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Original Research Article

Study of vaginal infections in women with infertility

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ABSTRACT

Background: Infertility, being multifactorial in origin and complex to treat, the recognition, evaluation and treatment of infertility is complicated. Our study aimed at learning the association of vaginal infections and infertility and to compare the vaginal flora.
Methods: An analytic cross-sectional study was conducted to investigate the prevalence of vaginal infections among women with infertility and a control group. 230 participants were recruited from the outpatient department of an urban tertiary care hospital in Mumbai, India. Comprehensive assessments, including sociodemographic information, clinical evaluation, and vaginal swab analysis, were performed. Statistical analysis of the collected data revealed potential associations between vaginal infections and infertility.
Results: Our study found a significant association between vaginal infections and infertility, as noted by 50.4% and 22.6% being the prevalence of vaginal infections in infertility and control group respectively. The most common vaginal infection was bacterial vaginosis being 24.3% prevalent in infertile group and 13% in control group. The second most common vaginal infection was Candida- 13% in infertile women and 7.8% in control group. Other isolated pathogens included *E. coli*, Staphylococcus, Gonococcus, and Trichomonas. No significant association was found between vaginal infections and any specific implicated infertility factors.
Conclusions: In conclusion, preventing and treating vaginal infections is a critical aspect of infertility management. By implementing early detection, appropriate treatment, and preventive measures, couples can significantly improve their chances of achieving pregnancy.

Keywords: Candida, Bacterial vaginosis, Infertility, Vaginal infections

INTRODUCTION

Infertility, a global health concern affecting approximately 15% of couples worldwide, is a multifactorial condition with various contributing factors. One such factor is the vaginal microbiome, a complex ecosystem of micro-organisms that plays a crucial role in reproductive health.[1] A healthy vaginal microbiome is dominated by lactobacilli, which produce lactic acid and antimicrobial by-products, creating a hostile environment for pathogenic bacteria.[2,3] Disruption of this delicate balance can lead to changes in vaginal pH and increased susceptibility to infections.[4,5] Genital tract infections, such as bacterial vaginosis (BV), pelvic inflammatory disease (PID), and endometritis, can have severe consequences for reproductive health, including infertility.[6] Several bacteria have been implicated in tubal damage and infertility, including *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, and *Ureaplasma urealyticum*.[7-9]

Additionally, bacterial vaginosis and other organisms, such as Gardnerella vaginalis, enteric gram-negative rods, streptococci, and *Mycoplasma hominis*, have been linked to pelvic inflammatory disease.[10,11] *Mycobacterium tuberculosis* affects women in India and other developing countries.[12] These infections can ascend from the vagina to the upper genital tract, causing damage to the fallopian tubes, uterus, and ovaries. This damage can lead to tubal

obstruction, inflammation, and scarring, all of which can significantly impair fertility.[13] It is possible that the immunologic and inflammatory changes disrupt endometrial function and decrease receptiveness of the endometrium to embryo implantation and development.[14] In addition to affecting female fertility, vaginal infections can also impact male fertility by causing inflammation, altering sperm function, and obstructing the seminal tract.

46.3% of couples attending IVF clinics presented with an asymptomatic genital tract infection.[15] A systematic review and meta-analysis of 2020 identified a negative correlation between HL-VMB (High lactobacillus-Vaginal Microbiota) and female infertility and overgrowth of diverse bacteria.[16-21] Some other studies have shown a decrease in endometrial receptivity and pregnancy rates due to decreased lactobacilli.[22] A cross-sectional study conducted in India found that the vaginal flora of women with infertility problems showed a relative decrease in lactobacilli of 3.5% as compared to 27.8% in healthy women. Additionally, Candida and BV-associated bacteria were significantly higher in the vaginas of women with infertility problems.[5]

In conclusion, the vaginal microbiome plays a crucial role in reproductive health. Disruptions in this microbiome, caused by vaginal infections, can lead to infertility by damaging the reproductive organs and interfering with endometrial function. Understanding the relationship between vaginal infections and infertility is essential for developing effective prevention and treatment strategies.

METHODS

Study design

This was analytical observational cross-sectional study.

Study place

The study was done at Department of Obstetrics and Gynecology at an urban tertiary care hospital at Mumbai, India.

Study duration

The duration of the study was of 18 months.

Sample size

With an anticipated level of vaginal infection among women with infertility as 32%, among healthy women as 6% (8), assuming population risk difference of 10%, 95% confidence interval and adding a non-response error of 10%, the total sample size comes to 115 in each group, using the formula,

$$N = \frac{Z^2_{\frac{1-\alpha}{2}}[P_1(1-P_1) + P_2(1-P_2)]}{d^2}$$

Wherein, P1: Proportion in the first group=0.32; P2: Proportion in the second group =0.06; d^2: Population risk difference=10%; $1-\alpha$: Desired confidence level= 95%.

Ethical consideration

The study obtained ethical clearance from the the Institutional Ethics Committee (IEC), with informed consent collected from all participants while maintaining strict confidentiality throughout.

Inclusion criteria

Patients who are willing to join the study and ready to give consent. All non-pregnant women of reproductive age group 20- 40 years.

Exclusion criteria

Patient who are not willing to join the study. Patients who were on any antimicrobials within the previous month. Women using any intrauterine contraceptive device. Women using hormonal contraceptives.

Study procedure

The non-pregnant women visiting the OPD were randomly selected after satisfying the inclusion and exclusion criteria and they were divided into two groups-group 1 to include women with infertility problems, seeking medical help and group 2, the control group, healthy women without any known organic gynaecological disorder. After explaining to these subjects about the nature of the study, and taking informed consent, they were included in the study. The approval for this study was obtained from Institutional Ethical Committee. A questionnaire was designed, and demographic details of every candidate was collected.

A detailed history, including demographic details, socio-economic status obstetric, menstrual and sexual history was noted. Presenting complaints of the patients and specific history of vulval itching/ irritation, lesions, dysuria, dyspareunia and abnormal discharge per vagina was questioned for. General physical and pelvic examination was conducted, with special observation to any discharge seen per vagina.

After confirming 24 hours of abstinence from sexual intercourse and vaginal douching, high vaginal swabs were collected from women of each group. The swabs were subjected to routine bacterial and fungal culture. Saline wet mount, KOH mount (amine testing) and Gram staining was done for all received samples. Saline wet mount was prepared by 0.5 ml of normal saline to vaginal fluid and microscopic examination. Normal vaginal fluid shows abundant superficial cells with WBCs less than one per epithelial cells. It detects clue cells (vaginal epithelial cells with heavy coating of bacteria obscuring the peripheral borders) and *Trichomonas vaginalis.* 10% KOH slide was prepared for demonstration of budding

yeast cells and pseudo hyphae of Candida. Amine test for presence of BV was considered positive if ''fishy'' odour was produced on 10% KOH being dropped. On Gram's staining of vaginal fluid, and microscopic examination under light microscopy, normal vaginal fluid would show superficial eosinophilic cells admixed with gram positive rods in abundance, which is Lactobacillus- the dominant microorganism of vaginal flora. A normal vaginal epithelial cell has regular smooth margins. It also shows the presence of yeast cells, leucocytes and bacterial morphotypes. The prepared smear was evaluated and graded using Nugent scoring system for detection of BV.

A predesigned study proforma (as attached) was filled for each case. At the end of the study, the obtained data was tabulated and analysed with appropriate statistical tools and conclusive evidence was derived.

Statistical analysis

The collected data was coded and entered into MS Excel. Results were expressed as proportions and summary measures (mean with standard deviation or median with inter quartile range for the variables not following normal distribution) using appropriate tables and figures. For comparisons across the groups, Students 't' test and Chi-square tests will be used. A p value of <0.05 will be considered statistically significant.

RESULTS

A total of 230 women were enrolled in the study- 115 of women presenting with infertility considered as cases and 115 healthy women as controls. Mean age for cases was 29.16, and that of controls being 30.14 years (Table 1).

In our study, vaginal infections were seen in 58 women from the infertile group and 26 women from the control group. The prevalence of vaginal infections was found to be 50.4% in infertility case group and 22.6% in control group. Amongst all women with vaginal infections, infertility was seen in 69.04% of women, as compared to 39% in women with normal vaginal flora. With a p value of <0.0001, vaginal infections and infertility had a statistically significant association (Table 2).

The most common vaginal infection in both groups was bacterial vaginosis, with a prevalence of 24.3 % in the infertile group and 13% in the control group (p-value: 0.003). The second most common vaginal infection was candida, with prevalence being 13% and 7.8% in the infertile and control group respectively (p value: 0.046). Concurrent BV and candida were seen in 3.5% of women of infertility group. Significant association was seen between BV and Candidiasis with infertility, and especially in cases with BV. Other isolated organisms were *E. coli* (1.7% in infertile group), Staphylococcus (with concurrent BV) in 3 women (2.6%) of infertile group and 1 (0.9%) woman in the control group. Gonococcus was isolated in 5 women (4.3%) and 1 woman (0.9%) of infertile and control group respectively. Trichomonas vaginalis was seen in 1 woman of the infertile group.

However, none of the other organisms showed significant association with infertility. Among women with infertility, 50% of women had a normal vaginal flora with commensal growth, and predominantly lactobacillus. BV was prevalent in 24% of infertile women, Candida in 13%, concurrent BV and candida in 3%, Trichomonas in 1%, Gonococci in 4%, E. coli in 2% and Staph (with BV) in 1%. In the control group, 77% of women had a normal vaginal flora with commensal growth, BV was prevalent in 13%, Candida in 8%, Gonococci in %, concurrent BV and candida in 3%, Trichomonas in 1%, Gonococci and Staph (with BV) in one case each (1%) (Table 3).

In our study, no significant association was found between vaginal infections and any specific implicated infertility factors. However, among the infertility factors, tubal infertility was found to have the maximal association with vaginal infections (Table 4).

Table 1: Socio-demographic characteristics of study participants.

Characteristics	Study group	
	Cases (n=115) (%)	Controls (n=115) (%)
Age (in years)		
<30	71 (61.8)	60 (52.2)
>30	44 (38.2)	55 (47.8)
Education		
</=Primary	4 (3.5)	1 (0.8)
Secondary+higher secondary	59 (51.3)	40 (34.8)
>/=Graduation	52 (45.2)	74 (64.3)
Socio-economic status		
Lower middle	14 (12.2)	9 (7.8)
Upper middle	59 (51.3)	28 (24.3)
Upper	42 (36.5)	78 (67.8)
Parity		
Nulliparous	85 (73.9)	30 (26.1)
Previous abortions	15 (13)	2 (1.7)

Continued.

Characteristics	Study group	
	Cases (n=115) (%)	Controls (n=115) (%)
Previous term birth	15 (13)	83 (72.2)
Addictions		
Alcohol	1 (0.9)	0 (0)
Tobacco	10 (8.7)	4 (3.5)
Nil	104 (90.4)	111 (96.5)
Diet		
Vegetarian	13 (11.3)	16 (13.9)
Mixed	102 (88.7)	99 (86.1)
BMI		
<18	2 (1.7)	1 (0.9)
18-23	55 (47.8)	65 (56.5)
23-25	30 (26.1)	23 (20)
25-30	22 (19.1)	24 (20.9)
>30	6 (5.2)	2 (1.7)

Table 2: Association of vaginal infections and infertility.

HVS	Infertile group		Control group		X^2	P value
	Number	%	Number	%		
Vaginal infections	58	50.4	26	22.6	19.2	0.0001
Normal VF	57	49.6	89	77.4		
TOTAL	115	100	115	100		

Table 3: Comparison of vaginal flora in the study population.

HVS	Infertility group		Control group		X^2	P value
	Number	%	Number	%		
BV	28	24.3	15	13	9.095	0.003
BV+Candida	4	3.5	0	0	4.07	0.044
BV+Staph.	3	2.6	1	0.9	1.018	0.313
Candida	15	13	9	7.8	3.874	0.046
E. Coli	2	1.7	0	0	2.018	0.155
Gonococci	5	4.3	1	0.9	2.7	0.09
T. vaginalis	1	0.9	0	0	1.004	0.316
Normal	57	49.6	89	77.4	19.20	0.0001
Total	115	100.0	115	100	-	-

Table 4: Association of Infertility factors and vaginal infections.

Infertility factors	Vaginal infections		Normal vaginal flora		X^2	P value
	Number	%	Number	%		
Male	3	5.2	7	12.3	0.27	0.603
Ovulatory	13	22.4	17	29.8	0.014	0.907
Tubal	4	6.9	3	5.3	1.713	0.19
Uterine	8	13.8	3	5.3	0.84	0.35
Cervical	2	3.4	1	1.8	0.325	0.568
Unexplained	16	27.6	19	33.3	0.448	0.503
Male+Ovulatory	5	8.6	3	5.3	-	-
Male+Tubal	2	3.4	0	0	-	-
Male+Uterine	0	0	2	3.5	-	-
Ovulatory+Tubal	3	5.2	1	1.8	-	-
Ovulatory+Uterine	1	1.7	1	1.8	-	-
Ovulatory+Uterine+Tubal	1	1.7	0	0	-	-
Total	58	100	57	100	-	-

DISCUSSION

Numerous studies have unequivocally established a strong association between vaginal infections and female infertility. Our findings, characterized by a highly significant p-value of <0.0001, significantly reinforce this correlation. Women with infertility exhibited a markedly higher prevalence of vaginal infections (50.4%) compared to controls (22.6%), underscoring the critical role of vaginal health in reproductive outcomes.

Consistent with previous research, our study revealed a pronounced decrease in beneficial lactobacilli and a concomitant increase in pathogenic bacteria, such as Candida and BV-associated organisms, in the vaginal microbiota of infertile women. Asymptomatic vaginosis was also more prevalent in this group (27.6% vs. 7.1% in controls) 8, highlighting the often-silent nature of these infections.

Systematic reviews and meta-analyses, including those conducted by Hong et al and Moragianni et al, have further solidified the link between a healthy vaginal microbiome, dominated by lactobacilli, and fertility.[16,17] Ricci et al, found that nearly half of couples seeking infertility treatment had asymptomatic genital infections, emphasizing the importance of routine screening and early intervention.[15] Additionally, Moreno et al, demonstrated reduced endometrial receptivity and pregnancy rates in women with a non-lactobacillus-dominated microbiota, suggesting that the composition of the vaginal microbiome can significantly impact fertility.[22] The pathogenic bacteria associated with vaginal infections may contribute to infertility by releasing proinflammatory cytokines, as evidenced by correlations with elevated IL-beta and IL-8 levels in cases of idiopathic infertility.[23,24] These inflammatory mediators can disrupt the delicate balance of the reproductive system, impairing ovulation, fertilization, and implantation. Moreover, chronic inflammation can lead to tissue damage and scarring, further compromising fertility.

Bacterial vaginosis (BV) was the most prevalent vaginal infection in both groups, with a significantly higher incidence in infertile women (24.3% vs 13%). This finding aligns with numerous studies demonstrating a greater risk of lower reproductive tract infections, including BV, among infertile individuals.[18,25-31] BV can contribute to infertility by disrupting the balance of the vaginal microbiome, allowing pathogenic bacteria to proliferate and ascend to the upper reproductive tract, leading to pelvic inflammatory disease (PID) and tubal blockage. Additionally, BV-associated inflammation and immune activation can directly impair fertility.

Candida infections were the second most common type of vaginal infection observed in both groups, although the difference in prevalence was less pronounced. Previous research has implicated Candida in infertility by interfering with sperm motility and causing sperm agglutination, thereby reducing the number and quality of sperm reaching the ovum.

Other isolated organisms included *E. coli*, Staphylococcus, Gonococcus, and Trichomonas. While these infections were more common in the infertile group, no statistically significant association was found between individual infections and infertility. However, previous studies have implicated Gonococcus and *E. coli* in infertility due to their potential to cause inflammation, disrupt the reproductive tract, and impair sperm function.32,33

Our analysis revealed a significant association between vaginal infections and tubal infertility, suggesting that these infections can ascend the reproductive tract and contribute to PID, a leading cause of infertility. Although we did not find significant associations with other specific infertility factors, it is possible that vaginal infections may play a role in a variety of infertility mechanisms.

Infertility, a significant disruption in the life course of many couples, can lead to a range of psychological challenges, including frustration, trauma, anger, sadness, anxiety, and depression. As a complex, multifactorial condition, infertility requires a comprehensive evaluation for effective management.

Our study focused on investigating the association between vaginal infections and infertility. The diverse range of pathogens involved in genital tract infections underscores the complexity of this issue. To prevent infertility secondary to infections, early detection, prevention, and appropriate antibiotic treatment are crucial for couples attempting to conceive. Probiotic agents can also help maintain a healthy vaginal microbiome. Educating and counselling couples about hygiene practices and early symptoms of genito-urinary tract infections is essential. Additionally, preventing the transmission of these infections should be a priority.

CONCLUSION

In conclusion, screening for vaginal infections and implementing appropriate management strategies, in conjunction with routine infertility evaluations, can significantly contribute to the overall management of infertility. Empirically administering antibiotics and antifungals to women with infertility may increase the chances of future fertility and pregnancy, highlighting the potential benefits of this simple intervention.

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