Non-spore forming Gram-positive bacilli
Corynebacterium

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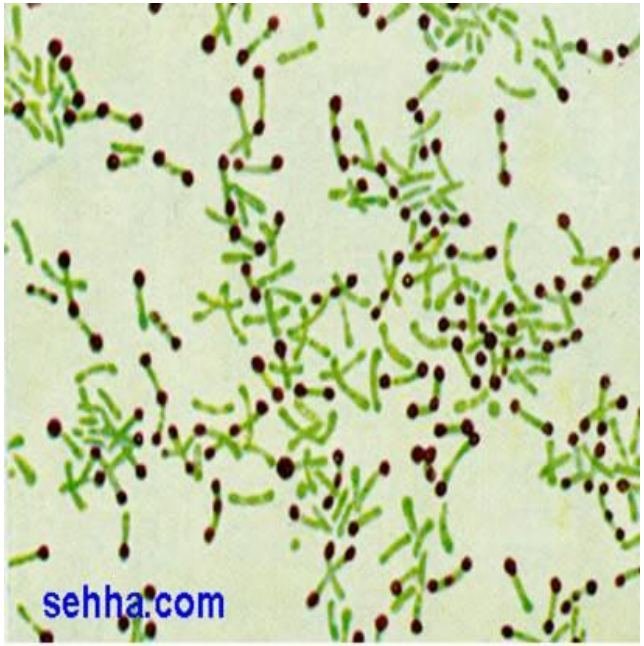
Many members of the genus corynebacterium are members of the normal flora of the skin and mucous membranes of the humans. *C. diphtheriae* is the most important member of the group, it can produce a powerful exotoxin that causes **diptheria** in human.

Morphology & identification:

These bacilli characteristically possess irregular swellings at one end (clopped shaped). The metachromatic granules are irregularly distributed within the rode (beaded appearance). In stained smear (**Alber's stain**) the bacilli tend to arranged in 89or at acute angle to one another. In stained smear from culture medium the bacilli arranged in **chine's letter appearance**

C. diphtheria grow aerobically on most ordinary media. On blood agar the colonies are small, granular and gray & may have small zone of hemolysis. On agar containing potassium tellurite, Potassium tellurite medium (McLeod's medium): black colonies. Because this agar contain tellurite that is reduced to elemental tellurium within the organism

On selective **Tinsale media**, black colonies surrounded by halo (**Diagnostic**).



خلايا ميكروب الدفتيريا بعد صبغها، ويلاحظ أنها تأخذ فى ترتيبها شكل الأحرف الصينية ، كما يوجد بها حبيبات تصطبغ أكثر من بقية الخلية

Pathogenesis, pathology & clinical signs:

Naturally, *C. diphtheria* may occurs in the respiratory tract, in wound or on the skin of normal carriers. It spread by droplets or by direct contact. The bacilli can grow on mucous membranes or skin wound and the toxigenic strains start producing **exotoxin.** The diphtheria exotoxin is **heat-labile** polypeptide, composed of two fragments. Fragment B is required for transport of fragment A into the cell. Fragment A inhibits protein synthesis through inhibition of polypeptide chain elongation factor leading to necrosis and neurotoxic effects.

The exotoxin is aborted into the mucous membranes and destroys the epithelium causing necrosis & inflammation. The necrotic epithelium embedded in exuding fibrin and RBCs & WBCs forming grayish psudomembrane, usually on the tonsils. Removal of pseudomembrane may result in bleeding.

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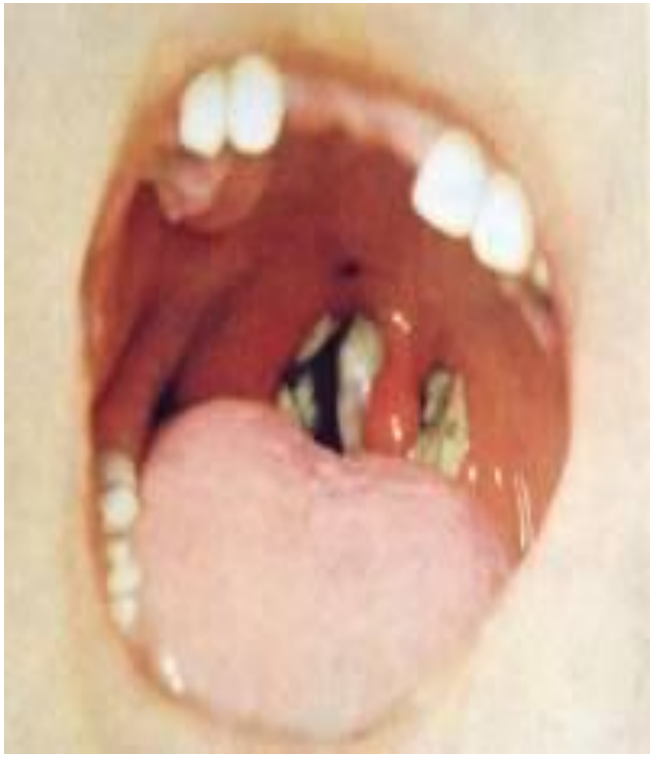
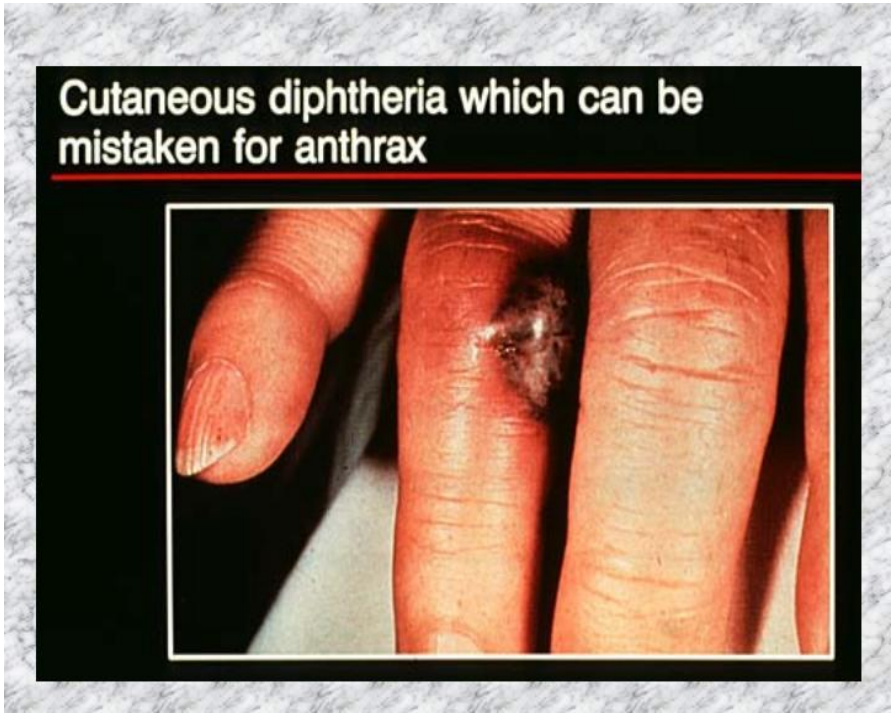
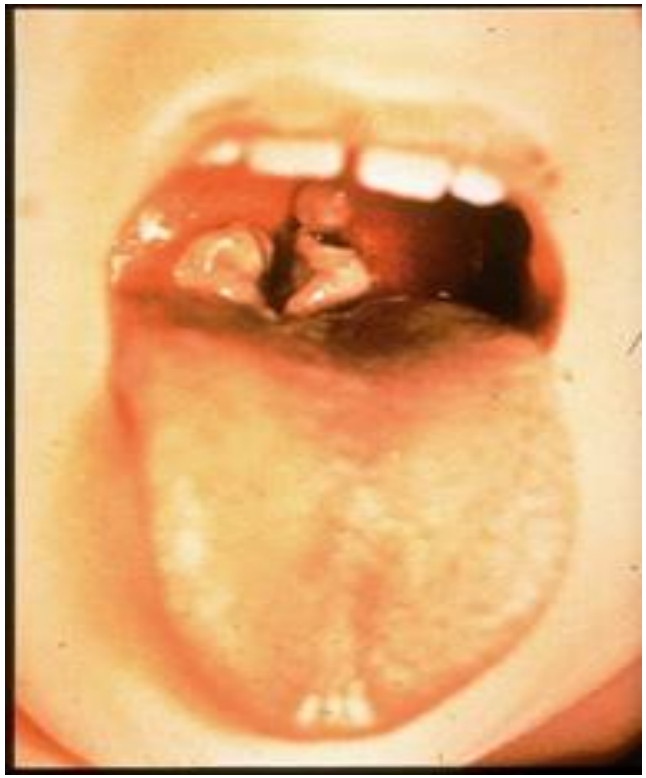
C. diphtheria



Clinical findings:

- ✓ *Respiratory diphtheria: Thick, gray, adherent **pseudomembrane** over the tonsils and throat.
- ✓ Cutaneous diphtheria: ulcerating skin lesion covered by a gray membrane.





Marked edema of the neck may noticed. The bacilli within the pseudomembrane continue to secrete exotoxin actively and result in distant toxic systemic damage particularly on paranchymatous organs (Heart, liver, kidney). It may also cause nerve damage resulting in paralysis of soft palate, eye or extremities. Formation of pseudomembrane may occur on infected skin wounds. *C. diphtheria* never invade deeper tissues & particularly never enter bloodstream.

In diphtheria, the inflammation begins in the respiratory tract, sore throat & fever & dyspnea usually enveloped. Difficult breathing or even suffocation may result due to inflammation and swelling in the area (relieved by intubation or tracheostomy). Untreated patients may continue shed the bacilli in the environment for weeks or months after recovery (Convalescent carriers).

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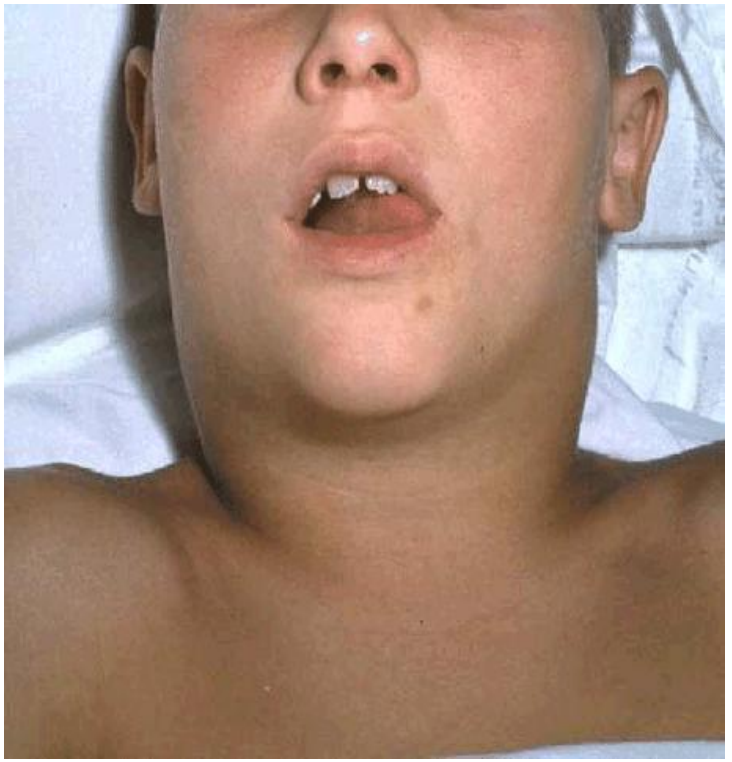
C. diphtheria



Laboratory diagnosis:

Note: specific treatment must not delayed for laboratory report if the clinical picture is highly suggestive of diphtheria.

1.Specimens: throat or nasal swabs or from other suspected lesions must be obtained before treatment.

2. Direct stained with Methylene blue (Alber's stain) . Staining by Gram or methylene blue (G +ve rods arranged as L or V shaped ,Chines letter appearance)

Albert stain: differential stain for metachromatic granules.

3. Culture on blood agar plate

Selective media: Loefler slants, tellurite plate or Tinsale medium.

4. Toxigenicity test for exotoxin production: -

1- in vivo: animal inoculation, Schick test.

2- in vitro: (Elek's test) antibody-based gel diffusion precipitation test to confirm toxin production.

3- tissue culture neutralization assay.

4- PCR assay: for the presence of toxin gene in the organism isolated from the patient.

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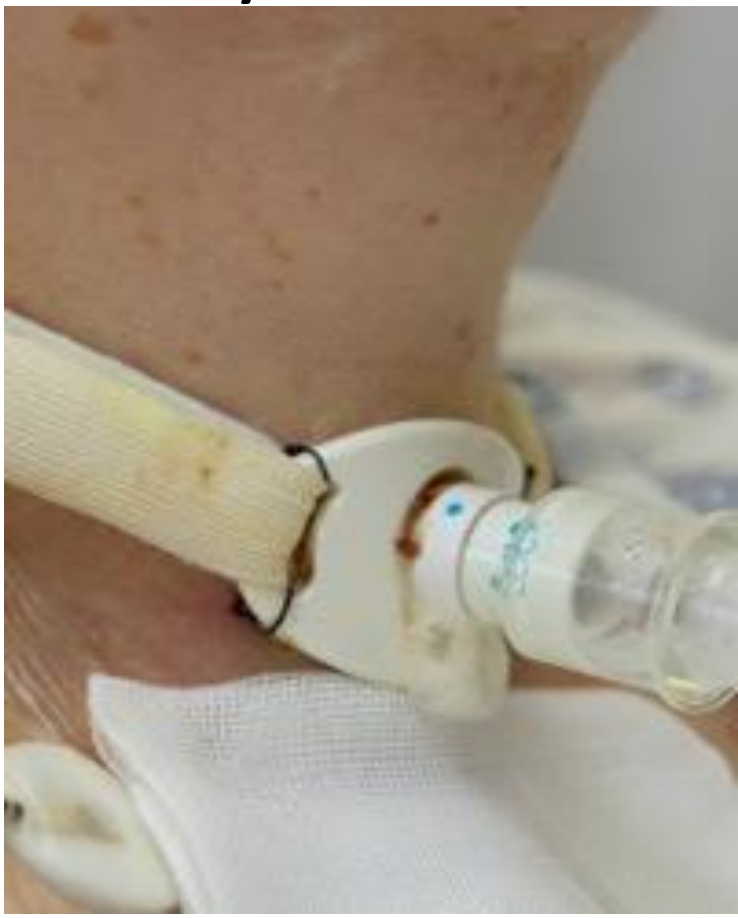

Schick test:

Intracutaneous skin test distinguishes between persons who are susceptible and those who are immune to diphtheria toxin and to test for sensitivity to toxoid.

Procedure: the test is performed by intradermal injection of 0.1 mL of purified standardized toxin, if the patient has no antitoxin the toxin will cause inflammation at the site of injection 4-7 days later. If no inflammation occurs, antitoxin is present and the person is immune.

Epidemiology:

Either clinical or subclinical infection at an early age yield protective levels of antitoxin. Thus most members of the population, except children are immune. By age 6-8 years about 75% of children in developing countries where skin infection with diphtheria are common have protective serum antitoxin. Absorption of small amount of *C. diphtheria* toxin does not produce a disease, but it can serve as antigenic stimulus for immune system to produce antitoxin.

Active immunization with *C. diphtheria* **toxoid** during childhood yield antitoxin levels that generally adequate until adulthood. Young adults in developed countries (Non **endemic** areas) should receive a booster dose because diphtheria bacilli are not sufficiently prevalent to provide a stimulus of subclinical infection. **DPT** is the combined of **diphtheria toxoid**, **tetanus toxoid** & **pertussis vaccine** used as a single injection for immunization of children. For booster dose in adults only **TD** is used.

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