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Antimicrobial Susceptibility Patterns of *Pseudomonas aeruginosa* Clinical Isolates at a Tertiary Care Hospital in Ambajogai

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Abstract

Introduction: Currently antibiotic resistance in bacterial populations is one of the greatest challenges to the effective management of infections. Infections caused by *Pseudomonas aeruginosa* are frequently life threatening & difficult to treat as it exhibits intrinsically high resistance to many antimicrobials & the development of increased, particularly multi-drug resistance in health care settings. *Objective:* The present study was done to determine the drug sensitivity pattern of *Pseudomonas aeruginosa* from various clinical specimens in our set up. *Materials and Method:* This study was conducted during October 2015 to May 2016. A total of 376 strains of Pseudomonas aeruginosa were isolated from various clinical specimens. *Pseudomonas aeruginosa* was identified by using standard microbiological techniques. Antimicrobial susceptibility patterns of all the isolates were carried out by Kirby- Bauer disk diffusion method as per CLSI guidelines. *Result:* A total 376 strains of *Pseudomonas aeruginosa* were isolated of which 267 (71%) were from indoor & 109 (29%) were from outdoor patients. Of the 376 isolates 206 were from males & 170 were from females. Majority of isolates of *Pseudomonas aeruginosa* were obtained from specimens of pus. Majority of the strains showed low level of susceptibility to Ceftazidime (38%), Ofloxacin (40.4%), Ciprofloxacin (42.1%), Piperacillin (47.7%) & Gentamicin (51%). We got good sensitivity with Tobramycin (82.1%) & with Amikacin (78.3%). All isolates were susceptible to Imipenem (100%). *Conclusion:* The result of our study suggests the occurrence of resistant strains of *Pseudomonas aeruginosa*. Periodic susceptibility testing should be carried out over a period of two to three years to detect the resistance trends & judicious, rational treatment regimen prescription should be followed by physician.

Keywords: Antibiotic Susceptibility Pattern; *Pseudomonas aeruginosa.*

Introduction

Pseudomonas aeruginosa is increasingly recognized as an emerging opportunistic pathogen of clinical relevance that causes infections in hospitalized patient particularly in burn patients, orthopaedicrelated infection, respiratory diseases, immunosuppressed and catheterized patients [1,2]. Nowadays more & more resistance of *Pseudomonas aeruginosa* are encountered in routine clinical practice, a serious problem, increase morbidity and mortality and also cost of treatment [3]. Members of thePseudomonad's genus are major agents of

nosocomial and community acquired infections, being widely distributed in the hospital environment where they are particularly difficult to eradicate [4]. It has also been observed that 28% of healthy people in hospital environment are carrier for*Pseudomonas aeruginosa* [5]. Mechanisms that cause antimicrobial drug resistance and multi-drug resistance in *Pseudomonas aeruginosa* are due to acquisition of resistance genes (e.g.those encoding beta-lactamase and amino-glycoside modifying enzymes) via horizontal gene transfer and mutation of chromosomal genes (target site, efflux mutations) are the target of the fluoroquinolones particularly ciprofloxacin [6]. *Pseudomonas aeruginosa* can survive harsh environmental conditions and displays intrinsic resistance to a wide variety of antimicrobial agents, two factors that facilitate the organism's ability to survive in hospital setting [7]. Biofilm formation in *Pseudomonas aeruginosa*, particularly in the case of pulmonary infections in patients with cystic fibrosis, contributes to its resistance to antimicrobial agents [8]. *Pseudomonas aeruginosa* demonstrates resistance to multiple antibiotics, thereby jeopardizing the selection of appropriate treatment [9]. Over period of time, we observed an increase in number of *Pseudomonas aeruginosa* among our laboratory isolates, and thereforethe present study was designed to find out the current antimicrobial susceptibility patterns of *Pseudomonas aeruginosa* in our set up.

Materials and Method

Place

The study was carried out in the Department of Microbiology, Swami RamanandTeerth Rural Government Medical College and Hospital, Ambajogai, Beed.

Time Period

The study was of 8 months, conducted from October 2015 to May 2016.

Samples

Sampleswere collected from patients who are hospitalized for more than week with all aseptic precautions. The various clinical samples were included such as pus, sputum, urine, blood etc.

Isolation and Identification

The samples were further processed on the basis of their growth on routine MacConkey medium which showed lactose non-fermenting pale colonies which were oxidase test positive and on Nutrient agar pigmented and non-pigmented colonies with oxidase test positive. A battery of tests were performed that included Gram's staining, motility tests, sugar fermentation tests and biochemical tests such as urease and Phenyl pyruvic acid test and IMViC (indole, methyl red, Voges-Proskauer and citrate) tests for the identification of *Pseudomonas aeruginosa*.

Antibiotic Sensitivity Testing

Pseudomonas aeruginosa strains were subjected to antibiotic susceptibility testing on Mueller- Hinton agar by Kirby–Bauer disc diffusion method following National Committee for Clinical Laboratory standards (NCCLS) guidelines. The following antibiotics were tested: Imipenem(10 mcg/disc), Gentamycin(10 mcg/disc), Amikacin(30 mcg/disc), Ceftazidime(30 mcg/disc), Ciprofloxacin(5 mcg/disc), Ofloxacin(5 mcg/disc),Tobramycin(10 mcg/disc) and Piperacillin(100 mcg/disc). *Pseudomonas aeruginosa ATCC 27853* was used as control strain.

Result

A total of 376 strains of *Pseudomonas aeruginosa* were isolated and identified by standard microbiological procedures out of total 3387 samples. Out of total 376 strains 267 were from indoor and 109 from the outdoor patients, while 206 (55%) were from males and 170 (45%) from females.

Maximum isolates of *Pseudomonas aeruginosa*, 138 (36.7%) were from pus sample only, followed by sputum 98(26%) and urine 55 (13.8%) [Table 1].

Antimicrobial susceptibility patterns of *Pseudomonas aeruginosa* varied markedly. *Pseudomonas aeruginosa* isolates showed minimum susceptibility to Ceftazidime (38%) and maximum susceptibility to Tobramycin (82.1%). All isolates were sensitive to the carbapenem drug- Imipenem (100%).

Table 1: Sex wise distribution of the *Pseudomonas aeruginosa* isolates

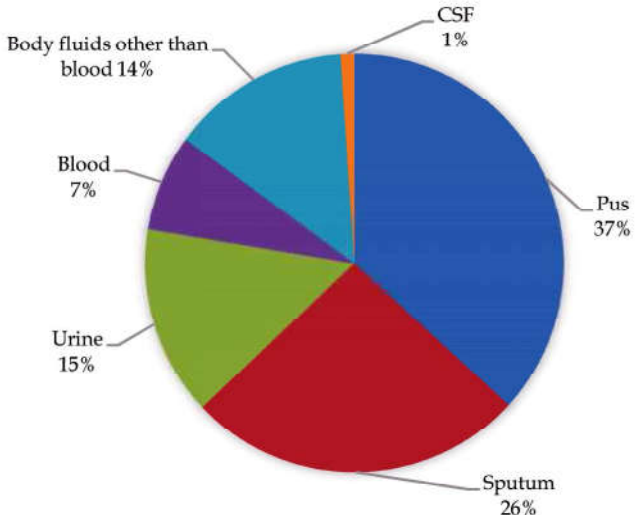
Sex	Total number	Percentage (%)
Male	206	55
Female	170	45
Total	376	100

Table 2: Isolation of *Pseudomonas aeruginosa* from various clinical specimens

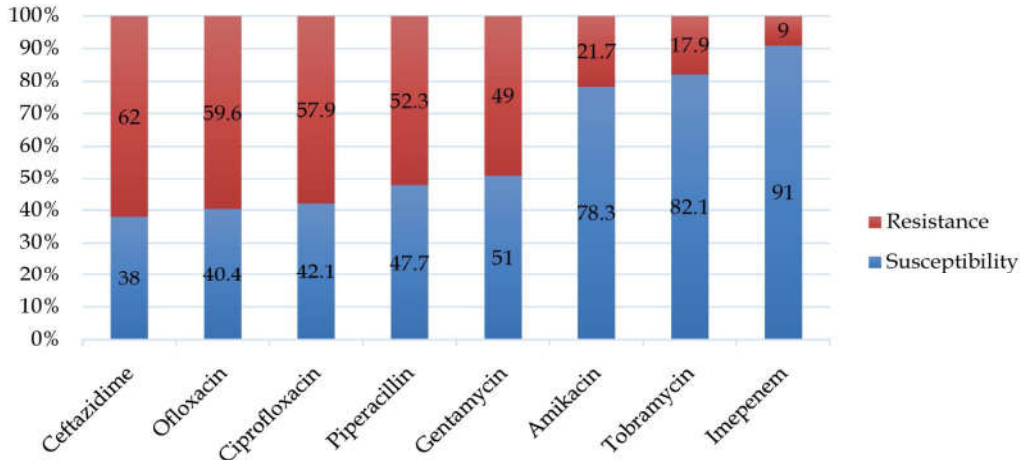
Specimen	Number of Isolates	Percentage (%)
Pus	138	36.70
Sputum	98	26.06
Urine	55	14.63
Blood	28	7.45
Body fluids other than blood	52	13.82
CSF	4	1.06
Total	376	100

Table 3: Antibiotic susceptibility of *Pseudomonas aeruginosa* isolated from various clinical samples

Antibiotic	Sensitivity (%)
Ceftazidime	38
Ofloxacin	40.4
Ciprofloxacin	42.1
Piperacillin	47.7
Gentamycin	51
Amikacin	78.3
Tobramycin	82.1
Imipenem	100



Graph 1: Isolation of pseudomonas aeruginosa from various clinical specimens



Graph 2: Bar diagram showing the percentage of susceptibility and resistance of *Pseudomonas aeruginosa* to various antimicrobials

Discussion

Out of 3387 clinical samples of various natures, *Pseudomonas aeruginosa* was isolated from 376 samples i.e. the isolation rate was 11.1%, so it is undoubtedly an important nosocomial pathogen.Being an extremely adaptable organism, it can survive and multiply even with minimum nutrients, if moisture is available. The isolation rate of *Pseudomonas aeruginosa*in our study is comparable with study done by More S.R. et alwho mentioned isolation rate as 9.19% [10].

We can say that duration of stay is directly proportional to the rate of infection as out of 376 strains of *Pseudomonas aeruginosa* in our study, 267(71%) were from indoor and 109 (29%) were isolated from outdoor patients which is in correlation with Shampa Anupurba study who also mentioned the indoor patients to outdoor patients ratio that is 1:0.36 [11].

In the present study infections caused by *Pseudomonas aeruginosa* were more common in males (55%) compared to females (45%). This finding is in comparison with study done by Rajat Rakesh et al [4], Jamshid et al [12] and Rashid et al [13].

In our study, the maximum isolates of *Pseudomonas aeruginosa* were from pus/swab (36.7%), followed by sputum (26.06%). These results are in line with studies of Vijaya Choudhari et al [14] who mentioned that pus samples (35.3%) showed highest culture positivity followed by sputum (20.8%) and urine (13%).

In our study, majority of the isolates were susceptible to Tobramycin by 82% followed by Amikacin *Pseudomonas aeruginosa* isolates were found to be sensitive to Imipenem. This may be due to the 78.3% which is comparable with S. Shenoy [15] study. One striking feature in this study was that all the restricted use of Imipenem in this hospital. This is consistent with a report published in 2002 in Mangalore, India [15] but other studies have showed varying degrees of resistance to Imipenem in recent years [16,17,18,19].

In our study, the isolates were less susceptible to fluoroquinolones such as Ciprofloxacin (42.1%) similar findings had been reported in a study done in North Kerala [20].

This study shows that the clinical isolates of *Pseudomonas aeruginosa* are Multidrug resistance (resistance to ≥3 different classes of antibiotics tested) and are becoming resistant to commonly used antibiotics and gaining more and more resistance to newer antibiotics. The antimicrobial agents are losing their efficacy because of the spread of resistant organisms due to indiscriminate use of antibiotics, lack of awareness, patient noncompliance and unhygienic condition.

Conclusion

Results of the present study clearly demonstrated the occurrence of resistance to various antipseudomonal agents among the *Pseudomonas aeruginosa* isolates. Amikacin seems to be a promising therapy for Pseudomonal infection. Hence, its use should be restricted to severe nosocomial infections, in order to avoid rapid emergence of resistant strains. The problem of increasing resistance to *Pseudomonas aeruginosahas* limited the use of other classes of antibiotics like the fluoroquinolones, tetracycline's, macrolides and chloramphenicol [21].

Imipenem was the only anti-pseudomonal drug against which all isolates of *Pseudomonas aeruginosa*were fully sensitive. To prevent the spread of the resistant bacteria, it is critically important to have strict antibiotic policies while surveillance programsfor multidrug resistant organisms and infection control procedures need to be implemented. We suggest that there is a need to emphasize the rational use of antimicrobials and strictly adhere to the concept of "reserve drugs" to minimize the misuse of available antimicrobials. The solution can be planned by continuous efforts of microbiologist, clinician, pharmacist and community to promote greater understanding of this problem.

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