Laboratory Haematology

OXFORD UNIVERSITY HOSPITALS NHS FOUNDATION TRUST

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Laboratory Haematology

About the Oxford University Hospitals NHS Foundation Trust (OUH)
One of the largest NHS teaching trusts in the country. It provides a wide range of general and specialist clinical services and is a base for medical education, training and research. The Trust is one of the largest employers in Oxfordshire, primarily based in Headington, Oxford and comprises:

The John Radcliffe Hospital
- accident and emergency
- Acute medical and surgical services, trauma, intensive care, cardiac, women’s services and children’s services.

The Churchill Hospital
- non-emergency specialist services
- Renal medicine and transplant, clinical and medical oncology, dermatology, chest medicine, infectious diseases and palliative care.

The Horton Hospital (Banbury)
- accident and emergency services
- general hospital service, maternity and paediatric services,

The Nuffield Orthopaedic Centre (NOC)
- Orthopaedics, rheumatology and rehabilitation.
About us:

The main Haematology Department, including Blood Transfusion, is situated on Level 4 of the John Radcliffe Hospital, Headington, Oxford, and provides a comprehensive service to Oxfordshire and other, more distant referring laboratories.

This service includes the following:

1. Routine Haematology
2. Routine Haemostasis
3. Blood Transfusion service
4. Morphology and Leukaemia Immunophenotyping service
5. A comprehensive molecular diagnostic service for a range of haematological conditions, including both the National Haemoglobinopathy Reference Laboratory and the Thames Valley Haemato-molecular Diagnostic Service. (More information can be found at: /haematology-laboratories/molhaem/).

On the Horton General Hospital site the department is located in the Pathology building (beyond E and F ward, near Gynaecology), provides the following services:

1. Routine Haematology and Morphology
2. Routine Haemostasis
3. Blood Transfusion service

This provides a comprehensive service to the hospital as well as local GP’s in the North of Oxfordshire, and South of Northamptonshire.

On the Churchill Hospital site the department provides the following services:

1. Routine Haematology, haemostasis and blood product issue support, 8am-7pm Monday to Friday, from the Laboratory Medicine Building. Located opposite Car Park 5. Use Air Tube Station 14 to send samples. This laboratory is referred to as Laboratory Medicine (LM)
2. Specialist Haemostasis and Thrombophilia diagnostic and monitoring service for patients with inherited haemostatic defects. (The Oxford Haemophilia and Thrombosis Centre, OHTC). The OHTC is located to the rear of the Churchill campus near John Warin Ward.

On the Nuffield Orthopaedic Centre site there is no Haematology or Blood Transfusion laboratory and all services are supported from the John Radcliffe.

24/7/365 services are maintained on both the John Radcliffe and the Horton sites.
Policies- Dept. of Laboratory Haematology Oxford University Hospitals Foundation Trust

Policy Statements

Animal Specimens
We do not accept animal specimens for laboratory testing

Billing
- Work to support NHS care of OUH patients will be managed following Trust budget setting process, with cross-charging from Directorate of Pathology and Laboratory Medicine to appropriate Division / Directorate based on monthly outcome against plan.
- Work to support Oxfordshire CCG NHS workload will be billed by OUH commissioning according to the agreed overall Contract on a monthly basis.
- In the absence of a specific exceptional contract, the standard Laboratory Medicine Terms and conditions will apply. ([http://www.ouh.nhs.uk/cellular-pathology/documents/terms-and-conditions.pdf](http://www.ouh.nhs.uk/cellular-pathology/documents/terms-and-conditions.pdf)).
- In case of initial referral of work for the first time to the laboratory, or a significant change (considered to be an increase of greater than 20 percent by activity), please contact the laboratory prior to referral, so that the laboratory can assure you that they have the capacity and resources to meet your request.
- Receipt of a sample for testing will be deemed to be acceptance of a contract with OUH Foundation Trust, the work will be invoiced within 30 days of completion and payment is expected within 30 days of invoice being raised. It is the sender’s responsibility to manage the process of raising purchase order numbers, if that is required by the sending organisation.
- Private Patients:
  - The OUH sees a variety of Private Patients, they need to be registered with the OUH Private Patient Office ([http://www.ouh.nhs.uk/privatehealthcare/payment/default.aspx](http://www.ouh.nhs.uk/privatehealthcare/payment/default.aspx)), and the bills for the Laboratory Medicine fraction of their care will be raised as part of their overall care. If insurance is involved they need to sign a Pre-authorisation with the relevant Insurance Company.
  - Non-OUH patients, the Department is happy to provide services for other organisations that provide care to Private Patients, but as the Department will have no relationship with the individual patient, any bill will be the responsibility of the sending organisation. The Department will not bill individual patients.
- Clinical Trials:
  - Clinical Trials at OUH if there is a laboratory component to the Trial should have agreed the financial aspects of the study with the Joint R&D office based at the Churchill Hospital, while obtaining ethical approval. Details of any support required from OUH laboratories should be agreed in advance of the Trial commencing, contact labtrials.ouh@nhs.net
  - Individual projects can be agreed directly with the laboratory on a case by case basis, please contact Laboratory Manager, Haematology and labtrials.ouh@nhs.net, well in advance.

Business Continuity and Contingency Plans
In the event of a local, regional or national disaster, Oxford University Hospitals NHS Trust have comprehensive contingency plans in place to ensure the impact on care and specifically on laboratory services is minimised. With standardisation of testing across our various sites we have worked to ensure the majority of the common workload can be performed from more than one site.
Chain of Custody

Chain-of-Custody is a record of disposition of a specimen to document who collected it, who handled it, who performed the analysis, is often required when results are to be used in a court of law, (e.g. in Paternity testing cases). The Dept. of Laboratory Haematology does NOT provide this service. In certain appropriate cases with relevant consent, samples or test results originally used for clinical care would be released if no longer required for clinical testing.

Confidentiality of Results

OUH Hospitals Laboratory Haematology Department is committed to maintaining the confidentiality of patient information. To work towards this goal, we aim to minimise the transmission of results by telephone or Fax and aim to maximise the use of electronic transmission of results to systems with audit trail of access to the results.

The Department follows the policy of the Oxford University Hospitals Foundation Trust, with regards to patient confidentiality and as such all staff are required to complete training in Information Governance and maintain competence.

Phone Enquiry Policy (Gen 001)

Results will only be released to a referring clinician or their approved representative. Third parties including patients or their relatives will be referred to the ordering clinician. We will want to ensure anyone phoning has legitimate authority to receive the results. Patients would need to be clearly identified by use of NHS or MRN number AND full name or Date of Birth. Provision of appropriate information before enquiring will greatly assist prompt and accurate response to enquiries and reporting.

Complaints / feedback

Although we always like to hear about the things we have done well, we would also like to hear about the things we could do better.

The Oxford University Hospitals NHS Foundation Trust (OUH) is committed to providing the very highest standards of care.

We will always try our best to get things right, but sometimes mistakes happen. When they do, it is vitally important to put things right as soon as possible, and to ensure that the same mistakes do not happen again. If as a user you feel that the department has not fulfilled its obligations to you, please contact the department as below. These contact details can also be used for feedback and questions about services provided

- Laboratory Manager: Dan Smith (Dan.smith@ouh.nhs.uk) 01865 220337
- Quality Manager: Andrew Platt (Andrew.platt@ouh.nhs.uk) 01865 857663

Making a Formal Complaint

Please put your complaint in writing to:

Chief Executive
Oxford University Hospitals NHS Foundation Trust
Headley Way
Headington
Oxford OX3 9DU
Email: complaints@ouh.nhs.uk
http://www.ouh.nhs.uk/about/complaints.aspx
Telephoning of Critical Values

The Critical Values policy is described below:

Definition of a Critical Value: - A Critical Value is defined as one which is such at variance with normal (expected values) as to be life threatening unless something is done promptly and for which some corrective action could be taken.

Abnormals are not considered Critical Values: Most laboratory tests have established reference ranges which are the results that are typically seen in a group of healthy individuals. While results outside these ranges may be considered abnormal, that is not the same as “critical”.

Action taken when a result exceeds the Critical Values: In addition to normal reporting staff will attempt to telephone or otherwise contact the ordering clinician as quickly as possible. For this reason, each request should be accompanied by contact details to allow the laboratory to contact a referring clinician. The following limits apply to inpatients and primary care at all times.

Critical Values for FBC include:
- Haemoglobin < 50g/l (when unexplained by clinical data/diagnosis)
- Platelet count < 30x10^9/l (when unexplained by clinical data/diagnosis)
- Neutrophil count < 0.5x10^9/l (when unexplained by clinical data/diagnosis)

Critical Values for Coagulation testing include:
- INR > 7.9 (patients on Warfarin)
- PT > 25.0 seconds (unless known to be receiving anti-coagulation therapy)
- APTT > 50.0 seconds (unless known to be receiving anti-coagulation therapy)
- Fibrinogen < 1.0 g/l (new presentation)

The appearance of significant features on a blood film (not diagnosed / unexpected change) to include but not limited to:
- New diagnosis of Malaria infection
- New diagnosis of Acute Leukaemia
- New diagnosis of red cell fragmentation syndrome (HUS, DIC, TTP)

Disclosure of Results

Results will only be released to a referring clinician or their approved representative. Third parties including patients or their relatives will be referred to the ordering clinician.

Infectious Material

All samples will be treated by the department as potentially hazardous; however, samples from patients who are known to pose an infection risk should have this information appropriately recorded on the request form and all samples should have an approved danger of infection sticker on them. It is the duty and responsibility of the sender aware of the risks and to arrange for appropriate packaging, labelling and transportation.

- Both the form and the specimen label must carry a common warning label indicating in black on a yellow background.
The label must be clearly visible to anyone handling the specimen but should not carry clinical details.

Apart from the common warning label, the request form must give sufficient clinical information to enable laboratory staff to know which precautions to take.

Because of the extra work and stress involved in processing 'high risk' specimens it is important that the category is limited to those specimens where it is a matter of medical opinion that the patient concerned is likely to be carrying a hazard group-3 pathogen.

- Each specimen must be sealed in a double plastic bag.
- Place labels:
  - One on the specimen container
  - One on the request form / EPR specimen envelope.
- Please ensure that the correct samples are sent, as no decanting will be possible on high-risk samples.

Blood taken from the following patient's must be treated as “high-risk” specimens:

1) Patient’s known to be HIV positive.
2) Patient’s known to be HTLV1 positive.
3) Patients who are known to be Hepatitis B or C positive.
4) Intravenous drug abusers.
5) Patients from other known high-risk groups.

- Please contact the department before sending samples from patients suspected of having any form of Viral Haemorrhagic fever (VHF), as these will require special processing and only minimal tests will be performed.
- Samples from patients with confirmed VHF must not be sent to the department, as we are not equipped to handle them.

For further information see ACDP.1995. Categorization of biological agents according to hazard and categories of containment London. HMSO.

Labels are available from: Oxuniprint, stock code – DOIL0001, Danger of Infection Label pk 250.
Distribution of testing on various sites within the department of laboratory Haematology
The department operates 4 laboratories on three sites within the OUH FT. this document describes the scope of practice on each site.

Churchill site
This has two laboratories (LM and OHTC)
LM:
- FBC (including Retic)
- Clotting screen / INR / D-Dimer
- Electronic issue of blood products (blood, platelets, plasma)

OHTC
- Clotting screen / INR / D-Dimer
- Factor assays
- VWD investigations
- Thrombophilia investigations
- Haemostatic investigations
- Platelet function investigations
- HIT screening

Horton site
- FBC (including Retic)
- ESR
- Malaria investigations
- Glandular Fever
- Sickle Screen
- Blood film examination
- Haptoglobin
- G6PD assay
- Clotting screen / INR / D-Dimer
- Blood Group and antibody screening / investigation
- Issue of blood products (blood, platelets, plasma)

JR site
- FBC (including Retic)
- ESR
- Malaria investigations
- Glandular Fever
- Sickle screen
- Plasma viscosity
- Blood film examination
- Bone marrow examination
- Clotting screen / INR / D-Dimer
- LMW Heparin testing
- Blood Group and antibody screening / investigation
- Issue of blood products (blood, platelets, plasma)
- Foetal-maternal haemorrhage estimation
- ABO titres
- Haemoglobinopathy diagnosis
- Flow cytometry: acute / chronic leukaemia investigations & PNH (low sensitivity only)
- Molecular investigations for
  - Cancer / leukaemia
  - Haemoglobinopathy / Anaemia investigations
  - Haemostatic / thrombotic disorders
  - Disorders of Iron regulation
The Department attained ISO 15189:2012 accreditation in July 2016 after assessment by UKAS against the Standard ISO-15189 Medical laboratories—Requirements for quality and competence

For accredited scope of practice please see below


Any tests referred to in this handbook which are not explicitly covered in the scope of practice above are by definition NOT part of the laboratories external accreditation. They should as far as practicable still be covered by the Laboratory Quality Management system, including QC and initial verification.

Update: July 2017.
The department as part of its performance improvement process has altered the technical processes used to perform a number of specific tests. These changes have been internally verified and do not alter the results as seen by requesting clinician’s. However as these changes have not as yet been reviewed by UKAS, they should be considered to be temporarily out of scope of ISO 15189 accreditation. The laboratory is intending to rectify this issue on the forthcoming surveillance visit in Q4 of 2017-18. If you would like to discuss this issue please contact the quality manager (Andrew.platt@ouh.nhs.uk)

The related tests are:
- Flow cytometry for chronic / acute leukaemia’s
- Flow cytometry for PNH screening
- HIT screening
- Collagen binding assay (CBA)
- TP53 mutation testing

The Department takes part in a wide variety of National External Quality Assurance Schemes.

The Department is annually assessed by the MHRA for conformance with the Blood Safety and Quality Regulations.

The Department contributes as required to the Foundation Trusts assessment by CQC against relevant standards.

Proficiency Testing
The laboratory is committed to participation at least 40 different external Quality assurance schemes as well as a variety of internal quality control process to ensure the consistent quality of results produced. External QA schemes selected are chosen on the basis of suitability of the laboratory’s needs. Where possible EQA schemes will be accredited to ISO 17043 international standard or hold equivalent markers of quality, participation in individual schemes is kept under regular review

Schemes we participate in as of July 2017 include:

<table>
<thead>
<tr>
<th>Scheme</th>
<th>Site</th>
<th>Frequency</th>
<th>Section</th>
</tr>
</thead>
<tbody>
<tr>
<td>NEQAS FBC</td>
<td>all</td>
<td>Every 4 weeks</td>
<td>Core automated</td>
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<tr>
<td>NEQAS Automated WBC differential</td>
<td>all</td>
<td>Quarterly</td>
<td>Core automated</td>
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<tr>
<td>Service</td>
<td>Frequency</td>
<td>Methodology</td>
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<td>----------------------------------------------</td>
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<tr>
<td>NEOAS Reticulocytes</td>
<td>Every 8 weeks</td>
<td>Core automated</td>
<td></td>
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<tr>
<td>NEOAS Sickle</td>
<td>Every 8 weeks</td>
<td>Core automated</td>
<td></td>
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<tr>
<td>NEOAS G6PD assay</td>
<td>Every 8 weeks</td>
<td>Core automated</td>
<td></td>
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<tr>
<td>NEOAS Haptoglobins</td>
<td>Every 4 weeks</td>
<td>Core automated</td>
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<tr>
<td>NEOAS Plasma Viscosity</td>
<td>Every 4 weeks</td>
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<tr>
<td>NEOAS ESR</td>
<td>Every 6 months</td>
<td>Core automated</td>
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<tr>
<td>LabQuality GF (commercial scheme)</td>
<td>Quarterly</td>
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<tr>
<td>NEOAS Nucleated RBC (pilot)</td>
<td>Every 6 months</td>
<td>Core automated</td>
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<tr>
<td>NEOAS Malaria RDT</td>
<td>CH</td>
<td>Haemostasis</td>
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<tr>
<td>NEOAS Haemoglobinopathy Adult</td>
<td>Every 8 weeks</td>
<td>Haemoglobinopathy</td>
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<tr>
<td>NEOAS Haemoglobinopathy Neonate</td>
<td>Every 4 weeks</td>
<td>Haemoglobinopathy</td>
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<tr>
<td>NEOAS Haemoglobinopathy Liquid Capillary</td>
<td>Every 8 weeks</td>
<td>Haemoglobinopathy</td>
<td></td>
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<tr>
<td>NEOAS Routine Coagulation</td>
<td>Every 8 weeks</td>
<td>Core automated</td>
<td></td>
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<tr>
<td>NEOAS Thrombophilia</td>
<td>CH</td>
<td>Haemostasis</td>
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<tr>
<td>NEOAS Morphology (Films)</td>
<td>CH</td>
<td>Haemostasis</td>
<td></td>
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<tr>
<td>NEOAS Morphology (Malaria)</td>
<td>CH</td>
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<tr>
<td>NEOAS Morphology (manual WBC differential)</td>
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<tr>
<td>NEOAS Digital Morphology</td>
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<td>NEOAS Immunophenotyping (part 1)</td>
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<td>NEOAS Immunophenotyping (part 2)</td>
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<tr>
<td>NEOAS PNH</td>
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<tr>
<td>NEOAS Special Stains (Iron stain only)</td>
<td>CH</td>
<td>Morphology</td>
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<tr>
<td>NEOAS ABO &amp; Rh D testing</td>
<td>CH</td>
<td>Blood Transfusion</td>
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<tr>
<td>NEOAS Antibody screening and identification</td>
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<td>Blood Transfusion</td>
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<tr>
<td>NEOAS Crossmatching</td>
<td>CH</td>
<td>Blood Transfusion</td>
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<tr>
<td>NEOAS Red cell phenotyping</td>
<td>CH</td>
<td>Blood Transfusion</td>
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<tr>
<td>NEOAS Foetal Maternal Haemorrhage</td>
<td>CH</td>
<td>Blood Transfusion</td>
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<tr>
<td>NEOAS ABO titration</td>
<td>CH</td>
<td>Blood Transfusion</td>
<td></td>
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<tr>
<td>DNA diagnostics for Haemoglobinopathies</td>
<td>CH</td>
<td>Molecular Haematology</td>
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<tr>
<td>NEOAS / CEQAS BCR-ABL screening</td>
<td>CH</td>
<td>Molecular Haematology</td>
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<td>NEOAS / CEQAS BCR-ABL quantitation</td>
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<td>Molecular Haematology</td>
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<tr>
<td>NEOAS Jak-2</td>
<td>CH</td>
<td>Molecular Haematology</td>
<td></td>
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<tr>
<td>NEOAS Clonality (IgH)</td>
<td>CH</td>
<td>Molecular Haematology</td>
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<tr>
<td>NEOAS Gastro-Intestinal Stromal tumours</td>
<td>CH</td>
<td>Molecular Haematology</td>
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<tr>
<td>NEOAS Colorectal tumours</td>
<td>CH</td>
<td>Molecular Haematology</td>
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<tr>
<td>NEOAS Lung Cancer</td>
<td>CH</td>
<td>Molecular Haematology</td>
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<tr>
<td>NEOAS Melanoma</td>
<td>CH</td>
<td>Molecular Haematology</td>
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<tr>
<td>NEOAS / CEQAS NPM-1 mutation analysis</td>
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<td>Molecular Haematology</td>
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<tr>
<td>NEOAS / CEQAS SCT Chimerism</td>
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<td>Molecular Haematology</td>
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<td>WFH / NEOAS Haemophilia genetics</td>
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<td>Molecular Haematology</td>
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<td>NEOAS Thrombophilia genetics</td>
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<td>Molecular Haematology</td>
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<tr>
<td>Service</td>
<td>Frequency</td>
<td>Department</td>
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<td>NEQAS FLT-3 mutation analysis</td>
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<td>Molecular Haematology</td>
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<td>NEQAS / Genomics England DNA extraction (pilot)</td>
<td>Annual</td>
<td>Molecular Haematology</td>
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<tr>
<td>NEQAS / EMQN Next Generation sequencing</td>
<td>Annual</td>
<td>Molecular Haematology</td>
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<tr>
<td>NEQAS HFE testing (replacing EMQN HFE)</td>
<td>Every 6 months</td>
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<td>NEQAS HFE interpretative results only</td>
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<tr>
<td>NEQAS Pathogenicity of Sequence Variants (PSV)</td>
<td>Every 6 months</td>
<td>Molecular Haematology</td>
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</tbody>
</table>
The Quality Policy of the Department of Laboratory Haematology; within the Directorate of Pathology and Laboratories; Oxford University Hospitals NHS Foundation Trust.

The Department of Laboratory Haematology provides a diagnostic and routine monitoring service for the OUH NHS Foundation Trust; local CCG's and acts as a referral centre for referring hospitals. The analytical profile of the department includes both routine tests and specialist molecular analysis. In addition, the department provides blood transfusion support for the trust and certain surrounding hospitals. The department is committed to providing a service of highest quality and is aware and takes into consideration the needs and requirements of its users. In order to ensure that the needs and requirements of users are met, the department of laboratory haematology will:

- Operate a quality management system to integrate the organisation, procedures, processes and resources required.
- Obtain and monitor data on user satisfaction and complaints.
- Review the existing services in relation to the needs and requirements using data gathered from the users of the service in order to achieve continual quality improvement.
- Set quality objectives and plans in order to implement this quality policy and to ensure that it is suitable and effective.
- Ensure that all personnel are familiar with this quality policy to ensure user satisfaction.
- Commit to the health, safety and welfare of its entire staff. Visitors to the department will be treated with respect and due consideration will be given to their safety while on site.
- Uphold professional values and remain committed to good professional practice and conduct.
- Comply with any relevant environmental legislation.
- Where such appropriate controls exist, comply with any relevant or appropriate national or international product storage legislation.

The Department of Laboratory Haematology will comply with the requirements of ISO 15189:2012; other relevant national and international standards and where appropriate, other regulatory bodies and is committed to:

- Staff recruitment, training, development and retention at all levels sufficient to provide a full and effective service to its users.
- Ensuring that all staff are familiar with the contents of the Quality manual and all procedures relevant or appropriate to their work.
- The correct and proper procurement and maintenance of such equipment and others resources as are needed for successful provision of service.
- The collection, transport and handling of all specimens where appropriate in such a way as to ensure the correct performance of laboratory examinations and where appropriate in compliance with relevant legislation.
- The selection and use of examination procedures that are fit for purpose and provide the highest achievable quality of all tests performed.
- Reporting results of examinations in ways that are timely, confidential, accurate and clinically useful.
- The assessment of user satisfaction, in addition to internal audit and external quality assessment, in order to produce continual quality improvement.

This policy is a controlled document and is reviewed annually within the department. Signed copies will be displayed within the laboratory.
Radioactive Samples
Specimens from patients receiving radioactive tracers or material should be marked as such. Please include the date and time of receipt and the isotope used. These samples will be handled in such a way as to protect the health and safety of our staff while meeting the needs of the patient. This may involve some delay in the processing of the sample. Please ensure testing is truly necessary before requesting testing on patients immediately after such treatment.

Record Retention
The Department retains requests sample material and test results for the retention periods recommended by the Royal College of Pathologists, in “Retention and Storage of pathological records and specimens 5th edition 2015”
http://www.rcpath.org/Resources/RCPath/Migrated%20Resources/Documents/G/G031_RetentionAndStorage_Apr15.pdf

Referral of Tests to another Laboratory
Although the vast majority of the analytical methods that the department offers are performed on one of the three sites, there are a number of tests that are referred to other external laboratories. This will happen for one of the following reasons:

1. The test requires expertise or equipment that the department does not have
2. The low number of request's is such that, it would not be possible to maintain suitable skills or competence in the analytical method, so they are referred to another referral laboratory
3. In extremis, routine tests may be referred in the case of catastrophic analyser failure, as part of the departmental service contingency plan

Referring laboratories are chosen that provide equal performance to our own and are regularly asked to provide evidence of continuing CPA / UKAS accreditation and acceptable EQA performance. A list of the referring laboratories is held in the department and is available on request to the quality manager.

Referred samples will be sent off by the laboratory using appropriate postal or courier methods and the laboratory will manage the dispatch and return of results process. A process of monitoring TAT for these samples is in place.

Where possible reports will be sent out using similar mechanisms used for internal processing; however, they will contain the name of the processing laboratory and reference ranges where appropriate.

OUH will invoice for samples referred to another laboratory at the price charged to OUH, plus a small administrative charge plus if required any courier costs.
Oxford University Hospital Foundation Trust Users
All results will be sent to the Cerner Millennium EPR System

What you need before you start
A computer that has been set up to connect to the EPR either from the OUH virtual desktop, or directly from a web page short cut on your desktop. You need to ensure that your smartcard has been properly enrolled and provisioned to give you access to Trust systems, and that you have an OxNet (hospital network) account.

Login process
You will need an NHS Smartcard and passcode, it is your responsibility to obtain this and a log-in before working your first shift, (even for locum staff!).

Tap&Go
For clinicians, most of the time, you will access clinical systems via the OUH vWorkspace virtual desktop, using your NHS smartcard to activate the proximity sensor attached to the PC, by gently tapping it with your card. This takes you to a desktop from where you can launch EPR simply by entering your passcode. You can secure your session by tapping the sensor again, and this allows you to retrieve your session at any other machine running vWorkspace (with a proximity sensor attached). After a pre-determined time should you not return to your session the EPR will time-out, and you will have to launch it again when you re-connect to your session.

- Always secure your session before leaving the PC.
- Never share your card or passcode – you would be in breach of the Data Protection Act.

Launching Millennium
To launch Millennium click on the TRUE icon in the task bar or double-click the icon on the desktop: Either will open the browser page:
After single-clicking the PowerChart icon (the middle one of the Applications) the login window Your Smartcard number is entered automatically by the single sign-on system – then enter your Millennium passcode (this can be set up to be the same as your Smartcard passcode – a minimum of 4 digits like a bank PIN), and press enter key or click OK

Forgotten your smartcard or its stopped working?
The Tap&Go infrastructure addresses a number of hardware usability and security issues that make the system easy for busy clinicians to use – faster login, fast user switching, without losing your active Millennium EPR session, secure session roaming, all using the NHS Smartcard with proximity sensor to automate the login process.
However, if your Smartcard fails (nothing happens when you tap the sensor) or you forget it at home, then you can log in to your vWorkspace desktop by using your OUH network username and passcode entered into the Windows login window. This is the same process you would use if you accessed vWorkspace from the web portal.

All Results should be Endorsed (signed) on receipt

- You can endorse results even if you have not requested them; you are part of a team and can sign results if you are tasked to check them.
- Endorsement ensures you have signed and if necessary actioned a result.
- This is all about Patient Safety
- It is Trust Policy that ALL results must be endorsed.
- NB Results for FBC and Coagulation Screens will be released prior to final authorisation and may be subject to change

- Once Results reviewed click Endorse Results button
Oxfordshire CCG GP Practices
Results are transmitted to relevant Practice IT systems after the result is authorised. Results are transmitted at 04:40, 07:50, 10:50, 12:50, 13:50, 14:50, 16:50, 17:50, 19:50 and 22:50 and would be available for collection by the practice IT after that time.

If there are any problems in receipt of results, please contact the Directorate IT team on (01865 2) 20463.
Contact Details jerry.dempsey@nhs.net, or Kevin.paddon@nhs.net

Oxford Haemato-Molecular Diagnostic Service
The integrated Bone Marrow Reports are available to colleagues in neighbouring partner hospitals by access to a secure website. If registered to access the Integrated Reports Website please click on: http://oxfordir.oxnet.nhs.uk. NB. It only works from an NHS networked (N3) computer.

To apply for access contact Mr Steve Harris steve.harris@ndcls.ox.ac.uk

Raid Anticoagulant Dosing Service
Results are posted direct to patients using First class post. Results which require a Recall test of the patient in less than 7 days will be phoned to the patient by the Nurse Specialist team.

All other users
Results will be produced hard copy and sent by second class mail.

NPEX
National Pathology Exchange, http://www.npex.nhs.uk/nationalexchange. The OUH is registered and has been since 2015. We are receiving requests and samples and sending reports to a variety of users by this route, to use this route please contact Mr Kevin Paddon, kevin.paddon@ouh.nhs.uk initially.
## Working Hours

<table>
<thead>
<tr>
<th></th>
<th>JRH Site</th>
<th>Horton site</th>
<th>Churchill site</th>
<th>OHTC</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>24/7/365</td>
<td>24/7/365</td>
<td>Monday – Friday</td>
<td>Monday – Friday</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>08:00 - 19:00</td>
<td>09:00 – 17:30</td>
</tr>
</tbody>
</table>

While the Routine laboratories are open for the receipt of samples and processing the most common tests some of the more specialist work will only be available when the more specialist staff in individual sections are on duty.

There is only one qualified Biomedical Scientist on duty at the following times:

<table>
<thead>
<tr>
<th>Day</th>
<th>JR2 site</th>
<th>Horton site</th>
<th>Churchill site</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saturday</td>
<td>20:30 hr – 8.30 hr</td>
<td>09:00 hr – 09:00 hr</td>
<td>N/A</td>
</tr>
<tr>
<td>Sunday</td>
<td>20:30 hr – 8.30 hr</td>
<td>09:00 hr – 09:00 hr</td>
<td>N/A</td>
</tr>
<tr>
<td>Monday – Friday</td>
<td>17:00 hr – 09:00 hr</td>
<td></td>
<td>8:00-10:30 hr</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>17:00 – 19:00 hr</td>
</tr>
</tbody>
</table>

**Specialist units:**

Are all staffed 08:30-17:00 Monday to Friday:

- Oxford Haemophilia and Thrombosis Centre, Churchill
- Molecular Haematology, JRH
- National Haemoglobinopathy Reference Laboratory, JRH
- Immunophenotyping Service, JRH
- Ante-Natal and Neonatal Screening Services, JRH

General Enquiries please see appendix II
Transfer of Specimens to the Laboratory

In the majority of clinical areas at John Radcliffe, Churchill and Horton Hospitals, samples may be sent to the laboratory in the Pneumatic tube system. Staff are encouraged to use the Air Tube system were available for all relevant samples. Use of the Air Tube system is covered by OUH policy, a copy of which is available on the OUH intranet site: (http://orhnet.orh.nhs.uk/estatesandfacilities/lib/1464/258/Air%20Tube%20procedure.pdf).

Portering services on the 4 hospital sites are marginally different and local policies should be followed but while there are some routine collections of samples from the retained estate of the Churchill Hospital in most areas there is no routine collection of samples and a portering job needs to be logged with the help-desk on each occasion.

Transfer between the sites of the Trust during routine hours is by vehicles operated by the South Central Ambulance Service (SCAS). A vehicle operates at xx:10 from Laboratory Medicine on the Churchill site via the NOC and arriving at the John Radcliffe laboratories at xx: 45, between the hours of 8:10 and 17:45 Monday to Friday. Most Haematology samples generated at the Churchill are processed on the Churchill site, but some specialist work will be sent to the John Radcliffe, all of the NOC workload will be sent to the John Radcliffe.

Between 7pm and 8am, Monday to Friday, and all day Sat, Sun and Bank Holidays a Commercial Courier “CitySprint”, provides an hourly service between Churchill, and NOC receptions and the Labs at the John Radcliffe. Acute clinical areas at the Churchill site can book additional CitySprint collections direct from the clinical areas and staff working there should make themselves familiar with the local process.

SCAS provides 4 routine collections from the Horton Laboratories at 8:00, 10:35, 14:25, and 16:30 to bring specialist work to the JRH. If necessary, the Laboratory Staff can organise additional deliveries using a local Taxi firm. Most Haematology samples generated at the Horton site will be processed at the Horton site but specialist work will be sent to the JRH.

A twice a day service from Oxfordshire G.P.’s operates for collection of samples and delivers to both the laboratories at the JRH and the Horton.

All samples that are transported by these services must be appropriately packaged and labelled. Transport of all samples should be such to guard against unauthorized access to specimens. These are examples of the approved transport boxes currently in use.
Samples sent by either Royal Mail or other courier services are the responsibility of the sender until the arrival in the Laboratory. It is their responsibility to ensure packaging meets the standard for the transport of specimens through the post. Royal Mail Group plc will accept Category B diagnostic specimens provided they are packaged to Packaging Instruction 650 requirements. Full details may be accessed on the Royal Mail website: (http://track.royalmail.com/portal/rm/content1.jsessionid=W1KZIBD3WTPKUF82IGVIFHQ?catId=40044&mediaId=36200675)

Specialist Samples:

Most samples can be sent by the best available means to arrive in the Laboratories as quickly as possible but some will be required to be processed in specialist areas of the laboratory which may not provide a 7-day service, and may not be able to be stored.

Examples include but are not limited to:

- Platelet Investigations including those linked to potential Non-Accidental Injury investigation, these samples must be tested within 2 hours of being bled. Please contact the laboratory (01865 2) 25311 in routine hours, or contact Duty SPR, via switchboard, before bleeding patient. In some cases, arrangements can be made to bleed the patient at the OHTC.
- Cold Agglutinins in Blood Transfusion, must arrive while still at 37°C, and only between 08.30 and 13.00 Mon-Friday. Contact laboratory on (01865 2) 20339 if sending sample.
- Pre-Natal diagnosis in NHRL or Haemophilia. Contact the laboratory IN ADVANCE, on (01865 5) 72769, safe details to provide result by Telephone and Fax MUST be provided.
Requesting

Blood should be correctly collected into vacutainers if at all possible (including children). The anticoagulant / sample tube required is listed in the section on normal ranges at the end of this handbook. Please remember to fully label samples with the following as a minimum:

- Full Name (first name and surname)
- Date of birth

(Blood Transfusion samples will require a higher standard of patient identification please see the relevant section of this handbook, and there is a Trust “Blood Transfusion Policies and Procedures”, a copy should be available on all wards or from on the intranet: [http://orh.oxnet.nhs.uk/BloodTransfusion/Pages/Default.aspx](http://orh.oxnet.nhs.uk/BloodTransfusion/Pages/Default.aspx)

You must make yourself familiar with it if you need to request blood for transfusion.)

Date and time of collection should be provided on all samples; as certain assays can only be performed on fresh samples.

Please use the patient's hospital number (MRN) or NHS number and give a location for the report and relevant clinical details. It is most important that requests for immediate investigations are reserved for those that are truly urgent; we will prioritize investigation of samples using information from request forms. Hospital In-patients are given priority followed by some outpatient clinics.

**OUH Requests**

The Trust has almost completed implementation of the Cerner Millennium EPR system, samples requested using this method do not require request cards. When implementation is complete only samples requested via EPR will be accepted from OUH users. Information about correct positioning of these labels can be found on the reverse of the specimen bags.

- Always check patient identification before labelling samples.
- Only put specimens from a single patient in each primary bag.
- Stick the specimen label directly over the tube label.
- Labels should be straight, vertical and as close to the cap as possible.
- If using paper forms, please ensure that on each request form, the individual collecting blood, signs the form.

**If Cerner is not used** a fully completed request form must accompany all samples (please see Appendix III, Examples of Haematology Request forms). Please ensure that the correct form is used. Information about the service and sample collection is printed on reverse of request form. It is very important to complete the request form fully and correctly. The use of Addressograph labels on request forms is encouraged, but all samples should be handwritten. Samples labelled with addressograph labels are unlikely to be processed, due to technical difficulties with the analytical equipment in use in the department.
Oxfordshire CCG GP Practices:
Locations are equipped to use the GP electronic requesting system (Sunquest-ICE) the system will identify which bottles are required for each test and produce labels suitable for use on the requests bottles and a larger label to go on the request card including eye-readable information in case of an IT-Link failure and eye readable clinical details. The date and time the patient was bleed should be added on each occasion.

It is CCG and OUH policy that all requests should include a valid NHS number.

All other locations:
Each request should be accompanied by a fully completed request form see Appendix III for examples. Patient demographics should include:
- Full name (first name and surname)
- Date of Birth
- Hospital Number
- NHS Number (or CHI number for Scottish requests)
- If your referral site expects the laboratory number to be reflected back to them, it must be included at this point.
- If your Trust has a policy of demanding Purchase Order Number for all external work it must be included at this point.
- Relevant Clinical Details
Use of addressograph labels on request cards is encouraged, but is strongly discouraged on bottles, as some designs of larger labels will interfere with the automated flow of samples through the analysers, and some referral sites (NHSBT) will only accept hand written samples because of evidence of an increased risk of patient misidentification in some circumstances with pre-printed labels.

Sample Requirements

<table>
<thead>
<tr>
<th>Tube Colour</th>
<th>Anticoagulant Used</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Trisodium Citrate" /></td>
<td>Trisodium Citrate: for all routine coagulation analysis</td>
</tr>
<tr>
<td><img src="image" alt="Potassium EDTA" /></td>
<td>Potassium EDTA: for routine Haematology analysis and transfusion investigations</td>
</tr>
<tr>
<td><img src="image" alt="Serum" /></td>
<td>Serum: for Haematinic analysis</td>
</tr>
</tbody>
</table>

Sample Types
The trust supplies BD Vacutainer tubes for the collection of adult samples, these contain a variety of different additives, either to stabilize an analyte, or to make a sample suitable for testing. It is very important that the correct tube is taken for the correct test requested. In almost all cases, if the wrong tube is received, the laboratory cannot provide a correct result. The table below displays the commonly encountered sample tube types.
If at all possible please use BD vacutainer tubes for the collection of all samples, regardless of patient age. If this is not possible, there is a selection of paediatric tubes available, these may have different lid colours to the adult tubes, so some degree of care is needed in selection. As with adult tubes, please send correctly filled samples for clotting studies.

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>Tube lid Colour (BD Vacutainer tubes)</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDTA</td>
<td></td>
</tr>
<tr>
<td>Citrate</td>
<td></td>
</tr>
<tr>
<td>Clot</td>
<td></td>
</tr>
<tr>
<td>Heparin</td>
<td></td>
</tr>
</tbody>
</table>

Please note, we still receive a number of paediatric EDTA tubes with Heparin lids on them, care must be taken not to mix up the red and orange lids.
General Haematology

For FBC, the department requires an EDTA sample, if at all possible this should be in a properly filled, well-mixed purple top vacutainer. 4.5 and 1.8ml tubes are available for use. For paediatric use, there is a 1.3ml screw top sample tube available. Please send full samples if possible, this allows for supplementary requested tests to be performed on the same sample. For other commonly requested tests please see the normal range section of this document.

As part of the Full Blood Count profile, results will be produced and reported for:

- White blood Cell Count
- Haemoglobin
- Haematocrit
- Red Cell Count
- Mean Cell Volume (MCV)
- Mean Cell Haemoglobin (MCH)
- Mean Cell Haemoglobin Concentration (MCHC)
- Platelets
- Nucleated red Cell Count

There is no need to request these tests in addition to a Full Blood Count.

As part of the Full Blood Count profile the aim will be to produce a 5-part White Cell Differential.

- Neutrophil
- Lymphocyte
- Monocyte
- Eosinophil
- Basophil

In most cases an accurate differential will be produced and reported by the analyser, in a minority of cases either the analyser will not produce a differential or will produce a report suggesting the presence of other cells in the circulation. In these cases, a Blood Film will be produced and examined by microscopy. There is no need to request a Blood Film to receive a White Cell Differential.

There are circumstances were examining a Blood Film will add value to the report if so please include clinical circumstances and why the Film is requested. Examples which would generate a Film request regardless of FBC result would include:

- ?Sezary Cells
- ?DIC
- ?Haemolysis
- ?Haemolytic Anaemia
- Persistent/ Unexplained Anaemia
- Persistent/ unexplained Thrombocytopenia

General Coagulation
- For a coagulation screen request PT and APTT or "Coagulation screen" on form
- For patients anticoagulated with warfarin request an INR
- For patients anticoagulated with unfractionated heparin request an APTT
- Patients anticoagulated with LMW heparin do not normally need monitoring

Coagulation citrate samples (blue top) need to be correctly filled due to the nature of the anticoagulant used. Partially filled, clotted or overfilled specimens will not be processed. For patients that are difficult to bleed paediatric (1.3ml) or short draw (1.8ml) vacutainers tubes are recommended. Please ensure that all samples are adequately labelled. Please see figure below for correct filling.

Some Conditions where it is appropriate to request a coagulation screen

- Unexplained bleeding or bruising
- Personal or family history of bleeding disorder
- Liver disease
- Prior to anticoagulant therapy
- Conditions associated with DIC e.g. septicaemia
- Massive transfusion
- Suspected NAI

Coagulation screens before surgery or invasive procedures


These recommendations can be summarized as follows:

- Indiscriminate coagulation screening prior to surgery or other invasive procedures for prediction of bleeding risk is not recommended
• **A comprehensive bleeding history** should be taken in **all patients** prior to surgery and invasive procedures

• If the bleeding history is **negative**, no further coagulation testing is indicated

• If the bleeding history is positive or there is a clear clinical indication (e.g. liver disease), a comprehensive **assessment guided by the clinical features is required**

**Some reasons not to request coagulation screen**

The use of coagulation screens has increased over recent years as a routine screen for haemostatic abnormalities. The tests were not designed for this purpose and their use in this manner is inappropriate for several reasons:

1. By definition, 2.5% of normal healthy subjects will have prolonged clotting times. This is likely to be higher in the patient population. A large proportion of such results will require further investigations, causing delayed operations, unnecessary anxiety to the individual, and unnecessary and expensive laboratory investigations. There are also occasions when this can erroneously precipitate the use of blood products.

2. All inherited bleeding disorders (and many acquired ones) have low prevalence. Indiscriminate screening results in low positive predictive value, and a high number of false positives.

3. The coagulation screen is insensitive to factor XIII deficiency, mild von Willebrand disease (the commonest congenital bleeding disorder in Caucasians) and platelet disorders and may give false reassurance.

4. Some factor deficiencies causing prolongation of the APTT are clinically irrelevant, e.g. factor XII deficiency. This and the lupus anticoagulant may suggest a bleeding risk when none exists, causing unnecessary postponement of surgical procedures.

5. Evidence in the literature shows that coagulation tests have both low sensitivity and specificity to predict bleeding.

6. 95% of potentially clinically significant abnormalities of coagulation or haemostasis in medical and surgical patients can be detected through a comprehensive history and physical examination.

7. Despite a large number of abnormal results being generated through indiscriminate use of the coagulation screen, studies have shown that patient management is rarely altered.

Requests for specialized bleeding investigations (especially those in patients where there is a suspicion of non-accidental injury) should be discussed with the clinical staff at the OHTC **before taking the samples**. The majority of these samples need to be reasonably fresh (<4-6 hours), so should be sent direct to the OHTC on the Churchill site.
Specialist Coagulation

Testing for Thrombophilia

Who to test for heritable thrombophilia

<table>
<thead>
<tr>
<th>Patient</th>
<th>Relative</th>
</tr>
</thead>
</table>
| • consider testing those with a strong family history of unprovoked thrombosis  
• women planning a pregnancy who have had a VTE due to a provoking factor should be tested and considered for antenatal prophylaxis if a thrombophilia is found | • Consider testing asymptomatic relatives in selected thrombosis-prone families with high risk thrombophilia (antithrombin, protein C or protein S deficiency). May be particularly helpful for counselling female relatives regarding COC and HRT.  
• women planning a pregnancy who have a family history of venous thrombosis should be tested if an event in a first degree relative was unprovoked, or provoked by pregnancy or COC exposure |

In patients, if testing is indicated it is usually performed one month after discontinuing anticoagulation with Warfarin. We do not recommend testing in the acute phase or when anticoagulated with warfarin.

Who to test for antiphospholipid antibodies

• Patients with unprovoked or recurrent VTE who are stopping anticoagulation.  
• Ischaemic Stroke < 50 years  
• Three consecutive spontaneous abortions < 10 weeks  
• Foetal death > 10 weeks  
• Premature birth due to (pre-)eclampsia or placental insufficiency

Samples required

• Hereditary thrombophilia - 4 citrate samples and 1 EDTA  
• Antiphospholipid antibodies - 1 citrate and 1 clotted  
• Both - 4 citrate samples, 1 EDTA and 1 clotted

Platelet Function Assays (Including those that form part of a Non-Accidental Injury investigation)

These are not routine tests and require medical discussion before testing. These need to be performed on fresh samples arriving at the Haemophilia centre (Churchill site) within 2 hours of collection; as such it is essential that clinical areas contact the laboratory before bleeding the patient.

In routine working hours (Monday – Friday) please phone OHTC on (01865 2) 25311  
Outside of this please bleep the on duty Haematology Specialist Registrar via switchboard

Factor assays, Nucleotides etc
Samples should be sent to:
**Haemophilia and Thrombosis Centre (OHTC)**
Old Road,
Headington,
Oxford,
OX3 7LE

Routine samples received from elsewhere should be frozen with suitable material to allow the sample to remain frozen for the entire journey. i.e. Dry ice is ideal although a frozen ice pack can be used for short (same day / fast track courier) transport. Plasma should be frozen in 2ml freezer vials with screw cap lids and labelled with -70°C freezer proof labels. Be aware samples will be thawed at 37°C in a waterbath and labels must also be resistant to this.

Samples for platelet nucleotides must be pre-prepared before being frozen as a platelet extract on dry ice. Please contact the Haemophilia and Thrombosis Centre beforehand to discuss the preparation proceed and to receive a copy of the SOP if necessary.

What will we accept?
Samples should be accompanied with a suitable request card and covering explanation letter if there are complex clinical details. If no local SLA is in place a purchase order number should be included.

Do we need Clinical information?
Yes, as much as possible including previous local results from your laboratory if available (including laboratory bar code numbers)
Specialist Haematology including Molecular Haematology

The Oxford University Hospitals NHS trust’s department of Haematology provides a comprehensive molecular diagnostic service for a range of haematological conditions. The services offered are divided into 4 main areas:

1) **Haemostasis**: Haemophilia and thrombophilia genetic testing

2) **Haemoglobinopathies**: A national service offering extensive molecular investigation of α-thalassaemia, β-thalassaemia, abnormal haemoglobins and the sickle cell syndromes.

3) **Iron Regulation**: Screening for the HFE gene mutations

4) **Haemato-oncology**: An integrated phenotypic (immunophenotyping) and molecular service for the management of haematological malignancies.

5) **Solid tumours**: Integrated pathology and genomics. CE marked diagnostics of response prediction and cancer gene mutation panel utilising next generation sequencing.

More detailed information on all aspects of our service can be obtained from our web site ([http://www.oxford-translational-molecular-diagnostics.org.uk/](http://www.oxford-translational-molecular-diagnostics.org.uk/)) or requested by e-mail from oxford.molecularhaem@nhs.net.

**General Information**

<table>
<thead>
<tr>
<th>Laboratory address for specimen reception</th>
<th>Molecular Haematology, Level 4, John Radcliffe Hospital, Headington Oxford, OX3 9DU</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lab service hours</td>
<td>9:00-5:00 Monday to Friday</td>
</tr>
<tr>
<td>E-mail for advice and enquiries:-</td>
<td><a href="mailto:oxford.molecularhaem@nhs.net">oxford.molecularhaem@nhs.net</a></td>
</tr>
<tr>
<td>Haematology molecular genetics laboratory</td>
<td>01865 572769</td>
</tr>
<tr>
<td>Immunophenotyping laboratory</td>
<td>01865 572827</td>
</tr>
<tr>
<td>Fax:</td>
<td>01865 572775</td>
</tr>
</tbody>
</table>

For more information please see [Appendix IV](#) at the end of this document.
National Haemoglobinopathy Reference Laboratory

Information for Users

Summary
The National Haemoglobinopathy Reference Laboratory was centrally funded by the DH from 1982 to 31st March 2006, after which the central funding ceased and the DH required the NHRL to charge for its service on a provider to provider basis. The central funding was devolved to all PCTs to pay for haemoglobinopathy DNA studies. The NHRL offers a service for the identification of haemoglobinopathy genotypes by the molecular analysis of DNA and haematological investigation. This includes the investigation of difficult/complex phenotypes and the identification of carrier states for antenatal patients. It also offers a prenatal diagnosis service by fetal DNA analysis for sickle cell disease, α-thalassaemia and β-thalassaemia.

DNA tests: There are 5 categories of molecular haemoglobinopathy investigations performed by the NHRL, described in detail in section 1 of this document. The five tests are:

1) α-thalassaemia mutation/s identification (all α+ and α0 types),
2) β-thalassaemia, mutation/s identification (all β, δβ-thalassaemia and HPFH deletions)
3) sickle cell disorders (all genotypes)
4) Hb variant identification (all α-chain and β-chain variants)
5) Prenatal diagnosis for α-thalassaemias, β-thalassaemias and sickle cell disorders

Contact Details
Email: lab.support@nhs.net
Telephone: (01865 5) 72769

For more information please see Appendix V at the end of this document.
## Retrospective Testing

In accordance with local policies, the department stores specimens for a period of time post analysis in conditions suitable for retrospective or additional test requests. With certain analysis however there is a time limit outside of which the stored sample is likely to unsuitable for processing. The following table will give information on such time limits for commonly encountered tests. For any tests that are not on this list, please contact the department for advice. Outside of the times stated on this list new samples will be required.

<table>
<thead>
<tr>
<th>Test</th>
<th>Availability</th>
<th>Storage comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>FBC</td>
<td>All sites,</td>
<td>Samples are suitable for processing however up to 48 hours from collection. Samples however are retained by the department for 24 hours post analysis, so may not be available for use.</td>
</tr>
<tr>
<td>ESR</td>
<td>Horton and JR sites</td>
<td>Samples if available are suitable for testing up to 24 hours post collection.</td>
</tr>
<tr>
<td>Plasma Viscosity</td>
<td>JR site</td>
<td>Samples if available are suitable for testing up to 5 days post collection, however samples are only stored for a minimum time period.</td>
</tr>
<tr>
<td>Glandular Fever Screen</td>
<td>Horton and JR sites</td>
<td>Serum samples, if available, are suitable for testing up to 72 hours post collection. EDTA samples, if available, are suitable for testing up to 24 hours post collection.</td>
</tr>
<tr>
<td>Blood Films / Malaria Film</td>
<td>Horton and JR sites</td>
<td>Samples are suitable for testing up to 24 hours post collection, on a suitable EDTA sample.</td>
</tr>
<tr>
<td>Flow Cytometry</td>
<td>JR site</td>
<td>Samples if available are suitable for testing up to 72 hours post collection, on a suitable EDTA sample.</td>
</tr>
<tr>
<td>Sickle Test</td>
<td>Horton and JR sites</td>
<td>Samples if available are suitable for testing up to 4 days post collection.</td>
</tr>
<tr>
<td>Haemoglobinopathy Screen</td>
<td>JR &amp; Churchill site</td>
<td>Samples if available are suitable for testing up to 4 days post collection.</td>
</tr>
<tr>
<td>PT / INR</td>
<td>All sites,</td>
<td>Samples are suitable for testing up to 24 hours post collection.</td>
</tr>
<tr>
<td>APTT</td>
<td>All sites,</td>
<td>Samples are suitable for testing up to 8 hours post collection.</td>
</tr>
<tr>
<td>Thrombin Time</td>
<td>All sites</td>
<td>Samples are suitable for testing up to 8 hours post collection.</td>
</tr>
<tr>
<td>Fibrinogen</td>
<td>All sites</td>
<td>Samples are suitable for testing up to 8 hours post collection.</td>
</tr>
<tr>
<td>Assay Type</td>
<td>Site</td>
<td>Sample Storage Criteria</td>
</tr>
<tr>
<td>------------------------------------</td>
<td>---------------------------</td>
<td>-----------------------------------------------------------------------------------------</td>
</tr>
<tr>
<td>D-Dimers</td>
<td>All sites, JR &amp; OHTC</td>
<td>Samples are suitable for testing up to 8 hours post collection</td>
</tr>
<tr>
<td>Factor Assays</td>
<td>JR &amp; OHTC, JR sites</td>
<td>Samples are suitable for testing up to 8 hours post collection, however fresh samples are strongly recommended</td>
</tr>
<tr>
<td>Haematinc Assays</td>
<td>Horton and JR sites</td>
<td>Samples if available are suitable for testing up to 72 hours post collection.</td>
</tr>
<tr>
<td>(including Red Cell Folates)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Haptoglobins</td>
<td>Horton site</td>
<td>Samples if available are suitable for testing up to 72 hours post collection.</td>
</tr>
<tr>
<td>DAT</td>
<td>Horton and JR sites</td>
<td>Samples if available are suitable for testing up to 72 hours post collection.</td>
</tr>
<tr>
<td>Molecular processes involving RNA testing</td>
<td>JR2</td>
<td>Samples are suitable for testing up to 72 hours post collection, on a suitable EDTA sample</td>
</tr>
<tr>
<td>Molecular processes involving DNA testing</td>
<td>JR2</td>
<td>Samples if available are suitable for testing for an unlimited period post collection, however samples are only stored for a minimum time period</td>
</tr>
</tbody>
</table>

**Sample Analysis Turnaround times**

This document provides information on turnaround times for commonly encountered assays offered by the department. During routine processing, the department will endeavour to process samples to within these stated time limits. In situation of reduced staffing or unexpected analyser failure, processing times may be longer. The times stated are in-laboratory turnaround times and do not take any account of delivery of sample to department or delivery of report to final location. For any process not covered and for samples stated as urgent, we will attempt to process them as rapidly as practical, within operational constraints.

<table>
<thead>
<tr>
<th>Analysis</th>
<th>Urgency</th>
<th>Turnaround (unless stated this refers to 95% of samples being processed in the given time period)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Routine Haematology</td>
<td>FBC: Out-patient / GP</td>
<td>98% &lt;48 hours ¹</td>
</tr>
<tr>
<td></td>
<td>FBC: In-patient</td>
<td>3 hours</td>
</tr>
<tr>
<td></td>
<td>FBC: A&amp;E</td>
<td>95% &lt; 60 minutes ⁵</td>
</tr>
<tr>
<td></td>
<td>Routine Plasma Viscosity</td>
<td>72 hours</td>
</tr>
<tr>
<td>Morphology</td>
<td>Malaria Screen</td>
<td>7 hours</td>
</tr>
<tr>
<td></td>
<td>Routine Blood Film</td>
<td>36 hours (Mon – Fri)</td>
</tr>
<tr>
<td>Coagulation</td>
<td>Pre-op coagulation screen</td>
<td>5 hours</td>
</tr>
<tr>
<td></td>
<td>Urgent coagulation screen (A&amp;E)</td>
<td>95% &lt; 1 hour ⁵</td>
</tr>
<tr>
<td>Test</td>
<td>Turnaround Time</td>
<td></td>
</tr>
<tr>
<td>----------------------------------------------</td>
<td>-----------------</td>
<td></td>
</tr>
<tr>
<td>Urgent INR (Warfarin Control)</td>
<td>1 hour</td>
<td></td>
</tr>
<tr>
<td>Urgent APTT (Heparin control)</td>
<td>1 hour</td>
<td></td>
</tr>
<tr>
<td>INR (GP only)</td>
<td>98% &lt; 24 hours $^1$</td>
<td></td>
</tr>
<tr>
<td>Urgent D-Dimer (DVT screen)</td>
<td>1 hour</td>
<td></td>
</tr>
<tr>
<td>Urgent Factor deficiency investigation</td>
<td>5 hours</td>
<td></td>
</tr>
<tr>
<td>Factor deficiency Investigation</td>
<td>1 week</td>
<td></td>
</tr>
<tr>
<td>Thrombophilia Screen</td>
<td>3 weeks</td>
<td></td>
</tr>
<tr>
<td>Haematinics</td>
<td></td>
<td></td>
</tr>
<tr>
<td>B12 / Folate / Ferritin</td>
<td>48 hours</td>
<td></td>
</tr>
<tr>
<td>Erythropoietin</td>
<td>1 week</td>
<td></td>
</tr>
<tr>
<td>Molecular</td>
<td></td>
<td></td>
</tr>
<tr>
<td>For molecular TAT, please see Appendix VI Oxford BRC Haemato-Molecular Diagnostic Service</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Immunophenotyping</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leukaemia diagnosis</td>
<td>Formal report 1 week</td>
<td></td>
</tr>
<tr>
<td>Blood Transfusion</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Routine Group &amp; Antibody Screen</td>
<td>24 hours $^2$</td>
<td></td>
</tr>
<tr>
<td>Urgent Group &amp; Antibody Screen</td>
<td>3 hours $^2$</td>
<td></td>
</tr>
<tr>
<td>Routine Antenatal Serology</td>
<td>48 hours $^3$</td>
<td></td>
</tr>
<tr>
<td>Routine Post Natal Rh Negative testing</td>
<td>36 hours</td>
<td></td>
</tr>
<tr>
<td>Urgent Crossmatch request</td>
<td>1 hour $^2$</td>
<td></td>
</tr>
</tbody>
</table>

$^1$Turnaround times, as agreed with local PCT’s
$^2$Turnaround times are only achievable on patients with no special transfusion requirements and a negative antibody screen.
$^3$This excludes any samples that require further investigations
$^4$Depending on the complexity of the investigation
$^5$Locally agreed in laboratory TAT with ED (January 2010)

**Key Factors known to affect result quality**
Although all analytical methods used by the department are appropriately controlled by internal and external quality assurance methods, there are some factors that can affect the specific analytical methods. This document will cover the factors affecting the most common tests; information on other tests is available from the relevant sections on request.

- Correct sample collection, storage and transport to the laboratory: can minimize sample degradation pre analysis. For any analysis that requires whole (EDTA) blood, samples which contain clots may not be suitable for processing.
- Correct sample volume: for certain tests (Coagulation screens), this is essential as the tubes contain a liquid anticoagulant. For other tests, there will be a minimum amount of sample required for correct analysis. Samples that do not contain this will not be processed.
- Sample age: in general samples should be processed without delay post collection. However, with some tests the delay is more critical than with others. Please refer to the retrospective testing table for information on this.
- Haemoglobin: this test includes a spectrophotometric process, so excessive jaundice and lipaemia may affect the result. This can be detected in the laboratory and corrected for before releasing results.
- Platelet clumping: this is a non-specific immunological process that can lead to a falsely low platelet count. The current analysers used in the department, check for this and the laboratory will amend results appropriately.
- Clotting tests: may be affected by the presence of anticoagulants; however, this affect will depend on the specific test and the specific anticoagulant. More information is available from the OHTC on ext 25311.
- Certain clotting tests (platelet function analysis) cannot be transported to the department using the trust air tube system. In addition, these samples need to be as fresh as possible, so please contact the OHTC on ext. 25311 before sample collection.
- Haemoglobinopathy screens: results will be affected by the patient receiving recent blood transfusions or Bone marrow transplant. This should be discussed with the section before sending the sample as it may be prudent to defer testing for a period of time.
- HbA2 levels: can be lowered in severe iron deficiency, clinical advice should be sought before testing.
- Sickle Screening: Results obtained from neonates (< 6 months old) may be unreliable because of the low percentage of HbS and the high percentage of HbF. Abnormally high levels of plasma protein (e.g. Myeloma) may cause false positive results. The kit may not reliably produce a positive result in patients who have an HbS level of less than 20%. Samples that have HGB less than 6.0 g/dl are deemed grossly anaemic and the sickle screen results may be affected due to the larger plasma: red cell ratio.
- Glandular Fever (infectious mononucleosis screen): False positive result may occur in serum samples from patients with a recent infection of cytomegalovirus, leptospirosis, hepatitis A and parvovirus. Some patients do not develop heterophile antibodies (<20% adults and 50% children).
- Haptoglobin: The assay is unsuitable as a measure of haemolysis in Children as they may have physiologically lower serum levels. In conditions where acute phase proteins are elevated a normal or high Haptoglobin level may not exclude intravascular haemolysis, similarly pregnancy, malignancy, biliary obstruction, steroid therapy and oral contraception may show normal or high levels despite haemolysis.
- Molecular testing for somatic mutations will be affected by undeclared bone marrow transplants; clinical advice should be sought before testing.
- Molecular testing for somatic BRAF mutations may be affected by the presence of endogenous melanin in FFPE samples and lead to an invalid test result.
- Blood transfusion samples: results obtained from these samples may be affected if the patient has recently received a blood transfusion or Bone marrow transplant. Because of this, it is very important to ensure that the blood bank is aware of this information.
Requests for Blood and Blood Products for Transfusion

There is a Trust “Blood Transfusion Policies and Procedures”, a copy should be available on all wards or from on the intranet: http://orh.oxnet.nhs.uk/BloodTransfusion/Pages/Default.aspx
You must make yourself familiar with it if you need to request blood for transfusion.

When trained and deemed competent doctors, nurses, phlebotomists and medical students may take blood specimens for grouping and crossmatching. Staff who are untrained are not permitted to take blood samples for transfusion.

Particular attention must be paid to positive identification of the patient based on interrogation of the patient where possible, a wristband and the patient's hospital notes.

As a further safeguard, in addition to patient's name and other relevant information on the specimen and request card, BloodTrack Tx system must be used. This is an electronic system for which ensures that the patient sample is labelled correctly. It uses bar-coded wristbands in combination with a handheld computer to ensure positive patient identification and produce an on demand printed patient identification label. These labels are suitable for labelling a blood transfusion sample (NOTE: an addressograph label is not suitable). The laboratory will not proceed with any incorrectly or inadequately labelled specimens.

There is an expectation that samples will be accompanied by an electronic EPR request. Only in exceptional circumstances will samples be accepted with a completed signed request card.

Please note that a routine group and crossmatch will take a minimum of 3 hours. For planned transfusions including pre-planned surgical procedures, the specimen should arrive at the laboratory a full working day before the blood is required. Routine specimens received in the laboratory after 20:00 hours may not be processed until the following day in this case blood will be available by 10:00 if no atypical antibodies are present.

Patients with atypical antibodies
If you require blood for a patient with known red cell antibodies, please ensure the laboratory are given as much notice as possible. Although red cells suitable for most commonly occurring antibodies are kept in stock, for more unusual antibodies/combinations blood will need to be ordered from the NHSBT. For some combinations obtaining suitable red cells is difficult and there will be a considerable delay.

Emergency Transfusion including Massive Haemorrhage
The trust has a Massive Haemorrhage Protocol (MHP) which is available at http://ouh.oxnet.nhs.uk/BloodTransfusion/Pages/MajorHaemorrhageProtocol.aspx

There is an expectation that clinical staff will be aware of how to activate the protocol and where a copy can be located.

Activation is by calling switchboard on 4444 – use the term activate the Major Haemorrhage Protocol and clearly state you site and location.

Other blood products

- 5% or 20% Human Albumin Solution,
- Platelets
- Fresh Frozen Plasma,
- Cryoprecipitate
- Prothrombin Complex concentrate (Octaplex)
- Prophylactic Anti-D

These are available on a named patient basis from the laboratory upon discussion with the staff on duty.
- FFP and Cryoprecipitate should only be ordered if it is to be used immediately.
- The department will not thaw out FFP and Cryoprecipitate on a "standby basis", although FFP may be available already thawed from within the laboratory.

**Blood Ordering Schedule**

The Trust has implemented a comprehensive system of remote blood issue in most of the theatre suites. This ensures that blood is available on request directly at the point of need for most operations. As such we no longer use a Blood Ordering schedule in its traditional sense.

**Transfusion Sample Requirements**

These are adult (> 7 years old) sample volume requirements, for children under 7 years old a minimum of 1.5 ml is required for group save and crossmatch. More may be required for patients requiring complex antibody investigations.

<table>
<thead>
<tr>
<th>Tube Colour</th>
<th>Investigation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><strong>Group and Save</strong> <em>(Full, 4.5 ml EDTA)</em> 2 x EDTA if patient known to have antibodies.</td>
</tr>
<tr>
<td></td>
<td><strong>Crossmatch Request</strong> <em>(Full, 4.5 ml EDTA)</em> 2 x EDTA if patient known to have antibodies</td>
</tr>
<tr>
<td></td>
<td><strong>Antenatal Serology</strong>: Full, 4.5 ml EDTA on mother. Other samples may need to be sent if haematology and haemoglobinopathy screening is required.</td>
</tr>
<tr>
<td></td>
<td><strong>Kleihauer test</strong> <em>(2ml EDTA)</em> Routine samples on Rhesus negative mothers at delivery (2 ml EDTA on mother and baby)</td>
</tr>
<tr>
<td></td>
<td><strong>Direct Coombs Test</strong> <em>(DAT)</em> <em>(2ml EDTA)</em></td>
</tr>
<tr>
<td></td>
<td><strong>Cold Agglutinins</strong> <em>(Clot delivered to laboratory at 37°C in thermos flask, discuss the test with laboratory staff prior to collecting a sample)</em></td>
</tr>
</tbody>
</table>
Samples for Antenatal Testing:
Samples sent to the laboratory for antenatal testing should comply with the labelling requirements indicated above. In addition, if a patient is not yet registered with the Trust and for whom there is no NHS number available, the laboratory will accept the patients FULL address as a patient identifier (this will not be acceptable on samples for compatibility testing)
### Appendix I Normal Ranges (Adult, 13 years+)
(Paediatric Normal ranges are available by contacting the JR2 site on extension 20336)

#### FBC

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Unit(s)</th>
<th>Male (13-70yr)</th>
<th>Female (13-70yr)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Haemoglobin (Hb) (EDTA)</td>
<td>g/l</td>
<td>130 - 170</td>
<td>120 - 150</td>
</tr>
<tr>
<td>Red cell count (RBC) (EDTA)</td>
<td>x10^{12}/l</td>
<td>4.5 - 5.5</td>
<td>3.8 - 4.8</td>
</tr>
<tr>
<td>Haematocrit (Hct) (EDTA)</td>
<td>l/l</td>
<td>0.40 - 0.50</td>
<td>0.36 - 0.46</td>
</tr>
<tr>
<td>Mean Cell Volume (MCV) (EDTA)</td>
<td>fl</td>
<td>83 - 101</td>
<td></td>
</tr>
<tr>
<td>Mean cell Hb (MCH) (EDTA)</td>
<td>pg</td>
<td>27 - 32</td>
<td></td>
</tr>
<tr>
<td>Mean cell Hb concentration (MCHC) (EDTA)</td>
<td>g/l</td>
<td>315-345</td>
<td></td>
</tr>
<tr>
<td>Red Cell Distribution Width (RDW) (EDTA)</td>
<td>%</td>
<td>11.5 – 16.0</td>
<td></td>
</tr>
<tr>
<td>White cell count (WBC) (EDTA)</td>
<td>x10^9/l</td>
<td>4.0-11.0</td>
<td></td>
</tr>
<tr>
<td>Differential</td>
<td></td>
<td>Neutrophil 2.0 -7.0</td>
<td>x10^9/l</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Lymphocyte 1.0 -3.0</td>
<td>x10^9/l</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Monocytes 0.2 -1.0</td>
<td>x10^9/l</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Eosinophil 0.02 -0.5</td>
<td>x10^9/l</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Basophil 0.02 -0.1</td>
<td>x10^9/l</td>
</tr>
<tr>
<td>Immature Granulocytes (IG) (EDTA)</td>
<td>x10^9/l</td>
<td>0 – 0.1</td>
<td></td>
</tr>
<tr>
<td>Platelets (Plt) (EDTA)</td>
<td>x10^9/l</td>
<td>150-400</td>
<td></td>
</tr>
<tr>
<td>Mean Platelet Volume (MPV) (EDTA)</td>
<td>fl</td>
<td>9 – 12.1</td>
<td></td>
</tr>
<tr>
<td>Reticulocytes (EDTA)</td>
<td>%</td>
<td>0.5 - 2.5</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.04 – 0.12</td>
<td>x10^{12}/l</td>
</tr>
<tr>
<td>Nucleated RBC (EDTA)</td>
<td>%</td>
<td>0 – 0.2</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0 – 0.5</td>
<td>x10^9/l</td>
</tr>
</tbody>
</table>

#### ESR

| Erythrocyte Sedimentation Rate (ESR) (EDTA) | Male (17-69yr) | <14 | mm/hr |
|                                           | Male (>70 yr.) | <30 | mm/hr |
|                                           | Female (17-69yr) | <20 | mm/hr |
|                                           | Female (>70) | <35 | mm/hr |
| Plasma Viscosity at 25°C (EDTA)           | 1.50 - 1.72 MPa/s |

#### COAGULATION
(all require citrate tubes)

<p>| Prothrombin time (PT) (Citrate) | 9.0 – 12.0 sec |
| Activated Partial Thrombinplastin Time (APTT) (Citrate) | 20.0 - 30.0 sec |</p>
<table>
<thead>
<tr>
<th>Test Description</th>
<th>Reference Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>International Normalised Ratio (INR)</td>
<td>2.0-4.0*</td>
</tr>
<tr>
<td>(*Discuss with anticoagulation nurse specialist)</td>
<td></td>
</tr>
<tr>
<td>Thrombkin time (TT)</td>
<td>14.0-19.0 sec</td>
</tr>
<tr>
<td>Reptilase Time</td>
<td>12.0 – 16.0 sec</td>
</tr>
<tr>
<td>Fibrinogen (Fib)</td>
<td>1.5-4.0 g/l</td>
</tr>
<tr>
<td>D-Dimers</td>
<td>&lt; 500 µg/l FEU</td>
</tr>
<tr>
<td>Protein C</td>
<td>0.70 – 1.40 IU/ml</td>
</tr>
<tr>
<td>Protein S Free</td>
<td>Male 0.70 - 1.5.0 IU/ml</td>
</tr>
<tr>
<td></td>
<td>Female 0.55 – 1.35 IU/ml</td>
</tr>
<tr>
<td>Antithrombin (AT)</td>
<td>0.80 – 1.20 IU/ml</td>
</tr>
<tr>
<td>Dilute Russell’s Viper Venom Tests (DRVVT) ratio</td>
<td>0.80 - 1.20</td>
</tr>
<tr>
<td>Actin FSA ratio</td>
<td>0.70 – 1.30</td>
</tr>
<tr>
<td>ADAMTS 13 activity</td>
<td>40 - 130 IU/dl</td>
</tr>
<tr>
<td>Anti-Xa assay (Peak treatment dose given BD)</td>
<td>0.50 - 1.00 U/ml</td>
</tr>
<tr>
<td>Von-Willebrand Factor (AG &amp; activity)</td>
<td>0.50– 2.00 IU/ml</td>
</tr>
<tr>
<td>Factor VIII (Citrate)</td>
<td>0.50 – 2.00 IU/ml</td>
</tr>
<tr>
<td>Factor IX (Citrate)</td>
<td>0.50 – 2.00 IU/ml</td>
</tr>
<tr>
<td>Factor XI (Citrate)</td>
<td>0.70 – 1.30 IU/ml</td>
</tr>
<tr>
<td>Factor XII (Citrate)</td>
<td>0.50 – 2.00 IU/ml</td>
</tr>
<tr>
<td>Antiplasmin (Citrate)</td>
<td>0.80 – 1.30 IU/ml</td>
</tr>
<tr>
<td>PFA (Citrate)</td>
<td>Collagen/ADP 55 - 112 sec</td>
</tr>
<tr>
<td></td>
<td>Collagen / Epinephrine 79 - 164 sec</td>
</tr>
<tr>
<td>Platelet Nucleotides (Citrate)</td>
<td>ATP 50-300 pMol/10⁹ pltts ratio</td>
</tr>
<tr>
<td></td>
<td>ADP 30-200 pMol/10⁹ pltts ratio</td>
</tr>
<tr>
<td></td>
<td>ratio 0.8 – 2.2 ratio</td>
</tr>
</tbody>
</table>

**HAEMATINIC ASSAYS**

<table>
<thead>
<tr>
<th>Test Description</th>
<th>Reference Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Erythropoietin (EPO) (Clot)</td>
<td>5.0 - 25.0 iu/l</td>
</tr>
<tr>
<td>Iron (Fe) (Clot)</td>
<td>Male 14-31 µmol/l</td>
</tr>
<tr>
<td></td>
<td>Female 11-30 µmol/l</td>
</tr>
<tr>
<td>Transferrin (Clot)</td>
<td>1.8 - 3.6 g/dl</td>
</tr>
<tr>
<td>Transferrin Saturation (Clot)</td>
<td>16-50 %</td>
</tr>
<tr>
<td>Serum Folate (Clot)</td>
<td>3 - 20.0 µg/l</td>
</tr>
<tr>
<td>Red Cell Folate (Heparin / EDTA)</td>
<td>125-650 µg/l</td>
</tr>
<tr>
<td>Vitamin B12 (B12) (Clot)</td>
<td>180-900 ng/l</td>
</tr>
<tr>
<td>Ferritin (Clot)</td>
<td>Male 20-300 µg/l</td>
</tr>
<tr>
<td></td>
<td>Female 10-200 µg/l</td>
</tr>
</tbody>
</table>
HAEMOGLOBINOPATHY

Sickle test (Heparin / EDTA)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>HbF</td>
<td>&lt; 1.0 %</td>
</tr>
<tr>
<td>HbA2</td>
<td>1.5 - 3.0 %</td>
</tr>
</tbody>
</table>

Thalassaemia screen (EDTA)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>HbF</td>
<td>&lt; 1.0 %</td>
</tr>
<tr>
<td>HbA2</td>
<td>1.5 - 3.0 %</td>
</tr>
</tbody>
</table>

RED CELL ENZYME ETC.

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glucose-6 Phosphate Dehydrogenase (G6PD) (Heparin / EDTA)</td>
<td>4.6 - 13.5 u/gHbg</td>
</tr>
<tr>
<td>Pyruvate Kinase (PK) (send away test) (Heparin / EDTA)</td>
<td>6.2-14.2 u/gHbg</td>
</tr>
<tr>
<td>Haptoglobins (Heparin / Clot)</td>
<td>0.4 - 2.4 g/l</td>
</tr>
</tbody>
</table>

Ranges updated: 04/01/17. Ranges Reviewed 04/01/17

Reference range source

Ranges have been derived from a number of reputable sources.

1. Adult Normal FBC & ESR Ranges derived from Practical Haematology; 11th edition Dacie & Lewis 2012; checked and verified as part of new equipment implementation 2015.
3. Coagulation and Haematinic ranges derived from manufacturers recommendations, but checked and amended as part of new equipment verification 2015.
4. PK range supplied by external referral laboratory
5. G6PD range supplied by manufacturer of assay and verified by laboratory
6. Haptoglobin reviewed and updated as part of a formal verification of method 2016
Appendix II, Departmental Telephone Numbers

### General Enquiries

<table>
<thead>
<tr>
<th>Service</th>
<th>Ext</th>
</tr>
</thead>
<tbody>
<tr>
<td>Combined Haematology / Biochemistry office (JRH)</td>
<td>(01865 2) 20336</td>
</tr>
<tr>
<td>Haematology / Coagulation (Horton)</td>
<td>(01295 2) 29369</td>
</tr>
<tr>
<td>Oxford Haemophilia &amp; Thrombosis Centre</td>
<td>(01865 2) 25311</td>
</tr>
<tr>
<td>Blood Transfusion Laboratory (JR2 &amp; Churchill)</td>
<td>(01865 2) 20339 / 20340</td>
</tr>
<tr>
<td>Blood Transfusion (Horton)</td>
<td>(01295 2) 29236</td>
</tr>
<tr>
<td>Molecular Diagnostics (JRH)</td>
<td>(01865 5) 72769</td>
</tr>
<tr>
<td>Immunophenotyping Laboratory (JRH)</td>
<td>(01865 5) 72827</td>
</tr>
</tbody>
</table>

### Medical Assistance – Monday to Friday 8:30-17:00 only

- Blood Transfusion Duty Registrar                                    (01865 741166) Bleep 6888
- Haemostasis Duty Registrar                                           (01865 741166) Bleep 5529
- Haematology Duty Registrar                                            (01865 741166) Bleep 1836

Duty Registrar outside of core hours ask Switchboard to bleep duty Haematology Registrar.

For **non-urgent** clinical enquiries please email: HaematologyRegistrarEnquiries@nhs.net
For general Molecular enquiries please email: oxford.molecularhaem@nhs.net
For all National Haemoglobinopathy Reference Service queries lab.support@nhs.net
For Haemoglobinopathy screening queries please hbopathy.screening@nhs.net

### Staff

<table>
<thead>
<tr>
<th>Role</th>
<th>Ext</th>
</tr>
</thead>
<tbody>
<tr>
<td>Registrars (JR)*</td>
<td>01865 (2) 20367 or bleep 1836</td>
</tr>
<tr>
<td>Registrars (Churchill)*</td>
<td>01865 (2) 35884 / 35885 or bleep 1836</td>
</tr>
<tr>
<td>Laboratory Manager - Mr. D Smith</td>
<td>01865 (2) 20337</td>
</tr>
<tr>
<td>Quality Manager – Mr. A Platt</td>
<td>01865 (8) 57663</td>
</tr>
<tr>
<td>Blood Bank Manager – Miss J Staves**</td>
<td>01865 (2) 20334</td>
</tr>
<tr>
<td>Morphology Lead – Mr K Leyden**</td>
<td>01865 (8) 51084</td>
</tr>
<tr>
<td>Coagulation Contact – Mr. P Baker**</td>
<td>01865 (8) 57096</td>
</tr>
<tr>
<td>Automated Haematology contact – Mrs. M McClure**</td>
<td>01865 (2) 21125</td>
</tr>
<tr>
<td>Molecular Lead – Dr S Henderson**</td>
<td>01865 (5) 72766</td>
</tr>
<tr>
<td>Laboratory secretary</td>
<td>01865 (5) 72824</td>
</tr>
<tr>
<td>Dr Hatton’s Secretary</td>
<td>01865 (2) 35886</td>
</tr>
<tr>
<td>Dr Keeling’s Secretary</td>
<td>01865 (2) 25318</td>
</tr>
</tbody>
</table>
Dr Littlewoods’s Secretary 01865 (2) 35882
Dr A Peniket's Secretary 01865 (2) 35259

*For non-urgent clinical enquiries please use the dedicated Haematology registrar email HaematologyRegistrarEnquiries@nhs.net. This will be checked on a daily basis only, for urgent enquiries please phone or bleep.

** These contacts should be used if there is a non-routine issue with a specific department, otherwise please use the general enquiries number.
Appendix III, Examples of Haematology Request forms

Please note: it is expected that the majority of requests made to the department will be made on either Cerner EPR (inpatients / outpatients) or via Sunquest ICE (GP). If available in your clinical areas the laboratory would urge you to use these systems instead of using paper forms.

If using paper forms, please ensure that on each request form, the individual collecting blood, signs the form.

Haematology / Biochemistry Joint Requesting form (in use from January 2007)

This form has replaced both JR and Horton Haematology & Biochemistry forms; this form is to be used, only if EPR ordering is not available

1. Please fill in the patients’ full name & Date of Birth; insufficiently labelled samples or cards will not be tested. If available, it is acceptable to use addressograph labels on request forms.
2. Please use either NHS or Hospital number on all requests; it enables the department to merge results with previous records on the patient.
3. Clinical information is very important, especially for some Biochemistry requests. Please do not leave this blank.
4. Please include a patient location on all request cards, it will enable the laboratory to telephone the results if abnormal or if the sample is unsuitable for testing. For GP patients, please remember to give the requesting GP’s name and location.
5. Please list the tests required, for Haematology & Biochemistry. For information on sample requirements, please refer to section in laboratory handbook. Please ensure that you have taken sufficient blood in suitable tubes for the required tests. If the sample is urgent, please make this clear on the form.

6. Please fill in date and time of sample collection.

**Blood Transfusion Form:** This form is being used on all sites not using EPR ordering

1. Patient’s full name must be on the form and on the sample, not the one that the patient “likes to be known by”. The correct hospital number or NHS number and date of birth must be on sample and form. If they are missing or do not match, the sample will not be processed. The use of addressograph labels on request forms (not samples) is encouraged.

2. If the SafeTx system is being used, there is no need to use the Red label system. Please ensure that a SafeTx label is on the request card and sample. If using the Red Label system, the red label must be on the form, sample, and patient’s wristband and in the patient’s notes. If the red label is missing from any of these, new samples will necessary. Incorrectly labelled forms will not be processed.

3. All forms must be signed, at least by the doctor requesting the transfusion: no signature, no processing of sample. It is important that we can work out who you are, so please write clearly!

4. It is essential to the blood bank that this section is filled in as completely as possible. It is of particular importance if the patient is thought to have red cell antibodies, or has been transfused in another hospital.
5. Finally, tell us what you want, when you want it and where you want it. If blood is truly required urgently then telephone the department to let us know that the sample is coming. If the blood is to cover surgery, consult the maximum blood-ordering schedule.

**JR & Horton Blood bank Antenatal Request form.**

As of January 2007, all antenatal (AN) samples will be processed on the JR site, please use this form for such samples

1. Please ensure that you put the following information as a minimum on the form in this section:
   a. NHS number (Hospital number is acceptable)
   b. Full patient name.
   c. Date of birth
2. Please enter the applicable ethnic code.
3. Please ensure that the following information is included on this form:
   a. Midwife location code
   b. GP name
   c. GP location
   d. Midwife name and contact details
   e. Date (and time) sample taken
4. Please ensure that all the questions are answered in this section
5. Please send sufficient samples for the requested tests. Current departmental policy does not allow samples to be shared for FBC and Blood grouping:
   a. Blood Group & Antibody Screen – 1x EDTA
   b. FBC / Sickle / Thal screen – 1 x EDTA
c. Please make sure that a family origins questionnaire is included with all requests for Sickle or Thal screens.

6. If the sample is from a partner of an AN patient, it is very important that the AN patient details are completed in this section. The majority of partner sample request forms will be labelled with a yellow sticker.

In addition to this information, please ensure that you read the back of the request form.

**Molecular Haematology**: a variety of specialist Request forms are downloadable from the specialist website [http://www.oxford-translational-molecular-diagnostics.org.uk/content/forms](http://www.oxford-translational-molecular-diagnostics.org.uk/content/forms)
Appendix IV, Oxford BRC Haemato-Molecular Diagnostic Service
Short User Guide

The Oxford University Hospitals NHS trust’s department of Haematology provides a comprehensive molecular diagnostic service for a range of haematological conditions. The services offered are divided into 4 main areas:

1) **Haemostasis:** Haemophilia and thrombophilia genetic testing

2) **Haemoglobinopathies:** A national service offering extensive molecular investigation of α-thalassaemia, β-thalassaemia, abnormal haemoglobins and the sickle cell syndromes.

3) **Iron Regulation:** Screening for the HFE gene mutations

4) **Haemat-oncology:** An integrated phenotypic (immunophenotyping) and molecular service for the management of haematological malignancies.

5) **Solid tumours:** Integrated pathology and genomics. CE marked diagnostics of response prediction and cancer gene mutation panel utilising next generation sequencing.

This document is intended as a brief and provisional introduction to our services. More detailed information on all aspects of our service can be obtained from our web site ([http://www.oxford-translational-molecular-diagnostics.org.uk/](http://www.oxford-translational-molecular-diagnostics.org.uk/)) or requested by e-mail from oxford.molecularhaem@nhs.net.

**General Information**

<table>
<thead>
<tr>
<th>Laboratory address for specimen reception</th>
<th>Molecular Haematology, Level 4, John Radcliffe Hospital, Headington Oxford, OX3 9DU</th>
</tr>
</thead>
</table>

Lab service hours 9:00-5:00 Monday to Friday

Enquiries and information:-
Website: [http://www.oxford-translational-moleculardiagnostics.org.uk/](http://www.oxford-translational-moleculardiagnostics.org.uk/)
E-mail for advice and enquiries:
Haematology molecular genetics laboratory
Immunophenotyping laboratory
Fax:

oxford.molecularhaem@nhs.net
01865 572769
01865 572827
01865 572775

Clinical and BRC Research leads
Scientific Director
Business Manager
Haematology Laboratory Manager
Dr Anna Schuh MD, PhD, MRCP, FRCPath
Dr Chris Hatton FRCP, FRCPath
Dr Shirley Henderson MSc PhD
Dr Nick Housby PhD
Mr Dan Smith C.Sci. FIBMS

Request Form and Samples

All samples should be accompanied by a completed request form (page 6). For haemoglobinopathy investigations, a more detailed NHRL request form is available from our website. Specimens and forms should have a minimum of 4 patient identifiers including patient surname, first name, dob and hospital number.

Please provide as much clinical and laboratory information as possible, including a brief clinical history and any others recent results available on the patient. Indicate on the form the sample type, date of collection and the investigation that you are requesting. Please remember to give full contact details for results and reports.

The sample type required for each investigation is shown in the appropriate section below. All samples should be addressed to Molecular Haematology and sent to the specimen reception of the Haematology Laboratory at the John Radcliffe Hospital, Level 4. Address is given in general information (page 1).

Investigations Offered

1) Haemostasis

<table>
<thead>
<tr>
<th>Disorder</th>
<th>Tests</th>
<th>Specimen Required</th>
<th>Turnaround Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Haemophilia A</td>
<td>$F_8$ gene intron 22 inversion (inverse PCR) $F_8$ gene intron 1 inversion (inverse PCR) $F_8$ mutations by direct sequencing Carrier analysis and full genetic screen Dosage analysis (MLPA) for partial/complete $F_8$ gene deletions/duplications</td>
<td>4ml EDTA peripheral blood</td>
<td>2-8 weeks depending on complexity</td>
</tr>
<tr>
<td>Haemophilia B</td>
<td>$F_9$ mutations by direct sequencing Carrier analysis and full genetic screen Dosage analysis (MLPA) for partial/complete $F_9$ gene deletions/duplications</td>
<td>4ml EDTA peripheral blood</td>
<td>2-8 weeks depending on complexity</td>
</tr>
</tbody>
</table>
VWD
- Detection of known VWF gene mutations by direct sequencing
- Targeted VWF gene screening for Type 2A, 2B, 2N and 2M by direct sequencing
- Full genetic screening for Type 1 and Type 3 VWD
- Dosage analysis (MLPA) for partial/complete VWF gene deletions/duplications

Other Disorders
- F5, F7, F10, F11, F13A by direct sequencing.
- Fibrinogenaemias (α, β and γ genes) Antithrombin deficiency (SERPINC1 gene)
- Carrier and full gene analysis Dosage analysis (MLPA) for partial/complete F7 and SERPINC1

Platelet Disorders
- gene deletions/duplications
  - May-Hegglin anomaly (MYH9 gene).
  - Glanzmann Thrombasthenia (ITGA2B & ITGB3 genes)
  - Bernard-Soulier syndrome (GpIba, Gp9 and GpIbb genes)
  - Platelet-type pseudo VWD (GpIba gene)

Thrombophilia
- Factor V Leiden and prothrombin 20210 mutations by multiplex PCR

<table>
<thead>
<tr>
<th>Disorder</th>
<th>Tests</th>
<th>Specimen Required</th>
<th>Turnaround Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>α-thalassaemia</td>
<td>Detection of deletions using Gap-PCR and MLPA. Detection of non-deletion α⁺ mutations by α-globin gene sanger sequencing.</td>
<td>2 x 4ml EDTA peripheral blood</td>
<td>2-6 weeks depending on complexity</td>
</tr>
</tbody>
</table>

*Prenatal diagnosis of haemophilia by DNA analysis available by prior arrangement with the laboratory. Turnaround time 3-5 working days.

2) **Haemoglobinopathies**

This service is provided by the National Haemoglobinopathy Reference Laboratory which provides a tertiary referral service for all hospitals throughout the UK, Ireland and abroad. Genetic tests for all known haemoglobinopathy mutations are available.
<table>
<thead>
<tr>
<th>Condition</th>
<th>Methodology</th>
<th>Sample Volume</th>
<th>Turnaround Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>β-thalassaemia</td>
<td>Detection of non-deletion mutations using sanger sequencing, pyrosequencing and ARMS-PCR. Detection of deletions using MLPA and Gap-PCR.</td>
<td>2 x 4ml EDTA peripheral blood</td>
<td>2-6 weeks depending on complexity</td>
</tr>
<tr>
<td>HPFH and δβ-thalassaemia</td>
<td>Detection of deletions by MLPA and Gap-PCR. Detection of non-deletion HPFH using sanger sequencing of the gamma gene promoters.</td>
<td>2 x 4ml EDTA peripheral blood</td>
<td>2-6 weeks depending on complexity</td>
</tr>
<tr>
<td>Sickle cell disease</td>
<td>Genotyping by sanger sequencing, pyrosequencing and ARMS-PCR.</td>
<td>2 x 4ml EDTA peripheral blood</td>
<td>2-6 weeks depending on complexity</td>
</tr>
<tr>
<td>Hb Variants</td>
<td>Identification by sanger sequencing of the α, β, γ and δ-globin genes.</td>
<td>2 x 4ml EDTA peripheral blood</td>
<td>2-6 weeks depending on complexity</td>
</tr>
</tbody>
</table>

*Prenatal Diagnosis of sickle cell disease, β-thalassaemia and Hb H/Bart's hydrops fetalis available by prior arrangement with the laboratory. Turnaround time 3-5 working days.*
### Indications: family history of iron overload, unexplained high ferritin.

<table>
<thead>
<tr>
<th>Indications</th>
<th>Test</th>
<th>Specimen Required</th>
<th>Turnaround Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>On all patients with suspected Haemochromatosis.</td>
<td>HFE gene mutation analysis: C282Y/H63D</td>
<td>4ml EDTA peripheral blood</td>
<td>2 weeks</td>
</tr>
<tr>
<td>Hyperferritinaemia with normal serum iron and Transferrin saturation.</td>
<td>SLC40A1 gene (Ferroportin) IRE 5'UTR of FTL gene. FTL gene. Carrier and full genetic screening by sequencing.</td>
<td>4ml EDTA peripheral blood</td>
<td>2-8 weeks depending on complexity</td>
</tr>
<tr>
<td>Hereditary Hyperferritinaemia cataract syndrome</td>
<td>HFE2, HAMP, SLC40A1, TFR2 and HFE gene mutation analysis. Dosage analysis (MLPA) for partial/complete HFE, HFE2, HAMP, TFR2 and SLC40A1 gene deletions/duplications</td>
<td>4ml EDTA peripheral blood</td>
<td>2-8 weeks depending on complexity</td>
</tr>
<tr>
<td>Patients with iron overload, lethargy, liver disease, cardiomyopathy, diabetes, endocrine problems, arthritis, abdominal pain, skin pigmentation.</td>
<td>Dosage analysis (MLPA) for partial/complete HFE, HFE2, HAMP, TFR2 and SLC40A1 gene deletions/duplications</td>
<td>4ml EDTA peripheral blood</td>
<td>2-8 weeks depending on complexity</td>
</tr>
<tr>
<td>Juvenile haemochromatosis: Severe iron overload, diabetes, cardiomyopathy, endocrine problems, hypogonadotrophic hypogonadism</td>
<td>Dosage analysis (MLPA) for partial/complete HFE, HFE2, HAMP, TFR2 and SLC40A1 gene deletions/duplications</td>
<td>4ml EDTA peripheral blood</td>
<td>2-8 weeks depending on complexity</td>
</tr>
<tr>
<td>Asian patients with suspected HC Severe iron overload, diabetes, infertility, endocrine problems, hypogonadotrophic hypogonadism</td>
<td>Dosage analysis (MLPA) for partial/complete HFE, HFE2, HAMP, TFR2 and SLC40A1 gene deletions/duplications</td>
<td>4ml EDTA peripheral blood</td>
<td>2-8 weeks depending on complexity</td>
</tr>
<tr>
<td>Iron overload (negative for the 2 common North European mutations) Non Caucasians with unexplained iron overload</td>
<td>NGS TSCA Iron Regulatory Gene Panel is based on the following gene sets:- TFR2, SLC40A1, HFE, HFE2, HAMP, TF, FTL, IRE of FTL, SLC11A2, TMPRSS6, HEPH, FTH1, CP, ALAS2, BMP4, BMP6, SMAD4.</td>
<td>4ml EDTA peripheral blood</td>
<td>8 weeks</td>
</tr>
</tbody>
</table>
Iron regulatory iron deficiency anaemia (IRIDA), unexplained anaemia
Hyperferritinaemia
Hereditary Hyperferritinaemia
Cataract Syndrome (HHCS)
Atransferrinaemia
Aceruloplasminaemia
Hereditary ferritinopathy
X-linked Sideroblastic anaemia

<table>
<thead>
<tr>
<th>Disorder</th>
<th>Test</th>
<th>Sample Type</th>
<th>Specimen Required</th>
<th>Turnaround Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acute lymphoblastic leukaemia</td>
<td>BCR-ABL t(9;22) by multiplex PCR and quantitative PCR</td>
<td>RNA*</td>
<td>20ml of EDTA peripheral blood or 2ml of bone marrow</td>
<td>3-5 working days</td>
</tr>
<tr>
<td>Acute Myeloid Leukaemia</td>
<td>NPM1, FLT3 ITD and D835TK</td>
<td>DNA</td>
<td>4ml of EDTA peripheral blood or bone marrow</td>
<td>2 weeks</td>
</tr>
</tbody>
</table>

4) Haemato-Oncology

There is a detailed Operational Policy which can be downloaded from the website at:

a) Molecular Genetics / minimum residual disease monitoring

Molecular genetic testing uses PCR (DNA) and RT-PCR (RNA) methodologies to detect common chromosomal abnormalities of clinical, diagnostic or prognostic significance in malignant haematological conditions.
<table>
<thead>
<tr>
<th>Disease</th>
<th>Assay Details</th>
<th>Required Material</th>
<th>Time Frame</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acute Myeloid Leukaemia</td>
<td>PML-RARA t(15;17), CBFB-MYH11 type A, RUNX1/RUNXT1</td>
<td>RNA* 20 ml of EDTA peripheral blood or 2 ml of bone marrow</td>
<td>3-5 working days</td>
</tr>
<tr>
<td>Lymphoma: B-cell clonality</td>
<td>IgH FR1, FR2, FR3 rearrangements</td>
<td>DNA Minimum *5 FFPE Rolled Sections</td>
<td>2 weeks</td>
</tr>
<tr>
<td></td>
<td>Igk Vk-Jk, Vk-Kde + intron-Kde rearrangements</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>IgL rearrangements using BIOMED 2 CE marked primers</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>IgH incomplete D-J rearrangements using BIOMED primers</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lymphoma: T-cell clonality</td>
<td>TCRB and TCRG and TCRD gene rearrangments using BIOMED 2 CE marked primers</td>
<td>DNA Minimum *5 FFPE Rolled Sections</td>
<td>2 weeks</td>
</tr>
<tr>
<td>Chronic lymphocytic leukaemia</td>
<td>Somatic hypermutation analysis using leader and biomed 2 primers</td>
<td>DNA 4 ml of EDTA peripheral blood or bone marrow</td>
<td>4 weeks</td>
</tr>
<tr>
<td></td>
<td>TP53 mutation analysis by FISH and sanger sequencing</td>
<td>Blood or BM slide, DNA</td>
<td></td>
</tr>
<tr>
<td>Chronic myeloid leukaemia</td>
<td>BCR-ABL t(9;22) by multiplex PCR and quantitative PCR</td>
<td>RNA* 20 ml of EDTA peripheral blood or 2 ml of bone marrow</td>
<td>2 weeks</td>
</tr>
<tr>
<td>Myeloproliferative disorders</td>
<td>JAK2 V617F mutation by allele specific PCR and pyrosequencing</td>
<td>DNA 4 ml of EDTA peripheral blood or bone marrow</td>
<td>2 weeks</td>
</tr>
<tr>
<td></td>
<td>JAK2 Exon 12 and MPL W515 mutation analysis</td>
<td>DNA 4 ml of EDTA peripheral blood or bone marrow</td>
<td>4 weeks</td>
</tr>
<tr>
<td>Disorder</td>
<td>Tests</td>
<td>Specimen Required</td>
<td>Turnaround Time</td>
</tr>
<tr>
<td>----------------------------------</td>
<td>-----------------------------------------------------------------------</td>
<td>------------------------------------------------------------------------------------</td>
<td>----------------------------------</td>
</tr>
<tr>
<td>Haemopoietic Stem Cell Transplant</td>
<td>BCR-ABL by multiplex PCR</td>
<td>RNA* 20ml of EDTA peripheral blood or 2ml of bone marrow</td>
<td>2 weeks</td>
</tr>
<tr>
<td></td>
<td>STR-Chimerism (total white cell, CD3, and CD34 positive cells).</td>
<td>DNA BM/PB</td>
<td>2 weeks</td>
</tr>
<tr>
<td>Non Hodgkin’s lymphomas (NHL)</td>
<td>FISH studies for chromosomal rearrangements (8;14); (14;18); (11;14); (11;18); (2;5); del17p13.1, 11q22.3 Performed only after histopathology review</td>
<td>FFPE Slides BM/PB 4ml of EDTA peripheral blood or bone marrow</td>
<td>2 weeks</td>
</tr>
</tbody>
</table>

* Must arrive in lab within 36 hours of collection.

b) **Immunophenotyping**

Immunophenotyping is performed on a six channel Becton Flow Cytometer. The following antibody panels are available:

<table>
<thead>
<tr>
<th>Disorder</th>
<th>Tests</th>
<th>Specimen Required</th>
<th>Turnaround Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acute leukaemia</td>
<td>T cell antibodies (CD2, CD3, CD7)</td>
<td>Blood (10ml EDTA) and/or Bone marrow (1-2 ml EDTA), CSF and Pleural Fluid (5-10 ml), as appropriate. Please note: blood/bone marrow smears should be unstained.</td>
<td>Usually processed within 24 hours*</td>
</tr>
<tr>
<td></td>
<td>B cell antibodies (CD10, CD19, CD79a, Cytoplasmic IgM</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Myeloid antibodies (CD13, CD14, CD33, CD64, CD117, MPO, CD11)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Others (CD34, CD56, HLA-DR, TdT, CD41, NGZ, Glycophorin A)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>B-lymphoproliferative</td>
<td>CD3, CD5, CD10, CD19, CD20, CD23, CD38, CD79b, kappa, lambda, FMC7</td>
<td>Blood (10ml EDTA) and/or Bone marrow (1-2 ml EDTA), CSF and Pleural Fluid (5-10 ml), as appropriate. Please note: blood/bone marrow smears should be unstained.</td>
<td>Usually processed within 24 hours*</td>
</tr>
</tbody>
</table>
### T/NK lymphoproliferative
- CD2, CD3, CD4, CD5, CD7, CD8, CD16, CD19, CD56, kappa, lambda, CD57, TCR, CD25, α, β, γ, δ
- Blood (10ml EDTA) and/or Bone marrow (1-2 ml EDTA), CSF and Pleural Fluid (5-10 ml), as appropriate. Please note: blood/bone marrow smears should be unstained.
- Usually processed within 24 hours*

### Hairy cell leukaemia
- B-lymphoproliferative panel, CD11c, CD22, CD25, CD103
- Blood (10ml EDTA) and/or Bone marrow (1-2 ml EDTA), CSF and Pleural Fluid (5-10 ml), as appropriate. Please note: blood/bone marrow smears should be unstained.
- Usually processed within 24 hours*

### Multiple myeloma
- CD19, CD38, CD45, CD56, CD138, kappa, lambda
- Blood (10ml EDTA) and/or Bone marrow (1-2 ml EDTA), CSF and Pleural Fluid (5-10 ml), as appropriate. Please note: blood/bone marrow smears should be unstained.
- Usually processed within 24 hours*

### PNH
- FLAER, CD14, CD24, CD59 (Assay provided has a lower sensitivity for clones of 1%. As such it is not appropriate for high sensitivity requests).
- Blood (10ml EDTA) and/or Bone marrow (1-2 ml EDTA), CSF and Pleural Fluid (5-10 ml), as appropriate. Please note: blood/bone marrow smears should be unstained.
- Usually processed within 24 hours*

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**SAMPLES ARE VALID FOR 72 HOURS FROM COLLECTION.**

*Results are communicated by e-mail and telephone. All results are discussed at the MDT meetings and authorised weekly.

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### 5) Solid Tumours

Identification of clinically actionable mutations utilising the COBAS system or tumour profiling using a clinically validated NGS based Cancer Panel.

<table>
<thead>
<tr>
<th>Disorder</th>
<th>Tests</th>
<th>Specimen Required*</th>
<th>Turnaround Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Colorectal Cancer</td>
<td>KRAS, BRAF, NRAS 50 gene NGS cancer panel</td>
<td>10 x 5um sections mounted on unstained slides</td>
<td>5 working days</td>
</tr>
<tr>
<td>Case Type</td>
<td>Gene Panel</td>
<td>Section Details</td>
<td>Duration</td>
</tr>
<tr>
<td>-----------------</td>
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</tr>
<tr>
<td>Lung Cancer</td>
<td>EGFR, KRAS, EML4-ALK 50 gene NGS cancer panel</td>
<td>10 x 5um sections mounted on unstained slides. For EML4-ALK, 3x 5um unstained sections on coated slides,</td>
<td>5 working days</td>
</tr>
<tr>
<td>Melanoma</td>
<td>BRAF, NRAS 50 gene NGS cancer panel</td>
<td>10 x 5um sections mounted on Unstained slides. Please note: melanin contamination may cause this test to fail.</td>
<td>5 working days</td>
</tr>
<tr>
<td>Other Cancer</td>
<td>50 gene NGS cancer panel*</td>
<td>10 x 5um sections mounted on unstained slides</td>
<td>10 working days</td>
</tr>
</tbody>
</table>

*Specimen requirements - 10 x 5um sections mounted on unstained slides (or 5 if marked neoplastic area >2cm²). Multiple sections can be placed on a single side. Please clean microtome blade and water-bath thoroughly before cutting sections to avoid cross-contamination and false positive results. Please include a H&E stained section from same block with tumour boundary marked. Tissue in this ring should be >70% neoplastic. Cytological material can be sent as for tissue blocks or send maximum available material (smears, touch preps etc.) on slides.

*Genes included in the 50 gene panel:- BRAF FGFR1 CTNNB1 SMO JAK3 EZH2 KRAS ERBB2 CDKN2A SMAD4 AKT1 GNA11 NRAS MET ABL VHL KDR GNAQ PDGFRA FGFR3 NOTCH1 NPM1 ALK IDH2 PIK3CA FLT3 ATM MPL JAK2 SRC KIT RB1 ERBB4 GNAS MLH1 APC PTEN CSF1R FGFR2 HNF1A HRAS CDH1 EGFR RET STK11 FBXW7 TP53 SMARCB1 PTPN11 IDH1

Consultant Haematologist, Head of BRC/NHS Translational Molecular Diagnostics:

Dr. Anna Schuh. *MD, PhD, MRCP, FRCPath*

Consultant Haematologist: Dr. Chris Hatton

Consultant Clinical Scientist/Scientific Lead: Dr. Shirley Henderson. *PhD.*

Address: Molecular Haematology, Level 4, John Radcliffe Hospital, Headington, Oxford, OX3 9DU
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Sec: 01865 572826
Immunophenotyping: 01865 572827 Fax: 01865 572775

Email: molhaem@ouh.nhs.uk.
Appendix V, NHRL Information for Users

Summary

The National Haemoglobinopathy Reference Laboratory was centrally funded by the DH from 1982 to 31st March 2006, after which the central funding ceased and the DH required the NHRL to charge for its service on a provider to provider basis. The central funding was devolved to all PCTs to pay for haemoglobinopathy DNA studies. The NHRL offers a service for the identification of haemoglobinopathy genotypes by the molecular analysis of DNA and haematological investigation. This includes the investigation of difficult/complex phenotypes and the identification of carrier states for antenatal patients. It also offers a prenatal diagnosis service by fetal DNA analysis for sickle cell disease, α-thalassaemia and β-thalassaemia.

**DNA tests:** There are 5 categories of molecular haemoglobinopathy investigations performed by the NHRL, described in detail in section 1 of this document. The five tests are:

1. α-thalassaemia mutation/s identification (all α^+ and α^0 types),
2. β-thalassaemia, mutation/s identification (all β, δβ-thalassaemia and HPFH deletions)
3. sickle cell disorders (all genotypes)
4. Hb variant identification (all α-chain and β-chain variants)
5. Prenatal diagnosis for α^0-thalassaemias, β-thalassaemias and sickle cell disorders

**Costs:** Tariffs are structured according to the complexity of the tests performed to identify and report the haemoglobinopathy mutations involved. The charging structure is based on the number of amplifications required according to the UKGTN /CMGS guidelines. Thus the costs of the tests for each of the five types of investigation are the same for each level of complexity.

**Indications for sample referral:** Guidelines for which samples should be sent for DNA analysis are listed in section 2. A more detailed document “Guideline for DNA referral” is available and can be obtained in electronic versions by email if required.

**Sample referral details:** Guidelines for the referral of blood samples to the NHRL for either phenotype/genotype investigations or prenatal diagnosis are presented below. A specific NHRL referral form for accompanying blood samples is available, by email if required, or can be downloaded from [www.oxford-translational-molecular-diagnostics.org.uk/content/forms](http://www.oxford-translational-molecular-diagnostics.org.uk/content/forms).

**Quality assurance:** The NHRL is part of the OUH haematology Dept and has full UKAS accreditation to ISO 15189: 2012. Turnaround target times are those of the DH Genetics White Paper and are monitored annually for UKGTN. Last year’s turnaround times for each test category are presented in section below.

**Opening Times.** The NHRL is staffed from 9.00 to 17.00 hours, Mon–Fri, except for bank holidays.

1) The Five Molecular Diagnostic Investigations

There are 5 investigations that can be requested on the referral forms. The molecular analyses included in each investigation are detailed below:
**α-thalassaemia:**

α-thalassaemia mutations in the α-globin genes are screened for as appropriate, using Gap-PCR, MLPA, RE-PCR and DNA sequencing. The α-thalassaemia investigation includes molecular analysis for:

- **α⁺-thalassaemia deletions:** -α³.⁷, -α⁴.²
- **α⁺-thalassaemia:** all non-deletion mutations
- **α⁰-thalassaemia:** --MED, --(α)²².⁶, --SEA, --THAI, --FIL
- **α⁰-thalassaemia:** --BRIT, --SA, and all rare/novel deletion mutations

**β-thalassaemia:**

All known β-thalassaemia mutations, and all δβ-thalassaemia / HPFH deletion mutations are screened in a referral for β-thalassaemia mutation analysis, using ARMS-PCR, RE-PCR, Gap-PCR, DNA sequencing and MLPA as appropriate. The β-thalassaemia investigation includes:

- **β-thalassaemia:** all point mutations in all ethnic groups.
- **δβ-thalassaemia, Aγδβ-thalassaemia and εγβ-thalassaemia**
- HPFH deletion genes: Black HPFH1, Ghanaian HPFH2, Indian HPFH3, Italian HPFH4/5, Vietnam HPFH6
- Non deletional HPFH - γ-promoter mutations
- Fusion Hbs Lepore and Kenya
- Triple α-gene analysis in thalassaemia intermedia cases and patients with unusually severe β-thalassaemia trait.
- Xmn1 and α-thalassaemia status of thalassaemia intermedia/major patients.
- Increased α-globin gene copy number by MLPA or Gap-PCR

**Sickle cell disease:**

Sickle cell disease genotypes are analysed by ARMS-PCR, RE-PCR, Gap-PCR, MLPA, DNA sequencing and pyrosequencing as appropriate. The sickle cell investigation includes:

- Hb S/S, Hb S/Hb D-Punjab, Hb S/Hb O-Arab, Hb S/C,
- Hb S/β-thalassaemia, Hb S/δβ-thalassaemia, Hb S/HPFH.
- Xmn1 status of affected patients with high Hb F levels
- α-thalassaemia status of affected patients if appropriate

**Hb Variant:**

All haemoglobin variants are identified by DNA analysis. Referrals for diagnosis of an unknown variant are analysed first by HPLC and IEF, and then identified/confirmed by DNA analysis using ARMS-PCR, RE-PCR, Gap-PCR, MLPA, DNA sequencing, as appropriate. The Hb variant investigation includes:

- **Confirmation of the clinically important Hb variants in antenatal patients:**
  The variants Hb D-Punjab, O-Arab and Lepore require confirmation by DNA analysis in all antenatal patients. The clinically important abnormal haemoglobins Hb S, C, E, are identified by DNA analysis only in couples requiring prenatal diagnosis.

- **Identification of unknown / rare Hb variants in antenatal patients:** These will be identified by DNA sequencing as a matter of urgency if partner is a haemoglobinopathy carrier.
• **Identification of unknown rare Hb variants in non-antenatal patients:** These are characterised first by HPLC and IEF, and then identified/confirmed by DNA sequencing of the $\alpha_1$, $\alpha_2$ and $\beta$-globin genes.

**Prenatal Diagnosis:**

The prenatal diagnosis investigation involves the molecular determination of a fetal genotype and identification/confirmation of the maternal and paternal genotypes. To provide the safest possible result and in accordance with best practise guidelines, **two** different molecular diagnostic techniques are used to arrive at the result. The PND investigation also includes a check for maternal DNA contamination of the fetal DNA sample by a study of the inheritance pattern of 11 polymorphic STR markers from the mother and father.

Prenatal diagnosis is performed by mutation analysis of parental DNA (prepared from fresh blood samples whenever possible) and fetal DNA (prepared from chorionic villi, cultured chorionic villi, amniotic fluid, cultured amniocytes or fetal blood).

The genetic risks for prenatal diagnosis are detailed in our DNA referral guidelines document and also found in the “Handbook for Laboratories” published on-line by the NHS Sickle Cell and Thalassaemia Screening Programme as detailed below.

In summary, we carry out prenatal diagnosis for couples at risk of having a child affected with:

- Hb Bart’s Hydrops fetalis (homozygous $\alpha^0$-thalassaemia)
- Hb H hydrops fetalis involving severe non-deletion $\alpha^*$-thalassaemia mutations
- homozygous $\beta$-thalassaemia,
- $\beta$-thalassaemia co-inherited with $\delta\beta$-thalassaemia, Hb Lepore, Hb E, Hb O-Arab

2) **Samples that require DNA investigation**

**Antenatal samples**

Comprehensive guidelines for which patient samples require further investigation for antenatal screening purposes are contained in the “Handbook for Laboratories” published by the NHS Sickle Cell & Thalassaemia Screening Programme Laboratory Subgroup. The guidelines can be downloaded from the Screening Programme website (www.sct.screening.nhs.uk/) or is available from us electronically by email request.

**Non-antenatal samples**

The following haemoglobinopathies should be referred for genotyping by DNA studies:

- patients with Hb H disease,
- $\beta$-thalassaemia major & intermedia
- sickle $\beta$-thalassaemia, sickle HPFH
- Hb E / $\beta$-thalassaemia
- Hb E / $\alpha$-thalassaemia
- Any other complex haemoglobinopathy

A definitive diagnosis of the following carrier states can only be made by DNA analysis. **If in doubt about a referral please telephone the lab for advice.**
• $\alpha^0$-thalassaemia & homozygous $\alpha^+$-thalassaemia (in high risk groups)
• Hb D-Punjab
• Hb E
• unknown Hb variants,
• $\delta\beta$-thalassaemia & HPFH trait,
• $\beta$-thalassaemia trait
• $\beta$-thalassaemia trait with borderline-raised Hb A$_2$ value (silent $\alpha$-thalassaemia),
• patients with a split Hb A$_2$ value which add up to >3.5%
• Any other complex haemoglobinopathy

3) Sample referral procedures

Non-Pre-Natal samples: carrier state/genotyping request:

A fresh 10ml EDTA blood samples should be sent with:

a. a completed genotype referral request form with patient information clearly supplied,

b. haematological details of the patient (full blood count, Hb A$_2$ and F values, iron status, Hb electrophoresis results)

There are separate referral forms for genotype analysis and prenatal diagnosis. The referral form must be filled in and returned with the samples, together with the family origin form.

For optimal DNA analysis results, blood samples should be less than 5 days old. However, samples up to one-month old that have been kept refrigerated usually give satisfactory results.

Samples should always be sent in appropriate packaging by first class post, or for urgent samples, by courier service.

Turnaround times: these depend on the urgency of the sample and how many molecular investigations are required to identify the mutations. The target turnaround time for genotyping urgent antenatal patients is two weeks. For non-antenatal patients, the target turnaround time is three to eight weeks, but a few very complex analyses may take longer.

Prenatal diagnosis request:

The lab must be telephoned in advance to make arrangements for the referral of a prenatal diagnosis case, including provision of safe contact details to report the result by telephone and fax.

Requirements: Fresh parental blood samples must be sent with the fetal sample, together with a completed prenatal diagnosis request form with the following parent information clearly supplied: haematological details of both parents (full blood count, Hb A$_2$ and F values, Hb electrophoresis results.

Parental samples: Fresh 10ml EDTA blood samples from mother and father should be sent from all couples requiring prenatal diagnosis at the time of fetal sampling. This is required for control samples and for testing for maternal contamination, even if the mutation has been characterised previously. If the father is unavailable for blood sampling, a copy of his laboratory results stating his haemoglobin genotype should be provided. If the paternal genotype is unknown please contact the laboratory for advice.
**Fetal sample:** CVs, amniotic fluid or fetal blood can be used to extract fetal DNA.

- **Chorionic villus biopsy sample (CVS):** The CVS must be cleaned by microscopic dissection to remove any contaminating maternal tissue before sending to the NHRL for DNA analysis. The referrer must arrange for this to be carried out at a local cytogenetics laboratory, and also instruct the lab to forward the sorted CVS by guaranteed post or courier to the NHRL with appropriate documentation.

  It is recommended that a CVS culture is set up by the cytogenetics lab for back up purposes if this is not to be done routinely by the lab for karyotyping. The NHRL will contact the cytogenetic laboratory if the backup cultures are required.

  The cleaned CVS should be sent to the NHRL in culture medium, saline or if possible, in CVS lysing solution (0.1-0.5ml depending upon size of the cleaned CVS sample). CVS lysing solution is 100mM NaCl / 25mM EDTA / 0.2% SDS / 0.4mg/ml Proteinase K.

- **Amniotic fluid sample:** Obstetric departments should aim to take approximately 20 mls of amniotic fluid. 10mls can then be forwarded directly to the NHRL for testing. The remaining 10mls can be sent to a local cytogenetics laboratory for back-up cultures. The NHRL will contact the cytogenetic laboratory if the backup cultures are required. If it is not possible to obtain 20mls of amniotic fluid please telephone the laboratory for advice.

- **Fetal blood:** On very rare occasions fetal blood sampling may be performed and a fetal blood sample sent in EDTA for analysis. For example, for the diagnosis of homozygous α-thalassaemia in a fetus diagnosed as hydropic by ultrasound.

**Turnaround times:**
The time taken for prenatal diagnosis is normally 3-5 working days upon receipt of fetal sample, provided the parental mutations are known beforehand.

**Patient consent:**
Specific patient consent obtained for medical investigations for a haemoglobinopathy in one laboratory will permit the referral of the blood sample to another laboratory for: additional investigations of the haemoglobinopathy, the storage of the patient's DNA sample for any further investigations related to the patient's diagnosis in the future, the use of the patient's DNA for quality assurance in laboratory tests, and the use of the patient's DNA for education and training of laboratory staff. It will not permit the analysis of the patient's DNA for any other genetic disorder without further specific consent for that test.

Each blood sample referred for DNA analysis should be accompanied by a referral form which has been signed by the requesting clinician/counsellor/nurse to state that patient consent for haemoglobinopathy DNA testing has been obtained and that a copy of the consent could be obtained if required. Alternatively, or a copy of the patient's consent form allowing testing for haemoglobinopathies should be provided. We are able to provide a blank patient consent form if required.
Reference ranges

Our reference ranges for haemoglobinopathy screening red cell indices are:

<table>
<thead>
<tr>
<th></th>
<th>Hb g/l</th>
<th>RBC $10^9$/l</th>
<th>MCV fl</th>
<th>MCH pg</th>
<th>Hb A₂ %</th>
<th>Hb F %</th>
</tr>
</thead>
<tbody>
<tr>
<td>men</td>
<td>130-170</td>
<td>4.5-5.5</td>
<td>80-98</td>
<td>27-32</td>
<td>2.0-3.2</td>
<td>&lt;1.0</td>
</tr>
<tr>
<td>women</td>
<td>120-150</td>
<td>3.8-4.8</td>
<td>27-32</td>
<td>2.0-3.2</td>
<td>&lt;1.0</td>
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Quality Assurance

UKAS accreditation: The NHRL is part of the OUH NHS Trust haematology dept quality management system which has been awarded UKAS accreditation to ISO 15189: 2012 status.

The NHRL participates in the NEQAS “DNA diagnostics for haemoglobinopathies” pilot scheme, the NEQAS “Hb A₂/Hb F & abnormal haemoglobins” scheme, newborn sickle screening scheme and the NEQAS full blood count scheme.