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Research Article

METHICILLIN-RESISTANT STAPHYLOCOCCI PREVALENCE IN CANCER PATIENTS AT A TERTIARY CARE CANCER CENTRE – A RETROSPECTIVE STUDY

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ABSTRACT

Objective: The objective of this study was to investigate the prevalence of Methicillin-resistant *Staphylococcus aureus* (MRSA) and MR-coagulase-negative staphylococci (CoNS), as well as their antimicrobial resistance, in various samples from cancer patients in North Kerala.

Methods: The retrospective study was conducted at a tertiary care cancer centre in North Kerala over a 4-year period from January 2016 to December 2019. During the study, data on all cultures from cancer patients was analyzed. This study was approved by Institutional Review Board (IRB). Non-duplicate isolates of staphylococci were included in the study obtained from various clinical specimens. Species identification and antimicrobial susceptibility testing was done using automated methods.

Results: During the period of 4 years (2016–2019), a total of 1176 isolates of staphylococci were analyzed, out of which 784 were *S. aureus* isolates (68%) and 392 (32%) isolates were CoNS. Among CoNS species, *Staphylococcus epidermidis* and *Staphylococcus haemolyticus* were the most common species of CoNS, representing 39% and 28% of the total CoNS identified. Overall prevalence of methicillin resistance in *S. aureus* and CoNS was found to be 50.7% and 55.6%, respectively. Methicillin-resistant staphylococci isolates showed higher resistance to multiple drugs than methicillin-sensitive staphylococci isolates.

Conclusion: This study demonstrates that MRS could also be a haul in cancer patients at North Kerala. A higher percentage of MR-CoNS isolates are multidrug resistant than MRSA isolates. Glycopeptides and linezolid still stay the mainstay for treatment for MRS infections.

Keywords: Methicillin-resistant staphylococci, Prevalence, Antibiotic resistance, Cancer patients.

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INTRODUCTION

Infections are common among cancer patients, lengthening hospital stays, and increasing health-care costs. Nosocomial infections account for the majority of infections in cancer patients [1,2]. Staphylococcus is a genus of bacteria that causes a wide range of infectious diseases in humans. It is divided into two types: *S. aureus* and coagulase-negative staphylococci (CoNS). CoNS are opportunistic pathogens that are generally associated with infections in patients that have medical devices or who are immunocompromised [3-6]. Among the CoNS, *Staphylococcus epidermidis* and *Staphylococcus haemolyticus* are much more virulent than the other CoNS species [4-6].

Methicillin-resistant staphylococci (MRS) are increasingly being classified as multidrug-resistant (MDR) due to their high resistance to a wide range of commonly used antimicrobials such as erythromycin, clindamycin, aminoglycosides, fluoroquinolones, and antibiotic medication, leaving few effective options. MDR among CoNS is on the rise, posing a significant challenge in the management of hospital-acquired infections and acting as a reservoir of antibiotic resistance [5,7].

There is no information available on the prevalence of Methicillin-resistant *S. aureus* (MRSA) and Methicillin-resistant CoNS (MR-CoNS) in cancer patients in Southern India. As a result, the current research was undertaken to investigate the prevalence of MRSA and MR-CoNS, as well as their antimicrobial resistance, in various samples from cancer patients in North Kerala.

METHODS

This retrospective study was conducted at a tertiary care cancer center at North Kerala, India. January 2016–December 2019 data on all microbial cultures from various clinical samples from cancer patients were analyzed during the period – February 2020–May 2020. This study was approved by Institutional Review Board (IRB) [1616/IRB-SRC/13/MCC/14-03-2020/4].

The study included only non-duplicate isolates of *S. aureus* and CoNS (clinically significant) was included in the study. The isolates were considered to be of clinical significance only when the following criteria were fulfilled: (1) Strains obtained in bacterial pure culture; (2) infection risk factors (immunosuppressive); (3) clinical symptoms of infection (hyperthermia >38°C, hypotension, tachycardia, tachypnea); and (4) infection site (respiratory, intestinal, joints, skin, and soft tissues).

These staphylococci species were isolated from varied clinical samples such as pus, sputum, urine, blood, and body fluids. Blood agar, MacConkey agar, and chocolate agar plates were used to culture the clinical specimens, which were incubated aerobically at 37°C for 24–48 h. Bactec 9050 (BD BactecTM 9050 Blood Culture System) and BacT/ALERT 3D (Biomerieux, USA) were used to process blood cultures according to the manufacturer's instructions.

Bacterial identification and Antibiotic susceptibility testing (AST)

After the initial colony isolation from the culture plates, these colonies were then subjected for species identification by automated identification system (Phoenix and Vitek 2 automated system) and from 2019 onward all the staphylococci were identified using Matrix-assisted laser desorption ionization time flight mass-spectroscopy (MALDI-TOF-MS - Biomerieux, France). After identification of staphylococcal species, antibiotic susceptibility was performed using automated system. These colonies were taken from the plate and suspension was made

and compared with the MacFarland standard before inoculating into the AST panel kits. AST was performed using BD PhoenixTM (Becton Dickinson, USA) instrument from January 2016 to October 2017 and thereafter using VITEK 2 Compact system (Biomerieux, France). The bacterial suspensions were added to the commercially available Gram-positive kit (GPC- P628) and loaded into the automated system for AST.

AST of those isolates was performed against a panel of antimicrobials. This automated system monitors the kinetics of bacterial growth, calculates it using a unique algorithm and follows a Clinical and Laboratory Standards Institute (CLSI) guidelines [8]. As a quality control, *S. aureus* ATCC 25923 was used.

Statistical analysis

WHONET 5.6 software was used to evaluate the isolated staphylococcal species and its antibiotic susceptibility pattern. Using SPSS version 21.0 software, a Chi-square test for linear trend was used to compare antibiotic resistance patterns (SPSS Inc., Chicago, IL, USA). A statistically relevant p value was <0.05.

RESULTS

A total of 1176 isolates of staphylococci were analyzed (Table 1). The skin and soft tissue infections (SSTI) (60%) in addition as lower respiratory tract (23%) were the most important sources of *S. aureus* isolates. On the other hand, majority of the CoNS were from SSTI (54%), followed by blood (25%) and lower respiratory tract (17%) (Table 2). Gender-wise distribution (male and female) among *S. aureus* was found to be 1:08, whereas CoNS is significantly higher in men population (1:06) than women.

All staphylococcal species were identified using automated methods. Among CoNS species, *Staphylococcus epidermidis* (n=351; 39%) was the predominant species, followed by *S. haemolyticus* (n=111; 28%), *S. hominis* (n=49; 12%), *S. saprophyticus* (n=22; 6%), and other CoNS species (n=59; 15%).

The sensitivity of all staphylococci to widely used antibiotics was tested. The prevalence of MRSA and MR-CoNS during the study period is given in Figs. 1 and 2. Overall prevalence of MRSA and MR-CoNS was found to be 50.7% and 55.6%, respectively. Our study shows increasing trend in MRS infection among cancer patients from 2016 to 2019. The prevalence of MRSA isolates was observed from 2016 (37%) with 2018 (58%) and 2019 (57.5%) showing a significant rise in MRSA incidence. An alarming upward trend in MR-CoNS when compared to MRSA. The prevalence of MR-CoNS strains was observed from 2016 (20%) to 2019 (87.5%) showing a significant rise in MR-CoNS incidence.

In the present study, MRSA and MR-CoNS represent variability in drug-resistant pattern. MRSA isolates revealed the highest resistance to ciprofloxacin (84%), followed by erythromycin (79%) and clindamycin (27%), whereas MR-CoNS isolates showed highest resistance to co-trimoxazole (77%), followed by erythromycin (69%) and ciprofloxacin (67%). About 1% and 2.3% of MRSA and MR-CoNS isolates were found to be resistant to linezolid, respectively. All the isolates of staphylococci were sensitive to glycopeptides (Figs. 3 and 4). The incidence of inducible ($iMLS_B$) and constitutive ($cMLS_B$) clindamycin resistance was higher in MRSA ($iMLS_B$ - 17%; $cMLS_B$ - 6.2%) than MSSA ($iMLS_B$ - 12.7%; $cMLS_B$ - 1.3%); whereas among MR-CoNS ($iMLS_B$ - 16%; $cMLS_B$ - 27.5%) and MS-CoNS ($iMLS_B$ - 3.4%; $cMLS_B$ - 1.7%) (Table 3).

DISCUSSION

MRS have disseminated worldwide and still be among the foremost common hospital pathogens. The prevalence and characterization of MRSA and MR-CoNS in hospitals are reported from different parts of the Globe [3,5,9-15]. In the present study, 4-year data of staphylococci infections in cancer patients and prevalence of MRS and

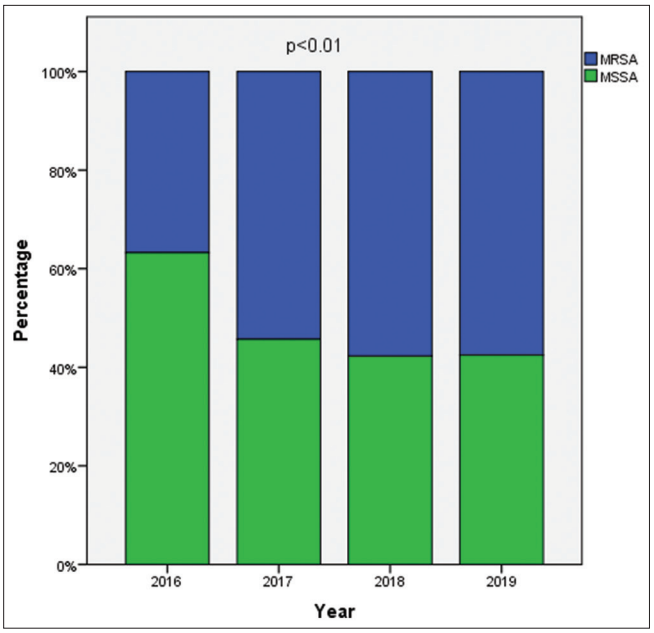

Fig. 1: Prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) and methicillin-sensitive *Staphylococcus aureus* (MSSA)

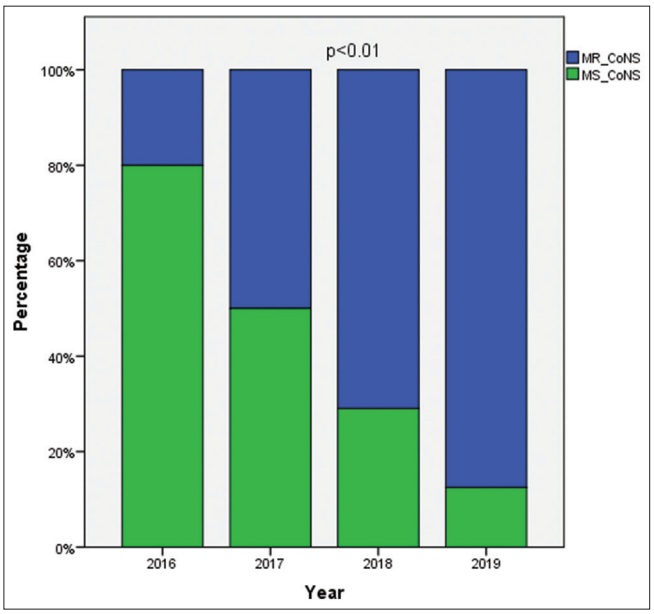

Fig. 2: Prevalence of methicillin-resistant coagulase-negative staphylococci (MR-CoNS) and methicillin-sensitive coagulase-negative staphylococci (MS-CoNS)

Table 1: Overall prevalence of staphylococci in our hospital

Staphylococci	2016 (n=324)	2017 (n=257)	2018 (n=394)	2019 (n=201)	Total prevalence n (%)
Staphylococcus aureus	234	151	246	153	784 (66.7)
CoNS	90	106	148	48	392 (33.3)

CoNS: Coagulase-negative staphylococci

Table 2: Sample-wise distribution of methicillin- sensitive staphylococci and methicillin-resistant staphylococci

Type of specimen	*Staphylococcus aureus* n (%)	MRSA n (%)	CoNS n (%)	MR-CoNS n (%)
Blood	59 (7.5)	36 (9)	97 (25)	42 (19)
Respiratory	182 (23)	105 (26)	68 (17)	40 (18)
Exudates	468 (60)	224 (56)	213 (54)	133 (61)
Urine	21 (3)	11 (3)	12 (3)	2 (1)
Body fluids	54 (7)	22 (6)	-	-

Table 3: Prevalence of inducible and constitutive clindamycin resistance among methicillin-sensitive staphylococci and methicillin-resistant staphylococci

Resistant phenotype	MSSA n (%)	MRSA n (%)	MS-CoNS n (%)	MR-CoNS n (%)
$_{i}MLS_{B}$	48 (13)	68 (17)	6 (3.4)	35 (16)
$cMLS_{B}$	5 (1.3)	25 (6.2)	3 (1.7)	70 (27.5)

MLSB: Macrolide-Lincosamide-StreptograminB

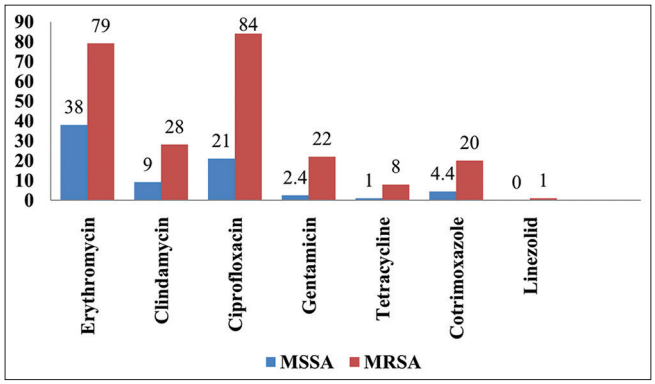

Fig. 3: Overall resistance pattern among methicillin-sensitive *S. aureus* and methicillin-resistant *S. aureus* isolates

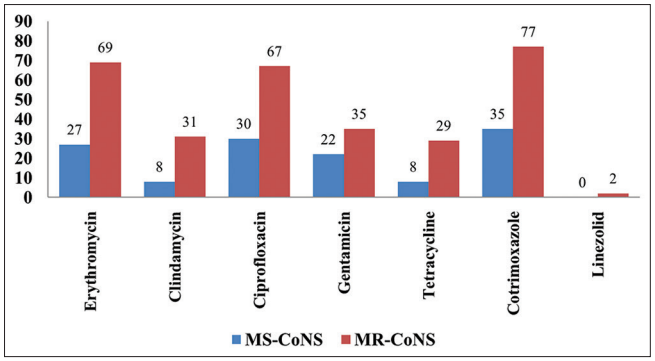

Fig. 4: Overall resistance pattern among methicillin-sensitive coagulase-negative staphylococci (MS-CoNS) and methicillin-resistant coagulase-negative staphylococci (MR-CoNS)

antibiotic susceptibility were analyzed. The incidence of MRS strains was evaluated in several cultures obtained from different cancer patient specimens. In the present study, around 60% of the staphylococci isolates were from exudates, followed by respiratory specimens and Blood. This finding was consistent with the previous reports from India and worldwide [11,16,17].

In our study, the overall prevalence of MRSA was 50.7%. The observed resistance was very low in 2016 (37%) in comparison to 2019 (57.5%). In our study, we found higher rate of MRSA (50.7%) when compared to the previous multicenter studies from India [10,13]. On the contrary, some studies [12,18,19] have found an alarmingly high incidence of MRSA in India and around the world. MRSA prevalence in India and elsewhere, which ranged from 21% to 82%, may be due to regional differences in study design, sample population, antibiotic policies, and thus infection control steps.

MDR patterns were more common in MRSA than MSSA. MRSA isolates showed higher resistance to ciprofloxacin, erythromycin, and clindamycin, though comparable rates of resistance were noticed in gentamicin and co-trimoxazole. The high and increasing level of resistance to ciprofloxacin, a routine antimicrobial, altogether the hospitals was may be expected due to its pervasive use. The findings throughout this study was according to the previous multicentric studies [11,20] and which was not concordance to the previous report [18]. Clindamycin is an efficient drug for staphylococcal skin and soft-tissue infections. Resistance to the macrolide group of antibiotics in staphylococci has been reported from different parts of the world. The overall prevalence of inducible clindamycin resistance among MRSA and MSSA isolates was 68 (17.5%) and 48 (12%), respectively. This was not according to the previous studies performed in South-India and worldwide [15,18]. In general, it is going to be risky to use clindamycin once erythromycin testing shows resistance or intermediate although the microorganism is sensitive to clindamycin. For this reason, routine D-testing may facilitate clinicians to retain.

The alarming increase in MR-CoNS is limiting the effectiveness of all β-lactam agents, limiting therapeutic options significantly. Linezolid could also be a viable solution for methicillin resistant staphylococcal species. According to multinational and multicenter surveillance studies, linezolid is effective against 99% of *S. aureus* and CoNS clinical strains [5,10,11,20]. Emerging resistance to linezolid, on the other hand, may be a major source of concern. It is time that we tend to acknowledge the way reaching consequences expose by such an excellent threat and closely monitor and track resistance to linezolid by prospective resistance surveillance studies, notably wherever frequent and extended linezolid therapy is employed.

MR-CoNS are one of the most common causes of human infections, and their MDR property is more noticeable than that of MS-CoNS. Further, it also serves as a reservoir for resistance genes. Among CoNS, *S. epidermidis* and *S. haemolyticus* were the predominant species reported. The identification of CoNS was performed using automated methods also as MALDI-TOF.

The species distribution of CoNS in our study was concordance with the previous study from India where *S. epidermidis* (39%) was the predominant species, followed by *S. haemolyticus* (28%) [9,10,16]. In contrast, few studies from India and worldwide reported *S. haemolyticus* as the most common species, followed by *S. epidermidis* [3,11,13]. The varying adaptability of different organisms to selective pressures such as biocides and antimicrobials within the ecosystem may be due to differences in colonization characteristics of patients in India and around the world.

Many reports have found a rise in the incidence of hospital-acquired infections caused by MR-CoNS strains in recent years. According to the surveys, the prevalence of MR-CoNS was higher (75–90%) than MRSA in the 1990s, and this trend continues today [4,5]. This was reflected within the present study, MR-CoNS (55.6%) displayed higher percentage in comparison to MRSA (50.7%). Furthermore, MR-CoNS showed high resistance to other non-β-lactam antimicrobial agents – co-trimoxazole (77%), erythromycin (69%), and ciprofloxacin (67%). An overall high prevalence of resistance to all or any antibiotics was observed with MR-CoNS - showing higher resistance to other non β-lactam antimicrobial agents as compared to MS-CoNS. Within the present study, the general incidence of inducible and constitutive MLS_{B} resistance phenotype among the CoNS isolates was 10% and 19%,

respectively. This is often not supported by other studies from India and worldwide [5,10].

As a result, routine monitoring of hospital-associated infections, as well as the antimicrobial susceptibility pattern of MRS and the formulation of a specific antibiotic strategy, may help reduce the burden of MRS infections within the hospital.

CONCLUSION

This study demonstrates that MRS may be a problem in cancer patients at North Kerala. A higher percentage of MR-CoNS isolates are MDR than MRSA isolates. Glycopeptides and linezolid still remain the mainstay for treatment for MRS infections. To reduce the prevalence of nosocomial infection in critical care areas, we recommend education and awareness among healthcare workers and clinicians, as well as adherence to simple guidelines for prevention of nosocomial infection.

Hand hygiene is also the most important tool for preventing infection in health-care settings. This current study will aid our hospital to educate the employees using their own data to drive home the value of their hand hygiene. This study's findings can also be used to help us refocus approaches in other fields.

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Nil.

AUTHORS' CONTRIBUTIONS

Dr. Saravanan M, Ms. Sujina TK, and Dr. Parthiban R contributed substantially to the conception, design of the study, analysis, and interpretation of data. All authors discussed the results and commented on the manuscript. Dr. Saravanan M and Dr. Parthiban R drafted the final manuscript.

CONFLICT OF INTEREST STATEMENT

None to declare.

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Nil.

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