

Original Research Article

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A Retrospective Evaluation of Vaginitis in Women of Reproductive Age Group in a Tertiary Care Hospital in Solapur, India

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ABSTRACT

Vaginitis in women can be caused by aerobic bacteria, *Trichomonas vaginalis*, yeast or it can be bacterial vaginosis which can lead to various gynaecological and obstetrical complications. This is a retrospective study aimed at determining the prevalence of vaginitis due to various microorganisms. A total of 500 women in the reproductive age group attending the tertiary care hospital over the last 2 years were included in the study. The vaginal swabs of these patients were subjected to Gram's staining & wet mount to identify *T.vaginalis*, budding yeast and clue cells. All vaginal specimens were cultured for aerobic bacteria and fungi and identified according to standard microbiological techniques. Out of 500 women, 185 (37%) had vaginal infection. The most common identified vaginal infections were aerobic bacteria i.e. *E. coli* (36.8%), *Klebsiella* spp. (13.05%), *Acinetobacter* spp. (8.65%), *Staph. Aureus* (6.49%), *Citrobacter* spp. (4.32%), *Pseudomonas* spp (3.8%) and *Group B Streptococci* (3.24%) followed by Candidiasis (21.62%), Trichomoniasis (9.73%) and Bacterial vaginosis (6.49%). Gram negative bacteria revealed high level of sensitivity to Meropenem (93%), Amikacin (81.9%) and Ciprofloxacin (61.7%) and were found resistant to Ceftriaxone, Ceftazidime, Cotrimoxazole and Ampicillin. Thus the study reveals that vaginal infections by *E. coli* and *Klebsiella* followed by *Candida* spp., Trichomoniasis and bacterial vaginosis are common problems in women of reproductive age group. The treatment option varies according to the aetiology. Therefore, diagnosis of vaginal infections in women of reproductive age group should be done to prevent the injudicial use of chemotherapeutic agents to avoid further complications.

Keywords

Vaginitis, Bacteria
vaginosis, Aerobic
bacteria, Yeast,
Trichomonas vaginalis,
Reproductive age group,
Gram stain, Wet mount

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Introduction

The complaint of vaginal discharge is very common, particularly in south East Asia where about a quarter of all adult women report this complaint. (Chaudhary *et al.*, 2012) Symptomatic vaginal discharge in the women of reproductive age group is responsible for 5-10 million OPD visits per year throughout the world (Masand *et al.*, 2015). The prevalence

of vaginal discharge in India is estimated to be 30% (Masand *et al.*, 2015).

Abnormal vaginal discharge also predisposes to significant morbidity in the form of pelvic inflammatory diseases, infertility, endometriosis, cuff cellulitis, urethral syndrome, pregnancy loss, preterm labour, increase susceptibility to sexually transmitted infections (STI), including HIV and to be

associated with low birth weight and preterm birth (Masand *et al.*, 2015).

Females are more prone to urinary and vaginal infections because of the anatomical and functional proximity to the anal canal and due to the short urethra (Swamy *et al.*, 2015). The vagina could be infected by a variety of pathogens including bacteria, fungi, viruses, and parasites (Abdelaziz *et al.*, 2014). The causative organisms can be endogenous, iatrogenic or sexually transmitted (Swamy *et al.*, 2015). But, many women believe that such infections are normal and part of the female experience and do not seek care due to shame or lack of information (Swamy *et al.*, 2015).

These gynaecological disorders have substantial impact on female reproductive health, mental health, and ability to work and to perform routine physical activities (Chaudhary *et al.*, 2012). Vaginal complaints such as Bacterial Vaginosis, candidiasis, trichomoniasis, and *Chlamydia trachomatis* infections are common among women of reproductive age, with high incidences during pregnancy (Abdelaziz *et al.*, 2014)/

Bacterial vaginosis (BV), an alteration of vaginal flora involving a decrease in *Lactobacilli* and predominance of anaerobic bacteria, is among the most common cause of vaginal complaints for women of childbearing age (Bhat *et al.*, 2012). It has been shown to increase the risk of obstetric and gynaecologic complications such as preterm labour and delivery, chorioamnionitis, post-caesarean endometritis, post-abortion pelvic inflammatory disease, and cervicitis (Bhat *et al.*, 2012).

Trichomoniasis is known to be the most common, curable, sexually transmitted infection among sexually active women and may be associated with the acquisition and transmission of HIV which is caused by a

parasitic protozoan *Trichomonas vaginalis* (Deivam *et al.*, 2014; Mulu *et al.*, 2015). Negative outcomes of this infection are especially significant during pregnancy (Deivam *et al.*, 2014).

In the majority of women, a diagnosis of Vulvovaginal candidiasis is made at least once during their childbearing years. Among the many causes of vaginitis, VVC is the second most common after bacterial vaginosis and is diagnosed in up to 40% of women with vaginal complaints in the primary care setting (Bhat *et al.*, 2012)/ Approximately 75 % of adult women will have at least one episode of vaginitis by *candida* spp. during their life time (Mulu *et al.*, 2015). Unfortunately, about 40 – 50 % of women who had a first episode are likely to present a recurrence and 5 % may present a form of “recurring” characterized by at least three or more episodes of infection per year (Mulu *et al.*, 2015).

Aerobic vaginitis has been identified for a smaller proportion of women whose microbiota (*lactobacilli*) is replaced by facultative anaerobic or aerobic bacteria especially *Staph. aureus*, *Group B streptococci*, *E. coli* and *Klebsiella* spp (Bhat *et al.*, 2012)/ In pregnant women, *Group B Streptococci* (GBS) is a cause of cystitis, amnionitis, endometritis, and stillbirth (Bhat *et al.*, 2012). Intrapartum colonisation of the mother’s vagina with coliforms, enterococci is also associated with neonatal sepsis and meningitis in the newborn (Bhat *et al.*, 2012).

Bacterial vaginal infections are often least understood and empirical antifungal therapy for any vaginal infection without high vaginal swab culture is still in practice. Thus this study was carried out to determine the microbiological profile of symptomatic vaginal discharge in our hospital and antimicrobial susceptibility profiles of aerobic bacterial isolate and its utility in the

management of genital tract infection in women of reproductive age group.

Materials and Methods

This is a retrospective study done at a tertiary care centre in Solapur over a period of last two years. Two high vaginal swabs were collected from 500 women suspected of vaginitis taking all aseptic precautions.

Direct saline wet mount, Gram staining and bacteriological cultures were carried out for all specimens. One drop of a vaginal swab suspension with physiological saline were placed on a slide and covered with a cover slip. The wet film were examined under bright field microscopy at 40× objective for the presence of motile *Trichomonas vaginalis*, pseudo hyphae and/or budding yeast cells indicative of *Candida* spp., granulocytes and clue cells.

Gram stained smears were prepared from vaginal swab and examined under oil immersion at × 1000 magnification to look for clue cells, budding yeast cells, granulocytes and Gram negative diplococci.

Candida spp. was identified by the presence of yeast cells in wet mount as well as identification of Gram positive budding yeast cells per oil immersion field. *T. vaginalis* was identified by its pear shaped morphology with characteristic jerky motility on wet mount of vaginal specimen.

BV was identified by the presence of 'Clue cells' which are nothing but the vaginal epithelial cells with a granular surface and blurred margins (because of attached bacteria) on a physiological saline (0.85 %) wet mount and in Gram's stain can be seen as Gram negative cocco-bacilli studded vaginal epithelial cells instead of normally predominant Gram positive *Lactobacilli*.

All vaginal specimens were plated on to 5 % sheep blood agar, MacConkey agar to isolate aerobic bacteria. The inoculated media incubated at 37 °C aerobically for 24-72 hours. Identification of the cultured isolate was done by conventional phenotypic and biochemical methods.

Antimicrobial susceptibility testing was performed by Kirby-Bauer disc diffusion method. The following antimicrobial agents were employed: Amikacin (30µg), Meropenem (10µg), Ciprofloxacin (5 µg), cotrimoxazole (25 µg), Ampicillin (10µg), Ceftriaxone (30µg), Ceftazidime (30µg), Piperacillin (100µg), Erythromycin (15 µg) and clindamycin (2 µg), Linezolid (30µg) and Vancomycin (30µg). Antibiotic susceptibility was interpreted according to Clinical and Laboratory Standards Institute (CLSI, 2015) Guidelines.

Reference strain of *E. coli* ATCC 25922, *Staph. aureus* ATCC 25923 and *Pseudomonas aeruginosa* ATCC27853 were used for quality control for antimicrobial susceptibility test.

Results and Discussion

A total of 500 women in the reproductive age group (15-50 years) are included in the study in which 185 (37%) had vaginal infection among which 33 (31.42%) women were pregnant (Table 1).

Most of the females i.e. 157 (84.86%) of 185 women were infected with only one microorganism whereas rest of the women, 28 (15.13%) were infected with more than one microorganism.

The most common vaginal infections were due to aerobic bacteria i.e. 66.48% among which *E. coli* was the predominant microorganism causing vaginitis accounting for 36.8% cases

which is followed by infections with *Klebsiella* spp. (14.05%), *Acinetobacter* spp. (8.65%), *Staphylococcus aureus* (6.49%), *Citrobacter* spp. (4.32%), *Pseudomonas* spp. (3.78%) and very few women were infected with *Group B Streptococci* i.e. in 3.24% cases (Table 2).

Moreover, significant number of women was infected with *Candida* spp. (21.62%) followed by *Trichomonas vaginalis* (9.73%) and few women were reported to have Bacterial vaginosis (6.49%) (Table 2).

Among the analysed samples, co-infection occurred in 28 of 185 positive cases i.e. in 5.6% cases. Co-infection of bacteria/bacteria and bacteria/*candida* spp. were most commonly seen. This accounted for more than 96% of co-infection cases. Co-infections of bacteria & bacteria, bacteria & *candida* and bacteria & *T.vaginalis* were 19(67.8%), 8(28.57%) and 1(3.57%) respectively (Table 3).

The most common aerobic bacterial isolates were Gram negative bacteria which showed high level of susceptibility to Meropenem (92.8%), Amikacin (80.8%) and Ciprofloxacin (60.8%) whereas very less sensitivity was observed for Ceftriaxone (22.4%), Ceftazidime (22.45%) and Co-trimoxazole (14.4%) (Table 4).

Overall, very few Gram positive aerobic bacteria were isolated in the present study and it is observed that all those organisms were sensitive to Vancomycin (100%) and Linezolid (100%) whereas most of them were sensitive to Erythromycin (83.3%), Clindamycin (77.7%) and Ciprofloxacin (66.6%) (Table 5). This study was undertaken to assess the prevalence of microorganisms isolated from vaginal secretions in a tertiary care centre in Solapur, Maharashtra. It shows that prevalence of vaginitis is 37% which is

very similar to the studies done by Mathew *et al.*, and Bhargava *et al.*, (2016) whereas Mulu *et al.*, (2015) showed very less cases of vaginitis which may be due to better hygienic conditions of the patients.

Enterobacteriaceae were the most common cause of vaginitis in women of reproductive age group in the present study among which *E. coli* was the predominant bacteria responsible for it. Comparable results were obtained in the studies of Zarbo *et al.*, (2013), Mohamed *et al.*, (2015), Swamy *et al.*, (2015) and Kumar *et al.*, (2015) where they concluded *E. coli* to be the leading cause of vaginitis. This may be related to the poor hygiene of the patient as the Enterobacteriaceae are a part of normal flora of human gastrointestinal tract and close proximity to anal canal in females which contributes to the entry of gut flora into the genital tract causing vaginal infection.

5.6% of women in the present study were infected with more than one microorganism. Co-infection of bacteria/bacteria and bacteria/*candida* accounted for maximum co-infection cases which is similar to the findings of Karou *et al.*, (2012).

Vaginal candidiasis in the present study was responsible for 21.62% cases of vaginal infection which is comparable with the results of Abdelaziz *et al.*, (2014), Zarbo *et al.*, (2013) and Bhargava *et al.*, (2016). Pregnancy related emotional stress, hormonal changes and suppression of immune system steps up the *candida* species overgrowth and infection. This is evident in the study done by Nelson *et al.*, (2013) where he encountered 90.38% cases of vaginal candidiasis as he included only pregnant women in his study.

The present study encountered 9.73% women with *Trichomonas vaginalis* infection.

Table.1 Showing frequency of vaginitis in pregnant and non-pregnant women

Pregnancy status	Total (n=500)	Vaginitis(n=185)	Percentage
Pregnant	105	33	31.42%
Non-pregnant	395	152	38.48%

Table.2 Types of vaginitis among females of reproductive age group

Sr. No	Causative organism	Number	Percentage
1	<i>E.coli</i>	68	36.8%
2	<i>Klebsiella spp.</i>	26	14.05%
3	<i>Acinetobacter spp.</i>	16	8.65%
4	<i>Staph. aureus</i>	12	6.49%
5	<i>Citrobacter spp.</i>	08	4.32%
6	<i>Pseudomonas spp.</i>	07	3.78%
7	Group B Streptococci	06	3.24%
8	Candidiasis	40	21.62%
9	<i>Trichomoniasis</i>	18	9.73%
10	Bacterial vaginosis	12	6.49%

Table.3 Showing types of co-infection

Sr. No.	Type of vaginal infection	Total = 28	Percentage
1	<i>E.coli</i> + <i>Klebsiella</i>	13	7.02 %
2	<i>E.coli</i> + <i>Candidiasis</i>	8	4.32 %
3	<i>E.coli</i> + <i>Acinetobacter spp.</i>	3	1.62 %
4	<i>E.coli</i> + <i>Pseudomonas spp.</i>	2	1.08 %
5	<i>E.coli</i> + <i>Trichomoniasis</i>	1	0.54 %
6	<i>E.coli</i> + Group B <i>Streptococcus</i>	1	0.54 %

Table.4 Showing % sensitivity of Gram negative aerobic bacteria to various antibiotics

Antibiotics	<i>E.coli</i> (n=68)	<i>Klebsiella</i> (n=26)	<i>Acinetobacter</i> (n=16)	<i>Citrobacter</i> (n=8)	<i>Pseudomonas</i> (n=7)	Overall Sensitivity
	S	S	S	S	S	
Meropenem	100%	100%	43.7%	100%	100%	92.8%
Amikacin	88.2%	88.4%	31.2%	87.5%	85.7%	80.8%
Ciprofloxacin	75%	23.5%	18.7%	50%	28.5%	60.8%
Ceftriaxone	10.2%	23.07%	37.5%	37.5%	85.7%	22.4%
Ceftazidime	8.82%	26.9%	37.5%	37.5%	85.7%	22.4%
Cotrimoxazole	8.82%	30.7%	12.5%	12.5%	-	14.4%
Ampicillin	11.7%	-	12.5%	25%	-	-
Piperacillin	-	-	-	-	100%	-

Table.5 Showing % sensitivity of Gram positive aerobic bacteria to various antibiotics

	<i>Staph. aureus</i> (n=12)	Group B <i>Streptococcus</i> (n=6)	Overall Sensitivity
	S	S	
Erythromycin	75%	100%	83.3%
Ciprofloxacin	66.6%	66.6%	66.6%
Clindamycin	75%	83.3%	77.7%
Linezolid	100%	100%	100%
Cotrimoxazole	66.6%	-	-
Ampicillin	91.6%	100%	94.4%
Vancomycin	100%	100%	100%

This can be correlated with studies done by Deivam *et al.*, (2014), Masand *et al.*, (2015) and Zarbo *et al.*, (2013). Bacterial vaginosis was responsible for 6.49% of vaginitis cases which is coherent with the findings of Zarbo *et al.*, (2013) and Mohamed *et al.*, (2015).

Susceptibility testing of aerobic bacteria in the present study reveals that Meropenem, Amikacin & Ciprofloxacin are the antibiotics of choice which act against the Gram negative aerobic bacteria encountered in most of the vaginal infections. These results can be compared with study of Swamy *et al.*, (2015) this study reveals that vaginitis is caused more frequently by aerobic bacteria than candida. Hence the practice of empirical antifungal therapy without investigating high vaginal swab needs to be revised to prevent its unnecessary use.

Vaginal infections caused by aerobic bacteria, *Candida* spp., Trichomoniasis and bacterial vaginosis are common problems in women of reproductive age group. Detection of vaginal infection during pregnancy by *Group B Streptococci*, *E. coli* and coliforms in pregnant women is of significance as it can get transmitted to the neonate during delivery & can cause sepsis. The management of vaginal discharge is dependent on the correct microbiological diagnosis of pathogen which can help clinicians to institute appropriate

antimicrobial therapy which will prevent the obstetrical & gynaecological sequel of vaginitis. And this in turn will help to prevent the injudicial use of antibiotics and development of antibiotic resistance.

Moreover, Ciprofloxacin, Amikacin and Meropenem are the recommended drugs for empirical therapy for Gram negative organisms. For Gram positive organisms, Ampicillin, Erythromycin and Clindamycin are the recommended drugs.

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