

## Office of Research and Development

# **Microbial water quality of the built environment & Development of an UV-LEDs POU device for water disinfection**

**Hodon Ryu, Ph.D.**

March 22, 2023

# EPA Disclaimer

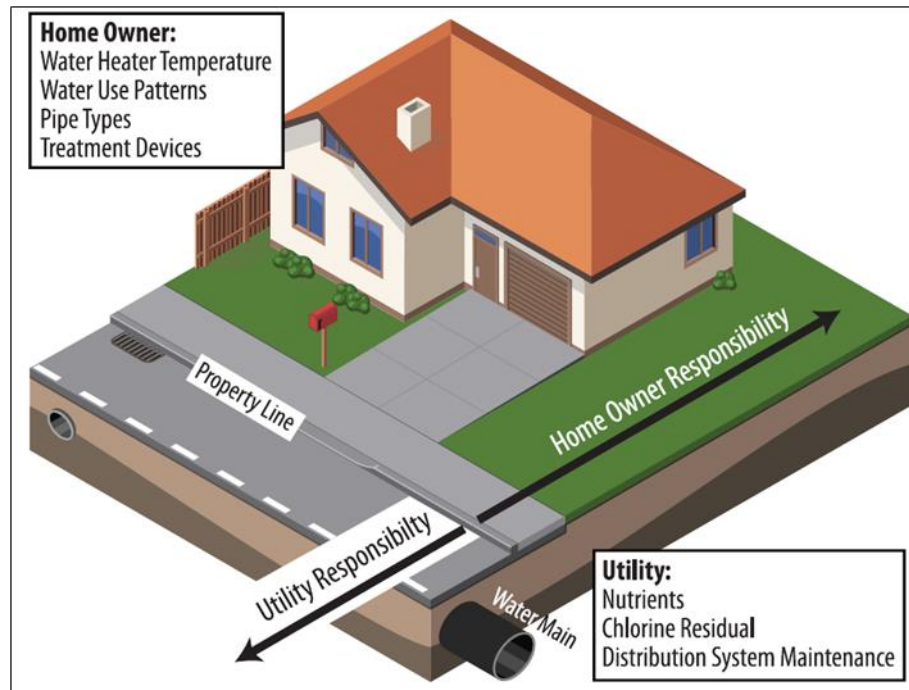
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# **1. Microbial water quality – built environment**

2. Microbial disinfection by UV-LED

# Premise Plumbing: why premise plumbing?

**Premise plumbing (PP)** includes that portion of the drinking water distribution system (DWDS) connected via the service line to houses and other buildings.



❖ *Public health data shows that a significant fraction of the nation's **waterborne disease outbreaks** are attributable to PP systems.*

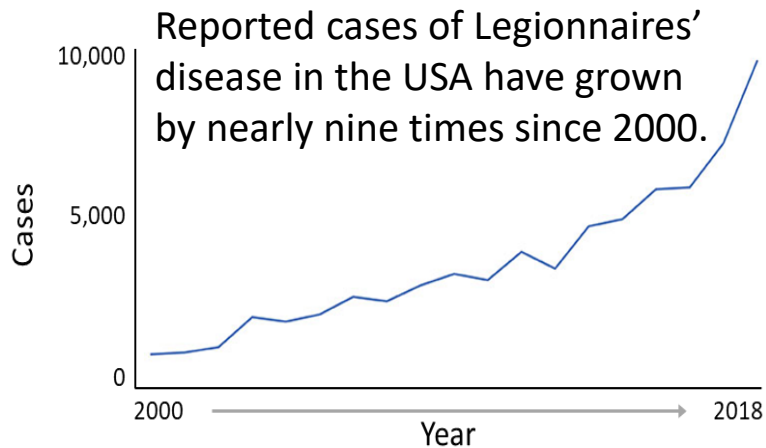
❖ *It is important to understand the characteristics of these systems which amplify the **potential public health risk** relative to the DWDS.*

# Built Environment: Opportunistic Premise Plumbing Pathogens (OPPPs)

## *Legionella*

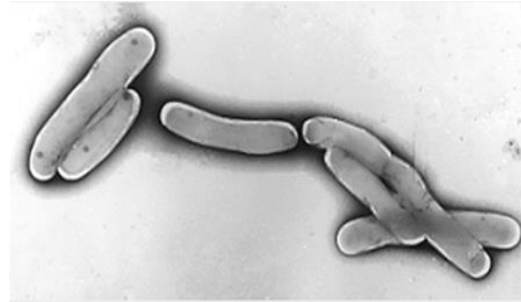


*Legionella pneumophila*



National Notifiable Diseases Surveillance System (CDC)  
<https://www.cdc.gov>.

## *Mycobacterium*



*Mycobacterium avium*

- Waterborne illness caused by **nontuberculous mycobacteria** (NTM) cost nearly \$1.8 B for in-patient and out-patient treatment in 2010. Thomson et al, 2015.
- Pulmonary NTM infections in the US are typically caused by *M. avium* and *M. intracellulare*.

## *Pseudomonas spp.*

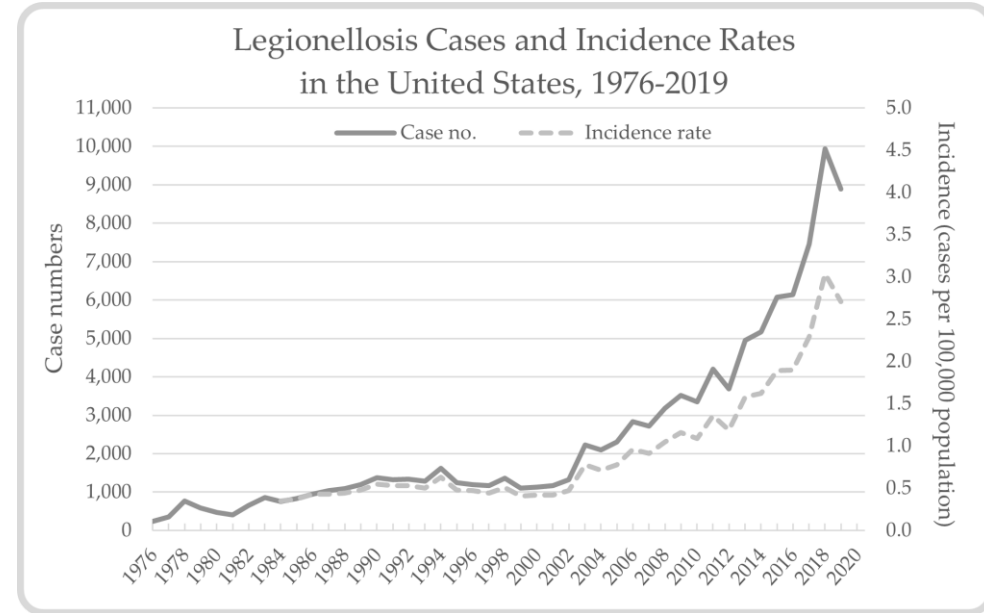


*Pseudomonas aeruginosa*

- Over 140 different species of pseudomonas.
- *P. aeruginosa* and *P. maltophilia* are associated with 80% of human infections.
- *P. aeruginosa* is most often found in POU areas, such as faucets, drains, and showerheads. Mena and Gerba, 2009

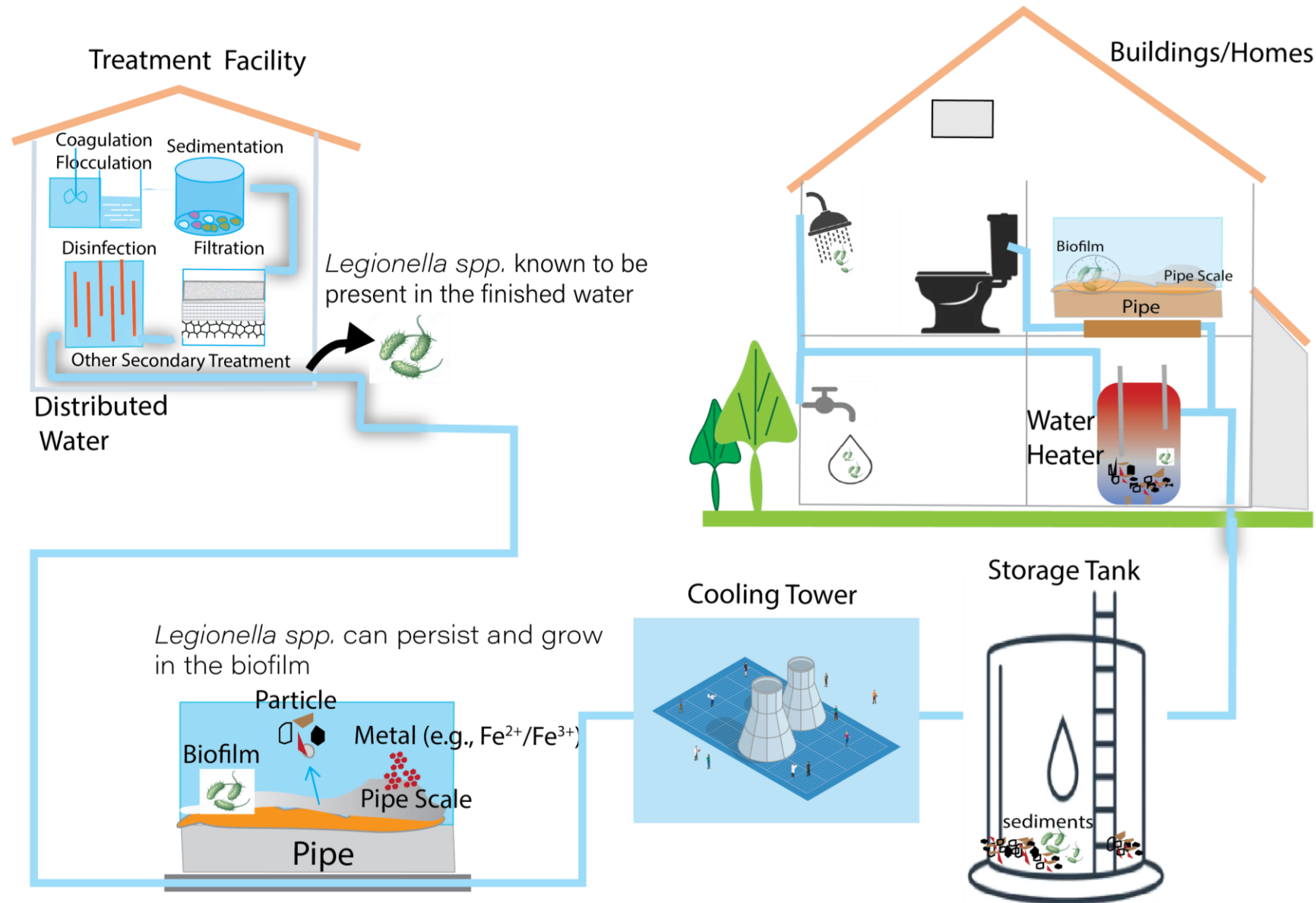
# U.S. EPA Regulation and Guidance for *Legionella*

- *Legionella pneumophila* caused the most drinking water outbreaks relative to other water-associated pathogens in the U.S.
  - Legionnaires' disease (LD) incidence has risen steadily from 0.48 cases/100,000 people during 1992-2002 to 2.71 cases/100,000 people in 2018
  - The CDC reported the highest hospitalization and death rates associated with LD amongst 7.15 million waterborne infectious disease cases
- Contaminant Candidate List 3, 4 and 5: *L. pneumophila*
- Surface Water Treatment Rule
  - Maximum contaminant goal for *Legionella*: zero
  - Require disinfection and for most systems, filtration
  - Disinfectant residual monitoring at entry point and in distribution system
- No regulation and monitoring of microbes beyond the meter



\*National Notifiable Diseases Surveillance System Graph credit to Helen Buse, U.S. EPA

# Legionella Occurrence In Engineered Environments



# Legionella Control in Premise Plumbing

## Temperature Control

- Keep hot and cold water lines at temperatures outside growth range of 25-45°C
- Heat shock: temporarily elevating the temperature at 60-70°C

## Manage Hydraulics

- Flushing has been shown to lower *Legionella* levels in drinking water
- High flow and turbulence can reduce biofilm formation
- There is no consensus on the optimal flushing frequency to prevent *Legionella*

## Disinfection

- Maintaining a disinfectant residual has demonstrated some degree of efficacy towards management of *Legionella*

## Plumbing Materials

- Each pipe material influences water chemistry and shapes the biofilms that colonize the plumbing in a unique manner

## Managing the Distal Portion of the Plumbing

- Small diameter piping in the distal portion, maximizing water circulation, and minimization of surface area
- Use of point of use devices

*National Academies of Sciences, Engineering, and Medicine, 2020; U.S. EPA, 2016*



# Manuals and Guidance of *Legionella* Control in Large Buildings



## Technologies for *Legionella* Control in Premise Plumbing Systems: Scientific Literature Review

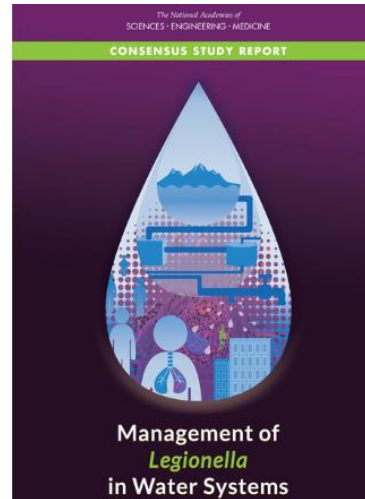
U.S. EPA, 2016



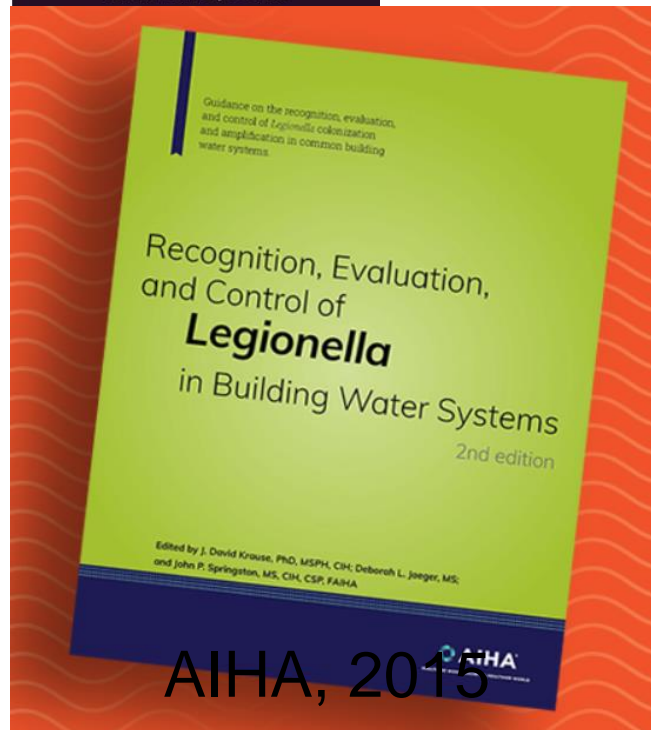
ANSI/ASHRAE Standard 188-2018  
(Supersedes ANSI/ASHRAE Standard 188-2015)  
Includes ANSI/ASHRAE addenda listed in Annex D

## Legionellosis: Risk Management for Building Water Systems

ASHRAE 188 Standard

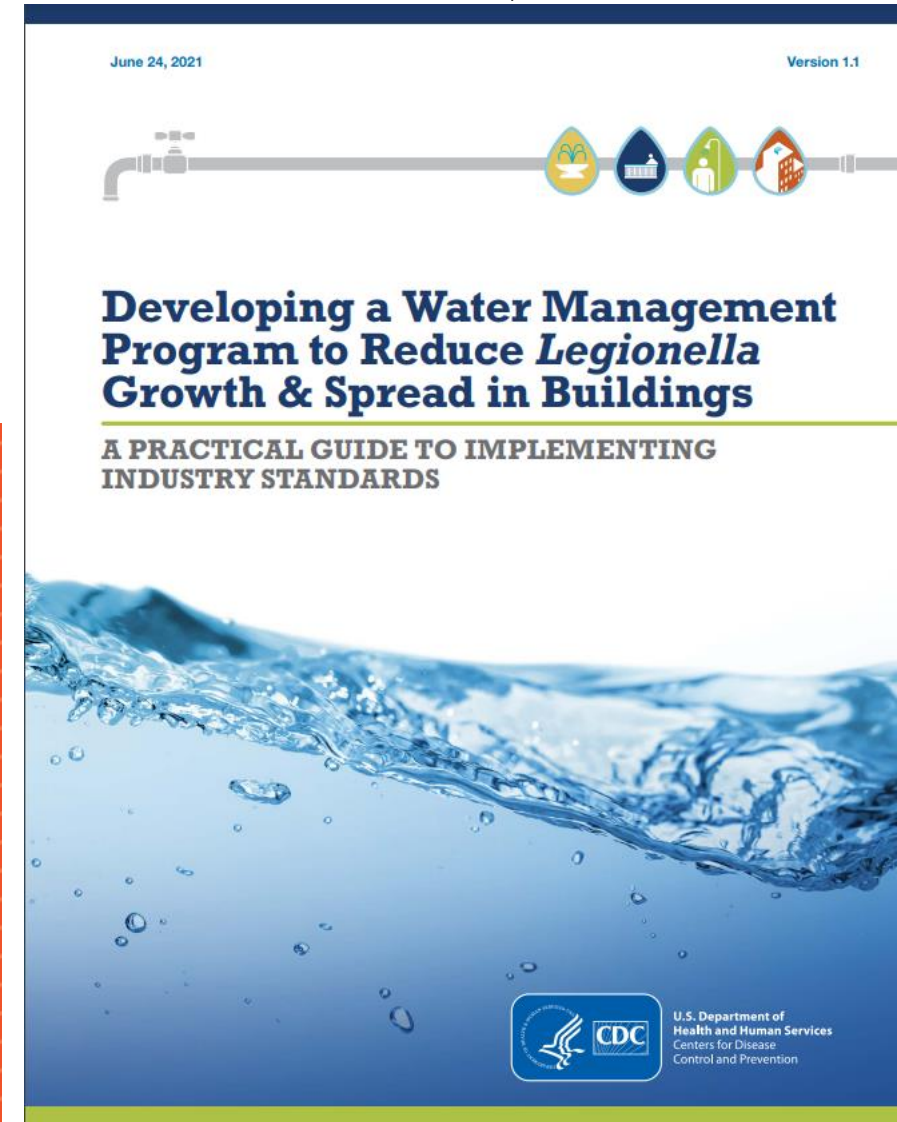


National  
Academies of  
Sciences,  
Engineering,  
and Medicine,  
2020



AIHA, 2015

CDC, 2021



# Guidance Lacking in *Legionella* Control in Residential Homes

- Hydraulic Management
  - No consensus on flushing frequency and protocol
- Temperature Control
  - Increase water heater set point to 60-70°C
  - Switch to tankless, gas water heaters
- Distal Devices
  - Cleaning of water heaters, showerheads, and faucet aerators
  - Use of point of use devices
- **Lack of data to guide homeowners to control *Legionella* in a contaminated home**

*Freije, 2010; Masters et al., 2018; Rhoads et al., 2020; National Academies of Sciences, Engineering, and Medicine, 2020; ASHRAE 188 Standard*

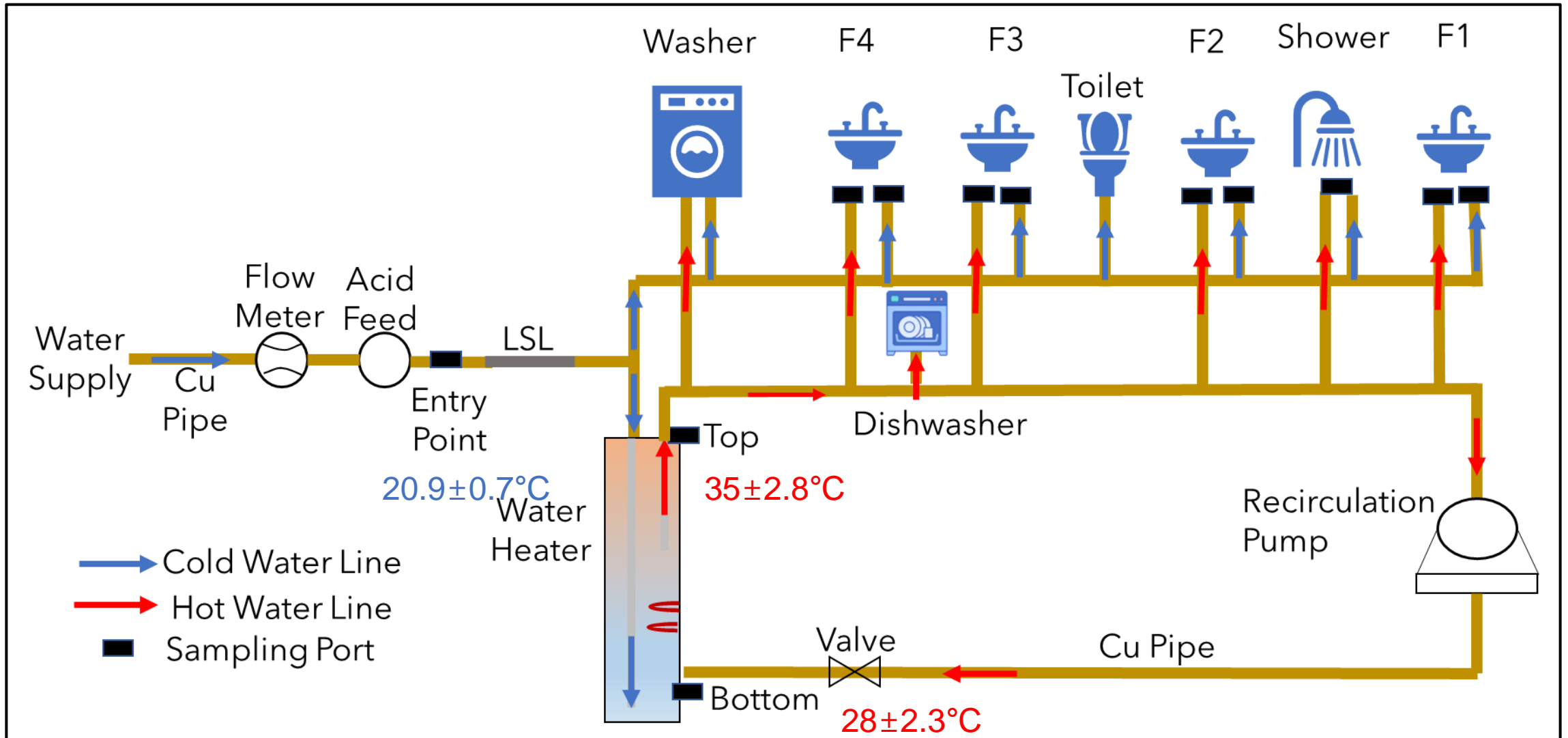
# Case study 1 - *Premise Plumbing: objectives*

This project was aimed at characterizing microbial water quality using molecular technology and its applicability for assessing treatment performance in a simulated home plumbing system.

**Obj1:** *Monitoring three major OPPPs and two phagocytic amoebae using **qPCR***

**Obj2:** *Investigating microbial communities and metabolic functional profile of bulk water and biofilms using **a metagenome and next generation sequencing technology***

# Model Home Apparatus



Located at the Andrew W. Breidenbach Environmental Research Center, Cincinnati, OH



# Model Home Apparatus



Water Heater



Washer



Dishwasher



Toilet



Shower



Faucet 1



Faucet 3

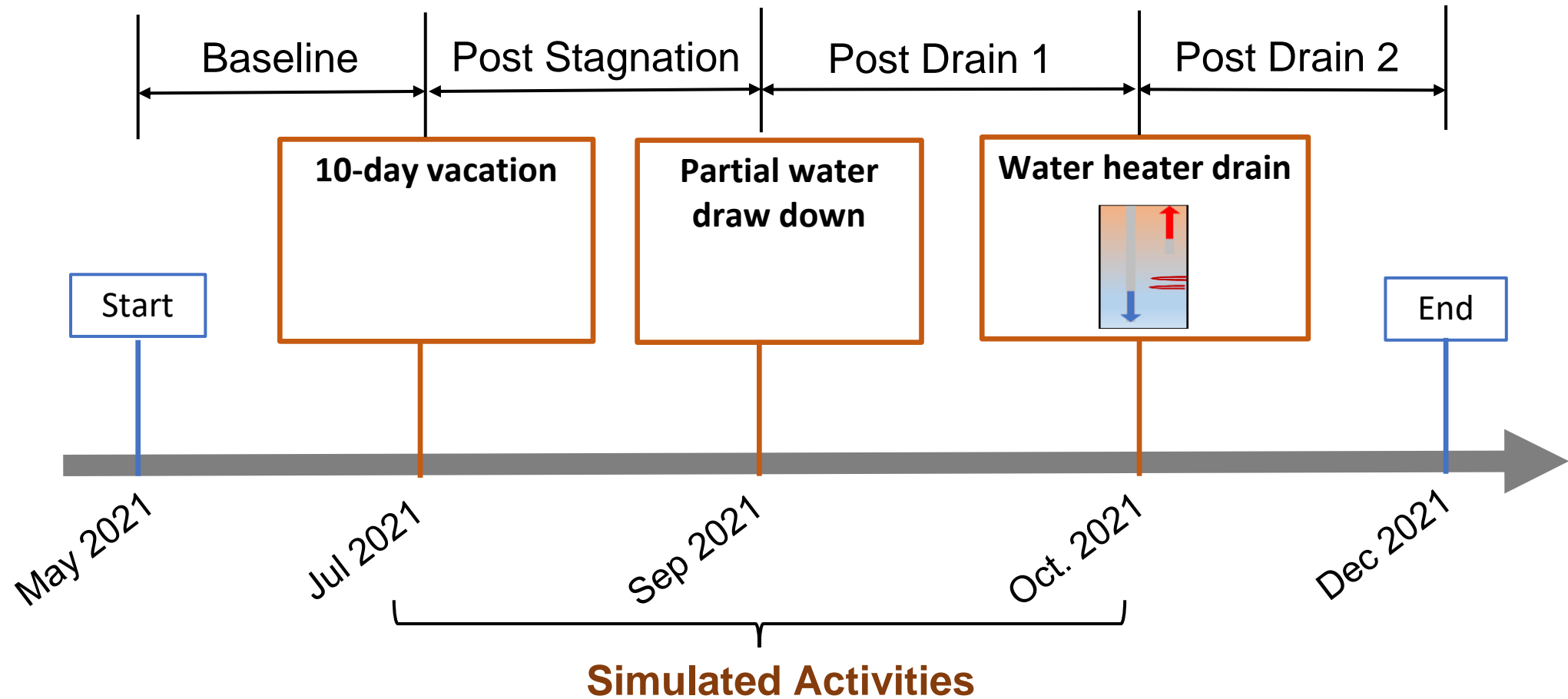


Faucet 2



Faucet 4

# Model Home Simulated Activities



# Microbial Analysis: molecular analyses

HPS Sampling

Sample Filtering

DNA Extraction

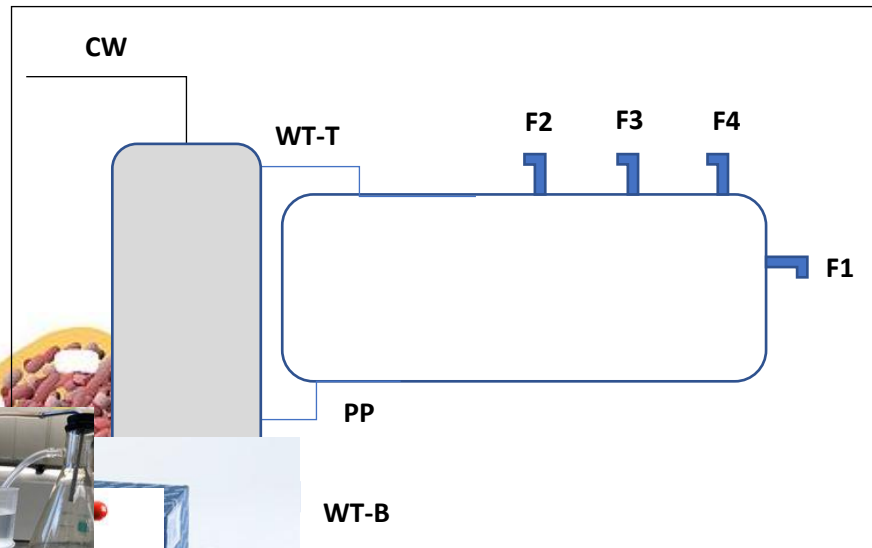
DNA Sequencing



Bulk  
water



Biofilm



DNA Extraction



13 qPCR assays  
(targeting OPPPs)



Illumina

Pacbio

Short reads

Long reads



Taxonomy and Functions

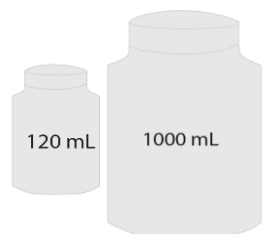
Taxonomy

# Water Sampling & Microbial Analysis

## Samples

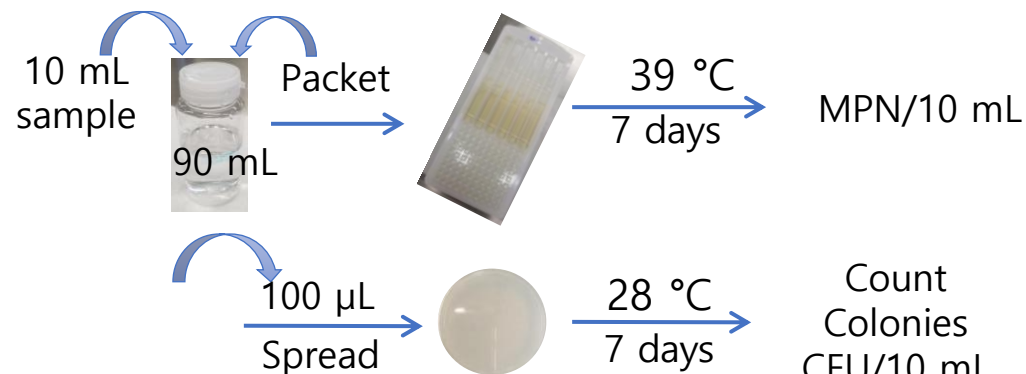
## Protocol

## Targeted Species



**Legiolert**

**Heterotrophic  
Plate Count  
(R2A agar)**

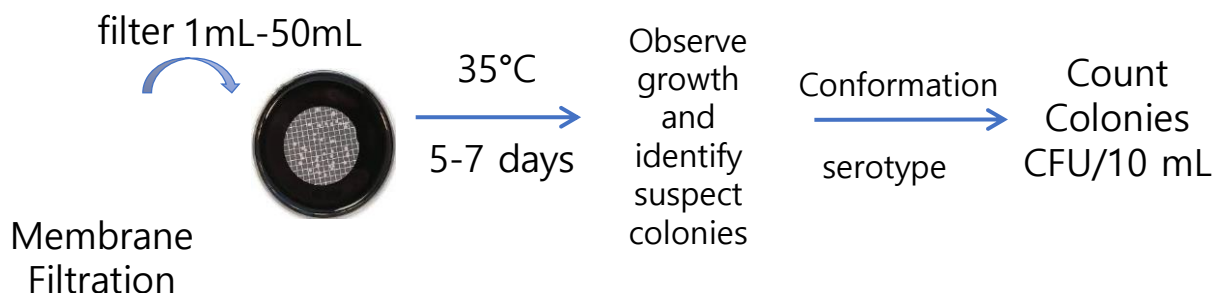


**Culturable *L. pneumophila***

**Sub-population of heterotrophs**



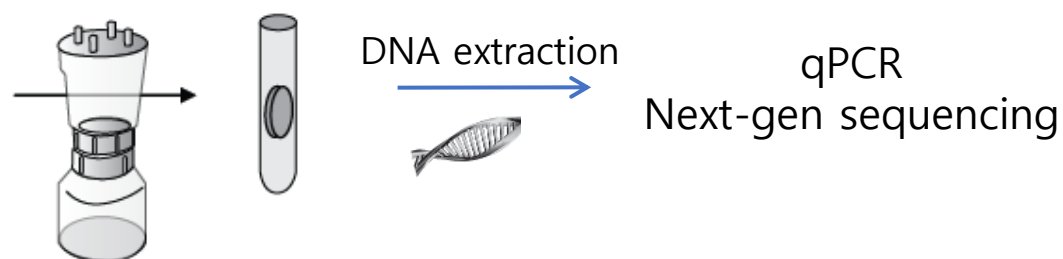
**Buffered  
Charcoal  
Yeast  
Extract Agar  
(BCYE)**



**Culturable *L. pneumophila***



**Molecular  
Analyses**

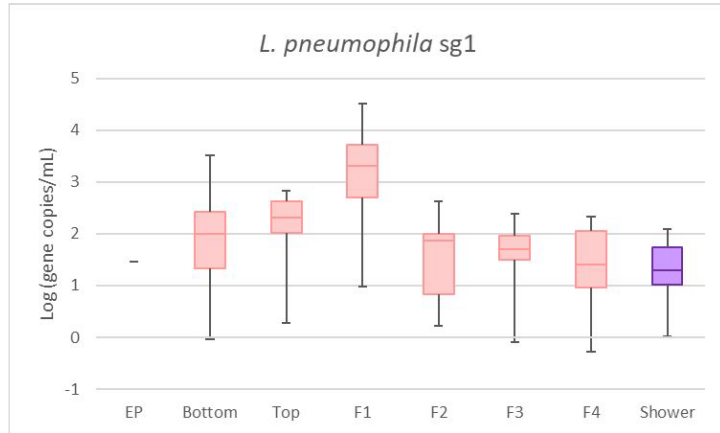


**OPPPs**  
*Microbial communities*  
*Metabolic functional profile*



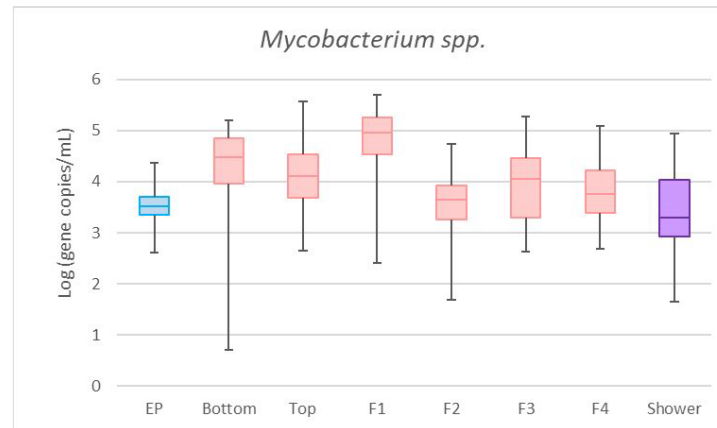
# Results - Prevalence of three major OPPPs

## *L. pneumophila*



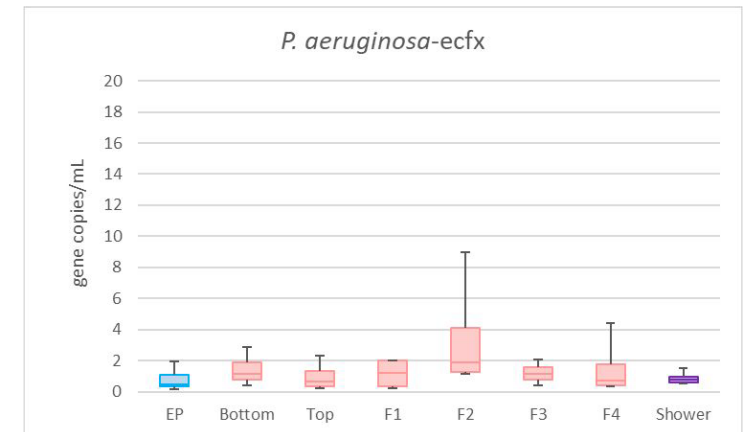
(86%, 187 positive out of 217)

## Mycobacteria



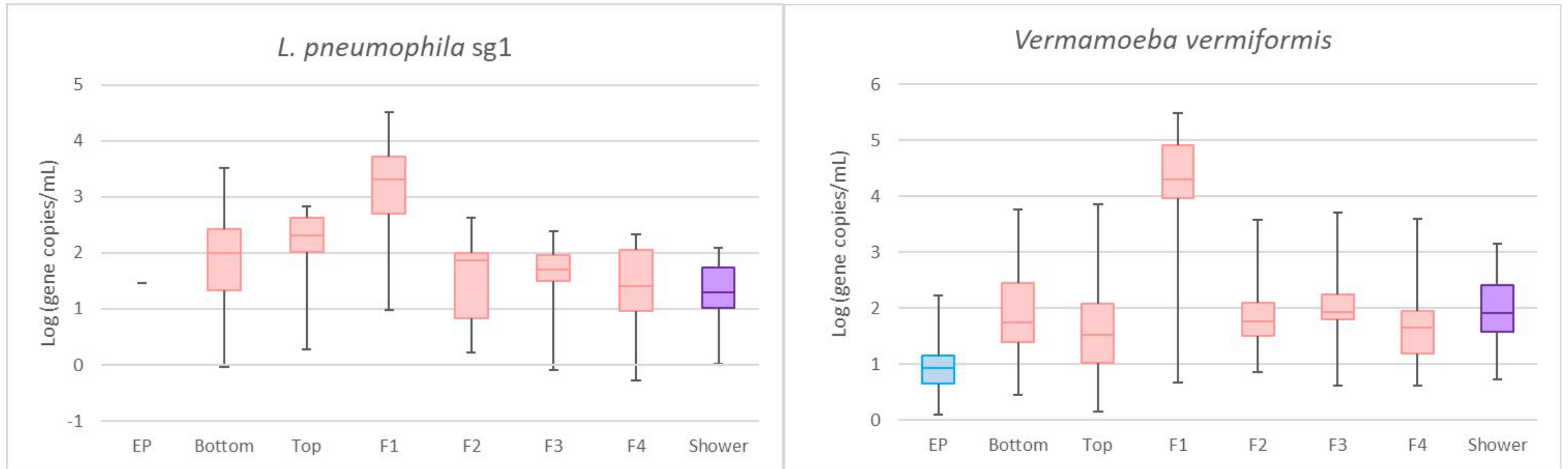
(94%, *M. intracellulare* – 67%)

## *P. aeruginosa*



(22%, 48 positive out of 217)

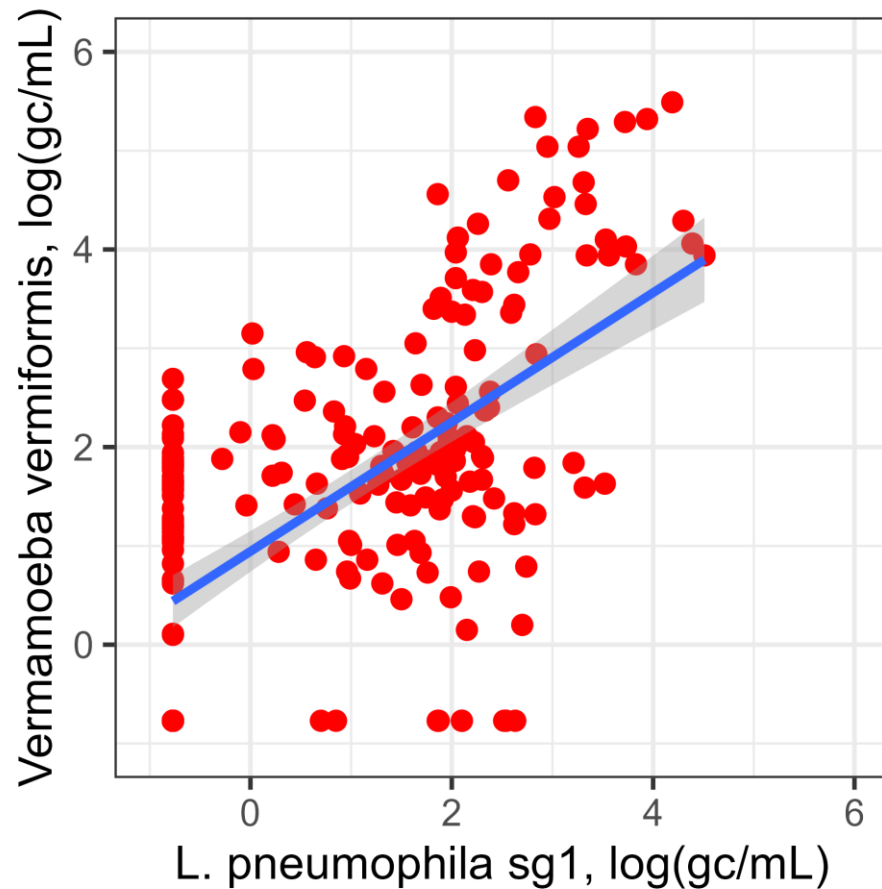
# ***F1: Potential growth of *L. pneumophila* within phagocytic amoeba***



# Correlation (Spearman's rho)

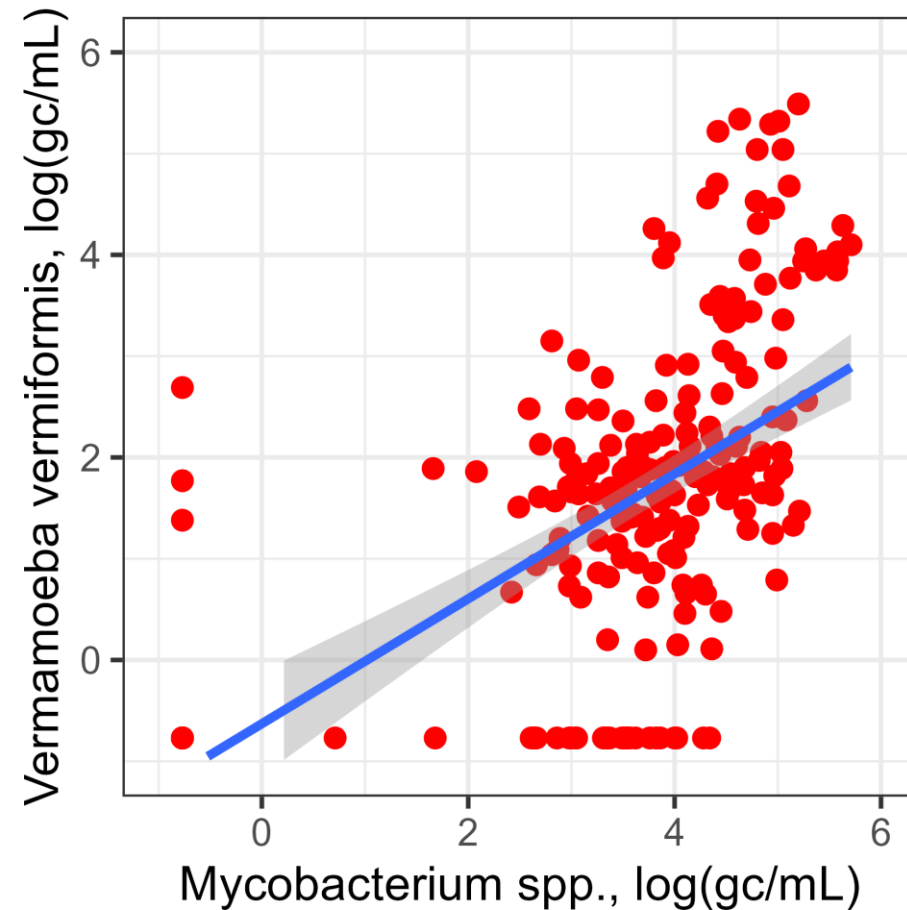
## Amoeba vs. *L. pneumophila*

(Spearman rho=0.57,  $p<0.05$ )



## Amoeba vs. *Mycobacterium* spp.

(Spearman rho=0.57,  $p<0.05$ )



# Case study 1 - Summary

- ✓ *L. pneumophila* were the most prevalent pathogen (86%), followed by *M. intracellulare* (67%) and *P. aeruginosa* (22%).
- ✓ Faucet 1 had a relatively high concentration of *L. pneumophila* and the highest detection of *V. vermiformis*. This suggests the potential growth of *L. pneumophila* within the phagocytic amoeba.
- ✓ Mycobacterium spp. were detected in all hot- and cold-water locations with consistently high levels over the entire sampling period, indicating that mycobacteria were very persistent in the chlorinated water of this system.
- ✓ Microbial community is highly diverse with evidence of spatial and temporal structuring influenced by environmental conditions.
- ✓ qPCR and sequencing methods proved to be an effective way to quantify the change in pathogen numbers and to monitor the microbial population in the HPS as different treatment methods are applied.



# Acknowledgements



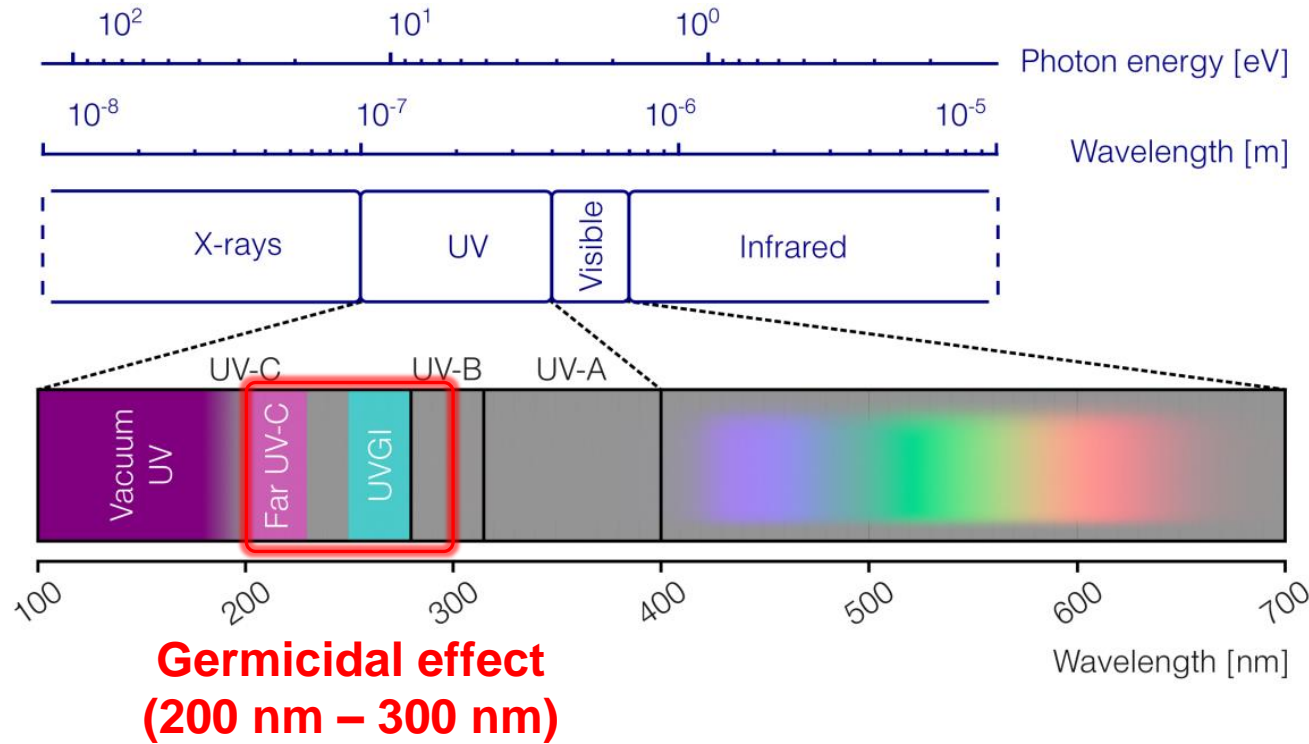
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*Shaping the Future of Science*



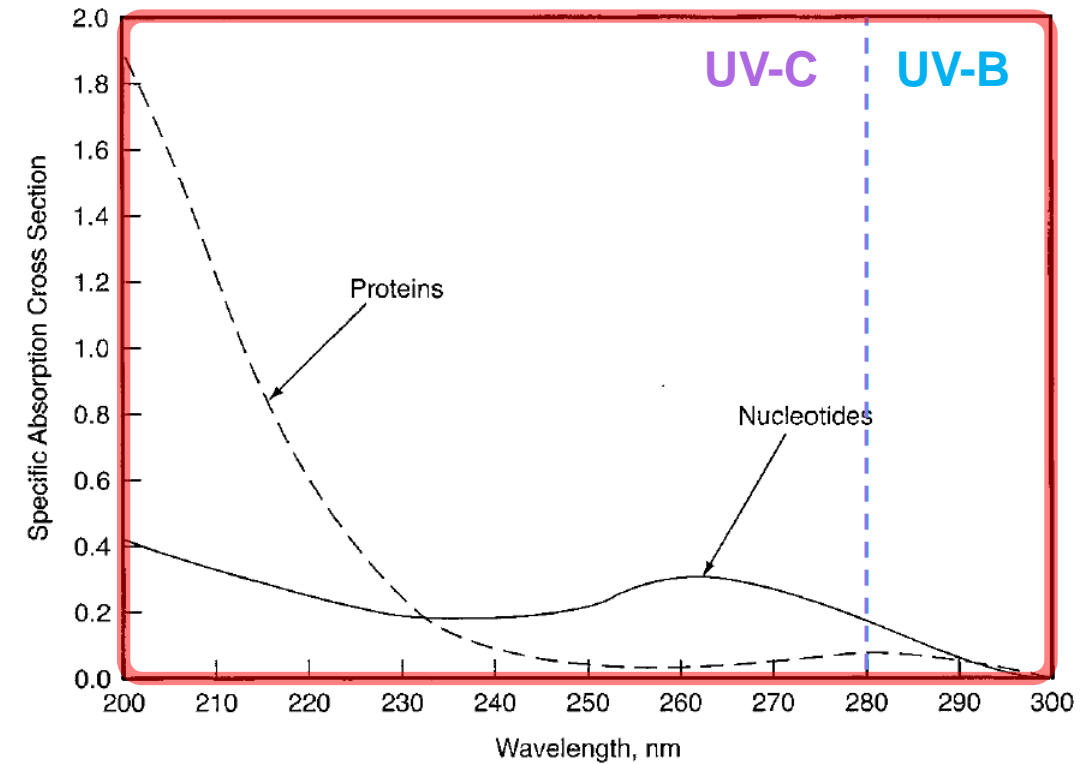
1. Microbial water quality – built environment

## **2. Microbial disinfection by UV-LED**

# Germicidal Ultraviolet



Source: IUVA White Paper

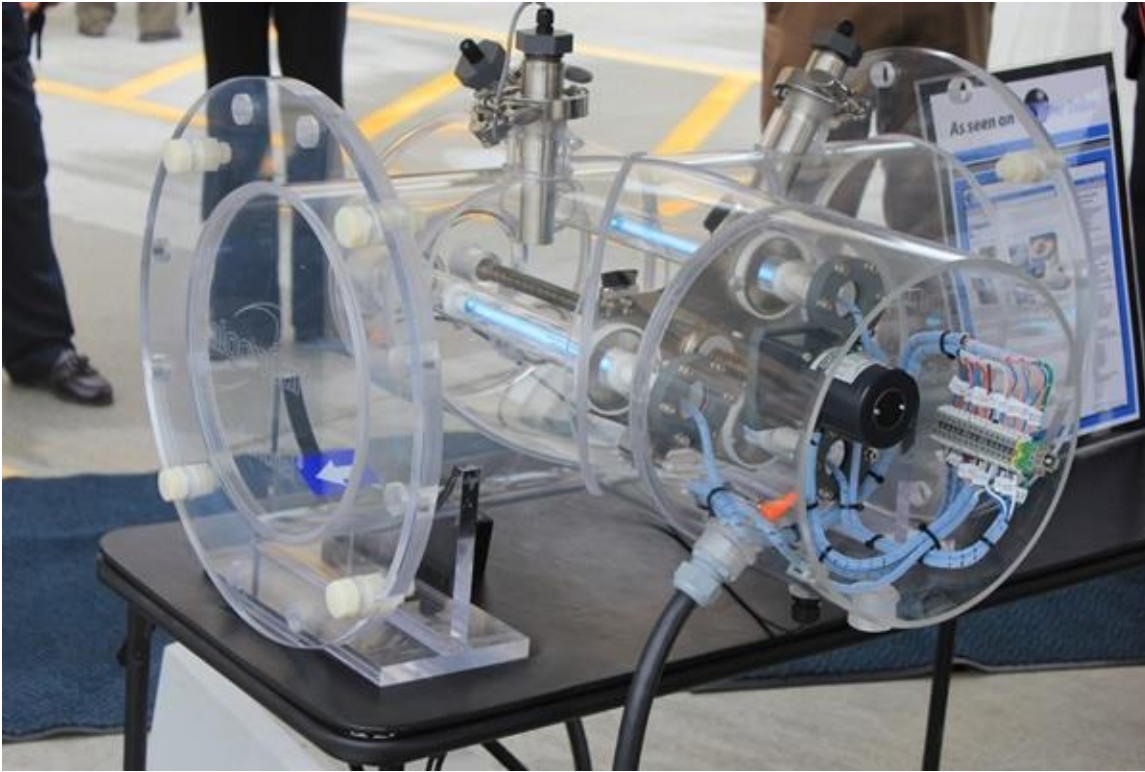


Source: Ultraviolet Disinfection Handbook

- UV can be subdivided into several categories, including UV-A, UV-B, UV-C, and vacuum UV.
- The 200 – 300 nm of UV is absorbed by the DNA, RNA, or protein of microorganisms, which breaks bonds in an organism's genome and structure.



# UV Water Disinfection

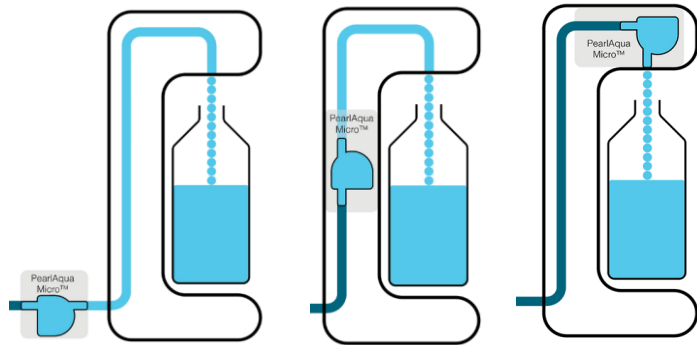


Source: <https://www.cincinnati-oh.gov/water/news/gcww-launches-uv-treatment-facility/>

- Effective against bacteria, viruses, and protozoa (*Cryptosporidium* and *Giardia*).
- No known chemical byproducts (e.g., carcinogenic DBPs), No residual chemical disinfectant.
- Emerging UV technology: Mercury-free UV-Light Emitting Diodes (UV-LEDs).



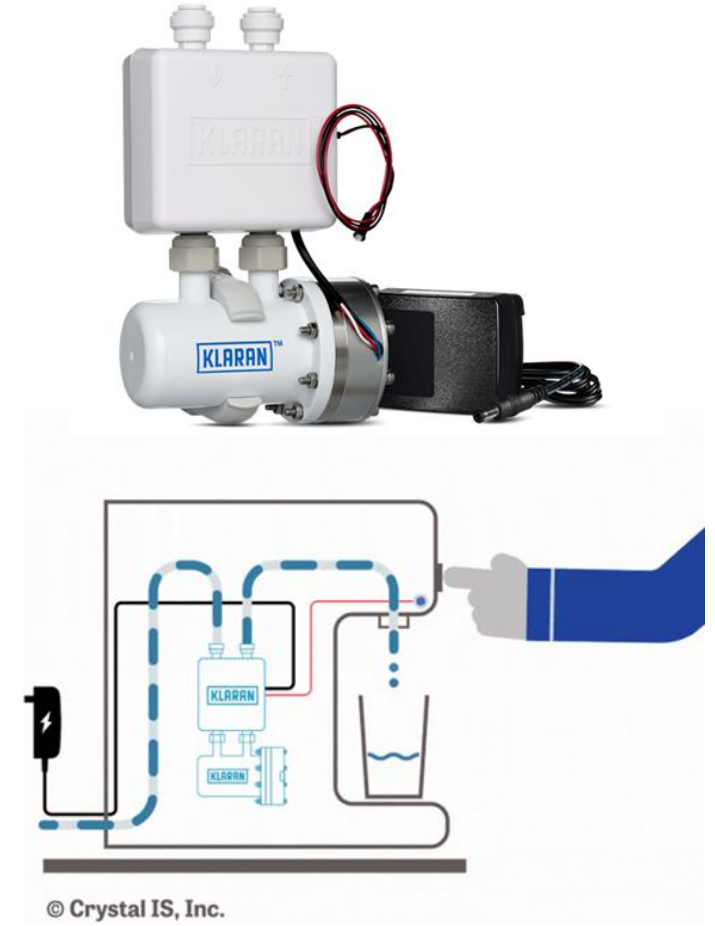
# Design Flexibility and Point-of-Use (POU)



Source: <https://www.aquisense.com/>



Source: <https://acuvatech.com/>

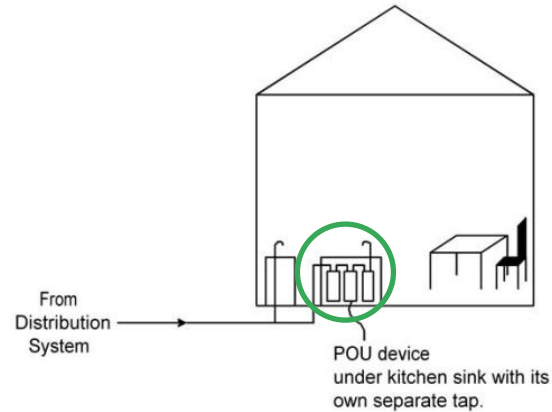


Source: <https://www.klaran.com/>

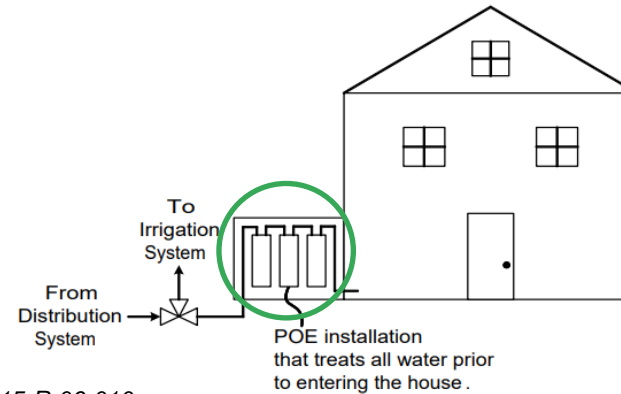
- UV-LED POU devices are ideal for small water disinfection systems.

# Decentralized Water Disinfection System

## Point-of-Use

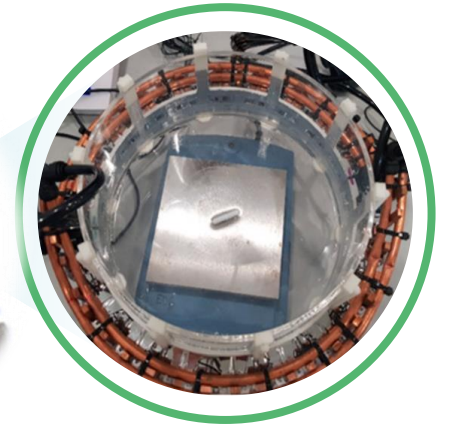
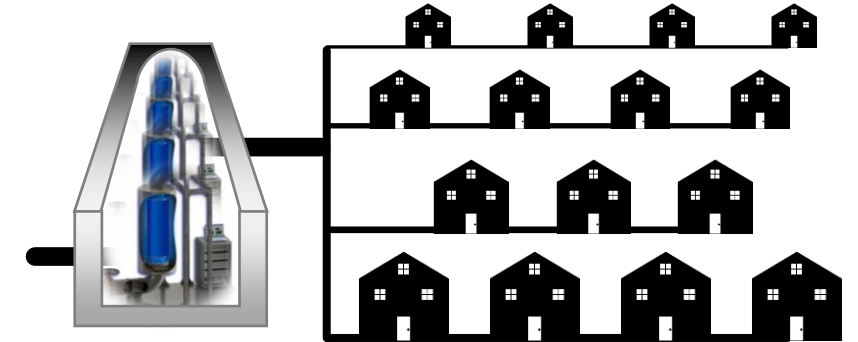


## Point-of-Entry



Source: EPA 815-R-06-010

## (small) Public Water Systems



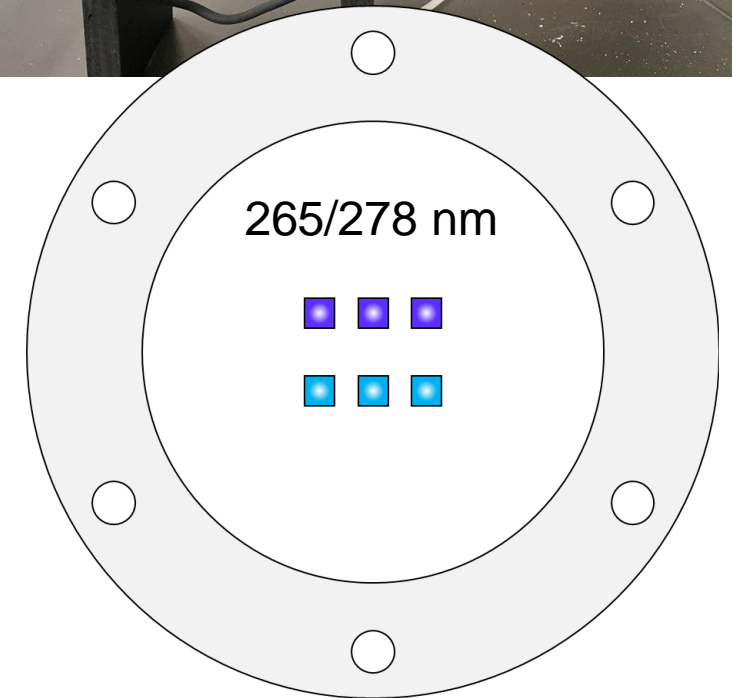
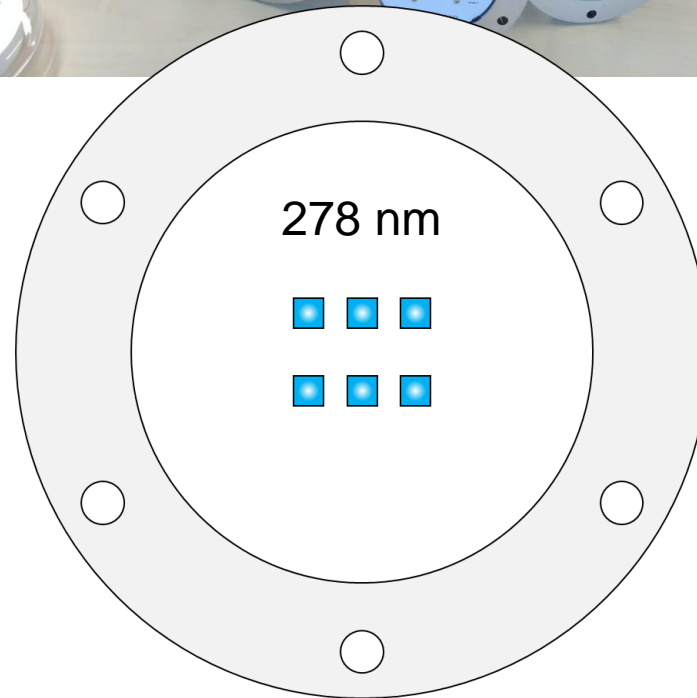
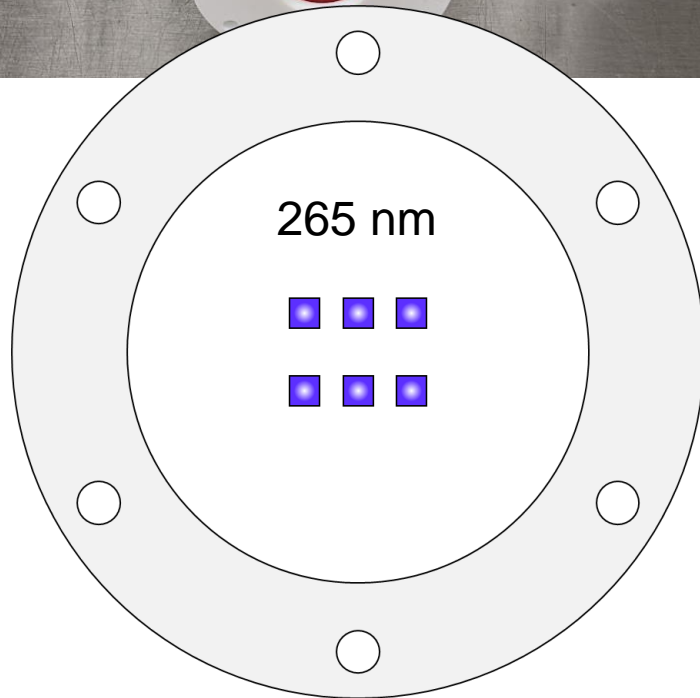
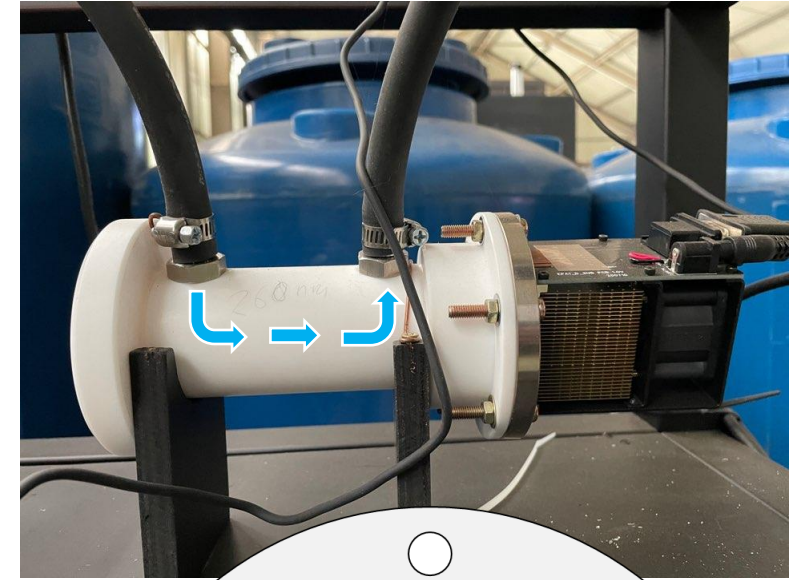
Source: Jarvis et al., Water, 2019

- The UV-LED decentralized water disinfection system can improve public health in rural communities.

# Case study 2: Research Motivation

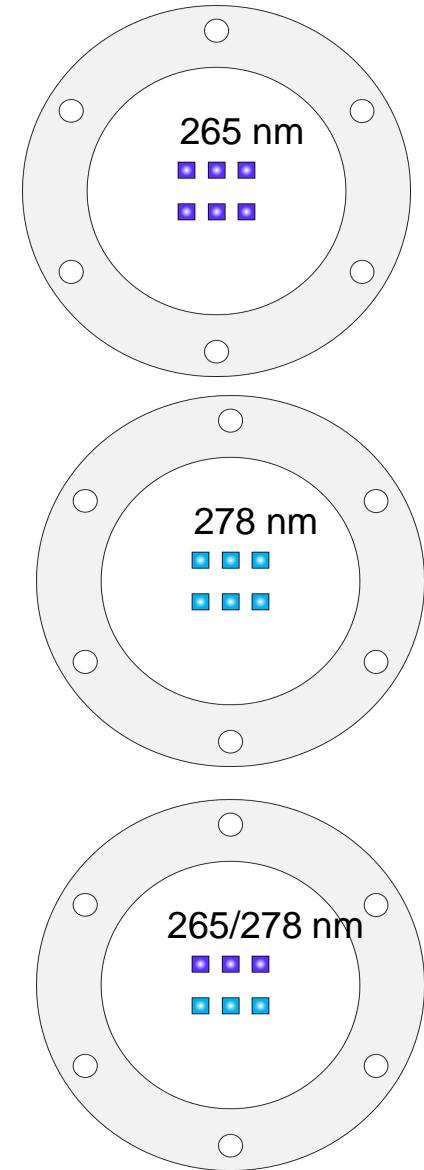
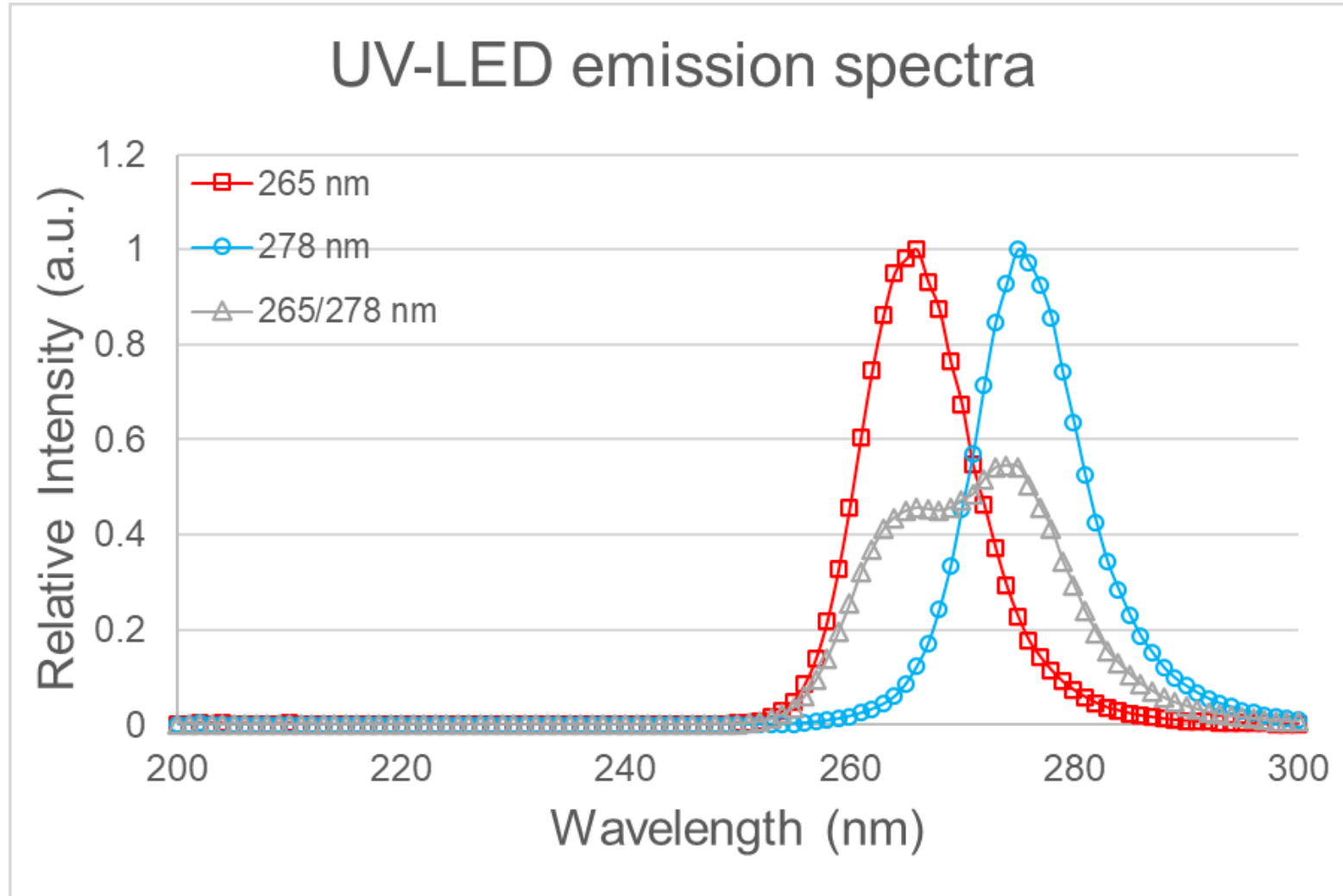
- Evaluate the performance of the UV-LED POU device for the inactivation of microbial indicators of waterborne pathogens under various operating conditions.
- Investigate potential synergistic effects of multi-wavelength on the inactivation of microbial indicators.
- Assess the specific energy consumption based on the inactivation performance of the UV-LED POU device.

# UV Reactor and UV-LED Plate



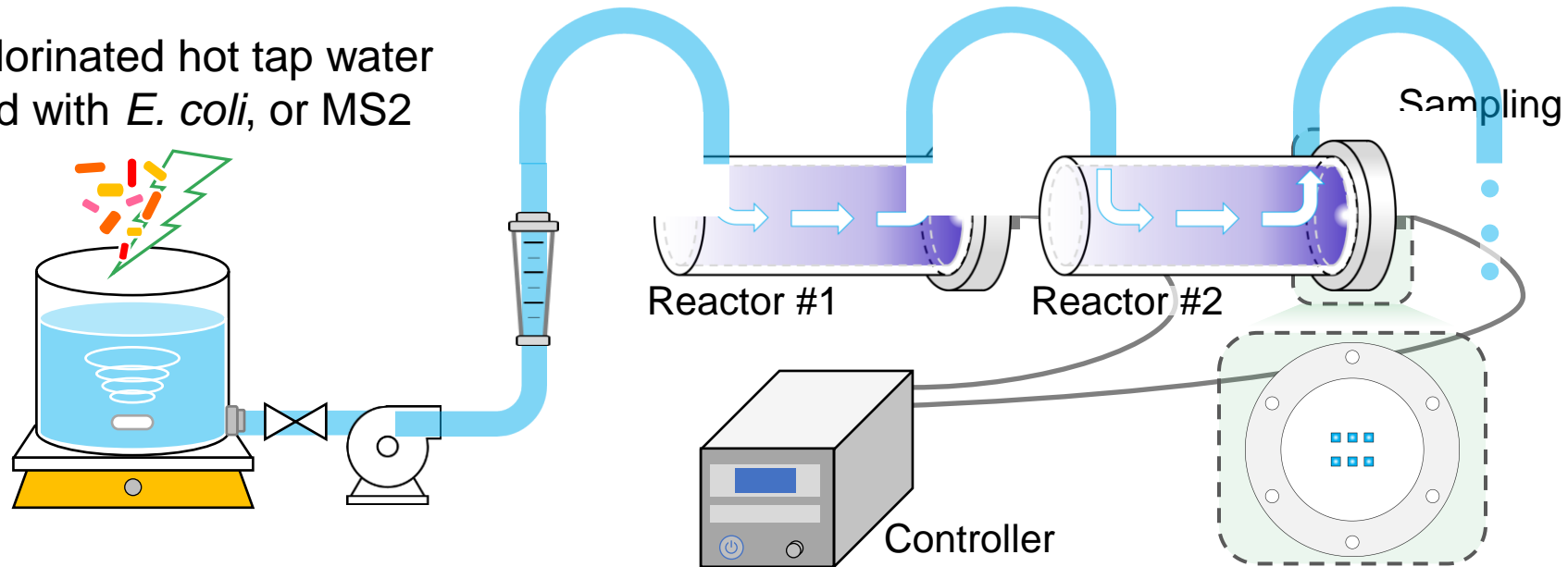


# Emission Spectra of UV-LEDs



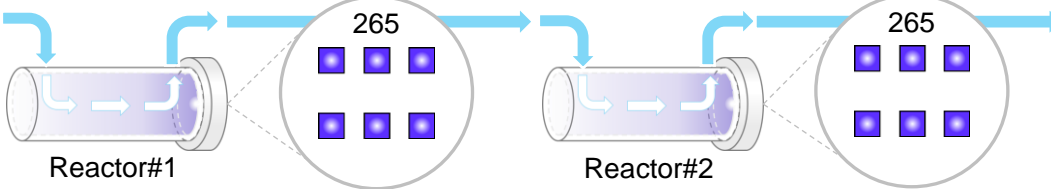
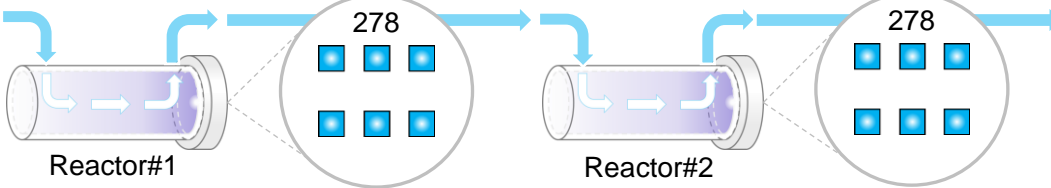
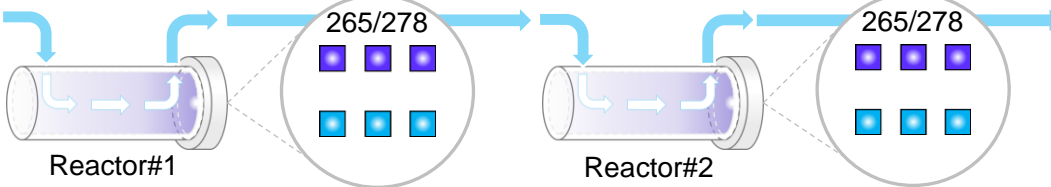
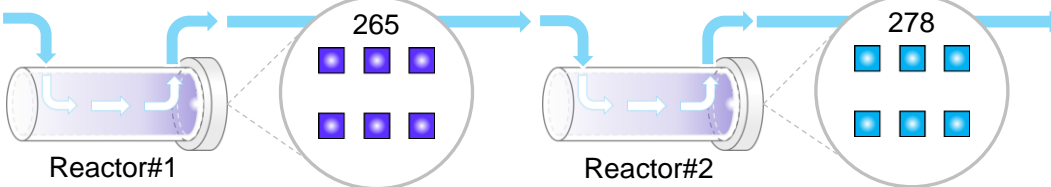
# Experimental Setup

1. Dechlorinated hot tap water containing HPC
2. Dechlorinated hot tap water spiked with *E. coli*, or MS2



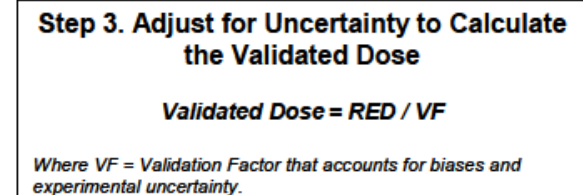
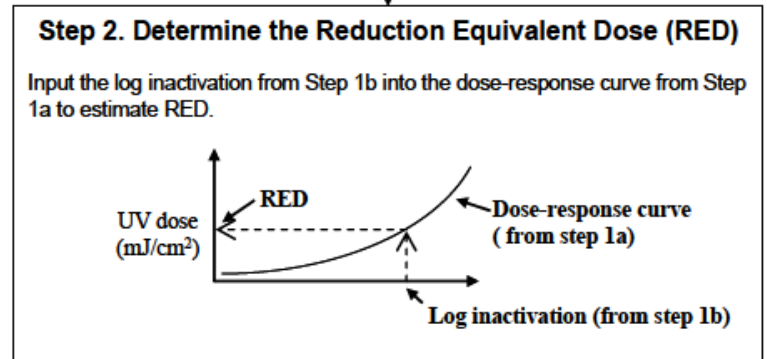
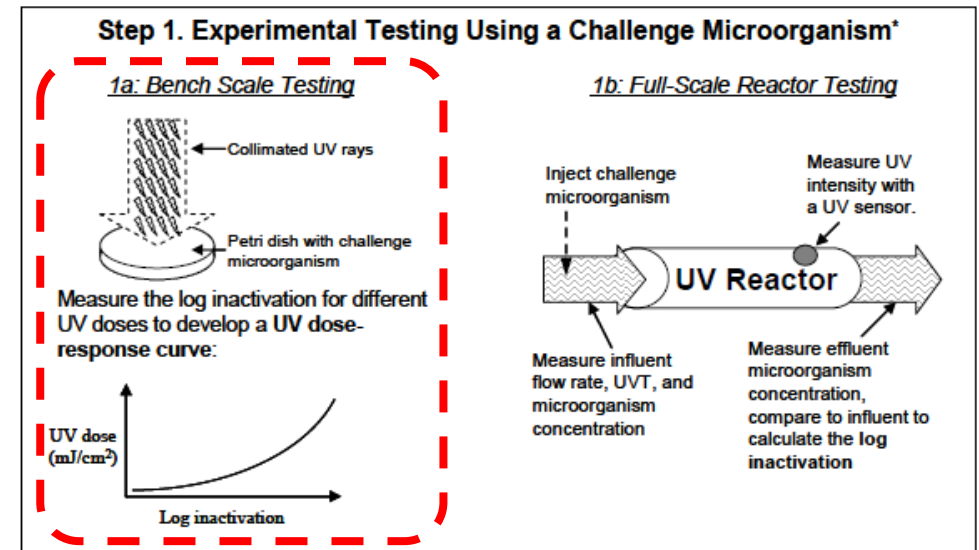
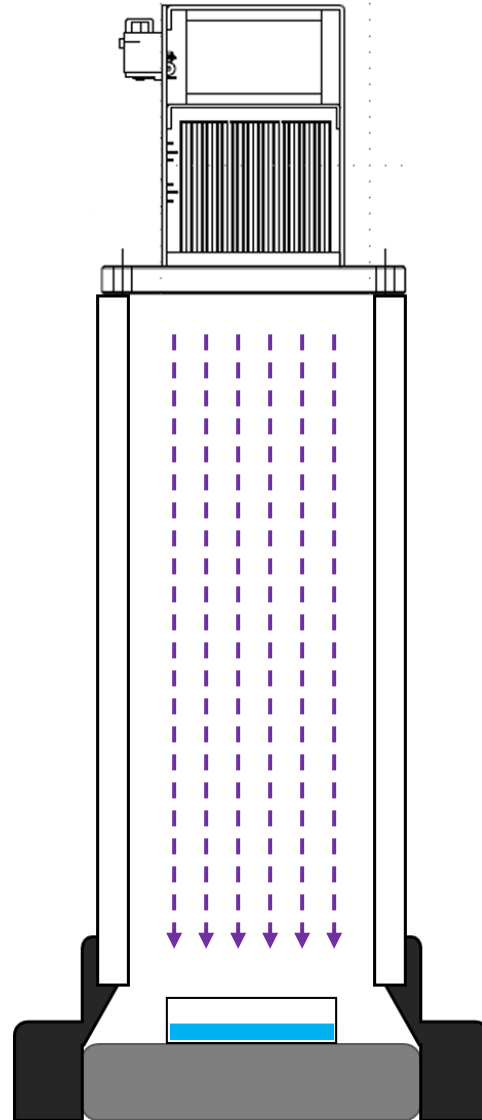
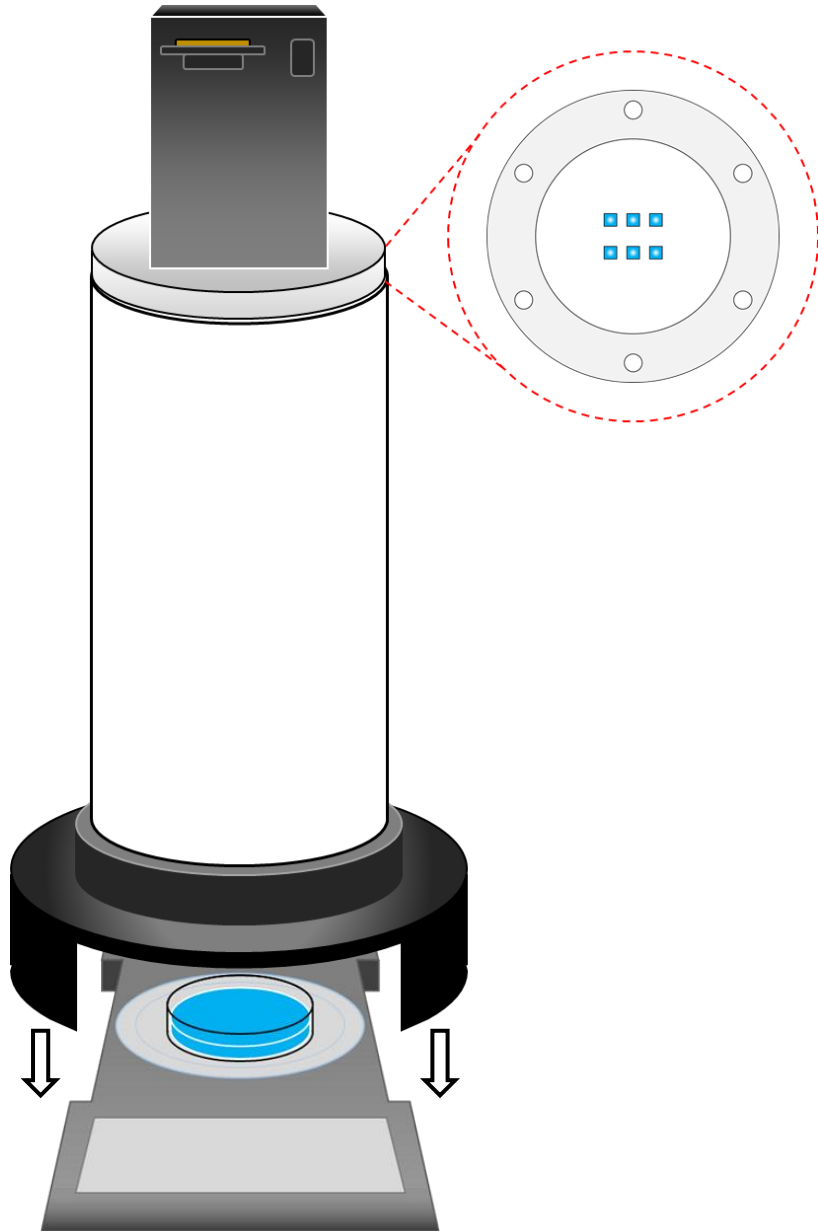
- Feed solution: (1) 20 L of dechlorinated hot tap water containing heterotrophic bacteria.  
(2) 20 L of dechlorinated hot tap water spiked with *E. coli* (ATCC 25922 , $10^6$  CFU/mL) or MS2 (ATCC 15597-B1,  $10^6$  PFU/mL).
- Flow rate: 1 L/min, 2 L/min, or 4 L/min.
- Different UV wavelengths, irradiation sequences, and UV intensities.

# UV Wavelength Combinations

No.	Reactor #1	Reactor #2	Configuration	
1	265 nm	265 nm		Single Wavelength
2	278 nm	278 nm		
3	265/278 nm	265/278 nm		Multiple Wavelength
4	265 nm	278 nm		

➤ To investigate synergistic effects between UV wavelength combinations on microorganisms inactivation.

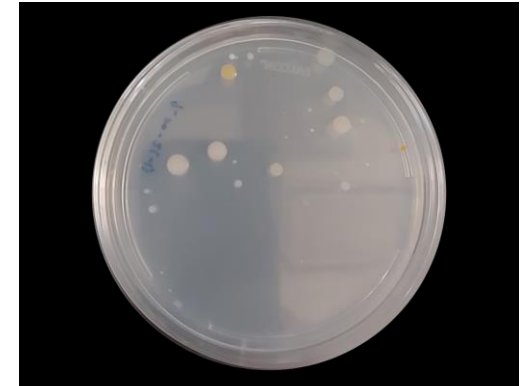
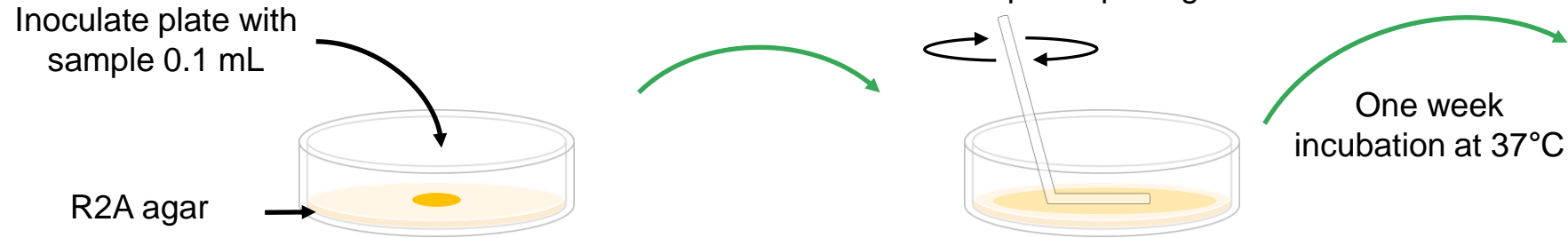
# Collimated Beam Testing



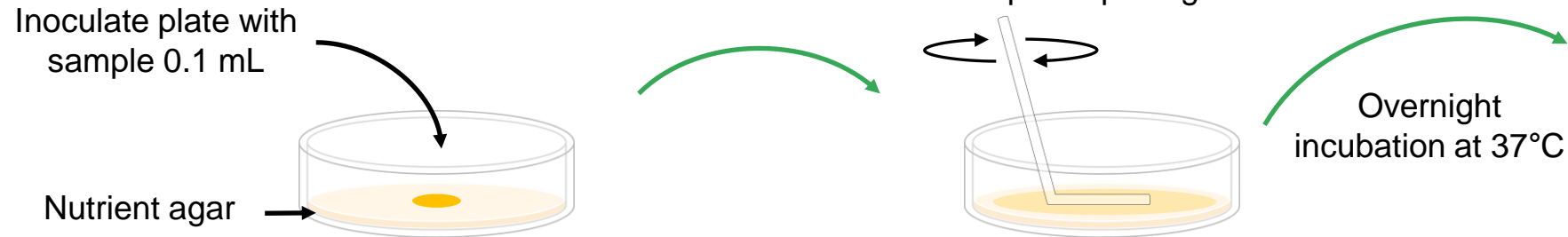


# Microbiology Methods

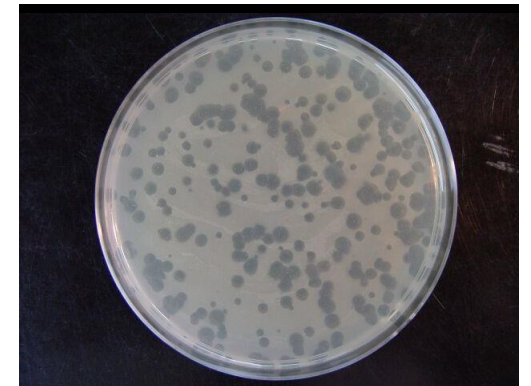
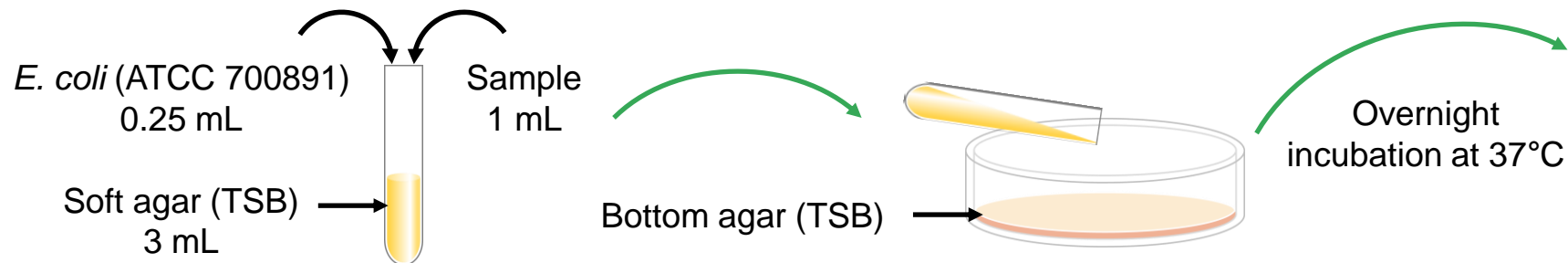
## Heterotrophic bacteria (Spread plating)



## *E. coli* (Spread plating)

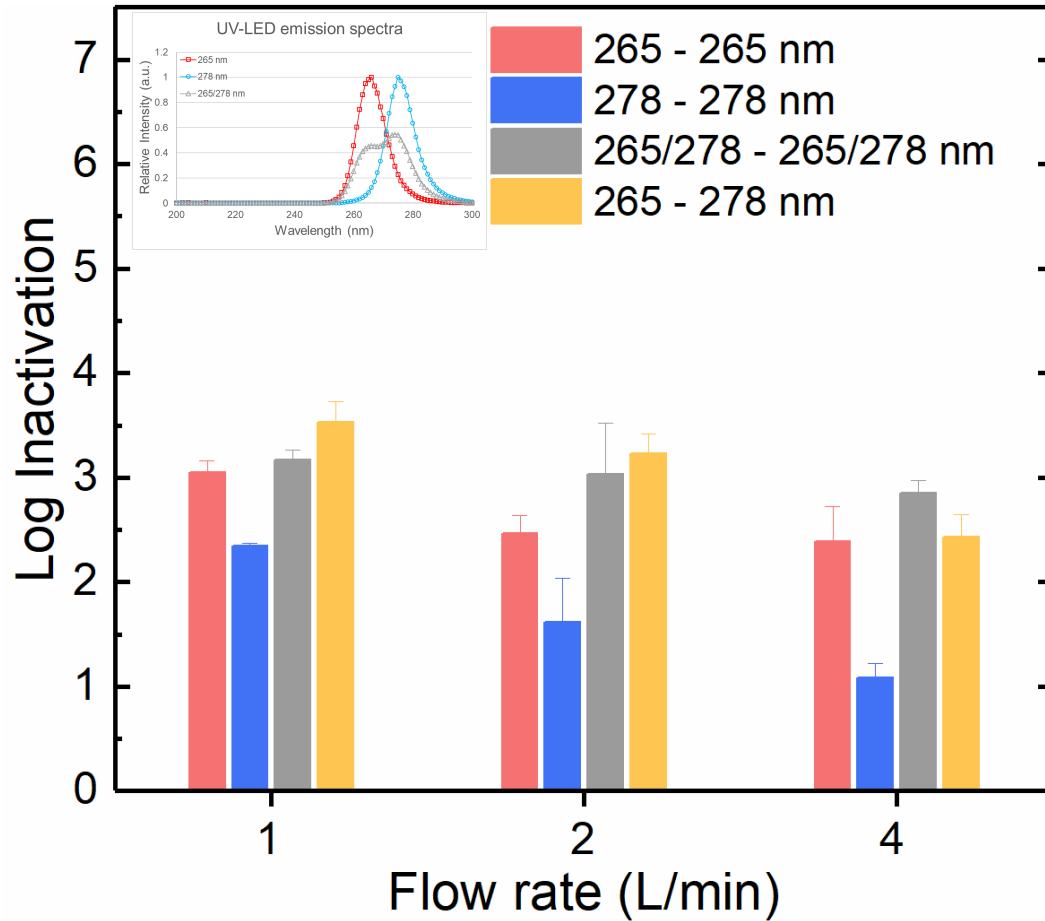


## MS2 (Double agar layer)



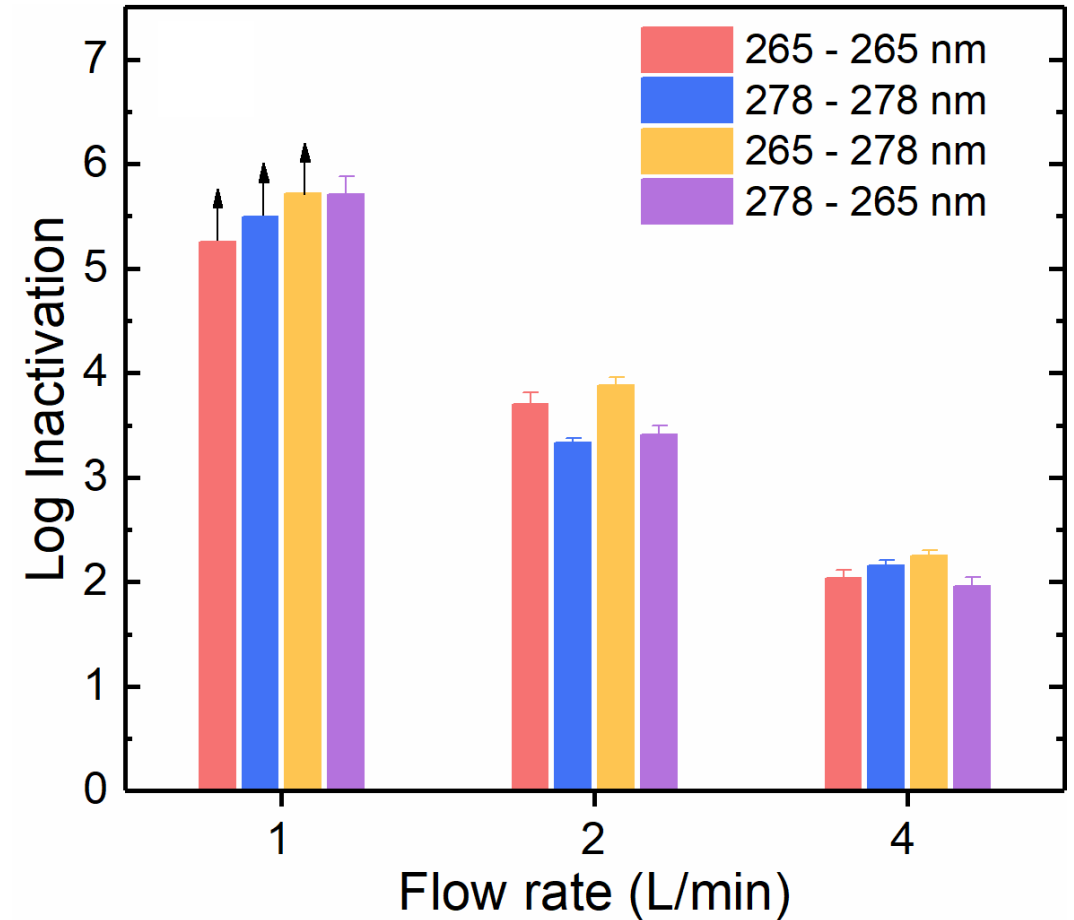
# Bacterial Inactivation

## HPC



- Possible synergistic effects of multi-wavelength due to different action spectra for diverse bacteria.

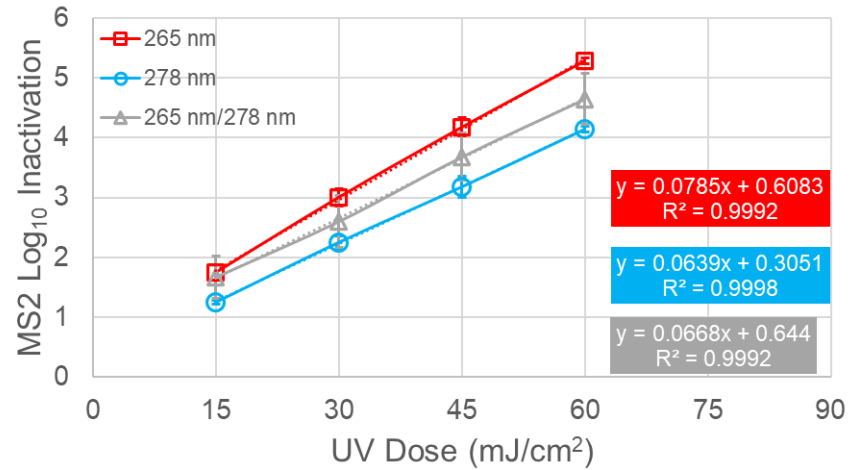
## *E. coli*



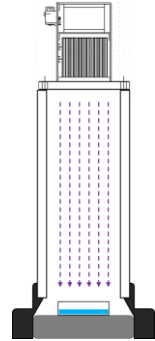
- Still greater than 2-log inactivation even at 4 L/min (0.9 sec of UV exposure time per each reactor).

# Inactivation of MS2 and RED

UV Dose - Response (MS2)

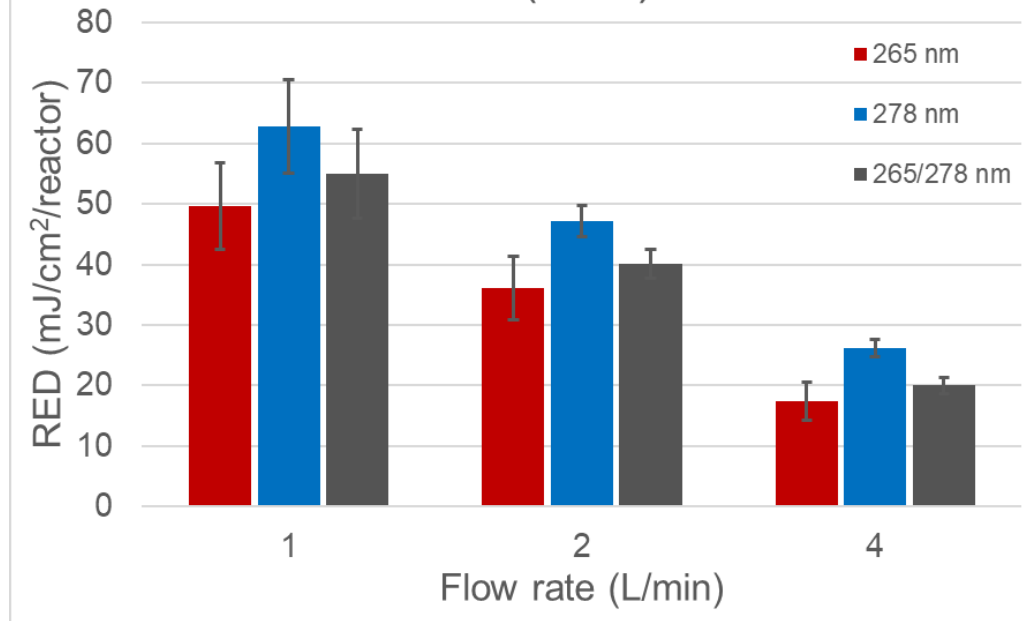


1a: Bench Scale Testing



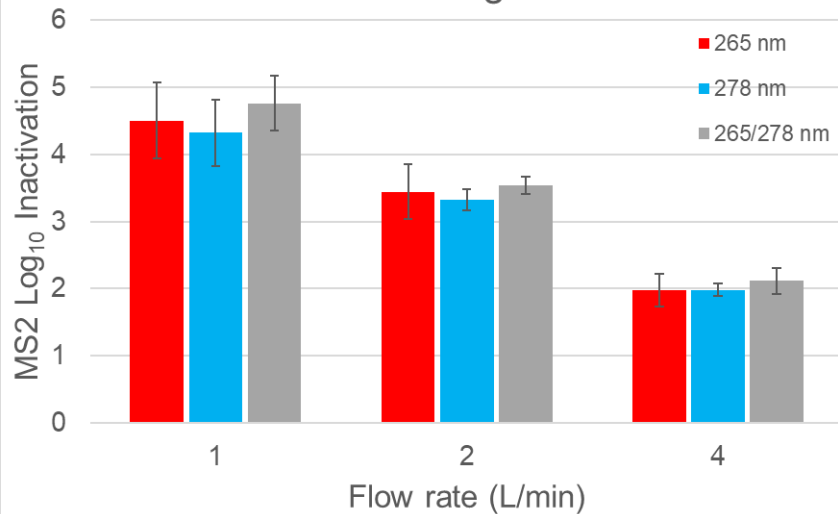
2. Determine the Reduction Equivalent Dose (RED)

RED (MS2)

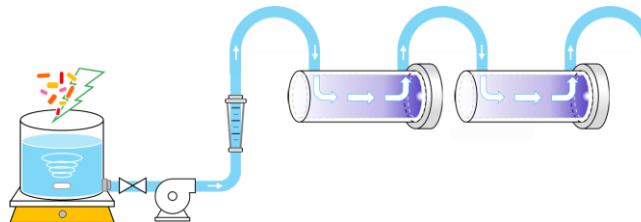


*\*All six (6) LEDs on*

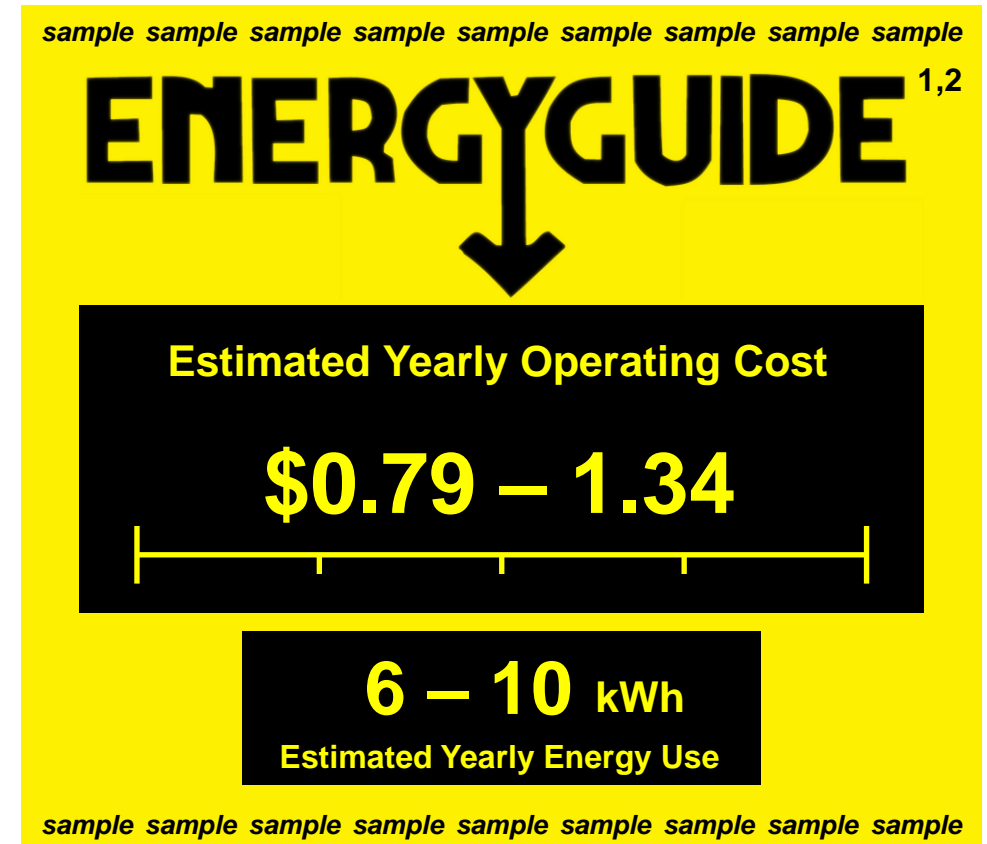
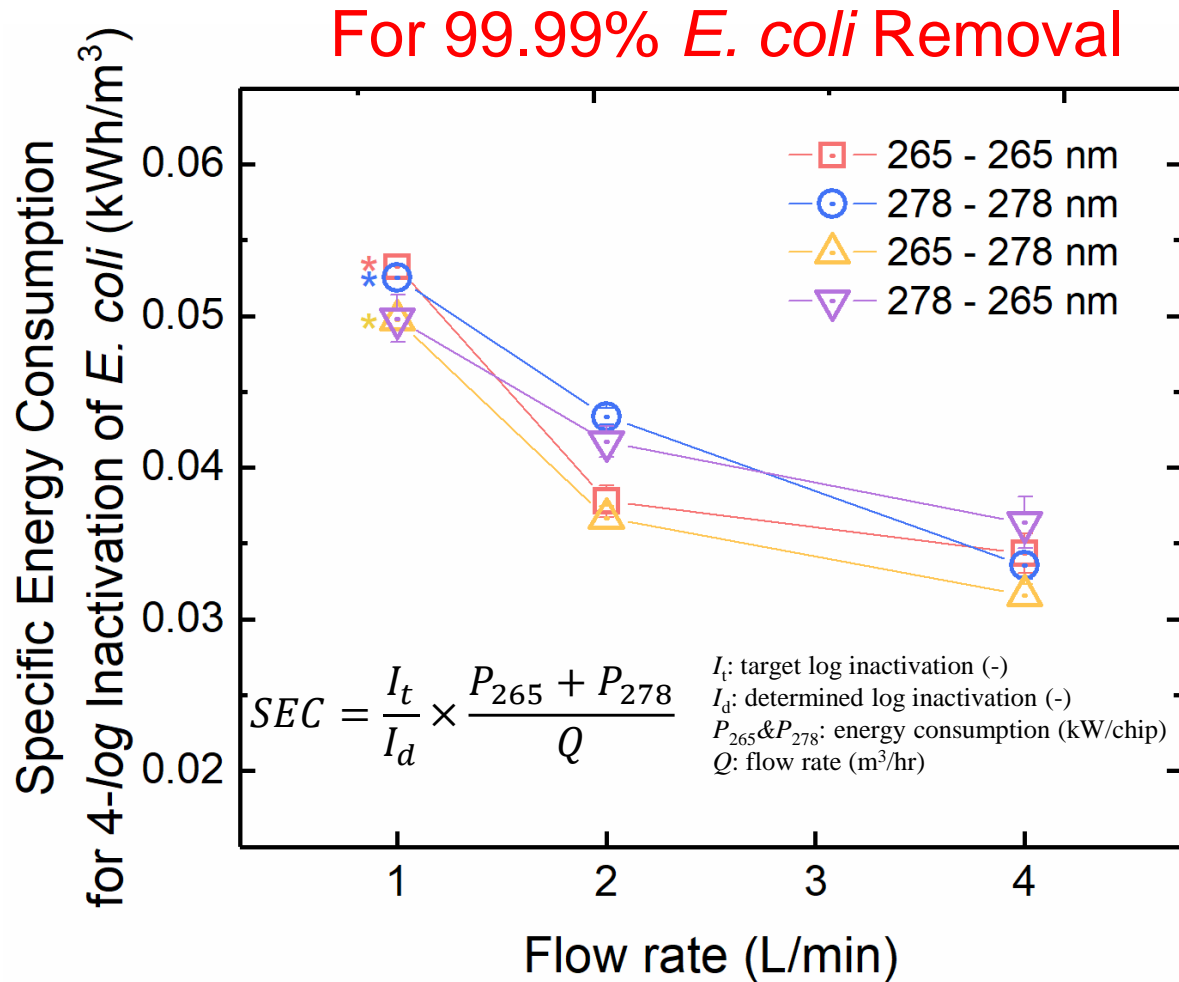
MS2 inactivation using UV-LED POU



1b: Full-Scale Reactor Testing



# Specific Energy Consumption



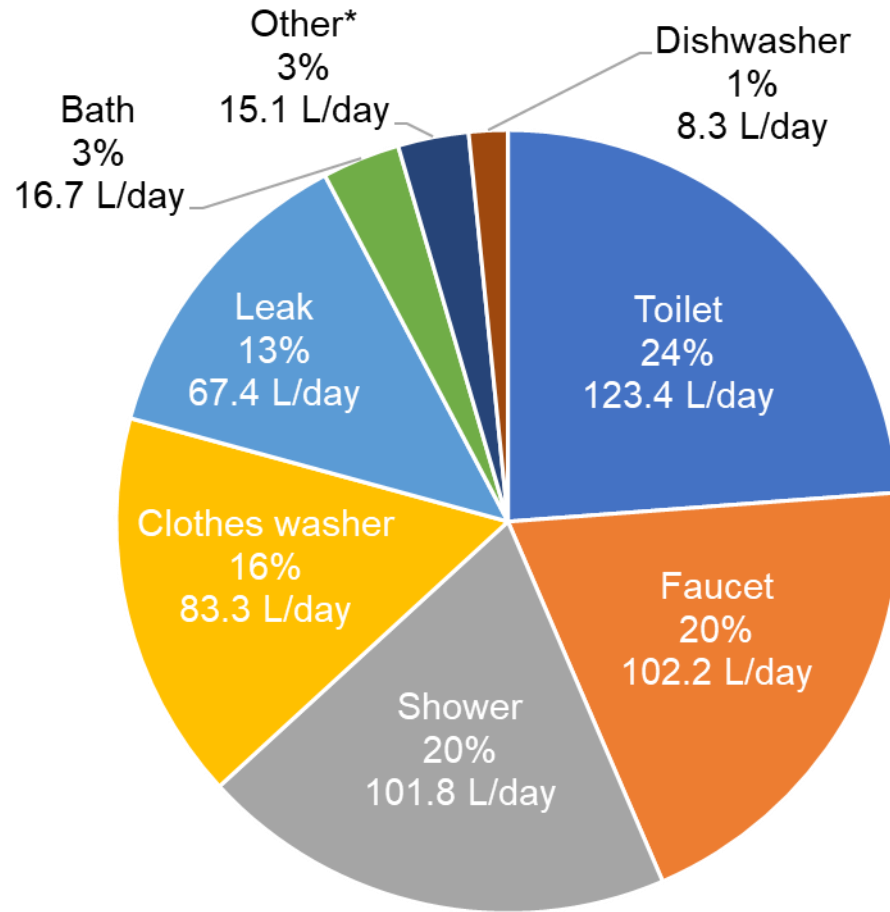
<sup>1</sup>Assuming 190 m<sup>3</sup> of UV-treated product water per year (520 L/day per indoor household water usage).

<sup>2</sup>13.21 cents per kWh of average price of electricity to ultimate customers, year 2021 (Source: U.S. Energy Information Administration (EIA), Electric Power Monthly).

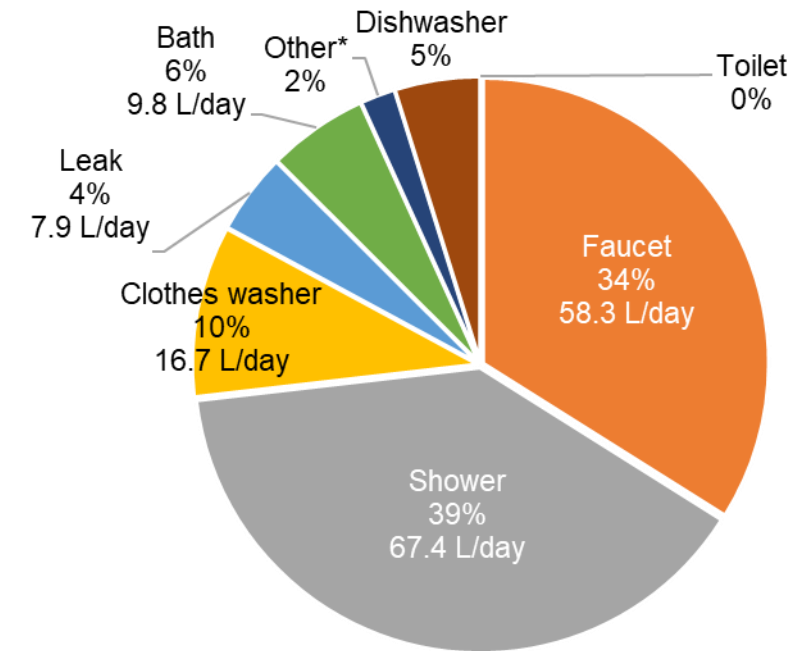
- Estimated annual operating costs: \$0.79 – 1.34 (depending on flow rates).
- Capital cost, electricity usages by other necessary equipment (controller), maintenance costs are excluded.

# U.S. Residential Water Use

Indoor household use by fixture (**about 515 L/day**)



Hot water usage (**about 172 L/day**)



\*The other category includes evaporative cooling, humidification, water softening, and other uncategorized indoor uses.

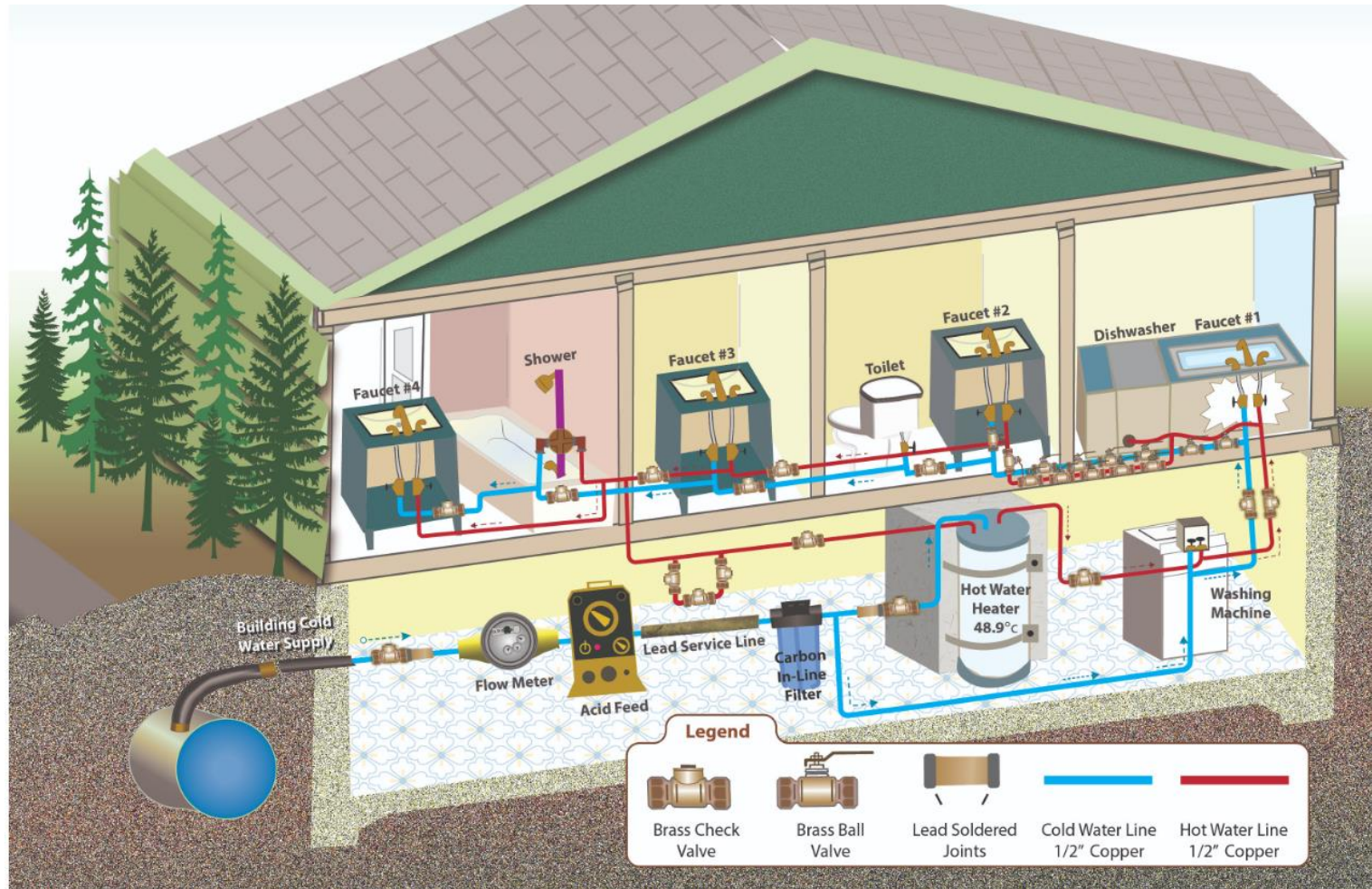
Source: Water Research Foundation, Residential Uses of Water, Version 2, 2016

# Case study 2: Summary

- This study demonstrated **the water disinfection performance of a flow-through UV-LEDs POU device** using heterotrophic bacteria, *E. coli*, and MS2 at various operating conditions.
- The results showed **a potential synergistic effect** of the combined UV-LED wavelengths on heterotrophic bacteria inactivation.
- The results of specific energy consumption revealed that increasing the flow rate can also improve energy efficiency, as demonstrated with the high flow rate *E. coli* test.
- **Further study:** the device will be installed and tested in the Home Plumbing System at EPA for real-life applications.



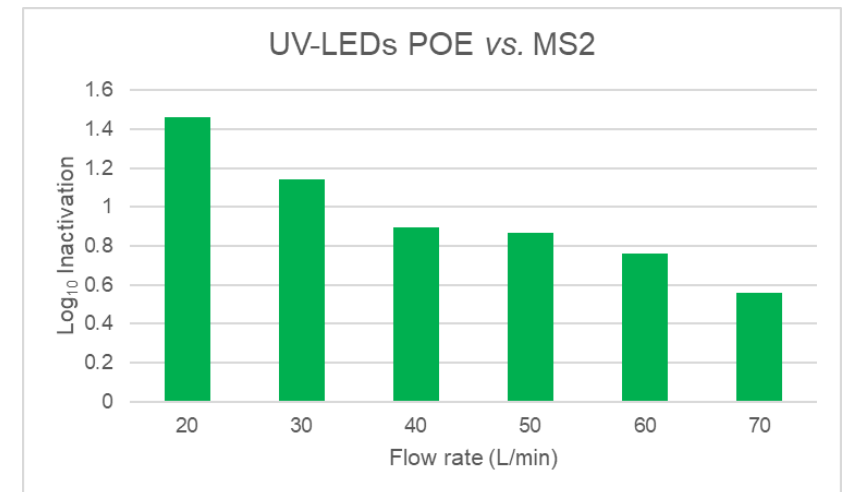
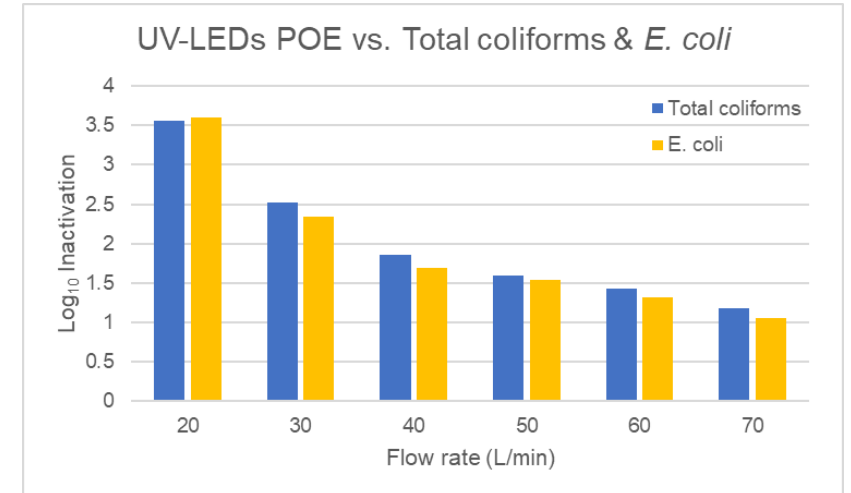
# Future Study



- Installing the device in the Home Plumbing System at EPA and testing for real-life applications.



# Wastewater Treatment



- Testing POE device for treating secondary effluent from wastewater treatment plant (US EPA T&E facility).
- $UVT_{254}$ : 64–69%



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# Q&A



THANK YOU



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