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Original Research Article

Prevalence of female genital tract tuberculosis in suspected cases attending Gynecology OPD at tertiary centre by various diagnostic methods and comparative analysis

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ABSTRACT

Background: The genital tract tuberculosis is one of the most common causes of tubal factor infertility. This study was conducted to compare the results of different diagnostic methods used in screening for female genital tuberculosis in suspected cases attending Gynecology OPD at RMC, Ajmer.

Methods: This prospective study was conducted in department of obstetrics and gynecology, J. L. N. Medical College, Ajmer, Rajasthan, for studying incidence of genital tuberculosis by various diagnostic methods (viz. AFB smear examination, AFB Lowenstein Jensen culture method, TB-PCR and CBNAAT).

Results: Prevalence of genital TB was 5.5% in study population of 200 selected women meeting the inclusion criteria. 72% women were in between 20-30 years age group. Oligomenorrhoea (24%) was found to be significant symptom with P value of <0.05. TBPCR and CBNAAT were found to be statistically significant with P value of <0.001 for diagnosing FGTTB.

Conclusions: We concluded that genital tuberculosis is paucibacillary disease, TBPCR and CBNAAT appears to be rapid and sensitive diagnostic modality.

Keywords: AFB, CBNAAT, Culture, FGTTB, Genital tuberculosis, TBPCR

INTRODUCTION

Tuberculosis (TB) is one of the oldest diseases as old as human civilization.¹ It is a chronic infectious disease and the morbidity associated with this condition has major health implications. In 1744, the first reported case of female genital tuberculosis was described by Morgagni – an anatomist. By polymerase chain reaction (PCR) analysis mycobacterial DNA has been detected in the genital areas of Andean Mummies date from A.D. 140-1200.² The manifestations of tuberculosis in gynaecology were published as a monograph by Hagar in 1886.³

Studies have shown that genital infection is seen in 12-24% of cases with pulmonary tuberculosis.⁴

The genital tract tuberculosis is one of the most common causes of tubal factor infertility. Female genital tuberculosis (FGTB) is an important cause of significant morbidity, short- and long-term sequelae especially infertility whose incidence varies from 3 to 16% cases in India. Mycobacterium tuberculosis is the etiological agent for tuberculosis. The fallopian tubes are involved in 90-100% cases, endometrium is involved in 50-80% cases, ovaries are involved in 20-30% cases, and cervix is involved in 5-15% cases of genital TB.

Tuberculosis of vagina and vulva is rare (1-2%). The diagnosis is made by detection of acid-fast bacilli on microscopy or culture on endometrial biopsy or on histopathological detection of epithelioid granuloma on biopsy. Polymerase chain reaction may be false positive and alone is not sufficient to make the diagnosis. Laparoscopy and hysteroscopy can diagnose genital tuberculosis by various findings.

The incidence of FGTB is 1-19% in various parts of India. In infertility patients, incidence of FGTB varies from 3 to 16% in India with higher incidence being from apex institutes like All India Institute of Medical Sciences (AIIMS), New Delhi, where prevalence of FGTB in women of infertility was 26% and incidence of infertility in FGTB to be 42.5%, which may be due to referral of difficult and intractable cases to this apex hospital from all over India, especially from states like Bihar where prevalence of TB is very high.⁵

METHODS

This prospective study was conducted at the department of obstetrics and Gynaecology, J. L. N. (Jawahar Lal Nehru) Medical College, Ajmer, Rajasthan from October 2017 to September 2018, in 200 selected women attending gynaecology OPD at RMC (Rajkiya Mahila Chikitsalaya) AJMER for studying incidence of genital tuberculosis by various diagnostic methods and to compare results of these non-invasive methods viz. AFB smear examination, conventional AFB Lowenstein Jensen culture method, TB-PCR and CBNAAT (Cartridge based nucleic acid amplification test) for diagnosing female genital tuberculosis.

The study was presented to Institutional Ethical Committee (IEC) for ethical clearance, after getting clearance from IEC the study was started. The data were collected from the women using a semi-structured questionnaire. The socio-demographic factors will be taken by direct interview. After enrollment, detailed history was obtained regarding demographic details, gynecological symptoms, and past history of TB. Details of previous investigations and treatment were noted to rule out other causes of infertility. Explanation of procedure to all women participating in study was done. Consent from every woman participating in this study was taken. After physical and gynecological examination all subjects underwent the following investigations:

Blood tests: CBC (Complete Blood Count)/ PBF (peripheral blood Film)/ ESR (Erythrocyte Sedimentation Rate)

- Mantoux (Tuberculin) test (poor sensitivity and specificity)
- Chest X-ray- PA view
- USG - PELVIS and TVS (Transvaginal sonography)
- HIV I AND II tests and VDRL

- Endometrial biopsy, curettage or aspirate- Premenstrual Endometrium sample was taken (with help of pipelle's sampling curette) for CBNAAT, AFB smear, AFB Culture
- Histopathology examination (HPE) (demonstration of epithelioid granuloma)
- Mycobacterial AFB smear and culture using Lowenstein-Jensen (LJ) medium
- Molecular methods (CBNAAT/ TBPCR)-
 - a. TBPCR (Polymerase Chain Reaction) - For TBPCR premenstrual endometrial biopsy sample taken in Normal Saline and was sent. PCR test was carried out using IS6110 and TRC4 probes. The test involved processing of samples, amplification and isolation of DNA. Demerit of TBPCR is that it's costly it was done only in affordable and feasible women.
 - b. CBNAAT9 (Cartridge Based Nucleic Acid Amplification Test/ Gene Xpert)- For CBNAAT (Cartridge Based Nucleic Acid Amplification Test/ Gene Xpert)
- Diagnostic laparoscopy: Laparoscopy criteria were grouped as either suggestive (type 1 or abnormal) or non-suggestive (type 2 normal) of GTB.

Inclusion criteria

The 200 selected suspected cases were of viz

- Menstrual irregularities- oligomenorrhoea, hypomenorrhoea, amenorrhoea, menorrhagia, dysmenorrhoea, metrorrhagia. After ruling out of other causes by routine investigations, usg and various serum hormonal assays viz. S.TSH/ S.prolactin/ FSH/ LH/ DHEA
- Infertility - unexplained infertility. Women in whom infertility was due to male factors, ovulation disorders, endometriosis, sexual dysfunction were excluded from study
- Chronic pelvic pain
- Chronic pelvic inflammatory disease resistant to standard antibiotic treatment according to CDC 2015 guideline for PID.⁶⁻⁸

Exclusion criteria

- Immuno-compromised (HIV) women
- Women suffering from other known granulomatous diseases like chronic diseases, leprosy, filariasis, silicosis
- Patient already treated for TB or extra-pulmonary TB
- Known or suspected malignancy
- Acute PID
- Women on steroid treatment
- Menstrual irregularities due to hormonal irregularities.

Women who were positive for mycobacterium tuberculosis by AFB smear, AFB culture, CBNAAT and TBPCR were treated with standard anti tuberculosis treatment (ATT) in category 1 under RNTCP and were followed up.

Statistical analysis

The data was coded and entered into Microsoft Excel spreadsheet. Analysis was done using SPSS version 20 (IBM SPSS Statistics Inc., Chicago, Illinois, USA) Windows software program. Descriptive statistics included computation of percentages, means and standard deviations. Chi-square test used for qualitative data whenever two or more than two groups were used to compare. Level of significance was set at $P \leq 0.05$.

RESULTS

In present study the mean age in this study was 27.91 ± 5.88 years and maximum 144 (72%) women in between 20-30 years age group, while 52 (26%) were in age group 30-40 years (Table 1). 74 (37%) were having normal menstrual cycle, Oligomenorrhoea was found to be statistically significant symptom in women with genital tuberculosis with P value of <0.05 .

68 (34%) women complained of white discharge per vagina resistant to treatment given according to CDC guidelines for >6 months and 132 (66%) women were not having discharge per-vaginal. 78.5% (157) women presented with complaint of infertility. 141 (89.8%) women out of 157 were of primary infertility and 16 (10.2%) were of secondary infertility.

Table 1: Distribution according to menstrual irregularities.

| Menstrual irregularity | Case No. | Culture | CBNAAT | TBPCR | P value |
|------------------------|------------|----------|-------------|----------|-------------|
| | | | FGTB | | |
| Normal cycle | 74 (37%) | 1 | 2 | 2 | > 0.05 |
| Oligomenorrhoea | 48 (24%) | 3 | 7 | 2 | <0.05 (S) |
| Menorrhagia | 26 (13%) | 0 | 0 | 0 | > 0.05 |
| Dysmenorrhoea | 27 (13.5%) | 0 | 0 | 0 | > 0.05 |
| Hypomenorrhoea | 25 (12.5%) | 0 | 1 | 0 | > 0.05 |
| Total | 200 | 4 | 10 | 4 | |
| P value | | > 0.05 | <0.05 (S) | > 0.05 | |

Test applied chi square test

Table 2: Relationship between diagnostic laparoscopy findings, FGTTB, AFB culture, TB-PCR and CBNAAT.

| Diagnostic laparoscopy findings | Total no of cases | AFB Culture | CBNAAT | TBPCR |
|---------------------------------|-------------------|-------------|----------|----------|
| | | | FGTTB | |
| Abnormal | 27 (39.7%) | 2 | 6 | 4 |
| Normal | 41 (60.3%) | 0 | 0 | 0 |
| Total | 68 | 2 | 6 | 4 |
| P value | | 0.08 | 0.001(S) | 0.001(S) |

Test applied Chi Square test

A total of 68 women went under diagnostic laparoscopy, out of them 41 (60.3%) were having normal findings and bilateral dye spillage was seen and 27 (39.7%) women were having abnormal findings suggestive of genital tuberculosis viz. Delayed dye spillage 6 (8.8%), hydrosalpinx present 7 (10.3%), beaded appearance of fallopian tubes 5 (7.3%), granulomas 1 (1.4%), pelvic congestion 1 (1.4%), pelvic adhesions 5 (7.3%) and bilateral dye spillage absent 15 (22%) and unilateral dye spillage seen 10 (14.7%). In 27 (39.7%) women with abnormal findings 2 were found positive for FGTTB by AFB culture, 6 found positive by CBNAAT and 4 found positive for FGTTB by TBPCR as shown in Table 2 and 3.

In TST test 77 (37.5%) women were found reactive and 123 (62.5%) were found non reactive. 162 (81%) women were having normal ESR and 38 (19%) were having increased ESR. Out of 38, 9 women were found positive for FGTTB by CBNAAT and 3 were found by TBPCR and 4 were found positive by AFB culture. Sensitivity of CBNAAT test was found to be 75%, specificity 96.42%, positivity rate of 5%, positive predictive value of 30% and negative predictive value of 99.42% and results were highly significant with P value of <0.001 . Sensitivity of TBPCR test was found to be 50%, specificity of 100%, positivity rate of 2.78%, with a P value of <0.001 . As PCR is a costly investigation, only 144 women underwent TBPCR. In present study prevalence of FGTTB

was found to be 5.5% when all mentioned tests were included.

Table 3: Diagnostic laparoscopy findings in infertility women suspected for FGTTB.

| Diagnostic laparoscopy findings in women suspected for FGTTB | Total no. of cases n = 68 |
|--|---------------------------|
| Bilateral dye spillage seen (normal findings) | 41 (60.3%) |
| Bilateral Dye spillage absent | 15 (22%) |
| Delayed dye spillage | 6 (8.8%) |
| Hydrosalpinx present | 7 (10.3%) |
| Beaded appearance | 5 (7.3%) |
| Granulations | 1 (1.4%) |
| Adhesions | 5 (7.3%) |
| Unilateral dye spillage | 10 (14.7%) |
| Pelvic congestion | 1 (1.4%) |

DISCUSSION

In Present study we found the prevalence of genital TB 5.5% (provided all investigations used in study are included) in study population of 200 selected women meeting the inclusion criteria. However, prevalence of

FGTTB were found by Shaheen et al, Pakistan, Nadgouda et al, India, Khanna and Agarwal, India, Abdelrub et al, Yemen, Pratibha et al, India, Thangappah et al, India 2.43%, 10%, 26%, 6.9%, 9.3% and 39% respectively as shown in Table 4.¹¹⁻¹⁶ In this study the mean age was 27.91±5.88 years and maximum 144 (72%) women in between 20-30 years age group however in Thanagappah et al¹⁶ 2018 reported in their study mean age of presentation was 27.35 years and Arpitha VJ et al, reported mean age of patients were 28.89±4.23 years.¹⁴ Oligomenorrhoea was found to be statistically significant symptom however, Radha BP Thangappah et al in reported menstrual disturbances such as secondary amenorrhoea, oligomenorrhoea, and menorrhagia were seen in 27.7% women and Arpitha VJ et al in reported that majority of women had a normal menstrual cycle 76.8% and hypomenorrhoea was the most common menstrual abnormality in 8.7% women followed by oligomenorrhoea (5.8%).^{14,16} 68 (34%) women complained of white discharge per vagina resistant to treatment however, Thangappah et al reported that vaginal discharge not responding to treatment for more than two years was seen in 5 (3%) cases out of 161 and Jindal UN et al reported that 6 (3%) patients out of 197 patients presented with white vaginal discharge.^{16,17}

Table 4: Studies on the prevalence of genital tuberculosis reported in literature.

| Author, country (year) | Study design | Number of participants | Prevalence of genital TB (%) |
|---|-----------------|------------------------|---|
| Tripathy and Tripathy, India ⁴ | Prospective | 91 | 3% (overall) 41 (in cases with tubal infertility) |
| Jindal, India ¹⁷ | Retrospective | 150 | 48% |
| Shaheen et al, Pakistan ¹¹ | Prospective | 534 | 2.43% |
| Singh et al, India ¹⁰ | Retrospective | 140 | 48.5% (in cases with tubal infertility) |
| Nadgouda et al, India ¹² | Prospective | 170 | 5.8% |
| Khanna and agarwal, India ¹³ | Cross sectional | 100 | 26% |
| Abdelrub et al, Yemen ¹⁴ | Prospective | 682 | 6.9% (overall) 31 (in cases with tubal infertility) |
| Pratibha et al, India ¹⁵ | Prospective | 160 | 9.3% |
| Thangappah et al, India ¹⁶ | Prospective | 153 | 25.4% |
| Present study, India | Prospective | 200 | 5.5% |

There were 77 (37.5%) Women were found reactive by TST Test however, Thangappah et al in reported that a positive TST test or Mantoux test with an induration of >10mm was seen in 37 (21.4%) cases out 161 total cases and In 29 of the 37 cases with positive tuberculin test, other diagnostic parameters were also positive.¹⁶ Therefore, TST test has been included as one of the diagnostic criteria to suspect Genital tuberculosis. Thangappah et al, in 2018 reported that 27 patients (15.6%) out 161 patients total were having abnormal ESR on investigating.¹⁶ Among the 27 cases with elevated

ESR, in 21 of them other diagnostic parameters were also positive. Thangappah et al, reported that 93 (53.8%) women were having normal findings on laparoscopy and 20 (11.6%) women were having definite evidence of TB findings such as granulomas, caseation, calcification and tubercles, in another 18 (10.4%) cases there was probable evidence of tuberculosis with hydrosalpinx dilated retort shaped tubes, tubes covered with white plaques and exudates, dense adhesions and loculated ascities and in remaining 42, there was suspicion of tuberculosis because of minimal adhesions, and cornual block.¹⁶

Table 5: Comparative parameters of present study with previous studies.

| Comparative parameters | Present study | Thangappah et al ¹⁶ | Jindal UN et al ¹⁷ | Arpitha VJ et al ¹⁸ | Mittal P et al ¹⁵ | Kandi et al ²⁰ |
|--|--|---|---|---|------------------------------|---------------------------|
| Age in year(range), mean, SD | 72% in between 20-30,27.91, 5.88 | 20-37, mean 27.35 | Mean 32.4, SD 8.3 | 28.89,4.23 | | |
| Menstrual irregularity | 37% normal, 24% oligomenorrhea, 13.5% dysmenorrhea, 13% menorrhagia, 12.5% hypomenorrhoea, P value was significant for oligomenorrhea p value<0.05 | Menstrual disturbances in 27.7%, (secondary amenorrhea, oligomenorrhea, menorrhagia | | 76.8% normal, 24.2% menstrual irregularity, hypomenorrhea most common in 8.7%, 5.8% oligomenorrhea | | |
| Systemic signs and symptoms (fever with night sweat, loss of appetite, weight loss | Present in 42%, Fever with night sweats 6.5%, loss of appetite 13%, weight loss in 24.5% P value was <0.001 by chi square test and significant | | Fever in 4.1% | | | |
| Vaginal discharge | 34% had white discharge | Persistent white discharge for more than 2yrs is seen in 3% cases. | (3%) patients presented with white discharge p/v. | | | |
| Infertility | Infertility was present in 78.5%, 89.8% have primary infertility and 10.2% secondary infertility 90% of female genital tract tuberculosis patients were from primary infertility | 93.1% patients had primary infertility, 56.6% had no symptoms other than infertility | 70% had primary infertility and 30% had secondary infertility | 65.2% had primary infertility | | |
| Diagnostic laparoscopy | Out of 68 patients in whom diagnostic laparoscopy done 39.7% have abnormal findings (Delayed dye spillage 6 (8.8%), hydrosalpinx present 7 (10.3%), beaded appearance of fallopian tubes 5 (7.3%), granulomas or granulations 1 (1.4%), pelvic congestion 1 (1.4%), pelvic adhesions 5 (7.3%) and bilateral dye spillage absent 15 (22%) and unilateral dye spillage seen 10 (14.7%), Chi square test show positive relation in between CBNAAT, TBPCR. P value<0.001 | 53.8% patients were having normal finding, 11.6% were having definite evidence of TB findings such as granulomas, caseation, calcification and tubercles. (10.4%), there was probable evidence of tuberculosis with hydrosalpinx dilated retort shaped tubes, tubes covered with white plaques and exudates, dense adhesions and loculated ascities. In (24.2%), there was suspicion of tuberculosis because of minimal adhesions, and cornual block. | | 40% normal, suprahepatic adhesions in (18.8%), pelvic adhesions in (34.8%), pelvic congestion in (5.8%), peritubal adhesions in (27.5%), omental and bowel adhesions in (15.9%) and TO-mass in (5.7%). On chromopertubation, delayed spillage of the dye was seen in (26%), cornual block in (21.7%), sacculations in (20.2%), hydrosalpinx in (15.9%), shortened tubes in (4.3%), rigid tubes in (4.3%) and fimbrial phimosis in (8.7%). Other incidental finding was endometrioma (in the form of endometriotic spots) was seen in (13.1%). | | |
| Per speculum cervix examination Unhealthy cervix (erosion, hypertrophy, irregular margins) | 30% had unhealthy cervix and chi-square test show positive relation in between unhealthy cervix and AFB Culture (3/4) and CBNAAT (6/10) P value<0.05 | | Cervical erosion was present in (3.4%) | | | |

| Comparative parameters | Present study | Thangappah et al ¹⁶ | Jindal UN et al ¹⁷ | Arpitha VJ et al ¹⁸ | Mittal P et al ¹⁵ | Kandi et al ²⁰ |
|--|---|---|--|---|---|---|
| Fornix/adnexal tenderness | Fornix tenderness present in 8% patients and AFB culture, CBNAAT, and TBPCR were positive in 1, 3, 2 patients and chi square test show P value <0.05 for CBNAAT and TBPCR. | | Reported (5.04%) patients presented with adnexal / fornicial tenderness. | | | |
| MT test | (37.5%) patients were found reactive, AFB culture, CBNAAT, and TBPCR were positive in 4, 7, 2 patients respectively and chi square test show significant p value <0.05 for CBNAAT and AFB culture | Reported that a positive TST test or Mantoux test with an induration of >10mm was seen in 37 (21.4%) cases. In 29 of the 37 cases with positive tuberculin test, other diagnostic parameters were also positive | | | | |
| ESR | (19%) patients were having increased ESR (Abnormal). 9 patients were positive for CBNAAT, 3 for TBPCR, AND 4 were for AFB culture and chi square test was statically significant p value <0.001 | 27 patients (15.6%) out 161 patients total were having abnormal ESR on investigating. Among the 27 cases with elevated ESR, in 21 of them other diagnostic parameters were also positive. | | | | |
| Incidence of female genital tract tuberculosis | Out of 200 total patients positive for TB was 11, 1 positive for smear, 10 by CBNAAT, 4 by AFB culture, and 4 by TBPCR with incidence of 5.5% | 4.6% of the endometrial samples were positive for AFB smear, AFB culture was positive in 3.5% cases. PCR was positive in 28.1% of endometrial samples. | | 69 patients positive for endometrial TBPCR none of the cases shown positive results on TB BACTEC (culture) or by ZN staining. | Out of 160 genital samples, a total of 9.3% (15/160) were positive, 53.3% (8/15) were detected by endometrial PCR, 33.3% (5/15) detected by BACTEC, 33.3% (5/15) cases detected by AFB culture and 6.66% (1/15) cases detected by direct AFB smear examination. | Reported that for extrapulmonary samples the sensitivity and specificity for CBNAAT samples were 85.7% and 93.5% respectively |

Arpitha VJ et al reported that 69 women went under laparoscopy and 40% had no evidence of genital tuberculosis on laparoscopy and in rest 60% women on laparoscopy had: suprahepatic adhesions in 13 (18.8%), pelvic adhesions in 24 (34.8%), pelvic congestion in 4 (5.8%), peritubal adhesions in 19 (27.5%), omental and bowel adhesions in 11 (15.9%) and TO-mass in 4 (5.7%),

on chromopertubation, delayed spillage of the dye was seen in 18 (26%), cornual block in 15 (21.7%), sacculations in 13 (20.2%), hydrosalpinx in 11 (15.9%), shortened tubes in 3 (4.3%), rigid tubes in 3 (4.3%) and fimbrial phimosis in 6 (8.7%), Other incidental finding endometrioma in 9 (13.1%).¹⁴ In present study prevalence

of FG TB was found to be 5.5% when all mentioned tests were included.

Out of 200 samples, a total of 11 (5.5%) were positive, 1/11 (9.09%) were detected by direct AFB smear examination, 10/11 (90.9%) were detected by CBNAAT, 4/11 (36.36%) were detected by AFB culture and 4/11 (36.36%) were detected by TBPCR. Out of 200 women 144 (72%) women were tested by TBPCR, by TBPCR the incidence of Female genital tuberculosis was 2.78 %. Kandi et al, reported that for extra pulmonary samples the sensitivity and specificity for CBNAAT samples were 85.7% and 93.5% respectively.²⁰ Table 5 shows the comparative parameters of present study with previous studies.

CONCLUSION

We concluded that genital tuberculosis is paucibacillary disease, TBPCR and CBNAAT appears to be rapid and sensitive diagnostic modality. But both can neither confirm nor exclude genital tuberculosis. But by combining all tests TST test, ESR, Laparoscopy, AFB culture, AFB smear examination, TBPCR and CBNAAT can effectively confirm an early diagnosis of GTB.

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