

HOT (or Warm!) TOPIC: *Legionella* Part 2

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September 9, 2024



Local Health Department (LHD) *Legionella* Case Review

- ❖ Early July storms
- ❖ Epidemiology had *Legionella* cases come their way
- ❖ Hot tub in question
 - Drained before sampling
 - Pool already went through hyperchlorination
- ❖ Reportable Disease section informed of outbreak
- ❖ LHD working on the fly with Kentucky Division of Laboratory Services (DLS) to prep for sample collection

General Lab Overview: Initial Inquiries

- ❖ Meeting of the Minds
 - Epidemiology
 - Environmentalists
 - Lab Analysts
- ❖ Transfer of Information
 - Background information
 - Determine potential sample locations
 - Create kits based on needs
 - » Lab cannot instruct *how* to collect
 - » Lab can assist with sample parameters upon arrival to lab



July 3, 2023

❖ Epidemiology

- Emails sent to Environmental Microbiology supervisor and Global Preparedness and Environmental Branch Manager giving heads-up that a Legionella case forming with a connection in Lexington, KY
 - » DLS approved to test if samples collected
 - » Tentative date was proposed for collection

July 4, 2023

❖ DLS closed for holiday



July 5, 2023

❖ Environmental

- Reached out to DLS for sampling kits
- Arranged pick-up



July 6, 2023

- ❖ Environmental
 - Quantities of supplies verified
- ❖ Laboratory
 - Kits created based on given quantities
 - Met with Environmental at DLS to exchange hands
 - Gave basic instructions on what would constitute satisfactory samples
 - Received and started processing samples
- ❖ Epidemiology/Environmental
 - Stayed in contact with DLS staff throughout collection process to gain assistance with questions as arose

July 7, 2023

- ❖ Environmental/Epidemiology
 - Reached out to DLS for assistance with Outreach entry
- ❖ Laboratory
 - Back and forth with internal Outreach partners and Environmental/Epidemiology to assist with Outreach issues until complete

July 13, 2023

❖ Laboratory

- Testing completed
- Results reported



Environmental Water Sampling



Request KDLS Outreach Account

- ❖ Two forms required for access to the Kentucky Laboratory Web Outreach (KY-LWO) system; allow a few days for account creation
 - ❖ HIPAA form
 - ❖ Kentucky Laboratory Web Outreach Access Request Form

- ❖ Outreach Test Code: WLEG *L. pneumophila* Testing

<https://outreach2.psychesystems.com/netoutreachks/>
<https://www.chfs.ky.gov/agencies/dph/dls/Documents/HIPAAForm.pdf>
<https://www.chfs.ky.gov/agencies/dph/dls/Documents/KDLSOutreachAccessRequestForm.pdf>
<https://www.chfs.ky.gov/agencies/dph/dls/Documents/OutreachManual.pdf>

Environmental Water Sampling Continued



Request Environmental Laboratory Kits

- ❖ Requisition form for ordering lab kits
 - Environmental: Water Bacteriology Kits
 - MUST specify *Legionella* samples

Collection Updates

- ❖ Determine if kits have intended pieces
- ❖ Collection
 - If any discrepancies, contact the lab to inform of changes
 - Lab may need to order additional media
- ❖ Lab can assist with Outreach sample submissions if needed



<https://www.chfs.ky.gov/layouts/download.aspx?SourceUrl=https://www.chfs.ky.gov/agencies/dph/dls/Documents/RequisitionforLaboratoryKits.docx>

Collection Kit

- ❖ 100ml Bottle w/ Sodium Thiosulfate ($\text{Na}_2\text{S}_2\text{O}_3$)
- ❖ Absorbent Sheets/Plastic Bottle Bag
- ❖ 1L (Extra Large) Bottle
- ❖ 15mL Centrifuge Tube w/ Sodium Thiosulfate
- ❖ Dry Swabs
- ❖ Tube Shuttle (Holds Tubes with Sample Swabs)
- ❖ Ice Packs
- ❖ Styrofoam Cooler
- ❖ Shipping Supplies
 - Primary/Secondary Containers

*Not all items will be needed for every case. Alternative items can be requested in advance if the case is expected to be unique.

Sample Receiving: In the Lab

- ❖ Pictures (if necessary)
- ❖ Check sample integrity
- ❖ Compare Outreach forms to samples received
- ❖ Double check information given to samples received
 - Supply check
 - Order missing supplies
- ❖ Complete paperwork
- ❖ Print worksheets
- ❖ Prep supplies for testing
- ❖ Decide on testing timeline





Cabinet for Health and Family Services
Department for Public Health
100 Sower Blvd, North Loading Dock
Frankfort, Kentucky 40602-2020

Incident Information:

Name (Last, First, MI): Spa, Test
Street:
City, State, Zip Code: KY 40601

Send Report to:

Submitter/Health Dept: DIVISION OF LABORATORY SERVICES
Location: 100 SOWER BLVD #204

Specimen Information:

Date of Collection: 2023-07-19
Time of Collection: 12:00

Specimen/Account #: 
197107213WLEG

Ask at Order: Sample sequence number?:1
Permit # (for Dairy only)?:NA
Type of water?:Drinking water- other
Water sample/source related to?:Su spect
Collector's remarks?:Room 123 Shower Head
Sample number?:1
Sanitarian ID?:R1234
Public Location code?:NA
Collector?:Ted Waterbear

WLEG

Documentation: Sample Outreach Form

- ❖ All information on this page is filled out by the collector
- ❖ Most fields are mandatory
- ❖ Code for *Legionella* is WLEG

Documentation: Lab Worksheet

Lab Worksheet

- ❖ Specific to tests
- ❖ Lab equivalent to Chain of Custody
- ❖ Documents start to finish of testing
 - Work and supplies
 - Equipment

Form 30a-Water Testing Legionella
Rev. 000 Feb. 2023

KENTUCKY DIVISION OF LABORATORY SERVICES
Water Analysis Worksheet - Legionella
Legiolert & Legionella Culture
ES.DLS.IDEXXL – IDEXX Legiolert Enzyme Substrate Test
ES.DLS.LCUL – Detection of Legionella Species in Water

Samples _____ Date Received _____ Page __ of __

Test Item	Lot Number	Expiration Date
Legiolert		
Legiolert Pre-treatment		
Sterile Reagent Grade Water		
Buffer Diluent		
Bottle		
Tray		
Legiolert Positive/Negative Controls (IDEXX)		
BCYE		
BCYE + GPVC		
BCYE Bi-plate		
Legionella LAT		
<i>L. pneumophila</i> ATCC 33152		
<i>L. anisa</i> ATCC 35292		
Pipette ID(s):		
Sealer ID(s): <input type="checkbox"/> QTP13190504836 (Potable) <input type="checkbox"/> QTP13174302999 (Non-potable)		
Incubator ID(s):		
Balance ID(s):	Timer ID:	

Legiolert Start Date/Time _____ End Date/Time _____
Isolation Media Start Date/Time _____ End Date/Time _____
Bi-plate Start Date/Time _____ End Date/Time _____
Legionella Latex Agglutination Test Date/Time _____

Analyst _____
Date Reported _____
Supervisor Review/Date _____

Key: P = potable NP = non-potable T = typical AT = atypical NS = no suspect NG = no growth

Sample #	Legiolert Result	MPN/100mL	BCYE	GPVC	Bi-plate	Legionella LAT Result
Type: <u>P/NP</u>	<input type="checkbox"/> Positive <input type="checkbox"/> Negative					<input type="checkbox"/> L. pneumophila Detected, <input type="checkbox"/> sg 1 <input type="checkbox"/> sg 2-14 <input type="checkbox"/> Other Legionella spp. Detected <input type="checkbox"/> Not Detected
Type: <u>P/NP</u>	<input type="checkbox"/> Positive <input type="checkbox"/> Negative					<input type="checkbox"/> L. pneumophila Detected, <input type="checkbox"/> sg 1 <input type="checkbox"/> sg 2-14 <input type="checkbox"/> Other Legionella spp. Detected <input type="checkbox"/> Not Detected
Type: <u>P/NP</u>	<input type="checkbox"/> Positive <input type="checkbox"/> Negative					<input type="checkbox"/> L. pneumophila Detected, <input type="checkbox"/> sg 1 <input type="checkbox"/> sg 2-14 <input type="checkbox"/> Other Legionella spp. Detected <input type="checkbox"/> Not Detected
Type: <u>P/NP</u>	<input type="checkbox"/> Positive <input type="checkbox"/> Negative					<input type="checkbox"/> L. pneumophila Detected, <input type="checkbox"/> sg 1 <input type="checkbox"/> sg 2-14 <input type="checkbox"/> Other Legionella spp. Detected <input type="checkbox"/> Not Detected
Type: <u>P/NP</u>	<input type="checkbox"/> Positive <input type="checkbox"/> Negative					<input type="checkbox"/> L. pneumophila Detected, <input type="checkbox"/> sg 1 <input type="checkbox"/> sg 2-14 <input type="checkbox"/> Other Legionella spp. Detected <input type="checkbox"/> Not Detected
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Type: <u>P/NP</u>	<input type="checkbox"/> Positive <input type="checkbox"/> Negative					<input type="checkbox"/> L. pneumophila Detected, <input type="checkbox"/> sg 1 <input type="checkbox"/> sg 2-14 <input type="checkbox"/> Other Legionella spp. Detected <input type="checkbox"/> Not Detected
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Type: <u>P/NP</u>	<input type="checkbox"/> Positive <input type="checkbox"/> Negative					<input type="checkbox"/> L. pneumophila Detected, <input type="checkbox"/> sg 1 <input type="checkbox"/> sg 2-14 <input type="checkbox"/> Other Legionella spp. Detected <input type="checkbox"/> Not Detected

Documentation

- ❖ Lab accession number
- ❖ Potable/Non-Potable
- ❖ Legiolert Results
 - Overall result
 - Quantitative result
 - Plate results
 - Overall plate results

Documentation: Interpreting Results

- ❖ Final report
- ❖ Top two sections carried over from Outreach form
- ❖ Lower section is the results with method information
- ❖ Marked final at bottom of page
- ❖ IDEXX Legiolert Test Limits for *L. pneumophila*:
 - Potable Water: <1 organism/10 mL
 - Non-Potable Water: <10 organisms/1 mL
- ❖ Acceptable limits for any water source: None Detected
- ❖ Accreditation stamp/seal

Kentucky Cabinet for Health and Family Services
Department for Public Health
Division of Laboratory Services
100 Sower Blvd. Suite 204, Frankfort, KY 40601
Vaneet Arora, MD, MPH, D(ABMM)
CLIA Laboratory Director

Laboratory Report

Send Report to Submitting Facility:

DIVISION OF LABORATORY SERVICES
100 SOWER BLVD #204
FRANKFORT, KY 40601

Date/Time Flagged Inlab: 2024-02-21 14:27:07
Lab Med Rec No.: 197107212
Lab Visit No: 197107213
ID Number: ENV2024-000001
Collection Date & Time: 2023-07-19 12:00

Name: **Spa, Test**

Street:

City

State: KY

ZIP: 40601

This report contains patient information that must be protected in accordance with the Health Insurance Portability and Accountability act (HIPAA).

General Information

WLEG:

Sample sequence number?:1

Permit # (for Dairy only?):NA

Type of water?:Drinking water- other

Water sample/source related to?:Suspect

Collector's remarks?:Room 123 Shower Head

Sample number?:1

Sanitarian ID?:R1234

Public: Location code?:NA

Collector?:Ted Waterbear



WLEG laboratory report
(top of page 1)

WATER

Overall Result: Detected

Accession # 501
Date and Time Received in Lab 07/19/2023 1300 AC
Date and Time Started (mm-dd-yy 24hr) 07/19/2023 1400 AC
Date and Time Finished (mm-dd-yy 24hr) 07/26/2023 1400 AC
IDEXX Water 256 per 10mL

Division of Laboratory Services (502) 564-4446

2024-02-22

12:49

FINAL

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WLEG laboratory report
(bottom of page 1)

Kentucky Cabinet for Health and Family Services
Department for Public Health
Division of Laboratory Services
100 Sower Blvd. Suite 204, Frankfort, KY 40601
Vaneet Arora, MD, MPH, D(ABMM)
CLIA Laboratory Director

Name: Spa, Test
Lab Med Rec No.: 197107212
Lab ID No.: ENV2024-000001
Collection Date & Time: 2023-07-19 12:00

Method: IDEXX Legiolert MPN. For potable water, the Legiolert test detects Legionella pneumophila at = 1 organism/10mL. For non-potable water, the Legiolert test detects Legionella pneumophila at = 10 organisms/1mL.

Acceptable limits for any water source: None Detected

Note: Any presence of L. pneumophila will require a follow up per The Centers for Medicare and Medical Services (CMS) for healthcare facilities according to each facility's specific water management plan in place.

These results relate only to the items tested as received. The laboratory is not responsible for any sampling information that is provided by the client that may be used to determine the final concentration of target analytes. This test report shall not be reproduced except in full and not without written approval of the laboratory.

Accession # 501

Date and Time Received in Lab 07/19/2023 1300 AC

Date and Time Started (mm-dd-yy 24hr) 07/26/2023 1430 AC

Date and Time Finished (mm-dd-yy 24hr) 07/30/2023 1430 AC

WATER LEGIONELLA Culture L. pneumophila Serogroup 2-14 isolated

Method: CDC Preparation of Specimens for Bacterial Examination Mod, 2018 or current revision. Legionella LAT kit by Oxoid. Analytical sensitivity is limited to the 11 detectable species of the LAT kit. Legionella species includes: L. pneumophila 1&2-14, L. longbeachae 1&2, L. bozemanii 1&2, L. dumoffii, L. gormanii, L. jordanis, L. micdadei, L. anisa.



WLEG laboratory report
(top of page 2)

Date and Time Received in Lab 07/19/2023 1300 AC

Date and Time Started (mm-dd-yy 24hr) 07/26/2023 1430 AC

Date and Time Finished (mm-dd-yy 24hr) 07/30/2023 1430 AC

WATER LEGIONELLA Culture No Legionella species isolated *See analytical sensitivity for full list.*

Method: CDC Preparation of Specimens for Bacterial Examination Mod, 2018 or current revision. Legionella LAT kit by Oxoid. Analytical sensitivity is limited to the 11 detectable species of the LAT kit. Legionella species includes: L. pneumophila 1&2-14, L. longbeachae 1&2, L. bozemanii 1&2, L. dumoffi, L. gormanii, L. jordanis, L. micdadei, L. anisa.

Acceptable limits for any water source: None Detected

Note: Any presence of L. pneumophila will require a follow up per The Centers for Medicare and Medical Services (CMS) for healthcare facilities according to each facility's specific water management plan in place.

These results relate only to the items tested as received. The laboratory is not responsible for any sampling information that is provided by the client that may be used to determine the final concentration of target analytes. This test report shall not be reproduced except in full and not without written approval of the laboratory.



Division of Laboratory Services (502) 564-4446

2024-02-22

12:49

FINAL

Page 2 of 2

WLEG laboratory report
(bottom of page 2)

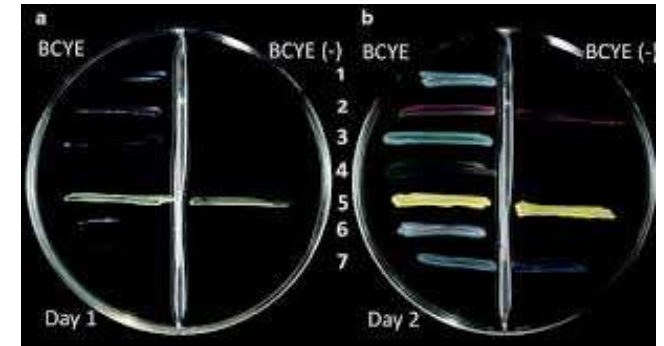
Testing Results: Culture



BCYE Agar



BCYE w/ GPVC



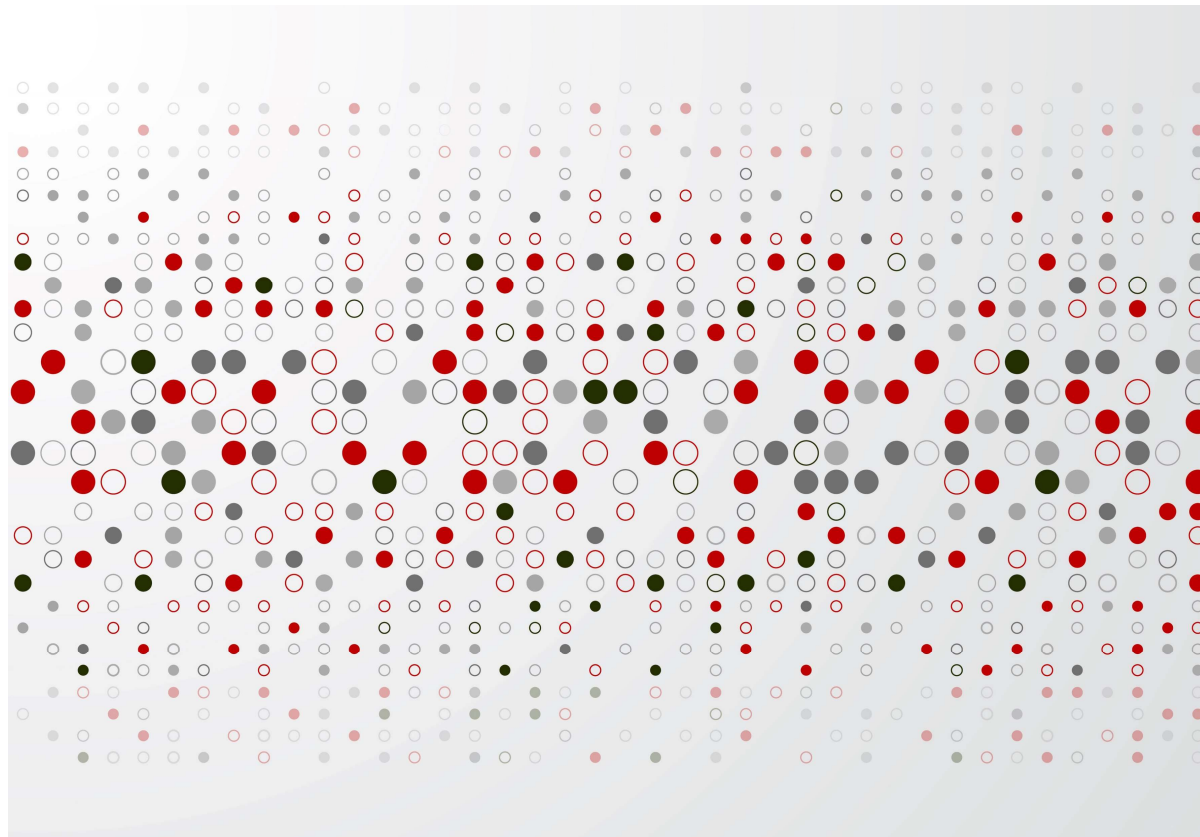
BCY w/wo Cysteine

Testing Results: Quantification



IDEXX Legiolert®

Testing Results: PCR Capabilities



DLS Takeaways

❖ Communication is Key!

- Legionella Response Group will be a beneficial step to include moving forward for future cases to help with initial steps.
- Contact directly between DLS and collectors helps the process run more smoothly and we can correct issues on the fly.
- We are creating a group email for our lab to better assist in the future.

❖ Updates Needed

- We were just as surprised about swab testing as the collectors. We were able to take their comments from the field and adapt moving forward.
- Larger containers to help with filter collection were new for us as well. We were able to find something to aid in the collection on site ASAP!
- Updated reports to clarify when multiple tests are shown.
- Triggered lab paperwork update as well to better adapt to new sampling types.

Thank you.

Ashley Cravens

ashley.aurandcravens@ky.gov

(502)782-7726



Kentucky Public Health
Prevent. Promote. Protect.



TEAM 
KENTUCKY[®]
CABINET FOR HEALTH
AND FAMILY SERVICES