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Prevalence And Antimicrobial Susceptibility Pattern Of enterococccs Species With Special Reference To Vancomycin Resistance enterocccus In Various Clinical Samples

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ABSTRACT

Enterococci species exists as normal microbiological flora in oral cavity, intestinal tract and vagina. In recent years Enterococcal infections have emerged as a great therapeutic challenge due to its intrinsic and acquired resistance to Vancomycin. Hence this study aims to characteriseEnterococcus species and its antimicrobial resistant patterns with special reference to Vancomycin in various clinical samples. A descriptive study was conducted in a tertiary care hospital in Chennai. A total of 110 Enterococcus was isolated from 2142 samples, out of which E.faecalis was 59% and E.faecium was 41%. Urine samples showed more Enterococcus than other samples. E.faecium showed more drug resistance than E.faecalis, there was increased resistance to Ampicillin, Ciprofloxacin and Cotrimoxazole (68.8%, 75.5% and 62.2 respectively). VRE among the isolates was 6.36%, in E.faecalis was 4.6% and E.faecium was 8.8%.Enterococcal isolates showed high susceptibility to Teicoplanin and Linezolid. Better infection control measures and judicious use of antimicrobials can reduce the spread of drug resistant Enterococcal strains.

Keywords: Enterococcus faecalis, Enterococcus faecium,Drug resistance, VRE.

INTRODUCTION

Enterococci speciesit is found to cause various infections like urinary tract infections, soft tissue infections, bacteraemia, abdominal infections, meningitis and pelvic infections[(1,2)]. It causes both community acquired and nosocomial infections[(3, 4)]. Emerging drug resistance to antimicrobials like aminoglycosides, β-lactam antimicrobials and more recently glycopeptides like Vancomycin has posed a threat to treatment strategies[(5)]. Identification and tracking the distribution of Enterococci and knowledge about their susceptibility to antimicrobials is up most important in treatment as well

as control of infection[6].Hence this study aims to know the prevalence of Enterococciin various samples and its resistance pattern.

Aims and objectives:

To determine the prevalence and antimicrobial susceptibility of Enterococcus species in different clinical sample

To determine the prevalence of Vancomycinresistant Enterococci (VRE).

MATERIALS AND METHODS

This study was conducted in a tertiary care hospital in Chennai in Tamilnadu from June 2019 to October 2020. Various sample like urine, pus, body fluids and blood were collected from different departments in all age groups, both gender and both in patients and out patients were included. All samples were processed in lab by inoculating in Blood agar, Mac Conkey agar, except urine samples which was inoculated in CLED medium.

Phenotypic identification

Gram stain, catalase test, Bile esculin test and growth at 6.5%, sugar fermentation tests of mannitol, sorbose, arabinose, sorbitol, and lactose was done for phenotypic identification of Enterococcus species [7].

Antimicrobial susceptibility testing

Antimicrobial susceptibility testingwere done for all Enterococcal isolates by Kirby Bauer disc diffusion method on Muller – Hinton agar by using inoculums with turbidity equivalent to 0.5McFarland Standard. Commercially available antimicrobial disc like Ampicillin (10 µg), Vancomycin (30µg), Teicoplanin (30µg), Erythromycin (15µg), Ciprofloxacin (5µg), Chloramphenicol (30 µg) and Linezolid (30µg), High level Gentamycin (120µg, Cotrimoxazole(1.25/23.75µg) were used along with ATCC control strains E. faecalis 29212. CLSI 2019 guidelines were used [8].

Determination of minimum inhibitory concentration

Agar dilution method was used to determine minimum inhibitory concentration (MIC) of Vancomycin. Different concentration of Vancomycin was added to Brain heart infusion agar and isolated Enterococci was grown in broth and turbidity was matched with 0.5 McFarland standards. By using 10µl of growth culture was spot inoculated. The plates were kept in incubation at 37^{o}C for 24hours.The minimum concentration of Vancomycin at which the bacterial growth was inhibited

was considered as MIC.E.faecalis ATCC29212 was used as control strains were used. MIC of susceptible ≤ 4, 8–16 intermediate and ≥ 32 resistant or VRE according to CLSI guidelines.

RESULTS

Out of 2142 culture positive samples, 110 Enterococci species were isolated with prevalence of 5.13%. Majority of isolate were from urinary specimen, followed by pus (76, 27 respectively) table1. Enterococcus faecalis species was predominant (59%) followed by Enterococcus faecium (41%). Enterococcus faecium showed higher resistance to antimicrobials than E.faecalis Table 2. High level Gentamycin resistance was 35.3% in E.faecalis and 46.6% in E.faecium. 4.6% of E.faecalis and 8.8% E.faecium showed Vancomycin resistance. Teicoplanin were sensitive in all isolates and Linezolid showed high susceptibility in both species (E.faecalis - 1.5% and E.faecalis - 2.2%).

Table 1: Distribution of Enterococcus isolates in various clinical sample

Samples	E. faecalis N(%)	E. faecium N(%)	Total
Urine	42	34	76
Blood	2	0	2
Pus	17	10	27
Other body fluids	4	1	5
Total	65 (59)	45(41)	110

Table 2: Antimicrobial resistance pattern of Enterococcal isolates

Antimicrobials	E. faecalis N (%)	E. faecium N(%)
Ampicillin	41(63)	31(68.8)
Erythromycin	36(55.38)	28(62.2)
Chloramphenicol	33(50.9)	22(48.9)
Cotrimoxazole	38(58.4)	28(62.2)
Ciprofloxacin	46(70.7)	34(75.5)
Teicoplanin	0	0
Vancomycin	3(4.6)	4 (8.8)

High level Gentamycin	23(35.38)	21(46.6)
Linezolid	1(1.5)	1(2.2)

Table 3: Distribution of Vancomycin resistant Enterococcal isolates

Isolates	Total Number(n)	VRE Number(%)
E.faecalis	65	3(4.6)
E.faecium	45	4 (8.8)
Total	110	7 (6.36%)

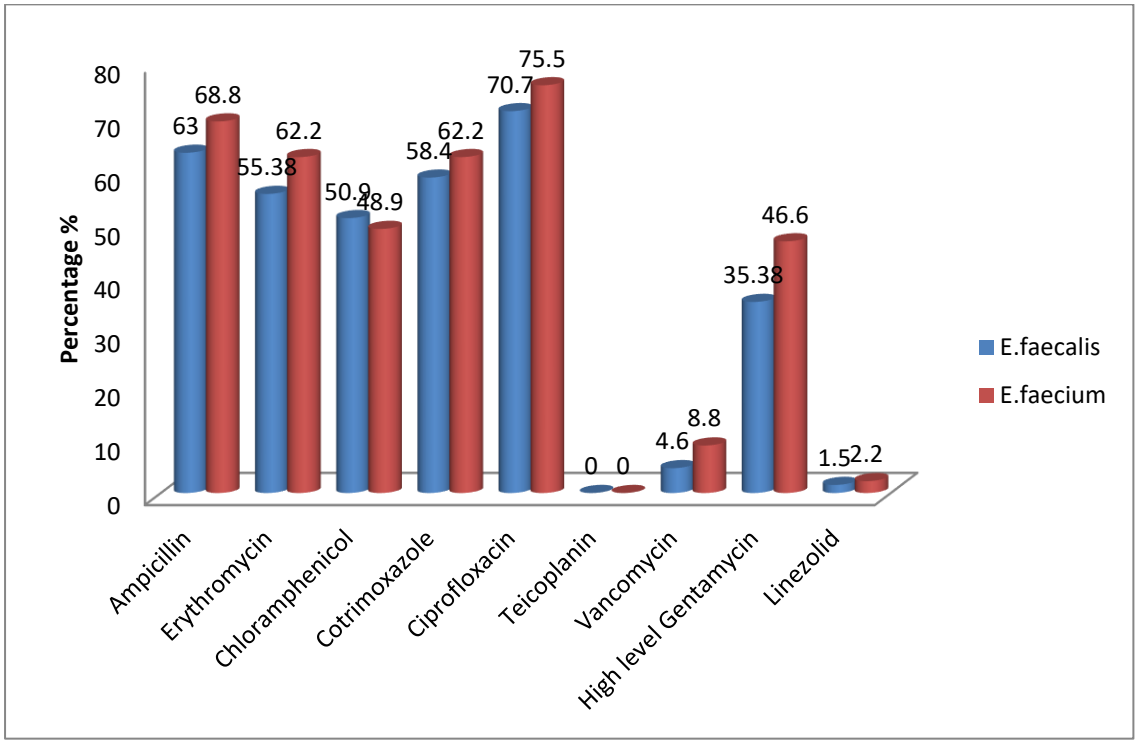

Fig 1: Antimicrobial resistance pattern of E.faecalis and E.faecium.

DISCUSSION

In recent years, there has been a increasing in Enterococcal infections worldwide and emergence of drug resistance among such isolates. This indicates the need of continuous surveillance of the bacteria [9].We found that prevalence of Enterococci was 5.13% in various samples, which was similar to study by Vidhylakshmi et al [10].Urine samples showed more isolation of Enterococci followed by pus and body fluids, this finding were concordant to study by Zavaryani et al[11] and Banerjee et

al[12].In our study,E.faecalis formed the major isolate 65(59%) followed by E.faecium 45(41%) which was similar to study by Arif et al[13]**.** Many recent studies have shown thatthere is increaseisolation rate of E.faecium and other Enterococcus species[14]. E.faecium showed increased antimicrobial resistance pattern compared to E. faecalis[15].

Resistant pattern among the Enterococcal isolates showed an increase in resistance to Ampicillin, Ciprofloxacin and Cotrimoxazole (68.1%, 76.2% and 62.4 respectively), which is similar to study by Sreeja et al[16].High level Gentamycin resistance was 35% in E.faecalis and 45% in E.faecium, the result were in concordance to study by Manimala et al[16], Sreeja et al[17]and Shah et al[18]., studies where HLGR was 34%, 47%, and53% respectively. In our study HLG resistance was greater among E.faecium than E. faecalis isolates, Mendiratta et al[19]. study also reported the same results.

Vancomyc in resistance

Vancomycin resistance is increasing in past two decades worldwide posing a threat in treatment of serious Enterococcal infections. The treatment options for Vancomycin resistant isolates are very limited and Vancomycin resistance genes are transferable to other Gram positive organisms like Staphylococcus aureus [10]. Prevalence of VRE is varied in different parts of India, 4% in Tamilnadu, 7.09% in Varanasi, 11.39% Nagpur and 14.29% in Indore [16, 12, 20, 21]. Our study showed VRE prevalence as 6.36%, which is concordance with the study by Harsha et al [22]. All theVancomycin resistant Enterococcal isolated strains showed sensitivity to Linezolid and Teicoplanin, hence these two drugs can be used in VRE isolates [23].

CONCLUSION

In coming years Vancomycin resistance can pose challenge to treatment options, hence it is essential to take necessary measures in all health care settings to contain the spread of resistant strains., routine lab detection of VRE, judicious use of Vancomycin and isolation of VRE suspected patients and effective surveillance mechanism will be able to contain the spread of VRE.

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