

Received:
21 September 2018
Revised:
7 December 2018
Accepted:
28 January 2019

Cite as: Feyissa Efa,
Yared Alemu,
Getenet Beyene,
Esayas Kebede Gudina,
Wakjira Kebede. Methicillin-
resistant *Staphylococcus*
aureus carriage among
medical students of Jimma
University, Southwest
Ethiopia.
Heliyon 5 (2019) e01191.
doi: [10.1016/j.heliyon.2019.e01191](https://doi.org/10.1016/j.heliyon.2019.e01191)



Methicillin-resistant *Staphylococcus aureus* carriage among medical students of Jimma University, Southwest Ethiopia

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Abstract

Objectives: Infections caused by methicillin-resistant *Staphylococcus aureus* (MRSA) are often difficult to manage due to its resistance to multiple antibiotics. This study aimed to determine the nasal carriage of MRSA and its antimicrobial susceptibility patterns among medical students at the Jimma University medical center (JUMC), Southwest Ethiopia.

Methods: An institution based cross-sectional study was conducted at the JUMC from May to August; 2016. A total of 371 participants were systematically selected. Demographic data was collected using pre-designed questionnaire. Nasal swabs were collected following standard microbiological methods. MRSA was detected using cefoxitin (30μg) disc (Oxoid, UK); and antimicrobial susceptibility tests were performed by disc diffusion method.

Results: A total of 371 students were included. Of these, 84.9% (315/371) were males. The overall prevalence of nasal carriage of *S. aureus* and MRSA among

medical students at JUMC were 22.1% (82/371) and 8.4 % (31/371), respectively. The carriage rate of MRSA among medical intern (20% (16/80)) was higher compared with clinical year-I (3.6% (6/166)) and year-II (7.2% (9/125)) students. Resistance against trimethoprim-sulfamethoxazole, tetracycline and ciprofloxacin were 83.9%, 64.5% and 51.6%, respectively. Longer stay in hospital was significantly associated with the acquisition of MRSA ($X^2 = 6.93$, P value = 0.031).

Conclusion: The prevalence of nasal carriage of MRSA was high. Longer stay in hospital environment was associated with the acquisition of MRSA. These findings suggest that infection control efforts focusing the performance of antimicrobial stewardship could have a significant impact on MRSA incidence in this setting.

Keywords: Epidemiology, Health profession, Infectious disease, Microbiology

1. Introduction

Staphylococcus aureus (*S. aureus*) is a Gram-positive bacterium. The human skin, throat, and nasal nares are the most prevalent site known to asymptomatically colonize by normal flora of *S. aureus* [1, 2]. Colonized individuals are at increased risk for developing infections, which range from mild skin infections to severe diseases, such as endocarditis, bacteremia, sepsis, and osteomyelitis [3, 4, 5]. Infections caused by methicillin resistant *S. aureus* (MRSA) strain known to be resistant to β -lactam antibiotics present a growing problem in the world and often difficult to manage due to its resistance to multiple antibiotics [6, 7].

Previous studies in Ethiopia have reported 20.3% and 28.8% MRSA carriage among health professionals at Mekele and Dessie referral hospitals, respectively [8, 9]. Existing data from the country also revealed antimicrobial resistance rates for ampicillin and tetracycline of 89.9% and 86.1%, respectively [8]. However, there is no similar report among medical students on clinical attachment in hospitals in Ethiopia. Therefore, this study aimed to determine the rate of MRSA carriage and their antimicrobial susceptibility patterns among healthy clinical year medical students of Jimma University on clinical attachment at JUMC, Southwest Ethiopia.

2. Materials and methods

2.1. Study setting

The study was conducted at Jimma University Medical Center, a tertiary teaching hospital located in Jimma town, 352km southwest of Addis Ababa. It is the largest hospital in Oromia Regional State with over 500 inpatient beds and over 15 million catchment populations. In the year 2016, the hospital had 1448 employees most of

which were health professionals. In the same year, the hospital served as the only clinical attachment site for 750 medical students. The hospital served about 15,000 inpatients, 160,000 outpatient visits and 11,000 emergency cases in that year [10]. This medical center has been serving as a practical attachment site for health science and medical students from an Institute of Health.

2.2. Study design, period and population

An institution based cross-sectional study was conducted from May to August; 2016. All clinical year medical students who have a clinical exposure for eight hours per day in JUMC were included. The participants included in the study were divided into three groups: clinical year I, clinical year II and medical intern. The clinical year I and II group include 4th and 5th year medical students, respectively. Medical intern referred to the graduating class (6th year), who had at least three years clinical exposure at the JUMC. Participants who had nasal infection and received any antibiotic therapy in the last two weeks prior to data collection were excluded.

2.3. Sample size and sampling technique

A single population proportion formula was used to calculate the sample size of 187, including 10% non response rate, considering the proportion of MRSA nasal carriage rate from the previous study done somewhere else [9]. However, in order to increase the yield of relevant findings, the sample size became doubled and increased to 374. A clinical year medical students attending clinical practice at the JUMC were stratified based on their academic years and the sample size was allocated for each stratum followed by proportional allocation sampling technique as clinical-I, $n = 166$, clinical-II, $n = 125$ and medical intern, $n = 80$. The first student was randomly selected and then the systematic sampling technique was used to recruit the study participants in each stratum.

2.4. Data collection procedure

Demographic and other related information from consenting students were collected using pre-tested questionnaires. Self-sampling technique [11] was used to collect non-repeated samples by inserting a sterile and moistened swab into the nostril to a depth of about 1.5–2 cm and rotating it against the interior surface of the nostril. The sterile cotton swabs were moistened prior to sampling to avoid discomfort. All data collection was performed at the end of the academic year to ensure subjects in the clinical group had at least one solid year of hospital exposure. The swabs with samples were inserted into the bottles containing Amies transport medium (Oxoid, UK), then transported to the Microbiology teaching laboratory of Jimma University using a cold chain within 2 hours of collection by nurse data collector.

2.5. Laboratory procedures

2.5.1. Bacterial culturing

The inoculated Mannitol Salt Agar (MSA) (Oxoid, UK) with nasal swabs was incubated at 37 °C for 48 hours. Visual inspection of golden-yellow colonies on MSA indicated the presence of presumptive *S. aureus* due to mannitol fermentation. Colonies which showed consistent results of positive reaction for gram stain and catalase test and the tube coagulase test were phenotypically confirmed as *S. aureus* [12].

2.5.2. Antimicrobial susceptibility test

Antimicrobial test was performed on Muller Hinton Agar (oxoid, UK) using the disc diffusion method as previously described by Clinical Laboratory Standard Institute [12]. The Standard antimicrobial disks (oxoid, UK) used were penicillin G (10 units), ciprofloxacin (5 µg), clindamycin (2 µg), gentamicin (10 µg), erythromycin (15 µg), chloramphenicol (30 µg), ampicillin (10 µg), Ceftriaxone (30 µg), tetracycline (30µg) and Trimethoprine-sulfamethaxazole (25 µg) and ceftiofur (30 µg). Moreover, isolates with a zone of inhibition of ≤ 21 mm to ceftiofur (30 µg) were considered to be MRSA phenotypically [13].

2.5.3. Quality control

S. aureus ATCC 700699 and ATCC 25923 reference strains obtained from the Ethiopian Public Health Institute, Addis Ababa, were used as positive and negative controls respectively for phenotypic identification of *S. aureus*.

2.6. Data management and analysis

Every study participant had a unique identification number. The demographic, related information in the questionnaires and laboratory results in the laboratory registration book were entered into Epi Data Manager for consistency checks and data cleaning. These in turn were exported to SPSS software version 20 for analysis according to the objectives of the study. Continuous variables were categorized and described as proportions and also analyzed to compare the significance of difference in the distribution by using the Chi square test. The difference in distribution was considered significant if p value was less than 0.05.

2.7. Ethical consideration

The protocol was ethical cleared by IRB of Jimma University Institute of Health. An official letter from IRB was submitted to JUMC for approval before data collection. The purpose of the study was clearly described to the study participants. Written informed consent was obtained from the eligible participants before enrollment.

Clinical and laboratory data collected were kept confidential through anonymity. For those with MRSA colonization, treatment was recommended.

3. Results

3.1. Participant characteristics

A total of 371 students were enrolled in the study with the response rate of 99.2%. Of these, 84.9% (315/371) were males. Over 80% (299/371) of the participants were 25 years old or younger. With respect to category of the students, the clinical year-I students were 44.7% (166/371), clinical year-II students were 33.7% (125/371) and medical interns were 21.6% (80/371) [Table 1](#).

3.2. MRSA colonization rates

S. aureus carriage rate among clinical year medical students on clinical rotation at Jimma University Medical Center for at least eight hours daily in the last one year was 22.1% (82/371). The overall MRSA carriage rate was 8.4 % (31/371). Moreover, among *S. aureus* carriers, 37.8% (31/82) were MRSA carriers. MRSA carriage rate was higher among medical intern, 20% (16/80) than clinical year-II 7.2% (9/125) and year-I 3.6% (6/166) [Table 2](#).

3.3. Antimicrobial susceptibility profiles

The susceptibility test was done for commonly prescribed antibiotics by medical students to treat the infections caused by *S. aureus* including MRSA and MSSA in the

Table 1. Distribution of *S. aureus* according to sex, age and clinical year of the students on clinical attachment for at least one year at the JUMC from May to August, 2016.

Variables	<i>S. aureus</i> colonization (n = 371)		Total, n (%)
	Yes, n (%)	No, n (%)	
Sex			
Male	69 (21.9%)	246 (78.1%)	315 (100%)
Female	13 (23.2%)	43 (76.8%)	56 (100%)
Age (in years)			
≤25	62 (20.7%)	237 (79.3%)	299 (100%)
≥25	20 (27.7%)	52 (72.3%)	72 (100%)
Category of the students			
Clinical year I	28 (16.9%)	138 (83.1%)	166 (100%)
Clinical year II	23 (18.4%)	102 (81.6%)	125 (100%)
Medical Interns	31 (38.8%)	49 (61.2%)	80 (100%)
Total	82 (22.1%)	289 (77.9%)	371 (100.0)

Table 2. Distribution of MRSA according to sex, age and category of year among healthy medical students at JUMC from May to August, 2016.

Characteristics	MRSA colonization		Total, n (%)
	Yes (n = 31), n (%)	No (n = 340), n (%)	
Sex			
Male	28 (8.9%)	287 (91.1%)	315 (100%)
Female	3 (5.4%)	53 (94.6%)	56 (100%)
Age (in years)			
≤25	22 (7.4%)	277 (92.6%)	299 (100%)
≥25	9 (12.5%)	63 (87.2%)	72 (100%)
Category of the students			
Clinical year I	6 (3.6%)	160 (96.4%)	166 (100%)
Clinical year II	9 (7.2%)	116 (92.8%)	125 (100%)
Medical intern	16 (20%)	64 (80%)	80 (100%)

MRSA = Methicillin resistance *staphylococcus aureus*.

hospital setting. The susceptibility percentages of 82 *S. aureus* isolates to ten antimicrobial drugs were presented in Table 3. Of the MRSA isolates, 83.9% (26/31) were found to be susceptible to clindamycin, the drug of choice to treat the MRSA infections in this clinical setting. Susceptibility of these strains to erythromycin was 70.9%, for example, among MSSA isolates, the susceptibility rate to clindamycin,

Table 3. List of antimicrobial drugs used and susceptibility percentages of 82 *S. aureus* isolates, stratified by MRSA and MSSA strain.

Antimicrobial agents	Susceptibility profiles of <i>S. aureus</i> isolated			
	MRSA (n = 31)		MSSA (n = 51)	
	Resistance n, (%)	Susceptible n, (%)	Resistance n, (%)	Susceptible n, (%)
DN, Clindamycin (2μg)	5 (16.1)	26 (83.9)	0 (0.0)	51 (100.0)
E, Erythromycin (15μg)	9 (29.1)	22 (70.9)	5 (9.8)	46 (90.2)
CIP, Ciprofloxacin (5μg)	16 (51.6)	15 (48.4)	6 (11.7)	45 (88.3)
TE, Tetracycline (30 μg)	20 (64.5)	11 (35.5)	28 (54.9)	23 (55.1)
SXT, Trimethoprim-sulfamethoxazole (25 μg)	26 (83.9)	5 (16.1)	28 (54.9)	23 (55.1)
AMP, Ampicillin	-	-	51 (100.0)	0 (0.0)
PEN, Penicillin G	-	-	51 (100.0)	0 (0.0)
CN, Gentamicine	-	-	8 (15.7)	43 (84.3)
C, Chloramphenicol	-	-	3 (5.9)	48 (94.1)
CTX, Ceftriaxone	-	-	2 (3.9)	49 (96.1)

All antimicrobial susceptibility breakpoints interpretation is according to British society for antimicrobial chemotherapy recommendation [13].

ceftriaxone and chloramphenicol was found to be 100%, 96.1%, and 94.1%, respectively.

3.4. Multi-drug resistance patterns of *S. aureus* isolates

Multidrug-resistant status of *S. aureus* isolates was performed for ten antimicrobial agents commonly used in the setting. MRSA appear less frequently multi-resistant in comparison to MSSA. Among the total MSSA isolates; 52.9% (27/51) were multi-drug resistant. Of those, 37.0% (10/27), 33.3% (9/27) and 29.6% (8/27) were isolated from nasal nares of medical interns, clinical year-I and clinical year-II students, respectively. Among MRSA isolates; 48.4% (15/31) were multi-drug resistant. Of these, 60.0% (9/15) and 26.7% (4/15) was determined among medical interns and clinical-I student nasal swabs, but the differences was not statistically significant. As described in Table 4 in details. No MRSA isolates showed 100% susceptible to the five drugs used in this study.

3.5. Factors associated with the acquisition of MRSA

More than half of study participants who had been exposed to JUMC environments for more than two years were colonized with MRSA. Duration of the year in hospital environments during medical practice was statistically significant for the acquisition

Table 4. Resistance profiles of 82 *S. aureus* isolates from medical students at the JUMC from May to August 2016.

Resistance profiles	Antimicrobial resistance	Source of isolates			No of isolates, n = 82
		C-I	C-II	Interns	
MRSA					
MRSA I	SXT, TE, CIP, E, DN	1	-	1	2
MRSA II	SXT, TE, CIP E	2	-	1	3
MRSA III	SXT, TE, CIP,	2	1	7	10
MRSA IV	SXT, TE	1	3	2	6
MRSA V	SXT, TE	-	5	5	10
MDR-MRSA					15/31 (48.4%)
Total MRSA					
					31/82 (37.8%)
MSSA					
MSSA I	AMP,PEN,TE,SXT, CN	-	-	3	3
MSSA II	AMP, PEN, TE,SXT	2	4	3	9
MSSA III	AMP, PEN,TE	7	4	4	15
MDR-MSSA					27/82 (32.9%)

C-I = Clinical year-I, C-II = Clinical year –II, Interns = medical interns, MDR-MRSA = multidrug-resistant MRSA. MRSA isolates showed resistance to three or more antibiotics tested referred to as MDR-MRSA.

of MRSA ($X^2 = 6.93$, P value = 0.031). Students who were using gloves while handling patients, treated with any antibiotics for the last one year before data collection and those who were frequently decontaminate their hands using disinfectants were less colonized by MRSA, but there is no statistically significant association ($p = > 0.05$). MRSA carriage rate in percentage in relation to associated factors is presented in Table 5.

4. Discussion

The *S. aureus* and MRSA nasal carriage rate among medical students at JUMC was 22.1% and 8.4 %, respectively. The *S. aureus* colonization rate in this study is comparable with the similar studies from Taiwanese University (21.9%) [14] and Tanzania (21.0%) [15]. However, this finding is lower compared with report from Czech Republic (30%) [16]. Moreover, the overall prevalence of MRSA from this study is relatively high compared to previous studies reported from Saudi Arabia, Tanzania and Nepal were (6.7%), (1.5%) and (4%), respectively [15, 17, 18]. The difference might be due to the variation in infection control and prevention policies across countries, the variation in perception and the reality of student's knowledge about MRSA epidemiology, the extent of risky environmental exposures and different geographical locations.

Table 5. Factors associated with the acquisition of MRSA among *S. aureus* colonized medical students on clinical attachment at JUMC from May to August, 2016.

Variables	Total number	MRSA colonization		P-value
		Yes (n = 31), n (%)	No (n = 51), n (%)	
Using gloves while handling a patient				
Yes	75	27 (36.0)	48 (64.0)	0.480
No	7	4 (57.1)	3 (42.9)	
Hand decontamination score				
High	11	4 (36.4)	7 (63.6)	0.41
Moderate	36	11 (30.6)	25 (69.4)	
Low	35	16 (45.7)	19 (54.3)	
Antibiotic use in the past one year				
Yes	8	4 (50.0)	4 (50.0)	0.71
No	74	27 (36.5)	47 (63.5)	
Duration of exposure to hospital environment				
<1year	27	5 (18.5)	22 (81.5)	0.03
1–2 years	24	10 (41.7)	14 (58.3)	
>2 years	31	16 (51.6)	15 (48.4)	

High decontamination score = always using sanitizers, Moderate score = always using water, soap with sanitizers sometimes, Low score = always using water only or water with soap sometime.

In relation to the category of the students, the present study found that a higher proportion of MRSA nasal carriage among medical intern students compared with clinical year I and II student. The variation could be due to the fact that medical intern students had a longer exposure in the hospital environments and frequent visits the wards and clinics to provide care to the patients where MRSA may more prevails. These may be attributed to increase in the possibility of *S. aureus* exposure and subsequent acquisition of MRSA.

The antimicrobial resistance profiles to the most commonly prescribed drugs such as ampicillin, penicillin and tetracycline and trimethoprim-sulfamethoxazole as opposed to less commonly used drugs like clindamycin and erythromycin shows increased resistance. The findings of this study was comparable to many other similar studies reported from outside of Ethiopia [15, 19]. Furthermore, all MSSA isolates identified in this study were completely resistant to ampicillin and penicillin. On the other hand, it is 100%, susceptible to clindamycin. More than half of MSSA isolates were developing resistance to tetracycline and trimethoprim-sulfamethoxazole. The finding was comparable with the previous studies reported from Ethiopian health care works who have equal exposure length of time with the students in a hospital environment [8, 9].

A multi-drug resistance profile in this study was taken as resistance to three or more antimicrobial drugs listed in Table 4. Approximately, half of *S. aureus* isolates were multi-drug resistant. MRSA isolates were resistant to three and more antimicrobial agents out of five selected drugs that commonly used to treat infections caused by MRSA. No resistance was reported with clindamycin in this study among MSSA isolates. That is good because this normal flora can protect individuals against pathogenic bacteria [3]. Therefore, visible measures should be focused on MRSA surveillance to prevent transmission in a hospital setting, particularly in resource-limited countries like Ethiopia [8, 9]. Resistant to TMP-SMX among MRSA isolates in this study was prevalent, this is also common in other African countries [20] especially, at high human immunodeficiency virus (HIV) areas [21]; because TMP-SMX drug was used as prophylactic therapy, this may increase antimicrobial resistance stewardship.

In this study, a longer hospital stay was prone the students to MRSA colonization. We found that significant association between long stays in clinical practice with the acquisition of MRSA among medical students. The highest proportion in the acquisition MRSA was seen among medical intern (6th-year) students who were spent more than two years in the hospital compared with clinical year-I (4th-year) medical students who were spent less than one year on clinical practice. The results were comparable with previous studies from Tanzania [15], Taiwan [14], Saud Arabia [17]. This might be due to the similarity of length of stay in hospital, frequent visits of wards and clinics, and the rate of the patient load. Moreover, during data

collection we witnessed that clinical year-I students were more adhered to basic infection prevention precautions, after contact with patients than clinical year-II and Medical intern students. However, there was no statistically significant association with the using of gloves while handling a patient, hand decontamination score and antibiotic usage in the past one year with the acquisition of MRSA among medical students.

The limitation of this study was the lack of investigating vancomycin resistance profile among the isolates. However, the strength of this study was clearly described that there is a high prevalence of MRSA and MSSA with their multi-drug resistance patterns among medical students, and this is a risk for the students, their colleagues and as well as for the patients under their care. Therefore, in view of these results, it is important to understand the perceptions and the reality of student's knowledge on epidemiology of MRSA in this hospital setting.

5. Conclusion

The prevalence of nasal carriage of *S. aureus* and MRSA in this study was high. The higher MRSA carriage rate was determined among medical interns than clinical year-I and II students, with more than half of *S. aureus* isolates were multi-drug resistant. Longer stay in hospital environment was significantly associated with the acquisition of MRSA colonization. These findings suggest that infection control efforts focusing the performance of antimicrobial stewardship could have a significant impact on MRSA incidence in the hospital setting.

Declarations

Author contribution statement

Feyissa Efa: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data.

Yared Alemu, Getenet Beyene: Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data.

Esayas Kebede Gudina: Analyzed and interpreted the data.

Wakjira Kebede: Conceived and designed the experiments; Analyzed and interpreted the data; Wrote the paper.

Funding statement

The study was supported by Jimma University.

Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

Acknowledgements

The authors would like to thank officials and study participants of the study for their cooperation during data collection by providing and/or facilitating collection of valuable information. Ethiopian Public Health Institute, Addis Ababa, is acknowledged for provision of reference strains.

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