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Bacteriological Profile and Antibiotic Susceptibility Pattern of Pus Isolates in Patients Attending a Tertiary Care Hospital in Kolkata, West Bengal

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Abstract: ***Objective: Aim of the present study is to determine the bacteriological profile of pus samples and their antibiotic susceptibility pattern in patients attending a tertiary care hospital in Kolkata, West Bengal. Materials and methods: The present study is a prospective study done in a tertiary care hospital in Kolkata. The study was done for a period of one year. Pus samples received for diagnostic Microbiology was subjected to culture and antibiotic susceptibility testing. Results: Our study showed that out of the 690 pus samples received in the Microbiology Department, 442 were culture positive (64.05%). Out of them, Klebsiella pneumoniae was found to be the most predominant organism (29.05%), followed by Escherichia coli (21.5%), Pseudomonas aeruginosa (18.16%), Staphylococcus aureus (17.6%), Enterococcus species (10.6%) and Acinetobacter species (3.63%). Conclusion: The changing antimicrobial resistance pose a challenge in treating the pyogenic infections. Microbiological analysis and antimicrobial susceptibility of pus isolates serve as a useful tool for judicial and appropriate use of antibiotics. This can minimize the emergence of drug resistant strains in future.***

Keywords: Bacteriological profile, antibiotic sensitivity, antimicrobial resistance, pus

1. Introduction

Pyogenic infections are characterized by local inflammation associated with pus formation.[1] These infections are generally caused by one of the pyogenic bacteria.

Pus is a collection of the white or yellow fluid, formed at the site of inflammation during an infection. It is made up of dead tissue, white blood cells and damaged cells.[2]

The occurrence of wound infections depends on multiple factors, like the host defense mechanism, microbial load and condition of the wound.[3] Effective treatment of wound infections depend upon the proper understanding of causative pathogen, pathophysiology of the infection process and pharmacology of the therapeutic agents.

Antibiotics are widely used for therapeutic and prophylactic purposes, but their irrational use in patients has led to a surge in antibiotic resistance.[4] Infections from resistant organisms are linked to increase mortality and economic loss.[5]

Antimicrobial resistance (AMR) especially in Gram Negative bacteria have emerged as a significant public health problem throughout the world. This concern is mainly due to the insufficient treatment options available. India faces one of the world's greatest burdens of drug-resistant pathogens.[6]

Routine surveillance is therefore essential for constant monitoring of antimicrobial resistance pattern in clinically important pathogens.

The present study aims to highlight the bacteriological profile of the pus samples and their sensitivity patterns to different antibiotics that will help the clinicians to provide an effective and rational treatment.

Setting: A cross-sectional study with secondary data collection was done in a tertiary care hospital in Kolkata.

Duration: The study was done for a period of one year from 1st December, 2023 to 30th November, 2024.
Pus samples received by the Microbiology Department were processed and identified using standard protocols.

Sampling methods: Secondary data collection from register taking all patients between 01.12.2023 to 30.11.2024 (Total enumeration).

Sample size calculation:
The sample size was calculated after studying the prevalence of risk factors from various studies and taking their average, which was 50%. The sample size was estimated at 5% level of significance with an allowable error of 5%, using the following formula:

$$n = (z\ \alpha)^2\ p\ q\ /\ L^2$$

Where,
n= Sample size, p= Prevalence, q= (p-1), L= Allowable error, Z Alpha = Z 0.05 = 1.96

So,
p = 0.51
q = 0.49
L = 0.05

So, n is (1.96 x 1.96 x 0.51 x 0.49) / (0.05 x 0.05) = 384

However, we took all the patient data from 01.12.2023 to 30.11.2024 (Total enumeration).

Inclusion criteria:
Pus samples (both in-patient and out-patient) received in the Microbiology Department for a period of one year.

Exclusion criteria:
Samples other than pus (blood, urine, body fluids) were not accepted for this study.
Collection and processing of samples.

Collection and processing of samples:
All the pus samples received in the Department of Microbiology were processed. Microscopical examination was done for the presence of pus cells, RBC, bacteria or any yeast. These samples were then inoculated in Blood agar, Mac conkey agar and Chocolate agar and finally incubated at 37 degree Celsius in aerobic conditions for 24 hours.

The plates were examined the next day, the colony count was noted as Scanty, Moderate growth or Heavy growth, the colony morphology was noted, Gram stain was performed for each colony, Catalase and Oxidase tests were done and then biochemicals were put up for further identification. Antibiotic susceptibility testing was carried out by Kirby-Bauer disc diffusion method.Vancomycin susceptibility was carried out by E-strip method for Staphylococcus and Enterococcus.

The biochemicals were read the next day along with the antibiotic sensitivity plate. Zone of inhibition of each drug was measured with calipers and final report was released according to the CLSI guidelines.

Drugs used:
For Gram positive organisms, drugs used were Penicillin, Ampicillin, Cefoxitin, Amoxycillin-Clavulanic acid, Cephalexin, Cefuroxime, Cefotaximee, Cefepime, Erythromycin, Azithromycin, Clindamycin, Vancomycin, Linezolid, Teicoplanin, Daptomycin, Ciprofloxacin, Levofloxacin, Ofloxacin, Doxycyline, Tetracycline and Tigecycline.

For Gram negative organism, drugs used were Penicillin, Ampicillin, Cefoxitin, Amoxycillin-Clavulanic acid, Piperacillin-Tazobactam, Cefoperazone-Sulbactam, Cephalexin, Cefuroxime, Cefixime, Cefotaxime, Ceftazidime, Ceftriaxone, Cefepime, Aztreonam, Imipenem, Meropenem, Ertapenem, Gentamicin, Amikacin, Netilmicin, Ciprofloxacin, Levofloxacin, Ofloxacin, Moxifloxacin, Doxycyline, Tetracycline, Tigecyclin and Cotrimoxazole.

Reporting was done after reading the zone size of each drug, following the CLSI guidelines.

Data Collection
The data was collected and analysed using Microsoft Excel sheet and other statistical softwares.

2. Results

Table 1

Organism isolated	*Percentage of distribution (%)*
Klebsiella pneumoniae	128 (28.95%)
Escherichia coli	95 (21.5%)
Pseudomonas aeruginosa	80 (18.09%)
Staphylococcus aureus	78 (17.64%)
Enterococcus species	45 (10.18%)
Acinetobacter species	16 (3.61%)

Table 2

Gender	*No. of patients (%)*
Male	386 (55.96%)
Female	304 (44.04%)

Table 3

Age group involved	*Percentage of distribution (%)*
< 20 years	13 (1.88%)
21-40 years	283 (41.01%)
41-60 years	196 (28.4%)
61-80 years	184 (26.66%)
>81 years	14 (2.02%)

Table 4: Antibiotic sensitivity pattern of Klebsiella pneumoniae

Antibiotic group	*Sensitivity percentage (%) (ICU)*	*Sensitivity percentage (%) (IPD/OPD))*
Fluoroquinolones	33.3%	40%
Aminoglycosides	66.7%	80%
Tigecycline	100%	100%
Carbapenems	33.3%	60%
Cefoperazone-Sulbactam	33.3%	60%
Piperacillin-Tazobactam	33.3%	78%
Colistin	98%	100%

Table 5: Antibiotic sensitivity pattern of Escherichia coli

Antibiotic group	*Sensitivity percentage (%) (ICU)*	*Sensitivity percentage (%) (IPD/OPD))*
Fluoroquinolones	12.5%	44.4%
Aminoglycosides	87.5%	100%
Tigecycline	87.5%	100%
Carbapenems	62.5%	77.8%
Cefoperazone-Sulbactam	37.5%	66.7%
Piperacillin-Tazobactam	40%	86%
Colistin	100%	100%

Table 6: Antibiotic sensitivity pattern of Pseudomonas aeruginosa

Antibiotic group	*Sensitivity percentage (%) (ICU)*	*Sensitivity percentage (%) (IPD/OPD))*
Fluoroquinolones	71.4%	60%
Aminoglycosides	85.7%	60%
Minocycline	100%	100%
Carbapenems	85.7%	80%
Cefoperazone-Sulbactam	85.7%	60%
Piperacillin-Tazobactam	85.7%	60%
Colistin	100%	100%

Table 7: Antibiotic sensitivity pattern of Staphylococcus aureus

Antibiotic group	*Sensitivity percentage (%) (ICU)*	*Sensitivity percentage (%) (IPD/OPD))*
Fluoroquinolones	0%	20%
Aminoglycosides	50%	100%
Tigecycline	100%	100%
Vancomycin	100%	80%
Teicoplanin	100%	100%
Linezolid	100%	100%
Oxacillin	0%	60%
Cotrimoxazole	100%	80%

Table 8: Antibiotic sensitivity pattern of Enterococcus species

Antibiotic group	*Sensitivity percentage (%) (ICU)*	*Sensitivity percentage (%) (IPD/OPD))*
Fluoroquinolones	0%	25%
High level Gentamicin	0%	50%
Tigecycline	100%	100%
Linezolid	100%	75%
Vancomycin	20%	100%
Teicoplanin	20%	75%
Penicillin	0%	50%
Amoxycillin-clavulanic acid	0%	50%

Table 9: Antibiotic sensitivity pattern of Acinetobacter species:

Antibiotic group	*Sensitivity percentage (%) (ICU)*	*Sensitivity percentage (%) (IPD/OPD))*
Fluoroquinolones	60%	100%
Aminoglycosides	20%	100%
Tigecycline	42.9%	100%
Carbapenems	40%	80%
Cefoperazone-sulbactam	11.6%	100%
Cotrimoxazole	50%	0%
Colistin	100%	100%
Minocycline	28.6%	100%

3. Discussion

Pyogenic infections are usually associated with local and systemic infections and lead to pus formation.

Gram negative bacteria such as Klebsiella pneumoniae, E.coli, Pseudomonas aeruginosa, Acinetobacter species and Gram-positive bacteria like Staphylococcus aureus and Enterococci are the common causative agents.[7]

In this study, the predominant pathogens were Gram negative bacteria (72.34%). This was in accordance with a study done by Shama et al [8], which also showed the predominance of Gram-negative bacteria.

This was also in accordance with a study done by Swati et al [9] in which Gram-negative bacteria were mostly isolated.

Similarly, predominance of Gram-negative bacteria (85.05%) in wound swabs has been shown in a study done by Kemebradikumo Ponder et al [10] conducted in Nigeria.

Regarding age distribution in the current study, the maximum number of pus samples came from the age group 21-40 years (43.82%).

This result is comparable with that of Rashid et al[11], where the main age group affected was 21-40 years (41.2%)

In our study, the Male: Female ratio was 1.27:1. This result has been corroborated by a study done by Bashir et al[12], where the ratio was 1.2:1.

According to the present study, Klebsiella species was found to be the most common pathogen isolated among the pus samples (29.05%). This is in accordance with a study done by Krishnamurthy et al [13], where Klebsiella pneumoniae was the predominant organism isolated (34.46%).

Among the Gram-positive organisms, the percentage of Staphylococcus aureus isolated was 17.6%. This corroborates with the study done by Mita et al [14], where the percentage of Staphylococcus aureus isolated was 22.9%.

Klebsiella isolated was found to be resistant to many drugs, only being 100% sensitive to Tigecycline, followed by Colistin. This is in accordance with a study done by Nand Kishore et al [15], which showed a high sensitivity to Tigecycline (87%) and Colistin (92%) respectively.

Acinetobacter species isolated especially from the samples received from Intensive Care Unit (ICU) were found to be highly drug resistant. [16]

Pseudomonas aeruginosa isolated from the pus samples were mostly sensitive to Carbapenems and Colistin. [17]

According to our study, Staphylococcus aureus was highly sensitive to Linezolid (100%), Vancomycin and Teicoplanin. Another similar study done by Kumar et al shows high sensitivity to Linezolid (91.7%.) [18]

Similar study done by Murugesan et al showed Staphylococcus aureus is highly sensitive to Vancomycin (94.25%).[19]

Enterococcus species isolated from pus samples were found to be highly sensitive to Linezolid. This is similar to a study done by Jabbari Shiadah et al which also showed a high sensitivity of Enterococcus to Linezolid. [20]

Our study showed existence of drug resistance to multiple antibiotics in Klebsiella pneumoniae, Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, Acinetobacter species and Enterococcus species isolated from pus samples.

Hence, formulation of antibiotic policies and proper infection control measures should be considered essential for the management of patients. [21]

4. Conclusion

Knowledge about the bacteriological profile of pus samples and their antibiotic susceptibility pattern can serve a s a useful tool for the clinicians to start an appropriate treatment. Since the frequency of multiple drug resistance among both Gram positive and Gram-negative bacteria is high, therefore, proper antimicrobial stewardship is required in Hospitals.

This will not only benefit the patients, but also assist the clinicians in selecting the appropriate antibiotics, help in de-escalation of antibiotics and reduce the risk of emergence of the multidrug resistance.

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