Antibiotic Susceptibility Pattern of Enterococci in a Tertiary Care Hospital of Kanpur, UP

Anjali Tiwari¹, R. Sujatha^{2*}, NashraAfaq³

Abstract:

Background: Enterococci are important agents of nosocomial infection, ranking as the second most common organisms causing complicated urinary tract infections, bacteremia, endocarditis, intra-abdominal and pelvic infections, wound and soft tissue infections, neonatal sepsis, and rarely meningitis. Infections by enterococci have traditionally been treated with cell wall active agents in combination with an aminoglycoside.

Material and methods: The present study was a cross-sectional study in which a total of 1132 clinical specimens, were obtained during the period of 6 months from January 2023 to June 2023. Antibiotic susceptibility testing was done by Kirby Bauer's disc diffusion method according to the CLSI guidelines 2022.

Results: During the study period of 6 months, 42 enterococci were recovered from 1132 different clinical samples, accounting for an infection rate of 3.07%. Among 42 enterococcal isolates, 22 (52.3%) were obtained from urine, 03 (7%) from blood 10 (6.8%) from pus 10(23.8%) from sputum 1(2.4%) and 6 (14.3%) from foley's tip. The predominant isolates were E. Faecalis 36(85.7%) followed by E. faecium06(14.2%).In the antibiotic susceptibility pattern, most of the enterococcal isolates were predominantly sensitive to 100% vancomycin and High-level gentamicin and high level streptomycin followed by linezolid.

Conclusion: Enterococcus faecalis and Enterococcus faecium were the predominant species. It is necessary to implement infection control measures like antimicrobial stewardship, especially restricting the use of antibiotics to a minimum.

Introduction

The majority of infectious diseases are bacterial in origin. The accelerated emergence of antibiotics resistance among the prevalent pathogen is of global health concern. Enterococci are Gram-positive, facultatively anaerobic cocci that may occur in pair or short chains [1]. They were distinguished from streptococci and related taxonomy by their ability to grow at 10°C and 45°C, and growth in the presence of pyrrolidonylarylamidase [1]. Enterococci are commensals of the gastrointestinal tracts of animals from simple invertebrates to humans. They are known to be relatively a virulent in healthy individuals, but have become important opportunistic pathogens, especially in hospitalized patients [2]. They have emerged as important nosocomial pathogens [3, 4]. The most frequent infections caused by these organisms include urinary tract infections, intra abdominal and intra pelvic abscesses [3, 5, and 6]. They also cause surgical wound infections, bacteraemia, endocarditis, neonatal sepsis, and rarely meningitis5.

Although about 23 species of Enterococci have been identified, there are two main species, Enterococcus faecalis and Enterococcus faecium that are responsible for most human enterococcal infections [3, 4, and 5].

This organism has been reported as the second leading cause of nosocomial urinary tract infections and third leading cause of nosocomial bacteraemia in hospitalized patients [5] .Risk factors for infections include frequent exposure to antimicrobial agents particularly the use of vancomycin and third-generation cephalosporins, decreased immunity orneutropenia, renal insufficiency, use of steroids and presence of an indwelling urinary catheter [4,5]. Vancomycin resistant enterococci infection rates are highest among critically ill patients admitted in Intensive Care Units (ICU) with limited treatment options [6]. Infections by enterococci have historically been treated with semi-permeable membrane active agents (e.g., penicillin or ampicillin) in association with an aminoglycoside (streptomycin/ gentamicin); however such combination treatment has failed to work due to emergence of resistance such as HLAR, beta-lactam antibiotics resistance or vancomycin resistance [7].

Material and Methods

The present study was a cross-sectional study conducted in the Department of Microbiology of Rama Medical college hospital and research centre, Kanpur, India for a period of 6 months from January 2023 to June 2023. Approval of ethical committee was obtained from the Institutional Ethical Committee.

Inclusion criteria: Patients of all age groups and gender with history of urinary tract infection, presence of prolonged urinary catheterization and wound infection attending out-patient and inpatient department of

PG Student¹, Department of Microbiology, Rama Medical College Hospital & Research Centre Kanpur

Professor & Head², Department of Microbiology, Rama Medical College Hospital & Research Centre, Kanpur.

Research Associate³, Dept of Microbiology, Rama Medical College Hospital and Research Centre, Kanpur.

medicine, surgery, obstetrics and gynaecology, paediatrics, orthopaedics and ICU, post off at RCM were considered as study population.

Exclusion criteria

faecal samples were excluded from the study.

All the clinical samples except blood were processed initially by plating on CLED agar, Blood agar and Mac Conkey Agar, and incubating at 37°C for 18-24 hours. Blood samples were inoculated first in Brain Heart Infusion (BHI) broth, incubated at 35-37°C for five days, examined daily for microbial growth (turbidity) followed by subculture on CLED agar, blood agar and MacConkey agar. Identification of the enterococcal isolates were performed by adopting standard protocols such as cultural characteristics, gram stain, motility testing, catalase test, bile esculin test, salt tolerance test. The antimicrobial susceptibility testing was performed on Mueller Hinton agar plate by the Kirby Bauer disc diffusion technique with commercially available Hi-Media antibiotic discs on Muller Hinton agar plates, the zone size was interpreted as Susceptible(S), Intermediate(I). Resistant[®] according to Central Institute(CLSI Laboratory Standard 2022) GUIDELINES. The antibiotics tested were as followsfor urinary isolates and catheter tip ampicillin 10 µg, ciprofloxacin 5 µg, linezolid 30 µg, nitrofurantoin 300 µg, vancomycin 30 µg and teicoplanin 30 µg. For isolates from other sites like pus, wound, bloodampicillin 10 µg, linezolid 30 µg, vancomycin 30 µg and teicoplanin 30 μ g. High level gentamycin (120 μ g) and high level streptomycin (300 µg) were used for all isolates.

Results

| Enterococcus | A | ЛР |] | Р | V | Ά | Т | Έ | N | IT | L | Z | HI | L |
|--------------|---|----|---|---|---|---|---|---|---|----|---|---|----|---|
| Spp. | S | R | S | R | S | R | S | R | S | R | S | R | S | Ο |

During the study period of 6 month, 42 enterococci were recovered from different clinical samples, accounting for an infection rate of 3.71 % of which 32 (76.1%) were from inpatients, from out-patient and 10 (23.8%). Among 42 enterococcal isolates, maximum number of isolates were obtained from urine 22 (52.3%) and minimum number from sputum 1(2.3%). (Table 1)

[Table/Fig-1]: Total no. of Enterococcal isolates in various clinical samples.

| Table Enterococcus species (n=42) | Urine (%) | Blood (%) | Pus (%) | Sputum (%) | Foley tip (%) | | | | |
|--|--------------|--------------|-------------|---------------|---------------------|--|--|--|--|
| <i>E.faecalis</i> (n=36) | 19 (52.7) | 3 (8.3) | 8 (22.2) | 1(2.7) | 5 (13.8) | | | | |
| $\begin{array}{c c c c c c c c c c c c c c c c c c c $ | | | | | | | | | |
| Total (n=42) 36(85.71%), 6 (14.28%) | | | | | | | | | |
| *n=No. of isolates | | | | | | | | | |

Females 29 (69%) were found to be more prone to Enterococcal infection as compared to males 13 (30%)

ISSN 2395-0757

(Graph 1). High prevalence of Enterococcal infection was seen in the age group 21-40 years (38%) followed by 61-above, 0-20 and 41-60 age groups (Table 2).

Table No 2: Age Wise Distribution

| Age group (years) | Number (%) |
|-------------------|------------|
| 0-20 | 8(19%) |
| 21-40 | 16(38%) |
| 41-60 | 5(11.9%) |
| 61-ABOVE | 13(30.9%) |



Graph No 1: Gender Wise Distribution

In the present study among the Enterococcus isolated E. faecalis was the predominant species (85.7%) followed by E. faecium (14.2%). On studying the antibiotic susceptibility pattern it was found that most of Enterococcus isolates were predominantly showed 100% sensitivity against linezolid followed by high sensitivity with vancomycin, High level gentamicin (Table 3& Graph 2). Among isolates least sensitivity was observed from clinical sample showed least sensitivity with Penicillin, amoxicillin and tetracycline.



Graph No: Antibiotic sensitivity pattern of enterococcus spp

| | [Table 3]: Antimicrobial | susceptibility pattern | of Enterococcus spp. |
|--|--------------------------|------------------------|----------------------|
|--|--------------------------|------------------------|----------------------|

| E. faecalis | 8 | 11 | 4 | 30 | 36 | 0 | 10 | 26 | 16 | 3 | 33 | 3 | 35 | 1 | 36 | 0 | 13 | 6 |
|--|---|----|---|--------------|----|---|----|----|----|---|----|---|----|---|----|---|----|---|
| E. faecium | 4 | 2 | 2 | 4 | 5 | 1 | 3 | 3 | 5 | 1 | 6 | 0 | 5 | 1 | 6 | 0 | 2 | 1 |
| Total(n=42) | | | | | | | | | | | | | | | | | | |
| Note: As, the number of isolates are not equal, all the antibiotics are not used for all isolates; n=no. of isolates, AMP: | | | | | | | | | | | | | | | | | | |
| Ampicillin; VA: Vancomycin; TE: Tetracycline ;P: Penicillin LZ: Linezolid; HLG: High level gentamycin; HLS: High | | | | | | | | | | | | | | | | | | |
| level streptomycin; *For All sampleExcept urine NIT: Nitrofurantoin; NX: Norfloxacin . *HLG and HLS were used | | | | | | | | | | | | | | | | | | |
| for all isolates | | | | [^] | - | | | | | | | | | | | | | |

[Table-4]: Distribution of clinical isolates in wards n=No. of isolates

| Enterococcus spp. (n=42) | Medicine (%) | Surgery (%) | Gynaecology (%) | Paediatrics (%) | | |
|-----------------------------|-----------------|----------------|--------------------|--------------------|--|--|
| E. faecalis | 16 | 9 | 7 | 4 | | |
| (n=36) | (44) | (25) | (19) | (11.1) | | |
| E. faecium | 3 | 1 | 2 | 0 | | |
| (n=6) | (50) | (16.6) | (33.3) | (0) | | |

Among the wards highest enterococcal infection was observed in Medicine (45.2%) followed by surgery ward (23.8%) and least was in pediatric ward.

Discussion

More females were found to be infected with Enterococci as compared to males. This is comparable with the study carried out by Bose et.al (2013) in which number of females infected with Enterococcal infection was more (68.93%) than number of males [14].

In the present study the highest prevalence of Enterococcal infection was seen in the age group 21-40 (38%) years. Similar findings of higher infection rate in age group 21- 30 yrs was reported by Bose et al. (2013) while, in another study Barros et al. (2009) have reported high prevalence of Enterococcal infection in the age group of 50-60 years[14.15].

E. faecalis and E. Faecium were the only species isolated in the present study. This is comparable with the findings of Bose et.al (2012) that isolated 82% E. faecalis and 18% E. faecium in their study. Mickeen et al. (2002) also isolated only three species i.e., E. faecalis, E faecium and E. durans in their study [16].

In the present study Vancomycin show (100%) sensitivity followed by Linezolid(92%), nitrofuration (50%). pepracillin(14.28%) and High level of gentamicin (99%).In the study of Srivastava et.al (2013)maximum sensitivity was Linezolid show (100%) sensitivity followed by vancomycin(91.5%), nitrofuration (88.5%), norfloxacin (77%), Gentamicin (60%), cefoperazone (54%), nettalin (52%), pepracillin (46%) and-+ciprofloxacin (42%) [17]. Enterococci has been reported as important cause of urinary tract infection (UTI), bacteraemia and few more clinical problems, mostly in hospital settings. Bactericidal synergy between beta-lactam and glycopeptides antibiotics is needed for management of serious enterococcal infections [18].

Conclusion

The present study highlights the prevalence of E. faecalis in our hospital. The data confirms vancomycin as a drug of choice. We further emphasize the need for constant monitoring of antibiotic susceptibility pattern in defined geographical areas which will be helpful in formulating local antibiotic policies.

References

- J. parameswarappa, v. p. basavaraj, and c. m. basavaraj, "isolation, identification, and antibiogram of enterococci isolated from patients with urinary tract infection," annals of African medicine. 2013; 12(3); 176–181.
- Procop gw, church ld, hall gs, janda wm, koneman ew, schreckenberger pc, et al. the gram positive cocci, part ii: streptococci, enterococci, and the "strepococcus like" bacteria. In: koneman'scolor atlas and textbook of diagnostic microbiology. 7th ed. philadelphia: lippincott williams and wilkins; 2017. pp. 768.
- Jenna i.wurster, jose t.saavedra, michael s.gilmore. Impact of antibiotic use on the evolution of enterococcus faccium. idsa. journalof infectious diseases; jan 2016.
- Adelehh.zadeh, mana shojapur, raiyehnazari, majid akbari, masumehsofien, hamid abtahi. Genotyping of vancomycin resistant enterococci in arak hospitals. jundishapur j microbiol.2015; 8(4):e16287
- Varsha gupta, nidhi single, preeti behl, tripti sahoo, jagdish chander. Antimicrobial susceptibility pattern of vancomycin resistant enterococci to newer antimicrobial agents. indian j med res. 2015; 141(4): 483-486.
- A tripathi, skshukla, a singh, kn prasad. a new approach of real time polymerase chain reaction in detection of vancomycin resisistantenterococci and is comparsion with other methods. indian j med microbiol. 2013; 31(1):47-52.
- S.shafiyabi, j.mariraj,ns.sumathi, shanmugam, s.krishna. emergenece of vancomycin resistant enterococci in a tertiary care hospital in south india. int j pharm biomed res. 2013; 4(2): p111-113
- S. c. fernandes and b. dhanashree, "drug resistance & virulence determinants in clinical isolates of enterococcus species," indian journal of medical research. 2013; 137, no. 5, pp. 981–985, 2013.
- Amaschieto, r.martinez, et al. "antimicrobial resistance of enterococcus sp. isolated from the intestinal tract of patients from a university hospital in brazil," memorias' do instituto oswaldo cruz. 2004; 99 (7); pp. 763–767, 2004
- Boyce jm. vancomycin resistant enterococcus. Detection, epidemiology, and control, journal of clinical and diagnostic research. 2021 may, vol-15(5): dc08-dc11

- 11. Carmeli y, eliopoulos gm, samore mh. Antecedent treatment with different antibiotic agents as a risk factor for vancomycin-resistant enterococcus. emerg infects dis. 2002;8:802-07.
- 12. National nosocomial infections surveillance system. National nosocomial infections surveillance (nnis) system report, data summary from january 1992 through june 2004, issued october 2004. Is j infecting control? 2004; 32:470-85.
- Arias ca, murry is. enterococcal species, streptococcus bovis group and leuconostoc species. In: mandell gl, douglas r and bennett je editors. Principles and practice of infectious disease, vol 2.7th ed. philadelphia: churchill livingstone; 2010. pp. 2643-53.
- 14. Bose s, ghosh ka &barapatre r. Prevalence of drug resistance among enterococcus species isolated from a tertiary care hospital. International journal of medical and health sciences. 2012; 1(3): 38-44.
- 15. Barros m, martinelli r, rocha h. enterococcal urinary tract infections in a university hospital:clinical studies. The braz j infects dis. 2009; 13(4): 294-6.
- Miskeen pa, deodhar l. antimicrobial susceptibility pattern of enterococcus species from urinary tract infections. j assoc physicians india. 2002 march; 50: 75-8.
- Srivastava et.al .prevalence and antimicrobial susceptibility of enterococcus species isolated from different clinical samples in a tertiary care hospital of north India: national journal of medical research. 2013; 03(04):389-391.
- 18. Amatya R, Jha B, Shrestha R, Adhikari RP, Timilsina S. Prevalence of high level gentamicin and vancomycin resistance among enterococci from a tertiary care hospital in central Nepal. Nepal Med Coll J. 2014; 16 (2-4): 125-7.