Bacteriological profile and antibiotic sensitivity patterns of aerobic pus isolates: A study conducted in tertiary care hospital of North India

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Abstract

Background: Wound infection is an important cause of morbidity and mortality among hospitalised patients. Therefore knowledge of pathogens causing wound infection is necessary and can be helpful in selection of appropriate antimicrobial therapy. This study was undertaken to identify the bacterial pathogens recovered from infected wounds and characterise their antimicrobial resistance profile.

Materials and Methods: It was a cross-sectional study, carried out for a period of two years, from January 2017 to December 2018, in the Microbiology department of a tertiary care hospital in North India. During this time period all pus/wound swab samples received were analysed for the identification of microbiorganisms and for the determination of their antibiotic susceptibility. For data analysis SPSS version 17.0 software and MS excel 2007 were used.

Results: Out of total 2516 pus/wound swab samples, 1672 (66.45%) were positive for bacterial growth and of these 1672 positive culture, 1312 (78.46%) were pure isolates and 360 (21.53%) cultures had grown more than one organism (poly-microbial). So a total number of isolates under study was 2032. Of these 2032 aerobic culture isolates, 594 (29.23%) were Gram positive cocci and 1438 (70.76%) were Gram negative bacilli. The most common pathogen isolated was Escherichia coli (29.23%) followed by Staphylococcus aureus (20.47%), Klebsiella pneumoniae (12.3%) and Pseudomonas aeruginosa (11.12%). Most of the isolates obtained were multi-drug resistant.

Conclusion: Since the frequency of multiple drug resistance among both gram positive and gram negative bacteria is alarmingly high, therefore periodic monitoring of antimicrobial susceptibility profile of the infectious agents causing wounds infections in hospital settings is needed. This will benefit not only the patient but also assists physician in selection of appropriate chemotherapy.

Keywords: Pus isolates, Bacteriological profile, Antibiotic sensitivity patterns.

Introduction

Pus formation is one of several cardinal indicators of suppurative infections caused by pyogenic bacteria, resulting in aggregation of dead leukocytes, bacteria and tissue debris.1 Wound is a breach in skin integrity caused due to injury. Exogenous wounds are usually associated with traumatic injuries, burns etc. whereas endogenous wounds and abscesses may be associated with appendicitis, cholecystitis etc.2 Colonization and proliferation of bacteria in wound may lead to wound infection. Most of the latter are hospital acquired and usually following an invasive procedure or a surgical intervention. Hospital acquired infections are a world-wide in problem being an important cause of morbidity and mortality among hospitalized patients. A WHO sponsored survey showed that the prevalence of nosocomial infections was 3-21% with wound infections accounting for 5-34%.3 Therefore the knowledge of infectious agents causing wound infection is necessary for selection of appropriate antimicrobial therapy. Previous data have shown that most common pathogens associated with wound infections are Staphylococcus aureus, Enterococcus spp, Escherichia coli, Klebsiella spp, Pseudomonas spp, Proteus mirabilis, Candida albicans.4,5 The present study was undertaken to know the bacteriological profile and antibiotic susceptibility patterns of pathogens causing pyogenic infections in our hospital in order to help clinician formulate an empirical treatment for the patients.

Aims and Objectives

To identify and characterize aerobic bacterial pus isolates onto specie level
To determine antimicrobial sensitivity patterns of the isolates

Material and Methods

Study Design and Data Collection

It was a cross-sectional study, carried out over a period of two years, from January 2017 to December 2018, in the Bacteriological section of Microbiology department of a tertiary care hospital in North India. During this time period all pus/wound swab samples (n=2516) received for aerobic culture & sensitivity testing, from various clinical departments of the Institute, were include in the study.

Data collection included information about, age & sex of the patients from whom samples were taken, a brief history of illness.

All pus samples were processed as per standard bacteriological techniques for aerobic cultures.6 They were inoculated on both Blood agar and Mac-Conkey’s agar and incubated aerobically for 24-48 hours at 37 degree, before reporting them as sterile. Gram staining was performed on growth obtained in positive cultures. Isolates were identified to specie level by Vitek-2 Compact (Biomerieux) using gram positive, gram negative, and yeast identification cards as per manufacturer’s guidelines.

Antimicrobial sensitivity testing was also determined by same system using AST cards. Antibiotic sensitivity results were interpreted as per CLSI guidelines.7
Ethical approval: Ethical approval was taken from the ethical committee of the institution.

Statistical analysis: SPSS version 17.0 software and MS excel 2007 were used for statistic analysis.

Results
The study comprised of a total 2516 samples of patients with wound infection. Among these, those with positive cultures were 1672 (66.45%) and of these 984 (58.85%) samples were from male patients and 688 (41.15%) from females. Majority of these patients were in the age groups of 20–40 years (Table 1&2, Fig 1&2).

Bacterial Isolates
Out of total 2516 pus/wound swab samples, 1672(66.45%) were positive for bacterial growth and of these 1672 positive culture, 1312(78.46%) were pure isolates and 360(21.53%) cultures had grown more than one organism (polymicrobial). So the total number of isolates under study was 2032 of which 594(29.23%) were Gram positive cocci and 1438(70.76%) were Gram negative bacilli. The most common pathogen isolated was Escherichia coli (29.23%) followed by Staphylococcus aureus (20.47%), Klebsiella pneumoniae (12.3%) and Pseudomonas aeruginosa (11.12%), (Table 3). The antimicrobial sensitivity patterns of gram negative isolates of Enterobacteriaceae family, Non-fermenting isolates and gram positive isolates are listed in Tables 4, 5 and 6 respectively. Most of the gram negative isolates were multi-drug resistance (MDR). However most of the strains of S. aureus were sensitive to Vancomycin and Linezolid (Fig 5-7).

Table 1: Sex wise distribution of aerobic culture positive pus/wound swab samples (n=1672)

| Sex         | Patients with positive cultures |
|-------------|---------------------------------|
| Male        | 984 (58.85%)                    |
| Female      | 688 (41.15%)                    |

OR: 2.079; CI : 1.757-2.460; p <0.001; Highly significant

Table 2: Age wise distribution of aerobic culture positive pus/wound swab samples (n=1672)

| Patient’s age group | Number of positive samples |
|---------------------|----------------------------|
| < 20 years          | 170                        |
| 20-40 years         | 898                        |
| 40-60 years         | 206                        |
| > 60 years          | 398                        |

OR: 1.702; CI: 1.439 – 2.014; p < 0.001; Highly significant

Table 3: Categorization of aerobic bacterial isolates obtained from positive pus cultures (n=2032)

| Gram positive cocci (GPC) | Gram negative bacilli (GNB) |
|---------------------------|-----------------------------|
| Isolate                   | Number | %    | Isolate               | Number | %    |
| Staphylococci aureus      | 416    | 20.47| Escherichia coli      | 594    | 29.23|
| CONS                      | 138    | 6.79 | Klebsiella pneumoniae| 250    | 12.3 |
| Enterococcus sp           | 36     | 1.77 | Enterobacter sp       | 100    | 4.92 |
| Streptococcus sp          | 4      | 0.2  | Citrobacter sp        | 28     | 1.37 |
|                           |        |      | Proteus sp            | 64     | 3.14 |
|                           |        |      | Pseudomonas aeruginosa| 226    | 11.12|
|                           |        |      | Acinetobacter sp      | 176    | 8.66 |

Table 4: Antibiogram enterobacteriace

| GNB Isolated | E. coli | Klebsiella sp | Enterobacter sp | Proteus sp |
|--------------|---------|---------------|-----------------|------------|
| No of isolates | 594     | 250           | 100             | 64         |
| Antibiotic    | % Sensitive | % Sensitive | % Sensitive | % Sensitive |
| Ampicillin    | 4.7     | 4.8           | 0.0             |            |
| Gentamicin    | 58.9    | 27.2          | 24.0            |            |
| Amikacin      | 78.8    | 29.6          | 44.0            |            |
| Amoxy-clavulate| 12.5   | 10.4          | 4.0             |            |
| Cefoperazone-Sulbactam | 46.7 | 20.8          | 34.0            | 62.5       |
| Piperacillin-Tazobactam | 37.0 | 14.4          | 28.0            | 62.5       |
| Cefepime      | 16.2    | 12.0          | 16.0            | 21.9       |
Table 5: Anti-biogram Non fermenters

| GNB Isolated            | Pseudomonas Aeruginosa | Acinetobacter sp |
|-------------------------|------------------------|------------------|
| No of isolates          | 226                    | 176              |
| Antibiotic              | % Sensitive            | % Sensitive      |
| Cefazidime              | 38.6                   | 5.3              |
| Gentamicin              | 56.8                   | 8.8              |
| Piperacillin-Tazobactam | 40.9                   | 9.7              |
| Amikacin                | 64.8                   | 15.0             |
| Aztreonam               | 22.7                   | 9.7              |
| Cefoperazone-Sulbactam  | 51.1                   | 13.2             |
| Cefepime                | 44.3                   | 8.0              |
| Doripenem               | 47.7                   | 11.5             |
| Imipenem                | 61.4                   | 11.5             |
| Meropenem               | 58.0                   | 12.4             |
| Ciprofloxacin           | 45.5                   | 8.8              |
| Levofloxacin            | 34.1                   | 15.0             |
| Tigecycline             | 21.6                   | 61.1             |
| Trimethoprim/Sulfamethoxazole | 84.0         | 13.2             |
| Colistin                | 79.6                   | 85.2             |

Table 6: Anti-biogram Staphylococcus Aureus

| GPC Isolated                  | Staphylococcus Aureus |
|--------------------------------|-----------------------|
| No of isolates                | 416                   |
| Antibiotic                    | % Sensitive           |
| Cefoxitin                     | 25                    |
| Erythromycin                  | 48                    |
| Clindamycin                   | 70                    |
| Trimethoprim/Sulphamethoxazole| 35                    |
| Daptomycin                    | 91.8                  |
| Linezolid                     | 98                    |
| Teicoplanin                   | 92.6                  |
| Vancomycin                    | 97.9                  |
| Tetracycline                  | 79.8                  |
| Tigecycline                   | 84.6                  |
| Rifampicin                    | 86.0                  |
| Ciprofloxacin                 | 5.7                   |
| Levofloxacin                  | 2.4                   |
| Gentamicin                    | 57.2                  |

Discussion

Of the total of 2516 study subjects bacterial pathogens were isolated from 1672; isolation rate being 66.45%. This was higher than the previous studies done in Gondar (52%), Bahir Dar (53%), but lower than Dessie (70.5%), and Muluye (70.2%). Also 21.53% of the total growths showed poly-microbial growth. Open wounds provide an environment conducive to the growth of bacteria so these wounds can easily be invaded and colonised by them. This might have been a reason for poly-microbial growth.

There was preponderance of male (58.85%) over females (41.15%) as well as gram negative isolates (70.76%) over gram positive (29.23%), which was also shown by previous studies from India by Biradar A et al, Basu et al and Mantravadi et al. The pre-dominant
isolates in the present study was found to be *Escherichia coli* (29.23%) which was also seen in a similar study by Deepali et al. Other isolates in the decreasing order of isolation being *Staphylococcus aureus* (20.47%), *Klebsiella pneumoniae* (12.3%) and *Pseudomonas aeruginosa* (11.12%). A study on wound microbiology conducted by Bowler et al also implies that the normal microbial flora of the gut, oral cavity, skin and genitourinary mucous membranes contain bacteria that can easily colonize wounds especially the ones in close proximity to those sites so this could be reason for *E. coli* preponderance.

Antimicrobial sensitivity profile showed most of the gram negative isolates as multi-drug resistance. *E. coli* isolates were conspicuously found to be resistant to ampicillin (95.3%), Cephalosporins (91.6%) and co-trimoxazole (74.8%) cases. Similar results were also shown by other studies nationwide. These MDR strains were found to be sensitive mainly to Tigecycline and Colistin.

Among gram positive cocci, whereas 75% isolates of *Staphylococcus aureus* were resistant to Cefoxitin (MRSA) only 2.1% to Vancomycin. These finding were in agreement with those in Ethiopia, Nepal, and Italy where 83%, 60.6%, 74.2% of *Staphylococcus aureus* were found to be Methicillin resistance, respectively. *Staphylococcus aureus* also showed a high resistance to erythromycin (58%) but high (98%) sensitivity to linezolid.

**Conclusion**

The frequency of multiple drug resistance among both gram positive and gram negative bacteria is alarmingly high. Thus, rational use of antimicrobials along with a strict compliance of hospital infection control practices can go a long way in fighting the menace of antibiotic resistance should be practiced.

**Conflict of Interest:** None.

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