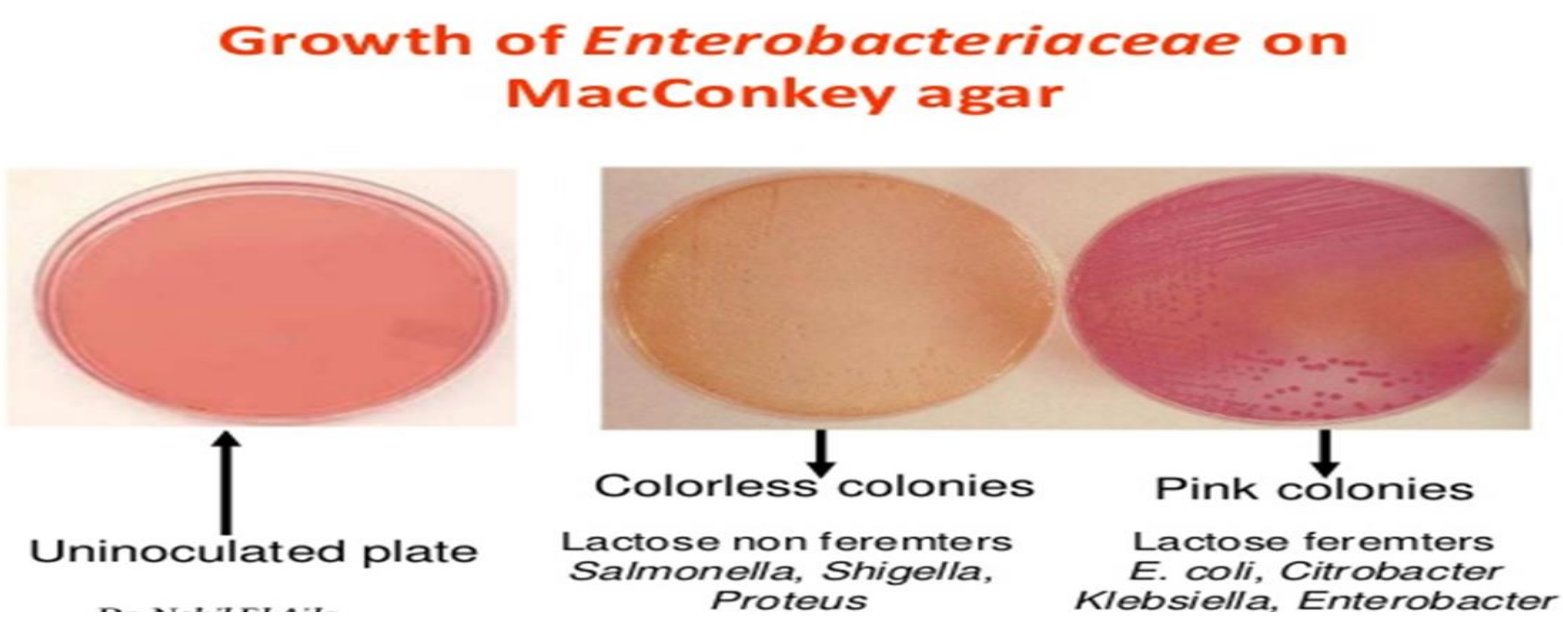
Enterobacteriaceae

Asst . Prof. Dr. Bara′ Hamid



Family *Enterobacteriaceae* often referred to as “enterics”:

- Gram negative bacilli or coccobacilli
- Non-spore forming
- 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.

Four major features:

1- All ferment glucose (dextrose).

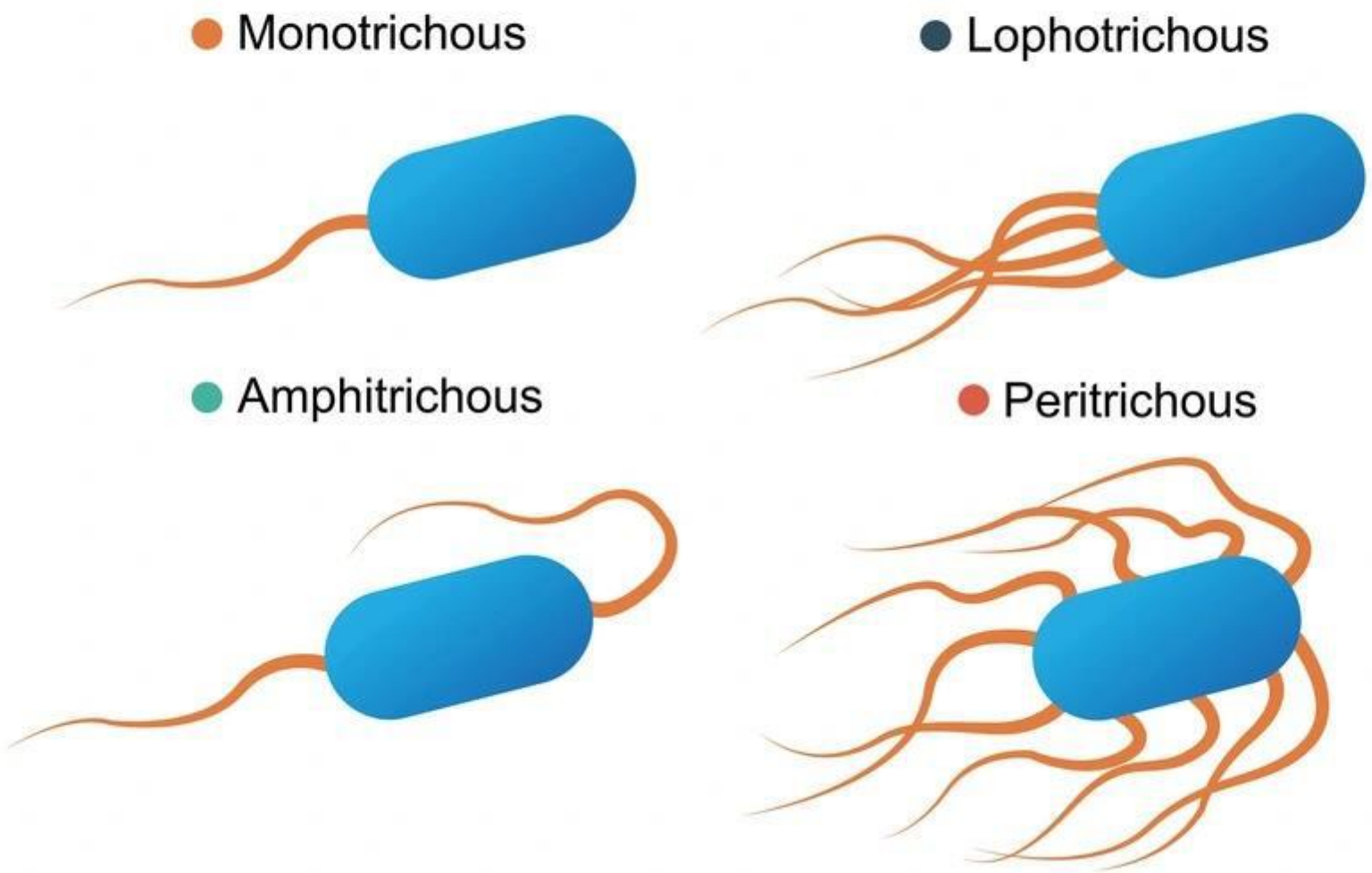
2- All reduce nitrates to nitrites (NO_3 to NO_2 or all the way to N_2).

3- All are oxidase negative.

4- All except Klebsiella, Shigella and Yersinia are motile by peritrichous flagella.

MICROBIOLOGY

Bacterial Flagella



Representative genera of Enterobacteriaceae that are Medically Important:

E. coli (Escherichia)

Klebsiella spp.

Enterobacter

Morganella spp.

Proteus spp.

Salmonella spp.

Shigella spp.

Citrobacter

Serratia spp.

Yersinia spp

Culture methods

- **blood agar/** Colony morphology on BAP of little value, as they look the same, except for *Klebsiella*.
- **selective/** differential medium (MacConkey).

Enterics classified as following:

1) Based on lactose fermentation(On MacConkey), enterics are divided into two groups:

A) Lactose fermenters = coliform (*E. coli, Klebsiella, Enterobacter)*

B) Non- lactose fomenters = other enteric (*Morganella*, *Proteus*, *Salmonella, Shigella* , *Citrobacter, Serratia* , *Yersinia*).

2) Based on clinical infections produced, enterics are divided into two categories:

A- Primary intestinal pathogens: *Salmonella*, *Shigella*, and *Yersinia* spp.

B- Opportunistic pathogens: normally part of the usual intestinal flora that may produce infection outside the intestine. Like normal flora of intestine/ *E. coli* infectious in urinary tract KES (*Klebsiell*a – *Enterobacter* – *Serratia*) *Proteus* – *Providencia* - *Citrobacter*.

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**:**

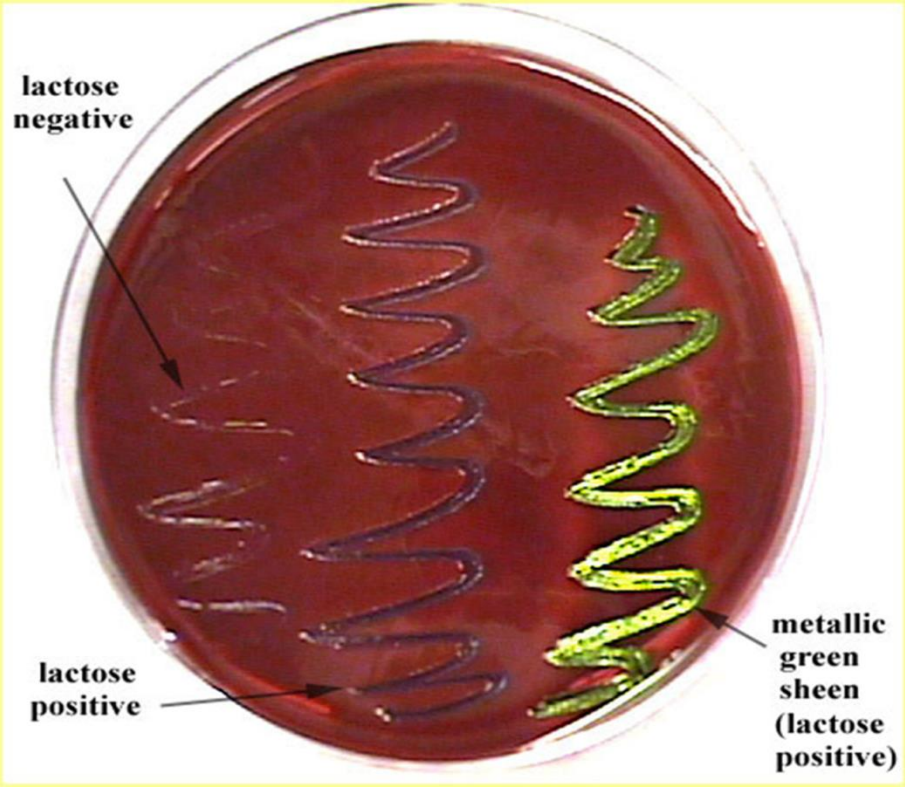
- O antigen – somatic, heat-stable antigen located in the cell wall.
- H antigen – flagellar, heat labile antigen.
-K antigen – capsular, heat-labile antigen.

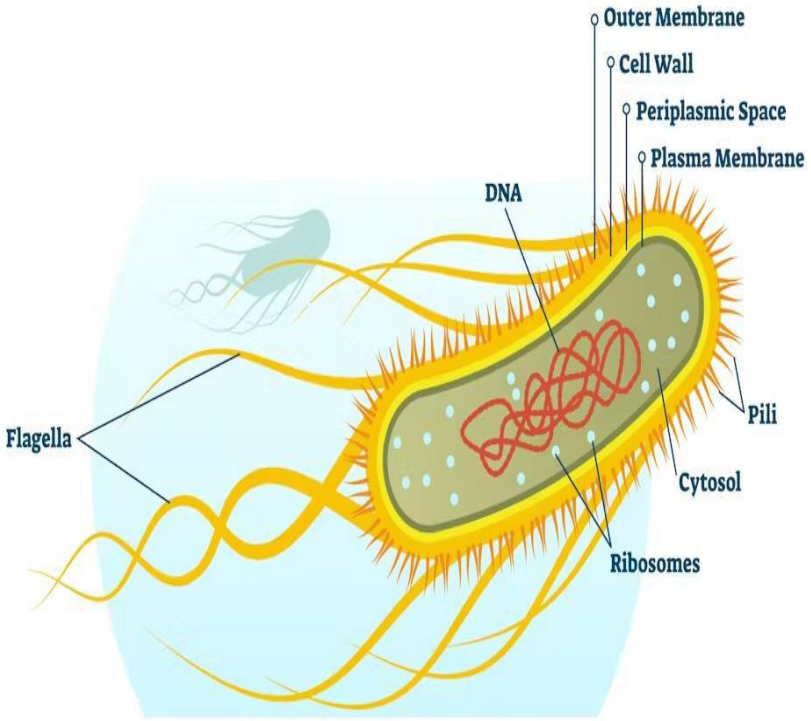
Summary of morphology, cultural characteristics, and biochemical reactions of *Enterobacteriaceae*

	TSI	Indole	MR	VP	Citrate	Urease	Motility
E. coli	A/A/-	+ve	+ve	-ve	-ve	-ve	Motile
Citrobacter freundii	A/A/-	+ve	+ve	-ve	+ve	-ve	Motile
Klebsiella pneumoniae	A/A/-	-ve	-ve	+ve	+ve	+ve	Non motile
Enterobacter cloacae	A/A/-	-ve	-ve	+ve	+ve	+ve	Motile
Salmonella typhi	A/Alk/+	-ve	+ve	-ve	+ve	-ve	Motile
Shigella boydii	A/Alk/-	-ve	+ve	-ve	-ve	-ve	Non motile
Proteus mirabilis	A/Alk/+	-ve	+ve	-ve	+ve	+ve	Motile Swarwing

Escherichia coli

- It is the most significant species in the genus. Important potential pathogen
in humans. Common isolate from colon flora, usually motile.
- Dry, pink (lactose positive) colony with surrounding pink area on MacConkey.





• Infections

- Bacteremia (most commonly isolated Gram-negative rod) , primarily from a genitourinary tract infection or a gastrointestinal source.

- Urinary tract infection (most common cause of bacterial UTIs); limited to bladder (cystitis) or can spread to kidneys (pyelonephritis) or prostate (prostatitis) .

- Neonatal meningitis (usually with strains carrying the K1 capsular antigen). *E. coli* is one of the most common causes of septicemia and meningitis among neonates; acquired from mother in the birth canal before or during delivery.

- Intraabdominal infections (associated with intestinal perforation). Most infections are endogenous(from patients own normal flora).

- Gastroenteritis At least five different pathogenic groups cause gastroenteritis (EPEC, ETEC, EHEC, EIEC, EAEC); most cause diseases in developing countries, although EHEC is an important cause of hemorrhagic colitis (HC) and hemolytic uremic syndrome (HUS).

Gastrointestinal Infections

Enteropathogenic (EPEC) – primarily in infants and children; outbreaks in hospital nurseries and day care centers; stool has mucous but not blood.

Enteroaggregative (EAEC) – cause diarrhea by adhering to the mucosal surface of the intestine; watery diarrhea; symptoms may persist for over two weeks.

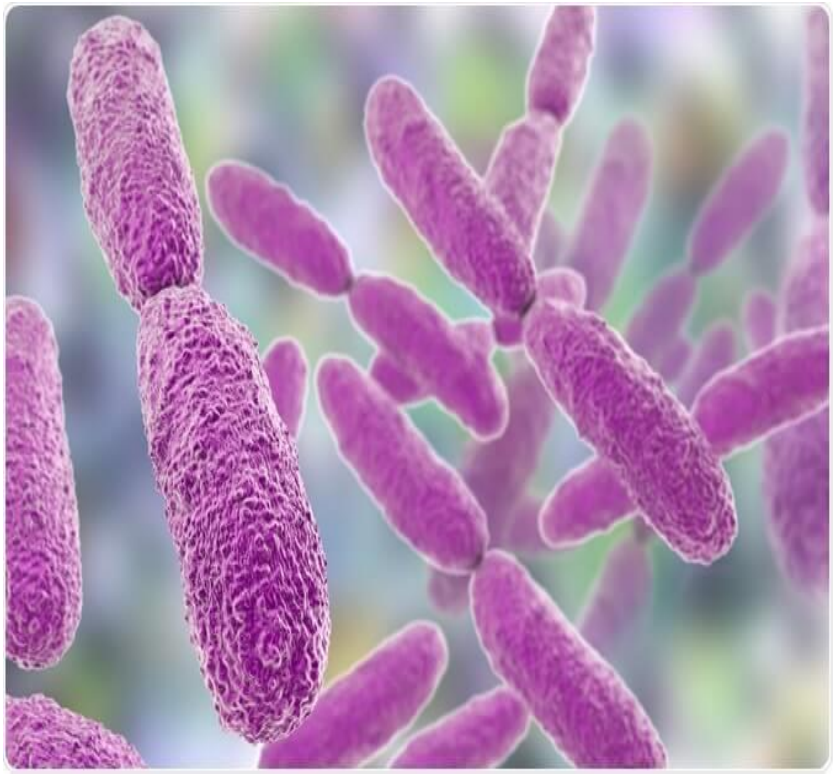
Enterotoxigenic (ETEC) – "traveler's diarrhea"; watery diarrhea without blood; self-limiting; usually not identified.

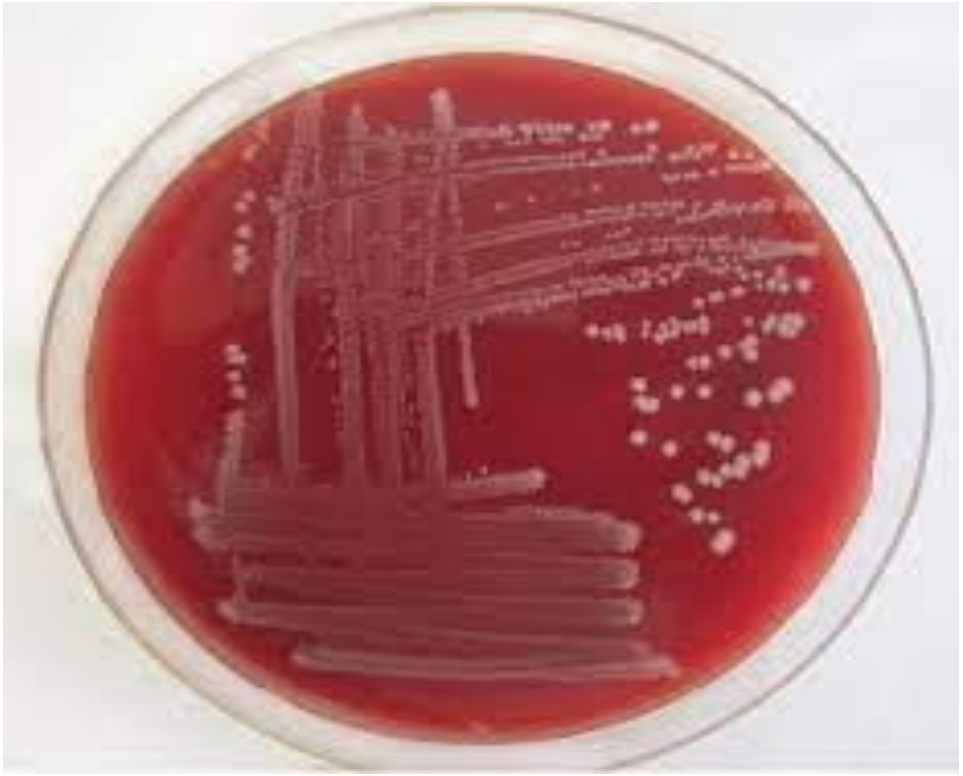
Enteroinvasive (EIEC) – produce dysentery with bowel penetration, invasion and destruction of intestinal mucosa; watery diarrhea with blood.

Enterohemorrhagic (EHEC serotype 0157:H7) – associated with hemorrhagic diarrhea and hemolytic-uremic syndrome (HUS), which includes low platelet count, hemolytic anemia, and kidney failure; potentially fatal, especially in young children; undercooked hamburger and unpasteurized milk have spread the infection.

Klebsiella species

Usually found in GI tract. Four major species, *K. pneumoniae* is mostly commonly isolated species. Possesses a polysaccharide capsule, which protects against phagocytosis and antibiotics and makes the colonies moist and mucoid. Has a distinctive “yeasty” odor. Frequent cause of nosocomial pneumonia.



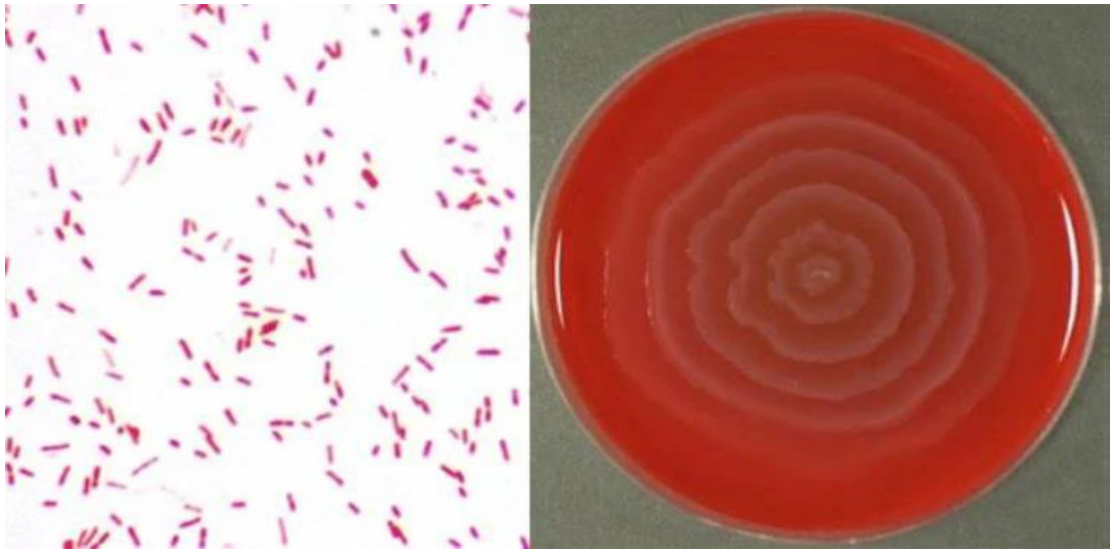


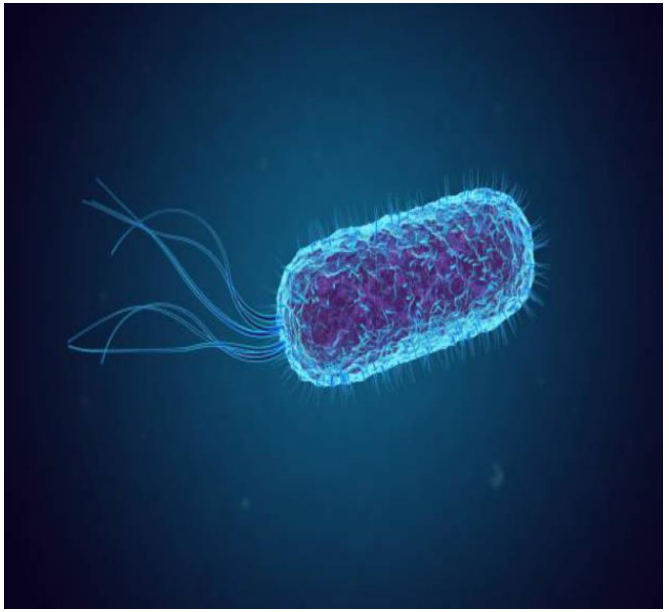
Proteus

All are:

- Normal intestinal flora
- Opportunistic pathogens
- Lactose negative

P. mirabilis and *P. vulgaris* are widely recognized human pathogens, isolated from urine, wounds, and ear and bacteremic infections. Both produce swarming colonies on non-selective media and have a distinctive "burned chocolate" odor. Both are strongly urease positive and exhibits characteristic "swarming"





Salmonella

S. typhi and *S. paratyphi* are the causes of human Typhi & paratyphi respectively. They are strict human pathogens . Infections occur worldwide, particularly in the warm months of the year.

Direct fecal-oral spread in children. Most infections are acquired by eating contaminated food products. Individuals at risk for infection include those who eat improperly cooked poultry or eggs, patients with reduced gastric acid levels, and immunocompromised patients.

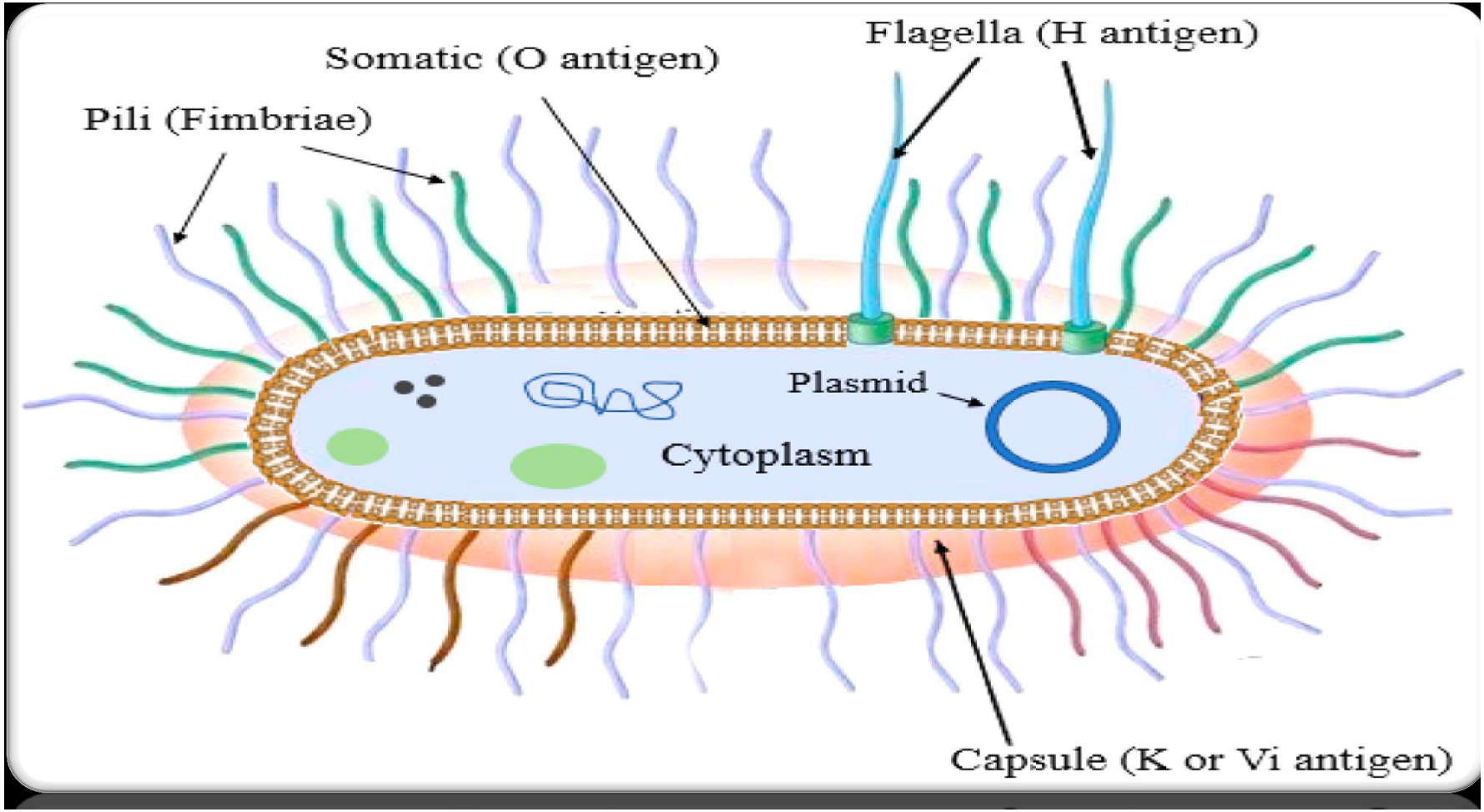
***Salmonella* possess:**

-Vi antigen positive(Capsular antigen of *Salmonella typhi* referred to as Vi antigen).

- Endotoxin

-Tolerant to acids in phagocytic vesicles. Can survive in macrophages and spread from the intestine to other body sites (particularly true of *S. typhi*).

Salmonella


Flagella (H antigen)
Somatic (O antigen)
Pili (Fimbriae)
Plasmid
Cytoplasm
Capsule (K or Vi antigen)

Clinical Manifestations

1-Gastroenteritis

2- Enteric fever , also called typhoid fever [*S. typhi*] or paratyphoid fever caused by [*S. paratyphi*]. Incubation 5-21 days.
Fever, relative bradycardia, leukopenia, anemia, constipation, rose spots, Neuropsychiatric manifestations. Mortality rate was 10-15% in preantibiotic era.

3-Bacteremia and vascular infections: (most commonly seen with *S. typhi, S. Paratyphi).*

4-Carrier state = Asymptomatic colonization (primarily with *S. typhi* and *S. paratyphi*).

Diagnosis of typhoid fever:

-Specimens: Stool, urine, blood

A- Culture/ on Selective media (MacConkey, SS agar)

►Typhoid fever / 1st.week blood culture

► Stool culture/ ≥3. Week

Blood cultures are positive during the first week and after the second week, while stool cultures and sometimes urine cultures are positive after the second week.

B- Widal test:

Anti-O Ab

Anti-H Ab

Anti-Vi Ab (in long term carriers)

The Widal test is a serological test for antibodies against *Salmonella typhi* & *S. paratyphi*. One looks for a 4-fold rise in titer between acute and convalescent stages.

10% of those infected become short term carriers and a smaller % become long-term carriers due to persistence of the bacteria in the gallbladder or urinary bladder.

When facilities for culturing are not available, the Widal test is the reliable and can be of value in the diagnosis of typhoid fevers in endemic areas.
Widal reaction is specific reaction consisting in agglutination of typhoid bacilli when mixed with serum from a patient having typhoid fever or other salmonella infection and constituting a test for the disease.
The antigens used in the test are H and O antigens of *Salmonella typhi* & *S. paratyphi*. The patient's serum is tested for O and H antibodies (agglutinins) against *Salmonella* antigens. The main principle of widal test is that if homologous antibody is present in patients serum, it will react with antigen in the reagent and gives visible agglutination in the tube. The titre of the patient serum using Widal test antigen suspensions is the highest dilution of the serum sample that gives a positive result. Agglutinin starts appearing in serum by the end of 1st week with sharp rise in 2nd and 3rd week and the titre remains steady till 4th week, after which it declines.

Shigella:

4 medically important species which are*:*

Shigella dysenteriae
Shigella flexneri
Shigella boydii
Shigella sonnei

Nonmotile, noncapsulated, H2S negative.

- *S. sonnei* is the most common cause of shigellosis in the industrial world.
- *S. flexneri* is the most common cause in developing countries.

Virulence factors & pathogenicity :

- Endotoxin and genes for adherence, invasion, and intracellular replication, permeability barrier of outer membrane.

Exotoxin (Shiga toxin) is produced by *S. dysenteriae;* disrupts protein synthesis and produces endothelial damage.

- Hemolytic colitis (HC) and hemolytic uremic syndrome (HUS) associated with *Shigella*.
- Relatively few organisms can produce disease (highly infectious).

Disease occurs worldwide with no seasonal incidence.

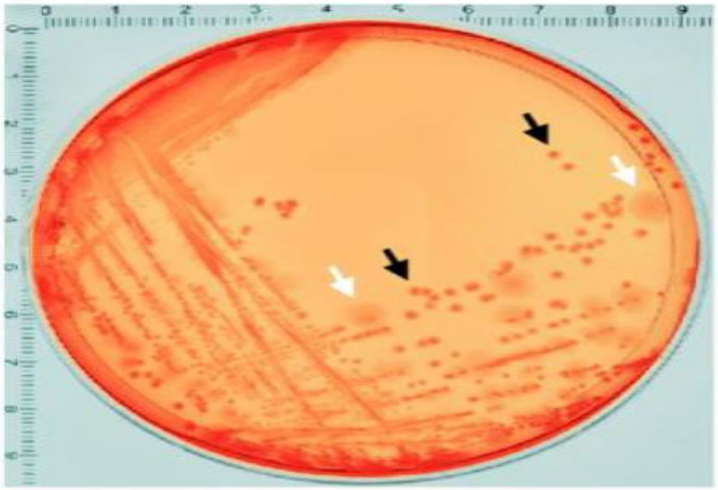
Humans are only reservoir for these bacteria. Disease spread person to person by fecal-oral route.

Gastroenteritis (shigellosis) symptoms:

Most common form is an initial watery diarrhea progressing within 1 to 2 days to abdominal cramps and tenesmus (with or without bloody stools).

Asymptomatic carriage develops in a small number of patients (reservoir for future infections). A severe form of disease is caused by *S. dysenteriae* (bacterial dysentery) as bloody diarrhea containing mucus, were seen.

(A)



(B)