Great Ormond Street Hospital for Children NHS Foundation Trust

IMMUNOLOGY LABORATORY CAMELIA BOTNAR LABS GOSH

USER MANUAL

October 2018
ICL 001

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Master copy held on Qpulse (Immunology Laboratory, Camelia Botnar Labs)

General Information

Address:

Clinical Immunology Laboratory
Level 4 Camelia Botnar Laboratories
Great Ormond Street Hospital for Children NHS Trust
Great Ormond Street
London
WC1N 3JH
UK

Hayes DX Box no: DX6640202

Key Contacts

	Phone	Email	Other
Enquiries	020 7829 8835	immunology@gosh.nhs.uk *	
Fax	020 7813 8268		
Clinical Lead/Consultant Clinical Scientist	020 7813 8466	Kimberly.Gilmour@gosh.nhs.uk	
Consultant Immunologist	020 7829 7820	Matthew.Buckland@gosh.nhs.uk	
Lab Manager	020 7813 8194	Fariba.Tahami@gosh.nhs.uk	
CTL Manager	020 7405 9200 x 0433 020 7813 8194	Jesmina.james@gosh.nhs.uk	
Hospital Switchboard	020 7405 9200		

*note: this email address is checked frequently during the day but **should not** be used for urgent queries or requests.

Laboratory hours

9:00 am - 5:30 pm Monday to Friday
No out of hours service: for non-urgent queries leave message on voicemail
Clinical advice available out of hours via switchboard (see below)

Range of tests

The Clinical Immunology Laboratory at GOSH provides a specialist immunology service with an emphasis on primary immunodeficiency and paediatric testing. It also provides a comprehensive range of investigations to assist clinicians caring for children with autoimmune, rheumatological, renal, allergic and gastrointestinal disorders. The Cell Therapy Laboratory (CTL), part of Immunology, undertakes manipulation of blood and marrow for transplantation and cell therapy.

Research Tests/Assays in development

The laboratory has an active programme of assay development, so new tests may be available that are not detailed in this guide. Full details of available tests can be discussed with the Consultant, Clinical scientist or scientific staff of the laboratory.

Clinical, requesting and interpretative advice

Within laboratory hours: Via the immunology laboratory or laboratory consultant or clinical scientist

Out of Hours: Via the immunology registrar (bleep via switchboard on 020 7405 9200)

Labelling and Requesting

Labelling samples

All samples must be taken and labelled in accordance with the Clinical Practice Guideline: Blood Tests: requesting, labelling and sampling requirements available on the intranet.

Samples must be clearly labelled AT THE BEDSIDE USING INFORMATION FROM THE PATIENT'S WRISTBAND with surname, first name, hospital no, date of birth, location and date of collection. Patient labels may be used.

Unlabelled samples will not be processed. The doctor or ward requesting the test will be informed by telephone that a new sample is required and an incident report submitted.

Request forms

Requesting is via PIMS for inpatients / outpatients at GOSH.

Samples from parents/siblings should be registered by the requester and a PIMs request generated form. Where this is not possible a "downtime" form must be used. **Under no circumstances should an amended patient form/sticker be used.**

External referrals require fully completed request forms with patient's surname, forename, DOB, contact details, requesting clinician, location address for results and invoice, referring hospital no and relevant clinical details.

External Molecular Immunology referrals must complete a molecular immunology referral form available from the laboratory or the internet (http://www.labs.gosh.nhs.uk/laboratory-services/immunology)

Control Bloods

Fresh control blood is required for a number of tests in the laboratory. These include functional tests and molecular tests and some phenotypic analysis. If a control sample is required this is clearly indicated on the PIMS form. It is the responsibility of the requesting clinician to ensure that the laboratory is supplied with control blood. As several tests may be being done on one day, the laboratory may ask you to take extra control blood to save more than one individual being bled. Further details of suitable controls and an information sheet for controls are available from the Immunology Laboratory. **If the correct control samples are not received, the laboratory will be unable to process the request.**

Urgent tests

Tests will not be processed urgently without prior direct discussion with Laboratory Clinical Lead. All requests for urgent tests to be performed must be referred to the Clinical Lead. If the Clinical Lead is unavailable, consult with the laboratory manager or senior healthcare scientist.

Referred tests

Samples to be sent away are separated, stored frozen and sent to the appropriate hospital every Monday (or Tuesday if Monday is a bank holiday) and Thursday. If a more urgent result is required, please contact the laboratory. Calprotectins are send daily Monday-Thursday.

Specimen Collection and Transport to the laboratory

Internal

Samples may be sent via the chute system (041) or portering system.
Neither of these systems is infallible, and late or urgent samples must be taken to the lab by hand. Molecular, phenotypic and functional assays must not be sent via the porters (use chute or deliver directly).

If a patient is sent away to be bled by their local hospital or to take a stool sample at home, these should be brought to the Outpatients department where sample labelling can be checked before sending to the Immunology laboratory as above. Samples should not be sent by post directly to the laboratory as this does not conform with the transport of infectious substances regulations.

External

Samples for ***serological*** tests may be sent via first class post or Hays DX (our DX number is: DX6640202)

Samples for some ***functional assays (eg neutrophil function assays and NBT)*** must be sent as urgent samples at room temperature via a courier to ensure that they arrive in time to be tested on the day of collection. Most samples for ***Molecular Immunology***, ***cell proliferation e.g. PHA*** and ***immunophenotyping*** must arrive within 24 hours and be sent at room temperature. The laboratory must be alerted at least 24 hours before that the sample is arriving. Some tests may not be available every day. Further detail of sending requirements is available from the laboratory.

Samples must be placed separately from the request forms in sealable plastic bags. Samples that have to be posted or couriered must be packaged according to transport of infectious substances regulations.

Blood Volumes and helpful tips

Correct blood volumes and bottles for GOSH patients are on the requesting system.

If a group of tests requiring the same bottle type are requested, it may be possible to reduce the volume of blood required or put all the blood into one tube. Please contact the lab for further details.

Many tests must be booked in advance with the laboratory or are not available on certain days. The lab may be unable to process the sample if the guidance given for each test is not adhered to.

Results

- Results are authorised daily.
- For internal users, authorised results are available via the GOSH web (Path results).
- For external requests, a printed report is sent to the requesting clinician. Web access may be requested (see http://www.labs.gosh.nhs.uk) so identified individuals can view results via a secure log-in.
- Results are not generally available over the phone unless urgent.
- Results can be emailed to nhs.net accounts, internal emails, or faxed to a secure fax in exceptional circumstances
- Unauthorised urgent results will only be issued to Consultant Immunology or Infectious diseases staff, or with the agreement of the laboratory clinical lead or her deputy.
- All results are issued with an interpretive comment, and normal ranges are printed on the reports where appropriate. For quantitative assays numerical reference ranges are provided in the user manual. For other assays including all specialist assays an interpretive comment is written on the report.

- Any tests without quoted reference ranges will have an individual comment added.
- Further advice on result interpretation is available from the laboratory if required
- Upon request, the laboratory can supply users with estimates of uncertainty.

Requesting additional tests and sample retention

If the sample is still available and sufficient in volume and quantity, additional tests may be requested by telephoning or emailing the laboratory. Note: requesters must send another request form, with details of the additional test required.

If the exact serological tests that will be required are not known at the time of sampling, request "save serum". Serum will be stored for 2-3 months. When tests are wanted, a new request form with test details and date of stored sample should be sent to the laboratory. Requests for 'save serum' from Immunology and Infectious diseases consultant's will be stored long term.

Lithium heparin and EDTA samples are viable for 1-2 days (depending on test required) and are discarded at the end of each week.

Serum samples are frozen for 2-3 months and discarded.

Patients on replacement immunoglobulin therapy should have serum and plasma stored before the start of treatment, then annually or at change of product ("Store Ig serum plasma" on PIMS). This is stored long term.

Peripheral blood mononuclear cells can also be saved in liquid nitrogen to enable future investigation of undefined immune deficiency (after discussion with the Consultant or Clinical Scientist). Request 'save cells in liquid nitrogen' on PIMS. High risk samples cannot be stored by the laboratory.

Test request management

A system for request management of tests is in place in the Immunology department. This is to stop duplication, unnecessary repeated testing, and other inappropriate requests. These tests are booked on Omni, and samples are kept for a period of time. The affected clinicians are contacted via Omni and email. This is to provide an opportunity for further discussion, and the possibility for unblocking of the tests when necessary. The frequency of testing for Immunology assays is listed in appendix 5.

The Immunology laboratory strives to ensure that all service user information provided through verbal and written communication including this user guide is relevant and up to date. In order to utilise our services in the most effective and efficient manner, users should not hesitate to contact the laboratory with any queries regarding the service provided.

Complaints

Any complaints regarding the service provided by the Immunology Laboratory should be directed to the Laboratory Manager and should follow the procedure laid out in the GOSH Complaints Policy (POL 10024).

Information Governance

The Immunology Department complies with all relevant professional, national and local guidance on confidentiality and data protection including but not limited to the GOSH Confidentiality Policy, Information Governance Policy, the Caldicott Guardian manual and the Data Protection Act 1998.

Addresses of referral labs

ADDENBROOKES Clinical Immunology Level E4 Addenbrooke's Hospital Hills Road Cambridge CB2 2QQ	CHURCHILL OXFORD Department of Immunology Churchill Hospital Old Road, Headington Oxford OX3 7LJ
CHARING CROSS Specimen Reception 1st Floor Lab Block Charing Cross Hospital Fulham Palace Road London W6 8RF	JOHN RADCLIFFE OXFORD Neuroscience Group Institute of Molecular Medicine John Radcliffe Hospital Oxford OX3 9DU
ROYAL FREE/ HSL Autoimmune Serology Lab Clinical Immunology 2nd Floor Royal Free Hospital Pond Street London NW3 2QG	QUEENS SQUARE Dept. of Neuroimmunology The National Hospital For Neurology and Neurosurgery Institute of Neurology Queen Square London WC1N 3BG
SHEFFIELD PRU Northern General Hospital Sheffield S5 7YT	ROYAL LONDON Immunopathology Department Barts and the London NHS Trust 2nd floor, Pathology and Pharmacy Building, 80 Newark Street London E1 2ES
Meningococcal Reference Unit Manchester Medical Microbiology Partnership PO Box 209 Clinical Sciences Building Manchester Royal Infirmary Manchester M13 9WZ DX6962410 Manchester 90M	University Hospital of Wales Department of Medical Biochemistry & Immunology Heath Park Cardiff CF14 4TW
PHE Respiratory & Systemic Infection Laboratory Colindale 61 Colindale avenue London NW9 5HT	Royal Devon & Exeter NHS Foundation Trust Clinical Biochemistry Barrack Road Exeter EX2 5DW DX6461102 Exeter92
Anthony Nolan Histocompatibility Laboratories Royal Free Hospital Pond Street	Royal United Hospital Immunology Dept. (B.I.R.D diagnostics) Level 1 entrance B38 Combe Park

Hampstead London NW3 2QG	Bath BA1 3NG

List of tests by type

Serological tests

Test	PIMS	Sample bottle	Minimum Volume	Sent to (in house if not indicated)	Completion time (days)	Repeat request	Reference Range	Comments
Humoral Immunity								
Immunoglobulins IgG, IgA, IgM	Immunoglobulins IgG, IgA, IgM Request under Chem Path specialised	Serum/ serum gel	1 ml	Chem Path GOSH	5	See Appendix 5	Age specific See Appendix 1	IgG may be lost via renal tract, gut etc. Check albumin is normal before interpreting IgG Note: test run by Chemical Pathology, clinical interpretation and advice from Immunology
IgG subclasses	IgG subclasses	Serum/ serum gel	2 ml	PRU, Sheffield	21	See Appendix 5	Age specific See Appendix 1	Will always be low if total IgG low. Will not routinely be done if patient < 2y of age, or with normal total IgG and IgA.
IgD	IgD	Serum/ serum gel	2 ml	PRU Sheffield	30	See Appendix 5	2-100 KU/L	May be non-specifically raised in a variety of inflammatory conditions. Not specific to hyper IgD syndrome
Tetanus antibodies	Tetanus antibodies	Serum/ serum gel	2 ml	Sheffield	21	See Appendix 5	>0.1 iu/ml	If low, a booster vaccine should be given and retest 4-6 weeks later
Pneumococcal abs (serotype specific)	Pneumococcal abs	Serum/ serum gel	0.5 ml	Addenbrookes	28	See Appendix 5	Putative protectivelevel 0.35 ug/ml, optimal level >0.5ug/ml	7 serotypes contained in Prevenar and 5 non-vaccine serotypes measured. Results must be interpreted with vaccine and exposure history.
Meningococcal W and Y		Serum/ serum gel	1 ml	Manchester	30	See Appendix 5	See comment on report	
Allergy								
IgE	IgE	Serum/ serum gel	1 ml		8	See Appendix 5	Age specific See Appendix 2	IgE may be elevated in many atopic and other conditions. Normal total IgE does not exclude allergy.
Specific IgE (please state allergens required in reason for request) (for panels see appendix 2)	Specific IgE	Serum/ serum gel	1 ml initially then 0.5 ml per allergen		10	See Appendix 5	See appendix 2 and comments on reports	Allergens should be selected on the basis of history. There is a significant false positive rate. Further advice on interpretation can be obtained from the laboratory. Level of IgE that is significant varies with allergen. Further advice available from the laboratory. A total IgE >5000 may interfere with the assay and reduces the specificity of the test
Mast cell tryptase	Tryptase	Serum/ serum gel	1 ml	Royal Free-HSL	21	See Appendix 5	2-11.4 mg/l	Raised after anaphylaxis. Samples must be taken at the time of the reaction, at 4-6 hours and 24 hours to allow interpretation. Include time of reaction and samples on form.

	Complement							
Complement C3 + C4	C3 C4 complement Request under Chem Path specialised	Serum/ serum gel	1 ml	Chem Path GOSH	5	See Appendix 5	C3 0.75-1.65g/l C4 0.14-0.54g/l	Consumed during sepsis. If low in this context should be rechecked. Low in SLE. Note: test run by Chemical Pathology, clinical interpretation and advice from Immunology
Functional Complement: classical */**	Functional Complement	Serum/ serum gel	1 ml		30	See Appendix 5	>40%	Measures the function of the classical (C1,2,4) and terminal complement pathways. If abnormal, recheck. Alternative pathway function will be measured in parallel with this test.
Functional Complement: Alternative */**	Functional Complement	Serum/ serum gel	1 ml		30	See Appendix 5	>10%	Measures the function of the alternative (C3, B, D, properdin) and terminal complement pathways. If abnormal, recheck. Classical pathway function will be measured in parallel with this test.
Factor H	Factor H	Serum/ serum gel	1 ml	Charing Cross	70	See Appendix 5	See Interpretative comment	Mutations in Factor H, with or without low protein, are associated with familial (D-) HUS
Factor I	Factor I	Serum/ serum gel	1 ml	Charing Cross	70	See Appendix 5	See Interpretative Comment	Deficiency associated with recurrent bacterial infections or occasionally with autoimmune disease (haemolytic anaemia, renal). AP50 will be low.
C3 Nephritic factor	C3 Nephritic factor	Serum/ serum gel	2 ml	PRU Sheffield	21	See Appendix 5	See Interpretative Comment	Stabilises the alternative pathway C3 convertase. Results in a low C3 and seen in membranoproliferative glomerulonephritis and partial lipodystrophy. Only done if C3 low.
C1q levels	C1q levels	Serum/ serum gel	2 ml	PRU Sheffield	60	See Appendix 5	50-250 mg/l	Classical pathway component. Deficiency gives autoimmune disease/recurrent infection. Only measure if CH100 low and AP50 normal. Other complement component measurements available on request
C2 levels	C2 complement	Serum/ serum gel	2 ml	PRU Sheffield	60	See Appendix 5	10-80mg/l	Classical pathway component. Deficiency gives autoimmune disease/recurrent infection. Only measure if CH100 low and AP50 normal. Other complement component measurements available on request
C1 Esterase inhibitor	C1 Esterase inhibitor	Serum/ serum gel	1 ml	Royal London	30	See Appendix 5	Level:150-350 mg/l Function: >84%	Low levels (85%) or non-functional protein (15%) are associated with hereditary angioedema. C4 low in patients during, and normally between attacks.

Mannose binding lectin *	MBL	Serum/ serum gel	1 ml		22	See Appendix 5	See appendix 3	Low levels correlate with polymorphisms. Increased risk/severity of infections.
	Autoimmunity *Note: a variety of other autoantibodies may be available on request. Please discuss with the laboratory before sending samples* *Note: for neuronal autoantibodies CSF may only occasionally provide additional information and many assays are not validated on CSF. Please discuss with the laboratory before sending CSF.*							
Autoimmune profile **(NB: does not include ANA) ***	Autoantibody screen	Serum/ serum gel	0.5 ml		8	See Appendix 5	Reported as positive/ negative	Detects the following antibodies: **Smooth muscle** (SM): often a non-specific finding. Occurs in type I autoimmune hepatitis (along with ANA). Also found in some patients with Hepatitis A and C infections. **Liver kidney microsomal** (LKM): found in Type 2 autoimmune hepatitis. Also occurs in Hepatitis C. **Mitochondrial** (MITO) : found in primary biliary cirrhosis (a rare disease in children) **Reticulin:** false positives frequently found. Positive in coeliac disease with low specificity and sensitivity **Gastric parietal cell** (GPC): found in pernicious anaemia. This is very rare in children
Anti nuclear antigen *	ANA/dsDNA/ENA	Serum/ serum gel	1 ml		8	See Appendix 5	Staining patterns are reported with titre. Positive >/=1:160 Strong positive >1:640	Titred 1:160 to 1:2560. May be non-specifically raised in infection. If positive, laboratory will automatically proceed to check antibodies to dsDNA (>1:160) and ENAs (>1:320)
Double stranded DNA antibodies *	ANA/dsDNA/ENA	Serum/ serum gel	ANA sample		8	See Appendix 5	Neg <10.0 IU/ml Equivocal 10-15 IU/ml Positive >15 IU/ml	Only performed if ANA positive. Levels of dsDNA antibody may correlate with disease activity in SLE.
Extractable nuclear antigens *	ANA/dsDNA/ENA	Serum/ serum gel	ANA sample	In house/ Royal London	Screen: 8 Type: 28	See Appendix 5	Reported as positive/ Negative	If ANA positive ENA screen will be done: Includes Ro, La, Sm, RNP, Jo-1, Scl-70, centromere. If screen positive, sample will be sent to Royal Free for typing. Not quantitative. ENAs may evolve during the course of paediatric connective tissue disease.
Anti C1q antibodies	Anti C1q antibodies	Serum/ serum gel	1 ml	Sheffield	60	See Appendix 5	0-15 U/ml	Described in hypocomplementaemic urticarial vasculitis, rheumatoid vasculitis, SLE (paed and adult), mesangiocapillary glomerulonephritis and IgA nephropathy.

Rheumatoid factor (RF)	Rheumatoid factor	Serum/ serum gel	1 ml	Royal Free-HSL	21	See Appendix 5	< 20 IU/ml negative >20 positive	Testing for rheumatoid factor is a poor screening procedure for juvenile idiopathic arthritis and of supportive diagnostic value only in the highly restricted population of older children with polyarticular arthritis (positive in up to 33%). Poor prognostic feature. Levels do not correlate with disease activity. Also found in many other connective tissue and inflammatory diseases.
Coeliac Screen *	Coeliac Screen (ttg)	Serum/ serum gel	0.5 ml		7	See Appendix 5	<7 IU/ml negative 7-10 IU/ml equivocal >10 IU/ml positive	IgA antibodies to tissue transglutaminase. If positive, endomysial antibodies are automatically done by lab to confirm. High positive predictive value for coeliac disease. May be falsely negative if IgA very low (included in test).
GAD antibodies	GAD antibodies	Serum/ serum gel	1 ml	John Radcliffe Oxford	28	See Appendix 5	0 – 5.0 U/ml neg 5.1-30 equivocal >30 positive	Antibodies may be positive pre/ early IDDM
Thyroid antibodies	Thyroid antibodies	Serum/ serum gel	0.5 ml		8	See Appendix 5	<60 Negative 60-100 Equivocal >100 positive	Presence consistent with autoimmune thyroid disease and indicates the need to monitor thyroid function. Not specific for any one thyroid disease.
TSHR antibodies	TSHR ab	Serum/ serum gel	2 ml	Sheffield	40	See Appendix 5	0 – 0.9 IU/ml	Antibodies may be an additional finding in patients where thyroglobulin abs neg but autoimmune thyroid disease possible. Only available in selected cases after discussion with lab
Glomerular basement membrane (GBM) antibodies *	Renal (GBM) antibodies	Serum/ serum gel	0.5 ml		8	See Appendix 5	<7 negative 7-10 equivocal >10 positive	Goodpastures syndrome. Patients with Wegner's may have a similar renal presentation (rapidly progressive glomerulonephritis) and ANCA should also be done. If urgent contact lab.
ANCA (anti neutrophil cytoplasmic abs) *	anti neutrophil cytoplasmic abs	Serum/ serum gel	0.5 ml		8	See Appendix 5	MPO: <3.5 negative 3.5-5.0 equivocal >5.0 positive PR3 <2.0 negative 2.0-3.0 equivocal >3.0 positive	Positive in Wegners and also other vasculitides. PR-3 levels may be a useful indicator of disease activity.
Anti ganglioside antibodies (includes GM1, GD1a, GT1b, GQ1b and sulphatides)	Anti ganglioside abs	Serum/ serum gel	0.5 ml	National Hospital Queens square	42	See Appendix 5	Reported as positive/ Negative	Found in a low proportion of patients with Guillain Barre syndrome, chronic demyelinating polyneuropathy and multifocal motor neuropathy. Because antibody incidence is low, these tests should not be a routine part of diagnosis.
Anti Acetylcholine receptor (ACHR)	Anti Acetylcholine receptor (ACHR)	Serum/ serum gel	1 ml	John Radcliffe Oxford	20	See Appendix 5	0-5	Occur in myasthenia gravis. The assay will detect antibodies to the foetal cholinesterase receptor. Musk antibodies also available on request (if AchR negative)

Adrenal Cortex Antibodies	Adrenal Cortex Antibodies	Serum/ serum gel	0.5 ml	Royal London	28	See Appendix 5	Reported as positive/ Negative	Positive in 50% of Addisons where there are other AI disease, less if adrenal disease occurs alone. Antibodies cross react with the steroid-producing cells in the testis and ovary
Insulin antibodies	Insulin antibodies	Serum/ serum gel	2 ml	Sheffield	28	See Appendix 5	0-5 mg/L	May occur in insulin resistance.
Cardiac muscle antibodies	Antimyocardial antibodies	Serum/ serum gel	1 ml	Royal London	28	See Appendix 5	Reported as positive/ Negative	Described in myocarditis, idiopathic dilated cardiomyopathy, rheumatic carditis and Dresslers syndrome.
Basal ganglia antibodies	Basal ganglia antibodies	Serum/ serum gel	0.5 ml	National Hospital Queens square	20	See Appendix 5	Reported as positive/ Negative	Found in a proportion of children with post-streptococcal movement disorders. Clinical utility still being evaluated.
Interferon beta neutralising antibodies	B interferon neut.abs	Serum/ serum gel	0.5 ml	National Hospital Queens square	21	See Appendix 5	<20 NU Negative 20 – 100 NU Low positive 100 – 600 NU Positive >600 NU High positive	May be found in patients who have become resistant to treatment with IFNb for MS
Aquaporin 4 antibodies	Aquaporin 4 antibodies	Serum/ serum gel	1 ml	Oxford	30	See Appendix 5	Reported as positive/ Negative	May be found in patients with neuromylitis optica.
Voltage gated potassium channel antibodies	VGKC abs	Serum/ serum gel	1 ml	Oxford	40	See Appendix 5	0 - 100 pM	Associated with limbic encephalitis, described in the literature as amenable to immune modulation
Voltage gated calcium channel antibodies		Serum/ serum gel	1 ml	Oxford	40	See Appendix 5	0 - 45 pM	May be found in Lambert-Eaton myaesthenic syndrome
MUSK antibodies	MUSK Abs	Serum/ serum gel	1 ml	Oxford	40	See Appendix 5	Reported as positive/ Negative	May be found in AChR negative myasthenia gravis
NMDA receptor antibodies	NMDA rec. antibodies	Serum/ serum gel	1 ml	Oxford	40	See Appendix 5	Reported as positive/ Negative	May be found in autoimmune encephalitis, typically in children and young people with prominent psychotic symptoms.
Paraneoplastic antibodies	Paraneoplastic antibodies	Serum/ serum gel	1ml	Oxford	40	See Appendix 5	Reported as positive/ negative	Screen includes anti yo, Hu, Ri,MA1, MA2, CV2, CRMP%, amphiphylin Detected in a variety of autoimmune neurological conditions, originally described as paraneoplastic in adults.
Myositis Profile	Myositis Profile	Serum/ serum gel	1ml	Royal United Bath Hospital	21	See Appendix 5	Reported as negative/positive for named ENA.	Looks for ENAs associated with myositis
MISC								

Calprotectin	Calprotectin	Faeces	50p size (apricot size)	Charing Cross	28	See Appendix 5	< 50.0 mg/kg	High levels may help in discrimination of inflammatory bowel disease from other GI symptoms If patient is collecting sample at home inform patient to keep sample refrigerated.
Myeloid Related Protein (MRP 8 and 14)	MRP	Serum/ Serum gel	1 ml		28	See Appendix 5	See appendix 4	MRP 8 and 14 are endogenous TLR-4 ligands that are expressed in granulocytes and monocytes and promote inflammatory processes in vivo. Used to predicate response to MTX or relapse when withdrawn for JIA patients
Beta 2 microglobulin	Beta 2 microglobulin	Serum/ Serum gel	1 ml	Royal Free	21	See Appendix 5	1.16 – 3.21	
Soluble CD25 *	Soluble CD25	Serum/ Serum gel	1ml		21	See Appendix 5	<2500 pg/ml	May be raised in HLH, declining with response to treatment.
Infiximab Levels and antibodies	Infliximab	Serum/ Serum gel	1ml	Royal Devon and Exeter	14	See Appendix 5	Levels reported in mg/L. Antibody reported as positive/negative	Monitors drug levels and presence of antibodies
Adalimumab Levels and antibodies	Adalimmumab	Serum/ Serum gel	1ml	Royal Devon and Exeter	14	See Appendix 5	Levels reported in mg/L. Antibody reported as positive/negative	Monitors drug levels and presence of antibodies
HLA typing B27	HLA typing B27	EDTA	4mls	Anthony Nolan, Royal Free Hospital	21	See Appendix 5	Reported as positive/ negative	HLA B27 is strongly associated with ankylosing spondylitis.
HLA typing DQ2 and DQ8	HLA typing DQ2 and DQ8	EDTA	4mls	Anthony Nolan, Royal Free Hospital	21	See Appendix 5	Reported as positive/ negative	HLA DQ2 and DQ8 are associated with coeliac disease and Type 1 diabetes.
Biomarkers	Biomarkers	Serum, specimens containing sodium azide should not be used.	1ml		93	See Appendix 5	<200 pg/ml	Soluble Fas Ligand is measured; this is part of the FAS/FASL apoptosis cascade. 85% of patients with elevated sFAS and VitB12 have FAS mutations.
Cytokines	Immunology cytokines	EDTA/ Serum	2ml		56	See Appendix 5	<50 pg/ml	Raised in inflammatory conditions and in patients undergoing CAR therapy. Please call lab if urgent.

- Some serological assays (denoted by *) may be affected by icteric/lipaemic/haemolysed samples. These should be avoided if possible.
- Heat inactivated samples are unsuitable for assays denoted by **.

Cell Phenotypic Analysis

- **All tests must be booked with the laboratory (x8835). The laboratory may not be able to process unbooked samples due to time and staffing constraints.**
- Tests should only be requested after discussion with an Immunology or Infectious diseases Consultant or Registrar or as part of a defined disease/ therapy monitoring protocol.
- Lymphocyte tests can be kept for 24 hours (e.g. can be taken on Monday for Tuesday analysis).
- Most inpatients should be tested Mon-Thursday
- High risk (HIV) inpatients and outpatients should be tested on Fridays
- Tests are run on Tuesday, Thursday and Friday. Samples ideally taken Mon/Weds for Tues/ Thurs testing.
- Samples must be in the lab by 2pm on the day of testing
- A full blood count MUST be taken the same day to allow enumeration and full evaluation of lymphocytes.
- Interpretative comments are provided with all results. There are limited reference ranges available for children. Interpretation is based on those in Comans-Bitter et al (1997) J Paediatrics 130 (3): 388.
- Routine panels are listed below; additional tests may be available on a research basis after discussion with the consultant or clinical scientist.

All tests on PIMS are listed under *Lymphocyte subsets*:

PIMS	Sample bottle	Volume	Completion time (days)	Comments
Basic	EDTA	0.5 ml	3	T4/8, B + NK
MEM	EDTA	0.5 ml	3	Memory T cells Naïve cells CD45RA+CD27+ Memory cells CD45RA-CD27+ Effector cells CD45RA+CD27-
TCT	EDTA	0.5 ml	4	T Cell Types Gamma delta T cells: may be raised in infection, autoimmunity, PID and may fail to express CD4 or CD8. Double negative T cells Increased numbers of CD3 positive, CD4 and CD8 negative T cells are seen in the peripheral circulation in patients with ALPS, but are also found in other inflammatory and immune dysregulated states. This is a screening test for ALPS
ACT	EDTA	0.5 ml	3	CD38 is a marker of early activation and in studies has been shown to be useful in monitoring HIV patients starting to fail therapy prior to observing decreased CD4 counts. DR (MHC II) is a useful marker of activation. MHC II deficient SCIDs may be CD4 lymphopaenic
CD25	EDTA	0.5 ml	3	CD25 is upregulated on activated CD4 positive T cells. Rare deficiency of this molecule also described
MHCI *plus a Healthy Control sample	EDTA	0.5 ml	4	Rare deficiency of this molecule described
Neutrophil *plus a Healthy Control sample	EDTA	0.5ml	4	CD11a, 18. CD18 is beta2 integrin which is abnormal in leucocyte adhesion deficiency type I. CD11a binds to CD18 in the membrane, and therefore will also not be expressed.
XBMEM	EDTA	1 ml	10	EUROCLASS method. Assesses proportion of naïve, marginal zone, switched memory and transitional B cells and plasmablasts. Abnormalities have been defined in patients with CVID and other immune deficiencies.
CD19/20	EDTA	0.5 ml	3	Routine B cell marker is CD19. Patients may fail rituximab (anti-CD20 antibody) treatment if they fail to express CD20. Lack of CD19 (but presence of CD20) is a cause of CVID.
V beta repertoire	EDTA	2 mls	8	Looks at surface expression of T cell receptor V beta families. In normal individuals at least 70 % of their T cells will be detected with the panel of antibodies used, and there will be a polyclonal distribution. Oligoclonal expansions on a polyclonal background are suggestive of infection or malignancy, whilst an oligoclonal distribution is seen in defects of T cell development.
ThyE	EDTA	0.5 ml	10	Looks for CD31 positive cells, which are a surrogate marker of thymic emigrants.
Treg	EDTA	0.5 ml	10	Regulatory T cells are defined as CD4+/CD25+/Cd127low/FoxP3+. This panel detects cells that are CD4+/CD25+/CD127low and are most likely T regulatory cells.

Lymphocyte proliferation

- **All tests must be booked with the laboratory (x8835). The laboratory may not be able to process unbooked samples due to time and staffing constraints.**
- Tests should only be requested after discussion with an Immunology or Infectious diseases Consultant or Registrar or as part of a defined disease/ therapy monitoring protocol.
- Lymphocyte proliferation tests can be kept for 24 hours at room temperature (eg can be taken on Monday for Tuesday analysis).Most inpatients should be tested Mon-Thursday
- High risk (HIV, Hep B, C) patients cannot be tested
- Tests are run on Tuesday, Thursday and Friday afternoons
- Samples must be in the lab by 2pm on the day of testing
- Interpretative comments are provided with all results.
- Routine panels are listed below; additional tests may be available on a research basis after discussion with the consultant or clinical scientist.
- In the week preceding a bank holiday, please check with the lab if they can perform proliferation assays

Test	Sample bottle	Volume	Completion time (days)	Comments
Whole Blood				
PHA stimulation *plus a Healthy Control sample	Lithium heparin	0.5 ml	14	May be abnormal if lymphocyte count low. Consider PHA: separated cells
Separated cells (peripheral blood mononuclear cells)				
CD3 stimulation *plus a Healthy Control sample	Lithium heparin	5 ml	14	
PHA (PHA: separated cells) *plus a Healthy Control sample	Lithium heparin	5 ml	14	
PHA +/- IL2 *plus a Healthy Control sample	Lithium heparin	5 ml	14	Synergy is normally seen between PHA and IL2. this may be lacking in patients with IL2 receptor/function defects
Candida stimulation *plus a Healthy Control sample	Lithium heparin	5 ml	14	May be low if the patient has not had thrush (i.e. not encountered the antigen)
Mixed lymphocyte culture *plus a Healthy Control sample	Lithium heparin	5-10 ml from patient and donor(s) (depends on the no. of donors to be tested)	14	Only undertaken to asses suitability of family donors for HSCT or 3rd party immune function.

Save Cells	Lithium heparin / EDTA	5 - 10 ml	10	Sample will depend on future tests required. Sample is ficolled and PBMCs frozen in Liquid Nitrogen. If sample is to be stored for subsequent therapeutic use, please book with lab well in advance as processing requirements different.

Neutrophil Function Tests

All tests must be booked with the laboratory (x8835). The laboratory may not be able to process unbooked samples due to time and staffing constraints.

- Tests should only be requested after discussion with an Immunology or Infectious diseases Consultant or Registrar or as part of a defined disease/ therapy monitoring protocol.
- Samples must be in the lab by 2pm on the day of testing and must have been taken within 8 hours
- Interpretative comments are provided with all results.
- Routine tests are listed below; additional tests may be available on a research basis after discussion with the consultant or clinical scientist.

Test	Sample bottle	Volume	Completion time (days)	Comments
NBT (nitrobluetetrazolium test) *plus a Healthy Control sample	EDTA	1 ml	3	Excludes all common forms of CGD
DHR (dihydrorhodamine) *plus a Healthy Control sample	LiHep	1 ml	8	May be abnormal in CGD, atypical forms of CGD and MPO deficiency.
Neutrophil phagocytosis *plus a Healthy Control sample	LiHep	1 ml	8	Tests the ability of neutrophils to phagocytose opsonised material over a time course
CD62L shedding *plus a Healthy Control sample	LiHep	1 ml	8	Will be abnormal in patients with IRAK4 deficiency or other defects in this pathway

Protein and Functional tests for Primary Immunodeficiency (Molecular Immunology)

- GOS hosts a national diagnostic centre for the molecular diagnosis of primary immune deficiency. This is run by the immunology and molecular genetics laboratories. Protein or functional testing is undertaken on patients, and genetic analysis then performed on patients with abnormal protein results or a very strong clinical history.
- These tests should only be undertaken under the direction of a Consultant Immunologist/ Senior Clinical Scientist. External referrals must be discussed with Dr Kimberly Gilmour (020 7813 8466) or her deputy and accompanied by a completed referral form (http://www.labs.gosh.nhs.uk/laboratory-services/immunology).
- The table below indicates when a separate EDTA sample needs to be sent for DNA storage and future analysis (part of the original sample can sometimes be used for this purpose). Sample volumes are minimum requirements, if the patient is lymphopaenic, an increased volume will be required.
- All of these tests require a control blood to be sent at the same time. **It is the responsibility of the referring clinician to ensure that a control is provided.** If the control is a parent or other family member, please indicate, label appropriately (e.g. mother's name and DOB), and DNA will be stored from them as well as the patient.
- More protein tests are available than are indicated, but these should be discussed on an individual basis. Genetic tests for these rarer conditions are often not available, or on a research basis only. Further details are available from Dr Gilmour.
- All samples must be booked with the Immunology lab at least 24 hrs before sample is sent as staff may not be available to perform assays on some days. The laboratory reserves the right to not analyse unbooked or late samples.
- Additional genetic tests are available for some conditions where protein testing is not available (eg RAG1/2, Artemis)
- 73 immunology genes are screened on a single panel (immunology TIGER). Further details are available from Dr Gilmour or NE Thames Regional genetics service.

Test	Sample bottle	Extra sample for DNA storage?	Volume (minimum)	Completion time (days)*	Comments
WASP *plus a Healthy control sample	EDTA	Yes -1 ml	2 mls	8	Flow cytometry Protein may be absent or reduced in quantity in Wiskott Aldrich syndrome. Small platelets are a consistent feature and platelet volume should also be requested on these patients.
Common gamma chain *plus a Healthy control sample	EDTA	Yes -1 ml	2 mls	8	Flow cytometry Abnormal in X-linked SCID (T-B+NK-). Stat-5 phosphorylation should also be done (see below).
Btk *plus a Healthy control sample	EDTA	Yes -1 ml	2 mls	8	Flow cytometry. Abnormal in X-linked agammaglobulinaemia (hypogammaglobulinaemia with absent B cells) For patients with absent B cells, protein test not done, only

					genetics. For patients with some B cells, protein assay used as a screen.
SAP *plus a Healthy control sample	EDTA	Yes -1 ml	2 mls	8	Flow Cytometry. Abnormal in X-linked lymphoproliferative disease 1.
XIAP *plus a Healthy control sample	EDTA	Yes -1 ml	2 mls	8	Flow Cytometry. Abnormal in X-linked lymphoproliferative disease 2.
CGD proteins: gp91 *plus a Healthy control sample	EDTA	Yes -1 ml	5 mls	8	Flow cytometry. X-linked CGD. The lab will select the proteins tested based on sex, maternal carrier status and family history,
CGD proteins:p47 *plus a Healthy control sample	EDTA	Yes -1 ml	5 mls	8	Flow cytometry. Autosomal recessive form. The lab will select the proteins tested based on sex, maternal carrier status and family history,
CGD proteins: p22, 67 *plus a Healthy control sample	EDTA	Yes -1 ml	10 mls	10	Immunoblot. Autosomal recessive forms. The lab will select the proteins tested based on sex, maternal carrier status and family history,
CD40 Ligand (CD154) *plus a Healthy control sample	Li Hep	Yes -1 ml	1 ml	8	Flow cytometry. Abnormal in X-linked hyperIgM syndrome.
CD40 *plus a Healthy control sample	EDTA	Yes -1 ml	1 ml	8	Research assay. Abnormal in one type of AR-HIM syndrome
Perforin *plus a Healthy control sample	EDTA	Yes -1 ml	2 mls	8	Flow cytometry. Perforin deficiency accounts for 30% of patients with familial HLH Other proteins that may be deficient in HLH are also available on request.
Stat 5 tyrosine phosphorylation *plus a Healthy control sample	EDTA	Yes -1 ml	2 mls	8	Measures signalling via the IL2, 7 or 15 receptors. Will be abnormal if components of this pathway are missing/non-functional (IL2: gamma chain, Jak-3, IL7: IL7Ra). Used as a functional screen of the pathways so that other specific tests can be undertaken after this. Please specify cytokines required
Granule release assay (GRA) *plus a Healthy control sample	EDTA	Yes -1 ml	10 mls	8	Tests the ability of T and NK cells to degranulate in response to a stimulus and express CD107a on their surface. Screens the pathway that includes syntaxin 11 and Munc13-4/18-2, lyst and Rab27a
Molecular Immunology Miscellaneous (specify test in reason for request) *plus a Healthy control sample	EDTA	Yes -1 ml	Varies with test	Varies with test	Requested after discussion with Clinical Lead, laboratory manager or senior healthcare scientist. Reason for request must be stated
NK killing assay *plus a Healthy control sample	Li Hep	Yes -1 ml	10 mls	8	LSS should be requested to ensure the presence of NK cells before assay setup. Low or absent natural killer (NK) cell activity is common prior to and during active disease, as well as after remission following chemotherapy in a significant proportion of individuals with Familial HLH.
Apoptosis functional	LiHep	Yes -1 ml	10 mls	30	Screening assay for defects in the apoptosis pathway (including Fas, FASL,

*plus a Healthy control sample					caspase 8 and caspase 10). Defects in this pathway are associated with ALPS
***If results are required urgently, please liaise with the immunology lab as many tests can be completed same day**					

Alphabetical list of tests

Test	PIMS	Sample Bottle	Minimum Volume	Sent to (in house if not indicated)	Completion time (days)*	Repeat request	Reference Range	Comments
Adalimumab Levels and antibodies	Adalimmumab	Serum/ Serum gel	1ml	Royal Devon and Exeter	14	See Appendix 5	Levels reported in mg/L. Antibody reported as positive/negative	Monitors drug levels
Anti Acetylcholine receptor (ACHR)	Anti Acetylcholine receptor (ACHR)	Serum/ serum gel	1 ml	John Radcliffe Oxford	20	See Appendix 5	0-5	Occur in myasthenia gravis. The assay will detect antibodies to the foetal cholinesterase receptor. Musk antibodies also available on request (if AchR negative)
Adrenal Cortex Antibodies	Adrenal Cortex Antibodies	Serum/ serum gel	1 ml	Royal London	28	See Appendix 5	Reported as positive/ Negative	Positive in 50% of Addisons where there are other AI disease, less if adrenal disease occurs alone. Antibodies cross react with the steroid-producing cells in the testis and ovary

ANCA (anti neutrophil cytoplasmic abs)	anti neutrophil cytoplasmic abs	Serum/ serum gel	0.5 ml		8	See Appendix 5	MPO: <3.5 negative 3.5-5.0 equivocal >5.0 positive PR3 <2.0 negative 2.0-3.0 equivocal >3.0 positive	Positive in Wegners and also other vasculitides. PR-3 levels may be a useful indicator of disease activity.
Anti nuclear antigen	ANA/dsDNA/ENA	Serum/ serum gel	1 ml		8	See Appendix 5	Staining patterns are reported with titre. Positive >/=1:160 Strong positive >1:640	Titred 1:160 to 1:2560. May be non-specifically raised in infection. If positive, laboratory will automatically proceed to check antibodies to dsDNA (>1:160) and ENAs (>1:320)
Apoptosis functional	Apoptosis functional	LiHep	10 ml + DNA smaple		30	See Appendix 5	Interpretative comment provided	Screening assay for defects in the apoptosis pathway (including Fas, FasL, caspase 8 and caspase 10). Defects in this pathway are associated with ALPS
Aquaporin 4 antibodies	Aquaporin 4 antibodies	Serum/ serum gel	1 ml	Oxford	30	See Appendix 5	Reported as positive/ Negative	May be found in patients with neuromylitis optica.
Autoimmune profile **(NB: does not include ANA)**	Autoantibody screen	Serum/ serum gel	0.5 ml		8	See Appendix 5	Reported as positive/ negative	Detects the following antibodies: **Smooth muscle** (SM): often a non-specific finding. Occurs in type I autoimmune hepatitis (along with ANA). Also found in some patients with Hepatitis A and C infections. **Liver kidney microsomal** (LKM): found in Type 2 autoimmune hepatitis. Also occurs in Hepatitis C. **Mitochondrial** (MITO) : found in primary biliary cirrhosis (a rare disease in children) **Reticulin:** false positives frequently found. Positive in coeliac disease with low specificity and sensitivity **Gastric parietal cell** (GPC): found in pernicious anaemia. This is very rare in children

Basal ganglia antibodies	Basal ganglia antibodies	Serum/ serum gel	1 ml	National Hospital Queens square	20	See Appendix 5	Reported as positive/ Negative	Found in a proportion of children with post-streptococcal movement disorders. Clinical utility still being evaluated.
Beta 2 microglobulin	Beta 2 microglobulin	Serum/ Serum gel	1 ml	Royal Free	21	See Appendix 5	1.16 – 3.21	
Biomarkers	Biomarkers	Serum, specimens containing sodium azide should not be used.	1ml		93	See Appendix 5	<200 pg/ml	Soluble Fas Ligand is measured; this is part of the FAS/FASL apoptosis
Btk	Btk	EDTA	5 ml		8	See Appendix 5	Interpretative comment provided	Flow cytometry. Abnormal in X-linked agammaglobulinaemia (hypogammaglobulinaemia with absent B cells) For patients with absent B cells, protein test not done, only genetics. For patients with some B cells, protein assay used as a screen.
C1 Esterase inhibitor	C1 Esterase inhibitor	Serum/ serum gel	1 ml	Royal London	30	See Appendix 5	Level:150-350 mg/l Function: >84%	Low levels (85%) or non-functional protein (15%) are associated with hereditary angioedema. C4 low in patients during, and normally between attacks.
C1q antibodies	Anti C1q antibodies	Serum/ serum gel	1 ml	Sheffield	60	See Appendix 5	0-15 U/ml	Described in hypocomplementaemic urticarial vasculitis, rheumatoid vasculitis, SLE (paed and adult), mesangiocapillary glomerulonephritis and IgA nephropathy.
C1q levels	C1q levels	Serum/ serum gel	1 ml	PRU Sheffield	60	See Appendix 5	50-250 mg/l	Classical pathway component. Deficiency gives autoimmune disease/recurrent infection. Only measure if Complement Functional : classical low and Complement Functional: Alternative normal. Other complement component measurements available on request
C2 levels	C2 complement	Serum/ serum gel	1 ml	PRU Sheffield	60	See Appendix 5	10-80mg/l	Classical pathway component. Deficiency gives autoimmune disease/recurrent infection. Only measure if Complement Functional : classical low and Complement Functional: Alternative normal. Other complement component measurements available on request

C3 Nephritic factor	C3 Nephritic factor	Serum/ serum gel	1 ml	PRU Sheffield	21	See Appendix 5	See Interpretative Comment	Stabilises the alternative pathway C3 convertase. Results in a low C3 and seen in membranoproliferative glomerulonephritis and partial lipodystrophy. Only done if C3 low.
Calprotectin	Calprotectin	Faeces	50p size	Charing Cross	28	See Appendix 5	< 50.0 mg/kg	High levels may help in discrimination of inflammatory bowel disease from other GI symptoms
Candida stimulation	Candida stimulation	Lithium heparin	5 ml		16	See Appendix 5	Interpretive comment provided	May be low if the patient has not had thrush (ie not encountered the antigen)
Cardiac muscle antibodies	Antimyocardial antibodies	Serum/ serum gel	1 ml	Royal London	28	See Appendix 5	Reported as positive/ Negative	Described in myocarditis, idiopathic dilated cardiomyopathy, rheumatic carditis and Dresslers syndrome.
CD3 stimulation	CD3 stimulation	Lithium heparin	5 ml		8	See Appendix 5	Interpretative comment provided	
CD40	Molecular Immunology Miscellaneous	EDTA	5 ml		8	See Appendix 5	Interpretative comment provided	Research assay. Abnormal in one type of AR-HIM syndrome
CD40 Ligand (CD154)	CD40 Ligand	Li Hep	1 ml		8	See Appendix 5	Interpretative comment provided	Flow cytometry. Abnormal in X-linked hyperIgM syndrome.
CD62L shedding	L-selectin shedding	LiHep	1 ml		7	See Appendix 5	Interpretative comment provided	Will be abnormal in patients with IRAK4 deficiency or other defects in this pathway
CGD proteins: gp91	CGD proteins	EDTA	5 mls		8	See Appendix 5	Interpretative comment provided	Flow cytometry. X-linked CGD. The lab will select the proteins tested based on sex, maternal carrier status and family history,
CGD proteins:p47	CGD proteins	EDTA	5 mls		8	See Appendix 5	Interpretative comment provided	Flow cytometry. Autosomal recessive form. The lab will select the proteins tested based on sex, maternal carrier status and family history,
CGD proteins: p22, 67	CGD proteins	EDTA	10 mls		10	See Appendix 5	Interpretative comment provided	Immunoblot. Autosomal recessive forms. The lab will select the proteins tested based on sex, maternal carrier status and family history,

Coeliac Screen	Coeliac Screen (ttg)	Serum/ serum gel	0.5 ml		8	See Appendix 5	<7 IU/ml negative. 7-10 IU/ml equivocal. >10 IU/ml positive	IgA antibodies to tissue transglutaminase. If positive, endomysial antibodies are automatically done by lab to confirm. High positive predictive value for coeliac disease. May be falsely negative if IgA very low (included in test).
Common gamma chain	Gamma chain	EDTA	5mls		8	See Appendix 5	Interpretative comment provided	Abnormal in X-linked SCID (T-B+NK-). Stat5 phosphorylation should also be done (see below).
Complement C3 + C4	C3C4 complement. Request under Chem Path specialised	Serum/ serum gel	1 ml	Chem Path GOSH	5	See Appendix 5	C3 0.75-1.65g/l C4 0.14-0.54g/l	Consumed during sepsis. If low in this context should be rechecked. Low in SLE.
Complement Functional : classical	Functional Complement	Serum/ serum gel	1 ml		30	See Appendix 5	>40%	Measures the function of the classical (C1,2,4) and terminal complement pathways. If abnormal, recheck. Alternative pathway function will be measured in parallel with this test.
Complement Functional: Alternative	Functional Complement	Serum/ serum gel	1 ml		30	See Appendix 5	>10%	Measures the function of the alternative (C3, B, D, properdin) and terminal complement pathways. If abnormal, recheck. Classical pathway function will be measured in parallel with this test.
DHR (dihyydrorhodamine)	DHR	LiHep	1 ml		7	See Appendix 5	Interpretative comment provided	May be abnormal in CGD, atypical forms of CGD.
Double stranded DNA antibodies	ANA/dsDNA/ENA	Serum/ serum gel	ANA sample		8	See Appendix 5	Neg <7.0 IU/ml. Equivocal 7-10 IU/ml. Positive >10 IU/ml	Only performed if ANA positive. Levels of dsDNA antibody may correlate with disease activity in SLE.

Extractable nuclear antigens	ANA/dsDNA/ENA	Serum/ serum gel	ANA sample	In house/ Royal Free	Screen 8 Type 28	See Appendix 5	Reported as positive/ Negative	If ANA positive ENA screen will be done: Includes Ro, La, Sm, RNP, Jo-1, Scl-70, centromere. If screen positive, sample will be sent to Royal Free for typing. Not quantitative. ENAs may evolve during the course of paediatric connective tissue disease.
Factor H	Factor H	Serum/ serum gel	1 ml	Charing Cross	70	See Appendix 5	See Interpretative comment	Mutations in Factor H, with or without low protein, are associated with familial (D-) HUS
Factor I	Factor I	Serum/ serum gel	1 ml	Charing Cross	70	See Appendix 5	See Interpretative Comment	Deficiency associated with recurrent bacterial infections or occasionally with autoimmune disease (haemolytic anaemia, renal). AP50 will be low.
GAD antibodies	GAD antibodies	Serum/ serum gel	0.5 ml	John Radcliffe Oxford	28	See Appendix 5	0 – 5.0 U/ml neg 5.1-30 equivocal >30 positive	Antibodies may be positive pre/ early IDDM
ganglioside antibodies (includes GM1, GD1a, GT1b, GQ1b and sulphatides)	Anti ganglioside abs	Serum/ serum gel	0.5 ml	National Hospital Queens square	42	See Appendix 5	Reported as positive/ Negative	Found in a low proportion of patients with Guillain Barre syndrome, chronic demyelinating polyneuropathy and multifocal motor neuropathy. Because antibody incidence is low, these tests should not be a routine part of diagnosis.
Glomerular basement membrane antibodies	Renal (GBM) antibodies	Serum/ serum gel	0.5 ml		8	See Appendix 5	<7 negative; 7-10 equivocal; >10 positive	Goodpastures syndrome. Patients with Wegner's may have a similar renal presentation (rapidly progressive glomerulonephritis) and ANCA should also be done. If urgent contact lab.
Granule release assay (GRA)	Granule release assay	EDTA	10 mls		8	See Appendix 5	Interpretative comment provided	Tests the ability of T and NK cells to degranulate in response to a stimulus and express CD107a on their surface. Screens the pathway that includes syntaxin and Munc13-4/18-2
HLA typing B27	HLA typing B27	EDTA	4mls	Anthony Nolan, Royal Free Hospital	21	See Appendix 5	Reported as positive/ negative	HLA B27 is strongly associated with ankylosing spondylitis.

HLA typing DQ2 and DQ8	HLA typing DQ2 and DQ8	EDTA	4mls	Anthony Nolan, Royal Free Hospital	21	See Appendix 5	Reported as positive/ negative	HLA DQ2 and DQ8 are associated with coeliac disease and Type 1 diabetes.
IgD	IgD	Serum/ serum gel	0.5 ml	PRU Sheffield	40	See Appendix 5	2-100 KU/L	May be non-specifically raised in a variety of inflammatory conditions. Not specific to hyper IgD syndrome
IgE: total	IgE	Serum/ serum gel	1 ml		8	See Appendix 5	Age specific See Appendix 2	IgE may be elevated in many atopic and other conditions. Normal total IgE does not exclude allergy.
IgE:Specific (please state allergens required in reason for request) (for panels see appendix 2)	Specific IgE	Serum/ serum gel	1 ml initially then 0.5 ml per allergen		10	See Appendix 5	See appendix 2 and comments on reports	Allergens should be selected on the basis of history. There is a significant false positive rate. Further advice on interpretation can be obtained from the laboratory. Level of IgE that is significant varies with allergen. Further advice available from the laboratory. A total IgE >5000 may interfere with the assay and reduces the specificity of the test
IgG subclasses	IgG subclasses	Serum/ serum gel	1 ml	PRU, Sheffield	21	See Appendix 5	Age specific . See Appendix 1	Will always be low if total IgG low. Will not routinely be done if patient < 2y of age, or normal total IgG and IgA.
Immunoglobulins IgG, IgA, IgM	Immunoglobulins IgG, IgA, IgM. Request under Chem Path specialised	Serum/ serum gel	1 ml	Chem Path GOSH	5	See Appendix 5	Age specific . See Appendix 1	IgG may be lost via renal tract, gut etc. Check albumin is normal before interpreting IgG
Infiximab Levels and antibodies	Infliximab	Serum/ Serum gel	1ml	Royal Devon and Exeter	14	See Appendix 5	Levels reported in mg/L. Antibody reported as positive/negative	Monitors drug levels
Insulin antibodies	Insulin antibodies	Serum/ serum gel	1 ml	Sheffield	28	See Appendix 5	0-5 mg/L	May occur in insulin resistance.

Interferon beta neutralising antibodies	B interferon neut.abs	Serum/ serum gel	1 ml	National Hospital Queens square	21	See Appendix 5	<20 NU Negative 20 – 100 NU Low positive 100 – 600 NU Positive >600 NU High positive	May be found in patients who have become resistant to treatment with IFNb for MS
Lymphocyte subsets Basic	Lymphocyte subsets Basic	EDTA	0.5 ml		3	See Appendix 5	Interpretative comment provided	T4/8, B + NK
Lymphocyte subsets Memory	Lymphocyte subsets Memory	EDTA	0.5 ml		3	See Appendix 5	Interpretative comment provided	Memory T cells Naïve cells CD45RA+CD27+ Memory cells CD45RA-CD27+ Effector cells CD45RA+CD27-
Lymphocyte subsets T cell Types	Lymphocyte subsets T cell Types	EDTA	0.5 ml		4	See Appendix 5	Interpretative comment provided	T Cell Types Gamma delta T cells: may be raised in infection, autoimmunity and may fail to express CD4 or CD8. Double negative T cells Increased numbers of CD3 positive, CD4 and CD8 negative T cells are seen in the peripheral circulation in patients with ALPS, but are also found in other inflammatory and immune dysregulated states. This is a screening test for ALPS
Lymphocyte subsets Activation	Lymphocyte subsets Activation	EDTA	0.5 ml		3	See Appendix 5	Interpretative comment provided	CD38 is a marker of early activation and in studies has been shown to be useful in monitoring HIV patients being to fail therapy prior to observing decreased CD4 counts. DR (MHC II) is a useful marker of activation. MHC II deficient SCIDs may be CD4 lymphopaenic
Lymphocyte subsets CD25	Lymphocyte subsets CD25	EDTA	0.5 ml		3	See Appendix 5	Interpretative comment provided	CD25 is upregulated on activated CD4 positive T cells. Rare deficiency of this molecule also described
Lymphocyte subsets MHC1	Lymphocyte subsets MHC1	EDTA	0.5 ml		4	See Appendix 5	Interpretative comment provided	Rare cases of deficiency are described

Lymphocyte subsets neutrophil panel	Lymphocyte subsets neutrophil panel	EDTA	0.5 ml		4	See Appendix 5	Interpretative comment provided	CD11a, 18. CD18 is beta2 integrin which is abnormal in leucocyte adhesion deficiency type I. CD11a binds to CD18 in the membrane, and therefore will also not be expressed.
Lymphocyte subsets Extended B memory	Lymphocyte subsets Extended B memory	EDTA	0.5 ml		10	See Appendix 5	Interpretative comment provided	EUROCLASS method. Assesses proportion of naïve, marginal zone, switched memory and transitional B cells and plasmablasts. Abnormalities have been defined in patients with CVID and other immune deficiencies.
Lymphocyte subsets CD19/CD20	Lymphocyte subsets CD19/CD20	EDTA	1 ml		3	See Appendix 5	Interpretative comment provided	Routine B cell marker is CD19. Patients may fail rituximab (anti-CD20 antibody) treatment if they fail to express CD20. Lack of CD19 (but presence of CD20) is a cause of CVID.
Lymphocyte subsets ThyE	n/a	EDTA	1 ml		10	See Appendix 5	Interpretative comment provided	Looks for CD31 positive cells, which are a surrogate marker of thymic emigrants.
Lymphocyte subsets Treg	Lymphocyte subsets Treg	EDTA	1 ml		10	See Appendix 5	Interpretative comment provided	Regulatory T cells are defined as CD4+/CD25+/Cd127low/FoxP3+. This panel detects cells that are CD4+/CD25+/CD127low and are most likely T regulatory cells.
Mannose binding lectin	MBL	Serum/ serum gel	1 ml		22	See Appendix 5	See appendix 3	Low levels correlate with polymorphisms. Increased risk/severity of infections.
Meningococcal W and Y antibodies	n/a	Serum/ serum gel	1 ml	Manchester	30	See Appendix 5	See Interpretative Comment	
Mixed lymphocyte culture	Mixed lymphocyte culture	Lithium heparin	5-10 ml from patient and donor(s) (depends on the no. of donors to be tested)		14	See Appendix 5	Interpretative comment provided	Only undertaken to asses suitability of family donors for BMT or 3rd party immune function.
Molecular Immunology Miscellaneous (specify test in reason for request)	EDTA		Varies with test		Varies with test	See Appendix 5		Requested after discussion with Clinical Lead, laboratory manager or senior healthcare scientist.

Myeloid Related Protein (MRP 8 and 14)	MRP	Serum/ Serum gel	1 ml		28	See Appendix 5	See appendix 4	MRP 8 and 14 are endogenous TLR-4 ligands that are expressed in granulocytes and monocytes and promote inflammatory processes in vivo.
MUSK antibodies	MUSK Abs	Serum/ serum gel	1 ml	Oxford	40	See Appendix 5	Reported as positive/ Negative	May be found in AChR negative myasthenia gravis
Myositis Profile	Myositis Profile	Serum/ serum gel	1ml	Royal United Bath Hospital	21	See Appendix 5	Reported as negative/positive for named ENA.	Looks for ENAs associated with myositis
NBT (nitroblue tetrazolium test)	NBT	EDTA	1 ml		3	See Appendix 5	Interpretative comment provided	Excludes all common forms of CGD
Neutrophil phagocytosis	Phagocytosis	LiHep	1 ml		7	See Appendix 5	Interpretative comment provided	Tests the ability of neutrophils to phagocytose opsonised material over a time course
NK Killing Assay	NK Killing	LiHep	5 mls		8	See Appendix 5	Interpretative comment provided	LSS should be requested to ensure the presence of NK cells before assay setup. Low or absent natural killer (NK) cell activity is common prior to and during active disease, as well as after remission following chemotherapy in a significant proportion of individuals with Familial HLH.
NMDA receptor antibodies	NMDA rec. antibodies	Serum/ serum gel	1 ml	Oxford	40	See Appendix 5	Reported as positive/ Negative	May be found in autoimmune encephalitis, typically in children and young people with prominent psychotic symptoms.
Paraneoplastic antibodies	Paraneoplastic antibodies	Serum/ serum gel	1ml	Oxford	40	See Appendix 5	Reported as positive/ negative	Screen includes anti yo, Hu, Ri,MA1, MA2, CV2, CRMP%, amphiphylin. Detected in a variety of autoimmune neurological conditions, originally described as paraneoplastic in adults.
Perforin	Perforin	EDTA	5 mls		8	See Appendix 5	Interpretative comment provided	Flow cytometry. Mutations in perforin are found in 30% of patients with familial HLH. Other proteins that may be deficient in HLH are also available on request.
PHA (PHA: separated cells)	PHA: separated cells	Lithium heparin	5 ml		14	See Appendix 5	Interpretative comment provided	

PHA +/- IL2	PHA +/- IL2	Lithium heparin	5 ml		14	See Appendix 5	Interpretative comment provided	Synergy is normally seen between PHA and IL2. this may be lacking in patients with IL2 receptor/function defects
PHA stimulation	PHA	Li Hep	0.5 ml		14	See Appendix 5	Interpretative comment provided	May be abnormal if lymphocyte count low. Consider PHA: separated cells
Pneumococcal abs (serotype specific)	Pneumococcal abs	Serum/ serum gel	0.5 ml	Addenbrookes	28	See Appendix 5	Putative protectivelevel 0.35 ug/ml, optimal level >0.5ug/ml	7 serotypes contained in Prevenar and 5 non-vaccine serotypes measured. Results must be interpreted with vaccine and exposure history.
RF (Rheumatoid factor)	Rheumatoid factor	Serum/ serum gel	1 ml	Royal Free	21	See Appendix 5	< 20 IU/ml negative. >20 positive	Testing for rheumatoid factor is a poor screening procedure for juvenile idiopathic arthritis and of supportive diagnostic value only in the highly restricted population of older children with polyarticular arthritis (positive in up to 33%). Poor prognostic feature. Levels do not correlate with disease activity. Also found in many other connective tissue and inflammatory diseases.
SAP	SAP	EDTA	5mls		8	See Appendix 5	Interpretative comment provided	Flow Cytometry. Abnormal in X-linked lymphoproliferative disease 1.
Save Cells in Liquid Nitrogen	Save cells in liquid nitrogen	Lithium heparin / EDTA	5 - 10 ml		n/a	See Appendix 5	n/a	Sample will depend on future tests required. If sample is to be stored for subsequent therapeutic use, please book with lab well in advance as processing requirements different.
Soluble CD25	Soluble CD25	Serum	1ml		21	See Appendix 5	<2500 pg/ml	May be raised in HLH, declining with response to treatment.
Stat 5 tyrosine phosphorylation	Stat5	EDTA	5 ml		8	See Appendix 5	Interpretative comment provided	Measures signalling via the IL2, 7 or 15 receptors. Will be abnormal if components of this pathway are missing/non-functional (IL2: gamma chain, Jak-3, IL7: IL7Ra). Used as a functional screen of the pathways so that other specific tests can be undertaken after this.
Tetanus antibodies	Tetanus antibodies	Serum/ serum gel	1 ml	Sheffield	42	See Appendix 5	>0.1 iu/ml	If low, a booster vaccine should be given and retest 4-6 weeks later
Thyroid antibodies	Thyroid antibodies	Serum/ serum gel	0.5 ml		8	See Appendix 5	<60 Negative; 60-100 Equivocal; >100 positive	Presence consistent with autoimmune thyroid disease and indicates the need to monitor thyroid function. Not specific for any one thyroid disease.

Tryptase (Mast cell)	Tryptase	Serum/ serum gel	1 ml	Royal Free	21	See Appendix 5	2-11.4 mcg/l	Raised after anaphylaxis. Samples must be taken at the time of the reaction, at 4-6 hours and 24 hours to allow interpretation. Include time of reaction and samples on form.
TSHR antibodies	TSHR ab	Serum/ serum gel	0.5 ml	Sheffield	40	See Appendix 5	See report	Antibodies may be an additional finding in patients where thyroblobulin abs neg but autoimmune thyroid disease possible. Only available in selected cases after discussion with lab
V beta repertoire	V beta repertoire	EDTA	2 mls		8	See Appendix 5	Interpretative comment provided	Looks at surface expression of T cell receptor V beta families. In normal individuals at least 70 % of their T cells will be detected with the panel of antibodies used, and there will be a polyclonal distribution. Oligoclonal expansions on a polyclonal background are suggestive of infection or malignancy, whilst an oligoclonal distribution is seen in defects of T cell development.
Voltage gated calcium channel antibodies	n/a	Serum/ serum gel	1 ml	Oxford	40	See Appendix 5	0 – 45 pM	May be found in Lambert-Eaton myaesthenic syndrome
Voltage gated potassium channel antibodies	VGKC abs	Serum/ serum gel	1 ml	Oxford	40	See Appendix 5	0 - 100 pM	Associated with limbic encephalitis, described in the literature as amenable to immune modulation
WASP	WASP	EDTA	10 mls		8	See Appendix 5	Interpretative comment provided	Flow cytometry Protein may be absent or reduced in quantity in Wiskott Aldrich syndrome. Small platelets are a consistent feature and platelet volume should also be requested on these patients.
XIAP	XIAP	EDTA	5 mls		8	See Appendix 5	Interpretative comment provided	Flow Cytometry. Abnormal in X-linked lymphoproliferative disease 2.
	***If results are required urgently, please liaise with the immunology lab**							

Bone marrow/ Peripheral Blood stem cell manipulations

All peripheral blood stem cells (HPC-A) or bone marrows (HPC-M) for administration to patients must be processed and issued by the Cell Therapy Laboratory (CTL) and should never be accepted directly by the ward.

Manipulation of bone marrow may be required to reduce volume or due to blood group incompatibility. Manipulation of bone marrow or PBSCs may be required to select stem cells (CD34+ cells), deplete T and B cells (alpha/beta and CD19 depletion) or to enrich/deplete another cell population.

All manipulations can only be performed if booked well in advance (at least 1 week) with the CTL and an approved protocol is provided. Please contact the laboratory as soon as the potential date is known.

CTL may only be able to perform **one** manipulation per day.

Depending on the manipulation needed, the time taken will range from 2 h to > 6 hours. Contact CTL (0433) for more details on individual patients.

If cells are to be stored the following must be undertaken:

- Virological and other infectious screening of donor
- Consent from Donor for storage
- Booking of the procedure in advance

All the relevant forms are available from the BMT team or the laboratories.

Appendix 1

Immunoglobulins G,A,M

AGE	IgG g/l	IgA g/l	IgM g/l
Cord	5.2 - 18.0	<0.02	0.02 - 0.2
0 - 2 weeks	5.0 - 17.0	0.01 - 0.08	0.05 - 0.2
2 - 6 weeks	3.9 - 13.0	0.02 - 0.15	0.08 - 0.4
6 - 12 weeks	2.1 - 7.7	0.05 - 0.4	0.15 - 0.7
3 - 6 months	2.4 - 8.8	0.10 - 0.5	0.20 - 1.0
6 - 9 months	3.0 - 9.0	0.15 - 0.7	0.40 - 1.6
9 - 12 months	3.0 - 10.9	0.20 - 0.7	0.60 - 2.1
1 - 2 years	3.1 - 13.8	0.30 - 1.2	0.50 - 2.2
2 - 3 years	3.7 - 15.8	0.30 - 1.3	0.50 - 2.2
3 - 6 years	4.9 - 16.1	0.40 - 2.0	0.50 - 2.0
6 - 9 years	5.4 - 16.1	0.50 - 2.4	0.50 - 1.8
9 - 12 years	5.4 - 16.1	0.70 - 2.5	0.50 - 1.8
12 - 15 years	5.4 - 16.1	0.80 - 2.8	0.50 - 1.9
15 - 45 years	6.0 - 16.0	0.80 - 2.8	0.50 - 1.9
45+ years	6.0 - 16.0	0.80 - 4.0	0.50 - 2.0

IgG Subclasses
Ranges:

AGE	IgG1 g/l	IgG2 g/l	IgG3 g/l	IgG4 g/l
Cord	3.6 - 8.4	1.2 - 4.0	0.3 - 1.5	< 0.5
6 months	1.5 - 3.0	0.3 - 0.5	0.1 - 0.6	< 0.5
2 years	2.3 - 5.8	0.3 - 3.9	0.1 - 0.8	< 0.5
5 years	2.3 - 6.4	0.7 - 4.5	0.1 - 1.1	<0.5
10 years	3.6 - 7.3	1.4 - 4.5	0.3 - 1.1	<1.0
15 years	3.8 - 7.7	1.3 - 4.6	0.2 - 1.2	<1.1
Adult	3.2 - 10.2	1.2 - 6.6	0.2 - 1.9	<1.3

Appendix 2

IgE

AGE	NORMAL RANGE KU IgE/L
Newborn	0-5
Newborn – 3 months	0-11
3 months – 1 year	0-29
1 year – 5 years	0-52
5 years – 10 years	0-63
10 years – 15 years	0-75
Adult	0-81

SIGE Specific IgE

No normal ranges. A positive result indicates sensitisation to that substance. Level at which allergic reactions are likely varies with allergen

Results are indicated as:-

Value		Grade	Interpretation
<0.35	KUA/l	0	Negative
0.35 – 0.69		1	Weak positive
0.70 – 3.49		2	Positive
3.50 – 17.49		3	Positive
17.5 – 52.49		4	Strong positive
52.5 – 100		5	Strong positive
>100		6	Strong positive

SIGE Panels

Food Screen	cows milk, egg, soya, wheat
Atopic dermatitis	egg, cows milk, wheat, soya, HDM
Atopic eczema	Egg white, cows milk, wheat, soya, peanut, HDM, cat, dog, mixed grasses, mixed trees, mixed moulds
Asthma/rhinitis	HDM, mixed grass, birch, cat, dog
Latex	
Peanut	
Tree nut	specify nuts in reason for request
Other	specify allergens in reason for request

Mixes contain the following:

Grasses	Cocksfoot, meadow fescue, rye grass, timothy grass, Meadow grass
Trees	Box-elder, silver birch, oak, elm, walnut
Mould	Penicillim chrysogenum, Cladosporium hebarum, Aspergillus fumigatus, Candida albicans, Alternaria alternate, Setomelanomma rostrara

Appendix 3

MBL Mannan Binding Lectin

From unpublished data under review from Prof.M Turner/Dr.N Klein

A tight correlation has been identified between MBL level and genotype.

>1300ng/ml	correlate with wild type alleles showing no deficiency
1300-400ng/ml	correlate with heterozygous variant alleles and may show deficiency associated with some increased risk of infection.
>400ng/ml	Correlate with functional MBL deficiency associated with increased risk of infection.
<75ng/ml	Correlate with homozygous variant alleles and non-functional MBL associated with the greatest risk of infection.

Appendix 4

In JIA, serum concentration of MRP has potential value in predicting outcome in two situations:

1. Risk of flare of arthritis after withdrawal of MTX. Applies to MTX when it is the sole DMARD, Foell et al JAMA 2010. Actual values are dependent on local assay and standard curve. For GOSH measurements, the values are –

a) < 4000 ng/mL = low ; low risk of arthritis flare after stopping MTX.

b) 4000- 6000 ng/mL, medium – some risk of arthritis flare after stopping MTX.

c) >6000 ng/mL, high- high risk of arthritis flare after stopping MTX.

Note: We do not yet know if this biomarker has value in predicting uveitis, or flare after stopping other drugs

2. **Chance of good response to MTX, being started for arthritis,** Moncrieffe et al 2013

a) if MRP is > 12,000 ng/ML child has a high chance of reaching >ACR70 response in first 4- 6 months of MTX .

b) if MRP< 12,000 ng/mL, the test at present cannot help you predict good response

Appendix 5: Frequency of testing

Test	Frequency	Exceptions	Reference
ABC PANEL	monthly		
ACHR ABS	3 months		RCPath
ADRENAL CORTEX ABS	12 months		RCPath
ANCA	3 months	First measurement, may repeat within 1 month. Plasmapheresis, may need more frequent repeat	RCPath and Brit Soc Rheum
ANTI IGA	12 months		RCPath
ANTI NUCLEAR ABS	protocol in place		
APOPTOSIS BIOMARKERS	No repeat if normal	If abnormal, may repeat x1 to confirm	
APOPTOSIS FUNCTIONAL	No repeat if normal	If abnormal, may repeat x1 to confirm	
AQUAPORIN 4	6 monthly		RCPath
AUTO IMMUNE PROFILE	12 months		RCPath
B CELL PANEL	only if antiCD20 failure		
B2 MICROGLOBULIN	monthly		RCPath
BASAL GANGLIA AB	6 monthly	based on clinical context	RCPath
B-INTERFERON NEUTRALISING ABS	6 monthly	based on clinical context	
BTK	No repeat if normal	If abnormal, may repeat x1 to confirm	
C1 ESTERASE INHIBITOR	No repeat if normal	If abnormal, may repeat x1 to confirm	
C1Q ABS	12 months		RCPath and Tarzi et al
C1Q LEVEL	No repeat if normal	If abnormal, may repeat max monthly	RCPath and Tarzi et al
C2 LEVEL	No repeat if normal	If abnormal, may repeat max monthly	RCPath
C3 NEPHRITIC FACTOR	1 month		Rcpath
CALPROTECTIN	monthly		
CANDIDA STIMULATION	3 monthly		
CD154 EXPRESSION monitoring	6 monthly		
CD154 Diagnostic	No repeat if normal	If abnormal, may repeat x1 to confirm	

CD25 PANEL	monthly		
CD3 STIMULATION	3 monthly		
CGD	No repeat if normal	If abnormal, may repeat x1 to confirm	
COELIAC SCREEN/IGA	3 monthly		RCPath
COMPLEMENT FUNCTIONAL PATHWAY	normal - once	pts on ecluzimab	RCPath
DHR	No repeat if normal	Abnormal, may repeat x1 to confirm	
DNA ABS	protcool in place		
ENA SCREEN	protcool in place		
ENA TYPING	protcool in place		
ENDOMYSIAL ABS	protcool in place		
EXTENDED B MEMORY PANEL	6 monthly		
FACTOR H	once		
FACTOR I	once		
FUNCTIONAL C1 ESTERASE INHIBITOR	No repeat if normal	If abnormal, may repeat x1 to confirm	
GAD ABS	3 monthly		RCPath
GAMMA CHAIN	once		
GAMMA CHAIN POST THERAPY PANEL	protol in place		
GANGLIOSIDE ABS	6 monthly		RCPath
GBM ABS	3 months	First measurement, may repeat within 1 month. Plasmapheresis, may need more frequent repeat	RCPath
GRA	No repeat if normal	If abnormal, may repeat x1 to confirm	
HLA TYPING	once		
IGD	once		
IGG SUBCLASSES	protocol in place		RCPath
IMMUNOBLOT	test dependent		
IMMUNOLOGY SAVE SERUM	test dependent		
INSULIN ABS	12 months		RCPath
L SELECTIN SHEDDING	No repeat if normal	If abnormal, may repeat x1 to confirm	
LSS BASIC PANEL	monthly	post biologics, trial patients	
MANNOSE BINDING LECTIN	No repeat		

MHC CLASS 1 EXPRESSION	No repeat		
MISC2S	test dependent		
MISCI	test dependent		
MISCS	test dependent		
MIXED LYMPHOCYTE CULTURE	once per donor		
MONOCYTE PANEL	trial dependent		
MUSK ABS	3 monthly		RCPath
MYOCARDIAL ABS	3 monthly		
NBT TEST	No repeat if normal	If abnormal, may repeat x1 to confirm	
NEUTROPHIL PANEL	No repeat if normal	If abnormal, may repeat x1 to confirm. Post HSCT in CD18 def may use as monitoring more frequently	
NEUTROPHIL PHAGOCYTOSIS	No repeat if normal	If abnormal, may repeat x1 to confirm	
NMDA RECEPTOR ABS	3 monthly		RCPath
NO CELLS	No block		
PARANEOPLASTIC AB	12 months		RCPath
PERFORIN	once		
PHA STIMULATION	3 monthly		
PHAIL2 STIMULATION	3 monthly		
PNEUMOCOCCAL ABS	6 weeks		
RESEARCH ASSAY	test dependent		
RHEUMATOID FACTOR	12 months		Rcpath
SAPH/XIAP	No repeat if normal	If abnormal, may repeat x1 to confirm	
SAPX/XIAPX	No repeat if normal	If abnormal, may repeat x1 to confirm	
SAVED CELLS IN LIQUID NITROGEN	request dependent		
SAVED IGS	yearly or at Ig change		
SAVED TISSUE	as requested		
sCD25	fortnightly		
SCLN MOLECULAR	request dependent		
SEPARATED CELLS PHA STIMULATION	3 monthly		
SPECIFIC IGE (extra allergens)	6 monthly		RCPath
SPECIFIC IGE (first allergen)	6 monthly	? Have 11m for specifics already	RCPath and local

STAT5	No repeat if normal	If abnormal, may repeat x1 to confirm	
T CELL ACTIVATION PANEL	monthly		
T CELL TYPES PANEL	monthly		
T MEMORY PANEL	monthly		
TEST NOT PERFORMED	No block		
TETANUS ABS	once if normal, 2xs pre and post vacc	2xs post hsct	
THY-E PANEL	monthly		
THYROID ABS	12 monthly		
TOTAL IGE	11 months	already set?	RCPath and local
Treg CELLS PANEL	monthly		
TRYPTASE	No block		RCPath
TSH RECEPTOR ABS	12 months		RCPath
TTG IGA	3 monthly		RCPath
V BETA PANEL	3 monthly		
VGKC ABS	based on clinical context		Rcpath
WASP	No repeat if normal	If abnormal, may repeat x1 to confirm	
ZAP70	No repeat if normal	If abnormal, may repeat x1 to confirm	