

## Micro Blk2 Review

\*\*\*NOTE: All slides shown in review came from regular class lecture slides. There are printouts of slides available to be picked up in the Micro department if you missed the review. The review was pretty much verbatim of what was on the slides with few additional comments.\*\*\*\*\*

### I. **Indigenous Flora of Humans**

#### A. **Normal Flora**

1. Beneficial vs. Harmful
  - a. harmful if it gets outside of its normal place
  - b. beneficial protection against exogenous pathogens
2. Bacterial colonization
  - a. occurs sequentially
  - b. characteristic for each animal
  - c. certain obstacles that must be overcome; bacteria can adapt to this
  - d. specific association between bacterial lectin (protein) and host receptors (glycoprotein or glycolipid)
  - e. Host specificity—certain bacteria will colonize certain species (rat, human, etc.)
  - f. Tissue specific—e.g. tongue vs. teeth

#### B. **Skin-Habitats for Colonization**

1. although appears flat, is tremendous area for colonization
2. keratin layer, sweat glands, and all components of hair follicle
3. has anaerobic bacteria such as propionibacterium

#### 4. **Staph epidermidis is normal flora in skin**

#### C. **Nose (NO-SLIDE-IN-HANDOUTS)**

1. nose has flora similar to skin with one addition
2. frequently carry pathogens in our nose, e.g. **Staph aureus** and some species of **corynebacterium**

#### D. **Oral Cavity**

1. very complex flora composed of both aerobic and anaerobic bacteria
2. dental plaque development
  - a. involves normal flora bacteria
  - b. salivary glycoprotein provides site for strep colonization
  - c. leading to synthesis of glucose polymers and additional bacterial colonization
  - d. Thus, production of organic acid which causes tooth decay and elimination of enamel

#### E. **Intestine**

1. From mouth towards stomach you see fewer and fewer bacteria
2. Stomach and upper small intestine have transient colonization (not normal flora)—pH is too low; mostly appear after meals
3. Ileum and large intestine, number of bacteria increase tremendously
  - a.  $10^{11}$  bacteria; largest bacteria site on human body
  - b. 500 different species
  - c. anaerobic bacteria outnumber aerobic bacteria by at least 100:1; some studies suggest 1,000:1

#### F. **Genitourinary Tract Normal Flora**

1. only place see normal flora is outer portion of urethra
2. upper portion of urethra and bladder are usually sterile because of flow of urine
3. urine flow interruption (such as by catheter) can lead to UTI
4. Vaginal flora is different with age
  - a. children have simple flora; high pH→more frequent UTI
  - b. puberty—hormonal changes and glycogen deposition on vaginal epithelial cells; lactobacilli and anaerobic bacteria can colonize in lower pH
  - c. women receiving anti-microbial agents can get vaginitis because of the disruption of the colonization by the normal flora
  - d. menopause—simple flora like childhood because of hormonal changes

**G. Beneficial Role of Normal Flora**

1. stimulation of immune system
2. germ free animals don't have a good immune system
3. normal flora bacteria will interfere with other bacteria that try to colonize

## II. Anti-microbial Agents

**A. Definitions**

1. Chemotherapeutic agent vs. antibiotic: **antibiotic is compound produced by one microorganism that inhibits another microorganism**; this is also true for chemotherapeutic agents except that chemotherapeutic agents can be produced in laboratory
2. Broad spectrum vs. narrow spectrum: arbitrary definition; who's to say which is what?
3. Bacteriostatic (inhibits growth) vs. bactericidal agents (kills the bacteria)
  - a. don't mix the two because in most cases bacteriocidal agents is only active if microorganism is multiplying
  - b. only exception is **POLYMIXIN**, which doesn't require bacteria to be multiplying to be bactericidal
4. Minimum Inhibitory Concentration (MIC) vs. minimum bactericidal concentration (MBC)
5. Selective toxicity—reason we can use anti-microbial agents because they target something in the bacteria cell which isn't present in eukaryotic cells

**B. Dangers and Misuse of anti-microbial agents**

1. can lead to drug resistance
2. drug toxicity
3. sensitization—can cause hypersensitivity reaction resulting from overuse of drugs
4. superinfection
5. masking of serious infection

**C. Beta Lactam Group—inhibit cell wall synthesis**

1. Need to be able to recognize structures of penicillin and cephalosporine
  - a. both contain beta lactam ring, which **MUST** remain intact
  - b. only difference is penicillin has 5-member ring while cephalosporine has 6-member ring
2. **Penicillins**: Structure
  - a. contains beta lactam ring and thiazolidine ring
  - b. can add radicals to side chain to get different penicillin compounds
  - c. natural penicillin (pen. G, V) has narrow spectrum—mostly inhibit gram positive bacteria
  - d. can inhibit gram negative with synthetic penicillin
  - e. Mechanism of Action
    1. penicillin crosses cell wall and binds to penicillin binding protein (enzymes such as transpeptidases or carboxypeptidases that function in peptidoglycan synthesis)
    2. peptidoglycan synthesis stops
    3. at the same time that it binds to the penicillin binding protein, it induces **autolysins** which eat away at the cell wall → leading to cell lysis
  - f. Side Effects
    1. **ALL ANTIMICROBIAL AGENTS HAVE GI EFFECTS!!!**—therefore, they're not listed in the slides; just the ones unique to the particular agent
    2. Penicillin shows hypersensitivity (about 5%)—very rarely result in anaphylactic reaction mediated by IgE, although possible; mostly just rashes
  - g. Resistance
    1. **beta lactamase—breaks beta lactam ring**
  - h. Tolerance
    1. same MIC but higher MBC; **penicillin is no longer bactericidal but remains bacteriostatic**
3. **Cephalosporins**
  - a. beta lactam ring and dihydrothiazine ring; two places to add side chain radicals
  - b. we are at 4<sup>th</sup> generation of cephalosporin

- c. Same mechanism of action as penicillin
- d. Same mechanism of tolerance as penicillin
  - 1. you will sometimes hear beta lactamase refer to as penicillinase (more specific for penicillin) or cephalosporinase (more specific for cephalosporin)
- e. Same side effects as penicillin
- f. Bactericidal just like penicillin

4. **Additional beta lactam**

- a. monobactams, penems and carbapenems—fairly new with lots of hope, but bacteria has already developed resistance
- b. beta lactamase inhibitors (clavulanic acid)—**don't have anti-microbial properties but can bind up beta lactamase**; used in combination with penicillin such as amoxicillin

5. **Vancomycin**

- a. bactericidal
- b. inhibits transfer of amino sugar to peptidoglycan
- c. very narrow spectrum=inhibits only G+ bacteria
- d. two mechanisms of resistance: alteration of cell wall and some bacteria becoming tolerant (low MIC, high MBC)

6. **Bactracin**

- a. bactericidal
- b. prevents attachment of amino sugars
- c. because of toxicity, **only administer orally or topically**—NOT IV
- d. side effect: nephrotoxicity

7. **Cycloserine**

- a. inhibits incorporation of D-alanine into peptidoglycan
- b. only used in treatment of **Mycobacterium tuberculosis**
- c. Remember: agents used to treat TB are always used in combination
- d. Side effect: CNS toxicity
- e. Only use when MT doesn't respond to something else; used as second line compound

**D. Protein Synthesis Inhibitors—directed against ribosomes**

1. **Aminoglycosides—aminocyclitols**

- a. irreversibly binds to 30S ribosomal subunit
- b. bactericidal
- c. poorly absorbed; not given orally if expect to get in bloodstream
- d. narrow spectrum: only G-negative bacteria
- e. side effects: ototoxicity and nephrotoxicity
- f. **therapeutic and toxic dose is very close**; must monitor patient closely for renal failure
- g. mechanism of resistance: mainly through **enzymatic modification** of acetyltransferase, phosphotransferase, and nucleotidyltransferase but also through blocking transportation and alteration of binding sites
- h. if bacteria is resistant to one aminoglycoside because of enzymatic modification, you may still be able to use another aminoglycoside, i.e. just because bacteria is resistant to Gentamicin doesn't mean it's resistant to all other aminoglycosides

2. **Tetracyclines**

- a. **reversible binding to 30S ribosomal subunit**
- b. bacteriostatic
- c. broad spectrum
- d. side effect: **discoloration of teeth or developing bones**
- e. Contraindicated in pregnancy and children less than 8-year-old
- f. Mechanism of resistance: less permeable

3. **Chloramphenicol**

- a. **reversible binding to 50S ribosomal subunit**
- b. bacteriostatic
- c. broad spectrum
- d. can be administered orally
- e. principle side effect: **aplastic anemia** (universally fatal)—1/10,000 to 1/20,000
- f. mechanism of resistance: altered cell wall

4. **Macrolides**
    - a. only one used is Erythromycin
    - b. binds 50S ribosomal subunit**
    - c. bacteriostatic or bactericidal
    - d. broad spectrum
    - e. side effects: GI
    - f. mechanisms of resistance: cross resistance with lincosamide, i.e. if you have a patient who is resistant to erythromycin, you don't want to switch them to clindamycin—chances are the microorganism will also be resistant to clindamycin
  5. **Lincosamides**
    - a. only one used therapeutically is clindamycin
    - b. binds to 50S ribosomal subunit
    - c. bacteriostatic or bactericidal
    - d. very narrow spectrum—only used in treating anaerobic bacteria
    - e. unknown mechanisms of resistance
  6. **Streptogramin**
    - a. Synercid—combination of two streptogramin
    - b. Broad spectrum
    - c. Used in **vancomycin-resistant enterococcus faecalis, methicillin-resistant staphylococcus, and penicillin-resistant pneumococcus**
- E. Metabolic Pathways Inhibitors—particularly folic acid pathway which eventually leads to synthesis of purines and pyrimidines**
1. **Sulfonamides**
    - a. competitively inhibit the conversion of p-aminobenzoic acid (PABA) to dihydrofolic acid structure is very similar to PABA
    - b. bacteriostatic; however, when **combined with trimethoprim they become bactericidal**
    - c. many sulfonamides available in the market
    - e. side effects: fairly safe compound, few with hypersensitivity and some blood abnormalities, but they reverse themselves when medication is discontinued
    - f. mechanism of resistance: if you know mechanism of action, you can almost guess mechanism of resistance
      1. overproduction of PABA
      2. alteration of tetrahydropteroyl synthetase so that it will recognize PABA better than sulfonamides
      3. microorganism can mutate to become folic acid dependent—normally bacteria cannot produce folic acid but must take in from environment (humans do produce own folic acid)
      4. decreased permeability
  2. **Sulfones**
    - a. only one used is diaminodiphenyl-sulfone (DDS) for treatment of leprosy
    - b. growing concern is that Mycobacterium lepre is becoming resistant to the compound
    - c. unknown mechanism of resistance; probably same as sulfonamides
  3. **Trimethoprim**
    - a. Primarily used in **combination with sulfamethoxazole=cotrimoxazole** (Bactrim)
    - b. Mix two bacteriostatic to yield bactericidal agent**
    - c. By itself, it is used to treat **UTI and traveler's diarrhea**
    - d. Side effects and mechanism of resistance are the same as sulfonamides except it's based on different enzyme (one step further in the folic acid pathway)
- F. Nucleic Acid Synthesis Inhibitors**
1. **Metronidazole**
    - a. mechanism of action: through reductive process produces very short lived, yet very toxic intermediates; kills cell by creating nicks in DNA
    - b. narrow spectrum: only effective against anaerobic bacteria or protozoa since it has to be reduced to be active
    - c. mutagenic and carcinogenic (never demonstrated in humans)

- d. cause peripheral neuropathy that is reversible when drug is discontinued
  - e. bacteria become resistant by no longer able to reduce metronidazole
2. **Rifamycin**
- a. compound used is called Rifampin in US (elsewhere is called Rifampicin)
  - b. inhibits DNA-dependent RNA polymerase**
  - c. broad spectrum but bacterial resistance develop so quick that it's not used to treat bacterial infection
  - d. used to treat Mycobacterium tuberculosis
  - e. side effects: skin rashes and reversible thrombocytopenia

3. **Nalidixic Acid**

- a. inhibits bacterial DNA gyrase**
- b. narrow spectrum; inhibits G-negative bacteria
- c. high concentration can be used to treat UTI
- d. its derivatives form quinolones

4. **Quinolone**

- a. derivatives of nalidixic acid**
- b. same mechanism of action
- c. at least 100X more active than nalidixic acid against bacteria
- d. mechanism of resistance include alteration of bacterial DNA gyrase, alteration of outer membrane proteins and development of efflux pump that can pump out as much quinolone as they are taking in

**G. Inhibits Cell Membrane—only one class**

1. **Polymixin**

- a. many type but all too toxic except for B & E
- b. function as cationic detergent**
- c. breaks apart lipids and cell membrane causing cell lysis
- d. only compound that is bactericidal even when organism is not multiplying**
- e. very narrow spectrum against G-negative bacteria
- f. only used as topical ointment because of high toxicity; neurotoxicity and nephrotoxicity

**H. Unknown Mechanism of Action**

- 1. **Isoniazid**—used to treat Mycobacterium tuberculosis
- 2. **Nitrofurans**—believed to inhibit oxidation of pyruvic acid
  - a. broad spectrum
  - b. used mostly as topical ointment although can also be used to treat UTI
- 3. **Ethambutol**—another anti-Mycobacterium tuberculosis agent

**I. Anti-microbial Susceptibility Testing**

- 1. Quantitative Test→gives specific MIC values
- 2. Semi-quantitative procedures—tell whether organism is very resistant, resistant, very susceptible, or susceptible
- 3. Qualitative procedures—classify organism as resistant or sensitive

**IV. Enterics**

A. **Enteric Bacteria**—normally in intestinal tract; cause trouble if gets out

- 1. G-negative rods
- 2. Facultative
- 3. Non-sporulating
- 4. Most belong to family Enterobacteriaceae
- 5. antigenicity can be divided into:
  - a. O-antigen—based on lipopolysaccharides
  - b. K-antigen—based on capsular antigen
  - c. H-antigen—based on flagella
  - d. Pili/fimbriae
- 6. **toxicity is due to endotoxin (lipid A part)**
- 7. **Coliform Organisms**

- a. include *E. coli*, *Klebsiella* species, *Citrobacter* species, *Enterobacter* species, and *Serratia* species
  - b. Urease producing species: *proteus mirabilis*, *proteus vulgaris*, *morganella morganii*, and *providencia rettgeri*
    1. neutralizes pH by breaking down urea to form ammonia
    2. recall, this is important in the pathogenesis of *helicobacter*
- 8. Pseudomonas aeruginosa**
- a. enteric bacteria but family is pseudomonadaceae
  - b. found everywhere; frequent contaminant of hospitals
  - c. does not ferment carbohydrates
  - d. produced variety of virulence factor; focus on exotoxin A which ribosylates elongation factor 2 (EF2)→shuts down protein synthesis
- 9. Three types of Intestinal Infections caused by Enteric Pathogens**
- a. enteric pathogens are gram negative bacteria we don't normally harbor in our intestinal tract
  - b. these include *Vibrio cholerae* and *E. coli*
    - 1. Vibrio Cholerae**
      - a. comma shaped
      - b. motile
      - c. gram negative rods
      - d. produces acid but no gas from carbohydrates
      - e. grows at high pH
      - f. no animal reservoir—only colonizes humans
      - g. waterborne transmission—from human to human through contaminated water supply
      - h. can be divided into several groups based on O-antigens or biotypes (El Tor and classical) and serotypes
      - i. attaches to small intestine
      - j. produces enterotoxin with A region (attachment) and B region (binding) causes overproduction of cAMP→massive outflow of fluid into intestine without reabsorption
      - k. no damage to intestine; just treat with fluid replacement although antibiotics can be administered to prevent carriers of *V. cholerae*
    - 2. Enterotoxigenic E. coli (ETEC)**
      - a. very similar to *V. cholerae*; produces **heat labile enterotoxin (HLE)** with identical mechanism of action as *V. cholerae*
      - b. also produce heat stable enterotoxin which stimulates guanylate cyclase activity→same result as increase in cAMP
      - c. only difference from *V. cholerae* is that ETEC has colonization factor antigen—which allows the organism to attach to intestinal epithelial cells
    - 3. Enteropathogenic E. coli (EPEC)**
      - a. Invades the cell; no inflammation
      - b. infantile diarrhea in bottle fed infants
      - c. adherence to enterocytes in small bowel
      - d. invasion and cytotoxin production→damage to small bowel
- 10. Enteric Pathogens that invade Colon Tissue**
- a. do not enter bloodstream
  - b. **Shigella species**
    - 1. Bacillary dysentery only occurs in primates**
      - a. species causing most dysentery in US is *S. sonnei*
      - b. **does not produce hydrogen sulfide (H<sub>2</sub>S)**—use this to distinguish from salmonella

- c. does not ferment lactose
  - d. ferment glucose with production of acid but no gas
  - e. transmitted by food and water
  - f. see this mostly in **institutional outbreaks** (hospitals, daycares, etc.)
  - g. **Only needs 10 microorganisms to cause infection**—easily transmitted between individuals
  - h. Produce **shiga toxin that inhibits protein synthesis** and has cytotoxic, neurotoxic, and enterotoxic activities
  - i. Can penetrate large epithelial cells and produce massive inflammatory response
  - j. **Classical bacillary dysentery** **bloody diarrhea**
  - k. Treatment is **fluid replacement**
  - l. Although it has activity in small and large intestine, it has no entry into the bloodstream
  - m. Student Question: So how does it have neurotoxicity?
  - n. Answer: it's the toxin that is absorbed not the organism itself.
- c. **Enteroinvasive E. coli (EIEC)**
- 1. produce Shigella like disease
  - 2. invasion of colonic epithelial cells
  - 3. don't see the shiga toxin production so **no small intestine diarrheal disease**
- d. **Vibrio parahemolyticus**
- 1. see lots in gulf coast; halophilic marine organism—likes high salt concentration
  - 2. contaminates **raw oysters and insufficiently cooked shellfish**
  - 3. produces **watery diarrhea** and sometimes produce shigella like symptoms
- e. **Enterohemorrhagic E. coli (EHEC)**
- 1. produced by **E. coli O157: H7**
  - 2. normal flora in cows, easily contaminates hamburger meat (Jack in the Box!)
  - 3. produces shiga toxin like called **verotoxin** that cause **hemorrhages in the colon**
  - 4. in children, see primarily **Hemolytic Uremic Syndrome (HUS)**—acute renal failure, anemia, and thrombocytopenia
  - 5. very hard to reverse once see HUS in children

## 11. Enteric Pathogens that Produce Systemic Infections—enters the bloodstream

- a. **Salmonella species**
- 1. does not ferment lactose
  - 2. **produce H<sub>2</sub>S**
  - 3. antigenic structure includes somatic (O) antigen, Flagella (H) antigen, and Capsular or K-antigen (referred to as Vi antigen)
  - 4. transmitted by contaminated food and water or animal and human carriers
  - 5. frequent contaminant of raw eggs
  - 6. three clinical manifestations:
    - a. **gastroenteritis** (food poisoning)—doesn't enter bloodstream
      - ingestion of contaminated food or water
      - adherence to epithelial cells to lamina propria
      - PMN leukocytes response→inflammation
      - Enterotoxin production
    - b. **enteric fever**
      - ingestion of contaminated food or water
      - organism invades epithelium cells of distal ileum and colon
      - multiply within macrophages
      - lysis the macrophages and enters the bloodstream
      - primary bacteremia (no gastroenteritis symptoms at this point)
      - multiply within fixed macrophages
      - secondary bacteremia and localize in various organs

- when localize in gall bladder, bacteria enters intestine via infected bile (now see gastroenteritis symptoms)
- early in the disease process, you can isolate organism from blood, but late in the process, you can isolate it from intestinal tract
- c. **septicemia**
  - same as enteric fever but doesn't localize in intestinal tract
  - can't diagnose this disease by looking at intestinal tract; must have blood specimen
  - vaccines available: oral, heat-phenol inactivated *Sal. typhi*, and capsular polysaccharide (Vi antigen)
  - Treatment is **fluid replacement**; use antibiotic for enteric fever and septicemia; don't need antibiotic for food poisoning—body's own response will get rid of it
- c. **Campylobacter jejuni**
  1. curved bacilli under the microscope
  2. **microaerophilic**—requires reduced O<sub>2</sub> concentration for growth
  3. ingestion of contaminated food or water
  4. organisms invade small and large intestine leading to abscess formation and enterotoxin formation
  5. can be isolated from bloodstream
  6. incubate at high temperature
- d. **Helicobacter pylori**
  1. idealogic agent in chronic gastritis and ulcers
  2. colonizes by **urease production** which neutralizes acid
  3. can cause depletion of gastric mucus resulting in gastric symptoms
  4. treatment by three **substance bismuth (pepto bismol), metronidazole, and amoxicillin or tetracycline**
- e. **Yersinia enterocolitica**
  1. not much said other than enteric disease similar to Camp. Jejuni

## V. Neisseria

- A. Pathogenic—fastidious nutrition; increased CO<sub>2</sub> for growth; grow best at body temperature: gram negative cocci; Catalase and oxidase positive (determine genus); Carbohydrate fermentation (determines species)
1. **N. gonorrhoeae**
    - a. only occurs in human; primarily by sexual transmission
    - b. virulence factors include endotoxin (cause the cellular damage), pili, IgA protease (allows survival in intestinal mucosa) and capsule
    - c. most males are symptomatic by urethral discharge; most female are asymptomatic
    - d. principle complication in untreated male is urethral stricture, in women is sterility, infertility
    - e. few cases the organism will enter bloodstream (disseminated gonococcal infection—DGI)
      1. clinical manifestation of this was destructive arthritis and dermatitis
      2. lesions primarily on extremities, i.e. hands and feet
    - f. **Gonococcal Ophthalmia Neonatorum**—infection the neonate as it passes through the birth canal
      1. Required by law that ointment (silver nitrate, erythromycin or tetracycline) be placed in infant's eyes to prevent gonococcal infection as well as chlamydia
    - g. can cause conjunctivitis in adults and vulvovaginitis in young girls before puberty (after puberty, vaginal cells are resistant to gonococcus)
    - h. diagnosis: oxidase positive, gram negative, intracellular, diplococci from urethral discharge of male; gram stain is not helpful in female—too many gram negative diplococci part of normal flora
    - i. **chocolate agar and Thayer Martin mediums (VCN)** are selective medium
    - j. treatment is antibiotic: whenever we treat gonococcus we also treat chlamydia since both tend to occur together and chlamydia is sometimes hard to diagnose; chemoprophylaxis is also possible
  - B. **N. meningitidis**
    1. predisposing factor is overcrowding
    2. susceptible individuals have to come in contact with a carrier

3. occurs sporadically in US although epidemics still occur worldwide
  4. transmitted by **upper respiratory tract secretions**
  5. virulence factors
    - a. **lipopolysaccharide (endotoxin)** is responsible for most of the clinical manifestation
  6. we divide organism into different groups based on capsular polysaccharide
  7. it's antibody against the capsular polysaccharide that protects us from the disease; lifetime immunity
  8. clinical manifestation
    - a. organism colonizes upper respiratory tract
    - b. develop upper respiratory tract infection
    - c. sometimes they enter the bloodstream and cause meningitis and arthritis
    - d. cause rash lesions primarily on trunk at site of entry into the blood stream
  9. diagnosis is very similar to gonococcal, using same media; got the diagnosis if gram negative intracellular diplococci in CSF
  10. treatment is penicillin; chemoprophylaxis is Rifampin; vaccine available that is being used for college students—only problem with vaccine is there is no Group B polysaccharide
- C. **Moraxella**—can cause **otitis media in children**
- D. **Acinetobacter calcoaceticus**—normal flora; hospital acquired pneumonia
- E. **Kingella kingae**—normal flora; endocarditis and arthritis
- F. **non-pathogenic** can also cause diseases
1. opposite characteristics of pathogenics (Dr. Rolfe said to refer to notes for details and summary)

## VI. Haemophilus

- A. Normal flora
- B. Small, gram negative bacilli, non-motile, non-sporeforming
- C. Not important to know which species use what factors, but it is important to know that we determine the species is by the requirement of either the X—factor (Hematin) or the V—factor (NAD)
- D. Primarily in children; **not common in children less than 6 months**—this is the difference from Bordetella because protection against Bordetella is primarily cell mediated (doesn't cross the placenta)
- E. Transmitted by **upper respiratory tract secretions**
- F. Virulence factors: polysaccharides capsule, IgA protease; **antibody against polysaccharide capsule provides immunity**
- G. Nasopharyngeal colonization, local infection (most stop here)
- H. Main complication is when it can gain entry into bloodstream to cause meningitis and makes us susceptible to secondary viral infection
- I. Diagnosis: isolate through conventional technique
- J. Treatment: not important to remember; now exist polysaccharide vaccine that is required for all kids in US; **based on capsular serogroup B**; passive immunization is also available
- K. **Haemophilus influenzae biogroup aegyptius** causes **conjunctivitis (pink eye) and Brazilian purpuric fever (more severe)**—can occur in epidemic proportions in elementary schools
- L. **Haemophilus ducreyi**—sexually transmitted disease; **gets chancre at site of inoculation**; swollen lymph nodes
- M. **Gardinerella vaginalis**—maybe sexually transmitted; **causes female genital tract infections and neonatal sepsis**; diagnosis is through vaginal swab for clue cells—just **epithelial cells with lots of gram negative bacteria attached**
- N. Other Haemophilus species—most are part of normal oral flora

## VII. Spirochetes

- A. four genre: **Treponema, Borrelia, Leptospira, and Spirillum**
- B. only **Borrelia and Spirillum take up stain** (Giemsa or Wright stains) for visualization
- C. can visualize all four genre without staining by using **silver impregnation or darkfield microscopy**
- D. none can be cultured in vitro except for leptospira which can also survive very long outside host
- E. **Treponema pallidum**—“**the Great Imitator**”
  1. primarily sexually transmitted disease or congenitally acquired as well as blood transfusion or direct inoculation

2. after initial penetration of mucous membrane or skin there's an **incubation period**
  3. first clinical manifestation is development of **hard chancre**; in male is easily recognized on external genitalia; unrecognized in female because usually on cervix and is painless; loaded with *Treponema pallidum*
  4. **enters primary latent stage** without any manifestation before entering **secondary stage** (stage of systemic involvement)—can develop skin and mucous membrane lesions (full of *Trep. Pallidum*)—highly infectious
  5. patient then enters a **latent period**—some can relapse between secondary stage until can no longer relapse
  6. then enters the **tertiary stage** or late syphilis; some stays in latent period for life, others go into tertiary stage where the organism localizes in neuro and cardio tissue
  7. some will go through all stages without any symptoms and then suddenly develops tertiary stage symptoms
  8. **late benign syphilis** is host response to *Treponema pallidum*→**granulomatous tissue formation**
  9. **Congenital Syphilis**—infant acquired from passage through birth canal; can be free of disease, still born, neonatal disease or death
  10. Clinical manifestations not much help in diagnosis of syphilis, especially towards the late stages; cannot isolate organism, but can directly demonstrate organism on chancre
  11. Mouth lesions are not helpful at all in diagnosis because too many spirochetes as part of normal flora
  12. **Non-specific test** is initial test used to screen individual for syphilis—**too many false positives**; as patient is treated for syphilis, non-specific antibody titer will decrease, thus, allowing one the ability to follow the course of the disease or use it to diagnose congenital syphilis
  13. Non-specific test positives are retested with **specific test** that has antibody specific for the *Treponema* (no false positives)
  14. Treatment is **penicillin**
  15. Secondary or tertiary stage syphilis can develop **Jarisch-Herxheimer Reaction**—penicillin causes lysis of spirochetes, releasing endotoxin to cause fever—condition is better once endotoxin is removed—should expect this reaction to occur and should continue treatment
- F. Non-venereal Treponematoses**
1. immunologically and morphologically the same as *treponema pallidum*
  2. treated with penicillin
  3. diagnose through clinical manifestation
  4. principle one seen in this part of country is PINTA, which causes depigmentation of skin
- G. Relapsing Fever**—primarily tick born disease in US; can also be louse born
1. incubation period, followed by fever, non-fever, fever, non-fever relapses
  2. relapse is due to organism's **ability to change antigenic structure**
  3. diagnosis: demonstration of *Borrelia* in blood during time of fever
  4. **louse-born relapsing fever (not seen in US)**
    - a. ***Borrelia recurrentis***
    - b. Occurs in epidemics
    - c. Transmitted by human body louse
- H. Lyme Disease--#1 tick born disease in US**
1. ***Borrelia burgdorferi***
  2. **Classical bulls-eye lesion** at site of inoculation
  3. after certain period of time develop flu like symptoms, ache all over, feel terrible
  4. symptoms can go away and enter second stage where bacteria can cause arthritis or cardiac or neurological problems
  5. symptoms may go away and enter tertiary stage, but by they patient will suffer mental loss and cardiac damage
  6. can be diagnosed by clinical criteria if patient is in situation where they were likely exposed to tick and develop characteristic primary lesions; however, most patients won't develop those characteristic lesions; clinical criteria can become difficult—good serologic tests available
  7. treatments can be difficult in long term—usually treated for weeks or months
- I. Fusospirochetosis**—symbiotic disease; **involves both *Borrelia vincentii* and *Fusobacterium* species**—both are part of normal oral flora

1. result from poor nutrition and injury to mouth
2. can cause **ulceromembranous stomatitis**—ultimate form of pseudomembrane in back of throat or **ulcerative gingivostomatitis** (“trench mouth”)—inflammation of the gums
3. diagnosis: stain smears, look for cigar shaped Fusobacterium and spiral shaped Borrelia
4. treatment is antibiotic to lower the level of organisms and correct the underlying condition

**J. Leptospirosis**

1. **Leptospira interrogans** (causes human diseases) and **Leptospira biflexa** (occurs in nature—doesn't cause human diseases)
2. Transmitted from direct contact of infected tissue or drinking contaminated water
3. Clinical manifestation depends on where the organism localizes—conjunctivitis or rash
4. Severe when localization is in kidneys or liver—called icteric leptospirosis—very high fatality rate
5. Early in the disease look in blood; late look in urine
6. Can be isolated using conventional media like the other spirochetes; serologic tests are also available

**K. Rat Bite Fevers**

1. spirillum minor is a very small spirochete; streptobacillus moniliformis is gram negative bacillus
2. epidemiologically, they both occur in oral cavity of rodents, primarily rats
3. spirillum minor is an actual infection whereas moniliformis is part of normal flora
4. site of bite becomes inflamed; patient develops soft tissue infection; organism can infect bone
5. both treated with penicillin

## VIII. Yersinia, Francisella, and Pasteurella

- A. **bipolar staining**; animal pathogens transmitted accidentally to human; all members of family enterobacteriaceae
- B. **Yersinia pestis** prefers **temperature below body temperature**
1. rat and rat flea are primary reservoir; in US rodents are reservoir
  2. transmission is direct handling of infected tissue, interruption of rodent flea rodent cycle, inhalation, or laboratory acquired
  3. produce **Murine toxin** that acts on peripheral vascular system; **endotoxin is responsible for many of clinical manifestations**
  4. from initial site of inoculation of organism into the host, it travels to regional lymph nodes and cause them to swell → Bubonic plague
  5. organism sits in lymph node and produce endotoxins and destroys human tissue—**50% survival rate among untreated Bubonic plague patients**
  6. in untreated individuals, organisms can enter blood stream and infect any organ (**septicemic plague**)—**fatality is 100%**
  7. one of the organs is the lung (**pneumonic plague**)—primary is when you inhale the organism from another infected individual; secondary is when organism gains entrance through blood stream—**100% fatality** but very infectious before death
  8. Diagnosis: gram stain of aspirate of swollen lymph nodes; look for bipolar staining; easily isolated
  9. Treatment is antimicrobial agents; vaccine available but mostly used for laboratory workers and those going into areas where plague is a problem (e.g. outbreak in India couple of years ago)
- C. **Francisella tularensis—reservoir is the hare (rabbit)**
1. primarily seen in hunters
  2. transmitted by direct handling of infected tissues, animal bites, blood sucking arthropods, or ingestion
  3. requires special media for growth; must have **increased sulfhydryl compounds (cysteine-glucose blood agar)**
  4. vaccine available but not used for general public
- D. **Pasteurella multocida**
1. reservoir is mainly **domesticated animals** (cattles)
  2. transmitted by animal bites, non-bite animal exposure
  3. soft-tissue infection; organism can enter blood stream (hemorrhagic septicemia) or bone

4. treatment takes a very long time

## IX. Bordetella

- A. **Bordetella pertussis—whooping cough**
- B. Bordetella parapertussis—less severe whooping cough
- C. Bordetella bronchiseptica—primarily animal pathogen transmitted by upper respiratory secretions to human; in **children and newborns even those younger than 6 months**
- D. Virulence factors:
  1. **filamentous hemagglutinin for binding**
  2. **tracheal cytotoxin**—destroys epithelial cells on trachea
- E. pathogenesis:
  1. incubation period—develops cough, mild upper respiratory infection
  2. **catarrhal, prodromal, preparoxysmal stage—most infectious**, children still can go to school and contaminate everyone with this
  3. paroxysmal, spasmodic stage—less infectious; deep whooping cough; stay home
  4. convalescent stage—takes a long time; cough becomes less and less severe
- F. Clinical complications is secondary infections due to destruction of epithelial cells
- G. Isolate on **Bordet-Gengou medium**
- H. Treatment is antimicrobial; not much effects on organism but for treatment of secondary infection
- I. **DTP vaccine**

## X. Brucella

- A. **B. Suis found in swine**
- B. Displays **viscerotropism**—infection is localized in specific tissue; for B. suis it is **animal placenta**, which contains **high levels of erythritol**—therefore, frequent cause of abortion and massive contaminant of environment
- C. Transmission is handling of infected tissues, ingestion, inhalation, conjunctiva, laboratory acquired—90% seen in slaughter house workers, 10% due to ingestion of unpasteurized milk
- D. Virulence factors: **lipopolysaccharides and capsule**
- E. Can multiply inside PMN; sits in lymph nodes and multiply; can invade bloodstream and getting into **reticuloendothelial system where it forms granulomatous lesions**
- F. **Strain 19 disease**—strain 19 is used to immunize animals; if accidental needle prick occurs, can cause debilitating diseases—make you feel bad for a very long time
- G. Identify by **Basic Fuchsin and Thionine, H<sub>2</sub>S production and Urease production**