



September 10, 2014 (2nd version*)

Interim Laboratory Guidelines for Handling/Testing Specimens from Cases or Suspected Cases of Hemorrhagic Fever Virus (HFV)

(*This revision has been made by a CLP subcommittee after consultation with CDC personnel.)

On August 5, 2014, the Centers for Disease Control and Prevention (CDC) issued an Interim Guidance for Specimen Collection, Transport, Testing, and Submission for Patients with Suspected Infection with Ebola Virus Disease which can be seen here <http://www.cdc.gov/vhf/ebola/hcp/interim-guidance-specimen-collection-submission-patients-suspected-infection-ebola.html>. The Committee on Laboratory Practice (CLP) of the ASM acknowledges that specimens from suspect HFV patients may arrive at the routine testing laboratory without the knowledge of the laboratory and agrees with the CDC that all laboratory testing must follow standard precautions. In addition, the CLP has drafted this document outlining enhanced precautions which some institutions may choose to adopt in an abundance of caution in order to assure the safety of their testing personnel and to provide appropriate medically necessary laboratory testing to suspect HFV patients. This document is presented as one possible approach to testing of patients with suspected HFV. Guidelines for testing should be thoroughly discussed with the appropriate medical personnel prior to implementation and may include significant modifications of protocols recommended in this document. Contact your State Health Laboratory for any questions regarding VHF Testing and submission of specimens to the CDC (**SEE NOTE BELOW**).

General Considerations:

- A. Initial testing of patients upon presentation may be limited to CDC required tests for confirmation of Ebola or other HFV diagnosis. Additional testing may be determined upon consultation with Infectious Diseases (ID) and Microbiology. Any referral testing from suspect patients should be discussed with the referral laboratory prior to submitting specimens.
- B. All specimens taken from the patient may be labeled as ‘**SUSPECTED HFV**’.
- C. See Table 1 below for detailed description of testing that may be considered after consultation.
- D. Testing that requires specimen removal from patient’s room and transport of samples to laboratory should be kept to a minimum (**do not use a pneumatic tube system**).
- E. Specimen processing may be performed in the patient’s room, nearby in a contained testing area, inside a biological safety cabinet [BSC] located in a negative pressure room (e.g., an AFB suite) or inside a BSC located in an isolated area of the laboratory. Specimen processing should be performed while wearing appropriate PPE (impermeable gown, double gloves, eye protection, N-95 mask, shoe covers). PPE should not be re-used or leave the testing area. Place all PPE into a double-bag which contains absorbent pads soaked with bleach, then placed in a rigid plastic, impervious container for disposal (see disposal instructions at the end of this document).

Table 1 - Testing and Laboratory Procedures for Consideration:

Test	Recommendation
	Wipe specimen containers with a laboratory bleach solution, place into a double-bag that contains absorbent pads soaked with bleach then place in a biohazard rigid transport container. All transport containers should be wiped down with laboratory bleach prior to leaving the patient’s room. Laboratory processing of specimens may take place in the patient’s room or as described above in General Considerations: E.
	AS AN OPTION: Document hand-off, receipt, referral and disposal of all specimens.
Chemistry, Coagulation,	Testing may be limited to iSTAT or equivalent POC testing systems and performed in the patient’s

Hematology	room, particularly for high risk or known positive patients.
Urinalysis	Urinalysis available as a urine dipstick may be performed in the patient's room.
<p>Malaria Testing: (Rapid Malaria antigen testing may be performed in the patient's room by qualified laboratory personnel; it should be noted that this assay is not as sensitive for diagnosis as malaria smears).</p>	<ol style="list-style-type: none"> 1. Collect in a lavender top (EDTA) blood tube. 2. Preparation of thin blood smears may be done inside or outside of the patient's room. Wipe the outside of the lavender (EDTA) blood tube with bleach prior to removing from the patient's room (be careful not to remove patient identifying information). The processing steps 3 & 4 below should be done as described above in General Considerations: E. 3. Remove stopper of lavender (EDTA) blood tube with a gauze wipe soaked in bleach to prevent aerosol formation. 4. Prepare a thin blood film, fix in methanol for 30 minutes then place in dry heat at 95°C for 1 hr. to inactivate the specimen. 5. The smears can then be stained with Giemsa and read as usual. 6. WBC and platelet count can be estimated from the stained blood film. <p>NOTE: Thick smears for malaria diagnosis may be done at the discretion of the laboratory director.</p>
<p>Blood Cultures: Perform only if required and minimize blood draws for blood cultures. Use plastic bottles if available.</p>	<p>Once received in the laboratory, all specimens should be opened as described above in General Considerations: E. Wipe the outside of the bottles with bleach and inspect for any signs of breakage and positivity before loading onto the blood culture instrument or placing into an incubator for manual incubation. If the blood culture bottles are flagged as positive, or if they show any sign of positivity upon visual inspection, unload the bottles from the instrument or remove from the incubator, place the bottle(s) into a double-bag that contains absorbent pads soaked with bleach, place in a biohazard rigid plastic impervious container and process as described above in General Considerations: E.</p> <ol style="list-style-type: none"> 1. Prepare slides for Gram stain examination and allow them to dry. 2. Fix the blood smear in methanol for 30 minutes, followed by dry heat at 95°C for 1 hour to inactivate the specimen. Perform testing of the gram stain QC smear in this same manner. 3. The smears can then be stained and read as usual. <p>Do not perform any direct testing on positive blood cultures.</p> <p>Inoculate plates as per protocol based on Gram stain result.</p> <ol style="list-style-type: none"> 1. Use shrink seal (Parafilm® or other suitable plate wrap) on all sub-cultured plates, place plates in a biohazard baggie and incubate in the AFB suite (if available) in the 35°C CO₂ incubator. 2. Examine plates for growth twice per day. 3. Perform all spot testing and inoculations of appropriate ID/AST systems from isolated colonies. AS AN OPTION: If any growth occurs, subculture the organism (as described above in General Considerations: E.) onto fresh plates and incubate overnight. Work only from the sub-cultured plates to minimize risk of contact with blood from the patient.
<p>Other specimens for bacterial culture: Unless critically needed, do not perform.</p>	<p>Prepare (and transport) all specimens leaving the patient's room as previously described using laboratory bleach. All specimens should be processed as described above in General Considerations: E.</p> <p>If centrifugation is necessary, use covered carriers as for AFB processing. If specimens show signs of breakage or leakage – do not open. Consult with the Laboratory Director.</p> <p>Gram stains may be prepared as directed in the Blood culture section above.</p> <p>Seal culture plates. Perform all spot testing and inoculations of appropriate ID/AST systems from isolated colonies. AS AN OPTION: If any growth occurs, subculture the organism (as described above in General Considerations: E.) onto fresh plates and incubate overnight. Work only from the sub-cultured plates to minimize risk of contact with blood from the patient.</p>

Specimen storage:	All specimen containers should be wiped with bleach, placed into double-bags that contains absorbent pads soaked with bleach, then placed in a rigid plastic, impervious container and isolated until they can be disposed of in an appropriate manner (see specimen decontamination and disposal section below). Long-term storage of specimens is not permitted for any known suspect HFV patient.
Specimen decontamination and disposal	All specimens should be autoclaved prior to disposal. If no autoclave is available on site, contact the laboratory director for procedures for discarding of specimens and other laboratory waste.

NOTE: Contact information for state health laboratories can be found here: <http://www.aphl.org/aphlprograms/preparedness-and-response/Pages/Emergency-Lab-Contacts.aspx>.

Additional information regarding procedures for the collection, handling, and testing of specimens for EVD (Ebola) and sending specimens to the CDC for EVD testing have been issued by the CDC and are posted at the following site:

<http://www.cdc.gov/vhf/ebola/hcp/interim-guidance-specimen-collection-submission-patients-suspected-infection-ebola.html> and <http://www.cdc.gov/ncezid/dhcpp/vspb/specimens.html>

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