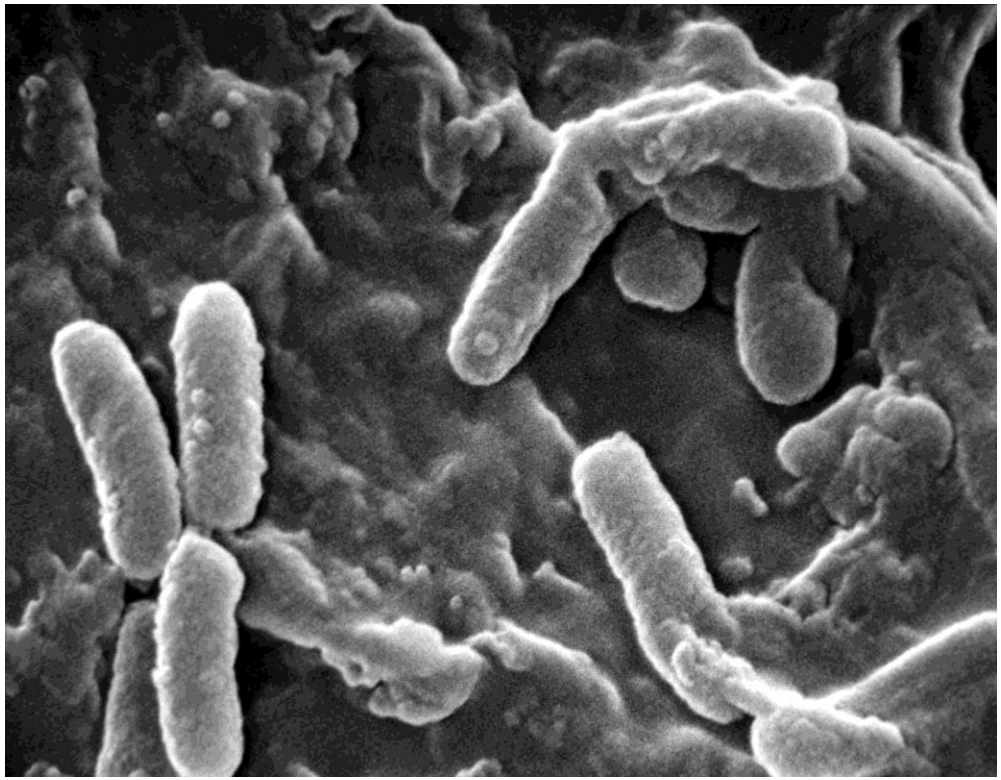
Pseudomonas Aeruginosa

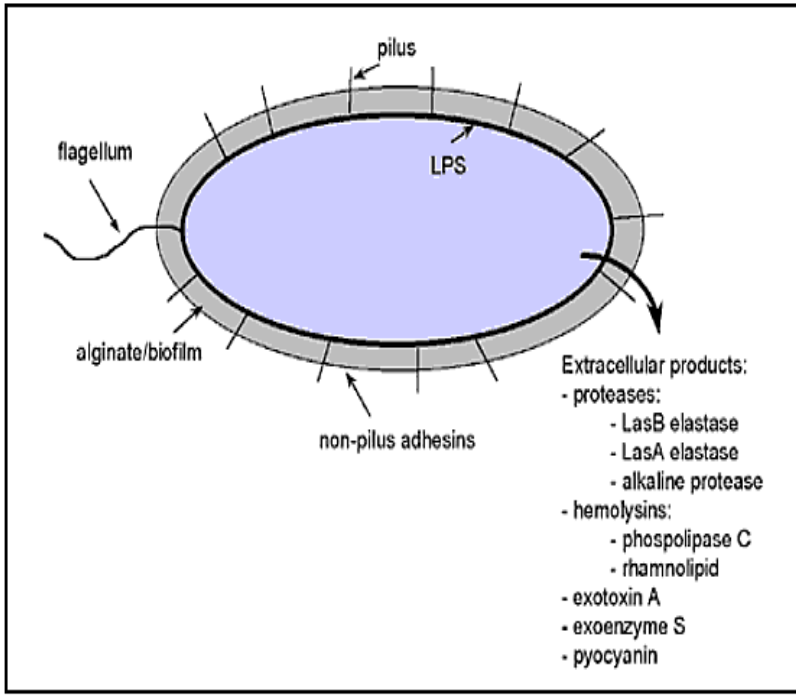


Pseudomonas aeruginosa

- Motile (by single or multiple polar flagella) gram-negative rods
- Obligate (strict) aerobes (most strains)
- Oxidase (usually) and catalase positive
- Nonfermentative chemoheterotrophic respiratory metabolism
- Minimal nutritional reqts.; Many organic compounds used as C and N sources, but only a few carbohydrates by oxidative metabolism
  - Glucose used oxidatively
  - Lactose negative on MacConkey's agar
- Some strains produce diffusible pigments:
  - Pyocyanin (blue); fluorescein (yellow); pyorubin (red)
- *P. aeruginosa* produces characteristic grape-like odor and blue-green pus & colonies
- Broad antibiotic resistance

Structural Components

- Adherence to host cells mediated by pili and nonpilus adhesins.
- LPS (lipopolysaccharide) inhibiting antibiotic killing and suppress neutrophil and lymphocyte activity
- Alginate – mucoid exopolysaccharide that forms a shiny biofilm protecting from antibodies, complement, phagocytosis, and antibiotics
- Pyocyanin – impairs ciliary function, mediates tissue damage through production of oxygen radicals



Epidemiology

- Primary habitat is the environment (water and soil)
- Normal flora 2-10% of healthy individuals
- Infections mostly in hospitalized patients with illnesses, such as CF, leukemia and burns.
- Minimal nutritional requirements and can tolerate broad temperature spectrum (20-42C) and thus contaminate respirator humidifiers, medications and contact lens solutions.
- Can transiently colonize the respiratory and GI tract of hospitalized patients
- Respiratory colonization of CF patients become chronic leading to high morbidity and mortality rates.
- No seasonal incidence

Pathogenesis

- **Require break of first-line defenses (wound or contaminated solution)**
- **Attachment by pili, flagella and extracellular polysaccharide**
- **Virulence Factors (toxins and enzymes):**
  - **Exotoxin A**
    - Similar in structure to Diptheria toxin
    - Inhibits protein synthesis by ADP-ribosylating EF-2 (G-protein)
    - Causes Dermatonecrosis in burn wounds, corneal damage in ocular infections, and tissue damage in chronic pulmonary infections.
    - Also this toxin is immunosuppressive
    - Its presence is associated with fatal outcome and Ab against it is with survival.
  - **Exoenzyme S**
    - ADP-ribosylates G-proteins including p21 RAS interfering with host cell growth
    - Cellular destruction and spread from burn wounds
  - **Elastase**
    - Destruction of elastin-containing tissues (blood vessels, lung tissue, skin), collagen, immunoglobulins (IgA and IgG), and complement factors
    - Can produce hemorrhagic lesions (ecthyma gangrenosum) associated with disseminated infection
  - **Endotoxins**
  - **Phospholipase C**
    - Breaks down lipids and lecithin causing tissue destruction
  - **Alkaline Protease**

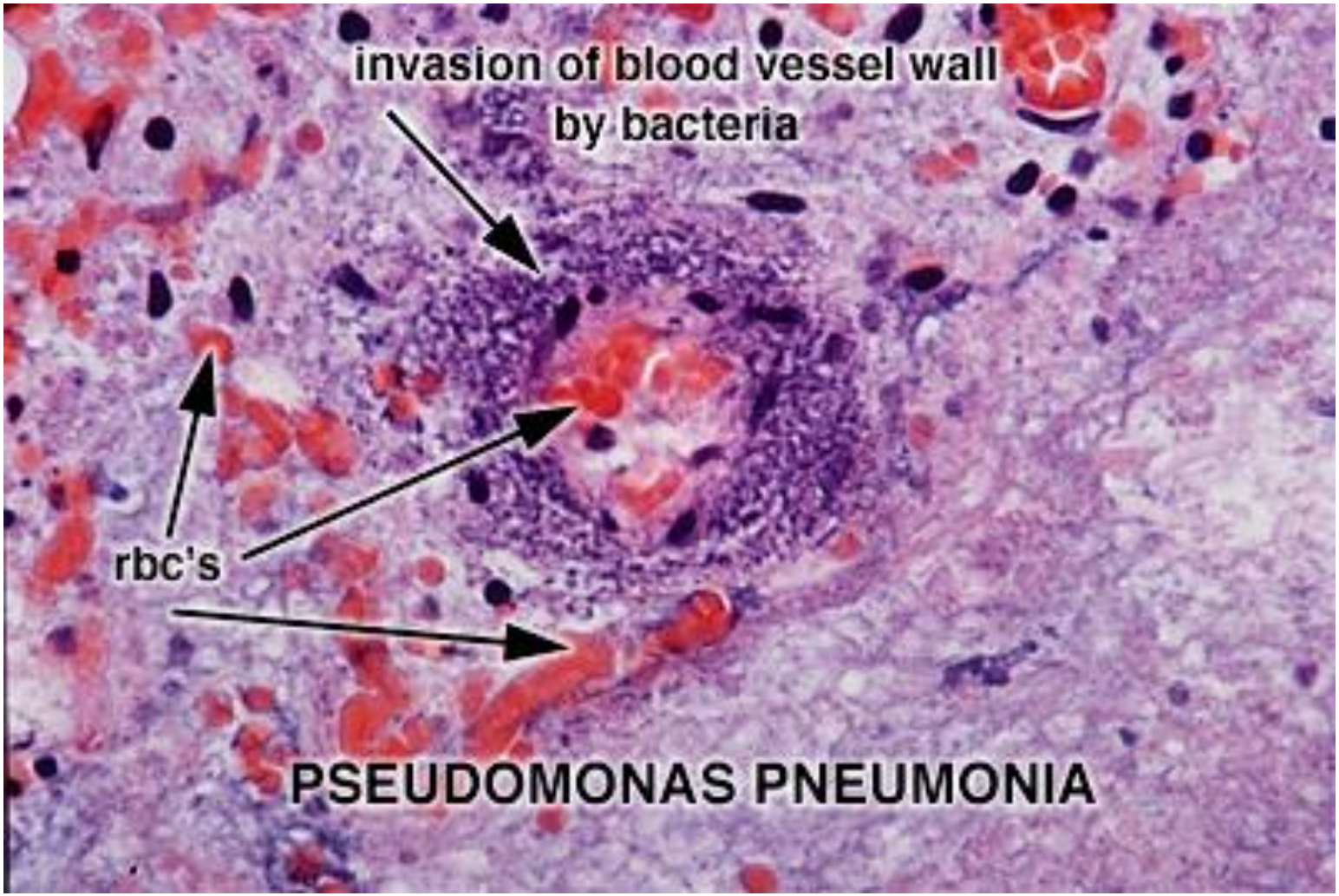
Clinical Disease

- Pulmonary Infections
- Burn Wound Infections and other skin and soft tissue infections (life threatening)
- UTI’s (especially catheterized)
- External Otitis (malignant OE, swimmer’s ear)
- Eye Infections and corneal ulceration via contaminated contact lens cleaning fluids
- Pseudomonal Endocarditis

Pulmonary Infections

- Can range from asymptomatic colonization to severe necrotizing bronchopneumonia
- Colonization is seen in patients with cystic fibrosis, chronic lung disease, and neutropenia
- Mucoid strains are commonly isolated from chronic pulmonary patients and are more difficult to eradicate
- Predisposing conditions include previous therapy with broad spectrum abx (disrupts normal protective bacteria population and use of respiratory therapy equipment (can introduce the organism to lower airways)
- Mortality rate can be as high as 70% for invasive bronchopneumonia

Pseudomonas PNA



Ecthyma Gangrenosum

- **Ecthyma gangrenosum is a well recognized cutaneous manifestation of severe, invasive infection by *Pseudomonas aeruginosa* that is usually seen in immunocompromised, burn patients, and other critically ill patients**

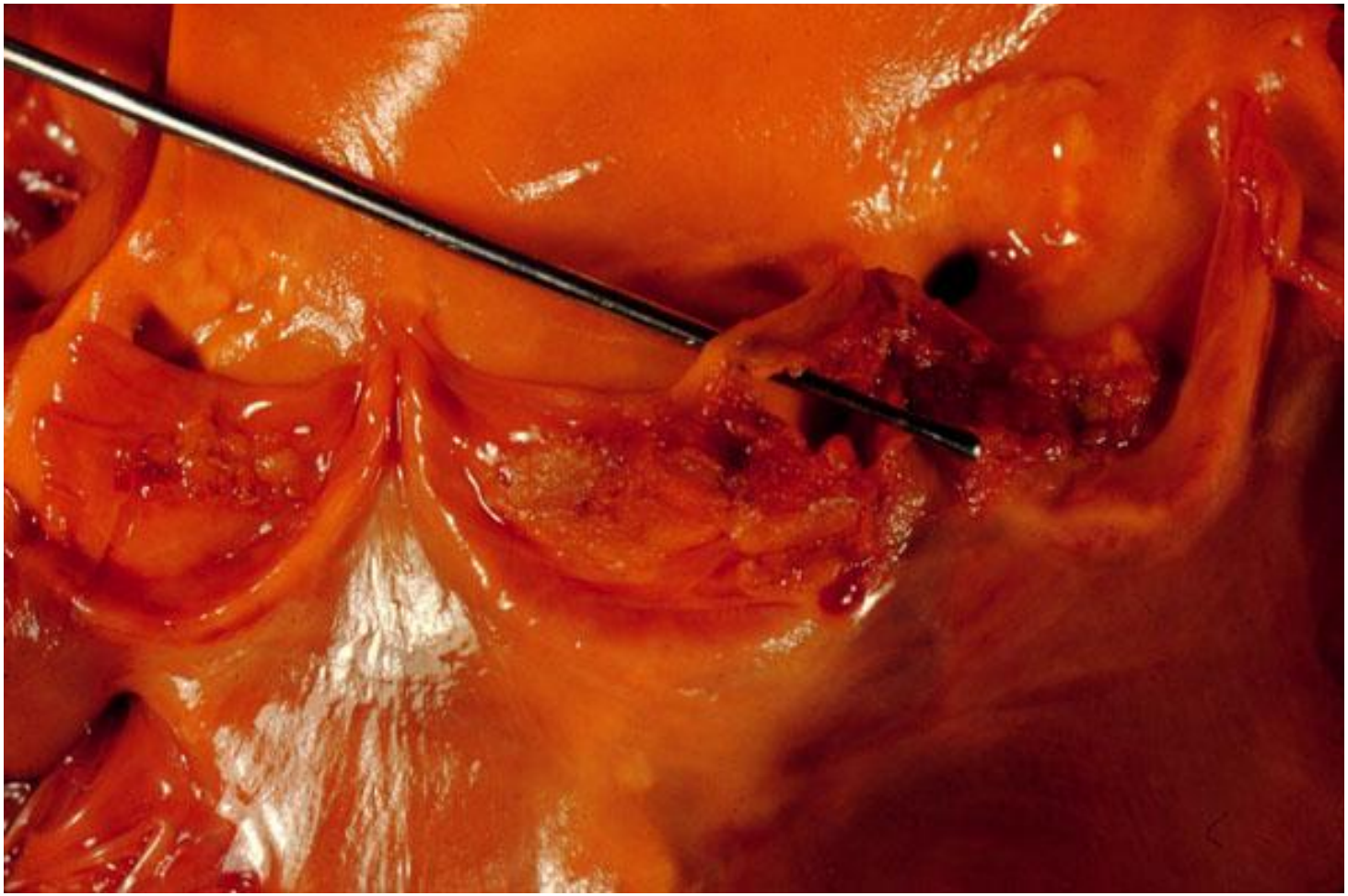
Black necrotic ulcer

Malignant Otitis Externa

Diabetic pts

Pseudomonas Keratitis and Corneal Ulceration

Endocarditis



Diagnosis and treatment

Dx

- Oxidase positive colonies, pyocyanin production and ability to grow at 42C are diagnostic
- Culture and sensitivity tests

Rx

- Inherently resistant to many antibiotics (penicillin, ampicillin, tetracycline, earlier aminoglycosides and sulfonamides)
- Can mutate to more resistant strains during therapy
- Penetration of antibiotic is highly dependent on outer membrane pores which can be altered (high dose carbenicillin and ticarcillin)
- Production of B-lactamases
- 3rd generation cephalosporins, carbapenems and monobactams drugs of choice
- Combination of active antibiotics generally required for successful therapy (Anti-β-lactam and aminoglycoside)

- No vaccine

Moraxella catarrhalis

GENERAL OVERVIEW

- Formerly classified as *Neisseria* & more recently *Branhamella*
- Gram negative, aerobic coccobacilli, nomotile
- Grows on blood or chocolate agar

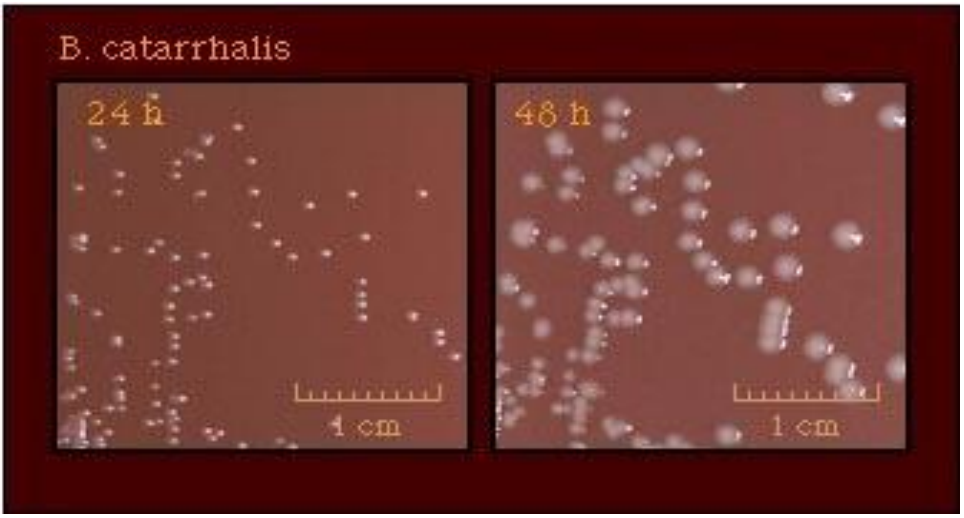
CLINICAL SYNDROMES

- In Elderly Patients with Chronic Pulmonary Disease
  - Bronchitis
  - Bronchopneumonia
- In Previously Healthy People (normal flora)
  - Sinusitis
  - Otitis

TREATMENT, PREVENTION, AND CONTROL

- Most strains produce β-lactamase; Penicillin Resistant
- Amoxicillin-clavulanate, second and third generation cephalosporin

Colony morphology



Catalase positive

Oxidase positive

Penicillin resistant

Mycoplasma

- Classification – order Mycoplasmatales; family Mycoplasmataceae; 2 medically important genera
  - *Mycoplasma*
  - *Ureoplasma*
  - Three common clinical isolates – *M. pneumoniae*, *M. hominis*, *M. genitalium* and *U. urealyticum*
- Morphology and cultural characteristics
  - Do not possess the distinctive cell wall of bacteria

Mycoplasma

- Plasma membrane is the outermost part of the organism and is unique among bacteria in that it has a high content of sterols (acquired from medium or tissue living in) that act to prevent osmotic lysis
- Very small in size (too small to be seen with an ordinary light microscope) and highly pleomorphic
- Don’t stain with a Gram stain
- Non-motile
- May possess a capsule
- Although some are free living, most are closely adapted parasites

Mycoplasma

- Grow on media enriched with serum (need cholesterol)
- Grow best at 35-37^0 C either aerobically or anaerobically
- *M. pneumoniae* grows in 5-14 days, *M. hominis* in 2-4 days, and *U. urealyticum* in 24-28 hours.
- M. pneumoniae colonies resemble fried eggs and can be stained with Dienes stain (they stain blue)

Mycoplasma colonies with Diene's stain

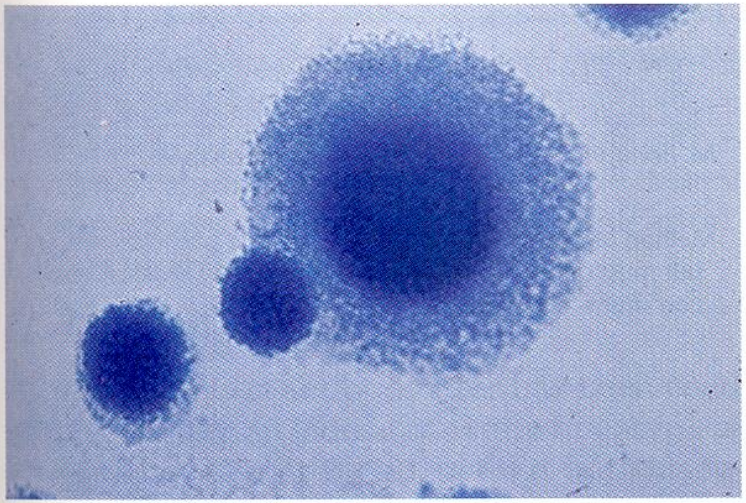


Figure 21-16

Diene's stain of *Mycoplasma* spp. colonies demonstrating typical "fried egg" appearance.

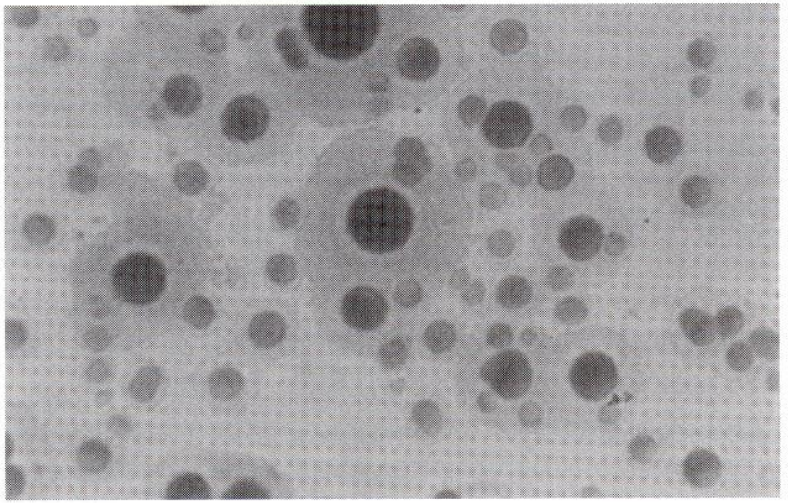


Figure 21-17

Typical mixed sizes of *Mycoplasma* organisms on primary isolation media: *Mycoplasma salivarium*. (Courtesy Bionique Testing Laboratories, Saranac Lake, N.Y.)

Mycoplasma

- Virulence factors
  - Not invasive and simply colonize cell surfaces through specific binding
  - It binds to cilia in the bronchus via surface mycoplasmal cytadhesin (P1 protein) to sialic acid leading to interference with the ciliary action resulting in its desquamation.
  - Damage to host tissues may be due to immune response rather than invasion by the organism.
  - Organism shed in UR secretions for 2-8 days before onset of symptoms and shedding continue up to 14 weeks

Mycoplasma

- Clinical significance
  - M. pneumonia – the major cause of primary, atypical pneumonia (walking pneumonia). 10% of all pneumonia
    - Transmitted by droplet infection
    - Age range: 5-15 yrs old, more severe in older children
    - After a 2-3 week incubation, the disease begins as a mild, upper respiratory tract infection and progresses to fever, headache, malaise, and a dry cough which is usually mild and self-limited.
    - 3-10% develop clinically apparent pneumonia with occasional complications of arthritis, rashes, cardiovascular problems, or neurological problems.
  - Pharyngitis with fever and sore throat
  - Myringitis or otitis media
  - Genital tract infections - caused by *M. hominis* and *U. ureolyticum* which may also be found as part of the NF in the genital tract
  - May cause nongonococcal urethritis, PID, post-partum fever, infertility, stillbirth, spontaneous abortion, and acute urethral syndrome

Mycoplasma DX

- M. pneumoniae
  - Isolation in culture: incubation for more than a week
  - Ability of colonies to hemolyze guinea pig RBCs
  - 4 fold rise in specific antibody titer
  - Cold agglutinin test – a nonspecific test in which the patient produces cold reacting antibodies that agglutinate type O human RBCs at 4^0 C, but not at 37^0 C
    - A single titer of 1:128 is significant and occurs in 7 days and disappears in 6 weeks.
- M. hominis
  - Isolation in culture
  - No hemolysis of guinea pig RBCs
- U. urealyticum
  - Urease production

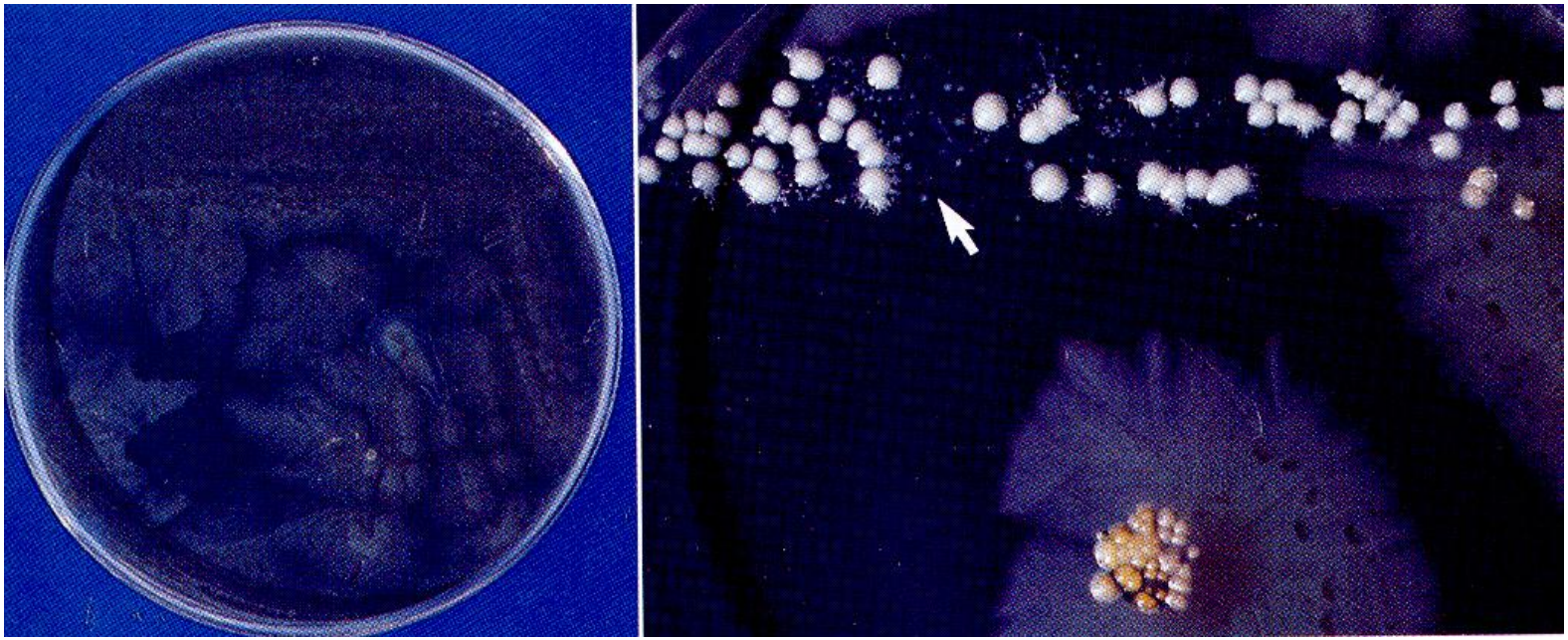
Mycoplasma

- Treatment
  - M. pneumonia – Clarythromycin and azithromycin and quinolones
  - Genital infections – **Tetracycline**, azithromycin and quinolones

Legionella

- Classification – family Legionellaceae with more than 21 species. We will only discuss *L. pneumophilia*
- Morphology/cultural characteristics
  - Small, G- pleomorphic rods that stain very poorly alternative Dieterle stain
  - Motile
  - Requires cysteine, ferric ions and PH 6.9 for growth and, therefore, won't grow on ordinary lab media
  - The best media for primary isolation is buffered charcoal yeast extract with alpha keto glutarate (BCYEα).
    - This can be made selective by the addition of cefamandole, anisomycin, and polymyxin B.

Legionella in BCYEα



Legionella

- Growth in enhanced by incubation in a candle jar or in 2.5% CO_2
- Growth might take 2-10 days
- Colonies are pinpoint with a ground-glass appearance

- Diagnosis
  - Inoculate BCYEα and CBA and look for growth versus no growth
  - Are relatively inert and nonfermenter
  - Catalase +
  - Direct fluorescent antibody testing (positive in 25-50%)
  - PCR

Legionella

- Virulence factors
  - Inhibit phagosome-lysosome fusion which allows for intracellular growth
  - Endotoxin (LPS) less toxic than other G-ve
  - Inhibit generation of bactericidal substances in phagocytic cells (peroxide)

Legionella

- Clinical significance
  - Acute pneumonia – Legionnaire's disease
    - Airborne transmission with an incubation of 2-10 days
    - Symptoms include fever, chills, malaise, myalgia, headache, dry cough, vomiting, diarrhea, and abdominal and chest pain. Hospitalization is usually required in 3-5 days.
    - Without antibiotics, the fatality rate is as high as 15%
    - The disease occurs more in males over 60 years of age and in the immunocompromised
    - No person to person transmission
    - Disease rate after exposure is low
    - The reservoir of infection is often in the cooling towers of air conditioning systems and in hot water lines as well as in soil and water

Legionella

  - Pontiac fever – an acute, self-limited febrile illness with an incubation of 24-36 hours. Symptoms include a high fever, chills, malaise, myalgia, and headache which lasts 2-5 days
  - Reaction to endotoxin or hypersensitivity to *Legionella* components
- Treatment/antimicrobic susceptibility
  - **Erythromycin**, rifampin, clarithromycin or azithromycin
- Prevention
  - Minimize production of aerosols in public places
  - Resistant to chlorine and heat