The Ebola Epidemic in West Africa
Institute of Medicine
- Forum on Microbial Threats -
Washington, DC, USA
March 24-25, 2015
Marburgvirus
Cuevavirus (no isolate)
Ebola virus
- Zaire (CFR 50-90%)
- Sudan (CFR ~50%)
- Taï Forest (CFR unknown)
- Bundibugyo (CFR <40%)
- Reston (CFR 0%)
Ebola HF seems to be restricted to the humid rain forest of Central Africa, whereas Marburg HF is found in the drier areas of Central Africa.
**EHF/MHF - clinical course**

**Survivors:** myalgia, arthralgia, asthenia, hepatitis, hearing loss, ocular diseases, psychosis

**Initial Symptoms:** fever, chills, headache, myalgia, arthralgia, asthenia, abdominal pain, diarrhea, sore throat, cough, pharyngeal/conjunctival injections, rash

**Fatal Cases:** bleeding signs, anuria, shock, tachypnea, obtundation, dyesthesia

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Antigen:

- 3
- 6
- 7
- 16

IgM antibody:

- 2
- 9
- 10
- 16

IgG antibody:

- 6
- 18
- 19
- 29
- 30
- 168

Days after onset of symptoms:

- 0
- 5
- 10
- 15
- 20
- 25
- 30
- 35
- 749

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Marburg hemorrhagic fever, 1967

Ebola hemorrhagic fever, 1995
Diagnostics

Detection of pathogen

- Nucleic acid detection
- Antigen detection
- Particle detection
- Virus isolation

Detection of pathogen-specific immune response

- IgM detection
- IgG detection
- Neutralization assays

Onset of symptoms

Recovery
WHO wants rapid, sensitive, safe and simple Ebola diagnostic tests

**Diagnostic tests**
- Ebola/Marburg abicap antigen detection
  (Senova, Corgenix, others)
- Ebola/Marburg PCR detection kits
  (Altona; Genekam, Vacunek, LIPSDIAG, Sacace, Genesia, others)
- currently diagnostics largely based on ‘in-house’ assays
- problems with accessibility of proper positive control material
- problems with test evaluation

**Test evaluation**
- no official evaluation system in place
- European Network for Imported Viral Diseases (ENIVD) performs quality assurance tests for Ebola and related viruses (http://enivd.net/)
CDC/NIH Lab ELWA, Monrovia
Molecular detection methods

qRT-PCR methods (thermal amplification)
- most widely used by mobile labs
- comes in multiple flavors (multiplex, probe, etc.)
- rapid (<1 hour)
- well evaluated
- requires sophisticated equipment

RT-LAMP methods (isothermal amplification)
- occasionally used by mobile labs
- rapid (<30 minutes)
- requires less sophisticated equipment
- not well evaluated
- more prone to fail due to use of multiple oligos

Isothermal recombinase polymerase amplification (RPA) method
- occasionally used by mobile labs
- rapid (<10 minutes)
- requires sophisticated equipment
- not well evaluated
Nucleic acid extraction

- multiple commercial sources offer kits
- most commonly used kits are based on chaotropic agents
- simple to use (can be automated)
- time-consuming (30–60 min; depending on sample volume)
- caution: potential for rest infectivity?

Buffers AVL & RLT only completely inactivate EBOV following ethanol addition
qRT-PCR diagnostics

- Aug 20th - present: ~ 5,000 samples processed
- JBAIDs (CDC Kenya):
- BioRAD (CDC Atlanta):
- Smart cycler / Lighycler (NIH):

EBOV-GP assay
EBOV-VP40 assay
EBOV-L screening assay
EBOV-NP confirmation assay

Takada & Kawaoka, Trends in Microbiology, 2001

>99% concordance among CDC & NIH assay!
Diagnostic algorithm needs to be adapted to local situation. Rapid test use will require altered diagnostic algorithm.

- initial test on admission (clinical symptoms)
  - isolation ward
  - retest within 24-72 hours
    - retest after 3 days without symptoms
      - discharge with 2 (3) consecutive negative test results

+ = two target qRT-PCR
Blood chemistry

“bedside”
**Limitation of rapid tests**

- **incoming patient**: a negative does not exclude an infection
- **survivor**: a negative does not exclude infectivity
- **disease progression**: viral load determination
- **treatment**: quantitative viral load determination

**Advantage**
- bedside application
- rapid result (<15 minutes)

**Problem:**
- sensitivity (compared to 'gold standard' qRT-PCR)

**Solution:**
- needs to be worked into a diagnostic algorithm

Format may also be used for antibody detection using viral antigens to capture human (host) antibodies
Serology

- ELISA format most commonly used (IgM-capture, IgG)
- rapid test based on the format of a pregnancy test
- ELISA, up to 2 hours; rapid test (<15 minutes)
- rapid tests not well evaluated
- caution: inactivation & rest infectivity?

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<td>Positive control</td>
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Buffer = 0.5%, NP40/1%, Tween20
Advances in diagnostics

Diagnostic: world reference centers
- most reliable and sophisticated (multiple approaches)
- shipping issues (international transport)
- time delay (weeks)
- reporting issues (weeks)
- confirmation not needed

Diagnostic: in-country reference centers
- reliable and sophisticated (multiple approaches)
- shipping issues (national transport)
- time delay (days)
- reporting issues (days)
- confirmation usually not needed

Diagnostic: on-site
- reliable (usually one approach)
- no transport issue
- time delay (<6 hours)
- reporting within hours
- confirmation usually not needed

Diagnostic: bedside
- less reliable (one approach)
- no transport
- no time delay (<15-30 min.)
- no reporting
- confirmation preferable and needed for low viral load
Acknowledgements

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The people of Liberia