e-ISSN: 0975-1556, p-ISSN:2820-2643

Available online on www.ijpcr.com

International Journal of Pharmaceutical and Clinical Research 2023; 15(10); 178-185

Original Research Article

PCR-Based Assessment of MRSA: mecA Gene Detection Among Patients and Healthcare Workers

Dheepa N[1], A.V.Mathivadhana[2], Sasikala Gunasekaran[3], Panneerselvam Periasamy[4]

[1]Assistant Professor, Department of Microbiology, Government Erode Medical College, Perundurai, Erode, Tamilnadu, India

[2]2nd year M.B.B.S Student, KMCH Institute of Health Sciences and Research, Coimbatore, Tamilnadu, India

[3]Department of Nursing, Government Erode Medical College Hospital, Perundurai, Erode, Tamilnadu, India

[4]Assistant Professor, Department of Physiology, Government Erode Medical College, Perundurai, Erode, Tamilnadu, India

Received: 25-07-2023 / Revised: 28-08-2023 / Accepted: 30-09-2023

Corresponding author: Dr. Dheepa. N

Conflict of interest: Nil

Abstract:

Methicillin-Resistant Staphylococcus aureus (MRSA) is a notorious pathogen responsible for a wide range of healthcare-associated and community-acquired infections. Timely detection and management of MRSA colonization among both patients and healthcare workers are essential to prevent its spread and associated adverse outcomes. This research aims to screen for MRSA colonization in both patient populations and healthcare workers while employing Polymerase Chain Reaction (PCR) technique for the identification of the mecA gene, a key genetic determinant of methicillin resistance.

The study will involve a cross-sectional approach, where nasal swabs will be collected from a representative sample of patients admitted to healthcare facilities and healthcare workers across diverse departments. The collected swabs will be subjected to MRSA screening using both traditional culture methods and PCR-based techniques targeting the mecA gene. The results will be analysed to determine the prevalence of MRSA colonization, assess the concordance between the two methods, and investigate potential risk factors associated with colonization.

This research aspires to provide valuable insights into the prevalence of MRSA colonization among patients and healthcare workers, shedding light on the effectiveness of PCR-based mecA gene detection in comparison to conventional culture techniques. The findings could contribute to the development of more efficient and rapid screening protocols, facilitating early intervention and infection control measures. Ultimately, this study seeks to enhance our understanding of MRSA transmission dynamics within healthcare settings and underscore the importance of rigorous screening procedures.

Keywords: Methicillin-Resistant *Staphylococcus aureus,* MRSA, mecA gene, PCR Technique, Screening, Healthcare-Associated Infections, Colonization, Infection Control.

Introduction

The primary objective of this study was to investigate the prevalence of Methicillin-Resistant Staphylococcus aureus (MRSA) colonization among both hospitalized patients and healthcare workers.

MRSA stands as a significant public health challenge, characterized by its widespread presence and formidable resistance to multiple antibiotics. Within healthcare settings, MRSA infections pose a substantial threat, contributing to heightened rates of morbidity, mortality, and increased healthcare expenditure[1].Of particular concern is the potential role played by both patients and healthcare workers as reservoirs for MRSA, thereby facilitating its transmission within healthcare facilities[2]. The timely and accurate identification of MRSA colonization is vital for the implementation of effective infection control measures.

The emergence and persistence of Methicillin-Resistant Staphylococcus aureus (MRSA) in healthcare facilities have raised considerable concerns in recent years.[3,4,5] MRSA's ability to withstand treatment with multiple antibiotics makes it a formidable adversary in clinical settings. Its prevalence not only prolongs patient hospital stays

but also increases the risk of severe, often life-threatening infections. Moreover, the financial burden associated with managing MRSA-related complications has become a significant challenge for healthcare systems worldwide [6,7,8].

One of the critical facets of MRSA transmission within healthcare facilities is the role played by patients and healthcare workers as potential carriers of this resistant bacterium [9,10]. Understanding the extent of MRSA colonization among these groups is pivotal in curbing its spread. Consequently, early identification of MRSA colonization is paramount for the implementation of stringent infection control measures, including isolation precautions and decolonization strategies, to protect both patients and healthcare workers.

This research study aspires to address these pressing issues by meticulously examining the prevalence of MRSA colonization within our healthcare facility. By employing a cross-sectional prospective design, we aim to capture a comprehensive snapshot of MRSA colonization over a one-year period. This investigation will encompass patients admitted to various departments and healthcare workers who are on the front lines of care delivery [11,12]. Our approach to this study will be multifaceted. Traditional culture techniques will be employed to isolate and identify MRSA colonies, providing us with a foundational understanding of MRSA prevalence. In parallel, we will harness the power of Polymerase Chain Reaction (PCR) technology to detect the mecA gene—a pivotal genetic marker responsible for methicillin resistance. This dual methodology will not only confirm the presence of MRSA but also delineate its resistance profile.By shedding light on the extent of MRSA colonization and its resistance patterns, we aim to provide valuable insights into the epidemiology of MRSA within our healthcare facility. Furthermore, we anticipate that this research will contribute to the formulation of evidence-based infection control strategies tailored to our specific context. Ultimately, our goal is to mitigate the burden of MRSA-related infections, enhance patient safety, and ensure the well-being of our dedicated healthcare workers.

Materials and Methods

In the Department of Microbiology at Vinayaka Mission's Kirupananda Variyar Medical College & Hospitals, Salem, a cross-sectional prospective study was conducted from December 2011 to December 2012. This study was conducted on one hundred consecutive isolates of Staphylococcus aureus from clinical specimens received in the microbiology department from various patients and swabs collected from three different sites that is, anterior nares, palm, and web spaces of each of the one hundred healthcare workers. The specimens received from patients were inoculated into routine conventional culture medias and incubated at 37°C for 24 hrs. The swabs collected from the healthcare workers were inoculated into mannitol salt agar and incubated at 37°C for 24 hrs. After incubation, colonies from both the conventional medias and mannitol salt agar were confirmed as Staphylococcus aureus by conventional identification techniques [13,14]. A cross-sectional study will be conducted in multiple healthcare facilities, involving patients admitted to various departments and healthcare workers. Nasal swabs will be collected from participants, and demographic and clinical data will be recorded.

Nasal swabs will be collected using sterile swabs and transported to the laboratory for processing. Swabs will be cultured on selective media for MRSA isolation, and DNA will be extracted from the swabs for PCR analysis.

MRSA Screening: Traditional culture methods will be employed for MRSA screening, involving incubation and identification of characteristic colonies. Additionally, PCR will be used to amplify the mecA gene, a genetic marker of methicillin resistance.

Data Analysis: Descriptive statistics will be used to determine the prevalence of MRSA colonization. The concordance between culture-based and PCR-based methods will be assessed using appropriate statistical measures. Multivariate analysis will be performed to identify potential risk factors associated with MRSA colonization.

Out of the 100 Staphylococcus aureus isolated from patients, 68 were from male patients and 32 were from female patients. All the 100 Staphylococcus aureus isolates from patients were subjected to disc diffusion tests using oxacillin discs (1 µg) and cefoxitin discs (30 µg) and also to oxacillin screen agar (1 µg oxacillin/mL) to detect methicillin resistance. Among the 68 Staphylococcus aureus isolates from male patients and 32 Staphylococcus aureus isolates from female patients, 46 and 24 Staphylococcus aureus isolates, respectively, were confirmed as Methicillin-Resistant Staphylococcus aureus (MRSA) (Fig-1).

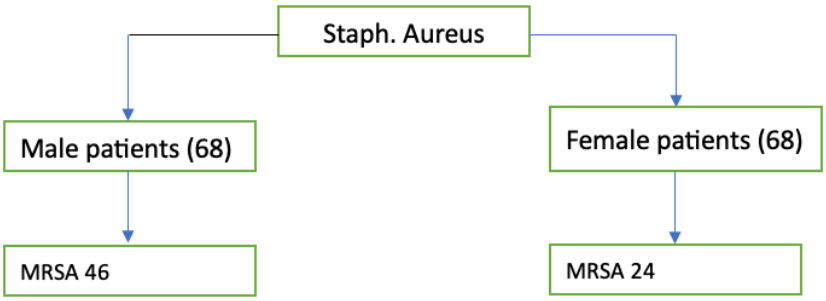

Figure 1: Detection of MRSA from Patient Sample

Out of the 70 MRSA isolates obtained from patients, the majority, 55 (78.57%), were derived from pus samples, while 10 (14.28%) were from urine, 3 (4.28%) from endotracheal tubes, and 2 (2.85%) from sputum specimens. In our assessment of 100 Staphylococcus aureus isolates from patients, we employed disc diffusion testing using oxacillin discs (1 µg) and cefoxitin discs (30 mg), in addition to oxacillin screen agar (6 µg oxacillin/ml) to detect methicillin resistance. Interestingly, only 64 (64%) isolates exhibited methicillin resistance when assessed solely by oxacillin disc diffusion, while 70 (70%) isolates demonstrated methicillin resistance when both oxacillin screen agar and cefoxitin disc diffusion tests were applied concurrently.

We subjected the 70 MRSA isolates from patients to PCR analysis for the detection of the mecA gene, considered the gold standard for this purpose. Remarkably, all 70 MRSA isolates (100%) were found to possess the mecA gene (Fig-2). the performance of the phenotypic methods for MRSA detection against the PCR-based detection of the mecA gene, we conducted a comparative analysis.

The results revealed that the oxacillin disc diffusion method exhibited a sensitivity of 92.1% and a specificity of 100%. Meanwhile, both the oxacillin screen agar and cefoxitin disc diffusion methods demonstrated a sensitivity and specificity of 100% each.

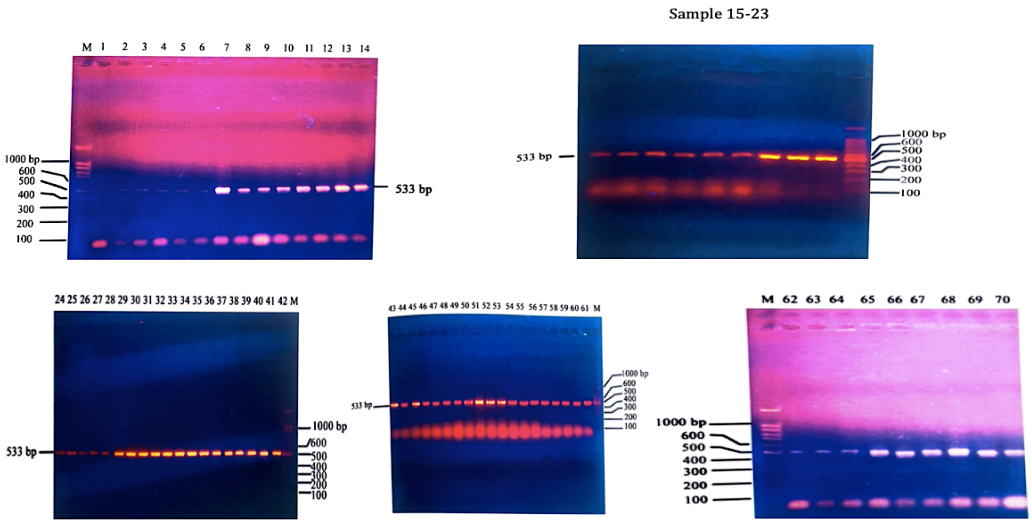

Figure 2: Agarose gel electrophoresis for mecA gene (533bp) in Patients sample 1-70

Table 1: Comparison of phenotypic methods with genotypic method of detection of Methicillin resistant Staphylococcus aureus from Patients

Test Method	Detected as MRSA	Sensitivity(100%)	Specificity (100%)
Oxacillin disc diffusion (1g)	64	92.1	100
Oxacillin screen Agar (6ng)	70	100	100
Cefoxitin disc diffusion (30ng)	70	100	100
PCR for mecA gene	70	100	100

Among the one hundred healthcare workers, 71 were female, and 29 were male. Among the female healthcare workers, 19 (26.7%) were carriers of Staphylococcus aureus, while among the male healthcare workers, 4 (13.8%) were carriers of Staphylococcus aureus. To assess methicillin

resistance, all 23 Staphylococcus aureus isolates obtained from healthcare workers underwent disc diffusion testing using oxacillin discs (1 mg) and cefoxitin discs (30 µg), as well as screening on oxacillin agar (6 µg oxacillin/ml). Out of the 10 Staphylococcus aureus isolates identified, 6 (8.45%) were methicillin-resistant Staphylococcus aureus (MRSA) among female healthcare workers, and 4 (13.78%) were MRSA among male healthcare workers (Fig3).

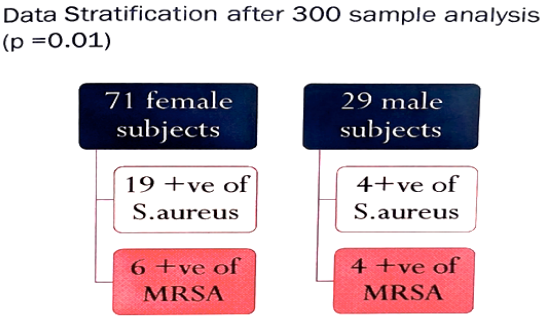

Figure 3: Isolation of MRSA from HCWs

Out of the 10 Methicillin resistant Staphylococcus aureus carriers in HCW's, 7 (70 %) were nasal carrier and 3 (30 %) were palm carriers (Fig-4).

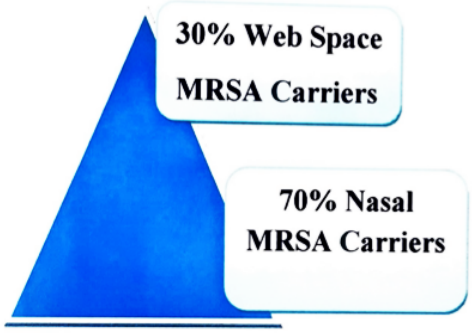

Figure 4: Percentage of MRSA carriers in HCWs

Out of 23 Staphylococcus aureus isolated in HCWs, 12 were from ICU, 7 were from Surgery ward, 3 were from Medicine ward and one was from OBG ward. Out of 12 Staphylococcus aureus isolates from ICU-5 (41.66 %) were methicillin resistant Staphylococcus aureus, out of 7 Staphylococcus aureus isolates from Surgery ward 3 (42.85 %) were methicillin resistant Staphylococcus aureus, out of 3 Staphylococcus aureus isolates from medicine ward 2 (66.6 %)were MRSA.(Chart -1)

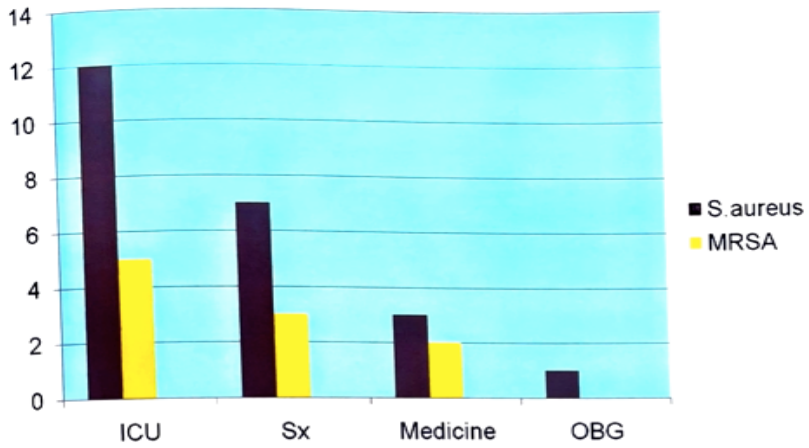

Figure 5: Isolation of MRSA among HCWs in various wards

Off the 23 Staphylococcus aureus isolates from health care workers subjected to disc diffusion test using oxacillin disc (lug) and cefoxitin disc (30 wg) and also to oxacillin screen agar (6 pg oxacillin/ml) to detect methicillin resistance, only 8 (34.78 %) isolates were detected as methicillin resistance by oxacillin disc diffusion and 10 (43.47%) isolates were detected as methicillin resistance by both

oxacillin screen agar test and cefoxitin disc diffusion test. Finally, the 10 MRSA isolates from health care workers were subjected to PCR assay for detection of mecA gene and al the 10 MRSA isolates (100 %) detected the presence of mecA gene. (Fig-9)

Table 2: Comparison of phenotypic methods with genotypic method of detection of Methicillin resistant Staphylococcus aureus from HCWs

Test Method	Detected as MRSA	Sensitivity(100%)	Specificity (100%)
Oxacillin disc diffusion (1g)	8	83.3	100
Oxacillin screen Agar (6ng)	10	100	100
Cefoxitin disc diffusion (30ng)	10	100	100
PCR for mecA gene	10	100	100

The phenotypic methods for detection of methicillin resistant Staphylococcus aureus were compared for their sensitivity and specificity with the detection of mecA gene by PCR which is the gold standard. The results showed that the sensitivity and specificity of oxacillin disc diffusion method was 93.3 % and 100 % respectively and that of both screen agar and cefoxitin disc diffusion methods were 100 % oxacillin and 100 % respective.

Discussion

This study, Aimed to investigate the prevalence of MRSA colonization among both hospitalized patients and healthcare workers. The research involved the comparison of traditional culture methods with PCR-based mecA gene detection, which is considered the gold standard for MRSA screening. The primary objectives were to validate the efficacy of molecular techniques in MRSA detection and to explore potential factors contributing to MRSA colonization. These findings hold the potential to inform targeted interventions for reducing MRSA transmission [15, 16]. Nosocomial infections continue to pose a significant burden on global healthcare systems, resulting in morbidity and mortality. MRSA, in particular, has become increasingly prevalent among hospitalized patients due to the overuse of common cephalosporins since the 1980s [17]. Recent data from the Centers for Disease Control and Prevention (CDC) have identified MRSA as the second most common cause of hospital-acquired infections, following only Escherichia coli [18,19]. In many American and European hospitals, MRSA accounts for 29% to 35% of all clinical isolates. It is important to note that the available data on MRSA prevalence primarily originates from developed countries, limiting our ability to estimate its global distribution accurately. Variations in prevalence rates have been observed both temporally and geographically, likely due to differences in clonal expansion and drug resistance pressures within communities [20,21].

In India, for example, studies have reported varying MRSA prevalence rates. Some studies indicate prevalence rates of approximately 38.44% among Staphylococcus aureus isolates, while others have reported rates as high as 90.89% in specific regions [22,23]. Over time, the burden of multidrug-resistant MRSA has shown an increasing trend, ranging from 23.2% in 2001 to 32% in 2006 [24].

In 2010, India's MRSA colonization rates were reported at 34% in Orthopaedics, 18% in Surgery wards, and only 1% in medical units. This highlights the variation in MRSA prevalence across different hospital settings [25].

Notably, MRSA carriage rates among healthcare workers (HCWs) in India have not been extensively studied, and available data vary. In our study, we found a MRSA prevalence of 10% among HCWs, with male HCWs exhibiting a higher prevalence, consistent with previous studies [26].

MRSA colonization rates on the hands of healthcare workers have shown a wide range from 10.5% to 78.3%. In our study, we observed similar results, and the sensitivity and specificity of various detection methods were consistent with previous research [27].

In recent years, molecular techniques have gained prominence in MRSA diagnosis due to their high sensitivity in identifying small numbers of resistant organisms and their ability to simultaneously detect Staphylococcus aureus and the mecAgene [28]. These techniques also offer the advantage of shorter turnaround times compared to traditional diagnostic methods [29].

In summary, our study contributes valuable insights into MRSA colonization rates among patients and healthcare workers, with a focus on comparing traditional culture methods and molecular techniques. These findings hold implications for infection control and prevention strategies in healthcare settings.

Conclusion

In conclusion, our comprehensive study provides valuable insights into the prevalence and characteristics of Methicillin-Resistant Staphylococcus aureus (MRSA) colonization among both patients and healthcare workers within our healthcare facility. The findings from our research shed light on several key aspects of MRSA epidemiology and detection methods.

Firstly, our study reveals a substantial MRSA colonization rate among patients, with a prevalence rate of 70%. This high prevalence underscores the importance of ongoing surveillance and infection control measures to manage and mitigate the impact of MRSA in healthcare settings. Among healthcare workers, the prevalence of MRSA colonization was comparatively lower, at 10%, emphasizing the need for continued monitoring and adherence to infection control practices among this group.

A notable gender disparity was observed, with males being predominant carriers of MRSA in both patients (46%) and healthcare workers (13.78%). This gender-based difference in MRSA colonization rates warrants further investigation to uncover potential underlying factors and inform targeted interventions.

Regarding the sites of MRSA isolation, our study identified that in patients, MRSA was primarily isolated from pus samples (14.28%), highlighting the significance of wound care and infection management protocols. Among healthcare workers, the anterior nares emerged as the most common site for MRSA colonization, with an 88.2% prevalence. This finding emphasizes the need for diligent nasal screening and decolonization efforts among healthcare workers to minimize the risk of MRSA transmission.

In terms of diagnostic methods, our study demonstrated that both cefoxitin disc diffusion and PCR exhibited similar sensitivity and specificity for the detection of MRSA. Importantly, the results of the cefoxitin disc diffusion test were in concordance with the PCR results for the mecA gene. This suggests that the cefoxitin disc diffusion test can serve as a viable alternative to PCR, particularly in resource-constrained settings where PCR may not be readily available.

Furthermore, our study underscores the value of PCR assays for detecting the mecA gene in Staphylococci. These molecular techniques provide a rapid and reliable means of identifying methicillin resistance, enabling timely and accurate patient management decisions. Incorporating PCR-based assays into routine susceptibility testing protocols can enhance our ability to identify intrinsic resistance patterns and guide appropriate antimicrobial therapy.

In summary, our study highlights the significant prevalence of MRSA colonization among patients and healthcare workers in our healthcare facility. It emphasizes the need for continued vigilance in infection control practices and the importance of gender-specific and site-specific surveillance. Additionally, our findings suggest that cefoxitin disc diffusion can be a practical alternative to PCR for MRSA detection, particularly in resource-limited settings. The incorporation of PCR-based assays enhances our capacity to identify methicillin resistance promptly, ultimately contributing to better patient care and infection control.

Limitations: Only a small group of the population included in the study. Large-scale studies from whole of Tamilnadu and India are needed to undertake. Another limitation of the study was that it did not have equal representative from various economic and occupational strata of society which could bias the result.

Acknowledgements: The authors would like to thank all the participated and the administration of Vinayaka Mission's Kirupananda Variyar Medical College, Salem, Tamilnadu for granting permission to carry out the research work.

Financial support and sponsorship: Nil.

Ethical statement: Institutional ethical committee accepted this study. All individuals who took part in the study gave their informed consent, and data confidentiality was ensured.

Author's contribution:

Dr. Dheepa N: Conceived, investigation, methodology, Data Collection, project administration, Responsible for Data's Integrity and Authenticity.

A.V. Mathivadhana: Literature Review, Discussion and Manuscript draft editing.

Dr.Sasikala Gunasekaran: Proof Reading, writing—original draft, writing—review and editing;

Panneerselvam Periasamy: Manuscript draft editing and statistical analysis.

All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work. All authors have read and agreed to the published version of the manuscript.

Data Availability: All datasets generated or analyzed during this study are included in the manuscript.

Informed Consent: Written informed consent was obtained from the participants before enrolling in the study.

References

1. Wang XR, Wu LF, Wang Y, Ma YY, Chen F-H, Ou H-L. Rapid detection of Staphylococcus aureus by loop-mediated isothermal amplification. ApplBiochemBiotechnol. 2015;175:882-91.
2. Khosravi AD, Khoshnood S, Montazeri EA, Jomehzadeh N, Moradi M, Shahi F. The application of the loop-mediated isothermal amplification method for rapid detection of

methicillin-resistant Staphylococcus aureus. New Microbes New Infect. 2022;45:100960.
3. Sudhaharan S, Vanjari L, Mamidi N, Nagapriyanka E, Vemu L. Evaluation of LAMP assay using phenotypic tests and conventional PCR for detection of nuc and mecA genes among clinical isolates of Staphylococcus spp. J Clin Diagn Res. 2015;9(8):DC06.
4. Chung Y, Kim TS, Min YG, Hong YJ, Park JS, Hwang SM, et al. Usefulness of multiplex real-time PCR for simultaneous pathogen detection and resistance profiling of staphylococcal bacteremia. BioMed Res Int. 2016;2016.
5. Notomi T, Okayama H, Masubuchi H, Yonekawa T, Watanabe K, Amino N, et al. Loop-mediated isothermal amplification of DNA. Nucleic Acids Res. 2000;28(12):e63-e.
6. Tian X, Feng J, Wang Y. Direct loop-mediated isothermal amplification assay for on-site detection of Staphylococcus aureus. FEMS Microbiol Lett. 2018;365(11):fny092.
7. Tomita N, Mori Y, Kanda H, Notomi T. Loop-mediated isothermal amplification (LAMP) of gene sequences and simple visual detection of products. Nat Protoc. 2008;3(5):877-82.
8. Nagamine K, Watanabe K, Ohtsuka K, Hase T, Notomi T. Loop-mediated isothermal amplification reaction using a nondenatured template. Clin Chem. 2001;47(9):1742-3.
9. Woźniakowski G, Kozdruń W, Samorek-Salamonowicz E. Loop-mediated isothermal amplification for the detection of goose circovirus. Virol J. 2012;9(1):1-11.
10. Diribe O, North S, Sawyer J, Roberts L, Fitzpatrick N, La Ragione R. Design and application of a loop-mediated isothermal amplification assay for the rapid detection of Staphylococcus pseudintermedius. J Vet Diagn Invest. 2014;26(1):42-8.
11. Panno S, Matić S, Tiberini A, Caruso AG, Bella P, Torta L, et al. Loop mediated isothermal amplification: principles and applications in plant virology. Plants. 2020;9(4):461.
12. Wong YP, Othman S, Lau YL, Radu S, Chee HY. Loop-mediated isothermal amplification (LAMP): a versatile technique for detection of micro-organisms. J ApplMicrobiol. 2018;124(3):626-43.
13. Huang Z-G, Zheng X-Z, Guan J, Xiao S-N, Zhuo C. Direct detection of methicillin-resistant Staphylococcus aureus in sputum specimens from patients with hospital-associated pneumonia using a novel multilocus PCR assay. Pathogens. 2015;4(2):199-209.
14. Rietmeyer L, Fix-Boulier N, Le Fournis C, Iannazzo L, Kitoun C, Patin D, et al. Partition of tRNA Glyisoacceptors between protein and cell-wall peptidoglycan synthesis in Staphylococcus aureus. Nucleic Acids Res. 2021;49(2):684-99.
15. Chambers HF, DeLeo FR. Waves of resistance: Staphylococcus aureus in the antibiotic era. Nat Rev Microbiol. 2009;7(9):629-41.
16. Centers for Disease Control (CDC) (2020). Identifying Healthcare-associated infections (HAI) for NHSN surveillance. Retrieved from https://www.cdc.gov/nhsn/pdfs/pscmanual/pcs manual_current.pdf.
17. Clinical and Laboratory Standards Institute (CLSI) (2022). "Performance Standards for Antimicrobial Susceptibility Testing," 32nd edition. CLSI supplement M100. Clinical and Laboratory Standards Institute, Wayne, PA.
18. Choopara I, Suea-Ngam A, Teethaisong Y, Howes PD, Schmelcher M, Leelahavanichkul A, et al. Fluorometric paper-based, loop-mediated isothermal amplification devices for quantitative point-of-care detection of methicillin-resistant Staphylococcus aureus (MRSA). ACS Sensors. 2021;6(3):742-51.
19. Chen C, Zhao Q, Guo J, Li Y, Chen Q. Identification of methicillin-resistant Staphylococcus aureus (MRSA) using simultaneous detection of mecA, nuc, and femB by loop-mediated isothermal amplification (LAMP). CurrMicrobiol. 2017; 74(8):965-71.
20. Köck R, Becker K, Cookson B, van Gemert-Pijnen J, Harbarth S, Kluytmans J, et al. Methicillin-resistant Staphylococcus aureus (MRSA): burden of disease and control challenges in Europe. Eurosurveillance. 2010;15(41).
21. Turner NA, Sharma-Kuinkel BK, Maskarinec SA, Eichenberger EM, Shah PP, Carugati M, et al. Methicillin-resistant Staphylococcus aureus: an overview of basic and clinical research. Nat Rev Microbiol. 2019;17(4):203-18.
22. Zaczek-Moczydłowska MA, Mohamed-Smith L, Toldrà A, Hooper C, Campàs M, Furones MD, et al. A single-tube HNB-based loop-mediated isothermal amplification for the robust detection of the ostreid herpesvirus 1. Int J Mol Sci. 2020;21(18):6605.
23. Zhao L, Huang X, Zhang T, Zhang X, Jiang M, Lu H, et al. A point-of-care test device for MRSA rapid detection. J Pharm Biomed Anal. 2022;209:114464.
24. Elshabrawy WO, Zaki ME, Kamel MF. Genetic and phenotypic study of methicillin-resistant Staphylococcus aureus among patients and health care workers in Mansoura University Hospital, Egypt. Iran J Microbiol. 2017;9(2):82.
25. Mahmood AR, Hussein NM, Hussein NM. Detection of biofilm-forming genes in Staphylococcus aureus clinical isolates by PCR assay. J Nat Sci Biol Med. 2022;13(1).

26. Garoy EY, Gebreab YB, Achila OO, Tekeste DG, Kesete R, Ghirmay R, et al. Methicillin-resistant Staphylococcus aureus (MRSA): prevalence and antimicrobial sensitivity pattern among patients—a multicenter study in Asmara, Eritrea. Can J Infect Dis Med Microbiol. 2019;2019.
27. Hormozi SF, Vasei N, Aminianfar M, Darvishi M, Saeedi AA. Antibiotic resistance in patients suffering from nosocomial infections in Besat Hospital. Eur J TranslMyol. 2018;28(3).
28. Abdel-Wahab F, Ghoneim M, Khashaba M, El-Gilany A-H, Abdel-Hady D. Nosocomial infection surveillance in an Egyptian neonatal intensive care unit. J Hosp Infect. 2013;83(3):196-9.
29. Abdel-Maksoud M, El-Shokry M, Ismail G, Hafez S, El-Kholy A, Attia E, et al. Methicillin-resistant Staphylococcus aureus recovered from healthcare- and community-associated infections in Egypt. Int J Bacteriol. 2016;2016.