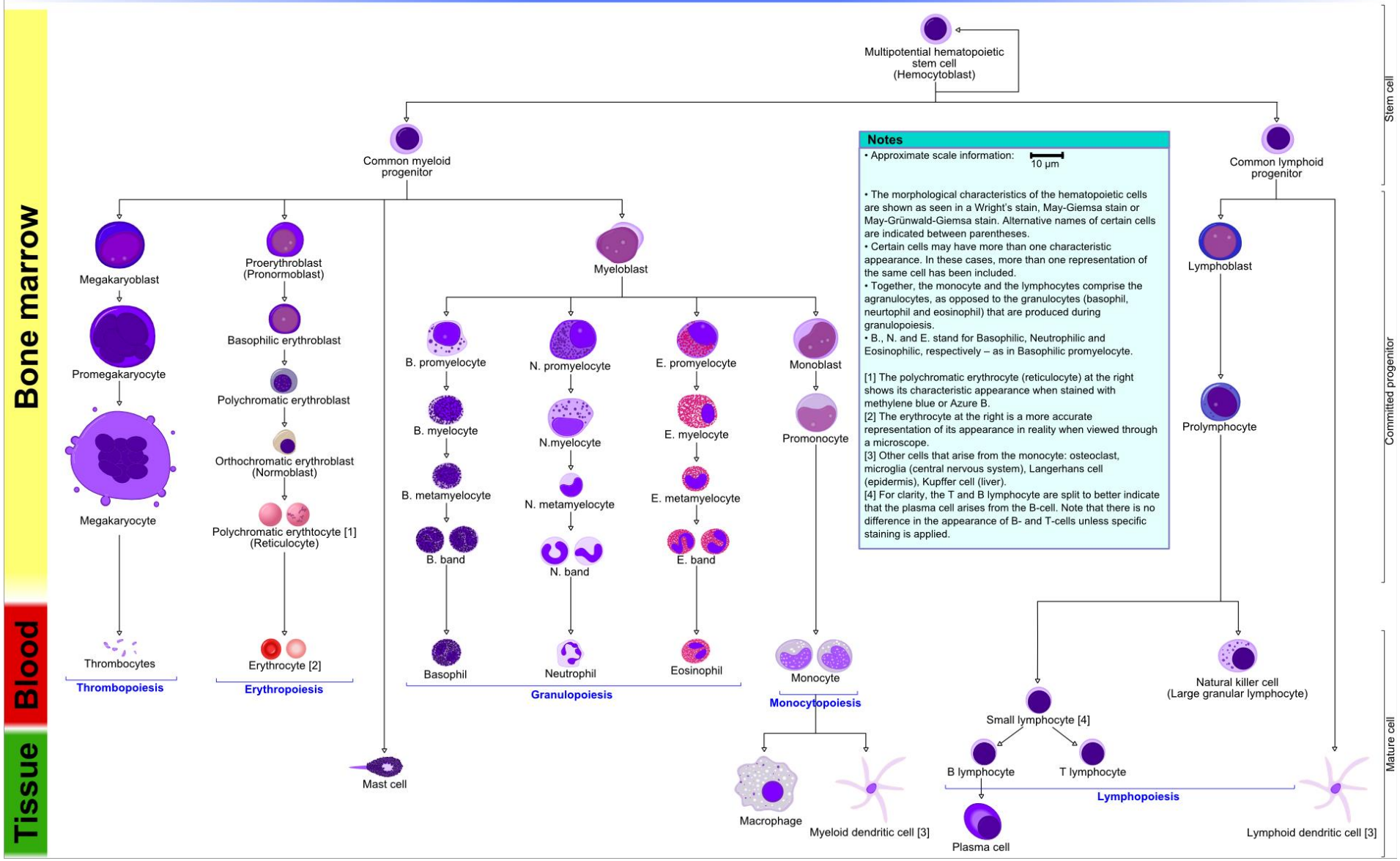




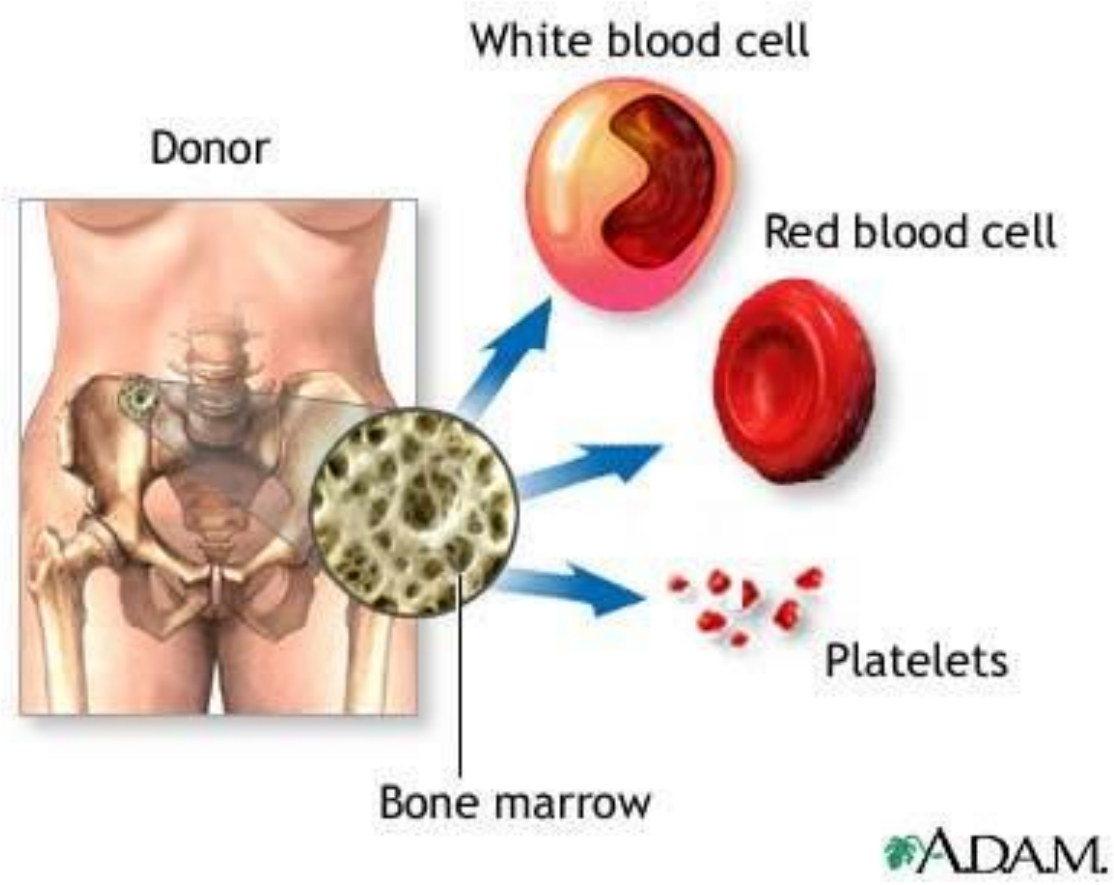


ALL ?? AML

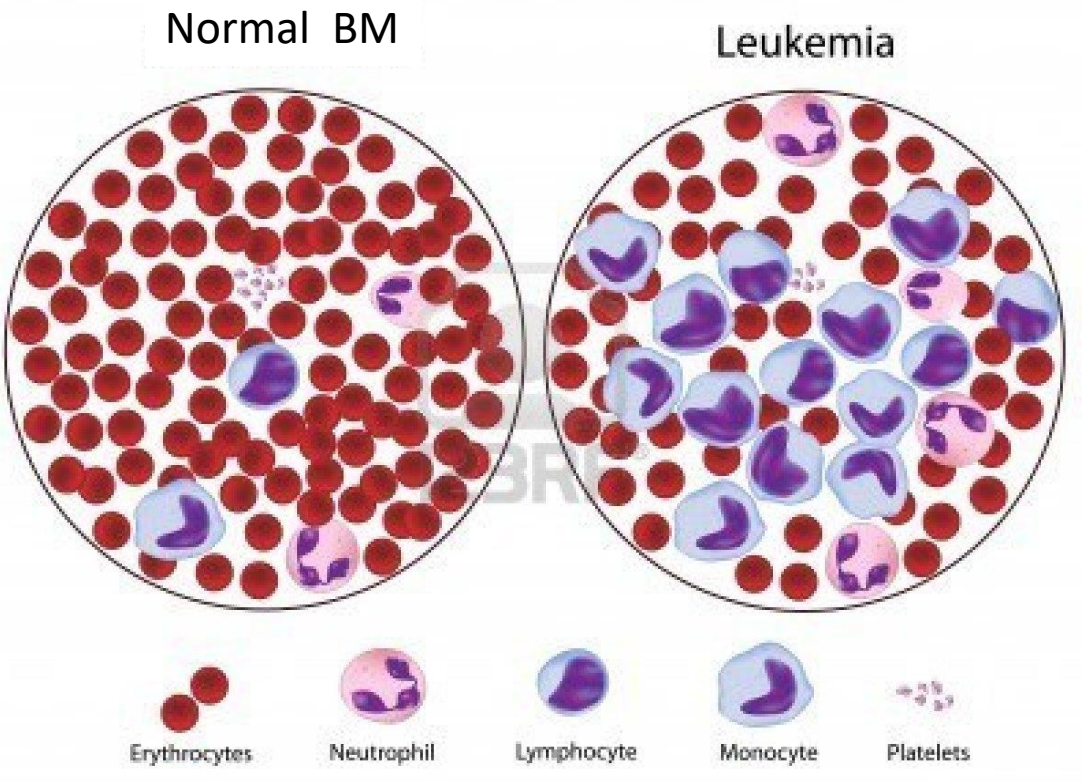
Hematopoiesis in humans

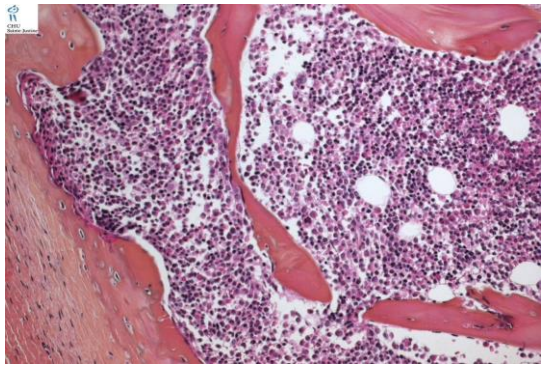


Leukemia

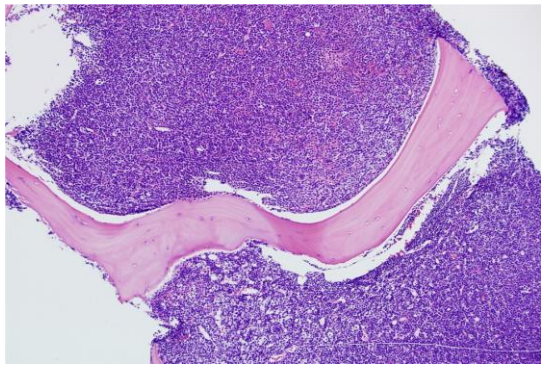

White blood cell
Donor
Red blood cell
Platelets
Bone marrow
A.D.A.M.

Acute Leukemia





Normocellular marrow



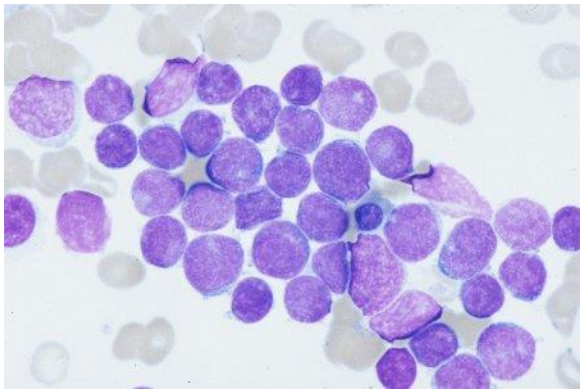
Hypercellular marrow

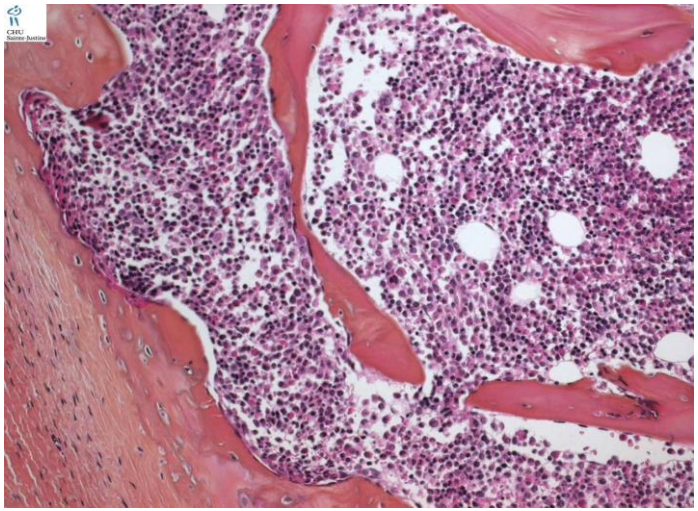
Clinical Presentations

Fatigue Weakness Anorexia	**RBC**
Fever Prolonged infection	**WBC**
Easy bruising Bleeding	**Platelets**
Lymphadenopathy Hepatosplenomegaly	**RE system**
Testicular & CNS involvement	

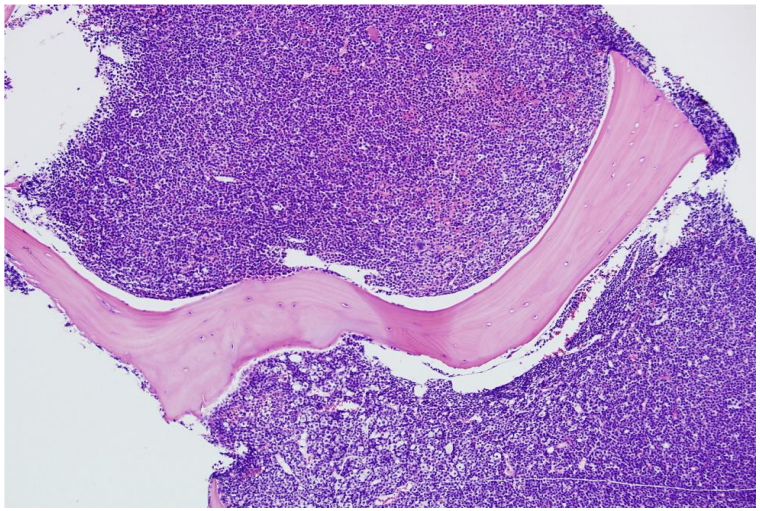
Acute Lymphoblastic Leukemia

- Most common malignancy of childhood
- 75% of all childhood leukemia
- Peak incidence ages 2-5 yrs
- Male : Female = 1 : 1.2
- Overall 5 year survival rate is >80%





Normal bone marrow



Bone marrow with leukemic infiltrates

Clinical Characteristics of 724 Children with ALL (CCSG)

Clinical characteristics	Percent (%)
Age (years) distribution	
<1 1 – 3 3 – 10 >10	6 18 54 22
General symptoms	
Fever Bleeding Bone pain	61 48 23
Lymphadenopathy	
None Moderate Extended	50 43 7
Splenomegaly	
None Moderate Extended	37 49 14
Hepatosplenomegaly	
None Moderate Extended	32 55 13
Mediastinal enlargement	7

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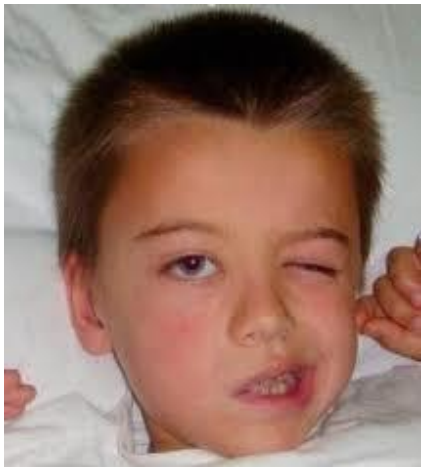
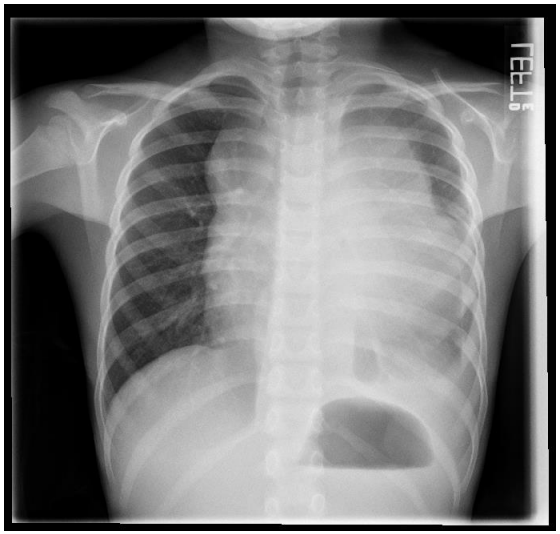
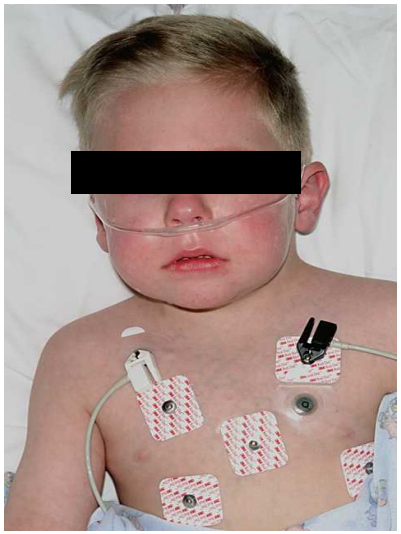
Specific Signs & Symptoms

Leukemia cutis (AML-M5 > ALL)



CNS leukemia (<5% at diagnosis)
- **CNS1 : no lymphoblasts**
- **CNS2 : <5 cells/cm3 with blasts on cytospin**
- **CNS3 : ≥5 cells/cm3 with blasts or CN palsy**





Anterior mediastinal mass with
Superior vena cava syndrome

ALL – Diagnosis

Supportive

- CBC
- PBS
- Type & crossmatch for blood and platelets
- Hemoculture & urine culture
- Tumor lysis labs
- CXR

Specific

- Bone marrow aspirate and biopsy
  - Morphology
  - Immunophenotyping
  - Cytogenetics
- Lumbar puncture
  - Cell count
  - Cytospin
  - Intrathecal chemotherapy

Blood Cell Counts in ALL

Blood cell counts	Level (s)	Percent (%)
Hemoglobin (g/dL)	<7 7 – 11 >11	43 45 12
WBC (cells/mm3)	<10,000 10,000 – 49,000 >50,000	53 30 17
Platelet (cells/mm3)	<20,000 20,000 – 99,000 >100,000	28 47 25

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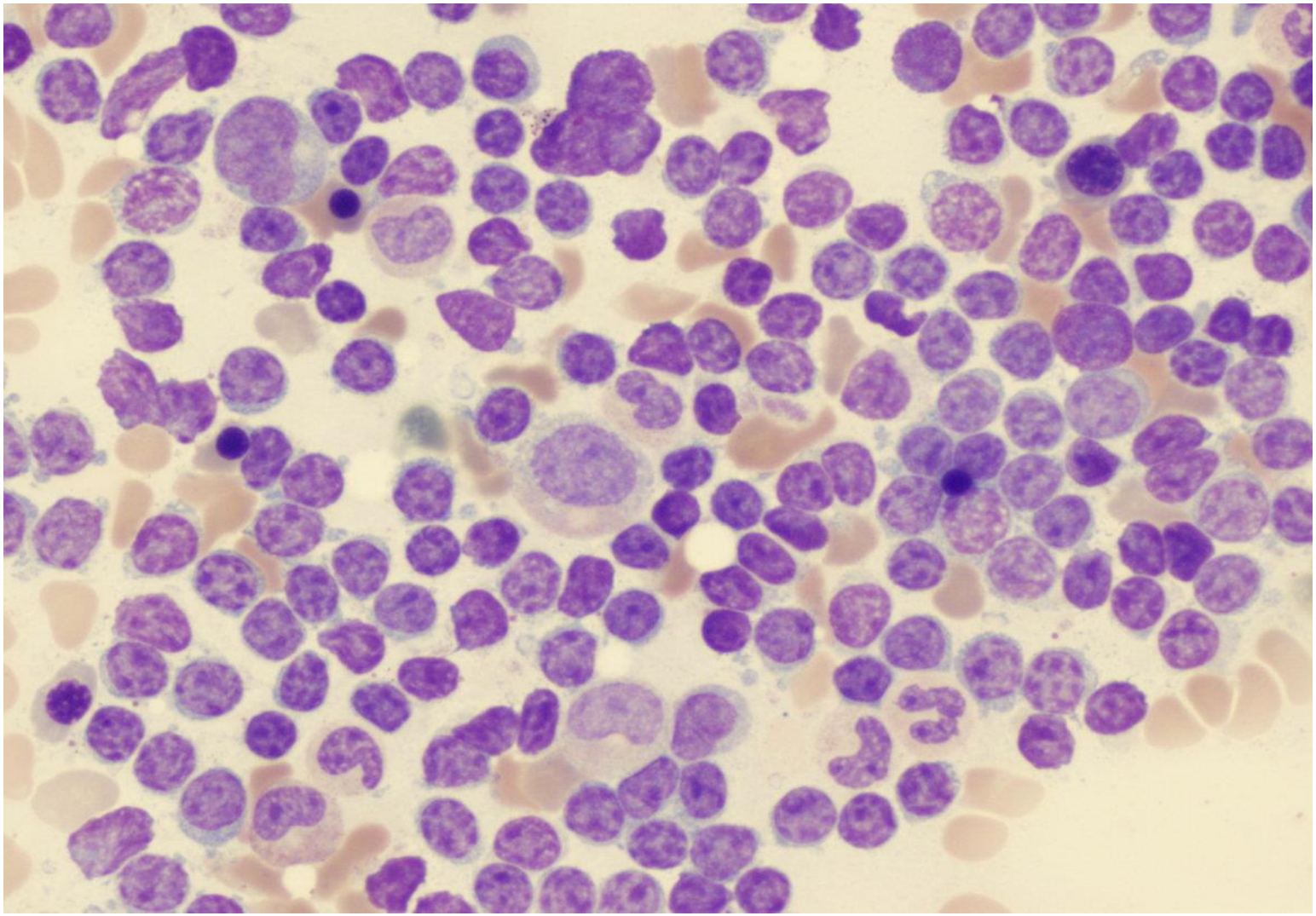
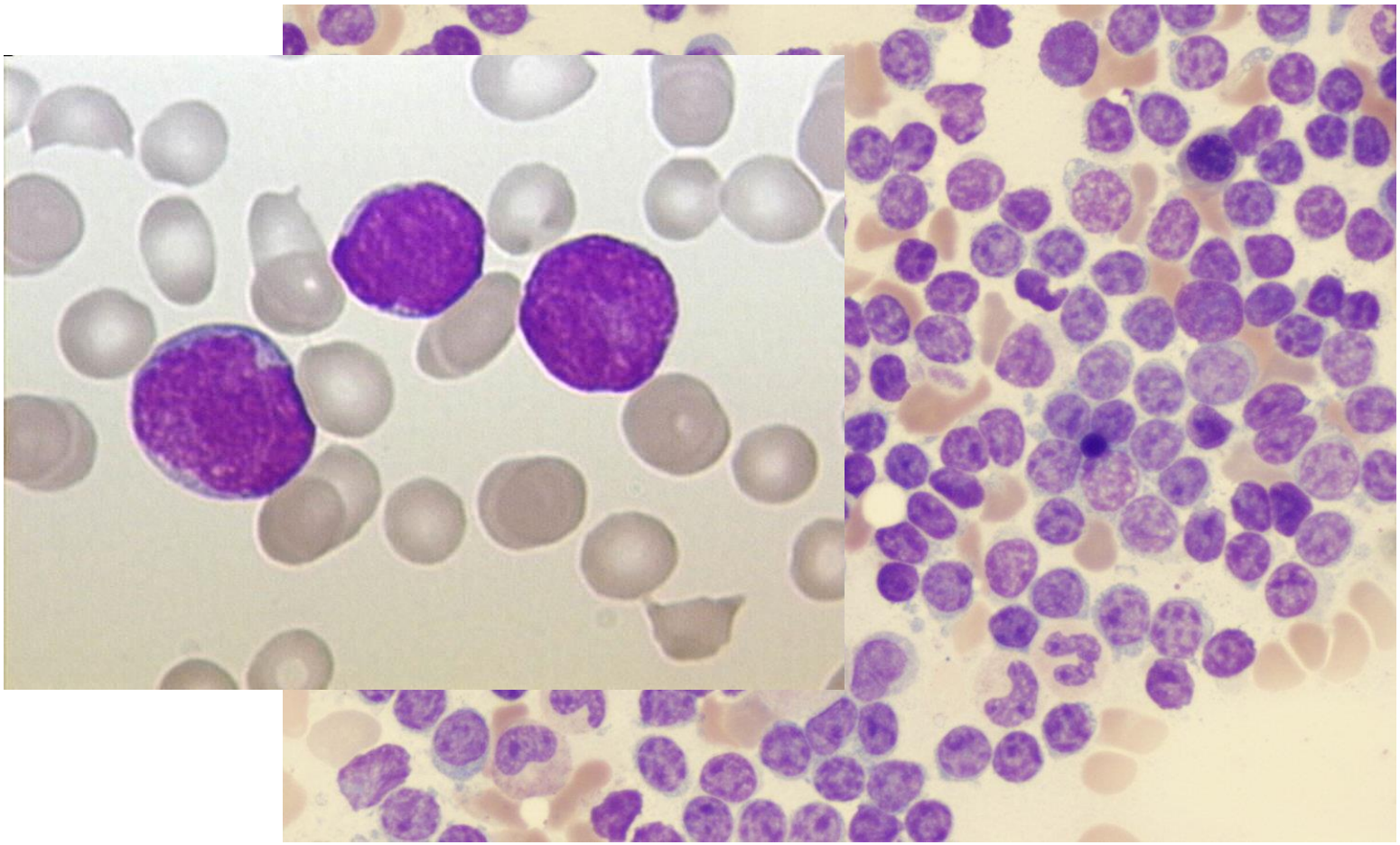
ALL – Blood smear

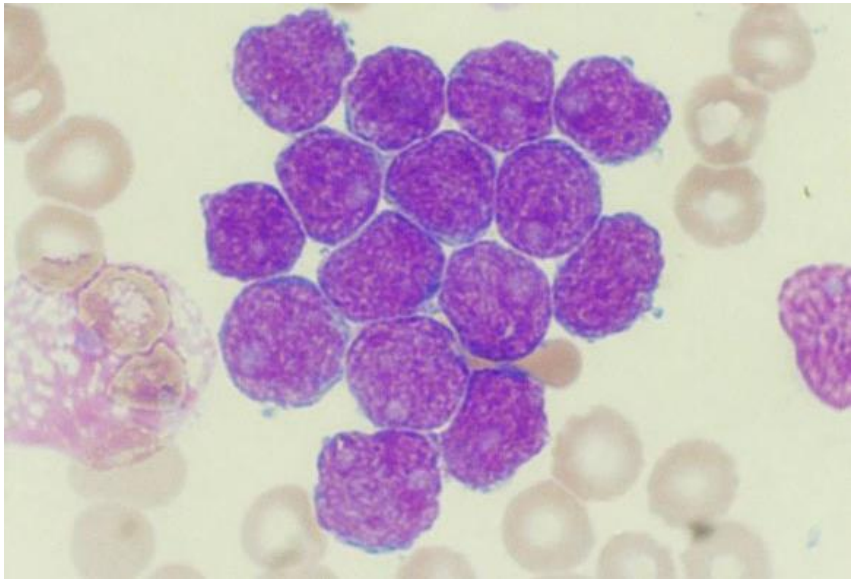


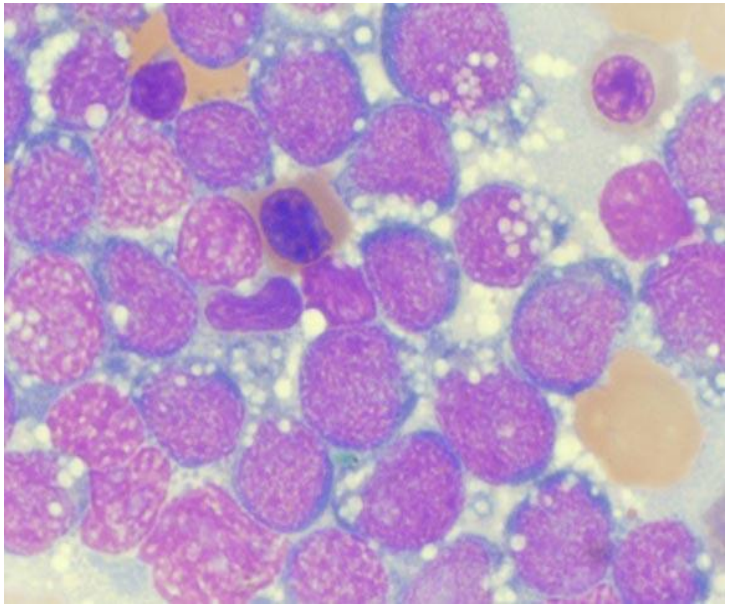
ALL – Blood smear

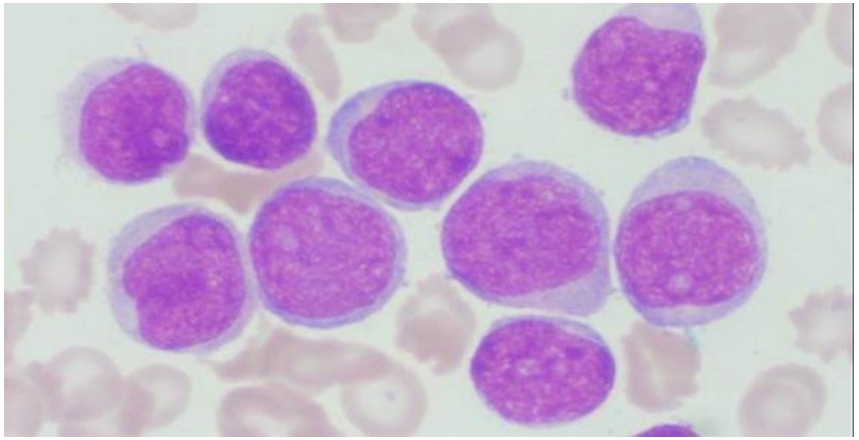


ALL – FAB

L-1	85%
L-2	14%
L-3	1%







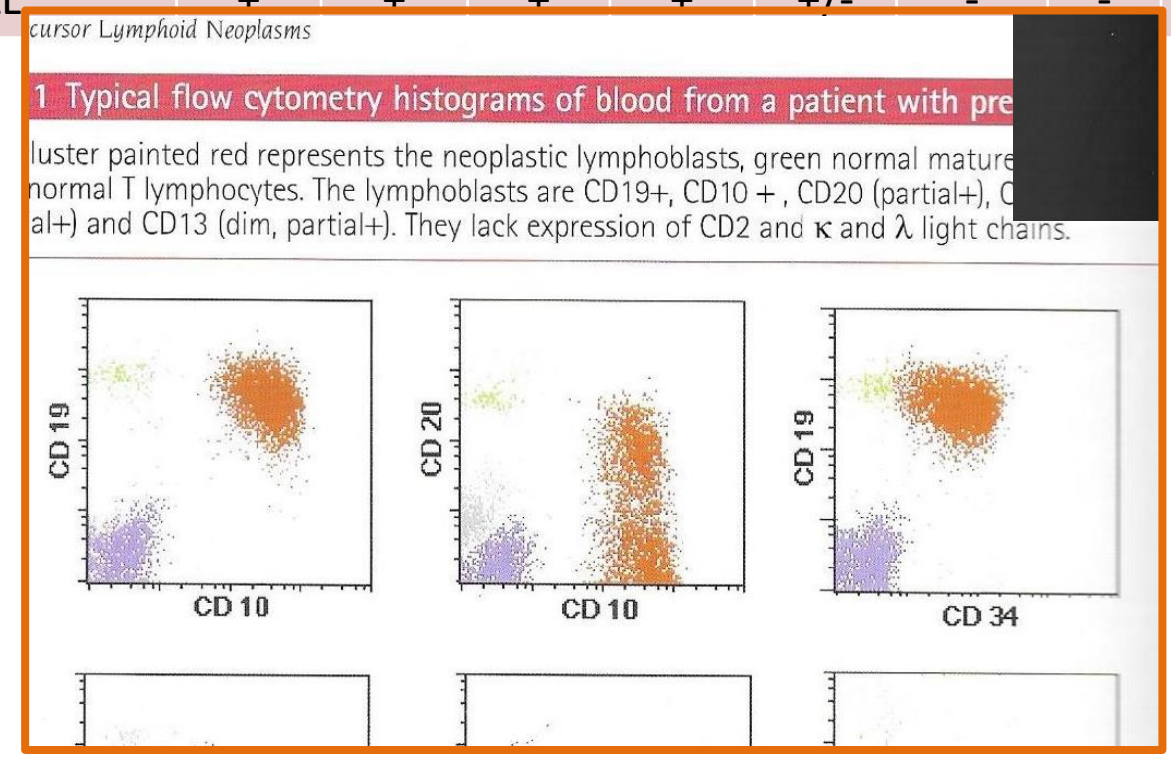
ALL - Immunophenotyping

B cell	CD19	CD20	CD22	CD24	CD10	CD34	Tdt	HLA-DR	SIg
Pre precursor B-ALL	+	-/+	-	+	+/-	+	+	+	-
Precursor B-ALL	+	+/-	-/+	+	+	+	+/-	+	-
Mature B-ALL	+	+	+	+	+/-	-	-	+	+

T cell	CD1	CD2	CD3	CD4	CD5	CD7	CD8	CD10	CD34	Tdt	HLA-DR
T-ALL	+	+	+	+	+	+	+	+/-	-/+	+	-/+

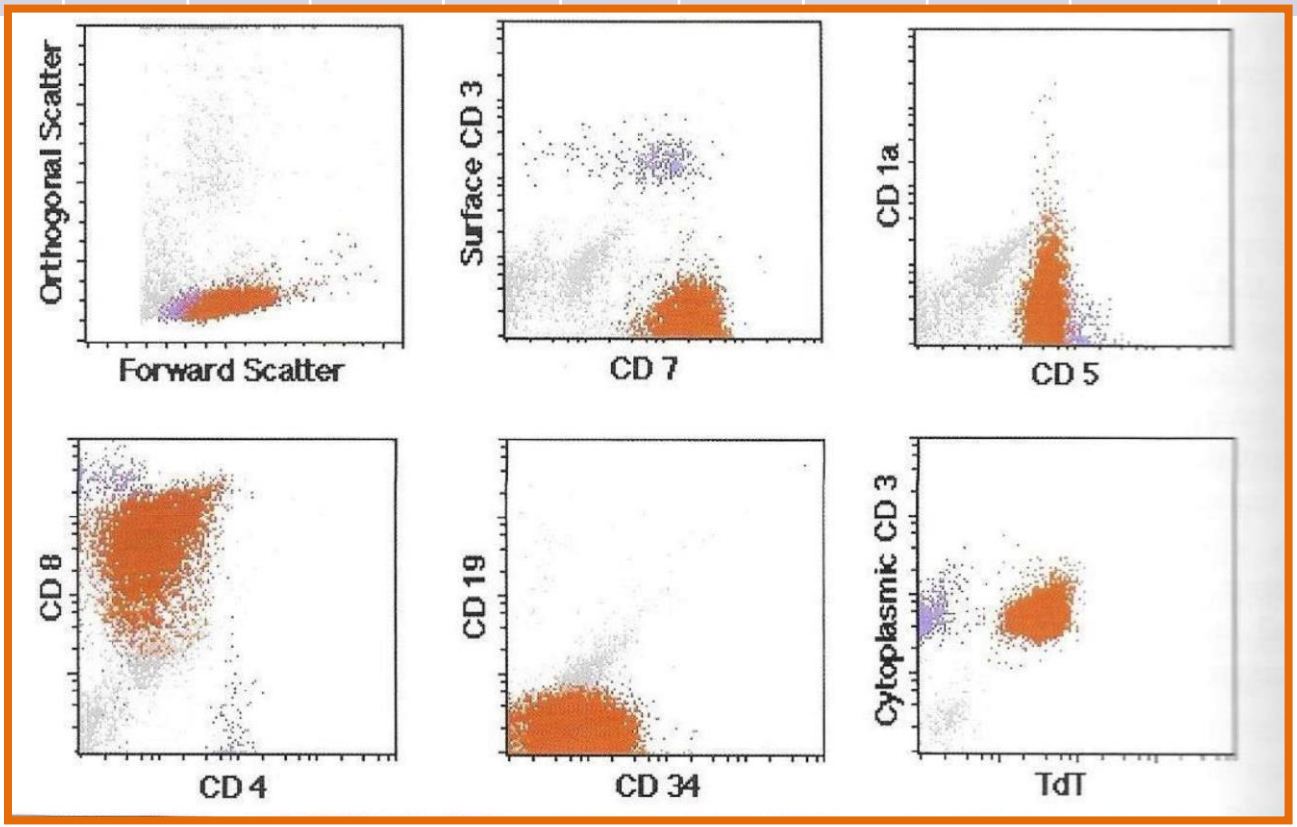
ALL - Immunophenotyping

B cell	CD19	CD20	CD22	CD24	CD10	CD34	Tdt	HLA-DR	SIg
Pre precursor B-ALL	+	-/+	-	+	+/-	+	+	+	-
Precursor B-ALL	+	+/-	-/+	+	+	+	+/-	+	-
Mature B-ALL	+	+	+	+	+/-	-	-	+	+



ALL - Immunophenotyping

T cell	CD1	CD2	CD3	CD 4	CD5	CD7	CD8	CD10	CD34	Tdt	HLA-DR
T-ALL	+	+	+	+	+	+	+	+/-	-/+	+	-/+

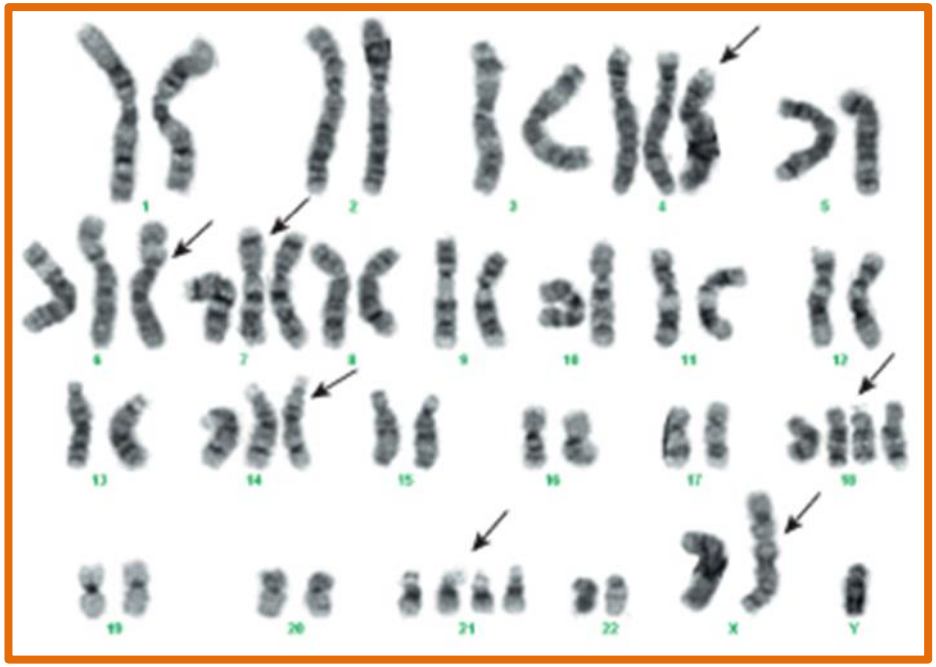


ALL - Cytogenetics

Prognosis	Cytogenetic Findings
Favorable	Hyperdiploidy > 50 chromosomes t(12;21)
Intermediate	Hyperdiploidy 47-50 chromosomes Normal (diploidy) del(6q) t(1;19)
Unfavorable	Hypodiploidy – near haploidy del(17p) t(9;22) t(11;23) t(4;11) 9p abnormalities t(17;19) t(5;14)

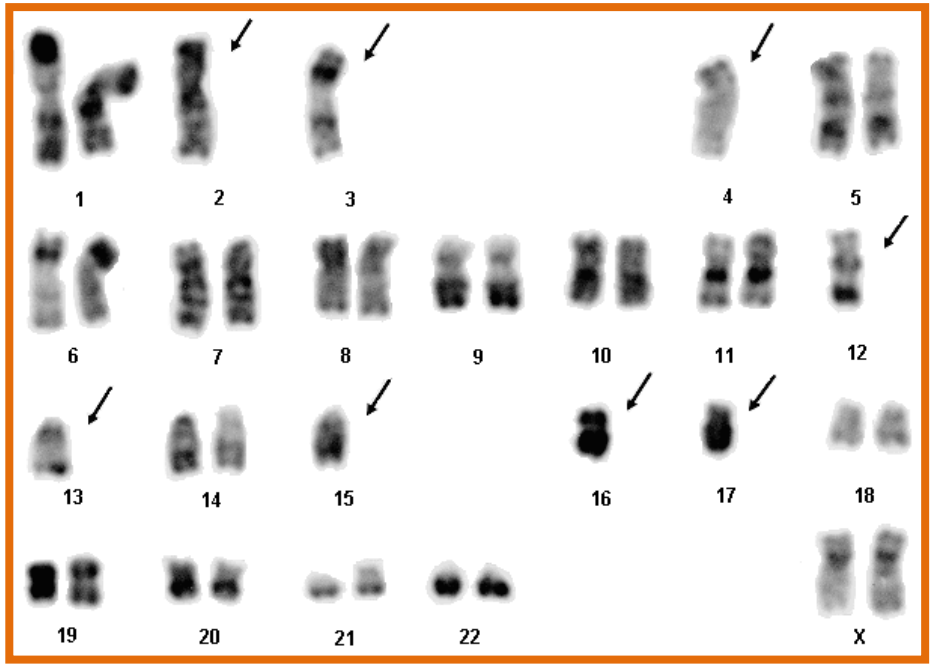
ALL - Cytogenetics

Hyperdiploidy



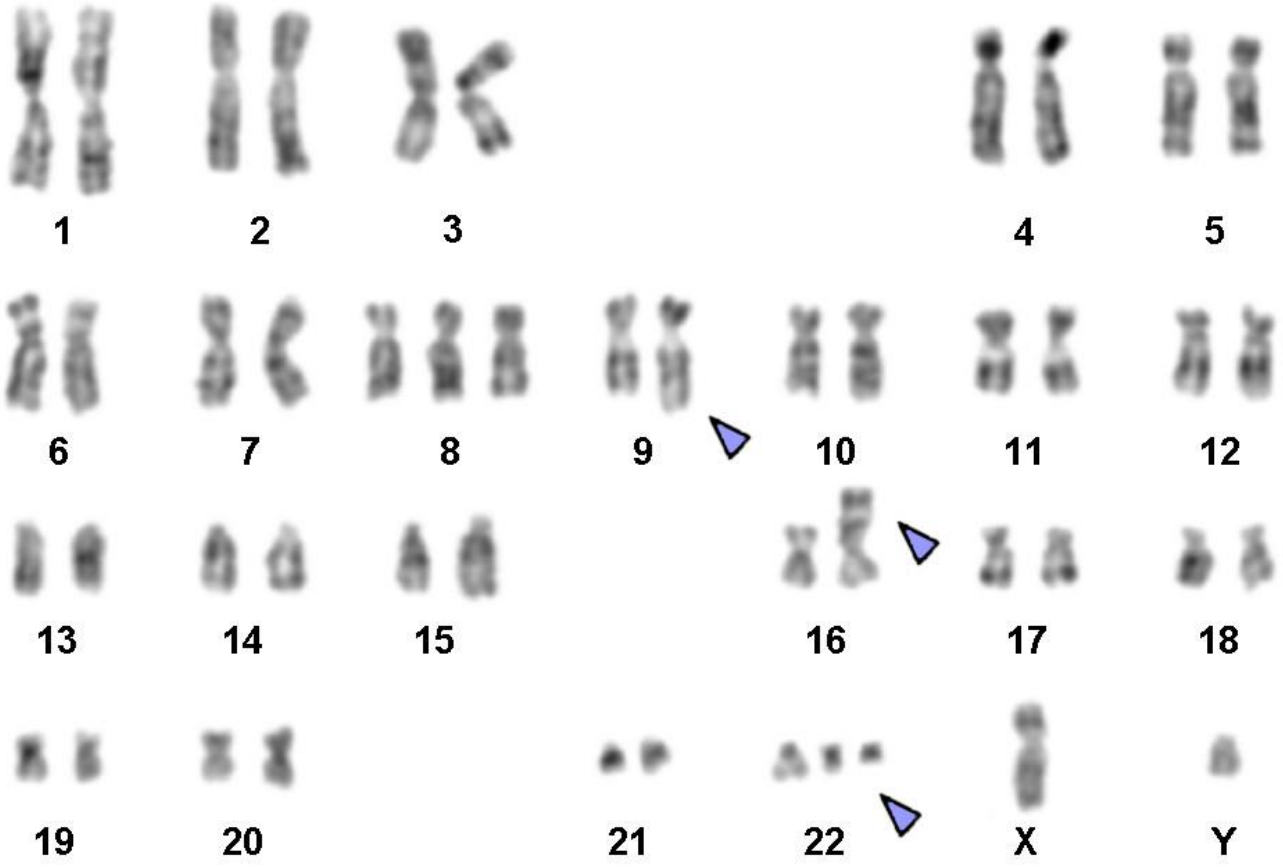
Good prognosis

Hypodiploidy



Poor prognosis

ALL - Cytogenetics



48,XY,+8,t(9;22)(q34;q11),der(16),t(16;17),+der(22)

ALL – WHO Classification

Immunologic subtype	% of cases	FAB subtypes	Cytogenetic abnormalities
Pre B ALL	75	L1, L2	t(9;22), t(4;11), t(1;19)
T cell ALL	20	L1, L2	14q11 or 7q34
Mature B cell ALL (Burkitt leukemia)	5	L3	t(8;14)

ALL – Prognostic factors

Prognostic factors	Favorable	Unfavorable
WBC (cells/mm^3)	<10,000	>50,000
Age (years)	2 – 7	<2 and >10 (esp. infant)
Gender	Female	Male
Response treatment	<4 weeks	>4 weeks
MRD	Negative day15	Positive day33+
Time to relapse after treatment ends	>6 months	<6 months
Surface markers	Precursor B-ALL	T-/B-ALL
Cytogenetics (DI)	Hyperdiploid	Hypodiploid
Structure		11q23/MLL-ALL gene rearrangement
FAB	L1	L2/L3
Mediastinal enlargement	-	(+)
Visceromegaly	+ to ++	+++
LDH	Moderate	High

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Risk Stratification for ALL - ThaiPOG

Standard Risk (SR)	High Risk (HR)	Very High Risk (VHR)
Clinical criteria ■ Pre-B ALL ○ Age 1-9 and ○ WBC <50,000 ■ Down Syndrome Molecular criteria (optional) ■ Day 29 BM MRD <0.01% ■ No unfavorable molecular feature	Clinical criteria ■ T-ALL ■ Pre-B ALL ○ Age 10-13 or ○ WBC ≥50,000 ■ Testicular disease ■ Steroid pretreatment Molecular criteria (optional) ■ Day 29 BM MRD ≥0.01% with favorable cytogenetic: ETV-6/RUNX-1 or double trisomy 4,10	Clinical criteria ■ Pre-B ALL ○ Age ≥14 ■ CNS-3 ■ Induction failure (M2 or M3 at day 29) Molecular criteria (optional) ■ Day 29 BM MRD ≥0.01 with no favorable cytogenetic ■ Unfavorable molecular feature ○ iAMP 21 ○ MLL rearrangement ○ Hypodipliody (<44 chromosome or DNA index <0.81) ○ Ph-chromosome (follow Ph-ALL protocol)

ALL – Treatment

Supportive

- Blood and platelet transfusion
- Antibiotics
- Antipyretics
- Pain meds
- IV fluid
- Prevent tumor lysis

Specific

- Chemotherapy (2.5-3 yrs)
  - Induction
  - Consolidation
  - Maintenance
- CNS-directed therapy
  - Intrathecal therapy
  - Cranial irradiation

ALL Therapy

- Risk-adapted – depending on
  - Clinical manifestations
  - Laboratory analysis: morphology, cytochemistry, immunology, molecular cytogenetics
- Divided into
  - Remission induction
  - Consolidation
  - Maintenance phase (including delayed intensification, interim maintenance, etc)

Induction of Remission

- **Remission** : disappearance of all signs of leukemia on clinical examination and peripheral blood analysis
  - Bone marrow analysis with <5% blasts morphologically
  - Normal hematopoiesis
  - MRD detection – increasingly used
- **Goal** : elimination of leukemia cells by a combination of chemotherapy (3 drugs vs. 4 drugs)
- **Success rate** : >90%
- Regression of organomegaly noted within the first 2 weeks
- **Duration** : 4-5 weeks

Consolidation Treatment

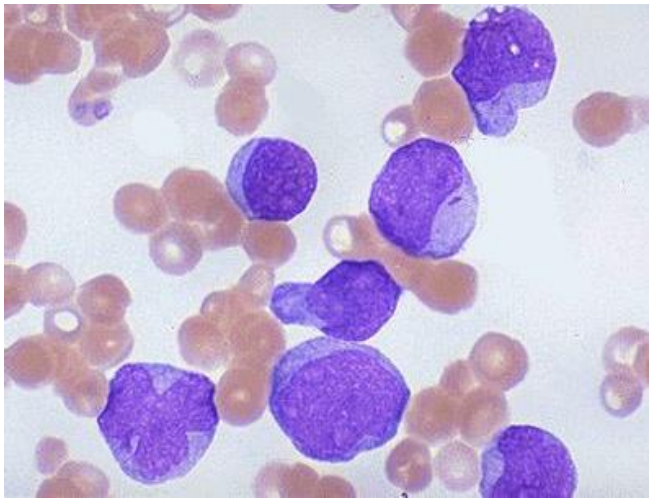
- Without treatment beyond induction, leukemia will reappear within weeks or months
- **Goal :** to administer further intensive chemotherapy to completely eradicate leukemic cells
- **How** : combinations of different chemotherapy to reduce the number of remaining leukemic cells and the development of chemotherapy resistance

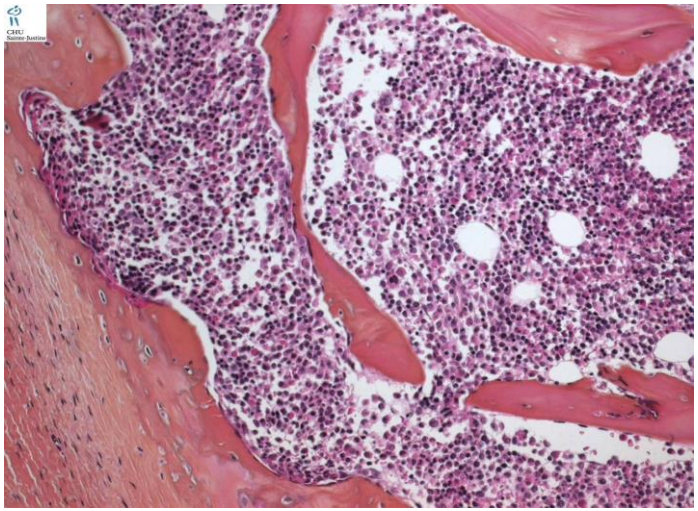
Maintenance Treatment

- **Goal** : prevents recurrence of ALL
- **Duration** : 1.5–2.5 years
- Most of the chemotherapy will be in oral form
- The dosage of chemotherapy will be adapted to the patient's condition and blood cell counts
- Can start being back to a normal lifestyle

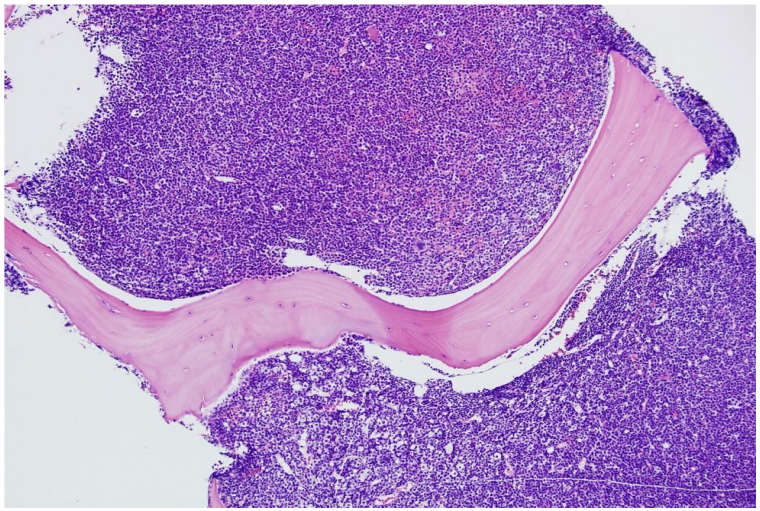
Acute Myeloid Leukemia

- 15-20% of all childhood leukemia
- Frequency slight increase during adolescence
- No gender difference
- Therapy is intense over 6-8 months
- Survival up to 60-70%



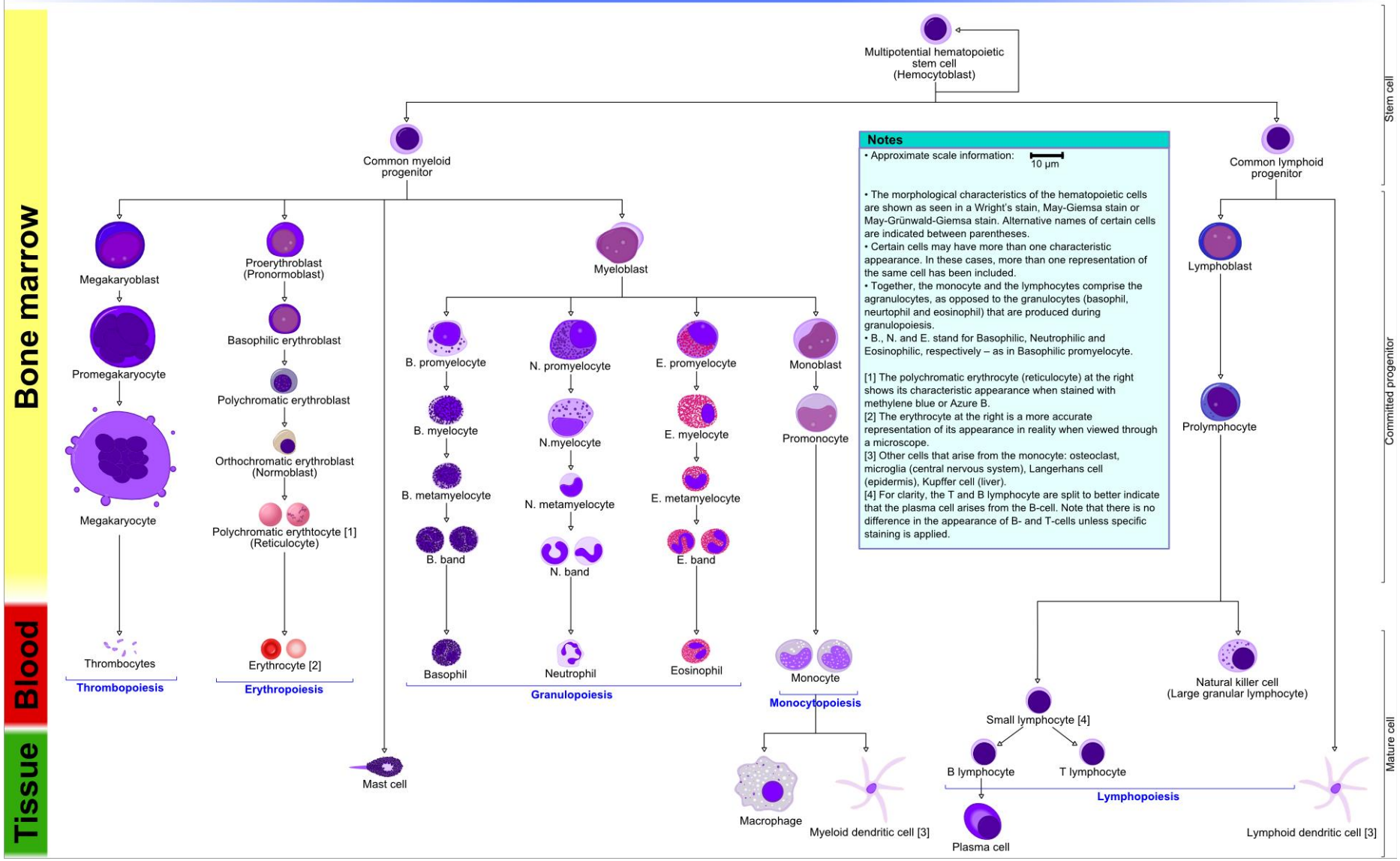


Normal bone marrow



Bone marrow with leukemic infiltrates

Hematopoiesis in humans



AML – Diagnosis

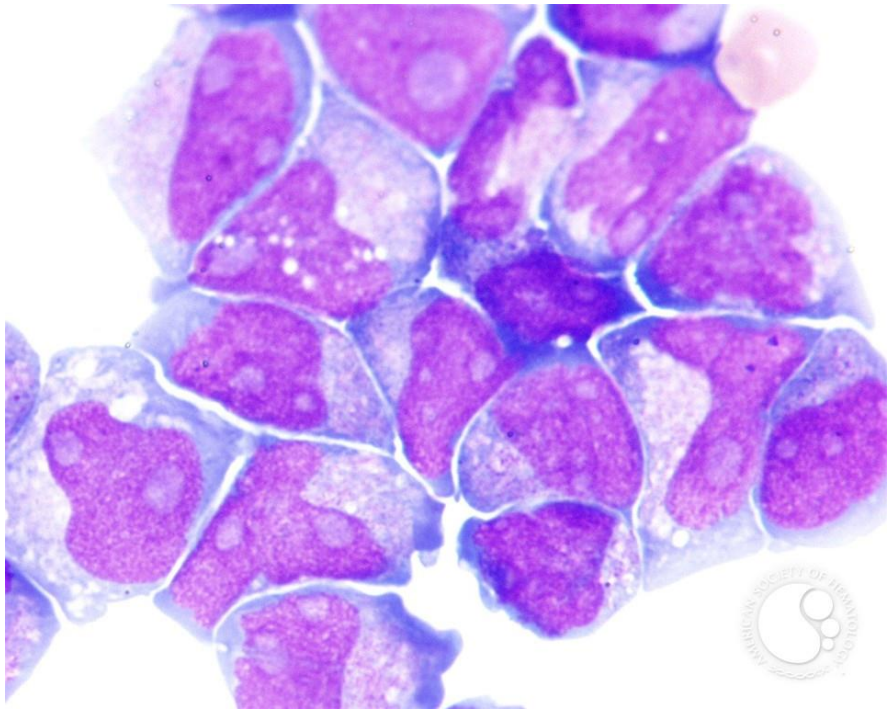
Supportive

- CBC
- PBS
- Type & crossmatch for blood and platelets
- Hemoculture & urine culture
- Tumor lysis labs
- CXR

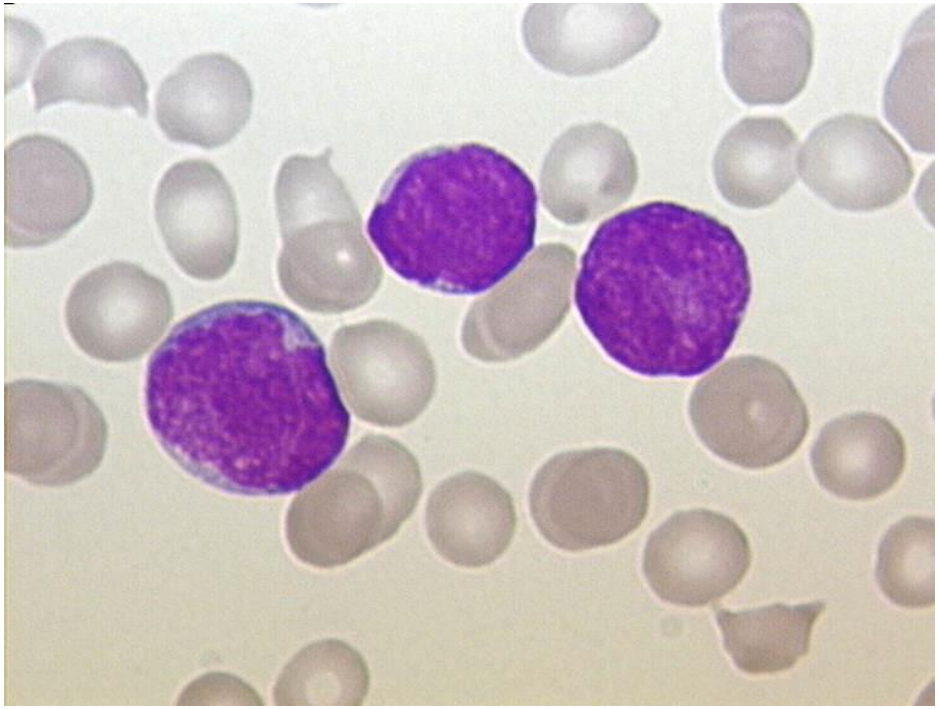
Specific

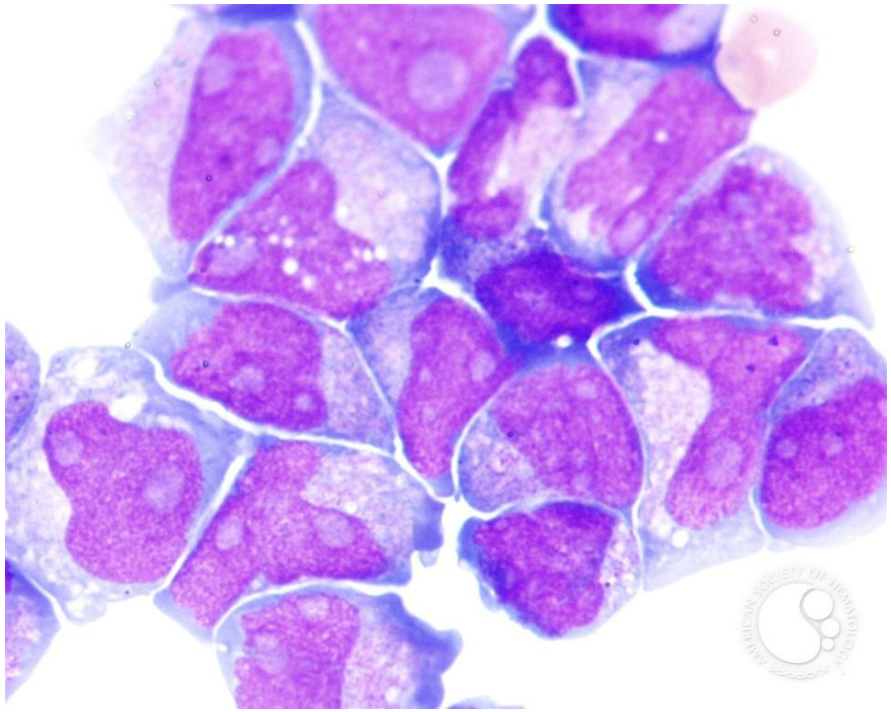
- Bone marrow aspirate and biopsy
  - Morphology
  - Immunophenotyping
  - Cytogenetics
- CNS-directed therapy
  - Intrathecal therapy

Acute Myeloid Leukemia



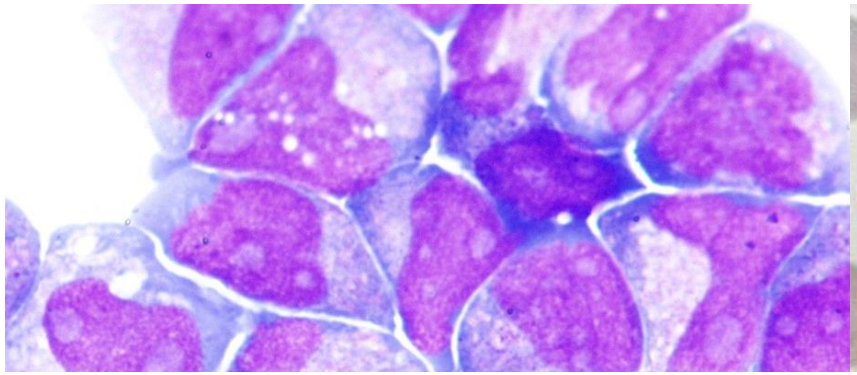
Acute Myeloid Leukemia

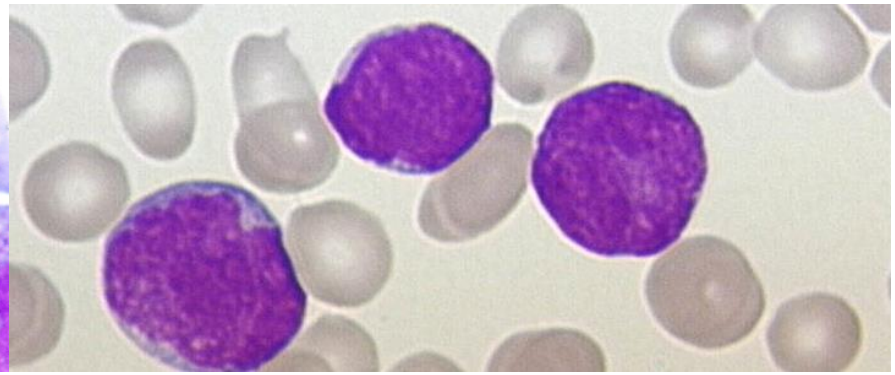




Cytologic Features of Blasts in AML vs. ALL

Feature	AML	ALL
Blast size	Large, often uniform	Variable, small to medium size
Nuclear chromatin	Usually finely dispersed	Coarse to fine
Nucleoli	1-4, often prominent	Absent or 1-2
Cytoplasm	Moderately abundant, granules often present	Usually scant, coarse granules sometimes present (7%)
Auer rods	Present in 60-70% of cases	Not present

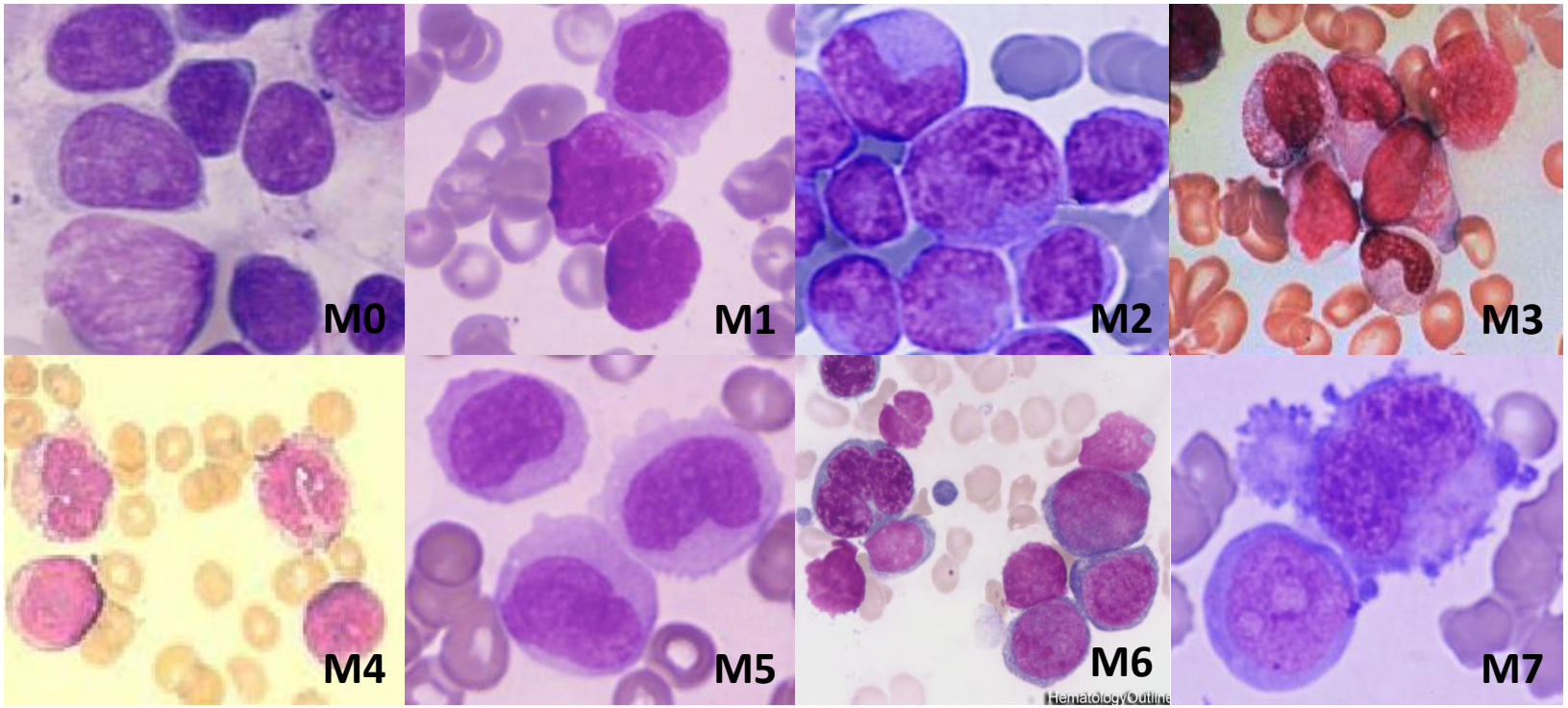




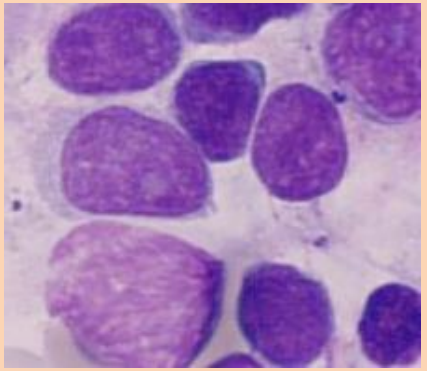
1976: FAB Classification

M0	Minimal myeloid differentiation (MPO-)
M1	Poorly differentiated myeloblasts
M2	Granulocytic differentiation
M3	Acute promyelocytic leukemia
M4	Myelomonocytic leukemia
M5	Monoblastic leukemia
M6	Erythroblastic leukemia
M7	Megakaryoblastic leukemia

AML – FAB Classification

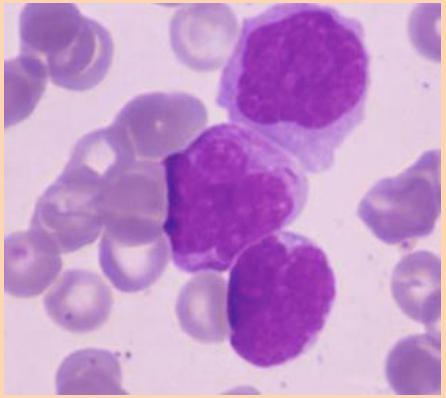


Acute Myeloblastic Leukemia with Minimally Differentiated (M0)



- Common in adult > children
- Accounts for <3% of all AMLs
- Diagnosis :
  - < 3% blasts positive for MPO, PAS and NSE
  - Blasts negative for B and T lymphocyte antigens, platelet glycoprotein and erythroid glycophorin A
  - Myeloid antigens : CD13, CD33 and CD11b positive

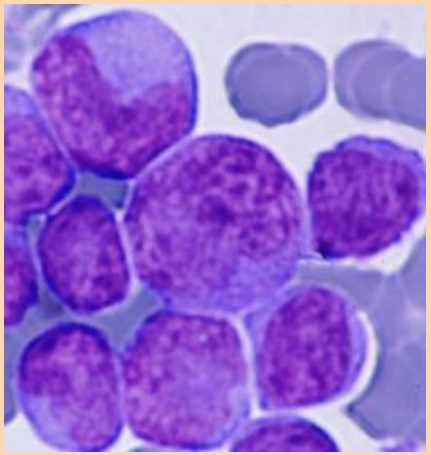
Acute Myeloblastic Leukemia without Maturation (M1)

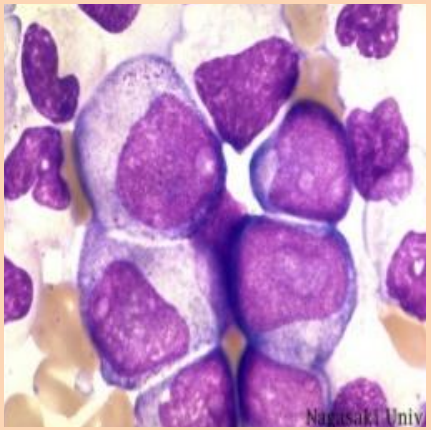


- Common in all age group
- > 90% cells are myeloblasts
- Auer rods are found in the blast of 50%
- If no evidence of granules or Auer rods, the blasts may resemble L2
- MPO or Sudan black stains positive in > 3% of the blasts indicating granulocytes differentiation
- Myeloid antigens : CD13, CD33 and CD11b positive
- Most common cytogenetic abnormalities : t(9; 22) (q34; q11)

Acute Myeloblastic Leukemia with Maturation (M2)

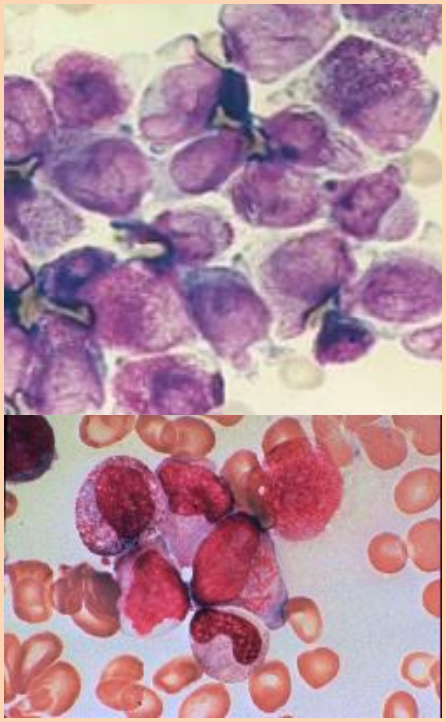
- Accounts for 25% of all AMLs
- 30 – 90% are myeloblasts
- Presence of maturation at or beyond promyelocyte stage (differ from M1)
- Monocytic component < 20% (differ from M4)
- 15% associated with t(8;21)





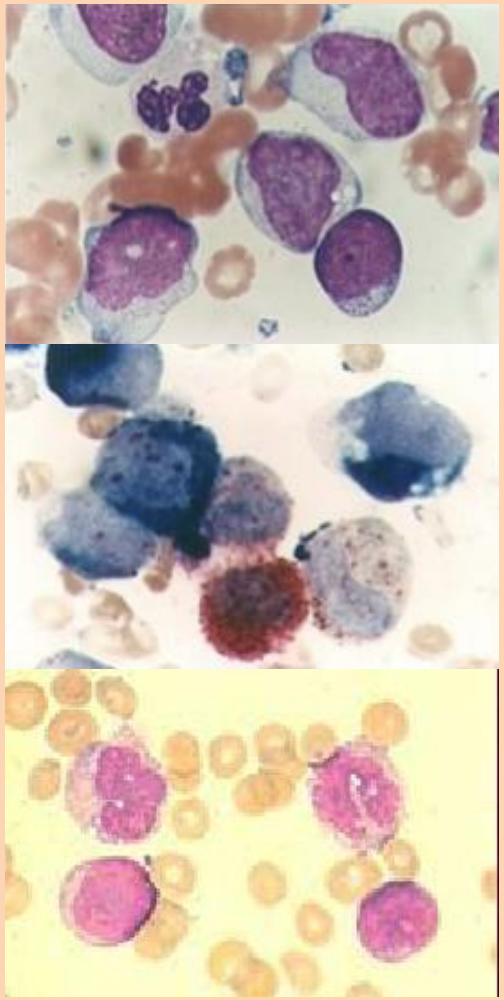
Acute Promyelocytic Leukemia (APL; M3)

- Accounts for 5 – 10% of all AMLs
- Marrow : hypergranular promyelocytes
- Classical – Hypergranular (80% leukopenia)
- Variant – Hypogranular (leukocytosis)
- Granules contain procoagulants (associated with DIC)
- Associated with t(15;17)
- High doses vitamin A (all-trans-retinoic acid; ATRA) can overcome the block in differentiation and induce remission



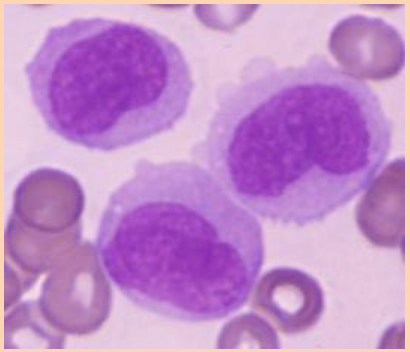
Acute Myelomonocytic Leukemia (AMML; M4)

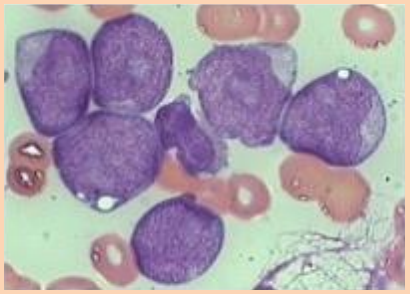
- Accounts for 25 – 36% of all AMLs
- Gingival hyperplasia with bleeding
- Increased incidence of CNS involvement
- Monocytes and promyelocytes 20 – 80% (differ from M1, M2, M3)
- M4 with eosinophilia (M4-Eo) associated with inv 16
- Marrow eosinophil from 6 – 35%

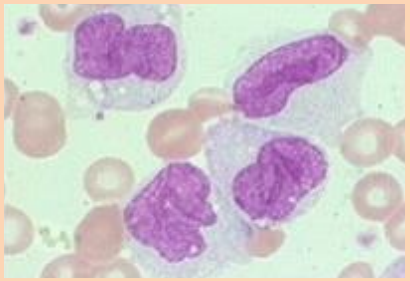


Acute Monoblastic Leukemia (AMOL; M5)

- Accounts for 15% of all AMLs
- Extramedulary infiltration of the lungs, colon, meninges, lymph nodes, bladder and larynx and gingival hyperplasia
- Common findings are weakness, bleeding and a diffuse erythematous skin rash
- > 80% are monocytic cells
- 2 forms : M5a (maturation index <4%)
  M5b (maturation index > 4%)
- Strong association between AML M5/M4 and deletion and translocations involving band 11q23

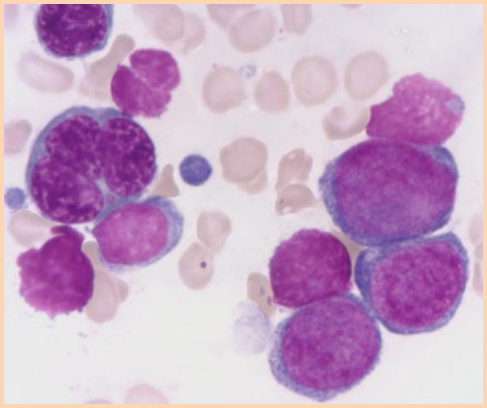
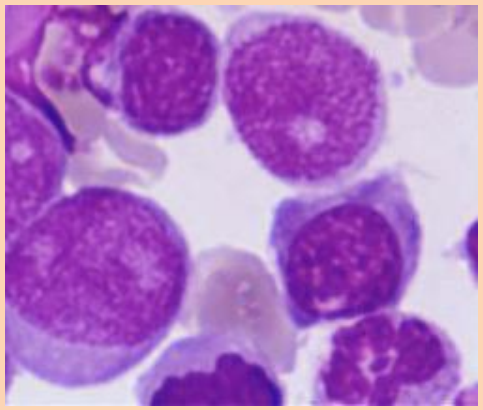
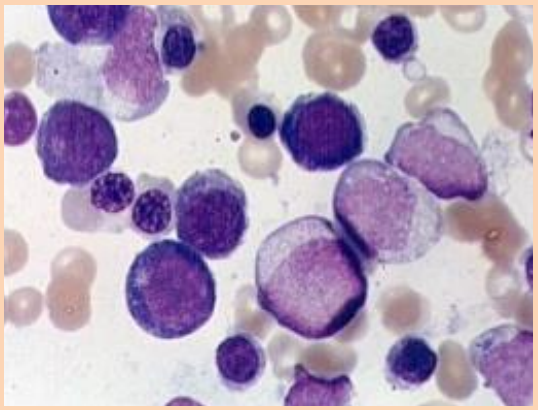






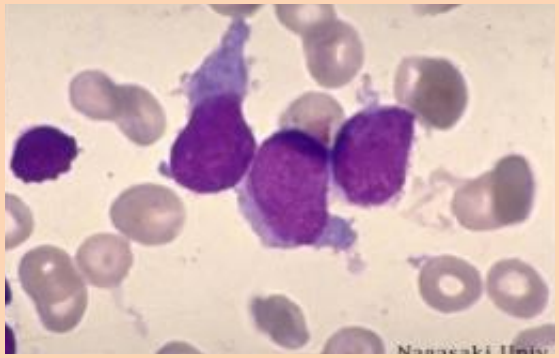
Erythroleukemia (M6)

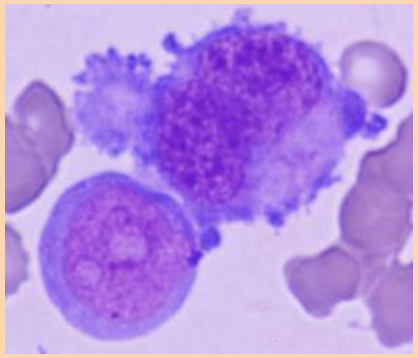
- Rare form of leukemia accounting for < 5% of all AMLs
- Clinical manifestations are similar to other types of AML
- Diagnosis : >50% of all nucleated bone marrow cells are erythroid and > 30% of the remaining nonerythroid cells are myeloblasts (if < 30% then myelodysplasia)
- Cytochemistry of erythroblasts are normally PAS negative but in AML-M6, erythroblasts are PAS positive

Acute Megakaryoblastic Leukemia (AMKL; M7)

- Rare form of leukemia accounting for 5-10% of all AMLs
- Pancytopenia is characteristic at initial diagnosis
- Associated with fibrosis & bone marrow dry tap is common
- Bone marrow biopsy show increased fibroblasts with > 30% blast cells
- Blast cells show cytoplasmic protrusion or budding
- Monoclonal antibodies reacts with platelet glycoprotein Ib, IIb/IIIa and IIIb & immunologic study shows CD41, CD42 and CD61 positive
- Associated with t(1;22)(p13;q13) in young children < 18 months who do not have Down's syndrome





Histochemical classification of AML

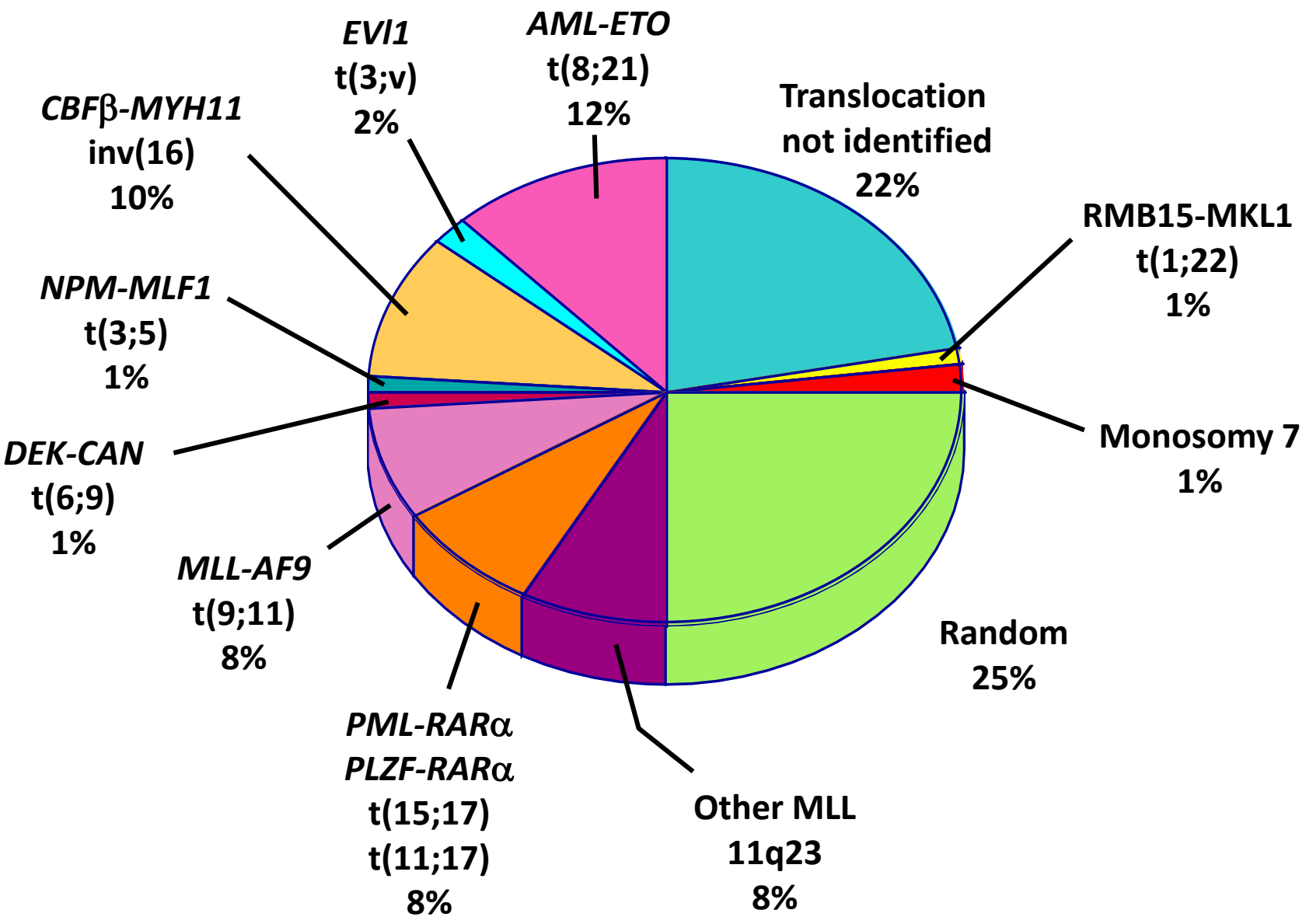
Histochemical characteristics							Frequency (%)
M0	-	SB	-	-	-	-	<3
M1	MPO	SB	-	-	-	-	20
M2	MPO	SB	-	-	-	-	25
M3	MPO	SB	-	-	(NSE)	-	5-10
M4	MPO	-	-	NASD	**NSE**	-	25-36
M5	MPO	-	-	NASD	-	-	15
M6	-	-	PAS	-	-	**Glycophorin A**	<5
M7	-	-	-	NASD	NSE	-	5-10

Pediatric Oncology : A Comprehensive Guide, 2nd Edition (2011)

Immunophenotyping of AML

	Cluster determination															
M0	-	13	-	15	33	34	(36)	31/61	42	65	117	HLA-DR	19	2	(4)	(7)	
M1	-	13	-	15	33	34	-	-	-	65	117	HLA-DR	-	2	-	(7)	(56)
M2	-	13	-	15	33	34	-	-	-	65	117	HLA-DR	-	2	-	7	(56)
M3	**11b**	13	-	15	33	34	-	-	-	65	117	(HLA-DR)	-	2	-	7	(56)
M4	11b	13	14	15	33	34	36	-	-	65	117	HLA-DR	-	2	4	7	56
M5	11b	13	14	15	33	34	36	-	-	65	(117)	**HLA-DR**	-	2	4	7	56
M6	-	13	-	-	33	-	-	-	-	65	(117)	-	-	-	-	7	-
M7	-	13	-	-	-	34	36	**41/61**	42	65	117	**HLA-DR**	-	2	4	7	56

Heterogeneity within FAB groups



WHO 2016

- AML with recurrent genetic abnormalities
  - t(8;21)(q22;q22); *RUNX1-RUNX1T1*
  - inv(16)(p13.1q22); *CBFB-MYH11*
  - t(15;17)(q22;q12); *PML-RARA*
  - t(9;11)(p22;q23); *MLLT3-MLL*
  - t(6;9)(p23;q34); *DEK-NUP214*
  - inv(3)(q21q26.2); *RPN1-EVI1*
  - t(1;22)(p13;q13); *RBM15-MKL1*
  - *Provisional entity: AML with BCR-ABL1*
  - *Mutated NPM1*
  - *Biallelic mutated CEBPA*
  - *Provisional entity: AML with mutated RUNX1*
- AML with myelodysplasia-related changes
- Therapy-related myeloid neoplasms

Arber et al., Blood, 2016

WHO 2016 (cont)

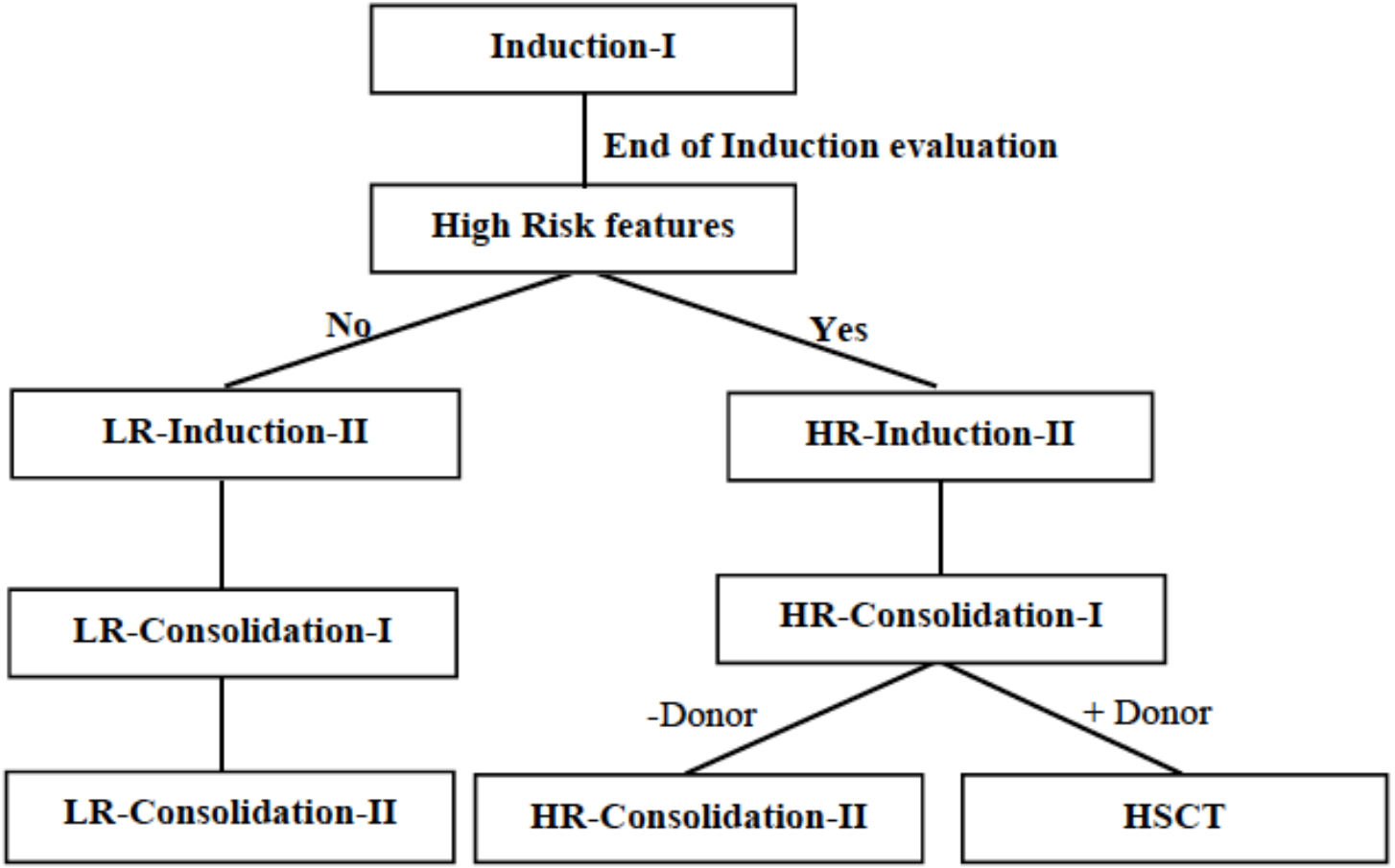
- Acute myeloid leukemia, not otherwise specified
  - AML with minimal differentiation (M0)
  - AML without maturation (M1)
  - AML with maturation (M2)
  - Acute myelomonocytic leukemia (M4)
  - Acute monoblastic/monocytic leukemia (M5)
  - Acute erythroid leukemia (M6)
  - Acute megakaryoblastic leukemia (M7)
  - Acute basophilic leukemia
  - Acute panmyelosis with myelofibrosis
- Myeloid Sarcoma
- Myeloid proliferations related to Down syndrome
  - Transient abnormal myelopoiesis (TAM)
  - Myeloid leukemia associated with Down syndrome

Arber et al., Blood, 2016

Risk Stratification for AML - ThaiPOG

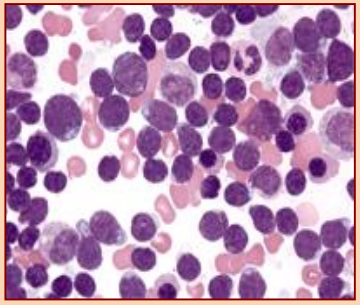
Low Risk (LR)	High Risk (HR)
■ Presence of low risk molecular marker: Inv 16 or t(8,21) regardless of monosomy 5, monosomy 7, -5q, MLL-gene rearrangement or MRD status at the end of induction-I ■ Normal cytogenetic with MRD < 0.1% at the end of induction-I ■ AML patient who has no molecular marker and cytogenetic information available	■ FLT3/ITD positive with high allelic ratio >0.4 regardless of low risk feature ■ Presence of monosomy 5, monosomy 7, -5q or MLL rearrangement without low risk molecular features ■ Normal cytogenetic with MRD ≥ 0.1% at the end of induction-I ■ Induction failure (M2, M3 at the end of induction-I)

Treatment Schema for AML Protocol

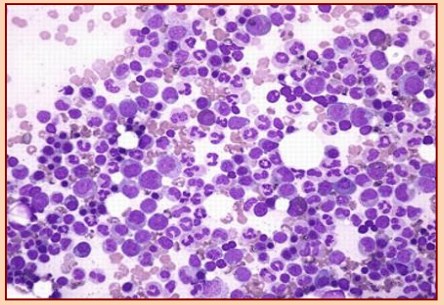


AML - Principles of Therapy

- 4-5 courses of intensive therapy
  - No role for maintenance therapy
  - Role of allogeneic SCT controversial
- Standard agents
  - Cytarabine and anthracyclines most active
  - Other agents include etoposide, cladribine, thioguanine, topotecan



Leukemia



Recovering bone marrow

Summary

- Each type of leukemias has their unique characteristics
- Acute leukemia associates with decreased numbers of other lineages
- Peripheral blood smear and further bone marrow examination are essential for definite diagnosis
- Immunophenotyping, cytogenetic and molecular studies are used to stratify patients in different risk group and appropriate treatment protocol

Thank you