Ebola PUI: Laboratory Testing & Biosafety Perspective

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Objectives

- DCLS overview and Ebola Virus response
- Specimen collection – DCLS testing
- Specimen transport
- Specimen packaging & shipping
- Diagnostic laboratory testing
Division of Consolidated Laboratory Services (DCLS)

- Serves as the public health, environmental, agriculture, and consumer protection laboratory for the Commonwealth of Virginia.
- Serves hundreds of local, state, and federal agencies.
- Perform >7 million tests annually with over 650 different types of analyses.
- Test services include:
  - Microbiology, Mycology, Molecular Biology, Immunology/Virology, Newborn Screening
  - Radiochemistry, comprehensive chemical analyses, food, water, feed, fertilizer analyses
  - Metal/pesticide analyses, motor fuels and commodities analyses, laboratory certification
- Operate a statewide courier that provides routine and emergency transportation for specimens from over 200 locations to DCLS.
- Training and education (hospitals and partner agencies)
DCLS’ LRN-Emergency Response Program

- Only Laboratory Response Network (LRN) Reference laboratory for the Commonwealth of Virginia.

- Prepare, identify, and respond 24/7/365 to any public health emergency concerning agents of bioterrorism or highly pathogenic, high consequence, or emerging infectious pathogens.

- Comprised of highly skilled scientists trained and competent to work in BSL-3 containment.

- Test methods include:
  - Nucleic acid extractions and Real-time PCR assays
  - Conventional, microbial assays
  - Immunoassays
  - Mouse bioassay
<table>
<thead>
<tr>
<th>Agent</th>
<th>Disease</th>
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<tbody>
<tr>
<td><em>Bacillus anthracis</em></td>
<td>Anthrax</td>
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<tr>
<td><em>Brucella</em> <em>spp.</em></td>
<td>Brucellosis</td>
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<tr>
<td><em>Francisella</em> <em>tularensis</em></td>
<td>Tularemia</td>
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<td><em>Burkholderia mallei</em></td>
<td>Glanders</td>
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<td><em>Burkholderia pseudomallei</em></td>
<td>Meliodiosis</td>
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<td><em>Yersinia pestis</em></td>
<td>Plague</td>
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<tr>
<td><em>Coxiella burnetii</em></td>
<td>Q fever</td>
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<td>Poxviruses</td>
<td>Smallpox, Vaccinia, Chickenpox</td>
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<tr>
<td>Ricin toxin</td>
<td>Ricin poisoning</td>
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<tr>
<td><em>C. Botulinum</em> &amp; toxins</td>
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Middle East Respiratory Syndrome Coronavirus (MERS-CoV)

**LRN – Bio Testing at DCLS**

- B. Anthracis
- Brucella spp.
- F. tularensis
- B. mallei & pseudomallei
- Y. Pestis
- C. Burnetii
- Poxviruses
- Ricin toxin
- C. Botulinum & toxins

**Health Advisory:**

Middle East Respiratory Syndrome (MERS)

**Developing Story:**

First MERS case in U.S.

Healthcare worker in Indiana recently was in Saudi Arabia

2nd case of MERS confirmed in U.S., 500 people may have been exposed
# Ebola Virus Disease

## LRN – Bio Testing at DCLS

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## Years of Outbreaks

- **2013** - Novel Coronavirus/ MERS
- **2014** - Ebola Zaire Virus
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Our Goal: Protecting the Public’s Health

Real-time Disease Surveillance and Prevention

RAPID Disease Investigation “Epidemiology”

RAPID Laboratory Testing
DCLS - Ebola Virus Response
Risk Mitigation - Enhanced PPE

Standard DCLS BSL-3 PPE:
- PAPR
- PAPR head cover
- Fluid-impervious, back closing gown
- shoe covers
- EC gloves (double while working in BSC)

Enhanced DCLS Ebola testing PPE:
- Tyvek suit
- PAPR (inside Tyvek suit)
- Full PAPR hood (inner bib inside Tyvek suit)
- Tyvek boots (inside Tyvek suit)
- Fluid-impervious, back closing gown
- EC gloves (double while working in BSC)
- Physician suspects EVD in a PUI with correlated symptoms, travel, and/or exposure history.
- Physician contacts VDH to discuss suspect case.
- VDH consults with the CDC.
- VDH and CDC Ebola SME determine risk classification and decide if Ebola virus testing is warranted.
- Once approved, VDH contact DCLS to request Ebola virus testing.
- DCLS contacts hospital to provide specimen collection/packaging guidance and arrange a STAT courier pickup at the healthcare facility.
- Courier delivers specimen to DCLS for emergency testing.
DCLS Ebola Virus Testing

DCLS Ebola Response Team
- Specialized BSL-3 trained scientists with molecular biology experience.
- Perform specimen handling, inactivation, and extraction in a restricted BSL-3 containment lab.

First Tier molecular testing:
- Ebola virus real-time RT-PCR – detects Ebola Zaire virus
- Malaria real-time PCR – detects 4 *Plasmodium* species

Results within 5 hours of specimen receipt at the laboratory
- False negative EVD results possible when specimen is collected less than 72 hours from symptom onset.
  - If patient remains symptomatic without alternative diagnosis, submit a later specimen to DCLS to rule-out EVD.
- Any positive EVD result will require confirmation at the CDC.
DCLS Enhanced Testing Capabilities

Second Tier molecular testing:
- Influenza real-time RT-PCR – detects 3 common circulating viruses
- Norovirus real-time RT-PCR – detects GI and GII genogroups
- Rapid, molecular GI pathogen panel – 22 GI pathogens (bacterial, viral, parasitic)

Additional Testing Capabilities – to provide a comprehensive testing menu in the absence of an alternative diagnosis
- Arbovirus testing
- Facilitate shipment of specimens to the CDC for additional or confirmatory testing.
DCLS outreach to sentinel hospital laboratories to discuss biosafety, specimen collection and packaging and shipping

- Provided EVD testing and submission instructions and FAQ documents - shared via DCLS website, sentinel lab communications, and in DCLS Ebola kits
- DCLS Training Coordinators provided on-site, risk assessment training to Virginia hospitals
- Offered free Packaging and Shipping training courses for hospital laboratories
- Provided Ebola Category A/UN2814 specimen collection and shipping kits to: - 35 VDH Health Districts, 5 regional epidemiologists, 4 OCME offices, >100 hospitals, 6 Ebola Assessment Centers, and 2 Ebola Treatment Centers
- Served on the VDH Ebola Assessment Team and participated in 2016 Ebola Summit
- Provided 24/7 transport of Ebola virus PUI specimens to DCLS
Specimen Collection & Transport
Coordinate Ebola PUI testing in collaboration with VDH and DCLS

- Whole blood preserved with EDTA
- Plastic tubes only (NO glass)
- 2 lavender top blood tubes - minimum volume of 4mL each
- Do NOT centrifuge or manipulate specimens
- Specimens should be stored and shipped at 2-8°C
Specimen Transport In-House

Pre-plan the route of the specimen from site of care to the testing area

- Familiarize staff
- Avoid high traffic areas
- Consider providing an escort
- Wear CDC recommended PPE
- Hand carry - DO NOT use pneumatic tube systems
- Use a durable, leak-proof transport container (29 CFR §1910.1030)
- Decontaminate specimen and outside of transport container with EPA approved disinfectant for Ebola virus before removing from site of care

Staff training is critical for all aspects of handling patient specimens
Ebola Virus is classified by the Department of Transportation (DOT) as a Category A infectious substance

- Packaged and shipped in accordance with DOT Hazardous Materials Regulations and International Air Transport Association (IATA) Dangerous Goods Regulations

Individuals packaging and shipping infectious substances must be trained and certified

- Packages require UN-certified materials, labels, marking, and documentation
- DOT requires recertification every 3 years
- IATA requires recertification every 2 years

DCLS Courier – Specimen pickup

- Where to park at the facility?
- Consider convenience to the pickup location
- Where and whom to meet?

DCLS Resources

- Ebola Virus Disease Testing and Shipping Instructions (# 23539) - DCLS website Hot Topics
- Micro/Viro Submission Form (# 16857) – DCLS website Resources/Submission Forms
- Ebola FAQs - DCLS website Hot Topics
Clinical Diagnostic Testing
Laboratory testing should NOT be delayed. Timely diagnostic testing is essential to ensure patient care is not compromised.

- Use a certified Class II biosafety cabinet (BSC)
- Proper donning and doffing of CDC recommended PPE is critical for safety
- Use manufacturer-installed safety features that reduce likelihood of exposure
- Plan the path of the specimen throughout the testing area and the order of tests
- Staff training and strict adherence to protocols is essential

Assessment hospitals must be capable of safely and timely performing the following:

- CBC, differential, platelet count
- Sodium, potassium, bicarbonate, BUN, creatinine, glucose
- Liver function tests (AST, ALT, total bilirubin)
- Coagulation testing, especially prothrombin time (PT)
- Blood culture for bacterial pathogens (manual or automated)
- Urinalysis (screening test, not requiring centrifugation)
- Malaria smear or rapid test
- Rapid Influenza virus testing
Instruments or methods used in dedicated laboratories to conduct Ebola virus testing

### Clinical Chemistry

<table>
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<th>Manufacturer</th>
<th>Device</th>
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<tbody>
<tr>
<td>Beckman-Coulter</td>
<td>DxC880i</td>
</tr>
<tr>
<td>Abbott Laboratories</td>
<td>ISTAT</td>
</tr>
<tr>
<td>Abaxis</td>
<td>Piccolo Xpress</td>
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### Microbiology

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<tr>
<th>Test</th>
<th>Method</th>
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<tbody>
<tr>
<td>Blood Culture</td>
<td>Plastic bottle/manual monitoring method</td>
</tr>
<tr>
<td>Malaria</td>
<td>Smear fixed in methanol for 15 mins</td>
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<td>Alere BinaxNOW</td>
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### Hematology

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<tbody>
<tr>
<td>Sysmex</td>
<td>XN 9000</td>
</tr>
<tr>
<td>Sysmex</td>
<td>pocH 100i</td>
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### Coagulation

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<tbody>
<tr>
<td>ITC</td>
<td>Hemochron Signature Elite</td>
</tr>
<tr>
<td>F. Hoffman-La Roche</td>
<td>CoaguChek</td>
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These lists are generated from institutions other than DCLS, are for information only and are not intended as a DCLS or CDC endorsement of these instruments or practices, nor should this be considered a complete list of all test instruments that may be acceptable.
# Site-specific Risk Considerations

## Administrative controls
- Institutional policies and SOPs, safe work practices, spill response, medical surveillance
- Training, demonstrating proficiency, spill response
- PPE selection and use

## Engineering controls
- BSCs, safety equipment, sealed centrifuge rotors, sharps containers
- Test area layout, ventilation, anteroom
- Limit staff, restrict testing area

## Infectious Waste Management
- EPA requirements for inactivating Ebola Virus

## Instrumentation
- Certification
- Ease of operation/use while wearing PPE
- Aim for closed tube/chamber systems
- Minimize generation of aerosols
- Assess potential routes of exposure
- Consider proximity to other operations
- Can it be segregated after use
- Decontamination
- Will EPA recommended disinfectants affect/interfere with performance or potentially void manufacturer’s warranty
After Ebola Virus

What will be next???

Public health and health care organizations are better prepared to handle the next emerging infectious pathogen.
Questions?


Email us at:

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latoya.griffin-thomas@dgs.virginia.gov

CDC Ebola Virus Disease Guidance for Laboratory Personnel: