Antibiotic Susceptibility Pattern of *Pseudomonas aeruginosa* Isolates from Wound Infections in a Tertiary Care Centre in South Kerala, India

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To cite this article:

Received: May 4, 2018; Accepted: June 14, 2018; Published: July 9, 2018

Abstract: Multiple antibiotic resistance in bacterial populations is a pervasive and growing clinical problem, which is recognized as a threat to public health. Drug resistance to *Pseudomonas* sp. has spread to such a level irrespective of the type of patients, that, its pattern of distribution and antibiotic resistance needs to be studied in detail, especially in trauma patients. Of the Gram negative bacilli, *Pseudomonas aeruginosa* has been of particular interest, the incidence of which in wound infection has increased compared to a decade back. The objective of this study was to know the antibiotic susceptibility pattern of *Pseudomonas aeruginosa* isolates from wound infections. Out of the total of 1404 cases from which pus samples collected, 204 (14.5%) yielded *Pseudomonas aeruginosa* isolates. Among the antibiotics tested, Imipenem was the most sensitive drug, showing susceptibility in 81.3% of the isolates, followed by Piperacillin + tazobactam (76.4%), Meropenem (70.5%) and Piperacillin (67.6%). Fifteen (7.4%) multidrug resistant strains were reported out of the 204 isolations. Increase in resistance to aminoglycosides and fluoroquinolones in this study, prompted evaluation of antibiotic susceptibility pattern of *Pseudomonas* isolates from clinical samples at regular intervals.

Keywords: *Pseudomonas aeruginosa*, Multiple Drug Resistance, Wound Infections, Gram Negative Bacilli

1. Introduction

Wound infection is universal and the bacterial type varies with geographical location, resident flora of the skin and clothing at the site of wound [1]. There are reports of considerable number of carriers for *P. aeruginosa* among the healthy people within the hospital environment, capable of transmitting infection to the patients undergoing treatment [2]. In the recent years, *Pseudomonas aeruginosa* has overtaken *Staphylococcus aureus* in causing wound infections [3]. In the past three years, some of the Indian studies shown *Pseudomonas* sp as the most predominant isolate from wound infections (Gunjam Shrivastava et al, 2016 [4], Vivek Kulkami et al, 2015 [5] and Richa Gupta et al, 2016 [6]. In one of the studies from Maharashtra, Sapana G Mundhada et al, 2015 [7] reported *Pseudomonas aeruginosa* as second dominant strain isolated, wherein *Klebsiella* sp was the most predominant isolation. In yet another study from Odisha, *Pseudomonas* trailed behind *Staphylococcus aureus* and *Klebsiella*, among the isolated bacterial pathogens from wound infections (Santa Otta et al, 2015 [8]. Suryakala R Nair et al, 2015 [9], showed that *Pseudomonas aeruginosa* was the second predominant isolation behind *Staphylococcus aureus*, from diabetic foot ulcers. The work was carried out from Trivandrum, Kerala. This organism demonstrates resistance to multiple antibiotics, there by jeopardizing the selection of appropriate treatment [10]. Carbapenem resistance is the dominant one due to loss of carbapenem specific porin OprD2 [11]. There are few studies from India that provide antimicrobial susceptibility of *P. aeruginosa* isolates [12, 13]. But, there are, so far, no reports of this nature from this part (Karakonam, Trivandrum district) of South Kerala, India.

2. Materials and Methods

This work was carried out in the department of Microbiology, Dr. SM CSI Medical College, Karakonam,
Trivandrum district, Kerala, India. Pus and discharges from various wound infections, mostly from inpatients, were collected for a 11 months period from January 2017 to November 2017. The samples collected were from surgery, ENT, medicine, intensive care units, critical care units, OG, dermatology, peripheral centre and OPDs. Out of the total of 1404 samples collected, 204 (14.5%) yielded *Pseudomonas aeruginosa* on culture. The media used were blood agar and MacConkey agar from HiMedia Mumbai. The isolations were identified based on colony and biochemical characteristics and also bluish-green diffusible pigment production (Figure 1).

![Figure 1. Pigment production and Antibiotic susceptibility pattern of Pseudomonas aeruginosa isolate- Kirby Bauer disc diffusion method.](image)

*Pseudomonas aeruginosa* strains were subjected to antibiotic susceptibility testing by Kirby-Bauer disc diffusion method in Mueller Hinton agar, HiMedia, Mumbai, (Figure 1), based on the CLSI guide lines [14, 15]. ATCC strains of *Pseudomonas aeruginosa* (ATCC 27853) and *Escherichia coli* (ATCC 25922) were used as quality control. All isolates in this study were non-duplicate. The antibiotic discs used were Amikacin (30 mcg), Gentamicin (20 mcg), Piperacillin (100 mcg), Piperacillin + Tazobactam (10 mcg), Imipenem (10 mcg), Cefoperazone (75 mcg), Ceftazidime (30 mcg), Netilmicin (30 mcg), Ciprofloxacin (5 mcg) and Meropenem (10 mcg), purchased from HiMedia, Mumbai. After performing the test, the plates were incubated at 37°C overnight. The diameter of the zone of inhibition was measured and compared to that of standard strains and the results were interpreted as sensitive or resistant, based on CLSI guidelines [16].

3. Results

Out of the total of 1404 samples cultured, 204 (14.5%) yielded *Pseudomonas aeruginosa*. Male patients predominated over the females in the number *Pseudomonas* isolates obtained (Table 1).

<table>
<thead>
<tr>
<th>Sex</th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of isolates</td>
<td>130</td>
<td>74</td>
</tr>
<tr>
<td>% of isolates</td>
<td>63.7</td>
<td>36.3</td>
</tr>
</tbody>
</table>

The maximum number of *Pseudomonas* isolations were made from the age group of >60 (33.8%), followed by 51-60 and 41-50 (Table 2).

<table>
<thead>
<tr>
<th>Age group</th>
<th>0-10</th>
<th>11-20</th>
<th>21-30</th>
<th>31-40</th>
<th>41-50</th>
<th>51-60</th>
<th>&gt;60</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of isolates</td>
<td>9</td>
<td>2</td>
<td>11</td>
<td>14</td>
<td>35</td>
<td>64</td>
<td>69</td>
</tr>
<tr>
<td>% of isolates</td>
<td>4.4</td>
<td>1.1</td>
<td>5.4</td>
<td>6.8</td>
<td>17.2</td>
<td>31.4</td>
<td>33.8</td>
</tr>
</tbody>
</table>

The most number of isolations were made from the samples received from male surgical ward (MSW) (31.9%), followed by a satellite multi-speciality hospital attached to this medical college, with equal distributions from female surgical ward and ENT department, in that order (Table 3).

<table>
<thead>
<tr>
<th>Ward</th>
<th>MSW</th>
<th>FSW</th>
<th>ENT ward</th>
<th>KK</th>
<th>MMW</th>
<th>MICU</th>
<th>SICU</th>
<th>CCU</th>
<th>ANW</th>
<th>Skin ward</th>
<th>OP</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of isolates</td>
<td>65</td>
<td>23</td>
<td>23</td>
<td>40</td>
<td>10</td>
<td>4</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>35</td>
<td></td>
</tr>
<tr>
<td>% of isolates</td>
<td>31.9</td>
<td>11.2</td>
<td>11.2</td>
<td>19.6</td>
<td>3.4</td>
<td>2</td>
<td>2</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>17.2</td>
</tr>
</tbody>
</table>

MSW- Male surgical ward, FSW- Female surgical ward, KK- peripheral multi-speciality hospital, MMW- Male medical ward

Among the antibiotics used, the maximum number of *Pseudomonas* isolates were found sensitive to Imipenem (81.3%), followed by Piperacillin + tazobactam (76.4%), Meropenem (70.5%) and Piperacillin (67.6%), (Table 4, 5) and (Figure 2).

<table>
<thead>
<tr>
<th>Antibiotics</th>
<th>Ak</th>
<th>G</th>
<th>Pc</th>
<th>Pt</th>
<th>Tb</th>
<th>Im</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of isolates</td>
<td>S</td>
<td>R</td>
<td>S</td>
<td>R</td>
<td>S</td>
<td>R</td>
</tr>
<tr>
<td>% of isolates</td>
<td>64.7</td>
<td>35.3</td>
<td>49</td>
<td>51</td>
<td>67.6</td>
<td>32.4</td>
</tr>
</tbody>
</table>

Ak- Amikacin, G- Gentamicin, Pc- Piperacillin, Pt- Piperacillin + tazobactam, Tb- Tobramycin, Im- Imipenem
## Table 5. Antibiotic sensitivity pattern of Pseudomonas isolates (n=204).

<table>
<thead>
<tr>
<th>Antibiotics</th>
<th>Cip</th>
<th>Mrp</th>
<th>Cpz</th>
<th>Caz</th>
<th>Nt</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S</td>
<td>R</td>
<td>S</td>
<td>R</td>
<td>S</td>
</tr>
<tr>
<td>No. of isolates</td>
<td>101</td>
<td>103</td>
<td>144</td>
<td>60</td>
<td>124</td>
</tr>
<tr>
<td>% of isolates</td>
<td>49.5</td>
<td>50.5</td>
<td>70.5</td>
<td>29.5</td>
<td>60.7</td>
</tr>
</tbody>
</table>

Cip- Ciprofloxacin, Mrp- Meropenem, Cpz- Cefoperazone, Caz- Ceftazidime, Nt- Netilmicin

## Figure 2. Antibiotic susceptibility pattern of Pseudomonas isolates in % (n=204).

Among the 3rd generation cephalosporins used, Ceftazidime and Cefoperazone recorded sensitivity of 65.6% and 60.7% respectively. Of the aminoglycosides, Amikacin, Netilmicin and Gentamicin recorded 64.7%, 61.7% and 49% respectively. Ciprofloxacin amounted 49.5% and Tobramycin showed 50% sensitivity. Fifteen (7.4%) multidrug resistant strains were reported out of the total 204 isolations.

### 4. Discussion

The isolation rate of *Pseudomonas aeruginosa* of 14.5% in our study was less than that of another study of wound infections in Varanasi, India (32%), [17]. The age group of >60 and 51-60 accounting for maximum number of isolations, in our study, more or less correlates with another study in North India [17]. Predominant number of *Pseudomonas* isolates made from cases in male surgical ward in this study differ from another report from New Delhi, India, wherein, neurosurgery cases dominated [18]. Most number of isolates being sensitive to Imipenem (81.3%), Piperacillin+ Tazobactam (76.4%), and Piperacillin (67.6%), in the decreasing order, in our study is in correlation with Deepak Juyal et al [19] and Viren A Javia et al [20], wherein, they showed a sensitivity pattern of 71.3%, 69.2% and 52% respectively. A novel finding in this study was that Imipenem, Piperacillin + Tazobactam, and Piperacillin were found to be sensitive to more number of isolates, than as reported by the studies quoted above. Among the aminoglycosides, Amikacin and Gentamicin recorded 64.7% and 49% sensitivity, respectively in our study, which is more or less in correlation with Deepak Juyal et al, wherein they reported 72% and 48.9% correspondingly [19]. Although, Ciprofloxacin in our study showing a sensitivity of 49.5%, is well above the 24.5% as mentioned in the same study as quoted above [19]. Anyway, as per our findings, there is a gradual and constant increase in resistance to aminoglycosides and fluoroquinolones, which is a matter of concern. 15 MDR strains also reported in this study added up to the agony, and as a result, evaluation of susceptibility pattern of *Pseudomonas aeruginosa* strains isolated at regular intervals is required for effective treatment for those cases.

### 5. Conclusion

In our study on antibiotic susceptibility pattern of *Pseudomonas aeruginosa* isolates, the percentage of resistant strains was 51 and 35.3 respectively against Gentamicin and Amikacin. 34.4% were resistant to Ceftazidime, a 3rd generation cephalosporin. 50.5% of the strains were resistant to Ciprofloxacin. Piperacillin showed 32.4% resistance. So, infections due to those *Pseudomonas* strains have to be treated only after performing the antibiotic susceptibility testing by including these drugs. 15 multidrug resistant strains emerging out of the 204 total isolates, prompts one to detect the gene responsible for such resistance in future studies. In our study, the most number of strains were susceptible to Imipenem (81.3%), Piperacillin + tazobactam (76.4%) and Piperacillin (67.6%), which could be used as reserve drugs in case of severe illnesses, due to *Pseudomonas* infection. Increase in resistance to antibiotics used other than Imipenem and Piperacillin + tazobactam and also emergence of 15 MDR strains of *Pseudomonas aeruginosa* in our study...
is a matter of concern in all health care establishments. As a result regular antibiotic susceptibility testing of *Pseudomonas* isolates is required for effective treatment of those cases.

Acknowledgements

I am thankful to Dr. S. Rema Devi, Professor and Head of the Department of Microbiology, Dr SM CSI Medical College and Hospital, Karakonam, Kerala, India, for permitting me, as well as extending work related expertise to complete this work. I am also grateful to the Director and the Principal of this Institution for the constant encouragement in research related activities.

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