

PUBLIC HEALTH LABORATORY NEWSLETTER

SARS-CoV-2 Sequencing Begins at the State Public Health Laboratory

Submitted by: Victoria N. Stone, PhD, PH Lab Consultant 2 and Christina Moore, MLS(ASCP)^{CM}, PH Laboratory Manager 2

At the beginning of July 2021, the TN State Public Health Laboratory began sequencing severe acute respiratory syndrome coronavirus 2, the cause of COVID-19. Sequencing of SARS-CoV-2 is an important public health tool for combating the virus. Early sequencing allowed researchers to decode the genome and identify SARS-CoV-2, which ultimately led to the rapid development of diagnostic assays and life-saving therapeutics. Sequencing also enhances epidemiological surveillance as it can help detect transmission patterns and provides data needed to identify outbreak clusters. But as the virus has continued to spread, it has evolved, making genomic sequencing even more critical. Some of these genetic changes or “variants” are of global concern because they may be able to

spread easier, cause a more severe infection, or evade a natural or vaccine-induced immune response. To track these variants, the CDC has established the National SARS-CoV-2 Strain Surveillance system that focuses on integrating next-generation sequencing into the national COVID-19 response plan.

To implement sequencing at the state lab, we validated a version of the ARTIC protocol (artic.network/ncov-2019) modified to work with our current PulseNet sequencing protocols. The ARTIC protocol relies on direct amplification of the nucleic acid extracted from the virus using tiled, multiplexed primers. This method has been adopted by numerous research and public health facilities worldwide as it has proven to have high sensitivity and works directly from clinical samples. Our modified protocol generates these ARTIC

amplicons but uses our current Illumina sequencing library prep method, providing us a capacity to sequence 94 samples per run. Data will be analyzed in-house using a bioinformatics pipeline developed by the Florida Bureau of Public Health Laboratories. This gives us the ability to submit data to national repositories such as GISAID and GenBank as well as provide SARS-CoV-2 lineage information in real-time. Additional sequencing and data analysis methods may be adopted in the future to expand our capacity.

The lab and state epidemiologists are working to develop a testing strategy, which will involve sequencing historical samples for national surveillance purposes. Guidance for new specimen submissions will be released soon and will include contact information, required sample criteria, and shipping instructions.

References:

Genomic sequencing of SARS-CoV-2: a guide to implementation for maximum impact on public health. Geneva: World Health Organization; 2021. License: CC BY-NC-SA 3.0 IGO.
<https://www.cdc.gov/coronavirus/2019-ncov/variants/variant-surveillance>

Rapid Automated Molecular Identification of *Candida auris*

The multidrug-resistant yeast *Candida auris* is a serious emerging global health threat. Increasing numbers of infections have been reported in over thirty countries, including the United States, since it was first identified in 2009. Outbreaks of *C. auris* are becoming problematic in long-term healthcare settings due to the ability of this organism to colonize patients for extended periods of time, persist on

Submitted by: Nicole Braun West, PhD, M (ASCP)^{CM} | ARLN PH Laboratory Scientist 4



Photo credit: www.bd.com

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contaminated surfaces for several weeks and be spread easily between patients. Identification of this resistant pathogen is often difficult via standard laboratory methods, creating a need for specialized technology to reduce any misidentification which may result in delayed treatment. The

TDH Antibiotic Resistance Laboratory Network Laboratory currently screens for *C. auris* colonization using a manual real-time PCR assay for molecular detection and has recently acquired new instrumentation to utilize an automated approach in addition to the current testing

platform. The BD MAX™ System is a rapid and automated sample-to-result real-time PCR assay which can be utilized for the identification of *C. auris*. Once fully implemented, this new technology will allow TDH ARLN public health scientists to rapidly and accurately identify *C. auris* with limited hands-on time.

References:

<https://www.cdc.gov/fungal/candida-auris/index.html>

Leach L, Russell A, Chaturvedi S, Chaturvedi V. 2019. A rapid and automated sample-to-result *Candida auris* real-time PCR assay for high throughput testing of surveillance samples with BD MAX™ open system. *J Clin Microbiol* 57:e00630-19.

SPOTLIGHT ON SAFETY

***Burkholderia pseudomallei*: Beware and Be Safe in the Clinical Laboratory**

Submitted by: Rolinda Eddings, MT(ASCP), Safety Officer

There are recent case reports of melioidosis from several states in the U.S. What would you do if you found *Burkholderia pseudomallei* from a culture in your laboratory? *Burkholderia pseudomallei* is the causative agent of melioidosis in humans. The organism is readily aerosolized from its environmental reservoirs and causes outbreaks of disease where it is endemic. While uncommon, laboratory-associated cases of melioidosis can occur. *B. pseudomallei* is considered a biothreat agent and needs to be treated as a select agent. Melioidosis is endemic in tropical regions of the world with most cases reported from rice-growing regions of SE Asia and northern regions of Australia. In the U.S., the disease is considered imported from endemic areas of the world. The disease can remain latent for years and reactivate. Since melioidosis has multiple signs and symptoms, it may be mistaken for other respiratory illnesses.

Burkholderia pseudomallei should be suspected and cannot be ruled out if the isolate meets the following criteria:

- Gram negative coccobacilli (may be slightly curved and bipolar)
- Growth on BAP as greyish-white colonies, becoming dry and wrinkled with age
- Growth on MAC in 48 hours
- Oxidase Positive
- Indole Negative
- Resistant to polymyxin B or colistin
- Colonies are non-hemolytic without violet pigment on Mueller Hinton
- May have musty odor (cannot be used to rule out and no sniffing of plates)



Use caution and follow ASM Sentinel Clinical Laboratory Guidelines

https://asm.org/Articles/Policy/Laboratory-Response-Network-LRN-Sentinel_Level-C

Contact your LRN Reference Lab to:

- Consult on performing rule-out testing with technical and biosafety guidance
- Decide if an isolate should be sent for additional testing with packaging and shipping guidance
- Discuss rule-out and confirmatory testing in BSL-3 laboratory space at the LRN reference lab
- In Tennessee, the State Public Health Laboratory is the LRN Reference Lab. The Bioterrorism Coordinator can be reached at 615-406-3792.

Use of automated identification algorithms (i.e. MALDI-TOF, 16s, VITEK-2) may misidentify *B. pseudomallei*. Misidentifications include: *Burkholderia thailandensis*, *Burkholderia cepacia*, *Chromobacterium violaceum*, *Ochrobactrum anthropic*, *Pseudomonas spp.* *Acinetobacter spp.* and *Aeromonas spp.*

References: APHL and ASM

Updated Requisitions

Clinical Submission Requisition (PH-4182)

- [https://www.tn.gov/content/dam/tn/health/program-areas/lab/requisitions/PH-4182 Clinical Submission Requisition rev 9-21.pdf](https://www.tn.gov/content/dam/tn/health/program-areas/lab/requisitions/PH-4182%20Clinical%20Submission%20Requisition%20rev%209-21.pdf)

COVID-19 Submission Requisition (PH-4360)

- [https://www.tn.gov/content/dam/tn/health/program-areas/lab/requisitions/PH-4360 COVID-19 Test Requisition Form rev 09-21.pdf](https://www.tn.gov/content/dam/tn/health/program-areas/lab/requisitions/PH-4360%20COVID-19%20Test%20Requisition%20Form%20rev%2009-21.pdf)

Please discontinue use of previous versions and use the updated versions for all future requests.

2020 Update to CDC's Gonococcal Treatment Guidelines

Submitted by: Zachary Perry, M(ASCP)^{CM}, PH Laboratory Manager 1

Nationwide, sexually transmitted infections of *Neisseria gonorrhoeae* have increased 63% since 2014. This increase, combined with *N. gonorrhoeae*'s ability to acquire antimicrobial resistance, necessitates evolving treatment recommendations. The Tennessee Department of Health, along with three other regional laboratories, provide antimicrobial susceptibility testing data to the CDC to assist with decisions regarding treatment guidelines.

TDH's ARLN gonococcal laboratory section performs AST testing on 2,000-3,000 *N. gonorrhoeae* isolates annually. Alert breakpoints are monitored for cefixime, ceftriaxone and azithromycin, to identify isolates of interest and potential emerging resistance. CDC's revised guidelines now recommend a single 500mg intramuscular dose of ceftriaxone for treatment of *N. gonorrhoeae* infection. This was updated from the 2010 recommendation of a 250mg

intramuscular dose of ceftriaxone and a single 1g oral dose of azithromycin. Through TDH's ARLN section and its partner labs' testing, CDC has seen increased incidence of azithromycin resistance, which led to the new recommendations. This data is illustrated in the figure below.

TDH continues to monitor antimicrobial resistance in *N. gonorrhoeae* and provide related data to the CDC.

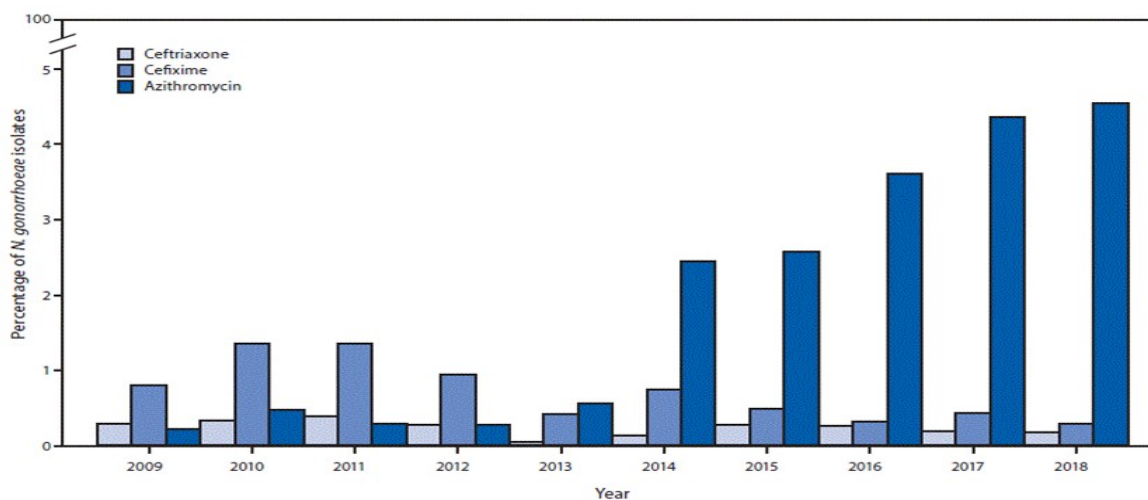


FIGURE. Percentage of *Neisseria gonorrhoeae* isolates with elevated minimum inhibitory concentrations (MICs)* to ceftriaxone, cefixime, and azithromycin — Gonococcal Isolate Surveillance Project, United States, 2009–2018

Reference

St. Cyr S, Barbee L, Workowski KA, et al. Update to CDC's Treatment Guidelines for Gonococcal Infection, 2020. *MMWR Morb Mortal Wkly Rep* 2020;69:1911–1916. DOI: <http://dx.doi.org/10.15585/mmwr.mm6950a6>

TRAINING NEWS

Packing and Shipping Infectious Substances Training

CDC/TDH Packing & Shipping Division 6.2 & 9 Infectious Substances Training

TDH has partnered with CDC to offer live, virtual Infectious Substance Packing and Shipping training. This two-day course is appropriate for those responsible for packing, marking, labeling and documentation of shipments for the transportation of Division 6.2 infectious substance and dry ice. This training satisfies a portion of the requirements however, each facility is responsible for assuring that appropriate packing instructions are adhered to as required by federal law and air transport standards.

This course will be held virtually over two, half-day sessions. Participants must attend both sessions and successfully complete the examination in order to receive credit for this course. This course is approved for 7.5 hours of P.A.C.E.® credit. Registration, attendance and examination will be facilitated through [CDC TRAIN](#). Admission preference will be given to laboratory staff who pack and ship infectious substances working in Tennessee laboratories. Please visit the TDH Laboratory Training and Workshop webpage for upcoming dates and more information.

Please see the [IATA Dangerous Goods Training Guidelines—Appendix H](#) regarding upcoming IATA Training Requirements changes effective January 1, 2023.

The Safe Transport of Division 6.2 Infectious Substances, Biological Specimens, Dry Ice & Related Materials

Delivered by Saf-T-Pak and supported by the Association of Public Health Laboratories

Participants in Saf-T-Pak's one-day virtual seminars receive the comprehensive Saf-T-Pak Training Reference Manual and take part in hands-on exercises to correctly pack, mark, label and document a shipment using actual UN-specification packaging. The training meets the requirements outlined by IATA, ICAO and US DOT 49 CFR regulations. This virtual seminar is designed for laboratorians who package, ship and transport Division 6.2 hazardous materials. Participants who successfully complete this program will be awarded 8.0 hours of P.A.C.E.® credit. Please visit the TDH Laboratory Training and Workshop webpage for upcoming dates and more information.

To register or for more information about these and other TDH training opportunities, please visit the TDH Laboratory Training and Workshop webpage:

<https://www.tn.gov/health/health-program-areas/lab/lab-education.html>

Rule Out or Refer Biothreat Preparedness Training

This all-day, intermediate-level workshop focuses on practical methods that clinical microbiology laboratories can use to remain alert for the agents of bioterrorism. Participants will learn about surveillance and evaluation procedures that can be integrated into the routine work of the clinical microbiology lab. Procedures for the referral of suspect cases will also be discussed. In this hands-on course, following appropriate safety precautions, participants will examine actual cultures and organisms in a laboratory setting.

Audience: Medical Laboratory Scientists and Medical Laboratory Technicians working in microbiology laboratories in Tennessee. Limited seats available. Please visit the TDH Laboratory Training and Workshop webpage for upcoming dates and more information.

EMPLOYEE NEWS

Welcome New Employees!

Robert Schell

PH Laboratory Scientist 1
Molecular Biology

Chelsea Dancy

PH Laboratory Scientist 2
Serology

Cherelle Jones

PH Laboratory Technician 2
Media Prep/Support Services

Brian Sally

PH Laboratory Scientist 1
Enterics

Devin Smith

PH Laboratory Scientist 1
Newborn Screening

Austin Hargrove

PH Laboratory Scientist 1
Sequencing

Jamahd Dias

PH Laboratory Scientist 1
Newborn Screening

Sakshi Sawarkar

PH Laboratory Technician 2
Environmental Chemistry

Lindsay Jolly

PH Laboratory Manager 4
Administration

Athena Lemon

PH Laboratory Scientist 2
Vectorborne Diseases/Molecular Biology

Kirsten Welch

PH Laboratory Scientist 1
Special Microbiology

Brian Selinsky

Contract Bioinformatician
Molecular Biology

Charles Lechner

APHL Bioinformatics Fellow
Newborn Screening

Rachel Wofford

CSTE Fellow
Vectorborne Diseases

Jillian Socea

APHL Antimicrobial Resistance Fellow
ARLN

Karen Beasley Maynard

Contract Scientist
Whole Genome Sequencing

Shcorey Shepard

Contract Scientist
Serology

Woody Dewberry

Contract Administrative Secretary
Knoxville Regional Laboratory

Kaela Gardner

Contract Scientist
Molecular Biology

Elora Fullerton

Contract Administrative Secretary
Nashville Central Laboratory

Natarina Gore

Administrative Assistant
Administration

Promotions

Charlotte Corter

Administrative Services Assistant 3
Administration

Penny Coe

PH Laboratory Scientist 2
ARLN

Nicole Braun West

PH Laboratory Scientist 4
ARLN

Dana Baker

PH Laboratory Scientist 2
Newborn Screening

Patzili Loera

PH Laboratory Scientist 2
ARLN

Christopher Turrill

PH Laboratory Scientist 1
ARLN

Retirements

Timothy Morris

PH Laboratory Consultant 1—Environmental
9 Years of Service

Interested in working at the Public Health Laboratory?

Visit www.TN.gov/careers to view current
employment opportunities!

*The Mission of Laboratory Services is to provide quality testing services through innovation,
collaboration, and education that protects and improves the health of all.*



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