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Haematology and Blood Bank Laboratory User Manual			

Belfast Health and Social Care Trust (BHSCT)

HAEMATOLOGY AND BLOOD BANK LABORATORY USER MANUAL

Additional Information & Cross References	
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Introduction to Laboratory Services

The Department of Haematology resides within the Belfast Health and Social Care Trust (BHSCT) and aims to provide a comprehensive routine and specialist regional diagnostic, interpretative and clinical advice service. The repertoire of investigations and required specimens are outlined in this document.

The department provides a general haematology and blood transfusion service across 3 sites, namely; The Royal Group of Hospitals (RGH) or commonly known as the Royal Victoria Hospital (RVH), Belfast City Hospital (BCH) and the Mater Infirmorum Hospital (MIH) with specialist and regional services [including the Stem Cell Bank] mainly located at the Belfast City Hospital laboratory.

The laboratory service provides an out of hours repertoire which is restricted to routine testing for example; FBC, coagulation screening, INR, APTT for monitoring of unfractionated heparin, D-dimers in specific circumstances, ESR for query temporal arteritis, blood grouping and the provision of blood and blood products. Other specialised investigations which may be required, for example; over long weekends and public holidays can be arranged as appropriate by prior discussion with the duty Consultant Haematologist.

The Haematology and Blood Transfusion laboratories are currently accredited by the United Kingdom Accreditation Service (UKAS) under the standards defined for Medical laboratories – requirements for quality and competence (ISO 15189:2022). The scope of the Haematology laboratory accreditation with UKAS is as outlined in the following link [customer reference: 8703] or by searching for accredited organisations on the main UKAS website.

www.ukas.com/search-accredited-organisations/

It is important for users to note that this document will be updated regularly and therefore any hard copies made of this information may not be valid beyond the time of printing. Similarly, locally-saved versions of this publication may only be valid at the time it was saved. For the most current version, refer to publications on the BHSCT websites (Intranet and Internet), Belfast Trust GP website and primary care intranet.

- ***Provision of Services on Multiple Sites***

The Belfast Trust Laboratories (BTL) for Haematology, Blood Bank and Stem Cell Bank currently operate under a single management system across 3 multiple sites, namely; the Royal Victoria Hospital, Belfast City Hospital and Mater Infirmorum Hospital and as outlined in 'GEN-1 General Principles for the Assessment of Conformity Assessment Bodies by UKAS'.

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All staff are managed by a central management team and are trained and competent to work on any site as and when required. The Belfast Trust Laboratories are part of a central Quality Management System (QMS) which includes control of documents, incidents, improvement audits, training/competency assessments, traceability, change management and verifications/validations.

The Laboratories within Haematology and Blood Bank operate a limited service from the BCH and MIH laboratories. Samples are transferred to the RVH site to accommodate a 24 hour cover. BHSCT Transport Service operate a frequent, 20 minute collection service, during 'out of hours' period to facilitate transfers between sites. In addition, there are motor cycle couriers available during peak periods, to ensure that urgent samples are delivered in a timely manner.

Identical analytical platforms are employed on all BHSCT sites and each site employs the same internal quality assurance materials and each is enrolled in the same external quality assessment schemes. The established reference ranges apply to all sites.

Under certain clinical circumstances, when an APTT value cannot be obtained using the routine coagulation analyser (ACL TOP 550), this test will automatically be performed on an alternative platform (Stago). Please refer to the Table further down for normal ranges.

Transferred tests are analysed and reported according to the same BHSCT standard operating procedures (SOPs).

Inter site comparison analyses for all transferred tests are regularly performed to ensure consistency of results.

Users should be advised that all telephone communications will be automatically diverted to the alternate sites as appropriate.

Please see table below:

TEST	MIH	BCH
FBC	Transferred to RVH 24 hours	09:00 – 16:00 *
INR	Transferred to RVH 24 hours	09:00 – 16:00 *
Coagulation Screen	Transferred to RVH 24 hours	09:00 – 16:00 *
D-Dimer	Transferred to RVH 24 hours	Transferred to RVH 24 hours

*** All samples received after 16:00 will be transferred to RVH for analysis.**

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Laboratory Contact Details

General Laboratory Enquiries	RVH	BCH	MIH
Blood Bank	028 961 50038 or 50039	028 950 40987 or 40988	028 950 41332 or RVH Blood Bank
Routine Haematology	028 961 51540 or 51541	028 950 40922 or 40921	028 950 41508 or Lab reception on 41328
Coagulation	028 961 51540 or 51541	028 950 40921	028 950 41508 or Lab reception on 41328
Paediatric Haematology (RBHSC)	028 961 50388		
Haemostasis & Thrombosis (BCH)	028 950 40910		
Flow Cytometry (BCH)	028 950 40913		
Stem Cell Bank (BCH)	028 950 40912		
Red Cell Investigations (BCH)	028 950 40915		
Laboratory Out of Hours Emergency	028 961 50038 Contact Royal Hospital, Blood Bank		
On-call Registrar &/or Consultant	028 9032 9241 BCH switchboard		
Secretaries office	BCH site: 028 950 47989	028 908 02223	

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Professional Contacts

Clinical Director		Dr Gary Benson	028 950 47977 gary.benson@belfasttrust.hscni.net
Service Improvement Lead (SIL) Haematology & Blood Bank		Dr Richard Gooding	028 950 45634 Richard.Gooding@belfasttrust.hscni.net
Blood Sciences Service Manager (Haematology, Blood Bank & Immunology)		Mr Stephen Kane	028 950 46792 stephenr.kane@belfasttrust.hscni.net
BTL Governance & Quality Manager		Mrs Gillian Foster	028 961 51562 gillian.foster@belfasttrust.hscni.net
Regional Haemovigilance Manager		Denise McKeown	07795801936 or e-mail Denise.McKeown@belfasttrust.hscni.net
Laboratory Managers	Blood Bank	Mrs Lyndsey Parker	028 961 51543 lyndsey.parker@belfasttrust.hscni.net
	Haematology & Stem Cell Bank <i>(Routine and Specialist)</i>	Mrs Frances McCauley	Office: 028 950 42154 Mobile: 07545212199 frances.mccauley@belfasttrust.hscni.net
Operational Managers	Blood Bank	Vacant	Vacant
	Routine Haematology	Ms Joy Gallagher	028 961 51498 Joy.Gallagher@belfasttrust.hscni.net
	Specialist Haematology	Ms Colleen Williamson	028 950 40915 Colleen.Williamson@belfasttrust.hscni.net
	Stem Cell Bank <i>(Designated Individual for SCB)</i>	Mrs Nicola McCormack	028 950 40912 Nicola.McCormack@belfasttrust.hscni.net
BHSCT Haemovigilance Service		028 950 40602 or e-mail: haemovigilance@belfasttrust.hscni.net	

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Clinical Contacts

Haematology Laboratory Registrar (By Rotation)	Mon-Fri 9 – 5pm (excludes bank hols)	1st Tel. 028 950 40911 No answer- Bleep 1660 or Tel. Office 028 950 5005 or 50054
	Outside normal hours (includes bank hols)	Phone BCH switchboard on 028 9032 9241
General Haematology (Adult)	Dr Jeremy Hamilton	028 961 54221
Lymphoma/Myeloma Service	Dr David Donaldson	028 961 56289
	Dr Sarah Lawless	077 405 39589
	Dr Michael Quinn	028 950 47831
	Dr Oonagh Sheehy	028 950 48069
	Dr David Waddell	028 950 50227
Leukaemia/BMT Service	Dr Claire Arnold	028 950 48036
	Dr Stephen Boyd	028 950 48016
	Dr Nicholas Cunningham	028 950 48016
	Dr Damian Finnegan	028 950 47940
	Prof MF McMullin	028 950 48008
Haemostasis & Thrombosis Service	Dr Gary Benson	028 950 47977
	Dr Claire Corrigan	028 950 45634 or 028 950 40444
	Dr Richard Gooding	028 950 45634 or 028 950 40444
Paediatric Haematology Service	Dr Christine Macartney	028 961 56958
	Dr Bethany Mitchell	028 950 48118

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Laboratory Locations and Opening Hours

Within the Haematology, Blood Bank and Stem Cell Bank Laboratories

Belfast City Hospital

(BCH)

Switchboard Telephone: 028 90 329241

Service: Routine and Specialist Haematology, Blood Bank & Stem Cell Bank

BCH Tower Block,
C Floor,
Lisburn Road,
Belfast,
BT9 7AB.

Opening Hours:

Monday to Friday

Haematology	09:00 - 16:00
Blood Bank	08:00 - 18:00
Stem Cell Bank (SCB)	09:00 - 17:00

Laboratory Contact Details (BCH)

Outside the above hours a reduced service is provided by the Haematology and Blood Bank Laboratories, Kelvin Laboratories, Royal Victoria Hospital - see Laboratory Contacts.

Blood Bank	028 950 40987 or 40988
Routine Haematology & Coagulation	028 950 40922 or 40921
Haemostasis & Thrombosis	028 950 40910
Flow Cytometry	028 950 40913
Stem Cell Bank	028 950 40912
Red Cell Investigations	028 950 40915

Royal Victoria Hospital

(RVH)

Switchboard Telephone: 028 90 240503

Service: Routine Haematology & Blood Bank

Kelvin Laboratories,
274 Grosvenor Road,
Belfast,
BT12 6AB.

Opening Hours:

Monday to Friday Open 24 hrs

(A reduced service is provided between 17:00-09:00)

Sat, Sun & Bank Holidays Open 24 hrs
(Reduced service is provided)

Laboratory Contact Details (RVH)

-see Laboratory Contacts.

In the event of an Emergency

Tel. 028 961 50038

(RVH, Blood Bank)

Blood Bank	028 961 50038 or 50039
Routine Haematology & Coagulation	028 961 51540 or 51541

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Mater Infirmorum
Hospital
(MIH)

Switchboard Telephone: 028 90 741211

Service: Routine Haematology & Blood Bank

Crumlin Road,
Belfast,
BT14 6AB

Laboratory Contact Details (MIH)

Service is provided by the Haematology and Blood Bank Laboratories, Kelvin Laboratories, Royal Victoria Hospital – see Laboratory Contacts.

Blood Bank	028 950 41332 or RVH Blood Bank
Routine Haematology & Coagulation	028 950 41508 or Lab reception on 41328

Royal Belfast Hospital for
Sick Children
(RBHSC)

Switchboard Telephone: 028 90 240503

Service: Paediatric Haematology
(satellite lab only)

274 Grosvenor Road,
Belfast,
BT12 6AB.

Opening Hours:

Monday to Friday 09:00 – 17:00

Laboratory Contact Details:

Outside the above hours a reduced service is provided by the Haematology and Blood Bank Laboratories, Kelvin Laboratories, Royal Victoria Hospital – see Laboratory Contacts.

Paediatric Haematology (RBHSC)	028 961 50388
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Outside hours Clinical staff should contact the RVH Laboratories on:

RVH Blood Bank: 028 961 50038 or 028 961 50039
RVH Haematology: 028 961 51540 or 028 961 51541

Weekdays after 5pm, weekends and Bank holidays this service must only be used for urgent requests which may influence immediate patient management.

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Clinical Advice and Interpretation

Where appropriate, interpretative information will be reported with numerical results.

For a more detailed interpretation or clinical advice on clinical indications and limitations of examination procedures the Belfast Trust Clinical Haematology Team have developed a new app to make routine referrals quick and easy, track their progress and receive advice without having to wait for a call back. This app is available to all users within the Belfast Trust only, on their phone, tablet or computer via the 'Loop' web page.

For emergencies and outside of Trust referrals **bleep #1660** or contact the duty laboratory Specialist Registrar (SpR) via switchboard at the Belfast City Hospital (BCH) or the appropriate consultant.

There is a 24-hour consultant-led medical on-call service available for interpretative advice and clinical consultation in urgent cases, including acceptance of patients who are found to have primary haematological disorders.

The Duty Specialist Registrar and/or consultant can be contacted via the BCH switchboard or via Ward 10 North, BCH.

Haematology Referral App:

On the BHSCT Internal Loop enter a search for '***Haematology app***' or '***Haematology referral***'.

Alternatively, click on 'Digital Apps' in the top menu, scroll down and select 'Clinical' from within the Trust Application Category. Scroll down and the Haematology Referrals App will be listed at the bottom right.

When running the app for the first time you may get a pop up box below, this is a standard requirement and nothing out of the ordinary, please 'allow' access and you will be able to access the app.

Any issues with using the new app please email: haemreferrals@belfasttrust.hscni.net

Please do not submit referrals through this email address as any referrals submitted in this way will **not** be actioned. All routine referrals should be submitted through the app only.

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How to make Comments, Informal Complaints and Compliments

The Haematology laboratory is committed to continuously improving the quality and range of services provided and welcomes any comments, suggestions or compliments from the service users. A User Satisfaction Survey is conducted every two years by the Belfast Trust Laboratories which forms part of the Management Review (MR).

Current users of the laboratory are invited to participate in this survey and responses are discussed and feedback given to users. The MR report will be made available to users of the service upon written request to the laboratory manager.

The Haematology laboratory is committed to fully investigating failures in service delivery to reduce the risk of recurrence, improve the service and ensure compliance with the Belfast Trust clinical governance policies.

Copies of the policy are available via:

Online via BHSCT website: <http://www.belfasttrust.hscni.net/>

Upon request from: LabFeedback@belfasttrust.hscni.net .

Please contact the Laboratory Service Manager (details as follows) if you have any concerns, informal complaints or compliments. All complaints will be recorded, reviewed, acted upon and feedback offered.

Making a complaint does not affect your rights and will not prevent you from accessing laboratory services.

For further information on raising a formal complaint against the service, please refer to the BTL User Manual C-345 for further contact details on the Complaints Department.

Blood Bank	Mrs Lyndsey Parker	Work Tel: 028 961 51543 E-mail: lyndsey.parker@belfasttrust.hscni.net
Haematology & Stem Cell Bank (Routine & Specialist)	Mrs Frances McCauley	Work Tel: 028 950 42154 Mobile: 07545212199 E-mail: frances.mccauley@belfasttrust.hscni.net

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Service Agreement and Patient Consent

Each request accepted by the Haematology laboratory service for examination(s) (will take into account the request, the examination and the report) shall be deemed as an agreement by the user for the Belfast Health & Social Care Laboratory services, or other accredited laboratories, as they may be used to perform testing outside the repertoire, to carry out the necessary testing and reporting function.

It also implies an acceptance of the conditions of preparation and transport as outlined in this manual. In some cases, where more specialised or invasive procedures are required or those with an increased risk of complication a more detailed explanation or recorded consent may be required and in emergency situations the laboratory may carry out necessary procedures in the best interests of the patient.

The Haematology department will endeavour to inform customers and users of the service as soon as possible if there are:

- Any deviations from the agreement that may impact upon the examination results
- Any issues that arise which may impact on the quality of the service or the results provided.

Data Protection

The Haematology department adheres to the Belfast Health & Social Care Trust Policy on the Data Protection Act 2018, Good Data Protection Regulations (GDPR) and Protection of Personal Information (Reference TP 026/08). These outline the legal requirement for both the Trust and its staff to treat personal information confidentially and ensure all information is held securely.

Further details, including how we use patient or service user information, can be found on the BHSC Trust website: <https://bhsct.sharepoint.com/sites/pm/SitePages/Welcome-to-Information-Governance-and-Data-Protection.aspx>

Haemovigilance Service

The Trust Haemovigilance staff ensure safe transfusion practice is an integral part of transfusion practice outside of the Laboratory.

This includes:

- Ensuring transfusion policies in the Clinical areas are up to date with current guidelines and best practice
- Provide education on Transfusion Practice to multi-disciplines and staff groups
- Be a resource to clinical area and support services and a link to the Blood Bank Laboratory
- Investigate incidents and report to appropriate bodies
- Monitor transfusion practice and component use & wastage
- Continuously strive to improve transfusion practice outside of the Laboratory

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More information can be found on the BHSCT Loop page, entering search option 'Haemovigilance Department'.

Contact details for the Haemovigilance Team:

E-mail: haemovigilance@befasttrust.hscni.net Telephone: 028 950 40602

Instructions on Completing a Haematology Request Form and Specimen Tube

➤ Haematology

Attention to detail is essential to ensure that the right result is sent out on the right patient. Specimens will not be accepted for analysis where the following essential criterion in the Minimum Data Set is not met on the form or the specimen container.

SAMPLE	REQUEST FORM
<ul style="list-style-type: none"> • H&C number ¹ • Patient Official First Name • Surname • Date of Birth (dd/mm/yyyy) • Name / signature of staff member taking the sample ⁵ 	<ul style="list-style-type: none"> • H&C number ¹ • Patient Official First Name • Surname • Sex • Date of Birth (dd/mm/yyyy) • Date & Time of Sample Collection • Full Name of Consultant / Authorised Healthcare Professional • HCP Code of Consultant / Authorised Healthcare Professional² • Source Name / encompass Location code³ • Test Requested • Specimen type and Anatomical Site ⁴ (where relevant) • Name / signature of staff member taking the sample ⁵
Footnote	
<ol style="list-style-type: none"> 1. The H&C Number must be used unless the patient is not registered with a GP in NI / is registered but does not yet have their H&C number (in which case, it must clearly state "No H&C number available" on the request form) or In an emergency situation when the identity of the patient is UNKNOWN (in which case, use the local hospital emergency numbering system) 2. HCP code must be registered by Information Standards Group. To request a HCP code please visit https://regional.sharepoint.hscni.net/sites/CCIS/SitePages/NIHCPCodeRequest.aspx 3. encompass location code should be provided on request form to ensure correct return of results. However it is noted not all areas area are in the position to provide an encompass location code at present and therefore provision of encompass code will not be enforced under MAC at this time. 4. Mandatory for Microbiology and Virology only 5. Mandatory for Blood Transfusion only 	

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The table above lists the new updated Minimum Acceptance Criteria (MAC 2.0) for sample and request forms. All points MUST be present and legible on the sample container and request form in order to progress to laboratory testing. It is responsibility of the requester to ensure that samples are correctly labelled and request forms are completed to agreed standards.

Note to encompass users: If the sample collection process is not properly followed on encompass, the laboratory will not receive an electronic request. Consequently, the MAC will not be met and the sample will not be processed.

Additional notes:

1. If the location/source is not specified, lab staff cannot telephone critical results.
2. Some tests are time-specific and if the date and time of sampling are not stated, the accuracy of such results cannot be assured.
3. For example, medication of anticoagulants such as Apixaban and Rivaroxaban (with time of last dose in relation to sampling) can be very relevant to interpretation of result.
4. If gender is not specified, the laboratory cannot provide gender-specific reference ranges.
5. Telephoning of critical results is further facilitated by identification of the requesting practitioner.

Precious samples:

In exceptional circumstances where samples are deemed precious e.g. samples which are unrepeatable or highly invasive e.g. CSF, bone marrow specimens etc. it should be documented on the sample request that it is a 'precious sample'. In the rare event that this type of sample arrives with missing information, it will be the decision of senior lab staff whether to analyse the sample and report any results. The report will contain an appropriate comment relating to the problem and the likely reliability of the results, alerting the requesting practitioner to take responsibility for the results and for any action taken as a result of the report.

OrderComms:

The essential criteria will all be fulfilled if the sample and request form information are sent in an electronically-created paper format (Ordercomms) and we strongly encourage the trust-wide use of OrderComms, with the exception of Blood Transfusion.

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Instructions on Completing a Blood Bank Request Form and Sample Labelling

➤ Blood Transfusion Request Form & Sample Labelling

Patient details must be confirmed against the patient's ID band, and by direct questioning of the patient according to Trust policy (*see [hyperlink below](#)*). Information on patient's ID band and sample must be identical.

BHSCT: Blood Transfusion Manual – Policy, Procedures and Guidelines is available on the intranet or link below:

[Blood Transfusion Manual, Policy, Procedures and Guidelines](#)

Better Blood Transfusion E-Learning- there is an on line teaching programme available. Go to:

Trust staff: LearnHSCNI - <https://learn.hscni.net>

Agency staff: ELFH (elearning for health) [Blood Transfusion - elearning for healthcare \(elfh.org.uk\)](#)

Requests for blood grouping and/or ordering blood products must be completed by a doctor or designated nurse. The collection and labelling of specimens for blood transfusion may be delegated to designated staff who have had the appropriate training.

The laboratory will undertake testing and issue of blood/blood products only on receipt of a legible Regional Request form and a correctly labelled specimen or, where applicable, using EPIC. Incorrectly completed forms or specimens will be rejected according to BSH Guidelines for Pre-Transfusion compatibility procedures in Blood Transfusion Laboratory, 2012.

An addressograph or EPIC label can be used on the **Request Form** provided the consultant and ward area are completed. Fill out all details on the **Request Form** as follows:

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Essential Information for a Blood Bank Request Form	
Patient Details	<ul style="list-style-type: none"> • Official First Name • Surname • DOB • H&C number (MRN if no H&C available) • Sex • Patient location • Full Name of Consultant / Authorised Healthcare Professional • HCP Code of Consultant / Authorised Healthcare Professional <p><i>Patient's address is desirable only</i></p>
Requestors signature	<p>Doctor or authorised staff member must sign the request.</p> <p>This section on the request form labelled "<i>I confirm that the patient identification details correspond with the details on the patients identification band and the sample tube. Within the last 3 years I am certified as competent in core competency in obtaining a venous sample for pre-transfusion testing</i>"</p> <p>Must be signed by the person taking the blood sample.</p>
Date and Time of sample	Required to assess suitability
Test Required	<p>Tick the box indicating what test is required:</p> <p>Group and Antibody screen - This is a blood group and antibody screen and sample is saved in case a crossmatch is required later, kept for 7 days.</p> <p>Direct Antiglobulin Test</p> <p>Kleihauer</p> <p>Transfusion Reaction Investigation</p>
Product or Component Requirement	Record the number of units required and product type in the table
Date and Time Required Delivery area	Record date and the time of start of operation or required transfusion time. Record where the blood needs to be delivered to.
Special Requirements	Tick the appropriate box indicating if the patient needs CMV neg, Irradiated, or HLA matched components
Clinical Details Fill in information as available	<p><i>The following is Desirable criteria:</i></p> <ul style="list-style-type: none"> • Blood Group • Previous transfusions • Known antibodies • Indication for transfusion • Known transfusion reactions • Any recent anti-D administration

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Essential Information for Labelling a Blood Bank Sample Tube

Once the sample has been taken the tube **must be labelled immediately, at the patient's bedside, with details transcribed from the patient ID band onto the sample tube OR the appropriate EPIC label affixed by the person who took the sample.**

An EPIC label is the **ONLY** type of printed label that will be accepted on a Blood Bank sample tube

N.B. All samples must be handwritten and signed, except when using EPIC label. Additionally, NIBTS referral sample tube must be signed by sample taker and have date and time taken on it.

Pre-labelling of samples is NOT allowed and is considered highly dangerous to the patient!

The following information **must** be on the sample tube, unless EPIC label* is used

Essential Information for a Blood Bank Sample Tube	
Patient Details	<ul style="list-style-type: none"> • Official First Name • Surname • DOB • H&C number (MRN only if H&C not available) • Signature of sample taker (not required for EPIC label, unless NIBTS referral*) • Date of specimen

Samples not meeting these minimum requirements will not be accepted. There is a 'zero tolerance' policy for incorrectly or incompletely labelled requests.

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Management of Unidentified Patients

A special protocol is applicable for unidentified patients in A&E and theatres. Check the Intranet for the most recent Trust policy titled: '**Management of Unidentified Patients**'.

Confirmation Sample in Blood Bank

When a patient has **no historic blood group** on record in Blood Bank and blood components are required; the lab will request a further blood sample (confirming blood group sample). Blood components can still be issued for the patient without this confirming sample at the request of the clinician caring for the patient e.g. if blood is required urgently and there is no time for a further sample to be processed.

Blood Tracking and Cold Chain

Blood/blood product tracking and maintenance of the "cold chain" are mandatory national requirements of the Blood Safety and Quality Regulations. All hospitals have a legal requirement to trace each individual unit of blood components/products, whether transfused or disposed of in accordance with the EU Directive 2002/98/EC. Blood must be stored only in designated blood refrigerators and not in any other ward refrigerators. Blood will be returned to stock 48 hours after the date/time requested as stated on the request form unless the blood bank is notified that it is still required.

Urgent Samples

The laboratory must be telephoned to arrange all urgent samples before the specimen is sent to the laboratory. It is not sufficient to mark the request form as urgent. The requesting clinician is responsible for arranging transport of urgent samples to the laboratory. Samples should not be left in the clinical areas over weekends and bank holiday periods.

Non-accidental Injury (NAI):

NAI screen - please see [Appendix 22](#)

Rejection of Requests for analysis or Restrictions on Reporting of Results

➤ **Blood Transfusion**

There is a '**Zero Tolerance**' policy in Blood Bank for incorrectly or incompletely labelled requests (this includes the sample and request form). This is in line with the British Blood Transfusion guidelines and patient safety.

➤ **Haematology**

The laboratory may be obliged to reject a request for analysis or restrict reporting of results when:

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- There is insufficient information supplied to enable unequivocal identification of the patient, the specimen, the tests required or the source of the request.
- The specimen has been collected inappropriately e.g. unsuitable anticoagulant or preservative.
- The integrity of the specimen is in question e.g. leaked in transit, undue delay in transport, a pattern of results that suggests significant error (eg. sample from a drip arm or contaminated with EDTA).
- There is interference present which invalidates one or more test results (excessive haemolysis, icterus or turbidity).

An appropriate comment will be added to the report, assuming a valid report can be entered into the patient record based on information supplied. Please contact the laboratory for a full explanation of the reason for rejection or restriction of reporting and advice on how to avoid this eventuality.

Precious Samples:

In exceptional circumstances where samples are deemed precious e.g. samples which are unrepeatable or highly invasive e.g. CSF, bone marrow specimens etc. it should be documented on the sample request that it is a 'precious sample'. In the rare event that this type of sample arrives with missing information, it will be the decision of senior lab staff whether to analyse the sample and report any results. The report will contain an appropriate comment relating to the problem and the likely reliability of the results, alerting the requesting practitioner to take responsibility for the results and for any action taken as a result of the report.

Requesting Additional Add-on Tests

➤ **Haematology**

Telephone the lab within 4 hours to request additional investigations on a specimen, for example; reticulocyte count, blood film, red cell investigations, coagulation tests etc.

For add on requests the requestor must ring the Laboratory with patient details and if possible the most recent laboratory accession number.

The Laboratory will require the name and source of the requestor in order to record a full audit trail on LIMS for the new test request.

➤ **Blood Transfusion**

Additional tests/examinations including blood products can be ordered depending on availability of a suitable valid sample in the Blood Bank.

A new sample will be required according to the following guidelines (this can be found on the reverse of the Blood Transfusion request form).

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Patient transfused or pregnant	Timing of Blood Sample
Within last 3 months	Within 72 hours of anticipated transfusion
More than 3 months ago	Within 7 days of anticipated transfusion

To request additional tests or products contact the blood bank.

Irregular Antibodies:

Please note that if a patient is found to have a red cell alloantibody or a positive Direct Antiglobulin Test (Coomb's test) it will take longer to select compatible blood. Extra samples are likely to be requested from Blood Bank to confirm the identified antibody with NIBTS. If antibody is known, please state antibody on the request form and contact the blood bank to discuss emergency cases or pre-operative cases for whom blood is required in theatre. Patients in the latter category should **not** be sent to theatre until you have confirmed that compatible blood is available.

Paediatric Transfusion:

- (i) Neonates & infants <4 months: An initial sample must be taken from infant & mother. When infant/mother incompatibility exists the blood will be cross-matched against the mother's plasma. The mother's blood group, antibody screen and NIBTS antenatal reference number must be sent to the lab.
- (ii) Older infants & children: Send only the child's sample, unless contacted by the lab.

Minimum Re-testing Intervals

Minimum re-testing intervals (MRI) are defined as the minimum time before a test should be repeated, based on the properties of the test and the clinical situation in which it is used. MRIs facilitate a more efficient use of pathology services and hence improve patient care.

Guideline:

The Royal College of Pathologists (RCPATH) have published a national document on the minimum retesting intervals in pathology.

***This document (attached below) should only be used as a guideline.**

Link to: [National minimum retesting intervals in pathology: A final report detailing consensus recommendations for minimum retesting intervals for use in pathology](#)

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Availability of Results

All laboratory results reported are based on professional judgements made by personnel who have the appropriate qualifications for their job role.

All personnel will have the applicable theoretical and practical background experience to perform the assigned managerial and technical tasks in accordance with national, regional and local regulations and professional guidelines.

Hospital patient results can be accessed on ward computers electronically via EPIC as soon as they become validated.

GP reports are transmitted by electronic download (EDI) to individual practice systems.

The issuing of results of a non-urgent nature over the telephone is discouraged and must be kept to an essential minimum in the interests of safety as verbal reports may lead to transcription errors.

In the event of a major delay to patient results a 'Labs Notification' will be generated and sent to all BTL users by senior management.

Turnaround Times (TAT)

- For an **extremely urgent** sample, please contact the laboratory to have it prioritised.
- Turnaround times are from day of receipt to issue of reports.
- See the list of tests for a more detailed version with expected TATs for each test.
- The times shown are the typical turnaround times achieved by the laboratory, but may be longer or shorter depending on the availability of staff and the complexity of the investigation.
- For more specialised tests these will be batched and results available two working days after completion.

If further tests, such as a manual differential white cell count are required, the results will not be displayed until they are validated.

The accession number will be displayed, and by telephoning the lab, a provisional result can be given.

Referral Tests

Specialised tests which are not available in the Belfast Trust may be sent to selected referral laboratories for analysis by arrangement. The referral centre names are provided with the laboratory reports. Further details are available upon request.

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Sample Stability

A delay in transport to the laboratory may affect sample quality and compromise the results produced. It is essential that the specimens reach the laboratory as soon as possible to allow processing before the sample quality deteriorates.

The sample may be rejected if it is received outside the recommended timeframe.

Measurement of Uncertainty

Measurement uncertainty recognises that all results are subject to measurement error and in order to compare a test result with a previous result or with a specific decision value it can be useful to have a feel for the reliability of the result. However, biological variation should be taken into consideration. The measurement of uncertainty is available to all service users on request. Any requests for information from a service user must be referred to the Laboratory Manager or deputy. The user will be supplied with a copy of the current measurement of uncertainty for the test(s) with an accompanying letter explaining how measurement of uncertainty can be utilised in the interpretation of laboratory results.

External Quality Assurance

All laboratory tests performed are externally quality assured; test performance can be obtained upon written request.

Sample Packaging and Transport

All blood samples must be transported in the specially designed once-only laboratory form/specimen carrier bags and in accordance with the BHSCT Laboratories Transport Policy.

Users should refer to the Trust policy document titled 'Transport of specimens to the Laboratory'.

This policy is available in the Belfast Trust Laboratories User Manual which is accessible on the home page of the intranet or via the internet at: <http://www.belfasttrust.hscni.net> and in the Services Tab select Adult Services, followed by Laboratory Services & moving the cursor to 'Belfast Trust Laboratories User Manual' (purple section) and click on the link.

Health and Safety

All specimens must be treated as a potential hazard.

Specimens of blood, serum and other body fluids from suspected carriers of Category 3 pathogens (hepatitis B or C and HIV) must be clearly marked with hazard stickers and enclosed in a sealed plastic bag. Request forms should also have a hazard sticker.

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For Laboratory Related Information: Guidance on COVID-19 Samples

Please refer to the COVID-19 section [Guide 1] of the 'Contents' page in the Belfast Trust Laboratories (BTL) User Manual (C-345) which is available via the BHSCCT website (see link below) and Trust intranet.

<https://belfasttrust.hscni.net/service/laboratory-services/laboratories-user-manual/>

The following pages provide specific information on the departmental test repertoire including sample requirements and interpretative information.

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BLOOD BANK

*** In an Emergency please contact Blood Bank to prioritise sample (TAT < 2hrs)**

TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TURN-AROUND TIME (TAT)
BLOOD GROUP & ANTIBODY SCREEN	Blood	EDTA	6mls			For more complex investigations sample may be referred to NIBTS Reference Lab [TAT is 24 -72hrs]. Refer to Appendix 1.	< 4hrs
COLD AGGLUTININ TITRE <i>This test is not UKAS accredited</i>	Blood	EDTA	6mls				24 hrs
CELL GROUP & DAT (< 4 MONTHS OLD)	Blood	EDTA	0.5mls				< 4hrs
* CROSS-MATCH / ELECTRONIC ISSUE	Blood	EDTA	6mls				< 4hrs
DIRECT ANTIGLOBULIN TEST (COOMB'S TEST)	Blood	EDTA	4 or 6mls				< 4hrs
KLEIHAUER TEST	Blood	EDTA	6mls			FMH sample sent to referral laboratory for confirmation of bleed >2mls. [TAT is 5 days]. See Referral Labs.	48 hrs

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HAEMATOLOGY * *FBC and ESR normal ranges are based on Dacie and Lewis, Practical Haematology.*

TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT															
BLOOD FILM	Blood	EDTA	4mls		See Appendix 2 & 4	By arrangement with laboratory staff. See Appendix 3 & 4 Sample collection tube must be an EDTA with black insert. Yellow inserts, containing a gel, will be rejected. See Appendix 21	< 4hrs															
ESR <i>(Erythrocyte Sedimentation Rate)</i> * <i>ESR</i> <i>Normal range is based on Dacie and Lewis, Practical Haematology</i>	Blood	EDTA	4mls	Insufficient samples (<1.5ml) will be rejected	<table border="1"> <thead> <tr> <th>Age (years)</th> <th>Adult Male</th> <th>Adult Female</th> </tr> </thead> <tbody> <tr> <td>17-50</td> <td>≤ 10mm</td> <td>≤ 12mm</td> </tr> <tr> <td>51-60</td> <td>≤ 12mm</td> <td>≤ 19mm</td> </tr> <tr> <td>61-70</td> <td>≤ 14mm</td> <td>≤ 20mm</td> </tr> <tr> <td>>70</td> <td>≤ 30mm</td> <td>≤ 35mm</td> </tr> </tbody> </table>	Age (years)	Adult Male	Adult Female	17-50	≤ 10mm	≤ 12mm	51-60	≤ 12mm	≤ 19mm	61-70	≤ 14mm	≤ 20mm	>70	≤ 30mm	≤ 35mm	Sample collection tube must be an EDTA with black insert. Yellow inserts, containing a gel, will be rejected. See Appendix 21	< 4hrs
Age (years)	Adult Male	Adult Female																				
17-50	≤ 10mm	≤ 12mm																				
51-60	≤ 12mm	≤ 19mm																				
61-70	≤ 14mm	≤ 20mm																				
>70	≤ 30mm	≤ 35mm																				
FULL BLOOD COUNT (FBC) (Causasians) Including DWCC <i>Normal range is based on Dacie and Lewis, Practical Haematology</i>	Blood	EDTA	4mls	FBC sample must be analysed <16 hours from the bleed time	See Appendix 2 Includes individual parameters.	Sample collection tube must be an EDTA with black insert. Yellow inserts, containing a gel, will be rejected. See Appendix 21	Urgent < 1hr Routine < 4hrs															

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
MALARIAL PARASITES	Blood	EDTA	4mls	<p>Sample must be received in RVH Haematology lab < 12hrs old.</p> <p>See Appendix 6</p>	See Appendix 6	<p>Sample collection tube must be an EDTA with black insert. Yellow inserts, containing a gel, will be rejected. See Appendix 21</p> <p><i>Plasmodium Knowlesi</i> species can't be identified by this laboratory.</p> <p>Therefore suspected cases are referred to the Malarial Reference Laboratory, London for PCR identification. Samples referred for confirmation by PCR TAT = 1-2 wks.</p>	< 4hrs
RETICULOCYTE COUNT <i>Normal range is based on Dacie and Lewis, Practical Haematology</i>	Blood	EDTA	4mls	<p>Sample must be <24 hours old from the bleed time.</p>	<p>0 day - Adult 0.2 -2.5% 50 – 100 x10⁹/L</p>	<p>Sample collection tube must be an EDTA with black insert. Yellow inserts, containing a gel, will be rejected. See Appendix 21</p>	< 4hrs

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SPECIALIST RED CELL

TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
ERYTHROPOIETIN	Blood (serum or plasma)	Serum or EDTA	6mls		2.5 – 10.5mIU/mL		< 3 weeks
G6PD SCREEN	Blood	EDTA	4mls		Reported as; Deficient or Not Deficient	By arrangement with laboratory staff	7 days
SICKLE CELL, HAEMOGLOBIN VARIANT and THALASSAEMIA SCREENING	Blood	EDTA	4mls		See Appendix 5	Laboratory testing is only provided Mon - Fri 9-4pm Patients <28 days old - please contact red cell laboratory before sending samples. Ethnicity must be clearly stated on all request forms and a FBC must be completed.	< 3 weeks
SICKLE CELL TEST Urgent	Blood	EDTA	4mls		Reported as; Positive or Negative See Appendix 7	Contact lab if required urgently Ethnicity must be clearly stated on all request forms and a FBC must be completed.	< 4hrs

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	EPIC CODE	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
SICKLE CELL TEST	Blood	EDTA	4mls	Sick	Reported as Positive or Negative See Appendix 7	All samples will be forwarded to red cell laboratory for confirmation screening by capillary electrophoresis. Ethnicity must be clearly stated on all request forms and a FBC must be completed.	< 3 weeks
HbSF LEVELS	Blood	EDTA	4mls	HbSF	Reported in %	Contact Red Cell lab if required urgently	<7 days

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ROUTINE COAGULATION

* **Coagulation reference ranges** have been regionally established based on the **ACL TOP 550 Coagulation analyser (see Appendix 8)**.

Samples will **not** be processed if over filled, under filled and if the sample tube is expired.

** Samples will **not** be processed if the sample is received >4hrs after venepuncture

For paediatric sample requirements please contact the laboratory

Prothrombin Time, APTT and Fibrinogen routine tests may be processed up to 12 hours post venepuncture on the **ACL TOP 550 Coagulation analyser**.

D Dimer & INR may be processed up to 24 hours, assuming the samples have been stored at room temperature under controlled conditions i.e. 15-24 °C. These times were based on a laboratory study undertaken under controlled conditions.

TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
** ANTI-THROMBIN	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	0.83 - 1.28 IU/mL	Only performed at BCH Mon-Fri 9am-5pm (excluding bank holidays) Also available as part of Thrombophilia Screen.	< 6 weeks as part of thrombophilia screen. Discuss with lab for urgent samples
* APTT	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	See Appendix 8 for age related ref. range	Also available as part of Coagulation Screen. Underfilled, haemolysed, lipaemic sample will be rejected.	Urgent < 2hrs Routine < 4hrs

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
* COAGULATION SCREEN	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	See Appendix 8 for age related individual analytes - PT, APTT, Fib.	Underfilled, haemolysed, lipaemic sample will be rejected.	Urgent < 2hrs Routine < 4hrs
* D-DIMERS	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	< 0.5µg/mL	Underfilled, haemolysed, lipaemic sample will be rejected.	< 4hrs
* FIBRINOGEN	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle.	See Appendix 8 for age related ref. range	Also available as part of Coagulation Screen. Underfilled, haemolysed, lipaemic sample will be rejected.	< 4hrs
* INR	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle.	Therapeutic target range dependent on clinical indication	Underfilled, haemolysed, lipaemic sample will be rejected.	< 4hrs
** PROTEIN C Activity	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle	0.74-1.53 IU/mL	Also available as part of Thrombophilia Screen	< 6 weeks

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
** Free PROTEIN S	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle	Male: 0.66-1.56 IU/mL Female in absence of oral contraception, HRT or pregnancy: 0.60 – 1.13 IU/mL	Also available as part of Thrombophilia Screen	< 6 weeks
* PROTHROMBIN TIME	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle.	See Appendix 8 for age related ref. range	Also available as part of Coagulation Screen. Underfilled, haemolysed, lipaemic sample will be rejected. Presence of anticoagulant	< 4hrs
** THROMBOPHILIA SCREEN	Blood	Citrate	3.0mls x 4	Fill sample tubes to line indicated on bottle	See individual analytes	BCH only Mon-Fri 9am-5pm excluding bank holidays **Not performed on patients >40yrs** See guidelines for Thrombophilia Screening	< 6 weeks

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SPECIALIST COAGULATION

➤ **Coagulation reference ranges** have been locally established based on the **Stago Coagulation analyser**.

Samples will **not** be processed if over filled, under filled and if the sample tube is expired.

** Samples will **not** be processed if the sample is received >4hrs after venepuncture

For paediatric sample requirements please contact the laboratory

D-Dimer & INR may be processed up to 24 hours, assuming the samples have been stored at room temperature under controlled conditions i.e. 15-24 °C. These times were based on a laboratory study undertaken under controlled conditions.

TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
** ADAMTS13 <i>This test is not UKAS accredited</i>	Blood	Citrate x2	3.0mls	Fill sample tubes to line indicated on bottle Please complete <u>ADAMTS13 request form</u> See Appendix 9	72 – 1113%	Test only available in BCH Mon-Fri 9am-5pm, (excluding bank holidays). For same day testing, sample must arrive in lab by 12pm. Clinical details must be discussed with Haematology prior to sending sample. See Appendix 9	Routine < 3 wks Emergency <6hrs
**ANTI- XA (Low Molecular Weight Heparin [LMWH])	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	Prophylaxis 0.2 – 0.4 IU/mL Treatment 0.5 – 1.0 IU/mL		< 4hrs

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
** Direct oral anticoagulants (DOACs) (APIXABAN/ RIVAROXABAN/ EDOXABAN) <i>This test is not UKAS accredited</i>	Blood	Citrate	3.0mls	Fill sample tubes to line indicated on bottle	See guidance in Appendix 10	Dose taken, time taken, sample date & time MUST be specified on request form.	Urgent < 4hrs Routine 1 week
** UNFRACTIONATED HEPARIN ASSAY (UFH)	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	Treatment 0.5 – 1.0 IU/mL	Only performed at BCH Mon-Fri 9am-5pm (excluding bank holidays)	Discuss with BCH lab
** APC RESISTANCE RATIO (APC-R)	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	> 2.0	Also available as part of Thrombophilia Screen.	< 6 weeks
➤ APTT	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle Sample must be analysed within 4 hours of the bleed time.	Stago 24 – 38 seconds	Also available as part of Coagulation Screen. Underfilled, haemolysed, lipaemic sample will be rejected.	< 4hrs

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
** COAGULATION FACTOR ASSAYS	Blood	Citrate	3.0 mls x2	Fill sample tubes to line indicated on bottle	See individual analytes	BCH only Mon-Fri 9am-5pm (exceptions are Factors 8, 9, and chromogenic 8. See below	Routine < 3 wks Urgent < 4hrs
➤ COAGULATION SCREEN	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	See individual analytes for PT, APTT and Fibrinogen	Underfilled, haemolysed, lipaemic sample will be rejected.	Routine < 4hrs Urgent < 1.5hrs
➤ D-DIMERS	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	Stago < 0.5 µg/mL	Underfilled, haemolysed, lipaemic sample will be rejected.	< 4hrs
** FACTOR INHIBITOR ASSAYS	Blood	Citrate	3.0 mls x2	Fill sample tubes to line indicated on bottle		BCH only - Mon-Fri 9am-5pm (excluding bank holidays) By arrangement with laboratory staff – see individual factors	Urgent requests - same day if received before 12pm. Routine < 3 wks
** FACTOR II	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	0.6 – 1.3 IU/mL	BCH only Mon-Fri 9am-5pm excluding bank holidays	Routine < 3 wks Urgent < 4hrs
** FACTOR V	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	0.6 – 1.3 IU/mL	BCH only Mon-Fri 9am-5pm excluding bank holidays	Routine < 3 wks Urgent < 4hrs

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
** FACTOR VII	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	0.6 – 1.3 IU/mL	BCH only Mon-Fri 9am-5pm excluding Bank holidays	Routine < 3 wks Urgent < 4hrs
** FACTOR VIII	Blood	Citrate	3.0 mls x2	Fill sample tubes to line indicated on bottle	0.6 – 1.3 IU/mL	Performed BCH 9am-5pm Mon-Fri OOH @ RVH site (urgent samples only)	Routine < 3 wks Urgent < 4hrs
** CHROMOGENIC FACTOR VIII	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle	0.6-1.5 IU/mL	Performed BCH 9am-5pm Mon-Fri OOH @ RVH site (urgent samples only)	Routine < 3 wks Urgent < 4hrs
** FACTOR IX	Blood	Citrate	3.0mls x2	Fill sample tubes to line indicated on bottle	0.6 – 1.3 IU/mL	Performed BCH 9am-5pm Mon-Fri OOH @ RVH site (urgent samples only)	Routine < 3 wks Urgent < 4hrs
** FACTOR X	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	0.6 – 1.3 IU/mL	BCH only Mon-Fri 9am-5pm excluding bank holidays	Routine < 3 wks Urgent < 4hrs
** FACTOR XI	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	0.6 – 1.3 IU/mL	BCH only Mon-Fri, must be received before 4pm Performed on fresh plasma.	< 4hrs
** FACTOR XII	Blood	Citrate	3.0 mls	Fill sample tube to line indicated on bottle	0.6 – 1.3 IU/mL	BCH only Mon-Fri, must be received before 4pm Performed on fresh plasma.	< 4hrs

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** FACTOR XIII	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle	0.7-1.4 IU/mL	BCH only	Routine < 6 wks <i>(contact lab if result is urgent)</i>
➤ FIBRINOGEN	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle. Sample must be analysed within 4 hours of the bleed time.	Stago Analyser 2-5 g/L	Also available as part of Coagulation Screen. Underfilled, haemolysed, lipaemic sample will be rejected.	< 4hrs
HITT ASSAY (Heparin Induced Thrombocytopenic Thrombosis)	Blood	Serum Clotted sample (Serum)	4 or 6mls	Clotted sample required (Red or yellow top)	Optical Density values of: ≥ 1.0 U/mL = Positive < 1.0 U/mL = Negative	Clinical details must be discussed with Haematology prior to sending sample.	< 72hrs
INR	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle	Therapeutic target range dependent on clinical indication	Underfilled, haemolysed, lipaemic sample will be rejected.	< 24hrs
** LUPUS ANTICOAGULANT	Blood	Citrate	3.0mls x2	Fill sample tube to line indicated on bottle	Negative	Also available as part of Thrombophilia Screen	< 3 wks <i>4-6 weeks as part of Thrombophilia</i>

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
PLATELET FUNCTION ASSAY <i>This test is not UKAS accredited</i>	Blood	Citrate	3.0mls x2	Fill sample tube to line indicated on the bottle. Must be received within 2 hours of venepuncture. Do not send in Pod system.	Col/ADP 62-110 seconds Col/Epi 82-150 seconds	Pre-arrange with laboratory [Tel: 028 950 40910] processed in BCH only Mon – Fri 9am-4pm excluding bank holidays. Will not be processed on Cat 3/high risk samples. Not suitable for infants under 1 year old. Unreliable results with low Hb/HCT and platelets <150. Samples that have had a difficult venepuncture should not be sent.	< 2 hours
PLATELET AGGREGATION <i>This test is not UKAS accredited</i>	Blood	Citrate	3mls x 5 Must include a 'Control' sample 3mls x 5. Contact lab*	Patients should ideally be bled at the haemophilia centre, BCH. Fill sample tube to line indicated on the bottle. Must be received within 2 hours of venepuncture. Do not send in Pod system. Samples that have had a difficult venepuncture should not be sent.		Pre-arrange with laboratory [Tel: 028 950 40910] processed in BCH only, Mon-Fri 9am – 3pm excluding bank holidays. Must be discussed with consultant haematologist AND the haemostasis laboratory before taking samples.	< 3 hours

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	REFERENCE RANGE	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
➤ PROTHROMBIN TIME	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle. Sample must be analysed within 4 hours of the bleed time.	Stago Analyser 12-17 seconds	Also available as part of Coagulation Screen. Underfilled, haemolysed, lipaemic sample will be rejected. Presence of anticoagulant	< 4hrs
** REPTILASE	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle	15 – 18 seconds	BCH only Mon-Fri 9am-5pm excluding bank holidays	Routine < 3 wks Urgent < 4hrs
** THROMBIN CLOTTING TIME	Blood	Citrate	3.0mls	Fill sample tube to line indicated on bottle	14 – 22 seconds	BCH only Mon-Fri 9am-5pm excluding bank holidays.	Routine < 3 wks Urgent < 4hrs
** VON WILLEBRANDS SCREEN <i>(Includes PT, APTT, Factor 8, VWF antigen, VWF activity)</i>	Blood	Citrate	3.0mls x 2	Fill sample tube to line indicated on bottle	0.7 – 2.0 IU/mL	BCH only Mon-Fri 9am-5pm excluding bank holidays	< 3 weeks Contact lab for urgent requests

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FLOW CYTOMETRY-haemato-oncology

All tests are available on Encompass under 'Flow Cytometry - Haemato-Oncology'

TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	EPIC INSTRUCTIONS	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
BONE MARROW INVESTIGATION	Bone Marrow	By arrangement with medical staff			<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ FLOW 	By arrangement with medical staff	2 weeks (verbal result available in 2 days)
CD34	Blood	EDTA	4mls		<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ CD34 	By arrangement with medical staff	2 weeks (verbal result available in 2 days)
PERIPHERAL BLOOD Flow cytometry For investigation of haematological malignancies NB Lymphocyte subsets for investigation of immunodeficiency states – refer to Immunology Lab.	Venous Blood	EDTA	4mls		<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ FLOW 	By arrangement with medical staff. See guidelines in Appendix 11	2 weeks (verbal result available in 2 days)

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	EPIC INSTRUCTIONS	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
BONE MARROW Flow cytometry	Bone Marrow	RPMI/ Heparin	3 – 5mls		<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ FLOW 	By arrangement with medical staff. See guidelines in Appendix 12	2 weeks <i>(verbal result available in 2 days)</i>
Body fluid Flow cytometry	Body fluid e.g. Pleural Fluid	EDTA	Up to 4mls		<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ FLOW 	By arrangement with laboratory staff. Store and transport at room temp asap. See guidelines in Appendix 12	2 weeks <i>(verbal result available in 2 days)</i>
Unfixed tissue biopsies Flow cytometry	Unfixed tissue	RPMI	N/A		<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ FLOW 	By arrangement with laboratory staff. Store and transport at room temp asap. See guidelines in Appendix 12	2 weeks <i>(verbal result available in 2 days)</i>
HEREDITARY SPHEROCYTOSIS (EMA BINDING STUDY)	Blood	EDTA	4mls		<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ EMA 	MUST be pre-booked by contacting flow cytometry staff. See guidelines in Appendix 13 & 14	2 weeks <i>(verbal result available in 2 days)</i>
PNH SCREEN	Blood	EDTA	4mls	See guidelines on PNH screening	<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ PNH 	By arrangement with medical staff. See guidelines in Appendix 15 Sample should be tested as soon as possible after collection up to a maximum of 48 hours.	2 weeks <i>(verbal result available in 2 days)</i>

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TEST	SAMPLE	SAMPLE TUBE	VOLUME	PRECAUTIONS	EPIC INSTRUCTIONS	KEY FACTORS AFFECTING PERFORMANCE / INTERPRETATION	TAT
CSF	CSF	Plain container	10-20 drops	Store and transport at room temperature	<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ FLOW 	By arrangement with medical staff. Transport to lab as soon as possible after collection.	2 weeks (<i>verbal result available in 2 days</i>)
URINARY HAEMOSIDERIN	Urine	Yellow top urine or plain universal container	10mls			None	2 weeks
PLATELET GLYCOPROTEIN	Blood	EDTA	4mls (NAI thumb prick)		<ul style="list-style-type: none"> ➤ Flow cytometry - Haemato-oncology ➤ PLATELET GLYCOPROTEIN 	By arrangement with medical staff. Transport to lab as soon as possible after collection.	2 weeks (<i>verbal result available in 2 days</i>)

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BHSCT Haematology Department

Referral Laboratories

Blood Bank

1. Test: Antibody Investigation

Northern Ireland Blood Transfusion Service (NIBTS)
 Belfast City Hospital Complex
 Lisburn Road
 Belfast
 BT9 7TS
 Tel: 028 9032 1414

2. Test: Foetal Maternal Haemorrhage (FMH)

Northern Ireland Blood Transfusion Service (NIBTS)
 Belfast City Hospital Complex
 Lisburn Road
 Belfast
 BT9 7TS
 Tel: 028 9032 1414

Haematology

3. Test: Malarial Parasites for confirmation

Malaria Reference Laboratory
 London School of Hygiene and Tropical Medicine.
 Keppel Street,
 London,
 WC1E 7HT
 Tel: 020 7927 2427
 Fax: 020 76370248

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APPENDICES

The following provide additional information or useful guidance referred from the individual test information or the laboratory reports.

[Appendix 1: Clinical Significance of Red Cell Antibodies](#)

[Appendix 2: FBC Test – Regional Reference Range](#)

[Appendix 3: Reporting of Blood Films by Medical Staff](#)

[Appendix 4: WBC in Africans and Caribbeans of African Lineage](#)

[Appendix 5: Haemoglobinopathy & Thalassaemia Investigation](#)

[Appendix 6: Investigation of Malarial Parasites](#)

[Appendix 7: Sickling disorders - Sickle solubility testing](#)

[Appendix 8: Age Related Coagulation Reference Ranges for the ACL Top 550 Analyser](#)

[Appendix 9: ADAMTS13 Activity Request Form](#)

[Appendix 10: DOAC Levels](#)

[Appendix 11: Strategy for handling peripheral blood samples](#)

[Appendix 12: Proposals for a revised strategy for handling bone marrow and body fluid samples](#)

[Appendix 13: Referral of samples for EMA binding studies](#)

[Appendix 14: Notes and Guidance on the Use of Eosin-5-maleimide \(EMA\) Binding in the Diagnosis of Hereditary Spherocytosis](#)

[Appendix 15: Guidelines on PNH Screening by Flow Cytometry](#)

[Appendix 16: Guidelines for Red Cell Volume Investigation](#)

[Appendix 17: Bone Marrow Aspirate and Trepine Biopsy](#)

[Appendix 18: Reticulocyte haemoglobin equivalent \(RET-He\) in the diagnosis of functional iron deficiency](#)

[Appendix 19: Methylene Blue Fresh Frozen Plasma or Cryoprecipitate](#)

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[Appendix 20: Provision of HLA selected platelet components for alloimmunised patients](#)

[Appendix 21: BHSCT Order of Draw for Blood Sciences](#)

[Appendix 22: Non-Accidental Injury Screen](#)

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Appendix 1

Clinical Significance of Red Cell Antibodies

- Clinically significant antibodies are those that are capable of causing patient morbidity due to the accelerated destruction of a significant proportion of transfused red cells.
- Anti-A, anti-B and anti-A,B must always be regarded as being of clinical significance.
- With few exceptions, red cell antibodies which are likely to be of clinical significance are only those which are reactive in the indirect antiglobulin test (IAT), performed strictly at 37°C.
- Recommendations for the selection of red cells for transfusion to patients with alloantibodies are given in table 1 below.

Table 1 - Likely clinical significance of red cell alloantibodies, and recommendations for the selection of blood for patients with their presence

System	Specificity	Likely clinical significance in transfusion	Recommendation for selection of red cells for transfusion *
ABO	Anti-A ₁	No	IAT crossmatch compatible at 37°C
Rh	Anti-D, -C, -c, -E, -e	Yes	Antigen negative (Rh & K antigen identical)*
Rh	Anti-C ^w	No	IAT crossmatch compatible **
Kell	Anti-K, -k	Yes	Antigen negative*
Kell	Anti-Kp ^a	No	IAT crossmatch compatible **
Kidd	Anti-Jk ^a , -Jk ^b	Yes	Antigen negative*
MNS	Anti-M (active 37°C)	Yes	Antigen negative*
MNS	Anti-M (not active 37°C)	No	IAT crossmatch compatible at 37°C
MNS	Anti-N	No	IAT crossmatch compatible at 37°C
MNS	Anti-S, -s, -U	Yes	Antigen negative*
Duffy	Anti-Fy ^a , -Fy ^b	Yes	Antigen negative*
P	Anti-P ₁	No	IAT crossmatch compatible at 37°C
Lewis	Anti-Le ^a , -Le ^b , -Le ^{a+b}	No	IAT crossmatch compatible at 37°C
Lu	Anti-Lu ^a	No	IAT crossmatch compatible at 37°C
Diego	Anti-Wr ^a (anti-Di3)	Yes	IAT crossmatch compatible **
H	Anti-HI (in A ₁ and A ₁ B patients)	No	IAT crossmatch compatible at 37°C
All	Others active by IAT at 37°C	Yes	Seek advice from Blood Centre

* Where antigen negative red cells are recommended these should also be compatible in an IAT crossmatch.

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** These recommendations apply when the antibody is present as a sole specificity. If present in combination, antigen negative blood may be provided by the blood centre, to prevent wastage of phenotyped units.

The above guidance is also suitable for patients undergoing hypothermia during surgery (Mollison, 2005b).

BSH Guidelines for Pre-Transfusion compatibility procedures in Blood Transfusion Laboratory, 2012.

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Appendix 2

FBC Test - Regional Reference Ranges

Reference ranges based on Dacie & Lewis

Test Analytes & Units	Current Gender	Age	Reference Range
Haemoglobin (Hb) g/L	Female	0-1 day	140 - 220
		2-3days	150 - 210
		4-7days	135 - 215
		0-1 month	115 - 165
		0-2 months	94 - 130
		0-1 year	111 - 141
		2-6 years	110 - 140
		7-12 years	115 - 155
		13-150 years	120 - 150
	Male	0-1 day	140 - 220
		2-3 days	150 - 210
		4-7 days	135 - 215
		0-1 month	115 - 165
		0-2 months	94 - 130
		0-1 year	111 - 141
		2-6 years	110 - 140
		7-12 years	115 - 155
		13-150 years	130 - 170
Haematocrit (HCT) L/L	Female	0-1 day	0.45 - 0.75
		2-3days	0.45 - 0.67
		4-7days	0.42 - 0.66
		0-1 month	0.33 - 0.53
		0-2 months	0.28 - 0.42
		0-1 year	0.3 - 0.38
		2-6 years	0.34 - 0.4
		7-12 years	0.35 - 0.45
		13-150 years	0.36 - 0.46
	Male	0-1 day	0.45 - 0.75
		2-3 days	0.45 - 0.67
		4-7 days	0.42 - 0.66
		0-1 month	0.33 - 0.53
		0-2 months	0.28 - 0.42
		0-1 year	0.3 - 0.38
		2-6 years	0.34 - 0.4
		7-12 years	0.35 - 0.45
		13-150 years	0.4 - 0.5
Red Blood Cell (RBC) x10¹²/L	Female	0-1 day	5 - 7
		2-3days	4 - 6.6
		4-7days	3.9 - 6.3
		0-1 month	3 - 5.4

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		0-2 months	3.1 - 4.3
		0-1 year	3.9 - 5.1
		2-6 years	4 - 5.2
		7-12 years	4 - 5.2
		13-150 years	3.8 - 4.8
	Male	0-1 day	5 - 7
		2-3 days	4 - 6.6
		4-7 days	3.9 - 6.3
		0-1 month	3 - 5.4
		0-2 months	3.1 - 4.3
		0-1 year	3.9 - 5.1
		2-6 years	4 - 5.2
		7-12 years	4 - 5.2
		13-150 years	4.5 - 5.5
Mean Cell Haemoglobin (MCH) pg	Female	0-1 day	31 - 37
		2-3days	31 - 37
		4-7days	31 - 37
		0-1 month	30 - 36
		0-2 months	27 - 33
		0-1 year	25 - 29
		2-6 years	24 - 30
		7-12 years	25 - 33
		13-150 years	27 - 32
	Male	0-1 day	31 - 37
		2-3 days	31 - 37
		4-7 days	31 - 37
		0-1 month	30 - 36
		0-2 months	27 - 33
		0-1 year	25 - 29
		2-6 years	24 - 30
		7-12 years	25 - 33
		13-150 years	27 - 32
Mean Cell Haemoglobin Concentration (MCHC) g/L	Female	0-1 day	300 - 360
		2-3days	290 - 370
		4-7days	280 - 370
		0-1 month	290 - 370
		0-2 months	285 - 355
		0-1 year	320 - 360
		2-6 years	310 - 370
		7-12 years	310 - 370
		13-150 years	315 - 345
	Male	0-1 day	300 - 360
		2-3 days	290 - 370
		4-7 days	280 - 370

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		0-1 month	290 - 370
		0-2 months	285 - 355
		0-1 year	320 - 360
		2-6 years	310 - 370
		7-12 years	310 - 370
		13-150 years	315 - 345
Mean Cell Volume (MCV) fl	Female	0-1 day	100 - 120
		2-3days	92 - 118
		4-7days	88 - 126
		0-1 month	92 - 116
		0-2 months	87 - 103
		0-1 year	72 - 84
		2-6 years	75 - 87
		7-12 years	77 - 95
		13-150 years	83 - 101
	Male	0-1 day	100 - 120
		2-3 days	92 - 118
		4-7 days	88 - 126
		0-1 month	92 - 116
		0-2 months	87 - 103
		0-1 year	72 - 84
		2-6 years	75 - 87
		7-12 years	77 - 95
		13-150 years	83 - 101
Red Cell Distribution Width (RDW) %	Female	0-<1	
		1-<14	
		13-150years	11.6 - 14
	Male	0-<1 yrs	
		1-<14 yrs	
		13-150years	11.6 - 14
Platelet Count (PLT) x10⁹ /L	Female	0-1 day	100 - 450
		2-3days	210 - 500
		4-7days	160 - 500
		0-1 month	200 - 500
		0-2 months	210 - 650
		0-1 year	200 - 550
		2-6 years	200 - 490
		7-12 years	170 - 450
	13-150 years	150 - 450	
	Male	0-1 day	100 - 450
		2-3 days	210 - 500
		4-7 days	160 - 500
		0-1 month	200 - 500
		0-2 months	210 - 650

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		0-1 year	200 - 550
		2-6 years	200 - 490
		7-12 years	170 - 450
		13-150 years	150 - 450
White Cell Count (WCC) x10⁹ /L	Female	0-1 day	10 - 26
		2-3days	7 - 23
		4-7days	6 - 22
		0-1 month	5 - 19
		0-2 months	5 - 15
		0-1 year	6 - 16
		2-6 years	5 - 15
		7-12 years	5 - 13
		13-150 years	4 - 10
	Male	0-1 day	10 - 26
		2-3 days	7 - 23
		4-7 days	6 - 22
		0-1 month	5 - 19
		0-2 months	5 - 15
		0-1 year	6 - 16
		2-6 years	5 - 15
		7-12 years	5 - 13
		13-150 years	4 - 10
Neutrophil Count x10⁹ /L	Female	0-1 day	4 - 14
		2-3days	3 - 5
		4-7days	3 - 6
		0-1 month	3 - 9
		0-2 months	1 - 5
		0-1 year	1 - 7
		2-6 years	1.5 - 8
		7-12 years	2 - 8
		13-150 years	2 - 7
	Male	0-1 day	4 - 14
		2-3 days	3 - 5
		4-7 days	3 - 6
		0-1 month	3 - 9
		0-2 months	1 - 5
		0-1 year	1 - 7
		2-6 years	1.5 - 8
		7-12 years	2 - 8
		13-150 years	2 - 7
Lymphocyte Count x10⁹/L	Female	0-1 day	3 - 8
		2-3days	2 - 8
		4-7days	3 - 9
		0-1 month	3 - 16

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		0-2 months	4 - 10
		0-1 year	3.5 - 11
		2-6 years	6 - 9
		7-12 years	1 - 5
		13-150 years	1 - 3
	Male	0-1 day	3 - 8
		2-3 days	2 - 8
		4-7 days	3 - 9
		0-1 month	3 - 16
		0-2 months	4 - 10
		0-1 year	3.5 - 11
		2-6 years	6 - 9
		7-12 years	1 - 5
		13-150 years	1 - 3
Monocyte Count x10 ⁹ /L	Female	0-1 day	0.5 - 2
		2-3days	0.5 - 1
		4-7days	0.1 - 1.7
		0-1 month	0.3 - 1
		0-2 months	0.4 - 1.2
		0-1 year	0.2 - 1
		2-6 years	0.2 - 1
		7-12 years	0.2 - 1
		13-150 years	0.2 - 1
	Male	0-1 day	0.5 - 2
		2-3 days	0.5 - 1
		4-7 days	0.1 - 1.7
		0-1 month	0.3 - 1
		0-2 months	0.4 - 1.2
		0-1 year	0.2 - 1
		2-6 years	0.2 - 1
		7-12 years	0.2 - 1
		13-150 years	0.2 - 1
Eosinophil Count x10 ⁹ /L	Female	0-1 day	0.1 - 1
		2-3days	0.1 - 2
		4-7days	0.1 - 0.8
		0-1 month	0.2 - 1
		0-2 months	0.1 - 1
		0-1 year	0.1 - 1
		2-6 years	0.1 - 1
		7-12 years	0.1 - 1
		13-150 years	0.02 - 0.5
	Male	0-1 day	0.1 - 1
		2-3 days	0.1 - 2
		4-7 days	0.1 - 0.8

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		0-1 month	0.2 - 1
		0-2 months	0.1 - 1
		0-1 year	0.1 - 1
		2-6 years	0.1 - 1
		7-12 years	0.1 - 1
		13-150 years	0.02 - 0.5
Basophil Count x10 ⁹ /L	Female	0-1 day	0.02 - 0.1
		2-3days	0.02 - 0.1
		4-7days	0.02 - 0.1
		0-1 month	0.02 - 0.1
		0-2 months	0.02 - 0.1
		0-1 year	0.02 - 0.1
		2-6 years	0.02 - 0.1
		7-12 years	0.02 - 0.1
		13-150 years	0.02 - 0.1
	Male	0-1 day	0.02 - 0.1
		2-3 days	0.02 - 0.1
		4-7 days	0.02 - 0.1
		0-1 month	0.02 - 0.1
		0-2 months	0.02 - 0.1
		0-1 year	0.02 - 0.1
		2-6 years	0.02 - 0.1
		7-12 years	0.02 - 0.1
		13-150 years	0.02 - 0.1

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Appendix 3

Reporting of Blood Films by Medical Staff

Department of Haematology, Belfast City Hospital

Introduction:

Examination of a blood film is an essential procedure in the clinical assessment, investigation, and interpretation of abnormal FBC results. Initial screening is undertaken by qualified Biomedical Scientist staff, who will refer samples to the lab registrar:

- (i) In new patients where there is a difficulty in interpretation
- (ii) In cases of suspected primary haematological disorders.

Specimen Preparation:

Blood films are spread from fresh EDTA samples in the laboratory. They should be prepared within two hours (but not exceeding 12 hours) of blood collection. Well-spread, well-stained films are required to ensure reliable information can be acquired.

Methods:

A systematic approach to blood film examination is essential to correct interpretation.

- FBC samples are selected for blood film examination according to numerical criteria, analyser flags, and clinical flags
- The blood film is referred by Biomedical Scientist staff along with the FBC request form &/or the morphology referral form
- Referrals are placed in a tray basket on the registrar's bench in the lab
- The lab registrar should liaise regularly – at least twice daily - with BMS staff, to look out for samples requiring urgent action

Results:

Interpretative comments are entered onto LIMS. Clinical interpretation is dependent on the patient's history, FBC results, other investigations, and comprehensive knowledge and experience of clinical haematological practice. Therefore, inexperienced registrars must discuss all cases with the duty lab consultant.

References:

- Bain, BJ. Morphology of blood cells. Ch 3. pp 61-174. In: Blood Cells: A Practical Guide. Blackwell Publishing Ltd, 4th Edition, 2006.
- Bain, BJ. Blood film morphology in health and disease. Ch5, pp 79-113. In: Practical Haematology. Churchill Livingstone Elsevier Ltd, 10th Edition, 2006.

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Appendix 4

WBC in Africans and Caribbeans of African Lineage

Department of Haematology, Belfast City Hospital

95 percentile ranges (x10⁹/L)

Origin	Male		Female	
	WBC	Neut	WBC	Neut
African	2.8-7.2	0.9-4.2	3.0-7.4	1.3-3.7
Caribbean	3.1-9.4	1.2-5.6	3.2-10.6	1.3-7.1

From Barbara Bain, *Blood Cells: A Practical Approach*, 2006.

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Appendix 5

Haemoglobinopathy & Thalassaemia Investigation

Department of Haematology, Belfast City Hospital

Haemoglobinopathy screening for haemoglobin variants (Hb V), alpha thalassaemia and beta thalassaemia is provided by Specialist Red Cell Investigations, Haematology, Belfast City Hospital.

All samples referred for haemoglobinopathy investigation are initially screened for haemoglobin variants and for HbA₂ quantitation by capillary electrophoresis (CE). Provisional results are confirmed by isoelectric focusing (IEF). These results are interpreted in conjunction with mean cell haemoglobin (MCH) values, family origin and with/without Hb F levels.

DNA analysis is only performed on a limited number of cases and when indicated by the criteria as outlined below.

N.B. Samples are referred to the Regional Molecular Diagnostic Service [RMDS] for DNA analysis.

Action Values

HbA₂: The action value for Hb A₂ has been set at 3.5%. A level of $\geq 3.5\%$ is indicative of beta thalassaemia and below suggestive of alpha thalassaemia.

MCH: The action value for MCH is below 27 pg.

Hb F: Action values for Hb F are above 10% when MCH is 27 pg or 5% when MCH is less than 27 pg.

Hb Variant

- All Hb variants are referred for DNA analysis except HbS
- Only HbS can be reported without DNA confirmation providing
 - Both CE and IEF indicate the presence of HbS
 - MCH is above 27 pg
- Any discrepancies between IEF and CE are referred for DNA analysis.

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Alpha thalassaemia

- If the HbA₂ is below 3.5% and MCH is less than 25 pg, the individual should be assessed for the possibility of α^0 thalassaemia heterozygosity or α^+ thalassaemia homozygosity and referred for DNA analysis by GAP-PCR and/or PCR-direct sequencing of the alpha globin gene.
- If the HbA₂ is below 3.5% but MCH is ≥ 25 pg but below 27 pg then α^+ thalassaemia carrier (heterozygous) state or possible iron deficiency is indicated and no DNA analysis is required

Beta thalassaemia

- A level of Hb A₂ $\geq 3.5\%$ in the presence of a MCH below 27 pg indicates heterozygosity for β thalassaemia and no DNA analysis is required.

Hb Variant and Thalassemia

- If the level of the Hb V detected by CE is greater than the Hb A level suggests possible co-existence of Hb V with β^+ thalassaemia especially if the MCH is below 25 pg. Refer for DNA analysis for both β thalassaemia and Hb Variant.
- If the Hb V is present at a level of below 30 % of total Hb (< 25% if Hb V is Hb E) and MCH is below 25 pg there is indication of a possible co-existing alpha thalassaemia or iron deficiency. Refer for DNA analysis for both α thalassaemia and Hb Variant.

Haemoglobin F

High HbF levels are associated with hereditary persistence of foetal haemoglobin (HPFH) or delta beta ($\delta\beta$) thalassaemia. HbF levels can also be higher in the presence of Hb Variants and in children under 1 year of age.

- If MCH is ≥ 27 pg, HbF is $\geq 10\%$ - suggests HPFH
- If MCH is < 27pg, HbF is $\geq 5.0\%$ - suggests $\delta\beta$ thalassaemia

Antenatal Screening

All women on booking are offered antenatal screening. Once accepted they are screened for the presence of a haemoglobinopathy by CE and IEF. These results are interpreted in conjunction with mean cell haemoglobin (MCH) values and family origin as indicated on the family origin questionnaire (FOQ). Baby's biological father is screened when the criteria as outlined below is met.

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- **Hb V**

- If one of the variants as listed in Box 1 is detected then screening of the baby's biological father is recommended.
- DNA analysis is performed as defined previously.

- **Alpha thalassaemia**

- Possible alpha⁰ thalassaemia heterozygosity or alpha⁺ thalassaemia homozygosity when the MCH is less than 25 pg and screening of the baby's biological father is recommended. Refer for DNA analysis to distinguish between alpha⁰ and alpha⁺ thalassaemia
- Possible alpha⁺ thalassaemia carrier status when the MCH is between 25 and 27 pg. No requirement for testing of baby's biological father or for DNA analysis.
- If the family origin of either parent is unknown and the MCH is less than 25 pg in the mother then DNA analysis of both parents is recommended

- **Beta thalassaemia**

- If beta thalassaemia has been identified in an antenatal patient according to the above criteria then screening baby's biological father is recommended
- Retain mother's antenatal sample until baby's biological father testing has been completed
- If baby's biological father also has β thalassaemia or is a carrier then both parental samples are referred for DNA analysis

- **Combined Hb Variant and Thalassaemia**

- If either alpha or beta thalassaemia has been identified with a significant maternal Hb variant then screen baby's biological father
- If baby's biological father also has a significant haemoglobinopathy or is a carrier then both parental samples are referred for DNA analysis

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Box 1: Maternal conditions requiring testing of the baby's biological father.

Significant maternal haemoglobinopathies

The following maternal haemoglobinopathies should be detected by antenatal screening and are important for maternal care:

- Hb SS
- Hb SC
- Hb SD^{Punjab}
- Hb SE
- Hb SO^{Arab}
- Hb S/Lepore and Hb Lepore/ β thalassaemia
- Hb S/ β thalassaemia
- Hb S/ $\delta\beta$ thalassaemia
- HbH disease ($--/\alpha$)
- β thalassaemia major/intermedia
- Hb E/ β thalassaemia

Carrier states in biological mother:

- HbS
- HbC
- HbD^{Punjab}
- HbE
- HbO^{Arab}
- Hb Lepore
- β thalassaemia
- $\delta\beta$ thalassaemia
- α^0 thalassaemia ($--/\alpha\alpha$)
- Hereditary Persistence of Fetal Haemoglobin (HPFH)

Any compound heterozygote state including one or more of the above conditions.
Any homozygous state of the above conditions.

Iron Deficiency

Severe iron deficiency anaemia (Hb <80 g/l) can reduce the Hb A₂ level slightly (by up to 0.5%). Outside of pregnancy, anaemia should be treated and the haemoglobin analysis repeated when the patient is iron replete. During pregnancy, there is no justification for delaying the investigation for haemoglobinopathies whilst treating iron deficiency presumptively, as this will delay the process of identifying at-risk carrier couples, who could be offered prenatal diagnosis. Therefore, all antenatal samples where the MCH is below 25 pg regardless of their iron levels, are referred for DNA analysis.

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Reporting of Results:

The molecular diagnostic report for haemoglobinopathies will be combined into one comprehensive format under the one request item of HTMS (Haemoglobin Variant and Thalassaemia Molecular Screen). Both haemoglobin variants and thalassaemia for alpha and beta globin genes to be simultaneously reported. The report will also contain interpretation of the molecular screening, subsequent actions appropriate for the results and information regarding partner screening. The HTMS report will be a follow on from the Specialist Red Cell initial screening report (HTS Haemoglobin variant Thalassaemia Screen).

References

NHS Sickle Cell and Thalassaemia Screening Programme: Handbook for antenatal laboratories. Public Health England, October 2017

NHS Sickle Cell and Thalassaemia Screening Programme: Handbook for newborn laboratories. Public Health England, January 2017

Ryan K et al (2010). Significant haemoglobinopathies: guidelines for screening and diagnosis. Brit J Haematol, 149: 35–49.

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Appendix 6

Investigation of Malarial Parasites

Department of Haematology, Royal Victoria Hospital

Introduction: Malaria is a blood borne parasite which can be identified by microscopically examination of blood films. There are currently five malarial parasite species which affect Humans, *Plasmodium Falciparum*, *Plasmodium Ovale*, *Plasmodium Vivax*, *Plasmodium Malariae* and *Plasmodium Knowlesi*. Infection with *P. falciparum* or *P. Knowlesi* is a medical emergency, therefore, ALL first time sample requests for investigation of malarial parasites are treated as URGENT.

All patients **must** have a **Viral Haemorrhagic Fever (VHF)** risk assessment performed before testing for Malaria as the laboratory needs to adopt health and safety protocols to protect against the higher category pathogen e.g. Ebola etc.

Please check the website below for information on current outbreaks, endemic countries and risk assessment if the patient has travelled to an at risk country

Risk Assessment

https://www.gov.uk/government/uploads/system/uploads/attachment_data/file/478115/VHF_Algo.pdf

The outcome of the above assessment should be recorded on the Malaria Request Form and should be sent to the RVH, Haematology laboratory (use the link below to print the form):

Request Form

<https://belfasttrust.hscni.net/download/615/haematology-blood-bank/15669/h-1038-request-for-malarial-investigation-2.doc>

Specimen Preparation: Fresh 4ml EDTA samples are required for the investigation of malarial parasites. Samples must be **less than 12 hours old** and taken after or during a temperature spike. Older samples are unsuitable for assessment of malaria by microscopy as the anticoagulant affects the morphology of the parasite.

Methods: The malaria investigation includes a qualitative screening test using a commercial kit (BinaxNow) and traditional microscopy screening and identification.

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Microscopy: Performed for all malaria requests. Thin films stained with 1/10 Giemsa stain and viewed under microscope. Parasitaemia will be expressed as the percentage of red cells infected and performed for *P. Falciparum* and *P. Knowlesi* only.

Results: Reported as either No malarial parasites seen, Malarial parasites seen- Awaiting identification please treat as *P. Falciparum* until species confirmed or as the actual species identified i.e. *Plasmodium Falciparum*, *Plasmodium Ovale*, *Plasmodium Vivax*, *Plasmodium Malariae*, *Plasmodium Knowlesi* or a mixed infection.

First time positives and unidentifiable species will be referred to the Malaria Reference Laboratory located at the London School of Hygiene and Tropical Medicine (LSHTM).

Interval of testing: When there is a strong clinical suspicion of malaria but the initial films are reported as no malarial parasites seen repeat films should be made and examined after 12-24 hours and again after a further 24 hours.

Clinical Advice:

Contact the Infectious Diseases Team of the Belfast trust or alternatively the Infectious diseases consultant in Newcastle via switchboard.

References:

- Bailey J.W., Williams J., Bain, B.J., Parker-Williams J., Chiodini P.L. Guideline: the laboratory diagnosis of malaria. British Journal of Haematology, 2013, 163, 573-580.

**Department of Haematology
Royal Victoria Hospital
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Appendix 7

Sickling disorders - Sickle solubility testing

Department of Haematology, Belfast City Hospital

Sickle solubility testing is available at RVH and BCH labs. This is a rapid screening test. A positive result indicates the presence of HbS but does not distinguish between sickle cell disease (Hb SS), sickle cell trait (Hb AS), and various compound heterozygous states. In the acute setting, before confirmatory results are available, one should manage the patient as sickle cell disease.

(a) Positive sickle solubility test and normal blood film: Assume sickle cell trait.

(b) Positive sickle solubility test and any sickle cells or target cells on blood film: Assume sickle cell disease, irrespective of Hb (for example patients with HbSC disease may have a normal Hb but still have a clinically significant acute sickling crisis).

N.B. Sickle solubility tests are often negative in infants with sickle cell disease (due to the protective effect of HbF).

False positive sickle solubility results:

- Severe leucocytosis
- Hyperproteinaemia, eg myeloma
- Hyperlipidaemia
- Unstable haemoglobins – especially after splenectomy

False negative sickle solubility results:

- Low Hb
- Infants <6 months
- Post-transfusion

RJG Cuthbert,

Last updated August 2014

Adapted from Practical Haematology, 10th Edition, Churchill Livingstone, 2006.

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Appendix 8

Age Related Coagulation Reference Ranges for the ACL Top 550 Analyser

Test	Age	Male Low	Male High	Female Low	Female High
PT (secs)	0-1 day	10.1	15.9	10.1	15.9
APTT (secs)	0-1 day	31.3	54.5	31.3	54.5
FIB (g/l)	0-1 day	1.67	3.99	1.67	3.99
PT (secs)	0-1 month	10	14.2	10	14.2
APTT (secs)	0-1 month	32	55.2	32	55.2
FIB (g/l)	0-1 month	1.62	3.78	1.62	3.78
PT (secs)	0-3 months	10	14.2	10	14.2
APTT (secs)	0-3 months	29	50.1	29	50.1
FIB (g/l)	0-3 months	1.5	3.79	1.5	3.79
PT (secs)	0-6 months	10.7	13.9	10.7	14.2
APTT (secs)	0-6 months	28.1	42.9	28.1	42.9
FIB (g/l)	0-6 months	1.5	3.87	1.5	3.87
PT (secs)	150 years	10.1	14.3	10.1	14.3
APTT (secs)	150 years	21	32	21	32
FIB (g/l)	150 years	1.8	4.3	1.8	4.3

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Appendix 9

ADAMTS13 Activity Request Form

Patient Name _____ Date of Birth _____ H&C _____

Local Hospital Number _____

Consultant in charge of patient: Name _____ Mobile _____ Email _____

Hospital & full address for report: _____

Contact name/number (must be available to take results): _____

Is this patient pregnant/ had a recent pregnancy?

YES/NO

Date & Time: Sample Collection _____

Last plasma infusion/exchange _____

Clinical details:

- Urgent request Routine request
- Suspected TTP (immune mediated)
- Suspected TTP (congenital)
- Suspected aHUS
- 1st acute presentation
- Acute relapse
- Monitoring
- Other (please specify) _____

FBC results

- RBCs _____
- WBCs _____
- Hb _____ Hct _____
- Platelets _____
- Blood Film Y/N
- Evidence of MAHA Y/N

Sample Requirements :

Standard Coagulation Tubes. Either as:

- 2 citrated blood samples despatched to arrive in the laboratory within 48 hours of sample collection as un-centrifuged whole blood (stored at room temperature)
- OR:
- Double spun plasma (at 2,000g) divided into 4 aliquots (minimum volume 150 µL in each). Freeze immediately at -70°C.

Send frozen / on dry ice to: HAEMOSTASIS LABORATORY, C FLOOR, BELFAST CITY HOSPITAL BELFAST BT9 7AB

- For urgent samples Tel: 028 950 40910
Samples must be in the lab by 12 noon if urgent and require testing on the same day. We normally test Mon-Fri, excluding weekends and bank holidays
- Please send a completed copy of this form with your request form.

ADAMTS13 LEVELS <10% WILL HAVE ALSO BE TESTED FOR THE PRESENCE OF AN ANTI-ADAMTS13 INHIBITOR

- If you suspect TTP please contact your regional TTP centre or the Haematologist on call for urgent advice Tel: 028 950 40444 (Mon-Fri 9am-5pm)
- If ADAMTS13 activity is >10% and you suspect aHUS contact the national aHUS service on Tel: 0191 2336161 and ask for the on-call consultant for aHUS

We will endeavour to ring results on the number you have provided. From time of receipt of sample in the lab urgent results will usually be available within 24 hrs. It remains the responsibility of the requestor to coordinate clinical management based on these results.

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Appendix 10

DOAC Levels

	Apix. C _{max} (ng/mL)	Apix. C _{min} (ng/mL)	Apix. Anti-Xa Activity Max (IU/mL)	Apix. Anti-Xa Activity Min (IU/mL)
	Median [5th, 95th Percentile]			
<i>Prevention of VTE: elective hip or knee replacement surgery</i>				
2.5 mg twice daily	77 [41, 146]	51 [23, 109]	1.3 [0.67, 2.4]	0.84 [0.37, 1.8]
<i>Prevention of stroke and systemic embolism: NVAf</i>				
2.5 mg twice daily*	123 [69, 221]	79 [34, 162]	1.8 [1.0, 3.3]	1.2 [0.51, 2.4]
5 mg twice daily	171 [91, 321]	103 [41, 230]	2.6 [1.4, 4.8]	1.5 [0.61, 3.4]
<i>Treatment of DVT, treatment of PE and prevention of recurrent DVT and PE (VTEt)</i>				
2.5 mg twice daily	67 [30, 153]	32 [11, 90]	1.0 [0.46, 2.5]	0.49 [0.17, 1.4]
5 mg twice daily	132 [59, 302]	63 [22, 177]	2.1 [0.91, 5.2]	1.0 [0.33, 2.9]
10 mg twice daily	251 [111, 572]	120 [41, 335]	4.2 [1.8, 10.8]	1.9 [0.64, 5.8]

* Dose adjusted population based on 2 of 3 dose reduction criteria in the ARISTOTLE study. Although treatment with apixaban does not require routine monitoring of exposure, a calibrated quantitative anti-Factor Xa assay may be useful in exceptional situations where knowledge of apixaban exposure may help to inform clinical decisions, e.g., overdose and emergency surgery.

Rivaroxaban plasma concentrations after therapeutic doses based on phase II data and simulated virtual data

Dose	Clinical setting	C _{trough} (µg/l)	C _{max} (µg/l)
2.5 mg bid	Acute coronary syndrome	16 (6–34)*	44 (28–66)*
10 mg od	VTE prevention after total hip replacement	9 (1–38) [#]	125 (91–196) [#]
15 mg od	Stroke prevention in patients with AF (CrCl ≤50 ml/min)	57 (18–136) [‡]	229 (178–313) [‡]
20 mg od	DVT treatment (continued treatment)	26 (6–87) [§]	270 (189–419) [§]
20 mg od	Stroke prevention in patients with AF (CrCl >50 ml/min)	44 (12–137) [‡]	249 (184–343) [‡]

*Estimated parameters at steady state – median values (5th–95th percentile range).

[#]Estimated parameters at steady state – median values (5th–95th percentile range) in patients undergoing hip replacement surgery. [‡]Estimated parameters at steady state – geometric means (5th–95th percentile range) in stroke prevention in patients with AF (Bayer HealthCare Pharmaceuticals and Janssen Research & Development, LLC: data on file). [§]Estimated parameters at steady state – geometric means (5th–95th percentile range) in phase II studies in the acute treatment of DVT.

Reference: ‘Summary of Product Characteristics, Bristol-Myers Squibb-Pfizer’ (Updated 30-Oct-2017).

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Appendix 11

Strategy for handling peripheral blood samples

**Department of Haematology, Belfast City Hospital
Flow Cytometry Laboratory**

1. Baseline data:

- Clinical history
- FBC results
- Blood film morphology

2. Peripheral blood flow cytometry indicated:

- Lymphocytosis $\geq 4.5 \times 10^9/L$ persisting for >3 months
- Circulating blasts or other abnormal cells
- Investigation of PNH and HS according to existing guidelines

Blood film morphology and interpretative comments on flow results will be reported.

3. Peripheral blood flow cytometry not indicated:

- No clinical details (flow will be done if FBC/blood film provide guidance)
- Isolated cytopenias
- Isolated leuco-erythroblastic change, ie without blasts
- Leucocytosis due to isolated neutrophilia, eosinophilia, basophilia or monocytosis, ie without blasts
- Inappropriate/speculative investigation of systemic disorders

Blood film morphology and a comment on why flow is not indicated will be reported, as well as any suggestions for further investigation/follow-up.

4. Lymphocyte subset requests will be despatched to the Immunology laboratory at RVH.

RJG Cuthbert, February 2014

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Appendix 12

Proposals for a revised strategy for handling bone marrow and body fluid samples

Department of Haematology, Belfast City Hospital Flow Cytometry Laboratory

1. Specific question on referring request form: eg CSF sample sent with “? lymphoma”

The morphology will be reviewed and the relevant flow panel will be done, for example a B-cell screen for B-cell lymphoma.

2. No specific question on referring request form: eg BM sample sent with “pancytopenia ?cause” or “anaemia ?cause”

The morphology will be reviewed. If there are any suspicious morphological findings a single tube TBNK screen will be done. This gives us the opportunity to examine the scatter plots which are helpful in deciding the pathway of investigation. If any abnormalities are detected on the screen an appropriate detailed flow investigation will be undertaken.

If there are no suspicious findings the morphology will be reported and flow will not be undertaken.

3. For “?myeloma”: A plasma cell panel – CD45/CD138/CD38/kappa/lambda – will be undertaken, but a routine B-cell screen will no longer be done.

4. For BCH marrows: Appropriate flow studies will be conducted and reported on LabCentre without morphology comments. The immunophenotype will be written on the integrated bone marrow report work-sheet. The BM aspirate morphology, trephine biopsy findings including immuno, and any molecular results will be reported as usual by the lab registrar/respective consultant.

RJG Cuthbert, January 2016

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Appendix 13

Referral of samples for EMA binding studies

Department of Haematology, Belfast City Hospital

Sample required: Peripheral blood in EDTA

This investigation is expensive, and time-consuming. It takes one member of lab staff out of all other duties for at least half a day. Due to restrictions on staff overtime it is not possible to process samples out of hours.

Please contact the flow lab (tel. no. 028-950-40913) to book a suitable time to have the test done.

Patient identification - Minimum acceptance criteria:

- First name and surname
- DOB
- Hospital number
- Referring consultant

Clinical Details:

Please provide clinical details and, if possible, a summary of relevant investigations. It is not enough to write “? Hereditary spherocytosis”.

**The reliability of results is poor in samples held at +4°C for more than 48 hours.*

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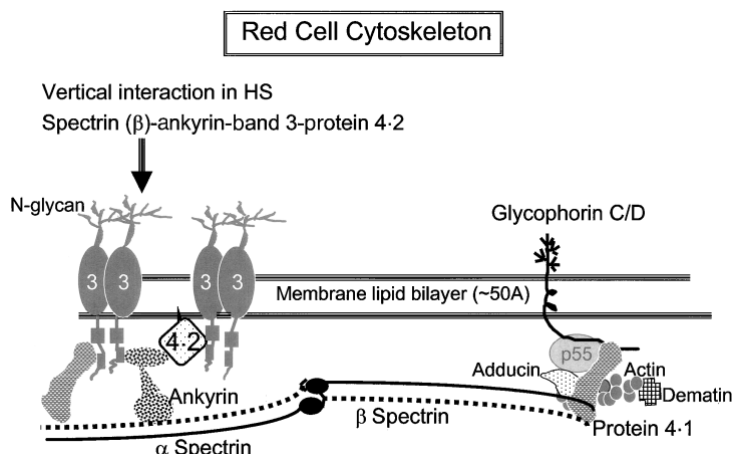
Appendix 14

Notes and Guidance on the Use of Eosin-5-maleimide (EMA) Binding in the Diagnosis of Hereditary Spherocytosis

Department of Haematology, Belfast City Hospital

The RC cytoskeleton is a spectrin-based network of proteins located on the inside of the RC membrane. Deficiency of any one component destabilises the cytoskeleton resulting in loss of RC membrane, and causing the characteristic RC cell morphology and shortened survival.

Eosin-5-maleimide (EMA) has a high affinity for Band 3 of the RC cytoskeleton. The maleimide moiety binds to a lysine residue in the extracellular portion of Band 3. Eosin lodges in the transmembrane core of Band 3. Up to 95% of EMA fluorescence is associated with Band 3 binding. The remainder is contributed by binding to other RC integral proteins. In HS, because of the membrane loss, there is reduced Band 3 expression no matter what underlying genotype is present.



The diagnosis of HS is usually straightforward, based on the clinical history, family history, physical examination (splenomegaly, jaundice) and laboratory data - especially RC indices, morphology, and reticulocyte count.

Other causes of haemolytic anaemia must be excluded, particularly autoimmune haemolytic anaemia. In neonates haemolysis caused by maternal alloantibody must be excluded.

A confirmatory test is indicated when the above diagnostic criteria are not met, and other causes of haemolysis have been excluded:

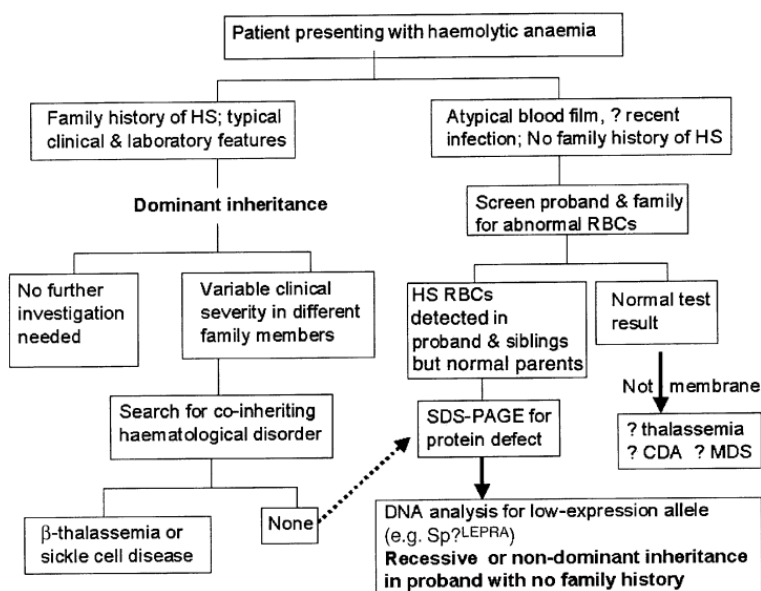
- Blood film is atypical
- No clear pattern of inheritance

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- Proband has an on-going mild haemolytic process with an apparently normal FBC
EMA has a specificity of approximately 99% and sensitivity of 92-95% for HS.
- Hereditary pyropoikilocytosis (HPP) and hereditary elliptocytosis (HE) can be differentiated from HS, based on the graded reduction in fluorescence intensity: HPP < HS < HE ≤ normal. Clearly, HPP and HE have distinctive RC morphology.
- EMA binding cannot distinguish HS from some rare RC disorders – CDA II, Melanesian ovalocytosis and cryohydrocytosis – but these have distinctive RC morphology and clinical features.

RJG Cuthbert, July 2010 (reviewed July 2015)

BCSH Recommended Approach to Investigation of HS



- Newly diagnosed patients with a family history of HS, typical clinical features (splenomegaly) and laboratory investigations (spherocytes, raised MCHC, increase in reticulocytes) do not require any additional tests.
- If the diagnosis is equivocal, for example, where there are a few spherocytes on the film but no other laboratory, clinical or family evidence, screening with the EMA binding test HS is helpful. The high predictive value can be improved further when used in conjunction with clinical information and red cell indices.
- Confirmation of the diagnosis may be necessary in selected cases if the EMA screening produces equivocal results: SDS-PAGE is recommended.
- Diagnosis of HS does not require molecular analysis of affected genes.

References:

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Bolton-Maggs PHB et al. Guidelines for the diagnosis and management of hereditary spherocytosis. Br J Haematol, 2004; 126: 455-474

Kar R et al. Evaluation of eosin-5-maleimide flow cytometric test in diagnosis of hereditary spherocytosis. Int J Lab Hematol, 2010; 32: 8-16

King M-J et al. Rapid flow cytometric test for the diagnosis of membrane-associated haemolytic anaemia. Br J Haematol, 2000; 111: 924-933

King M-J et al. Eosin-5-maleimide binding to band 3 and Rh-related proteins forms the basis of a screening test for hereditary spherocytosis. Br J Haematol, 2004; 124: 106-113

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Appendix 15

Guidelines on PNH Screening by Flow Cytometry

Department of Haematology, Belfast City Hospital

The proposed classification scheme for PNH has three main categories covering the spectrum of disease presentation:

1. Classical PNH which includes haemolytic and thrombotic patients
2. PNH in the context of other primary disorders such as aplastic anaemia
3. Subclinical PNH with a small PNH clone but no evidence of haemolysis or thrombosis

Indications for PNH Screening:

- Intravascular Haemolysis:
 - Unexplained haemoglobinuria and/or haemosidinuria
 - Unexplained Coomb's-ve haemolytic anaemia - When characteristic RBC abnormalities such as spherocytes, sickle cells, schistocytes, etc. are absent
- Thrombosis:
 - Thrombosis at unusual sites - Budd-Chiari syndrome, portal, splenic, splanchnic veins or cerebral sinuses
 - Thrombosis and associated intravascular haemolysis and/or cytopenias
- Cytopenias:
 - Aplastic anaemia
 - Hypoplastic MDS
 - Other unexplained cytopenias – PNH screening is indicated only if detailed bone marrow workup has proven uninformative

Routine PNH Screening Not Indicated:

- Coombs+ve haemolytic anaemia – PNH screening is not required in the absence of other indications
- Abdominal pain or dysphagia - PNH screening is not required, unless there is evidence of intravascular haemolysis
- Isolated anaemia - PNH screening is not indicated
- MDS other than hypoplastic MDS - PNH screening is not indicated, unless there is evidence of intravascular haemolysis

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Follow-up:

- Established diagnosis of PNH – Monitor clone size annually, or more frequently if there is any clinical change
- During ecluzimab therapy according to treatment protocol
- Aplastic anaemia with a small clone - Serial monitoring to predict progression to haemolytic PNH

Routine Follow-up Screening Not Indicated:

- MDS found to have a PNH clone – PNH monitoring is not indicated - rarely, if ever, progresses to clinical PNH

Notes on Interpretation of Results

PNH is a rare disorder with an incidence of approximately 1/ 10⁶/ year. However, screening of appropriate patients is important, because PNH is a chronic disease with a profound impact on quality of life and survival.

Diagnosis of PNH

Ham's test - Neither specific nor sensitive, and cumbersome to perform.

Complement lysis sensitivity – Laborious, difficult to standardise and poorly sensitive. However, the test led to the recognition of RBC's with intermediate complement sensitivity as well as the most abnormal PNH cells:

- Type I Normal RBC's
- Type II RBC's with intermediate complement sensitivity
- Type III RBC's with exquisite complement sensitivity

Flow cytometry is used to detect populations of GPI anchor-deficient cells, and is the method of choice for diagnosis and monitoring PNH.

The routine flow assay has a sensitivity of approximately 1% when 3000-5000 acquisition events are analysed. It is suitable for use as a screening test to detect patients with large clones associated with classical PNH, and can also detect smaller clones in patients with aplastic anaemia and subclinical PNH.

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Higher sensitivity assays, capable of detecting clones $\leq 0.01\%$, have highly variable performance characteristics, and their role in routine clinical practice has not been established.

RBC Analysis

Haemolysis and transfusion may lead to underestimation the size of a RBC clone. Thus WBC clones are frequently detected when RBC clones are not detectable. Comparing the relative sizes of RBC and WBC clones may provide useful clinical information.

WBC Analysis:

Assessment of PNH populations in WBC's is the best method for assessing the true size of a PNH clone. Both monocytes and neutrophils are suitable targets. The clone size measured in each population agrees relatively closely. Assurance gained by detecting the abnormality in both populations adds to the confidence in diagnosis.

Results:

When there is a high clinical suspicion of PNH, interpretation of immunophenotyping studies that demonstrate the presence of large PNH clones is straightforward. Patients with >20% Type III RBC's are likely to have overt intravascular haemolysis.

Patients with large Type II populations and absent or minimal Type III cells may have a reticulocytosis and modestly elevated LDH, but have less haemolysis than a patient with an equivalent number of Type III cells.

Small clones can be reliably detected in many patients with aplastic anaemia and hypoplastic MDS, though their prognostic value is uncertain.

Haemolytic anaemia associated with a small granulocyte clone should not be considered diagnostic of classical haemolytic PNH, but should trigger an investigation for other causes of haemolysis.

RJG Cuthbert, June 2010 (Reviewed June 2014)

This guideline and the notes on interpretation of results are adapted from: Borowitz MJ et al. Guidelines for the diagnosis and monitoring of paroxysmal nocturnal hemoglobinuria and related disorders by flow cytometry. Clinical Cytometry, 2010; 78B: 211-230.

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Appendix 16

Guidelines for Red Cell Volume Investigation

Department of Haematology, Belfast City Hospital

Red cell volume investigation involves iv administration of radiolabelled autologous red cells. It is undertaken to diagnose or exclude absolute erythrocytosis in patients who are negative for JAK-2 V617F mutation.

Indications:

4. Packed cell volume (haematocrit) elevated for >2 months:
 - Male PCV >0.52
 - Female PCV >0.48
 - Minimal or no venous occlusion when taking the blood sample
5. Erythrocytosis may be masked by Fe deficiency:
 - Typical FBC:
 - Hb usually upper end of normal range
 - PCV usually upper end of normal range
 - Low MCV
 - Raised RCC
 - Fe replacement should be undertaken only with extreme caution
 - PCV may rise rapidly and precipitate thrombosis
 - Monitor Hb and PCV weekly

Not indicated:

1. PCV normal (unless 2 above applies)
2. PCV grossly elevated:
 - Male: PCV >0.60
 - Female PCV >0.56

These patients have absolute erythrocytosis

Requesting the Investigation:

Requests should be submitted to:

Nuclear Medicine Department
Level 1 Imaging Centre
Royal Victoria Hospital

Use the appropriate radiology request form, and please submit giving full clinical details.

RJG Cuthbert Updated February 2014

This document is adapted from: Guidelines for the diagnosis, investigation and management of polycythaemia/erythrocytosis Br J Haematol, 2005;130:174-9.

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Appendix 17

Bone Marrow Aspirate and Trepine Biopsy

Department of Haematology, Belfast City Hospital

Introduction:

As part of the investigation of patients with a known or suspected haematological disorder, it is often necessary to perform a bone marrow aspirate and trephine biopsy procedure. The duty laboratory registrar undertakes the initial assessment and the results are scrutinised by the consultant haematologist before final authorisation.

Specimen Collection and Preparation:

Bone marrow aspirate slides are made at the patient's bedside using pink frosted glass slides and labeled with the patient's first name & surname, and the date of the procedure. Aspirate samples for flow cytometry and cytogenetics are placed in universal containers containing RPMI and heparin. Aspirate samples for molecular studies are placed in EDTA tubes. The trephine biopsy sample, if taken, is placed in a 5mL plain plastic tube containing 10% formalin labelled appropriately. The samples are delivered to the bone marrow/flow cytometry laboratory for processing.

Principles of Methods:

Various stains can be used to visualise blood cells and their precursors. These include Wright's stain and Perl's stain for haemosiderin performed on the aspirate smears, and H&E and Giemsa stains performed on the trephine sections. The stained slides are assessed, the various cell populations enumerated and then reported by suitably trained medical staff. The results help in the diagnosis and management of patients. Immunohistochemistry has an important role in trephine diagnosis, but is expensive. Judicious use of immunohistochemistry may be of diagnostic benefit to patients but must be used systematically.

Clinical Interpretation:

Examination of Wright's stained bone marrow aspirate slides, assessment of iron stores, examination of trephine sections and cytochemical stains is a complex procedure that requires specialist training and experience. It can affect a patient's diagnosis and treatment and as such should only be performed by appropriately qualified staff.

References:

Lee S-H et al. ICSH guidelines for the standardization of bone marrow specimens and reports. *Int J Lab Haematol*, 2008; 30: 349-64

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Appendix 18

Reticulocyte haemoglobin equivalent (RET-He) in the diagnosis of functional iron deficiency

Department of Haematology, Royal Victoria Hospital

Erythrocytes have a lifespan of 120 days. Therefore iron depletion in the bone marrow or changes in the iron supply cannot be detected early by classical haematological parameters, such as Hb, MCV, MCH, or even the proportion of hypochromic erythrocytes (%hypo).

When reticulocytes are released from the bone marrow into the peripheral blood they continue to mature for about two days. Thus, the measurement of the reticulocyte count allows “real-time” measurement and monitoring of the erythropoietic activity.

The Hb content of reticulocytes reflects the actual iron supply for Hb synthesis in the bone marrow and therefore provides qualitative information about these cells. With the measurement of RET-He an early detection of iron depletion in erythropoiesis is possible.

RET-He can help to distinguish between classical iron deficiency and functional iron deficiency (FID). In FID the iron stores are replete (normal ferritin levels), but the iron is not sufficiently available for Hb synthesis. Patients with anaemia of chronic disease (ACD) generally suffer from iron deficiency, with 20% of these patients showing a FID or a combined state of ID and FID.

In case of infection or inflammation it is very difficult to distinguish between depleted iron stores and functional iron deficiency. Classic biochemical markers such as ferritin and transferrin are influenced by the acute-phase-response. Low ferritin values may identify iron depletion, while normal or elevated levels do not give a clear indication of the actual iron available for erythropoiesis.

Reference range for RET-He is 28–35 pg

Indications for RET-He:

Diagnostic

- Distinguish classical and functional iron deficiency in ACD
- Detect early state of iron deficiency when biochemical markers are influenced (acute-phase response, pregnancy)

Therapeutic

- Monitoring of erythropoietin and/or iron therapy

RJG Cuthbert, August 2015

(Adapted from R Häusler: [Reticulocyte haemoglobin equivalent \(RET-He\)](#))

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Appendix 19

Methylene Blue Fresh Frozen Plasma or Cryoprecipitate

Department of Haematology, Belfast City Hospital

Anyone born from the 1st January 1996

Cytomegalovirus (CMV) negative red cells

- CMV negative red cells and platelet components should be provided for intra uterine transfusions and for babies up to the age of 20 weeks
- CMV negative blood components should be provided where possible for women, during pregnancy regardless of their CMV status (this applies during pregnancy but not labour or delivery). For emergency transfusions in this group, where CMV negative products are not readily available, leucodepleted components are recommended
- Granulocytes components should be CMV negative for CMV seronegative patients

Irradiated red blood cells

- Red cells for intrauterine transfusions
- Neonatal exchange transfusions
- Severe T lymphocyte immunodeficiency syndromes
- Top up transfusions if the baby has received intrauterine transfusions
- Bone marrow, stem cell transplant patients, and donors of bone marrow / stem cell
- Hodgkin's lymphoma
- Aplastic anaemia receiving ATG (and/or alemtuzumab)
- Patients on purine analogue (type of chemotherapy) drugs (fludarabine, cladribine and deoxycoformycin)
- Patient's on bendamustine, clofarabine, alemtuzumab
- HLA (human leucocyte antigen) selected units
- Transfusion from first or second degree relative

HLA matched red cells are indicated for:

- Patients awaiting a renal transplant
 - Those who have been transplanted in the past and are non-immunosuppressed.
- This is best practice however if in an emergency situation it is down to a clinical decision if HLA is not available.

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Patient groups for whom HLA matched platelets are indicated:

- Platelet dysfunction in chronic renal failure/uraemia
- Patients who have already demonstrated HLA antibodies and who may show platelet refractoriness

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Appendix 20

Provision of HLA selected platelet components for alloimmunised patients

Department of Haematology, Belfast City Hospital

Platelet refractoriness is defined as two consecutive failures of response to platelet transfusions, i.e. failure to achieve 24 hour post transfusion platelet count $> 20 \times 10^9/l$.

The majority of cases of platelet refractoriness are due to non-immune causes. These have been identified in studies by *Bishop et al* from the Melbourne Blood Centre and published in *Transfusion* as DIC consumption, anti-microbial therapy especially amphotericin B, pyrexia, hypersplenism, post BMT (immune dysregulation). Non-immune causes should be treated to optimise response to platelet substitution therapy. Immune causes which account for no more than 25% of cases are anti-ABO antibodies, anti-platelet specific antibodies and anti-HLA antibodies.

The first line of treatment should be ABO identical platelet components with high dose (NIBTS platelet components have the platelet yield on the label). Our quality monitoring data demonstrate average yield for single donor platelets (apheresis) of $280 \times 10^9/l$ but with reduced results for pools of buffy coat derived platelets of $238 \times 10^9/l$.

Where anti-platelet specific antibodies are identified, NIBTS will endeavour to provide HPA specific antigen negative platelet components.

Where anti-HLA antibodies are identified, NIBTS will search for matching donors but we often have to make use of selected mismatching because of lack of HLA identical or HLA homozygous haplotype donors.

It is imperative that NIBTS medical team receive follow up information on increment data and clinical response when HLA selected components are transfused. This will enable us to target donors and provide optimum support.

Therefore the following platelet support strategy will be adopted:

- Transfuse ABO identical platelet components (single donor platelets preferred).
- Select high yield dose single donor platelets.
- Provide HPA specific platelet components if appropriate.
- Provide HLA selected platelet components if appropriate. This would normally require confirmation of HLA antibodies and 3 month clinical assessment follow up.
- Return increment clinical response data to NIBTS medical team periodically.









Prepared by: Dr Morris and Dr Maguire Date: 10 September 2014

MP14/02

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Appendix 21

BHSCT Order of Draw for Blood Sciences (Updated Aug 2023)

Tube Type	Haematology Tests	Immunology Tests	Microbiology Tests
Blood Cultures			Blood Cultures*
 Sodium Citrate	ion, INR, XDP, Thrombophilia Lupus Anticoagulant, assays		
 Serum	Cold Agglutinin		
 Serum Separator		Autoimmune serology: Connective tissue disease antibodies (ANA, dsDNA/ENA), Hep2 anti-nuclear and centromere antibodies, Anti-phospholipid antibodies, Anti-CCP antibody, Coeliac screen, Gastric parietal cell antibody, Intrinsic factor antibody, Liver associated antibodies, Adrenal antibodies, Islet cell antibodies, Ovary/testes antibodies, Anti-acetylcholine receptor antibody, Anti paraneoplastic antibodies, Vasculitis screen (MPO/PR3/ANCA), Anti-GBM. Immunochemistry: Functional antibodies to PSSA, Antibodies to tetanus IgG, meningococcal C, haemophilus and diphtheria, CH50, AH50, C1 esterase functional, C1Q, Mannose Binding Lectin (MBL) Allergy: Total IgE, Allergen specific IgE, Extrinsic allergic alveolitis, Anaphylaxis, Mast cell tryptase.	Serology, Syphilis, ASOT.
 Lithium Heparin		Lymphocyte function (Prior arrangement only/requires control sample)	
 K3 EDTA	FBC, Retics, ESR, Maternal samples for Kleihauers, Lymphocyte, Immunophenotyping, PNH Screen, Malaria screening	Cellular: Lymphocyte subset phenotyping, Neutrophil function (requires control sample).	Samples for PCR
 Cross match	Group & Crossmatch, Direct Coombs, Card, Sample for Kleihauers.		
 Trace Elements			
 Fluoride Oxalate			

***NOTE:** For Blood Cultures - The rubber seal at top of blood culture bottle should be cleaned with alcohol wipe and allowed to dry before inserting needle to add blood.



For any test not listed above see the laboratory discipline manual for sample details. Laboratory manuals can be found at: <https://belfasttrust.hscni.net/service/laboratory-services/laboratories-user-manual/> or scan QR code opposite.

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Appendix 22

Non-Accidental Injury Screen (NAI)

