Original Article

Chlamydia Trachomatis Infection Among Infertile Women at the University College Hospital, Ibadan

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ABSTRACT

Background: Chlamydia trachomatis infection is reported as the commonest cause of tubal and pelvic infection and by proxy, the commonest cause of tubal damage and female infertility. Serotypes D-K are known to cause sexually transmitted genital tract and neonatal infection. Testing for the presence of Chlamydia trachomatis has been revolutionized by the development of monoclonal antibodies. This study sought to determine the prevalence of Chlamydia trachomatis infection among infertile women at the University College Hospital Ibadan and to determine the diagnostic value of Chlamydia antibody testing.

Methodology: A cross-sectional study conducted among two hundred and seventy-three women with infertility attending the gynaecology clinic of the University College Hospital Ibadan. Hysterosalpingography (HSG) was performed on all patients as part of their routine evaluation for infertility. Venous blood was obtained to detect Chlamydia Ig G antibodies using the diagnostic bio-probe (DIA.PROBE) enzyme-linked immunoassay (ELIZA) for the quantitative determination of IgG antibodies specific to chlamydia trachomatis.

Results: Positive test results for Chlamydia trachomatis were recorded amongst 136 women, giving a prevalence of 49.8%. 46(33.8%) women with normal tubal appearance and 90(66.2%) with tubal disease tested positive for Chlamydia antibodies. Bilateral tubal blockage was seen in 77(28.2%), while left and right tubal blockages were seen in 35(12.8%) and 21(7.7%) respectively. The accuracy of Chlamydia antibody testing in predicting tubal patency revealed a sensitivity of 67.7% and positive predictive value of 66.1% using HSG as the gold standard. Chlamydia trachomatis infection was found to be associated with age at sexual debut, history of ectopic pregnancy and previous history of abortion ($P<0.05$).

Conclusion: This study revealed a high prevalence of Chlamydia trachomatis amongst infertile women especially those with tubal disease. Risk factors associated with the infection such as early age of sexual debut, ectopic pregnancy and abortion further support the role of infectious morbidity especially with Chlamydia trachomatis.

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INTRODUCTION

Infertility refers to the inability of a couple to achieve conception after one year of regular, unprotected sexual intercourse. For women 35years and older a period of 6 months is sufficient to ascribe the diagnosis.[1] In many gynaecological clinics in developing countries, infertility constitutes a major presenting compliant and accounts for greater than 20% of consultations.[2] A high premium is placed on a woman's ability to reproduce; therefore, infertility can be a source of

socio-cultural, psychological, and financial disharmony in affected couples.[3,4] The aetiology of infertility is multi-factorial and has traditionally been classified into male and female factors with equal contributions from both. Causes of female factor infertility are further sub-divided into, tubal, ovarian, uterine, cervical, and peritoneal factors. Genital tract infections leading to tubal factor infertility is the leading causative factor in African and other developing countries. Most infections are acquired from sexual contacts, post-abortal sepsis and postpartum infections.[4,5,6] Tubal factor infertility account for 40% of the causal burden of infertility in Africa.[4,6,7] In Nigeria, the prevalence ranges from 5.5% - 48% in clinical settings.[8,9,10,11] The commonest infective agents implicated are Chlamydia trachomatis and Neisseria gonorrhoea. Additionally, genital tuberculosis has begun to emerge as an important aetiology, however, bacterial infections seem to be more prevalent.[4,5]

Chlamydia trachomatis is an intracellular bacterium that requires living cells to multiply. Out of the 18 distinct serotypes, serotypes D-K cause sexually transmitted genital tract and neonatal infections.[12] Chlamydia infection is reported as the commonest cause of tubal and pelvic infection and by proxy the commonest cause of tubal damage and infertility.[13] Presently, it has emerged as a global public health issue in the reproductive health of both sexes and there is an estimated annual incidence of around 92 million cases in the world.[14] In the United States alone the cost of treatment is estimated to be about 2 billion dollars per annum.[14] Most infections are asymptomatic and about 80% of cases and are cleared by the immune system. In those with persistent and chronic infection, the course is usually that of chronic inflammatory reaction. There is associated tubal and peritoneal damage, loss of tubal patency and motility. Infection of the female reproductive tract by Chlamydia trachomatis is one of the leading global causes of tubal and female factor infertility.[15] Testing for Chlamydia in women with tubal infertility has been well documented by several studies.[14,15] Cell culture was long the gold standard for diagnosis of Chlamydia trachomatis infection because of its absolute specificity. However, due to the labour-intensive methodology, turnaround time, cost, and requirements for infrastructure and technical expertise, cell culture facilities are limited to specialized research laboratories only. Other non-culture tests, such as enzyme immunoassays (EIAs) and serological tests, lack the high sensitivity and specificity needed for accurate diagnosis of ongoing infection. Molecular methods such as Polymerase Chain Reaction (PCR) are more specific and sensitive when compared to other methods.[16] Testing for the presence of Chlamydia trachomatis in clinical specimens has been revolutionized by the development of monoclonal antibodies. This test method is rapid and does not depend on a cold chain transportation system.[17]

The association between Chlamydia trachomatis infection and antibody testing in tubal factor infertility has been demonstrated by several studies.[15,17] A study carried out in Benin City Nigeria, revealed a high prevalence of Chlamydia trachomatis antibody titre in tubal factor infertility.[18] Multiple risk factors have been associated with acquiring genital tract Chlamydia trachomatis infection. Risky behaviours include excessive alcohol intake, use of illicit drugs, cigarette smoking and use of oral contraceptive pills.[19] In a recent study from Nigeria, factors such as previous sexually transmitted infection especially Neisseria gonorrhoea, multiple sexual partners, and lifetime partners greater than one all had strong associations with chlamydia infections.[20]

Chlamydia antibody testing therefore plays a significant role in screening women with tubal infertility and may serve as a baseline investigative tool to assess the risk of tubal pathology in infertile patients whilst offering treatment before other elaborate investigations for infertility.[21] This study aimed at determining the accuracy of Chlamydia trachomatis antibody testing (CAT) as a minimally invasive and cost-effective screening tool in predicting tubal factor infertility among infertile women. The sensitivity, specificity, positive,

negative predictive values, and accuracy of Chlamydia antibody testing in detecting tubal pathology were determined using HSG as the gold standard.

MATERIALS AND METHOD

A cross-sectional study conducted among women with infertility attending the Out-patient Gynaecological clinic of the University College Hospital, Ibadan between 1st September 2018 and 28th of February 2019. A total of 273 women were recruited consecutively during their clinic visit. The study was approved by the University of Ibadan and University College Hospital's Institutional Review Board. Informed consent was obtained after details of the study was explained to the participant.

A purpose-designed proforma was used to obtain their sociodemographic data, gynaecologic history, and other relevant information. Thereafter, patients were taken to a private room in the clinic for sample collection. Each patient was identified, and a sample was taken from the cubital fossa and placed in an EDTA (ethylenediaminetetraacetic acid) bottle labelled with the patient's identification details. The collected samples were stored at +2- 8^0 Celsius before analysis which allowed for sample pooling and cost-effective analysis. Chlamydia IgG antibodies were detected from the patient's serum using the diagnostic bio-probe (DIA.PROBE) enzyme linked immunoassay (ELIZA) for the quantitative determination of IgG antibodies specific to Chlamydia trachomatis. The assay was carried out according to the manufacturer's instruction. Following sample collection each patient was booked for a hysterosalpingogram as part of tubal patency evaluation.

Data obtained was entered into the Statistical Package for Social Sciences version 17 software (IBM SPSS, Chicago, IL. USA). Significance was set at ($P< 0.05$). Bivariate analysis was performed between the socio-demographic data, gynaecologic data, and the screening outcome of Chlamydia infection. Logistic regression analysis was used to identify the significant predictors after adjusting for cofounders. The Sensitivity, Specificity, Positive Predictive and Negative Predictive Values were calculated by comparing with HSG findings.

RESULTS

Background characteristics

Table 1: shows the participants background characteristics. The mean age of the respondents was 34.9 years (SD=6.2) and ranged between 18-62 years. Almost all respondents were married (94.9%) and belonged to the Yoruba ethnic group (90.1%). More than half of the respondents were Christians (60.4%) and had tertiary level of education (62.3%). This was followed by those with secondary (27.1%), primary (8.4%) and no education (2.2%).

Table 1. Participant's background characteristics

VARIABLE	FREQUENCY	PERCENTAGE
Age		
<30	50	18.3
30-34	83	30.4
35-39	84	30.8
40+	56	20.5
Marital status		
Single	10	3.7
Married	259	94.9
Separated	2	0.7
Cohabiting	2	0.7
Level of education		
None	6	2.2
Primary	23	8.4
Secondary	74	27.1
Tertiary	170	62.3
Tribe		
Yoruba	246	90.1
Ibo	11	4.0
Hausa	5	1.9
Others	11	4.0
Religion		
Christianity	165	60.4
Islam	108	39.6

Gynaecologic characteristics of participants

Table 2: The mean age at sexual debut was 22.5years (SD=5.3) and ranged between 8-46 years. Majority of the respondent had infertility greater than 49 months (42.5%). The least duration was between 37-48 months (9.9%). Almost half of the respondents had their sexual debut between 20-24 years (42.1%). About a third of the respondents were

diagnosed with secondary infertility (68.1%) and did not have any children (58.2%). About half had a previous history of pelvic infection (54.2%) while one fifth had a history of previous pelvic surgery (23.8%). Among respondents who had a previous surgery, Caesarean section (43.2%) and myomectomy (29.2%) constituted the bulk of surgeries. Contraceptive use was reported by (10.3%) of the respondents. Injectable (35.8%), condoms (28.6%) and oral contraceptives (14.3%) were the most common types of contraceptive methods in use. About half of the respondents (54.6%) reported a previous history of abortion with induced abortions constituting almost half (45.6%) of the types of abortions reported. Also, Spontaneous termination of pregnancy (STOP) was reported by (38.9%) while (15.4%) reported having had both spontaneous and induced abortions. Almost all respondents did not have a new sex partner (97.1%) or history of multiple sexual partners (90.1%). About (97.1%) had only one sexual partner while (97.4%) did not use a condom consistently during previous sexual encounters. Majority of the respondents tested negative for the HIV infection (75.5%) and did not have a history of vaginal discharge or abdominal pain in the preceding six months (75.5% and 70.0%) respectively.

Table 2: Gynaecological Characteristics of Participants

VARIABLE	FREQUENCY	PERCENTAGE
Age at sexual debut (years)		
<20	71	26.0
20-24	115	42.1
25-29	66	24.2
30+	21	7.7
Infertility type		
Primary	87	31.9
Secondary	186	68.1
Duration of infertility (months)		
≤ 12	37	13.6
13-24	57	20.9
25-36	36	13.1
37-48	27	9.9
49+	116	42.5
Parity		
0	159	58.2
1	63	23.1
2	37	13.6
3	11	4.0
4	3	1.1
Previous history of ectopic pregnancy		
Yes	14	5.1
No	259	94.9
Previous pelvic infection		
Yes	148	54.2
No	125	45.8
Previous pelvic surgery		
Yes	65	23.8
No	208	76.2
Surgery type (n=65)		
Appendectomy	1	1.5
Caesarean section	28	43.2
Ectopic	4	6.2
CS/Ectopic	6	9.2
Cerclage	1	1.5
Hysteroscopy	1	1.5
Myomectomy	19	29.3
Myomectomy/CS	1	1.5
Myomectomy/ectopic	1	1.5
Tubal recanalization	3	4.6
Contraceptive use		
Yes	28	10.3
No	245	89.7
Contraceptive type (n=28)		
Oral	4	14.3
Emergency	2	7.1
Condom	8	28.6
Implant	2	7.1
Injectables	10	35.8
IUD	2	7.1
Previous history of abortion		
Yes	149	54.6
No	124	45.4
Type of abortion (n=149)		
Spontaneous	58	38.9
Induced	68	45.6
Both	23	15.4

Table 3: 49.8% of the patients tested positive to Chlamydia trachomatis. About half of the women had a normal HSG report (51.3%). This was followed by those diagnosed with bilateral tubal blockage (28.2%), left tubal blockage (12.8%), and right tubal blockage (7.7%).

Table 4: Chlamydia trachomatis infection was found to be associated with age at sexual debut, history of ectopic pregnancy, and previous history of abortion ($P<0.05$).

Table 5: Following adjustment for confounders, significant predictors of patient's Chlamydia status were age at sexual debut and HSG status. Patients whose age at sexual debut was less than 20 years were twice more likely to test positive for chlamydia compared to those whose age at sexual debut was 25 years and above (OR= 2.547; 95%CI=1.216-5.334).

Table 3: Hysterosalpingogram and Chlamydia Test Result

VARIABLE	FREQUENCY	PERCENTAGE
HSG report		
Normal	140	51.3
Bilateral tubal blockage	77	28.2
Left tubal blockage	35	12.8
Right tubal blockage	21	7.7
Chlamydia antibody test	136	49.8
Positive	137	50.2
Negative		

Table 4: Association Between Chlamydia Status And Gynaecologic/ Obstetric Characteristics Of Participants

Variable	Chlamydia		Total (%)	Chi square	P value
	Positive (%)	Negative (%)			
Age at first sexual intercourse (years)					
<20	43 (60.6)	28 (39.4)	71 (100)	8.452	**0.015***
20-24	60 (52.2)	55 (47.8)	115 (100)		
25+	33 (37.9)	54 (56.1)	87 (100)		
Fertility type					
Primary	41 (47.1)	46 (52.9)	87 (100)	0.370	0.543
Secondary	95 (51.1)	9 (48.9)	186 (100)		
Duration of infertility (months)					
≤ 12	18 (48.6)	19 (51.4)	37 (100)	1.808	0.771
13-24	26 (45.6)	31 (54.4)	57 (100)		
25-36	16 (44.4)	20 (55.6)	36 (100)		
37-48	12 (48.0)	13 (52.0)	25 (100)		
49+	63 (54.3)	53 (45.7)	116 (100)		
Parity					
0	77 (48.4)	82 (51.6)	159 (100)	1.258	0.739
1	30 (47.6)	33 (52.3)	63 (100)		
2	21 (56.8)	16 (43.2)	37 (100)		
3+	8 (57.1)	6 (42.9)	14 (100)		
Previous history of ectopic pregnancy					
Yes	12 (85.7)	2 (14.3)	14 (100)		
No	124 (47.9)	135 (52.1)	259 (100)	7.606	**0.006***
Previous pelvic infection					
Yes	75 (50.7)	73 (49.3)	148 (100)	0.095	0.757
No	61 (48.8)	64 (51.2)	125 (100)		
Previous pelvic surgery					
Yes	38 (58.5)	27 (41.5)	65 (100)	2.550	0.110
No	98 (47.1)	110 (52.9)	208 (100)		
Contraceptive use					
Yes	15 (53.6)	13 (46.4)	28 (100)	0.176	0.675
No	121 (49.4)	124 (50.6)	245 (100)		

Also, patients who were diagnosed with tubal blockage on HSG were about four times more likely to test positive for Chlamydia compared to those with normal HSG (OR = 4.650; 95%CI=2.705-7.995).

Table 6: The accuracy of using Chlamydia trachomatis antibody testing in predicting tubal patency showed a fairly high Sensitivity and Specificity (67.7%& 67.1% respectively).

Table 5: Logistic Regression of Chlamydia Antibody Test on Significant Variables

Variable	Unadjusted OR (95%CI)	Adjusted OR (95%CI)
Age at sexual debut (years)		
<20	2.513 (1.320-4.782	2.547 (1.216-5.334) *
20-24	1.785 (1.013-3.147)	1.641 (0.873-3.083)
25+ ***(ref)***		
Previous history of ectopic pregnancy	6.532 (1.433-29.768)	3.134 (0.625-15.714)
Yes		
No ***(ref)***		
Previous history of abortion		
Yes	1.901 (1.173-3.081)	1.676 (0.955-2.939)
No ***(ref)***		
HSG		
Tubal blockage	4.277 (2.578-7.097)	4.650 (2.705-7.995) *
Normal ***(ref)***		

Table 6: Accuracy of the Chlamydia Test Iin Predicting Tubal Patency

Variable	HSG		Total (%)	Chi square	P value
	Tubal blockage (%)	Normal (%)			
Chlamydia Positive	90 (66.2)	46 (33.8)	136 (100)	33.063	<0.001*
Negative	43 (31.4)	94 (68.6)	137(100)		
	133 (49.8)	140 (50.2)	273		
Sensitivity	67.7%				
Specificity	67.1%				
Positive predictive value	66.1%				
Negative predictive value	68.6%				

DISCUSSION

This study sought to determine the prevalence of Chlamydia trachomatis infection among infertile women and to determine its accuracy in predicting tubal patency at a Nigerian tertiary institution. Findings were indicative of a high prevalence among infertile women, significant association with age at sexual debut, history of ectopic pregnancy and previous history of abortion (P<0.05). HSG was the gold standard for tubal evaluation in this study and our findings suggested that women with tubal disease had a four-fold likelihood of Chlamydia infection.

The prevalence of Chlamydia trachomatis infection among infertile women, attending the gynaecological clinic of the University College Hospital, Ibadan was 49.8%. This finding was similar to that reported by Mawak *et al*[22] in a study conducted among women attending a gynaecological clinic in North-Central Nigeria. A lower prevalence was reported by Morhason-Bello *et al*[23] in a study conducted in Ibadan. However, this study was conducted in a secondary health facility and had a smaller study population. Similarly, Tukur *et al*[24] reported a lower prevalence in women with tubal factor infertility. Some of these differences can be explained by variations in methodology and the fact that this study was conducted among a cohort of high-risk groups who have proven tubal factor infertility.This study revealed that the overall seroprevalence of Chlamydia trachomatis was higher in women with tubal factor infertility than in women without tubal factor infertility as confirmed with HSG. This was similar to a study by Jeremiah *et al*[25] which revealed a higher prevalence of Chlamydia trachomatis antibodies in infertile women with tubal factor infertility compared with fertile controls. Makled *et al*, also demonstrated that positive chlamydia antibody was significantly associated with tubal blockage.[22] This further emphasized the role of pelvic infection as a cause of tubal damage and highlights a correlation between chlamydia antibodies and tubal factor infertility.

Analysis from this study suggests that there was no correlation between age, marital status, level of education, tribe and religion with positive chlamydial antibody test. This was in contrast to a study in North-Western Nigeria in which age and marital status where associated with Chlamydia infections.[26] Though age was not significantly associated with Chlamydial infection in this study, the highest prevalence was in the third decade of life. In a population survey in the United States of America, Chlamydia infection was commoner in the adolescent population.[27] Adolescents and young adults fall into an age groups of high sexual activity and this may be responsible for the high prevalence observed. A study in Ibadan, Nigeria[23] suggested that Muslims and women with lower educational and social status were more likely to be at risk of Chlamydial infection. It was suggested that, these women may have a risky sexual behaviour coupled with poor health awareness and health seeking behaviour.[23] However, this was not consistent with our finding.

Previous history of ectopic pregnancy and abortion revealed statistically significant association with Chlamydia trachomatis infection. The finding of previous ectopic pregnancy is supported by a study in Uganda, in which Chlamydial antibodies were associated with a fourfold increase in the risk of ectopic pregnancy.[28] In contrast, a study in Netherlands found no association between chlamydial antibodies and ectopic pregnancy.[29] Chlamydial infection has been associated with endosalphingeal damage from pelvic inflammatory disease predisposing to ectopic pregnancy.

Following logistic regression, age at sexual debut and presence of tubal blockage on HSG, were significantly associated with chlamydial infection. Age at sexual debut less than 20 years, increased the risk of infection by two-folds. Previous studies have also found that early age at first sexual intercourse was associated with having genital Chlamydia infection.[30,31] Adolescents have a higher risky sexual behaviour and are less likely to use barrier

contraceptives especially in low resource countries where access to reproductive health is limited. Also, the immature epithelium of the endocervical region are more susceptible to infections from Gonococcal and Chlamydial organisms.[28]

Women with tubal disease were more likely to test positive to Chlamydial antibodies. In our study, these women had a four-fold likelihood of Chlamydia infection than those with normal findings. This was consistent with other studies done in Nigeria.[32,33]The accuracy of Chlamydia serology using HSG as standard was found to be highly specific and to have good positive predictive values.[34] This suggests that a positive Chlamydial serological test is highly suggestive of tubal blockage and can easily be used to predict tubal patency or complement part of the evaluation protocol of infertile women. In this study, the serological screening method used was found to have a moderately high Sensitivity and high Positive Predictive Value. It therefore suggests that Chlamydia trachomatis screening can positively predict about half of women with tubal blockage. Though, Laparoscopy, the gold standard for diagnosing tubal blockage was not done, reports have shown that tubal spasm may occur in some women reported as having bilateral tubal blockage on HSG. This was an important limitation of our study.It is therefore prudent that physicians combine their clinical skills with other ancillary investigations to accurately rule out tubal factor infertility.

CONCLUSION

This study revealed a high prevalence of Chlamydial antibodies in infertile women. This was well correlated with the presence of tubal disease and early age at sexual debut. Risky sexual practices have been documented in adolescents especially in low-income countries ultimately contributing to the development of tubal disease and consequent infertility in the adult years. It is prudent to screen all infertile women who present for clinical evaluation to determine the presence of Chlamydia trachomatis and offer appropriate treatment. Against this backdrop, we are therefore recommending that routine Chlamydia serological testing should be part of the infertility workup. We suggest that large scale preventive strategies especially among high risk groups such as adolescents should be established to prevent and control the spread of chlamydia trachomatis infection.

DECLARATION

Sponsorship- Nil
Conflict of Interest- Nil

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