Alder Hey Children's NHS
NHS Foundation Trust



# A User's guide to The Medical Microbiology Department

The Microbiology Department is a UKAS accredited medical laboratory, No 9091.

Link to schedule is permitted

# Contents

Last review date: December 2022

# 1. Introduction

This handbook is intended to give an overview of the microbiology service at Alder Hey, and to provide outline guidance for the collection of samples for the diagnosis of infection. It is not intended to be a complete guide to all situations and advice can always be sought from the microbiology consultants or scientists.

**We anticipate further review of this user handbook in 2022 and welcome suggestions on how it can be improved; please contact the Lead Biomedical Scientist Mrs Catherine Hatch or Head of Department Dr Christopher Parry (see the section "Contact Details" below).**

Where samples are processed by external providers, the details are recorded on the Alder Hey report. If additional tests are required on samples processed elsewhere please approach the Alder Hey laboratory, rather than going directly to the external laboratory.

**COVID-19**

For information on the transport and collection of specimens for COVID-19 (SARS Cov-2) please refer to the COVID-19 hub on the Trust intranet

## 1.1. Useful websites

- Alder Hey Laboratory Medicine (Intranet): http://intranet/ClinicalSupport/SitePages/Pathology.aspx
- Alder Hey Antimicrobial and Infection Guidance (Intranet): http://intranet/DocumentsPolicies/SitePages/Antimicrobials.aspx
- United Kingdom Security Health Agency (External): UKHSA provides information to both the public and healthcare professionals in respect to infectious diseases. https://www.gov.uk/government/organisations/uk-health-security-agency

- The European Committee on Antimicrobial Susceptibility Testing (External): EUCAST deals with antimicrobial breakpoints and technical aspects of susceptibility testing. www.eucast.org

- COVID -19 HUB on intranet https://alderheynhsuk.sharepoint.com/sites/ahcovidhub/

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- UKAS :1SO 15189 Scope of accreditation
  https://www.ukas.com/wp-content/uploads/schedule_uploads/00007/9091-Medical-Single-1.pdf

## 2. Notifiable Infections

The following diseases are notifiable under the Health Protection (Notification) Regulations 2010:

| | |
|---|---|
| Acute encephalitis | Measles |
| Acute infectious hepatitis | Meningococcal septicaemia |
| Acute meningitis | Monkeypox |
| Acute poliomyelitis | Mumps |
| Anthrax | Plague |
| Botulism | Rabies |
| Brucellosis | Rubella |
| Cholera | Severe Acute Respiratory Syndrome (SARS) |
| Diphtheria | |
| Enteric fever (typhoid or paratyphoid fever) | Scarlet fever |
| Food poisoning | Smallpox |
| Haemolytic uraemic syndrome (HUS) | Tetanus |
| Infectious bloody diarrhoea | Tuberculosis |
| Invasive group A streptococcal disease | Typhus |
| Legionnaires' disease | Viral haemorrhagic fever (VHF) |
| Leprosy | Whooping cough |
| Malaria | Yellow fever |

Report other diseases that may present significant risk to human health under the category 'other significant disease'.

A positive culture / PCR / serology result is not required for notification; notification should be performed as soon as possible if any of the above are clinically suspected. The Cheshire and Merseyside Health Protection Team can be contacted on 0344 225 0562.

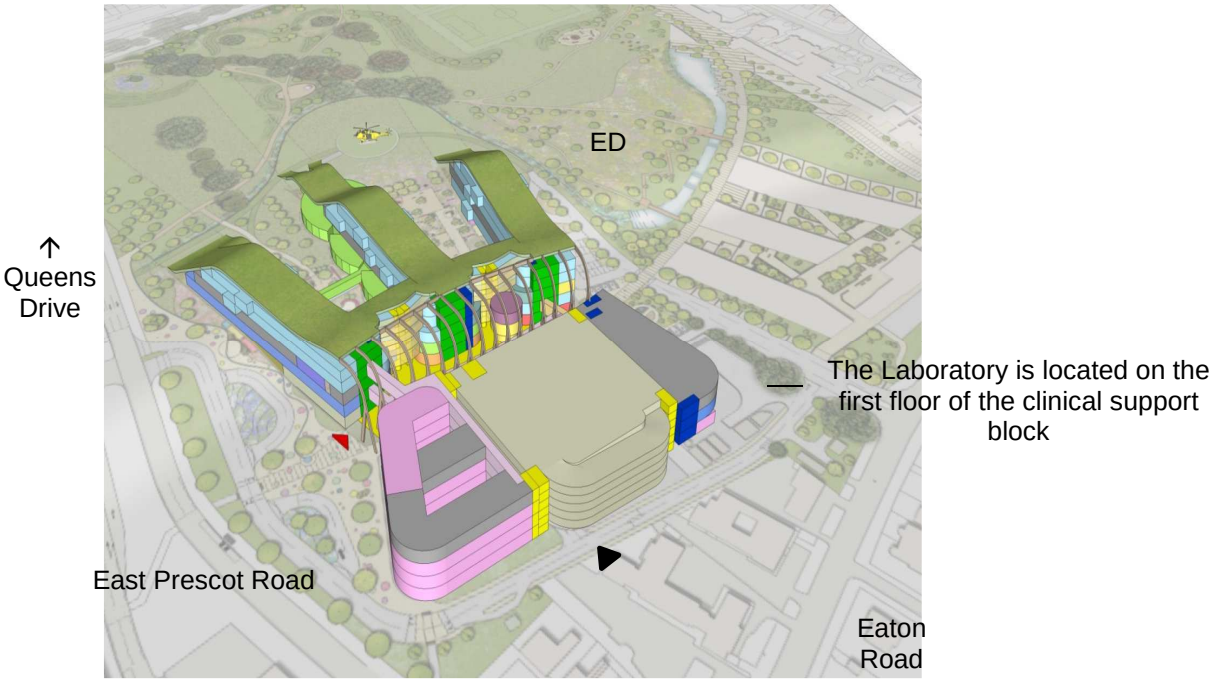
# 3. General Information

The Alder Hey Children's Medical Microbiology Department is a specialist paediatric Microbiology Laboratory which serves both the clinicians and patients of the hospital as well General Practitioners from Liverpool and the surrounding areas.

The aim of the laboratory is to provide an efficient patient-centred microbiology service which improves the investigation and management of infectious diseases in children.

Any samples that cannot be processed on-site are referred to other CPA or UKAS approved laboratories.

### 3.1. Where To Find Us:




The entrance to the multi-story visitor car park is located off East Prescot Road. There is a drop off point for A&E patients at the Eaton Road entrance.

External visitors should exit the atrium via the rotating doors next to WHSmith; there is an intercom button that connects to specimen reception next to the double doors to the right.

### 3.2. Contact Details

| | |
|---|---|
| **Postal Address** | Microbiology Department<br>Alder Hey Children's NHS Foundation Trust<br>Eaton Road<br>West Derby<br>Liverpool<br>L12 2AP |
| **Sat-Nav Postcode** | L14 5AB |
| **Telephone** | Main switchboard - 0151 228 4811 then request extension as indicated below |
| **Hays DX Address** | DX6961702<br>Old Swan 90L |
| **E-mail** | microbiology@alderhey.nhs.uk<br>Please note this is a generic e-mail address which may be seen by any of the microbiology medical or senior scientific staff. Please do not use for urgent clinical queries. |

### 3.3. Results / Enquiries

- **Monday – Friday: 8.30am – 5pm**
  Contact the laboratory on ext.5268 - direct line – 0151 293 5268 - external or internal 2268
  Out of hours external calls are referred through switchboard
  Internal calls contact the laboratory on ext. 2268

**Please note:** Results cannot be given to or discussed with family members.

# 4. Principal Services

Access to consultative and principal diagnostic services are available on a 24 hour basis.

## 4.1. Clinical Service

### 4.1.1. Clinical Microbiology Consultants

| Staff member | Title | Ext. | Direct (0151) | Email |
|---|---|---|---|---|
| Dr Róisín Mulqueen | Consultant Medical Microbiologist | 2566 | 252 5566 | Roisin.mulqueen@alderhey.nhs.uk |
| Dr Christopher Parry | Consultant Medical Microbiologist<br>Head of Department | 2566 | 252 5566 | Christopher.Parry2@alderhey.nhs.uk |
| Dr Mukul Acharya | Locum Consultant Medical Microbiologist | | | Mukul.Acharya@alderhey.nhs.uk |
| Prof. Nigel Cunliffe | Consultant Medical Microbiologist (Part-time)<br>Deputy Head of Department | | | NigelC@liverpool.ac.uk |
| Dr Anna Smielewska | Consultant Virologist & Strategic Lead for Virology | 4082 | Tuesdays | Anna.Smielewska@liverpoolft.nhs.uk |
| Dr Beatriz Larru | Consultant Paediatric Infectious Diseases. Trust Infection Control Doctor & DIPC | | | Beatriz.Larru@alderhey.nhs.uk |

### 4.1.2. Availability of Medical Consultant Services

Advice is available from either a Consultant Medical Microbiologist or a Consultant in Infectious Diseases and Immunology seven days a week. The Consultant Medical Microbiologists can be contacted using the telephone numbers above or via the hospital switchboard. The Paediatric Infectious Diseases and Immunology team can be contacted via switchboard or on bleep 244.

The out–of–hours clinical service is provided jointly by the Consultant Medical Microbiologists and the Paediatric Infectious Diseases Consultants.

- **A Consultant is always available for urgent clinical advice – contact is via switchboard.**

## 4.2. Diagnostic Service

The department provides a comprehensive microbiological service in medical bacteriology, mycology, virology, parasitology and serological investigations. Advice on the selection of appropriate diagnostic specimens, their collection and transport is available.

### 4.2.1. Senior Laboratory Scientists

| Staff member | Title | Ext. | Direct (0151) |
|---|---|---|---|
| Mrs Catherine Hatch | Lead Biomedical Scientist | 2267 | 252 5267 |
| Mrs Fiona Shaw | Senior Biomedical Scientist | 2268 | 252 5268 |
| Mrs Kathryn Ball | Senior Biomedical Scientist | 2268 | 252 5268 |

### 4.2.2. Laboratory Hours

Routine laboratory hours are Monday to Friday, 9 am to 5.30 pm, with a reduced service at weekends. An on-site service is available between 5.30 pm and 11 pm during the week and between 9 am and 11 pm at weekends and bank holidays to process any samples that are considered urgent. The BMS must be contacted in the laboratory on extension 2268 (or via mobile number through switchboard after 5.30pm), with the details of the request and how the specimen is being transported to the laboratory.

### 4.2.3. Urgent samples

Requests for urgent specimens to be processed after 11pm should be directed to the on-call Biomedical Scientist through switchboard. The following requests will be processed out-of-hours:

- Urine samples on children less than 6 months of age for microscopy, culture ± direct sensitivity (if positive for leucocyte esterase or nitrites on dipstick).
    - NB. Urines should be screened by dipstick by the requestor.
    - BMS staff are only expected to process dipstick positive samples out of hours.
- Urine samples from patients with known renal problems if the dipstick is positive – no age limits apply.
- CSF microscopy and culture
    - Please ensure the sample is ready to send to the laboratory before you contact the BMS or call them in from home.
- Material from sterile sites, e.g. synovial fluid, peritoneal fluid
- Pus from deep seated abscesses

Other requests can be discussed on a case-by-case basis with the Consultant on duty.

#### 4.2.4. Results of Particular Clinical Significance

Significant results are phoned through to the ward or relevant medical staff, irrespective of whether the original request is marked as urgent or routine. Significant results are also passed to the Paediatric Infectious Diseases team.

#### 4.2.5. Delays in the Examination Process

In the event of a significant delay in the examination of any sample, a comment will be added to the sample on Meditech and the Lead Biomedical Scientist or Consultant Microbiologist will inform the requesting doctor by email. Delays may be due to technical failure, failure of equipment or failure to supply by the manufacturer.

In the event of an extended delay samples will be sent to an accredited external laboratory for processing.

#### 4.2.6. Teaching and Training

The Department of Microbiology supports scientific and professional training for its staff, as well as the teaching of science students attending local universities and colleges.

The department welcomes enquiries from staff members who require a basic insight into microbiology services. Please contact the Lead Biomedical Scientist, Catherine Hatch, or the Head of Department, Dr Róisín Mulqueen, with any enquiries.

### 4.3. Environmental Microbiology

The Microbiology department undertakes a number of screening programmes throughout the Trust. Please note: This no longer includes water testing for the presence of *Pseudomonas aeruginosa* in water outlets and aerobiology of high risk areas, which has now been outsourced to a laboratory which is accredited for this testing with the United Kingdom Accreditation Service (UKAS).

### 4.4. Epidemiology

The laboratory contributes to national epidemiological surveillance via Public Health England.

### 4.5. Principal working relationships

#### 4.5.1. Infection Prevention and Control

The current Director of Infection Prevention and Control (DIPC) is the Paediatric Infectious Diseases Consultant Dr Beatriz Larru.

The Infection Prevention and Control Team:

| Role | Name | | Contact |
|---|---|---|---|
| DIPC/Infection Control Doctor Alder Hey | Dr Beatriz Larru | | Beatriz.Larru@alderhey.nhs.uk |
| Lead Infection Prevention & Control Nurse | Jo Keward | 2338 | Josephine.Keward@alderhey.nhs.uk |
| Infection Prevention & Control/PPE Nurse Specialist | Claire Oliver | 4175 | Claire.Oliver@alderhey.nhs.uk |
| Infection Prevention & Control Nurse Specialist | Tracey Styles | | Tracy.Styles@alderhey.nhs.uk |
| Infection Prevention & Control SSIS Nurse Specialist | Lisa Moore | | |
| Secondee | Hayley Clark | 2447 | Hayley.Clarke@alderhey.nhs.uk |
| Clinical services manager IPC/PPE | Cheryl Brindley | 3200 | Cheryl.Brindley@alderhey.nhs.uk |

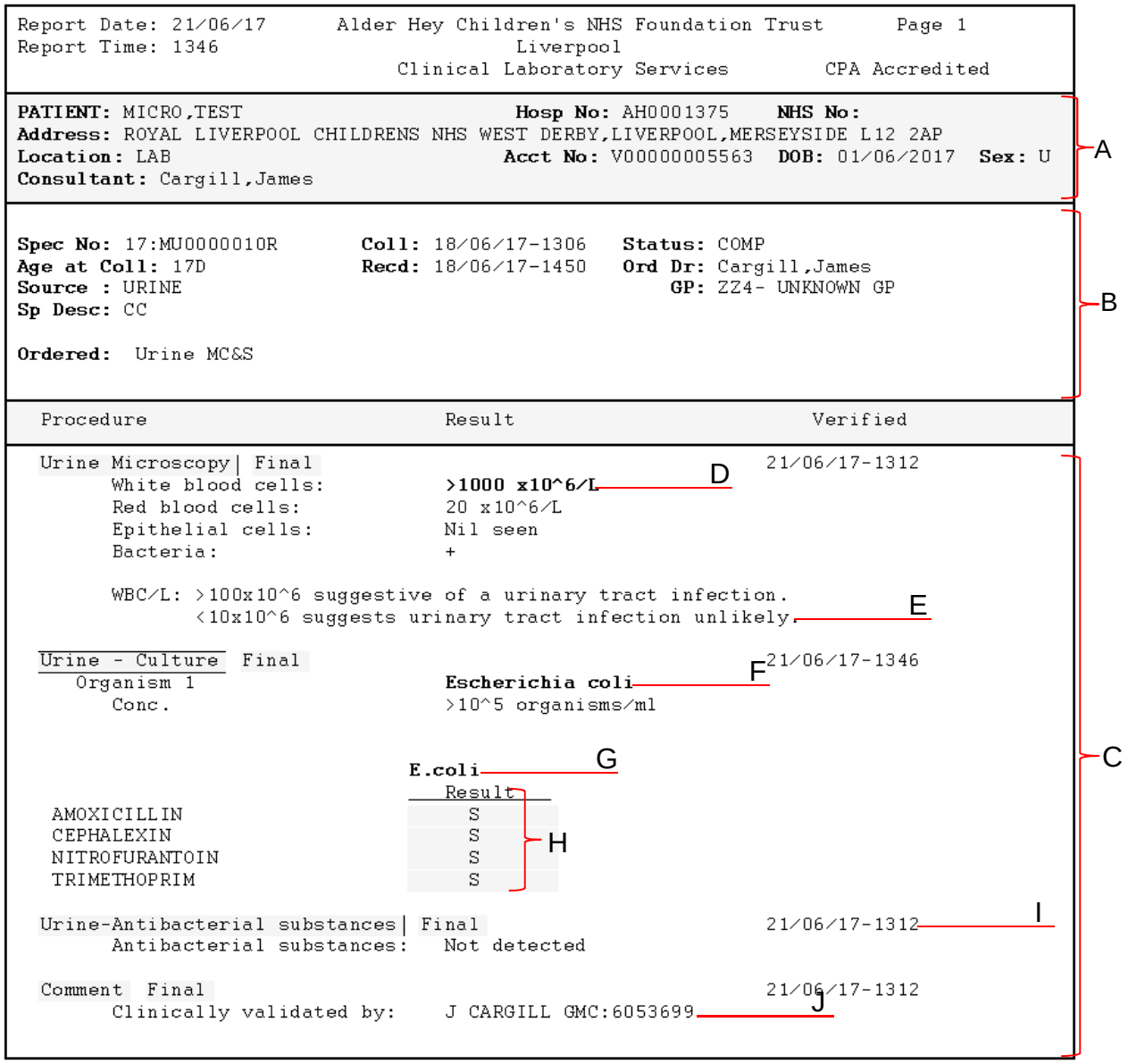
- The IPCT can be contacted by:
    - Email; infection.control@alderhey.nhs.uk,
    - Telephone; 4175 / 2485 / 2338 (Direct dial 0151 252 5485)
    - Bleep; 138
- Working hours 8am-5pm Monday-Friday

Where appropriate (e.g. respiratory viruses, multi-resistant Gram-negative isolates etc.) the microbiology report contains brief details about the appropriate infection control precautions to be taken.

#### 4.5.2. Paediatric Infectious Diseases and Immunology Team

The microbiology consultants work in cooperation with the Infectious Diseases clinicians. The Infectious Diseases team can usually be contacted on bleep 244.

# 5. The Microbiology Report Explained



Report Date: 21/06/17 Alder Hey Children's NHS Foundation Trust Page 1
Report Time: 1346 Liverpool
Clinical Laboratory Services CPA Accredited

**PATIENT:** MICRO,TEST **Hosp No:** AH0001375 **NHS No:**
**Address:** ROYAL LIVERPOOL CHILDRENS NHS WEST DERBY,LIVERPOOL,MERSEYSIDE L12 2AP
**Location:** LAB **Acct No:** V00000005563 **DOB:** 01/06/2017 **Sex:** U
**Consultant:** Cargill,James

A

**Spec No:** 17:MU0000010R **Coll:** 18/06/17-1306 **Status:** COMP
**Age at Coll:** 17D **Recd:** 18/06/17-1450 **Ord Dr:** Cargill,James
**Source :** URINE **GP:** ZZ4- UNKNOWN GP
**Sp Desc:** CC

**Ordered:** Urine MC&S

B

| Procedure | Result | Verified |
|---|---|---|
| Urine Microscopy \| Final | | 21/06/17-1312 |
| White blood cells: | **>1000 x10^6/L** | D |
| Red blood cells: | 20 x10^6/L | |
| Epithelial cells: | Nil seen | |
| Bacteria: | + | |
| WBC/L: >100x10^6 suggestive of a urinary tract infection. <10x10^6 suggests urinary tract infection unlikely. | | E |
| Urine - Culture Final | | 21/06/17-1346 |
| Organism 1 | **Escherichia coli** | F |
| Conc. | >10^5 organisms/ml | |

**E.coli** G

| | Result |
|---|---|
| AMOXICILLIN | S |
| CEPHALEXIN | S |
| NITROFURANTOIN | S |
| TRIMETHOPRIM | S |

H

| | | |
|---|---|---|
| Urine-Antibacterial substances \| Final | | 21/06/17-1312 I |
| Antibacterial substances: | Not detected | |
| Comment Final | | 21/06/17-1312 |
| Clinically validated by: | J CARGILL GMC:6053699 | J |

C

A standard microbiology report is shown below.

Key features are:

A. The patient details.

B. The sample details. This section includes the clinical details entered when the sample was ordered on Meditech.

C. The results for the Microbiology procedures performed in the laboratory.

D. Abnormal results are printed in bold text on the report.

E. Where available, normal ranges are displayed alongside the test results.

F. Bacterial and viral isolates are reported by name together with the concentration if applicable.

G. Where antibiotic susceptibility results are reported, a short form of the organism name is displayed.

H. Antibiotic susceptibility results are reported as:

S Sensitive to the agent

H Susceptible with Higher dosing*

*Please refer to the 'High Dose Antibiotic Table' in the Antimicrobial Prescribing Guidelines for optimal higher dose prescribing, when treating an infecting organism with a corresponding antimicrobial reported as “H”.

R Resistant to the agent

Where additional interpretation for the susceptibilities is indicated (e.g. to highlight specific resistance mechanisms of concern) these comments are reported below the susceptibilities.

Some additional tests (e.g. carbapenemase expression) may be reported as:

D Detected

N Absent (not detected)

Antimicrobial susceptibilities may also be reported with an MIC (Minimum Inhibitory Concentration) for that agent. These will be accompanied by an interpretation as above, and may include:

X No interpretation is possible

These MIC values are intended to be informative for the infectious diseases clinicians.

| | **S.aureus** |
|---|---|
| | MIC Result |
| VANCOMYCIN | 1.0 S |

I. The date the procedure was validated is displayed for each procedure

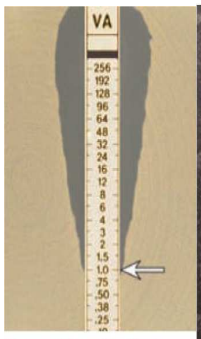
J. Positive results are clinically validated by one of the consultants; any additional interpretation is recorded here.

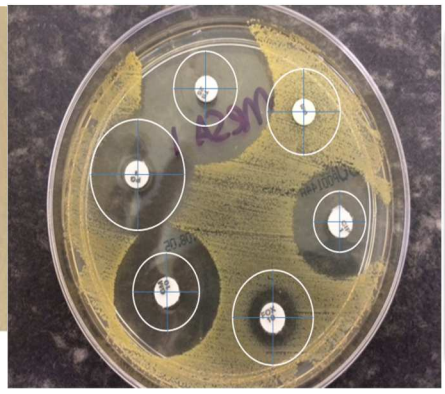
### 5.1. Antimicrobial Susceptibility Reporting

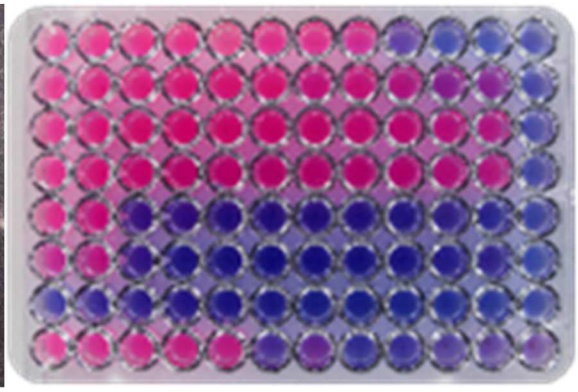
The department adheres to The European Committee on Antimicrobial Susceptibility Testing guidelines for the interpretation and reporting of antibiotic sensitivity results and antifungal sensitivity results. For some isolates susceptibility testing is referred to external laboratories (e.g. *Burkholderia* and similar species from cystic fibrosis patients, clinically significant moulds etc.).

The laboratory uses a combination of disc diffusion testing (where the zone of clearance around a disc containing a defined antibiotic concentration is used to determine susceptibility

or resistance), and gradient strips and broth micro dilution tests (where the specific MIC of an agent can be determined).





Disc diffusion Broth microdilution

Gradient strip

The laboratory is making increasing use of susceptibility testing on an automated platform. Sensitivity results are reported as:

- **S – Clinically Susceptible:** level of antimicrobial susceptibility associated with a high likelihood of therapeutic success.
- **H - Susceptible with Higher dosing***

  *Please refer to the 'High Dose Antibiotic Table' in the Antimicrobial Prescribing Guidelines for optimal higher dose prescribing, when treating an infecting organism with a corresponding antimicrobial reported as “H”.

  A microorganism is categorised as "Susceptible with Higher dosing" when there is a high likelihood of therapeutic success because exposure to the agent is increased by adjusting the dosing regimen or by its concentration at the site of infection.
  NB: EUCAST guidance calls this category: "Susceptible, Increased exposure*"
  *Exposure is a function of how the mode of administration, dose, dosing interval, infusion time, as well as distribution and excretion of the antimicrobial agent will influence the infecting organism at the site of infection.

- **R – Clinically Resistant:** level of antimicrobial susceptibility associated with a high likelihood of therapeutic failure.

The microbiology results are reported according to:

- Cheshire & Merseyside Antimicrobial Stewardship Group “Guidance for the Reporting of Sensitivity Results by Microbiology Laboratories” (November 2015)
- The Pan-Mersey “Antimicrobial Guide and Management of Common Infections in

Primary Care" (July 2017) where appropriate

- The Alder Hey "Antimicrobial Prescribing Guidelines"
- The current antibiotic prescribing for the individual patient (if appropriate).

Additional susceptibility results can be discussed with either the Microbiology or Infectious Diseases Consultants;

## 6. Antimicrobial assays

The biochemistry laboratory provides both the in-house and referral service for antibiotic, antifungal, and antiviral levels where required.

## 7. Referred tests

The laboratory refers a number of tests to external laboratories (details of these laboratories can be found at the end of the handbook).

## 8. Factors affecting the validity of results

- Microbiology tests can be affected by the concurrent use of antimicrobial agents; pathogens may be suppressed if samples are collected after antibiotics are started.
    - Please document clearly on the request form if samples are collected after antibiotics have been started; this is particularly important for meningitis cases.
    - In cases of suspected sepsis, prompt antimicrobials (within an hour) should be given according to National guidance; do not delay this for the collection of samples.
- The department undertakes duplicate testing of samples as a means of internal quality control and also participates in a number of external quality control schemes organised by UKHSA - UKNEQAS for Microbiology, Lab quality - Finland and INSTAND - Germany.
- Whilst internal and external quality assurance programmes are in operation to ensure accuracy and precision of results, occasionally random errors may occur and escape detection.
- The clinician is often best placed to detect such errors, if you doubt the validity of any result, it is vital that you contact the department at once so that we can investigate and re-test samples whenever possible.
- Certain factors may affect and possibly invalidate some test results, causing potential biological and analytical interference. For example, haemolysed blood samples, antibiotics and type of specimen tube used.
- Please give relevant clinical details at all times including details of recent travel abroad, current treatment etc. as this will ensure that samples are processed appropriately and reduce the need for additional test requests.

## 9. Specimen Containers

Please ensure that sterile containers are used when sending samples to the microbiology department and that the containers are labelled with the patient's full name, date of birth and the date of collection.

## 10. Labelling Requirements for Request Forms

Samples from General Practitioners and Walk-in-Centres must be accompanied by a hand written request form or a GP's letter. The form/letter should clearly state the following information for unequivocal identification of the patient and specimen:

- Patient name (in full – no abbreviations)
- GP name and address and Clinic name if applicable
- Date of Birth
- Sex
- Type of specimen
- Date and time specimen taken

**NB** It is **ESSENTIAL** that the laboratory knows the date on which a specimen is taken: processing delayed specimens can yield unhelpful or misleading results and they may be discarded (e.g. urine samples dated 2 days prior to day of receipt).

If patients are given a request form and asked to provide a specimen from home **they should be asked to write the date on which the specimen was collected on both the container and the form and to return the sample to the Alder Hey Out-Patients department and NOT the GP surgery.** The results of samples not returned to and processed at Alder Hey may not have interpretation from a paediatric microbiologist.

Also required:

- All relevant clinical details including any recent antibiotic treatment
- History of recent foreign travel, if applicable
- Risk status, if applicable
- Date of onset and duration of illness, particularly for serology
- Specify anatomical site from which "wound" specimens were taken.
- An indication of parental / legal guardian consent must be given for HIV testing, samples cannot be processed without consent in writing.

**If the laboratory cannot unequivocally identify the sample and match it to a form, then it will be discarded.**

## 10.1. Additional Test Requests

- Please telephone the laboratory before ordering additional tests to ensure that the sample is available and still suitable for examination.
- **Requests for additional tests on referred samples must be made to the microbiology department and not to the referral laboratory directly.**

The table below indicates how long samples are kept in the laboratory before they are discarded.

| Sample | Time Kept |
|---|---|
| Faeces – C&S | 2 weeks after primary culture |
| Faeces – *C. difficile* | 2 weeks after primary test, positive samples are stored for 3 months. |
| Respiratory samples for C&S | 2 weeks after primary culture |
| Swabs, fluids and aspirates | 2 weeks after primary culture |
| Urines | 1 week after primary culture |
| CSF Samples – routine | 2 weeks after primary culture at 4-8$^{O}$C then 12 months at -80$^{O}$C |
| Tissue | If there is any sample remaining after processing it will be stored for 1 month after primary culture. |
| Mycology samples | Not stored as samples are referred |
| Respiratory Virus PCR | 2 days |

Samples that are sent to referral laboratories are stored under local procedures and may not be available for additional tests.

Requests for additional tests will be accommodated if at all possible but in some instances samples are not suitable for additional analysis, these include:

| Initial request | Reason |
|---|---|
| MRSA Screen swabs | Swabs are cultured on selective media that may compromise the recovery of additional pathogens |
| Stool samples for C&S | Additional bacterial culture will only be performed within 3 days of receiving the sample due to potential overgrowth of normal flora. |
| Serology samples | The ability to add additional tests is dependent upon the test required and the initial volume of blood received. |

# 11. Labelling Requirements for Specimens

Samples received with a handwritten request form must be labelled in block capitals with:

- Full Name as registered on Meditech
- AH Number
- DOB
- Ward
- Specimen source
- Date and time of collection

Please Note: If the laboratory cannot unequivocally identify the sample and match it to a form, then it will be discarded. The laboratory will inform senders by means of an electronic or printed report when a specimen has been discarded for the above reasons

It is **ESSENTIAL** that the laboratory knows the date on which a specimen is taken: processing delayed specimens can yield unhelpful or misleading results, so they may be discarded (e.g. IGRA sample appropriately collected & transported to laboratory the same morning, but labelled with the Meditech order request date/time from the previous day).

**NB: If a Meditech sample label is accidentally printed at time of ordering rather than time of collection, it must be altered to the correct date/time of collection by the person who took the sample. A clear handwritten correction to the labels at time of collection will suffice.**

# 12. Transport of Clinical Specimens

GP Samples may be delivered to the outpatients department found next to the East Prescot Road entrance to the hospital.

## 12.1. Storage of Non-Urgent Samples

- Non-urgent samples should be refrigerated and sent to the department as soon as possible the next morning.

## 12.2. Packaging and Transport

- ALL specimens should be placed in a **separate** plastic bag and sealed.

## 12.3. Leaking or damaged samples

- The plastic transport bags, if properly sealed, are designed to contain accidental specimen leakage from the container.
- Most incidents of specimen leakage are due to the fact that neither the container nor the integral bag strips have been closed properly.
- Upon receipt of a sample whose integrity was compromised or which could have jeopardized the safety of the carrier or the general public the sender is contacted immediately. The sender will be informed of any measures that should eliminate recurrence; all incidents will be recorded on the Trust's recording system.
- Repeat specimens will be required from samples such as urine.

# 13. Standard Procedures for the Safe Collection of Specimens

These procedures concern all clinical staff who are qualified to collect diagnostic specimens from patients. **Staff must always follow aseptic techniques when handling blood, body fluids, excretions, or secretions,** even when these have not been specified as infectious.

## 13.1. Potential Hazards

All staff must be aware of the potential physical and infectious hazards, associated with the collection of samples for microbiological investigation.

- Follow all local procedures to protect personal safety, prevent injury and exposure to biological hazards.
- Follow all local procedures to reduce the risk to colleagues who are involved with the handling, transport and laboratory investigation of specimens.

## 13.2. Safety Precautions

- Staff collecting specimens must take care to prevent contaminating themselves, their environment, the external surfaces of the specimen containers, or the accompanying test request forms.
- If gross contamination of the hands with blood, faeces or other biological fluids is anticipated, then gloves should be worn. Hands should always be washed after taking specimens. If splashing into the eyes or onto mucous membranes is possible goggles should be worn.
- In addition, specimens should be collected aseptically, without allowing contamination by extraneous and, therefore, irrelevant micro-organisms.

Contaminated specimens can adversely affect the validity of many laboratory results. For example, the microbiological investigation of contaminated blood or other materials from sites, which are normally sterile, can commit patients to unwarranted courses of expensive and potentially toxic treatment.

All waste generated from obtaining a specimen should be disposed of according to local procedures.

Please disinfect the outside of any specimen containers if they are contaminated during sample collection.

## 14. Sepsis

Wherever possible blood cultures should be taken in all cases of suspected sepsis and before antibiotics are started. This may not always be possible, and antibiotics should not be unnecessarily delayed in septic patients if there are difficulties collecting cultures.

The NICE “Sepsis risk stratification tool: children and young people aged 12-17 in hospital”, “Sepsis risk stratification tool: children aged 5-11 years in hospital”, and “Sepsis risk stratification tool: children aged under 5 years in hospital” do not advise any additional routine microbiological investigations other than a venous blood culture.

See the NICE guideline NG51 “Sepsis: recognition, diagnosis and early management”.

https://www.nice.org.uk/guidance/ng51

## 15. Meningitis and Encephalitis

Meningitis is defined as inflammation of the meninges. Meningitis can be acute or chronic and can have both infective and non-infective causes. Encephalitis is an inflammatory process involving brain parenchyma. Over 100 causes have been associated with encephalitis; the majority are viral infections, but other infection and immune-mediated conditions (including post-infectious inflammatory processes) are possible. Over one third of cases have no identified aetiology. Viral encephalitis is usually acute and is often associated with some elements of meningitis (i.e. meningoencephalitis), although neck stiffness occurs in less than one in three cases.

Typical bacterial causes of meningitis include:

- *Streptococcus pneumoniae*
- *Neisseria meningitidis*
- *Haemophilus influenzae*
- *Escherichia coli*
- *Listeria monocytogenes*
- Group B *Streptococcus*

Typical viral agents of meningitis and encephalitis include:

- Herpes Simplex Virus
- Varicella Zoster Virus
- Enteroviruses (Throat swabs and faeces are additional appropriate sample types for consideration; detection of virus in these samples is suggestive, but not diagnostic, of the cause of illness. Blood for Enterovirus detection is unhelpful.)
- Parechovirus (Typically seen under the age of 3 in the immunocompetent; testing

outside of this age is not recommended. In the immunocompromised Parechovirus should be considered at all ages. Parechovirus PCR is included as standard in the CSF viral PCR tests used by the laboratory.)

HIV testing is recommended for all adults with meningoencephalitis; discussion with the infectious diseases team is strongly recommended if HIV infection is considered a possibility.

Clinical features, season and travel history provide important information and may affect the need for additional testing. Patients who are immunocompromised may have atypical clinical features due to an altered immune response or disseminated infection.



# 16. Respiratory Samples

Bronchiolitis is a common presentation, particularly during the winter months, for respiratory tract infections in young children. This is typically a viral infection, classically respiratory syncytial virus (RSV), and as such many of the pathogens can be diagnosed using respiratory PCR.

If Mycobacterial infection is suspected (e.g. TB) samples will be referred to Liverpool Clinical Laboratories, however the microbiology department strongly recommends discussing such patients with the Paediatric Infectious Diseases team.

*Bordetella pertussis* may be detected by culture, PCR, or serology. For samples sent from outside Alder Hey, serology is the preferred sample type.

## 16.1. Cystic Fibrosis

For patients seen in the community or in the Emergency Department, the cystic fibrosis team recommend that a respiratory sample is collected before starting antibiotics if possible. They also appreciate early contact regarding these patients as management can be complex.

The same sample types are processed from Cystic Fibrosis (CF) patients as for any other respiratory infection; however there are additional cultures routinely performed for specific CF-associated pathogens these include *Burkholderia* and other non-fermenting Gram-negative bacilli, rapid-growing mycobacterial species such as *Mycobacterium abscessus* and fungal culture to detect *Exophiala* and similar species.

Please include in the clinical details that the patient has cystic fibrosis.

### 16.2. Respiratory viral PCR

Respiratory PCR testing is only available for inpatients and children who attend A&E with respiratory symptoms and are likely to be admitted.

No repeat testing will be performed within a two week period.

This test now includes SARS CoV-2 , this test must not be used for asymptomatic screening – pre op, elective and non-elective admission and surveillance for SARS CoV-2 , a COVID-19 test should be requested

## 17. Urine Samples

Urine samples for **routine culture and microscopy** must be received in sterile containers, containers containing boric acid are accepted if any delay in delivery to the department is expected.

The best routine sample is a clean catch specimen.

**Schistosoma detection**: The sample must be received as fresh as possible, please include the appropriate clinical and travel history on the request form; samples may be referred to LSTM.

## 18. Urinary tract infections (UTI)

UTIs may present with nonspecific symptoms and signs, particularly in infants and young children.

### 18.1. Preterm infants

Clinical signs of UTI in preterm infants include:

- Feeding intolerance
- Apnoea and bradycardia
- Lethargy
- Tachypnea
- Abdominal distension
- Hypoxia with documented oxygen desaturation

### 18.2. Term infants

Signs and symptoms of UTI in neonates are typically nonspecific and can include lethargy, irritability, tachypnea, or cyanosis; neonates may appear acutely ill. Clinical signs include:

- Fever
- Failure to thrive
- Jaundice (typically conjugated hyperbilirubinemia related to cholestasis)
- Vomiting
- Loose stools

- Poor feeding

UTI may be the presenting manifestation that identifies a neonate with an underlying congenital anomaly of the kidney and urinary tract.

Neonatal urinary tract infection is associated with bacteraemia and congenital anomalies of the kidney and urinary tract. Upper renal tract infections may result in renal parenchymal scarring and chronic kidney disease. Neonates with UTI should be evaluated for associated systemic infection, and anatomic or functional abnormalities of the kidneys and urinary tract.

### 18.3. Younger children (up to 2 years of age)

Signs and symptoms UTI in children younger than two years include:

- History of previous UTI
- Temperature >40°C
- Suprapubic tenderness
- Fever >24 hours

Absence of another source for fever is not always helpful in diagnosing a UTI; likewise an identified alternate source of fever does not exclude UTI. Reports of foul-smelling urine or gastrointestinal symptoms are not typically reliable in diagnosis.

### 18.4. Older children

Symptoms of UTI in older children include:

- Fever
- Urinary symptoms (dysuria, urgency, frequency, incontinence, macroscopic haematuria)
- Abdominal and/or back pain
- New-onset urinary incontinence
- Suprapubic tenderness and costovertebral angle tenderness

The combination of fever, chills, and flank pain may suggest pyelonephritis.

### 18.5. Complicated versus uncomplicated (simple) infection

- Uncomplicated UTIs are limited to the lower urinary tract and may be typically seen in children older than two years with no underlying medical problems or anatomic or physiologic abnormalities.
- Complicated UTIs involve coexisting upper renal tract infection, multiple-drug resistant pathogens, or hosts with specific indication (e.g. anatomic or physiologic abnormality of the urinary tract.

It is important to indicate to the laboratory if a complicated infection is suspected as extended antibiotic sensitivity testing is performed.

Asymptomatic bacteriuria may occur in 1-3% of infants and preschool-age children, and ~1% of older children; the organisms are typically of low virulence and easily eliminated by antibiotics, however, in most case the asymptomatic bacteriuria resolves spontaneously without complication.

### 18.6. What urine sample should I collect?

The microbiology department recommend wherever possible that mid-stream or clean-catch urine be collected for the diagnosis of UTI. Alternately, samples from intermittent or newly introduced catheters are suggested.

National standards for the processing of urine samples state that catheters, bag urines, catheter tips and ureteric stents are not acceptable sample types; at Alder Hey we accept but strongly discourage bag urine samples.

#### 18.6.1. Mid-stream or clean catch urine

If a mid-stream urine can be obtained, the first part of the voided urine is discarded and, without interrupting the flow, the remaining urine is collected into an appropriate container. For a clean-catch specimen, the whole flow is collected and an aliquot sent to the laboratory.

- **Please ensure that the patient's peritoneal / genital area is physically clean before collecting a urine sample for culture.**
    - **For Girls**
      Clean from the front to the back and gently pat dry with clean dry sterile gauze.
    - **For Boys**
      Gently retract the foreskin and clean the entire surface with gauze soaked in the sterile water. Replace the foreskin and dry with clean sterile gauze.
- Catch the middle portion of the urine in a clean wide-mouth receptacle. Such a receptacle need not be sterile: any container, previously washed thoroughly with detergent and hot water and stored dry, is suitable.
- A sample of the middle portion of the urine must be poured into a 20ml universal container (white top). The pot should be labelled with the patient's details.

#### 18.6.2. CSU (catheter sample of urine) collection

Catheter samples may be obtained either from transient catheterisation or from indwelling catheters.

The specimen should be collected aseptically from a sample port in the catheter tubing or by aseptic aspiration of the tubing.

- For indwelling catheters, the distal end, or preferably the sampling port if present, must be disinfected with 70% isopropyl alcohol and urine aspirated with a sterile syringe.
- The urine must then be transferred to an appropriate container.
- The specimen should not be obtained from the collection bag.

Growth from indwelling catheters may not identify the genuine pathogen as bacterial biofilms develop on the catheter surface. Interpretation of specimens from long term-catheterised patients may be extremely difficult to impossible.



# 19. Eye, Ear, Nose, and Throat Samples

## 19.1. Eye Infections

### 19.1.1. Conjunctivitis

Infectious conjunctivitis is one of the causes of red or sticky eyes. Conjunctivitis may occur in association with infection of the eyelid (blepharoconjunctivitis), inflammation of the eye lid (blepharitis) or of the cornea (keratoconjunctivitis).

- An eye swab will typically be sufficient for conjunctival infections
- For neonatal infection consider sending a sample for gonococcal and Chlamydia PCR (*Neisseria gonorrhoeae* is a very sensitive organism and may not survive the culture process).
    - Please ensure that suspected cases of ophthalmia neonatorum are clearly indicated on the request so that specific culture plates can be set up.

### 19.1.2. Orbital cellulitis

Orbital cellulitis is the infection of orbital tissue resulting from trauma, surgery, or an extension of paranasal sinus infections. It is a serious infection and may cause blindness, septic thrombosis of the cavernous sinus or intracranial infections.

- Eye swabs are of limited value.
- Ideally send aspirates from the affected tissues if possible
- Blood cultures should be taken.

### 19.1.3. Collecting eye swabs

- Collect before antimicrobial therapy, where possible, and preferably before application of local anaesthetic or dye.
- Prior to collecting any samples for processing remove the exudate from the eye.

- Moisten the swab tip with normal saline to provide optimum collection for bacterial/viral/chlamydial detection.

**From infants:**

- Lay the child flat (on a bed or a parent's knee)
- Gently fold down lower eyelid and run swab across the inner surface rotating swab to ensure optimum specimen collection.

**From older children:**

- Sit or lay the patient with the head well supported, ask the patient to look up and gently pull down the lower lid exposing the conjunctiva.
- Gently sweep the swab stick along the lower eye from the inside out taking care not to touch the eyelids. Place swab immediately into the transport tube.

Unless otherwise stated, blue Transwabs should be used for bacterial culture, any available pus should be sampled as well as the lesion of interest.

#### 19.1.4. Chlamydia detection

- For chlamydial examination the cells from the inner canthus must be sampled.
- Separate samples must be collected into appropriate transport media for detection of viruses or chlamydiae.

### 19.2. Ear infections

#### 19.2.1. Otitis Externa

Infection of the external ear canal resembles skin and soft tissue infection elsewhere, however, as the canal is narrow foreign materials and fluid that enter can become trapped and cause irritation and maceration of the superficial tissue.

- A swab of the external ear canal is usually sufficient.

#### 19.2.2. Otitis Media

Middle ear infections may be caused by a number of bacterial species, often those found in the upper respiratory tract.

- An external ear swab is not useful unless there is perforation of the eardrum.

### 19.3. Nasal infections

#### 19.3.1. Sinusitis

The sinus cavities are usually sterile or may contain small numbers of bacteria that are continuously removed by the mucociliary system. Acute community acquired sinusitis is typically caused by upper respiratory tract bacteria, however viral infections are also an

important cause. Complications of sinusitis include orbital infection, and less commonly intracranial infection and osteomyelitis. Acute nosocomial sinusitis is often a complication of endotracheal intubation and mechanical ventilation, and often shows no clinical signs of infection. Nosocomial sinusitis is often polymicrobial.

- Superficial swabs are likely to be inadequate; scrapings or biopsy material are most likely to yield the diagnosis.
- Nose swabs are not a suitable sample type for sinusitis and should only be used for carriage detection.
- Specimens should be obtained by careful aspiration of the sinus (avoiding contamination by upper respiratory tract flora), and sent in a sterile universal container.

#### 19.3.2. Fungal Infection

Fungal infections are usually due to filamentous fungi, including *Aspergillus*, *Rhizopus* and *Mucor* species. In the immunocompromised, sinusitis caused by filamentous fungi may cause life-threatening infections and is often locally invasive. *Candida* and *Cryptococcus* species are also causes of infection in patients who are immunocompromised.

- Aggressive surgical debridement is often required in addition to systematic antifungal therapy and treatment of the underlying cause.
- Examination of tissue rather than pus is important in fungal sinusitis.
- Community-acquired chronic fungal sinusitis is a relatively common problem in some tropical and subtropical countries, e.g. in Africa and India, and imported cases may be encountered, therefore please provide an appropriate travel history where relevant (this is also important for laboratory safety as some of the potential fungal species need to be handled under higher safety precautions).

### 19.4. Throat infections

#### 19.4.1. Pharyngitis

Inflammation of the pharynx may be acute or chronic. Most cases are viral but it is difficult to differentiate between bacterial and viral pharyngitis on symptoms alone. The most common cause of bacterial pharyngitis is *Streptococcus pyogenes* (Group A *Streptococcus*). In children *S. pyogenes* carriage (not infection) rates have been reported as 20% - 30%. Groups C and G streptococci may also cause pharyngitis. Fungal infections may be seen in the immunocompromised, or in patients receiving antibiotics.

- A throat swab should be considered for culture.

#### 19.4.2. Diphtheria

Please indicate in the clinical details if any of the following are present:

- Membranous or pseudomembranous pharyngitis/tonsillitis
- Contact with a confirmed case of diphtheria within the last 10 days
- Travel to a diphtheria high risk area (Africa, South East Asia, South America) within the last 10 days
- Contact with someone who has been to a high risk area within the last 10 days
- Contact with any animals (e.g. household pets, farm, petting zoo) within the last 10 days
- Recent consumption of any type of unpasteurised milk or dairy products

For these patients a naso-pharyngeal swab may be considered if a throat swab is unobtainable for any reason.



#### 19.4.3. Epiglottitis

Inflammation of the epiglottis commonly affects children and is associated with fever, hoarseness of voice, stridor and difficulty in swallowing. Acute epiglottitis in young children is a rapidly progressive inflammation of the epiglottis and surrounding tissues and may result in complete airways obstruction.

- Because trauma from the swab may precipitate obstruction, throat swabs are contraindicated in cases of suspected acute epiglottitis.
- Blood cultures should be taken in all cases of suspected epiglottitis.

#### 19.4.4. Tonsillitis

Inflammation of the tonsils is usually due to a viral infection but may also be bacterial. It is a common type of infection in children, and symptoms include a sore throat that can feel worse when swallowing, fever, coughing and headache.

- A throat swab can be sent for culture.

#### 19.4.5. Quinsy

Quinsy (peritonsillar abscess) is a rare acute infection usually on one side of the throat only, with the swelling behind the tonsil near the back of the roof of the mouth. Symptoms are similar to that of tonsillitis, including dribbling, generally feeling unwell and neck swelling because of the abscess.

- Pus may be aspirated from the abscess and if so, should be sent in a sterile universal container.
- If pus is unobtainable send a throat swab for culture.
- Blood culture should be considered for severe infection.

#### 19.4.6. Laryngitis

Acute inflammation of the larynx may be caused by a viral infection (such as a cold), or voice strain or occasionally by bacterial infection. This eases without treatment within a week. This is known as acute laryngitis.

- A throat swab should be considered for culture.

# 20. Wound Swabs

## 20.1. Choosing between swabs, tissues and fluids

In general, the more sample that can be sent to microbiology, the more the laboratory can do. Therefore, where possible, send pus/fluid samples or tissue samples in preference to swabs.

#### 20.1.1. Collecting wound swabs

- Check the expiry date on the swab
- Decontaminate the skin to remove as much of the superficial flora.
- If taking a swab from another surface (e.g. abscess cavity at incision and drainage) superficial decontamination may not be indicated.
- Taking a Transwab (blue top), remove the swab and gently but firmly rotate it on the surface directly where infection is suspected.
- Do not take swabs from slough or necrotic tissue.
- Place the swab into the transport medium.
- If pus is present send as much as possible in a sterile universal container.

## 20.2. Superficial skin infections

Skin infections can be categorised typically as:

- Cellulitis – a diffuse spreading infection involving the deeper layers of the skin and subcutaneous tissues
- Erysipelas – a diffuse spreading infection involving the upper dermis and superficial lymphatic system.
- Impetigo – a superficial, intra-epidermal infection producing erythematous lesions that may be bullous or nonbullous[6].
- Paronychia – a superficial infection of the nail fold occurring as either an acute or chronic condition.
- Folliculitis – infection and inflammation of a hair follicle.
- Ecthyma gangrenosum – a focal skin lesion characterised by haemorrhage, necrosis and surrounding erythema.

Superficial swabs of intact skin are often unrewarding; consider sending swabs for suspected surgical site infections or where the skin has broken down. It may also be appropriate to consider superficial fungal infection.

#### 20.2.1. Necrotising skin and soft tissue infections

Necrotising skin and soft tissue infections are typically described as gangrene (e.g. gas gangrene, Fournier's gangrene) or necrotising fasciitis, depending on the clinical features and the tissue planes involved. In cases of necrotising infection swabs are not the appropriate samples to send; these conditions require urgent surgical intervention, as well as antimicrobial therapy.



### 20.3. Ulcers

A skin ulcer is a lesion of the skin with loss of the skin integrity, which can extend from the epidermis down to deeper layers. All ulcers are invariably colonised by a polymicrobial flora and microbiology samples should be taken only if a clinical diagnosis of infection has been made.

- When swabs are taken from infected ulcers, they should be taken after cleansing and debridement to reduce the presence of superficial colonising flora.

#### 20.3.1. Burns

Patients suffering from severe burns are at a higher risk of both local and systemic infection. Gram negative organisms cause the most severe infections; fungal infections on the other hand can spread quickly, but are more easily treated. A definitive diagnosis can be difficult to obtain.

- Skin swabs should be collected as from ulcers (see above).

### 20.4. Bite wounds and contact with animals

Bite wounds can become contaminated by oral flora and normal human skin flora. Most bites are due to cats and dogs, but some are due to other pets (including reptiles, rodents and birds), domesticated animals (including horses, sheep etc.) wild animals or other humans.

- Skin swabs should be collected from broken skin.
- If pus is present a sample of pus in a sterile universal is preferred.
- Some organisms associated with animals may not grow from wound swabs – consider blood cultures and joint aspirates as appropriate, e.g.:
    - *Capnocytophaga canimorsus* is associated with dog bites and causes septicaemia. This organism is usually isolated only from blood cultures.

- *Streptobacillus moniliformis* is associated with rat bites and diagnosis is confirmed by culturing the organism from blood or joint fluid.

- Please note some pets carry Salmonella (e.g. snakes & turtles).
- *Mycobacterium marinum* is associated with fish tanks.

## 20.5. Necrotising skin and soft tissue infections

Necrotising skin and soft tissue infections are typically described as gangrene (e.g. gas gangrene, Fournier's gangrene) or necrotising fasciitis, depending on the clinical features and the tissue planes involved. In cases of necrotising infection swabs are not the appropriate samples to send; these conditions require urgent surgical intervention, as well as antimicrobial therapy.



## 20.6. Orthopaedic infections

### 20.6.1. Septic arthritis

Septic arthritis occurs either via haematogenous spread or directly from contiguous lesions. Purulent arthritis and synovitis may also be caused by sodium urate crystals (gout) and calcium pyrophosphate crystals (pseudo-gout). If required, samples can be referred for microscopic examination.

### 20.6.2. Bursitis

Inflammation of a bursa is often accompanied with prominent overlying cellulitis. The olecranon and prepatellar bursae are the most commonly affected sites. They are often subjected to repeated trauma.

## 20.7. Intra-abdominal sepsis

Intra-abdominal sepsis is infection occurring in the normally sterile peritoneal cavity, and covers primary and secondary peritonitis, as well as intra-abdominal abscesses.

### 20.7.1. Peritonitis

Spontaneous primary bacterial peritonitis accounts for <1% of bacterial peritonitis and is seen most frequently seen in children, and particularly those with nephrotic syndrome.

Chronic peritonitis may develop as a result of abscess formation and persist for weeks or months unless drained. Chronic infection may also be caused by *M. tuberculosis*. Tuberculous peritonitis is a rare disease in the UK but is more common elsewhere. If TB is suspected then discussion with the infectious diseases team is strongly recommended.

#### 20.7.2. CAPD Peritonitis

Diagnosis is usually based on the presence of at least two of: cloudy dialysate effluent, symptoms of peritonitis, positive culture and/or Gram stain of peritoneal fluid.

If routine cultures are negative and abnormal dialysate findings persist after treatment of presumed or documented bacterial peritonitis, consider sending samples for mycobacterial culture.

#### 20.7.3. Solid organ abscess (liver, spleen, pancreas)

Pyogenic liver abscesses often present as multiple abscesses, and can be a due to, biliary tract disease, haematogenous spread from another focus, surgery, or trauma. Pancreatic abscesses are potential complications of acute pancreatitis, while splenic abscesses are typically due to haematogenous spread from another focus or trauma.

##### *20.7.3.1. Parasitic liver abscesses*

These arise typically as a result of the spread of *Entamoeba histolytica* via the portal vein from the large bowel which is the primary site of infection. Hydatid (tapeworm) cysts may also occur as fluid-filled lesions in the liver, and cysts may become super-infected with gut flora and progress to abscess formation. If parasitic infection is suspected then discussion with the infectious diseases team is strongly recommended.

#### 20.7.4. Subphrenic abscess

Subphrenic abscesses occur immediately below the diaphragm, often as a complication of gastrointestinal pathology (e.g. perforation) or trauma. Subphrenic abscesses are typically caused by mixed infections from the normal gastrointestinal flora.

### 20.8. Retroperitoneal collections

#### 20.8.1. Psoas abscess

Psoas abscesses may be seen as complications to gastrointestinal pathology (e.g. appendicitis), spinal infections (e.g. osteomyelitis, discitis), bacteraemia or perinephric abscesses.

#### 20.8.2. Renal abscess

Renal abscesses are typically caused by Gram negative bacilli and result from ascending urinary tract infection, pyelonephritis, renal calculi or bacteraemia. Pyuria may be present, but urine culture is usually negative.

#### 20.8.3. Perinephric abscess

Perinephric abscesses are an uncommon complication of UTI, which usually affects patients with one or more anatomical or physiological abnormalities. The abscess may be confined to the perinephric space or extend into adjacent structures. Pyuria, with or without positive culture, is normally seen on examination of urine.

# 21. Pericarditis

Most pericardial effusions are small in volume and are sterile. Infectious pericarditis can be separated into three groups:

1. purulent, which are caused by bacteria and is fatal if untreated.
2. benign, either due to viruses or post pericardiotomy syndrome
3. hypersensitivity or post-infectious

# 22. Respiratory Tract infections

## 22.1. Pleural effusion

Pleural effusions may be infective, e.g., as the result of pneumonia or the direct spread of infection into the pleural cavity from tuberculosis, or sterile, e.g., secondary to chronic heart failure, malignancy, or uraemia.

## 22.2. Empyema

Empyema thoracis most often occurs as a complication of either pneumonia or lung abscess. The most common cause is *S. pneumoniae*, however any organism may be isolated. In the immunocompromised there are a number or unusual infections that may present with empyema; please ensure that any immune compromise is noted in the clinical details.

## 22.3. Lung abscess

Lung abscesses involve the destruction of lung parenchyma and present on chest radiographs as large cavities often exhibiting air-fluid levels.

- If TB is suspected then discussion with the infectious diseases team is strongly recommended.
- If there is a history of foreign travel or immunocompromised/suppression please provide full details. Such patients may have unusual causes of lung abscess which may require additional cultures to be performed.

# 23. Head and neck abscesses

## 23.1. Brain abscess

Brain abscesses are serious and life-threatening, and as such should be referred to the neurosurgical team as soon as possible.

Treatment of brain abscesses involves the drainage of pus and appropriate antimicrobial therapy.

- If TB is suspected then discussion with the infectious diseases team is strongly recommended.
- If there is a history of foreign travel or immunocompromised/suppression please provide full details. Such patients may have unusual causes of lung abscess which may require additional cultures to be performed.

## 23.2. Dental abscess

- Aspiration of dental abscesses may be taken, where possible, to identify the infecting organism(s).
- In cases of intraosseal abscess, swabs can be useful, but only if taken from a disinfected site.

# 24. Faeces Samples

## 24.1. Community-associated gastroenteritis

Please notify the laboratory if there is a history of foreign travel as additional cultures may be required.

Routine faeces culture detects the typical causes of food poisoning. The laboratory's culture procedure is in line with Public Health England's protocol, for this reason bacterial culture will not be undertaken if a child has been in hospital for three days or more.

When collecting a specimen of faeces it should be obtained in a convenient container (potty, bedpan or nappy) and transferred into a blue Fecon™ container with a plastic spoon attached. The laboratory requires a "grape sized" sample for each procedure requested.

All faecal samples submitted from children with gastroenteritis will be subject to the following investigations:

- Routine microscopy for parasites*
- Routine bacterial culture for *Salmonella*, *Shigella*, *Campylobacter* and *E. coli* O157
- Routine virology for Enteric viruses (referral test) will be restricted to children under 5

years of age.

* If parasitic infection is seriously considered three stool samples are required on alternate days. (Please note stored samples are not suitable for some parasitic investigations.)

Additional bacterial culture for *Vibrio cholerae* and *Yersinia enterocolitica* will be undertaken if clinical details suggestive e.g. for *Vibrio* sp. recent travel to an endemic area (Asia, Africa or Latin America); and mesenteric adenitis for *Yersinia* sp.

### 24.2. Viral gastroenteritis

Enteric virus PCR is available following discussion with a consultant microbiologist. Enteric virus PCR to be performed on all oncology patients and children under 5 years of age



### 24.3. Clostridium difficile

C. difficile testing will be undertaken on request only on children over two years of age.

- Samples will only be accepted for *Clostridium difficile* toxin (CDT) detection from Children over two years of age **and** if the sample takes the shape of the container.
- Samples from CDT positive patients will not be re-tested within 28 days unless approved by the consultant microbiologist.
- Rectal swabs are not accepted as a substitute for faeces for the above tests.

### 24.4. Threadworm detection

Cellotape collection kits are available from the department for the detection of *Enterobius vermicularis* (threadworm).

- The female worm emerges from the anus at night to lay its eggs so diagnosis of threadworm infection is by anal sampling in the morning BEFORE the patient has bathed and in girls before urination if possible.
- Ova are seldom seen in faeces therefore stool samples are not suitable for the detection of threadworm.
- The worm can sometimes be persuaded to emerge to lay eggs by wrapping the child in a blanket and keeping still for around 30 minutes. Negative results obtained by this method are not totally reliable.

Full instructions for sample collection are included with the cellotape kits.

### 24.5. *Helicobacter* antigen detection

- It is recommended that the initial diagnosis of paediatric *H. pylori* infection is based on a positive histopathology plus a positive rapid urease test or a positive biopsy culture.
- The detection of *H. pylori* antigen in stool is a test of eradication.

- A 'test and treat' strategy is NOT recommended in children.
- See *Journal of Paediatric Gastroenterology and Nutrition* 2011;53:230-43

# 25. Mycobacterial investigations

Please refer patients to hospital if mycobacterial infection is suspected.
Specimens collected for the diagnosis of mycobacterial infection should be taken (whenever possible) before anti-tubercular treatment is started. Please note 'Other' antimicrobials may also have significant anti-mycobacterial activity, notably fluoroquinolones and macrolides.

## 25.1. Interferon Gamma Release Assays (IGRA)

The T-Spot TB test and Quantiferon TB test detect both active tuberculosis (TB) in patients who are not yet showing symptoms and latent TB in contacts during an outbreak situation or immunocompromised patients before reactivation occurs.

- The sample must be received in the laboratory before 11am on the agreed date, and must have the correct day & time of collection on the specimen label.
- IGRA tests are not accepted on Fridays, weekends and Bank Holiday Mondays or any day before a bank holiday.

# 26. Surveillance and screening for multi-resistant Gram-negative bacteria and Vancomycin resistant Enterococci (VRE)

These cultures are performed on inpatients only and are to detect the carriage of multi-resistant bacteria in the gastrointestinal tract. The preferred specimen type is faeces (or the equivalent, e.g. colostomy output). VRE screening is only performed on patients on 1B

Throat swabs are processed from ventilated patients and are also screened for staphylococcal and beta-haemolytic streptococcal carriage.
Rapid CPE PCR testing is available for patients who fit the PHE guidelines for screening, this is an inpatient test only.

## 26.1. What do all these abbreviations mean?

When reporting resistance mechanisms detected in Gram-negative bacteria, the following terms and abbreviations are used. Where a resistance mechanism has been identified, the comments include brief details of the appropriate infection control procedures to be followed.

### 26.1.1. AmpC

AmpC enzymes affect penicillin and first and second generation cephalosporins, and lead to resistance to agents such as amoxicillin and cefalexin. They are found on the chromosome in a number of species (e.g. *Enterobacter* and *Citrobacter* species), but can also be found on mobile pieces of DNA called plasmids which can be passed between species. Third generation cephalosporins (e.g. cefotaxime) are weak inducers and poor targets for AmpC enzymes, however the development of resistance is a possibility; these agents should therefore be used with caution in infection caused by AmpC-expressing isolates.

### 26.1.2. ESBL

Extended-spectrum beta-lactamase (ESBL) enzymes have evolved from a number of bacterial beta-lactamase enzymes. Different enzymes affect different antibiotics, however the microbiology department has opted to report ESBL-expressing isolates at resistant to cephalosporins as an antibiotic class on the reports. ESBL enzymes are commonly carried on plasmids and are therefore transferrable between species; these plasmids now often carry resistance to other agents such as aminoglycosides and quinolones.

### 26.1.3. CRE

Carbapenem-resistant Enterobacteriaceae (CRE) include species such as *Enterobacter*, *Klebsiella*, and *Citrobacter* that have developed resistance to at least one carbapenem antibiotic (typically ertapenem). ‘This resistance is usually due to the expression of either an AmpC or ESBL enzyme together with mutations that cause changes to channels in the cell wall. A CRE is always resistant to a carbapenem but does not carry an enzyme that specifically targets carbapenem agents (compare with CPE isolates below).

### 26.1.4. CPE

Carbapenemase-producing Enterobacteriaceae (CRE) include species such as *Enterobacter*, *Klebsiella*, and *Citrobacter* that are expressing an enzyme that breaks down carbapenem antibiotics. Carbapenemase enzymes may be carried on plasmids and be both transferrable and associated with additional resistances. Because different carbapenemase enzymes show different degrees of activity, a CPE is not always resistant to a carbapenem (compare with CRE isolates above).

### 26.1.5. MBL

Metallo-beta-lactamase (MBL) expression leads to resistance to anti-pseudomonal penicillins (e.g. piperacillin) and cephalosporins (e.g. ceftazidime) as well as carbapenems in *Pseudomonas* and *Acinetobacter* species. MBL enzymes may be carried on plasmids and be transferrable between species.

#### 26.1.6. OXA

Oxacillinase (OXA) enzyme expression is typically due to chromosomal genes in *Acinetobacter* species (unlike in *Enterobacteriaceae*) and lead to resistance to carbapenems.

#### 26.1.7 VRE

Vancomycin resistant Enterococci – Enterococci mainly Enterococci faecalis and faecium can be resistant to vancomycin due to the presence of plasmids. VanA and VanB type account for most significant infections in clinical settings

# 27. Mycology Investigations

## 27.1. Dermatophyte infection

Skin, hair and nails may be examined for superficial infections by fungi.

Dermatophyte infections include:

- Tinea barbae – a mild to severe pustular folliculitis of the beard which can be misidentified as a Staphylococcus aureus infection
- Tinea capitis – infection of the scalp which can range from mild scaling lesions to a highly inflammatory reaction with folliculitis, scarring and alopecia. A history of animal contact or a travel history can be helpful in identifying the causative agent.
- Tinea corporis – “ringworm” of the body and may involve the trunk, shoulders and limbs. Infection may range from mild to severe, commonly presenting as annular scaly lesions with sharply defined, raised, erythematous vesicular edges.
- Tinea cruris – infections of groin, perianal and perineal sites are the most common in adult males. Lesions are erythematous and covered with thin, dry scales and may have a raised, defined border and small vesicles.
- Tinea manuum – usually presents as a diffuse hyperkeratosis affecting the palms and interdigital areas of the hands are affected. Hands are also a likely site for infection with zoophilic or geophilic dermatophytes particularly if the lesions are inflammatory, and involvement can spread to other body sites by contiguous spread and scratching.
- Tinea pedis (athlete’s foot) – toe webs and soles of the feet are most commonly affected; particularly the spaces between the fourth and fifth toes. Alternately a chronic, squamous, hyperkeratotic infection covering the pink areas of the soles, heels and side of feet may be seen.
- Tinea unguium / onychomycosis – fungal infection of the nail.
- Pityriasis versicolor (tinea versicolor) – infection of the stratum corneum by lipophilic yeasts of the Malassezia genus.

## 27.2. How should I collect samples from superficial fungal infections?

### 27.2.1. Skin

Patients' skin and nails can be swabbed with 70% alcohol prior to collection of the specimen, this is especially important if creams, lotions or powders have been applied. The edges of skin lesions yield the greatest quantities of viable fungus. Lesions should be scraped with a blunt scalpel blade. Dermapak™ collection kits are available from the department or a sterile universal is also suitable.

### 27.2.2. Nails

Good nail samples are difficult to obtain. It should be specified whether the sample is from the fingernails or toenails. Material should be taken from any discoloured, dystrophic or brittle parts of the nail. The affected nail should be cut as far back as possible through the entire thickness and should include any crumbly material. Nail drills, scalpels and nail elevators may be helpful but must be sterilized between patients. If associated skin lesions are present, samples from these are likely to be infected with the same organism and are more likely to give a positive culture. Send samples to the department in a sterile universal container.

### 27.2.3. Hair

Samples from the scalp should include skin scales and plucked hairs or hair stumps. Cut hairs are not suitable for direct examination as the infected area is usually close to the scalp surface. Plastic hairbrushes, scalp massage pads or plastic toothbrushes may be used to sample scalps for culture where there is little obvious scaling but such samples do not replace a scraping for direct examination. Send samples to the department in a sterile universal container.

## 27.3 Referral of skin, hair and nail specimens for microscopy and culture

27.3.1 Specimens of skin, hair and nail are currently referred for microscopy and culture to the Mycology Reference Centre at UHSM – Manchester Foundation Trust

# 28. Virology

The majority of serology tests are referred to Liverpool Clinical Laboratories at the Royal Liverpool Hospital; a small number of tests are sent to the Manchester Medical Microbiology Partnership or to UKHSA laboratories.
The external laboratory where a test is performed is shown on the Microbiology report.

Please contact the Alder Hey Laboratory (0151 252 5268) if you require any further information or if any result is required urgently; we will notify the referral laboratory in advance of any urgent request.



## 28.1. Choosing serology or PCR tests

Serology tests are usually required for immunocompetent individuals. The combination of IgM (for acute infection) and IgG (for immunity), can be used to determine if the infection was acquired recently or in the more distant past.

PCR testing can demonstrate the presence of genomic material (DNA or RNA) from a pathogen, but this does not necessarily indicate an acute (primary) infection. Viral genomic material can be detected in immunocompetent individuals with no associated disease as a consequence of prolonged virus replication after acute infection, virus reactivation from latency (secondary infection) or reinfection.

In immunocompetent non-neonates, it is not appropriate to request both viral serology and PCR at the same time. For example, if screening for acute hepatitis B virus infection, by the time of presentation with hepatitis the infection is best detected by virus specific serology tests for antigens and antibodies (HBsAg and HBc antibodies). PCR tests may be appropriate following confirmation of infection to demonstrate (and quantify) ongoing viral replication (viral load tests) in chronic infections.

Serology tests are usually poor tests for the immunocompromised (both iatrogenic or disease-related) and neonates (where maternal IgG will be present) because these individuals do not develop good antibody responses. Therefore PCR tests are generally required to both diagnose virus infection and to follow the virological response of any antiviral treatment given.

### 28.2. Viral loads

Viral Load tests should not be requested unless the patient is known to be infected and you are monitoring the response to treatment. They are not appropriate tests to request for diagnosis.

To protect the integrity of the sample the specimens should be received in the laboratory before midday on the same day as they are taken.

## 29. Molecular (PCR) Tests

There are many molecular tests that can be performed on appropriate samples.
Please note inappropriate tests may be stopped either at the Alder Hey laboratory or at the receiving laboratory.

Individual test requests can be discussed with the consultant medical microbiologists. Please note that some tests are only appropriate on specific sample types, e.g. VZ PCR is appropriate on vesicle fluids (for the diagnosis of chickenpox or shingles) or on CSF, but is not an appropriate diagnostic test on blood, and hence there is no VZ PCR test option on EDTA blood samples.
Chlamydia/GC molecular tests – use Aptima collection system – orange container for swabs (not eye swabs) and yellow container for urine

## 30. Serological Tests

Please note the inappropriate tests may be stopped either at the Alder Hey laboratory or at the receiving laboratory.

Clinical details are important when requesting any serology tests, including date of exposure and onset of symptoms. This is particularly true for zoonotic and parasitic infection; without these details the samples may be cancelled. Paired samples may be required to assess change in antibody titre.

### 30.1. Zoonotic infection (e.g. Lyme disease) and imported diseases

Requests for zoonotic infection or imported pathogens are referred to the UKHSA Rare and Imported Diseases Laboratory at Porton Down.
Please ensure when requesting these tests that full clinical details are provided as the test may not be accepted by the referral laboratory if relevant details are missing.

# 31. Quality Control

## 31.1. External assessment

The microbiology department is accredited to United Kingdom Accreditation Service (UKAS) ISO 10189 standards.

## 31.2. External quality assurance

The microbiology laboratory is registered with the following external quality assurance schemes:

| Scheme | Clinical Applications Covered | Provider |
|---|---|---|
| General Bacteriology | Isolation of pathogens from simulated clinical material | UKNEQAS |
| Antimicrobial Susceptibility Testing | Determination of susceptibility to antibacterial agents | UKNEQAS |
| Clostridium difficile detection | Detection of *C. difficile* toxin | UKNEQAS |
| MRSA | Detection of MRSA | UKNEQAS |
| Viruses in CSF | Detection of virus | UKNEQAS |
| Faecal parasitology and teaching | Examination for the presence of parasites | UKNEQAS |
| Respiratory rapid: RSV | Detection of RSV | UKNEQAS |
| Molecular detection of respiratory viruses inc SARS CoV-2 | Detection of virus | UKNEQAS |
| SARS CoV-2 PCR | Detection of SARS CoV-2 virus | UKNEQAS |
| Antifungal Susceptibility Testing | Determination of susceptibility to antifungal agents | UKNEQAS |
| Viral Gastroenteritis | Detection of viruses in a simulated faeces specimen | UKNEQAS |
| Endoscopic Rinse Waters | Detection of *Pseudomonas* in a simulated water sample | FEPTU |
| Recreational and Surface Water | Swimming pool water contamination | FEPTU |
| *Helicobacter pylori* antigen detection | *H. pylori* antigen detection in faeces | Labquality |
| Gram stain – blood culture | Gram stain of simulated blood cultures | Labquality |

| Scheme | Clinical Applications Covered | Provider |
|---|---|---|
| Resistant Gram-negative bacilli | Detection of multi-drug resistance mechanisms | Labquality |
| Vancomycin resistant Enterococci (VRE) | Detection of VRE | Labquality |
| Streptococcal antibodies | Streptococcal ASOT and anti-DNase B detection | INSTAND eV |

- UKNEQAS is the UK National External Quality Assurance Scheme for microbiology operated by Public Health England.
- FEPTU are the Food and Environment Proficiency Testing Unit operated by Public Health England.
- Labquality are a Finnish provider of laboratory quality control materials.
- INSTAND eV is a German provider of laboratory quality control materials.

### 31.3. Internal quality assurance

- All test kits are verified for use before being used clinically.
- Antimicrobial susceptibility testing is subject to weekly control testing.
- There is an ongoing internal quality assurance scheme involving the duplicate testing of clinical samples, performed monthly.

### 31.4. Document Control

- All bench guides and standard operating procedures used in Microbiology are controlled and managed electronically using iPassport (Genial Genetics Ltd).
- Laboratory standard operating procedures are based on the Public Health England Standards for Microbiological Investigations.

## 32. Patient Confidentiality

- All Staff are aware of the importance of patient confidentiality; they are all required to complete the following Statuary and Mandatory training:
    - Information Governance
    - Safeguarding Level 1
    - Safeguarding Level 2
- All access to Meditech and email is controlled by secure passwords which must not be shared, staff must log off shared PC's when they leave or lock a personal PC if it is left unattended.

## 33. Requests for Results from External Agencies:

- Staff must ensure that the caller is genuine; they must take a number and telephone the caller back to confirm identity. If the caller refuses no result may be given.
- Any faxed results must be sent to a confidential secure fax to a named individual.
- Patient data may be sent by email internally between @alderhey.nhs.uk email addresses or externally between secure @nhs.net accounts.
- Personal information is not sent from Alder Hey accounts to external email accounts.

## 34. Complaints Procedure

- Please contact a consultant microbiologist or the laboratory Lead Biomedical Scientist if you have any concerns or cause for complaint regarding any aspect of the service provided by this department.
- We will endeavour to act on any concerns raised and will inform you of any actions taken.
- If appropriate, an incident will be logged on the Trust Ulysses system.

# 35. List of Referral Laboratories

The Microbiology Department refers work to the following CPA or UKAS accredited laboratories.

| Abbreviation | Laboratory | Address |
|---|---|---|
| GOSH | Department of Microbiology, Virology and Infection Control | Level 4 Camelia Botnar Laboratories<br>Great Ormond Street Hospital NHS Foundation Trust<br>Great Ormond Street<br>London<br>WC1N 3JH |
| LCL | Liverpool Clinical Laboratories<br>• Microbiology Department<br>• Immunology Department<br>• Virology Department | Royal Liverpool and Broadgreen University Hospitals NHS Trust<br>Duncan Building<br>Prescot Street<br>Liverpool<br>L7 8XP |
| LHTD | Department of Parasitology | The Hospital for Tropical Diseases<br>Mortimer Market<br>LONDON<br>WC15 6AU |
| LSTM | The Diagnostic Laboratory | Liverpool School of Tropical Medicine<br>Pembroke Place<br>Liverpool<br>L3 5QA |
| MRI | Molecular Diagnostic Laboratory<br><br>Meningococcal Reference Unit<br><br>Vaccine Evaluation Unit | Manchester Medical Microbiology Partnership<br>PO Box 209<br>Clinical Sciences Building<br>Manchester Royal Infirmary<br>Oxford Road<br>Manchester<br>M13 9WZ |
| UKHSA Birmingham | National Mycobacteria Reference Service | Public Health Laboratory Birmingham<br>Heart of England NHS Foundation Trust<br>Bordesley Green East<br>Birmingham<br>B9 5SS |
| UKHSA, Colindale | Antibiotic Resistance and Healthcare Associated Infections (AMRHAI)<br>Respiratory and Vaccine Preventable Bacteria Reference Unit (RVPBRU)<br>Gastrointestinal Bacteria Reference Unit (GBRU)<br>Sexual Transmitted Bacteria Reference Unit (STBRU)<br>Virus Reference Department (VRD) | Centre for Infection<br>Public Health England<br>61 Colindale Avenue<br>London<br>NW9 5EQ |

| Abbreviation | Laboratory | Address |
|---|---|---|
| UKHSA, Porton | Rare and Imported Pathogens Laboratory | Rare and Imported Pathogens Laboratory<br>Porton Down<br>Salisbury<br>Wiltshire<br>SP4 0JG |
| SNGH | Department of Immunology | Sheffield Northern General Hospital<br>PO Box 894,<br>SHEFFIELD,<br>S5 7YT |
| UHSM, MRC | University Hospital of South Manchester | Regional Mycology Laboratory<br>2nd Floor Laboratory,<br>Education and Research Centre<br>Wythenshawe Hospital<br>Southmoor Road<br>Manchester.<br>M23 9LT |

Bartonella serology tests are sent to Aix-Marseille Université, Marseille, France.

## 36. Acknowledgments

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