ASCALON

THE NEW STANDARD OF CLEAN

Ascalon International, Inc.

Elements of Technology Design

March 19, 2020

VISION

George Bernard Shaw once wrote, "Some people see things as they are and ask why? I dream things that never were and say, why not!"

Robert F. Kennedy, 1968

All our products promote human & animal health – WELL BEING

All our products are only made from plant-derived ingredients – SAFE

All our products have ZERO impact on the environment – SUSTAINABLE

OBJECTIVES

The New Standard of Clean

A Truly Household-Safe Biofilm Remover

A Bottle on Every Countertop

The ASCALON Difference:

PLANT-BASED INGREDIENTS, ECO-FRIENDLY, TOTALLY EFFECTIVE

THE PROBLEM

"Insanity: doing the same thing over and over again and expecting a different result."

Albert Einstein

Bacteria are everywhere and they generate BIOFILM.

- Biofilm protects the bacteria colonies from the outside environment.
- Disinfectants and cleaning products do not penetrate the biofilm.
- The bacteria survive and grow back – faster!
- There is no true Clean.

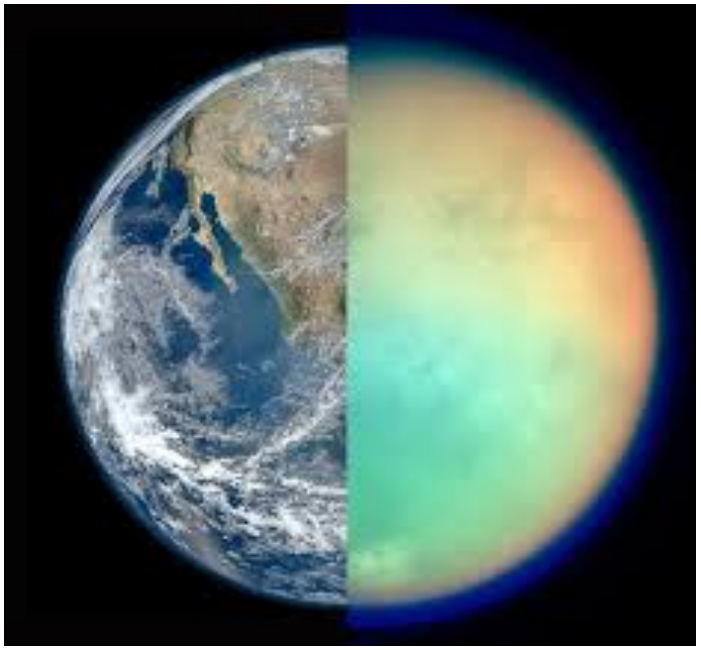
IT'S THE BIOFILM!!!
Get rid of the biofilm, and the surfaces are safe & CLEAN!

CURRENT SOLUTIONS

Disinfectants that kill 99.99% of bacteria leave hundreds of thousands of bacteria behind to reform, secrete new biofilm, and grow larger colonies.

The high rates of infections in US hospitals and food contamination are vivid examples of places where there is extensive cleaning using toxic products, yet people continue to get sick – We Need a Better Tool!

ORIGINS



Bacteria first appeared on Earth about 3.6 billion years ago as single cell organisms.

Bacteria became widespread and flourished for the last 2.5 billion years.

It was the action of bacteria as a blue-green algae, evolving the process of photosynthesis using water as a reducing agent and sunshine, that produced oxygen as a waste product; causing Earth's atmosphere, its waters, and plants to form into what they are today.

- Bacteria over time have adapted to many diverse environments and have given rise to numerous descendant forms.
- Among bacteria's numerous remarkable survival mechanisms are biofilms.
- Biofilms are the slimy inanimate layers formed when bacteria encase themselves in a hydrated matrix of polysaccharide and protein.
- In the primitive stage of Earth's formation, biofilms played a crucial role in bacteria's survival in the extreme heat and acidity using a genetic tactic of random mutation.

EVOLUTION

"Rust never sleeps."
Neil Young

Nomadic planktonic bacteria first attach to a compatible surface.

Then, they exude inanimate extracellular polymeric substances (EPS), which is biofilm, that hold the bacteria together as a community.

Once inside the slimy EPS, bacteria are free to grow, divide, and disperse.

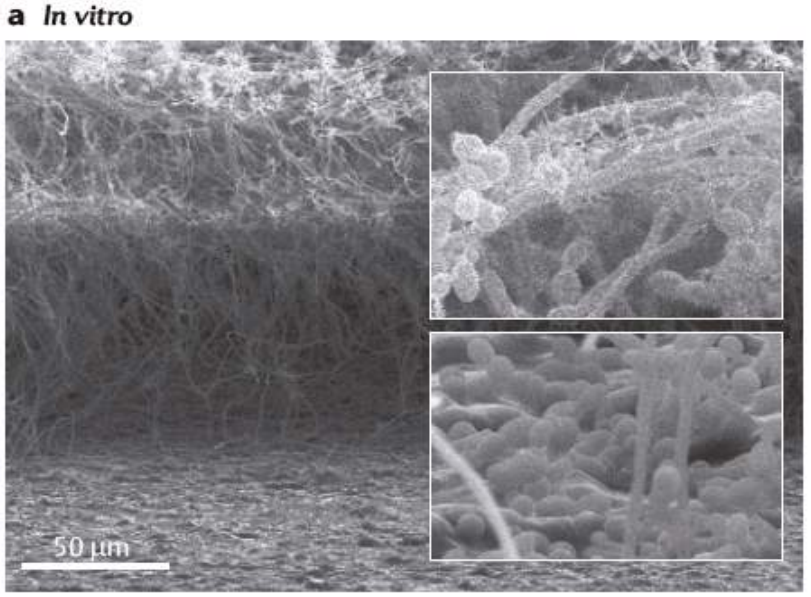
- Within their customized micro-niches, biofilm encased bacteria live in a primitive circulatory system that help it thrive in unfavorable environments.
- One part of the biofilm that aids bacteria's survival is the special channels it forms in which nutrients are generated and circulate.
- A fundamental mechanism behind its remarkable vitality is the adaptive evolutionary changes in biofilm encased bacteria.
- Depending on the environment and the situation, certain bacterial genes are selectively expressed in favor of prolonged survival.

FORMATION

What are Biofilms?

Biofilm is formed by bacteria communities encased in a self-produced matrix. Biofilm formation is a universal bacterial trait by excreting a slimy, glue-like substance.

Biofilms almost always consist of mixtures from many species of bacteria, as well as fungi, virus, algae, yeasts, protozoa, other microorganisms, and debris.



Scanning electron micrograph (SEM) of an *in vitro Candida albicans* biofilm. The biofilm sample was sliced to show three layers in a cross-sectional view.

- Biofilms are composed of DNA, proteins, and polysaccharides that are held together by sugary molecular strands, collectively EPS.
- The living cells produce EPS and are held together by these strands, allowing them to develop a complex three-dimensional meshed matrix of resilient, attached communities.
- Biofilm communities can be as thin as a few cell layers or many inches thick, depending on environmental conditions.

STRUCTURE

EPS Biofilm Structural Integrity

EPS bacterial communities are characterized by:

- Complex community interactions
- Structural heterogeneity = asymmetry
- Genetic diversity
- Extracellular matrix of polymeric substances = EPS

Bacteria can join together on any surface.

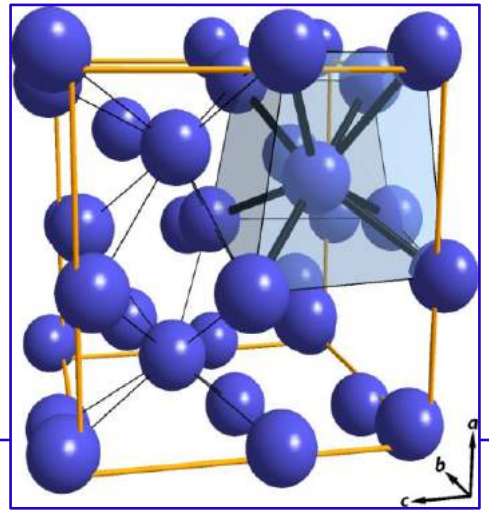
- Then, form a protective matrix around their group

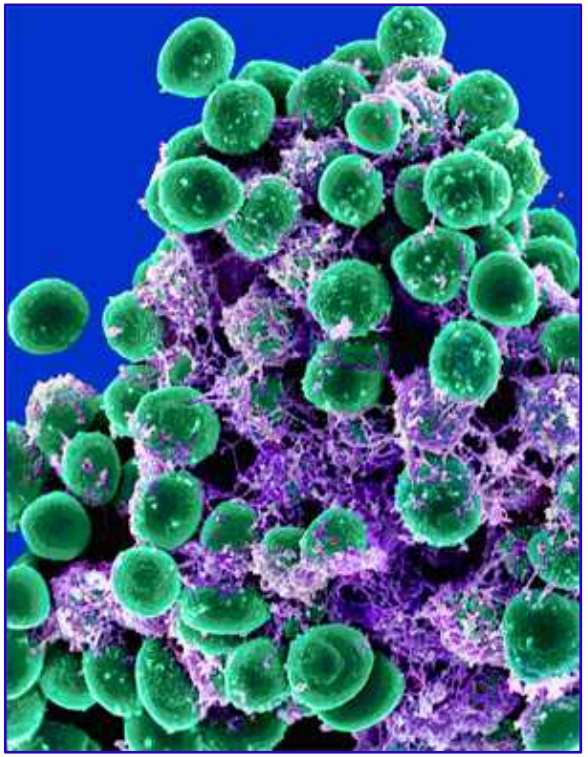
The matrix is a polymeric structure.

- Molecules with repeating structural units that are connected by chemical bonds

EPS communities exhibit capabilities that far exceed the capabilities of individual bacteria:

- Internal neural-like communication network via adaptive shifts in genetic expression
- Collective physical properties
- Diverse behaviors
- Robust survival strategies





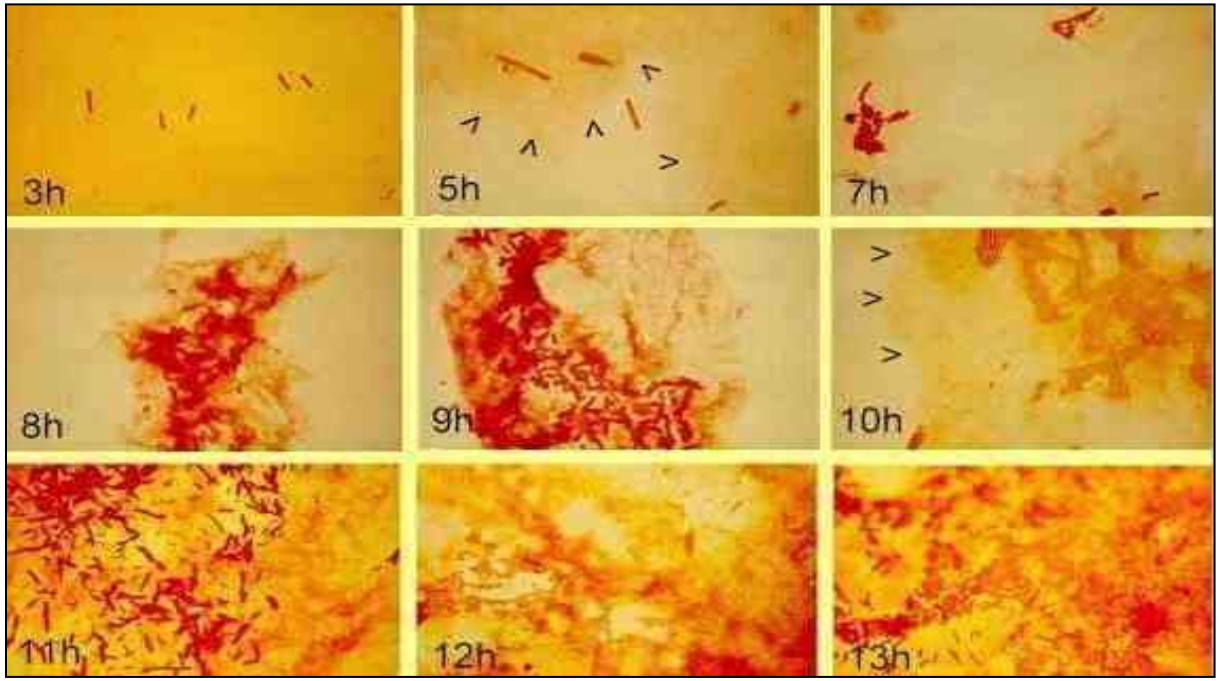
SEM of *Staphylococcus epidermidis* cluster embedded in exopolysaccharide matrix

NECESSITY

"Antimicrobial resistance is one of our most serious health threats."

Dr. Tom Frieden, Director CDC, September 2013

- 2 million people each year are infected by antibiotic-resistant bacteria.
- Even minor infections can be untreatable and deadly.
- The number of overall infections is over 20 million per year.
- Infectious bacteria are everywhere, and rapidly multiply.
- They hitchhike on our hands, shoes, etc; then carried into our homes.

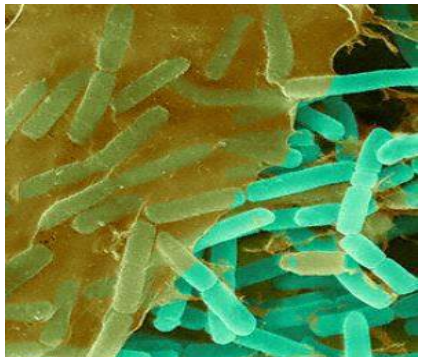


Salmonella bacteria colony time-lapsed biofilm growth cycle

- The solution to the looming bacterial health threat posed by "superbugs" is <u>prevention, not treatment</u>.
- Household disinfectants have very limited ability to penetrate biofilms, thus <u>many of the bacteria survive even after cleaning</u>.

RESISTANCE

"Resistance is futile."
The Borg



SEM of *E. coli* Biofilm Growth

FIVE mechanisms for EPS resistance & tolerance to disinfectants:

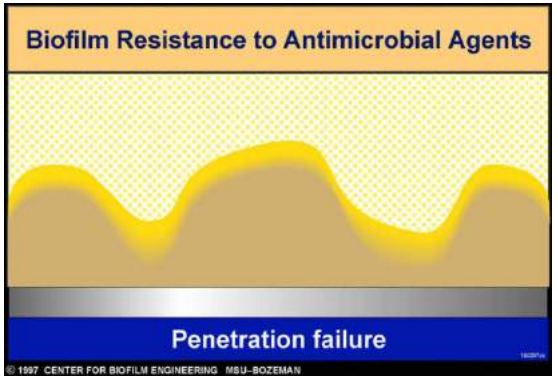
1. Antimicrobial Depletion (the agent does not reach the EPS encased cells)
2. Slow Penetration (giving EPS encased cells a chance to initiate stress responses)
3. Stress Response (cells change activity in response to environmental challenge)
4. Altered Microenvironment (metabolically inactive, though viable cells)
5. "Persister" Cells (spore-like cells that survive an antimicrobial challenge)

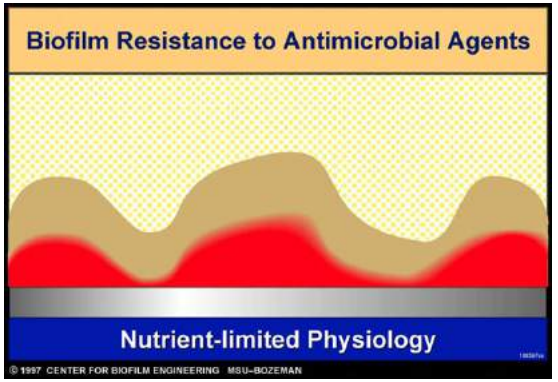
Biofilms construct multicellular protection from disinfectants for its cells. This is caused by shifts in genetic expression when bacterial cells attach to surfaces and begin to form biofilms.

One way for biofilm resistance to disinfectants is penetration failure. The disinfectant doesn't reach the interior portions of the biofilm within a reasonable amount of time. (See 1 above)

Another way for biofilm resistance to disinfectants is a chemical interaction in the outer layers of the biofilm that neutralizes the disinfectant before it can reach bacteria in the inner portions of the biofilm. (See 2 and 3 above)

And another way for biofilm resistance to disinfectants is nutrient-limited physiology. Some disinfectants target metabolically active bacteria. If the bacteria in the inner regions of the biofilm are metabolically slow or inactive, the disinfectant is not effective. (See 4 and 5 above)





TOOLS

"If I had asked people what they wanted, they would have said faster horses."

Henry Ford

Biofilms Enable Problems Caused By Bacteria

More than half of the earth's biomass is composed of biofilm.

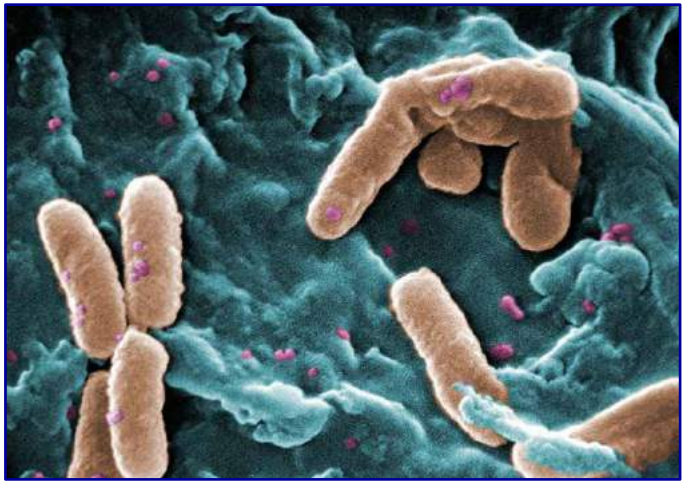
- Over 98% of all bacteria are found in biofilms.
- Biofilms are the dominant communities on planet Earth.

The EPS structure allows bacteria to cooperate and interact.

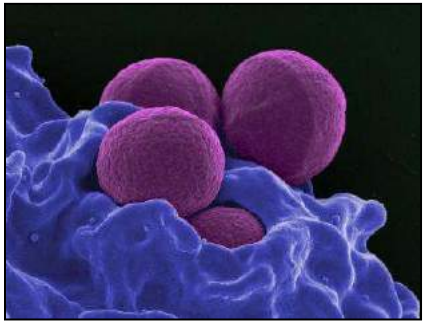
Resistance to disinfectants is caused by the EPS matrix that protects the bacteria community.

NIH reports the cause of over 80% of all infections are linked to EPS biofilm communities.

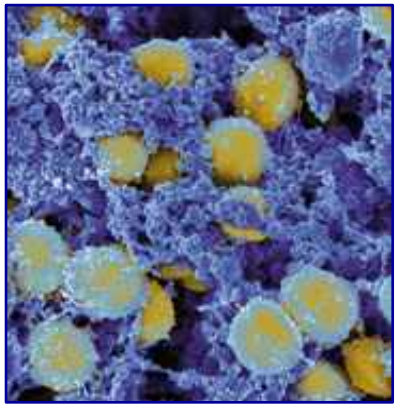
The EPS biofilm matrix allows highly antibiotic resistant microorganisms to be tenaciously bound to surfaces.



SEM of *Pseudomonas aeruginosa* attached to surface by its EPS matrix



SEM of mature methicillin-resistant *Staphylococcus aureus* (MRSA) biofilm embedded in extracellular matrix



EPS Remover

An EPS remover is a liquid that demonstrates sterilant level performance, in conjunction with removing the biofilm, when applied directly to pathogenic microorganisms that have been grown in their biofilm formations for at least 96 hours.

By removing EPS, the harmful pathogenic microorganisms are removed together with their biofilm host.

END PRODUCT

"If you can dream it, you can do it."
Walt Disney

A Biobased and Sustainable Liquid – Made with only food grade, plant-based ingredients.

- Safe for the home = No VOCS, no toxins, no pollutants.
- Safe for children, elders, pets, and for employees.
- Safe for the environment = all-inclusive ecologically favorable properties.

Kills All Odor-causing Bacteria – Proven repeatable results, superior cleaner.

USDA BioPreferred® – 95% Certified Biobased content = *Organic*.

EPA Safer Choice Program – Eco-friendly, biodegradable = *Sustainable*.

Effective Cleaner – Patent-pending formula removes the biofilm and the dirt it holds.

FDA Approved for Food Handling Use – Compliant.

BEFORE *AFTER*

Biofilm Build-Up on Concrete (Porous)

Biofilm Stains Removed From Concrete by Laurinex

Macroscopic Biofilm Demo

Laurinex was applied to this concrete curb and gutter and let sit for one minute.

The surface was lightly brushed, and then rinsed with fresh water.

The surfaces are free of biofilm and clean.

POINT OF DIFFERENCE

2016 BY THE NUMBERS

Food Recalls

USDA = 58M Lbs. – 764 – 22%

100% in 10 Years

$10M Now & $100M Long Term

Foodborne Illnesses

CDC = 48M – 128K – 3K – $55.5B

WHO = 600M – 420K – 125K – 40%

Hospital Acquired Infections

CDC = 722K – 75K – $45B

Adolescent Respiratory & Infections

EWG = 30% to 50% – 17 – $37B

NCHS = 23 Times in 4 Years – 40%

Employee Injuries

EPA = 6 Out of 100

USDA = 13 Out of 100

Toxic Cleaners & Disinfectants

CPA = 150 – 96%

NIH = 267K

THE NEW STANDARD OF CLEAN

SAFER CHOICE
Meets U.S. EPA Safer Product Standards
epa.gov/saferchoice

USDA CERTIFIED BIOBASED PRODUCT
PRODUCT 95%

THE ASCALON EFFECT

CREATED VALUE

- ✓ *Enhanced Well-Being*
- ✓ *Enhanced Safety in Food Supply*
- ✓ *Reduced Infections*
- ✓ *Reduced Illnesses*
- ✓ *Reduced Work-Related Injuries*
- ✓ *Reduced Environmental Footprint*
- ✓ *Reduced Liabilities*
- ✓ *Cost Effective*

CONTACT

"But when it has been shown by the researches of Pasteur that the septic property of the atmosphere depended not on the oxygen, or any gaseous constituent, but on minute organisms suspended in it, which owed their energy to their vitality, it occurred to me that decomposition in the injured part might be avoided without excluding the air, by applying as a dressing some material capable of destroying the life of the floating particles. Upon this principle I have based a practice."

Lord Joseph Lister, 1867

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