PASSION ACADEMIC TEAM

YU - MEDICINE

Sheet# 5 - MICROBIOLOGY

Lec. Date :

Lec. Title : Pseudomonas

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RESPIRATORY SYSTEM

Pseudomonas

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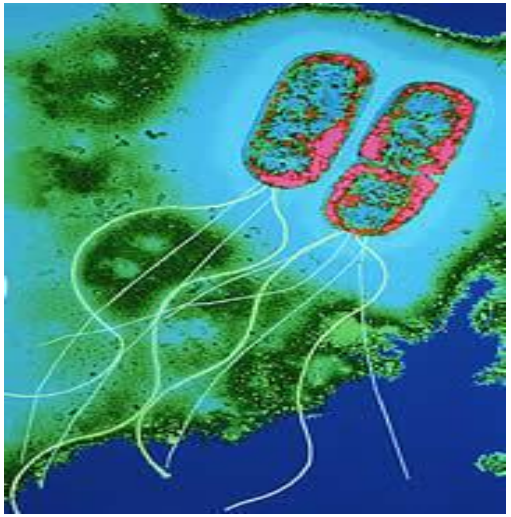
*Pseudomonas are everywhere around us (swimming pools, rivers, lakes Etc) and specially in wet places

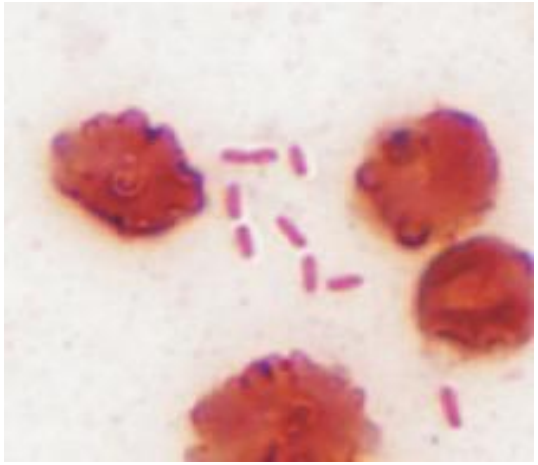
Pseudomonas



Structure and Physiology

- Gram-negative rods.
- Motile with polar flagella.
- Obligate aerobe.
- Oxidase-positive. * it's the only enterobacteria that is oxidase positive
- Do not ferment carbohydrates.
- Resistant to multiple drugs.

*it has a high mutation rate which makes it resistance to multiple drugs... We can solve this problem by giving combined drugs



Pseudomonas aeruginosa:

- Opportunistic infections of multiple sites

- 80% of human infections caused by aeruginosa species

*It can cause many infections starting with simple skin infection (Hot tub folliculitis) .. Pneumonia and many others infections

- Does not require enriched media

* can live in basal media.... بكفيها Agar يعني ال

- Can survive and multiply over a wide temperature range (20–42°C) in almost any environment

* it's wide temperature range is an important virulence factor

- P. aeruginosa can infect almost any external site or organ.
- P. aeruginosa is invasive and toxigenic.

* toxins are the major virulence factor

Clinical note :
These toxins can cause tissue damage.. So before we start the treatment procedure we have to remove a part from the damaged skin to reduce the microbial contant

- P. aeruginosa is resistant to many antibiotics
- Emits an intense “fruity” odor.

* it has a fruity odor like the fresh apple odor

- Most strains of P. aeruginosa produce multiple extracellular products, including exotoxins

EPIDEMIOLOGY

- The primary habitat of P aeruginosa is the environment.
- Colonization rates may be higher in hospitalized patients. * most of pneumonia in ICU is caused by Pseudomonas aeruginosa
- Pseudomonas aeruginosa complicates cystic fibrosis (CF) * in CF pt. The accumulation of mucous helps the p. aeruginosa to grow (by giving it the wet medium)
  * the most common - 70%- of isolated Pseudomonas in pneumonia in CF pt. Is p.aeruginosa

- Pseudomonas spp. normally inhabit soil, water, and vegetation and can be isolated from the skin, throat, and stool of healthy persons.

- Spread is via contact with fomites or by ingestion of contaminated food and water.

* أكثر مكان ممكن تنتشر فيه هو ال public swimming pools

* it can enter the body by ingestion, inhalation - the most dangerous - or attack the skin causing simple skin infection (Hot tub folliculitis) .... If it enters the :
Ear it'll cause otitis
Eye it'll cause chornial ulceration which can lead to lose of sight

- High risk population: patients receiving broad-spectrum antibiotics, with leukemia, burns, cystic fibrosis, and immunosuppression.
- Methods for control of infection are similar to those for other nosocomial pathogens.
- Special attention should be paid to sinks, water baths, showers, hot tubs, and other wet areas.

Pathogenesis and Immunity

This organism is widely distributed in nature

It is pathogenic only when

Distruption means infect or injury

1. Disruption of mucous membrane and skin.
2. Usage of intravenous or urinary catheters.
3. Neutropenia (as in cancer therapy).

Neutroprnia means low concentration of neutrophils

Pathogenesis

All of things that are mentioned are virulence factors

- Antigenic structure, enzymes, and toxins
- Pili and nonpilus adhesins.

Pili function : plays the major rule in attachment

- Capsule (alginate, glycocalyx): seen in cultures from patients with cystic fibrosis.

LPS : lipopolysacharides, a toxin that's found in gram negative bacteria that makes low blood pressure and thrombosis

- LPS- endotoxin, multiple immunotypes.

All are stains that are produced by toxins :

- Pyocyanin: catalyzes production of toxic forms of oxygen that cause tissue damage.

*Pyocyani is a blue-green chemical.. It has an advantage we can use it as anti-tumor

- Pyoverdin: a siderophore.

* pyoverdin is yellow green which makes chelation of iron

- Pyorobin

*pyorobin it's a red stain.. No formation available about it because it's not well understood

Proteases All are exoenzymes

- Serine protease, metalloprotease and alkaline protease cause tissue damage and help bacteria spread.
- Phospholipase C: a hemolysin

*phoshpholipase C : makes RBC'S Distruction

- Exotoxin A: causes tissue necrosis

*exotoxin A : cause tissue necrosis specially in trechea

- Exoenzyme S and T: cytotoxic to host cells.

*exoenzymes S and T : they do toxic effect in the cell itself

Pulmonary infection

- Tracheobronchitis
- Necrotizing pneumonia in CF patients: diffuse, bilateral bronchopneumonia with microabscess and necrosis.

***Pseudomonas aeruginosa* and cystic fibrosis. The** lungs of a young adult are shown at autopsy. There is both extensive inflammation and thick biofilm throughout

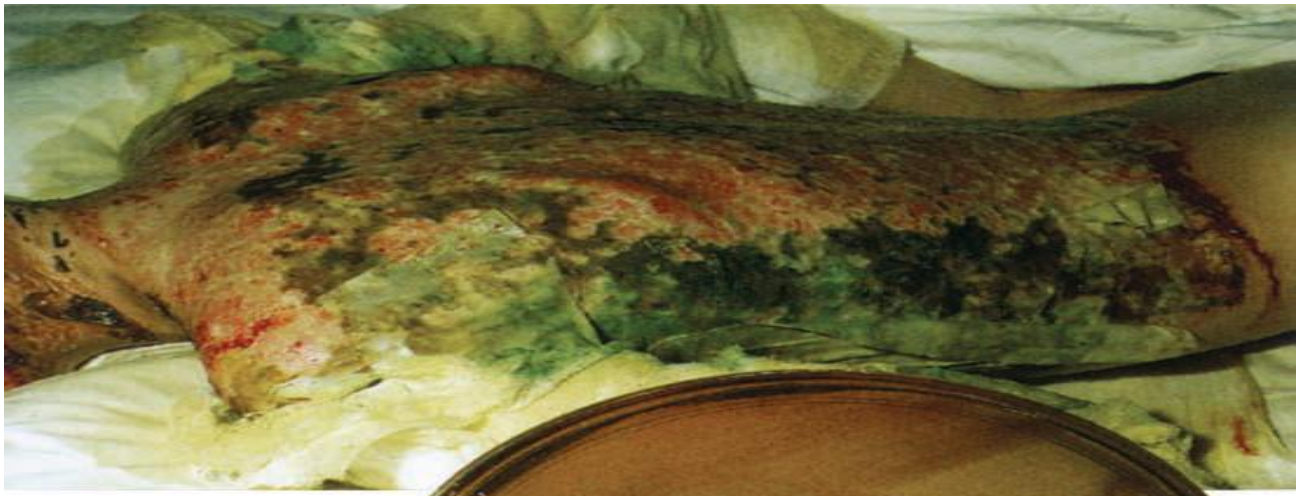
It needs a gene therapy to be treated

Ear infections

- Otitis externa: mild in swimmers; malignant (invasive) in diabetic patients.

- Chronic otitis media

Mild in good immune persons
Malignant (we said invasive to tell you that is a serious infection 💀☠️) in immunosuppressive people



** هاي الصورة بشوفها بالحروق.. بهاي الحالة بتروح ال p.aeruginosa لجزء المحروق وبتعمل microbial growth... طيب ليش؟ ببساطة عيني.. لأنه بحالة الحرق راحت ال skin يلي هي ال first defense line وصارت ال mucosa مكشوفة ف صار الوسط مناسب لنموها
**لون الحرق بكون حسب لون ال stain يلي هيفرزهم ال toxin
(ارجع سلايد 9 إذا ناسيهم 😔)
**نأسف إذا الكتابة مش كتير واضحة بس الصورة جاية بالنص شعمل يخوااااان 🤓💔

***Pseudomonas aeruginosa* pigment production.**

The blue color of pyocyanin when mixed with yellow tissue or media components typically produces a green discoloration.

Laboratory Diagnosis

- Specimen: pus, sputum.
- Culture: blood agar plate and differential media.

For diagnosis
1.oxidase reaction
2.the odor
3.the stain.. But here many bacteria species have the same stain, to know if this stain is from p.aeruginosa see below:

ممكن بعض البكتيريا تعمل نفس اللون لكن الفرق بال coloration هو انه ال :

P.aeruginosa will make diffusion in the agar

ف بصير ال plate كله ملون بنفس لون ال toxin بينما بالانواع الثانية من البكتيريا يلي بتلون هو ال colony بس

- Identification of P. aeruginosa is usually based on oxidase test and its colonial morphology:
- β-hemolysis
- The presence of characteristic pigments and sweet odor
- Growth at 42 ^{o}C.

P. aeruginosa

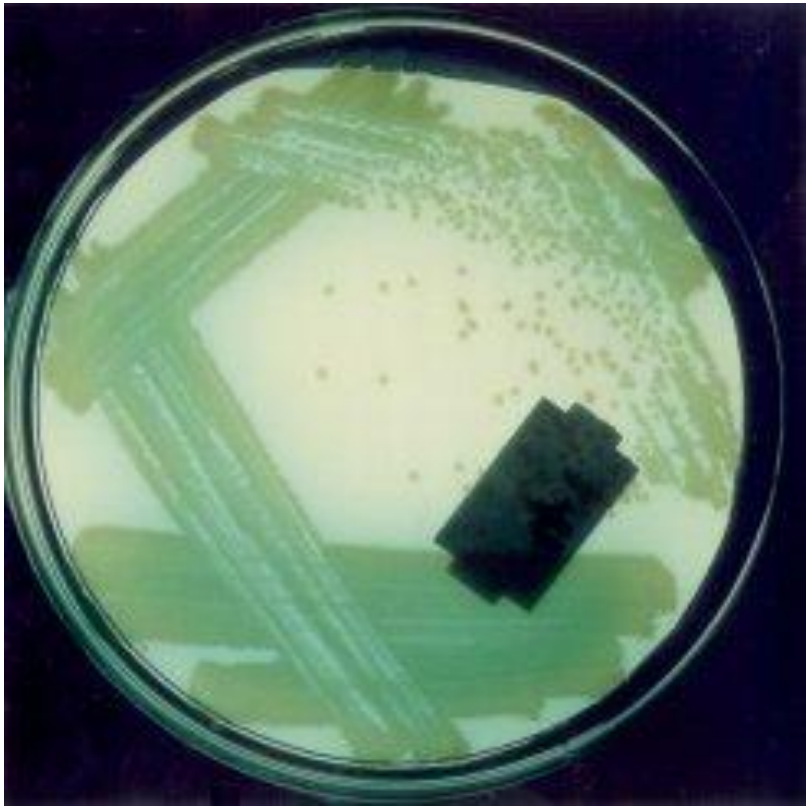
Forms round colonies with a fluorescent greenish color, sweet odor, and β-hemolysis.

Pyocyanin- nonfluorescent bluish pigment

pyoverdin- fluorescent greenish pigment

pyorubin red pigment

Some strains have a prominent capsule (alginate).



هون زي ما احنا شايفين حتى المنطقة يلي ما فيها growth ماخذة لون ال stain تاعت ال toxin
** ارجعوا تذكروا مين ال toxins يلي بتعمل اللونين

Blue-green // yellow -green 😉

Treatment

* it needs a long term therapy (6 weeks)

- Combined antibiotic therapy is generally required to avoid resistance that develops rapidly when single drugs are employed.

- Avoid using inappropriate broad-spectrum antibiotics, which can suppress the normal flora and permit overgrowth of resistant pseudomonads.

Prevention and Control

Control:

1. Patients at high risk should not be admitted to a ward where cases of pseudomonas infection are present.

2. Patients infected with *P. aeruginosa* should be isolated.

ركزوا على isolated.. ركزووووواااا

3. Sterilize *جميع الأدوات يلي بستخدمهم المريض لازم يصيرلهم sterilization

all instruments, apparatus, and dressing;

4. antimicrobial and other therapeutic substances.

5. Monitor clinically relevant isolates of *P. aeruginosa* by a suitable typing system to identify epidemic strains.

نوت الدكتور حكاه: \:
عندي اشي اسمه ال molecular diagnosis هو يلي بخلينا نعرف ال source of infection... بنعمل
molecular therapy يعني بنعزل ال Pseudomonas وبنحدد ال similarity بينها وبين ال other
Pseudomonas المعزولة بالمستشفى لحتى نحدد هل هاي ال Pseudomonas يلي مع المريض جاية من داخل
المستشفى ولا من خارجه.. بالانجليزي يعني

To know if it is nosocomial or non-nosocomial

Moraxella catarrhalis

- Previously known as *Branhamella catarrhalis*

*ال moraxella catarrhalis كان يصنف مع ال neisseria لأنه gram negative diplococcus

- Normal commensal of the respiratory tract

- Has become an important opportunistic pathogen
  - Predisposing factors
    - Advanced age, Immunodeficiency, Neutropenia, Other debilitating diseases

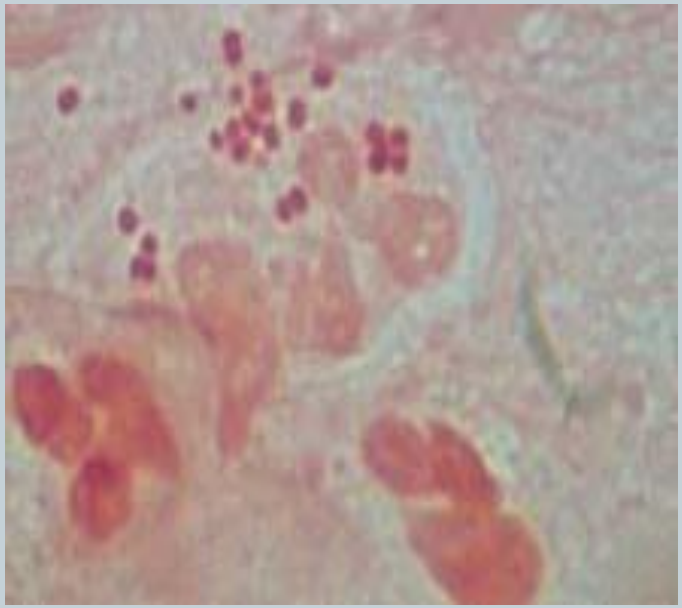
*معظم الإصابات وحدتها كلها بتعتمد ع الحالة المناعية للشخص (مش حاجة مهمة.. تنوروا بالعلم 🤓)،

- Clinical infections
  - Pneumonia
  - Sinusitis
  - Otitis media (3rd most common cause)

Virulence factors:

- Endotoxin
- Pili
- Beta-lactamase *beta lactamase : responsible for resistance of beta lactam ring drugs

Laboratory Diagnosis: *Moraxella catarrhalis*



Direct smear from an otitis media sample showing intracellular gram-negative diplococci

👀 focus on it ركزوا علي ملووون.. للناطقين بغير العربية

Laboratory Diagnosis: *Moraxella catarrhalis*

- Colonies appear smooth with a grayish- white color
- When colonies pushed with loop, they “scoot” across media

Scoot: if I pushed the colony with the loop it'll move with it

Moraxella catarrhalis **growing on chocolate agar after 24 hours of incubation**

Laboratory Diagnosis : *Moraxella catarrhalis*

- Oxidase positive
- Catarrhalis Disc (butyrate esterase)
  - Positive= blue-gren
- All CTA (Cystine tryptic agar) sugars negative
- Produce beta- lactamase

*if we made gram stain we will found intracellular gram negative diplococci

*butyrate disc : moraxella تفاعل مميز لل

* إذا أخذنا عينة من البكتيريا وحطيناها ع ال disc يلي عليه butyrate وتحول اللون إلى dark blue // purple estrase بكون الاختبار positive

يعني هاي البكتيريا فيها هذا الانزيم وبالتالي هي moraxella

Identification of Selected *Neisseria* Species & *Moraxella*

الفرق بين ال moraxella و ال neisseria species هو أنه :

Moraxella is a suger non-fermetive... Can't make fermentation for any type of suger

Species	Growth			Acid production			
	BAP	R.T	T/M	Gluc	Mal	Lac	Suc
N. gonorrhoeae	=/+	=	+	+	=	=	=
N. meningitidis	+	=	+	+	+	=	=
N. lactamica	+	v	+	+	+	+	=
N. sicca	+	+	=	+	+	=	+
M. catarrhalis	+	+	=	=	=	=	=

Treatment

Amoxicillin - clavulanate = Augmantine

ادري انكو بتدرسوش فارما... بس بلاش يجي بالامتحان 🌝🌵

- Amoxicillin-clavulanate, second- and third-generation oral cephalosporins, and trimethoprim-sulfamethoxazole (TMP-SMZ) are the most recommended agents.

- Alternatively, azithromycin or clarithromycin can be used

Bacillus

B. anthracis: anthrax of the animals and humans.

*Bacillus is the most dangerous group
*B.anthrax is the main and the most dangerous pathogen in it
* anthrax = الجمرة الخبيثة

Morphology and Physiology

* it's a Giant, long bacteria
*it has a sharp cut end (squeaky end)
* it's capsule made of polypeptide (glutamate polypeptide)

وهذا يؤدي لاختلاف ال immune response

- Aerobic or facultatively anaerobic.
- Large gram-positive rods, have square ends, arranged in long chains.
- Spore is located in the center of the cell.
- Most are saprophytic (soil, water, air, and on vegetation.)

Physiology and Structure

- *B.* anthracis is encapsulated and non-motile.

- Animal products contaminated with anthrax spores can be sterilized only by autoclaving.

*spores can be sterilized only by autoclaving

Pathogenesis and Immunity

- Primarily a disease of herbivores (sheep, cattle, horses); humans are rarely affected.

*spores formation starts with in dead body of the animal

- Being used by the terrorists as a biological warfare.

- In animals, portal of entry is mouth and GI tract.

*B.anthracis enters the animal by eating contaminated food with anthrax spores // may be enter by inhalation

- In humans, scratches in the skin (95% of infection), ingestion or inhalation lead to infection.

نركز حبتين هون شوي 🤓

*most common anthrax in human is the skin anthrax
*most dangerous anthrax in human is the pulmonary -inhalated- anthrax

- Inhalation is the most likely route for infection with biological warfare (LD50: 2,500-55,000).

- The spores germinate in the tissue at the site of entry, and growth of the vegetative forms results in gelatinous edema and congestion.

- Bacillus spread via lymphatics to the blood and other tissues.

Virulence factors

Capsule (encoded from a plasmid)

Exotoxins (A-B toxins encoded from another plasmid)

This toxin complex increases vascular permeability which leads to shock.

Human Transmission

- Cutaneous
  - Contact with infected tissues, wool, hide, soil
  - Biting flies
- Inhalational
  - Tanning hides, processing wool or bone
- Gastrointestinal
  - Undercooked meat

Human Transmission

- Tanneries
- Textile mills
- Wool sorters
- Bone processors
- Slaughterhouses
- Laboratory workers

*هذول همه الأكثر عرضة للإصابة

*people who work with animals should be vaccinated against anthrax

يعني الجماعة يلي فوق

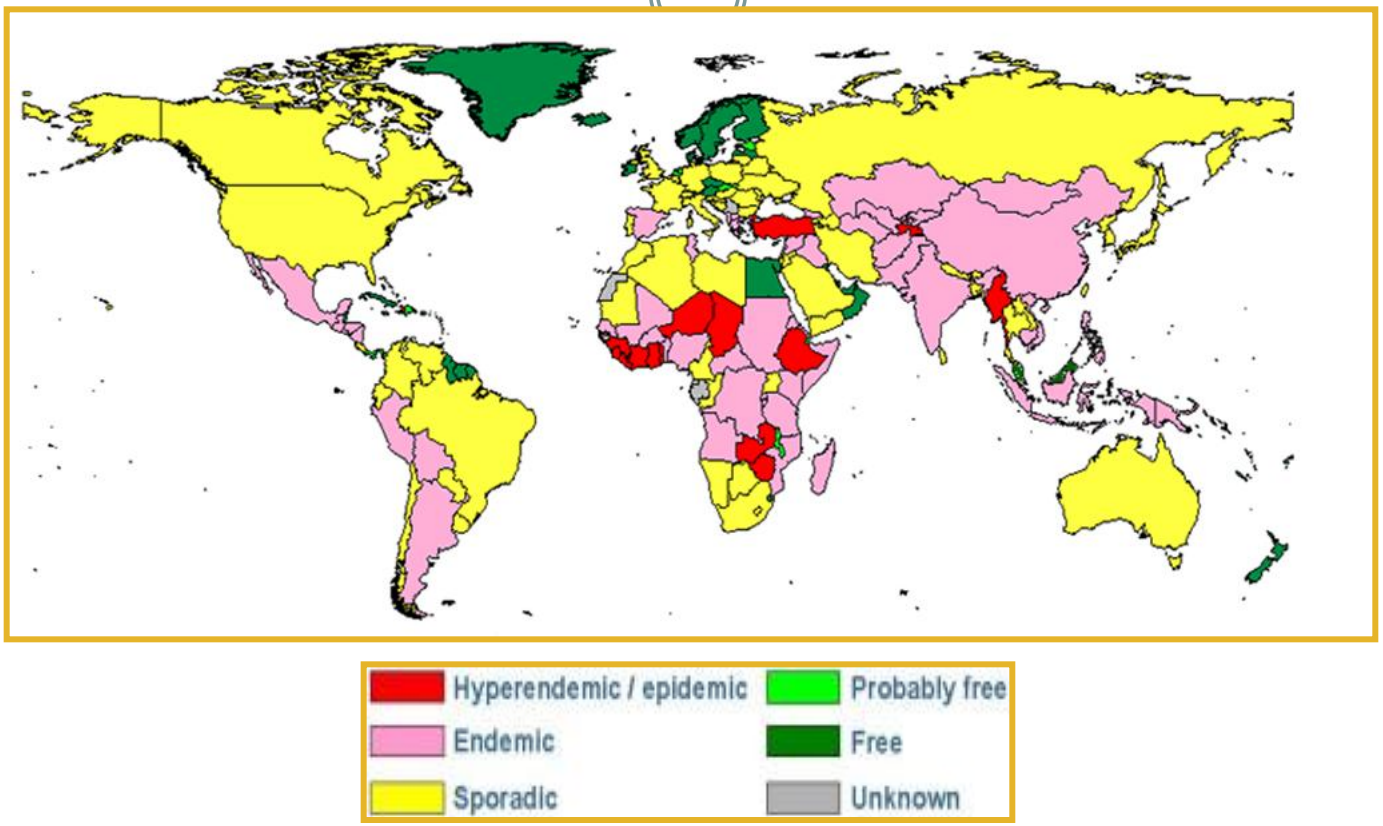
Animal Transmission

- Bacteria present in hemorrhagic exudate from mouth, nose, anus
- Oxygen exposure
  - Spores form
  - Soil contamination
- Sporulation does not occur in a closed carcass
- Spores viable for decades

Animal Transmission

- Ingestion
  - Most common
  - Herbivores
    - Contaminated soil
    - Heavy rainfall, drought
  - Carnivores
    - Contaminated meat
- Inhalation
- Mechanical (insects)

Anthrax Distribution



20,000 to 100,000 cases estimated globally/year

* أكثر الأماكن انتشارا هي في أفريقيا وشرق آسيا
* الحل الوحيد إذا ظهرت ماشية مصابة بالمرض هو التخلص منها.. قتلها ثم حرقها

Disease in Humans

Cutaneous Anthrax

Gastrointestinal Anthrax

Inhalational Anthrax

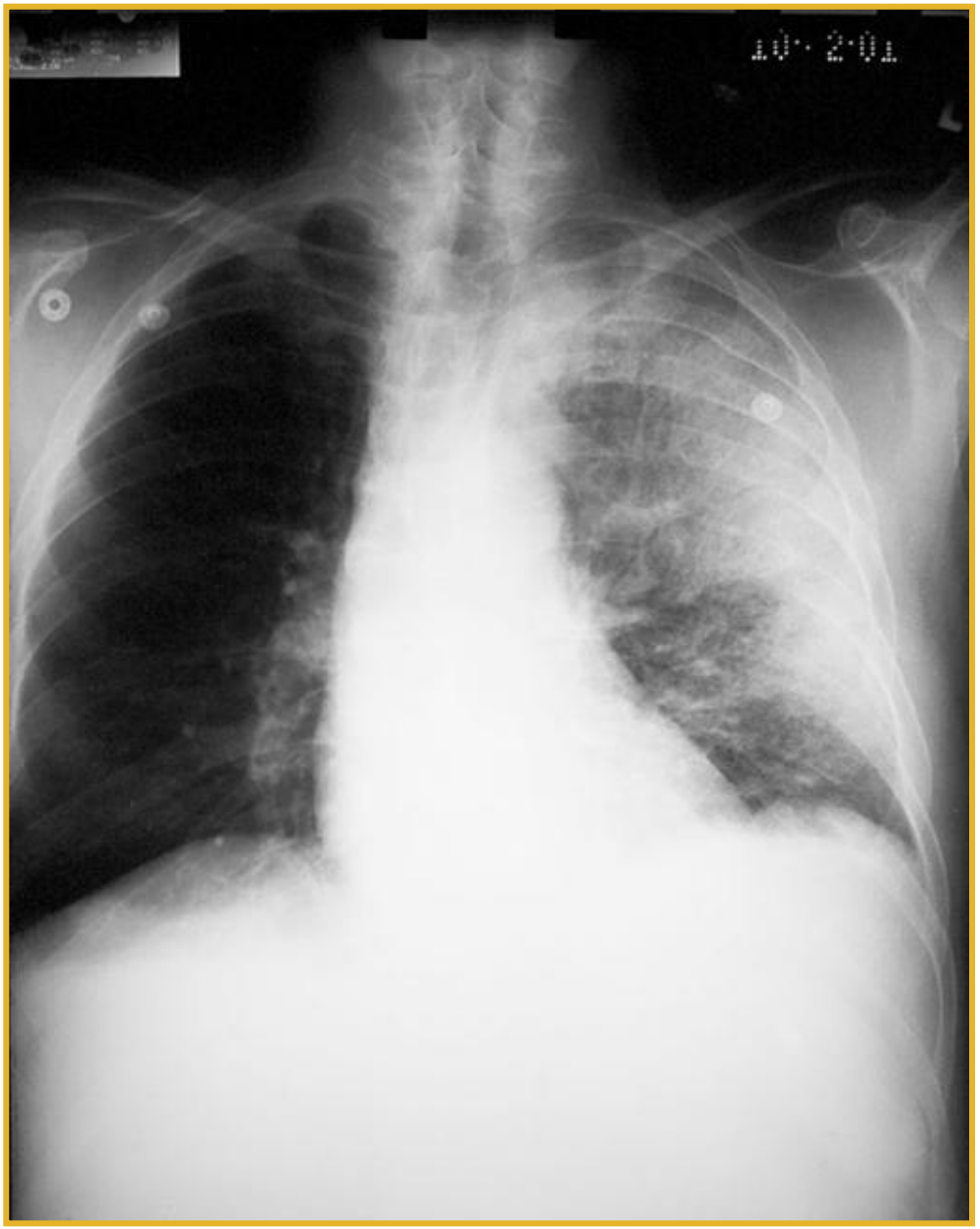
Inhalation means pulmonary
🤓🤓🤓

Inhalational Anthrax

- Incubation: 1 to 7 days
- Initial phase
  - Nonspecific (mild fever, malaise)
- Second phase
  - Severe respiratory distress
  - Dyspnea, stridor, cyanosis, mediastinal widening, death in 24 to 36 hours
- Case fatality: 75 to 90% (untreated)

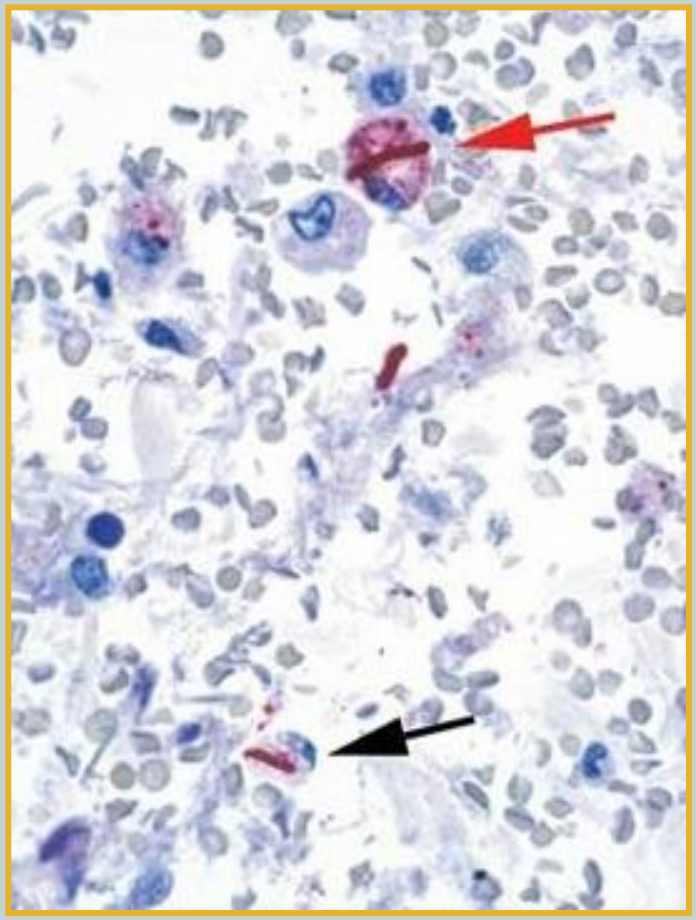
الأعراض بتبلش mild بعدين بتبلش تتطور لحتى توصل لمرحلة ال Respiratory distress - respiratory failure - عشان هيك لازم نحط المريض على ال respirator قبل ما يودّع

الاشي الواضح بالصورة ويلي بميز هو ال anthrax ال widding of mediastinum وبصير عندي infiltration for يعني the lungs الرئة بصير فيها سوائل بدل ال air sacs



Diagnosis in Humans

- Identification of *B. anthracis*
  - Blood, skin, secretions
- Culture
- PCR
- Serology
  - ELISA
- Nasal swabs
  - Screening tool



Treatment

- Penicillin
  - Most natural strains susceptible
- Additional antibiotic options
  - Ciprofloxacin
    - Treatment of choice in 2001
    - No strains known to be resistant
  - Doxycycline
- Course of treatment: 60 days

Prevention and Control

- Humans protected by preventing disease in animals
  - Veterinary supervision
  - Trade restrictions
- Improved industry standards
- Safety practices in laboratories
- Post-exposure antibiotic prophylaxis

اهم خطوة عشان نتجنب الإصابة هي ال vaccination بعدين يليها بالأهمية ال veterinary supervision

Vaccination

- Cell-free filtrate
- At risk groups
  - Veterinarians
  - Lab workers
  - Livestock handlers
  - Military personnel
- Immunization series
  - Five IM injections over 18-week period
  - Annual booster

Vaccine Side Effects

- Injection site reactions
  - Mild: 30% men, 60% women
  - Moderate:1 to 5%
  - Severe:1%
- Systemic effects rare
  - Muscle or joint aches, headache, rash, chills, fever, nausea, loss of appetite
- No long-term side effects noted

حكا الدكتور انصاب من البكتيريا و اعاني واموت ولا اتطعم
وحتى لو عانيت من الأعراض هتكون mild بالاغلب
لو كانت severe عاد عمر وخلص شو بدنا نعمل 😕💔

You are a hero just by reaching this slide 🤓❤️

اتمنى يكون التفريغ جيد 😂 موفقين باااشن واسفة ع اي خطأ غير مقصود... ❤️