ORIGINAL ARTICLE

# Antibiotic Sensitivity Pattern of Pathogens Isolated from Pus Culture- A Tertiary Care Hospital Based Study

Singh A[1], Verma V[2], Singh R[3]


## ABSTRACT

**Background:** Microbial pathogens cause human skin and soft tissue infections (SSTI) and surgical site infections (SSI) after surgical procedures. These can result in the production of pus, yellowish fluid comprising of dead WBCs and cellular debris. The microorganisms responsible for pus production vary greatly in relation to their spectrum of prevalence in different hospital and also in their antibiotic sensitivity. Further, the antibiotic sensitivity also changes because of the emergence of resistant strains. It is therefore, important that the common bacterial pathogens causing infection in a particular hospital and their sensitivity should be known. This will help in the choice of prophylactic antibiotic and in initiating the empirical antibiotic prescription for the infected cases before the culture sensitivity report is made available which takes about 2-3 days. **Objective:** To identify the spectrum of aerobic bacteria which are responsible for SSTI and SSI and their antibiotic sensitivity pattern. **Method:** This cross sectional hospital based study was conducted in Nepalgunj Medical College and Teaching Hospital (NGMCTH), Kohalpur from January 2019 to November 2019. These pus swabs were obtained from the Department of Surgery and Department of Gynaecology & Obstretics. Samples were cultured in the Microbiology laboratory of NGMCTH, Kohalpur. Identification and characterization of isolates were performed on the basis of Gram staining and cultural characteristics. Antibiotic sensitivity test was performed in vitro by Bauer-Kirby method. Collected data were statistically analyzed using SPSS 20.0 and Microsoft Excel 2015. **Results:** During the study period, a total number of 311 pus swabs were obtained among which only 164 (52.73%) pus swab showed bacterial growth. Out of 164 pus swabs, 150 pus swabs yielded monomicrobial growth (150 bacterial isolates) and 14 pus swabs yielded polymicrobial growth (33 bacterial isolates). Gram Negative Bacteria (60.1%) was more prevalent than Gram Positive Bacteria (39.9%). Combined together, the most common isolate was S.aureus (36.1%) followed by E.coli (24.0%), Klebsiella (14.2%), Enterobacter (11.5%), Pseudomonas (9.8%), S.pyogenes (3.3%) and Proteus (1.1%). S. aureus was highly sensitive to Doxycycline (90.6%), Chloramphenicol (81.5%), Amikacin (79.5%) and Ceftraixone (72.7%). S. pyogenes showed 100% sensitivity to Cefexime, Amikacin, Chloramphenicol, Azithromycin and 80% sensitivity to Doxycycline. Similarly, most common gram negative isolate E.coli showed higher sensitivity to Chloramphenicol(71.4%) and Amikacin (66.7%) , Klebsiella showed higher sensitivity to Doxycycline(92.3%), Gentamicin(87.5%) and Amikacin (81.0%), Enterobacter showed higher sensitivity to Amikacin(90.9%) and Pseudomonas was highly sensitive to Chloramphenicol (71.4%) and Amikacin (66.7%). Piperacillin, Amikacin, Gentamicin, Ofloxacin and Ceftriaxone showed 100% sensitivity to Proteus spp. Amoxyclav, Cefepime and Cefexime (except in S. pyogenes) showed least sensitivity in both gram negative and gram positive bacterial isolates. **Conclusion:** In our study, the most common isolate wasS. aureus. Amikacin, as a single drug was found to be effective for empirical therapy of both gram negative and gram positive bacteria whereas Doxycycline and Amikacin was found effective in gram positive isolates. Amoxyclav and Cefepimewas commonly resistant in all bacterial isolates.


***Keywords: Antibiotic sensitivity, Pus culture, SSTI, SSI***


1. Mrs. Ankita Singh
2. Dr. Vikash Verma
3. Dr. Ritesh Singh

**Address for Correspondence:**

Mrs. Ankita Singh
Lecturer, Department of Pharmacology
Nepalgunj Medical College, Chisapani
Contact no: 9841108538
E-mail: ankita.hb@gmail.com


## INTRODUCTION

The invasion of pyogenic bacteria during or after trauma, burn injuries and surgical procedures can result in inflammation and pus formation[1,2]. Body's defense mechanism recruits immune cells into the infection site to fight against bacteria. Accumulation of these cells produces pus, causing pyogenic infection which actually delays the wound healing and may cause complication like wound dehiscence or wound breakdown[3].

Clinicians who care for patients with skin and soft tissue infection (SSTI) and surgical site infection (SSI) are responsible for determining the presence of an infection, identifying the extent of infection, ascertaining the causative microorganism, administering the appropriate antibiotics, and deciding on surgical treatments for purulent and necrotizing infections[3,4]. Rational use of antibiotics can hasten the patient recovery, prevents severe complications such as skin deformation, body defect, and death and prevents the abuse and misuse of broad-spectrum antibiotics and expression of antibiotic-resistant bacteria[5].

The bacterial spectrum can vary in different hospital and the bacterial sensitivity to a particular antibiotic is also different in different hospitals. Further, the bacterial sensitivity is not a static phenomenon but many sensitivity changes can occur in bacteria with time. Therefore, this study has been conducted at NGMCTH to see the spectrum of bacteria in pus culture and its sensitivity to a particular antibiotic. Further, if the antibiotic sensitivity of particular bacteria is known then we can empirically prescribe the patient till the culture report is thrown. Thereafter, the culture report dictates the choice of the antibiotics.

**MATERIALS AND METHOD**

This is a cross sectional hospital based study conducted in Department of Bacteriology, NGMCTH. This study was for a period of eleven months i.e. from January 2019 to November 2019. Pus samples were obtained from the Department of Surgery and Department of Gynaecology/Obstretics. Pus samples were processed for aerobic culture, identification of bacteria and antibiotic sensitivity test in the Microbiology Laboratory, NGMCTH.

The samples were inoculated on blood agar and mac conkey agar and incubated overnight at 37°C in aerobic condition. Identification and characterization of bacteria were performed on the basis of gram staining and microscopic/ colony characteristics.

Antibiotic susceptibilities of bacterial isolates were determined by Kirby- Bauer disc diffusion method using Clinical and Laboratory Standards Institute (CLSI) guidelines[6].

For fastidious organisms such as Streptococci, agar was supplemented with 5% sterile defribinated blood. Antibiotic discs containing Piperacillin (100mcg), Cefexime (5mcg), Amikacin (30mcg), Gentamicin (10mcg), Ofloxacin (5mcg), Chloramphenicol (30mcg), Doxycycline (30mcg), Ceftriaxone (30mcg), Azithromycin (15mcg) and Cefepime (30mcg) were used. The numerical data obtained from the study were entered and analyzed in SPSS software version 20.0 and results were expressed in charts and tables.

**RESULTS**

Out of 311 pus swabs received for culture and sensitivity test in the Microbiology laboratory, NGMCTH, 164 cases (52.73%) cases yielded positive culture, and the rest of 147 (47.27%) cases were growth negative. Among the 164 culture positive pus samples (100%), 150 (91.46%) yielded pure bacterial isolates, 9 (3.05%) yielded two organism and 5 yielded (1.22%) three organisms; so, a total number of 183(58.84%) bacterial strains were isolated from 311 (100%) pus samples.

| Order of isolates | Frequency (%) |
|---|---|
| Aerobic monomicrobial growth | 150 (48.23%) |
| Aerobic polymicrobial growth | 14 (2.25%) |
| No growth | 147 (47.27%) |
| **Total** | **311 (100%)** |

*Table I: Shows Bacterial Yield in Culture of Pus Samples*

**Distribution of bacterial agents**

Out of 164pus swabs that showed bacterial growth, 183 bacterial strains were isolated. A total number of 110 (60.1%) Gram Negative Bacilli (GNB) and 73 (39.9%) Gram Positive Cocci (GPC) was observed. Most common organism isolated was Staphylococcus aureus 66(36.1%) followed by E.coli 44(24%). Other isolates included were Klebsiellaspp 26(14.2%), Enterobacterspp 21(11.5%), Pseudomonas spp 18 (9.8%), Streptococcus pyogenes 6(3.3%) and Proteus spp 2(1.1%).

| Gram Staining | Frequency | Percentage |
|---|---|---|
| Gram Positive Cocci | 73 | 39.9% |
| Gram Negative Bacilli | 110 | 60.1% |

*Table II: Shows the Frequency/Percentage of Gram Stained Bacteria*

| Bacteria | Frequency | Percent |
|---|---|---|
| S. aureus | 66 | 36.1% |
| E.coli | 44 | 24.0% |
| Klebsiella | 26 | 14.2% |
| Enterobacter | 21 | 11.5% |
| Pseudomonas | 18 | 9.8% |
| S. pyogenes | 6 | 3.3% |
| Proteus | 2 | 1.1% |
| **TOTAL** | **183** | **100%** |

*Table III: Shows Rate of Isolated Bacteria after Aerobic Culture*

| Antibiotics | Staph. aureus | E.coli | Kleb siella | Entero bacter | Pseudo monas | S. pyogenes | Proteus |
|---|---|---|---|---|---|---|---|
| Piperacillin | 43 | 31 | 20 | 14 | 12 | 3 | 1 |
| Cefexime | 42 | 20 | 19 | 6 | 9 | 3 | 1 |
| Amikacin | 44 | 27 | 21 | 11 | 9 | 2 | 1 |
| Amoxyclav | 31 | 23 | 17 | 12 | 7 | 2 | 1 |
| Gentamicin | 39 | 28 | 16 | 11 | 7 | 6 | 1 |
| Ofloxacin | 33 | 17 | 17 | 5 | 4 | 3 | 1 |
| Chloramphenicol | 27 | 21 | 12 | 7 | 5 | 3 | 2 |
| Doxycycline | 32 | 24 | 13 | 17 | 8 | 5 | 1 |
| Ceftriaxone | 33 | 22 | 18 | 8 | 3 | 2 | 1 |
| Azithromycin | 16 | 13 | 3 | 8 | 6 | 3 | 1 |
| Cefepime | 43 | 25 | 14 | 9 | 6 | 5 | 2 |

*Table IV: Shows Frequency Distribution of Antibiotics Screened in Pus Culture Reports*

**Pattern of sensitivity for Gram Positive Cocci**

Most common isolated organism S. aureus showed highest sensitivity to Doxycycline (90.6%) which was followed by Chloramphenicol (81.5%), Amikacin (79.5%) and Ceftriaxone (72.7%). Amoxyclav (3.2%), Cefexime (11.9%) and Cefepime (14.0%) were the least sensitive antibiotics. Similarly, S.pyogenes was 100% sensitive to Cefexime, Amikacin, Chloramphenicol, Azithromycin and 80% sensitive to Doxycycline. Amoxyclav and Ofloxacin were not sensitive at all. Both of the Gram Positive Cocci were highly sensitive to Doxycycline, Amikacin and Chloramphenicol and least sensitive to Amoxyclav, Cefepime and Ofloxacin.

| Antibiotics | S.aureus (Number/Percent) | S.pyogenes (Number/Percent) |
|---|---|---|
| Piperacillin | 27 (62.8%) | 1 (33.3%) |
| Cefexime | 5 (11.9%) | 3 (100%) |
| Amikacin | 35 (79.5) | 2 (100%) |
| Amoxyclav | 1 (3.2%) | 0(0%) |
| Gentamicin | 21 (53.8%) | 2 (33.3%) |
| Ofloxacin | 14 (42.4%) | 0(0%) |
| Chloramphenicol | 22 (81.5%) | 3 (100%) |
| Doxycycline | 29 (90.6%) | 4 (80%) |
| Ceftriaxone | 24 (72.7%) | 1 (50%) |
| Azithromycin | 10 (62.5%) | 3 (100%) |
| Cefepime | 6 (14.0%) | 1 (20%) |

***Table V: Shows the Antibiotic Sensitivity Pattern ofGram Positive Cocci***

**Pattern of sensitivity for Gram Negative Bacilli**

Most common gram negative isolate E.coli showed higher sensitivity to Chloramphenicol (71.4%) and Amikacin (66.7%) and least sensitivity to Amoxyclav (0%), Cefexime (5%) and Cefepime (8%). Klebsiella showed higher sensitivity to Doxycycline (92.3%), Gentamicin (87.5%) and Amikacin (81.0%) while showed least sensitivity to Amoxyclav (0%), Cefexime (10.5%), Ceftriaxone(16.7%) and Cefepime (28.6%). Similarly, Enterobacter showed higher sensitivity to Amikacin (90.9%) and least sensitivity to Amoxyclav (0%), Cefepime (11.1%), Ceftriaxone (12.5%) and Chloramphenicol (14.3%). Pseudomonas was sensitive to Chloramphenicol (71.4%) and Amikacin (66.7%) and least sensitive to Amoxyclav (0%), Cefexime (5%) and Ofloxacin (5.9%). Piperacillin, Amikacin, Gentamicin, Ofloxacin and Ceftriaxone showed 100% sensitivity to Proteus spp. Amikacin was the most effective antibiotic for all the isolated Gram Negative Bacilli.

| Antibiotics | E.coli (Number / Percent) | Klebsiella (Number / Percent) | Enterobacter (Number / Percent) | Pseudomonas (Number / Percent) | Proteus (Number / Percent) |
|---|---|---|---|---|---|
| Piperacillin | 20(64.5%) | 8(40%) | 4(28.6%) | 20(64.5%) | 1(100%) |
| Cefexime | 1(5%) | 2(10.5%) | 2(33.3%) | 1(5%) | 0(0%) |
| Amikacin | 18(66.7%) | 17(81.0%) | 10(90.9%) | 18(66.7%) | 1(100%) |
| Amoxyclav | 0(0%) | 0(0%) | 0(0%) | 0(0%) | 0(0%) |
| Gentamicin | 10(35.7%) | 14(87.5%) | 6(54.5%) | 10(35.7%) | 1(100%) |
| Ofloxacin | 1(5.9%) | 8(47.1%) | 1(20%) | 1(5.9%) | 1(100%) |
| Chloram phenicol | 15(71.4%) | 6(50%) | 1(14.3%) | 15(71.4%) | 1(50%) |
| Doxycycline | 7(29.2%) | 12(92.3%) | 6(35.3%) | 7(29.2%) | 0(0%) |
| Ceftriaxone | 6(27.3%) | 3(16.7%) | 1(12.5%) | 6(27.3%) | 1(100%) |
| Azithromycin | 8(61.5%) | 1(33.3%) | 3(37.5%) | 8(61.5%) | 0(0%) |
| Cefepime | 2(8%) | 4(28.6%) | 1(11.1%) | 2(8.0%) | 1(50%) |

***Tale VI: Shows Antibiotic Sensitivity Pattern of Gram Negative Bacilli***

**DISCUSSION**

Irrational use of antibiotics has been held responsible for the emerging resistant bacterial strains. Rational use of antibiotic would include the selection of right antibiotic, to right patient, at right dose, for right duration of time. However, empirical therapy of antibiotic has to be followed in infection if it is life threatening to avoid the complications and severity of pyogenic infections. The problem is that we do not know the bacteria responsible and its sensitivity to particular antibiotics. Appropriate empirical therapy of antimicrobials should beinitiated on the basis of knowledge of the common pathogens of the area, producing the particular disease and their antibiotic sensitivity. This is only possible by such a study conducted here.

In our present study, predominance of monomicrobial (48.23%) was observed while polymicrobial infections (2.25%) were uncommon. This result is in accordance with the study done by Hanuman thappa et al[7] (2016) where mono microbial infection was 54.2% and polymicrobial infection was 2.8%. However, Basuet al[8] (2009) reported86.54% of monomicrobial infections and 9.61% of polymicrobial infection and Sowmya et al[5] (2014) reported 91.7% of monomicrobial growth.

Our study showed 60.1% of Gram Negative Bacilli (GNB) and 39.9% of Gram Positive Cocci (GPC). A study by Sowmyaet al[5] (2014) also reported 60% of GNB and 40% of GPC. Zubairet al[9] (2011) yielded predominance of GNB (63.8%) in aerobic pus culture and Mantravadi et al[10] reported 59.3% of GNB growth which is consistent with our findings.

The commonest gram positive organism producing pus in our hospital patient was S. aureus (36.1%) followed by S. pyogenes (3.3%) in small number of cases. Our findings are in accordance with Pandeya et al11 that reported 32.3% of S. aureus and 2.40% of S.pyogenes. Sowmyaet al[5] (2014) reported 37.4%

of S. aureus and Mantravadi et al10 reported 37.2% of S. aureus and 2.2% of Streptococcus spp which was similar to our findings.

Our study showed that S. aureus was most sensitive to Doxycycline (90.6%) followed by Chloramphenicol (81.5%), Amikacin (79.9%) and Ceftriaxone(72.7%). Similarly S.pyogenes showed 100% sensitivity to Amikacin, Cefepime, Chloramphenicol, Azithromycin and 80% sensitivity to Doxycycline. Hanuman thappa et al[7] (2016) showed 76.1% sensitivity of S. aureus to Amikacinand Mantravadi et al[10] (2015) showed 71.4% sensitivity of S. aureus to Amikacin and 100% sensitivity of Streptococcus to Amikacin which are in accordance with our study results. Similarly, Chaudary et al[12] (2017) reported in their study that the most sensitive antibiotic for gram negative isolates was Amikacin (93%) followed by Chloramphenicol (92.6%). However, Naimi et al[13] (2017) showed that S. aureus had 77% sensitivity to Doxycycline and 84.8% sensitivity to Chloramphenicol which was different from our study with 90.6% sensitivity to Doxycycline and 79.5% sensitivity to Amikacin. Similarly, S. aureus was least sensitive to Amoxyclav (3.2%) and S. pyogenes showed no sensitivity to Amoxyclav. This was comparable to Trojan et al[14] that reported 89% of resistance to Amoxyclav and Mantravadi et al[11] reported 77.4% of Amoxyclav resistance in S. aureus but only 10% Amoxyclav resistance in S.pyogenes which was different from our findings.

Emperically for gram positive infections, the treatment in the life threatening condition should be started with Doxycycline as Chloramphenicol is not favored by Clinicians because of bone marrow suppressing action. In pediatric patients, Amikacin may be a better choice.

The commonest gram negative offender in our study is E.coli (22.6%) followed byKlebsiella (14.5%), Enterobacter (11.5%), Pseudomonas (9.8%) and Proteus (1.1%). In their study Basuet al[7] report that Pseudomonas (40.38%) and E. coli (15.38%) to be the most common isolates in wound infection which is inconsistent with our finding. Our finding is in accordance with the Pandeya et al[11] that reports 20.7% of E.coli and the least isolated bacteria was Proteusmirabilis (0.6%). Similarly, Mantravadiet al[10] also supports our findings withmost common gram negative organism being E.coli (21.7%) followed by Klebsiella (16.8%) and Pseudomonas (7.5%) among other isolates.

Antibiotic sensitivity of these microorganism showed that all of them are commonly sensitive to Amikacin. i.e. E.coli showed higher sensitivity to Chloramphenicol (71.4%) and Amikacin (66.7%), Klebsiella showed higher sensitivity to Doxycycline (92.3%), Gentamicin (87.5%) and Amikacin (81.0%), Enterobacter showed higher sensitivity to Amikacin (90.9%), Pseudomonas showed higher sensitivity to Chloramphenicol (71.4%) and Amikacin (66.7%) while Proteus spp.

showed 100% sensitivity to Piperacillin, Amikacin, Gentamicin, Ofloxacin and Ceftriaxone. A study by Chaudary R et al[12] (2017) reports Amikacin (93%) to be the drug of choice for gram negative bacterial isolates which was comparable with our study results with 81.06% sensitivity of Amikacin to gram negative isolates. Similarly, Timilsina et al[15] (2015) shows the sensitivity of Amikacin to be 93.62% followed by Gentamycin 89% for gram negative isolates. For E. coli, Timilsina et al15 found out that the most effective antibiotic was Amikacin (100%). Shrestha et al[16] (2009) also showed Amikacin (94.38%) to be the most sensitive antibiotic for E. coli. Abdullah et al[17] (2013) showed low sensitivity of Doxycycline (11.5%) and high sensitivity to Amikacin (89.4%) in Klebsiella isolates. However, a study by Khare et al[18] (2017) reported higher sensitivity of Klebsiella to Doxycycline (67%) and another study by Ravichitra et al[19] (2014) showed 87.3% of sensitivity of Gentamycin to Klebsiella which was similar to our findings. Pandeya et al[11] also reports higher sensitivity (80%) of Pseudomonas to Amikacin and Chloramphenicol and E.coli higher sensitivity to Amikacin (70.6%). Khanam et al[20] in their study concluded that Amikacin among the aminoglycosides showed good sensitivity whereas resistance to Gentamicin and Tobramycin is on the rise. Similarly, our study reports that all gram negative isolates were 0% sensitive to Amoxyclav and very less sensitive to Cephalosporins; Cefexime and Cefepime. E.coliwas 88.2% resistant to Cefepime in the study by Pandeya et al[10] which was similar to our findings with only 8% sensitivity of Cefepime to E.coli.Timilsina et al[15] also supports our finding with 88.46% resistance of Amoxicillin and 73% resistance of Cefixime. Stanley et al[21] (2018) also reports very high resistance in Amoxicillin-Clavulanate in E. coli and Klebsiella in his study in Uganda.

From above discussion, it is clear that Amikacin, as an empirical drug, is the drug of choice for life threatening infections which covers both gram negative & gram positive organism. Once a culture report is thrown, a proper antibiotic according to the report should be prescribed. The emerging resistance pattern of the established antibiotics threatens the treatment strategy for pyogenic infections. With the limited number of antimicrobial agents currently available and many other antibiotics that are still under the clinical trials, it is utterly important to rationally select the antibiotics.

## CONCLUSION

Study concludes that the most common isolated bacteria after aerobic culture of pus is the Staphylococcus aureus. The most effective antibiotic for gram positive isolates was Doxycycline and Amikacin for gram negative isolates. Amikacin, as the single drug was found to be most effective for both gram negative and gram positive infections.

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