

Enterobacteriaceae

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Classification – more than 15 different genera

Escherichia

Shigella

Edwardsiella

Salmonella

Citrobacter

Klebsiella

Enterobacter

Hafnia

Serratia

Proteus

Providencia

Morganella

Yersinia

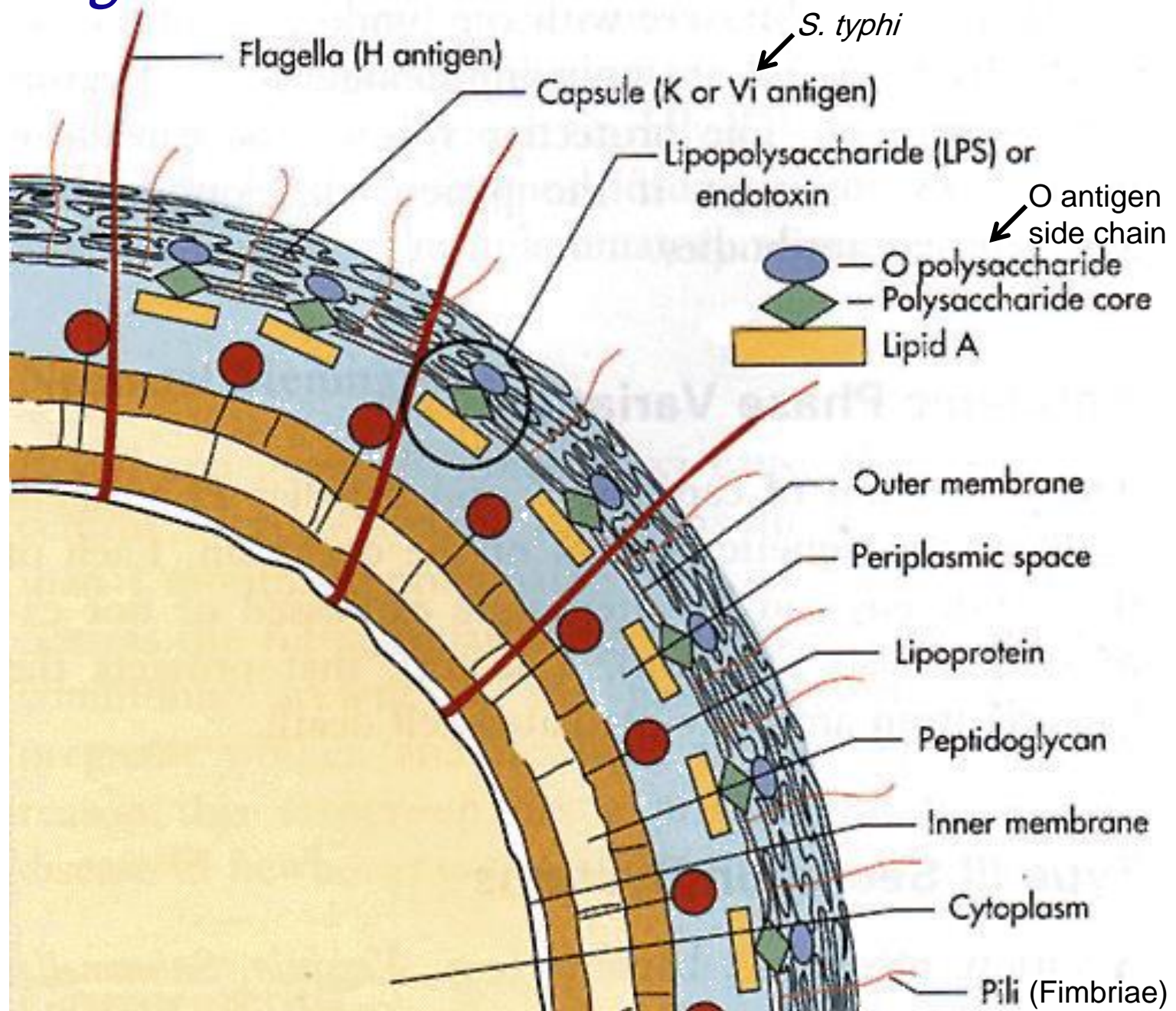
Erwinia

Pectinobacterium

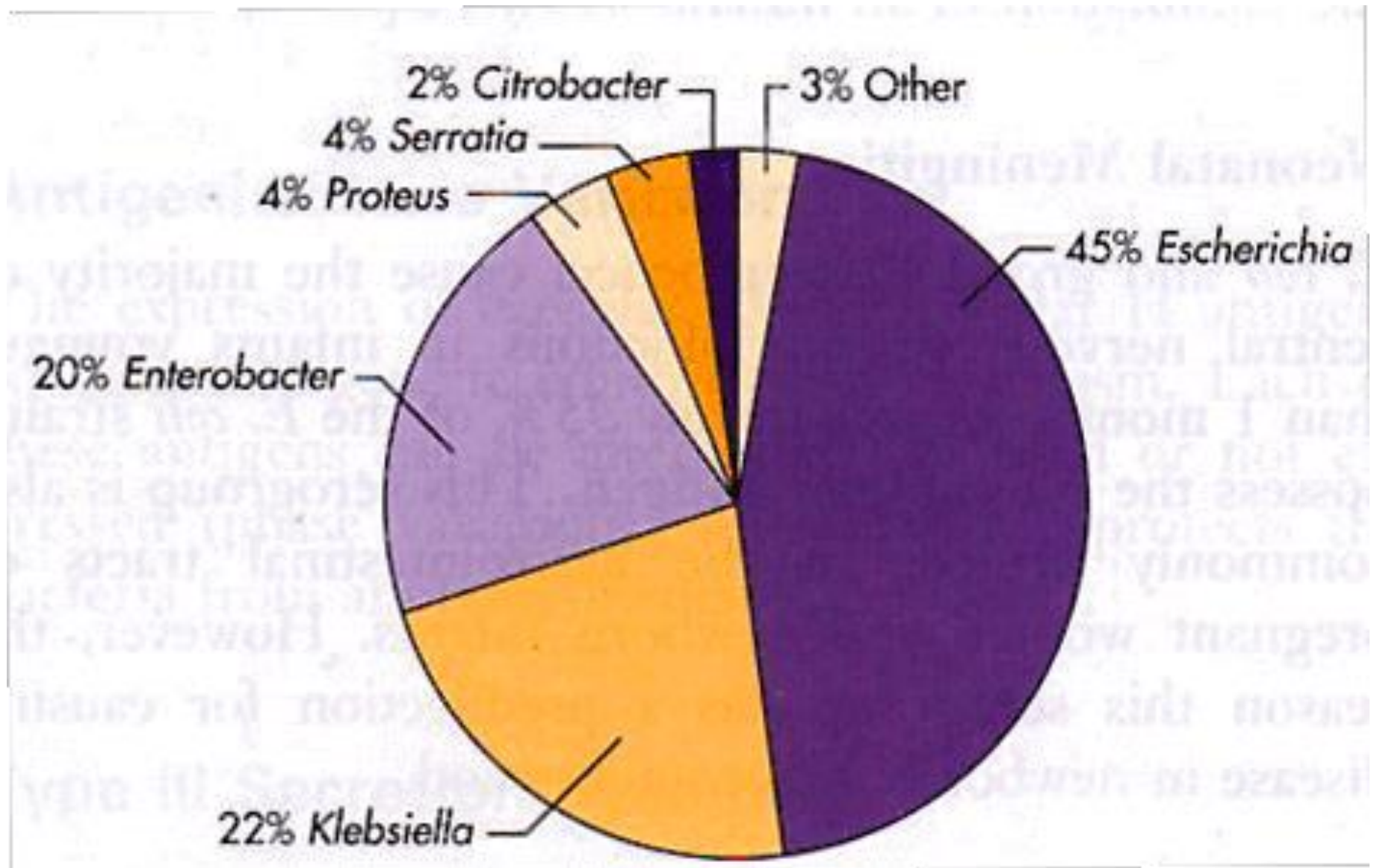
•Morphology and General Characteristics

- Gram-negative, nonsporing rod shaped bacteria
- Oxidase negative
- Ferment glucose and may or may not produce gas in the process (aerogenic or anaerogenic)
- Reduce nitrate to nitrite
- Are aerobes & facultative anaerobes
- If motile, motility is by peritrichous flagella
- 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

Antigenic Structure of Enterobacteriaceae



Incidence of Enterobacteriaceae Associated with Bacteremia



- Most grow well on ordinary lab media and selective and differential media originally developed for the selective isolation of enteric pathogens.
- Selective media: incorporation of dyes and bile salts that inhibit G+ organisms and may suppress the growth of nonpathogenic species of *Enterobacteriaceae*.
- Many are differential on the basis of whether or not the organisms ferment lactose and/or produce H₂S.

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Antigens

- Somatic O antigens – these are the heat stable polysaccharide & part of the LPS.
- Flagellar H antigens – are heat labile
- Envelope or capsule K antigens – overlay the surface O antigen and may block agglutination by O specific antisera. Boiling for 15 minutes will destroy the K antigen and unmask the O antigens.
- In *Salmonella* the K antigen is called the Vi (virulence) antigen.

• *Escherichia coli*

– Normal inhabitant of the G.I. tract.

– Some strains cause various forms of gastroenteritis.

– Is a major cause of urinary tract infection and neonatal meningitis and septicemia.

– May have a capsule.

• Pathogenic strains are usually hemolytic on BA

• Biochemical reactions:

– Ferments Glucose, Lactose, & Mannitol (acid & gas)
but Sucrose is not fermented [GLSM: + + - +]

– Motile

– Indole + ; Methyl red + ; Voges Prausker - ; Citrate – [IMViC + + - -]

- Urease negative; H₂S not produced
- TSI is A/A + gas
- There is an inactive biotype that is anaerogenic, lactose –, and nonmotile.
- Antigenic structure: has O, H, and K antigens. K1 has a strong association with virulence, particularly meningitis in neonates.
- Virulence factors
- Toxins
- Enterotoxins – produced by enterotoxigenic strains of *E. coli* (ETEC). *Causes movement of water and ions from the tissues to the bowel resulting in watery diarrhea.*

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–There are two types of enterotoxins:

1. LT – is heat labile and binds to specific Gm1 gangliosides on the epithelial cells of the small intestine where it stimulates adenylate cyclase to increase production of cAMP which causes increased outflow of water and electrolytes into gut lumen

2. ST – is heat stable and binds to specific receptors to stimulate the production of cGMP with the same results as with LT.

Both enterotoxins are composed of five B subunits (for binding) and one A subunit which has the toxic enzymatic activity.

–Shiga-type toxin – also called the verotoxin -produced by enterohemorrhagic strains of E. coli (EHEC) – is cytotoxic, enterotoxic, neurotoxic, and may cause diarrhea and ulceration of the G.I. tract. There are two types - shiga-like toxin 1 and shiga-like toxin 2.

–Enteroaggregative ST-like toxin – produced by enteroaggregative strains of E. coli (EAEC) – causes watery diarrhea.

- Hemolysins – two different types may be found: cell bound and secreted. They lyse RBCs and leukocytes and may help to inhibit phagocytosis when cell bound.

- Endotoxin

- Type III secretion system to deliver effector molecules directly into the host cells. Involved in inducing uptake of EIEC into intestinal cells.

- Adhesins – are also called colonization factors and include both pili or fimbriae and non-fimbrial factors involved in attachment. There are at least 21 different types of adhesins. Antibodies to these may protect one from colonization.
- Virulence factors that protect the bacteria from host defenses
 - Capsule
 - Iron capturing ability (enterochelin)
- Outer membrane proteins - are involved in helping the organism to invade by helping in attachment and in initiating endocytosis.

E. Coli

–Clinical significance

- Is the leading cause of urinary tract infections which can lead to acute cystitis (bladder infection) and pyelonephritis (kidney infection).

New evidence in women who suffer from recurrent UTIs suggests that this is due to the formation of pod-like *E. coli* biofilms inside bladder epithelial cells.

Bacteria living on the edges of the biofilms may break off leading to a round of infection.

Ascending urinary tract infection

Pod-like biofilm

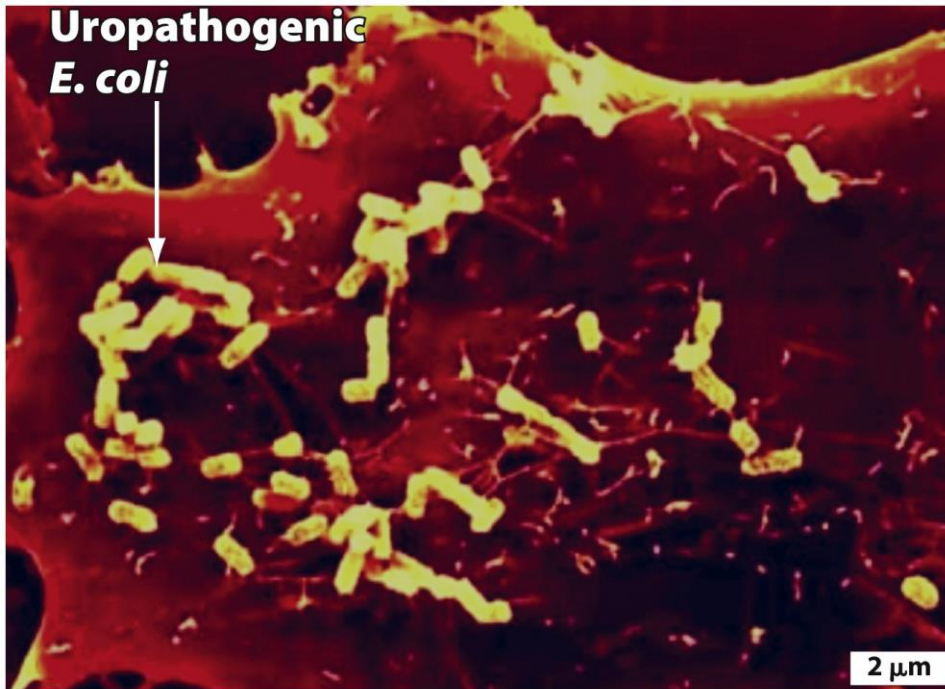
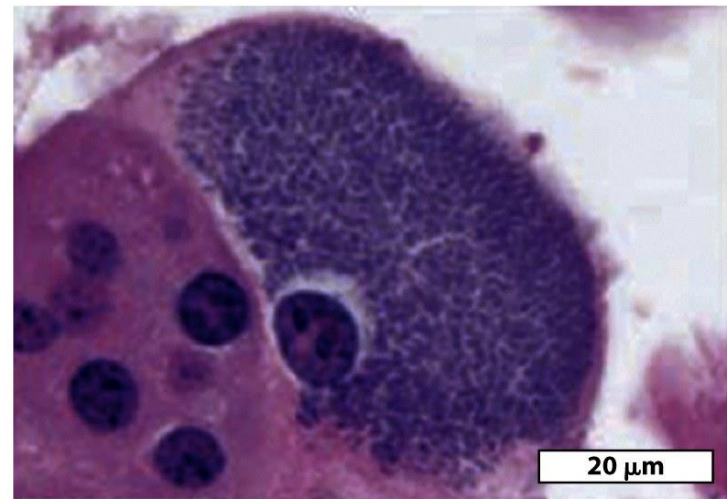
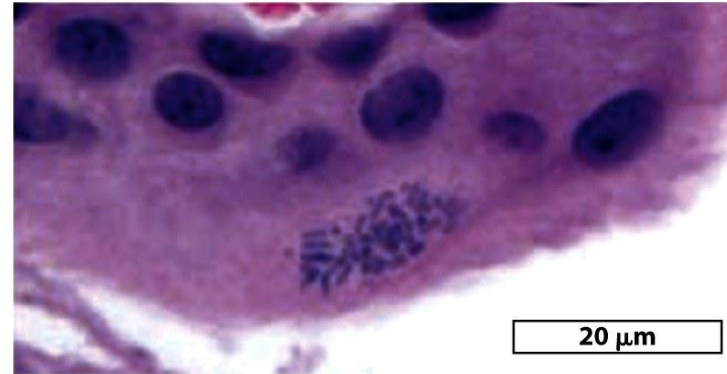
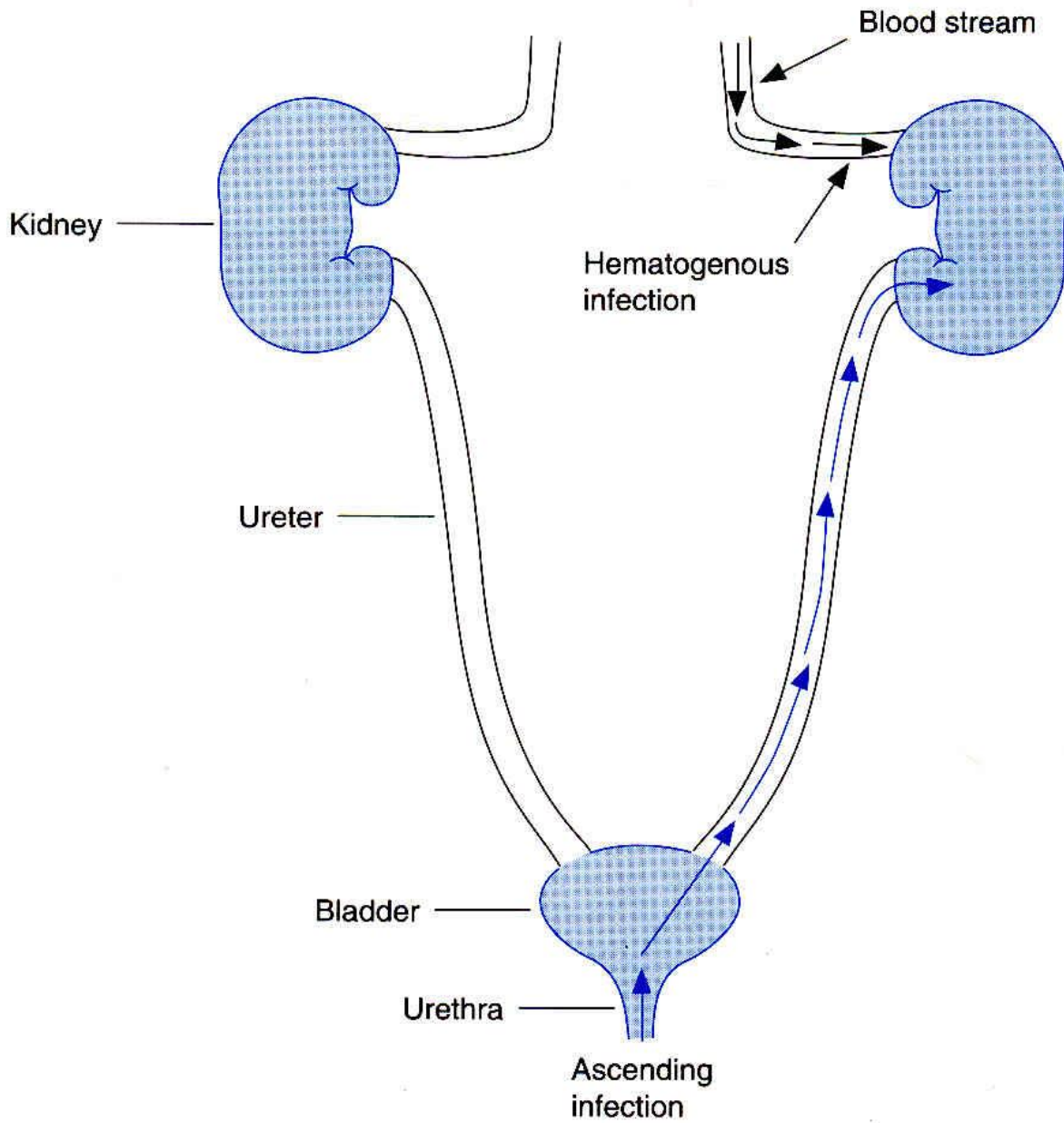


Figure 26.10a Microbiology: An Evolving Science
Courtesy of Hilde Merkert, University of Wuerzburg



Special Topic 26.1 figure 1c Microbiology: An Evolving Science
Gregory G. Anderson, et al. 2003. *Science* 301:105



- Predisposing factors: urinary obstruction due to prostatic enlargement, calculi or pregnancy
- 5-7% pregnant women may have asymptomatic bacteriurea – may lead to UTI, pyelonephritis and hypertension
- Pyelonephritis due to haematogenous spread by strains having K antigen
- Significant bacteriuria = $>10^5$ bacteria/ml
- Properly collected MSU
- Suprapubic aspiration in infants
- Indwelling catheter
- Antibody coated bacteria detected by immunofluorescence or staphylococcal coagglutination when Kidneys are infected

E. coli infections

- Neonatal meningitis –the leading cause of neonatal meningitis and septicemia with a high mortality rate. Usually caused by strains with the K1 capsular antigen.
- Gastroenteritis – there are several distinct types of *E. coli* that are involved in different types of gastroenteritis: enterotoxigenic *E. coli* (ETEC), enteroinvasive *E. coli* (EIEC), enteropathogenic *E. coli* (EPEC), enteroaggregative *E. coli* (EAEC), and enterohemorrhagic *E. coli* (EHEC).

E. coli gastroenteritis

- ETEC – is a common cause of traveler's diarrhea and diarrhea in children in developing countries.
- The organism attaches to the intestinal mucosa via colonization factors and then liberates enterotoxin.

-The disease is characterized by a watery diarrhea, nausea, abdominal cramps and low-grade fever for 1-5 days.

-Transmission is via contaminated food or water.

- ETEC diagnosed by detection of LT & ST

- LT: Tissue culture tests – rounding of Y1 mouse adrenal cells or elongation of CHO cells or by serological tests (ELISA) or in vivo tests (rabbit ileal loop) or by DNA probes

- ST: infant mouse test, ST-ELISA, DNA probes

-EPEC – Bundle forming pili are involved in attachment to the intestinal mucosa.

- This leads to changes in signal transduction in the cells, effacement of the microvilli, and to intimate attachment via a non-fimbrial adhesion called intimin.
- The exact mode of pathogenesis is unclear, but diarrhea with large amounts of mucous without blood or pus occurs along with vomiting, malaise and low grade fever.
- This is a problem mainly in hospitalized infants and in baby care centers.
- Diagnosis by serotyping



“Nonintimate” association:
Bacteria attach to host cell by
bundle-forming pili



Bacterial attachment:
Signal transduction event stimulated;
host cell tyrosine kinase activated;
 Ca^{2+} levels increase



“Intimate” contact:
Pedastallike structure (composed
of actin fibers) forms in host cell
under bacteria (intimin)

EPEC

Figure 16-4 Steps in the binding of EPEC strains to host cells and effect of binding on host cell functions.

E. coli gastroenteritis

- EIEC – The organism attaches to the intestinal mucosa via pili and outer membrane proteins are involved in direct penetration, invasion of the intestinal cells, and destruction of the intestinal mucosa.
- There is lateral movement of the organism from one cell to adjacent cells.
- Symptoms include fever, severe abdominal cramps, malaise, and watery diarrhea followed by scanty stools containing blood, mucous, and pus.
- Diagnosis: Serology test, cell penetration of He-La or ELISA test

E. coli gastroenteritis

- EAEC – Mucous associated autoagglutinins cause aggregation of the bacteria at the cell surface and result in the formation of a mucous biofilm.
- The organisms attach via pili and liberate a cytotoxin distinct from, but similar to the ST and LT enterotoxins liberated by ETEC.
- Symptoms include watery diarrhea, vomiting, dehydration and occasional abdominal pain.
- Diagnosis by cell culture technique
- EHEC – The organism attaches via pili to the intestinal mucosa and liberates the shiga-like toxin.
- The symptoms start with a watery diarrhea that progresses to bloody diarrhea without pus and crampy abdominal pain with no fever or a low-grade fever.

This may progress to hemolytic-uremic syndrome that is characterized by low platelet count, hemolytic anemia, and kidney failure.

- Most often caused by serotypes O157:H7.
- This strain of *E. coli* can be differentiated from other strains of *E. coli* by the fact that it does not ferment sorbitol in 48 hours (other strains do).
- A sorbitol-Mac (SMAC) plate (contains sorbitol instead of lactose) is used to selectively isolate this organism.
- Confirm that the isolate is *E. coli* O157:H7 using serological testing and confirm production of the shiga-like toxin before reporting out results.



