Comparison of the Geenius™ HIV-1/2 Supplemental Assay and HIV-1 Western blot for HIV rapid test confirmation from dried blood spots

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** The authors have no conflicts of interest to disclose **
HIV Referral Testing at Wadsworth

• NYSDOH AIDS Institute (AI) and Bloodborne Viruses Lab (BVL) partner to support HIV rapid testing in NYS
• AI: guidance, training and TA to non-clinical providers
• BVL: confirmatory testing services
Why HIV Rapid Testing?

• Community-based testing sites expand access to HIV testing
• Wide variety of venues: LHDs, health centers, mobile vans
• Some sites lack ability to perform venipuncture to collect blood
Two Rapid Test Confirmation Algorithms

Blood referral (plasma/serum)
- HIV-1/2 antigen/antibody combination immunoassay
  - Supplemental Ab on reactives
  - HIV-1 NAT on negatives/indeterminates

Dried blood spot referral (DBS)
- Supplemental Ab testing
  - HIV-1 NAT on negatives/indeterminates
• After transition to MS for serum/plasma, WB used for DBS only
• Not feasible to adapt MS for use with DBS
• Geenius testing for serum/plasma began 2016
• Geenius easier to modify for use with DBS
DBS Testing Methods (Western blot)

- ¼ in punch is eluted for 2 hrs at room temp or overnight at 2-8°C
- Eluates tested using the GS HIV-1 Western blot kit (Bio-Rad)
Western blot has many disadvantages

• Batching is needed for cost efficiency
  – High and low pos, neg control with each batch
• Interpretation is subjective, requires experience
• Manual result entry into LIMS
• Designed for HIV-1
  – May misclassify HIV-2 as HIV-1 due to antibody cross-reactivity
Geenius HIV 1/2 Supplemental Assay

FDA approved in 2014

• Confirms and Differentiates HIV-1 and HIV-2 Ab
• Single-use device
• Results within 30 min
• Reader and software for reading results
• Import results electronically into CLIMS
Transitioning to Geenius for DBS

• In 2016, began using Geenius for supplemental Ab testing of all serum/plasma specimens
• Western blot was limited to DBS referrals
  – Fewer WB specimens meant longer TAT
  – Added expense of maintaining WB
• Minimal modification of the Geenius procedure needed
• Validation data showed excellent correlation
  – Reviewed and approved by NYSDOH CLEP
Geenius DBS Method

- ¼” inch punch is eluted overnight at 2 - 8°C in 150µl PBS pH 7.2 + 0.05% Tween 20 + 0.005% NaN₃
- 40µl of eluate and one drop of buffer added to well 1.
- Wait 20 minutes after adding buffer to well 2.
Western blot vs Geenius for DBS

How did switching from WB to Geenius affect our DBS referral testing?

<table>
<thead>
<tr>
<th></th>
<th>WB</th>
<th>Geenius</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total tested</td>
<td>245</td>
<td>191</td>
</tr>
<tr>
<td># HIV-1 Positive</td>
<td>217 (89%)</td>
<td>165 (86%)</td>
</tr>
<tr>
<td># Indeterminate/Non-reactive</td>
<td>28 (11%)</td>
<td>26 (14%)</td>
</tr>
<tr>
<td>HIV-1 Positive TAT (Average)</td>
<td>4.9 days</td>
<td>2.6 days</td>
</tr>
<tr>
<td>Indeterminate/Non-reactive TAT (Average)</td>
<td>9.0 days</td>
<td>5.5 days</td>
</tr>
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Further improvements to Geenius DBS

• IQCP completed; Control now run monthly
• Validated 2-hr elution; TAT reduced to a few hours

<table>
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Dried Blood Spot Turn-Around Time to Final Result

% of specimens

Days to Final Result

- WB HIV-1 P
- G HIV-1 P, ON Elution
- G HIV-1 P, 2hr Elution
Dried Blood Spot Turn-Around Time to Final Result

% of specimens

Days to Final Result

WB Neg or Ind, TMA
G Neg or Ind, TMA
Geenius vs WB: Accuracy

• BVL conducts HIV-2 RNA testing (Qual, Quant)
• In 2018, received plasma for HIV-2 RNA testing
• In 2013, a DBS specimen from the same patient was submitted for rapid test confirmation
• Result was HIV-1 positive by Western blot in 2013
  – p24+, gp41+, gp120+/-, gp160+
• Re-tested the 2013 DBS by Geenius
  – Correctly identified it as HIV-2 positive
Conclusions

- Geenius significantly reduced TAT, including potential acute infections that require HIV-1 NAT
- Geenius eliminates subjectivity in interpretation
  - Automated reader and dedicated software
- Detects and differentiates Abs to both HIV-1 and HIV-2
  - HIV-2 can be reliably detected
- Easier and faster to perform than Western blot
- IQCP and 2 hr elution reduce TAT even more
Acknowledgments

Wadsworth BVL
- Monica Parker
- Linda Styer
- Bucky Carmichael
- Patty Dingman
- Phil Rivenburg
- Bob Blum
- Tim Rem
- Larry Roth
- Renee Hallack
- Tom Miller
- Lea Ryman
- Jean Rock

AIDS Institute
- Mara San Antonio-Gaddy
- April Richardson-Moore
- Thomas Sullivan

Wadsworth MLS Students
- Elizabeth Luke
- Samantha Frye