GI

Microbiology

LEC no.

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The Brucellae, Leptospira and Mycobacterium of the GIT

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In this lecture, we will cover another group of bacteria that cause infections in the Gastrointestinal tract which are:

1- Brucella ➔ the causative agent of Brucellosis

2- Leptospira ➔ the causative agent of Leptospirosis

3- Mycobacterium ➔ which could also cause GIT infections: Abdominal TB

THE BRUCELLAE

- The brucellae are obligate parasites of animals and humans and are characteristically located intracellularly.
- They are relatively inactive metabolically. Brucella melitensis typically infects goats; Brucella suis, swine; Brucella abortus, cattle; and Brucella canis, dogs. Other species are found only in animals.
- Although named as species, DNA relatedness studies have shown there is only one species in the genus, B melitensis, with multiple biovars.
- The disease in humans, brucellosis (undulant fever, Malta fever), is characterized by an acute bacteremic phase followed by a chronic stage that may extend over many years and may involve many tissues.

The species within the Brucellae genus that cause diseases in Humans are four:

1. Brucella Melitenisis

   ➔ Most common cause of symptomatic Human brucellosis
   ➔ Preferred host for B. Melitensis are Goats & Sheeps
   ➔ Brucellosis is a zoonotic disease, meaning it infects animals and then transmitted to humans

2. Brucella Abortus

   ➔ Found in cattle, mainly cows
   ➔ From its name, it causes abortion in cows, however it does not cause abortion in infected pregnant women. The reason why is because the placenta of cattle contains Erythritol which is a growth factor for Brucella, but the human placenta does not contain Erythritol

3. Brucella Suis ➔ Preferred host are swine/pig

4. Brucella Canis ➔ Preferred host are dogs

The disease caused by the Brucella species is called Brucellosis, it has many other names with different contexts, and those include:

1. Undulant fever (الحمّى المتموّجة)

   ➔ This name describes the fever associated with the disease: it typically rises during the afternoon, then it drops during the night (where it's coupled with profuse sweating), and the next day it rises during the afternoon and so on.

2. Malta fever (الحمّى المالطية)

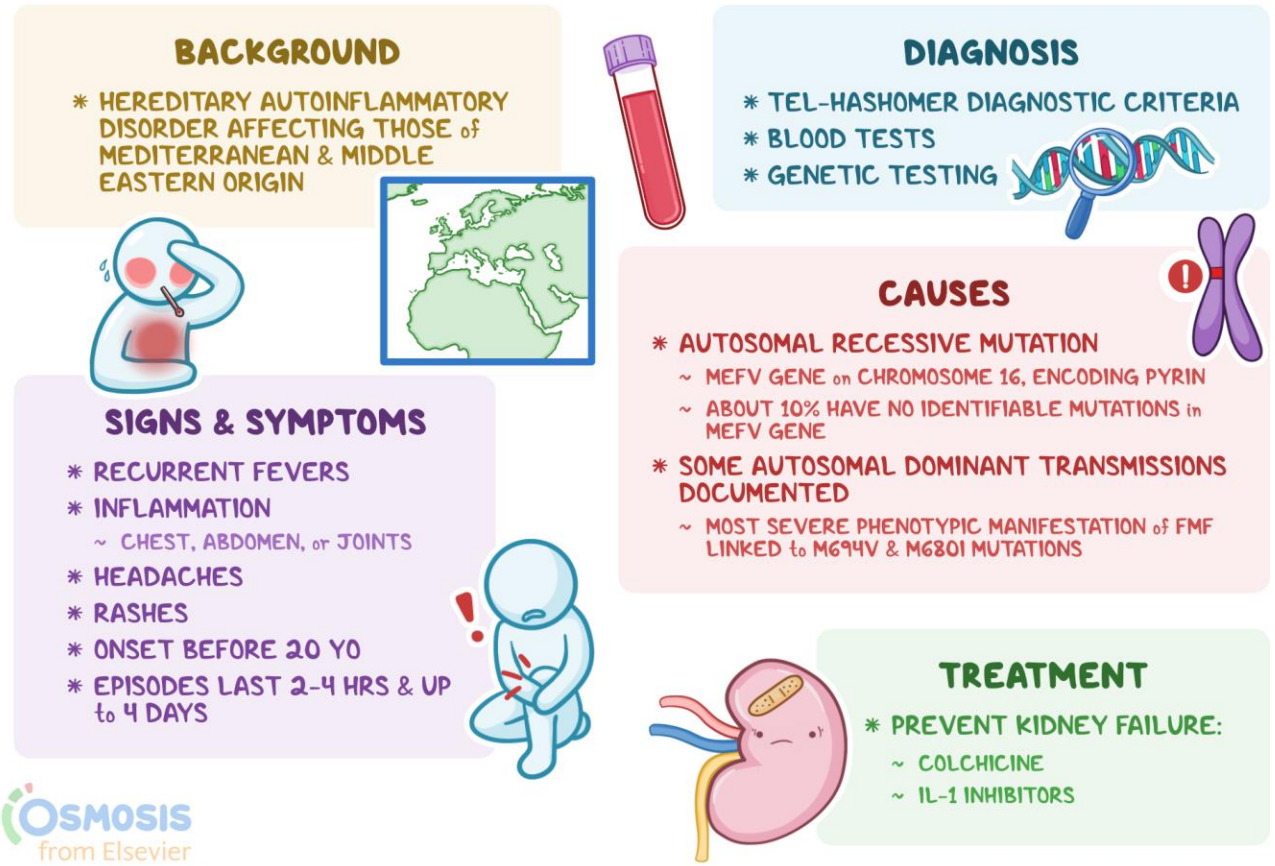
3. Mediterranean fever

   ➔ Which is different from Familial Mediterranean Fever. This is to show how Brucellosis was an endemic in the middle east including Jordan

4. Cyprus fever

Additional information: Familial Mediterranean Fever (FMF) is a genetic disorder that causes recurrent fevers, abdominal pain, and inflammation. It's linked to mutations in the MEFV gene, primarily affecting those of Mediterranean descent.

The picture on the right is also additional (للإستزادة!)



Morphology and Identification

- The appearance in young cultures varies from cocci to rods 1.2 µm in length, with short coccobacillary forms predominating. They are gram negative but often stain irregularly, and they are aerobic*, nonmotile, nonspore forming, **and non-encapsulated**

- Brucellae are adapted to an intracellular habitat, and their nutritional requirements are complex.

- Whereas B abortus requires 5–10% CO_2 for growth, the other three species grow in air.

- Catalase and oxidase are produced by the four species that infect humans. (positive)
- They are killed by boiling and pasteurization but are resistant to freezing and drying

*: Brucella species grow under aerobic conditions with the only exception of Brucella Abortus that require Capnophilic conditions

...Capnophilic: increased concentration of CO_2

➔ Although boiling and pasteurization kills the Brucella spp easily, a then-common mode of transmission in our region of Brucella spp such as Melitenisis was through drinking unpasteurized milk & other dairy products produced by cattle and goats infected with Brucella, as they are resistant to freezing & drying.

➔ Even though Brucella spp use carbohydrates, they do not ferment them nor produce acid or gas in sufficient amounts for classification.

Epidemiology

- Brucellae are animal pathogens transmitted to humans by accidental contact with infected animal feces, urine, milk, or tissues. The common sources of infection for humans are unpasteurized milk, milk products, and cheese as well as occupational contact (eg, farmers, veterinarians, and slaughterhouse workers) with infected animals. Cheese made from unpasteurized goat's milk is a particularly common vehicle for transmission of brucellosis.
- Brucellosis may be acquired by ingestion, inhalation, or mucosal or percutaneous exposure.
- Accidental injection of the live vaccine strains of B. abortus (S19 and RB51) and B. melitensis (Rev 1) can cause disease. B. melitensis and B. suis have historically been developed as biological weapons by several countries and could be exploited for bioterrorism.

➔Skin and Mucous membrane transmission of Brucella exposes health care workers to Brucellosis through, for example, needle stick injuries while taking blood samples from infected patients. Also, those working in slaughterhouses are at threat of infection with Brucella through skin abrasions and cuts that allow passage to the bacteria

➔ Splashes from samples taken from patients in the laboratory could also expose those working to infection

➔ Veterinarians can also contract Brucellosis through a needle stick injury through the live vaccine strains like REV 1 vaccine

➔ Transmission of Brucellosis via inhalation could be exploited for use in biological terrorism. The only recorded biological weapon that uses Brucella, especially B. Suis, was created by the USA, codenamed "The Menace." However, no use of Brucella in such nature was yet recorded.

➔ Human to Human transmission rarely occurs, except for two cases:

1. Vertical transmission: transmission from pregnant mother to susceptible fetus

2. Blood donations. Patients with Brucella are not allowed to donate blood

Pathogenesis

- Although each species of Brucella has a preferred host, all can infect a wide range of animals, including humans.
- The common routes of infection in humans are the intestinal tract (ingestion of infected milk), mucous membranes (droplets), and skin (contact with infected tissues of animals). Cheese made from unpasteurized goats' milk is a particularly common vehicle.
- The organisms progress from the portal of entry via lymphatic channels and regional lymph nodes to the thoracic duct and the bloodstream, which distributes them to the parenchymatous organs. Granulomatous nodules that may develop into abscesses form in lymphatic tissue, liver, spleen, bone marrow, and other parts of the reticuloendothelial system. In such lesions, the brucellae are principally intracellular.
- Osteomyelitis, meningitis, or cholecystitis also occasionally occurs. The main histologic reaction in brucellosis consists of proliferation of mononuclear cells, exudation of fibrin, coagulation necrosis, and fibrosis.
- The granulomas form and consist of epithelioid and giant cells, with central necrosis and peripheral fibrosis. **Cell-mediated immunity plays a big role in granuloma formation.**

Pathogenesis of Brucellosis is the same for all methods of transmission:

1. **Cross tissue barrier (skin, intestine, lung or liver)**

   ➔ The crossing of tissue barrier and into the bloodstream (bacteremia) causes the characteristic Undulant Fever of Brucellosis. The lipopolysaccharide (LPS) endotoxin activity plays a role in that.

2. **Get engulfed or actively internalized by phagocytes or APCs and multiply in them**

3. **Its main target is the Reticuloendothelial system where it circulates and establishes itself within it.**

   ➔ Manifestations in the RE system include abnormalities in its organs such as Spleen, Liver (Hepatosplenomegaly), and Lymph Nodes

4. **Other manifestations include those in the bone marrow (osteomyelitis) or meninges (meningitis) or Gallbladder (cholecystitis)**

Clinical Findings

- The incubation period ranges from 1–4 weeks. The onset is insidious, with malaise, fever, weakness, aches, and sweats.
- The fever usually rises in the afternoon; its fall during the night is accompanied by drenching sweat.
- There may be gastrointestinal and nervous symptoms. Lymph nodes enlarge, and the spleen becomes palpable. Hepatitis may be accompanied by jaundice.
- Deep pain and disturbances of motion, particularly in vertebral bodies, suggest osteomyelitis. These symptoms of generalized Brucella infection generally subside in weeks or months, although localized lesions and symptoms may continue.
- After the initial infection, a chronic stage may develop, characterized by weakness, aches and pains, low-grade fever, nervousness, and other nonspecific manifestations compatible with psychoneurotic symptoms.

➔ Chronic infection with brucellosis is a very debilitating condition with aches, pains and nervousness. For that reason, it's sometimes called "The Miserable Disease"

➔ As we mentioned earlier, Brucellosis (Mediterranean fever) was a very big endemic in our regions to the point where the simple signs of Fever & Abnormal Gait (way of walking) were diagnostic of being Brucellosis until proven otherwise.

➔ As much as was said about the findings, mainly 3 scenarios of Brucellosis are encountered:

1. Patient comes in with a similar or milder picture to Enteric (Typhoid) fever

2. Children patients usually present with Fever & Aute Monoarthritis, usually involving the Sacroilliac joint (which is the cause of the Abnormal Gait we mentioned)

3. Older patients present with Fever & Lower Back Pain due to Spondylitis of the spine. These patients are more than often the ones burdened with the "Miserable" feature of Brucellosis, with psychoneurotic symptoms such as nervousness and others.

Diagnostic Laboratory Tests

➔ **Brucella spp are fastidious bacteria: They require complex nutritional requirements, and they have a long incubation period (might take a month to see any growth on Brucella Agar)**

➢A. Specimens

- Blood should be taken for culture, biopsy material for culture (lymph nodes, bone, and so on), and serum for serologic tests.

➢B. Culture

- Brucella agar , specifically designed to culture Brucella species bacteria. The medium is highly enriched and—in reduced form—is used primarily in cultures for anaerobic bacteria.
- Brucella species bacteria grow on commonly used media, including trypticase-soy medium with or without 5% sheep blood, brain–heart infusion medium, and chocolate agar.
- The typical virulent organism forms a smooth, transparent colony; upon culture

➢C. Serology

➔ **Due to the fastidious nature of Brucella and prolonged culture growth times, Serology is relied upon for diagnosis**

- Immunoglobulin M (IgM) antibody levels rise during the first week of acute illness, peak at 3 months, IgG and IgA antibody levels rise about 3 weeks after onset of acute disease, peak at 6–8 weeks, and remain high during chronic disease.

❑Agglutination test : IgG agglutinin titers above 1:80 indicate active infection. Individuals injected with cholera vaccine may develop agglutination titers to brucellae.

❑ELISA assays— IgG, IgA, and IgM antibodies may be detected using enzyme-linked immunosorbent assay (ELISA), which use cytoplasmic proteins as antigens. These assays tend to be more sensitive and specific than the agglutination test especially in the setting of chronic disease.

➔ **The combination of both Agglutinating and non-Agglutinating tests (ELISA) is used for diagnosis. In Agglutinating, the antigen being looked for is the Brucella Smooth Antigen (S). While non-Agglutinating use cytoplasmic proteins**

➔ A common problem that might hinder diagnosis through serology by giving false negative to people who have a highly suggestive clinical picture of Brucellosis is the "Blocking Antibody" phenomenon, this is troubleshooted by doing one more step in the ELISA which is adding anti-human anti-globulins , overcoming the blocking antibody

➔ Serology fails to diagnose Brucella Canis

Explanation of Blocking Antibody phenomenon: In this phenomenon, the immune system produces antibodies that bind to the antigens which the serology tests look for. When those specific antibodies bind to the antigen, they exhibit a 'blocking' feature which doesn't allow other antibodies to bind to it, and therefore the ELISA test will come back as false negative.

Treatment & Immunity

- Brucellae may be susceptible to tetracyclines, rifampin, trimethoprim–sulfamethoxazole, aminoglycosides, and some quinolones. Symptomatic relief may occur within a few days after treatment with these drugs. However, because of their intracellular location, the organisms are not readily eradicated completely from the host.

- For best results, treatment must be prolonged. Combined treatment with a tetracycline (eg, doxycycline) and either streptomycin for 2–3 weeks or rifampin for 6 weeks is recommended.

➔ Doxycycline 100 mg twice every day, Rifampin 1 gram once every day for 6-8 weeks.

➔ The aggressive antibiotic use is due to the intracellular nature of this infection

Prevention, and Control

- Eradication of brucellosis in cattle can be attempted by test and slaughter, active immunization of heifers with avirulent live strain 19, or combined testing, segregation, and immunization. Cattle are examined by means of agglutination tests.
- Active immunization of humans against Brucella infection is experimental.
- Control rests on limitation of spread and possible eradication of animal infection, pasteurization of milk and milk products, and reduction of occupational hazards wherever possible.

➔ **No vaccine for Humans against Brucella, but there is one for Cattle.**

Leptospira

- Traditionally, the genus Leptospira comprised two species: the pathogenic L. interrogans and the free-living L. biflexa, now designated L. interrogans sensu lato and L. biflexa sensu lato, respectively.

- Leptospirosis; The disease is caused by pathogenic Leptospira species and is characterized by a broad spectrum of clinical manifestations, varying from asymptomatic infection to fulminant, fatal disease (Weil's Syndrome).

- Kidney involvement in many animal species is chronic and results in the shedding of large numbers of leptospirae in the urine; this is probably the main source of environmental contamination resulting in infection of humans.

- Human urine also may contain spirochetes in the second and third weeks of disease.

➔ The disease caused by Leptospira is Leptospirosis, which manifests as:

1. Asymptomatic or Mild disease in 90% of those infected
2. Weil's syndrome in less than 10% of those infected

➔ The species that cause Leptospirosis are L. Interrogans & L. Biflexia

➔ Weil's syndrome/disease is characterized by being "Biphasic" (similar to Brucella):

1. Leptospiremic phase is when the species are circulating in the bloodstream
2. Parenchymatous phase (or the Immune phase) is when they establish themselves in a parenchymatous organ, usually the liver or kidney

➔ People with Weil's syndrome present with a triad of Jaundice, Hemorrhage (lung mainly or other organs), and blood urea nitrogen retention. (More simply put as Hepatitis, Nephritis, and Hemorrhage)

➔ **Small point about transmission:**

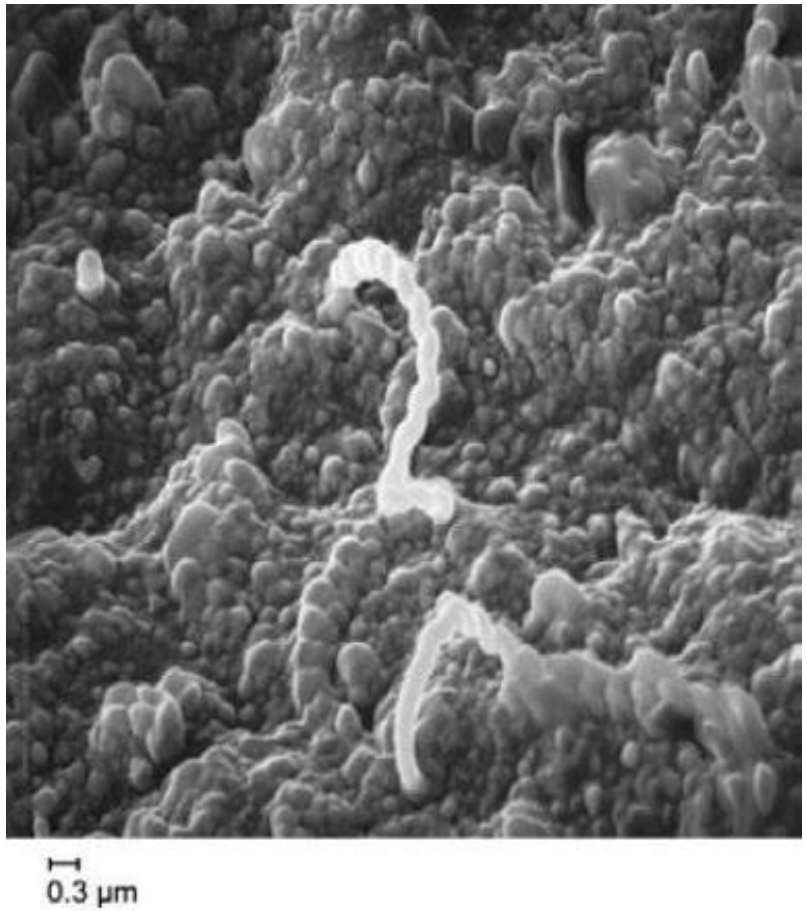
Most of the GIT infections we took during this course have been related to the feco-oral and oral-oral mode of transmission, while in Leptospirosis it's Urine exposure

The animal reservoir such as rodents, rats, mice as well as cattle, dogs and sheep commonly contract leptospirosis in the kidney, and they shed the species in their urine.

Leptospira interrogans

➔ **Other spirochetes we took were Treponema Pallidum (Syphilis) and Borrelia Burgdorferi (Lyme disease)**

- Leptospirae are tightly coiled, thin, flexible spirochetes 5–15 µm long, with very fine spirals 0.1–0.2 µm wide; one end is often bent, forming a hook (question mark appearance). They are motile. They have double membranes
- They are actively motile with two periplasmic flagella, which is best seen using a dark-field microscope.
- Leptospirae derive energy from oxidation of long-chain fatty acids and cannot use amino acids or carbohydrates as major energy sources. Ammonium salts are a main source of nitrogen.
- Leptospirae can survive for weeks in water, particularly at alkaline pH.



Epidemiology

- Leptospirosis has a worldwide distribution but occurs most commonly in the tropics and subtropics because the climate and occasionally poor hygienic conditions favor the pathogen's survival and distribution.

- Current information on global human leptospirosis varies but indicates that approximately 1 million severe cases occur per year, with a mean case–fatality rate of nearly 10%.

- The vast majority of infections with Leptospira cause no or only mild disease in humans. A small percentage of infections (~1%) lead to severe, potentially fatal complications.

Pathogenesis

➔ **Rarely ingestion and inhalation. Occupations with Urine exposure such as sewage workers are at risk.**

- Transmission occurs through cuts, abraded skin, or mucous membranes, especially the conjunctival and oral mucosa. After entry, and an incubation period of 1–2 weeks the organisms proliferate, cross tissue barriers, and disseminate hematogenously to all organs (leptospiremic phase)

- They then establish themselves in the parenchymatous organs (particularly liver and kidneys), producing hemorrhage and necrosis of tissue and resulting in dysfunction of those organs (jaundice, hemorrhage, nitrogen retention).

➔ **The paranchymatous phase/immune phase coincide with the disappearance of Leptospira from blood, leading to negative blood cultures during it.**

its commonly a chronic animal infection (accidental infection in humans)

Clinical Findings

➔ **Manifests at first as a Flu-like illness or Aspetic meningitis**

- The illness is often biphasic. After initial improvement, the second phase develops when the IgM antibody titer rises. It manifests itself often as “aseptic meningitis” with an intense headache, stiff neck, and pleocytosis of the CSF.

- Nephritis and hepatitis may also recur, and there may be skin, muscle, and eye lesions. The degree and distribution of organ involvement vary in the different diseases produced by different leptospirae in various parts of the world.

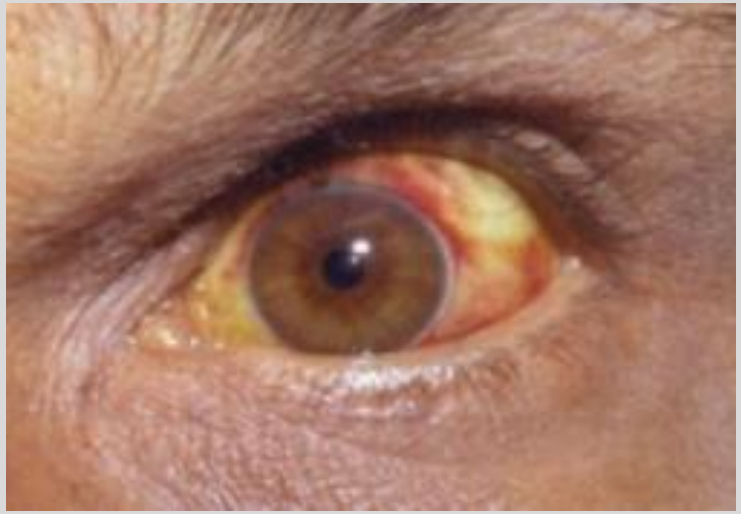
- Human urine also may contain spirochetes in the second and third weeks of disease.

- Many infections are mild or subclinical. Hepatitis is frequent in patients with leptospirosis.

➔ Another prominent characteristic of Leptospirosis (seen in sewage workers) is conjunctival suffusion, characterized by eye redness without exudate

➔ The image on the right shows what that looks like with the Jaundice that occurs with Leptospirosis (خوفتني شوي صراحة)

➔ Myalgia, Nausea, Vomiting are also seen in Leptospirosis and usually with NO fever (afibrile illness)



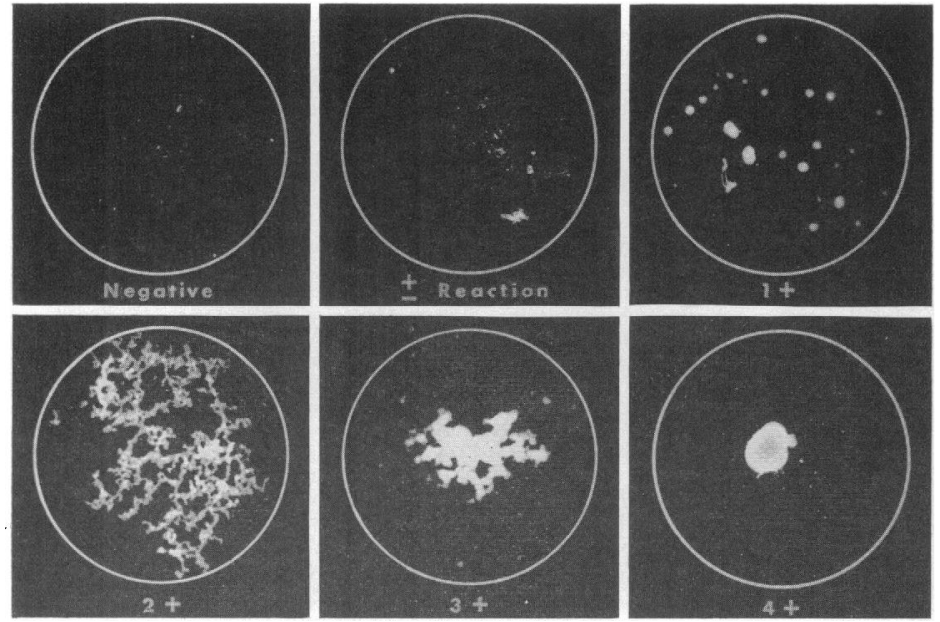
Diagnostic Laboratory Tests

- A. Specimens
- Specimens consist of blood, CSF, or urine and tissues for microscopic examination and culture.
- B. Microscopic Examination
- Dark-field examination or thick smears stained by the Giemsa technique.
- C. Culture
- Whole fresh blood ,CSF or urine or crushed tissue can be cultured. Leptospires grow best under aerobic conditions at 28–30 C in semisolid medium (eg, Ellinghausen-McCullough-Johnson- Harris EMJH) in 10 mL test tubes with 0.1% agar and 5-fluorouracil.
- Growth is slow, and cultures should be kept for at least 8 weeks.
- D. Serology
- The diagnosis of leptospirosis in most cases is confirmed serologically with microscopic agglutination test (MAT) and ELISA.

➔ Specimens taken from blood, CSF, or urine are examined to notice the characteristic question mark/hook appearance of Leptospira

➔ Cultures here can also take a long time to show results with up to 2 months to see growth on EMJH agar. Therefore, we rely less on Cultures for diagnosis but more on the MAT test (Microscopic Agglutination Technique)

Additional information: this is what the MAT test looks like:



Treatment &Immunity

- Treatment of mild leptospirosis should be with oral doxycycline, ampicillin, or amoxicillin.
- Severe leptospirosis should be treated with IV penicillin as soon as the diagnosis is considered.
- Serovar-specific immunity follows infection, but reinfection with different serovars may occur.

★ A question that should occur to you is why we are using Penicillin (which works by inhibiting cell-wall synthesis) to eliminate Gram-Negative bacteria? Answer is in the next slide

Prevention, and Control

★ **Because Leptospira contain penicillin-binding proteins**

- Leptospirae is excreted in urine both during the active illness and during the asymptomatic carrier state.
- Leptospirae remain viable in stagnant water for several weeks; drinking, swimming, bathing, or food contamination may lead to human infection. Persons most likely to come in contact with water contaminated by rats (eg, miners, sewer workers, farmers, and fishermen) run the greatest risk of infection
- Avoidance of exposure to urine and tissues from infected animals through proper eyewear, footwear, and other protective equipment. Targeted rodent control strategies could be considered.
- Vaccines for agricultural and companion animals are generally available, and their use should be encouraged.

➔ **No vaccine has yet been made for humans**

Mycobacterium Tuberculosis (Mtb)

- It was not until the 19th century, when Robert Koch utilized s new staining method (ZN stain) and applied it to sputum from patients discovering the causal agent of the disease Tuberculosis (TB); Mtb or Koch bacillus.
- Tuberculosis , consumption(consume patients ,weight loss), white plaque (extreme pallor seen among patients)
- The family mycobacterium tuberculosis complex(MTC) can cause Tuberculosis (TB) in humans and other livings.
- It includes M. tuberculosis (Mtb), Mycobacterium africanum, Mycobacterium bovis, Mycobacterium microti, Mycobacterium caprae, Mycobacterium pinnipedii, Mycobacterium suricatte, Mycobacterium mungi, Mycobacterium dassie, Mycobacterium oryx and Mycobacterium canetti.
- Mtb is a slow growing ,obligate aerobe, facultative intra- cellular bacterium .
- Non-spore forming, non-motile acid-fast bacilli.

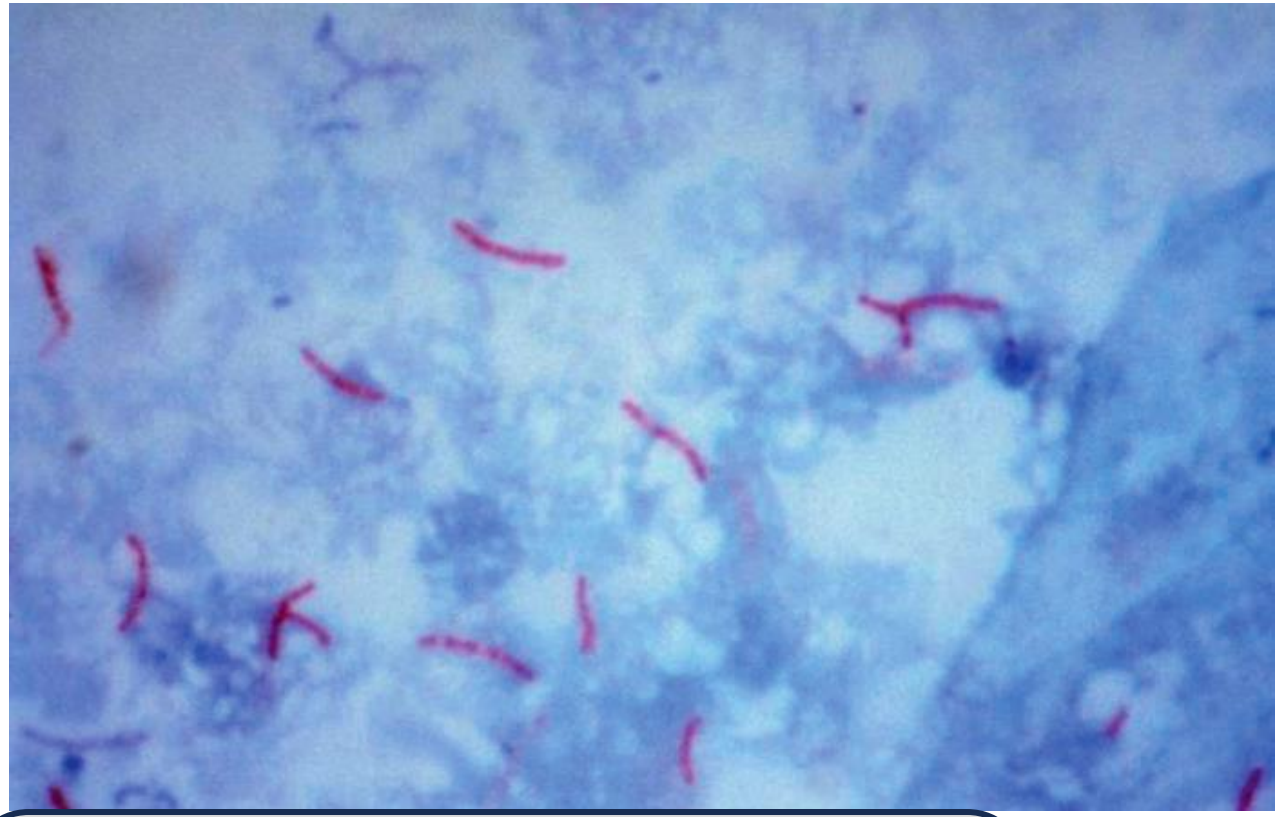
Overview: the last topic of this lecture is the Mycobacterium tuberculosis (MTB) –mainly- or consumption ((consume patients, significant weight loss in a short duration)and white plaque (extreme pallor seen among patients because of hypoxemia and chronic productive cough, and our concern on this lecture will be on the abdominal tuberculosis; as it is an important disease that dominated the human.

Mainly MTB is the principal member for tuberculosis, but any member of mycobacterium tuberculosis complex. They vary in geographical distribution, virulence, severity of disease. In addition, Mycobacterium bovis was a common cause of tuberculosis in humans specifically abdominal tuberculosis. The term bovis came from bovine," which refers to cattle; ingestion of unpasteurized milk and cheese made from unpasteurized milk can cause this infection.

Moreover, the only vcaccine for tuberculosis (BCG vaccine) is live attenuated mycobacteria bovis.

Tuberculosis is slowly growing bacteria; The growth rate is much slower than that of most bacteria. The doubling time of tubercle bacilli is about 24 hours. And its agar needs 8 weeks of incubation to be discarded as negative culture. They are acid fast not gram + nor gram - , obligate aerobe, facultative intra- cellular bacterium, Non-spore forming, non-motile bacilli.

Mycobacterium Tuberculosis (Mtb) staining



Here in ZN stain or Acid-fast stain, we use a red or pink pigment called carbonfyoxin stain, then we heat the sample to aid the penetration of the dye through the cell wall of MTB, which is of a high-level content of lipid. Then we treat it with acid alcohol which will remove ANY dye from the bacteria UNLESS it's acid-fast. So if we put methylene blue for example, it won't dye the sample since we have already used the the red and the acid-fast stain on the sample.

note that weight loss accompanying TB means that this disease always a differential diagnosis to all cancer types (similar presentation)

بنستنتج انه اسم الصبغة FAST-acid ألنه بتخلي العيّنة تصوم عن الصبغات الثانية (بتمنعها). يعني العيّنة انصبغت باللون االحمر أو الزهري وطالما استخدمنا ال fast-acid بعد هيك، رح يثبت اللون الزهري ولو حطينا صبغة تانية بعد هيك ما رح تشتغل (بتصير زي مانع أو درع)

Epidemiology

- Two TB-related conditions exist; latent TB infection (LTBI) and TB disease. If not treated properly, TB disease can be fatal. People who have latent TB infection do not feel sick, do not have any symptoms, and cannot spread TB to others

- About one third of the worlds population is infected with TB bacteria (TB latency).

- However, only small proportion of those infected will become sick with TB.

- TB remains a leading cause of infectious diseases morbidity and mortality. In 2015, an estimated 10.4 million new TB cases were seen world wide.

- TB is considered an airborne infectious disease although M. tuberculosis complex organisms can be spread through un-pasteurised milk, direct inoculation and other means.

There 2 clinical entities after the infection of MTB: 1. Active TB: acute inflammation, development of signs and symptoms 2. Latent TB: they have it but they don't feel sick or show symptoms or signs, NOT CONTAGIOUS. but once the host becomes immunocompromised, they might develop secondary reactivation for TB.

يعني الناس اللي انصابوا أول مرة ----> TB primary،
وثاني مرة ----> secondary TB

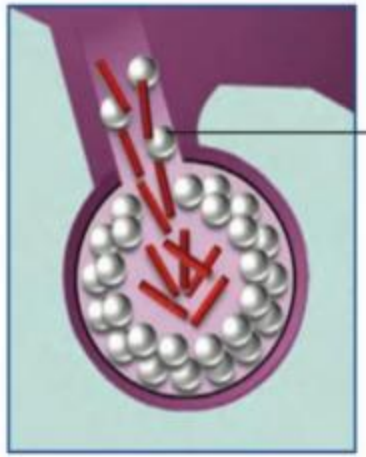
- The "10/3/1" formula has high consensus and is widely accepted, as it means that: Out of 10 people exposed to mycobacterium tuberculosis
- 3 of them will go through LTBI (Latent Tuberculosis Infection)
- and 1 out of 10 exposed people is going to develop active tuberculosis from the first exposure.
- Notice that there are 6 people left —out of the 10 exposed people— who somehow cleared the infection by their innate or adaptive immunity. However, there is a big debate regarding mycobacterium tuberculosis that our body cannot get rid of it and it remains present in our body as we don't have a sterilizing immunity against it.

This 1 who developed tuberculosis 70-80 % of these cases it will be pulmonary tuberculosis involving the lung while the 10-20 % of the cases cause any organ in the body (extrapulmonary TB). It can infect brain, heart, pleura and the abdomen.

Abdominal tuberculosis can be caused by different ways:

1. By Miliary TB; it a potentially fatal form of disseminated TB characterized by millet-seed-like granuloma formation in various organs. Miliary TB often arises from a primary pulmonary infection that spreads hematogenously or by lymphatics causing abdominal tuberculosis.
2. Swallowing the sputum that has active TB from the pulmonary type.
3. Direct extension: Like if it was unpasteurized milk or places near abdomen tuberculosis, like fallopian tube as well as the spine.(Potts disease)

Granuloma ruptures and TB enters lymphatics and blood circulation



Tuberculosis TB

- The primary site of TB is usually lung, from which it can get disseminated into other parts of the body. The other routes of spread can be contiguous involvement from adjacent tuberculous lymphadenopathy or primary involvement of extrapulmonary organ.
- TB bacteria can attack any part of the body such as the pleura ,L.N. ,pericardium, kidney, spine, brain and abdomen (abdominal Tuberculosis) collectively known as extrapulmonary TB.
- The abdominal TB, which is not so commonly seen as pulmonary TB, can be a source of significant morbidity and mortality and is usually diagnosed late due to its nonspecific clinical presentation.
- The abdominal TB usually occurs in four forms: tuberculous lymphadenopathy, peritoneal tuberculosis, gastrointestinal (GI) tuberculosis and visceral tuberculosis involving the solid organs

The primary site of TB is usually respiratory route (Pulmonary TB, the nuclei get regenerated whilst talking/singing/coughing), from which it can get disseminated into other parts of the body. The other routes of spread can be infectious from adjacent tuberculous lymphadenopathy or primary involvement of extrapulmonary organ

The abdominal TB usually occurs in four forms: 1. tuberculous lymphadenopathy (mesenteric lymph nodes), 2.peritoneal tuberculosis, 3.gastrointestinal (GI) tuberculosis(involving more than 1 area in GIT, from oropharynx to the anus , may cause oropharangeal TB) and 4.visceral tuberculosis (involving the solid organs like spleen, kidney & Liver).

Gastrointestinal (GI) tuberculosis pathogenesis

- Abdominal tuberculosis (TB) includes involvement of the gastrointestinal tract, peritoneum, lymph nodes, and/or solid organs .Abdominal TB comprises around 5 percent of all cases of TB
- Tuberculosis of the abdomen may occur via reactivation of latent TB infection or by ingestion of tuberculous mycobacteria (as with ingestion of unpasteurized milk, or sputum or undercooked meat). In the setting of active pulmonary TB or miliary TB, abdominal involvement may develop via hematogenous spread via contiguous spread of TB from adjacent organs (such as retrograde spread from the fallopian tubes) or via spread through lymphatic channels
- The mucosal layer of the GI tract can be infected with the bacilli with formation of epithelioid tubercles in the lymphoid tissue of the submucosa. After 2-4 wk, caseous necrosis of the tubercles leads to ulceration of the overlying mucosa which can later spread into the deeper layers and into the adjacent lymph nodes and into peritoneum. Rarely, these bacilli can enter into the portal circulation or into hepatic artery to involve solid organs like liver, pancreas and spleen

Gatrointestinal TB is uncommon as comprises only 5-10% of all cases of active TB. The whole pathology of mycobacterium tuberculosis infection is the granuloma formation.

Granuloma is a dynamic structure which is formed as a response of the immune system to contain intracellular infection, which might heal spontaneously or the condition might elevate, and caseous necrosis might occur through which immune cells in the granuloma rupture and the mycobacteria leave the granuloma and spread to invade adjacent structures.

Gastrointestinal TB clinical finding

- The clinical presentation tends to be non-specific, with abdominal pains and general complaints.

- Although any portion of the gastrointestinal tract may be affected, the terminal ileum and the cecum are the sites most commonly involved. Abdominal pain (at times similar to that associated with appendicitis) and swelling, obstruction, hematochezia, and a palpable mass in the abdomen are common findings at presentation. Fever, weight loss, anorexia, and night sweats are also common.

Clinically: It is not easy to diagnose someone with tuberculosis, as there is a lot of dilemma in the diagnosis, but usually, the patients share a few symptoms or signs. The symptoms mentioned above are associated with pulmonary tuberculosis; which is the most common clinical form and occurs in 85% of the cases of tuberculosis. If the tuberculosis is extrapulmonary; then the symptoms of the disease depend on the infected organ.

But the most common symptom in Abdomen is palpable mass and abdominal pain and hematochezia. Palpable mass can felt in the ileocecal junction.

Laboratory diagnostic methods

❖Smear microscopy

- Three specimens from each patient with suspected TB should be examined microscopically for Acid Fast Bacilli AFB (classically Ziehl-Neelsen) or mycobacteria can be demonstrated by yellow fluorescence after staining with auramine **(auramine staining).**

❖Culture

- Both liquid and solid mycobacterial cultures should be performed for every specimen, and recovered isolates should be Caccording to standard criteria (Lowenstein-Jensen or Middlebrook 7H10), Radiometric broth culture (BACTEC radiometric system). mycobacterial growth indicator tube (MGIT).
- Culture for acid fast bacilli is the most specific test for TB and allows direct identification and determination of susceptibility of the causative organism

❖A nucleic acid amplification test (NAAT), Tuberculin skin tests (TSTs), Interferon-gamma release assays (IGRAs) are commonly used as well.

Culture:

1. Semisynthetic agar media (semi-solid)— These media (eg, Middlebrook 7H10 and 7H11) contain defined salts, vitamins, cofactors, oleic acid, albumin, catalase, and glycerol .
2. Inspissated egg media (solid)— These media (eg, Löwenstein-Jensen) contain defined salts, glycerol, and complex organic substances (eg, fresh eggs or egg yolks, potato flour, and other ingredients in various combinations.
3. Broth media (fluid)— (eg, Middlebrook 7H9 and 7H12) support the proliferation of small inoculate.

The issue with culturing is that it takes quite a long time.

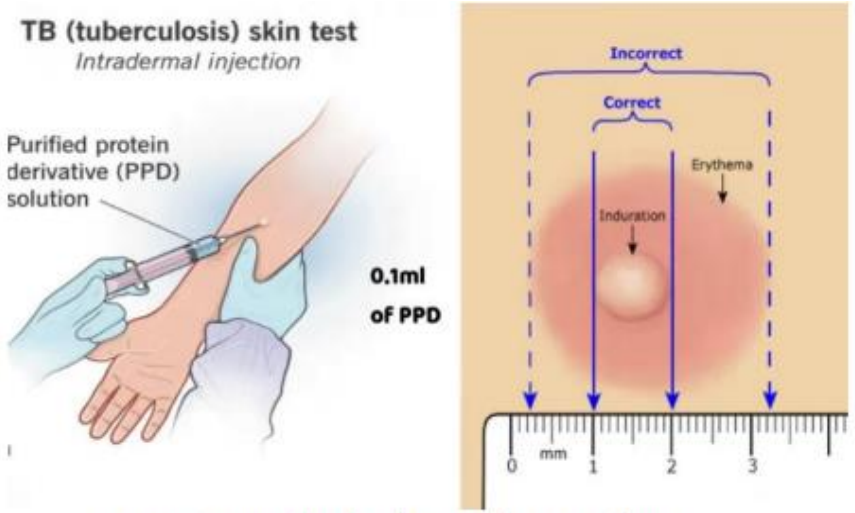
Imaging could also be used in the diagnosis of tuberculosis, such as CT or X-ray imaging which allow you to observe cavitary lung lesions in the cases of tuberculosis.

We use multiple methods instead of 1 in the diagnosis of tuberculosis + high index of clinical suspicion.

Tuberculin skin tests (TSTs):

- We take virulence factors from the mycobacteria, which are called PPD's (Purified Protein Derivatives) that are derived from the mycobacteria's cell wall.
- Then, we inject them intradermally, and we ask the patient to come back after 48 hours to measure the induration
- Then, we use a ruler to measure the diameter of the induration. And there is category for each induration.
- TST only tells if the tested person is exposed or has been previously exposed to Mycobacterium tuberculosis
- The issue with the TST is that it gives false positive results many times, as it may result in a positive result without the tested person being actually infected, and that is because:

1) The tested person is vaccinated with BCG
2) Environmental Mycobacteria (NTM's) contain mycobacterial antigens which could also lead to false positive TST.



Extra image for further understanding

Interferon-gamma release assays (IGRAs):

- **there are antigens specific only for Mycobacterium tuberculosis and mainly there is two of them found in the cell wall of Mycobacterium tuberculosis:**
  1) Culture filtrate protein (CFP-10/10KDa).
  2) Early secretory antigenic target (ESAT-6/6Kda).
- **Secondly, we put the antigen in the blood sample. If the monocytes in the blood sample recognize the Mycobacterium tuberculosis specific antigens, they will starting pouring huge amounts of interferon-gamma (IFN-gamma).**
- **We have a certain cutoff for IFN-gamma levels,**

if the IFN-gamma levels are below it → negative results
if the IFN-gamma levels are above it → positive results

- **It is very Important to know that whether the test is TST or IGRA, they both only tell us if there was a past exposure to Mycobacterium tuberculosis.**
- **Through TST and IGRA, we can't know if the infection occurred before or it is occurring now and in the case of current infection we can't know if it is in the latent or active phase.**

Treatment

- The course of TB treatment depends on whether the individual is in the latent or active stage, and on his or her probability of risk.

- Treatment of TB usually involves a drug cocktail, or a mixture of multiple drugs, with an intensive initial 2-month phase followed by a slower 4- to 6-month continuation phase the main anti-tuberculosis drugs used in the chemotherapy of TB are: isoniazid (INH), rifampin (RIF), pyrazinamide (PZA), and either ethambutol (EMB) or streptomycin (SM).
- Isoniazid preventive therapy IPT is the recommended treatment for LTBI but the regimen's main drawback is the duration of therapy

Treatment of tuberculosis takes about 6 months. The first 2 months are called intensive phase and rest 4 months are called continuation phase.
In the intensive phase ---> the patient takes 4 drugs isoniazid (INH), rifampin (RIF), pyrazinamide (PZA), and ethambutol (EMB) .
In the continuation phase --->the patient takes 2 drugs isoniazid (INH), rifampin (RIF)

Prevention

- The best way to prevent TB is to diagnose and isolate infectious cases rapidly and to administer appropriate treatment until patients are rendered noninfectious (usually 2–4 weeks after the start of proper treatment) and the disease is cured.

Usually in the past, the patient when diagnosed with TB they would sit in quarantine. They would sit for 3 weeks.

- Additional strategies include BCG vaccination and treatment of persons with LTBI who are at high risk of developing active disease.

- Mycobacterium bovis Bacillus Calmette–Guérin (BCG), an attenuated vaccine derived from M. bovis **(it is propagated in the laboratory and they lost their virulence factor, so they lose their ability to cause disease but they retained their immunogenicity)**, is the only licensed vaccine against tuberculosis (TB)

"Calmette" and "Guérin" are two French scientists who studied and worked on Mycobacterium bovis which causes bovine tuberculosis and it used to be transmitted to humans via milk before pasteurization.

The issue with the BCG is its low efficacy. Furthermore, It is mentioned in the books & articles that the efficacy of BCG is from 0% to 80% which means that out of 100 vaccinated people, the worst chance is that no one out of them is actually going to be protected, and the best chance is that 80 people out of the 100 vaccinated people are going to be actually protected. And that is why they stopped giving it in Europe and the USA. ممكن ٨٠ واحد يستفيد وممكن ولا واحد

However, BCG is still given in developing countries; because we believe that this way, we are protecting the children from the most severe forms of tuberculosis, which are tuberculous meningitis and miliary tuberculosis.

نرجوا منكم يا أفاضل بعد الدعاء لنا أن تدخلوا لهذا الرابط وتقوموا بتزويدنا بتغذية راجعة عن الشيتات والموديفايدز:

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The End