Prevalence of Multidrug Resistant (MDR) Proteus spp. in Burn Wound Infection of a Tertiary Care Hospital, Rajshahi

Anika Tasnim¹, Md. Shah Alam², Md. Abdullah Yusuf³, Fazlay Akbar Khan⁴, Jannatul Ferdose⁵, Mahmuda Sultana⁶

¹Department of Microbiology, Parkview Medical College, Sylhet, Bangladesh
²Department of Microbiology, Rajshahi Medical College, Rajshahi, Bangladesh
³Department of Microbiology, National Institute of Neurosciences and Hospital, Dhaka, Bangladesh
⁴Department of Orthopedic Surgery, Sylhet MAG Osmani Medical College, Sylhet, Bangladesh
⁵Pathology and Microbiology Department, National Institute of Diseases of Chest and Hospital, Mohakhali, Dhaka, Bangladesh
⁶Department of Anatomy, Parkview Medical College, Sylhet, Bangladesh

Email address: anika_46@yahoo.com (A. Tasnim), drmshahalam1963@gmail.com (M. S. Alam), ayusuf75@yahoo.com (M. A. Yusuf), fazlayakbarhakan@gmail.com (F. A. Khan), jferdose20@gmail.com (J. Ferdose), mahmudasultana90@gmail.com (M. Sultana)

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Abstract: Background: Multidrug resistant (MDR) bacteria are an emerging public health issue in all over the world especially in developing countries like Bangladesh. Objective: This study was aimed to determine the prevalence of MDR Proteus species in burn wound infected patients admitted in Burn and Plastic Surgery Unit, Rajshahi Medical College Hospital (RMCH). Methodology: This cross sectional study was conducted at Microbiology Department of Rajshahi Medical College Hospital, Rajshahi, Bangladesh. A total of 212 wound swabs were collected and processed as per standard protocol from January to December 2016 for a period of one year. The isolation and identification of Proteus species was done by conventional microbiological process and antimicrobial susceptibility was performed by Modified Kirby-Bauer disk diffusion method. The Proteus species was further classified into MDR on the basis of standardised international criteria. Result: Among the 196 isolates from 212 wound swabs, Proteus spp. was the 2nd most frequent isolated organism (23.47%) following Pseudomonas aeruginosa (34.18%). Two species of Proteus were identified; Proteus mirabilis (65.22%) and Proteus vulgaris (34.78%). More than 75.0% Proteus spp. showed resistance to ciprofloxacin and ceftriaxone where meropenem was the highest sensitive drug (>93%). MDR Proteus spp. were 63.04% among which Proteus mirabilis and Proteus vulgaris were 60% & 68.75% MDR respectively. Conclusion: This study culminates the high prevalence of MDR Proteus spp. in Burn Unit of RMCH and decreased sensitivity to commonly used antibiotics. Therefore a greater emphasis on prevention of MDR bacterial colonization and antibiotic stewardship are imperative.

Keywords: Burn Wound Infection, Multidrug Resistant (MDR), Proteus spp.

1. Introduction

Burn wound infection still persists as one of the main source of mortality and morbidity [1]. It provides moist and nourishing surroundings conducive to microbial colonization, proliferation and infection [2]. Burn patients are infected by hospital-acquired bacteria by numerous invasive and non-invasive procedures [1].

Different species of microorganisms are responsible for burn wound infection that varies in their frequency with geographical location. Among the Gram negative bacilli Proteus spp. is one of the culprits. The genus Proteus belongs to the tribe of Proteacea within the Enterobacteriaceae family consisting of Proteus, Providencia and Morganella genera. *P. mirabilis*, *P. vulgaris*, *P. penneri*, and *P. myxofaciens* are the four species that belongs to Proteus genus. The last one is
Insufficient in infections of human [3].

A variety of opportunistic nosocomial infections are caused by Proteus spp. that includes respiratory tract, ear, nose, skin, burns, and wounds [3]. Proteus mirabilis is the species most frequently recovered from human, particularly from wound infections and urinary tract accounting 90% of all infections caused by the Proteus spp. [4]. It is also familiar to find Proteus mirabilis bacilli in immunocompromised individuals like infected burn wounds [5].

Nowadays, majority of the bacteria that cause nosocomial burn infection shows resistant to at least one of commonly used antimicrobials [1]. A wide range of variables (biological, pharmacological and societal) are responsible antimicrobial resistance that occurs globally particularly in developing countries like Bangladesh [6]. The excessive and misuse of antibiotics in treating bacterial infections have evolved in the emergence of resistant strains that are difficult to treat [5].

At present the World Health Organization (WHO) has notified MDR bacteria as the most threatened issue that created negative impacts on prevention and treatment of bacterial infections [6]. The International Expert Proposal for Interim Standard Definitions for Acquired Resistance had defined MDR, which entailed non-susceptibility to three (3) or more specified groups of antimicrobials [7].

The significant evolution and rise of multidrug-resistance of numerous bacterial isolates is escalating day by day which has evoked a growing public health issue in the world. MDR Proteus spp. necessitates frequent monitoring of antimicrobial susceptibility pattern aiming to apply appropriate antimicrobial therapy [8]. Therefore, this study was performed to determine the prevalence of MDR Proteus spp. from burn wound infected patients in RMCH, Bangladesh.

2. Materials and Methods

This cross sectional study was carried out at Microbiology Department of Rajshahi Medical College (RMC) among burn wound infected patients admitted in Burn and Plastic Surgery Unit of RMCH, Rajshahi, Bangladesh from January to December 2016. Patients who had the history of burn more than 3 days irrespective of age, sex and duration of hospital stay were included in the present study. All relevant information was recorded in a predesigned data sheet.

2.1. Isolation and Identification of Proteus Species

The wound swabs were collected aseptically and were processed by standard microbiological methods at Microbiology Department of RMC. Samples were inoculated in Blood agar media, Nutrient agar media and MacConkey’s agar media aerobically at 37°C for 24 hours. Colonies that were non-lactose fermenting on MacConkey agar and showed swarming on Blood agar were isolated and identified by biochemical tests based on whether they were positive for phenylalanine deaminases production; H₂S gas production; and urease reactions. P. vulgaris produces indole which differentiated it from indole negative P. mirabilis.

2.2. Antimicrobial Susceptibility Test

Isolated bacteria were tested for antimicrobial susceptibility by modified Kirby-Bauer disk diffusion method using Mueller-Hinton agar media and commercially available antimicrobial disks following the guideline of CLSI 2015 [9]. Gram Negative bacteria were tested for amikacin (30 µg), gentamycin (10 µg), ciprofloxacin (5 µg), cefuroxime (30 µg), ceftriaxone (30 µg), ceftazidime (30 µg), amoxiclav (20/10 µg) and meropenem (10 µg). The isolates were considered as MDR when non-susceptibility to at least one agent in three or more antimicrobial categories [7]. Six frequently used classes of antibiotics were used to analyze MDR.

3. Result

Out of 212 wound swabs 89.62% yielded growth in culture and 196 bacterial species were isolated. Pseudomonas aeruginosa was the predominant organism (34.18%) followed by Proteus spp., the 2nd highest isolated organism (23.47%) in burn wound infection (Table 1). Proteus mirabilis and Proteus vulgaris were identified among 46 Proteus spp. Proteus mirabilis was most frequently isolated species (65.22%) followed by Proteus vulgaris (34.78%) (Figure 1).

| Isolated organisms          | No  | Percent |
|-----------------------------|-----|---------|
| Pseudomonas aeruginosa      | 67  | 34.18   |
| Proteus spp.                | 46  | 23.47   |
| Klebsiella spp.             | 29  | 14.80   |
| Escherichia coli            | 23  | 11.73   |
| Staphylococcus aureus       | 21  | 10.71   |
| Acinetobacter spp.          | 04  | 2.04    |
| CoNS                        | 04  | 2.04    |
| Enterobacter spp.           | 02  | 1.03    |
| Total Isolates              | 196 | 100     |

Note: N=Total sample (wound swab) number.

Figure 1. Distribution of isolated Proteus spp.

Distribution of Proteus spp. was analyzed in different age group. Proteus infection was highest in the age group 21-30 years followed by 31-40 years (Table 2).
Regarding antimicrobial susceptibility *Proteus* spp. were highly resistant nearly >75% to ciprofloxacin & ceftriaxone and more than 65% to gentamycin & ceftazidime. Meropenem was the highest sensitive drug (>90%) followed by amikacin (>60%) (Figure 2).

![Figure 2. Antimicrobial resistant pattern of Proteus spp.](image)

In this study the prevalence of MDR *Proteus* spp. was high. More than 60% of all isolates were found to be MDR among which *Proteus mirabilis* were 60% and *Proteus vulgaris* were 68.75% (Table 3).

### Table 2. Distribution of Proteus spp. in different age group (n=46).

| Age (years) | Proteus mirabilis | Proteus vulgaris | Total no |
|-------------|-------------------|------------------|----------|
| ≤10         | 04                | 01               | 05       |
| 11-20       | 05                | 02               | 07       |
| 21-30       | 09                | 07               | 16       |
| 31-40       | 08                | 03               | 11       |
| 41-50       | 03                | 01               | 04       |
| 51-60       | 01                | 00               | 01       |
| >60         | 00                | 02               | 02       |
| Total       | 30                | 16               | 46       |

Note: n=Total number of Proteus spp.

### Table 3. Multidrug resistant (MDR) Proteus spp.

| Organism      | Total no | No of isolates found to be MDR |
|---------------|----------|--------------------------------|
| *Proteus mirabilis* | 30       | 18 (60.00%)                    |
| *Proteus vulgaris* | 16       | 11 (68.75%)                    |
| Total         | 46       | 29 (63.04%)                    |

Figures in parenthesis represent percentage.

### 4. Discussion

The onset of infectious diseases and drug resistance are very common in developing and densely populated country like Bangladesh [10]. Therefore to manage and control the infection rate, species identification and antimicrobial resistance surveillance is essential.

Out of 196 isolates, *Pseudomonas aeruginosa* was the most frequent microbial isolates (34.18%) in this study. In Bangladesh, *Pseudomonas aeruginosa* was reported as the predominant organism of burn wound infection in 39% cases which was in agreement with this study [11]. *Proteus* spp. the 2nd most frequent bacterial isolate, were 23.47%, which was in accordance with other studies done in Bangladesh and India [12, 13]. However, in contrast to this findings, a very lower isolation of *Proteus* spp. was also reported [14, 15].

In the present study two species of *Proteus* (*Proteus mirabilis* and *Proteus vulgaris*) were identified to be responsible for burn wound infection. *Proteus mirabilis* was the most commonly isolated species (65.22%) and hence accountable for the most of the *Proteus* infection which was followed by *Proteus vulgaris* (34.78%). This finding was consistent with other studies conducted in Ghana [16] and India [9, 17]. According to Mordi and Momoh, a lower isolation of *P. mirabilis* and *P. vulgaris* was also reported [4]. The occurrence of burn wound infection in different age group by *Proteus mirabilis* and *Proteus vulgaris* was analyzed. The incidence was higher in 21-30 years age group which was quite similar with studies done in India [9].

Antimicrobial susceptibility pattern revealed that *Proteus* spp. were resistant to commonly used antibiotics like 3rd generation cephalosporin, quinolones etc which are being used indiscriminately on empirical basis for prolong duration of time. In this study *Proteus* spp. were >75% resistant to ciprofloxacin & ceftriaxone and >65% to gentamycin & ceftazidime. Meropenem was the highest sensitive drug (>90%) followed by amikacin. Previous reports within or outside Bangladesh showed similar findings [5, 12, 18]. Cent percent sensitivity to ciprofloxacin, gentamycin & >80% to ceftriaxone were also reported which was unlike to this study [6, 19].

*Proteus* spp. showed a high level of MDR (63.04%). Among them 60% of *Proteus mirabilis* and 68.75% of *Proteus vulgaris* were MDR which was in agreement with other studies published in India [9, 15], Ghana [16] and in Egypt [1]. The high incidence of MDR *Proteus* may be due to the inappropriate use of broad spectrum antimicrobial without sensitivity testing. This findings, however, in contrast with Pandey et al., who had reported a very low level of MDR *Proteus* spp. [17].

### 5. Conclusion

Immense bacterial proliferation and high prevalence of MDR *Proteus* spp. were observed in the present study. Commonly used 3rd generation cephalosporin and quinolone group of drugs were highly resistant whereas meropenem was the most sensitive drug. This highlights the alarming levels of antimicrobial resistance and need for applying effective antibiotic therapy. Therefore, MDR bacteria should be identified along with their antibiogram by every burn center in order to help clinicians to choose the most suitable antimicrobial therapy for patient benefit as well as to reduce emergence of drug resistant bacteria.

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