ANTIBIOTIC SUSCEPTIBILITY PATTERN OF ENTEROCOCCI CAUSING UTI REPORTED IN TERTIARY CARE CENTRE, TRIVANDRUM, KERALA, INDIA

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Abstract
Enterococci are the most common cause of urinary tract infections. The frequency of isolation of Enterococci from urinary tract of patients has risen. Vancomycin Resistant Enterococci along with other multidrug resistant bacteria are the main concerns for physicians. Enterococci may also act as vehicle for spread of resistant genes. The objective of this study was to determine the antimicrobial susceptibility pattern of Enterococci spp. isolated from urine samples. This cross-sectional study was conducted at Department of Microbiology Dr. Somervell Memorial CSI Medical College and Hospital, Karakonam, Trivandrum, Kerala, India. Thirty six Enterococci isolated from UTI cases were processed from August 2019 to October 2019. In-vitro drug-susceptibility tests of Enterococci isolates were performed on Mueller Hinton agar. A total of 36 urine specimens yielding growth of Enterococci were studied. Among these only 19.99% samples were from OPD, from the IP patients (80.56), out of that 24.14% from ICU and rest were from wards (75.86%). Females (83.33%) were found to be more prone to Enterococcal infection as compared to males (16.67%). High prevalence of Enterococcal infection was seen in the age group 61-70 years (25%) followed by 51-60 and 71-80 (16.67%) age groups. Enterococcus strains showed 100% sensitivity against vancomycin and linezolid. 50% of the isolates showed resistant towards high level aminoglycocides. Apart from the above sensitivity pattern the urinary isolates were observed with Nitrofurantoin (88.88%), Ampicillin (55.55%), Penicillin (52.77 %) and Norfloxacin (16.66%). In this study shows highly sensitive towards Nitrofurantoin. These data may aid health professionals in choosing the appropriate treatment for patients with Enterococcal UTI in the region and hopefully will prevent the misuse of antibiotics.

Key words: Antibiotic Susceptibility, Enterococci, UTI, Tertiary care centre, Kerala

Introduction
Urinary Tract Infection (UTI) is one of the most important causes of morbidity in the general population and is the second most common cause of morbidity among hospitalized patients. It has been estimated that symptomatic UTIs result in as many as 7 million visits to outpatient clinics, 1 million visits to emergency departments and 100,000 hospitalizations annually (Nisha and Ashalatha (2017); Wilson and Gaido, 2004).

Nowadays, it represents one of the most common diseases encountered in medical practice affecting people of all ages from the neonate to the geriatric age group (Kunin, 1994). Most infections are caused by retrograde ascent of bacteria from the faecal flora via the urethra to the bladder and kidney especially in the females who have a shorter and wider urethra and is more readily transfer by microorganisms (Jones et al., 2006). The structure of the females urethra and vagina makes it susceptible to trauma during sexual intercourse as well as bacteria been massaged up the urethra and into the bladder during pregnancy and or child birth (El-Sweih et al., 2008; Kolawale et al., 2009).

The risk of developing urinary tract infection increases significantly with the use of indwelling devices such as catheters and urethral stents or sphincters (Foxman, 2003). Urinary tract infections account for an estimated 25 to 40% of nosocomial infections and represent the most common type of these infections (Bagshaw and Laupland, 2006).
Enterococci are Gram-positive cocci, which are normal commensals of the gastrointestinal tract, genital tract and anterior urethra. It is the second most common cause of urinary tract infection (UTI) and third most common cause of bacteremia (Cetinkaya et al., 2000; Gordon et al., 1992). 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 (Foxman et al., 2000). UTIs are the most common of the Enterococcal infections: Enterococcus species have been implicated in approximately 10% of all UTIs and in up to approximately 16% of nosocomial UTIs (Centers for Disease Control and Prevention (CDC), 1993).

Most frequent infections caused by Enterococcus spp. are urinary tract infections followed by intra-abdominal abscesses and bloodstream infections (Low et al., 2001). In addition, studies have shown increasing resistance of Enterococci to anti-microbial agents such as β-lactams and high-level resistance to aminoglycosides and more recently to glycopeptides. This is possibly due to the use of broad-spectrum antibiotics or multi-antibiotic regimes, which allow for enterococcal overgrowth and super infection (Kaçmaz and Aksoy, 2005). β-lactams and aminoglycosides are generally the antibiotics of choice for treating the serious infections caused by Enterococci. High-level gentamicin-resistant (HLGR) enterococci frequently express additional resistance to multiple antibiotics, thereby causing therapeutic problems (Schouten et al., 2000). A high mortality rate of Enterococcal infections is due to increasing resistance of the organism to β lactam antibiotics, aminoglycosides and glycopeptides and inadequate response to the treatment (Shah et al., 2012).

The intrinsic resistance of Enterococci involves cotrimoxazole, aminoglycosides and cephalosporins which are commonly used to treat UTIs and other enterococcal infections. Acquired antimicrobial resistance is also important. It allows virulent Enterococci to survive for extended periods of time in hospital settings (Arias et al., 2011). Moreover, acquired vancomycin resistance is transferable to other organisms such as Staphylococcus aureus and Listeria monocytogenes (Shinde et al., 2012).

In almost all cases there is a need to start treatment before the final microbiological results are available. Area-specific monitoring studies aimed to gain knowledge about the type of pathogens responsible for UTIs and their resistance patterns may help the clinician to choose the right empirical treatment (Katarzyna et al., 2001).

Materials and Methods

A hospital based cross-sectional study was conducted at Dr. Somervell Memorial C.S.I Medical College and Hospital, Karakonam, Trivandrum, Kerala, India. Urine samples were collected from the patients with the symptoms of UTI attended in both IP and OP of health care centre for the period between: August 2019 to October 2019. Midstream specimens of urine (MSU) were collected in labeled and sterile wide mouth plastic container (Tille et al., 2014). After collection the specimens were transported to be analyzed in the laboratory of Microbiology Department. The samples were plated on Blood Agar (Microxpress, A Division of Tulip Diagnostics (P) Ltd. India) and Mac Conkey Agar (Hi Media Laboratories Private Limited, Mumbai) media by the semi-quantitative plating method using the calibrated loop technique (0.001 mL). Plates were incubated aerobically overnight at 37°C. Pure growth of an isolate in a count of ≥10 5 colony forming units (CFU) per milliliter of urine was considered as significant bacteriuria. Growth of ≥3 isolates in a sample was considered as contamination and a repeat sample was advised. Conventional methods of identification were used for identification of the bacterial isolates. (Baron et al., 2002).

Antimicrobial sensitivity test was done on Mueller Hinton agar by the Kirby-Bauer technique according to the CLSI guidelines (CLSI-2007) using Escherichia coli (ATCC 25922), Staphylococcus aureus (ATCC 25923) and Pseudomonas aeruginosa (ATCC 27853) as control strains. The antimicrobial agents used in antibiotic susceptibility testing were Penicillin (10µg), Erythromycin (15µg), Ampicillin (25µg), Linezolid (30µg), Vancomycin (30 µg), Chloramphenicol (30µg) and High level Gentamicin (30µg) (Hi Media Laboratories Private Limited, Mumbai).

Result

A total of Thirty six Enterococci strains were isolated from clinically suspected UTI patients the study period. Among these only 19.44% isolates were from OPD, from the IP patients (80.56), out of that 24.14% from ICU’s and rest were from wards (75.86%) table 1. Females (83.33%) were found to be more prone to Enterococcal infection as compared to males (16.67 %).

Table 1: OP and IP Distribution of Enterococcal UTI cases.

<table>
<thead>
<tr>
<th>Distribution</th>
<th>No. of Isolates</th>
</tr>
</thead>
<tbody>
<tr>
<td>OP (19.44%)</td>
<td>7</td>
</tr>
<tr>
<td>IP (80.56%)</td>
<td>ICU (24.14%) 7</td>
</tr>
<tr>
<td></td>
<td>Wards (75.86%) 22</td>
</tr>
</tbody>
</table>


High prevalence of Enterococcal infection was seen in the age group 61-70 years (25%) followed by 51-60 and 71-80 (16.67%) age groups table 3.

In the present study all Enterococci strains showed 100% sensitivity against vancomycin and linezolid. 50% of the isolates showed resistant towards high level gentamicin. Apart from the above sensitivity pattern the urinary isolates were observed with Nitrofurantoin (88.88%), Ampicillin (55.55%), Penicillin (52.77 %) and Norfloxacin (16.66%) Table 1.

### Discussion

The UTI causing Enterococci are gaining resistance at an amplified rate to usually used antimicrobial agents. The sensitivity pattern is varying in various geographical areas (Saleh et al., 2009). Regular survey of antimicrobial resistance plays a very important role in the empiric treatment of Enterococcal UTI (Pieore et al., 2012). The present study shows the antimicrobial susceptibility pattern of Enterococci isolated from UTI patients. A total of Thirty six Enterococci strains were isolated from clinically suspected UTI patients the study period. The results of Females (83.33%) were found to be more prone to Enterococcal infection as compared to males (16.67 %). This result is closely significant with the studies of Bharti et al., (2016) and Shrivastav et al., (2013). In this study, High prevalence of Enterococcal infection was seen in the age group 61-70 . The finding is coined with the results of Toshiki et al., (2015) a retrospective study conducted in Japan.

The present study, antibiotic susceptibility pattern of Enterococcal isolates towards vancomycin and linezolid concorded with the observations of Bharti et al., (2016) and Abdulla and Abdulla (2006).

The 50% of the isolates showed resistant towards high level gentamicin. Earlier studies in Iran have reported the prevalence of HLGR strains in clinical samples to be about 52% (Feizabadi et al., 2006). The treatment of choice for serious Enterococcal infections is an aminoglycoside in combination with a cell wall active agent (Landman and Quale, 1997). However, high-level aminoglycoside resistance (HLAR) is responsible for loss of synergy between agents active on the cell wall and aminoglycosides (Vakulenko et al., 2003).

The Enterococci isolates sensitivity pattern was observed with Nitrofurantoin (88.88%) and this finding is parallel to the studies of Ali et al., (2014).

### Conclusion

Enterococci have emerged from being harm less commensals to versatile lethal pathogens. Nitrofurantoin is effective in the treatment of Enterococcal UTIs. In this study shows highly sensitive towards Nitrofurantoin. These data may aid health professionals in choosing the appropriate treatment for patients with Enterococcal UTI in the region and hopefully will prevent the misuse of antibiotics.

### Acknowledgement

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**Table 2**: Sex Vs Isolation (%).

<table>
<thead>
<tr>
<th>Sex</th>
<th>Frequency</th>
<th>Percentage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>6</td>
<td>16.67</td>
</tr>
<tr>
<td>Female</td>
<td>30</td>
<td>83.33</td>
</tr>
<tr>
<td>Total</td>
<td>36</td>
<td>100</td>
</tr>
</tbody>
</table>

**Table 3**: Age Distribution of Enterococci spp. UTI cases.

<table>
<thead>
<tr>
<th>Age</th>
<th>Frequency</th>
<th>Percentage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;10</td>
<td>2</td>
<td>5.56</td>
</tr>
<tr>
<td>11-20</td>
<td>3</td>
<td>8.33</td>
</tr>
<tr>
<td>21-30</td>
<td>2</td>
<td>5.56</td>
</tr>
<tr>
<td>30-40</td>
<td>3</td>
<td>8.33</td>
</tr>
<tr>
<td>41-50</td>
<td>3</td>
<td>8.33</td>
</tr>
<tr>
<td>51-60</td>
<td>6</td>
<td>16.67</td>
</tr>
<tr>
<td>61-70</td>
<td>9</td>
<td>25</td>
</tr>
<tr>
<td>71-80</td>
<td>6</td>
<td>16.67</td>
</tr>
<tr>
<td>&gt;80</td>
<td>2</td>
<td>5.56</td>
</tr>
<tr>
<td>Total</td>
<td>36</td>
<td>100</td>
</tr>
</tbody>
</table>

P- Penicillin, AMP- Ampicillin, NIT- Nitrofurantoin, NOR- Norfloxacin, HLG- High level Gentamicin, LZ-Linezolid, VA- Vancomycin.
(Project Co ordinator, Dr. S.M.C.S.I.M.C & Hospital) and all the members of Research Department, Saveetha University, Dr. S.M.C.S.I.M.C & Hospital for the successful completion of this research work.

Ethical Approval

Ethical approval was obtained from the Institutional Human Ethics Committee, Dr. Somervell Memorial C.S.I Medical College and Hospital, Karakonam, Trivandrum, Kerala, India, on 25th of March, 2019.

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