South London Specialist Virology Centre

VIROLOGY

Laboratory User's Handbook April 2014 version



www.kch.nhs.uk

www.viapath.co.uk



www.clinicalvirology.org

South London Specialist Virology Centre in conjunction with Department of Medical Microbiology

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1. General information

Our laboratories are based at Kings College Hospital NHS Foundation Trust. We provide an extensive clinical microbiology service, including infection control services and specialist advice in microbiology, virology, parasitology and mycology to hospitals and General Practitioners. South London Specialist Virology Centre is part of the UK clinical virology network.

Epidemiological data are provided for the Communicable Disease Surveillance Centre in Colindale. Outbreaks of infectious disease are investigated in conjunction with the Consultants in Communicable Disease Control.

Virology is accredited by the Clinical Pathology Accreditation (CPA) scheme and MHRA. It participates in National Quality Assurance Schemes.

In October 2010, we joined the KingsPath team having previously been the HPA London Regional Laboratory, and along with our other colleagues have entered the joint venture with GSTS Pathology who provide pathology services at Guy's & St Thomas' NHS Foundation Trust, and are preferred partners for the KingsPath Partners Academic Health Science Centre. (on 1st May 2014 GSTS and KingsPath became Viapath.)

1.1 Where to find Virology

Postal addresses

South London Specialist Virology Centre Viapath LLP King's College Hospital NHS Foundation Trust Cheyne Wing, 2nd Floor (opposite Liver ITU) Denmark Hill London SE5 9RS

DX address:

South London (PHL) Kings DX 6570200 Peckham 90SE

Website contacts:

www.viapath.co.uk

www.clinicalvirology.org

1.2 Population served

King's College Hospital NHS Foundation Trust provides a wide range of services to the population of South East London. During 2009/10, approximately 360,000 specimens were received. The Trust serves Lambeth, Southwark and Lewisham with a population of 700,000.

King's College Hospital is a teaching hospital and centre of excellence with 950 in-patient beds, several specialist units of international repute, and units offering regional or supra-regional services. The Trust has a budget of £471 million, employs 6,000 staff with over 1 million patient contacts annually (including 85,000 A&E patients). King's College Hospital works closely with other healthcare organisations, such as local Primary Care Trusts. We also have strong academic links with King's College London together with our Academic Health Sciences Centre partners, Guy's and St Thomas' and the South London and Maudsley NHS Foundation Trusts.

The regional and supraregional services include:

- Haemato-oncology (including UK's largest bone marrow transplantation unit)
- Institute for Liver Studies (providing 30% of UK liver therapy, including transplantation and liver failure) for both adult and paediatric hepatology
- Variety Club Children's Hospital
- Regional Neurosciences and Neurosurgical services
- Renal unit (offering dialysis including HBV, HCV and HIV positive individuals)
- Harris Birthright Centre for Foetal Medicine
- Adult Intensive Care Unit, neonatal and paediatric intensive care
- Solid tumour oncology / cancer services (including skin, hepatobiliary, head and neck)
- Cardiac surgery (regional)

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- Paediatric and adult Accident and Emergency departments
- Obstetrics and gynaecology; assisted conception
- Genito-urinary medicine (Caldecot Centre)
- Reproductive and Sexual Health

1.3 Primary care

The laboratory serves more than 100 fund-holding general practitioners. The response of King's College Hospital to the evolving Primary Care Groups and Trusts has imposed further demands on the clinical virology service. The King's local Primary Referral Guide reflects general practitioners' expectations of direct access for prompt clinical advice, including virological advice. Specialist virology services and clinical advice are offered by virology to the London Boroughs of Lambeth, Southwark, Lewisham, Bromley, Bexley and Greenwich. The local authorities serviced by the laboratory are the London Boroughs of Bromley, Southwark, Bexley and Greenwich.

1.4 South London Specialist Virology Centre

Virology includes two Consultant Virologists, two Registrars, and a Principal Clinical Scientist and a team of other Healthcare Scientists, supported by a Health & Safety Manager, a Quality Manager and Administrative & Clerical team. The team manages a workload of over **150,000** samples of increasing complexity and requiring sophisticated laboratory processing and interpretation. The routine diagnostic work includes general serology, hepatitis and retrovirus serology, and an extensive repertoire of molecular based tests which include both qualitative and quantitative assays as well as antiretroviral resistance testing using automated sequence analysis, employing a range of different platforms and technologies underpinned by our Service Development research. General microbiology serological tests which are sent to the combined department are carried out in virology. In addition, all *Chlamydia trachomatis and N gonorrhoea* tests are carried out by APTIMA (TMA) technology on genital swab / urine samples. Virology receives specimens from a number of external microbiology laboratories for investigation.

1.5 Research

There are collaborations between clinical research groups and academic partners at Guy's, King's, and St Thomas' School of Medicine, the Institute of Psychiatry, The School of Nursing and Midwifery, King's College London and King's Division of Biomedical Sciences.

Research interests in the laboratory include:

The Principal Clinical Scientist, Dr Melvyn Smith along with team is involved in a variety of projects pertaining to technology transfer to the routine diagnostic service. Work carried out in the departments has been published in numerous peer-reviewed journals and presented at local, national and international meetings.

1.6 Surveillance activity in virology:

HIV HIV antiretroviral resistance data sequence for MRC database

HCV Reporting to the National HCV sentinel surveillance study

Norovirus Diagnosis and outbreak analysis

Influenza Diagnosis and typing

Pregnancy Screening in pregnancy

1.7 Key personnel and contact details

Virology direct lines: Results Phone: 020 3299 6155

Medical advice: 020 3299 6298

Microbiology Phone: 020 3299 3213 /020 3299 3565

Common Fax: 020 3299 3404

Virology only - 020 3299 9000 (switch) and extension				
Designation	Telephone extension			
Head of Virology - Consultant Virologist	Dr Mark Zuckerman	36298 / 36970		
Consultant Virologist	Dr M Sudhanva	36298 / 36971		
BMS 3 (Virology)	Ralph Henderson	36159		
Specialty Trainee	Vacant	36972		
Specialty Trainee	Dr Teresa Cutino	36973		

Microbiology only - 020 3299 9000 (switch) and extension				
Designation	Name	Telephone extension		
Clinical Director Consultant Microbiologist	Dr Jim Wade	33033		
Chairman Senior Lecturer	Dr John Philpott-Howard	33256		
Consultant Microbiologist	Dr Amanda Fife	33095		
Consultant Microbiologist	Dr lan Eltringham	33766		
Consultant Microbiologist and Antimicrobial Stewardship Lead	Dr Dakshika Jeyaratnam	32569		
Consultant Microbiologist and Infection Control doctor	Dr Surabhi Taori	334361		

Microbiology and Virology combined - 020 3299 9000 (switch) and extension				
Laboratory Manager	Mr Malcolm Goodwin	37777		
Principal Clinical Scientist	Dr Melvyn Smith	36155		
Laboratory Administrator	Mrs Linda Akkad	36260		
Virology Chief BMS	Ralph Henderson	36159		
Quality Manager	Mr Craig Smith	36140		

1.8 Normal laboratory opening times

Routine specimens are accepted at the virology laboratory from:

Monday to Friday: 9 AM to 5 PM

2. Use of the Laboratory

2.1 Test requesting procedure (routine, urgent and out of hours)

Routine requests can be made either by King's EPR system or any virology / microbiology request form. During normal laboratory hours please telephone urgent requests (020 3299 0000 and either extension 36155 or speak to the medical staff on 36298) to ensure priority processing. See section 3 for use of on-call services.

Please **DO NOT** call 36298 to obtain **RESULTS.**

2.2 Requesting Additional Tests

Sample Type	Time limit for requesting
	extra tests
Chlamydia trachomatis + N. gonorrhoae	14 days
Non-Blood Samples	
CSF	28 days
Fluids	28 days
Swabs	28 days
Faeces	28 days
Dry tissue (Skin and Nails)	28 days
Respiratory tract samples	28 days
Wet tissue	42 days
Whole blood samples	7 days
Plasma	
Medico-legal	30 Years
HIV viral loads (>40 cp/ml)	30 Years
Other plasma	14 days
Serum	
Other sera	~ 1 Year
Medico-legal	~ 2 Years
Pregnant booking	2 Years
Needlestick injuries	30 Years
Pre-transplantation sera	30 Years

All post-mortem tissues are returned to histopathology when testing is complete. For specific enquiries please contact the medical staff.

2.3 Completing the request form

EPR requests:

Most King's samples requests can be made via EPR (except Caldecot Centre and Occupational Health.) Please free text in the clinical details field if you can not see an intended test on EPR request.

Please telephone 36298 for specimens to be processed urgently.

Electronic requests from GPs

Many GP surgeries within the vicinity of King's send samples with electronic forms using T-Quest system. Please telephone 36298 for specimens to be processed urgently.

Types of paper based request forms:

Blue virology form ⇒ ⇒ ⇒ King's College Hospital usage

Yellow request form ⇒ ⇒ ⇒ Caldecot Centre / Reproductive and Sexual Health / GP / Antenatal

Multipart single pathology request ⇒ ⇒ GP use

Please telephone 36298 for specimens to be processed urgently.

The written form should be completed legibly. Please use labels whenever possible.

Minimum Required Data:

A request form must accompany all specimens sent to the laboratory. It should clearly state the following information.

Those in bold are a minimum requirement and without them the sample could be discarded or delayed.

- Patient name
- Unit number/NHS number
- Date of birth (age if DOB not known)
- Sex
- Ward or Address for report
- Requesting Medical Officer/GP name and number
- Date and time specimen taken
- Type of specimen (Specify anatomical site from which vesicle swab / fluid specimens were taken)
- Tests required

Other useful data

- Bleep number or mobile number, in order to phone results both before 5 PM and after 5 PM results
- Patient address
- All relevant clinical details including
 - any antimicrobial treatment (recent, current and intended)
 - History of foreign travel including return dates
 - Date of onset and duration of illness, particularly for serology
- Useful epidemiological information, e.g.:
 - Children and suspected influenza give the name of the school
 - Adults and suspected norovirus give the place and type of work, (e.g., catering, cruise liner
 - All patients and suspected viral haemorrhagic fever travel destination, date of return, signs and symptoms, malaria smear
- Viral Haemorrhagic Fever Risk status if applicable MUST BE DISCUSSED WITH MEDICAL VIROLOGY STAFF FOR RISK ASSESSMENT BEFORE SENDING SAMPLE

If uncertain about the exact test and terminology, please give a detailed clinical history as this can help the Virology medical staff to decide the most appropriate investigations.

2.4 Specimen labeling

- Use labels where ever possible.
- The specimen must be labelled with the same patient details as that on the request form
- Please ensure the full patient name, and the date of sample collection, are legible.
- Please note that unlabelled specimens do not guarantee authenticity of the sample and these cannot be processed and these may be discarded.

2.5 Specimen collection

The best results are obtained when an appropriate, well-taken specimen is in the proper container, is delivered to the laboratory promptly and relevant clinical information is provided on the request form. Please contact the laboratory if there is any doubt about the best specimen to take or you have questions about any test.

General guidelines on specimen collection include:

- Send specimens in sterile containers
- Collect specimens from the actual site of suspected infection. Please do not send just blood samples for 'viral serology' instead of vesicular fluid or throat swab or CSF, as the case may be.
- Take specimens that are representative of the disease process. For example, respiratory specimens are more appropriate than blood for serology in cases of acute respiratory infection.
- An adequate quantity of material should be obtained for complete examination especially vesicle fluid, CSF and NPA
- Take care to avoid contamination of the specimen by micro organisms normally found on the skin and mucus membranes. Sterile equipment and aseptic technique must be used for collecting specimens, particularly for those from normally sterile sites

- All swabs or material from swabs should be immersed in virus transport medium (VTM) and transported promptly to the laboratory. Viruses including (viral nucleic acid) may not survive prolonged storage at room temperature or may be overgrown by bacteria or fungi.
- In the absence of readily available VTM (laboratory issued or commercial), please immerse the swab tips in 1 ml of sterile saline in an universal container

2.6 Specimen limitations affecting assay performance

Factors that can affect assay performance are as follows

- inherent (age, gender, nutritional status, pregnancy, congenital immunological defects)
- acquired (passively acquired antibody, immune response to immunisation, immunosuppression)
- biological (lipaemic, haemolysed, high bilirubin content e.g. Liver ITU patients)
- collection (use of correct blood collection tubes e.g. serum from clotted blood may underestimate HIV-1 RNA load when compared to EDTA plasma)

2.7 Transport and receipt of specimens

During normal working hours, all routine King's specimens should be taken to central specimen reception at King's College Hospital.

For urgent testing bring the specimen DIRECTLY to the laboratory reception, which is on the 2nd floor of Cheyne Wing (opposite Liver ITU). *Please telephone 6298 for specimens to be processed urgently*.

<u>Do not</u> send samples to Central Specimen Reception for urgent testing, but contact laboratory or on-call BMS staff for directions before arranging urgent transport to laboratory reception.

See section 2.2.3 for samples referred from external institutions and laboratories.

2.8 Virology Cut-off times for receipt of specimens for a 24 hour TAT

If a sample needs to be processed urgently based on clinical ground, please contact the medical virologist on-call / on duty as detailed in section 3.3.

Virology cut-off times for processing samples for a 24-hour TAT					
Specimen type	Assays	Cut-off time for processing	Results available at:	Samples not processed on:	
Respiratory samples (combined throat and nasal swab, BAL, NPA, ETA etc.)	PCR for 11 respiratory viruses	12 noon	4.30 PM	Saturdays when there are no outbreaks in the community.	
Faeces	Norovirus RNA	10 AM	3.30 PM	Saturdays when there are no outbreaks in the community.	
CSF	HSV DNA, VZV DNA and enterovirus RNA. See below for CMV and EBV DNA [#]	10 AM	3.30 PM	Weekends and Bank Holidays.	
Vesicle fluid	HSV DNA, VZV DNA and enterovirus RNA	10 AM	3.30 PM	Weekends and Bank Holidays.	
EDTA whole blood	CMV DNA [#] , EBV DNA [#] and adenovirus DNA [#]	3 PM	1 PM next day	Weekends and Bank Holidays.	
EDTA plasma	HIV-1 RNA and HCV RNA	3 PM	1 PM next day	Weekends and Bank Holidays.	
Eye swab	HSV DNA, VZV DNA and adenovirus DNA	10 AM	3.30 PM	Weekends and Bank Holidays.	

[#]CMV DNA, EBV DNA and adenovirus DNA are assessed overnight during weekdays.

2.9 High risk specimens and safety

Pathogens are classified in hazard groups 1 to 4, with hazard group 1 being non-pathogenic to humans and hazard group 4 the most dangerous pathogens to humans.

All blood samples from suspected HIV and hepatitis patients are handled safely in the laboratory and consequently we do not require "DANGER OF INFECTION" labels. It is the responsibility of the sender to package samples safe enough for transportation according to regulations.

We assume all respiratory samples may potentially contain a hazard group 3 pathogen and treat samples accordingly.

Samples from patients with suspected viral haemorrhagic fevers, with a history of having returned within 21 days from Africa, Asia and South America are considered high risk. Contact virology medical staff before taking ANY sample. Special transport arrangements of these samples will be made in conjunction with Rare and Imported Pathogens Laboratory, PHE Public Health England Microbiological Services, Porton Down SP4 0JG

Great care must be taken in obtaining specimens. Equipment such as needles and blades must be immediately disposed of safely in locally approved "sharps" bins and NOT SENT TO LABORATORIES. Specimens should be transported to the laboratory without delay.

2.10 Courier and postal deliveries

When sending samples from an external institution or laboratory, it is the responsibility of the sender to ensure that the samples are packed in accordance with the current postal regulations, contain appropriate paper work and are labelled correctly. Courier / taxi / suitable transport should be arranged by sending institution or laboratory. You may have to contact the on-call BMS staff for out-of-hours' testing to indicate approximate time of arrival of sample at virology. Our experience shows that considerable amount of time is wasted by our on-call BMS staff just waiting for a sample to arrive because of lack of communication from test requesting person.

2.11 Printed results

Electronic reports are exported to EPR within 100 seconds of authorising.

Non-EPR reports are printed and dispatched every working day — Monday to Friday. The speed of reporting depends on the frequency of testing and the urgency of the request.

Copies of printed reports can be obtained by phoning extension 36155. Reports are <u>never</u> faxed. PDF of reports can be requested to @nhs.net email address in few select cases.

2.12 Telephoned results

Results of urgent requests and rapid requests that may aid the immediate patient management will be telephoned. Examples include:

- Respiratory virus detection by PCR in a respiratory sample.
- CSF nucleic acid test positive result
- Any acute infection diagnosis
- Blood borne virus infection such as new HIV, acute HBV, acute HAV and HTLV

2.13 Visitors

Visitors should introduce themselves at the laboratory reception (2nd floor, Cheyne Wing, opposite Liver ITU). The person they wish to see will come to meet them. It is best to make appointments in advance to ensure the right person is available.

2.14 Issue of VZIg and HBIg

The laboratory has limited stocks of varicella zoster immunoglobulin (VZIg) and hepatitis B immunoglobulin (HBIg). These are issued by the medical staff on a case-by-case basis and urgent laboratory testing if necessary.

The laboratory personnel cannot dispatch immunoglobulin by a taxi / courier or administer it.

It is the responsibility of the health care providers who require the immunoglobulin to:

- collect the immunoglobulin from Lab Reception, 2nd floor Cheyne Wing (opposite Liver ITU), King's College Hospital and
- administer the immunoglobulin

3. Out of hours' service

3.1 Out of hours' examinations provided in virology

An on-call service is provided by the Virology from 5pm to 9am Monday to Friday, all day Saturday, Sunday and bank holidays. For urgent specimens, contact the doctor or BMS on-call through the KCH switchboard (020 3299 9000).

Transplant coordinators should contact the King's College Hospital switchboard (020 3299 9000) and ask for the on call biomedical scientist.

Urgent / out of hours requests in virology

Assays / samples	Specimen type	Turn around time
HIV antibody /antigen	10 ml clotted blood	2 hours
Hepatitis A virus IgM	10 ml clotted blood	2 hours
Hepatitis B surface antigen	10 ml clotted blood	2 hours
Hepatitis B core total antibody	10 ml clotted blood	2 hours
Hepatitis B core IgM antibody	10 ml clotted blood	2 hours
Hepatitis B surface antibody	10 ml clotted blood	2 hours
Hepatitis C virus antibody	10 ml clotted blood	2 hours
HTLV I / II antibody	10 ml clotted blood	3 hours
CMV IgG antibody	10 ml clotted blood	2 hours
Treponemal antibody	10 ml clotted blood	2 hours
Toxoplasma IgG	10 ml clotted blood	2 hours
Measles virus IgG	10 ml clotted blood	4 hours
Norovirus RNA by PCR	Faeces	5-29 hours
Respiratory virus detection by PCR	Any respiratory sample	5-29 hours

Other tests can be carried out after discussion with one of the medical virologists.

3.2 Medical advice regarding the diagnosis and treatment of infection

During weekdays from 9 am to 5 pm medical advice on interpretation of virology results, antiviral management, blood borne virus exposure incidents and post exposure prophylaxis or any other relevant clinical circumstance can be sought from the virology specialist registrars or consultants on extension 36298.

During the day infection control advice can be obtained from the Micro Web or from the Infection Control Nurses on extension 4374.

Please **DO NOT** call 36298 to obtain **RESULTS** (for results call 3**6155**).

3.3 Out of hours' advice

An on-call service is provided by the Virology from 5pm to 9am Monday to Friday, all day Saturday, Sunday and bank holidays. To arrange for samples to be sent to please contact the King's College Hospital switchboard (020 3299 9000) and ask for the on call biomedical scientist. The on-call biomedical scientists will not look up results out of hours unless discussed and agreed with the medical virology staff.

Out of hours - a Specialist Registrar and/or Consultant Virologist can be contacted via KCH switchboard to discuss clinical, diagnostic and therapeutic problems with doctors.

Appropriate specimens for urgent examination on-call include:

- Urgent nasopharyngeal aspirate / BAL investigation for respiratory viruses
- Urgent duplicate combined throat and nasal swabs for avian influenza A H5N1 virus
- Screening of organ transplant donors
- Screening of organ transplant recipients who are urgently listed
- Returning travellers on renal dialysis
- Other tests are available after consultation with medical virology staff
- Faeces for norovirus RNA

4. Sections within the virology

Within the Virology Department there are two diagnostic sections viz. Virus Identification Section and Virus serology.

4.1 Virus Identification Section

Virus detection by molecular techniques is now a routine service for many viruses. Some are also performed during winter over the weekend. Please note that Virus isolation is now rudimentary and we no longer provide routine virus isolation service or direct immunofluorescence (DIF). Samples for electron microscopy (EM) are referred to another laboratory if needed.

Molecular diagnostics use real time PCR for the detection of viral nucleic acid in clinical samples. Nucleic acid tests performed routinely are for the detection and quantification of:

Blood-borne viruses (HCV RNA, HIV-1 RNA, CMV DNA and EBV DNA)

Influenza viruses

Other body sites/fluids (HSV DNA, VZV DNA, adenovirus DNA, enterovirus RNA)

Norovirus RNA testing of faeces

C. trachomatis & N gonorrhoea nucleic acid.

Multiplex PCR's are available for the following viruses as routine testing of all respiratory samples along with stand alone adenovirus DNA testing:-

multiplex 1: influenza A virus, influenza B virus and human metapneumovirus,

multiplex 2: RSV subgroup A and B, rhinovirus

multiplex 3: parainfluenza viruses 1, 2 and 3

multiplex 4: influenza A virus CDC matrix and pandemic influenza A H1N1 (2009)v H1

Antiretroviral resistance testing in HIV by nucleic acid sequencing and HBV DNA sequencing for resistance testing are also provided.

4.2 Virus serology

The virus serology section detects antibodies to viruses and viral antigens in serum using automated assays. In addition, a few bacterial antibodies are also assayed in this section. Automated machines include TECAN, Abbott Architect and VIDAS.

Blood Sciences Laboratory, Viapath www.viapath.co.uk

The Blood Sciences Laboratory (BSL) of Viapath completed in October 2006, is one of the largest, integrated, automated laboratories in Europe, ensuring Viapath remains at the forefront of developments in Pathology. Between Dec 2007 and March 2012, some virology markers were assessed in BSL but have now been repatriated back to the Virology department.

5. List of examinations performed in virology

5.1 Viral nucleic acid tests on non- blood samples

Specimen type	Tests performed	Schedule	Turn round time from receipt of sample during working week
Broncho-alveolar lavage, Combined nose and throat swab, Nasopharyngeal aspirate, endotracheal aspirate	Respiratory PCR: Influenza A virus Influenza B virus Parainfluenza 1 virus Parainfluenza 2 virus Parainfluenza 3 virus Respiratory syncytial virus (RSV) subgroup A Respiratory syncytial virus (RSV) subgroup B Rhinovirus Human metapneumovirus Adenovirus	Daily (week days). Weekends except Saturdays and Bank Holidays when there are no outbreaks.	2 – 4 hours
	Influenza A virus Oseltamivir resistance H275Y mutation	Whenever necessary	
Genital swab / Endocervical swab or urine or urethral swab	HSV 1&2 PCR (isolate for HSV drug resistance testing)	Daily (week days)	2 – 4 hours
Genital swab / Endocervical swab or urine or urethral swab	Chlamydia trachomatis and Neisseria gonorrhoea	Daily (week days)	3 - 5 days
Eye / conjunctival / corneal Swab	HSV DNA Adenovirus DNA VZV DNA	Daily (week days)	1 - 3 days
Eye / conjunctival / corneal Swab	Chlamydia trachomatis	Daily (week days)	1 - 3 days
CSF	CMV DNA EBV DNA HSV DNA Enterovirus DNA VZV DNA	Daily (week days)	1 - 3 days
CSF	JC virus DNA	Send away	7 - 14 days
CSF	HIV-1 RNA	Daily (week days)	5 - 14 days
Faeces (Stool)	Norovirus RNA	Daily (week days)	1 - 3 days
Urine / Blood	CMV DNA Adenovirus DNA BK virus DNA	Daily (week days) Daily (week days) Send away	7 - 14 days
Skin Swab	HSV DNA VZV DNA Enterovirus RNA	Daily (week days)	1 - 3 days
Skin vesicle	HSV DNA VZV DNA	Daily (week days)	1 - 3 days
Tissue / Biopsies	Site specific investigations	Daily (week days)	1 - 3 days

Specimen type	Tests performed	Schedule	Turn round time from receipt of sample during working week
Oracol mouth swab (Saliva)	Measles virus RNA		
	Mumps virus RNA	Send away	7 - 14 days
	Rubella virus RNA		
Please note the virus cu	lture and isolation and (direct immunofluores	cence are no longer

Please note the virus culture and isolation and direct immunofluorescence are no longer routine assays.

5.2 Viral nucleic acid tests on blood samples

Molecular assays on blood			
Test	Specimen type	Schedule	Turn round time from receipt of sample during working week
CMV DNA - quantitative	EDTA	Daily (week days)	2 - 4 days
EBV DNA - quantitative	EDTA	Daily (week days)	3 - 5 days
VZV DNA	EDTA	Daily (week days)	1- 3 days
HSV 1 and 2 DNA	EDTA	Daily (week days)	1 - 3 days
Enterovirus RNA	EDTA	Thrice a week	2 - 7 days
Adenovirus DNA	EDTA	Daily (week days)	3 - 5 days
HCV RNA viral load	EDTA / clotted	Thrice a week	5 - 14 days
HIV-1 RNA viral load	EDTA	Daily (week days)	5 - 14 days
HIV-1 proviral DNA (paediatric patients)	EDTA	Sent away	14 – 28 days
HIV-2 proviral DNA (paediatric patients)	EDTA	Sent away	14 – 28 days
HIV-1 antiretroviral resistance (inform medics for urgent testing)	EDTA	Weekly	14 – 28 days
HIV-2 antiretroviral resistance (inform medics for urgent testing)	EDTA	Sent away	14 – 28 days
HBV DNA antiviral drug resistance	EDTA blood	Whenever necessary	14 – 28 days
HIV-1 CCR5 / CXCR4 tropism assay trofile	EDTA blood	Weekly	On request with relevant clinical details
assay			relevant chilical details

5.3 Virus serology including bacterial serology

Test	Primary test performed at	Clotted blood	Schedule	Turn round time during working week
Hepatitis A virus IgM	Virology	10 ml	Daily	2 days
Hepatitis A virus IgG	Virology	10 ml	Daily	3 days
Hepatitis B surface antigen	Virology	10 ml	Daily	2 days
Hepatitis B surface antigen confirmation by neutralisation	Virology	10 ml	Daily	3 days
Hepatitis B core total antibody	Virology	10 ml	Daily	2 days
Hepatitis B core IgM	Virology	10 ml	As required	3 days
Hepatitis B e antigen	Virology	10 ml	As required	3 days
Hepatitis B e antibody	Virology	10 ml	As required	3 days
Hepatitis B surface antibody	Virology	10 ml	Daily	2 days
Hepatitis C virus antibody	Virology	10 ml	Daily	2 days
HIV-1 and 2 antibody / antigen	Virology	10 ml	Daily	2 days
HIV antibody - confirmation	Virology	10 ml	Daily	2 days
HIV-1 and 2 antibody on saliva	Virology	Saliva (not blood)	Weekly	10 days
HTLV 1 antibody	Virology	10 ml	Weekly	3 days
HTLV 1 antibody – confirmation (immunoblot)	Virology	10 ml	As required	7 days
CMV IgG	Virology	10 ml	2- 3 times a week	3 days
CMV IgG avidity	Virology	10 ml	As required	3 days
CMV IgM	Virology	10 ml	2 - 3 times a week	3 days
EBV VCA IgG	Virology	10 ml	2 - 3 times a week	3 days
EBV VCA IgM	Virology	10 ml	2 - 3 times a week	3 days
EBV VCA IgM – alternate assay	Virology	10 ml	As required	4 days
HSV IgG	Virology	10 ml	As required	7 days
HSV antibody type specific IgG	PHE- Colindale	10 ml	As required	14 days
Measles virus IgM	PHE - Colindale	10 ml	As required	1 – 14 days
Measles virus IgG	Virology	10 ml	As required	1 – 14 days
Mumps virus IgM	PHE - Colindale	10 ml	As required	1 – 14 days
Mumps virus IgG	Virology	10 ml	As required	1 – 14 days
Parvovirus B19 IgM	Virology	10 ml	1 - 2 times a week	4 days
Parvovirus B19 IgG	Virology	10 ml	1 - 2 times a week	4 days
Rubella virus IgM	Virology	10 ml	2- 3 times a week	3 days
Rubella virus IgG	Virology	10 ml	Daily	2 days
Varicella zoster virus IgG	Virology	10 ml	2 - 3 times a week	2 days

Test	Primary test performed at	Clotted blood	Schedule	Turn-around time during working week
Respiratory Complement Fixation tests				
CFT - Influenza A virus antibody	Virology	10 ml	Once a week	7 – 14 days
CFT - Influenza B virus antibody	Virology	10 ml	Once a week	7 – 14 days
CFT - Adenovirus antibody	Virology	10 ml	Once a week	7 – 14 days
CFT - RSV antibody	Virology	10 ml	Once a week	7 – 14 days
CFT - Q fever antibody	Virology	10 ml	Once a week	7 – 14 days
CFT – Chlamydia genus antibody	Virology	10 ml	Once a week	7 – 14 days
CFT - Mycoplasma antibody	Virology	10 ml	Once a week	7 – 14 days
Bacterial serology – testing service provide	ed by virology, clii		by microbiology	
Treponemal (syphilis) total antibody**	Virology	10 ml	Daily	2 – 5 days
Treponemal (syphilis) RPR	Virology	10 ml	Daily	2 – 5 days
Treponemal IgM	Virology	10 ml	Daily	2 – 5 days
Toxoplasma IgG antibody	Virology	10 ml	3 - 4 times a week	2 – 14 days
Anti-streptolysin O (ASO)	Virology	10 ml	Once a week	7 days

^{*}One 10ml clotted blood sample is usually sufficient for multiple serology tests

5.4 Serology battery tests

For certain patient groups the following tests will be performed:

Requests	Tests Performed
Acute hepatitis (jaundice, raised / abnormal LFTs)	HAV IgM, HBV surface antigen, HCV antibody – routine.
	EBV VCA IgM, CMV IgM and HEV IgM – are optional.
Miscarriage / TORCH	CMV IgM, CMV IgG, rubella virus IgM, rubella virus IgG, parvovirus B19 IgM, parvovirus B19 IgG (and CMV IgG avidity if IgG is positive)
Previous / past hepatitis	HAV IgG, HBV core total antibody, HCV antibody
Viral screen before transplantation	HCV antibody, CMV IgG, EBV VCA IgG, treponemal IgG, Toxoplasma IgG, HTLV 1 and 2 IgG, VZV IgG, HBV surface antigen, HBV core total antibody, HIV 1 and 2 antibody/antigen
Pregnancy screening (booking blood)	Treponemal antibody, HBV surface antigen, rubella virus IgG and HIV 1 and 2 antibody

6. Examinations referred to other laboratories

6.1 Virus isolation referrals

Test	Sample type	Reference Laboratory	Comments
Enterovirus typing	Enterovirus RNA positive sample	Centre for Infection, HPA, Colindale	Routinely sent to characterise the type of enterovirus
HSV antiviral resistance testing by phenotyping	HSV DNA positive original vesicle fluid / swab	Centre for Infection, VRD PHE, Colindale	On request if clinically indicated. Isolation of HSV in cell culture is required. Contact medical staff for advice
HSV antiviral resistance testing by genotyping	HSV DNA positive original sample	Centre for Infection, VRD PHE, Colindale	On request if clinically indicated. Contact medical staff for advice
Influenza typing	Influenza virus RNA positive original sample	Centre for Infection, VRD PHE, Colindale	Routinely sent as part of national surveillance of the isolation of influenza

6.2 Virus serology and molecular referrals

Virus	Sample type	Laboratory method		ethod	Reference Laboratory	Comments	
			IgG	PCR	,		
Dengue and other flaviviruses	10ml clotted blood	٧	٧	٧	Rare and Imported Pathogens Laboratory, HPA Microbiology Services, Porton	On request with relevant travel and clinical details. Contact medical staff for advice	
Haemorrhagic fever viruses	10ml clotted blood	٧	٧	٧	Rare and Imported Pathogens Laboratory, HPA Microbiology Services, Porton	Contact medical staff for advice	
Hantaan virus	10ml clotted blood	٧	٧	٧	Rare and Imported Pathogens Laboratory, HPA Microbiology Services, Porton	On request with relevant travel and clinical details. Contact medical staff for advice	
Hepatitis E virus (HEV)	10ml clotted blood	٧	٧	٧	Centre of Infection Colindale	On request with relevant travel and clinical details	
HSV type-specific serology	10ml clotted blood		٧		Centre of Infection, Colindale	On request with relevant clinical details	
Measles virus IgG, IgM and RNA	10ml EDTA blood and CSF	٧	٧	٧	Centre for Infection, Colindale	On request with relevant clinical details. Contact medical staff for advice	
	Virology has Oracol kits within King's.	٧	٧	V	Some kits are available in virology for local King's Hospital use via EPR. Inform virology.		
	patients, Oracol saliva kit available from HPU 020 3049 4280		-	-	Sample posted by patient / clinical staff to Centre for Infection, Colindale.		

		Laboratory method		nethod			
Virus	Sample type	IgM	IgG	PCR	Reference Laboratory	Comments	
Mumps virus IgG, IgM and RNA	10ml clotted blood	٧	٧	٧	Centre for Infection, Colindale	On request with relevant clinical details	
	Virology has Oracol kits within King's. For GP patients, Oracol saliva kit available from HPU 020 3049 4280	٧	٧	٧	Some kits are available in virology for local King's Hospital use via EPR. Inform virology. Sample posted by patient / clinical staff to Centre for Infection, Colindale.	use	
Polio	10ml clotted blood		٧	٧	Centre for Infection, Colindale	On request with relevant clinical details	
Q-fever (C. burnetii)	10ml clotted blood	٧	٧	٧			
Rabies	10ml clotted blood	٧	٧	٧			
Rickettsial antibody (Spotted fever group and epidemic typhus group)	10ml clotted blood	٧	٧	٧	Rare and Imported Pathogens Laboratory, HPA Microbiology Services, Porton	Contact medical staff for advice. On request with relevant clinical details	
West Nile virus, Japanese encephalitis virus, St Louis encephalitis virus	10ml clotted blood	٧	٧	٧			

6.3 Viral nucleic acid test referrals

Test	Sample type	Reference Laboratory	Comments
Adenovirus DNA - quantitative	10ml EDTA blood	Virology	Known adenovirus DNA positive samples only. This is a temporary arrangement before in-house assay is introduced
BK virus DNA quantitative assay in all other patients	10ml EDTA blood and urine	St Thomas's Virology	On request with relevant clinical details. Contact medical staff for advice
HTLV proviral DNA PCR	10ml EDTA blood	Centre for Infection, VRD PHE Colindale	On request with relevant clinical details. Contact medical staff for advice
JCV DNA PCR	CSF	Centre for Infection, VRD PHE Colindale	On request with relevant clinical details
Measles virus RNA PCR	10ml EDTA blood, saliva, urine, CSF Virology has Oracol kits within King's.	Centre for Infection, VRD PHE Colindale Some kits are available in virology for local King's Hospital use via EPR. Inform virology.	On request with relevant clinical details. Contact medical staff for advice
	For GP patients, Oracol saliva kit available from HPU 020 3049 4280	Sample posted by patient / clinical staff to Centre for Infection, Colindale.	
Mumps virus RNA PCR	Saliva, urine, CSF.	Centre for Infection, VRD PHE Colindale	On request with relevant clinical details. Contact medical staff for advice
	Oracol saliva kit available from Health Protection Unit: 020 3049 4280	Sample posted by GP patient to Centre for Infection, Colindale. Within Kings, this test is available on EPR	Sample posted by patient / clinical staff to Centre for Infection, Colindale
Parvovirus B19 DNA PCR	10ml clotted blood	Centre for Infection, VRD PHE Colindale	On request with relevant clinical details. Contact medical staff for advice
Rubella virus RNA PCR	10ml EDTA blood, saliva, urine, amniotic fluid	Centre for Infection, VRD PHE Colindale	On request with relevant clinical details. Contact medical staff for advice
HBV DNA viral load in Health Care Worker	10ml clotted blood	PHE Birmingham	On request with relevant clinical details
HIV-2 RNA load	2 x 10 ml EDTA blood	UCLH	On request with relevant clinical details. Contact medical staff for advice
HIV-2 RNA resistance	10ml clotted blood	PHE Birmingham	On request with relevant clinical details
HIV-1 antiretroviral resistance testing by phenotyping and virtual phenotyping	10ml EDTA blood	Virology	On request with relevant clinical details. Contact medical staff for advice. (Turnaround time: 4 weeks)

6.4 Antiviral assays

Test	Sample type	Reference Laboratory	Comments
Acyclovir and	10 ml clotted blood	Antimicrobial Reference Lab,	On request.
ganciclovir plasma	Label the specimen	Bristol	Contact medical staff for advice
levels	containers with time		with relevant clinical details
	of previous dose.		

7. Specimen collection methods

Aspirates and fluids from normally sterile sites

Collect the specimen with a sterile syringe. Transfer a maximum of 20ml into a sterile universal container. Ensure the cap is tightly screwed on.

Biopsies

All biopsies should be placed in sterile saline and not in formalin. Please state clearly the clinical diagnosis and the test needed. It may be better to send a sample for histopathology than for virology.

Bronchial washings

After collection remove the cap and the tubing of the sterile suction container and apply the screw cap to the container.

Bronchoalveolar lavage

A specialist will collect the specimen in a sterile container according to local protocol. Traps containing a specimen should be sealed using a loop of tubing.

Cerebrospinal fluid

An adequate amount is essential - send at least 2-3ml.

Cervical swabs

Chlamydia trachomatis and Neisseria gonorrhoea:

Take a *Chlamydia* specimen after the Microbiology swab. If there is discharge, mucus or pus in the cervix, then wipe it off. Insert the Chlamydia swab into the endocervical canal. Rotate firmly around the surface of the canal for 5-10 seconds; withdraw the swab without touching any vaginal surface. Place the swab in APTIMA sample collection tube, snip off the shaft and screw the cap on.

Neisseria gonorrhoea

This test is part of combined *Chlamydia trachomatis and Neisseria gonorrhoea* testing on the same sample using TMA technology by APTIMA

Virology:

Moisten the swab in sterile saline before taking the specimen. Never moisten swab in VTM. Follow procedures as for the Microbiology swab. Snap off the swab tip into VTM.

Ear swab

Place the swab in the ear canal over any vesicle. Rotate gently over the vesicles / ulcers. Place the swab in VTM.

Eye swab

Chlamydia trachomatis:

The aim is to collect epithelial cells and assess *Chlamydia trachomatis* nucleic acid by TMA. Scrape from conjunctiva and place the swab in APTIMA sample collection tube, snip off the shaft and screw the cap on.

Virology:

Moisten the swab in sterile saline before taking the specimen. Never moisten swab in VTM. Snap off the swab tip into VTM.

Faeces

Send a "plum-sized portion" or 5-10ml if liquid. Ask the patient to defecate into a clean bedpan or other convenient container if at home. Use the plastic spoon to transfer a portion of faeces into the pot. For liquid faeces use a plastic medicine spoon. Take care not to contaminate the outside of the faeces pot.

Genital tract swabs

Chlamydia trachomatis and Neisseria gonorrhoae:

The aim is to collect epithelial cells and assess nucleic acid by APTIMA (TMA). Scrape from the endothelium and place the swab in APTIMA collection tube, snip off the shaft and screw the cap on.

Virology:

Moisten the swab in sterile saline before taking the specimen. Never moisten swab in VTM. Snap off the swab tip into VTM.

High vaginal swabs

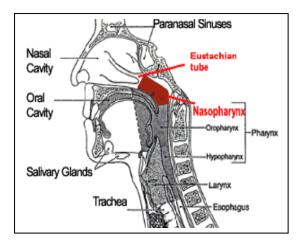
Introduce the speculum. Roll the swab firmly over the surface of the vaginal vault. Place the swab in VTM.

Mouth swabs

Sample any vesicle lesions or ulcers or inflamed areas. A tongue depressor or spatula may be helpful to aid vision and avoid contamination from other parts of the mouth. Place the swab in the VTM.

Nasopharyngeal aspirate (NPA)

Aspirate collected from the nasopharynx (see diagram) is ideal for viral upper respiratory tract infection as it contains epithelial cells, which harbour viruses.



Materials: Suction pump, sterile suction catheter (usually size 10, smaller for infants), sterile container usually 30ml urine jar, sputum pot or sterile container.

- 1. Attach catheter to suction pump, leaving wrapper on suction catheter. Turn on suction and adjust to a pressure of 100-150 mmHg.
- 2. Without applying suction, insert catheter into the nose, directed posteriorly and towards the opening of the external ear. Stop when you feel a resistance (you have reached the posterior nasopharynx).
- 3. Apply suction
- 4. Using a rotating movement, slowly withdraw catheter. Catheter should remain in nasopharynx no longer than 10 seconds.
- 5. Disconnect suction. Put the catheter in a sterile universal container and label.
- 6. Send to virology without delay. (Phone virology if you are sampling after 3pm or "out-of-hours".

Serology

Collect 10ml of blood in a blood collection tube (gold topped). Heparinised blood (green topped) may cause non-specific reactions in some antigen / IgM assays, but we can use this sample for routine serology and may lead to delays in reporting results.

Slides for immunofluorescence

NB: this is not a sensitive method and we no longer provide this service.

Sputum

Induced sputum or expectorated sputum can be used for virological assessment.

Do not collect shortly after the patient has been eating, drinking or cleaning their teeth. Ask a physiotherapist to assist if a patient has difficulty in producing satisfactory specimens.

Surface swabs and skin swabs

Rotate the swab on or in the required site. Place the swab in the VTM.

Throat swabs

Moisten the swab in sterile saline before taking the specimen. Never moisten swab in VTM. Snap off the swab tip into VTM. A combined throat and nose swab can also be taken (see above).

Throat & nasal swabs – (combined throat and nose swab)

This is the preferred to stand-alone throat swab for respiratory infections. Moisten the swab with sterile saline (never with VTM) before swabbing. Using two sterile saline moistened swabs, swab throat and nose sequentially. For nose - swab the anterior nares by gently rotating the swab in each nostril. Snap off the swab tips into VTM.

Tissues and biopsies

Under aseptic conditions transfer material to a sterile universal container that does not contain formalin as this inactivates pathogens very rapidly. Send in 0.5ml of sterile saline. Please specify which virus is being investigated for virology.

Urethral swabs

Avoid contamination with microorganisms from the vulva or the foreskin. Small swabs are available for this purpose.

Chlamydia:

Take this specimen after the Microbiology swab. Pass the swab through the urethral meatus and gently but firmly roll it over all the surfaces of the urethral epithelium for 1-2 seconds then withdraw. Snip off the swab into APTIMA sample collection tube.

Urine

Clean-voided midstream urine is preferred for virology.

It is recommended that in females the hands and the perineal area should be washed with soap and water prior to specimen collection. Part the labia and clean the area around the urethral meatus from front to back. Spread the labia with the fingers of one hand.

In males retract the foreskin, if present, and clean the skin surrounding the urethral meatus.

Start passing urine into the toilet, bedpan or urinal. When the urine is flowing freely, collect urine in a clean sterile container.

Special urine collection pouches are needed for collection in paediatric patients.

First catch urine

For Chlamydia trachomatis and Neisseria gonorrhoea, this is needed rather than mid-stream urine. The first 5-10 ml of voided urine is to be sent.

Vesicles, ulcers and genital lesions (see section 8)

Material required

Swabs preferably Dacron-tipped swabs with plastic shaft and viral transport medium (VTM): This is a pink fluid available from virology during normal working hours. Limited stocks may be available in the refrigerators in some wards and clinics. VTM has to be kept refrigerated and has a shelf life of 3 months at 4°C.

Method

Burst a vesicle using a sterile needle and collect with a swab or aspirate the fluid contents of the vesicle. Alternatively, scrape the base of the vesicle or ulcer with a swab so that cellular material is collected. Inoculate this fluid / cellular material into VTM.

Always state the site, distribution and nature of the vesicle. This is essential, as the laboratory may need to prioritise between HSV and VZV testing. In a case with suspected hand, foot and mouth disease we perform enterovirus RNA PCR.

8. Table of containers for transport of specimens Specimens should be transported and processed as soon as possible

Specimen/investigation	Container and comments
Aspirates from normally sterile sites (joint, ascites, peritoneal and pleural fluids)	Sterile universal container
Biopsies	Sterile container with sterile saline.
Blood	Serum in YELLOW topped container
	Whole blood in Purple / Mauve topped container (EDTA blood) (for CMV DNA, EBV DNA, adenovirus DNA, HIV pro-viral DNA)
	Plasma in Purple / Mauve topped container (EDTA blood) (HIV RNA, HBV DNA, HCV RNA, HIV genotypic resistance testing)
Bronchoalveolar lavage	Sterile container; e.g. 30ml sterile container or sterile universal container
and bronchial washings	
Cerebrospinal fluid (CSF)	Sterile universal container.
Cervical swab	For the investigation of Chlamydia trachomatis and Neisseria gonorrhoea use the APTIMA swab and the transport medium. Use swab in VTM for other viruses like HSV.
Conjunctival and corneal swabs	Use a Chlamydia swab in chlamydia transport medium APTIMA for Chlamydia trachomatis.
	Use swab in VTM for virology.
Ear swab	Send a swab in VTM.
Eye swab	Use a chlamydia swab in chlamydia transport medium APTIMA for
	Chlamydia trachomatis.
	Use swab in VTM for virology.
Faeces	With the spatula provided transfer a plum-sized portion of faeces, or equivalent volume of fluid, into a sterile universal container
High vaginal swab (HVS)	For Chlamydia trachomatis and Neisseria gonorrhoea investigations send swab in APTIMA transport medium
Mouth swab	Send the swab from lesion in VTM.
Nasal swab	Send the swab in VTM.
Nasopharyngeal aspirate	Traps containing a specimen should be sealed using a loop of tubing
Seminal fluid	Sterile universal container
Specimen/investigation	Container and comments
Sputum	Sputum from deep expectoration and not saliva is required. Send specimen in a 30ml sputum container or universal. It is usually difficult to perform respiratory PCRs on this sample.
Throat & nasal swab	Send the swab in VTM.
Tissues and biopsies	In at least 0.5 ml of sterile saline in sterile universal container.
Urine	Collect urine in a sterile universal container. Send first catch urine for Chlamydia trachomatis.
All other urine specimens	Sterile universal container
Urethral swab	For the investigation of <i>Chlamydia trachomatis and Neisseria gonorrhoea</i> use the APTIMA swab and the transport medium
Vesicles, ulcers and genital lesions	Send a swab in VTM.

• In the absence of readily available VTM (laboratory issued or commercial), please immerse the swab tips in 1 ml of sterile saline in an universal container