Immunophenotype abnormalities

NORMAL BLOOD CELL MATURATION

Normal myeloid differentiation

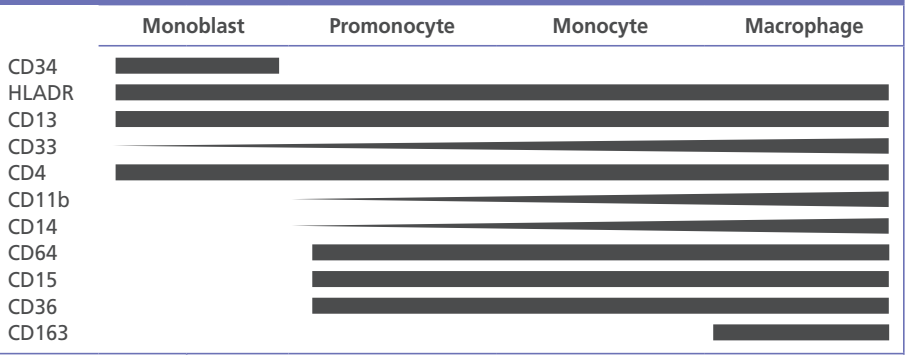
Erythroblast series				
	Pro-ERB	Basophilic	Polychromatophilic	Acidophilic
CD34	■			
CD117	■	■		
CD36	■	■	■	■
CD71	■	■	■	■
CD235a			■	■

Identification of RBC	• CD235a (specific to RBC) • CD36 (also expressed by the monocytic and platelet series) • CD71 (makes it possible to differentiate between erythroblasts which are positive and RBC which are negative)
Other markers of interest	• GPI-anchored proteins CD55 and CD59 (reduced in PNH) • Binding of EMA to protein band 3 (reduced in HS) • CV of CD36 and CD71 (included in the *Red score* which is a guide to the diagnosis of MDS)

Megakaryocytic series			
	Megakaryoblast	Promegakaryocyte	Megakaryocyte
CD34	■		
CD38		■	■
CD61		■	■
CD41		■	■
CD42			■

Identification of platelets	• CD41, CD42 and CD61 (specific to platelets) • CD36 (also expressed by the monocytic and erythroblast series)
Other markers of interest	• CD41 and CD61 (reduced or absent in Glanzmann's thrombasthenia) • CD42 (reduced or absent in Bernard-Soulier syndrome) • CD62p and annexin V (markers of platelet activation)

Monocytic series

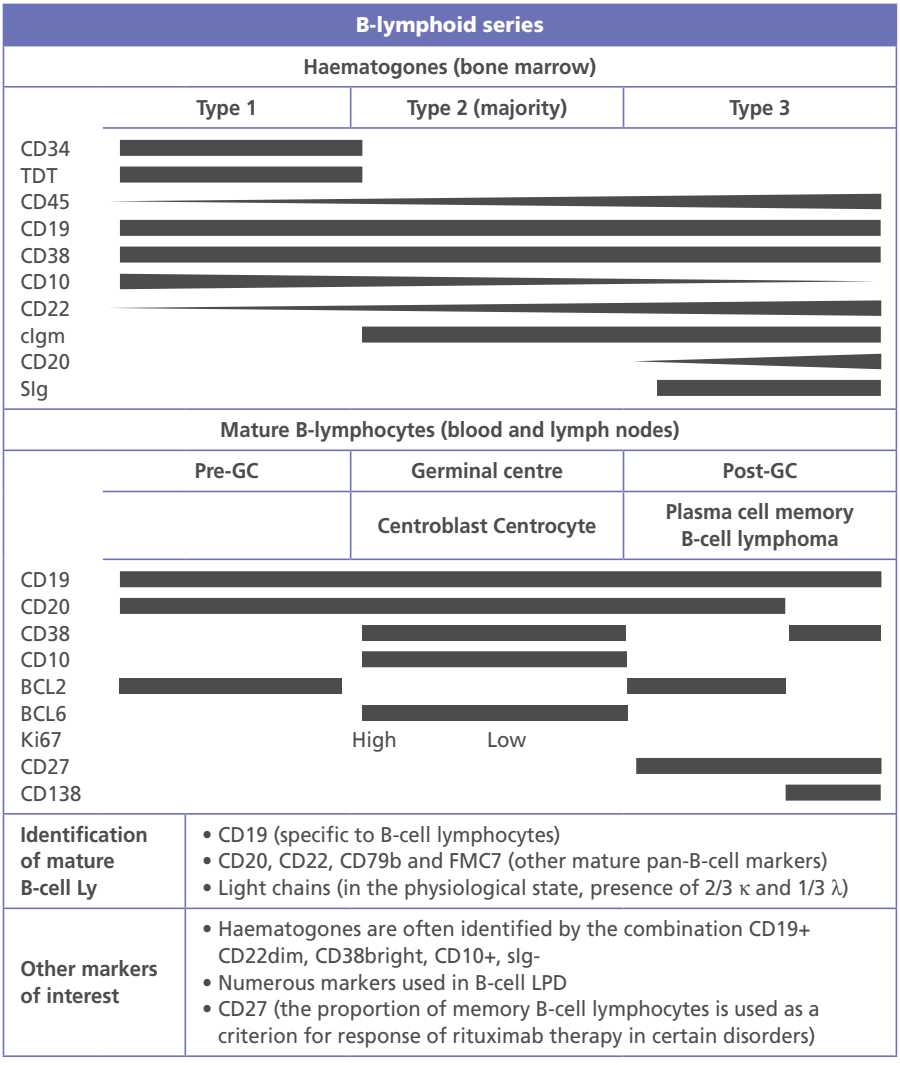


Identification of monocytes	• CD36 (also expressed by the erythroblast and platelet series) • CD13, CD33, CD15 and CD11b (also expressed by the granulocytic series) • CD4 (also expressed by T lymphocytes)
Other markers of interest	• GPI-anchored proteins CD14 and CD157 (reduced in PNH) • A distinction is made between classical CD14+ CD16- monocytes and non-classical CD14dim CD16+ monocytes (useful for diagnosis of CMML)

Granulocytic series

Identification of neutrophils	• CD13, CD33, CD15 and CD11b (also expressed by the monocytic series) • CD10 (also expressed by B lymphocytes derived from the germinal centre)
Other markers of interest	• GPI-anchored proteins CD16 and CD24 (reduced in PNH) • Expression of CD64 and reduction in CD10 in the event of sepsis • Reduction in neutrophil SSC (included in the Ogata score which is a guide to the diagnosis of MDS) • Basophils are differentiated from neutrophils due to their moderate expression of CD45, a lower SSC, a positive CD11b in 50% of cases, together with positive CD203c • Eosinophils are differentiated from neutrophils due to their stronger expression of CD45, negative CD16 and positive CD49d

Normal lymphoid differentiation



Identification of mature B-cell Ly	• CD19 (specific to B-cell lymphocytes) • CD20, CD22, CD79b and FMC7 (other mature pan-B-cell markers) • Light chains (in the physiological state, presence of 2/3 κ and 1/3 λ)
Other markers of interest	• Haematogones are often identified by the combination CD19+ CD22dim, CD38bright, CD10+, sIg- • Numerous markers used in B-cell LPD • CD27 (the proportion of memory B-cell lymphocytes is used as a criterion for response of rituximab therapy in certain disorders)

T-lymphoid series

Immature T-lymphocytes (bone marrow and thymus)

Bone marrow		Thymus (cortical)	Thymus (bone marrow)	
Pro-T	Pre-T	Cortical T	DP T	SP T

Mature T lymphocytes (periphery)

Naïve	Memory	Activated

Identification of mature T-cell Ly	• CD3s (specific to T-cell lymphocytes) • CD2 (also expressed by NK) • CD4 (auxiliary TL); CD8 (cytotoxic TL) • The T4/T8 ratio is generally between 2 and 4. There are < 5% double-positives and < 10% double-negatives
Other markers of interest	• The majority of mature T lymphocytes express TCR αβ (> 95%); these may be single-positive (4+ or 8+), double-positive or double-negative • A minority of T lymphocytes express TCR γδ (< 5%); these display strong expression of CD3 and are predominantly double-negative or CD8dim • CD25 and HLA-DR (activation markers) • NK markers (a variable proportion of CD8+ T lymphocytes physiologically express CD56. CD16 and CD57 are NK markers often found in LGL leukaemia) • Study of CD158 isoforms (KIR repertory restriction in T-cell LPD) • Ultraminority T lymphocyte population more common in subjects > 70 years (notably CD4+ CD8dim and CD4dim CD8+)

NK-lymphoid series	
Identification of NK-cell Ly	• CD2+ CD3- (combination allowing NK to be identified) • CD16 and CD56 (make it possible to differentiate between "CD16bright CD56dim" cytotoxic NK Ly, "CD16dim CD56bright" interferon-γ producing NK Ly and "CD16-CD56dim" antitumoural NK Ly) • CD57 (other marker expressed by NK Ly)

MAIN AB USED IN HAEMATOLOGY

The list of Ab is deliberately non-exhaustive. Only the most widely used and most useful Ab are stated.

Ab	Normal distribution	Clinical impact
CD1a	• T-lymphoid precursors	• T-cell ALL: used for classification (III)
CD2	• T and NK lymphocytes	• T-cell LPD: absent or weak in certain cases • AML: aberrant expression in certain cases • Systemic mastocytosis: aberrant expression in certain cases
CD3c	• T-lymphoid precursors • T and NK lymphocytes	• T-cell LPD: guides diagnosis of the CD3s negative forms • ALL: indicator for the T-lymphoid series
CD3s	• "Mature" T-lymphoid precursors • T lymphocytes	• T-cell LPD: absent or weak in certain cases • T-cell ALL: used for classification (IV)
CD4	• Certain T lymphocytes • Monocytes (weak CD4)	• T-cell LPD: guides diagnosis of T-cell LPD • AML: aberrant expression in certain cases
CD5	• T lymphocytes • Minority of B lymphocytes	• B-cell LPD: highly positive in CLL and MCL. Weakly positive in 5% of MZL and B-cell PLL. Sometimes positive in DLBCL (development of CLL into Richter syndrome or *de novo* form) • T-cell LPD: absent or weak in certain cases (notably LGL leukaemia and T-cell lymphoma NOS)
CD7	• T and NK lymphocytes	• AML: aberrant expression in certain cases • T-cell LPD: absent or weak in certain cases (notably Sézary syndrome and ATLL)
CD8	• Certain T lymphocytes (strong = cytotoxic; weak = Lγδ)	• T-cell LPD: guides the diagnosis of T-cell LPD and sometimes positive in NK LGL leukaemia
CD9	• B lymphocytes	• ALL: indicator for the B-lymphoid series

Ab	Normal distribution	Clinical impact
CD10	• T and B lymphocytes (positive for germinal centre precursors and a minority of mature cells) • Haematogones • Neutrophils	• B-cell LPD: positive in FL, Burkitt's lymphoma and certain DLBCL (indicator for the germinal centre) • T-cell LPD: sometimes positive in angioimmunoblastic lymphoma • B-cell ALL: used for classification (II ± III and IV)
CD11c	• Minority of B and T lymphocytes • Granulocytic and monocytic series	• B-cell LPD: highly positive in HCL and SRPL. Negative or weak in CLL and MZL
CD13	• Granulocytic and monocytic series	• AML: indicator for the myeloid series • B-cell LPD: indicator for lymphoplasmacytic differentiation
CD14	• Monocytes	• Indicator for monocytes • CMML: guides the diagnosis of CMML (in combination with CD16)
CD15	• Granulocytic and monocytic series	• Acute leukaemia: aberrant expression in certain B-cell ALL (+) and AML (-)
CD16	• NK lymphocytes • Certain CD8+ TL • Granulocyte precursors	• Indicator for the NK series • T-cell LPD: expressed in certain CD8+ T LGL leukaemia • AML: aberrant expression in certain cases (-)
CD19	• B lymphocytes • Plasma cells	• Indicator for the B series • B-cell LPD: absent or weak in certain FL and DLBCL
CD20	• B lymphocytes • "Mature" haematogones	• Indicator for the B series • B-cell LPD: absent or weak in CLL and certain DLBCL. Strong expression in HCL and SRPL • TT: may be "false-negative" during treatment with an anti-CD20 antibody
CD22	• B lymphocytes • Basophils	• B-cell LPD: reduced expression in CLL Increased expression in HCL and SRPL
CD23	• B lymphocytes (weak expression but increased during activation)	• B-cell LPD: helps differentiate between CLL (positive) and other B-cell LPD (negative or weak)
CD24	• B lymphocytes	• B-cell LPD: helps differentiate between MZL (often positive) and SRPL (often negative). Often negative in HCL

Ab	Normal distribution	Clinical impact
CD25	• Activated B and T lymphocytes	• B-cell LPD: positive in HCL and Waldenström macroglobulinemia (more rarely in CLL and MZL) • T-cell LPD: highly positive in ATLL and heterogenous fluorescence intensity in Sézary syndrome • Systemic mastocytosis: aberrant expression in certain cases • B-cell ALL: expression associated with the BCR-ABL+ forms • TT: therapeutic target of basiliximab
CD26	• T-lymphoid precursors • Majority of CD4+ TL • NK lymphocytes	• T-cell LPD: diagnostic criteria for Sézary syndrome (CD4+ CD26- > 30%)
CD27	• Memory B lymphocytes • Plasma cells	• B-cell LPD: helps differentiate between MZL (often positive) and SRPL (often negative) • MM: sometimes negative
CD28	• T lymphocytes	• MM: aberrant expression in certain cases
CD33	• Myeloid and monocytic cells	• AML: indicator for the myeloid series • MM: aberrant expression in certain cases (therapeutic impact)
CD34	• Marker of myeloid and lymphoid immaturity (T and B)	• Acute leukaemia: frequent expression in the majority of AML and B-cell ALL. Rare expression in T-cell ALL, AML3, AML5 and AML6 • Identification of myeloblasts which are "physiological" and associated with MPN
CD36	• Monocytes • Red blood cells • MK and platelets	• Identification of residual erythroblasts • AML: indicator for the erythroid, megakaryocytic or monocytic series according to the associated markers
CD38	• B/T lymphoid and myeloid precursors • Plasma cells (strong) • Certain B lymphocytes (derived from the germinal centre)	• MM: identification of plasma cells in combination with CD138 (absent from plasmablasts) • B-cell LPD: often positive MCL. Sometimes positive in MZL and Waldenström macroglobulinemia (indicator for lymphoplasmacytic differentiation). Positive in FL and certain DLBCL (indicator for the germinal centre)

Ab	Normal distribution	Clinical impact
CD41	• MK and platelets	• Glanzmann's thrombasthenia: negative or weak • AML: indicator for the megakaryocytic series (platelets sometimes adhere to AML blast cells and may yield false-positives)
CD42	• MK and platelets	• Bernard-Soulier syndrome: negative or weak
CD43	• Minority of B lymphocytes • T lymphocytes • Myeloid precursors	• B-cell LPD: often positive in CLL and MCL. Sometimes positive in MZL, DLBCL and Burkitt's lymphoma
CD45	• Pan-leukocytic	• Acute leukaemia: usually weak expression • B-cell LPD: usually strong expression
CD45 RA	• Certain B and T lymphocytes (majority of naïve TL)	• T-cell LPD: guides diagnosis
CD45 RO	• Certain B and T lymphocytes (majority of memory TL)	• T-cell LPD: guides diagnosis
CD52	• Lymphocytes • Monocytes	• TT: therapeutic target of alemtuzumab
CD56	• NK lymphocytes • Certain CD8+ TL	• T-cell LPD: expressed in certain CD8+ T LGL leukaemia • AML: aberrant expression in certain cases • MM: aberrant expression in certain cases
CD57	• NK lymphocytes • Certain CD8+ TL	• T-cell LPD: expressed in certain CD8+ T LGL leukaemia
CD58	• Haematopoietic and non-haematopoietic cells	• B-cell ALL: sometimes positive (utility for RD)
CD61	• MK and platelets	• AML: indicator for the megakaryocytic series (platelets sometimes adhere to AML blast cells and may yield false-positives)
CD62p	• Certain platelets	• Marker for platelet activation
CD64	• Monocytes • Granulocyte precursors	• AML: indicator for the myeloid series
CD65	• Granulocyte precursors	• AML: indicator for the myeloid series
CD68	• Monocytes and macrophages • CDP	• Acute leukaemia: expressed in the majority of AML and a few B-cell ALL
CD71	• Erythroid precursors (strong)	• AML: indicator for the erythroid series • Identification of residual erythroblasts
CD79a	• B lymphocytes	• ALL: indicator for the B-lymphoid series