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AEROBIC BACTERIAL PROFILE OF ASPIRATED PUS SAMPLES AT A TERTIARY CARE HOSPITAL, PATNA, BIHAR

Wasim Ahmad	Senior Resident, Department of Microbiology, Indira Gandhi Institute of Medical Sciences, Patna.
Pallavi Priya*	Senior Resident, Department of Microbiology, Indira Gandhi Institute of Medical Sciences, Patna.*Corresponding Author
Kumar Saurabh	Assistant Professor, Department of Microbiology, Indira Gandhi Institute of Medical Sciences, Patna.
Namrata Kumari	Professor, Department of Microbiology, Indira Gandhi Institute of Medical Sciences, Patna.

ABSTRACT Abscess is an accumulation of pus in tissue and it is caused by suppuration deep within a tissue, an organ or confined space. Though the bacterial profile from pus samples remains almost similar in various studies, the irrational use of antibiotics has lead to the emergence of various drug resistant pathogens. In this retrospective study we have taken 290 aspirated pus samples for aerobic culture and sensitivity and found 23 different aerobic bacterial isolates. Most common isolate was *Escherichia coli* (n-12) followed by *Staphylococcus aureus* (n-8) and one each was *Acinetobacter spp., Klebsiella pneumoniae* and *Streptococcus spp*. Sensitivity of *E coli* was found to be low for ciprofloxacin, amoxycillin+clavulanic acid, ceftazidime, cefepime and aztreonam; while sensitivity was high for amikacin, imipenem, colistin and tigecycline. *Staphylococcus aureus* was highly sensitive to most of the antibiotics that had been put for sensitivity testing. A continuous inspection should be carried out to monitor the antimicrobial susceptibility of bacterial isolates from aspirated pus samples to choose appropriate antibiotics for prophylaxis and treatment of infections.

KEYWORDS : Aspirated pus, Antibiotics, Sensitivity testing, Aerobic bacteria.

INTRODUCTION

Pus is a white to yellow fluid comprised of dead white blood cells (WBC), cellular debris and necrotic tissues, produced as a result of infection by any one of the pus producing bacteria (pyogenic infection). Abscess is an accumulation of pus in tissues and it is caused by suppuration deep within a tissue, an organ or a confined space.[1] Endogenous wounds and abscesses may be associated with different internal infections. Many of these processes are nosocomial (i.e acquired in health care institutions) and are contracted after invasive procedures, surgical manipulations or the placement of prostheses in hospital settings. Conversely, these may be derived from hematogenous spread of microorganisms from primary site of infection to distant (i.e., metastatic) sites. Finally , there may be a direct extension of bacteria from an adjacent site of infection or from ruptured viscera.[2] The commonest pyogenic bacteria are *Staphylococcus aureus, Streptococcus pyogenes, Pneumococcus* and coliform bacilli such as *Escherichia coli, Proteus species* and *Pseudomonas aeruginosa*.[3] Wide variety of aerobic and anaerobic species of bacteria may be present either singly or in combination in infections of wounds and other soft tissues. Some infections resolve without any specific therapy but some infections, especially mixed infections, can cause severe synergic infections calling for prompt antibiotic therapy .[4]

Different studies have been conducted globally from time to time to assess the bacterial profile and the antibiotic susceptibility pattern in pus samples. This is particularly relevant for the treating physicians, who needs to start empirical antibiotic therapy until the lab culture reports are awaited (Rameshkannan *et al.,* 2014).[5]

Though the bacterial profile from pus samples remain similar in various studies, there is considerable variation in the antibiotic susceptibility pattern of these isolates highlighting the increasing threat of emergence of resistant bacteria and hence there is a need for continuous surveillance of such changing trends.[6] Therefore, we designed this study to determine the common bacterial profile with special focus on antibiotic susceptibility pattern of isolated strains from the aspirated pus sample.

AIMS AND OBJECTIVE

1. To identify the common bacterial isolates from aspirated pus samples.
2. To determine their antimicrobial sensitivity pattern to guide the clinicians for starting appropriate empirical therapy.

MATERIAL AND METHODS

Type of study and duration - This is a cross-sectional institute based study where the samples were collected over a period of six months duration from January 2017 to June 2017.

Inclusion and Exclusion criteria - All aspirated pus collected from patients of different inpatient departments were included in the study, whereas, the pus from open wounds and pus collected by swab were excluded.

Methods - Needle aspiration was done following the standard precautions, which was either ultrasonography (USG) guided or by surgical procedure. Aspirated body pus were immediately transported to microbiology laboratory after collection and subjected to gram stain and aerobic culture. The sample was inoculated on Blood agar and McConkey agar for solid plate culture and simultaneously it was enriched by putting into Brain Heart Infusion (BHI) broth following standard laboratory protocol. Inoculated media were aerobically incubated overnight at 37°C. Identification of isolated bacteria was done by conventional methods and their antimicrobial sensitivity testing was performed by Kirby-Bauer disk diffusion method as per CLSI 2017 guidelines.[7] The culture was declared sterile if there was no growth after 48 hours of aerobic incubation.

Antimicrobial Agents - Antibiotic discs containing ciprofloxacin (5mcg), gentamicin (10mcg), amikacin (30mcg), amoxicillin-clavulanic acid (20/10mcg), ceftazidime (30mcg), cefepime (50mcg), tobramycin (10mcg), aztreonam (50mcg), piperacillin-tazobactam (100/10mcg), imipenem (10mcg), tigecycline (15mcg), colistin (10mcg), tetracycline (30mcg), erythromycin (15mcg), cotrimoxazole (25mcg), cefuroxime (30mcg), clindamycin (2mcg), doxycycline (30mcg), teicoplanin (30mcg), vancomycin (30mcg) and linezolid (10mcg), were obtained from Himedia Laboratories (Mumbai, India) and used as per manufacturer's instructions.

Antibiotics Susceptibility Testing - Antibiotic susceptibilities of bacterial isolates were determined according to the method recommended by the Clinical and Laboratory Standards Institute

2017. In short, inoculums were prepared for each bacterial isolate by adjusting the turbidity to 0.5 McFarland standard and lawm culture was done on Muller-Hinton agar plates. Antibiotic discs (Himedia, Mumbai, India) were placed on the agar plates and aerobically incubated overnight at 37°C. The zones of inhibition were then measured and noted. *Extended spectrum beta lactamase* (ESBL) production was observed using disc synergy test using ceftazidime and ceftazidime plus clavulanic acid discs, whereas *Methicillin Resistant Staphylpcoccus aureus* (MRSA) screening was done using cefoxitin disc diffusion method as per the CLSI guidelines. [7] Data were entered into Microsoft excel sheet and different percentages were calculated and presented in tabular form and in pie charts.

RESULTS AND DISCUSSION

Table 1: Different Aerobic Bacterial Isolates From Aspirated Pus Samples

Escherichia coli	12 (52.17%)	Total GNB 14 (60.87%)
Acinetobacter spp.	1 (4.34%)	
Klebsiella pneumonae	1 (4.34%)	
Staphylococcus aureus	8 (34.78%)	Total GPC 9 (39%)
Streptococcus spp.	1 (4.34%)	

Figure 1: Different aerobic Bacterial isolates from aspirated pus samples

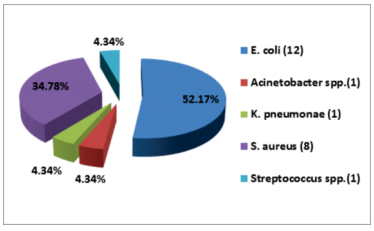

Table 2: Sensitivity In Percentage Of Aerobic Bacterial Isolates From Aspirated Pus Samples

Antibiotics	*E. coli*	*S. aureus*	*Acinetobacter sp.*	*K. pneumoniae*	*Streptococcus sp.*
Ciprofloxacin	00	37	00	00	00
Gentamicin	58	12	00	00	NT
Amikacin	75	87	00	00	100
Amoxycillin/ Clavulanicacid	8	NT	00	00	NT
Ceftazidime	17	25	00	00	00
Cefepime	8	NT	00	00	NT
Tobramycin	58	NT	00	00	NT
Aztreonam	25	NT	100	00	NT
Piperacillin/ Tazobactam	42	NT	00	00	100
Imipenem	92	NT	00	00	NT
Tigecycline	100	NT	100	100	NT
Colistin	92	NT	100	100	NT
Tetracycline	NT	75	NT	NT	100
Erythromycin	NT	87	NT	NT	100
Cotrimoxazole	NT	12	NT	NT	NT
Cefuroxime	NT	87	NT	NT	NT
Clindamycin	NT	87	NT	NT	NT
Doxycycline	NT	87	NT	NT	NT
Teicoplanin	NT	87	NT	NT	100
Vancomycin	NT	100	NT	NT	100
Linezolid	NT	100	NT	NT	100

(Abbreviation: NT-Not Tested, E. coli - Escherichia coli, K. pneumoniae - *Klebsiella pneumoniae*)

A total of 290 aspirated pus samples were received for culture and sensitivity over a period of six months duration in microbiology department of IGIMS Patna, out of which 23 (approximately 8%) showed growth of pathogenic bacteria whereas 267 (92%) showed no growth of any pathogenic bacteria. The percentage of isolation is low in comparison to other studies done by Dhiraj Kumar Chaudhary et al and Rugira T et al.[8,9] The lower isolation rate may be due to the presence of anaerobic or fastidious organisms lacking proper sample enrichment before culture, or due to the prior use of antibiotics by patients.

On the basis of conventional bacterial identification technique, such as gram stain, culture characteristic and biochemical reactions, five different bacterial pathogens were identified. Out of these 23 isolates, 14 (61%) were gram negative bacilli and 9 (39%) were gram positive cocci. Similar findings were also present in other studies.[8,9,10,11]

Most common isolate was *Escherichia coli* (52.17%), followed by *Staphylococcus aureus* (34.78%), *Acinetobacter spp.* (4.34%), *Klebsiella pneumoniae*. (4.34%), and *Streptococcus spp.*(4.34%)[Table/Fig-I]. A similar study done by Rugira T et al [9] also found that *Escherichia coli* was the commonest pathogen, while several other studies on pus showed that *Staphylococcus aureus* was the predominant organism isolated. [8,10,11,12]

Antibiotic susceptibility pattern of different isolates are shown in Table II. *Escherichia coli* was found to be highly sensitive to imipenem, colistin, tigecycline and moderately sensitive to amikacin, gentamicin, tobramycin and piperacillin/tazobactam. It showed lower sensitivity to amoxycillin/clavulanic acid, cetazidime, cefepime and aztreonam. Four of the *Escherichia coli* (i.e. 33.33%) isolates were found to be extended spectrum beta lactamase (ESBL) producer.

The isolated *Acinetobacter spp.* and *Klebsiella pneumoniae* were found to be sensitive to tigecycline and colistin only.

Staphylococcus aureus isolates showed higher sensitivity against most of the antibiotics that had been put for the testing such as amikacin, erythromycin, tetracycline, cefuroxime, clindamycin, doxycycline, teicoplanin, vancomycin and linezolid; while sensitivity against ciprofloxacin, gentamicin, ceftazidime and cotrimoxazole were lower in percentage. All the *Staphylococcus aureus* isolates were found to be *methicillin sensitive Staphylococcus aureus* (MSSA).

CONCLUSION

This study reports the most common organism encountered in aspirated pus is *E. Coli*, followed by *Staphylococcus aureus* and others. The susceptibility data from this report may be worth considering while implementing empirical treatment strategies for pyogenic infections. Aminoglycosides (amikacin and gentamicin), piperacillin/tazobactam, imipenem, doxycycline and vancomycin may be used as empirical therapy to cover these organisms. As the antibiotic sensitivity pattern may vary from region to region, continued monitoring of such patterns is needed. More studies need to be carried out with higher number of samples to detect the true burden of antibiotic resistance among organisms and prevent their further emergence by judicious use of drugs. At the same time, stringent health policies should also be formulated and implemented to regulate the purchase and prescription of antibiotics under strict supervision.

REFERENCES :

1. Tucker C. Acute and chronic inflammation. In: Cotran RS, Kumar V, Collins T, Robbins SL (editors). Robbins Pathologic Basis of Disease. 6th ed. Philadelphia: WB Saunders Company; 1999, 50-89.
2. Koneman's colour atlas and textbook of Diagnostic Microbiology, seventh edition, page 9.
3. Mackie & McCartney Practical Medical Microbiology, 14th edition.
4. Duguid J.D, Collee J.G & Fraser A.G lab strategy in diagnosis of infective Syndrome. Practical Medical microbiology, 13 thed Mackie &Mc Cartney 1989 p 617-618.
5. Rameshkannan, S., Nileshraj G, Rameshprabu S, Mangaiarkkarasi A, Meher Ali R. Pattern of pathogens and their sensitivity isolated from pus culture reports in a tertiary care hospital, Puducherry. Indian Journal of Basic and Applied Medical Research December 2014; 4(1): 243- 248.
6. Rozina Arshi Khan, Mahwish Jawaid and Mohammed Khaleel. 2018. Bacteriological Profile and Antibiogram of Isolates from Pus Samples in a Tertiary Care Centre.

Int.J.Curr.Microbiol.App.Sci. 7(01): 387-394.
7. CLSI. Performance Standards for Antimicrobial Susceptibility Testing, 27th ed. CLSI supplement M100. Wayne, PA: Clinical and Laboratory Standards Institute; 2017.
8. Dhiraj Kumar Chaudhary et al. "Bacteriological Profile and Antibiotic Susceptibility Pattern of Wound Infection in Children". EC Microbiology 5.3 (2017): 93-100.
9. Rugira Trojan, Lovely Razdan, and Nasib Singh. Antibiotic Susceptibility Patterns of Bacterial Isolates from Pus Samples in a Tertiary Care Hospital of Punjab, India. International Journal of Microbiology Volume 2016, Article
10. Arundhati Jamatia, Debasish Roy,Rubin Shil, Pranav Kumar Prabhakar. Bacteriological profile and antimicrobial resistance patterns isolates in pus samples at Agartala government medical college. Asian J Pharm Clin Res, Vol 10, Issue 1, 2017, 335-337.
11. Nandkishor Bankar, Archana Wankhade, R B Brahmane, Riddhi Hathiwala, Dhruba Hari Chandi. Bacteriological Profile of pus/ wound swab and Antimicrobial Susceptibility of Staphylococcus Aureus Isolated From pus & wound swab of Indoor Patients of Tertiary Care Hospital, Durg, Chhattisgarh India. International Journal of Innovative Research in Medical Science (IJIRMS) Volume 03 Issue 04 April 2018,1976-1980
12. R.Sujatha, Ankita Tripathi, Vineet, Nidhi Pal. Microbial characterisation of pus isolates and the changing trends in their sensitivity pattern at a tertiary care hospital in Kanpur city. RAMA Univ. J. Med Sci 2016;2(3):1-7