Detection of *Chlamydia Trachomatis* Antigen among Attendees of a Fertility Clinic in Abeokuta, Ogun State, Nigeria

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Abstract: Infertility is the biological inability of a man or woman to contribute to conception. *Chlamydia trachomatis* is the most implicated organism in infertility. The objective of our study was to determine the prevalence of recent *Chlamydia trachomatis* infection by antigen detection in couples with various forms of infertility. Three hundred and fifteen (315) subjects comprising 108 (24.3%) males and 207(75.7%) females, attending fertility clinic at Federal Medical Center, Abeokuta. High vaginal swabs and semen were collected and processed for microscopy; semen was analyzed for count and morphology following standard methods. *Chlamydia trachomatis* antigen was tested on all samples using an immunochomatographic rapid test kit. Overall prevalence rate of *Chlamydia trachomatis* was found to be 9.8%. Prevalence rate of *Chlamydia trachomatis* was highest in age group 31-40 years of age, when compared to other age groups. It also showed that *Chlamydia trachomatis* antigen was more prevalent among females (11.6%) compared to their male counterparts having 6.5% positivity. Antigen positivity was highest in subfertile group (26.3%) and lowest in secondary infertility group (8.3%). It was also higher in Azoospermic male subjects than Oligozoospermic or normal ranged subjects. From the findings of this study, it can be concluded that there was no strong independent evidence showing any association between *Chlamydia trachomatis* antigen positivity and infertility in couples at Abeokuta, Ogun State, Nigeria, although more case controlled studies are needed to further investigate any relationships between infertility and recent Chlamydia infection in our environment. However, there is strong evidence that adult couples in the prime of their reproductive age pose the risk of exposure to *Chlamydia trachomatis*. Thus, concerted efforts to address possible risk factors which are proxies to acquisition of *Chlamydia trachomatis,* and better health seeking behavior by couples intending to have children will likely reduce the burden of infertility in Nigerian couples.
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1. INTRODUCTION

Reproductive endocrinologists define infertility as the inability of a couple to conceive after 6 months of contraceptive free intercourse , if the female partner is less than 35years of age , or if the couple does not conceive after 12 months of contraceptive free intercourse or if the female partner cannot carry a pregnancy to full term (ARSM, 2008). Majority of infertity cases are diagnosable, but about 10% of cases remain unexplainable.

Chlamydia trachomatis is a major causative agent of sexually transmitted disease (STD), with an estimated 90 million cases worldwide (WHO, 2001) *Chlamydia trachomatis* is an obligate intracellular bacterium with 15 immunotypes, which are as follows: Types A-C cause trachoma (chronic conjunctivitis endemic in Africa and Asia); D-K cause genital tract infections; and serotypes L1-L3, *Lymphogranuloma venereum* (associated with genital ulcer disease in tropical countries).

Genital infection caused by *Chlamydia* trachomatis is generally asymptomatic. Approximately 50% of infected males and 80% of infected females show no symptoms, but infection may cause a mucopurulent cervicitis in females and urethritis in males (Ingalis et al., 1995). Commonly unrecognized and often poorly or inadequately treated, *Chlamydia* infections can ascend the reproductive tract resulting in pelvic inflammatory disease (PID) and, consequently, lead to chronic pelvic pain, ectopic pregnancy, and infertility. All Chlamydia have a group reactive antigen detectable in the supernatant of lysates (or heat suspension). The antigen is heat stable lipopolysaccharides with 2-keto-3 deoxycholic acid as an immunodominant component. This antigen can be detected by complement fixation test (Debatista et al.,

2002). This justifies the use of species specific monoclonal antibody to screen for different species so as not to have false positive results as complement fixation test will only detect the group reactive antigen. It also justifies a confirmation by culture methods.

Evidence of previous infection with *chlamydia trachomatis* has been associated with a reduction in semen quality (Agbolahor et al. 2007), this infection in male has a wide clinical spectrum which may lead to infertility such as non specific uretritis, epididymitis and prostititis leading to stenosis of the duct system (Eggart-Krusse et al., 1995). Recent evidence suggests that the bacteria by acting directly on sperm cell may cause infertility (Hosseinzadeh et al., 2001). Although some scholars believe that infection with clamydia has more adverse effect in females than in males (Ibadin et al., 2009).

Transmission of Chlamydia *trachomatis* is principally sexual. This makes it one of the high risk infections for easy transmission of HIV and also a major STI (Sexually transmissible infection). The invasive intracellular pathogenesis of *C. trachomatis* can cause significant damage to the genital epithelial layer, which may facilitate HIV infection. Conversely, the immunological changes due to HIV infection may favor *Chlamydia trachomatis* infection (Debattista et al., 2002).

Reports about the prevalence of *Chlamydia trachomatis* in various risk groups have recorded significant figures, for instance a study in Enugu recorded 26.4% prevalence rate in adult women (Ikeme et al., 2011) and about 45% prevalence in patients attending various gynaecological clinics in south east Nigeria (Okoror et al., 2007). These figures highlight the growing incidence of this infection in various parts of our country, hence our motivation to carry out this study. There is also lack of sufficient information as regards Chlamydia infection rates in Ogun state, Nigeria.

Therefore, this study was carried out to determine the presence of this infection in the one of the most exposed risk groups that is adult sexually active couples, to detect the presence of this infection and to show any link between infertility problems and presence of Chlamydia infections in Abeokuta, Ogun State, Nigeria.

2. MATERIALS AND METHODS

2.1. Study Design: The study was carried out on couples visiting infertility clinic in Abeokuta Metropolis, with the objective of screening for the presence of *Chlamydia trachomatis* in semen and Genital swabs of women who met the recruitment criteria were screened. Pregnant women were excluded from the study, while inclusion criteria were women presenting with Pelvic inflammatory disease and various forms of infertility.

2.2. Sample Collection and Processing: Male subjects were given written and oral instructions on semen sample collection, samples were obtained by masturbation and ejaculated into a clean wide mouthed plastic container, after the subjects must have passed urine and washed their hands and penis before producing the sample. High vaginal swabs were collected by family planning nurses, all samples were transported immediately to the Microbiology laboratory. Semen analysis was done following WHO protocol (WHO 1992). Microscopy was done on all HVS samples.

2.3. Chlamydia Antigen screening: The Chlamydia check-7 Test Unit (U.S.A) was used. The test principle is based on enzyme immunoassay based chromatographic rapid antigen detection. An initial extraction step was done according to the manufacturers protocol briefly 0.9ml of extraction solution was put in a tube; the swab was immersed into it and swirled vigorously for 10secs afterwards the extraction tube was kept for 10mins before testing was done.

3. RESULTS ANALYSIS

A total of 315 subjects were screened for the presence of *Chlamydia trachomatis* antigen, comprising 108 (24.3%) males and 207(75.7%) females, with an overall positivity rate of 31(9.8%). Table 1 shows the frequency of *Chlamydia trachomatis* antigen in relation to ages and sexes of subjects. It showed that subjects ages 31-40 years of age had the highest positivity rate (13.0%) for *Chlamydia trachomatis* antigen, this was closely followed by age group 41-50 and 20-30 years of age having 7.7% and 6.5% positivity respectively (Table 1). It also showed that *Chlamydia trachomatis* antigen was more prevalent among females (11.6%) compared to their male counterparts having 6.5% positivity (Table 1).

Table 1: Frequency of *Chlamydia trachomatis* in relation to ages and sexes of subjects

Age groups (years)	Total No. tested (%)	No. positive (%)	Males		Females	
			No. tested (%)	No. positive (%)	No. tested (%)	No. positive (%)
20-30	108(34.3)	7(6.5)	25(23.1)	0(0.0)	79(73.1)	7(8.8)
31-40	154(48.9)	20(13.0)	50(46.3)	6(12.0)	104(67.5)	14(13.5)
41-50	52(16.5)	4(7.7)	28(25.9)	1(3.6)	24(46.2)	3(12.5)
51 & above	5(1.6)	0(0.0)	5(4.6)	0(0.0)	0(0.0)	0(0.0)
Total	**315(100.0)**	**31(9.8)**	**108(24.3)**	**7(6.5)**	**207(75.7)**	**24(11.6)**

Table 2 shows the frequency of *Chlamydia trachomatis* antigen in relation to size of pus cell seen in the specimens of subjects. Percentage distribution of Chlamydia antigen positivity by pus cell size showed a higher distribution for large pus cells (40.4%) in comparison to small sized leucocytes (3.7%) among Chlamydia positive specimens (Table 2).

Table 2: Frequency of *Chlamydia trachomatis* Antigen in relation to sizes of pus cell

Specimens	Sizes of Pus Cell	
	Normal N (%)	Large N (%)
Chlamydia Negative	260(96.3)	31(59.6)
Chlamydia Positive	10(3.7)	21(40.4)

Table 3 shows the frequency of Chlamydia antigen in relation to type of infertility. Chlamydia positivity rate in relation to type of infertility showed that secondary infertility was (8.3%), primary infertility (9.5%) and the highest positivity rate was observed among those with subfertility (26.3%) as shown in Table 3.

Table 3: Frequency of Chlamydia Antigen in relation to type of Infertility

Age groups (years)		1^{O} Infertility		2^{O} Infertility		Subfertility	
	No. (%)	No. Tested (%)	No. Positive (%)	No. Tested (%)	No. Positive (%)	No. Tested (%)	No. Positive (%)
20-30	108(34.3)	24(22.2)	3(12.5)	70(64.8)	7(2.8)	14(13.0)	2(20.0)
31-40	154(48.9)	31(20.1)	2(6.5)	116(75.3)	10(8.1)	7(4.5)	2(28.5)
41-50	52(16.5)	8(15.4)	1(12.5)	42(80.8)	3(7.1)	2(3.8)	1(50.0)
51 & above	5(1.6)	0(0.0)	0(0.0)	5(100.0)	0(0.0)	0(0.0)	0(0.0)
Total	**315(100.0)**	63(20.0)	6(9.5)	240(76.2)	20(8.3)	19(6.0)	5(26.3)

Table 4 shows the frequency of Chlamydia antigen positivity in relation to sperm cell concentration. Positivity by sperm concentration showed highest positivity rate in Azoospermic male subjects 25(23.0%) and Oligozoospermic male subjects had the lowest Chlamydia positivity rate with 3(2.7%) as shown in Table 4.

Table 4: Frequency of Chlamydia Antigen Positivity in relation to Sperm Cell concentration

Specimens	Sperm concentration		
	Azoospermia N (%)	Normalozoo spermia N (%)	Oligozoospermia N (%)
Chlamydia Positive	25 (23.0)	3 (2.7)	3 (2.7)
Chlamydia Negative	37 (34.3)	40 (37.0)	31 (28.7)

4. DISCUSSION

Infertility is a global health issue, affecting about 8-10% of couples worldwide (Inhorn, 2003). In some societies in sub Saharan Africa, one third of all couples are unable to conceive throughout their reproductive lives (Crate et al., 1985). *Chlamydia trachomatis* infection has been reported to cause various forms of Gyneacological problems which can lead to infertility or contribute to it (Agbonlahor et al., 2007). The overall prevalence rate of *Chlamydia trachomatis* antigen positivity was (9.8%), this is lower to a recent study done at Enugu, Nigeria which reported an overall rate of 29.4% (Ikeme et al., 2011). Hence our result shows a high incidence of asymptomatic *C. trachomatis*, which agrees with findings of other reports (Strum-Ramirez et al., 2002; Nwaguma et al., 2009).

Our study reports that the highest incidence rate according to age was in age groups 31-40 years of age with females having 13.5% and males (12.0%). This is in agreement with earlier findings of Agbolahor et al. (2007) who reported that females were at more risk of

contacting Chlamydia infection, although the higher rate recorded for female subjects may not be unconnected with the fact that more females visited our clinic than males. The lopsided gender distribution was probably due to the fact that the female subjects were in house infertility patients, which made sample collection easier, and the fact that male partners refused presenting their samples (semen) for analysis. The high incidence seen in females of age group 31-40 can be attributed to the fact that previous sexual exposure to the Chlamydia infection at adolescent age and anatomy of the female genital tract puts them at increased risk.

Incidence of *Chlamydia trachomatis* in relation to infertility type reveals that subfertile subjects recorded the highest positivity rate to *Chlamydia trachomatis*, followed by subjects diagnosed with primary infertility and lastly by secondarily infertile subjects. The higher positivity rate observed in subjects with primary infertility as compared with secondary infertility, has been reported in a previous study (Larsen et al., 2004), who also reported that primary infertility is a serious issue in the developing world, as compared with the developed world where secondary infertility supersedes as the major fertility problem. Our study recorded a higher positivity rate in Large pus cell size with a rate of 40%, as compared with 3.7% for normal sized pus cells recorded for all analyzed samples, this is in agreement with an earlier report of Mania et al. (2001), which also proposed leucocytes size as a potential diagnostic indicator for infection with Chlamydia, although proper laboratory diagnosis by isolation of the bacteria is always the gold standard for confirming diagnosis. The method used in this study has been documented as an inexpensive and effective screening method in developing countries (Nair et al., 1999). The more accurate techniques such as nucleic acid amplification for detection of infected genital secretions were not used in this study.

Positivity by sperm concentration revealed that Oligozoospermic subjects had the highest rate followed by Normal and lastly Azoospermic subjects. This result is similar to a study done at Benin City, which reported a seropositive rate of 24.0% for male subjects with sperm abnormalties (Ibadin et al., 2009). This finding buttresses the pathological significance of *Chlamydia trachomatis*. Chlamydia infection in males has been reported to induce leucocytospermia, leading to leucocytes-derive reactive oxygen species, which causes peroxidation and damages sperm cells leading to reduced sperm count and impaired sperm motility (Hosseinzadeh et al., 2004).

5. CONCLUSION

In conclusion, there was no strong independent evidence showing any association between *Chlamydia trachomatis* antigen positivity and infertility in couples at Abeokuta, Ogun State, Nigeria, although more case controlled studies are needed to further investigate any relationships between infertility and recent Chlamydia infection in our environment. However, there is strong evidence that adult couples in the prime of their reproductive age pose the risk of exposure to sexually transmitted infections (STI) such as *Chlamydia trachomatis*, however, efforts to address possible risk factors which are proxies to acquisition of STI and better health seeking behavior by couples intending to have children will likely reduce the burden of infertility in Nigerian couples.

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REFERENCES

1. Agboniahor D. E., Okoro L. E., Esumeh F.L., Umolu P. I. (2007). Prevalence of Chlamydia in patients attending gyneacoloy Clinics in South E astern Nigeria: *Afr Health Sci*. 7(1): 18-24.
2. American Society of Reproductive Medicine (ARSM). http://arsm.org/patient/fags.html 10/23/2008.
3. Crates N., Farely T.M.M., Rowe P.J. (1995). Worldwide Pattern of Infertility: Is Africa different? *The Lancet* 2(8455):596-598.
4. Debattista J., Clementson C., Mason D., Dwyer J., Argen S., Wood ward C. (2002). Screening for Neisseria gonorrhea and Clamydia trachomatis at entertainment venues among men who have sex with men.SEX Trans Dis. 29: 216-21.
5. Eggert N. G., Rohr S., Bockem-Hellwing K., Huber M., Christmann-Edoga A., and R Runnebaum (1995). Immunological aspect of Sub-Fertility. Int. J. Androl, 18(2): 43-52.
6. Hosseinzadeh S., Eley A., Pacey A.A. (2004). Semen quality of men with asymptomatic Chlamydia Infection. *J. Androl*. 25(1): 104-109.
7. Ibadin K.O., Enabulele O.I., Eghafona N.O., Osemwenkha A. P. (2009). Seroprevalence of Chlamydia trachomatis infections in infertile men and its associations with semen quality in

U.B.T.H. Benin City, Nigeria. *Benin. J. Postgrad. Med.* 11(1):11-14.
8. Ikeme AC, Ezegwui HU, Ikeako LC, Agbata I, Agbata E. Seroprevalence of *Chlamydia trachomatis* in Enugu, Nigeria. *Niger J Clin Pract* 2011;14:176-80
9. Ingalis R.R., Rice P.A., Qureshi N., Takayama K., Lin J.S., Golenback D.T. (1995). Clamydia trachomatis is endotoxin mediated. *Infect Immun*. 63: 3125-30.
10. Inhorn M. C., (2003). Global Infertility and Globalization of new Reproduction technologies: Illustration from Egypt. *Social Science and Medicine*. 56: 1837-1851.
11. Larsen U. (2003). Infertility in central Africa. Trop Med Int Health. 8:357-67.
12. Mania J., Pramanik Y., Gokral D. K., Meherji (2001). Ghalmydia Trachomatis Infection among asymptomatic males in an Infertility Clinic. Idian J Derm Venerol Lerol. 67(5): 242-245.
13. Nair D., Bhalla P., Mathur M.D. (1999). Comprisim of Enzyme Linked Immunosorbent Assay and direct Flourescent antibody testfor the detection of Clamydia trachomatis in non Gonococcal Urethritis. Indian J. Med. Microbiol. 174: 184-6.
14. Nwanguma B. C., Kalu I., Ezeanyia L. U. (2009). Seroprevalence of anti Clamydia trachomatis IgA antibody in a Nigerian population. Diagnostic significance and implications for the transmission of HIV. Int J. Infec. Dis. 7:2.
15. Okoror L. E., Agbonlahor D. E., Esumeh S. I., and P.I. Umolu (2007). Prevalence of Clamydia in patients attending Gyneacological Clinics in South Eastern Nigeria. Afr Health Sc. 7(1): 18-24.
16. Sturm-Ramirez K., Hunter B., Diop K., Ibruhima N. (2002). Molecular epidemiology of genital Clamydia trachomatis Infection in high risk women in Senegal, Wet Africa. J. Clin. Microbiol. 32:138-45.
17. World Health Organization (1991). "Infertility: A tabulation of available data on prevalence of primary and secondary Infertility" WHO/MCH/91-9.
18. World Health Organization (1992). Mannual for the examination of Human Semen and Spem-Cervical mucus Interaction. University Press. Cambridge, U.K.

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