# 13th Global Measles and Rubella Laboratory Network Meeting

*Salle B, WHO/HQ Geneva, 29 June – 1 July 2015*

## Agenda

### Day 1: Monday 29 June

**Chairs:** Claude Muller (AM),
Annette Mankertz (PM)

<table>
<thead>
<tr>
<th>Time</th>
<th>Session 1 – Opening</th>
<th>Location</th>
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<tbody>
<tr>
<td>08:30</td>
<td>Registration</td>
<td>Salle B</td>
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<tr>
<td>09:00</td>
<td>Welcome and opening remarks</td>
<td>Michel Zaffran, Mick Mulders</td>
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<td></td>
<td>Introduction, meeting deliverables and recommendations</td>
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<td></td>
<td>from 12th meeting</td>
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<td></td>
<td>Administrative announcements</td>
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<td></td>
<td>Declaration of Interests</td>
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<td></td>
<td>Selection of chairs</td>
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<table>
<thead>
<tr>
<th>Time</th>
<th>Session 2 – Global updates Measles and Rubella Elimination</th>
<th>Location</th>
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<tbody>
<tr>
<td>09:20</td>
<td>01. Global Measles and Rubella programme – update</td>
<td>Robert Perry</td>
</tr>
<tr>
<td>09:40</td>
<td>02. GMRLN – update</td>
<td>Mick Mulders</td>
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<tr>
<td>10:00</td>
<td>03. Enhancing measles rubella surveillance</td>
<td>Marta Gacic-Dobo</td>
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<tr>
<td>10:30</td>
<td><em>Break</em></td>
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<table>
<thead>
<tr>
<th>Time</th>
<th>Session 3 – Region and selected country updates</th>
<th>Location</th>
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<tbody>
<tr>
<td>11:00</td>
<td>04. Eastern Mediterranean Region</td>
<td>Hinda Ahmed</td>
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<td></td>
<td>05. RRL/NL activities Institut Pasteur Tunis</td>
<td>Henda Triki</td>
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<td></td>
<td>06. RRL/NL activities CPHL Muscat</td>
<td>Said Al Baqlani, Sohail Zaidi</td>
</tr>
<tr>
<td>Time</td>
<td>Session</td>
<td>Speaker(s)</td>
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<tr>
<td>12:00</td>
<td>Western Pacific Region incl. update on the current outbreak in Mongolia</td>
<td>Yan Zhang</td>
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<td></td>
<td>GSL/RRL/NL activities – NIID Tokyo</td>
<td>Makoto Takeda</td>
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<td>RRL/NL activities – CCDC Beijing</td>
<td>Songtao Xu</td>
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<td>RRL/NL activities – DH Hong Kong</td>
<td>Janice Lo</td>
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<tr>
<td>13:00</td>
<td>Lunch</td>
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<td>14:00</td>
<td>African Region</td>
<td>Annick Dosseh/Charles Byabamazima</td>
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<td>RRL/NL activities – NICD Johannesburg</td>
<td>Sheilagh Smit</td>
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<td>RRL/NL activities – Institut Pasteur Abidjan</td>
<td>Hervé Kadjo</td>
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<td>RRL/NL activities – UVRI Entebbe</td>
<td>Barnabas Bakamutumaho</td>
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<td>NL and SNL activities – EHNRI Addis Ababa</td>
<td>Berhane Beyene Mentaye</td>
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<td>15:00</td>
<td>South East Asian Region</td>
<td>Sirima Pattamadilok</td>
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<td>RRL/NL/SNL activities NIH Bangkok</td>
<td>Atchariya Lukebua</td>
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<td></td>
<td>Building measles rubella laboratory network in India</td>
<td>Lucky Sangal</td>
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<td>Strengthening laboratory capacity Indonesia</td>
<td>Rusipah</td>
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<tr>
<td>16:00</td>
<td>Break</td>
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<tr>
<td>16:30</td>
<td>Region of the Americas</td>
<td>Not available</td>
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<td></td>
<td>RRL/NL activities – FioCruz Rio De Janeiro</td>
<td>Marilda Siqueira</td>
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<td>GSL/RRL/NL measles activities – CDC Atlanta</td>
<td>Paul Rota</td>
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<td></td>
<td>GSL/RRL/NL rubella activities – CDC Atlanta Project to Identify Laboratory Challenges for Rubella and CRS Surveillance</td>
<td>Joe Icenogle</td>
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<td>RRL/NL activities – PHAC Winnipeg</td>
<td>Joanne Hiebert</td>
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<td>SubRRL/NL activities – CARPHA Port of Spain</td>
<td>Pablo Martinez de Salazar</td>
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<tr>
<td>18:00</td>
<td>Adjourn followed by reception</td>
<td>Terrace</td>
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## Session 3 – Region and selected country updates (continued)

<table>
<thead>
<tr>
<th>Time</th>
<th>Session</th>
<th>Description</th>
<th>Speaker</th>
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<tbody>
<tr>
<td>08:30</td>
<td>27</td>
<td>European Region</td>
<td>Myriam Ben Mamou</td>
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<td></td>
<td>28</td>
<td>GSL/RRL/NL activities – PHE London</td>
<td>Kevin Brown</td>
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<td></td>
<td>29</td>
<td>Update on RRL Moscow/NIS subregion laboratory network activities 2014 – 2015</td>
<td>Sergey Shulga</td>
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<td></td>
<td>30</td>
<td>RRL/NL activities – RKI Berlin</td>
<td>Annette Mankertz</td>
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<td>31</td>
<td>RRL/NL activities – LIH Luxembourg</td>
<td>Judith Hübschen</td>
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## Session 4 – EQA

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<tr>
<th>Time</th>
<th>Session</th>
<th>Description</th>
<th>Speaker</th>
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<tbody>
<tr>
<td>09:45</td>
<td>32</td>
<td>Measles and Rubella IgM Proficiency Testing – Panel 01404 and new scoring algorithm for next panel 01503</td>
<td>Vicki Stambos</td>
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<td></td>
<td></td>
<td>Discussion</td>
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<tr>
<td>10:30</td>
<td>Break</td>
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<tr>
<td>11:00</td>
<td>33</td>
<td>WHO/CDC molecular EQA program, results of the 2014 survey and ways forward</td>
<td>Paul Rota</td>
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<td></td>
<td>34</td>
<td>Molecular EQA experience – China</td>
<td>Zhen Zhu</td>
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<tr>
<td></td>
<td>35</td>
<td>Molecular EQA experience – Japan</td>
<td>Katsuhiro Komase</td>
</tr>
<tr>
<td></td>
<td>36</td>
<td>New collaboration between WHO and INSTAND e.V. on molecular EQA for measles and rubella</td>
<td>Oliver Donoso Mantke</td>
</tr>
<tr>
<td>12:00</td>
<td>37</td>
<td>Report from Rubella IgG standardization working group</td>
<td>Christelle Vauloup</td>
</tr>
<tr>
<td>12:30</td>
<td>Lunch</td>
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### Session 5: Working group reports

<table>
<thead>
<tr>
<th>Time</th>
<th>Topic</th>
<th>Presenter(s)</th>
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<tbody>
<tr>
<td>13:30</td>
<td>Report from the working group on the 3rd revision of the WHO Laboratory Manual for the Diagnosis of Measles and Rubella – Status update</td>
<td>Jenny Rota</td>
</tr>
<tr>
<td>13:50</td>
<td>Global serosurvey guidelines</td>
<td>Ray Sanders</td>
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<tr>
<td></td>
<td>Point Of Care Testing for measles IgM and IgG – project update</td>
<td>David Featherstone</td>
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<td></td>
<td>Multiplex high throughput serology</td>
<td>Fiona van der Klis</td>
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<td></td>
<td></td>
<td>Paul Rota</td>
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<tr>
<td></td>
<td></td>
<td>Rob van Binnendijk</td>
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<td></td>
<td>IgM and IgG kit comparison study</td>
<td>Kevin Brown</td>
</tr>
<tr>
<td>15:00</td>
<td>Report from the working group on Extended and Whole Genome and Next Generation Sequencing (WGEWGNGS)</td>
<td>Alberto Severini on behalf of WG</td>
</tr>
<tr>
<td></td>
<td>Measles whole genome and next generation sequencing – PHE experience</td>
<td>Richard Myers</td>
</tr>
<tr>
<td>15:35</td>
<td>Introduction to break out groups</td>
<td>Mick Mulders</td>
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<tr>
<td></td>
<td><em>This session is organized for representatives of the laboratories belonging to the same Region to meet with their Laboratory Coordinator. The goal of this session is to discuss Region-specific laboratory performance and challenges (including budgetary), coordinate and plan activities for 2015 and 2016, including LabNet meetings, workshops and onsite visits to network laboratories in the Region, as well as strategies on how to improve performance.</em></td>
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<td></td>
<td></td>
<td><em>Regional Laboratory Coordinators to develop agenda and share before start of meeting</em></td>
</tr>
<tr>
<td>15:40</td>
<td>Break</td>
<td></td>
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<tr>
<td>16:00</td>
<td>Breakout groups</td>
<td>Group AFRO Salle B</td>
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<td>Group EMRO M209</td>
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<td>Group AMRO/PAHO Salle B</td>
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<td>Group EURO M405</td>
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<td>Group SEARO M120</td>
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<td>Group WPRO L243</td>
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<tr>
<td>18:00</td>
<td>Adjourn</td>
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### Day 3: Wednesday 01 July

**Chair:** Kevin Brown and Paul Rota

<table>
<thead>
<tr>
<th>Time</th>
<th>Session</th>
<th>Presenter(s)</th>
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<tbody>
<tr>
<td>08:30</td>
<td>Feedback from the breakout groups</td>
<td>Regional laboratory coordinators</td>
</tr>
<tr>
<td>09:45</td>
<td>Summary of workshop “Evidence needed to verify elimination of measles and rubella drawing on experience from Regional Verification Commissions”</td>
<td>Alya Dabbagh</td>
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<tr>
<td>10:00</td>
<td>Break</td>
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**Session 6: Molecular analysis**

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<thead>
<tr>
<th>Time</th>
<th>Topic</th>
<th>Presenter(s)</th>
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<tbody>
<tr>
<td>10:30</td>
<td>47. Measles genotyping – “Verification Quality Virologic Surveillance “Improving analysis and reporting of virologic surveillance data for national verification reports</td>
<td>Katsuhiro Komase</td>
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<td>48. Report from Japan (10 min max)</td>
<td>Janice Lo</td>
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<td></td>
<td>49. Report from Hong Kong (10 min max)</td>
<td>Sergey Shulga</td>
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<td>50. Simultaneous transmission of multiple measles virus lineages, NIS subregion 2013 – 2015 (10 min max)</td>
<td>Patch Incomserb</td>
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<td>51. SEAR measles genotype update and verification quality virologic surveillance of Thailand (10 min max)</td>
<td>Vicki Stambos</td>
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<td>52. Measles Vaccine-specific real-time PCR</td>
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<tr>
<td>11:30</td>
<td>53. Rubella genotyping</td>
<td>Moderator: Joe Icenogle</td>
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<tr>
<td></td>
<td>54. Rubella RT-PCR and Genotyping from Sera</td>
<td>Janice Lo</td>
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<td>55. Challenges in obtaining rubella genotypes in the EUR</td>
<td>Myriam Ben Mamou</td>
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<td></td>
<td>56. Genotypes from long term infections</td>
<td>Joe Icenogle</td>
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<tr>
<td></td>
<td>57. SEAR rubella genotype update</td>
<td>Patcha Incomserb</td>
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<tr>
<td>12:30</td>
<td>58. Status report MeaNS and RubeNS with meta-analysis data</td>
<td>Richard Myers</td>
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<tr>
<td>13:00</td>
<td>Lunch</td>
<td></td>
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<tr>
<td>14:00</td>
<td>General discussion - development of recommendations</td>
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<tr>
<td>15:30</td>
<td>Adjourn (coffee available)</td>
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1\textsuperscript{ST} MEETING OF THE WORKING GROUP ON EXTENDED, WHOLE GENOME AND NEXT GENERATION SEQUENCING


<table>
<thead>
<tr>
<th>16:00</th>
<th>Agenda</th>
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<tbody>
<tr>
<td></td>
<td>1. Welcome and introduction</td>
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<tr>
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<td>2. Summary of the working group activities to date</td>
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<td>3. Last call on ToR</td>
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<td>4. Catalogue of sequences, culture and methodologies</td>
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<td></td>
<td>a) What we have so far</td>
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<td></td>
<td>b) How we populate it</td>
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<td>c) Issues of sharing and publishing</td>
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<td>5. MeaNS; What it can do for measles WGS (Richard Myers)</td>
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<td>6. Let’s start a discussion about quality control</td>
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<td>7. Other issues mentioned in our teleconferences</td>
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<tr>
<td></td>
<td>a) Sequencing services and training for other laboratory of the MR LabNet</td>
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<td>b) Recommendations to the LabNet</td>
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<td>c) Publications</td>
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<td>d) Funding</td>
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<td>8. Planning for a possible 1-2 full day group meeting.</td>
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| Alberto Severini |
| Joanne Hiebert |
| Joe Icenogle |
| Claude Muller |
| Judith Hübschen |
| Kevin Brown |
| Ana Penedos (remote) |
| Richard Myers |
| Paul Rota |
| Bettina Bankamp (remote?) |
| Myriam Ben Mamou |
| Mick Mulders |

17:30 Adjourn
1st MEETING OF THE WORKING GROUP ON THE 3rd REVISION OF THE WHO MANUAL FOR THE LABORATORY DIAGNOSIS OF MEASLES AND RUBELLA


Session HC2 – Working group on the 3rd Revision of the WHO Manual for the Laboratory Diagnosis of Measles and Rubella
Chair: Marilda Siqueira; Minutes: Jenny Rota (All participants from GMRLN13 are welcome to join)

<table>
<thead>
<tr>
<th>Time</th>
<th>Agenda</th>
<th>Participants</th>
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| 08:30 | **Agenda**                                                                                                                                                                                               | Marilda Siqueira
|       | 1. Review comments and suggestions from labnet participants after presentation                                                                                                                                  | Jenny Rota |
|       | 2. Summary of first objectives for new edition of manual  
Q1. Do we have a reasonably complete Table of Contents?  
Q2. If not, what remains to be added or discussed?  
3. Next steps  
   a. Identify the chapters and annexes that  
      i. Require minor updating  
      ii. Have been greatly expanded in breadth of information  
      iii. Are new chapters and will require the most work  
   b. With the above in mind, assign responsibility for each chapter or sections, as appropriate, to WG member or members  
      i. Drafts of chapters or subsections  
      ii. Drafts of annexes and/or worksheets  
   c. Set deadline for first draft of chapters and protocols | Kevin Brown  
|       |                                                                                                                                   | David Brown (remote?)  
|       |                                                                                                                                                                                                  | Joe Icenogle  
|       |                                                                                                                                                                                                  | David Featherstone  
|       |                                                                                                                                                                                                  | Paul Rota  
|       |                                                                                                                                                                                                  | Mick Mulders |
| 10:30 | Adjourn (coffee break for those wanting to attend HC3)                                                                                                                                                |              |
# 2nd Joint MeaNS/RubeNS Steering Committee Meeting

**Salle B, WHO/HQ Geneva, 2 July 2015**

## Session HC3 – MeaNS/RubeNS Steering Committee

*Chair: Kevin Brown/ Joe Icenogle; Notes: TBD (All participants from GMRLN13 are welcome to join)*

## Agenda

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<th>Session</th>
<th>Topics</th>
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<tr>
<td>11:00</td>
<td></td>
<td>1. Roll call and Introductions</td>
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<td>2. Apologies</td>
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<td>3. Meeting of last meeting previously circulated</td>
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<td>4. Matters and actions arising: (not on the agenda)</td>
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<td>5. Major items for discussion</td>
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<td>a. Changes to MeaNS/RubeNS</td>
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<td></td>
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<td>i. New tools/updates implemented since last meeting (RM)</td>
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<td>ii. Update on server (RM)</td>
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<td>b. Handling GenBank numbers (JI)</td>
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<td>c. Handling vaccine strains (RM)</td>
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<td>d. WER report (PR)</td>
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<td>e. Outcome/implications of customer survey (KB/RM)</td>
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<td>f. Human resource issues (MM)</td>
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<td>g. Publication of MeaNS/RubeNS data without consent; How to handle? (MM)</td>
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<td>h. Requests for new implementations (All)</td>
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<td>13:00</td>
<td>Adjourn</td>
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SESSION 1 – OPENING

Michel Zaffran opened the meeting, welcoming the participants to the meeting. He commented about the relevance of the recent meeting, WORKSHOP ON MEASLES AND RUBELLA ELIMINATION VERIFICATION PROCESS, held on 26 June 2015 in Sitges, Spain, for the laboratory network. During the verification workshop in Spain, much emphasis was placed on the importance of the measles and rubella laboratory network for verification. In order to support the ongoing efforts of immunization programs and goals for elimination, high quality data and assurance of quality testing are critical. As new technology provides new opportunities, the laboratory needs to determine the best practices to guide and strengthen the network. The lab network must review the current status of the network and address future challenges for laboratory capacity and the verification of elimination.

The Global Laboratory Coordinator, Mick Mulders, presented the objectives and deliverables:

MEETING OBJECTIVES

1. To review and discuss the current status and management of the Global Measles and Rubella Laboratory Network (GMRLN) in order to develop and strengthen the technical capacity and structure of the network
2. To provide WHO staff and Measles and Rubella LabNet representatives with technical updates on laboratory aspects of measles and rubella surveillance and quality assurance
3. To determine how best to meet future challenges for the GMRLN

MEETING DELIVERABLES

To develop technical recommendations and a plan of action for further development and strengthening of the GMRLN for 2015-2016 and beyond

The meeting agenda was outlined and a brief summary of the recommendations from the 12th Global Measles and Rubella Laboratory Network 2013 was presented. The main meeting consisted of 6 sessions; an additional 3 sessions provided opportunities for discussion and planning among working groups or steering committees. For the core meeting, participants in attendance included 5 of the 6 regional laboratory coordinators, representatives of the global specialized/regional reference laboratories and selected national and sub-national laboratories, and two consultants. The laboratory coordinator from the AMR (PAHO) was unable to attend or participate remotely due to a concurrent meeting of the PAHO Technical Advisory Group on Vaccine-Preventable Diseases.
A summary of the presentations are included below. All presentations are available in full on the following link:

https://www.dropbox.com/sh/7qduoxk89634vx0/AACZntfSIcYnSvE9D-L0-OOa/Presentations?dl=0

SESSION 2 – GLOBAL UPDATES MEASLES AND RUBELLA ELIMINATION

GLOBAL MEASLES AND RUBELLA PROGRAMME — UPDATE

was given by Robert Perry, WHO, HQ. He reviewed the progress that has been made by setting global targets for reduction of measles mortality and cases. All 6 WHO regions have established target dates for measles elimination and 4 include dates for rubella elimination. However, one of the 2015 targets, ≥90% MCV1 coverage at the national level, continues to be a challenge, particularly in countries in the African, Eastern Mediterranean, and SE Asian regions. There is an urgent need to fill the gaps in MCV1 coverage. However, a second dose is needed for measles control and elimination, either through campaigns or routine systems. Considerable differences exist in MCV2 coverage across the regions and many countries in Africa have not yet introduced MCV2. In 2014, 28 countries conducted 43 vaccination campaigns with M or MR vaccine, some integrating with other interventions; these campaigns reached 218 million children. Seven countries conducted the campaigns over a wider target age group to address immunity gaps suggested by age distribution of measles cases.

In order to address the coverage gap of 21 million infants that did not receive MCV1 in 2013, activities are underway to focus on the 6 countries where 60% of these children live. The countries targeted are India, Nigeria, Ethiopia, Indonesia, Pakistan and DR Congo. The aim of the Global Routine Immunization Strategic Plan is to address these issues for all vaccines, and measles coverage is a critical indicator in this plan because it reflects coverage late in the first year of life. There is now a concerted effort by GAVI and global immunization partners to strengthen national immunization programs in these priority countries.

In addition, a working group on routine immunization has been organized to focus on how to break the 12 month barrier – targeting those countries that restrict routine measles vaccination to infants <12 months of age. Also efforts aimed at reducing the fear of wastage, and establishing a routine well-child visit in the 2nd year of life to deliver MCV2 and other child health interventions.

Reported measles cases have decreased from more than 800,000 in 2000 to a little over 265,000 in 2014. Calculating this as an incidence rate per million population, measles incidence decreased by 72% over this time period. Between 2013 and 2014, there was a decline in cases in Africa, but an increase in the W Pacific Region. However, the incidence of 41 per million in 2014 is still substantially higher than the target of <5 per million.

During the 12 months ending April 2015, the largest number of cases occurred in 14 countries based on the case-based system. The increase in cases in WPR was in China, the Philippines, PNG and Vietnam. In Somalia, DR Congo and PNG, aggregate disease surveillance systems report several times more cases than the case-based system. In PNG, the outbreak in late 2014 overwhelmed the case-based system, with 72,000 suspect cases reported, with about 5,000 tested and 2,000 confirmed.
There are variations in age distribution observed in outbreaks depending on the coverage achieved and the resulting level of vaccine-induced immunity. Where coverage is low, most cases are among children under 5 years of age. After 2012, a shift toward more cases in older age groups in Kenya was noted, a trend observed in other countries. In China, where there is very high coverage in vaccine-eligible children, cases occur at the extremes of the age distribution. The age distribution from the current post-elimination outbreak in Pernambuco State, Brazil – shows a bimodal age distribution. Although our understanding of age distribution and vaccination status of cases is incomplete, analysis of case-based data shows that the vast majority of cases in 2014 are still unvaccinated.

The reduction in mortality due to measles can be estimated based on the estimated deaths in the absence of vaccination. Mortality from measles has dropped over 75% and over 15.6 million lives have been saved since 2000 due to vaccination.

Turning to control targets, all regions but the Americas had incidence well above the 5 per million target. Progress has been stagnant in achieving MCV1 coverage. The 3 WHO regions with elimination targets at the end of 2015 lack adequate action or funds to achieve this goal. Although progress has been made, these regions are not on track to eliminate by 2015.

Based on MCV1 coverage in countries using MR or MMR vaccine, high RCV1 coverage has been achieved in many countries. Note that for rubella, 80% threshold of coverage is thought to be adequate to control rubella and CRS. Seven countries (Burkina Faso, Cameroon, India, Myanmar, Papua New Guinea, Viet Nam, Zimbabwe) are planning to introduce rubella vaccination. Many countries have yet to introduce rubella into their vaccination programs. Most countries in AFR and SEAR, as well as 3 large EMR countries still do not offer RCV in their routine system.

Rubella surveillance is not as complete as measles, a frustrating situation. For CRS, the number of countries reporting is even lower (141) and the gap between the number of reported cases compared to estimates is quite large (107 reported cases of CRS in 2014 vs. estimates of 30-100,000 cases of CRS each year).

The Americas region announced elimination of rubella in April of 2015- the first region to do so. EUR have announced a 2015 goal for rubella elimination and have met the goal in 43% of countries.

The 2014 Report on the Global Vaccine Action Plan made the recommendation that all Regions and countries establish verification commissions to “scrutinize and monitor progress towards elimination targets”. The Americas have eliminated measles in all but one country and EUR has achieved elimination of measles in 41% of countries. The W Pacific has met this goal in 22% of their countries. The African, E. Mediterranean, and SE Asian Regions and their member countries have yet to establish their verification mechanisms.

At the global level, guidelines have been developed for use by countries for introduction of a 2nd routine dose of measles vaccine and introduction of rubella vaccine into national immunization programs. Both these activities are being supported by GAVI with over $750M being made available to countries. Five of 41 remaining countries are scheduled to introduce MCV2 in 2015. For rubella vaccine introduction, 6 of 53 countries scheduled to introduce RCV will do so in 2015.
The approach being taken to improve the quality of SIAs is to strengthen country capacity to plan, implement and monitor their SIA performance. New seroprevalence survey methods will guide and evaluate SIAs. Efforts continue to fill the gap in funding for M/R control through 2020.

**In Summary**

- Rubella elimination has been achieved and verified in the Americas. However, a large inequity in access to rubella vaccine exists in numerous countries and regions which in turn bear the greatest burden of CRS.
- Based on current trends and program performance, 2015 global targets will not be achieved on time and 4 out of 6 WHO Regions are not on track to reach elimination goals. New tools, new funding, new plans can help resume progress.

Mick Mulders, WHO HQ Global Laboratory Coordinator, presented an

### UPDATE FROM THE GLOBAL MEASLES AND RUBELLA LABORATORY NETWORK (2014-2015)

As of June 2015, there are 723 laboratories in the GMRLN, an increase of 10 since the last GMRLN meeting in September 2014. The GMRLN is experiencing an increase in responsibilities and workload resulting from more case-based surveillance and elimination targets that require documentation of elimination with high quality data. There is a high incidence of measles in many regions however many countries are not reporting the percentage of suspected cases tested with a positive IgM result.

In 2014, there were 159,223 specimens tested for measles antibody and 59,136 positive IgM results were reported; in 2015 (as of 8 June) 47,121 specimens have been tested for measles IgM with 14,095 were reported positive by IgM. It was noted that in 2015 so far, over ¾ of cases were lab-confirmed and some testing in outbreak settings may increase workload unnecessarily. In 2014, 106,987 specimens were tested for rubella IgM, with 12,025 positive IgM results reported. Most negative measles cases are being tested for rubella, requiring additional funds for kits. Over 1.2 million Euro was spent for network labs. As costs are increasing for kits, available funds purchase fewer kits and expenses for molecular testing including EQA are also increasing.

Overall, labs are performing well in EQA. The results for the serology and the molecular panels have been very good, those results will be discussed at greater length in later sessions. None of the labs have failed accreditation, although some have passed provisionally.

The progress for accreditation for the GMRLN was summarized:

- Annualized in 4 Regions
- Desk review being implemented
- AMR (1GSL, 2RRL, 21NL): 18/24 full, 6 provisional
- EUR: (1GSL, 3RRL, 68 NL/SNL): 71/72 full, 1 provisional
- WPR: (1GSL, 3RRL, 52 NL): 52/56 full, 4 pending
- EMR: (2RRL, 21NL): 8 full, 1 provisional, 13 pending (sec.)
- SEAR: only onsite so far
- AFR: (3RRL, ESA 17NL, WCA 22NL): 26 full, 12 provisional, 3 pending
The GMRLN continues to improve molecular surveillance for measles. In the figure showing the distribution of measles genotypes for May 2014-April 2015, China should have a larger circle for H1 to reflect the proportion of cases (China experienced a large outbreak). However, other genotypes (B3, D8) have been detected in China, a reflection of sensitive surveillance to detect imported viruses. Globally, genotype D4 was predominant in recent years, however there has been an increase in detection of D8, along with B3. Submission of samples for rubella molecular surveillance continues to be a struggle; however, many labs are not reporting data.

Important work in progress includes development of 3 documents:

- Standardization of rubella IgG testing
- Seroprevalence guidelines
- Third edition of the WHO laboratory manual

A review of the potential for improved tracking of viruses by extended window, whole genome and next generation sequencing is underway. In the area of accreditation and EQA, the second round of molecular EQA (mEQA) is planned, along with an evaluation, partnering, and expansion of mEQA. Regarding IgM serology, a new web-based format for reporting of PT panel results will be introduced and additional stringency for completeness and timeliness are under consideration.

Inclusion of options for annualized accreditation by desk review is anticipated to reduce the burden on regional coordinators. Plans for on-site accreditation include Turkey, PHE-UK, India, Luxembourg, Pakistan, Côte d’Ivoire, Mali, and Indonesia. Laboratory assessments are planned for Thailand (Aug), Nigeria (Aug), Canada (Sep), China (Oct), Russia (Dec), and Myanmar (?).

Countries with tentative plans for assessments: Mongolia, Angola, Tanzania, Tunisia, Iran, Brazil (SP).

New technology that is under investigation and/or development:

- Alternative laboratory methods for assessing seroprevalence
- High throughput (MIA-Luminex)
- Point-of-care testing (lateral flow device)
- Vaccine-specific measles real-time RT-PCR

Trainings were conducted in Moscow (molecular workshop), in SEAR a lab management course was held, and CDC in Atlanta held a CIV training.

Noteworthy accomplishments for the LabNet:

- Laboratory capacity has improved in India, Indonesia, and Ethiopia
- Verification process in WPR and EUR
- Electronic linking of laboratory and epidemiological data is underway (EUR: MRLDMS) or under development (AFR, AMR, EMR, WPR, SEAR)
- Annual accreditation
- Monthly reporting of genotyping data (WPRO)
- Dried serum spots for QC testing; cross border sample transportation (Ebola)

Monthly bulletin for genotypes.
Longer term goals include integration with other VPDs, QC eQAvia web interface, Web-based and electronic training (eLearning), expansion of molecular methods for case confirmation and genotyping, and expansion of MeaNS/RubeNS databases.

Challenges for the LabNet and for MR control and elimination include:

- Expansion of elimination goals to include rubella
- Enhancing surveillance to support elimination goals and expansion of a second dose MR
- Adequate human resources in laboratories, turnover and training
- Genotype data, timeliness and completeness
- Continuing outbreaks and burden to laboratory (China, Philippines, DR Congo, Ethiopia, Mongolia- only laboratory confirmation- no epidemiologic linking)
- Competing priorities (Ebola, MERS-CoV)

In order to strengthen the LabNet and meet the challenges for case-based surveillance, the quality of data and performance of staff must be assured through EQA and annual accreditation. In addition, the following areas were identified for prioritization:

- Improve and expand capacity for genotyping as well as timeliness and completeness of data reporting
- Ensure sufficient laboratory capacity and coordination through staffing (regional coordinators)
- Identify additional funding resources, provide better mechanisms to project future funding needs to avoid lapses
- Improve genotyping surveillance, particularly for rubella
- Verification quality surveillance, including molecular surveillance
- Updated edition of lab manual to address current methodologies, goals and challenges
- Expansion of mEQA

Upcoming meetings and scientific conferences:

- 14th meeting of GMRLN (jointly with rotavirus, IBD?) TBD 2016
- SEAR LabNet – Oct, Bangkok
- Measles/rubella research symposium – 6-7 Oct, 2015, Atlanta
- ICM meeting – Nov, Amman
- AFR LabNet – TBD

Workshops and courses planned:

- SEAR: laboratory assessors’ course – Nov
- AFR: molecular workshop – Oct, Entebbe

SURVEILLANCE AND LABORATORY DATA

was presented by Marta Gacic-Dobo (HQ, IVB/EPI). Marta described the importance of linking together surveillance and laboratory data for a more accurate picture. Even so, only a small fraction of cases are reported, so the picture we get gives us a bias and many challenges exist for accurate and timely reporting. Data sources may be case-based or aggregate (lab data). Official routine data is comprised of aggregated data.
on suspected cases, confirmed cases, specimens tested and lab confirmed cases. Monthly data consists of case-based surveillance data and aggregate data from the laboratory.

The official annual data is not timely and different countries use different systems. All cases in outbreaks are not reflected in the data so reporting is not complete. For rubella, the information and reporting are very incomplete.

For the 12 month period (May 2014-April 2015) the incidence rate (per million population) for measles globally, based on available data, indicates that 75 countries (33%) have an incidence rate of <1 per million; 35 countries (18%) have rates over 1 but ≤5. Fifty countries (26%) have incidence rates between 5 and 49, and 17 countries (9%) have incidence rates ≥50. Seventeen countries, including India, have insufficient data or no data reported.

In order to gauge the sensitivity of measles surveillance by country, a target of 2 discarded cases per 100,000 population per year is used. Discarded cases are suspected measles cases which have been investigated and discarded as non-measles cases using laboratory testing and/or epidemiological-linkage. By this measure, 90 countries (46%) have adequate sensitivity. Thirty-six countries (19%) have ≤1 discarded case per 100,000 population. However, 68 countries (35%) have no case-based surveillance or insufficient data is reported to calculate the sensitivity indicator.

Cases in thousands, with method of measles confirmation by either laboratory, clinical, epi-link, or discarded, show different proportions, however all regions need more epi-linked confirmation. The monthly data by surveillance and laboratory were compared; certain lab data is more complete. The good news is that both systems are fairly similar but gaps exist in completeness so global situation can be determined. A better routine system is needed, one that is integrated, only a fraction of cases are reflected in case-based system. We need to identify the optimal method to share information and capture data from all sources in order to get a better picture of the situation.

IN SUMMARY
- Monthly data collection established for last 10 years, but still has limitations
- At global level surveillance and laboratory data are not linkable
- To achieve measles elimination, linked data from laboratory and surveillance system that is timely and reliable are critical
- Laboratory and surveillance needs to work together
- We need to work towards a reliable monthly/weekly global data but today we need to triangulate data from different sources
- Technology can help to build a better global data warehouse

Questions or comments:

Q. Is any effort underway to get information about CRS?

A. Not now at global level, but it is in the pipeline. Some efforts in some regions. There were 100 cases of CRS reported in 2014, trying to build capacity at national level. Guidelines are being developed for surveillance.
EMR continues to be a very challenging region due to conflicts (15/22 countries) and political issues. Ninety-eight percent of the unvaccinated children are in the areas with conflict. The target for the interruption of endemic transmission of measles has been proposed for 2017 with verification of elimination by 2020 in all countries of EMR. Nine of the countries have reported elimination target dates for rubella/CRS. SIAs have been conducted in EMR countries 2013-15, reaching 140 million people. In 19 countries, nationwide case-based surveillance was conducted in 2014; 3 countries conduct sentinel surveillance.

As of 2014, 8 of the countries (including the 2 RRLs) have the capacity to perform serologic testing, virus detection/isolation and sequencing.

In EMR the number of lab confirmed measles cases has decreased from 13,226 in 2013 to 5,855 as of Dec 2014. The number of rubella samples tested has remained relatively constant (~27,000) during 2012-2014 but the number of confirmed cases has doubled each year, with 284 confirmed in 2014.

During the first two quarters of 2015 on site accreditation review visits were conducted in Afghanistan, Pakistan, Kuwait, Qatar and Bahrain. In the last two quarters of 2015, onsite reviews are planned in Iran, SAA, Tunisia and UAE; desk reviews to be conducted in Iraq, Palestine and Syria.

There was good concordance of IgM results and accuracy in the practice panels for molecular detection and sequencing. Of the 5 surveillance performance indicators, 3 have improved between 2013 and 2014, but the sensitivity indicator of 2 discarded cases per 100,000 has decreased somewhat in 2014.

There is a high incidence of cases in many countries, particularly those affected by ongoing conflicts. There has been a spike in measles cases in Egypt in 2015.

Security concerns in many countries and funding problems continue to present challenges, along with ongoing outbreaks. However, moving forward, regional guidelines and commissions for verification of elimination are being developed and updated. The focus is on sustaining the lab technical capacity, maintaining quality performance, and improving surveillance indicators.

The priorities for 2015 include outbreak control in Egypt, Sudan, Somalia and verification of measles elimination in Bahrain, Palestine, and Oman.

**MEASLES AND RUBELLA ELIMINATION IN THE EMR, AN UPDATE ON THE ACTIVITIES OF RRL-TUNIS**

was given by Henda Triki,

The RRL in Tunis performs virus detection and genotyping and confirmation of serology results from NLs. Although the number of countries in the region that have forwarded serum for QC has declined since 2013, the results are good: the concordance is 96 and 97% for measles and rubella, respectively.

B3 and D8 are main genotypes detected since 2013. Palestine had previously never had a genotype identified, but in 2014 samples were sent and D8 was detected. Kuwait and Lebanon also had D8 detected in 2014. Through June 2015, B3 sequences were detected in Iran, Yemen, Oman, Saudi Arabia, and Somalia. The B3 strain has changed (now Harare lineage) from the previous B3s detected prior to 2012. Oman also had 2 D8 genotypes detected in 2015.

For rubella molecular surveillance, so far in 2015, there have been 27 samples from EMR, 21 of them from Tunisia. Some rubella sequences have not been reported.
There is a need to improve specimen collection from suspect cases of rubella. The problem arises for measles as well, frequently only serum is collected at the first contact.

**A REPORT FOR THE OMAN RRL AND NL ACTIVITIES**

was given by Said Al Baqlani.

There has been continuous improvement in timely testing (within 4 days) at the Oman NL since 2013. In 2015, (June), this indicator is 100%. Testing for IgM is done for both measles and rubella at the same time. Of 716 serum samples tested, 18 were positive for measles and 7 were positive for rubella.

For molecular surveillance in 2015, there have been more D9 genotypes detected compared to previous years. This cluster of D9 are similar to the strain identified in Japan. There were also 2 D8 sequences detected.

In regional activities in 2015 (June), confirmatory serology was performed for 177 samples from 5 of the 9 EMR countries that send samples to the Oman RRL, with overall 98% concordance. By country, all had concordance of >80%.

All 9 countries submitted samples for measles genotyping in 2014. In all of 2014, there were only 3 genotypes from 3 countries (Oman, Bahrain, Iran) determined for rubella (all 2B). In 2015 (June), 139 samples for measles have been sent from 4 countries including Oman; for rubella, 30 samples were received from UAE. Of the samples received, few PCR positive samples are able to be sequenced; for measles there were 44 (D8, D9, B3, and a single A genotype) and for rubella, only one (2A). In Oman most of the B3 sequences are related to the Harare strain.

An image of a gel was presented, there was CPE observed in Vero-SLAM cells for rubella, but this could not be rubella.

**AN UPDATE, MEASLES AND RUBELLA SURVEILLANCE IN PAKISTAN**

was given by Syed Sohail Zahoor Zaidi, subRegional Reference Laboratory at NIH Islamabad.

All performance indicators for the NL have been >80% since 2014. Indicator of reporting results used <7 days. There was a peak in measles cases in 2013 due to the outbreak when 10,000 samples were processed. The total samples processed in 2014 was 2,619 and these were tested for both measles (54% positive) and rubella (12% positive).

For the years 2014-2015, only B3 genotype has been detected from over 200 cases. The B3 sequence is closely related to that detected in Iran. There were also 22 samples with sequences from Afghanistan, all were B3.

**AN UPDATE FOR THE WESTERN PACIFIC REGION INCLUDING AN UPDATE ON THE CURRENT OUTBREAK IN MONGOLIA**

was given by Yan Zhang.

Cases of measles in WPR (mainly china) reached 80,576 in 2014, but by the 4th quarter of the year, cases had declined significantly only to rise again in 2015.
In March 2015, Brunei Darussalam, Cambodia, Japan were verified as having eliminated measles, for a total of 7 countries in WPR that have achieved this goal. New Zealand, Hong Kong (China), and Singapore may be ready to verify elimination in 2016.

The number of reported cases of rubella in WPR decreased each year since 2011. Almost half of reported cases in 2013 were in Japan. The highest proportion of CRS cases among 8 countries reporting CRS has shifted from Vietnam to Japan, but the overall number of cases have declined from 201 in 2011 to 12 in 2014.

Mongolia was certified by the Regional Verification Commission as having achieved elimination of measles in March 2014. A nationwide outbreak, affecting 21 provinces and the capital city (UB), was detected in early March 2015 despite high 2-dose MCV coverage. However the surveillance indicator of ≥2 per 100,000 discarded measles cases at 2nd level reporting units was less than 80%.

As of 5 June 2015, there were 11,046 suspected cases in UB; 2,067 were lab-confirmed. The highest number of confirmed cases have been among the <9 month old age group, however the highest proportion of suspected cases are among older groups, 15-24 years of age.

Among suspected cases tested for IgM by vaccination status that were tested Mar-May 2015 with known vaccination status, 80% of the IgM positive cases have occurred among unvaccinated persons. However, 19% of confirmed cases had 1 dose of MCV. Only 1 confirmed case among 154 lab-confirmed cases with known vaccination history had 2 doses of MCV. Nearly half of all suspected cases tested (Mar-May 2015) have been IgM negative. A proposal has been made to test IgM negative samples for IgG from suspected cases >6mo of age for which samples were collected within 3 days of rash.

Of 52 labs in WPR, 48 have maintained full accreditation, all labs participated in IgM PT and passed; 4 labs that participated in molecular PT passed.

Genotype H1 continues to be the most frequently detected genotype in the WPR; the proportion of H1 has increased from 2014 to 2015. H1 was also detected from the outbreak in Mongolia. In 2014, B3 spread from Philippines; in 2015, D8 genotype has been more prevalent than B3. There have been no D9 genotypes detected in 2015.

There was a WPR meeting held in Manila in March 2015. Priorities for strengthening WPR include improvement in the timeliness of reporting results with the target of 80% within 4 days and development of an outbreak management plan. Efforts are underway to improve virologic surveillance of rubella and to report the percentage of chains of transmission that have a genotype detected.

**Question and comments:**

**Q.** The cases in Mongolia- are most in children <9 months? What about measles immunity from maternal antibody?

**A.** Yes- in infants <9 months. Maternal antibody wanes in infants after 6 months.

**Comment:** Why are so many only clinically confirmed, in Mongolia that was verified as having eliminated measles. About the IgG testing <3 days post onset... why not do PCR?

**A.** After outbreak began, samples were collected, but later cases were epi-linked. The high workload resulted in many being clinically diagnosed. Yes, we will try to do both serum and PCR, but for some, they cannot get throat swab.

**Q.** In older groups, only 4% were lab confirmed? Have you looked at any other rash illness?

**A.** Yes rubella and measles.
Comment: H1 in Mongolia has been detected in other places.

**AN UPDATE OF THE GSL/RRL/NL ACTIVITIES OF NIID JAPAN**

was given by Makoto Takeda, in collaboration with Katsuhiro Komase and Yoshio Mori.

A training course, Laboratory Diagnosis Techniques for the Control of Vaccine Preventable Diseases, including Poliomyelitis and Measles was held Jan 19- Feb 13, 2015. The participants, from Nigeria, Vietnam, Mozambique and Egypt, were trained in M/R virus isolation in culture, detection by RT-PCR, and IFA (R), as well as detection of IgM by ELISA.

A seroepidemiology study using dried blood spots from a cluster sample set in Laos was conducted. In comparing detection of IgG for M/R, the measles antibody titer was lower than rubella. In order to rule out M/R variation in vaccine as a factor, testing was conducted to evaluate thermostability of vaccine components.

An analysis of rubella IgG kits was also conducted. Results are available at [http://www.nih.go.jp/niid/images/idsc/disease/rubella/RubellaHI-ElAtiter_Ver2.pdf](http://www.nih.go.jp/niid/images/idsc/disease/rubella/RubellaHI-ElAtiter_Ver2.pdf)

In Japan, IgM testing for M/R is provided by commercial labs, while RT-PCR is provided by 77 prefecture-level public health labs. In 2014, these labs tested 2,210 samples for measles by RT-PCR with 417 positives; for rubella there 984 were tested and 29 were positive.

In 2013, 232 measles cases were reported, as of week 38 in 2014, the number was 447. The increase in cases was due to an increase in measles activity in Southeast Asia. The genotypes determined in 2014 (Weeks 1-38) were H1, D8, D9 and B3.

As of 13 May 2015, there have been 67 reported cases of rubella and no cases of CRS detected. In 2014 there were 321 cases of rubella and 9 cases of CRS. During the peak of the rubella outbreak, rubella genotypes 2B-1 and 2B-2, as well as 1E were detected, in 2014 (Mar) only 2B-2 has been identified.

The Japanese Research Group Meeting for Measles and Rubella Elimination is held twice a year with participation from all prefectural laboratories, reference centers and the NIID. This year’s meeting will be 9-10 July.

A panel for molecular EQA was rolled out to 22 institutes with the aim of evaluating RNA extraction, the sensitivity of real-time RT-PCR to detect different genotypes, sequencing and molecular analysis. Positive results by RT-PCR varied according to the genotype, labs use different platforms.

Dr. Takeda presented data to show that a measles virus with 8 amino acid changes introduced into the H protein was neutralized by human serum to the same titer as the wild-type virus. These data suggested that multiple amino acid substitutions will not alter the recognition of measles H by polyclonal serum.

NIID studied CDV strains that were associated with outbreaks in monkeys. CDV does not bind to human SLAM but it does bind to human nectin 4. Recombinant CDV strains with site single directed mutations at amino acids 519, 541 or 541 in the CDV H protein could bind human SLAM suggesting that CDV could acquire the ability to infect humans.

NIID has plans to develop cells lines for isolation that can be shipped without CITES restrictions and that are highly sensitive to rubella virus.

In summary, measles cases are low, Japan is maintaining elimination status. The number of CRS cases in Japan looks high in the WPR because of the surveillance.
AN UPDATE OF THE CHINA CDC RRL/NL ACTIVITIES

was provided by Songtao Xu, in collaboration with Yan Zhang, Huiling Wang and Wenbo Xu, who is currently in Sierra Leon, working on Ebola.

In China, more than 50,000 cases were reported during 2014, and in 2015 (Jan-Apr), more than 20,000 cases have been reported. Since 2010, more than 90% of measles cases are lab-confirmed.

In 2014, among suspected sporadic cases, nearly 110,000 serum samples were tested and about 45% were IgM positive for measles and 3% were positive for rubella. There is concern about what is causing the rash and fever among the other 50%. This indicated that it is very necessary to do conduct fever and rash syndrome surveillance. Among cases associated with outbreaks, the serologic confirmation is much higher- over 90% were positive in 2014 and in 2015 (Jan-Apr).

Molecular surveillance for measles is good, 4,845 sequences were completed in 2014, 4,798 (99%) were H1 but non-endemic genotypes D8, D9, B3 and G3 were detected in 7 provinces. All provinces except Tibet are represented by molecular surveillance. So far in 2015 (Jan-Apr) only H1 has been detected in 21 provinces.

For proficiency panel testing, the NLM and 32 provincial laboratories passed the M/R serology IgM panel. For molecular EQA, RNA panels for RT-qPCR were distributed by NML to 21 provincial labs and all passed.

In 2014, the NML and 8 provincial labs participated in the WHO onsite review, and all passed with high scores.

With the support of the WHO and China CDC, a training and workshop was held in Wuhan 28-31 Oct 2014. This meeting promoted communication and sharing of data among participants from WHO, CDC, the NL/RRL, provincial laboratories, and EPI.

THE HONG KONG RRL/NL ACTIVITIES

reported by Janice Lo

The NL supports surveillance for M/M/R and CRS. In 2014, there were 50 cases reported for measles and 15 for rubella. About 74% of the measles cases were lab confirmed by IgM testing. There were no CRS cases reported. In 2014, 784 samples were tested for measles; 93% of the results were reported within 4 days. For rubella, 657 samples were tested in 2014.

In 2015 (May), 221 samples for measles and 209 for rubella have been tested. Of these, <10 cases for measles and rubella combined were IgM positive. In 2015 (May), 5 measles sequences have been identified as H1 (all similar) and 1 D8 was detected. For rubella, 2 of the 3 cases reported have also been genotyped, 2B and 1E. The HK lab has had some success with serum samples for rubella genotyping.

The HK RRL provides confirmatory testing for IgM for regional NLs. Concordance for both measles and rubella was >90% for 8 NLs in 2014 and 3 that have submitted in 2015. Samples submitted from 7 WPR regional NLs for RT-PCR and sequencing since 2014 have generated 163 measles genotypes from 292 samples submitted and 21 rubella genotypes from 97 samples submitted.

Several training courses have been completed already in 2015 covering laboratory techniques as well as laboratory surveillance and epidemiology. A molecular workshop is planned for Mar - Apr 2016 for WPRO countries.
THE UPDATE FOR THE AFRICAN REGION

was jointly presented by Charles Byabamazima followed by Annick Dosseh.

In 2014, of 44 reporting countries, there were 37,394 confirmed measles cases from 67,002 reported suspected cases. There were 77% of districts reporting at least 1 case with a serum sample, so somewhat below the 80% target.

There are 47 labs that include 2 at the sub-national level. Efforts continue to expand lab capacity and ensure quality control among the 47 labs in the region. Training for lab staff, monitoring of laboratory indicators and performing accreditation reviews are priority activities.

The 80% target for the 2 indicators for completeness and timeliness of sharing laboratory data remains a challenge, particularly the timeliness indicator. Some problems to overcome include unreliable internet connections, competing priorities, shortage of kits and staffing issues.

The workload is very high, although the number of samples received for surveillance at NLs in central AFR was lower in 2014 due to the Ebola outbreak and number of samples forwarded to the CIV RRL for confirmatory testing was greatly decreased. Couriers are rejecting samples from the area.

An algorithm for rubella testing from negative measles samples has improved rubella surveillance; a higher proportion of samples in 2014 were positive for rubella compared to measles.

A shortage of kits has been a big problem for the CIV lab, this RRL receives samples from 27 NLs for confirmatory testing.

There was very good performance from all but one lab in M/R IgM PT testing. Progress has been made in accreditation, however some are provisional due to failure to properly document and/or implement internal QC procedures.

Molecular surveillance is expanding with CIV initiating sequencing activities in 2015. B3 is the most commonly detected genotype in the region, followed by B2, D4, D2 and D10.

Funding is a major challenge for adequate staffing, training and supplies. Better coordination is needed between epi and lab to reduce unnecessary collection and testing of blood samples during outbreaks, while missed opportunities exist for collection of specimens for molecular surveillance.

AN UPDATE OF RRL/NL ACTIVITIES AT NICD, SOUTH AFRICA

was given by Sheilagh Smit.

In South Africa, the NL surveillance activities include testing measles samples for rubella (samples since May 2015) and expanding CRS surveillance to more provinces; 13 CRS cases have been identified in 2015 Five cases of SSPE were confirmed among children who were <9 months old when infected during outbreaks in 2009-11.

Most of the measles activity began in week 39 of 2014 and were concentrated in the Northern Cape and Gauteng. There was a total of 65 lab-confirmed cases of measles from 2 outbreaks as well as sporadic activity. Both infants <9 months of age and older age groups were affected in the sparsely populated Northern Cape. Genotype B3 was detected from both outbreaks.
So far in 2015 there have been 9 lab-confirmed measles cases. A cluster of 3 cases was investigated and evidence suggests the source was a HCW in an oncology ward who had traveled to Berlin 2 weeks prior to rash onset. One of the 3 spread cases was a fatal case. This case had no rash and never seroconverted. A family member developed measles 2 weeks later; the PCR from this case was negative. A second oncology patient was the 3rd case who developed measles 1 month later with no rash. Genotype D8 was identified from both oncology patients.

The RRL activities include confirmatory testing of serum from 9 NLs, samples should be submitted each quarter. Concordance since 2013 has remained >95%. Namibia ran out of kits so the RRL has been taking samples for primary testing. Samples for both measles and rubella testing were received from the Namibia NL. Over 150 of the 332 samples submitted were positive; most were rubella cases.

Principle challenges for the NL are delays in specimen transport to NCID, incomplete information on investigation forms or no form is submitted. Currently only blood is collected; viral samples are not incorporated into the protocol. There is a possible move for decentralized testing in 2016. All NLS are not submitting serum samples on a quarterly basis.

A PROGRESS REPORT OF MEASLES REGIONAL REFERENCE LABORATORY, CÔTE D’IVOIRE, 2014 -2015

was presented by Hervé Kadjo.

The RRL receives serum for confirmatory testing from regional labs, however 3 of these have been affected by Ebola outbreaks and have not submitted any samples, or very few, since 2013. Most of the results were good, but 5 of 16 countries that submitted samples for QC had some discordant results, however only 1 above 20% but they only sent 16 samples total.

More samples for PCR and genotyping need to be sent from the NLs. In 2014, only 3 countries submitted samples to the RRL. Nigeria and Togo have sent samples in 2015.

The capacity of the lab has improved since last meeting, due to training at CDC, thanks to Paul and Joe. Real-time RT-PCR has been implemented at CIV, and genotyping is pending receipt of reagents. Samples from AFR (CIV, Benin, Nigeria, Togo) were genotyped during the training at CDC. All were B3.

The RRL CIV also provides for transport of reagents and PT panels to 17 NLs.

The challenges for CIV are to fully implement sequencing before the end of the year and initiate molecular EQA. It is also necessary to raise awareness for the collection of samples for genotyping.

RRL/NL ACTIVITIES – PROGRESS TOWARDS MEASLES ELIMINATION, UGANDA

presented Barnabas Bakamutumaho

Following an SIA for measles in Uganda in 2011, the NL has supported both measles and rubella surveillance, demonstrating a sustained decrease in measles cases, while rubella cases have increased 2014-2015.

The indicator for testing of both NL testing for M/R surveillance and RRL confirmatory testing is excellent for timeliness based on 7 day turnaround.
Several of the countries in the Eastern Africa sub-region including Uganda have high incidence of rubella (>200 in 2014) but there is very little information on genotypes. Several initiatives for molecular surveillance of rubella and CRS surveillance are underway, as RCV introduction is planned. Support is needed to continue both measles and rubella sequencing.

Questions and comments:

Q. About CRS, could you explain the proposal for these projects, the IgG marker for CRS with Joe’s lab?

Joe: IgG subclass depends on the persistence of the infection. Mother, baby or both will have nonstandard IgG subclass, project to be done. There are many markers of congenital infection. The idea is to look for additional markers as result of long term exposure. It is really encouraging to see CRS research projects being planned and operationalized. Are these national surveillance projects? Or research?

A. In S. Africa- national. Tertiary hospitals, 31 sites. CRS is a reportable disease.

Q. Could you remark on the diagnosis of SSPE cases in S Africa?

A. Sheilagh: identification was by tertiary hospitals, by IgG and index of total IgG and albumen. The IgG was sky-high in both serum and blood.

**AN UPDATE ON MEASLES SURVEILLANCE AND LABORATORY EXPANSION IN ETHIOPIA NL AND SNL**

was given by Berhane Beyene.

In 2014, serum samples were collected from 91% of reported measles cases, and 53% of these were IgM positive.

For the period from May 2014 to May 2015, 192 outbreaks of measles have occurred, 1,050 outbreak cases were lab-confirmed and 5,682 cases were epi-linked. During the same time period, there were 349 confirmed cases of rubella.

There were 14,877 suspected cases of measles reported through May 2015; 19% were lab-confirmed, 80% were epi-linked, and 1% were clinically compatible. Although the <5 year age group was most affected, cases occurred in all age groups.

The NL is fully accredited and has demonstrated excellent performance through proficiency testing and accuracy of testing by EQC.

The establishment of a sub-national lab (sNL) had been proposed to reduce the workload burden of surveillance samples for a highly populated country. A sNL could reduce transport costs and improve the indicators for timeliness of results. Several criteria were assessed to determine the eligibility for a sNL, focusing on 4 labs serving 4 large regions. Two labs- the Bahir Dar and Hawassa Regional laboratories- were chosen to serve as sNLs. Parallel testing with the NL was ongoing until the sNLs achieved 100% concordance. In May 2015, the sNLs began testing and reporting.

Challenges include delays in results reaching outbreak response teams, assuring availability of kits for outbreaks, equipment maintenance and transport of samples.

Capacity expansion at both NL and the 2 sNLs are planned, efforts will continue to improve and maintain quality assurance activities and improve lab surveillance indicators.
AN UPDATE OF THE SOUTH EAST ASIAN REGION

was given by Sirima Pattamadilok.

The region must establish and maintain quality and the evolution of this process has increased the number of labs to provide surveillance to the region. The number of labs stands at 39 in 2015 but the goals of 2020 may see this increase even more. There are 5 proposed new labs - 3 in Indonesia, 1 in Myanmar, and 1 in Nepal to support national case-based surveillance.

As of May 2015, there are 36 proficient laboratories out of 39 laboratories in the SEAR MR LabNet (Timor-Leste, Chandigarh, Delhi have yet to receive accreditation).

Sequencing is performed in 3 labs: Pune, Bandung and Bangkok, all passed the 2014 molecular EQA testing.

In 2014, the SEAR MR lab network tested over 20,000 samples - Indonesia has the highest proportion of samples across the region.

For timeliness of measles reporting, the indicator in 2014 was 7 days, in 2015 it is 4 days. As of April 2015, in this year the 80% target has been reached for the more stringent 4 day reporting with the exception of India. All labs have ≥90% concordance for IgM for M/R serology panel testing.

For measles molecular surveillance, D5 is no longer being detected, B3 is new for India and Thailand, and the first genotyping in Bangladesh, also B3. For rubella, there have been 1E and 2B identified.

Progress has been made on many of the 2014 ITAG recommendations. A big challenge is to provide a genotype from 80% of the chains of transmission.

A training workshop is scheduled for laboratory aspects of CRS surveillance on Oct 2015.

Questions and comments:

Q. Yan asked Sirima about genotypes, need denominator.
A. Sirima: in progress, agree, need denominator.

Q. To all speakers - maternal immunity. Does it remain until 9 months?
A. Sirima - we follow a schedule, WHO guidance. In a cohort study, antibody titer declined around 6 months, there is a 6 to 9 months range, justification for EPI, 9-12 months for first vaccination.

Q. Lucky, would you consider taking more samples to confirm?
A. Will keep that in mind.


AN UPDATE OF THE RRL ACTIVITIES THAILAND

was given by Atchariya Lukebua, NIH Bangkok.

The RRL/NL at NIH Bangkok has developed strategies based on the recommendations made for 2016-2020 for measles elimination. These strategies include expanding the capacity for RT-PCR and meeting the surveillance sensitivity indicator for discarded cases in 80% of all areas.
In 2014, 414 samples were tested for measles and rubella. Compared to 2011-2013, the proportion of measles cases has decreased significantly (18% in 2014) and 5% were positive for rubella. In 2015 (Jan – May), of 190 cases tested, there have been 2% positive for measles and 12% positive for rubella- the first time that rubella cases have outnumbered measles cases since 2011.

For the indicator of timeliness of reporting, 95% of results of testing were reported within 4 days of receipt in 2014 and through May of 2015 has increased to 97%. All (100%) of genotype results have been reported within 2 months for 2014- through May of 2015.

Thailand had established 13 SNLs across the country. All of the labs achieved 100% concordance for the WHO IgM panel for both measles and rubella. The 2015 serology confirmatory QC testing showed very high (most 100%) concordance.

In addition to these EQC activities, the Thai NIH continues to provide training, conduct on-site visits, and assessing performance of the SNLs through either WHO accreditation or certification through ISO.

The RRL activities include training to support capacity for countries in the region. A training workshop for lab personnel from Pune, India NL was held from 2-20 Feb 2015.

Although confirmatory testing of IgM from NLs has not yet achieved full participation annually among a few NLs, all NLs have submitted samples in the period 2014-2015 (June). The IgM results for the 5 NLs that sent so far in 2015 had >94% concordance for measles and >90% for rubella and this included 2 countries that sent dried serum samples (DSS).

AN OVERVIEW OF ACTIVITIES TOWARD BUILDING MEASLES RUBELLA LABORATORY NETWORK IN INDIA

presented by Lucky Sangal

As the target of 2020 has been set for measles elimination and rubella/CRS control, strategies are in place to improve MCV2 vaccination coverage by the states across the country. This coverage increased in 2014.

Also, vaccination campaigns with M/R vaccination targeting wide age groups (9 months through <15 years) are planned. Routine doses of M/R will replace the MVC doses in the schedule in a phased manner.

In response to the M/R campaign, India will move from aggregate reporting to a case-based system. Sentinel sites will provide CRS surveillance and help evaluate the impact of the rubella vaccination.

In 2014, >54,000 cases were reported in 2014. In previous years, not fewer cases, just less surveillance, it is more sensitive now that the goal of measles elimination has been accepted. So far in 2015 (7 June), 36,511 measles cases have been reported. In 2014 and 2015 both measles and rubella cases have been confirmed within outbreaks.

In the lab network, it has expanded in northern part of the country, now there are 13 labs and 1 proposed lab in the islands, since it is difficult to ship samples. Seven labs are now performing molecular testing.

Over 5100 samples have been tested in 2015 (7 June); this is roughly equivalent to all cases tested in all of 2014. The workload of testing has shifted somewhat during 2013-15, but some labs receive more samples.
The timeliness indicator (4 days) has not reached the target among many labs, although there has been some improvement in 2015. Challenges are high workload, kits are used up. We hope to be more prepared next year.

The concordance for confirmatory results for IgM measles was very good in 2014, only 1 lab had problems. There were more problems with rubella IgM concordance. In 2015, this has improved across all labs for both M/R with 1 exception, but that lab had 100% concordance in 2014. No problem with DSS.

Genotypes were provided from 134 samples from 290 that were positive from >700 specimens received Jan 2014 through June 2015. Genotype D8 was detected from outbreaks in most districts, also some D4. There was an outbreak in northern India (Mathura), Nov 2014, with 19 cases. It was B3, Harare strain.

The outbreak in Mathura was notified on 11 Nov, but onsets of first cases began in early October. The outbreak affected mostly unvaccinated children <10 years of age. The last onset was Nov 14.

In order to transition to case-based surveillance, additional expansion of network may be needed in hard-to-reach areas and if necessary for political reasons. Some of the states are independent and want a lab of their own.

Also, collaboration with other institutes that engage in M/R diagnostics will be pursued. One of these, the Indian Council of Medical Research (ICMR) will supply proficiency panel in a step to decentralize EQA. ELISA kits manufactured by the government of India for M/R is being explored since Siemens kits are very expensive.

Future plans of the network include improving molecular surveillance by reducing the time needed for results and expediting the transport of positive PCR samples to labs performing sequencing.

Questions and comments:
Comment, Mick: regarding the classification of cases with an equivocal result, in an elimination setting, having an equivocal will require more testing, additional samples.

STRENGTHENING LABORATORY CAPACITY INDONESIA

Presented by Rusipah.

Indonesia is a challenging area for M/R surveillance – the country has >17,000 islands and has a population of >252 million. Our objectives are to increase case-based surveillance, beginning with 50% in 2015 and increasing each year until 100% in 2020.

Other objectives include increases in MCV1 and MCV2 coverage and campaigns in high risk areas. In addition, all measles outbreaks will be fully investigated and CRS surveillance will be rolled out to 12 provinces in 2015 and expanded in 2017-2019 to all provinces.

In the existing network, there are 4 NLs, with large variations in the proportion of the total population served. All labs have demonstrated good performance in proficiency and have been accredited in 2014, however only the lab in Bandung has met the target for timeliness of lab results. Timeliness indicator is low due to availability of reagents.

Molecular surveillance has been ongoing and shows a wide distribution of measles genotype D9. Information for rubella genotypes is restricted to only 3 provinces.
Lab expansion is needed to match the government commitments by 2020. With the geographic challenges and population, 3 new SNLs could provide some much needed support for surveillance and help spread the workload for serology testing more evenly.

There are many challenges including lack of coordination between lab and surveillance, and the need for human resources and operational cost. However, the 4 NLs can assist and support the 3 SNLs and the opportunity exists to enhance capacity through govt support and advocacy for the 2020 goals.

**AN UPDATE OF MEASLES OUTBREAKS IN NORTH EASTERN REGION IN BRAZIL 2013-2015**

was presented by Marilda Siquiera.

Brazil experienced outbreaks in 2013-15, beginning in epi weeks 11-12. The first state affected was Pernambuco and the first case occurred on 19 March 2013. The outbreak lasted 51 weeks, with the last onset on 14 March 2014, with a total of 244 cases, 198 of these were lab-confirmed. Fifty percent of the cases occurred among children <1 year of age. Genotype D8 was identified from outbreak cases.

The outbreak in Ceara began during the last week of 2013 and continued into 2015. In the early 1980s the vaccination coverage in Ceara had been quite low, but had reached 95% and after 1997 there had been no cases for almost 17 years.

The highest incidence occurred in the children <1 year of age, followed by the 20-29 age group.

Last year, in 2014, there were campaigns targeted to different age groups. However, surveillance was not complete, Brazil continued to have measles – 163 cases in 2015, 1 imported to another state on the northern coast. The last confirmed case was 2 June, although some suspected cases are still pending. The genotype D8 was also confirmed from this outbreak.

Among 161 confirmed measles cases in the Ceara outbreak, 47% were unvaccinated and 12% were <6 months of age. Vaccination status was unknown for 30% and the remaining 11% were vaccinated.

In order to control the outbreak, weekly negative reporting was provided by 90% of the reporting units. A second serum sample was collected from cases with inconclusive results.

These outbreaks showed a problem with consistent surveillance across the country. The outbreak had a negative impact on other health services and a high workload for dengue, zika, CKG, and flu. Work in progress includes a differential diagnosis and protocol for testing zika virus.

**AN UPDATE OF GSL/RRL/NL MEASLES ACTIVITIES FOR USA**

was given by Paul Rota, CDC, Atlanta.

To review the measles activity in the US in 2014, there were 2 instances in which measles outbreaks occurred primarily among unvaccinated individuals. The first was in a religious community in Ohio with over 400 cases. At the end of 2014, an outbreak was traced to an amusement park in California, where again, there were pockets of susceptibility.

In the US in 2015 (provisional data, June 2015) there have been 173 cases in 21 states and DC. Most have been in the western USA - California, Washington and Colorado. Most of the cases could be demonstrated to be import-associated (95%). Genotypes for which the source was identified included both D8 and D9 from India (4
separate imports) and 2 imports of H1 from China. D9 was associated with an import from Qatar. The cases in 2015 include 5 outbreaks, one of these had carried over from 2014, the amusement park in CA. The last onset for this outbreak was 2 March 2015.

In the US, 4 states have a lab designated as a VPD reference lab and these perform PCR and genotyping for measles. In 2014, the VPD ref labs tested approximately 500 samples.

Paul presented a preliminary report of an evaluation of FTA cards for specimen transport. The study used samples from the Democratic Republic of Congo (DRC). Specimens consisting of 2 throat swabs, 2 Oracol® swabs and a serum sample collected from suspected cases of measles or rubella in DRC during 2014. The study compared standard respiratory samples vs specimens that had been eluted and spotted on FTA cards and dried.

Of 182 cases (excluding those from patients with serum that tested IgM positive for rubella or those with negative results for measles from all 5 samples collected), the RT-PCR data showed comparable results for throat swabs vs Oracol® swabs. The sensitivity of real-time RT-PCR decreased using the FTA cards, (about 10-15% fewer positive), but overall the ability to detect measles RNA and also to genotype the virus using FTA cards was very good. The copy number was the limiting factor: about 1,000 copies were required for successful genotyping.

**GSL/RRL/NL RUBELLA ACTIVITIES FOR USA**

Presented by Joe Icenogle, CDC, Atlanta

As noted earlier in the meeting, the region of the Americas has been verified as having eliminated rubella. Although it is an infrequent occurrence, sporadic imported cases of rubella are detected in the US. In Feb of 2015 there was 1 imported case of rubella from the Philippines. The genotype was 1J. There have been no cases of CRS in 2015.

As the RRL, the CDC rubella team performs RT-PCR, IgM and IgG avidity testing to assist PAHO in case classification of challenging cases. The approximate yearly testing is <100 samples. CDC also provides reagents for RT-PCR kits and panels for molecular testing.

Other activities of the GSL include participation in workshops, in-house training of GMRLN members, collaboration in research studies and curation of RubeNS.

The rubella team at CDC is working to develop an ELISA-based assay for detecting serologic markers for CRS including the C protein recently described (Hyde et al., 2015, JID).

A research study is proposed to identify the challenges for developing and maintaining surveillance activities for rubella in the GMRLN to improve support for the goals of rubella and CRS control and elimination. We don’t have good data about what the challenges are. At present we are working with WHO/HQ, and regional epis, have not yet had discussions with national labs.

The initial proposal would focus on 2-3 regions, WPR, SEAR and AFR, targeting one or more countries in the regions. Stage 1 of the project would include background research, reaching out to stakeholders and determination of appropriate data collection tools. The aim is to address the turnaround time, transport of samples and the new challenges for CRS.

There is a new document from WHO on introducing rubella vaccine into national immunization programs. This guide in the works, not sure what the status of document is.
We have been studying persistence of rubella from different directions, to understand persistent infection. There is a lot to learn about persistence and identification of a target in CRS cases. Hope to have something in the future to aid surveillance.

AN UPDATE OF RRL/NL ACTIVITIES FOR CANADA

Joanne Hiebert, PHAC, Winnipeg, presented

The NL activities at the NML in Winnipeg include confirmatory testing and EQA and PT panels for the provincial labs (SNLs). The serology performed at the NML is reference testing and specialized testing. The NL also provides all molecular testing for rubella and for measles among the SNLs that do not perform RT-PCR. As an RRL, Vero-SLAM cells are distributed to regional labs that request them. The testing performed at the NL/RRL meet certification by ISO.

For the period Sep 2014- Jun 2015, 159 samples were tested by RT-PCR for rubella, all were negative. There were 50 tested for CRS surveillance. Ontario routinely sends still-birth fetal tissue to PHAC for testing. During this period 273 specimens were referred to NML and 86 RT-PCR positive samples were sent for genotyping.

The majority of the samples genotyped were vaccine strain (34 of 86 genotypes). Genotypes B3, D4, H1 and D8 were also identified from cases during the 10-month period.

There were 4 measles outbreaks consisting of 190 cases and 5 sporadic cases from Sep 2014- June 2015. The genotype was identified from the 4 outbreaks and the 5 sporadic cases. Only 1 outbreak with 18 cases had an unknown source. The other outbreaks were traced to the USA (CA outbreak, amusement park), India and China.

The initial 13 cases associated with the outbreak with unknown source did not have epi-links and the D4 genotype had no exact matches in MeaNS. Extended sequencing of 17 of the 18 cases added supporting evidence that all the cases were part of the same outbreak.

34 cases of vaccine sequences using the vaccine specific primers for real time RT-PCR, many requests for urgency for testing.

Development of a vaccine-specific (genotype A) real-time RT-PCR assay enabled the rapid identification of vaccine-associated measles cases. This assay was very useful since there were 34 post-vaccination cases suspected of measles and there is urgency for rapid results in these situations, particularly in the setting of an outbreak.

The NML has a good relationship with epi colleagues, and collaborate on weekly and annual report. It is necessary to align the epi weeks.

CARPHA, SUBRRL/NL ACTIVITIES IN THE CARIBBEAN SUB-REGION OF AMR

was given by Pablo Martinez de Salazar, CARPHA, Port of Spain, Trinidad and Tobago.

In addition to supporting the surveillance for M/R in the Caribbean community (CARICOM), CARPHA serves as RRL for Nicaragua, the Dominican Republic (DOR) and Haiti NLs. There are 700 sites reporting weekly from 19 countries.
The last accreditation for CARPHA was February 2014. The results for the 2014 PT M/R serology panel was 100% concordance.

Because of the high tourist traffic in the sub-region, surveillance for rash and fever and high MR coverage (95%) among birth cohorts are priorities. Surveillance for rash and fever in 2014 included testing 407 samples for measles and rubella IgM. Only 4 positives were obtained, and all were associated with recent vaccination.

Although the total number of samples sent for confirmatory testing from Nicaragua, Haiti, and DOR were low in 2014, the concordance was 100%.

For surveillance indicators, 82% of suspected cases had adequate sample collection and were obtained within 28 days. Twenty percent of samples arrived to the lab within 5 days after collection and 95% of the results for testing were generated within 4 days. The vaccination status information was missing from 47% of the samples.

There are significant challenges that affect the timely arrival of samples to the lab- many islands, regulations, shipment issues. Also there are rash illnesses that must be considered in the differential diagnosis including chikungunya, dengue and zika virus. There are concerns about the delays in rule-out of for M/R from samples that are initially suspected of dengue or the other rash-causing viral infections that are negative for those diseases.

Questions and comments:

Q. Can 1 outbreak have >1 genotype? In France, 3 genotypes.
A. An outbreak in DRC overlapped with a 2nd outbreak, one disappeared, the other appeared, but both at the same time, in parallel. Also can have multiple imported cases in same city with same genotype.

Some genotypes so widely distributed, same virus, same genotype, but from repeated importations.

Q. How do you dry blood spots on FTA card?
A. Samples were not blood- they were throat swabs and oral fluid. Dried on bench for several hours. Some had mold contamination because they were not completely dried when shipped.

Comment from Mick: Share protocols for FTA cards and the protocols for real-time RT-PCR targeting vaccine strains.

Q. Are most cases confirmed by PCR in the USA?
A. Paul: many are confirmed by PCR, not sure about serum. Much of the IgM is done in private sector. The database of reported cases from the state is sent to the MMR epis at CDC includes the method of confirmation from state epis- accuracy unknown. In the US, now moving toward more PCR confirmation.

Q. Can urine be use to spot on FTA cards?
A. (Paul) Probably could do urine, but have not done that. Difficult to work with since there are often precipitants.

Comment from Paul: Vaccine-specific primers/PCR very important, rush requests, many vaccine rashes. During the outbreak, exposure got to be anybody living in CA. Requirement for genotyping increases turnaround time for answer, can stand down if vaccine.

Q. About urine samples for virus on FTA… maybe could be utilized after centrifugation?
A. Maybe, but don’t know.

Q. About the cases in the outbreak in Brazil- vaccination coverage very high- what was their vaccination status?
A. (Marilda) Even with campaigns, still have susceptibles, is a major problem if follow-up of cases is not good, cannot stop the spread.

AN UPDATE FOR THE EUROPEAN REGION

was given by Myriam Ben Mamou.

In 2014, the EUR region continues to have a high incidence of measles. Ten of the 53 member countries had 91% of the cases in 2014. For rubella, Poland leads in the number of cases, with nearly 6,000 cases in 2014. However the total number of cases of both measles and rubella decreased in 2014 compared to 2013, by 50% and 84% respectively. Despite this, there are ongoing outbreaks, with cases in adults and some nosocomial transmission, highlighting susceptibility of HCWs.

The 4th meeting of the Regional Verification Commission (RVC) took place on 27-29 Oct 2015.

Some modifications to the verification process were proposed. In short, the countries are asked to provide the data that shows surveillance to be timely, complete, and sufficiently sensitive for verification purposes and the data should be available in a standardized format. In reviewing the status of the EUR region in 2013, the RVC noted that 9 countries have inconclusive evidence so determine if interruption of transmission of measles has occurred. However 15 countries had evidence to support the interruption of both measles and rubella.

Throughout the EUR LabNet, 11,000 specimens have been tested in 2015 (8 June). Most samples had a negative result, and very few of the positives were rubella cases.

The EUR labs that are scheduled for accreditation during July-Dec 2015 are Kazakhstan, Poland Georgia, and the RRL Moscow. Dec 2015. Desk reviews for accreditation in 2016 include 48 NLs and 20 SNLs.

The accreditation criteria that are below target are reporting IgM results in ≤4 days, adequate IQC procedures and timely reporting of genotypes.

Although progress has been made, the EUR region is not on track for the 2015 goals. Additional political commitment, improvements in surveillance, timely reporting, and coordination between lab and epi data are needed.

The WHO/Europe package for accelerated action identifies priority work areas on which the WHO European Regional Office will strengthen its technical support to Member States as they seek to eliminate measles and rubella. This package will set indicators and milestones on which to measure WHO/Europe and Member State progress.

The EUR LabNet performs at high levels of proficiency but have challenges to address the requirements for verification. Efforts are needed to provide better dissemination of measles genotyping data to describe chains of transmission, improve rubella laboratory confirmation and genotyping, and increase timeliness and completeness of lab results.

AN UPDATE OF GSL/RRL/NL ACTIVITIES FOR UK, PUBLIC HEALTH ENGLAND

was presented by Kevin Brown.

In the 1st quarter of 2015, coverage for MMR1 is estimated at 95%, an improvement but still a drop due to Wakefield. Cases were occurring in ages 11-20; a catch-up campaign in England targeted affected groups and
has had a dramatic effect. There were 2000 cases a month, now have decreases and positive cases have fallen in Wales and England.

For the first time in a number of years, England no longer has endemic measles. There have been multiple sporadic imported strains, but no evidence of endemic viruses.

As a consequence of the 2012-13 outbreak, there have been improvements in the coordination with labs in the UK. PHE initiated a program to roll-out measles PCR to PHLs. A validation panel was produced and all labs passed.

Surveillance for rubella continues, the UK had only 1 rubella case in 2014. So far in 2015, there have been 2 cases of CRS, the mothers acquired infection outside of the UK (Tanzania, Zimbabwe).

Kevin discussed the issues associated with testing for measles by PCR for case classification. Increasingly, labs want to do only PCR. If the result is negative, this outcome is not contributory to resolving the status of the case. One must consider the factors that could contribute to a negative result in a true case. Also, labs should include a cellular control for RNA.

There are concerns that the decrease of cases may result in reduced funding. Also, restructuring in PHE amid a trend toward decentralized testing in England as well as testing being done in Scotland, Ireland raise concerns about reporting and confirmatory testing.

THE RRL MOSCOW/NIS SUBREGION LABORATORY NETWORK ACTIVITIES SUMMARY 2014 – 2015

was given by Sergey Shulga.

The 24 labs in the NIS subregion supporting surveillance in 11 countries are all fully accredited. The NLs and the SNLs provide primary serologic testing, the NL in Belarus also does molecular testing and genotyping. All the labs participated in the 2014 serology proficiency panel and had 100% concordance.

There are 3 different IgM kits used among the labs. The confirmatory testing samples (n=1,639) sent from NLs to the RRL in 2014 showed 100% concordance despite sometimes using a different kit at RRL. There was no difference in results between Siemens and Euroimmune. However some labs only sent negative samples and one lab sent only 11 samples each for M/R retesting.

In 2014, about 7,000 samples were tested for both measles and rubella, half were positive for measles, no rubella positives.

In 2015, there has been 1 case of CRS; this case occurred in an unvaccinated female in a Roma traveling group in Astrakhan, it was a fatal case. The CRS case was IgM/PCR confirmed, the genotype is pending.

The NIS subregion has had large outbreaks in Ukraine, Georgia and the Russian Federation. In 2015 there has been increased measles activity in Kazakhstan with >800 cases, and Kyrgyzstan with >7,000 cases. The molecular surveillance in 2014 is incomplete, but genotype D8 was the most predominate with 304 samples sequenced. Imported cases were detected, B3 and D4.

A workshop for laboratory diagnosis for measles and rubella for 5 SNLs was held in Nov 2014. In April 2015, a workshop (ELIZA, quality assurance procedures), was held in Dushanbe Tajikistan and in June there was a molecular testing, genotyping and data management workshop for 6 participants (Kazakhstan, Azerbaijan, Tajikistan). On-site visits for these 3 NLs are planned July-Sept 2015.
There are ongoing efforts to develop proficiency and validation panels and internal controls for ELISA testing, primarily for the Vector kits. Challenges and priorities include staffing, obtaining needed supplies and maintaining quality assurance programs.

Annette Mankertz and Sabine Santibanez of the Robert Koch Institute, Berlin, presented the RRL Berlin activities.

The EQA for measles and rubella RT-PCR and genotyping has been very successful using FTA cards.

A guide for viral diagnosis for pregnant women has been published, which includes testing for measles, mumps, and rubella.

Germany has a special situation due to division of country for many years. In October 2014, the former eastern part of Berlin began to see increased measles activity. The outbreak moved from refugees to the main population. As of June 29, 2015, there have been 1,300 cases in Berlin. There was one fatal case, a boy with an unrecognized heart problem, who had not been vaccinated due to parental opinion that he was too fragile to receive vaccination. After attention in the news media, vaccination uptake increased. In Berlin, there was very high incidence among infants ≤1 year of age.

A national action plan for development of strategic and actionable goals for measles and rubella control is underway.

Sabena presented genotype data and analysis for D8 viruses, including tracking of a D8 variant detected in Russia (Rostov on Don). This strain has been detected in many EUR countries in 2015 including importations from Bosnia-Herzegovina. Germany has had sustained circulation of this strain.

Among the RRL activities, re-testing of serum for measles and rubella in 2014 was performed for the EUR region. Good concordance was obtained for measles using different kits; for rubella, there were substantial discordant results using the Diesse kit.

AN UPDATE FROM RRL LUXEMBOURG

was given by Judith Hübschen, LIH Luxembourg.

For 2014, 27 NLs participated in the PT serum panel IgM testing for measles and rubella. Most results were 100% concordant using 20/20 samples; however some countries tested only 18 or 19, citing low volume. More samples were received for confirmatory testing in 2015; a higher proportion are sending dried serum than in previous years. We also received dried blood spots. There was good compliance with new instructions, asking for only samples from suspected cases. There was good concordance for the re-testing. Many countries are receiving <50 samples from suspected cases per year.

In other RRL activities, specimens on filter paper were received for PCR and sequencing. Of 83 tested, 73 (88%) could be genotyped. Of those that had an IgM + result, the percent genotyped was 95%. Research activities include studies in Laos and complete genome sequencing.

Questions and comments:

Q. Paul: comment about PCR to Kevin: We should not use negative PCR to rule out a case, agree for the need to use an RNA control.

Comment: It was noted that some countries fail to meet criteria for collecting samples- they only collect throat swabs, not getting serum. There is work to do to define verification guidelines, including clarifying composition of adequate samples.
Kevin: oral fluid can be tested for IgM, however in UK, Scotland not even taking oral fluid sample. Negative PCR results are not reported. It is increasingly difficult to get serum samples.

Robert: throat swabs and oral fluid help with getting samples for sequencing. Some tests are more likely to be positive early, for verification needs to be clear. Information may go out of date quickly with new testing. Some labs have sophisticated systems for billing, is there any way to tap into those types of systems to see what they are doing? Raphael Harpaz looked into that in the US, Germany- incidence per million...always very high in young kids, there are fewer of them; there are more people in higher age groups.

Data in private labs, yes, they have the info, the positive information is for public health, the negative data are not available, no reason to collect; it is confidential, etc.

Mick: any definitions may change, what we are discussing are countries only doing PCR, will bring this forward on lab manual.

Joe: 3 comments and a Q. Kevin was talking about importation of CRS from C Africa...rubella incidence data is needed from C Africa, to help predict when cases will come. Oral fluid is measles-centric, IgM cannot be done for rubella from oral fluid, no test kit for rubella. Kevin, on your encephalitis case, was there infectious virus?

Kevin: yes there was infectious virus, sequence is in RubeNS. Regarding testing of rubella IgM from oral fluid, Microimmune has solved the problem with availability.

### SESSION 4: EQA

#### MEASLES AND RUBELLA IGM PROFICIENCY TESTING – PANEL 01404 AND NEW SCORING ALGORITHM FOR NEXT PANEL 01503

presented by Vicki Stambos, RRL, Australia, VIDRL, Melbourne

The panel 01404 consisted of 20 serum samples, 6 positive for measles and 7 positive for rubella. The 7 measles/rubella negative samples included 1 sample positive for dengue and 1 positive for parvovirus. Measles IgM results were returned from 214 labs and rubella results were returned from 213 labs. Most labs (98%) reported kit lot numbers, however many (about one-third) omitted filling in cut-off values. Validation criteria was generally complete (>90%).

In terms of timeliness for testing and for reporting (≤14 days), 86-100% of labs by region tested in a timely manner, however reporting within the 14-day timeframe was met by only 71-92% of the labs by region.

Following analysis of results, measles results for sample 003 (a rubella positive serum) were discrepant using different kits from 35 labs. Therefore, an adjustment was made for labs with discordant results for measles. Globally, 99% of the results had scores ≥90%.

Among the challenges for PT testing are issues involving shipment of serum. One possible method to address this problem would be to utilize dried blood spots (DBS). The use of DBS would provide cost saving for shipping as well as elimination of delays in customs. However, disadvantages include the necessity of an elution step, reagent and equipment needs and training. In addition, the use of DBS would require a large volume of whole blood to spike with IgM + serum.
Vicki introduced the work done at VIDRL to address some of the challenges in PT testing, namely, development of a new PT scoring algorithm and a web-based method for submission of results. The new algorithm for scoring would take raw data (OD), timeliness of testing and reporting, kit details and validation information into consideration when calculating the overall score. An in-house control may also be part of the algorithm. A weighted score method was described.

The web-based method for submission of results was introduced and web pages for viewing. The web-based method has advantages of required fields to capture information often omitted, as well as ease and efficiency of reporting results. The proposal is to have a transition period in 2015, with panels delayed until the report form and website have been completed (September).

Questions and comments:

Q. Lucky- Can SEAR consider using another measles/rubella kit?

A. Sirima, yes you are allowed to use any kit that is available in your market, you need to test the kit that you select (validity).

Claude Muller comment- the web-based option for PT would provide a framework to report data for each test, will help to record all the information, will also do that for all their testing. Bring all to a similar standard. The form could be printed out or put on computer to use as a result report form them have uniform reporting for results.

Every lab has own result reporting form or system. The form could have all the elements for information with every test. Could benefit some labs not doing this routinely.

Comments regarding dried blood samples:

Henda- agree with changes proposed, scoring and web submission, however dried blood samples, I think are not good because the process would be different from samples done routinely, and would be specific for PT.

David: same thing, and dried blood samples are validated for testing by Siemens only.

Lucky: same concerns for dried blood spots, shipments for dried blood vs dried serum, harsh conditions, would need to be verified.

Annick: yes same comment about dried blood samples.

Q. As to website for PT, we can use excel sheet using transition, then does that mean we are moving to internet? Africa has big challenge for internet connection, hope to keep spreadsheet for some countries, is it possible to have French version?

A. There can be considerations for slow internet, but our IT person said that transition as big as this, for first year to try the website, but if it is not feasible, should have option for excel spread sheet.

THE WHO/CDC MOLECULAR EQA PROGRAM, RESULTS OF THE 2014 SURVEY AND WAYS FORWARD

presented Paul Rota
The results of the first molecular PT panel were presented at 12th meeting of the GMRLN in Istanbul. In his presentation today, Paul Rota presented the results of the survey taken after the Istanbul meeting and described a plan for molecular PT testing in 2015. Based on the results of the survey, the 2015 proficiency testing will be similar to the testing in 2014 with a few exceptions. The number of laboratories will be expanded in 2015 and CDC will provide panels for SEAR, AMR, AFR and WPR with EUR participating in the PT panel provided by INSTAND. CDC anticipates shipping up to 50 panels in 2015 for distribution by regional laboratories. The composition of the panels (4 FTA punches for measles, 4 FTA punches for rubella), scoring criteria, and the feedback mechanism will be the same as in 2014. Panels should be distributed during November 2015 with all results completed by the end of January 2016.

MOLECULAR EQA EXPERIENCE – CHINA

was given by Zhen Zhu.

In China, panels for molecular PT are distributed to the provincial laboratories. Each panel contained 10 samples including 4 wild-type measles viruses, 4 wild-type rubella viruses, and 2 negative samples for both measles and rubella. The samples were lysates (0.5ml) of infected cells that had been inactivated at 56°C for 60 min. Each sample was transported to the provincial labs with dry ice by express courier (usually 1-2 days). The extracted RNA was tested with the assays (real-time RT-PCR, genotyping RT-PCR and sequencing) that are routinely used in the laboratory. Results were reported to NML by using the proficiency panel report form via e-mail. All the participants passed the 2014 molecular PT. Three different commercial real-time RT-PCR kits were used to detect measles/rubella in China and there was good correlation between the results from the different kits. Eleven provincial labs have the genotyping capability for measles and 7 provincial labs have genotyping capability for rubella. Real-time RT-PCR has been successfully carried out in the China Labnet.

MOLECULAR EQA EXPERIENCE – JAPAN

presented by Katsuhiko Komase

The PT samples, distributed on FTA cards, are similar in composition to the global panel. The panels were distributed to 22 Prefectural Institutes (PI). Most of the PIs have established highly sensitive, end-point RT-PCR methods, although RT-PCR protocols were modified at sites and 95% of PIs exceed the qualifying score. Results showed that the dual target PCR for measles improved sensitivity. There were some problems with genotyping including incomplete data, inaccurate alignments, outdated reference sequences and the use of BLAST to determine the genotype. Additional financial support is required maintain the EQA program, which results in an increased workload for NIID. EQA for real-time RT-PCR is planned.

NEW COLLABORATION BETWEEN WHO AND INSTAND E.V. ON MOLECULAR EQA FOR MEASLES AND RUBELLA

was given by Oliver Donoso Mantke.

INSTAND E.V. is a scientific society cooperating with national and international societies and organizations for the purpose of quality control including 65 programs in virology. INSTAND, in collaboration with the Robert Koch Institute, has been providing molecular EQA panels for measles and rubella since 2014 with analysis performed by Charité University Medicine, Berlin. The measles and rubella panels are similar to the panels distributed by CDC in that the samples are distributed on FTA cards. Results of pilot tests with the panels were
very good and a new panel was prepared in June 2015. This panel will be distributed to GMRLN laboratories in EUR.

Questions and comments:

Q. Kevin: were there cellular controls?

Paul: good point - some had cellular controls and some did not; the score was not affected. Recommend cellular controls and sources for different primer sets. Yes, need to scale up, as we move toward diagnosis, ask Japanese colleagues

Komase: I did not consider effect of RNA from serum.

Joe: kits that we send out have human DNA in them, distinction between them, RNA from cell line, rubella uses commercial source of human RNA.

Paul: multiplex kits in China- do these include cellular controls? Primer set for RNase P or actin, 2 kits do include RNAse P.

Songtao: This is an issue, RNase P control is negative and measles is positive, is that normal?

Most kits may deplete so cellular control may be negative.

Paul: Does everyone agree that we should do QA this year, proposed timeline in my slide, idea – rely on INSTAND program in Europe, Miriam to decide. CDC could supply to other countries, limited to what we can do this year. The compromise plan is to harmonize across regions. With the INSTAND program ahead of us, we can harmonize with them - strains reported and interpretation.

Can INSTAND prepare >70 panels? Financial considerations, shipment, by all RRLs.

Oliver: >70 samples not such a problem, management of external assessment, issues can be resolved, ask to continue with discussion, need other colleagues.

Q. (Said): for Oliver: can he give us the website to get details to see about participating as an institution.

A. Send an email to Oliver, or he can show this, presentation uploaded to Dropbox.

Q. (Joe) for Oliver: rubella tests- there are a number of tests that do not perform well with all genotypes, is the assessment part of this whether lab knows that the kit performs poorly with particular genotype? If we want to do diagnostics, may be lost on some. Have you seen any difference in stability of M R stability of RNA, assume it is the same?

Oliver: doing the stability testing internally to ensure quality of samples, always monitoring RNA - FTA samples from each phase of production, quite homogeneous.

A REPORT FROM RUBELLA IGG STANDARDIZATION WORKING GROUP

was given by Christelle Vauloup.

Several questions regarding testing and interpretation of rubella IgG are being investigated by the working group. These issues are particularly important for interpretation of rubella IgG results from pregnant women. It was demonstrated that the kinetics of IgG do not necessarily provide the means to distinguish between a
primary versus secondary infection. The presence or absence of IgM and avidity of IgG were found to accurately identify the type of response.

To address the question of whether a quantitative result for IgG would be helpful to define a protective titer, seronegative pregnant women were vaccinated following delivery. Analysis of the findings suggested that the current cut-off for IgG in international units (<10IU/ml) should be revised since lower levels are likely to be protective.

The other focus was on whether and to what degree results for IgG vary among kits. Substantial discrepancies between results obtained from 8 commercial ELISA assays were noted after a comparison study was conducted. Fifty-two percent of women considered susceptible with an ELISA assay had specific anti-E1 antibody detected by immunoblot.

Another finding suggested that equivocal results could be considered as positives and the cut-off can be reduced without losing specificity.

The epidemiologic situation of rubella in Finland, a country with very high vaccination coverage, shows decreased seroprevalence over time but no cases have occurred other than imported cases and no secondary cases have been detected.

Future plans include collection of a true IgG seronegative panel to evaluate kit sensitivity. Two articles describing these studies are complete and soon to be published, one additional article is in preparation.

SESSION 5: WORKING GROUP REPORTS

THE REPORT FROM THE WORKING GROUP ON THE 3\textsuperscript{rd} EDITION OF THE WHO LABORATORY MANUAL FOR THE DIAGNOSIS OF MEASLES AND RUBELLA – STATUS UPDATE

Was provided by Jenny Rota

The second edition of the Laboratory Manual was released in 2007. An updated version will be drafted by a consultant with input and feedback from the working group. The outline for the chapters and topics to be added has been developed. The manual will be distributed as a web-based document which will facilitate distribution in a timely manner.

Protocols, worksheets and training materials will be available as annexes that can be readily accessed and downloaded. Among the new chapters and content will be guidance for diagnostic challenges in near-elimination and elimination settings, and greatly expanded molecular methods. Another new chapter will focus on requirements for documentation and additional guidance for the verification of elimination of measles and rubella.

The goal is to have a draft of the third edition of the manual available for comments from the GMRLN by the end of 2015.
GUIDELINES ON THE USE OF SEROSURVEYS IN SUPPORT OF MEASLES AND RUBELLA ELIMINATION

were given by Ray Sanders.

The guidelines for serosurveys are envisioned to provide best practices for those involved in planning, performing, and analyzing serosurveys in support of measles and rubella elimination.

There are 5 primary sections with annexes that focus on the components of a serosurvey project. Each section contains advice that is important to consider to ensure the success and accurate interpretation of the data collected.

An advanced draft is now available, with revisions underway and a field test to be implemented among the next steps.

SEROSURVEILLANCE AND THE USE OF HIGH THROUGHPUT ASSAYS TO MEASURE IgG TO MEASLES AND RUBELLA

There were three presentations on this topic:

1: SEROPREVALENCE TESTING PROGRAM BY RIVM

Fiona van der Klis described the seroprevalence testing program used by RIVM in the Netherlands. It was emphasized that serosurveillance takes on even greater importance in settings of elimination since clinical surveillance is limited and monitoring vaccination coverage is critical. RIVM uses the Luminex platform technology to test for multiple antigens in a single reaction. RIVM has developed multiplex immunoassays (MIAs) including one for MMR and varicella.

Validation and population studies have been underway for a number of years. MIAs provide high throughput, reduced sample and reagent requirements as well as good correlation with ELISA. More studies are planned using other types of clinical samples including dried blood on filter paper.

2: BUILDING CAPACITY TO EVALUATE NEW SEROLOGIC ASSAYS IN THE WHO MEASLES AND RUBELLA LABORATORY NETWORK

Presented by Paul Rota.

This collaboration between RIVM, CDC and WHO/HQ will evaluate the measles-rubella microbead assay for use in seroprevalence studies by GMRLN laboratories. CDC has obtained reagents from RIVM and has set up the assay successfully. Scientists from RIVM are planning a visit to CDC in August 2015 to finalize the technology transfer. Comparison will be performed with a number of well-defined serum panels in late 2015.

3: MEASLES VIRUS NEUTRALIZATION: KEEP THE STANDARD ALIVE AND KICKING

Rob van Binnendijk described the need for a neutralization assay and the challenges with the standard WHO PRNT. Improvements and alternative methods for the neutralization assay were described along with observations of inconsistent behavior with the WHO III serum standard. Data was also presented from a
serologic study involving health care workers in the Netherlands which showed a good correlation between the Luminex microbead assay and PRN.

**POINT OF CARE TESTING FOR MEASLES IGM AND IGG – PROJECT UPDATE**

Presented by David Featherstone

The development of point-of-care test (POCT) for tetanus toxoid antibody and measles antibodies (IgM and IgG) initially focused on the redesign of the Oracol oral fluid collection device to allow the fluid to be collected, extracted and dispensed from one container.

The prototype collection device (Oralight) has been produced and studies are planned to assess sample quality. One study, scheduled for August 2015 in Rio, will assess sample quality of the oral fluid for measuring HIV antibody with serum using a locally produced POCT.

The design of the POCT for the project is a lateral flow device (LFD), which consists of a durable plastic case with a liquid port for addition of the oral fluid. Although the resulting band for the sample can be read visually, a portable device will be utilized for direct measurement of the band. In tests performed with Microimmune EIA and the LFD for measles IgG, sensitivity and specificity was 91%.

A trial batch of the measles IgM POCT are scheduled for release in July 2015. In August 2015 a study is planned in Rio to compare Siemens EIA and the measles IgM POCT using serum samples submitted for measles surveillance. Evaluation of oral fluid samples collected with Oralight and matched serum samples (Siemens EIA IgM) is planned for September 2015 in Ethiopia. Field trials for the tetanus and measles IgG POCTs are scheduled for October 2015 in Uganda.

**AN IgM AND IgG KIT COMPARISON STUDY**

By Kevin Brown

One of the recommendations from the 12th GMRLN meeting was to evaluate the performance of current IgM and IgG assays, particularly among cases of secondary vaccine failure and in low incidence settings where the positive predictive value is diminished. The format of the EIA assays have various advantages and disadvantages which should be included in the analysis. By far, the most data exists from proficiency panel testing for IgM with the Siemens kit, but other kits have been used and evaluated.

Data was presented that underscored the issues raised regarding the variability in rubella IgG results and poor correlation to the “immunity” cut-off of 120 IU/ml using different kits.

In conclusion, the GMRLN will need to elucidate the questions to be answered and the design of the comparison (type of samples, kits to be included). In addition, efforts to collect and share suitable samples will be required.

**Questions and comments:**

Comment: For vaccination vs wild-type serum, can’t distinguish between immunity stemming from vaccination and that from natural disease using serum.

Q. (to David, POCT), how long does it take to get a result- and what is the cost?
A. 15-20 minutes. Cost: Gates target is $1 a test, but probably closer to $6.

Q. Do we need to stick to the 10IU/ml cut-off for rubella IgG?

A. (Joe) people develop immunity by different means, can be vaccinated multiple times and have very low IgG but are clearly immune, they clear the vaccine virus. A rigid cut-off is a mistake, subtleties are important.

Comment (Joe): maternal antibody is active, pulls IgG into fetus, variable from person to person, some gone by 3 months, some there for 10 months. A lot of variability, does not precisely decline.

A REPORT FROM THE EXTENDED AND WHOLE GENOME AND NEXT GENERATION SEQUENCING (WGEWNGS) WORKING GROUP

was given by Alberto Severini.

As elimination proceeds, the diversity of the genome diminishes. A recommendation from the 12th GRMLN was to form a WG to explore a more discriminating method/target to improve ability to track transmission of measles and rubella. The targets to increase resolution include M/F and P for measles.

Other goals for the group are to determine how much variation is needed to exclude direct transmission between two measles cases and to develop a practical method to obtain WGS directly from the patients’ specimens.

For rubella, Joe at CDC has done much. The scope of our work is to facilitate sharing of isolates. A pipeline for rubella whole genome sequencing using Illumina Miseq has been developed. There were 3 teleconferences starting in March 2015 to discuss the terms of reference (TOR). In order to produce a global view, all the isolates need to be available. One of the TOR was to develop a method to culture rubella strains and to support the establishment of a rubella strain culture library.

BREAKOUT GROUPS

met to discuss plans for each of the regional laboratory networks. A representative from each group presented a summary of the meeting. A list of participants for each group has been included with the electronic files for the meeting.

AMR: MAIN ISSUES WERE PRESENTED BY MARILDA SIQUIERA.

1. 1. AMR is planning 6-8 on site visits to laboratories this year. Members of the team volunteered to assist with onsite visits
2. 2. Availability of kits- not all countries are using the Siemens kit. Need to determine which kits are been used in PAHO. NML, Canada can do comparative study depending on sera panel available
3. 3. Trainings and workshops:
   a. Individual trainings at RRL or GRL; onsite training
4. 4. Improved molecular surveillance; members discussed the need for dissemination of sequencing capacity and decided it is not necessary (since now in elimination phase)
5. 5. Proposed AMR LabNet meeting in 2016; suggestions for agenda were made:
   a. Committee or WG for case classification in elimination setting (national and international)
   b. Review and discuss the following topics:
      a) Algorithm for case classification versus analysis of individual case
b) Measles case definition versus rash and fever

c) Review referral system and improve interactions between labs

d) Structure of laboratory network in the region

Questions and comments:

Q. Mick: I did not understand the last item. RL and GL- just to say that these labs should interact with NL more actively.

Marilda: this year, there were 165 cases in Brazil, we review each one by telephone.

Joe: case classification is delayed by months sometimes when the samples are referred, idea to do more quickly, when there is a delay of 3-4 months, it is hard to collect additional information.

**AFR: MAIN ISSUES WERE PRESENTED BY ANNICK DOSSEH**

1. Measles and rubella kits
   - What is the distribution mechanism to avoid kits expiring?
   - How to avoid having all labs receiving deliveries with same expiration date?

2. Specimens for measles virus isolation/characterization
   - How to get specimens provided to RRL?

3. Virus isolation
   - Where to get new batch of Vero/hSLAM cells
   - Outbreak investigations

4. Rubella genotype (and implicitly Measles)
   - How to increase detection of circulating genotypes?
   - How to work with field to maximize opportunities for enhanced quality of specimens

5. Sub-national lab in Ethiopia
   - What is the optimum frequency of supervisory visit from NL?
   - How to perform quarterly confirmatory testing and annual PT

6. Human resources: data manager at NICD has left the job

7. Sequencing reagents & supplies
   - Delay in performing testing due to lack of supplies & reagents

8. 2015 planned trainings
   1. Molecular and sequencing workshop at UVRI for selected countries
   2. Onsite follow up support for sequencing at IPCIV

**Recommendations**

1. Measles & rubella kits
   - Link with lab coordinator(s) to re-distribute kits close to expiry date to needy labs
   - Avoid placing order for all labs at the same time or have them delivered at different times to spread out expiry dates?

2. Specimens for measles/rubella virus isolation/characterization
   - Work with field staff including training of surveillance teams
   - Work with NLS with capacity for isolation
   - Work with countries to improve quality of specimens sent for isolation

3. Vero/hSLAM cells for virus isolation
   - Get in touch with Hong Kong lab and check if provision of VERO/hSLAM is possible
   - Check with UVRI for distribution to some needy laboratories
4. Sub-national lab in Ethiopia
   - NL to perform quarterly supervisory visit to SNL the first year and then reduce frequency as necessary
   - Annual PT and quarterly shipment of QC specimens to NL to be done
   - Provide financial admin support from MoH/WHO

5. Human resources
   - Onsite training on data manager can be performed by IST/ESA
   - NICD to make official received

6. Rubella genotype
   - CDC planned research studies on specimen management; to be processed through AFRO

7. Molecular workshop at UVRI and follow up with IPCIV
   - Consider back to back (or separate visits if facilitators mobilized)
   - Finalize appropriate timing
   - Compile and share list of material needs early (training)

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**EMR: MAIN ISSUES WERE PRESENTED BY HINDA AHMED**

A. Laboratory Capacity
   1. RRL and labs in elimination phase to introduce additional diagnostic tools
      - For RRL, new sequencer
      - Avidity kits and IgG kits, (in addition to PCR)
   2. Development of testing algorithm for elimination phase
   3. Enhance rubella detection and genotyping; pilot new target developed by CDC: real time RT-PCR and sequencing
   4. Enhancing detection and characterization of circulating measles and rubella viruses
   5. Revise the surveillance guidelines to highlight at first patient contact collect both blood and throat swab/urine sample
   6. Establishment of subnational lab in Pakistan
   7. Training of new diagnostic tools and new staff due to staff turn over

B. Quality Control
   1. Implement both onsite visit for accreditation review and online submission
      - 2015 Q3 & Q4 onsite visit (Iran, SA, UAE and Tunisia), Online submission (Iraq, Palestine and Syria)
   2. Follow-up implementation of the new scoring of PT panel
   3. Expansion of mEQC: countries (Bahrain, Egypt, Lebanon, Iraq, Iran, Morocco, Pakistan, Oman, Tunisia)
   4. Ship specimens for test validation and genotyping to RRLs
   5. Annual EMR LabNet meeting and continue with EPI and surveillance 26-30 October, 2015.

C. Supplies
   1. Timely procurement of necessary laboratory supplies
      - Keeping enough stock to avoid stock-out
      - Shipment and distribution of lab supplies, PT panels and Vero/SLAM cell line
   2. Stringent follow-up of timely reporting of PTs, genotypes to MeaNS and RubeNS
   3. Strengthening laboratory data management
   4. Proper filing of laboratory data to support documentation of measles and rubella elimination

D. Funds
   1. Advocacy for resource mobilization; highlighting importance of lab surveillance for M and R elimination and data needs for verification
2. Utilization of available lab facilities
3. Report, prepare publications using the substantial amount of country laboratory data.
   - Funds required: minimum 80,000USD

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**EUR: MAIN ISSUES WERE PRESENTED BY MYRIAM BEN MAMOU**

1. Roll out of 2015 molecular PT for the European region
2. 2. Invite NRLs performing routine molecular testing to enroll in INSTAND molecular PT (Nov 2015 round)
   - NRLs to pay for the fees, WHO to support labs in need
   - INSTAND to expand protocols to get full set of results
   - Continue collaboration to harmonize INSTAND and CDC molecular PT in coming rounds
3. Operational arrangements for 2015 serology PT
   - PT panels to be distributed in September 2015 to RRLs
   - All labs to be encouraged to report online through VIDRL website
   - RRL Moscow to distribute to RUS / NIS during workshops and meetings Q4 2015- early 2016 and provide support to RUS SNLs for reporting results online
   - EUR Labnet strongly recommends to continue liquid serum panels (no DBS)
4. IgM retesting in 2015
   - RRLs to apply more strict conditions on data to be provided by NRLs
   - Feed-back received from RRLs on form and instructions
   - RLC to consolidate and share a harmonized form and instructions for final RRL/GSL review
5. Internal Quality Control procedures: training to be provided to all labs in 2016
6. Position of European MR Labnet on serosurveys in the verification context
   - The epidemiological situation including immunization gaps is well known in many countries, as well as priority interventions needed to fill immunity gaps
   - When evidence is lacking and cannot be provided by other sources, serosurveys can be an option to provide information
   - For that, the survey should answer a clear question tailored to a specific country context
   - Study design, protocol and lab tools to be adapted to the question
   - In resource limited settings, conducting serosurveys may compromise interventions having more impact on MR situation
   - Need to refer to global seroprevalence guidelines
7. Role of the WHO-accredited NRL vs other laboratories involved in MR testing
   - Laboratory data in national verification reports (annual country updates) should come from WHO-accredited lab and/or proficient labs
   - It is recommended that NRLs contribute to the annual verification report not only with their data, but also in facilitating information collection regarding other labs, and to document their proficiency (EQA / national accreditation)
   - When appropriate, the role of NRLs in supervising and/or coordinating other labs performing MR testing should be encouraged and formalized at country level
8. IgM Testing
   - Extend the review of serology PT data to rubella (GSL)
   - Recommend approved/valid ELISA kits based on serology PT data analysis
   - No need to conduct extensive ELISA comparison studies at this stage, as serology PT data review provides enough orientation

**Meetings**

RRL meeting: 1st or 2nd week of March 2016, Berlin
NRL meetings: 1st or 2nd week of September 2016, Montenegro

- English speaking countries / Russian speaking countries back to back
- Montenegro lab to be invited as observer
- Trainings: MRLDMS, IQC
- Opportunity to establish collaborations with NRLs for research studies

Accreditation visits as planned in 2015 (KAZ, POL, GEO, RUS); suggestions solicited for 2016.

**SEAR: MAIN ISSUES WAS PRESENTED BY SIRIMA PATTAMADILOK.**

1. 2015 ITAG recommendations related to the MR laboratory network
   - Timor Leste should enhance its current laboratory to ‘proficient’ status in order to support case-based surveillance.
   - All countries should collect adequate specimens to characterize measles and rubella genotypes and share information in a timely fashion. These data should be linked to epi case-based data to enable identification of chains of transmission
   - WHO proficient or nationally recognized laboratories compliant with WHO standards support CRS surveillance
   - ITAG recommends that by the end of 2016 establishing virologic surveillance for verifying interruption of indigenous transmission and identifying imported and import-related cases
   - Measles virus genotypes should be characterized in at least 80% of chains of transmission

2. Capacity building plan 2015
   - Regional Measles/Rubella Laboratory Network Workshop to Strengthen CRS Surveillance, October 26-30, 2015
   - Inter-country workshop on capacity building in measles and rubella lab accreditation, November 16-20, 2015
   - Onsite training for establishing molecular laboratory in Sri Lanka, Bangladesh, Bhutan and Myanmar
   - Provided sequencing training for NIV, Pune in February 2015

**WPR: MAIN ISSUES WERE PRESENTED BY YAN ZHANG.**

*Challenges*

1. Completeness: for MS with/close to measles elimination
   - Laboratories should attempt to obtain genetic information from at least 80% of chains of transmission
   - The national report should include the percentage of chains of transmission with genotype
2. Rubella and CRS surveillance, virologic surveillance should be strengthened.
3. Propose an outbreak management plan in priority country: work with EPI for the contingency planning
   - Outbreak management and response
   - Sample collection strategies
   - Use epi-link for the following case classification after the lab confirmation for the outbreak
   - Optimize the lab test algorithm
   - Maintain adequate supplies/reagents for continued testing
   - Provide additional manpower to manage its operations
   - Budgetary

*Priority needs*
1. Timeliness
   • Some countries should reach the new target indicator of timeliness of reporting (80% within 4 days)
   • Labs in countries experiencing large outbreaks should not be penalized for not meeting the 4 days turnaround but should communicate any timeliness issues with the Regional Lab Coordinator as soon as possible (PHL, VNM, PNG)

2. Confirmatory testing
   • All laboratories are requested to participate
   • Development of SOP to define concordance among different RRLs (HK and VIDRL)

3. Communication and coordination
   • Between regional Lab coordinator, national lab and RRL
   • Facilitate the process of confirmatory testing: send a draft sample list to regional coordinator and RRL before shipment for confirmation
   • Arranged scheduled shipment of specimens
   • National labs are suggested to provide feedback to RRL and regional coordinators for the follow up of discordant results
   • Relationship between private lab and national lab
   • Encourage NL to continue QA of the private labs
   • Recommendation from WHO to provide the framework to country about sharing data from private labs

Plans for 2015-2016

Regional LabNet meeting- implemented in May 2015

Molecular training course in HK lab in 29 Feb to 4 Mar 2016; 14 participants from 10 MS (new and experienced staff)

Measles and Rubella:
   • Sequence phylogenetic analysis
   • Match the sequences to named lineage in MeaNS to help identify transmission chains and to support implementation of the annual report of elimination verification

Accreditation:
   • By correspondence will be encouraged in the future
   • Onsite review: in priority countries and those which have not undergone an onsite review for 3 or more years
   • Facilitate to organize the workshop and training for subnational laboratories-China, PHL and VNM, if needed
   • Provide technical support for Mongolia outbreak

Recommendations for the Meeting
   • Laboratories should make sure that laboratory data are properly represented in the annual national reports to the RVC
   • Countries experiencing large outbreaks should follow the surveillance guidelines and use the mechanisms for epi-linking cases after each chain of transmission has been lab confirmed
   • National labs should ensure that their Country’s national surveillance/outbreak response plans include a contingency plan for sampling strategies in the event of a large outbreak to ensure the number of samples collected do not overwhelm the capacity of the lab
A SUMMARY OF THE WORKSHOP ON THE MEASLES AND RUBELLA ELIMINATION VERIFICATION PROCESS HELD IN SIEGES, SPAIN ON JUNE 26, 2015

Was presented by Alya Dabbah

The WHO framework for verification elimination was presented, including the three essential criteria and five supporting lines of evidence. Laboratory-related indicators (all with targets of ≥ 80%):

- The proportion of suspected cases with adequate specimens for detecting acute M or R infection collected and tested in a proficient lab
- Proportion of lab-confirmed chains of transmission with samples adequate for detecting measles or rubella virus collected and tested in an accredited lab
- Proportion of specimens received at the lab within 5 days of collection
- Proportion of results reported within 4 days of specimen receipt

The workshop was attended by RVC members from several regions along with experts on the verification process. Regional progress toward verification was reviewed with the aim of improving the verification framework. The participants agreed that the RVCs in different regions are aligned, had similar Terms of Reference and indicators, although there were some differences across regions. Country by country verification will occur in EURO and WPRO, while the other regions will verify as a region.

Questions and comments:

Q. Joe: regarding the limitations listed for rubella...why did the recommendations not mention rubella?
A. This is a draft, need to circulate issues and draft, excellent point.

Q. Annick: was AFR represented?
A. Yes, regional, no one from EMR, although potential members participated.

Comment: (Paul) working with WPR, interesting process, RVC, variations in reports, some detailed, some not, pay attention to lab data, need to give guidance to NL so to structure the data to VC in standardized manner, as much as possible, some need to incorporate this into workshops, this data needs to be organized, 5th line of evidence in particular, prompted theme for next session, indicators have private labs, RVCs willing to look at alternative indicators.

Comment (Hinda): One indicator- shipment to within 5 days of collection- labs cannot have any say in that, needs to be discussed, criteria, circumstances, some countries have lots of problems.

LABORATORY INVOLVEMENT IN VERIFICATION OF MEASLES ELIMINATION:
VERIFICATION QUALITY MOLECULAR SURVEILLANCE

presented by Paul Rota

The need to improve the analysis of genotype information and completeness of viral surveillance was highlighted. Laboratories preparing data summaries in support of measles elimination are encouraged to perform phylogenetic analysis of the sequences from all cases that have been genotyped and to match the sequences to named lineages ‘named strains’ in MeaNS to help identify transmission chains and potential sources of importation.
Laboratories were reminded that although the current recommendation is to obtain adequate samples for virologic detection from at least 80% of outbreaks, laboratories should attempt to obtain genetic information from at least 80% of chains of transmission and include percentage of chains of transmission with the list of genotypes in the report to the NVC.

Several laboratories were invited to give brief presentations of verification quality molecular surveillance including Japan, Hong Kong, Russia and Thailand. The presentations gave excellent examples of high quality virologic surveillance and served to demonstrate some of the challenges with using genotyping data to verify measles elimination.

Vicki Stambos described a measles vaccine-specific real-time RT-PCR that was developed at VIDRL. During 2014, parallel testing was performed for 20 samples collected from vaccine-associated cases. Of these, 12 that were sequenced as genotype A were positive using the measles A/vaccine real-time RT-PCR. Seven samples were negative by both assays (6 of which were genotype B3). One sample that was positive for measles A/vaccine by RT-PCR could not be sequenced (was genotype PCR negative due to a low Ct). These data are being compiled for publication.

Questions and comments:

Q. Alberto: have you tried to multiplex?
A. Vicki: not sure.

Q. Kevin: have you tried mixed wild-type and vaccine?
A. Vicki: Used all of last year, compiling for publication.

Q. Joe: robustness, or copy number dependent on vaccine-specific assay? Variation in copy number?
A. Vicki will get back to you, Thomas needs to address these questions.

Comment Paul: vaccine-associated cases tend to have higher Ct; this is subjective, but probably lower copy numbers, harder to genotype.

Sabine: About the vaccine sequences, are there consensus sequences on Genbank?

Kevin: back to other talks...point about named strains, assumption that the named strain is the source, is the first one that we could find in MeaNS, does not imply that it is the source.

A RUBELLA VIRUS GENOTYPE FROM A LONG TERM INFECTION

Joe Icenogle moderated a session on rubella genotyping and began the session with a presentation entitled:

The recommendations for increased emphasis on rubella virus genotyping in LabNet, going back for about 7 years, have not been effective in many countries. He proposed that a recommendation on rubella virus genotyping could be made more specific. The two suggestions were that countries with endemic rubella would obtain at least one rubella virus genotype determination; and at least one rubella virus genotype would be determined for each 2000 rubella cases reported by a country, to a maximum requirement of 3 genotypes. In this way, rubella genotyping would be a surveillance indicator.

Dr. Icenogle then gave an update on a rubella virus genotype from a long term infection from a case of Fuchs’ Uveitis Syndrome (FUS). Clinical signs of FUS are chronic inflammation of the middle layer of the eye which includes the iris, heterochromia (different iris color of the two eyes), and keratic precipitates, cataracts.
There is substantial evidence that rubella is associated with FUS. Studies have shown that a high proportion of FUS patients have antibodies directed against rubella virus and/or rubella virus RNA in their aqueous humor. An epidemiological study in the US correlated a recent sharp decline in the prevalence of FUS with the introduction of a population-wide rubella virus vaccination program.

The patient remembers having heterochromia starting at age 14, but does not remember having rubella as a child. Clinical samples were received at CDC. The results with E1 real-time RT-PCR were 1/3 positive (Ct=39). With real-time RT-PCR using primers and probe from the more highly conserved 5' end of the genome (Hubbschen et al., JVM, 2008), 2/3 replicates were positive (Ct=36.5). The 739 nt sequence window was amplified by 2 nested sets. The FUS sequence appears as a new branch with the highest similarity to genotypes 2C and 1a.

Findings support the association between rubella virus and FUS. Phylogenetic analysis provided evidence that this rubella virus was likely a previously undetected genotype which is no longer circulating. Since the patient had rubella prior to 1955, this sequence is from the earliest rubella virus yet characterized.

RUBELLA RT-PCR AND GENOTYPING FROM SERA – HONG KONG

By Janice Lo

Since 2008, 563 serum samples were tested for rubella RNA and 109 (19.4%) were positive. Genotypes detected were 1a, 1E, 1j and 2B. The primers were improved in late 2012 and 9/15 rubella PCR positive samples, which were negative with the old genotyping protocol, were positive with the new protocol (all except one yielded complete genotyping window). All 11 samples previously partially positive with old genotyping primer sets were positive with the new protocol, with complete genotyping results. Challenges include insufficient serum volume for RNA extraction, which may affect the RT-PCR positive rate, and low viral load in serum samples.

CHALLENGES IN OBTAINING RUBELLA GENOTYPES IN THE EUR.

was presented by Myriam Ben Mamou.

It was noted that insufficient sensitivity of rubella and CRS surveillance systems exist. In many countries, the rubella surveillance is passive and functions as a by-product of measles surveillance (e.g. measles IgM-negative sera are tested for rubella). NRLs frequently report antenatal screening of rubella immunity, but CRS surveillance is less developed in the region. In addition, case definitions may vary between countries. There is an extremely low rubella laboratory investigation rate of rubella suspected cases for rubella. There are also technical issues regarding clinical samples for RT PCR and genotyping issues.

AN UPDATE OF RUBELLA GENOTYPING IN SEAR

was presented by Patcha Incomserb.

In the last two years, genotype 2B was reported in Thailand and Bangladesh. Prior to 2014, genotypes 2B and 1E were detected in 6 countries.
Richard Myers from PHE presented

Mick Mulders circulated a link to a questionnaire about MeaNS and RubeNS earlier in the year and there were 44 respondents including 37 who used MeaNS and 17 who used RubeNS. The main findings related to low utilization were that there was no measles or rubella in the country, the laboratory does not perform sequencing, and lack of training or non-user-friendly issues. Features that are not available that were noted were password management (MeaNS), bulk upload capacity, lists of publications, and mapping tools.

MeaNS currently has 25,531 viral sequences and RubeNS has 1,241. Unfortunately, there continues to be submission of MeV vaccine sequences to MeaNS (~22 since the start of 2014). As this is not what MeaNS was designed for, laboratories are reminded not to submit vaccine sequences.

MeaNS has the facility for RRLs to submit sequences where a complete WHO name cannot be formed. This submission route requires country and year information only (e.g. MVs/RRLGBR/2015/2). WHO offices, GSLs and RRLs are now getting a weekly email from MeaNS and RubeNS that contains all new sequences submitted in that week.

Analysis of the genotype data in MeaNS appears to show a reduction in the number of genotypes. At the regional level, genotypes D8 and B3 are predominant for most regions though genotype H1 skews the distribution in WPR. However, some regions are under-reporting and diversity is not being recorded.

The MeaNS/RubeNS steering committees and the WGS group should consider that submitting whole genome sequences (WGS) is feasible, but WGS quality, depth, coverage will need to be considered. Widening the sequencing window allows better characterization of MeV outbreaks although N-450 is still the best approach in low-resource and endemic transmission settings. The greatest amount of detail can be obtained from WGS, but this is expensive and time-consuming. Analysis of the sequences of the M/F non-coding region may complement N-450 in well-resourced countries approaching elimination.

**Questions and comments:**

Q. Is there a decrease in sensitivity compared to N-450?
A. Only 5’ of noncoding region. Non-coding regions have a lower copy number.

Kevin: we are obtaining sequences from clinical samples- oral fluid. It is sometimes necessary to use 2 fragments. An advantage to N-450 is the high copy number.

Q. Lucky: not able to understand which lab, list of submissions? MeaNS has tools to make it easier for RRL to know what is happening, summary data for region. Is it possible to get line listing of all you have submitted, use for verification committee? To have some tables like shown earlier?
A. Richard: should be getting a line listing of samples rather than aggregate data by email.
SHORT-Term GOALS

- Ensure that all countries have access to verification quality laboratory support for surveillance for measles and rubella.
- Strengthen and maintain capacity among all NLs so that labs have the tools needed to support verification of elimination of measles and rubella including genotyping capacity or advocate for GMRLN partners to provide support for molecular surveillance activities.
- The availability of high quality laboratory data will be assured through EQA and annual accreditation as well support from WHO/HQ and partners to provide GMRLN staff with training in laboratory quality management.
- WHO/HQ will work with RLCs and NLs to identify and surmount barriers to the timeliness and completeness of reporting to meeting the established targets for these indicators.
- Laboratories will have the capacity to provide effective laboratory support during increased workload resulting from outbreaks.
- The GMRLN will continue to expand and improve quality control and quality assurance within the network.
- The GMRLN will continue to build capacity of laboratories to perform both serological and molecular methods.

LONGER TERM GOALS

- Fully integrate testing for measles and rubella within the GMRLN and include testing for other VPDs as required by national programs.
- The 3rd edition of the Laboratory Manual, with updated laboratory protocols, and electronic training materials will be available as web-based documents.
- Results from all EQA testing will be reported via a web interface.
- Laboratories will have access to a full range of laboratory methods needed for case classification/confirmation in pre and post elimination settings.
- All laboratories will routinely report to MeaNS and RubeNS. The GMRLN will have resources to fully support and expand the capacity and functionality of MeaNS and RubeNS.
- Laboratory and case-based surveillance data for suspected cases of measles and rubella (CRS?) will be linkable at the national level and reported on a weekly or monthly basis
- All laboratories will have the training and capacity to analyze sequence information from circulating strains of measles and rubella and to incorporate this information into national reports.

SPECIFIC RECOMMENDATIONS FOR 2015-2016
SURVEILLANCE

1. Timely reporting of results from laboratory testing for measles and rubella is essential for high quality surveillance. Most WHO Regions have adopted a maximum turn-around-time of 4 days, the time needed to report results of serologic testing (IgM) after receipt of the clinical specimen. For countries with endemic disease or those experiencing large outbreaks, reporting may be extended to 7 days. (Mick, RLCs)

2. Integration of case-based surveillance data with laboratory data is critical to improve the completeness and quality of surveillance. Efforts to improve communication and cooperation between laboratory and surveillance program staff are encouraged. Communication through channels such as, face to face, e-mail or by phone on a weekly basis (or suitable interval) to review cases and share information should be delegated to appropriate staff and implemented as a routine duty. (Robert, Mick, Marta)

3. Laboratory and case based surveillance data are currently reported on a monthly basis but the databases are not linked. The GMRLN should work with WHO/HQ and the Regional Offices to explore and implement methods to link laboratory data with case-based data. (Marta)

4. An indicator for adequate surveillance is testing at least 2 non-measles, non-rubella samples for every 100,000 population. In countries where a large proportion of the testing is performed by private commercial clinics or laboratories, RLCs and NLs should identify the major non-network laboratories that provide measles and rubella IgM testing and initiate efforts to obtain data regarding the number of IgM tests performed from suspected cases of measles and rubella. (Makoto)

5. The GMRLN should encourage and support countries introducing rubella vaccination by enhancing where needed, laboratory-based surveillance, laboratory testing and genotyping, as well as establishing or improving CRS surveillance. (Joe)

6. Laboratories in countries that have achieved elimination of measles and/or rubella are reminded that cases and outbreaks can still occur even though the country has achieved elimination. Laboratories must remain vigilant and be prepared to perform testing on samples from suspected cases. (Pablo)

7. The GMRLN will develop guidance for case confirmation and offer algorithms for discarding cases in near or post elimination settings which will be included in the next, third edition of the Laboratory Manual (Laboratory Manual WG)

8. The GMRLN recognizes that the use of RT-PCR for case confirmation must include caveats for the interpretation of negative results. Guidance for interpretation in different epidemiologic settings will be provided in the new edition of the laboratory manual.

9. Countries experiencing large outbreaks should utilize epidemiologic links to confirm cases once laboratory confirmation has been provided to avoid overwhelming the laboratory. National laboratories should ensure that their country’s national surveillance and outbreak response plan include strategies for laboratory testing that are appropriate for surveillance needs and do not result in unnecessary testing. The plan should also address surge capacity for assisting the NL when large numbers of samples need to be tested.

10. Because protracted outbreaks may involve unrecognized introductions of other genetic variants of the virus or even different genotypes, molecular testing of additional samples at appropriate intervals during an ongoing outbreak is recommended. The GMRLN will develop guidance for appropriately timed laboratory testing during outbreaks which will be included in the next, third edition of the Laboratory Manual. (Lab Manual WG)

SEROSURVEYS

11. In many countries, the epidemiologic situation including vaccination coverage are well known. However, when information is not available and cannot be obtained by other sources, serosurveys can help to identify immunity gaps and evaluate vaccination coverage. It is important that serosurveys are planned and conducted to assure that survey data addresses specific questions and the activities required to
conduct the survey do not present a burden to vaccination programs or laboratory activities. WHO/HQ has formed a working group to develop guidelines for conducting serosurveys and a draft version of the guidelines will be circulated in 2015. A pilot study to evaluate the guidelines will be conducted in 2016. Implications of the studies for the GMRLN will be included in the new edition of the Laboratory Manual. (Ray, Mick, Jim, Paul)

12. The GMRLN should continue to explore the use of alternative methods for assessing seroprevalence other than conventional/commercial EIA, such as multiplexed immunoassay (MIA) based on Luminex® technology, high throughput neutralization assays, and point of care assays. (Rob, Fiona, Paul)

QA/QC

13. RLCs should ensure that the appropriate number and representativeness of serum samples are sent from NLs to RRLs for confirmatory (quality control) testing of IgM results.

14. The molecular External Quality Control program that was initiated in 2014 by CDC on behalf of WHO will be continued and expanded in 2015 to all network laboratories performing molecular testing on a routine basis. The Measles and Rubella GSLs at CDC will produce panels for all regions except EUR which will use a semi-commercial panel. (Paul)

15. The participants agreed that the new scoring criteria for the serologic proficiency testing panels should be adopted, and the web interface for reporting results from proficiency testing to VIDRL should be rolled out in 2015 for evaluation. (Vicki)

16. The GLC and RLCs will ensure that all GMRLN laboratories are accredited annually either on-site or by desk review. (Mick, RLCs)

17. The GMRLN recommends the routine use of internal and in house controls in measles and rubella IgM EIAs. (Mick)

18. The GMRLN recommends that all laboratories routinely performing measles and rubella RT-PCR assays to detect RNA include controls to detect a reference gene to help assess sample specimen quality and to interpret results. (Kevin)

19. There is no need to conduct extensive comparison studies on EIA test kits used in the GMRLN. A comprehensive review of the data obtained from the serologic proficiency panels for measles IgM provided valuable information on the performance of the commercial IgM test kits used in the GMRLN and is adequate for making recommendations. This analysis should be now applied to data from rubella IgM testing. (Kevin, Mick)

MOLECULAR SURVEILLANCE, DETECTION AND CHARACTERIZATION OF MEASLES AND RUBELLA VIRUSES

20. All sequence data presented by laboratories at GMRNL13 must be submitted to MeaNS or RubeNS by September 1, 2015 and future sequence data generated for surveillance should be submitted in a timely manner and meeting the accreditation timeliness and completeness requirements.

21. All RRLs and selected NLs should have capacity for sequencing, sequence analysis and reporting to MeaNS and RubeNS. NLs without sequencing capacity should have a plan for referral of samples to a designated supervisory network lab (GSL, RRL or NL), and have an agreement about who will submit sequence data to MeaNS and RubeNS.

22. As all Member States from all Regions have adopted the goal for measles elimination and rubella control or elimination, Member States are requested to report timely and complete sequence data to WHO. Member States that are not currently submitting sequence data to MeaNS and RubeNS because of
national restrictions should work with WHO/HQ, the RLCs and national program staff to obtain official permission for timely reporting of sequence information for surveillance purposes to the global databases.

23. The GMRLN has formed a working group to explore the utility of extended sequencing windows and whole genome sequencing based on conventional Sanger sequencing or next generation methods for increasing the resolution of molecular epidemiologic studies. The outcomes of the findings of the working group should be used to guide future efforts of the GMRLN to evaluate the usefulness of these methods for the Global Measles Rubella Program by enhanced molecular epidemiology. The working group plans to have its first meeting in 2016. (Alberto)

24. To provide high quality molecular surveillance for measles and rubella, the genotype should be identified from at least 80% of all outbreaks.

25. In measles and rubella endemic countries, at least one genotype per year should be obtained for molecular surveillance. (Joe, Jim, Charles)

26. GMRLN laboratories are encouraged to use FTA cards as an alternative method to transport clinical samples and viral isolates for genotyping for molecular studies if transportation under reverse cold chain conditions is difficult to achieve. The protocol will be made available and will be included in the new edition of the Laboratory Manual. (Paul, Patcha)

27. Sequences obtained from serum samples that were IgM positive for measles have been valuable in expanding the sequence database in countries where samples for virologic detection are not collected routinely. GMRLN laboratories are encouraged to submit rubella IgM positive serum samples to RRLs for RT-PCR and genotyping to expand the rubella sequence database. (Annette)

28. Laboratories with sufficient capacity are encouraged to attempt to isolate measles and rubella viruses in cell culture from epidemiologically important cases. The successful isolation in culture of these viruses can be noted, along with the sequences submitted to MeaNS and RubeNS. This information will serve as a “virtual strain bank” and viral isolates will be forwarded upon request to GMRLN laboratories for whole genome sequencing or for inclusion in molecular proficiency test panels. (Kevin)

VERIFICATION OF ELIMINATION

29. Two of the five lines of evidence that support measles or rubella elimination are dependent upon the laboratory. High quality lab data and genotyping are necessary elements in confirming the absence of an endemic genotype. Laboratories should work with the National Verification Committees and Regional Verification Committees along with RLCs to provide complete laboratory data. A standard template should be adopted to include quality control and timeliness indicators, case based laboratory results, serosurveys, and genotyping information. (Paul, Alya)

30. Laboratories should utilize MeaNS and RubeNS to track genotypes and lineages within countries. For measles, laboratories should link sequences to the “named strains” listed on MeaNS so that sequence data can be compared between countries in the same region and globally. (Paul)

31. Laboratories should work with their National Verification Committees and RLCs to participate in the revision of the WHO framework for verification. (Paul, Alya)

CAPACITY BUILDING, COORDINATION, COMMUNICATION

32. Participation in activities that maintain the capacity required to perform high quality laboratory testing in support of global measles, rubella, and CRS surveillance is important for all members of the GRMLN. These activities include regional and national laboratory workshops and meetings, individual training visits, site visits, and consultations. RLCs should consider interregional training workshops to foster collaboration between regions. (RLCs)
33. All laboratory training activities should include a plan for follow-up with the participants/trainees to assess the effectiveness and utility of the training and to improve future training. (Paul)

34. The GMRLN should continue to explore all options for obtaining additional funding for critical laboratory activities. (Mick)

35. The GMRLN will post a new edition of the Laboratory Manual (web-based) in 2016. All GMRLN members are encouraged to provide feedback on drafts of the manual and to contribute protocols. After the new edition is posted, all laboratories are responsible for understanding the contents of the manual and for sharing the manual with members of their national surveillance program. (Lab manual WG)