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Prevalence and host-related factors of *Chlamydia trachomatis* infection among fertile and infertile people in Ankpa, Kogi State, Nigeria

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ABSTRACT

Background: Chlamydia trachomatis infection is one of the most common sexually transmitted bacterial infections in the world and can cause infertility in both males and females. However, most Nigerian health care facilities do not screen for Chlamydia antigen in gynecological and general out-patient clinics. The aim of this study was to determine the prevalence and possible host-related factors of C. trachomatis infection among fertile and infertile people in Ankpa, Kogi State. Methods: Urine samples, endocervical swabs and urethral swabs were collected from 116 fertile and 92 infertile patients between January and April 2023, after administering a structured questionnaire and obtaining informed consent. Samples were analyzed using Diaspot Chlamydia Kit, a rapid immunoassay test for the detection of genital Chlamydia antigen in urinogenital samples and by the McCoy cell culture technique. Samples (N=208) were collected from males (n=81) and females (n=127) aged between 19 and 50 years. Results: Overall, nineteen samples (9.1%) tested positive for C. trachomatis antigen. Infertile patients (63.2%) had higher C. trachomatis prevalence than fertile patients (36.8%). The highest prevalence was observed among infertile groups aged below 19 (33.3%) followed by fertile groups aged 20-29 (15.9%) and 30-39 (7.9%). Infertile divorced patients (14.3%) had a higher rate of infection than the single fertile patients (8.8%) and married patients (5.2%). C. trachomatis infection rate was higher among fertile men (66.7%) than among fertile women (23.1%). The seropositivity rate did not differ between fertile men and infertile men (χ 2=0.00; P=1.00). However, among women, the seropositivity rate differed marginally between fertile women and infertile ones ($\chi 2 = -0.174$; P=0.05). Infection was significantly associated with low-income level (R=0.179; P=0.01), history of sexually transmitted diseases (R=-0.264; P=0.00) and lack of condom use (R=0.150; P=0.031). Conclusion: The current study observed a high rate of C. trachomatis infection in the study population when compared with other epidemiological studies. Consequently, we suggest awareness campaigns, routine screening, and monitoring of all fertile and infertile people for early detection and treatment of confirm cases to check C. trachomatis infection threat to reproductive life.

Introduction

Chlamydia trachomatis is a Gramnegative, obligate intracellular bacterium that requires living cells to replicate [1]. *C. trachomatis* is the most prevalent sexually transmitted bacterial infection worldwide, with an estimated 4 -5 million

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new cases each year [2]. It is the most implicated organism in infertility. Genital infection caused by C. 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 [3]. 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 [4]. Chlamydia infections can occur in other areas besides the genitals, including the anus, eyes, throat, and lymph nodes. Repeated Chlamydia infections of the eyes that go without treatment can result in trachoma, a common cause of blindness in the developing world [5]. Factors associated with an increased risk of infection are numerous sexual partners, sexual intercourse with non-condom use, absence of barrier contraceptives, use of oral contraceptives, partner with concurrent partners or sexually-transmitted disease or non-gonococcal urethritis, gonococcal mucopurulent cervicitis, sterile pyuria, other sexually transmitted diseases, young women (<25 years old), unmarried status, African/American/Hispanic ethnicity and poor socio-economic condition [6].

In many developed countries, screening programs for Chlamydia have been set up to reduce transmission and reproductive tract morbidity [7]. The United States Centers for Disease Control and Prevention recommend annual screening of all sexually active women aged 25 or less as is screening of older women with risk factors (for example, those who have a new sex partner or multiple sex partners) [8]. In the United States, chlamydial genital infection is the most frequently reported infectious disease, and the prevalence is highest in persons aged <25 years [8]. Approximately 4 million cases of chlamydial infections are reported per year with an overall prevalence of 5%. A prevalence as high as 14% was reported in African-America females aged 18-26 years [9]. In Ethiopia the prevalence rate for Chlamydia infection of the cervix was 5.9% [10].

In most parts of Nigeria, *C. trachomatis* infections are not routinely screened for, and hence relative information about the frequencies of the organism is sparse [1]. Elsewhere in the country, a prevalence of 13.3% among unsuspecting women attending antenatal clinic in Benin City has been

reported [11] while Nwanguma et al. [12] reported a prevalence of 33% in asymptomatic volunteers in another Nigerian population. However, there is the dearth of infection regarding the occurrence of *C. trachomatis* in the study area. Therefore, the current study aimed to identify *C. trachomatis* prevalence in Ankpa, Kogi State, Nigeria, and explore which are the most important factors affecting prevalence estimates.

Materials and Methods

Study Area

The study was carried out in Ankpa-Kogi State, Nigeria, which is situated on latitude 7°14' – 7°22' N and longitude 7°31' -7°37'E. It covers an area of 1,200 km2 with a population of 267,353. It lies on altitude 163m above the sea level and has a heterogeneous population that is mostly dominated by the Igala people [13].

Study Design / Population

A cross-sectional study was conducted from January to April, 2023. One hundred and sixteen fertile and ninety-two infertile patients aged between 19 and 50 years were consecutively recruited by random sampling from gynaecological and general outpatient departments of some selected hospitals. A total of 208 patients attending five major hospitals in Ankpa, namely Amazing Grace Hospital, Bethel Hospital, Living Hope Hospital, Zonal Hospital and Bayoh Medical Diagnostic Laboratory participated in the study. Fertile and infertile persons accessing any kind of health services within Ankpa town who falls within the age bracket and indicated willingness to participate in the research were included in the study while people on antibiotics and those who were not willing to provide consent were excluded from this study. Ethical clearance for the study was obtained from the Kogi State Hospital Management Board health research officer in accordance with the code of ethics for biomedical research involving human subjects.

Sample Collection

In the female, endocervical swabs were collected with the assistance of the medical personnel (the nurse) as described elsewhere.1 Cusco vaginal speculum was inserted into the vagina for the visualization of the cervix. A swab stick was inserted through the speculum into the endocervical canal and rotated. This permitted acquisition of columnar or cuboidal epithelial cells which are the main reservoir of *Chlamydia*

organism. It was withdrawn without contamination from exocervical or vaginal cells. The swabs were transported promptly to the laboratory and processed within 30 minutes of collection. In males, the tip of the penis was clean. A special thin swab was inserted into the urethra and was gently twisted side to side and then remained still for few seconds in order to allow the swab to absorb enough fluid before it was removed. From the males, urine samples were also collected. Structured questionnaire was used to obtained demographic details and other relevant information such as number of sex partner, use of contraceptives, past STDs, educational status, marital status, etc. were obtained from the participants.

Sample analysis

The Chlamydia Rapid Test Device (Swab/Urine) is a qualitative, lateral flow immunoassay for the detection of Chlamydia antigen from female cervical swab, male urethral swab, and male urine specimens. The test procedure was in accordance with the kit manufacturer's instruction. [14]. In this test, antibodies specific to Chlamydia antigen is coated on the test line region of the test. During testing, the extracted antigen solution reacts with an antibody to Chlamydia that is coated onto particles. The mixture migrates up to react with the antibody to Chlamydia on the membrane and generates a colored line in the test line region. The presence of this colored line in the test line region indicates a positive result, while its absence indicates a negative result. To serve as a procedural control, a colored line will always appear in the control line region, indicating that proper volume of specimen has been added and membrane wicking has occurred. Samples were collected and processed according to the detailed instruction in the manufacturers leaflet. All positive and negative samples were further inoculated into coverslip cultures of McCoy cells treated with cycloheximide. The inoculums were centrifuged for one hour. Inoculated cultures were incubated at 370C for three days, washed, fixed in methanol, stained with giemsa, and screened by dark-ground microscopy for possible intracytoplasmic inclusion bodies.

Statistical Analysis

Microsoft excel was used to summarize the raw data. Statistical software package for Social Sciences (SPSS) version 25.0 for Windows (Inc., Chicago, IL) was used to analyze the data. Differences in proportion were compared using Chi-Square and Pearson correlation coefficient. Level of

statistical significance was set at p < 0.05 (95% confidence level).

Results

Out of 208 study participants, 19 had C. trachomatis infection giving a prevalence of 9.1%. One hundred and eighty-nine (189) of the participants did not have C. trachomatis infection (Table 1). Fertile patients with C. trachomatis infection were 7(36.8%) while the infertile participants with C. trachomatis infection were 12(63.2%). The fertile and infertile with no infection were 109(57.7%) detectable 80(42.3%) respectively. Although the infection with C. trachomatis, was more in infertile participants than the fertile category the difference was not statistically significant (P=0.082) (Table Similarly, C. trachomatis infection rate was higher among fertile men (66.7%) than among fertile women (23.1%) as shown in Table 3. Chlamydia seropositivity rate did not differ between fertile men and infertile men ($\chi 2 = 0.00$; p = 1.00). However, among women, the seropositivity rate differed marginally between fertile women and infertile ones $(\chi 2 = -0.174)$; p = 0.05). Table 4 displays the incidence of Chlamydia infections categorized by age bracket of participants. A comparison between fertile and infertile individuals was made with regards to infection rate across different age groups. The highest rate of infection among fertile participants (7.9%) was observed in those aged 30 – 39 years, while the infertile group had their highest incidence (33.3%) among those below 19 years old. Pearson's correlation coefficient analysis revealed no significant relationship between Chlamydia infections and age groups for both fertile and infertile individuals (r2 = 0.39; P = 0.577). C. trachomatis infection rate was higher among single fertile persons (8.8%) compared to married and divorced fertile persons, as indicated in Table 5. However, there was no significant statistical association between C. trachomatis infection and marital status of fertile patients (R=-0.022; P=0.815). Although the highest infection rate of, C. trachomatis was observed among infertile divorced persons (14.3%), it was not significantly associated with their marital status (R=0.010; P= 0.927). Patients with previous exposure to sexually transmitted diseases (STDs) recorded higher prevalence of C. trachomatis infection (15.4%) among the fertile persons as shown in Table 6. Infection was significantly associated with history of STD among the fertile group (R=-0.211; P=0.023). Likewise, C. trachomatis infection rate in the infertile group was also highest among patients with history of STD (27.6%) (R=-0.293; P=0.005). Overall, seropositivity of the infection was significantly correlated with history of STD (P=0.000). The prevalence rate of C. trachomatis infection was highest (11.4%) among fertile patients who attained secondary school education (Table 7). Nevertheless, infection was not correlated with educational level of the fertile patients (R=0.002; P=0.979). Similarly, highest C. trachomatis infection rate (16.1%) was recorded among infertile patients with secondary school educational level (R=-0.010; P=0.926). Generally, seropositivity rate of Chlamydia infections showed no relationship with educational level (P= 0.994). Fertile people whose monthly income ranged from ₹19000 to N50000 had higher prevalence rate (13.3%) of C. trachomatis infection as shown in Table 8. No significant correlation was found between the infection and the monthly income level of the fertile patients (R=0.057; P=0.541). Conversely, infertile patients demonstrated increased prevalence rate among those whose monthly income falls below №18000 (26.5%) exhibiting statistically significant correlation (R=0.324; P=0.002). In general, C. trachomatis infection displayed increased prevalence rate among those earning less than ₹18000 per month (13.2%), demonstrating

statistical significance in relation to their income level (P=0.01). As shown in Table 9, use of condom was not significantly correlated with infection rate in fertile group (R=0.049; P=0.602), unlike among infertile persons where use of condom was significantly correlated with *Chlamydia* infection (R=0.230; P=0.027). In this case, infection was absent among condom users but was 6.9% among non-users. Generally, *C. trachomatis* infection was higher among the non-condom users (12.1%).

Table 1. Distribution of *Chlamydia trachomatis* infection among study participants.

Infected	Frequency	Percent
Yes	19	9.1
No	189	90.9
Total	208	100.0

Table 2. Distribution of *C. trachomatis* infection among infertile and fertile patients.

Infected	Fertility		Total
infected	Fertile	not fertile	
Yes	7	12	19
	36.8%	63.2%	100.0%
No	109	80	189
	57.7%	42.3%	100.0%
Total	116	92	208
	55.8%	44.2%	100.0%
			P=0.082

Table 3. *Chlamydia trachomatis* infection in relation to gender of fertile and infertile patients.

	Sex							
	Male			Female				
Fertility	Infected		Total	Infected		Total		
	yes	No		Yes	no			
Fertile	4 (66.7%) 50 (66.7%)		54 (66.7%)	3 (23.1%)	59 (51.8%)	62 (48.8%)		
not fertile	2 (33.3%)	25 (33.3%)	27 (33.3%)	10 (76.9%)	55 (48.2%)	65 (51.2%)		
Total	6 (100.0%)	75 (100.0%)	81 (100.0%)	13 (100.0%)	114 (100.0%)	127 (100.0%)		
	X2 = 0.00' P	= 1.000		X2 = 0.079; P	X2 = 0.079; $P = 0.051$			

Table 4. Distribution of *Chlamydia trachomatis* infection in relation to age.

			C. tracho	matis Infection			
Fertility			Yes		No		Total number
J			N	%	N	%	(N) examined
Fertile	Age (yr)	=<19	0	0.0%	12	100.0%	12
		20 - 29	4	7.0%	53	93.0%	57
		30 – 39	3	7.9%	35	92.1%	38
		40 and above	0	0.0%	9	100.0%	9
	Total		7	6.0%	109	94.0%	116
			İ			R=016	P=.863
not fertile	Age (yr)	=<19	1	33.3%	2	66.7%	3
		20 - 29	7	15.9%	37	84.1%	44
		30 – 39	3	8.6%	32	91.4%	35
		40 and above	1	10.0%	9	90.0%	10
		Total	12	13.0%	80	87.0%	92
			İ			R=.124	P = .241
Total	Age (yr)	=<19	1	6.7%	14	93.3%	15
		20 - 29	11	10.9%	90	89.1%	101
		30 – 39	6	8.2%	67	91.8%	73
		40 and above	1	5.3%	18	94.7%	19
		Total	19	9.1%	189	90.9%	208
						R=.039	P = .577

Note: n=number of positive or negative samples in each category; N=total number of positive and negative samples

Table 5. Distribution of Screened Patients for Chlamydia trachomatis infection based on marital status.

			Infected	Infected					
			Yes	Yes					
Fertility			N	%	N	%	Total number (N) examined		
Fertile	Marital	Married	4	5.2%	73	94.8%	77		
	status	Single	3	8.8%	31	91.2%	34		
		Divorced	0	0.0%	5	100.0%	5		
	Total		7	6.0%	109	94.0%	116		
						R=022	P=.815		
not fertile	Marital	Married	10	13.0%	67	87.0%	77		
	status	Single	0	0.0%	1	100.0%	1		
		Divorced	2	14.3%	12	85.7%	14		
	Total		12	13.0%	80	87.0%	92		
						R=010	P=.927		

Note: n=number infected or not infected in each category; N=total number (both infected and noninfected participants)

Table 6. Distribution of *Chlamydia trachomatis* infection in relation to STD history.

			Infe	cted		Total	number		
		Y	es	N	No		ned (%)		
Fertility	STD history	Number	%	number	%			R	Sign.
Fertile	No	3	3.3%	87	96.7%	90	100.0%	211	0.023
	Yes	4	15.4%	22	84.6%	26	100.0%		
	Total	7	6.0%	109	94.0%	116	100.0%		
not fertile	No	4	6.3%	59	93.7%	63	100.0%	293	0.005
	Yes	8	27.6%	21	72.4%	29	100.0%		
	Total	12	13.0%	80	87.0%	92	100.0%		
Total	No	7	4.6%	146	95.4%	153	100.0%	264	0.00
	Yes	12	21.8%	43	78.2%	55	100.0%		
	Total	19	9.1%	189	90.9%	208	100.0%		

Table 7. Educational status of screened patients for *Chlamydia trachomatis* infection.

			Infe	cted				
Fertility	Educational	Y	es	N	Ю	Total number	R	sign.
1 0101110)	level	number	%	examined		8		
Fertile	illiterate	0	0.0%	2	100.0%	2	.002	.979
	primary school	0	0.0%	16	100.0%	16		
	secondary school.	5	11.4%	39	88.6%	44		
	tertiary school.	2	3.7%	52	96.3%	54		
	Total	7	6.0%	109	94.0%	116		
not fertile	illiterate	0	0.0%	4	100.0%	4	010	.926
	primary school	2	14.3%	12	85.7%	14		

	secondary school.	5	16.1%	26	83.9%	31		
	tertiary school.	5	11.6%	38	88.4%	43		
	Total	12	13.0%	80	87.0%	92		
Total	not educated	0	0.0%	6	100.0%	6	.000	.994
	primary school	2	6.7%	28	93.3%	30		
	secondary school.	10	13.3%	65	86.7%	75		
	tertiary school.	7	7.2%	90	92.8%	97		
	Total	19	9.1%	189	90.9%	208		

Table 8. Distribution of *Chlamydia trachomatis* infection based on income level.

		Infected						
Fertility	Income (N)	Yes		No		Total number	R	sian
rennity	per month	Count	%	Count	%	examined		sign.
Fertile	<₩ 18k	3	5.3%	54	94.7%	57	.057	0.541
	№19k to №50k	4	13.3%	26	86.7%	30		
	> N 50k	0	0.0%	29	100.0%	29		
	Total	7	6.0%	109	94.0%	116		
not fertile	<₩ 18k	9	26.5%	25	73.5%	34	.324	0.002
	№19k to №50k	3	10.0%	27	90.0%	30		
	> N50k	0	0.0%	28	100.0%	28		
	Total	12	13.0%	80	87.0%	92		
Total	<№ 18k	12	13.2%	79	86.8%	91	.179	0.01
	№19k to №50k	7	11.7%	53	88.3%	60		
	> N50k	0	0.0%	57	100.0%	57		
	Total	19	9.1%	189	90.9%	208		

k: = 000

Table 9. Chlamydia trachomatis infection based on condom use

		Infected				Total	R	Sign.
Fertility	Condom use	Yes		No				
		Count	%	Count	%	Count		
Fertile	no	5	6.9%	67	93.1%	72	.049	0.602
	yes	2	4.5%	42	95.5%	44		
	Total	7	6.0%	109	94.0%	116		
not fertile	no	12	17.6%	56	82.4%	68	.230	0.027
	yes	0	0.0%	24	100.0%	24		
	Total	12	13.0%	80	87.0%	92		
Total	no	17	12.1%	123	87.9%	140	.150	0.031
	yes	2	2.9%	66	97.1%	68		
	Total	19	9.1%	189	90.9%	208		

Discussion

Chlamydia trachomatis infection is a globally–distributed and sexually transmitted infection that may lead to infertility [15]. The World Health Organization estimated that one hundred and six million adults worldwide were newly infected with C. trachomatis, of which about nine million were seen in Africa [16]. In spite of the aforementioned, there is no documented evidence of the disease in the study area. Therefore, the present study was undertaken to determine the prevalence of C. trachomatis infection and explore its host–related factors among fertile and infertile people in Ankpa Local Government Area, Kogi State.

The overall results of the study showed about nine percent prevalence which suggest that Chlamydia infection is endemic in the study area. The prevalence of the infection in this study is consistent with studies conducted in North-West [3], North-central [17]. Similar prevalence rate was also reported in England [18], Netherlands [19], Spain [20], Shenzhen-China [21] and Mexico [22]. However, the prevalence of the infection in this study was higher than some other studies conducted in Southern Nigeria [2], Europe [7] and United States [23]. The high prevalence found in this part of the country in this study may be because Chlamydia infections usually present with no clearcut symptoms and are, as a result, left untreated or mistaken for other infections such as gonorrhoea. Other factors that could be attributed 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 in many health care centers in this part of the world [17].

In this study, infertile patients were more infected compared to fertile patients, although there was no significant association with *Chlamydia* seropositivity. This finding is in line with reports in Nigeria [24] and agrees with reports from Tehran, Iran [25]. This is indicative of the role of *C. trachomatis* causing infertility, being the most common bacterial sexually transmitted disease in the world.

Previous exposure to sexually transmitted diseases (STDs) was significantly associated with *chlamydia* positivity in this study. This finding is in consonance to previous reports in Kano State, Nigeria [3], Rivers State, Nigeria [26], Plateau State, Nigeria [1], Japan [27] and India [28]. However, it

is contrary to findings by other researchers [29]. This observation can be attributed to similar routes of transmission of chlamydial infection like other sexually transmitted infections.

Although age did not reach statistically significant level, our findings indicate a higher prevalence of *C. trachomatis* infection among age group less than nineteen and thirty to thirty-nine than in other age groups. Age of peak infection in this study is in line with reports from previous studies in Brazil [30], Argentina [31] and Nigeria [32]. This finding could be due to the tendency for increased sexual activity within these age groups, being the most sexually active group.

Inconsistent use of condom was significantly associated with *Chlamydia* positivity in this study. This finding is similar to previous reports in Rivers State [32], Kaduna State [33] and ilishan-remo [34] but at variance with the report in Oyo State [29]. This observation suggests that inconsistent use of condom may be the reason for the infection in the study population since *C. trachomatis* infection is transmitted through unprotected sexual intercourse.

Marital status was not significantly associated with *Chlamydia* positivity. However high prevalence was observed in fertile singles and divorced infertile than married fertile/infertile people. This finding is at variance with the report of Lagos State [35], Plateau State [1, 36] but agrees with report from Rivers State [32]. Possible explanation may be due to the fact that this set of people (singles and divorcees) may have multiple sexual partners since they are not married to particular sexual partners.

Socioeconomic levels were evaluated in our study using a wealth index. A lower income level was correlated with increased Chlamydia infection, and this is indicative of poverty. This observation is consistent with previous report in Jos [36] and Kaduna [33], Nigeria. This could be attributed to inability to access care or screening because of financial constraint, and young people attending clinics might not have previously consulted a doctor for Chlamydia screening and might be seeking treatment for the first time at those clinics for health problems. This study found educational status not statistically significant. This is in line with study reported in Northwest Nigeria [37] but in contrast to previous study reported in Kaduna [17]. This may be due to the fact that even among the educated citizens of this part of the country, there is little or no awareness of the presence of this infection, therefore measures to avoid the infection are not considered.

Conclusion

Genital *C. trachomatis* infection is highly prevalent in this part of Nigeria and should be considered a silent epidemic that needs urgent attention. To save families from the trauma of infertility occasioned by *Chlamydia*, efforts should be made to routinely screen people at risk and positive cases treated accordingly. Enlightenment campaign is also advocated.

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The study was not in receipt of any funding.

Ethical consideration:

Ethical approval for the study was obtained from the State Ministry of Health on issues related to human health in accordance with the code of conduct for biomedical research involving human subject

Competing Interests

Authors declare that there was no conflict of interest.

Financial disclosure

None declared.

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