International Journal of Current Advanced Research
ISSN: O: 2319-6475, ISSN: P: 2319-6505, Impact Factor: SJIF: 5.995
Available Online at www.journalijcar.org
Volume 6; Issue 10; October 2017; Page No. 7054-7056
DOI: http://dx.doi.org/10.24327/ijcar.2017.7056.1072

Research Article

DETECTION OF INDUCIBLE CLINDAMYCIN RESISTANCE AMONG CLINICAL ISOLATES OF ENTEROCOCCUS SPECIES

Dhinesh Kumar S[1] and Gopinath P[2]*

[1]BDS 2nd year, Saveetha Dental College, Chennai
[2]Senior Lecturer, Department of Microbiology, Saveetha Dental College, Chennai

ARTICLE INFO

Article History:

Received 26th July, 2017
Received in revised form 19th August, 2017 Accepted 25th September, 2017
Published online 28th October, 2017

Key words:

Enterococci, antimicrobial susceptibility test, inducible clindamycin resistance

ABSTRACT

Enterococci have transformed over the past century from being an intestinal commensal organism of little clinical importance to becoming the second most widespread nosocomial pathogen and is related with considerable mortality and morbidity. In this study a total of 20 different non-repetitive clinic isolates of Enterococci were collected from different clinical specimens and checked for inducible clindamycin resistance. Enterococcus isolates were subjected for susceptibility to erythromycin and other group of antibiotics by the Kirby-Bauer disc diffusion method. Of the 20 isolates, 18 (90%) of them were erythromycin resistance. The clinician must have a wide knowledge of inducible clindamycin resistance and report to laboratory immediately for prompt treatment. The D-test is a simple & reliable method to detect inducible & constitutive clindamycin resistance in routine clinical diagnosis setting.

INTRODUCTION

Enterococci have changed over the previous century from being an intestinal commensal life form of minimal clinical significance to turning into the second most boundless nosocomial pathogen and is connected with extensive mortality and horribleness. [1]

Among the distinctive types of Enterococci which have been recognized, Enterococcus faecalis, was the most widely recognized species related with the nosocomial contaminations, trailed by Enterococcus faecium. [2]

Clindamycin is utilized as a part of the treatment of skin and delicate tissue diseases, brought on by the staphylococcal and Enterococcal species. Great oral retention makes this medication a vital alternative in outpatient treatment or as a follow-up after intravenous treatment. Clindamycin is likewise utilized as an option for patients who are sensitive to penicillin. [3]

Imperviousness to macrolides (e.g. erythromycin) can happen by two unique instruments: efflux due to macrolide streptogramin resistance (msrA quality) and ribosome change because of erythromycin ribosome methylase (erm quality) [4]

Corresponding author:* **Gopinath P
Senior Lecturer, Department of Microbiology, Saveetha Dental College, Chennai

MATERIALS AND METHODS

Clinical isolates

A total of 20 different non-repetitive clinic isolates of Enterococci were collected from different clinical specimens were included in this study. These isolates were identified by standard biochemical parameters as described by elsewhere. Isolates were preserved in semi-solid brain heart infusion medium and stored at 4ºC until further use.

Antimicrobial susceptibility test

Antibiotic susceptibility test was determined for these strains to routinely used antibiotics such as ampicillin (10µ), vancomycin (30µ), teicoplanin (30µ), erythromycin (15µ), ciprofloxacin (5µ), amikacin (200µ), gentamycin (10µ), tetracycline (30µ) and linezolid (30µ) (Hi Media, Mumbai) by kirby-bauer disc diffusion method. [5]

Detection of inducible clindamycin resistance

Isolates which were resistant to erythromycin were further subjected to 'D test' as per CLSI guidelines. Enterococcus isolates were made into suspension and turbidity has been matched with 0.5 McFarland standard. These bacterial suspension were lawn cultured on Mueller Hinton agar (MHA). After a brief drying erythromycin (15 mcg) disc was placed at a distance of 15mm (edge to edge) from clindamycin (2 mcg) disc and was incubated at 37 ^{0}C overnight. Flattening of zone (D shaped) around clindamycin in the area between the two discs, indicated inducible clindamycin resistance. Three different phenotypes were appreciated after testing and

interpreted as follows:

1. MS Phenotype - Staphylococcal isolates exhibiting resistance to erythromycin (zone size ≤13mm) while sensitive to clindamycin (zone size ≥21mm) and giving circular zone of inhibition around clindamycin was labelled as MS phenotype.
2. Inducible MLS $_B$ Phenotype - Staphylococcal isolates showing resistance to erythromycin (zone size ≤13mm) while being sensitive to clindamycin (zone size ≥21mm) and giving D shaped zone of inhibition around clindamycin with flattening towards erythromycin disc were labelled as having this phenotype.
3. Constitutive MLSB Phenotype - this phenotype was labelled for those Enterococcal isolates which showed resistance to both erythromycin (zone size ≤13mm) and clindamycin (zone size ≤14mm) with circular shape of zone of inhibition if any around clindamycin. [6]

RESULTS

Sample wise distribution of clinical isolates of Enterococci:
We have isolated Enterococcus from different clinical specimens such as urine (60%), blood (20%), stool and wound swab (10%)

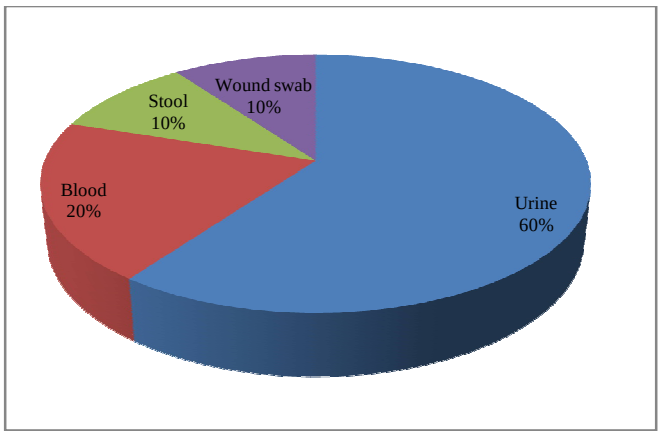

Fig 1 Pie chart showing the sample wise distribution of clinical isolates of Enterococcus spp

Bacterial isolates

In our isolates, 70% were identified as E. faecalis and 30% of them were E. faecium

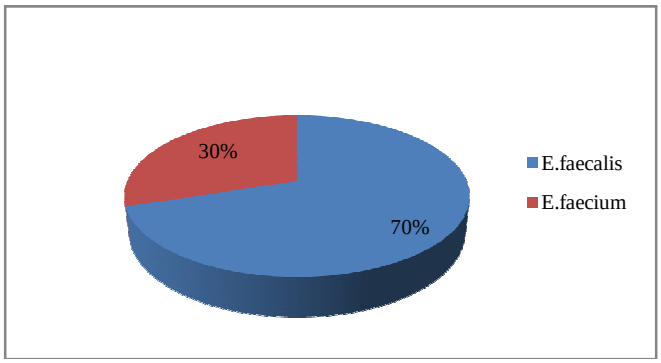

Fig 2 Pie chart showing species distribution of Enterococcus

Antibiotic susceptibility testing

Increased percentage of isolates were shown to be resistant to most of the drugs used. Wherein, 90% were resistant to erythromycin as well as to amikacin, 85% of isolates were resistant to ampicillin, 80% isolates were to gentamycin. The detailed result of antibiotic susceptibility pattern was shown in table 1.

Table 1 Results of antibiotic sensitivity pattern of Enterococci

Antibiotics	Sensitivity	Intermediate	Resistance
Ampicillin	1(5%)	2(10%)	17(85%)
Vancomycin	15(75%)	1(5%)	4(20%)
Teicoplanin	12(60%)	3(15%)	5(25%)
Erythromycin	2(10%)	0	18(90%)
Ciprofloxacin	6(30%)	0	14(70%)
Amikacin	1(5%)	1(5%)	18(90%)
Gentamycin	2(10%)	2(10%)	16(80%)
Tetracycline	4(20%)	4(20%)	12(60%)
Linezolid	18(90%)	1(5%)	1(5%)

Results of inducible clindamycin resistance

Enterococcus isolates were subjected for susceptibility to erythromycin and other group of antibiotics by the Kirby-Bauer disc diffusion method. Of the 20 isolates, 18 (90%) of them were erythromycin resistance. Results of D-test were projected in Table 2.

Table 2 Showing results of clindamycin resistance among Enterococcus isolates

Clindamycin resistance	Total (n=20)
ERY-S, CL-S	7 (35%)
ERY-R, CL-R	5 (25%)
ERY-R, CL-S(D-test positive, iMLS)	4 (20%)
ERY-S, CL-R (D-test negative, MS)	4 (20%)

DISCUSSION

Medication vulnerability information of the infecting organism is a fundamental figure settling on fitting helpful choices. The variety of components, which give imperviousness to MLS anti-microbials, mirrors the unpredictability of the safe phenotypes and additionally the clinical circumstance. The most boundless and clinically critical resistance components experienced with Gram-positive life forms are the creation of methylases and efflux proteins. The clinical disappointment of clindamycin treatment has been accounted for some time recently. [7,8,9] Hence, there is a need to distinguish the instruments that present imperviousness to MLS anti-infection agents concerning clindamycin treatment of staphylococcal contaminations.

The development of imperviousness to various anti-infection agents among Gram-positive cocci has left not very many helpful choices for clinicians. Despite the fact that half of our detaches were safe phenotypes, the other half were touchy to clindamycin, against which it would be sheltered and suitable to utilize clindamycin or different macrolides. In this manner, utilizing as a part of vitro erythromycin resistance as a surrogate marker for every one of the MLS anti-toxins and consequently dodging them as a treatment choice, would be unseemly. A helpful choice is unrealistic without the significant anti-toxin vulnerability information. This is the place the D-test gets to be distinctly huge.

Study conducted by Ciraj in 2009, 38% of strains were MLSBi phenotype and 15.3% were MLS B c phenotype among the MRSA strains. Results in regard to the MSSA group are similar to the studies from Turkey, where no constitutive resistance was reported and inducible resistance to clindamycin was approximately 11%. In contradiction, 20% isolates were ERY-R, CL-S (D test positive) whereas, 20% were negative for the same. [10]

CONCLUSION

The clinician must have a wide knowledge of inducible clindamycin resistance and report to laboratory immediately for prompt treatment. The D-test is a simple & reliable method to detect inducible & constitutive clindamycin resistance in routine clinical diagnosis setting.

References

1. Leclercq R. Mechanisms of resistance to macrolides and lincosamides: Nature of the resistance elements and their clinical implications. *Clin Infect Dis* 2002; 34:482-92.
2. Weisblum B. Erythromycin resistance by ribosome modification. *Antimicrob Agents Chemother* 1995; 39:577-85.
3. Roberts MC, Sutcliffe J, Courvalin P, Jensen LB, Rood J, Seppala H. Nomenclature for macrolide and macrolide-lincosamide-streptogramin B resistance determinants. *Antimicrob Agents Chemother* 1999; 43:2823-30.
4. 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; 41:4740-4.
5. Clinical and laboratory standards institute. Performance standards for antimicrobial susceptibility testing; seventeenth informational supplement. *Clinical Laboratory Standards Institute.* 2015;Vol.2(No.1)
6. Kloos WE, Banerman TL. Staphylococcus and Micrococcus. In: Murray PR, Baron EJ, Pfaller MA, Tenover FC, Yolken RH, editors. Chapter 22. Manual of clinical microbiology. Washington DC: ASM Press; 1999. pp. 264-82.
7. Watanakunakorn C. Clindamycin therapy of *Staphylococcus aureus* endocarditis: Clinical relapse and development of resistance to clindamycin, lincomycin and erythromycin. *Am J Med* 1976; 60:419-25.
8. Drinkovic D, Fuller ER, Shore KP, Holland DJ, Ellis-Pegler R. Clindamycin treatment of *Staphylococcus aureus* expressing inducible clindamycin resistance. *J Antimicrob Chemother* 2001; 48:315-6.
9. 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; 37:1257-60.
10. Ciraj AM, Vinod P, Sreejith G, Rajani K. Inducible clindamycin resistance among clinical isolates of staphycocci. *Indian Journal Of Pathology and Microbiology* 2009:52:49-51

How to cite this article:

Dhinesh Kumar S and Gopinath P (2017) 'Detection of Inducible Clindamycin Resistance Among Clinical Isolates of Enterococcus Species', *International Journal of Current Advanced Research,* 06(10), pp. 7054-7056.
DOI: http://dx.doi.org/10.24327/ijcar.2017.7056.1072

****