

## Coagulase negative staphylococci as a pathogen from wound infections in a Tertiary care Hospital

B. Nagasrilatha<sup>1,\*</sup>, M. Bharathi<sup>2</sup>, M. Sasidhar<sup>3</sup>, A. Shashikala<sup>4</sup>, S. Kusuma Bai<sup>5</sup>

<sup>1,2</sup>Associate Professor, <sup>3</sup>Professor & HOD, <sup>4,5</sup>Assistant Professor, Rajiv Gandhi Institute of Medical Sciences, Andhra Pradesh

**\*Corresponding Author:**

Email: lathabathala77@gmail.com

### Abstract

**Introduction:** Wound infections are a common type of infections that may contribute to longer hospital stay. Most of these infections are superficial and readily treated with a regimen of local care and antibiotics. Determination of the etiologic agent is vital in the final choice of antibiotics. CoNS are previously considered as contaminants and non-pathogenic have been identified as the etiologic agents in most hospital acquired infections. These were considered clinically significant when isolated in pure culture from infected sites and in repeated samples.

**Material and methods:** Retrospective statistical analysis from Jan – Dec 2015.

283 swabs from wound infections were processed by following standard operative procedures. All samples were subjected to direct microscopy (Gram staining) and culture. Culture positive samples were processed by doing Gram staining and Coagulase test. Coagulase negative staphylococcal isolates (CoNS) were subjected for antibiotic susceptibility testing.

**Results:** Of total 283 samples, 159 were culture positive (56.18%). Among 159 isolates 55 were Coagulase negative staphylococci (34.59%). The isolates showed high susceptibility to vancomycin (85.45%). But 80% of CoNS were MRCoNS. Susceptibility to gentamycin was high (69.09%) followed by Amoxiclav (amoxicillin and clavulanic acid - 58.18%), ciprofloxacin (52.72%), ceftriaxone (38.18%), cefepime and penicillin (36.36%) and cotrimoxazole (27.27%).

### Conclusions:

1. CoNS should be considered as an emerging pathogen in wound infections.
2. CoNS is becoming resistant to commonly used antibiotics.
3. CoNS is developing resistance even to vancomycin.

**Key words:** Wound infections, Pathogen, Antibiotic susceptibility testing, Methicillin resistance

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### Introduction

Wound is a breach in the normal tissue continuum, resulting in a variety of cellular and molecular sequelae. Infection of a wound may be defined as invasion of organisms through tissues following a breakdown of local and systemic host defenses.<sup>(1,2)</sup> Wound infections are a common type of infections that may contribute to longer hospital stay, significantly increase the cost of medical care and are likely to have an important role in the development of antimicrobial resistance. Most of these infections are superficial and readily treated with a regimen of local care and antibiotics. Determination of the etiologic agent is vital in the final choice of antibiotics.<sup>(3)</sup> Recent reports taken from the National Nosocomial Infections Surveillance System (NNIS) during the late 1980s and early 1990s have indicated that CoNS are among the five most commonly reported pathogens.<sup>(4)</sup>

CoNS are the indigenous flora of the human skin and mucous membrane<sup>(5)</sup> & has recently got attention as a potential pathogen, specifically for nosocomial infections<sup>(6,7)</sup>. CoNS are previously considered as contaminants and non-pathogenic have been identified as the etiologic agents in most hospital acquired infections<sup>(8,9,10)</sup>. These were considered clinically significant when isolated in pure culture from infected sites<sup>(5)</sup> and in repeated samples.

Antimicrobial resistance is an unavoidable consequence of the selective pressure of antibiotic exposure. Minimizing the antibiotic pressure is essential to control the emergence of resistant strains in the hospital and in the community.<sup>(3)</sup> Knowledge of the most common causative agents of infection and their antimicrobial susceptibility pattern is very essential for the judicious administration of empirical therapy before the culture results are available.<sup>(11)</sup> Therefore the present study was conducted to know the frequency of CoNS in wound swabs and their antibiotic susceptibility pattern in general and specially for methicillin.

### Aim

1. To know the frequency of CoNS in wound swabs
2. To know the antibiotic susceptibility pattern in general and specially for methicillin.

## Material and Methods

Retrospective statistical analysis from Jan – Dec 2015.

283 swabs from wound infections, received from various clinical departments were processed in bacteriology section of department of microbiology, RIMS, Kadapa. All samples were processed by following standard operative procedures. All samples were subjected to direct microscopy (Gram staining) and culture. Inoculation was done on to nutrient agar, blood agar and MacConkey agar and incubated at 37°C for 18 – 24 hrs aerobically. Culture positive samples were read by its colony morphology and Gram's staining. The isolates which showed Gram positive cocci in clusters in Gram's staining were considered as Staphylococci and were tested for Coagulase production. Coagulase negative staphylococcal isolates (CoNS) were further subjected for antibiotic susceptibility testing by Kirby – Bauer's disc diffusion method.

## Results

Of total 283 samples, 159 were culture positive (56.18%). Among 159 isolates 55 were Coagulase negative staphylococci (34.59%) as shown in Table 1.

**Table 1: Showing culture positivity and percentage of CoNS**

Total Samples	Culture Positive	CoNS
283	159(56.18%)	55 (19.43%)

Among 55 CoNS positive samples 28 and 27 isolates were from males & females respectively.

Out of 55 CoNS positive samples 42 were from inpatients. (76.36%) and 13 were from outpatients as shown in Table 2.

**Table 2: Showing gender wise and out-patient & in-patient wise distribution of samples**

	OP	IP	Total
Male	09	19	28
Female	04	23	27
Total	13	42	55

The isolates showed high susceptibility to vancomycin (85.45%). But 80% of CoNS were MRCoNS. Susceptibility to gentamycin was high (69.09%) followed by Amoxiclav (amoxicillin and clavulanic acid - 58.18%), ciprofloxacin (52.72%), ceftriaxone (38.18%), cefaperazone and penicillin (36.36%) and cotrimaxazole (27.27%) as shown in Table 3.

**Table 3: Showing Antibiotic Susceptibility pattern of CoNS**

	P	AMP	AMC	COT	CIP	G	CPZ	CTR	OX	VAN
OP/13	4 30.76	1 7.69	7 53.84	5 38.46	9 69.23	9 69.23	4 30.76	05 38.46	1 7.69	12 92.30
IP/42	16 38.09	1 2.38	25 59.52	10 23.8	20 47.61	29 69.04	16 38.09	16 38.09	10 23.8	35 83.33
Total(55)	20 36.36	02 3.63	32 58.18	15 27.27	29 52.72	38 69.09	20 36.36	21 38.18	11 20.0	45 85.45

P – Penicillin; AMP – Ampicillin; AMC – Amoxycrav; COT – Cotrimaxazole; CIP – Ciprofloxacin; G – Gentamycin; CPZ - Cefaperazone; CTR – Ceftriaxone; OX – Oxacillin; VAN - V ancomycin

## Discussion

Skin, the largest organ in the human body, plays a crucial role in the sustenance of life through the regulation of water and electrolyte balance, thermoregulation, and by acting as a barrier to external noxious agents including microorganisms, however, when the epithelial integrity of skin is disrupted, a wound results.<sup>(12)</sup> A break or abrasion in the skin can provide an entryway for these surface bacteria into the body, and they stick very well to the moist edges of a cut. The bacteria begin to multiply and extend into the cut. The body's defense mechanism includes bringing immune cells into the area to fight against the bacteria. Eventually, accumulation of these cells produces the thick whitish liquid that we call pus.<sup>(13)</sup>

Wound infection is a major problem in hospitals in developing countries as it increases morbidity and prolonged hospital stay.<sup>(14)</sup> Consequences of wound infection are increased trauma, rise in treatment costs and more resource demanding wound management.<sup>(15)</sup> Knowledge of the most common causative agents of infection and their antimicrobial susceptibility pattern is very essential for the judicious administration of empirical therapy before the culture results are available.<sup>(11)</sup>

Most of the studies revealed that Staphylococcus aureus, Esch.coli, Pseudomonas, Klebsiella are the most common pathogens of wound infections. But now the trend is changing towards Coagulase Negative Staphylococci (CoNS) which were considered as contaminants in the past, as some strains of Coagulase negative staphylococci produce some virulence factors. These include, among others, a surface Polysaccharide Adhesin (PS/A) and an Extracellular Slime Substance (ESS) which are responsible for biofilm formation. The biofilm thus formed includes encasement of bacteria.<sup>(8)</sup> Multi-resistant CoNS may adhere to medical devices and surfaces through slime which

secretes out of the cell and has a mucopolysaccharide structure, and in this way, they may easily colonize and spread within hospital environment. Furthermore, the slime factor protects the CoNS from antibiotics, phagocytosis and chemotaxis.<sup>(6)</sup>

In the present study we isolated CoNS in 19.43% of total samples (55/283) and 34.59% of total isolates (55/159). This figure was higher when compared to studies by Shittu et al (16.1%), Aizza et al (5.5%), Neelima et al (15.8%), DVMVSV Raghav Rao et al (9.35%), Khyati Jain et al (4.3%), and Vijeta et al (10%). The recognition of CoNS importance as pathogen in the recent past might be the reason for this higher percentage in the present study. Of the total 55 CoNS isolates, majority (76.36%) were isolated from inpatients showing their ability to form biofilms not only on intravenous catheters and various indwelling foreign devices but also their ability to colonize on tissues even in the absence of foreign bodies by a special attachment mechanisms involves specific interactions with various serum, plasma and tissue components of the host including connective tissue proteins and serum derived proteins<sup>(8,16)</sup> and their importance as causative agents in hospitalized patients who are vulnerable to get infections.

Susceptibility of CoNS to penicillin was 36.36% in present study which is at a higher percentage than by some studies.<sup>(4,17)</sup> Isolates from out patients showed a little more resistance than isolates from in patients. It might be due to selective pressure by penicillin as it is routinely prescribed in OP cases, on CoNS isolates, which represents presence of resistant strains in community. Whereas resistance to ampicillin was more in isolates from inpatients, which is inactivated by beta lactamases produced by the organisms. Again it might be due to selective pressure by using ampicillin in the treatment of inpatients.

Susceptibility to Amoxycyclin in present study is 58.18%, which is far less when compare to Vijeta et al (100%) and higher than Abdul hadi et al (27.8%). Most probably it might be due to prescribing commonly to majority of cases irrespective of OPD and IPD. Susceptibility to cotrimaxazole was high in the study by Vijeta et al (80%). Whereas the findings in present study (27.27%) showed more or less similar as Abdul hadi et al and Neelam et al. There is no difference in CoNS susceptibility to ciprofloxacin (52.72%) for in patients (52.38%) and out patients (53.84%) in our study but it is high in the study by Vijeta et al (80%) and less in a study by Koksals et al (45%).

Most of the CoNS isolates were susceptible to gentamycin (69.09%) in present study and it is still high in a study by Vijeta et al (100%). Resistance to gentamycin was only 14.5% in a study by Neelam et al. This represents CoNS are also highly sensitive to gentamycin including those resistant to penicillins.<sup>(18)</sup> CoNS isolates in present study showed less susceptibility to cephalosporins (cefpaperazone and

ceftraxone - 36.36% and 38.18% respectively) which might be due production of beta lactamases.

21.81% of CoNS isolates were methicillin sensitive in present study. Which is almost similar to Abdul hadi et al (24.1%), and Neelam et al study (33.3%) but it is high in DVMVSV Raghav rao et al (73.07%) and no resistant strains were found in Vijeta et al study(100% sensitive). It shows that the percentage of MRCoNS varies from place to place.

Vancomycin susceptibility was 85.45% in present study, but it was 100% in some studies.<sup>(4,6,11,13,17)</sup> Vancomycin has long been considered as an antibiotic of last resort for multi-drug-resistant staphylococci infections. On the other hand, vancomycin resistance has emerged first in enterococci and, more recently, in *S. aureus* and coagulase-negative staphylococci<sup>(19)</sup> Boneca and Chiosis, 2003; Palazzo et al., 2005). This condition has led CoNS to become a serious health problem that medical practitioners should be concerned about.<sup>(6)</sup>

In general the resistance of CoNS to various antibiotics was high in some studies than in the present study<sup>(4,17)</sup> and less in a study by Koksals et al.

As Coagulase negative staphylococci are acquiring virulence factors and emerging as pathogen, clinicians should follow Hospital antibiotic susceptibility before starting empirical treatment.

## Conclusions

1. CoNS should be considered as an emerging pathogen in wound infections.
2. CoNS is becoming resistant to commonly used antibiotics.
3. CoNS is developing resistance even to vancomycin.
4. Judicial use of antibiotics is a must to prevent development of resistance by the organism.

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