INTRODUCTION

Infections have been one of the major causes of mortality and morbidity worldwide among human population. All microorganisms namely Bacteria, Virus, Parasites and fungi caused variety of infections affecting every organ of the body. The role of microbiologists is, early identification of responsible pathogen and reporting with antibiotic susceptibility testing for speedy recovery of patients.

The staphylococci are important bacterial pathogens that can infect both animals and human and are responsible for numerous hospital and community acquired infections yearly. It is estimated that 30% of human population are long term carriers of S.aureus which can be found as part of the normal skin flora and in anterior nares of the nasal passages. S.aureus is the most common species to cause infections, ranging from minor illness like impetigo, boils, cellulitis, folliculitis, carbuncles, scalded skin syndrome and abscesses to life threatening infections such as pneumonia, meningitis, osteomyelitis, endocarditis, toxic stock syndrome, bacteremia and sepsis. Staphylococcal infections result in a significant burden both economically and clinically due to several factors including increasing antibiotic resistance and lack of effective vaccines.

Antimicrobial Resistance

Wide spread misuse of antimicrobial agents is one of the important factors for favouring the emergence of resistant bacterial strains. Genetic variability is essential for the development of microbial resistance which occurs through variable mechanisms such as 1. Point mutation occurring in the target site of antimicrobial agents. 2. Large scale rearrangements of the bacterial genome generated by integrons,
transposons or insertion sequences. 3. Acquisition of foreign DNA by plasmids, Bacteriophages or transposable genetic elements (17). This inheritance of foreign DNA contributes to organism’s genetic variability and its capacity to respond to the selection pressure imposed by the antimicrobial agents. Resistance is also mediated by microbial enzymes such as beta lactamase which inactivate the therapeutic agents.

Following resistance to penicillin, Methicillin, a betalactam antibiotic variant of penicillin class was introduced in 1959 by Beecham(9). Very soon after its discovery, Methicillin resistance to S.aureus (MRSA) was identified in 1961(31) and has been showing a increasing trend since that time. MRSA is prevalent world wide and varies from place to place ranging from 2 to 70%, except in Netherland the prevalence is very low < 0.5%. In India, the prevalence of MRSA is estimated to be 30 to 70%(3). Methicillin resistance in S.aureus is mediated by mec A gene which codes for a modified penicillin binding protein 2a (PBP-2a)(18). This resistance can be constitutive or inducible. Mec A gene is carried on mobile genetic elements, staphyloccocal cassette chromosome (SCC mec). The increasing prevalence of MRSA is a therapeutic threat and Clindamycin is an attractive therapeutic alternative to Vancomycin.

Clindamycin, the lincosamide antimicrobial is a frequent therapeutic option for staphyloccocal infections, particularly for skin, soft tissue infections as an alternative to penicillin allergic patients and infections in the abdomen. Pelvis and lung involving anaerobes. It has excellent tissue penetration except for the CNS,(31) The drug accumulates in abscess and no renal dosing adjustments needed. Clindamycin inhibit bacterial protein synthesis. Major target is the site of peptide bond formation in the 23s ribosomal RNA of the 50s ribosomal (19).

Clindamycin resistance is common among health care associated MRSA strains. Most Community acquired-MRSA remain susceptible todate, but resistance rates vary by region(20).Macrolide inducible resistance to clindamycin was first recognized in the laboratory in early 1960s (3). Clinical isolates resistant to clindamycin were first recognized in 1968 (22). Relapse of S.aureus infection in rabbit model of endocarditis during clindamycin therapy was observed in early 1970s(10) Clinical and bacteriologic relapse in a patient with S.aureus endocarditis during the fourth week of clindamycin therapy after initial improvement, was reported in 1976(33). The initial isolate was susceptible to clindamycin and erythromycin while that from the relapse was resistant to both. This led to avoidance of clindamycin for treatment of endocarditis. Erythromycin was introduced in 1952 as the first macrolide antibiotic. Within a year of its introduction, Erythromycin resistant Staphyloccoci from US, Japan, and Europe were described(21). A triple disc diffusion test using a Telithromycin disc along with Erythromycin and Clindamycin discs has been introduced(6). Telithromycin belongs to the recently developed Ketolides, a class of antibiotic belonging to the MLSb family with certain structural differences from macrolides(7).

Macrolide lincosamide streptogramin B (MLSb) antibiotics comprise separate classes of molecules that bind to the ribosome and inhibit protein synthesis. Resistance occurs by any of the mechanisms, modification of bacterial drug target, modification and inactivation of drug itself and decreasing intracellular accumulation of the drug. Staphyloccal resistance to clindamycin may be inducible (iMLSb) or constitutive. It is noted that patients harbouring iMLSb staphylococci with clindamycin leads to the development of constitutive resistance, subsequently leading to therapeutic failure. The erm gene determinants belongs to a family of methylase genes preferentially located on mobile elements such as transposons (ermA) or plasmids (ermc). An additional factor in S.aureus is that expression of erm is inducible. Among MLSb drugs only macrolides are good erm inducers (19).

Aims and Objectives

1. To study the prevalence of inducible Clindamycin resistance among Staphylococcus species isolated from clinical samples of inpatients, from Govt. Rajajihospital (GRH), Madurai.
2. To determine the distribution of inducible Clindamycin resistance among MRSA, MSSA and CoNS.
3. To compare the phenotypic methods in detection of inducible Clindamycin resistance with genotypic method (PCR).

MATERIALS AND METHOD

This prospective study was conducted in patients admitted to Government Rajaji Hospital, attached to Madurai Medical College, Madurai. The study was conducted between December 2012 to August 2013. Institutional Ethical committee clearance has been obtained and written informed consent was received from the patients or parents in case of pediatric patients before collecting the specimens. A total of 350 clinical samples that include Pus, Blood, Sputum, Throat swab and wound swab were collected from the patients who were admitted to various Clinical departments of Government Rajaji Hospital, Madurai.

Inclusion criteria

- Males and females of all age groups were included.
- Patients with provisional diagnosis of active infections like boils, folliculitis, cellulitis, abscesses, and osteomyelitis.
- Patients with Post-operative wound infections.
- Patients with diagnosis of bacteremia and pneumonia.
- Patients affected with burn wounds, non – healing ulcer.
- Diabetic patients with ulcer.

Exclusion criteria

Patients suffering from infections of Genitals, Eye, Ear, CNS and urinary tract infections were excluded.

Identification of Inducible Clindamycin Resistance By Phenotypic Methods

Both MRSA & MSSA and the Coagulate negative Staphylococcus subjected to D- test to identify the inducible Clindamycin resistance. Isolates showing Erythromycin resistance, zone size ≤13mm and Clindamycin susceptible, zone size≥21mm are subjected to D-test.
**D-TEST**

After making a lawn culture of the isolated Staphylococcus in MHA plates, Erythromycin 15µg and Clindamycin 2µg discs were spaced 1526mm edge to edge interdiskdistance, higher sensitivity reported with 15mm interdiskdistance. The plates were incubated at 37°C for 16-18hrs.

**Interpretation**

**MS Phenotype:** Staphylococcal isolates showing resistance to Erythromycin, zone size ≤13mm and sensitive to Clindamycin, zone size ≥21mm and giving circular zone of inhibition was labeled as having this phenotype.

**Inducible MLS\(_b\) (iMLS\(_b\)) Phenotype:** Staphylococcal isolates showing resistance to Erythromycin and sensitive to Clindamycin, giving D-shaped zone of inhibition around Clindamycin with flattening towards Erythromycin disc was labeled as having this phenotype.

**Constitutive MLS\(_b\) Phenotype:** this phenotype was labeled for those Staphylococcal isolates, showing resistance to both Erythromycin (zone size ≤13mm) and Clindamycin (zone size≤14mm).

**Quality control strains**

Control strains used for disc diffusion test were, Staphylococcus aureus ATCC 25923 and in house strains of Staphylococcus aureus showing D-test positive repeatedly was used as positive control for inducible Clindamycin resistance.

**Broth Microdilution**

Cationically adjusted Mueller Hinton broth(CAMHB) was taken in four test tubes, containing 4µg/ml Erythromycin and 0.5µg/ml Clindamycin in one first tube, followed by 4µg/ml Erythromycin in second tube and 0.5µg/ml Clindamycin in third tube and last tube with MHB alone as control. The test inoculum prepared by standard recommendations were added to the four tubes and incubated at 37°C for 18-24hrs.

**Interpretation**

Any growth in first and second tubes, with no growth in third tube – indicates inducible Clindamycin resistance.

**Agar Dilution**

**Preparation of media**

Mueller Hinton agar was prepared in tubes and autoclaved. It was then allowed to cool in a 50°C water bath. Dilution of Erythromycin was prepared in sterile distilled water to give a final concentration of 1mg/litre and Clindamycin was diluted with sterile saline to give a final concentration of 0.5mg/litre. After adding the 1ml of appropriate dilution of drug to the 14ml of medium at 50°C it was mixed well and poured in sterile petridishes. MHA plates containing 1ml of both Erythromycin and Clindamycin was prepared. A control plate containing the test medium without antibiotic was also prepared. The media was used immediately otherwise potency of drugs would be affected. Upto 9-12 different strains can be inoculated in a single plate.

**Inoculum preparation**

At least 3-5 well isolated colonies of the similar morphology were selected from an agar plate. Top of each colony was touched with a loop and then transferred into a tube containing 4-5 ml of peptone water and incubation was done at 37°C until reaches 0.5Mc Farland’s standard (usually 2-6 hrs). This results in growth corresponding to 150 million organisms/ml.

**Inoculation of test plates**

All the four plates were divided into 9 divisions.10µl of inoculum was put into appropriate quadrant and incubation was done at 37°C for 16-18hrs.

**Interpretation**

A test was deemed to be positive (i.e., inducible clindamycin resistance was present) if there was any visible growth on Erythromycin only and combined plates, but not on the Clindamycin only plate. A test was deemed to be negative (i.e, MS resistance phenotype) if growth was found on the Erythromycin only plate but not on the combined or Clindamycin only plate.

Detection of inducible clindamycin resistance in staphylococcus species by genotypic method – polymerase chain reaction (PCR) erm a / erm c gene assay

**Interpretation :** The presence of ermA and erm C gene was indicated by the amplification of 196 & 295 bp PCR product from the clinical isolates respectively.

The various phenotypic methods were compared with PCR and sensitivity, specificity, PPV and NPV were determined. The P value and 95% confidence intervals were obtained using SPSS (Statistical Package for Social Sciences) version 16. P value <0.05 was taken as significant and P value >0.05 indicates that there is no significant difference between the various tests compared with PCR.

**RESULTS**

A total of 350 clinical samples of Pus, Blood, Sputum, Throat swab and wound swab collected from the patients admitted to various clinical departments of Government Rajaji Hospital, Madurai were processed. Out of 350 clinical samples, 181 samples were Pus, 75 samples were blood, 32 samples were sputum, 40 samples were wound swab and 22 samples were Throat swab. 48 samples showed no growth and 302 showed growth. Among the 350 isolates, 186 were staphylococci, 5 were streptococci, 111 were Gram Negative Bacilli. From the above observation it is inferred that staphylococci species were the common isolates from all the specimens collected. Among the 186 Staphylococca I isolates, 101 were from Pus, 40 from blood, 21 from wound swab, 14 from sputum and 10 from throat swab. From the observation, it is inferred that more number of Staphylococcus species were isolated from Pus samples, followed by blood. Out of 186 isolates, 91 were isolated from males and 95 were isolated from females.

Among the 186 Staphylococcus species isolated, 132 were S.aureus and 54 were Coagulase negative Staphylococci, it is inferred that S. aureus is the most common isolate among the staphylococcus species.
Among the 186 Staphylococcus species, from 101 pus samples 59 were S.aureus & 42 were CoNS, from 40 blood samples, 32 were S. aureus & 8 were CoNS, from 21 wound swab, 17 were S. aureus and 4 were CoNS.Among the 132 S.aureus isolates, 62 were (46.96%), Methicillin Resistant S. aureus (MRSA) and 70 were (53.03%) Methicillin Sensitive S. aureus (MSSA).

The sample wise distribution of MRSA results were analysed which showed highest percentage of MRSA isolate in Pus samples(27.27%) followed by blood (10.60%), Wound swab (5.30%) and sputum & throat swab 2.27% & 1.51% respectively.

**Table 1** Comparison of different types of MLSB resistance among Staphylococcus on D-zone test

<table>
<thead>
<tr>
<th>Phenotype</th>
<th>MRSA N=62</th>
<th>MSSA N=70</th>
<th>CoNS N=54</th>
<th>Total 186</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inducible Clindamycin resistance</td>
<td>28-45.16%</td>
<td>19-27.14%</td>
<td>9 (16.66%)</td>
<td>56-30.10%</td>
</tr>
<tr>
<td>Constitutive Clindamycin resistance</td>
<td>10-16.12%</td>
<td>4 (5.71%)</td>
<td>2 (3.70%)</td>
<td>16-8.60%</td>
</tr>
<tr>
<td>MS Phenotype D test negative Susceptible to Ery &amp; clindamycin</td>
<td>16-25.80%</td>
<td>14-20.0%</td>
<td>5 (9.25%)</td>
<td>35-18.81%</td>
</tr>
<tr>
<td>Total</td>
<td>8 (12.90%)</td>
<td>33-47.14%</td>
<td>38-70.37%</td>
<td>79-42.47%</td>
</tr>
</tbody>
</table>

The Comparison of different types of MLSB resistance among Staphylococcal species on D – test, showed that inducible Clindamycin resistance was high among MRSA (45.16%), followed by MSSA (27.14%) and CoNS (16.66%). The Constitutive resistance was also high in MRSA (16.12%). Among the 186 Staphylococcus species 30.10% showed inducible Clindamycin resistance.

The specimen wisedistribution of the inducibleClindamycin resistance were analysed. The inducible Clindamycin resistance positive isolates in pus were 30, 14 from blood, followed by 8 in wound swab and 4 in sputum samples

The sex distribution of inducible Clindamycin resistance in Staphylococcal species was analysed, 32 isolates were from male patients and 24 isolates were from female patients.

The department wise isolation of inducible Clindamycin resistance among Staphylococcus species was analysed, 18 were from surgery, followed by 14 from Medicine, 10 from Ortho, 7 from OG and 7 from Paediatric wards.

**Table 2** Detection of inducible Clindamycin Resistance by various Phenotypic methods, n = 91.

<table>
<thead>
<tr>
<th>Method</th>
<th>No. of isolate</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>D test</td>
<td>56</td>
<td>61.53%</td>
</tr>
<tr>
<td>Broth micro dilution</td>
<td>48</td>
<td>52.74%</td>
</tr>
<tr>
<td>Agar dilution</td>
<td>51</td>
<td>56.04%</td>
</tr>
</tbody>
</table>

Out of the 91 isolates which were screening positive, D-test detected 56 isolates, followed by Agar dilution which detected 51 isolates and Broth microdilution detected 48 isolates as inducible Clindamycin resistance producers.

**Table 3** Inducible Clindamycin resistance detection by Genotypic Method, n = 91

<table>
<thead>
<tr>
<th>Gene</th>
<th>ermA / mC</th>
<th>Total positive</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.of isolates</td>
<td>60</td>
<td>65.93%</td>
</tr>
</tbody>
</table>

Amidst the 91 isolates which were processed for genotypic studies, 60 isolates showed the presence of ermA/ermC genes responsible for inducible Clindamycin resistance. No gene was detected in the remaining 31 isolates.

**Table 4** Comparison of phenotypic methods with PCR

Comparison of D – Test with Pcr n = 91

<table>
<thead>
<tr>
<th>Pcr</th>
<th>D test</th>
<th>True positive</th>
<th>False negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>56</td>
<td>4</td>
<td></td>
</tr>
<tr>
<td>Negative</td>
<td>1</td>
<td>0</td>
<td>30</td>
</tr>
</tbody>
</table>

Comparison of Agar Dilution With PCR n = 91

<table>
<thead>
<tr>
<th>Pcr</th>
<th>Agar dilution</th>
<th>True positive</th>
<th>False negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>51</td>
<td>9</td>
<td></td>
</tr>
<tr>
<td>Negative</td>
<td>2</td>
<td>29</td>
<td></td>
</tr>
</tbody>
</table>

Comparison of Broth Microdilution With PCR n = 91

<table>
<thead>
<tr>
<th>Pcr</th>
<th>Broth micro dilution</th>
<th>True positive</th>
<th>False negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>40</td>
<td>12</td>
<td></td>
</tr>
<tr>
<td>Negative</td>
<td>4</td>
<td>27</td>
<td></td>
</tr>
</tbody>
</table>

It was found that D – test method showed only 4 false negatives and 1 false positive, thus close to PCR in identifying True positives. The Agar dilution method showed 9 false negatives and 2 false positives. Broth micro dilution method showed 12 false negatives and 4 false positives.

**Table 5** Sensitivity and specificity of various phenotypic methods

<table>
<thead>
<tr>
<th>Methods</th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>PPV</th>
<th>NPV</th>
</tr>
</thead>
<tbody>
<tr>
<td>D test</td>
<td>93.33%</td>
<td>96.77%</td>
<td>98.2%</td>
<td>88.2%</td>
</tr>
<tr>
<td>Agar dilution</td>
<td>85%</td>
<td>93.54%</td>
<td>96.22%</td>
<td>76.31%</td>
</tr>
<tr>
<td>Broth microdilution</td>
<td>80%</td>
<td>87.09%</td>
<td>92.3%</td>
<td>69.2%</td>
</tr>
</tbody>
</table>

From the above table it is inferred that, D – test method has the highest sensitivity (93.33%) and specificity (96.77%) and the PPV and NPV are 98.2% and 88.2% respectively. Broth microdilution method has the least sensitivity (80%) and specificity (87.09%) with the PPV and NPV of 92.3% and 69.2% respectively. Agar dilution method showed sensitivity (85%) and specificity (93.54%) with PPV 96.22% and NPV 76.31%. Since P value in all the tests were not < 0.05, it was inferred that there was no statistically significant difference between PCR and other phenotypic methods in the detection of inducible Clindamycin resistance.

**DISCUSSION**

Staphylococcus are one of the leading causes of numerous hospital and Community acquired infections yearly. The resistance exhibited by Staphylococci to a wide range of antibiotics imposes a serious therapeutic problem. Clindamycin, the lincosamide antimicrobial is a frequent therapeutic option for Staphylococcal infections, particularly for skin and soft tissue infections as an alternative to Penicillin allergic patients. The Prevalence of inducible...
Clindamycin resistance among MRSA is of serious concern in the recent years worldwide. Treatment of patients, with inducible Clindamycin resistance strains will lead to therapeutic failure and also develop Constitutive resistance\(^{(10)}\). The incidence of Clindamycin resistance varies by geographical area and therefore local statistics are crucial to guide empiric therapy\(^{(30)}\). The possible variation in the prevalence of Constitutive, inducible Clindamycin resistance and MS Phenotype could be explained due to difficulty in bacterial susceptibility in different geographical areas and also due to varying antimicrobial subscribing patterns of Physicians. Hence early detection of inducible Clindamycin resistance is of great clinical significance in combating the resistance. In view of this, the present study was taken to identify a simple and reproducible screening method to detect inducible Clindamycin resistance in this institution.

A total of 350 samples were processed in the present study, the Predominant isolate among all the isolates were Staphylococci and out of 350 samples, 186 (53.14%) were Staphylococci, among the 186 Staphylococci, more number of isolates 101(54.30%) were isolated from Pus samples, followed by Blood & Wound swab. It was also inferred that more number of Staphylococci isolates were isolated from 16-40 years (49.46%) age, followed by 41-60 years (25.80%). Analysis of sex wise distribution showed that most of staphylococcal infections were associated with females (51.07%) than males (48.92%). The present study detected 49.96% MRSA. Pulimood & Lalitha (1993)\(^{(26)}\) in their study showed 24 % MRSA, Rajaduraiapandi et al (2001)\(^{27}\) documented 37.9 % MRSA and Anuparba et al (2006)\(^{(1)}\) documented 54.8 %, Van biekm et al\(^{(32)}\) explained that the incidence of MRSA was steadily increasing since its emergence in 1960s. The increased trend may be due to the adverse use of antibiotics resulting in faulty genetic background or poor infection control practices in the environment. The comparison of different types of MLS\(_B\) resistance among Staphylococcus species on D-test in the present study showed inducible Clindamycin resistance was high among MRSA (45.16%), followed by MSSA (27.14%) and CoNS (16.66%). Earlier study in 2004 by Navaneeth et al\(^{24}\) documented 19% inducible Clindamycin resistance in MRSA and 19.2% in MSSA. In 2008 Ajantha GS et al\(^{(2)}\) observed 74% of MRSA and 45% of MSSA isolates as D-test Positive. Similarly in study conducted by Matthew V. N. O’ Sullivan et al (2005)\(^{(23)}\) reported 27.12% in MRSA and 9.5% in MSSA isolates. The present study results were in concordance with few studies described above. On contrary, schreckenberger et al\(^{(29)}\) and Levin TP et al\(^{(15)}\) have shown a higher percentage of inducible Clindamycin resistance in MSSA as compared to MRSA. Different studies from different parts of India have reported that 20% to 64% of their MRSA strains were of the iMLS\(_B\) Phenotype. Various studies in India showing prevalence of Inducible Clindamycin Resistance in Staphylococcus aureus isolates.

<table>
<thead>
<tr>
<th>Author’s name</th>
<th>Constitutive resistant</th>
<th>iMLS(_B) pheno</th>
<th>Ms Phen</th>
<th>Constitutive resistant</th>
<th>iMLS(_B)</th>
<th>Ms Phen</th>
</tr>
</thead>
</table>

Among the 54 CoNS isolated, 9 (16.66%) isolates showed inducible Clindamycin resistance, 2 (3.70%) isolates constitutive resistance, 5 (9.25%) isolates demonstrated MS phenotype and 38 (70.37%) isolates were susceptible to both Erythromycin and Clindamycin. According to Manish Mane et al\(^{(21)}\) 24% had iMLS\(_B\) Phenotype, followed by 76% of isolates sensitive to both Erythromycin and Clindamycin and no isolate demonstrated constitutive resistance and MS phenotype. Lim et al\(^{(16)}\), reported 9.6% iMLS\(_B\) in CoNS whereas Schmitz et al, reported a high incidence of both constitutive resistance (69%) and (30%) inducible resistance. Hamilton et al\(^{(13)}\) reported higher incidence of inducible resistance than constitutive resistance which was concordance with our study. Ciraj et al, documented an incidence of 6.3% of inducible Clindamycin resistance in CoNS isolates, which is significantly lower than our results.

The Specimen wise analysis of inducible Clindamycin resistance showed increased incidence among the Pus samples (16.12%) the associated major risk factors are wound infection, abscesses, Burns, non healing ulcers. The Blood samples showed 7.52%, sputum 2.15%,wound swab 4.30% inducible Clindamycin resistance. The sex analysis showed increased incidence in Males (57.14%) than Females (42.85%). Various phenotypic methods like D-test, Broth microdilution, Agar dilution were done to detect then inducible Clindamycin resistance. Among all the method D-test detected 61.53% of inducible Clindamycin resistance, followed by Agar dilution 56.04% and Broth microdilution detected 52.74%. Genotypic studies were carried out to detect the ermA / ermC responsible for inducible Clindamycin resistance. Among the 91 isolates processed for PCR, 60 (65.93%) isolates demonstrated the ermA/ermC genes. No gene was detected in the remaining 31 (34.07%) isolates. In this study, the phenotypic methods like D-test, Broth microdilution, Agar dilution were compared with PCR and found that among the phenotypic methods, D-test showed highest sensitivity (93.33%) and specificity (96.77%) with the Positive Predictive Value (PPV) and Negative Predictive Value (NPV) of 98.2% and 88.2% respectively. Among the 3 phenotypic methods, Broth microdilution has the least sensitivity (80%) and specificity (87.09%) with PPV and NPV of 92.3% and 69.2% respectively. Clarence J Fernades et al\(^{(10)}\) documented 100% sensitivity and specificity in Agar dilution, while Christian Lavalle et al\(^{(5)}\) reported 100% sensitivity and 97% specificity, which is significantly higher than our results. The present study showed 85% sensitivity and 93.5% specificity with 96.27% PPV and 76.31% NPV. Since P value in all the tests were not < 0.05, it was inferred that there was no Statistically significant difference between
PCR and other phenotypic methods in the detection of inducible Clindamycin resistance.

The isolates of Staphylococcus Species. that are erythromycin resistant but Clindamycin susceptible should not be reported as Clindamycin susceptible Unless iMLS\textsubscript{B} resistance has been excluded. The only method currently recommended for testing of iMLS\textsubscript{B} resistance is disk approximation testing. D - test is a simple, cost effective and easiest method with sensitivity very close to PCR in detecting all the inducible Clindamycin resistance positive isolates. PCR is the gold standard test, but this could not be adopted as a routine method in all Laboratories, since it requires costlier, sophisticated equipments and trained personnel to do PCR. The D – test can be done as a routine test in all Microbiological Laboratories and hence guiding the clinicians regarding the judicious use of Clindamycin.

- The study showed a preponderance of 54.57% Gram positive infections among various samples and out of which 53.1% were Staphylococcal species.
- Among the Staphylococcal species, 70.96% were S.aureus and 29.03% were CoNS. 46.96% were MRSA, 27.27% MRSA was isolated from Pus.
- 30.10% showed inducible Clindamycin resistance among all isolated Staphylococcal species, of which MRSA showed 45.16% iMLS\textsubscript{B} and 16.12% constitutive resistance, followed by CoNS 16.66% iMLS\textsubscript{B} and 3.70% constitutive resistance.
- 16.12% of inducible Clindamycin resistance isolates were Pus, associated with risk factors like wound infection, abscesses, burns, non healing ulcers.
- 57.14% Males and 42.85% females reported with inducible Clindamycin resistance, showing Male predominance.
- Various phenotypic methods were used for detection of inducible Clindamycin resistance and compared with PCR (ermA/ermC) as gold standard.
- D – Test detected increased number of inducible Clindamycin resistance (61.53%) than other methods like Agar dilution (56.04%) and Broth microdilution (52.74%).

CONCLUSION
Staphylococcus is notorious for causing a wide range of Hospital and Community acquired infections and because of emergence of resistance to multiple antibiotics the therapeutic options available for Clinicians are limited. In this study, the inducible Clindamycin resistance in Staphylococcal species have been emerging predominately in Pus samples from Surgical wards at Govt. Rajaji Hospital, Madurai with a prevalence of 30.10% with a higher predilection in Males.

MRSA showed 45.16% iMLS\textsubscript{B} phenotypes.

Early detection of these strains are crucial to establish an appropriate antimicrobial therapy and thereby reducing the mortality and morbidity associated with these infections.
Bibliography
