Role of Genital Tuberculosis and Chlamydia Trachomatis as a Tubal Factor in Primary Infertility

MADIHA M. EL-ATTAR, M.D.*; HEBAT-ALLAH G. RASHED, M.D.*; HOSSAM THABET, M.D.**; SAFWAT ABD EL RADY, M.D.** and SALMA ABD EL HAMED, M.D.**

The Departments of Clinical Pathology*, Obstetrics & Gynecology** and Tropical Medicine & Gastroenterology***, Assiut University Hospital, Egypt.

Abstract

Background: Infertility has become nowadays not only a medical, but a social problem as well. Infertility in female is caused by various factors. Tubal disease is responsible for 25-35% of female infertility. Pelvic inflammatory disease (PID) is the most common cause of tubal disease.

Aim of the Study: To assess the rate of M. tuberculosis and C. trachomatis infections in patients with tubal factor infertility and to determine the most reliable method for laboratory diagnosis of genital tuberculosis and C. trachomatis using ELISA with respect to laparoscopic finding.

Methods: Ninety women with primary infertility and abnormal Hysterosalpingography were enrolled as a studied group and 30 infertile women with polycystic ovary as the only cause of infertility and normal Hysterosalpingography as a control group. Laparoscopy was performed and peritoneal biopsy was taken for histopathological study. ELISA detection for Mycobacterium tuberculosis and C. trachomatis IgG antibodies was done. PCR for detection mpt64 gene of M. tuberculosis and C. trachomatis plasmid DNA also were done in peritoneal fluid.

Results: IgGAB for M.tuberculosis were +ve in 72 cases (80%) of the studied group and laparoscopy was +ve in 23 cases (76.7%) of the control group with no statistical significant difference. PCR for mpt64 gene of M. tuberculosis in peritoneal fluid was done for all patients; only 17 cases (18.9%) of the study group showed +ve results. IgG for C. trachomatis were +ve in 43 cases (47.8%) of the study group and it was +ve in 5 cases (13.3%) of the control group with height statistical significant difference. The +ve titers range from >5IU to 40IU. PCR was done for Chlamydia in peritoneal fluid only 5 cases (5.6%) of the studied group was +ve for chlamydia PCR and it was –ve in all control groups.

Conclusions: Genital tuberculosis continued to be an important cause of infertility in Egypt. PCR for mpt64 gene of M. tuberculosis plays an important role in diagnosis of genital tuberculosis with results comparable with laparoscopy and histopathology.

Detection of C. trachomatis IgG antibodies by ELISA can play a significant role in detection of C. trachomatis in infertile women. Screening of women with primary infertility for C. trachomatis is recommended in the first year of infertility, so that early therapeutic intervention can be instituted to allow women to conceive naturally.

Key Words: mpt64 gene of M. tuberculosis – C. trachomatis plasmid DNA.

Introduction

INFERTILITY has become nowadays not only a medical, but a social problem as well [1]. The incidence of infertility in any community varies between 5% and 15% [2]. Infertility in female is caused by various factors. Tubal disease is responsible for 25-35% of female infertility [3]. Pelvic inflammatory disease (PID) is the most common cause of tubal disease, representing greater than 50% of cases, and may affect the fallopian tube at multiple sites [4]. Checking the tubal patency is one of the important steps in investigating the female partner. Hysterosalpingography, Hysteroscopy, laparoscopy, salpingoscopy and sonosalpingography are the different techniques available. Laparoscopy has an additional advantage that one can visualize the other abdominal and pelvic structures, can take biopsy and can do adhesiolysis and laser surgery as well [5]. Tubal pathology can be managed medically and surgically. Medical treatment includes antibiotics for pelvic inflammatory disease, antituberculous for pelvic tuberculosis and medical treatment of endometriosis [6]. The incidence of genital tuberculosis (TB) cannot be assessed in any population in spite of the truth that T.B remains a major health problem in developing and developed countries because it is discovered incidentally in many patients and in large number of symptomless patients it remains undiscovered [7]. It is estimated that 5-10% of infertile women the worldwide have genital tuberculosis although
this varies from less than 1% in the United States to nearly 18% in India [8]. Laboratory diagnosis of TB depends on demonstration of the causative organisms, the use of PCR for detection of tuberculosis was considered as a rapid, sensitive and specific molecular technique for detecting mycobacterium DNA in both pulmonary and extra pulmonary samples including fluids from suspected TB patients. PCR assays targeting various gene segments, including a 65kD heat shock protein-encoding gene [9], and the mpt 64 gene [10,11] have definite mycobacterium detection in the lab in 2-3 days, beside being more sensitive than conventional methods [12].

Chlamydia trachomatis, is a major reproductive health care issue in women. It is the most important preventable cause of infertility and adverse pregnancy outcome. The magnitude of morbidity associated with chlamydial genital infection is enormous; including salpingitis, endometritis, PID, ectopic pregnancy and tubal factor infertility [13]. Currently, there are several challenges to be addressed in the control of C. trachomatis. Infection; these include a misperception of the prevalence of infection in different population, the fact that infection is often asymptomatic or presents with nonspecific symptoms and lack of availability of sensitive diagnostic tests. The key to the prevention of C. trachomatis infection rests on the ability to make diagnosis based on accurate lab testing [13].

This study aimed to assess the rate of tuberculosis and C. trachomatis infection in patients with tubal factor infertility and to determine the most reliable method for laboratory diagnosis of genital tuberculosis and C. trachomatis using ELISA for detection of TB and C trachomatis IgG antibodies and PCR for detection of mycobacterium tuberculosis DNA and C. trachomatis plasmid DNA with respect to laparoscopic finding.

Patients and Methods

This study is a comparative hospital-based study included 120 infertile women with history of primary infertility for at least one year up to twenty years who were attending to the Obstetric and Gynecology Department in Assiut University Hospital from June 2005 to May 2007. Ninety women with primary infertility and abnormal Hysterosalpingography (tubal block) were enrolled as a studied group and 30 infertile women with polycystic ovary as the only cause of infertility and normal Hysterosalpingography (patent tube) as a control group. Their mean age was 27.6±5.93 years and 26.5±5.93 years respectively.

An informed consent was obtained from all patients and controls enrolled in the study. The Ethical Committee approved this study and the study was carried out according to the principles of the Declaration of Helsinki. A detailed history taking and clinical examination were performed for every patient. Abdominal, pelvic and vaginal ultrasonography to evaluate the uterus and ovaries was done.

Laparoscopy was performed according to Nooman et al. [14] only for the studied group. Aspiration of free fluid in Douglas pouch was done during laparoscopy. Biopsy from suspected tissue was taken if possible (tubercles, adhesion, and cyst) for histopathological study.

Specimen: Three ml venous blood was collected after 10-12 hours fasting and collected in plain tubes. The samples were centrifuged within 30 minutes at 3000 rpm for 10 minutes and the serum samples then collected and divided into aliquots and stored at 70ºC for further analysis.

ELISA detection for Mycobacterium tuberculosis and C. trachomatis IgG antibodies was performed using immuno-biological laboratories, INC (IBL-America) Cat. No IB 19202 and Cat. No IB79284 respectively. Peritoneal fluid was subjected to microbiologic and PCR analysis.

Peritoneal fluid sample: Fluid from Douglas pouch was aspirated during laparoscopy by a long canula adapted to a disposable syringe, transported to the laboratory in sterile vials.

Microbiological study: Peritoneal fluid decontamination was performed in 4% NaOH. About 250ul of concentrated sediment used for Direct smear by Ziehl-Neelsen’s (ZN) stain and growth on (LJ) medium at 37ºC. Growth was monitored for 8 weeks.

Polymerase chain reaction (PCR): DNA isolation was performed from the sediment by using a QIAmp DNA Mini Kit with a bacterial DNA extraction protocol.

- PCR for M.tuberculosis: A 240bp region of the mpt64 gene of M. tuberculosis was amplified using primers MPT1 (59-TCCGCGCCAGT CGTCTTCC-39; nt 460-479) and MPT2 (59-GTCCTCGCGTCTAGGCCA-39; nt 700-681). For amplification we used puReTaq Ready-To-Go PCR Beads (GE Health care lot number 27-9557-01). The PCR was carried out in 25ul volumes, (1ul of each primer, 5ul of DNA, 18ul of sterile high quality water and PCR Beads). Using thermal cycler (Express) supplied by Hy-
baid, amplification was carried out for 30 cycles each consisting of denaturation at 94°C for 2 min, annealing at 60°C for 2 min and extension at 72°C for 2 min.

- **PCR for C. trachomatis**: A 570bp C. trachomatis plasmid DNA detection was performed by PCR using the primer sequence as follow (5’-TCC GGA GCG AGT TGA GAA GA-3’) and 5’ -AAT CAA TGC CCG GGA TTG GT-3’). For amplification we used puReTaq Ready-To-Go PCR Beads (GE Health care kit) lot number (27-9557-01). The PCR mixture was as follow (1ul of each primer, 5ul of DNA, 18ul of sterile high quality water and PCR Beads). The amplification profile was 1 min at 95°C, 1 min at 64°C and 1 min at 72°C for 35 cycles and 5 min at 72°C for one cycle. Products were detected by electrophoresis on 2% agarose gel after staining with ethidium bromide.

**Statistical analysis**: Was performed with SPSS 12 for Windows (SPSS Inc., Chicago, IL, USA) Results were expressed as percentage. A p-value less than 0.05 was considered statistically significant. According to the data distribution, comparison between the groups was made using unpaired t-test (two-tail) or nonparametric analysis (Mann-Whitney U test).

**Results**

**Patients' characteristics**: A total of 120 female patients presented with primary infertility were recruited in this study. Ninety women with primary infertility due to tubal factor were included as study group and 30 infertile women due to other cause of infertility (polycystic ovary) as a control group. The mean age of the study group was 27.60 ± 5.1 years (range 18-40 years) and the mean age of control group was 26.25 ± 3.88 years (range 19-37 years). The mean value of duration of infertility in the study groups was 27.60±5.1 years (range 18-40 years) and the mean age of control group was 26.25 ± 3.88 years (range 19-37 years). The mean value of duration of infertility in the study groups was (6.89 ±4.34) years and (3.68±2.29) years in control groups. The presenting symptom of most cases was infertility. Only some cases positive for TB PCR were symptomatic (3 with pelvic pain, 4 with menstrual dysfunction, 3 with abdominal distension and one with vaginal discharge). All cases positive for Chlamydia (either antibodies or PCR) were asymptomatic.

The laparoscopic findings in studied infertile female (90 patients) Table (1) showed the laparoscopic findings in 90 infertile female. The most frequent presenting finding was tubal block with adhesion in 20 patients (22.22%), unilateral hydrosalpinx in 12 patients (13.33%) and block without adhesion in 11 patients (12.22%).

**Microbiological study**: Acid fast bacilli were detected by direct Ziehl-Neelsen’s (ZN) smear in 2 (2.2%) patients of the studied group (with abnormal hysterosalpingography). Positive growth on LJ medium at 37°C was detected 5 (5.5%) patients.

**Mycobacterium tuberculosis and of C. trachomatis IgG antibodies**: IgG antibodies against Mycobacterium tuberculosis was detected in 72 (80%) patients of the studied group and in control group it was detected in 23(76.7%) with no statistically significant difference. IgG antibodies against C. trachomatis was detected in 43 patients (47.78%) of the studied group and in control group it was detected in 4 patients (13.33%) with a statistically significant increase in the studied group (p<0.00) (Table 2).

**Mycobacterium tuberculosis DNA and C. trachomatis plasmid DNA**: Mycobacterium tuberculosis DNA and C. trachomatis plasmid DNA were detected in 17 patients (18.89%) and 5 patients (5.56%) respectively of the studied group. All controls were negative for Mycobacterium tuberculosis DNA and C. trachomatis plasmid DNA (Table 2).

**Laparoscopic findings in 17 patients with mycobacterium tuberculosis positive DNA patients**: Table (3) showed the laparoscopic findings in Mycobacterium tuberculosis DNA the frequent presenting finding was tubal block with adhesion (29.41%), tubo-ovarian mass in (23.53%) and frozen pelvis and hydrosalpinx in (17.6%).

<table>
<thead>
<tr>
<th>Laparoscopy finding</th>
<th>No. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal pelvis</td>
<td>11 (12.22%)</td>
</tr>
<tr>
<td>Tubal block with adhesion</td>
<td>20 (22.22%)</td>
</tr>
<tr>
<td>Tubal block without adhesions</td>
<td>11 (12.22%)</td>
</tr>
<tr>
<td>Tubal block with cystic ovary</td>
<td>4 (4.44%)</td>
</tr>
<tr>
<td>Tubo-ovarian mass</td>
<td>8 (8.88%)</td>
</tr>
<tr>
<td>Frozen pelvis</td>
<td>7 (7.77%)</td>
</tr>
<tr>
<td>Fitz Hug Curtis Syndrome</td>
<td>2 (2.22%)</td>
</tr>
<tr>
<td>Endometriosis</td>
<td>5 (5.55%)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Hydrosalpinx:</th>
<th>No. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unilateral</td>
<td>12 (13.33%)</td>
</tr>
<tr>
<td>Bilateral</td>
<td>6 (6.67%)</td>
</tr>
<tr>
<td>Salpingitis</td>
<td>4 (4.44%)</td>
</tr>
</tbody>
</table>

N.B.: More than one laparoscopic finding could be found in a single patient.
Table (2): Serological results (TB and C. trachomatis IgG antibodies) and PCR in the studied patients and control groups.

<table>
<thead>
<tr>
<th>Item</th>
<th>Studied patient group</th>
<th>Control group</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N (90)</td>
<td>N (30)</td>
<td></td>
</tr>
<tr>
<td>Direct smear by Ziehl-Neelsen’s (ZN) stain</td>
<td>2 (2.2%)</td>
<td>0</td>
<td>N.S</td>
</tr>
<tr>
<td>Culture in LJ medium</td>
<td>5 (5.5%)</td>
<td>0</td>
<td>N.S</td>
</tr>
<tr>
<td>TB IgG antibodies</td>
<td>72 (80%)</td>
<td>23 (76.7%)</td>
<td>N.S</td>
</tr>
<tr>
<td>TB PCR</td>
<td>17 (18.89%)</td>
<td>0</td>
<td>0.02</td>
</tr>
<tr>
<td>C. trachomatis IgG antibodies</td>
<td>43 (47.78%)</td>
<td>4 (13.33%)</td>
<td>0.00</td>
</tr>
<tr>
<td>C. trachomatis PCR</td>
<td>5 (5.56%)</td>
<td>0</td>
<td>N.S</td>
</tr>
</tbody>
</table>

Table (3): Laparoscopic findings in TB positive PCR studied patients group.

<table>
<thead>
<tr>
<th>Laparoscopic Finding TB positive PCR patients</th>
<th>No. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tubal block with adhesions</td>
<td>5 (29.41%)</td>
</tr>
<tr>
<td>Tubal block without adhesions</td>
<td>2 (11.76%)</td>
</tr>
<tr>
<td>Frozen pelvis</td>
<td>3 (17.64%)</td>
</tr>
<tr>
<td>Hydrosalpinx</td>
<td>3 (17.64%)</td>
</tr>
<tr>
<td>Tubo-ovarian mass</td>
<td>4 (23.53%)</td>
</tr>
</tbody>
</table>

Laparoscopic findings in C. trachomatis IgG antibodies positive patients: Table (4) showed that the most frequent presenting finding was tubal block with adhesion (34.88%), hydrosalpinx (27.91%) and tubal block without adhesion in (13.95%).

Histopathology: Tissue biopsy was taken from 10 cases from peritoneal tubercles and adhesions in cases with frozen pelvis for histopathology and it was positive in 5 cases for TB and nonspecific in 5 cases.

Table (4): Laparoscopic finding in C. trachomatis positive IgG antibodies plasmid DNA positive studied patients group.

<table>
<thead>
<tr>
<th>Laparoscopic finding</th>
<th>C. trachomatis IgG Ab No. (43)</th>
<th>Plasmid DNA No. (5)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tubal block with adhesions</td>
<td>15 (34.88%)</td>
<td>1 (20%)</td>
<td></td>
</tr>
<tr>
<td>Tubal block without adhesions</td>
<td>6 (13.95%)</td>
<td>1 (20%)</td>
<td></td>
</tr>
<tr>
<td>Frozen pelvis</td>
<td>2 (4.65%)</td>
<td>p=0.04</td>
<td></td>
</tr>
<tr>
<td>Hydrosalpinx</td>
<td>12 (27.91%)</td>
<td>1 (20%)</td>
<td></td>
</tr>
<tr>
<td>Salpingitis</td>
<td>4 (9.3%)</td>
<td>1 (20%)</td>
<td></td>
</tr>
<tr>
<td>Tubo-ovarian Mass</td>
<td>2 (4.65%)</td>
<td>1 (20%)</td>
<td></td>
</tr>
<tr>
<td>Fitz Hug Curtis syndrome</td>
<td>2 (4.65%)</td>
<td>-</td>
<td></td>
</tr>
</tbody>
</table>

Discussion

Infertility has become nowadays not only a medical, but a social problem as well. The incidence of infertility in any community varies between 5% and 15% [2]. Pelvic inflammatory disease (PID) is the most common cause of tubal disease, representing greater than 50% of cases and may affect the fallopian tube at multiple sites [4]. Checking the tubal potency is one of the important steps in investigating the female partner. Hysterosalpingography, Hysteroscopy, laparoscopy, salpingoscopy and sonosalpingography are the different techniques available. Laparoscopy has an additional advantage that one can visualize the other abdominal and pelvic structures, can take the biopsy and can do adhesiolysis and laser surgery as well [5,6].

Tuberculosis (TB) is an infectious disease that is involved in about 5-16% of cases of infertility among developing countries women [18], though the actual incidence may be under-reported due to asymptomatic presentation of genital tuberculosis and paucity of investigations. The laboratory diagnosis of TB depends on demonstration of the causative organisms, we use PCR for detection of mycobacterial DNA which is sensitive (for detecting DNA even in dead bacilli in infected area or exudates from infected organ) and rapid (did not wait for culture which take up to 8 weeks). The detection sensitivity of the PCR assay was evaluated and it was found that as little as 5 fg of M. tuberculosis DNA from H37Ra (corresponding to one organism) spiked in water and 25 fg (five organisms) spiked in the pouch of Douglas could be detected [16].

In the present study acid fast bacilli were detected by Direct by Ziehl-Neelsen’s (ZN) smear in 2 (2.2%), positive culture results were detected in 5 (5.5%) and PCR for mpt64 gene of M. tuberculosis were detected in 17 (18.9%) these findings in agreement with Roya et al., who reported that the sensitivity of the PCR assay, by comparing PCR with other tools of laboratory diagnostic techniques, AFB staining gave the lowest detection rate (8/65; 5.2%). Culture also gave a low detection rate (12/65; 7.8%) and required a long time to obtain an answer (minimum time to see colonies were 4 weeks) [17].

In our study the PCR for mpt64 gene of M. tuberculosis was detected in 17 (18.8%) patients of the studied group these findings in agreement with Bhanu et al., who assessed the role of PCR in the diagnosis of genital tuberculosis and reported 16% PCR positive cases out of 25 infertile women.
Bapna et al., also reported that the average incidence of genital tuberculosis in infertility clinics throughout the world varies depending on the country. It varies from less than 1% in the Australia and United States to 17.4% in India and this difference may be related to the area of study and type of samples [18]. Our results are less than those from Ethiopia who reporting 48% PCR positive biopsies and curettage samples from 25 women with complaints of infertility [19].

In the present work, the median age of women with confirmed genital tuberculosis was 27 years (range 18-40 years) this finding in agreement with Bapna et al. and Crofton et al., who reported that female genital tuberculosis has been described as a disease of young women, with 80-90% of patients diagnosed between 20 and 40 years of age [18,20].

In present study, patients with positive PCR for mp64 gene of M. tuberculosis presented mainly with infertility only in 6 patients, 3 with pelvic pain, 4 with menstrual dysfunction, 3 with abdominal distension and one with vaginal discharge; this finding were supported by Gupta et al., who reported that women with genital tuberculosis usually present with infertility (primary or secondary), chronic lower abdominal or pelvic pain, menstrual dysfunction, oligomenorrhea amenorrhea, and rarely, menorrhagia and sometimes asymptomatic [21].

In our study, laparoscopic findings in the 17 patients with genital tuberculosis showed tubal involvement in 88.2% and a frozen pelvis in 29.4%; these findings in agreement with the study of Bapna et al., who reported that the tubes show tubercular salpingitis, tubal block, hydrosalpinx, frozen pelvis, or adhesions [18]. Moreover Tripathy and Tripathy, reported the presence of adhesions, tubercles and hyperemia in 59.6% cases as well as adhesions in the pouch of Douglas in 11.3% of cases [22], while Bhide et al., observed pelvic adhesions in 48% cases, tubercles in 33.8%, unilateral adnexal mass in 11.3%, and bilateral adnexal masses in 22%, encysted effusion in 8.45%, and lesions on the bowel or omentum in 25.4% of cases [23]. In the present study, a very high prevalence of pelvic and abdominal adhesions was observed, which has also been reported by [21,24].

C. trachomatis has currently emerged as the most common sexually transmitted pathogen. Chlamydial infection produces less severe symptoms than other sexually transmitted diseases. These deceptively mild symptoms allow the infection to go unnoticed with minimal patient awareness until secondary or tertiary symptoms develop. The sequelae of undetected and thus untreated infections like acute salpingitis and pelvic inflammatory disease lead not only to significant morbidity but far more importantly to infertility. Infertility due to C. trachomatis represents a preventable type of infertility, if detected early. C. trachomatis is one of the most common sexually transmitted pathogens of humans, with an estimation of 92 million new cases occurring worldwide each year [25].

Therefore, we tried to compare recently developed nucleic acid amplification assays with enzyme immunoassay in diagnosis of C. trachomatis as a cause of infertility in this study.

In our study C. trachomatis IgG antibodies were detected in 47.8% of studied patients group and in 13.3% of control group this finding in agreement with Wilkowska-Trojniel et al., who reported that, Chlamydia- specific IgG antibody was detected 39.1% in women with tubal factor infertility [26]. While Sharma et al., reported that, Chlamydia-specific IgG antibody was significantly higher (70%) in women with tubal factor infertility, verified by hysterosalpingography and laparoscopy, than in healthy fertile women (35%) and infertile women with causes other than tubal factor infertility (55%) [24].

In the present work, the most frequent presenting finding in patients with positive C. trachomatis IgG antibodies was tubal block with adhesion (34.88%), hydrosalpinx (27.91%) and tubal block without adhesion in (13.95%). These findings supported by Mol et al., who performed a meta-analysis comparing chlamydia antibody testing with HSG for the diagnosis of tubal occlusion, using laparoscopic chromopertubation as the gold standard. They found that ELISA had sensitivity and specificity of 75%, as the discriminative ability of chlamydial antibody testing was comparable to that of HSG, the authors concluded that chlamydial antibody testing can be used instead of HSG in the initial screening for tubal disease. Yet, chlamydial antibody testing is limited by its inability to provide anatomical information with respect to the uterus or tubes and its lack of a potentially therapeutic effect [27]. Moreover, Johnson et al., compared serum chlamydial antibody titre with tubal status and pelvic findings in 1006 women undergoing laparoscopy for infertility. They observed a highly significant association between chlamydial antibody status and the likelihood of tubal damage. Women with positive titers were more likely to have pelvic adhesions than tubal occlusion unless titers were very high, when tubal damage was likely to be.
more severe. It was concluded that chlamydial antibody tests might be useful as a screening test for the likelihood of tubal damage in infertile women and might facilitate decisions on which women should proceed with further investigations without delay [28].

The duration of infertility in the chlamydia positive cases in our study was approximately 2-8 yr, which corresponds well with other reports. All the infected infertile women were asymptomatic. This highlights clinical inadequacy in diagnosing C. trachomatis. This finding supported by Puolakkainen et al., who found a high percentage (47.8%) of women positive for C. trachomatis had tubal infertility. Majority of these women had no symptoms of C. trachomatis [29].

In our study, the age range of patients with positive C. trachomatis IgG antibodies was 20-30 years were this finding in agreement with ElQouqa et al., who found that the age groups 25-31 years and 32-38 years were those with the highest rate of infection [30]. This finding is inconsistent with those of other studies that have shown a decline in the prevalence rate after 25 years of age [31].

In the present work C. trachomatis plasmid DNA by PCR in fluid from pouch of Douglas, was detected in 5 patients of the studied patient group and negative in control group. This is consistent with Wilkowska-Trojniel et al., who reported that, C. trachomatis infection was detected by PCR in 8.7% of patients with tubal factor infertility in Poland [26]. In another study that was performed to determine the prevalence of C. trachomatis infection among women attending gynecology and infertility centers in Gaza, using EIA and PCR assays for the detection of C. trachomatis in endocervical swab specimens, the overall prevalence of C. trachomatis in the study population was 20.2% [30]. Contrary to our finding, a study in Iran, did not find any infection by Chlamydia in the group of infertile women, who were evaluated by either ELISA or PCR as routine protocols for the investigation before assisted reproduction technology procedures [32]. The wide variation of Chlamydia prevalence in different studies could be due to several factors, such as study population (i.e. selection of high-risk groups), hygiene levels, socioeconomic status, and different techniques employed. Our findings show that specific C. trachomatis IgG antibodies are more frequently detected as compared to chlamydial infection by (PCR), that is mainly connected with the fact that specific anti-chlamydial antibodies, especially IgG, are usually detected in cases of chronic process [33]. Moreover, Wilkowska-Trojniel et al., suggested that C. trachomatis IgG is a highly predictive screening test for tubal infertility with similar diagnostic value to hysterosalpingography, and furthermore serology is a noninvasive test. Infertile couple should be offered, among all the diagnostic procedures, also those detecting sexually transmitted infections, including first of all C. trachomatis. Nevertheless, negative C. trachomatis study result does not exclude the possibility of tubal occlusion and many authors indicate that diagnostic laparoscopy should be performed in these cases if needed [26].

From this study it is concluded that, genital tuberculosis continue to be an important cause of infertility in Egypt and the diagnosis being often made from endometrial biopsy results, laparoscopy, or hysterosalpingography. PCR for mpt64 gene of M. tuberculosis plays an important role in diagnosis of genital tuberculosis with results comparable with laparoscopy and histopathology.

Detection of C. trachomatis IgG antibodies by ELISA can play a significant role in detection of C. trachomatis in infertile women. Screening of infertile women for C. trachomatis is recommended in the first year of infertility, so that early therapeutic intervention can be instituted to allow women to conceive naturally.

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