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Prevalence of Nasal Carriers of Methicillin Resistant Staphylococcus Aureus among Health Care Workers

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Prevalence of Nasal Carriers of Methicillin Resistant *Staphylococcus Aureus* among Health Care Workers

A
Project work Submitted to

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Abstract

Background and objectives: Staphylococcal infections occur regularly in hospitalized patients and have severe consequences. Due to an increasing number of infections caused by Methicillin-Resistant Staphylococcus Aureus (MRSA) strains, which are now most often multiresistant, these strains impose an alarming threat to medical science. This study attempts to find out the prevalence of nasal carriers of MRSA in health care workers of Janamaitri Hospital and nursing students of JFIHS/LACHS, Kathmandu, Nepal.

Material and methods: A total of 154 nasal swabs were collected from the anterior nares of the nursing students (118) and hospital staffs (36) of JFIHS. These swabs were inoculated onto blood agar and mannitol salt agar. After overnight incubation the agar plates were analyzed for bacterial isolates and antibiogram was produced. MRSA detection was done by Cefoxitin disk test method.

Results: Out of 154 nasal swabs, 34(22.07%) of them had Staphylococcus aureus, 102(66.23%) of them had Coagulase Negative Staphylococcus (CoNS), and 18(11.68%) of them had no growth. Out of 34 Staphylococcus aureus, 11 of them were MRSA.

Conclusion: The present study indicates high nasal carriage rate of MRSA (32.35%) among the nursing students and the hospital staffs. The carriage rate was higher in case of nursing students than the other hospital staffs. Resistance to drugs of other classes was also seen in *Staphylococcus aureus*.

Keywords: *Staphylococcus aureus*; Nasal carriers; MRSA; MDR

Abbreviations

AST: Antibiotic Susceptibility Test; BA: Blood Agar; BN: Bachelor in Nursing; BSc: Bachelor of Science; B.Sc.Nursing: Bachelor of Science in Nursing; B.Sc. MLT: Bachelor of Science in Medical Laboratory Technology; CoNS: Coagulase Negative Staphylococcus; H2O2: Hydrogen Peroxide; JFIHS: Janamaitri Institute of Health Science; LACHS: Little Angles College of Higher Studies; MDR: Multi Drug Resistance; MHA: Muller Hilton Agar; MRSA: Methicillin resistant Staphylococcus aureus; MSSA: Methicillin sensitive Staphylococcus aureus; MSA: Mannitol Salt Agar; RT: Room Temperature; SPSS: Statistical Package for Social Sciences

Chapter I

Introduction

Staphylococcus aureus is a Gram Positive Coccal bacterium. It is a common cause of skin infection such abscesses, respiratory infection such as sinusitis, and food poisoning. *Staphylococcus* was first identified in 1880 in Aberdeen, Scotland, by the surgeon Sir Alexander Ogston in pus from a surgical abscess in a knee joint. This name was later appended to *Staphylococcus aureus* by Friedrich Julius Rosenbach, who was credited by the official system of nomenclature at the time. *Staphylococcus aureus* has been recognized as a major pathogen of hospital and community acquired infections. Staphylococcal infections occur regularly in hospitalized patients and have severe consequences. Due to an increasing number of infections caused by Methicillin-resistant. *S.* Aureus (MRSA) strains, which are now most often multiresistant [1].

First identified in the 1960s, methicillin-resistant *Staphylococcus aureus* (MRSA)are now regarded as a major hospital acquired pathogen worldwide [2]. The term methicillin resistant is historically used to describe resistance to any of this class of antimicrobials. MRSA began as a hospital-acquired infection, but has developed limited endemic status and is now sometimes community-acquired. The terms HA-MRSA (healthcare-associated mrsa) and CA-MRSA (community-associated mrsa) reflect this distinction [3].

Methicillin-resistant *Staphylococcus aureus* (MRSA) is an important cause of health care associated infections worldwide. Multidrug-resistant bacteria, such as methicillin-resistant *Staphylococcus aureus* (MRSA), are endemic in healthcare settings in many countries of the world. Nosocomial transmission of MRSA serves as a source of hospital outbreaks [4].

Healthcare workers may acquire methicillin-resistant *Staphylococcus aureus* (MRSA) from patients, both hospital and home environments, other healthcare workers, family and public acquaintances, and pets. There is a consensus of case reports and series which now strongly support the role for MRSA-carrying healthcare personnel to serve as a reservoir and as a vehicle of spread within healthcare settings [5].

Antibiotic sensitivity or antibiotic susceptibility is the susceptibility of bacteria to antibiotic. Because susceptibility can vary even within a species (with some strains being more resistant than others), antibiotic susceptibility testing (AST) is usually carried out to determine which antibiotic will be most successful in treating a bacterial infection in vivo [6].

Multidrug resistance is a condition enabling a disease-causing organism to resist distinct drugs or chemicals of a wide variety of structure and function targeted at eradicating the organism. Organisms that display multidrug resistance can be pathologic cells, including bacterial and neoplastic cells. Multidrug-Resistant Organisms (MDROs) are defined as microorganisms that are resistant to one or more classes of antimicrobial agents [7].

Statement of the problem

Nosocomial acquisition of methicillin-resistant *Staphylococcus aureus* (MRSA) has been an increasing problem world-wide. It now has got a high prevalence in community and even higher prevalence in medical personnel [8]. Thus, the study focuses on disclosing the threat imposed by strains of MRSA in nosocomial environment.

Rationale

Both methicillin sensitive and methicillin resistant *Staphylococcus aureus* (MSSA, MRSA) can cause invasive and life-threatening infections such as osteomyelitis, septicemia, endocarditis, and pneumonia [9]. Healthcare associated MRSA is of particular clinical importance because it is not only predictably cross resistant to all penicillin's and cephalosporin, but is also typically resistant to multiple other antibiotics [10]. Possibly because of inadequate initial antibiotic treatment, infections with health care associated MRSA result in increased costs and worse outcomes compared to infections with MSSA. Studies show that diligent surveillance programs, where patients are screened upon admission, have a tremendous impact in reducing MRSA infection rates. Identifying MRSA carriers not only among the patients but also among the medical personnel, isolating them, and administering antibiotics is an effective method to stop the spread of MRSA Healthy hospital personnel may carry pathogenic hospital strains in their nose and skin and may spread these pathogens to the community leading to more dreadful condition [11].

Healthcare workers, who have direct contact with persistently colonized patients, or contaminated objects in the immediate environment around them can contaminate their hands and subsequently transmit the organism to other patients. A subset of these will remain as nasal carrier for a prolonged period of time and may spread the organism to patients by direct contact transmission [11]. So, study of *S.* aureus as nasal carrier is of importance, especially in people concerned with hospitals to explore the clear picture regarding its existence.

Objective

General objective

To find out the prevalence of methicillin resistant *Staphylococcus aureus* in medical staff of Janamaitri hospital & medical students of JFIHS.

Specific Objective

i. To detect *Staphylococcus aureus* from nasal carriers
ii. To detect methicillin sensitive *Staphylococcus aureus*
iii. To detect methicillin-resistant *Staphylococcus aureus*
iv. To compare prevalence of *Staphylococcus aureus* carriers among department, age, sex.

Operational definition of terms/ functional definitions

Staphylococcus aureus

Identification of organisms was carried out by standard laboratory operating procedures (Gram staining, Catalase test, Mannitol fermentation, Coagulase test) [12].

Methicillin Resistant *Staphylococcus aureus*

Staphylococcus aureus isolates were deemed methicillin resistant when the Zone Of Inhibition will be ≤10 mm with the oxacillin disk & ≤21 mm with the cefoxitin disk [13].

Chapter II

Review of the Literature

Staphylococcus aureus is carried in the anterior nares of 40% of healthy individuals [14] defined as persistent carriers, whereas the remaining part of the population does not carry or is only transiently colonized by this opportunistic pathogen [15]. Over the last 2 decades, rates of infection with MRSA have risen rapidly in hospitals. Currently, almost half of nosocomial *S.* aureus infections are resistant to methicillin [16].

Shortly after the introduction of methicillin, patient carriers and hospital outbreaks of MRSA were recognized, and the first citation of a medical staff carrier (nurse) was reported [17]. Since then, it has been generally accepted that the hands of healthcare personnel are critical vectors for transmission of MRSA [18-22].

From November 2010 to March 2011 in Dessie Regional Health Research laboratory, A cross sectional study was conducted on a total of 118 HCWs (hospital care workers). Of the 118 healthcare workers, 34 (28.8%) carried *S.* aureus of which 15 were methicillin resistant. Therefore, 12.7% of all HCWs were identified as MRSA carriers. The rate of methicillin resistance among all *S.* aureus isolates was44.1% (15/34). MRSA carriage was particularly high among nurses (21.2%). The highest rate of MRSA carriers (57.1%) were workers of surgical wards [23].

Prevalence of nasal carriage and genetic diversity of *S. aureus* were determined among hospital staff (HS) and inpatients (IP) at the largest hospital in Ghana. In total, 632 nasal swabs were obtained from 452 IP and 180 HS in the Child Health Department (CHD) and Surgical Department (SD). *S. aureus* carriage prevalences were 13.9% in IP and 23.3% in HS. The chance of being a carrier was higher in HS (P = 0.005) and IP staying ≤7 days in hospital (P = 0.007). Resistance to penicillin (93%), tetracycline (28%) and fusidic acid (12%) was more common than for other agents (<5%). A higher chance of multidrug-resistant *S. aureus* carriage was observed among IP compared with HS (*P* = 0.01) [24].

Between October 2007 to November 2007 in the surgical wards of a tertiary care hospital in India, out of 100 swabs 13 were isolated as *Staphylococcus aureus*, of which 2(15.4%) were resistant to methicillin [25].

In a study of Brazilian university students (pharmacy, nursing, dentistry and medicine), the percentage nasal carriage of *Staphylococcus aureus* was 40.8% (102/250). Of the isolates, MIC_{50} of methicillin was 0.5 µg/mL and MIC_{90} was 1µg/mL. Six (5.8%) isolates were methicillin-resistant and carried the *mecA* gene [26].

Study was carried out in Microbiology section of Department of Pathology at National Institute of Child Health Karachi over a period of 13 months from March 2008 to April 2009. Eighty seven isolates of staphylococcus aureus were recovered from various clinical samples. Sixty six (75.86%) were isolated from various swabs and 21(24.13%) from blood. Twenty isolates (22.9%) were methicillin resistant. In this group high resistant was found to cloxacillin (100%), cephradine (100%), co-trimoxazole (95%), erythromycin (70%), Chloromycetin (65%), gentamicin (55%) and low resistance was observed to ciprofloxacin (30%).In MSSA 0% resistance was seen to meropenem, ciprofloxacin and chloromycetin and high resistance found to co-trimoxazole (98.5%) and pencillin (73.13%). Low resistance was also observed with amoxiclav (20.89%) [7].

From July 2009 to July 2010 in Kathmandu Medical college-Teaching Hospital out of 111 *Staphylococcus aureus* isolates 29(26.12%) were identified to be MRSA [27]. A retrospective study was conducted from December 2010to December 2012 at Chitwan Medical College Teaching Hospital (a 600 bed teaching hospital), Chitwan, Nepal. From the processed samples, 306 isolates of Staphylococcus aureus were recovered. Methicillin resistance was observed in 43.1% of isolates [28].

A study was carried out at National Medical College & Teaching Hospital, Birgunj during the period between april to june 2011 with the objective to study the nasal carriage of *Staphylococcus aureus* among the staffs at the hospital. A total of 54 nasal swabs were taken from the hospital staff. All the samples were processed following standard microbiological method. Gram positive cocci that were mannitol fermenting, catalase positive and coagulase positive isolates were considered as *S.* aureus. Nasal carriage rate of *S.* aureus among hospital staff was found to be 20.37%. Carriage among male and female staff was 19% and 21.2% respectively (p>0.05). All nasal *S.* aureus isolates were sensitive to Amikacin and Vancomycin. Methicillin resistance rate was found to be 45.5%. High rate of nasal carriage of *S.* aureus indicates need for standard infection control practices to prevent transmission [29].

From May 2008 and October 2009 in China, a total of 2103 medical interns were randomly tested for nasal colonization of *S. aureus* and methicillin-resistant *Staphylococcus aureus* (MRSA). The prevalence of *S. aureus* among staphylococci specimens was 23.1%, and among the total *S. aureus* the MRSA prevalence was 9.4% [30].

Study was undertaken from July to December 2014 among the medical students and interns in a medical college in India, and completed in a period of 6 months (two months for sampling and culture for MRSA from all the 150 students) which showed the prevalence of 0.7% [31].

Between September 2014 and January 2015 the prevalence of MRSA among medical students at King Abdulaziz University (KAU), Jeddah, Saudi Arabia, using molecular approaches was studied. Out of 150 students screened, 38 were nasal carriers of *S. aureus*. The prevalence of methicillin-sensitive *S. aureus* (MSSA) carriers was 18.7% (n=28), whereas 10 students (6.7%) were *mecA*-positive, representing MRSA carriers. Interns carry MRSA more than 6th year students and students who were not exposed to clinical work ($p<0.05$), while MSSA is found more in students who were not exposed to clinical work ($p<0.01$) [32].

Chapter III

Materials & Methods

Materials

The materials & equipment that were used are mentioned below annexure.

Methods

Type of study: Prospective study

Study site: JFIHS

Study population:

The study populations were the staffs & students of JFIHS.

Sample size

All the targeted medical students (B. Sc. Nursing 2nd, 3rd, 4th year & BN 1st, 2nd, 3rd year) & staff of JFIHS.

Study variables:

The study variables included types of working departments, age, sex, antibiotic resistance pattern, MRSA.

Time/duration

Duration of the study was of 6 months

Inclusion & exclusion criteria

Inclusion

i. Medical students of JFIHS(B. Sc. Nursing 2nd, 3rd, 4th year & BN 1st, 2nd, 3rd year).

ii. Staffs of JFIHS.

iii. Healthy individuals without the history of recent (within 1 month) antimicrobial therapy.

iv. Individuals without any infections or abscesses in nasal cavity.

Exclusion

i. Person on antibiotic within 1 month and with any sort of infections or abscesses in nasal cavity.

ii. Person not exposed to hospital environment.

Limitation of the study

i. The study was limited only to medical (nursing) students & staffs of JFIHS

ii. Time framework for the study was only six months.

Validity & reliability (Ethical consideration)

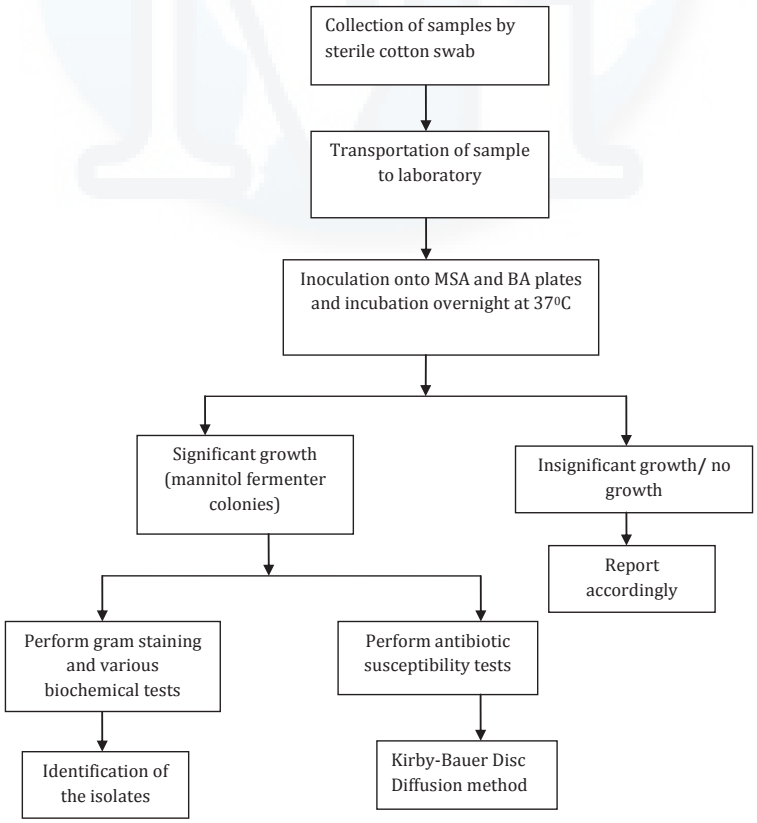
The ethical clearance was taken from the ethical committee of Janamaitri Hospital & NHRC (Ref. No. 1047) (Procedure).

Data analysis

All the results were entered in the worksheet of statistical package for social science (SPSS) software version 20.0

Budgeting

The allocated budget for the study and the details of expenditure in various topics are mentioned below annexure.



Chapter IV

Result

A total of 154 nasal swabs were collected from the anterior nares of the nursing students (118) and hospital staffs (36) of JFIHS. These swabs were inoculated onto blood agar and mannitol salt agar. After overnight incubation the agar plates were analyzed for bacterial isolates and antibiogram was produced. MRSA detection was done by Cefoxitin disk test method (Figure 1-8) (Table 1-5).

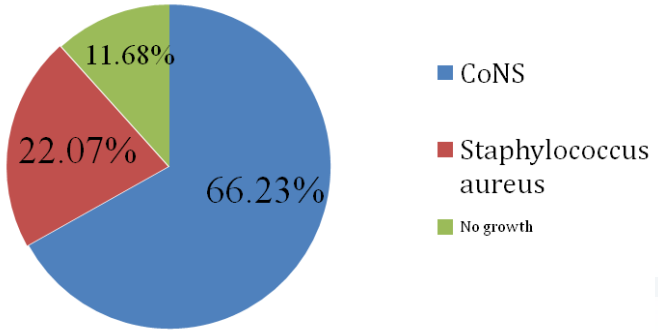


Figure 1: Pie chart showing distribution of various bacteria isolated in total population.

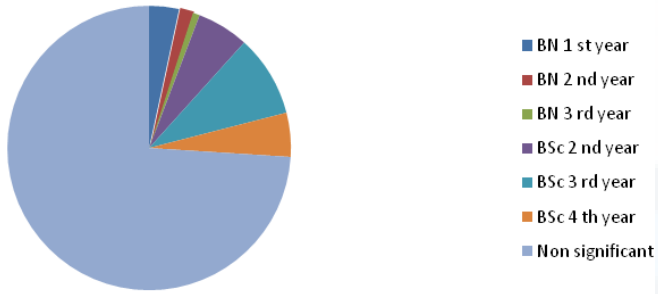


Figure 2: Pie chart showing distribution of *Staphylococcus aureus* in students.

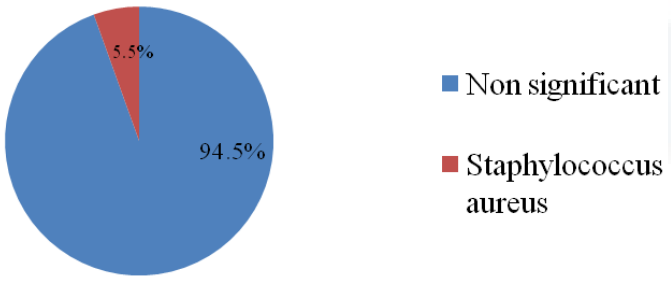


Figure 3: Pie chart showing distribution of *Staphylococcus aureus* in staffs.

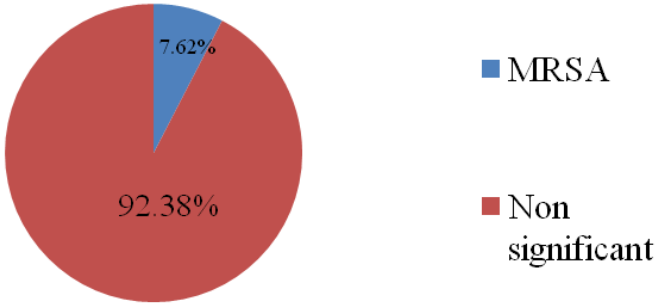


Figure 4: Pie chart showing distribution of *Staphylococcus aureus* in Student among MRSA in total population = 7.62% (9/118*100%).

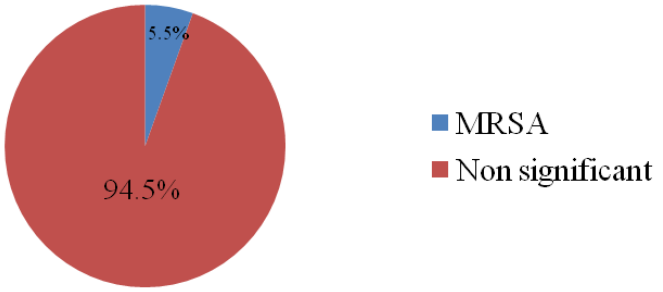


Figure 5: Pie chart showing distribution of *Staphylococcus aureus* in Staff among MRSA in total population = 5.55% (2/36*100%).

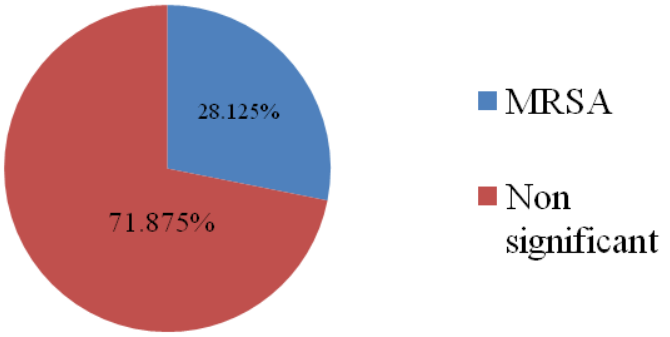


Figure 6: Pie chart showing distribution of *Staphylococcus aureus* in Student among MRSA in *Staphylococcus aureus* population = 28.125% (9/32*100%).

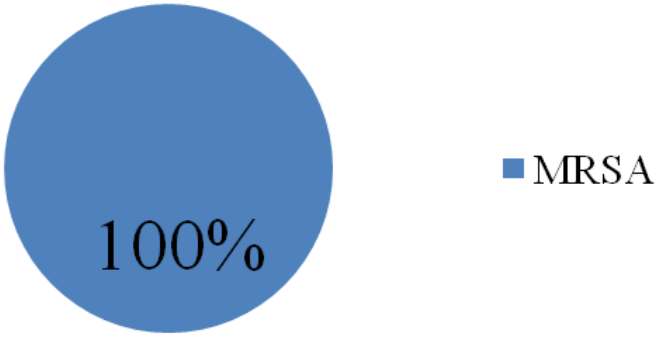


Figure 7: Pie chart showing distribution of *Staphylococcus aureus* in Staff among MRSA in *Staphylococcus aureus* population = 100% (2/2*100%).

Table 1: Group wise distribution of *S.aureus.*

Group	Total Population	No. of Positivity	Prevalence %
BN1	18	4	22.2
BN2	9	2	22.2
BN3	11	1	9.1
B.Sc. 2	25	7	28
B.Sc. 3	28	11	31.3
B.Sc. 4	27	7	25.9
Staff	36	2	5.55
Total	154	34	

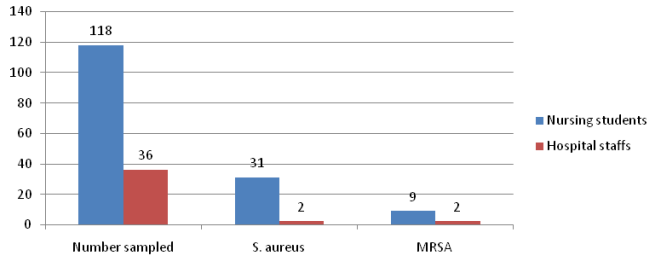


Figure 8: Distribution of *S. aureus* and MRSA carriage among nursing students and hospital staffs.

Table 2: Work experience wise distribution of *S.aureus*.

Work Experience	Total Population	Positive	Prevalence %
<=3	121	29	24
>3	23	5	15.2
Total	154	34	

Table 3: Age wise distribution of *S.aureus*.

Age	Total Population	Positive	Prevalence %
<=25	122	32	25.2
>25	22	2	7.4
Total	154	34	

Table 4: Sex wise distribution of *S.aureus*.

Sex	Total Population	Positive	Prevalence %
Male	12	1	8.3
Female	142	33	23.2
Total	154	34	

Table 5: Antibiotic Susceptibility Pattern.

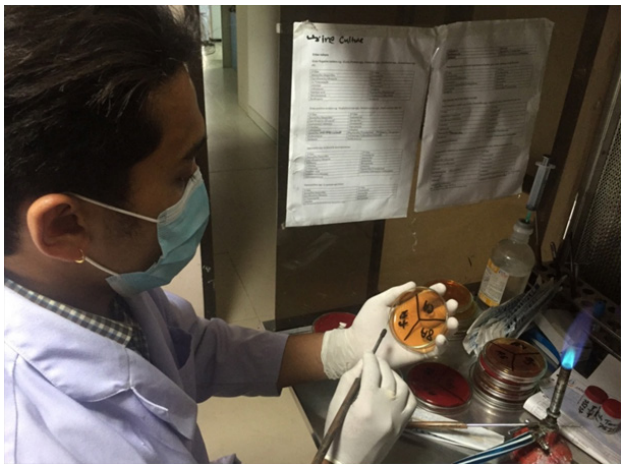
Antibiotics		Susceptibility Pattern	
Class	Name	Sensitive (%)	Resistant (%)
Aminoglycosides	Gentamicin	100	0
Quinolones	Ciprofloxacin	67.64	22.26
Macrolides	Erythromycin	44.11	55.99
Beta lactams	Penicillin	0	100
Beta lactams	Cefoxitin	67.65	32.35
Macrolides	Amikacin	97.05	2.95

Out of 154 nasal swabs, 34(22.07%) of them had Staphylococcus aureus, 102(66.23%) of them had Coagulase negative Staphylococcus (CoNS), and 18(11.68%) of them had no growth (Figure 1).

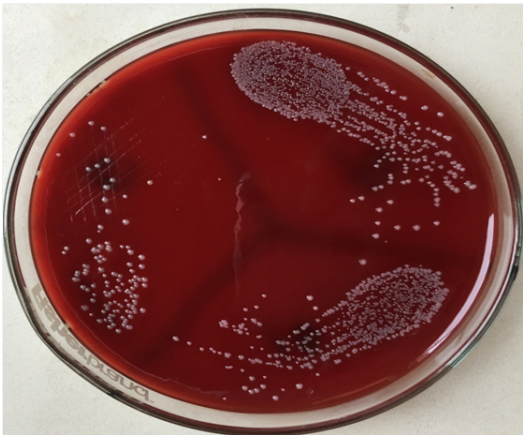
Of 118 students, 32(27.11%) had nasal carriage of *Staphylococcus aureus* (BN 1st =4 & BN 2nd =2 & BN 3rd =1 & B.Sc. 2nd =7 & B.Sc. 3rd =11 & B.Sc. 4th =7) (Figure 2).

And out of 36 staff tested, 2 (5.5%) had nasal carriage of *Staphylococcus aureus* (Figure 3).

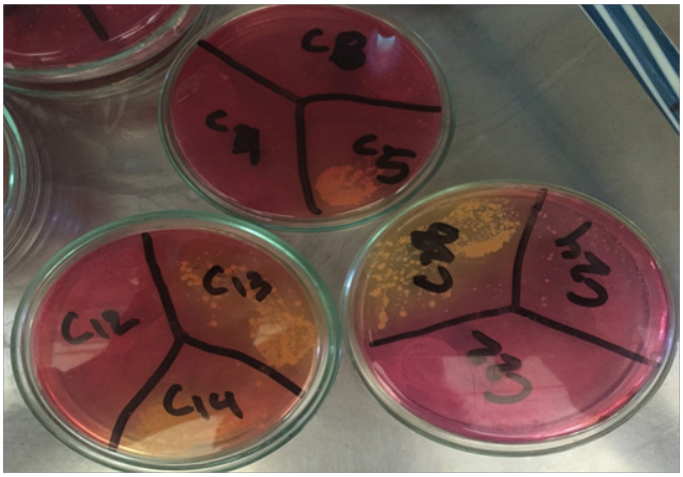
Out of 34 *Staphylococcus aureus*, 11 of them were resistant to Methicillin (MRSA). 9 were detected in the sample of nursing students and 2 were detected in the hospital staffs (Figure 4) (Photographs 1-4).



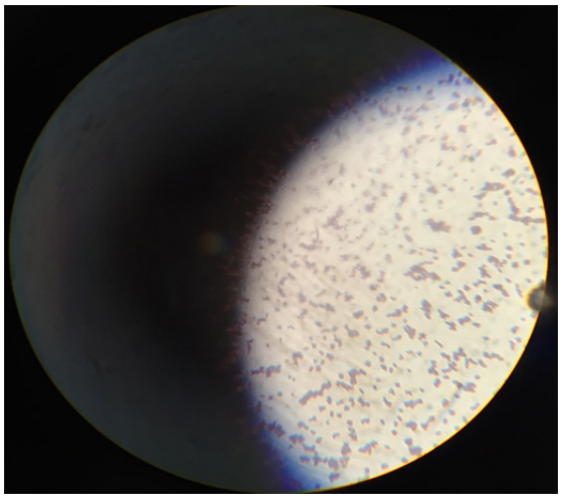
Photograph 1: Bench work activity.



Photograph 2: Blood Agar showing colonies of *Staphylococcus aureus*.



Photograph 3: Mannitol Salt Agar showing yellow colonies of mannitol fermenting *Staphylococcus aureus*.



Photograph 4: Microscopic observation of Gram Positive Cocci.

Chapter V

Discussion

i. Methicillin resistant *staphylococcus aureus* (MRSA) infection has increased over the years, becoming great threat in developing and non-developing countries. In the past it was usually found in hospital setting but now it has moved out into the community. MRSA can cause minor infection to serious ailment that sometimes becomes fatal.

ii. Present study showed prevalence rate of MRSA to be 32.35% among the nursing students and the hospital staffs in nasal swab. An important finding of this study is that there is the highest prevalence of nasal carriers of MRSA among the nursing students. This could possibly be explained due to the high frequency of patients contact with the nurses.

iii. A similar study was done in Northeast Ethiopia by Shibabaw et al. [23] which showed that the overall prevalence of nasal carriers of MRSA was 44.1% and the higher carriage was among the nurses (21.2%).

iv. Present study showed prevalence rate of *Staphylococcus aureus* to be 22.07% and MRSA to be 7.14% in our study population.

v. Higher prevalence rate of *S. aureus* in students (27.11%) as compared to staff (5.5%) can be attributed to factors like limited knowledge about nosocomial infection and their control measures, less awareness of the possible hazard of infectious agents, less development of professionalism etc. In general this could possibly be explained due to insufficient knowledge and inexperience of the students in the profession.

vi. Among 36 staff tested, only 2 were positive for *S. aureus*, but surprisingly both strains were MRSA. This low prevalence rate in staff might be due to easy access and prompt consumption of antibiotics in their routine life.

vii. The high prevalence of *S. aureus* among students of BSc Nursing third year (31.3%) and fourth year (25.9%) might be mainly attributed to their longer stay in wards during their clinical postings.

viii. Regarding distribution of *S.* aureus on work experience basis, less experienced individuals (≤ 3 years) are found to have higher prevalence of 24.0% in comparison to more experienced (>3 years) with 15.2 % prevalence. This also can be arguably explained on the fact that less experienced workers are less aware of the infection prevention habits which make them more prone to attain variety of bacteria.

ix. On the basis of sex, higher prevalence of *S. aureus* of 23.2% was seen in females, while only 8.3% males were positive. This distribution in more related to occupation rather than sex where majority of the female participants are nurses who work in close proximity to patients whereas males work in other departments.

x. With regard to age, the prevalence rate was found to be higher (25.2%) in younger candidates of ≤25 years in comparison to the prevalence rate (7.4%) of older candidates of >25 years age. This also can be due to longer working experience of the elder candidates in their respective profession.

xi. Multidrug resistance is one of the emerging problems along with the MRSA. In the present study, all the MRSA strains are found to be resistant to Penicillin along with the cefoxitin. Out of 11 MRSA, 4 of them are resistant to Erythromycin, Penicillin and Ciprofloxacin along with cefoxitin. Resistance to Erythromycin and Penicillin was seen in 3 cases. Resistance to only Penicillin along with cefoxitin was seen in 3 cases. All the MRSA were sensitive to Gentamicin and Amikacin.

xii. Erythromycin has been used extensively for the treatment of both minor and more serious staphylococcal infections. As a consequence, its role today is increasingly limited due to increasing resistance, which poses a great therapeutic challenge.

xiii. Another similar study done in TUTH, IOM, Maharajgunj detected 44.4% nasal carriage of MRSA among the patients admitted in different wards of the hospital.

xiv. Obviously, the above mentioned studies are not fully comparable because there may be the differences in the study design such as sample size and method of MRSA identification. In addition, carrier rates might be influenced by poor personal hygiene of study participants, poor sanitation of the hospital and difference in sampling techniques.

Chapter VI

Conclusion & recommendations

Conclusion

i. The present study indicates high nasal carriage rate of MRSA (32.35%) among the nursing students and the hospital staffs who were positive for *S. aureus*. The carriage rate was highest in case of nursing students than the other hospital staffs. There was also resistance to other drugs in *Staphylococcus aureus*.

ii. This high carriage rate *S.* aureus and particularly MRSA particularly in students and less experienced medical workers is a great threat for nosocomial infection transmission.

iii. MDR strains of *Staphylococcus aureus* are also emerging at an alarming rate which has been worsening the scenario.

Recommendations

i. Due to an increasing number of infections caused by methicillin-resistant *Staphylococcus aureus* (MRSA) strains, which are now most often multiresistant, staphylococcal infection has been a serious threat in modern medicine.

ii. There is a need for longitudinal surveillance of MRSA and its antimicrobial susceptibility profile in Nepal.

iii. Acts have to formulate so that the patients will have to suffer less from the nosocomial infection due to improper handling by inexperienced medical students.

iv. Effective implementation of hospital infection control and antibiotic policies to control antibiotic resistance in *Staphylococcus aureus*.

v. Staff screening should follow a strict protocol to ensure only persistent MRSA carriage is detected.

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Annexure

Materials

List of equipment and materials

Equipment

a) Autoclave b) Cotton swab c) Hot air oven d) Electric weighing pan

e) Microscope e) Inoculating loop f) Incubator g) Fridge

Media

a) Mannitol Salt Agar b) Blood Agar c) Mueller Hinton Agar

Chemical & Reagents

a) Gram staining reagents b) Antibiotics c) H2O2 d) Plasma

Glass wares and others

a) Conical flask b) Cotton c)Normal saline d) Glass slide e) Immersion oil

f) Lysol g) Petridish plates h) Pipettes i) Test tubes

Antibiotic discs

The antibiotics that were used for the susceptibility tests are as follows:

penicillin (10U), ciprofloxacin (5µg), erythromycin(15µg), co-trimoxazole (25µg), gentamicin (10µg),amikacin (30µg), cephalexin (30µg), ceftriaxone 30µg),

cefoxitin (30µg), oxacillin (1µg), [20]

Informed consent form

Consent form for Nasal Swab sample testing

This is to inform that I have been counseled about the nasal swab sample test to be conducted on me and have been explained about the implications of the test result. All details pertaining to Bacterial pathogens, its transmission, testing procedure, its limitations and interpretations of results have been explained to me in a manner that I can understand. I also understand that I am free to refuse the test and still get the help I need from this study without being discriminated against.

I hereby give my consent for the test to be conducted in order to know my status of nasal carriage of Staphylococcus aureus.

Code no. of the participant: … … … … … … … … … …

Signature of the participant: … … … … … … … … …

Date: … … … … … … … … … …

Proforma

Clinical Profile of the Participant

Name: … … … … … … … … Lab No: … … … … … …

Age/Sex: … … … … … … … Date: … … … … … …

Brief Clinical History:

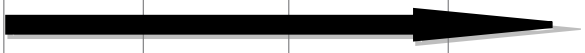
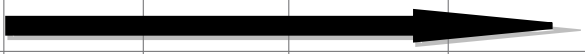
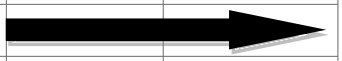
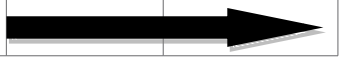
Participant on antibiotics: Yes ( ) No ( )

If Yes, Antibiotic(s) taken: 1) … … … … … … … … 2) … … … … … … …

Duration of treatment: … … … … … … … … … …

Operational Exposure: … …. … …. …. …. … .. … ….

Work Plan

Working plan	1st month	2nd month	3rd month	4th month	5th month	6th month
Collection of clinical samples from patients						
Samples processing						
Analysis of data						
Final report preparation & submission						

Budget

Item	Expenditure (N.C)
Laboratory Charge	10,000
Culture Media	10,000
Antibiotic Discs	10,000
Miscellaneous	15,000
Total	45,000