

20.3.2025

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Clostridium tetani

tetanus

C. perfringens

Causes gas gangrene; food poisoning

C. botulinum

botulism

C. difficile

pseudomembranous colitis

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### **Morphology and staining**



- General characteristics of Clostridium spp.
- Gram positive bacilli , spore former, motile except *C. perfringens*
- The location and shape of the spores is very important for the identification of this bacteria it may be in the middle, sub-terminal or terminal like in case of tetanus the spores looks dram-stick like bacilli.





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#### Clostridium tetani

- Causative agent of tetanus
- Found in soil, intestinal tracts, and feces of animals.
- Small rods, motile
- Spore-forming (drumstick appearance)
- Extremely sensitive to oxygen toxicity.



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# **Cultural Characteristics**



# **Culture** media

- Nutrient agar
- Blood agar
- Cooked meat broth/medium
- Thioglycolate medium



Cooked meat broth/medium



Thioglycolate medium

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- This type of bacteria grows on culture media containing blood or glucose in optimal temperatures 37 to 47 ° C.
- The macroscopic appearance of colonies is large, concave, rounded, shiny, and opaque with regular edges.
- Cooked meat broth is the best media for growth or thioglycollate medium.

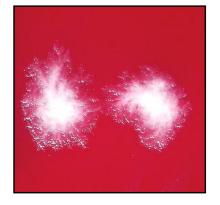
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# **Colony morphology**





C. perfringens

C. tetani

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# **Biochemical tests**



Species	Egg – yolk agar		gelatin	casein	tion	Acid production			
	Lecithinase	Lipase	Hydrolysis of gelatin	Digestion of c	Indole production	Glucose	Lactose	Sucrose	Maltose
C. tetani	-	-	+	-	٧	-	-	-	-
C. botulinum	-	+	+	+	-	+	-	-	+
C. perfringens	+	-	+	+	-	+	+	+	+

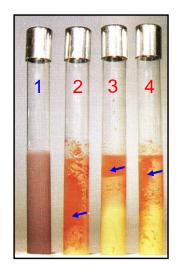
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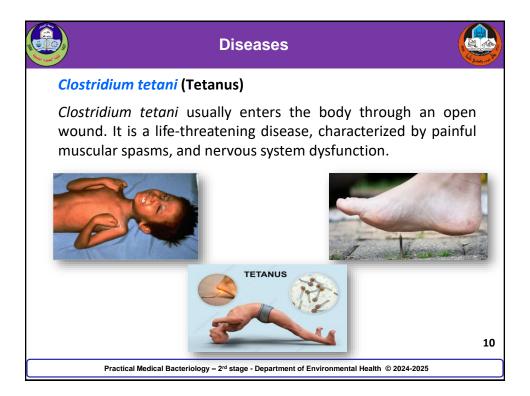


# **Litmus milk test**

1- un inoculated media2, 3, 4 the( stormy clot) reaction of three isolates with *C. perfringens* in litmus milk medium

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#### **Diseases**



#### **Clostridium Perfringens type A** (Soft tissue infections)

Causes gas gangrene (Myonecrosis) a life-threatening disease, and also causes food poisoning





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# **Diagnosis**



#### 1- Specimen: <u>Histological specimen</u> or <u>wound exudates</u>

- Histological specimens are transferred aseptically into a sterile screw-capped bottle.
- Specimens of exudates also specimens include: pus, necrotic tissue, feces, and food.

#### 2- Microscopical examination (Gram stain, Spore stain)

- Gram-positive bacilli and sporulated.
- The spore is oval, sub-terminal or drumstick (tetanus).

#### 3- Culture: Anaerobically at 37 °C

- Cooked meat medium → blackening of meat will observed with the production of H<sub>2</sub>S and NH<sub>3</sub>
- On blood agar  $\rightarrow$   $\beta$ -hemolytic colonies

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