

Aerobic Bacterial Pathogens causing Vaginitis in Patients Attending A Tertiary Care Hospital and their Antibiotic Susceptibility Pattern

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Abstract

Aerobic vaginitis (AV) is a condition in which the normal vaginal inhabitants are replaced by aerobic bacterial pathogens, triggering a vaginal immune response. AV should be treated promptly. The aim of the present study is to analyse the prevalence of AV in patients with clinical symptoms of vaginitis. High vaginal swabs were collected from 156 women with clinical suspicion of vaginitis, over a period of 1 year. The swabs were subjected to Gram stain and bacterial culture under aerobic conditions. The organisms were identified using standard microbiological methods and antibiotic susceptibility testing was performed for the bacterial isolates as per CLSI guidelines. Out of the 156 samples processed aerobically, 33 showed bacterial growth in culture. The common bacteria isolated in the study were *Staphylococcus aureus* (24.2%), β -hemolytic *Streptococcus* (21.2%), *Klebsiella* species (21.2%), *Escherichia coli* (15.2%). Antibiotics like β -lactams/ β -lactamase inhibitor combinations, Linezolid were effective against Gram positive bacteria. Gram negative bacteria were found to be more susceptible to Piperacillin Tazobactam, imipenem, meropenem, etc. The study emphasises the need for complete investigation of aerobic vaginal pathogens in patients with clinical symptoms of vaginitis.

Keywords: Aerobic vaginitis, *Staphylococcus aureus*, *Klebsiella*, antibiotics.

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(Received: 28 March 2019; accepted: 02 May 2019)

Citation: Lakshmi Krishnasamy, Chitralekha Saikumar and Govindasamy Kumaramanickavel, Aerobic Bacterial Pathogens Causing Vaginitis in Patients Attending A Tertiary Care Hospital and their Antibiotic Susceptibility Pattern, *J Pure Appl Microbiol.*, 2019; **13**(2):1169-1174. doi: 10.22207/JPAM.13.2.56

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INTRODUCTION

Vagina is equipped with normal physiological mechanisms to prevent the entry of pathogenic organisms¹. An intricate balance of microorganisms like lactobacillus maintain the physiological mechanisms of vagina. Factors influencing the growth of pathogenic organisms in vagina are alterations in pH, vascularity, glycogen content, hormonal status, etc.

Vaginitis is a clinical condition in which the patient complaints of abnormal vaginal discharge, vaginal irritation, itching, etc². Vaginitis is one of the frequent complaints of women visiting the gynaecology Out patients department³. Abnormal vaginal discharge in a patient could be due to infectious causes such as aerobic vaginitis, bacterial vaginosis (BV), trichomonal vaginitis (TV) and vulvovaginal candidiasis (VVC)²

In India, the prevalence of vaginal discharge is estimated to be 30%⁴. No definitive diagnosis could be made in many of the patients with clinical symptoms of abnormal vaginal discharge.² Improper diagnosis of aerobic vaginitis may lead to treatment failure and recurrence of infections. They may even lead to severe complications in pregnancy like miscarriage, chorioamnionitis, premature rupture of membranes and preterm delivery⁵⁻⁷. Therefore, there is a great need for correct diagnosis of the condition to initiate a proper therapy. Bacterial vaginal infections are least understood and often the patients with vaginitis are started on empirical antifungal drugs without proper investigations. Appropriate diagnosis and management of the cases would help in reducing the recurrence of the infections as well as decreasing the emergence of drug resistance among the pathogens. Thus, the main aim of the study is to evaluate the aerobic bacterial flora in patients with clinical symptoms of vaginitis and study their antibiotic susceptibility profile.

MATERIALS AND METHODS

The study was conducted in a tertiary care hospital over a period of 1 year from January 2018 to December 2018. The study has been approved by the institutional ethical committee. Hundred and fifty six patients with clinical symptoms suggestive of vaginal infections were enrolled in

the study. Their age ranged from 18 to 65 years. Patients with Bacterial vaginosis, trichomonal vaginitis, candidiasis were excluded from the study. A detailed history about the medical, surgical and other relevant history was obtained from all the patients using a Questionnaire after obtaining consent. Patients who were treated with any antibiotics in the past 1 month prior to the visit were excluded from the study.

Three high vaginal swab samples were collected aseptically from the lateral and posterior wall of the vagina of the patients, after inserting a sterile speculum. The swabs were transported immediately to the Microbiology laboratory and processed. First swab was used for Gram staining and the second swab for wet mount and the third was processed for bacterial culture aerobically. The swab was inoculated in Nutrient agar, Blood agar and Mac conkey agar and incubated at 37°C. After 24 hours of incubation, the plates were examined for the presence of growth.

The organisms grown in culture plates were identified using standard microbiological methods using biochemical reactions. The identification of the species of the bacteria were further confirmed by automated vitek 2 compact system. If no growth was observed, the plates were further incubated for 48 hours before reporting as no growth.

Antibiotic susceptibility testing of the bacterial isolates was performed using Kirby Bauer disc diffusion method using Muller Hinton agar plate as per Clinical and Laboratory Standards Institute (CLSI) guidelines. The antibiotics used were obtained from Hi Media Laboratories Private Limited, Mumbai. The antimicrobial agents used in antibiotic susceptibility testing were penicillin (10µg), erythromycin (15µg), ampicillin (10µg), amoxycillin clavulanic acid (30µg), clindamycin (2µg), cefoxitin (30µg), amikacin (30µg), imipenem (10µg), meropenem (10µg), cipro-floxacin (5µg), ceftriaxone (30µg), ceftazidime (30µg), piperacillin tazobactam (100/10µg), linezolid (30µg), cotrimoxazole (25µg), tobra-mycin (10µg), cefotaxime(30µg), ceftazidime (30µg) and gentamicin (10µg),

Microsoft excel was used for analysis of the results and the various data were expressed as percentages.

RESULTS

A total of 156 patients with clinical suspicion of vaginitis were included in the study. Out of the 156 high vaginal swab samples, 33 yielded bacterial growth under aerobic conditions. So, the prevalence rate of aerobic vaginitis in our study was 21.2%.

The study group included women of age ranged from 15 to 65 years. The rate of isolation of bacterial aerobic pathogens was found to be more in the age group of 21 – 30 years followed by 31-40 years, as depicted in table 1.

Table 1. Age wise distribution of cases

Age group (years)	No. of samples (n = 156)	No. of culture positive samples (n =33)
<20	3	1
21-30	72	17
31-40	48	9
41-50	18	3
51-60	12	2
>60	3	1

The common aerobic bacteria causing vaginitis isolated in the present study were *Staphylococcus aureus*, *Klebsiella species*, β -haemolytic streptococci. (Table 3)

There was higher rate of isolation of Gram positive organisms in the current study (Fig. 1).

The Gram Negative isolates were highly susceptible to meropenem and imipenem. They showed moderate susceptibility towards cephalosporins and aminoglycosides.

Table 2. Bacteria isolated in the present study

Organisms	No. of isolates	Percentage (%)
<i>Staphylococcus aureus</i>	8	24.2
β haemolytic streptococci	7	21.2
<i>Enterococcus sp.</i>	2	6
Coagulase Negative <i>Staphylococcus</i>	2	6
<i>Klebsiella sp.</i>	7	21.2
<i>Escherichia coli</i>	5	15.2
<i>Pseudomonas sp.</i>	2	6

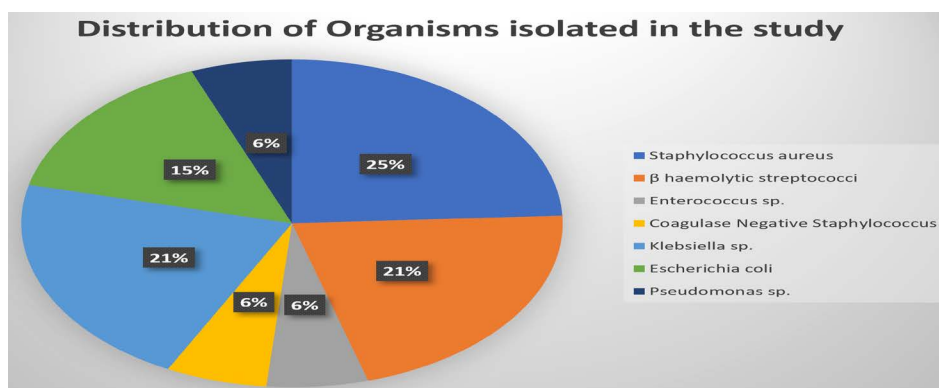


Fig. 1. Distribution of microorganisms

DISCUSSION

Vaginitis is a clinical condition which may lead to significant discomfort, morbidity and even recurrent medical visits. These infections, if left untreated may lead to complications, especially in pregnant women and women of child bearing age group⁸.

The prevalence of vaginitis due to aerobic bacterial pathogens in this study was 21.2%, which correlates well with the previous studies by Sangeetha et al. (2015) who reported 20.8% prevalence in patients of reproductive age and another study by Fan et al (2013) who reported a prevalence rate of 23.74% in their study^{9,10}.

Table 3. Percentage of sensitivity of Gram positive organisms to various antibiotics

Antibiotics	<i>Staphylococcus aureus</i> (n=8) %	β haemolytic streptococci (n=7) %	<i>Enterococcus</i> sp.(n=2) %	Coagulase Negative <i>Staphylococcus</i> (n=2) %
Penicillin	4 (50)	5 (71.4)	1 (50)	2(100)
Erythromycin	4 (50)	5 (71.4)	NT	1(50)
Ampicillin	6 (75)	6 (85.7)	2 (100)	NT
Amoxycillin	6 (75)	7 (100)	2 (100)	2(100)
clavulanic acid				
Ciprofloxacin	6 (75)	5 (71.4)	2 (100)	2 (100)
Clindamycin	6 (75)	6 (85.7)	NT	1(50)
Linezolid	8 (100)	7 (100)	2 (100)	2 (100)
Cefoxitin	6 (75)	4 (57.1)	NT	1(50)

NT : Not tested

Table 4. Percentage of sensitivity of Gram negative isolates to various antibiotics

Antibiotics	<i>Klebsiella</i> sp. (n=7) %	<i>Escherichia coli</i> (n=5) %	<i>Pseudomonas</i> sp. (n=2) %
Piperacillin	6 (85.7)	3 (60)	1 (50)
Tazobactem			
Imipenem	6 (85.7)	5 (100)	0
Meropenem	7 (100)	5 (100)	1(50)
Gentamicin	4 (57.1)	5 (100)	1(50)
Amikacin	4 (57.1)	3 (60)	1(50)
Cefotaxime	2 (28.5)	1 (20)	1(50)
Ceftazidime	3 (42.8)	1 (20)	0
Ciprofloxacin	2 (28.5)	4 (80)	1(50)
Cotrimoxazole	2 (28.5)	2 (40)	NT
Tobramycin	4 (57.1)	NT	1(50)

NT – Not tested

Another study from Solapur, Maharashtra showed 37% prevalence of bacterial vaginitis in their study which could be due to the variation in the hygienic conditions of the patients. All these studies highlight that aerobic bacteria are also one of the frequent causes of vaginitis and hence, the practice of empirical antifungal therapy for most of the cases of vaginitis without investigation needs to be revised.

In the present study, the prevalence of aerobic vaginitis was more in the women of reproductive age group of 20-30 years (51.5%) followed by 30-40 years (27.3%). This is in accordance with the previous studies by Kahn *et al.* and Sangeetha *et al.*^{8,9}

Staphylococcus aureus (24.2%) was found to be the most common Gram positive cocci

isolated in the present study followed by Group B streptococci (GBS) (21.2%). In the previous study by Yancey *et al.*, GBS was present in 26% of the cases¹¹. However, in a study done in Kerala, only 12% of the cases had GBS colonisation.¹² Coagulase negative *Staphylococcus* (CONS) is usually considered as a probable skin commensal. CONS has been isolated from 6% of the cases in our study.

Enterobacteriaceae are often isolated from patients with vaginitis in reproductive age group in various studies reported by Swamy *et al.*, (2015), Mohamed *et al.*, (2015) and Kumar *et al.*, (2015)¹³⁻¹⁵. This could be due to the poor hygiene and entry of the gut flora into genital tract causing vaginal infections¹³⁻¹⁵. Similar to the previous study by Lakshmi *et al.*, 2012, *Escherichia coli* has

been isolated from 15 % of the cases¹⁶. However, *Klebsiella* species were found to be the most pathogenic Gram negative bacteria in this study, unlike the previous report in which *Escherichia coli* was the most frequently isolated organism¹⁶. *Pseudomonas* species has been found in 6% of the cases.

Susceptibility testing of the isolates revealed that piperacillin tazobactam, imipenem, meropenem are the antibiotics of choice for treating the infections caused due to Gram negative isolates. The Gram negative bacteria also showed a moderate susceptibility rate towards gentamicin, amikacin and ciprofloxacin. For Gram positive bacteria, Linezolid, amoxicillin clavulanic acid, ampicillin, clindamycin showed a good susceptibility rate followed by ciprofloxacin, erythromycin and ceftioxin.

Appropriate detection of vaginal infections is important in all age groups for better management of the cases. However, the detection of vaginitis in reproductive age group especially during pregnancy is significant due to the high risk of transmission of bacterial infections to the neonates during their delivery period by Group B streptococci, *Escherichia coli* and other coliforms leading to sepsis in the newborn¹⁷. Thus, appropriate diagnosis and management of vaginitis cases would help in minimisation of complications as well as reduction in recurrence of such infections. Further, proper usage of antimicrobials helps in prevention of emergence of resistant organisms.

CONCLUSION

Vaginitis is one of the frequently occurring condition in women of all ages. The present study provides information regarding the prevalence of aerobic bacterial pathogens in patients suffering from vaginitis. The efficient management of the vaginitis cases depends on the appropriate diagnosis of the causative agent to initiate the correct therapy and prevent the consequences of the vaginitis. Further, this may help in judicious usage of the antimicrobial drugs preventing the emergence of resistant bugs. Appropriate diagnosis of the vaginitis cases using standard microbiological methods with complete antimicrobial susceptibility testing is highly recommended.

ACKNOWLEDGMENTS

None

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHORS' CONTRIBUTION

All authors have made substantial, direct and intellectual contribution to the work and approved it for publication.

FUNDING

None.

DATA AVAILABILITY

All datasets generated or analyzed during this study are included in the manuscript.

ETHICS STATEMENT

The study has been approved by the Institutional Research committee and Institutional Human Ethical committee.

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