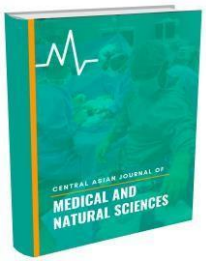


CENTRAL ASIAN JOURNAL OF MEDICAL AND NATURAL SCIENCES
https://cajmns.centralasianstudies.org/index.php/CAJMNS
Volume: 06 Issue: 04 | October 2025 ISSN: 2660-4159

.

Article

Identification and Antibiotic Sensitivity of *Staphylococcus aureus* Isolated from Burned-Wounded Patients

Mustafa D Younus*[1], Yahya Abbas Qasim[2], Ali Qasim Taha Muhammad[3], Maad Abdullah Mansur[4], Mohammed H Khalaf[5]

1,2,3,4,5. Department of Nursing Techniques, Kirkuk Technical Medical Institute, Northern Technical University, Iraq
* Correspondence: mustafathanoony@ntu.edu.iq

Abstract: Burns are the leading cause of death globally and the most severe type of trauma. resistant to methicillin One of the most prevalent bacteria associated with burn wound infections is *Staphylococcus aureus* (MRSA); nevertheless, antibiotic resistance in these strains has complicated therapy. Biofilm generation, a virulence factor that enhances antibiotic resistance, is the cause of treatment failure and recurrent staphylococcal infections in burn patients. In the current research, 50 samples were collected from burn and wound patients hospitalized to various hospitals in Erbil city. Thirty *S. aureus* isolates were identified using culture, morphological characteristics, biochemical tests, and Vitek's two compact methods. *S. aureus* generated yellow pigments on mannitol salt agar. *S. aureus* isolates were treated with several distinct antibiotics. the majority of isolates shown strong resistance to Ampicillin 100%, Ceftazidime 100%, Cefotaxime 100%, and Amikacin 6.6%. All *S. aureus* isolates were examined for biofilm production, and 71% of them produced robust biofilms. Impenem was the most efficacious antimicrobial drug against all *S. aureus* isolates.

Keywords: *Staphylococcus aureus*, Burns, Wounds, Biofilm, Antibiotic

Citation: Younus M. D., Qasim Y. A., Muhammad A. Q. T., Mansur M. A., Khalaf M. H. Identification and Antibiotic Sensitivity of *Staphylococcus aureus* Isolated from Burned-Wounded Patients. Central Asian Journal of Medical and Natural Science 2025, 6(4), 1707-1715.

Received: 23rd Jun 2025
Revised: 30th Jun 2025
Accepted: 20th Jul 2025
Published: 31th Jul 2025

1. Introduction

Staphylococcus aureus is the most dangerous species and the etiological source of many diseases that affect both humans and animals. It can cause nosocomial infections in hospitals and is isolated from the community, where it can affect the skin, bloodstream, lower respiratory tract, and urinary tract [1]. It is a type of natural plant that grows on the skin and in the nasal cavity. Nasal carriers of *S. aureus* comprise around 25–30% of the population in good health [2].

Burns are the most prevalent and dangerous type of trauma [3], and they continue to be a major public health issue and a leading cause of death globally [4,5]. In patients hospitalised with burns, the risk of microbial colonisation and infection is quite high because of the loss of protective barriers and the resulting decrease in cellular and humoral defence [6]. One of the most prevalent infections in burn injuries is *Staphylococcus aureus*. It has the potential to create a vast number of virulence factors, which are critical inpathogenesis and infectious invasion [7]. A major contributing cause to both acute and chronic infections that raise mortality rates and healthcare expenses is the global rise of organisms with multiple drug resistances (MDRs), such as methicillin-resistant *Staphylococcus aureus* (MRSA) [8]. The treatment of burn wound infections has been

hampered by the challenge of selecting the right medications because of antibiotic resistance [9, 10].

In relation to the research, biofilm formation serves a crucial role in the pathogenesis and drug resistance of *S. aureus* strains, resulting in highly harmful bacteria [11]. The severity of *S. aureus* infection is multifaceted, including its capacity to create biofilms and escape the human immune system [12]. Biofilm keeps pathogens from host defenses and hinders antibiotic administration, potentially slowing wound healing [13].

2. Materials and Methods

2.1 Collection of Specimens

A total of 50 burn patient specimens obtained from children, young people, adults attending (West Erbil Emergency Hospital) and the relevant data have been collected from every patient, Age and Gender, and residency. A swab was used containing a transportation media, and then all burn samples transferred directly to the laboratory within half an hour of collection for further processing.

2.2 Isolation and identification of *S. aureus*

Isolation and identification of *S. aureus* was based on the following characteristics:

2.3 Bacterial culture

Samples were inoculated on (Blood agar and Manitol salt agar), for this purpose all disposable swabs have been spreaded on these two media in a disposable petri plates, then incubated them overnight at 37 C.

2.4 Microscopic examination

This includes shape of the cell and reaction to gram stain. Smears prepared for isolated bacterial culture, stained with gram stain and examined under 1ight microscope using oil immersion objectives lens [14].

2.5 Biochemical Tests

Three biochemical tests were performed for confirming this bacteria is *Staphylococcus au*reus, Urease test, Catalase and Coagulase tests.

2.6 Vitek 2 compact system for identification of bacteria

The Vitek 2 system recognizes bacteria and other microbes by analyzing substrate consumption patterns. Card selection based on the organism's growing circumstances and gram stain. The Vitek 2 compact system delivers same-day, clinically relevant identification and susceptibility test findings for the majority of organisms encountered in the lab [15].

2.7 Antibiotic susceptibility test

2.8 Disk diffusion test

The disk diffusion sensitivity test was performed in accordance with the directions in the CLSI article. Three or four colonies were selected during a culture grown overnight on mannitol salt agar and transferred to 0.5 ml of phosphate buffered sa1ine. The suspension was adjusted to achieve turbidity leve1s equivalent to the 0.5 McFar and standard scale. Within 15 minutes of adjusting the turbidity, a sterile cotton swab was placed into the inoculum solution and spun numerous times against the front wall of the tube to get rid of excess liquid. Mueller-Hinton plates were streaked three times, and the plate flipped 60 degrees between each streak to ensure uniform inoculation. The inoculated plates were let to stand for 3 to 15 minutes prior adding the disks. Five antimicrobial drugs were utilized, spaced no closer than 24 mm apart. Following organism inoculation and disk placement, all inocu1ated plates were p1aced in an ambient-air incubator at 37°C for 18-24 hours. The zones of inhibition were measured from the back of the plate to the nearest whole millimeter using the ruler shown in table 3-6. The zone diameters of inhibition were interpreted using the NCCLS criteria [16].

Table 1. The standard inhibition zone of *S. aureus* for different antibiotics.

Antimicrobial agents	Concentration (mcg)	Symbol	Resistant mm or less	Intermediate mm	Sensitive mm or more
Ceftazidime	30	CAZ	≤ 14	15-17	≥ 18
Ampicillin	25	AM	≤ 28	------	≥ 29
Amikacin	10	AK	≤ 14	15- 16	≥ 17
Cefotaxime	30	CTX	≤ 14	15-22	≥ 23
Imipenem	10	AM	≤ 13	14-15	≥ 16

* Concentrations revealed above are according to Bioanalysis Company

2.9 Detection of hemolytic activity

The hemolytic activity of the bacterial isolates was tested by the blood agar plate's method. The bacterial isolates were cultured on blood agar (5 %) incubated at 37 °C for 24 hours. The zone of hemolytic production by the colony of bacteria was observed [17].

2.10 Biofilm formation of *S. aureus*

Biofilm formation in all isolates was detected by tube adherence method (TM). *S. aureus* were grown in TSB and then stained with crystal violet.

3. Results

3.1 Collection of *S. aureus* isolates

Fifty burn, wound samples were collected from patients admitted to West Erbil Emergency Hospital. Thirty isolates presumptively diagnosed as *S. aureus*, representing 60 % of total, a series of confirming tests were conducted to verify that all bacterial isolates reclaimed belong to species of *S. aureus*.

3.2 Identification of *S. aureus*

3.2.1 Microscopic examination of *S. aureus*

All isolates microscopically examined by gram stain, the result after staining showed that *S. aureus* is a gram-positive purple grape shaped bacterium non-spore forming.

3.2.2 Culture media

Staphylococci colonies appeared yellow colonies with yellow zones on Mannitol Salt Agar [18].

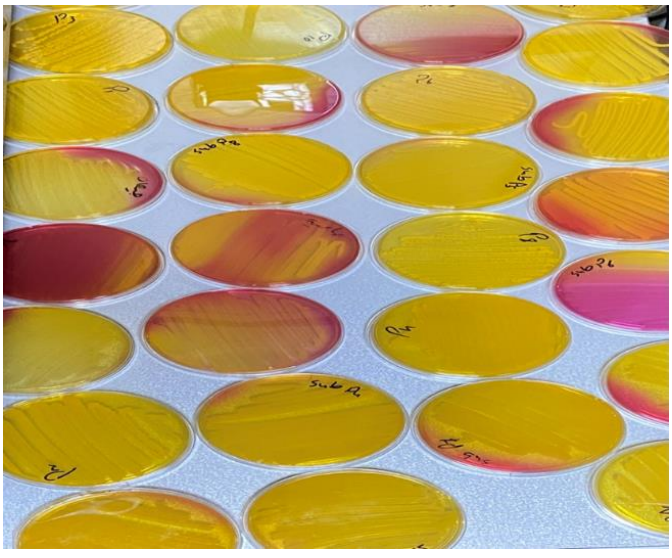


Figure 1. *S. aureus* on mannitol salt agar.

3.3 Biochemical Tests

All isolates of *S. aureus* shows 100% positive results for urease, catalases and coagulase as shown in the following figures below:

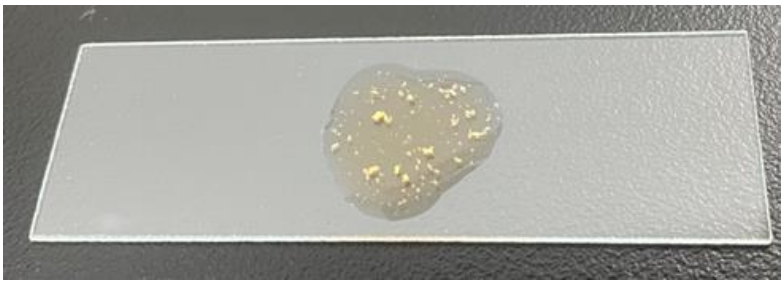
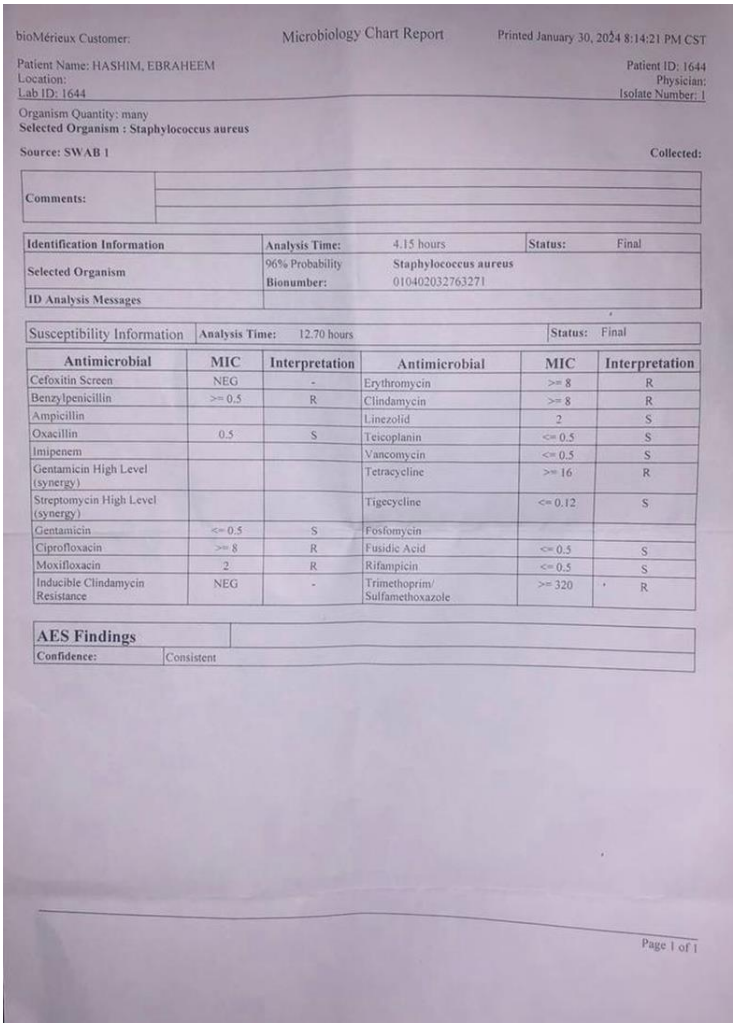


Figure 2. Shows *S. aureus* coagulases positive through slide method.

3.4 Identification of *S. aureus* by Vitek 2 system

All isolates of from burns samples have been subjected to Vitek 2 compact system test to confirm identification of this pathogen. Using this technique showed that all isolates were belong to one biotype according to 64 biochemical tests present in Vitek 2 compact system. This system selected the isolates to the 96% as S. *aureus.*



bioMérieux Customer: Microbiology Chart Report Printed January 30, 2024 8:14:21 PM CST

Patient Name: HASHIM, EBRAHEEM Patient ID: 1644
Location: Physician:
Lab ID: 1644 Isolate Number: 1

Organism Quantity: many
Selected Organism : Staphylococcus aureus

Source: SWAB 1 **Collected:**

Comments:

Identification Information	**Analysis Time:**	4.15 hours	**Status:**	Final
Selected Organism	96% Probability	**Staphylococcus aureus**		
	Bionumber:	0104020327632 71		
ID Analysis Messages				

Susceptibility Information	**Analysis Time:**	12.70 hours	**Status:**	Final

Antimicrobial	MIC	Interpretation	Antimicrobial	MIC	Interpretation
Cefoxitin Screen	NEG	-	Erythromycin	>= 8	R
Benzylpenicillin	>= 0.5	R	Clindamycin	>= 8	R
Ampicillin			Linezolid	2	S
Oxacillin	0.5	S	Teicoplanin	<= 0.5	S
Imipenem			Vancomycin	<= 0.5	S
Gentamicin High Level (synergy)			Tetracycline	>= 16	R
Streptomycin High Level (synergy)			Tigecycline	<= 0.12	S
Gentamicin	<= 0.5	S	Fosfomycin		
Ciprofloxacin	>= 8	R	Fusidic Acid	<= 0.5	S
Moxifloxacin	2	R	Rifampicin	<= 0.5	S
Inducible Clindamycin Resistance	NEG	-	Trimethoprim/ Sulfamethoxazole	>= 320	R

AES Findings
Confidence: Consistent

Page 1 of 1

Figure 3. Shows the identification probability and MIC for *S. aureus* by VITEK 2 compact system.

3.5 Antimicrobial sensitivity screening test for *S. aureus*

Thirty *S. aureus* isolates were screened for their resistance to (5) widely used antibiotics including (Ceftazidime, Amikacin, Ampicillin, Cefotaxime and Imipenem). Table 2 revealed the percentage of resistance for bacterial isolates to different antibiotics understudy. The resistance percentage for Ceftazidime, Ampicillin and Cefotaxime were 100%, and for Amikacin 6.6%, lastly for Imipenem shows no resistance, which means these antibiotics was the best among them.

Table 2. Percentage of resistance bacterial isolates to different antibiotics.

Antibiotics	Total No. of isolates	No. of resistant Isolates	% of Resistant
Ceftazidime	30	30	100%
Ampicillin	30	30	100%
Amikacin	30	2	6.6%
Cefotaxime	30	30	100%
Imipenem	30	0	0%

Figure 4. Antibiotic Susceptibility Test Performed against *S. aureus* by Using the Kirby-Bauer Disk Diffusion Method on Muller-Hinton Agar.

Hemolytic activity of *S. aureus* isolates

In the present study, All *S. aureus* isolates 30 (100%) were β- hemolytic producers.

Figure 5. Hemolytic activity of *S. aureus.*

3.6 Biofilm formation of *S. aureus*

Using the TM technique, biofilm development was detected in 21 (71%) of the *S. aureus* isolates. The incidence of antimicrobial resistance was higher in biofilm-producing *S. aureus* than in weak biofilm and non-producers. Crucially, all weak biofilm and non-producers of *S. aureus* were non-MDR, while the majority of biofilm-producing *S. aureus* were multidrug resistant (MDR). While a significant percentage of biofilm producers were confirmed to be MRSA, none of the biofilm non-producers were.

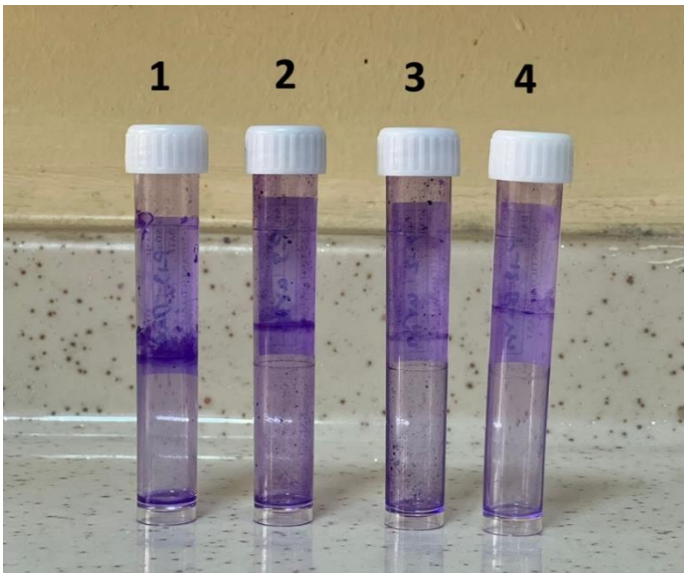


Figure 6. Biofilm formation of *Staphylococcus aureus*, tube No.1 showed very strong biofilm formation according to the grouping of the bacteria that forms a ring around the tube and at the bottom, tube No.2 also showed strong biofilm formation, unlike tube No.3 that showed moderate biofilm formation, lastly tube No4. Showed very weak biofilm production.

4. Discussion

This study was conducted for the isolation of *S. aureus* and determination of antimicrobial sensitivity test for 5 different types of antibiotics. Our results showed that the most effective antibiotics used in this study was imipenem, it is a class of carbapenems that work against the bacteria by inhibiting their growth.

A study by Gitau et al., 2018 [19] done on *Staphylococcus aureus* collected from clinical samples demonstrated that 50% of the samples were resistant to amikacin, which disagrees with our result. Another result by Onifade et al., 2018 [20] conducted in Nigeria on multidrug resistant bacteria agree with our findings, *Staphylococcus aureus* were sensitive to amikacin antibiotic.

Rasmi et al., 2022 [21] reported that along with wound infection isolates the percentage resistances of cefotaxime were 90% which agrees with our results, while other result obtained by Ghimire et al., 2020 [22] showed that the isolates were resistance to ceftazidime with 75% which was near to our results.

S. aureus demonstrated high resistance to Penicillin G, Chloramphenicol, Azithromycin, Ampicillin, Erythromycin, Trimethoprim, Clindamycin and Methicillin, which agrees with a result obtained by Maharjan et al., 2021 [23].

What makes MRSA different from a typical staph infection is its resistance to the antibiotic methicillin and other common antibiotics, such as amoxicillin, oxacillin, and penicillin. This means these antibiotics do not work on the infection. That's why a MRSA infection is so difficult to treat. Also Gram-positive bacteria acquire resistance to beta-

lactam antibiotics through the production of a protein called PBP2a (Penicillin-Binding Protein 2a), which is able to avoid the inhibitory effects of the antibiotics.

Mahmoudi et al., 2019; Shehade et al., 2025 [24, 25] and Rashid et al., 2022; Hemmati et al., 2024 [26, 27] reported that most of *S. aureus* were positive for biofilm formation, which both agree with our findings.

Bacteria form biofilms in response to environmental stresses such as UV radiation, desiccation, limited nutrients, extreme pH, extreme temperature, high salt concentrations, high pressure, and antimicrobial agents.

A research done by Al-Khamis et al., 2025 [28] on biofilm production of *S. aureus* in Iraq showed that all isolates were positive for strong biofilm formation which agrees with our findings. Another study performed by Hiawy and Mukharmish, 2019 [29] in Al-Kut city on molecular detection of biofilm genes in *S. aureus* like icaA, icaB and icaC, the study showed most of the *S. aureus* isolates were carrying these genes which also agrees with our results.

The agreement of biofilm production in *Staphylococcus aureus* across different provinces of Iraq suggests that biofilm formation is a common virulence trait among circulating strains. This may be due to shared genetic factors such as the icaADBC operon and widespread selective pressure in clinical settings.

For the Imipenem antibiotic one of the research was done by Jaafar and Shareef, 2025 [30] in Babylon city on relationship between *Staphylococcus aureus* biofilm formation and antibiotic resistance, their result were near to ours regarding the antibiotic resistance, because their isolates showed only 9.25% resistance to imipenem while our isolates were all resistant to imipenem.

The variation in antibiotic susceptibility of *Staphylococcus aureus* across provinces is likely due to differences in antibiotic usage patterns, prescribing habits, and local resistance development. In Erbil, the high effectiveness of imipenem may reflect lower exposure and less resistance pressure, while in other areas, overuse or misuse of the same antibiotic may have led to resistance. Additionally, the presence of different resistance genes and strain types, along with variations in infection control practices, can influence how effective certain antibiotics are from one region to another [31].

This study has several limitations, including a small sample size and restriction to hospitals in Erbil, which may limit the generalizability of the findings. The lack of molecular analysis of resistance and biofilm-related genes, absence of clinical outcome data, and limited antibiotic panel also reduce the depth of the study. Additionally, the method for biofilm detection was not specified, and the cross-sectional design prevents analysis of trends over time.

5. Conclusion

Staphylococcus aureus was identified as the most significant and leading cause of wound burn infections, making it the primary focus of this study. Antibiotic susceptibility testing was conducted using five different antibiotics, revealing that *S. aureus* exhibited high resistance to most of them. Among the tested antibiotics, Imipenem proved to be the most effective. Notably, biofilm-producing strains of *S. aureus* demonstrated a strong tendency toward antimicrobial and multidrug resistance. Therefore, regular monitoring of biofilm formation and antimicrobial resistance profiles of *S. aureus* is essential for enabling timely and effective treatment of wound infections.

Recommendations

a. Genetic site detection of *S. aureus* antibiotic resistance genes through molecular assay and transformation.
b. Identification of Biofilm formation genes through molecular.
c. Furthermore, study, such as DNA sequencing of *S. aureus*, to determine the exact function of certain virulence genes.
d. For more accurate results, large number of samples must be taken.

REFERENCES

[1] G. Li, M. J. Walker, and D. M. De Oliveira, "Vancomycin resistance in Enterococcus and Staphylococcus aureus," *Microorganisms*, vol. 11, no. 1, p. 24, Dec. 2022. DOI: 10.3390/microorganisms11010024.

[2] C. Pérez, T. Zúñiga, and C. E. Palavecino, "Photodynamic therapy for treatment of Staphylococcus aureus infections," *Photodiagnosis Photodyn. Ther.*, vol. 34, p. 102285, Jun. 2021. DOI: 10.1016/j.pdpdt.2021.102285.

[3] A. Marco-Fuertes, C. Marin, C. Gimeno-Cardona, V. Artal-Muñoz, S. Vega, and L. Montoro-Dasi, "Multidrug-resistant commensal and infection-causing Staphylococcus spp. isolated from companion animals in the Valencia region," *Vet. Sci.*, vol. 11, no. 2, p. 54, Jan. 2024. DOI: 10.3390/vetsci11020054.

[4] S. Samir, A. El-Far, H. Okasha, R. Mahdy, F. Samir, and S. Nasr, "Isolation and characterization of lytic bacteriophages from sewage at an Egyptian tertiary care hospital against methicillin-resistant Staphylococcus aureus clinical isolates," *Saudi J. Biol. Sci.*, vol. 29, no. 5, pp. 3097–3106, May 2022. DOI: 10.1016/j.sjbs.2022.03.019.

[5] F. de Lucca Melo, A. Gragnani, A. F. de Oliveira, and L. M. Ferreira, "Predicting mortality for critically ill burns patients, using the Abbreviated Burn Severity Index and Simplified Acute Physiology Score 3," *Injury*, vol. 53, no. 2, pp. 453–456, Feb. 2022. DOI: 10.1016/j.injury.2021.11.027.

[6] S. Kumar, R. P. Mahato, C. S., and S. Kumbham, "Current strategies against multidrug-resistant Staphylococcus aureus and advances toward future therapy," *The Microbe*, p. 100281, Feb. 2025. DOI: 10.1016/j.microb.2025.100281.

[7] B. B. Silva, M. Silva Júnior, F. G. Menezes, and E. J. Troster, "Factors associated with multidrug-resistant bacteria in healthcare-associated infections: A pediatric intensive care unit case-control study," *Einstein (Sao Paulo)*, vol. 20, eAO6704, Apr. 2022. DOI: 10.31744/einstein_journal/2022AO6704.

[8] C. Wang, H. Lu, X. Li, Y. Zhu, Y. Ji, W. Lu, G. Wang, W. Dong, M. Liu, X. Wang, and H. Chen, "Identification of an anti-virulence drug that reverses antibiotic resistance in multidrug resistant bacteria," *Biomed. Pharmacother.*, vol. 153, p. 113334, Sep. 2022. DOI: 10.1016/j.biopha.2022.113334.

[9] E. Hernández-Cuellar, K. Tsuchiya, R. Valle-Ríos, and O. Medina-Contreras, "Differences in biofilm formation by methicillin-resistant and methicillin-susceptible Staphylococcus aureus strains," *Diseases*, vol. 11, no. 4, p. 160, Nov. 2023. DOI: 10.3390/diseases11040160.

[10] A. Alsolami, N. S. ALGhasab, M. S. Alharbi, A. I. Bashir, M. Saleem, A. S. Syed Khaja, D. F. Aldakheel, E. Rakha, J. A. Alshammari, T. E. Taha, Z. Melibari, "Community-acquired methicillin-resistant Staphylococcus aureus in hospitals: Age-specificity and potential zoonotic–zooanthroponotic transmission dynamics," *Diagnostics*, vol. 13, no. 12, p. 2089, Jun. 2023. DOI: 10.3390/diagnostics13122089.

[11] M. M. Severn and A. R. Horswill, "Staphylococcus epidermidis and its dual lifestyle in skin health and infection," *Nat. Rev. Microbiol.*, vol. 21, no. 2, pp. 97–111, Feb. 2023. DOI: 10.1038/s41579-022-00780-3.

[12] A. Valliammai, A. Selvaraj, U. Yuvashree, C. Aravindraja, and S. Karutha Pandian, "sarA-dependent antibiofilm activity of thymol enhances the antibacterial efficacy of rifampicin against Staphylococcus aureus," *Front. Microbiol.*, vol. 11, p. 1744, Jul. 2020. DOI: 10.3389/fmicb.2020.01744.

[13] M. Bhattacharya and A. R. Horswill, "The role of human extracellular matrix proteins in defining Staphylococcus aureus biofilm infections," *FEMS Microbiol. Rev.*, vol. 48, no. 1, p. fuae002, Jan. 2024. DOI: 10.1093/femsre/fuae002.

[14] E. K. Perry and M. W. Tan, "Bacterial biofilms in the human body: Prevalence and impacts on health and disease," *Front. Cell. Infect. Microbiol.*, vol. 13, p. 1237164, Aug. 2023. DOI: 10.3389/fcimb.2023.1237164.

[15] M. Cheesbrough, *Laboratory Practice in Tropical Countries*, vol. 1, UK: Part, 2006, pp. 228–230.

[16] M. D. Younus, A. Z. Fage Ahmed Abdulrahman, F. O. Bahjat, N. L. Lutphy Ali, S. A. Rashid, A. S. Alkhouri, "Plasmid profiling, genetic site determination of antibiotic resistance gene of Pseudomonas aeruginosa isolated from burned patients," [Online]. Available: http://doi.org/10.24086/biohs2022/paper.538.

[17] L. J. Wang, X. Yang, S. Y. Qian, Y. C. Liu, K. H. Yao, F. Dong, W. Q. Song, "Identification of hemolytic activity and hemolytic genes of methicillin-resistant Staphylococcus aureus isolated from Chinese children," *Chinese Med. J.*, vol. 133, no. 1, pp. 88–90, Jan. 2020. DOI: 10.1097/CM9.0000000000000571.

[18] H. Zhang, J. Cao, Z. He, X. Zong, and B. Sun, "Molecular epidemiology of Staphylococcus aureus in a tertiary hospital in Anhui, China: ST59 remains a serious threat," *Infect. Drug Resist.*, vol. 2023, pp. 961–976, Dec. 2023. DOI: 10.2147/IDR.S395220.

[19] W. Gitau, M. Masika, M. Musyoki, B. Museve, and T. Mutwiri, "Antimicrobial susceptibility pattern of Staphylococcus aureus isolates from clinical specimens at Kenyatta National Hospital," *BMC Res. Notes*, vol. 11, p. 1–5, Dec. 2018. DOI: 10.1186/s13104-018-3337-2.

[20] A. K. Onifade, C. O. Afolayan, and O. I. Afolami, "Antimicrobial sensitivity, extended spectrum beta-lactamase (ESBL) production and plasmid profile by microorganisms from otitis media patients in Owo and Akure, Ondo State, Nigeria," *Karbala Int. J. Mod. Sci.*, vol. 4, no. 3, pp. 332–340, Sep. 2018. DOI: 10.1016/j.kijoms.2018.07.001.

[21] A. H. Rasmi, E. F. Ahmed, A. M. Darwish, and G. F. Gad, "Virulence genes distributed among Staphylococcus aureus causing wound infections and their correlation to antibiotic resistance," *BMC Infect. Dis.*, vol. 22, p. 652, Jul. 2022. DOI: 10.1186/s12879-022-07624-8.

[22] G. Ghimire, R. P. Chaudhary, and B. Lekhak, "Bacteriological profile and antibiotic susceptibility pattern of isolates of wound infection in children visiting Kanti Children Hospital," *Tribhuvan Univ. J. Microbiol.*, vol. 7, pp. 123–132, Dec. 2020. DOI: 10.3126/tujm.v7i0.33855.

[23] B. Maharjan, S. T. Karki, and R. Maharjan, "Antibiotic susceptibility pattern of Staphylococcus aureus isolated from pus/wound swab from children attending International Friendship Children's Hospital," *Nepal J. Biotechnol.*, vol. 9, no. 1, pp. 8–17, Jul. 2021. DOI: 10.3126/njb.v9i1.38645.

[24] H. Mahmoudi, M. Pourhajibagher, N. Chiniforush, A. R. Soltanian, M. Y. Alikhani, and A. Bahador, "Biofilm formation and antibiotic resistance in methicillin-resistant and methicillin-sensitive Staphylococcus aureus isolated from burns," *J. Wound Care*, vol. 28, no. 2, pp. 66–73, Feb. 2019. DOI: 10.12968/jowc.2019.28.2.66.

[25] S. M. Shehade, A. S. Juma, M. H. Ubeid, and M. D. Younus, "Perceptions and mindset toward infection control and prevention among medical microbiology students," *Cihan Univ.-Erbil Sci. J.*, vol. 9, no. 1, pp. 24–28, Jan. 2025. DOI: 10.24086/cuesj.v9n1y2025.pp24-28.

[26] S. A. Rashid, S. M. Sorchee, M. D. Yonus, and O. F. Bahjat, "Identification of biofilm producers in Staphylococcus aureus isolates and detection of their biofilm genes from gingivitis cases," *4th Int. Conf. Health Biol. Sci.*, 2022. [Online]. Available: http://doi.org/10.24086/biohs2022/paper.586.

[27] J. Hemmati, M. Chiani, B. Asghari, G. Roshanaei, S. S. Soleimani Asl, M. Shafiei, and M. R. Arabestani, "Antibacterial and antibiofilm potentials of vancomycin-loaded niosomal drug delivery system against methicillin-resistant Staphylococcus aureus (MRSA) infections," *BMC Biotechnol.*, vol. 24, p. 47, Jul. 2024. DOI: 10.1186/s12896-024-00874-1.

[28] A. R. Al-Khamis, J. M. Abed, H. K. Sameer, and M. A. Hassan, "Biofilm formation and antibiotic resistance of Staphylococcus aureus isolates," *Academia Open*, vol. 10, no. 1, pp. 10–21070, Apr. 2025. DOI: 10.1186/s13104-018-3337-2.

[29] A. R. Hiawy and J. H. Mukharmish, "Molecular study to detect the prevalence of biofilm genes and the effect of probiotics on Staphylococcus aureus isolates in Al-Kut city, Iraq," 2019, pp. 1141–1148.

[30] S. S. Jaafar and H. K. Shareef, "Relationship between Staphylococcus aureus biofilm formation and antibiotic resistance isolated from infections of the urinary tract in Babylon Province, Iraq," *Med. J. Babylon*, vol. 22, no. 1, pp. 123–128, Jan. 2025. DOI: 10.4103/MJBL.MJBL_593_23.

[31] P. A. Hamad, "Phenotypic and molecular detection of biofilm formation in methicillin-resistant Staphylococcus aureus isolated from different clinical sources in Erbil city," *Mediterr. J. Hematol. Infect. Dis.*, vol. 15, no. 1, p. e2023016, Mar. 2023. DOI: 10.4084/MJHID.2023.016.