Learning Objectives

After attending this presentation, participants will be able to:

- Compare the relative merits of different US Food and Drug Administration (FDA)–approved HIV tests for different circumstances
- Describe how new HIV tests fit in with current HIV testing recommendations
- Recognize the increasing role that HIV RNA tests play in HIV diagnosis

Outline

- The basics
- New tests: rapid, lab, & supplemental
- How differences between tests relate to accuracy
- On the horizon
HIV Infection and Laboratory Markers

- HIV RNA (plasma)
- HIV p24 Ag
- IgM
- IgG


HIV-1 antigens and RNA

- gp120
- gp 41
- p24
- RNA

CLIA-waived rapid HIV-antibody tests

- Oraquick Advance
- DPP HIV 1/2
- Chembio Sure Check
- INSTI HIV 1/2
- Chembio Stat Pak
- Uni-Gold Recombigen
Which antibodies do rapid tests detect?

1. p24 antibody
2. gp41 antibody
3. gp120 antibody
4. gp41 and gp120 antibody
5. All three: p24, gp41, and gp120
What's new?

- **Chembio SureCheck**
- **DPP HIV 1/2**

**Product name change:**
Clearview Complete is now Sure Check

**DPP HIV-1/2**
- Finger-stick, oral fluid
- Swab gums 4 times (15 seconds) or 10 µL whole blood
- Read time 10-25 min blood
  40 min oral fluid
Dual Path Platform Technology

- CLIA-waived for whole blood, finger-stick
- 50 µL specimen volume
- Results <1 minute
- Detects IgM antibodies


What’s new in the lab?
Abbott Architect Ag/Ab Combo 2010

“4th Generation”

Siemens Advia Centaur® CHIV 2015

Chemiluminescence Immunoassays

Patient Sample

Magnetic MicroParticles coated with antibody

Anti-p24 Monoclonal

Ag/Ab Combo

LITE Reagent

Antigens and antibodies labeled with AE

Trigger Solution

Relative light units

Anti-p24 Monoclonals

Anti-p24 Monoclonals

Random Access Multiplatform analyzers for HIV testing

On-board Refrigeration of Multiple Different Assays
Random Access Multiplatform analyzers for HIV testing

STAT sample requests without pausing
Results in <60 minutes

Abbott Architect Ag/Ab Combo
- Antibody gp41, gp120
- Antigen p24
- Control

Siemens Advia Centaur® CHIV
- CLIA-waived
- Whole blood (50µL)

Which factor is least important for the accuracy of a rapid HIV antibody test?

1. The type of specimen (serum, whole blood, oral fluid).
2. The volume of specimen the test requires.
3. The way the test is designed.
4. The time it takes to run the test.
5. The prevalence of HIV in the population tested.
Accuracy: Performance Characteristics

- **Sensitivity**
  - The ability of the test to identify correctly those who **have** the disease
    - HIV: ≥99.8%

- **Specificity**
  - The ability of the test to identify correctly those who **do not have** the disease
    - HIV: ≥99.8%

We Are Spoiled.

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**HIV RNA (plasma)**

- **HIV Ab**
- **HIV p24 Ag**

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**Relative Sensitivity during Seroconversion (plasma)**

- 26 seroconverters were analyzed with 14 tests
  - 17 seroconverters with Positive WB used for cumulative frequency analysis

- 50% Cumulative Frequency
Sequence of Test Positivity Relative to WB (plasma)

166 specimens, 17 Seroconverters - 50% Positive Cumulative Frequency

Bangkok Tenofovir Study:
Delayed HIV detection by oral fluid in patients on PrEP

Participants receiving tenofovir (who became HIV-infected) took longer to develop a reactive OraQuick (191.8 days) than participants receiving placebo (16.8 days)


Determine Combo Ag/Ab with Whole Blood

- In 6 studies involving >26,000 persons, Determine Combo failed to detect p24 antigen in whole blood from any of the 26 acute infections.

- CDC study: Eight seroconverters showed a median delay of 6 days between DC reactivity with plasma and reactivity with whole blood

Quick Case Study

- 25 year old MSM in an HIV-discordant relationship, on PrEP since 12/2015. Perfect adherence by history. Also had 2 other sex partners.

- 4th generation testing repeatedly negative until May 2016:
  - Positive 4th gen, negative Multispot, HIV RNA<20, signal detected

- Repeat 2 and 4 weeks later: same results.

2003 study: false-negative Oraquick and waning or absent gp41 titers in patients on early, effective therapy
Bangkok tenofovir study...

Genotype: Multiple drug resistance mutations (including TDF/FTC), virus not related to that of his virally suppressed partner.

- O’Connell et al, J Clin Micro 2003
**Geenius™ HIV-1/2 Lines**

**HIV-1 & HIV-2 Associated Lines**

**Control Band**

HIV-2  | HIV-1

*Need to read carefully*

**Dual Path Platform:**

Add 5 µL serum/plasma

Or 15 µL whole blood to specimen well

Add 5 drops buffer to buffer well
Insert test cassette in reader for automated interpretation.

Geenius Results: New Interpretations

<table>
<thead>
<tr>
<th>HIV-1 RESULT</th>
<th>HIV-2 RESULT</th>
<th>ASSAY INTERPRETATION</th>
</tr>
</thead>
<tbody>
<tr>
<td>Negative</td>
<td>Negative</td>
<td>HIV-1 NEGATIVE</td>
</tr>
<tr>
<td>Indeterminate</td>
<td>Negative</td>
<td>HIV-1 INDETERMINATE</td>
</tr>
<tr>
<td>Negative</td>
<td>Indeterminate</td>
<td>HIV-2 INDETERMINATE</td>
</tr>
<tr>
<td>Indeterminate</td>
<td>Indeterminate</td>
<td>HIV-1 INDETERMINATE</td>
</tr>
<tr>
<td>Negative</td>
<td>Positive</td>
<td>HIV-1 POSITIVE</td>
</tr>
<tr>
<td>Positive</td>
<td>Positive</td>
<td>HIV-2 POSITIVE</td>
</tr>
<tr>
<td>Negative</td>
<td>Positive</td>
<td>HIV-2 POSITIVE</td>
</tr>
</tbody>
</table>

1. HIV-1 Negative
2. HIV-1 Indeterminate
3. HIV-2 Indeterminate
4. HIV-1 Positive
5. HIV-2 Positive
6. HIV-1 Positive and HIV-2 Positive
7. HIV-1 Positive and HIV-2 Indeterminate
8. HIV-1 Indeterminate and HIV-2 Positive

Notes:
- HIV-1 and HIV-2 were detected by EIA and confirmed by Western blot.
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- Results are based on the manufacturer's guidelines.
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## On the Horizon...

### 4th generation HIV-1/2 immunoassay

<table>
<thead>
<tr>
<th>HIV-1 (+)</th>
<th>HIV-1 (-)</th>
<th>HIV-2 (+)</th>
<th>HIV-2 (-)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV-1 antibodies detected</td>
<td>HIV-2 antibodies detected</td>
<td>HIV antibodies detected</td>
<td>HIV-1 RNA viral load</td>
</tr>
</tbody>
</table>

**June 27, 2014**

**HIV-1/HIV-2 antibody differentiation immunoassay**

- Positive for HIV-1 and HIV-2 antibodies and p24 Ag
- Negative for HIV-1 antibodies and HIV-2 antibodies

### How about...

**4th generation HIV-1/2 immunoassay**

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<th>HIV-2 (+)</th>
<th>HIV-2 (-)</th>
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</thead>
<tbody>
<tr>
<td>HIV-1 RNA viral load</td>
<td>HIV-1/HIV-2 antibody differentiation assay</td>
<td>Useful clinical information</td>
<td>Negative</td>
</tr>
</tbody>
</table>

**Useful clinical information**

- HIV-1 infection
- HIV-2 infection
- (Viral suppression)
- HIV-2 infection

**HIV-1 RNA viral load**

- Acute HIV-1 infection
- Negative for HIV-1
"Point-of-Care" Nucleic Acid Tests

- Xpert HIV-1 viral load
  - 1 ml plasma
  - Results in 90 minutes
  - LOD 32 copies/mL
  - CE-marked December 2014

GeneXpert

Not available in U.S.

"Point-of-Care" Nucleic Acid Tests

- Xpert HCV viral load
  - 1 mL serum or plasma
  - Genotypes 1-6
  - Range 10 – 100,000,000 IU/mL
  - Results in 105 minutes
  - CE-marked April 2015

GeneXpert

Not available in U.S.

"Point-of-Care" Nucleic Acid Tests

- 25 μL whole blood specimen
- HIV-1 or HIV-2 viral load in 60 minutes
- CE-marked March 2015

Alere q HIV-1/2 Detect

Not available in U.S.
“Point-of-Care” Nucleic Acid Tests

- “Lab in a Tube”
- Influenza A/B – FDA cleared
- Strep A – FDA cleared
  - Results in 15 minutes
  - CLIA-waived May 2015
- HIV under development

Summary

- HIV screening tests keep getting better
- HIV RNA viral load will play an increasingly important role in diagnosis
- You never know what’s next
Suggested Readings


