Update on the development of Rapid Diagnostic Tests for meningitis

Olivier Ronveaux, WHO Geneva

London, MRF conference, November 2019
Meningitis in vitro diagnostics – three needs

1. **Global.** Key question on sick patient: is antibiotics/referral needed (yes/no). Point of care. Bacterial vs non bacterial infections

2. **Global.** Identify the pathogen – patient with meningoencephalitis syndrome - to determine the appropriate treatment, switch treatment or terminate inappropriate treatment.

3. **Meningitis belt.** Need to identify causative organism (Nm serogroup) rapidly at peripheral level for outbreak detection

[https://www.who.int/emergencies/diseases/meningitis/en/](https://www.who.int/emergencies/diseases/meningitis/en/)
1. **Global.** Key question on sick patient: is antibiotics/referral needed (yes/no). Point of care. Bacterial vs non bacterial infections

   - **Roadmap 2030:** by 2026, quality assured, affordable and accessible rapid diagnostic assay developed to rapidly detect invasive bacterial vs viral infection to support immediate medical decision-making at point of care
Challenge

Ideal biomarker/host marker not identified yet

Test profile

- No overlapping value between bacterial and viral
- Early detectable
- Blood or CSF without preparation
- Does not require highly trained staff
- High specificity – high negative predictive value
- Short time to result < 10 minutes
Host factors and biomarkers

- C-Reactive Protein
- Procalcitonin
- Lipocalin 2 (LCN2)
- Heparin Binding Protein
- Serum amyloid A Protein
- Cytokines / chemokines
Early detecting of LCN2 in CSF

<table>
<thead>
<tr>
<th></th>
<th>Confirmed acute bacterial meningitis n=90</th>
<th>Confirmed acute viral meningitis n=44</th>
</tr>
</thead>
<tbody>
<tr>
<td>CSF LCN2, pg/mL</td>
<td>127</td>
<td>2.4</td>
</tr>
<tr>
<td></td>
<td>108-146</td>
<td>0-6.2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>
**TRanscripts Identifying Meningitis - TRIM test**

- **Simple PCR assay**
  - Developed from transcript data (n > 500 samples)
  - lyophilised multiplex assay

- **Host transcripts in blood**
  - Distinguish bacterial meningitis from clinical mimics

  - **Sensitivity = 100%**
  - **Specificity = 90%**

  results of prototype assay
• Trim Assay being validated in a multi-site study in UK and Europe
  – ensure it works in hospital setting
  – recruiting > 696 patients, started in April 2019
• Systems component ready for use on standard hospital equipment
• Baseline data on TRIM data accuracy in India, Indonesia, Malawi and Brasil*

• Funded by MRC and industry

Future Benefits
• Compatible with point-of-care devices.
• Compatible for syndromic detection of sepsis and other bacterial syndromes
• Potential for treatment monitoring
• Potential to integrate with pathogen specific PCR testing

More information on the TRIM study
PI - Mike Griffiths
griffmj@liv.ac.uk

* supported by additional Newton and NIHR funded studies
2. **Global.** Identify the pathogen – patient with meningoencephalitis syndrome - to determine the appropriate treatment, switch treatment or terminate inappropriate treatment.

   – **Roadmap 2030:** by 2026, quality assured, affordable and accessible multiplex diagnostic test available to identify and distinguish the main pathogens responsible for meningitis
What do we want to have

• Cheap
• Reliable (high sensitivity and specificity)
• Capable of testing more and more pathogens
• Desk top machine that can be set up almost anywhere
  – Compact device, battery operated
  – Peripheral level
• Multi-pathogen detection in a single reaction or run
Multiplex PCR are already used...

- Commercially available: Xpert, Biofire, TAC etc
- In-house

Cape Town, 2017
- 6 pathogens
- Good performance compared to culture
- “potential to limit unnecessary therapy”

RESEARCH ARTICLE
Diagnostic accuracy of two multiplex real-time polymerase chain reaction assays for the diagnosis of meningitis in children in a resource-limited setting

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* colleen.bamford@uct.ac.za
Devices are available...

**Portable real-time PCR platforms**

<table>
<thead>
<tr>
<th>Platform</th>
<th>Features</th>
<th>Run Time</th>
<th>Power</th>
<th>Estimated Instrument Cost</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Q-POC (UK)</strong></td>
<td>Cassette Multiplex (up to 40 targets)</td>
<td>10-30 minutes</td>
<td>Battery powered</td>
<td>$3000</td>
</tr>
<tr>
<td><strong>Anitoa Maverick compact qPCR system (US)</strong></td>
<td>4-8 wells Multiplex (up to 4 targets)</td>
<td>~30 minutes</td>
<td>10V, battery backup option</td>
<td>$3500-$6000</td>
</tr>
<tr>
<td><strong>Coyote Mini8 Plus Real-Time PCR System (Germany)</strong></td>
<td>8 wells Multiplex (up to 2 targets)</td>
<td>&lt;2 hours</td>
<td>12V Battery pack</td>
<td>$6000-$8000</td>
</tr>
<tr>
<td><strong>Bio molecular Systems Mi qPCR (Australia)</strong></td>
<td>48 wells Multiplex (up to 4 targets)</td>
<td>~25 minutes</td>
<td>100-240V</td>
<td>$15000</td>
</tr>
<tr>
<td><strong>Q160 Mini Real-Time PCR System (China)</strong></td>
<td>16 wells Multiplex (up to 2 targets)</td>
<td>1-2 hours or less</td>
<td>85-265V</td>
<td>$4600</td>
</tr>
<tr>
<td><strong>Handheld real-time PCR device (prototype)</strong></td>
<td>4 wells Multiplex (up to 2 targets)</td>
<td>~35 minutes</td>
<td>?</td>
<td>?</td>
</tr>
</tbody>
</table>

Targets could be genes specific for pathogen and serotype/serogroup or associated with antibiotics

Table courtesy of Dr Xin WANG, US CDC
Meningitis multiplex test - developments

- WHO Expert meeting, September 2019 – Consensus on test specifications
  - Hospital, near-patient hospital laboratory which supports lumbar puncture and centrifugation, may support molecular testing
  - List A, universal, 13 pathogens, global level (bacterial, viral, parasite, fungi)
  - List B, Ideal, Regional specificities, 13 pathogens
- Target Product Profile to be published in December 2019
- Next steps:
  - Market review: identification of manufacturers in the pipeline
  - Define market size (demand): what is the global need
  - Access plan: Identification of barriers and incentives for assay development, production and accessibility
  - Design of a costing model: direct purchasing model versus alternate ones (need creative ideas to provide the machine/test/maintenance/reagents...access for LMICs)
Meningitis in vitro diagnostics – Use case 3

3. Meningitis belt. Need to identify causative organism (Nm serogroup) rapidly at peripheral level for outbreak detection

– Roadmap 2030: Adopt, integrate and implement minimum standards for surveillance of the main meningitis pathogens at country level on epidemiology, laboratory capacity (including the use of up to date diagnostic and AMR tests), and data management (SG 10)
Lateral flow immunochromatography

Two RDTs from Biospeedia

NmA, NmW, NmC, NmY, NmX

Thermostable
MeningoSpeed - PneumoSpeed

• Good performances under laboratory conditions: RDT vs PCR
  – Institut Pasteur Paris and Burkina Faso, CIV, CAR, Niger, Togo, Morocco
  – MeningoSpeed: sensitivity 95.6%, specificity 93.8% (545 samples)

• WHO: two levels of evaluation
  – Product suitability for procurement by WHO
    • Review of documentation, manufacturer practices, etc
  – External field validation
    • Burkina Faso and Niger, 2018-2019
Field validation study, Burkina Faso and Niger

- RDT at health centre level
  - Real situation. Districts in Alert -&gt; staff immediately trained
- National Reference Laboratory (NRL): repeat RDT and PCR as gold standard
- Semi structured interviews and questionnaires
- Concordance: control photography by blinded reviewer
- Ethical approval: WHO and two national ethical committees
327 patients included: Niger (246) and Burkina Faso (81)

<table>
<thead>
<tr>
<th>CSF positive - RDT health Centre</th>
<th>Neisseria (Nm)</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>106</td>
<td>32%</td>
<td></td>
</tr>
<tr>
<td>NmA</td>
<td>9</td>
<td>3%</td>
<td></td>
</tr>
<tr>
<td>NmC</td>
<td>56</td>
<td>17%</td>
<td></td>
</tr>
<tr>
<td>NmW</td>
<td>2</td>
<td>1%</td>
<td></td>
</tr>
<tr>
<td>NmX</td>
<td>40</td>
<td>12%</td>
<td></td>
</tr>
<tr>
<td>NmY</td>
<td>1</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td><em>S. pneumoniae</em></td>
<td>28</td>
<td>9%</td>
<td></td>
</tr>
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</table>
## Final results (October 2019)

<table>
<thead>
<tr>
<th>Health center</th>
<th>N</th>
<th>Sensitivity (%)</th>
<th>CI 95%</th>
<th>Specificity (%)</th>
<th>CI 95%</th>
<th>PPV (%)</th>
<th>NPV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>All Nm</td>
<td>327</td>
<td>95.3</td>
<td>88</td>
<td>99</td>
<td>90</td>
<td>86</td>
<td>94</td>
</tr>
<tr>
<td>Sp</td>
<td>334</td>
<td>92.9</td>
<td>77</td>
<td>99</td>
<td>99</td>
<td>97</td>
<td>100</td>
</tr>
<tr>
<td>All Nm</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>77</td>
<td>98</td>
</tr>
<tr>
<td>Sp</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>87</td>
<td>99</td>
</tr>
</tbody>
</table>
### NmA? Two by two tables

#### Health center

<table>
<thead>
<tr>
<th></th>
<th>PCR</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>+</td>
<td>0</td>
<td>9</td>
</tr>
<tr>
<td>+</td>
<td>0</td>
<td>329</td>
</tr>
</tbody>
</table>

2 out of these 9 tests positive also found positive by blind reviewers

#### NRL

<table>
<thead>
<tr>
<th></th>
<th>PCR</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>+</td>
<td>0</td>
<td>6</td>
</tr>
<tr>
<td>+</td>
<td>0</td>
<td>119</td>
</tr>
</tbody>
</table>

NmA migration line too close to the border of the device

Manufacturer indicated that this has been fixed
Good training is necessary
Study limitations

- Field conditions challenging (security)
  - PCR confirmation: hampered in Burkina Faso (strike)
  - RDT repeat at the NRL challenged in Niger
- Nm distribution: mainly C and X serogroups
  - Small numbers with other serogroups
Results suggest

• Good performance of the RDT overall
  – in particular for NmC and NmX (Se: 93% and 91%, respectively)
• Interpretation issues, specially associated with NmA
  – all false negatives were on the AW cassette
• Conditions for use need to be carefully implemented
Conclusions

• Quick wins welcome!
  – Lateral flow has a place now in the meningitis belt
  – Finalize the RDT suitability for procurement
  – Deployment to be discussed 29 November
  – Exploration outside the belt

• Ambitious agenda (use case 1 and 2)
  – Development money to be identified

• Fast moving context
  – Support of all stakeholders needed
Thank you
Group B Strep – roadmap 2030

• By 2026, a quality assured, affordable and accessible diagnostic assay available to identify (i) maternal GBS carriage and (ii) invasive GBS disease
Concordance: high agreement between blind reviewer and RDT reading at the Health centre

<table>
<thead>
<tr>
<th>RDT at the health centre</th>
<th>Total</th>
<th>Percentage of agreement (%)</th>
<th>Coefficient Kappa</th>
</tr>
</thead>
<tbody>
<tr>
<td>All Nm</td>
<td>65</td>
<td>82</td>
<td>61</td>
</tr>
<tr>
<td>NmA</td>
<td>73</td>
<td>90</td>
<td>32</td>
</tr>
<tr>
<td>NmC</td>
<td>75</td>
<td>96</td>
<td>39</td>
</tr>
<tr>
<td>NmW</td>
<td>73</td>
<td>95</td>
<td>32</td>
</tr>
<tr>
<td>NmX</td>
<td>70</td>
<td>94</td>
<td>83</td>
</tr>
<tr>
<td>NmY</td>
<td>71</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Sp</td>
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<td>97</td>
<td>82</td>
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