

**Tishk International University  
Faculty of Science  
Medical Analysis Department**



# ..... Medical Microbiology.....

**TOPIC:** Spore forming Gram positive bacteria

**2<sup>nd</sup> Grade- Spring Semester 2019-2020**



# CLASSIFICATION

Spore forming GPB

Aerobic

Bacillus

Anaerobic

Clostridium



# CLOSTRIDIUM

- Anaerobic, endospore former, gram positive rod
- Clostridia are ubiquitous in soil, water, and sewage
- Part of the normal microbial population in the GI tracts of animals and humans

## Medically important species are

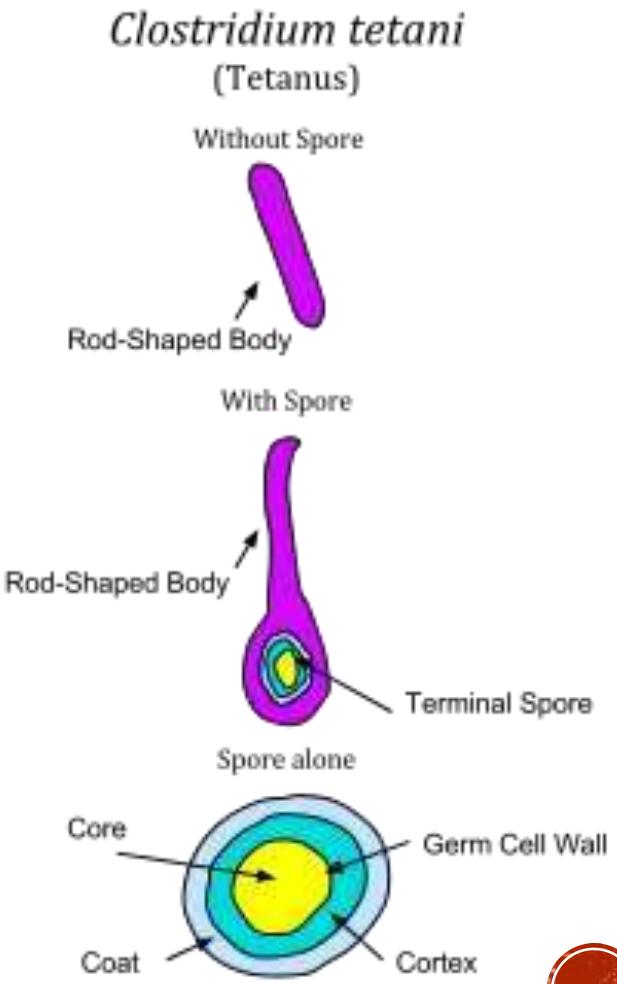
- *Clostridium tetani*
- *Clostridium botulinum*
- *Clostridium erfingen*
- *Clostridium difficile*



**CLOSTRIDIUM TETANI**

# MORPHOLOGY

- Large, motile, spore forming rod
- It produce round terminal spore (drumstick appearance)
- Spores are widespread in soil



# CLOSTRIDIUM TETANI GRAM STAIN

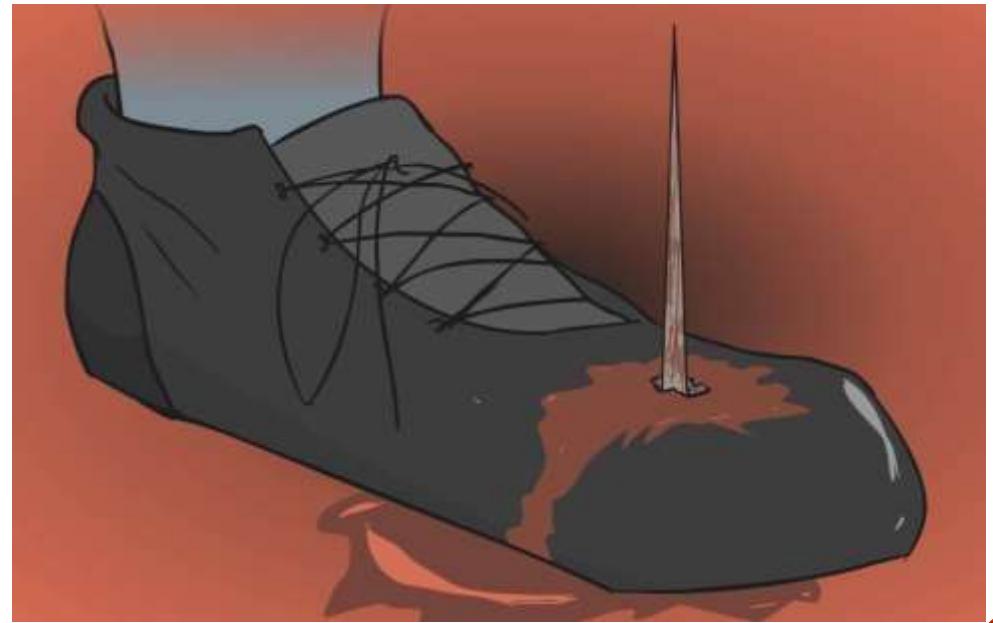


visuals:unlimited



# TRANSMISSION

- Portal of entry is wound site
- In neonatal tetanus, it may enter through contaminated umbilicus or circumcision wound.



## Two toxins

- Oxygen-labile hemolysin (tetanolysin)
- Heat-labile neurotoxin (tetanospasmin), Plasmid encoded



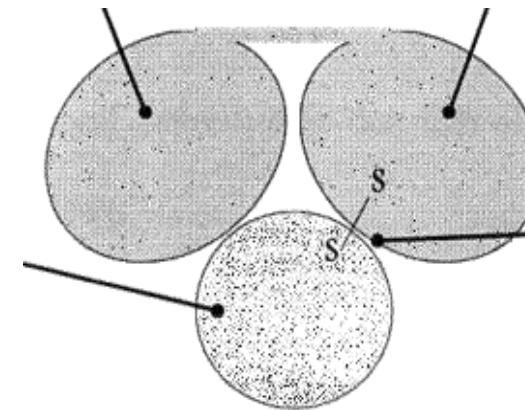
# TETANOLYSIN

- Clinical significance is unknown
- Inhibited by oxygen and serum cholesterol.
- serologically related to streptolysin O and the hemolysins.



# TETANOSPASMIN

- Released during stationary phase of growth.
- Responsible for clinical manifestations of tetanus.
- Two part (A-B)



■ Heavy chain (100 kDa)  
■ Light chain (50 kDa)

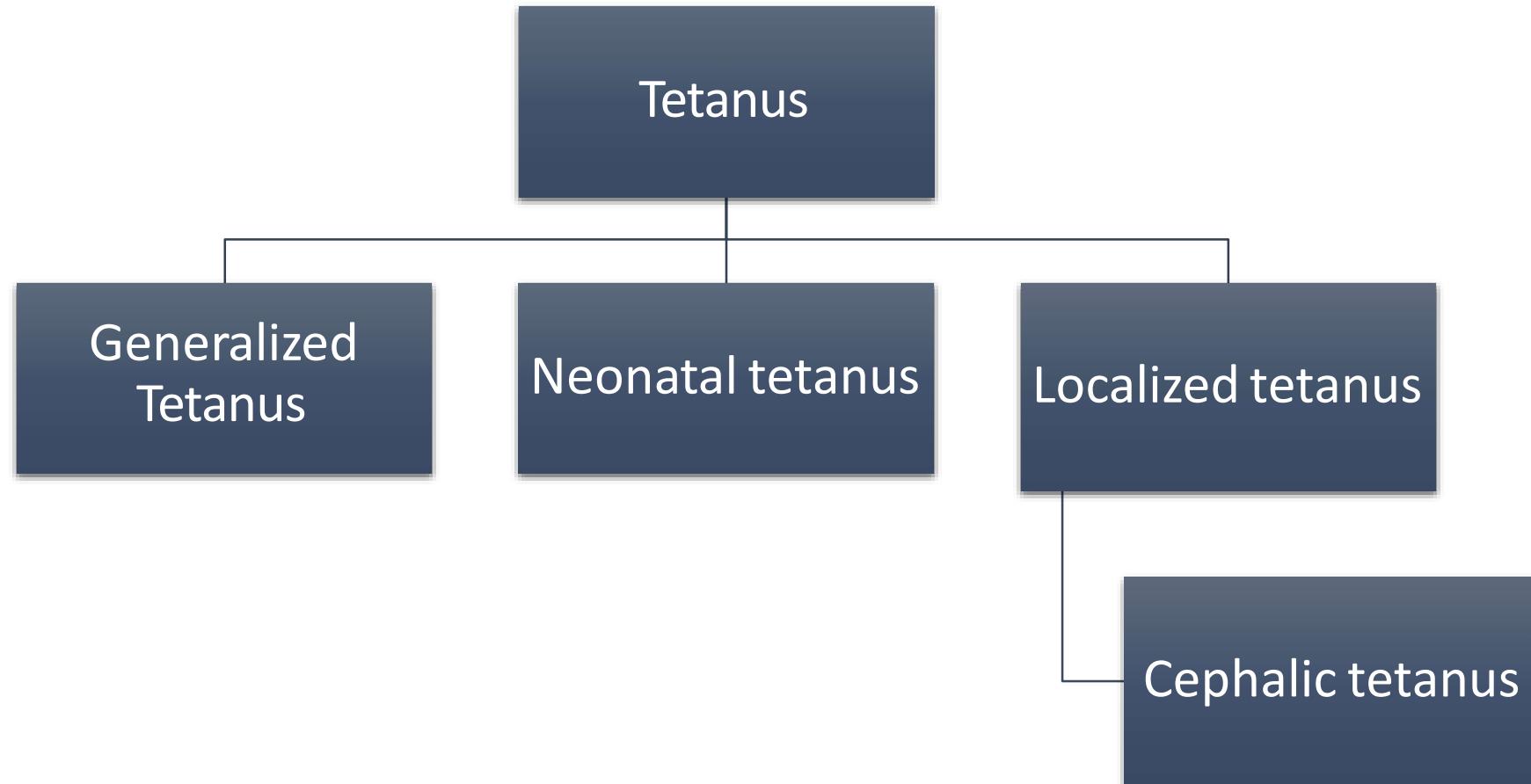


## Incubation period

- varies from a few days to weeks.
- The duration of the incubation period is directly related to the distance of the primary wound infection from the central nervous system.



# CLINICAL DISEASE



# GENERALIZED TETANUS

- Most common form.
- Involvement of the masseter muscles (trismus or lockjaw) is the presenting sign in most patients.
- characteristic sardonic smile that results from the sustained contraction of the facial muscles is known as *risus sardonicus*
- Other early signs are drooling, sweating, irritability, and persistent back spasms (opisthotonus)



# GENERALIZED TETANUS



**FIGURE 30-6** Facial spasm and risus sardonicus in a patient with tetanus. (From Cohen J, Powderly WG, Opal SM: *Infectious diseases*, ed 3, Philadelphia, 2010, Mosby.)



**FIGURE 30-7** A child with tetanus and opisthotonus resulting from persistent spasms of the back muscles. (From Emond RT, Rowland HAK, Welsby P: *Colour atlas of infectious diseases*, ed 3, London, 1995, Wolfe.)

# LOCALIZED TETANUS

- Disease remains confined to the musculature at the site of primary infection.

## Cephalic tetanus

- A variant in which the primary site of infection is the head
- The prognosis for patients with cephalic tetanus is very poor.



# NEONATAL TETANUS

## Neonatal tetanus (*tetanus neonatorum*)

- Typically associated with an initial infection of the umbilical stump
- progresses to become generalized.
- The mortality in infants exceeds 90%
- developmental defects are present in survivors.
- This is almost exclusively a disease in developing countries.



# NEONATAL TETANUS



# TREATMENT

- Debridement of the primary wound
- Use of penicillin or metronidazole to kill the bacteria and reduce toxin production

## Prevention:

- Vaccination with tetanus toxoid



**CLOSTRIDIUM BOTULINUM**

# **CLOSTRIDIUM BOTULINUM**

- large, spore forming, anaerobic rods.
- Spores are widespread in soil, contaminate vegetables and meat.
- If foods are canned without sterilization, spore will survive and germinate under anaerobic condition and bacteria will produce toxin



- Similar to tetanus toxin, C. botulinum toxin a protein (A-B toxin) consisting of a small subunit (light, or A chain)



# CLINICAL USE OF TOXIN

Botox is a commercial preparation of exotoxin A used to remove wrinkles on the face

**Effective in minute amount in certain muscle disorders**



# CLINICAL DISEASE

- Descending weakness and paralysis
- Diplopia
- Dysphagia
- Respiratory muscle failure
- No fever

## Botulism

### Wound botulism

Spores contaminate wound, germinate and produce toxin at site

### Infant botulism

Organism grow in gut and produce toxin. Ingestion of honey containing the organism is the cause

# TREATMENT

**Patients with botulism require the following treatment measures:**

- Adequate ventilatory support
- Elimination of the organism from the GI tract through the judicious use of gastric lavage and metronidazole or penicillin therapy,
- Use of trivalent botulinum antitoxin to inactivate unbound toxin circulating in the bloodstream.



# PREVENTION

- **Disease is prevented by**

- Destroying the spores in food (virtually impossible for practical reasons)
- Preventing spore germination (by maintaining the food in an acid pH or storage at 4° C or colder)
- Destroying the preformed toxin (Toxins are inactivated by heating at 60° C to 100° C for 10 minutes).
- Infant botulism has been associated with consumption of honey contaminated with *C. botulinum* spores, so children younger than 1 year should not eat honey.



**CLOSTRIDIUM PERFRINGENS**

# **CLOSTRIDIUM PERFRINGENS**

- Most frequent clinical isolate of clostridium
- Inhabits in soil and in intestine of animals and humans.
- Rapidly grows in tissues and in culture
- Large, rectangular, gram positive bacilli
- Capsulated, non motile with subterminal spore



# TOXINS AND ENZYMES

## Epsilon toxin

- Increase vascular permeability of gastrointestinal wall

## Iota toxin

- necrotizing activity
- increase vascular permeability

## Enterotoxin

- Produced primarily by type A strain
- Disrupts ion transport in ileum and jejunum by inserting into the cell membrane and altering membrane permeability.



## **Clostridial myonecrosis (gas gangrene)**

- Military settings- Gutshot injuries
- Civilian cases - accidental injuries, surgical complication, injection of medication such as epinephrine.



## ■ Simple wound contamination

- Care of wound
- removal of necrotic tissue
- cleansing
- Antibiotics - rarely

required

## ■ Anaerobic cellulitis:

- Opening the involved area
- removing all necrotic tissue
- cleansing thoroughly
- Antibiotics



# TREATMENT

## **Clostridial myonecrosis**

- Care of wound : surgical removal of all infected and necrotic tissue
- Amputation- rapidly progressive infection involving limb
- Antibiotics - Penicillin G/ Clindamycin or Metronidazole, Gentamicin or Tobramycin (for facultative Gram negative organisms)



# **CLOSTRIDIUM DIFFICILE**

# GENERAL CRITERIA

- Part of normal microbial flora of GIT (3 – 5% adult, 40 – 50% healthy neonates)

## Morphology:

- large gram positive rod.
- having oval subterminal spore.



# VIRULENCE FACTORS

## Enterotoxin (Toxin A)

- produce chemotaxis; induce cytokine production, with hypersecretion of fluid;
- produces haemorrhagic necrosis.

## Cytotoxin (Toxin B)

- induces depolymerization of actin with loss of cellular cytoskeleton.

## Adhesin factor

- binding to colonic cells.



# TRANSMISSION

- Exogenous - Person to person in hospital.
- Endogenous -Over growth of toxin producing strains after Rx of Antibiotics.

## **Antibiotics implicated in Cl.difficile Associated Diarrhoea and Colitis:**

- Cephalosporin
- Ampicillin and amoxicillin
- Clindamycin.
- Other Penicillins, Macrolides, Tetracyclines.

