

ORIGINAL RESEARCH

Inducible clindamycin resistant among Methicillin-Resistant *Staphylococcus aureus* isolated from Clinical Specimens**¹Dr. Prashant Peshattiwar, ²Dr. Bajarangi Lal Choudhary, ³Dr. Sonu Panwar, ⁴Dr. Dinesh Kumar**¹Associate Professor Department of Microbiology, Veerangana Avanti Bai Lodhi Autonomous State Medical College, Etah, Uttar Pradesh, India²Assistant Professor, Department of Microbiology, Hind Institute of Medical Sciences, Mau Ataria, Sitapur, Uttar Pradesh, India³Professor, Department of Microbiology, K. M. Medical College and Hospital, Mathura, Uttar Pradesh, India⁴Associate Professor, Department of Microbiology, K.M. Medical College and Hospital, Mathura, Uttar Pradesh, India**Corresponding Author:**Dr. Dinesh Kumar**Email:** dr.dineshkumarbhati@gmail.com

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Abstract:**Background:** For patients allergic to penicillin alternative drug is clindamycin which is preferred for mild to severe methicillin-resistant *Staphylococcus aureus* (MRSA) infections.**Objective:** To determine the clindamycin resistance by D-test to avoid treatment failure of the macrolide lincosamide group of drugs.**Materials and Methods:** Isolates were proceeded by bacteriological procedure, and antibiotic susceptibility, MRSA detection, and D-test were done by using the disk diffusion test as per clinical laboratory and standard guidelines (CLSI)**Results:** Of the total of 255 MRSA strains studied among them 101 (39.61%) were further preceded for D-test in which 80 (31.37%) isolates were positive. The resistance was obtained from pus 37 (46.25%) followed by Blood 31 (38.75%), and least from ET-secretion and sputum 6 (7.5%) and not resistant to vancomycin.**Conclusion:** The higher occurrence of inducible clindamycin resistance showed that treatment options were limited. So D-test should be checked to avoid treatment disappointment.**Keywords:** Clindamycin, D-test, Erythromycin, MRSA,**Introduction**

Staphylococcus aureus is a pluripotent pathogen that causes a variety of infection potentially fatal infections particularly in hospital environments due to its ability to develop drug resistance.[1] For individuals who are allergic to penicillins, the macrolide-lincosamides, and streptogramin B group of antibiotics is used as a substitute [2]. The antibiotics belonging to this group clindamycin is the suggested medication because of its better pharmacokinetic characteristics. However, the extensive usage of these antibiotics results in a rise in the prevalence of *Staphylococcus aureus* that are resistant to them [3].Resistance to these antibiotics has been associated with three mechanisms: target site alteration, antibiotic efflux, and drug modification [4]. An antibiotic sensitivity test of MRSA isolates which is positive for the D-test is necessary for effective treatment of infected patients. This is significant

because of the rise in antibiotic resistance and become difficulty in treatment [5]. Macrolide-resistant *Staphylococcus aureus* isolates may be clindamycin resistant either by default or by induction, or they may only be resistant to macrolide. Clindamycin and erythromycin discs are placed next to one another in disc diffusion tests, which identify inducible clindamycin resistance. To become the shape of the D-Zone adjacent to the erythromycin disk is an indication of induced clindamycin resistance [6]. Previous reports of *S. aureus* isolate D-test positive prevalence in cross-sectional study were range 7% to 94% [7, 8]. The infection due to D-test positive MRSA infection should not be treated with clindamycin. Inducible clindamycin resistance (ICR) MRSA isolates are sensitive to clindamycin but resistant to erythromycin, as routine tests. The D-test can be used to see if clindamycin may be used as a treatment option [9]. It has been suggested by Sedighi et al. [10] that D-test be utilized consistently in all microbiologic laboratories and that patients with infections produced due to inducible resistant *S. aureus* ought to stop clindamycin. Consequently, this research aimed to check if inducible clindamycin resistance was present, to assess any relationships that may exist between these two variables, and to evaluate how effective drugs work against MRSA.

Materials and Methods

Staphylococcus aureus was isolated and confirmed by gram stain, catalase, and coagulase test [11]. Every isolate performed antimicrobial susceptibility testing (AST) under CLSI criteria, utilizing Kirby Bauer's disc diffusion technique. Penicillin (10 units), cefoxitin (30 µg), erythromycin (15 µg), azithromycin (15 µg), tetracycline (30 µg), doxycycline (30 µg), levofloxacin (5 µg), norfloxacin (10 µg), ofloxacin (5 µg), nitrofurantoin (300 µg), clindamycin (2 µg), rifampicin (5 µg), linezolid (30 µg), moxifloxacin (5 µg), chloramphenicol (30 µg), and gentamicin were evaluated (10 µg) and the control strain was *Staphylococcus aureus* ATCC 25923. The D-test, which measures inducible clindamycin resistance, involves placing erythromycin (15 µg) and clindamycin (2 µg) discs at 15 mm. The zone of inhibition close to the erythromycin disc flattens, and this phenomenon is known as the D-test. Methicillin-resistant *S. aureus* (MRSA) bacteria were examined by disc diffusion technique with cefoxitin disc 30 µg/ml; resistance was defined as a zone of inhibition ≤ 21 mm. [12] and the bacterial colonies were cultured on MRSA Hi Crome agar and then after incubation the growth of green colonies considered as MRSA [13].

Results

1. Phenotypic test distribution (n=255)

Phenotypic test	Number	%
E-S, CD-R	10	3.92
E-S, CD-S	27	10.59
E-R, CD-R	117	45.88
E-R, CD-S (D- test) positive	80	31.37
E-R, CD-S (D- test) negative	21	8.24

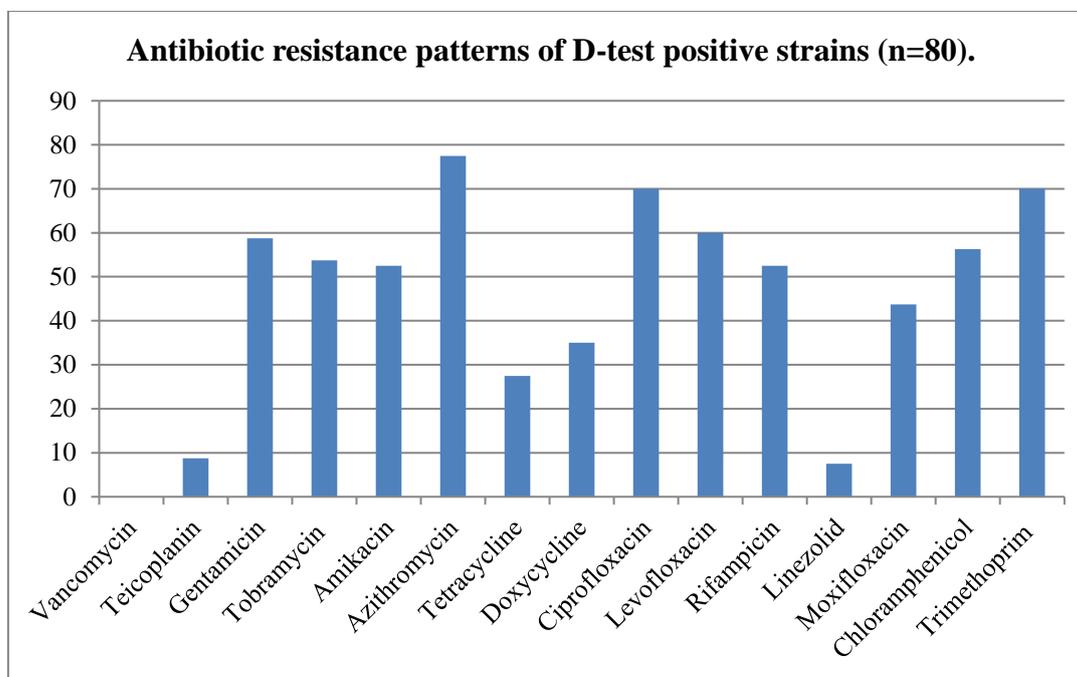
Of the total 255 MRSA isolates 101 (39.61%) were erythromycin resistant and clindamycin sensitive which were further tested for D-test in which 80 (31.37%) were found to be positive.

2. D- test positive specimen-wise distribution (n=80)

Specimen	Number	%
Blood	31	38.75

ET- secretion	06	7.5
Pus	37	46.25
Sputum	06	7.5

Of the total 80 (31.7%) inducible clindamycin resistance (D-test) isolates the highest result obtained was from pus 37 (46.25%) followed by Blood 31 (38.75%), least from ET-secretion and sputum 6 (7.5%).



The antibiotic resistance patterns of D-test positive showed no resistance to vancomycin followed by linezolid 6(7.5%) and teicoplanin 7(8.75%).

Discussion

Many nations have noticed an increase in attention to the use of MLSB antibiotics, specifically clindamycin, to treat *S. aureus* infections due to the increasing frequency of these infections and associated antimicrobial resistance[14]. In individuals allergic to penicillin drug clindamycin is an option for treating *Staphylococcus aureus* infections, particularly those affecting the skin and soft tissues. Treatment failures result from staphylococcal strains becoming more resistant to MLSB antibiotics, either constitutively or inducible, because of the extensive use of these antibiotics. The alteration of the binding site of clindamycin i. erm gene is the most known mechanism of clindamycin resistance. Routine antimicrobial susceptibility testing can identify these pathways by putting the clindamycin disc (D test) next to the erythromycin disc. [15-17]. The current research had a total of 255 MRSA strains and determined for phenotypic test for inducible clindamycin resistance (D-test) where results were found to be 101(39.61%) erythromycin resistance and clindamycin sensitivity which further proceeded for inducible clindamycin resistance (D-test) and found to be positive 80(31.37%). This study only included MRSA strains whereas other studies were done on Coagulase-negative *Staphylococcus* and methicillin-sensitive *Staphylococcus aureus*. Most of the studies supported higher results from MRSA 66% compared to MSSA 34% Patel et al. 54.54% MRSA and 13 of 35 (37.14%) MSSA isolates showed inducible clindamycin resistance [7]. Ghosh Soumya deep et al. [8] also show lower results compared to the present study Amira A. Kilany et al. [18] 5.1%, V Deotale, et al. 14.5% [19]. Of the total 80(31.37%) inducible clindamycin-resistant isolates on analysis of specimens-wise distribution found maximum isolates from pus 37(46.25%) followed by blood 31(38.75%), and least from ET-

secretion and sputum 6(7.5%). A similar study by Ghosh Soumyadeep et al. reported to exhibit the highest 58.82% and 16.67% [8]. isolates. This is like another study that showed the majority of strains were from wound infection 31.1% followed by bacteremia 27.3% and the least from respiratory tract infections 8.7% [20]. The antibiotic resistance patterns of D-test positive strains were not resistant to vancomycin and least to linezolid 6(7.5%), and teicoplanin 7(8.75%). The study by Adhikari RP et al. [21] also reported no resistance to vancomycin.

Conclusion

The higher prevalence of inducible clindamycin resistance showed that treatment options were limited. So, a D-test must be performed to check resistance and to avoid treatment failure. However, vancomycin remains the drug of choice.

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