Title: SE-HMDS Immunophenotyping User Guide

Version number	1.2
Author	Katy Jones
Authorised by	Robert Dunn
Issued on	January 2025

Contents

1. Introduction

Synnovis is a unique partnership of clinical, scientific and operational expertise, with a mission to transform pathology services in the UK. Our organisation is built on scientific expertise, providing a service that helps clinicians create better outcomes for their patients every day.

Our full-service, customer-focused offer is strongly rooted in the patient pathway. We serve our founding NHS Trusts, other NHS and private hospitals, and the GP community at large.

We are continually focused on innovation, finding new and improved ways to manage the logistics of high-volume pathology testing as well as specialist reference testing. We always strive to improve capabilities to better meet our customers' needs.

The SE-HMDS at King's College Hospital is a regional centre for diagnostic services, providing Immunophenotyping, Cytogenetic, Molecular Diagnostic and Histopathology services covering most of South-East England.

The Immunophenotyping laboratory at SE-HMDS offers an extensive flow cytometric panel repertoire, to aid the accurate diagnosis and monitoring of blood and bone marrow disorders as well as providing testing for the national PNH service.

The Synnovis Immunophenotyping laboratory at SE-HMDS is a UKAS accredited medical laboratory, no. 9597.

2. Contact Details

Correspondence Address:
Synnovis Immunophenotyping (SE-HMDS)
Ground Floor, Hambleden Wing
King's College Hospital
Denmark Hill
London SE5 9RS

Sample Address:
King's SE-HMDS Laboratory
c/o Central Specimen Reception
Blood Sciences Laboratory
Ground Floor, Bessemer Wing
King's College Hospital
Denmark Hill
London SE5 9RS

General Enquiries:
Email: synnovis.immunophenotypingkch@nhs.net

3. Main Contacts

Clinical enquires:

If you have any clinical queries please contact the SE-HMDS Consultants between 9-5pm Monday to Friday.

kch-tr.KHMDC-consultants@nhs.net

Dr Deborah Yallop (SE-HMDS Co-Director) Deborah.yallop@nhs.net

Dr Shireen Kassam (SE-HMDS Co-Director) Shireen.kassam@nhs.net

Results and sample requirement enquires:

If you have any general queries about results not yet received or sample requirements please contact the departmental email that is monitored between 9-5:30pm Monday to Friday.

Email: kch-tr.sehmdsreception@nhs.net

Scientific enquires:

If you require specific scientific advice or guidance of immunophenotyping testing or results; please contact the Head of laboratory or the departments Principal Clinical Scientist or Operations and Quality Lead:

Head of Laboratory

Katy Jones katy.jones28@nhs.net

Principal Clinical Scientist and Laboratory & Staff Development Lead

Lauren Jamieson lauren.jamieson1@nhs.net

Operations & Quality Lead

Charles Stanley-Manogaran charles.manogaran@nhs.net

4. Hours of Operation

Monday to Friday 9.00am to 5.30pm

Weekends: There is no routine service at weekends.

Bank Holidays: The department is not staffed on bank holidays. An email is sent to regular customers in advance detailing arrangements over extended bank holiday periods.

5. Sample Types

5.1 Type of sample

Peripheral blood is the only suitable sample type for PNH analysis.
Suitable sample types for Immunophenotyping include peripheral blood, bone marrow aspirate, cerebrospinal fluid (CSF) and other non-CSF fluids (appropriate to the clinical indication, disease suspected or being monitored).
Please see below for additive/tube specification.
Blood and bone marrow immunophenotyping requests should be accompanied by 3 slides for integrated interpretation.

5.2 Patient preparation

To obtain reliable results when performing immunophenotyping for haematological malignancies, obtaining good quality samples and testing them in a timely fashion is essential. Samples should be received within 24 hours of aspiration to allow for appropriate testing and quality acceptance.
Traumatic aspiration should be avoided. Clotted specimens are suboptimal for analysis, however, analysis may be attempted to identify clinically significant findings although full phenotypic analysis may not be achievable.

5.3 Type of container/additives and volumes

5.3.1 Additive

Blood and **bone marro**w specimens: Please use 4ml **EDTA** (purple top) tubes.

CSF: Please aspirate the specified volume into **TransfixCSF** tubes, according to product insert (various volumes available). (Stability internally verified up to 6 days). Also send an aliquot in a **conical bottom universal tube (no additive)** to allow for cytospin morphology and cell count analysis.

Other non-CSF fluids (e.g. pleural, ascitic, etc): Samples must be fluid, core needle biopsies are not suitable for flowcytometry. Aspirate into a **conical bottom universal tube** (no additive).

5.3.2 Volume

Blood and bone marrow: 1ml minimum required. Samples less than the stated volume are processed where volume permits.

CSF: Specified volume of TransfixCSF plus ideally at least 0.5ml in universal container. (Smaller volumes may be attempted however quality of results and full phenotyping may not be achieved).

Other fluids: Ideally at least 1ml in universal container. (Smaller volumes may be attempted however quality of results and full phenotyping may not be achieved).

5.3.3 Stability

Please note that samples should ideally be received within 24 hours of being taken to allow testing to occur within the recommended timeframe of less than 72 hours (within 24 hours for non-Transfixed fluids). However, panels have been internally validated for stability up to 5 days (depending on panel).

CSF samples taken into Transfix are stable for 6 days. Other fluids (for example pleural fluid and ascitic fluid) can be tested as required however cells rarely survive longer than 24 hours and so should be processed as a matter of urgency.

5.3.4 Labelling requirements

All specimens (including slides and TransfixCSF) should have the following minimum patient identifier labels; Forename(s), Surname, date of birth and hospital/identification number. Specimens should be dated and sample type must be stated.

6. Dispatch of Samples

To provide an accurate result, samples for the laboratory must be sent in accordance with guidelines to ensure they arrive in a suitable condition to be processed and analysed.

World Health Organisation (WHO) Guidance (2021)[1] states that: "Shippers of infectious substances shall ensure that packages are prepared in such a manner that they arrive at their destination in good condition and present no hazard to persons or animals during transport."

Similarly, under various dangerous goods transport/carriage regulations[2,3], it is the responsibility of the consignor (sender/requester) to ensure that all dangerous goods, including diagnostic specimens, are correctly classified and packaged into suitable containers that are correctly marked and labelled.

It is therefore the responsibility of the requestor to ensure that all samples are sent to Synnovis in accordance with the following instructions.

6.1 Packaging requirements

Potentially infectious samples from GPs transported by designated vehicles provided by Synnovis or the local NHS Trust must be carried out in compliance with the UK and European road transport regulations[2].

Infectious substances include material that is known to contain, or is reasonably expected to contain, pathogens. When in transport, infectious substances must be packaged according to the packing Instruction 650 of ADR as follows:

- All samples in containers (e.g. tube, pot known as the “primary”) must be placed in individual sample bags to avoid cross contamination. **Never send samples from different patients in the same sample bag.** Where the primary contains a liquid, then the primary container must be leak proof. Where the primary contains a solid, then the primary container must be sift proof (impermeable to dry contents).
- Individual sample bags should be placed into large, clear, sealable, leak proof, plastic, sample bags (known as the “secondary”) that, where the specimen is a liquid, contains absorbent material sufficient to absorb the entire quantity of the liquid present in the specimen container (e.g. a sufficient amount of paper toweling to absorb any leakage).
- The referral paperwork should be contained in the secondary packaging pocket.
- The large bag should be placed into a suitable rigid sample transport container that meets the testing requirements of the regulations and is correctly marked and labelled.
- Only rigid outer containers supplied by Synnovis or the local NHS Trust may be used to transport samples to the laboratory by road.
- There should be sufficient cushioning lining the outer rigid container to prevent samples becoming unstable.

N.B. Please send samples at the earliest opportunity; samples must be received within the timeframe specified above to ensure sample viability. Blood, bone marrow and Transfixed CSF samples not sent immediately should be

refrigerated at 4ºC and sent at the earliest opportunity. Non-transfixed fluids should remain at room temperature before sending.

6.2 Request/Referral Forms

Please use the King's SE-HMDS request form which may be retrieved from the South East Genomics website:
https://southeastgenomics.nhs.uk/glh/cancer-tests/haemato-oncology/

The clinical reason for referral is vital to determine the appropriate immunophenotyping panel(s) are selected, correct number of cells are analysed and how the sample is prioritised. All relevant clinical and haematological information and likely diagnosis can be included. If the patient is a participant of a research trial, it is important to give details as certain trials can have specific analysis requirements. Please include any treatments/therapies that the patient is or has recently received paying particular attention to targeted immunotherapies that may affect the immunophenotype.

The department operates a specimen acceptance policy. The following details are essential requirements for request cards. Samples referred without at least three patient identifiers may not be processed.

Request forms must contain the following information:

- Patient's forename and surname
- Patient's date of birth
- Patient's genetic sex
- Requestor's name and location:
  - Internal Request - location (ward code) and clinician details/code
  - External Request - address label/surgery and Consultant details.
- NHS and Hospital number
- Type of specimen(s)
- Date & time of specimen collection
- High risk for bacterial or viral infection or confirmed high risk infection; High risk specimens must be identified to the laboratory using the referral form (*Please note: without this information the specimen **will not** be processed by the laboratory*).
- Test(s) required
- Relevant clinical information, patient history and any transplant donor sex
- Request forms must be dated and signed by those taking the specimen. Please include appropriate contact details. (*Please note: without this information the specimen **will not** be reported by the laboratory*).

6.3 Rejection of Unacceptable Specimens

Specimens and request forms are checked on receipt to confirm the patient identification (PID) information provided on the form and specimen agree. A minimum of three PID data items (e.g. Full name, DOB, NHS number or hospital number) are required by the laboratory and must match for the specimen to be accepted. Please ensure PIDs and contact details are ***clear and legible*** on all referral forms sent to SE-HMDS.

Samples without any patient identifiers are discarded and **not processed**.

6.4 Policy for High Risk Samples

Samples accompanying cytogenetic requests from patients at high risk of infection referred for cytogenetic analysis **must** be identified to the laboratory.

The sample and request form must be clearly labelled as High Risk.

Please note: Specimens indicated as high risk without identification of the pathogen(s) will not be processed by the cytogenetic laboratory and communication with the referring provider will be attempted. If no response after 48 hours the sample will be disposed of.

HIGH RISK DISEASES

Anthrax
Brucellosis
Creutzfeldt-Jakob Disease
Ebola
E. coli 0157 Infection
Hepatitis B
Hepatitis C
HIV
Severe Acute Respiratory Syndrome (SARS)
SARS-CoV-2
TB
Typhoid or Paratyphoid fever
Viral haemorrhagic fever (VHF) of any type

Please note any ACDP (Advisory Committee on Dangerous Pathogens) category 3 pathogen (such as TB) or higher will not be processed by the laboratory as it does not have the sufficient containment level.
The Health & Safety Executive's approved list of biological agents can be found on their website:
http://www.hse.gov.uk/pubns/misc208.pdf

7. Reporting

7.1 Results-online

SE-HMDS laboratories offer test results online for NHS healthcare professionals via the KHMDC online portal. This is a free, secure, electronic, pathology results on-line service and is available to registered users. Results are available on this portal in real-time as that are completed and authorised. Please contact kch-tr.KHMDC-consultants@nhs.net or Synnovis Customer Support on 0203 299 3576 if you would like to register for access to this service as a new user at an existing referral site.

7.2 Posting / E-mailing Reports

Full copies of authorised reports are available on specific request or arrangement where online results access is not available. These will be securely emailed as PDFs using a group nhs.net account.

7.3 Additional Testing

Requests for additional tests on a specimen referral can be made by email if clinically relevant and agreed with a HCPC registered healthcare scientist. This is subject to sufficient sample material availability.

7.4 Samples requiring further information

Referrals that lack sufficient details for onward processing may be held pending further information which will be sought via email or telephone. Samples referred with urgent referral indications will be processed as appropriate for the disease until additional information is received that indicates tests should be cancelled.

N.B. Consultants are requested to co-operate as fully as possible with this policy; please respond to requests for further clinical information within 2 days otherwise samples may not be analysed. This is to avoid unnecessary work and helps the laboratory to process its large workload.

7.5 Reporting Times

Routine testing has a turnaround time of 5 working days, urgent testing has a turnaround time of 1 working day (see triaging section for designation of urgency).

8. Laboratory Storage of samples

All samples for immunophenotyping are stored in accordance with the guidelines issued by the Royal College of Pathologists in April 2015[5].

Immunophenotyping samples referred to SE-HMDS are stored for 2 weeks post receipt. Slides are stored onsite for 1 year before being sent for offsite archiving for 30 years. Digital flow cytometric data files are stored with maintained accessibility for a minimum of 30 years.

9. Techniques

Immunophenotyping, including measurable residual disease is performed using fully validated panels and where appropriate align with Euroflow recommendations. Panel descriptions are available on the UKAS website 9597 Medical Single

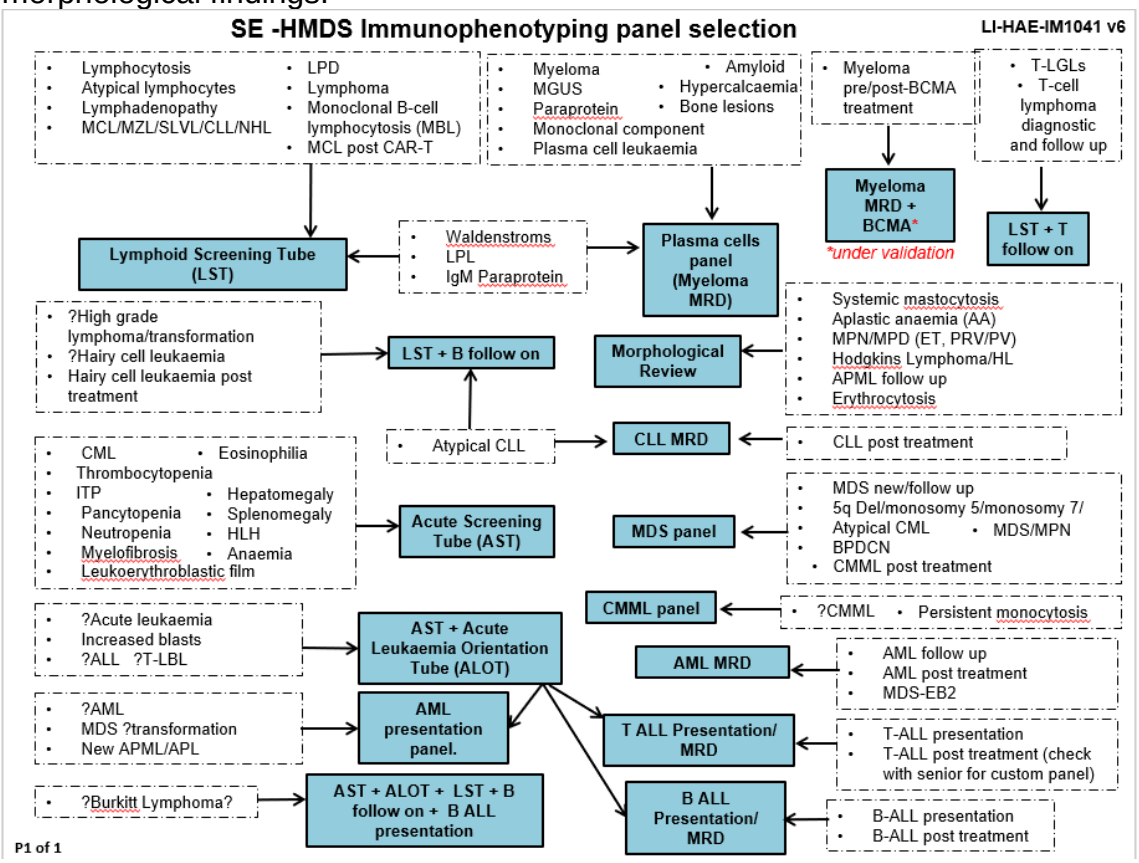
10. Immunophenotyping triaging and panel selection

10.1 Urgency triaging

Specimens are triaged for urgency depending on the clinical information provided. For example, highest priority is given to the presentation of new acute leukaemias and high grade lymphomas. Consideration is also given to specimen type and sample viability.

10.2 Immunophenotyping panel selection

Immunophenotyping panel are selected according to clinical information provided and where appropriate, further reflexed due to initial flow cytometry or morphological findings.



11. Complaints and Compliments

The department has procedures for logging compliments and complaints from service users. Please contact the Head of Service for further details if required.

12. References

1. Guidance on regulations for the transport of infectious substances 2021-2022. Geneva: World Health Organization; 2021. Licence: CC BY-NC-SA 3.0 IGO. **[PDC-HAE-CYT-TRANSREG]**
2. European Agreement concerning the International Carriage of Dangerous Goods by Road 2023 (ADR 2023) Volumes 1 & 2 **[PDC-HAE-CYT-ADR]**
3. The Carriage of Dangerous Goods and Use of Transportable Pressure Equipment Regulations (2009) as amended 2011. **[PDC-HAE-CYT-DGOODS]**
4. The Royal College of Pathologists: The retention and storage of pathological records and specimens (5th edition). 2015 **[PDC-HAE-CYT-RCPATH]**