

Legionella – Its Ecology, Epidemiology and Control ©

Diane Miskowski, MPH
Business Development Manager
Westmont, New Jersey



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Overview

- **History and Ecology of the Organism**
- **Diseases and Risk Factors**
- **Transmission**
- **Potable Water Systems**
- **Cooling Tower Water Systems**
- **Analytical Methods**
- **Proper Sampling and Shipping**
- **Choosing a Lab**

History of Legionella

- **Major outbreak of pneumonia at the 1976 American Legion Convention in Philadelphia**
- **240 cases of pneumonia with 34 fatalities**
- **Causative agent was unknown at the time-earlier outbreaks went undiagnosed**

The Disease-Legionellosis

- Eventually the bacterium was isolated, identified, and named *Legionella pneumophila*
- Bacterium must be inhaled deep into the lung via water droplets
- Water droplets must be between 1 and 5 microns in diameter

Legionellosis

- **Two illnesses of concern**
 - **Legionnaires Disease**
 - **Pontiac Fever**
- **Estimated 25,000 to 75,000 cases per year in US-most go unreported***
- **Reportable disease in US, Europe, Australia, Singapore**

Legionellosis – Different Illnesses

- **Legionnaire's Disease**
- **Non-pneumonia Legionellosis**
- **Pontiac Fever**
- **Asymptomatic Antibody Response**
- **No response**

Legionnaires Disease

Low Attack Rate 5%

- **High Mortality 15%**
- **Incubation 2-14 days**
- **High Fever 102⁰ -105⁰**
- **Difficulty Breathing**
- **Chest/Back Pain**
- **Pneumonia with multiple organ failure**
- **May have long term sequelae**

Pontiac Fever

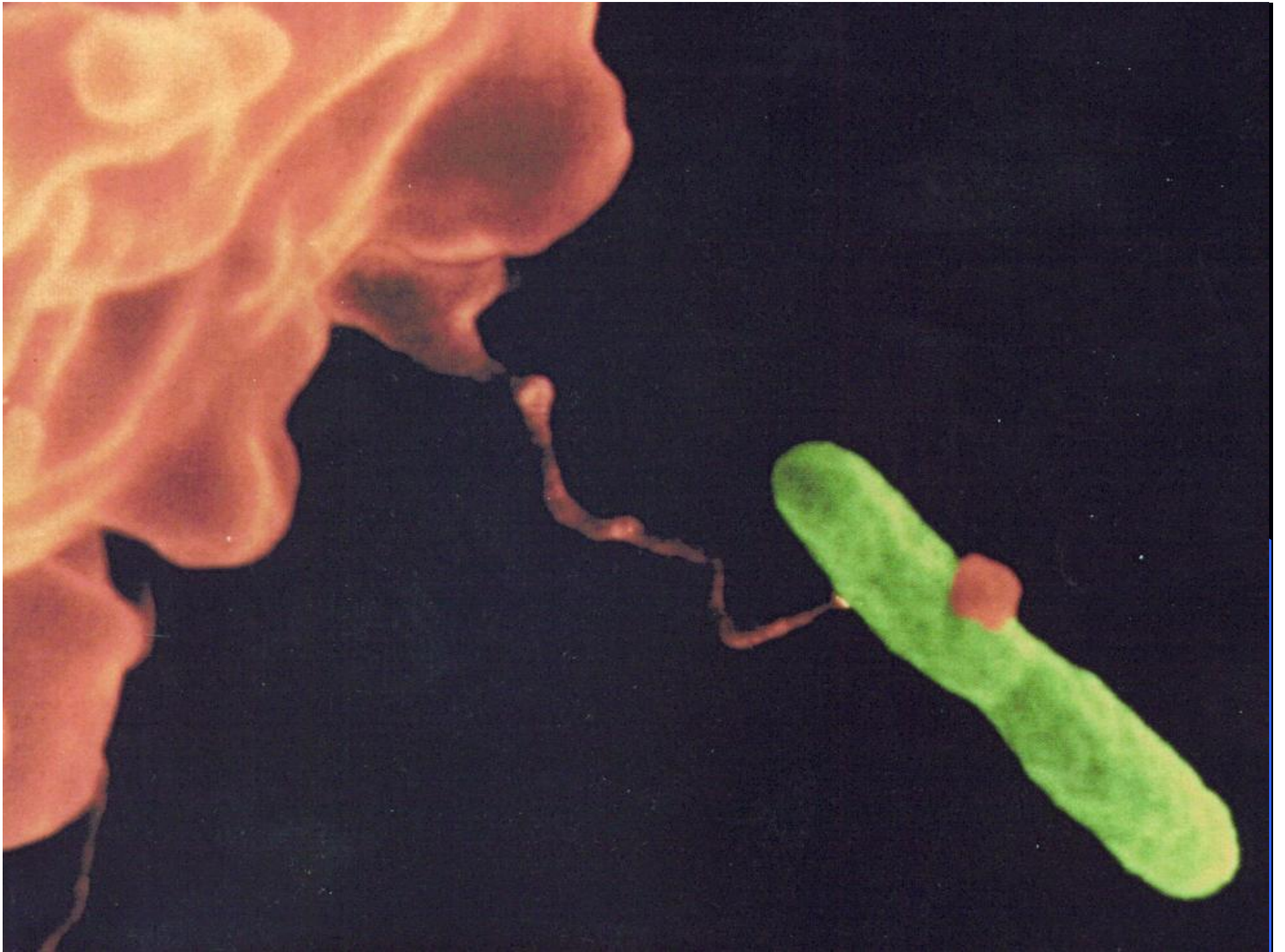
- **High Attack Rate** 95%
- **Low Mortality** 5%
- **Incubation** 1-3 days
- **Muscle Aches**
- **Flu-like symptoms**
- **No long term sequelae**

Legionellosis – Risk Factors

- **Community and Hospital Acquired**
- **Age**
 - . **Highest risk in elderly >65**
 - . **Not common in people <50**
 - . **Very rare in people <20**
- **Smoking**
- **Pre-existing COPD, diabetes**
- **Renal disease**
- **Compromised immune system**

Legionella Bacterium

- Naturally occurring with over 80 species and 60 serogroups –fresh and estuarine surface water and groundwater. Half are pathogenic.
- *L. anisa* and *L. pneumophila* in env. samples
- Gram negative, rod shaped bacteria with a flagella (tail)
- Symbiotic relationship with amoeba, protozoans, blue green algae
- May survive 135 + days in room temp. distilled water; over a year in tap water



Coiling phagocytosis

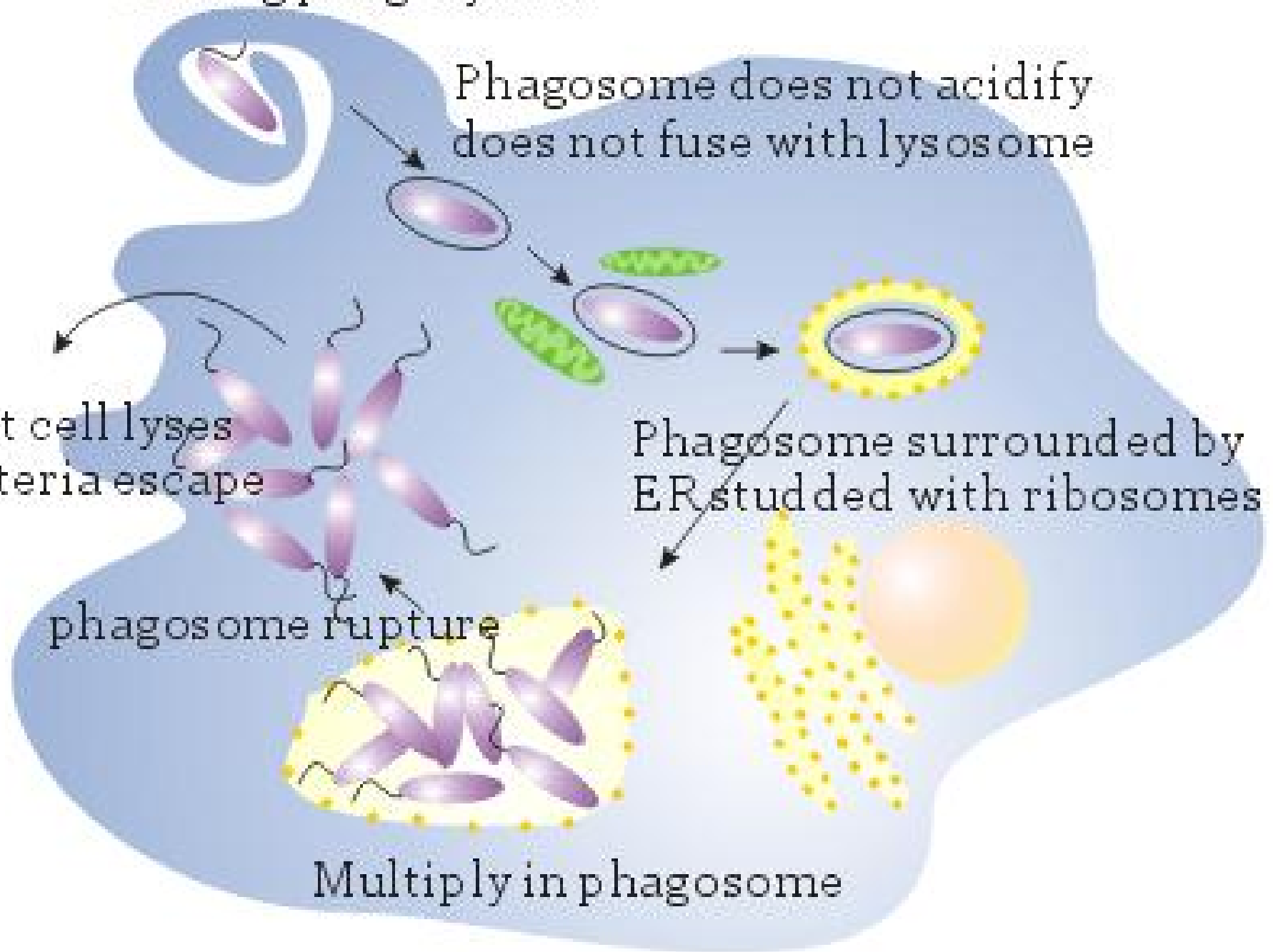
Phagosome does not acidify
does not fuse with lysosome

Host cell lyses
Bacteria escape

Phagosome surrounded by
ER studded with ribosomes

phagosome rupture

Multiply in phagosome



Legionella Bacterium-Characteristics

- **Fastidious Bacteria**
 - 68⁰ F to 120⁰ F for viability**
 - 77⁰ F to 113⁰ F for amplification**
 - Needs iron**
 - Needs L-Cysteine**
 - Lower Dissolved Oxygen**
 - pH of 4.5 to 10.5**
 - Rubber, some plastics support growth**
 - Copper inhibits growth**

Legionellosis Causation – Synchronicity

- **Viable bacteria in natural reservoir**
- **Amplification in water source-growth requirements**
- **Dissemination-respirable droplets formed by aerosolization**
- **Transmission-infected, respirable bioaerosols must travel thru air**
- **Inhalation by susceptible human host→alveoli→pulmonary macrophage**

Note: No person to person transmission

Transmission - Infected Aerosols Need to Survive the Environment

- **Bacteria won't survive in airborne droplets in the environment**
 - . **Low humidity**
 - . **Temperature**
 - . **UV light**
 - . **Distance to susceptible host**
- **If optimal conditions exist, infected aerosols can survive >1 mile***

Amplification - Role of Temperature

< 68⁰ F	Bacteria dormant but viable
68⁰ to 120⁰ F	Multiplication
122⁰ F	90% Kill in 2 Hours
140⁰ F	90% Kill in 2 Minutes
158⁰ F	100% Rapid Kill

Not absolutes-species, plumbing system, degree of biofilm, sediment, scale, etc.

Dissemination

Sources of Infected Bioaerosols

- **Cooling Tower Mist-down drift**
- **Evaporative Condensers**
- **Shower Heads and Faucets**
- **Heated Spa Baths and Whirlpool Baths**
- **Architectural Fountains and Waterfalls**
- **Misters, Air coolers, Humidifiers**
- **Metalworking Fluids-Water Based**
- **Nebulizers and Respiratory Therapy Equipment**

Potable Water System-Most Outbreaks Associated with Potable Water

- **Hot and cold water used for drinking and bathing**
- **City, well, or surface water**
- **Where water enters the building to where it exits the building**
- **Water main inlet, all piping, hot water heaters, storage tanks, distribution outlets and faucets, shower heads**
- **Semi-annual routine monitoring**

Legionella in Potable Water-What Influences Growth?

- **Chlorine concentration – 2mg/L***
- **77⁰ to 108⁰ F-ideal for amplification***
- **System Design-low flow, infrequent use**
- **Age of the System-biofilm, sediment***
- **Plumbing Materials-rubber, some plastics, leached organics**

Potable Water System-Maintenance/Control

- **Hospitals, Health care, Nursing Homes**
 - **Store cold water below 68⁰ F**
 - **Store hot water above 140⁰ F (scalding)**
 - **Install preset, thermostatic mixing valves, or**
 - **Periodically increase temperature within system to 150⁰ F or chlorinate**
- **Store hot water above 120⁰ F**
- **Bleach and rinse showers heads; remove aerator screens in faucets**

Potable Water System-Maintenance/Control

- **Maintain detailed engineering drawings or floor plans that include piping systems**
- **Hot water heaters/storage tanks-heating element located at bottom of tank**
- **Insulated recirculation loops and pipe runs**
- **Mix hot and cold water near the showerhead**
- **Warm water section of pipes should be self draining**
- **Short pipe runs**
- **Avoid dead legs**

Potable Water System – Disinfection after an Outbreak-Superheating

- **Super heat hot water system to 150° F**
- **Treat and trickle flush the system for one hour for every 10 years of age***
- **Numbers of outlets that can be trickle flushed simultaneously will depend on hot water system capacity and flow capacity**
- **Stresses old systems**
- **Recolonization occurs-treat periodically**
- **Treat the system when building occupancy is low-prevent scalding**

Potable Water System – Disinfection After an Outbreak-Chlorination

- **Chlorinate so that 2-5 mg/L of free chlorine exists throughout the system for ≥ 2 hours**
- **May require chlorination of the storage tanks to 50 mg/L**
- **Maintain water pH between 7.0 and 8.0**
- **Corrosive at 2mg/L**
- **Recolonization occurs-treat periodically**

Potable Water System – Disinfection After an Outbreak-Chlorination

- **UV Light***
- **Ozonation***
- **Maintenance chlorination**
- **Copper-silver ionization**
 - **Proper concentration varies**
 - **Very effective prevents re-colonization**
 - **pH dependent**

Emergency Water Systems

- **Safety showers, eye wash stations, fire sprinkler system**
- **Stagnant conditions at temperatures conducive to growth of Legionella**
- **Vigorously flush showers and eye washes monthly.**

Hot Tubs and Whirlpool Spas

- **Temperatures and nutrients optimum for Legionella growth;**
- **Increased Temperatures accelerate the loss of biocides**
- **Organics (nutrients) raise the level of free halogens needed for disinfection**

Hot Tubs and Whirlpool Spas- Maintenance

- **Follow filter flow rate guidelines and backflushing frequency; replace filter cartridges weekly**
- **Follow ANSI/NSPI chemical standards for pool disinfection**
- **Critical to maintain free available halogen level**
- **Automatic systems that monitor and dose offer best control of water chemistry**
- **Spas should be superhalogenated daily; replace spa water and scrub surfaces to remove biofilm weekly**

Cooling Towers and Evaporative Condensers -Maintenance

- **Historically, impacts largest number of people**
- **Keep the cold water basin and mechanical filters clean and free of dirt and debris**
- **Requires routine maintenance, inspection, and water treatment by professionals→minimize microbial growth, scale, corrosion, sediment accumulation**
- **Quarterly Routine Monitoring**

Cooling Towers and Evaporative Condensers-Shut Down and Emergency Decontamination

- **System Shut Down-** if cooling tower is down for 3 or more days, the entire system (tower, piping, heat exchanger) should be completely drained
 - | **If shut down is of shorter duration, stagnant cooling tower water must be treated with biocides before start up**
 - | **Emergency decontamination from outbreak-follow guidelines from the Cooling Tower Institute using chlorine-10mg/L for 24 hours**

Cooling Towers and Evaporative Condensers-Siting

- **Locate as far as possible away from fresh air intakes**
- **Do not locate near kitchen exhausts, plant manufacturing exhausts, truck bays**
- **Locate upwind of outdoor public area**

Sampling and Monitoring for Legionella

- **Currently no standards or regulations for controlling of Legionella;**
- **CDC and TX, NJ, NY, Allegheny County PA, Los Angeles County, CA and Canada have guidelines for monitoring healthcare facilities ; one state has guidelines for monitoring cooling towers (WI)**
- **Professional organizations and trade groups have guidelines-ASHRAE, AWT, Cooling Tower Institute, ASTM, ANSI/NSPI**

Choosing Sampling Methods

- Optimize Recovery of the bacteria by looking for its source
- Water Sampling where aerosolization may occur
- Swab Sampling of biofilm or slime
- Air Sampling
 - Not recommended
 - Legionella* do not survive drying or may be stressed

Quarterly Cooling Tower Sampling

- **Tower Makeup**
- **Tower Sump (away from makeup)***
- **Inlet to Heat Exchanger**
- **Outlet from Heat Exchanger**
- **Distribution Pack**
- **Tower Pack***

***Test routinely**

Test all items to establish baseline or during an outbreak

Semi-Annual Potable Water Sampling

- **City Water Main Entry Point**
- **Storage Tanks**
- **Hot Water Heater Drain Point***
- **Hot Water Return**
- **Last Point on Cold Water/First Point on Hot Water**
- **Last Point on Hot Water*/First Point on Cold Water**
- **Selected Outlets (10%)**

***Test routinely**

Test all items to establish baseline or during an outbreak

Sample Collection –Potable Water

- | Collect two if possible from hot and cold water supplies
 - > first draw- take first 1000 ml directly from the tap. Let water run for 60 seconds or longer then take the
 - > second draw sample
- | Obtain swabs from the lab. Remove aerators and swab inside faucet or showerhead. Get swabs from the lab
- | Sample every 6 months for routine monitoring

Sample Collection

- Use personal safety precautions during sampling-
high efficiency particulate respirators, gloves,
safety goggles
- Take care not to generate aerosols when sampling
- Obtain sterile, preserved bottles from the lab
 - 1000 ml bottles for potable (drinking) water
 - 250 ml bottles for non-potable water
- **Leave air space in the bottles!**

Proper Shipping

- **Overnight delivery to the lab***
- **Ship in insulated containers-avoid temperature extremes**
- **Use freezer packs only –NOT ICE**
- **Include Chain of Custody**
- **Proper identification**
- **Leak proof containers –seal lids with tape**

Selecting Analysis

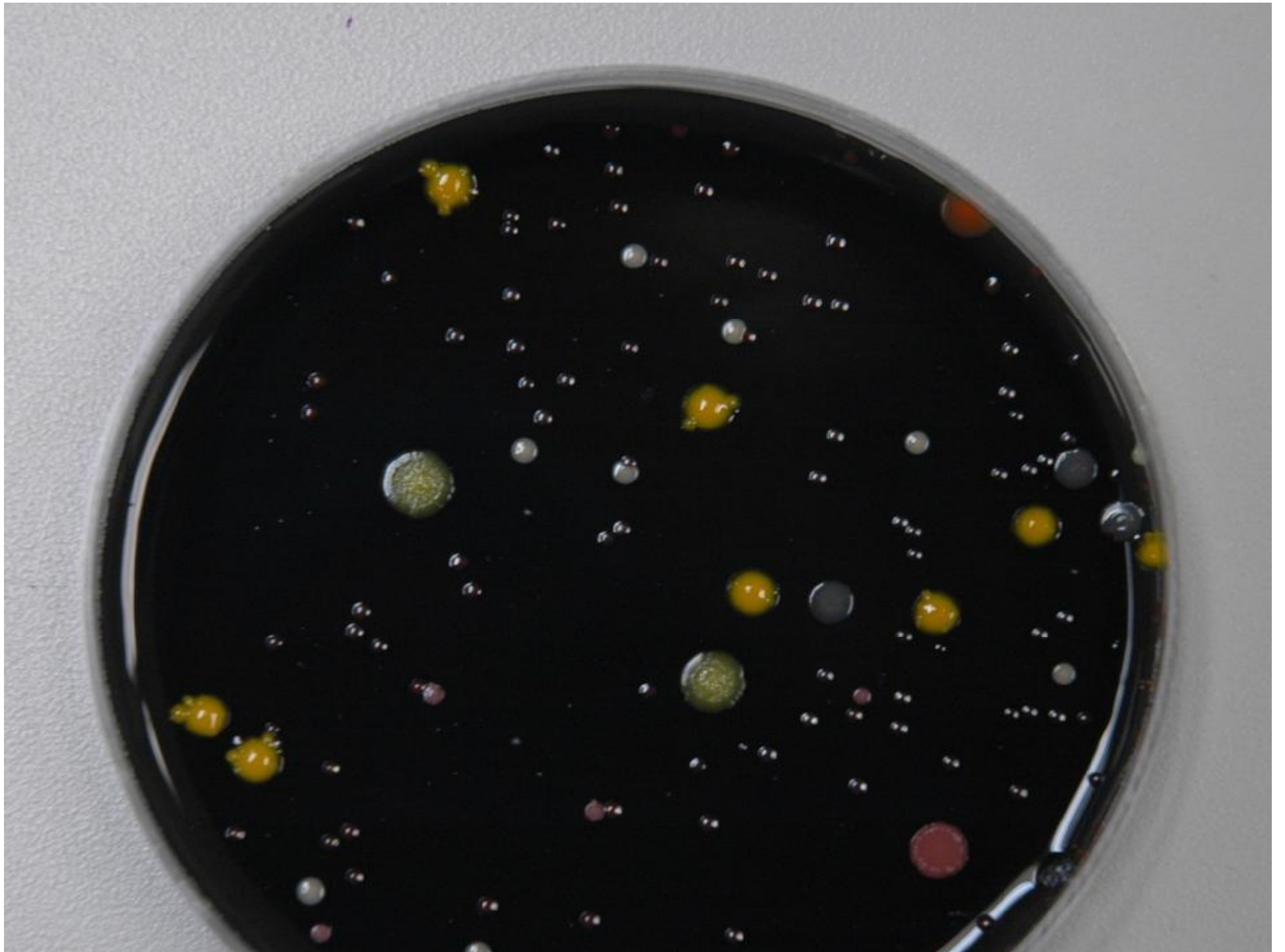
- Methods that are selective for *Legionella* bacteria
- Conventional culture methods versus PCR
 - Culture methods-24 species and associated serotypes. *L. pneumophila**
 - PCR 4 species *L. pneumophila, micdadei, maceachernii, sainthelensis/cincinnatiensis* *

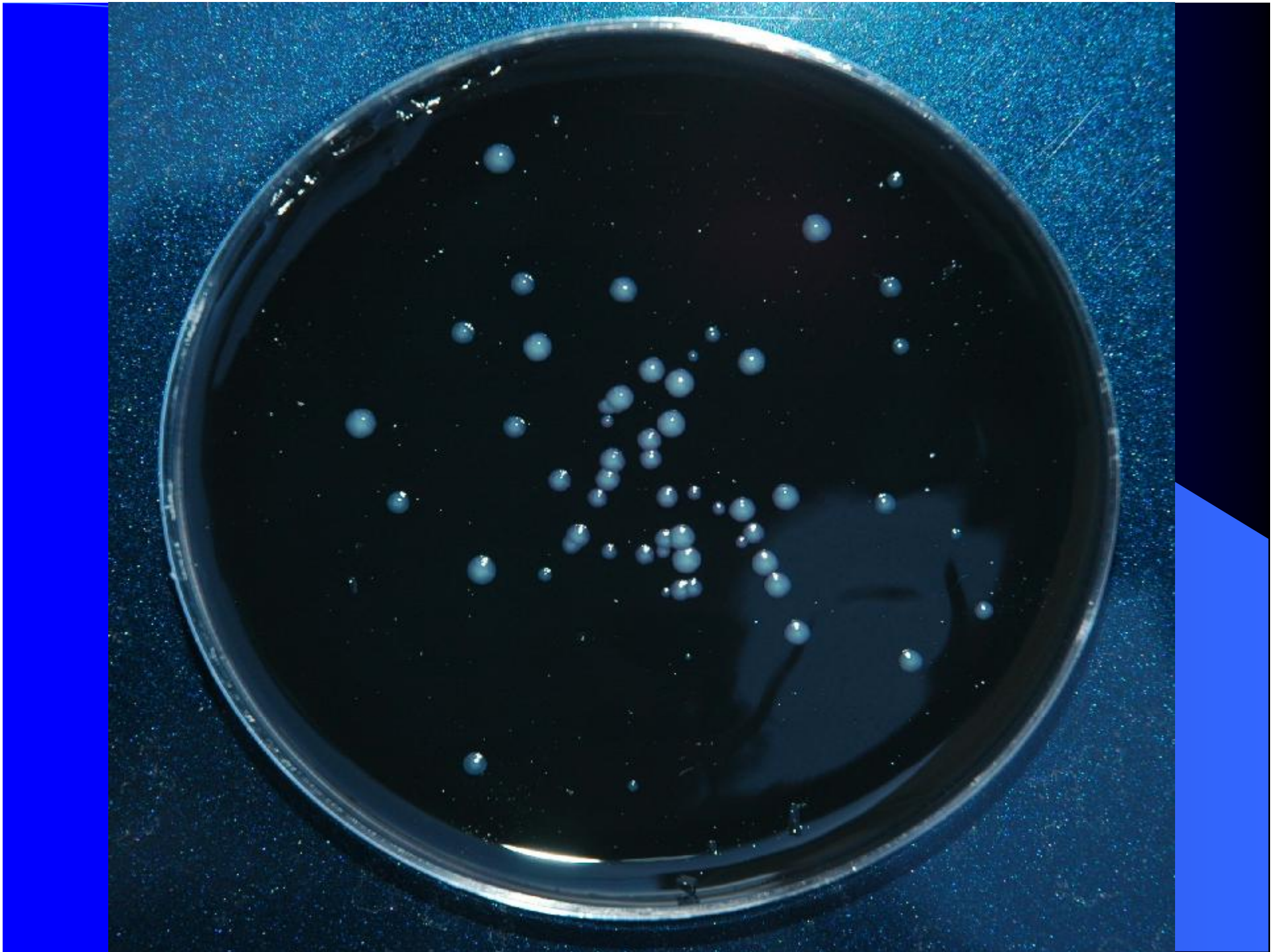
Selecting the Proper Analysis

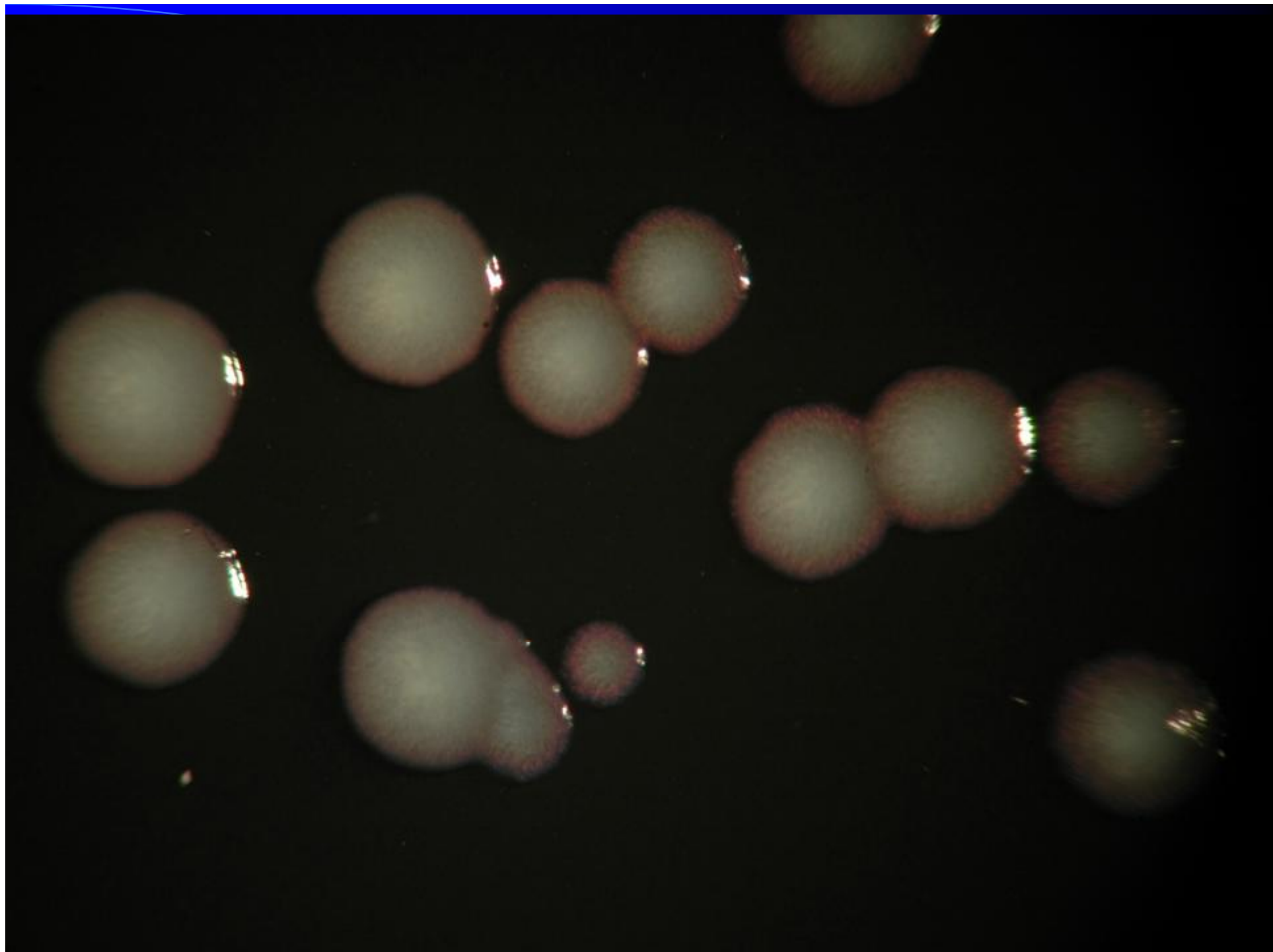
- Culture based method – still the recommended method worldwide. Quantitative
 1. Potable water – filtered or centrifuged to concentrate CFU/1000 ml or CFU/Volume sampled
 2. Non-potable water – acid or heat pretreated CFU/ml
 3. Heat Enrichment
 3. Detection limit varies depending upon amount concentrated

Culturable Analysis

- **Culture on selective media (4 types)- viable cells grow**
 - **Incubate at 35-37°C**
 - **Takes 10-14 days**
 - **ID to species and sero-types**
 - **PFGE or PCR-identify species and strains in tissue samples and env. samples after an outbreak to pinpoint source**







Direct Fluorescent Assay (DFA)

- **Not OK directly testing env. samples-false negatives and positives**
- **OK for clinical samples**
- **Used for species and serotype identification from pure cultures of Legionella grown from environmental samples**

Polymerase Chain Reaction (PCR)

- **Genetic test on DNA-species specific**
- **+/- Detection**
- **Very sensitive and high accuracy**
- **Rapid turnaround (same or next day)**
- **Detection limit-1 DNA fragment**

PCR

- **Sample matrix effects-false negatives**
- **Cannot distinguish between viable and non-viable cells**
- **Not a measure of infectivity-DNA not infectious**
- **Only 3 species**
- **Comparing results PCR>culturable**

Purpose of Sampling

- **Proactive-effectiveness of maintenance program**
 1. +/- or concentration
 2. Frequency of + are more predictive of problems than concentration
- **Reactive-outbreak sampling-Concentration of individual species & serotyping**
- **Concentrations are relative**

Result Interpretation

- **Non-detect-continue maintenance program and periodic testing***
- **Escalating course of action based on concentration**
 - § **Action Level 1**
 - § **Action Level 2**

OSHA Action Levels

Performance Level: Non-Detect

OSHA Action Levels	Non-Potable CFU/ml	Potable CFU/ml	Humidifiers CFU/ml
1	100	10	1
2	1000	100	10

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Selecting a Laboratory

- **High degree of skill and experience to be able to grow the organism and correctly identify it**
- **AIHA EMLAP-doesn't certify Legionella**
- **CDC Method**
- **ISO Method for EU or Canadian clients**
- **Be able to prepare pure culture isolates**
- **Store pure culture isolates (-70°C)**

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- | *Procedures of the Recovery of Legionella from the Environment*, US Center for Disease Control, January, 2005
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