Microbiology User Information

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Contents

1. Summary of revisions

Version	Summary of change
2.15	Update to new template
2.16	Update TAT include item for delayed results
2.17	1 year revision
2.18	More information on faecal sample collection, references to clinical scientist removed, general update in line with ISO15189. TAT revised
2.19	Transport of infectious substances MP PM 010 now obsolete so reference removed Use of Boric acid universal added Procalcitonin added Revised TAT's Reference lab for Rubella avidity amended
2.20	Inserted reference to lab confidentiality policy & complaints procedure TAT's updated to 2019 UoM section added
2.21	TAT'S updated to reflect 2018 data Mycoplasma serology section updated to reflect serology not done on patients >25yrs old Chlamydia test no longer offered singly all will be CTGC, CPE screening, opening times, tropism, M gen., remove resp serology and Mycoplasma PCR to NGH, chlamydia group serology-Bristol
2.22	Use of Chlamydia UPT tubes removed Culture for Helicobacter pylori from biopsies added as a referral test Urines moved to Barnsley
2.23	Covid info: updated to include Panther, Qiastat RPCR, and changes to CT/GC/HSV, EUCAST susceptibility testing.
2.24	Rejecting precious samples, lab times, consent waiver, M gen, document number
2.25	Remove mycoplasma serology Add hospital location Add T vag to repertoire Disposal of clinical materials Patient consent Remove ref to BD Viper
2.26	CSF PCR in house; Labelling policy update
2.27	Times, phone numbers, TAT, Covid, availability of tests

2. Contact numbers

If you have any queries, please do not hesitate to contact the laboratory for help and advice.

Contact telephone numbers

Bacteriology Laboratory:	01709 424242
Virology Laboratory:	01709 425237
Consultant Microbiologist:	01709 424742
Specialist Registrar	01709 427712

Clinical advice dect phone	01709 428280
Microbiology and Infection control secretary	01709 424743
Microbiology Manager:	01709 426957
Senior BMS office:	01709 427711

3. Purpose

The Microbiology Department comprises of Bacteriology, parasitology, mycology, molecular diagnostics and Virology/Serology, and offers a comprehensive range of diagnostic services including an on-call service for emergency samples outside routine hours.
The department is fully computerised with authorised reports accessible to Hospital & community staff via the ICE & EPR systems. The department is approved for training by the Institute of Biomedical Sciences, and is accredited by UKAS to ISO15189.
The microbiology laboratory can help to:

- Provide or confirm a diagnosis.
- Suggest appropriate antibiotics.
- Monitor response to treatment.

Failure to investigate may lead to:

- An increased use of antibiotics causing possible harm to patients.
- An increasing reliance on expensive broad-spectrum agents.
- Increasing antibiotic resistance in the community (and concomitant lack of knowledge of this).
- Difficulty in establishing a diagnosis when patients have failed to respond to treatment.

Doctors requesting investigations should bear in mind Asher's catechism (British Medical Journal 1954; ii: 460).

1. Why do I request this test?
2. What will I look for in the result?
3. If I find what I'm looking for will it affect my diagnosis?
4. How will this investigation affect my management of this patient?
5. Will this investigation ultimately benefit the patient?

This will ensure that laboratory services are used in a cost effective manner.

4. Laboratory Location:

The Rotherham NHS Foundation Trust
Moorgate Road, Rotherham, S60 2UD.
Pathology is situated on A-level (Top floor) within the main hospital building. Follow the signs to Pathology from the main entrance, take the corridor opposite the lifts on level A, Pathology is the first on the left. Pathology general reception area is straight ahead through the double doors. Microbiology is located within the secure pathology department, samples can be left in the general reception, but if needing access to the microbiology section, please ask at reception.

5. Laboratory opening times:

Normal service:
Monday - Friday 08:30- 1700 hrs
Saturday and Sunday - limited service between 09:00 and 17:00 hrs, on-call only 17:00-09:00

All samples received within the normal working day are processed on receipt. Request forms that ask for an urgent response will be analysed accordingly and any provisional report (direct microscopy etc.) will be telephoned ASAP. As many of these results can be complex it is useful if the requesting physician supplies a contact number on the request form.

The Medical and Scientific Staff of the department are available on the telephone numbers listed in section 2 for results, advice/interpretation.
The Microbiology Medical staff provide an on-call service out of hours and are contactable via the hospital switchboard.

The BMS staff MAY operate an extended day (Monday – Friday), where one BMS is available in the laboratory for urgent samples until 21:00, after this time the on-call BMS can be contacted via the hospital switchboard**. As of 13/1/25, due to unprecedented staffing issues we are unable to offer a late duty service so the lab is open until 17:00 when on call starts**

6. Requesting tests

Requests for work should be made via the ordercomms system (ICE or Meditech) where possible, or in the event of ordercoms failure or unavailablilty please use the manual request form or the reverse of the ICE paper.

If completing a manual request it is essential to complete the request legibly and fully in ballpoint pen. Every form should include the clinician name and location together with the following minimum data set, which should be identical on both the form and the sample:

- Surname
- Forename
- At least one of the following:
    - NHS Number (Trust primary identifier)
    - Registration Number (default unique identifier if NHS number not available)
    - Date of Birth

Where no unique identifier is available, other demographic information, if available e.g. address, should be used in conjunction with the surname / forename and date of birth to confirm the patient's identity. Failure to provide this information may lead to delays in the results being returned or the sample being rejected.

The request form must be accompanied by the correctly labelled sample which should be transported to the laboratory contained in the special specimen bag which should be attached to the request form.

7. Data protection

Laboratory Medicine is committed to ensuring the confidentiality of all patient sensitive information.

All data and information acquired while providing the services of the laboratory is handled in strict accordance with the Trust Confidentiality Policy. This ensures data is managed in compliance with all relevant legal obligations, standards and guidelines and professional codes of conduct.

The Pathology Confidentiality Policy builds on the Trust's Confidentiality Policy in giving clear guidelines on the transmission of patients' Pathology results and reports.

See also: MPL-PP-007.

The following link has instructions for patient collected samples, consent, personal information

protection and the laboratory's complaints procedure.

http://www.therotherhamft.nhs.uk/Pathology/Pathology/"

Patient consent is implied from the request form for microbiology samples.

8. Complaints procedure

If you have any concerns about the services provided by the laboratory, please let us know using any of the contact options provided above.

Formal complaints can be made through the Trust Patient Experience Team
http://www.therotherhamft.nhs.uk/yourexperience/

See also the Laboratory Medicine website.

9. Specimen rejection

If samples or request forms are not labelled correctly then they cannot be accepted by the laboratory, and a result will be issued as not tested.

Some unrepeatable samples (e.g CSF, sterile fluids and blood cultures) are treated as precious samples and the sample will be tested even if inadequately labelled, however a comment will be added to the result that the specimen was unlabelled, and the sender should take responsibility for the validity of the result. The requestor may be asked to attend the laboratory to confirm the identity of any mislabelled precious sample and sign a precious sample form. Precious samples, which have to be rejected, will be telephoned to the appropriate department, including, but not exclusively: CSF, fluids, BAL, Covid screening swabs & blood cultures.

10. Routine investigations

Specimens for routine investigations should be collected as early in the day as possible to ensure that they arrive in the laboratory during normal working hours.

Any specimens sent from patients suspected or diagnosed of blood borne viral infections, must be identified by using the yellow "biohazard" sticker both on the specimen and on the request form.
Requests for extra tests

These should be telephoned to the laboratory as soon as possible and a manual request form **MUST** be sent to confirm the request for the extra test saying "sample already in lab".

Please note that Microbiology cannot accept requests for extra tests on blood samples that have been already tested in Blood Sciences due to possible cross contamination from Blood science analysers, so a new sample and form should be sent to Microbiology. If the patient is particularly difficult to bleed then please contact the laboratory to discuss if add on tests are required.

Samples submitted for bacterial culture are only suitable for re-testing for 48 hours, so if extra tests are needed after this time a fresh sample should be sent.

Blood specimens for serology are stored for 2 years, however extra tests should be requested in relation to the current clinical episode. If unsure, please seek advice from the Consultant Microbiologist.

Any other specimens discuss with Laboratory staff /Consultant Microbiologist.

11. Uncertainty of measurement

Where a measurement step is involved in producing a result the laboratory has determined uncertainty, these values can be supplied to users on demand.

All areas of analysis that could contribute to loss of quality of the results have been addressed and minimised where possible.

12. Sending specimens to the laboratory

Please also refer to the Transport of samples documents and information on the general pathology web page.

All fluids, eg CSF, pleural fluid, joint fluids and pus require culturing without delay. Specimens should preferably be taken during normal laboratory opening hours and sent immediately to the Department.

The Microbiology Department (extension 4242) and **NOT** general pathology reception should be warned of the arrival of urgent and important, unrepeatable specimens.
If taken outside routine laboratory hours the Microbiology "BMS on-call" **must** be contacted via Switchboard.

Do not use the pneumatic tube system for high risk specimens or any unrepeatable specimens such as CSF samples (in case of tube malfunction), but bring them to the pathology reception and alert staff there that it is an urgent sample.

In general, all specimens should reach the laboratory as soon as possible after being taken. Microorganisms may be susceptible to drying, heat or cold (particularly freezing). If a significant delay is unavoidable then the samples should be placed in a refrigerator. (N.B DO NOT place blood cultures once taken into the fridge, if unable to transport to the laboratory immediately then leave at Room temperature).

In specimens such as sputum and urine bacteria can multiply to significant levels so the validity of the result may be compromised.

Genital pathogens such as *Neisseria gonorrheae*, anaerobic bacteria and other fastidious organisms are particularly sensitive to delays before culturing.

All bacterial swabs should be placed in transport medium which prevents drying, maintains pH and excludes oxygen; and kept at room temperature until delivery to the laboratory, if a significant delay is unavoidable (e.g overnight) refrigeration is advisable.

Urine for culture should always be collected in either a plain white topped sterile universal container or a red topped boric acid urine container. Boric acid is a preservative and will prevent bacterial overgrowth so preserving the sample quality so should be used wherever possible. In the event a dip-strip test is required prior to sending to the laboratory a plain white top container will have to be used, in this instance please ensure that the sample is received in the laboratory ASAP after testing or refrigerate until delivery is possible.

Disposal of consumables or contaminated items, used in the taking of clinical samples must be disposed of according to local policy by the person/department who are responsible for taking and requesting the sample.

Instructions for use of Boric acid containers (Red Topped)

MUST ONLY BE USED FOR MICROBIOLOGY MICROSCOPY AND CULTURE

The red topped containers contain a small amount of white powder – do not discard this!
Boric acid makes sure the urine is preserved and suitable for testing for cells and bacteria that indicate a UTI, if the sample cannot be analysed within 4 hours of collection.

While the powder is non-hazardous it is a chemical and can cause skin irritation so avoid skin contact with the white powder, do not eat and keep out of reach of children.

If you accidentally get powder on your hands, wash immediately with plenty of soap and water.
If you accidentally get it in your eyes, then rinse immediately with plenty of water and attend A&E as soon as possible.
If the powder is eaten, then attend A&E immediately.

Please ensure the container is filled with urine to the fill line, under filling may have an effect on the bacterial growth and the sample will be rejected if less than half full.

If only a small sample (<10mls or half a container full) is available then please use a white topped container, this should be sent to the laboratory within 4 hours or stored in the fridge until transport to the laboratory is available if collected after the last pick up.

If boric acid is used the sample will be suitable for microbiological analysis for up to 48 hours without the need to store in the fridge.

Specimens of clotted blood (brown top “serum separation” tubes) are suitable for most serological tests. Refrigerate until delivery: **DO NOT** freeze

13. Serology tests performed in Virology/Microbiology

Please note that from 1/4/16 Rubella IgG will no longer be tested if requesting an 'antenatal screen'.

If Rubella IgG test is required a separate request will need to be made when using electronic ordering, or write it on the request form if using paper based requesting.

Mycoplasma pneumonia serology will only be performed on patients under 25yrs old, please send a green topped viral throat swab for Mycoplasma PCR on other patients.

Serology/ Virology tests – sample type and Turnaround times

SPECIMEN TYPE	TEST	COLLECTION	EXPECTED TURN AROUND TIMES (90% completed)	COMMENT	TESTED AT
Venous Blood	Gentamicin assay	4.9 ml clotted Gel sample(brown top)	Same day		Test performed in Blood Sciences
Venous Blood	Other antibiotic assay	4.9 ml clotted Gel sample(brown top)	Results telephoned or authorised as soon as possible. Full result 8-12 days	Contact Consultant Microbiologist for advice	Sheffield Blood Sciences Bristol Southmead hospital
Venous Blood	Antenatal screening (HIV, Hepatitis B, Syphilis)	4.9 ml clotted Gel sample(brown top)	4 days		In-house (Barnsley)
Venous Blood	Antistreptolysin O Anti DNAse B	4.9 ml clotted Gel sample(brown top)	10 days		**This test is no longer available**
Venous Blood	Bartonella serology	Test no longer available		please contact the consultant microbiologist	
Venous Blood	B.pertussis serology Bordetella PCR	4.9 ml clotted (brown top)	12 days 10 days		Colindale STH
Venous Blood	Borrelia serology	4.9 ml clotted Gel sample(brown top)	21 days		Porton Down
Venous Blood	Brucella serology	4.9 ml clotted Gel	12days		Porton Down

		sample(brown top)			
Venous Blood	Covid antibody	4.9 ml clotted Gel sample(brown top)	7 days	Not routinely available, please discuss with Consultant Microbiologist before requesting.	**This test is no longer available**
Venous blood	Coxiella serology	4.9 ml clotted Gel sample(brown top)	15 days		Colindale
Venous Blood	CMV serology CMV PCR	4.9 ml clotted Gel sample(brown top) EDTA sample	8 days		In-house Sheffield Virology
Venous Blood	EBV serology EBV PCR	4.9 ml clotted Gel sample(brown top) EDTA sample (red top)	8 days 10 days PCR		In house / Sheffield Sheffield virology or Colindale
Venous Blood	Enterovirus serlogy Enterovirus PCR	Viral throat swab or faeces	10 days	Reliable serology tests are not available for enterovirus infections. Please send a viral throat swab, viral swab from any lesions or faeces sample for PCR, an EDTA or CSF sample can be tested for PCR in suspected systemic infections	**Serology no longer available** Sheffield virology
Venous blood or CSF or other sample	Fungal 18S RNA	4.9 ml clotted Gel sample(brown top)	14 days		Micropathology
Venous Blood	Farmers Lung antibodies	4.9 ml clotted Gel sample(brown top)	25 days		Sheffield Immunology
Venous Blood	Helicobacter serology	4.9 ml clotted Gel sample(brown top)	7 days		**This test is no longer available**
Venous Blood	Hepatitis A, B & C serology for screening	4.9 ml clotted Gel sample(brown	Hep A IgG 8 days Hep A IgM 7	Please use biohazard stickers	Barnsley site

		top)	days HBsAg 3 days Hep B core 6 days Hep B s Ab 8 days Hep c Ab 4 days		
Venous Blood	Hepatitis markers for confirmation	4.9 ml clotted Gel sample (brown top)	8 days	Please us biohazard stickers	Sheffield Virology
Venous blood	Hepatitis C Genotyping viral load/PCR	4.9 ml clotted Gel sample(brown top)	25 days 10 days		Sheffield Virology
Venous blood	Hepatitis D	4.9 ml clotted Gel sample(brown top)	25 days	Hepatitis D will only be tested if patient a known hepatitis B positive.	Colindale
Venous blood	Hepatitis E Hep E PCR	4.9 ml clotted Gel sample(brown top)	10 days	Hepatitis E only tested if ALT>500 >100 if immunocompromised	Sheffield Virology
Venous Blood	HIV serology HIV Viral Load, resistance testing HIV Pro-Viral DNA, Tropism	4.9 ml clotted Gel sample (brown top)EDTA sample (Red top)	24hours –3 days confirmation 8-45 days	Please use biohazard stickers	Barnsley site/NGH/ Colindale/Lab 21
Venous Blood	Leptospira serology	4.9 ml clotted Gel sample(brown top)	15 days		Colindale
Venous blood	Measles serology and PCR	4.9 ml clotted Gel sample(brown top) Viral throat swab	6-16 days		In-house /Sheffield virology
Venous Blood	Mumps serology and PCR	4.9 ml clotted Gel sample(brown top) Viral throat swab	6-18 days		In-house Sheffield virology
Venous Blood	Meningococcal PCR and Pneumococcal PCR	2.7 ml EDTA sample (red top)	10 days		Manchester

Venous Blood	Mycoplasma serology	4.9 ml clotted Gel sample(brown top)	14 days	No longer available	**No longer available**
Venous Blood	Parvovirus serology Parvovirus PCR	4.9 ml clotted Gel sample(brown top) EDTA (red top)	10 days 25 days		In-house/ Micropathology/NGH/Colindale
Venous Blood	Procalcitonin	4.9 ml clotted Gel sample(brown top)	Same day		In-house
Venous Blood	Psittacosis serology	4.9 ml clotted Gel sample(brown top)	26 days		Sheffield Virology/Bristol PHE
Viral swab	Respiratory PCR	Viral swab	Same day or 8 days		In house or Sheffield Virology
Venous Blood	Rubella serology	4.9 ml clotted Gel sample (brown top)	IgG 4 days IgM 10 days		In-house/VRD Colindale
Venous blood	Systemic fungal screen/galactomanan	4.9 ml clotted Gel sample(brown top)	14 days		Bristol mycology
Venous blood	Schistosoma/ Strongoloides serology	4.9 ml clotted Gel sample(brown top)	25 days		London school tropical med
Venous Blood	Syphilis serology	4.9 ml clotted Gel sample(brown top)	3 days confirmation 12 days		Barnsley site/ Sheffield Virology
Venous Blood	TB – Quantiferon T spot	4.9ml Lithium heparin 2 x 2ml lithium heparin (paed)	7 days	Please note a minimum of 4 ml of blood is required 4 working days	In-house Oxford Immunotech
Venous Blood	Toxoplasma serology	4.9 ml clotted Gel sample(brown top)	7 days Confirmation 20 days		In-house /Swansea
Venous Blood	Varicella Zoster serology	4.9 ml clotted Gel sample(brown top)	2 days Confirmation 8 days		In-house Sheffield Virology
Venous	HSV serology	4.9 ml clotted			Manchester

Blood		gel sample (brown top)	14 days		
Venous Blood	Brucella, Q fever, Leptospira,	4.9 ml clotted Gel sample(brown top)	12 -15 days		Porton Down
Venous Blood	Arboviruses eg Dengue, Zika etc	4.9 ml clotted Gel sample(brown top)	50 days		Porton Down
Naso-pharngeal aspirate (NPA)	Detection of respiratory viruses	Sterile universal container	If sent to NGH 8 days In-house same day	Results are telephoned. Result is dependent on the quality of the sample.	In-house or Sheffield Virology as a contingency
CSF	Viral PCR	Sterile universal container	8 days	A viral throat swab and faeces sample may also be useful	Test now available in house or may be sent to: Micropathology or Sheffield Virology
Faeces	Viral PCR	Universal container with spoon(blue top)	8 days		Sheffield Virology
Faeces	Norovirus	Universal container with spoon(blue top)	2 days (can be performed urgently same day)		In-house
Urine	Virology	Sterile universal container	8 days		Sheffield Virology
Urine/genital swab	Mycoplasma genitallium detection /resistance Trichomona vaginalis PCR	Yellow aptima container for urine or Orange aptima swab		In house 7 days 14 days resistance micropathology	In house PCR Resistance markers Micropathology/Colindale
Viral swab	Virology-PCR(Herpes, VZV , Mumps)	Swab in viral transport media -collect from lab	8 days if sent away to NGH 24 hours if		Sheffield Virology HSV/VZV/Mumps Respiratory viruses In-house or

	Adenovirus PCR Chlamydia pneumonaie/ Mycoplasma pneumonaie Respiratory viruses Covid PCR		tested in house All Covid PCR		Sheffield Virology as a contingency In house (resp panel) or STH
Viral swab	HSV PCR (genital site only) HSV (non-genital site)	aptima swab Swab in viral transport media -collect from lab	4 days 8 days		In-house Sheffield Virology
Viral swab or CSF	Bacterial 16S RNA	Swab in viral transport media -collect from lab	14 days		Micropathology

Viral Investigation

Swabs in viral transport media (Green topped Virocult), are available from the laboratory. The swabs must be returned to the laboratory ASAP.

Vesicle fluid, nose, throat, genital sites

Use a viral swab + VTM, use the swab to collect material from the affected area, place the swab in the liquid media in the transport tube and break the swab off and re-cap the tube securely.

Store in the fridge if transport to the laboratory is to be delayed.

Urine:

Send the urine in a white topped plain sterile universal container

Any other specimen type – such as a biopsy, please contact the Consultant Microbiologist for advice.

Paired sera are not usually required now PCR is more widely available, we need to know enough clinical details to decide which viruses to screen for, and a date of onset to decide whether collection of a second serum is appropriate.

Only a limited range of viruses are tested for, and often serology is unhelpful.

Viral serology is helpful when a specific virus is suspected (eg rubella, CMV), or with particular clinical scenario's such as rash, flu-like illness, and respiratory tract infections.

Patients with vague or long-standing problems ("lassitude" etc) almost never produce diagnostic results.

Blood samples can also be sent to Virology for Immunity status checking, e.g after routine vaccinations for Hepatitis B and as part of the antenatal screening for infectious disease or when a pregnant lady has been in contact with Varicella zoster (VZV or chickenpox).

Please supply as much clinical information as possible to allow us to perform the correct tests, to interpret the result correctly and to add appropriate comments.

Please be aware that if serology/virology tests are requested a separate sample should be sent to Virology, the request should not be added onto a blood sample already sent to Blood Sciences except in exceptional circumstances e.g a difficult to bleed patient, in these instances please ring the virology laboratory to discuss.

14. Samples for bacteriology

Antimicrobial Susceptibility Testing

The results generated by this department match the criteria stipulated by EUCAST (the European Committee on Antimicrobial Susceptibility Testing). Methods for AST include, disc sensitivity, breakpoint sensitivity (Vitek). Micro-broth dilution and gradient strip MIC (minimum inhibitory concentration).
Reporting follows national nomenclature:
S = Susceptible
R = Resistant
I = High dose treatment regime may be appropriate please see comments.
Customised comments will be provided where appropriate.

Urines for routine microscopy culture and sensitivity

The Microscopy method used in the laboratory uses an analyser to estimate the number of white and red blood cells, and the number of bacteria in urine to help to distinguish infection from contamination. If there is evidence of infection, then a culture and sensitivity will be performed.

A mid-stream urine, clean catch, SPA or catheter specimen should be sent to the laboratory in a sterile universal container /red topped boric acid urine container.

Urines received in inappropriate containers will not be processed.

Urines in plain white topped containers should be received within 1-2 hours of collection. If this is not possible then refrigeration at 4°C for up to 24 hours is possible for most specimens without much change in bacterial count, but the white cells may become unrecognisable.

Red blood cells may lyse in dilute urine shortly after the specimen being taken, an on-site "dip-stick" test will give an indication of the presence of blood.

Red top universals contain Boric acid as a preservative.
Please ensure the container is filled with urine to the fill line, under filling may have an effect on the bacterial growth. Samples received containing less than 10mls of urine (less than half full) will be rejected.

If only a small sample (<10mls or half a container full) is available then please use a white topped container, this should be sent to the laboratory within 4 hours or stored in the fridge until transport to the laboratory is available if collected after the last pick up.

If boric acid is used the sample will be suitable for microbiological analysis for up to 48 hours without the need to store in the fridge.

Suggestions for the collection of urine samples

Mid stream urines

Males

Retract the foreskin if necessary, and then pass the first part of the urine stream into the w.c. pan and catch the second part into a sterile universal/boric acid container.

Females

If there is a menstrual or vaginal discharge, use of a vaginal tampon is helpful. The patient should be instructed to clean the vulva from the front backwards using a cotton-wool swab soaked in sterile water, whilst separating the labia with two fingers of one hand. Antiseptics must be avoided. Keeping the labia separate, the patient passes the first part of the urine stream into the w.c. and catches the second part into a sterile universal container/boric acid container.

Babies and young children

A clean-catch specimen is preferred because urine in adhesive bags is frequently contaminated. A Supra-pubic aspirate (SPA) can be sent in cases where definitive diagnosis is providing difficult.

Urine for TB culture

Three complete consecutive early morning specimens are usually required. These may be refrigerated each day and taken to the laboratory together – the laboratory can supply suitable containers.

Urine for Legionella and Strep pneumoniae antigen testing

Urine samples should be collected in the normal way into a sterile white topped universal container. They can be kept at room temperature for up to 24 hours, but ideally should be sent to the laboratory as soon as possible. Alternatively, they can be stored at 2-8 C in a refrigerator.

For more information regarding specimen collection please see the internet site NHS choices.

Cannulae

The incidence of infection is related to the length of time the cannula remains *in situ*. The catheter tip may be infected secondarily by organisms already infecting the hub or insertion site which track down the catheter lumen or tunnel; but it may also acquire organisms from fluids passing through it or from the bloodstream itself.

Most central venous line-associated infections are caused by organisms from the skin near the exit site which gains access to the intravascular segment of the cannula.

Sample collection**:**

Disinfect the skin around the cannula entry site, remove cannula using aseptic technique, and cut off 4cm of the tip into a Universal container using sterile scissors and send this to the laboratory.

Cannulae should only be sent if there is evidence of infection.

Cannula site swabs

Cannula associated swabs (eg swabs of catheter insertion sites) may be employed as alternative specimens. However, routine investigation of cannula associated swabs from asymptomatic patients is of dubious value.

Sputum

Use 60ml or 30 ml containers with screw-on lids. Specimens obtained by deep coughing or post-physiotherapy specimen are preferred. Saliva and pernasal secretions are not suitable.

Culture of non-purulent material is not helpful as mouth flora inevitably predominates, and will be misleading except when M. tuberculosis is found. For M. tuberculosis and fungal culture, at least three early morning specimens are required.

Cerebrospinal Fluid (CSF)

IN ALL CASES the Microbiology Department must be informed when a CSF has been taken.

During normal working hours the Laboratory must be informed directly via extension 4242.

At all other times the **Microbiology BMS on call MUST** be contacted via switchboard

It is the responsibility of the Doctor initiating the request to ensure that CSF samples are expected by the Laboratory and delivered safely.

For routine Microbiological investigation at least 1 ml of CSF should be sent in sterile universal containers.

For cases of suspected subarachnoid haemorrhage (SAH) at least 2 samples should be sent to Microbiology (ideally the first and last specimen taken) for a cell count, with 1 sample being protected from light and sent to Biochemistry for xanthochromia (normally this should be sample 2).

For biochemical investigations (e.g protein and glucose) further specimens should be sent in accordance with their protocol. Please refer to the Biochemistry web page and user information. In certain circumstances CSF PCR both viral & bacterial can now be performed in house.

Faeces samples

Use blue topped universal container with a collecting spoon.

Samples for examination for Ova, Cyst and Parasites should be freshly passed and sent to the laboratory as soon as practical. Details of any foreign travel is essential.

Suggestions for collection of faeces specimens

When collecting faecal specimens, it is of paramount importance NOT to scoop the specimen from the WC basin as this will be contaminated and may lead to false results.

Pass the motion or part of the motion into a suitable container.

With the spoon attached to the blue lid, scoop some of the motion into the specimen container, taking care not to contaminate the outside of the container.
Do not fill more than half full.

For certain tests (e.g GDH/CDT) the container needs to be at least a quarter full.

Make sure the lid is securely fastened and please ensure that the person's identity (surname forename & DOB) is written clearly on the label of the specimen container, then place and seal it in the polythene bag at the back of the request form provided.

Send the specimen to the Microbiology Laboratory as soon as possible.

For further information, please refer to the NHS Choices website using the following link: NHS Choices- How Should I Collect a Stool (Faeces) Sample?

Specimen Collection Guidelines for *H.pylori* Faecal Antigen Testing

Specimen collection requirements

Solid, semi-solid or liquid cultures are approved for this test and should be transported in an airtight sterile container.

Specimen storage and transport

The specimen should be sent to the laboratory as soon as possible but may be stored in the fridge overnight.

Limitations of the faecal antigen test

Antimicrobials, proton pump inhibitors and bismuth preparations are known to suppress *H.pylori* and ingestion of these prior to testing may cause false negative results to occur. In such cases, the test should be repeated on a new specimen obtained two weeks after discontinuing treatment. A positive test result

for a patient ingesting these compounds within two weeks prior to performing the faecal antigen test, should be considered accurate.

Detection of Threadworms

The following methods are more reliable than examination of faeces sample for the detection of Threadworm infection.

Press a small piece of sellotape (approximately 5-6 cm in length) over the patient's anus – this should be carried out first thing in the morning. The sellotape should then be stuck onto a glass microscope slide, which must then be transported to the laboratory in a plastic carrier box.

A peri-anal swab may be taken. First thing in a morning rub the swab over the skin around the anus, place the swab in a plain universal container and send to the microbiology laboratory.

Please be aware that Threadworm ova are highly infectious, therefore hand-washing after this procedure is essential

Blood cultures

Patient ID stickers must not cover all of the bar code on the bottles, or cover the green detector pad on the bottom of the bottle. Please do not remove the peel off section of bar-code sticker from the bottle as this is required in the laboratory. Please ensure blood culture bottles and form are labelled with the line lumen where appropriate.

If possible, take cultures before starting antibiotics.

Only culture the blood once or twice during each clinical episode (three times for endocarditis from different sites).

Up to 20 ml of blood can be cultured per two-bottle set (minimum of 5ml in each bottle).

If the patient is difficult to bleed, use a Paediatric bottle as <5mls of blood can be used.

Bottles

A standard set of adult bottles consists of 2 bottles, a blue cap aerobic bottle and a purple capped anaerobic bottle

For Paediatric there is a single paediatric bottle for low-volume culture, (ie not more than 5 ml). These bottles have a yellow cap.

Bottles are supplied as part of a blood culture collection pack. This pack should be stored at room temperature and kept away from direct sunlight or heat sources. Always check the date of expiry of the bottles prior to use and return any out of date bottles to the laboratory.

Taking the blood

Please see the current guide to taking a blood culture on the Trust Intranet, instructions are also provided in the blood culture bag with the bottles, butterfly and adaptor cap.

Label the bottles and send them to the laboratory with a Microbiology request form **as soon as possible**.

Do not refrigerate them, or warm them on radiators before transportation to the laboratory.

If the patient is “high risk” then ensure the bottles and request form are labelled with biohazard stickers and **do not** use the air chute to transport these samples to the laboratory.

Fluids

Minimum volume required for microbiological investigation is 1ml, place fluid into a sterile universal container, and tighten the top securely.

Fluid may be inoculated directly into a blood culture bottle, but if a cell count and / or Gram Stain or microscopy is required, fluid in a universal container should be sent.

Inform the On-call Microbiology BMS via switchboard if an urgent cell count and Gram stain is required out of hours.

Skin, nail and hair samples can be tested for fungal infection (Mycology)

Clean the area with a 70% alcohol wipe before sampling, this minimizes contamination and is an aid to microscopy if greasy ointments or powders have been applied

Skin scrapes, nail clippings and small hair pieces should be taken and placed in a sterile universal or Dermapak, and sent to the laboratory as soon as possible. Samples can be stored at room temperature before transportation.

Please send as much material as possible to allow a full investigation to be completed to assist with a correct diagnosis.

Suggestions for taking samples:

Skin samples: using a sterile scalpel blade scrape the edge of the lesion and collect the skin scrapes into a dermapak, fold up and seal or alternatively onto a clean piece of paper, and then transfer this into a sterile universal.

Nail: If possible, collect the subungual debris in addition to nail clippings. Sample the discoloured, dystrophic or brittle parts of the nail only, sampling as far back as possible from the distal part of the nail.

Hair: Pluck hairs from the affected area with forceps (infected hairs come out easily) and scrape the scalp with a scalpel. Preferably, the sample should include hair roots, the contents of plugged follicles and skin scales. Hair cut with a scissors is unsatisfactory as the focus of infection is usually below or near the surface of the scalp.

Genitourinary samples

Nucleic Acid Amplification Test (NAAT)

The laboratory issues specific collection kits for Chlamydia/GC/HSV/Tvag/Mgen NAAT testing. Yellow Aptima tubes are designed for urine and orange aptima for swabs for CT/GC/HSV/Tvag/Mgen testing on the Hologic Panther

Cervical swab specimen collection:

Use the Orange Aptima collection kit for testing CT/GC/HSV/Tv/Mgen on the Hologic Panther

Remove the collection swab from packaging, hold by the 'snap-off' mark on the swab shaft

Insert the collection swab into the cervical canal and rotate for 15-30s.

Withdraw the swab carefully. Avoid contact with the vaginal mucosa.

Uncap the swab diluent tube and fully insert the swab into the tube.

Break the shaft of the swab at the score mark, take care not to splash the contents.

Tightly re-cap the tube and label with the patient information.

Vaginal/vulvovaginal swab patient collection procedure

Wash hands with soap and water.

Remove the swab from it's sheath, hold by the score mark on the swab shaft. (do not touch the soft tip or lay the swab down, if you drop it or lay it down discard and use another swab)

Hold the swab by the cap with one hand so the swab tip is pointing towards you, with your other hand gently spread the skin outside the vagina. Insert the tip of the swab into the vaginal opening. Point the tip towards your lower back and relax your muscles.

Gently slide the swab no more than 2 inches into the vagina, rotate gently as you push if it does not slide easily. If it is still difficult do not attempt to continue. Make sure the swab touches the walls of the vagina so that moisture is absorbed by the swab. Rotate for swab for 10-15 seconds.

Withdraw the swab without touching the skin and place in the tube and screw the cap on securely.

After collection wash hands with soap and water and dry.

Label the tube with patient identification.

Male urethral swab collection-clinic collection only

Remove the swab from the packaging

Insert the swab 2-4cm into the urethra and rotate for 3-5 seconds.

Withdraw the swab, uncap the Swab diluent tube and fully insert the swab into it

Break off the swab shaft at the score mark, use care to avoid splashing the contents.

Tightly re-cap the tube and label with patient information.

Urine collection

The patient should not have urinated for at least 1 hour prior to collection

Collect 2ml of the sample into a yellow aptima collection kit, you may use a collection vessel and there is a pipette within the kit to measure the urine. (this should be the first 20-60 ml of the urine NOT midstream)

Transfer to a plain sterile universal container, label with patient information.

RISH have produced some information videos for patients:

video demo for self-taken vaginal swab STI screen (can be patient or clinician taken)

https://youtu.be/AHrgIibibqA video demo for male urine sample STI screen

Specimen storage and Transport

	Female endocervical swab	Urine	Urine in Yellow Aptima tube
Temperature condition for transport to lab and storage	2-30^{o}C	2-8^{o}C	Room temperature (<30 C)
Process sample	Within 30 days of collection	Within 7 days of collection	within 30 days of collection

Swabs for routine bacterial culture

Place the swab in the tube containing charcoal transport medium, send to the laboratory as soon as possible, if same day transport is not available then they can be stored in a refrigerator overnight but DO NOT FREEZE.

Fine wire swabs (orange topped) are available for use when taking ear swabs.

For the diagnosis of Pertussis, fine flexible wire pernasal swabs (Blue top) are available.

Rectal and other screening site swabs may be sent for CPE/VRE/Candida auris screening.

Pus and swabs

Abscess pus, abscess swab, deep-seated pus swab, post-operative wound swab, wound exudates:

Collect specimens before antimicrobial therapy where possible.

The specimen will usually be collected by a medical practitioner.

Samples of pus are preferred to swabs. However, pus swabs are often received (when using swabs, the deepest part of the wound should be sampled, avoiding the superficial microflora). Swabs should be well soaked in pus.

Swabs for bacterial and fungal culture should be placed in the transport medium provided in the tube.

If possible a few ml of pus in a sterile universal bottle or even a few drops still in a syringe is much better than a swab. (N.B The syringe must be safely capped and needles should NOT be sent)

Ideally, a minimum volume of 1mL of pus should be sent.

The volume of specimen influences the transport time that is acceptable. Large volumes of purulent material maintain the viability of anaerobes for longer.

Numbers and frequency of specimen collection are dependent on clinical condition of patient.

If delays in transportation to the laboratory are unavoidable then the samples should be kept in a refrigerator.

The recovery of anaerobes is compromised if the transport time exceeds 3hr.

Surgical Specimens and tissue/bone samples/ biopsies

SPECIMENS MUST NOT BE PUT IN FORMOL SALINE for microbiology.

Use dry sterile universal containers.

If sending a biopsy for helicobacter culture a small amount of sterile saline should be added to the universal.

Make sure specimens are sent to the laboratory ASAP if this is not possible refrigeration is preferable to storage at ambient temperature; however the recovery of anaerobes is compromised if the transport time exceeds 3 hours.

The laboratory (or on-call BMS) MUST be informed if the specimen is urgent or requires processing out of hours.

Tests performed in Bacteriology and expected turnaround times

SPECIMEN	INVESTIGATION	Container	Expected turn around time (90% completed)	COMMENTS / KEY FACTORS	TEST PERFORMED
Blood culture	Culture & sensitivity	2 bottle set (adult) 1 bottle (paediatric) available from laboratory	5 days (for negative) Positive cultures are phoned ASAP. 10 days extended culture for certain clinical conditions	If possible take cultures before antibiotic therapy commences. Keep culture bottles at room temperature	In-house (Rotherham)
Biopsy	Culture for Helicobacter pylori Culture for other bacterial pathogens including TB	Sterile universal container with 1-2ml saline	15 days 5 days bacteria 50 days for TB		STH In house (Rotherham)
CSF	Microscopy Culture & sensitivity Cryptococccal antigen (PCR) Bacterial/ Viral PCR	Sterile universal container	Microscopy telephoned ASAP Culture 3 days 15days 8days		In-house (Rotherham) Micropathology or Sheffield
Corneal scrapings	MicroscopyCulture & sensitivity Acanthamoeba PCR	Inoculate directly onto agar plates and slides (collect from laboratory) Send contact lens + container	5 days 7 days	If suspecting Acanthamoeba , sending the contact lens container and solution is essential	In house (Rotherham) microscopy & culture Acanthamoeba Micropathology
Faeces	Culture & sensitivity	Universal container with spoon(blue top)	4 days		In-house (Rotherham)
Faeces	Detection of Rotavirus & Adenovirus	Universal container with spoon	2 days Normally done same		In-house (Rotherham)

	Norovirus	(blue top	day		
Faeces	Detection of Clostridium difficile toxin antigen (GDH)(Toxin test only performed if GDH pos)	Universal container with spoon (blue top)	GDH 1 day Toxin 2 days C.difficile ribotyping 14 days	Bristol stool chart 5, 6 or 7 should be sent, samples which are solid or formed and do not take the shape of the container will not be tested. Please ensure a representative sample is sent. Specimen containers must be at least ¼ full. Repeat tests will not be performed if tested negative within 7 days, or positive within 1 month.	In-house (Rotherham) C.difficile Ribotyping – leeds
Faeces	Microscopy for Ova, Cysts & Parasites Threadworm investigations	Universal container with spoon (blue top) Sellotape slide or perianal swab for threadworm	4 days 3 days		In-house (Rotherham)
Faeces	Helicobacter Antigen	Universal container with spoon(blue top)	7 days		In-house (Rotherham)
Genital swab	Culture & Sensitivity	Swab in transport media RISH – culture plates	Routine 4 days GC culture 4 days 4 days for Trichomonas and 14 days for Actinomyces sp	Send to the laboratory as soon as possible If gonorrhoea is suspected	In-house (Barnsley lab) RISH plates (in-house Rotherham)

Genital swab	Chlamydia LGV PCR	Specific swab for Chlamydia or first catch urine	4 days 15 days		In-house (Rotherham) Colindale
Joint/synovial fluid	Culture & Sensitivity Crystals (Histology)	Sterile universal container	4 days	Positive microscopies will be telephoned	In-house (Rotherham) Crystal microscopy - Histology
IUCD	Culture & Sensitivity	Sterile universal container	14 days	Cultured for Actinomyces spp.	In-house (barnsley)
IV Catheter tips	Culture & Sensitivity	Sterile universal container	4 days		In-house (Rotherham)
MRSA and MSSA screen swabs	MRSA/MSSA culture only	Swab in transport media	2 days		In-house (Rotherham)
Mycology – Hair, Nail & Skin scrapes	Fungal culture	Dermapak or sterile universal container	28 days	A provisional microscopy is usually available within 2 working days	In-house (Rotherham)
Pleural Fluid Ascitic fluid	Culture & sensitivity	Sterile universal container	4 days		In-house (Rotherham)
Pus/aspirate	Culture & sensitivity	Sterile universal container	10 days		In-house (Rotherham)
Sputum and Bronchial Alveolar lavage	Culture & Sensitivity	Sterile universal container	4 days 7 days if cystic fibrosis BAL 10 days		In-house (Rotherham)
Sputum	TB culture Mycobacterium PCR	Sterile universal container	50 days (for negative) Positives phoned ASAP 9 days	Send 3 consecutive early morning specimens Positive smears and cultures phoned ASAP	In-house (Rotherham) Birmingham
Swab – Ear	Culture & sensitivity	Fine wire or ordinary swab in transport media	4 days		In-house (Rotherham)

Swabs Eye, Nose, Throat, Skin	Culture & sensitivity	Swab in transport media	4 days		In-house (Rotherham)
Swab – Pernasal	Pertussis culture	Fine wire naso-pharyngeal swab in transport media(collect from lab)	5 days		In-house (Rotherham)
Swab – Pus, Wound, Ulcer	Culture & Sensitivity	Swab in transport media	4 days		In-house (Rotherham)
Tissue and bone	Culture & sensitivity	Sterile universal container – must **not** contain formalin Orthopaedic tissue/JF	10 days 50 days for atypical mycobacteria 18 days		In-house (Rotherham) STH
Urine	Culture & sensitivity	Sterile universal/red top boric acid container	Screen negative results 1 day 3 days for culture		In-house (Barnsley routine, in house Rotherham if urgent)
Urine	TB culture	24 hour container	50 days (for negative)	Send 3 complete, consecutive early morning specimens Positive results phones ASAP	In-house (Rotherham)
Urine	Legionella urinary antigen Pneumococcal urinary antigen	Sterile universal container	1 day		In-house (Rotherham)
Screening swabs including rectal, wound, surface	Screening for VRE, CPE, C. auris, Cipro resistance	Swab in transport media	3 days		In-house (Rotherham)
Antimicrobial susceptibility testing	Testing of pathogenic bacteria for susceptibility to appropriate antibiotics	In-house test	Various	Reports: S=susceptible R= resistant I= high dose	In- house Rotherham & Barnsley

				regime may be appropriate.	

15. Turn Around Times

These are based on the average expected turnaround time from receipt of sample to time of report. We aim to get 90% of results available in this stated time but there may be exceptions with samples processed over extended bank holiday periods and tests performed at specialist referral laboratories.
It is important to note that results are often available on ICE before the times quoted and in addition any urgent results that may affect the clinical management of the patient are telephone by the Consultant Microbiologist or Biomedical Scientific staff as soon as available.
Some serology tests which are performed in-house may require the specimen being sent to a reference laboratory for confirmation and further testing, this will result in a delay in the turn-around time, but all significant results are telephoned ASAP.

Should reporting the result from a sample be significantly delayed and thus compromise patient care, for example through equipment failure, supply problems or contamination issues, the user will be contacted, informed of the reasons and advised of a proposed date for resolution of the problem. An internal incident/internal investigation by CAPA and Datix reporting will be performed if appropriate.

16. Referal Laboratory Tests

Referral Lab	Sample Type	Test
Virology, NGH Sheffield	Serum	Hep B markers including: HBsAg, e Ag, eAb Hep B core total Ab and Hep B core IgM
		Hepatitis B viral load
		HIV Ab/Ag confirmation
		Hepatitis C Ab confirmation/viral load / genotype
		Hepatitis E IgM/IgG
		Syphilis confirmation
		Measles IgM NB: Measles PCR urine or T/S is preferred sample. **No availability**
		Mumps IgM NB: Mumps PCR urine or T/S is preferred sample **No availability**
		HTLV 1/2
	Faeces/ Viral throat swab	Enterovirus PCR,
	EDTA	HIV viral load
		CMV PCR (Generally EDTA for Adults and Urine for children)

		EBV PCR
		ADENO PCR
		HSV PCR
		HIV Resistance
		Parvovirus PCR
	Viral Swab	Respiratory inc Bordetella
		Herpes PCR – Non genital sites
		Eye PCR (Adenovirus & HSV)
	Viral swab	VZV Mumps/Measles PCR
	CSF	Viral PCR (HSV,VZV,Entero)
	Sputum BAL	Mycoplasma/Chlamydia PCR
Bristol Mycology (Myrtle Road)	Serum	Cryptococcal Ag
		Galactomanan/Aspergillus Ag/Beta glucan/fungal PCR. Chlamydia group serology
Micropathology Coventry	EDTA	Viral/Bacterial PCR (various)
	CSF PCR	Both Bacterial & viral PCR
		Bacterial PCR panel
		16S RNA
	Corneal scrape/swab /contact lens	Acanthamoeba
	BAL/ Sputum	Pneumocystis PCR/Tb PCR
Meningococcal Ref Unit, Manchester.	EDTA	Meningococcal PCR
		Pneumococcal PCR
	Serum	Meningococcal Ab
	Culture	Meningococcal typing
	Serum	HSV serology
Hospital of Tropical Diseases, London	Serum	Schistosoma Serology
		Filaria Serology
		Leishmania Serology
		Strongyloides Serology
		Amoebic Serology
		Hydatid Serology
		Toxocara serology
Toxoplasma ref Lab, Swansea.	serum	Toxoplasma confirmation / IgM /Dye test
	serum	Toxoplasma serology on HIV

		positive patients
Heartlands Hospital, Birmingham	sputum	TB confirmation/ susceptibilities/typing/PCR
Bristol Mycology	culture	Fungal identification, Candida sensitivity
Bristol	Serum	Chlamydia group Serology **No longer available**
Biochemisty Northern general, Sheffield	Serum	Teicoplanin assay Tobramycin assay Vancomycin assay
Southmead Hospital, Bristol	Serum	Other Antibiotic assays e.g Itraconazole /Voriconazole /other fungal treatment levels
Colindale, London Virus ref lab	EDTA	HIV-Proviral DNA (usually Mother and baby samples are sent together)
	Serum	Hepatitis D
		Bordetella antibodies
	Faeces	Rota Adeno PCR / Confirmation
General Bacteriology Reference laboratory (GBRU) Colindale	Culture	Pneumococcal typing
		Isolates for typing
		Isolates for identification/confirmation/typing
		Isolate for CPE confirmation
	Biopsy	Biopsies for Helicobacter culture and sensitivity
AMRHAI Colindale STRBU Colindale	Culture	Isolates for susceptibility testing
	Rectal swabs	LGV PCR
Rare & Imported Pathogens Unit (Porton Down)	serum	Borrelia (Lyme disease) serology
		Leptospira Antibodies
		Coxiella (Q fever) serology
		Brucella
		Arboviruses, Zika, Dengue etc
Leeds general Infirmary	Faeces	Clostridium difficile ribotyping
Anaerobic Reference lab University College Wales	cultures	Identification of anaerobic bacteria

Rabies Diagnostic unit	serum	Rabies serology
Dept of Clinical Parasitology Mortimer Market London	serum	Malaria antibodies