GRAM POSITIVE BACILLI

Clinically important Gram positive bacilli

Spore forming

1. **Bacillus (O_2)**
2. **Clostridium ($\not{O}_2$)**

Non spore forming

1.Listeria

2. Erysipelothrix

3.Corynobacterium

4. Propionibacterium

Acid-fast bacilli

1.Mycobacterium

2.Nocardia

Non-acid-fast branching filamenous bacilli

1.Actinomyces

Gram-Positive Spore-Forming Bacilli

- Gram positive, motile, rod shaped

-Bacillus

-Clostridium

-Sporolactobacillus

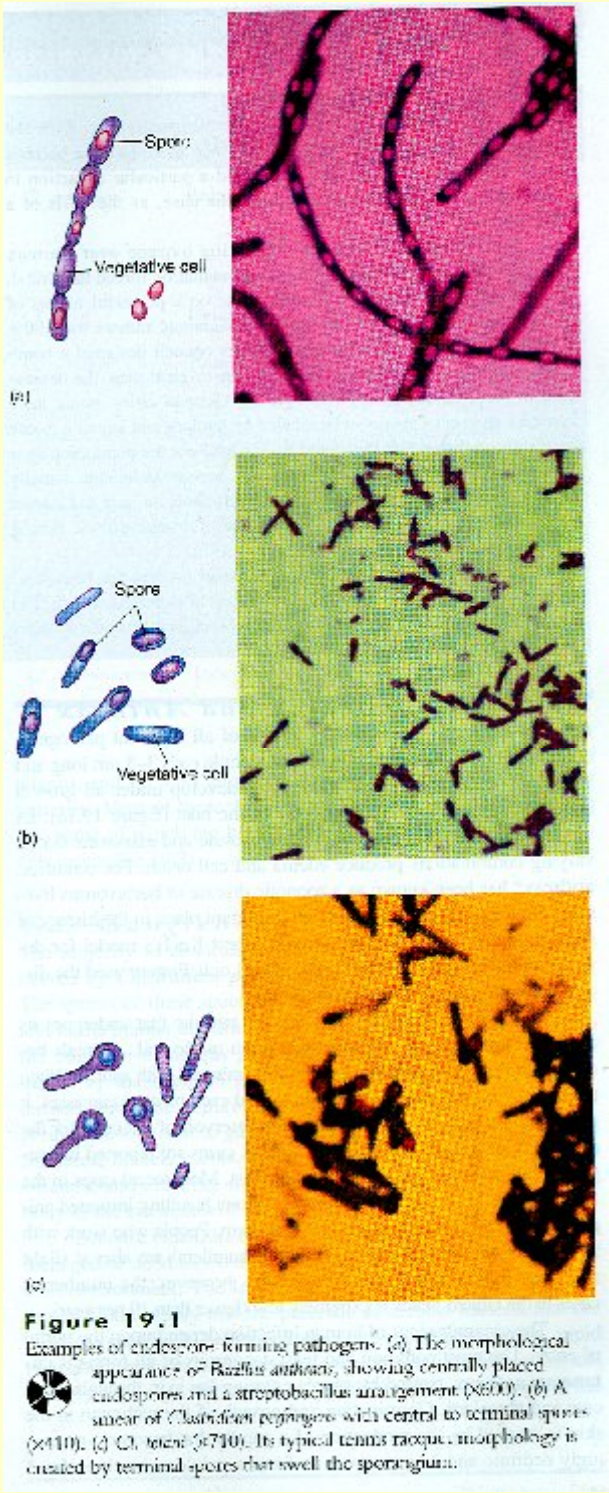


Figure 19.1

Examples of endospore-forming pathogens. (*a*) The morphological appearance of *Bacillus anthracis*, showing centrally placed endospores and a streptobacillus arrangement (×600). (*b*) A smear of *Clostridium perfringens* with central to terminal spores (×410). (*c*) *Cl. tetani* (×710). Its typical tennis racquet morphology is created by terminal spores that swell the sporangium.

GENUS BACILLUS

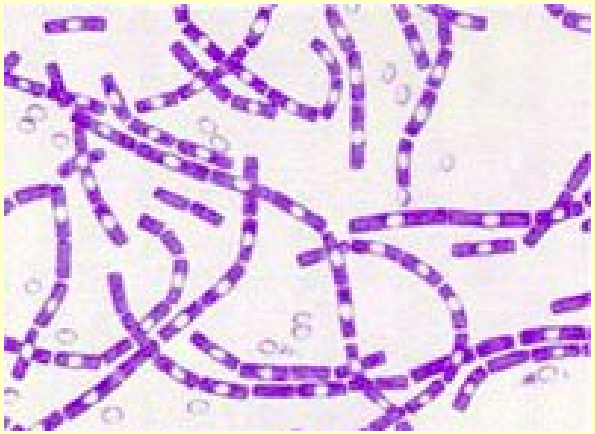
- Aerobic
- Catalase positive
- Not fastidious

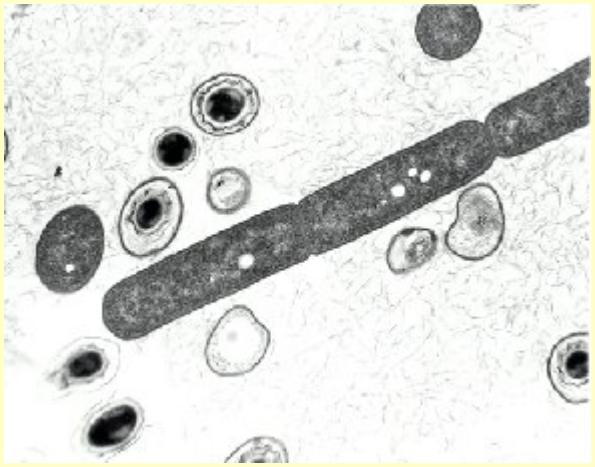
1.BACILLUS

- ***Bacillus anthracis***
  - Human pathogen
  - Isolation also considered to be clinically significant
  - Zoonosis
- ***Bacillus cereus***
  - Environmental organism
  - Contaminates food
  - Common cause of food poisoning
- ***Bacillus stearothermophilus***
  - Tolerates very high temperatures
  - Used for quality control of autoclaves

a.*Bacillus anthracis*

- **Large bacilli of 3-5 μm**
- **Single or paired in clinical isolates**
- **Polypeptide capsule and exotoxins**
- **Highly resistant central spores**

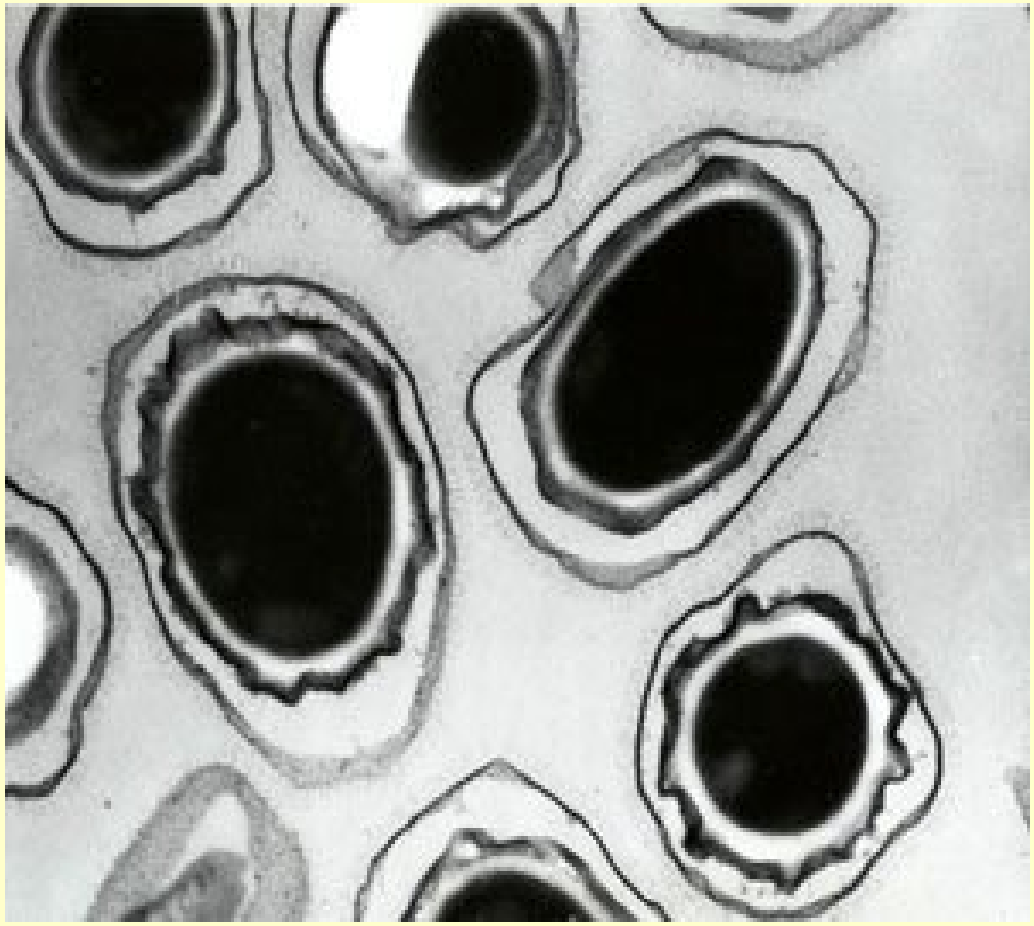




Anthrax

- Spore of B.anthracis
- Duration of life

(> 60 years) soil



Anthrax

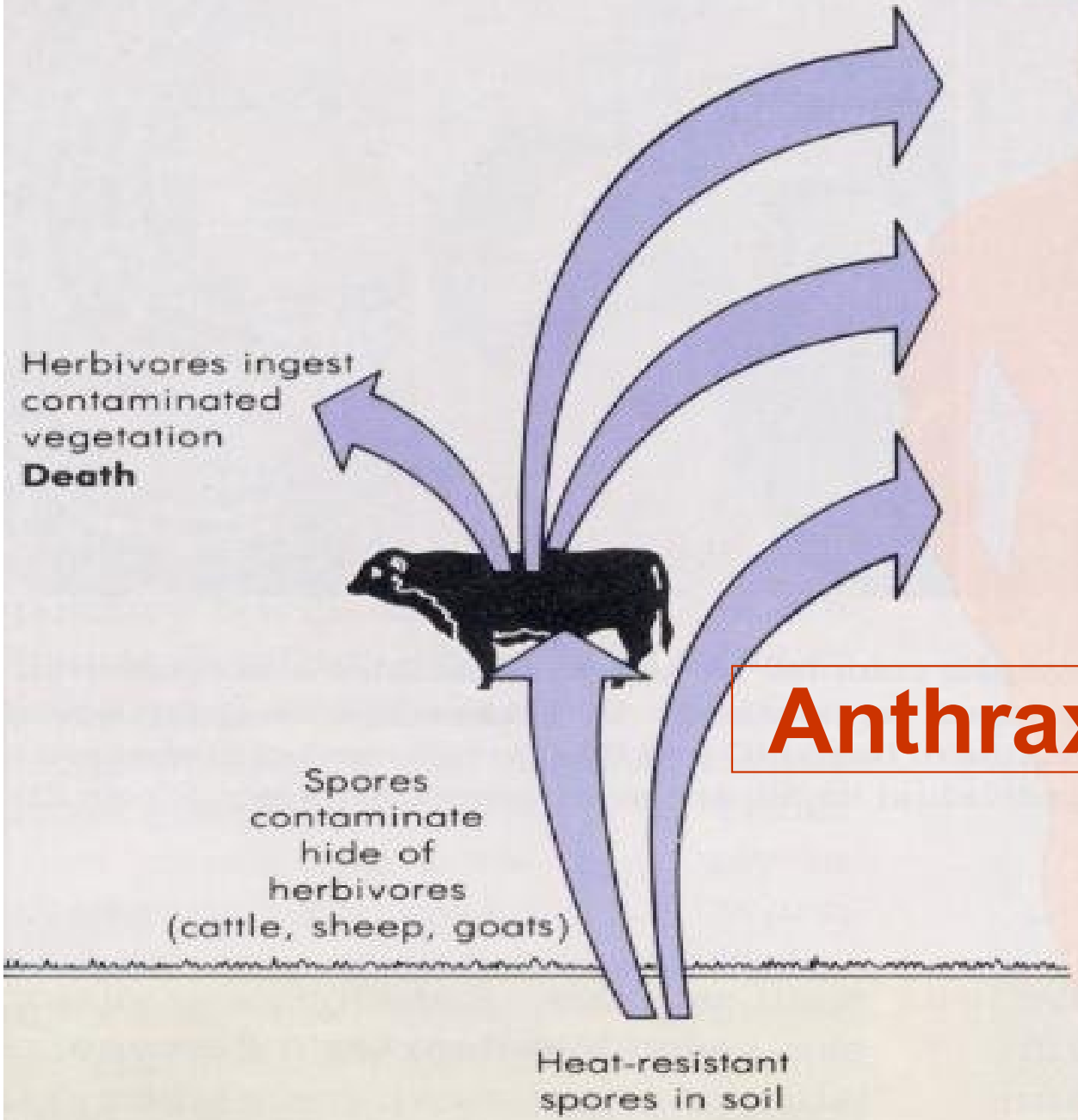
- Culture on rich media

Anthrax

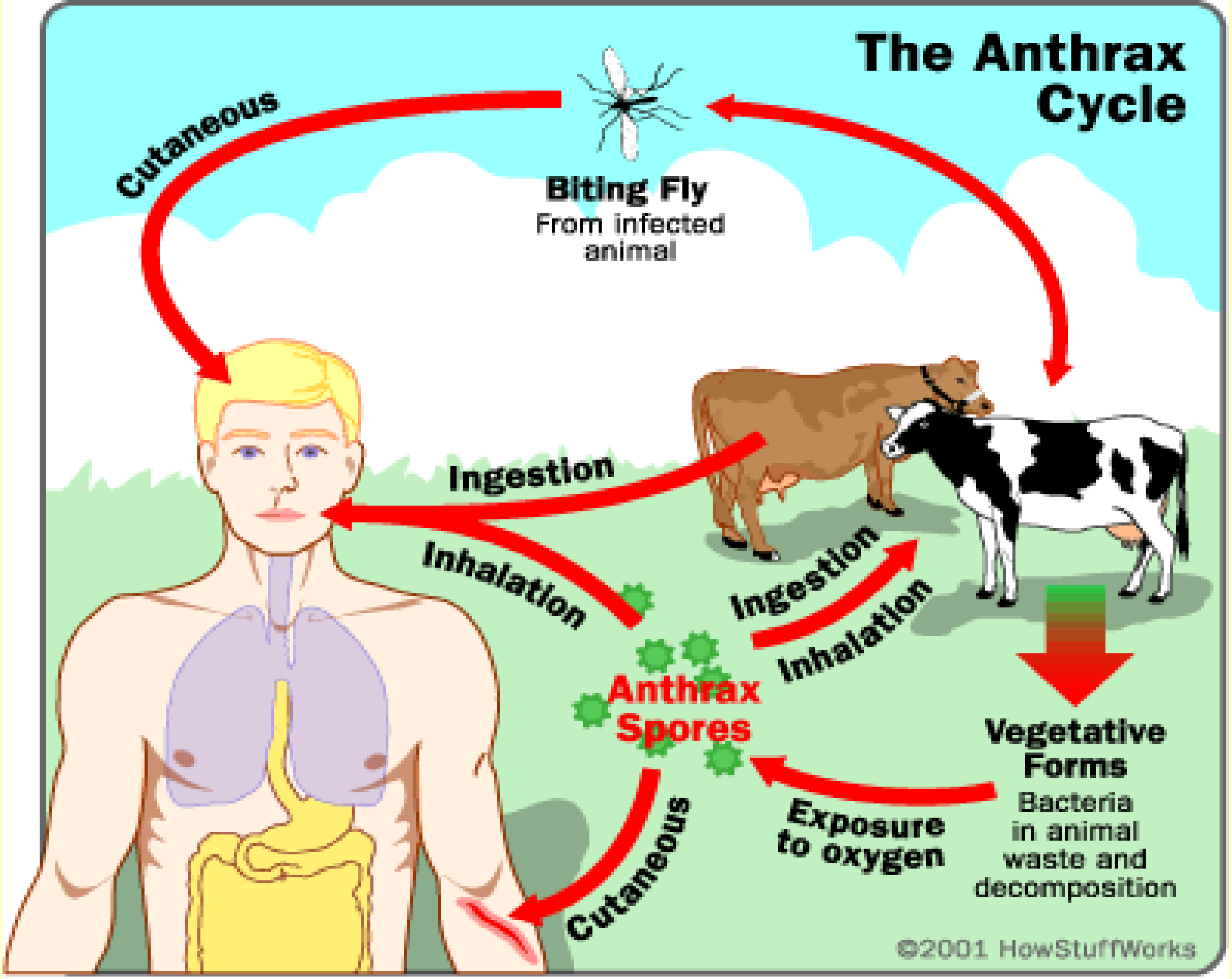
- Zoonotic disease of herbivorous livestock

Anthrax - Epidemiology

Primary hosts	Accidental host	
	Method of infection	Mortality
	*Contact with contaminated animal products (hides, wool, hair)	20%
	Ingestion of contaminated food products	95%
	Inhalation of contaminated dust (Woolsorter's disease)	95%



*Contact infections are generally on exposed skin surfaces (e.g., arms) and result in 95% of infections


The Anthrax Cycle
Cutaneous
Biting Fly
From infected animal
Ingestion
Inhalation
Ingestion
Inhalation
Anthrax Spores
Cutaneous
Exposure to oxygen
Vegetative Forms
Bacteria in animal waste and decomposition
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Anthrax

Pathogenesis and clinical presentations

Cutaneous anthrax

About 20% mortality

Virulence factors

Capsule

(antiphagocytic)

Toxin

(oedema & death)

Inhalation anthrax

High mortality

Gastrointestinal anthrax

High mortality

Cutaneous anthrax

- « malignant pustule »
- Incubation 2-3 days

erythematous papule

- Increasingly necrotic → later ruptures to form

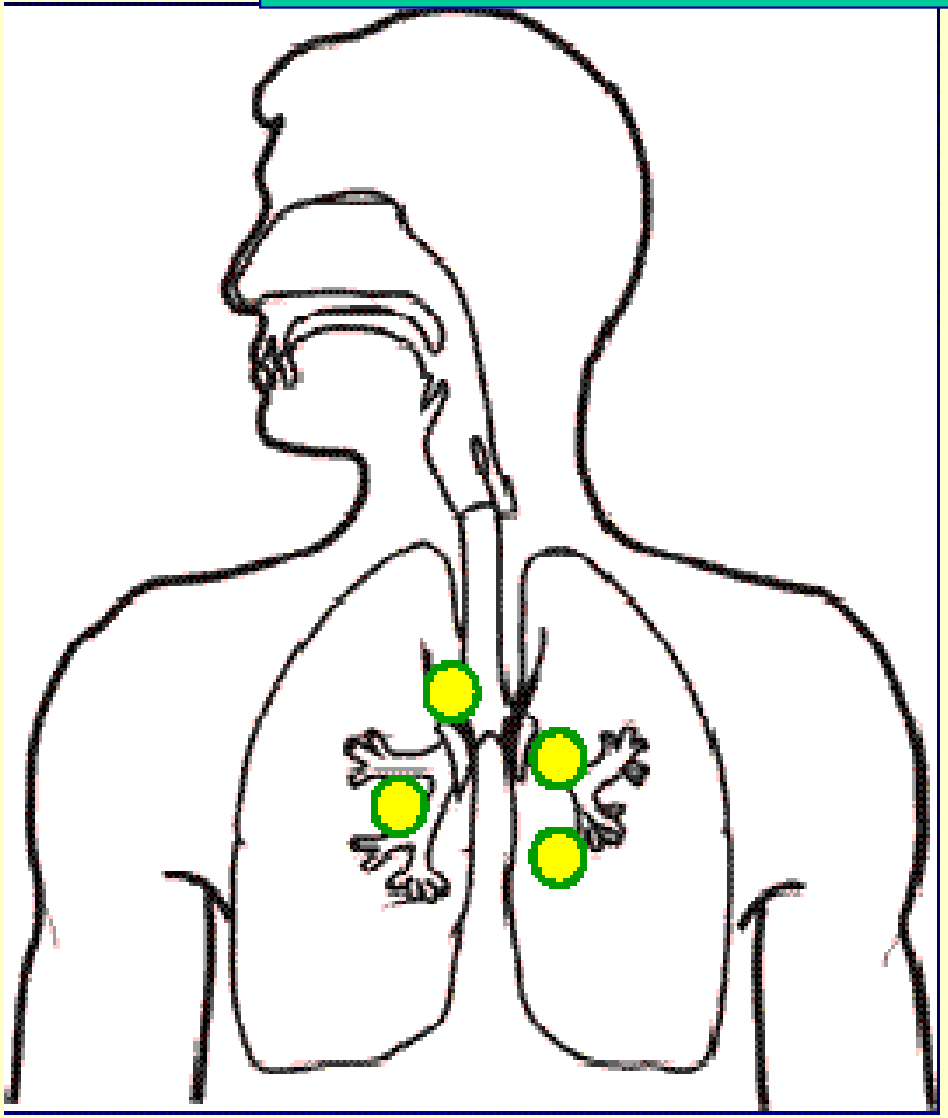
a painless black eschar

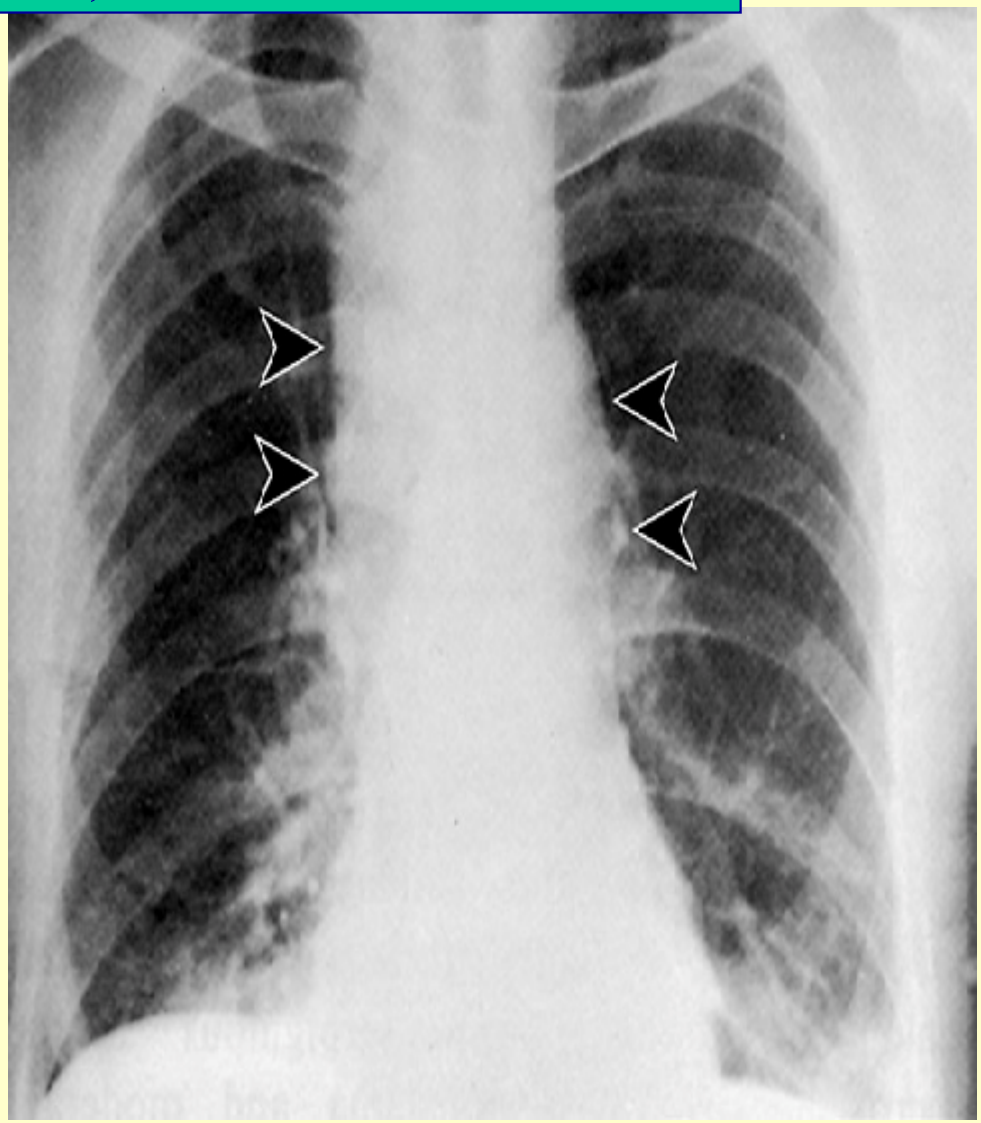
Gastro-intestinal anthrax

- Contaminated meat
- Asia, Africa
- Cardiovascular collapse within few hours and death

Pulmonary anthrax

Toxemia, capillary thrombosis, cardiovascular shock

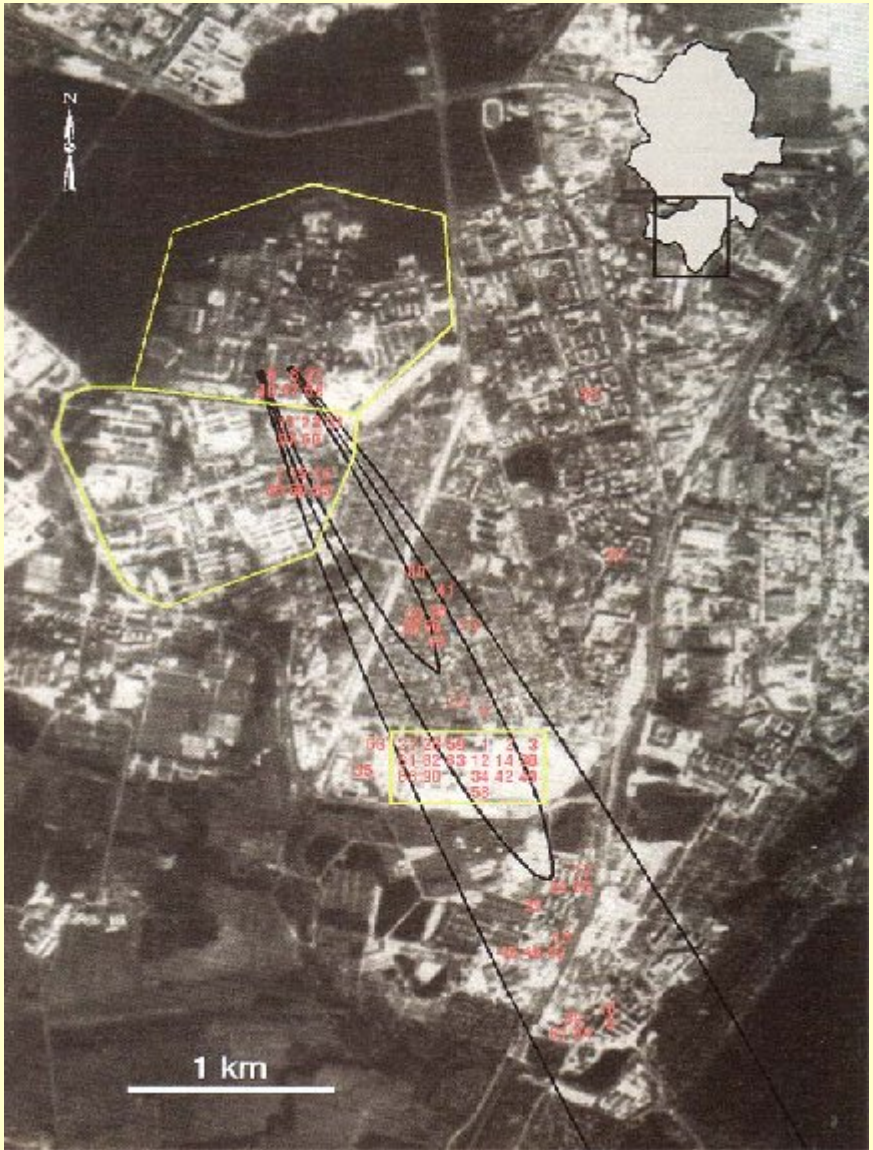




Sverdlovsk epidemy

- April 1979
- 1 200 000 habitants

Sverdlovsk epidemy



Sverdlovsk epidemy

Sverdlovsk epidemy

- **68 death « officials »**

Bioterrorisme
and Anthrax bacillus

Anthrax - Diagnosis

- Specimen
  - Aspirate or swab from cutaneous lesion
  - Blood culture
  - Sputum
- Laboratory investigation
  - Gram stain
  - Culture
  - Identification of isolate

Anthrax – treatment and prevention

- **Penicillin**
- **(Tetracycline /chloramphenicol)**
- Erythromycine,Clindamicine
- **Prevention**
  - **Vaccination of animal herds**
  - **Proper disposal of carcasses (burning or decontamination before burial)**
- **Active immunisation with live attenuated bacilli**


31ST MARINE EXPEDITIONARY UNIT
U.S.

b.*Bacillus cereus*

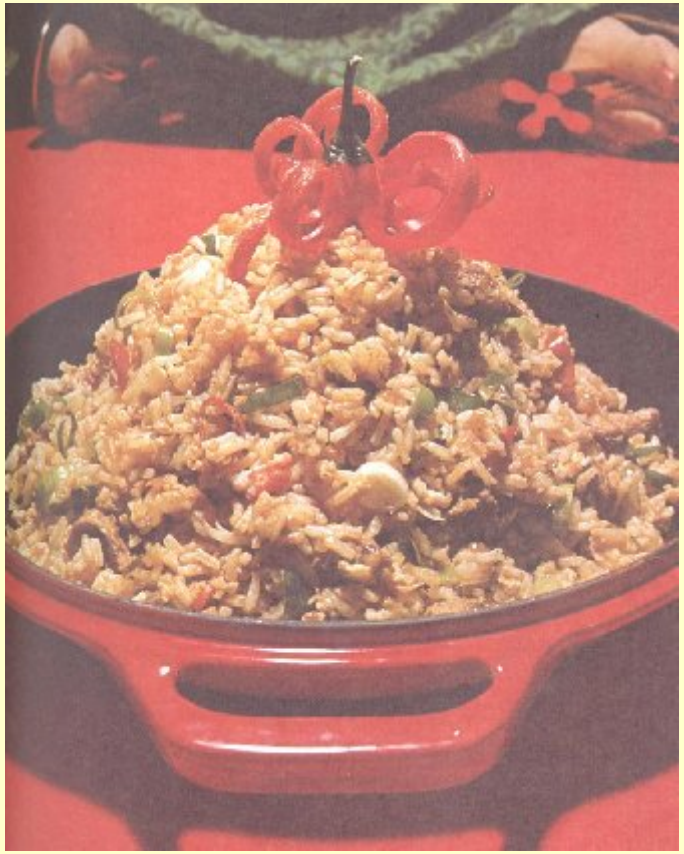
- Large, motile, saprophytic bacillus
- Heat resistant spores
- Airborne and dust-borne contaminants
- Pre formed heat and acid stable toxin (Emetic syndrome)
- Heat labile enterotoxin (Diarrhoeal disease)
- Multiply readily in cooked foods (Rice, potato and meat)
- Lab diagnosis – Demonstration of large number of bacilli in food

Bacillus cereus

clinical presentation

Gastroenteritis

EMETIC FORM



Incubation period < 6 hours
Severe vomiting
Lasts 8-10 hours

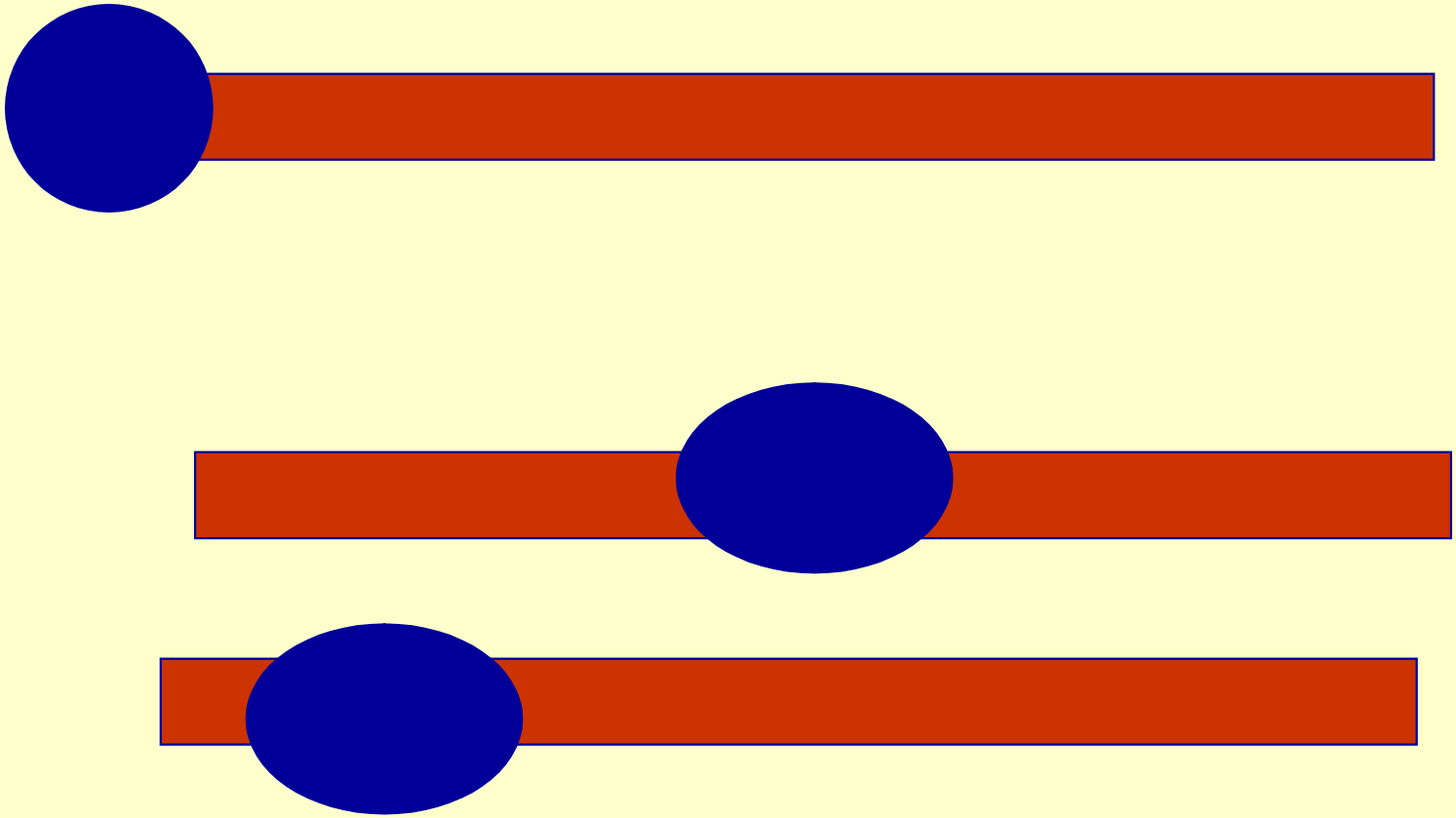
DIARRHOEAL FORM

Incubation period > 6 hours
Diarrhoea
Lasts 20-36 hours

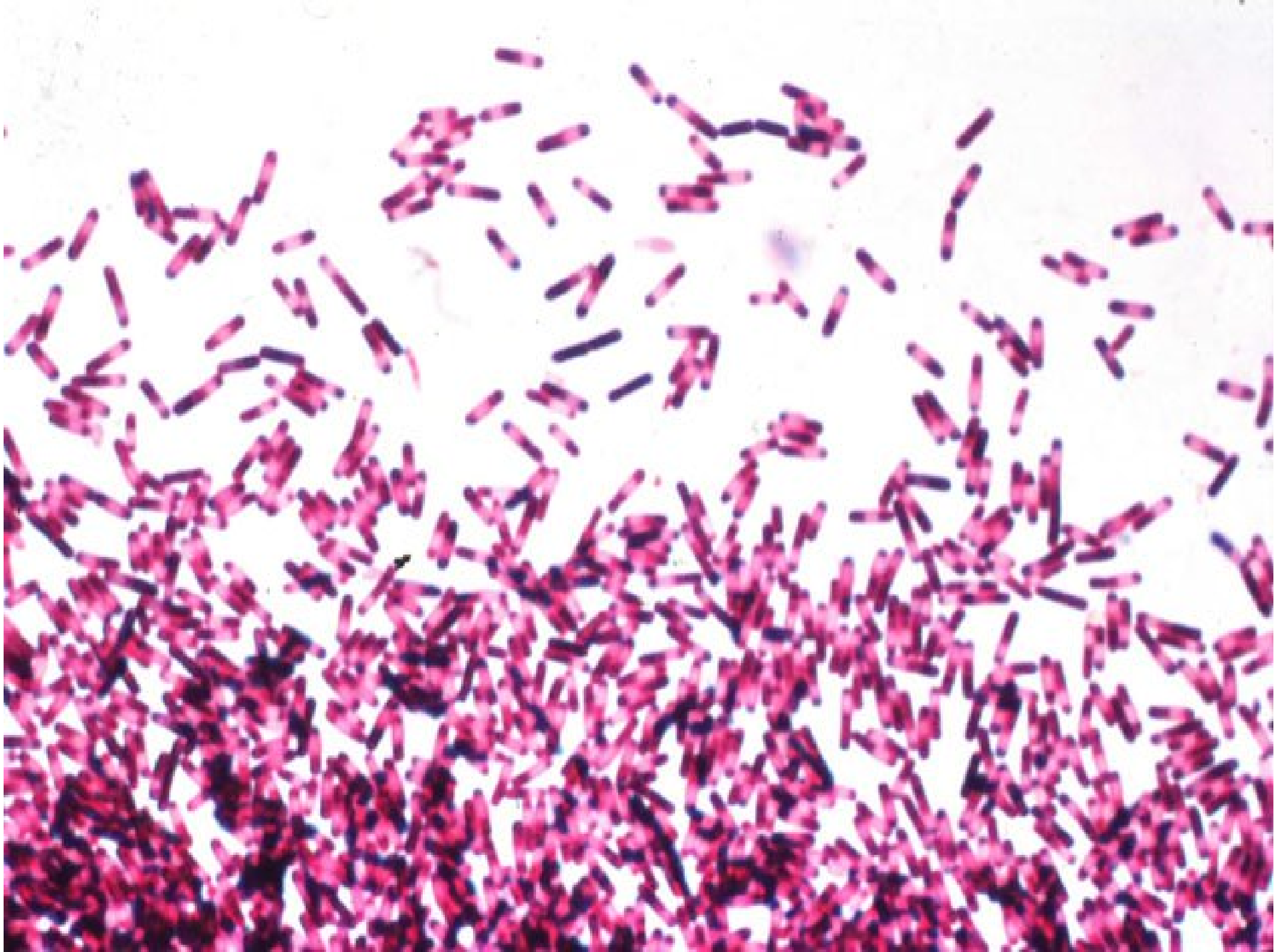
CLOSTRIDIUM (ANAROBES)

- **Anaerobic**
- **nonmotile**
- **Sporing**
- **Catalase negative**
- **Gram positive**
- **Diameter of the spore is larger than the cell resemble a spindle**
- **Clostridium is derived from Kloster meaning spindle**

- **Spores** Pleomrhic (elongated, spindle)



Most are obligate anaerobes produce neuro histo toxins



- Saprophytes - Most
- Some are opportunists - tetanus/gas gangrene/food poisoning
- *Cl. perfringens* - commensal of the intestine
- *Cl. sporogenes* -
  - Can invade the intestine after the death

CLASSIFICATION BASED ON THE TYPE OF DISEASE PRODUCED

- **A . Tetanus** *Cl. tetani* - Present in soil
- **B. Gas gangrene**
  - **Established** *Cl. perfringens* 'gut' organism
    Cl. septicum
    Cl. novyi
  - **Less pathogenic** *Cl. histolyticum*
    Cl. fallax
  - **Doubtful** *Cl. bifermentans*
    Cl. sporogenes

- **C. Food poisoning**
  - **1. Gastroenterritis - *Cl perfringens* Type A**
  - **2. Botulism - *Cl. botulinum/ Soil***
  - **3. Pig-bel *Cl. perfringens* type C**
- **D. Acute colitis - *Cl. difficile* / gut' organism (pseudomembranous colitis)**
  - Commonest cause of 'nosocomial' diarrhoea

Role of clostridia in infection and disease

- Wound and tissue infections
  - *Myonecrosis
  - *Antibiotic-associated colitis
  - *Tetanus
- Food intoxication of perfringens and botulism varieties.

Gas gangrene

- Anaerobic cellulitis or myonecrosis
- Predisposing factors:
  - surgical incisions
  - compound fractures
  - diabetic ulcers
  - septic abortions
  - puncture and gunshot wounds
  - crushing injuries

Lecithinase C (alpha toxin)

Dead tissue, blood clots, foreign matter aerobic organisms

↓

In an injury

DEVELOP ANAEROBIC CONDITION

↓

(*Exogenous infection*) Germination of spores

↓

Gas gangrene

↓

Rupture of RBC,oedema, necrosis, gas production, toxaemia, myositis

Crepitus

C Perfringens
C histolyticum
C septicum
C novyii

C Perfringens ⇨ **Alpha toxin (lecithinase)**

Collagenase
Hyaluronidase
DNAse

Clinical signs

- Pain
- Edema
- Bloody exudate in the lesion
- Fever
- Tachycardia
- Blackened necrotic tissue filled with bubbles of gas

Laboratory diagnosis

Good specimen ?/ Gram stain/ culture/ anaerobic incubation


Clostridium perfringens
Thioglycollate & ANABAP

Management

Provide an aerobic environment

↑

Surgical removal of all the dead and foreign matter

Antibiotics to cover **Clostridia** and other **contaminants**

Penicillin
Metronidazole
Aminoglycoside

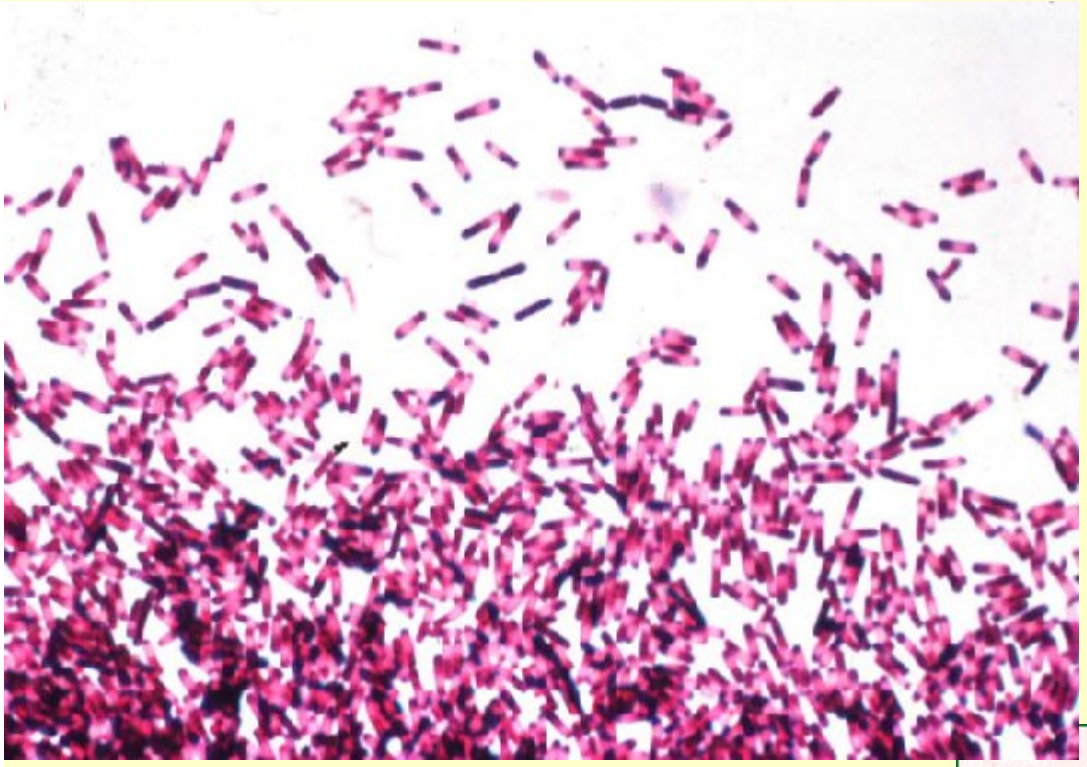
Or
Clindamycin

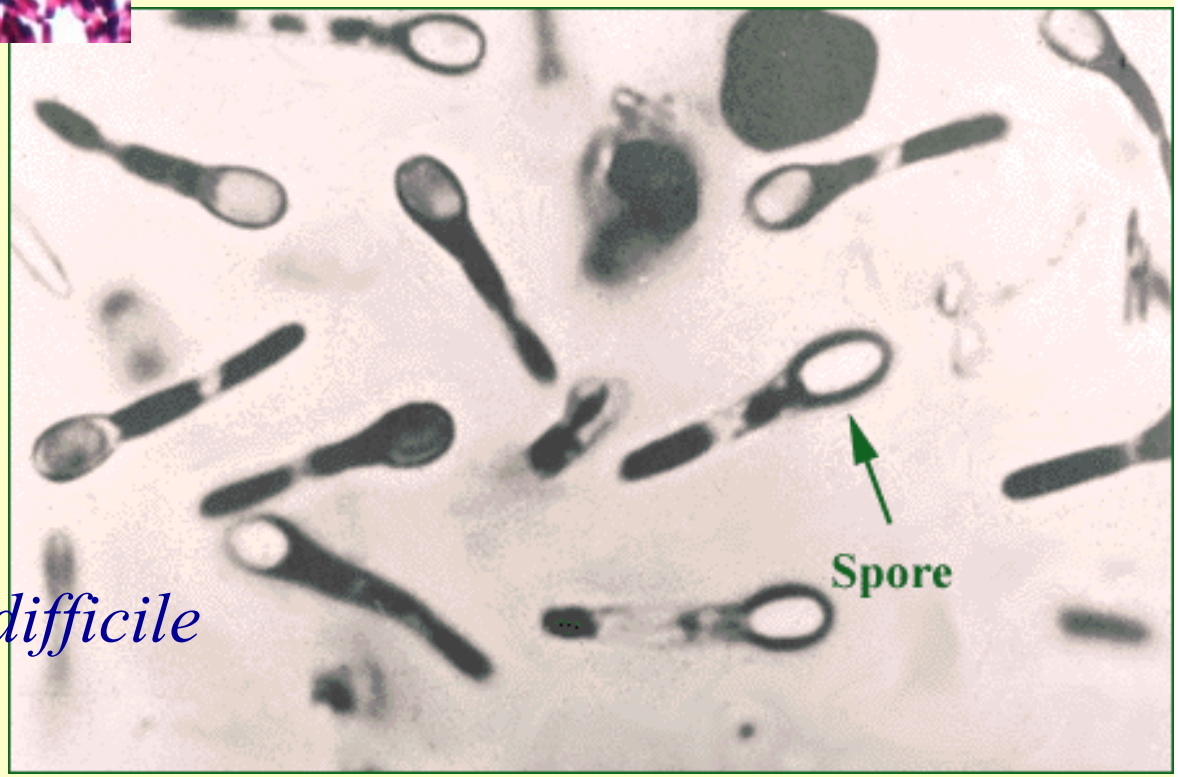
Or broad spectrum β lactum cefotaxime, imipenem

Used to give antitoxins to cover *C. perfringens*, *C. septicur* and *C. novyi*

Antibiotic-Associated Colitis

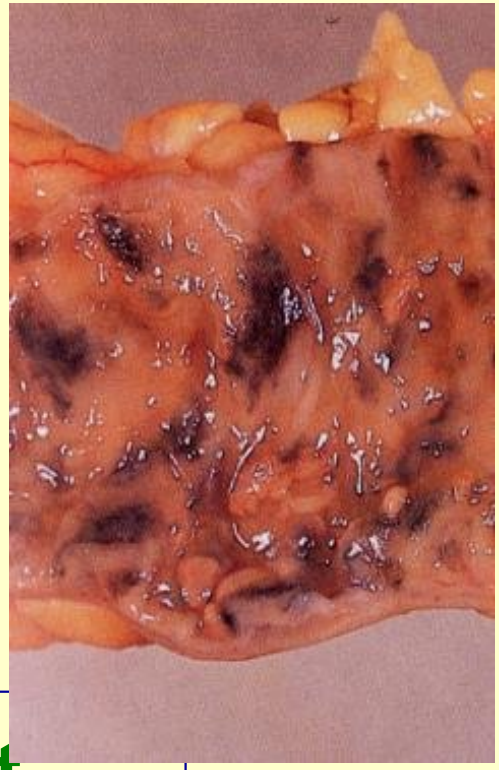
Clostridium difficile





C. difficile

PSEUDOMEMBRANOUS COLITIS



Virulence factors

Enterotoxin
(Toxin A)
Cytotoxin
(Toxin B)

Management

Discontinue antibiotics
Ampi/Tetra/Clinda
Oral metronidazole
Oral vancomycin

Diagnosis

Clinical suspicion
Culture of faeces
Detection of toxin

TETANUS (Lock-jaw)

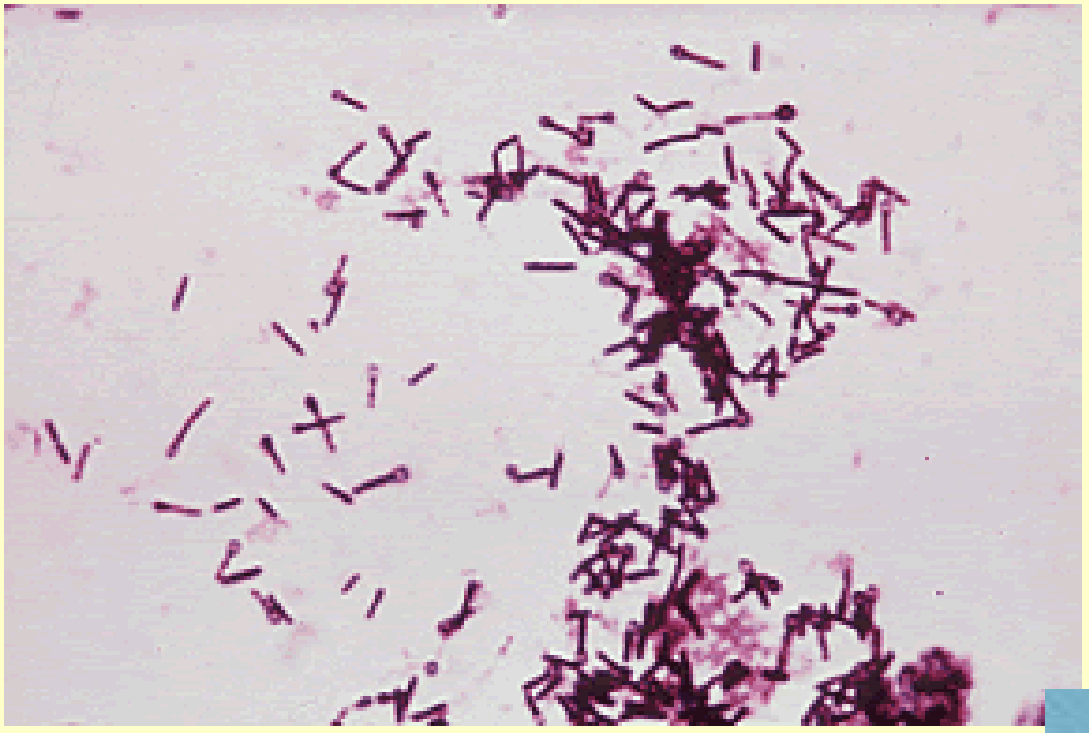
Cause tetanus in both man and animals disease which effect the nervous system of the host.

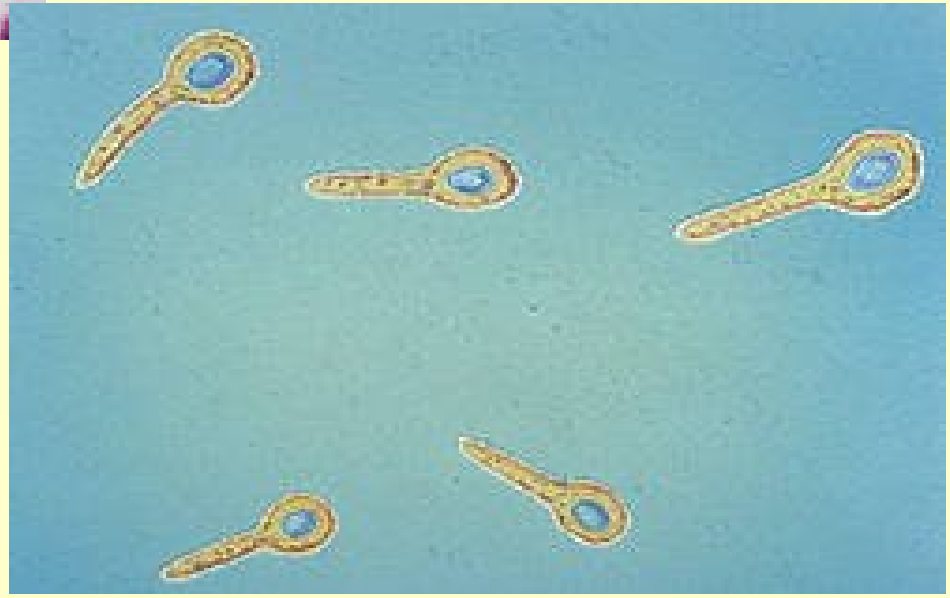
- Agricultural workers and gardeners and are more prone because the spores are present in the soil & intestine of animals.
- At birth under unhygienic conditions baby's can get – **tetanus neonatorum**.

Clostridium tetani

- Neuromuscular disease
- Entrance of spores through accidental puncture wounds
  - burns
  - Umbilical stumps
  - Frostbite
  - Crushed body parts

- Soil/Intestine/Vagina
- Drum stick appearance
- Motile with peritrichous flagella
- Obligatory anaerobes
- Grow on Robertson’s cooked medium





Clostridium tetani


D-113

– **What happens**

- Toxin acts at the synaptic junction – prevent the synthesis of acetylcholine. Thus, prevents synaptic transmission.

Toxins

- **Tetanolysin** - heat and oxygen labile/lyse RBC/
- **Tetanospasmin** - heat and oxygen stable/highly lethal (for mice 0.0000001 mg) dies within 1 - 2 days

get easily neutralize with antitoxin

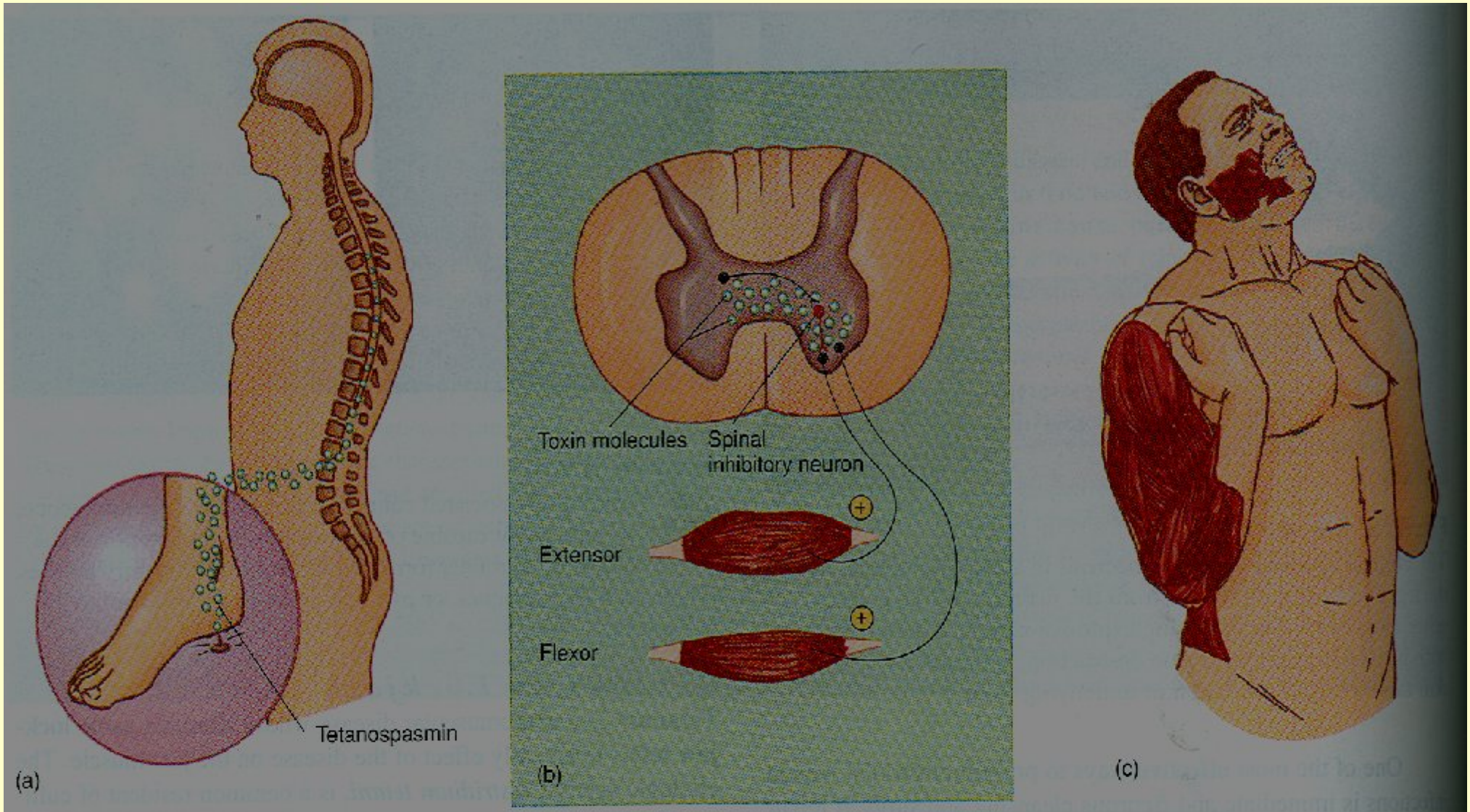
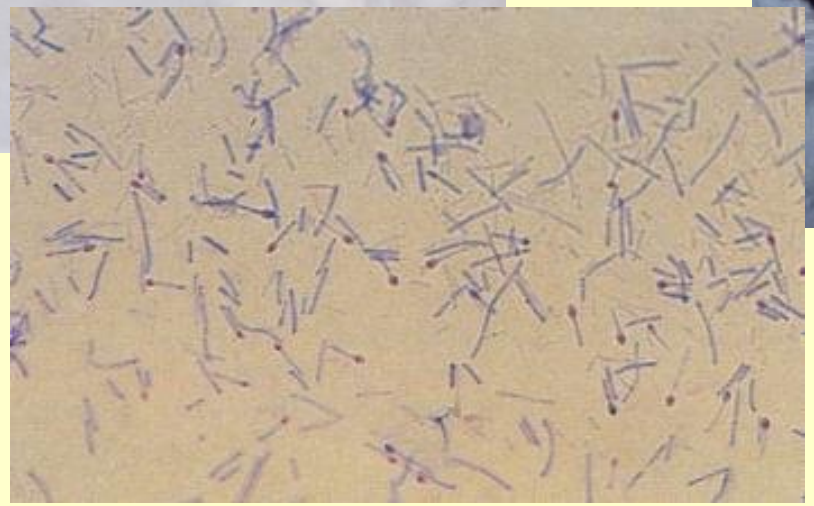
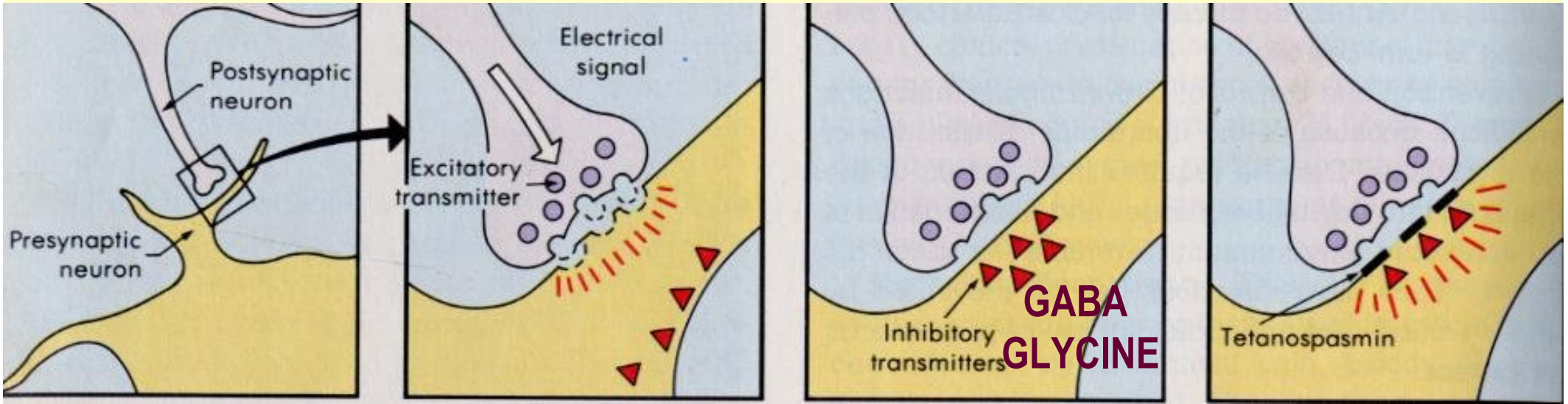


Figure 19.6

The events in tetanus. (*a*) Following traumatic injury, bacilli infecting the local tissues secrete tetanospasmin, which is absorbed by the peripheral axons and is carried to the target neurons in the spinal column. (*b*) In the spinal cord, the toxin attaches to the junctions of regulatory neurons that inhibit inappropriate contraction. Released from inhibition, the muscles receive constant stimuli and contract uncontrollably, even opposing members of a muscle group. (*c*) Muscles contract spasmodically, without regard to regulatory mechanisms or conscious control. Note the clenched jaw typical of risus sardonicus.




Postsynaptic neuron
Presynaptic neuron
Electrical signal
Excitatory transmitter
Inhibitory transmitters
GABA
GLYCINE
Tetanospasmin

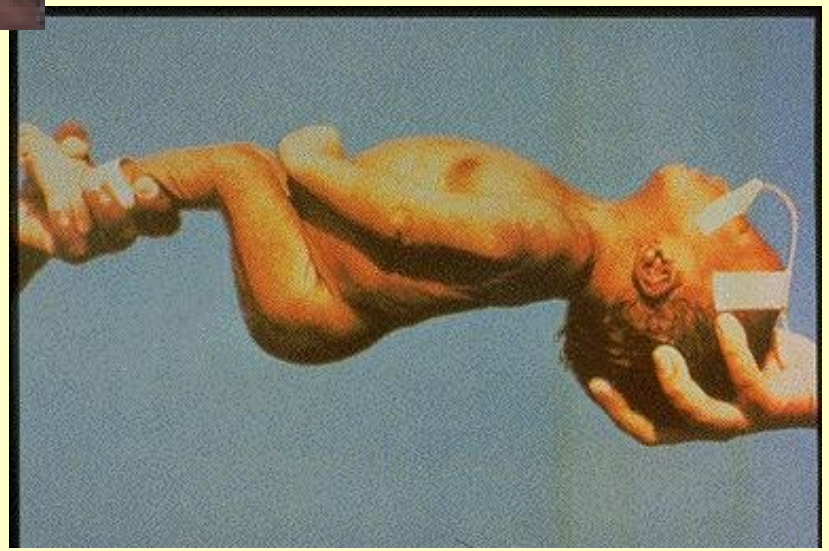
- **Susceptibility** -

  Some strains can withstand boiling for 3hrs/dry heat 160^0C for 1hr. but all will destroy at 121^0C/15 min.

- Spores germinate -------toxin-----motor nerve endings--------along the motor neurones of the peripheral nerve to the anterior horn cells-----***local tetanus*** (in the proximity of the wound).
- ***Ascending tetanus*** – when toxins spreads upwards along the spinal cord towards C.N.S. Gives generalized spasms.
- ***Descending tetanus*** – when toxin is given IV , spasms will appear in the muscles of the head, neck and spreads downwards.

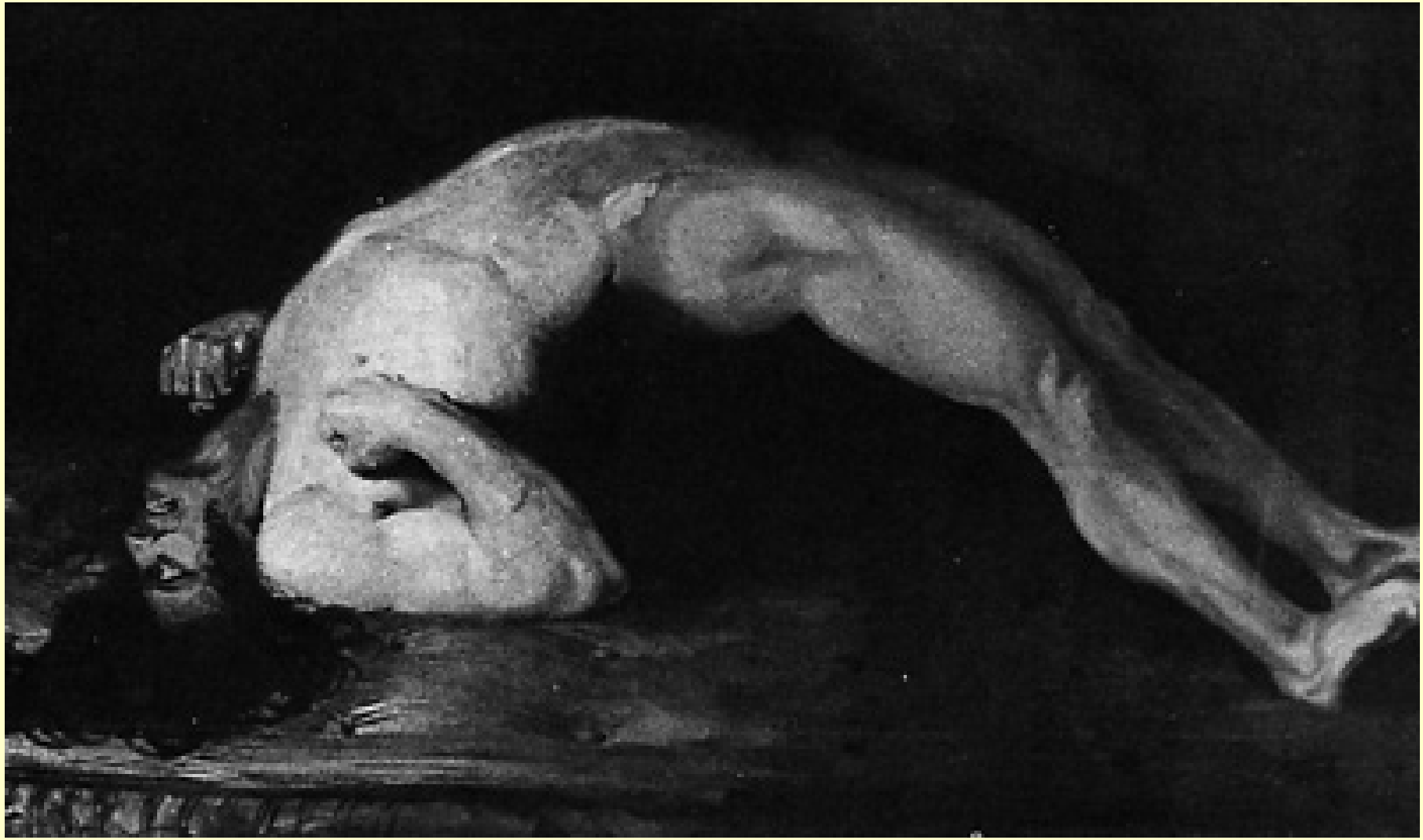
- **<u>Clinical symptoms</u>**
- Incubation period: 4-10 days
- Early symptom is **trismus** (lock jaw) – spasms of the masseter muscle
  - difficulty in opening of the mouth and masticating
  - rigidity spreads to muscles of the face, neck and truck

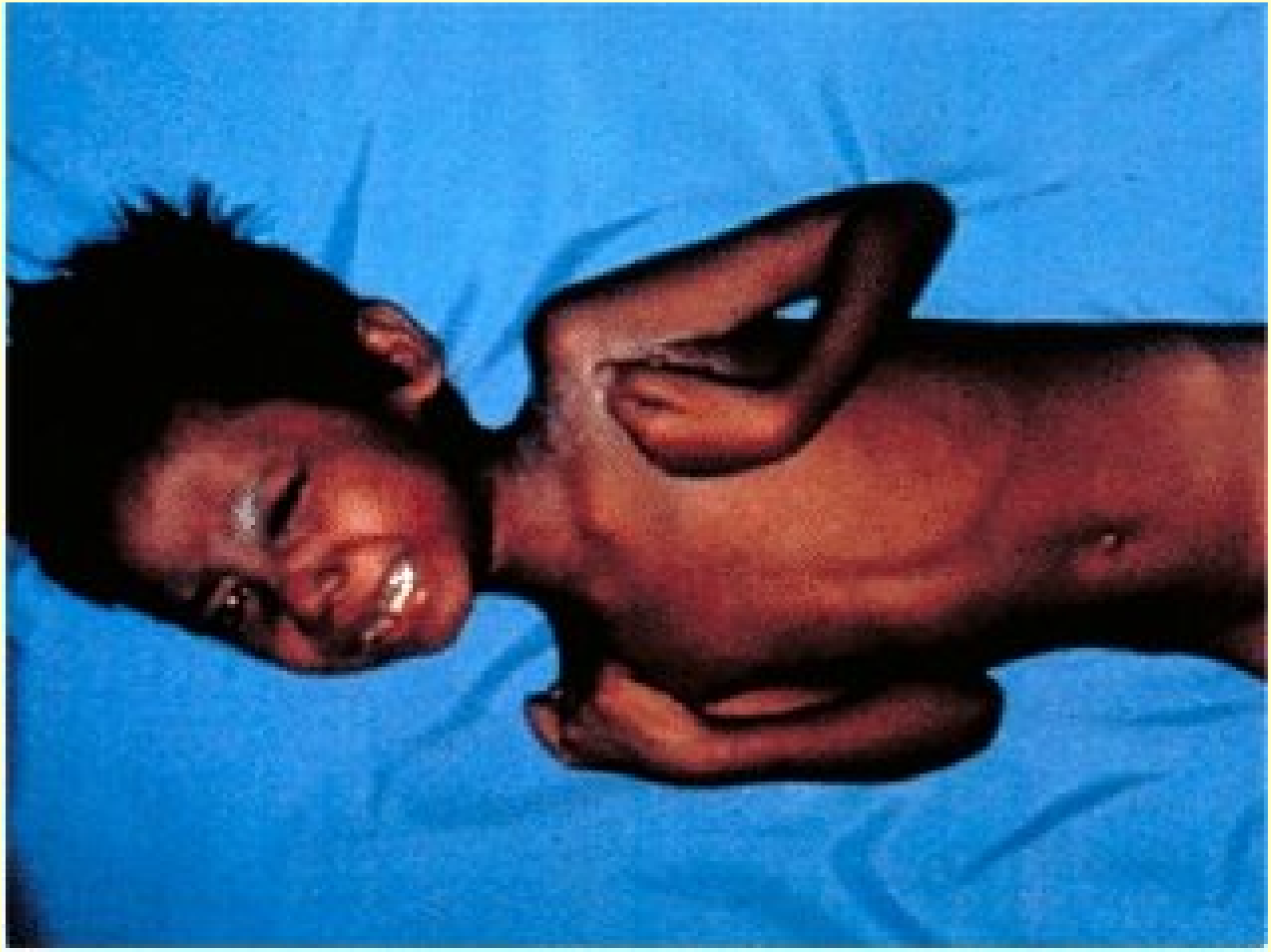
- **risus sardonicus** –The semblance of a grin caused by facial spasm especially in tetanus at the angle of the mouth
- back is usually slightly curved (Opisthonotus ?)
- In severe cases violent spasms will last for few seconds to 3-4 mins.
- If convulsions appear soon after the initial symptoms, it is very serious.
- The spasms gradually intensify and patient may die of
  - .Paralysis of respiratory muscles and respiratory collapse
- - fatality rate is 10-70%



Trismus

Opisthotonos





Clostridium tetani

Clinical diagnosis

- Treatment:
  - Symptomatic treatment

    cleansing and removing the afflicted tissue,

    Penicillin or tetracycline

    Muscle relaxants

    assitance of respiration(sometimes tracheostomy)

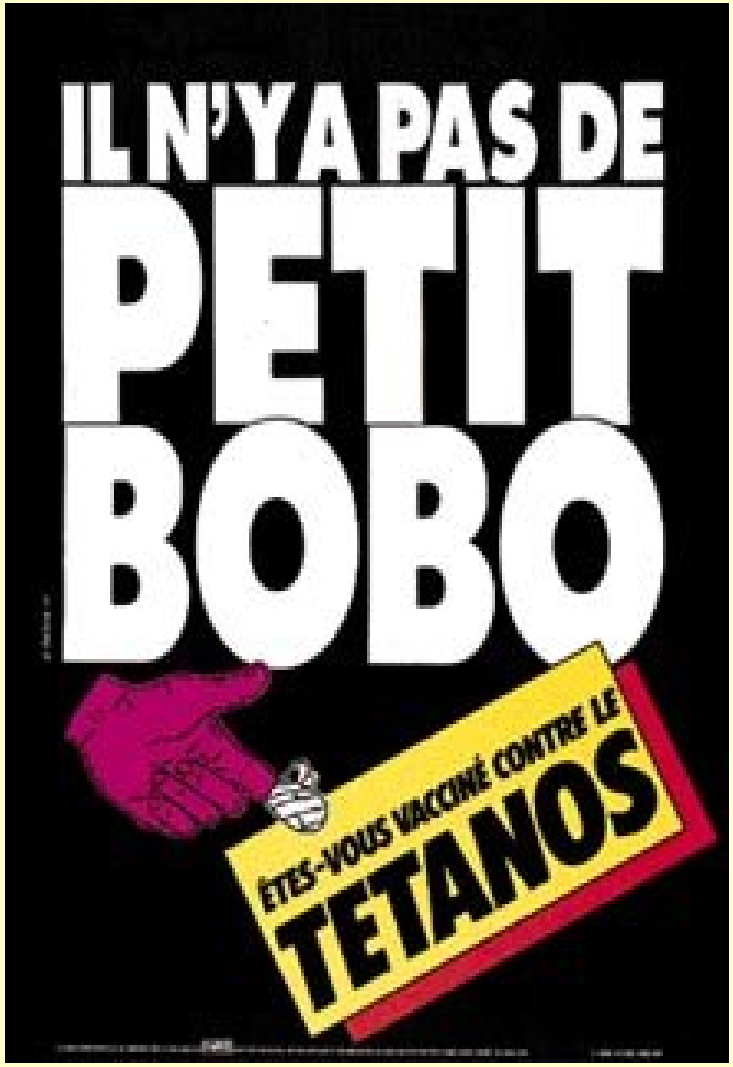
  - 10, 000 units of human tetanus immunoglobulin (**HTIG**)

Prevention and control

a. Immunization - HTIG 250 - 500 units (**to immune patient only**)
b. To non-immune toxoid followed by HITG

The recommended vaccination series for 1- to – 3-month-old babies: three injections given 2 months apart,
Booster doses about 1 and 4 years later.

Protection against neonatal tetanus:
Vaccination of pregnant women



Don't apply dung, ashes and mud to arrest bleeding

Clostridial food poisoning

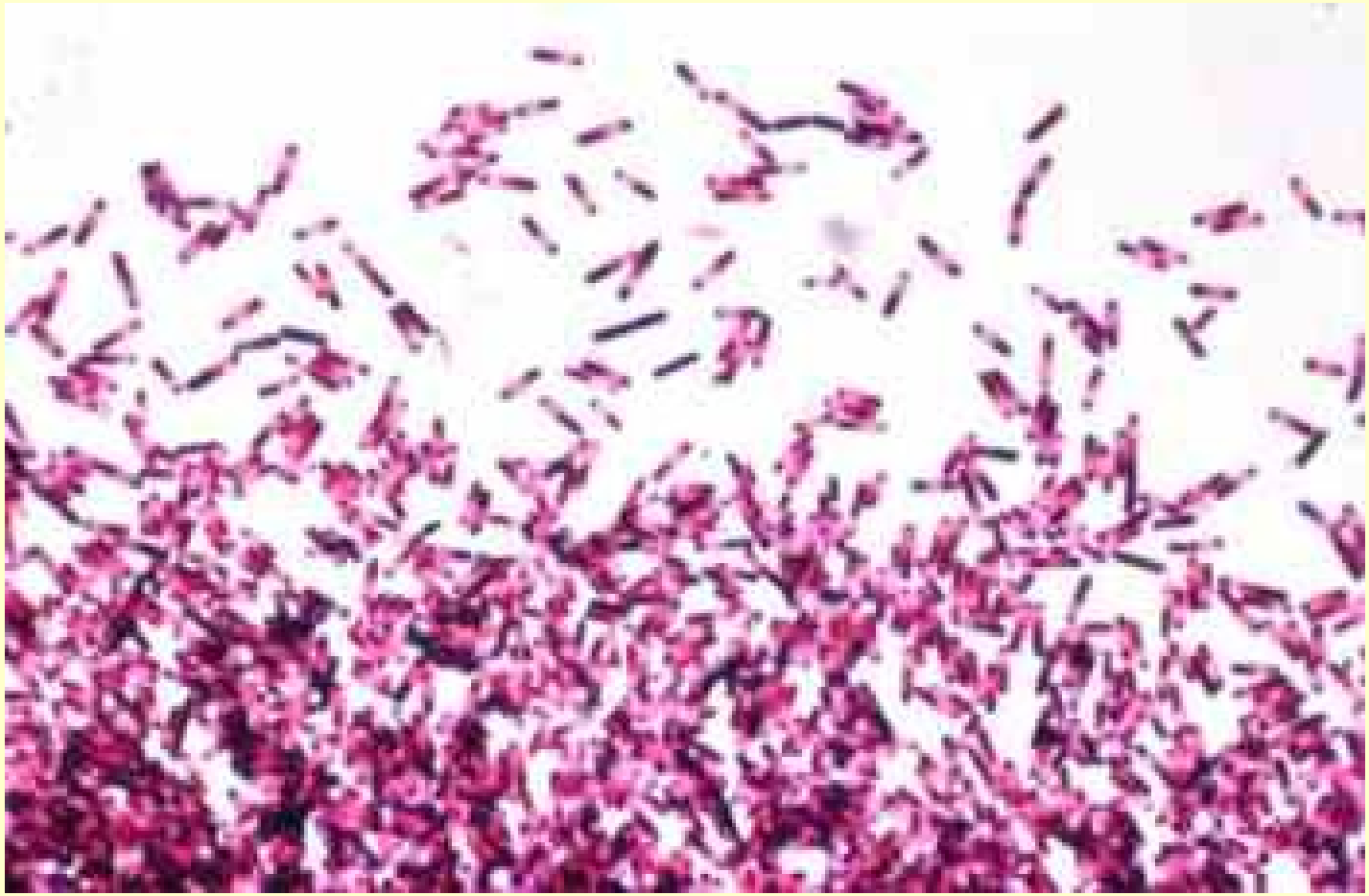
- C. perfringens, type A
- Carriers for food poisoning strains
- Survival of heat resistant spores in meals (meat, fish, beans etc.)
- Sporulation in gut - Short IP and watery diarrhoea, acute abdominal pain and vomiting for 24-48 hours
- Beta toxin production in C. prerfringens type C – Necrotizing enteritis(Pig bell)

BOTULISM
Sausage

8 toxins (A-G)

Food borne botulism
(IP 1-2 days)
Infant botulism
Wound botulism
(IP > 4 days)

Diagnosis
Isolation of organism in food/faeces
Detection of toxin in faeces / serum



C. botulinum

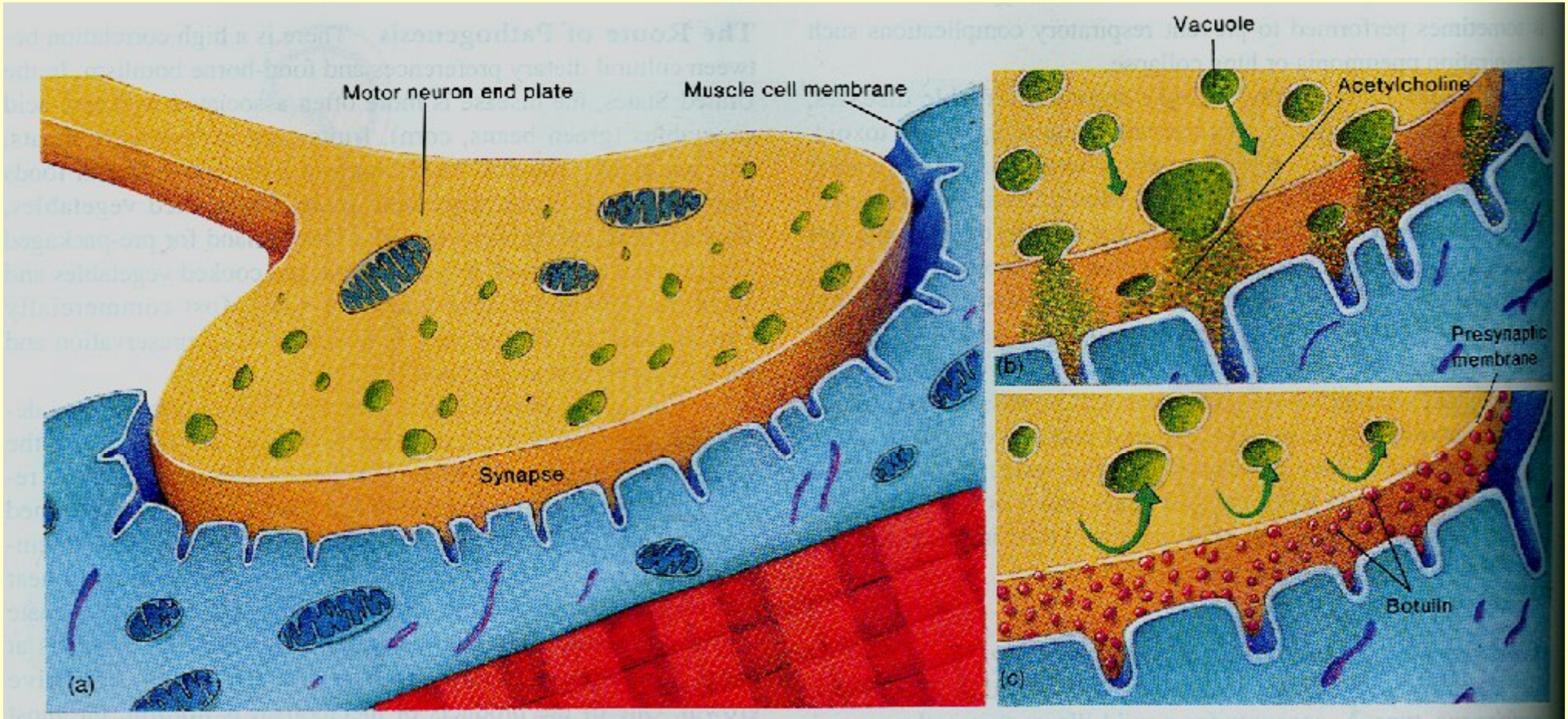


Figure 19.8

The physiological effects of botulism toxin (botulin). (*a*) The relationship between the motor neuron and the muscle at the neuromuscular junction. (*b*) In the normal state, acetylcholine released at the synapse crosses to the muscle and creates an impulse that stimulates muscle contraction. (*c*) In botulism, the toxin enters the motor end plate and attaches to the presynaptic membrane. This blocks release of the transmitter, prevents impulse transmission, and keeps the muscle from contracting.

Produces Botulism
World wide distribution
Found in soil and occasionally in animal feces
Sporese are highly heat resistant ,withstand 100C for 3-5 hrs.(120C for 5-10min)
Heat resistance is reduced by acid pH or high salt concentrations

Toxin
Released during growth and autolysis of bacteria.
It is found in 8 antigenic varieties.A-G
The principle cause for human disease A,B,E/F

A,B - Variety of foods
E - Fish products
C - Limberneck in birds
D - botulism in mammals
Toxin is neurotoxic protein
Destroyed by heating at 100C for 20 mins.
Action :Block release of Acetylecholine at synapses
and NMJ causing flaccid paralysis.
Pathogenecity
Illness is not an infection.
Botulism is an intoxication resulting from the ingestion of food in which C.botulinum has produced toxin.

Botulism

- **Incubation period 12-36 hours**
- **Diplopia**
- **Dysphagia and dysphonia**
- **Paralysis of muscles and respiratory system**
- **Mortality rate is 65-70%**

Botox

Infant botulism

- Spores germinate in the body and produce infection
- Raw honey has been implicated in some cases.
- Spores are common in dust and soil
- Spores germinate in the intestine and give off neurotoxin

Infant botulism

- Flaccid paralysis
- Weak sucking response
- Generalized loss of tone
- Weakness and cachexia
- Respiratory complications

Wound botulism

The spores enter a wound or puncture much as in tetanus

The symptoms are similar to those of food born botulism

More common in drug abusers

Treatment and prevention of botulism

-The CDC provides a source of type A, B, and E trivalent antitoxins

-Respiratory and cardiac support

-Penicillin

- Attention to home-preserved foods
- Addition of preservatives (sodium nitrate, salt and vinegar)
- Toxin in sensitive to 100C for few minutes