Developing diagnostics for resource-limited settings

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University of Cambridge

Trends in diagnostics: becoming a bimodal distribution

- Primary health clinics
- Emergency rooms
- Home
  - Near patient
  - Over the counter
  - Targeted
- Hospital labs
- Clinical labs
  - Remote
  - Centralised
  - Automated
  - Integrated
  - Panel testing

Low ↔ batch size ↔ High
## Prevalence of blood-borne viruses in 1,294 donors (Kumasi, Ghana)

<table>
<thead>
<tr>
<th>Virus</th>
<th>No. infected</th>
<th>% infected</th>
</tr>
</thead>
<tbody>
<tr>
<td>HBV</td>
<td>204</td>
<td>15.8</td>
</tr>
<tr>
<td>HCV</td>
<td>31</td>
<td>2.4</td>
</tr>
<tr>
<td>HIV</td>
<td>19</td>
<td>1.5</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>254</strong></td>
<td><strong>19.6%</strong></td>
</tr>
</tbody>
</table>

Allain et al. 2004

## Prevalence of viral markers in blood donors

<table>
<thead>
<tr>
<th>Country</th>
<th>Prevalence (%)</th>
<th>Cumulative %</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>HBV</td>
<td>HCV</td>
</tr>
<tr>
<td>Botswana</td>
<td>5</td>
<td>10.0</td>
</tr>
<tr>
<td>Cameroon</td>
<td>10.7</td>
<td>4.8</td>
</tr>
<tr>
<td>Egypt</td>
<td>5</td>
<td>12.0</td>
</tr>
<tr>
<td>Malawi</td>
<td>8</td>
<td>1.0</td>
</tr>
<tr>
<td>S. Africa</td>
<td>4</td>
<td>0.5</td>
</tr>
<tr>
<td><strong>UK (2003)</strong></td>
<td><strong>0.00073</strong></td>
<td><strong>0.00086</strong></td>
</tr>
</tbody>
</table>
Budget of blood bank at Kumasi, Ghana

- **Annual budget $70,000 (7,800 donors)**
- **Consumables (17%)**
- **Salary (13%)**
- **Overhead (10%)**
- **Equipment (10%)**
- **Test (15%)**
- **Blood bag (35%)**

**Imbalance of resources**

- **Ghana Kumasi Blood Bank**
  - **$ 1.5 for all testing**
- **Germany Red Cross**
  - **€ 3.8 million to detect 1 more HIV case**

Opare-Sem 2002
Bias of current test components for viral subtypes

<table>
<thead>
<tr>
<th>Virus</th>
<th>Subtype</th>
<th>Frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td>HCV</td>
<td>1a</td>
<td>&lt; 5%</td>
</tr>
<tr>
<td>HIV</td>
<td>B</td>
<td>&lt; 5%</td>
</tr>
<tr>
<td>HBV</td>
<td>A</td>
<td>&lt; 2%</td>
</tr>
</tbody>
</table>

- Test components optimal for developed world
- Insufficient sensitivity for certain geographic areas

HIV-1 subtype distribution in sub-Saharan Africa
Non-B HIV-1 subtype in French blood donors

Diagnostics for resource-limited settings
An unmet need

Academia

- Lack of product development expertise
  - Validation
  - Scale-up
  - Documentation
  - Patent protection

- Applied research not valued by environment
- Not their raison d'être
Diagnostics for resource-limited settings
An unmet need

Private Sector

- Low return of investment
- Low profit margin for rapid tests
- Imperative to use existing production line
- Technical inadequacies (stability, sensitivity)
- Problematic marketing/distribution channel
- Difficulties in service, repair and technical support

R & D for neglected diseases

<table>
<thead>
<tr>
<th>Skill &amp; infrastructure</th>
<th>Incentives</th>
</tr>
</thead>
<tbody>
<tr>
<td>Large company</td>
<td>Small company</td>
</tr>
<tr>
<td>Academic group</td>
<td>Academic group</td>
</tr>
</tbody>
</table>

Incentives: Skill & infrastructure
Creating a structure that blurs traditional boundaries

Goal - developing appropriate diagnostics for resource-poor settings

Assay format for developing countries

- Inexpensive, rapid and easy to use
- Stable for high humidity and poor storage/transport conditions
- Use of non-invasive sample types
- Flexible format to meet regional needs
- Designed for simplicity and large volume
- Incorporate cutting-edge technologies
How a dipstick assay works

Choosing the first target: Chlamydia trachomatis

- 90 million new cases worldwide
- Major cause of infertility and PID
- $4 billion in US, £100 million in UK
- Cost-effective treatment
- High % asymptomatic infections
- Lack of diagnostic screening tools
Signal amplification technology to improve sensitivity

<table>
<thead>
<tr>
<th>Concentration of Chlamydia LPS (picograms)</th>
<th>0</th>
<th>420</th>
<th>125</th>
<th>40</th>
<th>15</th>
</tr>
</thead>
<tbody>
<tr>
<td>Conventional rapid test</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Signal amplified</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Components of a diagnostic test

- Chemistry
- Detection
- Generic reagents
- Sample collection & preparation
FirstBurst™ - First void urine collector

- Unique collection & disposable device
- Reliable and convenient collection of first catch urine
- Sample with higher bacterial load
- ‘2003 Best Diagnostic Medical Futures Innovation’ award

Chlamydia Rapid Test

- Home use
- Clinical laboratory use
Field trial in Iloilo, Philippines

The ‘Living room’ project, Amsterdam
Field trial at Brook, Birmingham

• 98% of study participants found the instructions easy to understand
• 85% of women and 67% of men willing to wait 1 hour or more for their result

Preliminary results of Cambridge *Chlamydia* Rapid Test in vaginal swabs

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>Sensitivity (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCR</td>
<td>100</td>
</tr>
<tr>
<td>Endocervical swab</td>
<td>67 ± 10</td>
</tr>
<tr>
<td>Vaginal swab</td>
<td>70 ± 9</td>
</tr>
<tr>
<td>Clearview</td>
<td>50 ± 7</td>
</tr>
<tr>
<td>Cambridge</td>
<td>90 ± 15</td>
</tr>
</tbody>
</table>

* Clearview test performed in a high prevalence (sex workers) population in the Philippines
* Cambridge test performed in a high prevalence (sex workers) population and a low prevalence (OB-GYN) population in the Philippines
* Cambridge test performed in a young people’s sexual health clinic (16-25) in the United Kingdom
Diagnostics: only part of the equation

Diseases in resource-poor settings

**The rapid test paradox**
A less sensitive test results in more patients treated

**Nucleic acid tests**  2-3 weeks - test to results

- Sample
- Shipment
- Testing
- Notification
- Physician
- Treatment

Sensitivity (90%) x return rate (70%) = **63% treated**

**Rapid tests**  < 40 min

- Sample
- On-site testing
- Treatment

Sensitivity (70%) x return rate (100%) = **70% treated**

Modified from Gift et al. 1999
**Chlamydia prevalence in UK women 16-24 years old, 2003/04**

<table>
<thead>
<tr>
<th>Region</th>
<th>Prevalence</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>7.3%</td>
</tr>
<tr>
<td></td>
<td>10.7%</td>
</tr>
<tr>
<td></td>
<td>11.6%</td>
</tr>
<tr>
<td></td>
<td>11.9%</td>
</tr>
<tr>
<td></td>
<td>12.6%</td>
</tr>
<tr>
<td>London</td>
<td>7.3%</td>
</tr>
<tr>
<td></td>
<td>13.4%</td>
</tr>
</tbody>
</table>

**Blinding Trachoma**

- 146 million infected, 3 million blind or visually disabled
### Chlamydia Rapid Test performance in eye swabs

<table>
<thead>
<tr>
<th>PCR</th>
<th>+</th>
<th>-</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>+</td>
<td>49</td>
<td>10</td>
<td>59</td>
</tr>
<tr>
<td>-</td>
<td>3</td>
<td>202</td>
<td>205</td>
</tr>
<tr>
<td>Total</td>
<td>52</td>
<td>212</td>
<td>264</td>
</tr>
</tbody>
</table>

- **Sensitivity**: 83%
- **Specificity**: 98.5%

### Inadequacy of clinical signs in Trachoma

<table>
<thead>
<tr>
<th></th>
<th>Rapid test</th>
<th>Clinical signs</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCR+ without clinical symptom</td>
<td>35.9%</td>
<td></td>
</tr>
<tr>
<td>PCR - with clinical symptom</td>
<td>19.8%</td>
<td></td>
</tr>
</tbody>
</table>

- **Sensitivity**: 83%
- **Specificity**: 98.5%
Power of nucleic acid testing (NAT)

- High degree of analytical sensitivity
- Specific detection of infectious agent genome
- Applications
  - Early detection during window period
  - Diagnosis of infections in newborns from infected mothers
  - Anti-retroviral therapy monitoring
  - Improvement of the safety of blood supply

Complexity of current nucleic acid test (NAT)
Visual detection of nucleic acid

<table>
<thead>
<tr>
<th>HIV Viral load (IU/ml)</th>
<th>10^5</th>
<th>10^4</th>
<th>5 x 10^3</th>
<th>10^3</th>
<th>5 x 10^2</th>
<th>Neg</th>
</tr>
</thead>
<tbody>
<tr>
<td>First Gen. detection</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Second Gen. detection</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Triplex dipstick detection of co-infected clinical samples from Africa

Sample ID

<table>
<thead>
<tr>
<th>Sample ID</th>
<th>04008</th>
<th>KWADO</th>
<th>32</th>
</tr>
</thead>
</table>

Dineva et al. 2005
Comparison of TaqMan vs dipstick detection of HIV, HCV & HBV in 303 African samples

<table>
<thead>
<tr>
<th></th>
<th>No.</th>
<th>Range (IU/ml)</th>
<th>TaqMan Q-PCR (%)</th>
<th>Dipstick (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV</td>
<td>36</td>
<td>$2 \times 10^2 - 2 \times 10^6$</td>
<td>97.2</td>
<td>97.2</td>
</tr>
<tr>
<td>HCV</td>
<td>34</td>
<td>$3 \times 10^2 - 2 \times 10^7$</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>HBV</td>
<td>32</td>
<td>$2 \times 10^1 - 5 \times 10^8$</td>
<td>96.9</td>
<td>96.9</td>
</tr>
</tbody>
</table>

**SPECIFICITY**

|                | 201 negatives | 98.5% | 100% |

Dineva et al. 2005

Evolution of HIV infection

- **Diagnosis**
- **Monitor treatment**
  - Treat
  - Switch

- Levels:
  - CD4+ cells
  - Antibody
  - Viral RNA

- Time
Long cycle of product development ….

Technology development
Assay development
Scale-up validation
Field trial
Clinical trial
Regulatory approval

University of Cambridge
WHO, NIH, Wellcome Trust

Diagnostics for the Real World Ltd.
Wellcome Trust, Univ of Cambridge, scientists

Diagnostics for the Real World Ltd
Sunnyvale California
### CHLAMYDIA RAPID TEST

280 documents for manufacturing and QC

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#### How we will manufacture the products?

<table>
<thead>
<tr>
<th></th>
<th>150k tests/year</th>
<th>1 million tests</th>
<th>&gt; 1 million tests</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PHASE 1</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• pilot plant</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• all components</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>in-house</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>PHASE 2</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• key reagents in-house</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• packaging, labeling</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>outsourced to ‘Hope’</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>project</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>PHASE 3</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• partnerships or</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>subsidiary (China, India or Malaysia)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• manufacture all</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>components</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

- Clinical trials
- Marketing
- Early sales

Continuum of process development and scale-up
Commercialisation

- Product differentiation by technical innovation
- Good manufacturing process (GMP)
- Apply for FDA & EU license
- 2-tier pricing
  - ‘Cost plus’ in developing countries
  - High margin in developed countries
- Sales via selected distributors or bulk purchase

What are the challenges?

- Retain trained personnel
- Distribution and commercialisation
  - corruption
  - perception in developing countries
  - licensing is not the route
- Funding to develop additional tests
  - venture is not the route
- Sustainability
  - charity is not the route
Technology development & transfer by Fred Hollows Foundation

- 23 million blind worldwide due to cataract
- Modern cataract surgery too expensive and technically complex for developing countries
  - Developed robust, compact, portable operating microscope
  - Built intra-ocular lens manufacturing sites in Eritrea and Nepal
  - Manufacturing output of 520,000 lens/yr at 3.5% of cost
  - Export to > 50 countries
  - Broad based skill training programmes

Restored sight to 1,000,000 people

2006 Goal: test & treat 1 million women for Chlamydia
Resolving trade-offs along the way

- Academia vs Company
- Social responsibility vs Profitability
- Public health vs Private wealth
- Control vs Cash injection
- Business vs Idealism

Creating and maintaining a balance between doing well and doing good