CLOSTRIDIUM

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Clostridium

C. perfringens: gas gangrene; food poisoning
C. tetani: tetanus
C. botulinum: botulism
C. difficile: pseudomembranous colitis

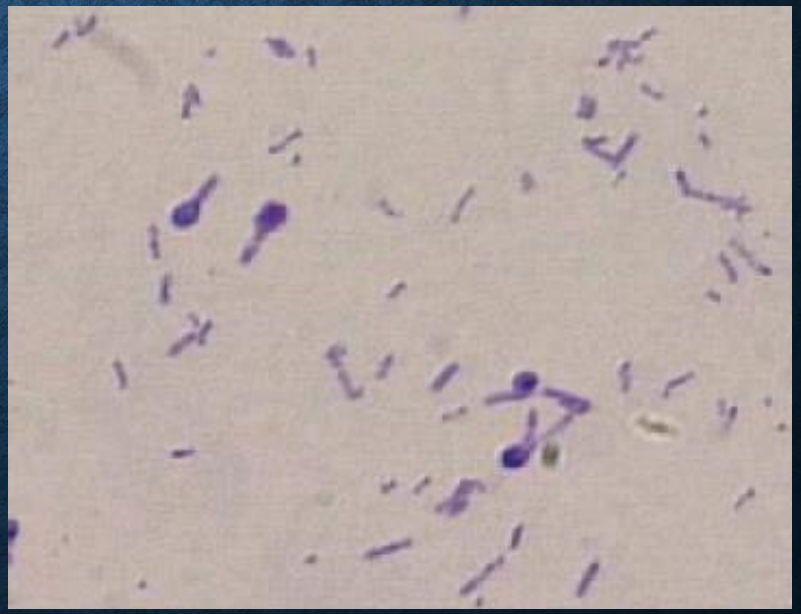
Physiology and Structure

Anaerobic.

Large gram-positive rods.

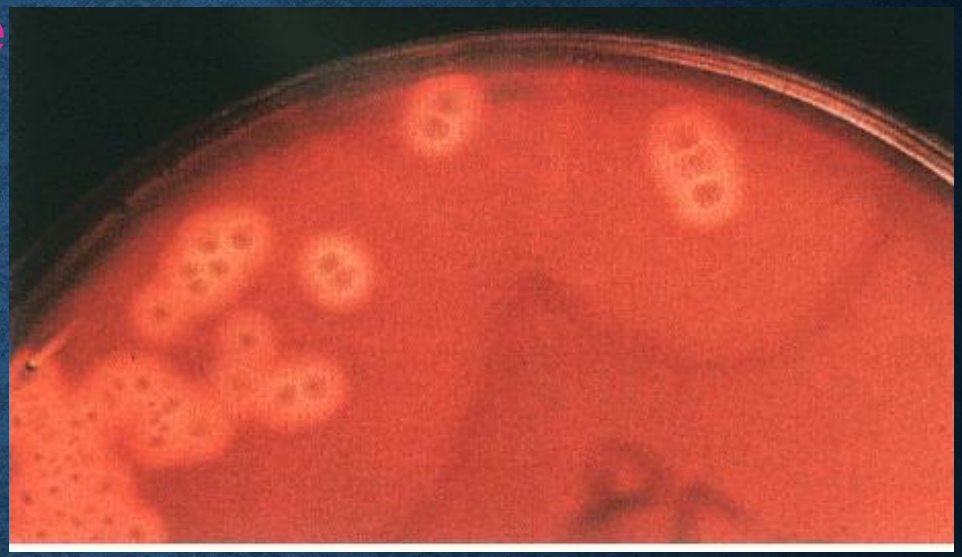
The spores are usually wider than the rods, and are located terminally or subterminally.

Most clostridia are motile by peritrichous flagella.



C. perfringens

Physiology and Structure



Large gram-positive bacilli.

Spores are rarely observed.

Non-motile; capsulated.

Hemolytic and metabolically active.

Subdivided into 5 types based on the four major lethal toxins they produce. Type A causes most of the human infections.

Type of Isolate	Lethal Toxins			
	α	β	ϵ	ι
A	+	−	−	−
B	+	+	+	−
C	+	+	−	−
D	+	−	+	−
E	+	−	−	+

C. perfringens

Pathogenicity and Immunity

Strains of *C. perfringens* are widely distributed in nature, and inhabit the intestine of humans and animals.

They (type A strains are most commonly isolated from human infections) cause a spectrum of diseases primarily by producing toxins and enzymes:

- α-toxin: lecithinase (phospholipase C) that lyses a variety of cells. Lethal, necrotizing and hemolytic. Increases vascular permeability, resulting in massive hemolysis and bleeding, tissue destruction, hepatic toxicity, and myocardial dysfunction.
- Other necrotizing and hemolytic toxins
- DNase, hyaluronidase
- Enterotoxin: produced primarily by type A strains.

C. perfringens

Clinical Diseases

Soft tissue infections

Portal of entry: trauma or intestinal tract.

Usually caused by mixed infection including toxigenic clostridia, proteolytic clostridia and various cocci and gram-negative organisms.

Three types of infections with increasing severity:

Cellulitis: gas formation in the soft tissue.

Fasciitis or suppurative myositis: accumulation of gas in the muscle planes.

Myonecrosis or gas gangrene: a life-threatening disease.

C. perfringens

Clinical Diseases

Gas gangrene

Spores germinate → vegetative cells multiply, ferment carbohydrates and produce gas in the tissue. This results in distension of tissue and interference with blood supply → the bacteria produce necrotizing toxin and hyaluronidase, which favor the spread of infection → tissue necrosis extends, resulting in increased bacterial growth, hemolytic anemia, then severe toxemia and death.

Incubation: 1-7 days after infection.

Symptoms: Crepitation in the subcutaneous tissue and muscle, foul smelling discharge, rapidly progressing necrosis, fever, hemolysis, toxemia, shock, renal failure, and death.

Can be also caused by other *Clostridium* species.

C. perfringens

Clinical Diseases

Food poisoning

The enterotoxin causes marked hypersecretion in jejunum and ileum.

Enterotoxin: a heat-labile protein produced by some strains of *C. perfringens* type A. When >10^8 cells in contaminated meat are ingested and sporulate in the small intestine, enterotoxin is formed. It disrupts ion transport in the enterocytes, and induces antibodies (non-protective) in adults.

Symptoms: diarrhea, usually without vomiting or fever.

Necrotizing enteritis (pig-bel): a fatal disease (acute necrosis in jejunum attributed to β-toxin) in children in New Guinea caused by type C *C. perfringens*.

Clotridium bacteremia usually occurs in patients with tumors.

C. perfringens

Laboratory Diagnosis

Specimens: pus, necrotic tissue, feces, food, etc.

Smears: large gram-positive rods with or without spores, usually in the absence of leukocytes.

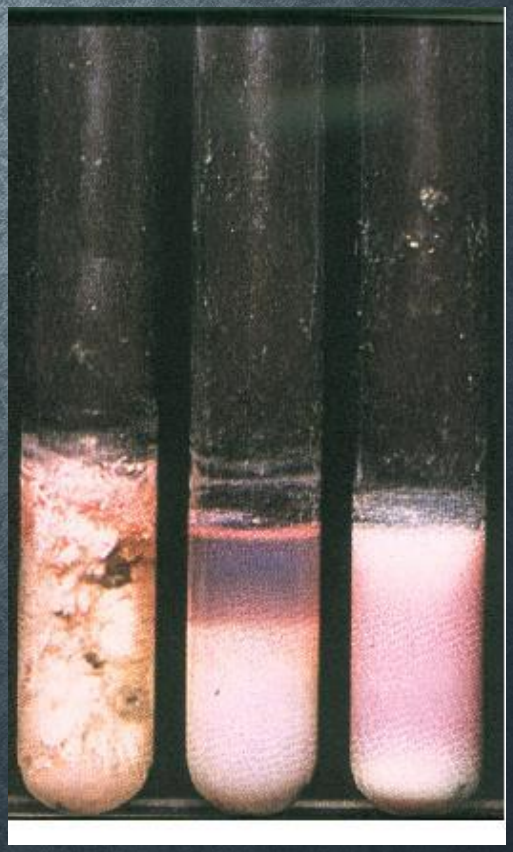
Culture: anaerobic culture on blood plate.

Identification:

"Storming fermentation"-- clot torn by gas in 24 hrs.

Lecithinase test-- precipitate formed around colonies on egg yolk media.

Biochemical tests.



C. perfringens

Treatment

Treatment for suppurative myositis or myonecrosis:

- Prompt and extensive débridement.
- Antibiotics (penicillin) administration.
- Hyperbaric oxygen may "detoxify" patients rapidly.
- Efficacy of antitoxins is doubtful.

C. perfringens food poisoning requires only symptomatic care.

Prevention, and Control

Preventive measures: surgical débridement and prophylactic antibiotics.

C. tetani

Physiology and Structure

Small, motile;

Spore-forming (drumstick appearance);

Extremely sensitive to oxygen toxicity.

Pathogenesis and Immunity

Tetanospasmin is responsible for clinical manifestations of tetanus.

An A-B toxin, released when the bacteria lyse.

Subunit A is a zinc endopeptidase that acts on CNS: Inhibits release of an inhibitory mediator (e.g., GABA or glycine) which acts on postsynaptic spinal neurons (causing spastic paralysis).

C. tetani

Pathogenesis and Immunity

Contamination of devitalized tissue (wound, burn, injury, umbilical stump, surgical suture) with the spores → germination of the spores → release of tetanospasmin → the toxin reaches CNS by retrograde axonal transport or via the bloodstream → the toxin is fixed to gangliosides in spinal cord or brainstem and exerts its actions.

Germination of the spore and production of toxin are aided by conditions that lead to low oxidation-reduction potential:

Necrotic tissue;

calcium salts;

associated pyogenic infections.

C. tetani

Clinical Diseases

Generalized tetanus

Incubation period: 4-5 days.

Symptoms: convulsive tonic contraction of voluntary muscles. Spasms involve first the area of injury, then the muscles of the jaw (trismus or lockjaw; risus sardonicus). Other voluntary muscles become involved gradually, resulting in generalized tonic spasms (opisthotonos). Death usually results from interference with respiration. The mortality rate of generalized tetanus: ~50%. In more severe cases, the autonomic nervous systems are also involved.

Localized tetanus (confined to the musculature of primary site of infection)

Cephalic tetanus (site of infection: head)

Neonatal tetanus (infection of the umbilical wound): mortality > 90%, and developmental defects are present in survivors.

C. tetani

Laboratory Diagnosis

Diagnosis depends on the clinical picture and a history of injury.

Proof of isolation of *C. tetani* from contaminated wounds depends on production of toxin and its neutralization by specific antitoxin.

Treatment, Prevention, and Control

Prevention is much more important than treatment:

1. Active immunization with toxoid.

'Booster shot' for previously immunized individuals. This may be accompanied by antitoxin injected into a different area of the body.

2. Proper care of wounds.
   Surgical débridement to remove the necrotic tissue.

3. Prophylactic use of antitoxin.

4. Antibiotic treatment.
   Metronidazole

*Patients with symptoms of tetanus should receive muscle relaxants, sedation and assisted ventilation.

C. tetani

Control

Active immunization with tetanus toxoid (toxin detoxified with formalin)

Aluminum salt-adsorbed toxoid

DPT vaccine

Course of immunization: as mentioned in *C. diphtheriae*.

Narcotics addicts are a high-risk group.

C. botulinum

Physiology and Structure

This species is a heterogeneous group of fastidious, spore-forming, anaerobic bacilli.

Produce at least eight (A-H) antigenically distinct toxins (botulinum toxins; Botox).

- Liberated during the growth and during autolysis of the bacteria.
- A-B toxins. The subunit A is a zinc endopeptidase blocking release of acetylcholine at peripheral cholinergic synapses.
- Destroyed by heating for 20 min. at 100 ^{o}C.
- Types A, B, E, and F are most commonly associated with human illness. These are composed of a neurotoxic protein with a lethal dose of 1-2 mg.

C. botulinum

Pathogenicity and Immunity

An intoxication.

Ingestion of food (spiced, smoked, vacuum-packed, or canned alkaline foods) in which *C. botulinum* has grown and produced toxin → the toxin acts on both the voluntary and autonomic nervous systems at synapses and neuromuscular junctions → flaccid paralysis.

C. botulinum

Clinical Diseases

Foodborne botulism

Incubation period: 18-24 hrs.

Symptoms: double vision, inability to swallow, speech difficulty, bulbar paralysis, constipation, and abdominal pain. Bilateral descending weakness of peripheral muscle. Death occurs from respiratory paralysis or cardiac arrest. No fever. Mortality is high.

Recovery may need months to years.

Patients who recover do not develop antitoxin.

C. botulinum

Clinical Diseases

Infant botulism

Occurs in the first month of life. Weakness, signs of paralysis, *C. botulinum* and its toxin are found in feces. May be caused by ingestion of the bacteria or spores which grow in the gut and produce toxin.

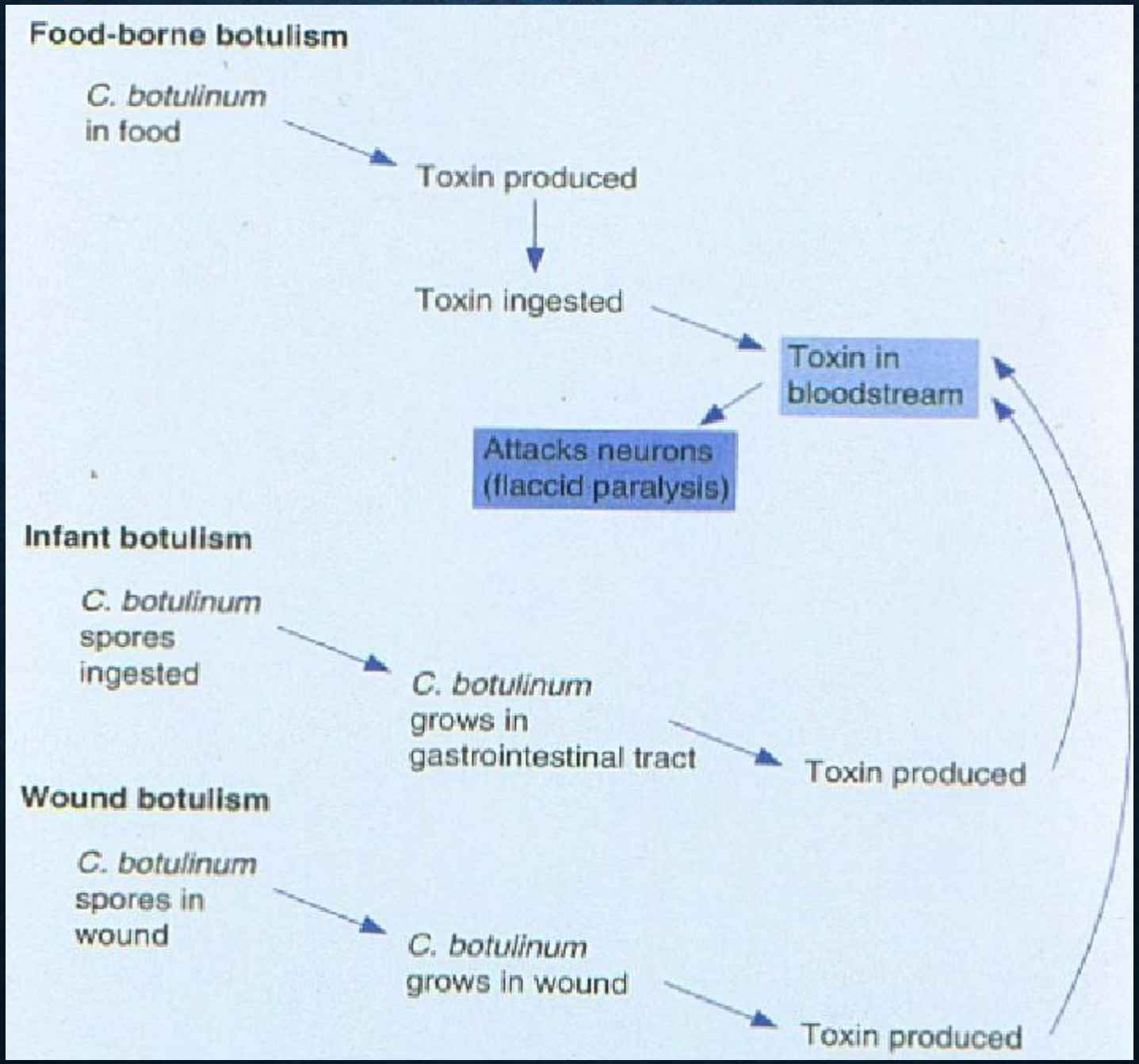
Feeding of honey has been implicated as a possible cause.

Patients recover with supportive therapy alone.

Wound botulism

Develops from contaminated wounds.

Symptoms similar to those of food borne botulism with longer incubation time. Less GI symptoms.


Food-borne botulism
C. botulinum in food
Toxin produced
Toxin ingested
Toxin in bloodstream
Attacks neurons (flaccid paralysis)
Infant botulism
C. botulinum spores ingested
C. botulinum grows in gastrointestinal tract
Toxin produced
Wound botulism
C. botulinum spores in wound
C. botulinum grows in wound
Toxin produced

C. botulinum

Laboratory Diagnosis

Culture of *C. botulinum* in patient feces and implicated food.

Detection of toxin in feces or serum from the patient and in leftover food: i.p. injection of mice → die rapidly. Toxin may also be detected by other serological tests.

Typing of toxin is done by neutralization with specific antitoxin in mice.

Treatment

Stomach lavage and high enemas.

Trivalent (A, B, E) antitoxin administered intravenously promptly.

Adequate ventilation by mechanical respirator.

C. botulinum

Prevention and Control

Spores of *C. botulinum* are widely distributed in soil and often contaminate vegetables, fruits etc.

Strict regulation of commercial canning has largely reduced the danger of widespread outbreaks. The chief danger lies in home-canned foods (vegetables, smoked fish or vacuum-packed fresh fish). The cans with toxic food may swell or may show innocuous appearance.

The risk from home-canned food can be reduced by boiling the food for 20 min.

Children younger than 1 year should not eat honey.

C. difficile

C. difficile is responsible for antibiotic-associated gastrointestinal disease ranging from self-limited diarrhea to severe, life threatening psudomembranous colitis.

It is a part of normal intestinal flora in a small number of healthy people and hospitalized patients. The spores can contaminate an environment for many months and can be a major source of nosocomial outbreaks.

This organism produces two toxins:

- Toxin A (an enterotoxin) induces release of cytokines, hypersecretion of fluid, and development of hemorrhagic necrosis.
- Toxin B (a cytotoxin) causes tissue damage.

C. difficile

Pseudomembranous colitis

Administration of antibiotics results in proliferation of drug-resistant *C. difficile*. Antibiotics that are most commonly associated with pseudomembranous colitis are ampicillin, cephalosporins, and clindamycin.

Disease occurs if the organism proliferates in the colon and produces toxins: watery or bloody diarrhea, abdominal cramps, leukocytosis and fever.

Laboratory diagnosis depends on isolation of *C. difficile* in the feces and detection of toxins with tissue culture cells (cytotoxicity assay).

The disease is treated by discontinuing the offending antibiotic, and orally giving either metronidazole or vancomycin in severe cases.

C. difficile

Antibiotic-associated diarrhea

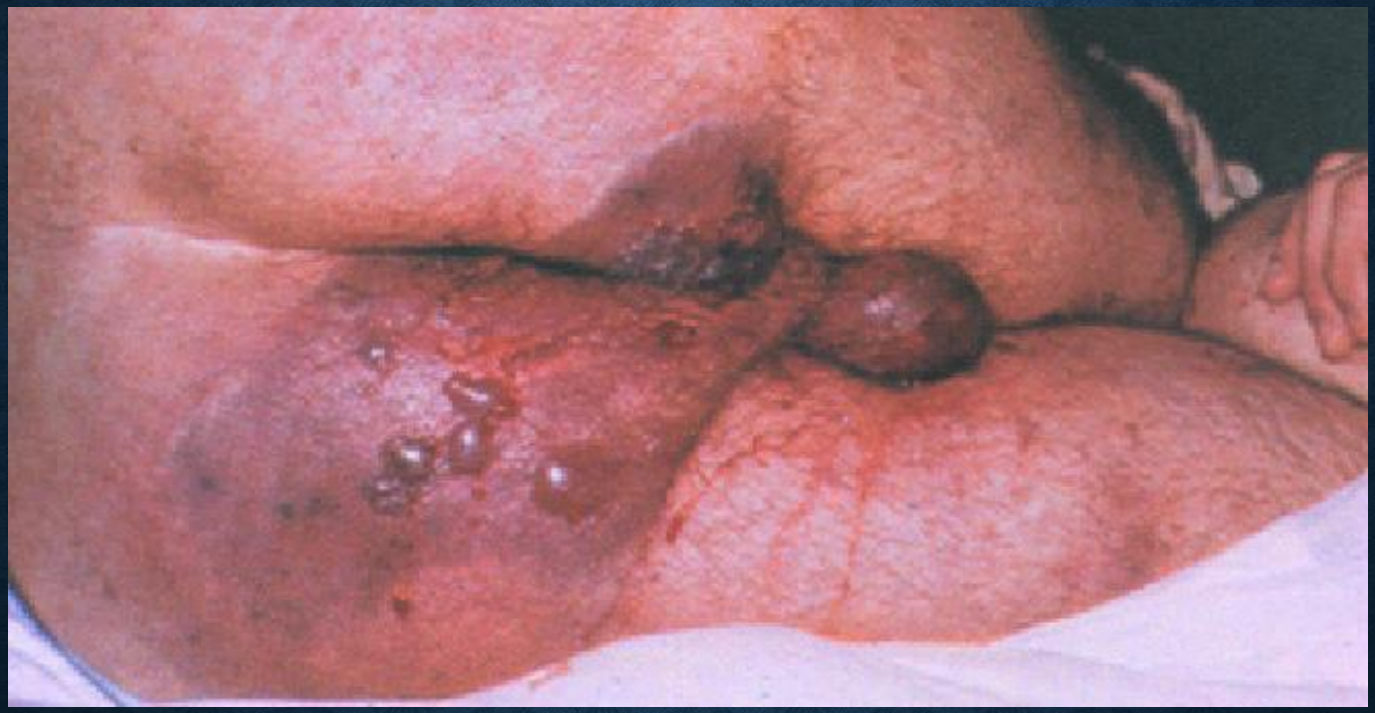
25% of cases are associated with *C. difficile*.

Mild to moderate diarrhea, less severe than the typical pseudomembranous colitis.

The role of the toxins is not well understood.

Other Clostridial Species

C. septicum is a cause of nontraumatic myonecrosis and frequently exists in patients with occult colon cancer, acute leukemia, and diabetes. It can spread from GI tract into tissue and result in fulminant infection with high mortality within 1 to 2 days.



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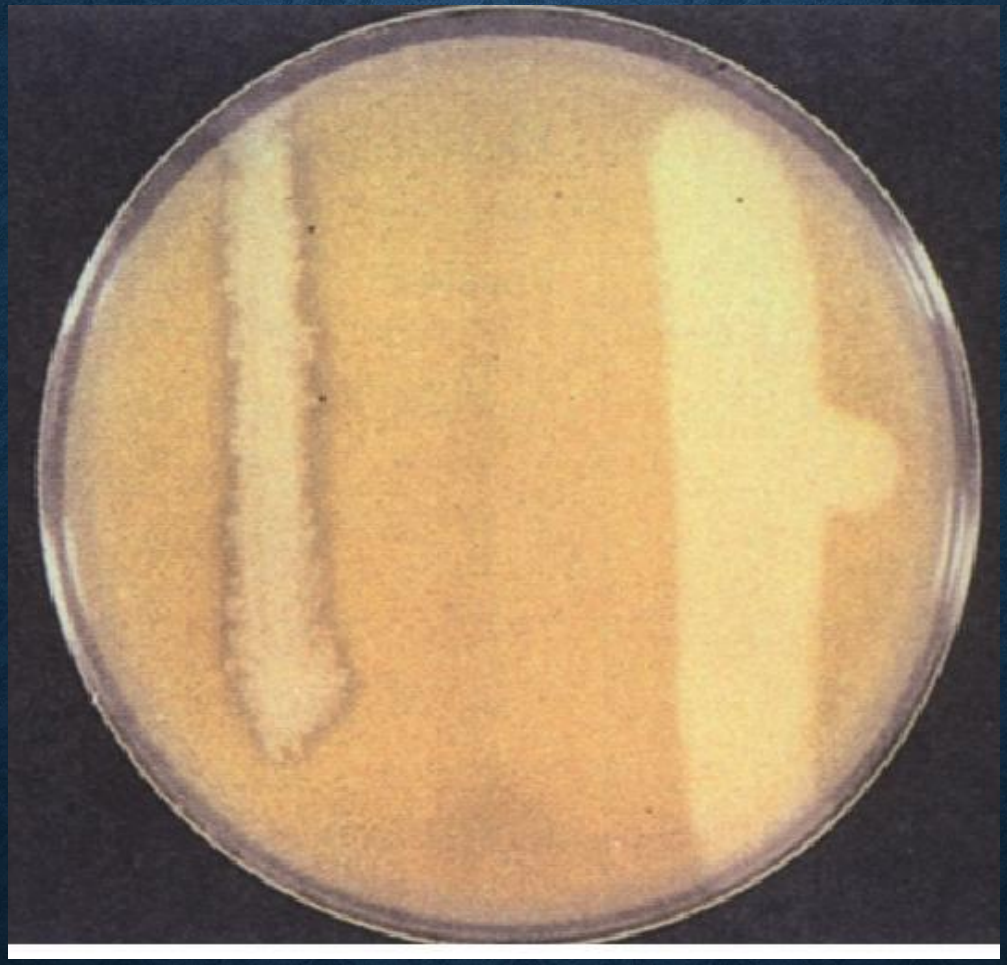

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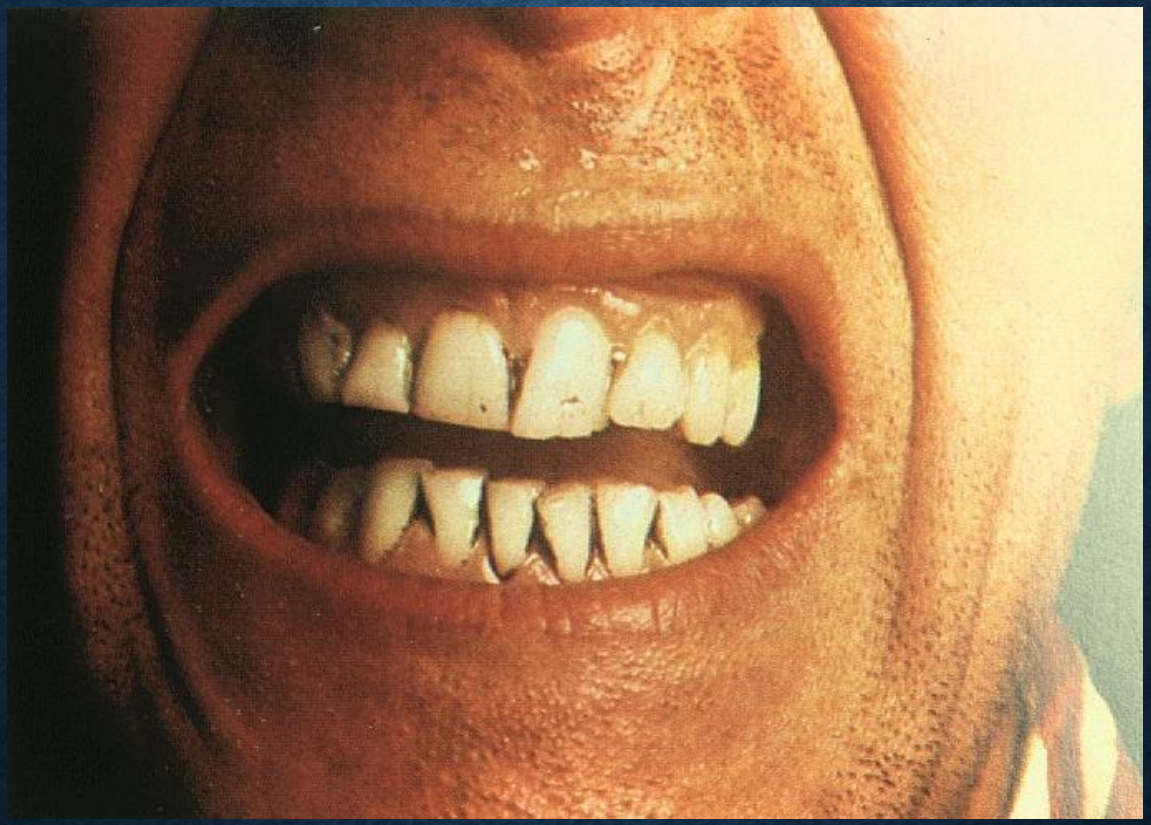
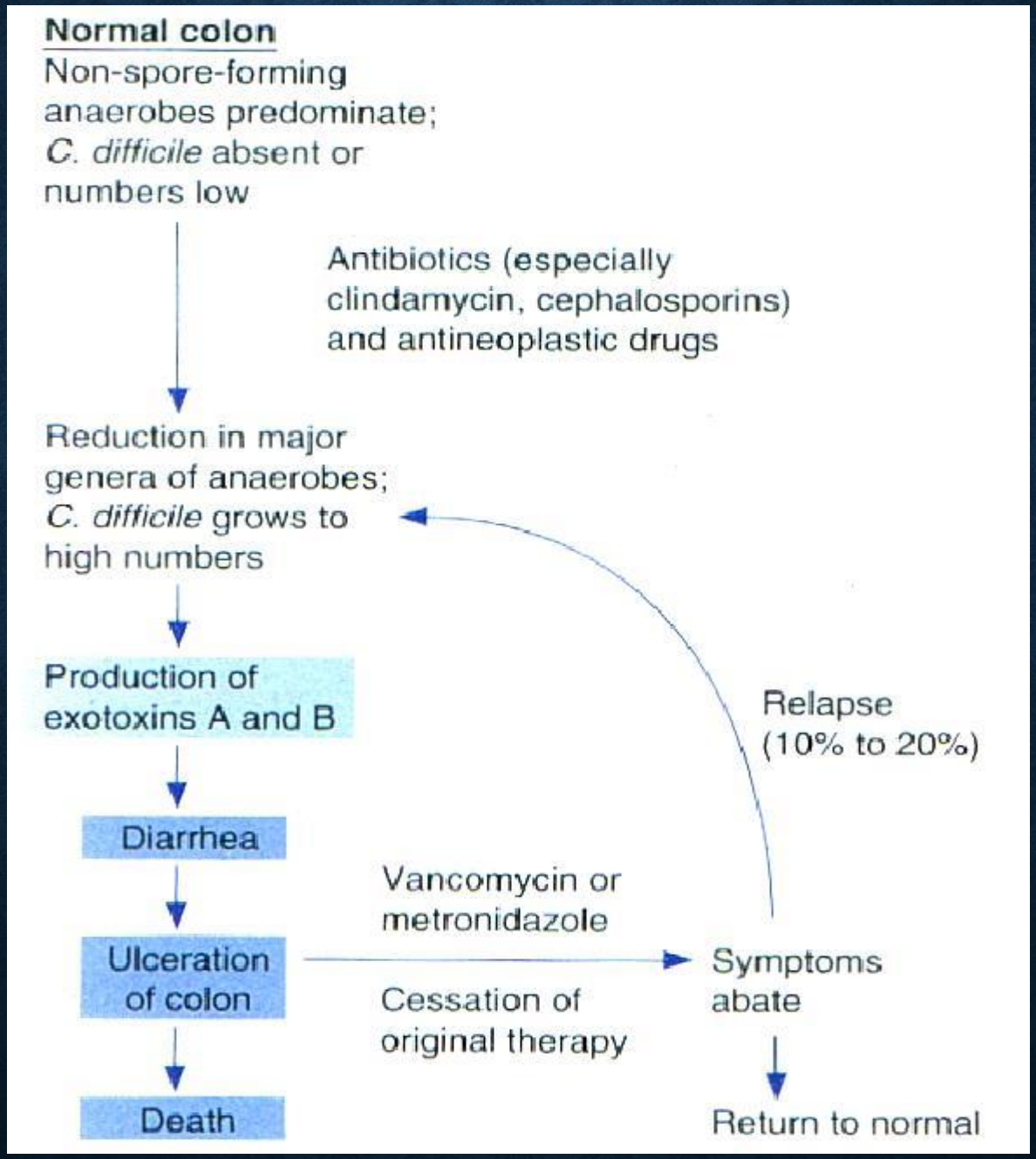
Nagler's reaction




Normal colon
Non-spore-forming anaerobes predominate; C. difficile absent or numbers low
Antibiotics (especially clindamycin, cephalosporins) and antineoplastic drugs
Reduction in major genera of anaerobes; C. difficile grows to high numbers
Production of exotoxins A and B
Diarrhea
Ulceration of colon
Death
Vancomycin or metronidazole
Cessation of original therapy
Symptoms abate
Return to normal
Relapse (10% to 20%)