ADDIS ABABA UNIVERSITY

COLLAGE OF HEALTH SCIENCES

DEPARTMENT OF MEDICAL LABORATORY SCIENCE

CARRIAGE RATE OF NASAL STAPHYLOCOCCUS AUREUS AND METHICILLIN-RESISTANT STAPHYLOCOCCUS AUREUS AND ASSOCIATED FACTORS AMONG DIABETIC PATIENTS AT TIKUR ANBESSA SPECIALIZED HOSPITAL DIABETIC CLINIC, ADDIS ABABA, ETHIOPIA

BY: ADANE AMERA

ADVISOR: KASSU DESTA (MSc, PhD CANDIDATE, ASSOCIATE PROFESSOR)

A research thesis submitted to the Department of Medical Laboratory Sciences, College of Health Science, Addis Ababa University, in partial fulfillment of Master of Science Degree in Clinical Laboratory Sciences (Diagnostic and Public Health Microbiology)

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Carriage rate of nasal *staphylococcus aureus* and methicillin-resistant *staphylococcus aureus* and associated factors among diabetic patients at Tikur Anbessa Specialized Hospital Diabetic Clinic, Addis Ababa, Ethiopia

Approved by the Examining Board

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ABBREVIATIONS

BAP	Blood agar plat
CA-MRSA	Community-associated MRSA
CI	Confidence interval
CLSI	Clinical and Laboratory Standards Institute
DM	Diabetic Mellitus
HA- MRSA	Hospital-associated MRSA
HD	Hemodialysis
MecA	Methicillin resistance gene
MHA	Muller Hinton agar
MRSA	Methicillin-resistant Staphylococcus aureus
MRSA	Methicillin-resistant Staphylococcus aureus
MSA	Mannitol salt agar
PBP2a	Penicillin-binding protein 2a
PI	Principal Investigators
QC	Quality control
SA	Staphylococcus aureus
SA	Staphylococcus aureus
SOP	Standard operating procedure
TASH	Tikur Anbessa Specialized Hospital
NCSA	Nasal carriage of S.aureus (NCSA)
CLSIC	Clinical and Laboratory Standard Institute
SPSS	Statistical Package for the Social Sciences

Abstract

Background: Currently the prevalence of diabetes mellitus is increased alarmingly in low and middle income countries. Concurrently the drug resistant *S. aureus* nasal carriage rate is increasing that may serve as a source of further infections in Diabetes mellitus patients. There is no study conducted on nasal carriage rate of *S.aureus* and Methicillin resistant *S.aureus* (MRSA) among diabetes patients in Ethiopia.

Objective: The aim of this study was to assess the rate of nasal carriage of S.aureus and MRSA and associated factors among diabetic patients at Tikur Anbessa Specialized Hospital Diabetic Clinic, Addis Ababa, Ethiopia from January to June, 2018.

Methods: A cross-sectional study was conducted from January to June, 2018. A total of 422 diabetic patients were recruited at TASH Diabetic Clinic. Nasal swab from the anterior nares was collected, and culture on both Manitol salt agar and blood agar. Antimicrobial susceptibility testing was done using disc diffusion method on Muller Hinton agar. Susceptibility to methicillin was phenotypically determined based on resistance of isolates to cefoxitin. Data was entered and analyzed using SPSS software version 20. Binary logistic regression analysis was used to assess the association of socio-demographic and associated factors for *S.aureus* culture isolates. P-value < 0.05 was considered as statistically significant.

Results: The overall prevalence of *S.aureus* in the study was 47/422(11.1%). Majority of the *S. aureus* isolates were resistant to penicillin 46(97.8) on the other hand, all of the *S. aureus* isolates were sensitive to clindamycin. Multidrug resistance were shown in 4(6.2%) of the isolates. Of these, one MRSA was isolated from retire man who had uncontrolled glucose level with Type II DM despite insulin treatment and had foot wound. *S.aureus* isolation rate was significantly higher among DM patients who currently used combination of insulin and oral drug for diabetes mellitus treatment (odds ratio [AOR], 3.985, 95% CI, 1.270-12.437; p<0.017). None of the assessed socio-demographic factors showed a significant association with *S.aureus* nasal colonization.

Conclusion: The nasal carriage rate of *S.aureus* among DM patients was low in this study. Also, the carriage rate of MRSA was very low in this study compared to other study; performing culture and sensitivity test for those DM patients would reduce the complication of infection. Further multicenter large scale studies should be conducted to identify the associated risk factors and antibiotic resistance pattern in diabetes mellitus patients.

Key words: *Staphylococcus aureus*, MRSA, diabetes mellitus patients

1. Introduction

1.1 Background

Staphylococcus aureus (*S. aureus*) is facultative anaerobic, Gram-positive cocci, catalase and coagulase positive bacterium. *S.aureus* is a part of genus *Staphylococcus*, which contains more than 30 species such as *S. epidermidis*, *S. saprophyticus*and *S. haemolyticus*. Among the *staphylococcal* species, *S. aureus* is by far the most virulent and pathogenic for humans. Infections caused by *S. aureus* remain a significant cause of mortality and morbidity in both in healthy and immune-compromised individuals (1).

The principal site of *staphylococcal* colonization is the anterior nares. It has been observed that if repeated cultures are performed, up to 80% of adults are found to harbor *S. aureus* in the nose at one time or the other. However, in most persons, the carrier state is transient, but 20 to 40% of adults remain colonized for months or even years (2,3).The nose, throat and oral cavity are the normal habitant of this organism. Approximately 50% to 60% of individuals are intermittently or permanently colonized with *S. aureus* and, thus, there is relatively high potential for infections. The anterior nares are the main ecological niches for *S. aureus*. The carriage has been identifying as a risk factor for subsequent infections (4).

Increased nasal colonization rates have been noted in DM patients, individuals on hemodialysis, those on ambulatory peritoneal dialysis, intravenous drug users and patients receiving routine allergy injections. It has also been suggested that patients with symptomatic human immunodeficiency virus infection have an increased colonization risks (5).

S.aureus has the ability to adapt to different environments and it may colonize the human skin, nails, back of the throat nares and mucus membranes and may thereby disseminate among recipient host populations via physical contact and aerosols. When an infected person coughs or sneezes, he or she releases numerous small droplets of saliva that remain suspended in air. These contain the bacteria and can infect others. Another common method of transmission is through direct contact with objects that are contaminated by the bacteria

S. aureus causes a wide range of infections from a variety of skin, wound and deep tissue infections to more life-threatening conditions such as pneumonia, endocarditis, septic arthritis and septicemia. This bacterium is also one of the most common species in nosocomial infections.

In addition, *S. aureus* may also cause food poisoning, scalded-skin syndrome and toxic shock syndrome,through production of different toxins.(1.6).

Diabetes is a serious, chronic non-communicable disease that predisposes to various infections such as bacteremia and foot ulcer infection (7). The source of these infections may be due to either endogenous or exogenous colonization. Studies have shown that nasal carriage of S.aureus (NCSA) is the most common source of infection for diabetes patients (8). One of the most common complications is diabetic foot ulcers (9).According to existing data, 15% of patients with DM during their lifetime suffered from the diabetic Foot ulcer (10). The pattern and epidemiology of MRSA infection in diabetic patients are not well defined (11).

Infectious diseases are more prevalent in individuals with DM. The main pathogenic mechanisms are: hyperglycemic environment increasing the virulence of some pathogens; lower production of interleukins in response to infection; reduced chemotaxis and phagocytic activity, immobilization of polymorphonuclear leukocytes; glycosuria, gastrointestinal and urinary dysmotility. Infectious diseases in DM may result in metabolic complications such as hypoglycemia, ketoacidosis, and coma. Hyperglycemia may compromise the immune system. Ex-vivo experiments, in which human cells are analysed in a laboratory environment outside of the body, show that innate cellular immunity may be compromised in hyperglycemic conditions. In a hyperglycemic or acidic environment neutrophils and macrophages malfunction, and restoring normoglycemia and a normal pH reverses these abnormalities. The resistance to antimicrobial agents is an increasingly global problem worldwide, especially among nosocomial pathogens. Staphylococci have become one of the most common causes of nosocomial infections. Multidrug-resistant staphylococci pose a growing problem for human health because of drug-resistant virulent strains of S. aureus, particularly MRSA (12).

Methicillin Resistant *Staphylococcus aureus* is a bacterium resistant to conventional therapies against Gram-positive organisms, in particular beta (β) - Lactam antibiotics (13). Methicillin resistant S. aureus acquires its resistance through the methicillin resistance gene mecA, which encodes a low-affinity penicillin-binding protein (PBP2a) that is absent in susceptible S. aureus strains (14, 15). This foreign penicillin-binding protein does not bind well to most β-lactams, and therefore allows MRSA to grow in their presence. The mecA gene is carried on a mobile genetic element called the Staphylococcal chromosomal cassette mec (SCCmec) (15).

1.2 Statement of the problem

Staphylococcus aureus is a major pathogen responsible for nosocomial and community-acquired infection. Methicillin resistant *S. aureus* has emerged as a nosocomial pathogen of major worldwide importance and is an increasingly frequent cause of community-acquired infections that cause significant morbidity and mortality. In 1961 there were reports from the United Kingdom of *S. aureus* isolates, which resisted to methicillin, and MRSA isolates were soon recovered from other European countries, and later from Japan, Australia, and the United States (16).

MRSA is a global public health problem. The MRSA infections can be found in both hospitals and the community. Generally, hospital-associated MRSA (HA-MRSA) infects hospitalized individuals with predisposing risk factors. On the other hand, community-associated MRSA (CA-MRSA) infects healthy individuals without any previous healthcare contact. CA-MRSA causes a paradigm shift in the management of staphylococcal infections in countries in which CA-MRSA is highly endemic (17).

According to the most recent statistics from WHO Global report on diabetes are globally the number of people living with diabetes and its prevalence are growing in all regions of the world. In 2014, 422 million adults (or 8.5% of the population) had diabetes, compared with 108 million (4.7%) in 1980. The epidemic of diabetes has major health and socioeconomic impacts, especially in developing countries. The complications of diabetes can lead to heart attack, stroke, blindness, kidney failure and lower limb amputation. Diabetes caused 1.5 million deaths in 2012. Higher than-optimal-blood glucose caused an additional 2.2 million deaths, by increasing the risks of cardiovascular and other diseases. In African region an estimated of 14.2 million adults have diabetes, Ethiopia is one of the high burden countries in Africa (18).

The significance of methicillin resistant *S.aureus* to public health is the ability of the pathogen to rapidly acquire resistance and virulence gene. This in turn, paving way for the emergence of new and highly pathogenic clones making treatment with antibiotics difficult and prolonging hospital admission stay (19).

Increased antibiotic resistance, frequency of invasive surgery, use of intravascular devices, and increased numbers of patients with immune-compromised status because of HIV infection or

immunosuppression after transplantation or cancer treatment and DM, has led to sharp increases in the incidence of *S.aureus* bacteremia and *S. aureus* infective endocarditis over the past 30 years. S. aureus in Ethiopia has gotten notoriously resistant to almost to all of antimicrobial agents in use including, penicillin, cephalosporins, tetracyclines, chloramphenicol, methicillin, vancomycin and sulphonamides(20).

The epidemiology of MRSA nasal carriage in diabetic patients is not well defined. Hence, early diagnosis is essential to provide appropriate treatment and prevent the spread of infection, especially with MDR S.aureus strains. However, investigations regarding to MRSA nasal colonization among diabetes population are limited in Ethiopia.

1.3 Significance of the study

Currently, the prevalence of multidrug resistance (MDR) and DM are the major health problem worldwide, particularly in developing countries including Ethiopia. Multidrug resistant organism such as *S.aureus* was mainly acquired in the hospital environments. DM patients frequently visit the hospital for their treatment follow-up and care, where there is high chance of being colonized by MDR bacteria, hence such study allow us to know the burden of the problem in this particular group of patients.

The result of this study benefit for DM patients early diagnosis and management with *S. aureus* infection, it gives additional information for physicians to reduce further complication and better management of diabetes patients. The finding also provide information for policy makers and other concerned bodies or designing an intervention and finding of this study can serve as source information for further studies.

2. Literature review

2.1 Epidemiology

S.aureus is the most frequently occurring bacterial pathogen among clinical isolates from hospital inpatients in the United States and is the second most prevalent bacterial pathogen among clinical isolates from outpatients (22). During the period 1997–2002, *S. aureus* was the most common cause of nosocomial bacteremia in North America 26.0% and Latin America (prevalence, 21.6%) and the second most common cause of nosocomial bacteremia in Europe 19.5%(23).

The prevalence rate of MRSA is various in different countries such as 1-5% in Northern Europe, 5-30% in Southern Europe, 5-40 % in Asian countries and 10-50 % in USA and in UK. The problems of treating such infections are mainly due to the multi-drug resistant nature of MRSA. The prevalence of MRSA in Nepal is reported to be 15-69% from different tertiary care hospitals (24, 25). Similar study conducted in Africa showed that the available evidence regarding the prevalence of methicillin resistance among *S. aureus* isolates collected in the African countries in different relevant studies yields variable findings, making the extrapolation of definitive relevant conclusions rather difficult. Certainly though, MRSA poses a visible threat in many African countries. The spread of MRSA in the African region can be worrisome, since there might be relatively limited availability of modern antibiotics effective against hospital-associated MRSA, like linezolid and daptomycin, in most of this part of the world. Furthermore, the implementation of infection control measures and the wide spread of HIV infection and tuberculosis, particularly in the sub-Saharan area, amplify the difficulty of dealing with the MRSA epidemic in Africa (32).

A study was conducted in turkey on the *staphylococcus aureus* nasal carriage rate and associated factor in Type II diabetic patient between march 2003 to December 2004 , showed that, the nasal carriage rate S.aureus on outpatient clinic type 2 diabetic patient insulin therapy dietetic treatment and oral anti diabetic agent and dietetic treatment were 35.3% and 15.8 % respectively(2).

A Prospective Cohort study was conducted in the endocrinology clinic of ShahidBehesty hospital, a central teaching hospital in Kashan, Iran on the Risk Factors of Methicillin-Resistant Staphylococcus aureus Colonization in Diabetic Outpatients, reported that nasal colonization of S. aureus among 494 DM patient was 42.5% (210). Of these, 122 (58%) were MRSA (24.6% of

all DM patients). The resistance rate of erythromycin, ciprofloxacin and clarithromycin was 81.9, 71.3 and 65.5%. The rate of multidrug resistance was 59% (72 isolates). Prevalence *s.aureus* 42.5 (26). A community-based cross-sectional study was conducted Prevalence and characteristics of Staphylococcus aureus and methicillin-resistant Staphylococcus aureus nasal colonization among diabetes population in China between April 2014 and May 2015, showed that from 529 diabetes participants, 46 (8.70%) were colonized with s. aureus and 22 (4.16%) were colonized with MRSA (27)

Another studied a cross-sectional study was conducted in Malaysia on healthy adult, Nasal carriage of Staphylococcus aureus among healthy adults showed that the prevalence of S. aureus nasal carriage was 23.4% from this One person was colonized with MRSA, which was different from the hospital strain. MRSA nasal colonization was found to be low outside of the health care environment (28)

A cross-sectional study conducted on 79 subjects with DFUs were assessed for nasal and DFU colonization with SA, including Methicillin-resistant-SA (MRSA) showed that Twenty-five (31.6%) subjects were positive for nares colonization with SA, while 29 (36.7%) subjects were positive for DFU colonization with SA . Seven (8.8%) subjects were positive for nares colonization with MRSA and seven (8.8%) subjects were positive for DFU colonization with MRSA. Longer duration of the ulcer was positively associated with the presence of MRSA in the ulcer (29) and similar studied conducted to assess on relationship between MRSA isolation from foot ulcers and nasal MRSA carriage Over a 12 month period, 65 consecutively attending patients with diabetic foot ulceration were recruited. There were 61% male and 85% with type 2 diabetes. Ulcers were neuropathic in 55%, ischemic in 14% and neuroischaemic in 31%. MRSA was isolated from 12 (19%) ulcers, and 11 (17%) had nasal carriage. Of the MRSA positive ulcer patients 7/12 (58%) had nasal MRSA carriage, compared with 4/53 (8%) with MRSA negative ulcers. the study conclude that nasal MRSA carriage in diabetic patients is a significant risk factor for foot ulcer MRSA infection(30).

The study was carried out between January 2004 and December 2004, to identify the frequency of *S. aureus* among diabetic and non-diabetic HD patients and to investigate resistance patterns against various antibiotics used broadly for treatment. The survey, 261 patients undergoing HD treatment from three HD units in Hatay were examined. A total of 148 *Staphylococcus aureus* strains were processed to assess their occurrence rates and antimicrobial susceptibility

profiles. *S. aureus* positivity was determined in 148 (56.7%) of the 261 HD patients and 26 (16.2%) of the 160 individuals in the control group. HD length was found to be 38.4 ± 24.3 months in the patients of *S. aureus* carrier and 27.3 ± 18.5 months in non-carrier patients. Significant correlation was also identified between durations those on HD and the isolation of *S. aureus* . However, the carrier state was unrelated to the presence of diabetes mellitus. age or sex. The studied conclude that nasal carriage of *S. aureus* was found to be more prevalent in HD patients than that in those in the control group. Also, it is concluded that DM was not a risk factor for the nasal carriage of *S. aureus*. In addition, the rates of antibiotic resistance of *S. aureus* strains were found to be quite higher in HD patients than in the control group **(31).**

The study conducted to assess the prevalence of methicillin-resistance among Staphylococcus aureus isolates in Africa. The study included articles published in 2005 or later reporting for the prevalence of MRSA among S. aureus clinical isolates. Thirty-two studies were included. In Tunisia, the prevalence of MRSA increased from 16% to 41% between 2002–2007, while in Libya it was 31% in 2007. In South Africa, the prevalence decreased from 36% in 2006 to 24% during 2007–2011. In Botswana, the prevalence varied from 23–44% between 2000–2007. In Algeria and Egypt, the prevalence was 45% and 52% between 2003–2005, respectively. In Nigeria, the prevalence was greater in the northern than the southern part and in the Ivory Coast, the prevalence was 39%, respectively. The prevalence of MRSA was lower than 50% in most of the African countries, although it appears to have risen since 2000 in many African countries, except for South Africa. A prospective study in both outpatients and inpatients of Ethiopia, which was performed during a 3-month period of 2006, reported a prevalence of MRSA of 55% (33).

2.2 Pathogenesis of *Staphylococcus aureus*

The nose is regarded as the major site of *S. aureus* carriage from where the organisms can spread to other parts of the body have shown that elimination of nasal carriage by using topical mupirocin also eliminates hand carriage. From these observations, it can be concluded that the nose provides an environment in which *S.aureus* can propagate and maintain itself for prolonged periods. The proposed pathogenesis for a number of endogenous infections would be that from the nose, the skin become colonized, causing subsequent infection in patients with impaired skin sites, e.g., in hemodialysis and CAPD patients and in patients with intravascular catheters (34).

In several studies, the elimination of nasal carriage reduced the incidence of *S. aureus* infections. a significant reduction in the rate of surgical-wound infection after intervention with mupirocin nasal ointment. Nasal treatment with mupirocin led to a reduction by a factor of four in the incidence of S. aureus bacteremia per patient year in carriers receiving hemodialysis. When the nares were treated topically to eliminate nasal carriage, *S. aureus* usually disappeared from other areas of the body. In patients receiving hemodialysis, 87 percent of those who carried *S. aureus* in their nares and on their hands carried the same strain at both sites. Treatment with topical mupirocin, which eliminates nasal carriage, also eliminates hand carriage. The proposed mechanism of pathogenesis for a number of endogenous infections is the colonization of the skin from the anterior nares, which causes subsequent infection in patients with areas of impaired skin, such as patients receiving dialysis and patients with intravascular catheters. (35).

The pathogenic process of *S. aureus* infection begins with colonization of host skin or mucosal surfaces and involves bacterial attachment to host cells often via components of the extracellular matrix. To persist, the organism produces molecules that decrease the effectiveness of complement mediated and antibody-mediated opsonophagocytosis and block effectors of host immune cell killing, such as reactive oxygen species and antimicrobial peptides. Ultimately, the organism expresses specific factors that damage host cells and degrade components of the extracellular matrix, contributing to persistence and facilitating spread within normally sterile sites of the host (36).

2.3 Methicillin Resistant *Staphylococcus aureus*

Strains of *S.aureus* have developed resistance to many commonly used antibiotics due to indiscriminate use of antibiotics. Staphylococcal resistance to penicillin is mediated by penicillinase (a form of β-lactamase) production: an enzyme which breaks down the β-lactam ring of the penicillin molecule. First report of a penicillin-resistant strain of *S. aureus* was published in 1945, revealing its association with penicillinase enzyme produced by the bacteria. The MRSA is a specific strain of the *S. aureus* bacterium that has developed antibiotic resistance to all penicillin‘s, including methicillin and other narrow-spectrum β-lactamase-resistant penicillin antibiotics (37).

3. Objectives

3.1 General objective

- To assess the rate of nasal *S. aureus* and MRSA associated factors among DM patients at Tikur Anbessa Specialized Diabetic clinic Hospital Addis Ababa, Ethiopia,

3.2 Specific objectives

- To determine the rate of nasal *S. aureus* among DM patients.
- To determine the rate of nasal MRSA among DM patients.
- To assess the associated factors for nasal carriage rate of *S. aureus* among DM patients.

4. Materials and methods

4.1 Study area

The study was conducted at Tikur Anbessa Specialized hospital Diabetic clinic, Addis Ababa, Ethiopia. Tikur Anbessa Specialized hospital is the largest hospital in the country compared to other hospitals, which is located at the center of the city in Lideta Sub city. The TASH, the largest referral hospital in the country, with 700 beds, was transferred to the School by the Federal Ministry of Health, and it has since become a University teaching hospital. Tikur Anbessa Specialized Hospital is now the main teaching hospital for both clinical and preclinical training of most disciplines. It is also an institution where specialized clinical services that are not available in other public or private institutions are rendered to the whole nation. The hospital receives referred patients from all parts of the country and provides local emergency service.

4.2 Study design and period

A facility based cross sectional was conducted at Tikur Anbessa Specialized hospital to determine the prevalence of *S.aureus* and MRSA and associated risk factor among DM patients attending in the hospital diabetic clinic from January to June, 2018.

4.3 Population

4.3.1 Source of Population

- All diabetic patients who sought medical services at TASH

4.3.2 Study population

- All diabetic patients attending in the diabetic clinic during the study period fulfilling the inclusion criteria.

4.4 Inclusion and Exclusion criteria

4.4.1 Inclusion

- All diabetic patients attending for care and treatment whose age greater than 18 years old and willing to participate during the study period were included.

4.4.2 Exclusion

- All diabetic patients who have taken antibiotic for the last 14 days and those patients who are seriously ill to give sample.

4.5 Study variables

4.5.1 Dependent variables

- Rate of S.aureus and MRSA

4.5.2 Independent variables

- Age
- Sex
- Educational status
- Occupation
- Type of DM
- Type of DM drug
- Hospital admission
- Blood glucose level
- Use of antibiotic
- Duration of DM

4.6 Measurement and data collection

4.6.1 Sample size determination

Sample size: it has to be determined using single population proportion sample size calculation formula, which is computed 422(including 10% non- response rate) by taking the prevalence is 50%.

$$n = \frac{(Z\ \alpha/2)^2 \times P\ (1 - P)}{d^2} = \frac{(1.96)^2 * 0.5(1-0.5)}{(0.5)^2} = 384 + 38 = 422$$

The sample size calculates are 422 participants will be needed.

P = prevalence

Z α/2=coefficient at level of significance (at 95% CI)

d = margin of error

4.6.2 Sampling technique

Systematic random sampling technique was used. The diabetes clinics provide their services three days per week but mainly on Mondays and Wednesdays. On average about 200 patients has

visited the DM clinic per week, about 800 patients visited per month. Based on the decision to collect data over the course of three months, the sampling interval has to be determined by dividing the expected number of diabetic patients over three months (2400) The sampling interval is determined by dividing the expected number of diabetic patients over three months (2400) by the sample size 422 which gives a sampling interval of about five. Sampling fraction is determined by selecting one number from 1-5 by lottery method. Thus, every 5th patient were selected

4.7 Data collection and laboratory methods

Data collection training was provided for nurses who are working at DM clinic, Tikur Anbessa Specialized Hospital. After obtaining an informed written consent, a pre-tested structured questionnaire was used to eligible participants for interview to obtain socio-demographic information and other information on risk factors for infection of *S.aureus* and MRSA.

4.7.1 Sample collection and laboratory analysis

Nasal swabs: Sterile cotton swabs were used to collect swabs from the anterior nares of the participants selected. Specimens for culture were obtained by firmly rotating a new pre moistened cotton-tipped swab against the anterior nasal mucosa for 3 second in each anterior nares (38)

Culture: Swabs were inoculated directly on to MSA (Oxoid). Plates were incubated at 37°C for 24 hours and left at room temperature to stimulate pigment formation. Then individual colonies was streak on to blood agar (oxoid) and incubated at 37°C overnight (39).

Identification of *S.aureus*

Identification of *S.aureus* were performed using observing color on MSA, Gram stain, observing hemolysis on BAP and production of catalase and coagulase using slide method and tube method. Those colonies showing yellowish golden pigment, beta-hemolysis on BAP, catalase and coagulase positive was considered as *S.aureus* (39).

Detection of MRSA strains by Cefoxitin disc diffusion methods

The isolated *S.aureus* was screened for methicillin resistance following the clinical Laboratory standard institute (CLSI, 2017) modified kirbaur disc diffusion method. Overnight cultures from

sheep blood agar plates was plated on Mueller-Hinton agar and Cefoxitin (30 µg) disk was placed on the inoculated plate. Zone diameters were measured using a ruler and a zone size of ≥ 22 mm after 24 hour incubation at 37 °C was considered as susceptible for Cefoxitin. While, those Isolates of *S. aureus* with an inhibition size of ≤21mm was considered as resistant to Cefoxitin and defined as MRSA.

Antibiotic Susceptibility testing

The antibiotic susceptibility testing was performed by modified Kirby-Bauer disc diffusion technique. The bacterial suspension was prepared by picking up 2-3 colonies from the pure culture with wire loop and emulsify in 2 ml of nutrient broth. The suspension was adjusted at 0.5% McFarland standard and streaked onto Mueller Hinton agar plates. The antibiotic susceptibility testing was performed on the following antibiotic discs; Cefoxitin (CXT, 30µg), Clindamycin (DA,2µg), Erythromycin (ERY,15µg) Trimethoprim Sulphamethoxazole (SXT, 1.25µg/23.75µg), Penicillin (PG, 10units) Ciprofloxacillin (CIP,5µg) and Tobramycine (TOB 10µg),Gentamicin (GEN, 10µg). After 18-24 hours incubation at 37^0c, zone of inhibition was measured and reported as susceptible (S) and resistance (R) based on the clinical and laboratory standard institute (CLSI) guidelines. Multidrug resistant (MDR) was defined as resistance to ≥3 antibiotic classes. *S. aureus* (ATCC-25923) was used as control for the antimicrobial susceptibility pattern (40).

4.8. Data Quality Assurance

The reliability of the study findings were guaranteed by implementing Quality Control (QC) measures throughout the whole process of the laboratory work. All materials, equipment, reagents and procedures were adequately controlled. Pre-analytical, analytical and post-analytical stages of quality assurance and Standard Operating Procedures (SOPs) were strictly followed. Pre-tested structured questionnaire guided interview were used for data collection on socio-demographic characteristics, clinical data and associated factors. Sterility of culture media and biochemical tests were checked by overnight incubation of un-inoculated media from each batch of preparation. A standard strain of *S. aureus* (ATCC-25923) was used as control during the biochemical and susceptibility testing. To standardize the inoculums density of bacterial suspension for the susceptibility test, 0.5 McFarland standard was used (40).

4.9 Data analysis and interpretation

Data were entered, cleaned and analyzed using SPSS version 20. Bivariate logistic regression analysis were used to determine the association between independent variables and the outcome variables. All independent variables with a p-value less than or equal to 0.05 in the analysis were included in the bivariate logistic regression model to identify variables which were associated independently. Odds ratio (OR) and their 95% confidence intervals (CI) was calculated and the result was considered statistically significant at $p < 0.05$.

4.10. Ethical considerations

Ethical approval of this study was reviewed and approved by the "Department of Research and Ethical Review Committee" of the Medical Laboratory Sciences, College of Health Sciences, and Addis Ababa University. Ethical clearance was obtained from Tikur Anbessa specialized hospital administration. The purpose and importance of the study was explained to each study participants. To ensure confidentiality of participant's information, code number was used where by the name of the participant and any identifier of participants was written on the questionnaire. Participant was interviewed alone to keep the privacy. All participants were pay for test. Test results were given to the clinicians who are working on Diabetic clinic of the Hospital for further diagnosis and management.

4.11. Dissemination of results

The findings were presented to Addis Ababa University College of health science, Department of Medical Laboratory Sciences. It will also be disseminated through presentation on local or scientific conferences and it would be published on journal.

Operational definition

Methicillin-resistant Staphylococcus aureus: Is defined as the strains of *S. aureus* that are resistant to cefoxitin ((30 µg) using disk diffusion method on Mueller Hinton agar.

Multi drug resistance: is antimicrobial resistance shown by a species of microorganism at least to one drug in three different classes of antibiotics

5. Results

5.1 Socio-demographic characteristics

A total of 422 DM patient 214 female and 208 male were recruited. The mean age of study participants was 49.8 (SD=15.9, range 18 to 90) years. The majority (387/422, 91.7%) was urban in residence; had attained college and university level (137/422, 32.5%) education and were employed (139/422, 32.9%) (Table 1).

Table1. Socio-demographic characteristics associated with *Staphylococcus aureus* isolate among diabetes mellitus patients at Tikur Anbesa Specialized Hospital, Addis Ababa, Ethiopia, 2018.

Socio-demographic characteristics		**DM patients N=422(%)**	**Total *S. aureus* isolates N (%), 47(11.1%)**		**COR(95%CI)**	***P value***
			Neg.	**Pos.**		
Sex	Female	214(50.7)	193(90.2)	21(9.8)	1	
	Male	208(49.3)	182(87.5)	26(12.5)	1.3(0.714-2.416)	0.381
Age in years	< 40	106(25.1)	96(90.6)	10(9.4)	1	
	≥ 40	316(74.9)	279(88.3)	37(11.7)	1.3(0.610-2.658)	0.520
Residence	Rural	35(8.3)	30(85.7)	5(14.3)	1.4(0.504-3.719)	0.538
	Urban	387(91.7)	345(89.1)	42(10.9)	1	
Education	Illiterate	56(13.3)	50(89.3)	6(10.7)	1.2(.0412-3.264)	0.779
	1-8	101(23.9)	87(86.1)	14(13.9)	1.6(0.685-3.531)	0.291
	9-12	128(30.3)	116(90.6)	12(9.4)	1	
	College and above	137(32.5)	122(89.1)	15(10.9)	1.2(0.534-2.646)	0.672
Occupation	Employee	139(32.9)	120(86.3)	19(13.7)	2.3(0.886-6.888)	0.084
	House wife	108(25.6)	95(88.0)	13(12.0)	2.1(0.729-6.249)	0.166
	Merchant	38(9)	34(89.5)	4(10.5)	1.8(0.464-7.259)	0.387
	Retire	84(19.9)	78(94.0)	5(6.0)	1.9(0.564-6.739)	0.291
	Others	54(12.8)	48(88.9)	6(11.1)	1	

5.2 Prevalence of *Staphylococcus aureus* and methicillin resistant *S. aureus* from Dm patients

Nasal swab culture was positive for 47/422(11.1%) of DM patients. Of these, methicillin resistant *S. aureus* was isolated from one DM patients. Even though none of the assessed socio-demographic factors showed a significant association with nasal *S.aureus* colonization in a bivariate logistic regression analysis, high prevalence of *S.aureus* was observed in 14.3% of rural resident, 12.5% of females, 11.7% of those who aged greater or equal to 40 years, 13.9% primary (1-8 grade) education level and 13.7% employees (Table 1).

Regarding the MRSA, only one (1/47) *S.aureus* isolate was resistant to cefoxitin, a surrogate marker for MRSA (Oxacilin). The MRSA strain was isolated from a 67 retire man who had uncontrolled blood glucose type II DM despite insulin treatment and had history of foot wound (data supplementary excel file).

5.3 Factors associated with *S. aurues* isolates among diabetes mellitus patients

Out of 422 DM patients, 312(73.9%) had Type II DM, 257(60.9%) had uncontrolled blood glucose level , 30(7.1%) had nasal infection and foot wound. Further, 54(12.8%) had hospital admission history in the last 6 months, 75(17.8% had previous history of antibiotic use in the last 6 months and 263(5.2%) were currently using insulin injection while 22(5.2%) use both insulin and other oral drugs for DM treatment. The rate of S.aureus isolation was significantly higher in those who are currently take a combination of both insulin and other oral drugs (odds ratio [AOR], 3.985, 95% CI, 1.270-12.437; $p<0.017$).than those who use oral drugs only. Use of combined DM drug and S.aureus isolation was found to be statistically significant (Table 2).

Table2. Factors associated with *S. aurues* among diabetes mellitus patients at Tikur Anbessa Specialized Hospital, Addis Ababa, Ethiopia, 2018.

Characteristics of DM patients		DM patients N=422(%)	Total *S. aureus* isolates N (%), 47(11.1%)		COR(95%CI)	*P value*
			Neg.	Pos.		
Type of DM	Type 1	110(26.1)	101(91.8)	9(8.2)	1	
	Type 2	312(73.9)	274(87.8)	38(12.2)	1.6(0.727-3.333)	0.255
Duration of DM	Less than 1year	22(5.2)	20(90.9)	2(9.1)	1.2(0.212-6.566)	0.850
	1-9 years	201(47.6)	174(86.6)	27(13.4)	1.8(0.674-4.972)	0.235
	10-19 years	135(32.0)	122(90.4)	13(9.6)	1.3(0.428-3.692)	0.677
	≥20 years	64(15.2)	59(92.2)	5(7.8)	1	
Type of DM treatment	Combination	22(5.2)	16(72.7)	6(27.3)	3.6(1.224-10.585)	**0.020**
	Insulin	241(57.1)	215(89.2)	26(10.8)	1.2(0.594-2.268)	0.662
	Oral drug	159(37.7)	144(90.6)	15(9.4)	1	
History of foot wound	No	392(92.90	350(89.3)	42(10.7)	1	
	Yes	30(7.1)	25(83.3)	5(16.7)	1.6(0.606-4.586)	0.323
Nasal infection	No	392(92.9)	347(88.5)	45(11.5)	1.8(0.418-7.879)	0.426
	Yes	30(7.1)	28(93.3)	2(6.7)	1	
History of hospital admission	No	368(87.2)	328(89.1)	40(10.9)	1	
	Yes	54(12.8)	47(87)	7(13.0)	1.2(0.517-2.884)	0.648
History of previous antibiotic taken	No	347(82.2)	307(88.5)	40(11.5)	1.3(0.544-2.946)	0.585
	Yes	75(17.8)	68(90.7)	7(9.3)	1	
Blood sugar level	Controlled	165(39.1)	151(91.5)	14(8.5)	1	
	Uncontrolled	257(60.9)	224(87.2)	33(12.8)	1.6(0.823-3.069)	0.168

DM, diabetes mellitus; N, total number of participants; COR, crude odds ratio; CI, confidence interval; neg, negative; pos, positive, 1 is reference category

Table 2 .Factors associated with *S. aurues* among diabetes mellitus patients at Tikur Anbessa Specialized Hospital, Addis Ababa, Ethiopia,

Characteristics of DM patients		**DM patients N=422(%)**	**Total *S. aureus* isolates N (%), 47(11.1%)**		**COR(95%CI)**	**P value**	**AOR(95%CI)**	**P value**
			Neg.	**Pos.**				
Type of DM	Type 1	110(26.1)	101(91.8)	9(8.2)	1			
	Type 2	312(73.9)	274(87.8)	38(12.2)	1.6(0.727-3.333)	0.255	1.414(0.63-3.263)	0.416
Duration of DM	Less than 1 year	22(5.2)	20(90.9)	2(9.1)	1.2(0.212-6.566)	0.850	1.731(0.285-0.494)	0.551
	1-9 years	201(47.6)	174(86.6)	27(13.4)	1.8(0.674-4.972)	0.235	2.040(0.726-5.735)	0.176
	10-19 years	135(32.0)	122(90.4)	13(9.6)	1.3(0.428-3.692)	0.677	1.316(.442-3.921)	0.622
	≥20 years	64(15.2)	59(92.2)	5(7.8)	1			
Type of DM treatment	Combination	22(5.2)	16(72.7)	6(27.3)	3.6(1.224-10.585)	**0.020**	3.985(1.27-12.437)	**0.017**
	Insulin	241(57.1)	215(89.2)	26(10.8)	1.2(0.594-2.268)	0.662	1.508(0.708-3.210)	0.287
	Oral drug	159(37.7)	144(90.6)	15(9.4)	1			
History of foot wound	No	392(92.90	350(89.3)	42(10.7)	1			
	Yes	30(7.1)	25(83.3)	5(16.7)	1.6(0.606-4.586)	0.323	1.808(0.625-5.225)	0.274
Nasal infection	No	392(92.9)	347(88.5)	45(11.5)	1			
	Yes	30(7.1)	28(93.3)	2(6.7)	0.551(.127-2.390)	0.426	0.638(0.143-2.845)	0.556
History of hospital admission	No	368(87.2)	328(89.1)	40(10.9)	1			
	Yes	54(12.8)	47(87)	7(13.0)	1.2(0.517-2.884)	0.648	0.498(1.221-2.994)	0.663
History of	No	347(82.2)	307(88.5)	40(11.5)	1.3(0.544-2.946)	0.585	1.600(0.659-3.886)	0.299

previous antibiotic taken	Yes	75(17.8)	68(90.7)	7(9.3)	1				
Blood sugar level	Controlled	165(39.1)	151(91.5)	14(8.5)	1				
	Uncontrolled	257(60.9)	224(87.2)	33(12.8)	1.6(0.823-3.069)	0.168	1.486(0.753-2.933)	0.254	

DM, diabetes mellitus; N, total number of participants; COR, crude odds ratio; AOR adjusted odds ratio CI, confidence interval; neg, negative; pos, positive, 1 is reference category

5.4 Antimicrobial susceptibility pattern

Out of the 47 *S.aureus* isolates, penicillin 46(91.5%), erythromycin 6(12.8%), Gentamicin 3(6.3) Tobramycin 3(6.3), cotrimoxazole 2(4.2%), Ciprofloxacin 2(4.2%) and cefoxitin 1(2.1%) were resistant. On the other hand, all the *S.aureus* isolates were sensitive to clindamycin (Table 3).

Table3.Antibiotic resistance pattern of *S.aureus* among DM patients at Tikur Anbessa Specialized Hospital, Addis Ababa, Ethiopia, 2018.

Type of antibiotics	Antibiotic susceptibility pattern of *S.aureus*	
	Resistant (%)	Susceptible (%)
Penicillin	46(91.5)	1(2.1)
Erythromycin	6(12.8)	41(87.2)
Clindamycin	0(0)	47(100)
Cefoxitin	1(2.1)	46(97.9)
Trimethoprim Sulphamethoxazole	2(4.2)	45(95.7)
Gentamicin	3(6.3)	44(93.6)
Tobramycin	3(6.3)	44(93.6)
Ciprofloxacin	2(4.2)	45(95.7)

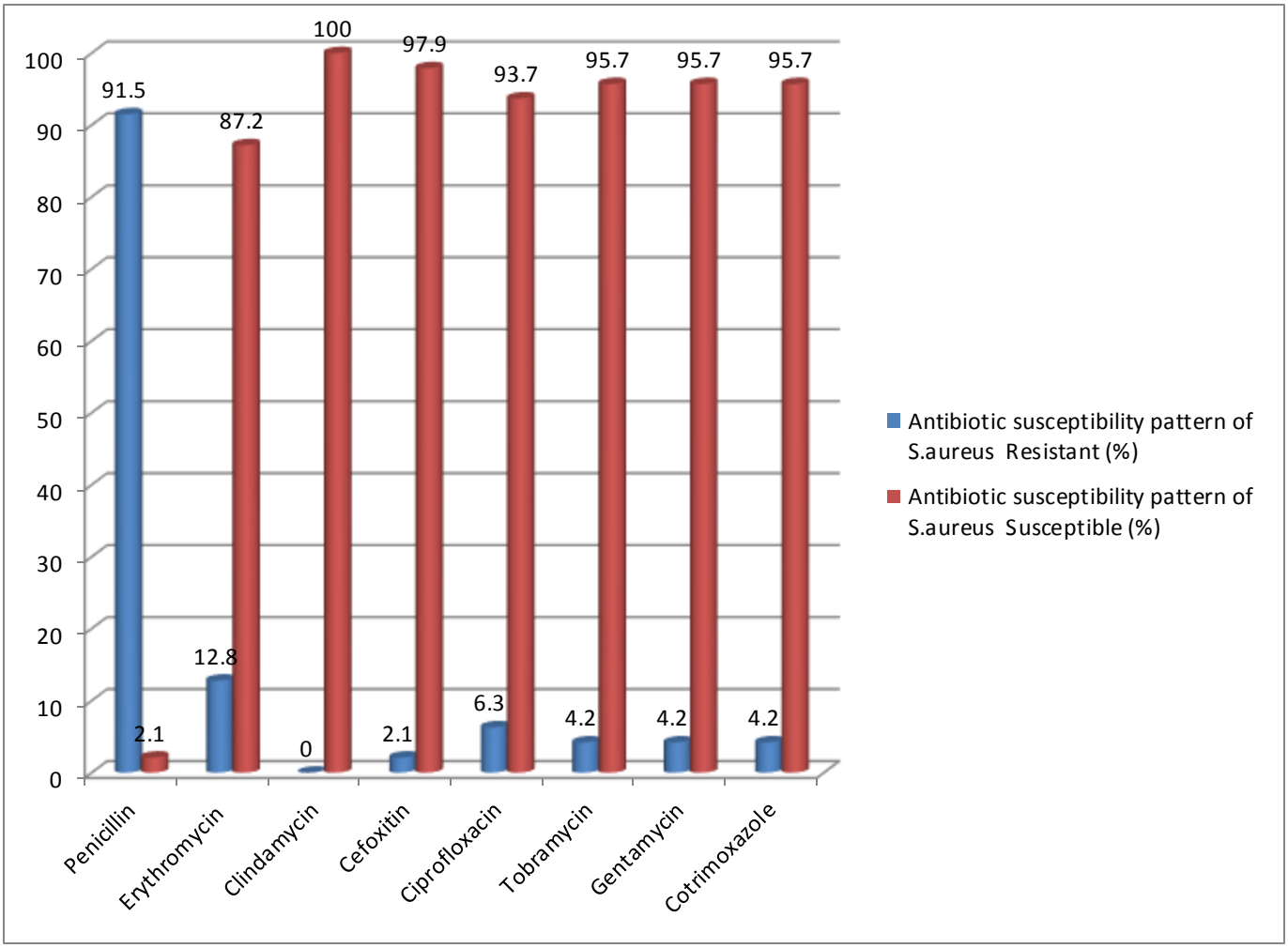

Figure 1: Antibiotic-resistance profiles of *S.aureus* isolates from nasal swabs

5.5 Multidrug resistance pattern of S.aureus

Out of the 47 isolates of *S.aureus*, 4/47 (8.5) isolates showed MDR. Multi drug resistance: is antimicrobial resistance shown by a species of microorganism at least to one drug in three different classes of antibiotics.

Table 4. Multidrug-resistance pattern of *S. aureus* among DM patients at Tikur Anbessa Specialized Hospital DM clinic, Addis Ababa, Ethiopia, 2018

No. of antibiotics	Drug resistance pattern	Resistance strains no (%)
3	Pen/E/SXT Pen/Genta/CIP	2(4.2)
4	Pen/Ery/SXT/Genta	1 (2.1)
5	Pen/Ery/Cip /Cefo/Genta	1 (2.1)

6. Discussion

S. aureus colonization is a risk factor for subsequent infection caused by the colonizing clone MRSA is a global public health problem. The MRSA infections can be found in both hospitals and the community. Generally, hospital-associated MRSA (HA-MRSA) infects hospitalized individuals with predisposing risk factors (17).

In this study, overall nasal carriage rate of *S.aureus* and MRSA from nasal swab culture was positive for *S.aureus* 47/422(11.1%) of DM patients. Of these, methicillin resistant *S. aureus* was isolated from one DM patients. It is lower than the study conducted in India by Rani et al **,**hospital based study on diabetic patients the prevalence of *S.aureus* was 38/60 (63.3%) and from this MRSA isolates from positive diabetic patients 38 (63.33%) were 7 (18.42%) (45) .Our study also lower than the study conducted in turkey on the *Staphylococcus aureus* nasal carriage rate and associated factor in diabetic patient showed that, the nasal carriage rate of *S.aureus* and MRSA on outpatient were 35.3% and 15.8 % respectively (2). And the study done in Colorado by Lipsky, B.A NIDDM Prevalence of nasal *Staphylococcus aureus* colonization was 27% (50/188)(42). The difference may be due to the sample size and method of detection.

Similar study done in Turkey by Tamer , on nasal carriage rate and associated factor among type- II diabetic patient, NCSA were 22.2 %(33/148) (43) and another Cross sectional community-based _study on diabetes_done by Hart J. in 660 DM patients 258 patients (39.1%) were positive for *S. aureus* and eight (3.1%) carried MRSA. It is higher than our study. The difference may be due to the sample size and study group also difference in technique of swab sample (47).

A study conducted in the endocrinology clinic of Shahid Behesty hospital in Iran on the Risk Factors of Methicillin-Resistant Staphylococcus aureus Colonization in Diabetic Outpatients, reported that nasal colonization of S. aureus among 494 DM patient was 42.5% (210/494). Of these, 122 (58%) were MRSA (24.6% of all DM patients) which is higher than our study (26).

Our study result was also lower than the study carried to identify the frequency of S. aureus among diabetic hemodialysis patients. The survey, 261 patients undergoing HD treatment from three units in Hatay was examined. A total of 148 *Staphy-lococcus aureus* strains were processed to assess their occurrence rates and antimicrobial susceptibility profiles. *S.*

aureus positivity was determined in 148 (56.7%) of the 261 HD patients (31) and the study done among patients with type-ii diabetes on dialysis done in Saudi Arbyia Overall prevalence was 38% (79/208) of HD patients were found to be nasal carriers of S. aureus including 9.6% (20/208) with MRSA (46). The difference might be due the study participants were also having renal failure and have prolonged hospital stays and isolation technique.

In this study, nasal carriage rate of *S.aureus* and MRSA from nasal swab culture was higher in China 46 (8.70%) were colonized with *s. aureus* and 22 (4.16%) were colonized with MRSA but nasal carriage rate of MRSA is high (27) this might be due to the method of using identification of MRSA and the community based study done in USA by Lin J et al on the prevalence of *S.aureus*,28.32% (286/1010) but the prevalence of MRSA 1.09% (11/1010) , is lower than our study, the difference might be due to the study method and the participants were selected from the community, and may not visit hospitals (44).

The prospective cohort study done in Turkey on the prevalence and risk factors for MRSA colonization in a population of outpatients with diabetes, 127/304 (41.9%) were colonized with S aureus and 30 (9.9%) were colonized with MRSA. Overall, 23.6% of all *S aureus* isolates was MRSA(41) and the study done in Taiwan by Lin S-Y et.al nasal *S. aureus* carriage rate was 16.4% (58/354) and nasal MRSA carriage rate was 2.8% (10/354 were higher than our study (48) the difference might be due to methodological difference.

Out of the 47 *S.aureus* isolates, penicillin 46(91.5%), erythromycin 6(12.8%), Gentamicin 3(6.3) Tobramycin 3(6.3), Cotrimoxazole 2(4.2%), Ciprofloxacin 2(4.2%) and cefoxitin 1(2.1%) were resistant. On the other hand, all the *S.aureus* isolates were sensitive to clindamycin

From the above statistics, we can know that the prevalence of *S. aureus* and MRSA nasal carriage among diabetic population were different in different countries and regions, and diabetic population might be more likely to carry *S. aureus* and MRSA.

7. Strength and limitation of the study

Strength

- As far as concern it is the first study of nasal carriage rate on diabetic patient
- Show the magnitude of *S.aureus* among diabetic patient and drug resistance pattern

Limitation

The study has some limitations. .

- The nasal swab results may give an indication of the prevalence of methicillin-sensitive *S. aureus* and MRSA in the community, but the MRSA strains in the community were not identified

8. Conclusion and recommendations

8.1 Conclusion:

The prevalence of *S.aureus* among DM patients was low in this study. Also, the carriage rate of MRSA was very low in this study compare to other study conducted; performing culture and sensitivity test for those patients reduce the complication of infection. Administering appropriate antibiotics have an important role in minimizing the resistance burden of bacterial species among diabetic patients. The rates of *S.aureus* isolation was significantly higher in those who are currently take a combination of both insulin and other oral drugs than those who use oral drugs only. Use of combined DM drug and S.aureus isolation was found to be statistically significant.

8.2 Recommendations

- Diabetic patients screened for nasal carriage *S. aureus* and MRSA,
- Effective methods of infection prevention control should be utilized to help reduce the incidence of *S.aureus* and MRSA infections.
- The importance of implementing strategies to eliminate nasal carriage of *S. aureus* to prevent the spread of infection is necessary.
- Further multicenter large scale studies should be conducted to identify the associated risk factors and antibiotic resistance pattern in DM patients.

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Annexes

Annex I: Information sheet

Purpose

We were conducting a research to assess prevalence of *S.aureus* and MRSA and associated factors among DM patients in Tiruk Anbessa Specialized Hospital, Addis Ababa. Your feedback on this research is important and will help to prevent co-infection and further complication of DM patients

Participation

We are asking you to voluntarily participate in this study. What is expected from you is to respond questions which may take 10 minutes and give nasalswab. The nasal swab sample is collected using sterile and moisturized cotton swab.

Benefits

If you have positive result with *S.aureus* and MRSA during investigation, an opportunities for treatment and follow up and prevent further complication due to infection.

Risks

There is no risk but there may be minimal discomfort during nasal swab collection.

Confidentiality

All the data obtained were kept strictly confidential by using only code numbers which is filled by the investigators and locking the data.

Right to refuse

Since participation in this study is entirely voluntarily, you can refuse or withdraw to participate in this study at any time. Your refusal will not affect your job or services given in the hospital.

If you have any question concerning the study you can ask with the following address

Principal investigator: Adane Amera

Address: Addis Ababa University College of Health science, Department Medical Laboratory Science.

Tel: 0963177054, Email address: ameraad@gmail.com

Addis Ababa, Ethiopia

Annex II: Consent Form

I have read the information above, or it has been read to me. I have been given the opportunity to ask questions and my questions have been answered to my satisfaction. I voluntarily consent that I would participate in this study, to give nasal swab and a participant in this study and understand that I have the right to withdraw from the study at any time

Name of participant _____date ____ /____ /____signature

Name of data collector _____date _____/____/_____ signiture _____

Annex IV: Questionnaire

A questionnaire prepared to assess the prevalence of nasal *S.aureus* and MRSA and associated factors among DM patients at TikurAnbessa Specialized Hospital, Addis Ababa.

PART I: Socio-demographic characteristics

Direction**:** Fill or use √ mark on box for response categories

S. No	Questions	Response of categories	Remark
1	Can you tell me your age?	_____year	
2	Sex	• Male • Female	
4	What is your educational status?	• Illiterate • 1-8 • 9-12 • diploma • University degree & above	
5	What is your occupational status?	1. Employed 2. House wife/home activities 3. Daily laborer 4. Merchant 5. Student 6. Other _____	
6	Monthly Income	• 1500 • 1501-3000 • 3001- 4500 • >4500	

Part II: Associated factors associated with *S.aurues* and MRSA among DM patients included in this study at TikurAnbessa Specialized Hospital, Addis Ababa, Ethiopia, 2018

S. No	Questions	Response of categories	Remark
1	Type of DM?	• DM Type 1 • DM Type 2	
2	Duration of DM (starting the date of confirmed DM)	• -------week • ---------month • ----------year	
3	Type of drug has been taken for treatment of DM	• Insulin • Oral drug • Combination(Insulin and Oral)	
4	Do you have a foot wound?	• Yes • No	
5	Do you have current nasal infection	• Yes, • No	
6	Do you have history of hospitalization for the last six month	• Yes • No	
7	Do you have taken antibiotic before six months?	• Yes • No	

Amharic translation of the consent information sheet, and consent form

Annex III:: Amharic Version Study Participant Information and Consent Form

የመረጃና የስምምነት ዉል ቅጽ

ነ የጥናቱ ዓላማ -የዚህ ጥናት አላማ በስኳር ህሙማን አፍንጫ ላይ ያለውን የስታፊሎ ኮከስ አሬስ የሚባለውን የባክቴሪያ እና ሜቲሲሊን የተባለዉን ፀረ-ባክቴሪያ የተቋቋሙ የባክቴሪያ ዝርያዎችን በአዲስ አበባ ጥቁር አንበሳ ስፔሻላይዝድ ሆስፒታል የስኳር ህሙማን ክሊኒክ በሚታከሙ ህመምተኞች ውስጥ መኖራቸውን መለየትና በምን ያህል መጠን እንደሚገኙ ማሳወቅ ነው፡፡ በዚህ ጥናት የእርሶ ተሳትፎ የነላ ጠቀሜታ ያለው ሲሆን ከስኳር ህመም በተጫማሪ በዚህ ባክቴሪያ ችግር ምክንያት የሚከሰተውን ሕመም ለመከላከል መሠረታዊ መረጃ ይሰጣል፡፡

2 የተሳትፎ ሂደት- ይህን ጥናት በአዲስ አበባ ጥቁር አንበሳ ስፔሻላይዝድ ሆስፒታል ለመስራት የጥናቱ ተሳታፊ እንዲሆኑ ተጋበዘዋል፡፡ለመሳተፍ ፍቃደኛ ከሆኑ የጥናቱን ዓላማ መረዳትና ፍቃደኝነትዎን መግለፅ ይጠበቃል፡፡ለዚህ ጥናት የሚያስፈልገው ከአፍንጫ ላይ ናሙና ከስኳር ታከሚወች በመውሰድ ሲሆን የሚወስድበዎት ጊዜም ነ0 ደቂቃ ብቻ ይሆናል፡፡

3. ጥቅሞች - በምርመራ ውጤትዎ መሠረት ተጨማሪ ሕክምና ያገኛሉ በተጨማሪም በዚህ ባክቴሪያ ምክንያት የሚመጣውን ተጨማሪ ህመም እንዳይፈጠር ይከላከላል፡፡

4. ከጥናቱ ጋር ተያይዞ የሚመጣ ጉዳት - በዚህ ጥናት ዝርዝር ላሰራር ሂደት ውስጥ አካላዊ ወይም አእምሮአዊ ጉዳት አይኖርም፡፡ ነገር ግን ናሙናዉ በሚወሰድበት ጊዜ መጠነኛ የሆነ የህመም ስሜት ሊሰማዎት ይችላል፡፡ ቢሆንም ይህ የህመም ስሜት ምንም አይነት ችግር አያመጣብዎትም፡፡

5. የጥናቱ መረጃ ሚስጥራዊነት- ሁሉም ከተሳታፊዎች የሚሰበሰቡ መረጃዎች በሚስጢር የሚያዙ እና የሚጠበቁ ይሆናሉ፡፡ በማንኛውም ምክኒያት ተሳታፊዎች እነማን መሆናቸውን የሚያሳይ በመጠይቁ ይሁን በሌላ ነገር አይኖርም፡፡ የተሰበሰቡ መረጃዎች ለሶስተኛ ወገን ተላልፎ አይሰጥም፡፡ በተጨማሪም ውጤቱ የሚለካዉ ይሁን ተሰብስቦ የሚያዘው በዋና አጥኚ ነው፡፡

7 የመዉጣት (የማቋረጥ) መብት - በዚህ ጥናት ላይ መሳተፍዎ በሙሉ ፍቃደኝነት ላይ የተመሰረተ ነው ።ጥናቱን የማቋረጥ ሙሉ መብት አለዎት ፡፡ ናሙናም ሆነ ለመጠይቁ መልስ ያለመስጠት ከሆስፒታሉ የሚያገኙትን ማነኛውንም አገልግሎት አይገድብም ፡፡

8 ጥያቄ ካለዎት ተጨማሪ መረጃ ከፈለጉ በማንኛዉም ጊዜ ከዚህ በታች የተጠቀሰዉን አድራሻ መጠቀም ይችላሉ፡፡

አዳነ አመራ ዋና ተመራማሪ:

ስ.ቁ: +251910922176 ኢ.ሜል ameraad@gmail.com

የስምምነት ዉል

የስምምነት ማረጋገጫ ፊርማ ቀን----------

እኔ ከዚህ በታች ስሜ የተጠቀሰዉና የፈረምኩት የጥናቱ ተሳታፊ በስኳር ህሙማን አፍንጫ ላይ ያለውን የስታፊሎ ኮከስ ኦሬስ የሚባለውን የባክቴሪ እንና ሜቲሲሊን የተባለዉን ፀረ-ባክቴሪያ የተቋቋሙ የባክቴሪያ ዝርያዎችን በአዲስ አበባ ጥቁር አንበሳ ስፔሻላይዝድ ሆስፒታል በሚታከሙ ህመምተኞች ውስጥ መኖራቸውን መለየትና በምን ያህል መጠን እንደሚገኙ ማሳወቅ የሚሰራውን ጥናት አላማና ጥቅም በሚገባ ተረድቻለሁ፡፡ ጥናቱ ላይ መሳተፍም ሆነ አለመሳተፍ በራሴ ፍቃድ የሚወሰን መሆኑም ተገልጾልኛል፡፡ በተጨማሪም ከጥናቱ ባልሳተፍም ሆነ አቋርጬ ብወጣ ከሆስፒታሉ በማገኘዉ የህክምና አገልግሎት ምንም አይነት ችግር እንደማይደርስብኝ ተነግሮኛል፡፡

በመሆኑም አስፈላጊውን መረጃ መስጠት እና ከአፍንጫዮ ናሙና መዉሰድ አስፈላጊ መሆኑን ስለተስማማሁበት ለመስጠት ሙሉ ፈቃደኛ መሆኔን በፊርማዬ እገልጻለሁ፡፡

የተሳታፊ ስምና ----------ፊርማ _____ቀን _____

መረጃ ሰብሳቢው ስም ----------ፊረማ----------ቀን _____

በአማርኛ የተዘጋጀ መጠይቅ

ክፍል1 የማህበራዊና ዲሞግራፊያዊ ሁኔታ

መመሪያ: የሚሰጠው ምላሽ በጽሁፍ መሙላት ወይም ምልክት መጠቀም

ባለፉት 14 ቀን ፀረ-ባክቴሪያ መድሀኒት የወሰዱ ከሆነ የጥናቱ ተሳታፊ አይሆኑም

የኮድ ቁጥር----------የካርድ ቁጥር ----------ናሙናየተወሰደበትቀን……

1.እድሜዎ ስንት ነው?_____ዓመት

2. ጾታ 1. ወንድ 2.ሴት

3. የሚኖሩትየትነው?

1. ከተማ
2. ገጠር

4.የትምህርትሁኔታ?

1.ያልተማረ 3. ሁለተኛደረጃ /9-12

2. የመጀመሪያያደረጃ /1-8/ 4. ዲፕሎማ 5.ዩኒቨርሲቲድግሪናከዚያበላይ

5.የሥራሁኔታ

1. ተቀጣሪ 3. የቀንሰራተኛ

2. የቤትእመቤት 4. ነጋዴ

5.ተማሪ 6.ሌላከሆነ ይግለጽ _____

6.ወርሃዊ ገቢ /በኢትዮጵያብር/

1.<1500 3. 3001-4500

2. 1501-3000 4. >4500

ክፍል II

1.የስኳርበሽታአይነት

1. አይነት 1 2. አይነት 2

2. ህክምና ክትትል ከጀመሩ ምን ያህል ጊዜ ነዉ……?

3. የሚወሰዱት የስኳር መድሃኒት አይነት

1. ኢንሱሊን 2. በአፍ የሚዋጥ 3. ኢንሱሊን እና በአፍ የሚዋጥ

4. የደም ስኳር መጠን FBS.....HgA1C.....

1. የተስተካከለ (controlled) 2. ያልተስተካከለ(uncontrolled)

5. የአፍነጫኢንፌክሽንታመዋል

1. አዎ 2. የለም

6. እግር ለይ ቁስል አለ

1. አለ 2. የለም

7. ባለፉት 6 ወራትፀረ-ባክቴሪያ መድሀኒትወስደል?

1. አዎ 2. የለም

3. መቼ……

3. ከወሰዱ የፀረ ባክቴሪያዉ መድሀኒት አይነት……

8.ባለፉት 6 ወራት ታመዉ ሆስፒታል ተኝተዉ ታክመዋል?

1. አዎ 2. የለም

አመሰግናለሁ!!!

Declaration

The undersigned declares that this thesis complies with the regulations of the University and meets the accepted standards with respect to originality and quality. The PI also agrees to accept responsibility for the scientific ethical and technical conduct of the research project and for provision of required progress reports.

Principal investigator (PI):

Adane Amera

Signature: _____

Date of submission: _____

This thesis has been submitted with my approval as an advisor.

Advisor:

Kassu Desta (MSc, PhD candidate.)

Signature: _____

Date: _____

Place: Addis Ababa, Ethiopia.