ORIGINAL ARTICLE

DETECTION OF CHLAMYDIA TRACHOMATIS ANTIGEN DIRECTLY FROM ENDOCERVICLE SPECIMEN

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ABSTRACT

Genital infections caused by Chlamydia trachomatis is the most common bacterial sexually transmitted disease (STD's) in world. Therefore it is important to screen high risk population and to diagnose the infection in early stage to prevent it's serious sequel
Aim : To detect Chlamydia trachomatis antigen directly from endocervicle specimen
Study design : It is cross sectional study conducted in S.S.G.Hospital ,Vadodara, Gujarat for one year.
Method : Endocervicle specimen from cases have been examined for Chlamydia trachomatis antigen by Immunochromatographic Immunoassay
Results: Out of 200 high risk female patients, Chlamydia trachomatis antigen was detected in 24 cases (12 %)
Conclusion : Study reports that C.trachomatis is a important cause of genital infections in females and Immunoassay is the rapid and accurate test for screening high risk population

Keywords: Chlamydia trachomatis, Sexually transmitted Disease, endocervicle specimen, antigen

INTRODUCTION

Genital infections caused by Chlamydia trachomatis is the most common Bacterial Sexually transmitted Disease (STD's) in world. An estimated 4 million cases occurs each year. (1)
In females C. Trachomatis is represented to cause 10-30 % of a spectrum of infections including cervicitis, urethritis, endometriosis, salpingitis with subsequent complications of ectopic pregnancy, infertility and recurrent spontaneous abortion and in pregnancy premature rupture of membrane, preterm delivery and stillbirth. (2)
The Chlamydial infections commonly are asymptomatic (usually 40%) or cause mild or nonspecific symptoms and signs. Therefore, screening of women as high risk is highly recommended. (3)
Women at highest risk often have the least access to healthcare facilities. The Laboratory diagnosis of C. Trachomatis genital infection is dependent upon specific laboratory identification. The medical history and physical examination while necessary in every instance are neither sensitive nor specific enough to identify patients. (2)
Therefore there is need for a rapid simple and accurate test to detect C. trachomatis infection, which can be performed outside the laboratories setting when the patient is still in clinical setting. (4)
Infection with Chlamydia Trachoma is the most prevalent bacterial sexually transmitted disease (STD's) and most often it is asymptomatic. Moreover in women it has serious sequel leading to significant morbidity. (5)
Since specific treatment for managing the condition is available, it is important to screen high risks population and to diagnose the infection in early stage to prevent it's further spread.(6)

METHOD

Females attending the obstetrics and gynecology out Patient Department of Shree Sayaji Rao General Hospital, Baroda were screened for Chlamydia Infection. The period of study was one year.
A standard questionnaire including Age, chief complains (if leucorrhoea than duration, amount, color, odor, itching),associated complains (Abdo Pain, urinary complain, disparunea, etc)
Besides this Educational status ,Socioeconomic background, Occupation, Menstrual history, Marital status ,Obstetric history ,Sexual activity ,Comptraceptive history,Previous H/o STD and Treatment if taken was asked to every patient
Accordingly 200 female patients with high risk are selected.

The study was thoroughly explained to the patient and a written consent was obtained before taking any specimen.

A complete gynecological examination for lesions on external genitalia and a speculum examination to detect any cervical lesion or inflammation was done by the gynecologist.

Clinical Specimen collection

The specimen was collected as per the instructions of the Kit **Biosign TM Chlamydia-II,** manufacturer.

To obtain a satisfactory specimen from cervix the patients was examined in lithotomy position and the cervix was visualized using bivalve speculum and anterior vaginal wall retractor.

First excess mucus was removed using large gauge moistened with sterile physiological saline. Than the swab provided with the kit was inserted approximately 1 cm in to the cervicle canal and rotated for 15-20 seconds before withdrawing. The swab was removed without touching the vaginal surface and placed in the sample collection tube.

The specimens were immediately transferred to the laboratory for EIA testing.

MATERIAL

"**Biosign TM Chlamydia II**" **Direct Chlamydia trachomatis antigen test** (manufactured by Princeton Biomeditech corporation (PBM)- USA) was used that is a direct binding monoclonal based immunochromatographic immunoassay for the qualitative detection of Chlamydia trachomatis antigen directly from endocervicle specimen.

Principle:

The "Biosign Chlamydia" test, it is a rapid qualitative immunoassay based on the solidphase immuno chromato graphic sandwich principle. The method employ a unique combinations of monoclonal dye conjugate and polyclonal solid antibodies to selectively detect lipopolysaccharide (LPS) antigen of Chlamydia trachomatis species with high degree of sensitivity.

In this test the endocervicle specimen is first treated with an extraction reagents to isolate Chlamydia trachomatis antigen if present. Following extraction of C. trachoma its antigen, the extract is added to the sample well of the test device with the aid of transfer pipette.

As the test samples flow through the absorbent pad, the labeled antibody dye conjugate binds to the genus specific LPS antigen of Chlamydia forming an antigen antibody complex. The pad is in contact with a chromatographic membrane strip which coated with immobilized polyclonal anti-LPS antibody in the test window. The antigen antibody complex moves by capillary action along the membrane strip forming a line of immobilized complex by a zone of antibody in the test window.

The presence of pink line in test window indicates the presence of Chlamydia antigen in the extract while the absence of line in the test window indicates negative results. The appearance of a pink line in the control window shows that the test has been carried out correctly.

RESULTS

Out of 200 female patients included in the study group , Chlamydia trachomatis antigen was detected in 24 cases

So the overall incidence of C. trachomatis in genital tract of female patients was found to be 12% in this study

Table 1: Different High risk Groups v/s C. Trachomatis Antigen

High risk Groups	Total no. of Patients	Positive by EIA	Incidence (%)
Chronic Cervicitis	92	12	13.04
PID	66	7	10.60
Infertility	36	3	8.3
Ectopic Pregnancy	2	-	0.0
Recurrent Spontaneous Abortion 1,2,3>	45	8	17.7

[Patients with two associated clinical conditions were included in more than one category]

Highest rate of recurrent spontaneous abortion followed by that with the history of cervicitis, PID and infertility 13.04%, 10.6% & 8.3% respectively.

Table 2: Age Distribution of Enrolled Subjects

Age Groups	Total no. of Patients	Positive by EIA	Incidence (%)
</=20	13	3	23.07
21-30	108	17	15.70
31-40	74	4	5.40
>/=40	5	0	0.00

Highest incidence of C. trachomatis antigen was found with younger age usually <20 year and majority of other cases were between age 20-40 year.

There was no single positive cases found above 40 year of age.

Table 3: Clinical Manifestations of C. Trachomatis Infections

Clinical Manifestations	Total no. of Patients	Positive by EIA	Incidence (%)
Symptomatic	178	22	12.30%
Asymptomatic	22	2	9.09%

89% of the total 200 cases studies were Symptomatic having H/O discharge, Abdominal pain, dysuria or dysparunia while 11% of patients were asymptomatic clinically and came to OPD for investigations of their infertility or recurrent abortions.

Table 4: Clinical Symptoms v/s C. Trachomatis Antigen

Symptoms	Total no. of Patients	Positive by EIA	Incidence (%)
Cervical Discharge	168	21	12.5%
Abdominal pain	130	18	13.8%
Dysuria	30	3	10.0%
Dysparunia	8	1	12.5%
None	22	2	9.09%

[Patients with multiple symptoms were included in more than one category]

Cervicle discharge and abdominal pain had strong association with c.trachomatis infections. The discharge was found usually yellowish white color, moderate in amount and usually not associated with foul-smelling or iching.

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DISCUSSION

EIA directed against various Chlamydial antigens have been introduced to facilitate laboratory screening and diagnosis of Chlamydial infections.

The prevalence of C.trachomatis in genital tract of women reported by various authors ranges from 8.0 % - 21.3 % (7,8), But overall prevalence varies depanding upon the Population studied and the sensitivity of the EIA used. Moreover EIA has been reported to give false positive results in cases of mixed infections in genital tract as endocervicle specimen may likely contaminated with vaginal secretions.

200 patients included in this study were attending Obs & Gyn OPD for various clinical complaints. All were married and were in the reproductive age group (18 – 48 yr).

Out of 200, 178 were symptomatic while 22 were asymptomatic, Therefore use of symptoms of genital tract infection as a diagnostic index for C.trachomatis infection has been found to be nonspecific because chlamydial infection may be asymptomatic in many cases.(9)

However the incidence of C.trachomatis was found to be higher In symptomatic patients than asymptomatic patients.(10)

Selection of the most appropriate laboratory test for C.trachomatis infection depends on local availability and expertise, the prevalence of infection in the test population and the intended purpose of the test, However in the population of women with prevalence of infection > 10% , EIA test is the most useful screening method with satisfactory sensitivity and specificity, rapid turnaround time and simplified transport condition.

CONCLUSION AND RECOMMENDATION

The overall incidence was found being 12% by "Biosign Chlamydia – II" immunoassay for C. Trachomatis antigen detection. The Chlamydial lipopolysaccharide (LPS) antigen was detected in 13.04% cases of cervicitis, 10.6% cases of PID, 8.3% cases of Infertility and 17.7%, Cases of recurrent spontaneous abortion.

The highest incidence was found with younger age usually <= 20 years suggesting that this age group are at high risk for C.trachomatis infection.

Out of 200 cases 11% were symptomatic and showed 9.09% of positivity for C.,trachomatis antigen. Other 89% were symptomatic but had nonspecific symptoms and they were mostly mild in nature. Thus it is worth while screening for this infection specially in high risk group to initiate prompt and complete treatment to prevent it's further spread and complications. EIA directed against various chlamydial antigen have been introduced to facilitate laboratory screening and diagnosis of C.trachomatis infection.

The test is cost effective and sensitivity being 70-90% as judged by reports in literature. Moreover it is rapid, accurate and easy to perform. So EIA recommenced as a diagnostic OR screening test in moderate to high risk population.

REFERENCES

1. Walter E.Stamm, Harrisons Principles of Medicine, Volume1,chapter 181,,Chlamydial infections , p 1055.
2. Robert B. Jones , Mendells infectious Diseases Volume II, chlamydial diseases , section c , p 1676.
3. H Hunter, Hands Field, Lora, L. Jasmar, Criteria for selective screening for Chlamydia trachomatis infection in women attending family planning clinics. JAMA 1986; 1730: 255.
4. R Malenie, P J Joshi, MD Mathur, Chlamydia trachomatis antigen detection in pregnancy and its verification by antibody blocking assay. IJMM2006;24 (2):97-100.
5. Stamm WE, Holmes K. K. Mardh PA., Public Health Implications and control of sexually transmitted Chlamydial infections. Sex.Tra.Dis.1990,181, p 93.
6. Stamm W.E. ,B.Cole. Asymptomatic Chlamydia trachomatis arthritis in men. S.T.D. 1986,13, p163.
7. Monica Arora, Sarala Malhotra, Merra Sahrma, Role of C.trachomatis in PID, Jan1992, I J M R (A), 89, p 87
8. Krishna Ray, S. Yadav, Prevalence of C.trachomatis and other sexually transmitted pathogens in female RTIs, Ind. Jr. Med. Micro. 1997;15(4):173
9. Barbara Mosscicki, mary-ann Shafa, Susan G.Millstein, The use and limitations of endocervicle gram stain and mucopurulent cervicitis as a predictor for C.trachomatis infection, Am.Jr.obs.Gyn.1987;157: p 65
10. Laurence S. Magder, Karl C. ,K Lontz, Linda H. Bush, Effect of patients characteristics on performance of an EIA for detecting cervical C.trachomatis infection, Jr. Clin. Micro. 1990;28(4):781.