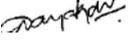


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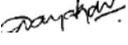
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1 Contact details:

Results and reception Tel no :	:	04712781212
Serology Tel no	:	04712781213
Isolation Tel no	:	04712781259

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2 Information for patients and users

Rajiv Gandhi Centre for Biotechnology, Bio-Innovation Centre (RGCB-BIC)
Thiruvananthapuram-695585

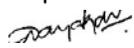
RGCB Offers PCR based testing for DNA and RNA viruses (CMV, EBV, HBV, HCV, HSV and H1N1) by qPCR and qRT PCR.

2.1 Working hours

Available 6 days a week

Monday to Saturday 9:00 a.m.-5:30 p.m.

S.No	Test Name	Patient Preparation	Type of sample	Type of container	Quantity of sample required	Stability	Sample retention time at the collection point	Method	Turnaround time (TAT)
1	Cytomegalovirus qPCR	None	EDTA Blood	Purple top Vacutainer	3ml	Whole blood collected, plasma separated and kept in - 80 for long	24 hrs	PCR	48-72hrs

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2	Epstein-Bar virus qPCR	None	EDTA Blood	Purple top Vacutainer	3ml	term storage Whole blood collected, plasma sepatated and kept in - 80 for long term storage	24 hrs	PCR	48-72hrs
3	Hepatitis B virus qPCR	None	EDTA Blood	Purple top Vacutainer	3ml	Separate Aseptically Plasma Within 1 hour of collection room NA, Refrigerated NA, Frozen 2 Weeks	24 hrs	PCR	48-72hrs
4	Hepatitis C virus qRT-PCR	None	EDTA Blood	Purple top Vacutainer	3ml	Separate Aseptically Plasma Within 1 hour of collection	24 hrs	PCR	48-72hrs

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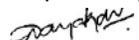
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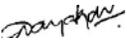
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						room NA, Refrigerated NA, Frozen 2 Weeks			
5	Herpes simplex virus 1 & 2 q PCR	None	EDTA Blood /CSF	Purple top Vacutainer	3ml	Whole blood collected, plasma separated and kept in - 80 for long term storage	24 hrs	PCR	48-72hrs
6	Influenza A/B and H1N1 (qRT PCR)	None	Nasal throat swab / throat	Viral Transport medium	5ml	Nasal throat swab samples refrigerated	24 hrs	PCR	24hrs
7	HIV1 qRT PCR	None	EDTA Plasma	Purple top Vacutainer	3ml	Whole blood collected, plasma separated and kept in - 80 for long term storage	24 hrs	PCR	24hrs

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Results and reception Tel no : 04712781212

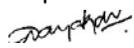
Isolation Tel no : 04712781259

List of tests offered by the Virology laboratory: Any test not listed – please phone laboratory for further instructions.

TEST	SAMPLE TYPE	SPECIAL INFORMATION
DNA OR RNA	EDTA Blood	Sample isolation and Preparation are Based on Kit Inserts. Corresponding kit inserts are used as Standard Operating Procedures
	Plasma	
	Urine	
	CSF	
	Swab	

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SPECIAL PATHOGENS

For special pathogens, please contact Virologist at lab or via e-mail system to discuss the case before sending specimens. In some cases, the reference laboratory will need to be notified in advance. Most cases of suspected viral haemorrhagic fever are due to other causes, and a clinical consultation may provide better information for both clinician and pathologist.

VIRUS	SAMPLE TYPE	SPECIAL INFORMATION
Viral haemorrhagic fevers	5ml EDTA blood and 5ml clotted blood	Consult with CLM
Arboviruses	Various	Consult with CLM

Laboratory Medicine and Molecular Diagnostics,

Tel: 04712529565

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3 Guide to Appropriate Specimens

3.1 General instructions:

All diagnostic information from the virology laboratory is contingent on the quality of specimen received. A poorly collected and/or poorly transported specimen can result in:

- Failure to isolate the causative virus, and
- Contamination with bacteria or fungi.
- Haemolysis of blood samples

Safety considerations with regard to the handling of specimens:

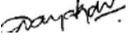
- Treat all specimens as potentially hazardous
- Do not contaminate the external surface of the collection container and/or its accompanying paperwork
- Minimize direct handling of specimens in transit from the patient to the laboratory. Ideally, specimens should be placed in plastic sealable bags with a separate pouch for the specimen request form.

Please ensure that samples are correctly labelled and that the request form is filled in with all the relevant data.

- The points listed below each specimen type are to enable clinicians, nursing staff, Lab Technician and patients to be able to take a good quality specimen.
- Clinicians, nursing staff, Lab Technician and patients are responsible for ensuring that these guidelines are followed.
- Please contact the laboratory if in any doubt as to the collection or transport of a specimen.

3.2 Urine specimens - collection and transport

Urine is normally a sterile body fluid. However, unless it is collected properly, it can become contaminated with micro-organisms from the perineum, urethra or vagina.

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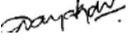
The following guidelines are provided to ensure proper specimen collection and subsequent, prompt delivery of urine samples to the laboratory.

3.3 Specimen collection

1. Midstream urine specimens (msu):

- ☐ The person obtaining the urine specimen should wash their hands with soap and water, rinse, and dry. If the patient is collecting the specimen, he/she should be given detailed instructions, including diagrams or a pictorial display.
- ☐ Females: Cleanse the urethral opening and the vaginal vestibule area with clean gauze pads soaked with sterile saline. Hold labia apart during voiding.
- ☐ Males: Cleanse the penis, retract the foreskin (if not circumcised), and wash with sterile saline. Keep foreskin retracted during voiding (to minimise contamination with skin flora).
- ☐ Both females and males: Allow a few millilitres of urine to pass (DO NOT STOP THE FLOW OF URINE) and collect the midstream portion of urine in a sterile container. In circumcised men, cleansing of the peri-urethral area does not improve the detection of bacteriuria and is therefore not necessary.

Collect voided urine directly into a sterile container; do not use a urinal or bedpan for collection.

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2. Catheter urine

A straight (non-indwelling) catheter is used by a physician to obtain urine directly from the bladder.

void contamination during urine collection from indwelling catheters.

This procedure is not routinely recommended because there is a risk of introducing microorganisms into the bladder.

Urine from an ileal conduit must be collected after removal of the external device and insertion of a catheter into the cleansed stoma.

Urine collected by suprapubic needle aspiration of the bladder avoids contamination associated with the collection of voided urine. This is the preferred method for infants and for patients for whom the interpretation of results of voided urine is difficult.

4 Specimen transport

Transport urine to the laboratory as soon as possible after collection.

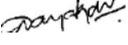
Urine specimens must be submitted for PCR within 2 hours after collection, or refrigerated within 24 hours whenever possible.

All specimen containers must be closed tightly to prevent leaking. If sample has grossly leaked from the container, the specimen will be rejected and not processed. If the specimen has leaked slightly,

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decontaminate the outside of the container with 70% alcohol prior to transport.

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5 Sterile body fluids including csf - collection and transport

A. CEREBROSPINAL FLUID (CSF)

Please note: CSF MUST BE COLLECTED PRIOR TO ANTIMICROBIAL THERAPY!

Collection considerations for Cerebral Nervous System (CNS) specimens:

Assay	Optimal volume	Comments
PCR	1-2ml	
Serology	1-2ml	NOT ideal specimen for serology.

Volumes are guidelines.

Greater volumes increase the chance of organism recovery.

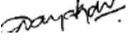
The laboratory, irrespective of the volume received, must process all CSF specimens.

CSF specimens should be transported to the laboratory promptly. Failure to do this may result in the non-viability of some viruses.

If prompt delivery is not possible CSF specimens should be kept at 4-8°C for viral PCR

CSF should not be added to viral transport medium.

The ideal tube for CSF specimens is a red-topped tube with no additives or clotting activators.

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6 Other sterile fluids

6.1 Vesicle fluid.

Vesicle fluid should be aspirated using a sterile technique, and inoculated into viral transport medium. Transport medium can be drawn up into the syringe and then expelled to flush the syringe and ensure that a maximum amount of vesicle fluid is obtained.

In the past, it has been permissible to use the aspirating syringe as the transport container provided the needle was capped. This practice is no longer acceptable because of the increased possibility of needle-stick injuries.

Other: Contact the virologist to discuss the clinical case and possible tests.

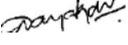
7 Specimen collection

Specimens should be collected with as little contamination from indigenous microbial flora as possible to ensure that the sample will be representative of the infected site.

Sterile equipment and aseptic technique must be used to collect specimens to prevent introduction of micro-organisms during invasive procedures.

In addition to routine information, it is essential that the patients' specimen label accurately reflects:

The specific body site from which the specimen was taken

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Provisional diagnosis

Collect specimens in sturdy, sterile, screw cap, leak-proof containers with lids that do not create an aerosol when opened.

8 Transport

8.1 Syringes:

Specimens obtained by a doctor using needle aspiration should be transferred to viral transport medium prior to transport to the laboratory. Transport medium can be drawn up into the syringe and then expelled to flush the syringe and ensure that a maximum amount of fluid is obtained.

8.2 Swabs

If a swab is taken it is essential that it be placed in viral transport medium. The swab should be placed into the bottle, and the shaft broken off. This will allow the bottle to close. Swabs for virological testing must not be put into the gel medium used for bacterial culture. Viral transport medium should be used instead.

8.3 Ulcers

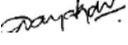
Specimens should be collected prior to the administration of antiviral therapy.

Swabs:

Remove overlying debris.

Vigorously swab or curette the base of the ulcer. Ulcer scrapings can be sent for culture.

If exudate is present from the ulcer, collect it with a syringe or a sterile swab.

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8.4 Tissue specimens:

Biopsies and tissue specimens:

Tissue should be sent in viral transport medium. If this is not available, use sterile water or saline. Do NOT use formalin.

GENERAL RECOMMENDATIONS FOR SPECIMEN COLLECTION FOR SEXUALLY TRANSMITTED DISEASES:

Cervical swabs: The cervix should be visualized via speculum examination and normal or inflammatory discharges should be removed with swabs. Swabs for Herpes Simplex Virus (HSV) should be collected from the ectocervix.

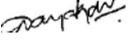
Genital Ulcer: Swabs should be used to obtain specimens from the ulcer base and placed into appropriate transport medium. If vesicles are also present in the same area, vesicle fluid may be collected after lancing the vesicle.

Vesicles: Vesicle fluid may be collected after lancing the vesicle, or aspirated from the vesicles.

8.5 **TRANSPORT**

All specimens should be transported to the laboratory promptly. Failure to do this may result in overgrowth of bacteria.

If prompt delivery is not possible specimens should be refrigerated at 4-8°C

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Syringes:

Specimens obtained by a doctor using needle aspiration should be transferred to viral transport medium prior to transport to the laboratory. Alternatively, and only if transferring it from the syringe will compromise the specimen, the doctor should remove the needle, using a protective device to avoid injury, and cap the syringe with a sterile cap prior to transporting it to the laboratory. If the latter procedure is followed it is essential that the specimen be submitted to the laboratory immediately after collection.

9 Sputum and respiratory tract specimens

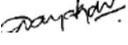
9.1 INTRODUCTION:

Infections of the lower respiratory tract are a major cause of morbidity and mortality. Diagnosis of these infections frequently is complicated by the contamination of specimens with upper respiratory tract secretions during collection.

9.2 SPECIMEN COLLECTION:

Specimens include sputum, tracheal aspirates, bronchial washings, bronchial brushes, bronchial biopsy specimens, bronchoalveolar lavage fluid, trans-tracheal aspirate, lung aspirate and lung biopsy specimens.

Throat (Pharyngeal specimens):

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Do not obtain throat samples if epiglottis is inflamed, as sampling may cause serious respiratory obstruction.

Depress tongue gently with tongue depressor.

Extend sterile swab between the tonsillar pillars and behind the uvula. (Avoid touching the cheeks, tongue, uvula, or lips).

Sweep the swab back and forth across the posterior pharynx, tonsillar areas, and any inflamed or ulcerated areas to obtain sample.

Nasopharyngeal swabs:

Carefully insert a swab through the nose into the posterior nasopharynx, and rotate the swab.

Nasopharyngeal aspirates

Attach syringe to tube and fill 5ml syringe with saline or viral transport medium. Instill saline into nostril and aspirate the recoverable nasal specimen immediately. Inject aspirated specimen into container containing virus transport medium

Tracheal aspirates

Broncheo-alveolar lavages

10 Guidelines for proper specimen transport:

All specimen containers must be tightly closed. Leaking specimens will compromise the quality of results.

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Specimens must be transported to the laboratory promptly. Failure to do this may result in the death of fastidious organisms and in overgrowth by more hardy bacteria.

If prompt delivery is not possible, specimens should be refrigerated at 4 – 8°C.

The longer the delay in reaching the laboratory, the lower the yield of virus due, and the less sensitive the culture.

11 Guidelines for blood specimens:

Please consult the list of tests to see which type of blood specimen is required.

In general, only two types of blood specimens are used – clotted blood for serology, and EDTA blood for other assays.

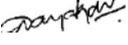
Serology – clotted blood (yellow or red-topped tube)

PCRs and viral loads done on blood samples – EDTA blood

Post-mortem blood samples are often haemolysed. Moderately haemolysed specimens might still be testable, but severely haemolysed specimens are often untestable.

11.1 Phlebotomy guide

This specimen sample manual is intended as a guide to all people taking specimens that are sent to the Laboratory Medicine and Molecular Diagnostics. This manual covers phlebotomy instructions as well as the correct sampling procedures for various other sample types. Please read the following instructions carefully before taking samples.

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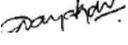
Please remember that all diagnostic information from our laboratory is dependent on the quality of specimen received.

11.2 General instructions:

This manual should be used as a training document and should be read and signed by all people who are responsible for taking samples that are to be sent to the Laboratory Medicine and Molecular Diagnostics.

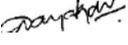
Please ensure that the correct procedure for the positive identification of the patient has been followed before taking any samples from a patient.

- The laboratory will not process samples if they are not labelled correctly.
- Do not pre-label samples—this may lead to erroneous labelling at times.
- Please ensure that laboratory specimens are stored out of direct sunlight.
- Please ensure that the correct sample container with correct anticoagulant (where relevant) is used. All the necessary information required is covered in this manual.
- Please ensure prompt, adequate mixing of blood samples taken into anticoagulant tubes (purple/blue top). These samples should be mixed by gently inverting at least 5 times. Do not shake! Failure to mix adequately may result in the sample clotting rendering it unsuitable for analysis.
- Please ensure that samples are stored safely for transport and handling.
- Please ensure that samples are not at risk to leak out or break, as the laboratory will not process these samples.
- Please check blood tubes for cracks before taking and sending samples.
- Please check expiry dates on tubes before taking specimens into tubes.

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- Please ensure that safety and infection control procedures are followed at all times.
- Please take note of the special precautions and storage instructions for certain tests. These are detailed under the relevant department doing the test.
- If a test requested is not covered in the sampling manual, please phone the laboratory for special instructions regarding correct specimen containers, special sampling procedures and requirements/ precautions to be taken.
- Please read the instructions at the beginning of each departmental section for individual tests as each department may have different instructions that need to be adhered to when taking certain samples types.
- Any after-request tests (tests not requested on original request form) must be telephonically requested with the relevant laboratory. The laboratory will inform you if the after-request can still be carried out.
- If in any doubt regarding any aspect of our service, please feel free to contact the Laboratory during working hours.
- For any additional test, the test request should be informed to the lab in writing, within 48 hours of sample reception in the lab
- Urgent test request will be accepted by the lab on the basis of verbal request by the clinicians, the test reports will be released on the same day. However, a written request needs to be communicated to the lab at the shortest possible time for documentation.

Following these instructions will ensure that a high level of service can be maintained by the Laboratory Medicine and Molecular Diagnostics to the benefit of our customers as well as to the patients.

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12 Routine venipuncture and specimen handling

12.1 Venipuncture procedure

The venipuncture procedure is complex, requiring both knowledge and skill to perform. Each phlebotomist generally establishes a routine that is comfortable for her or him. Several essential steps are required for every successful collection procedure:

Identify the patient.

Assess the patient's physical disposition (i.e. diet, exercise, stress and basal state).

Check the request form for requested tests, patient information and any special requirements.

Prepare the equipment, the patient and the puncture site.

Select a suitable site for venipuncture.

Perform the venipuncture.

Collect the sample in the appropriate container.

Label the collection tubes at the bedside or drawing area.

Assess the need for sample recollection and/or rejection.

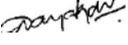
Recognise complications associated with the phlebotomy procedure.

Promptly send the specimens with the request form to the laboratory.

12.2 Patient identification

Verbal identification

Greet the patient and identify yourself.

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Always ask the patient to state his/her full name.

Never ask: "Are you vinod?"

Remember that many patients have a tendency to say yes to anything in the outpatients setting.

Ask the patient's date of birth and ask them to spell their names if you want to query the patient's identity.

Verifying Identification

Examining any of the following should follow verbal identification:

Identity book:

Wrist band (wards): All information on the wrist band should match the details provided on the request form. Note: a wrist band lying on the bedside table may NOT be used for identification.

Ankle band (paediatric& neonates)

Hospital/clinic card/book: should be inspected to confirm the patients name, hospital number, date of birth or age and doctor

Bed Number: a bed number on the request form cannot be used to identify ward patients.

13 Completing the request form

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A request form must accompany each sample submitted to the laboratory. This request form must contain the proper information in order to process the specimen. The essential elements of the request form are:

Patients surname and first name
 Patients hospital number, clinic number or ID number
 Patients date of birth and sex
 Requesting physicians complete name and contact number
 Person who took the specimen
 Date and time of collection
 Source and type of specimen
 Diagnosis
 Indicate the test(s) requested

13.1 Labelling the sample

Please note: the laboratory will not process unlabelled specimens. A properly labelled sample is essential to ensure that the results of the test match the patient. The essential elements in specimen labelling are:

Patient's surname and first name.

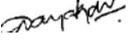
Patient's hospital number, clinic number or ID number.

Where available make use of the addressograph sticker provided.

14 Order of draw

Blood collection tubes must be drawn in a specific order to avoid cross-contamination of additives between tubes. Because blood is often taken for virology at the same time as for other disciplines, all the tubes are mentioned here. The recommended order of draw is:

First - blood culture bottles

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Second - non-additive tube (red stopper or SST)

Third - coagulation tube (light blue stopper). A light blue stopper (sodium citrate) tube is NEVER the first tube drawn. If a coagulation assay is the only test ordered, draw a non-additive tube (red stopper or SST) first, and then draw the light blue stopper tube.

Last draw - additive tubes in this order:

Heparin (dark green stopper)

Oxalate/fluoride (light grey stopper)

EDTA (lavender stopper)

NOTE: Tubes with additives must be thoroughly mixed (by gentle inversion and not shaking). Erroneous test results may be obtained when the blood is not thoroughly mixed with the additive. Overzealous mixing also results in haemolysis. Certain tests cannot be performed accurately with the presence of haemolysis.

15 Venipuncture site selection

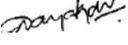
Although the larger and fuller median cubital and cephalic veins of the arm are used most frequently, wrist and hand veins are also acceptable for venipuncture.

Certain areas are to be avoided when choosing a site:

Extensive scars from burns and surgery - it is difficult to puncture the scar tissue and obtain a specimen.

The upper extremity on the side of a previous mastectomy - test results may be affected because of lymphedema.

Haematoma - may cause erroneous test results. If another site is not available, collect the specimen distal to the haematoma.

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Intravenous therapy (IV) / blood transfusions - fluid may dilute the specimen, so collect from the opposite arm if possible.

Cannula/fistula/heparin lock - hospitals have special policies regarding these devices. In general blood should not be drawn from an arm with a fistula or cannula without consulting the attending physician.

Oedematous extremities - tissue fluid accumulation alters test results.

15.1 Procedure for Vein Selection

Palpate and trace the path of veins with the index finger. Arteries that pulsate are most elastic and have a thick wall. Thrombosed veins lack resilience, feel cord-like, and roll easily.

If superficial veins are not readily apparent, you can force blood into the vein by massaging the arm from wrist to elbow. Tap the site with index and second finger, apply a warm, damp washcloth to the site for 5 minutes, or lower the extremity over the bedside to allow the veins to fill.

15.2 Performing venipuncture

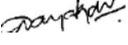
Approach the patient in a friendly, calm manner. Provide for their comfort as much as possible, and gain the patient's co-operation.

Do not rush at them, yelling, "BLOOD, BLOOD!!!" or "I am Count Dracula!"

Identify the patient correctly.

Properly fill out the appropriate request form(s), indicating the test(s) ordered.

Verify the patient's condition. Fasting, dietary restrictions, medications, timing, and medical treatment are all of concern and should be noted on the lab request slip.

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Position the patient. The patient should sit in a chair, lie down or sit up in bed.
Hyperextend the patient's arm.

Apply the tourniquet 3-4 inches above the selected puncture site. Do not place too tightly or leave on more than 2 minutes.

The patient should make a fist without pumping the hand.

Select the venipuncture site.

Prepare the patient's arm using an alcohol prep. Cleanse in a circular fashion, beginning at the site and working outward. Allow to air dry. Do not touch the site with your fingers after cleansing.

Grasp the patient's arm firmly using your thumb to draw the skin taut and anchor the vein. The needle should form a 15 to 30 degree angle with the surface of the arm. Swiftly insert the needle through the skin and into the lumen of the vein. Avoid trauma and excessive probing.

When the last tube to be drawn is filling, remove the tourniquet.

Remove the needle from the patient's arm using a swift backward motion.

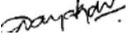
Press down on the gauze once the needle is out of the arm applying adequate pressure to avoid the formation of a haematoma.

Dispose of contaminated materials/supplies in the designated containers.

Mix and label all appropriate tubes at the patient's bedside. Label the tubes with the patient's name and hospital/clinic number.

Place specimens in the appropriate collection box for delivery to the laboratory.

For an urgent specimen, send to the laboratory immediately, and inform the laboratory staff.

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16 Additional considerations

How to prevent a haematoma:

Puncture only the uppermost wall of the vein

Remove the tourniquet before removing the needle

Use the major superficial veins

Make sure the needle fully penetrates the upper most wall of the vein. (Partial penetration may allow blood to leak into the soft tissue surrounding the vein by way of the needle bevel)

Apply pressure to the venipuncture site

16.1 How to prevent haemolysis

Mix tubes with anticoagulant additives gently 5-10 times

Avoid drawing blood from a haematoma

Avoid drawing the plunger back too forcefully, if using a needle and syringe, and avoid frothing the sample

Make sure the venipuncture site is dry

Avoid a probing, traumatic venipuncture

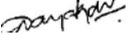
16.2 Indwelling lines or catheters

Potential source of test error

Most lines are flushed with a solution of heparin to reduce the risk of thrombosis

Discard a sample at least three times the volume of the line before a specimen is obtained for analysis

16.3 Haemoconcentration

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An increased concentration of larger molecules and formed elements in the blood may be due to several factors:

Prolonged tourniquet application (no more than 2 minutes)

Massaging, squeezing, or probing a site

Long-term IV therapy

Sclerosed or occluded veins

16.4 Prolonged Tourniquet Application

Primary effect is haemoconcentration of non-filterable elements (i.e. proteins). The hydrostatic pressure causes some water and filterable elements to leave the extracellular space.

Significant increases can be found in total protein, aspartate aminotransferase (AST), total lipids, cholesterol and iron

Affects packed cell volume (PCV) and other cellular elements

17 Safety and infection control

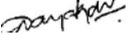
Due to contact with sick patients and their specimens, it is important to follow safety and infection control procedures.

Protect yourself

17.1 Practice universal precautions:

Wear gloves and a lab coat or gown when handling blood/body fluids.

Change gloves after each patient or when contaminated.

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Wash hands frequently.

Dispose of items in the appropriate containers.

Dispose of needles immediately upon removal from the patient's vein. Do not bend, break, recap, or re-sheath needles to avoid accidental needle puncture or splashing of contents.

Clean up any blood spills with a disinfectant such as freshly made 10% bleach.

If you stick yourself with a contaminated needle:

Remove your gloves and dispose of them properly.

Squeeze puncture site to promote bleeding.

Wash the area well with soap and water.

Record the patients name and ID number.

Follow your institutions guidelines regarding treatment and follow-up.

NOTE: The use of prophylactic antiretrovirals following blood exposure to HIV has shown effectiveness (about 79%) in preventing seroconversion

Protect the patient

Place blood collection equipment away from patients, especially children and psychiatric patients.

Practice hygiene for the patient's protection. When wearing gloves, change them between each patient and wash your hands frequently.

18 Troubleshooting guidelines

If an incomplete collection or no blood is obtained:

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Change the position of the needle. Move it forward (it may not be in the lumen)

Or move it backward (it may have penetrated too far).

Adjust the angle (the bevel may be against the vein wall).

Loosen the tourniquet. It may be obstructing blood flow.

Try another tube. There may be no vacuum in the one being used.

Re-anchor the vein. Veins sometimes roll away from the point of the needle and puncture site.

If the blood stops flowing into the tube:

The vein may have collapsed; resecure the tourniquet to increase venous filling. If this is not successful, remove the needle, take care of the puncture site, and redraw.

The needle may have pulled out of the vein when switching tubes. Hold equipment firmly and place fingers against patient's arm, using the flange for leverage when withdrawing and inserting tubes.

Ask the patient to take a deep breath. The patient may be holding their breath, causing vasoconstriction.

18.1 Problems other than an incomplete collection:

A haematoma forms under the skin adjacent to the puncture site- release the tourniquet immediately and withdraw the needle. Apply firm pressure.

The blood is bright red (arterial) rather than venous (dark red). Apply firm pressure for more than 5 minutes.

19 Blood collection on newborns

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The recommended location for blood collection on a newborn baby or infant is the heel.

Pre-warming the infant's heel (42° C for 3 to 5 minutes) is important to obtain capillary blood for blood gas samples and warming greatly increases the flow of blood for collection of other specimens. However, do not use too high a temperature warmer, because baby's skin is thin and susceptible to thermal injury.

Clean the site to be punctured with a 70% alcohol sponge. Dry the cleaned area with a dry cotton sponge. Hold the baby's foot firmly to avoid sudden movement.

Using a sterile blood lancet, puncture the side of the heel. Do not use the central portion of the heel because you might injure the underlying bone, which is close to the skin surface. Do not use a previous puncture site. Make the cut across the heel prick lines so that a drop of blood can well up and not run down along the lines.

Wipe away the first drop of blood with a piece of clean, dry cotton. Since newborns do not often bleed immediately, use gentle pressure to produce a rounded drop of blood. Do not use excessive pressure or heavy massaging because the blood may become diluted with tissue fluid.

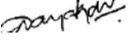
Fill the capillary tube(s) or micro collection device(s) as needed.

When finished, elevate the heel, place a piece of clean, dry cotton on the puncture site, and hold it in place until the bleeding has stopped.

Be sure to dispose of the lancet in the appropriate sharps container. Dispose of contaminated materials in appropriate waste receptacles. Remove your gloves and wash your hands.

20 Guidelines for Packaging and Transporting Samples

20.1 Transportation

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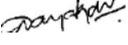
Regulations on the transportation of biological agents are aimed at ensuring that the public and the workers in the transportation chain are protected from exposure to any agent that might be in the package. Protection is achieved through

The requirements for rigorous packaging that will withstand rough handling and contain all liquid material within the package without leakage to the outside,

Appropriate labeling of the package with the biohazard symbol and other labels to alert the workers in the transportation chain to the hazardous contents of the package,

Documentation of the hazardous contents of the package should such information be necessary in an emergency situation, and

training of workers in the transportation chain to familiarize them with the hazardous contents so as to be able to respond to emergency situations.

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20.2 General Packaging Requirements for Transport of Biological Agents and Clinical Specimens

Figure 1 shows the generalized "triple" (primary receptacle, water tight secondary packaging, durable outer packaging) packaging required for a biological agent of human disease or materials that are known or suspected of containing them. This packaging requires the "Infectious Substance" label shown in Figure 2 on the outside of the package. This packaging must be certified to meet rigorous performance tests as outlined in the DOT, USPS, PHS, and IATA regulations.

Clinical specimens with a low probability of containing an infectious agent are also required to be "triple" packaged, but performance tests require only that the package shall not leak after a four-foot drop test. DOT, PHS, and IATA require a "clinical specimen" label on the outside of the package.

To monitoring temperature inside the package, a temperature monitoring device is used.

20.3 Transfer of Select Biological Agents of Human Disease

42 CFR Part 72.6 Additional Requirements for Facilities Transferring or Receiving Select Agents. Facilities transferring or receiving select agents must be registered with the CDC and each transfer of a select agent must be documented.

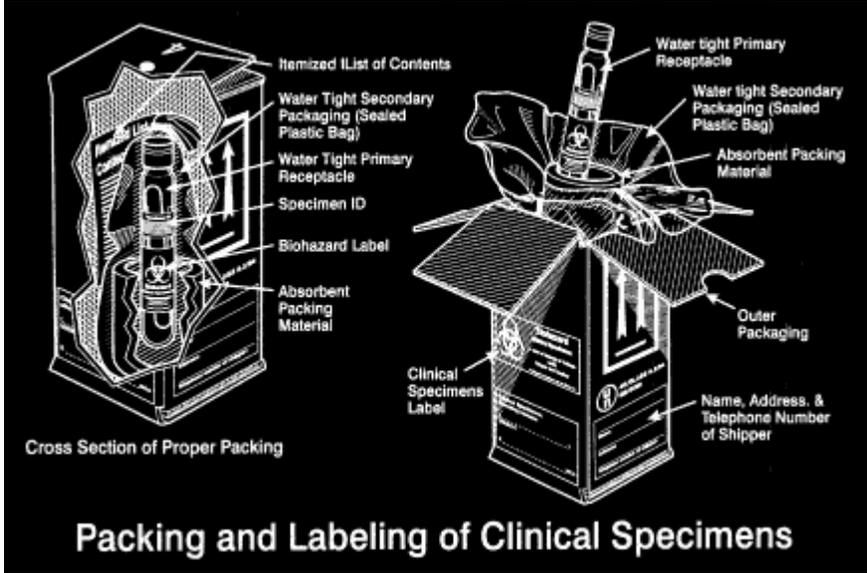
Figures 1 and 2 illustrate the packaging and labeling of infectious substances and clinical specimens in volumes of less than 50 ml..

Figure 1. Packing and Labeling of Infectious Substances

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Figure 2. Packing and Labeling of Clinical Specimens



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21 References

James Chin (2000). Control of Communicable Diseases Manual, 17th edition, American Public Health Association.

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