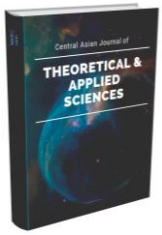

CENTRAL ASIAN JOURNAL OF THEORETICAL AND APPLIED SCIENCE

https://cajotas.centralasianstudies.org/index.php/CAJOTAS

*Volume: 06 Issue: 04 | October 2025 ISSN: 2660-5317*

*Article*

# Determining the Prevalence of Staphylococcus aureus Bacteria in Nasal Swabs from Restaurant Workers in Al- Kut City

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**Abstract:** Antibiotic resistance represents a global challenge to modern medicine, and among the most critical resistant pathogens is Staphylococcus aureus, particularly methicillin-resistant strains (MRSA). This study aimed to determine the prevalence of Staphylococcus aureus and the patterns of antibiotic resistance among nasal swab samples collected from restaurant workers in Al-Kut City. A total of 110 nasal swabs were obtained from food handlers and analyzed using standard microbiological techniques, including selective culturing on mannitol salt agar, coagulase testing, and Vitek2 identification. The D-test was used to assess inducible clindamycin resistance in MRSA isolates. Of the samples analyzed, 23 tested positive for Staphylococcus aureus, with 16 identified as MRSA and 9 as methicillin-sensitive S. aureus (MSSA). Antibiotic susceptibility tests revealed varying resistance rates among isolates, including 60.8% resistance to cefoxitin, 73.9% to oxacillin, 43.4% to vancomycin, 65.2% to erythromycin, 43.4% to clindamycin, 47.8% to ciprofloxacin, 17.4% to gentamicin, and 4.4% to chloramphenicol. These findings suggest a high prevalence of S. aureus carriage and antibiotic resistance among food handlers, underscoring the necessity of implementing regular screening and treatment programs in the food service industry to mitigate the risk of staphylococcal foodborne infections and protect public health.

**Keywords:** *Staphylococcus aureus*, Clindamycin Inducible Resistance (D-test), MRSA, MSSA

**Citation:** Aubed, M. H., Almeahy, A. W., Jabbar, M. R., Shahib, N. A., Makotuf, H. H., & Hamid, B. Determining the prevalence of Staphylococcus aureus bacteria in nasal swabs from restaurant workers in Al-Kut City. Central Asian Journal of Theoretical and Applied Science 2025, 6(4), 626-632.

Received: 30th Jun 2025
Revised: 07th Jul 2025
Accepted: 21st Jul 2025
Published: 03rd Aug 2025



## 1. Introduction

*Staphylococcus aureus* is usually a commensal organism, which has a residence on human mucosal or skin surfaces [1], [2], [3]. Nevertheless, it also serves as a notable one of the pathogens that causes the varied series of infections in the community and in hospitals, such as, recurrent tonsillitis, pneumonia, soft tissue, urinary tract infections, and bloodstream infections [4], [5], [6], [7]. Even though this bacterium infects various anatomical points, it is assumed that this bacterium has its main ecological niche in the form of the anterior nares. An estimate of 2030 percent of persons become persistent and become a carrier successfully in the nose and 30 percent are transient carriers [8]

*S. aureus* has over time become resistant to a number of classes of antibiotics [9], [10], [11]. The MRSA strains have become widespread worldwide and have been found to be resistant to practically all β-lactam antibiotics, such as penicillins, cephamycins and cephalosporins, the exception being ceftaroline, a fifth-generation cephalosporin which has FDA approval in the treatment of MRSA infections [12], [13], [14]. Moreover, Staphylococcus species are often developed to become resistant to the available classes of antibiotics like quinolones, macrolides, and aminoglycosides [15], [16], [17].

Resistance of *Staphylococcus aureus* to the majority of the antibiotics commonly used in the treatment of infection becomes an issue of concern. This has changed the antimicrobial susceptibility trend and thus prompted the thought of using clindamycin as a treatment agent [18]. However, it was only reported in the early 1960s that the methicillin-resistant Staphylococcus aureus (MRSA) - otherwise known as multidrug-resistant S. aureus - was the causative agent of hospital-acquired infections that were typically proved to be fatal in the vulnerable patients owing to their complications. *S. aureus* is Gram-positive, enzymatically catalase-positive, facultative anaerobic, nonmotile, and nonspore-forming bacterium [19], [20]. Although it naturally occurs in the human body, especially on the skin and through the gastrointestinal tract, it is a dangerous threat in the form of an opportunistic pathogen. Severe infections can be caused by any interruption in normal skin barrier, immune deficiency or gastrointestinal disorder.

*Staphylococcus aureus* continues to cause a significant proportion of diseases covering both health care and the community including skin and soft tissue infections, endocarditis, bacteremia, pneumonia, scalded skin syndrome, toxic shock syndrome and osteomyelitis [21], [22]. Many of its pathogenicity can be ascribed to a range of virulent factors which allow adherence to the host tissues, avoiding innate immunity, tissue destruction and the destruction of connective tissue. Some of them include the enzymes that enhance tissue invasion and dissemination, including lipases and hyaluronidase [23].

MRSA is also regarded as a significant global public health hazard because of its success and tenacity [24], [25]. Another group of antibiotics, which have been considered as a potential replacement of *S. aureus* treatments, is lincosamide antibiotics (clindamycin) whose main advantages are both oral and injectable routes, wide distribution across tissues, and the effectiveness against the wide range of *S. aureus* strains [26], [27]. Nonetheless, the widespread of over-usage of clindamycin and other antibiotics belonging to macrolide-lincosamide-streptogramin B (MLSB) group contributed with a significant increase of the rates of *Staphylococcus* strains demonstrating resistance to these substances

## 2. Materials and Methods

### Sample Collection

A hundred swab samples were taken on both genders of restaurant employees. Sterile nasal swabs were taken and the swabs were run immediately to the lab where they underwent microbiological analysis and were placed on the right culture media.

### Laboratory Diagnosis

The samples obtained were promptly inoculated, with blood agar and mannitol salt agar being inoculated upon reaching the laboratory one that is selective to Staphylococcus aureus. Occasionally cotton tipped swabs were used to roll over a localized area on the agar surface before streaking of a sample on the agar using a sterile inoculating loop as per quadrant streak method. The plates inoculated were then placed under 37°C at room temperature and left to develop in 24 h. The isolation and preliminary processing of bacterial identification was carried out on standard microbiological techniques as noted by references (17-19). Confirmation of S. aureus was done via traditional diagnostic procedures, which comprised analysis of colony shape and staining, hemolysis on blood-agar, catalase test, coagulase test (slides and tubes), and several biochemical reactions.

### Antimicrobial Sensitivity

protocol (Kirby-Bauer technique), following the provisions of the Clinical and Laboratory Standard Institute (CLSI) and the European Committee on Antimicrobial Susceptibility Testing (EUCAST) (21). Bacterial inocula were done by growing the isolates in nutrient broth culture and culturing them with 2 to 5 hours incubation until turbidity was observed. Sterile physiological saline was then added to the bacterial suspension to obtain a turbidity level of McFarland 0.5, equivalent to about 1.5 10 8CFU/mL.

In D-test, erythromycin and clindamycin antibiotic disks were brought to Mueller-Hinton agar in order to identify inducible clindamycin resistance. The presence of a flattened, D-shaped zone of inhibition alongside the clindamycin disk with a measurement of clindamycin inhibition zone being 21mm indicated a positive D-test result in erythromycin-resistant isolates. Transmitted light was used in measuring the diameters of inhibition zones of both oxacillin and vancomycin to be able to accurately assess them.

## 3. Results

The nasal carriage rate of Staphylococcus aureus timetable of food handlers of the 110 nasal swab specimens taken of the food handlers working in restaurants, 23 isolates of staphylococcus aureus were found giving an overall carriage level of 20.9 percent. Out of these, 14 isolates (53.33 percent) turned out to be methicillin-resistant S. aureus (mrsa), 9 isolates (39.22 percent) were methicillin-sensitive s. Aureus (mssa) as summarized in table 1.

**Table 1.** Prevelence of *Staphylococcus aureus* (MRSA & MSSA) in nasal carriage.

| Nasal carriage | isolates | No. (%) |
|---|---|---|
| Positive | *S.aureus* | 23 (20.9%) |
| | MRSA | 14(60.8%) |
| | MSSA | 9(39.22%) |
| Negative | - | 87(79.1%) |
| Total Sample | - | 110(100%) |

MRSA : Methicillin Resistance *S. aureus.*

MSSA: : Methicillin Sensitive *S. aureus.*

Our study showed that the number of male food handlers carriers were more than females number 78.25% and 21.73.66% in Table 2.

**Table 2.** Gender distribution of isolates and ratio of MRSA.

| SEX | MRSA | MSSA | Total |
|---|---|---|---|
| Mal | 12 (52.17%) | 6 (26.08%) | 18 (78.25%) |
| Female | 2 (8.69%) | 3 (13.04%) | 5 ( 21.73) |
| Total | 14 (60.86) | 9 (39.12) | 23 (100%) |

*Staphylococcal aureus* Nasal Carriage Distribution by Gender and Age Methicillin-sensitive S. aureus (MSSA) and methicillin-resistant *S. aureus* (MRSA) was more frequent in male food handlers than females. Most participants fell in the younger age category; this includes those aged between 2029 years and 3039 years, as Table 3 shows. In these age brackets, the rates of nasal carriage of MRSA and MSSA were comparatively high in males (21.73 and 8.69 percent respectively) than females (0 and 4.34 percent respectively). Conversely, its prevalence was very low in the older age group (5059 years) in which a small proportion of S. aureus colonies were isolated in samples of the male participants[28].

**Table 3.** Staphylococcus aureus (MRSA & MSSA) carriers and age group.

| Age Group in years | Male MRSA(%) | Male MSSA(%) | Female MRSA(%) | Female MSSA(%) | Total |
|---|---|---|---|---|---|
| 20-29 | 5 (21.73 %) | 1(4.34 %) | 0 | 1(4.34 %) | 7 (30.43%) |
| 30-39 | 5 (21.73 %) | 2(8.69 %) | 0 | 0 | 7 (30.43%) |

| | | | | | |
|---|---|---|---|---|---|
| 40-49 | 2 (8.69 %) | 1 (4.34 %) | 2(8.69 %) | 1 (4.34 %) | 6 (26.08%) |
| 50-59 | 0 | 2 (8.69 %) | 0 | 1(4.34 %) | 3 (13.04%) |
| Total | 12(52.17%) | 6(26.08%) | 2(8.69%) | 3(13.04%) | 23(100%) |

**Antibiotic Susceptibility Pattern of S. aureus**

Antimicrobial Resistance Pattern of Staphylococcus aureus strains Table 4 shows that resistance rates to the antimicrobial agents were variable among the S. aureus isolates (both in the MRSA and MSSA collection) against all of the measured antimicrobial agents. The greatest resistance was to ampicillin (95.6%), oxacillin (73.9%), erythromycin (65.2%), tetracycline and cefoxitin (both 60.8%). Ciprofloxacin, clindamycin and vancomycin had resistance rates of 47.8 (43.4 and 43.4), respectively [29]. Beating resistance rates were identified in the trimethoprim-sulfamethoxazole agent (39.2%), gentamicin (17.4%), and chloramphenicol (4.4%). It is important to mention that all isolates of S. aureus were sensitive to amoxicillin.

**Table 4.** Antimicrobial sensitivity pattern of Staphylococcus aureus isolates to various Antimicrobial agents.

| | ***Staphylococcus aureus* isolates** | |
|---|---|---|
| **Antimicrobial disc** | **Resistant (%)** | **Sensitive (%)** |
| Cefoxitin | 14(60.8%) | 9 (39.2%) |
| Oxacillin | 17( 73.9%) | 6 (26.1%) |
| Vancomycin | 10 (43.4%) | 13(56.6%) |
| Erythromycin | 15 (65.2%) | 8(34.8%) |
| Clindamycin | 10 (43.4%) | 13(56.6%) |
| Tetracycline | 14 (60.8%) | 9 (39.2%) |
| Gentamicin | 4 (17.4%) | 19 (82.6%) |
| Ciprofloxacin | 11(47.8%) | 12 (52.2%) |
| Trimethoprim sulfamethxazole | 9 (39.2%) | 14 (60.8%) |
| Rifampcin | 4 (17.4%) | 19 (82.6%) |
| Chloromphinicol | 1 (4.4%) | 22 (95.6%) |
| Ampicillin | 22 (95.6%) | 1(4.4%) |
| Amoxicillin | 0 | 23(100%) |

## 4. Discussion

Staphylococcus aureus is among the most common opportunistic pathogens which are found both in the community and in the health sector (2). The sample size of this study was composed of nasal swabs collected on 110 food handlers that are employed in seven different restaurants where S. aureus was isolated in 23 people, which translates to a carriage percentage of 20.9. This observation reflects the one reported in other parts of the country where nasal carriage in other regions was found to be 30 percent, 28 percent and 22.3 percent. Moreover, the findings can be observed in conjunction with the results of and according to which S. aureus was the most common pathogen isolated in food workers with a rate of 79%, 53.3 and 46 percent respectively [30], [31].

The sample of food handlers was mostly male (78.25 compared to 21.73 by females), which is also likely to be explained by the local cultural peculiarities and social constraints imposed on female activity as related to restaurant employment. Similar revelations have also been provided in a similar study. Furthermore, male gender may become another risk factor of community-acquired S. aureus infections, whereby a couple of retrospective case-control studies found it to be a possible risk factor. The difference in the results regarding the carriage of nasal isolates might depend on some factors such as the educational status,

hygienic behavior, job history, and policy of the institution about food security and health check [32], [33], [34].

Demographic factors in the carriage of S. aureus include sex and age [35]. The colonization seasons either with MRSA or MSSA seem to be more common in younger food-handling persons in the age categories of 2029 years. We found that MRSA was more common in male subjects 20-29 years old and 30-39 years old (21.73%), with a sharp decline in the increase of age (50-60 years). These findings concur with the findings in previous reviews, which show that the younger ones have a higher tendency of being occupied in the food sector, hence exposing them to more risk.

The general prevalence of MRSA in nasal S. aureus carrier in our study is 60.86% and can compared to the general prevalence estimated in other regions of 21.5 in Samarra, 27 in Duhok, 28.6 in Brazil, 83.3 in Thi-Qar and 92.5 in Ethiopia [36], [37], [38]. Such differences are called by variations in geographic, occupational and regulatory contexts, potential differences in hygiene standards and patterns of antibiotic use.

In Iranian food handlers, lower MRSA carriage rates have been reported, having a prevalence level of 14.3% (37), 5.3% (38), and 14.54% [39]. The prevalence of S. aureus and MRSA is affected by various factors among them is frequent use of antibiotics, poor hygienic conditions, and sharing of personal objects and utensils[40].

The outcomes of the present research indicated a significant antimicrobial resistance in the S. aureus isolates with a predominance to ampicillin (95.6 %), oxacillin (73.9 %), erythromycin (65.2 %), cefoxitin (60.8 %), and tetracycline (60.8 %). The rates of moderate resistance were recorded between 47.8 (ciprofloxacin) to 43.4 percent (vancomycin and clindamycin), trimethoprim-sulfamethoxazole (39.2 percent) and lower rates between 17.4 (gentamicin) to 4.4 percent (chloramphenicol). It is important to note that 100 percent of the isolates were sensitive to amoxicillin. The results are similar to the published results pertaining to different parts of the country, and this concurs with the findings of the studies [41], [42].

Vancomycin is still considered one of the last-line antibiotics against infections with S. aureus strain resistant to other antibiotics. New information, however, signifies that there is the emergence of resistant vancomycin S. aureus (VRSA) in adjacent countries [43, 44]. Causation of VRSA infections presents a major clinical challenge because the cross resistant nature of these strains to various classes of antibiotics. The current study also reported the prevalence of VRSA was 17.8 percent which was similar to the previous findings of other studies .

The increased rate of MRSA is probably related to the horizontal transmission of gene resistance especially plasmids that cause 8-lactamase enzymes. These enzymes deactivate the β-lactam antibiotics, including penicillins, which develops the extensive resistance to these antibiotics found in the MRSA strains [45].

## 5. Conclusion

This study revealed that the risk of developing and transmitting community-associated Staphylococcus aureus and multidrug-resistant and MRSA strains to the targeted population is a posible threat. It can be explained by the aspects like improper or overuse of antibiotics and poor hygiene standards. Food handlers may be a source of cross-over and a potential source of the spread of MRSA to the community at large due to their direct access to food. Hence, the risk can only be eliminated by ensuring that the food handlers receive formal training on food safety and personal hygiene. Also, additional research involving the use of molecular-based methods is advised to validate the presence of resistance genes and to enhance the knowledge of the genetic phenomena that support multidrug resistance in isolates of S. aureus.

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