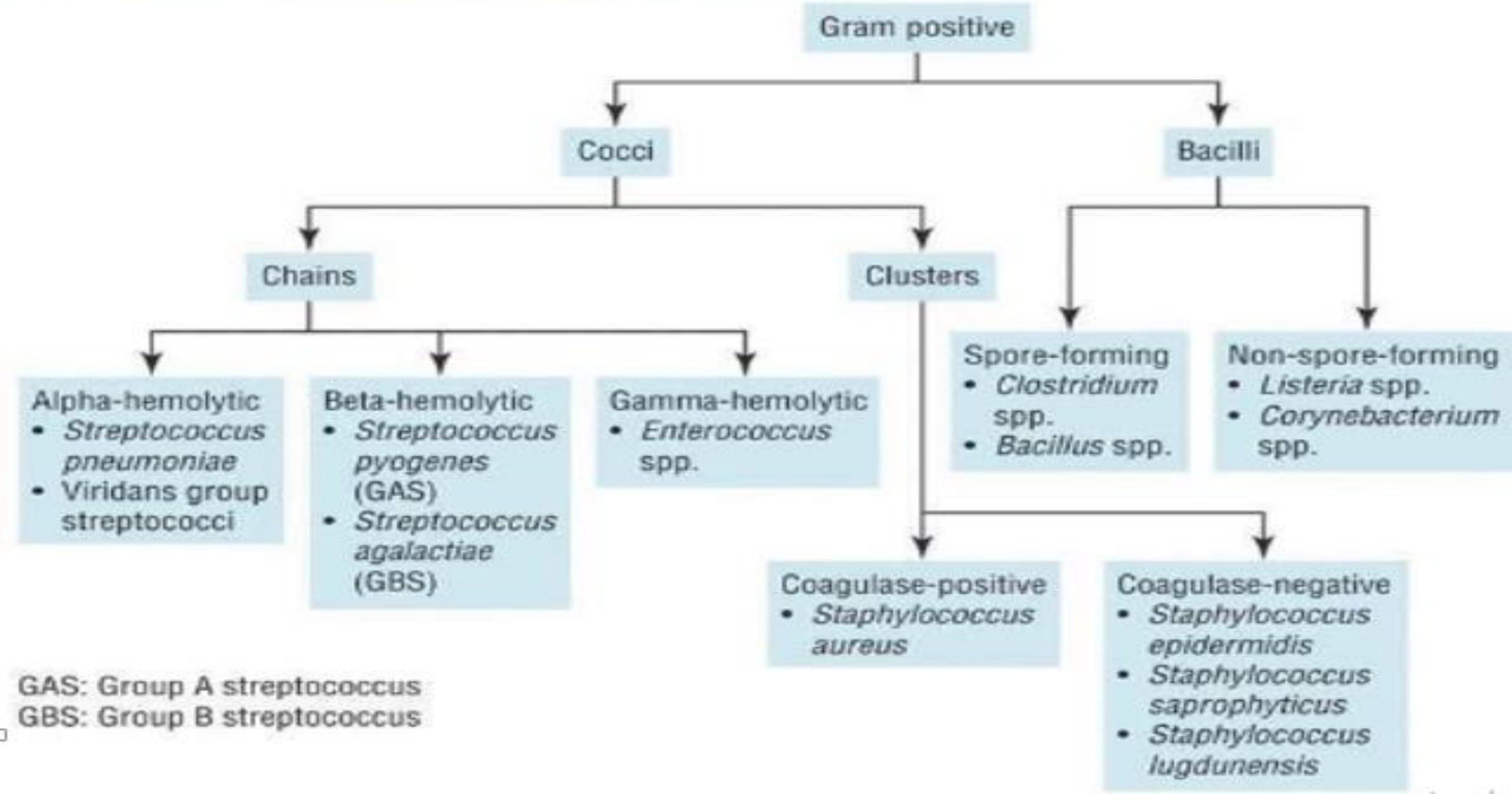
MICROBIOLOGY I
Pharmacy college
2nd stage
Lec 4
Gram Positive Bacilli

Dr. Noor Hassoon

Figure 24.1 Classification of Gram-Positive Bacteria



GAS: Group A streptococcus
GBS: Group B streptococcus

Introduction

- Gram-positive bacilli (rods) comprise a diverse group of bacteria.
- Unlike the cocci, these organisms exhibit significant variation in oxygen requirements, spore-forming capabilities, and pathogenic mechanisms.
- **Classification** based on oxygen requirement and endospore formation:

1- Spore-Forming Aerobes / Facultative Anaerobes:

Primarily represented by the genus **Bacillus** (e.g., *Bacillus anthracis, Bacillus cereus*).

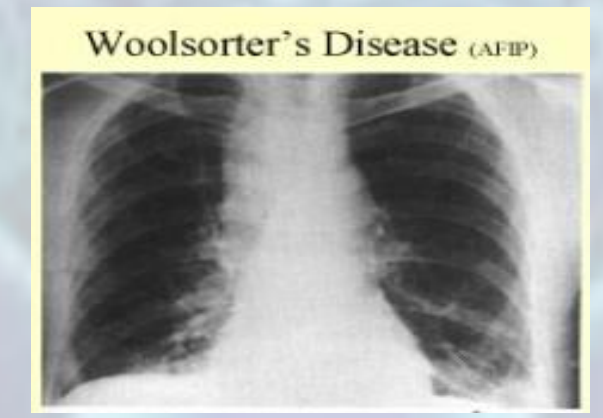
2- Spore-Forming Obligate Anaerobes:

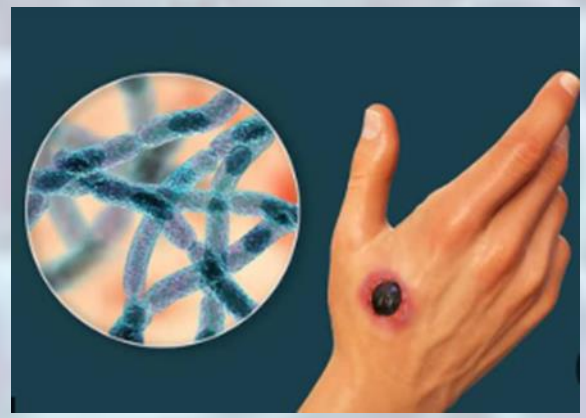
Primarily represented by the genus **Clostridium and Clostridioides** (e.g., *C. tetani, C. botulinum, C. perfringens, C. difficile*).

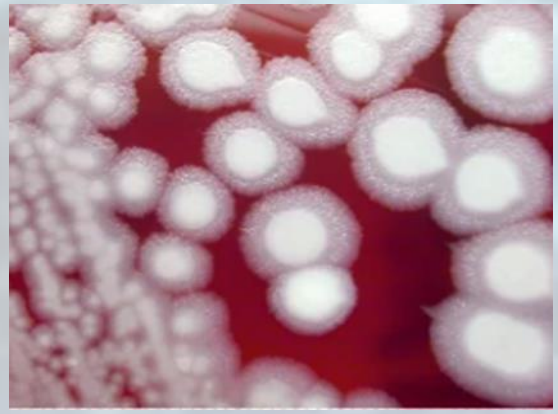
3- Non-Spore-Forming Rods: Includes key pathogens such as ***Corynebacterium diphtheriae*** (aerobic/pleomorphic) and ***Listeria monocytogenes*** (facultative anaerobe/psychrophilic (cold-loving)).

Spore-Forming Aerobes *Bacillus anthracis*: The Agent of Anthrax

- **Morphology:** Large, square-ended **"boxcar"** rods in chains, non-motile; 'Medusa-head' colonies.
- **Primary Niche:** Soil; Zoonotic pathogen (livestock/contaminated wool)
- **Virulence Factors:**
  - Antiphagocytic **Poly-D-glutamic acid capsule** (unique polypeptide capsule, not polysaccharide).
  - **Anthrax Toxin Trio**: Protective Antigen (PA), Edema Factor (EF - an adenylate cyclase), and Lethal Factor (LF - a zinc metalloprotease).







- **Clinical Manifestations:** Cutaneous (painless black eschar), Gastrointestinal, and Inhalation (**Woolsorter's disease**; highly fatal hemorrhagic mediastinitis).
- **Pharmacotherapy: Ciprofloxacin** or **Doxycycline** combined with an anti-toxin monoclonal antibody (e.g., Raxibacumab).

- **Resistance Mechanisms:**
  - ***Beta-Lactamase Production:*** Naturally encodes inducible **Bla1 (penicillinase)** and **Bla2 (cephalosporinase)** enzymes.
  - ***Fluoroquinolone Resistance:*** Driven by **target-site mutations** in the gyrA (DNA gyrate) and parC (Topoisomerase IV) genes, alongside the upregulation of active **efflux pumps** that pump the drug out of the cell.

Spore-Forming Aerobes – *Bacillus cereus*: "Reheated Rice" Syndrome

- **Primary Niche:** soil, contaminated food.
- **Pathogenesis:** Spores survive the boiling of rice.

If left at room temperature, spores germinate and release toxins.

- **Two Distinct Clinical Syndromes:**
  - ***Emetic Type:*** Rapid onset (1–6 hours); caused by a **heat-stable enterotoxin** (preformed in food); presents with severe vomiting.
  - ***Diarrheal Type:*** Delayed onset (8–16 hours); caused by a **heat-labile enterotoxin** (produced in the gut); presents with watery diarrhea.



- **Pharmacotherapy:** Usually **self-limiting**. Empiric antibiotics are not indicated for standard gastroenteritis. Severe systemic or ocular infections require **Vancomycin** or Clindamycin (inherently resistant to penicillins).
- **Resistance Mechanisms:**
  - ***Constitutive Beta-Lactamases:*** Produces highly potent, broad-spectrum **beta-lactamases (including metallo-beta-lactamases)**. This renders them intrinsically resistant to almost all penicillins and cephalosporins.
  - ***Aminoglycoside Modifying Enzymes (AMEs):*** Systemic strains can acquire enzymes that phosphorylate or acetylate aminoglycosides, rendering drugs like gentamicin ineffective.

Spore-Forming Anaerobes – *Clostridium tetani*: Spastic Paralysis

- **Morphology & Habitat:** Obligate anaerobe; terminal spores give a **"drumstick"** appearance; found widely in soil.
- **Pathogenesis:** Deep puncture wounds introduce spores into necrotic, anaerobic tissue.





- **Virulence:**
- **Exotoxin: Tetanospasmin** (neurotoxin). Cleaves SNARE proteins, blocking the release of inhibitory neurotransmitters (**GABA and Glycine**) from Renshaw cells in the spinal cord.
- **Clinical Presentation: Lockjaw (trismus)**, Risus sardonicus (sardonic smile), and opisthotonos (severe hyperextension of the spine).

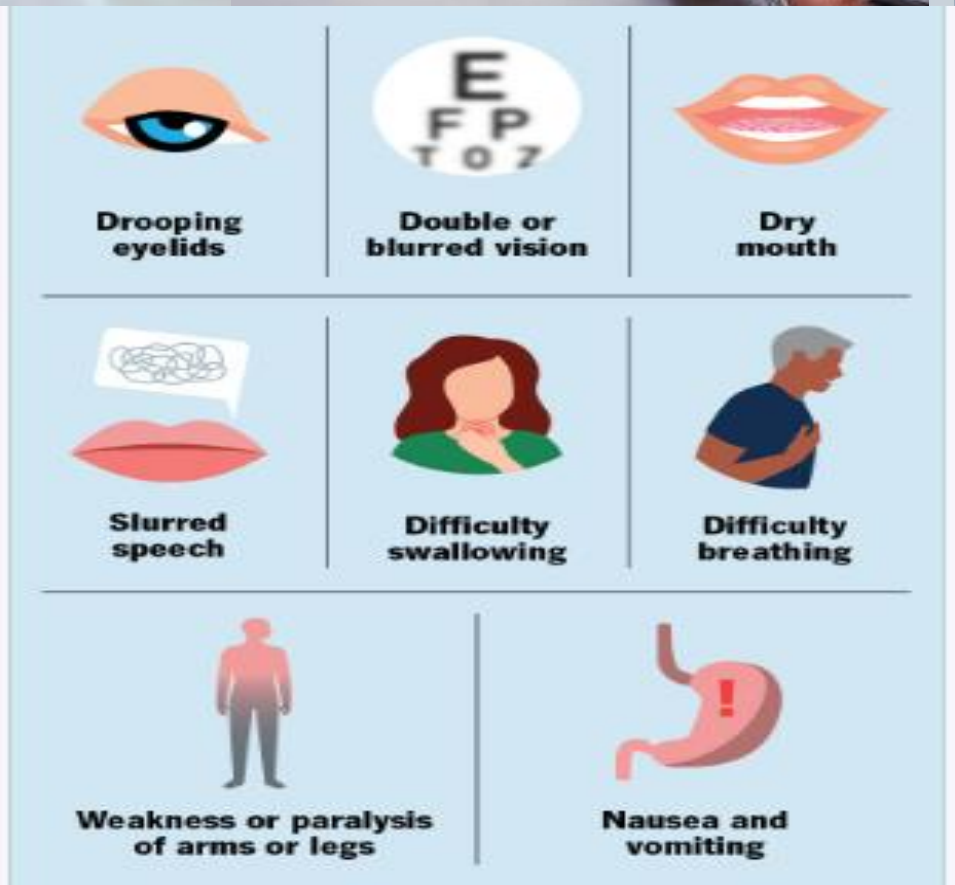
- **Pharmacotherapy:** Human tetanus immune globulin (HTIG) to neutralize toxin, wound debridement, and **Metronidazole**.

- **Resistance Mechanisms:**
  - **Nitroreductase Alteration*:*** Metronidazole requires intracellular reduction to its active free-radical form. Mutations that decrease this specific reductive metabolism cause resistance.

Spore-Forming Anaerobes – *Clostridium botulinum*: Flaccid Paralysis

- **Pathogenesis:**
  - *Adults:* Ingestion of **preformed toxin** from poorly preserved canned goods.
  - *Infants:* Ingestion of ***spores*** **from honey**, which germinate in the immature infant gut (Floppy Baby Syndrome).
- **Virulence:** Exotoxin -(Botulinum toxin)- Cleaves SNARE proteins at the neuromuscular junction, completely blocking **Acetylcholine (ACh)** release.
- **Clinical Presentation:** (Botulism) Flaccid, **descending paralysis** starting with cranial nerves (diplopia, dysphagia, ptosis) and leading to respiratory failure.







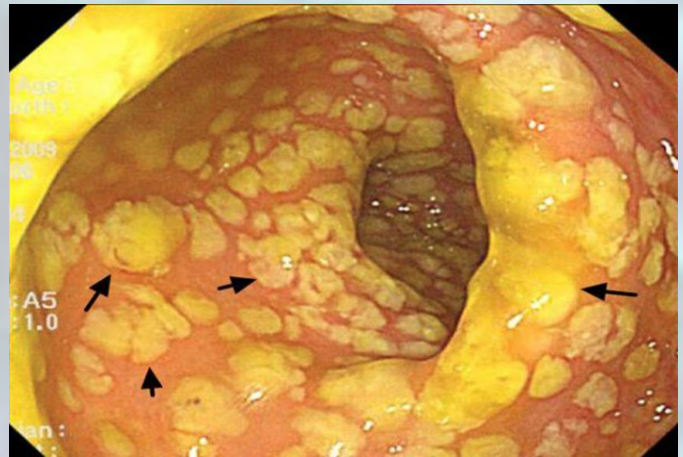
- **Pharmacotherapy:** Equine Antitoxin or Human Botulism Immunoglobulin.
- **Pharmacy Application:** Purified Botulinum Toxin Type A (Botox) used clinically for focal dystonias, migraines, and cosmetic applications.



- **Resistance Mechanisms:**
- Minimal Acquired Antibiotic Resistance: Like C. tetani, it displays minimal plasmid-mediated resistance.
- **Toxin Neutralization Resistance:** The primary 'resistance' issue in clinical settings is immunological resistance. Long-term cosmetic or therapeutic users of Botox can develop neutralizing antibodies against the heavy chain of the botulinum toxin, rendering the clinical protein therapy ineffective.

Spore-Forming Anaerobes – *Clostridioides difficile:* Antibiotic-Associated Colitis

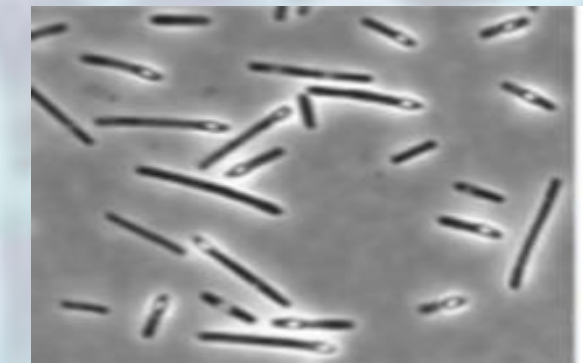
- **Pathogenesis:** Broad-spectrum antibiotic use (e.g., Clindamycin, Fluoroquinolones) killed normal colonic flora → *C. diff* spores germinate and overgrow.
- **Virulence Factors:**
  - *Toxin A (Enterotoxin):* Destroys brush border enzymes, causing watery diarrhea.
  - *Toxin B (Cytotoxin):* Induces actin depolymerization, leading to cellular apoptosis and plaque-like pseudomembranes.
- **Clinical Outcome: Pseudomembranous colitis**, severe abdominal pain, leukocytosis.
- **Pharmacotherapy Selection: Oral Vancomycin** or **Fidaxomicin**.



- **Resistance Mechanisms:**
  - *Hypervirulent BI/NAP1/027 Strain:* Displays high resistance to **fluoroquinolones** due to mutations in the **gyrA** gene, allowing it to rapidly outcompete other gut flora in hospitalized patients taking these drugs.
  - ***Vancomycin MIC Creep:*** Driven by alterations in cell wall synthesis pathways or thick biofilm formations, making the bacteria progressively less sensitive to standard vancomycin dosing.
  - ***Fidaxomicin Resistance:*** Can occur via point mutations in the **rpoB gene** (which encodes the beta subunit of RNA polymerase), directly altering the drug's target binding site.

Spore-Forming Anaerobes – *Clostridium perfringens*: Myonecrosis & Gas Gangrene

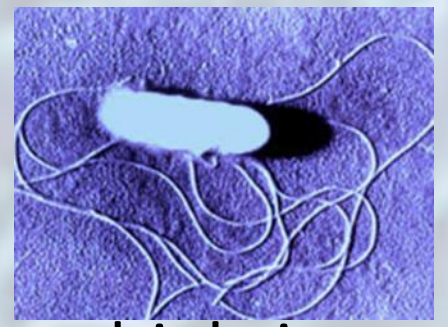
- **Morphology:** Large, boxcar-shaped rods , non-motile , double zone of hemolysis on blood agar.
- **Habitat:** Soil and human colonic flora. Introduced via traumatic, contaminated wounds.
- **Virulence Factor:** Alpha-toxin (Lecithinase). Splits lecithin into diglycerides, destroying cell membranes, RBCs (hemolysis), and muscle tissues.
- **Clinical Signs:** Gas gangrene (crepitus upon palpation due to bacterial gas production), foul-smelling discharge, and rapid tissue necrosis.





- **Pharmacotherapy:** High-dose Penicillin G combined with Clindamycin (Clindamycin is added because it suppresses bacterial protein synthesis and toxin production directly).
- **Resistance Mechanisms:**
- ***Clindamycin Resistance*** *(MLS_B Phenotype):* Mediated by the **erm** (Erythromycin Ribosome Methylase) gene. This gene encodes an enzyme that **methylates the 23S rRNA** component of the 50S ribosomal subunit. This alters the binding site, causing cross-resistance to Macrolides, Lincosamides (Clindamycin), and Streptogramins.
- ***Penicillin Resistance****:* Though rare, it can occur via modifications in Penicillin-Binding Proteins (PBPs), lowering their affinity for beta-lactam drugs.

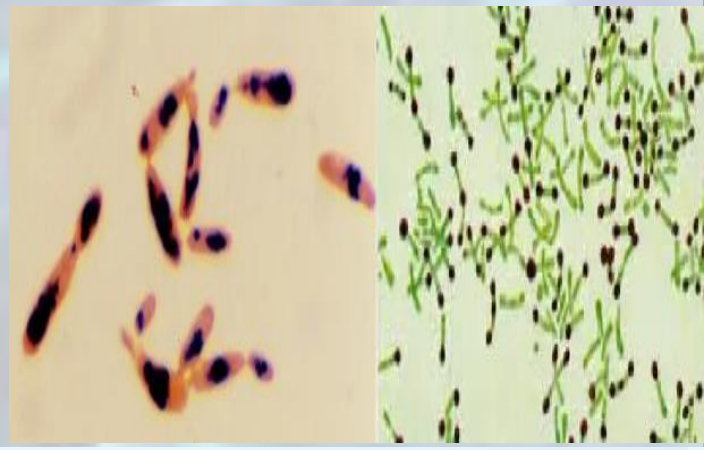
Non-Spore-Forming Rods – *Listeria monocytogenes*: Intracellular Opportunist



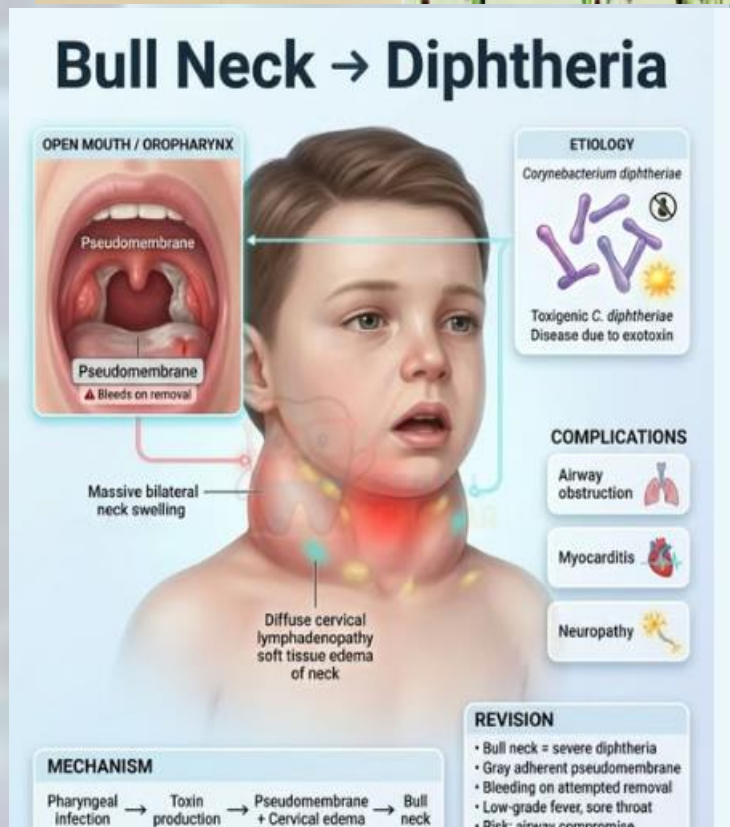
- **Characteristics:** Non-spore-forming, catalase-positive, exhibits "tumbling motility" at room temperature.
- **Environmental Adaptation:** Psychrotrophic (can grow and multiply in refrigerated temperatures); cross-contaminates unpasteurized milk, soft cheeses, and deli meats.
- **Pathogenesis:** Facultative intracellular pathogen. Escapes phagosomes using *Listeriolysin O*, then uses *ActA* proteins to polymerize host actin ("actin rockets") to move directly cell-to-cell.
- **At-Risk Populations:** Pregnant women (causes amnionitis, spontaneous abortion), newborns (neonatal meningitis), and immunocompromised individuals.

- **Pharmacotherapy:** Ampicillin (Note: Cephalosporins have no activity against *Listeria* due to altered penicillin-binding proteins).
- **Resistance Mechanisms:**
  - ***Intrinsic Cephalosporin Resistance:*** *Listeria* naturally possesses **low-affinity Penicillin-Binding Proteins (specifically PBP3)**. Cephalosporins cannot bind effectively to these proteins, making the entire drug class useless.
  - ***Efflux Pump Upregulation:*** Active multidrug efflux pumps (such as **MdrL**) pump out macrolides and fluoroquinolones, preventing them from building up inside the bacterial cell.

Non-Spore-Forming Rods – *Corynebacterium diphtheriae*: The Pseudomembranous Throat



- **Morphology:** Club-shaped, arranged in V or L shapes resembling "Chinese letters." Contains metachromatic granules.
- **Pathogenesis:** Acquired via respiratory droplets.
- **Virulence:**
- **Exotoxin:** Diphtheria Toxin, completely halting host protein synthesis.
- **Clinical Presentation:** Thick, gray-white **pseudo-membrane** over the pharynx/tonsils (risk of airway obstruction); severe cervical lymphadenopathy ("bull neck"); systemic myocarditis.



- **Pharmacotherapy & Prevention:** Erythromycin or Penicillin G, plus immediate Diphtheria Antitoxin. Prevented globally via the **DTaP** vaccine (toxoid).
- **Resistance Mechanisms:**
  - ***Macrolide Resistance:*** Driven by the acquisition of **erm(X) genes** via plasmids or transposons. Just like in *C. perfringens*, this causes ribosomal methylation at the 23S rRNA site, preventing erythromycin from stopping protein synthesis.
  - ***Penicillin Tolerance:*** Some strains show alterations in cell wall cross-linking enzymes, allowing them to survive standard bactericidal concentrations of penicillin without undergoing cell lysis.


Thank
you!