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Research article

Isolation, Identification, Speciation, and Antibiogram of Enterococcus species by conventional methods and Assessment of the Prevalence of vana genotype among VRE

Ashfaq Ahmad Shah[1], Syed Khursheed[2], Aadil Rashid[3], Amit Gupta[1]*

[1]Department of Life Sciences (Microbiology), Graphic Era (Deemed to be) University, Dehradun, India
[2]Department of Microbiology, SKIMS Medical College & Hospital, Srinagar, India
[3]Department of MLT, Sai Institute of Paramedical & Allied Sciences, Dehradun, India

ABSTRACT

Enterococci are Gram-positive, ovoid cocci that can be found as diplococci or in short chains. These species are found in the typical microbiota of the intestine, oral cavity, vagina, and other areas. Despite being commensals with moderate virulence, these species have lately emerged as important nosocomial infections with increasing drug resistance particularly to vancomycin. This has resulted in their changing patterns of infection that are resistant to conventional antimicrobial therapy. The main objective of this study was to assess the frequency of the VanA genotype among Vancomycin Resistant Enterococci, as well as the isolation, identification, speciation, and antibiotic sensitivity pattern of Enterococci from various clinical specimens**.** In this six-month study, 500 different clinical specimens such as urine, blood, and pus were collected aseptically from patients suffering from urinary tract infection (UTI), septicemia, and pyogenic illnesses, and a total of 94 Enterococci strains were identified. These isolates were identified and speciated using traditional tests and biochemical processes. Following culture, the disc diffusion technique was used to determine their antibiotic susceptibility pattern, as suggested for common antibiotics. RCR was also used to evaluate the prevalence of the vanA gene among VRE isolates. 88.29% were *E. faecalis*, 7.88% were *E. faecium*, 2.12% were *E. durans*, 1.06 % was *E. casseliflavus*, and 1.06 % was *E. gallinarum*, out of a total of 94 isolates. Urine (70.21 %) yielded the most isolates, followed by blood (17.02 %) and pus (17.02 %). Female patients had a higher isolation rate of 73.40 % (69/94) than male patients, who had a rate of 26.59 % (25/94), and the majority of the Enterococcal isolates were from inpatients (62.77 %) rather than outpatients (37.23 %). 30 % of the isolates were hemolytic, whereas 70% were non-hemolytic. The isolates sensitivity patterns revealed that they were resistant to antibiotics such as ampicillin, ciprofloxacin, and gentamicin**.** The susceptibility test also included 30g discs of vancomycin. Vancomycin resistance is greatest in *E. faecium* (28 %) 6/83, followed by *E. faecalis* (8 %) 2\7. All other isolates were vancomycin-susceptible. Vancomycin-resistant isolates made up 8.51 % of the total. Barely urine isolates were tested for nitrofurantoin resistance, which was found to be only 5%. Linezolid sensitivity was found in all of the Enterococci. According to the PCR test for the vanA gene, among total VRE isolates 5 *E. faecalis* and 1 *E. faecium* isolates had vanA genotypes with bands of 473bp. A number of studies have revealed an increment in the infection rate and antimicrobial resistance of Enterococci. Elevated isolation rate of VRE has posed threat to therapeutic strategies as now only few antibiotics are susceptible to such isolates. It is thus imperative to execute measures that could stable the VRE augmentation lest a normal commensal would get the status of SARS-CoV-2.

Keywords: Vancomycin resistant *Enterococci*, VanA genotype, Antimicrobial resistance, Antibiotic susceptibility pattern.

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***Correspondence:** Dr. Amit Gupta ✉ dr.amitgupta.bt@geu.ac.in **Orcid Id**: https://orcid.org/0000-0002-2003-8618

Department of Life Sciences (Microbiology), Graphic Era (Deemed to be University), Dehradun, Uttarakhand, India

INTRODUCTION

Enterococci belongs to the phylum Firmicutes, which is a vast genus of lactic acid bacteria, and is found as intrinsic microbiota in the human/animal intestinal system, oral cavity, and also reported in genitourinary tract [1]. They were previously called as faeces *Streptococci*, and this genus was categorized as group D *Streptococcus* until 1984. It poses a difficulty for a researcher to distinguish them from *streptococci* on physical characteristics solely [2]. Later genomic DNA analysis revealed that for its members a discrete genus classification would be pertinent [3]. *Enterococci* are gram positive, ovoid shaped cocci that mostly occur in short chains and pairs (diplococci). They are facultative anaerobic microorganisms losing the capability of spore formation. However,

they can tolerate relatively extreme environmental conditions like high salt concentrations (positive for salt tolerance test), high temperature (5-65^{0}C), and pH (4-10.0) [4]. Inspite of being avirulent in healthy individuals, members of this genus have recently emerged as nosocomial pathogens, significantly contributing to morbidity and mortality in hospitalized patients. Keeping an eye on the Enterococcal infection patterns has become urgent because of their increasing potential to cause fatal, refractory to treat infections (endocarditis, urinary tract, bacteremia, intraabdominal infections, wound infections, and meningitis) [5].

According to CDC survey of nosocomial infections, next to *Escherichia coli*, Enterococci accounts for 14% of nosocomial UTI's as well as are third most causative agents of nosocomial bacteremia [6]. Among the members of *Enterococci* genus, *E. faecalis* reported 80-90 % of human infections followed by *E. faecium* 5-10 % [7]. However, recent studies are now revealing an enhanced isolation rate of *E. faecium* and other *Enterococcus* species from different clinical samples. So, these *enterococcal* infections are traditionally treated with cell wall active antibiotics (e.g., penicillin or ampicillin) in combination with an aminoglycoside like streptomycin or gentamycin. However, in this day and age emergence in high-level aminoglycoside resistance (HLAR, intrinsic and acquired), resistance to cephalosporins of several generations and most importantly resistance to glycopeptide Vancomycin (VRE) have rendered them resistant to synergistic effects of combinational therapies [8]. VRE has recently arisen as a challenge to therapeutic methods, as infections are growing resistant to therapy and have a high death rate. The CDC has assigned multidrug-resistant *Enterococci* the same priority as MRSA and ESBL nosocomial infections. Due to its ability to survive and spread in a medical setting is largely due to inherent resistance against commonly used antibiotics, as well as their ability to acquire some resistance which is mainly through mutation or horizontal transfer of pathogenicity islands or genetic elements containing genes that contribute to resistance or code for virulence factors [9]. Enterococci's (VanA, VanB, VanC, and VanD) genes are well recognized to move between species and to other Gram-positive bacteria, making nosocomial infections even more difficult to treat [10, 11]. In order to reduce VRE-related mortality and morbidity, infection management methods should be developed for use in both the hospital and the community. As a result, tertiary care facilities must exercise extreme vigilance, and laboratory procedures to isolate identify, and speciate Enterococci as a first step toward understanding their role in nosocomial infections must be developed. In light of the foregoing, the current study was conducted in a tertiary care hospital in Srinagar India, to determine the frequency of Vancomycin Resistant Enterococci and their antibiotic susceptibility patterns in this region. The major objectives of this study were the isolation and identification of Enterococci from clinical specimens obtained from suspicious individuals; characterization of Enterococci (100 isolates) to the species level using established methods, determining the antibiotic susceptibility pattern of these clinical isolates, and using a polymerase chain reaction (PCR) assay for the VanA gene, determining the frequency of VanA genotype among Vancomycin Resistant Enterococci (VRE).

MATERIALS AND METHODS

Collection of samples

Over the course of six months, 500 different clinical specimens were investigated in the Microbiology department, SKIMS Medical College, and collect Enterococci strains (n=94) were isolated.

Inclusion criteria

Several Different samples like urine, pus, blood was screened for presence of *Enterococci*.

Exclusion criteria

Stool, oral throat swab, sputum, and other samples where they are present as normal commensals were excluded.

Laboratory procedure

Urine, pus, and blood was collected aseptically and processed for culture and sensitivity according to standard procedures. These isolates were screened and speciated using traditional tests including biochemical processes. The antibiotic susceptibility pattern for common antibiotics was established according to the recommendations. The frequency of VanA genotype among Vancomycin Resistant *Enterococci* was investigated using PCR test for VanA gene. All of the specimens that were brought in were subjected to the following tests.

Direct microscopy

In the smears, gram staining was performed to check the presence pus cells including gram-positive cocci (pairs and short chains) [10].

Culture

According to the package instructions, specimens were inoculated on agar (blood, chocolate agar, and MacConkey) media. For semiquantitative urine culture, Cystine lactose electrolyte deficient (CLED) medium was administered [9, 10]. The inoculation medium is incubated at 37^0 C overnight and checked for following characteristics.

- Occurrence of tiny transparent colonies with, α, β or no hemolysis on Blood agar [11].
- Occurrence of tiny grey translucent colonies on Chocolate agar [12].
- Occurrence of deep pink magenta-colored colonies on MacConkey agar [13].
- Small yellow colonies on CLED media [10].

Enterococci were presumptively identified using colony

smear and performing Gram's stain, Catalase test [14], Bile aesculin test [15], Salt tolerance test [15], and Heat tolerance test [16], in which Enterococci come out to be Catalase negative, hydrolyze aesculin to aesculetin, tolerate a salt concentration of 6.5 percent, and tolerate a temperature of 60^0 C for 30 minutes.

Colony smear

A smear of the colonies was prepared, and gram staining method was applied and determining gram-positive cocci i.e., in pairs and short chain along with controls [15].

PC: *Staphylococci* ATCC 25923

NC: *Escherichia coli* ATCC 25922

The colonies were subcultured onto nutrient agar for further biochemical testing.

Catalase test

A part of a colony was taken and dipped in a test tube with 3% hydrogen peroxide using a small, sealed capillary tube. Controls were also subjected to this test [10]. The quick and continuous effervescence indicates a positive test (nascent oxygen)

PC: *Staphylococcus aureus* ATCC 25923

NC: *Enterococcus faecalis* ATCC 29212

Enterococci come out to be catalase negative mostly but in some cases pseudo catalase positive [16].

Bile esculin test

Clinical isolates were streaked on bile esculin agar and kept at 37^0 C for 24 to 48 h. The presence of black pigmented colonies (Figure 2) indicates a positive test [16]. Enterococci convert esculin to esculetin, which interacts with ferric ions to produce black colonies.

PC: *Enterococcus faecalis* ATCC 292 I2

NC: *Streptococcus viridians*

Heat tolerance assay

Inoculating the suspected *Enterococcal* isolates, as well as the control strains, into BHI broth and incubating them at 60°C in a water bath for 30 minutes and was used to assess heat tolerance. Before incubation, the broth was subcultured on blood agar and MacConkey agar, as well as after incubation at 10 min, 20 min and 30 min intervals. As a positive control, *E. faecalis* ATCC 29212 was employed and take reading of the other isolates and its growth was verified. Both the broth (before and after heating) at 60°C for 30 minutes, the ATCC control strain thrived. Heat tolerant isolates were those that grew before and after 30 minutes of heat exposure. (Figure 3).

Salt tolerance test

Clinical isolates (1-2 colonies) samples were inoculated in brain hear infusion broth containing 6.5% NaCl and the indicator bromothymol blue (0.002%) and incubated at 37^0 C for 18-24 h. The broth thickens and becomes yellow when a positive test result is obtained [17].

PC: *Enterococcus faecalis* ATCC 292 I2

NC: *Streptococcus bovis*

Figure 1: Distribution of vanA genotype among the VRE isolates

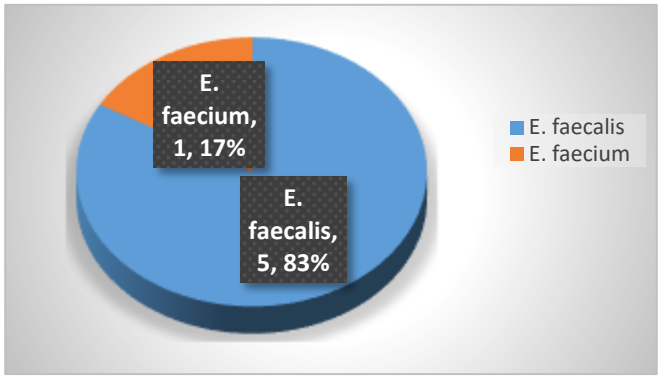


Figure 2: Blackish discoloration of medium- hydrolysis of esculin on be agar.

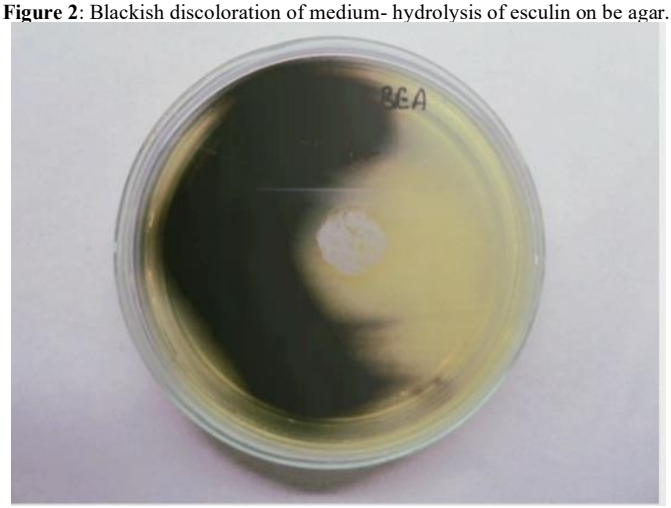
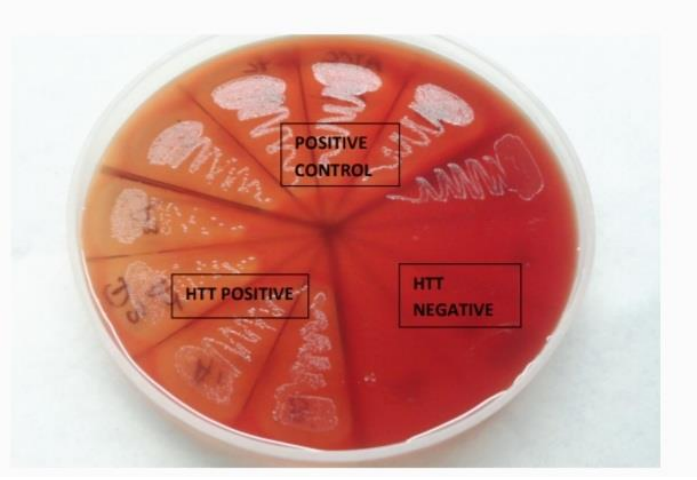


Figure 3: Heat tolerance test



PYR Test

Colonies from the isolate were inoculated into PYR broth (pyrrolidone- naphthylamide) and incubated at 37^0 C for 30 min. In the broth, one drop of PYR regent (p-Dimethylaminocinnamaldehyde) was added. The emergence of cherry red is an indication of positive test [19]. Enterococci come out to be PYR-positive

PC: *Enterococcus faecalis* ATCC 29212

NC: *Streptococcus agalactiae*

Speciation

Motility, Voges-Proskauer (VP) test, Arginine hydrolysis Sugar fermentation and tellurite reduction assays were used to speciate Enterococci (Figure 4) [20]. Table 1 shows how the results were interpreted.

Antibiogram

The antibiotic susceptibility of the isolated strain was determined using Kirby-Bauer disc diffusion technique and compared with established standards. To ensure the quality of the susceptibility testing, ATCC strains (*S. aureus, E. faecalis, E. coli,* and *Pseudomonas aeruginosa*) were utilized [20] (Figure 5).

Method

A lawn culture of *Enterococci* 0.5 McFarland standard was conducted on 5 percent Muller Hinton agar. Antibiotics were applied at the ideal distances and Petri plates incubated for 18 to 24 h at 37 ^{0} C. CLSI guidelines were used to measure and interpret zone sizes [20].

VRE detection by molecular method

The vancomycin resistance gene (VanA) was discovered in Enterococci (*E. faecium* and *E. faecalis*), using PCR method [21]. For this study, Prep Kit (The Helini Pure Fast Bacterial Genomic DNA Mini Spin) was used to extract DNA from Enterococcal isolates, which was then submitted to PCR and gel electrophoresis to confirm the gene product. As per primers incorporated the VanA gene is represented by a 473bp PCR product (Figure 6).

RESULTS

From a total of 500 clinical specimens, 94 Enterococci strains were identified. The overall incidence was found to be around 18% (88.29% isolates were *E. faecalis*, 7.88% *E. faecium*, 2.12% *E. durans*, 1.06 % *E. casseliflavus*, and 1.06 % *E. gallinarum*), out of a total of 94 isolates. The isolates came from urine (70.21%), pus (12.76%), and blood (17.02%). (Table 2). Thirty percent of the isolates were hemolytic, while seventy percent were non-hemolytic. Urinary isolates had the highest hemolytic activity (56.48 %), followed by pus group followed by 25.53% in 21-40yrs, 30.65% in 41-60 yrs. and 15.95% in 61 yrs. and above (Table 3). Males form 26.59% of the isolates and females 73.40%. A higher isolation rate (69/94) was observed among the female patients than male patients (23/94). Among females, the most enterococcus species were isolated from urine **(**Table 4). Among the isolates 27.65% are from 0-20 yrs. age group followed by 25.53% in 21-40yrs, 30.65% in 41-60 yrs. and 15.95% in 61 yrs. and above **(**Table 5**).** In pediatric age group 11.70% were neonates (less than 1 month) and 8.51% from 1 month to 12 yrs. In age group of 0 to 1-month Blood stream infection is more common while as in above 1 month it is Urinary tract infection because of Enterococcus **(**Table 6**).** The majority of the Enterococcal isolates in our investigation came from inpatients 62.77% than from outpatients 37.23%. Among 94 isolates, 59 isolates were from inpatients and 35 isolates were from outpatients.

Pattern of antibiotic susceptibility of enterococcal species (Kirby-Bauer disc diffusion method)

As indicated in **Table 7**, the isolates were resistant to a variety of antibiotics. Vancomycin 30µg discs were also used in the susceptibility test. *E. faecium* show highest resistance to Vancomycin (28%) 6/83 followed by *E. faecalis* (8%) 2\7. Total percentage of vancomycin resistant isolates was 8.51%. Same technique was applied to determine the pattern of antibiotic sensitivity of isolates to supplementary antimicrobials. Nitrofurantoin was used in only urine isolates and showed only 5% resistance. However, no isolate showed resistance against Linezolid (Table 8).

Detection of VRE using molecular methods

A PCR technique was used to determine the presence of the VanA gene in all eight vancomycin-resistant Enterococcal isolates, and the following results were obtained (Table 9)

According to the VanA gene PCR assay, 5 *E. faecalis* and 1 *E. faecium* isolates were with VanA genotype with 473bp bands. In the remaining two VRE isolates, no band corresponding to the VanA gene was found. The VanA genotype is found in 6.4 % of VRE isolates, according to our findings.

Figure 4: Biochemical reactions of *E. faecium* and *E. faecalis*

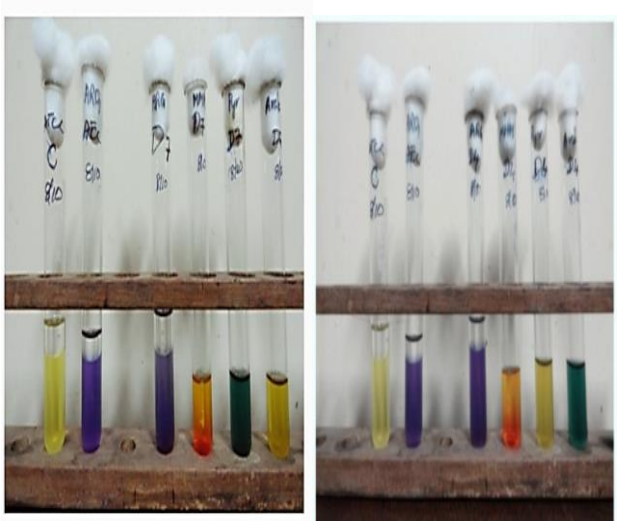


Figure 5: Antibiotic susceptibility pattern

Figure 6: PCR product-Agarose gel image, E+ positive control VanA, E1-E6 test isolates showing band of 473 bp.

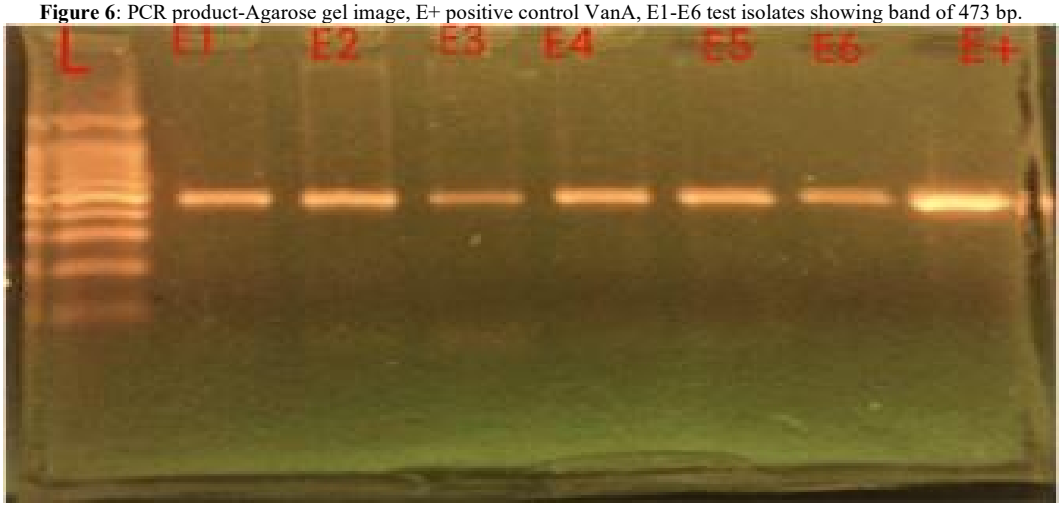


Table 1: Speciation of *Enterococci*

3	Arginine Hydrolysis	Motility	VPT	PTR	Lactose	Raffinose	Mannitol	Sorbitol	Sorbose	Arabinose	Sucr ose
E. faecium	+	-	-	-	+	-	+	+	-	+	NA
E. faecalis	+	-	-	+	+	-	+	+	-	-	NA
E. avium	-	-	-	-	NA	-	+	+	+	+	-
E. durans	+	-	-	-	-	-	-	-	-	NA	-
E. mundtii	+	-	+	-	+	NA	+	+	-	+	-
E. pseudovarium	-	-	+	ND	NA	-	+	+	+	-	-
E. solitarius	+	-	-	ND	-	-	+	+	-	-	NA
E. malodaratus	-	-	-	-	-	+	+	+	+	-	NA
E. casseliflavus	+	+	+	+	+	NA	+	-	-	+	NA
E. hirae	+	-	+	-	NA	-	-	-	-	-	NA
E. gallinarum	+	+	-	+	+	-	+	NA	-	+	-
ND- Not Detected; NA- Not Applicable; VPT- Voges Prausker's test; PTR-Potassium Tellurite Reduction; "+"- Sugar Fermented; "-"- Sugar Not Fermented											

Table 2: Isolates of Enterococci from different clinical samples

SPECIMEN	*E. faecalis*	E. *faecium*	*E. durans*	*E. casseliflavus*	*E. gallinarum*	Total%
URINE	60	5	1	0	0	66 (70.21%)
PUS	9	2	0	0	1	12 (12.76%)
BLOOD	14	0	1	1	0	16 (17.02%)
TOTAL%	**83 (88.29%)**	**7 (7.44%)**	**2 (2.12%)**	**1 (1.06 %)**	**1 (1.06 %)**	**94**

Table 3: β hemolytic and non-hemolytic isolates

SPECIMEN	B-hemolytic	Non-hemolytic
URINE	18 (19%)	48
PUS	8 (8.51%)	4
BLOOD	2 (2.13%)	14
	28 (30%)	66 (70%)

Table 4: Sex distribution among isolates

Sex	URINE	PUS	BLOOD	TOTAL
MALES	11	7	7	25 (26.59%)
FEMALES	55	5	9	69 (73.40%)

Table 5: Age distribution among different isolates

AGE GROUP	URINE	PUS	BLOOD	TOTAL%
O-20 YRS	12 (46.15%)	2 (7.69 %)	12 (46.15 %)	26 (27.65%)
21-40 YRS	21 (22.34%)	3 (3.19%)	0 (O%)	24 (25.53%)
41-60 YRS	22 (23.40%)	5 (6.66%)	2 (0%)	29 (30.65%)
61 YRS & ABOVE	11 (11.70%)	2 (33.34%)	2 (12.5%)	15 (15.95%)
TOTAL	**66 (70.21%)**	**12 (12.76%)**	**16 (17.02%)**	**94**

Table 6: Distribution of Enterococcal infection in pediatric age group

AGE GROUP	URINE	PUS	BLOOD	TOTAL
O-1 MONTH	2	0	9	11 (11.70%)
1MONTH-12 Yyars	5	1	2	8 (8.51%)
TOTAL %	**7**	**1**	**11**	**19 (20.21%)**

Table 7: Antibiotic sensitivity pattern of Enterococcus species from various specimen

Enterococcal species	Total no.	P (10 µg)		AMP (10 µg)		AMX (25 µg)		CIP (5µg)		OF (5µg)		LE (5µg)		AK (30µg)		AMC (20+ 10µg)		VA (30µg)	
		S%	R%	S%	R%	S%	R%	S %	R%	S %	R%	S %	R%	S %	R %	S %	R %	S%	R%
E. faecalis	83 (88.29 %)	20	80	80	20	21	79	20	80	12	88	10	90	38	62	15	85	92	8
E. *faecium*	7 (7.44%)	15	85	21	79	33	67	22	78	17	83	27	73	36	64	27	73	72	28
E. durans	2 (2.12%)	40	60	90	10	20	80	15	85	30	70	11	89	31	69	39	61	100	0
E. casseliflavus	1 (1.06 %)	38	62	94	6	24	76	23	77	23	77	14	86	28	72	25	75	100	0
E. gallinarum	1 (1.06 %)	55	45	100	0	30	70	21	79	19	81	16	84	40	60	33	67	100	0
P- PENICILLIN, AMP- AMPICILLIN, AMX-AMOXICILLIN, CIP- CIPROFLOXACIN, OF- OFLOXACIN, LE-LEVOFLOXACIN, AK-AMIKACIN, AMC-AMOXYCLAV, VA- VANCOMYCIN																			

Table 8: Antibiotic susceptibility pattern of isolates to supplemental drugs

	S%	R %	S%	R %	S%	R %	S %	R %	S %	R %	S %	R %	S %	R %	S%	R%
URINE (66)	100	0	85	15	0	100	11	89	11	89	10	90	20	80	98	0
PUS (12)	100	0	82	18	21	79	22	78	27	73	33	67	40	60	-	-
BLOOD (16)	100	0	73	27	25	75	24	76	25	75	12	88	36	64	-	-
LZ- LINEZOLID, TEI- TEICOPLANIN, TC- TETRACYCLINE, IPM- IMIPENEM, PIT- PIPERACILLIN, COT-TRIMETHOPRIM+ SULFAMETHOXAZOLE, GEN- GENTAMYCIN, NIT-NITROFURANTOIN																

Table 9: Distribution of VanA Genotype in the VRE isolates

VRE ISOLATES n-8	TOTAL ISOLATES TESTED	VanA GE NOTYPE	
		PRESENT	ABSENT
E. faecalis	6	5 (83%)	1 (17%)
E. faecium	2	1 (50%)	1 (50%)
TOTAL	8	6 (75%)	2 (25%)

DISCUSSION

Enterococci are rapidly becoming one of the most common causes of nosocomial infections in hospitals, as well as opportunistic infections in immunocompromised people [21]. Endocarditis, bloodstream infections, urinary tract infections, surgical site infections and wound infections are potentially life-threatening illnesses they can cause. [22] Their capacity to survive in harsh environments, as well as their natural and acquired resistance to a range of antibiotics, makes them a troublesome infection to treat with a high risk of death and morbidity [23]. The situation has worsened with the introduction of vancomycin-resistant Enterococci (VRE), leaving very few alternatives for selecting antibiotics to treat this multidrug-resistant pathogen.

As a result, it's critical to catch them early and provide appropriate treatment depending on antimicrobial susceptibility patterns. In this context, the purpose of our study was to see how common antibiotic resistance was among clinical isolates of *Enterococci* collected from patients in the area [23].

A total of 94 Enterococci strains were identified from 500 clinical specimens in this study. The overall rate of occurrence was estimated to be around 18%. Out of a total of 94 isolates, 88.29 percent were *E. faecalis*, 7.88 percent were *E. faecium*, 2.12 percent were *E. durans*, 1.06 percent were E. casseliflavus, and 1.06 percent were *E. gallinarum.* The isolates came from urine (70.21 %), pus (12.76 %), and blood (17.02 %). Our data are in harmony with previous studies and support the hypothesis that *E. faecalis* is the

most common Enterococcal isolate [18].

30% of the isolates were hemolytic, while 70% were non-hemolytic. Urinary isolates had the highest hemolytic activity (56.48%), followed by pus (29.33%), and blood (14.19 %). Female patients were shown to have a higher isolation rate (69/94) than male patients (23/94). Females had the highest number of enterococcus species isolated from urine, which is consistent with many other researches. Because of the small urethra, urinary tract infection is very common in women. As a result, a higher percentage is reflected here as well [17].

According to our study, among the isolates 27.65% are from 0-20 yrs. age group followed by 25.53% in 21-40 yrs., 30.65% in 41-60 yrs. and 15.95 % in 61 yrs. and above. In pediatric age group 11.70 % were neonates (less than 1 month) and 8.51% from 1 month to 12 yrs. In age group of 0 to 1-month Blood stream infection is more common while as in above 1 month it is Urinary tract infection because of Enterococci. In our study, majority of the Enterococcal isolates were from inpatients 62.77% than from outpatients 37.23%. Among 94 isolates, 59 isolates were from inpatients and 35 isolates were from outpatients. Such a result supports the generalization that Enterococci has emerged as nosocomial pathogens [22].

Antibiotic susceptibility was determined through Kirby Bauer disc diffusion method. As demonstrated in the table above, the isolates were resistant to a variety of antibiotics. Vancomycin 30 µg discs were also used in the susceptibility test. *E. faecium* show highest resistance to Vancomycin (28%) 6/83 followed by *E. faecalis* (8%) 2\7. Total percentage of vancomycin resistant isolates was 8.51%. Nitrofurantoin was used in only urine isolates and showed only 5% resistance. However, no isolate showed resistance against Linezolid. The presence of the Van A gene in all eight vancomycin-resistant Enterococcal isolates was determined using a PCR method. According to the VanA gene PCR assay, 5 *E. faecalis* and 1 *E. faecium* isolates were VanA genotype with 473 bp bands. In the remaining two VRE isolates, no band corresponding to the VanA gene was found. The VanA genotype is found in 6.4 % of VRE isolates, according to our findings. The findings of these parameters are in line with the findings of a number of studies.

CONCLUSIONS

The most prevalent isolates in this investigation were *E. faecalis* and *E. faecium*. However, recent research has demonstrated a shift in the species isolation from clinical specimens, with *E. faecium* and other species being isolated more frequently. A change in the pattern of *Enterococci* infections has occurred. Vancomycin-resistant strains are becoming more common over the world. In this study, eight of these strains were isolated. Multidrug resistance is common among Enterococci all around the world, according to more data in the literature. This suggests that the infection dynamics be evaluated continually and on a frequent basis. Controlling and preventing the spread of multidrug-resistant bacteria in hospitals necessitates a collaborative effort from multiple departments, which can only be achieved through hospital staff education, proper antimicrobial dosage administration, and early detection and reporting by laboratories and quick execution of infection control measures.

CONFLICTS OF INTEREST

There were no commercial or financial links that may be deemed a potential conflict of interest during the research, according to the authors.

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