Dallas County Health and Human Services Laboratory

Testing For Acute HIV-1 Infection

Brian Emerson
HIV-1 RNA Testing Supervisor
Dallas County Health and Human Services Laboratory
Ph: (214)-819-1950
Email: brian.emerson@dallascounty.org
Discussion Topics

- Introduction to HIV-1 RNA NAAT
- Dallas County HIV Algorithm
- Pooling Methods
- Dallas County Laboratory Experience
Introduction to HIV RNA Testing

- Dallas County Laboratory utilizes the Aptima HIV-1 RNA Qualitative Assay (manufactured by Gen Probe for Hologic® Inc.) for detection of acute HIV infection (AHI)

- Aptima HIV-1 RNA Qualitative Assay is approved by the FDA as a diagnostic test and can be used in place of Western Blot
Aptima HIV-1 RNA Assay is an *in vitro* nucleic acid-based amplification test (NAAT) used for the detection of HIV-1 RNA in human plasma and serum.

The presence of HIV-1 RNA in the plasma or serum of a patient without antibodies to HIV-1 is an indication of acute HIV-1 infection.
Figure. Time to detection of HIV RNA, p24 antigen, and antibody during early HIV infection.
Dallas County HIV Algorithm

Blood sample received for HIV test

ELISA or EIA is performed

- EIA + (Test repeated X2)
  - Western Blot
    - Western Blot Reactive
      - HIV Reactive
    - Western Blot Indeterminate
      - NAAT Testing
        - NAAT Reactive
          - AHI Protocol
        - NAAT Nonreactive
          - HIV – (Retest based on risk factors)
      - Western Blot Nonreactive
        - NAAT Testing
          - NAAT Reactive
            - AHI Protocol
          - NAAT Nonreactive
            - Repeat HIV testing in 1 month

- EIA Nonreactive
  - Pooled NAAT Testing
    - NAAT Reactive
      - AHI Protocol
    - NAAT Nonreactive
      - HIV – (Retest based on risk factors)

Bio-Rad Multispot HIV-1/HIV-2 Rapid Test
Dallas County HIV Algorithm

Blood sample received for HIV test

ELISA or EIA is performed

EIA Nonreactive

Pooled NAAT Testing

NAAT Reactive

AHI Protocol

NAAT Nonreactive

HIV Nonreactive (Retest based on risk factors)
Dallas County HIV Algorithm

- Blood sample received for HIV test
  - ELISA or EIA is performed
    - EIA + (Test repeated X2)
      - Western Blot
        - Western Blot Reactive
          - HIV Reactive
        - Western Blot Indeterminate
          - NAAT Testing
            - NAAT Reactive
              - AHI Protocol
            - NAAT Nonreactive
              - Repeat HIV testing in 1 month
              - Bio-Rad Multispot HIV-1/HIV-2 Rapid Test
Dallas County HIV Algorithm

Blood sample received for HIV test

ELISA or EIA is performed

EIA + (Test repeated X2)

Western Blot

Western Blot Nonreactive

NAAT Testing

NAAT Reactive

AHI Protocol

NAAT Nonreactive

Repeat HIV testing in 1 month

Bio-Rad Multispot HIV-1/HIV-2 Rapid Test
Pooling Methods

- The ability to combine specimens into pools greatly reduces the cost for detection of acute HIV-1 infection.
- Pooling specimens will reduce the cost by 90%. (using 30,000 specimen per year for analysis)
- Pooling is not part of the package insert. The pooling method was verified and validated within the Dallas County Health Laboratory.
- The pooling procedure is performed manually.
Pooling Methods

- The specimens requesting HIV testing are first tested using either the Bio-Rad HIV-1/2 Plus O EIA assay or Clearview Rapid HIV method (used in the STAT lab and Mobile Vehicle).

- The specimens are separated into High Risk or Low Risk category.

- High or Low Risk is determined by the patient risk factors and submitter.
  (Ex. STD Clinic = High Risk, Family Planning Clinic = Low Risk)

- Bio-Rad HIV Combo Assay was implemented in August 2012.
Pooling Methods

- Currently utilizing pools of 10 for all high risk EIA and Rapid Nonreactive (NR) specimens
  - Pooling is performed same day as the EIA
  - Nonreactive results are available 1 day after draw date
  - Reactive results are available 3 days after draw date

- Testing only High Risk specimens due to cost

- Western Blot Nonreactive and Indeterminate and AHI suspects are tested individually
Pooling Methods

- 10 individual specimens are assigned to a specific pool number
- 100 μl of serum is pipetted into labeled pool tube
Pooling Methods

- If pool result is reactive then the pool will be broken back down to 10 individual specimens and tested individually.
- Once the reactive specimen is determined, one more individual test will be performed before the specimen will be reported as an acute infection.
- The result of the specimen being reported as an acute infection will be given to the Medical Director (STD Clinic), DIS Manager, and HIV Surveillance.
Dallas County Laboratory Experience

Karen

Reginald
Dallas County Laboratory Experience

Daniel Serinaldi
Shayma Haq
Dallas County Laboratory Experience

- Dallas County Health Laboratory experienced a 44% increase in patients requesting a HIV test from 2007 to 2009
- The HIV-1 RNA program went live in July 2009
- A total of 118 HIV-1 RNA reactive specimens were detected out of 113,843 specimens analyzed (2009-2011)
# Dallas County Laboratory Experience

<table>
<thead>
<tr>
<th>Year</th>
<th>Total Specimens</th>
<th>HIV-1 RNA Detected</th>
</tr>
</thead>
<tbody>
<tr>
<td>2009</td>
<td>18,957</td>
<td>12</td>
</tr>
<tr>
<td>2010</td>
<td>36,760</td>
<td>44</td>
</tr>
<tr>
<td>2011</td>
<td>58,126</td>
<td>62</td>
</tr>
<tr>
<td>2012</td>
<td>30,924</td>
<td>43</td>
</tr>
</tbody>
</table>
Dallas County is experiencing more HIV-1 RNA reactive results when the gp160 band is present.
Dallas County Laboratory Experience

- EIA 4th Generation
  - A total of 64 reactive HIV-1 RNA specimens were analyzed
  - 24 were reactive 3rd generation EIA, reactive 4th generation EIA
  - 25 were nonreactive 3rd generation EIA, reactive 4th generation EIA
  - 15 were nonreactive on the 3rd and 4th generation EIA
Dallas County Laboratory Experience

- Bio Rad HIV Ag/Ab Combo Assay was implemented August 2012

- Currently performing the Bio Rad HIV Ag/Ab Combo assay and pooling the nonreactive high risk specimens for the HIV-1 RNA assay
Dallas County Laboratory Experience

- **October 2011: (Bio Rad 3rd Gen EIA)**
  - 57 EIA reactive
  - 48 confirmed with Western Blot
  - 3 confirmed with HIV-1 RNA

- **October 2012: (Bio Rad 4th Gen EIA)**
  - 48 EIA reactive
  - 45 confirmed with Bio Rad Western Blot
  - 3 confirmed by HIV-1 RNA
  - 1 of the HIV-1 RNA reactive was EIA reactive, Western Blot nonreactive
  - 2 of the HIV-1 RNA reactive was EIA nonreactive
Dallas County Public Health Follow-up

- The Public Health Follow-up discovered 12 new HIV-1 infections from the HIV-1 RNA reactive specimens.

- 22% of the HIV-1 RNA reactive specimens were found to have a dual diagnosis of syphilis and HIV.

- 12% were found to have a Chlamydia and/or gonorrhea infection.
Dallas County Public Health Follow-up

Age Grouping for the HIV-1 RNA reactive (2010-2011)

<table>
<thead>
<tr>
<th>Age Group (years)</th>
<th>HIV-1 RNA reactive</th>
</tr>
</thead>
<tbody>
<tr>
<td>15-24</td>
<td>40%</td>
</tr>
<tr>
<td>25-34</td>
<td>34%</td>
</tr>
<tr>
<td>35-44</td>
<td>14%</td>
</tr>
<tr>
<td>45+</td>
<td>12%</td>
</tr>
</tbody>
</table>
Dallas County Public Health Follow-up

Ethnicity Breakdown for the HIV-1 RNA reactive (2010-2011)

<table>
<thead>
<tr>
<th>Ethnicity</th>
<th>HIV-1 RNA reactive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Caucasian</td>
<td>54%</td>
</tr>
<tr>
<td>African-American</td>
<td>43%</td>
</tr>
<tr>
<td>Other</td>
<td>3%</td>
</tr>
</tbody>
</table>
Dallas County Public Health Follow-up

Risk Behavior Breakdown for the HIV-1 RNA reactive (2010-2011)

<table>
<thead>
<tr>
<th>Risk Category</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male-to-Male Sexual Contact</td>
<td>74%</td>
</tr>
<tr>
<td>High Risk Heterosexual</td>
<td>19%</td>
</tr>
<tr>
<td>Unknown</td>
<td>6%</td>
</tr>
<tr>
<td>Injection Drug Use</td>
<td>1%</td>
</tr>
</tbody>
</table>
Dallas County Laboratory

Thank you for your time
References

1. Gen-Probe Aptima HIV-1 & HCV Assay Training Manual; GenProbe Incorporated, San Diego, CA.

Acknowledgements

I would like to thank Dr. Nick Curry and Deborah Carr of Texas Department of State Health Services, Dr. Edward Bannister and Gloria Hardin of Dallas County Health and Human Services for providing me this exciting and rewarding opportunity.
Dallas County Health and Human Services Contact Information

Edward R. Bannister, Ph.D.
Laboratory Services Director
2377 N. Stemmons Freeway-Basement
Dallas, Texas 75207-2710
Tel: (214) 819-1952
Email: ebannis@dallascounty.org

Brian P. Emerson
HIV-1 RNA Testing Supervisor
2377 N. Stemmons Freeway-Basement
Dallas, Texas 75207-2710
Tel: (214) 819-1950
Email: brian.emerson@dallascounty.org