London Health Sciences Centre

St JOSEPH's HEALTH CARE LONDON

Pathology and Laboratory Medicine

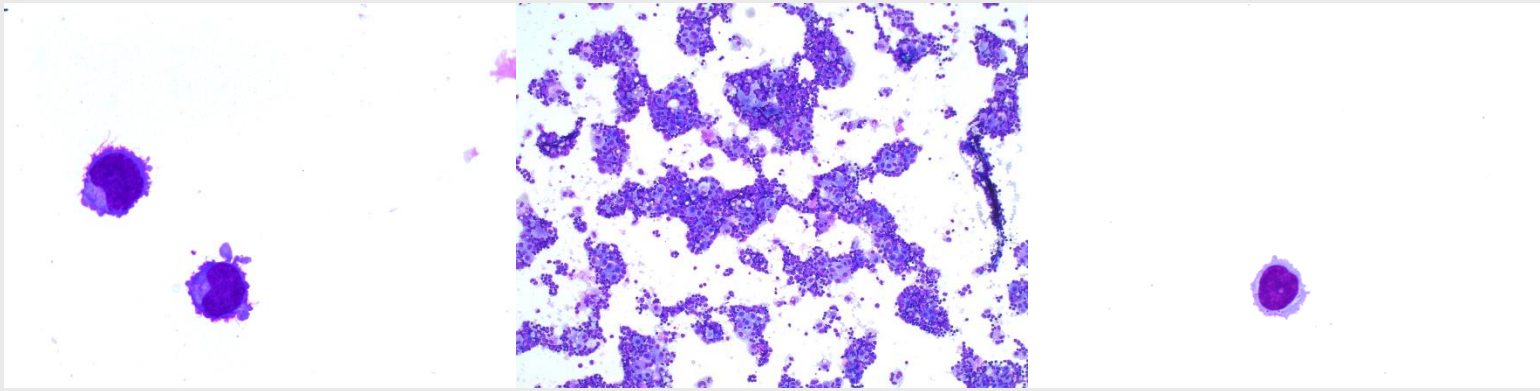
Non-Malignant Flow Cytometry in Hematology

Flow Cytometry Test Menu

- Leukemia/lymphoma immunophenotyping
- MRD (Lymphoid only)
- HIV
- CD34
- Solid organ Crossmatch
- Transplant monitoring
- PNH
- Hb F
- Basic immunology
- Research applications

Flow Cytometry in Hematology

- Role in hematologic disease characterization.
- 50% of samples are negative, doesn't meant the patient doesn't have disease.
- Immune function, red cell function and platelet function

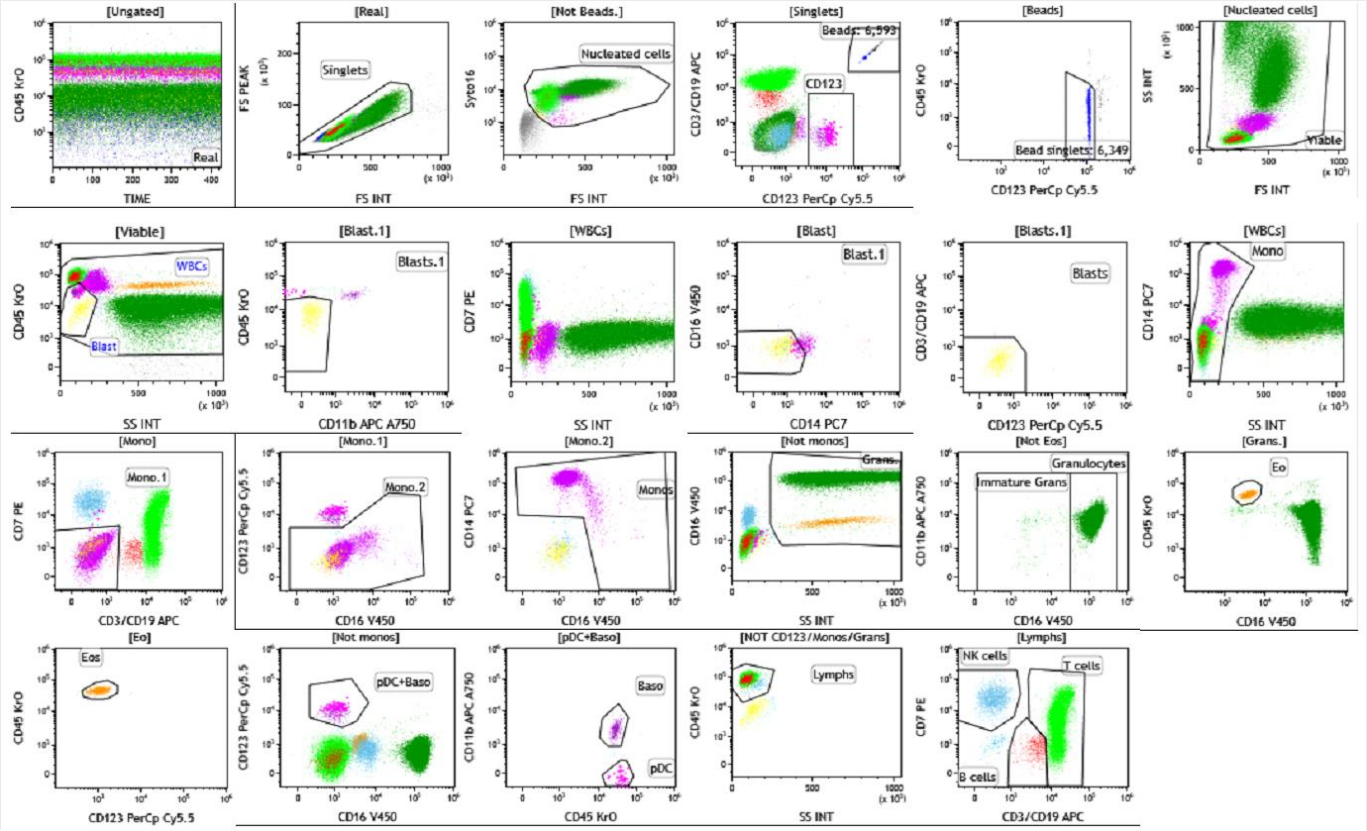


Flow Cytometry in Hematology

- Abnormal cell populations and does not mean malignant
- Context

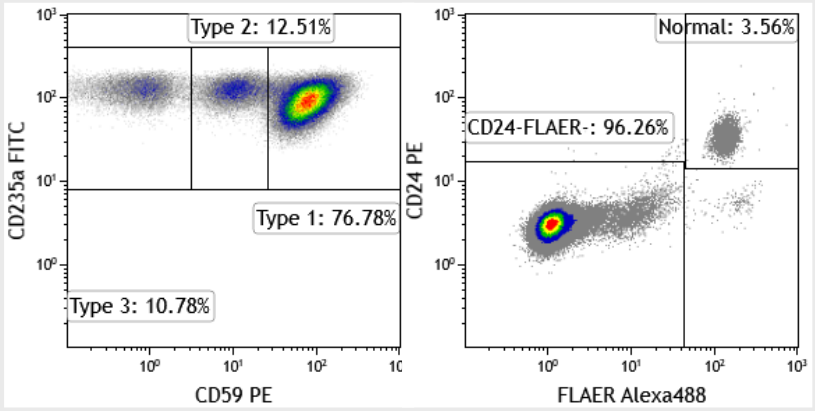
Importance of Non-Malignant Flow Cytometry

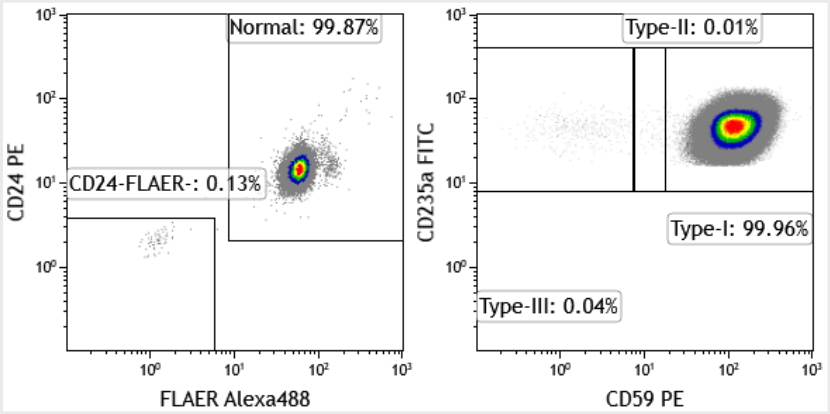
- Goal: Detect and classify abnormal cell populations without malignancy.
- Conditions covered: immune cytopenias, reactive lymphocytosis, immunodeficiencies.



Immune Cytopenias Overview

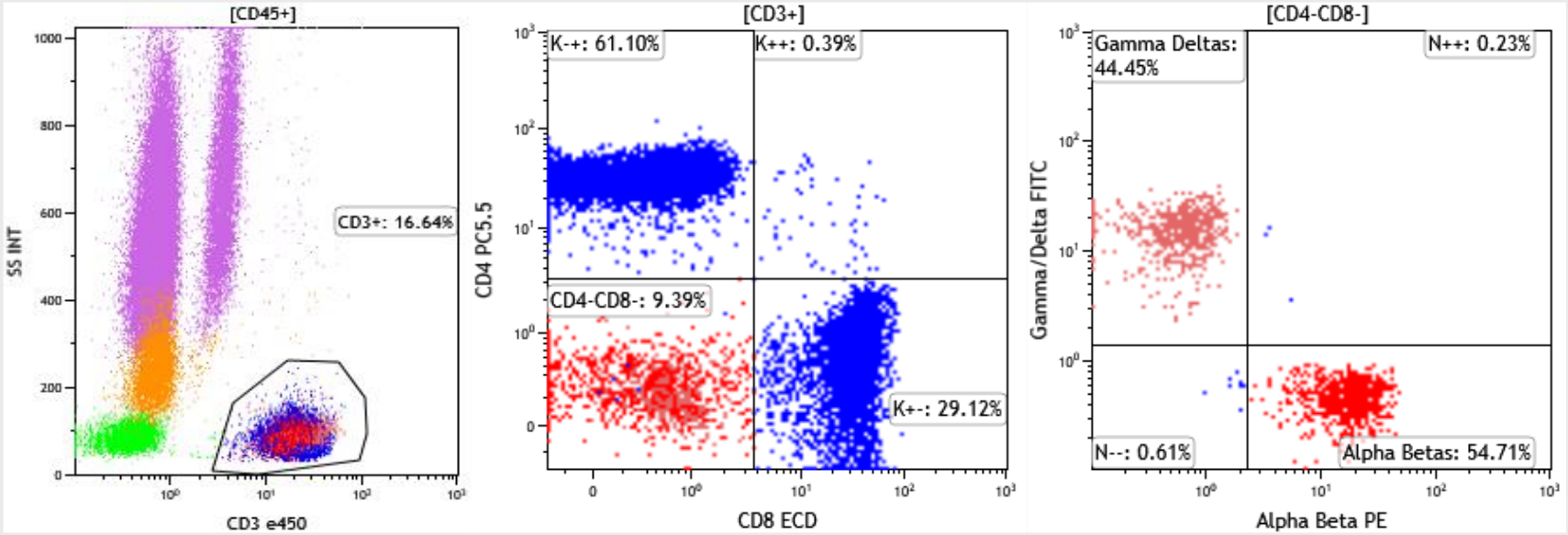
- Flow cytometry identifies autoantibodies or complement-bound erythrocytes.
- Helps in diagnosing autoimmune hemolytic anemia and immune thrombocytopenic purpura.
- Assessing for PNH clones.





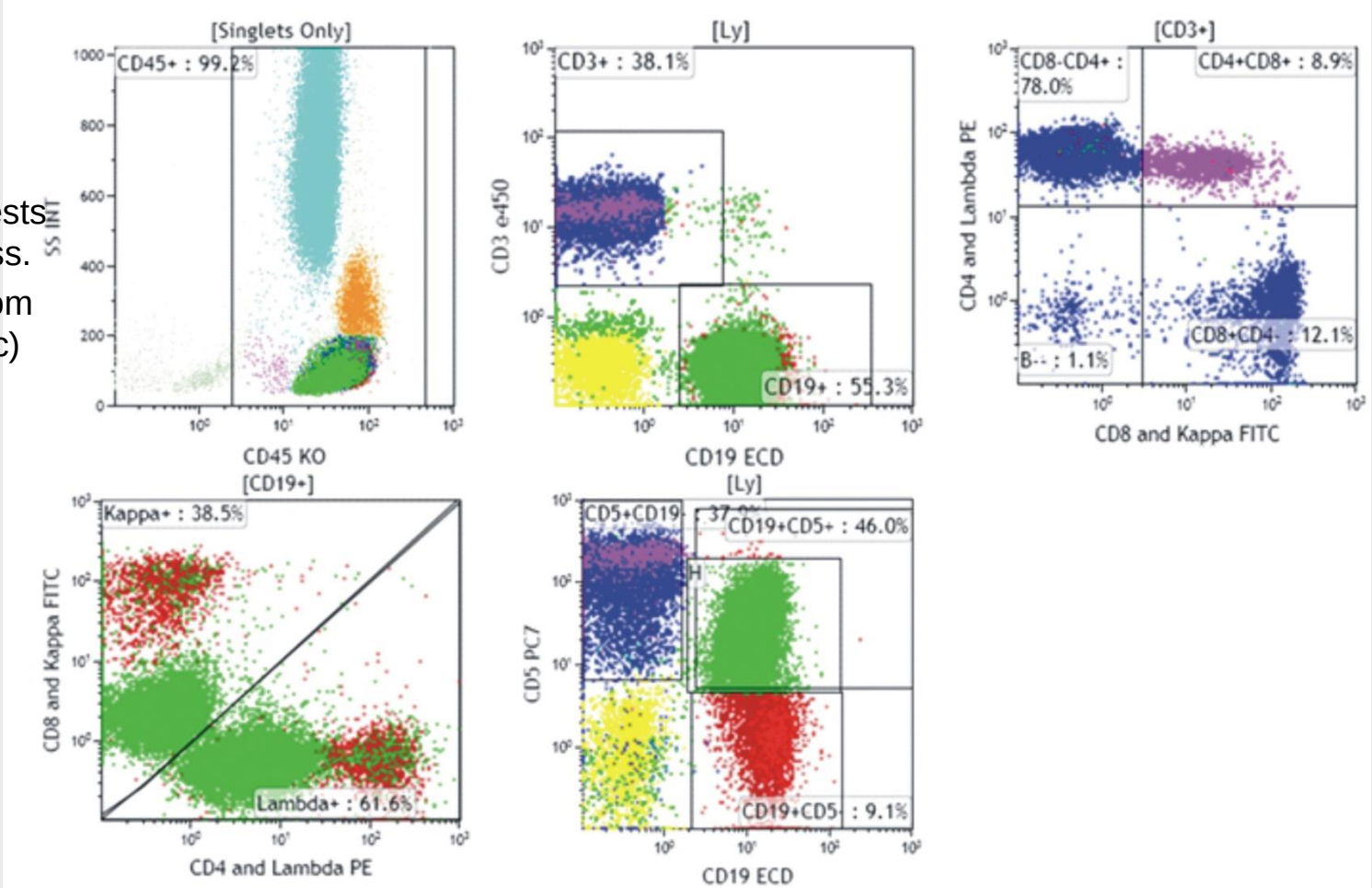
Flow Cytometry Markers for Immune Cytopenias

- CD4 discovered in 1970s
- Used in the 1980s for HIV monitor

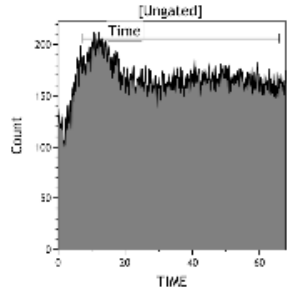


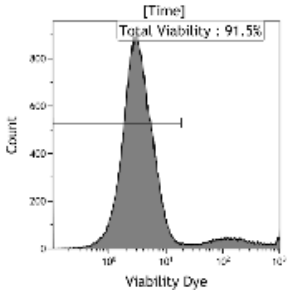
Reactive Lymphocytosis Overview

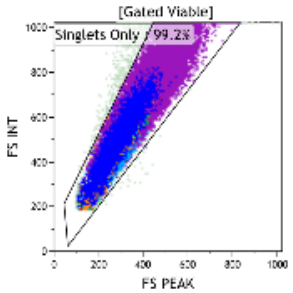
- Reactive lymphocytosis
- Polyclonal lymphocyte expansion suggests a reactive process.
- Differentiation from clonal (neoplastic) lymphocyte expansions.

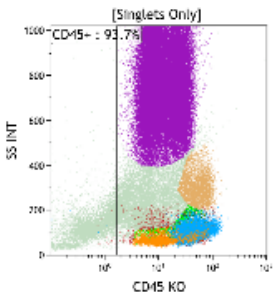


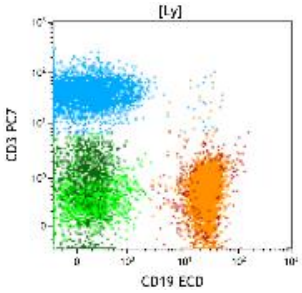
Screening of a normal bone marrow

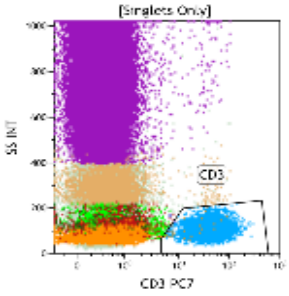


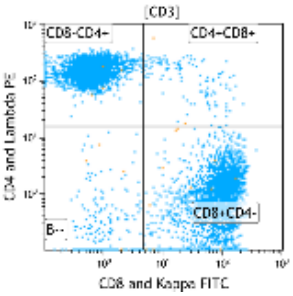


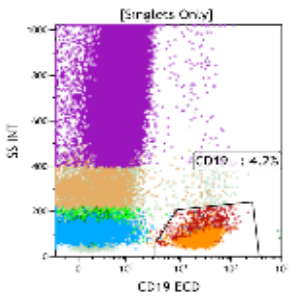


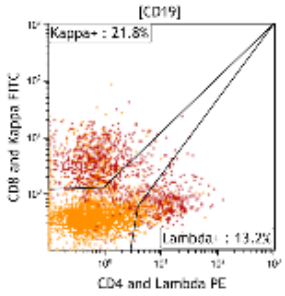


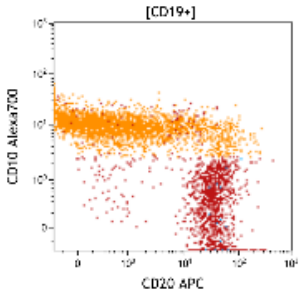


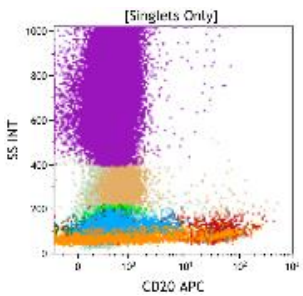


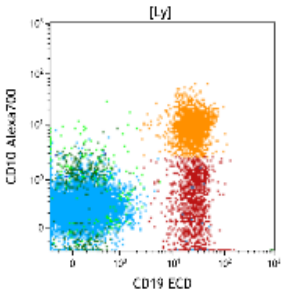


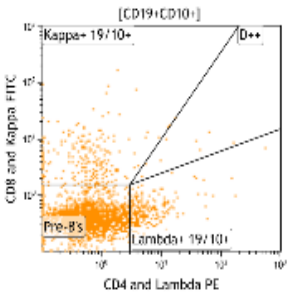


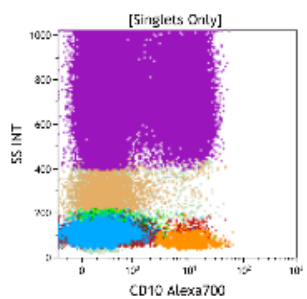


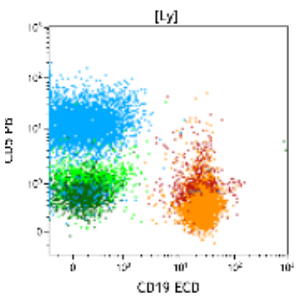


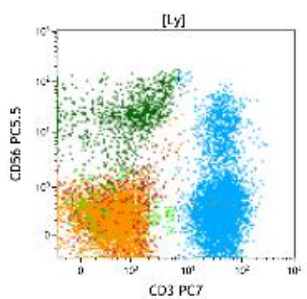






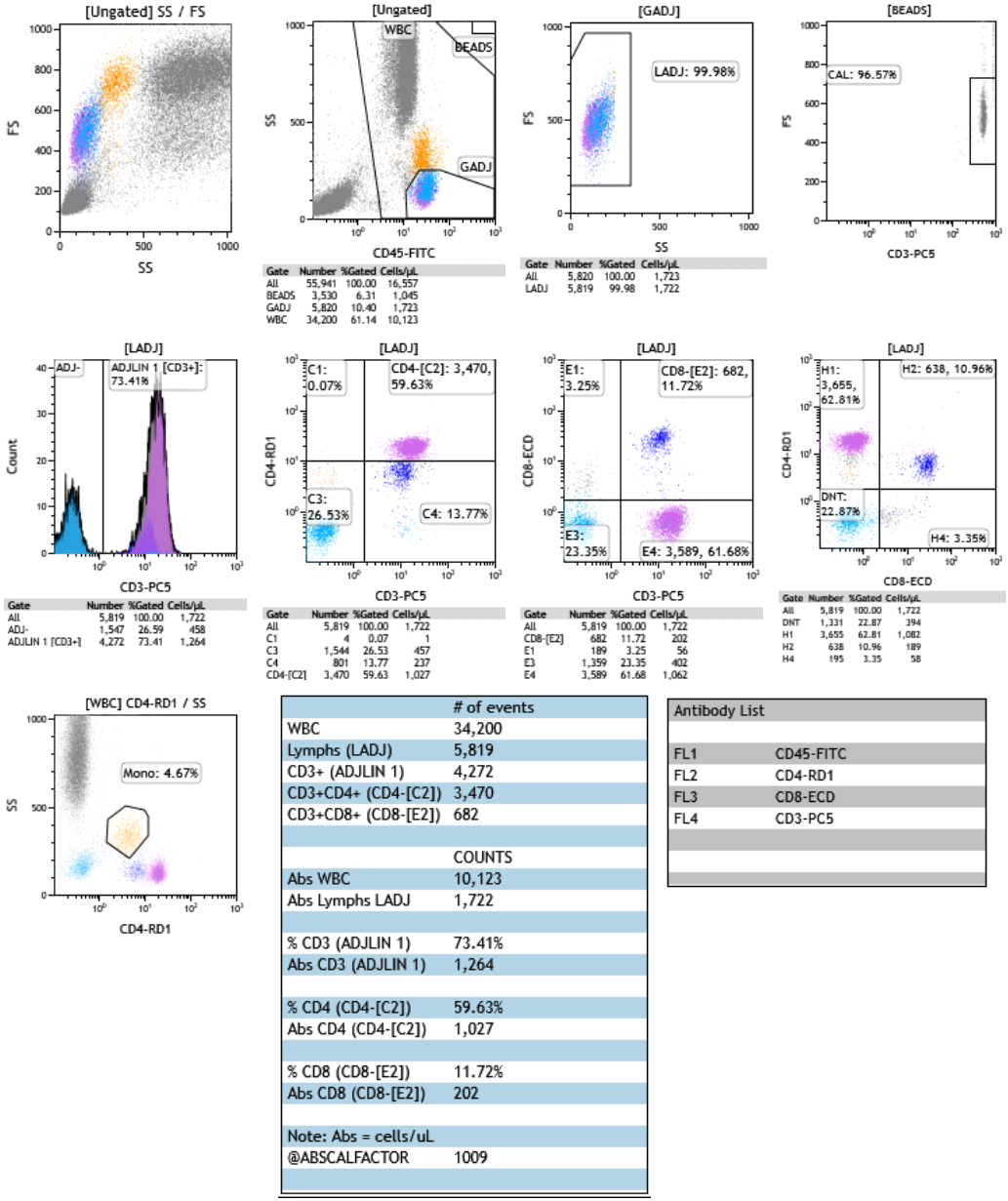






Flow Cytometry Markers for Immunodeficiencies

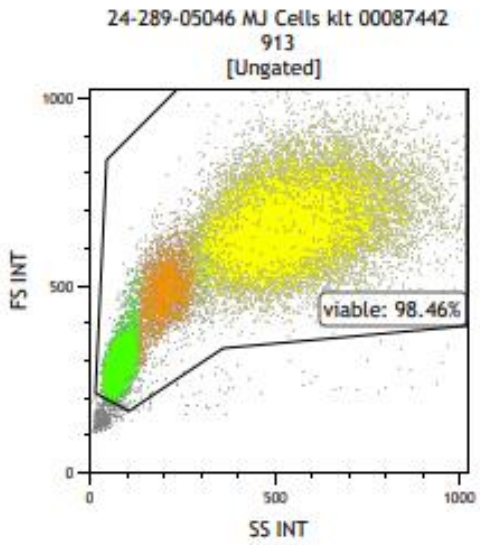
- Kaluza version with calculation of CD4 enumeration
- Platform agnostic
- IVD gating of a non-IVD test

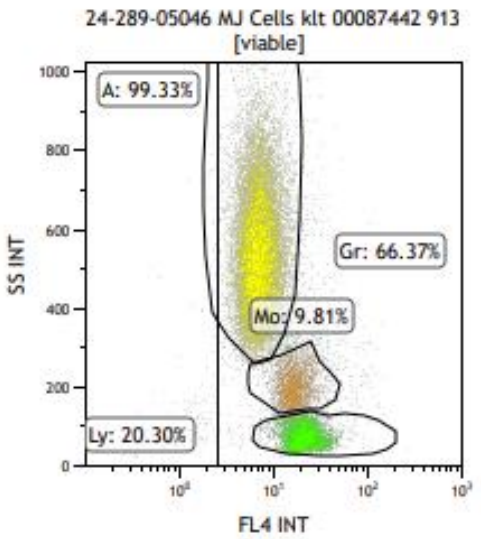


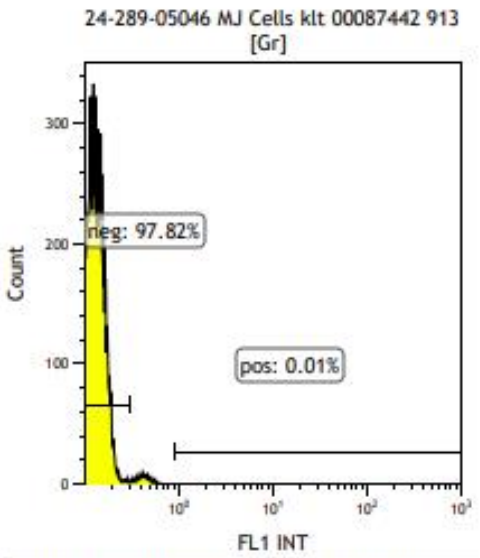
	# of events
WBC	34,200
Lymphs (LADJ)	5,819
CD3+ (ADJLIN 1)	4,272
CD3+CD4+ (CD4-[C2])	3,470
CD3+CD8+ (CD8-[E2])	682
	COUNTS
Abs WBC	10,123
Abs Lymphs LADJ	1,722
% CD3 (ADJLIN 1)	73.41%
Abs CD3 (ADJLIN 1)	1,264
% CD4 (CD4-[C2])	59.63%
Abs CD4 (CD4-[C2])	1,027
% CD8 (CD8-[E2])	11.72%
Abs CD8 (CD8-[E2])	202
Note: Abs = cells/uL	
@ABSCALFACTOR	1009

Antibody List	
FL1	CD45-FITC
FL2	CD4-RD1
FL3	CD8-ECD
FL4	CD3-PC5

CGD Assay

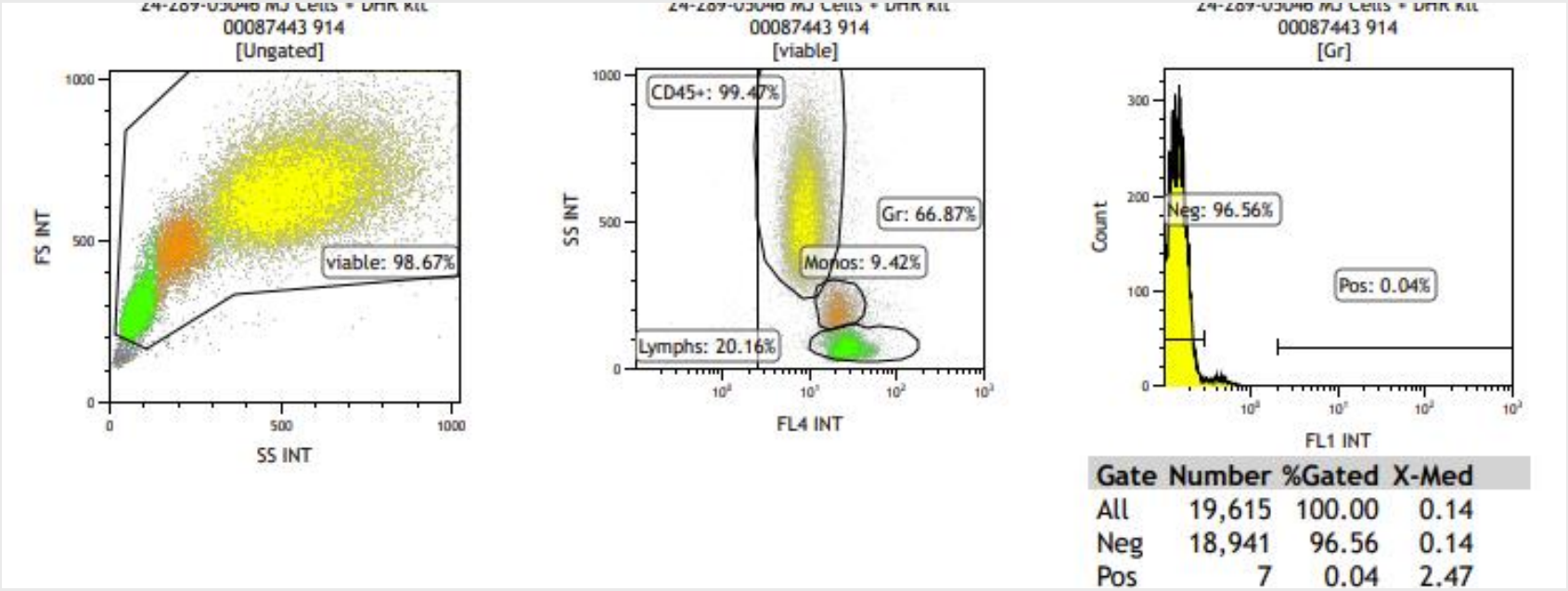






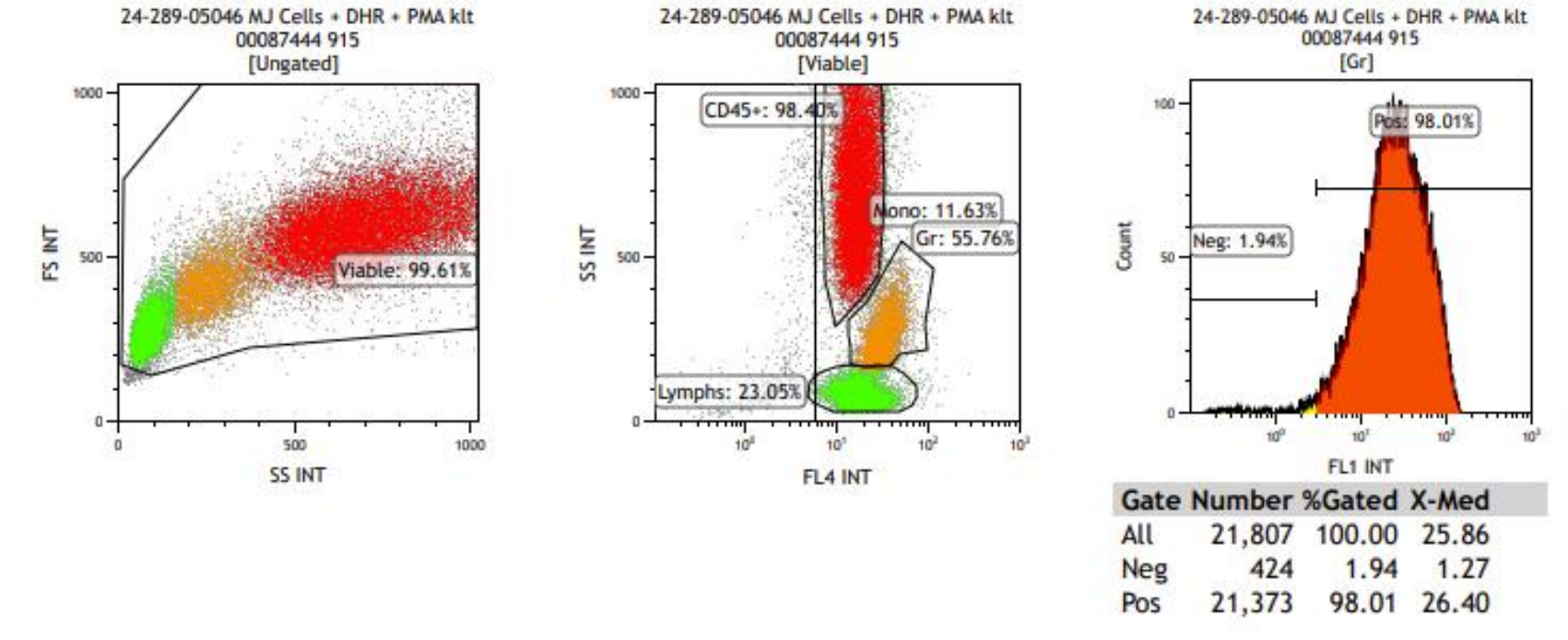
Gate	Number	%Gated	X-Med
All	19,672	100.00	0.13
neg	19,244	97.82	0.13
pos	2	0.01	1.29

CGD Assay



Gate	Number	%Gated	X-Med
All	19,615	100.00	0.14
Neg	18,941	96.56	0.14
Pos	7	0.04	2.47

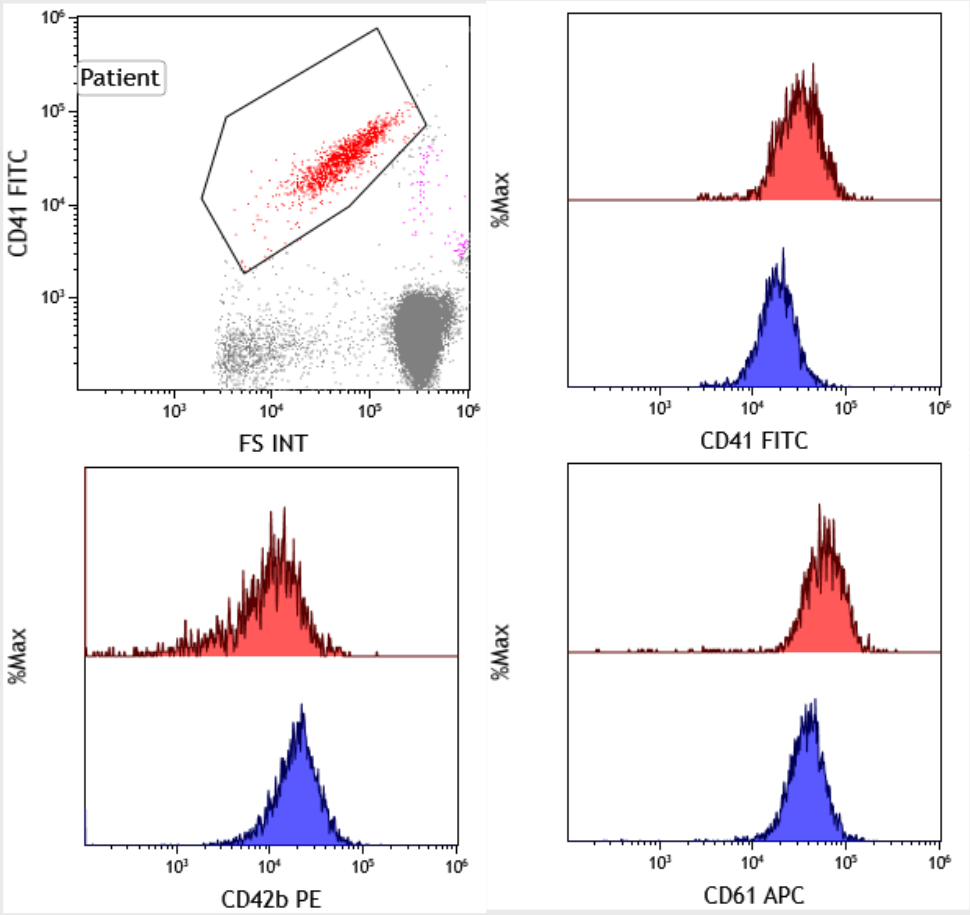
CGD Assay



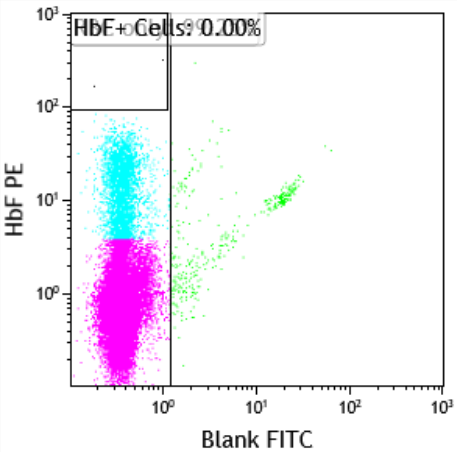
Gate	Number	%Gated	X-Med
All	21,807	100.00	25.86
Neg	424	1.94	1.27
Pos	21,373	98.01	26.40

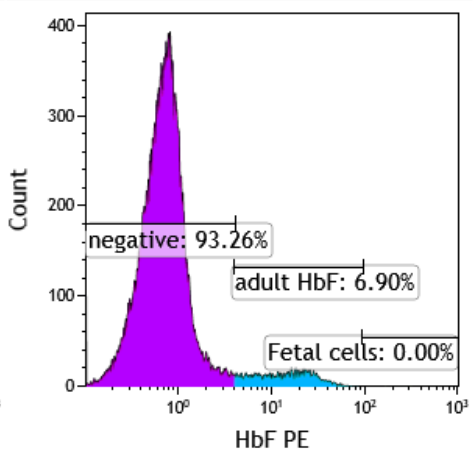
Additional Cell Populations: Red Cells and Platelets

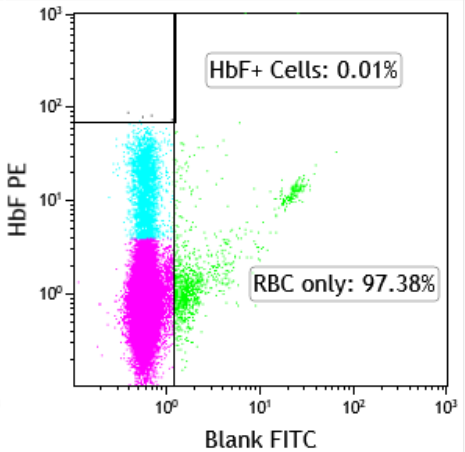
- Red cells and platelets can be analyzed in flow cytometry.
- Flow cytometry assesses abnormalities in these populations.
  - Importance in combination with other tests and clinical findings.

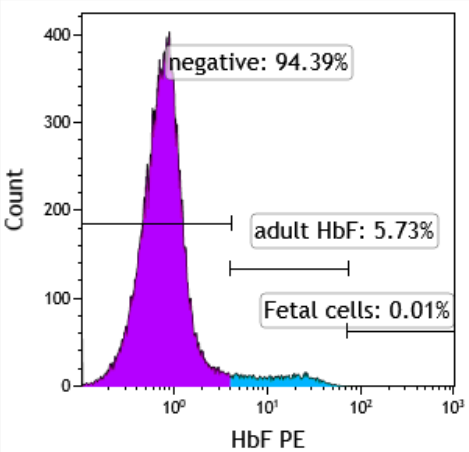


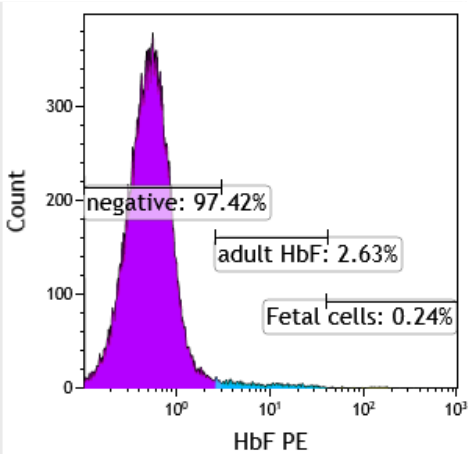
Case Studies

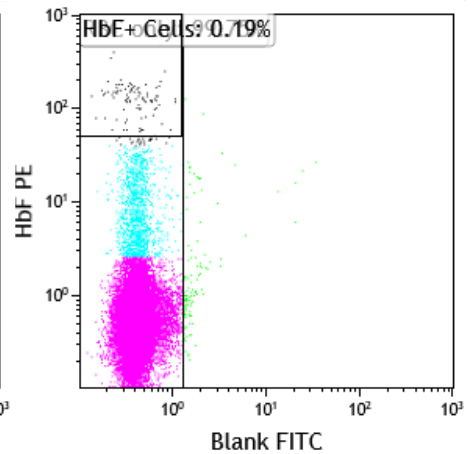






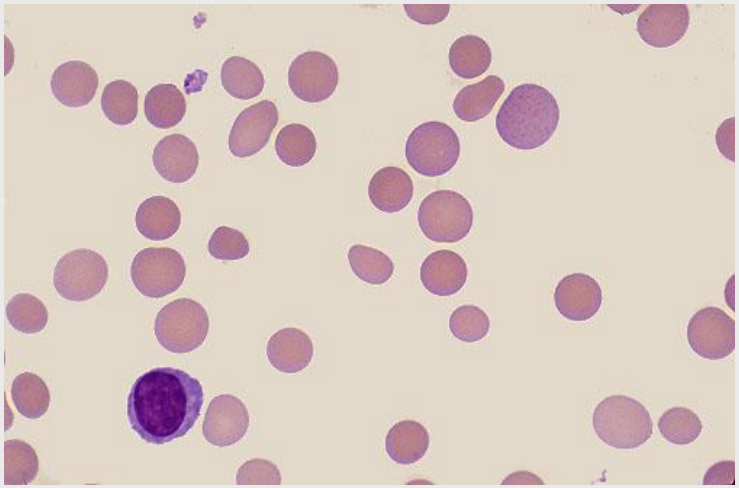




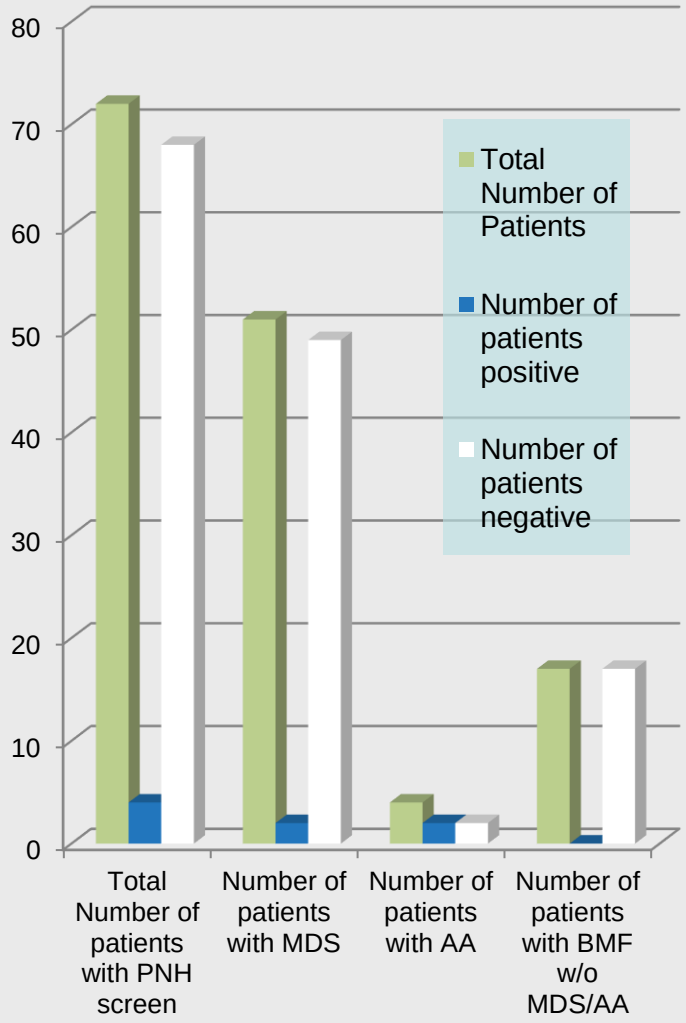


Advantages of Non-Malignant Flow Cytometry

- Specificity,
- Rapid diagnosis
- Monitoring.
- Non-malignant flow cytometry aids in providing accurate diagnoses and guiding treatment strategies.



Limitations and Considerations



- Sample quality
- Cost
- Marker availability.
- Areas for improvement
- Current challenges.

Emerging Trends in Flow Cytometry

- Advancements in automation
- Future directions that could enhance non-malignant disorder diagnostics
- Flow cytometry's role in non-malignant conditions and its importance in hematology
- Not for every laboratory
- Depend on the patient population

Acknowledgments