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Prevalence of Aerobic Vaginal Pathogens and Their Antibiotic Susceptibility Pattern in a Tertiary Care Hospital.

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ABSTRACT

Vaginitis is a very common disease of women in reproductive age group all over the world. Nearly 5-10 million women are affected every year. Vaginitis is commonly attributed to bacterial vaginosis, vulvovaginal candidiasis and Trichomoniasis. But there is high prevalence rate of aerobic vaginal pathogens from high vaginal swabs. This condition typically will not respond to antibacterial vaginosis medication. So it has to be managed based on culture and sensitivity reports. The present study was carried out in a tertiary care hospital from March 2014 to August 2014. A total of 215 high vaginal swabs from patients who were in reproductive age group were collected and processed. Significant growth was obtained in 118 samples. *Staphylococcus aureus* was the most prevalent organism (27.1%), followed by *Escherichia coli* (24.6%), Candida spp (22%), *Enterococcus* spp (8.5%), *Klebsiella* spp(4.2%), *Streptococcus* spp (4.2%), *Pseudomonas* spp (2.5%), *Acinetobacter* spp (2.5%), *Citrobacter* spp (0.85%)and *Enterobacter* spp (0.85%). There was a high prevalence of Methicillin Resistant Staphylococcus aureus (MRSA) (62.5%) among the isolates of *Staphylococcus aureus*. 100% sensitivity was noted with Vancomycin & Linezolid. The chemotherapeutic agents showing high sensitivity for gram negative rods include Amikacin, Levofloxacin, Imipenem, Cefoperazone-Sulbactum & Tigecycline. To conclude, all patients with symptoms of vaginitis should be investigated thoroughly. Culture & sensitivity should be mandatory and treatment should be based on invitro antibiotic susceptibility testing.

Keywords: vaginitis, multidrug resistance, highvaginal swab, aerobic vaginal pathogens.

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INTRODUCTION

The microbial flora of vagina is a complicated environment, comprising of many microbiological species in variable proportions. The normal vaginal flora is usually well maintained by a complex balance of organisms [1]. *Lactobacillus* species constitute the predominant microorganism among the normal vaginal flora and it is responsible for maintaining the acidic vaginal pH [2]. This gives protection from various pathogens which can invade the vaginal mucosa [3]. The other microorganisms which are part of normal vaginal flora include Coagulase negative Staphylococcus (CONS), *Diphtheroids* and *Micrococcus*. The microorganisms in the vagina which are potential pathogens include *Staphylococcus aureus*, *Enterococcus* species, beta haemolytic *Streptococcus*, *Neisseria* species, *Escherichia coli*, *Klebsiella* species & *Candida* species [4].

Vaginitis is one of the most common infections in women. Approximately 5-10 million females every year seek gynaecologic advice for vaginitis [5,6]. The most common etiology of infectious vaginitis is attributed to bacterial vaginosis, vulvovaginal candidiasis and trichomoniasis [4]. Etiological diagnosis may not be arrived in 7-72% of females with symptoms of vaginitis and such types of abnormal vaginal flora which can neither be considered as normal flora nor as bacterial vaginosis have been grouped as intermediate flora [7].

Bacterial vaginitis can occur in any age group but more commonly diagnosed in females of reproductive age group [4]. Diagnosis and treatment can be misleading if based only on clinical symptoms & signs [8]. The high prevalence of bacterial vaginitis demands thorough investigation of all symptomatic patients. Culture & sensitivity should be done invariably. Hence this study was conducted to throw light on the prevalence of aerobic vaginal pathogens and their invitro antibiotic susceptibility pattern in a tertiary care centre in India.

MATERIALS AND METHODS

The present study was carried out in a tertiary care hospital in Chennai from March 2014 to August 2014. A total of 215 high vaginal swabs from both inpatients and outpatients who were in reproductive age group were collected using sterile swabs after getting informed consent. The samples were processed immediately in the bacteriology lab. Direct Gram's stained smears were observed. The samples were inoculated into sheep blood agar and Mac Conkey agar. The culture plates were incubated aerobically at 37ºc for 24 to 48 hours. The isolates were identified with the help of colony morphology, Gram's staining and biochemical analysis [9].

The antibiotic susceptibility testing was done by Kirby-Bauer disc diffusion method using Mueller Hinton agar and sheep blood agar for fastidious organisms [10]. The antibiotics tested were cefoxitin (30mcg), amoxyclav (20/10mcg), vancomycin (30mcg), linezolid (30mcg), cefotaxime(30mcg), cefpirome (30mcg), cefixime (5mcg), cephalexin (30mcg), cotrimoxazole (25mcg), ciprofloxacin (5mcg), levofloxacin (5mcg), clindamycin (2mcg), azithromycin (30mcg), high level gentamicin (120mcg), gentamicin (10mcg), ampicillin (10mcg), piperacillin (100mcg), ceftazidime (30mcg), ertapenem (10mcg), amikacin (30mcg), imipenem (10mcg), tigecycline (15mcg) and cefoperazone-sulbactum (50/50mcg). Interpretation of the diameter of zone of inhibition was done using CLSI guidelines.

RESULTS

A total of 215 high vaginal swabs were processed for culture and sensitivity, out of which 11 samples (5.1%) were sterile, 86 samples (40%) grew normal vaginal flora and 118 samples (54.8%) grew pathogens. The most prevalent organism was *Staphylococcus aureus* from 32 swabs (27.1%). This was followed by *Escherichia coli* 29 (24.6%), *Candida* species 26 (22.03%), *Enterococcus* species 10 (8.5%), *Klebsiella* species 5 (4.2%), *Steptococcus* species 5 (4.2%), *Pseudomonas* species (2.5%), *Acinetobacter* species 3 (2.5%), *Citrobacter* species 1 (0.85%) and *Enterobacter* species 1 (0.85%).

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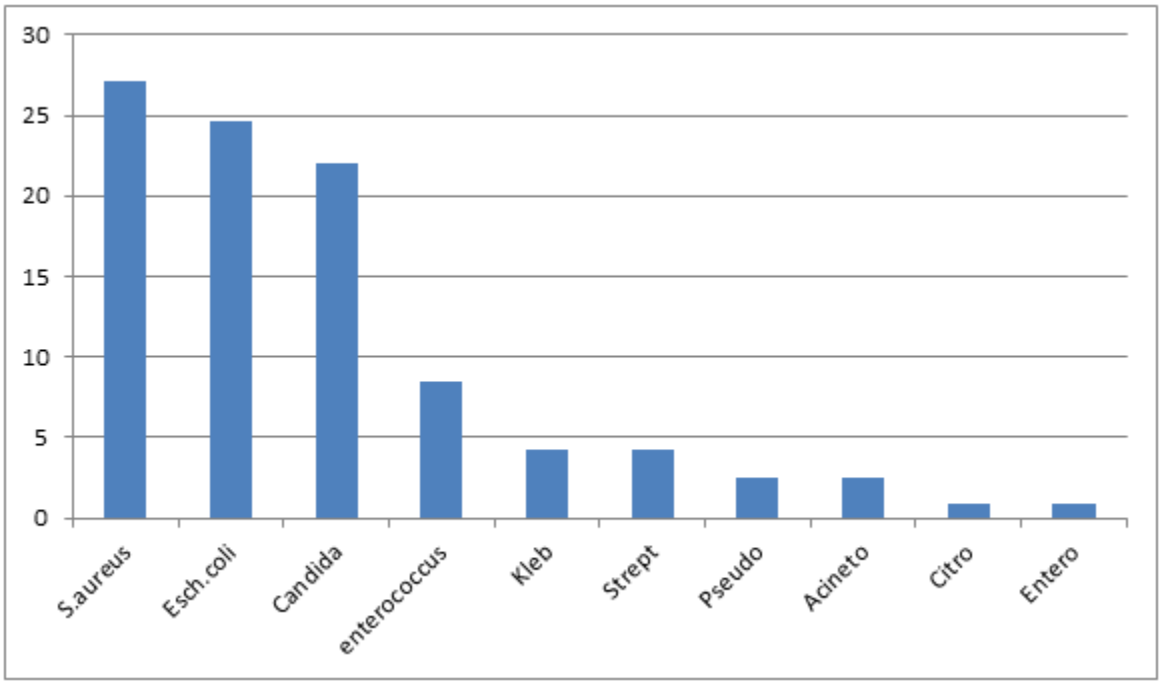
Table 1: Prevalence of aerobic vaginal pathogens from high vaginal swabs

Organism	Number of Isolates	% of Isolates
Staphylococcus aureus	32	27.1
Escherichia coli	29	24.6
Enterococcus spp	10	8.5
Klebsiella spp	5	4.2
Streptococcus spp	5	4.2
Pseudomonas spp	3	2.5
Acinetobacter spp	3	2.5
Enterobacter spp	1	0.85
Citrobacter spp	1	0.85
Candida spp	26	22.03
Normal flora	86	40

Figure 1: Prevalence of aerobic vaginal pathogens from HVS



The detailed results of the antibiotic susceptibility pattern of the various isolates are shown in Tables 2 and 3.

Table 2: Percentage of susceptibility of aerobic vaginal isolates (Gram positive cocci) to various antibiotics

Antibiotics	Staphylococcus aureus	Enterococcus spp	Streptococcus spp
Cefoxitin	37.5	-	-
Amoxyclav	25	70	80
Vancomycin	100	89.8	100
Linezolid	100	100	100
Cefotaxime	47.1	-	-
Cephalexin	18.75	-	-
Cefpirome	29.4	-	-
Cotrimoxazole	20	-	-
Ciprofloxacin	62.3	61.5	75
Levofloxacin	78.9	95.8	96.3
Clindamycin	70.6	-	-
Azithromycin	55.5	-	-
HL Gentamicin	-	90	80
Ampicillin	-	33	93.1
Piperacillin	-	79	94.9

Table 3: Percentage of susceptibility of aerobic vaginal isolates (Gram negative rods) to various antibiotics

Antibiotics	Escherichia coli	Klebsiellsa spp	Acinetobacter spp	Pseudomonas spp
Ceftazidime	71	72	65	66.6
Amoxyclav	47	59.7	-	-
Ertapenem	54.6	62.9	53	-
Amikacin	95.4	96	93	97
Gentamicin	77.3	79	67	69
Ciprofloxacin	66.6	79.8	62	64
Levofloxacin	-	96	97.6	89.5
Cefixime	75	60	-	-
Cefotaxime	87.3	82.9	54	49.6
Cefpirom	88.6	89.3	59	53
Imipenem	98	100	-	95
Tigecycline	100	100	100	-
Cefoperazone - sulbactum	100	100	92	91

DISCUSSION

The microbial ecology of vagina plays a crucial role in the prevention of any vaginal infection in women. *Lactobacillus* is mainly responsible for maintaining the acidic vaginal pH (below 4.5) and thereby preventing the multiplication of potentially pathogenic microorganisms. Usage of antimicrobials like broad spectrum penicillins and tetracyclines can suppress or eliminate the helpful bacteria in the genital tract there by leading to overgrowth of resistant organisms [11].

In our study *Staphylococcus aureus* was the most prevalent vaginal pathogen (27.1%). When the vaginal mucosa is colonized with this microorganism, it can predispose to a dreaded condition namely toxic shock syndrome [12-14]. Among the isolates of *Staphylococcus aureus* only 37.5% were sensitive to methicillin and 62.5% were methicillin resistant. All strains of MRSA were found to be 100% sensitive to vancomycin and linezolid. Most of the strains were multidrug resistant.

The second most prevalent organism isolated from the high vaginal swabs in our study was *Escherichia coli* (24.6%). Isolation of faecal microflora from vagina was correlated with unhygienic bowel practices in the past [15]. When the female introitus is colonized with these organisms there is a strong predisposition to recurrent urinary tract infection [(1)]. From our study we infer that *S.aureus* and *E.coli* are the most commonly isolated pathogens from patients with aerobic bacterial vaginitis and this correlates with Gilbert et al study.

Other Gram positive cocci isolated from HVS in our study include *Enterococcus* species (8.5%) and *Streptococcus* species (4.2%). The *Streptococcus* species are associated with second trimester miscarriages [16].

Other Gram negative rods isolated from HVS in our study include *Klebsiella* species (4.2%), *Pseudomonas* species (2.5%), *Acinetobacter* species (2.5%), *Enterobacter* species and *Citrobacter* species (0.85% each). Prevalence rate of *Candida* species was 22.03%.

Aerobic vaginitis typically has no response to bacterial vaginosis medications. So it has to be managed based on the reports of culture and sensitivity pattern.

The prevalence of MRSA is high among the vaginal isolates of *S.aureus.* Majority of the MRSA strains were multidrug resistant when compared to Methicillin Sensitive Staphylococcus aureus (MSSA) [17]. All the strains of MRSA were found to be sensitive to vancomycin and linezolid. Higher sensitivity rate was observed with clindamycin (70.6%), levofloxacin (78.9%) and azithromycin (55.5%). Lesser activity was noted with first generation cephalosporins (18.75%) & higher generation cephalosporins (47.1%).

Among *Streptococcus* species higher sensitivity rate was observed with ampicillin (93.1%), piperacillin (94.9%) and levofloxacin (96.3%). All the strains of *Streptococcus* species were sensitive to vancomycin and linezolid.

Among *Enterococci*, higher sensitivity rate was observed with amoxyclav (70%), levofloxacin (95.8%), HL gentamicin (90%), piperacillin (79%) and vancomycin (89.8%). All strains of *Enterococcus* species were susceptible to linezolid.

The most effective chemotherapeutic agents against fermenters group (Esch.coli and Klebsiella species) in our study were amikacin, levofloxacin, imipenem, tigecycline, cefoperazone – sulbactum and higher generation cephalosporins. The most effective antibiotics against nonfermenters (Pseudomonas species and Acinetobacter species) in our study were amikacin, levofloxacin, imipenem, cefoperazone – sulbactum and tigecycline.

CONCLUSION

As there is a high prevalence of bacterial vaginitis, all patients with gynaecological symptoms should be investigated thoroughly. Antibacterial vaginosis medications will not be helpful in patients with aerobic bacterial vaginitis. So culture & sensitivity has to be done invariably to find out the etiologic agent and should be treated accordingly. Multidrug resistant strains are on the rise and this scenario narrows down the choice of antimicrobial agents to only a few susceptible drugs. So irrational use of antibiotics should be avoided and treatment should be strictly based on invitro antibiotic susceptibility testing. Last but not the least, every hospital should formulate their own antibiotic policy based on local susceptibility patterns. This will be helpful for choosing antibiotics for empirical therapy by clinicians. This helps to curtail the emergence and spread of multidrug resistant organisms.

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