

**INTERNATIONAL JOURNAL OF ADVANCES IN
PHARMACY, BIOLOGY AND CHEMISTRY**

Research Article

**Seroprevalence of Syphilis among known HIV
Patients in Khartoum State - Sudan**

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ABSTRACT

Background: HIV and syphilis co-infection is complex and can occur through sexual intercourse, blood transfusion and vertical transmission.

Objective: The aim of this study was to detect serofrequency of syphilis among HIV Patients in Khartoum state during February to March 2016.

Method: A total of 90 HIV patients were enrolled in this study, serum tested for anti-syphilis total antibodies using ELISA.

The Results: Anti-syphilis rate among known HIV patients was 8(8.9%).The highest seroprevalence was observed among HIV Patients of 41-60 years age range with average 3(3.3%).

Conclusion: This study reflected the high frequency of syphilis among known HIV patients.

Keywords: Seroprevalence, Syphilis, HIV, ELISA, *Treponema pallidum* and Khartoum-Sudan.

INTRODUCTION

Syphilis is one of the oldest diseases caused by the bacterium *Treponema pallidum subspecies pallidum* and classified as acquired or congenital, The World Health Organization estimates that 12 million new infections of syphilis occur every year; of which, almost two-thirds are in sub-Saharan Africa and south/southeast Asia¹. *T.pallidum* is a spirochete bacterial with subspecies that cause treponemal diseases such as syphilis, pinta, bejel and yaws. treponemes are Gram negative, but they are so thin to be Gram stained, there are only visible using dark field microscopic examination².

Unprotected sexual contact, blood transfusion, vertical transmission from mother to baby are the main transmission routes for syphilis, cases of infection through blood transfusions are rare due to the limited survival of *T. pallidum* in collected blood and the serological screening of blood donation

candidates, whereby it is rapidly destroyed within a few minutes of exposure to heat, drying or air³. Syphilitic ulcers disrupt epithelium and mucosa, It could be transmissible by oral, vaginal, and anal sex^{4,5}. Transmission of bacteria represents the most frequent infectious complication of blood transfusion in the developed world and a major cause of transfusion associated mortality. Incubation time from exposure to development of primary lesions, which occur at the primary site of inoculation, averages 3 weeks but can range from 10-90 days. Regardless of the stage of disease, Donors can transmit the infection⁶.

T. pallidum organisms may be able to evade the acquired immune response by antigenic variation of bacterial surface proteins, consistent survive bacterial clearance of the primary lesion^{7,8}. Although its

treatment is accessible, its incidence remains a serious public health problem⁹.

Two types of serologic tests are required for diagnosis. First, there are treponemal tests, such as fluorescent treponemal antibody absorption (FTA-ABS), *T. pallidum* particle agglutination (TPPA) and enzyme immunoassay (EIA), chemiluminescence immunoassays, immunoblots and rapid treponemal assays. . Second, there are nontreponemal tests, such as the Venereal Disease Research Laboratory (VDRL) test or the rapid plasma reagin (RPR) test¹⁰. The interaction between syphilis and HIV infection is complex and remains incompletely understood, despite 30 years of clinical experience¹¹.

Syphilis is a complex disease, which is sexually transmitted. The incidence of syphilis is rising all over the world, partly due to the increased transmission in HIV patients^{12,13}. HIV can alter the clinical picture of syphilis and can cause more serious complications. Incidence of neurosyphilis in HIV infected individuals is high even when treated with recommended dosage¹⁴. Primary syphilis infection causes a genital ulcer, which disrupts the mucous membrane, making it more vulnerable to penetration by the HIV virus, During episode of Syphilis ,CD4 counts transiently decrease and HIV viral load increase. On the other hand, impaired cellular and humoral immunity in HIV infection lowers host defense against *T. pallidum*, leading to changes in the natural course and clinic progression of syphilis, shortened incubation time, and increased number and infectiousness of syphilis lesions^{15,16}.

In a systematic review of literatures from several regions of the world, a cause more serious complication median point-prevalence of syphilis among HIV-infected patients was shown to be 9.5%¹⁷. This study aimed to detect the frequency of syphilis among known HIV patients who attending Anti retroviral therapy clinics.

MATERIALS AND METHODS

The current descriptive, cross-sectional study carried out between January and March 2016. Ninety known HIV patients who attended clinics in Khartoum state, Sudan were recruited in this study. Approval was taken from Al Neelain University Ethical Committee Board and an informed consent was obtained from each patient before collecting the specimen, demographic and clinical data.

Experimental work:

Collection of Specimen:

Venous blood samples (5ml) in anticoagulant free container was taken and sera was separated and kept frozen at -20°C. All tested sera were tested for

antibodies by using ELISA Kit. The type of ELISA used was antigen sandwich ELISA (Fortress-diagnostics limited, United Kingdom).

Test procedure to detect anti-syphilis total antibodies:

All reagents were brought to room temperature before assaying, 100 micro liter of HRP conjugate were dispensed into all wells, except for the blank well, then 20µ of positive control, negative control , samples were dispensed into their respective wells, then the card board sealer was applied on to microtitters wells to prevent evaporation, and incubated for 1 hour at 37°C, and when incubation was completed, the card board was discarded, and the strips were washed by using automatic washer for 6 times , after that the strips mouth were turned down on to blotting paper to remove any liquid residue.

50 µ of chromogen A and 50 µ of chromogen B solution was dispensed into all wells and incubated for 15 minutes at 37°C away from intense light, then 50µ of stop solution was dispensed into all wells in the same order and at the same rate as for chromogen/ substrate, the absorbance of specimens were measured with photometer at 450/630 nm within 5 minutes after stopping the reaction.

Measuring the absorbance:

The plate reader was calibrated with blank well and the absorbance was read with micro well reader at 450 nm, the results were calculated by relating each sample optical density (OD) value to the Cut off value of plate.

Calculation of Cut off (C.O) value:

$$C.O = *Nc + 0.18$$

*Nc = the mean absorbance value for the three negative controls.

Interpretation of results:

Negative results: samples giving absorbance less than Cut-off value are negative for this assay.

Positive results: samples giving absorbance equal to or greater than Cut-off value considered initially reactive.

Borderline: samples with absorbance to cut-off ratio between 0.9 and 1.1 are considered borderline and retesting of those samples is recommended.

Data analysis:

All collected data were analyzed using SPSS. *P* values < 0.05 were considered statistically significant.

RESULT

Ninety patients diagnosed as HIV patients were enrolled in this study, the result revealed that Overall anti-syphilis rate among HIV patients using ELISA anti-syphilis total antibodies was 8(8.9%) (Figure 1). Most of them were female however all positive patients were males (Table 1). The age distribution of HIV patients ranged from 21 - 70 years, most of seropositive patients 3(3.3%) were belonged to both age groups 41-50 and 51-60 years (Table 2), were married 7(7.8%), The seroprevalence of syphilis among the married patients (7.8%) was higher than the unmarried 1(1.1%) from all patients .

Regarding HIV duration most of co-infected patients were belonged to 1-10 years group, only one patient has a duration of HIV <1 year (Figure 2). 7(7.8%) of positive patients initiate ART treatment about 1-10 years after diagnosis , only one patient has treatment for one week (< 1 year).

Among total population, 27(30%) had a past history of STIs, however most of positive results observed among who had no history of STIs 87.5% (7 out of 8), and the remain positive patient had a history of STIs before 5 months (<1 year). 23(85.2%) out of all studied patients take treatment to treat STIs symptoms, only one of them has syphilis positive result (Figure 3).

There was statistically significant difference in prevalence among age groups. Nevertheless, there was no significant difference in Residence, Occupation and past history of STDs

DISCUSSION

Syphilis and HIV co-infection presents a complex interaction. The increased incidence of HIV individuals afflicted with STD, including syphilis is epidemiologically documented in numerous studies^{18,19}.

In this study all known HIV infected patients were screened for syphilis , many of whom were on ART treatment, The overall frequency of anti syphilis antibodies was 8.9% positive, all of them were male. the age range HIV Patients was 21-70 years. The high prevalence in males could have been due to

polygamous relationships and lack of awareness of syphilis, This implies that syphilis infection is more common in adults. This finding was higher than the 2% found by Sunitha *et al* (2016) in India²⁰, 2% found by Arti *et al* (2016) also in India²¹ and 0.9% found by Mwumvaneza *et al* (2016) in Rwanda²² and slightly higher than the 6.7% found by Konda *et al* (2016) in South India²³, 6.8% found by Ozlem *et al* (2015) in Istanbul, Turkey²⁴ and 7.3% found by Techalew *et al* (2015) in Ethiopia²⁵ but less than 11.4% that showed by Shweta *et al* (2014) in India²⁶ and 10% found by Gerald *et al* (2015) in Uganda among HIV patients²⁷.

When compared with other studies in Sudan, it is slightly high to the result obtained by Ayman *-et al* (2015) a 7% of anti syphilis antibodies was detected among blood donors in Khartoum state²⁸, 6.8% obtained by Elsharif *et al* (2015) among blood donors in White Nile State²⁹ and 5.4% obtained by Nmarig *et al* (2016) among Blood donors in Khartoum Central Sudan³⁰. However, the obtained seropositivity was lower than the result of (42.2%) obtained by Eltayib-*et al* (2013) among Homeless in Khartoum state³¹ and 21.6% obtained by Osman *et al* (2014) among Adult Rural Residents in Red Sea State³².

CONCLUSION AND RECOMMENDATION

In this study we conclude the high frequency of syphilis in HIV infected population attending ART clinics so a screening of all patients for syphilis is recommended as a routine.

The appropriate diagnosis and treatment of syphilis in the HIV population could lead to better response to ART and lower transmission rates for both diseases. a variety of strategies may be needed in different localities, education and intervention programs need to be developed , which also lead to reduction in prevalence of syphilis in HIV infected patients.

ACKNOWLEDGEMENTS

We would like to thanks all participants of this study, laboratory staff of Omdurman ART clinic especially Mrs. Dina and all doctors. We also grateful to all staff of Microbiology Department AL Neelain University.

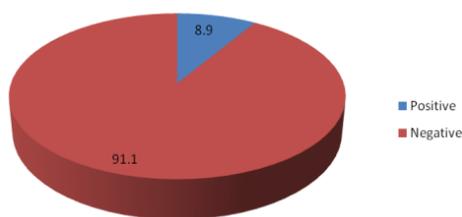


Figure 1
Frequency of Syphilis among known HIV patients (n=90)

Table 1
Frequency of syphilis among HIV patients (n=90) according to their gender

Gender	Number	Result		Total
		Positive	Negative	
Male	Count	8	36	44
	% within Result	100.0%	43.9%	48.9%
	% of Total	8.9%	40.0%	48.9%
Female	Count	0	46	46
	% within Result	0%	56.1%	51.1%
	% of Total	0%	51.1%	51.1%
Total	Count	8	82	90
	% within Result	100.0%	100.0%	100.0%
	% of Total	8.9%	91.1%	100.0%

Table 2
Frequency of syphilis among HIV patients (n=90) according to their age distribution

Age Range	Number	Result		Total	
		Positive	Negative		
Age	<=30 years	Count	0	16	16
		% within Result	0%	19.5%	17.8%
		% of Total	0%	17.8%	17.8%
	31-40 years	Count	2	31	33
		% within Result	25.0%	37.8%	36.7%
		% of Total	2.2%	34.4%	36.7%
	41-50 years	Count	3	28	31
		% within Result	37.5%	34.1%	34.4%
		% of Total	3.3%	31.1%	34.4%
	51-60 years	Count	3	5	8
		% within Result	37.5%	6.1%	8.9%
		% of Total	3.3%	5.6%	8.9%
	61-70 years	Count	0	2	2
		% within Result	0%	2.4%	2.2%
		% of Total	0%	2.2%	2.2%
Total	Count	8	82	90	
	% within Result	100.0%	100.0%	100.0%	
	% of Total	8.9%	91.1%	100.0%	

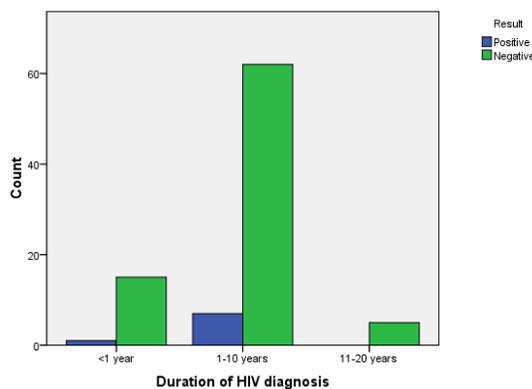


Figure 2
Frequency of Syphilis among HIV patients according to Duration of HIV diagnosis

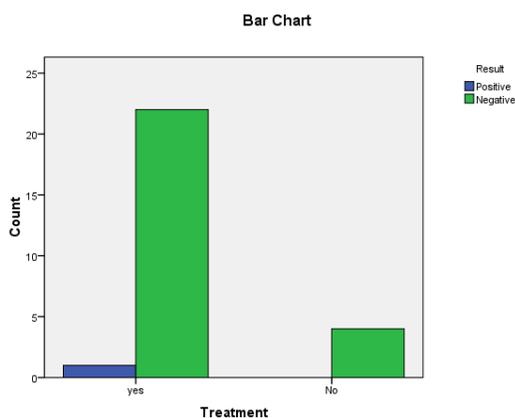


Figure 3
Frequency of Syphilis among HIV patients (n=90) regarding antiretroviral treatment

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