

PHE Microbiology Services Colindale Bacteriology Reference Department User Manual

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About Public Health England and the Bacteriology Reference Department

We work with national and local government, industry and the NHS to protect and improve the nation's health and support healthier choices. We address inequalities by focusing on removing barriers to good health.

We were established on 1 April 2013 to bring together public health specialists from more than 70 organisations into a single public health service.

PHE's Bacteriology Reference Department (BRD) is a national and international reference centre for a wide range of bacterial infections. We receive clinical samples and bacterial isolates from public health departments, National Health Service and commercial laboratories across the UK and internationally for specialist testing, bacterial characterisation and susceptibility testing.

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PHE – Bacteriology Reference Department (BRD)

The BRD Department is made up of four units:

1. The Antimicrobial Resistance and Healthcare Associated Infections Reference Unit (**AMRHAI**) is the national reference laboratory for investigation of antibiotic resistance in, and characterization of, healthcare associated bacterial pathogens. AMRHAI seeks to define outbreaks and identify transmission pathways using established and developmental phenotypic and genotypic methods to type isolates, to identify biomarkers associated with virulence, “fitness” etc. and to determine their susceptibility to relevant antibiotics. AMRHAI undertakes surveillance, advises on outbreak investigations, antimicrobial agents that may be appropriate for therapy, and on any public health risk. The Unit also provides identification service for difficult to identify bacteria; information and advice on infection control issues; investigation of healthcare- and community-associated infection, aspects of laboratory safety and other related matters.
2. The Gastrointestinal Bacteria Reference Unit (**GBRU**) works at local, regional, national and international levels to reduce the burden of gastrointestinal infection. Activities include national microbiological reference services for a range of gastrointestinal pathogens as well as the provision of specialist testing for the microbiological examination of clinical, food, water and environmental samples. The laboratory also undertakes research into the genetic diversity of pathogens and the development of improved detection and characterisation techniques for food, water and environmentally borne diseases. GBRU is able to offer expert advice, education and training on public health aspects of food microbiology and safety.
3. The Respiratory and Vaccine Preventable Bacteria Reference Unit (**RVPBRU**), provides national and international reference services for a number of bacteria causing respiratory, systemic and vaccine-preventable bacterial infections including *Streptococcus pneumoniae*, *Bordetella* spp., *Corynebacterium diphtheriae*, *Haemophilus* spp. *Legionella* spp. *Mycoplasma* and *Streptococcus* spp. RVPBRU receives bacterial isolates and clinical samples which are analyzed by a wide range of methodologies in accordance with customer needs. RVPBRU also performs surveillance and advises on incident/outbreak investigation.
4. The Sexually Transmitted Bacteria Reference Unit (**STBRU**) works at a national and international level to reduce the burden of bacterial STIs and receives bacterial isolates and specimens for a number of bacteria responsible for sexually transmitted infections. The diagnostic and reference samples received are analysed by a wide range of methodologies in accordance with customer needs.

STBRU also performs surveillance of antimicrobial resistance and investigates molecular epidemiology of bacterial STIs through various programmes and projects.

The information in this manual was correct at the time it was written but is subject to changes due to the internal and external PHE reorganisation.

Disclaimer

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Amendment History

Version No.	Date	Sections Affected	Pages Affected
1	April 2014	All units combined into one BRD user manual following internal reorganization.	All

Key personnel and contact details

Name	Designation	Telephone
Prof Maria Zambon	Director Reference Microbiology	020 8327 6269
Dr Nandini Shetty	Clinical Governance Lead and Medical Training lead	020 8327 6033
Steve Harbour	Reference Microbiology Operations Manager	020 8327 6432

Medical Microbiologist at Colindale

Name	Contact details
Dr Nandini Shetty	
Dr Meera Chand	if you need to contact the Medical Microbiologist please
Dr Gauri Godbole	email to: ColindaleMedMicro@phe.gov.uk
Dr John Holton	or telephone 0208 327 6736
To be appointed	

Reference Microbiology Services Colindale has recently appointed several Medical Microbiologists to offer advice on diagnosis, clinical interpretation of results and management of infections.

We now have a system in place where a Medical Microbiologist is available every weekday during working hours for advice on medical management of cases, incidents or outbreaks. An on-call Public Health / Microbiology or Virology service is available through the Colindale switchboard.

This service is **not** to access laboratory results.

If you have any issues accessing the microbiologist please contact Jocelyn Krajewska at Jocelyn.krajewska@phe.gov.uk and she will troubleshoot.

BRD General Office

Telephone: 020 8327 7887 (staffed 9am – 5.30pm Mon-Fri)

PHE MS Colindale switchboard: 020 8200 4400

Department addresses

DX address:

PHE Colindale Bacteriology
DX 6530002

Postal Address:

Public Health England
Bacteriology Reference Department
61 Colindale Avenue
London NW9 5EQ [View map](#) (opens external web site)

How to obtain services

Hours of service

The Department is open from 9am to 5.30pm, Monday to Friday. Telephone enquiries via the Department general office are available from 9am to 5.30pm, Monday to Friday. No routine services are available outside these hours. The Department is closed on public holidays.

Services to the public

BRD does not offer diagnostic services to members of the public except via a registered medical practitioner. Results can only be issued to the requesting physician or medical unit and will not be given to patients directly under any circumstances. We reserve the right to check the authenticity of callers in order to protect the confidentiality of patients' personal data.

There are no clinical facilities at PHE Colindale and we are unable to see patients or give telephone medical advice directly to members of the public.

Specimen Submission Guidelines

Specimens

All clinical specimens **MUST** be labelled with at least two of the following unique identifiers:-

- Surname/forename or other unique patient identifier **and/or**
- Date of birth
- Sender's sample number

All environmental specimens **MUST** be labelled with the following:-

- Unique specimen identifier/sender's sample number

Request Forms

Request Forms * **MUST** match and include the above information on the sample

Plus

- Name and contact information of requester (vital for urgent requests)
- Tests required
- Specimen type and site
- Hazard group, if known, or suspected to be Category 3
- Sender's sample number
- Consultant or GP name (if applicable)

Request Forms should also have:-

- Date of sample

Sex
Relevant clinical information
Date of onset
Vaccination history (if relevant to test requested)
NHS number

Please complete the forms in BLACK or BLUE pens (NOT red or any other colour).

Requests for work on presumptive isolates that fall into ACDP Hazard Group 3 MUST be clearly marked to show the findings of the sending laboratory.

If an additional test is required, please discuss with the relevant Unit by telephone. The turnaround time in this instance will vary.

Please use the current versions of request forms where possible and complete all relevant sections. BRD specific request forms are available from the PHE website with guidelines for the completion of forms.

Urgent specimens

If a test result is required urgently, prior telephone contact with the receiving Unit will ensure priority. Always mark 'URGENT' clearly on the request form.

Medico-legal specimens

If referring medico-legal specimens to STBRU please ensure that a chain of evidence form accompanies all specimens. Due to the legal sensitivities of these types of specimen they will only be processed if the laboratory has been contacted in advance and if all paper work is correctly completed. Please contact STBRU for further details [STBRU@phe.gov.uk or 020 8327 6464].

Specimen Transportation

Specimens sent by post or by courier must be in a sealed container, surrounded by sufficient absorbent packing material to take up any leakage in the event of damage during transit, sealed in a plastic bag and placed in an approved outer container which meets current postal or other transport regulations.

Contact the Departmental Safety Manager (Marlette Vigille 020 8327 6447) or the Bacteriology Specimen Reception Manager (Fiona Clode 020 8327 6063) for further information.

Guidance on the transport of infectious substances (including links to current European agreements and information from the HSE) may be found on the following web page:

<http://www.dft.gov.uk/vca/dangerousgoods/useful-links.asp>

Specimen Quarantine Policy

Failure to comply with our specimen submission guidelines and the following quarantine policy may lead to specimen rejection and/or delay of reports.

Please complete request forms as fully as possible. Failure to do so may result in delays or rejection. Some specimens may be rejected if lack of information could expose staff to “high risk” pathogens at the incorrect containment level. Requests for work on presumptive isolates that fall into ACDP Hazard Group 3 must be clearly marked to show the findings of the sending laboratory. See specimen submission guidelines for more details. If a specimen is submitted to BRD for an investigation that we do not offer we archive the sample/isolate and issue a report to the sender explaining the reasons for the sample’s rejection. The sample will be returned if requested (within mainland UK) or discarded after 14 days.

The time taken to perform bacterial identification and typing tests is dependent on the receipt of pure cultures. Cultures that require purification or that cannot be retrieved because they are no longer viable may increase turnaround time significantly or require repeat submission.

Serology tests:

For serological tests, separated serum is preferred. Samples which are highly haemolysed or hyperlipaemic should not be sent as lysed blood or heavily blood stained samples can interfere with serological testing.

Heat-inactivated samples may give rise to erroneous results in a number of assays and should not be sent – please contact the relevant Unit prior to sending the specimen if no other sample is available.

Services available

The department undertakes tests as listed on the following pages. Key factors affecting individual tests are noted against the relevant test, including minimum sample volumes where relevant.

Turnaround times

Turnaround times are from day of receipt to issue of reports in calendar days. The times shown are the typical turnaround times (TAT) achieved by the laboratory, but may be longer or shorter depending on the availability of staff and the complexity of the investigation. BRD staff are committed to the fastest possible issue of reports, consistent with accuracy, on the specimens they examine. Turnaround times may vary during seasonal outbreaks; testing may be conducted more frequently during epidemic seasons. We seek to process at least 75% of specimens received within the published TATs.

Requests for additional tests: time limits and specimen retention

If additional laboratory testing is required on a sample previously submitted to BRD, please contact the relevant Unit in the first instance. Original specimens are normally retained for at least one month (up to several years in the case of certain specimens) but further testing may not be possible due to sample volume constraints, specimen viability or other factors. The Unit will be able to advise on the feasibility of using the original specimen for analysis.

Services	Test type	Sample required	Target turnaround time	Test schedule & Request form	Contact Unit
<i>Achromobacter</i>	Species identification, molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Acinetobacter</i>	Species identification, molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Actinomycetes</i>	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
Antibiotic Resistance Surveys	European Antibiotic Resistance Surveillance Scheme (EARSS) & Surveillance of resistance	Pure culture, Agar slope	14 days	H1	AMRHAJ
Antibiotic Susceptibility Testing	New antimicrobials, Susceptibility testing service, Beta-lactamases, Endocarditis	Pure culture, Agar slope	14 days	H1, H2	AMRHAJ
<i>Bacillus (other than B.anthraxis)</i>	Identification	Pure culture on agar slopes	9 days	L4	GBRU
	Molecular typing	Pure culture on agar slopes	10 days	L4	GBRU
	Detection of emetic toxin gene by PCR.	Pure culture on agar slope	4 days	L4	GBRU
	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Bartonella henselae and B.quintana</i>	Serology	Not less than 400 µL serum in a sterile container	12 days	R1	RVPBRU
<i>Bordetella spp.</i>	Identification	Pure culture on a suitable agar slope or growth from a plate in charcoal transport medium	Varies	R3	RVPBRU
	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Bordetella pertussis</i>	Serology - NOT suitable for immune status	Not less than 400 µL serum in a sterile container	10 days	R3	RVPBRU
	PCR - only offered for infants <= 12 months old who have been admitted to hospital)	Pernasal swab flexible wire shaft and rayon/Dacron/nylon bud), nasopharyngeal swab (preferably NOT in charcoal transport media) or NPA not less than 400 µL in sterile container	24 hours	R3	RVPBRU
<i>Burkholderia spp.</i>	Species identification, molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Burkholderia pseudomallei</i>	Serodiagnosis			Contact laboratory	AMRHAJ
<i>Campylobacter</i>	Identification	Pure culture sent on Amies charcoal swab (preferably) or other suitable medium (e.g. blood or chocolate agar	12 days	Wednesday L4	GBRU

Services	Test type	Sample required	Target turnaround time	Test schedule & Request form	Contact Unit
		slope)			
Chlamydia (respiratory)	<i>C.pneumoniae/C.psittacci/C.abortus</i> - PCR assay	Minimum 200 µl of respiratory sample	Contact laboratory before sending	Urgent phoned	RVPBRU
Chlamydia trachomatis	<i>C.trachomatis</i> :/LGV multiplex - PCR	A confirmed clinical specimen, Residual sample from unprocessed NAAT swab transport medium (>=400uL) or fresh dry swab	5 days	Any day B7	STBRU
	<i>C.trachomatis</i> culture	Fresh specimen taken from site of infection	10-14 days Contact laboratory	Mon – Thurs B5	STBRU
Clostridium botulinum	Detection and identification of C. botulinum from clinical, food or environmental samples by PCR and culture	<ul style="list-style-type: none"> • Serum (≥ 5mL) to be collected close to the onset of symptoms (preferably < 3 days) and before antitoxin is given. Note: lysed or EDTA treated blood specimens are not suitable • Faeces (10g) or rectal washout • Food/Drink samples (10g or 10 mL) 	9 days	Contact laboratory L4	GBRU
	Detection of botulinum neurotoxins in clinical specimens or food		5 days	Contact laboratory L4, L7	GBRU
Clostridium perfringens	Identification of enterotoxigenic C. perfringens by PCR	Pure culture in anaerobic broth or transport swab	5 days	L4	GBRU
	Molecular typing	Pure culture in anaerobic broth or transport swab	10 days	L4	GBRU
	Detection of C. perfringens enterotoxin in faeces by ELISA	≥1g or 1ml of faeces from cases of diarrhoea collected as close to the onset of symptoms as possible (preferably <3 days)	3 days	L4	GBRU
	C. perfringens Toxin typing by PCR	Pure cultures of <i>C. perfringens</i> in anaerobic broth or transport swab	5 days	L4	GBRU
Clostridium tetani	Detection and identification of C. tetani by PCR and culture	<ul style="list-style-type: none"> • Pure cultures of <i>C. tetani</i> in anaerobic broth • Tissue/wound swab inoculated into anaerobic broth 	5 days	L4	GBRU
	Detection of C.tetani neurotoxin in serum (Note: serum will be first tested for tetanus immunity by	Serum (≥ 2mL) to be collected close to the onset of symptoms (< 3 days) and	5 days (following antibody results)	L4	GBRU

Services	Test type	Sample required	Target turnaround time	Test schedule & Request form	Contact Unit
	RVPBRU)	before antitoxin is given. Note: lysed or EDTA treated blood specimens are not suitable			
	Tetanus immunity: serum antibodies	Not less than 200 µL serum in a sterile container	21 days unless urgent	As required <i>R3</i>	RVPBRU
<i>Corynebacterium</i>	Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory <i>H1, H2</i>	AMRHAJ
<i>Corynebacterium diphtheriae</i>	<i>C.diphtheriae</i> and other potentially toxigenic corynebacteria: Identification and Toxin testing by PCR and Elek	Pure culture on blood or Loeffler slope	Within 24 hours (PCR) (6 day service)	Contact laboratory <i>R3</i>	RVPBRU
	Diphtheria immunity: serum antibodies	Not less than 200 µL serum in a sterile container (<5 days post-onset)	21 days unless urgent	<i>R3</i>	RVPBRU
<i>Corynebacterium jeikeium</i>	<i>C.jejkeium</i> Antimicrobial sensitivity	Pure culture		2-3 times weekly <i>H2</i>	AMRHAJ
<i>Cronobacter</i>	<i>C.sakazakii</i> : Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory <i>H1, H2</i>	AMRHAJ
Cystic Fibrosis (CF) Pathogens	Identification	Pure culture, Agar slope	15 days	<i>H2</i>	AMRHAJ
	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	<i>H1, H2</i>	AMRHAJ
<i>Enterobacter spp.</i>	Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory <i>H1, H2</i>	AMRHAJ
<i>Enterococcus spp.</i>	Species identification, molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory <i>H1, H2</i>	AMRHAJ
<i>Escherichia</i>	<i>E.coli</i> : Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory <i>H1, H2</i>	AMRHAJ
	Identification, serotyping, Phage typing, MLVA	Pure culture on Dorset's egg or nutrient agar slope	14 days for non-VTEC 6 days for VTEC	<i>L4</i>	GBRU

Services	Test type	Sample required	Target turnaround time	Test schedule & Request form	Contact Unit
			O157		
	<i>E.coli</i> O157: Serodiagnostic service	Sera aliquots of not less than 500µL	8 days	L5	GBRU
	PCR and Culture detection from faeces	Faecal sample in standard sealed container ≥1 gram	5 days	L4	GBRU
Gram-negative bacteria non fermenter & fastidious organisms	Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory H1, H2	AMRHAJ
Gram-positive bacteria (except <i>C.diphtheriae</i>)	Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory H1, H2	AMRHAJ
<i>Haemophilus</i>	<i>Haemophilus</i> spp. (excluding <i>H.ducreyi</i>) : Identification	Pure culture on chocolate agar slope with cap securely screwed down	12 days	R3	RVPBRU
	<i>H.influenzae</i> : Sero typing and capsular genotyping of <i>H.influenzae</i>	Pure culture on chocolate agar slope with cap securely screwed down	12 days	R3	RVPBRU
<i>Haemophilus</i> spp. & <i>Aggregatibacter</i> spp.	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Helicobacter</i>	<i>H.pylori</i> & <i>H.heilmannii</i> -like organism: Identification, antibiotic susceptibility& PCR	Heavy suspension or Gastric biopsies in saline or transport medium	15 days	Avoid Friday L4	GBRU
Infection prevention and control	Infection prevention and control			Contact laboratory	AMRHAJ
<i>Legionella</i>	<i>L.pneumophila</i> : In-house urinary antigen EIA assay (confirmation of sending lab testing results only)	Not less than 2mL urine sample (soon after onset), mid-stream, early morning with/without preservatives in a sterile container	8 days unless urgent	R1	RVPBRU
	<i>L.pneumophila</i> PCR (from urinary antigen positive patients only)	Lower respiratory tract samples (sputa, BAL, tracheal aspirate etc.) and other clinical samples in a sterile container	Urgent samples should be notified by phone	R1	RVPBRU
	Identification and epidemiological typing of clinical or outbreak associated isolates	Pure culture on either BCYE medium or a dense suspension in sterile distilled water or Page's saline	Varies	R1	RVPBRU
<i>Leuconostoc</i> spp.	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Listeria</i> spp.	Identification, serotyping and molecular typing of	Pure culture on agar slopes	5 days (PCR)	L4	GBRU

Services	Test type	Sample required	Target turnaround time	Test schedule & Request form	Contact Unit
	<i>L.monocytogenes</i>		10 days (fAFLP)		
	Species identification of <i>Listeria</i> by PCR	Pure culture on agar slopes	5 days	L4	GBRU
	Non-cultural detection of <i>L. monocytogenes</i> by PCR	<ul style="list-style-type: none"> CSF ≥50 µL full blood ≥ 1 mL serum ≥ 1 mL 	3 days	Contact laboratory L4	GBRU
	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	H1, H2	AMRHA1
<i>Klebsiella</i>	Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory H1, H2	AMRHA1
<i>Mycoplasma</i>	<i>M.hominis</i> and <i>Ureaplasma</i> spp.: PCR and/or culture	Minimum 200 µL of respiratory , CSF, joint and wound, aspirates in a sterile container	PCR - 5 days Culture – up to 42 days	R1	RVPBRU
	Mycoplasma/Ureaplasma: Biochemical characterisation and molecular methods	Pure culture on mycoplasma medium or chocolate/blood agar slope	Varies	R1	RVPBRU
	<i>M. pneumoniae</i> : PCR	Minimum 200 µL of respiratory sample (LRT or throat swab) in a sterile container CSF with paired respiratory samples	5 days	R1	RVPBRU
	<i>M. genitalium</i> : Molecular detection of the adhesion MgPa gene	Residual specimen from unprocessed NAAT swab transport medium (min volume =400uL), Fresh dry swab, Urine	5 days	Any day B7	STBRU
	Other species: Culture, PCR and sequencing when relevant		PCR - 5 days Culture – up to 6 weeks for some sp.	R1	RVPBRU
<i>Neisseria</i> spp.	<i>N.gonorrhoeae</i> : Confirmation of identification by Phenotypic and molecular	Isolate on chocolate slope or Tran swab	3-5 days	Monday Tuesday Wednesday B2	STBRU
	Susceptibility testing for third generation antibiotics	Isolate on chocolate slope or Tran swab	5-7 days	Monday Tuesday Wednesday B2	STBRU

Services	Test type	Sample required	Target turnaround time	Test schedule & Request form	Contact Unit
	Molecular confirmation of GC NAAT result	Residual NAAT specimen (min volume 400µL)	3 days	Any day B2	STBRU
	Molecular typing for linked isolates by NG-MAST	Linked isolates	Contact laboratory	Contact laboratory	STBRU
	Programme: Gonococcal Resistance to Antimicrobials Surveillance Programme (GRASP)	Sample submission is based on a pre-agreement between the laboratory and STBRU.	GRASP Annual Report	Please contact the laboratory before sending samples.	STBRU
	Programme: European Gonococcal Antimicrobials Surveillance Programme (Euro-GASP)	Sample submission is based on a pre-agreement between the laboratory and STBRU.	ECDC annual report	Please contact the laboratory before sending samples	STBRU
<i>Nocardia</i> spp.	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Pandoraea</i> spp.	Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Pseudomonas</i> spp.	<i>P. aeruginosa</i> antibodies (Serodiagnosis)	Serum (Not less than 200µL in 2 mL micro-tubes)	7-10 days Friday	H3	AMRHAJ
	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
<i>Ralstonia</i> spp.	Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
Resistance Mechanisms	Molecular detection and confirmation	Pure culture, Agar slope	14 days	H1, H2	AMRHAJ
<i>Salmonella</i>	Identification to genus and species level and serotyping	Pure culture on Dorset's egg or nutrient agar slope	17 days	L4	GBRU
	Phage typing	Pure culture on Dorset's egg or nutrient agar slope	10 days	L4	GBRU
	Identification to genus and species level, serotyping and phage typing	Pure culture on Dorset's egg or nutrient agar slope	27 days	L4	GBRU
	Antimicrobial susceptibility testing	Pure culture on Dorset's egg or nutrient agar slope	By arrangement	L4	GBRU
	Molecular subtyping to support outbreak	Pure culture on Dorset's egg or nutrient	By arrangement	L4	GBRU

Services	Test type	Sample required	Target turnaround time	Test schedule & Request form	Contact Unit
	investigations	agar slope			
Serratia	Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory <i>H1, H2</i>	AMRHAJ
Shigella	Identification to genus and species level and serotyping	Pure culture on Dorset's egg or nutrient agar slope	14 days	<i>L4</i>	GBRU
	Molecular typing by MLVA or PFGE (on request)	Pure culture on Dorset's egg or nutrient agar slope	14 days	<i>L4</i>	GBRU
Staphylococcus	Staphylococcal antibodies (Serodiagnosis)	Serum (Not less than 200µL in 2 mL micro-tubes)	12 days	Monday <i>H3</i>	AMRHAJ
	<i>S.aureus</i> : Molecular typing	Pure culture, Agar slope	1-15 days	<i>H1, H2</i>	AMRHAJ
	Staphylococcus coagulase negative: Species identification & Molecular typing	Pure culture, Agar slope	1-15 days	<i>H1, H2</i>	AMRHAJ
	Antimicrobial susceptibility testing	Pure culture, Agar slope	15 days	<i>H1, H2</i>	AMRHAJ
	Resistance gene detection	Pure culture, Agar slope	1-6 days	<i>H1, H2</i>	AMRHAJ
	Virulence gene detection	Pure culture, Agar slope	1-7 days	<i>H1, H2</i>	AMRHAJ
	Detection of staphylococcal enterotoxins A, B, C, D or E in foods or beverages.	Foods/beverages ≥10mL or 10g: keep refrigerated or frozen in a cooled or insulated container. Sample must arrive refrigerated	3 days	Contact laboratory <i>L7</i>	GBRU
Stenotrophomonas	<i>S.maltophilia</i> : Molecular typing and antimicrobial resistance	Pure culture, Agar slope	15 days	Contact laboratory <i>H1, H2</i>	AMRHAJ
Streptococcus	Streptococcal antibodies (Serodiagnosis)	Serum (Not less than 200µL in 2 mL micro-tubes)	12 days	Monday <i>H3</i>	AMRHAJ
	<i>Streptococcus spp.</i> and related genera or Gram positive rods: Identification	Pure culture on blood or chocolate agar slope	12 days	<i>R1</i>	RVPBRU
	<i>S.pyogenes</i> (Lancefield Group A) typing	Pure culture on blood or chocolate agar slope. Charcoal swabs not suitable.	12 days	<i>R1</i>	RVPBRU
	<i>S.agalactiae</i> (Lancefield Group B) typing	Pure culture on blood or chocolate agar	10 days	<i>R1</i>	RVPBRU

Services	Test type	Sample required	Target turnaround time	Test schedule & Request form	Contact Unit
		slope			
	Lancefield Group C & G typing	Pure culture on blood or chocolate agar slope	Contact laboratory	R1	RVPBRU
	<i>S. pneumoniae</i> : Serological typing	Pure culture on blood or chocolate agar slope	10 days	R3	RVPBRU
	Antimicrobial susceptibility	Pure culture, Agar slope	15 days	H1, H2	AMRHAJ
Tetanus	Tetanus immunity: serum antibodies	Not less than 200 µL serum in a sterile container	21 days	R3	RVPBRU
Treponema	<i>T.pallidum</i> (syphilis): Serological	Serum or plasma -Minimum volume 300ul and should be free of lysed blood	7 days	Any day B3	STBRU
	<i>T. pallidum</i> (syphilis): Serological	CSF: Minimum 300uL and should be free of lysed blood	7 days	Wednesdays B3	STBRU
	<i>T.pallidum</i> / <i>Haemophilus ducreyi</i> /Herpes Simplex Virus (HSV) complex (Genital ulcer disease) - PCR	Fresh dry swab or swab in viral transport medium is optimal taken from genital or oral ulcer	5 days	Any day B3	STBRU
Trichomonas	<i>T. vaginalis</i> : Molecular detection by amplifying a 92bp segment of a <i>T. vaginalis</i> specific repeat DNA fragment and is confirmed with molecular detection of the β -tubulin genes	Residual specimen from unprocessed NAAT swab transport medium (min volume =400uL), Fresh dry swab, Urine	5 days	Any day B7	STBRU
Ureaplasma	Refer to Mycoplasma			R1	RVPBRU
Vibrio (including <i>Plesiomonas shigelloides</i>)	Identification to genus and species level and serotyping	Pure culture on Dorset's egg or nutrient agar slope	14 days	L4	GBRU
Yersinia	Identification	Pure culture on Dorset's egg or nutrient agar slope	14 days	L4	GBRU
	<i>Y.enterocolitica</i> and <i>Y.pseudotuberculosis</i> : serodiagnosis	Sera aliquots of not less than 500µL	5 days	L5	GBRU

Reports

Reports will be delivered electronically as PDF documents via E-lab, or will be printed and delivered by post if the referring laboratory is not registered to E-lab.

Details on how to register for E-lab and further information on the system can be found at <http://www.hpa.org.uk/elab>

Policy on faxing and emailing reports containing patients' data

The following guidelines have been prepared having taken into account the Code of Practice on reporting patients' results by fax prepared by the Department of Health and Caldicott recommendations.

- It is PHE Microbiology Services policy that reports containing patients' data should **not** be sent by fax or email.
- Emails cannot be relied on to guarantee security of patients' data because they can be intercepted by a third party en route (except for those sent within the PHE network, within the NHS.Net network, or by encrypted e-mails)
- In **exceptional** circumstances it may be necessary to send a result by fax but not by email. In this case, the following conditions must be adhered to after telephone discussion with the Laboratory. Refer also to "PHE Microbiology Services - recognition of Caldicott recommendations" on page 20 of this manual.
- The report must be sent to a "safe-haven" fax machine. This means that, if the location is in general use, consideration must be given to ensuring that unauthorised personnel are unable to read reports, accidentally or otherwise. Also, the room housing the fax machine must be kept in a secure location which is locked if it is likely to be unattended at the time the fax is sent.
- Assurance must be sought from the intended recipient of the faxed report, preferably in writing, that the receiving fax machine is a "safe-haven".
- Measures must be taken to minimise the risk of mis-dialling, either by double-checking numbers or having frequently used numbers available on the fax machine's memory dial facility.
- Confirmation must always be sought from the intended recipient that the fax is expected and has been received.

Quality assurance in BRD: participation in EQA and IQA schemes

BRD participates in numerous EQA schemes, including those run by the UK National External Quality Assurance Scheme (NEQAS), the World Health Organisation (WHO), Quality Control for Molecular Diagnostics (QCMD) and European Reference laboratories (EURL). Details of participation in specific schemes are available on request.

The quality of our systems is also checked by our IQA schemes, which require selection of referred samples for "blinded" testing at a later date. After processing, the results for BRDW0078.01

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IQA samples are unblinded and are assessed against the results originally reported to the sending laboratory. Any discrepancies are fully investigated as to their root cause before any remedial action is implemented. Results of our EQA and IQA performance are discussed at Management Review meetings, and also at Unit meetings, as appropriate.

Complaints procedure: If there is a problem, or you are not satisfied with the service you have received:

In the first instance contact the appropriate Unit or Section Heads. Contact details are given on the following pages against each Unit, and in a summary list of contacts at the end of the user manual.

We endeavour to be responsive to the changing needs of all users of our services. We welcome comments on how we can improve the provision of these services. Please contact the Department if you have any queries.

PHE Microbiology Services - recognition of Caldicott recommendations

The recommendations of the Caldicott Report (1997) and the subsequent Information Governance Review (2013) have been adopted by Public Health England and by the National Health Service as a whole. These recommendations relate to the security of patient identifying data (PID) and the uses to which they are put. PHE Microbiology Services observes [Caldicott guidance](#) in handling PID and has appointed its own Caldicott Guardian. The Caldicott Guardian advises the Director of Reference Services and others on confidentiality issues and is responsible for monitoring the physical security of PID in all parts of the Colindale site. This also applies to the transfer of results of investigations to and from the site whether by mail services, telephone or fax. The value of 'safe haven' arrangements or other means of the sender and receiver of information identifying themselves to each other before data are transferred is emphasised (page 19 policy on faxing and e-mailing reports containing patients' data).

Customers are asked to draw to the Microbiology Services Caldicott Guardian's attention any instances where PID security has been threatened or has broken down. Uses that PID are put to outside clinical diagnostic services generally allow patient identifiers to have been removed beforehand, and when PID is used for research purposes the proposals are considered first by the PHE Research Ethics Committee. All enquiries about the security and use of PID at PHE Microbiology Services Colindale should be addressed to PHE Caldicott Guardian, Paul Cosford (paul.cosford@phe.gov.uk) or MS Associate Caldicott Guardian, Mubby Husain (mubby.husain@phe.gov.uk).

Compliance with the Human Tissue Act: Submitting tissue samples from deceased people

PHE Microbiology Services Colindale is licensed by the Human Tissue Authority (licence number 12459) to store tissues from deceased people for scheduled purposes. Post mortem samples are submitted by coroners or pathologists for examination to help them determine the cause of death.

Obtaining consent to remove, store and use human tissues for a scheduled purpose is one of the underlying principles of the Human Tissue Act. Microbiology Services Colindale receives post-mortem samples from coroners' post-mortems or from NHS establishments across the UK and therefore we are performing the examination under the authority of the coroner. Unless consent has been obtained or the coroner has requested that samples are retained for further testing, samples are disposed of within three months of the initial test being performed.

When tissue samples from deceased people are received at Microbiology Services Colindale they are retained securely and confidentiality is maintained in compliance with Caldicott principles as are all samples received at this centre. It is normal practice for tissue samples from the deceased to be disposed of in the same way that all other clinical samples we receive are disposed of. However, we will adhere to any specific requirements regarding disposal or returning of tissue samples if requested by the sending coroner or pathologist.

ANTIMICROBIAL RESISTANCE AND HEALTHCARE ASSOCIATED INFECTIONS REFERENCE UNIT (AMRHAI)

Key staff & contact

Head of UNIT	Professor Neil Woodford	020 8327 6511
Opportunistic Pathogens Section	Dr Jane Turton	020 8327 7224
Staphylococcus Reference Section	Professor Angela Kearns	020 8327 7227
Susceptibility Testing Section	Dr Robert Hill	020 8327 7237
Antibiotic Resistance Surveys	Dr David Livermore	020 8327 6511
Resistance Mechanism Section	Dr Katie Hopkins	020 8327 7061
Infection Control Section	Mr Peter Hoffman	020 8327 7274
AMRHAI FAX:		020 8200 7449
E-MAIL ADDRESSES:	Generic:	amrhai@phe.gov.uk
	Individuals:	firstname.surname@phe.gov.uk

Key Services

OPPORTUNISTIC PATHOGENS SECTION

Identification

Phenotypic and sequence-based identification is offered for fermenters, non-fermentative Gram-negative organisms, fastidious Gram-negative organisms, Gram-positive rods and other bacteria with no national reference facility.

Key factors affecting our ability to offer a timely and clinically relevant service:

- Lack of clinical information
- Lack of sender's test results.

Requests for work on presumptive isolates must include:

- Full details of sending laboratory's results
- An indication of whether the isolate(s) may be a hazard group 3 organism
- Full clinical details, including clinical and contact history
- An indication of any recent travel abroad
- Failure to provide necessary information on the form can result in an isolate being handled at Containment Level 2 instead of Containment Level 3, putting staff at risk. In these instances, a report of the incident will be sent to the Health and Safety Executive.
- Failure to provide necessary clinical information on the form can also result in an isolate being tested using inappropriate methods which will have an effect on clinical interpretation and delaying reporting.

Molecular (PCR/sequence based) species identification

These techniques are available to assist with the identification of the following genera and species: *Acinetobacter*, *Burkholderia*, *Enterococcus*, *Klebsiella*, *Achromobacter xylosoxidans*, *Stenotrophomonas maltophilia*, *Burkholderia pseudomallei*, *Cronobacter sakazakii*, *Pandora* and *Ralstonia* species and medically-important pseudomonads.

Molecular (DNA-based) typing

For inter-strain comparative purposes, a molecular typing service is available for all the organisms listed above, plus any other species involved in suspected outbreaks of healthcare-associated infection. Techniques used are pulsed-field gel electrophoresis (PFGE) or Variable Number Tandem Repeat (VNTR) analysis (*Pseudomonas aeruginosa*, *Klebsiella pneumoniae*). In addition, the following are offered:

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Further characterisation of isolates of *Acinetobacter baumannii* by detection of *bla*_{OXA} carbapenemase genes, identification of isolates belonging to the major clonal lineages (international clones I, II and III), and determination of repeat numbers at VNTR loci with small repeat units, that can provide discrimination within a PFGE type.

PCR identification of capsular types K1, K2, K5, K54, and K57 of *Klebsiella* spp., associated with invasive disease, and of two putative virulence factors (*rmpA* and *wcaG*).

PCR identification of epidemic strains of *Pseudomonas aeruginosa* known as the Liverpool, Midlands 1 and Manchester strains, associated with patients with cystic fibrosis.

VNTR comparison of isolates belonging to the *Mycobacterium abscessus* complex

Key factors affecting the performance of the test:

- Poor growers
- Isolates where DNA degrades
- Autolytic enzymes
- Single isolate with no indication of what it should be compared with

Serotyping

Capsular types K1, K2, K5, K54 and K57 of *Klebsiella* spp., associated with invasive disease and virulence, are detected by PCR using serotype specific targets

Serodiagnosis

Serodiagnostic reference services for *Pseudomonas aeruginosa*. The assay is charged for (please refer to latest price lists).

Key factors affecting the performance of the test:

- Whole and lysed blood can affect the test (haemolysed samples are not suitable for testing)

STAPHYLOCOCCUS REFERENCE SECTION

spa typing

This technique is available for the characterisation of *Staphylococcus aureus* (MSSA and MRSA) and involves DNA sequence-based typing of part of the protein A gene. The *spa* repeat succession can often be used to infer which MLST clonal complex an isolate belongs to.

Key factors affecting the performance of the test:

- Some isolates may appear to be non-typable by *spa*; these are rare and can generally be typed using alternative PCR primers
- Some isolates may include repeat units of a “non-standard” length (e.g. 25-28bp as opposed to 24bp). These can still be typed but will include a “??” notation in the repeat succession

Fine strain typing

PFGE-based analyses are available for inter-strain comparative purposes, including suspected outbreaks of MSSA, MRSA or coagulase negative staphylococci (CoNS) in healthcare or community settings.

Key factors affecting the performance of the test:

- Poor growers
- Isolates where DNA degrades
- Autolytic enzymes

A range of PCR and DNA sequence-based techniques for the characterisation of strains of *S. aureus* from healthcare and community-based infections is available. Please contact the relevant personnel to discuss (see list of contacts).

Molecular detection of resistance

PCR-based detection of resistance mechanisms is available, including

- *mecA* and its homologue *mecC*, which confer resistance to oxacillin (*mecA* is charged; *mecC* is free to NHS laboratories)
- *mupA* and *mupB* which confer high-level resistance to mupirocin (Charged)

Toxin gene detection

Toxin gene profiling of isolates of *S. aureus* is available, providing insights into strain virulence. We undertake PCR-based screening for 14 toxin genes: exfoliative toxins A, B and D; enterotoxins A-E and G-J; toxic shock syndrome-1 and Panton-Valentine Leukocidin (PVL).

Non-enteric disease: When requesting *S. aureus* toxin gene testing, select either PVL-testing only or extended toxin gene profiling (the latter includes all 14 toxin genes listed above). Where the toxin request is NOT diagnostic we will not charge but the free text field MUST contain the relevant previous referral details of related isolates e.g. MS - Colindale Laboratory reference numbers or details of the outbreak/diagnostic isolates etc. sent previously. If this is not included, we will assume it relates to primary diagnosis and will charge accordingly (please refer to latest price lists).

Enteric disease: Isolates of *S. aureus* from foods and/or cases of suspected food poisoning are screened for 9 enterotoxin genes (A-E and G-J). The detection of staphylococcal enterotoxins in samples of food or beverages is undertaken by the Gastrointestinal Bacteria Reference Laboratory (GBRU).

Identification of coagulase-negative staphylococci (CoNS)

Phenotypic (biochemical-based) and genotypic (PCR-based) techniques are available for the identification of CoNS.

Key factors affecting the performance of these tests:

- Slow growers
- Organisms with specific growth requirements

Serodiagnosis

Serodiagnostic reference services for *Streptococcus pyogenes* and *S. aureus*. These assays are charged for.

Key factors affecting the performance of the test:

- Whole and lysed blood can affect the test (haemolysed samples are not suitable for testing)

***Staphylococcus aureus* 'phages**

A 'phage typing service for *S. aureus* is no longer available. The International Set of *S. aureus* 'phages, together with their propagating strains, are available from NCTC.

ANTIMICROBIAL SUSCEPTIBILITY TESTING AND RESISTANCE MECHANISMS **SECTION**

Confirmation of unusual resistances

AMRHA1 investigates isolates found by diagnostic laboratories to have unusual resistances, aiming to identify (i) treatment options (ii) emerging resistance of public health importance (iii) underlying

resistance mechanisms, clonal spread of resistant strains. We have the capacity to determine the activity of most antibiotics available in the UK. Please state your requirements clearly on the request form.

Whilst AMRHAI is willing to examine a wide range of resistance phenotypes for customers, we view the following combinations of organism and resistance as exceptional and we advise referral of the isolate:

Organism	Resistance Phenotype
Carbapenem-resistant bacteria*	<p>All Enterobacteriaceae suspected to produce a carbapenemase. #.</p> <p>Do NOT send isolates of <i>Enterobacter</i> that have borderline resistance to ertapenem, but remain fully susceptible to other carbapenems.</p> <p>Do NOT send isolates of <i>Serratia</i>, <i>Morganella</i> or <i>Proteus</i> spp. that are borderline resistant to imipenem, but susceptible to other carbapenems.</p> <p>All <i>Pseudomonas</i> sp. suspected to produce a carbapenemase i.e. isolates resistant to carbapenems, ceftazidime and piperacillin-tazobactam AND with strong imipenem-EDTA synergy (irrespective of susceptibility or resistance to aztreonam).</p> <p>Do NOT send isolates of <i>Pseudomonas</i> spp. resistant only to carbapenems and susceptible to other β-lactams.</p> <p>Do NOT send isolates of <i>Pseudomonas</i> spp. that are resistant to ertapenem, but susceptible to other carbapenems. Ertapenem resistance is inherent in the genus.</p> <p>All <i>Acinetobacter</i> sp. suspected to produce a <u>metallo</u>-carbapenemase i.e. with strong imipenem-EDTA synergy.</p> <p>Do NOT send isolates of <i>Acinetobacter</i> that are resistant to ertapenem, but susceptible to other carbapenems. Ertapenem resistance is inherent in the genus.</p> <p>Despite all of the above, microbiology laboratories are encouraged to have a high index of suspicion, at least for Enterobacteriaceae, and we accept that we won't find a carbapenemase in all referred carbapenem-resistant isolates. There is no penalty charge when we don't (unless isolates turn out to be fully susceptible).</p> <p>Do NOT send isolates of <i>Stenotrophomonas maltophilia</i>, <i>Aeromonas</i> spp. and 'chryseobacteria' for investigation of carbapenem resistance (though note final bullet below), because metallo-carbapenemase production is an intrinsic characteristic of these bacteria.</p> <p>In addition, we seek representatives of any carbapenem-resistant strains (irrespective of suspected mechanism, and including species with intrinsic carbapenem resistance) that are associated with clusters or outbreaks of infection or colonization.</p>

Organism	Resistance Phenotype
<i>S. aureus</i>	Any of: MICs of oxacillin between 2 and 8, vancomycin, teicoplanin, linezolid, quinupristin/dalfopristin, clindamycin-R when erythromycin-S, daptomycin, tigecycline
Coagulase-negative staphylococci	Any of: vancomycin, linezolid, quinupristin/dalfopristin, daptomycin, tigecycline
<i>Corynebacterium jeikeium</i> .	Any of: vancomycin, teicoplanin, linezolid, quinupristin/dalfopristin
<i>S. pneumoniae</i>	Any of: meropenem, vancomycin, teicoplanin, linezolid, quinupristin/dalfopristin, tigecycline, penicillin (MICs >4 mg/L), cefotaxime (MICs >2 mg/L), moxifloxacin
Group A, B, C, G - haemolytic streptococci	Any of: penicillin, vancomycin, teicoplanin, linezolid, quinupristin/dalfopristin, quinolones, tigecycline
Enterococci	Any of: linezolid, daptomycin (<i>E. faecium</i> MICs >4 mg/L; <i>E. faecalis</i> MICs >2 mg/L), tigecycline. Also any isolates resistant to both ampicillin and quinupristin/dalfopristin or to teicoplanin, but not vancomycin.
Enterobacteriaceae, including members of the genera <i>Enterobacter</i> , <i>Escherichia</i> , <i>Citrobacter</i> , <i>Klebsiella</i>	Colistin (except <i>Serratia</i> spp., <i>Proteus</i> spp., <i>Morganella</i> spp.). Also, for <i>E. coli</i> only, tigecycline.
<i>Acinetobacter</i> spp.	Colistin
<i>Pseudomonas aeruginosa</i>	Colistin; MBL-test +ve
<i>H. influenzae</i>	Any third-generation cephalosporin, or carbapenem
<i>M. catarrhalis</i>	Ciprofloxacin, any third-generation cephalosporin
Organism	Resistance Phenotype
<i>S. aureus</i>	Any of: MICs of oxacillin between 2 and 8, vancomycin, teicoplanin, linezolid, quinupristin/dalfopristin, clindamycin-R when erythromycin-S, daptomycin, tigecycline
Coagulase-negative staphylococci	Any of: vancomycin, linezolid, quinupristin/dalfopristin, daptomycin, tigecycline
<i>Corynebacterium jeikeium</i> .	Any of: vancomycin, teicoplanin, linezolid, quinupristin/dalfopristin
<i>S. pneumoniae</i>	Any of: meropenem, vancomycin, teicoplanin, linezolid, quinupristin/dalfopristin, tigecycline, penicillin (MICs >4 mg/L), cefotaxime (MICs >2 mg/L), moxifloxacin
Group A, B, C, G β-haemolytic streptococci	Any of: penicillin, vancomycin, teicoplanin, linezolid, quinupristin/dalfopristin, quinolones, tigecycline
Enterococci	Any of: linezolid, daptomycin (<i>E. faecium</i> MICs >4 mg/L; <i>E. faecalis</i> MICs >2 mg/L), tigecycline. Also any isolates resistant to both ampicillin and quinupristin/dalfopristin or to teicoplanin, but not vancomycin.
Enterobacteriaceae, including members of the genera <i>Enterobacter</i> , <i>Escherichia</i> , <i>Citrobacter</i> , <i>Serratia</i> , <i>Proteus</i> , <i>Providencia</i> , <i>Klebsiella</i> , <i>Morganella</i> , <i>Salmonella</i> #, <i>Shigella</i> #	Ertapenem, meropenem, doripenem, imipenem (except <i>Proteus</i> spp. resistant at low level to imipenem only), colistin (except <i>Serratia</i> spp., <i>Proteus</i> spp., <i>Morganella</i> spp.). Also, for <i>E. coli</i> only, tigecycline.
<i>Acinetobacter</i> spp.	Colistin
<i>Pseudomonas aeruginosa</i>	Colistin; MBL-test +ve
<i>H. influenzae</i>	Any third-generation cephalosporin, or carbapenem

Organism	Resistance Phenotype
<i>M. catarrhalis</i>	Ciprofloxacin, any third-generation cephalosporin

Salmonella and *Shigella* will be processed in the Gastrointestinal Bacterial Reference Unit

*We are regularly asked to define criteria for referring carbapenem-resistant bacteria to us for investigation. These referral criteria have been expanded from those that we included in the UK Standards for Microbiology Investigations document, "[Laboratory Detection and Reporting of Bacteria with Carbapenem-Hydrolysing \$\beta\$ -lactamases \(Carbapenemases\) \(2013\).](#)" They are subjective and under constant review.

We are happy to examine other unusual combinations of resistance(s) and organism(s) and cases where the sender has obtained conflicting results by different methods (e.g. where an automated system identifies an isolate as having a particular resistance but this cannot be confirmed by classical methodology).

We do **not** seek the routine submission of (i) penicillin-resistant pneumococci unless penicillin MICs are >4 mg/L; (ii) glycopeptide-resistant enterococci; (iii) ESBL producers *for confirmation of resistance*.

Determination of minimum inhibitory concentrations (MICs) for referred or survey isolates is done by agar dilution and occasionally by E-test. Interpretative reading of these antibiograms allows assessment of the likely dominant underlying resistance mechanisms.

For the correct interpretation of susceptibilities, use of appropriate breakpoints and interpretation of mechanisms, isolates must be correctly identified to species level. You will be charged if unidentified 'coliform/gram-negative rod' isolates are submitted, unless also formally sent for reference identification. If an isolate is submitted for 'confirmation of results', please be aware that we can only comment if the results requiring confirmation are stated!

If you have a query about a report, please telephone the validator, using the contact details listed on our reports.

Key factors affecting the performance of the test:

- Slow growers
- Organisms with specific growth requirements
- Organisms which have not been identified

Therapeutic guidance

By determining MICs of appropriate antibiotics on submitted isolates, AMRHAI aims to elucidate the most suitable options for treatment. To evaluate susceptibility, we use published clinical breakpoints or, in their absence, advise on the best evidence for any potential antibiotic treatment. Where multiply-resistant isolates are submitted for therapeutic guidance, susceptibilities already established by the sender should be recorded on the submission form, along with appropriate clinical details. Any significant resistance mechanisms relevant to treatment will be interpreted from MIC profiles and reported. Resistances may be further investigated by molecular investigations, which may lead to revisions in data and/or advice. We also undertake interpretation of hospital laboratory data on the telephone when there is an urgency. For urgent referrals, please see below.

Endocarditis

AMRHAI determines MICs for endocarditis isolates to provide therapeutic guidance, as some laboratories choose not to maintain MIC testing capacity. Since this work does not entail investigating exceptional resistance it is charged. To maximise the speed of our response, submission forms must be clearly marked 'ENDOCARDITIS' and the appropriate telephone number for reporting

the results must be given. Users are advised to sign-up to receive reports electronically (as PDFs) via the eLab system, which allows access to results shortly after validation in AMRHAI.

Molecular investigation of resistance

Genes and mutations sought as an uncharged reference service are those that confer resistance to agents of last resort, including carbapenems and linezolid, because resistance to these agents is of public health concern.

Services currently offered include detection of:

- *mecA* and *mecC* in referred *S. aureus* with borderline methicillin / oxacillin resistance or oxacillin/cefoxitin mis-matches (i.e. suspected MRSA giving equivocal results in phenotypic tests). (*mecA* is charged; *mecC* is free to NHS laboratories)
- *mupA* and *mupB* in mupirocin-resistant *S. aureus*; these genes confer high level, clinically-significant resistance. (Charged)
- 23S rRNA mutations responsible for linezolid resistance in enterococci, staphylococci or streptococci.
- Genes encoding carbapenemases in *Acinetobacter*, Enterobacteriaceae or *Pseudomonas* spp.
- Genes encoding acquired (plasmid-mediated) AmpC β -lactamases in *E. coli* and *Klebsiella* spp. resistant to cephalosporins, but with no synergy with clavulanic acid.

New antibiotics

AMRHAI liaises with pharmaceutical companies to test new antibiotics against representative or unusually resistant referred isolates, possibly revealing new treatment options.

Surveys of resistance

Point prevalence surveys of antibiotic resistance are undertaken, giving measures of the extent and nature of critical resistance problems.

Other useful Information relevant to susceptibility testing services

BSAC Site (www.bsac.org.uk)

BSAC Survey Site (www.bsacsurv.org.uk)

INFECTION PREVENTION AND CONTROL ADVICE SECTION

Information and advice is available on infection control issues; education and training; research and audit; disinfection and sterilisation; investigation of healthcare- and community-associated infection, aspects of laboratory safety and other related matters.

GASTROINTESTINAL BACTERIAL REFERENCE UNIT (GBRU)

Key staff & contact

Head of Unit	Dr Kathie Grant	020 8327 7117
Foodborne Pathogens Reference Services	Dr Corinne Amar	020 8327 7341
Gastrointestinal Infections Reference Service	Dr Claire Jenkins	020 8327 6035
Salmonella Reference Services	Elizabeth De Pinna	020 8327 6136
GBRU FAX:	020 8905 9929	
E-MAIL ADDRESSES:	Generic: gbru@phe.gov.uk	
	Individuals: firstname.surname@phe.gov.uk	

Key Services

FOODBORNE PATHOGENS REFERENCE SERVICES (FPRS) SECTION

The Food Borne Pathogens Reference Service (FPRS) provides the national reference facility for the epidemiological typing and toxin testing for a range of Gram positive organisms associated with foodborne infection and intoxication. There is also an active research program on various aspects of foodborne infections and intoxications including the development of improved methods for the detection and characterisation of food borne pathogens.

On identification of a presumptive potential pathogen or high level of toxin, FPRS is required to notify the appropriate Environmental Health Officer, Consultant in Communicable Disease and all other relevant people. Notification will be through a designated, competent senior member of staff.

Bacillus species

Identification of *Bacillus* species, other than *B. anthracis*, and molecular typing of *Bacillus* isolates associated with foodborne outbreaks and other healthcare associated incidents.

What Samples or Specimens to Send

Pure cultures of *Bacillus* on agar slopes isolated from:

- Vomitus, faeces or foods suspected to be or linked with cases of food poisoning.
- Isolates from blood cultures, or from sites that are normally sterile, or other sites where invasive or other diseases are confirmed or suspected.
- Clinical and environmental sources where cross-infection is suspected.
- Foods or beverages with levels of *Bacillus* species including *B. cereus* of $\geq 10^4$ cfu per g or ml.

As foods may be contaminated simultaneously with several species of *Bacillus*, a selection of different colonial types should be sent.

Please fill in the correct request form as completely as possible including your address and telephone number; your specimen/sample reference number; specimen/sample details; your presumptive identification of the isolate together with the testing you require. Brief clinical and epidemiological information including patient details should be included with cultures from cases of infection.

Clostridium botulinum

Diagnostic service for botulism including the detection of botulinum neurotoxin and PCR detection and isolation of *Clostridium botulinum* from clinical specimens, food and environmental samples associated with suspected cases of botulism.

Tests on food and referred isolates are UKAS accredited (UKAS testing laboratory No. 1595).

What Specimens and samples to send

There are five routes by which botulism can arise in humans: foodborne, intestinal colonisation, wound, accidental or deliberate. Details on clinical presentation, diagnosis and laboratory tests for *C. botulinum* are available on the PHE website. Antitoxin for treatment is available on request through the Colindale Duty Doctor System (24 hours telephone: 020 8200 4400) for treatment of foodborne and wound botulism. Advice on treatment and prevention of infant botulism can also be obtained through the Duty Doctor System or from the Infant Botulism Treatment and Prevention Programme, California Department of Health (<http://www.infantbotulism.org/>).

Suspected cases of all forms of botulism should be discussed with the Foodborne Pathogen Reference Service prior to the sending of clinical specimens or samples. This is to ensure that the most appropriate samples are taken and sent under optimal conditions. Specimens should be sent immediately to the reference laboratory and the reference laboratory notified of their arrival so that necessary preparations for testing can be made.

- 10 g or 10 mL or suspected food and drink samples to be sent refrigerated.
- Serum: At least 5mL to be collected as close to the onset of symptoms as possible. Serum specimens must be collected before antitoxin is given. **Lysed or EDTA treated blood specimens are not suitable**
- Faeces: 10g faeces or rectal wash out for toxin detection and a pea sized portion inoculated into cooked meat broth or other anaerobic media for rapid PCR detection and isolation of *C. botulinum*
- Vomitus, gastric washings or gut content: At least 10 g in a sterile container.
- Pus or debrided tissue: To be placed as soon as possible into Cooked Meat Broth or other anaerobic culture medium. If pus is not available, a swab of the lesion should be taken and put immediately into a transport medium or anaerobic culture medium.
- Post mortem specimens: Heart blood, if not haemolysed. Specimens of faeces, gut contents or infected wounds may be useful.
- All cultures suspected of being *C. botulinum* should be sent in a cooked meat medium.

What information to send

Please complete the correct request form in full including your address and telephone number, patient (specimen) details or food (sample) details including your reference number, major patient symptoms, recent travel history, your identification of the isolate and what testing you require. Brief clinical and epidemiological information should be included. **If botulism is suspected, by any route, it is essential that the local CCDC is notified immediately.**

Emergency Situations

During working hours contact a senior member of staff for appropriate urgent attention. Outside working hours, contact the Colindale Duty Doctor (020 8200 4400). Urgent transport of samples to FPRU by taxi or courier should be considered if a clinical diagnosis of food botulism is suspected.

The local CCDC should be notified if a diagnosis of botulism is suspected. Please also notify the Microbiology Services Division, Colindale, Duty Doctor, Phone 020 8200 4400

Turnaround times are shown in calendar days and reflect the proportion of tests requiring prolonged observation in order to establish a negative result. They are dependent upon receipt of sample and request as described above and may vary dependent upon the clinical or public health urgency. If neurotoxin is detected, a turnaround time of >5 days may be required to establish the toxin type.

Rapid PCR detection of *C. botulinum* can be performed within 3 hours of the appropriate sample being received into the laboratory during the working day.

Positive results will be reported immediately by telephone.

For further information or for tests requiring urgent attention, please contact the appropriate member of staff.

Clostridium perfringens

Identification of *C. perfringens* toxin genes in cultures, typing of enterotoxigenic *C. perfringens*, and the detection of *C. perfringens* enterotoxin in faeces

- Pure cultures of *C. perfringens* in Cooked Meat Medium (CMM) isolated from:
- Faeces from cases of diarrhoea obtained after alcohol shock treatment or on direct isolation
- Faeces, gut contents or gut biopsy in cases of suspected necrotising enterocolitis
- Foods
- Faeces and food may be contaminated with several types of *C. perfringens*. It is recommended that in cases of suspected food poisoning several colony picks (three to five in separate CMM) should be sent from faeces and food.
- Faeces for enterotoxin detection in cases of diarrhoea, minimum sample \geq 1g or mL collected as close to the onset of symptoms as possible.
- In cases of suspected necrotising enterocolitis : faeces, gut contents or gut biopsy

What Information to Send

Please complete the correct request form including your address and telephone number, patient (specimen) details including your reference number, major patient symptoms, your identification of the isolate and what testing you require. Brief clinical and epidemiological information should be included from cases of *C. perfringens* diarrhoea. Please indicate if a relationship with other cases by common source is suspected and if the cases are suspected to be food-borne or as a result of person to person spread.

Clostridium tetani

The isolation and identification of toxigenic *C. tetani* (toxin gene detection) and the diagnosis of tetanus in humans by the detection of *C. tetani* neurotoxin in serum

What Samples or Specimens to Send

- Pure cultures suspected to be *C. tetani* in a Cooked Meat Medium (CMM).
- Tissue/wound swab to be placed into a CMM.
- Serum at least 2ml collected as close to the onset of symptoms as possible. Serum specimen must be collected before antitoxin is given and will be tested for the presence of tetanus antibodies before toxin detection is performed.

What Information to Send

Please complete the correct request form including your address and telephone number, patient (specimen) details including your reference number, major patient symptoms, your identification of the isolate and what testing you require. Brief clinical and epidemiological information should be included with cultures/specimens from cases of infection.

Listeria

Identification of *Listeria* species, typing of *L. monocytogenes* isolates and non-cultural diagnosis of listeriosis

What Samples or Specimens to Send

- Pure cultures on agar slopes:

Isolates from all cases of human listeriosis should be sent for sub-typing. All reports are incorporated into a database for national surveillance of listeriosis.

Isolates of *L. monocytogenes* from foods should be sent for sub-typing in the following circumstances:

- When the organism is present at >100 cfu of *L. monocytogenes* per g;
- If the isolates form part of a co-ordinated survey or follow up investigation;
- If there is a particular concern with a specific food product;
- If there is an association with a case of listeriosis.
- CSF (>50µL), full blood or serum if full blood is not available, (>1mL) samples for non-cultural diagnosis by PCR.

FPRS offers a service for the identification of *Listeria* species and this may be helpful when laboratories are experiencing difficulties in this area. Isolates of *Listeria* species other than *L. monocytogenes* where these are present at high numbers in food should also be sent.

Foods may be contaminated simultaneously by several species of *Listeria*, or several strains of *L. monocytogenes*, multiple (ideally three to five) subcultures should therefore be examined for each sample.

What Information to Send

Please complete the correct request form including your address and telephone number, patient (specimen) details including your reference number, major patient symptoms, your identification of the isolate and what testing you require. Brief clinical and epidemiological information should be included with cultures from cases of human listeriosis. A more detailed surveillance questionnaire for completion will be sent for each case.

Molecular sub-typing is performed on all isolates of *L. monocytogenes* submitted to the FPRS for surveillance purposes and to assist in outbreak investigations. Molecular subtyping results are not reported routinely but are available on request.

Food samples and clinical material referred to FPRS

- Clinical specimens and food samples that test negative are kept for minimum of six months whilst those that test positive are kept indefinitely.

Staphylococcal enterotoxin detection

The Foodborne Pathogen Reference Service offers a service for the detection of Staphylococcal enterotoxins A, B, C, D or E in samples of food or beverages.

What Samples or Specimens to Send

- Foods where *S. aureus* was recovered at $\geq 10^4$ cfu per g
- Food or beverages where staphylococcal food poisoning was suspected

Minimum sample size ≥ 10 mL or 10 g

Food samples should be kept refrigerated and sent in a cooled or insulated container to arrive refrigerated.

What Information to Send

Please complete the correct request form with your address and telephone number, details of the sample, your reference number and any epidemiological information.

Campylobacter

We do not provide a serodiagnostic service for *Campylobacter*.

Preston Microbiology Services offer *Campylobacter* serology testing (telephone 01772 522100). GBRU will impose a handling charge for dealing with such requests.

What Specimens or Samples to Send

- Pure culture sent on Amies charcoal swab (preferably) or other suitable media (i.e. blood or chocolate agar slope).

It is advisable to pick campylobacter isolates from a non-selective medium to minimize overgrowth by contaminants. If an overnight delay before posting is anticipated then the isolate should be stored at 4°C. It is not advisable to post samples on a Friday as these will remain at ambient temperature over the weekend.

What Information to Send

Please complete the correct request form including your address and telephone number, patient (specimen) details including your reference number, major patient symptoms, recent travel history, your identification of the isolate and what testing you require.

Helicobacter pylori & Helicobacter species

What Specimens or Samples to Send

- ***H. pylori* cultures:** should be harvested from a 48 to 72 hour culture. A heavy suspension (visibly cloudy) should be prepared in Dent's transport medium* or any rich broth (e.g. Brain Heart Infusion). Alternatively Amies charcoal swabs may be used. Isolates should be transported as soon as possible after harvesting.
- **Gastric biopsies** for culture of *H. pylori* should be sent without delay, preferably within 24 hours. Ideally biopsies should be sent in Dent's transport medium*. Alternatively biopsies can be sent in sterile physiological saline. If a biopsy is not posted / couriered on day of receipt in your laboratory then please store at 4°C. If delays of >96 hours are anticipated (or if biopsies are culture-negative in the sender laboratory), these samples may only be suitable for detection and antibiotic susceptibility testing by PCR. Such biopsies should be frozen (-20°C)

What Information to Send

Please complete the correct request form including your address and telephone number, patient (specimen) details including your reference number, major patient symptoms, recent travel history, your identification of the isolate and what testing you require.

*Dents transport medium is available free of charge from GBRU on request.

We do not provide a serodiagnostic service for *Helicobacter*. The *Helicobacter* service is not supported by grant in aid and there will be a charge for its use.

GASTROINTESTINAL INFECTIONS REFERENCE SERVICE (GIRS) SECTION

The range of reference services includes: identification to genus and species level, phenotypic and molecular typing, resistance typing and antimicrobial susceptibility testing. The section also offers a serodiagnostic service for *E. coli* O157 and other selected serogroups of Vero cytotoxin-producing *E. coli* (VTEC), *Yersinia enterocolitica* and *Yersinia pseudotuberculosis*. Also offered are a primary

diagnostic service for *Helicobacter pylori* and detection of VTEC in faeces from clinically appropriate cases.

Escherichia coli

- Species identification of the genus *Escherichia*
- *E. coli* serotyping
- *E. coli* Verotoxin-producing (VTEC) O157 phage typing
- Typing of VTEC O157 by variable number tandem repeat analysis (VNTR) for surveillance and epidemiology
- Detection, typing and subtyping of Verocytotoxin (VT) genes by PCR
- Identification by PCR of virulence genes in VTEC and in strains that may belong to other groups of *E. coli* associated with diarrhoeal illness. These enterovirulent *E. coli* include: enteropathogenic (EPEC), enteroaggregative (EAggEC), enterotoxigenic (ETEC), enteroinvasive (EIEC), and diffusely-adherent (DAEC) strains.
- Testing of faecal samples for VTEC and other enterovirulent *E. coli*.

What Specimens or Samples to Send

- Pure culture on Dorset's Egg or Nutrient agar slopes.
- Faecal sample In standard sealed container ≥ 1 gram

When submitting a culture to GBRU please pick from a non-selective medium or check the purity before sending. Submitting a pure culture significantly reduces sample processing time.

What Information to Send

Please complete the correct request form including your address and telephone number, patient (specimen) details including your reference number, major patient symptoms, recent travel history, your identification of the isolate and what testing you require. If you have any reason to suspect that the agent being submitted is an ACDP HG3, please indicate this clearly on form.

Shigella, Vibrio & Yersinia species

- Species identification of the genus *Shigella*
- Serotyping of *Sh. dysenteriae*, *Sh. flexneri* and *Sh. boydii*
- Phage typing of *Sh. sonnei*
- Species identification of the genus *Yersinia* (including *Yersinia pestis*)
- Serotyping of *Y. enterocolitica* and *Y. pseudotuberculosis*
- Species identification of the genus *Vibrio* (including *Plesiomonas shigelloides*)
- *V. cholerae* serotyping

What Specimens or Samples to Send

- Pure culture on Dorset's Egg or Nutrient agar slopes.

When submitting a culture to GBRU please pick from a non-selective medium or check the purity before sending. Submitting a pure culture significantly reduces sample processing time.

What Information to Send

Please complete the correct request form including your address and telephone number, patient (specimen) details including your reference number, major patient symptoms, recent travel history, your identification of the isolate and what testing you require.

Serodiagnosis

A serodiagnostic reference service is offered for the following bacteria:

- *Escherichia coli* O157
- *Yersinia enterocolitica*

- *Yersinia pseudotuberculosis*

We do not provide a serodiagnostic service for *Campylobacter* or *Helicobacter*.

Preston Microbiology Services offer *Campylobacter* serology testing (telephone 01772 522100). GBRU will impose a handling charge for dealing with such requests.

Please note that if a patient has undergone renal dialysis or received a blood transfusion as part of their current therapy, this can adversely affect tests for detecting antibodies to *E. coli* O157. Please provide information on request form if any of these treatments have been performed.

What Information to Send

Please complete the correct request form including your address and telephone number, patient (specimen) details including your reference number, major patient symptoms, recent travel history, your identification of the isolate and what testing you require.

SALMONELLA REFERENCE SERVICE (SRS) SECTION

The Salmonella Reference Service (SRS) provides the national reference facility for the epidemiological typing of Salmonella.

Salmonella

- Salmonella identification to genus and species level
- Serotyping of all *Salmonella* species.
- Phage typing for *Salmonella enterica* serotypes Typhi, Paratyphi A and B, Paratyphi B var. Java, Agona, Enteritidis, Hadar, Pullorum, Thompson, Typhimurium, Virchow.
- Supply of phage-typing reagents to WHO affiliated reference laboratories and other approved laboratories within the European Union.
- Training in phage-typing techniques for staff from the above laboratories.
- Monitoring of resistance to antimicrobial drugs of therapeutic and epidemiological relevance;
- Molecular sub typing of Salmonella to support outbreak investigations including PFGE and MLVA
- Investigation of the genetic basis of antibiotic resistance in enteric bacteria.

What Samples to Send.

- **Suspect Salmonella cultures:** should be submitted on nutrient agar or Dorsets' egg slopes in screw-capped containers
- **Urgent submissions:** Advise the Section by telephone of any urgent specimen that is being dispatched to SRS.

What Information to Send

Please complete the correct request form including your address and telephone number, patient (specimen) details including your reference number, major patient symptoms, recent travel history, and your identification of the isolate including the hazard group and what testing you require.

RESPIRATORY AND VACCINE PREVENTABLE BACTERIAL REFERENCE UNIT (RVPBRU)

Key staff & contact

Head of Unit	Dr Timothy Harrison	020 8327 6906
Respiratory and Systemic Bacterial Section	Dr Vicki Chalker	020 8327 6636
Vaccine Preventable Bacterial Section	Dr Norman Fry	020 8327 6776
International Section	Prof Androulla Efstratiou	020 8327 7270
E-MAIL ADDRESSES:	Individuals:	firstname.surname@phe.gov.uk

The laboratory is happy to discuss and advise upon particular clinical or epidemiological problems and outbreak investigations, ask for above Unit/Section Heads in the first instance.

Specimen submissions regarded by the sending laboratory as especially important or urgent should be notified to the unit by telephone to ensure that the appropriate level of priority is accorded to these specimens immediately upon receipt.

Turnaround times will vary depending on the nature of the enquiry and the complexity of the investigation required. Priority will **always** be given to clinical and outbreak associated isolates.

Where services are offered as reference services (i.e. free of charge) for customers in England and Wales, they are offered on the assumption that the primary diagnostic work has been undertaken already. Evidence of such primary testing should be noted on the specimen request forms or a charge will be levied.

Key Services

RESPIRATORY AND SYSTEMIC BACTERIAL SECTION (RSBS)

Lancefield Group A Streptococci (GAS), *Streptococcus pyogenes*

Genotypic and serological classification and epidemiological typing of Group A streptococci (GAS), *Streptococcus pyogenes*.

Typing of GAS is useful in the investigation of both community and hospital outbreaks of GAS infection.

The laboratory requests submission of ALL GAS isolated from blood culture or other normally sterile sites as part of the surveillance of invasive disease due to GAS.

What Specimens or Samples to Send

Pure culture on blood or chocolate agar slope. Charcoal swabs not suitable. **Charcoal swabs are not suitable**

Other information

- Typing of GAS is based upon determination of the M-protein by sequencing the *emm* gene.
- *Emm* genotyping (genotypic detection of the *emm* gene, which encodes M protein) is performed by sequencing the 5'-hypervariable region of the *emm* gene. More than 130 *emm* sequence types, ST(s) have been identified. Results are reported as an *emm* sequence type, which usually correlates with the M protein type, for example: *emm* ST12 = M type 12.

References

Further information and guidelines on group A streptococcal infection are available from the PHE website:

BRDW0078.01

Authorised by: Steve Harbour

Effective Date: 11.04.2014

Lancefield Group B Streptococci (GBS), *Streptococcus agalactiae*

Serological classification and epidemiological typing of Lancefield group B streptococci (GBS).

GBS are a relatively common cause of puerperal and neonatal infections, which may be nosocomially acquired. Epidemiological typing may assist in the investigation of apparent clusters or outbreaks of GBS sepsis in all age groups.

The laboratory requests submission of ALL group B streptococci isolated from blood culture or other normally sterile sites of neonates as part of the surveillance of invasive disease due to GBS in this age group (0-90 days).

GBS may also cause systemic infection in adults (non-pregnancy related). We are pleased to receive blood culture or other "sterile site" isolates for typing and surveillance purposes.

Other information

- The serological classification of GBS is based upon the identification of polysaccharide and protein antigens. There are currently ten polysaccharide antigens designated, Ia, Ib, II, III, IV, V, VI, VII, VIII, IX.
- The most common polysaccharide antigens are serotypes Ia, Ib, II or III. Serotype III is most commonly associated with neonatal infections.
- Molecular typing of GBS is available upon written request and discussion with the unit.

References

Further information on group B streptococcal infection is available from the PHE website:

<http://www.hpa.org.uk/web/HPAweb&Page&HPAwebAutoListName/Page/1202487089475>

Lancefield Group C and Group G Streptococci

We are no longer able to offer this reference service on a "routine basis".

However, the service will continue to be available for urgent public health including outbreak, investigations and in other relevant clinical circumstances but only after discussion and agreement with Unit/Section Heads.

Group C and G streptococci may cause both nosocomial (e.g.: burns unit cross-infection episodes) or institutional outbreaks.

Group C and G streptococci may also cause systemic infections in adults and in particular the taxonomy of group C streptococci may have clinical implications, as (with the exception of the human species *S. dysgalactiae* subsp *equisimilis*) they are all primarily animal species.

Group C streptococci of animal origin e.g.: *S. equi* subsp *zooepidemicus* may cause severe systemic infections in humans. Such infections may occur in clusters and have been associated with the consumption of raw milk.

The current typing methodology for these streptococci is based upon the detection and sequence of the *emm* gene, which encodes the major virulence factor, the M protein. The human group C and group G streptococci carry M protein antigens that are both serologically and genotypically distinct from those carried by the Lancefield group A streptococcus and are useful epidemiological markers.

Emm sequencing is based upon the heterogeneity of the 5' terminus of the *emm* gene which gives rise to the different sequence types. More than 40 *emm* types of group C and group G have been identified and information on these types can be found at: http://www.cdc.gov/ncidod/biotech/strep/M-ProteinGene_typing.htm

References

Further information on group C/G streptococcal Infection is available from the PHE website:

<http://www.hpa.org.uk/web/HPAweb&Page&HPAwebAutoListName/Page/1202487089506>

Identification of Streptococci and related genera

Referred (charged for) taxonomic identification service for streptococci and other related Gram positive, catalase negative genera from systemic and other significant infections.

However, a free-of-charge reference service will continue to be available for urgent public health investigations, outbreaks and incident management, either nosocomial or community based. This should be discussed and agreed with the section head.

Isolates that needs MIC/MBC and that are not streptococci and may be an enterococcus or a Gram positive rod will be referred to the AMRHAI Unit. The turnaround time in this instance will vary.

Other information

- An identification scheme incorporating updated taxonomic methodologies is used.
- Updated nomenclature based upon both the UK and USA classification schemes is used to subdivide streptococci into many species e.g. the 'sanguinis group' is subdivided into *S.sanguinis*, *S.parasanguinis*, *S.gordonii* and *S.cristatus*; *S.australis*; the 'anginosus group' is subdivided into *S.anginosus*, *S.constellatus* subsp *constellatus*, *S.intermedius* and *S.constellatus* subsp *pharyngis*.

References

Further information is available from the HPA website:

<http://www.hpa.org.uk/web/HPAweb&Page&HPAwebAutoListName/Page/1202487089434>

Legionella

A range of reference and confirmatory tests useful in the investigation of individual cases and outbreaks of legionella infection

The laboratory works very closely with the colleagues responsible for national surveillance and reports all clinically relevant results to them. National surveillance of Legionnaires' disease is undertaken by Prof. Nick Phin who may be contacted on 020 8327 6989.

If samples are submitted as part of an outbreak or incident investigation please ensure this is made clear on the request form and the relevant Health Protection Team is identified.

Legionella pneumophila sgp 1 urinary antigen detection

The laboratory encourages and requests the submission of **all** urine specimens for reference and confirmatory testing which have been found to be positive, equivocal or unexpectedly negative using commercially available *L. pneumophila* urinary antigen kits. The specificity of the RVPBRU “in-house” assay enables both confirmation of the submitting laboratory’s findings and determination if the infecting strain was *L. pneumophila* serogroup 1 (mAb2+ve)¹ or not. Samples submitted as UAG positive but found to be negative in our in-house assay will be re-examined using commercial kits to determine if they are genuinely positive (when the infecting strain is either *L. pneumophila* serogroup 1 mAb2-ve or non-serogroup 1) or are falsely positive.

Specimens should be collected as soon as possible after onset of symptoms. Antigen excretion typically continues for 7 - 14 days after onset but may continue for longer in severe cases.

Almost any urine specimen is suitable for examination, but clean-catch, mid-stream, early morning samples, with or without preservative, are most suitable for examination. Grossly contaminated samples may not be suitable.

Please supply details of the assay used and results obtained from primary testing otherwise you will be charged for these tests

Legionella genome detection and culture from clinical material

These services are provided to assist diagnostic laboratories in the investigation of outbreaks of legionella infection and other incidents of potential Public Health significance. Submission of any lower respiratory tract samples from all *L. pneumophila* urinary antigen positive patients is particularly encouraged as such samples are likely to yield useful epidemiological typing data.

These services are not usually offered for primary diagnosis although *L. pneumophila* PCR may be requested as a referred (charged) service after discussion and agreement with the laboratory.

The most commonly referred specimens are sputum and bronchoalveolar lavage (BAL), though the laboratory is pleased to receive any clinical specimens for examination from patients with **other evidence of legionella infection.**

Please supply details of the assay used and results obtained from primary testing.

Identification and epidemiological typing of legionella isolates

The laboratory encourages submission of **ALL** legionellae isolated from clinical material for confirmation and national surveillance purposes. We are also happy to receive any putative legionella isolate from clinical and other sources which is of public health significance.

¹ Most human disease is caused by a subset of *L. pneumophila* serogroup 1 strains that have a virulence associated epitope detected by a monoclonal antibody (designated mAb2). This mAb is used in the RVPBRU assay.

Currently, there are 54 named species of legionellae comprising more than 60 serogroups. *L. pneumophila*, the most frequently encountered species, comprises 16 serogroups.

Identification is made by nutritional characteristics and genotypic methods.

Specialised typing methodologies including monoclonal antibody subgrouping and DNA-sequence based typing are available as part of epidemiological investigations or, when appropriate, **after discussion** with the laboratory.

Please supply details of the assay used and results obtained from primary testing.

Legionella pneumophila serology

The Unit will only undertake *L.pneumophila* serogroup 1 serology at the request of HPTs in support of outbreak or incident investigations. This service is not offered for primary diagnosis or for the confirmation of results obtained using commercial assays.

Mycoplasma

The Unit offers confirmatory and referred services useful in the investigation of individual cases and outbreaks of mycoplasma and ureaplasma infection. These are genome detection and/or culture from clinical material and identification of referred isolates.

QUICK VIEW TABLE (further details below)

Target	Test	Turnaround time	Preferred specimen	Minimum specimen Volume
<i>M. pneumoniae</i>	PCR	5 days	Respiratory sample (LRT or throat swab)	0.2mL
Neonate screen: <i>M hominis</i> <i>Ureaplasma spp.</i>	PCR with culture on PCR positives	5 days	ETS, NPA	0.2mL
Other species	Culture, PCR and sequencing when relevant	Species dependant (see below)	Case dependant (e.g. respiratory, CSF, joint and wound, aspirates)	0.2mL
Isolates	Culture, PCR and sequencing when relevant	Species dependant (see below)	Culture on blood agar or in VTM	N/A

The detection of *Mycoplasma pneumoniae* DNA in clinical samples

This referred (charged) service is available where *M. pneumoniae* infection is of increased likelihood or would be of major clinical significance.

The presence of *M.pneumoniae* DNA in clinical material taken from an acutely ill patient is determined by using a PCR directed against the P1 adhesin gene. Any respiratory specimen is suitable for this test, preferably a lower respiratory tract specimen or throat swab.

CSF samples are rarely, if ever, positive for *M.pneumoniae* and are therefore not routinely tested for *M.pneumoniae* DNA.

The detection of mycoplasma / ureaplasma from clinical material

Detection and culture of mycoplasma is laborious and expensive. This referred (charged) service is not intended for the routine investigation of respiratory illness, but is available where mycoplasma infection is of increased likelihood or would be of major clinical significance.

Neonate screen

U. urealyticum, *U. parvum* and *M.hominis*, may be involved in respiratory infection or rarely meningitis/septicemia in neonates, especially low birth weight infants. The presence of *U. urealyticum*, *U. parvum* and *M. hominis* DNA in clinical material is determined using PCR amplifying the urease gene in ureaplasmas with species-specific probes (Yi *et al.*, 2005) and the glyceraldehyde-3-phosphate dehydrogenase (gap) gene in *M.hominis* (adaption of Baczynska *et al.*, 2004 with an house probe design). Culture will be attempted on all PCR positive specimens.

Other specimens

Mycoplasma and ureaplasmas may cause respiratory and other infections in the immunocompromised. Respiratory specimens from such patients are suitable for investigation. Mycoplasmas have occasionally been isolated from other extra-pulmonary sites including CSF, blood cultures, wound and joint aspirates. The presence of mycoplasmas will be determined using PCR, sequencing and culture when relevant for all human and zoonotic mollicute species except haemoplasmas.

Relevant PCR, sequencing and culture results will be available dependant on the organism in question. Culture results will be available ASAP following successful isolation. Some species such as *M. hominis* take only a few days whilst others such as *M.pirum* may take as long as 6 weeks to isolate.

The identification of putative isolates of mycoplasmas and ureaplasmas

This reference service is undertaken by biochemical characterisation, growth inhibition studies, and molecular methods including 16S rDNA sequencing.

The laboratory is pleased to receive any putative isolates from **clinical material**. The most frequently referred species include *M. hominis*, *U. urealyticum*, *U. parvum* and *M. pneumoniae*.

Priority will **always** be given to isolates of current clinical relevance.

References

Further information is available from the PHE website:

http://www.hpa.org.uk/infections/topics_az/mycoplasma/menu.htm

Bartonella

The Unit offers a referred (charged for) serological service for the diagnosis of infections such as Cat Scratch Disease, and endocarditis which may be caused by *Bartonella henselae* or *B.quintana*

Bartonella henselae and Bartonella quintana serology

The assay used determines antibody levels against *B.henselae* and *B.quintana*: IgM and IgG are estimated separately.

References

Further information is available from the PHE website:

http://www.hpa.org.uk/webw/HPAweb&HPAwebStandard/HPAweb_C/1195733782342?p=1200577814562

Respiratory chlamydiae

The Unit offers a reference service for chlamydia DNA detection by PCR, which may be useful in the investigation of individual cases and outbreaks of respiratory chlamydia infections. This service is only offered where there is a clear Public Health need to establish the diagnosis. Please contact the laboratory to discuss before sending any samples.

This is not a routine service and turnaround times will therefore vary depending on the nature of the enquiry and the complexity of the investigation required.

References

Further information is available from the PHE website:

http://www.hpa.org.uk/web/HPAweb&HPAwebStandard/HPAweb_C/1195733811051

VACCINE PREVENTABLE BACTERIAL SECTION (VPBS)

Bordetella pertussis and other bordetella

A range of reference, enhanced surveillance, and referred tests useful in the investigation of individual cases and outbreaks of pertussis infection. These are serology, genome detection by PCR (for *B.pertussis*), identification and, where appropriate, phenotypic and genotypic characterisation of isolates, including other *Bordetella* spp.

The laboratory works very closely with the Immunisation, Hepatitis and Blood Safety Department of Health Protection Services: Colindale and reports all clinically relevant results to them. National surveillance of pertussis is undertaken by Dr Gayatri Amirthalingam who can be contacted on 02083276407.

Bordetella pertussis serology

The Unit offers a referred (charged for) serological service for the diagnosis of pertussis. Anti-*pertussis* toxin (PT) IgG antibody levels are determined using an in-house EIA.

This service is offered where the following criteria are met: single samples taken >2 weeks after onset of cough for any individuals with a history of prolonged cough.

Please note: This service is **NOT** suitable for assessment of immune status.

Bordetella pertussis genome detection

Currently two regions of the *B. pertussis* genome are targeted. One is directed against the *B. pertussis* 'pertussis toxin promoter' (*ptxP*) and the second is directed against the 'insertion element' IS481, which occurs in *B. pertussis*, *B. holmesii* and some strains of *B. bronchiseptica*.

This service is offered free of charge in England and Wales where the following criteria are met: PERNASAL swab (PNS)/nasopharyngeal swab (NPS) or nasopharyngeal aspirate from an acutely ill child age ≤ 12 months admitted to PICU or paediatric ward with respiratory illness compatible with pertussis. It is offered as a referred (charged) service for colleagues in Scotland and Northern Ireland.

Correct specimen types for this test are pernasal swab (with flexible wire shaft and rayon/Dacron/nylon bud)/nasopharyngeal swab or nasopharyngeal aspirate. Please do not submit nose, nasal or throat swabs. We prefer PNS/NPS for PCR to be submitted **NOT** in transport media, but in sterile container. If submitting NPA, prefer at least 400 μ L in a sterile container.

Do not submit samples that have been collected more than 72 hours previously without first discussing this with the laboratory.

We try to process samples daily and therefore results will usually be available the same day for specimens received by 10am. **An accurate contact telephone number for receipt of results must be provided.** Written confirmation of telephone reports will be provided, usually within 6 days.

The identification and characterisation of *Bordetella pertussis* isolates

The laboratory encourages submission of all *Bordetella pertussis* isolates for confirmation and national surveillance purposes.

Identification and characterisation of *Bordetella* species

The Unit is pleased to receive putative isolates of *Bordetella* spp. from any human source. These will be fully characterised by a range of phenotypic and genotypic methods.

Other information

In conjunction with the Immunisation, Hepatitis and Blood Safety Department of Health Protection Services: Colindale, the VPBS will provide laboratory support for any investigations into pertussis outbreaks. Contact the laboratory before sending any samples.

References

Further information is available from the PHE website:

http://www.hpa.org.uk/infections/topics_az/whoopingcough/menu.htm

Pneumococci (*Streptococcus pneumoniae*)

Serological classification and epidemiological typing of pneumococci

There are currently over 90 different pneumococcal capsular polysaccharide serotypes based upon the Danish classification scheme.

We request submission of ALL blood, CSF and other "sterile site" isolates from episodes of invasive disease for this national surveillance function of our laboratory. Results of serotyping of these isolates

are shared with the Immunisation, Hepatitis and Blood Safety Department of Health Protection Services: Colindale and contribute to National Surveillance.

Presently available and likely future pneumococcal vaccines contain specific, common, capsular polysaccharide antigens. For this reason it is important to monitor the capsular type distribution of isolates from invasive disease in both adults and children.

Capsular typing of pneumococci may also be helpful in the investigation of instances of suspected cross-infection in hospitals, other residential institutions and day care centres (or similar) for children.

The Unit liaise closely with the AMRHAI in studies of antibiotic resistant pneumococci.

In period of very heavy workload priority will be given to isolates referred from children (see below).

Other information

- Some of the 90 serogroups/serotypes may be divided into specific serotypes or subtypes i.e.; types carrying the same number but different letters, e.g. 6A, 6B, 9A, 9L, 9V. Subtyping is undertaken on all sterile site isolates, in particular for any episode of systemic infection associated with possible vaccine failure.
- The more common serotypes are currently (October 2011), in order of prevalence, 7F, 3, 19A, 8, 22F, 1, 6C and 15A. RSIL, together with the Immunisation, Hepatitis and Blood Safety Department of Health Protection Services: Colindale, are actively following up all cases of invasive pneumococcal disease in the childhood age groups targeted for vaccination in order to ascertain immunisation history and determine vaccine effectiveness. This applies to anyone born after 4th September 2004.

References

Further information is available from the PHE website:

<http://www.hpa.org.uk/web/HPAweb&Page&HPAwebAutoListName/Page/1203008863939>

Identification and toxigenicity testing of *Corynebacterium diphtheriae* and other potentially toxigenic corynebacteria (*C. ulcerans* and *C. pseudotuberculosis*)

Identification/confirmation and toxigenicity testing of potentially toxigenic *Corynebacteria* is performed initially by realtime PCR (qPCR) on a DNA extract of the submitted isolate. Isolates which are qPCR positive for the toxin gene (*tox*) will also be tested by the Elek test for toxin expression. Although *C. diphtheriae*, *C. ulcerans*, and *C. pseudotuberculosis* toxin gene PCR positive results will be confirmed by the Elek test, a toxin gene PCR positive result should be acted upon without waiting for the Elek result. A toxin gene PCR negative result is final and no further toxigenicity testing will be undertaken on these isolates.

This service is available Monday to Friday during normal working hours. A Saturday service is also available for urgent isolates received before 12 noon on the Saturday. For advice on laboratory diagnosis of diphtheria and/or submission of samples to Colindale during normal working hours contact Unit/Section Heads.

Out of hours and weekends, advice and details of service available from the Duty Doctors via the Colindale main switchboard operator/Vodafone answering service (Tel. 020 8200 4400).

Cultures should be submitted to the laboratory by above to inform them that a culture is on its way.

Advice on immunisation against diphtheria, provision of vaccine and provision of diphtheria antitoxin for **therapeutic** use is available from the Immunisation, Hepatitis and Blood Safety Department of PHE Colindale on 020 8200 6868 (Dr M. Ramsay, 020 8200 7085 or Joanne White 020 8327 7446) during normal hours. Out of hours via Colindale Duty Doctors on 020 8200 4400.

Toxigenic *C.diphtheriae* is very uncommon within the UK and is almost always imported. A travel and immunisation history should **always** be obtained from suspected cases of diphtheria and, if feasible, their close contacts.

Some strains of *C.ulcerans* (and very rarely *C.pseudotuberculosis*) may produce diphtheria toxin and the illness caused may present as clinical diphtheria. Such infections should be treated as diphtheria with the important proviso that person-to-person transmission is extremely rare. Infection is usually acquired from raw milk and/or contact with farms and farm animals and also perhaps close contact with companion animals.

UK microbiological laboratories are encouraged to submit all isolates of *C.diphtheriae* and other potentially toxigenic corynebacteria to RVPBRU for surveillance and monitoring purposes. The Unit is a designated WHO Collaborating Centre for reference and research on diphtheria.

Urgent isolates received by 12 noon will be processed on the same day. All results are communicated by telephone. Under normal circumstances, a final written report is issued within 5 days of receipt and all interim results are given by telephone usually within 24 hours.

Notify RVPBRU (telephone 0208 327 7887) before sending an isolate for toxigenicity testing within working hours on a weekday. Outside these hours, please notify the Colindale duty doctor on 0208 200 4400. If possible always use the RVPBRU Request Form (R3) and ensure full contact telephone numbers are provided on the form.

References

Further information is available from the PHE website.

Guidelines for the Public health control and management of diphtheria (in England and Wales) have recently been updated to take account of recent changes in confirmatory testing and the new Public Health England (PHE) structures established in April 2013. The updated interim guidelines will be available on the website.

Haemophilus influenzae

Identification, serological typing and capsular genotyping of strains of *Haemophilus influenzae* isolated from cases of invasive disease.

Conjugate *H.influenzae* type b vaccine is routinely offered to all infants in the UK. Typing of strains of *H.influenzae* is invaluable in determining whether the strain is *H.influenzae* type b that is a vaccine preventable serotype, a non-type b serotype or non-capsulated strain.

The Unit requests submission of **ALL** *H.influenzae* isolated from blood culture or other normally sterile sites in patients of **ALL** ages as part of the surveillance of invasive disease due to *H.influenzae* and in

children aged 0 – 16 years as part of the surveillance of invasive disease due to *H. influenzae* and Hib vaccine failures in children. This surveillance is being conducted in collaboration with the Immunisation, Hepatitis and Blood Safety Department, PHE, Colindale

The laboratory is happy to discuss and advise upon particular clinical or epidemiological problems.

Other information

- Identification of *H.influenzae* is based upon X and V factor requirement and lack of haemolytic activity on blood agar.
- There are 6 capsular serotypes of *H.influenzae* (a-f) based on the capsular polysaccharide of the organisms. The majority of serious human infections are caused by *H.influenzae* type b, for which a conjugate vaccine is now available.
- Other capsular serotypes, notably types e and f and non-capsulated strains can cause serious infections.
- The Unit will refer requests for antimicrobial susceptibility testing to AMRHAI.
- The Unit does not offer a routine service for typing or susceptibility testing *H.influenzae* strains from non-invasive infections. Non-invasive isolates of *H.influenzae* (i.e. isolates from eye swabs, sputum, etc.) will only be examined if there are sound clinical or epidemiological reasons for the investigations. The laboratory is happy to discuss any clinical problem that may warrant further investigation.
- The Unit does **NOT** carry out tests for Hib antibodies. Hib serology is performed by Professor Ray Borrow, Meningococcal Reference Unit, PHE Microbiology Services, Manchester Medical Microbiology Partnership, Clinical Sciences Building 2, Manchester Royal Infirmary, Oxford Road, Manchester, M139WL Please contact Professor Ray Borrow (0161 276 6793) or Unit/Section Heads for further advice on Hib serology.

Identification of Haemophilus species (excluding *Haemophilus ducreyi*)

Identification service for strains of *Haemophilus* species isolated from cases of invasive disease.

For isolates not confirmed as *Haemophilus spp* a preliminary report will be issued and the isolate forwarded to AMRHAI for full identification, who will issue a report in due course.

References

Further information is available from the PHE website:

http://www.hpa.org.uk/infections/topics_az/Haemophilus_influenzae/menu.htm

Diphtheria immunity/vaccination studies

A referred (charged for) service for the determination of serum antibodies to diphtheria toxin

Diphtheria immunity status is determined by a tissue culture toxin neutralisation assay of serum antibodies specific for diphtheria toxin. Test plates are incubated for up to six days before a final report is issued. This assay is more reliable than ELISA, particularly for detecting susceptible individuals.

Results are reported in International Units/mL and classified as:

BRDW0078.01

Authorised by: Steve Harbour

Effective Date: 11.04.2014

- Individual is susceptible: <0.016 IU/mL;
- Levels conferring some protection: 0.016 – 0.09 IU/mL;
- Protective levels: 0.1 – 0.9 IU/mL;
- Levels conferring long-term protection: \geq 1.0 IU/mL.

Tests are batched every three weeks, unless a sample is deemed to be urgent.

Please supply details of vaccination history (if known) with all requests plus relevant clinical details.

References

Further information is available from the PHE website:

<http://www.hpa.org.uk/webw/HPAweb&Page&HPAwebAutoListName/Page/1191942152928?p=1191942152928>

Tetanus immunity

A referred (charged for) service for the determination of serum antibodies to tetanus toxin

Tetanus immunity status is determined by an ELISA for serum antibodies specific for tetanus toxin.

Provided serum is collected prior to therapeutic administration of antitoxin, determination of tetanus immunity status can be useful in confirming a clinical diagnosis of tetanus. Absence of detectable antibody or levels below or close to the minimum protective level lends support to the clinical diagnosis whilst higher levels do not.

Results are reported in International Units/mL. Minimum protective level is presently defined as 0.1 IU/mL.

According to demand tests are normally batched every three weeks, on occasion less frequently. If a sample is deemed to be urgent, a same day result can be produced.

Please supply details of vaccination history (if known) with all requests plus relevant clinical details.

References

Further information is available from the HPA website:

http://www.hpa.org.uk/infections/topics_az/tetanus/menu.htm

SEXUALLY TRANSMITTED BACTERIA REFERENCE UNIT (STBRU)

Key staff & contact

Head of Unit	To be appointed	020 8327 6464
Specialist and Reference Services	Dr Sarah Alexander	020 8327 6462
Molecular Epidemiology and Surveillance Section	Dr Stephanie Chisholm	020 8327 7326
E-MAIL ADDRESSES:	Individuals:	firstname.surname@phe.gov.uk

The Sexually Transmitted Bacteria Reference unit (STBRU) is an expanding laboratory that provides reference and specialist services for the bacterial sexually transmitted pathogens, *Neisseria gonorrhoeae*, *Treponema pallidum*, *Chlamydia trachomatis*, *Trichomonas vaginalis*, *Haemophilus ducreyi* and *Mycoplasma genitalium*. The laboratory is happy to discuss and advise upon particular clinical or epidemiological problems and outbreak investigations, ask for above Unit/Section Heads in the first instance.

Key Services

SPECIALIST AND REFERENCE SERVICE (SRS) SECTION:

STBRU currently provides a full reference service for the identification and typing of *N. gonorrhoeae*, serological confirmation of *T. pallidum*, molecular diagnostics for *C. trachomatis*, *T. pallidum*, *H. ducreyi*, *T. vaginalis* and *M. genitalium*.

Neisseria

STBRU offer two *N. gonorrhoeae* reference services:

- GC NAATS Molecular Confirmation Service
- *N. gonorrhoeae* Culture Confirmation Service (identification, sensitivity testing, etc.)

GC NAATS Molecular Confirmation Service

STBRU offer a *N. gonorrhoeae* molecular confirmation service for the direct detection of GC specific DNA from clinical samples.

This service is aimed at laboratories that are using GC NAATs and wish to comply with the United Kingdom current guidelines. These recommend that GC NAATS should only be used if positive predictive value (PPV) of >90 % can be achieved. The low prevalence of gonorrhoea in most settings, means that for many labs this is not achievable.

- The PPV of GC NAATs can be increased by confirming all positive results using a second, confirmatory assay which targets a different genetic sequence.
- The STBRU GC NAATs confirmatory service is aimed at aiding laboratories -who only have access to a GC NAAT platform with a single target- by confirming their GC NAAT positive specimens.
- This service is available for any clinical specimen, which has been determined to be *N. gonorrhoeae* positive at the local laboratory using a NAAT.
- STBRU will accept the residual processed NAAT specimen from any specimen site.
- STBRU will also accept for confirmation any clinical specimen, which is found to be discordant between *N. gonorrhoeae* culture and NAAT.

Please note that this service is intended as a confirmatory service and not a primary diagnostic service. Any clinical specimens which are received for this service that have not (i) been tested using a GC NAAT and produced a reactive result or (ii) have insufficient information on the referral form, will be charged.

See STBRU webpage for useful documents:

- Guidance for GC testing in England and Wales. (Department of Health)

- GC NAATs National SOP - If you require a hard copy of QSOP62 please email Standards@PHE.gov.uk

Referral of Putative *N. gonorrhoeae* Cultures

STBRU will accept viable cultures on a chocolate slope or on a transport swab.

Reference service is available for isolates that require:

- Confirmation of identification because results were anomalous;
- Confirmation of identification for medico-legal purposes (please contact lab before sending samples);
- Susceptibility testing for third generation cephalosporins, ceftriaxone, and cefixime and azithromycin.

Identification of *N. gonorrhoeae* will be confirmed using:

- Gram Stain and Oxidase test
- Biochemical methods: API NH
- Immunological methods: Phadebact
- Molecular identification: by amplification of PorA gene pseudogene and OPA gene multiplex
- Susceptibility testing for ceftriaxone, and cefixime and azithromycin; when requested.

Any samples received that do not meet our referral criteria will be charged.

Other services available include:

- Typing of medico-legal isolates from linked cases. This is performed using NG MAST which examines diversity into hyper variable genes (Por & tppB). For further information please contact the laboratory directly with details.
- If referring medico-legal specimens to STBRU please ensure that a chain of evidence form accompanies all specimens. Due to the legal sensitivities of these types of specimen they will only be processed if the laboratory has been contacted in advanced and if all paper work is correctly completed. Please contact STBRU for further details [STBRU@phe.gov.uk or 020 8327 6464].

Charges will be incurred for medico-legal work and tests not covered by the reference service.

Treponema

Syphilis serology

STBRU will accept serum and CSF specimens from patients with suspected syphilis infection for confirmation.

- We will require a minimum volume of 500µl
- All referred samples will be tested by:
 - Total antibody Enzyme Immunoassay (EIA)
 - IgM EIA (except CSF)
 - Treponemal Pallidum Particle Agglutination (TPPA)
 - Rapid Plasma Reagin (RPR)

Any samples for which discrepant results are obtained an Inno-LIA (Inno-LIA Syphilis Score) will be performed.

Please refer to the STBRU webpage for Charges of the Referral of specimens for Syphilis Serology.

Molecular testing of samples from genital ulcer disease (*T. palladium* and *H. ducreyi*)

Note; this test also includes a PCR for Herpes simplex virus, which is used as differential diagnosis during the investigation of genital ulcers.

STBRU will accept a fresh swab (sterile, cotton or dacron tipped) taken from the patient's ulcer (genital or anal).

Roll the swab over the base of the ulcer twice if possible. Send the swab dry, or put into transport buffer, as soon as possible to STBRU. If there is any delay in sending, store the swab in a fridge. Please complete our standard syphilis referral form. This is a charged test.

Chlamydia

Lymphogranuloma Venereum (LGV) Diagnostic Service

- Lymphogranuloma Venereum is a sexually transmitted infection caused by the L serovars of *Chlamydia trachomatis* (L1, L2 and L3), these strains are associated with a more chronic and invasive infection than standard genital chlamydia. As patients infected with LGV have a requirement for extended antimicrobial therapy, laboratory tests that can differentiate LGV from non-LGV-associated serovars of *C. trachomatis* can be clinically important.
- In 2003 as a response to an outbreak of LGV in men who have sex with men (MSM) that had been reported in Western Europe, STBRU launched a diagnostic service for the detection of LGV directly from clinical specimens.
- Since 2003 there has been a clear and steady rise in LGV cases within a core population of HIV positive MSM and it is evident that rather than being considered as an outbreak infection, LGV has now become endemic within this cohort. Therefore as of April 2013 this service will be charged.

STBRU are however very keen to help local NHS laboratory's to establish their own LGV diagnostic service and are able to provide both guidance and support. Please contact STBRU for further details: STBRU@phe.gov.uk

C. trachomatis Culture Service

STBRU offer a free of charge primary diagnostic service for the detection of viable *C. trachomatis* infection. The service is aimed at patients with persistent infections who have failed previous treatment with a first-line therapeutic regime (azithromycin or doxycycline) and have been confirmed as *C. trachomatis* positive at the local laboratory.

STBRU will accept swabs from patients fitting the above criteria. Correct storage of the swabs is essential to preserve viability of the organisms. It is therefore requested that specimens are sent to STBRU **only** with prior notification.

For more information please refer to the STBRU Chlamydia Culture Service Guidelines on the STBRU webpage.

Mycoplasma

Molecular detection of *M. genitalium*

Molecular detection of *M. genitalium* in clinical samples is determined by real time PCR directed against the MgPa adhesin gene and confirmed by a different target.

Samples accepted by STBRU:

- STBRU will accept specimens and DNA extracts for *M. genitalium* from patients with clinical signs or known contact cases.
- Specimens accepted include;
 - Extracted DNA
 - Rectal and genital swabs
 - Urine

Please note, charges will be levied for this service.

Trichomonas

Molecular detection of *T. vaginalis*

Molecular detection of *T. vaginalis* in clinical samples is determined by real time PCR against a 92bp segment of a *T. vaginalis* specific repeat DNA fragment and is confirmed with molecular detection of the β -tubulin genes

Samples accepted by STBRU:

- STBRU will accept specimens and DNA extracts for *T. vaginalis*
- Specimens accepted include;
 - Extracted DNA
 - Rectal and genital swabs
 - Urine

Please note that this service is intended as a confirmatory service and not a primary diagnostic service. Charges will be levied for this service.

MOLECULAR EPIDEMIOLOGY AND SURVEILLANCE SECTION (MESS):

This section of STBRU performs surveillance of antimicrobial resistance and investigates molecular epidemiology of bacterial STIs through various programmes and projects.

The Gonococcal Resistance to Antimicrobials Surveillance Programme (GRASP) is a sentinel surveillance scheme monitoring antimicrobial resistance in *N. gonorrhoeae* across England and Wales, annually.

Participating laboratories refer all gonococcal isolates identified over a three-month period (July – September) to STBRU for susceptibility testing. Isolates can be referred as follows:

- Stored in glycerol broth/beads at -80°C (frozen batches collected by courier by arrangement).
- Or
- On chocolate slopes or VCM transport swabs, by prior agreement with STBRU.

For more information please contact the laboratory.

SUMMARY LIST OF CONTACTS

Antimicrobial Resistance and Healthcare Associated Infections Reference Unit (AMRHAU)			
Prof Neil Woodford	Unit Head	neil.woodford@phe.gov.uk	020 8327 6511
Susceptibility testing, interpreting antibiograms, treatment			
Dr Robert Hill	Section Head	Robert.hill@phe.gov.uk	020 8327 7237
Infection prevention and control, site visits			
Mr Peter Hoffman	Section Head	Peter.hoffman@phe.gov.uk	020 8327 7274
Resistance mechanisms, inferring mechanism from antibiograms, commercial opportunities			
Dr Katie Hopkins	Section Head	Katie.hopkins@phe.gov.uk	020 8327 7061
Staphylococci ID & typing, PVL / other toxins, staph/strep serodiagnosis			
Prof Angela Kearns	Section Head	Angela.kearns@phe.gov.uk	020 8327 7227
Commercial opportunities (esp. antibiotic evaluations), surveys			
Prof David Livermore	Section Head	David.livermore@phe.gov.uk	020 8327 6511
Gram-negative typing and Serodiagnosis, identification			
Dr Jane Turton	Section Head	Jane.turton@phe.gov.uk	020 8327 7224
Gastrointestinal Bacteria Reference Unit (GBRU)			
Dr Kathie Grant	Unit Head	Kathie.grant@phe.gov.uk	020 8327 7117
<i>E. Coli</i>, Shigella, Vibrio, Yersinia, serodiagnosis			
Dr Claire Jenkins	Section Head	Claire.jenkins@phe.gov.uk	020 8327 6035
Salmonella identification, serotyping, phage typing			
Elizabeth De Pinna	Section Head	Elizabeth.depinna@phe.gov.uk	020 8327 6136
Bacillus, Clostridium, Listeria, Staphylococcus, Campylobacter, Helicobacter			
Dr Corinne Amar	Section Head	Corinne.amar@phe.gov.uk	020 8327 7341
Respiratory and Vaccine Preventable Bacteria Reference Unit (RVPBRU)			
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Streptococcus, Legionella, Mycoplasma/Ureaplasmas, Bartonella			
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Neisseria, Treponema, Mycoplasma genitalium			
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Dr John Holton		john.holton@phe.gov.uk	0208 327 6772