

## Association of Chlamydia serology with HIV in Nigerian women

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### ABSTRACT

This research was carried out to detect the presence of Chlamydia in pregnant women and gynaecologic patients in the North-central geopolitical zone of Nigeria. Blood samples were collected and analysed by ELISA techniques. The blood samples were also screened for HIV infection. A sero-prevalence of 59.0% was recorded for the study area. The sero-prevalence was higher among the gynaecologic patients (62.0%) than the pregnant women (57.5%). The difference was statistically significant ( $P = 0.0001$ ). Of the four centers chosen for the study, the Federal Capital Territory (Abuja) had the highest prevalence (84.7%), while Niger State had the least (28.7%). The difference was also statistically significant ( $P < 0.0001$ ). The prevalence rate of HIV among the participants in the study center was found to be 17.2%. Abuja had the highest prevalence rate of 24.6%, followed by Benue, 16.7%, then Kogi, 12.0% and finally, Niger, 4.7%. Chlamydia was found to be associated with HIV ( $p < 0.0001$ ). The sero-prevalence of chlamydia in the North-Central zone of Nigeria was found to be high. Chlamydia was found to be correlated with HIV in the study area and may have contributed to the zone emerging with the highest HIV prevalence in the country. For an infection that is largely asymptomatic but has devastating effects on populations, only a preventive approach would have beneficial effects in controlling the disease and its effects on women's health in the country.

**Keywords:** *Chlamydia trachomatis*, HIV, pregnant women, gynaecologic patients, co-infection

### INTRODUCTION

Chlamydial infection is the most common curable bacterial sexually transmitted disease (CDC, 2006; WHO, 2011). The incidence of chlamydial infections in women has increased dramatically from 79 to 467 per 100,000 between 1987 and 2003 (Sexually transmitted disease surveillance, 2004). According to the World Health Organization (WHO, 2011), 101 million chlamydial infections are detected annually worldwide. In the U.S. the Centres for Disease Control and Prevention (CDC) estimates that 2.8 million people are infected each year (CDC, 2006). In some parts of the third-world countries, more than 90 per cent of the population is infected (Gomes *et al.*, 2007). Chlamydial infection is caused by *Chlamydia trachomatis*, a coccoid bacillus closely related to Gram negative bacteria (Cheesbrough, 2000). *Chlamydia trachomatis* belongs to the genus *Chlamydia* which includes organisms previously called the Psittacosis Lymphogranuloma venerum Trachoma group (PLT) or the TRIC (TRachoma Inclusion

Conjunctivitis group) (Mackie & Mac Cartney, 1989).

*Chlamydia trachomatis* has 15 immunotypes (serotypes) viz; A-C which cause trachoma (chronic conjunctivitis endemic in Africa and Asia), D-K, which cause genital tract infection and L1-L3 responsible for lymphogranuloma venerum (associated with genital ulcer disease in tropical countries). *Chlamydia trachomatis* serovars D-K cause curable sexually transmitted disease called Chlamydia (CDC, 2006). The disease which arises from the infection of the lower genital tract is one of the most prevalent sexually transmitted diseases in the world (Gerbase *et al.*, 1998; Beagley and Timms, 2000). It is usually asymptomatic, in fact 50% of men and 80% of women are asymptomatic, and for this reason it is referred to as silent disease (Gaydos *et al.*, 1998; CDC, 2006; Ward *et al.*, 2007). In cases where symptoms are present they may last only a few days and may not be noticed or considered significant (Kidshealth, 2006). Clinical symptoms in women, where present include

vaginal discharge, dysuria, easily induced endocervical bleeding, irregular menstruation or intermenstrual bleeding, dyspareunia, lower abdominal pain, genital itching, increased urination frequency and sore in the vagina (CDC, 2006; Al-Mutairi *et al.*, 2007).

Chlamydia is one of the non – ulcerative sexually transmitted infections which elicit localised inflammations and immune responses characterised by the infiltration and accumulation of immune cells expressing CD4 surface proteins essential for the binding of HIV prior to entry, thus also facilitating the entry of HIV (Altes *et al.*, 2002; Joyee *et al.*, 2005; Wodarz and Hamer 2007). In a study conducted in the South- Eastern part of Nigeria by Nwaguma *et al.* 2009, it was found out that the prevalence of *C. trachomatis* infection observed in the HIV – seropositive subjects (50%) was much higher than the prevalence in the HIV-seronegative subjects (17.6%) (Nwaguma *et al.*, 2009).

Chlamydia, if left untreated can persist for at least 15 months (Mc Cormack *et al.*, 1979) and can have potentially serious lifetime consequences which include pelvic inflammatory disease (PID) (Stamm *et al.*, 1984, Land, 2010). PID is a general term that refers to infection and inflammation of the upper genital tract in women. It can affect the uterus (womb), fallopian tubes, ovaries and other organs related to reproduction (Moss, 2001). In USA, approximately 20-30 per cent of PID cases have been attributed to *C. trachomatis* (Soper, 2010). The incidence rates of PID in Nigeria vary between 0.28 and 4.4% of deliveries (Iloabachi, 1990). Sequelae of PID include ectopic pregnancy and tubal infertility (Buchan *et al.*, 1993). The risk of developing sequelae is dependent on the number of PID episodes (Westrom, 1994). Epidemiological studies have shown that sexually transmitted pathogens, including non-ulcerative agents such as *C. trachomatis*, may serve as biological cofactors for human immunodeficiency virus (HIV) seroconversion (Plummer *et al.*, 1991; Laga *et al.*, 1993; Grosskurth *et al.*, 1995). However, basic data, such as the incidence and

prevalence of many sexually transmitted diseases (STDs), which are necessary for obtaining estimates of their impact on sexual transmission of HIV, are relatively scarce in developing countries. In many areas, diagnosis of *C. trachomatis* genital infection is only performed in selected populations and is often based on the presence of clinical symptoms. Considering the high rate of asymptomatic chlamydial infection, particularly in women, a substantial “silent” or undetected epidemic of *C. trachomatis* infections could put this population at significant risk for HIV infection. A reliable epidemiological data is needed to determine the prevalence rate of the disease in the populations which will help in devising an effective chlamydia control program. It is in view of the above that this research work was carried out to detect the presence of Chlamydia in pregnant women and gynaecologic patients in the North-central geopolitical zone of Nigeria and to check the rate of occurrence of Chlamydia – HIV co-infection.

## **METHODOLOGY**

By simple random sampling method, three states (Benue, Kogi and Niger) and the Federal Capital Territory were selected from the North-Central geo-political zone. The Federal Medical Centers, Makurdi, and Kogi, the National Hospital, Abuja and the General Hospital Minna were chosen for the study. The study groups comprised 400 pregnant women and 200 gynaecologic patients attending the gynecological clinics for PID, infertility and ectopic pregnancy. Only those who had not been on antibiotics (azithromycin, erythromycin, doxycycline, or tetracycline) in the past three months before sampling were included in the study. Each subject gave her consent and the study was approved by the Ethical Committees of the Hospitals. Data were analyzed using Chi-square test and  $P \leq 0.05$  was considered statistically significant.

### **Blood sample collection**

Five (5) ml of blood sample was collected from each participant by venepuncture and stored in venoject vacutaneers and allowed to clot. The sera were separated by spinning the blood in a

centrifuge at 3000 rpm and stored at -20° C till use.

**Anti – Chlamydia trachomatis ELISA**

*Chlamydia trachomatis* IgG ELISA test kit by DIAGNOSTIC AUTOMATION, INC, Calabasas was used. It employs the LGV type 2 broadly reacting antigen of *Chlamydia trachomatis*. It detects *Chlamydia trachomatis*, *Chlamydia psittaci* and *Chlamydia pneumonia* (TWAR) antibodies. It was performed according to the manufacturer’s instruction.

**Test for HIV**

Determine kits for the test of HIV1/2 manufactured by Alere Medical Co. Japan were used to screen the blood samples. It is an in vitro, visually read, qualitative immunoassay for the detection of antibodies to HIV-1/2 in human serum, plasma or whole blood. The Determine positive samples were further tested using HIV 1/2 STAT-PAK DIPSTICK kit manufactured by Chembio Diagnostic Systems, INC, USA. Both tests were carried out according to the manufacturers’ instructions.

**RESULTS**

The study population was made up of two groups, pregnant women and gynaecologic patients. Of the 400 pregnant women tested, 230 were positive for Chlamydia IgG with a prevalence rate of 57.5% while 124 out of the 200 gynaecologic patients tested positive giving

a prevalence rate of 62.0%. The results are as shown in Table 1.

The six hundred blood samples constituted one hundred and fifty (150) from each of the Federal Capital Territory (Abuja), Benue, Kogi and Niger States. Of the 150 samples from each study center, 100 were from pregnant women and 50 from gynaecologic patients. Of the 150 samples assayed from each of the centers, 127, 107, 77 and 43 samples were found positive for the Federal Capital Territory (Abuja), Benue, Kogi and Niger States respectively giving prevalence rates of 84.7%, 71.3% 51.3% and 28.7% respectively. The sero-prevalence of Chlamydia IgG antibodies in parts of the North-Central Nigeria is shown in Table 2.

All the participants in the study groups were also screened for HIV to determine the relationship between Chlamydia and HIV in them. Of the 600 samples tested for HIV, 103 were positive giving a prevalence rate of 17.2%. Abuja had the highest prevalence rate of 24.6% (37 in 150), followed by Benue, 16.7%, (25 in 150) then Kogi, 12.0% (18 in 150) and finally, Niger, 4.7% (07 in 150). The results of HIV prevalence in the North Central are shown in Table 3.

As for relationship between Chlamydia and HIV, 61 subjects tested positive for both Chlamydia and HIV, 293 were positive for Chlamydia only, while 18 had only HIV. Results are shown in Table 4.

**Table 1:** Seroprevalence of Chlamydia IgG antibodies in parts of the North-Central Nigeria based on the study groups

Group	No. Screened	No. Pos	% Prevalence
Pregnant	400	230	57.5
Gynaecologic	200	124	62.0
Total	600	354	59.0

X<sup>2</sup>=1.11 P= 0.292

**Table 2:** Seroprevalence of Chlamydia IgG antibodies in parts of the North-Central Nigeria

Part	No. Screened	No. Positive	% Prevalence
Abuja	150	127	84.7
Benue	150	107	71.3
Kogi	150	77	51.3
Niger	150	43	28.7

X<sup>2</sup> = 111, (P<0.0001)

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Table 3: Prevalence of HIV 1/2 among the study participants in parts of the North-central

	No. Screened	No. Positive	% prevalence
Abuja	150	37	24.6
Benue	150	25	16.7
Kogi	150	18	12.0
Niger	150	07	4.7
Total	600	87	14.5

Table 4: Relationship between Chlamydia and HIV in parts of the North Central

	No. Screened	Positive	%prevalence
All Chlamydia	600	354	59.0
All HIV	600	79	13.2
Chlamydia only	600	293	48.8
HIV only	600	18	3.0
Chlamydia and HIV	600	61	10.2

$X^2 = 779.7$

$p < 0.0001$

### DISCUSSION

The seroprevalence of Chlamydia IgG antibody in parts of the North central Nigeria was found to be 59.0% in this study. This high prevalence is in line with the report by (CDC) in 2005 that in the United States, chlamydial genital infection is the most frequently reported infectious disease, and the prevalence is highest in persons aged <25 years (CDC, 2005). It is also similar to the findings of Mawak *et al.*, 2011 and Dibua *et al.*, 2013 which was 56.1% among gynaecologic clinic attendees in Jos and Nsukka respectively. A 51.0% prevalence among pregnant and non-pregnant women and their spouses attending pre and antenatal clinic in the College of Medicine of the University of Lagos has been reported (Okoror *et al.*, 2000). A slightly lower prevalence of 40.7% of *C. trachomatis* has also been reported from the South-Eastern part of Nigeria (Okoror *et al.*, 2007). Other lower prevalences of 38.3% in Zaria (Tukur *et al.*, 2006) and 13.3% in Benin City (Isibor *et al.*, 2005) have also been reported.

The high prevalence found in this part of the country in this study can be considered as an evidence of endemicity in the population and this may be due to the fact that Chlamydia infections usually present with no clear cut symptoms and are as a result left untreated or mistaken for other infections such as gonorrhoea. This often leads to serious sequelae such as pelvic inflammatory disease

(PID), endometritis, salpingitis, ectopic pregnancy and infertility (Mabey *et al.*, 1992).

Other factors that could be attributable to the high prevalence and endemicity are sociocultural inhibition that prevents women from reporting sexual symptoms and non-availability of facility to detect the causative agent (*Chlamydia trachomatis*) in many health care centres in this part of the world (Okonofua *et al.*, 1995). The high prevalence could have also been caused by the high prevalence of HIV-AIDS in the area. According to the Technical Report of the 2010 National HIV Sero-prevalence Sentinel Survey, the North-central geopolitical zone had the highest prevalence rate of AIDS and studies have shown the association between the two infections (chlamydia and HIV) such that the presence of one facilitates that of the other. Genital Chlamydia infection has been linked to an increasing risk for acquisition of HIV disease (Laga *et al.*, 1993; Ho *et al.*, 1995; Brunham, *et al.*, 1996; Stamm, 1999; Joyee, *et al.*, 2005) while on the other hand, immunosuppression due to HIV may lead to more severe Chlamydia disease condition like PID in those who are infected with *Chlamydia trachomatis* (Thomas, *et al.*, 2002). Thus early diagnosis and treatment of Chlamydia infections is important to prevent HIV risk and devastating clinical consequence. The two infections (Chlamydia and HIV infections) have also being reported to have common risk factors, for instance, Joyee,

*et al.*, (2005) reported that both HIV and Chlamydia are significantly associated with some risk factors such as multiple sexual partners.

The possible relationships between HIV infection and *Chlamydia trachomatis* are that the invasive intracellular pathogenesis of *Chlamydia trachomatis* can cause substantial damage to the genital epithelial layer which may facilitate HIV infection (Hitchcock, 1999) and immunological changes due to HIV infection may favor *Chlamydia trachomatis* (Debattista *et al.*, 2002). Chlamydia is one of the non – ulcerative sexually transmitted infections which elicit localised inflammations and immune responses characterised by the infiltration and accumulation of immune cells expressing CD4 surface proteins essential for the binding of HIV prior to entry, thus also facilitating the entry of HIV (Altes *et al.*, 2002; Joyee *et al.*, 2005; Wodarz and Hamer, 2007).

The results of this study also showed that the prevalence of Chlamydia was slightly higher among gynaecologic cases (62.0%) than in the pregnant women (57.5%). This is similar to an earlier report in Delta State by Omo-Aghoja *et al.* (2007), that the prevalence of serum Chlamydia antibody was significantly higher in cases (65.8%) (tubal infertility) compared with controls (pregnant women) (17.3%;  $P < 0.01$ ). Although it is not recommended for the diagnosis of lower genital tract infections, or for screening in asymptomatic patients, serological testing has been shown to be useful for diagnosing LGV, neonatal pneumonia and upper genital tract infections, and for the evaluation of tubal-factor infertility (Persson, 2002; Be'be'ar and de Barbeyrac, 2009).

Conclusively, the sero-prevalence of chlamydia in the North-Central zone of Nigeria was found to be high and it was found to be associated with HIV in the study area and may have contributed to the zone emerging with the highest HIV prevalence in the Country. Future studies are required to investigate the genotypes of chlamydia in circulation in the study population and to investigate the role of

*Chlamydia trachomatis* genotypes in disease manifestations.

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