

## Prevalence of nasal carriage of Methicillin Resistant *Staphylococci* in the asymptomatic healthcare workers, at a tertiary care hospital of Pakistan

Alavia Batool<sup>1</sup>, Maheen Imdad Chaudhry<sup>1</sup>, Hafsa Anjum<sup>1</sup>, Faisal Hanif<sup>2</sup>, Nadia Tayyab<sup>2</sup>, Amina

1. National University of Science and Technology.
2. Army Medical College, Rawalpindi.

Corresponding Author: Alavia Batool, Student of BS Applied Biosciences (ASAB), National University of Sciences and Technology (NUST). Email: [alavianaqvi@gmail.com](mailto:alavianaqvi@gmail.com)

### Abstract

The presence of Methicillin Resistant *Staphylococci* is very common in hospital environment. It includes, *Staphylococcus aureus* and Coagulase Negative *Staphylococci* (MRSA and MRCoNS). These are likely to cause deadly nosocomial infections in immunocompromised patients. They can be transmitted by direct contact through the Healthcare workers to the hospitalized patients.

**Aim:** To know the nasal carriage prevalence of Methicillin Resistant *Staphylococci* among asymptomatic healthcare workers in a tertiary care hospital of Pakistan.

**Methodology:** Nasal swabs were collected, cultured and tested for MRCoNS and MRSA by the clinical and laboratory standards institute (CLSI) protocols using Blood agar medium. Later Gram stain, disc diffusion method with cefoxitin and DNase test were performed as per CLSI standards.

**Results:** A total of 200 nasal swabs were collected and processed for culture from the healthcare workers. Out of 188 plates showing bacterial growth, 140 (74.6%) were Gram positive cocci & 48 (25.4%) were gram negative rods. All the gram-positive cocci showed positive catalase reaction. Coagulase testing was then performed to differentiate between *CoNS* and *S. aureus*. Ninety-six samples (68.6%) were found to be coagulase negative (CoNS) and 44 (31.4%) were coagulase positive. Sensitivity pattern showed that 14/96 and 6/44 among identified CoNS and *S. aureus* respectively were methicillin resistant.

**Conclusion:** The prevalence of MRCoNS exceeds the number of MRSA positive scenario among the healthcare workers but is still in a high ratio.

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## Introduction

Staphylococci are cluster-forming, gram-positive bacteria that are categorized into two major groups termed as coagulase-positive and coagulase-negative staphylococci (CoNS) based on the coagulase testing (1). Some staphylococci are commensals and some are colonizers of human skin (1). However, few of these skin-dwelling staphylococci have opportunistic nature and orchestrate pathogenesis in the hosts (1). *Staphylococcus aureus* resides on human skin, nares, and other mucous sites asymptotically (2), but can cause serious infections when invades the host as it possesses various virulence factors (1). *S. aureus* acquires methicillin resistance due to the formation of a mutant protein and acquires more lethality (3), inducing serious morbidity in the patients because it gets resistant to many commonly used antibacterials (4). *S. aureus* is the leading cause of nosocomial infections and its resistance to methicillin makes it clinically more significant (4). MRCoNS although not as virulent as *S.aureus*, also have potential to cause serious infections when they enter sterile body cavities especially in patients with implanting devices and have become one of the leading causes of hospital- acquired infections which is why they are also a clinical concern (6) (7).

Many multidrug resistant organisms are mostly transmitted through the hands of the healthcare workers (8). MRSA and MRCoNS are also transmitted through the contaminated hands of a healthcare personnel (5) (9) (10). The patient database of a tertiary care hospital in Pakistan was reviewed for the bacteria isolated from blood culture specimens of indoor patients. The data from the January 2021- August 2021 was collected and a significant number of the isolates were either MRSA or MRCoNS. A survey was conducted among the healthcare workers of a tertiary care hospital to know the prevalence of the MRSA and MRCoNS in the clinical staff. A very high rate of these infections motivated this research which aims at investigating the prevalence of these bugs in a tertiary care hospital. Hand hygiene and following standard infection prevention and control practices can eliminate the risk of their transmission from healthcare workers to immunocompromised patients.

## Methodology

The study was conducted at a tertiary care hospital of Pakistan. A special permit application was drafted and issued to the Heads of the respective departments and the sampling of the staff was initiated after their approval. The duration of this study was from 1<sup>st</sup> August till 31<sup>st</sup> December, 2021. Nasal swabs were taken from the anterior nares of 200 health care workers (HCWs) of different wards including adult ITCs, Neonatal Intensive Care Unit (NICU), Liver Transplant Unit, Accident & Emergency Detention, Gynecology wards, Operation Theater, Microbiology lab and Female Medical ward. It was descriptive cross-sectional study with non-probability consecutive sampling. Nasal samples (swabs) received in laboratory were collected, cultured and tested for MRCoNS and MRSA by the standard clinical and laboratory standard institute (CLSI) protocols using Blood agar medium. All the gram-positive cocci were tested for catalase reaction and Coagulase testing was then performed to differentiate between CoNS and *S. aureus*. Repeat specimens from same person were excluded from the study.

These samples were inoculated on the sheep blood agar and were incubated at  $35 \pm 2$  °C for 24 hrs. Identification of isolates was done by standard microbiological methods including colony morphology, Gram staining and biochemical reactions. Gram-positive cocci, positive for catalase, DNase and slide coagulase tests were considered as *S. aureus*. Furthermore, 0.5% McFarland suspension of isolated CoNS and *S. aureus* was inoculated on Muller-Hinton Agar

for detection of methicillin resistance by using cefoxitin 30 ug disc as surrogate marker as recommended by the Clinical and Laboratory Standard Institute (CLSI) guidelines. The data was analyzed by SPSS software.

**Results:**

In this study 200 healthcare workers participated. Out of these 200 workers, 94 were males and 106 were females. The participants were further divided based on their profession; 40 were doctors, 84 were nurses & 76 were lab technicians. All the candidates had clinical roles in the respective departments of the hospital.

Dept.	sam ples	(M) ♂	(F) ♀	Profession based division		
				Doc tors	Nur ses	Tec hnic ians
NICU	34	12	22	12	18	04
LTU	24	16	8	-	16	08
A&E	28	14	14	08	16	04
Lab	26	26	-	-	-	26
OT	26	26	-	-	-	26
Gyn	62	-	62	20	34	08
<b>Total</b>	<b>200</b>	<b>94</b>	<b>106</b>	<b>40</b>	<b>84</b>	<b>76</b>

**Table 1**

NICU: Neonatal Intensive Care Unit

LTU: Liver Transplant Unit

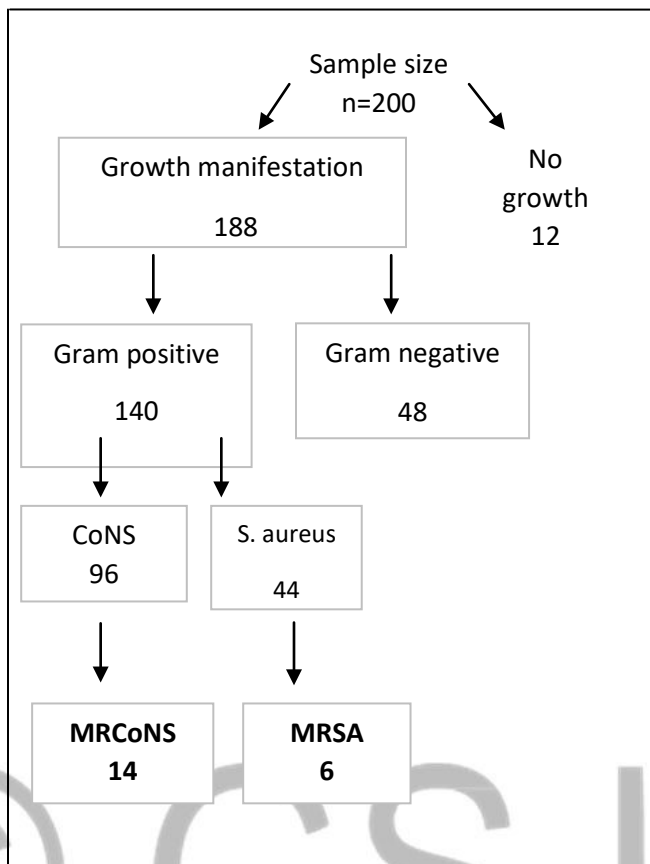
A&E: Accident & Emergency (detention)

OT: Operation Theatre

Gyn: Gynae Wards

Samples were collected from the anterior nares of the healthcare workers by sterile nasal swabs. These swabs were then inoculated on Blood agar medium for isolation of staphylococci. Out of 200 samples, growth was observed on 188 plates and 12 inoculations showed no growth. Golden yellow colonies were seen on the agar plates manifesting the characteristic feature of *S. aureus*. Out of 188 plates showing bacterial growth, 140 (74.6%) were Gram positive cocci & 48 (25.4%) were gram negative rods. All the gram-positive cocci showed positive catalase reaction. Coagulase testing was then performed to differentiate between CoNS and *S. aureus*. Ninety-six samples (68.6%) were found to be coagulase negative (CoNS) and 44 (31.4%) to be coagulase positive. Confirmation of *S. aureus* was done by using the DNase agar.

Kirby-Bauer Disk Diffusion Susceptibility Testing was carried out to detect bacterial resistance to methicillin. Cefoxitin disk was placed on the Mueller Hinton agar and plates were incubated for 18-24 hrs. at  $35 \pm 2^\circ$  C. The isolates with zones  $\leq 24$  mm and  $\leq 21$  mm as per recommended by the CLSI guidelines were considered methicillin-resistant CoNS (MRCoNS) and *S. aureus* (MRSA) respectively (11). Out of total 96 CoNS positive samples, 14 were declared methicillin-resistant and out of 44 *S. aureus*, 6 were found MRSA. The information of the HCWs was kept confidential and the workers tested positive for MRSA and MRCoNS were prescribed to use mupirocin ointment to anterior nares twice daily along with chlorhexidine gluconate bath once daily for five days followed by repeat testing and they were advised to strictly follow the hand hygiene practices.



The present study showed that the prevalence of methicillin-resistant isolates of *Staphylococcus* spp. is a health concern and invites an investigative study to evaluate the hand hygiene practices being followed by clinical staff.

A questionnaire was also circulated among these healthcare workers to evaluate their knowledge and expertise regarding the standard hygiene protocols and patientcare. The results of the questionnaire, however, did not match up with the experiment result. For instance, 100% of the candidates were literate in terms of hand-hygiene and infection control procedures, yet the practice frequently contradicted application of this knowledge. The major limitation of this research is that it is based on data collected from only one hospital. Maximum samples were collected from the clinical staff at Gynecology wards because the incidence of MRSA & MRCoNS infection from there was found to be maximum.

### **Conclusion:**

The present study showed the prevalence of methicillin-resistant strains of Staphylococcus species in the nasal swab samples of health-care workers. 56% of the CoNS isolated from these nasal samples was found to be resistant to methicillin. Out of the total sample size, there was significant prevalence of MRCoNS (27%) and MRSA (10%). These resistant strains are contagious and pose a threat to patients. Methicillin-resistant staphylococcus is generally resistant to a wide range of antibiotics and are sensitive to vancomycin.

### **Conflicts of Interest:**

None.

### **Source of funding:**

None.

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