



Seroprevalence of *Chlamydia trachomatis* Antibodies and Associated Risk Factors Among HIV Positive Patients Seen in Enugu.

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ABSTRACT

A cross-sectional study of *Chlamydia trachomatis* antibody seroprevalence was carried out among persons living with HIV in Enugu, Nigeria. One hundred and fifty persons diagnosed of HIV and receiving treatment at the University of Nigeria Teaching Hospital, Enugu were recruited for the study. Enzyme-linked immunosorbent assay kits were used to assess the seroprevalence of *Chlamydia trachomatis* antibodies among the participants. A structured PROFORMA was used to collect data on demographic factors and high-risk behaviours. The results showed a 60.7% (91/150) of *Chlamydia trachomatis* antibodies among the study participants. Among the participants with antibodies, the IgM prevalence was 51.6% (47/91), while IgG was 35.2% (32/91) and both IgM and IgG were 13.2% (12/91). Logistic regression showed that the risk of chlamydia infection was increased with the history of STD (OR = 1.4, (0.7 – 2.7)), history of multiple sexual partners (OR = 1.9, (0.9 – 3.7)), those currently sexually active (OR = 1.5, (0.7 – 3.1)), the use of unsterilized objects (OR = 4.9, (2.3 – 10.4)) and the irregular or non-usage of condoms (OR = 10.2 (4.6 – 22.7)). The findings of the study showed a high prevalence of *C. trachomatis* antibodies among the participants. Although these may not all represent active *C. trachomatis* infection, it can be inferred that a high percentage of the population has been exposed to the infection and many of them may still be harbouring this infection inadvertently. This may contribute to the high rate of spread of this infection as well as HIV in Nigeria.

INTRODUCTION

Chlamydia trachomatis is one of the 4 bacterial species in the genus *Chlamydia* family-*Chlamydiaceae*; phylum- *Chlamydiae*. It is a Gram-negative, obligate intracellular organism and the first chlamydial agent discovered in humans in 1907¹. Two biovars of *C. Trachomatis* are responsible for human infections-the trachoma biovar which causes localised infection on the epithelial surfaces of the conjunctiva

and urogenital system and the lymphogranuloma venereum biovar responsible for genital ulceration and spread via lymphatic channels². Serovars in both *C. trachomatis* biovars cause trachoma, sexually transmitted disease, some form of arthritis, neonatal inclusion conjunctivitis and pneumonia. Infection is primarily through penetrative sexual intercourse, however infection can be detected in the conjunctiva and nasopharynx without concomitant genital tract infection³. In females, *C. trachomatis* causes cervicitis

and pelvic inflammatory disease which may lead to such complications as ectopic pregnancy and tubal factor infertility. *Chlamydia trachomatis* in the cervix can be transmitted to a neonate during passage through an infected birth canal resulting in neonatal pneumonia and conjunctivitis. In males, urogenital serovars cause non-gonococcal urethritis and epididymitis. *Chlamydia trachomatis* can also induce Reiter's syndrome, proctitis, and conjunctivitis in both sexes^{4,5}.

Over 127 million new cases of chlamydia infection were expected to have occurred in 2016 with females having a slightly higher prevalence than males⁶. A prevalence of 3.8% was reported in females in the African region, which is second only to the Americas⁷. Within Nigeria, a prevalence of 9.6% has been reported in Kano amongst patients attending infertility and sexually transmitted disease clinics⁸. Another study reported 30.2% among female undergraduates in Port Harcourt⁹. A seroprevalence study in Enugu, Nigeria reported 42.9% in 2011¹⁰. Among HIV patients, a 29.4% prevalence rate has been reported in Abuja, Nigeria¹¹ and 17.3% in Lagos¹². Although the pathogenesis of *C. trachomatis* genital infection is still poorly understood, evidence indicates that active *C. trachomatis* infection is an important risk factor for HIV transmission and vice versa^{13,14}.

However, it is often difficult to compare prevalence rates reported for *C. trachomatis* in various studies because of the wide variations in the sensitivities of the different methods used for its detection. Direct cytological testing by Giemsa stain of fixed smears is rapid and has a sensitivity of 90% for detection of Chlamydial conjunctivitis in neonates but is not sensitive enough for diagnosing genital infections or conjunctivitis in adults¹⁵. Culture for years was considered as the most sensitive test for detecting *C. trachomatis* and is the only diagnostic procedure that confirms the presence of viable organisms^{15,16}. It is 100 per cent specific but is less sensitive when compared to nucleic acid amplification tests¹⁵. Serology tests such as complement fixation (CF), microimmunofluorescent (MIF) and tests based on enzyme immunoassays (EIA) are used mainly for neonates, patients with tubal factor infertility and occasionally for LGV infections when bubo aspirates are not available¹⁵. A major disadvantage of these tests is that they also detect antibodies to other chlamydial species and not just *C. trachomatis* in addition, antibody response to previous Chlamydial infections may not even have been elicited at all¹⁶. Antigen detection tests include the Direct Fluorescent Antibody (DFA) tests and the Enzyme Immunoassays (EIA) tests that use monoclonal or polyclonal antibodies to detect chlamydial lipopolysaccharide (LPS) or the major outer membrane protein (MOMP). The DFA tests are however no longer recommended for testing genital specimens due to variations in their sensitivity¹⁶. Currently, the most sensitive tests for detecting *C. trachomatis* are the Nucleic Acid Amplification Tests (NAATs)^{16,17}. Another advantage of these NAATs is their non-reliance on

viable organisms, so samples collected in the field and transported to a laboratory can still be tested.

In view of the of the rising prevalence rates of *C. trachomatis* infection worldwide, the importance of early diagnosis and treatment in the control of the HIV/AIDS epidemic cannot be overemphasised. *C. trachomatis* disease of the genital tract is often asymptomatic, especially in women. This contributes to the high rate of spread of the disease as well as HIV by carriers if co-infected by HIV. The aim of this study was to determine the prevalence of and risk factors for *C. trachomatis* antibodies as an indicator of *Chlamydia* sp seroprevalence among HIV patients.

METHODS

Study Population

The study population consisted of people living with HIV attending the HIV clinic of the University of Nigeria Teaching Hospital, Enugu.

Study Sample

A minimum sample size of 122 was calculated using Cochran formula based on a prevalence of 17%¹². The sample size was however increased to 150 to allow for attrition. All patients who had HIV and gave their consent were included in the study.

Ethical Consideration

Ethical approval for this study was obtained from the ethical board of the University of Nigeria Teaching Hospital, Enugu.

Data Collection

Pre-tested interviewer-administered PROFORMA data collection sheets were used to obtain the biodata of the participants as well as history of any previous sexually transmitted infection (STI), HIV and *Chlamydia* risk factors such as multiple sexual partners, unprotected sexual intercourse, history of blood transfusion, use of sharp objects, etc.

Specimen collection and Analysis

Three millilitres of venous blood samples was then obtained from each participant by venepuncture and the sera separated and stored at -20°C. The IgG and IgM in the sera were detected by ELISA (Diagnostic Automation, Inc). Analysis of serum samples were performed according to the manufacturers instructions^{18,19}.

Data Analysis

The data was analysed using the statistical programme for social science (SPSS) version 25. Mean, standard deviation and range were calculated for quantitative data. Qualitative and categorical data were compared using Chi square tests. The risk of

Chlamydia infection was determined by binomial logistic regression. All analysis was done at a 95% confidence interval and a p-value less than 0.05 was considered significant.

RESULTS

Table 1 shows the demographic distribution of the study participants. The mean duration of HIV infection

for the cases was 3.92 years (SD±2.34, range=0.5-12 years).One hundred of the cases (66.67%) of the cases were females while 50 (33.33%) were males. Male to female ratio=1:2. The ages of the study participants ranged from 16-60years with a mean age of 35.9 (SD ± 9.22).Ninety-three (61.3%) were married, 38(25.3%) single, 18 (12.0%) widowed and only 1(0.75%) were divorced.

Table1: Demographic Distribution of Study Participants

VARIABLE	Frequency (n)	Percentage (%)
Age Group (years)		
15-25	16	10.67
26-35	72	48.00
36-45	37	24.67
46-55	18	12.00
56-65	7	4.67
Mean Age (±SD)	35.9 (SD ± 9.22).	
Gender		
Male	50	33.33
Female	100	66.67
Marital Status		
Married	93	62.00
Single	38	25.33
Widowed	18	12.00
Divorced	1	0.67
Occupation		
Farmer	12	8.00
Civil servant	20	13.33
Student	20	13.33
Self employed	62	41.33
Unemployed	7	4.67
Others	29	19.33

Figure 1 shows that 91 (60.7%) of the participants were had *Chlamydia* antibodies and 59 (39.3%) did not have antibodies.

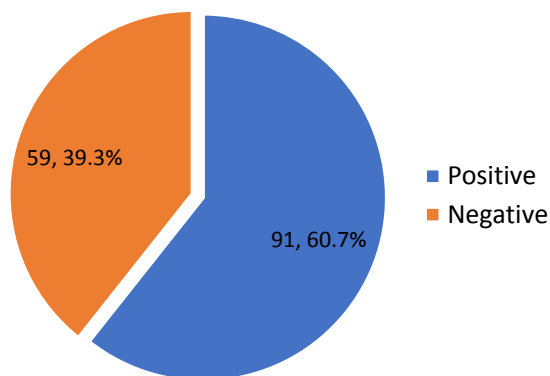


Figure 1: Seroprevalence of Chlamydia infection

Table 2 shows the distribution of the *C. trachomatis* antibodies among the study participants found to have *C. trachomatis* antibodies. The IgM prevalence was 51.6% (47/91), followed by the IgG prevalence of

35.2% (32/91) and a 13.2% (12/91) prevalence of the IgM and IgG combination among the 91 participants with *C. trachomatis* antibodies.

Table 2: Serology of *C. trachomatis* antibodies in study participants

Serology	Frequency	
	n = 91	Percent
<i>C. trachomatis</i> IgM	47	51.6%
<i>C. trachomatis</i> IgG	32	35.2%
<i>C. trachomatis</i> IgM + IgG	12	13.2%

Table 3 shows the distribution of the risk factors for *Chlamydia* infection among the study participants. There was a significant association of indiscriminate use of parenteral drugs, history of blood transfusion, use of sharp unsterilised objects and irregular or non-usage of condoms with the occurrence of *Chlamydia* antibody seroprevalence. Logistic regression showed

that the risk of chlamydia infection was increased with the history of STD (OR = 1.4, (0.7 – 2.7)), history of multiple sexual partners (OR = 1.9, (0.9 – 3.7)), those currently sexually active (OR = 1.5, (0.7 – 3.1)), the use of unsterilized objects (OR = 4.9, (2.3 – 10.4)) and the irregular or non-usage of condoms (OR = 10.2 (4.6 – 22.7)).

Table 3: Distribution of Risk Factors for Chlamydia Seroprevalence in Subjects

RISK FACTOR	Positive (N=91)	Negative (N=79)	Chi-square	Odds Ratio	p-value
History of STD	42(64.6%)	48(56.4%)	1.01	1.4 (0.7 - 2.7) ^R	0.3125
Current symptoms of STD	45(63.4%)	46(58.2%)	0.41	1.2 (0.6 - 2.3) ^R	0.518
History of multiple sexual partners	48(67.6%)	41(51.9%)	3.82	1.9 (0.9 - 3.7) ^R	0.051
Indiscriminate use of parenteral drugs	23(32.4%)	60(84.5%)	39.69	0.1 (0.03 - 0.20) 0.01 (0.001 -	0.00001*
History of blood transfusion	13(18.3%)	76(96.2%)	94.02	0.03)	0.00001*
Currently sexually active	49(69.1%)	42(59.2%)	1.49	1.5 (0.7 - 3.1) ^R	0.2207
Use of sharp unsterilised objects	57(80.2%)	32(45.1%)	18.81	4.9 (2.3 - 10.4) ^R	0.0001*
Irregular or non-usage of condoms	48(67.6%)	12(16.9%)	37.4	10.2 (4.6 - 22.7) ^R	0.0001*

*Statistically significant (p<0.05), R: Risk is increased when O.R> 1.0

DISCUSSION

Chlamydia trachomatis is a common STD worldwide and its role in the transmission and acquisition of HIV cannot be overemphasised. It is usually underdiagnosed which is due to its asymptomatic presentation in majority of cases.^{11,13} Several studies performed in the past have demonstrated an association between the presence of an STD (especially if untreated) and the predisposition to acquire and transmit HIV infection.^{6-9,16} The *C. trachomatis* seroprevalence rate of 60.7% obtained in HIV infected persons in this study when compared to previous studies is higher than that obtained in some other studies.^{2,6,9,11}

The distribution of the *C. trachomatis* antibodies among the 91 study participants found to have *C. trachomatis* antibodies showed that IgM prevalence was 51.6% (47/91), followed by the IgG prevalence of

35.2% (32/91) and a 13.2% (12/91) prevalence of the IgM and IgG combination among the 91 participants with *C. trachomatis* antibodies. Although several studies have demonstrated an increased prevalence of *C. trachomatis* infection among HIV seropositive patients.²⁰⁻²² In the current study, there was a relatively high prevalence of IgG antibodies which is indicative of past infection. The higher seroprevalence rate of *C. trachomatis* IgM in the HIV positive cases is an indication of a recent or current infection of *Chlamydia trachomatis*. Similarly, the relatively high prevalence of IgG may be attributed to the fact that it persists in the blood long after the infection has been cleared and therefore may not reflect the actual number of cases currently infected.^{21,23,24}

Logistic regression showed that the risk of chlamydia infection was increased with the history of STD, history of multiple sexual partners, those currently sexually active, the use of unsterilized

objects, and the irregular or non-usage of condoms. These are consistent with the findings of epidemiological studies which have consistently detected the main risk factors for *C. trachomatis* infection as frequent multiple sexual relationships and failure to use or erratic use of condoms.^{20–22,25}

CONCLUSION

A relatively high seroprevalence of *C. trachomatis* antibodies was observed among people living with HIV. Although these may not all represent active *C. trachomatis* infection, it can be inferred that a high percentage of the population have been exposed to the infection and may have been recently infected as well. This has implications for continued transmission of this STI in Nigeria.

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