ISSN: 0975-833X

Available online at http://www.journalcra.com

International Journal of Current Research
Vol. 11, Issue, 06, pp.4235-4238, June, 2019

DOI: https://doi.org/10.24941/ijcr.35339.06.2019

INTERNATIONAL JOURNAL OFCURRENTRESEARCH

RESEARCH ARTICLE

EVIDENCE OF COMMUNITY ACQUIRED MRSA OCCURRENCE AMONG INDIAN PATIENTS WITH PRIMARY PYODERMA ATTENDING TERTIARY CARE HOSPITAL – A RETROSPECTIVE STUDY

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ARTICLEINFO

Article History:
Received 18th March, 2019
Received in revised form
20th April, 2019
Accepted 26th May, 2019
Published online 30th June, 2019

Key Words:
MRSA, Community,
India, Pyoderma,
Impetigo,
Furunculitis

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ABSTRACT

Introduction: The CA MRSA gained much attention during mid 1990, after death of four children in Minnesota and North Dakota. The first reports in India on CA MRSA were reported during mid-2000s, whereas the present study reports MRSA among Community acquired pyoderma cases during mid-1990s. **Methods:** A total of 100 coagulase positive *Staphylococcus aureus* isolates, obtained from 185 pyoderma cases. Retrospectively, it is understood that the studied cases fulfilled the criteria for community acquired MRSA including no history of hospitalization, contact with hospitalized patients and previous antibiotic treatment. **Results:** Of 100 *S. aureus* isolates tested for methicillin resistance, 42% were found to be MRSA and 58% were MSSA. MRSA skin infections (89%) were found predominantly among children and young youths. The demographic details of the pyoderma cases show that, 67% of cases had contact with the crowded community setups (school going children). Impetigo (46%), folliculitis (32%) and furunculosis (20%) were found to be the commonest infections in this study. Phage typing of *S. aureus* showed all four phage groups and mixed groups. **Conclusion:** Retrospectively, it is evident from the existing demographic data and scientific data of the study, that in India the MRSA were circulating in community settings during 1990s, its first reports in USA.

***Citation: Suresh Chander, V.C. and Kavin Kumar, A.* 2019.** "Evidence of community acquired MRSA occurrence among Indian patients with primary pyoderma attending tertiary care hospital – A retrospective study", *International Journal of Current Research*, 11, (06), 4235-4238.

INTRODUCTION

MRSA is a common nosocomial infection associated with hospital and accounts for more than 50% of *S aureus* isolates from intensive care units in the western countries (Lindsay, 2013). There are reports of 30-85% MRSA prevalence in hospitals from different parts of India (Anupurba *et al.,* 2003). MRSA traditionally considered being a strict hospital pathogen has now known to cause infections among community settings, where the population has no history of risk factors of hospital associated infections. These MRSA isolates causing infections in the community were known as Community Acquired MRSA (DeLeo *et al.,* 2010; Mediavilla *et al.,* 2012). CA MRSA gained much attention in 1990s, when it was first reported in Minnesota and North Dakota causing death of 4 children. The CA MRSA isolates were found to be USA 300 and USA 400 clonal lineages (PFGE types) (Centers for Disease Control and Prevention, 1999). The later reports on CA MRSA confirmed that it is a heterogenous mix of strains. CA MRSA has become the most common pathogen of skin and soft tissue infections accounting up to 67% of *Staphylococcus aureus* infection in the community (Moran *et al.,* 2015). In Indian scenario, there are few reports on CA MRSA infections among community settings and also among hospital settings, which has now become endemic in Indian hospitals. The first report of CA MRSA in India is evidenced from the study by Saxena et al in 2003, where it showed about 18% of nasal carriage isolates from healthy adults to be MRSA (Saxena *et al.,* 2003). The first reports of clinical isolates of CA MRSA were published by Nagaraju et al., in 2004 and Patil *et al.* in 2006 (Umashankar *et al.,* 2004; Patil *et al.,* 2006). This study, which is a part of the approved post graduate thesis submitted to the Kuvempu University, India, evidence that during mid 1990s itself, MRSA was circulating in Indian community settings causing skin infections. This study on MRSA from primary pyoderma warrants a wider audience to have a better understanding of the epidemiology of CA MRSA in India.

METHODS

Study population: A total of 185 cases of primary pyoderma cases attending skin and veneral diseases and pediatric outpatient facilities of Chigateri General Hospital attached to JJ Medical College, Devangere during the period 1996 – 97, were included in the study. The demographic data of patients including name, age and gender, type of skin lesion, socioeconomic status, occupation, previous history of hospitalization, underlying medical conditions, antibiotic treatment and contacts with crowded community settings such

as schooling and hospitalized family members were recorded. The study was approved by institutional committee of the JJ Medical College and informed consents were obtained from the study participants.

Exclusion criteria: The patients with previous history of hospitalization for underlying medical conditions, contact with hospitalized patients, antibiotic therapy (oral, parenteral and topical) were excluded from the study.

Sample collection: Aseptically, pus (pus exudes) samples were collected from the infected sites using sterile swabs in duplicates and labelled appropriately. The swabs were immediately transported to the microbiology laboratory of JJ Medical College, Devangere. Control swabs were also collected and included in the study.

Microbiological methods: One of the pus swabs collected was used for direct smear and staining and other swab was utilized for culture on blood agar and glucose broth, incubated at $37^{O}C$ for overnight. Blood agar and glucose broth were observed for the growth of *Staphylococcus aureus* using staining and standard biochemical tests. Gram positive cocci in clusters were confirmed by tube coagulase test using citrated rabbit plasma (Elmer *et al.,* 1992). *S. aureus* ATCC 25923 was used as positive control for biochemical test.

Antibiotic susceptibility testing: Antibiotic susceptibility testing was carried out on Muller Hinton Agar by Kirby Bauer disc diffusion method (Biemer, *et al.,* 1973) and interpreted using NCCLS (CLSI), 1997 guidelines (Wayne, 1997). Following commonly used antibiotics at the hospital amikacin (30 µg), co-trimoxazole (25 µg), cloxacillin (µg), ciprofloxacin (5), chloromycetin, erythromycin (15µg), gentamicin (10 µg), tetracycline (3 µg) and penicillin (P-2U) were test for sensitivity.

Screening of methicillin resistance: Screening of methicillin resistance among 100 confirmed cases of coagulase positive *S. aureus* was done by Kirby Bauer Disc Diffusion test using methicillin disc (1µg) (Biemer, 1973) and interpreted using NCCLS, 1997 guidelines (Wayne, 1997).

Phage typing: A total of 86 coagulase positive *S. aureus* isolates were sent to the National Reference Phage Typing Centre at Moulana Azad Medical College. The phage typing was at RTD and RTDx100 dilutions and results were received.

RESULTS

Staphylococcus aureus was isolated from a total of 105/185 (56.75%) community acquired primary pyoderma cases included in the study. The age of the study participants ranged from 3 months to 66 years, with mean age of 14 years. Majority of the cases 76% were below 20 years and 91% were below 25 years. The male female ratio was found to be 60:40. 81% of the cases were found to fall under the low economic status category. 67% of cases were found to be school going children and 33% had no contact to community setup other than family contact. The types of the pyoderma among studied cases were found to be impetigo (46%), folliculitis (32%), furunculosis (20%) and ecthyma (2%) (Table 1). 5 *S. aurues* isolates were lost during subculture. 100 isolates were tested for antibiotic resistance and their resistance to tested antibiotic were as follows penicillin (86), cloxacillin (50), tetracycline (46), erythromycin (49), ciprofloxacin (25), cotrimoxazole (23), amikacin (17) and gentamicin (16) [Figure 1]. A total of 42 isolates were found to be methicillin resistant by oxacillin disc diffusion method. 51/85 strain were typable by phage typing and 34 were non typable at RTD and RTD 100 dilutions. Results of phage typing were as follows; phage group I – 7, phage group II – 7, phage group III – 4, phage group IV – 4 and mixed phage group – 31 (Table 2).

Table 1. Demographic details of community acquired pyoderma cases with *S. aureus*

Demographic details	MRSA (N)	MSSA (N)
Age		
Infant (up to 12months)	1	0
Children (2-18 years)	32	39
Young adults (18 – 25)	8	12
Adults (25 – 40 years)	1	11
Gender		
Male	29	31
Female	13	17
Socioeconomic status		
Lower level	36	45
Middle level	6	12
Upper level	0	1
Contact with community setups		
School going children	32	35
Family contacts	10	23
Type of Community acquired pyoderma		
Impetigo	21	25
Folliculitis	14	16
Furunculosis	6	14
Ecthyma	1	1

Table 2. Phage Typing of *S. aureus* (n=85)

Phage Group	No of isolates	% of isolates	At RTD	RTD x 100
Group I	7	8.2	6	1
Group II	7	8.2	7	
Group III	4	4.7	4	
Group IV	4	4.7	3	1
Mixed Group	31	36.4	31	
Not typable	32	337.6	32	

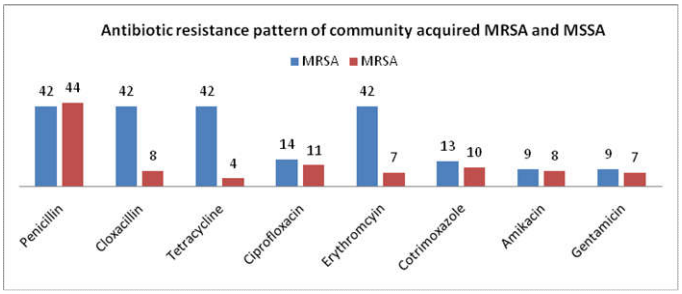

Fig. 1. Antibiotic resistance pattern of Community acquired MRSA (n=42) and MSSA (n=58)

DISCUSSION

The study was undertaken to determine the prevalence of *S. aureus* among community acquired primary pyoderma cases, to screen their methicillin resistance status and find their phage group. Impetigo was found to be commonest primary pyoderma (46%), followed by folliculitis (32%) and furunculitis (20%), source of *S. aureus* in this study. Other Indian studies show that folliculitis to be the commonest pyoderma cases of source of *S. aureus* infection (Umashankar *et al.,* 2004; Patil *et al.,* 2006). The current study shows that, healthy individuals from crowded community setup, school children (67%) and economically backward sections (81%) of

the community were found to be significantly susceptible to community acquired *S. aureus* infections. A total of 42% of *S. aureus* isolates in this study was found to be MRSA, which is high when compared to the other reported studies on community acquired pyoderma from India and abroad, during that period (Umashankar *et al.,* 2004; Patil *et al.,* 2006; Herold *et al.,* 1998), which reported 11%, 1.4% and 27% respectively. In-depth analysis of the present study showed that the MRSA among children attending school was high as 47.7% and for individuals with no known contact with community setup other than family contact was 30.3%. The prevalence of MRSA among children was very high 45.1%, compared to the previous reports on MRSA from community acquired infections (Herold *et al.,* 1998; Kale and Dhawan, 2016; Alvarez-Uria and Reddy, 2012; Gorak *et al.,* 1999; David and Daum, 2010), which reports upto 30% of the community acquired MRSA infections. The emergence of antibiotic resistance to available antibiotics poses a threat in deciding the empirical therapy. In the present study, the antibiotic susceptibility testing was done to detect resistance pattern of *S. aureus* to the routinely used antibiotics. The anbiotic resistance to commonly used antibiotics was found to be high for MRSA than MSSA. Of the tested antibiotics lowest resistance was detected for gentamicin (16%), followed by amikacin (17%), cotrimoxazole (23%) and ciprofloxacin (25%), whereas highest resistance was detected for penicillin (86%), followed by cloxacillin (50%), erythromycin (49%) and tetracycline (46%). Resistance to vancomycin, linezolid, fusidic acid and mupirocin were detected in this study. MRSA isolates from community pyoderma showed multidrug resistance, resistant to erythromycin, tetracycline and cloxacillin. Resistance to ciprofloxacillin, gentamicin, amikacin, cotrimoxazole and penicillin were found even among MRSA and MSSA from community primary pyoderma cases. The current study was in correlation with the previous first reports in that, the community acquired MRSA isolates were susceptible multiple antibiotics, especially cotrimoxazole, ciprofloxacin and aminoglycosides (David and Daum, 2010). The phage typing at National reference centre showed that 60% were typable, of which 34 were mixed types. The phage typing was found not significant with respect to MRSA typing, it is unfortunate that MRSA phage type were not available during the study period, which is a disadvantage of the study.

The current study was conducted during mid-1990s, taking account of the risk factors of CA MRSA infections in the inclusion criteria and excluding the risk factors of hospital acquired infections. The strength of the study was that it was conducted in the period, when the risk factors and occurrence of CA MRSA is not so popular in India. The CA MRSA gained much attention after it caused the loss of four children in Minnesota and North Dakota in USA during 1997 (Centers for Disease Control and Prevention CDC, 1999). In India during mid-2000s, few studies reported CA MRSA among healthy nasal carriers of *S. aureus* from community and also from patients with primary pyoderma infections attending outpatient facilities (Umashankar *et al.,* 2004; Patil *et al.,* 2006). The present study reveals that the CA MRSA isolates were in circulation during mid-1990s causing community acquired primary pyoderma among the healthy children of socioeconomically backward population and remain unnoticed until mid-2000s. Hence, the present study warrants a wider publicity among the scientific community in order to better understand the evolution and epidemiology of the CA MRSA in Indian population.

Conflict of Interest – None declared

Acknowledgement: We thank the participants for providing the clinical samples for the study. We thank our beloved Prof. Dr. Maheshwarrappa for his valuable guidance throughout the study.

Funding: The above study is not funded.

Abbreviations used in this Articl

MRSA	– Methicillin resistant Staphylococcus aures
MSSA	– Methicillin susceptible Staphylococcus aures
CA MRSA	– Community acquired Methicillin resistant Staphylococcus aures
HA MRSA	– Hospital acquired Methicillin resistant Staphylococcus aures
NCCLS	– National committee for Clinical Laboratory Standards
CLSI	– Clinical Laboratory Standards Institute
PFGE	– Pulse Field Gel Electrophoresis
ATCC	– American Type Culture Collection
RTD	– Routine test dilutions

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