J Clin Res Paramed Sci. 2023 December; 12(2):e143681. https://doi.org/10.5812/jcrps-143681.

Published online 2024 January 12. **Research Article**

Inducible Clindamycin Resistance in *Staphylococcus aureus* Isolates in Kermanshah, Iran

Zahra Jahanbakhshi [1], Jamileh Nowroozi [1], Zahra Kahrarian [2], Azin Tariniya Gilani [3], Mohadeseh Ahmadvand [3] and Nasrollah Sohrabi [4,*]

[1]Department of Microbiology, Collage of Science, North Tehran Branch, Islamic Azad University, Tehran, Iran
[2]Department of Microbiology, School of Medicine, Kermanshah University of Medical Sciences, Kermanshah, Iran
[3]Department of Biology, School of Science, York University, Toronto, Canada
[4]Department of Medical Laboratory Sciences, School of Allied Medical Sciences, Kermanshah University of Medical Sciences, Kermanshah, Iran

[*]*Corresponding author*: Department of Medical Laboratory Sciences, School of Allied Medical Sciences, Kermanshah University of Medical Sciences, Kermanshah, Iran. Email: nsohrabi@kums.ac.ir

Received 2023 December 20; **Accepted** 2024 January 01.

Abstract

Background: *Staphylococcus aureus* is the most common agent of nosocomial infections. Macrolide Lincosamide-Streptogramin B (MLS_B) antibiotics are the therapeutic choices for treatment of infections due to methicillin resistant *S. aureus* (MRSA) isolates. The most frequent mechanism for inducible resistance in *S. aureus* is modification in target site by *erm* (*erythromycin ribosome methylase*) genes.

Objectives: The aim of this research was to determine inducible MLSB ($iMLS_B$) and detection the *erm* genes in clinical samples of *S. aureus* isolated from hospitalized patients in the Imam Reza hospital of Kermanshah, west of Iran.

Methods: This study performed on 126 samples of *S. aureus*. Identification of isolates were performed using microbiological and biochemical procedures. Inducible resistance to clindamycin was tested by D-test. The prevalence of genes, such as *femB*, *mecA*, *ermA* and *ermB* was assessed by polymerase chain reaction (PCR).

Results: Eighty-three cases (65.9%) of isolates were methicillin resistant *S. aureus* (MRSA). The resistance rate against erythromycin and clindamycin was 67.4% and 52.2%, respectively. Totally, 49 cases (38.9%) of isolates were resistant to both erythromycin and clindamycin indicating constitutive MLSB phenotype ($cMLS_B$); 20 cases (15.9%) isolates showed positive D test indicating inducible MLSB phenotype ($iMLS_B$), while 16 cases (12.7%) were negative for D test indicating MS phenotype. Among 20 cases with $iMLS_B$ phenotype, *ermC* and *ermA* genes were showed in 7 cases (35%) and 4 cases (20%) isolates, respectively. The *ermB* gene is not detected in any cases and 9 cases (45%) isolates did not have any *erm* genes.

Conclusions: In general, findings of this study showed high frequency of resistance to clindamycin and erythromycin among *S. aureus* isolates and $cMLS_B$ to be the most pattern phenotype and *ermC* gene is the most common gene in $iMLS_B$ phenotype. Because variation of antimicrobial resistance pattern in geographic regions obtaining local results is useful for detecting and more appropriate control of nosocomial infection due to *S. aureus* isolates.

Keywords: *Staphylococcus aureus*, Inducible Resistance, Clindamycin, *ermA*

1. Background

Staphylococcus aureus is the gram-positive bacterium that causes many infections and syndromes. *Staphylococcus aureus* is the most common agent of nosocomial infections. The highest rate of infections due to *S. aureus* were seen among hospitalized patients with predisposing factors (1-3).

Methicillin-resistant *S. aureus* (MRSA) is commonly emerging in the types of nosocomial and community aquired infections (1-3). The growing up of prevalence of MRSA isolates is an increasing problem (1). In this situation we need to use other antibiotics. Macrolide lincosamide-streptogramin B (MLS_B) antibiotics are the therapeutic choices for treatment of infections due to MRSA isolates. Clindamycin also has some advantages, such as less cost and inhibition of production of some toxins and virulence factors in staphylococci (4, 5). Clindamycin is a common antibiotic to treatment of respiratory tract, bone, joint, skin and, soft tissue infections. This antibiotic has low side-effects. Thus, it

is appropriate for prolonged therapy (6, 7). Different mechanisms are responsible for bacterial resistance to macrolides including efflux pump, enzymatic antibiotic inactivation and target site modification (5). One of the prevalent mechanism of resistance to clindamycin is inducible resistance ($iMLS_B$) and the most frequent mechanism for inducible resistance in *S. aureus* is modification in target site by *erm* (erythromycin ribosome methylase) genes. The treatment by clindamycin in patients with $iMLS_B$ resistance phenotype may lead to constitutive MLSB phenotype ($cMLS_B$) and treatment of this infections is failed. The common laboratories cannot detect inducible resistance (erythromycin-resistant and clindamycin-sensitive) by routine laboratory procedure. The common phenotypic method to detect inducible clindamycin resistance is the method is known as D-test because flattening of zone (D-shaped) around clindamycin disk in the area between the two disks (erythromycin and clindamycin) that reveal inducible clindamycin resistance (D-test positive) (7, 8).

2. Objectives

In this study, we evaluated inducible clindamycin resistance in *S. aureus* strains isolated from hospitalized patients in the Imam Reza Hospital in Kermanshah Province, west of Iran.

3. Methods

3.1. Study Design

This descriptive study was performed on 126 strains of *S. aureus* strains isolated from hospitalized patients in the Imam Reza Hospital in Kermanshah Province, west of Iran, from June to December 2019.

3.2. Sample Collection and Laboratory Identification

The samples were obtained from all of clinical specimens, such as blood, cerebral spinal fluid (CSF), urine, vaginal, nasal, pus, tracheal and, wound swabs. The samples were inoculated in Blood agar and Mannitol Salt agar plates and incubated at 35°C for 24 - 48 hours. Gram stain, catalase, coagulase and DNAase tests were used for identification of *S. aureus* isolates. The confirmation of *S. aureus* isolates was performed by demonstration of *femB* gene presence using polymerase chain reaction (PCR) method (9).

3.3. Antibiotic Resistance Rate to Clindamycin and Erythromycin

The disk diffusion method was used for determination of the antibiotic resistance rate of isolates against clindamycin and erythromycin based on Clinical and Laboratory Standard Institute (CLSI) guidelines (10). We used clindamycin (2 μg) and erythromycin (15 μg) disks. Antibiotic disks were placed 30 mm apart on surface of the plates. They were incubated at 37°C for 18 - 24 h. Findings on the diameter of the halo created around the antibiotic disk were measured using a millimeter ruler and interpreted based on CLSI guidelines (10).

3.4. Cefoxitin Disk Diffusion Test

For detection of MRSA isolates by phenotypic method, we used cefoxitin disk (30 μg). Inhibition zones diameter of $\leq$ 21 mm were considered as MRSA (10).

3.5. Inducible Resistance to Clindamycin by D-test

In this test inducible resistance to clindamycin was detected by D-test based on CLSI guidelines (10). Briefly, erythromycin (15 μg) disk was placed apart on 15 mm from clindamycin (2 μg) disk on a Mueller-Hinton agar plate. After overnight incubation at 37°C, flattening of zone (D-shaped) around clindamycin in the area between the two disks, reveal inducible clindamycin resistance (D-test positive). There are three different phenotypes for interpretation of this test.

(1) Inducible MLSB ($iMLS_B$), phenotype (D+): Resistance to erythromycin and sensitive to clindamycin (zone size $\geq$ 21 mm) with a D-zone of inhibition around the clindamycin disk.

(2) Constitutive MLSB phenotype ($cMLS_B$): Resistance to both erythromycin and clindamycin

(3) MS phenotype: Resistance to erythromycin and sensitive to clindamycin, D-test negative.

(4) The susceptible phenotype (S phenotype): Sensitive to both clindamycin and erythromycin (11).

3.6. PCR Method

All *S. aureus* isolates were investigated for *femB*, *mecA*, *ermA*, *ermB*, and *ermC* genes using specific primers (Table 1) (12-16). Total DNA was extracted using a High Pure PCR Template Preparation Kit (Roche, Basel, Switzerland). PCR amplification was performed using automated thermal cycler (Applied Biosystems, USA). The used PCR conditions were including: Initial denaturation at 95°C for 5 minutes, denaturation at 1 minute, annealing at different degrees for each gene (Table 1) for 1 minute, extension at 70°C for 1 minute for 35 cycles and final extension at 70°C for 10 minutes. Gel electrophoresis of PCR products carried out

in 1.5% agarose gel at 85 V for 45 minutes and visualized on an ultraviolet (UV) transilluminator (BioRad, USA).

3.7. Data Analysis

Statistical calculations were performed using SPSS software version 16 for descriptive statistics of data. Statistical significance of differences between findings was evaluated by chi-square (χ^2) test. A P-value $<$ 0.05 was considered statistically significant.

4. Results

In this study 126 isolates were confirmed as *S. aureus* using phenotypic methods and presence of *femB* gene. 53 cases (42.6%) and 73 cases (57.4%) were isolated from males and females, respectively. According to presence of *mecA* gene, 83 cases (65.9%) were MRSA and 43 cases (34.1%) were methicillin sensitive *S. aureus* (MSSA). The resistance rate against erythromycin and clindamycin were 67.4% and 52.2%, respectively. In all *S. aureus* isolates 49 cases (38.9%) of isolates resistant to both erythromycin and clindamycin indicating constitutive MLSB phenotype ($cMLS_B$ phenotype); 31 cases (24.6%) isolates were sensitive to both erythromycin and clindamycin, 20 cases (15.9%) isolates showed positive D-test indicating inducible MLSB phenotype ($iMLS_B$ phenotype), while 16 cases (12.7%) were negative for D test indicating MS phenotype, 10 cases (7.9%) isolates were sensitive to erythromycin and resistance to clindamycin (S phenotype). The rate of inducible clindamycin resistance in MRSA isolates was higher than in MSSA isolates (P-value $<$ 0.05). The rate of $cMLS_B$ phenotype and $iMLS_B$ phenotype among MRSA isolates were 50.6% and 19.3%, respectively. Among MSSA isolates rate of $cMLS_B$ phenotype and $iMLS_B$ phenotype were 16.3% and 9.3%, respectively (Table 2).

These iMLSB resistance phenotype isolates were investigated for the presence of the *erm* genes. The *ermC* and *ermA* genes were showed in 7 cases (35%) and 4 cases (20%) isolates, respectively. All 11 positive cases for *erm* genes, were isolated from MRSA isolates. The *ermB* gene is not detected in any cases and, 9 cases (45%) isolates did not have any *erm* genes.

5. Discussion

Staphylococcus aureus is the most common agent of nosocomial infections. In recent years, prevalence of MRSA isolates has been increased and treatment of infections due to these isolates has been harder than before (1). In this situation we need to use other antibiotics. Macrolide Lincosamide-Streptogramin B antibiotics are the therapeutic choices for treatment of infections due to MRSA isolates. Clindamycin also has some advantages such as less cost and inhibition of production of some toxins and virulence factors in staphylococci (4, 5). The presence of clindamycin resistance phenotype in *S. aureus* clinical isolates could render effectiveness of this antibiotic for treatment of infections due to *S. aureus* isolates. This resistance may be constitutive or inducible (7, 8). In this present study we aimed to evaluate prevalence of resistance pattern to erythromycin and clindamycin and also *erm* genes occurrence in *S. aureus* isolates.

In this study, we used a D-test for phenotypically detection of several susceptibility pattern using erythromycin and clindamycin disks that located apart on surface of culture medium. There are several different phenotypes for interpretation of this test. Totally $cMLS_B$, $iMLS_B$, S and MS phenotypes were seen in 38.9%, 15.9%, 24.6 and 12.7% of *S. aureus* isolates, respectively.

In this study, $iMLS_B$ phenotype D-test positive (resistance to erythromycin and sensitive to clindamycin) was seen in 15.9% isolates. This result is higher than the result of other studies were conducted by Kilany in Egypt and Rahbar and Hajia in Iran that $iMLS_B$ phenotype were detected in 7.7% and 10.8% of *S. aureus* isolates, respectively (17, 18), and lower than other previous studies were performed by Raut et al. and Bobenchick et al. that they reported $iMLS_B$ phenotype in 25.6% and 22.3% isolates, respectively (19, 20). In some studies, the $iMLS_B$ rate was reported very high (82% and 88%) (21, 22). Findings of this study revealed the prevalence of the $iMLS_B$ phenotype between MRSA and MSSA isolates was statistically different (P $<$ 0.05). Resistant strains with $iMLS_B$ phenotype can lead to $cMLS_B$ phenotype and cause failure in treatment with clindamycin. The most prevalent clindamycin resistant phenotype in this study was $cMLS_B$ phenotype (resistance to both erythromycin and clindamycin) (38.9% of isolates) and this finding showed that $cMLS_B$ phenotype was higher than $iMLS_B$ phenotype similar to other study was conducted by Mahesh et al. (23). In the present study, the prevalence of $cMLS_B$ phenotype in MRSA isolates was significantly more than MSSA isolates, which in consistent with studies were performed in other countries (23, 24). The high frequency of $cMLS_B$ phenotype in MRSA isolates may be due to the selection pressure after therapeutic failure of methicillin and utilization of erythromycin and clindamycin. The $iMLS_B$ strains may be changed to $cMLS_B$, thus laboratories have to detect $iMLS_B$ strains by D-test and eradicate these strains by effective therapeutic agents. In present study, MS phenotype (resistance to erythromycin and sensitive to clindamycin, D-test negative) was observed in 12.7% *S. aureus* isolates. In accordance to our study, the prevalence of MS phenotype

Table 1. Primer for the Determination of *ermA*, *ermB*, *mecA*,*femB* and the Amplified Product Size

Genes and Primers	Sequences (5' to 3')	Product Size, bp	Annealing Temperature (°C)	References
mecA		147	62	14
F	GTGAAGATATACCAAGTGATT			
R	ATGCGCTATAGATTGAAAGGAT			
femB		388	55	15
F	CGTGAGAATGATGGCTTTGA			
R	TTAATACGCCCATCCATCGT			
ermA		190	54	12
F	AAGCGGTAAACCCCTCTGA			
R	TTCGCAAATCCCTTCTCAAC			
ermB		142	55	13
F	CTATCTGATTGTTGAAGAAGGATT			
R	GTTTACTCTTGGTTTAGGATGAAA			
ermC		297	52	16
F	AATCGTCAATTCCTGCATGT			
R	TAATCGTGGAATACGGGTTTG			

Abbreviations: F, forward primer; R, reverse primer.

Table 2. Susceptibility to Erythromycin (ERY) and Clindamycin (CL) Among all *Staphylococcus aureus* Isolates [a]

Susceptibility Pattern	Total (n = 126)	MRSA (n = 83)	MSSA (n = 43)	P-Value
ERY-R, CL-R (cMLSB phenotype)	49 (38.9)	42 (50.6)	7 (16.3)	< 0.05
ERY-S, CL-S (S phenotype)	31 (24.6)	8 (9.6)	23 (53.5)	< 0.05
ERY-R, CL-S (D test positive, iMLSB phenotype)	20 (15.9)	16 (19.3)	4 (9.3)	< 0.05
ERY-R, CL-S (D test negative, MS)	16 (12.7)	13 (15.7)	3 (7)	< 0.05
ERY-S, CL-R	10 (7.9)	4 (4.8)	6 (13.9)	0.48

Abbreviations: ERY, erythromycin; CL, clindamycin; S, sensitive; R, resistant; MRSA, methicillin resistant *S. aureus*; MSSA, methicillin susceptible *S. aureus*; S phenotype, sensitive to both erythromycin and clindamycin; $cMLS_B$, constitutive resistance macrolide, lincosamide, and streptogramin B; $iMLS_B$, inducible resistance macrolide, lincosamide, and streptogramin B; D test positive, MS phenotype: resistant to erythromycin and sensitive to clindamycin; D test negative, S phenotype, sensitive to both erythromycin and clindamycin.
[a] Values are expressed as No. (%).

is low in other study (25).

We investigated frequency of *erm* genes (*ermA*, *ermB* and *ermC*) in *S. aureus* isolates with $iMLS_B$ phenotype by PCR method. Our findings of this study showed of 20 MRSA isolates with $iMLS_B$ phenotype, the *ermA* and *ermC* genes were found in 4 cases (20%) and 7 cases (35%) isolates, respectively. The *ermB* gene is not detected in any cases. The *ermC* gene was the most prevalent gene in *S. aureus* isolates with $iMLS_B$ phenotype in present study. This result is similar to many studies revealed that *ermC* gene was associated with the majority of resistance to erythromycin among the MRSA isolates (26, 27). Contrary to results of this study, in some studies, *ermA* gene more frequent than *ermC* gene such as the study were conducted by Saderi in Iran that he reported prevalence of *ermA* and *ermC* in 60.3% and 54.8% of isolates that these results are higher than our findings in this study (28). Schmitz detected the *ermA* gene in 67% isolates and the *ermC* gene in 23% (29). In Korea, Jung et al. identified *ermA* gene in 89% and *ermC* gene in 5% isolates (30). In Iran, Moosavian et al. detected the *ermA* and *ermC* genes in 41.1% and 17.7% of *S. aureus* isolates, respectively (31). The *ermB* gene is not detected in any cases in this study. Cetin et al similar to our study found no *ermB* gene in *S. aureus* isolates (32). Other study reported low prevalence of *ermB* gene (33). The *ermB* gene usually detected in staphylococci spp. of animal origin and spread between streptococci and enterococci (33). These difference in prevalence of *S. aureus* isolates with $iMLS_B$ phenotype may be correlated to patients age, geographical area, rate of using antibiotics, bacterial species, source of

specimens and community or nosocomial infections. The presence of other mechanisms of resistance leading to the complexity of resistance in *S. aureus* to MLS_B antibiotics.

5.1. Conclusions

In general, results of this research showed high frequency of resistance to clindamycin and erythromycin among *S. aureus* isolates and $cMLS_B$ to be the most pattern phenotype. The *ermC* gene is the most isolated gene. Because variation of antimicrobial resistance pattern in geographic regions, obtaining local results is useful for detecting and more appropriate control of nosocomial infection due to *S. aureus* isolates. We recommended conducting the D-test for detection of $iMLS_B$ phenotypes in microbiology laboratories because the D-test is simple and cheap method that can be done in every routine laboratory.

Acknowledgments

This research was performed as the thesis of Mrs. Zahra Jahanbakhshi to get a master degree in microbiology. We are thankful to the research laboratory of Department of Medical Laboratory Sciences in Paramedical School, Kermanshah University of Medical Sciences.

Footnotes

Authors' Contribution: Study concept and design: N. S., and J. N., and Z. J.; acquisition of data: N. S., and Z. J.; analysis and interpretation of data: N. S., and Z. J., and Z. K., and A. T., and M. A.; drafting of the manuscript: N. S., and Z. J.; critical revision of the manuscript for important intellectual content: N. S., and Z. J., and Z. K., and A. T., and M. A.; statistical analysis: N. S., and J. N.; administrative, technical, and material support: N. S., and J. N., and Z. J.; study supervision: N. S., and J. N.

Conflict of Interests: The authors have no conflicts of interests.

Data Availability: The dataset presented in the study is available on request from the corresponding author during submission or after publication.

Funding/Support: There is no funding/support.

References

1. Tong SY, Davis JS, Eichenberger E, Holland TL, Fowler VJ. Staphylococcus aureus infections: Epidemiology, pathophysiology, clinical manifestations, and management. *Clin Microbiol Rev*. 2015;**28**(3):603–61. [PubMed ID: 26016486]. [PubMed Central ID: PMC4451395]. https://doi.org/10.1128/CMR.00134-14.
2. Lowy FD. Staphylococcus aureus infections. *N Engl J Med*. 1998;**339**(8):520–32. [PubMed ID: 9709046]. https://doi.org/10.1056/NEJM199808203390806.
3. Stefani S, Chung DR, Lindsay JA, Friedrich AW, Kearns AM, Westh H, et al. Meticillin-resistant Staphylococcus aureus (MRSA): Global epidemiology and harmonisation of typing methods. *Int J Antimicrob Agents*. 2012;**39**(4):273–82. [PubMed ID: 22230333]. https://doi.org/10.1016/j.ijantimicag.2011.09.030.
4. Deotale V, Mendiratta DK, Raut U, Narang P. Inducible clindamycin resistance in Staphylococcus aureus isolated from clinical samples. *Indian J Med Microbiol*. 2010;**28**(2):124–6. [PubMed ID: 20404457]. https://doi.org/10.4103/0255-0857.62488.
5. Ajantha GS, Kulkarni RD, Shetty J, Shubhada C, Jain P. Phenotypic detection of inducible clindamycin resistance among Staphylococcus aureus isolates by using the lower limit of recommended inter-disk distance. *Indian J Pathol Microbiol*. 2008;**51**(3):376–8. [PubMed ID: 18723962]. https://doi.org/10.4103/0377-4929.42515.
6. Fiebelkorn KR, Crawford SA, McElmeel ML, Jorgensen JH. Practical disk diffusion method for detection of inducible clindamycin resistance in Staphylococcus aureus and coagulase-negative staphylococci. *J Clin Microbiol*. 2003;**41**(10):4740–4. [PubMed ID: 14532213]. [PubMed Central ID: PMC254362]. https://doi.org/10.1128/JCM.41.10.4740-4744.2003.
7. Saderi H, Oulia P, Eslami M. Prevalence of Macrolide-Lincosamide-Streptogramin B (MLSB) resistance in S. aureus isolated from patients in Tehran, Iran. *Iran J Pathol*. 2009. Persian.
8. Siberry GK, Tekle T, Carroll K, Dick J. Failure of clindamycin treatment of methicillin-resistant Staphylococcus aureus expressing inducible clindamycin resistance in vitro. *Clin Infect Dis*. 2003;**37**(9):1257–60. [PubMed ID: 14557972]. https://doi.org/10.1086/377501.
9. Mahon CR, Manuselis G. *Textbook of diagnostic microbiology*. **355**. Saunders; 2000.
10. Clinical Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing; twenty-second informational supplement. *CLSI Document M 100-S22*. 2012.
11. Steward CD, Raney PM, Morrell AK, Williams PP, McDougal LK, Jevitt L, et al. Testing for induction of clindamycin resistance in erythromycin-resistant isolates of Staphylococcus aureus. *J Clin Microbiol*. 2005;**43**(4):1716–21. [PubMed ID: 15814990]. [PubMed Central ID: PMC1081368]. https://doi.org/10.1128/JCM.43.4.1716-1721.2005.
12. Ghanbari F, Ghajavand H, Havaei R, Jami MS, Khademi F, Heydari L, et al. Distribution of erm genes among Staphylococcus aureus isolates with inducible resistance to clindamycin in Isfahan, Iran. *Adv Biomed Res*. 2016;**5**:62. [PubMed ID: 27135031]. [PubMed Central ID: PMC4832884]. https://doi.org/10.4103/2277-9175.179184.
13. Zhang K, Sparling J, Chow BL, Elsayed S, Hussain Z, Church DL, et al. New quadriplex PCR assay for detection of methicillin and mupirocin resistance and simultaneous discrimination of Staphylococcus aureus from coagulase-negative staphylococci. *J Clin Microbiol*. 2004;**42**(11):4947–55. [PubMed ID: 15528678]. [PubMed Central ID: PMC525205]. https://doi.org/10.1128/JCM.42.11.4947-4955.2004.
14. Patel M, Waites KB, Moser SA, Cloud GA, Hoesley CJ. Prevalence of inducible clindamycin resistance among community- and hospital-associated Staphylococcus aureus isolates. *J Clin Microbiol*. 2006;**44**(7):2481–4. [PubMed ID: 16825368]. [PubMed Central ID: PMC1489468]. https://doi.org/10.1128/JCM.02582-05.
15. Liu D. *Molecular Detection of Human Bacterial Pathogens*. 2011. https://doi.org/10.1201/b10848.
16. Fasihi Y, Saffari F, Kandehkar Ghahraman MR, Kalantar-Neyestanaki D. Molecular detection of macrolide and lincosamide-resistance genes in clinical methicillin-resistant Staphylococcus aureus Isolates from Kerman, Iran. *Arch Pediatr Infect Dis*. 2016;**5**(1). https://doi.org/10.5812/pedinfect.37761.

17. Kilany A. Inducible clindamycin resistance among clinical isolates of Staphylococcus aureus. *Menoufia Med J*. 2016;**29**(2). https://doi.org/10.4103/1110-2098.192418.
18. Rahbar M, Hajia M. Inducible clindamycin resistance in Staphylococcus aureus: A cross-sectional report. *Pak J Biol Sci*. 2007;**10**(1):189–92. [PubMed ID: 19070014]. https://doi.org/10.3923/pjbs.2007.189.192.
19. Raut S, Bajracharya K, Adhikari J, Pant SS, Adhikari B. Prevalence of methicillin resistant Staphylococcus aureus in Lumbini Medical College and Teaching Hospital, Palpa, Western Nepal. *BMC Res Notes*. 2017;**10**(1):187. [PubMed ID: 28577365]. [PubMed Central ID: PMC5457603]. https://doi.org/10.1186/s13104-017-2515-y.
20. Bobenchik AM, Hindler JA, Giltner CL, Saeki S, Humphries RM. Performance of Vitek 2 for antimicrobial susceptibility testing of Staphylococcus spp. and Enterococcus spp. *J Clin Microbiol*. 2014;**52**(2):392–7. [PubMed ID: 24478467]. [PubMed Central ID: PMC3911353]. https://doi.org/10.1128/JCM.02432-13.
21. Gardiner BJ, Grayson ML, Wood GM. Inducible resistance to clindamycin in Staphylococcus aureus: Validation of Vitek-2 against CLSI D-test. *Pathology*. 2013;**45**(2):181–4. [PubMed ID: 23277176]. https://doi.org/10.1097/PAT.0b013e32835cccda.
22. Lavallee C, Rouleau D, Gaudreau C, Roger M, Tsimiklis C, Locas MC, et al. Performance of an agar dilution method and a Vitek 2 card for detection of inducible clindamycin resistance in Staphylococcus spp. *J Clin Microbiol*. 2010;**48**(4):1354–7. [PubMed ID: 20164285]. [PubMed Central ID: PMC2849538]. https://doi.org/10.1128/JCM.01751-09.
23. Mahesh CB, Ramakant BK, Jagadeesh VS. The prevalence of inducible and constitutive clindamycin resistance among the nasal isolates of staphylococci. *J Clin Diagn Res*. 2013;**7**(8):1620–2. [PubMed ID: 24086856]. [PubMed Central ID: PMC3782913]. https://doi.org/10.7860/JCDR/2013/6378.3223.
24. Prabhu K, Rao S, Rao V. Inducible clindamycin resistance in staphylococcus aureus isolated from clinical samples. *J Lab Physicians*. 2011;**3**(1):25–7. [PubMed ID: 21701659]. [PubMed Central ID: PMC3118052]. https://doi.org/10.4103/0974-2727.78558.
25. Nashwa M, Noha TAE. Phenotypic and genotypic detection of macrolide-lincosamide-streptogramin B resistance among clinical isolates of Staphylococcus aureus from Mansoura University Children Hospital, Egypt. *African J Microbiol Res*. 2017;**11**(12):488–94. https://doi.org/10.5897/ajmr2017.8471.
26. Khodabandeh M, Mohammadi M, Abdolsalehi MR, Alvandimanesh A, Gholami M, Bibalan MH, et al. Analysis of Resistance to Macrolide-Lincosamide-Streptogramin B Among mecA-Positive Staphylococcus Aureus Isolates. *Osong Public Health Res Perspect*. 2019;**10**(1):25–31. [PubMed ID: 30847268]. [PubMed Central ID: PMC6396818]. https://doi.org/10.24171/j.phrp.2019.10.1.06.
27. Schmitz FJ, Sadurski R, Kray A, Boos M, Geisel R, Kohrer K, et al. Prevalence of macrolide-resistance genes in Staphylococcus aureus and Enterococcus faecium isolates from 24 European university hospitals. *J Antimicrob Chemother*. 2000;**45**(6):891–4. [PubMed ID: 10837446]. https://doi.org/10.1093/jac/45.6.891.
28. Saderi H, Emadi B, Owlia P. Phenotypic and genotypic study of macrolide, lincosamide and streptogramin B (MLSB) resistance in clinical isolates of Staphylococcus aureus in Tehran, Iran. *Med Sci Monit*. 2011;**17**(2):BR48–53. [PubMed ID: 21278685]. [PubMed Central ID: PMC3524716]. https://doi.org/10.12659/msm.881386.
29. Schmitz FJ, Petridou J, Fluit AC, Hadding U, Peters G, von Eiff C. Distribution of macrolide-resistance genes in Staphylococcus aureus blood-culture isolates from fifteen German university hospitals. M.A.R.S. Study Group. Multicentre Study on Antibiotic Resistance in Staphylococci. *Eur J Clin Microbiol Infect Dis*. 2000;**19**(5):385–7. [PubMed ID: 10898143]. https://doi.org/10.1007/s100960050500.
30. Jung YH, Kim KW, Lee KM, Yoo JI, Chung GT, Kim BS, et al. Prevalence and characterization of macrolide-lincomycin-streptogramin B-resistant Staphylococcus aureus in Korean hospitals. *J Antimicrob Chemother*. 2008;**61**(2):458–60. [PubMed ID: 18156279]. https://doi.org/10.1093/jac/dkm483.
31. Moosavian M, Shoja S, Rostami S, Torabipour M, Farshadzadeh Z. Inducible clindamycin resistance in clinical isolates of Staphylococcus aureus due to erm genes, Iran. *Iran J Microbiol*. 2014;**6**(6):421.
32. Cetin ES, Gunes H, Kaya S, Aridogan BC, Demirci M. Macrolide-lincosamide-streptogramin B resistance phenotypes in clinical staphylococcal isolates. *Int J Antimicrob Agents*. 2008;**31**(4):364–8. [PubMed ID: 18206352]. https://doi.org/10.1016/j.ijantimicag.2007.11.014.
33. Aktas Z, Aridogan A, Kayacan CB, Aydin D. Resistance to macrolide, lincosamide and streptogramin antibiotics in staphylococci isolated in Istanbul, Turkey. *The Journal of Microbiology*. 2007;**45**(4):286–90.