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STUDY OF PREVALENCE AND SUSCEPTIBILITY PATTERN OF METHICILLIN-RESISTANT *STAPHYLOCOCCUS AUREUS* (MRSA) AT SREE GOKULAM MEDICAL COLLEGE, TRIVANDRUM.

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ABSTRACT

Background

Methicillin-resistant *Staphylococcus aureus* (MRSA) one of the most widespread causes of nosocomial infection's worldwide. Recently, they have been recovered from community. This study was undertaken to analyze the prevalence of methicillin resistance among isolates at Sree Gokulam Medical College,Trivandrum, and document the current resistance profile of MRSA and methicillin-sensitive*Staphylococcus aureus* (MSSA) to the commonly used anti-staphylococcal agents.

Methods

Over a 2-year period we analyzed 1215 isolates of *S. aureus* strains recovered from various clinical sources, from hospital and community practices. Antimicrobial susceptibility testing was done according to CLSI guidelines.

Results

The prevalence of MRSA from surgical/burn wounds/pus, urine and miscellaneous others were 60.1%, 15.5% and 6.6%, respectively. The major sources of MSSA were surgical/burn wounds, pus/abscess and upper respiratory tract specimens with rates of 32.9%, 17.1% and 14.3%, respectively. The greatest prevalence of resistance of MRSA along with beta lactams was seen for erythromycin (86.7%), and clindamycin (70%). Resistance rates among MSSA were highest for ampicillin (70%).

Conclusion

The prevalence of MRSA in the hospital increased from 12.5% in 2011 to 20.8% in 2013. Most isolates were associated with infected surgical/burn wounds which may have become infected via the hands of health care workers during dressing exercises. Infection control measures aimed at the proper hand hygiene procedures may interrupt the spread of MRSA. health care workers may also be carriers of MRSA in their anterior nares. Surveillance cultures of both patients and health care workers may help to identify carriers who would be offered antibiotics to eradicate the organisms. Most MRSA are resistant to several non-β-lactam antibiotics. Frequent monitoring of susceptibility patterns of MRSA and the formulation of a definite antibiotic policy may be helpful in decreasing the incidence of MRSA infection.

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INTRODUCTION

Staphylococcus aureus is and always will be one of the most frequently isolated pathogens in both community and hospital practices. It is the common bacterial agent recovered from blood stream infections, skin and soft tissue infections, pneumonia and hospital-acquired post-operative wound infections [1-4]. Complications involved are bacteremic seedings of several organs, causing endocarditis, osteomyelitis and septic arthritis[5,6]. Changes in the pattern of antimicrobial susceptibility of *S. aureus* and other organisms have been reported worldwide, especially in developing countries [7-9], making antimicrobial agents increasingly less effective in treating bacterial infections. There have been dramatic changes in the susceptibility of *S. aureus* in both hospitals and community settings in India. [10-12]. The older β-lactams, penicillin and ampicillin are ineffective against more than 80 % of isolated strains, and resistance to many of the non-β-lactam agents such as the tetracyclines, gentamicin, , erythromycin and clindamycin has gradually increased and reached alarming levels by the 1990s in many parts of the world [13-15]. Several mechanisms for the methicillin resistance seen in *S. aureus* have been elucidated. The most important is the production of a unique penicillin-binding protein (PBP) that has a low affinity for β-lactam antibiotics and whose effects are determined by several structural genes (*mec, mec RI, mec I*) [16,17]. Other known mechanisms of methicillin resistance are the production of the usual PBPs, but with modified affinities for the β-lactam drugs, and the hyper production of penicillinase enzyme [17,18]. Spread of Methicillin-resistant *Staphylococcus aureus* (MRSA) and methicillin-sensitive *Staphylococcus aureus (MSSA)* from infected patients to medical staffs are often become transient carriers [19]. MRSA, are usually also resistant to other non-β-lactam antibiotics, infections with them are life-threatening in immune compromised patients, often difficult to manage, and problematic to eradicate. The primary importance is to decrease the prevalence of MRSA by measures like rapid and reliable identification of the organisms along with their susceptibility patterns to other antibiotics, isolation and treatment of patients and carriers, and strict adherence to proper hand washing practices by health care providers. The purpose of this study was to review and document: (1) the current prevalence of methicillin resistance among isolates of *S. aureus* recovered from hospital and community sources and (2), the pattern of antimicrobial susceptibilities of MSSA and MRSA isolates to the commonly prescribed antibiotics in Sree Gokulam Medical College, Trivandrum.

METHODS

Between May 2011 to May 2013, specimens were derived from patients on the wards and from those attending outpatients' clinics at the Sree Gokulam Medical College, Trivandrum. No differentiation was between isolates from inpatients and outpatients, all being classified as "hospital practice" isolates. *S. aureus* recovered from patients attending health centers in the community and those seen by general practitioners were classified as "community practice" isolates. For purposes of gathering infection control surveillance data, other organisms and duplicates of clinically significant isolates were excluded from the study. *S. aureus* isolates were identified via colonial morphology on blood agar plates supplemented with 5% sheep blood, gram stain characteristics, catalase test, tube coagulase test, and DNAse test. Antimicrobial susceptibility testing was done on Mueller-Hinton agar using the disc diffusion technique as outlined by CLSI [20]. The following drugs and concentrations (in brackets) were used to determine the antibiogram of the strains: penicillin (10 µg), gentamicin (10 µg), erythromycin (15 µg), tetracycline (30 µg), co-trimoxazole (trimethoprim-sulfamethoxazole) (1.25/23.75 µg), amikacin (10 ug)ciprofloxacin 5 µg), clindamycin (2 µg), linezolid (30 ug) and vancomycin (30 µg). Methicillin-resistance was tested using a 30 µg Cefoxitin disc. Zone diameters were read after incubation at 35°C for

a full 24 hrs. Strains with zone sizes of < 23 mm for Cefoxitin was regarded as methicillin resistant. ATCC *S. aureus* strains 25923 and 29213 were used as quality control. Statistical analysis of data was done using SPSS software.

RESULTS

During the two-year study period, a total of 1215 *S. aureus* isolates were recorded. The major sources of *S. aureus* were from pus/abscess, which, accounted for 903 (74.3%) of all isolates (Table 1). Fifty nine *S. aureus strains* (4.5%) were from patients with pneumonia, 6 (1.1%) from the blood of patients with sepsis. The rest of the isolates were from vaginal, ear, CNS and miscellaneous infections, each accounting for <6.0% of the total. The major source of MRSA was pus/burns/surgical wounds (60.1%) followed by urine (15.5%), and sputum samples. MSSA was recovered more frequently from surgical/burn wounds/pus (50%), and the upper respiratory tract (14.3%).

Table 1
Clinical sources 1215 strains of Staphylococcus aureus isolated at Sree Gokulam Medical College, Trivandrum

	MRSA		MSSA	
Source/Specimen	N	%	N	%
Pus/abscess	354	78.7	542	70.9
Urine	49	9.3	62	8.9
Sputum	13	2.4	46	6.6
Throat swab	20	3.8	27	3.9
Blood	6	1.1	9	1.2
°Miscellaneous	23	4.3	57	8.2
Total	450	100	765	100

The antimicrobial susceptibility patterns of MSSA isolates are shown in Table 2. Throughout the study period resistance to the older β-lactam antibiotics increased significantly ($p < 0.001$) in both hospital and community practices. Resistance patterns of erythromycin, ampicillin, gentamicin and clindamycin were higher among hospital isolates when compared to community isolates, but these differences (except for gentamicin) were not statistically significant. The reverse situation was seen among community strains versus hospital strains where resistance rates were considerably higher for ciprofloxacin.

Table
Percentage of 693 [a]MSSA strains resistant to various antimicrobial agents at the Sree Gokulam Medical college

Antimicrobial	Hospital Practice (n = 556)	Community Practice (n = 137)	Significance (P-value)
Ampicillin	444 (80)	93 (68.2)	< 0.001
Tetracycline	237(42.7)	101 (73.5)	< 0.001
Erythromycin	80 (14.4)	4 (6.1)	NS
Clindamycin	58 (10.5)	11 (8.3)	NS
[1]Co-trimoxazole	56 (10.2)	5 (4.3)	NS
Gentamicin	108(19.5)	1 (1.2)	< 0.001
Ciprofloxacin	11 (2.0)	10(7.5)	NS
Cephalexin	0	0	—
Vancomycin	0	0	—
Amoxyclav	0	0	-
Amikacin	0	0	-

All MRSA isolates were fully sensitive to vancomycin, linezolid and teicoplanin, while the greatest prevalence of resistance was seen for Pencillin/Cephalexin/Amoxyclav/Cloxacallin where according to the label it was 100%, this was followed by erythromycin (86.7%) followed by tetracycline (78.7%),clindamycin (70%), and gentamicin (67%).

Table 3
Distribution of resistance pattern of MRSA isolates

Antibiotics	Present study
Pencillin/Cephalexin/Amoxyclav/Cloxacallin	100
Erythromycin	86.7
Tetracycline	78.7
Clindamycin	70
Gentamicin	67
Co-trimoxazole	53
Amikacin	5
Rifampacin	3
Vancomycin	0
Teicoplanin	0
Linezolid	0
	0

Table 4
Distribution of 1215 Staphylococcus aureus strains isolated from hospital practice and community practice at the Sree Gokulam Medical College, Trivandrum

Methicillin	No of hospital isolates	No of community isolates	Total
Sensitive	628	137	765
Resistant	410	40	450
Total	1048	177	1215

DISCUSSION

The present study has shown a steady increase in the prevalence rate of MRSA isolation (37%) over the previous studies of 29% done in other studies conducted in South India. Similar patterns have been seen worldwide as evident from the many recorded surveillance studies [3,4,18]. Despite this observation however; there are considerable differences

between individual countries and also from different states within a country [22]. The highest rates of methicillin resistance among *S. aureus* were observed among isolates from the western Pacific region [15]. In Korea, the rate was 64 % [22], in a major Taiwanese hospital resistance rate was found to be about 77% [23]. In North America [4,24,25], the Middle East [7,9,13,26], India [27] and the Canary Islands [28] rates of 38 – 50%, 33–40%, 18 – 43% and 25% respectively, have been reported. MRSA rates from Trivandrum, Kerala are scanty, but data from a previous report have shown rates of isolation to be < 10%. From South America [4,32-34] and Europe [18,35,36], rates of 10 – 66% and 25 – 58% respectively, have been seen. These different rates among MRSAs from different countries may be attributed to variations in patient populations, the biological characteristics of the *S. aureus* strains, and/or infection control practices. Although MRSA remains largely a nosocomial pathogen, 8.1% of isolates were community-acquired. Several factors may be at play in the increasing prevalence of community-acquired MRSA. One reason for this increase maybe lateral dissemination of MRSA from hospital to the community from discharged patients diagnosed with MRSA, and the discontinuation of therapy because of the high cost of prescribed drugs at local pharmacies. These strains of MRSA are frequently multi-resistant. Two reports from Australia suggests that while lateral dissemination from hospital to community may be a major factor, strains of MRSA isolated from people in a remote western area of the country shows no similarity to hospital-derived strains. These people have had little or no contact with urban cities, health care facilities or health care providers, and these MRSA are generally non-multi-resistant [34,35]. Although molecular techniques were not applied in this study, the multi-resistance as seen among community strains would suggest that these strains may have originated from the hospital.

Although a protocol exists for the management of infected patients and colonized health care providers (HCPs), it is rarely implemented. The protocol requires the physical separation of infected patients and their HCPs from uninfected patients, therapy for eradicating MRSA, stringent environmental disinfection of areas harboring MRSA and the application of barrier isolation precaution measures, including strictly enforced hand hygiene to interrupt spread patterns. A previous report from this country revealed that proper hand washing practices and sanitation techniques of HCPs at all levels of service were not strictly adhered to [36]. The situation is further complicated by the fact that HCPs frequently complain of overcrowded wards, scarce material resources and overworked personnel. MRSA is a well known cause of hospital acquired infection and in India, Community Acquired-MRSA strain is now an emerging pathogen.This fact should be borne in mind when standard regimens fail, although the existing data do not justify empirical use of anti MRSA drugs in treatment.[36] Magnitude of surgical wound infection problem may be increasing because many of the causative organisms have started to develop some form of drug resistance to currently used antibiotics. It is essential to give special attention in reducing surgical site infections.[37]

CONCLUSION

The study has shown that the prevalence of MRSA infections over the past 2 years to be high in comparison to studies done earlier. The principal source may have been the hands of health care professionals during wound dressing exercises. Infection control measures such as proper hand hygiene and surveillance cultures may help in arresting the spread of MRSA in the hospital setting. An antibiotic policy and the monitoring of susceptibility patterns of MRSA may also help in decreasing the prevalence of MRSA and antibiotic resistance.

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