National Guidelines for Ebola Preparedness and Response

National Institute of Virology (NIV) Pune

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Overview:

Purpose:

This document is intended to provide guidance regarding surveillance, containment, and clinical management including support of laboratories for Ebola diagnosis and clinical follow up.

Scope:

Surveillance, prevention of entry/spread of this virus in India along with clinical aspects, sampling strategies, transport, and laboratory diagnosis

Audience:

Healthcare professionals including hospital personnel, public-health staff, laboratory staff as well as other response workers

General information:

Ebola virus disease (formerly known as Ebola haemorrhagic fever) is a severe, often fatal illness, with an average case fatality ratio [CFR] of 50%, ranging from 25 to 90%. The virus was first isolated in 1976. The illness affects humans and nonhuman primates (monkeys, gorillas, and chimpanzees). Ebola is transmitted to humans from bats, monkeys and other animals. (http://www.who.int/mediacentre/factsheets/fs103/en/). Genus Ebola Virus is 1 of 3 members of the Filoviridae family (filovirus), along with genus Marburgvirus and genus Cuevavirus. Genus Ebola Virus comprised 5 distinct species:(http://mohfw.gov.in/showfile.php?lid=2887). The current epidemic is caused by the Zaire strain causing wide-spread human-to-human transmission.

The incubation period in humans is 2-21 days with an average of 8-10 days. The illness starts with fever, muscle-aches, weakness, vomiting, diarrhoea, abdominal pain etc. Haemorrhages are reported in most of the cases. Case definitions for surveillance have been recommended by GoI. For confirmation of diagnosis, blood sample is the preferred specimen. [Case definition by GoI- http://mohfw.gov.in/showfile.php?lid=2887]

Transmission occurs only after development of symptoms. Patients are infectious as long as their blood and secretions contain the virus. Hence infected patients are closely

monitored by medical professionals and undergo laboratory tests to ensure that the virus is no longer circulating in their systems before they return home. Men who have recovered from the illness can still spread the virus to their partner through their semen for up to 7 weeks after recovery.

Current status:

As on 14th November 2014, widespread transmission of Ebola has been reported in Guinea, Liberia and Sierra Leone as per the WHO update. Limited transmission is reported from Nigeria, Spain and United States. Travel associated cases were seen in Mali and Senegal. So far, 14413 cases have been reported from eight affected countries, with 5177 deaths. A total of 570 healthcare workers are reported to be infected so far and 324 have died. [http://who.int/csr/disease/Ebola/situation-reports/en/].

As on 18th November 2014, a confirmed and cured Ebola case, an Indian national from Liberia, has been isolated at the Delhi International Airport quarantine Centre. [GoI Press release - http://pib.nic.in/newsite/PrintRelease.aspx?relid=111541]

Risk assessment:

Healthcare workers, family members or others in close contact with infected people, and mourners who have direct contact with the bodies of the deceased as part of burial ceremonies are at higher risk of infection.

In unaffected countries like India, people at risk of acquiring infection include persons having history of direct close contact with symptomatic persons during visit or stay in affected countries or during close contact with symptomatic traveller.

Considering the report of confirmation of Ebola case in India, transmission to close contacts including health professionals needs to be prevented on priority basis. Appropriate use of Personal Protection Equipment (PPE) in addition to standard precautions needs to be observed meticulously. Transmissibility of the case needs to be monitored. Decision to discharge would depend on evidence of virus absence in body fluids. This is to prevent further spread of transmission by sexual route.

Advisory for screening at ports of entry and surveillance of travellers:

Advisory to all airports has been issued by the GOI for any traveller who after visiting African countries, primarily west African countries which are affected with EVD (link),develops symptoms suggestive of Ebola, up to 30 days of arrival in India, should immediately visit the nearest designated hospital [*Up dated list of countries can be seen from WHO website www.who.int. State surveillance officers [SSOs] are provided with an advisory. (http://mohfw.gov.in/showfile.php?lid=2890)

Prevention and control:

Currently, there is no licensed medicine or vaccine for Ebola virus disease, but several products are under development.

Good outbreak control relies on applying a package of interventions, namely case management, surveillance and contact tracing, a good laboratory service, safe funeral and social mobilisation. Measures to be applied include gloves and appropriate personal protective equipment when taking care of ill patients in designated isolation facilities. Regular hand washing is required after visiting patients in hospital, as well as after taking care of patients. **(http://www.who.int/mediacentre/factsheets/fs103/en/).**The healthcare workers need to follow standard precautions. Refer to Hospital infection control guideline for preventing infection among health workers. (http://mohfw.gov.in/showfile.php?lid=2899).

Priority activities for Ebola response :

- Provide advice to travellers to Ebola affected areas with relevant information on risks, measures to minimize those risks, and steps to take following a potential exposure.
- Identify isolation units where any suspect Ebola case could be properly investigated and managed at designated hospitals near each port of entry.
- Verify access to a diagnostic capacity in a WHO-recognized laboratory.
- Establish a strategy for identifying and monitoring the contacts of any suspect Ebola case
- Where appropriate, ensure preparedness activities including contingency planning for health centres, schools and other vital infrastructure and services.

- Reinforce the capacity to manage travellers who arrive at international airports with unexplained febrile illness and potential exposure to Ebola
- Ensure a protocol, and identify an isolation unit, for the investigation and management of any suspect Ebola case
- Indicate additional scenario on confirmed case

Guidelines for airlines, travellers, screening, case detections, case / contact / person under investigation definitions, investigations and management

Travellers returning from affected areas:

Ideally, all the travellers leaving countries where continued transmission of Ebola is established should have gone through 'exit screening'. The risk of a traveller visiting the affected areas and developing disease after returning is extremely low. Transmission requires direct contact with blood, secretions, other body fluids or tissues of infected persons, or with infected dead bodies or animals, all unlikely exposures for a traveller. Travellers are strongly advised to avoid all such contacts. In view of the prevailing situation it is advisable to avoid / defer travel to countries affected with outbreak of Ebola Virus disease. Guidance is also provided for Indian families living in these countries as well as those returning to India.

Travellers visiting family and friends in affected countries:

The risk for travellers visiting family and friends in affected areas is similarly low, unless the traveller has direct physical contact with a sick or dead person or animal infected with Ebola virus. Government of India has issued a travel advisory for such travellers. [MoHFW guidelines for air travellers: http://mohfw.gov.in/showfile.php?lid=2886]

Asymptomatic traveller:

All travellers returning from Ebola affected countries should be screened by health personnel at ports of entry, the screening should include history of travel, symptoms, potential contact with Ebola patients, and thermal scanning.

The airlines and local staff need to maintain contact details [addresses and phone numbers in India] of all passengers. These patients should be given a contact phone number of

central/state control rooms and asked to call immediately in case they develop symptoms suggestive of Ebola.

State health officials need to contact these persons frequently to check if any one of them has developed symptoms within recommended surveillance period. Such a telephonic surveillance is necessary to avoid further transmission.

In case such a person develops any symptoms, they should inform the state health officials immediately. They should stay away from family members and other people. Care of such patients at home is not advisable and they need to be isolated and tested as soon as possible. Such patients need to be admitted to wards for 'Suspected' patients and not in wards for 'Ebola confirmed' cases.

If a person diagnosed during his stay in affected countries but declared 'cured' and discharged, travels to India, his body fluids need to be tested for presence of the Ebola virus. This is especially required for containing transmission possibility due to presence of virus in semen.

Persons travelling with symptoms and fellow travellers:

All travelers are required to declare their travel itinerary and symptoms if any. There is a possibility that a person who has been exposed to Ebola virus and developed symptoms may board a commercial flight or other mode of transport, without informing the transport company of his/her status. A person may also develop symptoms during travel.

On-board the aircraft:

- The person should be made to sit beyond a distance of 3 feet from other passengers.
- The person should be sitting at the closest seat to the toilet.
- No-one should enter the toilet that the person has used. It could be used again after cleaning and clearance from the health authorities.
- The symptomatic traveler should be asked to cover his/her mouth and nose while coughing and sneezing.
- Only one crew member should attend to the symptomatic traveler/s and that member should not attend other travelers.

In case of vomiting / haemorrhage / other medical emergency:

- It is mandatory that all the other passengers on-board should avoid direct contact with any body fluids from the Ebola suspected traveler.
- Disinfectants available on-board aircraft need to be poured at the site where such body fluids are there on the surface and the crew should avoid direct contact with these fluids.

In case of death on-board:

- Ideally body needs to be put into a leak-proof body-bag.
- If this is not possible, cover the body with plastic sheets and avoid further contact till the airport health authorities bring in trained hospital staff to take care of the body.

The aircraft should be made available for next flight only after disinfection and clearance by airport health officials.

At the time of disembarkation:

- The pilot should intimate the airport health authorities immediately and arrange for evacuation.
- Health personnel should examine the person and see if s/he fits the case definition.
- On verification, such person should be transferred to designated facility in a designated ambulance by competent staff only.
- It should include thermal scanning.
- It may also include verification of clinical symptoms and signs for requirements of case definitions. [Case definitions have been provided by GoI: http://mohfw.gov.in/showfile.php?lid=2887]
- Although the **risk to fellow travelers** in such a situation **is very low**, **contact management** is recommended in these circumstances.
- The crew and fellow travelers should be advised to self monitor for development of symptoms suggestive of Ebola for 21 days and report to health authorities immediately.

[WHO Guidelines: http://apps.who.int/iris/bitstream/10665/132168/1/WHO_EVD_Guidance_TravelTransportRisk_14.1_eng.pdf; MoHFW Guidelines for airlines: http://mohfw.gov.in/showfile.php?lid=2885]

Screening at ports of entry:

Staff working at airport healthcare facilities at the 'ports of entry' needs to be sensitized.

Case definitions advocated by GoI need to be followed.

Case Definition of Ebola Virus Disease (EVD):

Suspected (clinical) case:

- Any person ill or deceased who has or had fever with acute clinical symptoms and signs of hemorrhage, such as bleeding of the gums, nose-bleeds, conjunctival injection, red spots on the body, bloody stools and/or melena (black liquid stools), or vomiting blood(haematemesis) with the history of travel to the affected area. Documented prior contact with an EBVD case is not required.

Probable case (with or without bleeding):

- Any person (living or dead) having had contact with a clinical case of EHF and

with a history of acute fever.

OR

- Any person (living or dead) with a history of acute fever and three or more of the

following Symptoms: headache/ vomiting/nausea/ loss of appetite/ diarrhea/ intense fatigue/ abdominal pain/ general muscular or articular pain/ difficulty in swallowing/ difficulty in breathing/hiccoughs.

OR

Any unexplained death.

- The distinction between a suspected case and a probable case in practice

  Relatively unimportant as far as outbreak control is concerned.

Contact:

- A person without any symptoms having had physical contact with a case or the body fluids of a case within the last three weeks. The notion of physical contact may be proven or highly suspected such as having shared the same room/bed, cared for patient, touched body fluids, or closely participated in a burial (e.g. physical contact with the corpse).

Confirmed Case:

- A suspected or probable case with laboratory confirmation (positive IgM

antibody, positive PCR or Viral isolation).

Protocols need to be in place for communication to responsible authorities. Ensure basic training of staff working at points of entry or in emergency teams, and health-care workers on principles of infection prevention and control, including hand hygiene, waste management, injection safety, and use of personal protective equipment, and other precautions to apply when in close contact with a suspected or confirmed case of EVD.

Isolation/Quarantine facility at the airport:

A designated, well-equipped isolation and quarantine facility at the airport needs to be manned by trained personnel.

Ambulance services:

A designated ambulance manned by trained professionals with infection control measures required to limit transmission possibility should be used to transport the person to designated isolation facility.The ambulance should be used for transporting another Ebola suspect only after proper disinfection.

Hospital Preparedness:

General Preparedness:

- Hospitals that will handle suspected/confirmed cases of EVD must be identified -at least one in each city with an international airport.
- The hospital must plan for capacity to manage at least 10 EVD patients.

Set up of the Isolation Facility:

- Isolation facility to be prepared and kept ready for admission of suspected EVD cases. This may be upgraded existing facility or newly created one. Specific signage to be put up indicating that the room contains a patient with or at high risk of Ebola There should be adjoining separate changing rooms for donning and doffing of PPE.
- Only single patient must be admitted in one room. Room must have adjoining dedicated toilet or latrine with showers, sink equipped with running water, soap and towels,
- Mattress and pillow should be covered with plastic or other impervious covering so that no fluid can seep through.
- The isolation area must be clearly divided into suspected and confirmed area. Patients will be first admitted in the suspected area. They will be moved to the confirmed area only when laboratory test turns out to be positive. There should otherwise be no movement of patients between the two zones.
- Staff should be dedicated to the two separate zones. If this is not possible and same staff has to care for suspected and confirmed cases, move from suspected to confirmed zone, while maintaining proper hand hygiene and PPE.
- A small laboratory facility must be set up adjacent to the isolation room, to provide routine laboratory services. This must be equipped with minimal essential equipment for routine hematology and biochemistry testing. Only trained staff should work in this laboratory

Access Control:

Access to the room must be strictly restricted. If necessary, personnel to be posted at the door to ensure this.

- Visitors are not to be allowed, except in rare situation, where a child may be admitted. In such case, one parent may be allowed, who will also need to use PPE.
- Entry and exit logs of all healthcare workers who enter the room of an Ebola patient must be maintained.

Maintenance of adequate supplies:

There should be adequate provision of PPE, alcoholic hand rub with dispensers, soap, running water, disposables, consumables, disinfectants, cleaning equipment etc. Orders need to be placed well in advance and there should be some arrangement for scaling up of supply in case of emergency. Some stock should be maintained in the isolation area itself-separate for suspected and confirmed areas.

Personnel:

All categories of hospital staff who will deal with Ebola suspect patients must be previously identified and trained.

- These include doctors, nurses, support staff like therapists, technicians, housekeeping staff, laundry service, dietician, catering staff etc. who will be involved in different activities including patient care, laboratory testing and support activities.
- They should be provided information on the virus, mode of transmission as well as guidelines and training on precautions (standard, contact and droplet) to be followed, use of PPE especially method of donning and doffing & hand hygiene.
- Specific personnel must be provided additional training on safe injection practices, use and disposal of sharps, precautions for aerosol generating procedures. After training, personnel will need to demonstrate their competency especially in donning and doffing of PPE.
- Health care workers must also be taught what to do if they themselves suspect any exposure to EVD patient or suspected /confirmed breach in procedure. (Annexure)
- Personnel coming off duty must provide written checklist of activities during the shift. They should also be assessed for fatigue, illness by medical personnel.

Hospital Infection Prevention & Control:

(http://mohfw.gov.in/showfile.php?lid=2901 &http://mohfw.gov.in/showfile.php?lid=2900)

The purpose of following infection prevention and control practices is to prevent the nosocomial transmission of the virus.

- In healthcare settings, Ebola virus spreads through direct contact (e.g., through broken skin or through mucous membranes of the eyes, nose, or mouth) with blood or body fluids of a person who is sick with Ebola virus.
- In addition, needles, syringes and material contaminated with infected fluids, can cause infections in health staff and patients.
- When medical items are re-used without adequate sterilization on patients attending a health facility, numerous people and health staff can get infected.
- If no hand washing takes place in between caring for patients, infections can spread between health staff, and from health staff to other patients and contacts.
- Standard, contact and droplet precautions need to be strictly followed while caring for the patients.

Standard Precautions are the minimum infection prevention practices that apply to all patient care, regardless of suspected or confirmed infection status of the patient, in any setting where healthcare is delivered. These practices are designed to both protect Health Care Personnel (HCP) and prevent HCP from spreading infections among patients. Standard Precautions include: 1) hand hygiene, 2) use of personal protective equipment (e.g., gloves, gowns, masks), 3) safe injection practices, 4) safe handling of potentially contaminated equipment or surfaces in the patient environment, and 5) respiratory hygiene/cough etiquette.

(Further details available at http://www.who.int/csr/resources/publications/EPR_AM2_E7.pdf)

Prevention Precautions:

Precautions	Use	Requirements
Contact precautions	Patients known or suspected to have serious illnesses easily transmitted by direct patient contact or by contact with items in the patient's environment	Gloves Gown
Droplet precautions	Barrier to stop infections spread by large (>5 microns), moist droplets produced by people when they cough, sneeze or speak	Contact precautions Well-fitting mask Eye protection

Contact Precautions:

Apply to patients known or suspected to be infected with a pathogen that can be transmitted by direct or indirect contact. Perform hand hygiene before touching patient and prior to wearing gloves

PPE use:

- Wear gloves when touching the patient and the patient's immediate environment or belongings.
- Wear a gown if substantial contact with the patient or their environment is anticipated.
- Perform hand hygiene after removal of PPE; note: use soap and water when hands are visibly soiled (e.g., blood, body fluids), or after caring for patients with diarrhea.

Droplet Precautions

Apply to patients known or suspected to be infected with a pathogen that can be transmitted by droplet route.

- Place the patient in a room with a closed door as soon as possible (prioritize patients who have excessive cough and sputum production);

PPE use:

- Wear a facemask, such as a procedure or surgical mask, for close contact with the patient; the facemask should be donned upon entering the exam room
- If substantial spraying of respiratory fluids is anticipated, gloves and gown as well as goggles (or face shield in place of goggles) should be worn
- Perform hand hygiene before and after touching the patient and after contact with respiratory secretions and contaminated objects/materials; note: use soap and water when hands are visibly soiled (e.g., blood, body fluids).
- Instruct patient to wear a facemask, avoid coming into close contact with other patients, and practice respiratory hygiene and cough etiquette.
- Clean and disinfect the room accordingly.

Hand Hygiene:

HCW should perform hand hygiene frequently, including before and after all patient contact, contact with potentially infectious material, before putting on and upon removal of PPE, including gloves. (http://www.who.int/gpsc/5may/Your_5_Moments_For_Hand_Hygiene_Poster.pdf , http://www.who.int/gpsc/tools/GPSC-HandRub-Wash.pdf).

Personal Protective Equipment User's Guidelines (Annexure-I):

- ✓ PPE protects the person wearing it and reduces the risk of becoming infected in the course of patient care.
- ✓ It should completely cover the body and leave no space open.

- ✓ The most vulnerable areas are the mucous membranes of the nose, mouth and eyes, and the hands as they are most frequently in direct contact with the patient.
- ✓ A good understanding of the different components of PPE is necessary for correct usage
- ✓ Patient's support staff who may have no or limited contact can use an adapted set of protective clothing, that is easier and more comfortable.
- ✓ This must however include N95 mask or face shields, one pair of gloves, a gown, head cap and boots.
- ✓ Heavy duty rubber gloves will be needed for cleaning staff.
- ✓ Goggles may be used but personnel should be warned that touching them increases the risk of getting infected. A competent staff member must supervise donning and doffing of PPE.
- ✓ However patient's attendants that will help to care of patients (e.g. parents of a sick child) will need to be dressed in complete PPE.

Hospital Management of Case:

When the patient reaches hospital in ambulance, he/she should immediately be shifted to designated isolation facility.

Admission procedures should be minimized and completed without direct exposure of the patient to reception staff, billing clerks etc

Patient Care:

- ✓ Patient charts and records should be kept outside the isolation rooms/areas to avoid their contamination.
- ✓ Wherever possible, single use disposable material to be used in patient care. (E.g. suction catheters, needles and syringes, IV tubings.etc)
- ✓ Non disposable equipment and material (e.g. stethoscope) used in patient care must not be shared with other patients. These should be thoroughly disinfected after use.

- ✓ Aerosol generating procedures: Procedures which can generate aerosols (open suctioning, bronchoscopy, intubation/ extubation, sputum induction etc) must be avoided if possible. Whenever essential, perform with great care
- ✓ Use of sharps must be restricted and safe injection practices are to be followed.

Safe Injection Practices

- Each patient should have dedicated injection and parenteral medication equipment.
- These are to be disposed at point of care.
- Use of sharps; must be restricted as much as possible e.g. phlebotomy procedures for laboratory testing to be limited to the minimal requirement.
- Whenever sharps are used, ensure that following precautions are maintained:
  - Never replace the cap on a used needle.
  - Never direct the point of a used needle towards any part of the body.
  - Do not remove used needles from disposable syringes by hand, and do not bend, break or otherwise manipulate used needles by hand.
  - Never re-use syringes or needles.
  - Dispose of syringes, needles, scalpel blades and other sharp objects in appropriate, puncture-resistant containers.
  - Ensure that containers for sharps objects are placed as close as possible to the immediate area where the objects are being used ('point of use') to limit the distance between use and disposal, and ensure the containers remain upright at all times.
  - Ensure that the containers are securely sealed with a lid and replaced when ¾ full.
  - Closed, resistant shoes (e.g. boots) should be used by all individuals in the patient care area to avoid accidents with misplaced, contaminated sharp objects.
- Phlebotomy procedures for laboratory testing must be limited to the minimal requirement. These need to be performed only by trained health care worker.

Clinical Management:

Presently no specific therapy or prophylaxis is available for EVD. GoI guidelines to be followed. (http://mohfw.gov.in/showfile.php?lid=2899)

- ✓ Supportive therapy has to be provided.
- ✓ Intravascular volume, electrolyte balance and nutrition has to be maintained
- ✓ Hydration needs to be maintained, especially in case of severe vomiting/diarrhea.
- ✓ Fever may be treated with paracetamol.
- ✓ There is no role of steroid therapy.
- ✓ Antibiotics are not to be given except in case of secondary bacterial infection.
- ✓ Surgical intervention must be strictly avoided.

Routine Investigation:

- ✓ Any other sample for routine testing must be drawn at same time. Avoid multiple pricks. Do not perform finger pricks for routine tests.
- ✓ Specimen tubes must be labeled prior to drawing the sample. Vacuum containers may be used. Use of gel tubes reduces the need for centrifuging blood for serum separation.
- ✓ Only authorized staff must draw the specimens. PPE must be worn and standard precautions followed. (Ref Guidelines for Specimen Collection, packing and transport)
- ✓ Routine hematology and biochemistry testing to be done in dedicated laboratory
- ✓ Testing for other infectious diseases could be done based on public health considerations and clinical presentation.

Testing for Ebola Virus

Case identification and verification needs to be done as per the case definition recommended by the Government of India. These activities are to be undertaken by the state/district/local health authorities in consultation with the state nodal/surveillance officer.

Sampling of suspect case-patients needs to be performed only in designated patient isolation facilities. Contacts should be sampled only if they become symptomatic within 21 days of exposure which is the surveillance period.

Timing for specimen collection:

Specimens should be collected when the patient shows symptoms that meet the case definition of EVD. Specimens collected less than 3 days post onset of disease (POD), if tested negative, second specimen should be collected within 48hr of first specimen. For IgM antibody testing, specimens of 8 POD and above would be ideal.

Types of specimens:

Whole blood collected in EDTA is the ideal specimen for detecting RNA. The specimen should be stored at 0-5^{o}C until transport to the laboratory within 24-48 hours for processing. In case the evacuated gel tubes are not available; with extreme precaution blood may be collected in routine syringe and transferred to EDTA bulb. This may be done only in extremely rare situations because the sampling is supposed to take place in designated isolation facilities which are expected to be well equipped.

PPE during Sampling:

Basic precautions include protection for head, eyes, respiration, body, hands and feet, mucosal membranes. Different sets of PPE are available based on risk of potential contamination by blood, body fluid, excretions, etc. Ideally for Ebola, powered air purifying respirators (PAPR) must be used while handling suspected patients (Pl see **Annexure I for** donning and doffing of PPE and PAPR – this annexure can also be given as a hand-out). PAPR with a hood or helmet offers many advantages over an N95 filtering face-piece or similar respirator, being more protective, comfortable, and cost-effective in the long run. In case of non-availability of PAPR, health care workers are advised to use fit-tested N95

respirator in combination with single-use (disposable) surgical hood extending to shoulders and single-use (disposable) full face shield and impermeable gown and gloves.

Collection of blood specimen:

The following steps are to be followed along with recommended logistics: [WHO guidance – http://www.who.int/entity/csr/resources/publications/Ebola/blood-collect-en.pdf?ua=1

Key steps are -

1. Before entering the patient's room, assemble all equipment
    a. Specimen collection materials.
    b. PPE.
    c. Packaging materials
    d. Waste management materials during specimen collection and shipment
    e. Fill out all the patient documentation including forms/labels, etc. [NIV Specimen request form as **Annexure-III**,]
2. Put on all PPE after careful hand-wash as mentioned in the WHO SOP.
3. Collect blood sample [5ml] from patient – preferably use pre-labelled [8-10 ml] vacutainer tubes that are leak-proof containers. Glass syringes/vials are not allowed.

   Procedures like centrifugation, aliquoting, etc at the site of specimen collection are not allowed to avoid accidental transmission.
4. Prepare the sample for packaging/transport.
5. Remove PPE as per the SOP.

Additional Samples

Semen: Since Ebola virus is known to persist in semen for more than seven weeks after being tested negative for blood.

Oral swab: If blood collection is not possible such as elderly people, infants, and death casesetc, oral swabs can be collected for screening/testing. The swabs should be transferred to chilled viral transport medium (VTM) immediately and stored at 0-5^{o}C until processing.

Breast milk: Ebola virus is also detected in breast milk of infected women after recovery, and therefore, breast milk should also be tested for the virus even after recovery.

Tests/techniques used for detection of Ebola virus :

- Real Time RT-PCR
- Reverse transcriptase polymerase chain reaction (RT-PCR,) assay
- Antigen detection tests
- Antibody capture enzyme linked immunosorbent assay (ELISA)

Shipment/transport:

1. Prepare all shipping equipment.
2. Prepare the sample [applicable only if primary container is other than plastic/leak-proof]
3. Package the sample.
4. Mark and label the box.
5. Finalize the shipment – after contacting the reference laboratory [list to be included in Annexure IV along with contact details], Transport Company and issuance of shipping and tracking receipt.
6. Laboratory in-charge of reference lab may be contacted for any clarifications. Prior communication to the laboratory is mandatory before dispatching the samples.

   The role of laboratory begins with receipt of specimens.

   [Additional guidelines at WHO site - http://www.who.int/csr/resources/publications/Ebola/blood-shipment-en.pdf?ua=1

Hospital Waste Management:

Personnel handling disposal of waste must wear PPE including heavy duty rubber gloves and impermeable aprons**.**

a. Waste should be segregated at point of generation to enable appropriate and safe handling.
b. Sharp objects (e.g. needles, syringes, glass articles) and tubing that has been in contact with blood or body fluids should be placed inside puncture resistant waste containers (as described above). These should be located as close as practical to the patient care area where the items are used. Ifthe sharps container is far, sharps should be carried in a covered kidney dish to carry to the sharps container.
c. Collect all solid, non-sharp, infectious waste using leak-proof waste bags and covered bins. Bins should never be carried against the body (e.g. on the shoulder).
d. Waste, such as faeces, urine and vomit, and liquid waste from washing, can be disposed of in the sanitary sewer. No further treatment is necessary

Waste Disposal:

Onsite inactivation: Ebola-associated waste may be inactivated by autoclaving in dedicated machines not used for any other purpose. Other methods of inactivation (e.g., chemical inactivation) have not been standardized.

Onsite incineration: Ebola-associated waste may be incinerated. It is essential to ensure that total incineration has taken place. Caution is also required when handling flammable material and when wearing gloves due to the risk of burn injuries if gloves are ignited.

Deep Burial: If above two facilities are not available, deep burial may be considered. Waste should be placed in a designated pit of appropriate depth (e.g. 2 meters or about 7 feet) and filled to a depth of 1–1.5 m (or about 3–5 feet). After each waste load, the waste should be covered with a layer of soil 10 –15 cm deep.

Discharge Policy :

(http://apps.who.int/iris/bitstream/10665/130883/2/WHO_HSE_PED_AIP_14.05.pdf?ua=1)

Discharge of the patient is planned by the clinical management team based on clinical and laboratory findings. Lab findings for planning a discharge include:

- A negative blood PCR regardless of any other serologic tests) on day 3 or later following onset of symptoms.
- For patients with previous positive blood PCR tests, this means a subsequent negative test 48 hours from the initial test (regardless of serology).
- If a patient continues to suffer symptoms and/or their condition is not improving, but this is not thought to be due to Ebola virus, 2 negative blood PCR tests 48 hours apart, with at least one test being done 3 days or more after onset of symptoms.

Advice on Discharge :(http://mohfw.gov.in/showfile.php?lid=2899)

- ✓ As convalescent patients will be weak and take 2-3 months to recover, proper nutrition with vitamin supplementation to be provided.
- ✓ 2-3 months must elapse before full resumption of routine activities.
- ✓ The virus can be found in semen up to 3 months after onset of disease, so theoretically infecting sexual partners is possible. Therefore use of condoms should be advised for sexual intercourse.
- ✓ Patients must report back regularly for follow-up care.
- ✓ Patients discharged back home often face stigmatization and/or rejection, sodischarge should be accompanied by the necessary psychosocial support and community awareness activities

Environmental Infection Control:

Ebola virus transmission occurs through direct contact and the role of the environment in transmission has not been established. However, because of the apparent low infectious dose, potential of high virus titers in the blood of sick patients, and disease severity, environmental infection control measures are needed to reduce the potential risk due to contaminated surfaces in the patient care environment.

- Appropriate PPE must be worn.

- Environmental surfaces or objects contaminated with blood, other body fluids, secretions or excretions should be cleaned and disinfected as soon as possible using standard hospital detergents/disinfectants (e.gFreshly prepared 1% Sodium Hypochlorite or 5% Lysol)

Management of Spillage:

- Vomitus, blood and other overt spillages on floors and similar impervious surfaces should be flooded with 10% sodium hypochlorite, covered with paper towels and left for 30 minutes before removal
- Avoid splashes while cleaning.
- All the material to be discarded appropriately.
- Cleaning personnel must wear proper PPE and perform hand hygiene after the activity.

(Interim guidelines for the clinical recognition, diagnosis and management of Ebola virus disease (EVD) in South Africa)

- Prepare fresh cleaning and disinfectant solutions every day. Change cleaning solutions and dusters, swabs, other cleaning equipment frequently while being used during the day, as they will quickly become contaminated. Any standard detergent may be sued for cleaning purpose.
- Clean and disinfect bed rails and over bed tables, housekeeping surfaces such as floors and counters at least once a day. Cleaning with a moistened cloth helps to avoid contaminating the air and other surfaces with air-borne particles.
- Dry sweeping with a broom should never be done. Rags holding dust should not be shaken out and surfaces should not be cleaned with dry rags.
- Cleaning should always be carried out from "clean" areas to "dirty" areas, in order to avoid contaminant transfer.
- Do not spray (i.e. fog) occupied or unoccupied clinical areas with disinfectant. This is a

  potentially dangerous practice that has no proven disease control benefit.

- Routine cleaning of the PPE doffing area should be performed at least once per day and after the doffing of grossly contaminated PPE. Cleaning should be performed by

a HCW wearing clean PPE. When cleaning and disinfection are complete, the HCW should carefully doff PPE and perform hand hygiene.

Management of Used Linen:

Linen that has been used on patients can be heavily contaminated with body fluids (e.g. blood, vomit) and splashes may result during handling. When handling soiled linen from patients PPE should be used.

- Soiled linen should be placed in clearly-labeled, leak-proof bags placed inside a solid container at the site of use and the container surfaces should be disinfected (using 0.5% chlorine solution) before removal from the isolation room/area.
- If there is any solid excrement such as faeces or vomit, scrape off and flush it down the toilet or in the sluice before linen is placed in its container. If the linen is transported out of the patient room/area for this procedure it should be put in a separate container – it should never be carried against the body.
- Linen should be then transported directly to the laundry area in its container and laundered promptly with water and detergent.
- For low-temperature laundering, wash linen with detergent and water, rinse and then soak in 0.05% chlorine solution or solution containing 500 ppm available free chlorine) for approximately 30 minutes.
- Linen should then be dried according to routine standards and procedures.
- Washing contaminated linen by hand should be discouraged. However, if washing machines are not available or power is not ensured, take the soiled linen out of the container and empty it into a large drum container of hot water and soap. Soak the linen in this drum and make sure it is totally covered with water. Use a stick to stir; then throw out the water and refill the drum with clean water and add chlorine 0.1% (a solution containing 1 000 ppm available free chlorine) and allow to soak for 10 – 15 minutes. Remove the linen and then rinse in clean water. Remove excess water and spread out to dry. Avoid as much splashing as possible. If safe cleaning and disinfection of heavily soiled linen is not possible or reliable, it may be prudent to incinerate the linen to avoid any unnecessary risks.

Potential exposure of Healthcare Workers (HCW):

1.Persons with percutaneous or mucocutaneous exposures to blood, body fluids, secretions, or excretions from a patient with suspected/confirmed infection with Ebola virus should stop working and immediately wash the affected skin surfaces with soap and water. Mucous membranes (e.g., conjunctiva) should be irrigated with copious amounts of water or eyewash solution. The incident must be immediately reported to the supervisor for further assessment.

2. HCW who develop sudden onset of fever, intense weakness or muscle pains, vomiting, diarrhea, or any signs of hemorrhage after coming in contact with a suspected/confirmed Ebola virus infection case or infectious material, must not report to work or if at work, immediately stop working. In addition he/she should notify the supervisor, hospital and health authorities for further action.

3. Asymptomatic HCW who had an unprotected exposure (i.e. not wearing recommended PPE at the time of patient contact or through direct contact to blood or body fluids) to a patient with suspected/confirmed infection with Ebola virus should receive medical evaluation and follow-up care including fever monitoring twice daily for 21 days after the last known exposure.

Hospital authorities must ensure that there is a system/plan/policy in place for monitoring and management of potentially exposed personnel and their contacts.

Disposal of Dead Body: (http://mohfw.nic.in/showfile.php?lid=2899)

- Safe and dignified disposal of dead body should be done with adequate precautions for prevention of transmission of EVD.
- Autopsy should not be performed.
- Relatives should be counselled properly regarding the mode of transmission of EVD and instructed strictly not to perform any ritual activities after death.
- Disposal should be done by trained staff using PPE.
- Dead body should be packed in impermeable leak proof body bags for safe disposal .
- Anyone who has accidently come in contact with blood or body fluids should be kept observed for mandatory period.

Sample processing and testing:

[*Could be separated and provided on request]*

Specimen acceptance/receipt:

Only designated trained personnel should receive the specimens. SOP for receipt need to be prepared by the laboratory according to the situation [BSL-3 /4 lab, availability of trained staff.

Processing of blood samples for ELISA, RT-PCR/real time RT-PCR:

Whole blood collected in EDTA is the ideal specimen for detecting RNA. The specimen should be stored at 0-5^{o}C until transport to the laboratory within 24-48 hours for processing. Aliquoting the sample is carried out inside a bio-safety cabinet (Class III), the content is distributed in equal quantities in sterile vials and stored at -80^{o}C. Separate aliquots are given for different tests by the in-charge of the laboratory. The detailed protocol for different tests is given below.

Diagnostic tests:

(i) IgM Capture ELISA (As per CDC kit)

Detection of IgM antibodies against Ebola virus in humans can be performed using this indirect Sandwich ELISA. The ELISA plates are coated with Goat anti-human IgM antibodies. After overnight incubation, plates are washed and incubated with blocking buffer to inhibit non-specific binding. The test serum is added along with controls. The incubation plates are washed and Ebola virus antigen is added. After 1 hour incubation, Rabbit α-Ebola antibody is added. Finally Goat α-Rabbit IgG antibody labeled with HRP is added and K & P ABTS substrate is added. The reaction is read at wavelength 414 nm. The detailed protocol is given below. All the volumes used in the test are 100µl.

- Coat the plate with Goat anti-human IgM (1:100) in 0.05M carbonate buffer, pH - 9.6, Incubate overnight at 4^{o}C.
- Wash X 4 with wash buffer
- Add 300 µl/well Blocking buffer (2% Skimmed Milk) and Incubate 2hr at RT
- Wash X 4 with wash buffer

- Add patient's serum, Positive Control, Negative control (1:100) in serum diluent (5% Skimmed milk & 0.01% Tween-20 in 1X-PBS), Incubate 1hr at 37ºC
- Wash X 4 with wash buffer
- Add Ebola-antigen and Normal-antigen (1:4) in serum diluent + (1:150) normal human serum, Incubate 1 hr at 37ºC.
- Wash X 4 with wash buffer
- Add Rabbit α-Ebola Serum (1:2000) in serum diluents, Incubate 1hr at 37ºC.
- Wash X 4 with wash buffer
- Add Goat α-Rabbit IgG-HRP (1:8000) in serum diluents Incubate 1hrat 37ºC
- Wash X 4 with wash buffer
- Add K & P ABTS substrate (1+1) Incubate 1hr at 37ºC.
- Terminate the reaction with 1% SDS solution and read the result at 414nm.

(ii) IgG capture ELISA (As per CDC kit)

This protocol describes the detection of IgG antibodies against Ebola virus from serum.Ebola- positive and normal antigen is coated to the polystyrene plate. After overnight incubation, block buffer is added to inhibit non-specific binding. Then test serum is added to it, and after incubation, detector antibody labeled with conjugate is added. The reaction is read at a wavelength of 414 nm.

The detailed protocol is given below. All the volumes used in the test are 100µl.

- Coat the plate with Ebola Positive & Normal Antigen (1:2000) in 0.05M carbonate Coat buffer, pH-9.6. Seal the plate with adhesive film, Incubate overnight at 4ºC.
- Wash X 4 with wash buffer
- Add 300µl/well Blocking buffer (2% Skimmed Milk) Incubate 2hr at RT
- Wash X 4 with wash buffer
- Add test serum, Positive Control, Negative control (1:100) in serum diluent (5% Skimmed milk & 0.01% Tween-20 in 1X-PBS), Incubate 1hrat 37ºC.
- Wash X 4 with wash buffer
- Add Anti-Swine IgG-HRP conjugate (1:8000) in serum diluents. Incubate 1hr. at 37ºC.

- Wash X 4 with wash buffer
- Add K & P ABTS substrate (1+1) Incubate 1hrat 37ºC.
- Terminate the reaction with 1% SDS solution & Read the result at 414 nm

(iii) Reverse transcriptase polymerase chain reaction (RT-PCR):

RNA Extraction:

Extract Ebola viral RNA from 140µl of patient's serum sample using QIAamp Viral RNA Mini Kit (Qiagen) as per the manufacturer's instructions. Elute RNA in 50µl AVE buffer from the kit. Samples from patients are an extreme biohazard risk. All the procedures should be carried out in a BS)-3 or 4 laboratory if the sample is non-inactivated.

Primers: Information can be provided by NIV on request.

FIRST PCR MIX:

(Super script III one step RT-PCR Kit with platinum Taq DNA polymerase)

- Molecular grade water 11.0µl
- 2X reaction mixture 25µl
- 20 pM Forward primer 2.5µl
- 20 pM Reverse primer 2.5µl
- Superscript III enzyme mix 1.0µl
- $MgSO_4$ 3.0µl

SECOND PCR MIX (SEMI NESTED PCR)

- Molecular grade water 13.0µl
- 2X reaction mixture 25µl
- 20 pM Forward primer 2.5µl
- 20 pM Reverse primer 2.5µl
- Superscript III enzyme mix 1.0µl
- $MgSO_4$ 3.0µl

PROCEDURE

- Add 5µl RNA in respectively labeled tube containing 45µl first PCR master mix.
- Perform Amplification in an automated thermal cycler according to reaction conditions as follows reverse transcription 50^0C for 30min, PCR activation

94^0C for 2 min, 35 cycles of 94^0C for 1min, 50^0C for 1 min, 68^0C for 2 min and a final extension step at 68^0 C for 10 min.

- Transfer 3µl of the first PCR product to PCR tube containing 47ul of second PCR mix.
- Perform Amplification in an automated thermal cycler according to reaction conditions as follows PCR activation 94^0C for 4 min, 35 cycles of 94^0C for 1min, 50^0C for 1 min, 68^0C for 2 min and a final extension step at 68^0 C for 10 min.
- Perform GEL electrophoresis of first and second PCR products along with 100bp marker.
- Expected PCR product size mentioned in primer table

(iv) Real time RT-PCR:

Real time RT-PCR is one of the sensitive techniques to diagnose Ebola virus. It is a specific and quantitative method of detection. RNA is extracted from the sample as described above and 5µl RNA is mixed with 45µl master mix containing respective forward and reverse primer. The mixture is amplified in a real time PCR apparatus (ABI 7300, 7500,7700; Strategene Mx3005 etc) according to reaction conditions (Refer WHO guidelines) and the data is interpreted. The algorithm calculates the threshold cycle (Ct) at which each PCR amplification reached a ΔRn threshold value that is inversely proportional to the log number of target copies present in the sample

Waste disposal (laboratory):

Disposal of Biological waste

Efficient containment of pathogen at work is achieved by proper disposal of biological wastes and proper decontamination of work areas.

DISINFECTION AND STERILIZATION:

A basic knowledge of disinfection and sterilization is crucial for bio-safety in the laboratory; therefore, it is important to understand the principle of disinfection and sterilization. This chapter deals with the general principles applicable to all known classes of microbial pathogens.

Specific decontamination requirements depend on the type of experimental work and the nature of the infectious agent(s) being handled. Contact times of disinfectants vary as per material. Therefore, all recommendations for use of disinfectants should follow manufacturers' instructions.

As per the Department of Biotechnology (DBT) guidelines, bio-waste pose hazard in the laboratory and can be dealt by disinfectants, autoclaving and incineration. The bio-waste disposal is governed/monitored by Central Pollution Control Board (CPCB) and by State Pollution Control Boards. The following elements are considered for a waste management policy:

1. Dedicated and trained staff.
2. Availability of autoclaves, incinerator, shredders, wormiculture.
3. Contract agencies to collect plastic and wet non-hazardous waste.
4. Effluent Treatment Plant (ETP) for BSL-3 and BSL-4.

Decontamination of Biological Safety Cabinets:

To decontaminate the used bio-safety cabinets, formaldehyde gas or paraformaldehyde (final concentration of 0.8% paraformaldehyde in air) is used. If the relative humidity is below 70%, an open container of hot water should also be placed inside the cabinet before the front closure is sealed in place with strong tape (e.g. duct tape). Heavy gauge plastic sheeting is taped over the front opening and exhaust port to make sure that the gas cannot seep into the room. Penetration of the electric leads passing through the front closure must also be sealed with duct tape.

The plate for the paraformaldehyde pan is plugged in. It is unplugged when all the paraformaldehyde has vaporized. The cabinet is left undisturbed for at least 6 hours. The plate for the second pan is then plugged in and the ammonium bicarbonate is allowed to vaporize. This plate is then unplugged and the cabinet blower is switched on for two intervals of approximately 2 seconds each to allow the ammonium bicarbonate gas to circulate. The cabinet should be left undisturbed for 30 minutes before the front closure (or plastic sheeting) and the exhaust port sheeting are removed. The cabinet surfaces should be wiped down to remove residues before use.

Decontamination/disposal of consumables and biological waste: After fumigation, all the used material is autoclaved (double door autoclave with one door inside the experimental room and the other opening to the washing room). The disposable items and biological samples will then packed in special waste disposal bags (Marked with Ebola suspected sample) and will be incinerated. Reusable items such as scissors, forceps, glass bottles, petridishesetc will be washed, sterilized and go to the storage cupboards.

Autoclaving

Autoclave should be equipped with continuous, computerized recording system throughout length of the autoclave cycle for parameters; Dates, Time of day, Load identification number and Operating parameters. It should completely and consistently kill the approved biological indicator (*Bacillus stearothermophilus*) at the maximum design capacity of each autoclave unit (Spores using vials or spore Strips; with at least 1 X 10^4 spores per milliliter). Under no circumstances will an autoclave have minimum operating parameters less than a residence time of 30 minutes, regardless of temperature and pressure, a temperature less than 121°C or a pressure less than 15 psi.

Precautions while operating autoclaves: The following rules can minimize the hazards inherent in operating pressurized vessels.

1. Responsibility for operation and routine care should be assigned to trained individuals.
2. A preventive maintenance program should include regular inspection of the chamber, door seals and all gauges and controls by qualified personnel.
3. The steam should be saturated and free from chemicals (e.g. corrosion inhibitors) that could contaminate the items being sterilized.
4. All materials to be autoclaved should be in containers that allow ready removal of air and permit good heat penetration; the chamber should be loosely packed so that steam will reach the load evenly.
5. For autoclaves without an interlocking safety device that prevents the door being opened when the chamber is pressurized, the main steam valve should be closed and the temperature allowed falling below 80°C before the door is opened.

6. Slow exhaust settings should be used when autoclaving liquids, as they may boil over when removed due to superheating.
7. Operators should wear suitable gloves and visors for protection when opening the autoclave, even when the temperature has fallen below 80°C.
8. In any routine monitoring of autoclave performance, biological indicators or thermocouples should be placed at the centre of each load. Regular monitoring with thermocouples and recording devices in a "worst case" load is highly desirable to determine proper operating cycles.
9. The drain screen filter of the chamber (if available) should be removed and cleaned daily.
10. Care should be taken to ensure that the relief valves of pressure cooker autoclaves do not become blocked by paper, etc. in the load.

Incineration

Incineration is useful for disposal of animal carcasses as well as anatomical and other laboratory waste, with or without prior decontamination. Incineration of infectious materials is an alternative to autoclaving only if the incinerator is under laboratory control. Proper incineration requires an efficient means of temperature control and a secondary burning chamber. Many incinerators, especially those with a single combustion chamber, are unsatisfactory for dealing with infectious materials, animal carcasses and plastics. Such materials may not be completely destroyed and the effluent from the chimney may pollute the atmosphere with microorganisms, toxic chemicals and smoke. However, there are many satisfactory configurations for combustion chambers. Ideally the temperature in the primary chamber should be at least 800^{o}C and that in the secondary chamber at least 1000^{o}C. Materials for incineration, even with prior decontamination, should be transported to the incinerator in bags, preferably plastic. Incinerator attendants should receive proper instructions about loading and temperature control. It should also be noted that the efficient operation of an incinerator depends heavily on the right mix of materials in the waste being treated. There are ongoing concerns regarding the possible negative environmental effects of existing or proposed incinerators, and efforts continue to make incinerators more environmentally friendly and energy-efficient.

Disposal

The disposal of laboratory and medical waste in the institute is governed by the CPCB, the State Pollution Control Boards) as per the National guidelines provided by the Ministry of Environment and Forest. The gazette of India, part-2, section-3, subsection-2, dated 24 August 2011, Gazette number 1626 by Environment and Forest Ministry of India, provides detailed information in this regard.

Annexure I

Donning and Doffing of PPE

Steps for donning of PPE

a. Put on surgical scrubs in changing room. No personal items (e.g., jewelry, watches, cell phones, pagers, pens) should be brought into patient room.
b. Put on gum boots. If not available, make sure you have closed, puncture and fluid resistant shoes and put on overshoes.
c. **Inspect PPE Prior to Donning**: Visually inspect the PPE to be worn to ensure that it is in good condition, that all required PPE and supplies are available, and that the sizes selected are correct for the healthcare worker
d. Place the gown over the scrubs. Ensure gown is large enough to allow unrestricted freedom of movement.
e. Put on PAPR with a full face-shield, helmet, or headpiece.
f. If a PAPR with a self-contained filter and blower unit integrated inside the helmet is used, then a single-use (disposable) hood that extends to the shoulders and fully covers the neck must also be used. Be sure that the hood covers all of the hair and the ears, and that it extends past the neck to the shoulders.
g. If a PAPR with external belt-mounted blower unit and attached reusable headpiece is used, then a single-use (disposable) hood that extends to the shoulders and fully covers the neck must also be used. Be sure that the hood covers all of the hair and the ears, and that it extends past the neck to the shoulder
h. If PAPR is not available use N95 or higher respirator. Over the N95 respirator, place a surgical hood that covers all of the hair and the ears, and ensure that it extends past the neck to the shoulders. Be certain that hood completely covers the ears and neck. Put on full face shield over the N95 respirator and surgical hood to provide additional protection to the front and sides of the face, including skin and eyes.
i. Put on outer full-body apron to provide additional protection to the front of the body against exposure to body fluids or excrement from the patient.
j. Perform hand hygiene

k. Put on first pair of gloves; tape them securely to the sleeves of gown.

l. Put on second pair of gloves.

m. Use ***heavy duty/rubber gloves*** for environmental cleaning and waste management.

n. Verify that the PPE has been donned correctly. The healthcare worker should be comfortable and able to extend the arms, bend at the waist and go through a range of motions to ensure there is sufficient range of movement while all areas of the body remain covered. A mirror in the room can be useful for the healthcare worker while donning PPE.

Steps for Doffing off PPE

Before entering the PPE removal area, inspect and disinfect any visible contamination on the PPE with disinfectant wipe. As a final step, disinfect outer-gloved hands with either an alcohol based hand rub and allow to dry. Verify that the trained observer is available in the PPE removal area before entering and beginning the PPE removal process.

a. Remove and discard apron taking care to avoid contaminating gloves by rolling the apron from inside to outside.
b. Following apron removal, inspect the PPE to assess for visible contamination or cuts or tears. Then disinfect affected PPE using disinfectant wipe.
c. Disinfect outer-gloved hands with alcohol based hand rub and allow to dry
d. Disinfect and remove outer gloves: Disinfect outer-gloved hands with alcohol based hand rub remove and discard outer gloves, taking care not to contaminate inner glove during removal process.
e. Inspect and disinfect inner gloves: Inspect the inner gloves' outer surfaces for visible contamination, cuts, or tears. If an inner glove is visibly soiled, cut, or torn, then disinfect the glove with alcohol based hand rub. Then remove the inner gloves, perform hand hygiene with alcohol based hand rub on bare hands, and don a clean pair of gloves.
f. **Remove Respirator** If a PAPR with a self-contained filter and blower unit integrated inside the helmet is used, then wait until Step K for removal and go to next

step. If a PAPR with an external belt-mounted blower unit is used, then all components must be removed at this step.

Remove and discard disposable hood.

Disinfect inner gloves with alcohol based hand rub.

Remove headpiece, blower, tubing, and the belt and battery unit. This step might require assistance from the trained observer.

Disinfect inner gloves with alcohol based hand rub.

Place all reusable PAPR components in an area or container designated for the collection of PAPR components for disinfection.

g. **Remove Gown *or* Coverall**: Remove and discard. Avoid contact of scrubs or disposable garments with outer surface of gown during removal. Pull gown away from body, rolling inside out and touching only the inside of the gown.

To remove coverall, tilt head back and reach under the PAPR hood to reach zipper or fasteners. Use a mirror to help avoid touching the skin. Unzip or unfasten coverall completely before rolling down and turning inside out. Avoid contact of scrubs with outer surface of coverall during removal, touching only the inside of the coverall.

h. **Disinfect Inner Gloves**: Disinfect inner gloves with alcohol based hand rub

i. **Disinfect Washable Shoes**: Sitting on a new clean surface (e.g., second clean chair, clean side of a bench) use a disinfectant wipe to wipe down every external surface of the washable shoes.

j. **Disinfect Inner Gloves**: Disinfect inner gloves with alcohol based hand rub

k. **Remove Respirator (if not already removed)**: If a PAPR with a self-contained filter and blower unit that is integrated inside helmet is used, then remove all components.

Remove and discard disposable hood

Disinfect inner gloves with alcohol based hand rub

Remove and discard inner gloves taking care not to contaminate bare hands during removal process

Perform hand hygiene with alcohol based hand rub

Don a new pair of inner gloves

Remove helmet and the belt and battery unit. This step might require assistance from the trained observer.

l. **Disinfect and Remove Inner Gloves**: Disinfect inner-gloved hands with alcohol based hand rub. Remove and discard gloves taking care not to contaminate bare hands during removal process.

m. **Perform Hand Hygiene**: Perform hand hygiene with alcohol based hand rub.

n. **Inspect**: Perform a final inspection of healthcare worker for any indication of contamination of the surgical scrubs or disposable garments. If contamination is identified, immediately inform supervisor.

o. **Scrubs**: Healthcare worker can leave PPE removal area wearing dedicated washable footwear and surgical scrubs or disposable garments. Scrubs are to be removed in change room

Shower: Showers are recommended at each shift's end for healthcare workers performing high-risk patient care (e.g., exposed to large quantities of blood, body fluids, or excreta). Showers are also suggested for healthcare workers spending extended periods of time in the Ebola patient room.

Protocol Evaluation/Medical Assessment: the healthcare worker should be assessed to review the patient care activities performed in order to identify any concerns about patient care protocols and to record healthcare worker's level of fatigue.

Place all PPE waste in a leak-proof infectious waste container.

Annexure II

Specimen request form

[Available At: http://www.niv.co.in/EBOLA_SPECIMEN_REQUEST_FORM_NIV.pdf]

Attached as a .pdf document separately.

Annexure III

Logistics Required

PPE - Examination/Surgical gloves

Heavy duty rubber gloves

Disposable overalls/gowns

N95 masks ?PAPRs

Disposable caps (head covers)

Scrub suits

Boots

Gum boots

Gowns

Heavy duty rubber Aprons

Goggles

Waste Management

Garbage bags

Waste containers-colour coded

Sharps containers

Cleaning mops/buckets

Changeable mop heads

Cloth dusters

Disinfectant-bleach,chlorinesolution,Lysol

Autocalve/incinerator

Hand Hygeine

Liquid soap, running water

If running water not available-provision of sufficient stored water

Alcoholic hand rub with dispenser

Alcoholic hand wipes/disinfectant wipes

Patient care

Mattress

Pillow

Sheets& Blankets

Absorbent pads

Impermeable cover

Covered emesis pan

Stock of disposables required-IV sets, cannulas,medicinesetc

Dedicated equipment: Stethoscope,Sphygmomanometer,thermometer,urinal,bed pan etc

Specimen Logistics:

1. Tourniquet
2. Holder
3. Evacuated EDTA tubes
4. Needle
5. PPE
6. Packaging materials
7. Transport materials
8. Specimen referral forms
9. Disinfectants
10. Discard containers

List of contributors: [alphabetically]

1. AB Sudeep
2. Avinash R Deoshatwar
3. Babasaheb V Tandale
4. Pragya D Yadav
5. Rajlakshmi Viswanathan
6. Shailesh D Pawar
7. Yogesh K Gurav

Reviewers:

1. A Basu
2. MS Chadha
3. DT Mourya