Acinetobacter- MDR A Bug in Burn Patients: A Retrospective Study Conducted In a Tertiary Care Hospital

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Abstract:
Background: Acinetobacter recently gained interest as a cause of serious wound infection particularly in burn patients. Despite considerable advancements in burn wound care and infectious control practices, infection remains the leading cause of death. The most frequently recovered organism depends on a patient’s normal flora, duration of hospitalization, technique of selection.
Objectives: The purpose of this study was to evaluate the prevalence and improve our knowledge about increased emergence of MDR-Acinetobacter in burn patients and difficulty in treatment due to large number of virulence factors and antimicrobial resistance genes.
Material & Methods: Acinetobacter isolated from the burn patients in Christian Medical College & Hospital, Ludhiana were investigated on the basis of biochemical tests and their ability to antimicrobial susceptibility patterns. Isolates that were susceptible to antimicrobial agents varied according to the class of antimicrobial drugs. Only 4.16% isolates were sensitive to Ampicillin, 6.5% sensitive to Gentamycin, 48% sensitive to Amikacin, 27% to Cefotaxime, 21% to Cepherazone, 39% to Imipenem, 58% to Polymyxin B etc.
Conclusion: So, this can be concluded from the study that Acinetobacter contributed significantly to morbidity and mortality in burn patients. The prevalence of Acinetobacter gained interest during last few decades due to its resistance to all antimicrobial agents.
Keywords: MDR: Multidrug resistance, XDR: Extensively drug resistance, PDR : Pan drug resistance

I. Introduction
Acinetobacter recently gained interest as a cause of dreadful wound infection [¹-⁴]. It is also a hospital acquired pathogen. Its propensity to contaminate the hospital environment and to become resistant to most of the antibiotics is a major concern [⁵-⁶].

Acinetobacter is a gram-negative coccobacillus that has emerged as a new threat to burn patients. It is non-motile, encapsulated and non-fermentative [⁷]. It belongs to family Neisseriaceae and sometimes it can be misidentified as Neisseria or Moraxella on gram staining [⁸].

Nowadays, there are more than 20 species of Acinetobacter [⁹]. The most common among them is Acinetobacter baumannii, formerly called as Acinetobacter calcoaceticus. This species makes up to 80% of total Acinetobacter clinical isolates [¹⁰, ¹¹, and ¹²]. Acinetobacter can be grown from several human sources that include skin, pharynx, sputum, urine, vaginal secretions and stool [¹³]. It cause wide spectrum of infections including pneumonia, meningitis, bacteremia, soft tissue infections, surgical site infections, peritonitis, endocarditic, catheter – related infections and urinary tract infections [¹⁴-¹⁵].

The most striking feature of Acinetobacter is their ability to develop multdrug resistance mechanism against major antibiotic classes. Interest in Acinetobacter, from both the scientific and public community, has risen sharply over recent years. Significant advances have been made in understanding of this organism. The present study was conducted with the purpose to evaluate the prevalence and improve our knowledge about increased emergence of MDR-Acinetobacter in burn patients and difficulty in treatment due to large number of virulence factors and antimicrobial resistance genes.

II. Material & Methods
The present study was a retrospective study conducted in Burn unit of Christian Medical College & Hospital (CMC&H) Ludhiana from January 7 – March 16 2013. A total of 320 samples were processed according to all standard microbiological protocol. Inclusion criteria: Patients from all age-group and both sexes were included in the study and patients who were already on antibiotic therapy were excluded from the study.
Sample collection and Processing: A wound swab (pus) was the most common type of specimen collected under all aseptic conditions. All the samples were incubated for 24hrs at 37°C and were cultured on Blood agar and MacConkey agar. The culture plates were then observed for growth of bacteria and were subjected to preliminary biochemical tests for identification. Suspected colonies were identified by Gram’s stain. Gram stained smear of pus was prepared to observe relative number of polymorphs and bacteria, different morphological forms of gram positive and gram negative bacteria. Acinetobacter isolated from clinical specimens were assessed using Kirby Bauer disk diffusion test by inoculation on Muller Hilton agar and incubating the plates at 37°C for 24hrs. According to the zone size of antibiotics used, reporting was done and organisms were considered resistant, sensitive and intermediate.

III. Result

The present study was conducted from January 2013 to March 2013. A total of 320 bacterial isolates were processed. 22 patients were admitted to burn unit. 48 isolates were having Acinetobacter. Most of Acinetobacter isolates were resistant to broad spectrum antibiotics. Mean age of the patients was 21-58 yrs with Mean burn level range from 20% - 85%.

<table>
<thead>
<tr>
<th>Name of organism</th>
<th>No. of organism</th>
<th>%age</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pseudomonas</td>
<td>140</td>
<td>43.75%</td>
</tr>
<tr>
<td>Acinetobacter</td>
<td>48</td>
<td>15%</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>46</td>
<td>14.3%</td>
</tr>
<tr>
<td>Klebsiella</td>
<td>41</td>
<td>12.8%</td>
</tr>
<tr>
<td>E.coli</td>
<td>23</td>
<td>7.18%</td>
</tr>
<tr>
<td>Proteus</td>
<td>6</td>
<td>1.8%</td>
</tr>
<tr>
<td>Enterobacter</td>
<td>6</td>
<td>1.8%</td>
</tr>
<tr>
<td>Citrobacter</td>
<td>9</td>
<td>2.8%</td>
</tr>
<tr>
<td>Candida</td>
<td>3</td>
<td>0.93%</td>
</tr>
</tbody>
</table>

Positive cultures that were having mixed growth in 37 isolates (77.08%) with Gram – positive organisms in 6 isolates and Gram – negative organism in 31 isolates. Patients had a positive culture with Acinetobacter in 11 isolates (22.91%)

Broad spectrum antibiotics used were carbapenems, fluoroquinolones, aminoglycosides, polymyxin, cephalosporins, commercially named as: (Ampicillin, Gentamycin, Amikacin, Cefotaxime, Cefperazone, Ceftazidine, Imipenem, Meropenem, Cefper sulbactum, Ciprofloxacin, Netromycin, Ofloxacin, Tobramycin, Ticarcillin, Polymyxin B, Colisitin and Azithromycin)
As per our study, the isolate displayed resistance to:

- Ampicillin = 100%, Gentamycin = 95.83%, Amikacin = 77.08%, Cefotaxime = 50%, Cefperazone = 75%, Ceftazidime = 75%, imipenem = 25%, Meropenem = 60%, Cefper sulbactum = 56.25%, Ciprofloxacin = 85.41%, Netromycin = 79.16%, ofloxacin = 97.91%, Tobramycin = 60.41%, ticarcillin = 93.75%, Polymyxin B = 64.58%, Colisitin = 62.5%, azithromycin = 97.91%

- Defining MDR (multi drug resistance) as resistance to cephalosporin, fluoroquinolones and aminoglycosides in 16% isolates.

- Defining XDR (extensively drug resistance) as resistance to MDR Acinetobacter + resistance to carbapenems in 26% isolates.

- Defining PDR (pan drug resistance) as resistance to XDR Acinetobacter + resistance to Polymyxins in 3.63% isolates.

According to the data given above it is clear that Acinetobacter isolated showed highest resistance against Ampicillin (100%), azithromycin (97.91%), ofloxacin (97.91%), gentamycin (95.83%), ticarcillin (93.75%) and showed less resistance against imipenem (25%). Maximum activity was showed against imipenem.

IV. Discussion

Infection in burned patients remains one of the main contributors to morbidity and mortality. In past few years, Acinetobacter has emerged as a common pathogen in burn units. Acinetobacter strains are nonfermenting, aerobic, gram negative coccobacillary organisms. They can be found occasionally colonizing skin, gastrointestinal tract [16]. Multidrug resistance Acinetobacter has emerged as a problem worldwide [17]. Such strains are resistant to all beta – lactam and fluoroquinolones and require therapy with colisitin and amikacin but in our study resistance to colisitin and amikacin were found. Emergence of resistance to multiple antimicrobial agents in Acinetobacter has become a significant public health threat. Multidrug resistance is labeled as such because of their resistance to more than one antimicrobial agent. Infection with MDR’s can lead to delayed antimicrobial therapy.

As per our study, 26% isolates show XDR Acinetobacter. Bacteria that are classified as XDR are epidemiologically significant not only due to their resistance to multiple antimicrobial agents but also resistance to almost all approved antimicrobial agents [18]. In our study, 3.63% isolates show PDR (pan drug resistance) means resistance to all antimicrobial agents. ‘PAN’ is a Greek word meaning ‘ALL’.

According to one study, the majority of Acinetobacter isolates were sensitive to cephalosporins and imipenem. In 1995, Lyttikainen et al. [19] described their experience during the course of 4 years (1989 – 1993) and found resistance to imipenem increased from 1.5% to 7% and resistance to tobramycin increased from 5% to 12% [20]. Pothers have reported resistance to imipenem in range of 11% to 24% [21]. In another report by Hansberger in 1999 analyzing data from 5 European countries, Acinetobacter species were found to have highest increase in resistance to antibiotics [22].

Figure 2 showing increase in resistance to Imipenum between 1997 to 2002
Similarly Paul and colleagues from Israel found an increase in resistance to imipenem from 10% - 34% between 1997 and 2002 [23]. Comparing to our data, 25% isolates show resistance to imipenem.

In our study Acinetobacter displayed much higher resistance to Ampicillin (100 %). It may be because of frequent use of ampicillin; 16% isolates showed MDR, 26% showed XDR and 3.63% showed PDR.

In MDR Acinetobacter, Colisitin is drug of choice and can be used in last resort. Colisitin was first introduced in 1952 and became available in 1960’s but was replaced in 1970’s with other antibiotics due to its toxicity [24]. There are 2 forms of colisitin available in market: colisitin sulfate and colisitin methanosulfonate [25]. Colisitin methanosulfonate or CMS was replaced by aminoglycosides due to its neurotoxicity.

Figure 3 represents 'colisitin 'or 'colisitin resistance 'from 1960 to middle 2011.

This graph shows the trend in colisitin use, which increased from 21st century. According to our study 62.5% isolates show resistance to colisitin. The mechanism of resistance might be loss of lipopolysaccharide and increased use has led to the discovery of resistant strains. This study is based on retrospective design. To the best of our knowledge, this is the largest series of Acinetobacter in burn patients to date and it throw light on new ideas for management of infections related to Acinetobacter. Acinetobacter infections do not affect mortality independently. Clinicians should consider these data when treating Acinetobacter infections.

V. Conclusion
Acinetobacter has emerged as a highly troublesome pathogen worldwide. Due to its immense ability to acquire antibiotic drug resistance, it has been propelled to the forefront of scientific attention. Apart from its predilection for the seriously ill patients within intensive care units, Acinetobacter has more recently caused a range of infectious syndromes in burn patients. This study details the significant advances, including current taxonomy, mechanisms of antibiotic resistance, epidemiology, clinical manifestation of infection, and treatment regarding Acinetobacter.

References
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