To study the Prevalence and Antimicrobial Susceptibility Pattern of Coagulase-negative Staphylococci (CoNS) isolated from various clinical sample at a Tertiary care centre, Uttar Pradesh.

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Abstract:

Introduction: Coagulase negative Staphylococci (CoNS) are one of the most common bacteria found on human skin and on mucous membranes as a normal flora. The presence of CoNS in clinical specimens is frequently associated with an infectious aetiology or contamination. CoNS are significant and commonly encountered pathogens in hospitals and they are occurring as the most preponderant isolates of all nosocomial infections.

Aim and Objective: To study the Prevalence and Antimicrobial Susceptibility Pattern of Coagulase-negative Staphylococci (CoNS) isolated from various clinical sample at a Tertiary care centre.

Material and Methods: This was a cross sectional study conducted in the Department of Microbiology at RMCHRC, Mandhana, Uttar Pradesh for a period of 1 year i.e, October 2021 to October 2022. A total of 58 CONS were isolated from 426 clinically relevant samples. The organisms were identified using the biochemical test from the clinical samples such as pus, wound swab, blood, throat swab and urine . The strains were identified as CoNS by colony morphology, Gram stain, catalase test and coagulase test. Bacitracin sensitivity was done to exclude Micrococci and Stomatococcus species. The antibiotic sensitivity testing was performed on Muller-Hinton agar by the Kirby-Bauer's disc diffusion method according to the CLSI guidelines.

Results: A total of 58 CONS were isolated from 426 clinically relevant samples. Out of the 58 isolates, 39(67.2%) were from wounds, 7(12%) from sputum and least for 1(1.7%) from throat swabs. The highest number of CoNS isolates were Staphylococcus epidermidis 31 (53.4%) and least for Staphylococcus saprophyticus 5 (8.6%). The CoNS in various clinical samples displayed Staphylococcus epidermidis as the predominant species isolated from pus & wound. The antibiotic sensitivity pattern of the isolates revealed 72.4% resistance to ampicill1in, 53.4% resistance to Co-trimoxazole and 37.9% resistance to cefoxitin (MRCoNS). There was no resistance to vancomycin and gentamicin observed.

Conclusion The increase in the resistance patterns and prevalence of CoNS is a result of frequent use of intravascular devices and raise in the number of immunocompromised patients in hospitals. This scenario emphasizes the need for rapid identification and speciation of CoNS with their antibiotic susceptibility for improved management of such cases and prevent emergence of drug resistance

Keywords: Coagulase Negative Staphylococcus, Antibiotic susceptibility patterns, CLSI, MRCoNS

Introduction

Coagulase-negative Staphylococci (CoNS), are the normal skin flora, emerged as predominant pathogens in hospital-acquired infections. CoNS are significant and commonly encountered pathogens in hospitals and they are occurring as the most preponderant isolates of all nosocomial infections [1]. Coagulase-negative staphylococci (CoNS) are systematically distinguished from S. aureus by the lack of coagulase. This enzyme promotes blood clotting, and the resulting fibrin coat on the bacterial surface may facilitate immune evasion. CoNS comprise a multitude of species, many of which are opportunistic pathogens [2]. Staphylococcus epidermidis is the most frequently encountered CoNS species on human skin and by far the most frequent source of CoNS infections [3]. S. epidermidis and other CoNS differ from S. aureus in that they are less virulent and typically cause chronic rather than acute infections. In fact, they may have an important role in contributing to immunity of the skin and mucous surfaces toward more harmful pathogens. However, serious complications can arise from chronic CoNS infections, particularly in immune-compromised, hospitalized, and very young or old patients..

These species have been documented as a cause of nosocomial bacteremia, wound infections, urinary tract infections, and pediatric and neonatal infections. These infections are not easy to treat because of the risk factors and the multiple drug resistance displayed by these organisms [4]. The drug resistance is highest in nosocomial infections because in hospitals, repeated

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contact with antibiotics leads to elimination of sensitive organisms from the flora and their substitution with resistant strains acquired by cross infection, especially CoNS.

Therefore, this study is undertaken to study the Prevalence and Antimicrobial Susceptibility Pattern of Coagulase-negative Staphylococci (CoNS) isolated from various clinical sample at a Tertiary care centre, Uttar Pradesh.

Material and Methods

This was a cross sectional study conducted in the Department of Microbiology at RMCHRC, Mandhana, Uttar Pradesh for a period of 1 year i.e, October 2021 to October 2022. A total of 58 CONS were isolated from 426 clinically relevant samples. The organisms were identified using the biochemical test from the clinical samples such as pus, wound swab, blood, throat swab and urine etc. The strains were identified as CoNS by colony morphology, Gram stain, catalase test and coagulase test. Bacitracin sensitivity was done to exclude Micrococci and Stomatococcus species. The antibiotic sensitivity testing was performed on Muller-Hinton agar by the Kirby-Bauer's disc diffusion method using ampicillin, amoxyclav, ceftriaxone. cotrimoxazole, cefotaxime, cefoxitin, gentamicin, amikacin and vancomycin according to the CLSI guidelines 2021 [5].

Results

A total of 58 CONS were isolated from 426 clinically relevant samples. Out of the 58 isolates, 39(67.2%) were from wounds, 7(12%) from sputum, 7(12%) from blood, 4(6.8%) from urine and 1(1.7%) from throat swabs.

The highest number of CoNS isolates were Staphylococcus epidermidis 31 (53.4%) followed by Staphylococcus haemolyticus 12 (20.6%), Staphylococcus lugdanensis 10 (17.2%), and Staphylococcus saprophyticus 5 (8.6%). (Table 1).

The species wise distribution of CoNS in various clinical samples displayed Staphylococcus epidermidis as the predominant species isolated from pus & wound swabs in our study. The other common species of CoNS in various clinical samples is displayed in Table No.2

Table No. 1: Incidence of different species of CoNS in the study

Species of CoNS	Number of isolates	Percentage
Staphylococcus epidermidis	31	53.44%
Staphylococcus haemolyticus	12	20.6%
Staphylococcus lugdanensis	10	17.2%
Staphylococcus saprophyticus	5	8.6%



Graph No 1: Graphical representation of Incidence of different species of CoNS in the study

Table No	2:	Species	of	CoNS	in	various	clinical
samples							

CoNS Species	Pus And Wou nd Swa bs	Sput um	Blo od	Uri ne	Thr oat Swa bs	Tot al
Staphyloc occus epidermidi s	21	3	4	2	1	31
Staphyloc occus haemolyti cus	9	1	1	1	-	12
Staphyloc occus lugdanens is	6	2	2	-	-	10
Staphyloc occus saprophyti cus	3	1	-	1	-	5

There was no specific gender predisposition for isolation of CoNS in our study. The maximum number of isolates was observed in the age group of 51-60 years of age followed by 31-40 years of age. The least number of isolates was observed in the age group of patients above 61 years of age.

Table No3: Age wise distribution of CoNS

S.No.	Age (in years)	No. of Cases	Percentage
1	0- 10	1	1.70%
2	20-Nov	9	15.50%
3	21-30	8	13.70%
4	31-40	12	20.60%
5	41-50	8	13.70%
6	51-60	17	29.30%
7	≥61	2	3.40%

The antibiotic sensitivity testing was performed on Muller-Hinton agar by the Kirby-Bauer disc diffusion method using a panel of nine antibiotics, which included Ampicillin (A), Amoxyclav (Amc), Ceftriaxone (Ci), Cotrimoxazole (Co), Cefotaxime (Ce), Cefoxitin (Cn), Gentamicin (G), Amikacin (Ak) and Vancomycin (Va). The maximum number of isolates was resistant to ampicillin (72.4%). All isolates were sensitive to gentamicin and vancomycin. The incidence of MRCoNS (Methicillin resistant CoNS) in this study was found to be 22(37.9) %.

Table No 4: Antibiotic sensitivity pattern of CoNS isolates in the study

Antibiotic	Resistance pattern (%)	Number of isolates
Ampicillin	72.4%	42
Amoxyclav	32.7%	19
Ceftriaxone	37.9%	22
Co- trimoxazole	53.4%	31
Cefotaxime	32.7%	19
Gentamicin	NILL	0
Amikacin	31%	18
Cefoxitin	37.9%	22
Ciprofloxacin	22.4%	13
Vancomycin	NILL	0

 Table No 5: Methicillin resistance pattern of CoNS species

Species of CoNS	Number of Isolates Tested	Number of MRCoNS
Staphylococcus epidermidis	31	12
Staphylococcus haemolyticus	12	8
Staphylococcus lugdanensis	10	2
Staphylococcus saprophyticus	5	0
TOTAL	58	22

The maximum number of isolates of MRcoNS was observed for Staphylococcus epidermidis 12 and least for Staphylococcus lugdanensis. There was no MRcoNs observed in Staphylococcus saprophyticus.

Discussion

Coagulase-negative Staphylococci (CoNS), which are the normal skin flora, have emerged as predominant pathogens in hospital-acquired infections. CoNS are significant and commonly encountered pathogens in hospitals and are occurring as the most preponderant isolates of all nosocomial infections [1]. Coagulase Negative Staphylococci (CoNS) are a major component of the normal flora of human skin and the oral flora found on mucous membranes [1, 2]. These Gram positive bacteria have been found to be the third most common causative agent of nosocomial infections. Clinical isolates of CoNS to the species level are usually not identified by many laboratories as they are considered normal inhabitants of skin and anterior nares. They are known to be causing only opportunistic infections. Moreover the methods used for speciation of CoNS utilize a series of biochemical reactions which are burdensome and often give inconsistent results.

In the present study a total of 58 CONS were isolated from 426 clinically samples. Out of the 58 isolates, 39(67.2%) were from wounds, 7(12%) from sputum, 7(12%) from blood, 4(6.8%) from urine and 1(1.7%)from throat swabs. This study was in support with the study performed by C. Roopa et al., [6] where maximum number of CoNS was isolated from the surgical ward and from pus samples.

The highest number of CoNS isolates were Staphylococcus epidermidis 31 (53.4%) followed by Staphylococcus haemolyticus 12 (20.6%), Staphylococcus lugdanensis 10 (17.2%), and Staphylococcus saprophyticus 5 (8.6%). This study was parallel to the study performed by Usha et al., [7] wherein Staphylococcus epidermidis was the predominant species, followed by Staphylococcus haemolyticus.

In the present study the antibiotic sensitivity pattern of the isolates revealed 72.4 % resistance to ampicillin, 53.4 % resistance to Co-trimoxazole and 37.9% resistance to cefoxitin (MRCoNS). There was no resistance to vancomycin and gentamicin observed in the present study which was similar to the study by the other author [6] [7] and also with no gender difference observed. The other Study by Sheikh and Mehdinejad, 2012 was also similar to our study [8].

For a long time, CoNS were considered as commensals and were rarely reported to cause severe infections. However, probably as a result of the combined effect of increased use of intravascular devices and the increase in the number of hospitalised immunocompromised patients, CoNS has emerged as a major cause of nosocomial blood stream infections [9]. In addition, as Staphylococcus epidermidis makes up a significant part of normal bacterial flora of the human skin and mucous membranes, it is easily introduced as a contaminant during the surgical implantation of polymeric devices [10].

The antibiotic resistance pattern of CoNS in our study shows resistance to multiple antibiotics like ampicillin, cotrimoxazole and cefoxitin. It will also be necessary for the hospital authorities to adapt specific antibiotic policies for treatment of all staphylococcal infections in accordance with their antibiogram reports and avoid using drugs in inadequate doses and for inappropriate duration.

Conclusion

CoNS will continue persist to be an infective agent in the future and hence studies on CoNS will facilitate in formulating and implementing particular antibiotic policies for treating CoNS infections and to control further emergence of drug resistant strains, in future.

References

- Mulu, W, Kibru, G, Beyene G, Damtie, M. Postoperative nosocomial infections and antimicrobial resistance pattern of bacteria isolates among patients admitted at Felege Hiwot Referral Hospital, Bahirdar, Ethiopia. Ethiopian J. Health Sci. 2012; 22(1): 7–18.
- 2. Otto M. Virulence factors of the coagulase-negative staphylococci. Front Biosci .2004; 9: 841–63.
- Otto M. Staphylococcus epidermidis the "accidental" pathogen. Nat Rev Microbial. 2009; 7:555–67.
- Asangi, S.Y., Mariraj, J., Sathyanarayan, M.S., Nagabhushan, R. Speciation of clinically significant Coagulase Negative Staphylococci and their antibiotic resistant pattern in a tertiary care hospital. Int. J. Biol. Med. Res. 2011; 2: 735–9.
- Clinical Laboratory Standards Institute. Performance Standards for Antimicrobial Susceptibility Testing: Twenty-Fifth Informational Supplement M100-S25. CLSI; Wayne, PA, USA: 2021.
- C. Roopa, Sunilkumar Biradar. Incidence and Speciation of Coagulase Negative Staphylococcus Isolates from Clinically Relevant Specimens with their Antibiotic Susceptibility Patterns. International Journal of Current Microbiology and Applied Sciences. 2015; 4(9): 975-980
- Usha, M.G., Shwetha, D.C., Vishwanath, G. Speciation of coagulase negative Staphylococcal isolates from clinically significant specimens and their antibiogram. Indian J. Pathol. Microbiol. 2013; 56: 258–60.
- Sheikh, A.F., Mehdinejad, M. Identification and determination of coagulase negative Staphylococci species and antimicrobial susceptibility pattern of isolates from clinical specimens. Afr. J. Microbiol. Res. 2012; 6: 1669–74
- 9. Becker K, Heilmann C, Peters G. Coagulase-negative staphylococci. Clin Microbiol Rev .2014; 27: 870–926.
- Sommerstein R, Kohler P, Wilhelm MJ, et al.Factors associated with methicillin-resistant coagulase-negative staphylococci as causing organisms in deep sternal wound infections after cardiac surgery. New Microbes New Infect .2015; 6: 15–21.