Original article

Clinically significant coagulase negative staphylococci speciation and their antibiogram patterns in tertiary care hospital

Pavithra DP1, Divya P*2

1Assistant Professor, Department of Microbiology, Bangalore Medical College & Research Institute, Bangalore, Karnataka.
2Assistant Professor, Department of Microbiology, Vijayanagar Institute of Medical Sciences, Ballari, Karnataka.
*Corresponding author: Dr. Divya P, Assistant Professor, Department of Microbiology, Vijayanagar Institute of Medical Sciences, Cantonment, Ballari-583104, Karnataka, India.

Abstract

Background and objectives: Coagulase-negative Staphylococci (CONS), which are the normal flora of skin and mucous membrane, have emerged as predominant pathogens in hospital-acquired infections. The infections are difficult to treat because of the risk factors and the multiple drug resistant nature of the organisms.

Materials and methods: Study was carried for period of 6 months. Clinical samples were collected from different sites were subjected to biochemical characterization and antimicrobial susceptibility test were done by using Disc diffusion method using Mueller–Hinton agar plates.

Results: Out of eighty four isolates Staphylococcus epidermidis 35 (41.6%) was most frequently isolated, followed by S. saprophyticus 24 (28.6%), S. hemolyticus 17 (20.2%), S. lugdunensis 2 (2.4%), S. warneri 2 (2.4%), S. cohnii 1 (1.2%) and others 3 (3.6%). Antibiotic susceptibility testing showed maximum resistance to ampicillin and penicillin with 85%-95% and sensitivity to piperillin/tazobactam, vancomycin and linezolid (85%-100%).

Conclusion: Due to emergence of multiple drug resistance among CONS isolates, there is a need to adopt simple laboratory procedure to identify and determine the prevalence and antibiotic resistant patterns of CONS.

Key words: Coagulase-negative Staphylococci, normal flora, hospital-acquired infections, antimicrobial susceptibility test

Introduction

Coagulase-negative Staphylococci (CONS), which are the normal flora of skin and mucous membrane, have emerged as predominant pathogens in hospital-acquired infections1. It belongs to family Micrococcaceae and genus Staphylococcus. They are gram positive, cluster forming cocci2. Previously CONS were generally considered to be contaminants having little significance. Over the past four decades, these organisms have become recognized as important agents of human disease3. CONS are divided into more than 44 species and more than a dozen subspecies, of which approximately half have been associated with humans4. Community- acquired coagulase negative staphylococcal infections are mostly due to S. saprophyticus, which is an important cause of urinary-tract infections in younger, sexually active women whereas hospital-acquired infections are due to S. epidermidis, usually results from the colonization of prosthetic materials in patients with vascular catheters or implanted prostheses5. CONS have become the 3rd cause of nosocomial bloodstream infections as a result of the combination of increased use of intravascular devices and an increase number of hospitalised immunocompromised patients6. These infections are difficult to treat because of the risk factors and the multiple drug resistant nature of the organisms7. Hence the present study was carried out to
determine the species distribution and antibiotic susceptibility pattern of CONS isolated from various clinical specimens.

**Materials and methods**

The study was carried for a period of 6 months at Department of Microbiology, VIMS, Ballari. A total of 84 consecutive non repeated clinically significant pure CoNS isolates were collected from various clinical samples like pus, sputum, urine, blood, fluid, ear swab, and throat swab. The isolates were considered clinically significant when isolated in pure culture from infected site or body fluid or if the same strain was isolated twice. The isolates were identified as CONS by colony morphology, Gram stain, catalase test and coagulase test (slide and tube coagulase). Bacitracin susceptibility was performed to exclude Micrococc and Stomatococcus species. Speciation of CoNS was done by Novobiocin resistance test, Urease activity, Ornithine decarboxylase and aerobic acid production from mannose. The antimicrobial susceptibility profiles of all isolates were done by Disc Diffusion Method using Mueller – Hinton agar plates according to the CLSI guidelines.

**Results**

A total of 84 CoNS were isolated from various clinical samples. Maximum number of CoNS were isolated from pus samples (30.9%), followed by sputum samples (29.7%), from urine samples (27.3%), from blood samples (5.9%), from ear swabs (5.7%), from fluid (1.1%) and from throat swab (1.1%). Identification of CoNS by simple scheme showed S.epidermidis (41.6%) as the most frequent isolate followed by S.saprophyticus (28.6%), S.haemolyticus (20.2%), S.warneri (2.4%), S.cohnii (1.2%) and unidentified (3.6%) in urine, pus and blood because of aberrant reactions. The isolation were more in males (64.2%) than females (35.7%) and in males majority of CoNS (55.5%) were found in the age group of >40 years and in females (30%) were found in age group of 30-40 years. Antibiotic susceptibility testing of the isolates showed maximum resistant to penicillin (94%) and ampicillin (85.7%), followed by erythromycin (70.2%), cefoxitin (65.4%), oxacillin (61.9%), piperclillin/tazobactam (28.5%) and linezolid (16.6%). No resistance to vancomycin was seen.

**Table 1**

<table>
<thead>
<tr>
<th>SPECIES</th>
<th>SPUTUM n (%)</th>
<th>URINE n (%)</th>
<th>PUS n (%)</th>
<th>BLOOD n (%)</th>
<th>EAR n (%)</th>
<th>THROAT n (%)</th>
<th>FLUID n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S.epidermidis 35(41.6)</td>
<td>13 (37.14)</td>
<td>10 (28.6)</td>
<td>8 (22.8)</td>
<td>1 (2.85)</td>
<td>2 (5.7)</td>
<td>-</td>
<td>1 (2.8)</td>
</tr>
<tr>
<td>S.saprophyticus 24(28.6)</td>
<td>8 (33.3)</td>
<td>7 (29.1)</td>
<td>7 (29.1)</td>
<td>1 (4.16)</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>S.haemolyticus 17(20.2)</td>
<td>3 (17.6)</td>
<td>6 (35.2)</td>
<td>5 (29.4)</td>
<td>2 (11.7)</td>
<td>-</td>
<td>1 (5.8)</td>
<td>-</td>
</tr>
<tr>
<td>S.lugdunensis 2(2.4)</td>
<td>-</td>
<td>-</td>
<td>2 (100)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

**Table 2**

A significant percentage of isolates resistant to penicillin (94%) followed by ampicillin (88.5%), erythromycin (40%) and cefoxitin (45.7%).

---

www.ijbamr.com P ISSN: 2250-284X, E ISSN: 2250-2858
<table>
<thead>
<tr>
<th>SPECIES</th>
<th>P</th>
<th>AMP</th>
<th>E</th>
<th>CX</th>
<th>OX</th>
<th>LZ</th>
<th>PIT</th>
</tr>
</thead>
<tbody>
<tr>
<td>S.epidermidis,35</td>
<td>33(94%)</td>
<td>31(88.5%)</td>
<td>14(40%)</td>
<td>16(45.7%)</td>
<td>11(31.4%)</td>
<td>4(11.4%)</td>
<td>6(17%)</td>
</tr>
<tr>
<td>S.saprophyticus,24</td>
<td>23(95.8%)</td>
<td>21(87.5%)</td>
<td>21(87.5%)</td>
<td>14(58.3%)</td>
<td>16(66.6%)</td>
<td>10(41.6%)</td>
<td>2(8.3%)</td>
</tr>
<tr>
<td>S.haemolyticus,17</td>
<td>16(94%)</td>
<td>15(88.2%)</td>
<td>15(88.2%)</td>
<td>15(88.2%)</td>
<td>17(100%)</td>
<td>-</td>
<td>11(64.7%)</td>
</tr>
<tr>
<td>S.lugdunensis,2</td>
<td>2(100%)</td>
<td>2(100%)</td>
<td>2(100%)</td>
<td>1(50%)</td>
<td>2(100%)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>S.warneri,2</td>
<td>2(100%)</td>
<td>2(100%)</td>
<td>-</td>
<td>1(50%)</td>
<td>1(50%)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>S.Cohnii,1</td>
<td>1(100%)</td>
<td>-</td>
<td>1(100%)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Others,3</td>
<td>2(66.6%)</td>
<td>2(66.6%)</td>
<td>2(66.6%)</td>
<td>2(66.6%)</td>
<td>1(33.3%)</td>
<td>-</td>
<td>1(33.3%)</td>
</tr>
<tr>
<td>TOTAL</td>
<td>79(94%)</td>
<td>72(85.7%)</td>
<td>59(70.2%)</td>
<td>55(65.4%)</td>
<td>52(61.9%)</td>
<td>14(16.6%)</td>
<td>24(28.5%)</td>
</tr>
</tbody>
</table>

P=penicillin, AMP=ampicillin, E=erythromycin, OX=oxacillin, LZ=linezolid, PIT=piperillin/tazobactam, CX=cefoxitin
Discussion

Coagulase-negative Staphylococci species formerly known as contaminants bacteria, but are now considered as important possible pathogens, recognized as important etiologic agents of a wide variety of human nosocomial infections accounting for about 9% of nosocomial infections\(^{12,13}\). In our study, out of 84 samples maximum isolation was from pus sample 26(30.9%), followed by sputum 25(29.7%) and urine 23(27.3%) which can be comparable with the similar study done by Asangi et al\(^{14}\). But in a study conducted by GoliaS et al\(^{6}\) showed that out of 134 samples 19 (14.2%) from blood cultures and 5 (3.7%) from fluid. In another study done by YedlaKavitha et al\(^{7}\) showed maximum number of CoNS were isolated from blood samples 25(31.65%) followed by urine 24(30.38%).

In our study *S.epidermidis* was the most frequent isolate 35(41.6%) followed by *S.saprophyticus* 24(28.6%), *S.haemolyticus* 17(20.2%), *S.lugdunensis* 2(2.4%), *S.warneri* 2(2.4%), *S.cohnii* 1(1.2%) which is similar to the other study conducted by GoliaS et al\(^{6}\) which showed *S.epidermidis* was the most frequent isolate 62 (46.3%) followed by *S.saprophyticus* 38 (28.4%), *S.hemolyticus* 27 (20.1%), *S.lugudensis&S.warneri* 3 (2.2%), *S.cohnii* 1 (0.7%). This study also correlates with another study done by Shubra Singh et al\(^{15}\) where Staphylococcus epidermidis was isolated in 40% of their clinical isolates followed by Staphylococcus haemolyticus(14%), Staphylococcus saprophyticus(12%), Staphylococcus lugdunensis(6%) and Staphylococcus hominis(6%). Manikandan et al\(^{16}\) showed *S.epidermidis* as the most predominant CONS (57%) followed by Staphylococcus hominis (22.8%).

Our study reported, 30(35.7%) cases were in >40 years of age group. The isolates were more in males 54(64.2%) than females 30 (35.7%) which is
comparable with a study by Larry M. Baddour, David L. et al\textsuperscript{17} incidence of CoNS was significantly higher among those 60 and older. Similar study done by Golia S et al\textsuperscript{6} which showed out of 134 CONS 52 (38.8\%) cases >40 years of age group. The isolation was more in males 87 (64.9\%) than females 47 (37.07\%). In this study, \textit{S.epidermidis} showed a significant percentage of isolates resistant to penicillin (94\%) followed by ampicillin (88.5\%), erythromycin (40\%) and cefoxitin (45.7\%). This is similar to the study conducted by Asangi et al\textsuperscript{14} In our study, antibiotic susceptibility testing showed maximum resistance to penicillin 79 (94\%), ampicillin 72 (85.7\%), and 52 (61.9\%) strains showed resistance to oxacillin which is correlated to study conducted by Golia S et al\textsuperscript{6} showed resistance to penicillin 95.5\%, ampicillin 88\% and cefoxitin 66.4\%. In this study, piperacillin/tazobactam 24 (28.5\%) and linezolid 14 (16.6\%) showed least resistance and no resistance to vancomycin was observed. This correlates with the study conducted by Goyal R et al\textsuperscript{8} and Yedla Kavitha et al\textsuperscript{7}. Hence, it is therefore recommended to assess the importance of CONS, speciate the clinically relevant CoNS and perform the antibiotic susceptibility testing before any typing procedure for epidemiological studies are undertaken\textsuperscript{8}.

**Conclusion**

CONS have become the major cause of nosocomial infections as a result of the combination of increased use of intravascular devices and an increase in the number of hospitalized immunocompromised patients. Emergence of multiple drug resistance among them demonstrates the need to adopt simple laboratory procedure to identify and determine the prevalence and antibiotic resistant patterns of CONS and also glycopeptides have been considered as the drug of choice for the management of these organisms.

**References**

17. Larry M. Baddour, David L. Comparison of microbiologic characteristics of pathogenic and saprophytic coagulase-negative staphylococci from patients on continuous ambulatory peritoneal dialysis. Diagnostic Microbiology and Infectious Diseases. 1986, 5(3): 197-205.