



Public Health and Environmental Laboratories Technical Guidance

January 2020

Update: Ebola Testing Algorithm, NJDOH Recommendation Regarding FDA EUA Tests and Clinical Laboratory Testing

Links:

[Packaging and Shipping Online Training brochure](#)

[NJDOH BioThreat Response Laboratory LAB-05 with Chain of Custody](#)

[CDC Viral Special Pathogens Branch form](#)

Attachments:

NJDOH Shipper’s Certification for Ground Transport – Attachment I

World Courier Account Application Form – Attachment II

Table of Contents:

Acronyms _____ page 2

Purpose _____ page 3

PREPAREDNESS: Acute Health Care Facility Designations _____ page 3

PREPAREDNESS: Clinical Laboratory Testing _____ page 4

PREPAREDNESS: Clinical Laboratory Risk Assessment and Mitigation _____ page 4

PREPAREDNESS: Risk Assessment – Summary of Clinical Laboratory Hazards _____ page 5

RESPONSE: Persons Under Investigation (PUI) – Initial Assessment _____ page 7

RESPONSE: Consultation with Public Health Authorities: CDS and CDC EOC _____ page 8

RESPONSE: Specimen collection, packaging and shipping to NJ PHEL _____ page 8

RESPONSE: Ebola virus testing at NJ PHEL _____ page 11

RESPONSE: Follow-up Testing at CDC Viral Special Pathogens Branch _____ page 13

References _____ page 14

Acronyms:

APHIS	Animal and Plant Health Inspection Service
BSC:	Biosafety Cabinet
BTRL:	BioThreat Response Laboratory of the PHEL
CDC:	Centers for Disease Control and Prevention
CDS:	New Jersey Communicable Disease Service
DASH	Data and Specimen Handling Form
DSAT:	Division of Select Agents and Toxins (CDC/APHIS)
EAH	Ebola Assessment Hospital
ETC	Ebola Treatment Center
EUA	Emergency Use Authorization.
EVD	Ebola Virus Disease
FDA	Food and Drug Administration
Frontline	Acute Care Hospital, emergency care settings, or urgent care clinic
IZDP:	Infectious and Zoonotic Disease Program of the CDS
LRN	Laboratory Response Network
PHEL:	New Jersey Public Health and Environmental Laboratories
PHL	Public Health Laboratory
PUI	Persons Under Investigation
RT PCR	Real Time Polymerase Chain Reaction
TAT	Turn Around Time from specimen receipt to test results
USDOT	United State Department of Transportation
VHF	Viral Hemorrhagic Fever
VSPB	Viral Special Pathogens Branch (CDC)

Purpose:

This guidance provides specific recommendations that will assist healthcare facilities, state and local health departments in developing preparedness plans for patients under investigation (PUIs) for Ebola virus disease (EVD). Context for this guidance document is provided in CDC's [Interim Guidance for U.S. Hospital Preparedness for Patients under Investigation and with Confirmed Ebola Virus Disease: A Framework for a Tiered Approach](#).

To create a coordinated, networked approach, state and local health officials, in collaboration with hospital and healthcare facility executives, designated healthcare facilities across the state to serve in one of three tiers as outlined in this guidance document.

PREPAREDNESS: Acute healthcare facilities designations

- [Frontline healthcare facilities](#)
- [Ebola assessment hospitals](#)
- [Ebola treatment centers](#)

1. Frontline Healthcare Facilities

Most U.S. acute care facilities that are equipped for emergency care (such as hospital-based emergency departments and other emergency care settings including urgent care clinics and critical access hospitals) are in this tier. Frontline healthcare facilities do not include primary care offices and other nonemergent ambulatory care settings. Preparedness guidance for these settings can be found at: [CDC Guidance for Clinicians](#).

It is possible that persons with unrecognized EVD will present to a Frontline healthcare facility (an acute care hospital or other emergency care setting including urgent care clinic, or critical access hospital) without prior notification. These facilities should be prepared to promptly identify and isolate these patients according to the [CDC's guidance for emergency departments](#). Frontline healthcare facilities are not expected to provide prolonged care (>12–24 hours) for a severely ill patient. It is important to remember that because of the potential stigma associated with EVD, patients returning from affected countries may be reluctant to disclose their travel history. Clinical laboratories should be prepared to provide sufficient testing to ensure patient care is not compromised while patients undergo assessment. The clinician should determine specific testing according to the patient presentation and travel history.

2. Ebola Assessment Hospitals

Ebola assessment hospitals are facilities prepared to provide up to 96 hours of evaluation and care for PUIs until the diagnosis is either confirmed or ruled out and until discharge or transfer is completed. Ebola assessment hospitals should be prepared to transport patients with confirmed EVD to an Ebola treatment center. Ebola assessment hospitals may also receive patients transferred from frontline healthcare facilities that are not prepared to provide evaluation, arrange for testing, and care for PUIs. Ebola assessment hospitals should ensure there is no delay in the care for these patients by being prepared to test, manage, and treat alternative etiologies of febrile illness (malaria, influenza) as clinically indicated.

3. Ebola Treatment Centers

Ebola treatment centers are facilities that plan to care for and manage a patient with confirmed EVD for the duration of the patient's illness. State and local decisions to designate Ebola treatment centers are informed by the results of a CDC site visit conducted by an interdisciplinary team of subject matter experts. Site visits assess the hospitals' ability to meet the minimum criteria (including infection control capacity, physical infrastructure, staffing resources, PPE supplies, waste management processes, worker safety training, environmental services, and laboratory set up). Staff

must be trained in and have practiced putting on and taking off (donning and doffing) PPE for Ebola, as well as providing clinical care using PPE.

PREPAREDNESS: Clinical Laboratory Testing

Clinical laboratories, especially those in Ebola Assessment and Ebola Treatment Hospitals, should be prepared to provide a timely and minimum menu of testing to ensure that medical evaluation is not delayed for any patient. Aside from the clinical symptoms the decision to perform the Ebola assay is based on some key clinical laboratory test results. The basic diagnostic tests of interest are:

1. A complete blood count (CBC), including differential, and platelet count
2. Sodium, potassium, bicarbonate, blood urea nitrogen, creatinine, and glucose concentrations
3. Liver function tests
4. Coagulation testing, specifically prothrombin time (PT), expressed as international normalized ratio (INR)
5. Urinalysis (dipstick)
6. Blood culture for bacterial pathogens
7. Malaria testing (smear or rapid testing or PCR)
8. Influenza virus testing during periods when influenza prevalence is high

***NOTE: Diagnostic immunoassays and molecular tests for Ebola virus infection are commercially available. Some laboratories may thus have the capability to perform testing for Ebola virus infection. While a decision to pursue such capability ultimately lies with individual laboratories, these tests should not be used as a replacement for testing performed at NJ PHEL and CDC (see RESPONSE section). The steps outlined below for diagnostic testing of PUI for Ebola infection should still be followed despite the results of any in house testing that is performed to evaluate for Ebola virus infection.**

If frontline healthcare facilities cannot immediately transfer the patient to another facility, they should have plans in place to ensure that routine laboratory tests needed to determine alternative diagnoses are performed while the patient is being evaluated for EVD.

Provisions should be made to perform these tests on site without delay when clinically necessary. CDC has repeatedly indicated that proper donning and doffing with PPE, strict adherence of laboratory staff to standard laboratory safety precautions and decontamination procedures are adequate for [safe processing](#) in the laboratory of specimens from patient under investigation (PUI) for Ebola.

A risk assessment should be initiated by each institution to determine isolation procedures, location and placement of PUI, specimen collection, on site test menu, and patient and specimen transport.

PREPAREDNESS: Clinical Laboratory Risk Assessment and Mitigation

Laboratory risk assessment is a process used to identify:

1. the hazards associated with a known or potentially infectious agent and the activities being conducted with them;
2. the likelihood of a person's exposure to that agent or material;
3. the consequences of such an exposure to personnel or equipment (e.g., a laboratory acquired infection or the need to take a machine off-line for extended periods).

A risk assessment of all processes, procedures, and activities must be performed to determine the potential for exposure to the specimen through generation of aerosols, sprays, splashes, or spills. Based on the assessment, a plan to mitigate the

identified risks should be implemented using engineering controls, administrative controls (including work practices), and the use of appropriate PPE.

CDC is aware of hospitals that have safely used instruments in their core laboratories to test specimens when EVD is a concern. However, following risk assessment, laboratories may choose to use point of care testing or other alternative procedures to minimize disruption to the core laboratory and minimize risk to laboratory personnel.

items for clinical laboratories to focus on during their site-specific risk assessment should include:

- [Specimen management and transport](#), including the path of the sample through the laboratory particularly avoiding transport through high-traffic areas or pneumatic tube systems
- Equipment hazards (e.g., the potential for creating aerosols, sprays, splashes of the specimen when performing testing and using equipment)
- Biological Safety Cabinet certification, operation and safe work practices
- Decontamination procedures, including spill response, and methods for [decontamination of equipment](#)
- Infectious waste management
- Laboratory design
 - Laboratories that have open room designs should also consider the risk of exposure to workers present in the area but that are not directly involved with testing of that sample
 - Some recommended measures to minimize the risk of laboratory transmission when testing patient specimens include: limiting the number of staff engaged in testing, evaluating and segregating equipment used for testing, and performing testing in a dedicated space
- Engineering controls and safety equipment
- Laboratory communication protocols
- Laboratory entry and exit procedures
- PPE selection and use
- Facility ventilation and filtration
- Employee medical surveillance and exposure response
- Safe sharps handling
- Personnel safety training and competencies

PREPAREDNESS: Risk Assessment Summary of Clinical Laboratory Hazards

Procedure	Recommendation
Centrifugation	Should be performed with biohazard sealed buckets or sealed rotor. The buckets or rotor should be opened inside a certified and operable BSC.
Homogenization	Procedures requiring homogenization of any specimen type should be avoided or performed with extreme care due to the risk of spray or splash, and should be performed inside a certified and operable BSC
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 ¹⁵ . For automated instruments, decontamination procedures should be those advised by the manufacturer or vendor for enveloped viruses.

Malaria testing	<p>Thin blood smears should be fixed in methanol for 15-30 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 parasite morphology.</p> <p>Thick blood films should not be hemolyzed with water, but should be stained with Giemsa stain that includes Triton X-100 to inactivate Ebola virus.</p> <p>Validated malaria PCR assays that have been approved by the Clinical Laboratory Evaluation Program for clinical use may be used to detect malarial parasites.</p> <p>Malaria antigen detection kits may assist with initial urgent assessment but must be recognized as being inherently less sensitive than smear microscopy or PCR, at least one of which must be performed as soon as possible.</p> <p>The effects of some inactivation/decontamination procedures on the performance of some rapid antigen tests for malaria have been investigated¹⁶.</p> <p>For more detailed guidance, see the CDC recommendations on Malaria testing for suspected Ebola patients at: http://www.cdc.gov/malaria/new_info/2014/malaria_ebola.htm</p>
Blood Cultures	Systems using plastic blood culture bottles are preferred. Blood culture in glass bottles should be avoided.
Other specimens for bacterial culture	"Pan-cultures" should not be performed. Procedures essential for patient management should be performed in a BSC with the use of appropriate PPE. Identification or characterization of subsequently cultured bacteria or fungi can be performed with standard precautions.
Wet preps	Should be avoided.
Viral cultures	DO NOT perform viral culture, including the use of rapid culture systems, on any specimen.
Pre-transfusion testing	Please refer to the American Association of Blood Banks' Ebola information sheet www.aabb.org/programs/clinical/Pages/Infection-Control-for-Handling-Blood-Specimens-from-Suspected-Ebola-Patients.aspx
Post-mortem examinations	Should only be performed under the explicit recommendation of the CDC and with their guidance. In the event of a fatality in a suspected or confirmed EVD patient, the Northern Regional Medical Examiner's Office must immediately be contacted at 973-648-4500 and request to have someone from the Office of the Chief State Medical Examiner contact them due to an EVD decedent.

Specimen storage	<p>With the exception of circumstances where retention is required by regulations, <u>long-term storage of specimens is discouraged</u>. It is recommended that specimens collected from suspected or presumptive positive EVD cases be isolated from other specimens in the laboratory and if stored, refrigerator or freezer unit must be kept locked at all time. A chain of custody form must ALWAYS accompany the specimen. Immediately after testing has been completed and the sample has been confirmed by the CDC, the samples should be disposed of in compliance with Waste management protocol below.</p> <p>Note – details of specimen decontamination and disposal should be documented for any samples from a presumptive or confirmed positive EVD patient, or a PUI of unknown status. The CDC <u>does not</u> classify samples tested and resulted as presumptive positive for Ebola Zaire RNA by the NJ PHEL Ebola Virus NP Real-Time RT-PCR Assay; Ebola Virus VP40 Real-Time RT-PCR Assay, as select agent samples, that classification being reserved for positive cultures confirmed by the CDC only, they do reserve the right to request information and confirmation of destruction/disposal.</p>
Specimen decontamination and disposal	<p>Wet a piece of gauze with a U.S. Environmental Protection Agency (EPA)-registered hospital disinfectant with a label claim for a non-enveloped virus (e.g., norovirus, rotavirus, adenovirus, poliovirus) and wipe the outside of the specimen container. The gauze and the disinfected specimen container should then be placed in a plastic bag and packaged with other contaminated waste for appropriate disposal or autoclaving.</p> <p>A list of EPA-registered disinfectants can be found at: http://www.epa.gov/oppad001/list-l-ebola-virus.html</p> <p>Note: Bleach or acidic chemicals must NOT be mixed with Trizol or any other reagent containing guanidine isothiocyanate, nor should they be disposed of together in the same container, as reactive compounds and toxic gases are formed if they interact.</p>

RESPONSE: Persons Under Investigation (PUI) – Initial Assessment

A person who has both consistent signs or symptoms and risk factors as follows should be considered a [Person Under Investigation \(PUI\)](#):

1. Elevated body temperature or subjective fever or symptoms, including severe headache, fatigue, muscle pain, vomiting, diarrhea, abdominal pain, or unexplained hemorrhage; **AND**
2. An [epidemiologic risk factor](#) within the 21 days before the onset of symptoms.
<https://www.cdc.gov/vhf/ebola/pdf/could-it-be-ebola.pdf>

If a diagnosis of EVD is being considered, the patient should be isolated in a single room (with a private bathroom), and healthcare personnel should follow [standard, contact, and droplet precautions](#), including the use of appropriate [personal protective equipment \(PPE\)](#). Infection control personnel should be contacted immediately.

If EVD is suspected, contact the local or state health department immediately for consultation and to assess if testing is indicated and the need for initiating identification of contacts.

RESPONSE: Consultation with Public Health Authorities: CDS and CDC Emergency Operation Center

NJ Procedure for communication and approval of specimen submission to PHEL

1. Contact Public Health Authorities

Hospitals should contact their state and/or local health department before contacting CDC.

Each patient will require evaluation on a case by case basis. Hospital officials should contact the local health department within the patient's jurisdiction. See link for current directory: <http://nj.gov/health/lh/directory/lhselectcounty.shtml> The local health department will contact the Communicable Disease Service.

If the hospital is unable to reach local health officials, contact the [Communicable Disease Service](#) directly at 609-826-5964 (business hours) or 609-392-2020 (after hours).

Local health officials, the Communicable Disease Service and the CDC will determine the need for testing and where the specimens should be sent. The New Jersey Public Health Laboratory has the capacity to perform the Ebola Virus NP Real-Time RT-PCR Assay; Ebola Virus VP40 Real-Time RT-PCR Assay and will be the site for initial testing of the blood specimen.

Specimens **other than blood** may be directly submitted to the CDC after consultation with the Viral Special Pathogens Branch (470-312-0094).

2. Receipt of Testing Approval

If public health authorities determine a need for testing the patient, the submitter will be given a CDS approval (case) number. This number should be used on all paperwork and specimen labels and will be used to track information regarding the case.

Alert the laboratory to prepare materials for blood draw and shipment and to make sure that they have 2 certified shippers on hand to package the specimen for transport. Order the test in your system and create specimen labels.

RESPONSE: Specimen collection, packaging and shipping to NJ PHEL

It is advisable to prepare materials and alert staff prior to specimen receipt in the clinical laboratory

a. Materials Needed:

- 3 or 4 ml lavender top EDTA **plastic** tubes
- Small clear biohazard Ziploc bags
- Absorbent material

- Ambient Category A shipping system: [Triple Packaging system](#) - Outer UN Certified Class 6.2 box with internal support for secondary container, secondary container (rigid plastic screw cap), bubble wrap to support specimens inside secondary container, USDOT Infectious Substance label, e.g. Infecon 3000
 - Overpack box - Styrofoam lined to accept the inner ambient container e.g. Infecon 6000 plus Overpack label
 - Sufficient cooled gel packs to fill Infecon 6000 (note, gel packs may be refrigerated or frozen – specimens is required to be at 2-8 C, however, freezing is not a problem for this analysis
 - Dangerous Goods Shippers Declaration and LAB-05 form
- b. **Two persons certified within the past two years** to ship Category A Infectious substances – persons should be on call to ship 24/7. Note: FREE online Packaging and Shipping training available at www.cdc.gov/labtraining.
- c. **Materials Needed Prior to Specimen Collection**

Prepare two Ebola Go-Kit - one for PATIENT ROOM and one for STAGING AREA.

- **For PATIENT ROOM (Hot Zone, Full PPE is required)**
 - EDTA tubes,
 - two Ziploc bags, each containing sufficient absorbent material to absorb the entire contents of the tube
 - Specimen labels
 - spray bottle with 10% v/v bleach prepared fresh daily.
- **For STAGING AREA (Warm Zone, Full PPE required if adjacent to patient room, laboratory PPE required if remote from patient room)**
 - Separate spray bottle with 10% v/v bleach prepared fresh daily
 - Disposable gloves
 - rigid specimen carrier (to carry specimen from patient area to staging area)
 - Rigid plastic screw cap secondary container from UN certified Class 6.2 ambient shipping system
- The remainder of the preparation for specimen collection can be done in a clean area in the laboratory with no gloves and a clean lab coat. The remainder of the materials for shipping should remain in a clean area in the laboratory and should NOT be brought into either the patient's room or the staging area where specimen collection and decontamination occur.

Specimen Collection

Have the laboratory prepare an "Ebola Go Kit" to bring into the patient area.

- a. Supply patient care area with two purple top tubes, Ziploc bags with absorbent material, specimen labels and spray bottle with freshly prepared 10%v/v bleach (prepared fresh daily)

INSIDE THE PATIENT ROOM

- b. Draw two (3 or 4 ml) plastic EDTA tubes. Fill tubes.
- c. Decontaminate the outside of the tubes.
- d. Label the tubes with patient name, hospital ID#, DOB, date, time, collectors initials and CDS approval number.
- e. Place each tube in a separate Ziploc bag with absorbent material.
- f. Remove all air from the bag.
- g. Decon the outside of the bags with 10%v/v bleach.
- h. Hand the bags to person transporting specimen to staging area to place inside a rigid specimen carrier.
- i. Staging area may be near patient care area or in the laboratory.

Package Blood: Begins in Staging Area, Completed in Clean Area

STAGING AREA:

- a. Staff should always wear PPE appropriate to the risk. If a Biological Safety Cabinet is available, the next step should be conducted in the cabinet.
- b. Gloves, impermeable lab coat and face protection are required if working outside the BSC. Skin, eyes, nose and mouth should be barrier protected.
- c. Gloves, impermeable lab coat are required if working inside the BSC. The BSC can serve as a shield, however, face protection should be worn if desired.
- d. Remove the deconned Ziploc bags from the rigid specimen carrier.
- e. Have a second person read the patient ID information to you to check against the labels. Do not touch any paperwork.
- f. Decon the outside of the bags again with 10%v/v bleach and change gloves before handling again.
- g. Wrap each Ziploc bag individually in bubble wrap.
- h. Place both Ziploc bags inside the Category A rigid plastic secondary container.
- i. Screw the container shut and decon the outside of the container with 10% bleach. Remove gloves and hand container to person in clean area to finish packaging.

CLEAN AREA SPECIMEN PACKAGING:

- a. Finish packaging the specimen as Category A according to the IATA packaging instruction. Use an overpack for cold packs to maintain the temperature at 2-8 C.
- b. Prepare three copies of the Shipper's Certification for Ground Transport and one copy of the LAB-05 with Chain of Custody.

- c. Specimen Storage: Accession, package and ship immediately after collection. If there is a delay in shipping, the package may be stored at 2-8°C until picked up by the courier. Secure package in a locked refrigerator until signed for by the courier.
- d. Have the courier sign the Chain of Custody. Keep a copy of all paperwork.

SPECIMEN TRANSPORT TO PHEL

- Specimen may be transported via hospital or PHEL emergency courier for same day delivery OR FedEx overnight.
- **All specimens that are being transported from the hospital laboratory to NJ PHEL should be labelled as Infectious substance, affecting humans (Suspected Category A Infectious Substance) on the shipping papers and on the outer container.**
- Couriers must use the COC form.
- The bag with labels/forms must be attached to the package.
- The sentinel laboratory must notify the BTRL Manager before courier leaves the pickup site,

RESPONSE: Ebola virus testing at NJ PHEL

In response to the international threat that an Ebola outbreak poses, the CDC have developed an assay, Ebola Virus NP Real-time RT-PCR Assay and Ebola Virus VP40 Real-time RT-PCR Assay that gained FDA approval for use under Emergency Use Authorization (EUA). This assay is intended for the in vitro qualitative detection of Ebola virus RNA (species *Zaire ebolavirus*) in clinical specimens from individuals meeting Ebola virus clinical and/or epidemiological criteria, in qualified laboratories designated by the CDC, including NJDOH PHEL.

The New Jersey Department of Health (NJDOH) Public Health and Environmental Laboratories (PHEL) performs the Laboratory Response Network (LRN) and Centers for Disease Control & Prevention (CDC) Ebola Virus NP Real-Time RT-PCR Assay; Ebola Virus VP40 Real-Time RT-PCR Assay. Note that this test is specific for *Zaire ebolavirus* species) and does not detect other Ebola species or other VHF etiologies.

This assay is to be used for testing individuals designated by the NJ Communicable Disease Service in conjunction with the CDC as Persons under Investigation (PUI) for Ebola Viral Disease (EVD). Additional assays for testing known Ebola cases or those suspected of having other viral hemorrhagic fevers are available at the Centers for Disease Control and Prevention (CDC) Viral Special Pathogens Branch (VSPB).

Two full, 3 or 4 mL lavender top EDTA tubes of blood are required. One tube is used for initial test for Ebola at the State Public Health Laboratory; the second tube is for confirmatory testing at CDC for positives or when special studies at CDC are required on negative samples. These samples cannot be shared or aliquoted for other laboratory tests.

Turn Around Time of the Ebola Assay at NJ PHEL

Within 24 hrs. after receipt at PHEL (most specimens received by 8 am are tested and resulted by PHEL by 6 pm):

- TAT by definition is the longest time that it could take to “turn a specimen around” and produce a result after a specimen is received in the laboratory. The TAT for the initial Ebola Virus NP Real-Time RT-PCR Assay; Ebola Virus VP40 Real-Time RT-PCR Assay **after the specimen is received in the PHEL laboratory will not exceed 24 hours.** The TAT will vary (within the 24 hours), depending on:
 - When the specimen is received (between 8AM and 5PM or after regular PHEL business hours).

- Whether there are problems associated with the specimen collection, handling, packaging and paperwork requiring correction before analysis can proceed.
- Time the alert to on-call laboratory staff is provided for off hours testing requests. Time is needed to communicate to all partners in the chain to assemble staff for testing during off peak hours and for staff travel time to the laboratory.
- Samples that are positive in the initial Ebola Virus NP Real-Time RT-PCR Assay; Ebola Virus VP40 Real-Time RT-PCR Assay are shipped to the CDC using World Courier. Given the limitation in shipping positive Ebola samples it is expected that sample may not arrive at the CDC until 24 to 48hrs after the initial result is obtained on a weekday and longer if initial result is obtained on a Friday unless the case is emergent enough that the CDC provides a mechanism for urgent delivery of the specimen. **Once the sample is received at the CDC a panel of tests is performed and reported within 24 hours.**
- All of the above emphasize the importance of strict adherence to protocols, rechecking at all steps in the collection packaging, shipping, completion of forms and submission to minimize problems that could delay the TAT and impact patient care.

Results interpretation

Results will be reported using LRN Results messenger and will be emailed to the submitter.

If the initial specimen tests negative: A negative test presumes that Ebola virus RNA was not present in the specimen at the detection level of the assay. However, negative results do not rule out Ebola, and should not be used as the sole basis for treatment or other patient management decisions. The clinical features of the illness and the type and risk of exposure are the keys to making patient management and isolation decisions. A negative EBOV VP40 rRT-PCR Assay test result should not be interpreted as demonstrating that the patient does not have Ebola, particularly if it has only been a few days since the onset of symptoms. The possibility of a false negative result should especially be considered if the patient's recent exposures or clinical presentation indicate Ebola is likely, and diagnostic tests for other causes of hemorrhagic illness are negative. Risks to a patient of a false negative result include: delayed treatment, potential lack of treatment, or stopping treatment too soon. If Ebola is suspected by exposure history together with clinical findings, re-testing should be considered in consultation with public health authorities.

For negative results on specimens collected less than 3 days post-onset of symptoms, and if the patient is still symptomatic, repeat testing is recommended, unless EVD is no longer in the differential diagnosis. Requests for repeat testing must be approved through the Communicable Disease Service. Testing for other viral hemorrhagic fevers at the CDC Special Pathogens Branch must be arranged through the Communicable Disease Service.

If the initial specimen tests positive:

Samples that test positive using this assay are considered presumptive positive for Ebola Virus NP Real-Time RT-PCR Assay; Ebola Virus VP40 Real-Time RT-PCR Assay and will be submitted to CDC for confirmatory testing.

A positive test result from the EBOV VP40 rRT-PCR indicates that RNA from Ebola virus was detected, and the patient is presumed to be contagious. Laboratory test results should always be considered in the context of clinical observations and epidemiologic data in making a final diagnosis and patient management decisions. For information on Ebola and guidelines on patient management, please refer to: <http://www.cdc.gov/vhf/ebola/index.html>.

RESPONSE: Follow-up Testing at CDC Viral Special Pathogens Branch

The need for follow-up testing will be determined in consultation with the New Jersey Communicable Disease Service and the CDC. If further testing is required, the New Jersey Public Health Laboratory will arrange the transport of presumptive positive specimens to the CDC via World Courier.

In the event of an emergency where laboratories receive instructions directly from the CDC or the NJ DOH CDS to ship specimens directly to the CDC (World Courier has been identified as the sole source for transporting samples to the CDC VSPB).

Specimens other than blood may be directly submitted to the CDC after consultation with the Viral Special Pathogens Branch at 470-312-0094.

Packaging and Shipping Specimens from PUI to the CDC

- **Reminder NO specimens will be accepted without prior consultation with the CDC or NJCDS and should only be shipped directly to the CDC if instructed to do so.**
- Maintain a minimum of two certified shippers as described above.
- Establish an account with World Courier. Currently World Courier is the only courier which will carry presumptive positive Ebola specimens. New account application form is attached.

Contact:

Donald Derle, World Courier, an AmerisourceBergen company, Director, Business Development/RTP
378 Center Point Circle, Altamonte Springs, FL 32701
Tel: 1-407-695-6501, Fax: 1-407-695-6507
www.worldcourier.com

- Include the CDC Infectious Disease (CDC Form 50.34) and [specimen submission forms pdf icon](#)[PDF – 182KB, 508].
- Label the outer packaging in accordance with I.A.T.A. regulations to prevent leakage (triple packaging).
- All specimens – being transported to the CDC should be labelled as Infectious substance, affecting humans (Suspected Category A Infectious Substance) on the shipping papers and on the outer container.
- On the **outside** of the box, specify how the specimen should be stored: **refrigerated** or **frozen**.
- Send specimens by overnight courier (World Courier). International submitters should consider door-to-door shipment via air transport to expedite specimen delivery to CDC.

Forms required for follow-up testing at CDC:

- CDC DASH pre-filled with NJ Contact information) and
- CDC VSPB form, (Linked) and hospital chain of custody form. NOTE; CDC DASH FORM IS SEPARATE FROM THIS DOCUMENT

The CDC DASH form must be used AS IS - with all the NJDOH contact information in the top right corner intact. Altering this NJDOH information in the form may result in delays in analysis

References

1. Bausch DG, Towner JS, Dowell SF, et al (2007): Assessment of the risk of Ebola virus transmission from bodily fluids and fomites. *J Infect Dis* 196 Suppl 2: S142-7.
2. Alimonti J, Leung A, Jones S et al (2014): Evaluation of transmission risks associated with in vivo replication of several high containment pathogens in a biosafety level 4 laboratory. *SciRep* 4:5824. doi: 10.1038/srep05824.
3. Dowell SF, Mukunu R, Ksiazek TG, et al (1999): Transmission of ebola hemorrhagic fever: a study of riskfactors in family members, Kikwit, Democratic Republic of Congo, 1995. *J Infect Dis* 179 (Suppl 1): S87-91.
4. Francesconi P, Yoti Z, Declich S, et al (2003): Ebola Hemorrhagic Fever Transmission and Risk Factors of Contacts, Uganda. *Emerg Infect Dis* 9:1430-37
5. Centers for Disease Control and Prevention (CDC) et al (2004): Imported Lassa Fever – New Jersey, 2004. *MMWR* Oct 1, 53(38): 894-97.
6. Centers for Disease Control and Prevention (CDC) et al (2009): Imported case of Marburg Hemorrhagic Fever – Colorado, 2008. *MMWR* Dec 18, 58(49): 1377-81
7. Timen A, Koopmans MPG, Vossen ACTM et al (2009): Response to imported case of Marburg Hemorrhagic Fever, the Netherlands. *Emerg Infect Dis* 15: 1171-75.
9. Amorosa V, MacNeil a, McConnell R et al (2010): Imported Lassa Fever, Pennsylvania, USA, 2010. *Emerg Infect Dis* 16: 1598-1600.
10. Richards G, Murphy S, Jobson R, et al (2000): Unexpected Ebola virus in a tertiary setting: Clinical and epidemiologic aspects. *Crit Care Med* 28: 240-44.
11. Personal communication, Aaron Devries, Minnesota Department of Health.
12. UK Department of Health, Advisory Committee on dangerous pathogens, Management of Hazard Group 4 viral hemorrhagic fevers and similar human infectious diseases of high consequence. Appendix 7: Laboratory Procedures.
13. Emond RTD, Evans B, Bowen ETW and Lloyd G (1977): A case of Ebola virus infection. *BMJ* 2: 541-44.
14. Formenty P, Hatz C, Le Guenno B, et al (1999): Human Infection Due to Ebola Virus, Subtype Côte d'Ivoire: Clinical and Biologic Presentation. *J Infect Dis* 179(Suppl 1): S48-53.
16. Rollin et al. 2011. Arenaviruses and Filoviruses. In: *Manual of Clinical Microbiology. (10th ed)*. ASM Press.
17. Hersberger M, Nusbaumer C, Scholer A et al (2004): Influence of Practicable Virus Inactivation Procedures on tests for frequently Measured Analytes in Plasma. *Clin Chem* 50: 944-46.
19. Loutfy MR, Assmar M, Burgess DCH, and Kain KC (1998): Effects of Viral Hemorrhagic Fever Inactivation Methods on the Performance of Rapid Diagnostic Tests for *Plasmodium falciparum*. *J Infect Dis* 178: 1852-55.

Shipper's Certification for Ground Transportation of Hazardous Materials

(To be completed when transporting hazardous materials by hospital courier)

(One signed copy retained by shipper for 375 Days)

Shipper's Reference Number(s) _____

Shipper: (Name) _____

Shipper's Address: _____

(Include Street number- Street- City- State- Zip Code)

Consignee: (Name) _____

Consignee's Address: _____

(Include Street number- Street- City- State- Zip Code)

Nature and Quantity of Hazardous Material:

Hazardous Materials Identification				Quantity, Number and Type of Packages	Packaging Instruction (#)
UN No.	Proper Shipping Name (technical name)	Hazard Class or Division	Packing Group		

Emergency response telephone number: _____

SHIPPER'S CERTIFICATION: "This is to certify that the above-named materials are properly classified, described, packaged, marked and labeled, and are in proper condition for transportation according to the applicable regulations of the US Department of Transportation."

Printed Name/Title of Signatory: _____

Place: _____

Signature: _____ Date: _____

Attachment II

**New Customer Account
Application Form**



CUSTOMER BILLING INFORMATION

COMPANY NAME:

TAX ID / FEIN Number:

Company Registered Address	Street:		
	Additional Add:		
	City:	State:	Post Code:

Billing Address (if different to Registered)	Street:		
	Additional Add:		
	City:	State:	Post Code:

Payer / Parent Details (if different to Registered & Billing)	Payer Company Name:		
	Street:		
	Additional Add:		
	City:	State:	Post Code:

ACCOUNTS PAYABLE CONTACT DETAILS and INVOICE REQUIREMENTS

CONTACT NAME:

EMAIL ADDRESS:

LOCATION ADDRESS: REGISTERED BILLING PAYER

Telephone:

Fax:

STATEMENT email address:

INVOICE email address (if different):

World Courier Net Terms (From date of invoice) is 30 Days.

Monthly Est.Sales Revenue: 4k or less

More than 4k (Please indicate amount)

New Customer Account Application Form



Reference Required on the Invoice: Yes No

If YES, please specify.

Any other special INVOICE requirements i.e. PO Number etc...? Yes No

If YES, please specify:

World Courier will not be responsible for providing missing or incorrect Purchase Order numbers or other required references. Payment cannot be withheld for this reason.

CCSF&CONDITIONS OF CARRIAGE

Is this account an IAC (Indirect Air Carrier) Yes NO

If YES, Please provide IAC#

Is the account a CCSF (Certified Cargo Screening Facility)? YES NO

Have you applied to become a CCSF? Yes NO

Shipments are subject to World Courier's current CONDITIONS OF CARRIAGE, refer to: www.worldcourier.com

GENERAL INFORMATION

STATUS: Public Ltd Private Ltd. Partnership Sole Trader

Non- Profit Organisation Other

Is the Company part of a group? Yes No if yes,(please complete below)

Parent Company Details:

NATURE OF BUSINESS:

Type of commodities to be shipped with WORLD COURIER (please tick all that apply):

Electronics Chemicals (non Pharma) Spare Parts / AOG

Pharmaceuticals* Food

Biological - Animals Biological Human Synthetic Material

Page 2 of 3

New Customer Account Application Form



Temperature – Controlled Dangerous Goods Radioactive

Other (please specify)

CONDITIONS OF CARRIAGE

All shipments are subject to World Courier's current CONDITIONS OF CARRIAGE, refer to: www.worldcourier.com

I/We confirm that the above information is both true and correct. I/We understand that all work undertaken by World Courier is subject to the conditions of carriage.

I/We understand that advanced payment may be required for the initial shipment with World Courier.

Authorized Signature:		Date:	
Printed Name:			
Position:			

Applicant authorizes World Courier and any credit agency or any service engaged by World Courier to obtain, verify or otherwise investigate any information, reference, statements, credit reports or other information obtained with respect to Applicant as World Courier deems appropriate.

Attachment III LAB-5 Clinical Specimen Rule out for Select Agent and Chain of Custody



New Jersey Department of Health, Public Health and Environmental Laboratories
REQUEST FOR TESTING OF CLINICAL SPECIMENS FOR SUSPECTED PATHOGENS OF PUBLIC HEALTH SIGNIFICANCE AND CHAIN OF CUSTODY

PHEL Use Only
BT-H

IMPORTANT: All sample/specimen submitters must email a copy of the completed LAB-5 form to DOH-BTEPI-PHEL@doh.nj.gov prior to shipment in addition to the required hard copy. Specimens must be pre-approved by the Communicable Disease Service (800-826-5964) prior to submission. Additional sheets or documentation may be attached if needed.

CDRSS Case number: _____

Name of requesting agency/institution: _____

Address: _____

City: _____

State: _____ Zip: _____

Phone: _____ Fax: _____

Email: _____

Name of Submitter: _____

Specimen/sample collected by: _____

Collection pickup site: _____

Collection Date: _____ Time: _____

Date shipped to PHEL: _____

Attending Physician: _____

Physician address: _____

Physician Email: _____

Physician Phone: _____

SPECIMEN INFORMATION:

Suspected agent(s):

- Bacillus anthracis*
- Brucella* spp.
- Burkholderia* spp.
- Coxiella burnetii*
- Ebola Virus
- Francisella tularensis*
- Orthopox
- Yersinia pestis*
- Antibiotic resistant isolate
- Other: _____

Type of specimen/sample:

- Culture-Bacteria
- CSF
- Other: _____
- Whole Blood
- Serum
- Urine

PATIENT INFORMATION:

Patient Name: _____

Sex: M F DOB/Age: _____

Travel in the past 6 months (locations & dates): _____

Date of symptom onset: _____

Pregnancy status at onset (trimester): 1st 2nd 3rd N/A

Is the patient hospitalized? Yes No

Is the patient alive? Yes No

Did the patient experience skin lesions? Yes No

Lymphadenopathy? Yes No

Dyspnea? Yes No

Fever? Yes No

Were there any positive blood cultures? Yes No

Other signs/symptoms: _____

Were any specimens handled outside of a biosafety cabinet?

Yes No

Biochemical Information (bacterial isolates):

Gram positive Yes No

 Large rods Yes No

Gram negative Yes No

Coccobacilli Rods Curved

Rapid growth on blood agar Yes No

Poor growth after 24h Yes No

Growth on MacConkey Agar Yes No

 Lactose fermentation Yes No

Hemolytic Yes No

Motile Yes No

Oxidase positive Yes No

Catalase positive Yes No

Urease positive Yes No

Indole negative Yes No

Satellite negative Yes No

β-lactamase positive Yes No

Antibiotic Resistant Yes No

Colistin Polymixin B Penicillin _____

Growth Temperatures 25°C 37°C 42°C



Attachment III LAB-5 Clinical Specimen Rule out for Select Agent and Chain of Custody



New Jersey Department of Health, Public Health and Environmental Laboratories
**REQUEST FOR TESTING OF CLINICAL SPECIMENS FOR SUSPECTED
 PATHOGENS OF PUBLIC HEALTH SIGNIFICANCE AND CHAIN OF CUSTODY**

Culture Description: _____

Colony Morphology (if applicable): Check all that apply

Growth medium used: <input type="checkbox"/> BAP <input type="checkbox"/> CHOC <input type="checkbox"/> MAC <input type="checkbox"/> EMB <input type="checkbox"/> Other: _____			
Time of growth when observation took place: _____ hours			
Form		Margin	
Elevation		Color	

- REJECTED: (PHEL Use Only)
 Improper package Unannounced No case number Improper documentation Other _____

CHAIN OF CUSTODY (Required for suspected Select Agents)

X _____ Relinquished by (Print)	Date: _____ Time: _____	X _____ Received by (Print)	
X _____ Relinquished by (Signature)		X _____ Received by (Signature)	
X _____ Relinquished by (Print)	Date: _____ Time: _____	X _____ Received by (Print)	
X _____ Relinquished by (Signature)		X _____ Received by (Signature)	
X _____ Relinquished by (Print)	Date: _____ Time: _____	X _____ Received by (Print)	
X _____ Relinquished by (Signature)		X _____ Received by (Signature)	
X _____ Relinquished by (Print)	Date: _____ Time: _____	X _____ Received by (Print)	
X _____ Relinquished by (Signature)		X _____ Received by (Signature)	
X _____ Relinquished by (Print)	Date: _____ Time: _____	X _____ Received by (Print)	
X _____ Relinquished by (Signature)		X _____ Received by (Signature)	