

FREQUENCY AND ANTIMICROBIAL RESISTANT PATTERN OF MDR ENTEROCOCCUS ISOLATED FROM CLINICAL SAMPLES AT MARDAN MEDICAL COMPLEX

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Abstract

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Introduction: Enterococcus species have recently become the major nosocomial pathogen exhibit resistance to various antimicrobials, particularly to fluoroquinolone and sulfonamide.

Aim: The study aimed to determine the frequency and antibiotic resistance pattern of *Enterococcus*, bring awareness, propose further study on antibiotic resistance and treatment strategy.

Material and Methods: The study was conducted at Microbiology department of Mardan Medical Complex from July to December in 2024. The samples, included blood, urine, and pus, were aseptically taken from patients with pyogenic infections, septicemia and urinary tract infections (UTIs). Culture and antibiotic sensitivity tests were conducted in accordance with accepted guidelines.

Result: A total of 1317 clinical sample were processed out of these 37 were *Enterococcus* species from July to December their frequency rate of infection were 2.8%. Among the patients highest ratio were observed in female

28 (75.7%), male 9 (24.3%) as shown in Table 1. Highest ratio were observed in middle age 21 to 40 years of people 15 (40.5%), followed by 41 to 60 years 13 (35.1%) and age between 1 to 20 year were 4 (10.8%), same result were observed in age between 61 to 80 and 1 isolate is found in age >80 (2.7%) as shown an Table 1, isolates were sensitive to linezolid and resistant to fluoroquinolone and sulfonamide.

1. Introduction

Enterococcus are Gram-positive, ovoid-shaped cocci that form short chains or pairs. Despite being natural flora of the digestive tract, oral cavity, and vagina, they have evolved as nosocomial pathogens. Enterococcus is a global issue that contributes significantly to morbidity and mortality worldwide. They are a growing threat due to their resistance to antimicrobials such as β lactam antibiotics, aminoglycosides, and glycopeptides like vancomycin. They can cause serious infections such as endocarditis, bacteremia, and urinary tract infections (1). Enterococci which is normal flora of GIT tract, are also leading cause of hospital-acquired infections because due to their opportunistic pathogenicity (2). Enterococci is the second leading cause of UTI and the third leading cause of bacteremia in hospital-acquired infections. Between 2009 and 2010, Enterococci

were the second most common cause of nosocomial infection, as reported by the National Healthcare Safety Network. According to the report, enterococci made up 14% of all bacteria, second only to *S. Aureus* (16%), and 3% of those Enterococci were resistant to vancomycin(3). They have a remarkable capacity to develop resistance to other antibacterial, such as high-level of resistance to aminoglycosides and glycopeptides, and a notable ability to adapt to exposure to antibacterial while retaining intrinsic resistance to penicillin and low-level resistance to aminoglycosides(4). The most often isolated pathogen is *Enterococcus faecalis* which is followed by *E. faecium*. Because antimicrobial susceptibility varies by species to species identification is helpful for both clinical decision-making, especially when it comes to therapy, and epidemiological study of an outbreak (5). According to the 2018 national study for antimicrobial resistance in Europe, the percentage of vancomycin-resistant *Enterococcus* species in various nations increased from 10.4% in 2014 to 14.9% in 2017(6). The misuse and overuse of antibiotics has been a major contributor to the development of antimicrobial resistance. Antibiotic resistance is expected to spread not only in hospitals, but also in communities (7). Infections caused by Enterococci species were traditionally treated with a combination of cell wall active agents (e.g., penicillin) and aminoglycosides (streptomycin). However, this combination therapy failed due to aminoglycoside and vancomycin resistance (8). The antimicrobial susceptibility patterns and concurrent resistance to chosen antibiotics were determined using the disk diffusion method in accordance with CLSI standards (9).

2. Materials and Methods

2.1. Samples Collection

~~The study was a descriptive cross-sectional one on frequency and antibiotic resistance among *Enterococcus* spp.~~ strains. The data of this study were collected from July to December of 2024 at Mardan Medical Complex Mardan. Clinical samples were collected from patients diagnosed with infection suspected to be caused by enterococcus at the Mardan Medical Complex. These specimens included urine, pus, blood, sputum and other relevant biological samples.

2.2. Identification of Bacteria

Specimens were inoculated on selective medium, such as blood agar, MSA, and bile esculin agar, upon arrival at the laboratory in order to isolate *Enterococcus*. Following 18 to 24 hours of incubation at 37°C, the plates were examined using morphology, Gram staining, and biochemical tests such as bile esculin hydrolysis, catalase, and growth in 6.5% NaCl to identify typical *Enterococcus* colonies.

2.3. Antimicrobial Susceptibility Testing

The Kirby-Bauer disk diffusion method was used to identify the pattern of antibiotic resistance in *Enterococcus*. Amikacin, Clindamycin, Fosmycin, Fusidic Acid, Linezolid Nitrofurantoin, Penicillin, Vancomycin, Cephrodine, Chloramphenicol, Ciprofloxacin, and Norfloxacin Trime Sulphmethoxazole are examples of common antibiotic discs were employed to evaluate patterns of resistance. The Clinical and Laboratory Standards

Institute recommendations were followed in the measurement and interpretation of zones of inhibition.

2.4. Statistical Analysis

Data were entered, cleaned, and confirmed in MS Office Excel® 2016. After that, the data was exported to SPSS version 27. The tables and figures were created using Microsoft Word and Excel. Enterococcus isolates were analyzed, and the findings are based on descriptive statistics. The data are displayed as percentages, and frequencies.

3. Results

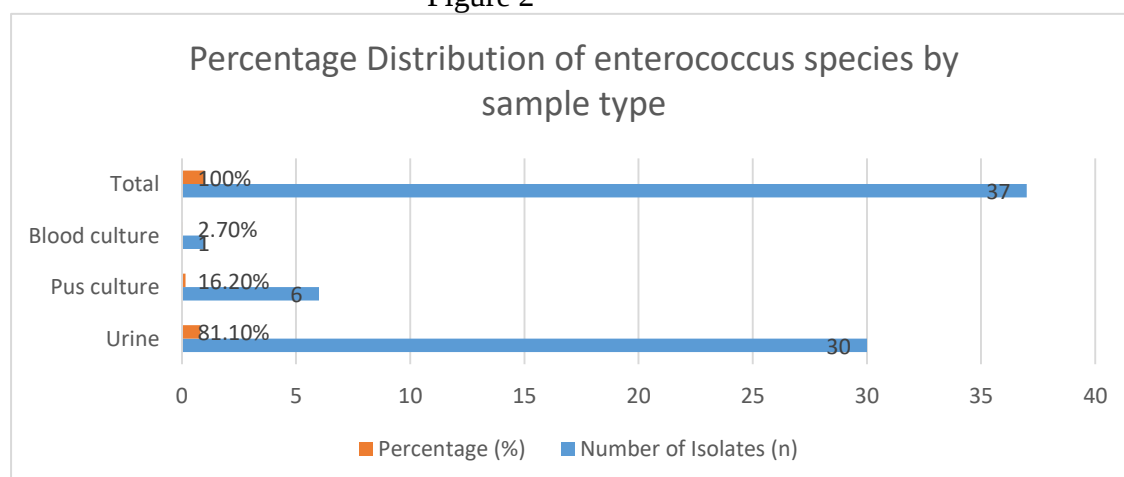
A total 1317 clinical sample were processed out of these 37 were *Enterococcus* species from July to December their prevalence rate of infection were 2.8%. Among the patients who were part of this study highest ratio were observed in female 28(75.7%), male 9(24.3%) as shown in Table 1. Result based on age, highest ratio were observed in middle age 21 to 40 years people 15 (40.5%), followed by 41 to 60 years 13 (35.1%) and age between 1 to 20 year were 4(10.8%), same result were observed in age between 61 to 80 and 1 isolate is found in age >80 (2.7%) as shown an Table 1.

Table 1

Demographics		Values, n %
Gender	Male	9(24.30)
	Female	28(75.7)
Age in years	1 to 20	4(10.8)
	21 to 40	15(40.5)
	41 to 60	13(35.1)
	61 to 80	4(10.8)
	>80	1(2.7)

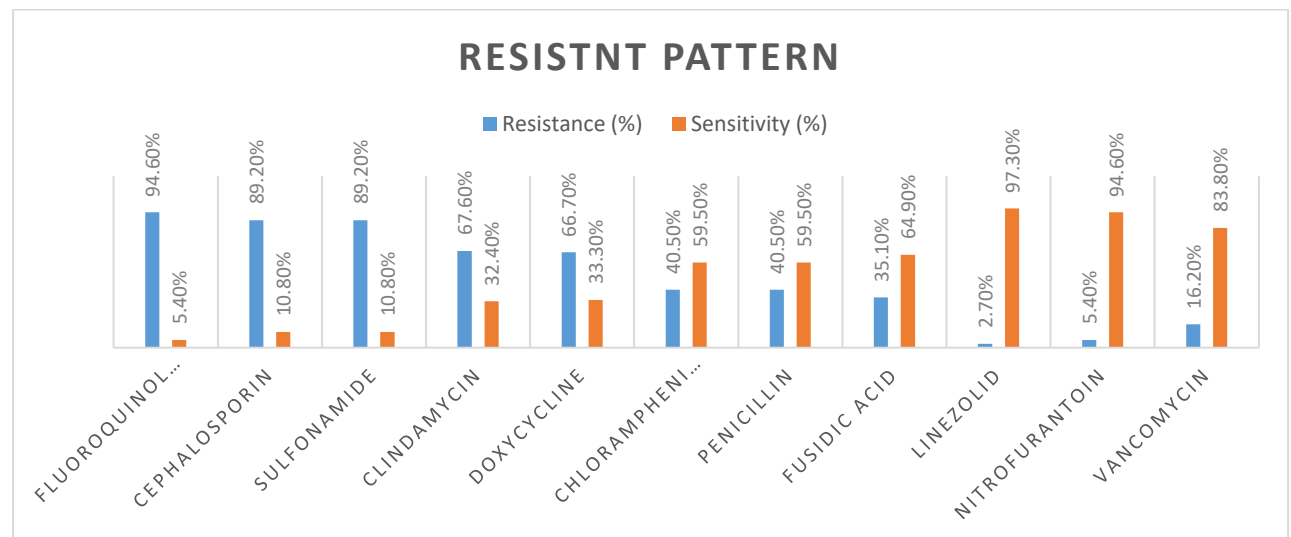
Bacterial species, isolated from different samples: urine 30(81.1%), pus culture 6(16.2%) and blood culture 1(2.1%) (Figure 2).

Figure 2



Enterococcus faecalis showed high resistant to Fluoroquinolone which was 94.6% followed by Cephalosporin and Sulfonamide 89.2% increase resistant to Clindamycin 67.6% Doxycycline 66.7% Chloramphenicol and Penicillin 40.5% and Fusidic acid 35.1%. While it was highly sensitive to Linezolid 97.3% Nitrofurantoin 94.6%

Vancomycin and Formycin 83.8% Amikacin 67.6%. These isolates show MDR pattern. Figure (3)



4. Discussion and Recommendations

This study was conducted at Mardan Medical Complex from July to December 2024. *Enterococcus* species are a major cause of nosocomial infections in hospitals, spreading easily among healthcare personnel and patients (6). Common antibiotic resistance has grown over the last 20 years as a result of overuse of antibiotics (50). *Enterococci* were present in 2.8% of the various clinical samples in our investigation. This prevalence rate was in line with the results of other writers who discovered that the prevalence rates in Asia-Pacific, Bangladesh, India, and Egypt were 3.3%, 3.2%, and 2.3%, respectively (3.6%) (50). In our study isolates were highest from urine 30(81.1%), followed by pus 6(16.2%) and blood 1(2.1%) (Figure 2). Most of the studies done on *Enterococci* support the same findings as isolates were highest in urine 80% followed by pus and blood *Enterococci* identified as the most frequent uropathogen (51). All *Enterococcus* specie isolates, in the present study, were sensitive to linezolid 97.3%, nitrofurantoin 94.6%, vancomycin 83.8% and fosmycin 83.8% This outcome was consistent with research conducted in Bangladesh and India that found no enterococci isolates to be linezolidresistant (52). In present study isolates were highest in female 28(75.7%) followed by male 9(24.3%) as shown an Table 1. Result were also estimated on the bases of age isolates were highest in the middle age between 21 to 40 year people 15(40.5%), followed by age between 41 to 60 year 13(35.1%) and followed by age between 1 to 20 year 4(10.8%) and same number of isolates were found in age between 61 to 80 and 1 isolate is found in age >80 (2.7%) Table 1. This is an arrangement with the similar study showed majority of isolates in the age group 31-50 years (53). In our study *Enterococcus faecalis* showed high resistant to Fluoroquinolone which is 94.6% followed by Cephalosporin and Sulfonamide 89.2% and increase resistant to Clindamycin 67.6% this study was in agreement with study in Pakistan for the last few years, *Enterococcus* has shown resistance against cell wall inhibitors and nucleic acid inhibitors, e.g., sulfonamide and fluoroquinolones (54). XDR is non-susceptibility to at least one agent in all but two or fewer antimicrobial classes, whereas MDR is non-susceptibility to at least one agent in three or more antimicrobial classes (32). Our study's findings indicate that 100% of *E. faecalis* and *E. faecium* strains were multidrug resistant (MDR), which is fairly consistent with 90.5% of MDR isolates (55)

5. Conclusion

This study demonstrates a significant burden of multidrug-resistant (MDR) *Enterococcus* isolates among clinical samples at Mardan Medical Complex, reflecting an emerging challenge for effective antimicrobial therapy. The isolates exhibited **marked resistance to Fluoroquinolones, Cephalosporins, and Sulfonamides**, indicating the reduced usefulness of these antibiotic classes for empirical treatment. However, the study also identified **high sensitivity to Linezolid, Nitrofurantoin, Fosfomycin, and Vancomycin**, suggesting that these agents remain the most reliable treatment options and can be considered drugs of choice for enterococcal infections in this setting. Overall, the findings emphasize the need for continuous surveillance, strict antibiotic stewardship, and updated local treatment guidelines to limit the spread of MDR *Enterococcus* and preserve the effectiveness of available therapeutic options.

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