The Republic of Uganda
Ministry of Health

Laboratory diagnosis of an Ebola Patient

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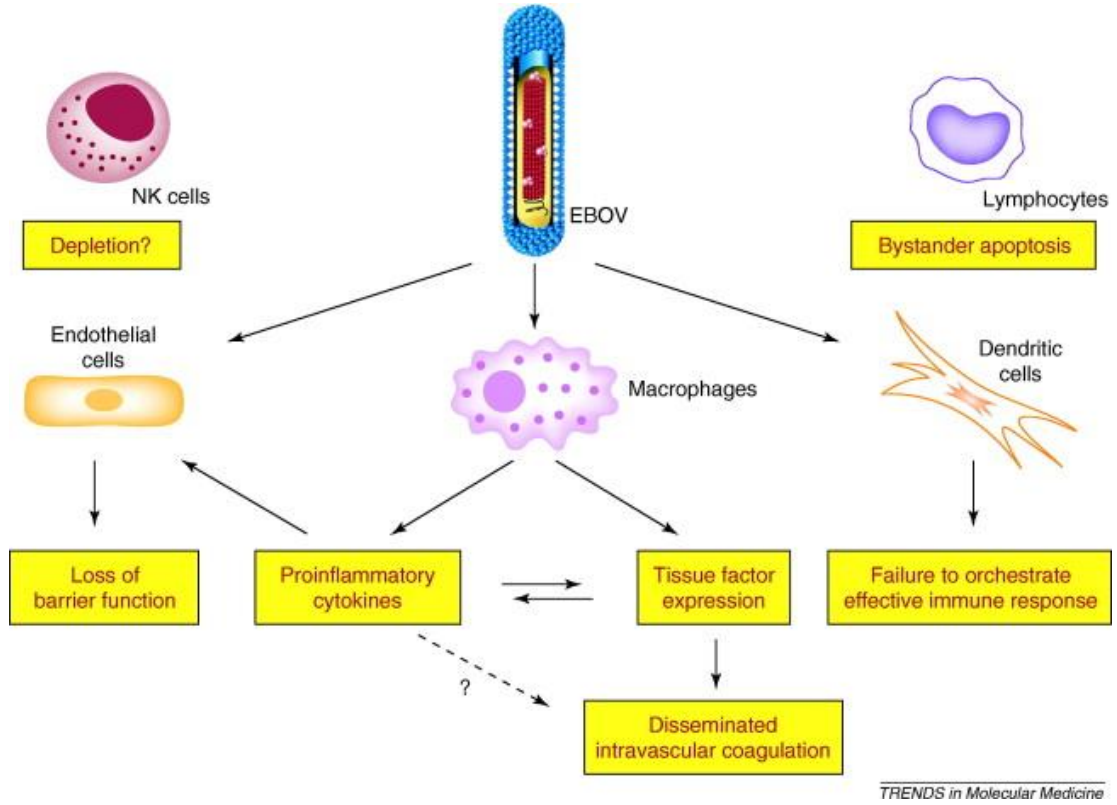
Presentation outline

- Pathogenesis of Ebola
- Testing Criteria
- Preparation Before Testing
- Sample Collection
- Sample Management
- Ebola-specific Tests
- Supportive Tests
- Summary

Pathogenesis of ebola

- Entry through mucosal surfaces or breaks in the skin
- Viral replication in macrophages and dendritic cells
- Spread to lymph nodes, liver, spleen, and other organs
- Immune system evasion and widespread inflammation
- Organ failure and hemorrhage



Testing criteria

- High fever, severe headache, muscle pain
- Unexplained hemorrhage
- Recent travel history to affected areas
- Contact with infected individuals or animals
- Clinical/laboratory personnel handling Ebola patients or samples

Preparation before testing (checklist)

- Necessary PPE
- BSL-4 containment/enhanced BSL3
- Protocols for handling and disposing of biohazardous materials
- Ebola-specific diagnostic kits and reagents
- Person meets the Ebolavirus testing criteria
- Personnel are trained in sample collection
- Do you have or need a partner
- Readily available disinfectant
- A plan for waste collection
- Safe donning/doffing procedures

Recommended specimens

- Whole blood in EDTA (a minimum volume of 4mL), collected in plastic tubes from live patients;
- Oral swabs stored in a universal transport medium, collected from deceased patients* or in situations where blood collection is not possible e.g. children.

Swab collection from live patients is not recommended due to lower sensitivity for reverse transcription polymerase chain reaction (RT PCR) and antigen detection.

Sample collection

- Sample type: blood, saliva, urine, and tissue samples
- Most PCR tests use whole blood collected in an EDTA tube (**purple top**).
- Always use plastic (not glass) collection tubes that do not contain heparin.
- Collection timing: early stages of symptom onset
- Use of appropriate containers and labeling
- Aseptic techniques

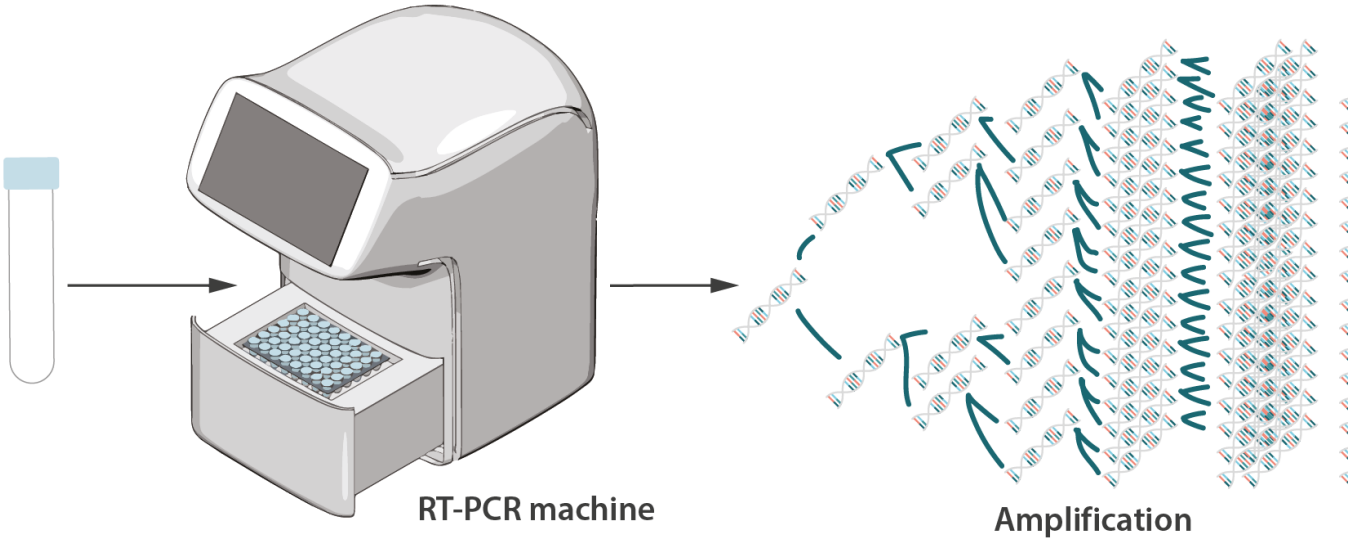
If the person under investigation's (PUI's) symptoms have been present for <3 days, a second specimen may be required and collected after 72 hours of symptom onset to definitively rule out Ebola.

Sample Management

- Immediate transport to the laboratory under cold chain conditions
- Strict chain of custody documentation
- Use of double-layer packaging for biohazardous materials
- Compliance with national and international transport regulations

Ebola-specific tests - RT-PCR

- Reverse transcription polymerase chain reaction (RT-PCR) as the **gold standard**
- Detection of viral RNA in blood samples
- High sensitivity and specificity
- Faster turnaround time and easy-to-interpret results



Specific Tests for Ebola - ELISA

- Enzyme-linked immunosorbent assay (ELISA) for antigen detection
- Detection of Ebola virus antigens in blood or tissue samples
- Use in initial screening and confirmation
- Limitations and cross-reactivity considerations

Specific Tests for Ebola - Virus Isolation

- Culturing the virus in BSL-4 laboratory conditions
- High sensitivity but requires specialized facilities
- Used for research and confirmation purposes
- Time-consuming and resource-intensive

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Specific Tests for Ebola - IgM and IgG Serology

- Detection of Ebola-specific antibodies (IgM and IgG)
- Indication of recent or past infection
- Role in epidemiological studies and vaccine trials
- Limitations in acute diagnosis

SUPPORTIVE TESTS FOR EBOLA

CBC and Coagulation Profile

- Complete blood count (CBC) to monitor white blood cell and platelet counts
- Coagulation profile to assess bleeding risk (PT, aPTT, INR)
- Liver function tests and renal function tests for organ involvement
- Importance in patient management and prognosis

Electrolytes and Blood Chemistry

- Monitoring electrolytes (sodium, potassium, chloride) for dehydration and electrolyte imbalance
- Blood chemistry (glucose, lactate, bicarbonate) to assess metabolic status
- Indicators of organ function and disease severity
- Role in supportive care and treatment decisions

Sample management post-esting

- Proper disposal of used materials and PPE
- Decontamination of work surfaces and equipment
- Documentation and storage of test results
- Compliance with regulatory guidelines for biohazardous waste

Summary

- Importance of accurate and timely diagnosis of Ebola
- Adherence to safety protocols and guidelines
- Integration of specific and supportive tests for comprehensive evaluation
- Continuous training and capacity building for laboratory personnel

Question: What is the gold standard test for detecting Ebola virus in blood samples?

a) Enzyme-linked immunosorbent assay (ELISA)

b) Complete blood count (CBC)

c) Reverse transcription polymerase chain reaction (RT-PCR)

d) IgM and IgG Serology

Correct Answer: **c) Reverse transcription polymerase chain reaction (RT-PCR)**

Question: Which type of collection tube should be used for PCR tests when collecting whole blood samples for Ebola testing?

a) Heparinized glass tubes

b) EDTA plastic tubes (purple top)

c) Serum separator tubes

d) Improvised stool containers or glass jars with lids

- Correct Answer: **b) EDTA plastic tubes (purple top)**