

## BIOFILMS

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## OBJECTIVES

1. Describe biofilms.
2. Discuss the infections associated with biofilms.
3. Identify problems with treatment and prevention.

## WHAT ARE BIOFILMS?

- Communities of organisms attached to a solid surface
  - Can be nonliving or living tissue surface
- Evolve over time consisting of many species
- Embedded in extracellular matrix
- Located at phase interface—flow
- Most important, they are a multiorganism cooperative population
- In nature 95-99% of microbes in biofilms



## EXAMPLES OF BIOFILMS

- Water pipes
- Ventilator system of airplanes or convention centers
- Wine casks causing spoilage
- Serious lung infections of cystic fibrosis (CF) patients



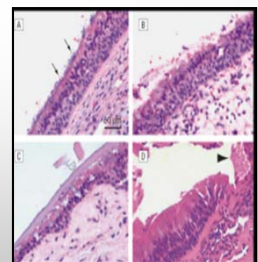
## DENTAL UNIT WATER LINES

- Legionella spp.
- Nontuberculous Mycobacteria (NTM)
- Pseudomonads
- Grow and multiply in biofilm to reach infective concentrations
- Potential for inhalation leading to respiratory infections
- Direct contamination of surgical wounds



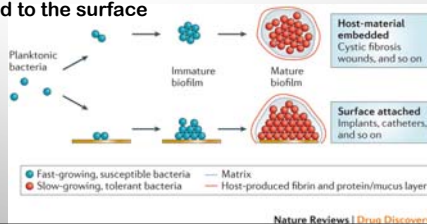
## BIOFILMS

- Account for over 80% of microbial infections in the human body (NIH)
- 17 million new biofilm infections with 550,000 fatalities each year
- Chronic infections and longer hospital stays



## BIOFILMS

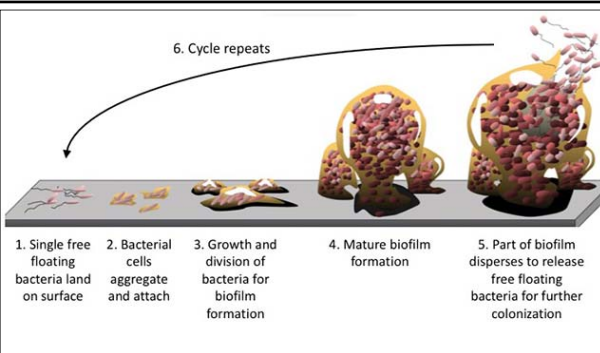
- Protect from oxygen & other harmful factors
- Two main types of biofilms
- Sessile
  - Permanently anchored to a surface
  - Covalently bonded to the surface
- Planktonic
  - Free floating
  - Movement to new habitats



## STEPS

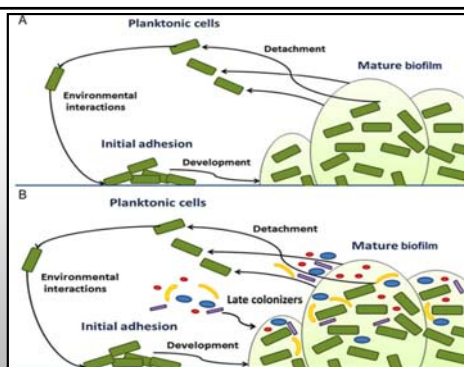
- Single cell layer—initial attachment to an abiotic or biotic surface
  - Production of reversible adhesion—living or dead
- Maturation of the biofilm
  - Irreversible adhesion—can move at first but lose motility and adhere to each other and excrete matrix to become microcolonies
  - Secondary colonizers become attached and pores, cavities, channels, outgrowths form
- Dispersal
  - Disintegration, degradation, loss, liberation—single cells or microcolonies

6. Cycle repeats



## STRUCTURES

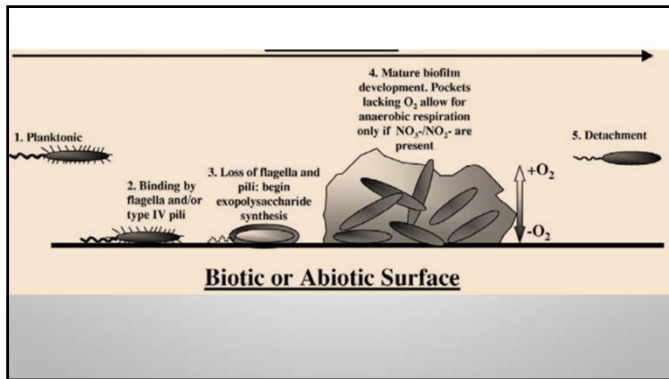
- Primitive-simple cell layer
- Mats of photosynthetic, methanogenic, and sulfate reducing communities (waste water)
- Dental biofilms (plaques) a complex community of many microorganisms and hundreds of species
- Bandlike outgrowths, formed by mixed bacterial populations under conditions of turbulent flow—torn off & disseminated
- Mature—3 dimensional structure with pores, channels, voids, signal components of “quorum sensing” system



## ARCHITECTURE



- Outer layer
  - Most dynamic and metabolically active cells
- Intermediate layer
  - Still active but less so
  - Genetic reservoir for genes involving nutrient utilization and drug resistance
- Inner surface layer
  - Persister cells
- Allow growth in hostile environment—stress



### GENETICS

- Exchange of genetic information high
- Scanning confocal laser microscopy—reporter gene encode fluorescent to see plasmids
- Microarrays for mRNA demonstrate differences in planktonic and embedded species

### GENETICS

**Bacterial conjugation**

- Gene transfer
  - Transformation
  - Conjugation
- Greater genetic potential as a group than alone
  - Eventually the virulence factors cluster, causing a worsening of disease.
- Disaggregation
  - Potential to transmit already upregulated resistant aggregates of microorganisms to other body sites

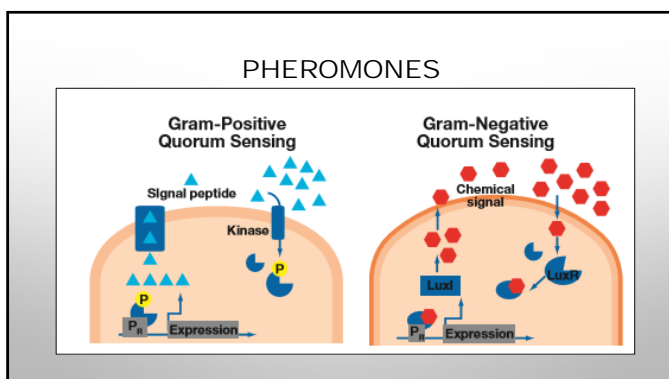
(a) Transformation with DNA fragments: Bacterial chromosome, Uptake of DNA, Integration by homologous recombination, Stable transformation, Degradation, Unsuccessful transformation.

(b) Transformation with a plasmid: DNA plasmid, Bacterial chromosome, Uptake of plasmid, Stable transformation.

### ROLE OF QUORUM SENSING

- Cell communication widely used to coordinate expression of traits once a population threshold is reached
- Metabolism
- Production of multiple virulence factors
- Chemotaxis
- Biofilm formation
- Swarming motility

Harmless bacteria → Quorum signals → Pathogenic bacteria → Virulence factors

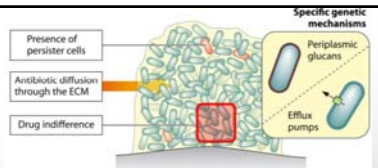


### PERSISTERS

Antibiotic Treatment → Decrease in Antibiotic Concentration → Persisters

- Metabolically inert cells present in all biofilms
- Disabled apoptosis
- Maintain gene pool
- Resist environmental stress (antimicrobials)

## RECALCITRANCE

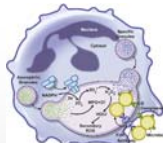


- **Survival after physicochemical aggression**
  - UV light, heavy metals, acidity, changes in hydration or salinity, and phagocytosis
  - Withstand antibiotic-mediated killing even when planktonic cells are susceptible

## RECALCITRANCE

- **Impaired antibiotic diffusion—subinhibitory levels**
  - Increases resistance
  - Increases mutations through conjugation
  - Rearrangement of integrin gene cassette
  - Increases persisters
- **Hypermutability—60-fold higher in *S.aureus***
  - Induces breakage in DNA
  - Down-regulates repair genes
- **Small-colony variants**
  - Better piliation, adhesion, adherence to cells

## RESISTANCE TO IMMUNE SYSTEM



- Matrix act as decoy molecules that prevent efficient microbial recognition by neutrophils
- Impaired oxidative burst and neutrophil killing
- Conceals b-glucans from recognition by innate immune pattern recognition receptors
- Protects from neutrophil extracellular traps (
- Down-regulation of flagellin expression and motility because flagellin is a ligand for Toll-like receptor
- Motility facilitates host cell invasion and phagocytosis

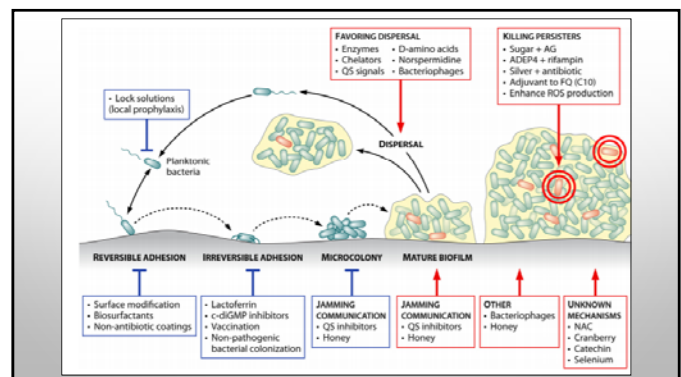
## PREVENTATIVE STRATEGIES



- **Hygiene, training, reduction in devices**
- **Removal of unnecessary devices**
- **Antibiotic prophylaxis during insertion**
- **Antibiotic coating—local high concentration**
- **Mechanical removal**

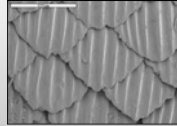
## OPTIMIZATION OF ANTIMICROBIALS

- **Choose best antimicrobial for organism**
- **Lock solutions for catheters—12 hours high concentration antimicrobials**
  - High risk patients
  - Can prevent blood stream infections
  - With systemic antimicrobials if not removed
  - **Must remove catheter if *S. aureus* or *Candida***



## PREVENTIVE STRATEGIES

- **Inhibiting adhesion**
  - **Material modifications and biosurfactants**
    - Zirconium oxide rather than pure titanium implants
    - Silicon coating
    - Sharkskin pattern
    - Preventing protein and platelet adherence
    - Bioactive antibodies, mannose
    - Lactoferrin
    - Acoustic waves in urinary catheters



## PREVENTIVE STRATEGIES

- **Jamming quorum sensing**
  - RNAIII-inhibiting peptide with *Staph. Aureus*
  - Azithromycin with *Pseudomonas aeruginosa*
  - Garlic and horseradish, green tea
- **Vaccination to biofilm antigens**
  - Before implantation
  - CF patients
  - Chronic UTI

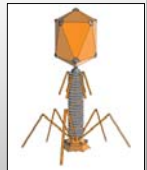


## ERADICATING BIOFILMS

- **Antimicrobials**
  - Up to 50% treatment failures, resistance, prolonged treatment
- **Non-antimicrobials—alone or in combination**
  - Inducing dispersal to return to planktonic state
  - Enzymes
  - Divalent cation chelators—EDTA & citrate
  - QS signals
  - *Bacillus subtilis*
  - NO

## ERRADICATING PERSISTERS

- **Aminoglycosides and mannitol or fructose**
  - Stimulation of PMF leads to increased aminoglycoside uptake
- **Silver—↑ membrane permeability to ↑ effect of gentamicin, ofloxacin, or ampicillin**
- **Cocktail of bacteriophages**

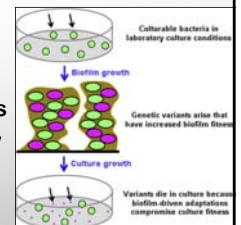


## KEY CHARACTERISTICS

Key steps in surface-attached biofilm formation:	<ul style="list-style-type: none"> <li>• Surface attachment to biotic and/or abiotic surface</li> <li>• Production of matrix including exopolysaccharide</li> </ul>
Mechanisms to increase resistance to host defenses:	<ul style="list-style-type: none"> <li>• Concealment or down-regulation of pathogen-associated molecular patterns or antigens</li> <li>• Resistance to phagocytic activity, host antimicrobial defenses, and NET killing</li> </ul>
Mechanisms to increase resistance to antimicrobial drugs:	<ul style="list-style-type: none"> <li>• Physiologic heterogeneity in biofilms, leading to subpopulations that are metabolically quiescent, slow growing, or that have induced stress responses</li> <li>• Limited diffusion or sequestration of antimicrobials by biofilm matrix</li> <li>• Increased expression of antimicrobial efflux pumps</li> </ul>

## WHY ARE OUR CULTURE PLANKTONIC?

- **Rich media and optimal conditions**
- **Fast growing**
- **Biofilms protect from stress factors so organisms adapt phenotypically**



## LABORATORY CONSIDERATIONS

## • Cultures

- Require growth to get colonies
  - Problem is colonies won't grow under normal conditions



## • False negatives

- Improper sample collection
  - Swabs or culturing outer surface of equipment

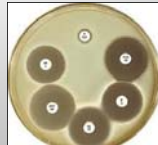
## LABORATORY CONSIDERATIONS

## • Aggregates of organisms

- Single colonies can represent up to 100,000 bacteria of mixed origin
- Thus amounts of each organism are greatly underestimated or not considered significant

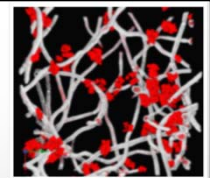
## CULTURE AND SUSCEPTIBILITY

- Sonication of removed hardware to remove biofilm
- Antibiotic susceptibility
  - Single isolates that are members of a biofilm may not represent the genetic potential or resistance of a community



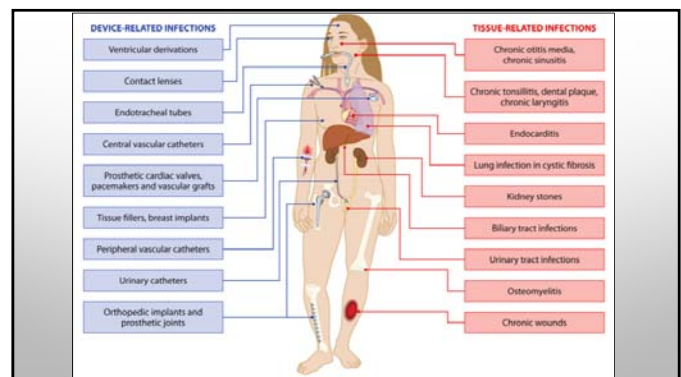
## DETECTION

- Biopsies of tissue or removed device
- PCR
- Pathogen-specific probes
- Confocal laser scanning microscopic imaging



## CHRONIC, DIFFICULT TO ERADICATE INFECTIONS

- Cystic fibrosis and *Pseudomonas aeruginosa*
- Indwelling medical devices with *Staphylococcus* and *Candida*

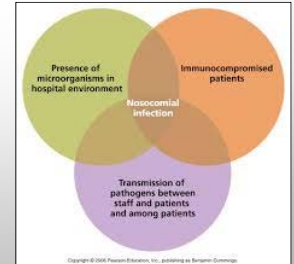


## INDWELLING MEDICAL DEVICE-RISK RELATED INFECTIONS IN THE UNITED STATES

Device	Usage	Infection Risk (%)
Bladder catheter	Tens of millions	10-30
Cardiac-assisted devices	700	50-100
Cardiac pacemakers	400,000	1-5
Central venous catheters	5 million	5-8
Dental implants	1 million	5-10
Fracture fixators	2 million	5-10
Joint prostheses	600,000	1-3
Penile implants	15,000	2-10
Prosthetic heart valves	85,000	1-3
Vascular grafts	450,000	2-10

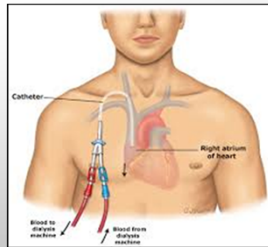
## BIOFILMS: A MICROBIAL RESERVOIR FOR NOSOCOMIAL INFECTIONS

- Hospital environment
  - Water distribution system
  - Contaminated surfaces
  - Biocides ineffective
  - Resistant to desiccation



## INDWELLING CATHETER-ASSOCIATED INFECTIONS

- Central venous catheters in ICU
- ↑cost, ↑length of stay, ↑mortality
- Skin commensals:
  - Coagulase negative staph
  - *Staphylococcus aureus*
  - *Candida*
- Removal necessary



## TREATMENT AND PREVENTION

- Surface coatings
  - Antimicrobials
  - Metal (silver, bismuth) nanoparticle
- Disruption of biofilm—removal
- Antimicrobial treatment—lock therapy



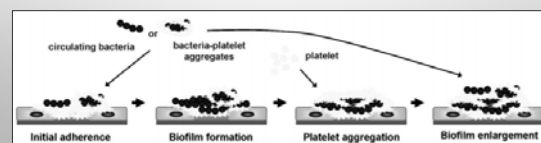
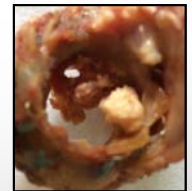
## INFECTIVE ENDOCARDITIS

- Biofilm associated with commensal strep on damaged heart valves
- Fibrin-platelet complex embedded with bacteria on heart valve

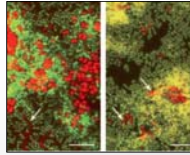


## ENDOCARDITIS

- Platelets essential
- Biofilm induced aggregation
- Platelets increase resistance



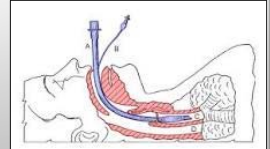
## AIRWAY BIOFILMS



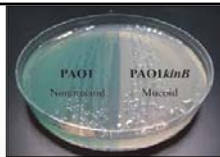
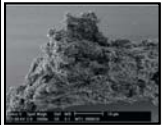
- Cystic fibrosis , diffuse panbronchiolitis and bronchiectasia with *Pseudomonas aeruginosa*
- Acute exacerbations from planktonic bacteria that dispersed from biofilm
- Slow progressive disease that induced by harmful immune reactions

## ENDOTRACHEAL TUBE COLONIZATION AND VENTILATOR-ASSOCIATED PNEUMONIA

- Readily accumulate within hours
- Aerosolization releases to cause pneumonia
- Major reservoir—50% of pneumonia caused by biofilm organisms
- Associated with treatment failure
- Oral and enteric organisms
- Under estimated by current cultures

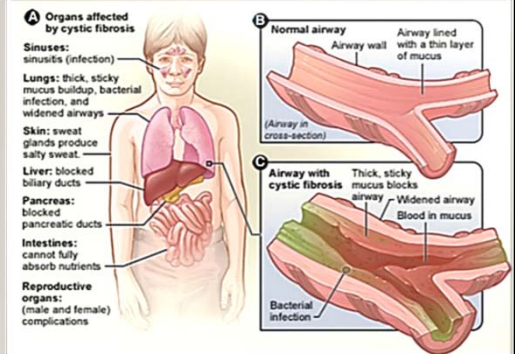


## CYSTIC FIBROSIS

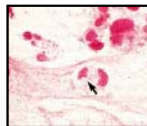


- Dehydrated and thickened airway surface liquid
- Hinders mucociliary clearance
- Colonize and cause an initial acute infection and vigorous inflammatory response
- Thickened ASL severely impairs the immune response
- Chronic lung inflammation

## CF



## BIOFILMS IN COPD



- 24 million in U.S. 3<sup>rd</sup> cause of death worldwide
- Intermittent exacerbations-50% infections
- Nontypeable *Haemophilus influenzae*



## ORAL BIOFILMS (PLAQUE)

- Influenced by: age, dietary sugar, oral hygiene, systemic and immune conditions, hyposalivation
- Sugar crevices, attracts pathogens causing inflammation and gingivitis
- Plaque accumulates at the sulcus
- Undisturbed spreads over teeth
- Good oral hygiene removes this plaque





## DENTAL BIOFILMS



- If accumulation occurs:
  - Switch from gram-positive to gram-negative bacteria
    - Anaerobic and facultative anaerobic, gram-negative bacilli and spirochetes
  - Pathogens
    - *Porphyromonas gingivalis*, *Bacteroides forsythia*, *Aggregatibacter actinomycetemcomitans*, *Treponema denticola*
  - Causes periodontitis—destroys bone and tissue

## BIOFILMS ON CONTACT LENSES



- May lead to microbial keratitis—*Ps. aeruginosa*—*Serratia marcescens*, *Staph. epidermidis* and *Staph. aureus*
  - Corneal scarring and vision loss
  - 12-66% contact lens wearers
- Acute red eye—34% of continuous wear
- Peripheral ulcer
- Infiltrative keratitis



## BIOFILMS ON CONTACT LENSES



- Influenced by
  - Length of wear
  - Pathogen
  - Deposited proteins
  - Lens material (hydrophobicity, roughness)

## OTITIS MEDIA



- 80% of children develop OM before age 3
- Recurrent, nonresponsive or chronic—biofilm
- Mixed pathogen: *Streptococcus pneumoniae* and nontypeable *Haemophilus influenzae*
- Biofilm more likely with combination and greater resistance
- *S. pneumoniae* differs by serotype

## TYMPANOSTOMY TUBE



- 600,000 placed in children per year
- Tube otorrhea (83%) and occlusion (74%)
- Acute infections: *Streptococcus pneumoniae* and *Haemophilus influenzae*
- Chronic: *Staphylococcus aureus* and *Pseudomonas aeruginosa*
- Financial and operative burden
- Organoselenium coating lessens *S. aureus* biofilm

## URINARY TRACT INFECTIONS

- Cause relapses, reinfection and chronic prostatitis
- 20% of UTIs
- *Escherichia coli* cause ~80%
- Urinary catheters—80% nosocomial—all colonized by day 30
- Treatment with nanoparticles

## CHRONIC WOUNDS

- Obesity, diabetes, cardiovascular
- \$10.9 billion and precede 85% of amputations
- Wound > 1 month
- Diabetic foot ulcers, pressure or decubitus ulcers, venous leg ulcers, and nonhealing surgical-site infections.



## CHRONIC WOUNDS



- Multiple organisms
- *S. aureus*, *P. aeruginosa*
- *P. aeruginosa*—larger wounds, delay or prevent healing

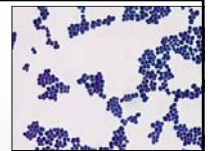
## TREATMENT OF CHRONIC WOUNDS

- Debridement
- Antibiotics and anti-inflammatory drugs
- Moisture imbalance corrected with dressings
- Epithelialization and tissue formation promoted specific therapies, such as growth factors



## STAPHYLOCOCCUS

- *S. aureus* and *S. epidermidis*
- Osteomyelitis, endocarditis, medical device implants, and persistence in cystic fibrosis
- 10,000-fold lower # needed to colonize foreign body than to cause skin abscess
- Secreted polysaccharide (slime) required



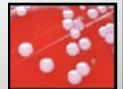
## STAPHYLOCOCCUS AUREUS

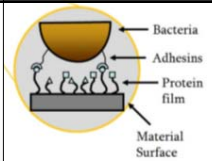
- High morbidity and mortality in endovascular
- Infective endocarditis, osteomyelitis, arthritis
- Sublethal doses of vancomycin can induce biofilms in MRSA
- Foci for metastatic spread and toxin release
- Matrices consist of proteins, DNA, and polysaccharide
- Polysaccharide not essential—protein in highly virulent



## STAPHYLOCOCCUS EPIDERMIDIS

- Normal microbiome
- Causes ~ 20% of orthopedic device-related infections
- Increasing up to 50% in late-developing infections
- Prominent in any implanted device infection
- Low level of virulence factors
- Triggers low levels of pro-inflammatory cytokines and high levels of interleukin-10
- May contribute to the sub-acute and persistent nature



*S. EPIDERMIDIS* BIOFILM

- Protein coating required—fibronectin
- Polysaccharide intercellular adhesion
- Or accumulation associated protein (less robust)
- Form different structures

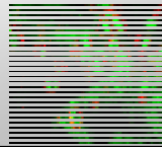
*S. EPIDERMIDIS* AS A PATHOGEN

- Preterm neonates
- Immunocompromised
- Indwelling medical devices



## STAPHYLOCOCCAL EVASION OF THE HOST IMMUNE SYSTEM

- PMNs can attack biofilms by phagocytosis, release of toxic granule components, and production of NETs
- Extracellular polysaccharide prevents attachment
- agr adhesion molecule kills PMNs
- Extracellular nuclease can degrade NETs

*STAPHYLOCOCCUS LUGDUNENSIS*

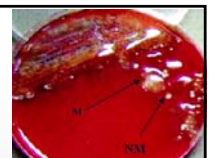
- Most similar to coagulase-positive Staphylococci
- Skin commensal and opportunistic pathogen
- 0.8%–7.8% of infectious endocarditis cases in nondrug users with mortality rates 38%–42%
- Infections of medical devices, such as catheters and prosthetic joints
- Significant cause of skin and soft-tissue infections

## PSEUDOMONAS AERUGINOSA

- Ventilator-associated pneumonia, cystic fibrosis meningitis, abscess, infections of skin and soft tissues (including diabetic foot), urinary tract, bone and joint, bacteremia, corneal infections, systemic diseases
- Immunosuppressed patients
- MDR and avid biofilm producer
- Catheters (urinary & vascular), ventilator tubes, chronic leg wounds



## ALGINATE



- Soft loosely adhered polymer that surrounds the cells
- Protects *P. aeruginosa* from harsh environments in CF lungs
- Provides extracellular matrix in biofilms—up regulated
- Virulence factors and motility downregulated

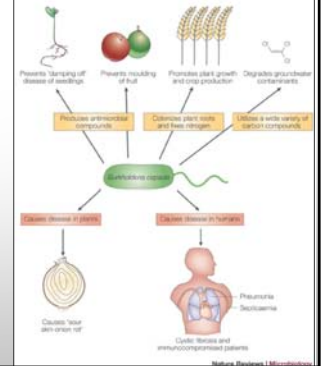
## ALGINATE



- Inhibits immune defenses
  - Inhibition of bacterial uptake and killing by macrophages
  - Prevents activation of the complement alternative pathway
  - Reduces opsonophagocytosis
- Structural stability and protection of biofilms
- Necessary for water and nutrient retention

## BURKHOLDERIA CENOCEPACIA

- Beneficial OR
- Opportunistic pathogen
- Severe lung infections, necrotizing pneumonia and septicaemia in CF
- 9 species (genomovars)
- *B. cenocepacia* (genomovar III) and *B. multivorans* (genomovar II)



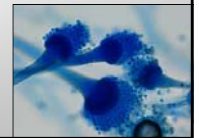
## BURKHOLDERIA CENOCEPACIA

- Decline in CF lung function
- May develop into systemic infection--cepacia syndrome
- Major cause of premature death and lung transplant
- Post transplant infections morbidity & mortality
- Antibiotic resistance and numerous virulence factors

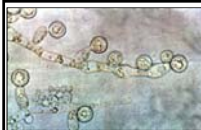


## ASPERGILLUS FUMIGATUS BIOFILMS

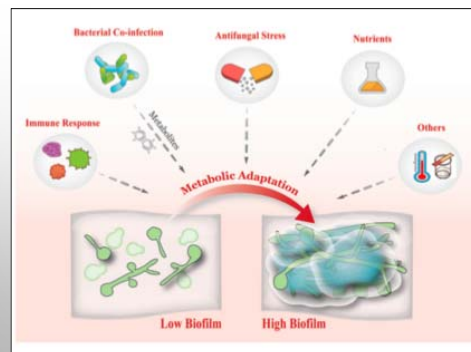
- Newly identified biofilm producer
- Conceals  $\beta$ -glucans from recognition by an innate immune pattern recognition receptor
- Protects hyphae from neutrophil extracellular traps
- Contributes to high treatment failure rate



## CANDIDA ALBICANS

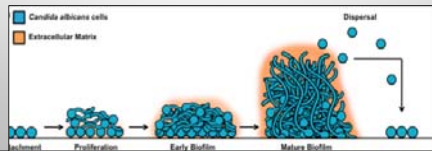


- Most common human fungal pathogen--ranging from mucosal to systemic infections
- Asymptotically colonizes mucosal surfaces
- Disruption in the host environment or immune dysfunction, proliferates and invades any site
- Adheres to catheters and indwelling medical implants
- 3<sup>rd</sup> most commonly isolated bloodstream pathogen in hospitalized patients with a mortality up to 50%



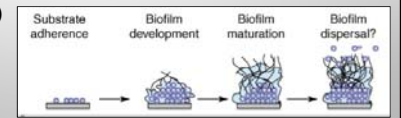
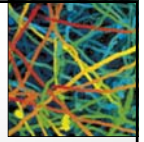
*CANDIDA ALBICANS*

- Majority of infections associated with biofilms
- Indwelling medical devices--high morbidity and mortality
- Significant drug resistance



## BIOFILM

- Production of hyphae is a hallmark of initiation formation
- Accumulation of extracellular polysaccharide matrix as matures-- -mannan & glucan
- Dispersal of yeast seeds to other organs (differ from planktonic yeast)

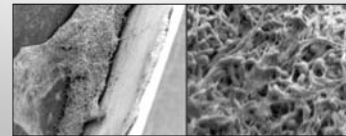
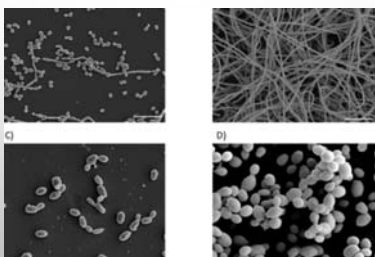


## THRUSH

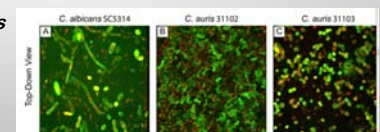
- Pseudomembranous candidiasis
- Most common oral opportunistic infection in HIV+ and other immunocompromised individuals
- Denture stomatitis occurs in up to 70% of denture wearers--chronic

*CANDIDA* IN VASCULAR CATHETERS

- Most dangerous—20% polymicrobial
- Up to 20,000-fold increase in antifungal MICs
- ↓ growth rate, cell density, modified target, efflux
- Persists and extracellular matrix

MIXED *CANDIDA* BIOFILMS*CANDIDA AURIS*

- New multi-resistant invasive yeast with high mortality (60%)
- Ability to attach to silicon elastomer catheter significantly less than *C. albicans*
- Did not produce hyphae in biofilms & had much less extracellular matrix
- Half as thick as *C. albicans*



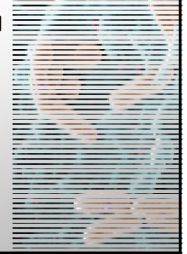
### *KLEBSIELLA PNEUMONIAE*

- Pneumonia, UTI, liver abscess
- Most frequently associated with nosocomial infections in US-indwelling urinary catheters
- ESBL and CRP
- Polysaccharide capsule
- Fimbriae important in UTI biofilms



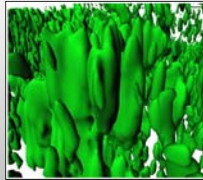
### *KLEBSIELLA PNEUMONIA* BIOFILM IN UTI

- Resistant to long exposure to ampicillin and ciprofloxacin, gentamicin, cefotaxime
- Polysaccharide capsule, fimbriae
- Enhanced in mixed esp. with *Ps. aeruginosa*, *E. coli*, *P. mirabilis*, *organella*, *Enterobacter*, *C. albicans*, *Streptococcus*



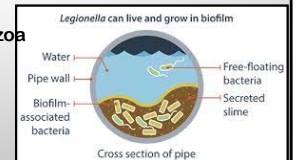
### *ESCHERICHIA COLI* UTI BIOFILMS

- Biofilm in GI tract also—250 serotypes
- From harmless gut commensal to pyelonephritis and sepsis
- Primary urinary biofilm producer
- Wide array of genetic tools, fimbriae and flagella



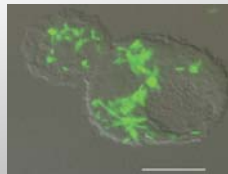
### *LEGIONELLA PNEUMOPHILA*

- Severe respiratory illness with fatality from 5% to 80%
- Aerosols
- Ubiquitous in natural and anthropogenic water systems
- Biofilms essential in water systems
- Usually monospecies or with protozoa
- Enhanced virulence



### *LEGIONELLA PNEUMOPHILA*

- Amount of biofilm directly correlated with the biomass of protozoa
- Replicate inside protozoa
- Adhere well to plastic, not copper
- *Pseudomonas* inhibitory
- *Acanthamoeba castellanii*



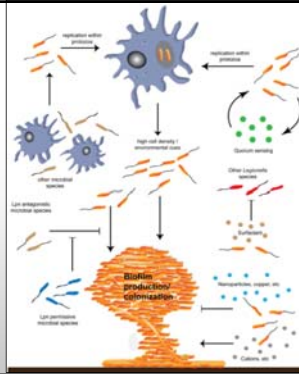
### FACTORS THAT INFLUENCE BIOFILMS

- CA, Zn, Mn, Fe AND Mg facilitate attachment
- Lactoferrin can directly kill *L. pneumophila*
- Carbon at 20° and stagnation
- Heat to 55° reduces
- 37–42 °C, monospecies mycelial mat-like and filamentous bacteria
- 25 °C are thinner and made up of rod shaped cells
- Summer in cooling towers



### ELIMINATING *L. PNEUMOPHILA*

- Extremely resistant to disinfectants and biocides
- Resistant to chloride
- Nanoparticles better

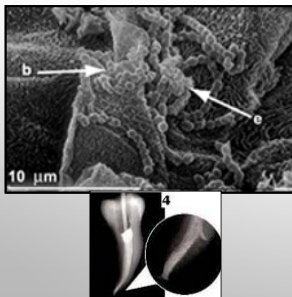


### *STREPTOCOCCUS PYOGENES*

- Pharyngitis, cellulitis, and impetigo to puerperal sepsis, myositis, toxic shock, necrotizing fasciitis
- Post sequelae rheumatic fever and acute glomerulonephritis
- Large amounts of *S. pyogenes* cells and a lack of neutrophils in necrotizing fasciitis demonstrate biofilms



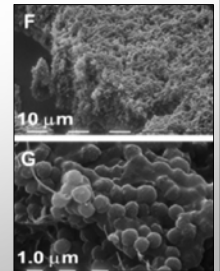
### *STREPTOCOCCUS PYOGENES*



- Biofilm necessary for carriage state in tonsils
- Account for survival with antimicrobial therapy
- Biofilms form on gutta percha points (material used in tooth cavity repair and root canal procedures)

### *STREPTOCOCCUS PYOGENES*

- Ability differs among serotypes (M types)
- M protein necessary to initiate
- Macrolide susceptible strains produce significantly more biofilm than resistant strains
- Penicillin can kill biofilms
- Sub-lethal concentrations of fluoroquinolones can inhibit



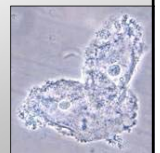
### *ENTEROCOCCUS*

- *E. faecalis* twice the biofilm producer of *E. faecium*
- Catheter-related bloodstream infections
- Endocarditis isolates
- 50% of root canals



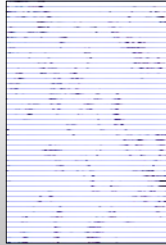
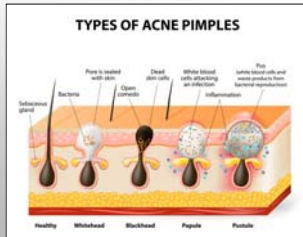
### *GARDNERELLA VAGINALIS* AND ANAEROBES IN BACTERIAL VAGINOSIS BIOFILM

- Most common vaginal disorder in women of reproductive age (60%)
- Causes PID, postoperative infections, preterm birth and susceptibility and spread of HIV
- ↓ lactobacilli & ↑ pathogens, primarily anaerobes
- *Gardenerella vaginalis* first? Most important?
- Different strains?



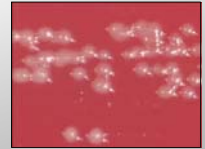
## CUTIBACTERIUM (PROPIONIBACTERIUM) ACNES

- Skin commensal



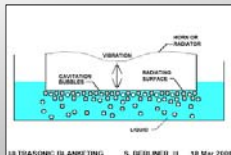
## BIOFILM PRODUCER

- Implant-associated infections
- Shoulder prosthetic joint infections
- Cerebrovascular shunt infections
- Fibrosis of breast implants
- Cardiovascular devices



## C. ACNES IDENTIFICATION

- Improved diagnostic procedures
  - Sonication
  - Prolonged cultivation time of up to 14 days
  - Improved molecular methods
    - Broad-range 16S rRNA gene PCR



## COMPARISON OF BIOFILM INFECTIONS WITH ACUTE AND COMMENSAL

Features of biofilm infections	Necessary condition for biofilm infection	Also found in acute planktonic infection	Also found in colonization/normal flora on skin and mucosal membranes
Aggregates of bacteria embedded in a self-produced polymer matrix	Yes	No	No/Yes
Tolerant of clinically relevant PK/PD* doses of antibiotics, despite the susceptibility of planktonic cells	Yes	No	No/Yes
Tolerant of innate and adaptive immune responses	Yes	No	No/Yes—unknown
Inflammation	Yes	Yes	No
<b>Chronic infections</b>	Yes	No	No
Foreign body-associated infections	No	Initial	No
Located on surfaces	No	Yes	Yes
Localized infection	Yes	Yes	Yes
Focus of spreading or local exacerbation	Yes	Yes	Yes

\*PK, PD, pharmacokinetic/pharmacodynamic [adapted from (16)]

## SUMMARY OF PATHOGENICITY

- Attachment to a solid surface
- “Division of labor” increases metabolic efficiency
- Evades host defenses
- High density of microorganisms
- Horizontal gene transfer—more virulent strains
- Produces large amount of toxins
- Protects against antimicrobials
- Dispersion transmits organisms to other body sites

## SURVEY MONKEY

PROGRAM EVALUATIONS  
NO SCANTRON SHEETS

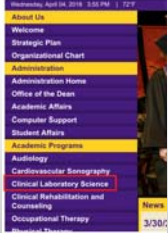


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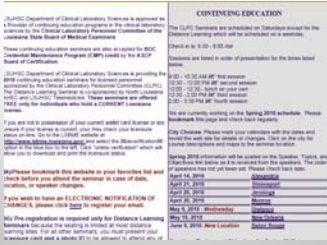


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INSTRUCTIONS

- QUESTION 1 - LICENSE NUMBER
- QUESTIONS 2 AND 3 - SPEAKER 1
- SAME QUESTIONS AS SCANTRON SHEETS
- CLICK NEXT TO SAVE
- QUESTIONS 4 AND 5 - SPEAKER 2
- QUESTIONS 6 AND 7 - SPEAKER 3
- QUESTIONS 8 AND 9 - SPEAKER 4
- QUESTION 10 - RECOMMENDATIONS
- QUESTION 11 - GENERAL COMMENTS/FUTURE PROGRAMS
- CHANGES CAN BE MADE TO ANY PAGE UNTIL YOU CLICK DONE AFTER QUESTION 11.

1. Please fill in your CLPC license number in the space provided below.

2. Speaker 1:

	Excellent	Good	Average	Poor
Speaker Evaluation - Overall quality of presentation	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Speaker Evaluation - Knowledge of subject matter	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Speaker Evaluation - Organization of presentation	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Course Evaluation - Rate the session	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Course Evaluation - Material covered/selected?	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>

3. Please comment on the speaker and course strengths/weakness.

Next

10. Would you recommend these presentations to others?

	YES	NO
Presentation 1	<input type="radio"/>	<input type="radio"/>
Presentation 2	<input type="radio"/>	<input type="radio"/>
Presentation 3	<input type="radio"/>	<input type="radio"/>
Presentation 4	<input type="radio"/>	<input type="radio"/>

11. Please comment on the program in general and list suggestions for future programs.

PREV DONE