

Original Research Article

Staphylococcus aureus nasal carriers and the prevalence of methicillin resistant *Staphylococcus aureus* among medical students

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ABSTRACT

Background: *Staphylococcus aureus* is a dynamic and adaptable bacterium that has an incredible talent to attain antibiotic resistance. Nasal colonisation of *S.aureus* increases with greater exposure of students to the hospital environment. The prevention of staphylococcal infection and reduction of spread and emergence of MRSA are essential. Medical students would be a key target group to introduce awareness of hospital-acquired infections. Therefore, the present study aims at understanding the prevalence of carrier rate of *S.aureus* and MRSA among Medical students during their clinical postings.

Methods: Nasal swabs were taken from 150 medical students. Their *Staphylococcus aureus* nasal carriage was determined by plating on sheep blood agar and MRSA carriage by to Kirby Bauer Disc Diffusion method using Cefoxitin disc (30 µg).

Results: The present study showed a 40% nasal carriage of *S.aureus* amongst medical students. Of the 60 *S. aureus* isolates 28%, 36% and 56% isolates were from 1st, 2nd and 3rd year respectively. The colonisation rate of MRSA in the present study was found to be 12.66% amongst the medical students of 6%, 10%, 22% belonged to 1st, 2nd, and 3rd respectively

Conclusions: The nasal carriage of *Staphylococcus aureus* and methicillin resistant *Staphylococcus aureus* (MRSA) among medical students increases as their exposure to patient care increases. Medical students should be made aware of the risks of carrying *S.aureus* and MRSA and educated about the hand washing protocol and safety precautions to be followed while handling patients.

Keywords: *Staphylococcus aureus*, Methicillin resistant *staphylococcus aureus*, Medical students

INTRODUCTION

Staphylococcus aureus is a dynamic and adaptable bacterium that has an incredible talent to attain antibiotic resistance.¹ Nosocomial infections by *S.aureus* result in morbidity of hospitalized patients, prolonging the duration of hospitalisation and increasing the cost of health care. *S.aureus* has been associated with surgical

wound infections; hospital-associated pneumonia, catheter-associated infections and bacteraemia.²

Methicillin resistant *Staphylococcus aureus* (MRSA) strains had rapidly emerged and became a major problem in hospitals immediately after the Methicillin was introduced. It became a major nosocomial pathogen in community, hospitals, long term care facilities and tertiary care hospitals. MRSA colonisation precedes

infection, anterior nares being the ecological niches of *S. aureus*.¹

The prevalence rate of MRSA had reached 50% in the United States hospitals. In the United Kingdom, MRSA accounted for 44% of *S. aureus* isolated from health care workers. In Japan, MRSA accounted 60-70% of *S. aureus* isolated from inpatients. In Saudi Arabia, the MRSA nasal carriage rate among healthcare workers was reported as 76%.³ In Delhi, India, MRSA nasal colonization among healthcare workers was reported to be 37.3%.⁴ In South India, the MRSA positivity of 8.5% was reported among health care workers.⁵

Thus, healthcare workers are at a higher risk of colonization by MRSA than the general public, apparently due to increased exposure to this organism. The presence of MRSA in health institutes is directly proportional to high rate of infections caused by this strain. This may lead to a relative increase in treatment cost and length of hospital stay. Therefore, screening for MRSA in hospitals is an important factor for building up successful infection control strategies. Medical students would be a key target group to introduce awareness of hospital-acquired infections.³ Few studies have analysed whether the patient exposure creates a risk of MRSA carriage in medical students, especially in the Indian setting.¹ Some of the published studies further raised concern, as they reported that nasal colonisation of *S. aureus* increased with greater exposure of students to the hospital environment.⁶

Therefore, prevalence studies need to be carried out to screen this group to assess their carriage status during their clinical rotations.³ Hence, the present study aims at understanding the prevalence of carrier rate of *S. aureus* and MRSA among Medical students.

METHODS

A total of 150 medical students i.e. 50 students each from first, second and third year of their MBBS course were included in a cross-sectional study by purposive nonprobability sampling. The study was carried out in a tertiary care hospital in Aurangabad from August to October 2015. The approval and clearance of ECRHS (Ethics Committee for Research on Human Subjects) for the research was taken from the ethical committee before initiation of the research activity. Consent was taken from the students. Both male and female students were included in the study. Students suffering from any clinically evident infection or who have used antibiotics in the past 30 days were excluded from the study.

Specimen collection

First of all, the person collecting the sample washed his hands with soap and water and then a disinfectant hand rub was used. Gloves were worn and then the sample was collected. One swab was used for each student. The

subject was made to sit comfortably. The sample was collected from both the anterior nares by sterile disposable cotton swab sticks after moistening it with few drops of distilled water from a distilled water ampule. The swabs were rubbed very well 5 times over the inner wall of ala and nasal septum.¹ The swab was then placed back into its container. This procedure was done with utmost precaution to avoid the swab being touched by any other body part or surface. The container was labelled properly with the student's name and academic year. The students were grouped according to their year and their containers were labelled as A, B, C for 1st, 2nd, 3rd year respectively and were also numbered for identification during microbiological procedures. The swabs were brought to the Microbiology Department for further processing without any delay.

Microbiology procedures

All swabs were aseptically plated on Sheep Blood Agar. Two swabs each from a different student were plated on a single plate, each swab occupying half of the plate. The swabs were streaked to get growth. The plates were incubated at 37°C for 18-24 hours. The plates were read and the staphylococcal colonies were identified from colony morphology. Opaque, pale yellowish, circular convex, smooth colonies, 1-2mm in diameter, with or without haemolysis were subjected to gram staining and catalase test. Gram positive cocci in clusters giving positive catalase test were considered as Staphylococci.⁷ These were further processed by coagulase test to identify *S. aureus*. Those which gave positive slide coagulase test were labelled as *S. aureus* and those who gave a negative coagulase test were subjected to tube coagulase test. Isolates giving a positive coagulase test were taken as *S. aureus*. Those who were negative for slide and tube coagulase were labelled as coagulase negative Staphylococci (CoNS).⁸



Figure 1: Cefoxitin disc test for MRSA.

B15 - (2nd year student) shows zone of inhibition of 19 mm - MRSA. B16 - (2nd year student) shows zone of inhibition of 30 mm - MSSA.

Detection of methicillin resistance

As per the CLSI Guidelines (2014), all isolates of *S. aureus* were subjected to Kirby Bauer Disc Diffusion

method using 30 µg Cefoxitin disc.⁹ For this a part of the colony was inoculated in peptone water and incubated at 37°C for a few hours, till the turbidity matched with 0.5 McFarland's turbidity standard to get a standard inoculum. A sterile swab was taken and immersed in the standardised inoculum and excess inoculum squeezed by pressing the swab on the sides of the test tube. Mueller Hinton agar was taken and the swabs were streaked in three directions on one half of the plate so as to get a uniform confluent growth. The other half of the plate was used for another swab. Care was taken during streaking so that the two inoculates on the same plate do not mix. Plates were incubated at 33- 35°C for 24 hours.

Interpretation was done as per CLSI Guidelines.⁹

Zone of Inhibition ≤ 21 mm = Methicillin Resistant *Staphylococcus aureus* - (mecA positive)

Zone of Inhibition ≥ 22 mm = Methicillin Sensitive *Staphylococcus aureus* (MSSA) - (mecA negative)

Control strains used

S.aureus ATCC 25923 = mecA negative (cefoxitin zone =23-29 mm)

S.aureus ATCC 43300 = mecA positive (cefoxitin zone ≤ 21 mm) (Figure 1)

Statistical analysis

Data was collected in the case record form. Then it was compiled and Chi square test was applied to the data as a test of significance and a $p < 0.05$ was deemed statistically significant.

RESULTS

In the present study, a total of one hundred and fifty medical students were enrolled, comprising of 50 students each from first, second and third year of their MBBS course. They were screened to find out the prevalence of nasal carriage of *Staphylococcus aureus* and methicillin resistant *Staphylococcus aureus*. Of these 94 (62.66%) were females and 56 (37.33%) were males in an age group of 18-23 years.

Table 1: Academic year wise distribution of *Staphylococcus aureus* carriage among medical students.

<i>S.aureus</i>	1st year	2nd year	3rd year	Total
	Students	Students	Students	Students
Isolated	14 (28%)	18 (36%)	28 (56%)	60 (40%)
Not Isolated	36 (72%)	32 (64%)	22 (44%)	90 (60%)
Total students	50 (100%)	50 (100%)	50 (100%)	150 (100%)

Chi square value =8.666, degree of freedom =2, p value=0.013 (Significant).

Table 2: Academic year wise distribution of methicillin resistant *Staphylococcus aureus* nasal carriage among medical students.

MRSA	1st year students	2nd year students	3rd year students	Total students
Isolated	3 (6%)	5 (10%)	11 (22%)	19 (12.66%)
Not Isolated	47 (94%)	45 (90%)	39 (78%)	131 (87.33%)
Total students	50 (100%)	50 (100%)	50 (100%)	150 (100%)

Chi square value =6.267, degree of freedom =2, p value=0.043 (Significant).

Table 3: Result of *Staphylococcus aureus* colonisation among preclinical and clinical groups.

<i>S.aureus</i> colonisation	Preclinical group (1st year)	Clinical group (2nd year + 3rd year)	Total students
Isolated	14 (28%)	46 (46%)	60 (40%)
Not Isolated	36 (72%)	54 (56%)	90 (60%)
Total students	50 (100%)	100 (100%)	150 (100%)

Chi square value =4.5, degree of freedom =1, p value=0.033 (Significant).

Table 4: Result of methicillin resistant *Staphylococcus aureus* colonisation among preclinical and clinical groups.

MRSA colonisation	Preclinical group (1st year)	Clinical group (2nd year + 3rd year)	Total students
Isolated	3 (6%)	16(16%)	19(12.66%)
Not Isolated	47(94%)	84(84%)	131(87.33%)
Total students	50 (100%)	100 (100%)	150 (100%)

Chi square value =3.013, degree of freedom =1, p value =0.082 (Not Significant).

Out of the 150 swabs collected, 60 swabs were identified as having *S. aureus*. Thus the *S. aureus* nasal carriage was 40% among 150 medical students. Of the 60 *S. aureus* isolates 14, 18 and 28 isolates were from 1st, 2nd and 3rd year respectively. The p value for these observations was significant ($p < 0.05$). An increasing prevalence is seen among medical students from 1st year to 3rd year (Table 1).

Out of the 60 swabs positive for *S. aureus*, 35 swabs were from females and 25 swabs were from males. The p value was not significant with respect to gender.

MRSA was isolated from 19 out of 150 students i.e. MRSA carriage was of 12.66%. Of the total 19 MRSA isolates 3, 5 and 11 isolates were from 1st, 2nd and 3rd year respectively. Thus, MRSA carriage for 1st year was 6%, for 2nd year it was 10% and for 3rd year it was 22%. The probability value is significant for these results. The prevalence of MRSA carriage is increasing from 1st to 3rd year (Table 2).

Out of the 19 MRSA isolates, 11 swabs were from females and 8 swabs were of males. The association of gender with MRSA carriage was not significant.

In the Indian MBBS course, 1st year students are not exposed to patient care; thus we can consider them to be a preclinical group. From 2nd year onwards and into the 3rd year, the students are exposed to patient care, thus we can consider them as a clinical group. Accordingly we have evaluated the results of nasal carriage of *S. aureus* and MRSA in these groups. The prevalence of *S. aureus* colonisation was more in the clinical group (46%) as compared to the preclinical group (28%). The p value was significant ($p < 0.05$) for these observations (Table 3)

Higher prevalence of MRSA carriage was found in the clinical group (16%) as compared to the preclinical group (6%) but the probability value for these observations was not significant (Table 4).

DISCUSSION

The present study showed a 40% nasal carriage of *S. aureus* amongst medical students. An increasing trend of prevalence of *S. aureus* was seen over the academic years. The p value for these observations was significant ($p < 0.05$). Our results were comparable to those found by López-Aguilera who reported a *S. aureus* nasal colonisation rate of 39% among medical students. Various studies have reported *S. aureus* nasal carriage in medical students ranging from 10% - 39%.^{3,6,10-13}

The colonisation rate of MRSA in the present study was found to be 12.66% amongst the medical students. The nasal carriage rate of MRSA in international studies in students ranged from 0% - 6.7%.^{3,9-11,14} Amongst the Indian studies, a study by Bhaskaran reported a MRSA

colonization of 8.6% among medical students while another study by Shrivastav et al 2015 reported the MRSA carriage rate to be 1.1%, which is much lower than our study.^{6,15}

The present study showed no significant difference in carriage rate of *S. aureus* and MRSA among male and female students. This is in correlation with other studies³ who have also not shown significance of colonization in relation to the gender of students. However, Bischoff et al reported a higher prevalence of *S. aureus* colonization among male students.¹⁶

The prevalence of *S. aureus* was more in the clinical group (46%) as compared to the preclinical group (28%) suggesting that *S. aureus* colonisation increases with exposure of the student to the hospital environment. These observations were statistically significant ($p < 0.05$). Shrivastav et al 2015 reported the carriage rate of *S. aureus* to be 20% among the clinical group which was much lower as compared to our study (46%).⁶ Contrary to our study and other studies, Syafinaz et al observed a higher prevalence of *S. aureus* in the preclinical group as compared to the clinical group.² Similar was the case for MRSA carriage which was more in the clinical group (16%) as compared to the preclinical group (6%). Our results for MRSA were not significant for preclinical and clinical students ($p = 0.08$). Similar findings on the prevalence of nasal carriage of *S. aureus* and MRSA were obtained in Saudi Arabia, Agra and Michigan.^{3,6,10}

Eradication of MRSA colonisation from patients and healthy carriers has been tried with topical and oral antibiotics.¹¹ International guidelines recommends local therapy with Mupirocin to eliminate MRSA nasal colonisation in certain groups of patients and health-care workers colonized with MRSA. However, this antibiotic is only effective at removing the bacterium from the nose over a few weeks, and recolonisation often occurs after therapy is discontinued. Another drawback observed is the increasing trend in resistance of Mupirocin and the subsequent decrease in effectiveness.⁹

Further studies need to be carried out to find the efficacy of Mupirocin therapy by rescreening these MRSA positive students after treating them with Mupirocin and testing the MRSA strains for mupirocin resistance by disc diffusion method. At present the best option to avert further MRSA colonisation among medical students and other healthcare workers is to make them aware of the risks of colonisation *S. aureus* and MRSA and educate them about the standard hand washing protocol and safety precautions to be followed while handling patients.

CONCLUSION

This study shows that the nasal carriage of *Staphylococcus aureus* and Methicillin resistant *Staphylococcus aureus* (MRSA) among medical students

increases as their exposure to patient care increases. These students may act as vectors in transmitting the infection in the hospital and may also carry these notorious bacteria into the community. So, it is of utmost importance that the medical students should be made aware of this periodically during their clinical postings of MBBS course preferably at the inception. They should be educated through lectures and workshops on hand hygiene and other safety precautions during patient care. This will definitely control the carriage rate and infection rate in the hospital. Such awareness in the medical students will help in preventing these dreadful bacteria from being transmitted to the community.

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