Laboratory Outreach Communication System (LOCS) Call

Monday, September 19, 2022, at 3:00PM EDT

- **Welcome**
  - Sean Courtney, Division of Laboratory Systems, CDC
- **Monkeypox Update**
  - Serena Carroll, Monkeypox Response, CDC
- **Monkeypox Virus Sequencing in the 2022 Outbreak Reveals Multiple Independent Lineages and Unique Mutational Signatures**
  - Crystal Gigante, Division of High-Consequence Pathogens and Pathology, CDC
- **National Wastewater Surveillance System (NWSS): Opportunities and Challenges for Wastewater Surveillance for Monkeypox**
  - Carly Adams, Division of Foodborne, Waterborne, and Environmental Diseases, CDC
- **FDA Update**
  - Tim Stenzel, US Food and Drug Administration (FDA)

About DLS

Vision

Exemplary laboratory science and practice advance clinical care, public health, and health equity.

Mission

Improve public health, patient outcomes, and health equity by advancing clinical and public health laboratory quality and safety, data and biorepository science, and workforce competency.

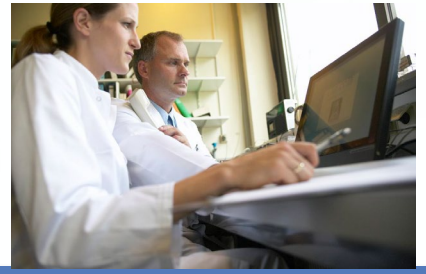
Four Goal Areas

Quality Laboratory Science

- Improve the quality and value of laboratory medicine and biorepository science for better health outcomes and public health surveillance

Highly Competent Laboratory Workforce

- Strengthen the laboratory workforce to support clinical and public health laboratory practice



Safe and Prepared Laboratories

- Enhance the safety and response capabilities of clinical and public health laboratories

Accessible and Usable Laboratory Data

- Increase access and use of laboratory data to support response, surveillance, and patient care

LOCS Calls

https://www.cdc.gov/locs/calls

Find LOCS Call information, transcripts, and audio recordings on this page

Next Scheduled Call

Monday, October 17

@ 3 PM to 4 PM ET

We Want to Hear From You!

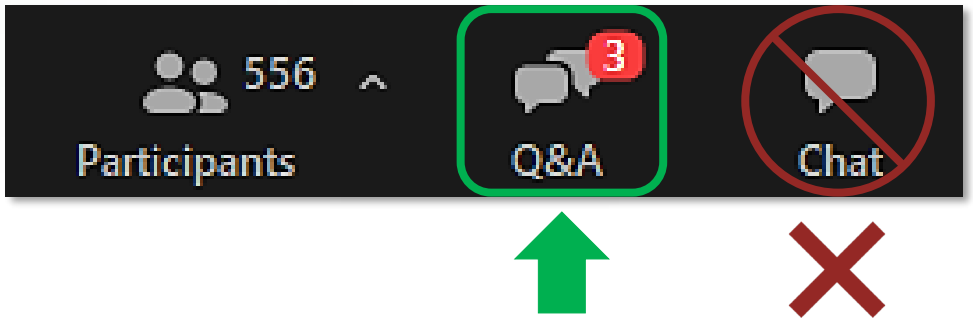
Training and Workforce Development

Questions about education and training?

Contact LabTrainingNeeds@cdc.gov

How to Ask a Question

- **Using the Zoom Webinar System**
  - Click the **Q&A button** in the Zoom webinar system
  - Type your question in the **Q&A box** and submit it
  - Please do not submit a question using the chat button



- For media questions, please contact CDC Media Relations at media@cdc.gov
- If you are a patient, please direct any questions to your healthcare provider

Slide decks may contain presentation material from panelists who are not affiliated with CDC. Presentation content from external panelists may not necessarily reflect CDC's official position on the topic(s) covered.

Monkeypox Update

Serena Carroll
Monkeypox Response, CDC

Monkeypox virus sequencing in the 2022 outbreak reveals multiple independent lineages and unique mutational signatures

Crystal M. Gigante

Centers for Disease Control and Prevention

Laboratory Outreach and Communication System (LOCS) Call
Monday, 19 September 2022

The findings and conclusions in this report are those of the presenter and do not necessarily represent the views of the Centers for Disease Control and Prevention

Monkeypox

- In 2017, the largest monkeypox outbreak in western Africa occurred in Nigeria after decades of no identified cases in the country.
- During 2018 to 2021, eight cases were imported from Nigeria to non-endemic countries.
- Since the re-emergence of MPXV in Nigeria in 2017, there had only been two reported events of person-to-person transmission of MPXV outside of Africa.
- In May of 2022, this pattern of monkeypox cases in travelers from Nigeria shifted, and multiple countries reported monkeypox among persons who had not travelled to monkeypox-endemic countries.

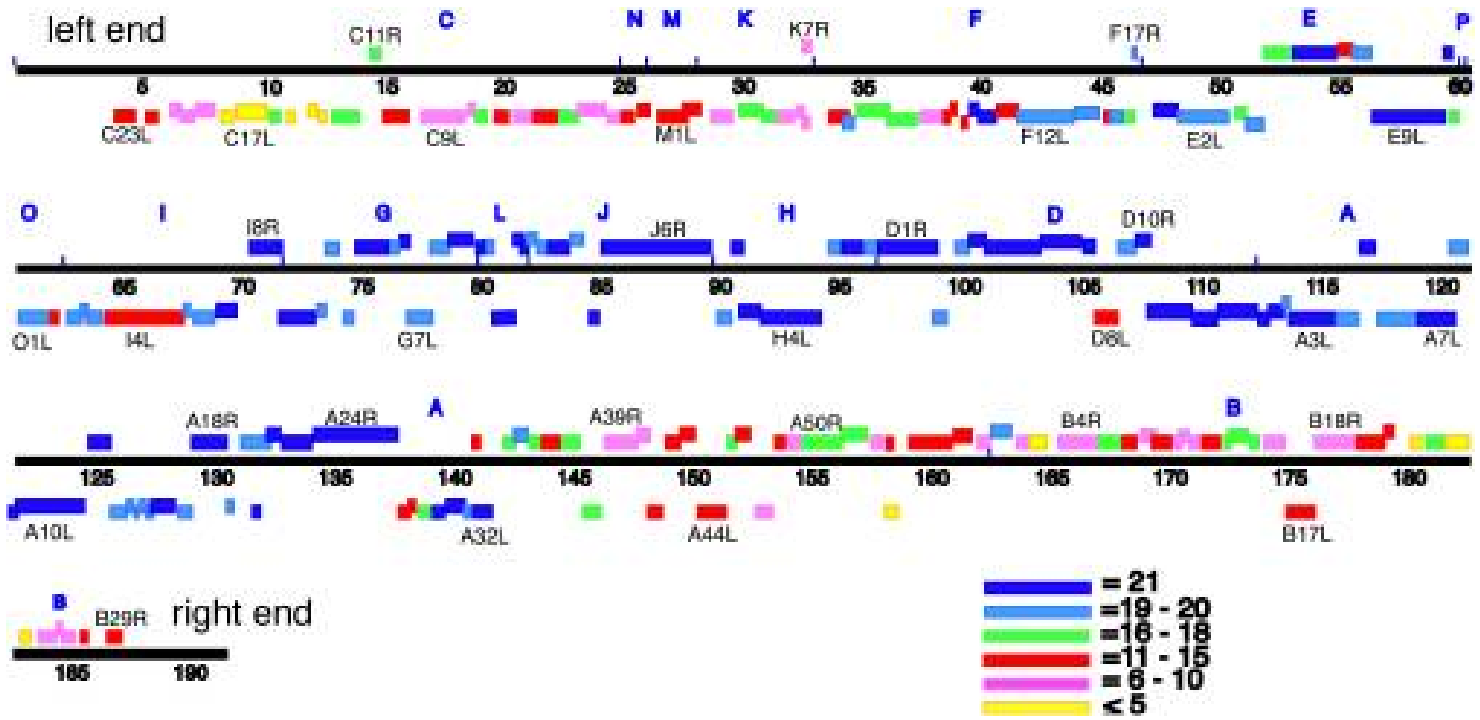
2022 Monkeypox virus sequencing update

- All publicly available MPXV genomes from the 2022 Monkeypox outbreak to date belong to Clade IIb (formerly West African) MPXV

- Genomes published during the 2022 monkeypox outbreak share a common ancestor with MPXV from Nigeria; however, sequences from surrounding countries are limited and most of our understanding of these relationships comes from viruses linked to or identified in Nigeria

Monkeypox virus

- 200 kb dsDNA genome
- Several repeat regions
- Inverted terminal repeats
- ~200 predicted coding sequences (CDS)
  - Colored boxes indicate conservation of CDS across orthopoxviruses
  - Blue: highly conserved
  - Yellow: less conserved



Upton *et al*. Poxvirus orthologous clusters: toward defining the minimum essential poxvirus genome. *J Virol*. 2003

Monkeypox virus sequencing: Clinical samples

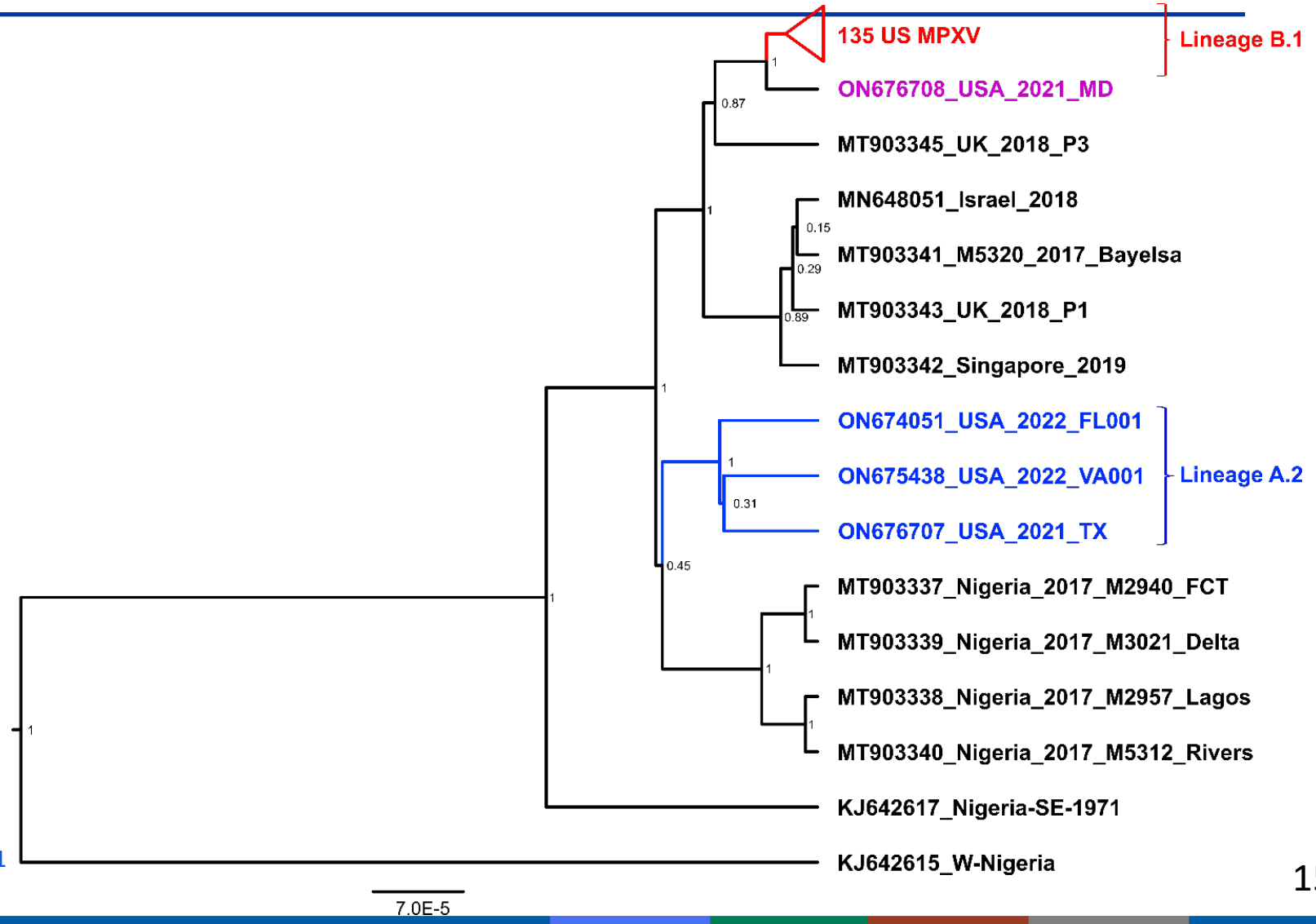
- Sample type: lesion swab
- Total DNA very low
- Monkeypox virus DNA 0.1% - 5% of total DNA

Approach:

Screening: Clade II MPXV PCR Ct < 30, Average coverage > 35
Sequencing: Metagenomics Sequencing, Illumina DNA prep, NovaSeq
Assembly: de novo assembly (SPAdes), polishing ivar, minimum coverage 20, -q 20, 2/3 majority
Sequencing 2: Direct sequencing Ligation kit LSK109 on ONT MinION or GridION
Assembly 2: hybrid assembly (Unicycler), polishing ivar, minimum coverage 20, -q 20, 2/3 majority

U.S. Monkeypox virus sequencing update

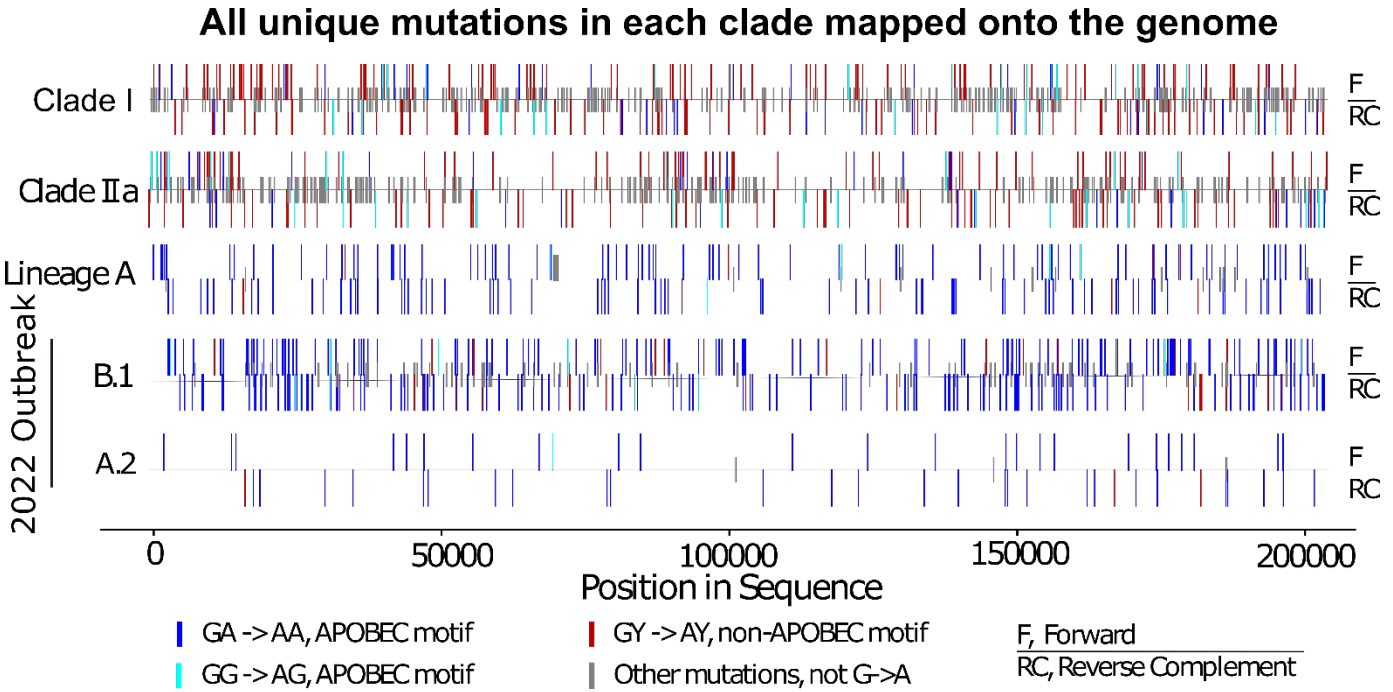
- 2022 U.S. monkeypox virus sequences fall into two lineages (red and blue)
- Lineage B.1 (red)
  - Most U.S. sequences
  - Most global sequences
- Lineage A.2
  - Three 2022 U.S. cases (VA, FL, PA)
  - One 2021 case (TX)
  - Also reported in India and Thailand
  - Most had travel to Western Africa or Middle East



https://www.biorxiv.org/content/10.1101/2022.06.10.495526v1

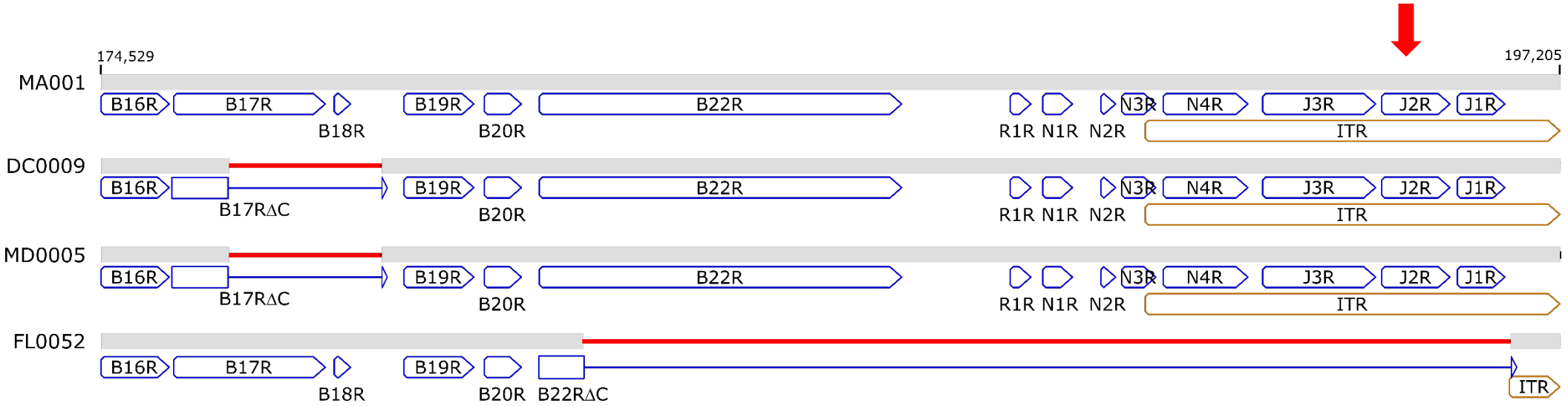
Increased mutations observed in 2022 MPXV, evidence of host protein ABOBEC3 activity

- Many of the observed mutations in the 2022 outbreak MPXV sequences are GA-to-AA
- This is indicative of Apolipoprotein B mRNA Editing Catalytic Polypeptide-like3 (APOBEC3) deaminase activity
- APOBEC3 proteins are part of vertebrate innate immune system that restrict the replication of viruses through cytosine-to-uracil deaminase activity



Preliminary finding: evidence of large deletions in 2022 MPXV

- ~3% of MPXV genomes contain evidence of deletions
- Deletions have potential to impact diagnosis and efficacy of therapeutics
- Deletions are more common in the terminal regions of the genome, where non-essential genes are located
- MPXV Clade II (West African)-specific, MPXV generic assays target a terminal gene: soluble TNF receptor CrmB/ J2R



Summary

- Genomic surveillance revealed multiple lineages of MPXV in the US in 2021 and 2022
- MPXV sequences since 2017 exhibit evidence of host antiviral editing
- ~3% of MPXV genomes contain large deletions or rearrangements
- Analyses are expected to continue as more sequences are reported

Acknowledgements

- National Center for Emerging and Zoonotic Infectious Diseases, Centers for Disease Control and Prevention, Atlanta, Georgia, USA
- Leidos Inc., Reston, VA 20190, USA
- Los Alamos National Laboratory
- Massachusetts Department of Public Health
- Maryland Department of Health
- Minnesota Department of Health
- Florida Department of Health
- Washington DC department of Health
- Maryland department of Health
- Rhode Island department of Health
- Dallas County Health and Human Services Public Health Laboratory
- Florida Department of Health Bureau of Public Health Laboratories-Jacksonville
- Florida Department of Health in Broward County
- Virginia Department of General Services
- Virginia Department of Health
- Utah Department of Health and Human Services
- Sacramento County Public Health
- Leidos Inc., Reston, VA 20190, USA

References

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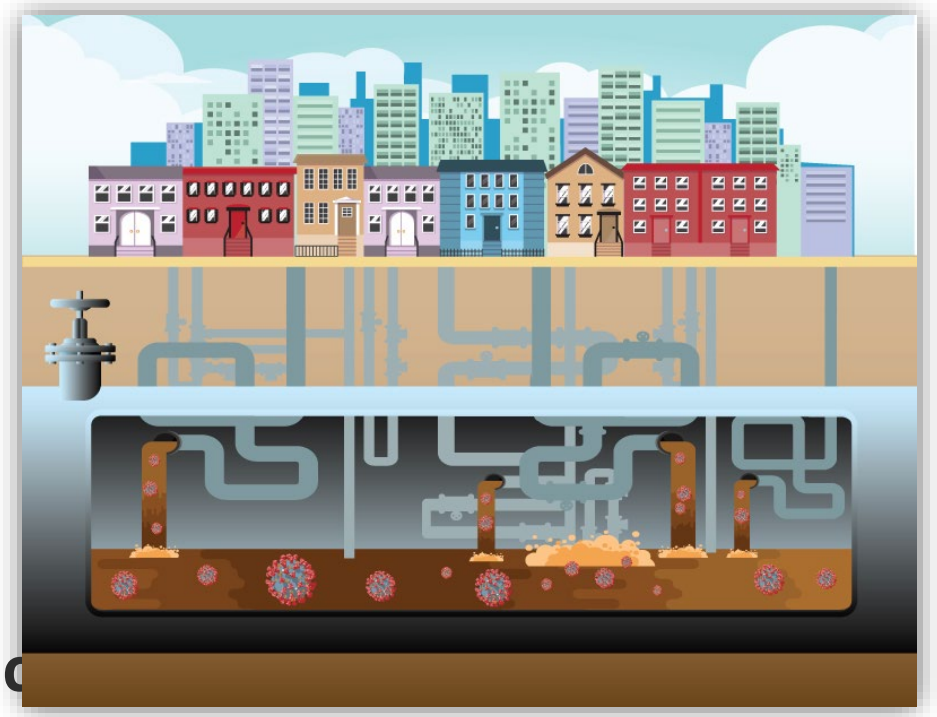
20. https://www.biorxiv.org/content/biorxiv/early/2022/08/01/2022.07.30.502168.full.pdf

National Wastewater Surveillance System (NWSS): Opportunities and Challenges for Wastewater Surveillance for Monkeypox

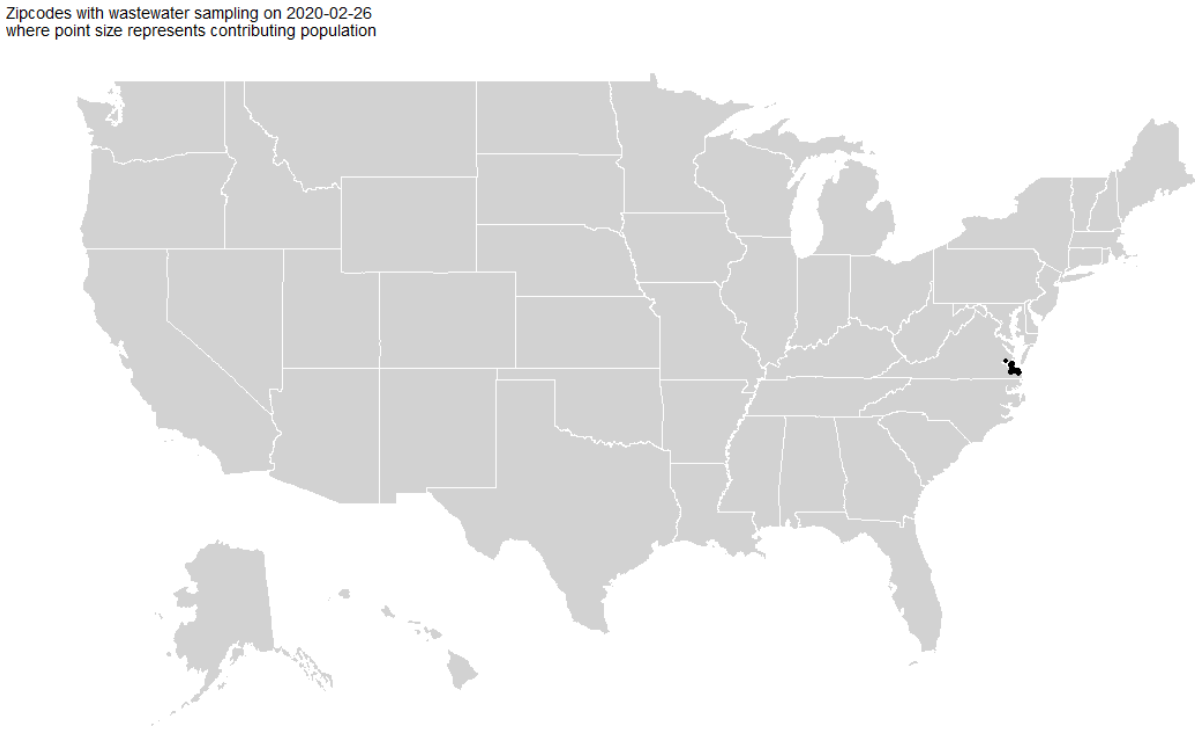
Carly Adams, PhD
Epidemic Intelligence Service (EIS) Officer
Waterborne Disease Prevention Branch
Division of Foodborne, Waterborne and Environmental Diseases

LOCS Call
September 19, 2022



NATIONAL ™ WASTEWATER SURVEILLANCE SYSTEM

NWSS implementation has grown rapidly

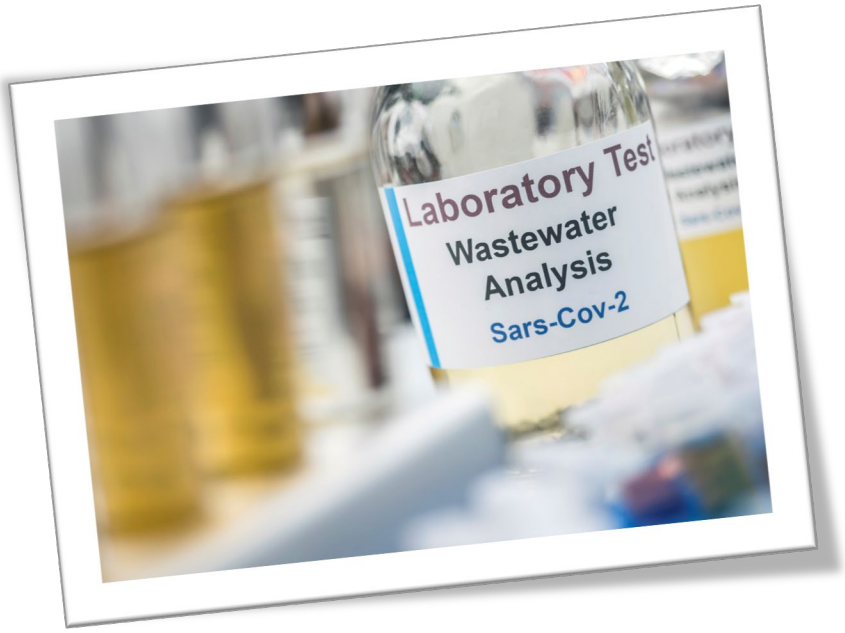


- 46 states, 5 major cities and 2 territories using CDC funds for wastewater surveillance
- Commercial testing contract services 367 sites nationwide, prioritizing vulnerable communities
- Over 83,000 unique wastewater samples from represent over 130M people, which is about 50% of Americans on a sewer system.

Wastewater Surveillance | Public Health Toolbox

- Can capture asymptomatic infections
- Independent of healthcare-seeking behavior and testing access
- Wastewater serves as an efficient pooled sample of community (or sub-community) infection levels
- Data available quickly
- Flexible platform that can be leveraged for multiple pathogen targets

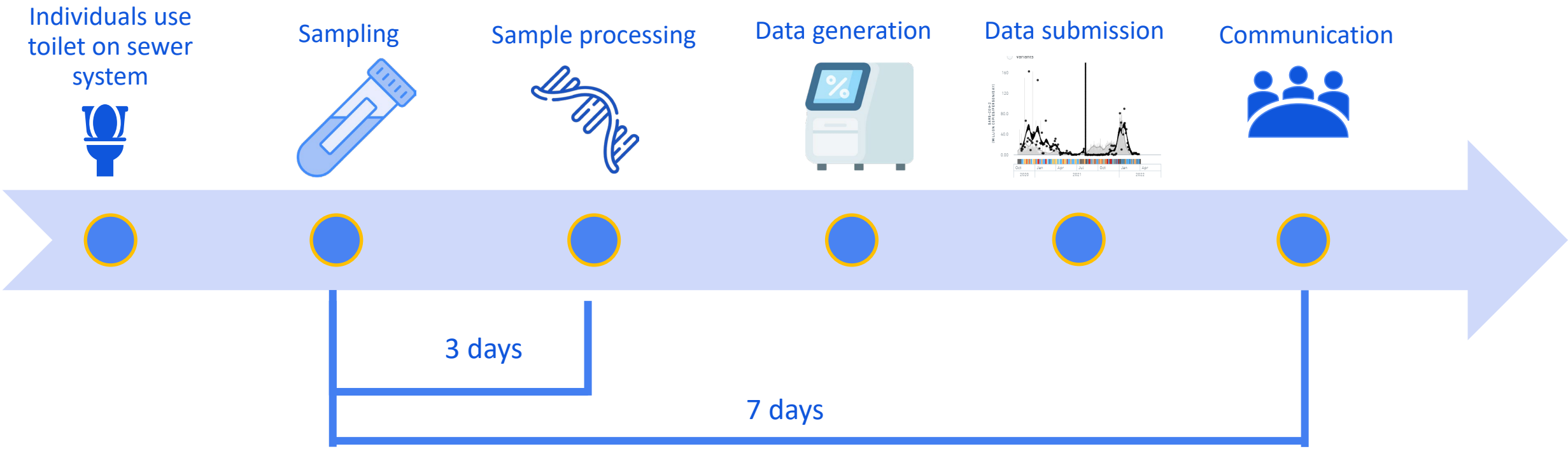
Successful Use of Wastewater Data for Response



State and local jurisdictions have used COVID-19 wastewater data to inform response decisions:

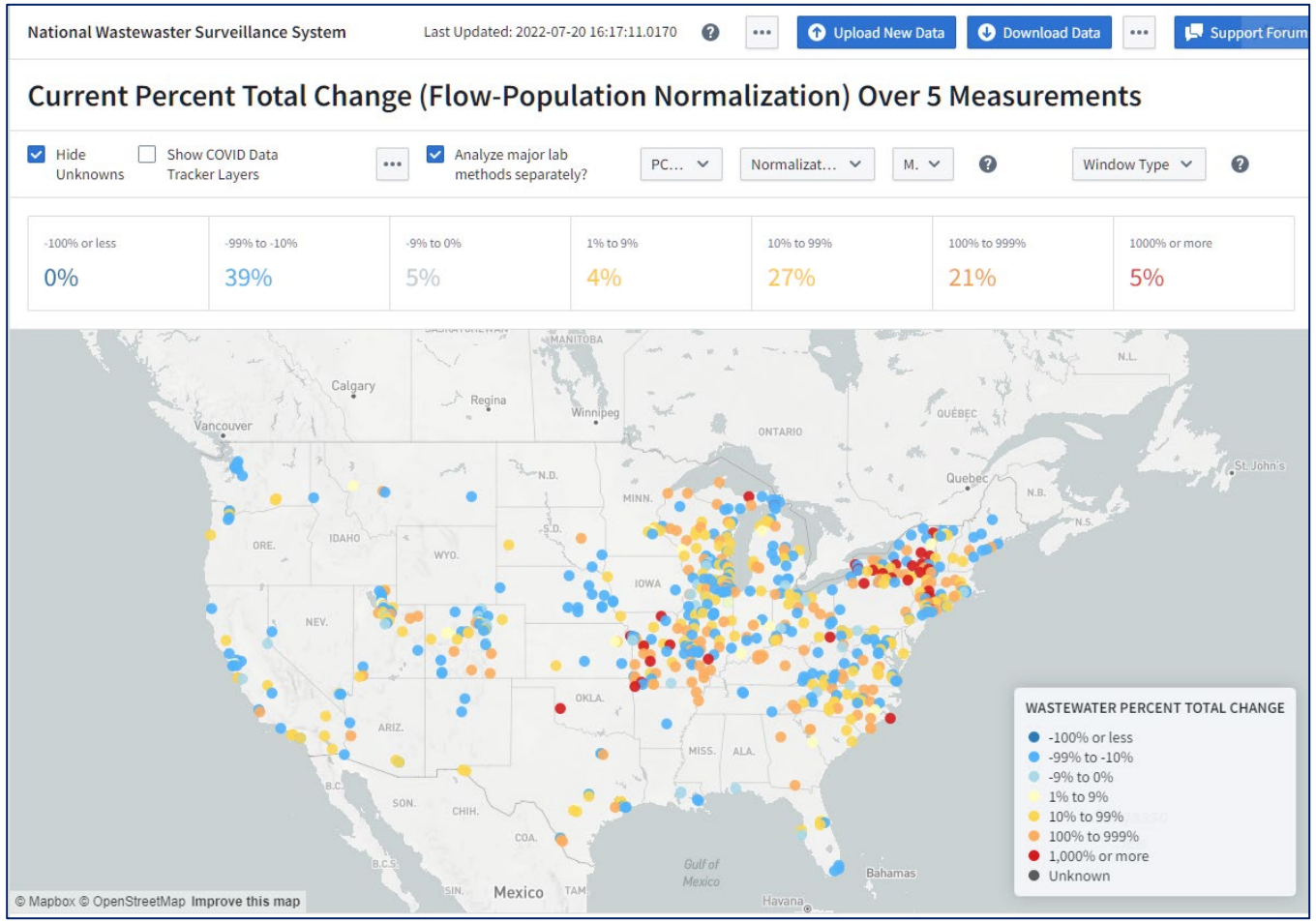
- ✓ Independent confirmation of true increases or decreases in cases
- ✓ Public health messaging
- ✓ Distribution, siting of test capacity
- ✓ Surveillance data in communities where clinical testing is limited or not available
- ✓ Near-term forecasting of cases or hospital utilization
- ✓ Monitoring the impact of home testing
- ✓ Detecting the emergence of Variants of Concern

Timeliness and quality of wastewater data



- Rapid turnaround time is key for public health action
- DCIPHER provides automated reports to ensure that data are high quality
- Data submission options include APIs for ease of use
- Fast, quality data can give lead times of up to 4-6 days on clinical cases

Wastewater surveillance data: the NWSS DCIPHER Data Dashboard

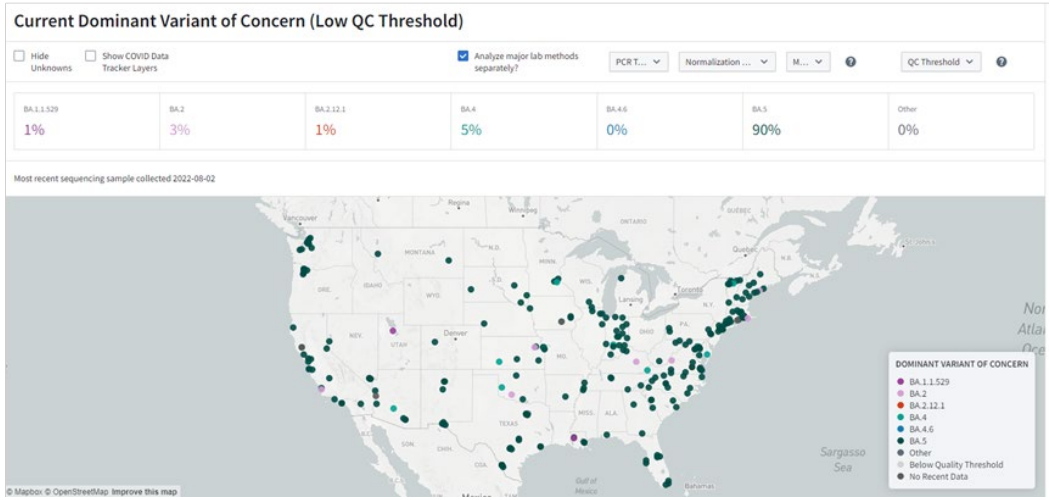


Metric	What does this show us?
Percentiles	Relative levels of virus present in a community over time
Percent Change	Magnitude and direction of virus levels in a community
Detection Proportion	How frequently is the virus detected in a community
Variant Specific Metrics	If a known variant is present, and at what proportion

NWSS Sequence Data Visualization Dashboard

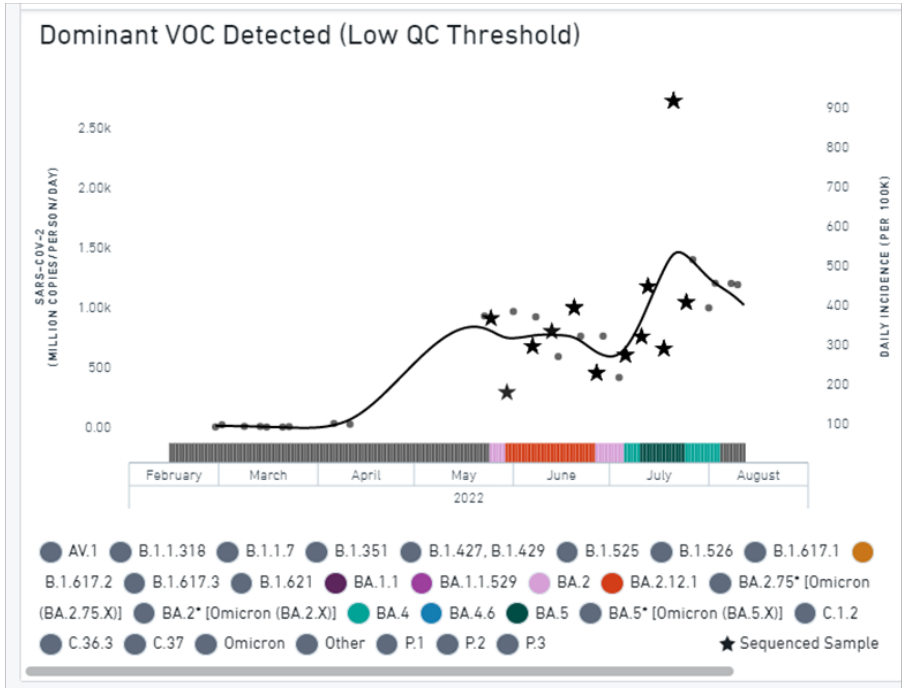
1

DCIPHER Dashboard Dominant Variant of Concern Map of US



2

Wastewater Dominant Variant per Sewershed for a wastewater sampling location



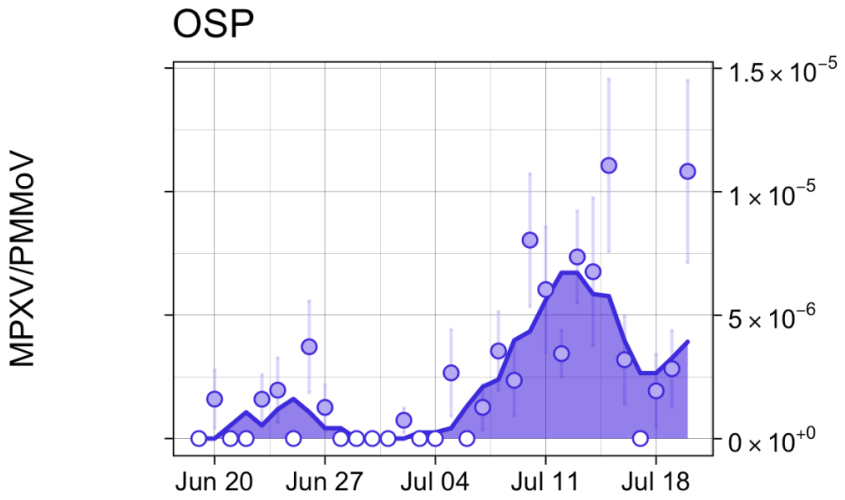
NWSS and Monkeypox

- Monkeypox virus DNA may be shed in stool and urine, as well as other fluids, that could be captured in untreated wastewater
- The testing platform is flexible: the NWSS team is now working to validate a protocol for testing of influent wastewater for monkeypox virus DNA
  - This would give us quantitative levels of DNA over time to monitor for trends
  - The test cannot be used to determine if virus is viable or infectious
- The data pipeline is adaptable: NWSS has developed a method for integration of monkeypox wastewater data into DCIPHER

NWSS collaborators and monkeypox wastewater surveillance

- There have been recent reports of detection of monkeypox virus DNA in wastewater in multiple states.
- CDC scientists are working with academic groups and health departments to learn more about these reports and have confirmed that evidence of monkeypox virus DNA is present in the wastewater samples

MPX Virus DNA Levels over time (normalized values of MPX to human fecal indicator, in cp/g)



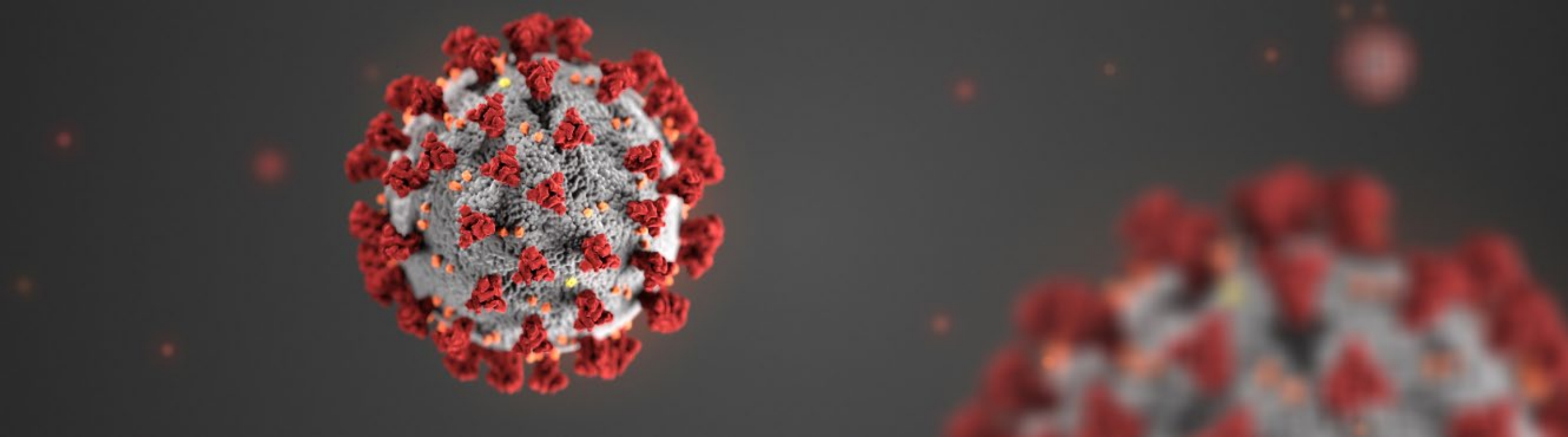
Adapted from Wolfe et al. 2022, preprint

Future directions: how to adapt NWSS to support the monkeypox response

- The NWSS testing and data platform can be adapted to monitor for monkeypox virus in addition to SARS-CoV-2
- Wastewater surveillance can help us monitor for disease at the community level—with a single sample representing communities of hundreds to millions of individuals
- Close partnership with case surveillance and health departments will be key

Challenges and research gaps

- How many people in a community would need to be infected for scientists to be able to detect the virus in wastewater?
- How long do positive signals last and how they correlate to clinical case counts?
- What does it mean for a community if the virus is detected in their wastewater?
- What method would be best for detecting the virus in wastewater, maximizing scientists' ability to detect infections but minimizing the risk for false positives?
- Is monkeypox virus viable and/or infectious in untreated wastewater?



For more information, contact CDC
1-800-CDC-INFO (232-4636)
TTY: 1-888-232-6348 www.cdc.gov

For more information: NWSS@cdc.gov
www.cdc.gov/NWSS

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.

FDA Guidance on Monkeypox Diagnostic Tests

Office of In Vitro Diagnostics
Office of Product Evaluation and Quality (OPEQ)
CDRH | Food and Drug Administration

- MPXDx@fda.hhs.gov
- https://www.fda.gov/about-fda/center-devices-and-radiological-health/subscribe-cdrh-email-lists
- CDRHWebinars@fda.hhs.gov

*These slides present high level discussion points. Please refer to the guidance document for the details and official policies.

Actions Taken September 7, 2022

FDA Takes Significant Action to Help Expand Access to Testing
www.fda.gov/news-events/press-announcements/monkeypox-update-fda-takes-significant-action-help-expand-access-testing

- HHS Secretary issues 564 declaration for the emergency use of IVDs for monkeypox
  www.hhs.gov/about/news/2022/09/07/hhs-secretary-becerra-issues-564-declaration-to-expand-the-availability-of-testing-for-monkeypox.html
- FDA Guidance: Policy for Monkeypox Tests to Address the Public Health Emergency
  www.fda.gov/regulatory-information/search-fda-guidance-documents/policy-monkeypox-tests-address-public-health-emergency
- FDA issues first monkeypox emergency use authorization (EUA)
  www.fda.gov/medical-devices/emergency-use-authorizations-medical-devices/monkeypox-emergency-use-authorizations-medical-devices

*These slides present high level discussion points. Please refer to the guidance document for the details and official policies.

Assistance for Monkeypox Test Developers

- EUA Templates to assist developers
  www.fda.gov/medical-devices/emergency-use-authorizations-medical-devices/monkeypox-emergency-use-authorizations-medical-devices#templates

- Frequently Asked Questions on Testing for Monkeypox
  www.fda.gov/medical-devices/monkeypox-and-medical-devices/faqs-testing-monkeypox

- Virtual Town Halls / Webinar on the Policy for Monkeypox Tests
  www.fda.gov/medical-devices/workshops-conferences-medical-devices/medical-device-webinars-and-stakeholder-calls
  Submit questions in advance to: CDRHWebinars@fda.hhs.gov with "*Question for the Monkeypox Tests Webinar*" in the subject line.

- Email questions, pre-EUAs, and EUA submissions to: MPXDx@fda.hhs.gov

Policy for Monkeypox Tests To Address the Public Health Emergency

- Describes FDA's review priorities of emergency use authorization (EUA) requests for monkeypox diagnostic tests
- Describes FDA's enforcement policies for certain diagnostic tests that are developed by and performed in a single-site laboratory certified under the Clinical Laboratory Improvement Amendments (CLIA) that meets the requirements to perform tests of high complexity
- Provides recommendations for diagnostic test validation
- Describes FDA's enforcement policies for FDA-cleared or authorized monkeypox diagnostic tests that are modified
- Describes FDA's enforcement policies for certain serology tests

EUA Request Review Priorities

For monkeypox (MPX) diagnostic tests, FDA intends to prioritize review of EUA requests for:

- **High-throughput diagnostic** tests
- Tests with **home specimen collection**
- **Rapid diagnostic tests**

All from experienced developers with high manufacturing capacity that:

- Inform FDA (within 30 days of the guidance) of their intent to submit an EUA request

*Details in Section IV.A.1 of the Monkeypox Test Guidance

Overview for Laboratories Developing MPX Diagnostic Tests

- **FDA will *not* expect EUA requests** for *certain* MPX diagnostic tests when the laboratory notifies FDA (within 5 business days of offering test or from date of guidance if already being offered)
  - **Developed and performed in a single site CLIA-certified laboratory** certified to perform tests of **high complexity**;
  - Molecular **PCR** technology;
  - **Lesion swabs samples**; and
  - Appropriately **validated.**

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nce.

- **FDA will *not* expect EUA requests** for *certain* validated modifications to a cleared or authorized MPX diagnostic test with notification to FDA

*Details in Section IV.A.2 & IV.A.3 of the Monkeypox Test Policy

Overview for Commercial Manufacturers of MPX Diagnostic Tests

- FDA expects developers to **submit** an EUA request or premarket submission and **receive authorization** or clearance **prior to offering or distributing** a monkeypox test
  - Inform FDA within 30 days of guidance of intent to submit an EUA request – MPXDx@fda.hhs.gov

- **FDA does not intend to object to implementation** of *certain* modifications to a developer's own cleared or authorized MPX diagnostic test while FDA conducts its review

*Details in Section IV.A.1 & IV.A.3 of the Monkeypox Test Policy

Monkeypox Diagnostics: Validation & Templates

- Validation Recommendations in Voluntary Templates
  www.fda.gov/medical-devices/emergency-use-authorizations-medical-devices/monkeypox-emergency-use-authorizations-medical-devices#templates
  - EUA Summary Template for Developers of Molecular Diagnostic Tests for Monkeypox
  - EUA Template for Developers of Molecular Diagnostic Tests for Monkeypox
- FDA intends to update recommendations as appropriate as the outbreak evolves

*Details in Section IV.B and Section V of the Monkeypox Test Policy

Monkeypox Serology Tests

- Not used to diagnose, or aid in the diagnosis of, an active infection
- Not tests of immunity
- May further understanding of the disease process

FDA does not intend to object to the use of monkeypox tests developed and performed in a high-complexity CLIA-certified laboratory that is part of an entity that conducts research on diseases and is integrated into the direct medical care of the patient (often referred to as academic medical center laboratories) where:

- the laboratory **notifies FDA** of validation, and
- certain information is included in the test reports.

*Details in Section IV.C of the Monkeypox Test Policy

First Monkeypox EUA

Quest Diagnostics Monkeypox Virus Qualitative Real-Time PCR
www.fda.gov/medical-devices/emergency-use-authorizations-medical-devices/monkeypox-emergency-use-authorizations-medical-devices#molecular

- Intended to detect monkeypox and other non-variola *Orthopoxvirus* DNA using lesion swab specimens

Resources for Monkeypox Test Development and Validation

How to Receive Updates/Alerts and Ask Questions by Email:

- Subscribe to CDRH Email Lists: www.fda.gov/about-fda/center-devices-and-radiological-health/subscribe-cdrh-mailing-lists
  - **Monkeypox and Medical Devices**
  - **In Vitro Diagnostics**
- For questions about Monkeypox IVD EUAs, email: MPXDx@fda.hhs.gov

Where to Find Information:

- Monkeypox and Medical Devices: www.fda.gov/medical-devices/emergency-situations-medical-devices/monkeypox-and-medical-devices
- Monkeypox Emergency Use Authorizations for Medical Devices: www.fda.gov/medical-devices/emergency-use-authorizations-medical-devices/monkeypox-emergency-use-authorizations-medical-devices
- Policy for Monkeypox Tests To Address the Public Health Emergency: www.fda.gov/regulatory-information/search-fda-guidance-documents/policy-monkeypox-tests-address-public-health-emergency

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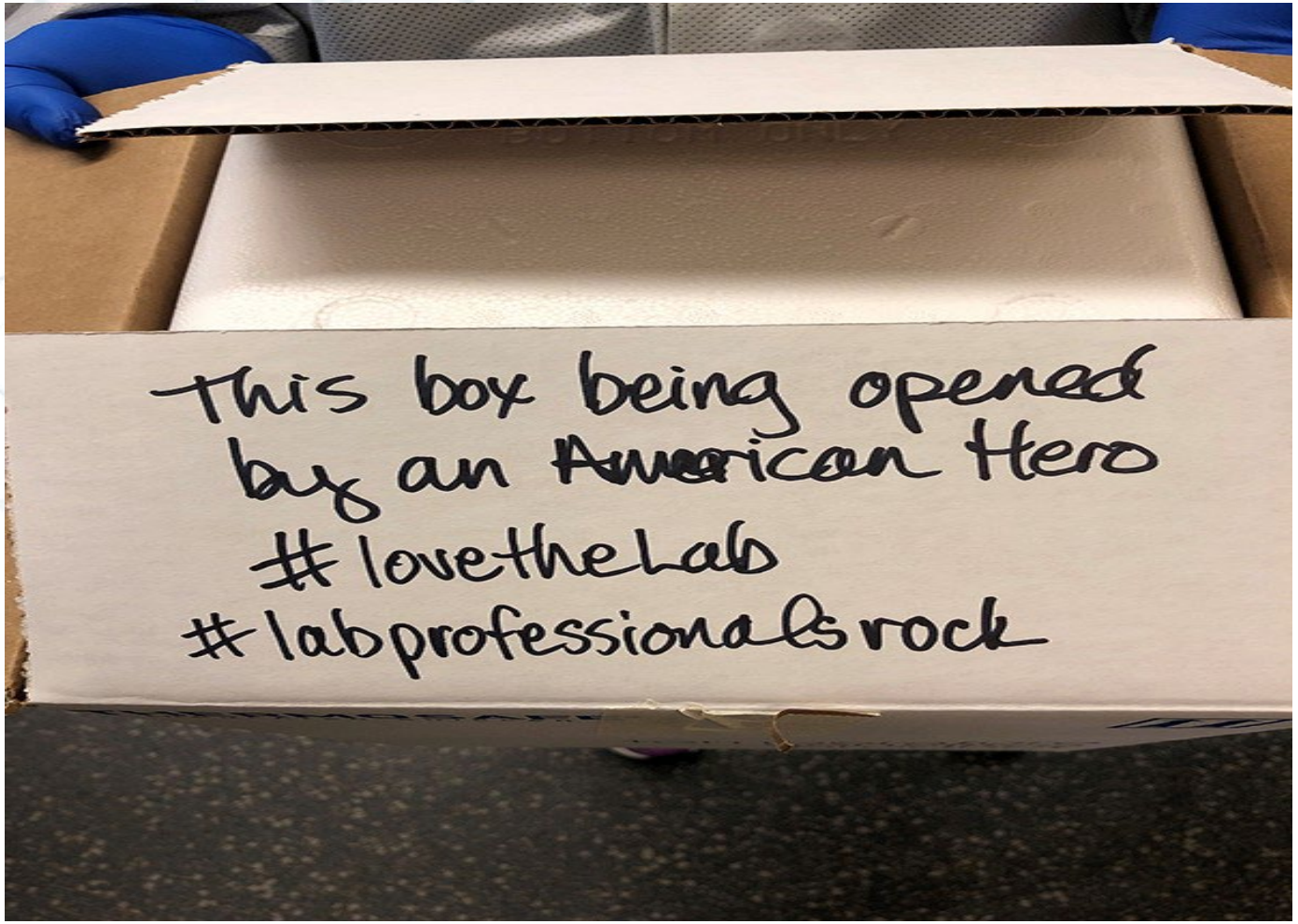

Photo submitted by the Microbiology Laboratory at The University of Pittsburgh Medical Center

For more information, contact CDC
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