

Original Article**Pattern of Aerobic Bacterial Organisms Isolated From Pus and Their Antibiogram in Jahurul Islam Medical College & Hospital**

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Abstract

Aim: To analyze the pattern of aerobic bacterial pathogens and their antibiogram isolated from pus specimen in Jahurul Islam Medical College and Hospital.

Methodology: This retrospective study was conducted in the Department of Microbiology, Jahurul Islam Medical College and Hospital. The data included were the results of positive pus culture and the sensitivity test. The study period extended from 1st January, 2020 to 15th February, 2020. The results were retrieved from the records of Microbiology department of Jahurul Islam Medical College and Hospital. The samples were inoculated into blood agar and MacConkey's agar media and incubated aerobically at 37^oc for 24 hours. Isolation and identification of different bacteria were done by standard microbiological procedure, including colony morphology, Gram staining and biochemical tests. Antimicrobial susceptibility testing was carried out by 'Kirby-Bauer disc diffusion technique' using Mueller- Hinton agar media and zones of inhibition were measured and interpreted as recommended by the Clinical and Laboratory Standard Institute (CLSI) guidelines.

Result: In the present study, a total of 100 culture positive pus specimen results were analyzed. *Staphylococcus aureus* was the most frequent organism isolated from pus specimen (N=45,45%) followed by *Escherichia coli* (N=33, 33%) and *Pseudomonas aeruginosa* (N=13,13%). Other organisms like *Proteus mirabilis* (N=07,07%) and *Streptococcus sp* (N=02, 02%) were also isolated. Both *Staphylococcus aureus* and *Streptococcus sp* showed high (100%) resistance to Amoxyclav. *Streptococcus sp* alone was highly (100%) resistant to Azithromycin, Cotrimoxazole and Clindamycin. *Staphylococcus aureus* was 84.4% resistant to Azithromycin and 15.6% sensitive to Azithromycin. *Staphylococcus aureus* and *Streptococcus sp* showed almost equal sensitivity (53.3% and 50.0% respectively) to Cefuroxime. Similar to Gram positive cocci, Gram negative bacilli *Escherichia coli*, *Pseudomonas aeruginosa* and *Proteus mirabilis* showed high (100%) resistance to Amoxyclav. Colistin was 93.9% sensitive against *Escherichia coli* and 100% sensitive against *Pseudomonas aeruginosa* but it was only 14.3% sensitive against *Proteus mirabilis*. *Escherichia coli* and *Proteus mirabilis* showed fairly good sensitivity to Meropenem (78.8% and 85.7% respectively) whereas *Pseudomonas aeruginosa* was only 30.8% sensitive to it. All bacteria isolated from pus sample were mostly sensitive to Piperacillin-Tazobactam.

Key words: Pus culture, Isolation pattern, Antibiogram.

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Introduction

Infectious diseases remain the most common cause of morbidity and mortality worldwide¹. Bacteria and viruses cause most diseases, but diseases are also caused by other microorganism, protozoa and other parasites². Pyogenic infections are characterized by local and systemic inflammation usually with pus formation. These may be endogenous or exogenous. A break in the skin can provide entry to the surface bacteria which thereby starts multiplying locally. The body's defense mechanism includes bringing of immune cells into the area to fight against bacteria. Eventually, accumulation of these cells produces pus which is a thick whitish liquid^{3,4}. The most common pus producing bacteria are *Staphylococcus aureus* (*S. aureus*), *Klebsiella sp*, *Pseudomonas*, *Escherichia coli* and *Streptococci* in which *S. aureus* is the most common bacteria that produces pus⁵. These infections result in delayed healing and may cause several complications like wound dehiscence or wound breakdown^{6,7}. The use of antimicrobial agents locally or systemically for prevention or treatment of infections in any dose and over any time period causes a "selective pressure" on microbial population⁸. The routine use of antibiotics has resulted in widespread antibiotic resistance by development of antibiotic resistant genes in many organisms⁹. The current spread of multi drug resistant bacteria from clinical isolates has increased the need for regular updates in the knowledge of the bacteriological review of pus culture reports so as to avoid the unguided empirical treatment which appears to differ in various environment¹⁰. Penicillin, the first antibiotic to be used on a large scale, was first put to use during the World War II¹¹. It was considered the magic bullet as just a single injection could cure a life-threatening infection¹². Since its discovery and consequently, with the advent of more antibiotics, there was a belief in the medical fraternity that this would lead to the eventual eradication of infectious diseases. On account of erratic use, malpractices or for natural causes, in recent years, drug resistance of many human pathogenic bacteria is being commonly reported from all over the world¹³. Although pharmacological

industries have produced large number of newer antibiotics in the last three decades, the situation is alarming in developing as well as developed countries mainly because of their indiscriminate use^{14,15}. The present study was undertaken to analyze the pattern of aerobic bacterial pathogens involved and their antibiotic sensitivity isolated from pus specimen in Jahurul Islam Medical College and Hospital.

Materials and Methods

This retrospective study was conducted in the Department of Microbiology, Jahurul Islam Medical College and hospital. The data included the positive pus culture results that were analyzed for the period from 1st January 2020 to 15th February 2020. Records were retrieved from the Microbiology department of Jahurul Islam Medical College. All the significant isolates were identified and studied by standard procedure. The samples were inoculated into blood agar and MacConkey's agar media and incubated aerobically at 37°C for 24 hours. Isolation and identification of different bacteria were done by standard microbiological procedures, including colony morphology, Gram staining and biochemical tests¹⁶.

Antimicrobial susceptibility testing was carried out by 'Kirby-Bauer disc diffusion technique' using Mueller Hinton agar media and zones of inhibition were measured and interpreted as recommended by the Clinical and Laboratory Standard Institute (CLSI) guidelines¹⁷.

Results

In the present study, a total of 100 positive culture results of pus specimen were analyzed. *Staphylococcus aureus* was the most frequently isolated organism from pus specimen (N=45,45%) followed by *Escherichia coli* (N=33,33%) and *pseudomonas aeruginosa* (N=13,13%). Other organisms like *Proteus mirabilis* (N=07, 07%) and *Streptococcus sp* (N=02,02%) were also present. The type and frequency of isolation of different organisms are shown in Table 1.

Table 1: The isolation pattern of organisms and their percentage.

Organism isolated (N=100)	Number	Percentage
<i>Staphylococcus aureus</i>	45	45%
<i>Streptococcus sp</i>	02	02%
<i>Escherichia coli</i>	33	33%
<i>Pseudomonas aeruginosa</i>	13	13%
<i>Proteus mirabilis</i>	07	07%

Staphylococcus aureus and *Streptococcus sp* were completely (100%) resistant to Amoxyclav. *Streptococcus sp* showed high(100%) resistance to Azithromycin, Cotrimoxazole and Clindamycin. *Staphylococcus aureus* was 84.4% resistant and 15.6% sensitive to Azithromycin. *Streptococcus sp* showed 100% sensitivity to Piperacillin-Tazobactam but

Staphylococcus aureus was only 52.2% sensitive to it. Both *Staphylococcus aureus* and streptococcus sp showed almost similar type of sensitivity to Cefuroxime. The comparative antibiotic sensitivity and resistance pattern of *Staphylococcus aureus* and *Streptococcus sp* is shown in Table 2.

Table 2: The antibiotic sensitivity pattern of Gram-positive cocci.

Antibiotics	Staphylococcus aureus (N=45)		Streptococcus sp (N=02)	
	Sensitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)
Amoxyclav	0	100	0	100
Azithromycin	15.6	84.4	0	100
Cotrimoxazole	42.2	57.8	0	100
Clindamycin	33.3	66.7	0	100
Cefuroxime	53.3	46.7	50	50
Cloxacillin	66.7	33.3	50	50
Ceftriaxone	51.1	48.9	50	50
Gentamycin	46.7	53.3	100	0
Linezolid	46.7	53.3	50	50
Levofloxacin	46.7	43.3	50	50
Piperacillin -Tazobactam	52.2	37.8	100	0

Similar to Gram-positive cocci, Gram-negative bacilli -*Escherichia coli*, *Pseudomonas aeruginosa* and *Proteus mirabilis* showed highest (100%) resistance to Amoxyclav. Azithromycin was 100% resistant against *Proteus mirabilis*, 72.7% resistant for *Escherichia coli*, and 61.5% resistant against *Proteus mirabilis*. *Escherichia coli* showed high (27.3%) degree of resistance to Cefixime. *Pseudomonas aeruginosa* exhibited 69.2% resistance to Cefixime whereas *Proteus mirabilis* was 42.9% sensitive to Cefixime. Colistin showed high level (93.9%) of sensitivity

against *Escherichia coli* and 100% sensitivity for *Pseudomonas aeruginosa* but it was only 14.3% sensitive against *Proteus mirabilis*. Meropenem showed good sensitivity against *Escherichia coli* and *Proteus mirabilis* (78.8% and 85.7% respectively) but it showed only 30.8% sensitivity for *Pseudomonas aeruginosa*. All bacteria isolated from pus sample were highly sensitive to Piperacillin-Tazobactam. The antibiotic sensitivity and resistance pattern of Gram negative bacteria is shown in Table 3.

Table 3: Antibiotic sensitivity pattern of Gram negative bacteria.

Antibiotics	<i>Esch coli</i> (N=33)		<i>Ps. aeruginosa</i> (N=13)		<i>Pr. mirabilis</i> (N=07)	
	Sensitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)	Sensitive (%)	Resistant (%)
Amoxyclav	0	100	0	100	0	100
Amikacin	63.6	36.4	76.9	23.1	71.4	28.6
Azithromycin	27.3	72.7	38.5	61.5	0	100
Cotrimoxazole	51.5	48.5	30.8	69.2	28.6	71.4
Cefuroxime	42.4	57.6	15.4	84.6	42.9	57.1
Cefixime	27.3	72.7	30.8	69.2	57.1	42.9
Ceftriaxone	39.4	60.6	23.1	76.9	71.4	28.6
Colistin	93.9	6.1	100	0	14.3	85.7
Gentamycin	90.9	9.1	46.2	53.8	71.4	28.6
Levofloxacin	63.6	36.4	38.5	61.5	42.9	57.1
Meropenem	78.8	21.2	30.8	69.2	85.7	19.3
Piperacillin Tazobactam	66.7	33.3	69.2	30.8	100	0

Discussion

Pyogenic infection is referred to bacterial infection that leads to severe local inflammation with pus. The invasion and multiplication pathogens in tissue will cause cell damage and leads to loss of integrity of tissue and skin. This will leads to subcutaneous infection to life threatening complications. The present study is aimed to isolate the bacterial pathogens which cause pyogenic infection and study their antibiotic resistance pattern. In this study, both gram positive and gram negative pathogens were isolated from a total of 100 samples. In a study from Bangladesh, *S. aureus* is the commonest organism isolated in 40.5% of the cases of wound infections¹⁸. It is the commonest organism in most of the studies^{19,20,21}. In a study by Bowler et al. *S.aureus* is the most common cause of cutaneous abscess in 25-30% of the cases and mostly associated with acute soft tissue infections. It is also associated with bite wound infections and diabetic wound ulcers. It is one of the causes of delayed wound infection²². In this study *S. aureus* was 45 in number out of 100 samples. The antibiotic resistance pathogens were rapidly increased due to the frequent use of antibiotics. Now a day it became great difficulty to manage or control the pyogenic pathogen and one of the major problems faced by the physicians²³. In this study *Staphylococcus aureus* showed 100% resistance to Amoxycylav, 84.4% resistance to Azithromycin, 57.8% resistance to Cotrimoxazole and 66.7% resistance to Clindamycin. *Staphylococcus aureus* showed 66.7% sensitivity to Cloxacillin, 52.2% sensitivity to Piperacillin-Tazobactam and 51.1% sensitivity to Ceftriaxone. Another Gram positive cocci that is isolated from pus sample was *Streptococcus sp.* *Streptococcus sp* was found to be 100% resistant to Amoxycylav, Azithromycin, Cotrimoxazole and Clindamycin. This organism showed 100% sensitivity to Piperacillin-Tazobactam and Gentamycin and intermediate towards Levofloxacin(50%), Cloxacillin (50%), Linezolid (50%) and Cefuroxime (50%). Among Gram-negative bacilli, *Esch.coli* was the predominant organism isolated from pus samples as is seen in several studies²⁴. It is the most common Gram negative bacilli isolated in post-operative infections²⁵. In this study among the Gram negative bacilli *Escherichia coli* (N=33) was predominant organism isolated from pus sample. Antibiotic resistance is of increasing concern as it is transmitted among patients leading to further difficulty in treatment. Most of our Enterobacteriaceae

were from surgical site infections like intra-abdominal surgeries, abscesses like perianal abscess, intra-abdominal abscess, perinephric abscess and pyonephrosis, but as this was a retrospective study, the source could not be determined²⁶. The Present study, the second most commonly isolated Gram negative bacilli was *Pseudomonas aeruginosa* (N=13) followed by *Proteus mirabilis* (N=07). Widespread and non-judicious use of these antibiotics without sensitivity testing and abuse of these drugs by self-medication to treat all kind of infections due to low cost, may have promoted development of resistance to these antibiotics²⁷. *Escherichia coli*, *Pseudomonas aeruginosa*, *Proteus mirabilis* showed 100% resistance against Amoxycylav. *Escherichia coli* was 72.7% resistant, *Pseudomonas aeruginosa* 61.5% and 100% resistant to Aithromycin. Another drugs third generation Cephalosporin, Cefixime that has been used for long period of time in our country showed 72.7% resistance against *Escherichia coli* and 69.2% against *Pseudomonas aeruginosai* was 57.1% sensitive and 42.9% resistant to Cefixime. Study Shows, Colistin, Meropenem, Piperacillin-Tazobactam and Gentamycin showed good sensitivity against most of the pathogens isolated from pus sample. This may be because of the fact that these are less commonly prescribed drugs as they have only parenteral preparation and they are used in hospitalized patients according to the sensitivity report. Sensitivity pattern of pathogens is changing radically. This prominent and significant increase in resistance of organisms to commonly used antibiotics is alarming. Clinicians should look for the recent trend of susceptibility pattern, particularly of that locality while choosing a treatment regimen to prevent emergence of antibiotic resistance²⁷.

Conclusion

Antimicrobial sensitivity and resistance pattern of pathogens varies from time to time and from place to place. For this reason, regular monitoring of bacterial susceptibility to different antibiotics is necessary. In this study bacterial isolates from pus samples exhibited high to moderate levels of resistance against different classes of antibiotics. Regular monitoring of bacterial susceptibility and using antibiotics according to sensitivity whenever it is rational may reduce the emergence of antimicrobial drug resistance of microorganisms.

References

1. Hossein AH, Ali AR, Akram H, Farhad M. Infectious Diseases in hospitalized Children of Central Iran. *Pak J Med Sci.* 2010;26(4):901-4.
2. Ananthanarayan and Paniker. Infection. In test book of microbiology, 7th edition, CKJ paniker. Orient Longman. 2006;64-6.
3. Koneman WK, Allen SD, Janda WM, Schreckenberger PC, Propcop GW, Woods GL et al. Philadelphia Color Atlas and Textbook of Diagnostic Microbiology, 6th ed. Lippincott-Raven 2005; 624-62.
4. Chopra A, Puri R, Mittal RR, Kanta S. A clinical and bacteriological study of pyodermas. *Indian J Dermatol Venereol Leprol.* 1994;60:200-2.
5. Kumar AR. Antimicrobial sensitivity pattern of *Klebsiella pneumoniae* isolated from pus from tertiary care hospital and issues related to the rational selection of antimicrobials. *Journal of chemical and pharmaceutical research.* 2013; 5 (11): 326-331.
6. Garner JS, Jarvis WR, Emori TG, Horan TC, Hughes JM. CDC definitions for nosocomial infections. *Am J Infect Control.* 1988;16(3):128.
7. Terry BA. Cost-effective application of the Centres for Disease Control: guideline for prevention of surgical wound infections. *AM J infect Control.* 1985;13(3):232.
8. Sharafati-chaleshtori R, Sharafati-chaleshtori F, Karimi A. Antibiotic resistance pattern of staphylococcus strains isolated from orange and apple juices in Shahre-kord. Iran. *Pak J Med Sci.* 2010;26:615-8.
9. Sengupta S, Human P, Girag AM, Shivananda PG. Acinetobacterbac: An emergency nosocomial pathogen in the burns unit, Manipal. *India Burns.* 2001;27:140-4.
10. Mohammed A, Adeshina GO, Ibrahim YKE. Retrospective Incidence of wound infections and antibiotic sensitivity 162 pattern: A study conducted at the Aminu Kano Teaching hospital. Kano, Nigeria. *Int J Med Sci.* 2013;5:60-6.
11. Cohen ML. Epidemiology of drug resistance: implications for a post-antimicrobial era. *Science.* 1992; 257:1050-5.
12. Mumtaz S, Akhtar N, Hayat A. Antibigram of aerobic pyogenic isolates from wounds and abscesses of patients at Rawalpindi. *Pak J Med Res.* 2002; 41(1):16-18.
13. Jahan F, Lawrence R, Kumar V, Junaid M. Evaluation of antimicrobial activity of plant extracts on antibiotic susceptible and resistant *Staphylococcus aureus* strains. *J Chem Pharma Res.* 2011;3:777-89.
14. Ahmad I, Beg AZ. Antimicrobial and phytochemical studies on 45 Indian medicinal plants against multi-drug resistant human pathogens. *J Ethno pharma col.* 2001;74:113-23.
15. Harbottle H, Thakur S, Zhao S, White DG. Genetics of antimicrobial resistance. *Anim Biotechnol.* 2006;17:111-24.
16. Collee JG, Marr W. Specimen collection, culture containers and media. In: Collee JG, Duguid JP, Fraser AG, Marmion BP eds. *Mackie & McCartney Practical Medical Microbiology*, Vol. 2, 14th ed. Churchill Livingstone, New York. 1996: 85-111.
17. Wayne P. CLSI performance standard of antimicrobial susceptibility testing: twenty-fourth international supplement. CLSI Document M100S24. *Clin Lab Std Instit.* 2014;34.
18. Sultana S, Mawla N, Kawser S, Akhtar N, Ali MK (2015). Current microbial isolates from wound swab and their susceptibility pattern in a private medical college hospital in Dhaka city. *Delta Med Col J.* 3: 25-30.
19. Tiwari P, Kaur S (2010) Profile and sensitivity pattern of bacteria isolated from various cultures in a tertiary care hospital in Delhi. *Indian J Public Health.* 54: 213-215.
20. Duggal S, Khatri PK, Parihar RS, Rajat A. Antibigram of various bacterial isolates from pus samples in a tertiary care centre in Rajasthan. *Int J Sci Res.* 2015; 4: 1580-1584.

21. Rennie RP, Jones RN, Mutnick AH. SENTRY Program Study Group (North America) . Occurrence and antimicrobial susceptibility patterns of pathogens isolated from skin and soft tissue infections: report from the SENTRY Antimicrobial. 2003.
22. Bowler PG, Duerden BI and Armstrong DG. Wound microbiology and associated approaches to wound management. Clin Microbiol Rev.2001; 14: 244-269.
23. Sujata Singh, MayuriKhare, Rakesh Kumar Patidar, Swati Bagde, Sahare KN, Deepak Dwivedi and Vinod Singh. Antibacterial activities against pyogenic pathogens. International Journal of Pharmaceutical Sciences and Research. 2013; 4(8): 2974-2979.
24. Biradar A, Farooqui F, Prakash R, Khaqri SY, Itagi I. Aerobic bacteriological profile with antibiogram of pus isolates. Indian J Microbiol Res. 2016; 3: 245-249.
25. Ananthi B, Ramakumar M, Kalpanadevi V, Sopia A, Karthiga L, Kalavathy VH. Aerobic bacteriological profile and antimicrobial susceptibility pattern in postoperative wound infections at a tertiary care hospital. Int J Med Sci Clin Inv. 2017; 4: 2702-2706.
26. Sukanya S, Kanne P, Chavali P, Vemu L. Aerobic bacteriological profile and antimicrobial susceptibility pattern of pus isolates from tertiary care hospital in India. J Infect Dev Ctries. 2018; 12(10):842-848.
27. Afroz S, Sarkar D, Khatun K, Khan TM, Paul S. Bacterial pathogens in wound infection and their antimicrobial susceptibility pattern in a medical college hospital, in Dhaka, Bangladesh. Int J Res Med Sci. 2020;8(6):2105-2109.