Point of Care Testing Updates

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National Association of County & City Health Officials

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Objectives

1. To discuss POC tests that are current
2. To update new tests in the pipeline
3. To discuss the accuracy of older and new POC tests
4. To mention impact of POC testing—advantages and barriers
Laboratory and Point-of-Care Tests for STIs

*Chlamydia trachomatis (CT)*

*Neisseria gonorrhoeae (NG)*

*Trichomonas vaginalis (TV)*

Syphilis

Herpes Simples Virus (HSV)

HIV

Gaydos, C. Rapid Tests for STDs Current Infect Dis Reports 2006;8:115-124

Huppert et al. Point of Care tests for STIs: What’s the Point? Point of Care Journal, 2009

Laboratory Test vs. POC Test

• Laboratory performed test may take 2-4 days turn around time or longer

• Lab test FDA rated highly complex or moderately complex; require a lab

• POC test (CLIA Waived) take 10-20 min-1 hour; minimal equipment

• POC usually rated simple enough to be performed by a trained health care worker (minimal training)
What Are Current POCs?

- **CT**: Clearview (Inverness);
  Cx 49.7% sens; vag 32.8% sens

- **NG**: None FDA cleared but Gram Stain

- **Trichomonas**: Wet preparation - OSOM

- **Syphilis**: RPR, VDRL; sensitive, not specific

- **HSV**: Tzanck Smear (80% sensitive, non-differentiating between simplex, zoster, and varicella)

- **HIV**: Orasure oral fluid antibody test; Many others

New POC tests for STIs

- Chlamydia
- Gonorrhea
- Trichomonas
- Syphilis
- HSV
- HIV
Needs Assessment of Clinicians

Which organisms do Clinicians want a POC test?

How sensitive; how specific?

How fast does it have to be?

What about cost?

What about equipment?

What about Patients Needs?

- Willingness to wait is important
- Willingness to self-collect specimens is important
- Willingness to pay is important

Rompalo et al. Sexual Health 2013;10:541-545
Patient Focus Group and Clinic Questionnaire about POC Tests (N =371)

<table>
<thead>
<tr>
<th>Specimen Type Preference</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cervical</td>
<td>15.4%</td>
</tr>
<tr>
<td>Vaginal</td>
<td>50.9%</td>
</tr>
<tr>
<td>Urine</td>
<td>33.7%</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Willingness to Wait</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>20 min</td>
<td>59.0%</td>
</tr>
<tr>
<td>40 min</td>
<td>20.8%</td>
</tr>
<tr>
<td>60 min</td>
<td>10.8%</td>
</tr>
<tr>
<td>90 min</td>
<td>9.4%</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Willingness to Pay</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>$10</td>
<td>46.6%</td>
</tr>
<tr>
<td>$20</td>
<td>31.0%</td>
</tr>
<tr>
<td>$30</td>
<td>10.8%</td>
</tr>
<tr>
<td>$40</td>
<td>2.7%</td>
</tr>
<tr>
<td>$50</td>
<td>8.9%</td>
</tr>
</tbody>
</table>

Self–collected vaginal swabs

Barnes et al. 2014 CDC STD Conf, Atlanta GA
New: “Near Patient” Test for Chlamydia and Gonorrhea

GeneXpert® CT/NG, Cepheid (90 minutes)

Urine or female Swab samples in Transport Reagent → Transfer the sample to the cartridge → Insert cartridge and start assay

Total hands-on time: <1 Minute

## Results CT/NG

1,722 female & 1,387 males

### Xpert CT/NG vs. Patient Infected Status

<table>
<thead>
<tr>
<th>Specimen</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td>CT Cervical</td>
<td>97.4%</td>
<td>99.6%</td>
</tr>
<tr>
<td>CT Vaginal</td>
<td>98.7%</td>
<td>99.4%</td>
</tr>
<tr>
<td>CT Female Urine</td>
<td>97.6%</td>
<td>99.8%</td>
</tr>
<tr>
<td>NG Cervical</td>
<td>100%</td>
<td>100%</td>
</tr>
<tr>
<td>NG Vaginal</td>
<td>100%</td>
<td>99.9%</td>
</tr>
<tr>
<td>NG Female Urine</td>
<td>95.6%</td>
<td>99.9%</td>
</tr>
<tr>
<td>CT Male Urine</td>
<td>97.5%</td>
<td>99.9%</td>
</tr>
<tr>
<td>NG Male Urine</td>
<td>98.9%</td>
<td>99.9%</td>
</tr>
</tbody>
</table>

### Sensitivity and Specificity of POC/near patient tests for CT & NG

<table>
<thead>
<tr>
<th>Organism</th>
<th>Test</th>
<th>Sample Type</th>
<th>Sensitivity*</th>
<th>Specificity*</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Chlamydia trachomatis</strong></td>
<td>Biostar OIA Chlamydia test</td>
<td>Cervical</td>
<td>59.4-73.8%</td>
<td>98.4-100%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Male Urine</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Clearview Chlamydia</td>
<td>Cervical</td>
<td>49.7%</td>
<td>97.9%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Vaginal</td>
<td>32.8%</td>
<td>99.2%</td>
</tr>
<tr>
<td></td>
<td>Quick Vue</td>
<td>Cervical</td>
<td>25-65%</td>
<td>100%</td>
</tr>
<tr>
<td></td>
<td>Chlamydia Rapid Test</td>
<td>Vaginal</td>
<td>83.5%</td>
<td>98.9%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Male Urine</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>X-pert CT/NG</td>
<td>Cervical</td>
<td>97.4%</td>
<td>99.6%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Vaginal</td>
<td>98.7%</td>
<td>99.4%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Female Urine</td>
<td>97.6%</td>
<td>99.8%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Male Urine</td>
<td>97.8%</td>
<td>99.9%</td>
</tr>
<tr>
<td><strong>Neisseria gonorrhoeae</strong></td>
<td>Biostar OIA GC test</td>
<td>Cervical</td>
<td>60%</td>
<td>89.9%</td>
</tr>
<tr>
<td></td>
<td>PATH GC-Check</td>
<td>Cervical</td>
<td>70%</td>
<td>97.2%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Vaginal</td>
<td>54.1%</td>
<td>98.25</td>
</tr>
<tr>
<td></td>
<td>X-pert CT/NG</td>
<td>Cervical</td>
<td>100%</td>
<td>100%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Vaginal</td>
<td>100%</td>
<td>99.9%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Female Urine</td>
<td>95.6%</td>
<td>99.9%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Male Urine</td>
<td>98.9%</td>
<td>99.9%</td>
</tr>
</tbody>
</table>


Adapted from Huppert et al. (2010). * Sensitivity and specificity compared to NAATs
What about new CT/NG tests coming along?

1. MAMEF-based DNA detection (microwave accelerated metal enhanced fluorescence)
2. Atlas Velox TM System
3. MobiLab
1. MAMEF-based DNA detection

- Microwave-based lysing
- Ultra-rapid and sensitive detection of biomolecules

Fluorescent Probe
22 nt

Target Sequence
47 nt

Anchor Probe
21 nt

Microwave-Accelerated Metal-Enhanced Fluorescence DNA detection
Clinical evaluation of CT MAMEF

Blind Evaluation of the Microwave-Accelerated Metal-Enhanced Fluorescence Ultrarapid and Sensitive *Chlamydia trachomatis* Test by Use of Clinical Samples

<table>
<thead>
<tr>
<th></th>
<th>NAAT+ / MAMEF +</th>
<th>NAAT+ / MAMEF -</th>
<th>NAAT- / MAMEF +</th>
<th>NAAT- / MAMEF -</th>
<th>Clinical Sensitivity (%)</th>
<th>Concordance (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cryptic plasmid</td>
<td>37</td>
<td>8</td>
<td>15</td>
<td>197</td>
<td>82.2</td>
<td>91.1</td>
</tr>
<tr>
<td>16S rRNA</td>
<td>34</td>
<td>11</td>
<td>15</td>
<td>197</td>
<td>75.5</td>
<td>89.9</td>
</tr>
<tr>
<td>Both assays</td>
<td>33</td>
<td>12</td>
<td>15</td>
<td>197</td>
<td>77.3</td>
<td>89.5</td>
</tr>
</tbody>
</table>

- 257 vaginal swabs – 245 adolescents and young women

- Less than 10 minutes
- $1.50 per test
- $2,500 reader
2. Atlas Velox TM System

Platform/Equipment
- Small footprint
- Low cost
- No reagents on board
- No fragile optical sensors
- Portable – Robust reader for POC settings

Disposable cartridge
- Reagent stabilised on card
- 20 minutes
- Simple to use system - designed to meet CLIA Waiver
  - Chlamydia (lead product)
  - Chlamydia & Gonorrhea

• Electrochemical label released from probe hybridised to target by nuclease enzyme
100 patient samples determined to be positive or negative for Chlamydia using the BD test

<table>
<thead>
<tr>
<th></th>
<th>Disease Present</th>
<th>Disease Absent</th>
<th>Totals</th>
</tr>
</thead>
<tbody>
<tr>
<td>positive</td>
<td>49</td>
<td>0</td>
<td>49</td>
</tr>
<tr>
<td>negative</td>
<td>1</td>
<td>50</td>
<td>51</td>
</tr>
<tr>
<td>Total</td>
<td>50</td>
<td>50</td>
<td>100</td>
</tr>
</tbody>
</table>

98% sensitivity  
100% specificity

306 patient samples determined to be positive or negative for Chlamydia using Roche or Gen-Probe test

<table>
<thead>
<tr>
<th>Johns Hopkins Results</th>
<th>GeneProbe/Roche Assay Result</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Positive</td>
</tr>
<tr>
<td>Atlas Genetics Assay Result</td>
<td>Positive</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
</tr>
</tbody>
</table>

98% sensitivity  
98% specificity

3. MobiLab

- A Smartphone-enabled microfluidic NAAT for CT

- Three discrete units: a droplet pendant microfluidic cartridge, a battery-powered instrument for droplet manipulation and amplification, and a smartphone for user interface, data acquisition and processing.

- A single-stream loop-mediated isothermal amplification (LAMP) assay to operate in tandem with the mobiLab platform.

- Polyhistidine-coated magnetic particles capture nucleic acid targets from sample lysate via electrostatic interaction.
Trichomonas vaginalis

- Wet Preparation showing motile trichomonads
- Stained Trichomonas
- Electron microscope view of trichomonas on epithelial cell

- Wet Preparation
- Affirm
- OSOM POC
## Trichomonas Test Comparisons

<table>
<thead>
<tr>
<th>Test Type</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wet prep</td>
<td>55%–65%</td>
<td>100%</td>
</tr>
<tr>
<td>Culture</td>
<td>75%</td>
<td>100%</td>
</tr>
<tr>
<td>POCT (OSOM)</td>
<td>&gt;83%</td>
<td>&gt;97%</td>
</tr>
<tr>
<td>PCR (LDT)</td>
<td>83-92%</td>
<td>100%</td>
</tr>
<tr>
<td>TMA AptimaTV</td>
<td>100%</td>
<td>100%</td>
</tr>
<tr>
<td>ProbTec TVQ</td>
<td>98.3%</td>
<td>98.3%</td>
</tr>
</tbody>
</table>

References:


Van Der Pol; Schwebke; Taylor: Posters STI & AIDS, 2013.
<table>
<thead>
<tr>
<th>Organism</th>
<th>Test</th>
<th>Sample Type</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Trichomonas vaginalis (TV)</em></td>
<td>OSOM TV Rapid Test</td>
<td>Vaginal Swabs</td>
<td>83.3-90%*</td>
<td>98.8%-100%*</td>
</tr>
<tr>
<td></td>
<td>Affirm VPIII Microbial Identification Test</td>
<td>Vaginal Swabs</td>
<td>46.3%**</td>
<td>100%**</td>
</tr>
</tbody>
</table>


*-The range reflects reported sensitivities and specificities reported in Huppert et al. (2010), Nye et al. (2009), Patullo et al. (2009) and Madhivavan et al. (2013).*

**-Affirm compared to nucleic acid amplification test, JCM, Cartwright et al. (2013).
OSOM Rapid TV Antigen Test

- Immunochromato-graphic detection
- TV membrane proteins
- Mouse antibodies
- Latex beads/capillary action

Huppert et al, JCM 2005; STI 2007: Sensitivity 83-90%, Specificity 98-100%

**POSITIVE**
- A blue Test Line and a red Control Line is a positive result

**NEGATIVE**
- A red Control Line but no blue Test Line is a negative result.
Why is the impact of TV important?
Syphilis: Serologic DX requires detection of two types of antibodies

- Non-Treponemal: RPR, VDRL (Can be POC)
- Treponemal: FTA-abs, TPPA, Many new POC

- Biologic false positive non-treponemal test
- Falsely reactive treponemal test due to cross-reacting serum antibodies
- Both test types have imperfect specificity

- Reactive treponemal test cannot distinguish active from inactive infection
Syphilis serologic screening algorithms

**Traditional**

- Quantitative RPR
  - RPR+
    - TP-PA+ or other trep. test
      - TP-PA+ Syphilis (past or present)
      - TP-PA- Syphilis unlikely
  - RPR-

**Reverse sequence**

- EIA or CIA
  - EIA/CIA+
    - Quantitative RPR
      - Evaluate clinically
        - RPR+ Syphilis (past or present)
        - TP-PA+ Syphilis (past or present)
        - TP-PA- Syphilis unlikely
      - RPR-
        - TP-PA
  - EIA/CIA-

CDC recommended algorithm for reverse sequence syphilis screening followed by nontreponemal test confirmation. If at risk for syphilis, repeat RPR in several weeks.
Treponemal tests

- Fluorescent treponemal antibody absorbed (FTA-ABS) test
- Treponema pallidum particle agglutination (TP-PA) test
- Enzyme immunoassays (EIAs)
  - Trep-Chek
  - Trep-Sure
  - Captia G
- Chemiluminescence immunoassays (CIAs)
  - LIAISON
  - Architect
- Multiplex flow immunoassays (MFI)
  - BioPlex 2200 Syphilis IgM and IgG
  - AtheNA Multi-Lyte
  - ADVIA Centaur SYPH test
- POC---Immunochromatographic strip tests (ICS)
  - Syphilis Health Check
  - Dual Path Platform (DPP) Syphilis Screen & Confirm Assay (SSCA)
POC Syphilis Health Check™

Syphilis Antibody Rapid Immunochromatographic Test

• Rapid qualitative screening for human TP antibodies in whole blood, serum or plasma
• Results in 10 minutes; 2 steps; room temperature
• 98% agreement to other treponemal tests
• Serum, plasma or whole blood or finger-stick

Negative: 1 colored band in control area
Positive: Colored bands in test area and control area
Inconclusive: No distinct color bands in either area

FDA Cleared
CLIA Waived
Evaluation of an Immunochromatographic Point-of-Care Test for the Simultaneous Detection of Nontreponemal and Treponemal Antibodies in Patients With Syphilis

Rita Castro, MD, PhD,∗† Ângela Lopes, Bsc,‡ and Filomena da Luz Martins Pereira, MD, PhD§

### DPP Syphilis Screening & Confirm Assay (SSCA)

<table>
<thead>
<tr>
<th>SSCA</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Compared to RPR</td>
<td>98.8%</td>
<td>94.7%</td>
</tr>
<tr>
<td></td>
<td>171/173</td>
<td>71/75</td>
</tr>
<tr>
<td>Compared to TPHA</td>
<td>95.5%</td>
<td>88.8%</td>
</tr>
<tr>
<td></td>
<td>184/185</td>
<td>56/63</td>
</tr>
<tr>
<td>Compared to FTA-abs</td>
<td>98.9%</td>
<td>93.2%</td>
</tr>
<tr>
<td></td>
<td>187/189</td>
<td>55/59</td>
</tr>
</tbody>
</table>
A solid phase immunochromatographic assay
Qualitative detection of antibodies of all isotypes (IgG, IgM, IgA) against Treponema pallidum
Simultaneously in serum, plasma, or whole blood
Recombinant TP (15kDa, 17kDa) antigens used as captures and detectors; 2-30°C Storage
Standard Diagnostics-Alere
Determine™ Syphilis TP
POC Test


**Detects**
Antibodies to *Treponema pallidum* at POC

**Rapid**
Provides accurate and reliable results in 15 minutes

**Convenient**
No refrigeration required (storage 2-30°C)
No power or water source is needed to run test

**Flexible**
Uses serum, plasma or whole blood by venipuncture or finger prick
What about Combination Syphilis and HIV POC tests?

These are coming soon—stay tuned

Here is a preview
DPP® HIV-Syphilis Assay


Video whole Blood Sample

http://www.youtube.com/watch?v=DE4Wxy4byQE&x-yt-ts=1401912551

- Chembio Diagnostic Systems has developed a dual HIV 1/2 and Syphilis Treponemal antibodies POC test (Dual Path Platform (DPP®) technology)

- Immunochromatographic rapid screening POC test

- Fingerstick whole blood, venous whole blood, serum, and plasma

18 month shelf-life at 2-30°C
No refrigeration or cold chain required
No timers required
Results are easy to interpret
No specialized equipment required

3 minute test procedure
Whole blood, serum or plasma specimens
No specialized training required
Built-in procedural and reagent control line
• SD BIOLINE HIV/Syphilis Duo test is a solid phase immunochromatographic assay
• Qualitative detection of antibodies to all isotypes (IgG, IgM, and IgA) specific to HIV-1/2 and/or Treponema palladium (TP)
• Serum, plasma, or whole blood
• 1-30°C for 24 months
HIV

CLIA-Waived Point-of-Care Rapid HIV Tests

OraQuick Advance

Uni-Gold Recombigen

Clearview Complete

Clearview Stat Pak

INSTI
DPP HIV-1/2 Assay

- CLIA moderate complexity for serum, plasma, oral fluid
- “SampleTainer” = residual specimen after testing
- FDA-approved Dec 21, 2012
## Sensitivities and Specificities for POC Tests for HIV

<table>
<thead>
<tr>
<th>Organism</th>
<th>Test</th>
<th>Sample Type</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV*</td>
<td>OraQuick Advance Rapid HIV-1/2 Antibody Test</td>
<td>Oral Fluid, Whole blood/Serum</td>
<td>99.6%</td>
<td>100%</td>
</tr>
<tr>
<td></td>
<td>Reveal G3 Rapid HIV-1 Antibody Test</td>
<td>Serum/Plasma</td>
<td>99.8%</td>
<td>99.9%</td>
</tr>
<tr>
<td></td>
<td>Multispot HIV-1/HIV-2 Rapid Test</td>
<td>Serum/Plasma</td>
<td>100%</td>
<td>99.9%</td>
</tr>
<tr>
<td></td>
<td>Uni-Gold Recombigen HIV Test</td>
<td>Whole blood/Serum/Plasma</td>
<td>100%</td>
<td>99.7%</td>
</tr>
<tr>
<td></td>
<td>Clearview HIV-1/2 Stat-Pak or Clearview Complete HIV ½</td>
<td>Whole blood/Serum/Plasma</td>
<td>99.7%</td>
<td>99.9%</td>
</tr>
<tr>
<td></td>
<td>Chembio DPP HIV1/2 Assay</td>
<td>Oral Fluid, Whole blood/Serum/Plasma</td>
<td>99.8%</td>
<td>99.9%</td>
</tr>
<tr>
<td></td>
<td>INSTI HIV-1 Antibody Test</td>
<td>Whole blood/Plasma</td>
<td>99.8%</td>
<td>99.5%</td>
</tr>
</tbody>
</table>


*Adapted and Updated from Huppert et. al. (2010) and Branson (2007)
**New HIV Diagnostic Algorithm**

4th generation HIV-1/2 immunoassay

- **(-)** Negative for HIV-1 and HIV-2 antibodies
- **(+)** Negative for HIV-1 and HIV-2 antibodies and p24 Ag

HIV-1/HIV-2 antibody differentiation immunoassay (i.e. Multispot rapid)

- **HIV-1**
  - HIV-1 antibodies detected
  - Initiate care (and viral load)

- **HIV-2**
  - HIV-2 antibodies detected
  - Initiate care

- **HIV-1 +/HIV-2 +**
  - HIV antibodies detected

- **HIV-1&2 (-) or indeterminate**

  - **RNA**
    - **RNA (+)** Acute HIV-1 infection
      - Initiate care
    - **RNA (-)** Negative for HIV-1

---

Herpes Simplex Assays
Serology vs. Virus Detection

- Serology for Antibody
- Tzanck Smear for inclusions
- Virology Culture for virus
- ELVIS (Diag.Hybrids) HSV type
- New FDA NAAT test for virus (HSVQ\textsuperscript{x})
  - HSV-1 and 2 for lesions

- IsoAmp POC for virus genital and oral swabs
# Sensitivities and Specificities for Serology & POC Diagnostics for HSV-2

<table>
<thead>
<tr>
<th>Herpes Simplex Virus 2 (HSV-2)</th>
<th>Test</th>
<th>Sample Type</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Serology</strong></td>
<td><strong>HerpeSelect</strong></td>
<td>Serum</td>
<td>80-100%*</td>
<td>41-100%*</td>
</tr>
<tr>
<td></td>
<td><strong>HerpeSelect</strong></td>
<td>Serum</td>
<td>91%**</td>
<td>97%**</td>
</tr>
<tr>
<td><strong>Serology</strong></td>
<td><strong>Kalon HSV-2 gG2</strong></td>
<td>Serum</td>
<td>84-98.6%*</td>
<td>83.2-100%*</td>
</tr>
<tr>
<td><strong>Virus</strong></td>
<td><strong>Rapid Real-Time PCR LDT ABI7500 Fast</strong></td>
<td>Genital lesions</td>
<td>96.7%***</td>
<td>99.6%***</td>
</tr>
<tr>
<td><strong>Virus</strong></td>
<td><strong>Qx PCR (BD)</strong></td>
<td>Lesion vs. PIS</td>
<td>95.9-97.3%†</td>
<td>95.7-100%</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Lesion vs. PCR</td>
<td>95.7-100%</td>
<td>95.8-100%</td>
</tr>
<tr>
<td><strong>Virus</strong></td>
<td><strong>IsoAMP HSV POC (Biohelix)</strong></td>
<td>Genital swabs</td>
<td>97.1%***</td>
<td>93.4%****</td>
</tr>
</tbody>
</table>


Adapted from Biaro et al. (2011). Sensitivity and specificity are expressed as a range from multiple studies over multiple years from the meta-analysis. **Adapted from Zahariadis et. al. (2010); ***Adapted from Gardella et. al. (2010); †Van Der Pol et al. J Clin Microbiol 2012;51:3466-3471.

****Adapted from Lemieux et. al. (2012)
New Virus POC HSV Test- IsoAMP

- FDA cleared POC assay for the detection of HSV in lesions; IsoAMP® HSV (Biohelix Corporation, Beverly, MA)

- Technology utilizes isothermal helicase dependent amplification (HAD), which uses Bst DNA polymerase, and by obviating the nucleic acid extraction process, can offer results in 1.5 hours

- From five study sites in the U.S., and after discrepant analysis, overall agreement of IsoAmp with ELVIS was 98.8%, with a 37.0% overall prevalence


A rapid and simple isothermal nucleic acid amplification test for detection of herpes simplex virus types 1 and 2

Hyun-Jin Kim, Yanhong Tong, Wen Tang, Louisito Quimson, Vicki A. Cope, Xiaojing Pan, Aurelie Motre, Richard Kong, Jian Hong, Debbie Kohn, Nancy S. Miller, Melinda D. Poulter, Huimin Kong, Yi-Wei Tang, Belinda Yen-Lieberman

• **Analytical sensitivity of the assays was 5.5 and 34.1 copies/reaction for HSV-1-2**

• **Viral culture was used as the reference standard, the clinical sensitivity and specificity of the IsoAmp® HSV assay were 100.0% and 96.3% respectively.**
The IsoAmp® HSV Assay (Biohelix Corp)

- FDA-cleared for HSV in genital and oral lesions
- The IsoAmp HSV has a test-to-result time of <1.5 hr.
- Isothermal helicase-dependent amplification (HDA) technique; no nucleic acid extraction
- The rapid and simple characteristics of the IsoAmp HSV assay make it potentially suitable for POC testing

Lemieux et al. Expert Reviews Ltd. 437-443, 2012;
Why do POCTs?

- Improve patient satisfaction
- Treat patients before leave clinic
- Improve clinical practice efficiency
- Revenues from testing
- Improve medical outcomes

Published data are few to substantiate these points
Barriers to implementation of POCTs

• Financial viability
• Money for instruments and consumables
• Obtaining CLIA certificate
• Validating the test(s)
• Policies and procedures (training manuals)
• Operator training (recertification)
• Getting results into the EMR (interface- ??7K?)
• Space
• Billing and Reimbursement
Money for instruments and consumables

- **Instruments**
  - For many small devices the price of the instrument is included in the supplies
  - Larger instruments may cost $5-20 K

- **Consumables**
  - Consumable costs must be covered by billing revenues which will vary by test, payer and volume used
<table>
<thead>
<tr>
<th>Test Complexity</th>
<th>Categorization of Test Complexity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Waived</td>
<td>• Many are approved by FDA for home use</td>
</tr>
<tr>
<td></td>
<td>• Uses basic methodologies</td>
</tr>
<tr>
<td></td>
<td>• Present little risk if improperly performed</td>
</tr>
<tr>
<td>Provider Performed Microscopy (Physician, Nurse, Physician Assistant) as part of physical exam</td>
<td>• Specimen is labile and any delay will impact accuracy</td>
</tr>
<tr>
<td></td>
<td>• No controls available for monitoring the complete test process</td>
</tr>
<tr>
<td></td>
<td>• Limited to unstained microscopic examinations</td>
</tr>
<tr>
<td>Moderate and High Complexity</td>
<td>Assigned based on 7 criteria</td>
</tr>
<tr>
<td></td>
<td>• Knowledge required to perform</td>
</tr>
<tr>
<td></td>
<td>• Training required to perform</td>
</tr>
<tr>
<td></td>
<td>• Complexity of test preparation</td>
</tr>
<tr>
<td></td>
<td>• Complexity of operational steps</td>
</tr>
<tr>
<td></td>
<td>• Calibration of quality control and proficiency testing</td>
</tr>
<tr>
<td></td>
<td>• Complexity of troubleshooting and maintenance</td>
</tr>
<tr>
<td></td>
<td>• Level of judgment required to do test</td>
</tr>
</tbody>
</table>
Workflow

• Ideally, test will be available at time physician sees patient without need to return to examining room
  – Requires test are predicted as necessary and performed ahead of time
  – Test is so fast it does not disrupt encounter

• Deciding which tests will be required and when depends on the patient type: New patient, check-up, management visit, sick visit
Conclusions

POCTs in primary/STI care have potential:

• Improve patient outcomes
• Improve practice efficiency
• Reduce total tests ordered
• Improve practice revenues

But there are barriers to successful implementation that need to be overcome which can be costly, time consuming, and require learning new skill sets
Conclusion: The Promising Future

• Better POC tests; self-collection; self-testing
• Testing outside a clinic
  ✓ Internet recruitment, pharmacy, vans
• Cheaper test kits
• Use of research to remove barriers to testing
• Learn how to effectively use new tools and how new research can improve the detection of STDs
  ✓ Provide cost-effective ways to increase the number of patients being tested and treated
Acknowledgements

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• Jeff Holden
• Laura Dize
• Perry Barnes
• Billie Masek

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410-614-0932
Use of POC in Clinical Settings

• Immediate treatment before patient leaves the clinic; no loss to follow-up

• Impact on disease epidemic?
  – Decrease interval of disease spread

• Impact on behavior?
  – Counseling on risk reduction

• **ASSURED** Criteria by WHO
  – When is a test good enough?

http://www.who.int/std_diagnostics/about_SDI/priorities.htm
Affordable by those at risk of infection

Sensitive few false negatives

Specific few false positives

User-friendly simple to perform: 3-4 steps, with minimal training

Rapid and Robust
  rapid: to enable treatment at first visit
  robust: no requirement refrigerated storage

Equipment-free easily collected non-invasive specimens, e.g. urine, saliva

Delivered delivered to end-users

http://www.who.int/std_diagnostics/about_SDI/priorities.htm
POCT – Build Your Own Test

• First Priority of Needs Assessment Survey
  – Chlamydia (62%); HIV – Early Seroconversion (14%)
  – Syphilis (8%)

• Overall, participants selected sensitivity as their top priority, followed by cost, specificity, and time

• Choices (statistically significant)
  Sensitivity: 90-99% > 80-90% > 70-80%
  Cost: $20 > $35 > $50
  Specificity: 99% > 95% > 90%
  Time: 5 > 15 > 25 minutes

## Preferences in Attributes by Prioritized Test

<table>
<thead>
<tr>
<th>Attributes</th>
<th>Odds Ratios * all p-values &lt;0.05</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Priority Force Choice Questions</strong></td>
<td></td>
</tr>
<tr>
<td>ALL N=218</td>
<td>Chlamydia N= 136</td>
</tr>
<tr>
<td>Chlamydia N= 136</td>
<td>Early HIV Seroconversion N=30</td>
</tr>
<tr>
<td>Early HIV Seroconversion N=30</td>
<td>Syphilis N=21</td>
</tr>
<tr>
<td><strong>Sensitivity (%)</strong></td>
<td>90-99</td>
</tr>
<tr>
<td>90-99</td>
<td>13.6*</td>
</tr>
<tr>
<td>136</td>
<td>18.2*</td>
</tr>
<tr>
<td>30</td>
<td>10.6*</td>
</tr>
<tr>
<td>21</td>
<td>11.8*</td>
</tr>
<tr>
<td><strong>Specificity (%)</strong></td>
<td>99</td>
</tr>
<tr>
<td>99</td>
<td>3.7</td>
</tr>
<tr>
<td>136</td>
<td>3.7</td>
</tr>
<tr>
<td>30</td>
<td>4.7*</td>
</tr>
<tr>
<td>21</td>
<td>5.9*</td>
</tr>
<tr>
<td><strong>Cost ($)</strong></td>
<td>20</td>
</tr>
<tr>
<td>20</td>
<td>4.5*</td>
</tr>
<tr>
<td>136</td>
<td>5.2*</td>
</tr>
<tr>
<td>30</td>
<td>3.2</td>
</tr>
<tr>
<td>21</td>
<td>4.3</td>
</tr>
<tr>
<td><strong>Time (minutes)</strong></td>
<td>5</td>
</tr>
<tr>
<td>5</td>
<td>3.0</td>
</tr>
<tr>
<td>136</td>
<td>3.2</td>
</tr>
<tr>
<td>30</td>
<td>2.5</td>
</tr>
<tr>
<td>21</td>
<td>3.6</td>
</tr>
</tbody>
</table>

Forced Choice Questions used in a survey with multivariate analysis

What qualities do providers identify as best for POC STI tests: Do opinions differ by practice, region and country?

Results:
• 190 subjects replied to the survey: 46% male and 54% female
• Europe (27%), Oceana (26%), America (22%), Africa (11%), Asia (11%)
• The majority (61%) were from developed countries
• Unreliability (19.5%) was the characteristic considered the greatest barrier for use of POCTs, followed by a technology that was laboratory-driven (12%) and complexity (12%) of performing the test.
<table>
<thead>
<tr>
<th>Disease</th>
<th>% Respondents from Developed Country</th>
<th>% Respondents from Resource Constrained Countries</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlamydia</td>
<td>56</td>
<td>26</td>
</tr>
<tr>
<td>Gonorrhea</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td>Hepatitis B/C</td>
<td>2</td>
<td>7</td>
</tr>
<tr>
<td>Herpes Simplex Virus</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>HIV (early Seroconversion)</td>
<td>21</td>
<td>31</td>
</tr>
<tr>
<td>Human papillomavirus</td>
<td>2</td>
<td>9</td>
</tr>
<tr>
<td>Syphilis</td>
<td>8</td>
<td>16</td>
</tr>
<tr>
<td>Trichomonas</td>
<td>1</td>
<td>1</td>
</tr>
</tbody>
</table>
The greatest barrier for use of POCTs (n=190)

- Unreliability: 19%
- Laboratory-driven: 12%
- Complexity: 12%
- Time Frame: 12%
- Patient wait time: 10%
- workflow interruption: 10%
- other: 6%
- 24%

(no differences between developing and developed for the top 3)
Important Drivers of POC Adoption

- Sensitivity, specificity, and time are important
- Willingness to pay the cost is important
- Willingness to self-collect specimens is probably important
- What about the patient’s needs?
Point-of-care tests for sexually transmitted infections: what do “end users” want? (N=58)

- Five focus groups (Baltimore & Cincinnati)
- Favorable POCTs (Rapid, Easy to read, Simple to use)
- Home testing acceptable – better privacy
- Clinic-based- definitive results & immediate treatment
- Barriers- cost and ability to read and perform tests
- Hispanic patients questioned home test reliability, wanted high sensitivity and bi-lilingual instructions

Table 1. Advantages of having access to a STI point-of-care test in the clinic or at home

<table>
<thead>
<tr>
<th>Settings</th>
<th>Advantages</th>
</tr>
</thead>
<tbody>
<tr>
<td>In the clinic</td>
<td>Know the results right away</td>
</tr>
<tr>
<td></td>
<td>Saves time</td>
</tr>
<tr>
<td></td>
<td>Quicker services</td>
</tr>
<tr>
<td></td>
<td>Do not have to wait</td>
</tr>
<tr>
<td>At home</td>
<td>Convenience-</td>
</tr>
<tr>
<td></td>
<td>Confidentiality</td>
</tr>
<tr>
<td></td>
<td>Confidentiality</td>
</tr>
<tr>
<td></td>
<td>Privacy</td>
</tr>
<tr>
<td></td>
<td>No appointment needed</td>
</tr>
<tr>
<td></td>
<td>No need to go to clinic</td>
</tr>
<tr>
<td></td>
<td>Less embarrassment</td>
</tr>
<tr>
<td></td>
<td>Increased health awareness</td>
</tr>
<tr>
<td></td>
<td>Empowerment</td>
</tr>
</tbody>
</table>

Rompalo et al. Sexual Health 2013;10:541-545
New CDC Recommendations for HIV Testing in Laboratories
A step-by-step account of the approach

CDC’s new recommendations for HIV testing in laboratories capitalize on the latest available technologies to help diagnose HIV infections earlier – as much as 3-4 weeks sooner than the previous testing approach. Early diagnosis is critical since many new infections are transmitted by people in the earliest (“acute”) stage of infection.

By putting the latest testing technology to work in laboratories across the United States, we can help address a critical gap in the nation’s HIV prevention efforts.

**Step 1:** “Fourth generation” HIV test
*Detecting HIV sooner*
Detects HIV in the blood earlier than previously recommended antibody tests by identifying the HIV-1 p24 antigen, a viral protein which appears in the blood sooner than antibodies.

**Step 2:** HIV-1/HIV-2 antibody differentiation immunoassay
*Diagnosing HIV-1 vs. HIV-2*
Produces results faster than the previously recommended Western Blot.
Distinguishes between HIV-1 and HIV-2, which the previously recommended Western Blot cannot do – this distinction can have important treatment implications for a patient.

**Step 3:** Nucleic Acid Test (NAT)
*Acute HIV-1 infection or “false positive”?
Ensures accurate detection of early infection or indicates a false positive from the fourth generation test.

This graphic is designed to illustrate key concepts of the new testing approach in laboratories. For more detail, please see the full guidelines here: http://www.cdc.gov/hiv/pdf/HIVtestingAlgorithmRecommendation-Final.pdf.