Prepared by

Dr. Hala Sayed Hassan

Assistant professor of Bacteriology, Mycology and Immunology

Fac. of Vet. Med

Beni Suef University

Family Corynebacteriaceae

- Aerobic Non-Spore Forming Gram-Positive Bacilli
  It includes 3 genera; G. Corynebacterium, G. Listeria and G. Erysipelothrix

General characters of genus Corynebacterium

- 1-Gram positive small pleomorphic (coccoid ,club or rod forms), nonspore formers, in Gram's stained film they appear single , acute angles, in palisades of parallel cells and chineese letters appearance.
- 2- pathogenic members are non motile.

- 3-Facult. anaerobes,require enriched media such as blood agar and Loeffler's serum media.
- 4- Most Corynebacteria are catalase-positive, oxidase-negative
- Urease is produced by all pathogenic Corynebacteria with the exception of *C. bovis*.

Pathogenic members

1- C.pseudotuberculosis

caseous lymphadenitis in sheep and goat

Ulcerative lymphangitis in horse

Odematous skin disease in buffaloe

2-C. bovis

summer mastitis in cattle and suppurative infection in other animals

3-C. renale

urinary tract infection in cattle and occasionally other species.

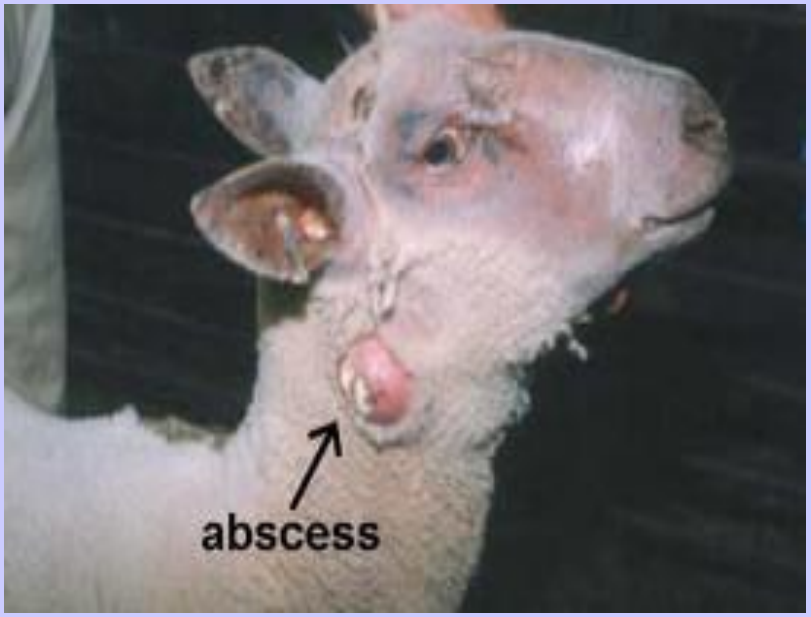
Human Corynebacteria

- C. diphteriae

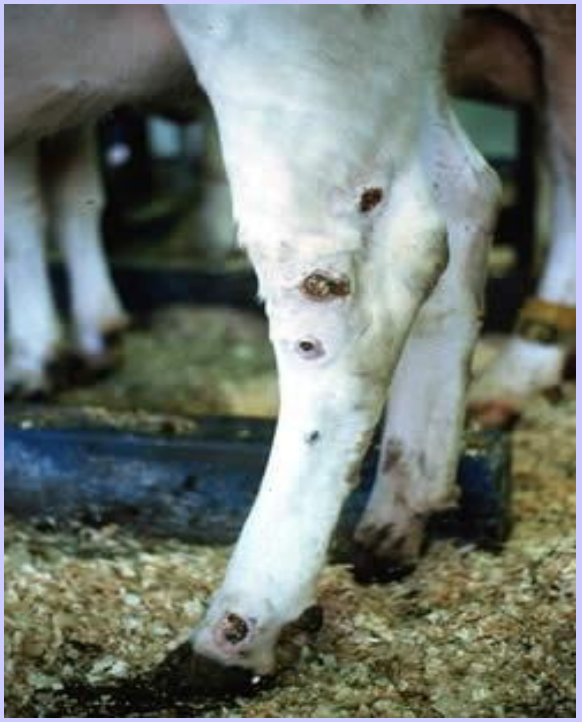
Diphteria in children

- C. acnes
- C. ulcerans

C. pseudotuberculosis



Caseous lymphadenitis in sheep



Ulcerative lymphangitis in horse

Odematous skin disease in buffaloe

Endemic disease transmitted by bitting of the forest fly in hairless areas of skin

It begin as swellings which initially started from a hairless area as axillary and groin regions then extends oedema to the hind or fore limbs, the belly and brisket region. It is firm in consistency and usually involves the drainage lymph nodes which may enlarge to attain a size of watermelon. In affected cases, OSD may be associated with haematuria or respiratory distress and recumbancy and almost ends with death.

Laboratory diagnosis

- **Samples** (sampling from the periphery of the abcess as the abscess pathology appear as onion like structure)
- **Culture**

Grow relatively slow on blood agar (white opaque flat colonies surrounded by a narow zone of hemolysis (its cell wall contain high percent of lipids upto 11.3% ,spattered on flame and easily pushed on agar surface)

selective media (BHIA+ nalidixic acid+fosfomycin)

- **Morphology:**
- **As previously mentioned in general characters**
- In addition to it contains volutin granules which can be stained by Albert stain and Neisser's stain (beaded in cytoplasm)

Biochemical characters

- Catalase and urease positive.
- Ferment glucose, galactose, maltose and mannose.
- According to nitrate reduction they are classified into 2 biotypes.
- (The ovine/caprine strains lack nitrate-producing capacity,
- while the equine and buffaloe strains usually reduce nitrate)

Starch agar hydrolysis test

medium =25 g of starch powder +1 L of purified water, mixed thoroughly and boiled for 1 min

then 15 g of brain heart agar were added, mixed thoroughly, then autoclaved at 121°C for 15 min.

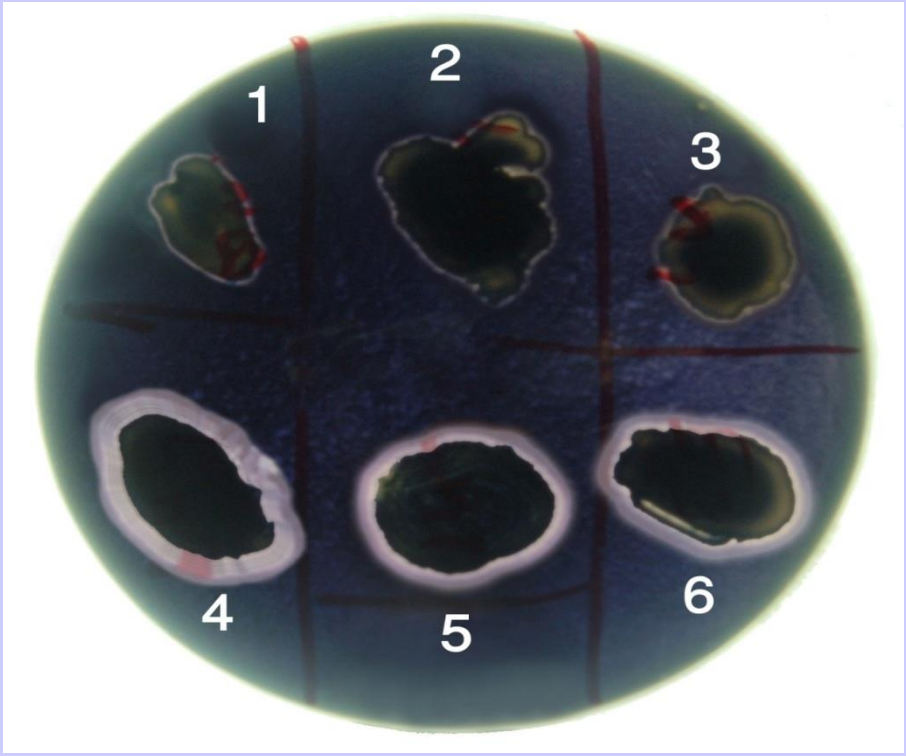
Isolates were streaked on plates of starch agar and incubated for 48 hours at 37°C.

The surfaces of inoculated media were flooded with iodine solution .

Positive result

clear zone around the colonies against a dark blue back ground (starch hydrolysis)

- As a result, when flooded with iodine solution, transparent clear zones are formed around the colonies, as the hydrolysed products formed around them do not form dark blue colour with iodine. On the other hand, the rest of the areas of the plates become dark blue, as iodine forms dark blue colour with the unhydrolysed starch in these areas.



Starch hydrolysis plate showing clear zone of hydrolysis around colonies of *C. pseudotuberculosis* of buffalo origin(No.4, 5, 6) while sheep isolates (No1, 2, .3) showing no hydrolysis.

- The most chracteristic test is the synergism of hemolysis with R. equi (modified CAMP test) using FNR medium(BHIA+7% defebrinated sheep blood+fosfomycin+ nalidixic acid+ R.equi filterate)

Synergistic hemolysis between C. pseudotuberculosis and R. equi

Pathogenicity test

- Biotype 1…….I/P inoculation into male guinea pig

Orchitis with localization of the organism in the scrotal sac (strauss reaction)

Biotype 2……. rapid haemorrhagic lesions at site of inoculation associated with rapid death of the experimental animals as a result of the presence of other toxigenic factor(s) beside PLD

Virulence factors

- 1. Phospholipases (dermonecrotic toxins):
- It has sphingomyelinase activity increasing vascular permeability. It is produced by C.ovis
- Type D (PLD) is responsible for CLA in sheep and goat and
- ulcerative lymphangitis in horses while type C is responsible for OSD in buffaloes.

- 3. Haemolysin
- 4. Cord factor -Toxic trehalose
- It is corynemycolic acid, corynemyolenic acids. They are granulomagenic and may mediate intracellular survival

C. bovis

- summer mastitis in cattle and suppurative infection in other animals
- *Corynebacterium bovis* is a lipophilic bacterium which produces small, white, dry, non-haemolytic colonies.
- Absence of growth on MacConkey agar

C. renale group

- opportunistic agents of urinary tract infections in cattle and occasionally other species.
- Produce small non-hemolytic colonies after 24 h incubation.
- Pigment production after 48 hours
- Ferment glucose, variable in nitrate reduction, urease test positive

Differentiation of the species in the *Corynebacterium renale* group

Feature	*C. renale* (type I)	*C. pilosum* (type II)	*C. cystidis* (type III)
Colour of colony	Pale yellow	yellow	White
Growth in broth at pH 5.4	+	_	_
Nitrate reduction	-	+	-
Casein digestion	+	_	_
Acid from Xylose	_	+	_
Hydrolysis of tween 80	_	_	+

Identification of corynebacteria of veterinary importance

	C. pseudotuberculo-sis	C. renale	C. bovis
hemolysis	+	-	-
Nitrate reduction	v	v	-
Urease	+	+	-
acid from maltose	+	+	-
acid from glucose	+	+	+

R. equi

- Former members of genus Corynebacteria
- Garm positive rods or cocci
- Grow on non enriched media producing mucoid (capsule production), salmon pink colonies
- Non-hemolytic on blood agar
- Modified CAMP test positive