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of Health**

**Wadsworth  
Center**

# **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



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# 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)

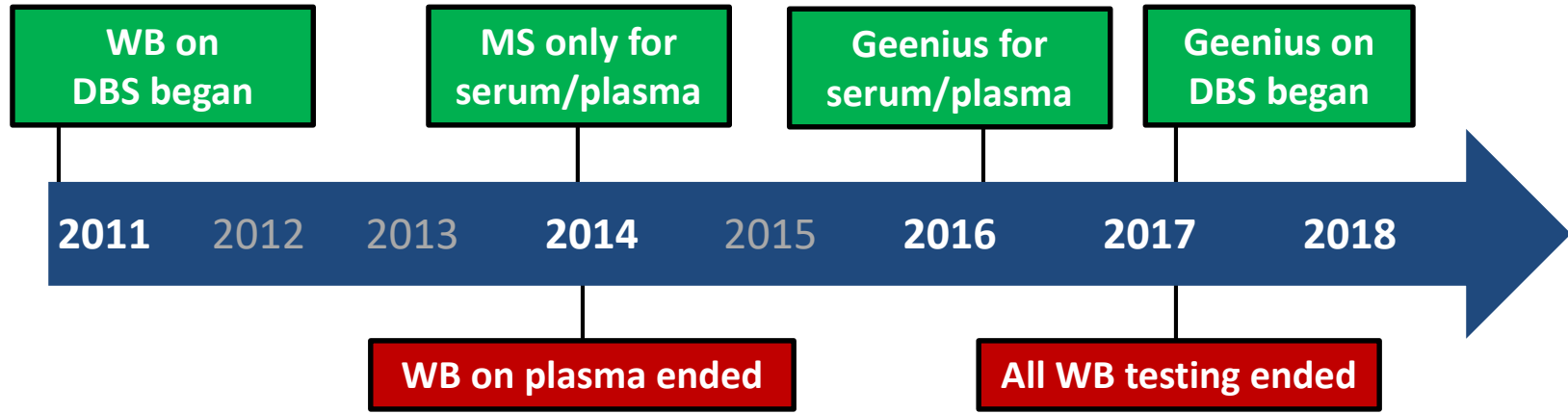
Supplemental Ab testing



HIV-1 NAT on  
negatives/indeterminates



# Supplemental Ab Testing Timeline

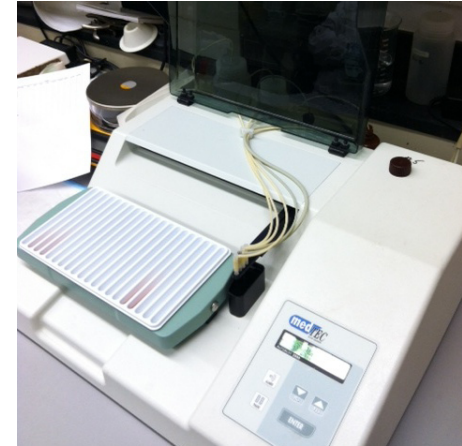
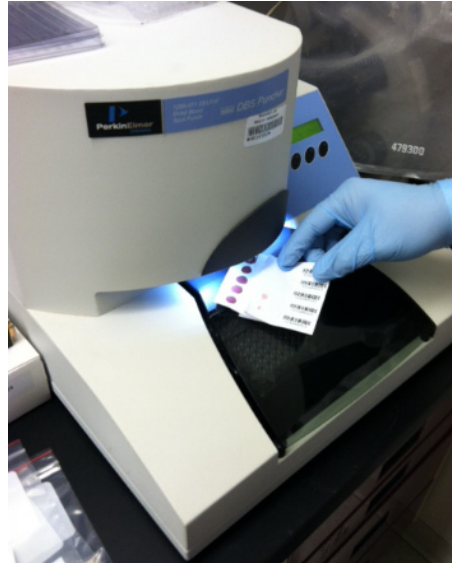


- After transition to MS for serum/plasma, WB used for DBS only
- Not feasible to adapt MS for use with DBS
- Geenius testing for serum/plasma began 2016
- Geenius easier to modify for use with DBS



# DBS Testing Methods (Western blot)

- $\frac{1}{4}$  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

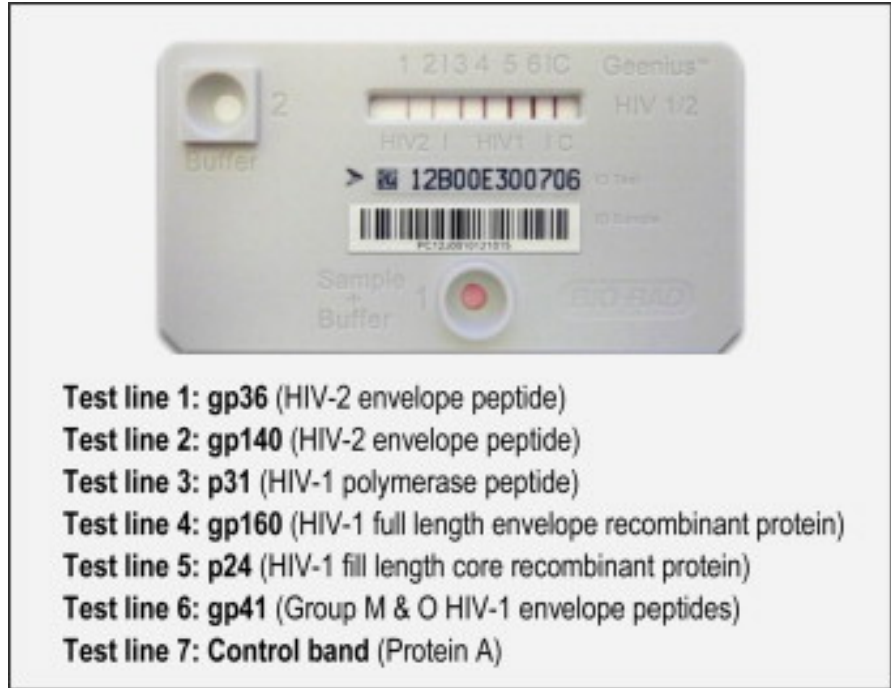
- Batching is needed for cost efficiency
  - High and low pos, neg control with each batch
- Interpretation is subjective, requires experience
- Manual result entry into LIMS
- Designed for HIV-1
  - May misclassify HIV-2 as HIV-1 due to antibody cross-reactivity



# 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



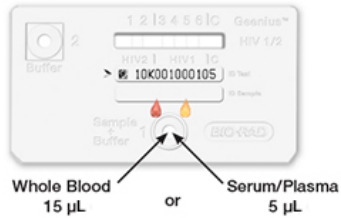


# Transitioning to Geenius for DBS

- 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



Dispense 15  $\mu\text{L}$  of whole blood or 5  $\mu\text{L}$  of serum/plasma into Well 1



**40 $\mu\text{L}$  DBS eluate**

Add 2 Drops of buffer into Well 1 (immediately but no longer than 5 minutes)

**1 drop for DBS**



Wait 5-7 minutes



Add 5 Drops of buffer into Well 2



Wait 15 to 20 minutes

**Wait 20 min**



Read, interpret and report results



Geenius<sup>®</sup> Reader

Notebook

(validated for Geenius<sup>®</sup> Software)

## Geenius DBS Method

- $\frac{1}{4}$ " inch punch is eluted overnight at 2 - 8 $^{\circ}\text{C}$  in 150 $\mu\text{L}$  PBS pH 7.2 + 0.05% Tween 20 + 0.005%  $\text{NaN}_3$
- 40 $\mu\text{L}$  of eluate and one drop of buffer added to well 1.
- Wait 20 minutes after adding buffer to well 2.



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# Western blot vs Geenius for DBS

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

	WB	Geenius
Total tested	245	191
# HIV-1 Positive	217 (89%)	165 (86%)
# Indeterminate/Non-reactive	28 (11%)	26 (14%)
HIV-1 Positive TAT (Average)	4.9 days	2.6 days
Indeterminate/Non-reactive TAT (Average)	9.0 days	5.5 days



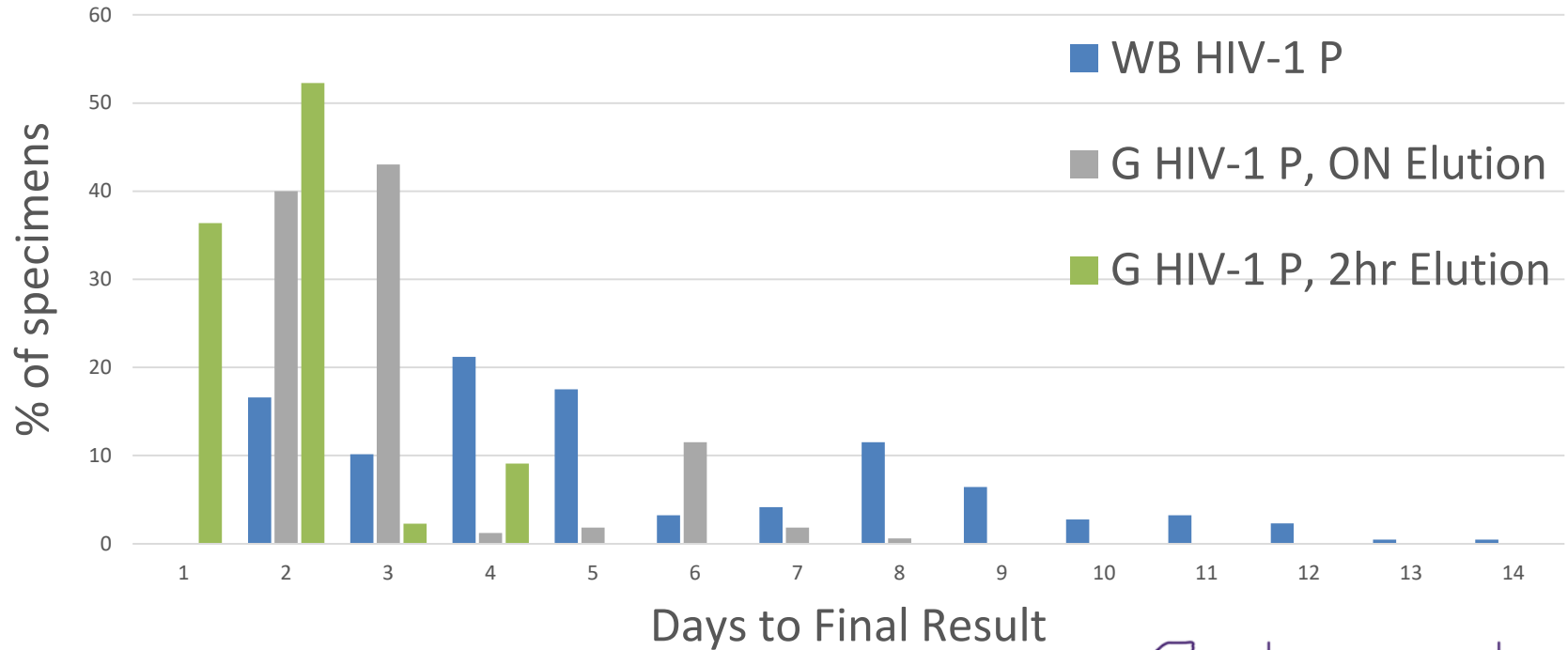
# Further improvements to Geenius DBS

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

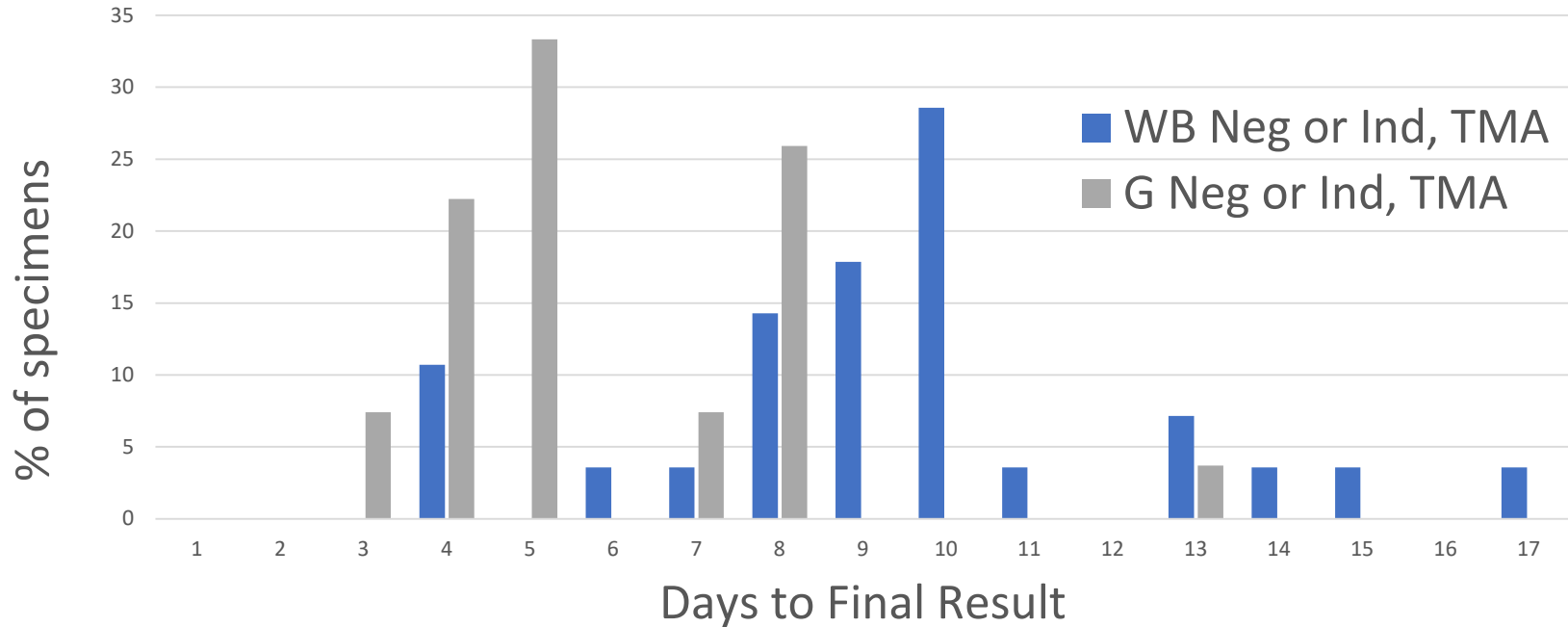
	Western Blot	Geenius (O.N.)	Geenius (2 hr.)
Total tested	245	191	45
# HIV-1 Positive	217 (89%)	165 (86%)	44 (98%)
# Indeterminate/Negative	28 (11%)	26 (14%)	1 (2%)
HIV-1 Positive TAT (Average)	4.9 days	2.6 days	1.3 days
Indeterminate/Neg TAT (Average)	9.0 days	5.5 days	2.6 days



## Dried Blood Spot Turn-Around Time to Final Result



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# 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





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