

# LABORATORY GUIDELINES

## FOR TESTING OF

### EBOLA VIRUS DISEASE

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#### 1. Introduction and situation update

The Ebola virus has been confirmed as the cause of an outbreak of haemorrhagic fever in West Africa. Ebola virus disease (EVD, also commonly known as Ebola haemorrhagic fever) is not commonly described in West Africa, and this is the first large outbreak of the disease in this region. Lassa fever, also a haemorrhagic fever, is commonly reported from this region. The most recent outbreaks of Ebola virus disease were reported from the Democratic Republic of Congo and Uganda in 2012. This outbreak is reported to have started early February 2014 in Guinea, but cases have since been confirmed from Liberia, with suspected cases reported from other neighbouring countries (Sierra Leone, and Mali). Partial molecular sequencing of the virus genome has indicated that the virus has high level of homology with Zaire Ebola virus. Zaire Ebola virus is highly lethal with case fatality rate of up to 90% reported in previous outbreaks. Regular outbreak situation updates are posted on the NICD website, under the 'Alerts' section ([www.nicd.ac.za](http://www.nicd.ac.za)).

The incubation period of the disease is 2- 21 days. A prodrome of fever and gastro-intestinal symptoms will be followed by progressive multisystem disease with bleeding a feature in a significant number of patients but not all.

The World Health Organization and a number of international organizations are supporting government authorities in the affected countries to respond to the outbreaks. While the risk of introduction of Ebola virus into South Africa is considered low, we strongly recommend that surveillance for viral haemorrhagic fevers (and at present, particularly EVD), be strengthened. This should be done primarily through Port Health services, but it is also extremely important that public and private practitioners are on the alert for any ill persons that have travelled to viral haemorrhagic fever risk areas. There needs to be a high index of suspicion for EVD in health workers from the affected region with unexplained fever. Any ill persons reported on flights from Guinea, Liberia, Sierra Leone or Mali and neighbouring countries will need to be evaluated by the relevant Port Health officials. All requests for medical evacuation of persons from Guinea, Liberia, Sierra Leone or Mali with febrile illness or suspected infectious disease will need careful evaluation by the Port Health officials.

Testing for EVD in South Africa is only available at NICD.

## 2. Case definition

Any person* presenting with an acute onset of fever that has:	
•	Visited or resident in Guinea, Liberia, Sierra Leone or Mali in the 21 days prior to onset of illness
	AND
•	Had direct contact or cared for suspected/confirmed EVD cases in the 21 days prior to onset of illness, or been hospitalised in Guinea, Liberia, Sierra Leone or Mali
	OR
Has unexplained multisystem illness that is malaria-negative	
*Healthcare workers in particular are at high risk	

It is critical to maintain a very high index of suspicion for common causes of febrile illness in persons who have travelled to Guinea, Liberia, Sierra Leone, Mali and surrounding countries, including: malaria, dengue fever, Lassa fever and other endemic diseases (e.g. typhoid fever). These may be severe and life-threatening, and healthcare workers are urged to do appropriate tests and institute appropriate therapy as a matter of urgency. Malaria is the most likely cause of an acute febrile illness in returning travellers from most African countries and has to be prioritised for testing. However, Lassa fever is endemic in certain West African countries, including Nigeria, Sierra Leone, Guinea and Liberia - and needs to be considered in the differential diagnosis for any traveller from these countries who has unexplained febrile illness and has visited rural areas.

Lassa fever virus is transmitted to humans through direct contact with urine and droppings of infected multi-mammate rats, which contaminate the environment and food items. Transmission can also occur through the inhalation of aerosolised infected rodent excreta. Person-to-person transmission is also important, being common in both village and healthcare settings, and occurs through direct contact with blood, tissue, secretions or excretions of an infected person; therefore, VHF isolation precautions are recommended for nursing patients with Lassa fever. The incubation period is 1-3 weeks; symptoms include fever, retrosternal pain, sore throat, back pain, cough, abdominal pain, vomiting,

diarrhoea, facial swelling and mucosal bleeding. Mortality rates approach 20%, with pregnant women in their third trimester being at highest risk for severe disease and death. Given that the incubation periods and clinical presentations of Lassa fever and EVD may overlap, both diseases must be excluded in persons who have a suggestive travel history and present with a febrile illness.

Ebola virus disease testing is neither warranted nor useful for patients that are not suffering from a clinical illness compatible with Ebola haemorrhagic fever, even in the event of compatible travel histories. The tests cannot be used to determine if the patient has been exposed to the virus and may develop the disease later.

**3. Procedure for submission of specimens for testing**

**Step 1: Report the suspected case**

- Contact the NICD Hotline ☎ +2782-883-9920

**Step 2: Submit specimens for laboratory diagnosis**

- All suspected cases of EVD should have a serum (either clotted blood/red top tube or SST/yellow top tube) taken for viral RNA detection and antibody determination.
- The specimens should be packaged in accordance with the guidelines for the transport of dangerous biological goods (triple packaging using absorbent material) and transported directly and urgently to:

**Centre for Emerging and Zoonotic Diseases**  
**Special Viral Pathogens Laboratory**  
**National Institute for Communicable Diseases (NICD)**  
**National Health Laboratory Service (NHLS)**  
**No. 1 Modderfontein Rd**  
**Sandringham, 2131**

- ALL specimens should be accompanied by a fully completed VHF specimen submission form (see page 4).
- Samples should be kept cold during transport (cold packs are sufficient, but dry ice may be needed if longer transport times are involved).

**Table 1:** Summary of procedure to submit specimens for EVD investigation

Pathogen, clinical symptoms & comments	Sample collection	Tests to request	Send to	Forms to complete
<b>Viral haemorrhagic fevers</b>  Fever, headache, arthralgia, myalgia, rash, haemorrhagic manifestations, gastrointestinal symptoms, pathology lab indicators, travel history or other exposure events	Blood:	VHF/Ebola	Centre for Emerging and Zoonotic Diseases, Special Viral Pathogens Laboratory, NICD,  1 Modderfontein Road, Sandringham, 2131.	All cases to be notified to the NICD Hotline.  Cases will be referred accordingly.  Complete the NICD VHF Request Form
	Serum or clotted blood (red/yellow top tube)			

**4. Laboratory tests available at NICD**

**Table 2:** Summary of laboratory tests available at the NICD for EVD

Available tests	Special Instructions	Turn-around time	Testing laboratory
Serology: fluorescent antibody test, IgG and IgM	Tubes in sealed in plastic bags and secondary containers with absorbent material. Follow international regulations for transportation of biohazardous materials. Transport at on cold packs or dry ice (if longer transport times anticipated, where cold packs will no longer be sufficient)	24-48 hrs	Centre for Emerging and Zoonotic Diseases, Special Viral Pathogens Laboratory
Serology: ELISA, IgG and IgM		3-5 days	
PCR		24-48 hrs	+27 11 386 6376 + 27 82 903 9131



Special Viral Pathogens Laboratory

+27 82 903 9131 or +27 11 386 6376

NICD Hotline +27 82 883 9920

**REQUEST FORM: INVESTIGATION OF SUSPECTED VIRAL HAEMORRHAGIC FEVER**  
Completed form must accompany specimens

Referring doctor .....	Patient .....
Address .....	Age .....Sex.....
..... Code .....	Hospital number.....
Tel (W) .....	Hospital .....Ward.....
Tel (A/H or cellular) .....	Date of admission .....
Fax.....	Medical fund ..... No.....
Account to: ..... Code .....	

Specimen/s ..... **Date taken** .....

Disease suspected/tests required.....

Possible exposure to viral haemorrhagic fever (e.g occupation, urban or rural resident, history of travel, contact with animals or human patient, insect/tick bites - give dates).....

**N.B. ACCURATE DATE OF ONSET OF ILLNESS**.....

Clinical history and examination .....

Treatment (antibiotics/antimalarials).....

Results of laboratory investigations already performed:

Date				
Total leukocytes				
Differential %N/L				
Platelets				
Haemoglobin				
Coagulation test/s				
ALT				
AST				
Malaria parasites				
Blood culture				