Screening Strategies: Serology/NAT screening algorithms

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NRL, Melbourne, Australia

IPFA 4rd Workshop on Plasma Quality and Supply
Hanoi, Vietnam
Established in 1985
Not-for-profit organisation that exists to support laboratories that perform testing for the diagnosis and management of human infectious disease
Contracts with the Australian Government
NRL – Major Activities

- NRL Evaluations
- NRL Quality Assurance
  - EQAS
  - QC Program
  - Specificity Monitoring
- NRL Training
- NRL Testing
- NRL Workshop
Insight into donor screening

How does NRL gain insight into donor screening?
- NRL provides QC and EQAS to the majority of donor screening laboratories
- Globally – over 1000 participants
- ‘The Region’

NRL has a leading role in ‘The Region’
- Not just provision of Quality Assurance programs
- Laboratory Strengthening
- Training (QMS, testing algorithms, QA programs)

WHO Collaborating Centre for Diagnostics and Laboratory Support for HIV and AIDS and other Blood-Borne Infections
What is ‘The Region’?

The Asia Pacific Region comprises two regions as defined by WHO:
- South-east Asia Region (SEARO)
- Western Pacific Region (WPRO)

20 countries in NRL QA programs
“The Region” – SEARO

- Bangladesh
- Bhutan
- Democratic People’s Republic of Korea
- India
- Indonesia
- Maldives
- Myanmar
- Nepal
- Sri Lanka
- Thailand
- Timor-Leste

http://www.searo.who.int/countries/en/
“The Region” – SEARO

Bangladesh
Bhutan
India
Indonesia
Maldives

Myanmar
Nepal
Sri Lanka
Thailand

http://www.searo.who.int/countries/en/
“The Region” - WPRO

http://www.wpro.who.int/countries/en/
“The Region” - WPRO

American Samoa
Australia
Brunei Darussalam
Cambodia
China
Cook Islands
Fiji
French Polynesia (France)
Guam (USA)
Hong Kong (China)
Japan
Kiribati
Lao People’s Democratic Republic

Macao (China)
Malaysia
Marshall Islands
Micronesia, Federated States of
Mongolia
Nauru
New Caledonia (France)
New Zealand
Niue
Northern Mariana Islands
Palau
Papua New Guinea

Philippines
Pitcairn Islands (UK)
Republic of Korea
Samoa
Singapore
Solomon Islands
Tokelau* (New Zealand)
Tonga
Tuvalu
Vanuatu
Viet Nam
Wallis and Futuna

http://www.wpro.who.int/countries/en/
“The Region” - WPRO

Australia

Cambodia

Macao (China)

Malaysia

Mongolia

New Zealand

Hong Kong (China)

Japan

Papua New Guinea

Philippines

Republic of Korea

Singapore

Viet Nam

http://www.wpro.who.int/countries/en/
Asian Region

- Has about one third of world population
- Disproportionate burden of disease
- Geographically disparate
  - Large land masses
  - Archipelagos
  - Small island nations
- Vast differences in economies, political stability
- 10 million units
- >3,500 blood banks in eleven countries*

*Choudhury; Asian J Transfus Sci 2011 5(2):117-120
WHO Recommendations

- Each country should have a *national policy* on blood screening
- National programme for blood screening which sets out the *strategy* for screening, with *algorithms that define the actual tests* to be used in each screening facility
- Where feasible, blood screening should be *consolidated in strategically located facilities*
Screening of all blood donations should be mandatory for

- HIV 1 and HIV 2
- HCV
- HBV
- Syphilis
WHO Recommendations

- Adequate resources, including sufficient number of qualified and trained staff be available.
- A national system for the evaluation, selection and validation of all assays.
- The minimum evaluated sensitivity and specificity levels of all assays used for blood screening should be as high as possible and preferably not less than 99.5%.
- Quality-assured screening of using serology should be in place before nucleic acid testing are considered.
Blood Screening in the Region
Blood Screening in the Region
## Examples of Blood Transfusion Services

<table>
<thead>
<tr>
<th>Country</th>
<th>No of labs</th>
<th>Centralised services</th>
<th>Type of services</th>
<th>National policies</th>
</tr>
</thead>
<tbody>
<tr>
<td>Australia</td>
<td>4</td>
<td>Yes</td>
<td>Red Cross</td>
<td>Yes</td>
</tr>
<tr>
<td>Bhutan</td>
<td>27</td>
<td>Yes</td>
<td>Hospital</td>
<td>Yes</td>
</tr>
<tr>
<td>Bangladesh</td>
<td>&gt;100</td>
<td>No</td>
<td>Private/Red crescent</td>
<td>Yes</td>
</tr>
<tr>
<td>China (mainland)</td>
<td>&gt;400</td>
<td>Partial</td>
<td>Government</td>
<td>Yes</td>
</tr>
<tr>
<td>India</td>
<td>&gt;2750</td>
<td>No</td>
<td>Provincial, hospital, private, charitable, government</td>
<td>Yes (+/-)</td>
</tr>
<tr>
<td>Indonesia</td>
<td>&gt;200</td>
<td>No</td>
<td>Red cross, hospital</td>
<td>?</td>
</tr>
<tr>
<td>Malaysia</td>
<td>14</td>
<td>Yes</td>
<td>Government</td>
<td>Yes</td>
</tr>
<tr>
<td>Myanmar</td>
<td></td>
<td>Yes</td>
<td></td>
<td>Yes</td>
</tr>
<tr>
<td>Nepal</td>
<td>~70</td>
<td>?</td>
<td>Hospital, district</td>
<td>Yes</td>
</tr>
<tr>
<td>New Zealand</td>
<td>2</td>
<td>Yes</td>
<td>Red Cross</td>
<td>Yes</td>
</tr>
<tr>
<td>Thailand</td>
<td>13</td>
<td>Yes</td>
<td>Red Cross</td>
<td>Yes</td>
</tr>
<tr>
<td>Timor-Leste</td>
<td>1 +/-</td>
<td>Yes</td>
<td>Government</td>
<td>Yes</td>
</tr>
<tr>
<td>Sri Lanka</td>
<td>~80</td>
<td>Yes</td>
<td>Hospital, private</td>
<td>Yes</td>
</tr>
</tbody>
</table>
## Serology Screening – anti-HIV

<table>
<thead>
<tr>
<th>Assay</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abbott Architect</td>
<td>100%</td>
<td>99.62%</td>
</tr>
<tr>
<td>Abbott PRISM</td>
<td>100%</td>
<td>99.98%</td>
</tr>
<tr>
<td>DiaSorin Liaison XL</td>
<td>100%</td>
<td>99.69%</td>
</tr>
<tr>
<td>DiaSorin Murex EIA</td>
<td>100%</td>
<td>99.91%</td>
</tr>
<tr>
<td>Ortho VITROS</td>
<td>100%</td>
<td>99.92%</td>
</tr>
<tr>
<td>Roche Elecsys (combi PT)</td>
<td>100%</td>
<td>99.88%</td>
</tr>
<tr>
<td>Siemens ADVIA Centaur</td>
<td>100%</td>
<td>99.74%</td>
</tr>
<tr>
<td>Siemens Enzygnost EIA</td>
<td>100%</td>
<td>99.3 – 100%</td>
</tr>
</tbody>
</table>
Serology Screening
## Serology Screening – anti-HIV

<table>
<thead>
<tr>
<th>Rapid Tests used in NRL EQAS</th>
<th>WHO Prequalified</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACON HIV 1/2 Ultra Rapid Test Device (Serum/Plasma)</td>
<td>No</td>
</tr>
<tr>
<td>Alere Determine HIV-1/2 Ag/Ab Combo Rapid Test</td>
<td>Yes</td>
</tr>
<tr>
<td>Alere Determine HIV-1/2 Rapid Test</td>
<td>Yes</td>
</tr>
<tr>
<td>Bio Focus BioTracer HIV 1/2 Rapid Card</td>
<td>No</td>
</tr>
<tr>
<td>Blue Screen HIV 1/2/O Tri-line HIV Rapid Test Device (WB/S/P)</td>
<td>No</td>
</tr>
<tr>
<td>Core Diagnostics Core HIV 1&amp;2</td>
<td>No</td>
</tr>
<tr>
<td>Green Cross GENEDIA HIV 1/2 Rapid 3.0</td>
<td>No</td>
</tr>
<tr>
<td>Human HEXAGON HIV Rapid Test</td>
<td>No</td>
</tr>
<tr>
<td>Orgenics ImmunoComb II HIV 1 &amp; 2 CombFirm</td>
<td>No</td>
</tr>
<tr>
<td>PMC FIRST RESPONSE HIV 1-2.O Rapid Card test (v.3.0)</td>
<td>No</td>
</tr>
<tr>
<td>Standard Diagnostics SD Bioline HIV-1/2 3.0 Rapid Test</td>
<td>Yes</td>
</tr>
<tr>
<td>J. Mitra HIV TRI-DOT</td>
<td>No</td>
</tr>
<tr>
<td>Wondfo One Step HIV 1/2 WB/S/P Rapid Cassette Test (2 Lines)</td>
<td>No</td>
</tr>
</tbody>
</table>
Serology Screening
# Testing Reported in NRL EQAS

<table>
<thead>
<tr>
<th>Country</th>
<th>Serology</th>
<th>Nucleic Acid Testing</th>
</tr>
</thead>
<tbody>
<tr>
<td>AUSTRALIA</td>
<td>ChLIA, EIA, Blot, PAA</td>
<td>Yes</td>
</tr>
<tr>
<td>BANGLADESH</td>
<td>ChLIA, EIA,</td>
<td>No</td>
</tr>
<tr>
<td>BHUTAN</td>
<td>EIA, PAA</td>
<td>No</td>
</tr>
<tr>
<td>CHINA (mainland)</td>
<td>ChLIA, EIA, Blot, PAA</td>
<td>Yes</td>
</tr>
<tr>
<td>INDIA</td>
<td>ChLIA, EIA, Blot, PAA, RTD</td>
<td>Partial</td>
</tr>
<tr>
<td>INDONESIA</td>
<td>ChLIA, EIA, Blot</td>
<td>Yes</td>
</tr>
<tr>
<td>JAPAN*</td>
<td>ChLIA, EIA, Blot, PAA</td>
<td></td>
</tr>
<tr>
<td>MALAYSIA</td>
<td>ChLIA, Blot, PAA</td>
<td>Yes</td>
</tr>
<tr>
<td>MONGOLIA</td>
<td>ChLIA, EIA, Blot, PAA, RTD</td>
<td>Yes (+/-)</td>
</tr>
<tr>
<td>NEPAL</td>
<td>EIA, RTD</td>
<td>No</td>
</tr>
<tr>
<td>NEW ZEALAND</td>
<td>ChLIA</td>
<td>Yes</td>
</tr>
<tr>
<td>VIETNAM</td>
<td>ChLIA, PAA, RTD</td>
<td>Yes</td>
</tr>
</tbody>
</table>
# Testing Reported in NRL EQAS

<table>
<thead>
<tr>
<th>Country</th>
<th>Serology</th>
<th>Nucleic Acid testing</th>
</tr>
</thead>
<tbody>
<tr>
<td>PR OF CHINA</td>
<td>ChLIA, EIA, Blot, PAA, RTD</td>
<td>Yes</td>
</tr>
<tr>
<td>PHILIPPINES</td>
<td>ChLIA, EIA, Blot, PAA, RTD</td>
<td></td>
</tr>
<tr>
<td>REPUBLIC OF KOREA</td>
<td>ChLIA, EIA, Blot, PAA,</td>
<td>Yes</td>
</tr>
<tr>
<td>REPUBLIC OF MALDIVES</td>
<td>ChLIA</td>
<td></td>
</tr>
<tr>
<td>REPUBLIC OF SINGAPORE</td>
<td>ChLIA, PAA</td>
<td>Yes</td>
</tr>
<tr>
<td>SRI LANKA</td>
<td>ChLIA, EIA, Blot</td>
<td></td>
</tr>
<tr>
<td>THAILAND</td>
<td>ChLIA, EIA, Blot, RTD</td>
<td>Yes</td>
</tr>
<tr>
<td>UNION OF MYANMAR</td>
<td>ChLIA, EIA, RTD</td>
<td></td>
</tr>
<tr>
<td>VIETNAM</td>
<td>ChLIA, EIA, Blot, PAA, RTD</td>
<td>Yes</td>
</tr>
</tbody>
</table>
Window Period for HIV and HCV

Testing Strategies in Asia Pacific

- Diverse
- Dangerous
- Difficult
- Developing
The Himalayan

In India, Blood Transfusions Have Caused Thousands of
HIV Infections

The Himalayan Times > Nepal > Blood-borne Transfusion happening without testing blood for deadly diseases

12,000 Chinese blood plasma treatments contaminated
with HIV

Traces found in both used for immune deficiencies caused by illnesses including
HIV, Hepatitis A, B, C, and patients advised to report their stocks of the treatment and closely monitor
patients already treated.
Serology

More common – RTD and PAA used for screening
- Multiple instances in HIV, HCV and HBsAg RTDs

Anti-HIV only tests (3rd Gen)
- Many participants use these with HIV combo as dual screen
- Some performed anti-HIV only and risk not detecting early infection
- Many laboratories using RTDs in this situation

Evidence EQAS is commonly used but QC is not!!!
QC in the Region

**Pros**
- Vast number of results reported
  - Both serology and NAT
- Overall laboratory performance – Good
- Data generated supports quality improvements
- Often the only positive samples seen frequently

**Cons**
- Not enough participants for IVDs in resource limited labs
- Reports of in-house QC use – common
- Commercial QC cost-prohibitive – a major issue
Two types of QC provided

- Single viral NAT QC for Grifols
  - QConnect HIV RNA
  - QConnect HCV RNA
  - QConnect HBV DNA

- Multiplex NAT QC for Roche
  - QConnect TriScreen
HIV RNA

- Peer Group Data by Lab
- Grifols Ultrio Elite on Panther
HIV RNA

- Roche cobas MPX v2.0
- Data by reagent lot
- Typical Lot Variation
Lab 275 - HCV RNA

- Peer Group Data by Lab
- Roche cobas MPX
Lab 275 – HCV RNA

- Roche cobas MPX
- Data by Instrument
- Spikes caused by pooling of left over QC samples
HCV RNA

- Roche cobas MPX v2.0
- Peer Group Data
- Unusually high Ct values for Lab 365
Lab 365

- Roche cobas MPX v2.0
- Data by reagent lot
- Spikes in Ct values
Lab 365

Roche cobas MPX v2.0

Data by Instrument

Spikes in Ct values – switch in instruments reported?
TriScreen Study

Multi-site, multi-country study on suitability of TriScreen to monitor Roche cobas MPX and cobas TaqScreen MPX v2.0
Results

Generally, HCV RNA MPX v2.0 testing demonstrated greater imprecision.

No detectable differences in imprecision between participants testing HCV RNA on cMPX on the c6800/c8800 systems (past participants).

QC was sensitive enough to see different protocols were being used for routine and cadaveric testing.
Results – Investigation into unexpected results
Mini-Pool and QC Testing

- Use of QC for cadaveric protocol demonstrated dilution possible
- Many donor centres using Roche cobas MPX v2.0 test in mini-pools of 6

Question -

*Can NRL QConnect QCs be used in mini-pool format?*
HIV RNA

Roche cobas MPX v2.0 using minipool protocol
**HCV RNA**

- Roche cobas MPX v2.0
- HCV RNA “0” Ct values
- Few for two labs
- More for lab in red box

| Laboratory | 40.00 | 38.00 | 36.00 | 34.00 | 32.00 | 30.00 | 28.00 | 26.00 | 24.00 | 22.00 | 20.00 | 18.00 | 16.00 | 14.00 | 12.00 | 10.00 | 8.00 | 6.00 | 4.00 | 2.00 | 0.00 | -2.00 |
|------------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| Analyte: HCV RNA | 40.00 | 38.00 | 36.00 | 34.00 | 32.00 | 30.00 | 28.00 | 26.00 | 24.00 | 22.00 | 20.00 | 18.00 | 16.00 | 14.00 | 12.00 | 10.00 | 8.00 | 6.00 | 4.00 | 2.00 | 0.00 | -2.00 |
| EQC List Number: 140817D1; | 40.00 | 38.00 | 36.00 | 34.00 | 32.00 | 30.00 | 28.00 | 26.00 | 24.00 | 22.00 | 20.00 | 18.00 | 16.00 | 14.00 | 12.00 | 10.00 | 8.00 | 6.00 | 4.00 | 2.00 | 0.00 | -2.00 |
| Grouped by Laboratory: | 40.00 | 38.00 | 36.00 | 34.00 | 32.00 | 30.00 | 28.00 | 26.00 | 24.00 | 22.00 | 20.00 | 18.00 | 16.00 | 14.00 | 12.00 | 10.00 | 8.00 | 6.00 | 4.00 | 2.00 | 0.00 | -2.00 |
| Assays: Roche COBAS TaqScreen MPX v2.0 (multiplex) | 40.00 | 38.00 | 36.00 | 34.00 | 32.00 | 30.00 | 28.00 | 26.00 | 24.00 | 22.00 | 20.00 | 18.00 | 16.00 | 14.00 | 12.00 | 10.00 | 8.00 | 6.00 | 4.00 | 2.00 | 0.00 | -2.00 |

All Labs - All Date: Mean=38.57; SD=4.03; 3SD=33.99
Lab 481

- Roche cobas MPX v2.0
- HCV RNA “0” Ct values
- Samples stored incorrectly on receipt
Conclusion

- Dilution possible but needs more work
- Limit of detection differences between MPX on 6800 and MPX v2.0 on s201 may play a role
- Use of QC for mini-pool testing highlighted variation was detectable even at high Ct values
General conclusions

NAT screening – the ‘Big Two’
- Grifols and Roche
- Some ‘local’ manufacturers successful

Serology
- Diverse technologies and manufacturers
- Assay selection driven by cost, availability and practicality
- Higher error rates seen in manual testing
Where to From Here?

- Assay evaluation and selection
- Assay procurement
- Validated testing algorithms
- Quality management systems
- Quality assurance / quality control
- Training and competency
Thank-you!

Acknowledgements

- NRL EQAS team
- NRL QC team
- All the program participants