



Processing Water - I Thought It Was Sanitary

Moderator: Josie Greve-Peterson PSSI Food Safety Solutions, United States
Organized by the Water Safety and Quality PDG

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Today's Presenters



Josie Greve-Peterson
PSSI Food Safety Solutions, United States

Josie Greve-Peterson is the Corporate Microbiologist for PSSI Food Safety Solutions where she develops and implements strategies and programs to mitigate food safety risks, which includes providing microbiological and technical support. Josie has a B.S. and M.S. degree in microbiology, more than 11 years of experience in the food industry in various quality management and food testing roles, and serves as Vice-Chair of the Dairy PDG.



Diane Walker
Montana State University, United States

Diane Walker is a Research Engineer at Montana State University's Center for Biofilm Engineering where she works with an interdisciplinary team of engineers, microbiologists and statisticians in the Standardized Biofilm Methods Lab (SBML), developing methods to grow, treat, sample and analyze biofilms for use by academia, industry and the regulatory agencies. Diane has degrees in biology, bio-resources engineering, and environmental engineering from MSU, and has worked with the SBML for more than 15 years.

Today's Presenters



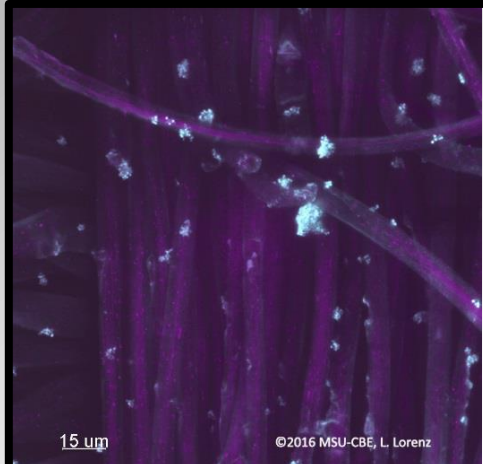
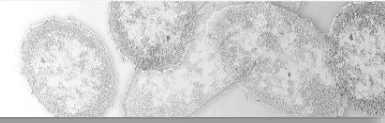
Phyllis Posy
Posy Global, Israel

Phyllis Posy has extensive experience working with city, state, federal and international governments, and stakeholders, bringing environmental, water and reuse technologies to market, and implementing them to improve water and food processing. She is President of Posy Global in Jerusalem, Israel, and Secretary of the Water PDG.



Neil Bogart
Ecolab, United States

Neil Bogart is an Executive Area Technical Support Coordinator for the Food & Beverage division of Ecolab. Prior to Ecolab, he worked in food manufacturing in Quality and Regulatory and was a Food Safety Consultant specializing in food safety program development and sanitation systems. He is also a Chef. Neil holds a Food Science degree from Mississippi State University and his Le Cordon Bleu from Monroe County Community College.



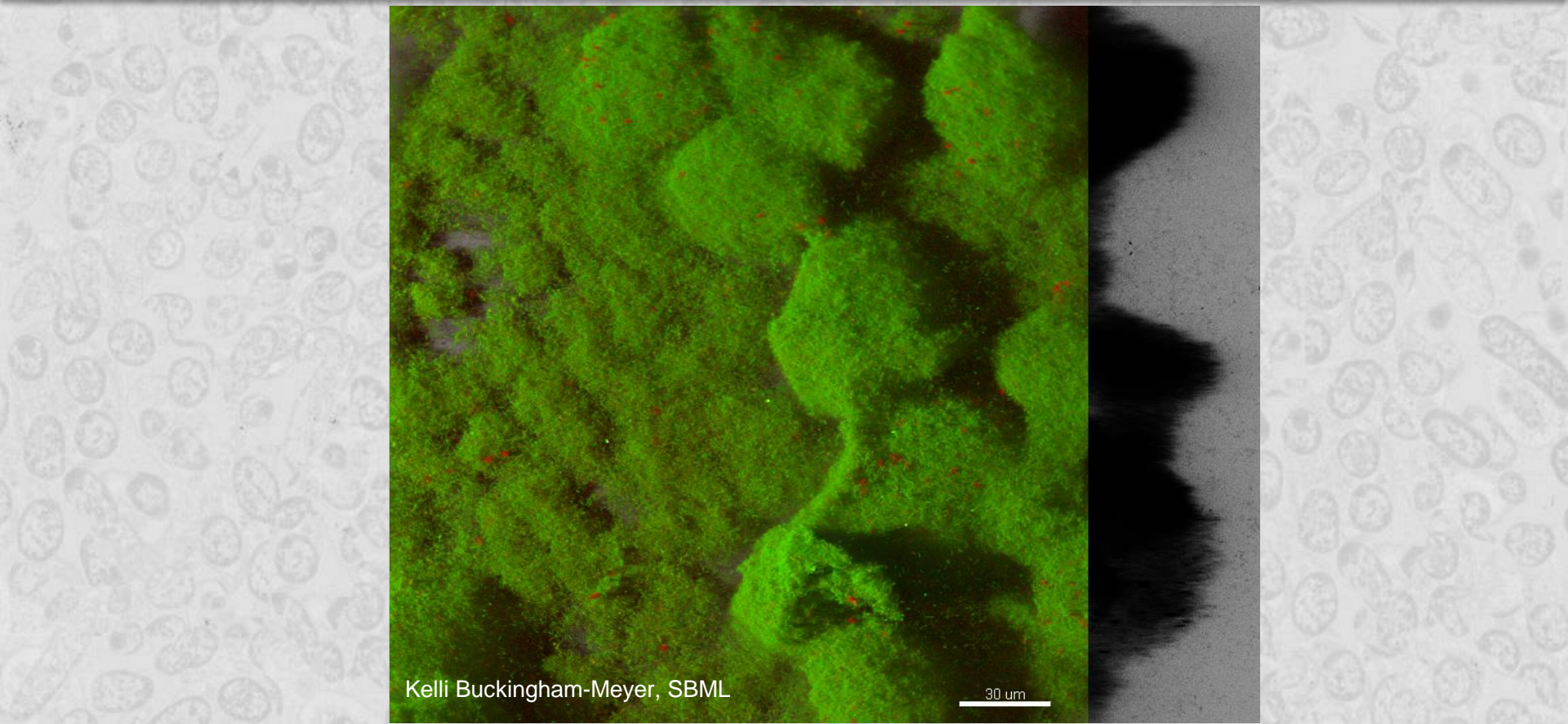
Biofilms

Diane K. Walker
Research Engineer
Standardized Biofilm Methods Lab
Montana State University



IAFP Webinar | June 8, 2021

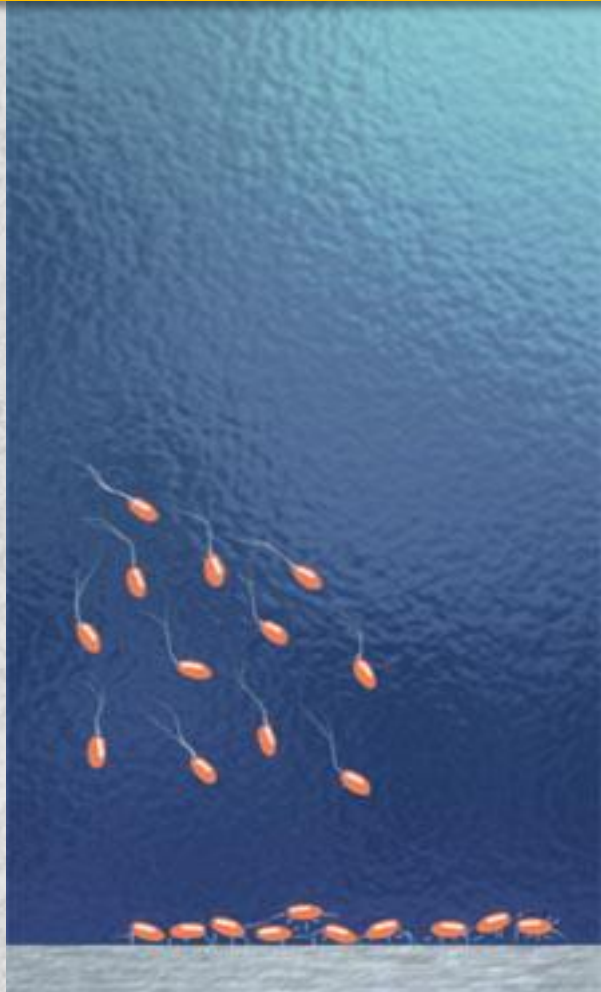




Kelli Buckingham-Meyer, SBML

30 µm

Biofilms are a self-organized, community of microorganisms embedded in a matrix of extracellular polymeric substances.



**Free-swimming
cells alight on a
surface and attach**

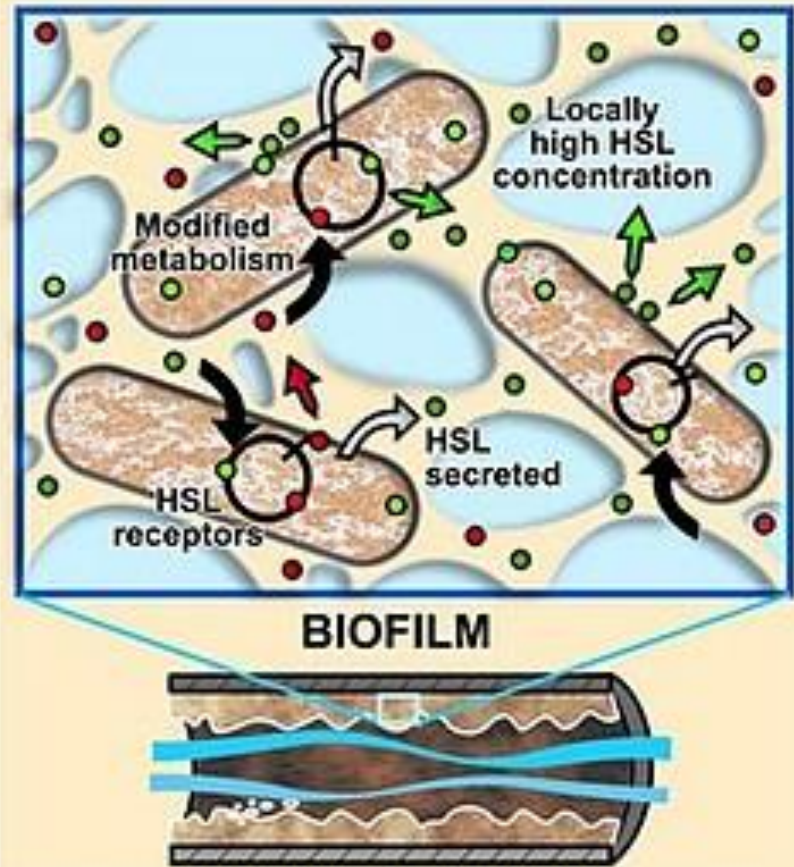
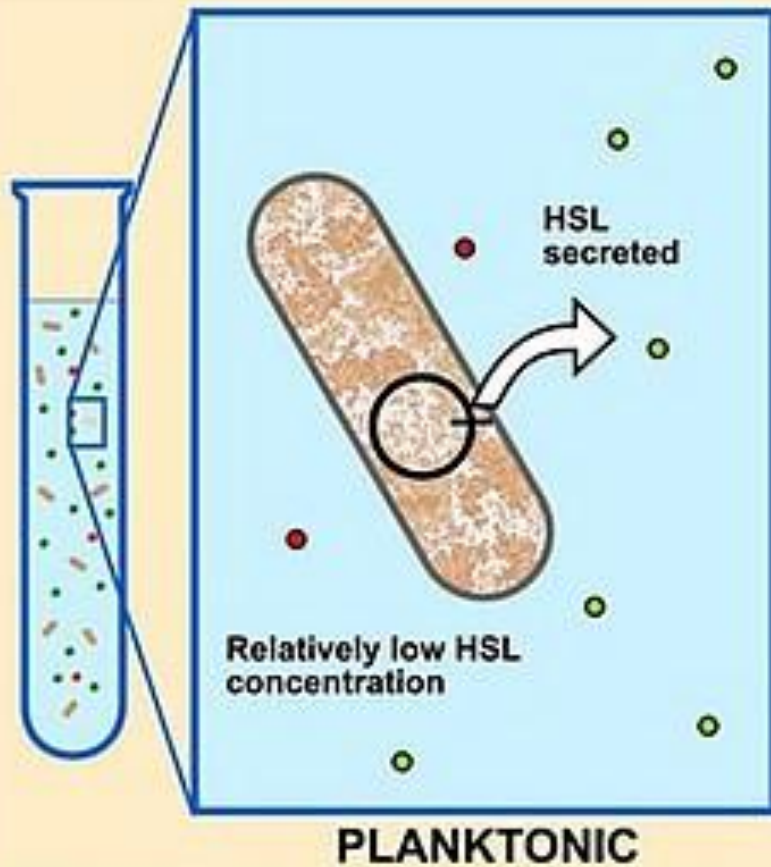


**New genes are
expressed to synthesize
matrix polymers**

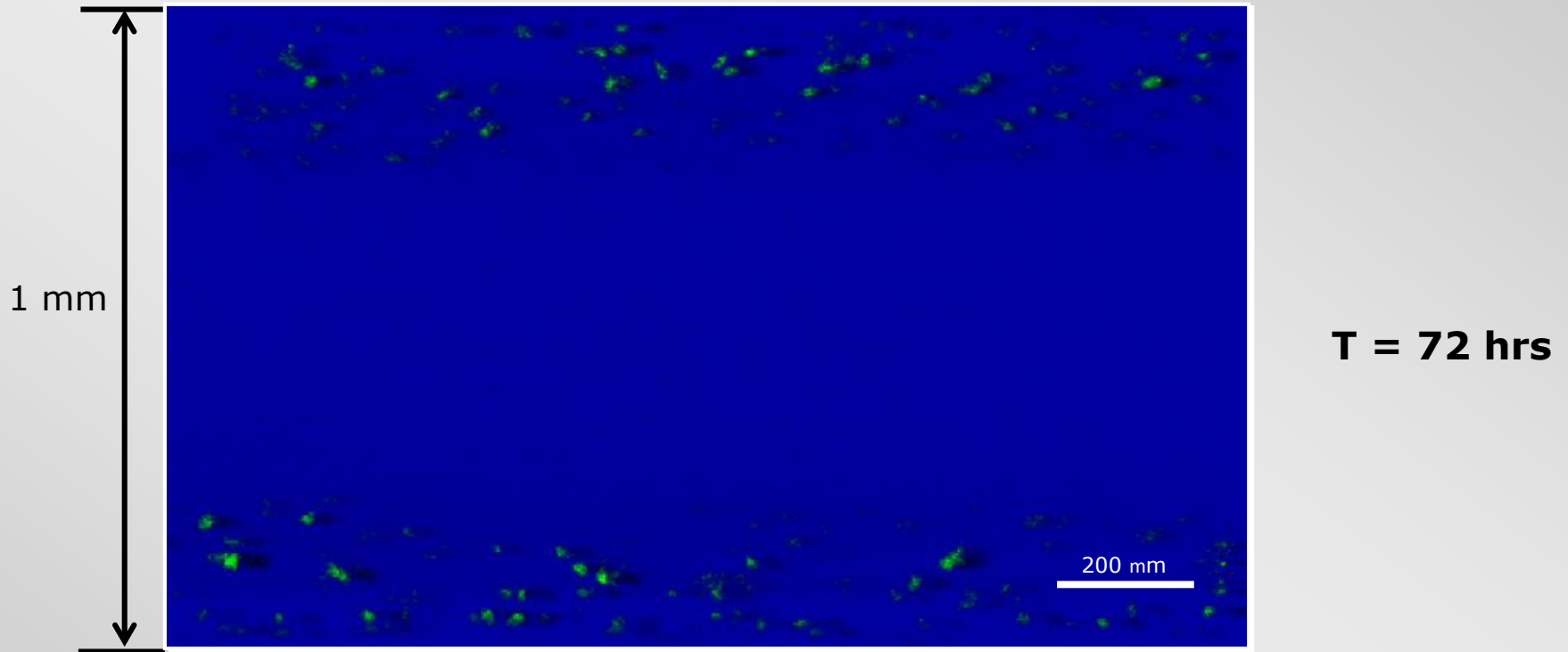


**Cells coordinate
by exchanging
signaling molecules**

Quorum Sensing



Pathogen Survival in Biofilm



Green is *gfp* *P. aeruginosa* PA01

Red is *dsRed* *E. coli* 0157:H7

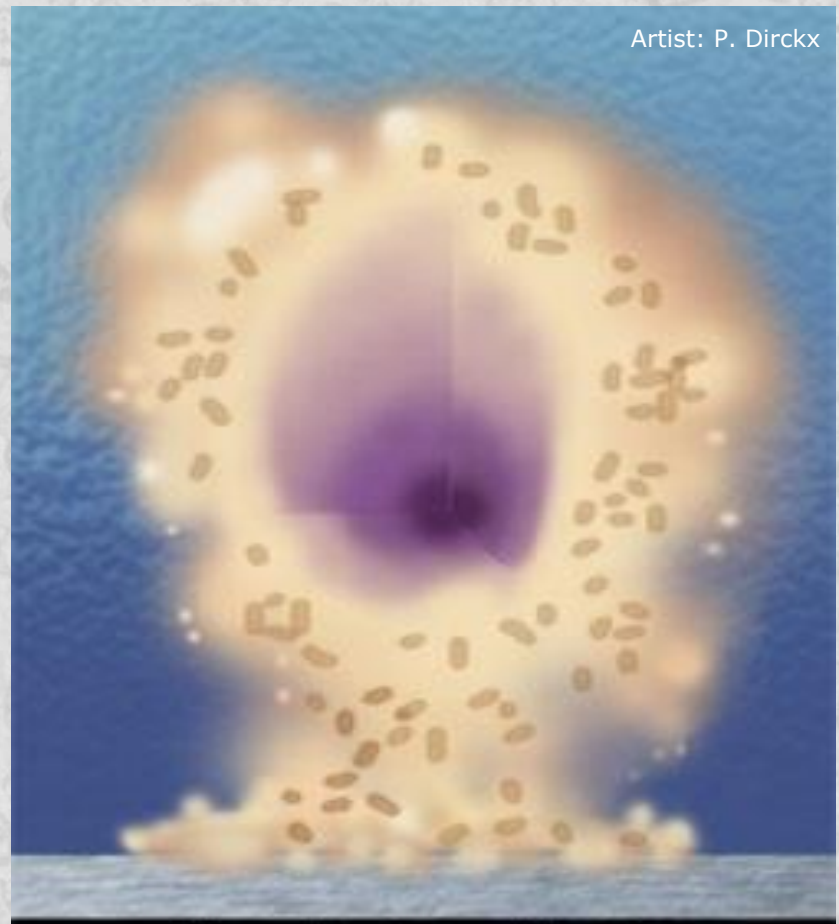
Klayman BJ, Volden PA, Stewart PS, Camper AK, "Escherichia coli O157:H7 requires colonizing partner to adhere and persist in a capillary flow cell," *Environ Sci Technol* 2009; 43(6):2105-2111

Ben Klayman

Artist: P. Dirckx

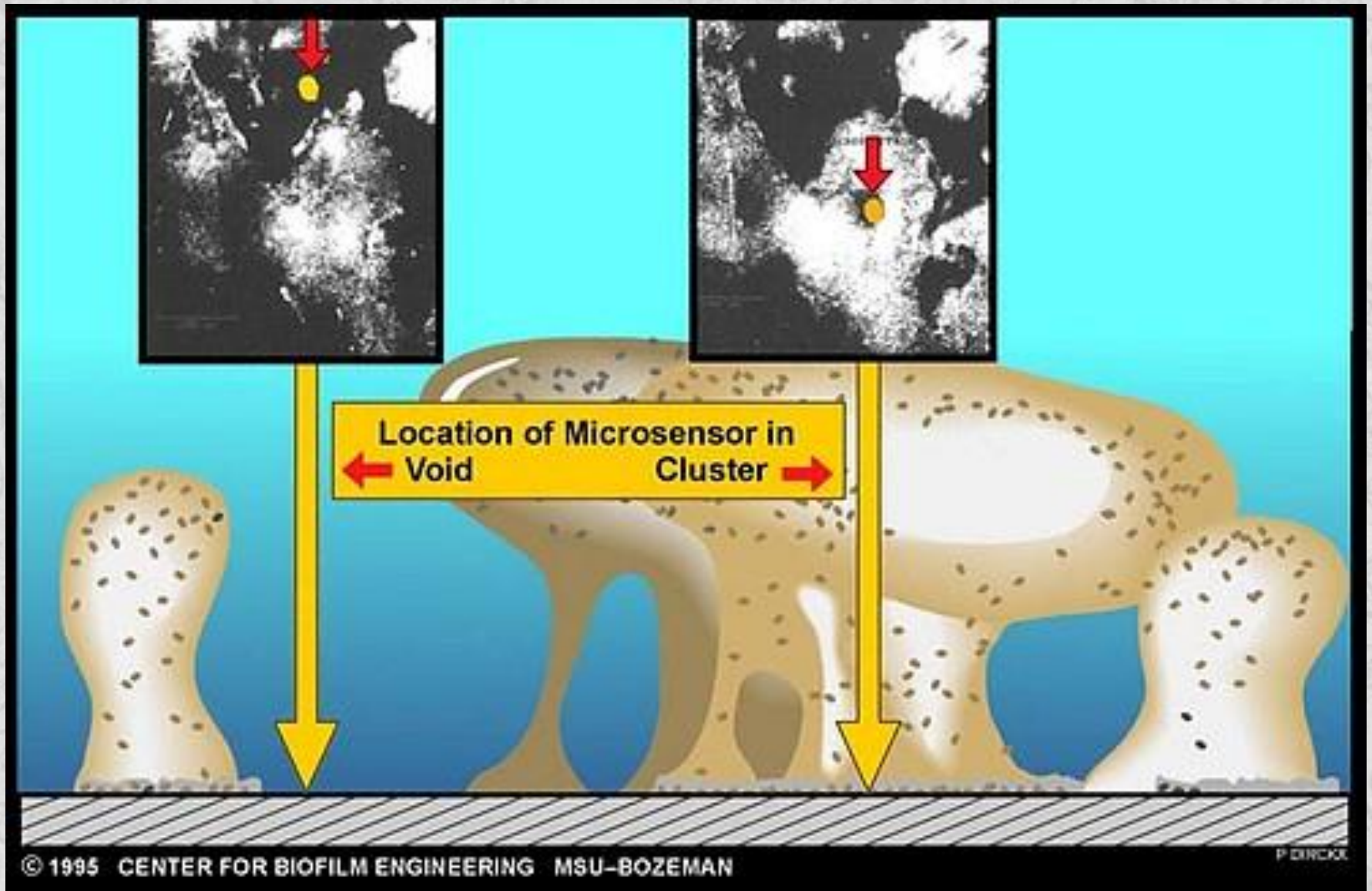


**Bacteria reproduce
and form
microcolonies**

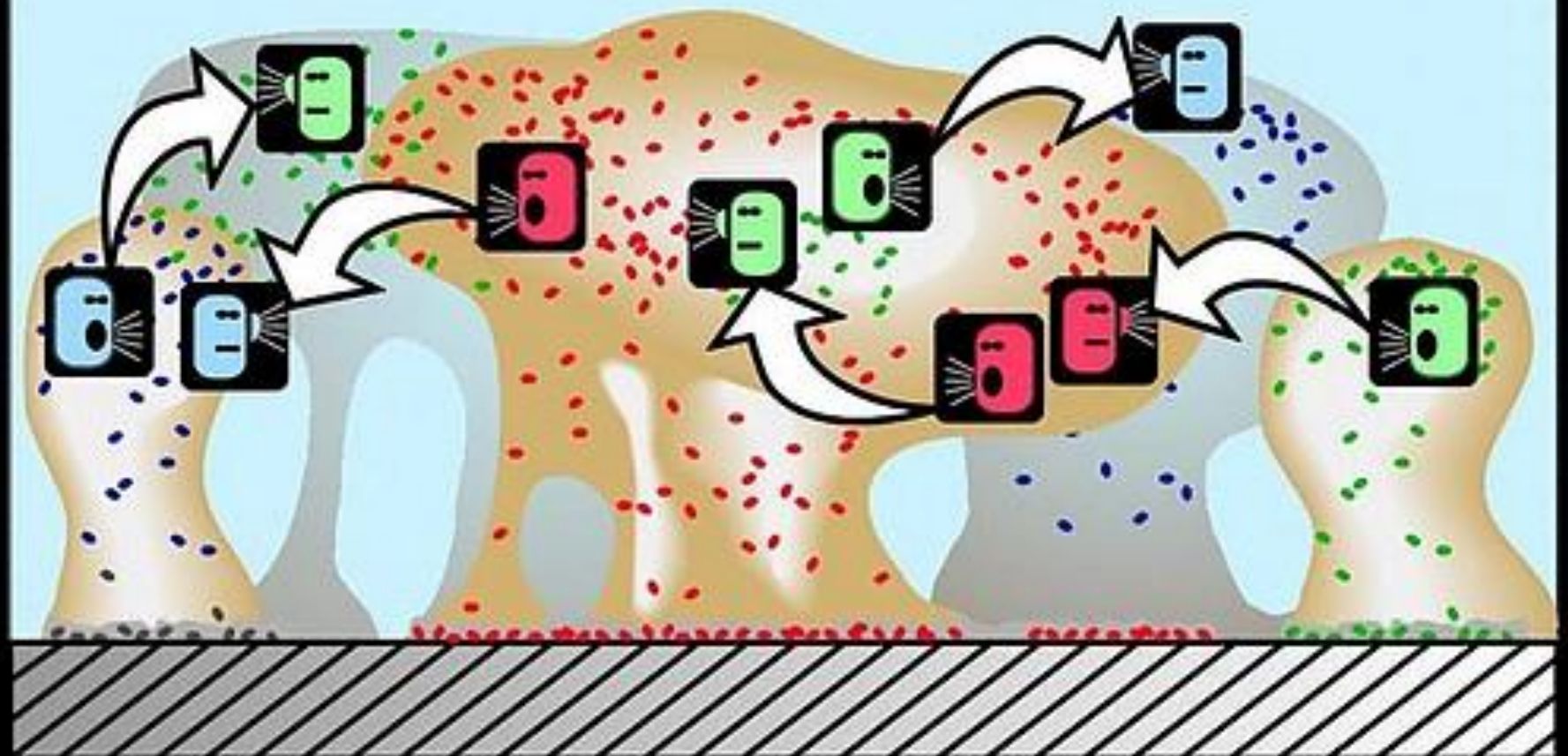


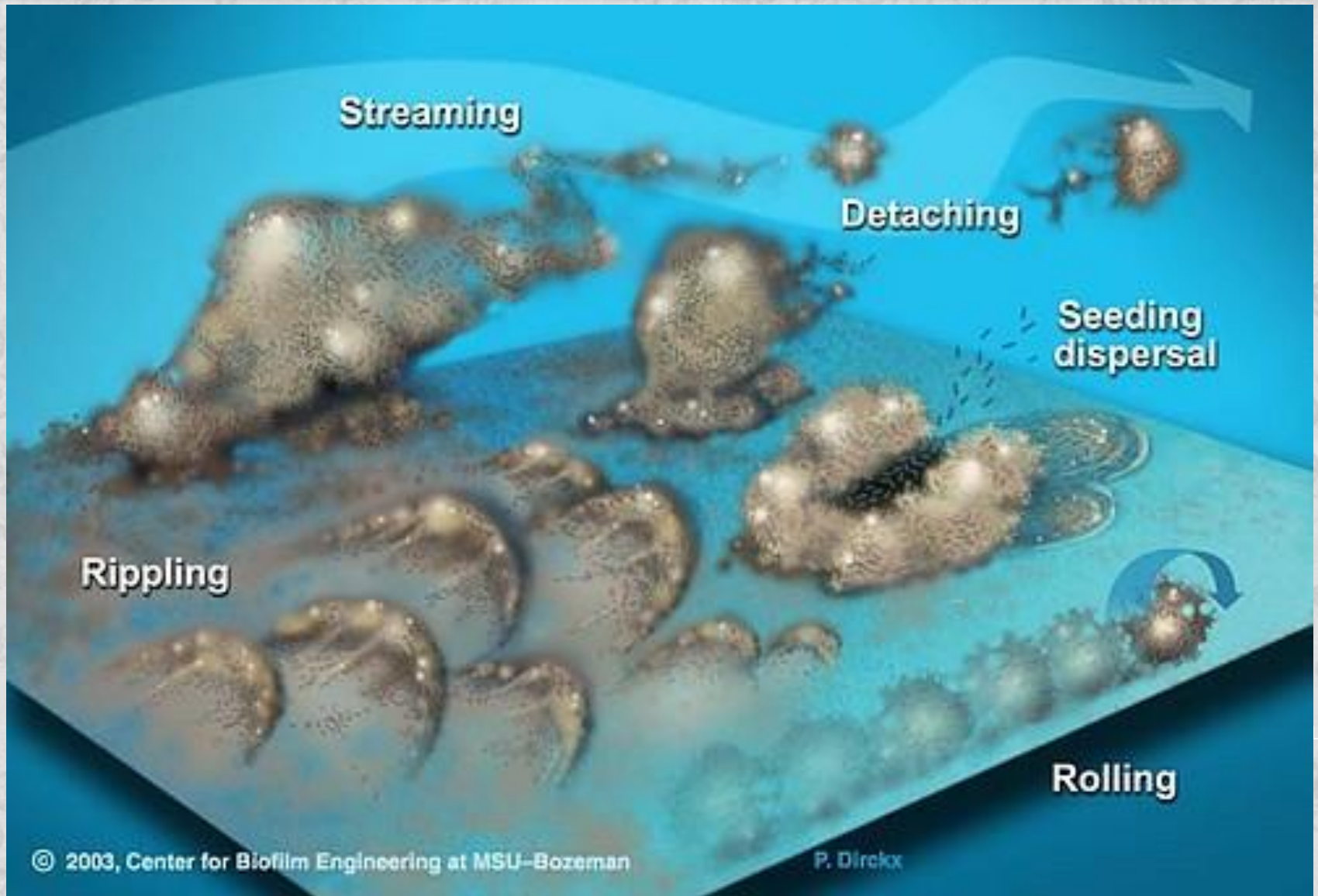
**Chemical gradients
are established**

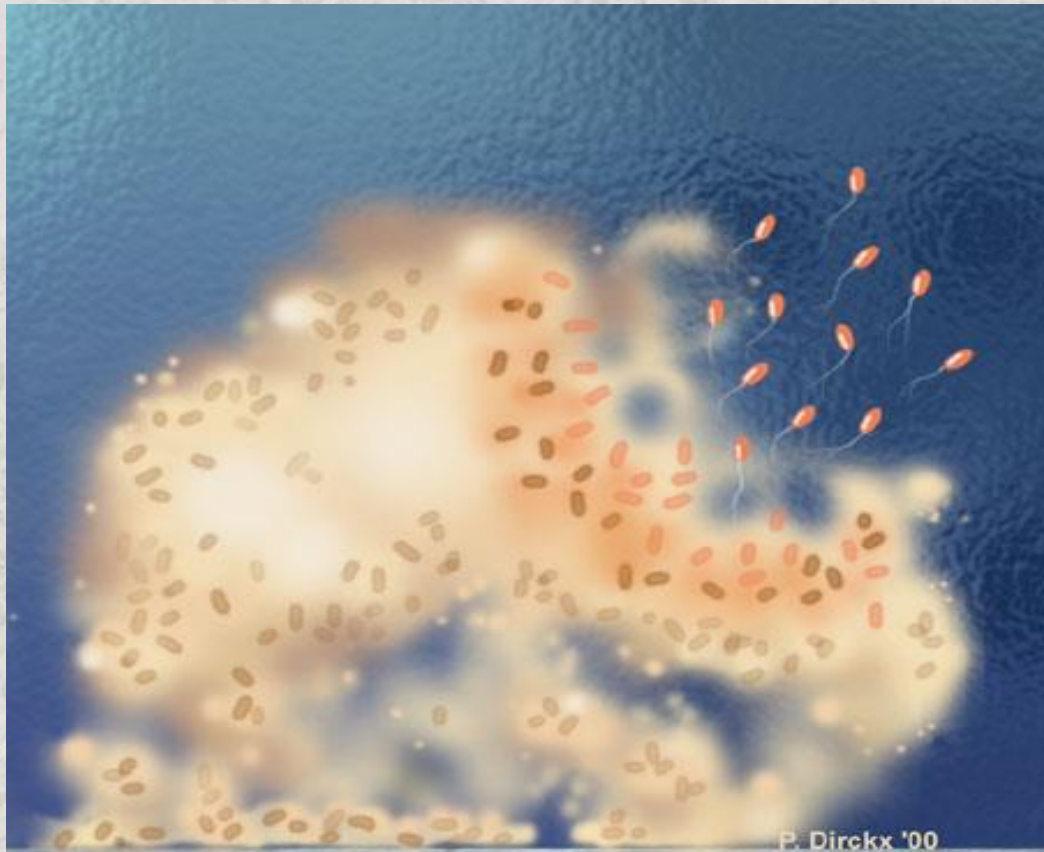
SBML



Cell-Cell Communication

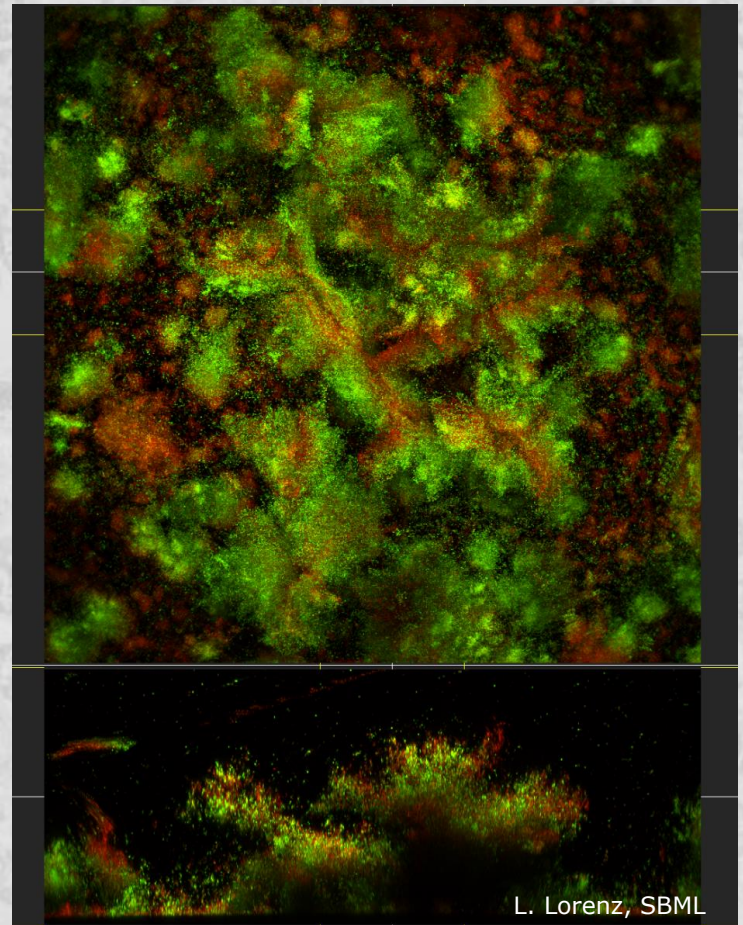






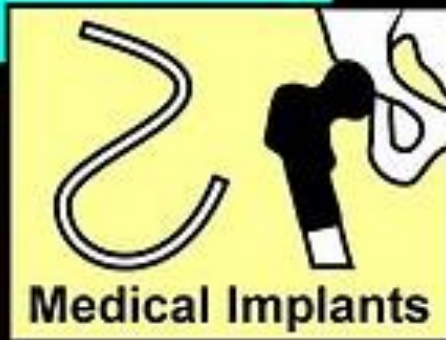
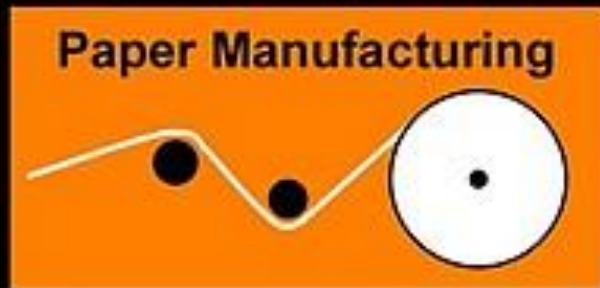
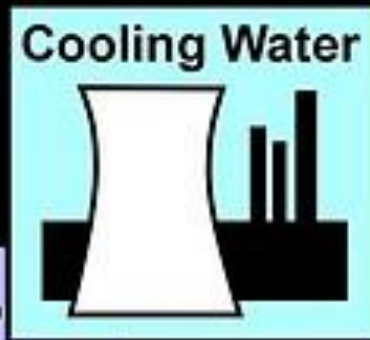
P. Dirckx '00

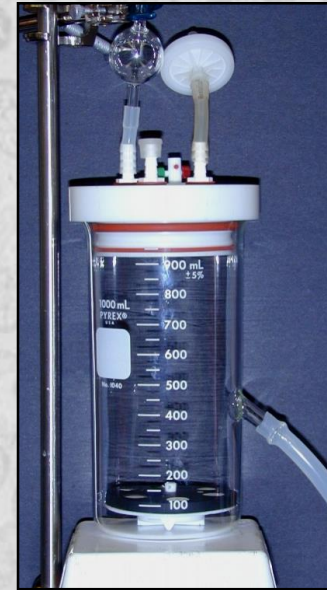
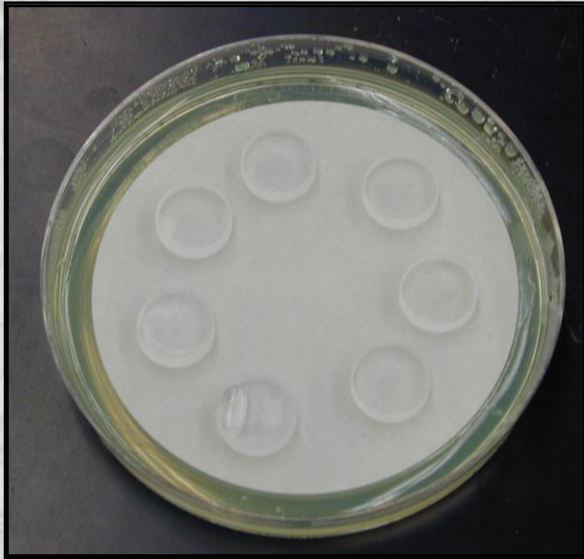
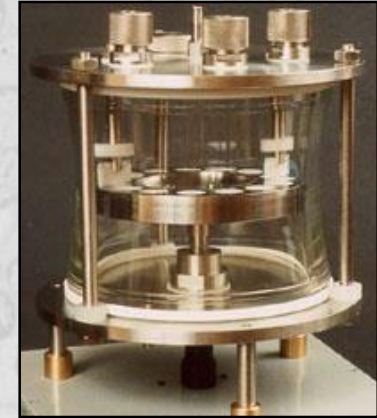
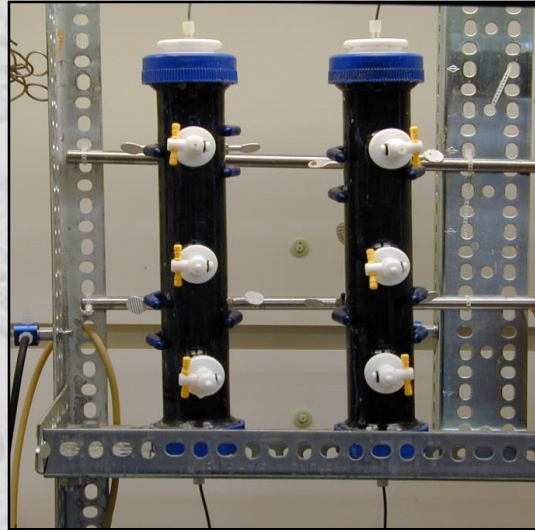
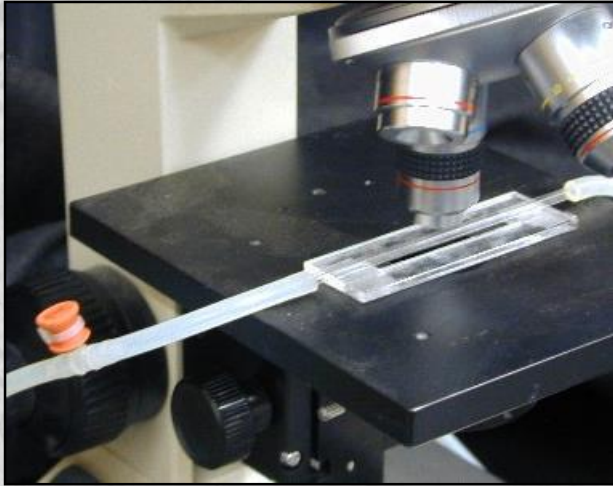
**Cells dissolve matrix
and are released**



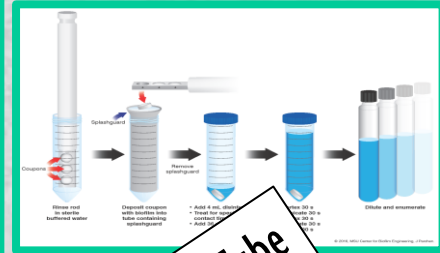
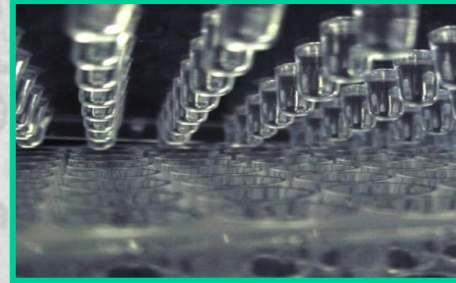
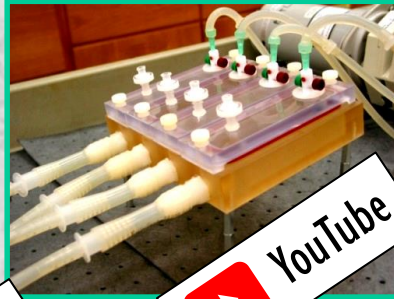
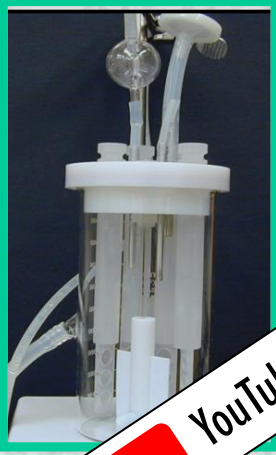
L. Lorenz, SBML

Biofilms Impact . . .





ASTM Biofilm Methods



2002
Rotating
Disk
Reactor
E2196

2007
CDC
Reactor
E2562

2008
Drip
Flow
Reactor
E2647

2011
MBEC
Assay
E2799

2013
Single Tube
Method
E2871

Fluid shear is important

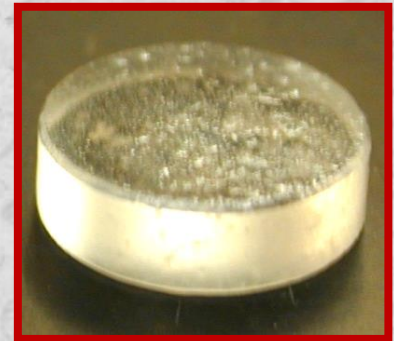
Static Biofilm
No Shear



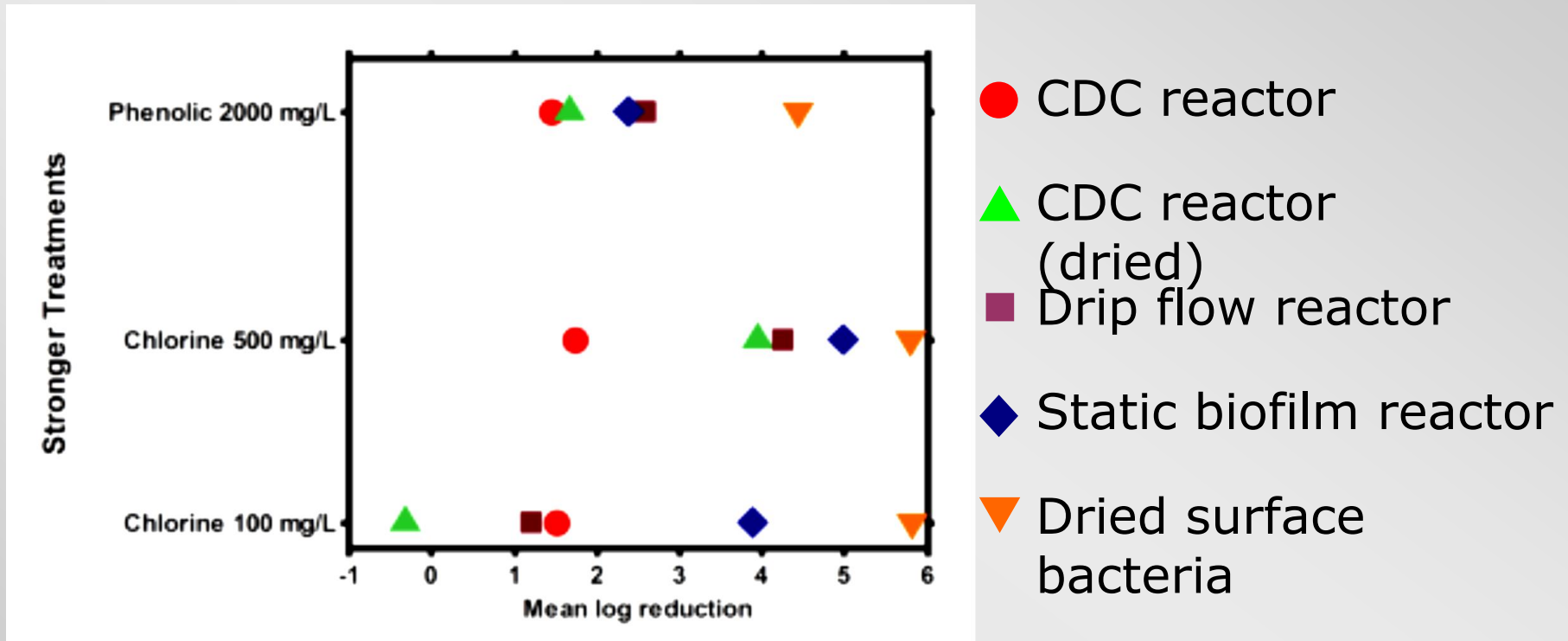
Drip Flow Biofilm
Low Shear



CDC Biofilm
High Shear

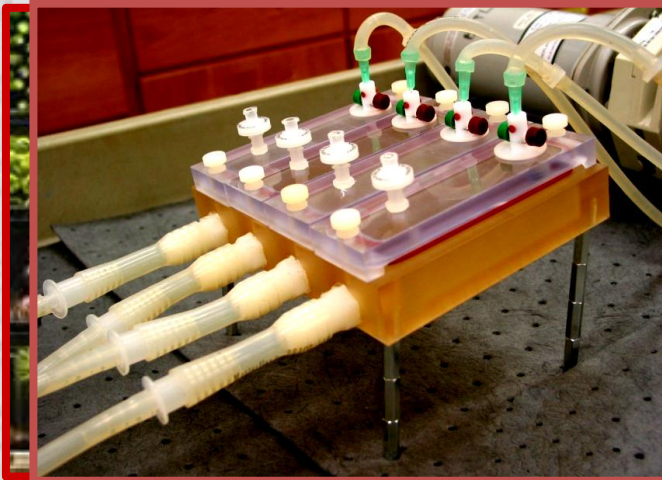
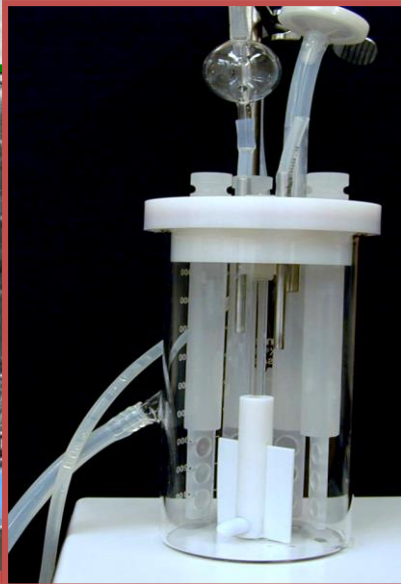


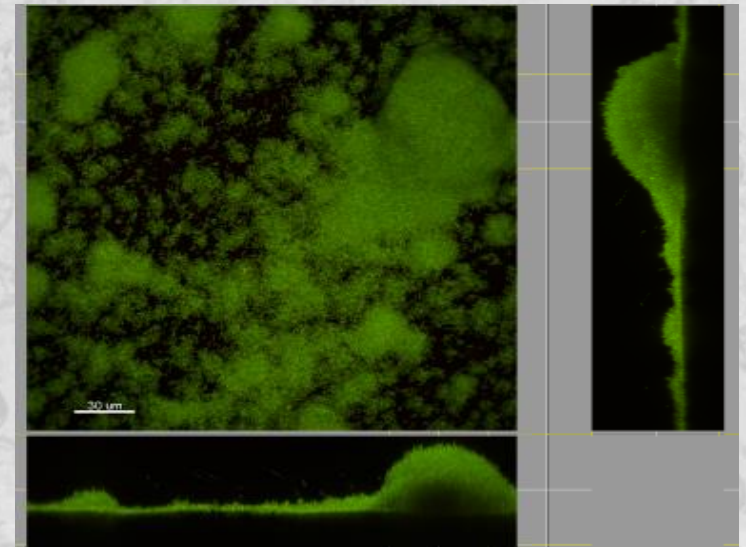
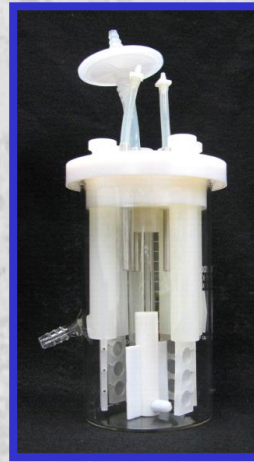
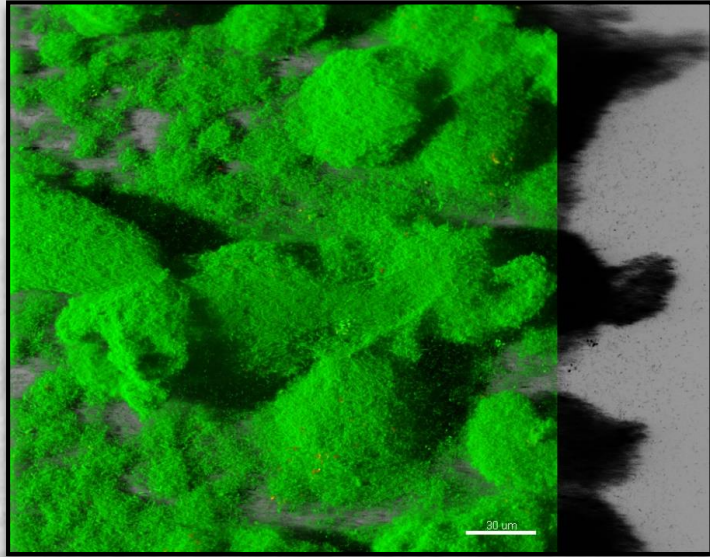
Parallel Reactor Efficacy Study



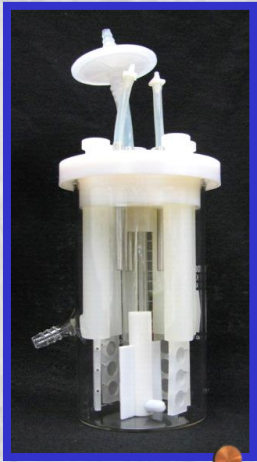
Disinfectant efficacy depends upon how the biofilm was grown





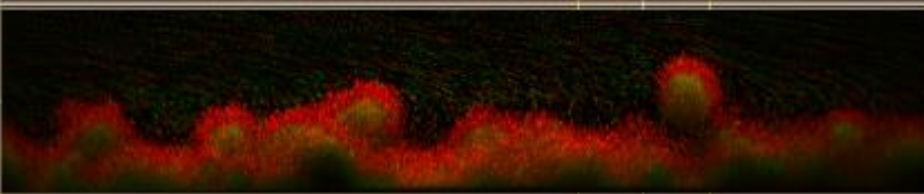
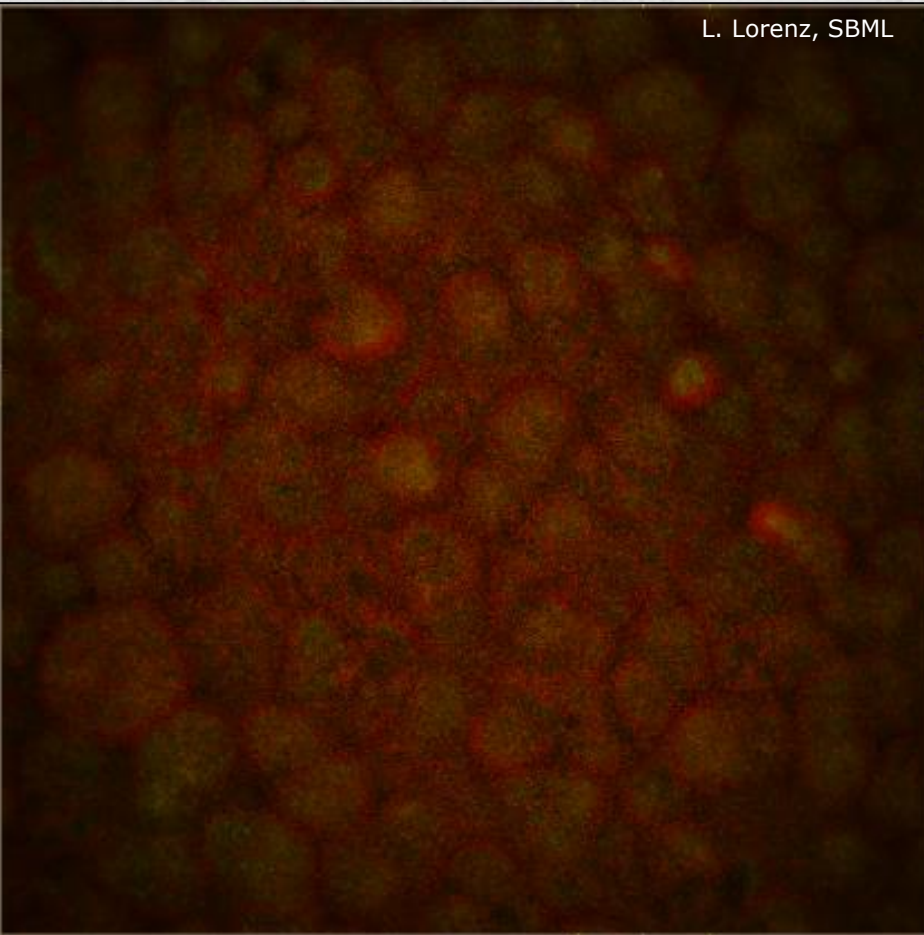


Surface Material	Log Density (\log_{10} CFU/cm ²)
Polycarbonate (left)	8.58
Stainless Steel (right)	7.89
Polypropylene	8.01
Borosilicate Glass	8.23

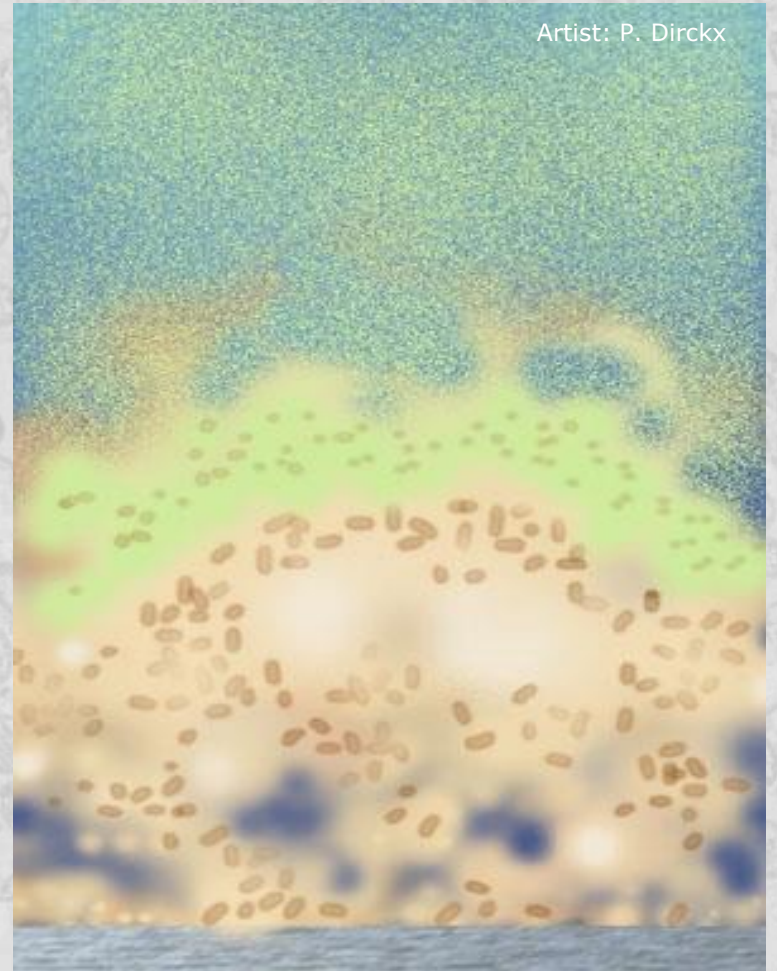


Surface Material	Log Density (\log_{10} CFU/cm ²)
HALAR (ECTFE)	7.40
PEEK	6.78
UHMWPE	7.09
ETFE	7.71
Ceramic	8.50
PVDF	9.25

L. Lorenz, SBML

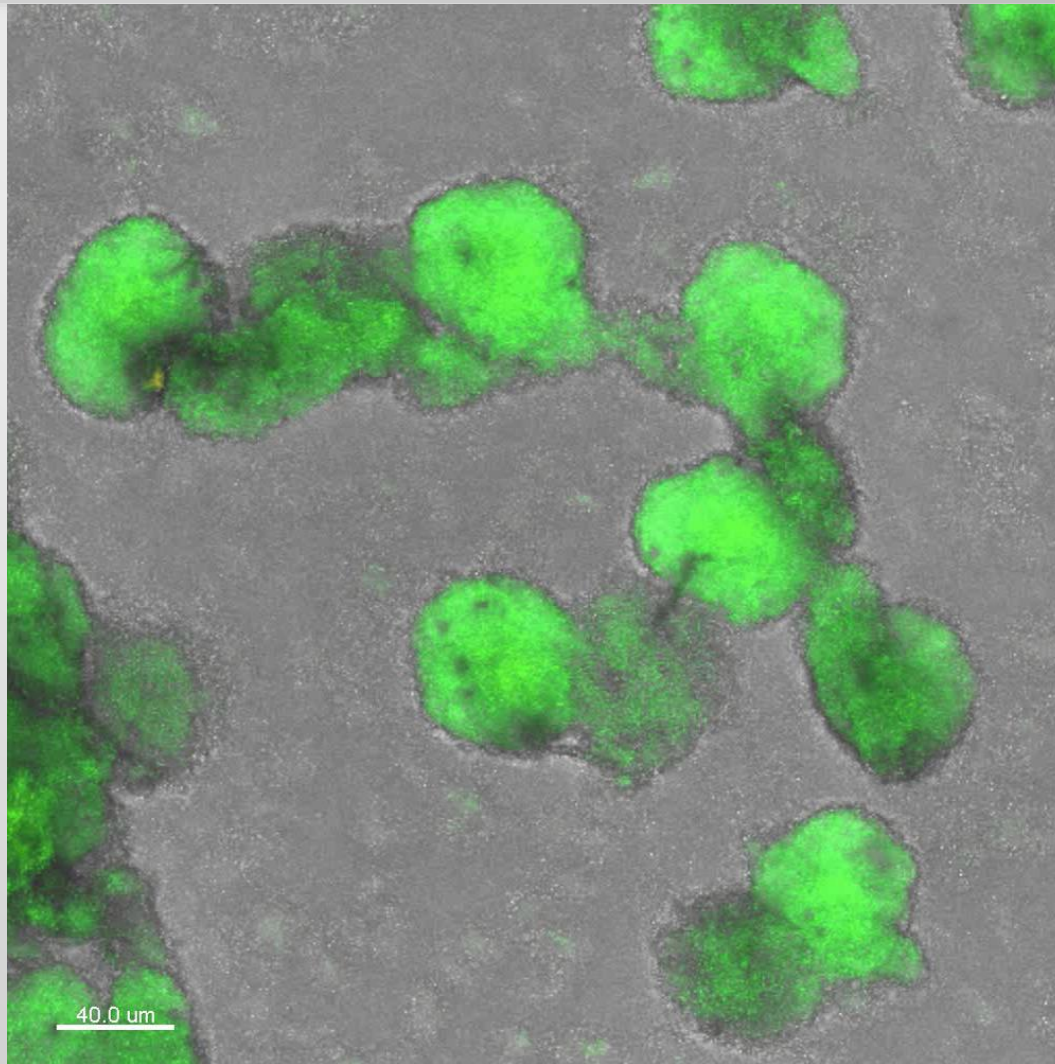


Artist: P. Dirckx

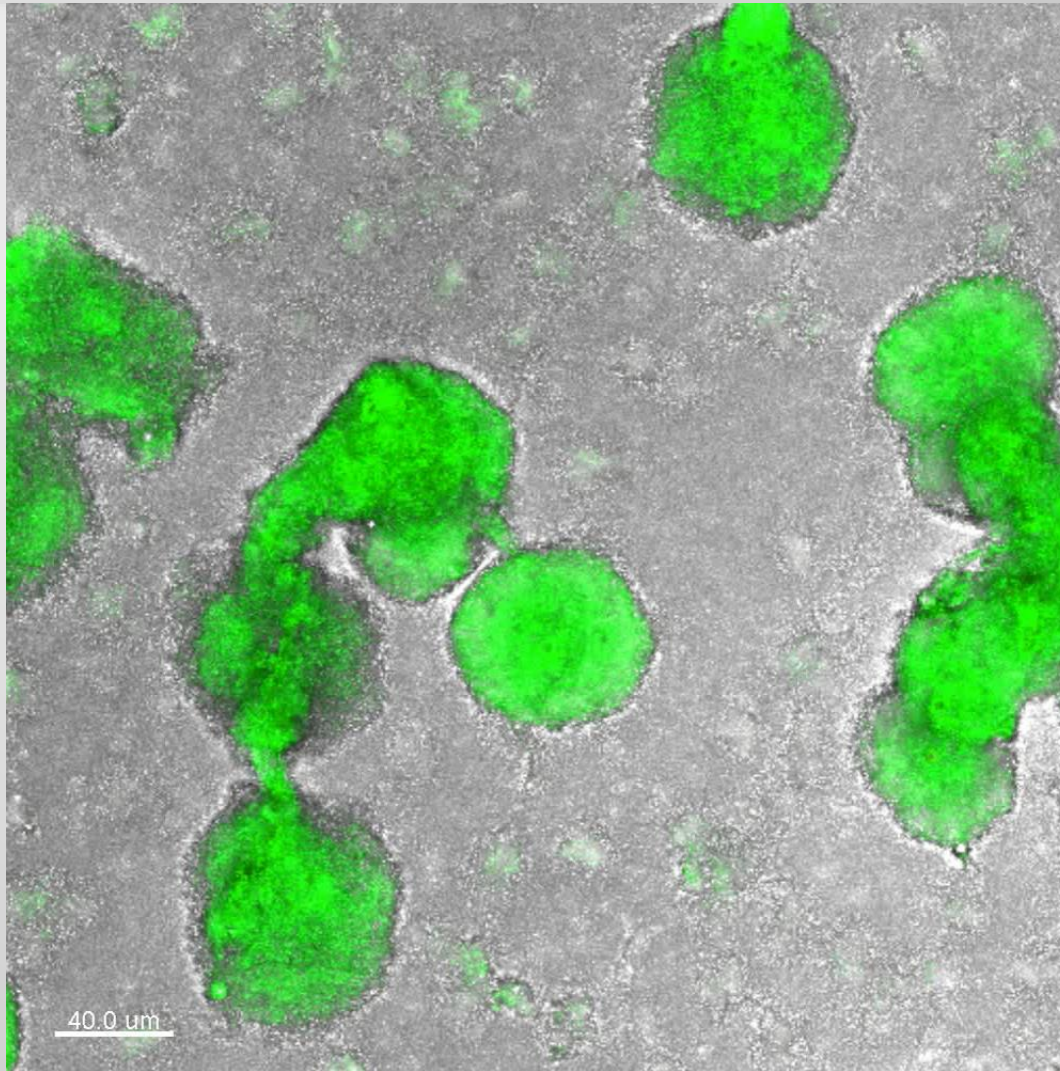


**Biofilm affords
protection from
antimicrobial agents**

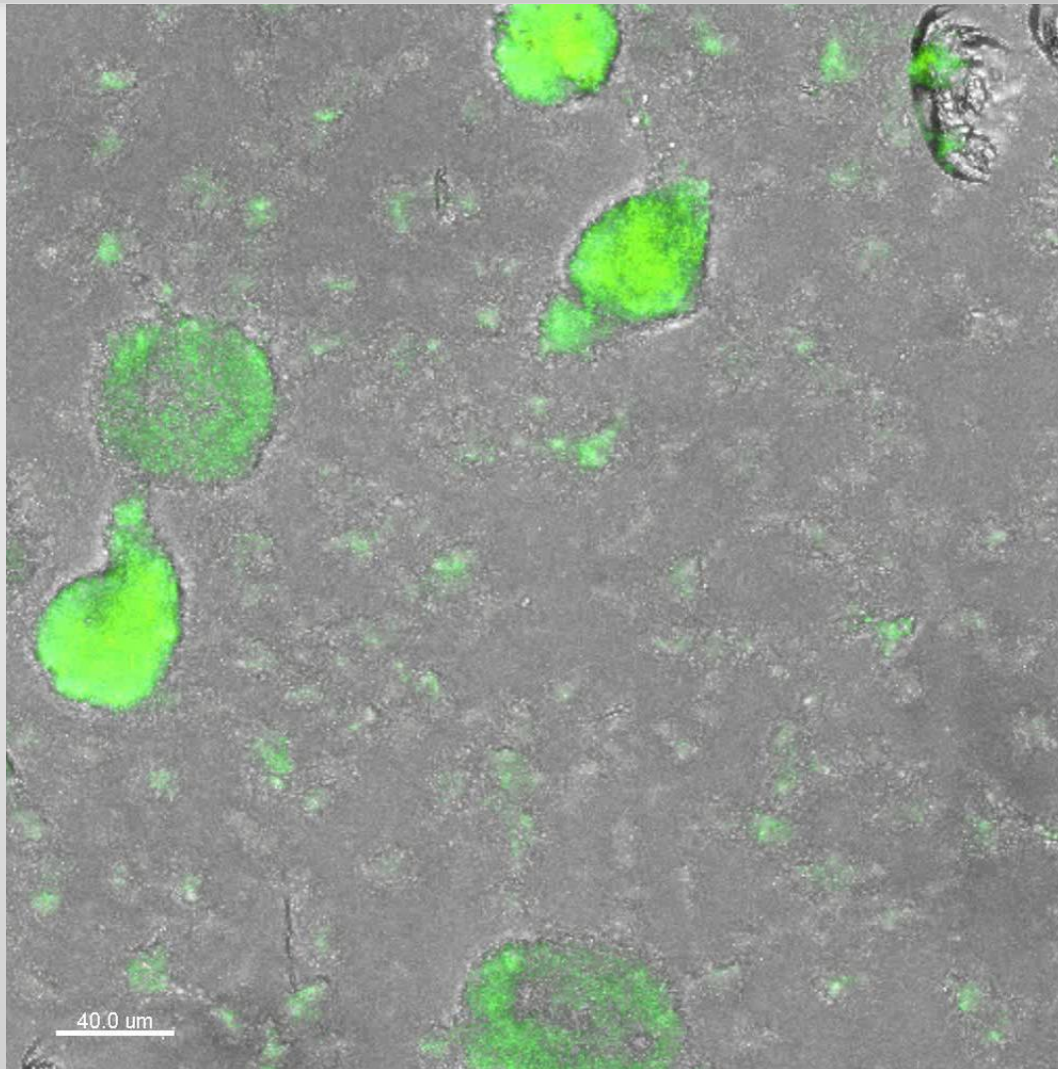
Movie: Alcohol/Quat blend (undiluted)



Movie: Phenolic disinfectant (1:16)

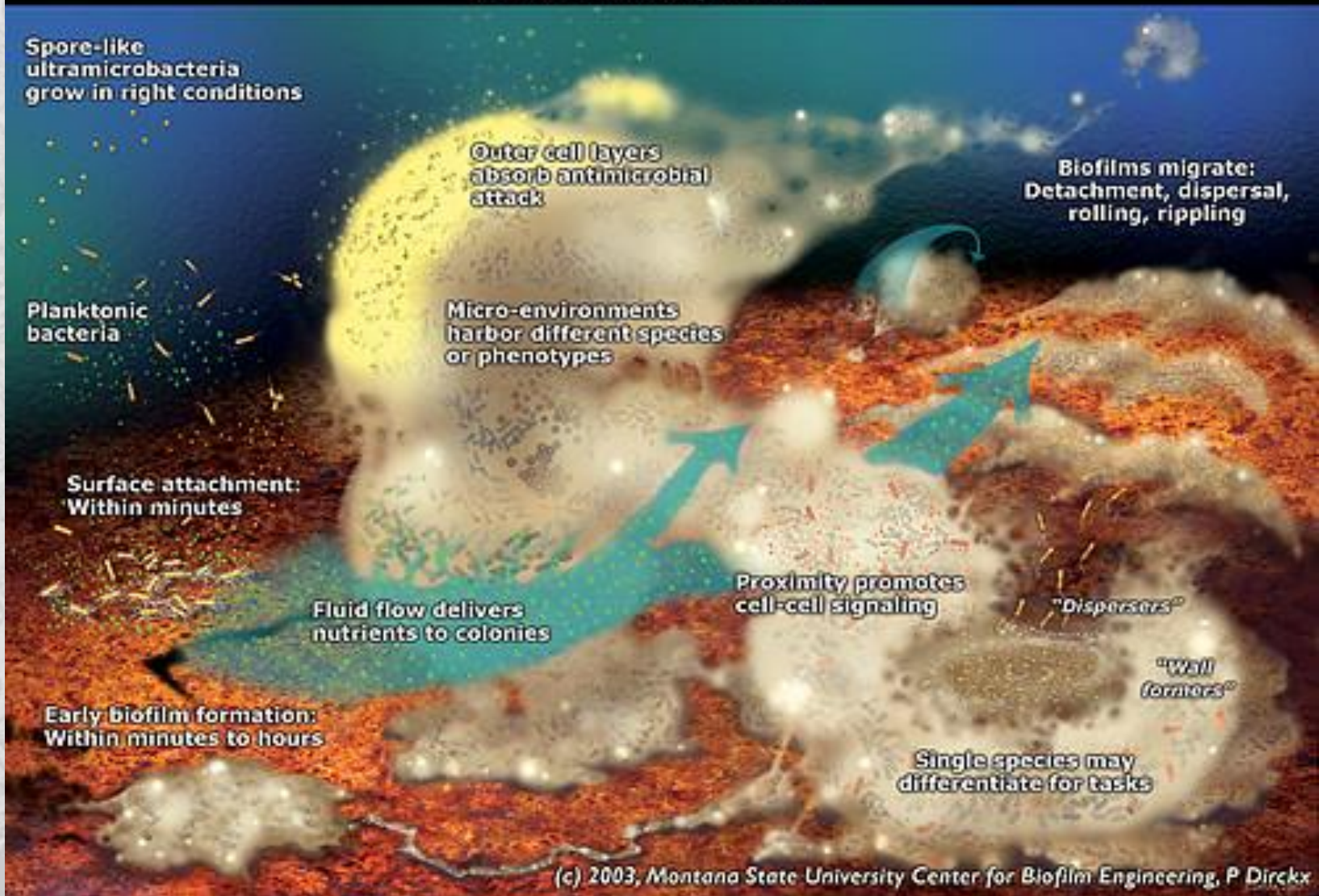


Movie: Chlorine (1:20)



Microbial Biofilms: Sticking Together for Success

Single-celled microbes readily form communities in resilient structures that provide advantages of multicellular organization.



Thank You!



CENTER FOR
BIOFILM
ENGINEERING

biofilm.montana.edu

Standardized Biofilm Methods Lab:

Darla Goeres
Paul Sturman
Al Parker
Kelli Buckingham-Meyer
Lindsey Lorenz (Miller)
Diane Walker

“See” you in Phoenix!



**Processing Water:
Assessing Risk for Biofilms in Food/Dairy Processing:
Why and How?**

**Phyllis B. Posy
President, PosyGlobal**



International Association for
Food Protection,
WEBINAR



PosyGlobal

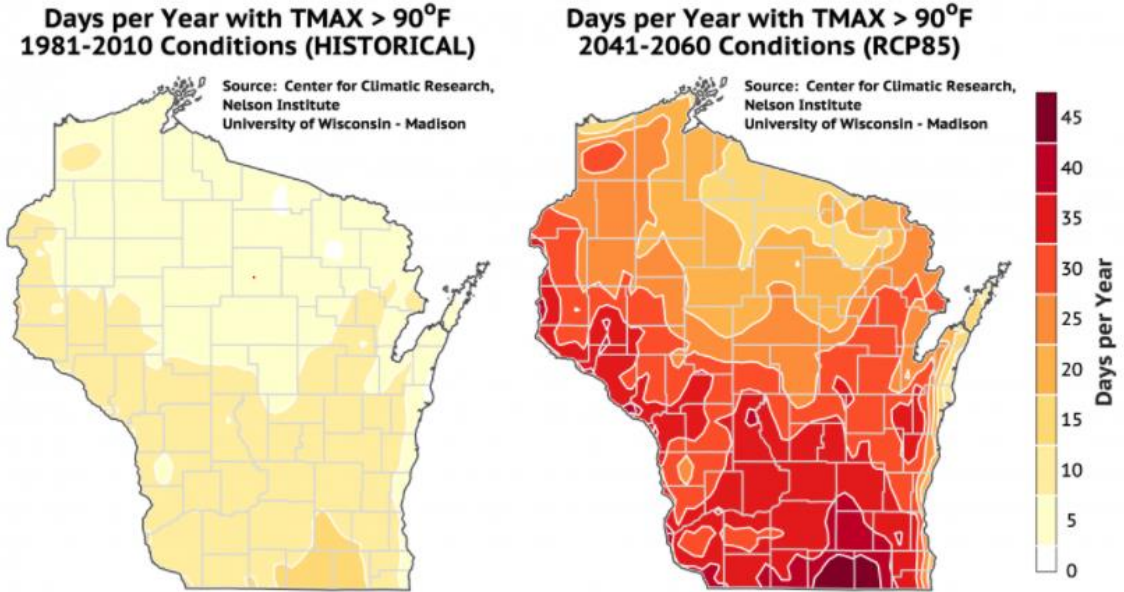
Leveraging 5 decades of experience answering the need for practical ideas that make things work better. Helping dairy companies turn water into a resource.

- **Context: Biofilm RA in a changing world**
- **Step 1: Choose a focus**
- **Step 2: Analyze your water supply**
- **Step 3: Evaluate your vulnerability**
- **Step 4: Develop/Evaluate strategies**



Customer requirements have changed

- Clean
- Different
- Research
- Lots of specific
- Before
- WGS



Graph courtesy of Dan Vimont, director of the Center for Climatic Research

ions”
away!
46 on
ies

Climate Change

- Temperature extremes; precipitation extremes
- More feed contamination
- Somatic cell counts rising require more antibiotics
- Gene transfer promoting resistance to treatments

○ Similarities with Standard RA:

- Aggregate; data-based; action oriented

○ Critical Differences:

- MRA: compliance indicators; bacteria; additive; only amounts above the threshold carry real risk
- BF RA: interactive; cumulative; any amount carries risk
slow operations; spoil product; cause loss

○ Does current system address history or today?

○ Key directional questions: Are you

- Relying on municipal water without further testing?
- Using coliform testing as the key indicator?
- Reducing time dedicated to cleaning/QA?
- Extended operational runs since the last HACCP Plan?
- Updating facilities regularly?

- Context: Biofilm RA in a changing world
- **Step 1: Choose a focus**
- **Step 2: Analyze your water supply**
- **Step 3: Evaluate your vulnerability**
- **Step 4: Develop/Evaluate strategies**

○ Recognize your culture:

- Never have any processing lapses
- A little more chlorine will kill it all off....
- We had some occasional counts... nothing serious
- Climate Change won't affect us
- If it is not completely broken, trying to fix it will make it worse
- Rinsing with lots of water will prevent biofilms

○ Dizzying array of tools

○ Understand your organization

- What would be meaningful?

○ Define a corollary goal

- Intractable even nuisance problem
- Operational optimization
- Reuse water planning

○ Generate support for a serious risk assessment

○ Keep focus on the target and how the RA ties to it!

- Context: Risk Assessment in a changing world
- Step 1: Choose a focus
- **Step 2: Analyze your water supply**
- **Step 3: Develop a Data Plan**
- **Step 4: Make it a habit over the long term**

- **Depend on Municipal /WHO potable water?**
- **Differ with official FDA/FSMA position (as does the EPA)**
- **Can standards designed to make water safe for individuals be applied to food processing facilities?**
 - US rules make no distinction between food manufacturing facilities and a single home: a service connection is a service connection.
 - A PWS serving 10 households and a large food processing/manufacturing facility has the same testing/monitoring requirements as a supplier that serves 11 households and no manufacturing facility.
 - PWS focus on Fecal e coli, will not indicate salmonella, listeria, bacillus or pseudomonads, masking biofilm, spoilage, even potential food safety risk.
 - Sampling, analysis and reporting timing mean that pipes would be inoculated and even product shipped before you can take action.
- **If you depend on external water sources without further testing/tracking you are at high risk for biofilms**



Me
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Navigation

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Between RTCR (Revised Total Coliform Rule) and FSMA: The Hole in the Middle

Wednesday, July 12, 2017: 8:30 AM-10:00 AM

Primary Contact: Ewen Todd
 Organizers: Phyllis Posy, Dorothy Wrigley and Ewen Todd
 Convenor: Phyllis Posy

This session will confront the generally accepted position that if water is good enough to drink, it is good enough to use for food processing. Speakers will analyze the data on whether/how the gap between EPA Drinking Water and FDA FSMA policies leave a hole in the middle that can compromise food safety and dialogue about solution models. The Revised Total Coliform Rule (RTCR; effective 4/2016) refocuses the Safe Drinking Water Act criteria on fecal *Escherichia coli* as the exclusive indicator for compliance. It changes requirements for public notification when samples are positive in favor of triggered assessments ("find and fix"). Users could never know that microbially contaminated water was provided, except from the annual water quality report a year or more later. While impact on individuals drinking might be minimal, food processors, especially small and medium manufacturers relying on municipal water, could be contaminating their pipes and products. Municipal water can be used for processing (food contact or even ingredient water) without any risk assessment because FSMA specifically excluded municipal water users from requirements to address water in their written Food Safety Plans. In reality, not all municipal water is disinfected and public water suppliers must be compliant with treatment requirements only 95% of the time. Recent research in Minnesota and Wisconsin found EPA compliant water, when not disinfected, can be contaminated with Salmonella and other organisms of concern. Viable pathogens can infiltrate, through non-point sources or through the hydraulic action of high service pumps, and go undetected by EPA standard testing. Our current statistics do not capture the link between food outbreaks where the underlying transmission agent or amplification is in the "drinking water". Here, EPA, FDA, and FSIS panelists discuss: How big is the hole in the middle and can it compromise food safety? What should we do about it?

Presentations

- 8:30 AM What is in Drinking Water that Could Matter: The Minnesota Virus Study
Mark Borchardt, US Dairy Forage Research Center, USDA-Agricultural Research Service
- 9:00 AM Do We Only Find What We Are Looking for?
Vincent Hill, Division of Foodborne, Waterborne and Environmental Diseases Centers for Disease Control and Prevention
- 9:30 AM Solutions Panel: Is There an Addressable Gap and What are Options and Models for addressing it?

Moderator: Phyllis Posy

Strategic Services & Regulatory Affairs Atlantium Technologies, Vice Chair Water Safety and Quality PDG

EPA Perspective: Dr. Julie Javier

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■ Timing is not on their side



Wisconsin WAHTER Study

Does Groundwater-borne Illness Risk Meet EPA Standards?

- Acceptable EPA risk for waterborne disease is 1 infection in 10,000 people/year
- Assume every infection leads to an illness, then the acceptable illness rate is 0.0001 illness/person-year
- In the spring of 2006 the WAHTER Study measured 0.44 illness/person-year in children < 5 years old that was attributed to groundwater
- 4,400 times higher than EPA acceptable risk

- Follows basic concept of the WATHER study
 - 74% of the population relies on municipal groundwater
 - 567 PWS do not disinfect; 243 community; 324 NTNC
 - 82 wells in study – 14.7% of the systems
 - All compliant – no e coli (none triggered the GWR)
- Tested for: Human Enteric Viruses Adenovirus Group A –F; **Enterovirus**; **Norovirus GI & GII**; Hepatitis A; Human Polyomavirus; **Rotavirus**; **Salmonella spp.**; **Campylobacter jejuni**; **Enterohemorrhagic E. coli (EHEC)**; Bovine *Bacteroides*; M3 *Bacteroides*-like; Bovine polyomavirus; Pepper mild mottle virus; Total coliforms and *E. coli*
- **66% wells positive for a target; 20% positive for salmonella;**
- **Of those, 60% TCR positive but NONE positive for e coli**



Overall compliance gaps triggered April 2021 Compliance Advisory

- FY2020: 34% violated at least 1 standard
- 7% health violations (21 million people)
- Reporting violation are “cheaper” than a hit
- Many violations under the radar: 5% rule

Survey of Compliance in 2015 in 3 top dairy states

- State Detection Database used; survey of dairies/ farms and processors
- In one state, over 430 dairy facilities were in areas served by public water systems that had **TCR detections**, did not exceed 5%, so no violations:
 - Detections report pathways
- Second state, of 38 dairies; CCR reports showed half had detections
 - 3 had violations- so three get public notice a year later
- Third state, 54 dairies were listed as “public water systems” from 2012-2015: 18 had one or more hits/and/or violations.
- **Check: <https://echo.epa.gov> ; ewg.org/tapwater**

Where does your water come from?

- Does your water supplier buy finished water or treat it?
- Ground Water or Surface? blend or use only one source?
- How much Defacto Reuse?
- Understand the treatment process!
- Does your system disinfect ? With what? To what level?

Determine what percent of their output you get?

Where on the distribution system are you located

- Are you the end of the line?
- Are there sampling points nearby that will provide information?

Check on your state database for recent detections, violations; check your CCR

- Have been any in the past 18 months? Did you know at the time?
- Determine if there is a realistic way you can stay in the loop so you can – on a timely basis -- evaluate, monitor, verify issues and variability in your specific supply (esp. blending)



Develop a data plan

- Speciate incoming water to create a benchmark
- Check your QA data against FoodTracker (Thank You Cornell!!)

<i>Listeria monocytogenes</i>	14561 isolates
<i>Pseudomonas spp.</i>	3809 isolates
<i>Vibrio parahaemolyticus</i>	160 isolates
<i>Streptococcus spp.</i>	2938 isolates
<i>Streptococcus agalactiae</i> only	1233 isolates
Lactic acid bacteria	453 isolates
<i>Salmonella spp.</i>	14928 isolates
<i>Bacillus spp.</i>	4161 isolates
<i>Paenibacillus spp.</i>	2502 isolates
<i>Klebsiella spp.</i>	634 isolates
Total isolates	88599 isolates

Counted at 6/6/2021 3:56 AM Eastern Standard Time.

- Speciate in Zones 3&4 before a vacation or break;
- Generic gram neg?

- **QA: MAP Micro trail for the last 2 years**
 - Incoming water variability
 - Anomalies can mean persistence
- **MAP Maintenance – repeats/preventive -2 years**
 - Cooling/heating systems: pinhole leaks, joint fixes
- **MAP Engineering –go back 2 years; ahead 2**

“Normal changes” can be opportunities

 - Deadlegs often ignored in the rush to end
- **MAP Utilities: Water use volume; variability**
 - CIP the most constant, process critical
 - By product/by time of day/by season
 - Flowmeter, records point to gaps
 - Power too!

- **Consistency: cleaners & sanitizers evaluated/changed?**
 - Chemicals may remove biological fouling
 - Enzymatic cleaners better for residual elements
 - Frequent changes will control biofilms more than consistency
- **Any inherent risks in your specific products or process?**
 - Storing water(fiberglass, temperature, with/without residual) vents?
 - Ct time and pipe flow rates- will the chemicals really work?
 - Pipe materials- some materials have natural pits
 - Disinfection by products
- **Have you reviewed your system for:**
 - Cross-contamination possibilities: sequential use for diff products
 - Water used in the packaging process
 - Any special relevant incidents or factors?

- **In process contamination/cross contamination/backflow**
 - Power anomalies
 - GAC and other filters? optimum niches for microbial growth
 - Untreated water hoses for cleaning?
 - Water Storage tanks? especially without hepa filters; fiberglass in the sun
 - Heat exchangers with pinhole leaks
 - Ice chillers and cooling processes recirculate
 - RO membranes foul; haven for biofilms; watch flux clean earlier
- **Consider how dynamic your system is**
 - Frequency of product or process changes?
- **Post process contamination**
 - Thermophilic and psychotolerant spores
 - Flushes for post-pasteurization pipes
 - Rely on sheer forces or volume to do the job? Vary!
 - Any pre-rinse water used in later stages?

- Is chlorine a silver bullet or simply overused?
- In a study by the University of George on resistance of *Listeria* individual cells(planktonic) and biofilms to chlorine, 13 strains of *Listeria* were tested for their resistance to chlorine. The results of the study are as follows:

Bo

The screenshot shows a PubMed search result for the article: "Chlorine resistance of *Listeria monocytogenes* biofilms and relationship to subtype, cell density, and planktonic cell chlorine resistance." The authors are Folsom, JP and Frank, JE. The abstract states: "Strains of *Listeria monocytogenes* vary in their ability to produce biofilms. This research determined if cell density, planktonic chlorine resistance, or subtype are associated with the resistance of *L. monocytogenes* biofilms to chlorine. Thirteen strains of *L. monocytogenes* were selected for this research based on biofilm accumulation on stainless steel and rep-PCR subtyping. These strains were challenged with chlorine to determine the resistance of individual strains of *L. monocytogenes*. Planktonic cells were exposed to 20 to 80 ppm sodium hypochlorite in 20 ppm increments for 5 min in triplicate per replication, and the experiment was replicated three times. The number of tubes with surviving *L. monocytogenes* was recorded for each isolate at each level of chlorine. Biofilms of each strain were grown on stainless steel coupons. The biofilms were exposed 60 ppm of sodium hypochlorite. When in planktonic culture, four strains were able to survive exposure to 40 ppm of chlorine, whereas four strains were able to survive 80 ppm of chlorine in at least one of three tubes. The remaining five strains survived exposure to 60 ppm of chlorine. Biofilms of 11 strains survived exposure to 60 ppm of chlorine." A red circle highlights the sentence: "The number of tubes with surviving *L. monocytogenes* was recorded for each isolate at each level of chlorine."

Biofilms of each strain were grown on stainless steel coupons. The biofilms were exposed 60 ppm of sodium hypochlorite. When in planktonic culture, four strains were able to survive exposure to 40 ppm of chlorine, whereas four strains were able to survive 80 ppm of chlorine in at least one of three tubes. The remaining five strains survived exposure to 60 ppm of chlorine. Biofilms of 11 strains survived exposure to 60 ppm of chlorine.

- **Key technologies come in various sizes/combos**

- **Coordinate with Corollary goal**

- **Chemicals may be useful for specific need/processes**

- Understand what they leave behind, potential impact

- Change regularly to prevent resistance/assure efficacy

- **Chlorine Dioxide/Ozone, other aggressive treatments**

- Consider potential product contact; worker exposure

- **Tools**

- Remember your generator and power supply

- Filtration – new media/ technologies invented daily

- Consider particle size, most resistant microbe

- Investigate synergies; treatment order may be important

UV: Inactivation/ disinfection for water

- Non-validated tech: high risk
- Polychromatic cover broader spectrum, no regrowth
- Reliable flow meter; control;
- Pay careful attention to process limits: flow, water quality
- Computerized monitoring/tracking;
- Coordinate power
- Develop good startup/shut down process

Validate, Verify and Document

- Paper validation; real time verification
- Continual documentation; watch trends carefully



- **Risk is part of Life**
- **Life is not a Lab**
 - Biofilms won't disappear
 - Don't be afraid of new tech
 - Start with peer reviewed lit; Translate your conditions
 - Credible validation: base paper analysis on process limits
 - Be alert to climate, environmental, water variability
- **Achieving Corollary goal will make the RA work**
- **Pay for Service: more realistic cost comparison**
- **Document from the beginning**
 - If it is not documented, it did not happen

Thank you for your attention!

Please address any reactions, comments and questions to:

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PosyGlobal
Regulatory & Compliance Services

REAL-LIFE MYTHS & COMMON MISTAKES

Food Manufacturing Biofilms

Neil Bogart – Executive Area Technical Support Coordinator

JUNE 8, 2021

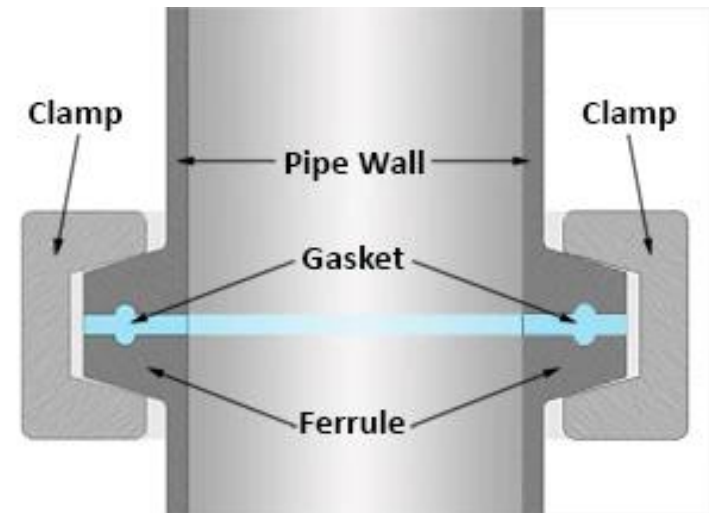
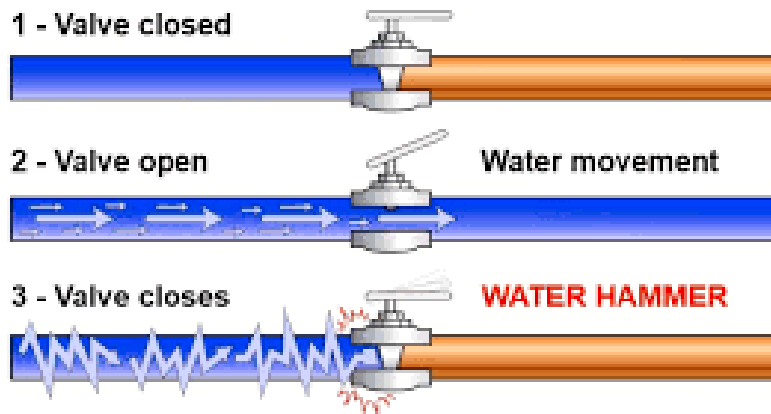


MYTHS & MISTAKES

Water Hammering & Excessive Processing Vibration

My system has hammered for as long as I have been here.

- What is the excessive pressure doing at connections?
 - Could it be creating niches?



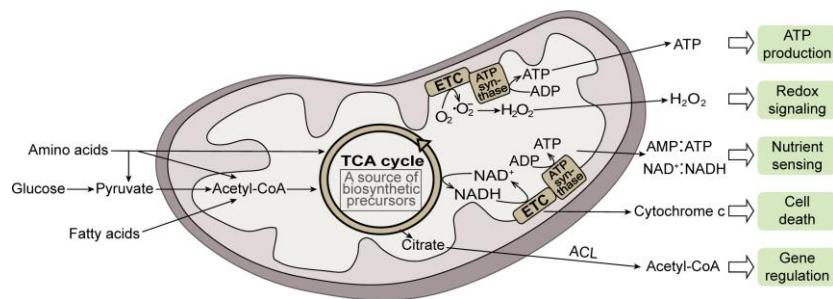
MYTHS & MISTAKES

ATP Verification

I swabbed my equipment after the post rinse and got counts.

Plausible

- Inadequate rinsing times
- Biofilms in processing water
 - PM of water header
 - CIPable water header
 - Water nozzles
 - Flex hoses
- UV inactivates, but ATP can still be picked up

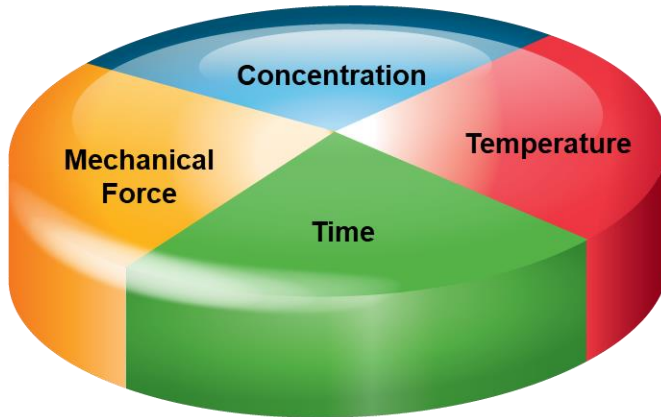


MYTHS & MISTAKES

Mechanical Action - Turbulent Flow

Chemistry alone will remove a biofilm

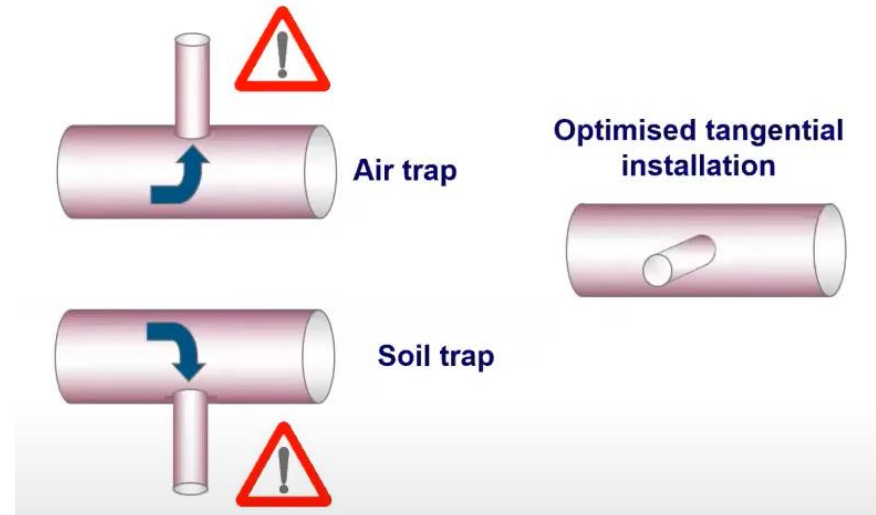
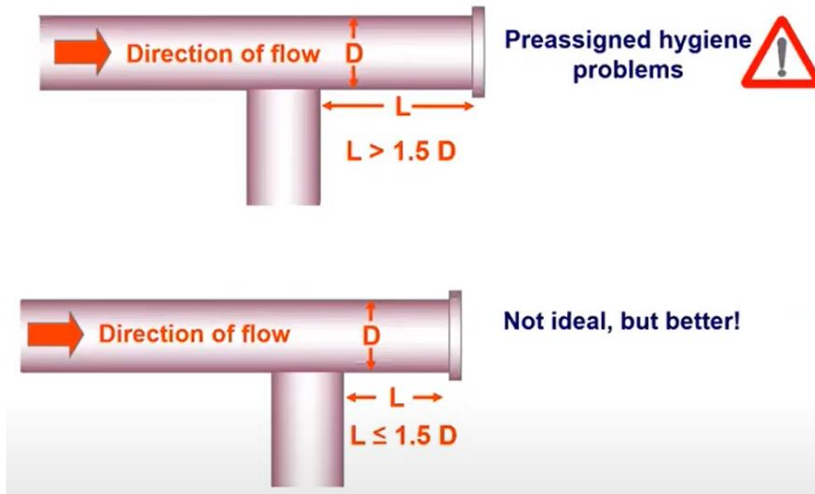
- False: Biofilms can survive saturation with disinfectants.
 - Mechanical action is needed to completely remove a biofilm
- Biofilms attachment to the surface is based off the environment



MYTHS & MISTAKES

Dead Leg Rules

- Any drops or unused portion of any length of piping has the potential for the formation of a biofilm and should be eliminated if possible or have special sanitizing procedures



MYTHS & MISTAKES

Preventative Maintenance Program

Insufficient preventive maintenance

Elastomers

Torque requirements



- PTFE (Teflon)
- Viton
- EPDM
- Buna-N



Typical Applied Torque Settings

Gasket Material	Torque (in-lbs)
Silicone	25-30
EPDM	30-44
PTFE & PTFE Blends	50+ ¹

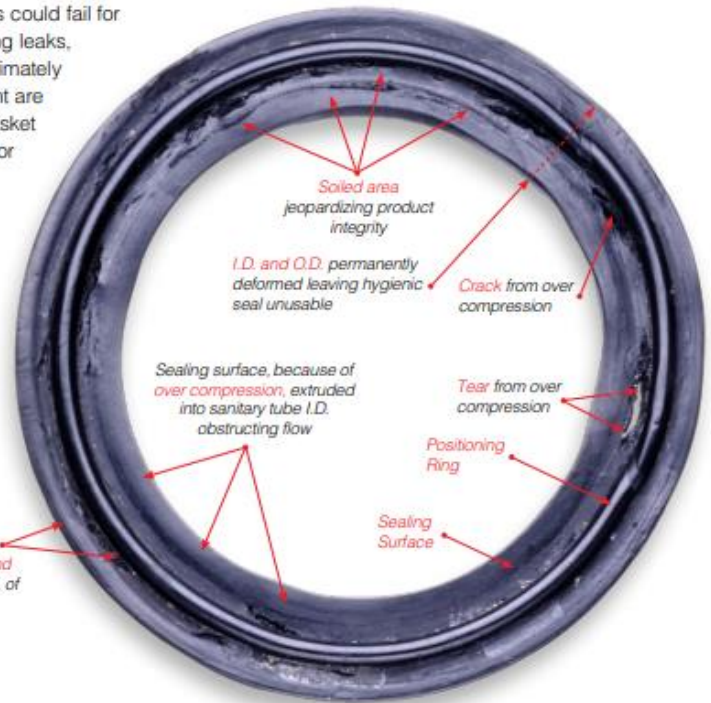
1. Consult clamp manufacturer for maximum allowed torque settings.

MYTHS & MISTAKES

Preventative Maintenance Program



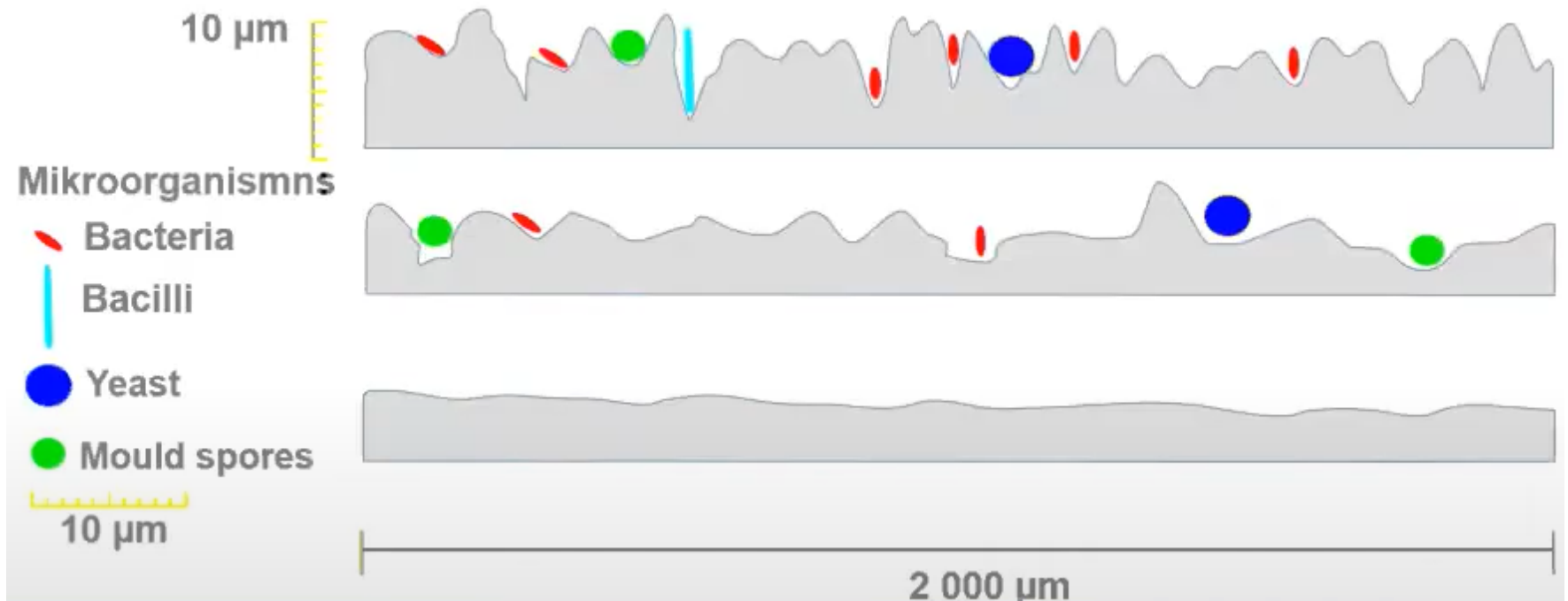
i-Clamp® Gaskets could fail for a number of reasons causing leaks, product entrapment and ultimately product loss. To the right are reasons why a gasket fails and what to look for when inspecting gaskets in your line.



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MYTHS & MISTAKES

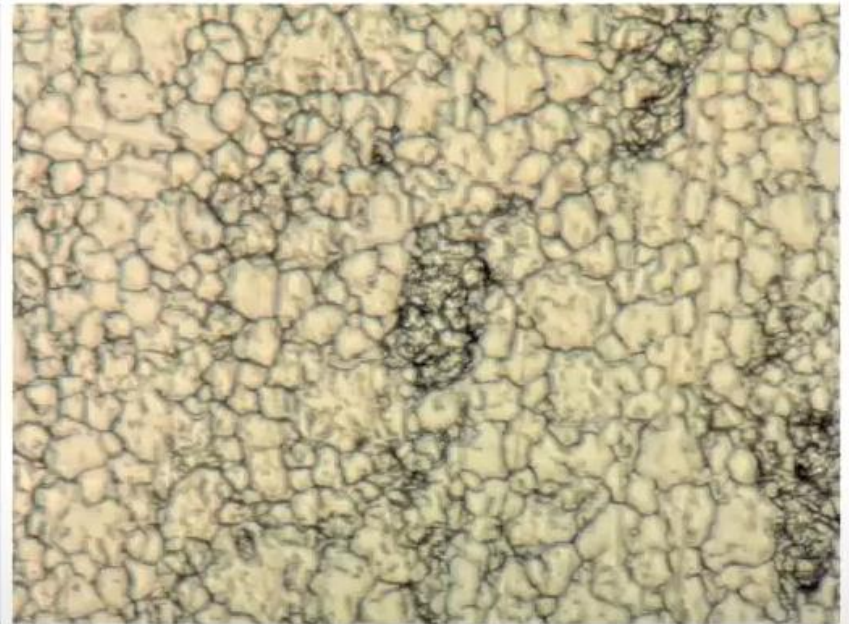
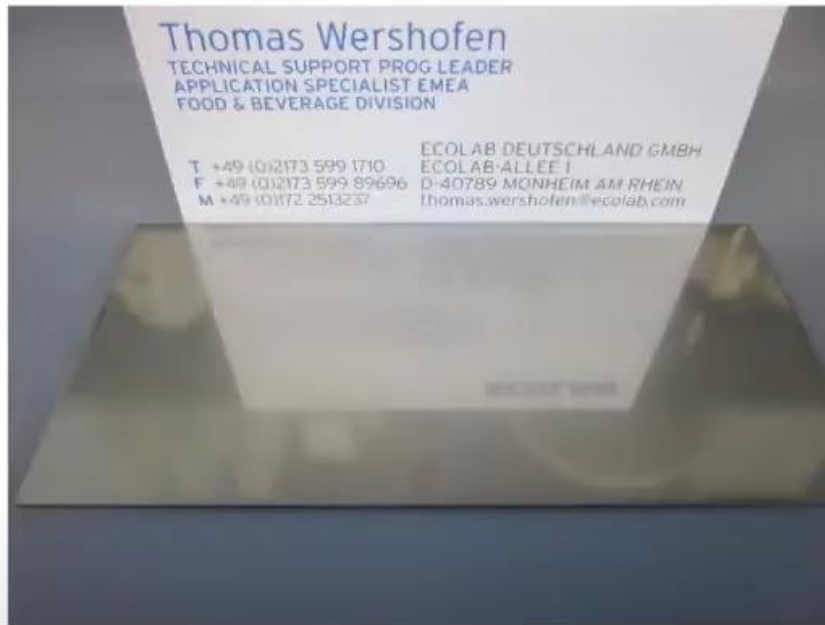
Smooth Surfaces - Stainless Steel Surface



MYTHS & MISTAKES

Smooth Surfaces - Stainless Steel Surface

Photos of a stainless steel surface ($R_z = 0,8$)



MYTHS & MISTAKES

Smooth Surfaces - Stainless Steel Surface

2B Finish

- This is achieved by cold rolling, heat treating and pickling, along with the application of a light rolling at the end in order to achieve a smooth and reflective sheen. Considered the most widely used surface finish, 2B is the basis for most polished and brushed finishes. Most stainless-steel grades 304, 304L and 316L come in a standard 2B finish.

Pit Free Dairy (PFD)

- These types of finishes use an intense polishing method to eliminate any unseen pits, scratches or imperfections. Eliminating these imperfections early could also make the finished product easier to clean without the need to use harsh chemicals to eliminate the bacteria that develops.



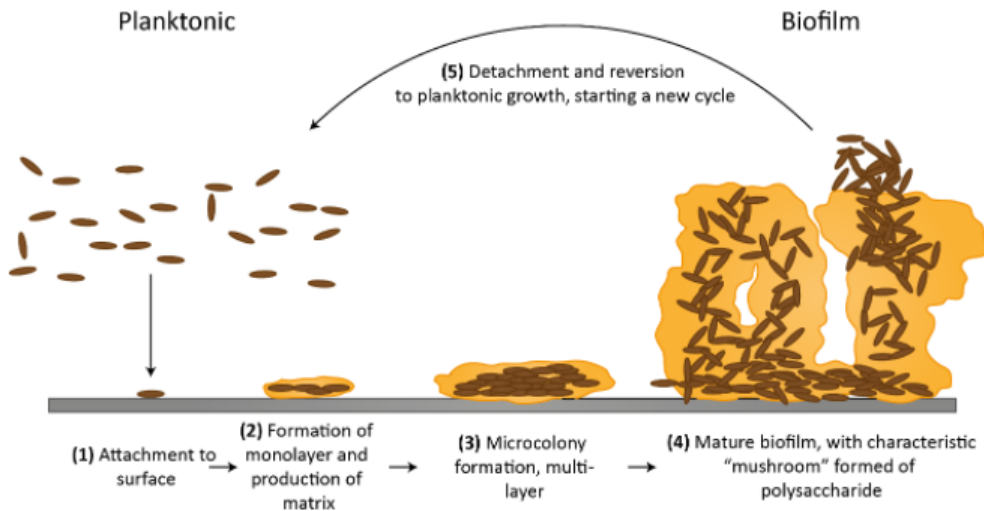
Sourced: <https://www.ryerson.com/resource/the-gauge/three-layers-of-stainless-steel-polish#:~:text=Considered%20the%20most%20widely%20used,in%20a%20standard%202B%20finish.>

MYTHS & MISTAKES

Chemistry & Micro Counts

I washed my system with a procedure that should have killed the biofilm, but my micro counts went up. How could this be?

- Plausible
 - An established biofilm can take numerous washes with the correct 4X4 process to completely remove an established biofilm.



Adapted from Vasudevan, 2014, J Microbiol Exp 1(3): 00014. DOI: 10.15406/jmen.2014.01.00014.

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Questions?

Questions should be submitted to the presenters via the **Questions section** at the right of the screen.



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