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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

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Abstract

Wound infection can be caused by variety of organisms like bacteria, virus, fungi and protozoa and may co-exist as poly microbial communities. Even though the bacterial profile of pus samples in many studies remain the same, the antibiotic resistance pattern of these isolates has shown a lot of variations. This study was conducted in the department of microbiology of a private tertiary care hospital CCM Medical College Durg, Chhattisgarh over a period of one year from January 2017 to December 2017. Antibiotic sensitivity testing of Staphylococcal isolates was performed by modified Kirby Bauer's disc diffusion method. Total of 387 samples were processed for aerobic culture and sensitivity, of which in 304 (78.55%) samples single growth was observed. Of the total 304 isolates, 274 isolates were from surgical units and 30 isolates from medicine units. Isolation of Gram positive and Gram negative organisms was 144 (47.36%) and 158 (51.97%) respectively and 2 (0.65%) species were Candida. Antibiotic susceptibility testing was done on 104 Staphylococcus aureus isolates for MRSA detection and antibiotic resistance pattern. 93.3% isolates were resistant to penicillin followed by cotrimoxazole (67.3%) and erythromycin (50%). Resistance to linezolid was observed in 2.9% cases while vancomycin and teicoplanin showed 1.9 % and 6.7% resistance. 26.9 % isolates were MRSA. This study gives an outline of antibiotic susceptibility of Staphylococcal isolates to formulate the local antibiotic policy to start the appropriate empirical antibiotic treatment before the culture reports are available.

Introduction

Pyogenic infections are characterized by local and systemic inflammation, formation of pus, generally caused by one of the pyogenic bacteria, which result in the aggregation of dead leukocytes as well as pyogenic bacteria commonly known as pus, "a white to yellow fluid".[1,2] Wounds are result of loss of intact skin due to injury caused by external forces such as surgical wounds, burns, bites, abrasions, minor cuts and more severe traumatic wounds such as lacerations and crush or gunshot injuries such discontinuity in skin is good environment for microbial colonization as there is presence of moisture, warmth and nutrition for their growth. Colonization with proliferation of bacterial flora may lead to wound infection which may be serious even sometimes lead to death. These may be endogenous or exogenous. A break in the skin can provide entry to the surface bacteria which thereby start multiplying locally. The body's defense mechanism includes bringing immune cells into the area to fight against bacteria. Eventually, accumulation of these cells produces pus which is a thick whitish liquid.[3]

In most of the studies it is seen that majority of the isolates are aerobes which includes Gram positive cocci like Staphylococcus aureus, Staphylococcus epidermidis, Streptococcus pyogenes, Enterococci and Gram negative bacilli like Escherichia coli, Klebsiella pneumoniae, Proteus and Pseudomonas.[4] Wound infection can be caused by variety of organisms like bacteria, virus, fungi and protozoa and may co-exist as poly microbial communities especially in wound margins and in chronic wounds[5] and in many

cases there is a mixed infection with more than one bacterial species.[6]

Even though the bacterial profile of pus samples in many studies remain the same, the antibiotic resistance pattern of these isolates has shown a lot of variations.[7,8] The inadvertent use of antibiotics leads to emergence of drug resistant pathogens, which in turn acts as a great challenge to the health services. Moreover, highly virulent strains and capacity to adapt quickly to changing environment worsens the situation and draws a matter of concern.[9]

The emergences of multi-drug resistant strains have resulted in prolonged illness, higher health care expenditures and higher risk of death due to infection. Antimicrobial resistance in addition hampers the control of infectious diseases by reducing the effectiveness of treatment thus patients remain infectious for a long time increasing the risk of spreading resistant microorganisms to others.[10,11,12,13]

During the last few decades, multidrug-resistant Gram-negative bacterial strains such as Acinetobacter baumannii, Esherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, and Gram-positive methicillin-resistant Staphylococcus aureus(MRSA) were increasingly associated with pus infections under hospital settings due to extensive misprescription and inadequate dose regimen of antibiotics.[10,13]

Objectives of Study

To study Bacteriological profile of pus /wound swab samples and antibiotic susceptibility pattern of Staphylococcus aureus isolated from the pus.

Material and Methods

Sample collection

A study was conducted in the department of microbiology of a private tertiary care hospital Chhattisgarh, India. A Total of 304 samples were collected by sterile syringe aspiration (n=297) and by sterile swab (n=7) from inpatients of CCM medical college Kachandur Durg, which were received from various clinical department of the hospital over a period of one year from January 2017 to December 2017, and were processed for aerobic culture and sensitivity. Only age and sex of the patients was considered for study purpose.

Pus samples were collected using sterile cotton swabs placed in screw capped tubes and pus aspirates were collected by using sterile disposable syringes and were immediately sent to the microbiology laboratory. All samples were processed aerobically on Blood Agar, MacConkeys agar, Nutrient agar and incubated aerobically at 37°C for 24 hours. Identification of isolates from positive culture was done as per standard tests for identification, by Gram staining, motility and biochemical tests like catalase, coagulase, indole, methyl red, Voges- Proskauer, citrate, urease, phenyl pyruvic acid test and oxidase test[6]

Antibiotic sensitivity testing

Antibiotic sensitivity testing of Staphylococcal isolates was performed by modified Kirby Bauer's disc diffusion method on Mueller Hinton agar using antibiotics as per CLSI guidelines. Standard antibiotics like, Cefazoline(30mcg), penicillin (2 U), cotrimoxazole (25mcg) , cefoxitin (30mcg), clindamycin (2mcg), , erythromycin (15mcg), linezolid (30mcg), teicoplanin (30mcg), and vancomycin (30mcg) were tested. All the culture media, biochemical media and antibiotic discs used were obtained from Hi Media.

Analysis

Analysis was done by using sort, filter and percentages applications in MS Excel, 2013 version.

Results

Total 387 samples were received in the microbiology department and were processed for aerobic culture and sensitivity, out of which in 304 (78.55%) samples single growth was observed.

Among 304 culture positive samples, isolation of Gram positive and Gram negative organisms was 144 (47.36%) and 158 (51.97%) respectively and 2 (0.65%) species were Candida.

Table1: Gram positive isolates

Coagulase Negative Staphylococci (CoNS)	4
Enterococcus species	27
Staphylococcus aureus	104
Streptococcus species	7
Streptococcus pyogens	2
Total	144

Table 2: Gram negative isolates

Acinetobacter species	3
Citrobacter koseri	2
Citrobacter species	3
Escherichia coli	70
Klebsiella pneumoniae	2

Klebsiella species	21
Proteus mirabilis	10
Proteus species	1
Proteus vulgaris	6
Pseudomonas aeruginosa	28
Pseudomonas species	12
Total	158

Of the total 304 isolates, 274 isolates were from surgical units and 30 isolates from medicine units.

Table 3: Unit wise isolates

Surgical ward	274	90.13%
Medical ward	30	09.86%
Total	304	

Table 4: ward wise (Department wise) isolates

DERMATOLOGY	5	1.64 %
ENT	11	3.62%
GEN MEDICINE	19	6.25%
GEN SURGERY	207	68.09%
OBGY	25	8.22%
ORTHOPEDICS	31	10.20%
PEADIATRICS	3	0.99%
TB & CHEST	3	0.99%
	304	

TABLE 5: Age and sex wise distribution of isolates

Age group (in years)	0 to 5	5 to 15	16 to 25	26 to 35	> 35	Total	%
Male	2	10	30	35	104	181	59.53
female	2	4	24	13	80	123	40.46

TABLE 6: Antibiotic resistance pattern in Staphylococcus aureus

Resistogram	Antibiotics	Resistant strain	%
Staphylococcus aureus (n=104)	Cefazoline	53	51.0
	Cotrimoxazole	70	67.3
	Penicillin	97	93.3
	Erythromycin	52	50.0
	Cefoxitin	28	26.9
	Linezolid	3	2.9
	Clindamycin	13	12.5
	Vancomycin	2	1.9
	Teicoplanin	7	6.7

Screening of MRSA (Methicillin resistant Staphylococcus aureus)
Cefoxitin 30 µg disc was used as surrogate marker for MRSA detection. Zone of inhibition < 21 mm is considered as Methicillin resistant.

Discussion

Staphylococcus aureus is one of the most prominent organisms causing skin or soft tissue abscesses. In addition to form pus it is also responsible for the pneumonia, toxic-shock syndrome, exfoliative skin disease, and enteritis. S. aureus colonize the human skin, nails, and nares and disseminate via physical contact and aerosols. Staphylococcal invasion is generally caused due to breaches in local defense mechanism, like skin cuts or hair follicle trauma, which manifests as abscess formation and severe inflammation of surrounding tissues.[15] Improper use of antibiotics over the past few decades has led to drug-resistant strains, designated MRSA (methicillin-resistant S. aureus).[16] Vancomycin has been used as the last-resort antibiotic for MRSA infections, but use of vancomycin also lead to the vancomycin resistant (VRSA) .

In our study out of 387 samples 304 (78.55%) were culture positive, this positivity rate was similar with the study by

Nithya et al.[17] However, lower rate was also reported by Shrestha et al. (50%).[18]

In our study Gram positive and Gram negative organisms isolated were 47.36% and 51.97% respectively. There was a preponderance of Gram negative organisms observed in our study. This was in accordance with the study by Nithya et al.[17] while Bhatta et al.[19] & Shrestha et al.[18]reported Staphylococcus aureus to be the most prevalent bacteria isolated from the cases of wound infections.

90.13% isolates were from the surgical units while only 9.86% isolates were from the medicine wards, these results were in accordance with the study by Asmabegaum Biradar et al.[4] Also in our study maximum isolates were from the age group of >35 years similar results were observed by Asmabegaum Biradar et al.,[4] while Salurai et al. observed maximum isolates from the < 1 year age group.[20]

Different studies have been performed to assess the bacterial profile and the antibiotic susceptibility pattern in pus samples. This is particularly relevant for the treating physician who needs to start empirical treatment of patient until the lab culture reports are awaited.[21]

We have performed the antibiotic susceptibility testing on Staphylococcus aureus to see the MRSA and resistance pattern to start the empirical treatment before the culture reports are available.

We observed that maximum (93.3%) isolates were resistant to penicillin, followed by Cotrimoxazole (67.3%) and erythromycin (50%).

Resistance to Linezolid was observed in 2.9% cases while vancomycin and Teicoplanin showed 1.9 % and 6.7% resistance.

In our study it was observed that 26.9 % isolates were MRSA which was similar with the MRSA rate observed by Zulfiqar A Naqvi et al.[22]

The prevalence of MRSA in our study was higher than the study by Subedi and Brahmadathan,[23] while higher rates of MRSA were shown by Pushpalatha et al.[24]

Difference in the isolation of MRSA may be due to differences in the empirical treatment or lack of culture facilities in the remote areas.It is of great concern that in our study some strains showed resistance to linezolid and vancomycin, so these drugs should be used very cautiously to prevent emergence of antibiotic resistance.

Conclusion

This study gives an outline of antibiotic susceptibility of Staphylococcal isolates which will help in formulating the local antibiotic policy for the hospital and to start the appropriate empirical antibiotic treatment before the culture reports are available. However more studies from the area are required to formulate the policy. The present study provides one-time information about the antibiotic susceptibility of Staphylococcus aureus, and the periodic review of the bacteriological profile and antibiotic sensitivity pattern is highly essential.

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