Section: Microbiology

Original Research Article

ENTEROCOCCAL BURDEN IN URINARY TRACT INFECTIONS IN DARBHANGA MEDICAL COLLEGE, DARBHANGA, BIHAR, INDIA

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Abstract

Background: 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:** In this study, 690 patients were showed to be urine culture positive of which there were 135 Enterococcus (19.61%) and 555 other bacteria (80.79%). Out of 690 patients positive urine culture, the isolation of bacterial species were found as Escherichia coli 259 (38.64%), Staphylococcus aureus 133 (19.88%), coagulase-negative Staphylococci 29 (4.43%), Enterococcus 135 (19.61%), Proteus 20 (3.67%), Enterobacter 21 (3.14%), Citrobacter 14 (2.44%), Pseudomonas 20 (3.02%), Acinetobacter 15 (2.38%), and Klebsiella pneumonia 45 (6.76%). **Conclusion:** It is concluded that Enterococcus are Gram-positive cocci presenting as a harmless commensal, causes UTI infection and which is highly prevalent among females than males. Early and proper treatment can decrease the antibiotic resistance.

INTRODUCTION

In each year, millions of people are affecting by urinary tract infection (UTI), it is a serious health problem. In the world, all age groups across the lifespan are affected and it is the most cause of mortality and morbidity.[1] The lower urinary tract or both the upper and lower tract is involved.[2] 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 ofthe 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.[8] 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.[9] 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,

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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, Darbhanga Medical College and Hospital from February 2022 to January 2023; 135 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).

RESULTS

In this study, 690 patients were showed to be urine culture positive of which there were 135 Enterococcus (19.61%) and 555 other bacteria (80.79%). Out of 690 patients positive urine culture, the isolation of bacterial species were found as Escherichia coli 259 (38.64%), Staphylococcus aureus 133 (19.88%), coagulase-negative Staphylococci 29 (4.43%), Enterococcus 135 (19.61%), Proteus 20 (3.67%), Enterobacter 21 (3.14%), Citrobacter 14 (2.44%), Pseudomonas 20 (3.02%), Acinetobacter 15 (2.38%), and Klebsiella pneumonia 45 (6.76%) [Table 1].

The Gram-positive bacteria and Gram-negative bacteria proportion were 269 (38.97%) and 421 (61.07%), respectively. Female 86 (63.63%) were found to be more prone to enterococcal infection as compared to male 53 (38.36%) [Table 2]. The high prevalence of enterococcal infection was seen in the age group 21-30 years 53 (38.36%) followed by 31-40, 51-60, 41-50, 61-70, 11-20 and <10 age groups.

In this study, Enterococcus showed that against vancomycin 91.81% sensitivity which is followed by teicoplanin (93.43%) and ampicillin/sulbactam (75.39%), ampicillin (54.89%), amoxicillin (54.89%), rifampicin (54.89%), and cefotaxime (54.04%), among urinary isolates, least sensitivity was observed with clindamycin (13.12%) [Table 3]

Table 1: Bacterial isolates in positive culture

Bacteria	Frequency (n)	Percentage
Enterococcus	135	19.61%

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E. coli	259	38.64%
Klebsiella	45	6.76%
S. aureus	133	19.88%
CONS	29	4.34%
Enterobacter	21	3.14%
Citrobacter	14	2,44%
Pseudomonas	20	3.02%
Proteus	20	3.06%
Acinetobacter	15	2.38%
Total	690	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	84	63.03%
Male	51	38.36%

Table 3: Antibiotic sensitivity pattern of Enterococcus isolated from urine sample

Antibiotics	Sensitive n (%)
Vancomycin	91.81
Teicoplanin	93.43
Linezolid	94.25
Ampicillin/sulbactam	75.39
Ampicillin	54.89
Amoxicillin	54.89
Rifampicin	54.89
Cefotaxime	54.04
Penicillin-G	41.77
Chloramphenicol	45.10
Cephalexin	24.61
Tobramycin	24.61
Tetracycline	25.42
Ofloxacin	26.24
Norfloxacin	27.05
Pristinomycin	17.78
Ciprofloxacin	17.78
Co-trimoxazole	19.68
Gentamicin	19.68
Levofloxacin	19.68
Erythromycin	12.30
Amikacin	12.30
Clindamycin	13.12

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 sitesof 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]

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 84 (63.63%) more prone to enterococcal infection as compared to male 53 (38.36%). 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 53 (38.36%) 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 (91.81%) and linezolid (94.25%). 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

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treatment. Early and proper treatment can decrease the antibiotic resistance.

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