Lab Handbook Blood Science Analyte (Test) Proforma

Analyte update: Lymphocyte Subset Quantitation (TBNK)

Prepared for update of the Liverpool Clinical Laboratories Pathology Handbook website entry.

Field	Detail
Test Name (Analyte)	Lymphocyte Subset Quantitation
Alternative Name(s) and Keywords	Lymphocyte subsets; TBNK; CD3/CD4/CD8; CD19; CD16/56 NK cells; CD348; HIV348; ATG348; LYMSUB; flow cytometry; EDTA blood.
Discipline/Specialty	Blood Sciences - Clinical Immunology.
Description	Flow cytometric enumeration of T cells, B cells and NK cells in EDTA whole blood using BD Multitest reagents on the BD FACSLyric. Samples may be prepared manually or using the BD FACSDuet.
Clinical Indication	Investigation of suspected cellular immunodeficiency, monitoring immune function in HIV, monitoring immune suppression or ATG therapy, and monitoring B cell depletion or recovery where clinically indicated. Non-HIV related cellular immunodeficiency requests should include clinical details and may require discussion with Immunology medical staff.
Patient Preparation	No specific patient preparation required. Please provide relevant clinical details, including HIV status or monitoring indication, ATG therapy, rituximab, immunosuppressive therapy, transplant context, acute infection, or suspected primary immunodeficiency.
Specimen Container	Purple / lavender top EDTA tube. Do not use heparin or citrate tubes for routine lymphocyte subset quantitation. Do not store in the fridge.
Container Image	Purple / lavender EDTA tube. Website image may use existing EDTA tube image. 
Primary Sample Type	Whole blood.

Minimum Volume Required (µL for serum//blood/urine etc. unless otherwise stated)	3 mL EDTA blood preferred for routine testing. The analytical procedure requires 50 µL whole blood per BD Trucount tube; a minimum of 300 µL per tube is required.
Special Precautions / Requirements	Routine HIV348 and LYMSUB testing is not available at weekends. Samples for HIV348 and LYMSUB must be receipted in the laboratory by 16:30 on Friday. For urgent ATG weekend requests, contact the Laboratory Manager in advance by email and mobile to arrange testing. Requests are time-sensitive and must include sufficient clinical information.
Transport and Storage Requirements	Store and transport at room temperature. Do not refrigerate. Samples should be received by the laboratory within 24 hours of venepuncture. Samples should be stained within 48 hours of collection and acquired within 24 hours of staining, in line with BD method requirements.
Telepath Test Code	Telepath Blood Science / Blood Sciences codes: LYMSUB, HIV348 and ATG348. LYMSUB reports Tube 1 and Tube 2. HIV348 and ATG348 report .
National Pathology Code (READ/SNOMED CT)	To be supplied by LCL I.T. / LIMS team if required.
Reference Interval(s)	Age-related reference intervals apply. See Appendix 1. Absolute counts are displayed as x10^6/L. The numeric values are equivalent to cells/mm3 or cells/µL in the source reference table. Caveat: due to LIMS age-band limitations, age labels around 9 to 15 months may not exactly match the source age bands; see Appendix 1.
Telephone Action Limit(s)	No routine numerical telephone action limit. Urgent ATG monitoring or clinically unexpected critical results should be escalated to the Senior BMS and, where appropriate, the Consultant Immunologist or clinical team.
Measurement Units	Percentages are reported as %. Absolute counts are reported as x10^6/L. T4:T8 ratio is unitless.
Clinical Interpretation	Age, acute infection and immunosuppressive drugs can alter T and B lymphocyte numbers. CD4 count is used to monitor immune function in patients known to be infected with HIV. It does not diagnose HIV infection and must not be used as a substitute for HIV viral studies. B-cell counts may support monitoring after rituximab or other B-cell depleting therapy. Results requiring clinical interpretation are sent to Clin queue.
Useful Links / Guidelines	Pathology Handbook page: http://pathlabs.rlbuht.nhs.uk/lymphocytesubsetquantitationfacsanalysis__i.htm Reference interval source: Comans-Bitter WM et al. Immunophenotyping of blood lymphocytes in childhood. Journal of Pediatrics. 1997;130:388-393. Internal documents: Lymphocyte subset analytical SOP, non-analytical SOP, CC-LCL-79, LIMS/ICE implementation evidence.
Common Interferences / Causes of Spurious Results	Clotted, haemolysed, refrigerated or lipaemic samples may give unreliable results. Blast cells, abnormal lymphocyte populations, severe lymphopaenia, immunosuppressive drugs and monoclonal antibody therapy may affect interpretation. Insufficient mixing, incorrect tube, delayed transport or failed IQC may invalidate results.
Availability of Clinical Advice	Clinical advice is available from the Consultant Immunologist / Immunology medical staff via the Clinical Immunology laboratory. Urgent ATG weekend testing must be arranged in advance with the Laboratory Manager by email and mobile.
Significant Change Values	Not formally established. Interpret serial changes in the context of clinical condition, treatment, previous results and local measurement uncertainty.
Testing Frequency / Minimum Re-testing Interval	Repeat frequency should be based on clinical indication. For suspected immunodeficiency, repeat at significant change of clinical symptoms. HIV and ATG monitoring should follow the relevant clinical protocol.

Related tests	Full blood count and differential; immunoglobulins; vaccine responses; HIV viral load; CD19 monitoring; immunophenotyping panels; primary immunodeficiency investigations.
Technology & Analytical Principle Used	Flow cytometry using BD Multitest direct immunofluorescence reagents, BD Trucount tubes and BD FACSLyric with BD FACSuite Clinical software. Manual preparation or BD FACSDuet automated preparation may be used.
EQA Scheme	UK NEQAS LI Immune Monitoring.
Laboratory Performed	Liverpool Clinical Laboratories, Clinical Immunology laboratory, Royal Liverpool University Hospital site.
UKAS Accreditation Status	Method/platform update under CC-LCL-79. UKAS status to be confirmed following controlled review and submission/assessment evidence for the BD FACSLyric and BD FACSDuet implementation.

Form completed by: Immunology Manager Date: 21 May 2026

Change control completed by: CC-LCL-79 Date: 21 May 2026
(QMS-EXTD-160, LCL Laboratory Handbook)

Appendix 1: Reference intervals for Lymphocyte Subsets

Reference intervals are sourced from the attached Immunodeficiency T&B Lymphocyte Subsets reference table. Paediatric intervals are from Comans-Bitter WM et al. Immunophenotyping of blood lymphocytes in childhood. Journal of Pediatrics. 1997;130:388-393. Absolute counts in the source are cells/mm3. They are displayed here as µL, with the same numeric values.

LIMS age-band caveat

Due to LIMS age-reference limitations, the report may display operational age bands rather than the exact source age bands. The source "5-9 mo" interval is displayed operationally as "Up to 40 weeks". The source "9-15 mo" interval is displayed operationally as "Up to 1 year". For patients aged 9 to 15 months, the actual source reference interval is "9-15 mo" and this should be used for clinical interpretation. For patients aged 12 to 15 months, do not interpret solely against the operational "After 1 up to 2 years" label. The source "15-24 mo" interval applies after 15 months up to 24 months.

Subset	Source age band	%	Absolute Count (µL)
CD3+	9-15 mo	54-76	1600-6700
CD3+CD4+	9-15 mo	31-54	1000-4600
CD3+CD8+	9-15 mo	12-28	400-2100
CD19+	9-15 mo	15-39	600-2700
CD3-CD16/56+	9-15 mo	3-17	200-1200

CD3+

Website / LIMS age band	Source age band	%	Absolute Count (µL)
Up to 7 days	Neonates	28-76	600-5000
Up to 8 weeks	1wk - 2mo	60-85	2300-7000
Up to 20 weeks	2-5 mo	48-75	2300-6500
Up to 40 weeks	5-9 mo	50-77	2400-6900
Up to 1 year	9-15 mo	54-76	1600-6700
After 1 up to 2 years	15-24 mo	39-73	1400-8000
After 2 up to 5 years	2-5 y	43-76	900-4500
After 5 up to 10 years	5-10 y	55-78	700-4200
After 10 up to 16 years	10-16 y	52-78	800-3500
Adult	Adults	58-86	700-2100

CD3+CD4+

Website / LIMS age band	Source age band	%	Absolute Count (µL)
Up to 7 days	Neonates	17-52	400-3500
Up to 8 weeks	1wk - 2mo	41-68	1700-5300
Up to 20 weeks	2-5 mo	33-58	1500-5000
Up to 40 weeks	5-9 mo	33-58	1400-5100
Up to 1 year	9-15 mo	31-54	1000-4600
After 1 up to 2 years	15-24 mo	25-50	900-5500
After 2 up to 5 years	2-5 y	23-48	500-2400
After 5 up to 10 years	5-10 y	27-53	300-2000
After 10 up to 16 years	10-16 y	25-48	400-2100
Adult	Adults	31-59	300-1400

CD3+CD8+

Website / LIMS age band	Source age band	%	Absolute Count (µL)
Up to 7 days	Neonates	10-41	200-1900
Up to 8 weeks	1wk - 2mo	9-23	400-1700
Up to 20 weeks	2-5 mo	11-25	500-1600

Up to 40 weeks	5-9 mo	13-26	600-2200
Up to 1 year	9-15 mo	12-28	400-2100
After 1 up to 2 years	15-24 mo	11-32	400-2300
After 2 up to 5 years	2-5 y	14-33	300-1600
After 5 up to 10 years	5-10 y	19-34	300-1800
After 10 up to 16 years	10-16 y	9-35	200-1200
Adult	Adults	12-36	200-900

CD19+

Website / LIMS age band	Source age band	%	Absolute Count (µL)
Up to 7 days	Neonates	5-22	40-1100
Up to 8 weeks	1wk - 2mo	4-26	600-1900
Up to 20 weeks	2-5 mo	14-39	600-3000
Up to 40 weeks	5-9 mo	13-35	700-2500
Up to 1 year	9-15 mo	15-39	600-2700
After 1 up to 2 years	15-24 mo	17-41	600-3100
After 2 up to 5 years	2-5 y	14-44	200-2100
After 5 up to 10 years	5-10 y	10-31	200-1600
After 10 up to 16 years	10-16 y	8-24	200-600
Adult	Adults	3-25	100-500

CD3-CD16/56+

Website / LIMS age band	Source age band	% interval	Absolute Count (µL)
Up to 7 days	Neonates	6-58	100-1900
Up to 8 weeks	1wk - 2mo	3-23	200-1400
Up to 20 weeks	2-5 mo	2-14	100-1300
Up to 40 weeks	5-9 mo	2-13	100-1000
Up to 1 year	9-15 mo	3-17	200-1200
After 1 up to 2 years	15-24 mo	3-16	100-1400
After 2 up to 5 years	2-5 y	4-23	100-1000
After 5 up to 10 years	5-10 y	4-26	90-900
After 10 up to 16 years	10-16 y	6-27	70-1200
Adult	Adults	1-19	90-600

T4:T8 Ratio

Age band	Ratio interval	Units
Adult	0.92-3.72	Ratio