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Syphilis and advancements in its treatment

Yangyang Chen

Shanghai Experimental Foreign Language School, Shanghai, 200000, China

18321733998@163.com

Abstract. Syphilis is an infectious disease caused by the bacterium Treponema pallidum. Pallidum and its subspecies is a spirochete microaerophilic virus that sensitive to oxygen and temperature. T. pallidum is responsible for causing syphilis through sexual exposure and vertical transmission from pregnant women to their fetuses. The invasiveness and immunoevasiveness of syphilis is caused by its lack of outer membrane immune targets and with fewer surface transmembrane protein. These features explain why syphilis causes millions of people suffered from this disease and the incidence is still increasing. For the four stages of syphilis, there are different symptoms and courses of treatment approach should be taken. The administration of penicillin has reached a level of maturity and clarity in its therapeutic application. An alternative way of treatment involves the utilization of DNA vaccine. The more refined DNA vaccine technic suggests potential utility of the DNA vaccine in T.pallidum treatment. Furthermore, this review explores the current study on vaccine mRNA which holds substantial promise as a valuable avenue for syphilis treatment. T.pallidum, syphilis, transmission .

Keywords: syphilis, treatment, vaccine, T.pallidum

1. Introduction

Syphilis presents a significant threat to global public health with an alarming increase of approximately 5.6 million new cases reported worldwide each year. In 2012, there are already 17.7 millions of syphilis infection cases worldwide. Meanwhile, syphilis has also increased the possibility of infection and transmission HIV [1]. Another contributing factor to the heightened risk associated with syphilis is the lack of an outer membrane immune target on Treponema pallidum, resulting in reduced immune response efficiency. The timely diagnosis of syphilis remains a challenge, with potential delays. Syphilis primarily spreads through sexual and vertical transmission. In sexual transmission (acquired syphilis), it predominantly affects men who engage in unprotected sexual intercourse, although transmission can occur even with condom use due to mucous membrane breaks. Vertical transmission, known as congenital syphilis, occurs when pregnant women pass the infection to their fetus, often resulting in a high risk of stillbirth when left untreated. Blood transmission, once a route of transmission, has become increasingly rare [2]. Syphilis consists of four stages, including the primary, secondary, latent (early latent and late latent) and tertiary stage. For primary syphilis, the duration of incubation is 9-90 days and the characteristic single painless chancre forms at the site of inoculation. The chancres usually develop in 3 weeks after exposure. Typical chancre is painless so the diagnosis of syphilis may be delayed. However, in some cases, chancres may be painful and multiple in some individuals [3]. Secondary syphilis may go undetected due to the inconspicuous secondary lesions, typically developing

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within 3 months of the primary infection. Symptoms during this stage include condylomata lata, mucocutaneous lesions, and generalized lymphadenopathy. The manifestation would present about 8 weeks after transmission and in the first two years of infection, disseminated mucocutaneous rash and the multisystem involvement would appear. The latent syphilis is, during the period, symptoms of syphilis are absent after secondary stage. It occurs at different intervals. For patients who are in the first year of infection, they are considered at the early latent syphilis, while later latent syphilis is defined as has longer than one year or unknown duration of syphilis. Latent syphilis can be halted with treatment, while untreated patients with latent syphilis will progress into the last stage, tertiary syphilis. Approximately one-third of latent syphilis patient will advance to tertiary syphilis without timely treatment [4,5]. Treatment for different stages of syphilis involves varying doses and durations of intramuscular penicillin G benzathine. After several courses of treatment, clinical follow-up is essential to ensure a decrease in T.pallidum counts[6]. Currently, development of DNA vaccine for T.pallidum are ongoing. Better immunogenicity of flagellin plasmid DNA is promising to prevent the T pallidum transmission [7]. In addition, the mRNA vaccine that use Group A Streptococcus (GAS, Streptococcus pyogenes) and Group B Streptococcus (GBS, Streptococcus lactis) as extracellular pathogen holds potential in the future [8].

2. Mechanism

2.1. Pathophysiology

T.pallidum is a spirochete microaerophilic virus with length of 6 to 20 um and diameter of 0.10 to 0.18 um. It has a central cylinder of protoplasts enveloped by cytoplasmic membrane, peptidoglycan, and outer membrane. The out membrane does not consist of polyscharrides and the surface transmembrane protein that exposed on the surface is relatively few. The small number of immune targets that position on the T.pallidum's outer membrane is considered as the reason of its invisibility.[9-11]. Meanwhile, because of its lacking outer membrane polyscharrides and membrane protein, the immune system cannot identify and fight against its infection quickly. The genome of T.pallidum is a cyclic chromosome with 1138 kilo base pairs and contains 1041 open reading frame (ORFs) ,which makes it form a relatively small bacterial genome and also strengthens a hypothesis that many metabolism in T.pallidum requirement would rely on the biosynthetic pathway of the host. Among animals, syphilis present after human is infected by the pathogens in vivo or vitro. As to current understanding, human is the only host and the T.pallidum has no animal hosts.[12,13]. Porin is considered as the key group of memberane protein to target for. TPR gene is a T.pallidum repeated gene family that recently identified. Its code protein acts as porin and mediates its attachment to host tissuesa [14,15].With each consecutive transmission, the tpr kv part are variable. The antigenic variation through gene transformation in the infection has been assumed to be another mechanism that allow the pathogen to escape the immune response of host. by the strong host respond, the time of infection will be enlonged and continue present [15,16].

2.2. Syphilis Transmission

In the global context, Treponema pallidum subspecies pallidum stands out as the most pathogenic species within the Treponema genus. Infections typically occur through the exchange of bodily fluids during sexual contact, leading to what is commonly known as acquired syphilis. Additionally, pregnant women can transmit the disease to their unborn fetuses, resulting in congenital syphilis. Although rare today, syphilis can also be transmitted through blood transfusions, a decrease in cases largely attributed to advancements in blood bank procedures [2].

It is worth noting that aside from congenital syphilis, the majority of cases are transmitted through sexual contact. T. pallidum is transmitted through direct contact with painless syphilitic sores, known as chancres. These chancres typically manifest on the external genitalia of both genders but can also appear on the mouth, hands, anus, or even internally in areas such as the cervix or rectum [17]. An alarming statistic reveals that 30% to 60% of sexual partners of individuals with early-stage syphilis are at risk of

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contracting the disease themselves. T. pallidum can gain entry through areas with minimal barriers, typically involving mucous membranes. Recent observations have shown that anal and oral sex may play pivotal roles in the resurgence of syphilis, particularly among homosexual males. Even individuals who employ condoms during penetrative sexual intercourse may remain susceptible to infection or transmission. Such instances are most prevalent during primary syphilis, secondary syphilis, or when mucous membrane lesions are present [2].

2.3. Invasion

Despite lacking metabolic capacity, being oxygen sensitive, and having a reduced ability to survive at high body temperatures, T. pallidum can nonetheless infiltrate and persist in various tissues and organs. T.pallidum is found in large amount of primary syphilis patients' CSF. In addition, the clinical manifestation of spreads of secondary, tertiary and congenital syphilis provides evidence of the high invasion capacity of this pathogen. In infected rabbits, it has been observed that T. pallidum enters the bloodstream following either intratesticular or intradermal vaccination. The presence of the organisms has been detected on mucous membranes within deeper tissues within a matter of hours [18-20].

2.4. Immune responses

Regarding the unclear role of cell-mediated immunity in protective immunity against syphilis, the characteristic feature of syphilis lesions includes vascular abnormalities and local tissue infiltration of lymphocytes, macrophages, and plasma cells at all stages of the disease. In primary syphilis, the predominant cell types are CD4+ T cells and macrophages, whereas CD8+ T cells become the majority in secondary syphilis. This is surprising since T. pallidum is considered an extracellular organism. The expression of human Th1 cytokines such as IL-2 and IFN-γ increases, as observed in rabbit models [10,22,23].

3. Stages of syphilis

3.1. Priamary syphilis

Syphilis 'characteristic is ulcer(chancre) and local lymph node disease. Its incubation time is 9 to 90 days but the primary lesion commonly happens around 3 weeks after the transmission. Chancres usually present in the anogenital area as single, painless and indurated sores with clean base. They may excrete transparent serum. However, chancres may also exhibit variations in their presentation. In some cases, they may appear as multiple sores and are painful, suppurative, destructive, particularly when affecting the external genitalia (usually in oral cavity). Furthermore, they can also lead to a condition known as syphilitic balanitis of Follman [3]. The infection initiates when T.pallidum penetrate the micro abrasion in dermis or complete mucous membrane, it also results in the development of a solitary chancre at the site of immunization.

Mild regional syphilis is predominantly linked to the initial phase of the infection. During this stage, chancres typically develop from a sclerotic lesion and progress into ulcers, but they typically do not suppurate. In male homosexual individuals, primary chancres are commonly found on the penis, but a significant portion, approximately 32% to 36%, may also present in other locations such as the rectum, oral cavity, or anal canal [24,25].

Furthermore, research indicates that individuals co-infected with HIV have a notably higher probability, approximately 2 to 3 times greater, of progressing to both primary and secondary syphilis compared to those without an HIV diagnosis [26,27].

3.2. Secondary syphilis

Multisystem involvement, the characteristic of secondary syphilis in the initial two years of infection and it may first present after 8 weeks of transmission. The initial presentation of secondary syphilis often includes a rash that appears rose or macular in shape, with longer lesions potentially developing into papules or nodules. This type of rash is typically not irritating but may be associated with pruritus,

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especially in individuals with darker skin tones. Secondary syphilis is characterized by various manifestations, including condylomata lata, mucocutaneous lesions, generalized lymphadenopathy [3]. These symptoms are often associated with the presence of secondary syphilis.

Systemic symptoms of secondary syphilis may include a throat and muscle soreness, discomfort, and weight loss. Approximately 85% of cases exhibit systemic non-tender lymphadenopathy, with the most common phenomenon being a disseminated mucocutaneous rash. On the trunk and proximal extremities, there are initially pale and distinct macular lesions., followed by a variety of lesion types. In terms of diagnosis, maculopapular lesions are present in 50% to 70% of patients, making them the most common secondary syphilis lesions (12% popular, 10% macular, and 6% to 14% annular popular) [28,29]. It's worth noting that some cases may present with inconspicuous secondary lesions that go unnoticed [28]. Rashes frequently manifest on the palms and soles on feet, with approximately 4% to 11% of infected individuals encountering hair loss as a result of T. pallidum infection within the hair follicles [25,29].

The condyloma lata is developed at the same time with the secondary syphilis lesion among approximately 10% of patients. These expanded lesions in the warm and moist regions in body like perineum and anus are highly contagious. Inflammation in the oral cavity, tongue, and genital mucosa can lead to formation of mucous patches [25,28]. Secondary syphilis rarely accompanies stomach and kidney damage, as well as hepatitis [30,31]. Nephrotic syndrome may also occur in some cases. Early neurosyphilis symptoms may be present in about 5% of secondary patients [19,28].

3.3. Latent syphilis

The diffuse lesion and other symptoms of secondary syphilis spontaneous regress usually in a period of three months and has no manifestation at different time among people who has not been treated. The latent syphilis can be divided into two stages by the approximate duration of infection. Patients are typically considered to have early latent syphilis during the first year of infection, and 25% of those who do may experience recurrent secondary symptoms [4]. The later latent syphilis is defined as the non-symptom infection for more than one year or longer; In the late latent stage, no transmission via sexually intercourse could occur but serological tests would show positive result. In this phase, organism may be found in the blood and can infect fetuses during pregnancy. The latent syphilis will be ended when curative antibiotic therapy is applied to patients who have entered tertiary syphilis. For individuals who have not been treated and progressed to latent syphilis, syphilis can show up in multiple organs so would be called neurosyphilis, cardio- vascular syphilis and gummatous syphilis while this syphilis exits at the same time [3].

3.4. Tertiary syphilis

In the retrospective Oslo study [5], about one third of latent syphilis patients developed to tertiary syphilis. After 20 to 40 years of infection, syndromes have broken out. In 15% of untreated syphilis individuals, gumma caused by progressive inflammation (late benign syphilis) which is a kind of localized bone and tissue deterioration and 10% of patients have developed cardiovascular syphilis that typically seen as an aneurysm or aortic insufficiency; 6.5% patients are with the symptomatic late neurosyphilis. And in the Table 1, it shows the summary of information of the four syphilis stages.

Table 1. Information of four syphilis stages.

Stage of syphilis	Time of stage initiation	Incubation	Symptoms
Primary	/	9-90 days	Ulcer(chancre) and local lymph node disease. Chancres may be multiple, painful, suppurative, destructive, at external genitalia

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Table 1. (continued)

Secondary	within three months of initial infection, manifestation forms		multisystem involvement, condyloma lata, mucocutaneous lesions, generalised lymphadenopathy and patchy alopecia, meningitis, cranial nerve palsies, hepatitis, anterior uveitis, periostitis splenomegaly, and glomerulonephritis associated with the present of secondary syphilis. sore throat, muscle soreness, discomfort, and weight loss
Early latent	first year of infection	/	diffuse lesion and other symptoms of secondary syphilis.
Late latent	more than one year or the unknown duration of infection	/	non-symptom infection. when no treatment applied, sphilis categorised into neurosyphilis, cardio- vascular syphilis and gummatous syphilis
Tertiary	after the untreated latent syphilis	20-40years after infection	15% of gumma 10% of cardiovascular syphilis

4. Prevention

Syphilis continues to make challenge to the public health worldwide, especially it has heavily increased the risk of infection and transmission HIV [2]. According to the recent estimate from WHO, there were 17.7 million syphilis individuals among 15 to 49 years old in 2012 and an increase of 5.6 million new cases each year. The estimated epidemicity and morbidity rate of syphilis may differ among different countries. The highest currency of this disease occurs in Africa, over three fifth of new cases happen in LMIC [1,33].

The Centers for Disease Control and Prevention (CDC) and the American Academy of Pediatrics (AAP) have jointly developed an algorithm for assessing data concerning infants born to mothers infected with syphilis during pregnancy. In cases where mothers have tested positive for reactive non-treponemal testing (RPR) at any point during pregnancy, it is imperative that the new-born infants undergo syphilis testing via RPR immediately after birth.

The evaluation of infant health status is based on various factors, including the comparison of infant RPR results with maternal RPR levels at the time of delivery, the mother's treatment history, and maternal RPR titres. These assessments help in classifying infants into distinct risk scenarios, as outlined by the CDC, or categories as defined by AAP.

It is important to emphasize that early syphilis diagnosis before and during pregnancy is a key role in preventing increase of i congenital syphilis cases [34,35].

5. Treatment

5.1. Antibiotics

Currently, the primary treatment strategy for primary, secondary syphilis and early latent syphilis are still treated with a single 400 000 U dose of penicillin G benzathine intramuscularly. For late latent syphilis or uncertain syphilis and tertiary syphilis, 2400 000 U intramuscular penicillin G benzathine

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need to be administered weekly for 3 weeks. Depending on the stage of the illness, patients who are allergic to penicillin can take oral doxycycline 100 mg twice day for 14–28 days. Desensitisation therapy is advised for pregnant women who are allergic to penicillin so they can continue receiving treatment with penicillin G benzathine. For a year, patients should have a clinical and laboratory follow-up every three to six months. The decrease of incidence in non-periodic experiments is considered as a positive result for treatment. Meanwhile, the possibility of nerosyphilis need to be considered. Also, for the patients whose value do not decrease by at least 2 diluents in non-syphilis tests should be retreated as this may indicate the possibility of reinfection [6].

5.2. Vaccine

With the ongoing advancement of DNA vaccine research, investigations into T. pallidum DNA vaccines have deepened. Researchers have employed the MTT method that use PCR and ELISA to assess vaccine titers. The findings of these studies indicate that combined immunization with multiple vaccine components yields superior immune activity and protective effects compared to single-gene vaccine administration. However, it's noteworthy that the use of CS nanoparticle packaging did small positive effect to the vaccine's performance [36].

Recent research reports have highlighted the significantly enhanced immunogenicity of flagellin plasmid DNA compared to recombinant flagellin vaccines. This discovery suggests that leveraging flagellin plasmid DNA may represent an effective strategy for combating T. pallidum transmission. Additionally, a separate study identified a limitation in DNA vaccine immunization: although it induces the production of high levels of antibodies, it tends to elicit a relatively weak cellular immune response, resulting in suboptimal immunity.

6. Prospect

The exploration of mRNA-based vaccines holds promise in the context of combating infections caused by extracellular pathogens such as Group A Streptococcus and Group B Streptococcus [8]. Notably, these pathogens primarily induce a TH1-like immune response. This TH1-like response is of particular significance as it has been associated with the effective clearance of pathogens like T. pallidum and serves as a valuable defense mechanism against bacterial infections. In efforts to enhance the immunogenicity of vaccines, recent developments include the utilization of messenger ribonucleic acid (mRNA) vaccines targeting Mycobacterium tuberculosis, incorporating Toll-like receptor 4 (TLR4) agonist peptide (RpfE) as an adjuvant [37,38]. It is worth noting that, as of now, there is no provision for the mRNA vaccine against the T. pallidum. Nevertheless, the substantial practical experience gained in the creation of vaccines for mRNA targeting viruses and other infectious pathogens underscores the potential benefits of applying this knowledge to address this unmet need.

7. Conclusion

This review introduces the Treponema pallidum that cause human syphilis and talks about the invasion, mechanism of infection and the immune response after the immune cells detect the Treponema pallidum . Also, the symptoms, duration and the incubation time of four different stages of syphilis are listed and summarized. For the different stages, the recent preventions and treatments takes in each stages are discussed. A promising mRNA vaccine is worth to treat syphilis because of its higher immunogenicity. Although the mRNA vaccine has no provision recently, the better effect of the mRNA vaccine in study is prove the potential benefit of vaccine.

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