

## Original Article



# Staphylococcus Aureus Isolated from Surgical Site Infections: Evaluation of Biofilm and Biofilm-Related Genes

Zaker Saeedi Nejad <sup>1</sup> , Farzad Mazloomirad <sup>2</sup>

<sup>1</sup> Department of Internal Medicine, School of Medicine, Shahid Beheshti Hospital, Yasuj University of Medical Sciences, Iran

<sup>2</sup> Student Research Committee, Yasuj University of Medical Sciences, Yasuj, Iran

✉ **Corresponding Author:** Tel: +989138740342; Email: kmazlumirad@gmail.com

**Received:** October 20, 2025

**Revised:** December 29, 2025

**Accepted:** February 16, 2026

**Citation:** Saeedi Nejad Z, Mazloomirad F.

Staphylococcus Aureus Isolated from Surgical Site Infections: Evaluation of Biofilm and Biofilm-Related Genes. J Surg Trauma. 2026;14(2):86-92.

DOI:10.61882/jsurgtrauma.14.2.86



## Abstract

**Introduction:** *Staphylococcus aureus* is the most common bacterium causing surgical site infections (SSIs). The biofilm formation of *S. aureus* leads to increased survival and bacterial persistence. This study aimed to evaluate the frequency of biofilm formation and the presence of *icaD*, *icaA*, *fnbA*, *clfA*, and *cna* genes in *S. aureus* from SSIs isolated from patients in two hospitals in southwest Iran.

**Methods:** This cross-sectional study included 77 *S. aureus* isolates from SSIs. The ability of *S. aureus* for biofilm formation was assessed by the Congo Red Agar method. The prevalence of *icaD*, *icaA*, *clfA*, *fnbA*, *cna*, and *mecA* genes was determined by PCR. Data were analyzed using SPSS software (version 20) through chi-square test and Fisher's exact tests.

**Results:** Biofilm formation was observed in 55 isolates (71.4%), including 30 (54.5%) strong and 25 (45.5%) weak biofilm producers' isolates. The ability to form biofilm was higher among MRSA isolates (83.7%) compared to MSSA isolates (50%). The frequency of the *icaD*, *fnbA*, *clfA*, *cna*, and *icaA* genes was 62.3%, 42.9%, 28.6%, 20.8%, and 19.5%, respectively. There was a significant association between the presence of *fnbA* ( $P=0.000$ ), *clfA* ( $P=0.000$ ), *icaD* ( $P=0.02$ ) genes and biofilm formation; however, it was not observed for *icaA* ( $P=0.054$ ), and *cna* ( $P=0.132$ ) genes.

**Conclusions:** Our findings reinforce the role of Biofilm-related genes (*fnbA*, *clfA*, *icaD*) and biofilm formation. Given the role of genes in biofilm formation, we can pursue the development of scientific solutions to control biofilms, which ultimately lead to improved public health.

**Key words:** Polymerase Chain Reaction, *Staphylococcus aureus*, Surgical Wound Infection

## Introduction

The Centers for Disease Control and Prevention defines surgical site infections (SSIs) as infections that occur at the incision site within 30 days of surgery. SSI is the third most common cause of nosocomial infections. Moreover, it is one of the most common problems for patients undergoing surgery. Surgical wound infection leads to an increase in the length of hospital stay, leading to an increased rate of mortality. SSIs are usually caused by exogenous and/or endogenous microorganisms that enter the wound during surgery (primary infection) or after surgery (secondary infection) (1). Among organisms, *S. aureus* is currently the most

common organism isolated from surgical wound infection, which causes 37% of SSIs in the community (2,3). *S. aureus* is a common human bacterium and an opportunistic pathogen that causes a wide range of nosocomial and community-acquired infections (4,5). These include some skin infections, postoperative infections, wound infections, soft tissue infections, pneumonia, toxic shock syndrome, sepsis, infective endocarditis, as well as bone and joint infections (6-8). Nosocomial pathogens can infect patients by forming biofilms on foreign implanted objects in the body (9). More than 65% of Nosocomial infections are caused by the infecting organisms that have the ability to produce biofilms (10). The most common microorganism



isolated from SSIs is *S. aureus* (11). Biofilm formation increases the severity of *S. aureus*-related infections and also leads to increased resistance to antimicrobial drugs (12). Biofilm formation by organisms is a potential factor of delay in the healing of surgical wounds. Various factors contribute to the formation of *S. aureus* biofilms. These include the *icaADBC* operon, which encodes enzymes required for polysaccharide intercellular adhesin (PIA) synthesis (13), and adhesion-related proteins known as *MSCRAMMs*, such as collagen-binding protein (*cna*), fibronectin-binding proteins (*fnbA* and *fnbB*), laminin-binding protein (*Eno*), fibrinogen-binding protein (*clfA*), biofilm-associated protein (*Bap*), and elastin-binding protein (*EbpS*), all of which facilitate bacterial adhesion (14–16). The primary objectives of this study were to assess biofilm formation in *S. aureus* isolates using the Congo Red Method and to determine the frequency of biofilm-associated genes, including *icaA*, *icaD*, *clfA*, *fnbA*, and *cna*, in strains isolated from surgical site infections in patients admitted to two hospitals in Yasuj, southwest Iran.

## Methods

This cross-sectional study was performed on 77 isolates of *S. aureus* isolated from patients with SSI who were hospitalized at Imam Sajjad and Shahid Beheshti Hospitals, Yasuj, Iran, between January and December 2019. The bacteria studied in this study are taken from a research project with a code of ethics (IR.YUMS.REC.1397.124), approved by the Student Research Committee of the University of Medical Sciences of Yasuj.

### Detection of Biofilm Formation by Congo Red Method

In order to prepare this medium, first, Brain Heart Infusion Broth medium was prepared along with agar, and Concord reagent was prepared separately in the form of an aqueous solution. Then the two solutions were autoclaved. After autoclaving, the sucrose was filtered from the medium and added to the medium. Biofilm formation by the Congo Red Method was performed according to Arciola et al.'s method (17). A positive result was indicated by black colonies. Weak biofilm producer isolates remained pink, and occasional darkening at the centers of colonies was also observed. Bright red colonies were considered negative.

### PCR assay

According to Cayci et al.'s method, DNA

extraction was performed by the boiling method (18). Detection of *methicillin-resistant S. aureus* (MRSA) was based on the *mecA* gene. PCR was performed for the detection of *icaD*, *icaA*, *fnbA*, *clfA*, and *cna* genes using primers displayed in Table 1. The PCR mixture was prepared separately for each gene in a total volume of 25 µL, including 12.5 µL of Master Mix (Amplicon, Denmark), 2 µL of each primer, 5 µL of bacterial DNA, and 3.5 µL of sterile distilled water. After amplification, 10 µL of the PCR products were electrophoresed (Major Science MP300, Taiwan) on 1.5% agarose gel (Pishgam, Iran) at 90 V for 45 minutes. The PCR products were stained with Gel Stain (Pishgam, Iran). They were then visualized by Gel Documentation (Major Science, Taiwan).

### Statistical analyses

The data were analyzed using SPSS software (version 20) through the Chi-square and Fisher's exact tests.  $P < 0.05$  were regarded as statistically significant.

## Results

Of the 77 *S. aureus* isolates, according to the *mecA* gene, 63.6% and 36.4% isolates were MRSA and MSSA, respectively. Biofilm formation was noted in 55 (71.4%) isolates, including 30 (54.5%) strong and 25 (45.5%) weak biofilm producers' isolates. In total, 65 (84.4%) *S. aureus* isolates were positive for at least one of the studied biofilm-related genes (*icaD*, *icaA*, *fnbA*, *clfA*, and *cna*). None of the genes involved in biofilm formation were observed in 12 isolates. Of the 22 isolates that were not able to form biofilms, 12 isolates were positive for at least one of the biofilm-related genes. Frequency of biofilm-related genes in biofilm-positive isolates and biofilm-negative isolates. In this study, the prevalence of *icaD*, *fnbA*, *clfA*, *cna*, and *icaA* genes was (48/77, 62.3%), (33/77, 42.9%), (22/77, 28.6%), (16/77, 20.8%), and (15/77, 19.5%), respectively. The significant association was observed in phenotypic production of biofilm, and the presence of *fnbA*, *clfA*, and *icaD* genes in *S. aureus* isolates ( $P < 0.05$ ); however, this connection was not observed for *icaA* and *can* ( $P > 0.05$ ) genes (Table 2).

The frequency of biofilm formation among MRSA isolates (83.7%) was higher than that among MSSA isolates (50%). The statistical difference between MRSA and MSSA regarding the frequency of the *clfA*, *fnbA*, *cna*, *icaA*, and *icaD* genes was not significant (Table 3).

**Table 1.** The primers used in this study

| Target Gene | Primer Sequence (5' → 3')                                            | Amplicon Length, bp | Reference | PCR program                                                                                                                                    |
|-------------|----------------------------------------------------------------------|---------------------|-----------|------------------------------------------------------------------------------------------------------------------------------------------------|
| <i>cna</i>  | F- AAAGCGTTGCCTAGTGGAGAC<br>R- AGTGCCTTCCCAAACCTTTT                  | 192                 | (19)      | 94°C – 5 Minutes<br>94°C - 30 Seconds<br>30 Seconds at 54°C<br>72°C - 1 Minutes<br>32X<br>72°C - 10 Minutes<br>annealing:<br>60°C - 30 Seconds |
| <i>clfA</i> | F- CCGGATCCGTAGCTGCAGATGCACC<br>R- GCTCTAGATCACTCATCAGGTTGTTCA<br>GG | 1000                |           |                                                                                                                                                |
| <i>fnbA</i> | F- GATACAAACCCAGGTGGTGG<br>R- TGTGCTTGACCATGCTCTTC                   | 191                 |           | annealing:<br>30 Seconds at 52°C                                                                                                               |
| <i>icaD</i> | F- AAACGTAAGAGAGGTGG<br>R- GGCAATATGATCAAGATAC                       | 381                 | (20)      | annealing:<br>30 Seconds at 49°C                                                                                                               |
| <i>icaA</i> | F- CCTAACTAACGAAAGGTAG<br>R- AAGATATAGCGATAAGTGC                     | 1351                |           | annealing:<br>30 Seconds at 49°C                                                                                                               |
| <i>mecA</i> | F- GTGAAGATATACCAAGTGATT<br>R- ATGCGCTATAGATTGAAAGGAT                | 147                 | (21)      | According to the<br>reference                                                                                                                  |

**Table 2.** The relationship between the strength of biofilm formation and the presence of biofilm-related genes

| Genes                   | <i>icaA</i>   |               | <i>icaD</i>   |               | <i>clfA</i> |              | <i>fnbA</i>   |               | <i>Can</i>    |               |
|-------------------------|---------------|---------------|---------------|---------------|-------------|--------------|---------------|---------------|---------------|---------------|
| Biofilm                 | Positive      | Negative      | Positive      | Negative      | Positive    | Negative     | Positive      | Negative      | Positive      | Negative      |
| <b>Positive (n= 55)</b> | 14<br>(25.5%) | 41<br>(74.5%) | 39<br>(70.9%) | 16<br>(29.1%) | 22<br>(40%) | 33<br>(60%)  | 31<br>(56.4%) | 24<br>(43.6%) | 14<br>(25.5%) | 41<br>(74.5%) |
| <b>Negative (n= 22)</b> | 1<br>(4.5%)   | 21<br>(95.5%) | 9<br>(40.9%)  | 13<br>(59.1%) | 0<br>(0 %)  | 22<br>(100%) | 2<br>(9.1%)   | 20<br>(90.9%) | 2<br>(9.1%)   | 20<br>(90.9%) |
| <b>P-value</b>          | 0.054*        |               | 0.02**        |               | 0.000*      |              | 0.000**       |               | 0.132*        |               |

\* Fisher's exact tests

\*\* Pearson Chi-square

**Table 3.** The relationship of MRSA and MSSA with the presence of biofilm-related genes

| Genes       |          | MRSA (n= 49) | MSSA (n= 28) | P-value |
|-------------|----------|--------------|--------------|---------|
| <i>icaA</i> | Positive | 12 (24.5 %)  | 3 (10.7 %)   | 0.142** |
|             | Negative | 37 (75.5 %)  | 25 (89.3 %)  |         |
| <i>icaD</i> | Positive | 29 (59.2 %)  | 19 (67.9 %)  | 0.45**  |
|             | Negative | 20 (40.8 %)  | 9 (32.1 %)   |         |
| <i>clfA</i> | Positive | 17 (34.7 %)  | 5 (17.9 %)   | 0.116** |
|             | Negative | 32 (65.3 %)  | 23 (82.1 %)  |         |
| <i>fnbA</i> | Positive | 23 (46.9 %)  | 10 (35.7 %)  | 0.338** |
|             | Negative | 26 (53.1 %)  | 18 (64.3 %)  |         |
| <i>cna</i>  | Positive | 11 (22.4 %)  | 5 (17.9 %)   | 0.633** |
|             | Negative | 38 (77.6 %)  | 23 (82.1 %)  |         |

\*\* Pearson Chi-square

Table 4. Patterns observed from gene

| Related genes patterns           | Number in pattern | MRSA / MSSA | Biofilm + / Biofilm - |
|----------------------------------|-------------------|-------------|-----------------------|
| <i>icaA</i>                      | 3                 | 2 / 1       | 2 / 1                 |
| <i>icaA – icaD</i>               | 4                 | 3 / 1       | 4 / 0                 |
| <i>icaA – fnbA</i>               | 2                 | 2 / 0       | 2 / 0                 |
| <i>icaA – icaD – fnbA</i>        | 3                 | 3 / 0       | 3 / 0                 |
| <i>icaA – icaD – clfA</i>        | 3                 | 2 / 1       | 3 / 0                 |
| <i>icaA – icaD – fnbA – clfA</i> | 2                 | 2 / 0       | 2 / 0                 |
| <i>icaD – fnbA – clfA – can</i>  | 3                 | 2 / 1       | 3 / 0                 |
| <i>icaD</i>                      | 10                | 4 / 6       | 3 / 7                 |
| <i>icaD – fnbA</i>               | 11                | 5 / 6       | 10 / 1                |
| <i>icaD – clfA</i>               | 2                 | 2 / 0       | 2 / 0                 |
| <i>icaD – can</i>                | 4                 | 2 / 2       | 3 / 1                 |
| <i>icaD – fnbA – clfA</i>        | 5                 | 3 / 2       | 5 / 0                 |
| <i>icaD – clfA – can</i>         | 3                 | 3 / 0       | 3 / 0                 |
| <i>fnbA – clfA – can</i>         | 2                 | 2 / 0       | 2 / 0                 |
| <i>fnbA</i>                      | 3                 | 2 / 1       | 2 / 1                 |
| <i>fnbA – clfA</i>               | 2                 | 2 / 0       | 2 / 0                 |
| <i>fnbA – can</i>                | 2                 | 1 / 1       | 2 / 0                 |
| <i>clfA</i>                      | 1                 | 0 / 1       | 1 / 0                 |
| <i>clfA – cna</i>                | 1                 | 1 / 0       | 1 / 0                 |
| <i>Can</i>                       | 1                 | 0 / 1       | 0 / 1                 |
| without genes                    | 12                | 7 / 5       | 2 / 0                 |

According to the results of this study, 20 different patterns were observed for the distribution of studied genes in the isolates. None of the isolates had all genes simultaneously. Two genes were observed in 28 isolates, three genes in 16 isolates, and 4 genes in 5 isolates simultaneously (Table 4).

## Discussion

Biofilm-producing *S. aureus* isolates cause bacterial resistance to antibiotics and host immune response, and cause chronic infections (22). Biofilm formation was observed in 71.4% of studied isolates, which was similar to the studies of Yousefi and Goudarzi et al. (23,24). Moreover, the ability to produce biofilm was higher in MRSA isolates,

Overall, the results of this study showed that the highest rate of gene isolation was related to *icaD* and *fnbA*, in 64.9% and 45.5% of *S. aureus* isolates, respectively. In terms of gene prevalence, their frequency rate was more than that of the study by Nourbakhsh et al. (27). and lower than that of the study by Demir et al. (28). The *icaA* gene was

compared to MSSA isolates, consistent with the findings of a study performed by Goodarzi et al. (24). In a study by Zalipour et al., the rate of biofilm formation in *S. aureus* isolates was 54.4% by the Congo Red Method (25). Karki et al. showed the ability to produce biofilm in more than 90% among *S. aureus* isolates (26). Differences in the biofilm properties of isolates among various studies might be due to the origins of bacteria, the geographical area, and genetic characterization. Several studies have shown that biofilm formation ability in *S. aureus* isolates is associated with the presence of *ica* locus (especially *icaA* and *icaD*) and *MSCRAMMs* proteins (14,16).

detected in 19.5% of the isolates, which is close to the results of a study by Keikhaie et al. (29). In the study of Azmi et al., the rate of gene *clfA* isolation was higher than that of our study (30). The lowest frequency was related to the *cna* gene, which is similar to Garcia et al. (31). In the present study, there was a significant association between the

attendance of *fnbA*, *clfA*, *icaD* genes and biofilm formation in *S. aureus* isolates; however, this connection was not observed for *icaA* and the *cna* gene. According to the results of this study, the frequency of the *icaA*, *icaD*, *fnbA*, *clfA*, and *cna* genes in *MRSA* and *MSSA* isolates was lower than that of the study by Ghasemian et al., and the statistical difference between *MRSA* and *MSSA* regarding the frequency of the genes (*clfA*, *fnbA*, *cna*, *icaD*, *icaA*) was not significant (32).

*S. aureus* is one of the bacteria that is of particular importance in nosocomial infections, especially surgical site infections. Biofilm formation can lead to increased pathogenicity, protection of microorganisms against the host's defense system and antibacterial agents, and the lack of early recovery of such patients. Therefore, it is necessary to observe the hygienic standards and proper sterilization of treatment tools to prevent the formation of biofilms in these infections.

## Conclusions

The results of the present study showed that the most common genes associated with biofilm were *icaD* and *fnbA*. There was a significant association between the presence of genes and biofilm formation in *S. aureus* isolates, but this connection was not observed for the *cna* gene.

## Ethics Approval and Consent to Participate

This study was approved by the Ethics Committee of the University of Medical Sciences of Yasuj, Iran (Ethics Code: IR.YUMS.REC.1397.124). Written informed consent was obtained from all participants before their enrollment in the study.

## Consent for Publication

Not applicable.

## Data Availability Statement

The datasets generated and/or analyzed during the current study are available from the corresponding author upon reasonable request.

## Funding Statement

The authors did not receive any specific grant from the government, commercial, or not-for-profit sectors.

## Acknowledgements

The authors express their gratitude to all participants for their involvement in this study.

## Authors' Contribution

ZSN contributed to the study design, supervised data acquisition procedures, and drafted the initial manuscript. FM conceived and designed the study, supervised the entire research process, interpreted the results, and was the primary contributor to the writing and revision of the final manuscript. The authors read and approved the final version of the manuscript and agreed to be accountable for all aspects of the work.

## Conflict of Interest

All authors declared that there are no conflicts of interest to report.

## Declaration of Generative Artificial Intelligence (AI) in Scientific Writing

Artificial intelligence was not used at any stage of this study.

## References

1. Chaudhary R, Thapa SK, Rana JC, Shah PK. Surgical site infections and antimicrobial resistance pattern. 2015. [DOI: [10.3126/jnhrc.v15i2.18185](https://doi.org/10.3126/jnhrc.v15i2.18185)]
2. Pal S, Sayana A, Joshi A, Juyal D. Staphylococcus aureus: A predominant cause of surgical site infections in a rural healthcare setup of Uttarakhand. J Family Med Prim Care. 2019 Nov 1;8(11):3600-6. [DOI: [10.4103/jfmpe.jfmpe\\_521\\_19](https://doi.org/10.4103/jfmpe.jfmpe_521_19)]
3. Mardanpour K, Rahbar M, Mardanpour S, Mardanpour N. Surgical site infections in orthopedic surgery: incidence and risk factors at an Iranian teaching hospital. CTOD. 2017;2(4):132. [DOI: [10.4103/2542-4157](https://doi.org/10.4103/2542-4157)]
4. Gao P, Ho PL, Yan B, Sze KH, Davies J, Kao RY. Suppression of Staphylococcus aureus virulence by a small-molecule compound. PNAS. 2018 Jul 31;115(31):8003-8. [DOI: [10.1073/pnas.1720520115](https://doi.org/10.1073/pnas.1720520115)]
5. Choudhary KS, Mih N, Monk J, Kavvas E, Yurkovich JT, Sakoulas G, Palsson BO. The Staphylococcus aureus two-component system AgrAC displays four distinct genomic arrangements that delineate genomic virulence factor signatures. Front microbiol. 2018 May 25;9:1082. [DOI: [10.3389/fmicb.2018.01082](https://doi.org/10.3389/fmicb.2018.01082)]
6. Antri, K., et al., High levels of Staphylococcus aureus and MRSA carriage in healthy population of Algiers revealed by additional enrichment and multisite screening. Eur J Clin Microbiol Infect Dis. 2018 Aug;37(8):1521-9. [DOI: [10.1007/s10096-018-3279-6](https://doi.org/10.1007/s10096-018-3279-6)]
7. Chikkala R, Ch S, Divyakolu S, Ratnakar KS, Sritharan V. A Simple Sample Processing Protocol and Multiplex PCR for Direct Detection of MRSA

- from Uncultured Clinical Samples—A Pilot Study. *AID*. 2019 Mar 7;9(1):25-38. [DOI: [10.4236/AID.2019.91003](https://doi.org/10.4236/AID.2019.91003)]
8. Maleki DT, Ghalavand Z, Nikmanesh B, Hashemi A, Kadhoda H, Eslam G. Identification of Virulence Genes in *Staphylococcus aureus* Isolates Segregated from Children's Wounds. *Pejouhesh dar Pezeshki*. 2019 Oct 9;43(1). [URL: <http://pejouhesh.sbmu.ac.ir/article-1-1935-en.html>]
  9. Habib MB, Shah NA, Amir A, Kamran M. Antimicrobial resistance and biofilm formation in implants related infections: Pathogens profiling and implants susceptibility. *Diagn Microbiol Infect Dis*. 2025 Aug 12;117061. [DOI: [10.1016/j.diagmicrobio.2025.117061](https://doi.org/10.1016/j.diagmicrobio.2025.117061)]
  10. Assefa M, Amare A. Biofilm-associated multi-drug resistance in hospital-acquired infections: a review. *Infect Drug Resist*. 2022 Jan 1;5061-8. [DOI: [10.2147/IDR.S379502](https://doi.org/10.2147/IDR.S379502)]
  11. Pal S, Sayana A, Joshi A, Juyal D. *Staphylococcus aureus*: A predominant cause of surgical site infections in a rural healthcare setup of Uttarakhand. *J Family Med Prim Care*. 2019 Nov 1;8(11):3600-6. [DOI: [10.4103/jfmpe.jfmpe\\_521\\_19](https://doi.org/10.4103/jfmpe.jfmpe_521_19)]
  12. Pajohesh R, Tajbakhsh E, Momtaz H, Rahimi E. Relationship between Biofilm Formation and Antibiotic Resistance and Adherence Genes in *Staphylococcus aureus* Strains Isolated from Raw Cow Milk in Shahrekord, Iran. *Int J Microbiol*. 2022 Oct 25;2022:6435774. [DOI: [10.1155/2022/6435774](https://doi.org/10.1155/2022/6435774)]
  13. Armoon, M., Babapour, E., Mirnejad, R., Babapour, M., & Taati Moghadam, M. (2024). Evaluation of *icaA* and *icaD* genes involved in biofilm formation in *Staphylococcus aureus* isolates from clinical sources using Reverse Transcriptase PCR. *Arch. Razi Inst*. 2024 Dec 31;79(6):1329. [DOI: [10.32592/ARI.2024.79.6.1329](https://doi.org/10.32592/ARI.2024.79.6.1329)]
  14. Moormeier DE, Bayles KW. *Staphylococcus aureus* biofilm: a complex developmental organism. *Mol Microbiol*. 2017 May;104(3):365-76. [DOI: [10.1111/mmi.13634](https://doi.org/10.1111/mmi.13634)]
  15. Khoramian, B., et al., Comparison of virulence factors and biofilm formation among *Staphylococcus aureus* strains isolated from human and bovine infections. *Microb Pathog*. 2015 Nov 1;88:73-7. [DOI: [10.1016/j.micpath.2015.08.007](https://doi.org/10.1016/j.micpath.2015.08.007)]
  16. Eftekhari F, Rezaee R, Azad M, Azimi H, Goudarzi H, Goudarzi M. Distribution of adhesion and toxin genes in *staphylococcus aureus* strains recovered from hospitalized patients admitted to the ICU. *Arch Pediatr Infect Dis*. 2017 Jan;5(1):e39349. [DOI: [10.5812/PEDINFECT.39349](https://doi.org/10.5812/PEDINFECT.39349)]
  17. Halim RM, Kassem NN, Mahmoud BS. Detection of biofilm producing staphylococci among different clinical isolates and its relation to methicillin susceptibility. *Open Access Maced J Med Sci*. 2018 Aug 5;6(8):1335. [DOI: [10.3889/oamjms.2018.246](https://doi.org/10.3889/oamjms.2018.246)]
  18. Cayci YT, Coban AY, Gunaydin M. Investigation of plasmid-mediated quinolone resistance in *Pseudomonas aeruginosa* clinical isolates. *Indian J Med Microbiol*. 2014 Jul 1;32(3):285-9. [DOI: [10.4103/0255-0857.136567](https://doi.org/10.4103/0255-0857.136567)]
  19. Zmantar T, Chaieb K, Makni H, Miladi H, Abdallah FB, Mahdouani K, Bakhrouf A. Detection by PCR of adhesins genes and slime production in clinical *Staphylococcus aureus*. *J Basic Microbiol*. 2008 Aug;48(4):308-14. [DOI: [10.1002/jobm.200700289](https://doi.org/10.1002/jobm.200700289)]
  20. Darwish SF, Asfour HA. Investigation of biofilm forming ability in *Staphylococci* causing bovine mastitis using phenotypic and genotypic assays. *Sci World*. 2013;2013(1):378492. [DOI: [10.1155/2013/378492](https://doi.org/10.1155/2013/378492)]
  21. Japoni A, Jamalidoust M, Farshad S, Ziyaeyan M, Alborzi A, Japoni S, et al. Characterization of SCCmec types and antibacterial susceptibility patterns of methicillin-resistant *Staphylococcus aureus* in Southern Iran. *Jpn J Infect Dis*. 2011;64(1):28-33. [PMID: [21266752](https://pubmed.ncbi.nlm.nih.gov/21266752/)]
  22. Parastan R, Kargar M, Solhjoo K, Kafizadeh F. *Staphylococcus aureus* biofilms: Structures, antibiotic resistance, inhibition, and vaccines. *Gene Rep*. 2020 Sep 1;20:100739. [DOI: [10.1016/j.genrep.2020.100739](https://doi.org/10.1016/j.genrep.2020.100739)]
  23. Yousefi M, Pourmand MR, Fallah F, Hashemi A, Mashhadi R, Nazari-Alam A. Characterization of *Staphylococcus aureus* biofilm formation in urinary tract infection. *Iran J Public Health*. 2016;45(4):485. [URL: <https://ijph.tums.ac.ir/index.php/ijph/article/view/6579>]
  24. Goudarzi M, Mohammadi A, Amirpour A, Fazeli M, Nasiri MJ, Hashemi A, et al. Genetic diversity and biofilm formation analysis of *Staphylococcus aureus* causing urinary tract infections in Tehran, Iran. *J Infect Dev Ctries*. 2019;13(09):777-85. [DOI: [10.3855/jidc.11329](https://doi.org/10.3855/jidc.11329)]
  25. Zalipour M, Ebrahim-Saraie HS, Sarvari J, Khashei R. Detection of biofilm production capability and *icaA/D* genes among staphylococci isolates from Shiraz, Iran. *Jundishapur J Microbiol*. 2016 Dec 1;9(12):1L. [DOI: [10.5812/jjm.41431](https://doi.org/10.5812/jjm.41431)]
  26. Karki S, Sah AK, Lamichhane J, Maharjan A, Sharma L, Rajbhandari R, et al. Biofilm Formation and Detection of *icaD* Gene in *Staphylococcus aureus* Isolated from Clinical Specimens. *Open Microbiol J*. 2019 Aug 30;13(1). [URL: <https://www.sciencedirect.com/org/science/article/pii/S1874285819000293>]
  27. Nourbakhsh F, Namvar AE. Detection of genes involved in biofilm formation in *Staphylococcus aureus* isolates. *GMS Hyg Infect Control*. 2016 Mar 22;11:Doc07

- [PMID: 4804124]
28. Demir C, Demirci M, Yigin A, Tokman HB, Yildiz SC. Presence of biofilm and adhesin genes in *Staphylococcus aureus* strains taken from chronic wound infections and their genotypic and phenotypic antimicrobial sensitivity patterns. *PD-PDT*. 2020 Mar 1;29:101584 [DOI: [10.1016/j.pdpdt.2019.101584](https://doi.org/10.1016/j.pdpdt.2019.101584)]
  29. Keikhaie R, Sargazi A, Hassanshahian M, Shahi Z. Detection of Intercellular Adhesion Genes (*icaA* and *icaD*) in *Staphylococcus aureus* Clinical Isolates in Zabol-Iran. 2017;5. [DOI: [10.18869/acadpub.rmm.5.1.40](https://doi.org/10.18869/acadpub.rmm.5.1.40)]
  30. Azmi K, Qrei W, Abdeen Z. Screening of genes encoding adhesion factors and biofilm production in methicillin resistant strains of *Staphylococcus aureus* isolated from Palestinian patients. *BMC genomics*. 2019;20(1):578. [DOI: [10.1186/s12864-019-5929-1](https://doi.org/10.1186/s12864-019-5929-1)]
  31. Uribe-García A, Paniagua-Contreras GL, Monroy-Pérez E, Bustos-Martínez J, Hamdan-Partida A, Garzón J, et al. Frequency and expression of genes involved in adhesion and biofilm formation in *Staphylococcus aureus* strains isolated from periodontal lesions. *J Microbiol Immunol Infect*. 2021 Apr 1;54(2):267-75. [DOI: [10.1016/j.jmii.2019.05.010](https://doi.org/10.1016/j.jmii.2019.05.010)]
  32. Ghasemian A, Peerayeh SN, Bakhshi B, Mirzaee M. Comparison of biofilm formation between methicillin-resistant and methicillin-susceptible isolates of *Staphylococcus aureus*. *Iran Biomed J*. 2016;20(3):175. [PMID: 26948126]