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Research Article

Seroprevalence of *Treponema pallidum* Among Residents of Yemetu, A Semi-Urban Community in Ibadan, Southwest Nigeria

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ARTICLE INFO

Article history: Received: 29 August, 2020 Accepted: 30 September, 2020 Published: 6 November, 2020

Keywords: Treponema palladium/syphilis asymptomatic prevalence semi-urban Ibadan Nigeria

ABSTRACT

Introduction: Syphilis is a sexually transmitted bacterial disease transmitted during vaginal, anal and oral sex. Pregnant women can also pass the disease to their unborn babies. However, it thrives in dirty and unhealthy environment which has made its spread uncontrollable. Therefore, this community-based study was carried out in Yemetu community in Ibadan to determine the burden of syphilis infection among the asymptomatic residents of this community.

Methodology: Blood samples were aseptically collected from consenting one hundred and fifty male (m=49 and female (f=101) participants, age ranged 15 to \geq 55 years who volunteered to take part in the study. A structured questionnaire was used to capture demographic data and other relevant information from these participants. Sera from these blood samples were tested for syphilis antibodies using a 3rd generation Enzyme Linked Immunosorbent-Assay (ELISA) kit, syphilis Ab version Ultra. Data were analysed using Chi-squared and ANOVA with p<0.05 considered as significant.

Results: An overall seroprevalence rate for *Treponema pallidum*/syphilis in this study was 1.3%. Syphilis infection was higher among male (2.1%) than in female (1.0%), 1.6 times higher in male compared to their female counterparts (OR=1.57, 95% CI 1.43-2.81) and also statistically significant (p=0.0313). Highest rate (4.1%) of the infection was detected in age group >55 years while lowest rate (3.8%) of syphilis infection was found in age group 15-24 years. A total of 46 (30.7%) participants claimed to practice multiple sexual partnership with 2 (4.3%) of them positive for *Treponema pallidum* and this was found to be statistically significant (p=0.043). Furthermore, distribution of *Treponema pallidum* infection by marital status in the study community showed that highest prevalence (2.4%) was found among the single while lowest rate (1.2%) was detected among married participants. Among the occupational groups, highest *Treponema pallidum* infection rate (3.6%) was found among artisans and the lowest rate of 3.5% was among the students.

Conclusion: Although this study reported low prevalence for syphilis infection in this community among asymptomatic population, this rate is however of public health importance. Therefore, public awareness for hygiene, high risky behaviour including voluntary counseling and testing for syphilis should be advocated.

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Introduction

Syphilis is a sexually transmitted disease caused by a spiral-shaped bacterium, *Treponema pallidum* [1]. It is classified under phylum spirochaeta, order spirochaetales, family spirochaetaceae and genus *Treponema*. It can enter the body through minor cuts or abrasions in the

skin or through mucous membranes, most often sex. Syphilis is passed from person to person through direct contact with an infected ulcer [2]. These ulcers occur most frequently on the external genitals, vagina, and anus or in the rectum. Occasionally the sores also occur on the lips and in the mouth. The genital ulcer of early syphilis can increase the risk of HIV transmission [3, 4].

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In addition, syphilis is transmitted during vaginal, anal and oral sex. Pregnant women can pass the disease to the babies they are carrying. Syphilis cannot be spread from toilet seats, pools, hot tubs, shared clothing or eating utensils [5]. There are at least three more known species causing human treponema diseases such as *Treponema pertenue* that causes Yaws, *Treponema carateum* causing Pinta and *Treponema pallidum endemicum* responsible for bejel or endemic syphilis. Yearly, millions of people are exposed to avoidable, life-threatening risks through the transfusion of unsafe blood. A global database indicates 6 million of 81 million units of blood collected annually in 178 countries are not screened for transfusion transmissible infections in which syphilis is one [6].

Syphilis is believed to have infected 12 million people in 1999, with greater than 90% of cases in the developing world [7]. After decreasing dramatically since the widespread availability of penicillin in the 1940s rates of infection have increased since the turn of the millennium in many countries often in combination with Human Immunodeficiency Virus (HIV). This has been attributed partly to increased promiscuity, prostitution, decreasing use of condoms, and unsafe sexual practices among men who have sex with men [8, 9]. Between 700,000 and 1.9 million pregnancies are affected yearly as a result of syphilis infection. Also, WHO has estimated that 1.9 million pregnant women had active syphilis and it is higher in women who did not receive adequate prenatal care [10]. The rate of infection is also higher in sub Saharan Africa, where syphilis is found among drug users and HIV infected patients [8, 11]. Syphilis can present in at least one to four stages: primary, secondary, latent and tertiary [7, 12].

Congenital syphilis (CS) too can be classified into early and late CS and this is manifested by factors such as gestational age, stage of maternal syphilis, maternal treatment, and immunological response of the fetus [13]. Early CS is characterized by hepatomegaly, splenomegaly, hemolytic anemia, osteochondritis, nephritic syndrome and hydrops fetalis are seen in early CS while late CS is characterized by Hutchinson's triad, which is a constellation of Hutchinson's teeth, interstitial keratitis and eight nerve deafness [13]. There are four members of the bacterial family and none of them can be differentiated with morphological, chemical or immunological methods [14]. Of the aforementioned bacteria, syphilis is the sole sexually transmitted treponema disease, as the other conditions are transmitted via direct contact with an infected individual [15].

The study location (Yemetu) is a semi-urban community and a densely populated area in Ibadan. It has many social joints or locations where some people are engaged in alcoholic consumptions and certain drugs, exposing them to risky sexual behaviours and other vices and age is no barrier. Awareness of syphilis infection is very low with poor hygiene and self-medication is common among the inhabitants of Yemetu community. Hence there is need to carry out an epidemiological survey to determine the knowledge/awareness of the inhabitants of Yemetu as regards syphilis infection and the correlation of their social behaviour with this infection. Therefore, this study was designed to assess the burden of syphilis among sexually active inhabitants of Yemetu community in Ibadan, Nigeria.

Methodology

Study Area as described by: Briefly, this descriptive community-based study was carried out in Yemetu, a semi-urban community located in an area covering about 27 square kilometer, in Ibadan North Local Government of Oyo State [16]. The residents of the community are dominated by Yorubas with mixtures of fewer tribes such as Igbos, Hausas and fewer Togolese. They are majorly into petty trading and peasant farming, mostly artisans with few public servants and students. Many of them practice African traditional religion while others are Christians and Muslims. The community can boast of a State-owned secondary health facility (Adeoyo Maternity Hospital) which is not too far from the University College Hospital (a tertiary healthcare facility and a teaching hospital for the University of Ibadan), public and private schools, and a Police station.

The population of this community is less than 1/10th of the population of Ibadan. The community is a semi-urban setting in the heart of Ibadan. Major characteristics of the area of study (Yemetu community) are poor housing and environmental sanitation and improper waste management. Majority of the inhabitants of the city are low income earners. Ibadan is endowed with business viability which attracts people from many other parts of the country. Ibadan is an urban city in southwest Nigeria which is densely populated and the capital of Oyo State. It has a population of over 2.5 million people according to the 2006 National Population Commission Census. Enrolment of the participants and study population: A total of 150 consenting apparently healthy low-income community dwellers between ages 15 and 60 (Median age = 27.3) years, without prior knowledge of their HBV status were recruited into the study. The study was based upon availability of participants in their residence together with their willingness to take part in the study. The key assumption for using this approach was anchored on the ground that the target population is homogeneous and share same characteristics and lifestyle.

They were provided with health education relating to modes and routes of contracting HBV including preventive measures before enrolment into the study in order to assess their knowledge and to provide information on HBV. Participants were educated and enlightened about the dangers of HBV infection, mode and risk factors for its transmission, including ways of preventing HBV infection. Following this, a semi-structured questionnaire was used to capture each participant's socio-demographic information and history of vaccination. Although many prospective participants indicated their willingness to be enrolled into the study but only 150 of them showed interest to give their blood samples collected by a trained phlebotomist.

I Sample Collection and Laboratory Analysis

The aseptically collected samples from each participant by venipuncture into sterile EDTA tubes and reagents were allowed to attain room temperature. The samples were initially spun at 2,500 rpm for 10 mins to separate the plasma. Required numbers of microwells were placed on the microwell holder. One hundred microliters each of negative control and calibrator in duplicates and positive control in single well were dispensed appropriately. This was followed by dispensing 100µl of each of the samples appropriately. The microplate was incubated for 60 minutes at 37°C followed by washing, using an automatic washer. About 100 μ l enzyme conjugate was dispensed into each well, except the 1st blanking well, and covered with the sealer. The microplate was incubated for 45 minutes at 37°C and was washed with an automatic washer again. 100 μ l TMB/H₂O₂ mixture was dispensed into each well including the blank one (the reagents were checked to be correctly added). The microplate was incubated for 15 minutes at room temperature (18-24°C).

One hundred microliters sulphuric acid was dispensed into all the wells using the same pipetting sequence to stop the enzymatic reaction which turned the positive control and positive samples from blue to yellow/brown. The colour intensity was measured in each well with a microplate reader (Molecular Devices, California, USA) at 450 nm (reading) and at 620-630 nm (background subtraction was strongly recommended).

II Principle of the Test

Microplates were coated with purified *Treponema pallidum* synthetic antigens. The solid phase was first treated with the sample and anti Tp Ab was captured, if present, by the antigens coated on the microplate. After washing out all the other components of the sample, in the second incubation bound anti Tp total antibodies, these were detected by the addition of Tp synthetic antigens, labeled with peroxidase (HRP). The enzyme captured on the solid phase, acting on the substrate/chromogen mixture, generated an optical signal that was proportional to the amount of anti Tp antibodies present in the sample. After blocking the enzymatic reaction, its optical density was measured by an ELISA reader. The version ULTRA was particularly suitable for automated screenings.

III Data Analysis

Data were analysed using SPSS version 21. Mean, median and tables were used to describe the results using Chi-square and ANOVA at 95% CI for Odd Ratio. P-value < 0.05 was considered significant at 95% CI.

Results

Association of socio-demographic information and clinical correlates of *Treponema pallidum* rates of the study participants showed that sixty-three (42.0%) participants had prior knowledge of syphilis while 87 (58.0%) were ignorant. Fifty-eight (38.7%) of them had no incisions on their bodies while ninety-two (61.3%) were incised at one point in their lives. A total of 46 (30.7%) participants claimed to practice multiple sexual partnership with 2 (4.3%) of them positive for *Treponema pallidum* and this was found to be statistically significant (p=0.043) (Table 1). A total of 61 (40.7%) of the participants had only primary education, 41 (27.3%) did not have any formal education while the rest 36 (24.0%) and 12 (8.0%) had secondary and tertiary education respectively.

The subjects also consisted of 46 (30.7%) individuals with multiple sexual partners and 104 (69.3%) people with one or no sexual partner. The prevalence rate of *Treponema pallidum* was higher in male (2.0%) than in female (1.0%) and was 1.6 times higher in male compared to their female counterparts (OR=1.57, 95% CI 1.43-2.81). Of the 49 male participants tested, 1 (2.0%) was positive for *Treponema pallidum* infection while 1 (1.0%) female participant also tested was positive.

Characteristics	No Tested (N=150)	No (%) Positive	p-value	OR (95% CI)
Age range (years)				
15-24	26(17.2)	1(3.8)	0.61	1.22((1.39-2.74)
25-34	29(19.3	0(0.0)		
35-44	40(28.7)	0(0.0)		
45-54	31(20.7)	0(0.0)		
≥ 55	24(16.0)	1(4.1)		
Marital Status				
Single	41(27.3)	1(2.4)	0.0579	1.14((0.86-1.77)
Married	85(56.7)	1(1.2)		
Separated	16(10.7)	0(0.0)		
Widowed	8(5.3)	0(0.0)		
Sex				
Male	49(32.7)	1(2.0)	0.0313*	1.57(1.43-2.81)
Female	101(67.3)	1(1.0)		
Level of Education				
Primary	61(40.7)	3(4.9)	0.066	0.83(0.9662-1.55
Secondary	36((24.0)	2(5.6)		
Tertiary	12(8.0)	1(8.3)		
None	41(27.3)	5(12.2)		
Knowledge about Syphilis				
Yes	63 (42.0)	1(1.6)	0.157	
No	87(58.0)	1(1.1)		

Table 1: Association of sociodemographic information and clinical correlates with Treponema pallidum infection among the study population.

Having multiple sexual partners				
Yes	46((30.7)	2(4.3)	0.043*	
No	104(69.3)	0(0.0)		
Have tattoo, incision/tribal in any part of the body				
Yes	92(61.3)	1(1.1)	0,066	
No	58(38.7)	1(1.7)		

*P<0.05 was considered statistically significant (using Chi square test).

However, despite the same number of positive by gender in this study, the rate of infection was statistically significant (p=0.0313) (Table 1). An overall rate of 1.3% of *Treponema pallidum* infection was found in this study. Highest rate (4.1%) of the infection was detected in age group >55 years while lowest rate (3.8%) of syphilis infection was found in age group 15-24 years. However, *Treponema pallidum* infection was not detected in age range 25-44 years (Table 2). Furthermore, distribution of

Treponema pallidum infection by marital status in the study community showed that highest prevalence (2.4%) was found among the single while lowest rate (1.2%) was detected among married participants. The separated and widowed participants did not have *Treponema pallidum* infection (Table 1). Among the occupational groups, highest *Treponema pallidum* infection rate (3.6%) was found among artisans and the lowest rate of 3.5% was among the students (Table 3).

Table 2: Age	Distribution of	of syphilis	infection b	by Gender in	Yemetu	Community.
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Age Range	Male			Female		Total	
	No Tested	No (%) Positive	No Tested	No (%) Positive	No Tested	No (%) Positive	
15-24	14	0(0.0)	12	1(8.3)	26	1(3.9)	
25-34	9	0(0.0)	20	0(0.0)	29	0(0.0)	
35-44	10	0(0.0)	30	0(0.0)	40	0(0.0)	
45-54	8	0(0.0)	23	0(0.0)	31	0(0.0)	
<u>></u> 55	8	1(12.5)	16	0(0.0)	24	1(4.2%)	
Total	49	1(2.0)	101	1(1.0)	150	2(1.3)	

Table 3: Occupational distribution of syphilis infection in Yemetu Community, Ibadan.

No. Tested (%)	No. (%) Positive
29 (19.3)	1 (3.5)
18 (12.0)	0 (0.0)
72 (48.0)	0(0.0)
03 (2.0)	0(0.0)
28 (18.7)	1 (3.6)
150 (100)	2 (1.3)
	No. Tested (%) 29 (19.3) 18 (12.0) 72 (48.0) 03 (2.0) 28 (18.7) 150 (100)

Discussion

This study showed an overall prevalence of 1.3% for Treponema pallidum (syphilis) infection among the inhabitants of Yemetu in Ibadan who participated in the study. This rate was higher than a prevalence of 0.13% reported by carried out among pregnant women in Ibadan and 0.3% rate for syphilis among Nigerian pregnant women reported by the Federal Ministry of Health (FMOH) which indicated that, the national prevalence for syphilis among Nigerian pregnant women was put at such value [17, 18]. Similarly, the prevalence 1.3% in this study was slightly higher than 1.0% rate reported in a study carried out among pregnant women in Osogbo [19]. Also, a lower prevalence of 0.4% when compared with the reported 1.3% detected in this study was reported among pregnant women, attending antenatal clinic in North-eastern Nigeria and 0.13% found by in Enugu, South-eastern Nigeria [20, 21]. Therefore, this indicates that syphilis infection in this region is endemic and it is spreading very fast among asymptomatic population in this study community.

However, this rate for syphilis reported in this community was lower than the 19.3% overall prevalence for syphilis reported among similar population in Ilorin by Sule *et al.* [22]. Previous studies in different parts of Nigeria also indicated higher rates for syphilis infection when compared with 1.3% detected in this study. In Calabar, a south-eastern part of the country, a seroprevalence of 6.5% was reported by while reported 1.7% in their work in Ilorin, North-central Nigeria [23, 24]. Furthermore, in other African countries, prevalence of syphilis has been reported to be higher than 1.3% reported in the present study which was lower than 2.0% in Cotonou, 4.0% in Kisumu, 6.0% in Yaounde, and 14.0% among Ndola women [22]. Likewise, 12.5% in Zambia, Mozambique (18.3%) and Malawi (5%) have been reported [25-27].

Nevertheless, Todd *et al.* did not find any case of syphilis infection in the 4,452 pregnant Afghan women receiving antenatal care at three government maternity hospitals in Kabul [28]. The inability of the study to detect syphilis infection from such large population could depend on the sensitivity of the test kit used or the public sensitization or awareness on the mode and route of transmission of this pathogen. Some studies have attributed the spread of syphilis to the socioeconomic status and the sexual behaviour of the residents of the community in which the infection is rampant [22, 28]. This therefore reaffirms the spread of primary syphilis infection as synonymous to multiple sexual partnership and environmental sanitation of the inhabitants of such community. Furthermore, the difference in the various reports could be attributed to variation in sexual practices, sexual behaviour of the community where studies were conducted, geographical variation, cultural practices, differences in accessibility to treatment of STIs (Sexually Transmitted Infections), and differences in the laboratory techniques employed to detect syphilis infection [20]. The much higher seroprevalence rates found for syphilis from Southern Africa (Zambia, Malawi and Mozambique) might be due to the higher prevalence rates of HIV/AIDS infection in that part of Africa [20].

Gender distribution of Treponema pallidum infection in this study indicates that the infection was higher among the male (2.0%) than their female (1.0%) participants (p=0.313; p<0.05) and 1.6 times higher risk of contracting syphilis infection in male compared to their female counterparts (OR=1.57, 95% CI 1.43-2.81). This rate was comparable with 4.5% rate of syphilis infection among male population in a study carried out among patients attending General Hospital, Calabar. However, despite the fact that the rates for HBV infection by gender in the present study were lower, they are still comparable since the two studies were carried out among asymptomatic population. Other factors could be environmental factors such as hygiene and sexual exposure as earlier reported in a previous study carried out at the General Hospital, Calabar in which 2.0% rate was detected among the female participants [23]. Also, other studies reported various rates of syphilis infection by gender while others carried out gender specific studies which did not give room for comparison [17, 20].

Age specific data indicates that syphilis infection is rampant among sexually active population which may implicate sexual transmission of the pathogen [17, 23]. However, this study shows a highest rate of 4.2% for syphilis infection detected in older population among the participants and this portrays the spread of the infection across all ages irrespective of gender [17, 22]. Nevertheless, this infection was not detected in older age category in an earlier study carried out among patients attending General Hospital Calabar [23]. Other factors may also be responsible for the spread of syphilis infection which include socioeconomic and cultural myth among others [26, 27].

Additionally, while blood transfusion can be life-saving, there are associated risks, particularly the transmission of blood-borne infections [29]. This might be responsible for the alarming rate for syphilis infection among blood donors as earlier reported [30]. From this study, syphilis among blood donors is of alarming rate (8%). Syphilis among blood donors in this study was higher than the 3.6% found by Chikwem *et al.* in Maiduguri, North-eastern Nigeria; the 7.5% found by Adjei *et al.* among Tanzanian donors; and lower than the 12.7% reported by Matee *et al.* among Sudanese donors [31-34]. The implication of syphilis in voluntary blood donors is the risk of transmission of this infection to recipients of blood and blood products. This can contribute to the everwidening pool of infection in the wider population.

Occupational distribution of syphilis infection also indicates that some professions are at higher risk of acquiring this infection and these include health workers particularly those in STI clinic [17]. The risk for syphilis infection is also higher among commercial sex workers, polygamy and multiple sexual partnership and others living in dirty environment with poor sanitary practice [17]. A higher risk was also noted in this study among participants with multiple sexual partnership for a statistically significant value (p=0.043) was found. Also, in this study, artisans had higher rate for syphilis infection than the students. The Artisans in this community had low educational background with very low knowledge of this infection and the risk of acquiring it. Socio-demographic information from the study population also implicates married and unmarried participants with syphilis infection.

Conclusion

Despite the low prevalence of syphilis infection in this study community, it is still considered a threat to public health. It is important to note that the index case in this study was asymptomatic. Multiple sexual partnerships also play a high factor in circulating the pathogens within the community. Although the same number of male positive for Syphilis infection was also the same for their female counterparts, the rate of infection was however higher in male and this difference was statistically significant (p<0.05), affecting sexually active population who are the artisans and students. This therefore indicates that syphilis infection is endemic in this community and the region in general. Hence, necessary steps including public awareness and promotion of high level of hygiene are desirable.

Acknowledgments

All authors wish to express their gratitude to all the study participants for their co-operation.

Author Contributions

- i. Study design (All authors)
- ii. Sample collection (BAS, DAC)
- iii. Reagent acquisition, laboratory and data analysis (All authors)
- iv. Wrote first draft of the manuscript (BAS)
- v. Revised the manuscript (All authors)
- vi. Read and approved the final draft (All authors)

Ethical Approval

Ethical approval for the study was obtained from Oyo State Ethical Review Board committee of the Ministry of Health (AD3/479/349). Participation in this study was voluntary, and individuals who did not consent at any stage were excluded in order to follow the principle of autonomy. They were provided with full assurance of anonymity, confidentiality, privacy together with all the information supplied including the samples to be collected.

Conflicts of Interest

None.

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