Welsh Blood Service Transplantation Laboratories

USER GUIDE

Transplantation Laboratories
Welsh Blood Service
Ely Valley Rd, Pontyclun CF72 9WB
Telephone: (01443) 622000
e-mail: wtail@wales.nhs.uk
Website: welsh-blood.org.uk/wtail/

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TRANSPLANTATION LABORATORIES USER GUIDE

1. Contact details

Transplantation Laboratories
Welsh Blood Service
Ely Valley Road, Pontyclun
Wales
CF72 9WB
Tel: 01443 622186

In the event of telephone line failure on the above number, during working hours the Transplantation laboratory office can be contacted on 01443 226055.

Consultant Clinical Scientist
Deborah Pritchard Head of Transplantation Services Email: Deborah.Pritchard3@wales.nhs.uk

Laboratory Contacts

For general enquiries please contact the general Serology laboratory on 01443 622186.

Transplantation Service	Primary Lead	Tel.	Section
Renal and Pancreas Transplantation	Emma Burrows	01443 622186	Head of Solid Organ Transplantation Services
HLA Antibody Investigations			
Platelet Investigations			
Haemopoietic Stem Cell Transplantation	Jennifer Pepperall	01443 622041	Head of Stem Cell Transplantation Services
HLA-B27 typing in the spondyloarthropathies			
Miscellaneous Disease Investigations (excluding HLA-B27)			
Fetal RhD screening			
General Service and Research Enquiries	Deborah Pritchard / Felicity May	01443 622175 / 622181	Head of Transplantation Services / Deputy Head of Transplantation Services

Additional Services Hosted by the Laboratory

Service	Primary Lead	Ext.	Website Link
WBMDR	Christopher Harvey	622025	WBMDR
UK NEQAS for H&I	Amy De'Ath	622185	UK NEQAS for H&I

2. Introduction

Transplantation Laboratories, together with the Welsh Bone Marrow Donor Registry (WBMDR), form Transplantation Services (formerly WTAIL). The UK National External Quality Assessment Services for Histocompatibility and Immunogenetics (UK NEQAS for H&I) is a hosted organisation within this department.

Transplantation Laboratories provides Histocompatibility and Immunogenetics (H & I) support to a variety of health care providers ranging from specialist transplant units to General Practitioners. This service is critical to both solid organ and blood stem cell transplant programmes. The services provided include HLA typing, patient/donor crossmatching, HLA antibody testing, solid organ and stem cell donor/recipient matching, platelet typing and platelet antibody testing. Our staff work closely with clinicians in the management of solid organ transplant, blood stem cell transplant and thrombocytopenic patients.

3. Service Provision

Histocompatibility Testing for Solid Organ Transplantation

Transplantation Laboratories provide laboratory testing and clinical advice for patients requiring a pancreas or kidney who are transplanted in Wales. The service supports transplant of various donor types including:

- Local deceased donors
- Deceased donors allocated via NHSBT Organ Donation and Transplant (ODT)
- Potential living renal donors
- ABO incompatible donors
- HLA incompatible donors
- Altruistic donors
- Paired-pool exchange donors

Laboratory testing encompasses HLA typing, crossmatching and HLA antibody detection/identification. HLA typing is performed by polymerase chain reaction (PCR)- based methods at the resolution required for patient registration and donor allocation by NHSBT ODT. Additional typing is performed using next generation sequencing to assist in antibody definition.

HLA antibody screening is routinely performed on potential transplant patients every 3 months, and following any sensitisation events, by Luminex based methods. Testing schedules for patients receiving antibody incompatible transplants are discussed and agreed with the transplant team on an individual basis. Donor specific antibody testing is performed for transplant patients where antibody mediated rejection is suspected and to monitor effectiveness of treatment.

Allogeneic and autologous crossmatching is performed by Flow Cytometry.

The Laboratory manages patient registration and updates with ODT.
Clinical advice in relation to risk and donor selection is provided.

Histocompatibility Testing for Haematopoietic Stem Cell Transplants

Transplantation laboratories supports services to the local transplant centre for allogeneic stem cell transplants from related and unrelated donors. HLA-typing of patients and potential donors is performed by PCR-based methods, and next generation sequencing. If required, the patient is tested for HLA antibodies by Luminex based methods.

A graft identification advisory service is provided and Transplantation Laboratories liaise with WBMDR staff to facilitate local, national and international unrelated donor searches, verification typing sample provision and stem cell products. WBMDR is licensed by the Human Tissue Authority to import haemopoietic stem cells.

Platelet Immunology Service

The laboratory provides investigations for a variety of immune thrombocytopenia which include:

- Platelet refractoriness
- Neonatal alloimmune thrombocytopenia
- Post transfusion purpura

HLA and HPA antibody screening is performed by Luminex based methods and the Monoclonal Antibody Immobilization of Platelet Antigen (MAIPA) test, respectively. HLA and HPA typing is performed by PCR-based methods.
The Laboratory works in close collaboration with the Welsh Blood Service, selecting suitable HLA/HPA selected platelets when clinically required.

The 'All Wales Guidelines for HLA and HPA Selected Platelets' and forms for requesting platelets and providing increment data can be accessed from the Blood Health Team Transfusion Resources pages (https://wbs-intranet.cymru.nhs.uk/bht/policies-guidance-forms/)

Disease Diagnosis and Drug Hypersensitivity

HLA typing is provided to support disease diagnosis for a number of diseases e.g. HLA B27 and ankylosing spondylitis, DQA/DQB for coeliac disease and HLA-B*57:01 for Abacavir sensitivity.

Transfusion Related Acute Lung Injury

Investigations for this include HLA antibody screening of the implicated donors and the patient using Luminex technology and HLA typing where appropriate. Forms can be obtained from Blood Health Team Transfusion Resources pages (https://wbs-intranet.cymru.nhs.uk/bht/policies-guidance-forms/forms/). Investigations for HNA antibodies are referred to the NHSBT.

Miscellaneous

The laboratory is happy to discuss requirement for ad-hoc HLA typing or antibody definition e.g. for appropriate research projects. Please contact Deborah Pritchard for further information.

Fetal RhD Screening

The laboratory provides an all-Wales Fetal RhD testing service for RhD-negative expectant mothers to reduce the use of anti-D immunoglobulin (a human blood product) in the management of Rhesus disease. This testing is performed by a non-invasive (cell-free) methodology, wherein the fetal RhD type of a singleton or twin pregnancy is determined using a maternal peripheral blood sample. Expectant mothers who are found to be carrying a RhD negative baby are not at risk of Rhesus disease and do not require prophylaxis.

4. Laboratory Tests

The current range that can be undertaken are given below:

Serological Crossmatching and Antibody Screening Tests

- **Flow cytometry crossmatch (FCXM)**
  This is a more sensitive crossmatch than CDC and uses donor or recipient lymphocytes and recipient sera using a flurochrome labelled secondary antibody in order to detect IgG donor-specific antibodies against T and/or B cells. Donor specific antibodies detected using these techniques are considered an intermediate/high risk to transplant.

- **HLA Class I & Class II antibody detection by Luminex based methods**
  This is specific for IgG HLA class I and/or II antibodies, including both complement and non-complement fixing sub classes, utilising Luminex technologies. This technique is more sensitive that CDC, is highly specific and can resolve complex antibody profiles in highly sensitised patients using beads coated with a single HLA antigen. This technique is used to detect and specify antibodies in solid organ recipients, potential HLA mismatched HSCT recipients, platelet refractoriness and for monitoring HLA antibodies in HLA antibody incompatible transplants.

- **HNA antibody screening by Luminex based methods**
  This is specific for IgG HLA class I and/or II antibodies, including both complement and non-complement fixing sub classes, utilising Luminex technologies. This technique is used to detect HNA antibodies in solid organ recipients and as part of TRALI risk reduction measures.

- **MAIPA (Monoclonal Antibody Immobilisation of Platelet Antigen)**
  This sensitive technique detects antibodies directed to different platelet glycoproteins. This allows the identification of mixtures of platelet specific antibodies in the presence of HLA

antibodies. The patient's serum is incubated with a panel of HPA typed platelets and defined antibodies reported.

DNA-based HLA and HPA typing – using RT-PCR and NGS methodology

- **HLA Class I & II DNA typing: low resolution**
  Determination of HLA-A, -B, -C,-DR, -DQ and DP alleles or groups of alleles by DNA analysis giving definition comparable to serological typing, for the purpose of HLA typing in solid organ transplantation, as an aid to disease diagnosis and for selection of HLA compatible platelet transfusions.

- **HLA Class I & II DNA typing: high/Allelic level resolution (up to 3rd field)**
  Determination of HLA-A, -B, -C,-DR, -DQ and -DP alleles by DNA analysis giving high resolution definition. This is used for matching in haemopoietic stem cell transplantation in the related and unrelated setting and assists in HLA antibody specification.

- **Human Platelet Antigen (HPA) Typing**
  Determination of HPA 'antigens' by typing for HPA alleles using RT-PCR based techniques.

- **HLA B*57:01 typing**
  Determination of HLA-B*57:01 (positive or negative) and differentiation of the 'common' HLA B57 subtypes (subtypes determined by NGS). This is used to determine patients at risk of hypersensitivity type reactions to Abacavir.

- **HLA-B27 typing by RT-PCR**
  Determination of HLA-B27 (positive or negative) status using an RT-PCR based technology.
  Associations of HLA antigens with rheumatic diseases have been well established over the last two decades. Notably HLA-B27 typing has been particularly useful in the diagnosis of the atypical spondyloarthropathies.

Additional tests

- **HLA and disease association testing by molecular techniques**
  The laboratory performs routine HLA typing in relation to disease association investigations for specific patients, e.g. patients being investigated for suspected coeliac disease.
  In consultation with our customers we can undertake any specific HLA typing request, or family studies. We also undertake disease susceptibility/association projects via prior agreement with the Head of Transplantation Services.
  Determination of HLA-A, B, C, DRB1/3/4/5, DQA1/DQB1, DPB1 HLA types in combinations and resolution appropriate to user requirements.

- **ABO titres**

This is performed using donor cells and recipient plasma for assessing and monitoring ABO antibody removal for ABO incompatible kidney transplants. A diamed gel card method is used to determine titres by saline and AHG.

- **Fetal RhD screening**
  The laboratory uses a commercial kit (Devyser RHD) to perform non-invasive fetal RhD screening on cell-free DNA obtained from RhD-negative maternal plasma. The presence of circulating RhD DNA is indicative of the mother expecting a RhD-positive baby.

In addition to these tests, expert H&I advice is available from our staff.

5. Laboratory Hours

Routine

Monday to Friday 09.00-17.00 (excluding Bank Holidays)

Out of Hours Requests

The H&I service for deceased kidney and pancreas donor HLA typing and crossmatching is available 24 hours a day, 365 days a year.
The out of hours service is staffed by HCPC registered Clinical Scientists and Biomedical Scientists, all of whom are employed within Transplantation Services.
For each on call period there are three members of the team available: two laboratory staff are available, who perform HLA typing and patient/donor crossmatching, and a Consultant Clinical Scientist.

Contact Arrangements out of hours - On-call Scientist

Contact WBS main switchboard (01443 622000) and ask for either the 'HLA Typer', 'Crossmatcher' or Consultant on-call for Transplantation Services.
In the event of telephone line failure please contact reception/security on 01443 237952 or Transplantation laboratory office on 01443 226055.
Consultant advice may be obtained from the Head of Transplantation Services or Duty Consultant for WBS, both available via WBS switchboard as above.

6. Sample Requirements and Reporting

Request Forms

Please use the standard Transplantation request forms for renal/pancreas transplantation, stem cell transplantation and disease association testing available from the Transplantation website: (https://portal.welsh-blood.org.uk/wtail/request-forms/). Complete ALL sections as appropriate. Links to 'Platelet and White Blood Cell Investigation' request forms (including TRALI) can also be found here: https://wbs-intranet.cymru.nhs.uk/bht/policies-guidance-forms/forms/.

Request Form requirements

Details (Please note patient details on sample & request form must match exactly)	Requirement	Outcome if missing from request form
Patient name (Surname & Forename)	Mandatory	Discard
Date of birth	Mandatory	Discard
Hospital number/NHS number	Mandatory	Discard
Clinical details/ tests required	Mandatory	Discard if we can not ascertain required details
Diagnosis	Desirable	Process
Date and where applicable time (e.g. solid organ rejection cases)	Desirable	Discard if no information on sample tube
Requesting person and place to report.	Desirable	Process and identify requesting clinician/GP, e.g. by contact requesting hospital/GP

Any samples that are not labelled in accordance with the requirements detailed above will be discarded, with the following exceptions:

In the case of individuals whose identity must remain confidential (e.g. GUM patients, research study patients, bone marrow registry donors) samples do not need to meet the requirements listed above but must be labelled with a unique identification number.

In cases of clinical urgency or where it is not possible to obtain repeat samples (i.e. deceased donor samples), the laboratory may agree to process a sample referred without all requested identifiers in accordance with locally documented procedures (copy available on request).

In some cases, prior to processing the sample, the laboratory will need verbal/written clarification that the referring clinician will accept responsibility for processing the requests.

Results will only be released to requesting clinician (or healthcare professional),and will not be released to patients/carers.

Generated reports will clearly state which identifiers were missing and whether these were from the sample, the request form or both. It is the responsibility of the clinician receiving the report to check the identifiers given to confirm this is the correct patient/donor.

Urgent testing of samples must be by prior arrangement by contacting the laboratory.

Please note, all materials used during the collection process should be disposed of as per local Trust or Health Board guidance.

Additional requests

Requests for additional services can be made using standard haematology forms. The forms must clearly state the required investigation and referral laboratory in addition to the information required below.

For fetal RhD screening, the specific antenatal request card must be used.

The Transplantation Laboratories are category B laboratories and therefore samples that fall within this classification can be sent to and tested within the laboratory. Known high risk samples must be labelled as such on both the sample tubes and request form.

Please ensure that samples for each patient are packaged separately, with their respective request form, and that packaging conforms with the requirements detailed below.

Labelling

The following information is required on each **sample tube**:

Details (Please note patient details on sample & request form must match exactly)	Requirement	Outcome if missing from request form
Patient name (Surname & Forename)	Mandatory	Discard
Date of birth	Mandatory	Discard
Hospital number/NHS number	Mandatory	Discard

Please note, all samples for platelet immunology testing must be handwritten in accordance with the requirements for the acceptance of pre transfusion samples in Wales.

7. Consent and identification

Please note that it is the responsibility of the requestor to obtain informed consent and verify identification of the patient for the requested tests. Surplus material may also be stored for further diagnostic testing to benefit the individual and anonymously for quality control, education and training and approved research and development.

8. Confidentiality

The laboratories have access to the data and information needed to provide a service that meets the needs and requirements of internal and external customers. The laboratory information system (whether computerised or paper-based) provides for the collection, processing, recording, storage, and retrieval of data, and has documented procedures in place to ensure the confidentiality of patient information and the security of the data during each step of the process. WBS laboratories comply with the General Data Protection Regulation (GDPR) (Regulation (EU) 2016/679) and the Caldicott principles.

9. Important Factors That May Affect Histocompatibility & Immunogenetics (H&I) Tests

The following factors may affect any of the H & I tests that are performed by the Laboratory:

Factors that may affect testing	How to minimise effects
Sample storage and transportation temperature	Store and transport samples at ambient temperature.
Sample storage time	Ensure samples arrive within 24 hours of collection.
Anticoagulant	Ensure correct blood tubes are used - see the 'Standard Tests and Samples Required' Table
Immunosuppressants, e.g. Rituximab, IVIg	Note medication/treatments on sample request form
Low white cell count	Note low count on sample request form

Standard Tests and Samples Required

Testing Required		Samples Required		
		EDTA	Clotted	Other
Renal/ Pancreas Recipient HLA-typing	New patients & re-type samples	10ml 6ml	12ml	-
Virtual Crossmatch – Live	Recipient	10ml 6ml	12ml	-
	Donor	10ml 6ml	-	-
Full Crossmatch – Live	Recipient	30ml 6ml	12ml	-
	Donor	30ml 6ml	-	-
Deceased donor testing	Donor - typing	12ml	-	Lymph/Spleen
	Donor – crossmatching	20ml	-	
Renal Antibody Monitoring Samples	Routine Transplant list Post Transplant Rejection HLA incompatible	-	12ml	-
ABO Titres	Recipient/Donor	6ml	-	-
HSCT	Recipient	20ml 6ml	10ml	-
	Family member	10-20ml	-	-
	HLA-B27 typing	5-10ml	-	-

Disease Association Investigations	Miscellaneous e.g. Coeliac disease, Abacavir sensitivity	5-10ml	-	-
Platelet Investigations	Refractory PTP	40ml	10ml	-
	NAIT - Mother	10-20ml	10ml	-
	NAIT - Partner	10-20ml	-	-
Transfusion investigations	TRALI investigation	2 x 6ml	2 x 6ml	-
	Transfusion reaction	-	6ml	-
Fetal RhD Screening	Maternal peripheral blood	10ml	-	-

*Please contact Transplantation Laboratories for sample requirements relating to paediatric patients

10. Additional Samples and Testing

Transplantation Laboratories also offer a range of other investigations for patients by request. For further information please contact Deborah Pritchard, Head of Transplantation Services.

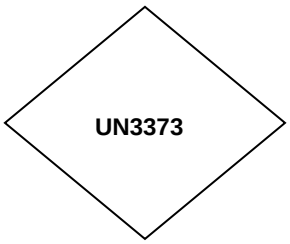
Additional testing that may be requested on an existing sample received by the laboratory must be discussed with the relevant departmental lead. See Section 1 for contact details.

11. Blood Sample Transportation

Following collection of samples according to local trust/health board policies and procedures, blood samples should normally be transported at ambient temperature and delivered to the Laboratory in as timely a manner as possible, and wherever possible within 24 hours of collection. Please contact the laboratory for advice if samples cannot be sent within this time-frame.

Please note that sample transport is the responsibility of the test requestor. The requestor must assure themselves that their transport mode is effective and appropriate to meet the above requirements.

All specimens must be packaged in accordance with the current UN3373 Sample Transport Compliance Regulations to prevent breakage or spillage in transit. They must be, clearly labelled in accordance with the regulations and display the hazard diamond as illustrated below.



In addition, the statement '**BIOLOGICAL SUBSTANCE CATEGORY B'** must be noted on the packaging. Packaging materials must be suitable for transporting UN3373 Biological substance Category B samples and clearly addressed to:

Transplantation Laboratories
Welsh Blood Service
Ely Valley Rd,
Pontyclun
CF72 9WB

For other arrangements please contact the Laboratory for help and advice.

12. Procedures for Specimen Handling

Our procedures for handling blood samples and other specimens are available on request.

13. Turnaround Times

The following target response times relate to working days, where applicable, after the sample/request is received at the laboratory. If requested, a copy of the report can be faxed or emailed as soon as it is available.

For urgent requests please contact the laboratory to arrange testing and reporting arrangements.

	Report (written unless otherwise stated)	**Operational Lead**	**Target Turnaround Times (90% within)**
Renal Transplantation	Live Donor Work-up	Head of Solid Organ Transplantation Services	10 working days
	Urgent Live Donor Testing		Contact laboratory to discuss requirements
	Deceased donor HLA Typing	Head of Stem Cell Transplantation Services	4 hours (90% target)
	Deceased donor crossmatching	Head of Solid Organ Transplantation Services	4 hours (80% target)
	Recipient HLA typing		10 working days

Haematopoietic Stem Cell Transplantation	Potential family donor work-up-matched donors	Head of Stem Cell Transplantation Services	10 working days from receipt of last sample
	Potential family donor work-up no matched donors		10 working days from receipt of last sample
	Unrelated donor work-up		10 working days from receipt of last sample
Platelet Immunology	Refractory investigation	Head of Solid Organ Transplantation Services	Verbal - 1 working day
			Written – 7 working days
	NAIT investigation		5 working days (10 working days if expanded panel testing required)
	Platelet antibody investigations (including PTP)		10 working days
Additional Services	Low resolution HLA type	Head of Stem Cell Transplantation Services	5 working days
	HLA-B27 typing	Head of Stem Cell Transplantation Services	10 working days
	HLA antibody investigations - urgent	Head of Solid Organ Transplantation Services	1 working day
	Day of transplant HLA antibody report		10 working days
	Fetal RhD screening	Head of Stem Cell Transplantation Services	10 working days

The requestor will be notified when an examination is delayed that could compromise patient care.
Please note, when successive testing or testing of multiple individuals is required, e.g. typing of potential haemopoietic stem cell transplant donors, the turnaround times may not apply.
The Laboratory issues results to Clinicians only, and not to patients/carers. Requests from patients/carers will not be fulfilled.

14. Compliments and Concerns

If you have any complaints, or suggestions for improvements, contact can be made:

- Via telephone on 01443 622175;
- via e-mail using Deborah.Pritchard3@wales.nhs.uk;

- in writing to Deborah Pritchard, Head of Transplantation Services, Welsh Blood Service, Ely Valley Road, Talbot Green, Pontyclun, CF72 9WB.
- or via one of the Transplantation operational leads (detailed in Section 1)

Although we are committed to providing our customers with an excellent service there may be times when we fail to fully meet your requirements. As a learning organisation we firmly believe that every complaint is to be valued as an opportunity to discuss with our customers how the service can be improved. Complaints will be processed in line with the Duty of Candour regulations, where appropriate.

Acknowledgement of the concern will be made and will indicate the probable timescale of the investigation. On completion of the investigation, the complainant will be notified in writing of the result of the investigation and any corrective actions that have been instigated.

15. Terms and Conditions

The WBS standard terms and conditions of service shall apply where a Service Level Agreement has not been signed between Transplantation Laboratories and the customer. A full set are available on request, the key elements of which are detailed below:

Invoicing
Velindre University NHS Trust shall invoice the customer for work undertaken on completion of the individual task, where appropriate. Payment due within 30 days of receipt of invoice.

Any Debts not paid within 30 days shall be considered to be outstanding and may incur interest charges at a daily rate of 0.05%, of the outstanding balance, until the amount has been settled.

Method of payment
The general method of payment shall be in accordance with current policy as stated on the reverse of the Invoice. No receipt will be given unless specifically requested.

Any customer requesting a service supplied by Transplantation Laboratories shall be deemed to agree to these terms and conditions.

16. Quality Assurance

The Transplantation laboratories are committed to effective quality management at every level. All work is undertaken within the framework of a documented quality system and according to good laboratory and good manufacturing practice (GLP and GMP respectively).

Techniques and procedures are validated, described in standard operating procedures (SOPs), and conducted by staff whose proficiency is regularly assessed. Where relevant, measurement of uncertainty is calculated for examinations. Further information is available on request.

An internal audit schedule is an integral part of the Quality Management System (QMS) and supports current external licensing and accreditation inspections by the Medicines and Healthcare products

Regulatory Agency (MHRA), United Kingdom Accreditation Services (UKAS) and other relevant accreditation bodies.

Transplantation Services is committed to continuously improving the quality and range of services provided and welcomes any comments or suggestions from our users. A questionnaire is distributed every 2 years as a minimum to monitor customer opinions.

17. Quality Assessment

Standards of testing are maintained by the rigorous use of internal quality assurance protocols and through participation in appropriate External Quality Assessment Services (e.g. UK NEQAS) and International Exchange Schemes.

A copy of last year's participation certificate and results summary is available, on request, for each of the following:

- **UK NEQAS for Histocompatibility and Immunogenetics**
- **University of California, Los Angeles International HLA DNA Exchange**
- **UK NEQAS for Blood Transfusion Practice**
- **INSTAND**

18. Accreditation and Regulation

Transplantation Laboratories is a UKAS accredited Medical laboratory No.9326 (ISO 15189:2022). The full schedule of accreditation may be viewed at UKAS Schedule of Accreditation.

The laboratory is also accredited by the European Federation for Immunogenetics (EFI). See https://efi-web.org/accreditation/efi-accredited-laboratories for details.

Accreditation categories and techniques covered are available on request.

Transplantation Laboratories, as part of the WBS, is also regulated by the Medicines and Healthcare Products Regulatory Agency (MHRA) License Holder WDA(H)17853.

19. Further information

More detailed information on any of these services can be obtained, on request.

Please contact Deborah Pritchard, or one of the Operational Leads (see Section 1).