

October 7, 2014

Guidelines for Pennsylvania Laboratories Handling Specimens from Patients with Suspected or Confirmed Ebola Virus Disease (EVD)

Purpose

The following guidelines are provided for Pennsylvania laboratories that may receive and test specimens from patients who are either:

- Suspected of having Ebola Virus Disease (EVD) and report LOW or HIGH risk exposure^a or
- Confirmed as having EVD with a laboratory test.

For patients with NO known exposures for EVD^b, routine clinical specimens should be received, processed and tested in accordance with usual and standard procedures for laboratory testing.

A suspected EVD patient who reports either a LOW or HIGH Risk exposure^a, and for whom a definitive diagnosis has not yet been determined, will be tested for Ebola virus **only upon** approval by both the Pennsylvania Department of Health as well as the Centers for Disease Control and Prevention (CDC). See current definitions at <http://www.cdc.gov/vhf/ebola/pdf/ebola-algorithm.pdf>.

Molecular EVD testing in Pennsylvania

Molecular diagnosis for EVD is available at the Pennsylvania Department of Health Bureau of Laboratories with a real-time RT-PCR assay that has been FDA-cleared under Emergency Use Authorization (EUA).

- **Before collecting samples** for testing, contact the Pennsylvania Department of Health, to obtain the **required prior approval for testing and instructions for specimen collection and transportation.**
- For negative results on specimens collected less than 3 days post onset of symptoms, repeat testing is recommended unless a definitive alternative diagnosis has been made and EVD is no longer in the differential diagnosis.

EVD transmission and decontamination

Please note the following points with regard to EVD:

- A person infected with Ebola virus is not contagious until symptoms appear.
- EVD is transmitted through direct contact (via broken skin or mucous membranes) with blood or body fluids from an EVD patient, or through contact with objects contaminated with blood or body fluids from an EVD patient. There is no evidence of airborne transmission.
- Ebola virus is readily inactivated by standard chemical decontamination procedures used in laboratories. Current guidance from the CDC <http://www.cdc.gov/vhf/ebola/hcp/environmental-infection-control-in-hospitals.html> suggests using either a 1:10 dilution of bleach or an EPA-registered disinfectant labeled for non-enveloped viruses such as norovirus, rotavirus, adenovirus, or poliovirus, as these viruses are more difficult to destroy than Ebola.

Ebola virus is present in numerous body fluids of patients with EVD¹. Although detected much less frequently, there is some evidence that environmental samples contaminated with blood or body fluid from an EVD patient may pose some risk of transmission¹.

Biosafety classification

The CDC has provided clarification related to two issues pertaining to Ebola virus biosafety classifications (<http://www.cdc.gov/vhf/ebola/hcp/safe-specimen-management.html>):

- While Ebola virus culture, which is commonly performed at high volume and can attain extremely high titer, is required to be performed at biosafety level 4, the handling of primary clinical specimens from EVD patients need not be restricted to this level of containment.
- According to the [Interim Guidance Regarding Compliance with Select Agent Regulations for Laboratories Handling Patient Specimens that are Known or Suspected to Contain Ebola Virus](http://www.cdc.gov/vhf/ebola/hcp/select-agent-regulations.html) (<http://www.cdc.gov/vhf/ebola/hcp/select-agent-regulations.html>), specimens from suspected EVD patients are **not** classified as select agents. For patients with confirmed EVD, select agent classification of specimens will be determined following additional testing and consultation with the CDC.

*See Supporting information for Biosafety Classification at the end of the document.

CDC guidance

Guidance from the CDC recommends that suspected EVD patients who report LOW or HIGH Risk exposure, or laboratory confirmed cases, be managed in US hospitals with standard contact and droplet precautions. Laboratory personnel are advised to adhere strictly to safety procedures for the prevention of transmission of blood borne pathogens when handling specimens from these patients (<http://www.cdc.gov/vhf/ebola/hcp/interim-guidance-specimen-collection-submission-patients-suspected-infection-ebola.html>) including the following:

- Specimen collection
 - full face protection (mask plus either goggles or face shield), gloves, impermeable gown
- Laboratory testing
 - full face protection (mask plus either goggles or face shield), gloves, impermeable gown
 - use of certified class II Biosafety cabinet or splash shield

Note, the above guidance refers to all laboratory work including the routine hematology and clinical chemistry testing that is essential for the appropriate care and treatment of patients.

Nevertheless, Ebola virus is indisputably a highly pathogenic agent². All laboratory directors should review their circumstances, facilities, resources and procedures, as well as the training and experience of their staff, in order to perform a thorough biohazard risk assessment and implement appropriate procedures for risk mitigation. However, any additional precautions or procedures should not interfere with the ability to provide appropriate medical care for suspected or confirmed EVD patients.

In light of all of the above, the following additional guidance is provided for consideration for the handling of laboratory specimens from suspected EVD patients reporting HIGH or LOW risk exposures or laboratory confirmed EVD cases.

General laboratory comments

- Laboratory testing should be limited to those tests essential to patient care. However, patient care and well-being should not be compromised.
- If available, the use of Point-of-Care instruments and methods inside or nearby the patient's isolation room is a preferred option, to provide reduced specimen transport and limit the need for testing in routine laboratories.
- Facilities that use Point-of-Care instruments should maintain a log of personnel handling specimens from suspected EVD patients.
- For testing that requires transport of specimens to the hospital laboratory, specimens should be double-bagged, placed in a durable, leak-proof biohazard transportation container, and **hand-carried** to the laboratory. **DO NOT** use a pneumatic tube system. At a minimum, specimens should be labeled to indicate that they should be hand-carried and not be placed in a pneumatic tube system. Each laboratory may wish to discuss additional labeling consistent with institutional policy that specimens have originated from a suspected LOW or HIGH risk, or confirmed EVD patient that require special transporting and handling.
- Laboratories should review their protocols for occupational exposure. If an occupational exposure occurs, employees should follow procedures for occupational exposures for their institution and make arrangements to consult with the Pennsylvania Department of Health (1-877- PAHEALTH) immediately if a potential exposure occurs.

Comments on specific laboratory procedures

Procedure	Recommendation
Centrifugation	Should be performed with sealed buckets or sealed rotor.
Homogenization	Procedures requiring homogenization of any specimen type <u>should be avoided</u> or performed with extreme care (e.g. Stomacher and sealed bag if available) due to the risk of spray or splash.
Clinical chemistry and hematology	Numerous issues pertaining to routine testing in these areas need to be considered and are highly variable depending on the type of equipment used, volume of testing performed, laboratory workflow and layout, and many other factors. A full risk assessment should be made at each site, including options for decontamination ³ . For automated instruments, decontamination procedures should be those advised by the manufacturer or vendor for non-enveloped viruses.

Procedure	Recommendation
Malaria testing	<p>Rapid antigen tests or thin blood smears are preferred: recognizing that rapid tests are inherently less sensitive but positive results are generally reliable.</p> <p>The effects of some inactivation/decontamination procedures on the performance of some rapid antigen tests for malaria have been investigated⁴.</p> <p>Thin blood smears should be fixed in methanol for 30 minutes and dried prior to staining. The use of additional heat inactivation is not considered necessary for Ebola decontamination and has been found by some parasitologists to cause disruption to the morphology of the parasites.</p> <p>Thick blood films are not recommended.</p>
Blood Cultures	Systems using plastic blood culture bottles are preferred. Blood cultures in glass bottles should be avoided if possible or handled with extreme care.
Other specimens for bacterial culture	“Pan-cultures” should not be performed. Procedures essential for patient management should be performed in a Class II Biosafety Cabinet with PPE.
Wet preps	Should not be performed.
Viral cultures	DO NOT perform viral culture , including any rapid culture systems (including shell vials), on any specimen.
Pre-transfusion testing	Please refer to the American Association of Blood Banks’ Ebola information sheet http://www.aabb.org/press/Pages/Infection-Control-for-Handling-Blood-Specimens-from-Suspected-Ebola-Patients.aspx
Post-mortem examinations	Should not be performed.
Specimen storage	With the exception of circumstances where retention is required by regulations, long-term storage of specimens is discouraged. It is recommended that specimens collected from suspected or confirmed EVD cases be isolated from other specimens in the laboratory and disposed of in an appropriate manner (see below) as soon as is practical after testing has been completed.
Specimen decontamination and disposal	Consult with the microbiology laboratory to discuss the optimal methods to decontaminate and dispose of specimens available in each specific facility. Possibilities include autoclaving specimens if facilities are available. An alternative is to decontaminate specimens in 10% bleach for 24 hours, then place in standard biohazard infectious waste disposal.

Supporting information for Biosafety Classification

Information in support of these recommendations is provided below.

- Recent experiments in Canada have demonstrated the absence of airborne Ebola transmission in non-human primate experiments⁵.
- An investigation of 173 contacts in 27 households demonstrated Ebola transmission only to those with direct physical contact or exposure to body fluids of the ill household member, and no transmission to the 78 household members who had no physical contact with the ill person⁶.
- An investigation of three generations of Ebola transmission during an outbreak in Uganda, demonstrated direct contact with patient body fluids as the strongest risk factor for transmission, with contaminated fomites as a possible lesser risk factor⁷.
- Several patients with viral hemorrhagic fever (VHF) have been cared for prior to being recognized as having VHFs in US and Western European medical facilities during the last several years. Although subsequently diagnosed as Lassa or Marburg fever, extensive follow-up of hundreds of potentially exposed healthcare workers including laboratory personnel, have found no instances of transmission of infection^{8,9,10,11}.
- To assist with the current outbreak in West Africa, laboratory personnel have been deployed to the European field laboratory in Guinea since mid-March, the Canadian field laboratory since June, and the two CDC laboratories since early August. Additionally, three other field laboratories set up by international partner groups are operational there. These laboratories process 200-300 specimens per day, yet there have been no documented cases of Ebola transmission to any of the laboratory scientists working at them. Earlier in the outbreak, some local West African laboratory personnel who were not wearing appropriate PPE and were performing procedures such as blood smear preparations without gloves, did acquire EVD. However, this has not occurred in any personnel wearing correct PPE and adhering to recommended procedures.
- In 1996, a physician who had been working in West Africa and an anesthetics assistant previously involved in his care, became severely ill in Johannesburg, South Africa. Despite hospitalization for more than a week before being diagnosed with Ebola, and the performance of some potentially high risk medical procedures, none of the more than 300 exposed healthcare workers, including laboratory personnel, contracted the virus¹².
- Reports in the literature of laboratory-acquired Ebola infections refer to events prior to the implementation of universal precautions and the availability of relevant safety devices such as retractable needles¹³ or to infections acquired during the performance of animal necropsy and other animal experiments¹⁴.
- Guidance documents from the UK note that one to two patients per year are diagnosed there with VHFs¹⁵. Some are not initially recognized as having VHF and are managed with standard precautions, yet there have been no reports of transmissions to health care workers. While VHF refers to a list of agents, not Ebola specifically, all are considered pathogens of “high consequence”.
- On average, routine laboratory testing is performed on a few patients per year collectively at healthcare facilities in the UK, US and Europe. In some cases dozens of samples per case are processed and tested before the patient is diagnosed with VHF. Therefore collectively in these countries since the implementation of universal precautions approximately 30 years ago, it would appear that hundreds of samples have been tested in laboratories using these procedures routinely, with no documented transmission to laboratory workers.

This document was prepared following the model developed by the New York State Department of Health and the New York City Department of Health and Mental Hygiene in

consultation with more than 40 microbiology, clinical chemistry, and hematology laboratory directors, infectious disease clinicians, epidemiologists, and scientific specialists in VHF at the CDC¹⁶.

References

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^aSuspected cases who meet the CDC criteria for Persons Under Investigation include i) travel within 21 days before illness onset to an EVD outbreak affected area (See <http://www.cdc.gov/vhf/ebola/resources/distribution-map-guinea-outbreak.html#areas> for the current list of affected areas; ii) fever (> 38.6 °C or 101.5 °F); and iii) compatible symptoms for EVD (e.g., severe headache, myalgia, vomiting, diarrhea, abdominal pain or unexplained hemorrhage).

High risk exposure is defined as either i) percutaneous, mucous membrane or direct skin contact with blood or body fluid from a confirmed or suspected EVD patient without appropriate personal protective equipment (PPE); ii) laboratory handling of body fluids from a confirmed or suspected EVD



patient without appropriate PPE or biosafety precautions, or iii) participation in funeral rites which include direct exposures to human remains in the geographic area where outbreak is occurring without appropriate PPE.

Low risk exposures are defined as i) healthcare workers in facilities that have treated confirmed or suspected EVD patients or ii) household members or others with direct contact with a confirmed or suspected EVD patient.

^bNo known exposures are defined as residence or travel to an EVD affected area without either High or Low risk exposures.