

NASAL COLONISATION OF METHICILLIN & MUPIROCIIN RESISTANT *STAPHYLOCOCCUS AUREUS* AMONG INTERNS OF TERTIARY CARE HOSPITAL SET UP.



Pharmacology

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ABSTRACT

Aim: To determine the nasal colonisation of methicillin resistant *S. aureus* among the interns posted in indoor-patient wards of the tertiary care hospital set up.

Material and method: The present study was a prospective observational study carried out in the Department of Microbiology among the interns posted in different indoor-patient wards of tertiary care hospital. The swab was inserted approximately 2 cm and rotated for 3 sec in the anterior nares and then re-inserted in transport tube. After collecting swabs, they were labelled properly, and transported to the laboratory. Methicillin resistance among *S. aureus* was determined using cefoxitin 30 µg discs by Kirby–Bauer disc diffusion method as per Clinical and Laboratory Standards Institute (CLSI) 2016 guidelines. After incubation, zone of inhibition was measured and the isolates were categorized as sensitive, low-level resistant and high-level resistant.

Results: Overall *Staphylococcus aureus* was isolated in 44% of the samples. Out of 44 *S. aureus* isolates, 23 (23%) were detected as MRSA strains. Nursing orderlies (13%) showed a higher carrier rate of MRSA as compared to other HCWs (10%). Maximum resistance was reported for ciprofloxacin for both MRSA (78.2%) and mupirocin-resistant MRSA isolates (88.2%).

Conclusion: The results of the present study will help to understand the mode of transmission of infection through the health care personnel such as interns.

KEYWORDS

Health care workers; methicillin-resistant *Staphylococcus aureus*; nasal carriage

INTRODUCTION:

Nosocomial infections are a serious cause of concern worldwide, especially in developing countries like India. Various agents are responsible, with *Staphylococcus aureus* being one of the commonest among them¹. The danger due to *S. aureus* infection increases many fold when they became resistant to methicillin. Such strains are called as Methicillin-resistant *S. aureus* (MRSA)².

S. aureus/MRSA is a part of commensal flora and it most commonly colonises in the anterior nares. Thus health care workers colonising MRSA become the potential source of infection to the patients. Development of mupirocin resistance among *S. aureus* left the infection control committee with few options to decolonise³. Mupirocin, a topical glycopeptide antibiotic is now commonly used for nasal decolonization of the HCWs as part of a routine surveillance to prevent the emergence and transmission of MRSA in health care facilities. The increased pressure of antibiotic use has led to the emergence of mupirocin resistant *S. aureus* (MupRSA), and the clinicians are left with few alternatives to prevent the spread of MRSA⁴.

The health care workers (HCWs) serve as a link between hospitals, long-term care facilities, and nursing homes on one hand and the community on the other. They may serve as reservoirs, vectors, or victims of MRSA cross-transmission⁵. In India the increasing prevalence of health care-associated MRSA (HA-MRSA) has been documented by several studies which showed its range from 17.4%-49%^{6,7}. Although vancomycin is currently used as a drug of choice for treatment of MRSA infections, but emergence of vancomycin-resistant *S. aureus* (VRSA) has limited the treatment options⁸.

Interns among the HCWs might crucially transmit the MRSA infections throughout the hospital as they are posted in different wards from time to time. Thus identifying the carrier state among them is important in limiting the infection. To the best of our knowledge, in our region there is no information on the methicillin & mupirocin resistant *Staphylococcus aureus* among interns. The aim of the study was to determine the nasal colonisation of methicillin resistant *S. aureus* among the interns posted in indoor-patient wards of the tertiary care hospital set up.

METHODOLOGY:

Study design: The present study was a prospective observational study carried out in the Department of Microbiology.

Ethical considerations: The proposed study was approved by the Ethical Committee of the institute.

Study population: The interns posted in different indoor-patient wards of tertiary care hospital set up were included for the present study.

Sample size: 100 non-duplicate samples.

Selection criteria: Following inclusion and exclusion criteria was used:

• Inclusion criteria:

1. Interns posted in different indoor-patient wards.

• Exclusion criteria:

1. Interns taking antibiotics during past week.
2. Interns underwent hospitalisation during last month.
3. Interns not posted in indoor-patient wards.

Sample collection:

After obtaining written informed consent from each intern, nasal swab was collected using a sterile cotton swab. The swab was inserted approximately 2 cm and rotated for 3 sec in the anterior nares and then re-inserted in transport tube. After collecting swabs, they were labelled properly, and transported to the laboratory.

Sample processing:

Nasal swab was inoculated on 5% blood agar and a selective medium, mannitol salt agar (MSA) using a sterile inoculating loop and incubated at 37°C for 24 h. After incubation, the media was observed for growth and on the basis of colonial morphology, Gram staining features and below mentioned biochemical test was identified as *S. aureus*. Biochemical test performed were catalase, coagulase (slide and tube), dimethyl sulfoxide oxidase and DNase test.

Determination of methicillin-resistant *Staphylococcus aureus*:

Methicillin resistance among *S. aureus* was determined using cefoxitin 30 µg discs by Kirby–Bauer disc diffusion method as per Clinical and Laboratory Standards Institute (CLSI) 2016 guidelines⁹. Inoculum was prepared in 4 ml sterile saline to obtain a suspension of 0.5 McFarland standards. Mueller Hinton agar (MHA) was inoculated and incubated at 37°C for 24 h. After incubation, zone of inhibition was measured by unaided eye and size of ≤21 mm was taken as resistant and ≥22 mm as sensitive.

Determination of mupirocin-resistant *Staphylococcus aureus* (MupRSA): Mupirocin resistance among MRSA strains was determined by using 5 µg and 200 µg mupirocin discs by Kirby–Bauer disc diffusion method. Isolates to be tested was inoculated on MHA culture medium and both the discs was applied and incubated at 37°C for 24 h. After incubation, zone of inhibition was measured and the isolates were categorized as sensitive, low-level resistant and high-level resistant. Isolates with zone > 14 mm for both 5 µg and 200 µg discs was considered as sensitive, those with zone < 14 mm for 5 µg but > 14 mm for 200 µg disc was considered as low-level resistant (MuL), and those with zone < 14 mm for both 5 µg and 200 µg discs was considered as high-level resistant (MuH).

Determination of antibiotic susceptibility pattern of MRSA and MupRSA: It was done by Kirby–Bauer disc diffusion method with the help of various antibiotic discs i.e. Penicillin (10 units), oxacillin (1 µg), ciprofloxacin (5 µg), levofloxacin (5 µg), clindamycin (2 µg), linezolid (30 µg), tetracycline (30 µg) and cotrimoxazole (1.25/23.75 µg). After incubation, zone of inhibition was measured by unaided eye and interpretation was determined as sensitive, intermediate or resistant as per CLSI guidelines⁹.

STATISTICAL ANALYSIS:

Data so collected was tabulated in an excel sheet, under the guidance of statistician. Data was analyzed using IBM SPSS. Statistics Windows, Version 20.0. (Armonk, NY: IBM Corp) for the generation of descriptive and inferential statistics. The statistical significant difference among groups was determined by the Chi square test and the level of significance was set at $p < 0.05$.

RESULTS:

In the current research, 100 interns working in the hospital were included in the study. Out of these interns, 55 (55%) were females and 45 (45%) were males. The age ranged between 18 and 30 years. Of the 100 non-duplicate nasal swabs processed in the laboratory, *Staphylococcus aureus* was isolated in 39.62% and 48.94% of doctor and nursing interns sample's respectively with statistically insignificant difference. Out of 44 *S. aureus* isolates, 23 (23%) were detected as MRSA strains. Nursing orderlies (13%) showed a higher carrier rate of MRSA as compared to other HCWs (10%) which was statistically insignificant ($P < 0.05$) as shown in table 1.

Table 1: Distribution of nasal swabs growing *Staphylococcus aureus* and MRSA and MSSA isolates on the basis of source

Intern	No. of samples	Number of S. aureus		Chi square	p value
		N	%		
Doctor students	53	21	39.62	0.88	0.35
Nurses students	47	23	48.94		
Total	100	44	44		
Source	MRSA	MSSA			
Doctor students	10	11		0.35	0.56
Nurses students	13	10			
Total	23	21			

High level mupirocin resistant (MuPR) *Staphylococcus aureus* was found a little higher by E test and agar diffusion method (52.1%) as compared to disc diffusion testing method (47.8%). 56.5% and 58.8% of MRSA and mupirocin-resistant MRSA isolates was sensitive to penicillin respectively. Nearly 39% and 41% MRSA and mupirocin-resistant MRSA isolates were resistant to cotrimoxazole respectively. MRSA and mupirocin-resistant MRSA isolates resistant to linezolid were 21.7% and 23.5% respectively. Maximum resistance was reported for ciprofloxacin for both MRSA (78.2%) and mupirocin-resistant MRSA isolates (88.2%) as shown in table 2.

Table 2: Mupirocin resistant (MuPR) *Staphylococcus aureus* among MRSA isolates along with antibiotic resistance

Methods	Low level MupR		High level MupR	
	N	%	N	%
Disc diffusion testing	05	21.7	11	47.8
E test and agar dilution	05	21.7	12	52.1
Chi square	0.01			
p value	0.91			
Antibiotics	MRSA (%)		MuPR (%)	
Penicillin	56.5		58.8	

Linezolid	21.7	23.5
Oxacillin	32.5	38.4
Levofloxacin	31.6	34.2
Tetracycline	40.4	50.8
Cotrimoxazole	39.1	41.1
Cefuroxime	52.1	68.8
Clindamycin	46.7	48.4
Ciprofloxacin	78.2	88.2

DISCUSSION:

The prevalence of *S. aureus* nasal carriage among HCWs is 44% in the present study which is higher than the study conducted by Truong et al¹⁰ (35.8%) and Yazgi et al¹¹ (34.9%) and comparable to other studies conducted by Singh et al¹² (47.5%) and Al-Abdli and Baiu¹³. Norazah et al¹⁴ in a study conducted in Malaysia in 2002, reported a higher prevalence of *S. aureus* nasal carriage which varied from 45% to 76%.

In our study, nasal carriage of MRSA was found in 23% of the HCWs. This is little higher with the study conducted in different hospital setting worldwide which has been reported in the range of 5.8% to 17.8%¹⁵⁻¹⁶. Lower percentage of MRSA carriage has been reported from Nepal¹⁷. This difference in prevalence of *S. aureus* and MRSA in various hospitals may be attributed to the inter-laboratory variation in the methods of detection as well as the effectiveness of hospital infection control policy.

In the present study, the MRSA carriage was particularly high among the nursing orderlies. This result was similar to the findings reported by Loveleena Agarwal¹⁸. The higher prevalence of MRSA among this group of HCWs could be due to their lack of knowledge with regard to hand hygiene, contact precautions, and infection control policies.

Nasal formulation of mupirocin, a topical antibiotic agent that interferes with bacterial protein synthesis, is recommended by the Food and Drug Administration of United States for use in the eradication of nasal carriage of *S. aureus* in adult patients and HCWs. Though the use of mupirocin is limited for infection control and other prophylactic measures, emergence of MupRSA is being reported from across the globe with the prevalence of 0.5% in Nigeria to 14.6% in India^{19,20}. In our study, 16 (16%) and 17 (17%) isolates were found to be mupirocin resistant by E test and agar diffusion and disc diffusion testing method respectively of which 11 and 12 isolates respectively were high levels resistant. Loveleena Agarwal¹⁸ reported 4 (2%) isolates were found to be mupirocin resistant of which three isolates were high levels resistant. In the presence of mupirocin resistance strains, treatment with mupirocin may be ineffective, especially with high-level resistance strains. Although low-level mupirocin resistant strains can be controlled by normal dosage schedule of mupirocin but few studies suggest that treatment failure may occur after few weeks. This emphasizes the importance of identification of both high and low-level resistant strains²¹.

MRSA nasal colonization of HCWs in our hospital is high, particularly among the nursing orderlies, who are in prolonged contact with the patient leading to the high possibility of nurse-to-patient transmission of these bacteria and dissemination of them in hospital setting. As a routine, screening and treatment of HCWs should be done for MRSA status in every hospital. It is also advisable to detect mupirocin resistance among the isolates obtained from the HCWs so that in case of resistance, alternative treatment options should be initiated.

CONCLUSION:

There is no substantial data in this region regarding the nasal colonisation of MRSA and mupirocin resistant *S. aureus* among interns. The present study will help to understand the mode of transmission of infection through the health care personnel such as interns. It will also create awareness among the clinicians regarding the resistant to most commonly used agent of decolonisation of MRSA such as mupirocin. The student will be able to understand and learn about nosocomial infections, method of sampling, isolation and identification of MRSA and mupirocin resistant *S. aureus*.

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