



## Measles and Rubella genotyping workshop

### México D.F., México, September 11<sup>th</sup> to 15<sup>th</sup>, 2023

#### Agenda (draft)

**Objective:**

Strengthen the capacities of national measles-rubella laboratories to carry out genotyping (sequencing and analysis of sequences) of measles and rubella viruses in accordance with the technical procedures defined in the Global Measles and Rubella Laboratory Network (GMRLN).

**Participants:**

Responsible of measles-rubella laboratories and sequencing laboratories in five countries of the region.

**Facilitators:**

The workshop is leading for technical experts from USA-CDC, staff of PAHO will provide support to the training.

#### DAY 1 – SEPTEMBER 11<sup>th</sup> (Monday)

<i>Time</i>	<i>Activity / Presentation</i>	<i>Responsible</i>
	Welcome and opening remarks	PAHO
	Introduction of participants and administrative announcements	Participants
8:00 - 9:30	Training objectives	PAHO
	Pre-Assessment	Participants
	Coffee break	
	Introduction to the laboratory facilities	InDRE
9:30 - 11:00	Situation of the measles and rubella elimination	PAHO-WDC
	Testing scheme and workplan	CDC

#### Session 1: Genotyping RT-PCR

11:00 - 12:00	Lecture: MV and RV genotyping RT-PCR	CDC
	Discussion regarding aliquoting kit primers, controls for genotyping kits	CDC
12:00 - 13:00	Lunch	
13:00 - 17:00	Practical 1: Setting up PCR for measles genotyping	Half participants
	Practical 2: Setting up PCR for rubella fragment genotyping	Half participants



**DAY 2 – SEPTEMBER 12<sup>th</sup> (Tuesday)**

**Session 2: PCR products**

8:00 - 12:00	Overview of the laboratory exercise for Day 2: gel electrophoresis, PCR product clean up	CDC
	Practical 3: Preparation of agarose gels	Participants
	Coffee break	
	Practical 4: Analyze genotyping RT-PCR by agarose gel electrophoresis	Participants
12:00 - 13:00	Lunch	
13:00 - 17:00	Practical 5: Clean up PCR Products Exo-SAP-IT	Half participants
	Practical 6: Clean up PCR Products Invitrogen PureLink	Half participants
	Practical 7: Analyze cleaned up products by gel electrophoresis-Lonza	Participants

**DAY 3 – SEPTEMBER 13<sup>th</sup> (Wednesday)**

**Session 3: Sequencing reactions**

8:00 - 12:00	Overview of the laboratory exercises for Day 3: Sequencing reaction set-up and clean-up	CDC
	Practical 8: Big Dye sequencing reaction set up	Participants
	Coffee break	
	Lecture: MV and RV sequencing	CDC
12:00 – 13:00	Lunch	
13:00 – 17:00	Practical 9: Sequencing reaction clean up Agencourt	Half participants
	Practical 10: Sequencing reaction clean up CentriSep	Half participants
	Demo: Setup and run the Sequencer instrument	InDRE/CDC

### DAY 4 – SEPTEMBER 14<sup>th</sup> (Thursday)

#### Session 6: Analysis of sequences

8:00 - 12:00	Collect sequence raw data from the ABI	
	Demo of Recall	CDC
	Coffee break	
	Practical 11: chromatogram analysis using ReCall	Participants
12:00 - 13:00	Lunch	
13:00 - 17:00	Lecture: what to do if poor sequences	CDC
	Practical 12: chromatogram analysis using ReCall (“trouble” sequences brought by each lab)	Participants
	Lecture: MEGA, RubeNS, MeaNS, and Named Strains	CDC
	Demo of MEGA	CDC
	Practical 13: MEGA	Participants

### DAY 5 – SEPTEMBER 15<sup>th</sup> (Friday)

#### Session 7: Genotyping, MeaNS and RubeNS

8:00 - 11:00	Lecture: Exact Matches, Distinct Seq ID, and visualization tools	CDC
	Demo of MeaNS / RubeNS	CDC
	Practical 14: MeaNS / RubeNS submission	Participants
	Coffee break	
11:00 - 12:00	Discussion about real time PCR	ALL
12:00 - 13:00	Lunch	
13:00 - 16:30	Review of all test procedure, results, next steps	ALL
	Discussions	WHO
	Post-Assessment	Participants
	Participant Workshop Evaluation	Participants
	Final questions, comments, concerns by participants of the workshop	ALL
16:30	Wrap-up and adjourn	