A grayscale electron micrograph of Legionella bacteria. The bacteria are rod-shaped with a distinct outer membrane and internal structures. One large, elongated bacterium is on the left, and another is partially visible on the right. The background is a fine, granular texture.

# Legionella: Detection, Surveillance & Prevention

Julie B. Trivedi, MD

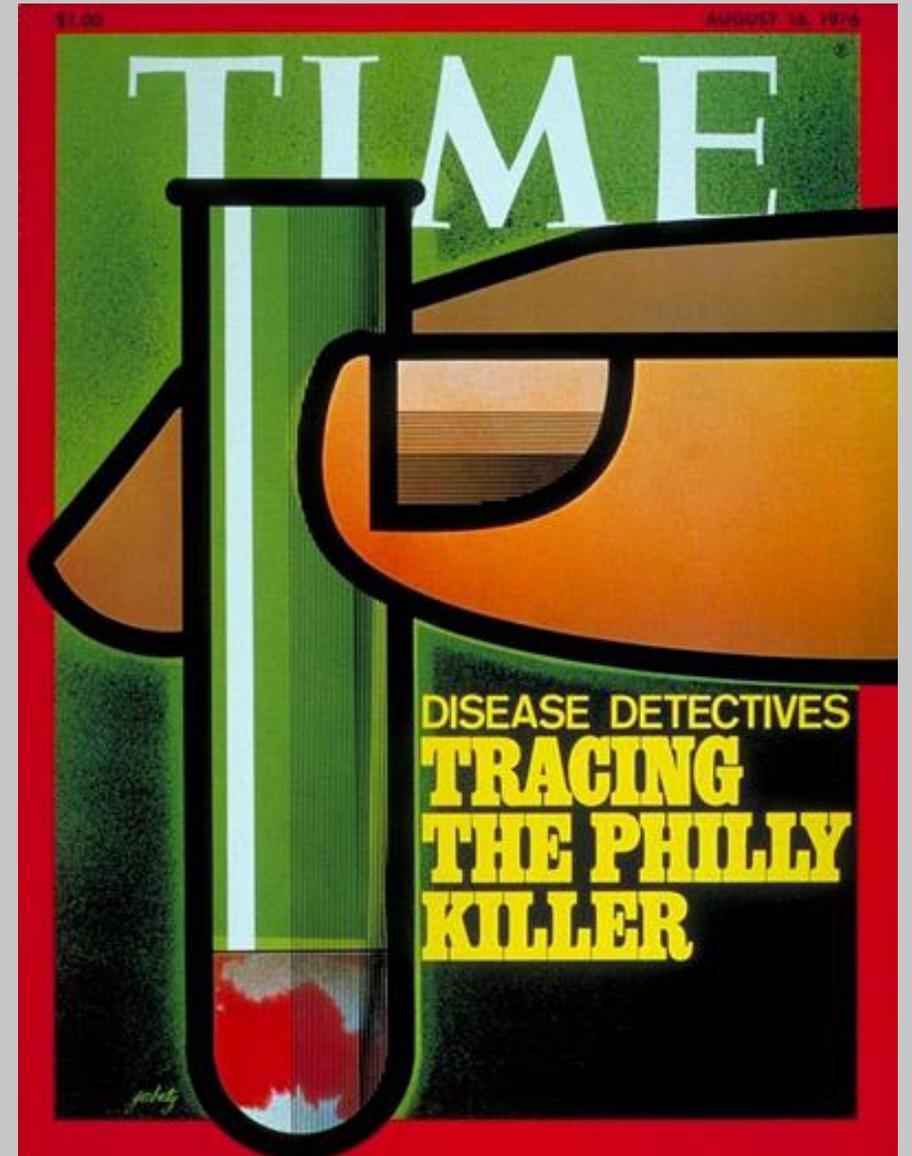
Assistant Professor of Medicine

Medical Director of Infection Prevention, University hospitals

UT Southwestern Medical Center, Dallas TX

# Outline

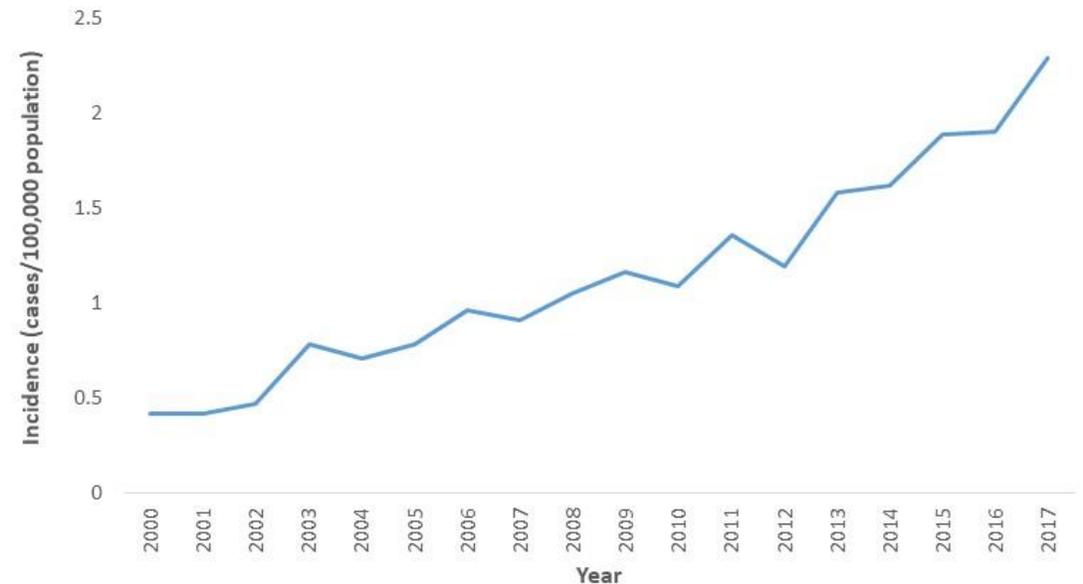
- Epidemiology
- Transmission & Clinical syndromes
- Case definitions
- Diagnostic testing
- Prevention



# Background

- Discovered in 1976 at an American Legion Convention in Philadelphia
- Rising incidence in Europe, North America
- Nearly 7500 cases of Legionnaire's disease in 2017 in US (6100 in 2016)

## Legionnaires' disease is on the rise in the United States



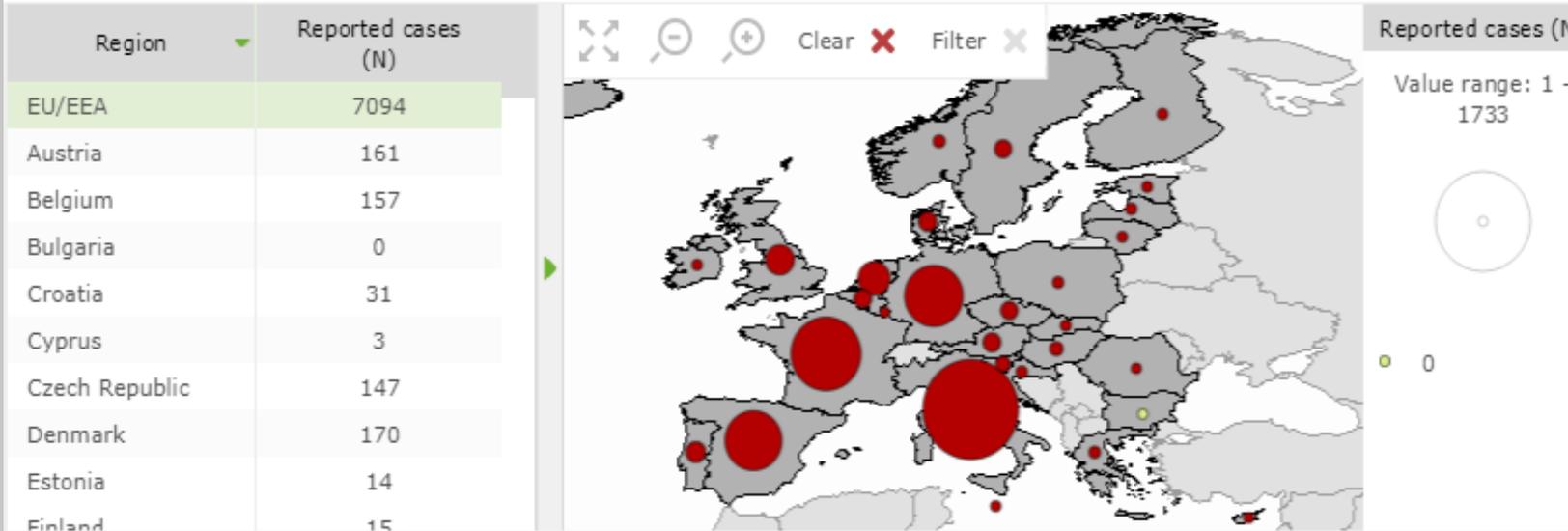
**Rate of reported cases increased 5.5 times (2000–2017)**

Source: National Notifiable Diseases Surveillance System

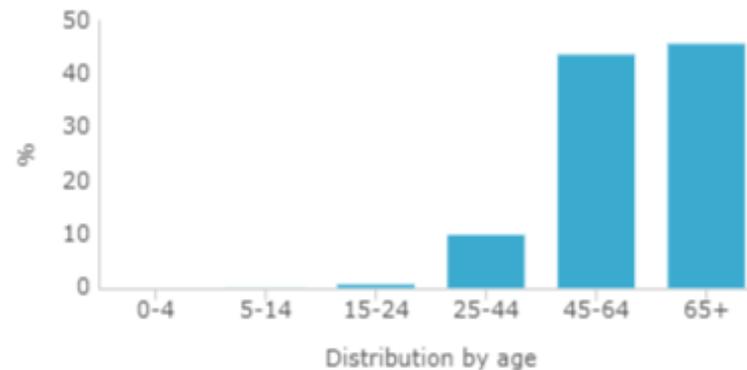
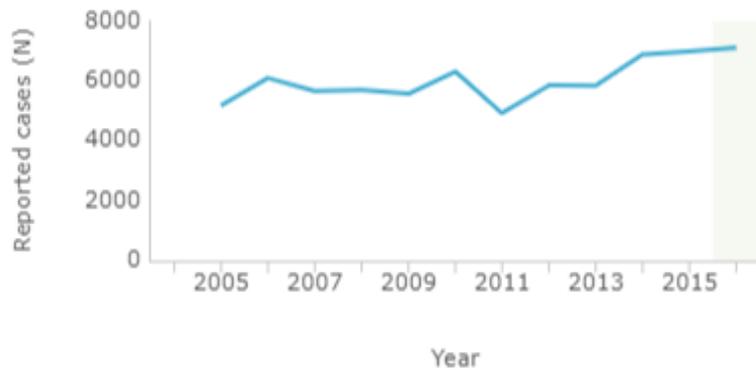
Centers for Disease Control and Prevention (CDC)

# Surveillance Atlas of Infectious Diseases

Legionnaires' disease ▾ | All cases ▾ | Reported cases ▾ | 2016 ▾



Legionella surveillance in Europe



# Background

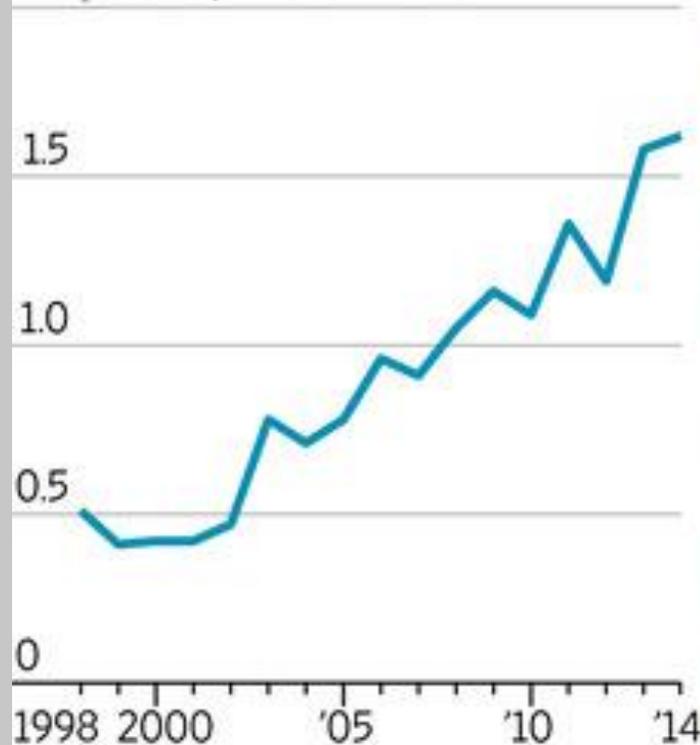
- 2000-2014, US, outbreaks:
  - 19% associated with LTCF
  - 15% with hospitals
- >20% of cases: travel related
- Risk factors in host
  - Age >50
  - Smokers
  - Chronic lung disease
  - Immunosuppression
- Exposures
  - Hotels
  - Cruise ship

# Disease Profile

In 27 outbreak investigations conducted between 2000 and 2014, the CDC found that potable water was the most frequent source of exposure and resorts and hotels were the most frequent places where outbreaks occurred.

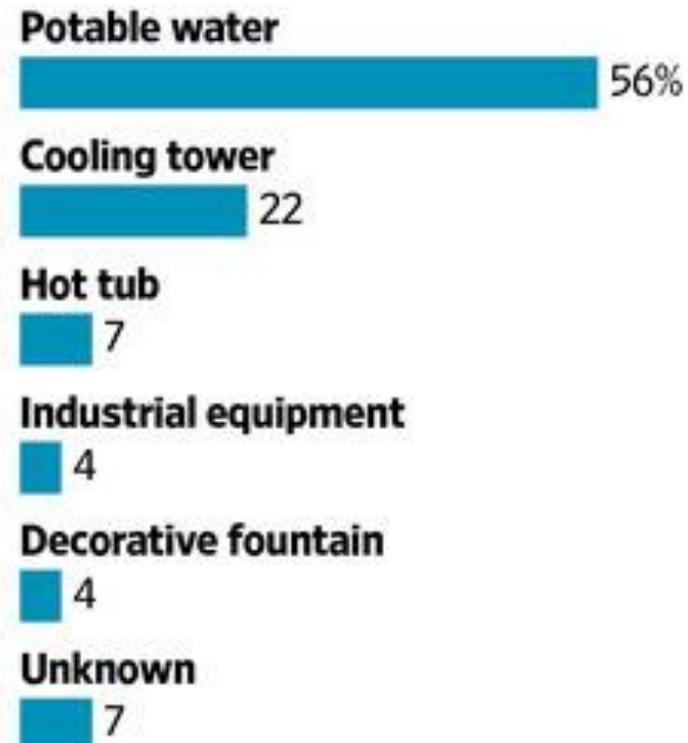
## U.S. reported cases of Legionnaires' disease

2.0 per 100,000 residents

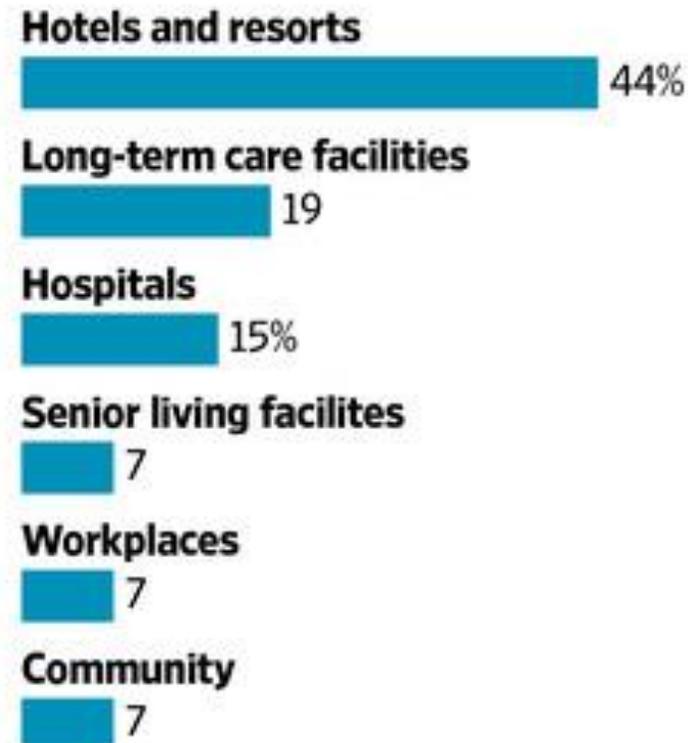


Source: Centers for Disease Control and Prevention

## Source of exposure



## Outbreak settings



THE WALL STREET JOURNAL. 6

# Pathogenic Legionella species (humans)

- **L. pneumophila (15)**
  - L. micdadei
  - L. dumoffii
  - L. bozemanii (2)
  - L. gormanii
  - L. feeleii (2)
  - L. hackeliae (2)
  - L. israelensis
  - L. jordanis
  - L. sainthelensi (2)
- **L. longbeachae (2)**
  - • L. maceachernii
  - • L. oakridgensis
  - • L. wadsworthii
  - • L. birminghamensis
  - • L. cincinnatiensis
  - • L. anisa
  - • L. tusconensis
  - • L. lansingensi

# Legionellosis: Two Clinical Syndromes

	<b>Legionnaires' disease</b>	<b>Pontiac fever</b>
Clinical features	Pneumonia: cough, fever, chest pain	Flu-like illness (fever, chills, malaise) without pneumonia
Radiographic pneumonia	Yes	No
Incubation period	2-14 days after exposure	24-48 hours after exposure
Etiologic agent	<i>Legionella</i> species	<i>Legionella</i> species
Attack rate*	< 5%	> 90%
Isolation of organism	Possible	Virtually never
Outcome	Hospitalization common Case-fatality rate: 5-40%**	Hospitalization uncommon Case-fatality rate: 0%

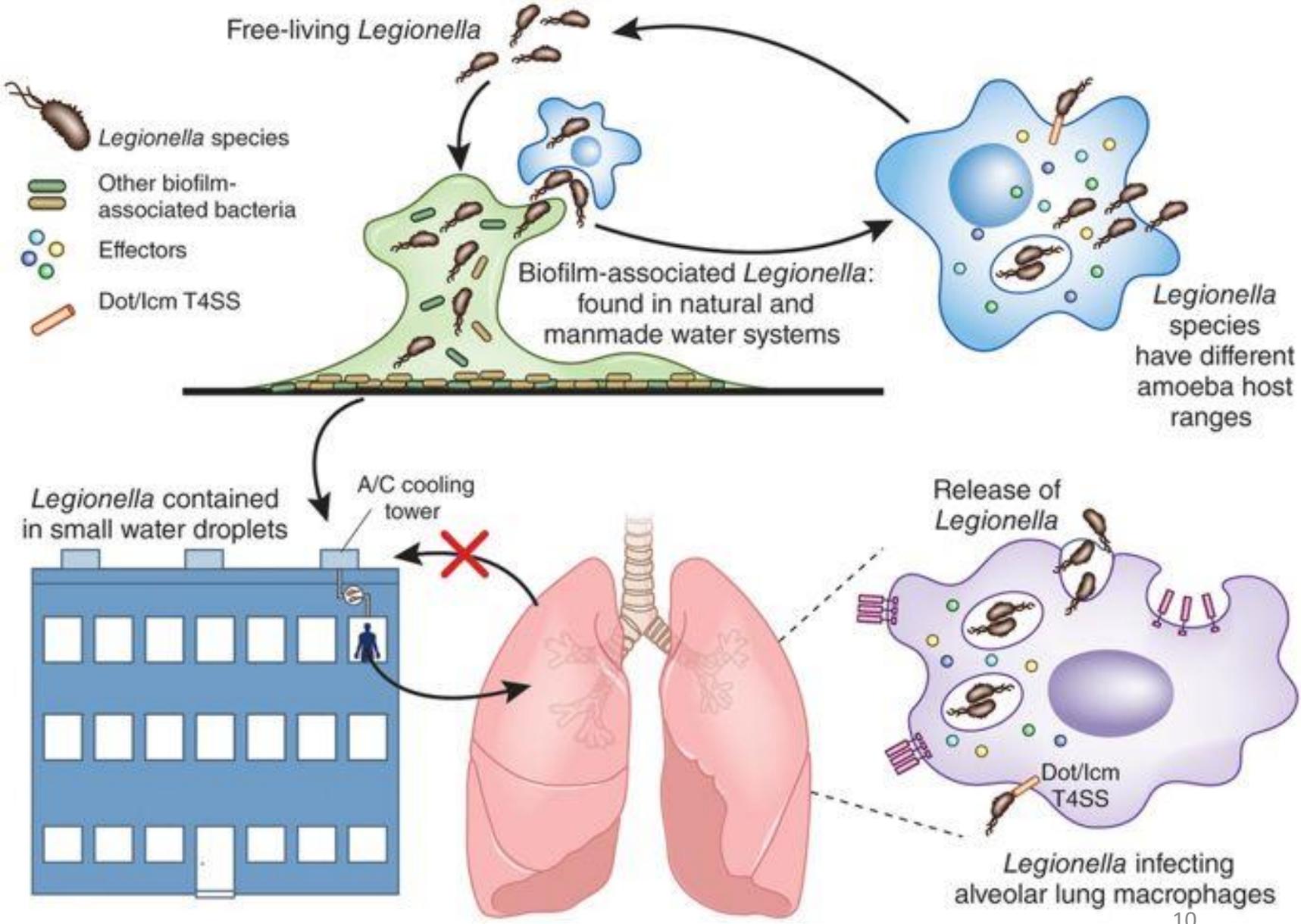
\* Percent of persons who, when exposed to the source of an outbreak, become ill.

\*\* Percent of persons who die from Legionnaires' disease or Pontiac fever.

# Microbiology

- Fastidious, aerobic, non-spore forming gram-negative bacilli
- Biphasic life cycle
  - nonmotile replicative  $\Leftrightarrow$  virulent, flagellated transmissive phase
- Facultative intracellular pathogens
- Naturally found in freshwater environments: rivers, lakes, streams, hot springs
- Known to form complex biofilm communities
- Can live within free-living amoeba

# Legionella Life cycle

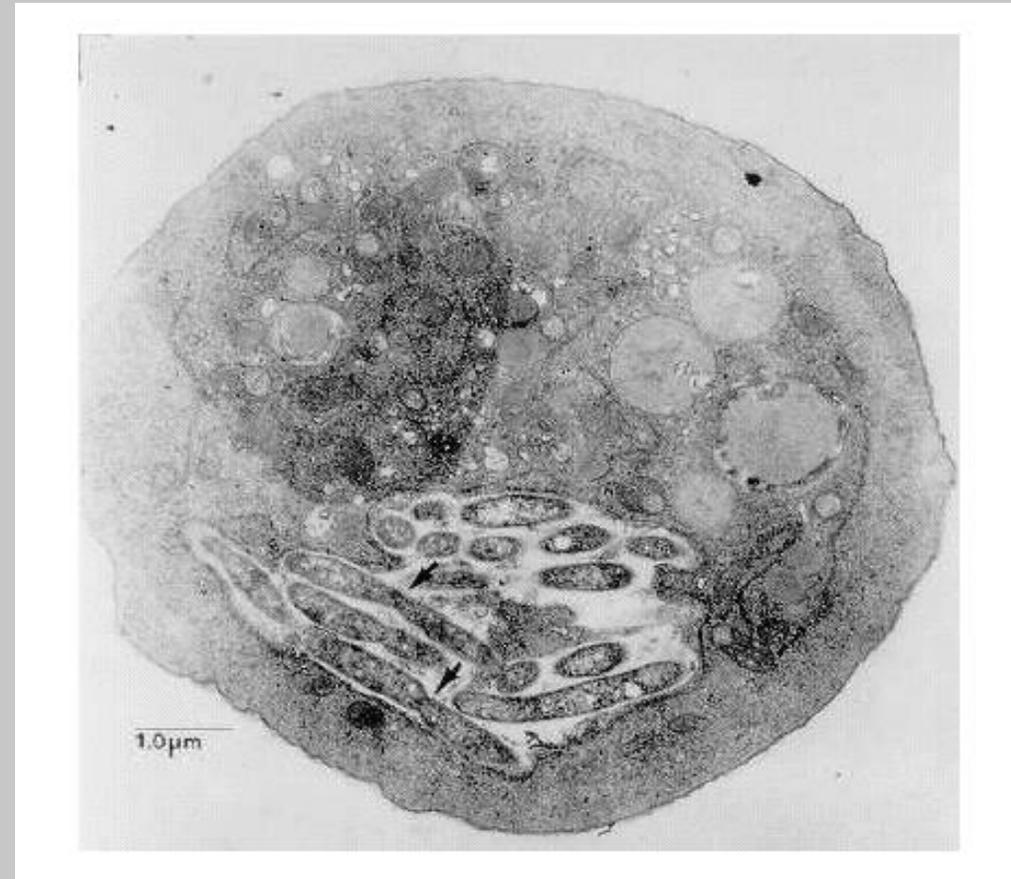


# Intracellular growth

**Lung fibroblast**

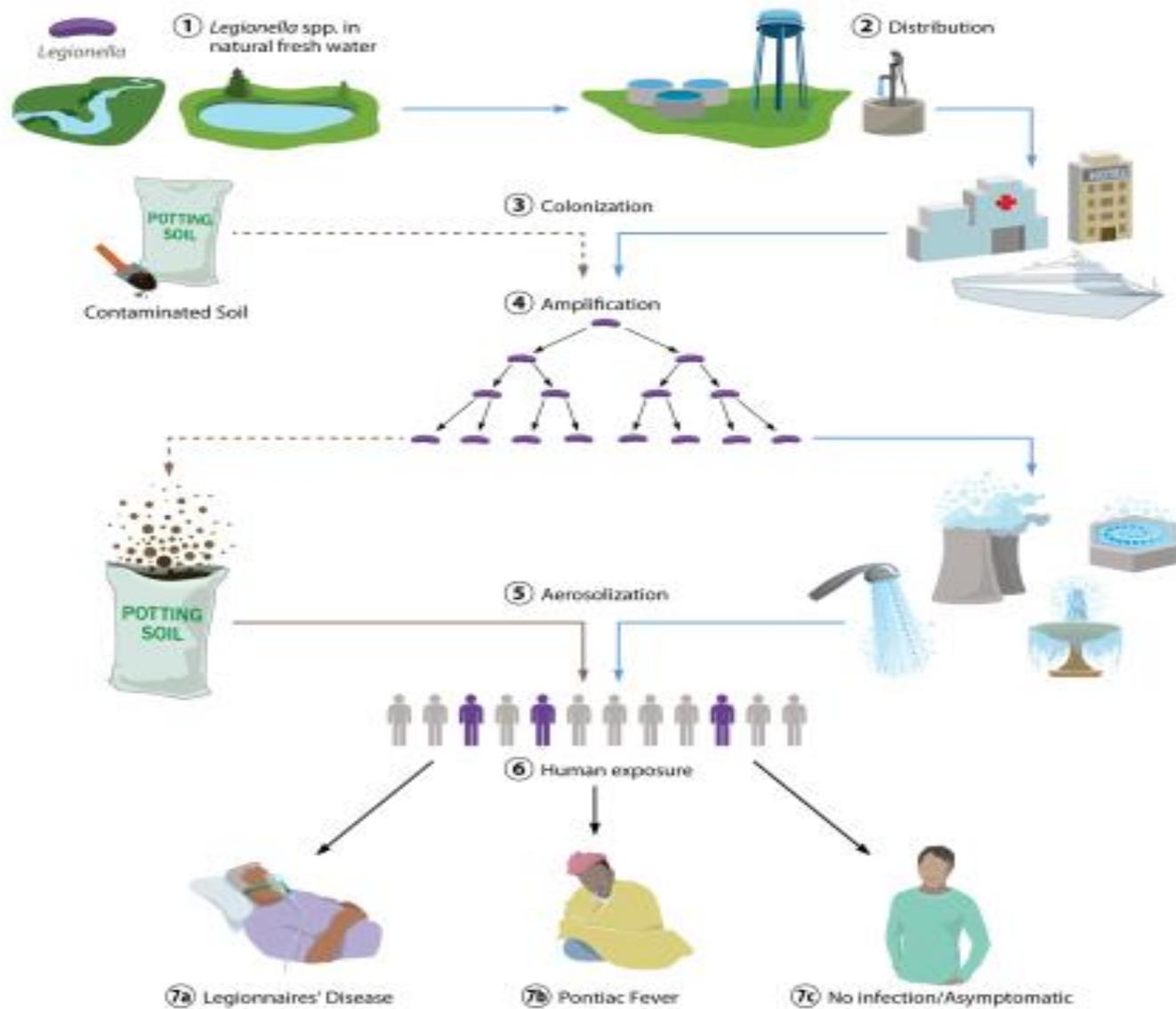


**Amoeba**



# Mode of Transmission

- Aerosolization of contaminated water
- Inhalation of contaminated aerosolsdroplets via steam, mist, air
- Person-to-person transmission very rare if at all.



**Legend**

- Established
- Established (Water)
- Established (Soil)
- - - - - Not established (Soil)



More susceptible/  
high-risk host



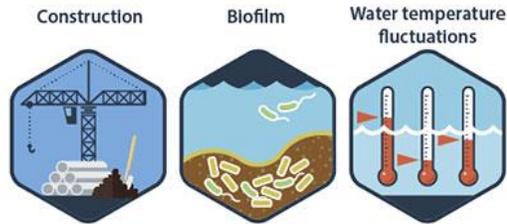
Less susceptible/  
low-risk host

# Scenario 2..continued

- Your IP director, who knows everyone in the hospital, reassures you that there have been no issues with **Legionella**. But a hospital on the other side of town has had some cases of Legionella.

# How *Legionella* affects building water systems and people

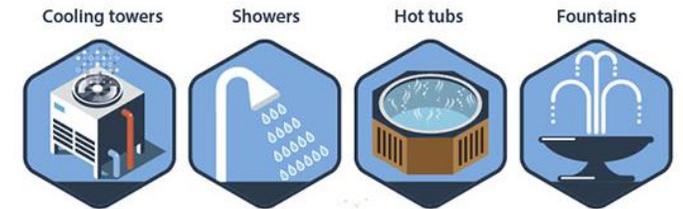
1. Internal and external factors can lead to *Legionella* growth in building water systems.



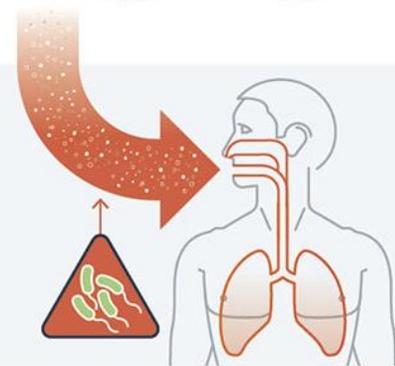
2. *Legionella* grows best in large, complex water systems that are not adequately maintained.



3. Water containing *Legionella* is aerosolized through devices.



4. People can get Legionnaires' disease when they breathe in mist or accidentally swallow water into the lungs containing *Legionella*. Those at increased risk are adults 50 years or older, current or former smokers, and people with a weakened immune system or chronic disease.



# Where is it found?

- Potable and non-potable water systems
- Thermal Conditions
  - Thrive in tepid water (25-37°C) but may survive <20°C and >55°C
  - Can persist & recolonize even after thermal shock treatments (70°C x 30 min)
- Building water systems
  - Plumbing: Pipes/valves & fittings
  - Cooling towers
  - Decorative fountains
  - Whirlpool spas or hot water spring spas
  - Humidifiers
- Fixtures
  - Faucets (electronic & manual)
  - Aerators
  - Showerheads & hoses
  - Ice machines
  - Eyewash stations
  - Water filters
- Medical Devices
  - CPAP machines
  - Bronchoscopes
  - Hydrotherapy equipment



Center for Clinical Standards and Quality/Survey & Certification Group

Ref: S&C 17-30-*Hospitals/CAHs/NHs*  
**REVISED 06.09.2017**

**DATE:** June 02, 2017  
**TO:** State Survey Agency Directors  
**FROM:** Director  
Survey and Certification Group  
**SUBJECT:** Requirement to Reduce *Legionella* Risk in Healthcare Facility Water Systems to Prevent Cases and Outbreaks of Legionnaires' Disease (LD)  
*\*\*\*Revised to Clarify Provider Types Affected\*\*\**

**Memorandum Summary**

- ***Legionella* Infections:** The bacterium *Legionella* can cause a serious type of pneumonia called LD in persons at risk. Those at risk include persons who are at least 50 years old, smokers, or those with underlying medical conditions such as chronic lung disease or immunosuppression. Outbreaks have been linked to poorly maintained water systems in buildings with large or complex water systems including hospitals and long-term care facilities. Transmission can occur via aerosols from devices such as showerheads, cooling towers, hot tubs, and decorative fountains.
- **Facility Requirements to Prevent *Legionella* Infections:** Facilities must develop and adhere to policies and procedures that inhibit microbial growth in building water systems that reduce the risk of growth and spread of *legionella* and other opportunistic pathogens in water.

- Mandate that facilities are required to put forth protocols to prevent bacterial growth in building water systems

# Surveillance Goals

- Incidence & trends
- Rapidly recognize potentially related cases
- Identify risk factors for infection
- Opportunities for prevention
- Monitor effectiveness of interventions

# Surveillance

## Legionella (Legionnaires' Disease and Pontiac Fever)

CDC > Legionella Home > Prevention with Water Management Programs > Special Considerations for Healthcare Facilities

### Legionella Home

About the Disease +

Fast Facts

For Clinicians +

For Health Departments +

For Laboratories

Prevention with Water -

## Water Management Program Validation

According to the CDC/Healthcare Infection Control Practices Advisory Committee (HICPAC) [Guidelines for Infection Control in Health-Care Facilities](#)  and [Guidelines for Preventing Health-care-associated Infections](#) [179 pages], healthcare facilities have two options for validating the efficacy of their water management program (confirming that the water management program is working as intended): 1) performing environmental sampling for *Legionella* or 2) performing active clinical surveillance for infections due to *Legionella*.

## Routine Environmental Sampling

Routine environmental sampling for *Legionella* (i.e., sampling that is performed proactively as part of a water management program, not in the context of an outbreak of *Legionella* growth and transmission in building water systems, not in the context of an outbreak

# CSTE Surveillance case definitions

- Created 2005
- 2010: approved by Council of State & Territorial epidemiologists (CSTE)
- Clinical description:
  - Legionnaire's disease: fever, myalgias, cough, pneumonia
  - Pontiac fever: flu-like illness, no pneumonia
- **Confirmed:** Clinically compatible + 1 Lab criteria
- **Suspect:** Clinically compatible + 1 presumptive lab criteria
- Travel associated: history of 1+ night away from home (domestic or international) within 10d prior to illness onset

# Healthcare Exposure case classifications

- Definite healthcare: **entire** 10d prior to symptom onset in a healthcare facility
- Portion of 10 days prior to symptom onset spent in specific setting
  - Possible healthcare
  - Assisted living
  - Senior living
- Travel: at least 1 night away from home in the 10d prior to symptom onset.

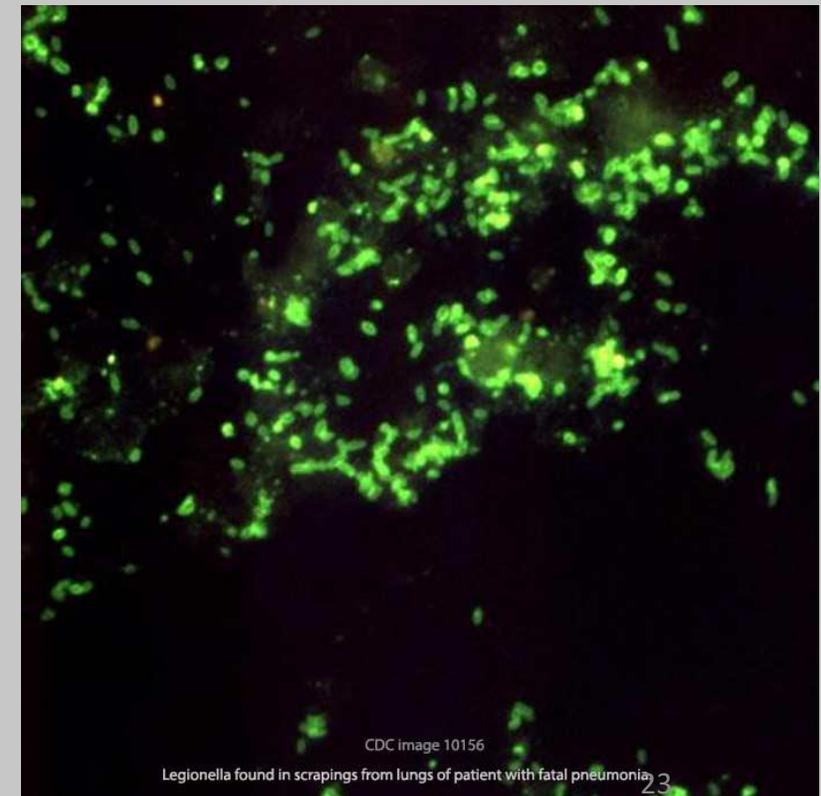
# Lab criteria: Confirmed

- Culture: Legionella isolated from respiratory secretions, lung tissue, pleural fluid or other normally sterile fluid (buffered charcoal yeast extract or BCYE agar at 35°C)
- Urine: detection of *Legionella pneumophila* serotype 1 Antigen
- Seroconversion: 4-fold rise in specific Ab titer to *Legionella pneumophila* serotype 1

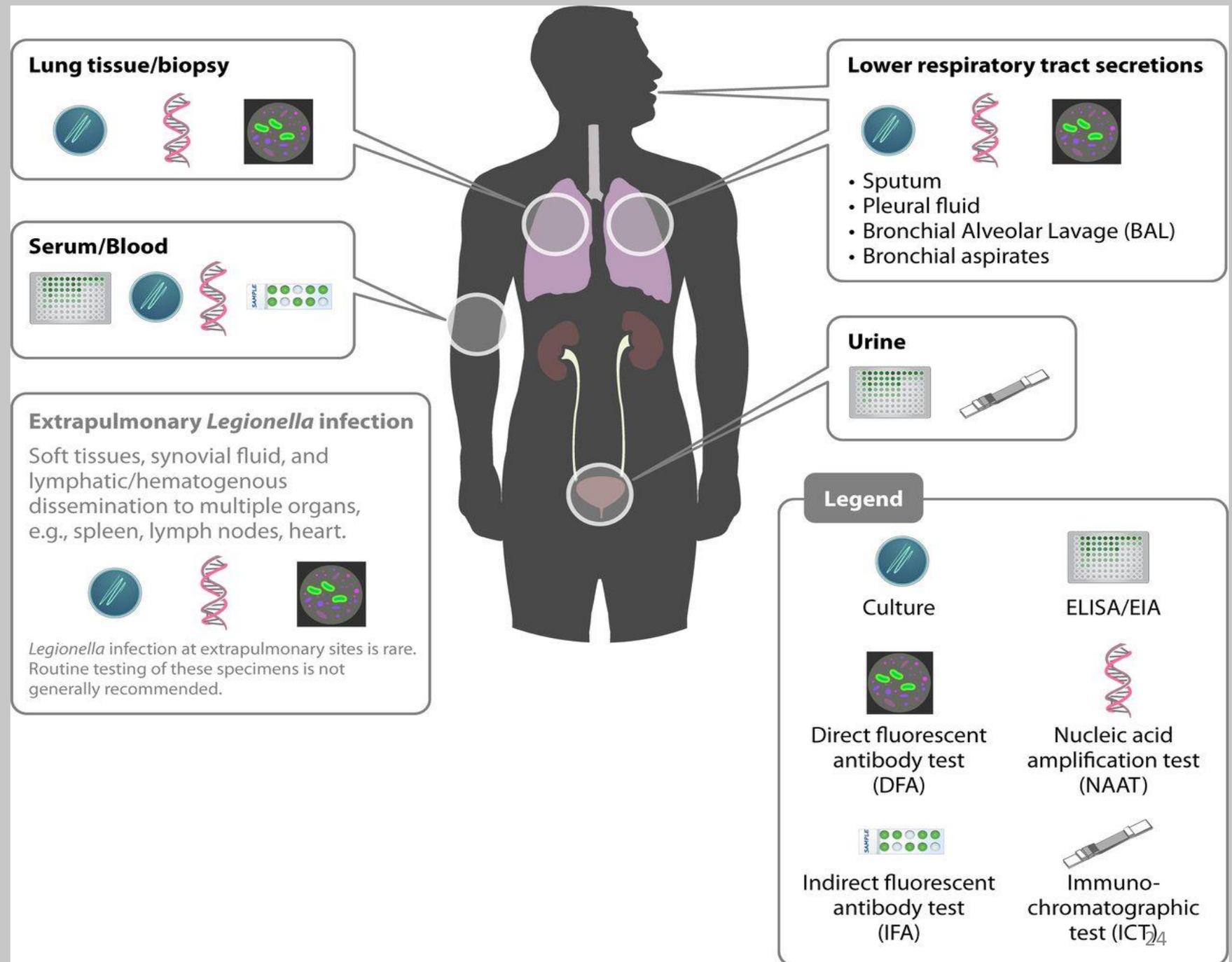
# Lab definition: Suspect

- Seroconversion:
  - Species/serogroups specific:  $\geq 4$ -fold rise in Ab titer, non-*L.pneumophila* type 1
  - Multiple species:  $\geq 4$ -fold rise in Ab titer using pooled antigens
- Histopath: detection of specific Ag or staining of organism in respiratory secretions, lung tissue or pleural fluid (DFA, immunohistochemistry etc)
- Nucleic acid assay: detection of *Legionella* species

CDC, DFA lung tissue



# Diagnostics



## *Legionella* diagnostics

Test	Sensitivity	Specificity
Resp culture	80	100
DFA	33-70	96-99
Urine antigen	70 (95)*	100
Serology	40-60	96-99

\*Higher value for *L. pneumophila* serogroup 1 only

- 1/3 of clinical labs unable to grow pure *Legionella* cx
- Decrease in culture-based detection in US & Europe

TABLE 1 Past, current, and emerging diagnostic tests for *Legionella* and LD<sup>a</sup>

Test or diagnostic method	Specimen type(s)	Assay time to result (sample collection time)	Information provided by positive assay result	Use for confirmative or presumptive LD diagnosis (U.S.)	FDA-cleared or -approved <i>in vitro</i> diagnostic test or reagents commercially available	Advantage(s)	Disadvantage(s)	Sensitivity (%)	Specificity (%)	Notes
Culture and isolation	Sputum, respiratory secretions or tissue, and, more rarely, blood, synovial/joint fluid, or soft tissues	3–14 days for growth plus cysteine biplate test	Together with testing for cysteine auxotrophy, identification of bacteria belonging to the <i>Legionella</i> genus	Confirmatory for <i>Legionella</i> species	Yes	Can detect all <i>Legionella</i> serogroups and species; supports epidemiological investigations	Long incubation and growth times; greater success in experienced laboratories; different specimen types associated with variable sensitivity	<10–80	~100	
Urinary antigen test EIA/ELISA  ICT	Urine	3–4 h (negligible)  15–30 min (negligible)	Infection by <i>L. pneumophila</i> sg1 only	Confirmatory for <i>L. pneumophila</i> sg1 only	Yes	Relatively rapid; availability of sample; may give positive results for long periods even after antibiotic treatment	Only FDA approved for Lp1; less sensitive for other serogroups; variable <i>Legionella</i> antigen excretion	70–90	95–100	
Serology- and antibody-based assays										
IFA (slide and ELISA formats)	Serum	2 h–1 day (3–10 wk for paired sera)	Detects increase in antibody titer against several <i>L. pneumophila</i> serogroups	Confirmatory for <i>L. pneumophila</i> sg1 but presumptive for other serogroups	Yes	Useful when pathogen is not cultured; retrospective epidemiological studies; inexpensive	Does not provide timely, POC information; single titers can be misleading because of high preexisting seroprevalence; not specific for unknown strains or species; technically demanding, and results are subjective; not all cases seroconvert; potential cross-reactivity issues	40–80	95–100	Acute- and convalescent-phase sera collected 3–10 wk apart; sensitivity is lower early in disease
DFA	Sputum, respiratory secretions or tissue, blood	~2 h (negligible)	<i>Legionella</i> serogroup and/or species discrimination	Presumptive	Yes	Inexpensive; commercially available reagents for identification and typing	Cross-reactivity may complicate interpretation; technically demanding	25–75	95–100	Can also be used for serogrouping and species identification

Slide agglutination	Culture isolate	1–2 h (same as for culture)	<i>Legionella</i> serogroup and/or species discrimination	Not applicable	Unknown	Inexpensive; commercially available reagents for identification and typing	Cross-reactivity may complicate interpretation; requires a culture isolate	≥89	≥97	Other variants include latex and serum agglutination; test performance typically based on <i>L. pneumophila</i> detection only; likely less sensitive for non- <i>pneumophila</i> <i>Legionella</i> species
MAb blotting	Culture isolate	1–2 h (same as for culture)	<i>Legionella pneumophila</i> sg1 and subtype discrimination	Not applicable	No	Simple procedure for <i>L. pneumophila</i> sg1 subtyping	Limited availability; requires a culture isolate	Unknown	Unknown	The EMD Duopath and Vircell VIRapid ICT devices report <i>Legionella</i> identification at specificities and sensitivities of ~93–100%; limited availability

Molecular assays

PCR (conventional and real time)	Any sample from which nucleic acid can be isolated	4–6 h (negligible)	Identification and typing of <i>Legionella</i> species and <i>L. pneumophila</i> sg1	Presumptive	No	Rapid; inexpensive; sensitive and specific; validated protocols widely available	Not FDA approved; can be sensitive to inhibitors; requires sophisticated and expensive equipment	30–100	95–100	Sensitivity and specificity dependent on assay design and specimen source
MALDI-TOF mass spectrometry	Culture isolate	~15 min (3–14 days for initial growth)	Identification of bacteria as <i>Legionella</i> spp.	Not yet addressed	Yes	Rapid; inexpensive for established facility and trained personnel	Requires sophisticated and expensive equipment; serogrouping/subtyping not possible; requires a culture isolate	90–99 <sup>c</sup>	Unknown	Sensitivity dependent on species analyzed; pure-culture isolates are used
Isothermal amplification	Any sample from which nucleic acid can be isolated	~1 h (negligible)	Identification of bacteria as <i>Legionella</i> species and <i>L. pneumophila</i> sg1	Presumptive	No	Rapid; inexpensive; less sensitive to inhibitors than conventional PCR	Not FDA approved; still in early stages of development; not widely available	~100 <sup>b</sup>	>90 <sup>b</sup>	Performance and efficiency dependent on assay design

<sup>a</sup> ICT, immunochromatographic test; IFA, indirect fluorescent antibody; DFA, direct fluorescent antibody; EIA, enzyme immunoassay; MAb, monoclonal antibody, MALDI-TOF, matrix-assisted laser desorption ionization–time of flight; sg, serogroup.

<sup>b</sup> Very few studies for comparison.

<sup>c</sup> Compared to *in situ* sequencing.

# Outbreak investigation

- When  $\geq 1$  **definite** case of healthcare associated LD...OR  $\geq 2$  cases of **possible** HCA LD within 12 months of each other
  1. Case identified and reported to health department
  2. Review all patients with healthcare associated pneumonia and test with lower respiratory tract culture and Legionella urine antigen
  3. Begin to identify potential exposure areas, risk factors
  4. Perform environmental sampling as appropriate
  5. Recommendations for immediate control measures
  6. Review/Create water management program

# Is there a water management program?

- Yes: Validate efficacy of program, i.e. Is it working?
  - Routine environmental sampling for Legionella → if >30% positive then all patients with hospital acquired pneumonia should have testing.
  - Active clinical surveillance for infections due to Legionella
- No: time to put a program together (session IV)
  - What can be done in the interim?

# Prevention

# 4 Key principles of WMP to prevent Legionella\*

1. Maintaining water **temperatures** outside the ideal range for *Legionella*\* growth
2. Preventing water **stagnation**
3. Ensuring adequate **disinfection**
4. Maintaining **devices & plumbing** to prevent scale, corrosion, and biofilm growth, all of which provide a habitat and nutrients for *Legionella*\*

\*and other pathogens such as non-tuberculous mycobacteria and gram-negative organisms

## Preventing Legionnaires' Disease: A Training on Legionella Water Management Programs (PreventLD Training)

- <https://www.train.org/main/course/1081923/>
- Hands on implementation of toolkit recommendations

# Routine Environmental Sampling

- THAB\*—total heterotrophic aerobic bacteria counts
- Quantitative Legionella PCR: detected vs not-detected
  - Does not specify viability
  - Newer molecular viability analyses can be helpful
- Culture: Viable vs Non-viable & Culturable vs Non-culturable
  - Non-viable, non-culturable: GOOD!
  - Viable, culturable: BAD!
  - Viable but non-culturable (VBNC) is of biggest concern.
    - Studies have demonstrated that organisms can turn active if placed in the right environment and thus be a potential issue.

\* Does not replace actual testing for Legionella

<https://www.cdc.gov/legionella/health-depts/environmental-inv-resources.html>

# Prevention of Legionella

## Primary prevention:

- Temperature
  - Cold water storage/distribution: <math><20^{\circ}\text{C}</math> (<math><68^{\circ}\text{F}</math>) ideally
  - Water heated to <math>66^{\circ}\text{C}</math> (><math>150^{\circ}\text{F}</math>)
- Prevent water stagnation → reduces biofilm
- Water restrictions

## Secondary Prevention

- Focal disinfection
  - UV light
  - Ozone
  - Point of use filters
- Systemic Disinfection
  - Super heat & flush (2-3 months)
  - copper-silver ionization
  - Hyperchlorination (corrosion)
  - chlorine dioxide
  - monochloramine

# Water temperature

- Hot water stored at 60°C (140°F) and circulated  $\geq 124^{\circ}\text{F}$  ( $\geq 51^{\circ}\text{C}$ ) → install thermostatic mixing valves to minimize risk of scalding
- Cold water temperature at  $< 20^{\circ}\text{C}$  ( $< 68^{\circ}\text{F}$ )
- If unable to allow hot water temperature as above, then intermittently hot water heating  $\geq 150^{\circ}\text{F}$  ( $\geq 66^{\circ}\text{C}$ ) or chlorine flush through the system

# Efficacy of Chlorination

- Actual concentration of chlorine
- contact time
- pH (efficacy decreases  $>7$ )
- Temperature
  - At high temp: bactericidal activity increases but free chlorine degrades
- turbidity
- buffering capacity of the water,
- concentration of organic matter, iron
- the number and types of microorganisms in the water system (in biofilms and free-living)

# Chlorine Dioxide

- Fast acting
- More effective at higher temperatures
- Less corrosive
- Wide pH range 4-10
- Penetrates biofilms
- Requires registering with local regulatory bodies

# Scenario: update

- After meeting with facilities, campus wide testing revealed a few sinks with detectable Legionella via PCR and culture non-viable, non-culturable.
- The decision is made to formally establish a water management program to continue annual testing for Legionella and to evaluate the high incidence of non-tuberculous mycobacteria.

# References

- Comprehensive list of websites and articles will be provided at the end of my last talk.

Thank you!

Questions?

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