PREVALENCE AND ANTIMICROBIAL SUSCEPTIBILITY OF METHICILLIN RESISTANT STAPHYLOCOCCUS AUREUS AND COAGULASE-NEGATIVE STAPHYLOCOCCI IN A TERTIARY CARE HOSPITAL

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ABSTRACT

Background: Globally nosocomial infection is a major problem. Prevalence and antibiotic resistance of methicillin-resistant Staphylococcus aureus (MRSA) strains is reported to be increasing globally. MRSA and methicillin resistant coagulase negative staphylococci (MRCoNS) are the important agents causing nosocomial infections.

Objective: The study was conducted to find out the prevalence rate of MRSA and MRCoNS and antibiotic susceptibility pattern.

Materials and Methods: This was a retrospective study conducted from June 2011 to November 2012 in a tertiary care hospital in south India. All isolates were identified by Clinical and Laboratory Standards Institute (CLSI) guidelines and antibiotic susceptibility pattern determined by modified Kirby Bauer disc diffusion method. The information was recorded and analyzed using Microsoft Excel (2007 version).

Results: A total of 210 Staphylococcus strains were isolated from various clinical samples, 180 were coagulase positive staphylococcus (CoPS) and 30 were coagulase negative staphylococcus (CoNS). Among 180 CoPS, 58 (32.22%) were Methicillin resistant and among CoNS, 12 (40%) were methicillin resistant. In MRSA maximum resistance was seen with oxacillin (93.2%) and least with vancomycin (3.5%). In MRCoNS maximum resistance was seen with oxacillin (91.7%) and least with vancomycin (0%).

Conclusion: There is need for continuous monitoring of the antimicrobial susceptibility pattern of methicillin staphylococcus aureus and methicillin resistant coagulase negative staphylococci for the selection of appropriate therapy, developing the antibiotic policy and for limiting the use of powerful antibiotics.

Keywords: Methicillin-resistant Staphylococcus aureus (MRSA), Methicillin resistant coagulase negative staphylococci, Vancomycin, Nosocomial infection, Susceptibility pattern

INTRODUCTION

Staphylococcus aureus (S.aureus) is one of the most important pathogens affecting humans, has acquired resistance to various antibiotics and is a leading cause of hospital and community acquired infections, manifesting from minor skin diseases to life-threatening infections [1,2]. Methicillin resistant Staphylococcus aureus (MRSA) was first described in 1961, reported after one year of introduction of methicillin and has emerged as one of the most important nosocomial pathogens specially in the last two decades [3].

MRSA is now endemic in India. The incidence of MRSA varies according to the region, 25% in western part of India [4] to 50% in South India [5]. MRSA is of serious concern not due its sole resistance to methicillin, but also because of resistance to many other antimicrobials that are used on a regular basis in hospitals. Current therapeutic options for MRSA are limited few expensive drugs like vancomycin, linezolid, teicoplanin, daptomycin and streptogramins. Another alarming sign is that emergence of resistance to Vancomycin, although at a low level has been reported [6]. Glycopeptides and linezolid continue to remain the mainstay of treatment for MRSA.

Both endemic and epidemic MRSA infections occur globally as infected and colonized patients in hospitals mediate the dissemination of these isolates and hospital staff assists further transmission [7]. Although many studies have been done on prevalence and antibiogram of staphylococcus, but many of these studies have concentrated only on methicillin resistant staphylococcus aureus and not on methicillin resistant coagulase negative staphylococci (MRCoNS) which are equally important. This study has been carried to determine the prevalence and antibiotic

susceptibility of staphylococcus aureus and MRCoNS in order to utilize the information to formulate antibiotic policy and appropriate control measures.

MATERIAL AND METHODS

This was a retrospective study done in a tertiary care hospital in south India from June 2011 November 2012. The records were taken from the Microbiology department. Staphylococcus strains were identified based on Gram's stain morphology, colony characteristics, and positive catalase and coagulase tests. All isolates were identified as S. aureus according to standard methods [8].

These were isolated from inpatients, the various clinical samples included were pus, sputum, urine, blood, cervical swab and catheter tip. Testing for methicillin resistance was performed using the cefoxitin disc diffusion method recommended by the Clinical and Laboratory Standard Institute [8]. The isolates were considered methicillin resistant if zone of inhibition was 10 mm or less.

Antibiogram was performed by modified Kirby Bauer Disc Diffusion method as per CLSI Standards against the following antibiotics-Amikacin (30µg), Vancomycin (30µg), Gentamicin (10µg), Ampicillin (10µg), Cefuroxime (30µg), Cefotaxime (3µg), Chloramphenicol (30 µg), Erythromycin (15µg), Oxacillin (1.0µg), Co-trimoxazole (1.25/23.75 µg), Clindamycin (2µg) and Norfloxcin(10µg).

Staphylococcus aureus ATCC 25923 was used as the control strain. The data obtained was recorded on Microsoft excel (2007 version) and analyzed. The results are explained in frequency (number) and in percentage (%).

RESULTS

The total isolates collected in the present study were 210, out of which 180(85.7%) were coagulase positive staphylococci and 30 isolates (14.3%) were coagulase negative staphylococci. Out of 180 coagulase positive staphylococci, 58(32.2%) were methicillin resistant. Among 30 isolates of coagulase negative staphylococci, 12 (40%) were methicillin resistant.

The distribution of MRSA among various clinical samples is shown in table 1.

Table 1: Distribution of MRSA and MSSA from different clinical samples

Clinical Number	Total Percentage	MRSA	samples specimens
Pus	52	16	27.5
Urine	36	10	17.2
Cervical swab	21	9	15.2
Blood	22	8	13.7
Sputum	17	6	10.3
Catheter tip	20	5	8.6
Other	13	4	6.8
Total	**180**	**58**	**32.22**

MRSA = Methicillin resistant Staphylococcus aureus

Highest numbers of samples were obtained from pus and least from sputum

The antibiotic susceptibility pattern of MRSA and MSSA is shown in table 2.

Table 2: Antibiogram of MRSA and MSSA.

Antibiotic	Sensitive Staphylococcus aureus isolates			
	MRSA isolates (n=58)		MSSA isolates (n=122)	
	Number	Percentage	Number	Percentage
Amikacin	54	93.1	115	94.2
Vancomycin	56	96.5	122	100
Gentamicin	19	32.7	112	91.8
Ampicillin	08	13.7	42	34.4
Cefuroxime	36	62	93	76.2
Cefotaxime	16	27.5	59	48.3
Chloram phenicol	19	32.7	62	50.8
Erythromycin	19	32.7	101	82.8
Oxacillin	04	6.8	13	10.6
Co – trimo xazole	18	31	109	89.3
Clindamycin	17	29.3	102	83.6
Norfloxacin	12	20.6	40	32.7

MRSA = Methicillin resistant Staphylococcus aureus
MSSA = Methicillin sensitive Staphylococcus aureus

The antibiotic susceptibility pattern of methicillin resistant coagulase negative staphylococcus (MRCoNS) is shown in table 3.

Table 3: Antibiogram of methicillin resistant coagulase negative staphylococcus (MRCoNS)

Antibiotic	MRCoNS Isolates sensitive (n=12)	
	Number	Percentage
Amikacin	9	75
Vancomycin	0	100
Gentamicin	3	25
Ampicillin	2	16.6
Cefuroxime	7	58.3
Cefotaxime	5	41.6
Chloramphenicol	4	33.3
Erythromycin	5	41.6
Oxacillin	1	8.3
Co – trimoxazole	4	33.3
Clindamycin	3	25
Norfloxacin	6	50

Highest resistance was seen with oxacillin and least with vancomycin

DISCUSSION

There is a growing concern about the rapid rise in resistance of S. aureus to antimicrobial agents [9]. In India, the importance of MRSA as a problem has been recognized relatively late [10]. The prevalence of MRSA varies in different parts of India and is not uniform. Reports from a Delhi hospital showed a prevalence rate of 51.6% in 2001, whereas it was reported as 38.44% in the same hospital in 2008 [11].

A recent study [12] found the prevalence to be 42% in 2008 and 40% in 2009. In a study at Aligarh, India [13] it was shown that 35.1% of *S.* aureus and 22.5% of coagulates-negative staphylococcal isolates were resistant to methicillin. In another study [10] conducted in Tamilnadu, out of 906 strains of *S.* aureus isolated from clinical samples, 250 (31.1%) were found to be methicillin resistant. Our study had MRSA prevalence of 32.22%. This variation in prevalence may be because of several factors like healthcare facilities available in the particular hospital, implementation and monitoring of infection control committee, rationale antibiotic usage which varies from hospital to hospital.

Maximum samples of the MRSA isolated were from the pus samples i.e. 16(27.5%) followed by urine, cervical swab and blood samples as shown in table 1. This is in accordance with other studies [14,15] and also different with other studies where throat swabs and wound swabs were the main source [10]. This difference may be because of the same reasons stated above.

MRSA strains were more resistant to all antibiotics than MSSA strains except for amikacin and vancomycin. The present study shows high resistance to ampicillin, oxacillin, cefotaxime, and clindamycin. This is in accordance with other studies [1,15,16]. MSSA isolates show higher sensitivity than MRSA strains to gentamicin (32.7% vs 91.8%), erythromycin (32.7% vs 82.8%), co trimoxazole (31% vs 89.3%) and clindamycin (29.3% vs 83.6%). Resistant to quinolones (norfloxacin) was high (80%) in the present study. In a recent study [17] the resistant rate was also high (87.5%). But a previous study [18] conducted in 2003, reported the resistant rate of ciprofloxacin to be only 32.6%. The rapid emergence of quinolones is probably due to the indiscriminate empirical use of these drugs.

Another concern is of vancomycin resistance, our study reported 3.5% resistance to vancomycin among MRSA. Although this is low when compared to a recent study [19], but nevertheless is an alarming sign. Vancomycin-intermediate and vancomycin resistant S. aureus strains have been reported recently from various parts of the country [6, 20, 21]. Regular monitoring of vancomycin sensitivity and routine testing of other newer glycopeptides like teicoplanin should be done to prevent rapid emergence of resistance.

Most common reason for multi drug resistant MRSA is indiscriminate use of antibiotics without drug sensitivity testing which may be due to due to lack of advanced laboratory facilities or negligence on the part of medical practitioners or patients poor economic status. There is a difference between antibiogram of MRSA and MSSA isolates and routine testing of methicillin resistance should be done using cefoxitin disc which at present is the most sensitive method.

Coagulase-negative staphylococcus (CoNS) is a group of opportunistic pathogens causing wide spectrum of diseases in humans. Recently MRCoNS have been associated with increased number of infections in hospitalized patients [22-24].

In the present study out of 30 CoNS, 12 (40%) were MRCoNS. The results were comparable to other studies [25, 26], which reported a

resistance of 48% and 43.8%. Maximum resistance was seen with ampicillin, oxacillin and gentamicin (table 3). Similar results were obtained in the previous study [25]. No resistance was seen with vancomycin. Resistance was also seen with Cefuroxime (41.7%) and Cefotaxime (58.4%) and erythromycin (58.4%). Gentamicin is a most commonly used drug, because of its low cost and synergistic activity with beta lactum antibiotics. In the present study 75% resistance was seen with MRCoNS, which is slightly higher than the previous study [26].

After exposure to multiple antibiotics, surgical prophylaxis, indiscriminate use of antibiotics patients become colonized with multi-drug resistant strains of CoNS species such as Staphylococcus epidermidis. This has led to use of glycopeptides in high risk patients and thereby its low level resistance in hospital strains [27]. There are about 33 CoNS species, but only a few of them have been associated with an increase in hospital acquired infections.

In a study done among catheter related blood stream infections, 96% were Staphylococcus epidermidis [28]. In another study, 51% were Staphylococcus epidermidis, 18% were Staphylococcus haemolyticus, 16% Staphylococcus saprophyticus [29]. Ideally we should have identified the individual strains of CoNS and reported the resistance to individual species of CoNS. Multiple resistances to antibiotics has been reported in many CoNS isolates, making infections they cause difficult to treat [30-32]. Although there are many studies done on prevalence of MRSA, but these studies have concentrated only on S.aureus and not on CoNS which are also important in the present scenario of staphylococcus strains developing multi drug resistance.

Limitations of the study

The limitation of the present study is that it was a retrospective study, and the sample size was small. Another limitation is, we did not report the individual CoNS species. Future studies should be done with large sample size and identify the individual CoNS species to know the prevalence and changing trends of antibiogram of MRSA and MRCoNS.

CONCLUSION

The present study showed a high level prevalence of MRSA and MRCoNS strains resistance against widely used antimicrobial agents. The regular surveillance of MRSA and MRCoNS will also be useful for selecting an appropriate antibiotic, to know the changing trends of antibiotic susceptibility pattern, for developing hospital antibiotic policy and for limiting the use of powerful antibiotics like Vancomycin as initial treatment and save it for the treatment of resistant and life-threatening staphylococcal infections.

Conflict of interest: None

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