ASM Webinar: Measles Update 2025: How to Avoid Rash Decisions

Introduction to measles virus and laboratory testing

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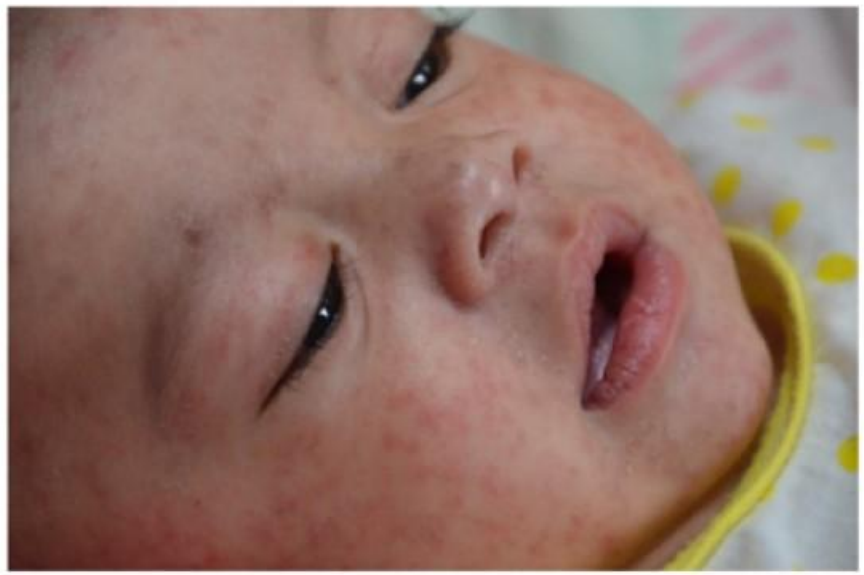
June 12, 2025

Introduction: Measles Virus

- Genus: Morbillivirus Family: Paramyxoviridae, Subfamily: Paramyxovirinae
- Genome is single stranded, negative sense RNA about 16Kb in length, helical nucleocapsid, 18 nm diameter
- Viral envelope contains surface glycoproteins for attachment and fusion
- Monotypic virus, but various genotypes have been described
- Infects only humans and non-human primates
- Live-attenuated vaccine has been in widespread use since the late 1960s, now part of MMR
- Severe outcomes of infection include inclusion body encephalitis (MIBE) and subacute sclerosing panencephalitis (SSPE)

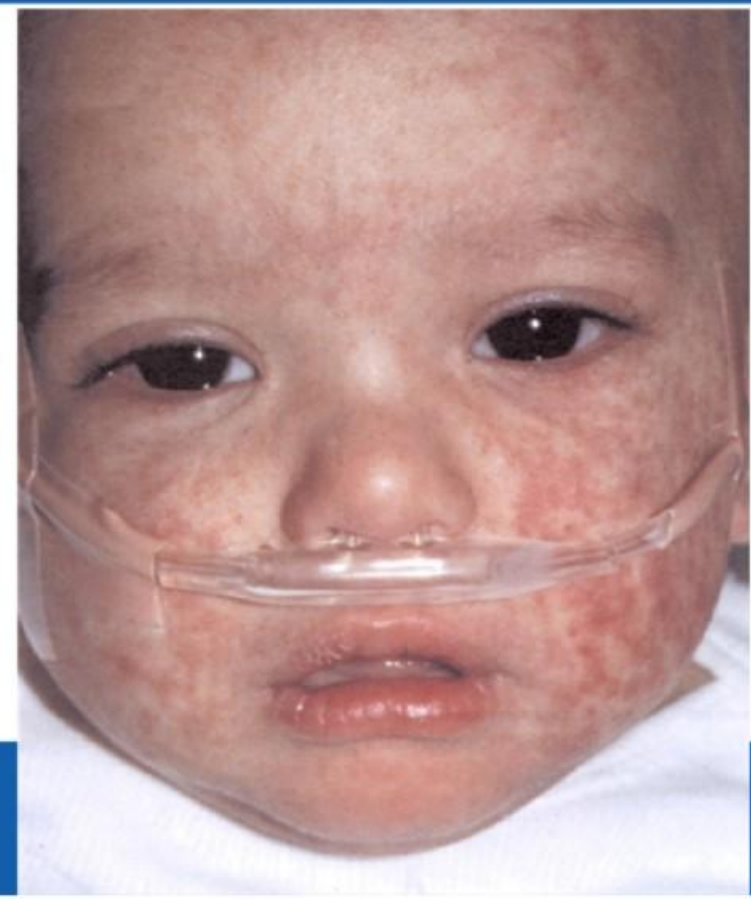
Measles

- **Acute, febrile rash viral illness**
  - High fever, rhinorrhea, conjunctivitis
- **Most contagious of the vaccine preventable diseases (R_0 = 12-16)**
- **Incubation period 10-14 days**
- **Infectious period 4 days prior through 4 days after rash onset**



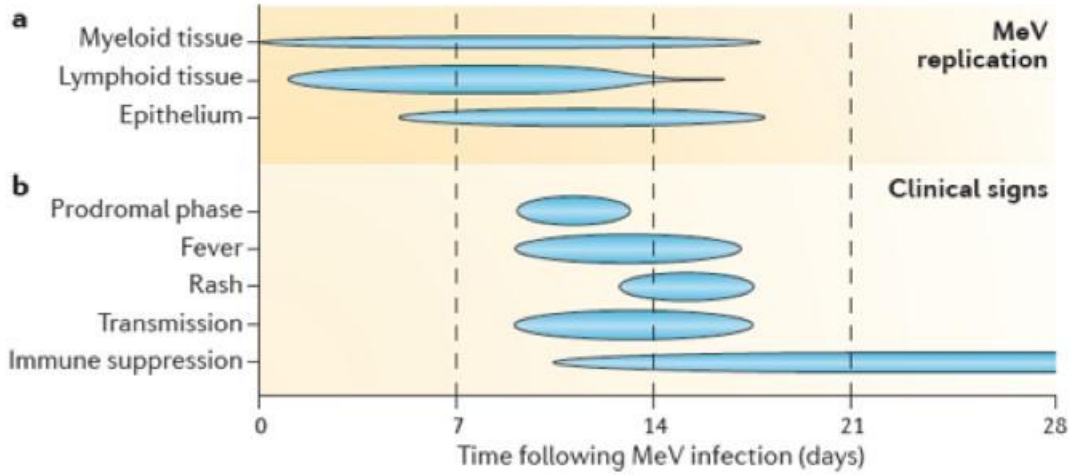
Measles complications

- Diarrhea (8%)
- Otitis media (7 – 9%)
- Pneumonia (1 – 6%)
- Hospitalized (10 – 25%)
- Encephalitis (1 per 1,000)
- Death (1 – 3 per 1,000)
- Subacute Sclerosing Panencephalitis (1 per 100,000)

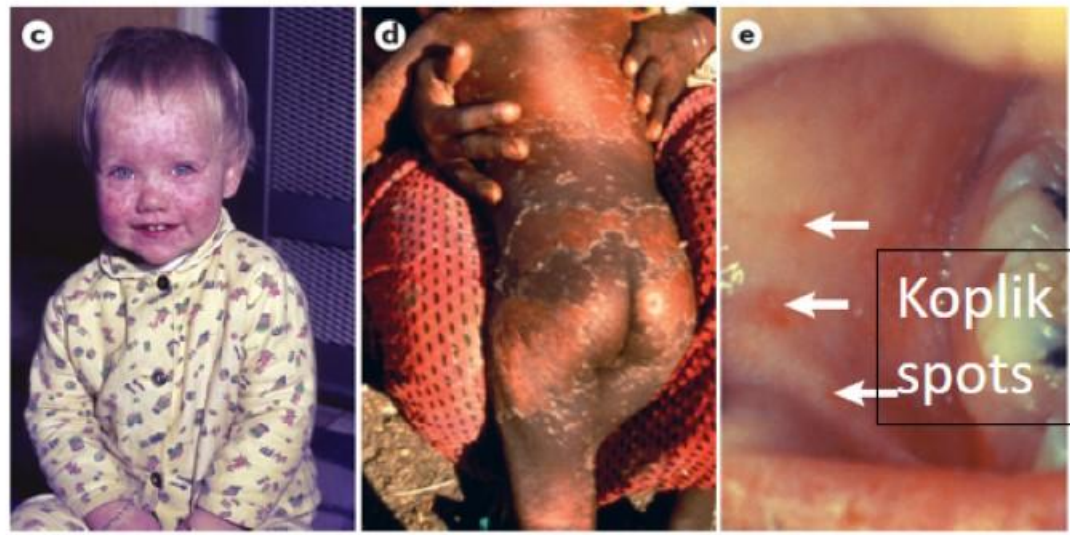


*SSPE: 1:5,000-10,000 cases

Pathogenesis of Measles



Immunosuppression causes high morbidity and mortality, secondary infections



Koplik spots

Rota, P. A. *et al.* (2016) Measles
Nat. Rev. Dis. Primers doi:10.1038/nrdp.2016.49

Measles in the US-2025 (data from https://cdc.gov/measles June 6, 2025)

U.S. Cases in 2025

Total cases

1168

Age
Under 5 years: **339 (29%)**
5-19 years: **439 (38%)**
20+ years: **381 (33%)**
Age unknown: **9 (1%)**

Vaccination Status
Unvaccinated or Unknown: **95%**
One MMR dose: **2%**
Two MMR doses: **3%**

U.S. Hospitalizations in 2025

12%

12% of cases hospitalized (137 of 1168).

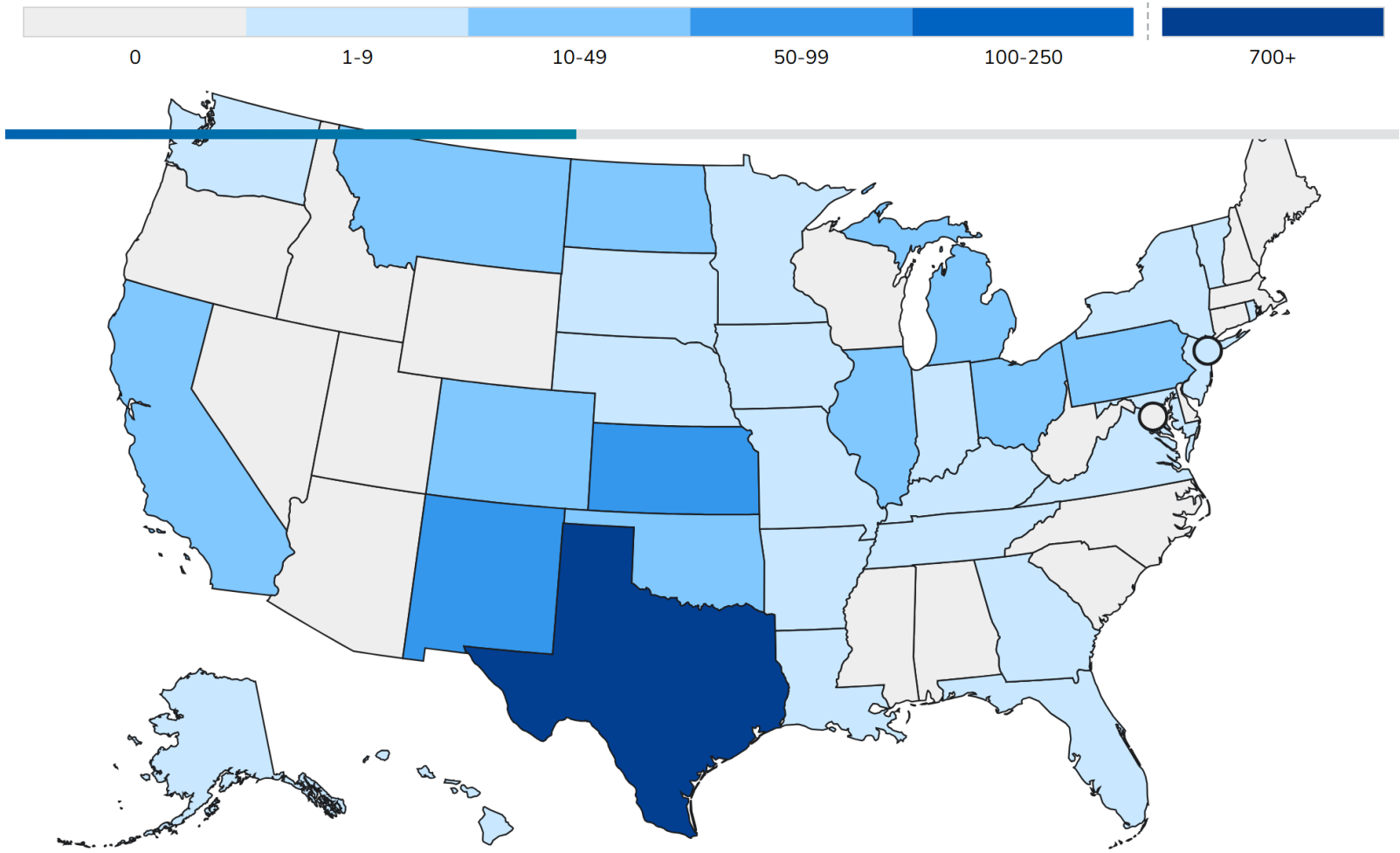
Percent of Age Group Hospitalized
Under 5 years: **21% (71 of 339)**
5-19 years: **8% (34 of 439)**
20+ years: **8% (31 of 381)**
Age unknown: **11% (1 of 9)**

U.S. Deaths in 2025

3

There have been 3 confirmed deaths from measles.

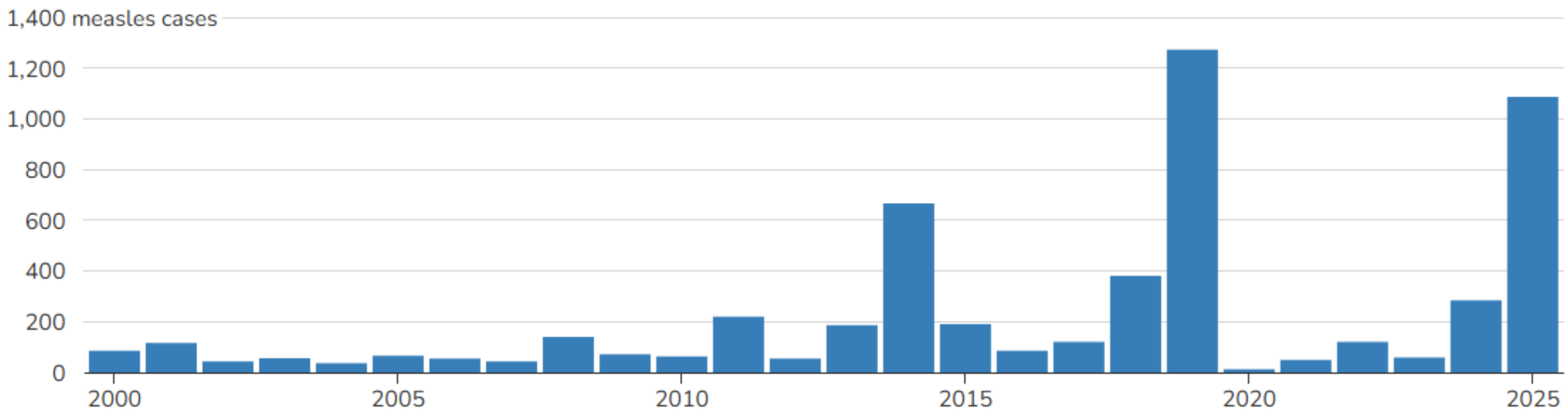
Map of measles cases in 2025



Yearly measles cases

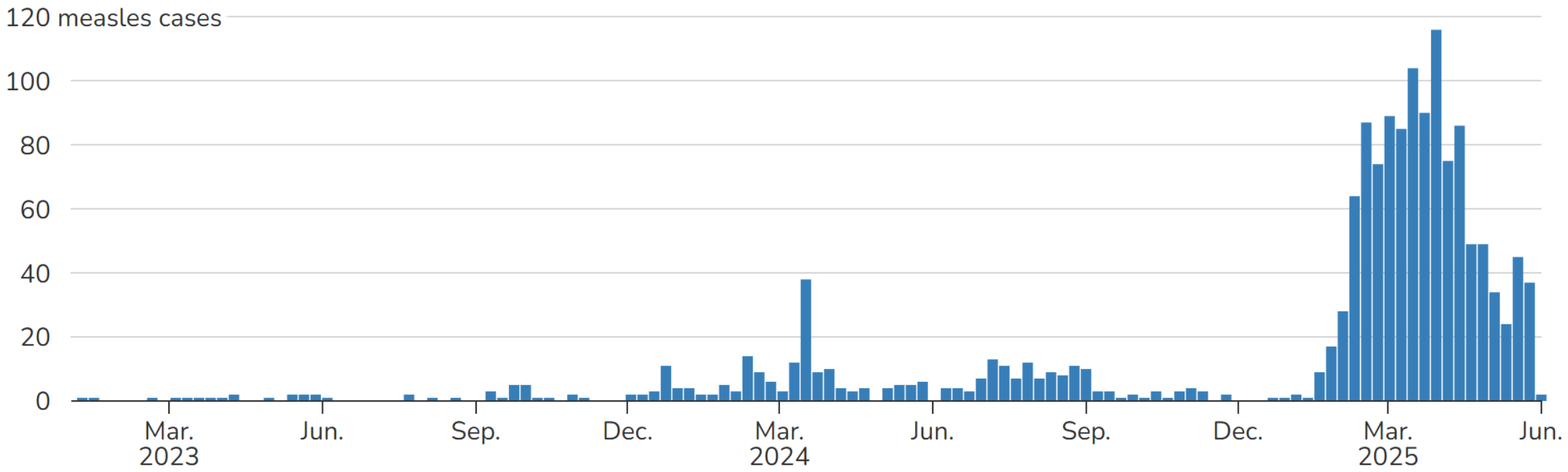
as of May 29, 2025

2000-Present* 1985-Present*



Weekly measles cases by rash onset date

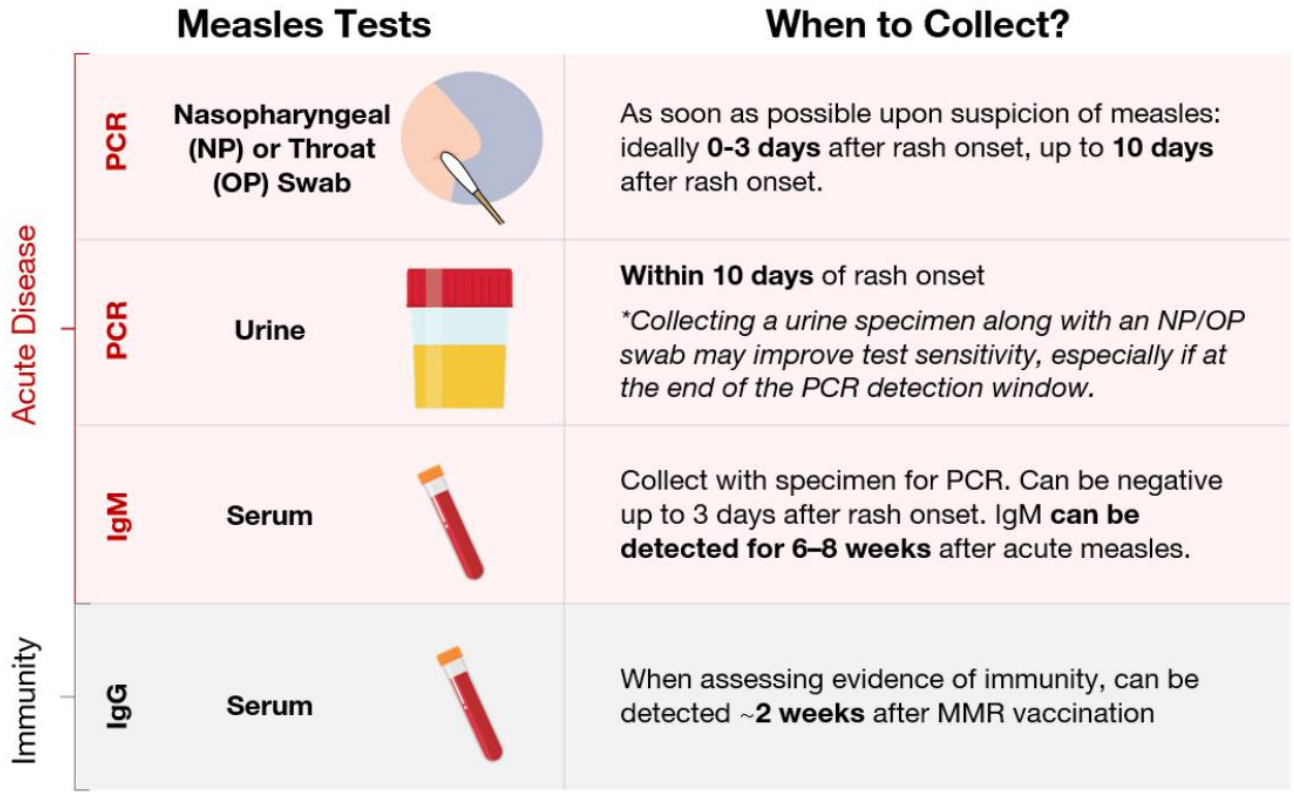
2023–2025* (as of June 5, 2025)



Laboratory testing for measles

Measles diagnosis and laboratory testing-1

- **Laboratory confirmation of measles infection is essential for all suspected measles cases whether sporadic or outbreak-associated.**
  - Real-time RT-PCR in respiratory (NP or throat swab) and urine specimens
  - Serologic testing to detect measles-specific IgM antibody in serum specimens



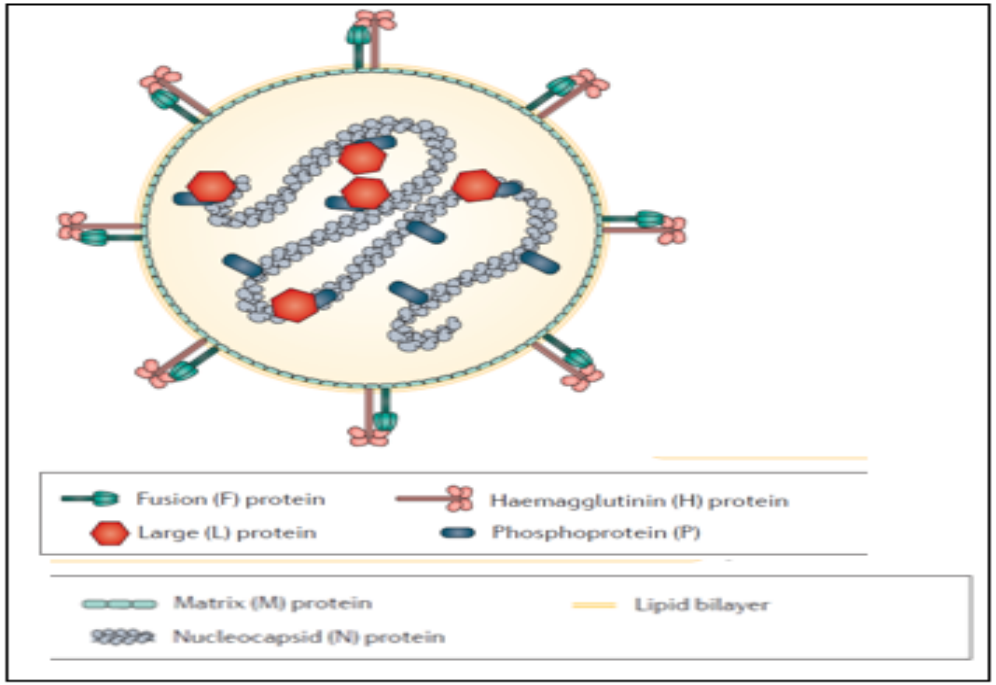
MMRV-Testing-for-Clinicians_Jan2025.pdf

Measles diagnosis and laboratory testing-2

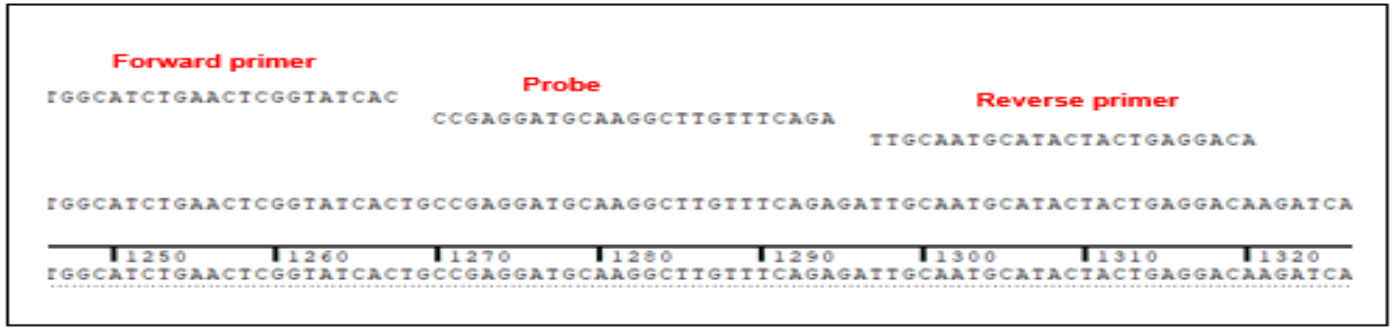
- **Rt-PCR is widely available.**
  - Need to collect specimen as close to rash onset as possible.
  - Specimens must be collected, stored and shipped properly to maintain specimen integrity
  - Detection of a cellular reference gene is ideal

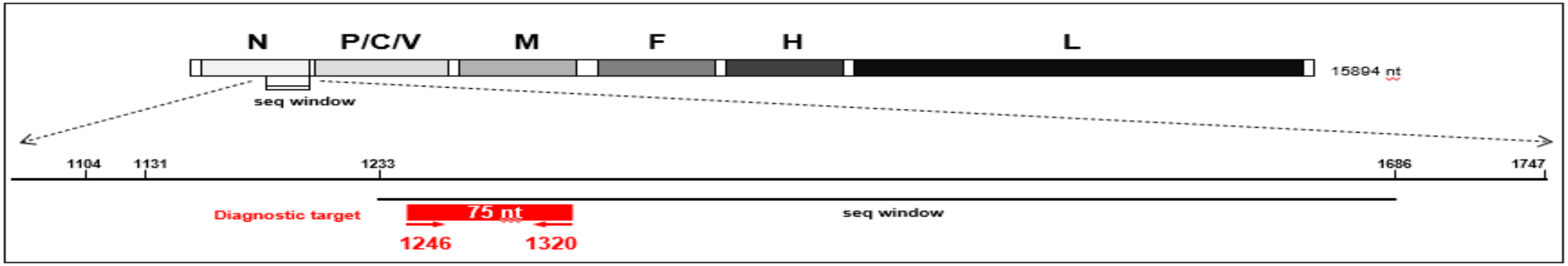
- **Serologic testing to detect measles-specific IgM antibody**
  - IgM test kits are not always available
  - Limited availability of testing, CDC and some state PHLs and commercial laboratories
  - Difficult to obtain specimens for validation
  - Can detect IgM up to one month after rash onset
  - False positive can occur with other infections which may complicate surveillance in low incidence settings

Overview: Measles Real Time RT-PCR Assay



CDC assay amplifies a 75 nucleotide (nt) region located close to the 3’ end of the N gene.





Measles Real Time RT-PCR Assays

- CDC test is a laboratory developed test (LDT)
  - Singleplex, Taqman assays:
    - Detects measles RNA and RNaseP RNA in separate reactions
    - Detects wild-type and vaccine strains of measles virus
    - ABI 7500 or ABI 7500 fast DX

- Many state and county laboratories perform measles RT-PCR
- Measles RT-PCR is offered through the APHL Vaccine Preventable Disease Reference Centers (VRCs)
  - https://www.aphl.org/programs/infectious_disease/Pages/VPD.aspx
- Testing is available from many commercial laboratories in various formats

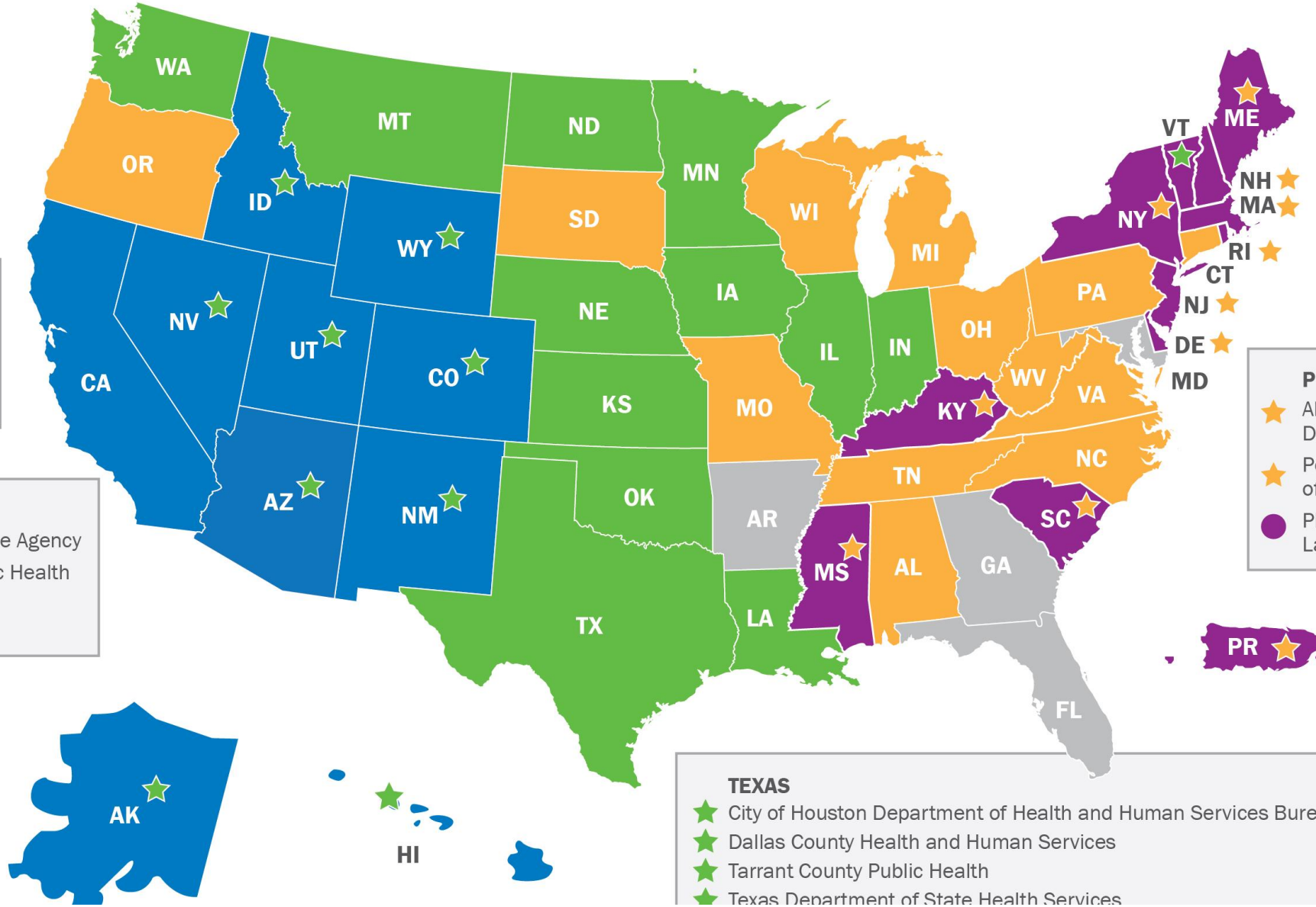
ENROLLED PHL VPD RC ASSIGNMENTS: VIRAL AND BACTERIAL

APHL™

OREGON
- Lane County Public Health Laboratory

NEVADA
- Nevada State Public Health Laboratory
- Southern Nevada Public Health Laboratory

CALIFORNIA
- Orange County Health Care Agency
- San Joaquin County Public Health Laboratory
- San Mateo County



PENNSYLVANIA
- Allegheny County Health Department
- Pennsylvania Department of Health Laboratories
- Philadelphia Public Health Laboratory

TEXAS
- City of Houston Department of Health and Human Services Bureau of Laboratories
- Dallas County Health and Human Services
- Tarrant County Public Health
- Texas Department of State Health Services

Identification of measles vaccine reactions: vaccine specific real-time RT-PCR assay (MeVa)

- Approximately 5% of individuals vaccinated with a measles-containing vaccine develop fever and rash that can be clinically indistinguishable from measles infection. Rapid differentiation of vaccine reactions from infections with wild-type virus is critical for guiding the public health response to outbreaks.

- Measles RT-PCR detects all genotypes of measles including vaccine strains.

- The Measles Vaccine Assay (MeVA) targets the N gene and distinguishes wild-type from vaccine strains. Must be performed conjunction with the standard measles RT-PCR.

- MeVA testing is available at CDC, the APHL-VPD Reference Centers, and ARUP

When should MeVA testing be considered?

Should be performed only on specimens (respiratory and urine) collected from patients with suspected vaccine reaction (febrile-rash illness within 21 days after vaccination) and potentially exposed to wild-type measles.

MeVA assay info:
https://www.aphl.org/Materials/VPD18_MeVA_Assay_CDC_Infosheet_032718.pdf

Onboarding measles molecular testing

- Measles and MeVA real-time RT-PCR assays are laboratory developed tests, and each laboratory has to perform their own validation prior to starting clinical testing.

- APHL Laboratory Test Verification and Validation Toolkit

- CDC measles laboratory and APHL-VPD Reference Centers available for technical consultations and/or questions labs may have while validating/onboarding measles testing.

- Limited availability of quantified positive control material through CDC's International Reagent Resource (IRR). Contact info to request: irr-mr@cdc.gov

- Measles viral isolates available through ATCC. CDC is planning on submitting additional strains in the near future including the strain with 3UC mutation. Can be used to create contrived specimens for validation studies.

Sending specimens to CDC and/or APHL-VPD RC

- Specimens that test positive for measles RNA should be submitted to CDC or an APHL Vaccine Preventable Disease Reference Center (VPDRC) for genotyping.

- Commercial laboratories are encouraged to submit positive specimens to CDC for genotyping
  - CSTOR: CDC Specimen Test Order and Reporting Web Portal
    - https://www.cdc.gov/infectious-diseases-labs/php/cstor-web-portal/index.html

- If requested by the submitter, specimens should also be sent for Measles Vaccine Assay (MeVA) testing.

- For sending specimens for genotyping and/or MeVA testing:
  - Contact the state or local health department to determine where to submit specimens and how to ship them

  - If sending to CDC, refer to the Infectious Diseases Laboratories Test Directory for each test
    - Measles detection instructions (Test CDC-10543)
    - Measles vaccine virus detection instructions (Test CDC-10528)
    - Measles Genotyping instructions (Test CDC-10240)

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Article

The Global Measles and Rubella Laboratory Network Supports High-Quality Surveillance

Paul A. Rota [1], Roger Evans [2], Myriam Corinne Ben Mamou [3], Gloria Rey-Benito [4], Lucky Sangal [5], Annick Dosseh [6], Amany Ghoniem [7], Charles R. Byabamazima [6], Maurice Demanou [6], Raydel Anderson [1], Gimin Kim [1], Bettina Bankamp [1], R. Suzanne Beard [1], Stephen N. Crooke [1], Sumathi Ramachandran [1], Ana Penedos [8], Vicki Stambos [9], Suellen Nicholson [9], David Featherstone [10] and Mick N. Mulders [11,*]

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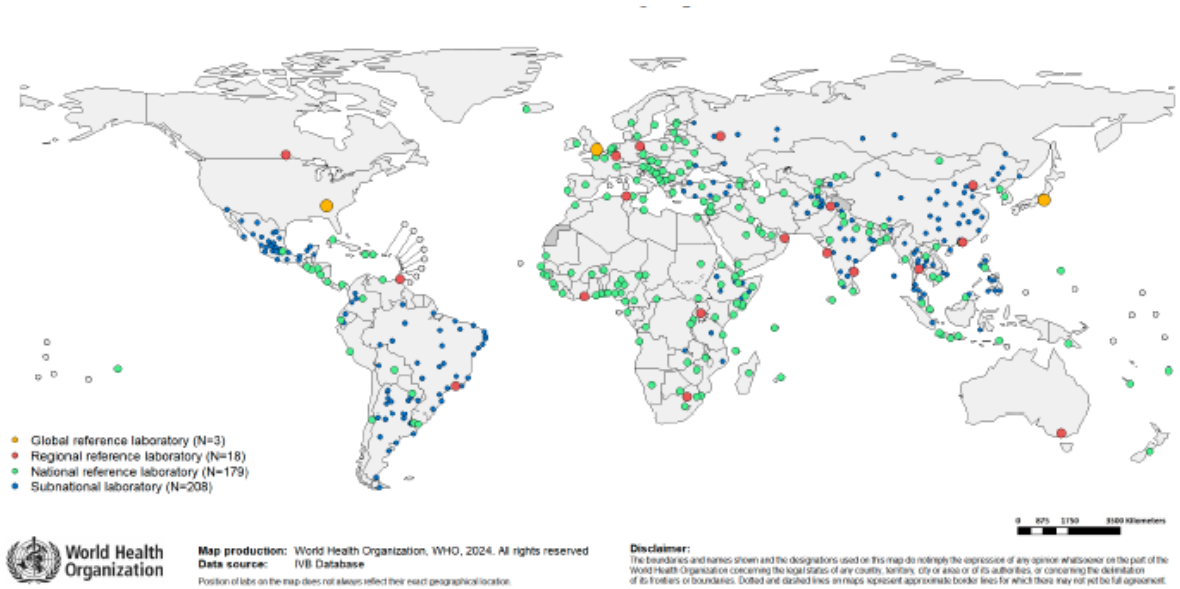

Figure 2. Map of GMRLN. Yellow circles indicate the locations of GSLs. Red circles indicate RRLs. Light blue circles indicate NLs. Blue circles indicate SNLs.

Genetic characterization of measles viruses

- Measles virus genotyping and sequence analysis can play an important role in tracking transmission pathways during outbreak investigations and is used for global surveillance of wild-type measles strains. Should be used in conjunction with epidemiological information.
- Measles positive upper respiratory/urine specimens should be sent for genotyping. In large outbreaks, prioritization of certain specimens for genotyping may be considered.
- **Genotyping by sequencing N450 region (Standard genotyping procedure)**
  - Available at CDC and the APHL-VPD Reference Centers
  - Distinct Sequence IDs (DSIds) assigned based on N450 sequence
  - MeaNS global database for measles sequences (majority N450)
- **Whole genome sequencing (WGS):** Analysis of WGS adds additional sequence information but may not be able to resolve all transmission pathways. Multiple importations of viruses with nearly the same WGS are possible given the lack of sequence diversity and frequent transmission from large outbreaks.

vaccines

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Review

Global Update on Measles Molecular Epidemiology

Bettina Bankamp [1,*,†], Gimin Kim [1,†], Derek Hart [2,†], Andrew Beck [1,†], Myriam Ben Mamou [3], Ana Penedos [4], Yan Zhang [5], Roger Evans [6] and Paul A. Rota [1,*,†]

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* Correspondence: bfb9@cdc.gov (B.B.); par1@cdc.gov (P.A.R.)
† CDC disclaimer: The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the Centers for Disease Control and Prevention.

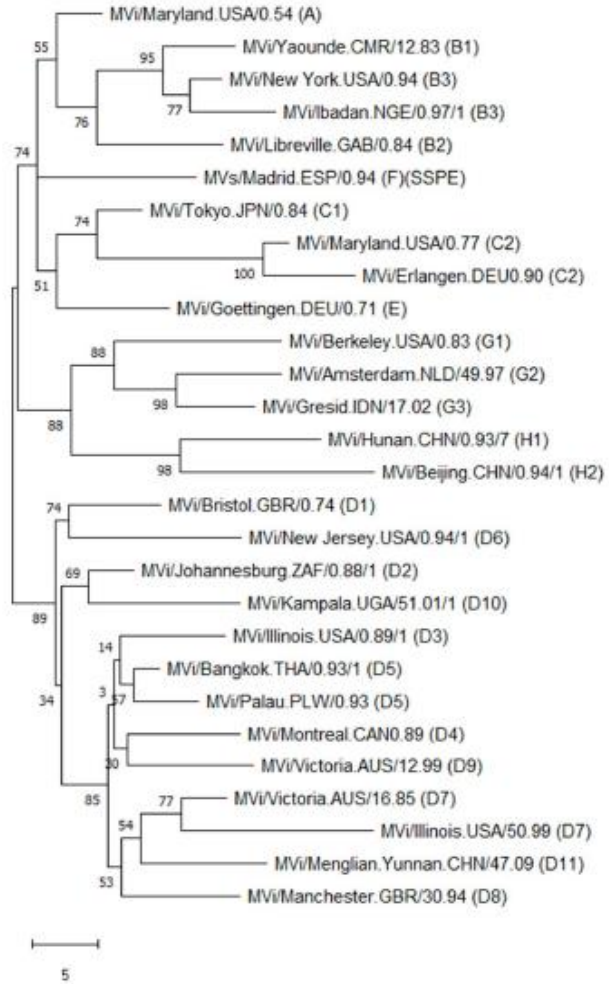

Figure 1. Phylogenetic tree with 28 measles N450 reference sequences. Data from MeaNS [8]. The evolutionary history was inferred using the maximum parsimony method. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (500 replicates) are shown next to the branches. The tree is drawn to scale, with branch lengths calculated using the average pathway method and are in the units of the number of changes over the whole sequence. Scale bar indicates number of nucleotides.

Detection of measles genotypes over time

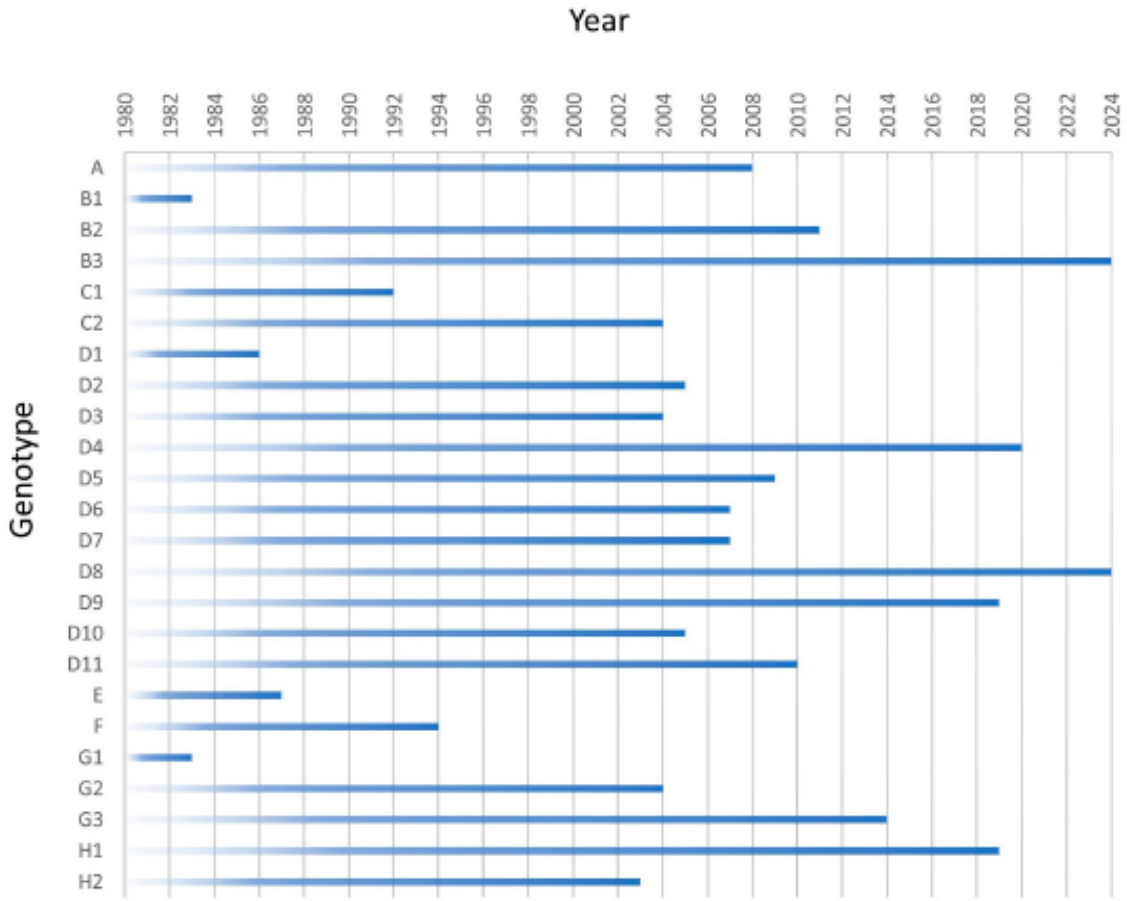

Figure 2. Year of last detection of measles genotypes. The last year of documented circulation reporte to MeaNS [8] for all 24 genotypes is shown. Viruses with a date of 2024 are currently circulating. A other viruses have had transmission interrupted in the year depicted. Note that the chart does no specify the first year of detection of any genotype.

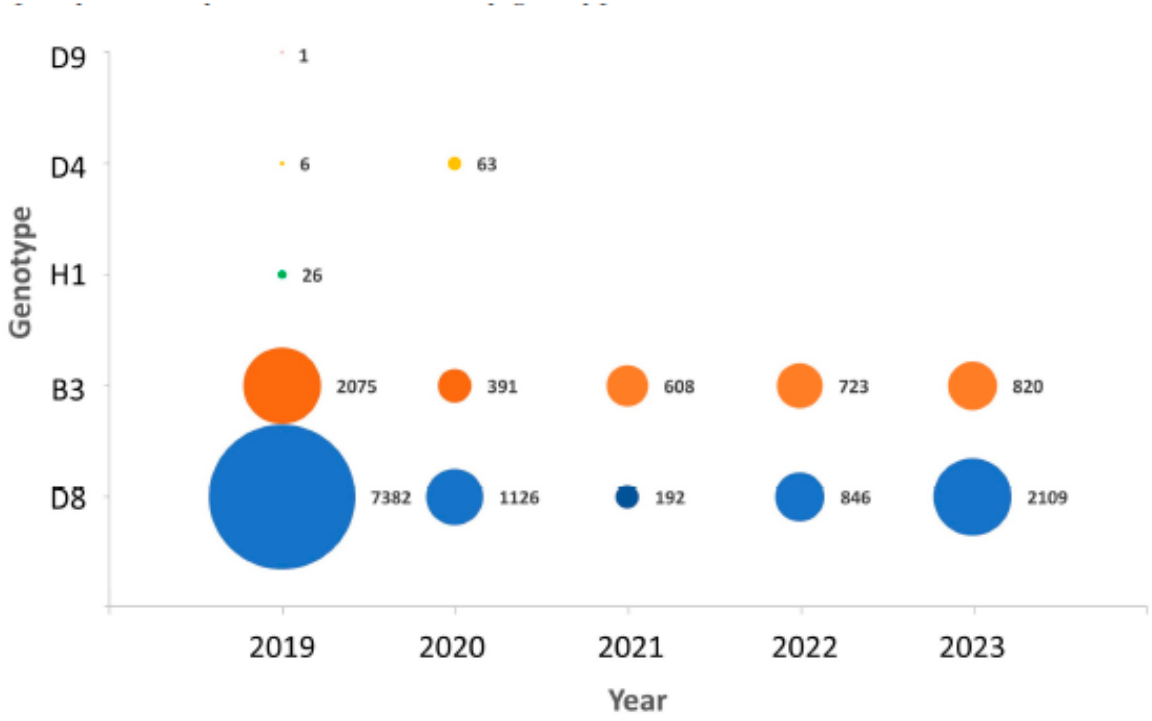

Figure 3. Number of submissions per genotype reported to MeaNS [8] 2019–2023.

Distinct Sequence Identifier (DSId): Each unique N450 sequence is assigned a DSId. Viruses assigned the same DSId have identical N450 sequences.

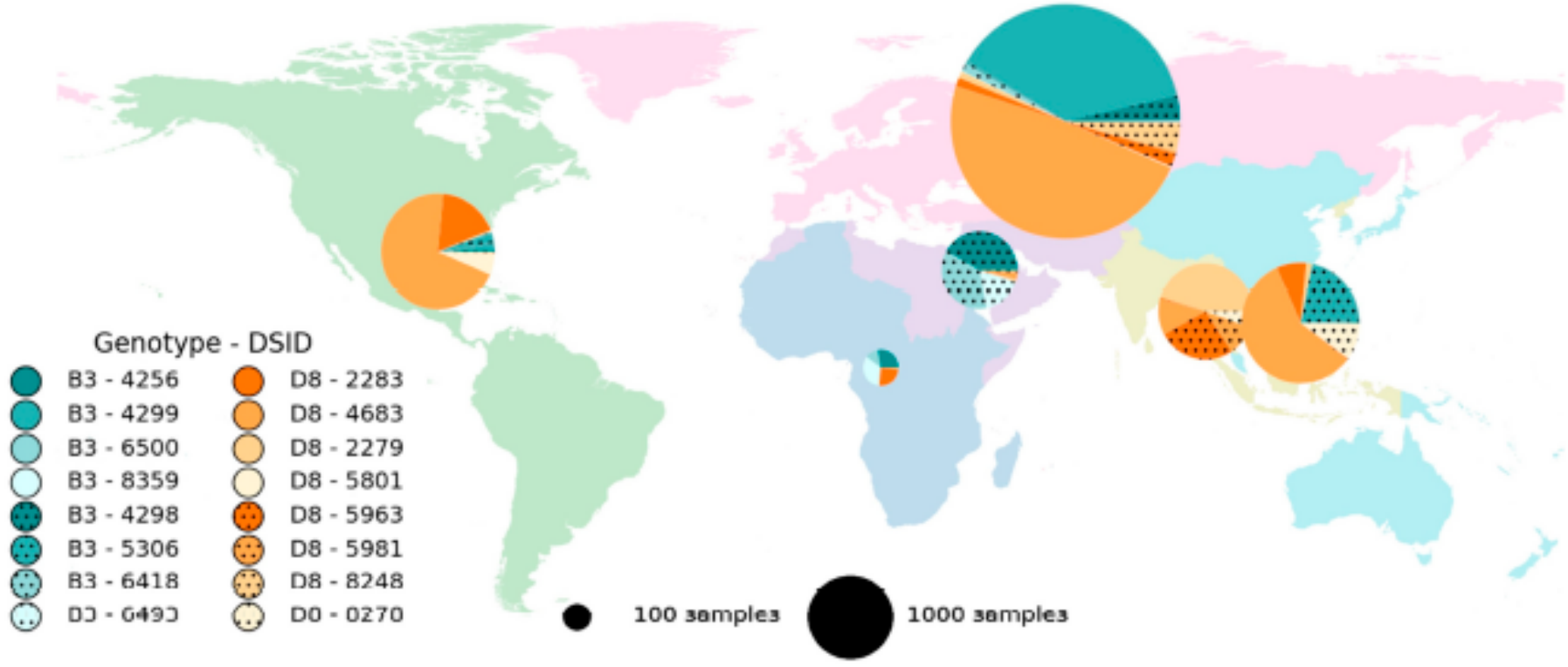

Figure 4. Global distribution of major DSIds according to the WHO region, 2019–2023. The genotype is indicated by the color and pattern in each pie chart. The size of the pie chart indicates the number

Thank you