ENTEROBACTERIACEAE

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ENTEROBACTERIACEAE

- The Enterobacteriaceae are a large, heterogeneous group of Gram-negative rods whose natural habitat is the intestinal tract of humans and animals.

- The family includes many genera (Escherichia, Shigella, Salmonella, Enterobacter, Klebsiella, Serratia, Proteus, and others).

- Some enteric organisms, such as *Escherichia coli*, are part of the normal microbiota and incidentally cause disease, but others, the salmonella and shigella, are regularly pathogenic for humans.

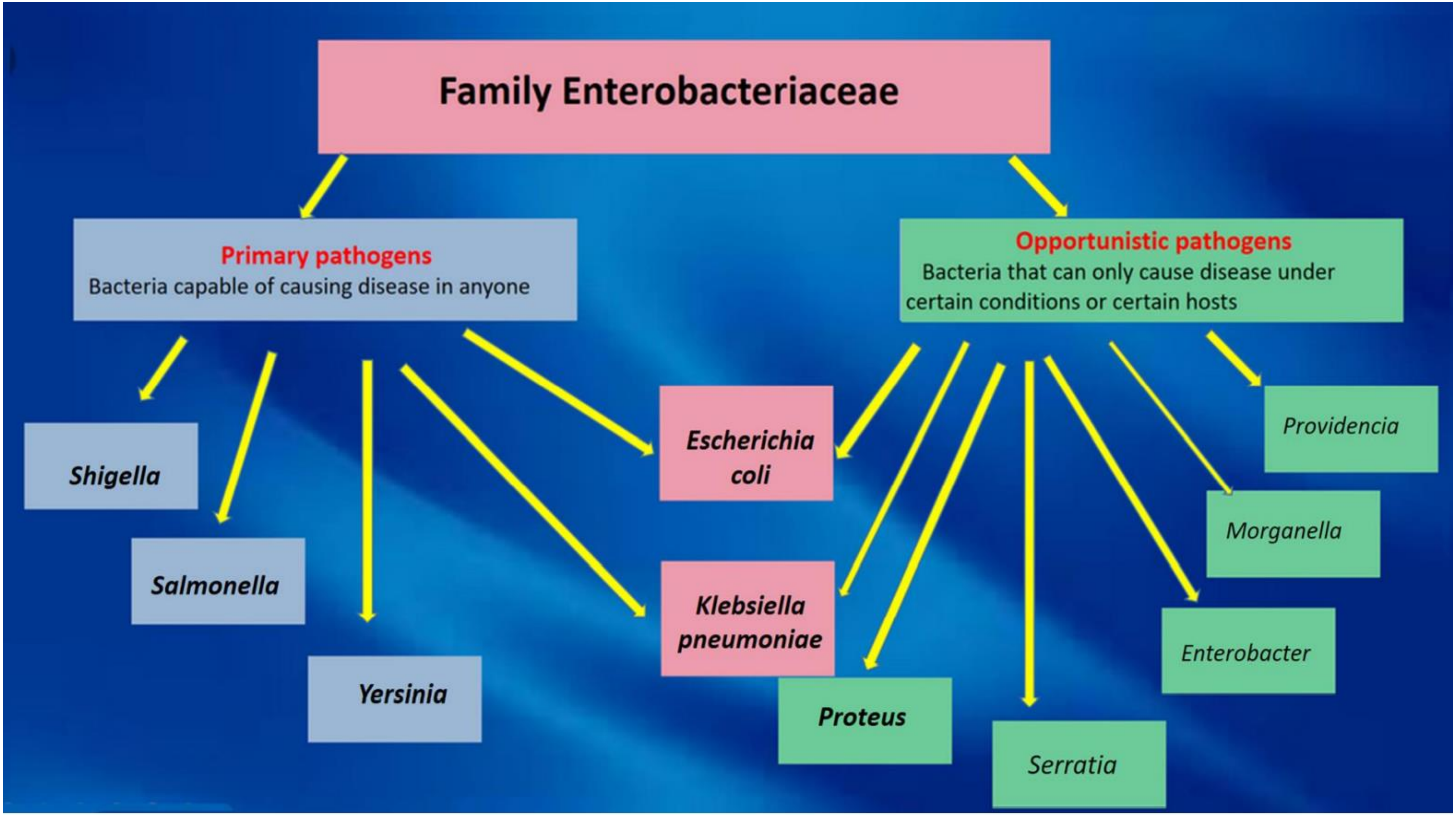
Habitat

- digestive tube (colon) of human and animals
- Enterobacteriaceae currently has 53 genera (and over 170 named species)
- Facultative anaerobes
- Diarrheal illnesses
- 3 million death/year
- 4 billion infectoions/ worldwide

Enterobacteriaceae Classification

Escherichia
Shigella
Citrobacter
Klebsiella
Hafnia
Enterobacteriaceae
Providencia
Edwardsiella
Pectinobacterium

Salmonella
Morganella
Enterobacter
Serratia
Proteus
Yersinia
Erwinia



Family Enterobacteriaceae
Primary pathogens
Bacteria capable of causing disease in anyone
Opportunistic pathogens
Bacteria that can only cause disease under certain conditions or certain hosts
Shigella
Salmonella
Yersinia
Escherichia coli
Klebsiella pneumoniae
Proteus
Serratia
Enterobacter
Morganella
Providencia

Enterobacteriaceae

Lactose fermenter

- *Echerichia coli*
- Klebsiella
- Enterobacter
- Serratia

Non Lactose fermenter

- Salmonella
- Shigella
- Proteus

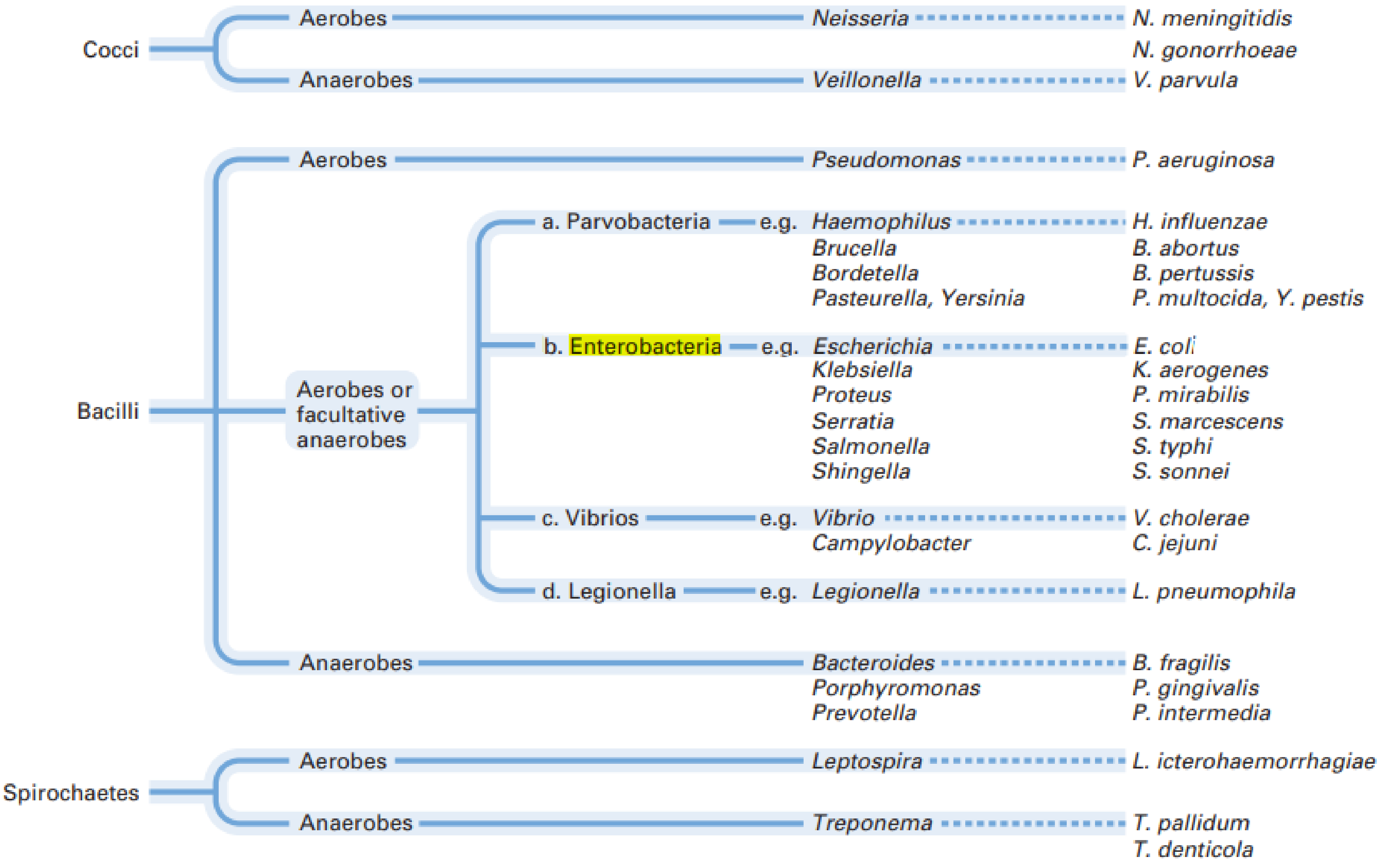


Fig. 2.8 A simple classification of Gram-negative bacteria.

Morphology and General Characteristics

- Facultative anaerobe (bacteria that can grow in the presence or absence of oxygen).

- Gram-negative, non-sporing, rod-shaped bacteria

- If motile, motility is by peritrichous flagella(Except Shigella & Klebsiella)

- Grow on bile-containing media (MacConkey agar)

- Many are normal inhabitants of the intestinal tract of man and other animals.

- Some are enteric pathogens and others are urinary or respiratory tract pathogens.

- Differentiation is based on **biochemical reactions** and differences in **antigenic structure**.

Morphology and physiology

- Grow readily and rapidly on simple media.
- **Klebsiella spp**. have large capsules (form large and very mucoid colonies); those of **Enterobacter** have smaller capsules; the others produce diffusible slime layers (form circular, convex and smooth colonies).

Enterobacteriaceae physiology

- Glucose is fermented with strong acid formation and often gas
- Reduce nitrates to nitrite
- Do not liquefy alginate
- Oxidase negative
- All species are endotoxigenic because of the liposaccharide outer cell wall

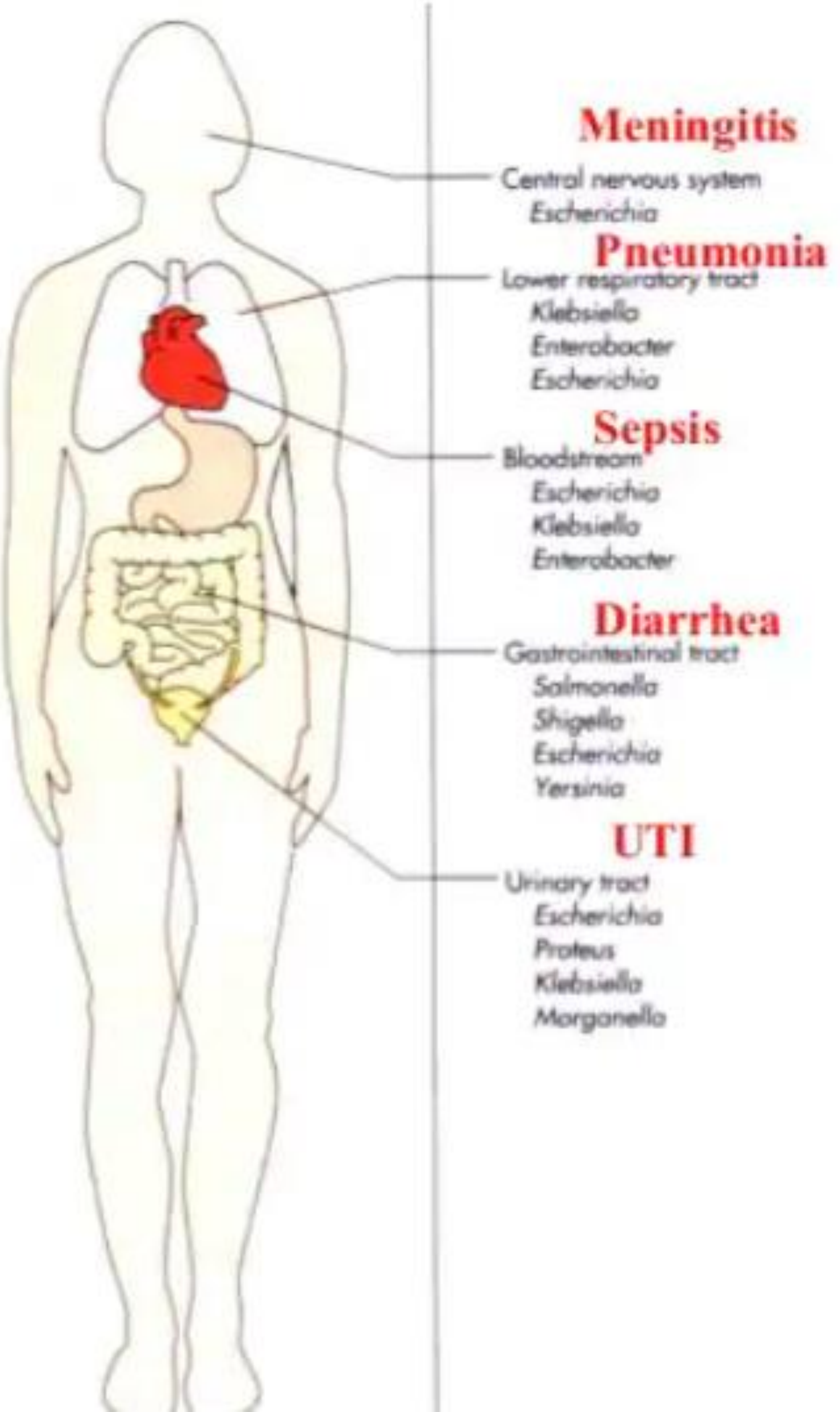
Enterobacteriaceae

Opportunistic pathogens

Escherichia coli
Klebsiella pneumoniae
Enterobacter aerogenes
Serratia marcescens
Proteus spp.
Providencia spp.
Citrobacter spp.

Obligate pathogens

Salmonella spp.
Shigella spp.
Yersinia spp.
Some *E. coli* strains

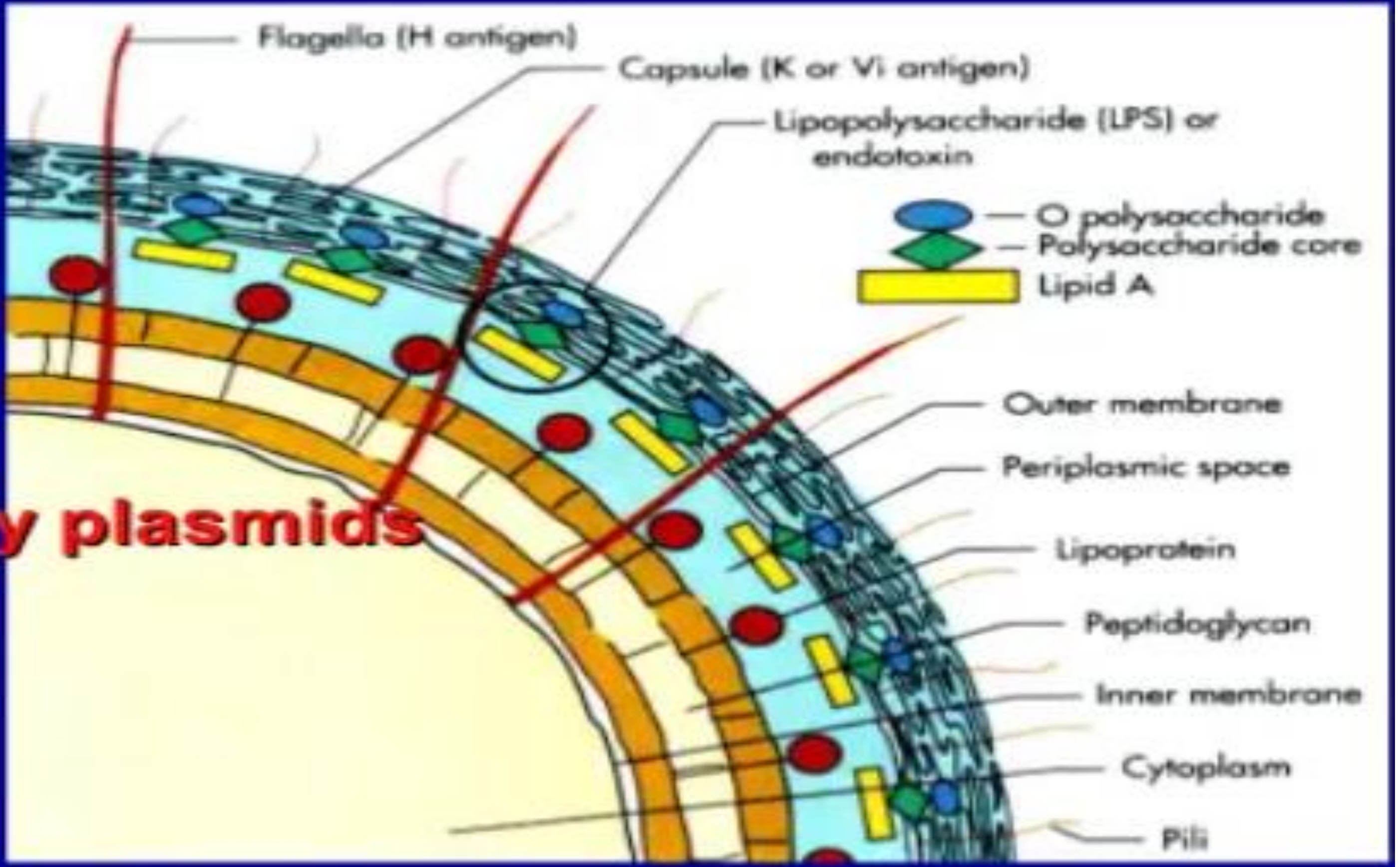


Modes of Infection

- Contaminated food and water (Salmonella spp., Shigella spp., *Yersinia enterocolitica*, *Escherichia coli* O157:H7).
- Endogenous (urinary tract infection, primary bacterial peritonitis, abdominal abscess).
- Abnormal host colonization (nosocomial pneumonia).
- Transfer between debilitated patients.
- Insect (flea) vector (unique for *Yersinia pestis*).

Antigenic Structure

- Most are motile by **peritrichous flagella** --H antigens.
- **Capsule –** K antigen (Vi for Salmonella).
- **Cell envelope (wall)**
- **LPS (endotoxin) –** O antigen.
- various outer membrane proteins.
- **Pili** - various antigen types, some encoded **by plasmids**



Escherichia coli

live in the human gut and are usually harmless but some are pathogenic causing diarrhea and other symptoms as a result of ingestion of contaminated food or water.

The organisms can disseminate into the bloodstream producing systemic hemolytic-uremic syndrome (hemolytic anemia, thrombocytopenia and kidney failure) which is often fatal.

The commonest community-acquired urinary tract infection is caused by *E. coli*.

Escherichia coli

1. Urinary tract infection (UTI):- *E. coli* is the most common cause of urinary tract infection and accounts for approximately 90% of first urinary tract infections in young women. The symptoms and signs include urinary frequency, dysuria, hematuria, and pyuria. Flank pain is associated with upper tract infection. None of these symptoms or signs is specific for *E. coli* infection. Urinary tract infection can result in bacteremia with clinical signs of sepsis.

2. *E. coli*-associated diarrheal diseases:- *E. coli* that cause diarrhea is extremely common worldwide. These *E. coli* are classified by the characteristics of their virulence properties. Each group causes disease by a different mechanism, at least six of which have been characterized.

- **Enteropathogenic *E. coli* (EPEC)**

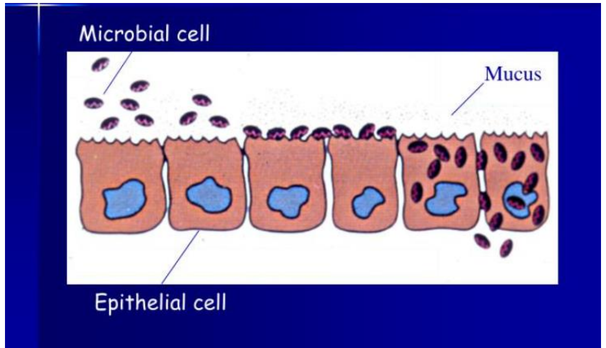
- Are an important cause of diarrhea in infants, especially in developing countries.

- EPEC adhere to the mucosal cells of the small bowel. Characteristic lesions can be seen on electron micrographs of small bowel biopsy lesions. The result of EPEC infection in infants is characterized by severe, watery diarrhea, vomiting, and fever, which are usually self-limited but can be prolonged or chronic.

Enterotoxigenic *E. coli* (ETEC) are a common cause of “traveler's diarrhea” and a very important cause of diarrhea in children less than 5 years of age in developing countries. ETEC colonization factors (pili known as colonization factor antigens [CFAs]) specific for humans promote adherence of ETEC to epithelial cells of the small bowel. Some strains of ETEC produce a heat-labile enterotoxin (LT) that is under the genetic control of a plasmid and **is closely related to cholera toxin**. The result is an intense and prolonged hypersecretion of water and chlorides and inhibition of the reabsorption of sodium. The gut lumen is distended with fluid, and hypermotility and diarrhea ensue, lasting for several days. Some strains of ETEC produce the heat-stable enterotoxin (ST). The strains with both toxins produce a more severe diarrhea.

Shiga toxin-producing *E. coli* (STEC) are named for the cytotoxic toxins they produce. There are at least two antigenic forms of the toxin referred to as **Shiga-like toxin 1** and **Shiga-like toxin 2**. STEC has been associated with mild non-bloody diarrhea, hemorrhagic colitis, a severe form of diarrhea, and with hemolytic uremic syndrome, a disease resulting in acute renal failure, microangiopathic hemolytic anaemia, and thrombocytopenia.

Enteroinvasive *E. coli* (EIEC) produce a disease very similar to shigellosis. The disease occurs most commonly in children in developing countries and travelers to these countries. Similar to Shigella, EIEC strains are non-lactose or late lactose fermenters and are nonmotile. EIEC produce disease by invading intestinal mucosal epithelial cells.



Enteroaggregative *E. coli* (EAEC) causes acute and chronic diarrhea (>14 days in duration) in persons in developing countries. These organisms also are the cause of foodborne illnesses in industrialized countries and have been associated with traveler’s diarrhea and persistent diarrhea in patients with HIV. **Diffusely adherent *E. coli* (DAEC)** have been considered a diarrheagenic group of *E. coli* (DEC). They are characterized by the diffuse adherence pattern on cultured epithelial cell

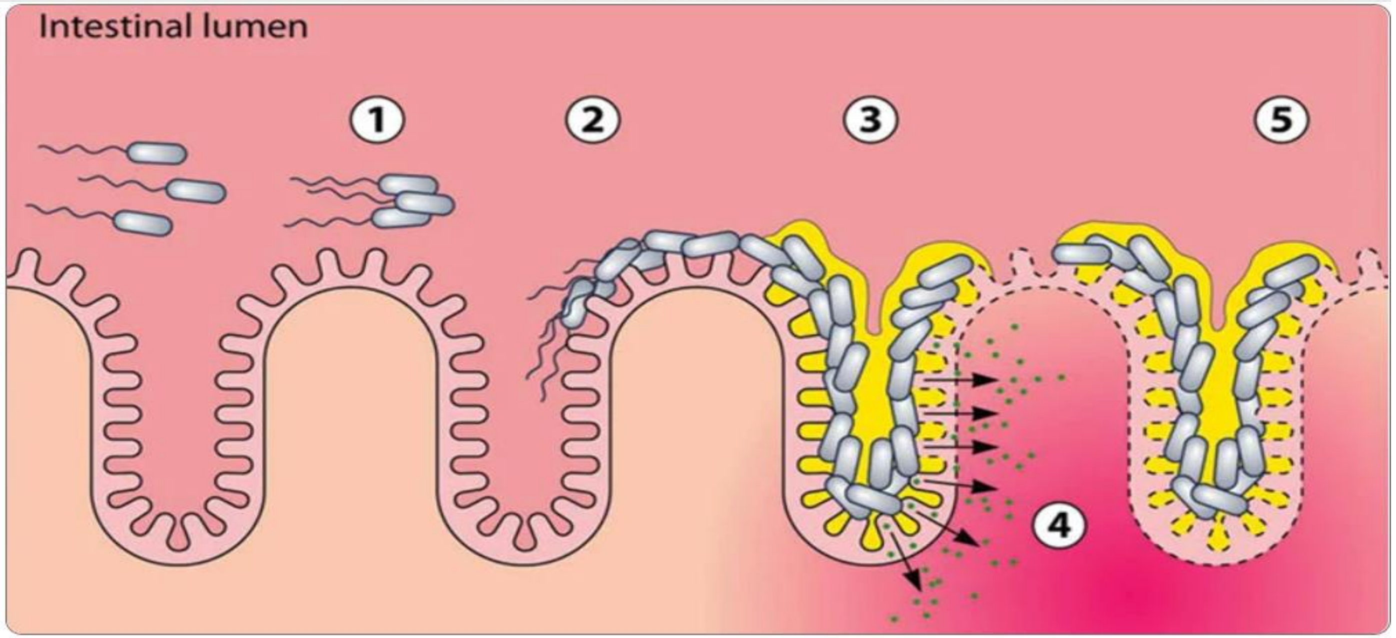


Figure: Stages of pathogenesis of EAEC. Numbers in circles show the progression of EAEC pathogenesis. (1) Agglutination of planktonic EAEC bacteria. (2) Adherence to the intestinal epithelium and colonization of the gut. (3) Formation of biofilm. (4) Release of bacterial toxins, inducing damage to the epithelium and increased secretion. (5) Establishment of additional biofilm. **Source: DOI: 10.1128/CMR.00112-13**

3. Sepsis:- when normal host defenses are inadequate, *E. coli* may reach the bloodstream and cause sepsis. Newborns may be highly susceptible to *E. coli* sepsis because they lack IgM antibodies. Sepsis may occur secondary to urinary tract infection.

4. Meningitis:- *E. coli* and group B streptococci are the leading causes of meningitis in infant.

Diagnosis:-

1. **Stool culture**
2. **Blood culture:** Performed when *E. coli* is suspected to have caused a bloodstream infection, such as sepsis.
3. **Urine culture:** Performed when *E. coli* is suspected to have caused a urinary tract infection (UTI).
4. **Serological tests:** These tests are used to detect specific antibodies or antigens in the blood or other body fluids. For example, enzyme-linked immunosorbent assay (ELISA) can detect the presence of Shiga toxin produced by some strains of *E. coli*.

5. **Imaging studies:** Imaging studies such as ultrasound or computed tomography (CT) scans may be performed in cases where complications of *E. coli* infections are suspected, such as renal abscess or peritonitis.

6. **Polymerase Chain Reaction (PCR):** PCR can be used to detect specific genetic markers of *E. coli*, including virulence genes and antibiotic resistance genes. It provides rapid and sensitive identification, particularly for pathogenic strains.

7. **Antimicrobial Susceptibility Testing:** Performed on *E. coli* isolates to guide appropriate antibiotic treatment. T

TREATMENT

Acute uncomplicated UTIs are often treated empirically. Because of widespread resistance to earlier agents like ampicillin, the use of trimethoprim/sulfamethoxazole (TMP-SMX) and fluoroquinolones for this purpose rose steadily.

In cases of empiric treatment failure, the selection of other antimicrobials must be guided by antimicrobial susceptibility testing of the patient's isolate.

Because most *E coli* diarrheas are mild and self-limiting, treatment is usually not required.

Shigella

- Shigellosis is a strictly human disease with no animal reservoirs.
- Shigella germs spread easily from one person to another, and it only takes a small amount to make someone sick with a disease called shigellosis
- Worldwide, it is consistently one of the most common causes of infectious diarrhea with over 150 million cases and 600 000 deaths per year.

The four species of Shigella are:

Shigella sonnei (the most common species in the United States)

Shigella flexneri

Shigella boydii

Shigella dysenteriae

- The organism invades the epithelial lining layer but does not penetrate.

- Enterotoxin plays an important role.

- Usually, within 2 to 3 days, dysentery results from bacteria damaging the epithelial layers lining the intestine, often with the release of mucus and blood (found in the faeces) and attraction of leukocytes (also found in the faeces as "pus").

- However, watery diarrhea is frequently observed with no evidence of dysentery.

- Shiga toxin such as neurotoxic, enterotoxic and cytotoxic, plays a role in disease production

Treatment

- Several antimicrobial agents have proved effective in the treatment of shigellosis.
- Because the disease is usually self-limiting, the beneficial effect of treatment is in shortening the duration of the illness and the period of excretion of organisms.
- Ampicillin was once the treatment of choice, but resistance rates as high as 50% have caused a shift to other agents;
- in recent years, ciprofloxacin, ceftriaxone, and azithromycin have been used, depending on susceptibility testing

Prevention

- Standard sanitation practices such as sewage disposal and water chlorination are important in preventing the spread of shigellosis.

- In certain circumstances, insect control may also be important, because flies can serve as passive vectors when open sewage is present.

- Basic personal hygiene and frequent handwashing.

Diagnosis:-

1- **Clinical Presentation and History:**
The initial step in diagnosing Shigella infections is to consider the clinical symptoms and obtain a detailed medical history.

2- **Stool culture:**
Is the gold standard diagnostic test for shigellosis. It involves isolating and identifying Shigella species from stool samples. Selective agar media, such as MacConkey agar or Hektoen enteric agar, are used to inhibit the growth of normal gut flora and promote the growth of Shigella species.

3- **Biochemical tests:**
Such as lactose fermentation, indole production, and agglutination with specific antisera, are performed to confirm the presence of Shigella species.

4- **Serological Testing:**

Such as enzyme immunoassays (EIAs) or latex agglutination tests, can be used to detect Shigella-specific antibodies in blood serum.

5- Polymerase Chain Reaction (PCR):

- PCR can detect specific genetic markers of Shigella species, providing rapid and sensitive identification.
- **6- Antimicrobial Susceptibility Testing:**

Salmonella

- Salmonella infection (salmonellosis) is a common bacterial disease that affects the intestinal tract.
- Salmonella bacteria typically live in animal and human intestines and are shed through feces.
- Humans become infected most frequently through contaminated water or food.
- Infections most often cause vomiting or diarrhea, sometimes severe. In rare cases, Salmonella illness can lead to severe and life-threatening bloodstream infections.
- Complexity of O, K, and H antigens leads to many serotypes.
- <u>Salmonella species vary in a preferred host.</u>

<u>Non-typhoidal</u> salmonellosis is a worldwide disease of humans and animals.

- Animals are the main reservoir, and <u>the disease is usually foodborne</u>, although it can be spread from person to person.

- *Salmonella enteritidis* is transmitted from contaminated food (such as poultry and eggs).
- It does not have a human reservoir and usually presents as a gastroenteritis (nausea, vomiting and non-bloody stools).

- The disease is usually self-limiting (2 - 5 days).
- Like Shigella, these organisms invade the epithelium and do not produce systemic infection.

- **ENTERIC (TYPHOID) FEVER**
- Typhoid fever is a strictly human disease.
- Spreads mainly from person to person via the fecal-oral route.
- Enteric fever: This includes both Typhphoid fever and Paratyphoid fever caused by *S. Typhi* and *S. Paratyphi*.
- Has no animal reservoirs.
- *S. Typhi* and *S. Paratyphi are* found only in humans.
- Causes three types of diseases in humans. i.e enteric fever, enterocolitis and septicaemia

- *Salmonella typhi*

- Transmitted from a human reservoir or in the water supply (if sanitary conditions are poor) or in contaminated food.

- initially invades the intestinal epithelium.

-The organism penetrates (usually within the first week) and passes into the bloodstream where it is disseminated in macrophages.

-Typical features of a systemic bacterial infection are seen.

-The Vi (capsular) antigen plays a role in the pathogenesis of typhoid.

-Antibiotic therapy is essential.

Virulence factors

- Endotoxin – may play a role in intracellular survival
- Capsule (for *S. typhi* and some strains of *S. paratyphi*)
- Adhesions – both fimbrial and non-fimbrial
- Type III secretion systems and effector molecules

Different systems may be found responsible for pathogenesis

- One type is involved in promoting entry into intestinal epithelial cells
- The other type is involved in the ability of Salmonella to survive inside macrophages such as the outer membrane proteins

- Flagella – help bacteria to move through intestinal mucous
- Enterotoxin - may be involved in gastroenteritis

Diagnosis

Salmonella typhi and *S.paratyphii* are the two major pathogens that cause enteric fever.

In which the following are possible specimens taken for the routine diagnosis of the disease as blood, feces, urine and sputum which are less used in diagnosis.

A. Specimens

a) Enteric fever: blood, bone marrow, stool, urine.

b) Food poisoning: stool, vomitus, suspected food

c) Septicemia: blood.

B. Culture and identification

C. Widal test

YERSINIA DISEASES

- Yersiniosis is a disease caused by bacteria called Yersinia. Although many species of Yersinia are found worldwide, most human illnesses are caused by *Yersinia enterocolitica*.

- Other species of Yersinia affecting humans are *Y. pseudotuberculosis*, which causes an illness similar to *Y. enterocolitica*, *and Y. pestis* which causes plague.

- Yersinia are hardy bacteria that can survive in adverse conditions like refrigeration and environments low in oxygen.

Yersinia entercolitica - the organisms are invasive (usually without systemic spread).

- *Y. enterocolitica* may cause acute mesenteric lymphadenitis, a syndrome involving fever and abdominal pain that often mimics acute appendicitis.

- *Y. enterocolitica* infections are seen most often in young children.

- Transmitted by ingestion from animal source

-*Y. enterocolitica* can be transmitted by fecal contamination of water or milk by domestic animals or from eating meat products.

Treatment of yersiniosis

- usually involves treating the symptoms only. For example, persons with diarrhea should generally drink lots of liquids to avoid dehydration. More severe or complicated cases may require antibiotics.

Diagnosis of Yersinia Diseases:

1- **Clinical Presentation and History:** The first step in diagnosing Yersinia diseases is to consider the clinical presentation and obtain a detailed medical history. include fever, gastrointestinal symptoms (such as diarrhea, abdominal pain), lymphadenopathy, and systemic symptoms in severe cases.

2- **Laboratory Tests:**

- **Stool Culture:** Yersinia enterocolitica and Yersinia pseudotuberculosis infections can be diagnosed by isolating the bacteria from stool samples. Selective agar media, such as CIN (Cefsulodin-Irgasan-Novobiocin) agar, are used to inhibit the growth of normal gut flora and promote the growth of Yersinia species.

- **Blood Culture:** In cases of suspected systemic Yersinia infections or Yersinia septicemia.

- **Serological Testing:** Serological tests, such as enzyme-linked immunosorbent assay (ELISA) or indirect fluorescent antibody (IFA) tests, can be used to detect antibodies against Yersinia species.

- **Polymerase Chain Reaction (PCR)** PCR can be performed on clinical samples such as stool, blood, or tissue samples.

- **Imaging Studies:** such as X-rays, ultrasound, or computed tomography (CT).

4- **Molecular Typing and Genomic Analysis:** such as multilocus sequence typing (MLST) can be utilized for further characterization of Yersinia isolates.

Klebsiella

- Species are present in the nasopharynx and faeces of about 5% of normal individuals.
- known to cause both community-acquired and nosocomial infections, including liver abscesses, pneumonia, urinary tract infections and bacteremia worldwide.
- The most common isolates species are *K. pneumoniae* and *K. oxytoca*.
- *Klebsiella pneumoniae* is often involved in respiratory infections.
- Recently a particular clone of *K. pneumoniae* has emerged as a cause of community-acquired pyogenic liver abscess that is seen mostly among Asian males worldwide.
- **Klebsiella species rank among the top 10 bacterial pathogens responsible for hospital-acquired infections.**

- In the past two decades, a distinct hypervirulent variant of *K. pneumoniae*, has emerged as a clinically significant pathogen responsible for highly invasive infections
- Unlike classical *K. pneumoniae*, hypervirulent *K. pneumoniae* (hvKp) can spread from the original site of infection to other organs. Once invasive dissemination occurs, patients often suffer severe and irreversible sequelae, such as blindness and central nervous system damage.
- The conditions of patients infected with hypervirulent *K. pneumoniae* causing liver abscess are serious, posing a great threat to public health and have attracted the attention of clinicians.

- The pathogenicity of *K. pneumoniae* mainly arises from various virulence factors which allow it to overcome innate host immunity and maintain infection in a mammalian host.
- <u>The main virulence factors that play an important role in pathogenicity are</u> :-

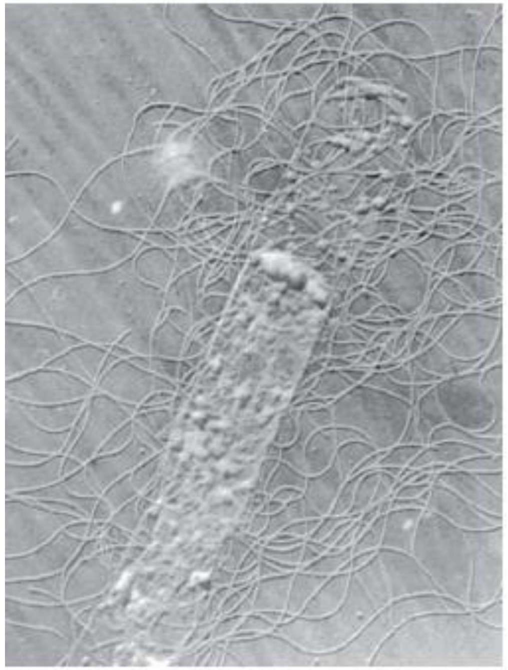
- capsular polysaccharide
- Lipopolysaccharide
- pili
- Iron capturing ability

Treatments

- The recommended treatment regimen is azithromycin 1 g orally once per week (or 500 mg daily) for at least three weeks and until all lesions have completely healed.
- Alternative antimicrobial treatment regimens exist, using ciprofloxacin, doxycycline, or trimethoprim–sulfamethoxazole.

Proteus

- species are widespread in the environment and are normal inhabitants of the human intestinal tract.
- The two species to most commonly produce infections in humans are *P. mirabilis* and *P. vulgaris*.
- Both species produce urease, resulting in rapid hydrolysis of urea with liberation of ammonia. Thus, in urinary tract infections, the urine becomes alkaline, promoting stone formation.
- in addition, the rapid motility of Proteus may also contribute to its invasion of the urinary tract.



Electron micrograph of *Proteus vulgaris*, showing peritrichous flagellation (9000×)

P. mirabilis causes urinary tract infections and occasionally other infections, such as bloodstream infections (frequently secondary due to a UTI) and respiratory tract infections.

P. vulgaris is probably more frequently implicated in wound and soft tissue infections than UTIs.

Pathogenicity of Proteus

They can cause –

- Urinary infection
- Abdominal and wound infections
- Septicemia
- Infection of ear
- Respiratory infections
- Nosocomial infections

Treatments

Strains of Proteus **vary greatly in antibiotic susceptibility**.

- *P. mirabilis* is resistant to nitrofurantoin but most often susceptible to penicillins (eg, ampicillin and amoxicillin), trimethoprim–sulfamethoxazole, cephalosporins, aminoglycosides, and imipenem;
- *P. vulgaris* is generally more resistant to various antibiotics (specifically ampicillin, amoxicillin, and piperacillin; the most active antibiotics for *P. vulgaris* and other members of the group are aminoglycosides and broad-spectrum cephalosporins.

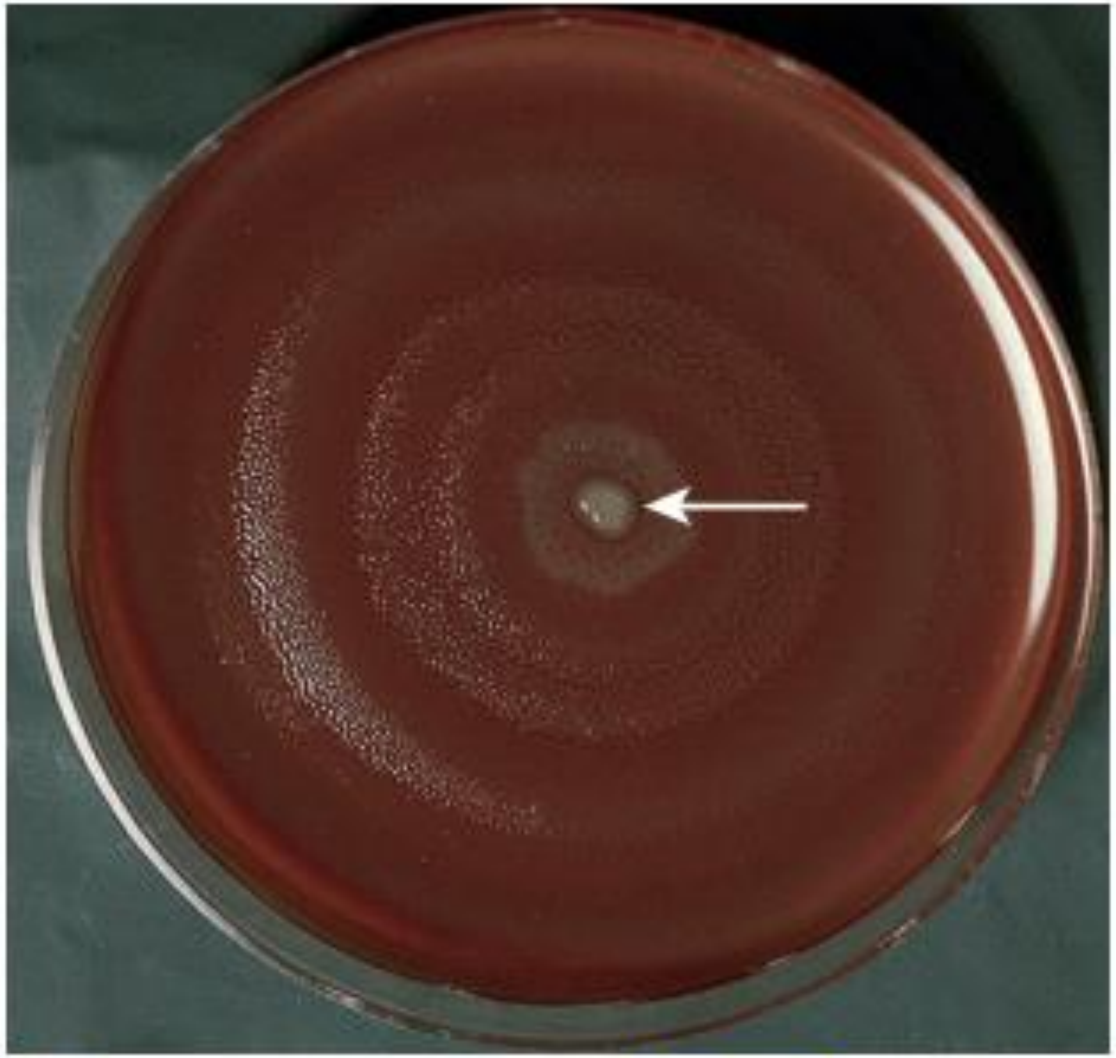
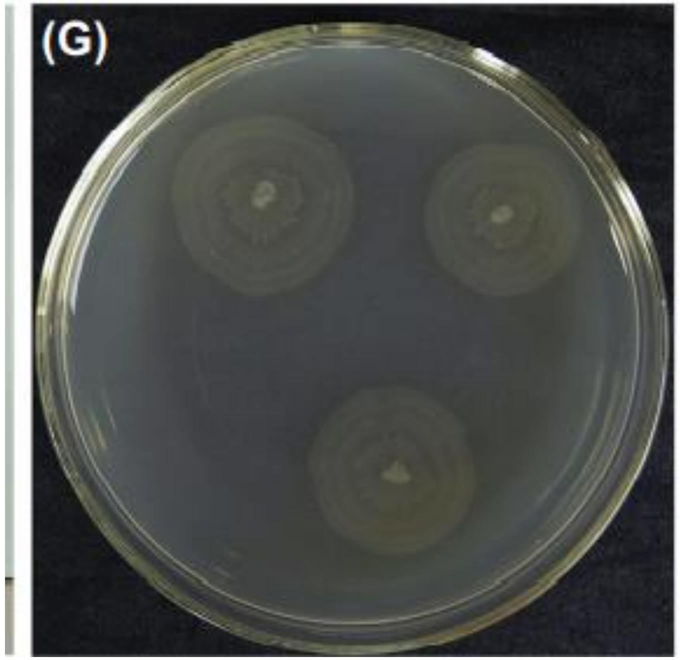


FIGURE 8-9 Swarming colonies of *Proteus* spp. The organism was inoculated in the middle of the blood agar plate *(arrow)*.



Enterobacter species

- Enterobacter species are indigenous to the intestinal tract but can be found on plants and as free-living saprophytes.
- They may cause nosocomial urinary tract infections and very rarely a primary infection.
- *Enterobacter cloacae* and *Enterobacter aerogenes* are the most frequently isolated as transients in the oral cavity.
-

- There are 26 species and 2 subspecies, of which recently, 6 has been reclassified to other genera. Only 10 have been isolated from clinical material.
- They grow readily on ordinary media, ferment glucose with the production of acid and gas, and are motile by peritrichous flagella.
- Some strains with a K antigen possess a capsule.
- Colonies of Enterobacter strains may be slightly mucoid. They are catalase positive and oxidase negative. Nitrates are also reduced. They also ferment glucose and lactose with the production of acid and gas.

- Enterobacter has the general characteristics of Klebsiella species but can be differentiated because they are motile and ornithine positive.
- Enterobacter species are widely distributed in nature. They are found in the soil, water, dairy products, and the intestines of animals as well as humans.

Treatment

- Enterobacter infections typically involve antibiotics. However, the choice of antibiotics may depend on the specific strain of Enterobacter and its susceptibility to different drugs.

Commonly used antibiotics for Enterobacter infections include:

1. Carbapenems (e.g., imipenem, meropenem): These are broad-spectrum antibiotics and are often considered the first-line treatment for severe Enterobacter infections.

2. Third-generation cephalosporins (e.g., ceftriaxone, cefotaxime)
3. Aminoglycosides (e.g., gentamicin, amikacin)

4. Fluoroquinolones (e.g., ciprofloxacin, levofloxacin)


Thank You.