ORIGINAL ARTICLE

CHLAMYDIA TRACHOMATIS INFECTION IN PELVIC INFLAMMATORY DISEASE PATIENTS – A SNAP SHOT

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ABSTRACT

Background: *Chlamydia trachomatis* is a sexually transmitted infection which due to its asymptomatic nature remains undiagnosed and presents as pelvic inflammatory disease in females. The objective of the study is to find prevalence and risk factors associated with positive testing of *Chlamydia trachomatis* infection in women suffering from pelvic inflammatory disease visiting a tertiary care hospital in Pakistan.

Material and Methods: This cross-sectional study was carried out at Department of Microbiology, Army Medical College/ National University of Medical Sciences from March 2019 to September 2019. This cross-sectional study was conducted among women presenting with pelvic inflammatory disease over a period of seven months. Socio-demographic and behavioral data were collected and real-time PCR diagnostic method was used to detect the presence of *Chlamydia trachomatis* infection in urine samples. Samples were collected by non-probability convenient sampling technique; Samples were analyzed for risk factors identification and association with *Chlamydia trachomatis* infection was determined.

Results: The prevalence of *Chlamydia trachomatis* was 12 (20%) out of total 60 participants. Maximum positive cases were seen in 20-24 years of age group. Risk factors i.e., age, number of sexual partners, gross monthly income, level of education, place of residence and past sexual history were evaluated. Positive association p =0.046 was found between number of sexual partners and *Chlamydia trachomatis* infection.

Conclusion: *Chlamydia trachomatis* a sexually transmitted infection is deadly infection because of its wide range of complications. Therefore, it should be diagnosed promptly and treated effectively. Due to its asymptomatic nature individuals having risk factors should be screened as early possible to avoid complications. Information about *Chalamydia trachomatis* risk factors and education about how to avoid infection should be given to individuals on large scale.

Keywords: *Chlamydia trachomatis*, Pelvic inflammatory disease, Polymerase chain reaction

BACKGROUND

Sexually transmitted infections (STIs) is a multiple faceted public health problem having consequences on health care systems globally.(1) STIs directly affect the health of individual and indirectly effect health care system due to economic liability they cause the health care system.[2] According to World Health Organization (WHO) 2018 report nearly 350 million individuals get infected with sexually transmitted infection every year of which seven percent cases of *Chlamydia trachomatis* infection are reported.[3]

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Chlamydia trachomatis presents as diagnostic dilemma due to its asymptomatic nature and remains undetected in 60 -70 percent of cases.[4] Patient presents to clinics when complications have developed and its uncontrolled transmission in community has occurred. Complications like pelvic inflammatory disease is common in females which is considered fatal as it effects mainly younger population and lead to infertility, ectopic pregnancy and death.[5] Infection with *Chlamydia trachomatis* increases the risk of being co infected with other STIs also.[6] This problem is more significant in South East Asia especially Pakistan where most cases are not diagnosed timely due to non-availability of diagnostic services, lack of awareness about safe sexual practices and social constraints. Therefore the need of hour is to conduct proper screening at Government level in resource limited countries and to create awareness about safe sexual practices among general public and to identify at risk

populations to reduce the spread of *Chlamydia trachomatis* infection and long term consequences associated with it.[7,8]

The main objective of our study was to identify and stratify risk factors associated with *Chlamydia trachomatis* infection in women suffering from pelvic inflammatory disease visiting a tertiary care hospital.[9]

In our study we performed polymerase chain reaction on urine specimen due to its noninvasive nature instead of endocervical swab which is routinely done. This method can be used as screening tool on larger scale as it requires noninvasive specimen collection technique, is a cost effective method and requires less expertise.[10, 11]

MATERIAL AND METHODS

This cross-sectional observational study was carried out at the Department of Microbiology, Army Medical College (National University of Medical Sciences), Pak Emirates Military hospital from March 2019 to September 2019 after obtaining ethical approval from institutional review board of institute.

Sixty females diagnosed with pelvic inflammatory disease were included in the study over a period of seven months after obtaining consent and confidentiality of participants were maintained at all times. Sample size was calculated by using the WHO calculator for sample size calculation by keeping Anticipated population proportion of 0.04 and confidence level of 95% (8) Inclusion criteria included all sexually active females between age of 16-49 years presenting with pelvic inflammatory disease. Males were excluded from the study as screening for males is not recommended. Females with other co-morbidities and who have already received treatment and duplicate samples from the same patient were excluded from the study.

For the purpose of this study, we defined Pelvic inflammatory disease as a clinical spectrum compromising of endometritis, salpingitis, tubo-ovarian abscess or pelvic peritonitis which was assessed through history of severe lower abdominal pain, dysuria, dyspareunia, fever, intermenstrual bleeding and prolonged menstrual cycle. We used imaging modalities to demonstrate the presence of tubo-ovarian abscess or thickened tubes with or without free pelvic fluid.

A total of sixty urine samples were collected from sixty female participants who fulfilled the study criteria and were tested for *Chlamydia trachomatis* using polymerase chain reaction assay. Urine samples were collected following aseptic guidelines and transported to laboratory in one hour otherwise specimen was stored at 2-8ºC. Specimens were stored at – 20 till tested for *Chlamydia trachomatis.* Once sample was received in laboratory 10 ml of urine was centrifuged at 1680g for 20 minutes. The supernatant was discarded, pellet processed according to manufacturer's instructions.

DNA extraction was done manually DNA was extracted from each urine sample using Pure Link Micro biome DNA purification kit Cat A29790 M/s Invitrogen, a commercially available kit. Real time PCR was done which amplified the target sequence of *Chlamydia trachomatis* cryptic plasmid gene a 71 bp DNA using a forward's primer and a reverse primer (5'–CATGAAAACTCGTTCCGAAATAGAA-3), (5'TC AGAGCTTTACCTAACAACGCATA-3') respectively following manufacturers guidelines. Instrument used was Smart cycler by Cepheid PCR system 16 well system. DNA was extracted from 200 µl thawed or from 200 µl aliquot of pellet from 10 ml of centrifuged urine.

Socio-demographic and behavioral data were collected and real-time PCR diagnostic method was used to detect the presence of *Chlamydia trachomatis* infection in urine samples. Samples were analyzed for risk factors identification and association with *Chlamydia trachomatis* infection using Statistical Package for the Social Sciences (SPSS) Version – 24.

RESULTS

Sixty female participants who fulfilled the study criteria were enrolled in the study. Frequency of *Chlamydia trachomatis* in urine specimen of this high-risk population was found to be twelve (20 percent) remaining forty-eight (80 percent) were negative for *Chlamydia trachomatis*.

Age was main factor which played a role in acquisition of *Chlamydia trachomatis* infection with maximum number of individuals in age group 20-24 being affected followed by 25- 29 years age group as shown in Table-I. Participant's age ranged from 16- 40 years

with mean age of 24.38 ± 4.85 years. Youngest participant was 16 years with oldest being 40 years.

Several risk factors were evaluated age, level of education, socioeconomic status, life time number of sexual partners, marital status, place of residence either urban or rural and past sexual history as shown in Table-I.

Socioeconomic level a major risk factor for acquiring *Chlamydia trachomatis* infection was evaluated as shown by table I. We grouped participants according to socioeconomic level. We stratified the study population as high, middle and lower socioeconomic level according to general gross income statistics of country. Around 54 (90%) subjects with low socioeconomic status while 5 (8.3%) subjects belonged to middle class and 1 (1.7%) of subjects were categorized as having upper middle socioeconomic status. Out of 12 positive subjects 10 (83.3 %) belonged to low socioeconomic status, 1 (8.3 %) belonged to middle socioeconomic status and 1 (8.3%) belonged to higher middle socioeconomic status as shown by Figure-1.

Table-I: Different factors affecting acquisition of *Chlamydia trachomatis* infection.

Age (range)	**16 -40 years**	
Age Groups in Years	**Positive n (%)**	**Negative n (%)**
15-19	2 (28.5)	5 (71.5)
20-24	5 (33)	10 (67)
25-29	4 (21)	15 (79)
30-34	nil	5 (100)
35-40	nil	4 (100)
40-44	1 (20)	4 (80)
45-49	nil	nil
Education		
Grade 5 or less than grade 5 (Elementary School)	5 (38.5)	8 (61.5)
Grade 5- Grade 8 (Middle School)	2 (11.7)	15 (88.3)
Grade 8- Grade 10 (High School)	4 (28.5)	10(71.4)
Grade 10- grade 12 (College)	1 (9.1)	10 (90.1)
More than grade 12 (Masters / University)	-	5 (100)
Socioeconomic Status (On basis of gross income)		
Lower income group	10 (18.5)	44 (81.5)
Middle income group	1 (20)	4 (80)
High income group	1 (100)	nil
Marital Status		
Married	12 (20)	48 (80)
Unmarried, divorced or widowed	-	-
No of life time sexual partners		
1	11 (20.4)	43 (79.6)
2	1 (16.5)	5 (83.5)
More than 2	-	-
Past STI History		
Present	7 (100)	-
Absent	5 (9.4)	48 (90.5)
Place of living		
Urban	10 (20)	40 (80)
Rural	2 (20)	8 (80)

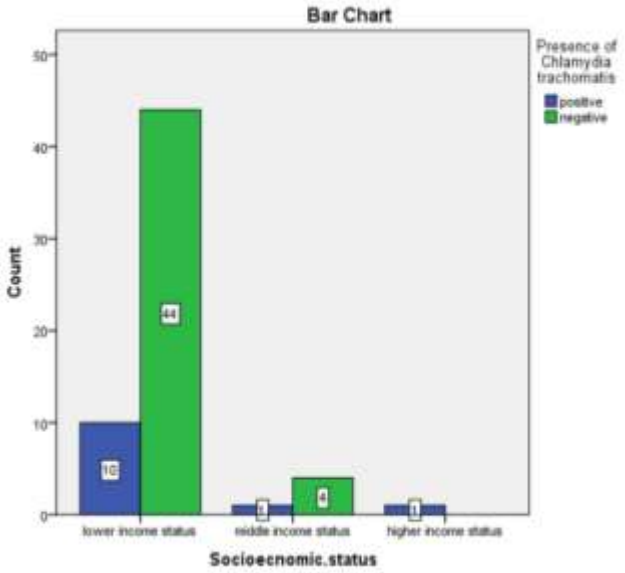

Figure-I: Frequency of *Chlamydia trachomatis* distribution among study participants according to socioeconomic status.

DISCUSSION

STIs effect not only the individuals health but also place economic burden not only on individual but also effect health care system of the country in particular and society in general by causing its uncontrolled transmission if not detected and treated in time. *Chlamydia trachomatis* is one of the most frequently occurring and easily treated STI worldwide. Around 131 million estimated cases of *Chlamydia trachomatis* occur annually raising global concern about its public health importance.[13,14]

Asymptomatic nature of infection makes diagnosis of *Chlamydia trachomatis* difficult. A study conducted at Karachi showed the prevalence to be 14.49 percent in symptomatic cases and 11.9 percent in asymptomatic persons[15] PID is the most commonly associated with *Chlamydia trachomatis* infection as a complication of its asymptomatic nature which can cause ectopic pregnancy which has significant mortality and morbidity.[16,17] It is responsible for 5-10 percent of all maternal deaths.[18]

A lot of research effort is taking place worldwide to assess risk factors associated with *Chlamydia trachomatis* infection so as to screen high risk groups early and to prevent its progression and further transmission. In Pakistan, however not much data is available that deals with *Chlamydia trachomatis* risk factor association. High risk sexual behaviours in our study was only studied in females who were heterosexuals so we could not evaluate these like male having sex with males, bisexual all these factors predispose to Chlamydial infection[19] 8.3 percent of Chlamydia positive females had more than one life time sexual partner. In our study positive association was found between no of sexual partners and diagnosis of *Chlamydia trachomatis* which correlates with international studies.[20]

In our study 20- 24 years age group was mainly affected which is consistent with other studies carried out in south east Asia.[21] A study carried out in Malaysia having similar socio demographic similarities as Pakistan showed infection was common in younger age group mainly under the age of 28 years.[22] The mean age of females infected by *Chlamydia trachomatis* was 24.38 years consistent with other studies on same subject. High risk groups are screened regularly in developed countries same should be done in Pakistan.[23] Screening should be compulsory for individual in age group from 20 to 30 years.

World over infection and complications occur more in socially disadvantaged population due to lack of knowledge about disease and accessibility to diagnosis and treatment. In our study maximum cases were seen in females belonging to disadvantaged socioeconomic status and education similar to studies carried out in Europe , North America and Australia.[24,25] There was no significant association found between education in our study mainly because major proportion of our population belonged to socially disadvantaged background as our hospital mainly caters for people belonging to lower socioeconomic status so a significant comparison cannot be done.

CONCLUSION

Due to asymptomatic nature of *Chlamydia trachomatis* infection, its ability to develop wide variety of complications high risk individuals should be identified and treated as soon as possible. Education about prevalence, risk factors and safe sexual practices should be made freely available to high-risk population.

LIMITATIONS

The main limitation of our study is it has small sample size, and is a single Centre study. A similar multi centre-based study on large scale should be carried out to determine the risk factors associated with *Chlamydia trachomatis* infection in our population.

RECOMMENDATIONS

Prevalence of *Chlamydia trachomatis* and its risk factors data is scarcely available from Pakistan, low economic status of majority of population and being a densely populated country Pakistan is high risk country

for it. Although our study was conducted for eighth months at a tertiary care hospital in one of the largest cities of Pakistan still a multicentre cohort study on larger scale should be carried out to identify risk factors.

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CONFLICTS OF INTEREST

The authors declare no conflicts of interest

AUTHOR CONTRIBUTION

Afnan Naeem: Principal investigator
Hafsa Waseem: Data collection
Sakhawat Ali: Review of study
Javaid Usman: Conception of idea and study design
Faisal Hanif: Review of study
Warda Furqan: Statistical analysis

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