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A HOSPITAL BASED SURVEILLANCE STUDY OF URINARY TRACT INFECTIONS CAUSED BY PSEUDOMONAS AERUGINOSA AND ACINETOBACTER SPECIES WITH SPECIAL EMPHASIS ON DRUG RESISTANCE

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ABSTRACT

BACKGROUND: Urinary tract infection (UTI) is the commonest bacterial infection in community practice. The most common microorganisms causing UTI include E.coli, Klebsiella, Staphylococcus aureus, Coagulase negative staphylococci, Pseudomonas, Proteus and Acinetobacter. The increase in multidrug resistance in bacterial uropathogens is an important and emerging public health problem in non-fermenting isolates. So this study focuses the surveillance of Pseudomonas *aeruginosa,* and Acinetobacter *species in UTI* and also focuses the drug resistance of the isolates.

METHOD: The study was conducted at the Department of Microbiology, DM WIMS,Meppadi,Wayanad, starting from May 2019 to July 2019. A total of 200 urine samples were taken for identifying the significant urinary tract infections. Organisms were isolated and identified using standard microbial techniques. Antibiotic sensitivity was studied using Kirby Bauer disc diffusion method and EDTA double disc synergy test.

RESULT: Out of the 200 urine samples studied, 87 showed significant bacteriuria, with 26 (29.9%) *Pseudomonas aeruginosa* and 6(6.9%) *Acinetobacter species*. Other isolates were E.coli (24), klebsiella (22), enterobacter (4), Citrobacter (3) and one each were Serratia and Morganella. Among *these isolates 15 Pseudomonas aeruginosa* and 2 *Acinetobacter species were MBL producers*.

CONCLUSION: The study reports that other than E.coli, *Pseudomonas aeruginosa* has a higher prevalence in urinary tract infection and more than half of the isolates are showing drug resistance to the commonly used drugs. Most of the infection with such strains were treated successfully with combination of drugs such as Tigecycline with colistin, colistin with a carbapenem, fosfomycin with a carbapenem, fosfomycin with aminoglycoside, and a carbapenem with an aminoglycoside have been reported as antibiotic combinations effectively administered to series of patients infected with carbapenemase producing organisms.

Urinary tract infections (UTIs) present microbial colonization of urine. They pose a major public health problem, due to the growing phenomenon of bacterial resistance to a wide range of antibiotics [1]. *Pseudomonas aeruginosa* and *Acinetobacter baumannii* are aerobic Gram-negative bacteria that do not ferment glucose and are ubiquitous in the environment. These are responsible for 12% of development of nosocomial bacteriuria, related to atheterization [3,4]. The mortality and morbidity associated with *P. aeruginosa* induced UTIs remain significantly high. *P. aeruginosa* has an innate tendancy to stick to the surfaces of catheters and form biofilms leading to higher incidence of UTIs in patients with long-term indwelling. Hence the study was aimed to detect the surveillance of *Pseudomonas aeruginosa* and *Acinetobacter* species in urine with emphasis on drug resistance, so that an effective antimicrobial treatment can be formulated and also reduces the risk of antimicrobial resistance.

MATERIALS AND METHODS

The study was conducted at the Department of Microbiology ,DM WIMS ,Meppadi, Wayanad, for a 3 months period starting from May 2019 to July 2019 to assess the surveillance of *Pseudomonas aeruginosa* and *Acinetobacter* species in urine. A total of 200 urine samples from patients of all age groups received in the microbiology laboratory for routine examination and culture, during the study period.

All the samples were processed by standard microbiological operating procedure for the isolation and identification of microorganisms following the manual of Clinical microbiology [4]. The samples were inoculated in routine culture media (blood agar, Mac conkey agar) , the colonies were subjected for microscopic examination and the bacteria were identified using colony charecteristics ,Gram staining and biochemical reactions. Antibiotic susceptibility testing of all isolates was performed by Kirby-bauer disc diffusion method and interpretation of the result was made in compliance with CLSI guidelines[5].

DETECTION OF METALLO-BETALACTAMASE PROD UCTION

In this study phenotypic detection method was followed for the detection of metallobetalactamase isolates.

SCREENING TEST

Phenotypic detection of metallobetalactamase among the uropathogens was carried out using Imipenem (10µg) and Imipenem + EDTA (750µg) discs [6].The metallobeta lactamase producing isolates was showed a greater than 7mm variations between the inhibition zone around the Imipenem discs alone and the inhibition zone around the Imipenem + EDTA discs.

The isolates were tested for the antibiotic susceptibility testing by Kirby bauer disc diffusion method on Muller-Hinton Agar as per CLSI [5].A suspension of bacteria equivalent to 1:10 dilution of 0.5 Mc Farland standard were used to prepare Lawn culture in Muller-Hinton agar and subsequent application of antibiotic discs was carried out[6,7].

EDTA-DOBLE DISC SYNERGY TEST

The imipenem-EDTA double disc synergy test was

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performed as described by Lee *et al* and Arakawa *et al*. 0.5 M EDTA solution was prepared by dissolving 186.1 g of disodium EDTA.$2H_2O$ (RANCHEM, New Delhi, India) in 1, 000 ml of distilled water. The pH was adjusted to 8.0 by using NaOH (HI-MEDIA, Mumbai, India) and was sterilized by autoclaving.

EDTA disks (6 mm in diameter, Whatman filter paper no.1) were prepared by incorporating 10 L of 0.5 M solution of EDTA on blank disks (equivalent to 750 g per disk). The test organisms were adjusted to a 0.5 McFarland turbidity standard and inoculated on Mueller Hinton agar plates as recommended by the CLSI. The EDTA disk was placed 20 mm apart edge to edge from the imipenem disk and the plates were incubated overnight at 35°C. A zone of synergy between the antibiotic disk and EDTA was taken as a positive result [8].

RESULT

Among the 200 isolates 87 showed significant growth with 35 MBL positive isolates(35/87). Among this 15(42.85%) were MBL *Pseudomonas aeruginosa,* being the most prevalent one followed by 2(5.71%) *Acinetobacter* species and 52(52/87) were non MBL isolates. Among this 11(21.2%) *Pseudomonas aeruginosa and* 4(7.7%) were *Acinetobacter* species. The highest prevalence of the infection was noted in female than the male. Patients with the age group of above 60 years were more prone to bacterial infection. In the present study all of the MBL producing *Pseudomonas aeruginosa* and *Acinetobacter species* were sensitive to PolymixinB and colistin and resistant to carbapenem (Meropenem, Imipenem) and cephalosporins (cefoxitin, cefaperazone, cefuroxime and ceftazidime) .

DISCUSSION

In this study, total of 200 urine samples were processed in the lab; out of these 87(36%) samples were identified with significant bacteriuria and from these 26 (29.9%) samples were identified as *Pseudomonas aeruginosa* and 6(6.89%) *Acinetobacter species*. The present study reported that the highest prevalence of urinary tract infections are caused by MBL(15/87) *Pseudomonas aeruginosa* as compared to *Acinetobacter* species. Mohibur *et al*.,2017 reported in their study that prevalence of MBL production was high in *Pseudomonas aeruginosa* (28%) which is consistent with present study.

A study by Rahn *et al*.,2010 reported that the highest rate of MBL were in male(66.8%) and also reported that the age group 61-80 years were more affected with MBL infections. In the present study , most of the infected patient were females (65.7%) than the male(34.3%) which is not consistent with Rahn's study and also my study detected the age prevalence of MBL infection is higher in the age group above 60 years which is consistent with Rahn's study.

In the present study , all the MBL producing *Pseudomonas aeruginosa* and *Acinetobacter species* were sensitive to Polymixin B and colistin, but were resistant to Carbapenems; Meropenem and Imipenem, Cephalosporins ,amoxycla vulinate ,Piperacilin-Tazobactam and Norfloxacin. The sensitivity to other classes of are as 77.2% each of ciprofloxacin and nitrofurantoin, 71.4% of each cotrimoxazole ,amikacin and gentamycin and 65.7% tobramycin.

In this study Double disc synergy test and combined disc method was found to be effective in the detection of MBL isolates.In a study conducted by Dardi Charan Kaur *et al.,*2015,MBL Gram negative isolates in urine showed 93-100% Sensitive to Polymixin and colistin, 71.42% sensitive to Amikacin and Gentamycin which is consistent with the present study. Present study showed 100% resistance to Carbapenems(ImipenemandMeropenem)Penicillin,Ampicillin,Cephalosporins(Cefuroxime, Ceftazidime and Cefaperazone) and Norfloxacin. The antimicrobial sensitivity and resistance pattern varies from community to community and from hospital to hospital. This is because of the emergence of resistant strains as a result of indiscriminate use of antibiotics.

CONCLUSION

The present study concluded that the *Pseudomonas aeruginosa* (26/87) is found to be the most predominant isolates followed by *Acinetobacter baumannii* (6/87) in urinary tract infection. Among the 26 *pseudomonas aerug inosa* 15 (57.7%) were MBL positive isolates and from 6 *Acinetobacter species* 2(50%) were MBL positive isolates. The study concluded that the *Pseudomonas aeruginosa* is the prevalent organism in urinary tract infection. The present study reveals that the urinary tract infection caused by the *Pseudomonas aeruginosa* and *Acinetobacter speciesare* mostly seen in elderly population. This may be due to their immune status and sedentary lifestyle. Tigercycline with colistin, colistin with a carbapenem, fosfomycin with a carbapenem, fosfomycin with an aminoglycoside, and a carbapenem with an aminoglycoside have been reported as antibiotic combinations effectively administered to series of patients infected with carbapenemase producing gram negative organisms.

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