

Unit 5 Lecture 12

Gram-Negative Bacilli

Enteric gram negative bacilli make up most of the isolates encountered in the clinical setting. Biochemical testing is necessary to identify the various organisms. Some organisms are true pathogens whereas others are opportunistic pathogens. Only a few of the enterobacteriaceae are presented.

To belong to the family Enterobacteriaceae, the organism must exhibit the following characteristics:

- [Gram negative bacilli](#)
- Ferment glucose
- Facultative anaerobes
- Reduce Nitrates to Nitrites
- Oxidase negative

Escherichia coli is a gram negative bacillus with peritrichous flagella. It is a facultative anaerobe easily cultured on most media. It can be identified on differential EMB and MacConkey media. Even though it is primarily a non-pathogen in the GI tract, certain strains cause gastroenteritis.

- ETEC (Enterotoxigenic *E. coli*): childhood diarrhea and traveler's diarrhea with virulence factor consisting of enterotoxins that stimulate hypersecretion of fluids and electrolytes.
- EIEC (Enteroinvasive *E. coli*): causes a dysentery resembling shigellosis whereby the organism penetrates the villi of the large intestine.
- EPEC (Enteropathogenic *E. coli*): causes infantile diarrhea in developing countries. Disease is primarily found in places where people have low income and poor living conditions. Children have chronic diarrhea.
- EHEC (Enterohemorrhagic *E. coli*): causes hemorrhagic enteritis which may lead to Hemolytic Uremic Syndrome (HUS); Strain *E. coli* O157:H7 and others produce a shiga-like toxin (virulence factor).

Other infections may have their origin in the GI tract, especially cystitis and pyelonephritis of the urinary tract, cholecystitis of the gall bladder, pneumonia, peritonitis in the abdominal cavity, septicemia, and neonatal meningitis. It is the most common case of nosocomial infections via the transfer of normal flora. Treatment must be based on the results of a susceptibility test because of much resistance now found in isolates. Most GI cases are self-limiting requiring only supportive care. There are no vaccines.

Klebsiella pneumoniae is a nonmotile gram negative diplobacilli. Many strains are mucoid because of its ability to produce a capsule. *Klebsiella* is a facultative anaerobe that is easily cultured. It is normal flora (GI tract). *Klebsiella* causes the second most number of cases of bacterial pneumonia

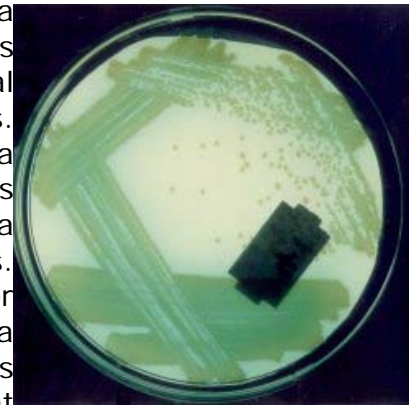
with extensive lung consolidation and high mortality rate without treatment and is important as a nosocomial infection. Transmission is via respiratory droplet. Treatment is often difficult with much antibiotic resistance and is dependent on the site of infection. Vaccines are not available.

Salmonella typhi is a true pathogen. It causes typhoid (enteric) fever. Although rare in the U.S., this organism is estimated to cause 21 million cases worldwide with about 200,000 deaths annually. This motile gram-negative bacillus is identified by biochemical testing. The POE is ingestion into the GI tract. After a two week incubation period invasion of the bloodstream (cultures positive early) occurs and is spread to various organs (liver, spleen, etc.), finally getting back to the small intestine. Endotoxins cause constipation and/or diarrhea. Organisms invade and multiply in lymph tissues. One can find organisms in blood, sputum (from lungs), urine (from kidneys), and in the spleen and liver. Hemorrhages in GI and lungs cause death from complications. Transmission is via poor sanitation and contact with any body secretion and contaminated water. Patients can develop into carrier state with the organism localizing in the gallbladder. Treatment is usually a third generation cephalosporin, chloramphenicol, or ampicillin. There is a vaccine available for those going to endemic areas.

Infections with other *Salmonella* species (salmonellosis) usually vary from mild typhoid to severe food poisoning. This gram-negative bacillus has peritrichous flagella. There are over two thousand strains based on H and O antigens (Kaufmann-White schema). The O antigens are found on the cell wall and the H antigens are found in the flagella. Septicemia can occur. Transmission is fecal/oral route. Sources especially include poultry, eggs, and dairy products. Reptiles and amphibians are reservoirs. Treatment with broad spectrum antibiotics should be done only if septicemic. The disease is self-limiting in most cases. There are an estimated 1.4 million cases per year in the U.S. Most cases go unreported.

Shigella species cause bacterial dysentery, a diarrheal disease. Shigellas are nonmotile gram negative bacilli, facultative anaerobes. Of the four species, *S. sonnei* and *S. flexneri* are most common in this country. The POE is GI with adherence and invasion of epithelial cells of large intestine. Symptoms usually appear after a 1-4 day incubation period. An exotoxin (Shiga toxin) elicits symptoms of ulceration and dysentery. An endotoxin causes abdominal pain, cramps, and fever. Septicemia is rare. Neurotoxins accumulate in CNS. Transmission is due to poor sanitation (food, fingers, feces and flies). Treatment is with ampicillin, TMP-SXT, or a fluoroquinolones. In the U. S. only 22,500 cases of the estimated 450,000 annual cases are reported. Worldwide, there are an estimated 150 million cases per year.

Pseudomonas aeruginosa is a motile (single polar flagella) [gram-negative bacillus](#) that is oxidase positive and a strict aerobe. They are easily cultured on most media and produce a variety of pigments (pyocyanin: blue-green) and odors. Pseudomonades are associated with a wide variety of primary infections. Cutaneous lesions occur via breaks in skin, cuts, and surgical wounds. Burns account for 25% incidence of cases. If a burn patient develops an infection, there is a 50% chance of mortality. Pulmonary infections occur via the respiratory tract. This organism has a major impact in cystic fibrosis patients. *Pseudomonas aeruginosa* causes UTI in hospital or nursing home patients, ulcerated corneas via contact lenses, outer ear infections (swimmer's ear), and rare meningitis infections. If the patient is immunocompromised and develops a septicemia, there is a 85% mortality rate. There has been a relative increase in number of infections, superinfections, and nosocomial infections since the introduction of antimicrobics. Transmission occurs from soil or water (hot tubs). Pseudomonas are ubiquitous, especially in hospital environments. Nosocomial transmission occurs because of poor techniques and the organism's ability to survive due to resistance to many disinfectants and antibiotics. Treatment involves use of semisynthetic penicillin used synergistically with aminoglycosides. A polyvalent (7) vaccine is used for burn patients.



Yersinia pestis is a nonmotile, gram-negative, pleomorphic bacillus that grows best on a blood based media. *Y. pestis* is the causative agent of plague, a disease that has ravaged civilization many times over in the past two millennia. The Plague of Justinian (540-590), Black Death (1346-1360) where one-third of the population (25 million) of Europe died, and the Great plague of London (1665-1666) are just a few of the major plagues that have occurred. If you think that plague is gone, you are mistaken. An outbreak in Surat, India in 1994 killed one hundred. Primary infections are of two kinds, cutaneous and pulmonary. In cutaneous infections the POE requires few organisms, uses rat flea (*Xenopsylla*) as vector, resides in lymph nodes to create [buboes](#), and is known as the bubonic plague. In pulmonary infections the organisms inhaled, the lungs become infected, and the disease is called the pneumonic plague. The primary means of transfer is human-to-human via droplets. In some cases the blood is invaded from either plague resulting in subcutaneous hemorrhages causing "black death" and is called "[septicemic plague](#)." Prevention involves rodent controls and vector (flea) controls. Treatment consists of streptomycin and tetracycline. There is a 90% mortality rate in untreated cases. A vaccine of killed organisms is available for travelers to highly endemic areas. This organism has been used in germ warfare (Japan v. China in the 1930s) and is considered by many to be the ideal organism for biological warfare because of its ability for human-to-human spread.

Yersinia enterocolitica causes enteritis (mesenteric lymphadenitis) which can mimic appendicitis

Francisella tularensis is a fastidious, nonmotile, pleomorphic, gram-negative rods that is an obligate aerobe. Because it is fastidious it grows best on Chocolate agar. *Francisella* is the causative agent of tularemia (rabbit fever). The incubation period is 3 hours to three weeks. Lesions and inflammation mark the POE. Systemic/pulmonary involvement results in a 10% fatality rate without treatment. POE is through the skin or mucous membrane causing an ulceroglandular or oculoglandular lesion. *Francisella* is one of a few organisms having the capability to penetrate intact skin. Other POE occurs through the bite of ticks, biting flies, mites and mosquitoes. Ingestion and inhalation via water, dust, undercooked meat, etc. lead to typhoidal or pulmonary tularemia. As few as ten organisms are needed to cause infection. There is no human-to-human transmission. Treatment is with intracellular antibiotics. An attenuated vaccine is available for those in susceptible careers.

Another zoonosis is caused by *Brucella*. These tiny gram negative coccobacilli are oxidase positive and very fastidious organisms. They cause Brucellosis which has various names depending on the infected species by one of four causative agents (Bang's disease, Malta fever, undulant fever). *Brucella* can cause spontaneous abortion and miscarriage in infected animals. The disease is acquired primarily through ingestion of unpasteurized milk or cheese and causes a fluctuating fever pattern. This organism also is a cause of laboratory infection through the inhalation of the microbe. Both *Brucella* and *Francisella* are potential agents of biological warfare and bioterrorism.

Haemophilus influenzae is a gram-negative coccobacillus. It produces a capsule that serves as a virulence factor. *Haemophilus* is difficult to culture and requires enriched media with blood. Specifically it needs [factors X \(Hemin\) and V \(NAD\)](#). *Haemophilus* causes infection of respiratory system, a mild respiratory infection in children, otitis media, and the highly contagious pink eye. In unvaccinated children under five it can cause acute bacterial meningitis or an acute epiglottitis. Both diseases usually are caused by the type b strain. In untreated meningitis there is a 90% fatality rate and a 33% residual impairment rate even in treated cases. Rarely are infections found in adults, but when found usually cause a bacterial pneumonia. In most people nontypeable *Haemophilus* is part of the respiratory normal flora. Transmission comes from carriers. Treatment is with ampicillin, chloramphenicol, or a third generation cephalosporin. A [vaccine \(Hib\)](#) was licensed in 1986 and since then the number of infections has decreased dramatically.



Another childhood disease that has been brought under control through vaccination is pertussis which is caused by *Bordetella pertussis*. However, since immunity wanes over time, more cases are seen in older children and

adults. This highly contagious gram-negative coccobacillus is a strict aerobe and is very difficult to culture. It requires special media (Regan-Lowe). Because of the difficulty and length of time it takes to recover this organism by cultural means, PCR is now the recommended approach to diagnosis. The organism produces a number of virulence factors; fimbriae, pertussis toxin, and tracheal cytotoxin. In the primary infection there is an invasion of the upper respiratory mucosa. After a 1-3 week of incubation the child produces a mild cough, coryza and sneezing. Endotoxin irritates bronchi. Cytotoxin destroys ciliated epithelial cells. Prolonged coughing ends in the characteristic 'whoop' due to rapid deep inspiration. The 'whoop' may be absent in babies or older adults. Fatalities result from complications, not toxins produced by the microbe. *Streptococcus pyogenes* or pneumococcal pneumonia can cause a secondary infection. Chronic cough is observed in older teens to adults. Transmission is by droplet spray from patients in early stages of disease. Treatment involves intensive care for very young children and erythromycin therapy. Prevention is available through a vaccine DPT. Because of rare side effects of the original DPT (killed organisms) a newer vaccines (acellular) were developed and are safer. These newer vaccines protect against disease but not infection because the vaccine targets the fimbriae and the pertussis toxin.

An outbreak of *Legionella pneumophila* in 1976 amongst American Legion members brought this organism to the attention of the world. The organism gram stains irregularly and is a pleomorphic rod and is a strict aerobe. It is easily cultured from watery environments and air conditioning units, cooling towers, humidifiers, whirlpools which serve as a reservoir. Legionnaire's disease is a lobar pneumonia with consolidation found generally in the immunosuppressed and producing a 20% fatality rate in the above patient. Often there are many sub clinical cases. Pontiac fever was determined to be caused by the organism years afterward by retrospectively looking at serological studies. Transmission is via watery aerosols and is not communicable from man to man. Treatment is with erythromycin. Vaccines are not available. Culture has been replaced by immunoassay and nucleic acid amplification methods as a means of diagnosis.