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A study on antimicrobial susceptibility of *Pseudomonas aeruginosa* isolated from skin and soft tissue infections with special reference to carbapenem resistance

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Abstract

Background: *Pseudomonas aeruginosa* is an aerobic gram negative rod which is widely implicated in skin and soft tissue infections. It is also one of the most important cause of hospital acquired infections. Development of Antimicrobial resistance in *Pseudomonas* is a very big concern as it poses difficulty in treating infections

Aim: The aim of this study is to analyse antimicrobial susceptibility of *Pseudomonas aeruginosa* isolated specifically from skin and soft tissue infections for a period of 3 months in a tertiary care hospital. The study also focuses on Carbapenems' (Imipenem, Meropenem) resistance of *Pseudomonas aeruginosa*.

Materials & Methods: Antimicrobial susceptibility testing of *Pseudomonas aeruginosa* isolated from (207) wound swab and pus samples from both inpatient and outpatient samples are done by Kirby Bauer disc diffusion method and interpreted as per CLSI (Clinical Laboratory Standard Institute) guidelines.

Results: In this study, Majority of the *Pseudomonas* isolates are susceptible to Piperacillin-tazobactam (91%) Imipenem (89%), Meropenem (91%). A comparatively lesser percentage of susceptibility is seen towards cephalosporins and fluoroquinolones. Maximum Percentage of *Pseudomonas* isolates were obtained from Burn Intensive Care unit (48.3%). Percentage of *Pseudomonas* isolates resistance to Imipenem, Meropenem are 9%, 12% respectively.

Conclusion: *Pseudomonas* is gaining resistance to various group of antibiotics especially cephalosporins and fluoroquinolones. It has also started slowly gaining resistance to Carbapenem group of antibiotics.

Keywords: *Pseudomonas aeruginosa*, skin and soft tissue infections, carbapenems

Introduction

Pseudomonas aeruginosa is a gram negative bacillus that causes wide variety of infections that cause serious illness to humans. It exists in water bodies, sand, as microbiota in humans. It majorly causes hospital acquired infections and acts a opportunistic microbe ^[1].

Pseudomonas aeruginosa is widely implicated in skin and soft tissues and specifically burns wound infections. This is because there is a loss of protective skin barrier; studies reveal that wound infections caused by *pseudomonas aeruginosa* will increase the risk of rejection of allografts and skin grafts. *Pseudomonas* infections are also most common among diabetic patients with burns wound infections ^[2]. *Pseudomonas* are the most troublesome agents causing nosocomial infections. Hospital infections caused by *Pseudomonas* are infections of wounds, bedsores, eye infections, urinary infections due to catheterisation, pneumonia following ventilator use are the common ones. *Pseudomonas* causes iatrogenic meningitis following lumbar puncture, post tracheostomy pulmonary infection. Ecthyma gangrenosum, green nail syndrome and many other types of skin lesions are also caused by *Pseudomonas*. *Pseudomonas aeruginosa* are extremely adaptable organism, they can survive and multiply even with minimal nutrients, if moisture is available ^[3].

Virulence of *Pseudomonas aeruginosa* are contributed by wide variety of factors. Bacterial pilli (favours adhesion), neuraminidase, Alginate glycoalyx slime help in colonisation. Enzymes like LasA and LasB elastases, cytotoxins, exotoxin A, exoenzyme S, lipopolysaccharide (cell wall) and hemolysins help in invasion into human tissues and establish infection [4]. Ability to form biofilms is also a virulence factor.

Pseudomonas is notorious for its wide range of mechanisms for antibiotic drug resistance. These modes of antibiotic drug resistance can be intrinsic or acquired. The inherent known mechanisms of antibiotic resistance are by decreasing the permeability of cell membrane to antibiotics, presence of effluent pumps or antimicrobial modifying enzymes. The acquired mechanisms of antibiotic resistance are mutations in chromosomes genes causing structural changes and transfer antibiotic resistance genes through plasmids [5].

In 2019, 1.27 million deaths were attributed to bacterial Antimicrobial resistance (AMR). Out of those 1.27 million deaths *Pseudomonas* accounted for 84,600 deaths for resistance to more than 1 drug. There is an increasing resistance to drugs which poses a fatal risk to public health [6].

This study focuses on antimicrobial susceptibility testing of *Pseudomonas* isolates from skin and soft tissue samples and determination of prevalence of isolates that are specifically resistant to carbapenems.

Materials and Methods

A total number of 207 *Pseudomonas aeruginosa* strains were isolated from different specimens of skin and soft tissue

infections like pus, wound swabs. The study was approved by Institutional Ethical Committee (IEC). This cross sectional study was conducted in the Department of Microbiology in a tertiary care hospital, Chennai.

- **Study Period:** 3 months
- **Inclusion Criteria:** The clinical specimens of skin & soft tissue infections from outpatient departments (OPD) as well as In Patient departments (IPD) were included in the study.
- **Exclusion Criteria:** Patients on prior antibiotics were excluded from the study.

Methodology

Pseudomonas aeruginosa strains isolated from skin & soft tissue infections were identified by standard conventional methods like gram staining, motility, culture, pigment production and biochemical tests especially catalase and oxidase test etc.

Antimicrobial susceptibility profile of *Pseudomonas aeruginosa* strains was done by Kirby-Bauer disc diffusion method on Mueller-Hinton agar with antibiotic discs like: 1) Piperacillin Tazobactam (PTZ) 2) Levofloxacin (LE) 3) Ceftazidime (CAZ) 4) Cefepime (CPM) 5) Imipenem (IPM) 6) Meropenem (MRP) and were interpreted as per Clinical Laboratory Standard Institute (CLSI) guidelines. The socio-demographic data - gender, ward, culture results and their antibiotic susceptibility were analysed with the help of Ms Excel 2013 version.

3. Results

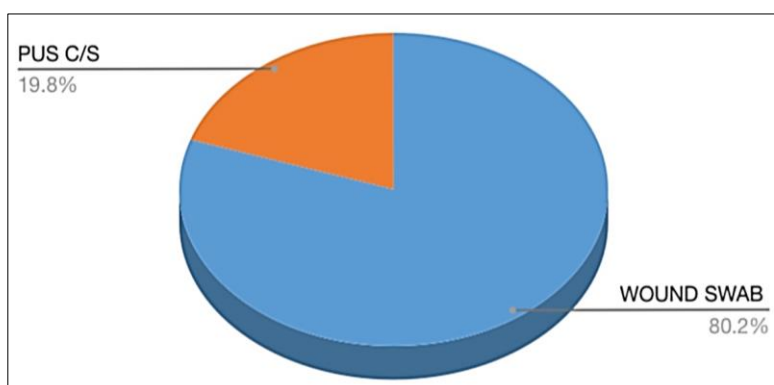


Fig 1: Shows different types of samples.

Fig 1 shows the isolation of *Pseudomonas aeruginosa* strains from different clinical samples. 166 (80.2%) were isolated

from wound swab followed by 41 (19.8%) from pus samples.

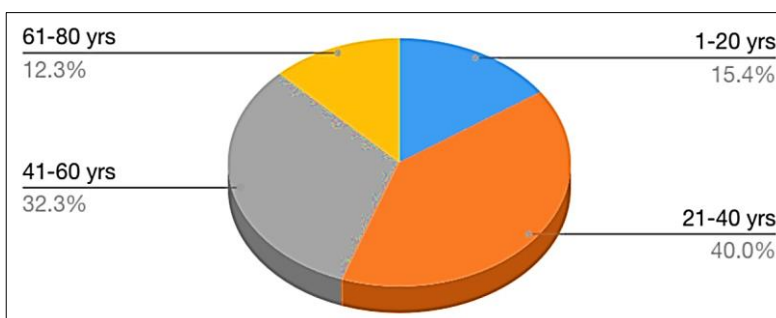


Fig 2: Shows distribution of isolates across age groups.

The Figure 2 & Table 1 tell us that a total of 207 isolates distributed across four age groups. The majority of isolates were found in the 21-40 years age group, accounting for 40% of the isolates. This was followed by the 41-60 years group with 32.30%, 1-20 years group had 15.40% of the

isolates and 61-80 years group with 12.30% of the isolates. Gender wise, males constituted a higher proportion with 183(88.4%) out of 207 isolates, while females accounted for 11.6%. This gender difference was consistent across all age groups.

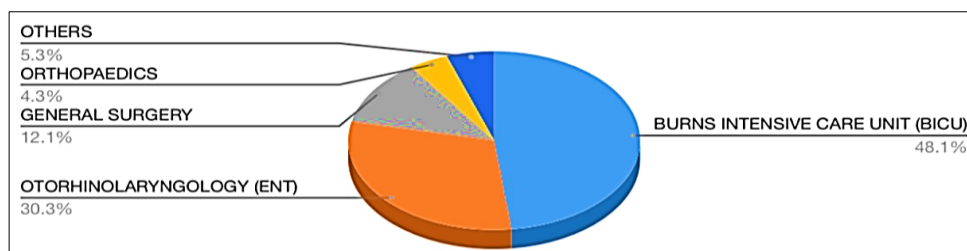


Fig 3: Shows distribution of samples

Figure 3 and Table 2 shows that highest proportion of isolates came from the Burns Intensive Care Unit (BICU), accounting for 48.30% of the total. This was followed by the

Otorhinolaryngology (ENT) with 30.40%, and General Surgery with 12.10% and Orthopaedics (4.3%).

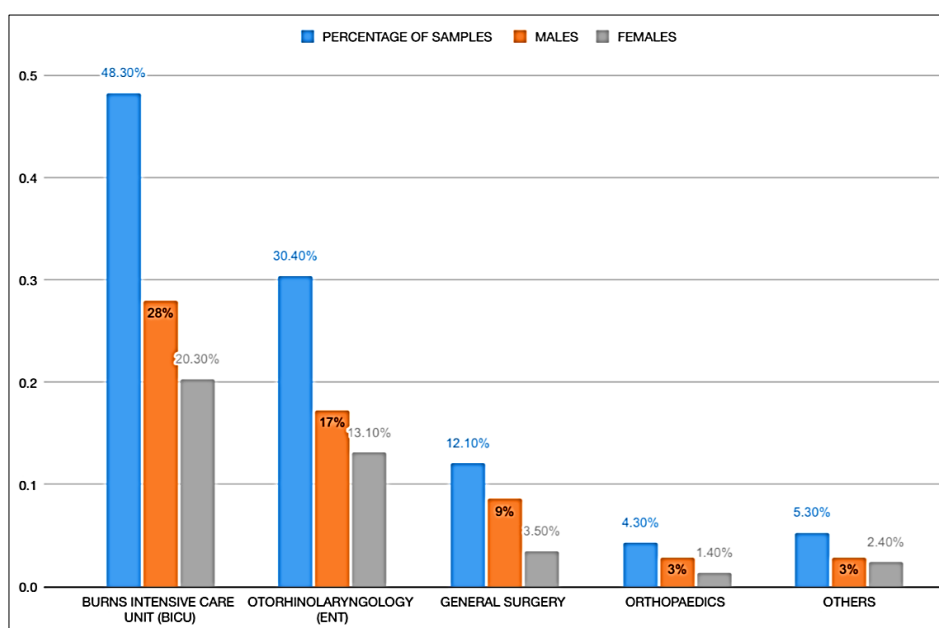


Fig 4: Shows Gender-wise distribution of isolates

Figure 4 and Table 2 show the Gender-wise distribution across various wards majority of the isolates are obtained from male patients in all wards.

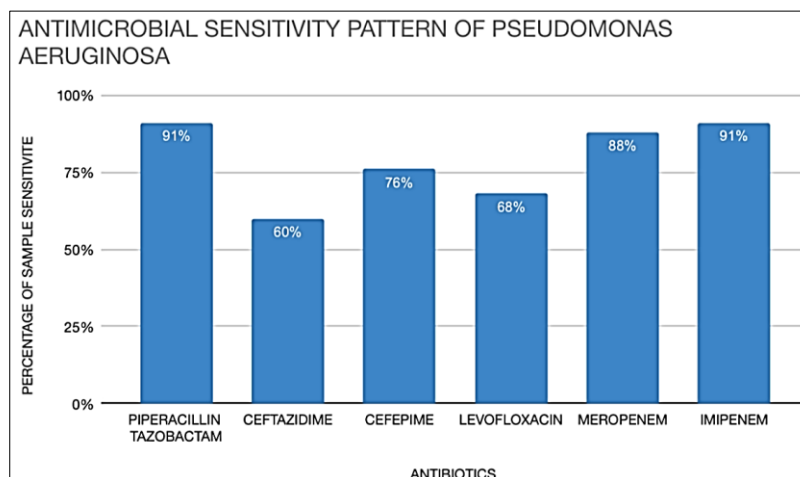


Fig 5: Shows overall antimicrobial susceptibility of *Pseudomonas aeruginosa*

The Figure 5 and Table 3 show the overall antimicrobial sensitivity of *Pseudomonas Aeruginosa*. The sensitivity to

Piperacillin-Tazobactam and Carbapenems are relatively higher than other antibiotics.

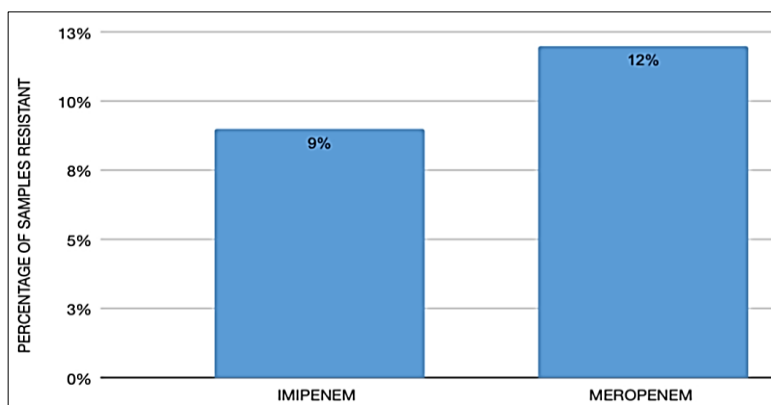


Fig 6: Shows overall percentage of *Pseudomonas* showing carbapenem resistance

Figure 6 shows 19 (9%) samples are resistant to imipenem, 25(12%) samples are resistant to Meropenem. Maximum number of carbapenem resistant isolates are identified in BICU, that is Meropenem & Imipenem are (78.4%) and (79%) respectively.

4. Discussion

In this study, most of the isolates of *pseudomonas aeruginosa* are from the Burns intensive care unit(BICU) similar to the study done by Mukhopadhyay R *et al.*. The main reason behind this is loss of protective skin barrier and moist environment which provides favourable growth conditions for *pseudomonas* [7]. The next most number of isolates were from General surgery, ENT, orthopaedics which is similar to the study done by Ingale HD *et al.*, [8]

In this study, the majority of isolates obtained were from males which is similar to findings reported from the study done by Bangera D *et al.*, and Gill MK *et al.*, [13, 17]. When the isolates of *pseudomonas* are analysed in terms of age groups, the maximum percentage of isolates were present in the age groups 21-40 years(40%) which is similar to the finding obtained in Gill MK *et al.*, (37%) [17].

Overall analysis of Antimicrobial sensitivity pattern of *Pseudomonas* isolated showed that the maximum percentage of isolates (91%) were susceptible to piperacillin-tazobactam which is similar to studies done by Ramakrishna MS *et al.*, (98.1%), V Sheeba *et al.*, (89%), Bangera D *et al.*, (85.26%), Deboral A *et al.*, (81.5%) [9, 11, 13, 16]. The percentage of isolates sensitive to cefepime is (76%) is comparatively lesser to results obtained Ramakrishna MS *et al.*, (98%) [9]. The percentage of isolates that are sensitive to ceftazidime is (60%) which is in accordance with results reported by Nirmala S *et al.*, (58%), UKEY P *et al.*, (53.64%), Gill MK *et al.*, (52.50%), Bangera D *et al.*,

(68%), Deboral A *et al.*, (71.3%) [15, 10, 17, 13, 18]. Antibiotic susceptibility testing also shows that (68%) of isolates were susceptible to levofloxacin which is comparatively more than the result obtained in study done by Rajput K *et al.*, (33.33%) [16].

Analysis of specifically carbapenem sensitivity rates show that, total the number isolates which are sensitive to Meropenem is (88%) and is similar to the reports from studies done by Ramakrishna MS *et al.*, (90.5%), V Sheeba *et al.*, (87%), Deboral A *et al.*, (83.30%), UKEY P *et al.*, (77.25%), j Bhardwaj H *et al.*, (91%) [9, 10, 11, 12, 16]. Sensitivity to imipenem is (91%) which is found similar to the results obtained from reports of Ramakrishna MS *et al.*, (93.5%), V Sheeba *et al.*, (92%), Nirmala S *et al.*, (100%), Bangera D *et al.*, (92.85%), Deboral A *et al.*, (82.40%) [9, 11, 13, 15, 16].

Carbapenem resistant isolates are given special importance in our study. The percentage of isolates resistant to Imipenem and Meropenem (9%) and (12%) respectively which are similar to the results obtained from studies done by Devi KD *et al.*, Yadav R *et al.*, (18, 19) and also we have lower percentage of Meropenem resistance (12%) as compared to the study done by Bangera D *et al.*, (33.03%) [13]. The difference in percentage of resistant isolates can be attributed to difference in hospital environment, geographical location etc.

Tables

Table 1: Shows distribution of isolates across age groups.

S. No	Age Group	Percentage
1	1-20 yrs	15.40%
2	21-40 yrs	40%
3	41-60 yrs	32.30%
4	61-80 yrs	12.30%

Table 2: Shows distribution of samples along with gender-wise stratification

S. No.	Speciality	Percentage of Samples	Males	Females
1	Burns Intensive Care Unit (BICU)	48.30%	28%	20.30%
2	Otorhinolaryngology (ENT)	30.40%	17.30%	13.10%
3	General Surgery	12%	8.60%	3.50%
4	Orthopaedics	4%	2.90%	1.40%
5	Others	5.30%	2.90%	2.40%

Table 3: Shows overall antimicrobial susceptibility of *Pseudomonas aeruginosa*

Antibiotics	No. Of sensitive samples (N=207)	Percentage Of Samples That Are Sensitive
Piperacillin Tazobactam	188	91%
Ceftazidime	125	60%
Cefepime	158	76%
Levofloxacin	141	68%
Meropenem	182	88%
Imipenem	188	91%

Conclusion: The majority of isolates of *Pseudomonas* from skin and soft tissue infections are from Burns intensive care unit (BICU). The Antimicrobial susceptibility pattern of *Pseudomonas aeruginosa* showed maximum sensitivity to Piperacillin-tazobactam and Carbapenems and lesser sensitivity to cephalosporins and fluoroquinolones.

Prevention of *Pseudomonas aeruginosa* cross-infection in hospitals requires constant vigilance and strict attention to asepsis. Infection control measures like hand hygiene should be effectively practiced in all intensive care units and antimicrobial stewardship should be meticulously followed.

Authors' Contribution

All authors have contributed equally in the formulation of study and preparation of the manuscript.

Competing Interest

Authors declare that there are no competing interests

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