

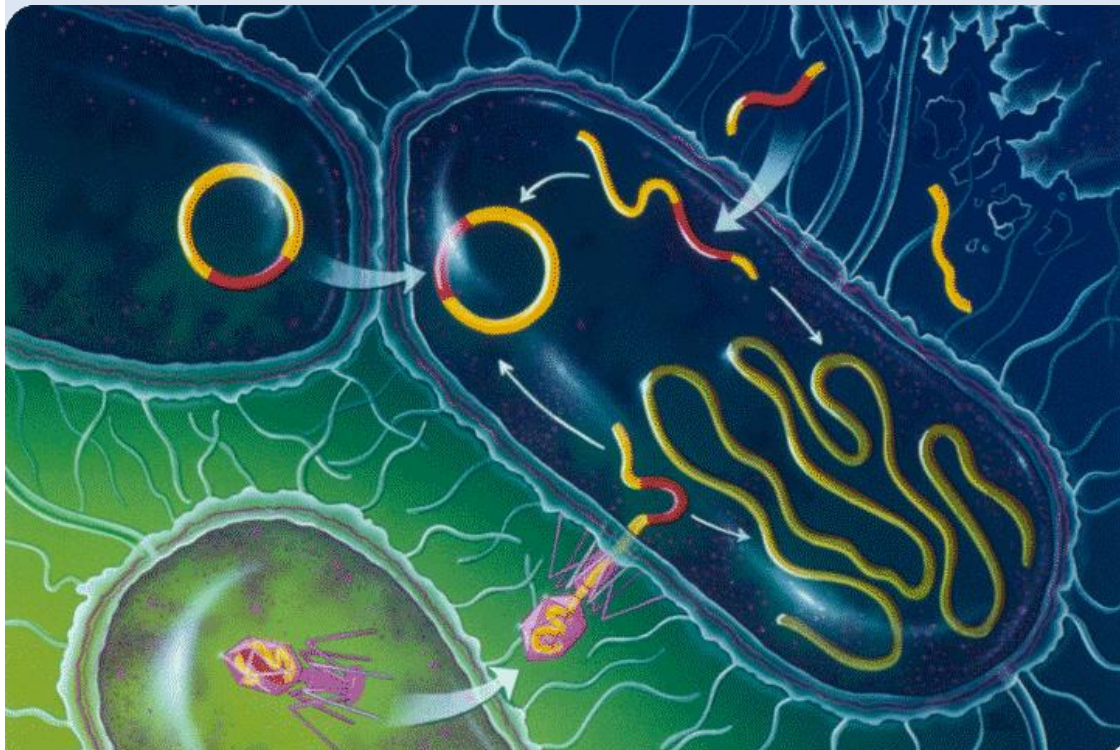
431 *Microbiology Team*

Chlamydia, Syphilis & Gonorrhea

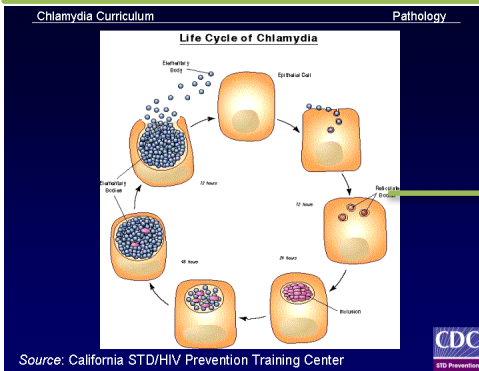
Leaders:

Faisal Al Rashed & Eman Al-Shahrani

Done By: Faisal Al Rashed, Sama Al Ohali



- The Genus **Chlamydia** is divided into 3 species: **C.trachomatis**, **C.psittaci**, and **C.pneumoniae**. **C.trachomatis** infections cause diseases of the genitourinary tract and the eye, including **trachoma, which is a major cause of blindness**. **C.psittaci** and **C.pneumoniae** infect various levels of the respiratory tract. **C.trachomatis** is divided into a number of **serotypes**, which correlate with the clinical syndrome they cause → **A-C, D-K, and L1-3**.
- **Chlamydia** have no peptidoglycan or mumaric acid in its cell wall → therefore, **it can not be stained with gram stain, nor can be treated with B-lactams**.
- β-Lactam antibiotics are bactericidal, and act by inhibiting the synthesis of the peptidoglycan layer of bacterial cell walls.
- Chlamydiae possesses ribosomes and synthesize their own proteins and, therefore, are **sensitive to antibiotics that inhibit this process, such as tetracycline (Doxycycline) and macrolides (Azithromycin, Erythromycin)** → they are all protein synthesis inhibitors.



Pathogenesis: chlamydiae have a unique life cycle, with morphologically distinct infectious and reproductive forms. The extracellular **infectious form, the elementary body**, can survive extracellular cell-to-cell passage. Once it is inside the host cell, the particle reorganizes into a reticulate body, which become metabolically active and divides repeatedly within the cytoplasm of the host cell. **As they divide, the reticulate bodies form an inclusion body**. After that, multiplication stops and all the reticulates become new infectious elementary bodies. They are then released from the cell, ending in host cell death. **(These Inclusions can be stained with Geimsa or Iodine stain!!)**

Chlamydia

- Chlamydia Spp. Are obligatory intracellular organisms → because they uses host cell metabolism for growth and replication. They don't have a rigid cell wall. Therefore, **Beta lactam antibiotics cannot be used to treat Chlamydia infections**.
- Fail to grow on artificial media; therefore, they cannot be isolated in cultures.

	Species	Disease
Chlamydia	C. trachomatis Serotype A, B, C	Trachoma (eye infection, causes Blindness)
	C. trachomatis Serotype D, K (the one associated with STDs)	Inclusion conjunctivitis, Genital infection (=NGU*)
	C. trachomatis Serotype L1, L2, L3 (not common in our area)	Lymphogranuloma venerum
	C. psittaci	Psittacosis (from parrots)
	C. Pneumoniae	Respiratory infection

Psitacosis (=ornithosis): a zoonotic disease that is transmitted to humans by inhalation of dust contaminated with respiratory secretions of feces of infected birds. The human disease usually targets the lower respiratory tract. There is an acute onset of fever, dry cough, and flulike symptoms, with hepatomegaly.

*NGU= non-gonorrheal urethritis.

The most common cause of urethritis is gonorrhea.
The most common cause of non-gonorrheal urethritis (NGU) is *C. trachomatis*

- Chlamydia Trachomatis is a common cause of sexually transmitted disease (STD).
- Spread by genital secretions, anal or oral sex.
 - Wide spread, 5-20 % among STD clinic in USA.
- Human are the sole reservoir. (It is completely a human disease, unlike *C. psittaci*)
- NGU: in males, the urethra is the principal locus of the infection. Females may present with cervicitis and/or urethritis. **Contiguous spread of infection may involve the epididymis in men and fallopian tubes "salpingitis" and adjacent tissues in women (Pelvic Inflammatory Disease).**
- Chlamydial NGU is symptomatically similar to infections caused by *N. gonorrhoeae*. Although the incubation time is longer (2-3wks) with chlamydia and the discharge tends to contain fewer pus cells.
 - Repeated or chronic episodes may lead to sterility in both sexes and to ectopic pregnancies

Infants born to genitally infected women:

If the mother is infected with *C. trachomatis* → the infant show evidence of infection during the first year of life "inclusion conjunctivitis"
If the mother is infected with *N. gonorrhoeae* → within the first day the baby will show severe infection of the eye "Ophthalmia Neonatorum"

Pathogenesis

- Chlamydia have **tropism** for epithelial cells of **endocervix** (This is why if we want to investigate we take endocervical swab) and upper genital tract of **women, urethra, rectum and conjunctiva** of both sexes.
- LGV can enter through skin or mucosal breaks
- Release of proinflammatory cytokines, leads to tissue infiltration by inflammatory cells, progress to necrosis, fibrosis then scarring.

Genital infections caused by *C. trachomatis*

- **In men:** urethritis (non gonococcal Urethritis, NGU), epididymitis & proctitis.
- **In women:** cervicitis, salpingitis, urethral syndrome, endometritis & proctitis.
- Urethritis present as **dysuria and thin urethral discharge** in 50% of men.
- Uterine cervix infection may produce vaginal discharge but is asymptomatic in 50-70% of women.
- **Salpingitis and pelvic inflammatory disease can cause sterility and ectopic pregnancy.**
- 50% of infants born to mothers excreting *C. trachomatis* during labor show evidence of infection during the first year of life. Most develop **inclusion conjunctivitis**, 5-10% develop infant pneumonia syndrome.
- LGV (Lymphogranuloma venereum) caused by *C. trachomatis* strains L1, L2, L3. LGV is Common in S. America and Africa. LGV presents as papule and inguinal lymphadenopathy. Chronic infection leads to abscesses, strictures and fistulas.

Diagnosis

- **Polymerase chain reaction (PCR)** is the most sensitive method of diagnosis. Performed on vaginal, cervical, urethral swabs, or urine.
- Isolation on tissue culture (**McCoy cell line**) but rarely done "culture on living tissue".
- *C. trachomatis* inclusions can be seen by **iodine or Giemsa stained smear**.

Treatment

CANNOT USE BETA LACTAMS!

Azithromycin, Erythromycin and Doxycycline.

*Doxycycline should not be used in small children and pregnant women because of the effects of tetracycline on calcification (bones)

Gonorrhea

* The Genus *Neisseria* consist of gram-negative, aerobic diplococci. Two *Neisseria* species are pathogenic for humans: *N.gonorrhoeae* (=gonococcus), the causal agent of gonorrhea, and *N.meningitidis* (=meningococcus), a frequent cause of meningitis.

*The bacteria is classified as pyogenic cocci because infections by these organisms are characterized by the production of purulent (puslike) material compromised largely of WBCs

*Microscopically --: they resemble a pair of kidney bean. It is frequently observed inside polymorphonuclear leukocytes of clinical samples obtained from infected patients "Intracellular"

* The organism produce IgA proteases that cleave IgA.

* Patient usually present with a yellow, purulent discharge and dysuria

* Pelvic Inflammatory disease can also occur here and fibrosis → leads to infertility

* Sometimes it invade the bloodstream and may result in a disseminated infection in which the organism can cause fever, a painful purulent arthritis, and skin lesions or rash. Gonococcal infection is the most common cause of septic arthritis in sexually active adults.

* Culture of *N.gonorrhoeae* on Thayer-Martin agar is THE GOLD STANDARD for diagnosis.

**N.gonorrhoeae* is resistant to penicillin. = PPNG: penicillinase-producing *N.gonorrhoeae*. so penicillin is not recommended for treatment of gonorrhea . however, most still respond to treatment with third generation cephalosporin, one dose of IM ceftriaxone.

*Incubation period is very short "days" compared to chlamydia "weeks"

*Case scenario: young, sexually active male, history of travel outside, and illegal sexual contact.

* pharyngitis is in those who practice oral sex

*pelvic inflammatory disease occurs in cases which are not treated early or properly

* why *N.meningitidis* does not require selective media but *N.gonorrhoeae* require selective media? Because the samples are usually taken from the genital tract in gonorrheal cases which is full of normal flora. The sample is a urethral discharge, and if not available, we can collect a urine sample.

N.Gonorrhoeae* ferment only **Glucose. *N.Meningitidis* ferment both **G**lucose and **M**altose.

- *Neisseria Gonorrhoeae* is a gram negative diplococci which causes the STD gonorrhea. It can be cultured.
- STD acquired by direct genital contact. It is localized to mucosal surfaces with infrequent spread to blood or deep tissues. Caused by *Neisseria gonorrhoeae*.
- Clinical manifestations: incubation period [IP] 2-5 days.
- Men: acute urethritis and acute profuse purulent urethral discharge,
- Women: mucopurulent cervicitis, urethritis with discharge.
- In both sexes: urethritis, proctitis.
- Symptoms similar to Chlamydia infection.
- Pharyngitis may occur.
- Pelvic inflammatory disease (PID) in women.
- **A Gram negative diplococci** grows on **chocolate agar** and on **selective enriched media** and CO₂ required. **Not a normal flora.**

Pelvic Inflammatory Disease

- PID occurs in 10-20% of cases, including fever, lower abdominal pain, adnexal tenderness, leukocytosis with or without signs of local infection.
- **Salpingitis and pelvic peritonitis cause scarring and infertility.**
- Disseminated gonococcal infection (DGI) due to spread to the bloodstream.

Disseminated Gonococcal Infection

- Due to spread of the bacteria to the bloodstream.
- Clinically: Fever, migratory arthralgia and arthritis. Purulent arthritis involving large joints. **Petechial, maculopapular rash.**
- Metastatic infection such as **Endocarditis, Meningitis & Perihepatitis** may develop.

Epidemiology of Gonorrhea

- Rates among adolescents are high, about 10% increase per year in USA.
- Inability to detect asymptomatic cases such as women and patient fail to seek medical care hampers control.
- Major reservoir for continued spread are asymptomatic cases.
- Nonsexual transmission is rare.

Pathogenesis

- Mainly a localized infection of epithelium, leads to intense inflammation.
- Possess pili and outer membrane proteins that mediate attachment to non-ciliated epithelium.
- Invasion by IA and Opa proteins.
- it can nullify IgA, thus, it is able to prevent an immune response.

Diagnosis

- Transport media required unless transfer to the lab is immediate.
- Direct smear for Gram stain of urethra and cervical specimens to see **Gram negative intracellular diplococci**, more sensitive in men.
- Culture on **Thayer-Martin** or other **selective medium**.
- Isolates identified by sugar fermentation of glucose only (does not ferment maltose or sucrose) or **Coagglutination test**.



Treatment

- Guided by local resistance pattern and susceptibility testing. Partner should be treated as well.
- **Ceftriaxone** IM (or oral Cefixime recommended).
- Ciprofloxacin or Ofloxacin.
- **Azithromycin, Doxycycline** (orally for 7 days) **both cover C.trachomatis** infection as well.
- Counselling.

Syphilis

Syphilis is a primarily a sexually transmitted disease caused by the spirochete *T. pallidum*. Starting with a small lesion (chancre), several progressive stages of the disease can span a period of 30 years or more, often ending in syphilitic dementia or cardiovascular damage. *T. pallidum* is so thin that it cannot be observed by conventional light microscopy, therefore, **can not be gram stained**, but requires IF or dark-field techniques to be seen.

Whenever there is a lesion on the skin that we can see and touch like a skin rash or chancre → this means that the person is highly infectious.
When the problem is inside the body "CNS OR CVS damage" → then the patient is NOT infectious.

- The disease can cause long term serious complications
- We must screen all blood donors for HIV, hepatitis, and syphilis.
- Serology is **VERY IMPORTANT** in syphilis.
- *Treponema pallidum* is a Spirochete bacterium responsible for Syphilis. Generally the main diagnosis method is serology.
- A chronic systemic infection, sexually transmitted, caused by a spiral organism called **Treponema pallidum** subsp. *pallidum*.
- The organism grow on cultured mammalian cells only, **not stained by Gram** stain but **readily seen only by immunofluorescence** (IF), dark field microscopy or silver impregnation histology technique.

Epidemiology

- An exclusively human pathogen.
- Transmission by contact with mucosal surfaces or blood, less commonly by non-genital contacts with a lesion, sharing needles by IV drug users, or transplacental transmission to fetus.
- **Early disease is infectious.** "Because it is Painless"
- Late disease is not infectious.

Pathogenesis

- Bacteria access through in-apparent skin or mucosal breaks.
- Slow multiplication produces **endarteritis** "the Pathology is mainly due to endarteritis" & **granulomas**.
- Ulcer heals but spirochete disseminate.
- Latent periods may be due to surface binding of host components.
- Injury is due to delayed hypersensitivity responses to the persistence of the spirochetes.

Clinical Manifestations / Stages

- **Primary syphilis:** **chancre is a painless**, indurated ulcer with firm base and raised margins on external genitalia or cervix, anal or oral site appear after an **IP of about 2-6 weeks**.
 - **Enlarged inguinal lymph nodes** may persist for months.
 - Lesion heals spontaneously after 4-6 weeks.
- **Secondary Syphilis:** Develops 2-8 weeks after primary lesion healed.
 - Characterized by symmetric mucocutaneous **rash, mouth lesions (snail track ulcers)** and generalized non-tender lymph nodes enlargement (full of spirochete) with **bacteremia causing fever, malaise** and other systemic manifestations.
 - Skin lesion distributed on trunk and extremities often palms, soles and face.
 - **1/3 develop condylomata lata:** which are painless mucosal warty erosions on genital area and perineum.
 - Secondary lesion resolve after few days to many weeks but disease continue in 1/3 of patients. Disease enter into a latent state.
- **Latent syphilis:** a stage where there is no clinical manifestations **but infection evident by serologic tests**. Relapse cease.
 - **Risk of blood-borne transmission** or from relapsing infection or mother to fetus continue.
- **Tertiary syphilis:** in 1/3 of untreated cases. Manifestations may appear after 15-20 years or may be asymptomatic but serological tests positive.
- **A-Neurosyphilis:** chronic meningitis, with increased cells and protein in CSF, leads to degenerative changes and psychosis. Demyelination causes peripheral neuropathies. Most advanced cases result in paresis (personality, affect, reflexes, eyes, senorium, intellect, speech) due to the effect on the brain parenchyma and **posterior columns "Tabes Dorsalis"** of spinal cord and dorsal roots.

- **B-Cardiovascular Syphilis:** Due to arteritis leads to aneurysm of aorta and aortic valve ring.
- + Localized granulomatous reaction called **gumma** on skin, bones, joints or other organs leads to local destruction.

Summary of syphilis stages:

Primary syphilis → painless chancre at site of inoculation (penis, labia, vagina)

Secondary syphilis → Rash + other systemic manifestations

Latent syphilis → No symptoms “may last 3-30yrs” but serology is still positive

Tertiary syphilis → CNS symptoms “degeneration of nervous system”, CVS symptoms “cardiovascular problems”, as well as gumma of skin, bones, joints.

- **Congenital Syphilis:** develop if the mother not treated, fetus susceptible **after 4th month of gestation**. Fetal loss or congenital syphilis result. Rhinitis, rash and bone changes (, **saber shine**), anemia thrombocytopenia, and liver failure.

saddle nose



saber shine

Diagnosis

Although treponemal spirochetes from primary and secondary lesions can be detected microscopically using IF stain or dark field illumination, **syphilis is usually diagnosed serologically**. Infection with T.pallidum elicits two kind of antibodies: 1) antitreponemal antibodies that are specific to the treponemal surface proteins; and 2) non-treponemal antibodies “regain”, that are directed against the protein cardiolipin, which is released by the injured cells. Serologic tests using both kinds of antibodies are available. Antitreponemal antibody tests are more specific than regain-based tests, but remain positive during and after successful treatment and, are, therefore, not useful for monitoring therapy. Cardiolipin-based tests are less specific, nevertheless, they are useful in screening and for monitoring therapy because tests for regain become negative about one year after successful treatment

Non-treponemal test (RPR , VDRL) → they use proteins released by the injured cells “not specific” → they only stay in blood during infectious period, so they are excellent in screening and following up the patient after treatment
Treponemal tests (FTA-ABS , MHA-TP) → they use proteins from the bacteria itself “very specific” so they are excellent in diagnosis → stay in the blood throughout life,so are not used for following up the patient after treatment

- **Serologic tests is the MOST IMPORTANT diagnostic method.**
- **Nontreponemal tests:** antibody to cardiolipin (lipid complex extracted from beef heart) called reagin. The tests are called rapid plasma reagin (RPR) and **venereal disease research laboratory (VDRL)**. Become positive during the primary stage (possible exception HIV), antibody peak in secondary syphilis. Slowly wane in later stages.
 - Used for screening and titer used to follow up therapy.
- **Treponemal tests:** treponemal antigen used.
 - Detects specific antibody to T.pallidum eg:
 - **Fluorescent treponemal antibody (FTA-ABS).**
 - **Microhemagglutination test (MHA-TP)** (antigen attached to erythrocytes)
 - Positive results confirm RPR and VDRL.
- **IgM used to diagnose congenital syphilis.**

	Test	Stage
Syphilis	Non-treponemal tests (RPR & VDRL)	POSITIVE during primary stage. screening, follow up therapy
	Treponemal tests (FTA-ABS) & (MHA-TP)	POSITIVE at all stages. if positive, confirm RPR & VDRL
	IgM antibody	Congenital syphilis

Treatment and Prevention

- Treponema is sensitive to **Penicillin**.
- Penicillin hypersensitive patients are treated with **Tetracycline, Erythromycin or Cephalosporins**.
- Prevention: counselling.

Take Home message

- Syphilis, Chlamydia and Gonorrhoea are the main STDs, caused by delicate organisms, cannot survive outside the body.
- Infection may not be localized.
- Clinical presentation may be similar (urethral or genital discharge, ulcers).
- One or more organisms (Bacteria, Virus, and Parasite) may be transmitted by a sexual contact.
- Screening for HIV required.
- If not treated early may end in serious complications.

Questions

1) A 31-year-old Haitian woman is evaluated for infertility. Pelvic examination shows a markedly enlarged vulva, inguinal lymph node enlargement, and rectal stricture. Biopsy of an inguinal lymph node reveals necrotizing granulomas, neutrophilic infiltrates, and inclusion bodies within macrophages. Which of the following is the most likely etiology of infertility in this patient?

- A. Chlamydia trachomatis
- B. Gardnerella vaginalis
- C. Molluscum contagiosum
- D. Mycobacterium tuberculosis
- E. Treponema pallidum

2) A 19-year-old woman presents with vague lower abdominal pain and a swollen, painful right knee. She denies any trauma to the knee or history of arthritic disorders. Physical examination reveals an enlarged joint that is red, warm, and painful. Pelvic examination is exquisitely painful and reveals an illdefined thickening in both adnexae. A green-yellow purulent vaginal discharge is noted. The patient is febrile and has an elevated WBC count of 15,000/ μ L. Which of the following etiologic agents is most likely responsible for this patient's condition?

- A. Escherichia coli
- B. Neisseria gonorrhoeae
- C. Streptococcus pyogenes
- D. Treponema pallidum
- E. Yersinia pestis

3) All of the following are true statements about chlamydia trachomatis EXCEPT which one?

- A. It infects several types of epithelial cells.
- B. It has a reservoir in domestic fowl.
- C. It has a number of serotypes that correlates with the syndrome produced on infection
- D. It is one of the most common causes of sexually transmitted diseases

Ans: 1:A , 2:B , 3:B