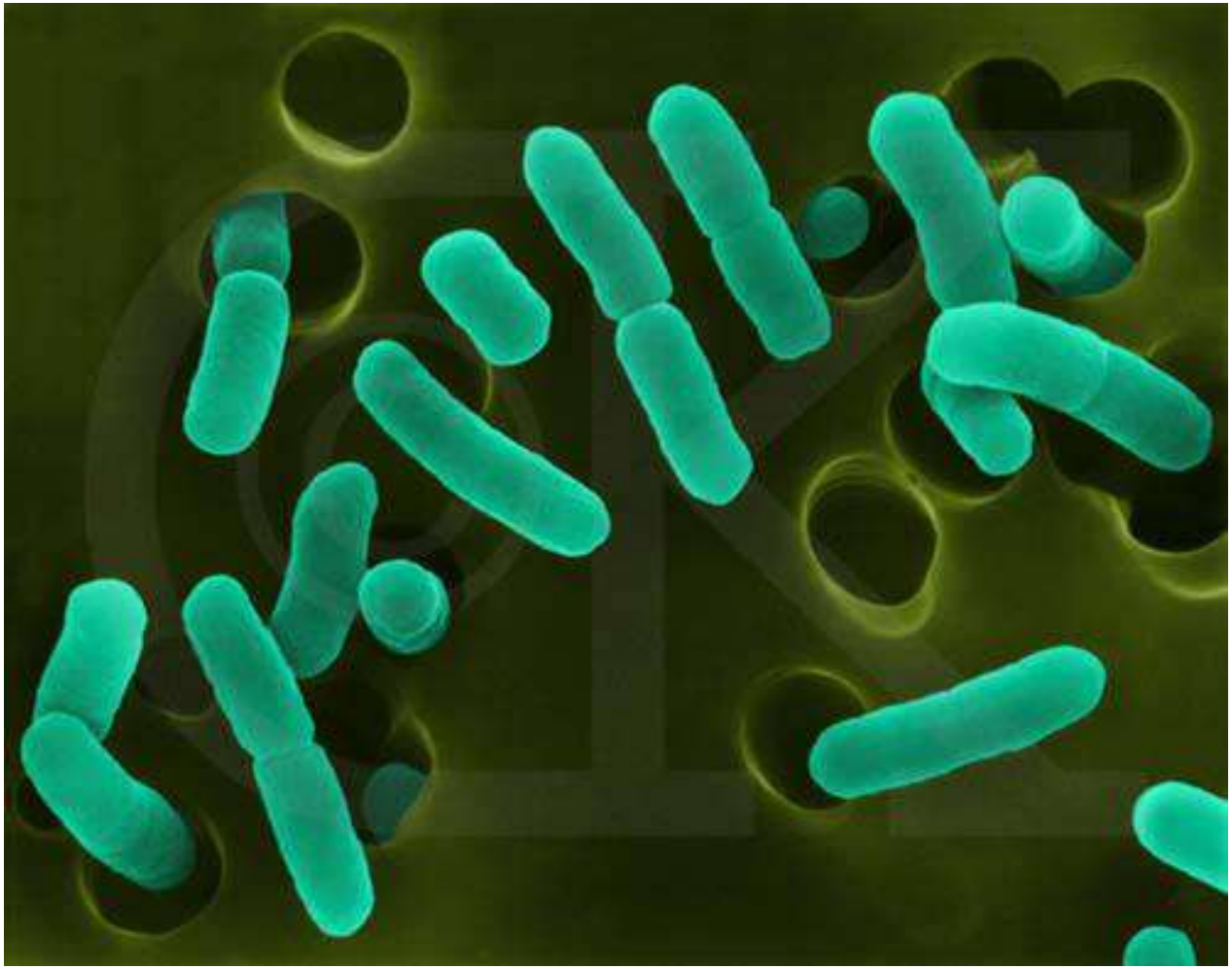


ENTEROBACTERIACEAE

Morphology & Identification

- **Gram-negative non-spore forming rods. When motile, by peritrichous flagella.**
- **Primarily normal flora of gastrointestinal tract. *E. coli>Klebsiella>Proteus>Enterobacter***
- **Free living, also transient colonizers of skin.**
- **Facultative anaerobes: mixed acid fermentation**
- **All ferment glucose; all reduce nitrates to nitrites; all oxidase negative.**
- **Lactose fermentation: normal flora positive and pathogens negative.**
- **Primary isolation media include eosin-methylene-blue (EMB) and MacConkey agar.**
- **Differential selective media for specific organisms including dyes and bile salts. (Salmonella-Shigella (SS) medium, bismuth sulfite media.)**

Enterobacteriaceae

- Small **gram-negative rods** (2-5 by 0.5 microns)
- Most motile with **peritrichous flagella**
  - *Shigella* and *Klebsiella* are nonmotile
- **Oxidase-negative facultative anaerobes**
- **Reduce nitrate**
- **Ferment glucose** and other carbohydrates
- Many genera
  - *Escherichia*, *Salmonella, Shigella, Klebsiella, Proteus, Enterobacter, Yersinia*, etc.
- Some strains **opportunistic** pathogens
- Some strains **true pathogens**
  - *Salmonella*, *Shigella*, *Yersinia*, some strains of *E. coli*

Classification

~29 genera, over 100 species.

- *Escherichia*
- *Shigella*
- *Edwardsiella*
- *Salmonella*
- *Citrobacter*
- *Klebsiella*
- *Enterobacter*
- *Hafnia*
- *Serratia*
- *Proteus*
- *Providencia*
- *Morganella*
- *Yersinia*
- *Erwinia*
- *Pectinobacterium*

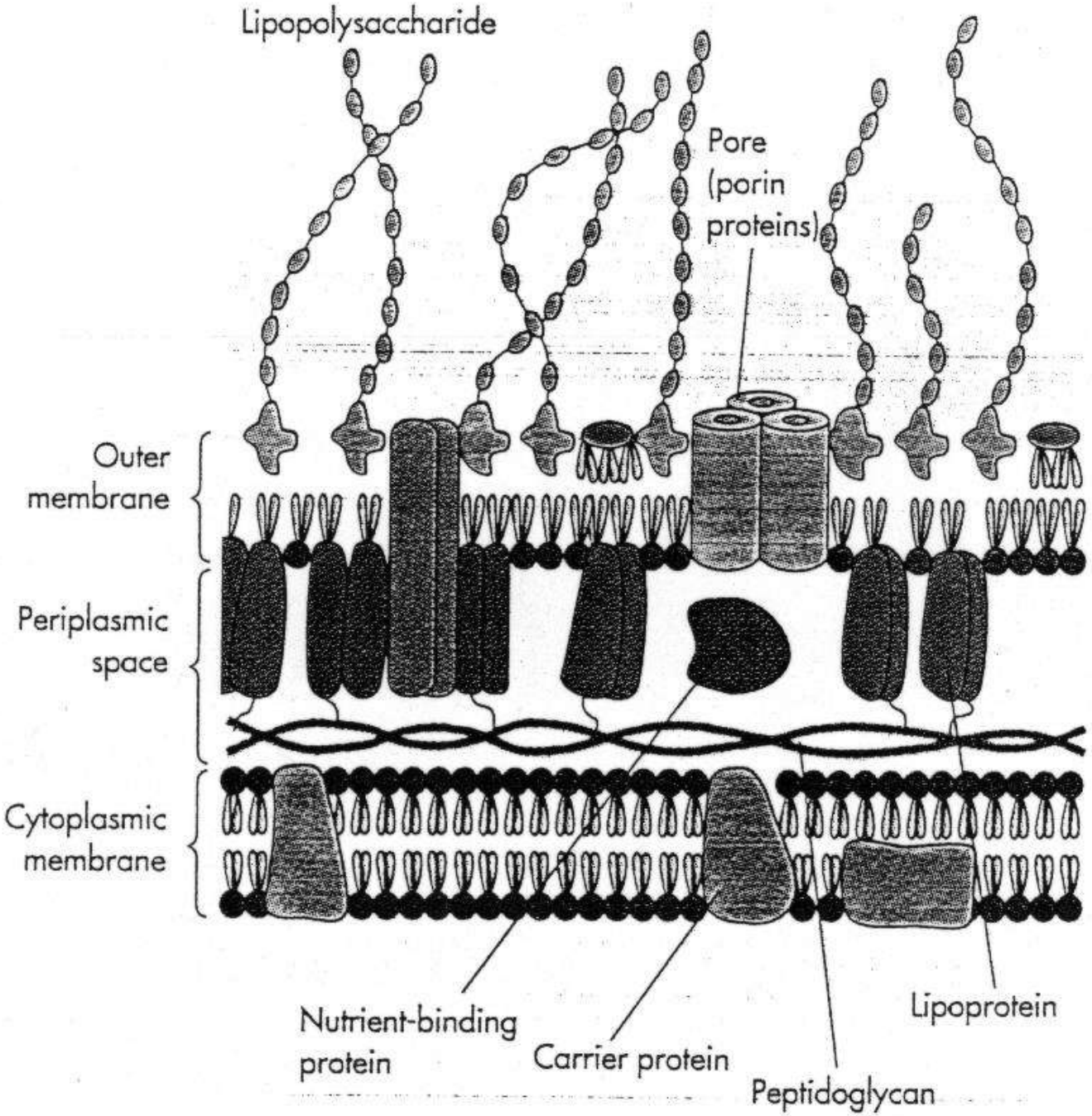
Antigenic Structure

- 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

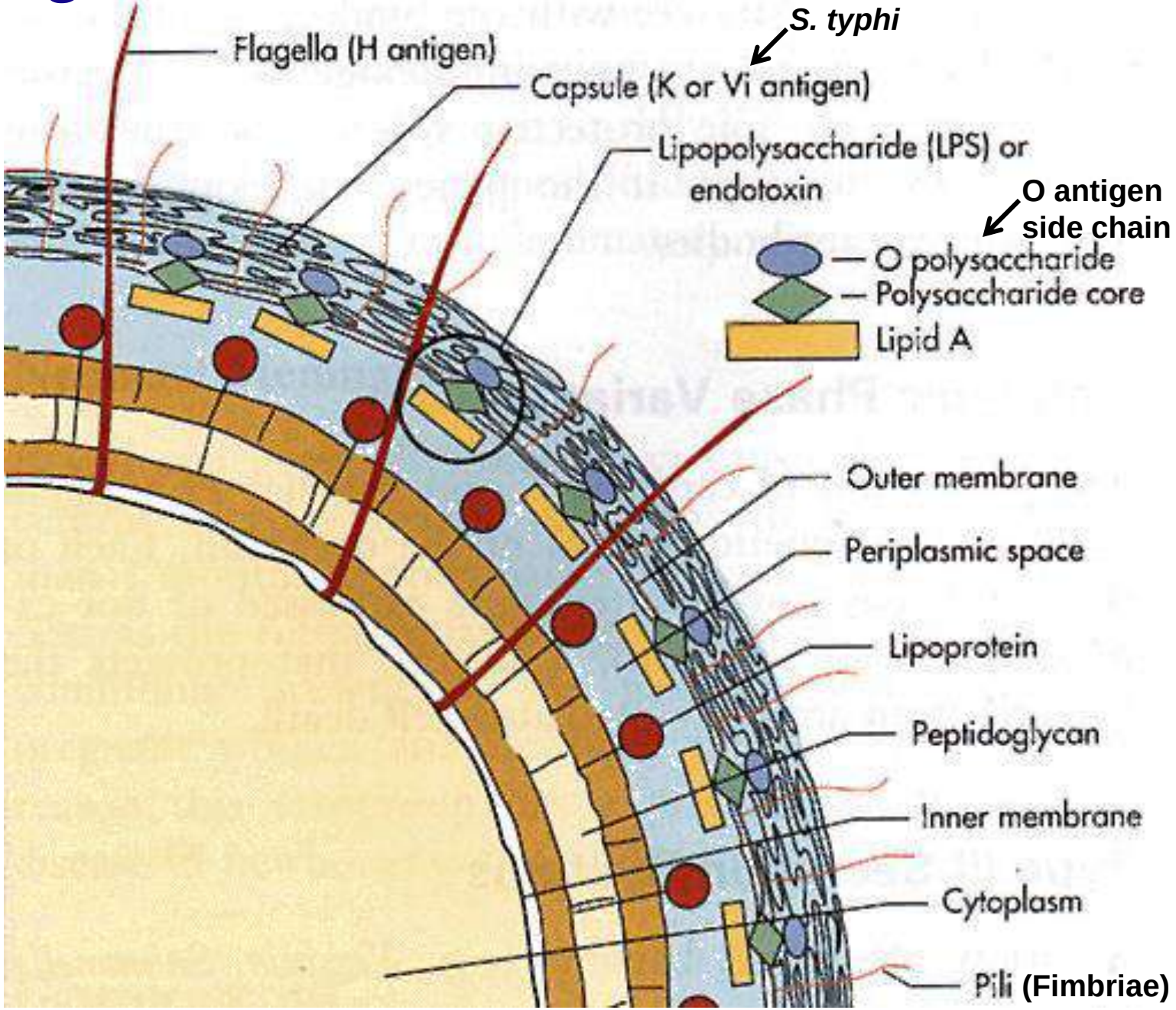
Distinguishing Properties Associated with All Enterobacteriaceae:

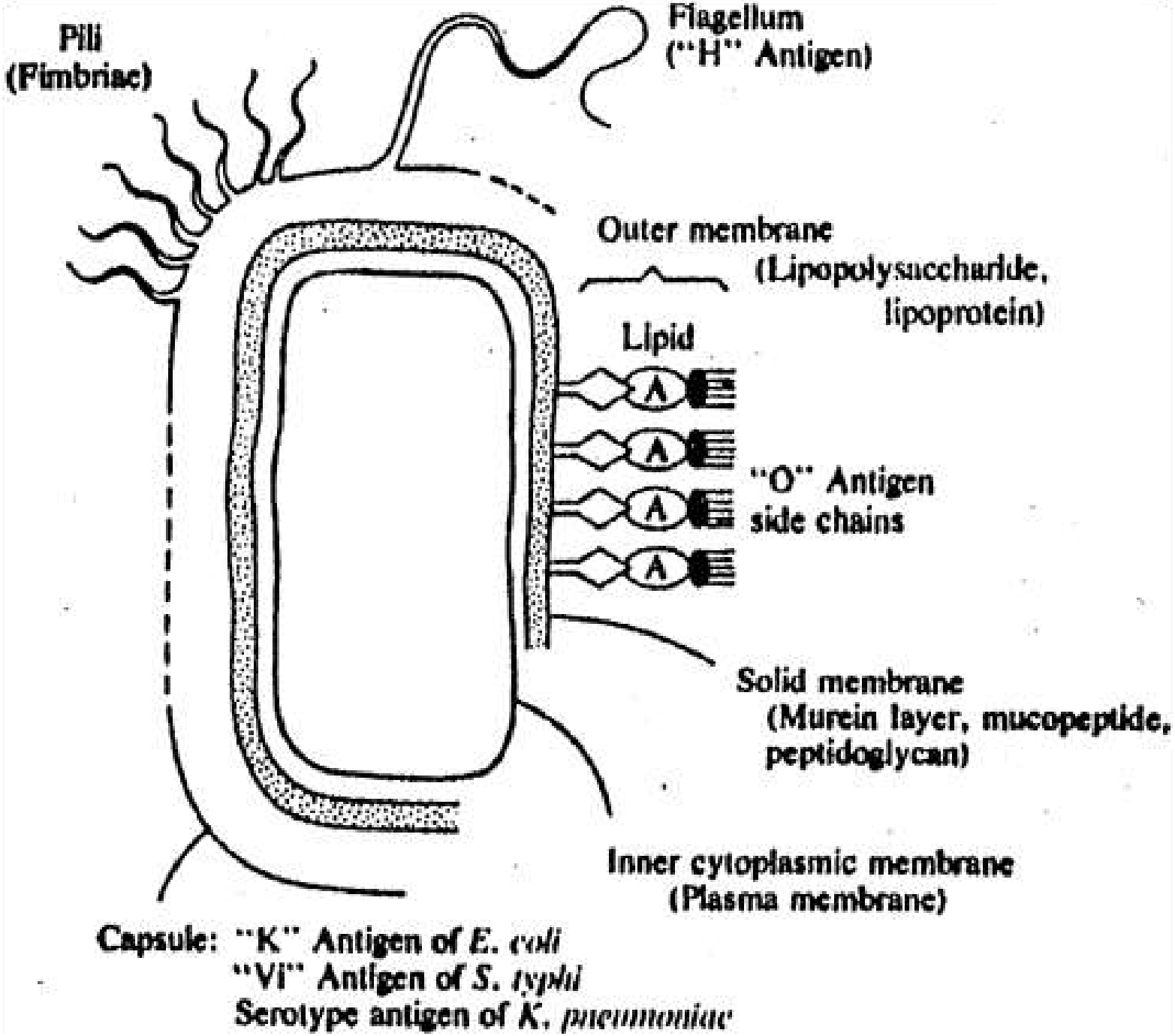
- **Ferment glucose**
- **Reduce nitrates**
  - NO_3 to NO_2 or all the way to N_2
- **Oxidase negative**

Gram-Negative Cell Wall

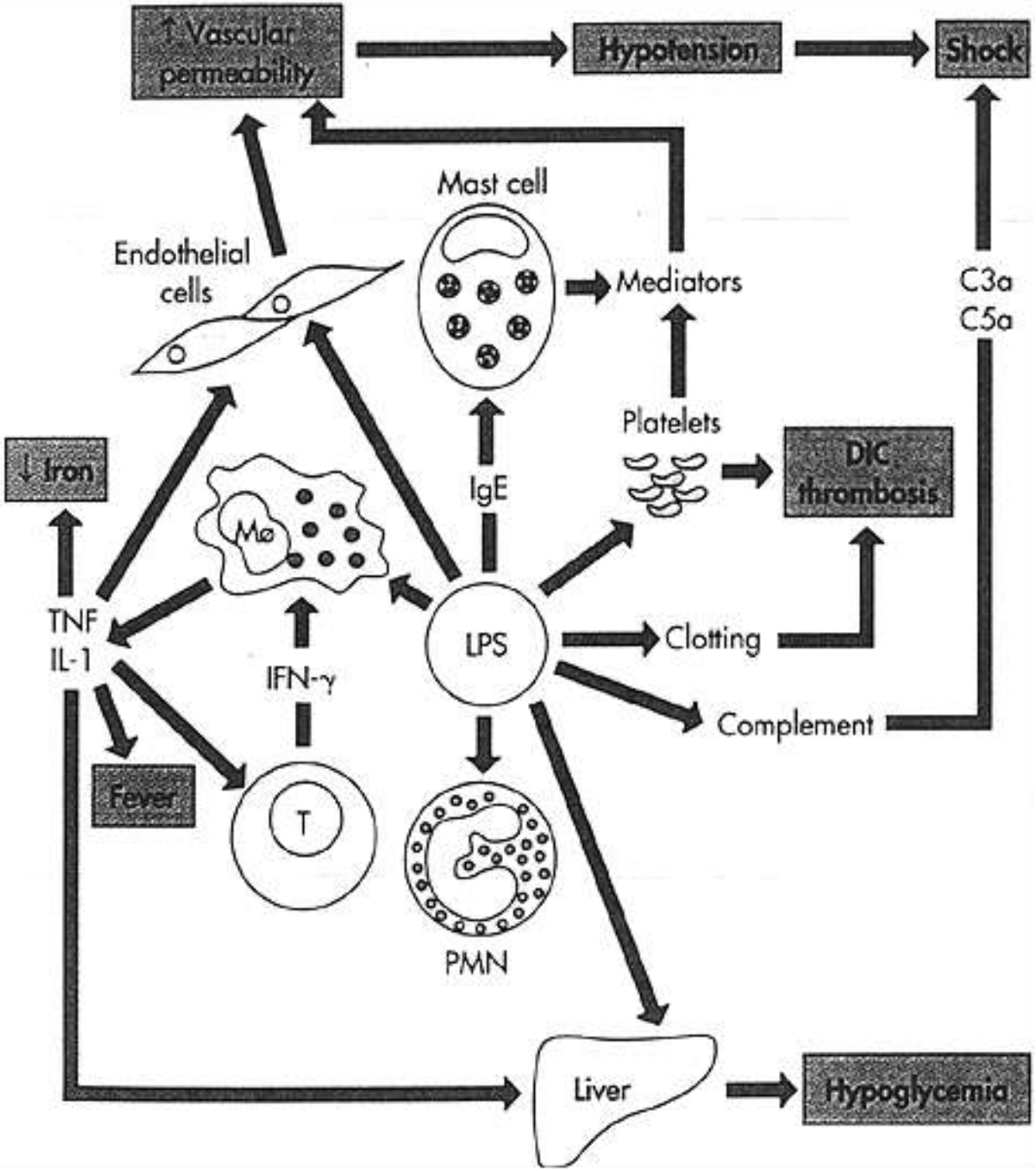


Antigenic Structure of Enterobacteriaceae




Pili
(Fimbriae)
Flagellum
("H" Antigen)
Outer membrane
(Lipopolysaccharide,
lipoprotein)
Lipid
"O" Antigen
side chains
Solid membrane
(Murein layer, mucopeptide,
peptidoglycan)
Inner cytoplasmic membrane
(Plasma membrane)
Capsule: "K" Antigen of E. coli
"Vi" Antigen of S. typhi
Serotype antigen of K. pneumoniae

Diversity of Activities Associated with LPS



Chapter 16 - Enterics

- Family *Enterobacteriaceae* often referred to as "enterics"
- Four major features:
  - All ferment glucose (dextrose)
  - All reduce nitrates to nitrites
  - All are oxidase negative
  - All except Klebsiella, Shigella and Yersinia are motile

Enteric Gram Negative rods

- Lactose Fermenting
  - Escherichia Coli
  - Klebsiella
  - Enterobacter
  - Citrobacter
- Lactose Non Fermenting
  - Salmonella
  - Shigella
  - Morganella
  - Providencia
  - Serratiia

Microscopic and Colony Morphology

- Gram negative bacilli or coccobacilli
- Non-spore forming
- Colony morphology on BAP or CA of little value, as they look the same, except for *Klebsiella*
- Selective and differential media are used for initial colony evaluation (ex. MacConkey, HE, XLD agars)

Classification of Enterics

- Due to the very large number of organisms in the Family *Enterobacteriaceae* (see Table 16-11), species are grouped into Tribes, which have similar characteristics (Table 16-1, page 466)
- Within each Tribe, species are further subgrouped under genera

Virulence and Antigenic Factors of Enterics

- Ability to colonize, adhere, produce various toxins and invade tissues
- Some possess plasmids that may mediate resistance to antibiotics
- Many enterics possess antigens that can be used to identify groups
  - O antigen – somatic, heat-stable antigen located in the cell wall
  - H antigen – flagellar, heat labile antigen
  - K antigen – capsular, heat-labile antigen

Clinical Significance of Enterics

- Enterics are ubiquitous in nature
- Except for few, most are present in the intestinal tract of animals and humans as commensal flora; therefore, they are sometimes call "fecal coliforms"
- Some live in water, soil and sewage

Clinical Significance of Enterics (cont'd)

- Based on clinical infections produced, enterics are divided into two categories:
  - Opportunistic pathogens – normally part of the usual intestinal flora that may produce infection outside the intestine
  - Primary intestinal pathogens – *Salmonella*, *Shigella*, and *Yersinia* sp.

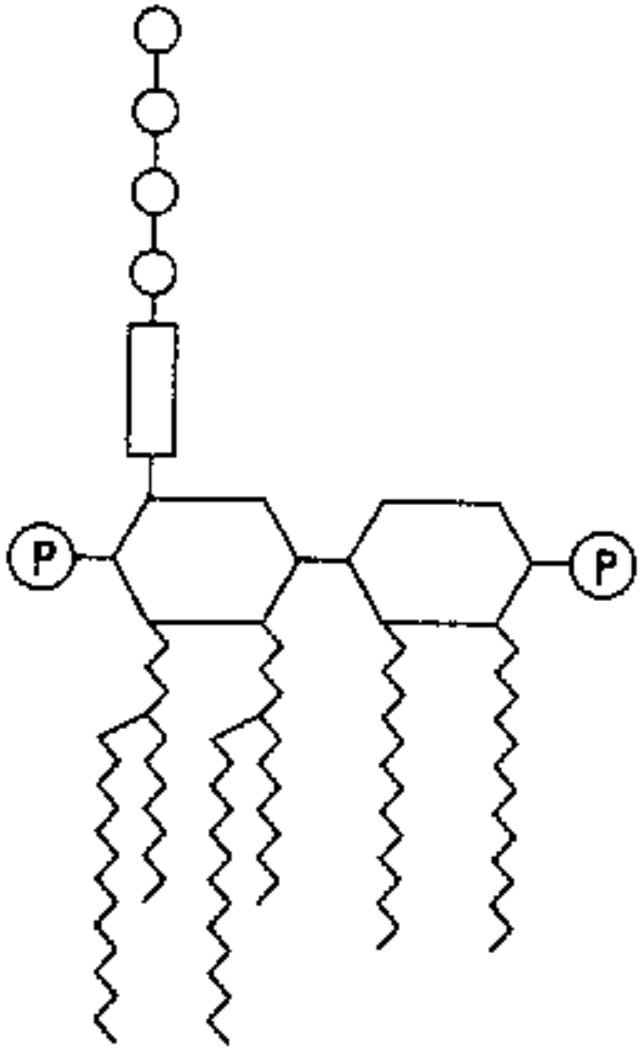
EXOTOXIN

1. Released from the cell before or after lysis
2. Protein
3. Heat labile
4. Antigenic and immunogenic
5. Toxoids can be produced
6. Specific in effect on host
7. Produced by gram-positive and gram-negative organisms

ENDOTOXIN

1. Integral part of cell wall
2. Endotoxin is LPS; Lipid A is toxic component
3. Heat stable
4. Antigenic; ??immunogenicity
5. Toxoids cannot be produced
6. Many effects on host
7. Produced by gram-negative organisms only

Structure of Lipopolysaccharide



Repeat units (up to 40)

Core

Disaccharide-diphosphate

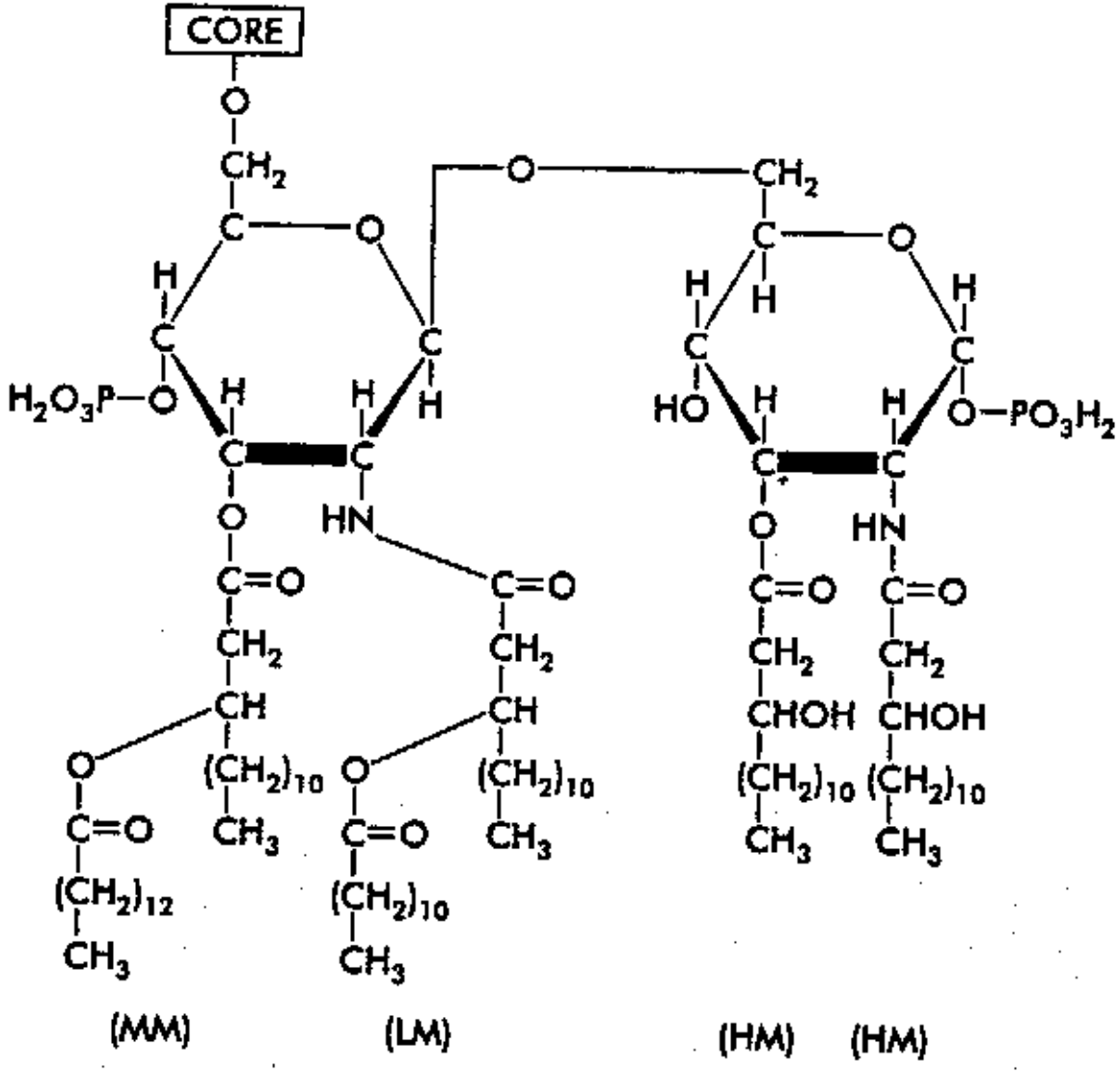
Fatty acids

O Antigen

Core polysaccharide

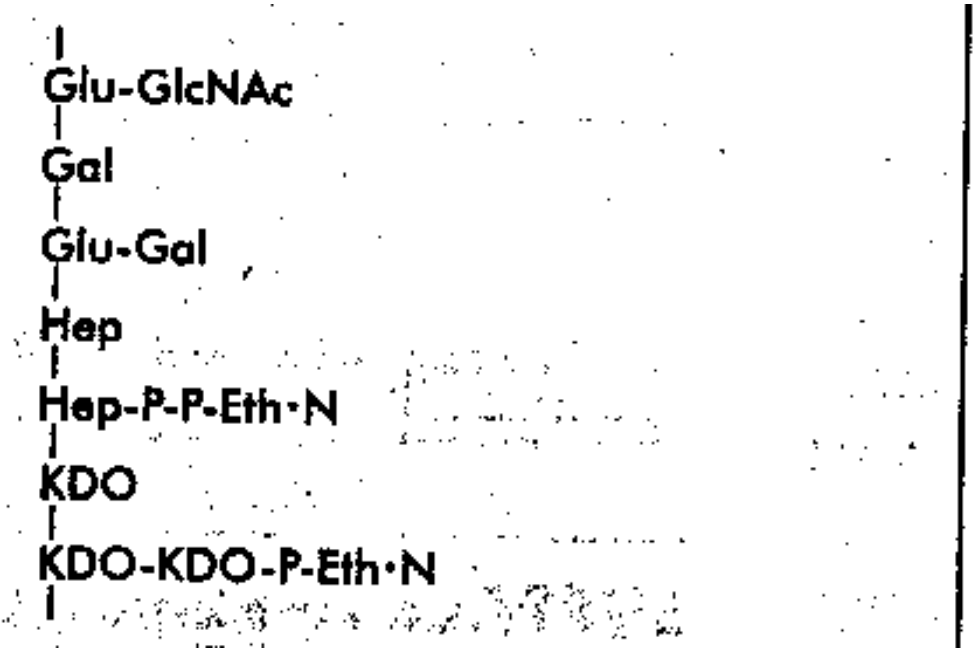
Lipid A

Structure of Lipid A



- **Hydrophobic** Lipid A is **endotoxic component**

Structure of Core Polysaccharide

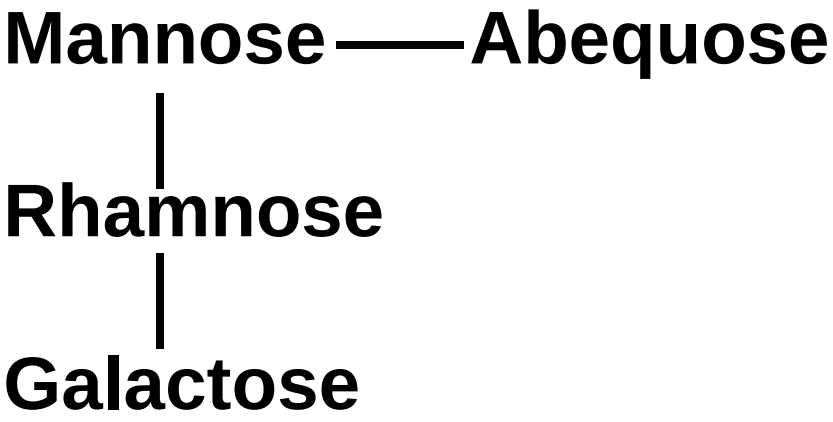


KDO = Keto-deoxy-octulonate
Hep = L-Glycero-D-mannoheptose
HM = β-Hydroxymyristic acid (C_{14})
LM = Lauroxymyristic acid
MM = Myristoxymyristic acid
Eth·N = Ethanolamine
Glu = Glucose
GlcNAc = N-Acetyglucosamine
Gal = Galactose

- **KDO** is **distinctive sugar** moiety in core polysaccharide

Repeat Units of O Antigen Side Chain

Example: (Repeated up to 40 times)



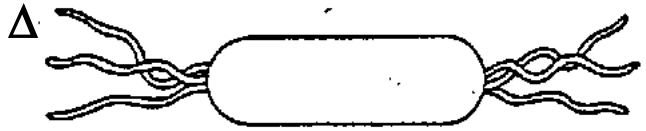
- **Heat stable** O antigen is often used to **serotype**

Taxonomic Descriptions of Prokaryotes Based on Numbers and Arrangements of Flagella

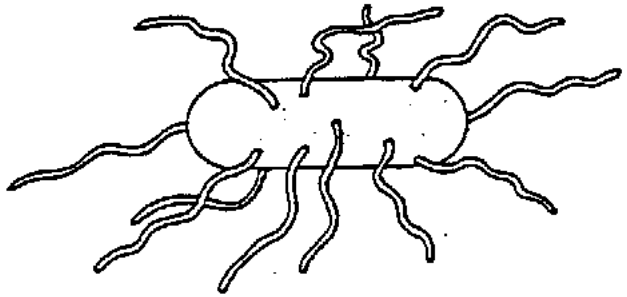
Terminology	Flagellum arrangement
Atrichous	No flagella
Monotrichous	One flagellum at one end

Amphitrichous	One or more flagella at each end

Lophotrichous	Tuft of Two or more flagella at one or both ends

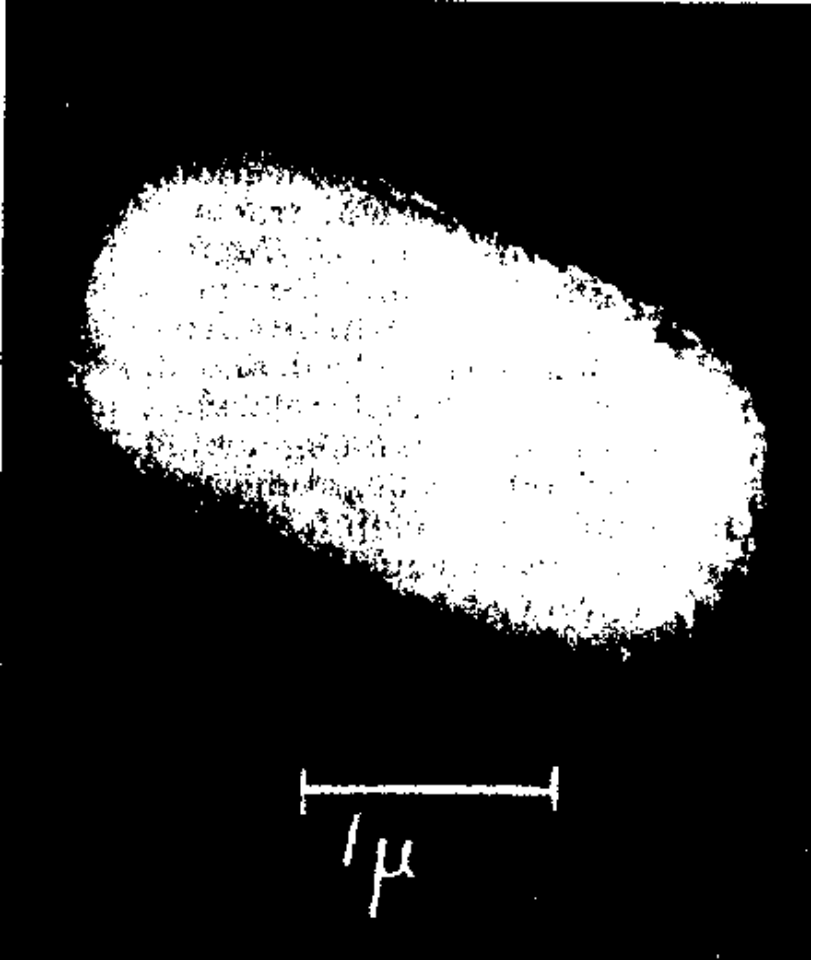


Peritrichous	Flagella surrounding the cell



Taxonomic Descriptions of Prokaryotes Based on Numbers and Arrangements of Flagella (cont.)

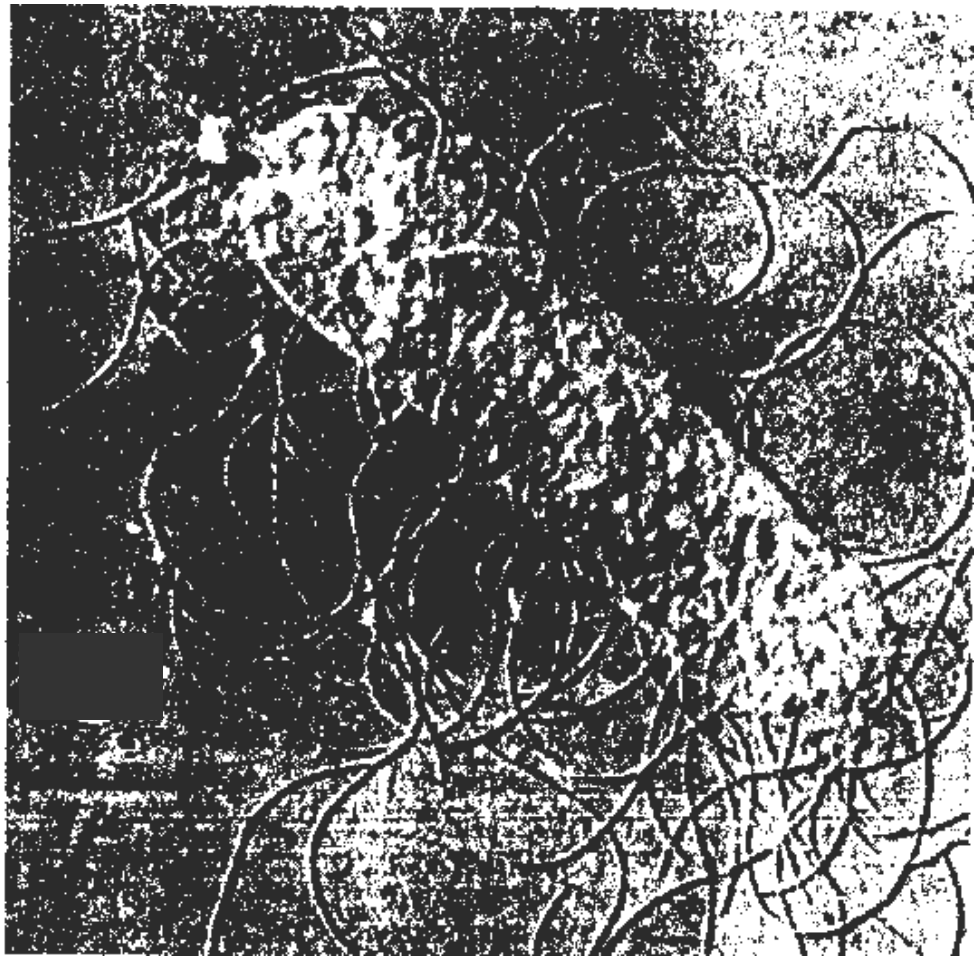
Polar	**Monotrichous**, a single flagellum at one or both ends of the cell
	Multitrichous, two or several flagella at one or both ends of the cell
Lateral	Flagella arise predominantly from the **middle pole** of the cell
	Monotrichous, one flagellum
	Multitrichous, several flagella in the form of a tuft originating from the midportion of the cell
Peritrichous	**Random**, haphazard arrangement of flagella scattered around the bacterial cell
Mixed	Two or more flagella exhibiting distinctly different physical properties in different regions of the bacterial cell



Escherichia coli

Proteus vulgaris

- **Hypermotile**
- **Swarming growth**



Family Enterobacteriaceae

	Genus	No. of species
	Citrobacter	4
	Edwardsiella	4
	Enterobacter	13
Certain *E .coli* strains can be considered true pathogens →	*Escherichia*	5
True pathogen →	*Shigella* (nonmotile)	4
	Ewingella	1
	Hafnia	2
	Klebsiella (nonmotile)	7
	Kluyvera	2
	Morganella	2
	Proteus	4
	Providencia	5
True pathogen →	*Salmonella*	7 subgroups
	Serratia	10
True pathogen →	*Yersinia*	11

Medically Important Enterobacteriaceae

Citrobacter species

Enterobacter spp.

Escherichia spp.

Klebsiella spp.

Morganella spp.

Proteus spp.

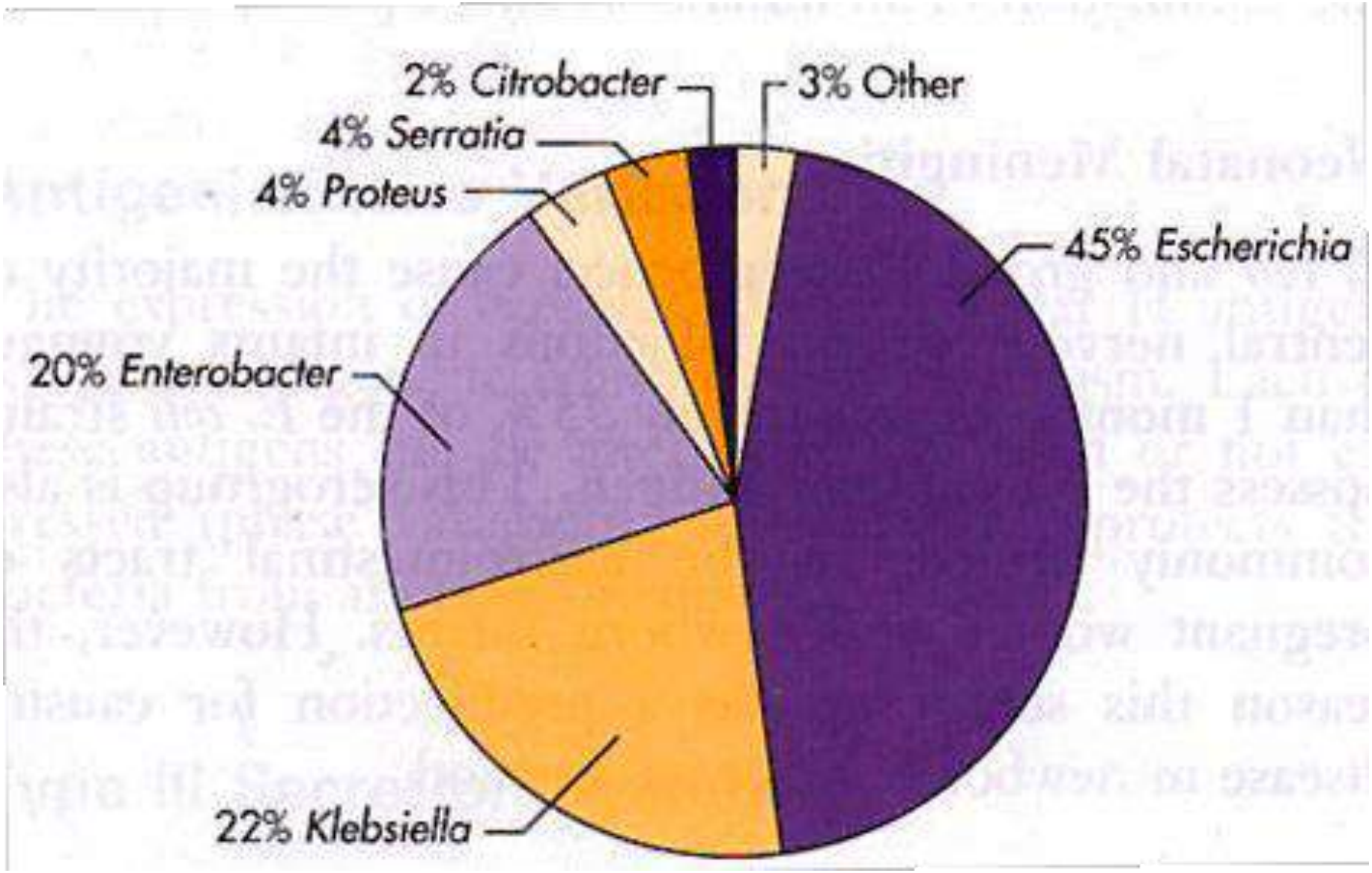
Salmonella spp.

Serratia spp.

Shigella spp.

Yersinia spp.

Incidence of Enterobacteriaceae *Associated with Bacteremia*



Enterobacteriaceae: Types of Infectious Disease

- **Intestinal (diarrheal) infection**
- **Extraintestinal infection**
  - **Urinary tract (primarily cystitis)**
  - **Respiratory (nosocomial pneumonia)**
  - **Wound (surgical wound infection)**
  - **Bloodstream (gram-negative bacteremia)**
  - **Central nervous system (neonatal meningitis)**

Enterobacteriaceae: Urinary Tract Infection, Pneumonia

- **Urinary tract infection: *Escherichia coli*, *Klebsiella pneumoniae*, *Enterobacter* spp., and *Proteus mirabilis***
- **Pneumonia: *Enterobacter* spp., *Klebsiella pneumoniae*, *Escherichia coli*, and *Proteus mirabilis***

Enterobacteriaceae: Wound Infection, Bacteremia

- **Wound Infection: *Escherichia coli*, *Enterobacter* spp., *Klebsiella pneumoniae*, and *Proteus mirabilis***
- **Bacteremia: *Escherichia coli*, *Enterobacter* spp., *Klebsiella pneumoniae*, and *Proteus mirabilis***

Triple Sugar Iron (TSI) Agar

- **Yeast extract 0.3% (% = grams/100 mL)**
- **Beef extract 0.3%**
- **Peptone 1.5%**
- **Proteose peptone 0.5%**

Total Protein = 2.6%

- **Lactose 1.0%**
- **Sucrose[1] 1.0%**
- **Glucose 0.1%**

Carbohydrate = 2.1%

[1]Absent in Kligler Iron Agar

Triple Sugar Iron (TSI) Agar

- **Sodium thiosulfate**
- **Sodium chloride**
- **Agar (1.2%)**
- **Phenol red**
- **pH = 7.4 Ferrous sulfate**

TSI Reactions of the *Enterobacteriaceae*

- **Yellow deep, purple slant: acid deep due to glucose fermentation , no lactose or sucrose fermentation with alkaline slant due to production of amine's from protein**
- **Black deep, purple slant: acid deep due to glucose fermentation with H_2S production, no lactose or sucrose fermentation**
- **Yellow deep and slant: acid deep and slant due to glucose as well as lactose and/or sucrose fermentation**
- **Black deep and yellow or black slant: acid deep and slant with glucose and lactose and/or sucrose fermentation with H_2S production**
- **Fracturing or lifting of agar from base of culture tube: CO2 production**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TSI Reactions of the *Enterobacteriaceae*

- **A/A + g = acid/acid plus gas (CO_2)**
- **A/A = acid/acid**
- **A/A + g, H_2S = acid/acid plus gas, H_2S**
- **Alk/A = alkaline/acid**
- **Alk/A + g = alkaline/acid plus gas**
- **Alk/A + g, H_2S = alkaline/acid plus gas, H_2S**
- **Alk/A + g, H_2S (w) = alkaline/acid plus gas, H_2S (weak)**

A/A + g

- ***Escherichia coli***
- ***Klebsiella pneumoniae***
- ***Klebsiella oxytoca***
- ***Enterobacter aerogenes***
- ***Enterobacter cloacae***
- ***Serratia marcescens*[1, 2]**

[1]Non-lactose, sucrose fermenter

[2]55% + g

A/A

- ***Serratia marcescens*[1, 2]**
- ***Yersinia enterocolitica*[2]**

[1]45% of strains

[2]Non-lactose, sucrose fermenter

A/A + g, H_2S

- ***Citrobacter freundii***
- ***Proteus vulgaris***[1]

[1]**Non-lactose, sucrose fermenter**

Alk/A

- ***Shigella***
- ***Providencia***

Enterobacteriaceae- Opportunistic diseases

Citrobacter
Enterobacter
Escherichia
Hafnia
Morganella
Providencia
Serratia

- **septicemia,**
- **pneumonia,**
- **meningitis**
- **urinary tract infections**

Enterobacteriaceae:

gastrointestinal diseases

- ***Escherichia coli***
- ***Salmonella***
- ***Shigella***
- ***Yersinia entercolitica***

Enterobacteriaceae

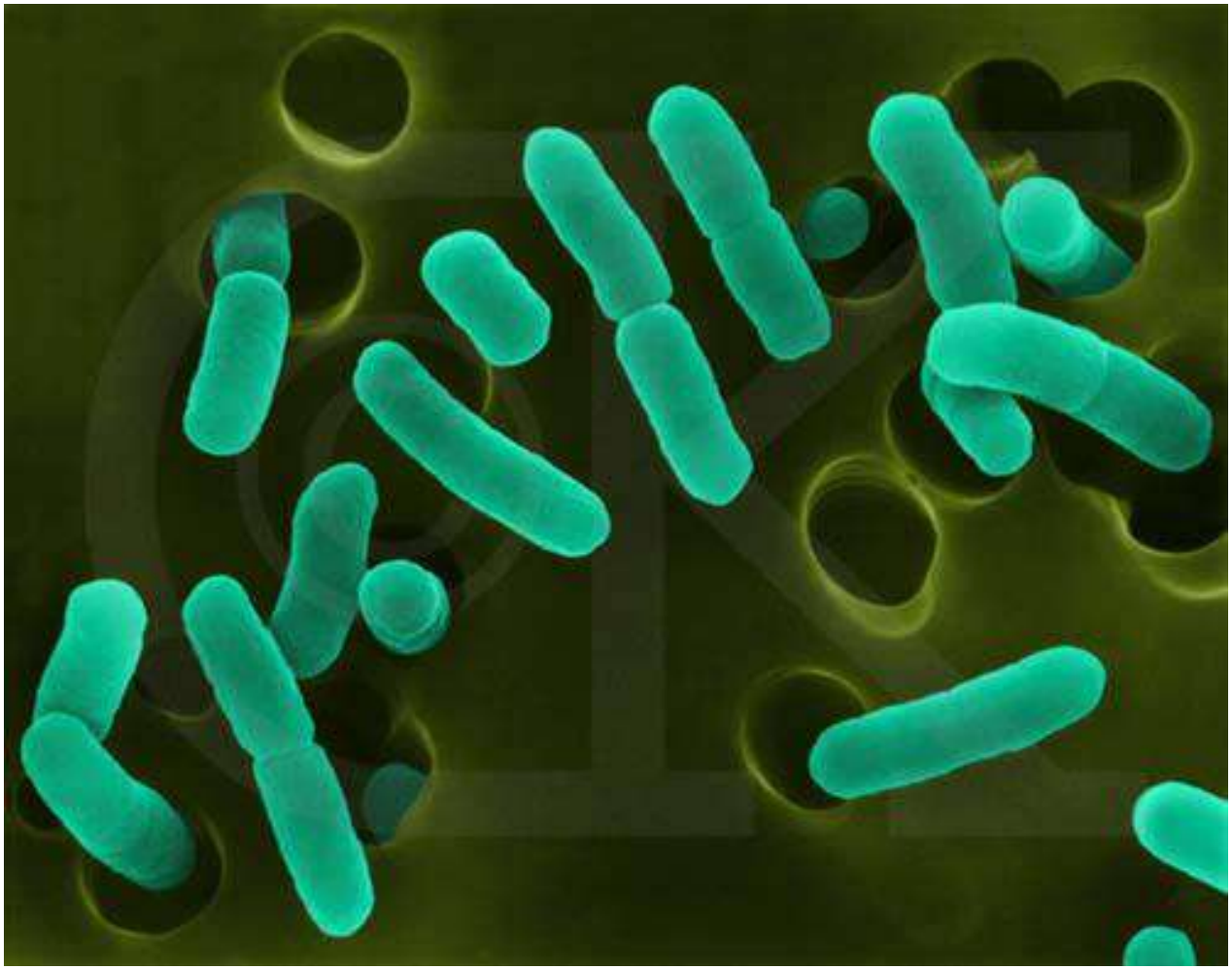
- **community acquired**
- **otherwise healthy people**
  - *Klebsiella pneumoniae*
    - * respiratory diseases
    - * prominent capsule

  - **urinary tract infection**
    - **fecal contamination**
    - ******E. coli*
    - ******Proteus*
      - **urease (degrades urea)**
      - **alkaline urine**

Enteric Gram Negative rods

- Lactose Fermenting
  - Escherichia Coli
  - Klebsiella
  - Enterobacter
  - Citrobacter
- Lactose Non Fermenting
  - Salmonella
  - Shigella
  - Morganella
  - Providencia
  - Serratiia

Escherichia coli



E. coli

Pathogenesis and clinical diseases

Urinary tract infection

E. coli is the most common cause of urinary tract infection.

Community- vs. hospital-acquired UT infection

Most infections originate from colon; the bacteria contaminate the urethra, ascend into the bladder, and may migrate into the kidney or prostate.

Symptoms: urinary frequency, dysuria, hematuria, and pyuria. Can result in bacteremia and sepsis.

Uropathogenic *E. coli* strains produce P (Pyelonephritis-associated) pili, which is associated with renal colonization and may induce protective immunity, and hemolysin HlyA.

E. coli

- Certain strains of *E. coli*
- ingested in sufficient quantities by host.
- enteritis, enterocolitis, and colitis
- pathogenic strains of E. coli which cause disease in the intestine are obligate pathogens.

six distinct "pathotypes" of E. coli (which cause disease in intestine)

- Enteropathogenic E. coli (EPEC),
- Enterotoxigenic E. coli (ETEC),
- Shiga toxin-producing E. coli (STEC)/enterohemorrhagic E. coli (EHEC), Verocytotoxin-producing E. coli (VTEC)
- Enteroinvasive E. coli (EIEC),
- Enteroaggregative E. coli (EAEC) (EAggEC)
- diffusely adherent E. coli (DAE)

Enteropathogenic *E. coli*

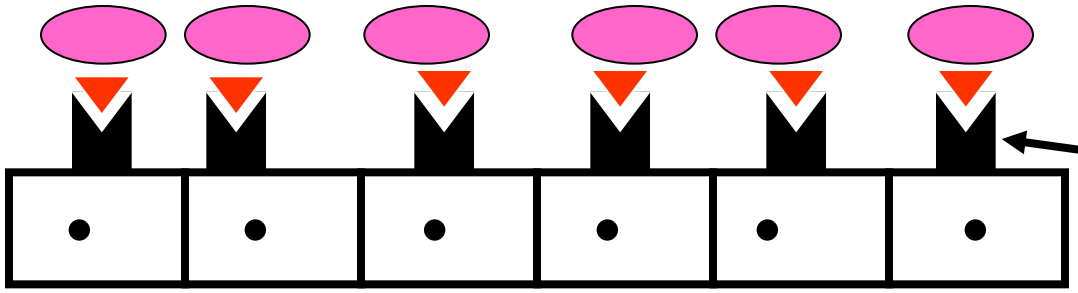
- EPEC is an important cause of infant diarrhea
- Breast-feeding diminishes the incidence of EPEC infection.
- Rapid person-to-person spread may occur.
- After infection with EPEC there is loss of microvilli.
- Sometimes the EPEc enter the intestinal cells.

Escherichia coli

- **Toxins: two types of enterotoxin; Shiga-type toxin; Enteroaggregative ST-like toxin; Hemolysins; Endotoxin**
- **Type III secretion system**
- **Adhesions –colonization factors ; both pili or fimbriae ;non-fimbrial factors involved in attachment. There are at least 21 different types of adhesions.**
- **Virulence factors that protect the bacteria from host defenses: Capsule/Iron capturing ability (enterochelin)**
- **Outer membrane proteins**

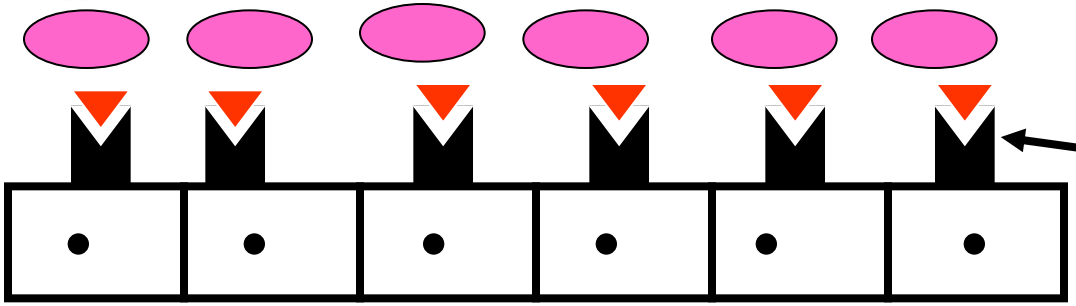
E. coli fimbriae

Type 1



mannose

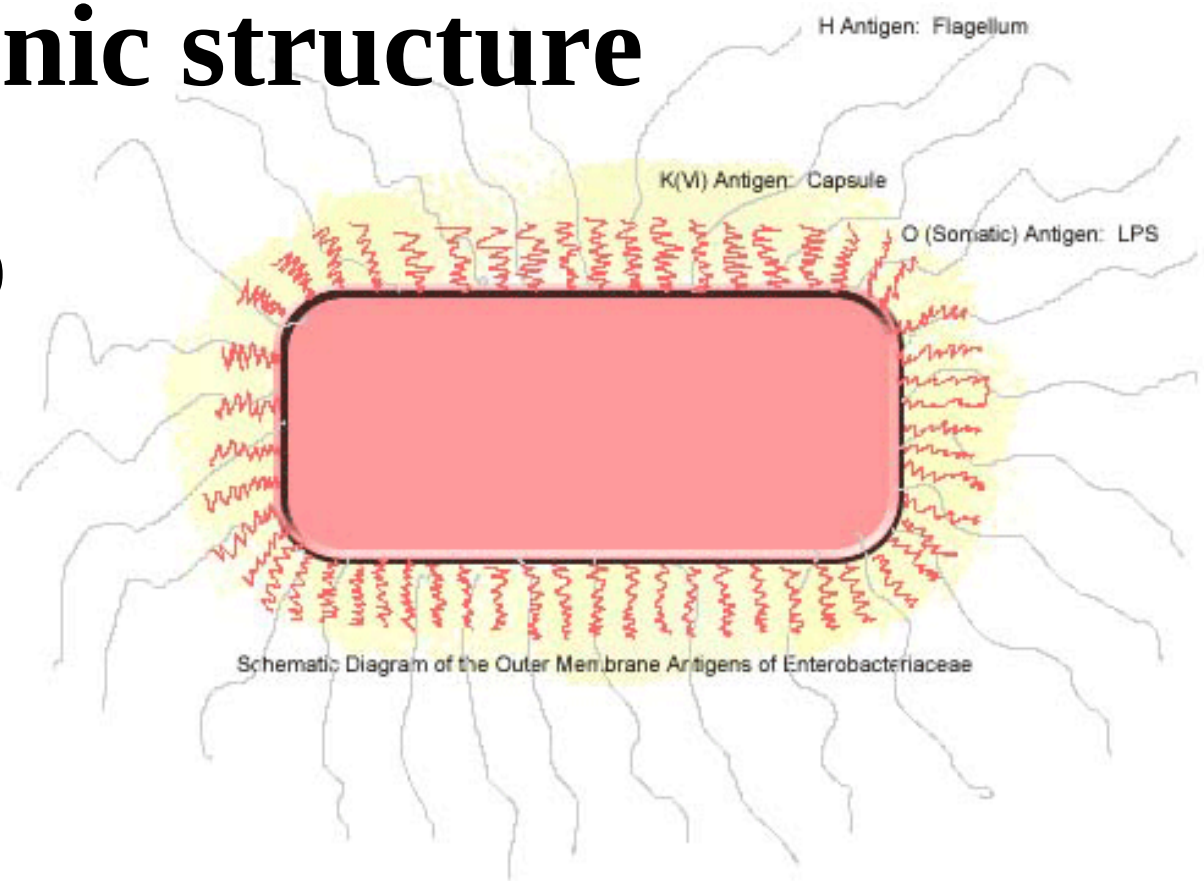
P



- galactose
  - glycolipids
  - glycoproteins

Antigenic structure

- O antigen
  - Somatic (on LPS)
  - 171 antigens
- H antigen
  - Flagella
  - 56 antigens
- K antigen
  - Capsule and or fimbrial antigen
  - 80 antigens



O18ac:H7:K1

18th O antigen

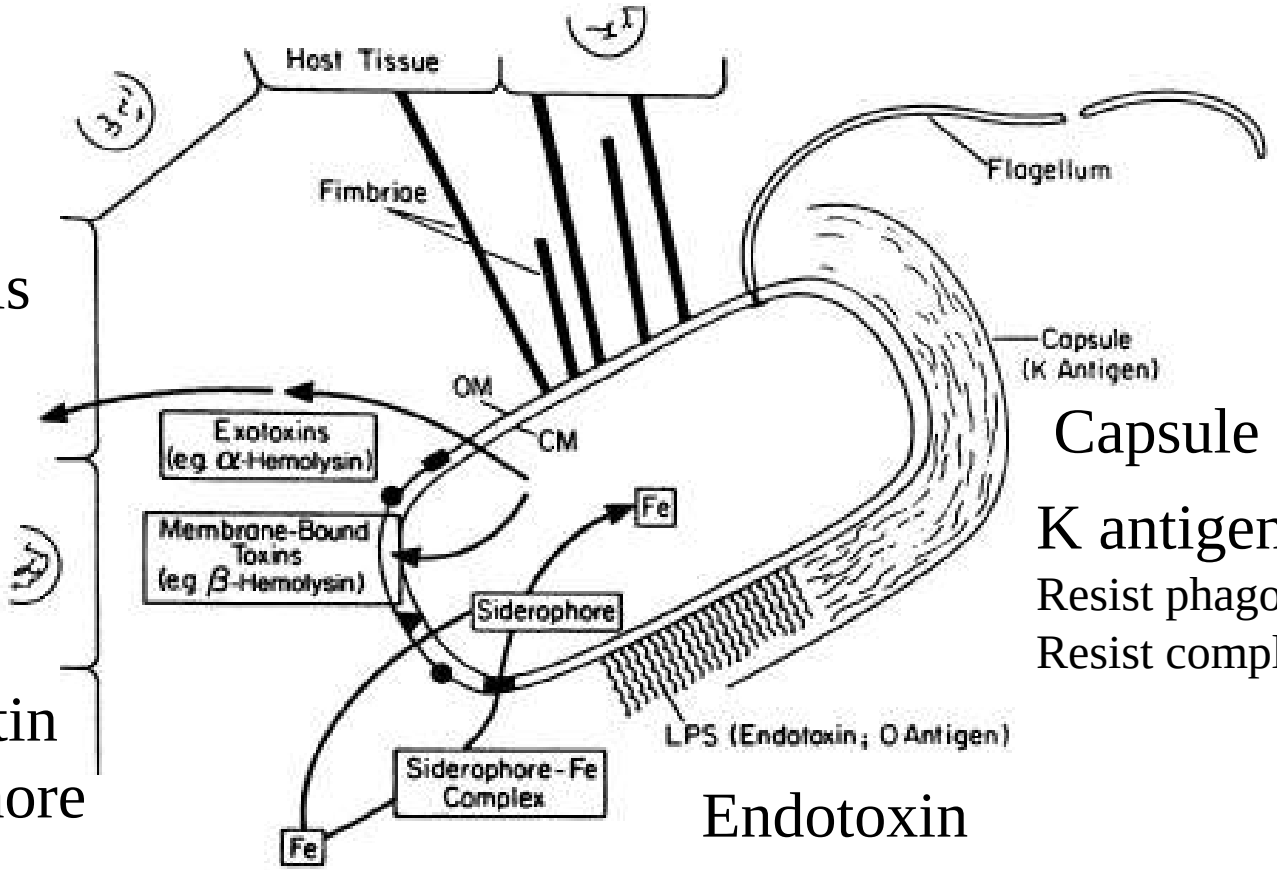
7th H antigen

1st K antigen

Urovirulence Factors

Adherence
Fimbriae

Hemolysis
α
β



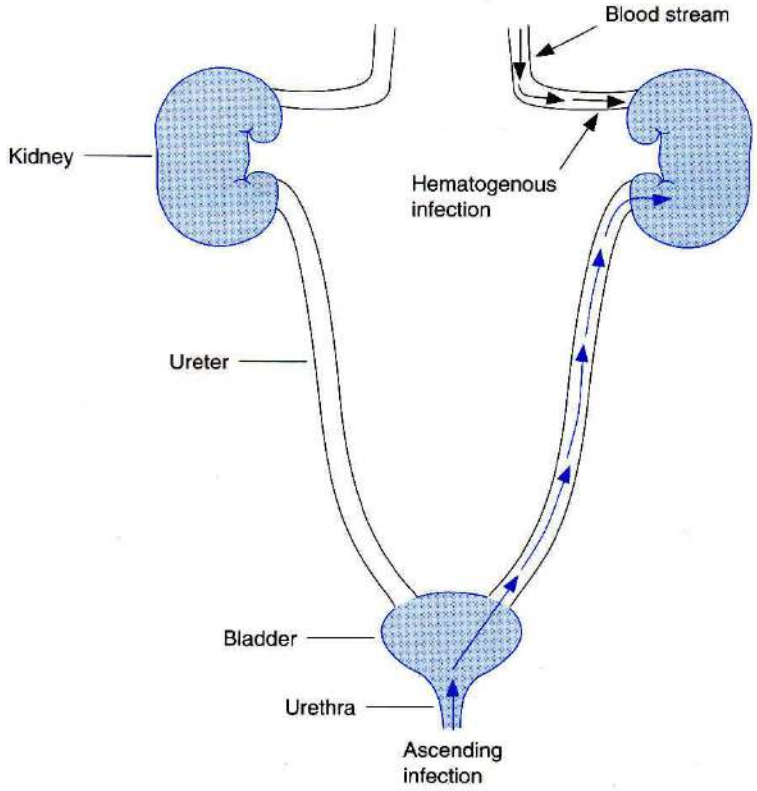
Capsule

K antigens
Resist phagocytosis
Resist complement proteins

Aerobactin
Siderophore

Endotoxin

FIG. 1. Schematic representation of an *E. coli* cell interacting with host tissue, highlighting features relevant to bacterial pathogenicity. Membrane proteins involved in transport, serum resistance, etc., are indicated by solid black circles, triangles, and rectangles. OM, Outer membrane; CM, cytoplasmic membrane; LPS, lipopolysaccharide. Adapted from reference 111, with permission from the publisher.



E.coli-urinary tract infection

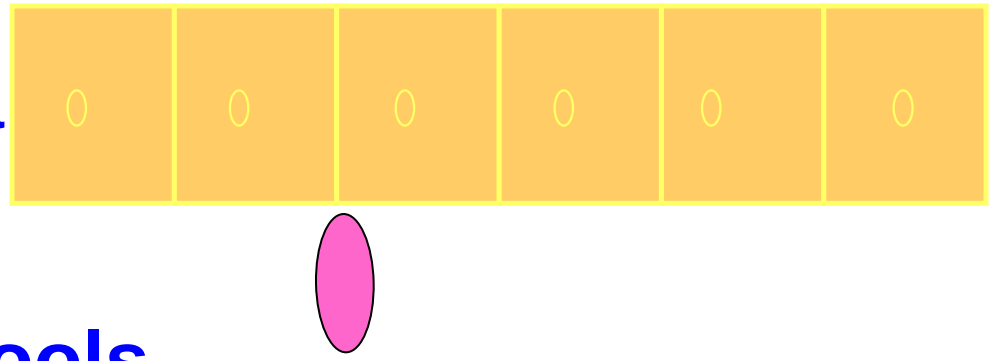
Is the leading cause of urinary tract infections which can lead to acute cystitis (bladder infection) and pyelonephritis (kidney infection).

E.coli-Meningitis and Sepsis

- **Neonatal meningitis – is the leading cause of neonatal meningitis and septicemia with a high mortality rate. Usually caused by strains with the K1 capsular antigen.**

Enteropathogenic *E. coli*

- fever
- infant diarrhea
- vomiting
- nausea
- non-bloody stools
- Destruction of surface microvilli



1. loose attachment mediated by bundle forming pili (Bfp);
2. Stimulation of intracellular calcium level;
3. rearrangement of intracellular actin,

Enterotoxigenic *E. coli*

- A watery diarrhea, nausea, abdominal cramps and low-grade fever for 1-5 days.
- Travellers diarrhea and diarrhea in children in developing countries
- Transmission is via contaminated food or water.

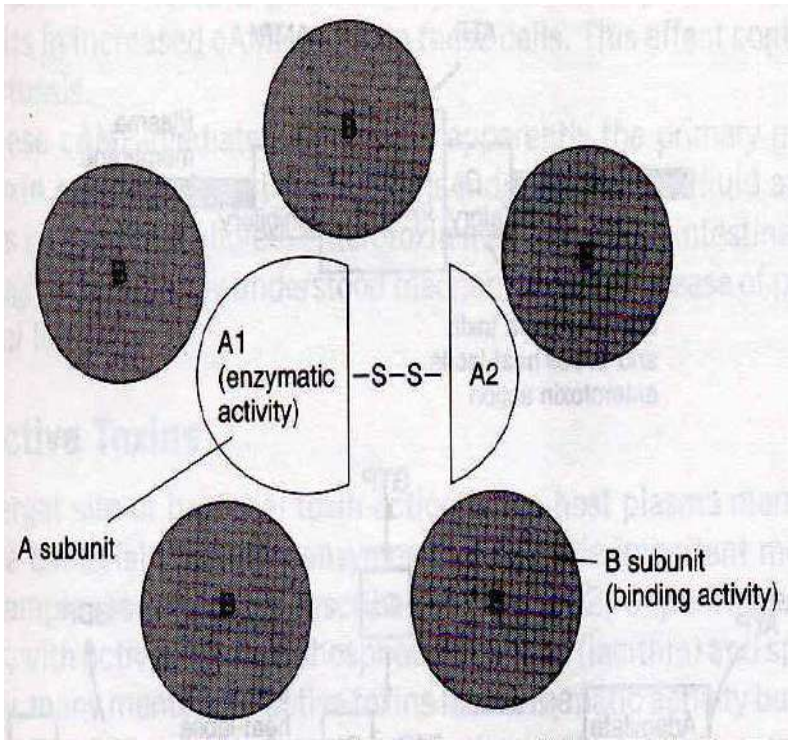
Enterotoxigenic *E. coli*

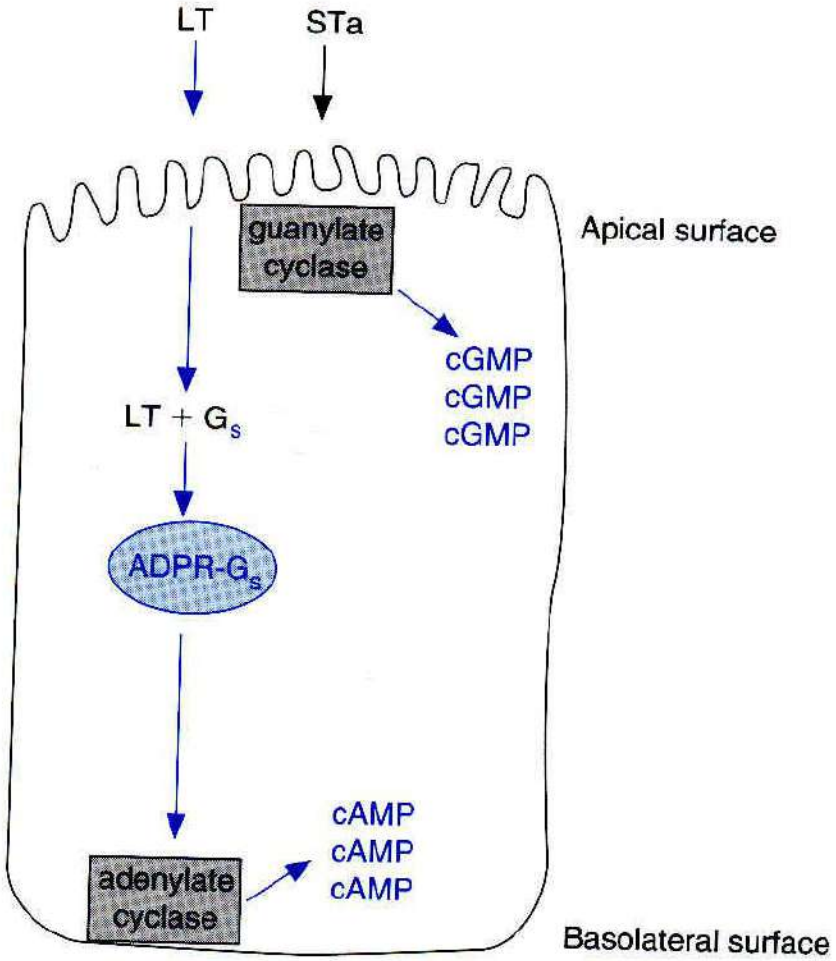
- diarrhea like cholera
- milder
- nursery travellers diarrhea
- caused by LT, ST, or LT/ST.

Enterotoxigenic *E. coli*

- **Heat labile toxin**
  - **like choleragen**
  - **Adenyl cyclase activated**
  - **cyclic AMP** ↑
  - **secretion water/ions** ↑
- **Heat stable toxin**
  - **Guanylate cyclase activated**
  - **cyclic GMP** ↑
  - **uptake water/ions** ↓

LT vs ST activity



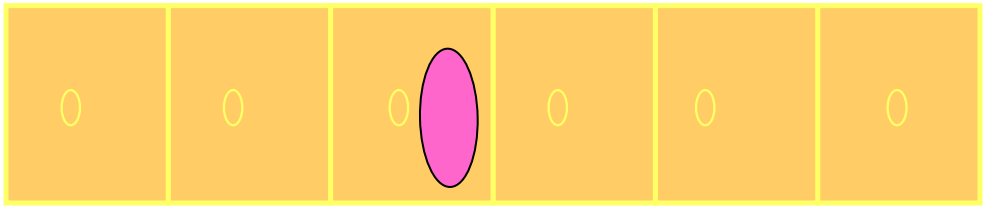


E.coli-Enteroinvasive (EIEC)

- **The organism attaches to the intestinal mucosa via pili**
- **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.**
- **resembles shigellosis**

Enteroinvasive *E. coli* (EIEC)

- **Dysentery**
  - **resembles shigellosis**
  - **elder children and adult diarrhea**



E.coli-c. Enteropathogenic (EPEC)

- **Malaise and low grade fever diarrhea, vomiting, nausea, non-bloody stools**
- **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.**
- **This is a problem mainly in hospitalized infants and in day care centers.**

"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

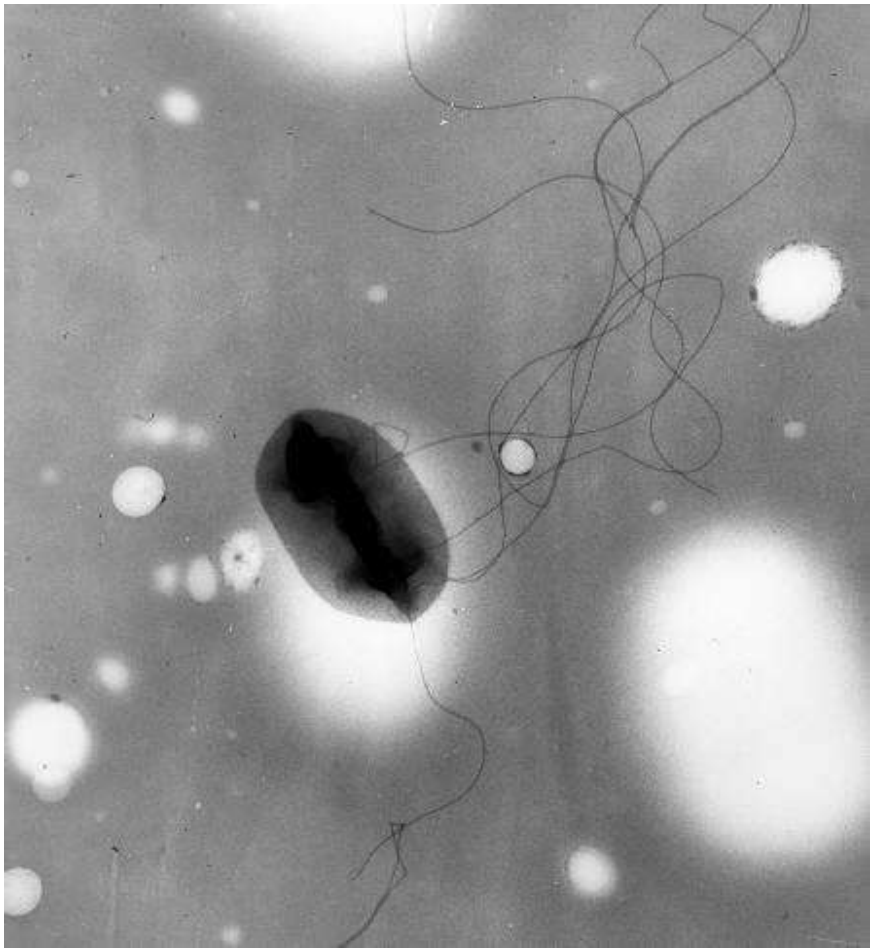
"Intimate" contact:
Pedastallike structure (composed of actin fibers) forms in host cell under bacteria (intimin)

E.coli-d. Enterohemorrhagic (EHEC)

- **Hemorrhagic**
  - **bloody, copious diarrhea**
  - **few leukocytes**
  - **afebrile**
- **hemolytic-uremic syndrome**
  - **hemolytic anemia**
  - **thrombocytopenia (low platelets)**
  - **kidney failure**

Enterohemorrhagic *E. coli*

- **Usually O157:H7**



Transmission electron micrograph

Enterohemorrhagic *E. coli*

- Vero toxin
  - "shiga-like"
- Hemolysins
- younger than 5 years old,causing hemorrhagic colitis

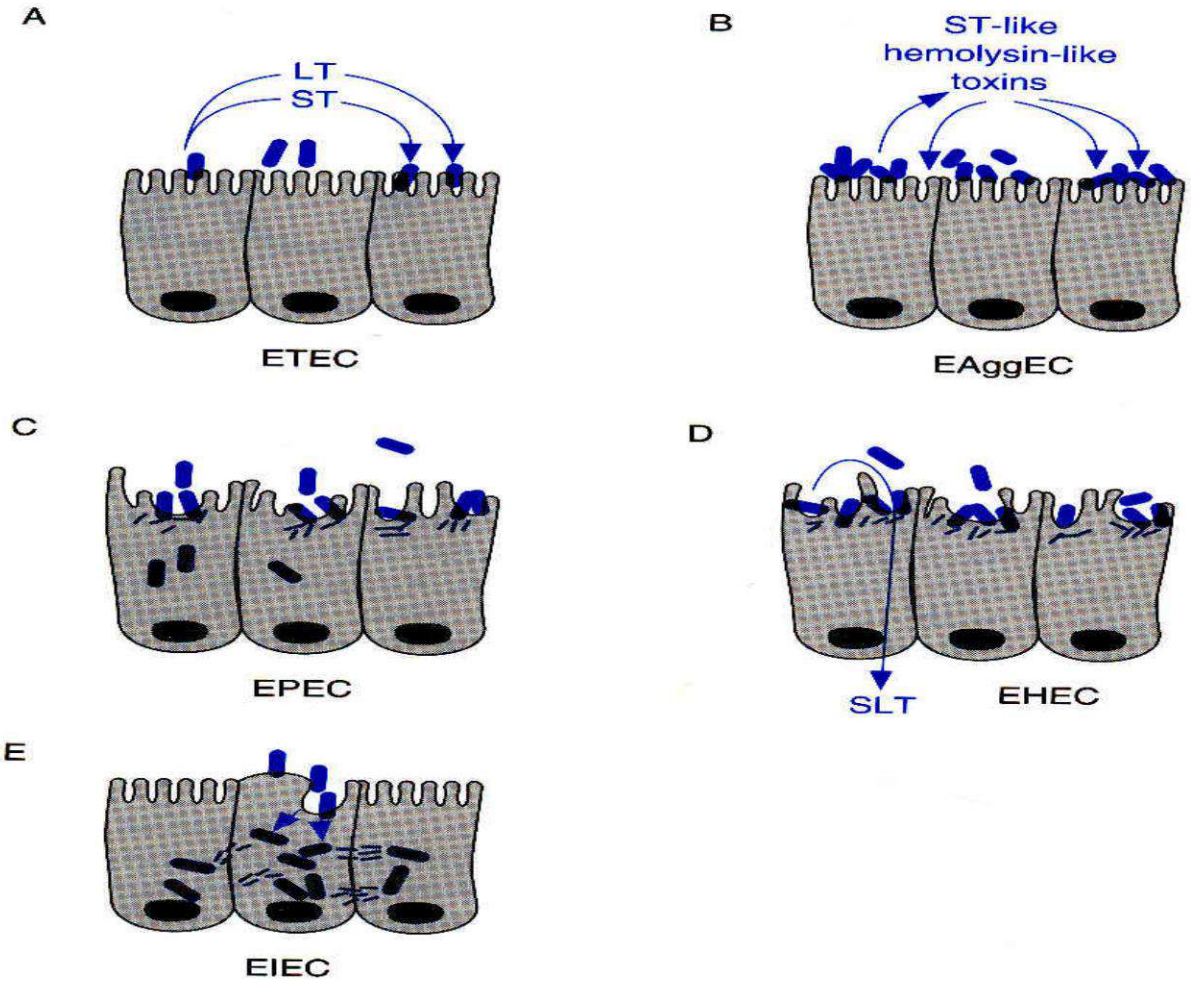
Enteroaggregative *E. coli*

- a cause of persistent, watery diarrhea with vomiting and dehydration in infants.
- That is autoagglutination in a ‘stacked brick’ arrangement.
- the bacteria adheres to the intestinal mucosa and elaborates enterotoxins (enteroaggregative heat-stable toxin, EAST).
- The result is mucosal damage, secretion of large amounts of mucus, and a secretory diarrhea.

E.coli-Enteroaggregative (EAggEC)

- **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 incluse watery diarrhea, vomiting, dehydration and occasional abdominal pain.**

Various Types of E. coli



	Virulence Factors			
Disease	**Adherence**	**Toxins**	**Siderophores**	**Invasion**
Enterotoxigenic *Escherichia coli* (ETEC)	Colonization factors of adherence (CFAs) Type 1 pili	Endotoxin Heat-labile enterotoxin (LT) Heat-stable enterotoxin (STa)	Enterochelin	Noninvasive
Enteropathogenic *E. coli* (EPEC)	Bundle-forming pili (BFP) Type 1 pili Intimin	Endotoxin	Enterochelin	Poorly invasive
Enteroaggregative *E. coli* (EAEC)	Mucus-associated autoagglutination Type 1 pili	Endotoxin Cytotoxin (enteroaggregative ST-like toxin [EAST])	Enterochelin	Noninvasive
Enteroinvasive *E. coli* (EIEC)	Type 1 pili Afimbrial adhesins	Endotoxin	Enterochelin	Type III secretion system Very invasive
Enterohemorrhagic *E. coli* (EHEC)	Type 1 pili Afimbrial adhesins	Shiga toxin Endotoxin	Enterochelin Heme uptake system	Probably poorly invasive

Sanitary significance

- **Totoal bacterial number: number of bacteria contained per ml or gm of the sample; the standard of drinking water is less than 100.**
- **Coliform bacteria index: the number of coliform bacteria detected out per 1000 ml sample; the standard of drinking water is less than 3**

General Characteristics of Salmonella

- **Coliform** bacilli (enteric rods)
- Motile gram-negative facultative anaerobes
- **Non-lactose fermenting**
- **Resistant to bile salts**
- H_2S producing

Classification and Taxonomy of Salmonella (Confused)

➢ **Old:** **Serotyping & biochemical assays used to name individual species within genus**
(e.g., *Salmonella enteritidis, S. choleraesuis, S. typhi*)

- **Over 2400 O-serotypes** (referred to as species)
(Kauffman-White antigenic schema)
- **Bioserotyping** (e.g., *S. typhimurium*)

➢ **New:** **DNA homology** shows only two species ***Salmonella enterica*** **(six subspecies)** and *S. bongori*

- Most pathogens in *S. enterica ssp. enterica*

Epidemiology of Salmonella Infection

DISEASE/BACTERIAL FACTORS

Enteritis, septicemia, enteric fever, asymptomatic carriage
Animals are main reservoir of human disease except for bacteria responsible for typhoid and paratyphoid fevers
Numerous virulence factors

TRANSMISSION

Ingestion of contaminated food products (especially poultry, eggs, dairy products)
Direct fecal-oral spread in children

WHO IS AT RISK?

Anyone consuming foods contaminated with large numbers of *Salmonella*, particularly children younger than 1 year old, elderly, patients with reduced gastric acids, and patients with AIDS
S. typhi: foreign travelers or individuals exposed to carriers

GEOGRAPHY/SEASON

Worldwide
More common in warm months

MODES OF CONTROL

Symptomatic treatment rather than antibiotics
Proper preparation and refrigeration of foods
Improved hygiene

Clinical Syndromes of Salmonella

Salmonellosis = Generic term for disease

Clinical Syndromes

- **Enteritis** (acute gastroenteritis)
- **Enteric fever** (prototype is **typhoid fever** and less severe paratyphoid fever)
- **Septicemia** (particularly *S. choleraesuis, S. typhi,* and *S. paratyphi*)
- **Asymptomatic carriage** (gall bladder is the reservoir for ***Salmonella typhi***)

Epidemiology and Clinical Syndromes of Salmonella (cont.)

Enteritis

- **Most common** form of salmonellosis with major **foodborne outbreaks** and **sporadic disease**
- **High infectious dose** (10^8 CFU)
- **Poultry, eggs**, etc. are sources of infection
- **6-48h incubation period**
- Nausea, vomiting, nonbloody diarrhea, fever, cramps, myalgia and headache common
- ***S. enteritidis*** bioserotypes (e.g., *S. typhimurium*)

Pathogenesis of Salmonella

Enteritis (cont.)

Virulence attributable to:

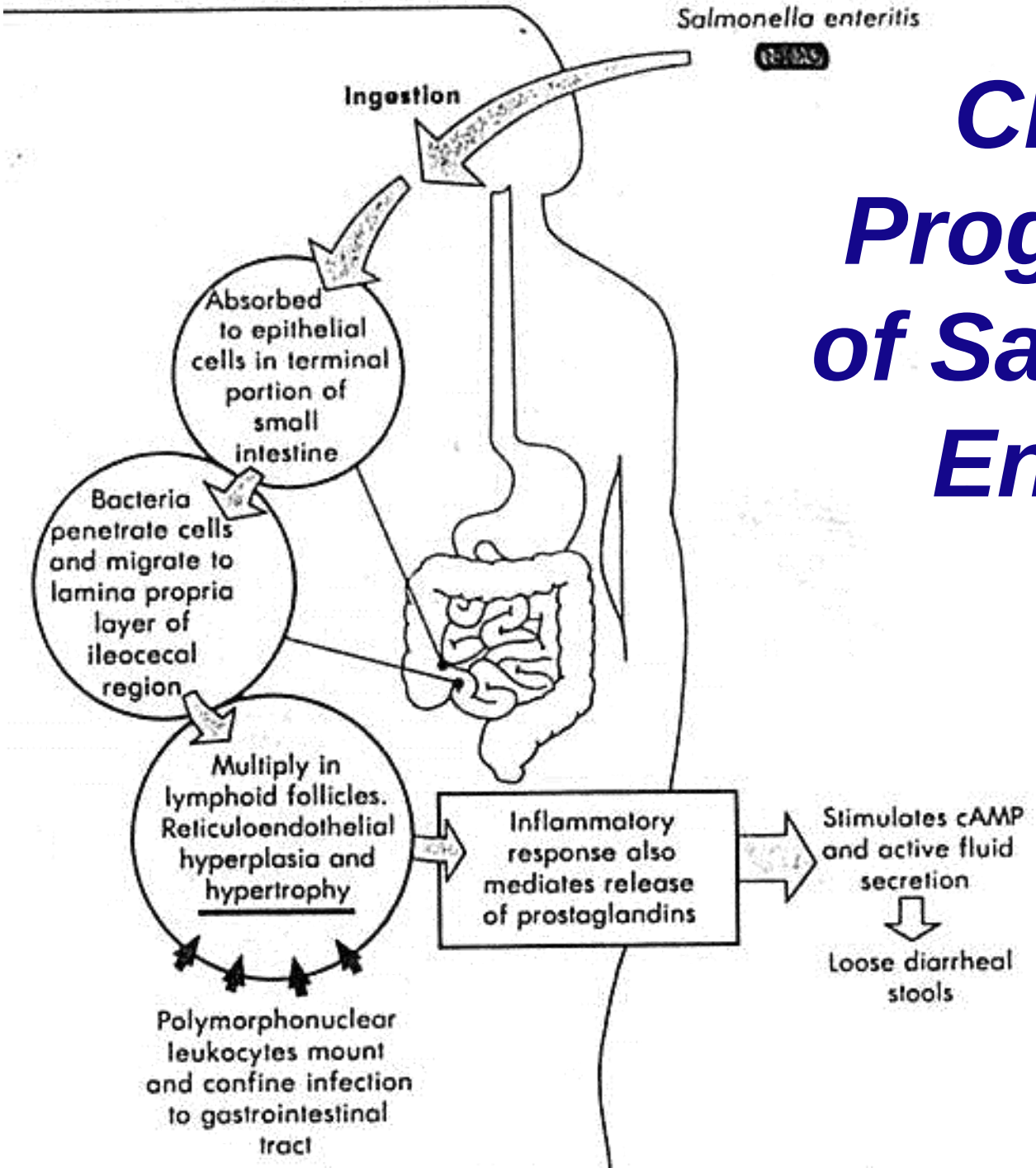
- Invasiveness
- Intracellular survival & multiplication
- Endotoxin
- Exotoxins: Effects in host have not been identified
  - Several *Salmonella* serotypes produce enterotoxins similar to both the heat-labile (LT) and heat-stable enterotoxins (ST), but their effect has not been identified
  - A distinct cytotoxin is also produced and may be involved in invasion and cell destruction

Pathogenesis of Salmonella (cont.)

Invasiveness in Enteritis (cont.)

- Penetrate mucus, **adhere to and invade into epithelial layer** (enterocytes) **of terminal small intestine** and further into subepithelial tissue
- Bacterial cells are **internalized in endocytic vacuoles** (intracellular) and the organisms **multiply**
- PMN's confine infection to gastrointestinal (GI) tract, but organisms **may spread hematogenously** (through blood, i.e., septicemia) to other body sites
- **Inflammatory response** mediates release of prostaglandins, **stimulating cAMP and active fluid secretion** with loose **diarrheal stools**; **epithelial destruction occurs during late stage of disease**

Clinical Progression of Salmonella Enteritis



Lamina propria = thin membrane between epithelium & basement layer

Hyperplasia = abnormal increase in # of normal cells

Hypertrophy = abnormal increase in normal tissue/organ size

Prostaglandins = potent mediators of diverse set of physiologic processes

Epidemiology & Clinical Syndromes (cont.)

Enteric Fevers

- ***S. typhi*** causes **typhoid fever**
  S. paratyphi A*, *B (*S. schottmuelleri*) and ***C*** (*S. hirschfeldii*) cause milder form of enteric fever
- **Infectious dose** = 10^6 CFU
- **Fecal-oral route of transmission**
  - Person-to-person spread by chronic carrier
  - Fecally-contaminated food or water
- **10-14 day incubation period**
- Initially signs of **sepsis/bacteremia** with **sustained fever** (delirium) for ≥ one week before abdominal pain and **gastrointestinal symptoms**

Pathogenesis of Salmonella (cont.)

Enteric Fevers (cont.)

Virulence attributable to:

- Invasiveness
  - Pass through intestinal epithelial cells in ileocecal region, infect the regional lymphatic system, invade the bloodstream, and infect other parts of the reticuloendothelial system
  - Organisms are phagocytosed by macrophages and monocytes, but survive, multiply and are transported to the liver, spleen, and bone marrow where they continue to replicate
  - Second week: organisms reenter bloodstream and cause prolonged bacteremia; biliary tree and other organs are infected; gradually increasing sustained fever likely from endotoxemia
  - Second to third week: bacteria colonize gallbladder, reinfect intestinal tract with diarrheal symptoms and possible necrosis of the Peyer's patches

Microbial Defenses Against Host Immunological Clearance

ENCAPSULATION and
ANTIGENIC MIMICRY, **MASKING** or **SHIFT**

CAPSULE, GLYCOCALYX or **SLIME LAYER**

Polysachharide capsules *Streptococcus pneumoniae, Neisseria meningitidis, Haemophilus influenzae*, etc.

Polypeptide capsule of *Bacillus anthracis*

→ **EVASION** or **INCAPACITATION** of **PHAGOCYTOSIS** and/or **IMMUNE CLEARANCE**

PHAGOCYTOSIS INHIBITORS: mechanisms enabling an invading microorganism to resist being engulfed, ingested, and or lysed by phagocytes/ phagolysosomes

→ **RESISTANCE** to **HUMORAL FACTORS**

RESISTANCE to **CELLULAR FACTORS**

See Chpt. 19

Methods That Circumvent Phagocytic Killing

METHOD	EXAMPLE
Inhibition of phagolysosome infusion	*Legionella* species, *Mycobacterium tuberculosis*, *Chlamydia* species
Resistance to lysosomal enzymes	*Salmonella typhimurium*, *Coxiella* species, *Ehrlichia* species, *Mycobacterium leprae*, *Leishmania* species, ***Salmonella typhi***
Adaptation to cytoplasmic replication	*Listeria* species, *Francisella* species, *Rickettsia* species

See Chpt. 19

Epidemiology & Clinical Syndromes (cont.)

Septicemia

- Can be caused by all species, but **more commonly associated with** *S. choleraesuis, S. paratyphi, S. typhi,* and *S. dublin*
- **Old, young and immunocompromised** (e.g., AIDS patients) at increased risk

Epidemiology & Clinical Syndromes (cont.)

Asymptomatic Carriage

- **Chronic carriage in 1-5% of cases** following *S. typhi* or *S. paratyphi* infection
- **Gall bladder** usually the **reservoir**
- Chronic carriage with other *Salmonella* spp. occurs in <<1% of cases and does not play a role in human disease transmission

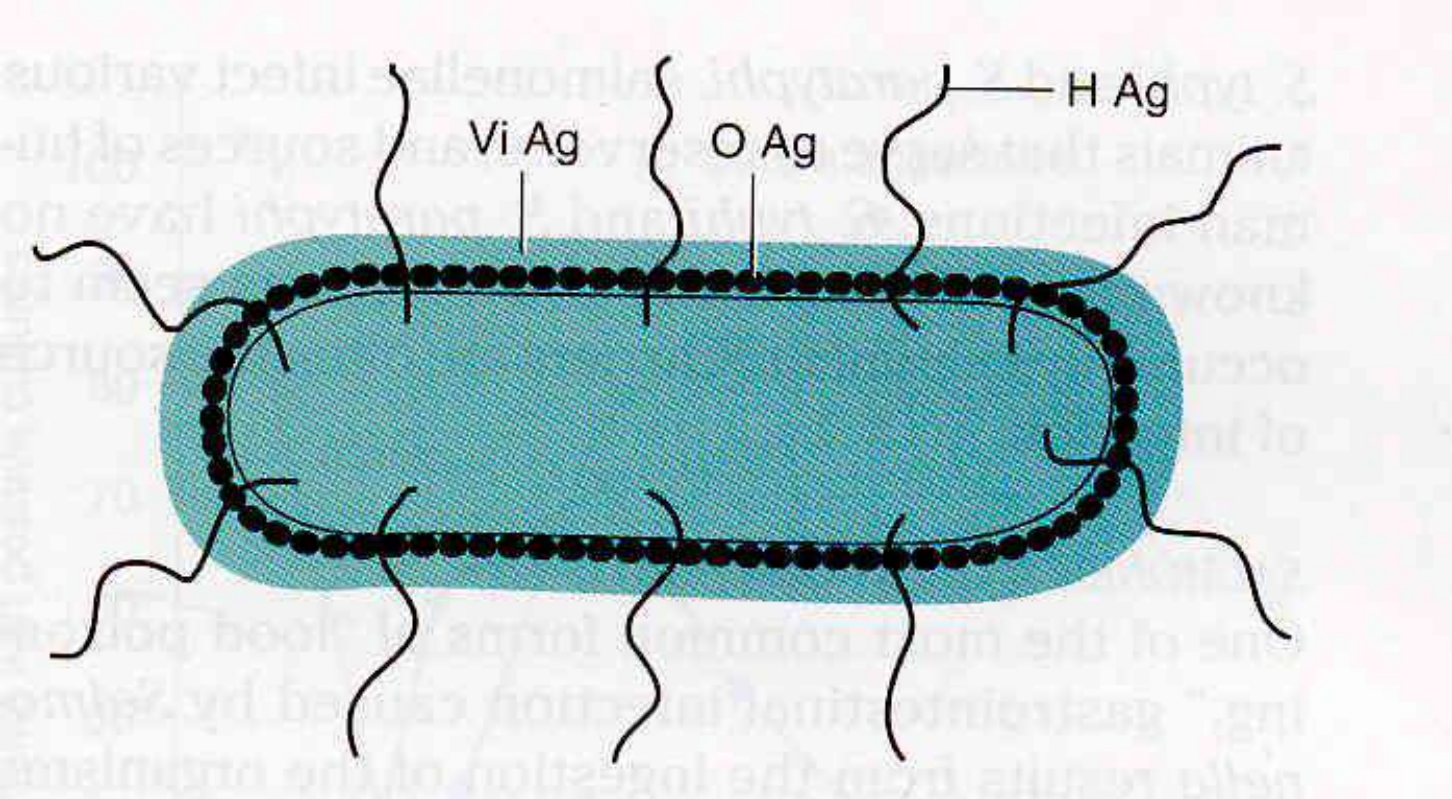
Treatment, Prevention and Control of Salmonella Infections

Enteritis:

- **Antibiotics not recommended** for enteritis because prolong duration
- Control by **proper preparation of poultry & eggs**

Enteric fever:

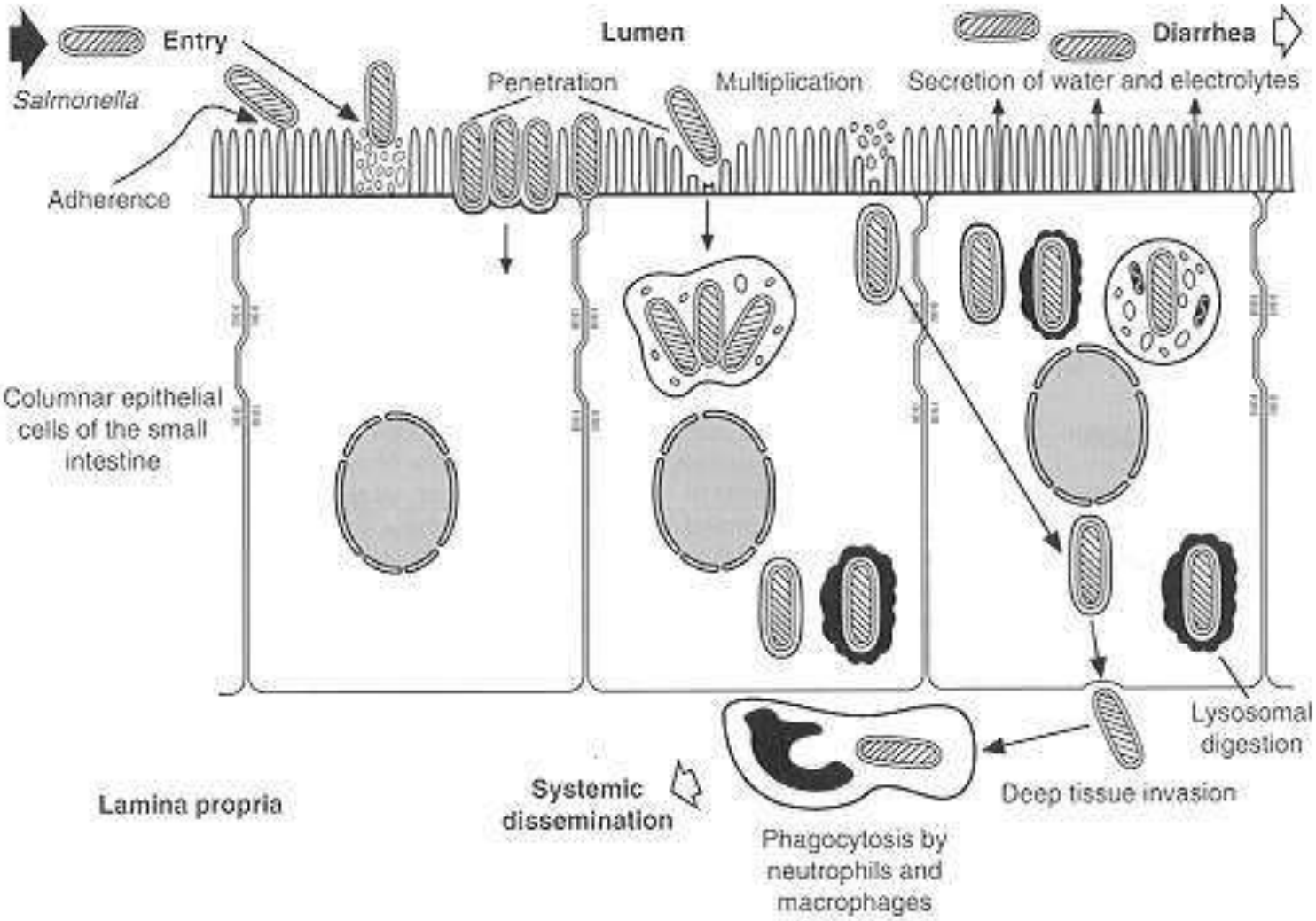
- **Antibiotics to avoid carrier state**
- **Identify & treat carriers** of *S. typhi & S. paratyphi*
- **Vaccination** can reduce risk of disease for **travellers in endemic areas**



The antigenic structures of salmonellae used in serologic typing

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 – 2 different systems may be found:
  - 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
- Outer membrane proteins - involved in the ability of *Salmonella* to survive inside macrophages
- Flagella – help bacteria to move through intestinal mucous
- Enterotoxin - may be involved in gastroenteritis
- Iron capturing ability

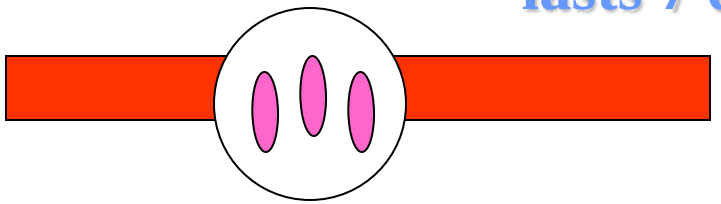

Entry
Salmonella
Lumen
Diarrhea
Penetration
Multiplication
Secretion of water and electrolytes
Adherence
Columnar epithelial cells of the small intestine
Lysosomal digestion
Lamina propria
Systemic dissemination
Deep tissue invasion
Phagocytosis by neutrophils and macrophages

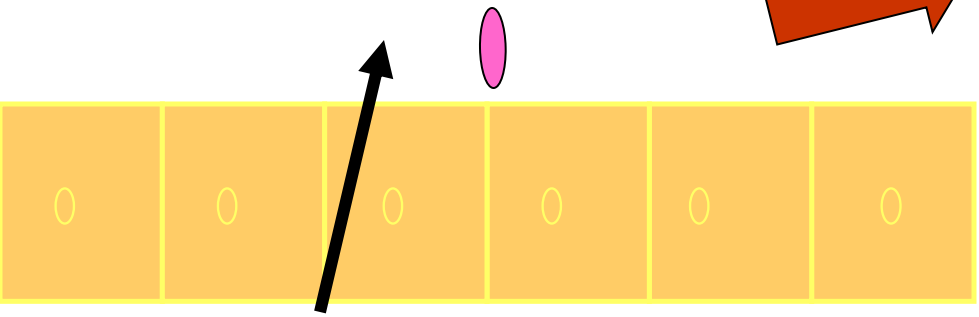
Enteric or typhoid fever

- Enteric or typhoid fever occurs when the bacteria leave the intestine and multiply within cells of the reticuloendothelial system.
- The bacteria then re-enter the intestine, causing gastrointestinal symptoms.
- Typhoid fever has a 10-14 day incubation period and may last for several weeks.
- Salmonella typhi is the most common species isolated from this salmonellosis.
- Human reservoir:carrier state common
- Contaminated food:water supply
- Poor sanitary conditions

Typhoid

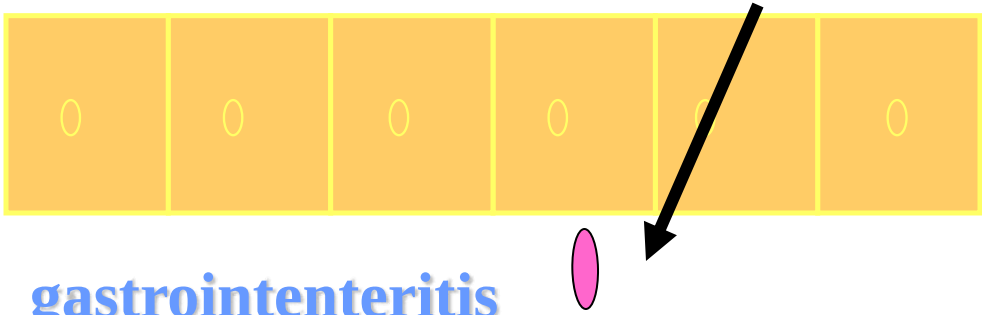
Septicemia
-occurs 10-14 days
– lasts 7 days





•acute phase, gastroenteritis

gall bladder
–shedding, weeks



gastrointenteritis

Salmonella gastroenteritis

- Salmonella gastroenteritis is the most common form of salmonellosis and generally requires an 8-48 hour incubation period and may last from 2-5 days.
- Symptoms include nausea, vomiting and diarrhea (non-bloody stool). Salmonella enteritidis is the most common isolate.
- poultry家禽, eggs. no human reservoir
- self-limiting (2 - 5 days)

Salmonella septicemia

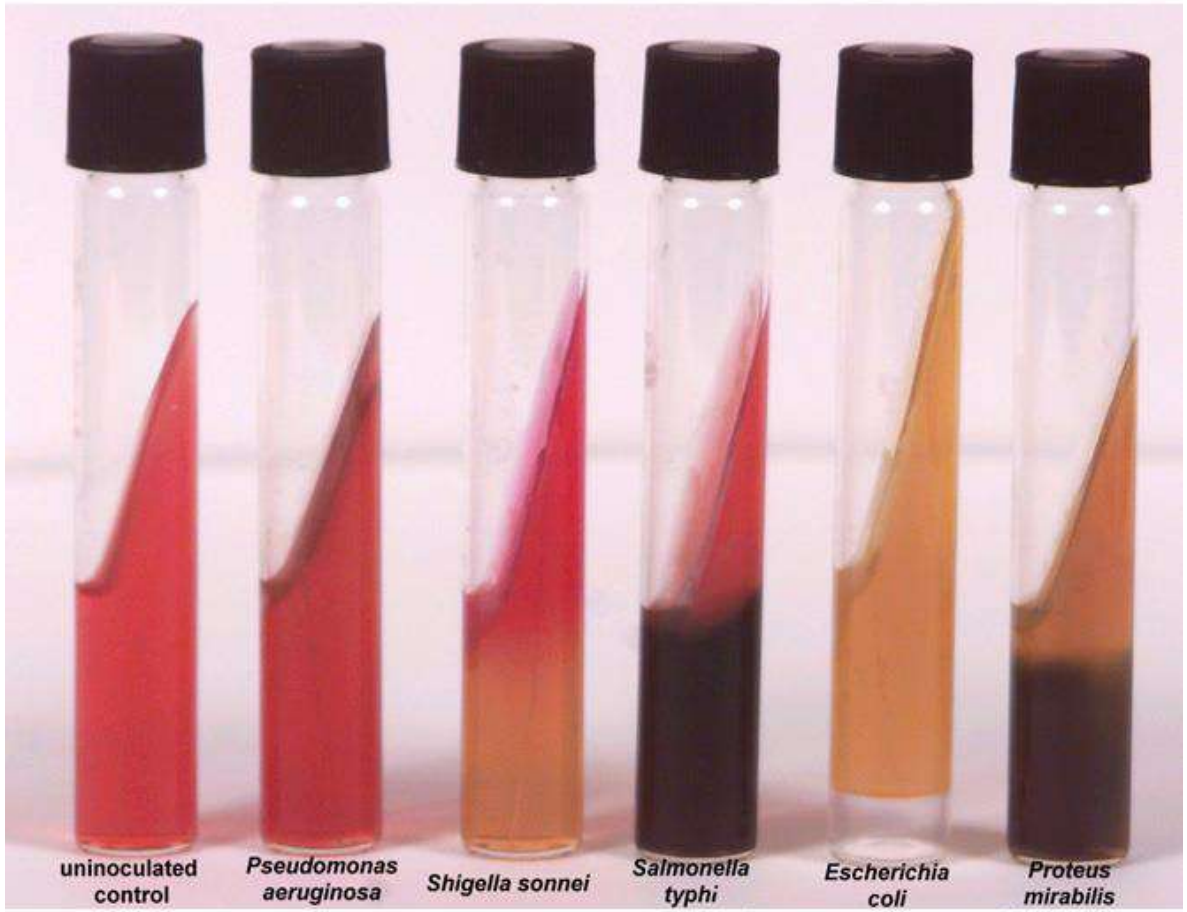
- ***Salmonella* septicemia (bacteremia) may be caused by any species but *S. cholerae-suis* is common. This disease resembles other Gram-negative septicemias and is characterized by a high, remittent fever with little gastrointestinal involvement.**

Immunity (S. typhi)

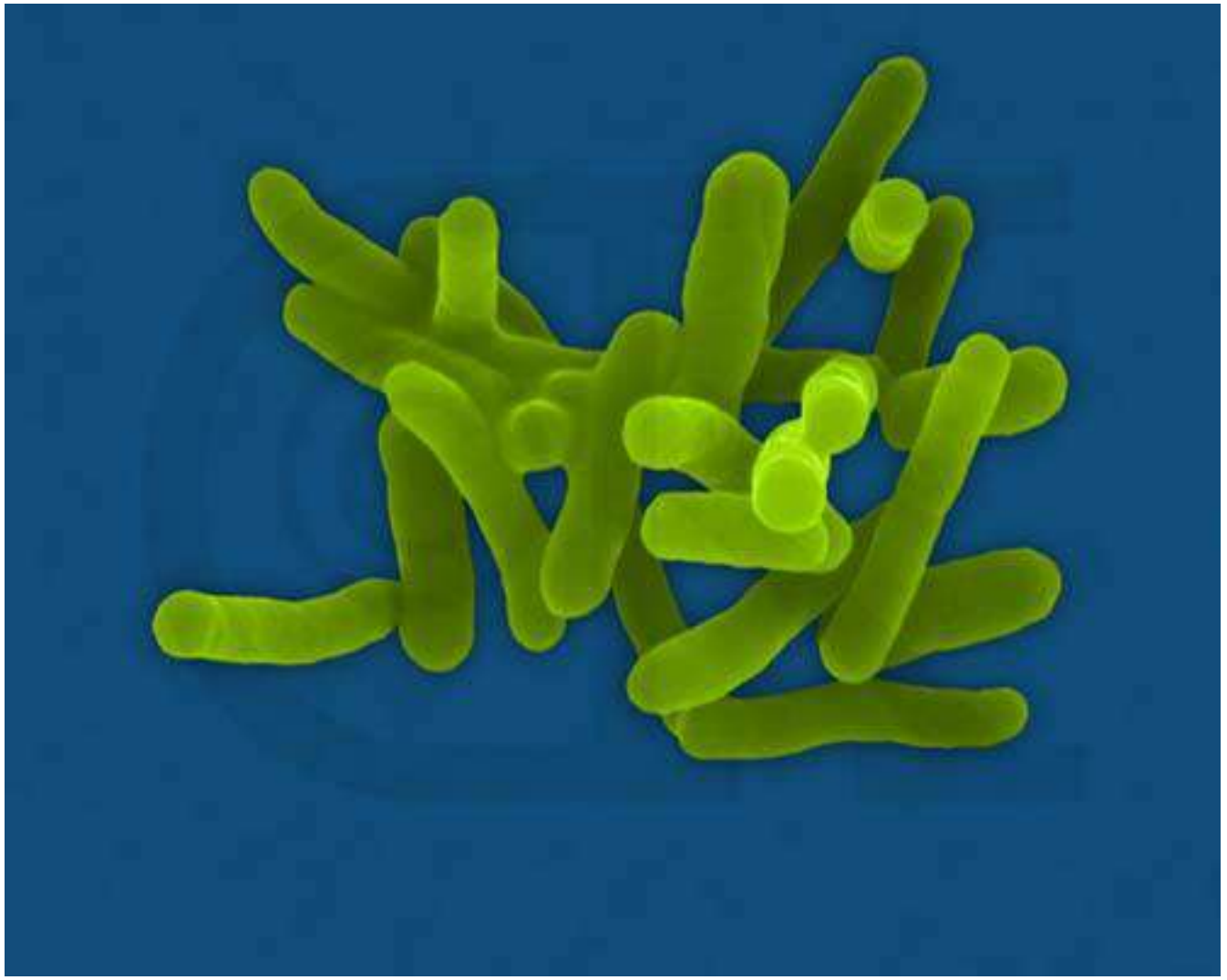
- **Vi (capsular) antigen**
  - **protective**

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**


uninoculated control
Pseudomonas aeruginosa
Shigella sonnei
Salmonella typhi
Escherichia coli
Proteus mirabilis
ASM MicrobeLibrary.org©Chamberlain

Shigella



Shigella

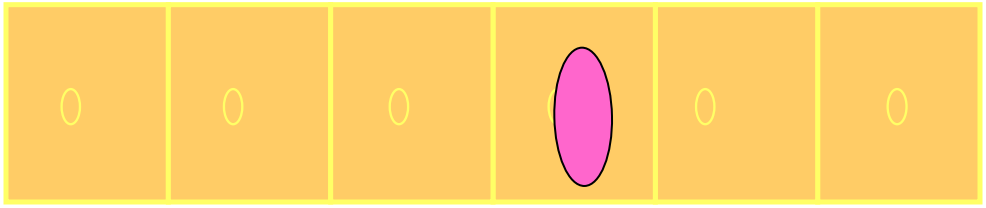
- ***S. flexneri, S. boydii, S. sonnei, S. dysenteriae***
  - **bacillary dysentery**
  - **shigellosis**
    - **bloody feces**
    - **intestinal pain**
    - **pus**

Genral features

- Pili.
- Most strains can not ferment lactose; S. sonnei can slowly_ ferment lactose.
- According to O antigen, 4 groups
- Easily causing drug-resistence.

Shigellosis

- **within 2-3 days**
  - **–epithelial cell damage**

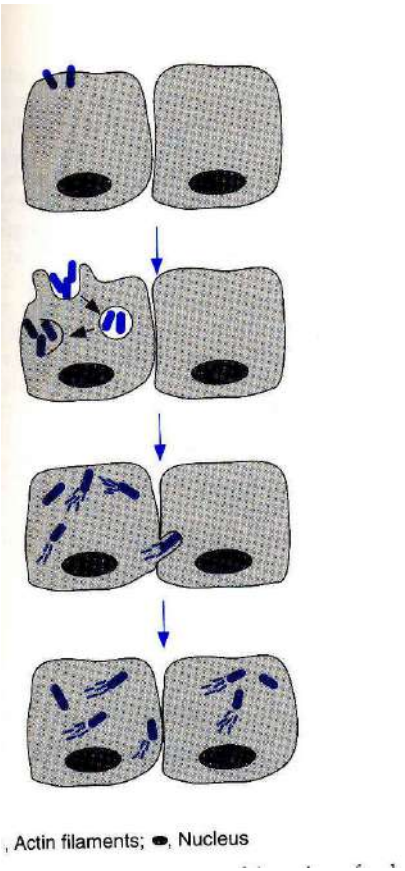


Shiga toxin

- enterotoxic
- cytotoxic
- inhibits protein synthesis
  - lysing 28S rRNA

Shigella attachment and penetration

- **Within 2-3 days**
- **Epithelial cell damage**



Clinical significance

- man only "reservoir"
- mostly young children
  - fecal to oral contact
  - children to adults
- transmitted by adult food handlers
  - unwashed hands

Clinical significance

- **The infective dose required to cause infection is very low (10-200 organisms).**
- **There is an incubation of 1-7 days followed by fever, cramping, abdominal pain, and watery diarrhea (due to the toxin)for 1-3 days.**
- **This may be followed by frequent, scant stools with blood, mucous, and pus (due to invasion of intestinal mucosa).**
- **Is is rare for the organism to disseminate.**
- **The severity of the disease depends upon the species one is infected with. *S. dysenteria* is the most pathogenic followed by *S. flexneri*, *S. sonnei* and *S. boydii*.**

Immunity

- **SIgA.**

Diagnosis of Shigella infection

- **Specimen: stool.**
- **Culture and Identification**
- **Quick immunological methods:**

1. **Immunofluorescent “ball” test;**
2. **Coagglutination**.

Prevention

- **streptomycin dependent (SD) dysentery vaccine.**

Treating shigellosis

- **manage dehydration**
- **patients respond to antibiotics ,** Problem of drug-resistance
  - **disease duration diminished**

Klebsiella

- NF of GI tract, but potential pathogen in other areas
- Virulence factors
  - Capsule
  - Adhesions
  - Iron capturing ability
- **Clinical significance**
  - **Causes pneumonia, mostly in immunocompromised hosts. Permanent lung damage is a frequent occurrence (rare in other types of bacterial pneumonia)**
  - **A major cause of nosocomial infections such as septicemia and meningitis**

Klebsiella

- **K. pneumoniae (Friedlander bacilli): may cause primary pneumonia, urinary tract and wound infections, bacteremia, meningitis, etc.**
- **K. rhinoscleromatis: pathogen of granumatous destruction of nose and pharynx.**
- **K. ozaenae: causes chronic atrophic rhinitis.**

Proteus

- **General characteristics: “swarming” phenomenon on nonselective agar (P.vulgaris; P.mirabilis and P.myxofaciens)**
- **Antigenic structure: O antigen (group-specific); H antigen (type-specific); certain P.vulgaris strains (OX-19, OX-K, OX-2)have common antigen with Rickettsia (Weil-Felix test).**
- **Clinical manifestations: urinary tract infections; food poisoning.**