Iranian Journal of Microbiology
Volume 17 Number 4 (August 2025) 577-585
DOI: http://doi.org/10.18502/ijm.v17i4.19272

ORIGINAL ARTICLE

The emerging threat of multidrug-resistant *mecA* gene-positive coagulase-negative Staphylococci

Samira Fattah Hamid[1*], Aza Bahadeen Taha[2], Shler Qasim Hussien[3]

[1]Department of Medical Microbiology, College of Health Sciences, Hawler Medical University, Kurdistan Region, Iraq
[2]Department of Laboratories, Medical Research Center, Hawler Medical University, Erbil, Kurdistan Region, Iraq
[3]Department of Medical Laboratory Technology, Erbil Technical Health and Medical College, Erbil Polytechnique University, Kurdistan Region, Iraq

Received: December 2024**, Accepted:** May 2025

ABSTRACT

Background and Objectives: Coagulase-negative staphylococci (CoNS), previously classified as normal bacterial flora, have recently been associated with serious infectious diseases. The clinical isolation rate of these bacteria has increased in parallel with a rising prevalence of antibiotic resistance. Therefore, this study aims to determine the prevalence and species diversity of CoNS and their antibiotic susceptibility patterns.
Materials and Methods: Two hundred samples were collected from patients attending outpatient clinics. Bacterial genus, species, and antimicrobial susceptibility patterns were confirmed by the Vitek2 system. The *mecA* gene was then detected in all isolated bacteria using a polymerase chain reaction.
Results: The most frequently isolated bacterium was *Staphylococcus haemolyticus* accounting for 37.83% of the isolates and was identified in different specimens. The antibiotic susceptibility profile illustrated the highest resistance against cefoxitin, followed by erythromycin, tetracycline, gentamicin, levofloxacin, clindamycin, and tobramycin. The *mecA* gene was detected in 95.49%, and all isolates demonstrated resistance to one or more classes of antibiotics. The highest degree of multiple resistance involved six classes of antibiotics.
Conclusion: Methicillin resistance in coagulase-negative staphylococci is alarmingly high. Periodic surveillance of multidrug-resistant CoNS is essential to monitor changes in their antimicrobial susceptibility and to prevent their transition from opportunistic pathogens to regular pathogens.

Keywords: Staphylococcal infections; Coagulase; Multidrug resistance; *mecA*; Methicillin resistance

INTRODUCTION

Within the *Staphylococcus* genus, *S. aureus* has historically been the only recognized pathogenic species (1). However, recent studies have increasingly focused on coagulase-negative staphylococci (CoNS) and highlighted their clinical significance. It is important to recognize that CoNS are generally regarded as non-pathogenic bacteria that inhabit the skin and mucous membranes (1, 2). Nevertheless,

*Corresponding author: Samira Fattah Hamid, Ph.D, Department of Medical Microbiology, College of Health Sciences, Hawler Medical University, Kurdistan Region, Iraq. Telefax: +964-750491021 Email: samira.hamid@hmu.edu.krd

these organisms can act as opportunistic pathogens, particularly in immunocompromised individuals and patients with implanted medical devices (3). Currently, more than 70% of CoNS worldwide are resistant to methicillin and are classified as methicillin-resistant CoNS (MRCoNS) (4, 5). The emergence and spread of MRCoNS in the hospital setting has increasingly become problematic. It's important to note that, according to the literature, the prevalence of MRCoNS is generally lower than that of methicillin-resistant *S. aureus* (MRSA). This lower prevalence may also be due to the infrequent occurrence of CoNS infections compared to *S. aureus*, and CoNS infections are often not accurately diagnosed as the cause of the disease (6, 7).

CoNS can also develop resistance to methicillin, similar to coagulase-positive staphylococci. There are two main ways in which staphylococci species develop resistance to β-lactam antibiotics. One way is by producing β-lactamase enzymes that can break down the antibiotic. The other way is to change the target site of β-lactams through making a new penicillin-binding protein (PBP2a) with low affinity for all β-lactam antibiotics. The *mecA* gene encodes PBP2a protein and is located on the staphylococcal cassette chromosome mec (SCCmec) mobile genetic element. This protein is responsible for the most clinically relevant antibiotic resistance mechanism in *S. aureus.* Analysis of the evolution of this gene suggests that it likely originated from the native *mecA* gene present in the *S. sciuri* group, which has undergone recombination and mutation events (2, 7-9).

The pathogenic potential of CoNS has increasingly being recognized, especially as multidrug resistance, and particularly MRCoNS has become more common. It's important to identify different species of CoNS and determine their antimicrobial resistance patterns (10). CoNS species show resistance to commonly used antibiotic classes such as macrolides and aminoglycosides, as well as to last-resort antibiotics such as glycopeptides (11).

MRCoNS are currently receiving increased attention due to their identification as the source of the *mecA* gene that can be transferred to *S. aureus* (10). Despite their clinical significance and impact on patient outcomes, multidrug-resistant and *mecA*-positive CoNS have not received as much attention as other antimicrobial-resistant pathogens such as MRSA. Therefore, there is a critical need for comprehensive studies focused on understanding the epidemiology, antimicrobial resistance profiles, and clinical implications of these emerging pathogens. Consequently, this study aimed to isolate and identify CoNS from different hospitals in Erbil city, investigate their antimicrobial susceptibility patterns, and detect the methicillin resistance-coding gene (*mecA*) among the CoNS isolates.

MATERIALS AND METHODS

Specimen collection. Specimens were collected from patients attending outpatient consultation clinics at Hawler Teaching Hospital, Rizgary Teaching Hospital, and Raparin Teaching Hospital in Erbil, Iraq, from January 2024 to June 2024. Clinical specimens obtained included blood, urine, sputum, throat swabs, wound swabs, aspirated fluids, pleural fluids, and pus. The specimens were cultured on blood agar and mannitol salt agar media (Lab M / UK), incubated at 37°C, and tested after 18 to 24 hours.

Primary identification and confirmation. All isolates were primarily identified based on their general cultural characteristics observed on blood agar and mannitol salt agar. This identification process included the examination of blood lysis and mannitol sugar fermentation. Staphylococci that showed mannitol fermentation on mannitol salt agar and exhibited complete lysis of blood surrounding colonies on blood agar were confirmed as *S. aureus* and subsequently excluded from the study (12-14). Other staphylococci were further analyzed for identification using the Vitek2 system (BioMérieux, France) with the gram-positive (GP) card for confirmation of bacterial identification (15).

Antibiotic susceptibility testing. The antibiotic susceptibility profile of all bacteria to 14 antibiotics belonging to 9 classes of antibiotics and the minimum inhibitory concentration of antibiotics were determined using the Vitek2 AST-GP card according to the manufacturer's instructions.

Phenotype confirmation of methicillin resistance. The penicillin-binding protein (PBP2a) latex agglutination test kit (Oxoid, Hampshire, UK) was used to confirm methicillin-resistant bacteria according to the manufacturer's protocol.

Detection of *mecA* gene. Genomic DNA from isolated bacterial strains was extracted using the AddPrep Bacterial Genomic DNA Extraction Kit (ADD Bio INC, Republic of Korea) according to the manufacturer's protocols. After extraction, the purity of the DNA was checked using a NanoDrop spectrophotometer (Thermo Scientific, USA). The DNA was then amplified in a thermocycler to determine the presence or absence of the *mecA* gene. The primers used for amplification were: Forward primer 5´-CTC AGG TAC TGC TAT CCA CC-3´ and Reverse primer 5´-CAC TTG GTA TAT CTT CAC C-3´ (Taha et al., 2019). The thermal cycler program was modified accordingly and performed as follows: initial denaturation for 5 minutes at 95°C, followed by thirty cycles consisting of 30 seconds of denaturation at 94°C, 30 seconds of annealing at 42°C, and 30 seconds of extension at 72°C, culminating in a final extension period of 10 minutes at 72°C. The amplified gene was then visualized by 1.5% agarose gel electrophoresis using a DNA molecular marker (100 bp DNA Ladder; GeNet Bio/South Korea) to confirm the presence of the expected 448 bp bands.

Data analysis. Statistical analyses were performed using SPSS version 26.0 (SPSS, Inc., Chicago, IL, USA). Descriptive statistics, including frequencies and percentages, were calculated to summarize the distribution of categorical variables. Frequency counts were used to quantify the number of occurrences for each category, while percentages represented the proportion of each category.

RESULTS

Profile of isolated bacteria. Of the 200 Staphylococcus species specimens, 89 isolates were identified as *S. aureus* and subsequently excluded from the study. Specimen types that did not include any species of CoNS were also excluded from subsequent analysis. The remaining 111 isolates were classified as CoNS, representing 14 different species within the *Staphylococcus* genus. Wound specimens were the most common type of specimen collected, accounting for 34.2% of the total samples, followed closely by urine specimens at 30.6%. Burn specimens accounted for 18.9%, while skin lesions accounted for 8.1%. Other specimen types included blood (3.6%), eye specimen (2.7%), and ear specimen (0.9%), with joint fluid also accounting for 0.9% (Table 1).

The most commonly isolated bacteria were *S. haemolyticus*, accounting for 37.83% of the total isolates, followed by *S. epidermidis* at 19.81% and *S. hominis* at 13.51%. Other species, such as *S. lentus* (8.10%) and *S. warneri* (6.30%), were also present, while the remaining species individually accounted for less than 4% each. Among the bacterial isolates, *S. haemolyticus* was the most diverse, occurring in multiple specimen types, including blood (9.5%), urine (21.4%), burn (23.8%), and wound (38.1%) specimens. *S. epidermidis* was predominantly isolated from burns (31.8%) and urine (31.8%) samples also occurred in skin lesions (27.3%). *S. hominis* was predominantly detected in skin lesions (60.0%) and urine (26.7%). Other notable isolates included *S. lugdunensis*, *S. pasteuri*, and *S. warneri*, each of which was isolated exclusively in specific specimen types, while *S. saprophyticus* was solely found in urine specimens (100%) (Table 2).

Antibiotic susceptibility pattern of CoNS bacteria. The antibiotic susceptibility profile showed that the highest resistance was seen in the cefoxitin screen test, with 92.8% of the isolates showing resistance. Other notable resistance rates included erythromycin (73.0%), tetracycline (66.7%), and gentamicin (62.2%). Levofloxacin resistance was found in 54.1% of the isolates, closely followed by clindamycin (52.3%) and tobramycin (51.4%). Additionally, moxifloxacin resistance was noted in 46.8%, while trimethoprim/sulfamethoxazole and teicoplanin showed resistance rates of 31.5% and 27.9%, respectively. Vancomycin resistance was observed in 24.3% of isolates, and resistance to tigecycline was low at 6.3% (Table 3).

Table 1. Distribution of specimens infected with CoNS

Type of Specimen	n (%)
Wound	38 (34.2%)
Urine	34 (30.6%)
Burn	21 (18.9%)
Skin Lesion	9 (8.1%)
Blood	4 (3.6%)
Ocular	3 (2.7%)
Ear	1 (0.9%)
Joint Fluid	1 (0.9%)
Total	111

Table 2. Isolation of CoNS from different clinical specimens

CoNS	n (%) of isolation	Wound n (%)	Urine n (%)	Burn n (%)	Skin lesion n (%)	Blood n (%)	Ocular n (%)	Ear n (%)	Joint Fluid n (%)
S. haemolyticus	42 (37.83)	9 (21.43)	16 (38.10)	10 (23.81)	0	4 (9.52)	2 (4.76)	0	1 (2.38)
S. epidermidis	22 (19.82)	6 (27.27)	7 (31.82)	7 (31.82)	0	0	1 (4.55)	1 (4.55)	0
S. hominis	15 (13.51)	4 (26.67)	9 (60.00)	2 (13.33)	0	0	0	0	0
S. lentus	9 (8.11)	5 (55.56)	2 (22.22)	2 (22.22)	0	0	0	0	0
S. warneri	7 (6.31)	7 (100)	0	0	0	0	0	0	0
S. lugdunensis	4 (3.60)	4 (100)	0	0	0	0	0	0	0
S. saprophyticus	3 (2.70)	3 (100)	0	0	0	0	0	0	0
S. intermedius	2 (1.80)	2 (100)	0	0	0	0	0	0	0
S. xylosus	2 (1.80)	2 (100)	0	0	0	0	0	0	0
S. capitis	1 (0.90)	1 (100)	0	0	0	0	0	0	0
S. chromogenes	1 (0.90)	1 (100)	0	0	0	0	0	0	0
S. galinarum	1 (0.90)	1 (100)	0	0	0	0	0	0	0
S. pasteuri	1 (0.90)	1 (100)	0	0	0	0	0	0	0
S. pseudointermedius	1 (0.90)	1 (100)	0	0	0	0	0	0	0
Total	111 (100)	38 (34.23)	34 (30.63)	21 (18.92)	9 (8.11)	4 (3.60)	3 (2.70)	1 (0.90)	1 (0.90)

Table 3. Antibiotic-resistant profile of CoNS

Antibiotic	Resistant n (%)
Cefoxitin Screen Test	103 (92.79%)
Erythromycin	81 (72.97%)
Tetracycline	74 (66.67%)
Gentamicin	69 (62.16%)
Levofloxacin	60 (54.05%)
Clindamycin	58 (52.25%)
Tobramycin	57 (51.35%)
Moxifloxacin	52 (46.85%)
Trimethoprim/Sulfamethoxazole	35 (31.53%)
Teicoplanin	31 (27.93%)
Trimethoprim	29 (26.13%)
Vancomycin	27 (24.32%)
Rifampicin	24 (21.62%)
Tigecycline	7 (6.31%) *

Table 4. Resistance of CoNS to different classes of antibiotics

Number of antibiotic classes	Resistant bacteria number (%)
1	5 (4.50%)
2	7 (6.31%)
3	11 (9.91%)
4	12 (10.81%)
5	22 (19.82%)
6	25 (22.52%)
7	18 (16.22%)
8	8 (7.21%)
9	3 (2.70%)

The resistance of CoNS to various classes of antibiotics is presented in Table 4, highlighting the distribution of resistance from the lowest to the highest. The resistance of bacteria to at least one antibiotic in three classes is defined as multidrug resistance. Accordingly, 95.4 percent of bacteria illustrated multidrug resistance. The highest resistance was observed in 25 isolates (22.52%) against six antibiotic classes, followed by 22 isolates (19.82%) resistant to five classes. Next, 18 isolates (16.22%) showed resistance to seven classes, while 12 isolates (10.81%) were resistant to four classes. There were 11 isolates (9.91%) resistant to three classes, and 8 isolates (7.21%) exhibited resistance to eight classes. Resistance was lower for two classes, with 7 isolates (6.31%), and was least prevalent in 5 isolates (4.50%) against one class, and 3 isolates (2.70%) against nine classes.

Analysis of 111 CoNS isolates revealed significant variability in antibiotic resistance between species, as shown in Table 5. Notably, *S. haemolyticus* had the highest resistance to 5 and 6 antibiotic classes (26.19% each), while *S. epidermidis* had the highest resistance

Table 5. Percentage of multidrug resistance profile for each species of CoNS.

CoNS	n of isolations	Percentage of resistance to antibiotic classes								
		1	2	3	4	5	6	7	8	9
S. haemolyticus	42	2.38	0	4.76	7.14	26.19	26.19	21.43	4.76	7.14
S. epidermidis	22	4.55	9.09	9.09	18.18	13.64	31.82	9.09	4.55	0
S. hominis	15	6.67	6.67	20	0	13.33	20	13.33	20	0
S. lentus	9	0	0	11.11	22.22	22.22	0	33.33	11.11	0
S. warneri	7	0	0	28.57	14.29	28.57	14.29	14.29	0	0
S. lugdunensis	4	0	75	0	25	0	0	0	0	0
S. saprophyticus	3	0	0	0	33.33	33.33	33.33	0	0	0
S. intermedius	2	0	0	0	0	0	100	0	0	0
S. xylosus	2	0	0	50	0	50	0	0	0	0
S. capitis	1	0	0	0	0	0	0	100	0	0
S. chromogenes	1	0	0	0	0	0	0	0	100	0
S. galinarum	1	100	0	0	0	0	0	0	0	0
S. pasteuri	1	0	100	0	0	0	0	0	0	0
S. pseudointermedius	1	100	0	0	0	0	0	0	0	0
Total	111	4.5	6.31	9.91	10.81	19.82	22.52	16.22	7.21	2.7

to 6 classes (31.82%). Other species, such as *S. hominis*, *S. lentus*, and *S. lugdunensis*, showed significant resistance to specific antibiotic classes, with *S. lugdunensis* reaching 75% resistance to 2 classes. Overall, the average resistance rates indicated that the 5 and 6 antibiotic classes had the highest resistance, at 19.82% and 22.52%, respectively.

Phenotype confirmation and *mecA* gene detection. Of 111 CoNS identified by Vitek2, methicillin resistance was confirmed phenotypically in 91 (81.98%) of the isolates through the detection of PBP2a. Twenty bacteria tested negative for the PBP2a protein were related to the species *S. haemolyticus*, *S. xylosus*, *S. chromogenes*, and *S. gallinarum*. All 111 isolates underwent molecular confirmation for the *mecA* gene by conventional PCR, and only 5 of *S. epidermidis* tested negative for the *mecA* gene, while the remaining 106 (95.49%) tested positive for the gene (Fig. 1).

Among the CoNS that tested, 95.49% of isolates were found to be positive for the *mecA* gene, which is a marker for methicillin resistance. The cefoxitin screen test showed positive results for 92.8% of the isolates. Additionally, the PBP2a test revealed a positivity rate of 81.98%. Furthermore, the cefoxitin screen test demonstrated a sensitivity of 97.26% and a specificity of 40%, while the PBP2a test exhibited a sensitivity of 85.85% and a specificity of 80% (Table 6).

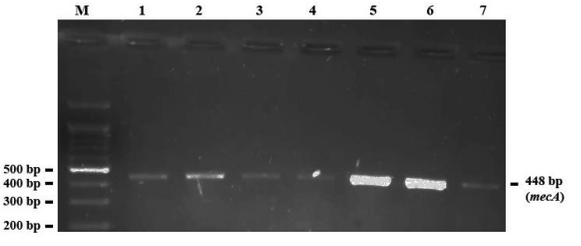

Fig. 1. PCR product of *mecA* gene in CONS bacteria. M: DNA marker (100bp ladder), Lane 1-7: PCR amplicon for *mecA* gene.

Table 6. Sensitivity and susceptibility of confirmatory tests.

Test	Positive n (%)	Negative n (%)
mecA gene	106 (95.49%)	5 (4.51%)
Cefoxitin Screen Test	103 (92.8%)	8 (7.2%)
PBP2a	91 (81.98%)	20 (18.02%)

DISCUSSION

In this study, CoNS isolates were obtained from different clinical specimens collected from three hospitals in Erbil, Iraq. The isolated bacteria showed a high diversity of CoNS species, including *S. haemolyticus*, *S. epidermidis*, *S. hominis*, *S. lentus*, *S. warneri*, *S. lugdunensis*, *S. saprophyticus*, *S. intermedius*, *S. xylosus*, *S. capitis*, *S. chromogenes*, *S.*

galinarum, *S. pasteuri*, and *S. pseudointermedius*. Among the CoNS species, *S. haemolyticus* was the most prevalent, followed by *S. epidermidis* and *S. hominis*. These staphylococci isolates were previously reported by other works (16, 17). This study reveals the prevalence and antimicrobial resistance profiles of CoNS in clinical samples, highlighting their significance as opportunistic pathogens. Among the isolated species, *S. haemolyticus* was the most frequently detected. This finding is consistent with previous research indicating that *S. haemolyticus* is a common contaminant and pathogen in clinical settings, especially in immunocompromised individuals. Its high prevalence raises concern about its role in hospital-acquired infections.

The distribution of CoNS across different specimen types has important clinical implications. The majority of isolates were obtained from wound and urine samples, indicating that CoNS are significant contributors to skin and soft tissue infections, as well as urinary tract infections. The notable presence of *S. epidermidis* in wound samples supports its role as a common skin flora and potential pathogen in postoperative infections. In contrast, the lower frequencies of CoNS in blood and ear specimen suggest a less common occurrence of these organisms in bloodstream infections, although their presence can- not be overlooked, especially in patients with com- promised immune systems. Our finding of *S. haemo- lyticus* as the predominant species differs from the findings of Nasaj et al, who reported *S. epidermidis* as the most commonly identified CoNS species (18). Bacteria were most frequently isolated from wound specimens, followed by urine and burn specimens. This finding is not consistent with any other research results due to differences in the types of patients at- tending the hospitals from which the specimens were collected (4, 19, 20).

The antibiotic resistance patterns observed in this study are concerning. A high percentage of isolates were resistant to cefoxitin, indicating a significant prevalence of methicillin-resistant strains. Additionally, resistance to commonly used antibiotics such as erythromycin and tetracycline raises significant concerns about treatment options. Genetic testing results support these findings, revealing that a significant proportion of isolates carry the *mecA* gene associated with methicillin resistance. This level of resistance complicates treatment and highlights the urgent need for robust antimicrobial stewardship programs to reduce the risk of infection and resistance. Multidrug resistance was present in almost all isolates, with a higher rate of resistance to 5 and 6 classes of antibiotics, which is consistent with a study conducted by Silva et al. who found high resistance to 4 and 5 classes of antibiotics (21).

The study found that a significant number of isolates were identified as MRCoNS by cefoxitin screening using the Vitek 2 system and were positive for the *mecA* gene. This finding is consistent with the results reported by Saenhom et al. who observed similar levels of methicillin resistance (22). In contrast, Shrestha et al. found a lower prevalence of *mecA* gene positivity (4). This difference highlights the variability in resistance patterns and emphasizes the need for ongoing surveillance and comparison across different studies. The presence of the *mecA* gene was strongly associated with methicillin resistance. Among all the isolates, only a few species were *mecA*-negative. However, this study found very few *mecA*-negative strains that were phenotypically resistant to methicillin. This could be due to a mechanism not dependent on penicillin-binding proteins, such as the overproduction of β-lactamase, or the presence of other low-affinity penicillin-binding proteins, which could explain this occurrence. In addition to the presence of the *mecA* gene in the vast majority of isolated strains, we found that a significant number of them were multidrug resistant. Chon et al. demonstrated that *mecA*-positive CoNS strains possessed multidrug resistance genes (23).

The PBP2a latex agglutination test shows low sensitivity and unsatisfactory performance with 20 isolates showing negative, the same poor performance was noted in the previous study by Chapin and Musgnug (24). The current study highlights the effectiveness of the mecA gene, the cefoxitin screen test, and the PBP2a test in detecting MRSA. While *mecA* offers the highest sensitivity, the cefoxitin screen's high sensitivity but low specificity raises concern about false-positive results. Combining these tests may improve diagnostic accuracy and guide appropriate treatment strategies.

The CoNS isolates showed varying degrees of susceptibility and resistance to the non-beta-lactam antibiotics tested, with particularly high resistance to gentamicin and erythromycin. These results are consistent with those reported by Al-Sultany and Al-Charrakh (25). However, our study revealed a

higher rate of resistance to tetracycline compared to their findings, which may be attributed to differences in the sample size or the number of bacterial isolates analyzed. In contrast, our results differ from those of a study conducted in South Africa by Asante et al. who reported a gentamicin resistance rate of only 2.2% (26). Additionally, the resistance rate to levofloxacin in our study was similar to the report by Khan et al. (27). This suggests a concerning upward trend in resistance to fluoroquinolones over time. However, the rate of resistance to clindamycin observed in our study was higher than that illustrated by Nicolosi et al. (28). Nicolosi and co-authors considered clindamycin as one of the antibiotics that could be used for treatment with a high success rate.

The study demonstrated that vancomycin and rifampin were the most effective antimicrobials against CoNS isolates. These findings suggest that these antimicrobial agents may be considered for empiric therapy in cases of CoNS-associated infections in the hospital setting. The results are consistent with those of Asante et al., who also identified vancomycin as the most effective antimicrobial agent against CoNS isolates (11). Furthermore, our results are consistent with those of Ashagrie et al., who reported no resistance to vancomycin, confirming its status as the most effective antibiotic against CoNS (29). The observed decrease in susceptibility to previously effective antimicrobial agents may be due to increased or cumulative use, as well as insufficient adherence to infection control practices within the studied hospitals.

The study's results suggest that the antimicrobial agents used are not recommended for initial empirical therapy against CoNS infections, possibly due to the overuse of these antibiotics in our country.

This study has several limitations, including a small and geographically limited sample size, which may limit the generalizability of the findings. Additionally, although *mecA* gene-positive CoNS were identified, other resistance mechanisms were not explored, and the reliance on phenotypic tests may have caused the misclassification of resistant isolates. The absence of genomic typing, biofilm analysis, and clinical outcome data further limits the understanding of strain spread, pathogenicity, and the impact on patient health. Future research should address these gaps by incorporating molecular typing, biofilm characterization, and clinical correlation to enhance understanding of multidrug-resistant CoNS.

CONCLUSION

The study revealed that methicillin resistance in CoNS was alarmingly high, both phenotypically and genotypically, with increased resistance rates to multiple classes of antibiotics. However, the isolates remained highly susceptible to vancomycin and rifampicin, suggesting that these antibiotics can still be considered viable empirical treatment options. Given the increasing incidence of CoNS as a significant cause of infection, there is a critical need to focus on the identification, speciation, and resistance patterns of these isolates to improve patient management. In addition, the implementation of regular active surveillance for multidrug-resistant CoNS will be essential to track changes in their epidemiology and antimicrobial susceptibility profiles.

REFERENCES

1. Grazul M, Balcerczak E, Sienkiewicz M. Analysis of the Presence of the Virulence and Regulation Genes from *Staphylococcus aureus (S. aureus)* in Coagulase Negative Staphylococci and the Influence of the Staphylococcal Cross-Talk on Their Functions. *Int J Environ Res Public Health* 2023; 20: 5155.
2. Bonvegna M, Grego E, Sona B, Stella MC, Nebbia P, Mannelli A, et al. Occurrence of Methicillin-Resistant Coagulase-Negative Staphylococci (MRCoNS) and Methicillin-Resistant *Staphylococcus aureus* (MRSA) from Pigs and Farm Environment in Northwestern Italy. *Antibiotics (Basel)* 2021; 10: 676.
3. Chenouf NS, Mama OM, Messaï CR, Ruiz-Ripa L, Fernández-Fernández R, Carvalho I, et al. Detection of methicillin-resistant coagulase-negative staphylococci and PVL/*mecA* genes in cefoxitin-susceptible *Staphylococcus aureus* (t044/ST80) from unpasteurized milk sold in stores in Djelfa, Algeria. *J Dairy Sci* 2021; 104: 2684-2692.
4. Shrestha LB, Bhattarai NR, Rai K, Khanal B. Antibiotic Resistance and mecA Gene Characterization of Coagulase-negative Staphylococci isolated from Clinical Samples in Nepal. *Infect Drug Resist* 2020; 13: 3163-3169.
5. Wang J, Meng Y, Zhang R, Yan H, Xu G, Zhu Y, et al.

Coagulase-negative staphylococci are the main causes of bacterial meningitis in duck. *Poult Sci* 2024; 103: 103592.
6. Deyno S, Fekadu S, Seyfe S. Prevalence and antimicrobial resistance of coagulase negative staphylococci clinical isolates from Ethiopia: a meta-analysis. *BMC Microbiol* 2018; 18: 43.
7. Martins A, Cunha Mde L. Methicillin resistance in *Staphylococcus aureus* and coagulase-negative staphylococci: Epidemiological and molecular aspects. *Microbiol Immunol* 2007; 51: 787-795.
8. Cave R, Misra R, Chen J, Wang S, Mkrtchyan HV. Whole genome sequencing revealed new molecular characteristics in multidrug resistant staphylococci recovered from high frequency touched surfaces in London. *Sci Rep* 2019; 9: 9637.
9. Medis S, Dissanayake T, Weerasekera M, Namali D, Gunasekara S, Kottahachchi J. Distribution of *mecA* and qacA/B genes among coagulase negative staphylococci isolated from central venous catheters of intensive care unit patients. *Indian J Med Microbiol* 2022; 40: 505-509.
10. Dhaouadi S, Soufi L, Campanile F, Dhaouadi F, Sociale M, Lazzaro L, et al. Prevalence of meticillin-resistant and -susceptible coagulase-negative staphylococci with the first detection of the *mecC* gene among cows, humans and manure in Tunisia. *Int J Antimicrob Agents* 2020; 55: 105826.
11. Asante J, Hetsa BA, Amoako DG, Abia ALK, Bester LA, Essack SY. Multidrug-Resistant Coagulase-Negative Staphylococci isolated from Bloodstream in the uMgungundlovu District of KwaZulu-Natal Province in South Africa: Emerging Pathogens. *Antibiotics (Basel)* 2021; 10: 198.
12. Sharp SE, Searcy C. Comparison of mannitol salt agar and blood agar plates for identification and susceptibility testing of *Staphylococcus aureus* in specimens from cystic fibrosis patients. *J Clin Microbiol* 2006; 44: 4545-4546.
13. Gaillot O, Wetsch M, Fortineau N, Berche P. Evaluation of CHROMagar Staph. aureus, a new chromogenic medium, for isolation and presumptive identification of *Staphylococcus aureus* from human clinical specimens. *J Clin Microbiol* 2000; 38: 1587-1591.
14. Bautista-Trujillo GU, Solorio-Rivera JL, Rentería-Solórzano I, Carranza-Germán SI, Bustos-Martínez JA, Arteaga-Garibay RI, et al. Performance of culture media for the isolation and identification of *Staphylococcus aureus* from bovine mastitis. *J Med Microbiol* 2013; 62: 369-376.
15. Spanu T, Sanguinetti M, Ciccaglione D, D'Inzeo T, Romano L, Leone F, et al. Use of the VITEK 2 system for rapid identification of clinical isolates of Staphylococci from bloodstream infections. *J Clin Microbiol* 2003; 41: 4259-4263.
16. Pung HP, Mat Azis N, Abdul Rachman AR, Jamaluddin TZMT, Amin Nordin S, Suhaili Z, et al. Presence of methicillin resistance and heteroresistance among Coagulase Negative Staphylococci (CoNS) isolates obtained from Health Sciences students at a Public University. *Trop Biomed* 2017; 34: 84-88.
17. Chajęcka-Wierzchowska W, Gajewska J, Zadernowska A, Randazzo CL, Caggia C. A comprehensive study on antibiotic resistance among coagulase-Negative Staphylococci (CoNS) strains isolated from ready-to-eat food served in bars and restaurants. *Foods* 2023; 23: 514.
18. Nasaj M, Saeidi Z, Asghari B, Roshanaei G, Arabestani MR. Identification of hemolysin encoding genes and their association with antimicrobial resistance pattern among clinical isolates of coagulase-negative Staphylococci. *BMC Res Notes* 2020; 13: 68.
19. Osman K, Alvarez-Ordóñez A, Ruiz L, Badr J, El-Hofy F, Al-Maary KS, et al. Antimicrobial resistance and virulence characterization of *Staphylococcus aureus* and coagulase-negative staphylococci from imported beef meat. *Ann Clin Microbiol Antimicrob* 2017; 16: 35.
20. Manandhar S, Singh A, Varma A, Pandey S, Shrivastava N. Phenotypic and genotypic characterization of biofilm producing clinical coagulase negative staphylococci from Nepal and their antibiotic susceptibility pattern. *Ann Clin Microbiol Antimicrob* 2021; 20: 41.
21. Silva V, Caniça M, Ferreira E, Vieira-Pinto M, Saraiva C, Pereira JE, et al. Multidrug-Resistant Methicillin-Resistant Coagulase-Negative Staphylococci in healthy poultry Slaughtered for Human consumption. *Antibiotics (Basel)* 2022; 11: 365.
22. Saenhom N, Kansan R, Chopjitt P, Boueroy P, Hatrongjit R, Kerdsin A. Evaluation of in-house cefoxitin screening broth to determine methicillin-resistant staphylococci. *Heliyon* 2022; 8(2): e08950.
23. Chon JW, Lee UJ, Bensen R, West S, Paredes A, Lim J, et al. Virulence Characteristics of *mecA*-Positive Multidrug-Resistant Clinical Coagulase-Negative Staphylococci. *Microorganisms* 2020; 8: 659.
24. Chapin KC, Musgnug MC. Evaluation of penicillin binding protein 2a latex agglutination assay for identification of methicillin-resistant *Staphylococcus aureus* directly from blood cultures. *J Clin Microbiol* 2004; 42: 1283-1284.
25. Koksal F, Yasar H, Samasti M. Antibiotic resistance patterns of coagulase-negative staphylococcus strains isolated from blood cultures of septicemic patients in Turkey. *Microbiol Res* 2009; 164: 404-410.
26. Asante J, Amoako DG, Abia ALK, Somboro AM,

Govinden U, Bester LA, et al. Review of clinically and epidemiologically relevant coagulase-negative Staphylococci in Africa. *Microb Drug Resist* 2020; 26: 951-970.

27. Khan MM, Faiz A, Ashshi AM. Clinically significant Coagulase Negative Staphylococci and their antibiotic resistance pattern in a tertiary care hospital. *J Pak Med Assoc* 2014; 64: 1171-1174.
28. Nicolosi D, Cina D, Di Naso C, Dangeli F, Salmeri M, Genovese C. Antimicrobial resistance profiling of coagulase-negative staphylococci in a referral center in South Italy: A surveillance study. *Open Microbiol J* 2020; 14: 91-97.
29. Ashagrie D, Genet C, Abera B. Vancomycin-resistant enterococci and coagulase-negative staphylococci prevalence among patients attending at Felege Hiwot Comprehensive Specialized Hospital, Bahir Dar, Ethiopia. *PLoS One* 2021; 16(4): e0249823.