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Microbiology

Aerobic Bacteriological Profile with Antibiogram of Pus Isolates in a Tertiary Care Hospital, Guntur, Andhra Pradesh

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Abstract Original Research Article

Pus is a major sample received to the microbiology laboratory. A diverse group of pathogens will cause wound infections and the antibiotic sensitivity and resistant pathogens will be different in different hospitals. This may increase the antimicrobial resistance creating a great hazard globally. *Aims and Objectives*: To isolate and identify the bacteria from pus samples and perform their antibiotic susceptibility testing of the isolates. *Materials and Methods*: A total of 621 samples were collected from different departments irrespective of age and gender and processed in the department of Microbiology Guntur medical college, Guntur according to standard guidelines. Antimicrobial susceptibility testing was done by Kirby Bauer Disc Diffusion Testing and further ESBL detection was done by Disc Synergy test and results were interpreted. *Results*: More number of males participated in the present study with maximum age group being 45 - 60 years. More number of samples were from Surgery. The pathogens isolated in the present study were Klebsiella, Escherichia coli, Pseudomonas, and Staphylococcus. The most antibiotics susceptible were Piperacillin and Tazobactum, Imipenem and Gentamycin. ESBL production in the present study was 44.5%. *Conclusion*: The present study revealed the pathogens causing pyogenic infections and their role in emergence of antimicrobial resistance which is becoming a global pandemic.

Keywords: Antimicrobial resistance, ESBL, Kirby Bauer, Disc Synergy.

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Interduction

Pyogenic bacteria forms pus with aggregation of dead leukocytes. Wounds occur as a result of cuts, abrasions, burns or after surgery and these provide an environment for microbial colonization because of presence of moisture, warmth and nutrition for the growth of bacteria. Colonization along multiplication leads to many complications which may sometimes endanger life of the patient. Various etiological agents include bacteria, virus, fungi, parasites cause these wound infections. Sometimes they may progress to chronic infections. The pathogens may differ depending on the problem, type of surgery. The most common causative agents include Staphylococcus aureus, Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Proteus species (Asmabegum Birdar et al., 2016) [1]. Incidence of wound sepsis range from 10 – 33% in India (Mantravadi H et al., 2015) [2]. Methicillin Resistant Staphylococcus Aureus is one of the most important causes of Hospital Acquired

Infections. The spread of multi drug resistant organisms plays an important role in the wound healing. Development of antimicrobial resistance is an urgent threat which is high in resource poor settings due to easy access of over-the-counter antibiotics (Asmabegum Birdar *et al.*, 2016) [3]. The current study has been conducted to isolate and identify aerobic bacteria from pus samples and to study their antimicrobial susceptibility pattern.

Materials and Methods: This prospective cross sectional study has been conducted in the department of Microbiology from March to August 2024 for the pus samples received from different departments from Government General Hospital, Guntur. A total of 621 samples from both outpatient and inpatient were included in the study. Patients who were on antibiotics were excluded from the study. Pus samples were collected with sterile disposable cotton swabs and aspirates in syringe. They were transported to the

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microbiology laboratory immediately. They inoculated on to Blood agar (BA), MacConkey agar (MA). Culture plates were incubated at 37°C for 24 hrs to 48 hrs in aerobic condition. After incubation, identification of bacterium from positive cultures was done with a standard microbiological technique which includes motility testing by hanging drop preparation, gram staining and biochemical reactions such as catalase, coagulase, indole, methyl red, Voges-Proskauer, citrate, urease, phenyl pyruvic acid test and oxidase test, Triple sugar Iron agar test. Antibiotic susceptibility testing was carried out by Kirby Bauer 's Disc Diffusion method on Muller Hinton Agar and interpreted as per Clinical Laboratory and Standard Institute guidelines.

The isolates were tested against Ampicillin (10 mg), Amoxyclav (20/10mg), Gentamicin (10mg), Amikacin (30 mg), Ciprofloxacin (5 mg), Ceftazidime (30 mg), Cefotaxime (30 mg), Imipenem (10 mg) and Piperacillin – Tazobactum (100/10mg). For grampositive isolates, Erythromycin (5mg), Clindamycin (2mg), Doxycycline(30mg), Ciprofloxacin (5mg), Linezolid (30mg), Vancomycin(30mg). ESBL was detected by Disk Synergy test as per EUCAST

guidelines. Methicillin Resistant Staphylococcus aureus (MRSA) was detected by Cefoxitin disc diffusion test. Lawn culture was done onto Mueller– Hinton agar plate. A 30 μ g cefoxitin disc was placed and incubated at 37°C for 24 hrs. The zone of inhibition of S. aureus \leq 21 mm was considered as methicillin resistant as shown in fig 4. *S. aureus* ATCC 25923 and *E. coli* ATCC 25922 were used as quality control.

RESULTS

In the present study males were 62% where as females were 38%. Maximum age group affected were 45-60 years followed by 31-45 years. Majority of the samples were from Surgery, Orthopedics followed by Obstetrics and Gynaecology. Among the total samples processed pus samples collected with sterile swabs revealed 205 with bacterial growth where as 309 were reported with no bacterial growth. 77 showed growth for aspirates whereas 30 were sterile. The isolation rate of bacterial pathogens was shown in below fig 1. Antibiotic Susceptibility testing results for Gram negative bactyeria and Gram positive bacteria were shown in fig2& 3 respectively.

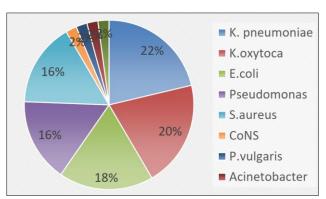


Fig. 1: Isolation rate of bacterial pathogens

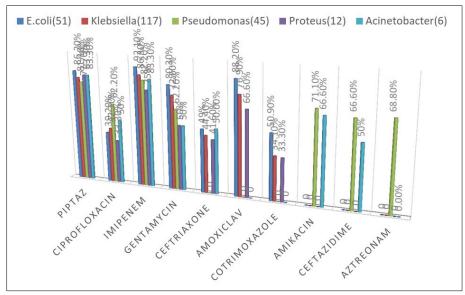


Fig. 2: Antibiotic susceptibility testing of Gram Negative Bacteria

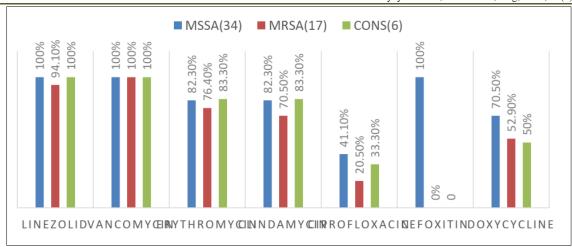


Fig. 3: Antibiotic Susceptibility Testing of Gram Positive bacteria

Table 1: ESBL Production rate

Table 1. ESDE 1 roduction rate	
ESBL PRODUCING ORGANISMS AMONG THE ISOLATES	PERCENTAGE OF ESBL PRODUCERS
KLEBSIELLA spp.	51.3%
E.coli	44.4%
Pseudomonas aeruginosa	40%
Proteus spp.	33.3%
Total	44.55%



Fig. 4: Cefoxitin Disc Diffusion Testing

DISCUSSION

This study has been conducted in the department of Microbiology, Guntur medical college, Guntur. A total of 621 samples were collected from different departments of Govt General Hospital, Guntur were processed. 282 samples (45%) showed bacterial growth which is similar to a study done by Rao *et al.*, (2014) [4]. The male to female ratio was 1.6:1. Maximum number of samples received in the present study were in the age group of 46-60 years. Asmabegaum Biradar [1], *et al.*, reported maximium samples in the age group of 21-30 years. Maximum number of samples

were from Surgery followed by Orthopedics and Gynaecology which is similar to a study by Vikas Jain et al 2016⁵. Major bacterial isolate was Klebsiella followed by E.coli in gram negative bacteria. Staphylococcus aureus was isolated in maximum among the gram positives. Among the Staphylococcus isolated MSSA were 34%, MRSA 17% followed by CoNS 7%. Wadekar et al., 2020 [6], reported the most common pathogens as Pseudomonas and E.coli which is in similar proportion to the present study. They reported Klebsiella as 5.1% where as the present study reported at its highest extent. These organisms are commonly found in hospital environment.

They tend to be resistant to common antiseptics and are also multidrug resistant [6]. The present study reported Staphylococcus as most common gram positive which is similar to a study by Kumari PH et al., [7]. MRSA strains are resistant to Beta lactam antibiotics -Penicillins and Cephalosporins. It is due to acquisition of mec A gene. In the present study the maximum susceptibility was shown to Piperacillin and Tazobactum and Imipenem which is correlating with a study by Rameshkannan et al., 2014 [8]. ESBL resistance in the present study was 44.5% where as Wadeker et al showed it at 61.2%. Staphylococcus were highest susceptible to Vancomycin and Linezolid which is similar to Shittu A O et al., [9]. In the present study 16% were Pseudomonas isolates which showed greater sensitivity to Piperacillin and Tazobactum and this finding is similar to Jain et al., [10]. As antibiotic resistance is increasing and is due to inadvertent use of antibiotics and the present study showed the role of different etiological agents in causing wound infections and the most common isolate among gram negatives is Klebsiella and among gram positives is Staphylococcus. Hence antibiotic policies should be formulated and infection control measures should be appropriate to avoid emergence of multi drug resistant organisms [11].

CONCLUSION

The present study revealed emergence of multi drug resistant organisms which is of greater concern. Irrational use of antibiotics should be addressed and strict surveillance on infection prevention and control measures should be adopted.

Conflicts of Interest: Nil

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