Bacteriological Profile and Antibiogram of Cultures Isolated from Tracheal Secretions

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Abstract

Background
Respiratory infections are associated with high morbidity and mortality, especially in critically ill patients. The excessive use of broad-spectrum antibiotics has led to the development of drug resistance, thus resulting in the emergence of pathogens which are difficult to treat. The aim of this study was to identify common pathogens in tracheal secretions and to study the patterns of their sensitivity and resistance to various antibiotics.

Materials and methods
This descriptive cross-sectional study was conducted in the Department of Pathology and Microbiology, Holy Family Hospital, Rawalpindi, Pakistan, from August 2017 to December 2017, using the convenient sampling technique. Tracheal secretions from patients in the intensive care unit (ICU), tested in the Pathology and Microbiology Department of Holy Family Hospital, were included in the study. The culture was done on blood and MacConkey agar and the sensitivity pattern was performed on Muller Hinton agar. Data were analyzed using SPSS v.23.0.

Results
Out of the bacteria isolated from positive growth cultures, Acinetobacter (45; 53.6%) was the most common isolate followed by Klebsiella (11; 13.1%). Acinetobacter was most sensitive to tigecycline (94.7%), and gram-negative bacteria such as Acinetobacter, Klebsiella, and Pseudomonas showed resistance to higher generation cephalosporins.

Conclusion
Acinetobacter was the most common gram-negative bacilli isolated. Tigecycline was found to be effective against Acinetobacter.

Introduction
Tracheobronchial secretions are produced by mucous glands and goblet cells of the tracheobronchial tree [1]. These secretions are not only involved in the protection of the respiratory tract but are also responsible for the exchange of heat and water during breathing [2].

Respiratory infections are associated with high morbidity and mortality, especially in critically ill patients [3]. Such patients are usually maintained using invasive devices which themselves tend to be a major reservoir for hospital-acquired infections [4,5]. About 15% of hospital-acquired infections (HAIs) are caused by ventilator-associated pneumonia (VAP). This is the second-most-common HAI having the highest morbidity and mortality [6].

Moreover, the advent and increase in multi-drug resistant (MDR) pathogens serve as a major problem [7]. The excessive and indiscriminate use of broad-spectrum antibiotics has led to the development of these resilient microbes which are difficult to treat [8,9].

The aim of this study was to identify the common pathogens in tracheal secretions and to study the patterns of their sensitivity and resistance to various antibiotics, which can serve as guidelines to physicians for empirical treatment with proper antibiotics.
Materials And Methods
This descriptive cross-sectional study was conducted in the Department of Pathology and Microbiology, Holy Family Hospital, Rawalpindi, Pakistan, from August 2017 to December 2017. All patients whose tracheal secretion samples were tested in the Pathology and Microbiology Department of Holy Family Hospital were included in the study. In total, 186 samples were selected, regardless of age and gender, by using the convenient sampling technique. Tracheal secretions from patients admitted in the intensive care unit (ICU) for more than 48 hours were obtained by sterile suctioning using an endotracheal tube and suction catheter tip. Samples were inoculated on agar plates. The culture was done on blood and MacConkey agar and was incubated at 37°C for 24 to 48 hours. Microbes were identified under a microscope by observing morphological characteristics after gram staining and applying biochemical tests. Antibiotic sensitivity pattern was done on Muller Hinton agar using the Kirby Bauer disk diffusion method. Antibiotic discs containing amikacin, amoxicillin clavulanate (augmentin), aztreonam, ceftriaxone, ceftazidime, cefoperazone + sulbactam (sultane), ciprofloxacin, cefepime, imipenem, tigecycline, piperacillin + tazobactam (taxocin), vancomycin, penicillin, gentamicin, linezolid, chloramphenicol, erythromycin, methicillin, ampicillin, and moxifloxacin were obtained and used as per the manufacturer’s instructions.

Data were entered and analyzed using the Statistical Package for Social Sciences (SPSS) v.23.0 (IBM, Armonk, US). Descriptive statistics were applied to find frequencies and percentages. Charts and tables were constructed.

Results
A total of 186 samples were collected during this study period with 85 (45.7%) samples from males, 95 (51.1%) from females, while gender was missing for six (3.2%) patients. Positive growth was observed in 84 (45.2%) samples. The gram-negative bacilli contributed a major number of isolates (75; 68.4%), the remaining nine (31.6%) were caused by gram-positive cocci.

Out of the bacteria isolated from positive growth cultures, *Acinetobacter* (45; 55.6%) was the most common isolate followed by *Klebsiella* (11; 13.1%). There were seven cases (8.3%) of *Staphylococcus aureus* which included one (1.3%) methicillin-resistant *Staphylococcus aureus* (MRSA). The least common isolate was *Enterococcus* (2; 2.4%) (Figure 1).

![FIGURE 1: Bacterial growth in tracheal secretions (n=84)](image)

*Acinetobacter* was most sensitive to tigecycline (94.7%) while the gram-positive cocci, i.e., *Staphylococcus aureus* and *Enterococcus* showed susceptibility to vancomycin (100%) (Table 1).
| Antibiotics | **Staphylococcus aureus** | **Enterococcus** |
|-------------|--------------------------|-----------------|
| Vancomycin  | 100%                     | 100%            |
| Penicillin  | 0%                       | -               |
| Gentamicin  | 100%                     | 0%              |
| Linezolid   | -                        | 100%            |
| Chloramphenicol | 100%                   | -               |
| Erythromycin | 25%                     | 0%              |
| Methicillin | 0%                       | -               |
| Ampicillin  | -                        | 0%              |
| Moxifloxacin| -                        | 50%             |

**TABLE 1: Antibiotic sensitivity pattern of gram-positive cocci in tracheal secretions**

The gram-negative bacilli such as *Acinetobacter*, *Klebsiella*, and *Pseudomonas* showed resistance to higher generation cephalosporins such as ceftriaxone and cefepime (100%). The susceptibility pattern of gram-negative bacilli is shown in Table 2.

| Antibiotic                          | **Acinetobacter** | **Klebsiella** | **Pseudomonas** | **Escherichia coli** | **Coliforms** |
|-------------------------------------|-------------------|----------------|-----------------|----------------------|---------------|
| Amikacin                            | 4.7%              | 54.5%          | 22.2%           | 80%                  | 80%           |
| Amoxicillin clavulanate (augmentin) | 0%                | 0%             | -               | 0%                   | 40%           |
| Aztreonam                           | -                 | -              | 28.6%           | -                    | -             |
| Cefazidime                          | 0%                | 0%             | 20%             | 0%                   | -             |
| Cefoperazone + sulbactam (sulzone)  | 7.7%              | 81.8%          | 66.7%           | 50%                  | 60%           |
| Ciprofloxacin                       | 0%                | 0%             | 22.2%           | 20%                  | 50%           |
| Ceftriaxone                         | 0%                | 0%             | 0%              | 25%                  | 60%           |
| Cefepime                            | 0%                | 0%             | 0%              | 33.3%                | 50%           |
| Imipenem                            | 2.5%              | 70%            | 28.6%           | 100%                 | 40%           |
| Tigecycline                         | 94.7%             | -              | -               | -                    | -             |
| Piperacillin + tazobactam (tazocin) | 2.5%              | 71.4%          | 12.5%           | 50%                  | 75%           |

**TABLE 2: Antibiotic sensitivity pattern of gram-negative bacilli in tracheal secretions**

**Discussion**

The resistance to conventional antibiotics is severely increasing in bacteria in clinical and non-clinical settings [10]. The rate of nosocomial infection is also steadily mounting in the patients admitted in the ICU due to excessive invasive procedures performed including artificial ventilator support [11]. This constantly emerging resistance is a serious situation implying the need for new regulations for the cautious use of antibiotics and refining the conditions of hospitals to prevent further exacerbation of resistance shown by the bacteria.

The percentage of samples showing positive growth in our study was 45.2%. In a study conducted by Gupta et al., the percentage of positive growth was 55% [7]. In another study by Chandra et al., the positive samples were 72.3% [12]. In a study conducted in the setting of Pakistan by Malik et al., the positive cultures came out to be 83% [13]. This marked decrease in our study can be attributed to the better infection control measures in the ICU setup of our hospital. However, the convenient sampling technique used in our study...
Gram-negative bacilli were predominant in tracheal secretions with population cannot be carried out. The limitation of our research included the decreased time duration of the study. Moreover, the study was by Gupta et al. and Malik et al. The resistance to drugs such as carbapenems. This was in consistency with the pattern shown in the research cefepime. The broad-spectrum antibiotics showed sensitivity to an extent, but there was an alarming rise in the remaining gram-negative bacteria in our study displayed resistance to the higher generations of cephalosporins, fluoroquinolones, penicillin, and drugs such as ceftazidime, ceftriaxone, ciprofloxacin, and cefepime. The broad-spectrum antibiotics showed sensitivity to an extent, but there was an alarming rise in the resistance to drugs such as carbapenems. This was in consistency with the pattern shown in the research by Gupta et al. and Malik et al. [7,17].

The limitation of our research included the decreased time duration of the study. Moreover, the study was carried out in a single tertiary care hospital due to which the generalization of results to the whole population cannot be carried out.

**Conclusions**

Gram-negative bacilli were predominant in tracheal secretions with *Acinetobacter* being the most common...
species isolated. Tigecycline was found to be effective against Acinetobacter. The resistance to cephalosporins and penicillins is shown to be established by Acinetobacter, Pseudomonas, and Klebsiella. One of the grave concerns related to hospital-acquired infections is the upsurge of multi-drug resistance among the respiratory pathogens which have also extended into the community. There is a need for limited and objective use of antibiotic therapy according to the new guidelines modified under the light of such researches.

**Additional Information**

**Disclosures**

**Human subjects:** Consent was obtained by all participants in this study. Institutional Research Forum, Rawalpindi Medical University issued approval RSRS-2018-P-05. This research forum certifies that: 1. The above-mentioned study is within the ethical boundaries. 2. There is no duplication of the above-mentioned study in this institution. 3. The forum acknowledges the importance of the study. 4. None of the investigator and co-investigator participating in this study took part in the decision making and voting procedure for this study.

**Animal subjects:** All authors have confirmed that this study did not involve animal subjects or tissue.

**Conflicts of interest:** In compliance with the ICMJE uniform disclosure form, all authors declare the following:

- **Payment/services info:** All authors have declared that no financial support was received from any organization for the submitted work.
- **Financial relationships:** All authors have declared that they have no financial relationships at present or within the previous three years with any organizations that might have an interest in the submitted work.
- **Other relationships:** All authors have declared that there are no other relationships or activities that could appear to have influenced the submitted work.

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