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RESEARCH PAPER

Occurrence, Associated factors, and Antibiogram Profiles of *Staphylococcus aureus* and *Salmonella* Typhi Among Food Handlers in Restaurants in Hawassa City, Ethiopia

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Abstract

Staphylococcus aureus and Salmonella Typhi are among the most common foodborne pathogens transmitted by asymptomatic food handlers. This study aimed at determining the occurrence and antibiogram profiles of S. Typhi and S. aureus among food handlers in restaurants in Hawassa City, Sidama Regional State, Ethiopia. A cross-sectional study was conducted from August to November 2020, utilizing laboratory analysis of nasal swabs and stool samples from 100 volunteer food handlers working in restaurants in Hawassa City based on standard culture and antibiotic sensitivity tests. Ten (10%) of the 100 food handlers were nasal carriers of S. aureus whereas no S. Typhi was found. Five of the ten S. aureus isolates were resistant to one or more of the seven antibiotics tested. Of the isolates that showed resistance, one showed single resistance to penicillin while four (40%) showed resistance to two or more drugs. Two isolates showed double resistance, and one isolate showed triple resistance. Of the multiple resistant isolates, one belonged to Methicillin-resistant S. aureus (MRSA), which was resistant to penicillin, tetracycline, erythromycin, and oxacillin. All the S. aureus isolates were vancomycin sensitive. This finding implies that food handlers may be a potential source of foodborne disease outbreaks in the community, and the finding of multiple resistant isolates suggests inappropriate use of antibiotics. This highlights the importance of educating food handlers about the necessity of hhygienic food handling and responsible use of antimicrobials.

Keywords: Antibiogram profiles, asymptomatic carriers, food handlers, Salmonella, Staphylococcus

Introduction

Food safety is the conditions and practices that encompasses all measures taken throughout the food chain, from production to consumption, to ensure that food is safe, free from harmful contaminants, and fit for consumption. It is a widespread public health issue in developing countries, where personal hygiene and food safety measures are not well understood and received little attention (Lund and OBrien, 2011). Each year, an estimated 30% of the population in developing countries suffers from foodborne diseases, and up to two million of them die (Steven *et al*., 2019). Foodborne diseases are also common in Ethiopia because of the prevailing high rate of adulteration, poor food hygiene and sanitation practices, inadequate food safety laws, and weak regulatory systems in the production and supply value chains (USAID, 2021). Moreover, the lack of formal education and training of food handlers often results in mishandling and inappropriate hygiene measures that allow contamination of food by pathogenic microorganisms. As a result, food handlers with no formal education or training in food safety and hygiene serve as sources of transmission of foodborne infections (Castro *et al*., 2016; Angelo *et al*., 2017; Beyene *et al*., 2019).

The problem is exacerbated by the existence of food handlers who harbor foodborne pathogens without showing signs and symptoms of diseases. Some opportunistic pathogens are harbored as part of the normal flora of the human body, as in the case of *Staphylococcus aureus,* while others, such as *Salmonella enterica serovar* Typhi, may lead to an asymptomatic carrier state after bouts of typhoid fever in a significant proportion of individuals (Levine *et al*., 1982). Food handlers, who asymptomatically harbor pathogenic microorganisms*,* make the prevention and control of foodborne disease outbreaks caused by these pathogens difficult because they are unlikely to seek medical care (Smith and Fratamico, 2018). Epidemiological evidence shows that most outbreaks of staphylococcal food poisoning follow contamination of cooked foods by persons carrying enterotoxigenic staphylococci in their nares or skin, and outbreaks of foodborne salmonellosis come from raw or cooked foods not subjected to further cooking after contact with bare hands (Angelillo *et al*., 2000). Food handlers are crucial to safety and prevention of foodborne illnesses, because they may introduce pathogens into foods during production, processing, distribution, and preparation.

This problem is a dire public health issue when foodborne pathogens are multidrug-resistant strains. As is true for many bacterial infections, the evolution and emergence of antimicrobial resistance complicates the problem of foodborne microbial diseases. The emergence of antimicrobial-resistant (AMR) bacteria threatens the effective and successful treatment of infectious diseases worldwide. The World Health Organization (2018) reported that the global consumption of antibiotics by humans has increased in the past two decades, primarily in low- and middle-income countries. Furthermore, there has been a shift towards the use of broad-spectrum and last-resort antibiotics. Over-the-counter sales and inappropriate use of antibiotics are major problems in Ethiopia and have contributed to the emergence of drug resistance in bacterial pathogens (Oumer, 2019). Several studies conducted across Ethiopia have also indicated increasing rates of drug resistance in *Salmonella* species *S. aureus* and several other pathogenic bacteria to commonly prescribed antibiotics (Reta *et al*., 2015; Setegn *et al*., 2016; Bayeh *et al*., 2019; Tadesse *et al*., 2019; Rajiha *et al*., 2019).

Some institution-based studies (mostly university cafeterias) in Ethiopia have reported the presence of *S. aureus* and *S.* Typhi circulating among food handlers (Dagnew *et al*., 2012; Diriba *et al*., 2020). However, there is little published data on the epidemiology of *S.* Typhi and *S. aureus* carrier status among food handlers in public food-catering establishments. The occurrence of foodborne disease outbreaks in public food catering establishments is likely to have far-reaching ripple effects that may have economic repercussions in countries with budding Hotels and Tourism Industry. In the absence of surveillance data and food safety risk management, periodic cross-sectional studies may serve as an early warning system to prevent potential foodborne disease outbreaks by prophylactic treatment of asymptomatic carriers. Therefore, this study aimed to assess the incidence and antibiogram of asymptomatic *S. aureus* and *S.* Typhi carrier status among food handlers working in restaurants in Hawassa City, southern Ethiopia.

Materials and Methods

Description of Study Areas

The study was conducted in Hawassa City, at the Hawassa University Comprehensive Specialized Hospital (HUCSH) Clinical Microbiology Laboratory. Hawassa is located in the Great East African Rift Valley on the shores of Lake Hawassa, 275 km south of Addis Ababa. Hawassa is the administrative center of Sidama Regional State of Ethiopia and situated within the geographic coordinates 7^{o} 3’N - 7.050^{o} N and 38^{o} 28’ E - 38.467^{o} E, at an altitude of about 1708 meters above sea level. According to the Ethiopian Central Statistical Agency (CSA, 2017), the estimated population of Hawassa in 2017 was 351,567, and considering the annual growth rate of 2.6% for the country, the current population of

Hawassa can be estimated to be 408,225. The city is a tourist hotspot for both locals and foreigners. Therefore, it is home to scores of international hotels, resorts, and large restaurants that serve both local and international visitors.

Study Population

Food handlers working in different mass-catering establishments/restaurants in Hawassa were enrolled in this study. The study participants were selected from different restaurants.

Study Design

A cross-sectional study, based on microbiological laboratory analysis of nasal swabs and stool specimens, was conducted among volunteer food handlers working in purposively selected restaurants in Hawassa City from August to November 2020. A semi-structured questionnaire was also used to collect the socio-demographic backgrounds of the food handlers to assess possible predisposing factors.

The restaurants for this study were selected purposively, based on a series of circumstantial observations relating to customer service, location, overall cleanliness, and hygienic reputation. We aimed to include a diverse range of establishments to ensure a comprehensive assessment of *Staphylococcus aureus* and *Salmonella Typhi* carriage among food handlers. This targeted approach allowed us to gather data from a representative sample of food handling environments, enhancing the reliability and relevance of our findings.

During the the study, we conducted visits to various restaurants to engage with managers and food handlers regarding our research objectives. The aim of the study was explained clearly, emphasizing the importance of assessing the carriage status of *Staphylococcus aureus* among food handlers and *Salmonella* Typhi among individuals at potential risk. Food handlers who were willing to participate voluntarily in the study were informed about the collection processes, the significance of their participation, and the potential impact on public health. Informed consent was obtained from all participating food handlers, ensuring they understood their rights and the study's implications.

Inclusion Criteria

All food handlers who worked in the selected mass-catering restaurants in Hawassa City, who voluntarily participated after being informed of the objectives of the study, were included in the study until the required sample size was obtained.

Exclusion Criteria

Food handlers who had taken antibiotics within three weeks before the study and those who were not willing to participate were excluded from the study.

Study Variables

Dependent variable: The incidence and antibiogram of *S. Typhi* and *S. aureus* carrier state among food handlers

Independent variable: Socio-demographic data (Age, Sex, educational status, and Service year)

Sample Size and Sampling Procedures

We followed a non-probabilistic convenient sampling method to determine the sample size, considering limited resources and time. A total number of 20 restaurants were purposively selected, consisting of 10 large international hotels and 10 medium-sized restaurants. Five volunteer study participants (food handlers) were included based on their willingness from each, resulting in a total sample size of 100 participants.

Samples Collection and Preparation

Nasal swabs and stool samples were collected from all 100 volunteer food handlers to determine the incidence of *S. aureus* and *S.* Typhi, respectively. For all the study participants, nasal swabs were obtained from both anterior nares (CLSI, 2020). We conducted nasal swab collections from food handlers using a sterile technique to minimize contamination. Each participant was instructed to tilt their head back slightly. The swabs were initially moistened with sterile distilled water, gently inserted into each nostril, rotated for a few seconds, and then aseptically returned to a labeled (with code), screw-cupped test tube. Following collection, all used materials, gloves, and masks were disposed of in biohazard bags, and hand hygiene was maintained throughout the process. Then specimens were immediately transported, using an ice box, to the clinical microbiology laboratory of Hawassa University Comprehensive Specialized Hospital (HU CSH) for bacteriological analysis. When the specimen could not be processed within 4 h, it was placed in a Stuart transport medium and stored in a refrigerator (CLSI, 2020).

Similarly, stool specimens were collected from each food handler with a suitable, labeled plastic container with a leak-proof lid and a clean wooden applicator stick. The participant was instructed to collect the stool specimen on a piece of toilet tissue and to transfer approximately 1 g of it to the container using two sticks. The lid was sealed once the specimen was placed in the specimen container. When quick delivery (within two hours of collection) of the specimen to the laboratory was not possible, a small amount of the stool specimen was collected on two or three swabs and placed in a tube containing Stuart's transport medium (CLSI, 2020). The stool samples were transported in an ice box to the Hawassa University Comprehensive Specialized Hospital (HUCSH) Clinical Microbiology Laboratory, for bacteriological analysis.

Bacteriological Analysis of Samples for Detection Of Target Organisms

The isolation and identification of *S. aureus* from the nasal swab samples was performed based on conventional methods and according to the standard operational procedure of the HUCSH laboratory (Anyanwu and John, 2013). Upon arrival at the microbiology laboratory, all nasal swab samples were directly inoculated onto Mannitol Salt Agar (Oxoid, UK), and the inoculum was streaked with wire loop. Mannitol Salt Agar (MSA) is a selective and differential medium designed to isolate staphylococci and distinguish *Staphylococcus aureus* based on its ability to ferment mannitol, producing yellow colonies with surrounding yellow zones. After incubation of the MSA plates at 37°C for 48 hours, presumptive colonies were examined by Gram staining, revealing Gram-positive cocci arranged in grape-like clusters. These isolates were further screened using the catalase test, in which a drop of 3% hydrogen peroxide was added to a colony; the immediate release of oxygen bubbles confirmed catalase positivity, consistent with staphylococcal species. Definitive identification of *S. aureus* was achieved using the slide coagulase test, wherein bacterial suspensions were mixed with plasma on a glass slide; visible clumping within 30 seconds indicated the presence of bound coagulase, a hallmark of *S. aureus*. All procedures were performed under aseptic conditions and following standard microbiological protocols (CLSI, 2020).

Likewise, conventional phenotypic methods were followed for the isolation and identification of *Salmonella Typhi* from the stool samples (Srijan *et al*., 2015). Accordingly, a portion of each stool sample was first inoculated into Selenite F broth, a selective enrichment medium that suppresses competing intestinal flora while promoting the growth of *Salmonella* spp., and incubated aerobically at 37°C for 24 hours. Following enrichment, aliquots were streaked onto Hektoen Enteric Agar and MacConkey Agar, both of which serve as differential and selective media for enteric pathogens. Hektoen Enteric Agar facilitates the identification of *Salmonella* through its characteristic production of hydrogen sulfide (black-centered colonies) and non-lactose fermentation, while MacConkey Agar aids in differentiating non-lactose fermenters (colorless colonies) from coliforms (pink colonies). After incubation at 37°C for 24 hours, suspected colonies were subjected to further biochemical screening and confirmed using serological agglutination tests with *Salmonella Typhi*-specific O antiserum, factor

9/group D (Andrews, 2021). This classical workflow remains a reliable approach for the detection of S. Typhi in stool specimens, particularly in resource-limited settings.

Antibiotic Sensitivity Testing

Antimicrobial susceptibility tests were performed using the Kirby-Bauer disk diffusion technique, following standard protocol (CLSI, 2020). The inoculum was prepared by selecting well-isolated colonies of the test organisms with a sterile wire loop and suspending them in sterile normal saline. The density of the suspension was adjusted to 0.5 McFarland turbidity standard using a barium sulfate solution. The test organisms were uniformly seeded on the Mueller-Hinton agar (Oxoid UK) surface and exposed to a concentration gradient of antibiotics diffusing from antibiotic-impregnated paper disks into the agar medium, and then incubated at 37°C for 24-48 h. The antimicrobial disks tested included Penicillin-G (10 IU), Oxacillin (1µg) Tetracycline (30µg), Erythromycin (15µg), Clindamycin (2µg), Cotrimoxazole (25µg) and Vancomycin. The diameters of the zone of inhibition around the discs were measured to the nearest millimeter with a ruler and classified as sensitive, intermediate, or resistant based on a standardized table (Callan and Westblade, 2020). A resistant (ATCC 25923), and susceptible (ATCC 29213) *S. aureus* strains were used in a parallel run as quality controls during identification by biochemical tests (Callan and Westblade, 2020).

Socio-demographic Data

A semi-structured questionnaire was used for collecting information on the demographic characteristics of the food handlers (Age, Sex, educational status, and Service year)

Data Quality Assurance and Analysis

Data quality was maintained throughout the study by adhering to the standard operating procedures of the HUCSH Clinical Laboratory. Culture Media were prepared according to the manufacturer's instructions, and sterility was checked by incubating the representative batch at 37°C overnight and observing growth. A uniform code was used for each sample for both the specimens and the questionnaire.

The data from the laboratory results and questionnaire were recorded in Microsoft Excel 2010 and then exported and analyzed using SPSS version 23. Descriptive statistics (percentages or frequency) were calculated to summarize the information collected during this study in frequency tables and percentages. The incidence of *S. aureus* and *S.* Typhi was calculated as the number of positive isolates divided by the total number of samples examined. The chi-square test ($\chi 2$) was used to compare the incidence among each socio-demographic category. Statistical significance (p) of less than 0.05 was considered. Bonferroni's post hoc test for chi-square was used to make multiple comparisons of the proportions (incidence among the various socio-demographic groups).

Ethical Issues

An ethical clearance letter was received from the Institutional Review Board (IRB) of Hawassa University, College of Medicine and Health Sciences. The participants (food handlers) were given complete information about the objectives of the study. Only volunteer participants were requested to sign an informed consent form prepared in English and Amharic. Signed informed consent was obtained from all the participants before sampling. Participants' confidentiality was maintained throughout the study.

Results

The Occurrence of *S. aureus* Nasal Carriage among Food Handlers

Of the 100 participants, 54 belonged to the 29-39 age group, whereas 31 and 15 belonged to the 18-28 and > 40 age groups, respectively (Table 1). Females accounted for 58% and males for 42%. Based on educational level, the study participants were classified into four categories: illiterate (17%), primary education (49%), secondary education (27%), and tertiary education (7%). Based on service years, 18% had 1-5 service years, 59% had 6-10 years, and 15% had >11 service years (Table 1).

Of the 100 nose swab samples, *S. aureus* was isolated from 10 samples, resulting in an overall occurrence of 10%. Of these 10 carriers, six were females and four were males. The sex-specific incidence of *S. aureus* carrier status was 9.5% (6/58) among female food handlers and 10.3% (4/42) among male (Table 1). Regarding age, among the 10 carriers of *S. aureus,* six were found in the 29-40 age group, while 3 and 1 were in the 18-28 and >40 age groups, respectively. Therefore, the age-specific incidence of the *S. aureus* carrier state was 11.1% (6/54) among the 29-39 age group, 9.7% (3/31) among the 18-28 age group, and 6.7% (1/15) among those aged >40 years (Table 1). The observed differences in the incidence rate with sex and age of the food handlers were not statistically significant (p.>0.05).

Regarding the educational status of the carriers, five were illiterate, three were at the primary level, one was secondary and one was at the tertiary educational level. Therefore, the educational level-specific incidence rate of *S. aureus* among food handlers was 5/16 among illiterates, 3/49 among those at the primary level, and 1/28 and 1/7 among those with secondary and tertiary educational levels, respectively (Table 1). The observed difference in the incidence of *S. aureus* nasal carrier state among food handlers with different educational statuses was statistically significant (p< 0.05). However, the observed differences in the nasal carriage rate of *S. aureus* among the literates with different educational levels were not statistically (p>0.05) significant (Table 2).

Based on experience, five of the 10 *S. aureus* nasal carriers had 6-10 years of service, three had 1-5 years and two of them had >11 years of service. The work experience-specific incidence of *S. aureus* was 8.5% (5/59) among food handlers who had 1-5 service years, 3/26 among those with 6-10 service years, and 2/15 among those with >11 service years (Table 1). However, the observed differences in the incidence of *S. aureus* nasal carrier state among food handlers with different service years were not statistically (p>0.05) significant (Table 1).

Antimicrobial Sensitivity Pattern of *S. aureus*

The antibiotic susceptibility profiles of the *S. aureus* isolates from the nasal swabs of the food handlers are shown in Table 3. Five of the ten isolates were resistant to one or more of the seven antibiotics tested. Of the isolates that showed resistance, one showed single resistance to penicillin, while four showed resistance to two or more drugs. Of the multiple antibiotic-resistant isolates isolate, HUB2 was found to be MRSA and showed resistance to penicillin, tetracycline, erythromycin, and oxacillin. Two isolates (isolates HUB6 and HUB8) showed double resistance, and one isolate (isolate HUB4) showed triple resistance (Table 3). Resistance to penicillin was the most frequent drug resistance exhibited by five of the ten (50%) *S. aureus* isolates, while all the isolates were sensitive to vancomycin. Resistance to clindamycin, erythromycin, and oxacillin was exhibited by single isolates (1 in 10) whereas resistance to tetracycline and cotrimoxazole was exhibited by two isolates in each case (Table 3).

Table 1. Incidence of *S. aureus* nasal carriage among food handlers of Hawassa City, Ethiopia, from August to November 2020, n=100

Characteristic		Frequency n (%)	Incidence of *S. aureus* (%)	Model test x^2 and p –value
Gender	Male	42 (42)	4 (9.5)	x^2= 0.018
	Female	58 (58)	6 (10.3)	p = 0.893
Total		100 (100)	10 (10)	
Age (years)	18-28	31 (31)	3 (9.7)	x^2= 0.693
	29-40	54 (54)	6 (11.1)	p = 0.707
	>40	15 (15)	1 (6.7)	
Total		100 (100)	10 (10)	
Education level	Illiterate	16 (16)	5 (31.3)	x^2= 10.275
	Primary education	49 (49)	3 (6.1)	p = 0.016
	Secondary education	28 (28)	1 (3.6)	
	Tertiary education	7 (7)	1 (14.3)	
Total		100 (100)	10 (10)	
Service year	5-Jan	26 (26)	3 (11.5)	x^2= 0.567
	10-Jun	59 (59)	5 (8.5)	p= 0.753
	> 11	15 (15)	2 (13.3)	
Total		100 (100)	10 (10)	

Table 1. Post hoc test for multiple comparisons of incidence of *S. aureus* among food handlers of different educational level (Bonferroni's method)

Socio-demographic group		*S. aureus* incidence				
(Educational level)	n	Negative	%	Positive	%	P value
Illiterate	16	11	68.80%	5	31.30%	0.002
Primary	49	46	93.90%	3	6.10%	0.204
Secondary	28	27	96.40%	1	3.60%	0.181
Tertiary	7	6	85.70%	1	14.30%	0.697
Total	100	90	90	10	10	

COR = crude odd ratio; CI = Confidence interval, n = number of study subjects

Table 3. Antimicrobial sensitivity pattern of *S. aureus* isolates from nasal swab samples of food handlers working in some restaurants in Hawassa city, southern Ethiopia

Isolate	Pen	Clind	Tet	Cotr	Ery	Oxa	Vanco	Number of R
HUB1	S	S	S	S	S	S	S	0
HUB2	R	S	R	S	R	R	S	4
HUB3	S	S	S	S	S	S	S	0
HUB4	R	S	R	R	S	S	S	3
HUB5	S	S	S	S	S	S	S	0
HUB6	R	R	S	S	S	S	S	2
HUB7	S	S	S	S	S	S	S	0
HUB8	R	S	S	R	S	S	S	2
HUB9	S	S	S	S	S	S	S	0
HUB10	R	S	S	S	S	S	S	1
Number of R	5	1	2	2	1	1	0	5

R: resistant; S: Sensitive; Pen: penicillin; Clind: Clindamycine; Cotr: Cotrimoxazole; Ery: Erythromycine; Oxa:Oxacilline;Vanco:Vancomycine

Discussion

The Occurrence of *Staphylococcus Aureus* and *Salmonella* Typhi Carrier State

The occurrence rate of the *S. aureus* nasal carrier state in this study (10%) was lower than the average nasal colonization of healthy adults reported from other countries previously (Mehraj et al., 2016; Hu *et al*., 2024). The nasal carriage rate of *S. aureus* among food handlers in the present study was also lower than the 20.5% rate reported for food handlers working in the Gondor University cafeteria, Gondor, northern Ethiopia (Dagnew *et al*., 2012). Other studies also reported higher rates than the present study, including 53% in Kuwait City (Udo *et al*., 2009) and 38.8% in Ekpoma Edo, Nigeria (Eke *et al*., 2015). On the other hand, a slightly lower carriage rate than the present study was reported in a study conducted in Jimma Town, southwest Ethiopia, where the rate of *S. aureus* nasal carriage was 9% (Beyene *et al*., 2019). Another similar study among food handlers in Canton City, Bosnia, and Herzegovina reported a slightly lower rate (7.1%) than the present results (Segalo *et al*., 2020). In contrast, the nasal carriage rate of *S. aureus* among food handlers in this study was significantly higher than the 0.77% reported in a similar study conducted in Turkey (Gunduz *et al*., 2008).

Several factors may explain the differences in the reported incidence rates of nasal carriage among food handlers reported in different studies. These differences might be due to variations in the study design such as sample size, study period, season, and methods of study (Mehraj *et al*., 2016). Furthermore, the results may show variation in different populations living in different geographical areas and their demographics (Sivaraman *et al*., 2009). Likewise,, occupational exposure to raw meat can make food handlers more susceptible to contamination and raise their risk of colonization (Ho et al., 2014). Certain host factors, which can be broadly divided into host genetic and immune factors, are more likely to be associated with nasal carriage; however, robust data on this association are scarce (Mehraj *et al*., 2016; Sivaraman *et al*., 2009).

A statistically significant difference in the incidence of *S. aureus* nasal carriage was observed between illiterate and literate food handlers. However, no significant differences were found in the incidence of nasal carriage among food handlers based on sex, age, or service year. This is contradictory to the study reported in Jimma Town by Beyne *et al.*

(2019), in the Gondar University by Dagnew *et al.* (2012), and in Amassoma City, Nigeria, by Onanuga and Temedie, (2011), who reported no statistically significant difference in the incidence of *S. aureus* nasal carriage among literate and illiterate food handlers.

On the other hand, the findings of this study were concordant with a study conducted among food handlers in the Wollo University student cafeteria by Gashaw (2019), where the observed differences in *S. aureus* incidence rates among educated and uneducated individuals were statistically significant ($P < 0.05$). Likewise, it was also supported by Adugna *et al.* (2018)], who suggested that the lower educational level of food handlers could contribute to higher *S. aureus* contamination. This could be explained by the fact that literate food handlers have better adherence to personal hygiene practices than do illiterate food handlers. The education of food handlers may be crucial in preventing bacterial carriage (Segalo *et al*., 2020). The acquisition of nasal colonization through contaminated hands may have a more significant effect on transient nasal carriers (Saadatian-Elahi *et al*., 2018). The observed discrepancy in these results could be due to differences in study design, population, and demographics.

Regarding *Salmonella* Typhi, none of the food handlers were found to be carriers of the pathogen based on stool culture. This finding was in line with several institutional studies conducted in different parts of Ethiopia. The reported incidence of *S*. Typhi was zero in a study conducted among food handlers at the University of Gondar (Andargie *et al*., 2008) and Hawassa University students' cafeterias by Desta *et al*. (2014). In contrast, the reported incidence of *S*. Typhi is 0.9% among food handlers in Dilla, southern Ethiopia (Misganaw and Williams, 2013), 1.3% among those working in Bahar Dar, northern Ethiopia (Bayeh *et al*., 2010), and 2.7% among food handlers in Ghana (Feglo and Sakyi, 2012). Variations in the incidence of *S*. Typhi among food handlers across different studies may be attributed to differences in personal hygiene, sociodemographic determinants, geographical regions (WHO, 2015), microbiological methods used to detect Salmonella, and study designs (Tadesse *et al*., 2019). While epidemiological risk factors for becoming a persistent carrier have not been throroughly studied, potential risk factors may be influenced by the proportion of the population with gallbladder abnormalities (Gal-Mor, 2019).

Antimicrobial Sensitivity Pattern of *S. aureus*

Considering the drug sensitivity patterns of *S. aureus* isolates, 5 of the 10 (50%) isolates were resistant to penicillin. This was concordant with a study conducted in Gondar, Northwestern Ethiopia, where 51.2% of nasal swab isolates were reported to be resistant to penicillin (Dagnew *et al*., 2012). Similarly, another study in Portugal revealed that 48% of nasal isolates of *S. aureus* were resistant to penicillin (Castro *et al*., 2016). However, the penicillin resistance rate among the *S. aureus* nasal isolates in the this study was significantly lower than the reported penicillin G resistance rate of 97.2 % from Jigjiga University (Wolde *et al*., 2015) and the 90.7% from Jimma town (Beyene *et al*., 2019).

In this study, one in 10 (10%) of the *S. aureus* isolates was resistant to oxacillin (Methicillin-resistant – MRSA) which is in line with studies from other parts of Ethiopia, including the reported oxacillin resistance rate of 9.8 % from Gondar University (Dagnew et al., 2012), the 7% from Jimma town (Beyene *et al*., 2019) and the 13.8% from Debre Markos (Reta *et al*., 2015). However, the incidence of MRSA isolates in this study was higher than that in a study from Turkey, where none of the isolates were found to be MRSA (Vatansever *et al*., 2016). These contradictory results could be due to differences in the methods, sample sizes, populations, and geographical locations in which the study was conducted.

The frequency of *S. aureus* isolates resistant to erythromycin and clindamycin was one in 10 (10%) cases and two in 10 (20%) cases for tetracycline and cotrimoxazole. This resistance frequency was lower than that reported by Dagnew *et al*. (2012) at Gondar University, where the reported frequencies of isolates resistant to tetracycline, cotrimoxazole, and erythromycin were 31.7%, 26.8%, and 14.6% respectively. Likewise, another study conducted in Jimma Town, southwestern Ethiopia, found that 14.6% of *S. aureus* isolates exhibited resistance to erythromycin (Beyene *et al*., 2019). A similar study conducted in Turkey reported that 9% of *S. aureus* isolates were resistant to tetracycline, 8% to erythromycin, and 0% to clindamycin (Vatansever et al., 2016). A much higher frequency of *S. aureus* isolates was reported from food handlers in Portugal with a resistance rate of 96% to tetracycline and 68% to erythromycin (Castro *et al*., 2016). The variation in the results could be mainly due to sample size and geographical area.

In contrast, all isolates (100%) were sensitive to vancomycin. This was in line with a study, in Gondar, where all isolates of *S. aureus* (100%) were reported to be sensitive to vancomycin (Dagnew *et al*., 2012). A similar finding has been reported in Turkey (Vatansever *et al*., 2016). In contrast, a study conducted in Jimma reported that 93% of *S. aureus* nasal swab isolates from food handlers were sensitive to vancomycin (Beyene *et al*., 2019). These variations could be due to differences in study populations and methods.

A multi-drug resistance (MDR) pattern, (resistance to at least one antimicrobial drug in three or more different categories of antimicrobial drugs) was observed in two of the 10 (20%) *S. aureus* isolates, a frequency that is slightly higher than that reported in a study from Jimma town (Beyene *et al*., 2019) where 14.3% MDR was reported, but lower than the 86.4% reported in Bosnia and Herzegovina (Segalo *et al*., 2020). The emergence of MDR *S. aureus* is attributed to the inappropriate use of broad-spectrum antibacterial drugs over-the-counter sell without the prescription of qualified health workers. This is common in most developing countries where antibiotic use is generally unregulated. Therefore, drug administration authorities and other public health officials should be vigilant to enforce existing regulations and control the prudent use of antibiotics by the public.

Conclusion

The study revealed that 10% of the food handlers were nasal carriers of *S. aureus*, while no *Salmonella Typhi* was detected in any stool samples. Notably, illiterate food handlers were significantly more likely to be nasal carriers of *S. aureus* compared to their literate counterparts. However, no statistically significant associations were found between nasal carriage and factors such as sex, age, or years of service. Among the *S. aureus* isolates, five of 10 exhibited resistance to one or more of the seven antibiotics tested, with one isolate showing single resistance to penicillin and four others demonstrating resistance to multiple drugs. Specifically, two isolates displayed double resistance, and one isolate exhibited triple resistance, including identification as MRSA, which was resistant to penicillin, tetracycline, erythromycin, and oxacillin. Importantly, all *S. aureus* isolates were found to be sensitive to vancomycin. The finding of multiple antibiotic-resistant *S. aureus* isolates from the nasal swabs of food handlers implies the possibility of antibiotic misuse or overuse and the potential risks of foodborne disease outbreaks. This calls for raising the awareness of food handlers about the need for hygienic food handling with an emphasis on illiterate ones, judicious use of antimicrobials, and curbing over-the-counter sell and abuse of drugs without the prescription of qualified physicians.

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Ethics Approval and Consent to Participate

Ethical approval with regards to all experimental protocols for the work was granted by Institutional Review Board (IRB) of Hawassa University, College of Medicine and Health Sciences. All the study participants were informed about the objectives of the study and were included in the study only after they gave their verbal consent to participate.

Competing Interests

The authors have none to declare

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