

**SUMMARY OF WHO CLASSIFICATION OF
TUMORS OF HEMATOPOIETIC AND LYMPHOID TISSUES**

**CHRONIC MYELOPROLIFERATIVE
DISEASES**

Chronic myelogenous leukemia
Chronic neutrophilic leukemia
Chronic eosinophilic leukemia/
hypereosinophilic syndrome
Polycythemia vera
Chronic idiopathic myelofibrosis
Essential thrombocythemia
Chronic myeloproliferative disease,
unclassifiable

**MYELODYSPLASTIC/
MYELOPROLIFERATIVE DISEASES**

Chronic myelomonocytic leukemia
Atypical chronic myeloid leukemia
Juvenile myelomonocytic leukemia
Myelodysplastic/ myeloproliferative diseases,
unclassified

MYELODYSPLASTIC SYNDROMES

Refractory anemia
Refractory anemia with ringed sideroblasts
Refractory cytopenia with multilineage dysplasia
Refractory anemia with excess blasts
Myelodysplastic syndrome associated with
isolated del(5q) chromosome abnormality
Myelodysplastic syndrome, unclassifiable

ACUTE MYELOID LEUKEMIAS

**Acute myeloid leukemias with recurrent
cytogenetic abnormalities**

AML with t(8;21) (q22; q22), (AML1/ETO)
AML with inv(16)(p13q22) or t(6;16)(p13;q22)
(CBF β /MYH11)
Acute promyelocytic leukemia
(AML with t(15;17) (q22;q12), (PML/RAR α)
and variants)
AML with 11q23 (MLL) abnormalities

**Acute myeloid leukemia with
multilineage dysplasia**

with prior myelodysplastic syndrome
without prior myelodysplastic syndrome

**Acute myeloid leukemia and
myelodysplastic syndrome,
therapy related**

Alkylating agent related
Topoisomerase II inhibitor-related

**Acute myeloid leukemia not otherwise
categorized**

Acute myeloid leukemia,
minimally differentiated
Acute myeloid leukemia without maturation
Acute myeloid leukemia with maturation
Acute myelomonocytic leukemia
Acute monoblastic & monocytic leukemia
Acute erythroid leukemia
Acute megakaryoblastic leukemia
Acute basophilic leukemia
Acute panmyelosis with myelofibrosis
Myeloid sarcoma

Acute leukemia of ambiguous lineage

B-CELL NEOPLASMS

Precursor B-cell neoplasm

Precursor B lymphoblastic leukemia/
lymphoma

Mature B-cell neoplasms

Chronic lymphocytic leukemia/
small lymphocytic lymphoma
B-cell prolymphocytic leukemia
Lymphoplasmacytic lymphoma
Splenic marginal zone lymphoma
Hairy cell leukemia
Plasma cell myeloma

Solitary plasmacytoma of bone
Extraosseous plasmacytoma
Extranodal marginal zone B-cell
lymphoma of mucosa-associated
lymphoid tissue (MALT lymphoma)
Nodal marginal zone B-cell lymphoma
Follicular lymphoma
Mantle cell lymphoma
Diffuse large B-cell lymphoma
Mediastinal (thymic) large B-cell lymphoma
Intravascular large B-cell lymphoma
Primary effusion lymphoma
Burkitt lymphoma/leukemia

**B-cell proliferations of uncertain
malignant potential**

Lymphomatoid granulomatosis
Post-transplant lymphoproliferative disorder,
polymorphic

T-CELL AND NK-CELL NEOPLASMS

Precursor T-cell neoplasms

Precursor T lymphoblastic leukemia/lymphoma
Blastic NK cell lymphoma

Mature T-cell and NK-cell neoplasms

T-cell prolymphocytic leukemia
T-cell large granular lymphocytic leukemia
Aggressive NK cell leukemia
Adult T-cell leukemia/lymphoma
Extranodal NK/T cell lymphoma, nasal type
Enteropathy-type T-cell lymphoma
Hepatosplenic T-cell lymphoma
Subcutaneous panniculitis-like
T-cell lymphoma
Mycosis fungoides
Sezary syndrome
Primary cutaneous anaplastic
large cell lymphoma
Peripheral T-cell lymphoma, unspecified
Angioimmunoblastic T-cell lymphoma
Anaplastic large cell lymphoma

**T-cell proliferation of uncertain
malignant potential**

Lymphomatoid papulosis

HODGKIN LYMPHOMA

Nodular lymphocyte predominant
Hodgkin lymphoma
Classical Hodgkin lymphoma
Nodular sclerosis classical Hodgkin lymphoma
Lymphocyte-rich classical Hodgkin lymphoma
Mixed cellularity classical Hodgkin lymphoma
Lymphocyte-depleted classical
Hodgkin lymphoma

**HISTIOCYTIC AND DENDRITIC-CELL
NEOPLASMS**

Macrophage/ histiocytic neoplasm

Histiocytic sarcoma

Dendritic cell neoplasms

Langerhans cell histiocytosis
Langerhans cell sarcoma
Interdigitating dendritic cell sarcoma/tumor
Follicular dendritic cell sarcoma/ tumor
Dendritic cell sarcoma, not otherwise
specified

MASTOCYTOSIS

Cutaneous mastocytosis
Indolent systemic mastocytosis
Systemic mastocytosis with associated
clonal, hematological
non-mast cell lineage disease
Aggressive systemic mastocytosis
Mast cell leukemia
Mast cell sarcoma
Extracutaneous mastocytoma

CYTOCHEMICAL STAINS USEFUL IN DELINEATING IMMATURE MYELOID ELEMENTS

Cytochemical Stain	Utility/Features
Myeloperoxidase	<p>Most specific cytochemical stain for granulocyte precursors</p> <p>May be weakly positive in monocyte precursors but generally negative</p> <p>Mature dyspoietic granulocytes may lack myeloperoxidase in some cases of acute myelogenous leukemia</p>
Sudan black B	<p>Stains lipid membrane of primary granules and peroxisomes in granulocyte precursors</p> <p>May be weakly positive in monocyte precursors but generally negative</p> <p>Rare cases of ALL are weakly positive</p>
Chloracetate esterase	<p>Stains primary granules of granulocyte precursors</p> <p>Reports of positivity in ALL (notably granular ALL) and acute monocytic leukemias</p> <p>Abnormally expressed in eosinophils in patients with acute myelomonocytic leukemia with eosinophilia</p>
Nonspecific esterase α -Naphthyl acetate	<p>Variably strong staining of monocytic cells (diffuse) and megakaryoblasts (punctate)</p> <p>Sodium fluoride inhibition of monocytic esterase</p> <p>Weak reaction may be found in granulocytic elements</p> <p>Reports of moderate positivity in some acute promyelocytic leukemia cases that are purely granulocytic by all other features</p>
α -Naphthyl butyrate	<p>Stains granules of monocytic cells (reaction may be weak to strong)</p> <p>May be weakly positive in granulocytic precursors but generally negative</p>
Periodic acid-Schiff*	<p>Large granules and globules of periodic acid-Schiff positive material* (glycogen or glycoprotein) in leukemic erythroblasts, while more mature neoplastic erythroid elements exhibit diffuse cytoplasmic positivity</p> <p>Diffuse or globular positivity also found in megakaryoblasts</p> <p>Weak diffuse positivity common in granulocytic and monocytic precursors</p>

Toluidine blue

Positive in both mast cells and basophils
Mast cells also chloracetate esterase positive

Ultrastructural myelo-
peroxidase

By electron microscopy, myeloperoxidase can be identified in peroxisomes and early granulocytic precursors (not detectable by light microscopy)

Platelet peroxidase in endoplasmic reticulum and perinuclear cisternae characteristic of megakaryoblasts (not detectable by light microscopy)

*Global positivity also characteristically present in lymphoblasts.

CLUSTER DESIGNATIONS (CD's) OF ANTIGENS USED IN THE DIAGNOSIS OF BONE MARROW DISORDERS

CD Number	Cellular Distribution/Utility in Diagnosis
CD1	Immature T cells, Langerhans' cells, occasional myeloid leukemias
CD2	T cells, natural killer cells, and myeloid leukemias
CD3	T cells
CD4	Helper T cells, monocytes/macrophages
CD5	T cells, B-cell subsets, B-cell chronic lymphocytic leukemia
CD7	T cells, occasional myeloid leukemias, natural killer cells
CD8	Suppressor/cytotoxic T cells, natural killer cells
CD10	Mature and immature B lymphocytes, some myelomas, some T-cell acute lymphoblastic leukemias
CD11a	Adhesion molecule on many cell types
CD11b	Granulocytes, monocytes/macrophages, and natural killer cells
CD11c	Adhesion molecule on monocytes, granulocytes, natural killer cells, and hairy cells
CD13	Myeloid elements
CD14	Myeloid/monocytic elements
CD15	Granulocytes, Reed-Sternberg cells
CD16	Fc γ receptor on natural killer cells, granulocytes, monocytes
CD19	B cells
CD20	B cells
CD21	Mature B cells (Epstein-Barr virus receptor)
CD22	B cells
CD23	B-cell subsets, B-cell chronic lymphoproliferative disorders
CD25	Interleukin-2 receptor on activated B cells, T cells, macrophages, and hairy cells
CD30	Reed-Sternberg cells, anaplastic large cell lymphoma cells, activated B and T cells, nonhematopoietic neoplasms
CD33	Myeloid elements
CD34	Hematopoietic progenitor cells, endothelial cells
CD38	Plasma cells, activated cells, progenitor cells, natural killer cells
CD41	Megakaryoblasts (glycoprotein IIb/IIIa)
CD43	T cells, myeloid cells, and some low-grade B-cell lymphoproliferative disorders
CD45	Pan leukocyte
CD56	Adhesion molecule, natural killer cells, some myelomas, activated T cells
CD57	Natural killer cells, T-cell subset
CD79a	B cells (mb-1 gene product, Ig α ; component of B cell antigen receptor "BCR" complex)
CD117	Hematopoietic Progenitor cells (C-Kit: Stem Cell Factor Receptor "SCFR")

B-Cell Differentiation

T-Cell Differentiation

**ONCOLOGY PROBES AVAILABLE IN
CYTOGENETICS AND MOLECULAR GENETICS
FOR FISH ANALYSIS AT KFSH&RC**

Type of Probe

Translocation Probes:

MYC/IGH for t(8;14)(q24;q32)	ALL, Burkitt's
AML1/ETO for t(8;21)(q22;q22)	AML M2
BCR/ABL t(9;22)(q34;q11.2)	CML, some ALL
CCND1/IGH t(11;14)(q13;q32)	mantle cell lymphoma
TEL/AML1 t(12;21)(p13;q22)	ALL <12y
IGH/BCL2 t(14;18)(q32;q21)	non-Burkitt's/Hodgkin's
PML/RARA t(15;17)(q22;q21.1)	AML M3

Amplification Probes

nmyc (MYCN) on chromosome 2p23-24	neuroblastoma
cmyc (MYC) on chromosome 8q24.2-q24.3	ovarian, cervical, and prostate cancer

Other Oncogene Probes:

MLL on chromosome 11q23	AML <12y, ALL <12y
RB-1 (retinoblastoma) on 13q14	Retinoblastoma
CBFB on 16q22 for inv(16)	AML M4EO

CLL Probes:

12 centromere
RB1 (Retinoblastoma)

Multiple Myeloma Probes:

6 centromere
17 centromere
RB1 (Retinoblastoma)

Post BMT Probes :

X centromere/Y q12

Routine Samples for Hematological Malignancies

1. Bone Marrow is the standard choice for bone marrow cytogenetics analysis (1 cc of bone marrow in a bone marrow transfer media is required).
2. Leukemic blood samples may be sent in cases where BM is difficult to obtain (5-10 cc blood in a sodium heparin tube)
3. For CLL, the choice of sample is peripheral blood, however BM may also be sent.
4. For Lymphoma, lymph nodes biopsy samples are required.

FRENCH-AMERICAN-BRITISH CLASSIFICATION OF ACUTE MYELOGENOUS LEUKEMIA

Subtype	Bone Marrow Definition
AML-M0	$\geq 30\%$ blasts (type I) $< 3\%$ Sudan black B/myeloperoxidase positivity Myeloid antigen expression by immunophenotyping or myeloperoxidase expression by electron microscopy
AML-M1	$\geq 30\%$ blasts (types I, II, rarely III) $\geq 3\%$ Sudan black B/myeloperoxidase positivity in blasts $< 10\%$ cells exhibiting maturation beyond blasts stage
AML-M2	$\geq 30\%$ blasts (types I, II, III) $\geq 3\%$ Sudan black B/myeloperoxidase positivity in blasts $> 10\%$ granulocytic cells exhibiting maturation beyond blasts stage $< 20\%$ monocytic cells
AML-M3	$\geq 30\%$ blasts (types I, II, III) + hypergranular promyelocytes † Intense myeloperoxidase/Sudan black B reaction in virtually all cells
AML-M3v	Same criteria as AML-M3, except that granules within most promyelocytes are inconspicuous and nuclei are highly grooved and reniform
AML-M4	$\geq 30\%$ myeloblasts + monoblasts + promonocytes $\geq 20\%$ Sudan black B/myeloperoxidase positive cells $\geq 20\%$ nonspecific esterase-positive cells
AML-M4 eos	Same criteria as AML-M4, with the addition of abnormal eosinophils in the bone marrow
AML-M5	$\geq 30\%$ myeloblasts + monoblasts + promonocytes $< 20\%$ Sudan black B/myeloperoxidase positive cells $\geq 80\%$ nonspecific esterase-positive cells Monoblasts predominate in M5a Promonocytes predominate in M5b
AML-M6	$\geq 30\%$ of nonerythroid cells are myeloblasts (types I, II, III) $> 50\%$ erythroid elements
AML-M7	$\geq 30\%$ blasts (myeloblasts + megakaryoblasts) $> 30\%$ megakaryocytic elements defined by immunophenotyping or ultrastructural electron microscopy

The National Cancer Institute recommends classification as acute leukemia when peripheral blood blasts exceed 30% regardless of bone marrow blast percentage. Peripheral Blood definitions are variable except for AML-M4, which requires monocytosis ($> 5,000/\text{mm}^3$ [$\geq 5 \times 10^9/\text{L}$]) and increased lysozymes.

†Promyelocytes usually predominate in AML-M3.

**CORRELATION OF FAB CLASSIFICATION
OF ACUTE MYELOGENOUS LEUKEMIA
WITH IMMUNOPHENOTYPIC FEATURES**

FAB SUBTYPE	M1	M2	M3	M4	M5	M6	M7
HLA-DR	+	+	-	+	+	+/-	+/-
CD 33 (MY 9)	+	+	+	+	+	+/-	+/-
CD 13 (MY 7)	+/-	+	+	+	+	-	NR
CD 15 (MY 1, M1)	+/-	+	+/-	+	+	+/-	NR
CD 14 (MY 4, M3)	-	+/-	-	+	+	-	NR
GLYCOPHORIN A	-	-	-	-	-	+	-
PLATELET GP IB/ IIB/ IIIA	-	-	-	-	-	-	+

NR: Not reported

+ : Positive reaction in $\geq 20\%$ of gated blasts

+/- : Positive or negative

Reaction in majority of cases is noted, but exceptions may occur.

FAB CLASSIFICATION OF ACUTE LYMPHOBLASTIC LEUKEMIA

<u>CYTOLOGY</u>	<u>L-1</u>	<u>L-2</u>	<u>L-3</u>
Cell size	Small	Large	Large, homogenous
Nuclear Chromatin	Homogenous	Variable	Finely, Stippled
Nuclear Shape	Regular	Irregular	Oval to round
Nucleoli	Rare	Present	1-3
Cytoplasm	Scanty	Moderate	Moderate, vacuolated
Cytoplasmic Basophilia	Moderate	Variable	Intense
Incidence in Children	85%	13%	2%
Incidence in Adults	35%	63%	2%
Immunologic Markers leukemia/lymphoma	Early B or thymic T	Early B or thymic T	Differentiated B (SIg positive), Burkitt-type

IMMUNOPHENOTYPIC FEATURES OF ALL (Crosswise)

WORKING FORMULATION CLASSIFICATION OF NON-HODGKIN'S LYMPHOMA

Low-grade

- A. Malignant lymphoma, small lymphocytic
Consistent with CLL
Plasmacytoid
- B. Malignant lymphoma, follicular, predominantly small cleaved cell
- C. Malignant lymphoma, follicular, mixed, small cleaved and large cell

Intermediate grade

- D. Malignant lymphoma, follicular, predominantly large cell
- E. Malignant lymphoma, diffuse, small cleaved cell
- F. Malignant lymphoma, diffuse, mixed, small and large cell
- G. Malignant lymphoma, diffuse, large cell

High grade

- H. Malignant lymphoma, large cell, immunoblastic
Plasmacytoid
Clear cell
Polymorphous
Epithelioid cell component
- I. Malignant lymphoma, lymphoblastic
- J. Malignant lymphoma, small noncleaved cell
Burkitt's
Follicular areas

Miscellaneous

- Composite
- Mycosis fungoides
- Histiocytic
- Extramedullary plasmacytoma
- Unclassifiable
- Other

KIEL CLASSIFICATION OF NON-HODGKIN'S LYMPHOMAS

B	T
Low-grade malignant lymphomas	
Lymphocytic	Lymphocytic
Chronic lymphocytic leukemia	Chronic lymphocytic leukemia
Prolymphocytic leukemia	Prolymphocytic leukemia
Hairy cell leukemia	Small cell, cerebriform
Lymphoplasmacytic/ cytoid (immunocytoma)	Mycosis fungoides/ Sèzary's syndrome
Plasmacytic	Lymphoepithelioid (Lennert's lymphoma)
Centroblastic-centrocytic (follicular ± diffuse; diffuse)	Angioimmunoblastic (AILD)
Centrocytic (mantle cell)	T-zone lymphoma
Monocytoid, including marginal zone cell	Pleomorphic, small cell (HTLV -1 ±)
High-grade malignant lymphomas	
Centroblastic	Pleomorphic, medium sized and large cell (HTLV -1 ±)
Immunoblastic	Immunoblastic (HTLV -1±)
Burkitt's lymphoma	Large-cell anaplastic (Ki -1+)
Large-cell anaplastic (Ki -1+)	
Lymphoblastic	Lymphoblastic

Other rare types of lymphoma may be separately identified for T- and B-cell lymphomas, respectively.