Prevalence of Chlamydia Trachomatis Infection among Female Undergraduates of the University of Port Harcourt Using Strand Displacement and Amplification [SDA] Technique

Type of Article: Original

***Kennedy Tamunomiebam Wariso, **John Odigie, **Sunny Eyaru**

Departments of *Medical Microbiology and Parasitology and **Pharmacology and Toxicology, University of Port Harcourt, Port Harcourt, Nigeria.

ABSTRACT

Background: Chlamydia trachomatis infection, being largely asymptomatic, is difficult to diagnose using the common diagnostic methods which have varying degrees of sensitivity and specificity. There is a paucity of data on the prevalence of Chlamydia trachomatis infection in Nigeria. The aim of this research is to determine the prevalence of and predictive risk factors for Chlamydia trachomatis infection among female undergraduate students of the University of Port Harcourt.

Methods: Four hundred undergraduate, non pregnant, asymptomatic female students below the age of 30 years were randomly selected and given questionnaires with self administrable vaginal swab sticks. The participants completed the questionnaires and provided vaginal swab samples which were analyzed using Strand Displacement and Amplification Technique.

Results: Of the 400 sexually active participants, 44 tested positive [prevalence rate of 11%] for Chlamydia trachomatis. Some of the associated risks factors identified were, having multiple sexual partners especially in the last 90 days, irregular contraceptive usage and past history of sexually transmitted infections.

Conclusion: There is an urgent need for a national policy on routine screening for Chlamydia trachomatis as treatment is cheap and effective, while the morbidity resulting from delayed diagnosis is more difficult to manage and associated with severe sequelae.

Key Words: – Prevalence; Chlamydia trachomatis; Strand Displacement and Amplification [SDA] Technique

Correspondence: DR K.T. Wariso

Email - ktwariso@yahoo.com

INTRODUCTION

Genital Chlamydia trachomatis infection is the most common curable sexually transmitted infection [STI] worldwide[1]. Chlamydia trachomatis is the most prevalent bacterial cause of STI in men and women worldwide, with about 50 million new infections yearly[2,3]. It mostly runs an asymptomatic course in women[4] and men[5], therefore it remains undetected and subsequently untreated. While the main burden of morbidity falls upon women[6], the challenge of Chlamydia control remains the identification of asymptomatic individuals. The inherent morbidity that results from asymptomatic Chlamydia trachomatis infection has necessitated the screening for Chlamydia trachomatis among women who are aged 25 years and above[5]; unmarried, separated or divorced[6]; or with past histories of STI's[7]; or with new or multiple sexual partners within the past 90 days[8] and; women who inconsistently use barrier form of contraception[9].

Although the major impact of disease caused by Chlamydia trachomatis is on the female reproductive tract, this agent also causes infections in men and children. The prevalence of Chlamydia infection in sexually active adolescent women, the population considered most at risk generally exceeds 10%[10], and in some STI clinic populations of women, the prevalence can reach 40%[10]. The prevalence of Chlamydia trachomatis infection ranges from 4 to 10% in asymptomatic men and from 15 to 20% in men attending STI clinics[11].

Although Chlamydia trachomatis can be easily treated with antibiotics, control of the infection has been impeded, primarily because the symptoms of the infection are often absent or insufficient to lead to diagnosis, especially among many affected women[12]. If untreated, it may lead to severe sequelae ranging from pelvic inflammatory disease [PID], through tubal scarring to ectopic pregnancy and tubal infertility[7]. Also, about one-third of women with untreated Chlamydial infection go on to develop PID, by ascending infection[13] and PID is the predominant infectious cause of chronic pelvic pain and tubal infertility[2,14,15]. Chlamydial genital infection during pregnancy increases the risk of spontaneous abortion, premature delivery and ectopic pregnancy. Neonates delivered vaginally from infected mothers may contract Chlamydial conjunctivitis or pneumonia[14]. Chlamydial infections in newborns occur as a result of perinatal exposure. It is known that about 65% of babies born from infected mothers become infected during vaginal delivery[16].

Chlamydia trachomatis is a spherical or ovoid, non motile, gram negative, obligate, intra cellular bacterium thought initially to be a virus because of the intracellular life cycle. It is metabolically deficient in its ability to synthesize ATP and thus requires an exogenous source of this high energy compound. This is why it is called an energy parasite. It undergoes a unique biphasic developmental cycle, forming distinctive intracellular inclusions that permit identification by light or fluorescent microscopy.

Chlamydia trachomatis is susceptible to a broad spectrum of antibiotics, particularly the tetracyclines and macrolides. Because the Chlamydial cell wall is different from that of many bacteria, beta-lactam antibiotics such as penicillin lack bactericidal activity against these microorganisms. Chlamydia trachomatis includes the agents of trachoma, lymphogranuloma venereum [LGV], urogenital tract disease, and inclusion conjunctivitis.

Strand Displacement and Amplification [SDA] technique is a form of Nucleic Acid Amplification Technique [NAAT] with a high specificity and sensitivity in the diagnosis of Chlamydia trachomatis infection[20,21]. The technique allows for the use of not only urine but also self administered vaginal swab as the clinical specimen. NAAT has revolutionized the diagnosis of Chlamydia trachomatis. Urine based screening using NAAT has comparable sensitivity and specificity to cervical and urethral specimens[21,22].

This study is aimed at determining the prevalence of Chlamydia trachomatis infection, and identification of the risk factors associated with genital Chlamydia infection among female undergraduates of the University of Port Harcourt.

MATERIALS AND METHODS

This study was carried out between January and June 2009 at the University of Port Harcourt Teaching Hospital, Port Harcourt. Ethical approval was obtained from the relevant boards at the University of Port Harcourt and the University of Port Harcourt Teaching Hospital.

Inclusion criteria were that subjects should be healthy, female undergraduates aged between 16 and 30, should not be on any antibiotics prior to the collection of samples, and should respond to all or most of the questions asked in the questionnaire. The sample size determined using the Kish formula was 400 with prevalence rates of 10.5%, at 95% confidence interval, and degree of accuracy as 0.05.

Informed and written consent was obtained from every participant. Those who consented were required to fill in questionnaires which detailed the predisposing/risk factors associated with Chlamydia trachomatis. They were also given vaginal swab for self administered vaginal swabbing. This process was explained to each participant by trained clinical students. All the self admissible vaginal swabs [Genital BBL Culture swab EZ] were quickly transported to the laboratory and analyzed by SDA.

RESULTS

Of the 400 students screened, only 44 of them were positive for Chlamydia trachomatis infection. These constitute about 11% of the total students screened. From the data obtained, students aged between 28 and 30 years have a significantly increased chance of contracting Chlamydia trachomatis infection compared to the other age ranges [Table 1], with a p value of <0.05, $X^2 = 22.081$ for linear trend and Odds ratio of 0.6.

Table 1: Prevalence of students with Chlamydia trachomatis infection according to age range

	Chlamydia trachomatis	
No. of Sex Partner	[Positive] Infected	[Negative] Not Infected
None	4[5.3%]	72[94.7%]
One	4[1.8%]	224[98.2%]
Two	8[16.7%]	40[83.3%]
Three	24[54.5%]	20[45.5%]
Four	4[100%]	0[0%]
TOTAL	44[11.0%]	356[89.0%]

The data obtained also showed that about 5.3% of those with no sexual partners at the time of study were infected compared to those with up to four sexual partners who had 100% infectivity [Table 2], with a p value of <0.05 using Fischer's test of exactness, $X^2 = 28.10$, RR = 19.00 ($7.32<RR<49.32$).

Table 2: Prevalence of students with Chlamydia trachomatis infection in relation to number of sexual partners

	Chlamydia trachomatis	
Age Range	[Positive] YES	[Negative] NO
[16-18]	8[33.3%]	16[66.7%]
[19-21]	4[5.3%]	72[94.7%]
[22-24]	12[8.8%]	124[91.2%]
[25-27]	0[0%]	120[100%]
[28-30]	20[45.5%]	24[54.5%]
TOTAL	44[11.0%]	356[89.0%]

Among the students that judiciously use condom during sexual intercourse, none of the 112 student respondents were infected while 39(17.5%) of those who do not use condom regularly were infected. This was statistically significant with a p value of <0.05, $X^2 = 21.99$.

The prevalence of Chlamydia infection among students with previous sexually transmitted infections (STI) is shown in Table 3, with 15.2% of subjects with previous STI testing positive them compared to 8.8% of those without a previous history of sexually transmitted disease who were also infected. This was statistically significant with a p value of <0.05, $X^2 = 4.58$.

TABLE 3. Prevalence Chlamydia Trachomatis Infection in Students With A Previous History Of Sexually

PREVIOUS STD	CHLAMYDIA TRACHOMATIS	
	INFECTED	NOT INFECTED
Yes	20 (15.2%)	112 (84.8%)
No	20(8.8%)	228(91.2%)

Table 4 shows the prevalence of Chlamydia trachomatis infection in relation to the number of sexual partners in the past 90days. Subjects with 3 and 4 or more sexual partners had significantly higher rates of Chlamydia positivity with a p value of <0.05, $X^2 = 12.30$, RR = 3.99($1.15<RR<10.01$).

Table 4: Prevalence of students with Chlamydia trachomatis infection in relation to number of sexual partners in the past 90 days.

	Chlamydia trachomatis	
No. of sex partners in past 90days	YES[infected]	NO[not infected]
None	0[0%]	12[100%]
One	4[2.0%]	196[98.0%]
Two	0[0%]	52[100%]
Three	4[14.3%]	24[85.7%]
Four or more	36[56.3%]	28[43.7%]

DISCUSSION

The present study revealed a prevalence rate of 11% [44 of the 400 subjects] for Chlamydia trachomatis infection. Possible predictive risk factors gotten as result of the study include increasing age [28 to 30 years], multiple sexual partners, inconsistent contraceptive usage, and a past history of STI.

The prevalence of Chlamydia infection in our study exceeds that from reported studies, which indicate a prevalence of 10% in sexually active adolescent women[10], considering that sexual activity was not an inclusion criterion in our study.

The documented prevalence of Chlamydia infection differs considerably depending on the methodologies for the identification leading to different prevalence rates even in similar populations. Techniques used in identification include antibody detection, antigen detection, culture, and nucleic acid amplification. Amplification methods use different approaches to achieve the amplification of low copies of nucleic acid to amounts that can subsequently be detected. This method has a better sensitivity than culture and allows the detection of infection in individuals with a low number of infectious units[20].

The probable predictive risk factors from the present study are in keeping with those from previous studies; increasing age[5], sexual activity[6]; past histories of STI's[7]; multiple sexual partners[8]; and poor contraceptive usage[9].

Treatment of Chlamydial infections, save for being cheap and effective, have an added benefit: treatment of Chlamydial infections could delay the spread of human immunodeficiency virus [HIV] as Chlamydia trachomatis infections are known to increase the risk for HIV infection[17], with women infected with Chlamydia up to five times more likely to become infected with HIV[17] because Chlamydia induces inflammation which results in an increased recruitment of CD_4 lymphocytes into the genital tract leading to an increase in HIV targets and an increased HIV replication is thought to contribute to enhanced transmission[17].

The biggest challenge to the treatment and control of Chlamydial disease is that as many as 70 to 80% of women infected with Chlamydia trachomatis are asymptomatic but still contagious, resulting in a large reservoir of unrecognized, infected individuals who are capable of transmitting the infection to sexual partners. Contributing to this challenge is the fact that immunity following infection is thought to be type specific, not organism specific and only partially protective[18]; hence, recurrent infections are common. Evidence suggests that the risk of developing sequelae such as ectopic pregnancy or infertility increases with each successive episode of infection[19].

Considering that all subjects that were positive for Chlamydia trachomatis were asymptomatic and the morbidity caused by Chlamydia trachomatis infection, the relevance of routine screening for Chlamydia trachomatis is strongly highlighted as treatment is cheap and effective, while the attendant morbidity resulting from delayed diagnosis is more difficult to manage and associated with severe sequelae. Screening should be cheap, readily accessible with acceptable degrees of sensitivity and specificity, and non-invasive.

The SDA provides high specificity and sensitivity in the diagnosis of Chlamydia trachomatis infection[20], allows for the use of urine and vaginal swabs, with urine based screening having comparable sensitivity and specificity to cervical and urethral specimens[12, 21,22]. Though various questions have been asked concerning the cost-effectiveness and eligibility of routine screening[23], age-based routine screening provides the greatest cost-saving strategy to identify those infected with Chlamydia trachomatis[24,26], considering the economic importance of long term sequelae from infection.

The paucity of data on the prevalence of Chlamydia trachomatis infection in Nigeria using SDA is due primarily to the lack of policy on routine screening for Chlamydia trachomatis using NAAT, unlike in the first world where it is done for populations where prevalence exceeds 3%[26]. There is an obvious need to assess the trend of STI including genital Chlamydia trachomatis infection among young, asymptomatic, non-pregnant females.

CONCLUSION

There is an urgent need for a national policy on routine screening for Chlamydia trachomatis as treatment is cheap and effective, while the attendant morbidity resulting from delayed diagnosis is more difficult to manage and associated with severe sequelae.

REFERENCES–

1. World Health Organization. Global prevalence and incidence of selected sexually transmitted infections. Overview and estimates. Geneva: WHO, 2001.
2. Awwad Z M, Al-Amarat AA , Shehabi AAA. Prevalence of genital Chlamydial infection in Symptomatic and Asymptomatic Jordanian patients. Int J. Infect Dis. 2003; 7: 206–209.
3. Mpiga P , Ravaoarimoro M. Chlamydia trachomatis persistence: An update. Microbiological Research. 2006; 161: 9–16.
4. World Health Organization Task Force on Prevention and Management of Infertility. Tubal infertility: Serological relationship to Past Chlamydia and Gonococcal infection. Sex Transm Dis. 1995: 22: 71–77.
5. Scieh, ISD, National Public Health Service for Wales. Renewing the focus, HIV and other sexually transmitted infections in the United Kingdom in 2002. London: Health Protection Agency, 2002.

6. Howell MS, Quinn TC, Braithwaite W, et al. Screening women for Chlamydia trachomatis in family planning clinics. The cost effectiveness of DNA amplification assays. Sex Transmit Dis .1998: 25: 108-117.
7. Adams E. J, Charlett A, Edmunds WJ, et al. Chlamydia trachomatis in the United Kingdom: A systematic review and analysis of prevalence studies. Sex Transm Infect 2004: 80: 354–362.
8. Committee for Quality Assurance. State of health care quality report, 2003. Washington DC: National Committee for Quality Assurance, 2003: 1–60.
9. Meyers DS, Halvorson H, Luckhaupt S. Screening for Chlamydial infection: An evidence update for the United States Preventive Services Task Force. Ann Intern Med. 2007: 147: 100-200
10. Center for Disease Control and Prevention. Recommendations for the prevention and management of Chlamydia trachomatis infections. Morbid. Mortal. Weekly Rep. 1993; 42 [No, RR-12]:1-39.
11. Stamm WE, Koutsky LA, Benedetti JK, Jourdeu JL, Brunham RC and Holmes KK. Chlamydia trachomatis urethral infections in men. Prevalence, risk factors and clinical manifestations. Am. Intern. Med. 1984; 100: 47-51.
12. Cheng HM, Macaluso SH Vermund HH , Hook EW. Relative accuracy of nucleic acid amplification test and culture in detecting Chlamydia in asymptomatic men. Journal of Clinical Microbiology. Nov, 2001; 39. II: 3927–3937.
13. Obunge OK, John CT. The role of genital Chlamydial infection in Acute Pelvic Inflammatory Disease. Afr. J. Cln. Exper. Microbio. 2007; 8910: 23–27
14. Parratt JR, Hay DP. Sexually transmitted infections. Current Obstetrics and Gynecology, 2003; 13: 224–230.
15. Debattista J, Timms P, Allan J, Allan JA. Immuno Pathogenesis of Chlamydia trachomatis infections in women. Fertility and Sterility.2003; 79, 6: 1273-1287.
16. Schachter JM, Grossman M, Sweet RL, Holt J, Jordan C , Bishop E. Prospective study of perinatal transmission of Chlamydia trachomatis. JAMA. 1986; 255: 3374- 3377.
17. Laga M, Mamoka M , Kivuvu B, et al. Non- ulcerative sexually transmitted diseases as risk factors for HIV-1 transmission in women: Results from a cohort study. AIDS 1993; 7: 95-102.
18. Pearlman MD , McNeeley SG. A review of the Microbiology, Immunology and Clinical implications of Chlamydia trachomatis infections. Obstet. Gynecol. Surv. 1992; 47: 448-461.
19. Hills SD, Nakashima A, Marchbanks PA, Addiss DG , Davis JP. Risks factors for recurrent Chlamydia trachomatis in women. Am. J. Obstet. Gynecol.1994; 170: 801-806.26. Jungkind DS, Direnzo KG, Beavis R and Silverman NS. Evaluation of automated COBRAS AMPLICOR PCR system for detection of several infectious agents and its impact on laboratory management. J. Clin. Microbiol. 1996; 34 : 2778–2783.
20. Black CM. Current methods of laboratory diagnosis of Chlamydia trachomatis infections. Clin. Microbiol. Rev. 1997; 10: 160-184.
21. Schepetiuk S, Kok T , Martin L, et al. Detection of Chlamydia trachomatis in urine samples by nucleic acid tests. Comparison with culture and enzyme immunoassay of genital swab specimens. J. Clin. Microbiol 1997; 35: 3355–3357.
22. Cook RL, Hutchison SL, Ostergard L, Braithwaite RS and Ness RB. Systematic review, non invasive testing for Chlamydia trachomatis and Nesseria gonorrhea. Ann Intern Med 2005; 142: 914–925.
23. Paavonen J. Is screening for Chlamydia trachomatis cost effective? Genitour Med. 1997; 73: 103–104.
24. Howell RM, Thomas CQ, Charlotte AG. Screening for Chlamydia trachomatis in Asymptomatic Women Attending Family Planning Clinics: A Cost-Effectiveness Analysis of Three Strategies. Annals of Internal Medicine 1998; 128: 277–284.
25. Nyari T, Wood MW , Kovacs L. Should all sexually active young women in Hungary be screened for Chlamydia trachomatis? European Journal of Obstetrics and Gynecology and Reproductive Biology 2003; 106: 55–59.
26. Jungkind DS, Direnzo KG, Beavis R, Silverman NS. Evaluation of automated COBRAS AMPLICOR PCR system for detection of several infectious agents and its impact on laboratory management. J. Clin. Microbiol. 1996; 34 : 2778–2783.