

## Original article

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

Pavithra DP<sup>1</sup>, Divya P<sup>\*2</sup>

<sup>1</sup>Assistant Professor, Department of Microbiology, Bangalore Medical College & Research Institute, Bangalore, Karnataka.

<sup>2</sup>Assistant 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.

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## 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 piperacillin/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

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## Introduction

Coagulase-negative Staphylococci (CONS), which are the normal flora of skin and mucous membrane, have emerged as predominant pathogens in hospital-acquired infections<sup>1</sup>. It belongs to family Micrococcaceae and genus Staphylococcus. They are gram positive, cluster forming cocci<sup>2</sup>. 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 disease<sup>3</sup>. CONS are divided into more than 44 species and more than a dozen subspecies, of which approximately half have been associated with humans<sup>4</sup>. 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 prostheses<sup>5</sup>. 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 patients<sup>6</sup>. These infections are difficult to treat because of the risk factors and the multiple drug resistant nature of the organisms<sup>7</sup>. 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 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 Isolate were considered clinically significant when isolated in pure culture from infected site or body fluid or if the same strain was isolated twice<sup>8</sup>.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 Micrococci and Stomatococcus species<sup>9</sup>. Speciation of CoNS was done by Novobiocin resistance test,Urease activity, Ornithine decarboxylase and aerobic acid production from mannose<sup>3,10</sup>.The antimicrobial susceptibility profiles of all isolates were done by Disc Diffusion Method using Mueller –Hinton agar plates according to the CLSI guidelines<sup>11</sup>.

**Results**

A total of 84 CoNS were isolated from various clinical samples. Maximum number of CoNS were isolated from pus samples 26(30.9%), followed by

sputum samples 25(29.7%), from urine samples 23(27.3%),from blood samples 5(5.9%),from ear swabs 2(2.5%),from fluid 1(1.1%) and 1(1.1%) from throat swab. Identification of CoNS by simple scheme showed S.epidermidis35(41.6%) as the most frequent isolate followed by S.saprophyticus24(28.6%),S.haemolyticus17(20.2%),S.lugdunensis2(2.4%),S.warneri2(2.4%),S.cohnii1(1.2%) and unidentified 3(3.6%) in urine, pus and blood because of aberrant reactions.(**Table.1**)

The isolation were more in males 54(64.28%) than females 30(35.7%) and in males majority of CoNS 30(55.5%) were found in the age group of >40years and in females 9(30%) were found in age group of 30-40years.

Antibiotic susceptibility testing of the isolates showed maximum resistant to penicillin79 (94%) and ampicillin 72(85.7%), followed by erythromycin 59(70.2%),cefoxitin 55(65.4%),oxacillin52(61.9%), piperacillin/tazobactam24(28.5%) and linezolid 14(16.6%). No resistance to vancomycin was seen.(**Table.2& Graph.1**)

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

**TABLE.1 SHOWING FREQUENCY AND PERCENTAGE OF CONS IN VARIOUS CLINICAL SAMPLES**

SPECIES n (%)	SPUTUM n (%)	URINE n (%)	PUS n(%)	BLOOD n(%)	EAR n(%)	THROAT n(%)	FLUID n(%)
<b>S.epidermidis 35(41.6)</b>	<b>13 (37.14)</b>	<b>10 (28.6)</b>	<b>8(22.8)</b>	<b>1(2.85)</b>	<b>2(5.7)</b>	<b>-</b>	<b>1(2.8)</b>
<b>S.saprophyticus 24(28.6)</b>	<b>8(33.3)</b>	<b>7(29.1)</b>	<b>7(29.1)</b>	<b>1(4.16)</b>	<b>-</b>	<b>-</b>	<b>-</b>
<b>S.haemolyticus 17(20.2)</b>	<b>3 (17.6)</b>	<b>6(35.2)</b>	<b>5(29.4)</b>	<b>2(11.7)</b>	<b>-</b>	<b>1(5.8)</b>	<b>-</b>
<b>S.lugdunensis2(2.4)</b>	<b>-</b>	<b>-</b>	<b>2(100)</b>	<b>-</b>	<b>-</b>	<b>-</b>	<b>-</b>

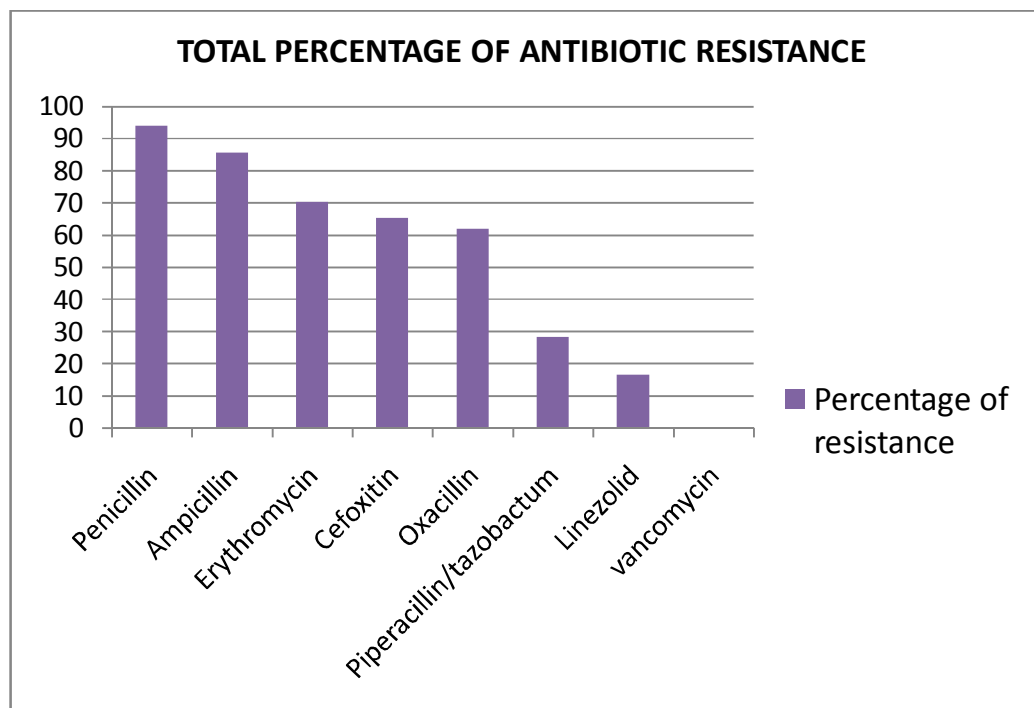
<b>S.warneri</b> 2(2.4)	-	-	<b>2(100)</b>	-	-	-	-
<b>S.Cohnii</b> 1(1.2)	<b>1(100)</b>	-	-	-	-	-	-
<b>Others</b> 3(3.6)	-	<b>1</b>	<b>1</b>	<b>1</b>	-		-
<b>TOTAL</b>	<b>25(29.7)</b>	<b>23(27.3)</b>	<b>26(30.9)</b>	<b>5(5.9)</b>	<b>2(2.5)</b>	<b>1(1.1)</b>	<b>1(1.1)</b>

**TABLE.2 RESISTANCE PATTERNS OF CONS TO DIFFERENT ANTIBIOTICS**

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

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

Graph.1



### 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<sup>12,13</sup>. 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<sup>14</sup>. But in a study conducted by GoliaS et al<sup>6</sup> 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<sup>7</sup> 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<sup>6</sup> 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. lugdunensis* & *S. warneri* 3 (2.2%), *S. cohinii* 1 (0.7%). This study also correlates with another study done by Shubra Singh et al<sup>15</sup> 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<sup>16</sup> 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<sup>17</sup> incidence of CoNS was significantly higher among those 60 and older. Similar study done by Golia S et al<sup>6</sup> 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, *S. epidermidis* showed a significant percentage of isolates resistant to penicillin (94%) followed by ampicillin (88.5%), erythromycin (40%) and ceftiofloxacin (45.7%). This is similar to the study conducted by Asangi et al<sup>14</sup>

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<sup>6</sup> showed resistance to penicillin 95.5%, ampicillin 88% and ceftiofloxacin 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<sup>8</sup> and Yedla Kavitha et al<sup>7</sup>. Hence, it is therefore recommended to assess the importance of CONS, speculate the clinically relevant CoNS and perform the antibiotic susceptibility testing before any typing procedure for epidemiological studies are undertaken<sup>8</sup>.

### 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.

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