The Gram-Positive and Gram-Negative of Medical Importance

Dr. Zaid Shaker Naji

Lec (7)

- Bacteria are a large group of unicellular, microscopic organisms, which have been classified as prokaryotic cells, as they lack a true nucleus. These microscopic organisms comprise a simple physical structure, including cell wall, capsule, DNA, pili, flagellum, cytoplasm and ribosomes.

- Bacteria can be gram-positive or gram-negative depending upon the staining methods.

- Gram-positive and gram-negative bacteria are classified based on their ability to hold the gram stain. The gram-negative bacteria are stained by a counterstain such as safranin, and they are de-stained because of the alcohol wash.
- Hence under a microscope, they are noticeably pink in colour. Gram-positive bacteria, on the other hand, retains the gram stain and show a visible violet colour upon the application of mordant (iodine) and ethanol (alcohol).

- Gram-positive bacteria constitute a cell wall, which is mainly composed of multiple layers of peptidoglycan that forms a rigid and thick structure.

- Its cell wall additionally has teichoic acids and phosphate. The teichoic acids present in the gram-positive bacteria are of two types – the lipoteichoic acid and the teichoic wall acid.

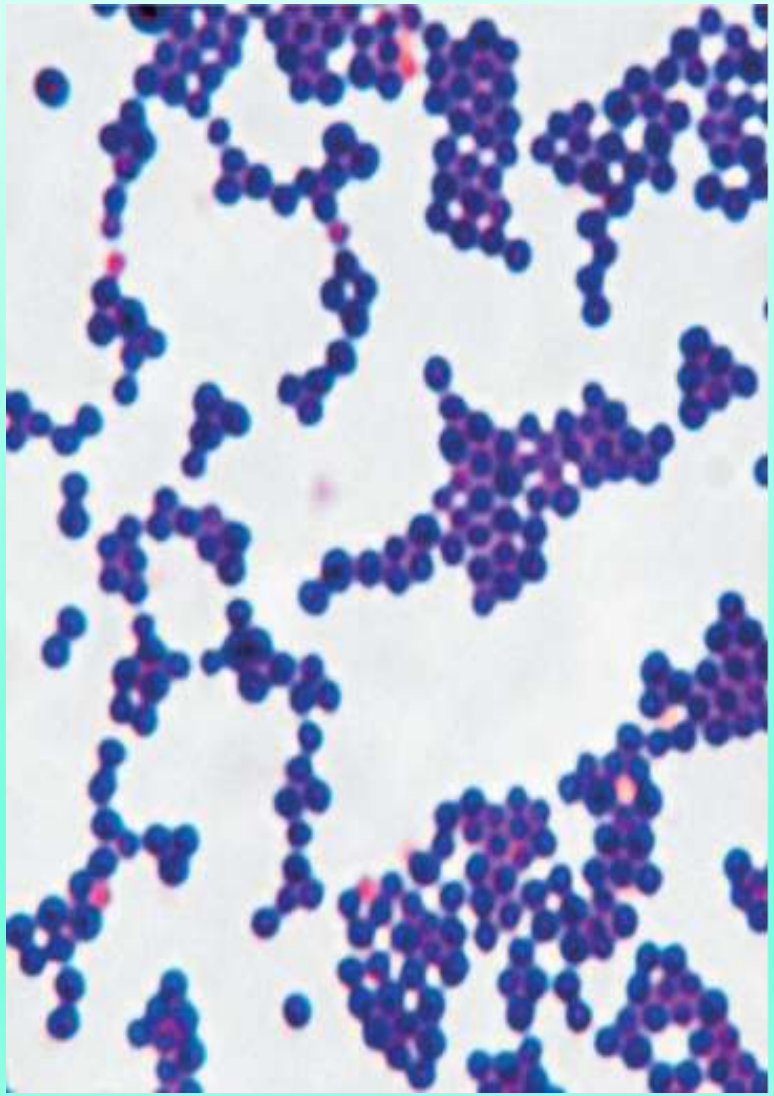
- In gram-negative bacteria, the cell wall is made up of an outer membrane and several layers of peptidoglycan. The outer membrane is composed of lipoproteins, phospholipids, and LPS. The peptidoglycan stays intact to lipoproteins of the outer membrane that is located in the fluid-like periplasm between the plasma membrane and the outer membrane.

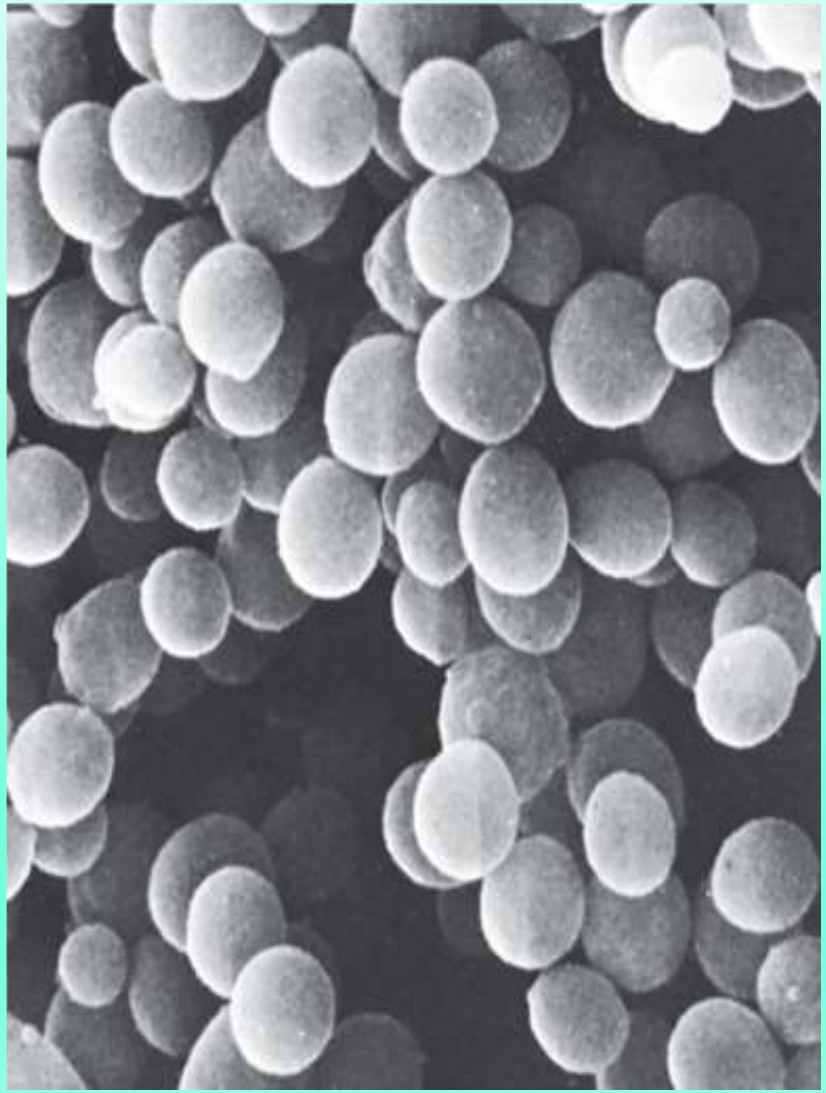
- The periplasm is contained with proteins and degrading enzymes which assist in transporting molecules.

- The cell walls of the gram-negative bacteria, unlike the gram-positive, lacks the teichoic acid. Due to the presence of porins, the outer membrane is permeable to nutrition, water, food, iron, etc.

Staphylococci General Characteristics

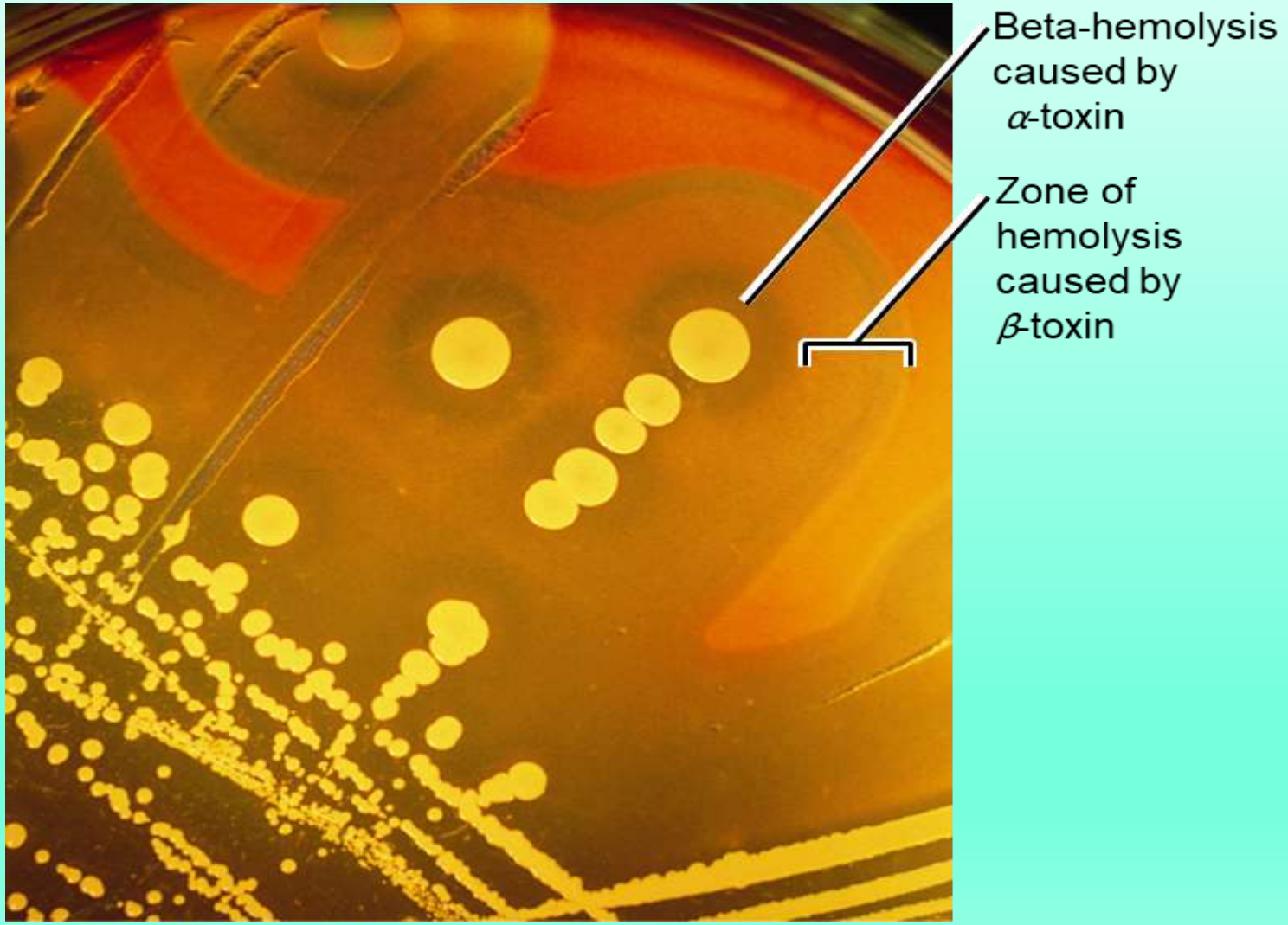
- Common inhabitant of the skin and mucous membranes
- Spherical cells arranged in irregular clusters
- Gram-positive
- Lack spores and flagella
- May have capsules
- 31 species





Staphylococcus aureus

- Grows in large, round, opaque colonies
- Optimum temperature of 37^{o}C
- Facultative anaerobe
- Resist high salt, extremes in pH, and high temperatures
- Produces many virulence factors



Virulence factors of *S. aureus*

Enzymes:

- **Coagulase** – coagulates plasma and blood; produced by 97% of human isolates; diagnostic
- **Hyaluronidase** – digests connective tissue
- **Staphylokinase** – digests blood clots
- **DNase** – digests DNA
- **Lipases** – digest oils; enhances colonization on skin
- **Penicillinase** – inactivates penicillin

Virulence factors of *S. aureus*

Toxins:

- **Hemolysins** (α, β, γ, δ) – lyse red blood cells
- **Leukocidin** – lyses neutrophils and macrophages
- **Enterotoxin** – induce gastrointestinal distress
- **Exfoliative toxin** – separates the epidermis from the dermis
- **Toxic shock syndrome toxin** (TSST) – induces fever, vomiting, shock, systemic organ damage

Epidemiology and Pathogenesis

- Present in most environments frequented by humans
- Readily isolated from fomites
- Carriage rate for healthy adults is 20-60%
- Carriage is mostly in nose, skin, nasopharynx, intestine
- Ability to infection include: poor hygiene and nutrition, tissue injury, already exist a primary infection, diabetes, immunodeficiency
- Increase in community acquired methicillin resistance (**MRSA)**

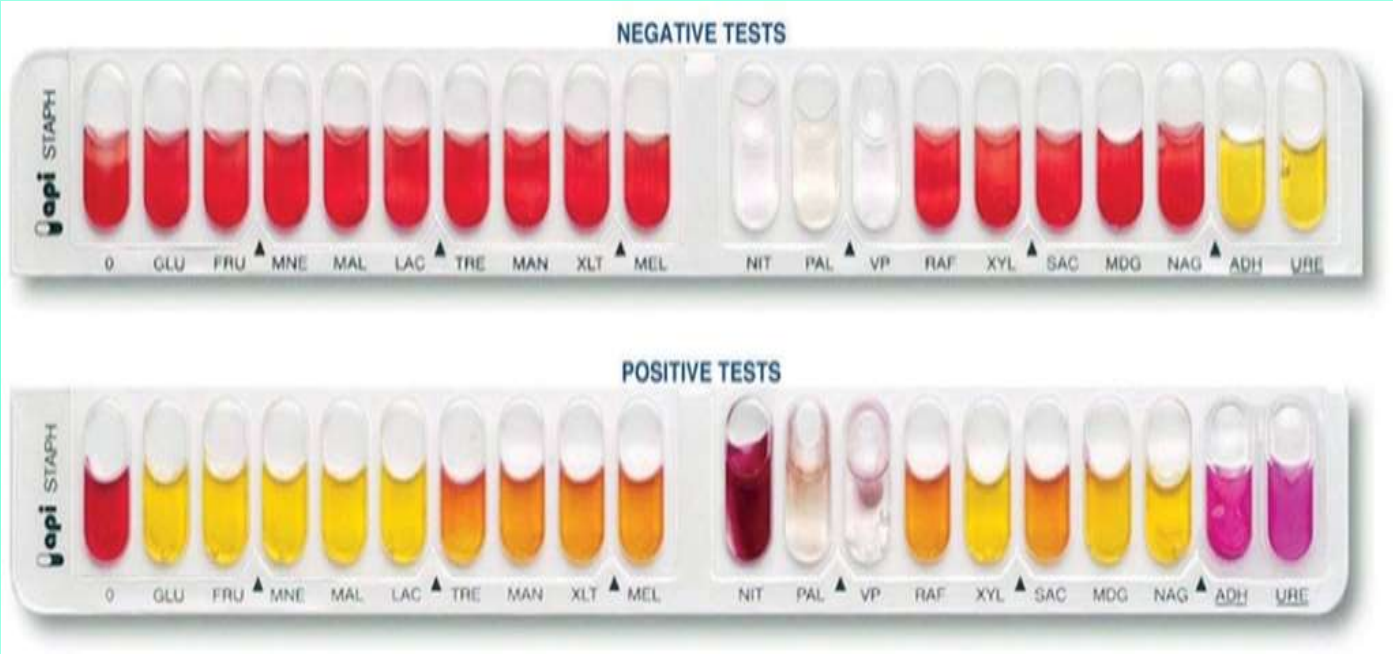
Staphylococcal Disease

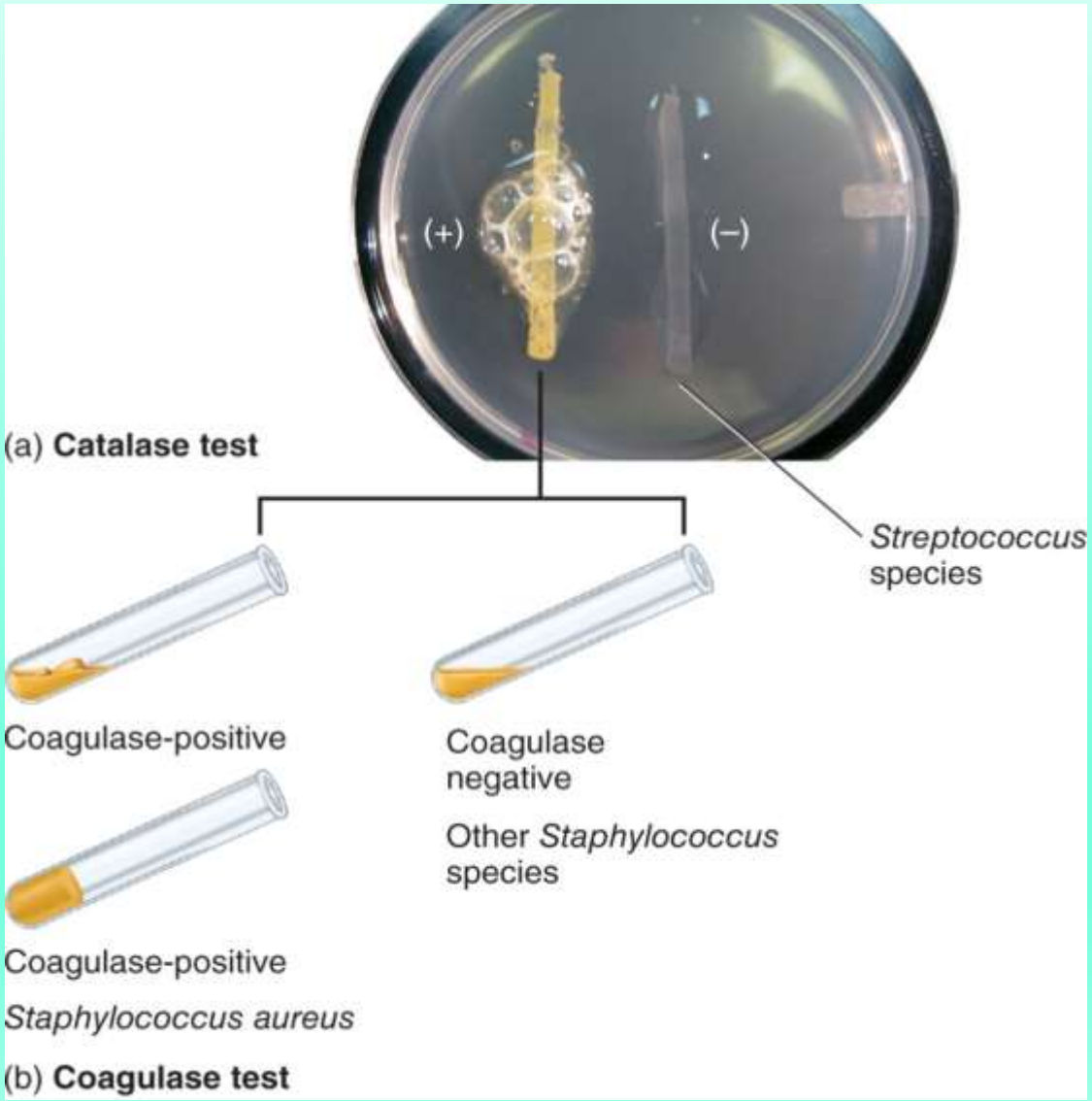
Range from localized to systemic

- **Localized cutaneous infections** – invade skin through wounds, follicles, or glands
- **Systemic infections**
  - **Osteomyelitis** – infection the bone and forming abscess
  - **Bacteremia** – primary origin is bacteria from another infected site or medical devices; endocarditis possible

Identification of *Staphylococcus*

- Frequently isolated from pus, tissue exudates, sputum, urine, and blood
- Cultivation, catalase, biochemical testing, coagulase





Enterobacteriaceae Family

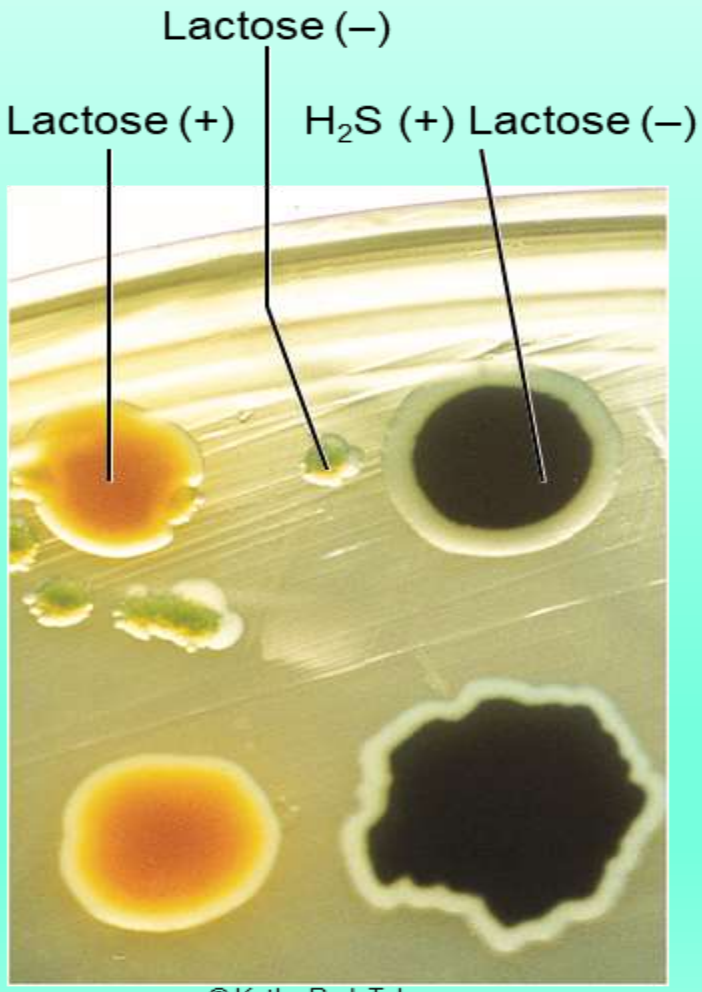
- Large family of small, non-spore-forming gram-negative rods
- Many members inhabit soil, water, decaying matter, and are common occupants of large bowel of animals including humans
- Most frequent cause of diarrhea through **enterotoxins**
- Enterics, along with *Pseudomonas sp.*, account for almost 50% of nosocomial infections

Enterobacteriaceae Family

- Facultative anaerobes, grow best in air
- All ferment glucose, oxidase negative, and catalase positive

- Divided into **coliforms** (lactose fermenters) and **non-coliforms** (non-lactose fermenters)
- Enrichment, selective and differential media utilized for screening samples for pathogens

(a)

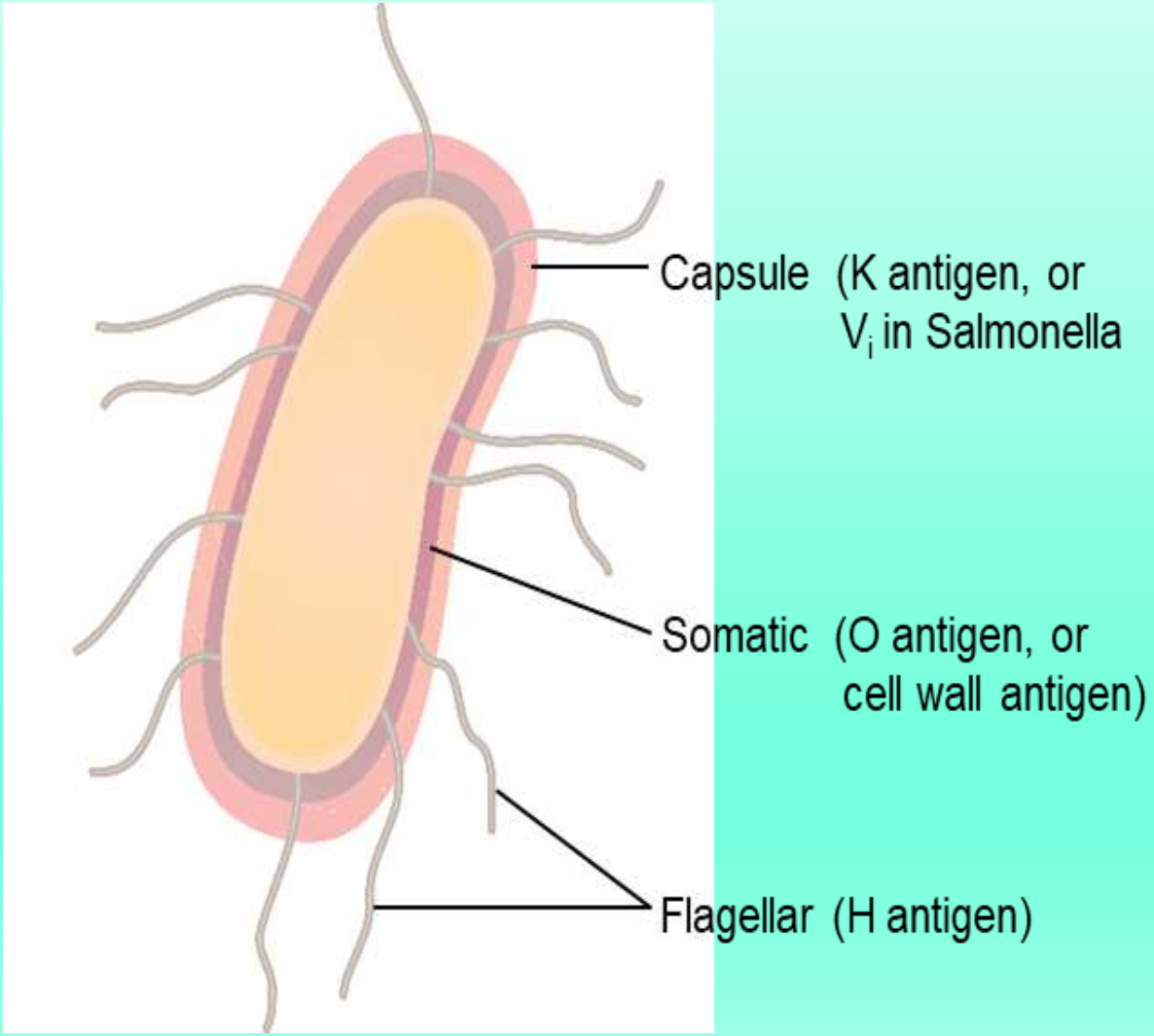


(b)

Antigenic Structures and Virulence Factors

Complex surface antigens contribute to pathogenicity and trigger immune response:

- **H** – flagellar Ag
- **K** – capsule and/or fimbrial Ag
- **O** – somatic or cell wall Ag
- **Endotoxin**
- **Exotoxins**



Escherichia coli: The Most Prevalent Enteric Bacillus

- Most common aerobic bacterium in gut
- 150 strains
- Some have developed virulence through plasmid transfer, others are opportunists

Pathogenic Strains of *E. coli*

- **Enterotoxigenic** *E. coli* causes severe diarrhea due to heat-labile toxin and heat-stable toxin – stimulate secretion and fluid loss
- **Enteroinvasive** *E. coli* causes inflammatory disease of the large intestine
- **Enteropathogenic** *E. coli* linked to wasting form infantile diarrhea
- **Enterohemorrhagic** *E. coli,* causes hemorrhagic syndrome and kidney damage

Escherichia coli

- Pathogenic strains frequent agents of infantile diarrhea – greatest cause of mortality among babies
- Causes ~70% of traveler's diarrhea
- Causes 50-80% UTI
- Coliform count – indicator of fecal contamination in water

نهاية المحاضرة