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Bacteriological (Aerobic) Study of Deep Seated Abscess and Its Antibiotic Susceptability Pattern in Mahatma Gandhi Memorial Hospital, Warangal Dr. Janakiram Rao Boothpur^{*1}, Dr. Archana B², Dr. Kondal Rao³, Dr. Padmaja⁴, Dr. Jyothi⁵, Dr. Sreedevi⁶,

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Original Research Article

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Abstract: Pus samples from Deep Seated Abscess from various sites of a total of 332 patients in Mahatma Gandhi Memorial Hospital, Warangal are taken for my study from January 2016 to December 2016 (12 months). The objective of present study is to assess the most common microorganisms causing deep seated infections and their antimicrobial susceptibility to routinely used antibiotics in this locality. The material for present study consisted of 332 pus samples isolated from various abscess, ie. Perotinitis, Pyopneumothorax, Deep Neck Space Infections, Osteomyelitis, Pyogenic Liver Abscess, Breast Abscess, Prostatic abscess, Psoas abscess] and Parotid abscess.Samples were collected under aseptic conditions and subjected to culture and antibiotic susceptibility testing. Descriptive statistics were provided.Out of total 332 pus samples taken from various abscess 119 were culture positive. Organisms from different abscesses are concluded and the outcome of the study shows out of 332 pus samples taken 201 were males and 131 were females, and 119 were culture positive.A total of 30/119 (25.21%) Staphylococcus aureus were obtained in all the abscesses combined, 25/119 (21 %) Proteus Vulgaris, 23/119 (19.32%) Klebsiella Pneumonia, 22/119 (18.48%) Escherichia Coli, and 19/119 (15.96%) Pseudomonas aeruginosa. Among Staphylococcus aureus there are two Vancomycin resistant MRSA strains which are resistant to linezolid. Every gram negative bacilli isolated in this study are sensitive to carbapenem ie.. Meropenem. Maximum sensitivity is shown by Meropenem (100%) and least sensitivity is shown by Ceftazidime (42%). Though pus samples were brought to the dept. of Microbiology, we have collected samples in aseptic precautions mostly from operation theatres, and from Emergency Operation Theatre where Incision & Drainage is commonly done to avoid contaminants and to avoid the delay of transport of specimens to the laboratory. This helps clinicians to choose appropriate antibiotic as early as possible and helps the patients to decrease the days of Hospital stay.

Keywords: Intra-abdominal infections (IAI), Deep neck space infection (DNSI), Pyogenic liver abscess (PLA), osteomyelitis, peritonitis, Psoas abscess.

INTRODUCTION

Pus samples from Deep Seated Abscess from various sites of a total of 332 patients in Mahatma Gandhi Memorial Hospital, Warangal are taken for my study from January 2016 to December 2016 (12 months).Here is brief introduction about abscesses taken for my study.

Pyopneumothorax

Parapneumonic effusions and empyema are common clinical problems with good variety of treatment options, occasionally having poor prognosis [1]. Empyema is usually a complication of pneumonia but may arise from infections at other sites[2,3].

Peritonitis

Generalized peritonitis due to intra-abdominal infections (IAIs), despite advances in surgical techniques, comprise more than 25% of emergency operations in our hospitals [3]. Decline in morbidity and mortality in IAI is a welcome observation in Indian scenario.

Deep neck space infections (dnsi)

Deep neck space infection (DNSI) means infection in the potential spaces and fascial planes of the neck, either with abscess formation or cellulitis [4]. In past, these infections were fairly common; however, with the advent of broad spectrum antibiotics, the incidence of these infections has decreased [8]. In spite of the extensive use of antibiotics, DNSI still remains serious with significant morbidity.

Osteomyelitis

The word "osteomyelitis" originates from the ancient Greek words osteon (bone) and muelinos (marrow) and means the infection of the medullary portion of the bone [6]. It can be classified as acute, subacute, or chronic, depending on the clinical presentation. The decline in prevalence can be attributed to the increased availability of antibiotics and the progressively higher standards of oral and dental health [7].

Pyogenic liver abscess (pla)

Pyogenic liver abscess (PLA), is a potentially life-threatening disease, recognized since the time of Hippocrates [5]. It presents with considerable morbidity and mortality.

Breast abscess

Breast abscess is defined as an acute inflammatory lump which yields pus on incision/aspiration.^[9] It is a common soft tissue infection which is characterized by localized pain, swelling, and redness associated with a mass that may or may not be fluctuant [10].

Prostatic abscess

Prostatic abscess results from focal accumulation of pus within the prostate gland. Untreated abscess may resolve or burst spontaneously. The incidence of prostatic abscess has decreased markedly because of the widespread use of antibiotics and decrease in the incidence of gonococcal urethritis [12].

Psoasabscess

Psoas abscess is regarded as a rare disease in the medical literature. The reported incidence is 0.4/100,000, but it has probably increased in recent years [13]. The causes of psoas abscess in India have also changed in the last decades. At the beginning of the 20 thcentury, psoas abscess was mainly caused by tuberculosis of the spine (Pott's disease).

Parotid abscess

Suppurative parotitis is usually unilateral and may progress to abscess formation. Among salivary glands; the involvement of parotid gland is more common than submandibular gland [14,15].

MATERIALS AND METHODS

The patients included in the study were either in-patients or out patients admitted into M.G.M Hospital, Warangal. Ethical approval was obtained.

Cefoxitin disk screen test

By definition, all methicillin-resistant S. aureus (MRSA) isolates carry the mecA gene, which confers resistance to all beta-lactam antibiotics, including cephalosporins and carbapenems. Apart from using molecular methods to detect the mecA gene directly, the most accurate phenotypic test for the presence of the mecA gene in S. aureus is the cefoxitin disk diffusion test. Cefoxitin is used because it is a more potent inducer of mecA expression than other agents such as oxacillin and the test results are relatively easy to interpret. Testing of ESBL:ESBL detection was done by phenotypic method as per CLSI (2010) recommended clavulanate combined disk and double disk synergy test using cefotaxime (30 µg), ceftazidime (30 µg) and combination of cefotaxime (30 μg)-/ceftazidime (30 μg) and clavulanic acid (10 μg). The zone diameter given by the disks with clavulanate was ≥ 5 mm or larger than those without the inhibitor if an ESBL was produced.

Statistical analysis

The results were tabulated and analyzed by SPSS 16 software using the chi-square (χ^2)-tests. A 'P' value of less than 0.05 was taken as significant. The study was approved by Ethical Committee of our hospital.



Fig-1: Swarming of proteus vulgaris on blood agar



Fig-2: Pigmentation produced by pseudomonas aeruginosa on Nutrient agar



Fig-3: MRSA strains showing oxacillin resistance



Fig-4: MSSA strain showing >22mm zone of inhibition for cefoxitin disc



Fig-5: Detection of ESBL producing organisms with cephalosporin showing small zone of inhibition compared to Amoxicillin/clavulanic acid



Fig-6 :Double disc synergy test showing the inhibition zone around Ceftazidime disc (CA) increasing toward the Ceftazidime plus Clavulanic acid disc (CAC), confirming an ESBL producer.



Fig-7: Triple sugar iron agar control, A/A, K/A and K/K



Fig-8: Amino acid decarboxylase tests



Fig 9: Kirby bauer disc diffusion method on Pesudomonas plate



Fig-10: Kirby- Bauer disc diffusion method

RESULTS

Out of total 332 pus samples taken from various abscess 119 were culture positive. Different abscess have varied results and they were shown here. In this study of Pyopneumothorax we found that staphylococcus aureus was found in (5/21) followed by proteus vulgaris (5/21), Klebsiella pneumonia (4/21),

Escherichia coli (4/21) and Pseudomonas aeruginosa (3/21). In over past 30 years, aerobic Grampositive organisms have been the most frequent isolates in acute thoracic empyema. Males are 42 and females are 16 out of 58 samples from which 21 are culture positive.



Graph-1: Organisms isolated in the entire abscess combined

ESBLs are beta-lactamases that hydrolyze extended-spectrum cephalosporins with an oxyimino side chain. These cephalosporins include cefotaxime, ceftriaxone, and ceftazidime, as well as the oxyimino-monobactam aztreonam.

Table -1: MRSA & MSSA strains of Staph aureus

	MRSA	MSSA
S.AUREUS	12	18
(30/119)	(40%)	(60%)

Among staphylococcus aureus there are two vancomycin resistant MRSA strains which are resistant to linezolid.

	K.PNEUMONIA	E.COLI	P.AEKUGINUSA	P.VULGARIS	
	(23/119)	(22/119)	(19/119)	(25/119)	
AMIKACIN	17 (73.91%)	12	11 (57.89%)	13 (52%)	
		(54.54%)			
CIPROFLOXACIN	15 (65.21%)	15	12 (63.15%)	16 (64%)	
		(68.18%)			
CEFTAZIDIME	13 (56.52%)	11 (50%)	8 (42.10%)	11 (44%)	
CEFTRIAXONE	14 (60.86%)	14	8 (42.10%)	10 (40%)	
		(63.63%)			
CEFTRIAXONE/SULBACTUM	21 (91.30%)	20	17 (89.47%)	22 (88%)	
		(90.90%)			
PIPERACILLIN/TAZOBACTUM	22 (95.65%)	22 (100%)	19 (100%)	25 (100%)	
CEFTAZIDIME/CLAVULANIC	22 (95.65%)	21	18 (94.73%)	24 (96%)	
ACID		(95.45%)			
MEROPENEM	23 (100%)	22 (100%)	19 (100%)	25 (100%)	
TETRACYCLIN	18 (78.26%)	15	10 (52.63%)	18 (72%)	
		(68.18%)			

Table -2: Sensitivity and resistance patterns of gram negative bacilli

DISCUSSION

An abscess (Latin: *abscessus*) is a collection of pus that has built up within the tissue of the body. Signs and symptoms for those close to the skin include: redness, pain, warmth, and swelling that when pressed feels like it is fluid filled. The area of redness often extends beyond the swelling.

Signs and symptoms

The main symptoms and signs of an abscess are redness, heat, swelling, pain and loss of function. Abscesses may occur in any kind of solid tissue but most frequently on skin surface (where they may be superficial pustules (boils) or deep skin abscesses), in the lungs, brain, teeth, kidneys and tonsils. Major complications are spreading of the abscess material to adjacent or remote tissues and extensive regional tissue death (gangrene).

Abscesses in most parts of the body rarely heal themselves, so prompt medical attention is indicated at the first suspicion of an abscess. An abscess could potentially be fatal (although this is rare) if it compresses vital structures such as the trachea in the context of a deep neck abscess. The prevalence of multidrug-resistant gram negative bacilli, especially ESBL producers, has increased worldwide with marked regional variations of their distribution. In our study, ESBL producers were seen to the extent of 45.05% in E. coli and 52.07% in Klebsiella spp. Stephen P. Hawser et al. [18] reported 67 % and 55 % ESBL production coli and K. respectively in *E*. pneumoniae isolates in a SMART study from India.

Apart from SMART global surveillance programme, several other local reports have also described variable ESBL rates in India, for example, Aggarwal *et al.* [16] reported ESBL rates as 22 % in *E*.

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coli and *Klebsiella pneumonia* in Pune, while Sinha *et al.* [17] showed the rates as high as 64.8 % in Jaipur.

Study by Brook et al. showed that Staphylococcus aureus and Streptococcus pneumoniae accounted for ~70% of all the aerobic Gram-positive isolates. Similar findings were observed in the present study. MRSA, MSSA and Streptococcus pneumoniae (24, 8, and 4 isolates, respectively) accounted for 91.42% (32/35) of Gram-positive isolates. Gram-negative aerobic bacteria constituted ~30% (15/50)of isolates. Klebsiella the *pneumoniae* has been reported earlier as the predominant isolate in other studies.

Gerd Jórgen et al. reported streptococci, anaerobes, and S. aureus as the most common bacterial organisms. This finding was consistent with other reports in literature. Salih Bakir et al. showed that 20 (58.8%) of 34 culture positive patients had polymicrobial growth and the most common organisms anaerobic Peptostreptococcus (21.3%) were and Staphylococcus epidermidis(19.7%). However, in our study, there were 18 patients with positive cultures,most common organism cultured was Staphylococcus aureus (5/18), followed by pseudomonas aeruginosa (4/18), Proteus Vulgaris (4/18) Klebsiella pneumonia (3/18) and Escherichia coli (2/18).

The negative cultures in the remaining patients were probably because of use of antibiotics at the time cultures were sent. Worldwide, management of Deep neck Space Infections is usually early surgical drainage of purulent abscesses via an external approach. In contrast to this, Plaza Mayor *et al.* recommended broad-spectrum intravenous antibiotics along with highdoses of oral or intravenous corticosteroids for virtually all cases with Deep neck Space Infections.

Salih Bakir *et al.* in their study on 173 cases of Deep neck Space Infections showed that 78 patients (40.5%) were treated successfully with only intravenous antibiotics and in the remaining 95 patients (59.5%), surgical intervention was required. It has been identified in the literature that in patients with cellulitis or minimal abscesses, IV antibiotics alone suffice in most of Deep neck Space Infections cases.

In adults a history of trauma is usually present in most of the cases of Osteomyelitis *S aureus* is perhaps the most common organism responsible for causing acute osteomyelitis of the patella, femur and tibia. Tuberculous osteomyelitis of patella is also an entity of which one should be aware, but it often tends to be multifocal. *S. aureus* have been isolated from the tissues of patellar osteomyelitis and it was found in our case also. *Staphylococcusaureus* is the most common isolate in all types of bone infection and is implicated in 50-75% cases of chronic osteomyelitis. Alcoholism was the most common risk factor associated with Pyogenic liver abscess (PLA). Leukocytosis and raised alkaline phosphatase were the most common laboratory abnormalities raised in 22 (61.11%) and 19 (52.77%) of patients, respectively.

Age group (30 to 40) has the highest incidence in our study consisting. This is in contrast with similar works done by Mgbor *et al.* and Mukhopadhyay *et al.*, where the highest incidence was in the age group (31×40). However, Baig *et al.*, noted a change in pattern to the 5 th decade probably due to changing etiology from suppurative appendicitis to hepatobiliary causes. However, we did not see any associated hepatobiliary disease in our study.

Breast abscess is mainly caused by *Staphylococcus aureus*. There are case reports of other organisms causing breast abscess. They include *Enterococci, Salmonella tophi, Mycobacterium tuberculosis, Atypical mycobacteria, Klebsiella pneumonia* and *Fusarium solani.*

In our study of 28 cases, 21 (75%) cases were lactational and 7 (25%) cases were non-lactational breast abscesses which is comparable with the findings in the series of Schwarz *et al.* with 83% (lactational) and 17% (non-lactational). However, Elagili *et al.* reported 53.3% of non-lactational and 46.7% of lactational in his study.

The mean age of our patients was 32 years (range: 19-80 years) which is consistent with the finding of Elagili et al. and Ulitzsch et al. with the mean age of 31.93 years and 32 years in lactational and non-lactational cases. S. aureus was the most common pathogen isolated in this study next to proteus vulgaris in which is comparable with findings by Elagili et al., Ulitzsch et al. and Dixon et al. Of the 21 patients aspirated in our study, 78% resolved well without recurrence, 16% cases failed to repeated aspiration and underwent incision and drainage and 6% cases had recurrent abscess occurring 1 month after the last aspiration. Most of the urinary pathogens are also causative agents of acute and chronic prostatitis and prostatic abscess. E. coli predominates as a cause of culture positive prostatitis. Therefore, if diagnostic aspiration is done and the pathogen is established, it helps the clinician to institute appropriate treatment. S. aureus is found in 90% of the cases of primary psoas abscess followed by E. coli (3%) as demonstrated by Yago et al. In the literature, there are other reported causes of primary psoas abscess such as brucellosis, pneumococcus etc.

Alexandru *et al.* reported case of primary psoas abscess caused by β -hemolytic streptococci. Our study on Psoas abscess is similar to Yago et al. Staphylococcus aureus was isolated in 3 samples out of 4 culture positives of Psoas abscess (total of 18 pus samples) which is contrast to Alexandru *et al.* Klebsiella pneumonia was isolated from one sample out of 4.

The bacteriology of secondary psoas abscess usually reflects the underlying condition and enteric organisms (Escherichia coli, Enterobacter and Salmonella) predominate. In case of Parotid Abscess, additional patient information would be beneficial, ie., biood leucocyte count, CRP, and erythrocyte sedimentation rates together with bacterial culture results. Considering the most common pathogens involved in in parotid abscess, penicillin or cephalosporins together with anaerobic coverage would possibly be the first- line treatment.

The presence of MRSA may mandate the use of Vancomycin. However bacterial culture should be taken whenever possible and antibiotic therapy choosen in accordance to the culture results. We have taken a total of 11 samples of Parotid abscess, out of which 2 were culture positive, with one Staphylococcus aureus and another Proteus vulgaris.

CONCLUSION

In a tertiary hospital like M.G.M Hospital, Warangal, it is common to get 5 to 8 pus samples daily from different departments like General Surgery, Orthopedics, Otorhinolaryngology, and Hospital of Tuberculosis and Chest Diseases. There are nearly 40 % pus samples from Deep Seated Abscess among the total pus samples. Here patients mostly come from rural background and will have history of usage of antibiotics from a Registered Medical Practitioner or may have taken treatment from a Quack.

So it is necessary to do bacterial cultures for every pus sample to help clinicians for giving appropriate treatment to the patients. It is necessary to know the incidence of microorganisms from Community acquired and Hospital acquired infectionsand it makes mandatory to know their antibiotic susceptibility patterns.

Though pus samples were brought to the dept. of Microbiology, we have collected samples in aseptic precautions mostly from operation theatres, and from Emergency Operation Theatre where Incision & Drainage is commonly done to avoid contaminants and to avoid the delay of transport of specimens to the laboratory. This helps clinicians to choose appropriate antibiotic as early as possible and helps the patients to decrease the days of Hospital stay.

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