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A surveillance of MRSA nasal carriage in Community and Health Care Workers

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Abstract

Staphylococcus aureus (S. aureus) is both a human commensal & an important pathogen in disease. It is a frequent cause of infections in both community and hospital. Being the principal habitant in human nares, S. aureus is known to disseminate to various body parts through hands, contributing to its infectious epidemiology. The current study was designed to evaluate the frequency of nasal carriage by Methicillin Resistant S. aureus (MRSA) in healthy volunteers of Mirpur-Khas region. Both, Community Associated Population (CAP) and Hospital Associated Population (HAP) were targeted. A total of 207 nasal swab samples were processed for the isolation of S. aureus. 27% (n=56) of the samples were positive for S. aureus, of which 59% (n=33) were identified as MRSA strains. The OR for nasal carriage by S. aureus and colonization by MRSA in HAP was calculated to be 4 and 12.05, with statistically significant p-values as (P = 0.0001) and (P = 0.001) respectively. The OR for nasal carriage by S. aureus and colonization by MRSA in male population was calculated to be 0.77 and 1.04, with p-values as (P = 0.194) and (P = 0.938) respectively, suggesting statistically insignificant evidence for gender association.

Key words: MRSA, Nasal Carriage, Community, Hospital.

Abbreviations: HAP = Hospital Associated Population, CAP = Community Associated Population, OR = Odds Ratio, MRSA = Methicillin Resistant *S. aureus*, MSSA = Methicillin Sensitive *S. aureus*

Introduction

Staphylococcus aureus (S. aureus) are gram positive cocci that behave both as commensals (Kluytmans et al 1997) and widespread pathogens (Wertheim et al 2005). They are frequently associated with hospital acquired infections. Infections due to *S. aureus* are increasingly being reported around the world (Boucher & Corey 2008). In many cases, the infections originate from hospital acquired antibiotic resistant *S. aureus*. The methicillin resistant *Staph. aureus*

(MRSA) are the most common accounting for about 50% of hospital acquired infections in many countries (Khan et al 2014). In Pakistan, the infections due to MRSA have greatly increased over the years. In 1989, 5% cases were reported, then a dramatic increase up to 40% has been witnessed (Hafiz et al 2002), (Perwaiz et al 2007). *S. aureus* are the common inhabitants of various body sites (Wertheim et al 2005), and are most prevalent in anterior nares. In-effect the nares provide the principal reservoir for these organisms (Rongpharpi et al 2013).

Studies have established that nasal carriers of *S. aureus* have an increased risk of acquiring an infection with this pathogen (Gupta et al 2013), (Wertheim et al 2004)(Herwaldt et al 2004). A sequence of events have also been postulated for the initiation of infection with nasal carriage, where by the organisms are disseminated via hands(Gebreyesus et al 2013) to other body sites where infections can occur (Wenzel & Perl 1995). A number of cross-sectional and longitudinal studies have been undertaken to explore the nasal carriage rate in patients, hospital staff, and healthy persons (Eriksen et al 1995). Consistent studies have reported that the carriage of *S. aureus* in the anterior nares is an important human reservoir for *S. aureus*(Kluytmans et al 1997), (Wenzel & Perl 1995).

The nasal carriage rate of *S. aureus* resistant to methicillin has never been studied for Mirpur-khas region of province Sindh. The current study was undertaken to investigate the frequency of the nasal carriage of methicillin resistant *S. aureus* in Mirpur-khas region, and to study the association of MRSA carriage with health care associated population.

Healthy volunteers working in hospital associated and non-hospital associated environment were included in the study. A comparative analysis was done to understand the current trends in nasal carriage for MRSA. Statistical analysis was done to evaluate the levels of significance for MRSA carriage between HAP and CAP.

Materials and Methods

Chemicals and Media

The media such as Manitol Salt Agar, Sheep Blood Agar, Muller Hinton Agar, Nutrient Broth were purchased from Oxide. Ames Transport Medium Swabs were purchased from Cito while the oxacillin discs were purchased from oxide.

Methodology

Mirpur-Khas is fourth largest city in the province Sindh of Pakistan and with estimated population of about 488,590= (2009). To ensure satisfactory

representation of the city, the samples of healthy volunteers were randomly collected from various private/government hospitals, pathological labs, and from residential and commercial areas. The study duration was one year from January 2015 to January 2016. A total of 207 nasal swabs were collected using sterile swabs. The samples were immediately brought to laboratory and inoculated on Mannitol Salt Agar and Sheep Blood Agar. The suspected colonies of *S. aureus* were further identified microscopically, and by biochemical tests. The antibiotic sensitivity testing against methicillin was done using *oxacillin* discs by standard *Kirby-Baur* disc diffusion method.

Collection of Nasal Swabs

The nasal swab samples were aseptically collected from both nares. The swabs were inserted in both nares up-to 5 cm and rotated gently about four-five times in nares (Konvalinka et al 2006) (Nouwen et al 2004) and put into transport medium for further processing.

Antibiotic Sensitivity Test

Antibiotic sensitivity testing was done using standardized *Kirby-Bauer* Disc Diffusion method. All *S. aureus* isolates were tested against *oxacillin* antibiotic. Overnight grown bacterial culture was diluted to $OD_{600} = 0.5$. 500 μ l of the diluted culture was inoculated on Muller Hinton Agar and spread evenly using sterile cotton swab. The antibiotic discs were placed on the agar surface and pressed gently to attain even contact with the agar surface. The plates were then incubated at 37°C for 24 hours. Clear zones (zones of inhibition) around the discs were noted and measured according to Clinical and Laboratory Standard Institute (CLSI).

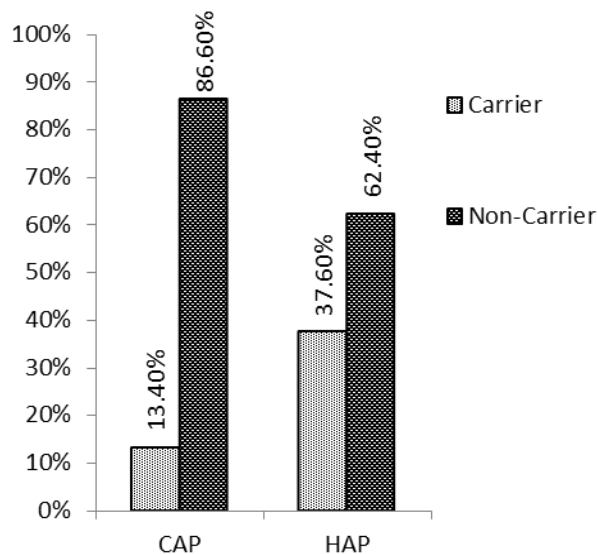
Statistical Analysis

IBM SPSS version 20 was used for data analysis. P-values were calculated using a chi squared test. The ORs were calculated manually.

Results

Prevalence of nasal carriage relies on hospital exposure.

Assuming if the prevalence of nasal carriage had nothing to do with the hospital exposure we processed about 207 nasal swab samples. Healthy volunteers working in Hospital environment and Non-hospital environment were screened. The samples were collected irrespective of gender and ethnicity. 27% (n=56) of the samples were positive for *S. aureus*. The *S. aureus* nasal carriage for CAP and HAP was calculated to be 13.4% and 37.6% respectively (Figure 1). The OR for carriage in hospital environment was calculated to be 4. A chi-squared test using IBM SPSS version 20 was also performed to get P-values (P=0.0001). This difference by conventional criteria is considered to be extremely statistically significant.

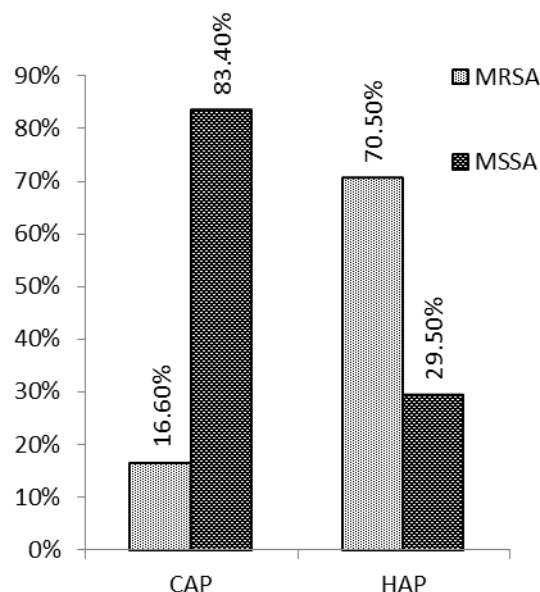


		Population	
		HAP	CAP
Carriage	Carrier	44	12
	Non-Carrier	73	78
	Total	117	90

Figure 1: (Top) Bar diagram displaying the percentage of *S. aureus* nasal Carriage among CAP and HAP.(Bottom) Population wise *S. aureus* nasal carriage.HAP = Hospital Associated Population, CAP = Community Associated Population

MRSA nasal colonization is dependent on hospital exposure.

To test if the nasal colonization by MRSA was independent of hospital exposure we analyzed 56 nasal swab samples which were positive for *S. aureus*. 59% of that was MRSA strains. The MRSA nasal colonization for CAP and HAP was calculated to be 16.6% and 70.5% respectively (Figure 2). The OR for MRSA carriage in hospital environment was calculated to be 12.05. The p-value using chi-squared test were calculated to be P = 0.001, which is significant to associate MRSA nasal colonization with hospital exposure.



		Population	
		HA	CA
<i>S. aureus</i>	MRSA	31	2
	MSSA	13	10
	Total	44	12

Figure2: (Top) Bar diagram displaying the percentage of MRSA and MSSA colonization among CAP and HAP.(Bottom) Population wise MRSA nasal carriage.HAP = Hospital Associated Population, CAP = Community Associated Population, MRSA = *Methicillin Resistant S. aureus*, MSSA = *Methicillin Sensitive S. aureus*

***S. aureus* nasal carriage and MRSA nasal colonization is independent of gender.**

The percentage for male and female nasal carriage was calculated to be 24% and 33% respectively.

The MRSA nasal colonization was 59% and 58% (Figure 3). To probe if the differences were significant enough for gender to effect the *S. aureus* nasal carriage and MRSA colonization we performed statistical analysis. The OR and P-values for nasal carriage and MRSA colonization with respect to gender was calculated to be; OR=0.77, P=0.194 and OR=1.04, P=0.938 respectively. These values are suggestive of a neutral effect of gender on *S. aureus* nasal carriage and MRSA colonization.

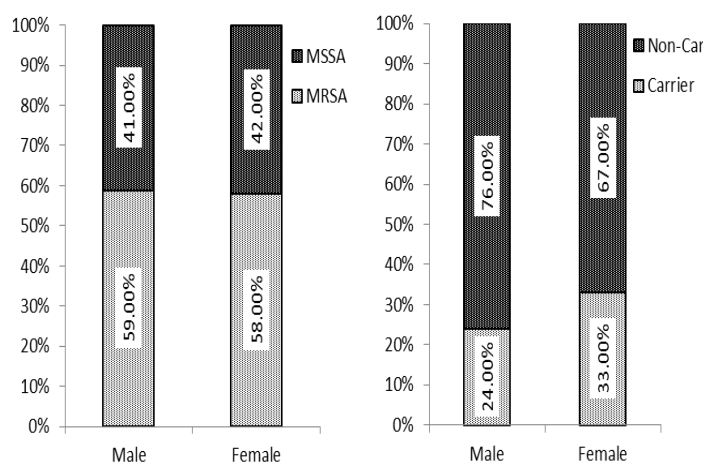


Figure3: Gender wise *S. aureus* and MRSA nasal carriage. MRSA = *Methicillin Resistant S. aureus*, MSSA = *Methicillin Sensitive S. aureus*

Discussion

S. aureus are the common inhabitants of skin, mucosae, perineum, pharynx (Wertheim et al 2005), gastrointestinal tract (Rimland & Roberson 1986), vagina, (Guinan et al 1982), axillae and the anterior nares (Dancer & Noble 1991), however the nares provide the principal reservoir for these organisms. About 20-30 % healthy individual harbour *S. aureus* in their nares (Costello et al 2009). The colonization of *S. aureus* especially MRSA in the nares is an endemic risk factor for infectious diseases. The current study designed for the surveillance of MRSA nasal carriage in healthy volunteers belonging to health care environment and non-health care environment revealed that overall MRSA carriage was 16%. The MRSA carriage for health care workers and community was calculated to be 26% and 2% respectively (Table

4). This upon statistical analysis was found to be an extremely significant difference. Suggesting an urgent need for the rational strategies to eradicate MRSA from hospital settings.

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Authors Contributions

Dileep Kumar processed the samples to isolate *S. aureus* and did the antibiotic sensitivity testing against *oxacillin*. The samples were processed at Muhamad Medical College, Mirpur-Khas and antibiotic sensitivity testing was done at Institute of Microbiology, University of Sindh, Jamshro. Data analysis was done by *Bushra Patoli* and *Atif Patoli*. *Atif Patoli* wrote the manuscript and was reviewed by *Bushra Patoli*.