Manchester University
NHS Foundation Trust

03-GB-009.991

7878

A Guide to the Histocompatibility and Immunogenetics Services Provided for Wythenshawe Cardiothoracic Transplant Unit

This guide outlines the Histocompatibility and Immunogenetics (H&I) services provided by the Transplantation Laboratory, Manchester Royal Infirmary in support of the Wythenshawe Cardiothoracic Transplant Unit Programme. The guide is of use to clinical and support staff in the cardiothoracic transplant unit.

**Revised by Marie Hampson, Steven Jervis, Dr Judith Worthington,
Stephen Sheldon & Julie Johnson.**

Valid until Dec 2025

Contents

1. Introduction

The Transplantation Laboratory is a regional specialty pathology service and as such offers a wide range of high quality, efficient and cost-effective services using state of the art technologies to Manchester University NHS Foundation Trust, other regional Trusts, and healthcare providers. The main services provided by the Transplantation Laboratory are described below:

a) Solid Organ Transplantation

The laboratory provides H&I support for:

- Kidney, kidney and pancreas, pancreas and islet cell transplantation programmes at Manchester Royal Infirmary.
- Cardiothoracic organ transplantation at Wythenshawe Hospital
- There is a 24 hour on-call service for kidney / kidney and pancreas / pancreas only / islet transplantation and all thoracic organ transplants.

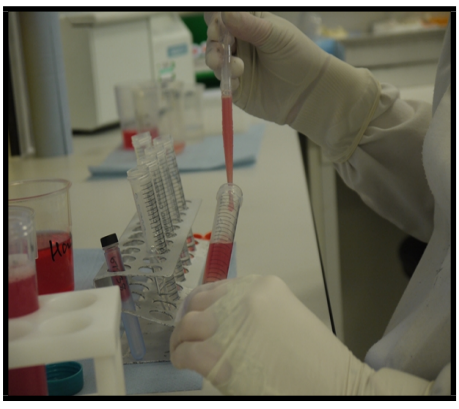
b) Haematopoietic Progenitor Stem Cell Transplantation

The Transplantation Laboratory provides Histocompatibility & Immunogenetics (H&I) support for the haematopoietic progenitor stem cell transplantation programmes at Manchester University NHS Foundation Trust (MRI and RMCH) and other regional trusts. The laboratory utilises state of the art molecular HLA typing technologies for patients and their potential donors who may need a stem cell or bone marrow transplant. The laboratory is one of the leading laboratories in the country in the application of chimaerism monitoring using short tandem repeats post progenitor stem cell transplantation. The laboratory provides additional KIR typing and interpretation of results for haploidentical stem cell transplants.

The laboratory offers a rapid and professional Graft Information and Advisory Service (GIAS) to undertake donor selection. This service is delivered by highly qualified and experienced HCPC registered staff and is led by an RCPath qualified H&I Clinical Advice Scientist.

c) Immunogenetics testing

The Transplantation Laboratory provides testing to support disease diagnosis and management for the Manchester University NHS Foundation Trust, Primary Care Centres and hospitals. A range of tests is provided, including HLA-B*27 and HLA-B*57:01 determination and HLA typing to support the diagnosis of Actinic Prurigo, Uveitis, Birdshot Retinopathy, Narcolepsy and Coeliac Disease. On request the laboratory can perform additional HLA typing to aid disease diagnosis and drug hypersensitivity investigations for tests in addition to those outlined.



d) Research and Innovation

The Transplantation Laboratory participates in research and innovation relevant to the clinical services provided to ensure that we continually improve our service provision in line with the current clinical evidence base. Projects are closely tailored to local clinical practice to ensure the most appropriate services are provided for the patients.

The Transplantation Laboratory is part of a network, which is cross-directorate and is known as the Manchester Institute of Nephrology and Transplantation (MINT). MINT is a multi-professional body of physicians, surgeons, nursing staff, scientists, other professions allied to medicine and managers. Its aim is to improve and develop the research and educational activities of the transplantation, nephrology and dialysis services to achieve the best possible care for transplant patients.

e) Audit

The Transplantation Laboratory is actively involved in audit related to laboratory activities as well as clinical audit in conjunction with the services we support. The process of clinical audit directly relates to the Trust's Clinical Effectiveness Strategy that aims to improve the quality and outcome of patient care. The laboratory also has an internal audit cycle against ISO 15189:2012 and European Federation for Immunogenetics standards to ensure continual compliance and continual improvement in transplant outcome.

f) Quality assurance

The Transplantation Laboratory is a UKAS accredited medical laboratory No.7878 and has European Federation of Immunogenetics accreditation (EFI No: 03-GB-009.991).

The laboratory has a well-established quality management system in operation which allows the laboratory to be focused on continual improvement in line with needs and requirements of our users. The QMS provides a structured framework for the laboratory and is monitored and maintained by the Laboratory Operations and Quality Manager. The Quality Policy which is reviewed annually describes the aims of the services.

Any test performed in the laboratory is subject to a variety of factors that may influence the outcome of the result. Some of these factors include the sample itself, the test method, reagents used and different operators carrying out the same process. Variations can also be caused by procedures that involve the measurement of analytes and reagents whereby environmental factors such as temperature and humidity may affect results. Any equipment used in the process will further introduce the opportunity for variation. To provide a measure of confidence in results produced it is necessary to identify all factors which may contribute to variation in a process and assess their potential to influence uncertainty. Once identified these factors must be reduced or controlled to an acceptable level and a value for the range of acceptable uncertainty assigned where possible.

The Transplantation Laboratory has chosen, where possible, to utilise internal Quality Control material and data to establish Uncertainty of Measurement where applicable. Upon request the laboratory shall make its estimates of measurement of uncertainty available to laboratory users.

Participation in external quality assurance programmes such as UK NEQAS and UCLA schemes, together with continual internal quality assessment monitoring of our tests, ensures that the laboratory's high-quality standards are maintained.

UK NEQAS schemes conform to high standards of professionalism, impartiality, clinical relevance and strict financial accountability across all disciplines and specialities, so that all concerned with the quality of laboratory investigations may have confidence in the service provided.

A highly experienced consultant team offers support to clinicians and service users 24 hours a day, seven days a week. The team provides information related to using the service, interpretation of test results and clinical advice. Reviews and changes to the service provision will be in consultation with our users and will be clearly defined in revised Service Level Agreements (SLAs), where applicable.

The Transplantation Laboratory actively supports and encourages staff training and continual professional development. It is recognised by both the Royal College of Pathologists, the National School for Healthcare Scientists and the British Society for Histocompatibility and Immunogenetics as a training laboratory in Histocompatibility and Immunogenetics. Where appropriate, staff members are registered with the Health and Care Professions Council (HCPC).

Details of our accreditation, including current certificates and performance data, are available upon request from the Laboratory Operations and Quality Manager (julie.johnson2@mft.nhs.uk).

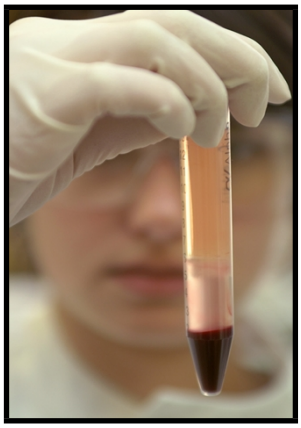
In order to help us improve our service, you may be asked to complete a questionnaire. We greatly appreciate and value your input and would like to thank you for your assistance and suggestions.

g) Complaint Procedure

The Transplantation Laboratory is continually aware of and takes into consideration the requirements of its users and staff, whilst striving to create the best standards of professional care. According to Trust policy, any complainants are referred to the Patient Advice and Liaison Service (PALS) who can support staff and patients to achieve speedy solutions. Also, complaints can be directed to the Laboratory Director, a Clinical Advice Scientist or any Transplantation Laboratory representatives at Multidisciplinary Team meetings. Please make any concerns you have about the quality of the service known to us as soon as possible; we take your complaints seriously.

Any suggestions from users regarding any aspect of our service provision, or indeed how the User Guide could be improved, are very welcome. Please forward any suggestions to the Laboratory Operations & Quality Manager (julie.johnson2@mft.nhs.uk).

h) Clinical Liaison and Advice



A Clinical Advice Scientist or deputy will always be available to attend multi-disciplinary team meetings as required in order to ensure optimum communication between the laboratory and clinical teams and provide advice relating to Cardiothoracic Transplant Service provision.

An experienced consultant team offers support to clinicians and service users 24 hours a day, seven days a week. The team provides interpretation of test results and clinical advice.
A 24-hour, 365-day on-call service is provided for deceased donor HLA typing and crossmatching and a Clinical Advice Scientist is always available for the provision of advice.

I) Confidentiality and Personal Information

The Transplantation Laboratory adheres to Manchester University NHS Foundation Trust's policies on data protection and disclosure.

2. General Information

2.1 Postal Address

Transplantation Laboratory
2nd Floor, Purple Zone
Manchester Royal Infirmary
Oxford Road
Manchester
M13 9WL
Tel: 0161 276 6397
Fax: 0161 276 6148

2.2 Business Hours

Opening Hours for routine work:	08.30 – 17.00 hrs
Out of hours, weekends, and Bank holidays:	On call staff & Clinical Advice Scientist can be paged via MFT switch Tel: 0161 276 1234

2.3 Laboratory Key Personnel

Laboratory Director

Prof Kay Poulton PhD, FRCPath
Consultant Clinical Scientist,
0161 276 6397
Email: kay.poulton@mft.nhs.uk

Consultant Clinical Scientists

Mr Stephen Sheldon, FRCPath
Tel: 0161 276 6397
Email: stephen.sheldon@mft.nhs.uk

Ms Natalia Diaz Burlinson, FRCPath
Tel: 0161 276 6397
Email: Natalia.DiazBurlinson@mft.nhs.uk

Dr Helena Lee PhD, FRCPath
Tel: 0161 276 6323
Email: helena.lee@mft.nhs.uk

Cardiothoracic Support Services Enquires

Dr Judith Worthington PhD FRCPath
Principal Clinical Scientist
0161 276 7988
Email: judith.worthington@mft.nhs.uk

Mr Steven Jervis DipRCPath
Senior Clinical Scientist
0161 276 6656
Email: steven.jervis@mft.nhs.uk

Mrs Marie Hampson DipRCPath
Senior Clinical Scientist
0161 276 7919
Email: marie.hampson@mft.nhs.uk

Mr Patrick Flynn DipRCPath
Senior Clinical Scientist
0161 276 6651
Email: Patrick.flynn@mft.nhs.uk

Laboratory Operations and Quality Manager

Julie Johnson MSc. HCPC
Principal Clinical Scientist
0161 276 6424
Email: julie.johnson2@mft.nhs.uk

General Enquiries

Business and Administration Manager
Judith Spencer
Tel: 0161 276 6397
Fax: 0161 276 6148
Email: judith.spencer@mft.nhs.uk

2.4 Essential Telephone Numbers

Specimen Reception:	0161 276 6471
Admin office:	0161 276 6397
Histocompatibility Team – General Enquiries	0161 276 6656 / 7919 / 6651 / 7988

2.5 Essential Email Addresses

Cardiothoracic Patient Listings: **OnCall.TLab@mft.nhs.uk**

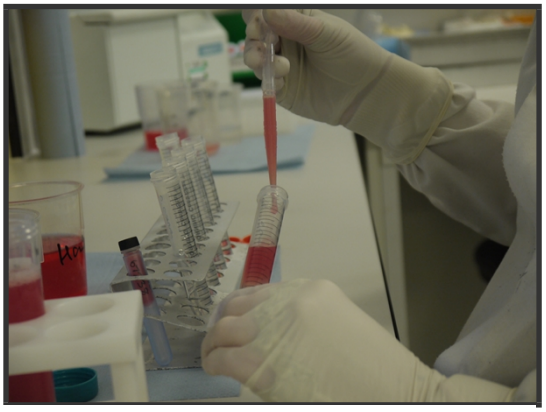
Solid Organ Enquiries: **cmm-tr.Histocompatibility@nhs.net**

2.6 Internet page

https://mft.nhs.uk/mri/services/transplantation-laboratory

3. Use of the Laboratory

3.1 Service Availability



The laboratory is open for receipt of routine specimens from 08:30 to 17:00 between Monday to Friday. Internal on-site samples may be sent directly to the laboratory using the pneumatic pod system (Transplantation Pod No 805).

There is an on-call service provision available outside of normal working hours provided by an on-call team consisting of a HCPC registered Clinical Scientist, a technologist and a Clinical Advice Scientist. This service is generally restricted to the solid organ transplant programme.

The on-call team can be contacted through the paging service provided by PageOne (**Page One User Guide (**www.pageone.co.uk/support/downloads) or directly via the MRI Switchboard (0161 276 1234).

3.2 Labelling of sample containers

The Transplantation Laboratory will make every effort to ensure requests are processed in a safe and timely manner, but it is essential that request forms and samples are labelled appropriately and legibly. The minimum acceptance criteria for request are normally **3 key identifiers** that should include at least:

- Patient's name (forename and surname)
- Date of birth
- Hospital number and or MRI District number
- NHS number
- Home Address of the patient.

These are all identifiers specific to the patient which help us to confirm identity and are essential.

It is also important to clearly identify the investigations required when completing the request card, please only select the test required and send only the appropriate sample tube.

If you have any concerns regarding this, please ring 0161 276 6471 / 6397 for further advice.

Specimens will not be accepted for analysis if:

- There are insufficient unique identifiers for the patient as specified.
- Incorrect sample type or tube
- Incorrect transportation conditions mean that the sample is not viable for testing
- Sample is received in a hazardous condition e.g. leaking or sharps attached.
- Mismatch of details between the form and sample(s)
- The information provided is illegible

Samples that fail to meet the above criteria will be discarded as unsuitable for analysis, and the sender will be informed. The only exception to this is for patients whose identity is anonymous and they have their own unique identifier, for example patient samples from Genitourinary Medical Centres or potential stem cell donors. In other circumstances samples may be accepted without the 3 key identifiers at the discretion of the laboratory.

3.3 Transportation of routine samples to the laboratory

All users are advised to refer to P650 Packaging Instruction which applies to UN No. 3373 (Diagnostic Specimens) for information on the correct procedures for packaging and transporting samples. When sending samples to the laboratory it is important to follow the correct courier and postal procedures and ensure the specimens are appropriately packaged. (**See appendix 8.1**)

All specimens should be transported at room temperature **(22^{o}C - 25^{o}C)**, unless otherwise instructed, avoiding where possible prolonged over exposure to heat. The samples should be transported directly to the laboratory as quickly as possible after collection to maintain the integrity of the sample and avoid compromising the results.

Internal on-site specimens may be transported directly to the Transplantation laboratory via the porter's rounds during the normal working day or by pneumatic pod system to Pod No 805. Samples should be placed in a specimen bag with the request for transportation around the trust.

Please contact the laboratory on 0161 276 6471 / 6397 if there are specific questions regarding transportation of specimens.

3.4 Urgent samples

If a result is required urgently and the sample will arrive during working hours the laboratory MUST be notified by telephone so that we can prioritise your request.

All samples should be packaged and transported as above. If you need to submit a sample out of normal working hours for testing on-call please contact the Clinical Scientist on-call via the hospital switchboard (0161 276 1234) or via the paging service provided by PageOne.

Page One User Guide **(**www.pageone.co.uk/support/downloads).

3.5 Acceptance time limit after sample drawing

Time limits and storage temperature requirements are imposed to maintain the integrity of the sample, to ensure accuracy and reliability of the testing and reduce the need for repeat samples. For all tests complete the request card or an ICE request for users within MFT. See **Appendix 8.2** for a full list of test and samples required

HLA genotyping

Whole blood – minimum 3 ml EDTA blood	**No time limit**
Buccal swab*	**No time limit**
Dried Blood Spot*	**No time limit**

(See pages 25-26 for instruction on how to take buccal swabs/ dried blood spot samples).

HLA antibody/ donor specific antibody (DSA) testing

5 ml Clotted blood (no anticoagulant)	**Up to 48 hours**

Crossmatching

EDTA/ Heparinised blood for crossmatching	**Up to 24 hours**

Storage conditions prior to sending

Clotted samples can be kept overnight at 4^{o}C but sent immediately the next morning to the laboratory for testing.

EDTA blood samples should be kept at room temperature whilst waiting and during transport to the laboratory, avoiding any excessive heat exposure.

Request cards can be obtained from the Business and Administration Manager, please call on 0161 276 6397 or email *judith.spencer@mft.nhs.uk*. These request cards are also available in electronic format upon request.

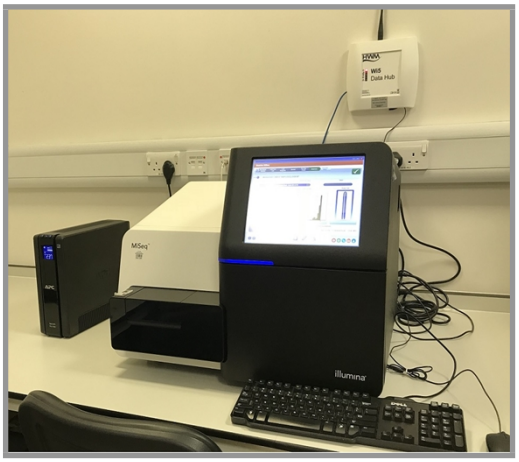
*see appendix for instructions of how to obtain sample

4. General Information regarding services available

4.1 Descriptions of standard tests

What is HLA typing (Tissue Typing)?
HLA typing is performed predominantly to match a donor and recipient for solid organ or haemopoietic progenitor stem cell transplantation (HPCT). Minimising the number of HLA mismatches between donor and recipient maximises the opportunity for optimal transplant survival.

HLA molecules are crucial to normal immune processes enabling the cells involved in immune responses to recognise foreign organisms and react against them. Following transplantation, the recipient's immune cells can recognise donor HLA molecules as foreign and react against them causing rejection.



HLA typing refers to the series of laboratory tests whereby the HLA molecules expressed on the surface of an individual's body cells are identified. HLA molecules are on the surface of all nucleated cells (i.e in humans, all cells apart from red blood cells) but lymphocytes are routinely used for tests because they can easily be isolated from anti-coagulated peripheral blood.

The technique used for testing varies according to the clinical requirement. For example, a rapid, but intermediate resolution technique (LinkSēq™) may be used to HLA type a potential donor in a solid organ transplant setting.

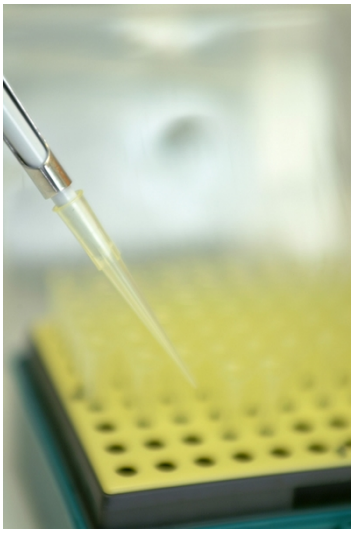
LABType® SSO offers intermediate level resolution and facilitates rapid batch testing of samples for routine testing. All routine cardiothoracic patients also receive extensive next generation sequence (NGS) based typing analysis to provide a high-resolution HLA type. The Transplantation Laboratory employs state of the art NGS HLA typing technology to assist in interpretation of HLA antibodies and thus permit suitable donor identification.

An individual's HLA type defines the combination of HLA molecules on the surface of their body cells. These are determined genetically. The HLA genes are unique in the human genome because of their considerable variability, which results in many different HLA types. The HLA genes are identified by letters (e.g. HLA-A) and the different gene products (specificities) by numbers (e.g. HLA-A2). Each individual inherits one set of HLA molecules from each parent thus they have two HLA-A, two HLA-B types and so on.

What is Antibody Screening?

Individuals can produce antibodies directed against HLA specificities that they do not possess. This can happen following exposure to non-self HLA during pregnancy, blood transfusion or transplantation. These antibodies are detected in serum and can potentially react with a donor organ or graft and cause transplant rejection.

It is important that cardiothoracic patients are screened for the presence of HLA antibodies and that the specificities of any antibodies detected are defined prior to transplantation. **Samples for antibody screening should be sent to the laboratory every three months from patients on the transplant list and approximately two weeks after a known sensitising events (e.g. blood transfusions).** When a patient is known to have antibodies against a particular HLA specificity, that specificity is listed as an unacceptable donor antigen. When donor directed HLA specific antibodies are identified, the level of risk associated with proceeding to transplant can be assessed on request.



HLA specific antibodies are detected and defined by microbead array techniques, which are highly sensitive and specific. They are referred to as Luminex assays and are semi quantitative.

For sensitised parous female patients being listed for transplantation, it is our policy to HLA type the father of the children or the children themselves in order to fully define pregnancy-related sensitisation.

Some patients have non-graft damaging “autoantibodies” that can cause false positive donor crossmatches, the laboratory will request samples to specifically test for these as necessary.

What is Crossmatching?

Crossmatching is an assay in which donor lymphocytes are tested against serum samples from the potential recipient(s) to ascertain whether any donor–reactive antibodies are present that would cause transplant rejection. Donor-reactive antibodies that cause a positive crossmatch test are normally a contraindication to transplantation.

- The *cytotoxic crossmatch* is a cell killing test. It is carried out for all potential recipients.
- The *flow cytometry crossmatch is* a more sensitive test that uses fluorescence to detect antibody binding to donor cells and is used for “high-risk” sensitised recipients.

Virtual Crossmatching

A "*virtual crossmatch*" assessment is performed pre-transplant for solid organ patients, whereby the donor HLA type is reviewed against the patient's HLA antibody profile (antibodies are listed as unacceptable antigens) to determine whether the patient has any donor-directed antibodies that could cause a positive crossmatch test result. In cases where all unacceptable antigens have been clearly defined, sensitised patients can be transplanted without the need for a prospective crossmatch as long as all unacceptable antigens are absent from the donor HLA type. This is the basis of the current solid organ allocation process in the UK and the low frequency of unexpected positive crossmatches for patients suggests that patients who have been rigorously assessed and defined as negative for donor-directed antibodies can safely proceed to transplant before the crossmatch test result is available. The purpose of this approach is to reduce the cold ischaemia time without compromising the safety of transplantation. For these recipients the crossmatch test is performed retrospectively.

Potential recipients will be assessed for their suitability for a "virtual crossmatch" against a particular donor and a "virtual crossmatch negative" report issued if appropriate. The transplant unit must be able to confirm that no potential sensitisation events have occurred since the date of the last patient serum sample tested for HLA antibodies.

Risk Assessment Crossmatching

For cardiothoracic patients with donor-directed antibodies, a pre-transplant "*risk assessment crossmatch*" can be performed on request. In a "risk assessment crossmatch" the level of risk associated with proceeding to transplant with a particular donor is determined by calculating the cumulative mean fluorescence intensity (MFI) value of all the donor-directed antibodies as detected by Luminex bead assays following national CTAG guidelines.

It must be noted that the validity of virtual and risk assessment crossmatch results for antibody positive patients is dependent on the donor HLA type being correct. In 2017, nationally, discrepancies were detected in 0.5% of donor HLA types after the organs had been allocated.

A flow diagram outlining the process used to provide a virtual crossmatch result and a risk assessment can be seen in (**Appendix 6)**. All crossmatch results are reported to the on-call Transplant Co-ordinator verbally and in writing by encrypted email. **No patient should be transplanted without first contacting the Laboratory or, out of hours, the on-call scientist to confirm the suitability of the donor.**

Post-transplant Donor Specific Antibody (DSA) testing

Post-transplant recipients can produce specific antibodies associated with transplant rejection. A post-transplant antibody monitoring service is available, including testing for donor-directed antibodies. Post-transplant samples for DSA testing to support the diagnosis of rejection should be sent when clinically indicated to support the diagnosis of antibody-mediated rejection.

5. Requesting Tests/Samples Required

The Transplantation Laboratory has its own distinctive request cards which can be obtained from the departmental business and administration manager (judith.spencer@mft.nhs.uk). Internal on-site requests should be made using the ICE system using the Transplantation Laboratory tab following the MFT standard operating procedure. All other requests should be made using the request cards shown below following the procedure described below for the test required.

Patient Test Requests

As a minimum requirement, include patient surname, forename, date of birth, hospital number/MRI district number, referring hospital, diagnosis, consultant, person requesting the test and the date sample taken. It is essential that the patient is clearly identified on the card and on the specimen. Please tick the relevant tests required box or state clearly what testing is required.

Cardiothoracic Patient 1st set of bloods:

- 5ml EDTA blood for initial HLA typing
- 10ml clotted blood for antibody screening

Cardiothoracic Patient 2nd set of bloods:

- 5ml EDTA blood for verification HLA typing
- 10ml clotted blood for antibody screening

Cardiothoracic Patient antibody testing:

- 10ml clotted blood for antibody screening

Cardiothoracic Patient Post-transplant DSA testing:

- 10ml clotted blood for antibody screening

Deceased organ donor crossmatching samples

Donor spleen and lymph node specimens for retrospective crossmatching should be taken by the retrieval surgical team and sent by taxi to the Transplantation Laboratory the next working day. A time of transplant 10ml clotted blood sample from the recipient should be sent at the same time as the donor material.

For urgent prospective deceased donor crossmatching, the 1st On Call scientist must be notified of when to expect samples. The 1st On Call can be contacted through the paging service provided by PageOne or directly via MRI Switchboard (0161 276 1234).

Page One User Guide **(**www.pageone.co.uk/support/downloads)

Current Cardiothoracic Request card

NHS
Manchester University
NHS Foundation Trust

THE TRANSPLANTATION LABORATORY, MANCHESTER ROYAL INFIRMARY
TEL: 0161 276 6397 FAX: 0161 276 6148

SURNAME*	FORENAME*	DATE OF BIRTH*	SEX	HOSPITAL*

HOSPITAL NUMBER*	NHS NUMBER	REQUESTED BY* (BLOCK CAPITALS)	CONSULTANT*

NHS PATIENT	BLOOD GROUP	BLOOD TRANSFUSION	No. PREGNANCIES	SAMPLE DRAW DATE*
☐ Yes ☐ No	ABO ____ Rh ____	Date ____ No. Units ____		

DIAGNOSIS*

RECIPIENT ☐ HEART ☐ SINGLE L ☐ DOUBLE L ☐ H+L **DONOR** ☐ **OTHER** ☐

TESTS REQUIRED*

RECIPIENT HLA TYPING
☐ HLA TYPING (5ml EDTA)

URGENT THORACIC ASSESSMENT
☐ SEND 10ml EDTA BLOOD + 10ml CLOTTED BLOOD

CYTOTOXIC ANTIBODIES
☐ SEND 10ml CLOTTED BLOOD

DONOR PROSPECTIVE CROSSMATCH
☐ SEND 40ml EDTA

PREGNANCY RELATED UNACCEPTABLE ANTIGENS
☐ 5ml EDTA

RECIPIENT_____

POST-TPX DONOR SPECIFIC ANTIBODIES
☐ 10ml CLOTTED BLOOD

*** ESSENTIAL INFORMATION REQUIRED IN ORDER TO PROCESS REQUEST**

FOR LABORATORY USE ONLY

DATE	CELL NO.	DNA NO.	SERUM NO.	PATIENT NO.

Samples booked in by_____

☐ **HLA TYPING** **Reviewed By**_____

☐ HLA-A ☐ HLA-B ☐ HLA-C

☐ HLA-DRB1 ☐ HLA-DRB3/4/5 ☐ HLA-DQB1 ☐ HLA-DPB1

Request on HLA Typing Database By_____

Request on the Serum Screening Database By_____

CELLS: FROZEN / DISCARDED BY_____

DISPOSAL OF RESIDUAL DONOR MATERIAL

☐ XM Material By_____ Date_____ ☐ Additional Material By_____ Date_____

CM11762

THORACIC ORGAN TRANSPLANT REQUEST

6. Reporting of Results

To maintain patient confidentiality and comply with General Data Protection Regulations (GDPR) and other legal requirements all results are reported via encrypted email or in writing only to an authorised individual. They are signed by a Clinical Advice Scientist or named deputy. Other results are only reported by telephone after agreement by a Clinical Advice Scientist. Provision of non-urgent results by fax is available on request during office hours and Clinical Advice Scientist advice is available on a 24hr basis. Users shall be informed of deviations from agreement that may impact on results. The Measurement of uncertainty (MoU) shall be considered for all examinations which include a measurement step where it has influence on the reported result. Estimates of the MoU will be made available to users upon request.

All times are quoted as working days from the receipt of the sample in the Transplantation Laboratory

Intermediate level HLA typing of patients:	**10 days**
High resolution HLA typing of patients:	**10 days**
HLA antibody screening of patients:	**10 days**
HLA antibody specificity testing of positive patients:	**15 days**
Donor specific antibody (DSA) testing:	**7 days**
Urgent HLA antibody screening of patients requiring urgent listing:	**5 hours***
Urgent Donor Specific Antibody (DSA) testing:	**5 hours***
Urgent deceased donor/ recipient crossmatching:	**6 hours***
Retrospective crossmatching	**3 days of transplant**

Turnaround Time (TAT)
Over 90% of the results are reported within **the specified working days** from receipt of samples. Special arrangements requiring a shorter turnaround may be established on a user-specific basis, by arrangement with the Clinical Advice Scientist and subject to the technical limitations of the assays. The turnaround times quoted are supported by audit data.

*Testing of clinically urgent samples via prior arrangement with the laboratory only.

7. Standards

All aspects of the services provided for the cardiothoracic transplant programme are compliant with the relevant standards/guideline.

BSHI BTS: Guidelines for the detection and characterisation of clinically relevant antibodies in allotransplantation. July 2015

HLA specific antibodies in cardiothoracic transplantation: Standardisation of testing, reporting and crossmatch protocols in the UK. (CTAG (13) S9) Sensitised Patients. September 2013.

Standards for Histocompatibility Testing Version 8.0 European Federation for Immunogenetics (EFI). January 2020

8. Appendix

8.1 Requirements for sending specimens by post

In order to comply with UN code number UN3373 there should be three layers of packaging.

1. The primary container containing the specimen

2. Secondary packaging e.g. a sealable plastic bag that contains enough absorbent material to contain the entire contents of the primary container without leakage occurring.

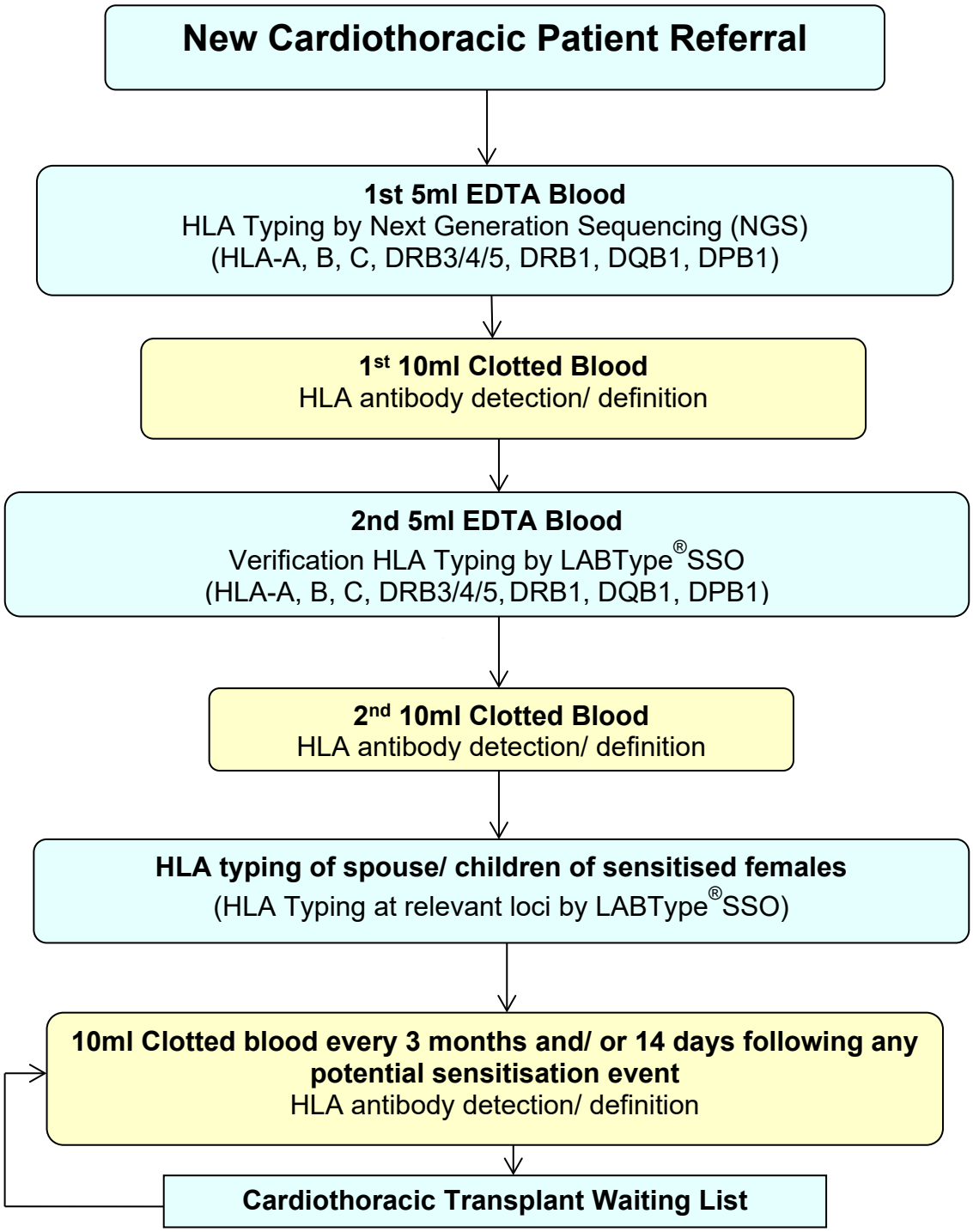
3. Outer packaging, to be labelled with the destination address, the name of the sending department and address, and be clearly marked “Diagnostic Specimen”

Appropriate packaging is available from suppliers including the Royal Mail, Royal Mail Safebox, FREEPOST, SWC1 143, Ross–on-Wye, HR9 7ZB.

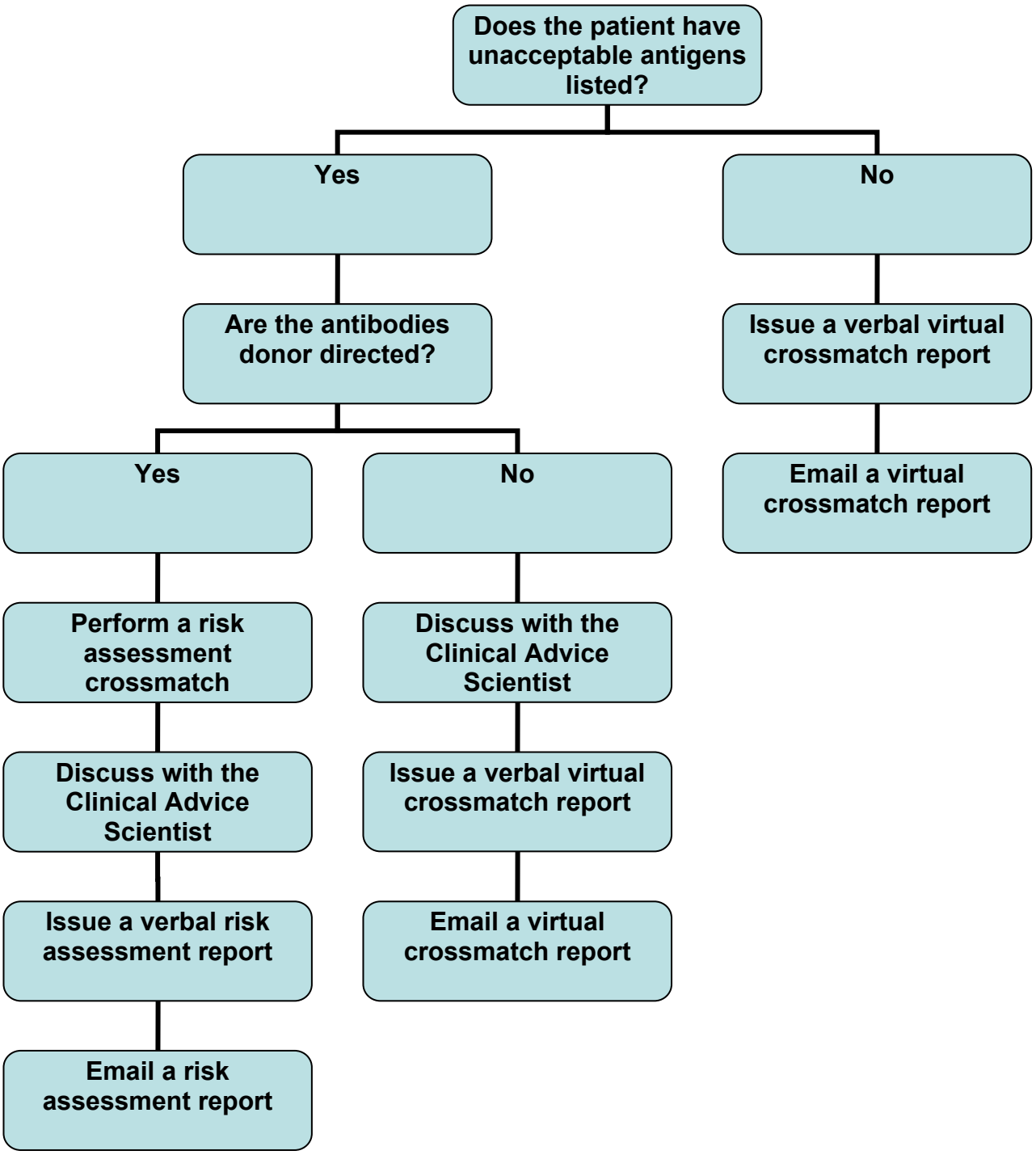
8.2 List of Tests and Samples Required

Test	Sample Required
Recipient HLA Typing (Initial HLA type)	5ml EDTA
Recipient HLA Typing (Verification HLA type)	5ml EDTA
Recipient HLA-Specific Antibody testing	10ml clotted blood
Post-transplant Donor specific antibody (DSA) testing	10ml clotted blood
Relative of sensitised females (child/ spouse) HLA Typing	5ml EDTA blood
Donor/ recipient Crossmatching	Donor: spleen and lymph node samples Patient: 10ml clotted blood
Recipient Autoantibody testing	20ml preservative free heparin/ EDTA and 10ml clotted blood

8.3 Cardiothoracic Patient Laboratory Work Up For Transplantation



8.4 Virtual Crossmatch Flow Chart Used by the On Call Clinical Scientist.



8.5 MFI risk level definitions for IgG HLA antibodies using LABScreen Single Antigen beads as defined by national CTAG guidelines

Risk Level	Cumulative MFI	Definition
Level 1	MFI = 0	No detectable antibody. Standard risk
Level 2	MFI <2,000	Minimum risk of hyperacute rejection but greater than standard risk of rejection
Level 3	MFI 2,000 – 5,000	Low risk of hyperacute rejection but significant risk of early rejection and antibody mediated graft damage. Immediate pre-transplant antibody reduction advised.
Level 4	MFI > 5,000	Transplant veto apart from exceptional cases

8.6 Laboratory risk level definitions for IgM HLA antibodies using LABScreen Single Antigen beads

MFI< 5000 negative
MFI>= 5000 <10000 listed as unacceptable - Risk level 2

All IgG and IgM MFI data are based on current (most recent sample) values.

8.7 Instructions for the collection of a Sample using a Buccal Swab

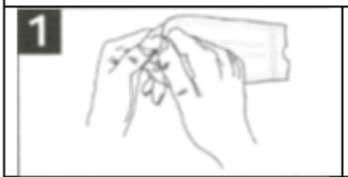
Buccal alone

NHS
Manchester University
NHS Foundation Trust

INSTRUCTIONS FOR COLLECTION OF BUCCAL CELL SAMPLES USING SWAB TECHNIQUE

Preparation:

- **Do not** use swab within 1 hour of eating, drinking or cleaning your teeth.
- Wash your hands thoroughly.
- Rinse your mouth out with water immediately before collecting sample
- Complete sticker label included in the pack with the individual's **Forename, Surname** and **Date of Birth**.
- Complete the sample card with the individual's **surname, forename, date of birth** and **date of specimen collection**.

	1. Pull the swab package open from one end.
	2. Remove the swab from the tube, taking care not to touch the white pad head.
	3. Insert the swab into your mouth. Using firm pressure, repeatedly rub against the inside of your cheek for **1 minute**.
	4. Place the swab back into the tube, taking care not to touch the white pad head.
	5. With the swab head in the tube, place your thumbnail in the small groove set in the swab handle (approximately 1 inch from swab head), then snap the handle in two by bending to one side. Let the swab head fall into the tube. a. Seal the tube with the cap provided. Affix the completed label to the tube, ensuring the excess sticker from both sides of the label stick together to form a flag-like formation - this will ensure the entirety of the label is visible. b. Put the tube in an envelope and send to the address stated on the accompanying letter.

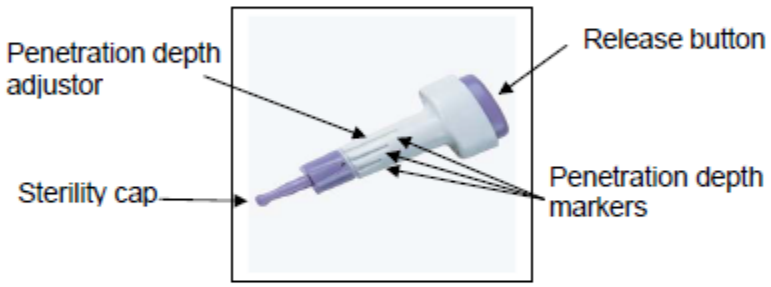
Send the collected sample to the Transplantation Laboratory (address overleaf) by first class in approved packaging.

8.8 Instructions for the collection of a Sample using a Blood Spot

NHS
Manchester University
NHS Foundation Trust

Transplantation Laboratory
Manchester Royal Infirmary, Oxford Road, Manchester, M13 9WL
Tel: 0161 276 6397, **Fax:** 0161 276 6148

COLLECTION OF BLOOD SPOT SAMPLES USING THE ACCU-CHEK® SAFE- T-PRO PLUS STERILE SINGLE-USE LANCING DEVICE



1. Ensure the collection paper is fully labelled with **surname, forename, date of birth** and **date of specimen.**
2. Wash hands thoroughly using soap and warm water and dry well to ensure a clean puncture site.
3. Twist the sterility cap and remove it.
4. The penetration adjustor is pre-set to the medium depth (~1.8mm) and is suitable for most adults. For children adjust to the low penetration depth (~1.3mm).
5. Hold the lancing device between the middle and index finger with the thumb on the release button.
6. Press the lancing device firmly against the puncture site, the side of the finger is recommended, and press the release button.
7. Squeeze the finger to encourage blood flow and collect at least 4 blood drops on each of the four circles (labelled 1-4) on the collection card (ensure the whole circle is filled with blood). A second puncture of the finger may be necessary.
8. Cover puncture site(s) with a plaster, dispose of the lance in an appropriate medical waste container and allow the blood spots to dry completely before packaging.