

Original Research Article

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## Bacteriological Profile and Antibiotic Sensitivity Pattern of Bacterial Pathogens Isolated from Pus Samples

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### ABSTRACT

Bacterial infections are considered as the common causes for the pyogenic infections. Various studies have shown that there is an inconsistency in bacteriological profile of the isolates and antibiotic resistance pattern. This demands for the periodic monitoring of the bacterial isolates and their antimicrobial sensitivity pattern. The present study aims to analyze the bacterial pathogens isolated from the pus samples and their antibiotic susceptibility pattern. This is a prospective, observational, single-centered study, conducted in a tertiary care hospital in Hyderabad for a period of 2 years, from August 2016 to 2018. A total of 410 pus samples were brought to the Microbiology lab from different inpatient and outpatient departments and were processed to identify the pathogens and their anti-microbial sensitivity pattern using Kirby Bauer disc diffusion method. The data analyzed was presented in percentages. Out of 410 samples analyzed, 257 (62.68%) were found to be culture positive. Among 257 culture positive isolates, 56.78% were gram negative bacteria and the rest 43.16% were gram positive bacteria. However, the most commonly isolated pathogens were *Staphylococcus aureus* followed by *Klebsiella* and *E. coli*. The gram positive bacteria were found to be most susceptible to vancomycin and linezolid, whereas gram negative isolates were most susceptible to imipenem, meropenem and colistin. The most common pathogen identified was *Staphylococcus aureus*. However, gram negative bacteria were predominant. The emerging bacterial resistance for various antimicrobial agents complicates the management of pyogenic infection. Periodic susceptibility data from various studies helps clinicians to select proper antibiotic for the isolates, thus preventing the emergence of the drug resistance stains.

### Keywords

Antimicrobial susceptibility, Bacteriological profile, Pyogenic infection, Pus Culture, Antibiogram

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### Introduction

Skin acts as an effective deterrent for infections. Although, various bacteria inhabit our skin, nasopharynx, stomach and other parts of the body, they are unlikely to cause

infections due to the presence of first line defense mechanism of the body.<sup>(1)</sup> This defense mechanism can be altered by various factors such as surgical procedures, trauma, burns and diseases leading to the skin penetration by the pathogens.<sup>(2)</sup>

Skin infections are not uncommon and the severity ranges from less significant pyoderma infections to very rare and life-threatening necrotizing infections. Various microorganisms (such as bacteria, fungi and parasites) can cause skin infections.<sup>(3)</sup>

Pus formation commonly encountered in bacterial infections, is usually due to the systemic inflammation. Bacterial pathogens leading to purulence or pus formation are called as pyogenic bacteria and such infections are called as pyogenic infections.<sup>(4,5)</sup>The most prevalent gram positive pyogenic bacteria include – *Staphylococcus aureus*, *Streptococcus pyogenes* and *Enterococci* and gram negative pyogenic bacteria include – *Escherichia coli*, *Klebsiella* species, *Proteus* and *Pseudomonas* species.<sup>(6)</sup>

Antibiotic therapy plays a cardinal role in the treatment of such infections. Many studies have shown that though the bacteriological profile from pus samples is consistent, but there are variations in the antibiotic resistance profile obtained from pyogenic pathogens.<sup>(7)</sup>Mis-prescriptions and improper use of antibiotics has led to the continuous increase in the emergence of the multi drug resistant microorganisms. As a result, there is increase in the rate of prolonged illness and risk of spreading the resistant strains.<sup>(8,9)</sup>

The aim of this study is to identify the pyogenic bacteriological profile and antibiotic sensitivity of the isolates from pus samples at a tertiary care hospital.

## **Materials and Methods**

This is a prospective, single centered study conducted for a period of 2 years from 2016 to 2018 in the Microbiology Department of Princess Esra Hospital, Hyderabad. The ethical committee approval was obtained

before the initiation of the study. A total of 410 samples were collected by sterile syringe aspiration and swabs from various inpatient and outpatient wards of the hospital, and were analyzed for bacterial growth isolates and their sensitivity profile for antibiotics in Microbiology laboratory as per the standard microbiological protocols.

The pus samples were inoculated on blood agar and Mac Conkey's agar plates at 35-37°C for about 24-48 hours. The primary identification of the bacterial colonies from the positive cultures was done with the help of colony appearance and hemolysis. Furthermore, identification and characterization of the bacterial isolates is performed by using colony morphological characteristics, hemolysis, gram staining and biochemical tests such as catalase test, coagulase test, indole test, methyl red (MR) test, citrate utilization test, urease hydrolysis test and triple sugar iron agar.

Antibiotic susceptibility pattern of the isolates was determined by using Kirby-Bauer disc diffusion test recommended by Clinical and Laboratory Standards Institute (CLSI). An inoculum was prepared for each bacterial isolate by adjusting the turbidity to 0.5 McFarland standard, which was then spread on Muller-Hinton agar plates. The antibiotic discs were then set on the agar plates and incubated overnight at 35-37°C for 24-48 hours. The zones of inhibition for the antibiotics were measured in mm and were further classified according to CLSI tables and guidelines.

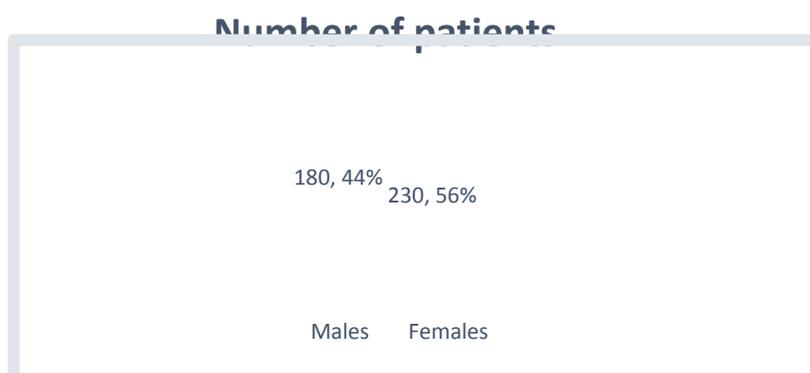
## **Results and Discussion**

The results were analyzed statistically using MS excel and was presented in terms of percentages.

**Table.1** Age distribution

Age group (years)	Number of patients	Percentage of patients
0-10	15	3.65
11-20	61	14.87
21-30	85	20.73
31-40	64	15.60
41-50	76	18.53
51-60	62	15.12
61-70	27	6.58
71-80	15	3.65
81-90	5	1.21

**Fig.1** Gender distribution



**Table.2** Department wise distribution of pus samples

S.No.	Department	No. of isolates	Percentage of isolates
1.	General Surgery	113	27.5
2	Dermatology	5	1.21
3	Pediatrics	15	3.65
4	Casualty	44	10.73
5	Plastic surgery	38	9.26
6	General Medicine	34	8.29
7	Gynaecology	25	6.09
8	Gastroenterology	20	4.87
9	Orthopedics	27	6.58
10	Urology	20	4.87
11	Cardiology	12	2.92
12	Neurology	15	3.65
13	CCU	9	2.19
14	MICU	13	3.17
15	Respiratory	12	2.92
16	ENT	8	1.95

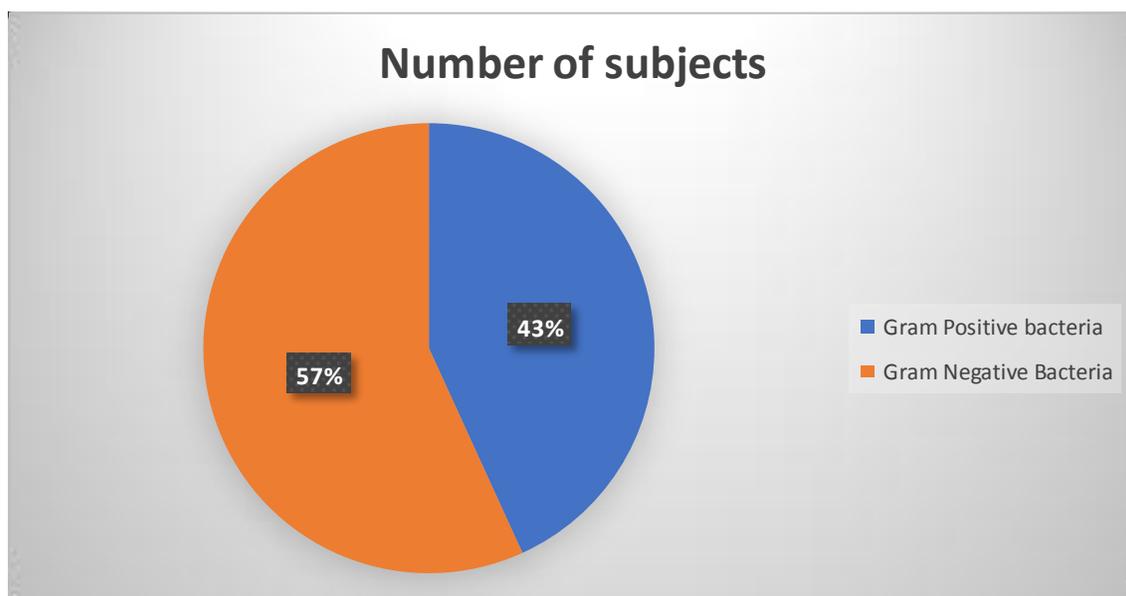
**Table.3** Distribution of samples according to the growth of isolates

S. No.	Sample Distribution	Number of Samples	Percentage of samples
1.	Culture Negative	153	37.31
2.	Culture Positive	257	62.68
3.	Total	410	100

**Table.4** Distribution of gram positive and gram-negative bacteria

S.No.	Isolated Bacteria	Number of subjects	Percentage of subjects (%)
<b>I.</b>	Gram Negative Bacteria	146	56.78
	Klebsiella species	96	37.35
	Escherichia coli	28	10.89
	Pseudomonas species	11	4.28
	Proteus species	5	1.94
	Acinetobacter species	2	0.77
	Citrobacter species	4	1.55
<b>II.</b>	Gram Positive Bacteria	111	43.16
<b>1</b>	Staphylococcus species		
	Methicillin resistant <i>Staphylococcus aureus</i> (MRSA)	14	5.44
	Methicillin sensitive <i>Staphylococcus aureus</i> (MSSA)	83	32.29
	Methicillin resistant coagulase negative <i>Staphylococcus</i> (MRCONS)	4	1.55
	Methicillin sensitive coagulase negative <i>Staphylococcus</i> (MSCONS)	9	3.50
<b>2.</b>	Streptococcus species	1	0.38
	Total	257	100

**Fig.2** Percentage of gram-positive and gram-negative isolates



**Table.5** Antibiotic sensitivity of gram negative isolates

S.No	Antibiotic	<i>Klebsiella</i> n=96	<i>E. coli</i> n=28	<i>Pseudomonas</i> n=11	<i>Proteus</i> n=5	<i>Citrobacter</i> n=2
1	Ampicillin	25 (26.04)	8 (28.57)	3 (27.27)	1 (20)	0
2	Ampicillin/Salbactam	64 (66.67)	21 (75)	6 (54.54)	3 (60)	2 (100)
3	Cefodroxil	42 (45.1)	13 (46.42)	5 (45.45)	2 (40)	1 (50)
4	Cefoperazone	56 (58.34)	16 (57.14)	5 (45.54)	5 (100)	1 (50)
5	Ceftazidime	39 (40.62)	10 (35.71)	4 (36.36)	2 (40)	1 (50)
6	Ceftazidime / clavulaunic acid	63 (65.62)	18 (64.28)	7 (63.63)	3 (60)	1 (50)
7	Cefotaxime	45 (46.87)	13 (46.42)	5 (45.54)	3 (60)	1 (50)
8	Aztreonam	57 (59.37)	16 (57.14)	6 (54.54)	3 (60)	1 (50)
9	Cefipime	42 (43.75)	13 (46.42)	5(45.54)	3(60)	1 (50)
10	Amikacin	67 (69.79)	20 (71.42)	8 (72.72)	5 (100)	2 (100)
11	Cotrimoxazole	29 (30.20)	10 (35.71)	4 (36.36)	2 (40)	1 (50)
12	Levofloxacin	46 (47.91)	15 (53.57)	3 (27.27)	3 (60)	2 (100)
13	Imipenem	85 (88.54)	27(96.42)	7 (63.63)	5 (100)	2 (100)
14	Piperacillin / Tazobactam	77 (80.20)	26 (92.85)	7 (63.63)	5 (100)	1 (50)
15	Colistin	90 (93.75)	26 (92.85)	6(54.54)	NA	2 (100)
16	Tigecycline	85(88.54)	24 (85.71)	NA	5(100)	2 (100)
17	Meropenem	85 (88.54)	27 (96.42)	7 (63.63)	4 (80)	2 (100)

\*NA=Not applicable

**Table.6** Antibiotic sensitivity of gram-positive isolates

S.No.	Antibiotics	<i>Staphylococcus aureus</i>		MRCONS n=4	MSCONS n=9	<i>Streptococcus pyogenes</i> n=1
		MRSA n=14	MSSA n=83			
1	Penicillin	0	18 (21.68)	0	2 (22.23)	1 (100)
2	Azithromycin	7 (50)	33 (39.75)	2 (50)	6 (66.67)	1 (100)
3	Amikacin	10 (71.42)	75 (90.36)	3 (75)	9 (100)	1 (100)
4.	Cefoxitin	0	83 (100)	0	9 (100)	1 (100)
5.	Ciprofloxacin	7 (50)	47 (56.62)	4 (100)	5 (55.56)	1 (100)
6	Vancomycin	14 (100)	83 (100)	4 (100)	9 (100)	1 (100)
7	Linezolid	14 (100)	83 (100)	4 (100)	9 (100)	1 (100)
8	Teicoplanin	10 (71.42)	73 (87.95)	3 (75)	7 (77.78)	1 (100)
9	Doxycycline	14 (100)	73 (87.95)	3 (75)	7 (77.78)	1 (100)
10	Cotrimoxazole	3 (21.42)	25 (30.12)	1 (25)	4 (44.45)	1 (100)
11	Daptomycin	10 (71.42)	71 (85.54)	3 (75)	6 (66.67)	NA
12	Pristinomycin	10 (71.42)	71 (85.54)	3 (75)	6 (66.67)	NA
13	Piperacillin / Tazobactam	10 (71.42)	83 (100)	1 (25)	9 (100)	NA
14	Meropenem	12 (85.71)	75 (90.36)	3 (75)	8 (88.89)	NA

\*NA=Not applicable

In this study, out of 410 pus samples collected and analyzed for bacterial growth, 257 (62.68%) samples were found to be culture positive. Highest number of samples were from department of general surgery (n=113) followed by casualty (n=44) and plastic surgery (n=38). Majority of the samples were of patients in the age group of 21-30 years. Additionally, male patients (n=230) were higher in number than females (n=180). Biradar *et al.*, has shown similar results with males comprising majority of the study samples.<sup>(10)</sup>

From 257 growth positive samples, 12 different species of bacteria were identified,

out of which 56.78% (n=146) were gram negative and 43.16% (n=111) were gram positive. The predominant presence of aerobic gram negative isolates compared to gram positive has been supported by previous studies conducted by Ghosh *et al.*, and Zubair *et al.*<sup>(11,12)</sup> Furthermore, almost all studies conducted in developing countries like India and Africa, have reported more growth of the gram negative isolates compared to gram positive bacterial isolates in pus samples.<sup>(10,13,14)</sup>

Different gram negative bacteria identified from pus cultures were as follows: *E. coli*, *Klebsiella sp.*, *Acinetobacter*, *Pseudomonas*

sp., and *Proteus* sp. *Streptococcus* sp., *Staphylococcus aureus*, MRSA, MSSA, MCONS, *Citrobacter* and *Proetus* sp. were the gram positive bacteria to be isolated from pus cultures. The most frequently found pathogens were *Staphylococcus aureus* (n=97) and *Klebsiella* spp (n=96), which belongs to the gram positive and negative bacteria respectively. This is followed by *E.coli* (n=28), CoNS (n=13) and *Pseudomonas* spp (n=11). Studies conducted by Mantravadi *et al.*, Lee *et al.*, and Rao *et al.*, has also reported *Staphylococcus aureus* to be the most frequently isolated bacterial pathogen from pus cultures.<sup>(15,16,17)</sup> In contrast to results presented in this study, Aghnihotri *et al.*, Duggal Swati *et al.*, and Basu *et al.*, has reported *Pseudomonas* to be the most commonly isolated pathogen which is followed by *Staphylococcus*.<sup>(18,19,20)</sup> Another study by Rameshkannan *et al.*, found *E.coli* to be the most frequently isolated organism from pus cultures.<sup>(21)</sup>

Among *Staphylococcus aureus* 75.45% isolates were MSSA and the rest 12.72% were MRSA. *Staphylococcus aureus* showed highest sensitivity for vancomycin (100%), linezolid (100%) and doxycycline (100%). Chauhan *et al.*, and Jain *et al.*, has reported similar results regarding antibiotic sensitivity of the *Staph. aureus*, where it showed higher sensitivity for aminoglycosides such as vancomycin and clindamycin.<sup>(22,23)</sup> Taiwo *et al.*, showed highest sensitivity for vancomycin (100%) and slightly less for linezolid (98%)<sup>(24)</sup>. However, Hanumanthappa *et al.*, has shown slightly lesser sensitivity of vancomycin (87.3%) among *Staph. aureus*.<sup>(25)</sup> The gram negative isolates were most sensitive towards imipenem (86.30%) followed by meropenem (85.61%) and colistin (83.56%). Rao *et al.*, has reported similar results with imipenem showing highest sensitivity among gram negative isolates.<sup>(17)</sup>

In conclusion, pyogenic infections are one of the cardinal causes of morbidity across the globe. There is a growing concern regarding the treatment of such infections as the multidrug resistant strains are evolving rapidly, thereby delaying the recovery of the infected individuals. The present study has shown that though the *Staphylococcus aureus* remains to be the dominant cause of such infections, the role of the gram negative isolates cannot be diminished. Antibiotic resistance further complicates the treatment. Thus, an empirical therapy based on the sensitivity data helps clinicians to select the appropriate antibiotic for the pathogen. Our study helps clinicians in selecting the antibiotics that are susceptible for the bacteria and also in preventing further resistance among pathogens by its appropriate use.

### **Limitations**

In this study only limited drugs were analyzed for the sensitivity of antibiotics in various microorganisms in a single hospital setting. However, multi-center study with larger sample size will provide significant results. Further, constantly changing antibiotic resistance patterns in bacteria requires periodic monitoring of their sensitivity profile.

### **Conflict of interest**

Authors have no conflict of interest.

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