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Incidence of Community acquired MRSA in the nasal swabs of general population of Gulbarga region

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Abstract

Background: *Methicillin resistant S. aureus (MRSA) has emerged as a major pathogen in both community and hospital settings. Nasal carriage of S. aureus represents a major risk factor for the development of infection with this bacterium.*

Objective: *The aim was to study community acquired MRSA incidence among nasal swabs of general population of Gulbarga.*

Methods: *Nasal swabs from anterior nares were collected for the isolation of S. aureus from 224 individuals including Microbiology Department students and general population of Kusnoor village in Gulbarga region, Karnataka. The nasal swabs were enriched in nutrient broth for enrichment of S. aureus then streaked on Mannitol Salt Agar (MSA) and incubated at 37 °C for 24 hours. Conventional methods such as growth characteristics on mannitol salt agar, Gram's staining, and biochemical characteristics have been performed for characterization of the S. aureus. Antibiotic sensitivity test was carried out for the isolated S. aureus using different antibiotics.*

Results: *A total of 224 nasal swabs were screened and the incidence of S. aureus was observed to be 41.96% (94/224). Of the total 94 S. aureus isolates, 1 (1.06%) isolate was found to be MRSA and 93(98.93%) isolates were MSSA. A total of 3 (3.22%) S. aureus isolates were found to be multi-drug resistant (MDR) in this study.*

Conclusion: *Our results suggest that general populations are at risk and are potential carriers of S. aureus and in particular MRSA.*

Keywords: *Methicillin resistant S. aureus (MRSA), anterior nares, S. aureus, community-acquired MRSA (CA-MRSA)*

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INTRODUCTION

Staphylococcus aureus is one of the most common human pathogens with ability to cause a wide range of infections. An average 20-40% of the adults carry of *S. aureus* in the anterior nares [1]. Methicillin-resistant *Staphylococcus aureus* (MRSA), a pathogen which is responsible for many nosocomial infections was first reported in 1960. Over the past few decades it has emerged in the community as well, and it is currently considered a threat to public health [2]. The anterior nares have been shown to be the main reservoir of *S. aureus* in both adults and children. The *S. aureus* is transmitted to nares by contaminated hands and from surfaces where it can survive for months. Nasal carriage of *S. aureus* acts as endogenous reservoir for clinical infections in the colonized individual but also as a source of cross-colonization for community spread. The spread of colonization occur especially in close contact areas like schools, pre-schools or households probably by the contaminated hands and surfaces [3]. Strains of MRSA that causes sudden rise in the incidence of infections and are grouped into healthcare- associated (HA) or community-associated (CA) MRSA. HA-MRSA is supposed to be of nosocomial origin and usually causes infections upon prolonged hospitalization or in patients with indwelling catheters and devices, or those undergoing dialysis, surgery, or antimicrobial or immunosuppressive therapy. On contrast, CA-MRSA strains originate from healthy individuals with no risk factors or previous healthcare contact and constitute a major proportion of skin and soft-tissue infections (SSTIs). Antimicrobial studies reveal that CA-MRSA is typically susceptible to most antibiotics except methicillin and β-lactams [4].

The present study was aimed to understand prevalence of nasal colonization by *S. aureus* and the potential risk factors for MRSA nasal colonization in general population of Gulbarga region. The present study helps to describe the evolving epidemiology of nasal colonization by *S. aureus* and MRSA.

MATERIALS AND METHODS

Overall a total of 224 nasal swabs were collected from general population of Gulbarga region. In the present study, to know the incidence of community acquired MRSA, different populations with different age groups, occupation and places were selected. The samples were collected using sterile cotton swab from the anterior nares of Microbiology Department post –graduate students and research scholars and general population of selected places within Gulbarga region.

All the details of the individual person were filled in a datasheet which contained brief information of name, age, sex, occupation, living status, sample number and history about health status.

Collection of samples

Required samples were collected from the anterior nares with the help of sterile cotton swabs. Before taking samples the swabs were dipped in sterile saline in propylene tubes (Hi-Media Pvt. Ltd; Mumbai). These swabs were then rubbed in both the nares of the individuals. The collected swabs were immediately kept into the sterile propylene tubes provided with the swabs and put in the transport box. These swabs were transported to laboratory for processing.

Isolation of *S. aureus* from the collected Specimens

In the laboratory, these samples were enriched by incubating for 6-12 hrs in the nutrient broth [5] then it was inoculated on to the mannitol salt agar (MSA) medium which serves as the selective medium for isolation of *S. aureus*.

Phenotypic Identification

For characterization of the *S. aureus*, conventional methods like growth characteristics on mannitol salt agar, Baird Parker agar, Grams staining and biochemical characteristics such as coagulase test and catalase test have been performed [6]. Antibiogram was carried out using 12 different antibiotics including methicillin and vancomycin. Antibiotic sensitivity tests had been performed as per CLSI guidelines [7, 8]

Antibiotic Sensitivity Test

Antibiotic Sensitivity test was done for each *S. aureus* isolate by the Kirby-Bauer disc diffusion method against ciprofloxacin (5µg), erythromycin (15µg), cloxacillin (30µg), vancomycin (30µg), ceftizoxime (30µg), ampicillin (10µg), penicillin (10µg), methicillin(5µg), amikacin(30µg), cefoxitin(30µg), oxacillin(1µg), and gentamycin(10µg). The *S. aureus* confirmed isolates were inoculated to 2-3 ml of nutrient broth. The tubes were allowed for incubation for about 6 hours then turbidity was adjusted to 0.5 MacFarland's standard. To the turbid solution a sterile cotton swab was dipped and a uniform lawn was made by spreading the swab evenly on Mueller Hinton Agar plates. The plates were allowed to dry for 5 minutes. After that the antibiotic discs were kept in such a way that space between two dices was 12-15 mm on the plates with the help of sterile forceps. Then the plates were incubated for about 18-24 hours in inverted position in an incubator at 37^0 C. After the incubation, individual antibiotic sensitivity was measured with the help of zone measuring scale (CLSI Guidelines). Then the tested all *S. aureus* isolates were preserved in tubes containing nutrient broth and 25% glycerol for further study.

RESULTS

Overall incidence of *S. aureus* in the community samples

A total of 94 (41.96%) *S. aureus* were isolated from the 224 nasal swab samples collected from the general population of Gulbarga. We observed highest incidence of 64.70% in the university students and lowest of 32.05% incidence was observed in the village people. Over all incidence of S. aureus was higher in males (49.56%) than in females (33.94%). Interestingly it was more than double in males (44.74%) compared to females (20%) among the samples collected from the general population of a village (Table 1).

Table 1: *Incidence of S. aureus in the community samples.*

Sample	Sample Collection			Incidence of *S. aureus*		
	Male	Female	Total	Male (%)	Female (%)	Total (%)
Microbiology Dept	39	29	68	23 (58.97)	21 (72.41)	44 (64.70)
Kusnoor Village	76	80	156	34 (44.74)	16 (20.00)	50 (32.05)
Total	115	109	224	57 (49.56)	37 (33.94)	94 (41.96)

Overall incidence of MRSA in the community samples

The incidence of MRSA was very low of 1.06% among the 94 *S. aureus* isolated from the 224 nasal swab samples collected from the general population of Gulbarga. Interestingly MRSA incidence was more in the samples collected from university students (2.27%) than in village population where not even single isolate tested to be MRSA (0%) among the 50 S. aureus isolates. Only 1 isolate was reported to be MRSA which was tested to be MDR strain while less than 13% of MSSA isolates were tested for MDR (Table-2).

Table 2: *Incidence of MRSA in the community samples.*

Source of Sample	Total Samples	*S. aureus* Isolates	MRSA (%)	MDR-MRSA (%)	MSSA (%)	MDR-MSSA (%)
Microbiology Dept	68	44 (64.70)	01 (2.27)	01 (2.27)	43 (97.72)	09 (20.93)
Kusnoor Village	156	50 (32.05)	00 (0.00)	00 (0.00)	50 (100.00)	03 (6.00)
Total	224	94 (41.96)	01 (1.06)	01 (100%)	93 (98.93)	12 (12.76)

Age group wise incidences of *S. aureus* in the general population

Total of 224 samples was collected from the general population. We observed maximum incidence of *S. aureus* in the age group 21-30 years (51.78%) and lower incidence was found in the age groups 51-60(0%) and and 61 and above age group (0%).Interestingly incidence was more than double in males than in females (Table-3).

Table 3: *Incidence of S. aureus among the general population nasal swabs (Age group wise).*

Age group in years	Samples collected			Incidence of *S. aureus*		
	Male	Female	Total	Male (%)	Female (%)	Total (%)
0-10	28	23	51	12 (42.86)	6 (26.08)	18 (35.29)
11-20	19	18	37	10 (52.63)	4 (22.22)	14 (37.83)
21-30	59	53	112	32 (54.23)	26 (49.05)	58 (51.78)
31-40	5	3	8	2 (40.00)	1 (33.33)	3 (37.50)
41-50	1	7	8	1 (100)	0 (00.00)	1 (12.50)
51-60	1	2	3	0 (00.00)	0 (00.00)	0 (00.00)
61 and +	2	3	5	0 (00.00)	0 (00.00)	0 (00.00)
Total	115	109	224	57(49.56)	37(33.94)	94 (41.96)

Incidence of MRSA among the general population nasal swabs (Age group wise)

Overall incidence of MRSA was found to be 1.06%. Maximum MRSA incidence was found in the age group 21-30 (1.72%).

Table 4: *Incidence of MRSA among the general population nasal swabs (Age group wise)*

Age Group (In years)	Total Samples	*S. aureus* Isolates	MRSA	MDR-MRSA	MSSA	MDR-MSSA
0-10	51	18	00 (0.00)	00 (0.00)	18 (100.00)	01 (5.55)
11-20	37	14	00 (0.00)	00 (0.00)	14 (100.00)	01 (7.14)
21-30	112	58	1 (1.72)	1 (1.72)	57 (100.00)	09 (15.51)
31-40	08	03	00 (0.00)	00 (0.00)	03 (100.00)	01 (33.33)
41-50	08	01	00 (0.00)	00 (0.00)	01 (100.00)	00 (0.00)
51-60	03	00	00 (0.00)	00 (0.00)	00 (0.00)	00 (0.00)
61 and above	05	00	00 (0.00)	00 (0.00)	00 (0.00)	00 (0.00)
Total	224	94	1 (1.06)	1 (100.00)	93 (98.93)	03 (3.22)

DISCUSSION

Staphylococcus aureus has long been recognized as an important pathogen in human

disease and nasal carriage appears to play a key role in the epidemiology and pathogenesis of infection. Methicillin (MRSA) is now being frequently encountered in the community and their emergence has led to the development of multi-drug resistant strains. Increase in the reports of such MDR strains in the hospital settings has given rise to the alarm of such strains spreading to the community [9, 10].

Community-associated methicillin-resistant *Staphylococcus aureus* (MRSA) is a leading cause of skin and soft-tissue infections in many parts of the world and many cases have been reported in healthy community individuals with no traditional risk factors for MRSA infection [11, 12].

In the present study 41.96% of *S. aureus* in nasal carriage rate has been observed which is much higher than compared to the study by Yukti Sharma et al with 12%, however, the quoted incidence rates for *S. aureus* nasal carriage is 20–40% [13, 14].

The study conducted by Fomda *et al*, 2014 showed an overall prevalence of 27.92% of *S. aureus* in the nostrils of the healthy subjects [13, 15, 16].

Study on healthy contacts of outpatient from an urban community of paediatric cases at Delhi showed a rate of 5.3 % [17]. A study from Japan reported 36% of *S. aureus* in nares of Japanese adults and 32.4% in nasal cavity of adults in USA, which is lower in comparison to results of our study [18].

Our study showed a low prevalence of 1.06% of MRSA nasal carriage while prevalence was found to be 5% and 19% in the study conducted by by Yukti Sharma et al., and Ramana et al., respectively [13, 15]. While, our study showed almost similar prevalence of MRSA nasal colonisation compared to1.83% a study conducted by Fomda et al., [16].

Munckhof et al. found a prevalence of only 0.7% in Queensland, Australia [19]. However, a higher propensity was observed amongst MRSA strains reported in USA with 24.15% [20]. Taiwan and Zaria, Nigeria have also reported a prevalence of 13.6% and 14.85%, respectively, from anterior nares of healthy population, adults and school children [21].

Prevalence of *S. aureus* nasal carriage among healthy adults ranges from approximately 20% to 30%, with higher prevalence in overcrowded population [22, 23, 24]. Carriage rates varying in different places may be attributed to the characteristics of the population under study, sampling sizes and culture techniques. Single sample of S aureus isolation further result in under estimating the number of intermittent carriers and hence to improve multiple nasal swabs should be taken from both anterior nares.

Studies in the developed world suggest that factors associated with CA-MRSA carriage include prior antibiotic usage, contact with health care facility, poor socioeconomic conditions and overcrowding [25, 26]. This finding is of great importance since the nasal carriage of *S. aureus* play an important role in the epidemiology and pathogenesis of infection. Continuing surveillance is needed to more accurately assess the prevalence, geographic distribution and epidemiology of community acquired infections. The results emphasise the need to improve personal hygiene and discourage antibiotics abuse so as to prevent the return of the consequences of pre-antibiotic era.

CONCLUSION

In conclusion, our results suggest that in general population the incidence rate of *S. aureus* nasal carriage was found to be 41.96% (94/224) and MRSA nasal carriage was found to be 1.06% (1/94). The general population is potential carrier of S. *aureus*.

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