Incidence of Enterococcal Urinary Tract Infection and it's Sensitivity Pattern among Patients Attending Teerthanker Mahaveer Medical College and Research Centre, Moradabad, India

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Abstract

Introduction: *Enterococcus* is an opportunistic pathogen which is a major causative agent of nosocomial infection and it is especially in hospitalized patients. It causes urinary tract infection (UTI). The aim of this study is to isolate enterococci from cases of UTI and to know the antibiotic susceptibility pattern of the isolates.

Materials and Methods: Early morning clean catch midstream urine sample was collected into a wide-mouthed sterile screw capped container then transported to the laboratory. Then, culture and antibiotic susceptibility were performed.

Result: Out of 627 bacteriuria patients, 106 *Enterococcus* were isolated and 521 other bacteria (83.09%). Out of 627 patients positive urine culture, the isolation of bacterial species were found as *Escherichia coli* 231 (36.84%), *Staphylococcus aureus* 119 (18.98%), coagulase-negative *Staphylococci* 21 (3.34%), *Enterococcus* 106 (16.91%), *Proteus* 13 (2.07%), *Enterobacter* 26 (4.15%), *Citrobacter* 16 (2.55%), *Pseudomonas* 19 (3.03%), *Acinetobacter* 24 (3.83%), and *Klebsiella pneumonia* 48 (7.66%). Females 70 (66.04%) were more prone to enterococcal infection as compared to male 36 (33.96%) and high prevalence was seen in the age group of 21-30 years 36 (33.96%) followed by 31-40, 51-60, 41-50, 61-70, 11-20, and <10. *Enterococcus* showed 98.11% sensitivity against vancomycin followed by teicoplanin.

Discussion: The commensal of the intestinal human flora are enterococci. In this study, enterococcal UTI was more prevalent than the other studies. In this study, females were 70 (66.04%) more prone to enterococcal infection as compared to male 36 (33.96%), and the highest prevalence of enterococcal infection was seen in the age group 21-30 36 (33.96%) years and most sensitive to vancomycin (98.11%), linezolid (92.45%).

Conclusion: It is concluded that *Enterococcus* are Gram-positive cocci presenting as a harmless commensals, causes UTI infection and which is highly prevalent among females than males. Early and proper treatment can decrease the antibiotic resistance.

Key words: Coagulase-negative *Staphylococci*, *Escherichia coli*, *Staphylococcus aureus*, Urinary tract infection, Vancomycin resistant *Enterococcus*

INTRODUCTION

In each year, millions of people are affecting by urinary tract infection (UTI), it is a serious health problem. In the

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world, all age groups across the lifespan are affected and it is the most cause of mortality and morbidity. The lower urinary tract or both the upper and lower tract is involved.

Within urinary tract, the most frequent sites of infection are urethra and urinary bladder.³ The risk of infection related to the frequency of sex was found that women were more prone to UTIs than man.⁴ A significant number of microorganism, usually >10⁵ cells/ml of urine is said to exist by UTI, are detected in properly collected mid-stream "clean catch" urine.^{5,6} The detection and identification of

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the causative pathogen in the urine are the gold standard for diagnosis.⁷

The important opportunistic pathogens and indigenous flora of the intestinal tract, oral cavity and genitourinary tract of human are enterococci especially in hospitalized patients. The human pathogen capable of causing UTI, intra-abdominal infection, pelvic wound infection biliary tract infection, respiratory infection, neonatal sepsis accompanied by bacteremia or meningitis or both are *Enterococcus faecalis* (80-90%) and *Enterococcus faecium* (5-10%) which are two commonly prevalent species.

Now enterococci are included in the genus *Streptococcus* but originally, it was classified as enteric Gram-positive cocci. Enterococci were classified as Group D streptococci by different biochemical characteristics in the late 1930s. In the late 1930s, it was suggested that for streptococci the term enterococci should be used specifically that grow at pH 9.6, 10 and 45°C both, in the presence of 6.5% NaCl (sodium chloride), hydrolyze esculine and survive at 60°C for 30 min.

Studies were involved during the mid-1980s, that includes fatty acid composition, nucleic acid hybridization and comparative oligonucleotide cataloguing of 16s ribonucleic acid led to the acceptance that enterococci were merited their own genus and sufficiently distinct from other streptococci.

Although approximately a dozen Enterococcus species have been recognized, and out of those only two are responsible for the majority of human infections. There are at least 12 species that causes enterococci infection that includes E. faecalis, Enterococcus avium, Enterococcus casseliflavus, Enterococcus mundtii, Enterococcus pseudoavium, Enterococcus raffinosus, Enterococcus durans, Enterococcus gallinarum, E. faecium, Enterococcus hirae, Enterococcus malodoratus, and Enterococcus solitarius. Addition to this list, some additional species have been also proposed such as Enterococcus columbae, Enterococcus cecorum, Enterococcus saccharolyticus, Enterococcus dispar, Enterococcus seriolicida, Enterococcus sulfureus, and Enterococcus flavescens. Either E. faecalis or E. faecium causes infections most clinically. Until recently the predominant enterococcal species are E. faecalis and E. faecium that accounting for 80-90% and 5-15% of all clinical isolates, respectively.¹⁰

A part of the normal flora of bowel, genital tract anterior urethrae of humans of humans are enterococci¹¹ among hospitalized patients, the organism can cause serious infection like endocarditis UTI, wound infection is enterococci, have been considered of relatively low virulence.¹¹ Symptoms include frequency, dysuria,

suprapubic pain along with loin pain. There may be fever with rigors. UTI due to the enlarged prostate is relatively common cloudy, dark, bloody, or unusual smelling urine.

The most common infection caused by bacteria in the urinary tract is an enterococcal infection but it also causes bacteremia, wound infection, intra-abdominal abscesses, infective endocarditis, and infrequently meninigitis. ¹² All over the word, particularly in developing countries, UTIs were treated by antibiotics though resistance to antibiotics is highly prevalent in bacterial isolates. ¹³ Due to the increasing level or antimicrobial resistance, treatment of UTI has thrown up a lot of challenges in over the years. ¹⁴

In UTI, endocarditis, bacteremia, intra-abdominal and intra-pelvic abscesses are the serious relevant infection of nosocomial and among *Enterococcus* genus; the main causative agents are *E. faecium* and *E. faecalis*. ¹⁵

The commensal organisms are enterococci. In the bowel, the normal inhabit are enterococci. They become opportunistic pathogens. Human infection caused by enterococcal species includes *E. avium*, *E. gallinarum*, *E. casselifavus*, *E. durans*, *E. raffinosus* and *E. mundtii.* ¹⁶ It increase virulence elements in hospitalized colonize patients of nosocomial enterococci. ¹⁷

MATERIALS AND METHODS

The study was conducted in Department of Microbiology, Teerthanker Mahaveer Medical College and Research Centre, Moradabad from February 2015 to January 2016; 106 isolated *Enterococcus* were collected from the urine sample.

Early morning clean catch midstream urine sample was collected into a wide-mouthed sterile screw capped container from clinically suspected patients.

With a calibrated micro loop, 0.001 ml of urine was cultured onto cystine lactose electrolyte lactose deficient agar, blood agar, MacConkey agar, and Muller Hinton Agar Plates. After overnight incubation at 37°C for 24 h, colony counts yielding bacterial growth of ≥10⁵/ml was considered as significant in the urine sample.

Heat tolerance test, i.e., growth for 30 min at 60°C, in the presence of sodium chloride of 6.5% (salt tolerance test), growth occur, *Enterococcus* genus were confirmed by catalase negative and blackening of bile-esculin agar among all the Gram-positive cocci. Sugar fermentation test including, lactose, mannitol, glucose, arabinose, potassium tellurite reduction, motility testing species of *Enterococcus* were further identified.

As per CLSI guidelines, on Mueller Hinton agar, antimicrobial susceptibility testing was performance: Vancomycin (30 μg), penicillin-G (10 μg), ampicillin (10 μg), amoxicillin (20 μg), ampicillin/sulbactam (10/10 μg), norfloxacin (10 μg), co-trimoxazole (25 μg), erythromycin (15 μg), gentamicin (120 μg), chloramphenicol (30 μg), ciprofloxacin (5 μg), clindamycin (2 μg), levofloxacin (5 μg), linezolid (15 μg), tobramycin (10 μg), amikacin (30 μg), pristinomycin (15 μg), teicoplanin (30 μg), tetracycline (30 μg), rifampicin (5 μg), ofloxacin (5 μg), and cefotaxime (30 μg).

RESULT

In this study, 627 patients were showed to be urine culture positive of which there were 106 Enterococcus (16.91%) and 521 other bacteria (83.09%) (Figure 1). Out of 627 patients positive urine culture, the isolation of bacterial species were found as Escherichia coli 231 (36.84%), Staphylococcus aureus 119 (18.98%), coagulase-negative Staphylococci 21 (3.34%), Enterococcus 106 (16.91%), Proteus 13 (2.07%), Enterobacter 26 (4.15%), Citrobacter 16 (2.55%), Pseudomonas 19 (3.03%), Acinetobacter 24 (3.83%), and Klebsiella pneumonia 48 (7.66%) (Table 1) (Figure 2).

The Gram-positive bacteria and Gram-negative bacteria proportion were 250 (39.87%) and 377 (60.13%), respectively. Female 70 (66.04%) were found to be more prone to enterococcal infection as compared to male 36 (33.96%) (Table 2). The high prevalence of enterococcal infection was seen in the age group 21-30 years 36 (33.96%)

Table 1: Bacterial isolates in positive culture

Bacteria	Frequency (n)	Percentage
Enterococcus	106	16.91
E. coli	231	36.84
Klebsiella	48	7.66
S. aureus	119	18.98
CONS	21	3.34
Streptococcus	4	0.64
Enterobacter	26	4.15
Citrobacter	16	2.55
Pseudomonas	19	3.03
Proteus	13	2.07
Acinetobacter	24	3.83
Total	627	100

E. coli: Escherichia coli, S. aureus: Staphylococcus aureus, CONS: Coagulase-negative Staphylococci

Table 2: Distribution of *Enterococcus* according to gender

Gender	Frequency	Percentage		
Female	70	66.04		
Male	36	33.96		

followed by 31-40, 51-60, 41-50, 61-70, 11-20 and <10 age groups (Table 3).

In this study, *Enterococcus* showed that against vancomycin 98.11% sensitivity which is followed by teicoplanin (94.33%) and ampicillin/sulbactam (73.59%), ampicillin (58.49%), amoxicillin (58.49%), rifampicin (58.49%), and cefotaxime (50.94%), among urinary isolates, least sensitivity was observed with clindamycin (11.32%) (Table 4) (Figure 3).

DISCUSSION

The commensal of the intestinal human flora is enterococci. The site such as genitourinary tract, oral cavity and skin especially in the perineal area are less often colonized by these organisms. In the hospitalized patients, the main sites

Table 3: Distribution of *Enterococcus* is according to age and sex

Age	Female <i>n</i> =70 (%)	Male <i>n</i> =36 (%)	Total n=106 (%)
0-10	2 (2.86)	-	2 (1.89)
11-20	14 (20)	4 (11.11)	18 (16.98)
21-30	32 (45.71)	6 (16.67)	38 (35.85)
31-40	14 (20)	8 (22.22)	22 (20.75)
41-50	4 (5.71)	4 (11.11)	8 (7.55)
51-60	-	10 (27.78)	10 (9.43)
61-70	4 (5.71)	4 (11.11)	8 (7.55)
Total	70	36	106

Table 4: Antibiotic sensitivity pattern of Enterococcus isolated from urine sample

Antibiotics	Sensitive n (%)
Vancomycin	104 (98.11)
Teicoplanin	100 (94.33)
Linezolid	98 (92.45)
Ampicillin/sulbactam	78 (73.59)
Ampicillin	62 (58.49)
Amoxicillin	62 (58.49)
Rifampicin	62 (58.49)
Cefotaxime	54 (50.94)
Penicillin-G	50 (47.17)
Chloramphenicol	44 (41.50)
Cephalexin	28 (26.41)
Tobramycin	28 (26.41)
Tetracycline	26 (24.52)
Ofloxacin	24 (22.64)
Norfloxacin	22 (20.75)
Pristinomycin	20 (18.87)
Ciprofloxacin	20 (18.87)
Co-trimoxazole	18 (16.98)
Gentamicin	18 (16.98)
Levofloxacin	18 (16.98)
Erythromycin	14 (13.20)
Amikacin	14 (13.20)
Clindamycin	12 (11.32)

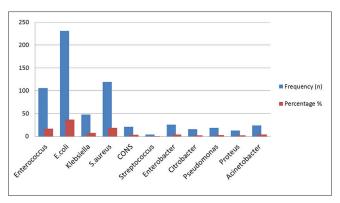


Figure 1: Distribution of different bacteria

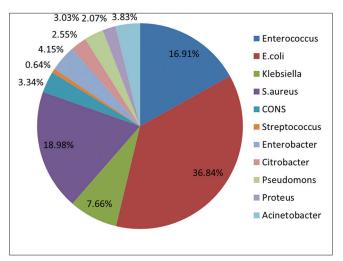


Figure 2: Distribution of different bacterial percentage

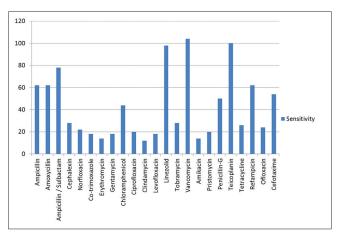


Figure 3: Antibiotic sensitivity pattern of Enterococcus

of colonization are ulcers, gastrointestinal tract and soft tissue wounds. In the recent years, important causes which are increasingly of nosocomial infections are enterococci which are regarded as low-grade pathogens traditionally.¹⁰

In the current study, enterococcal UTI was more frequent than has been reported in other studies. In present study 106 (16.91%) *Enterococcus* was isolated from the urine

sample. This study is comparable with the study of Tayebi *et al.*¹⁸ and Jombo *et al.*, ¹⁹ in which *Enterococcus* sp. was 8.7% and 12.4%, respectively.

In this study, females were 70 (66.04%) more prone to enterococcal infection as compared to male 36 (33.96%). This is comparable with the study carried out by Shrivastav *et al.* in which the number of females infected with enterococcal infection was more (72%) than number of males.²⁰

In this study, the highest prevalence of enterococcal infection was seen in the age group 21-30 was 36 (33.96%) years. Similar findings of higher infection rate in the age group 21-30 years were reported by Bose *et al.*²¹

In this study, *Enterococcus* isolates from the various clinical samples were sensitive to vancomycin (98.11%) and linezolid (92.45%). This study is comparable with the study carried out by Abdulla and Abdulla,²² in which sensitivity of vancomycin and teicoplanin was 99.1% each.

CONCLUSION

It is concluded that *Enterococcus* are Gram-positive cocci presenting as harmless commensal to multifaceted deadly pathogens. It causes UTI infection, which is highly prevalent among females than males. Thus, in present study *Enterococcus* showed the highest susceptibility to the vancomycin, hence, it is the drug of choice, limiting the use of linezolid only in the case of vancomycin-resistant *Enterococcus*.

The positive urine culture with the antibiotic sensitivity of the isolates is very important to an antimicrobial therapy, as antibiotic resistance is a worldwide problem which causes ineffectiveness of treatment. Early and proper treatment can decrease the antibiotic resistance.

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