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Environmental Screening of Organisms from High-Touch Areas of Critical Care Units in a Tertiary Care Hospital, Amalapuram - The Silent Invaders

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INTRODUCTION:

Environmental cleaning has long been identified as an important measure in prevention of Health care Associated Infections(HAI)[1.] HAI's pose a significant challenge to patient safety, particularly in critical care departments, where patients are highly susceptible to infections due to their critical condition and invasive procedures. The high-touch surfaces in these units have the potential to harbour harmful organisms, which could aid in the spread of illnesses. In recent years, the relationship between hospital environmental pollution and nosocomial infections has received increasing attention. Failure to maintain a clean environment also enhances the risk of multidrug resistant organisms (MDRO's) transmission, which pose a great challenge in terms of hospital infection control practices.[2] The risk of healthcare-associated infections is estimated to be two to twenty times higher in developing countries compared to resource-rich countries, with over 25% of patients affected[3].

In these settings, high-touch surfaces frequently contacted by healthcare personnel, patients and visitors, serve as reservoirs for pathogenic microorganisms[4]. These surfaces, including bed rails, door knobs/handles, medical equipments, washing sinks and light switches are pivotal points for the potential transmission of infectious agents. Hence, environmental screening of high-touch areas in all critical care departments is essential for identifying the presence of microorganisms, including multi-drug resistant organisms.

Objectives:

- To identify the microorganisms present on high-touch surfaces in critical care departments..
- To recommend measures to prevent environmental surface transmission of health care associated pathogens.

MATERIALS AND METHODS :

Study Design : A cross-sectional study

Study Setting : All critical care departments, including medical, surgical, neonatal, paediatric, cardiac and post operative wards and the Department of Microbiology, Konaseema Institute of Medical Sciences & RF, Amalapuram, Andhra Pradesh.

Duration : From January 2024 to December 2024

Sample collection method:

- After obtaining approval from Institutional Ethics Committee, samples were collected from high-touch areas including patient immediate environment like bed side rails, side table, trays, trolleys; commonly used surfaces like door knobs/ handles, light switches, washing sink; medical equipments like IV stands, BP apparatus, Stethoscopes, from all critical care units like medical, surgical, neonatal, paediatric, cardiac ICUs and post-operative ward.

- Sterile swabs moistened with sterile saline were used to swab approximately 10 cm^2 of each high-touch surface and were placed in a sterile tube and cultured on appropriate media and incubated at 37°C for 24-48 hours.
- Isolated colonies were identified using standard microbiological techniques, including gram stain, biochemical tests and Antibiotic susceptibility testing (AST) [6].
- Gram-negative bacteria were identified using a series of biochemical tests like oxidase test, catalase test, indole production, methyl red test, V-P test, citrate test, urease test, decarboxylases test, triple sugar iron agar test. On the other hand, Gram-positive bacteria were identified based on Gram reaction, hemolytic pattern, coagulase test and other tests. [7].
- Antibiotic susceptibility testing was performed using the Kirby-Bauer disk diffusion method according to CLSI guidelines[8].

DATA ANALYSIS:

- The prevalence of different microorganisms and their AST patterns were calculated and the data was analyzed using Statistical Package for the Social Sciences (SPSS®) software version 24

Conclusion:

The implications of environmental screening of high-touch areas of all critical care units will be significant in identifying prevalent microorganisms, including MDROs and can inform targeted cleaning and disinfection protocols. This information will be valuable to bridge the gap in existing knowledge and provide evidence-based recommendations to enhance patient safety and reduce infection rates[9]. Enhanced infection control practices based on the results can improve patient outcomes, help in reducing the risk of healthcare-associated infections (HAIs), decrease the spread of resistant pathogens and promote a safer hospital environment.

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Evidence that high-touch surfaces will work as an extra source of microbial pathogens accumulated over the years, e.g., several microorganisms can survive on medical equipment for hours to months, improved cleaning and disinfection of surfaces decrease the rate of HAI, and hospital environmental screening results and the study of clonal outbreaks, all have given support to the role of contaminated high-touch surfaces in the transmission of pathogens between patients and healthcare personnel[5].

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- Sterile swabs moistened with sterile saline were used to swab approximately 10 cm^2 of each high-touch surface and placed in a sterile tube and cultured on Mac-conkey and blood agar media and incubated at 37°C for 24-48 hours.
- Isolated colonies were identified using standard microbiological techniques, including gram staining, biochemical tests and Antibiotic susceptibility testing (AST)[6].
- Gram-negative bacteria were identified using a series of biochemical tests like oxidase test, catalase test, indole production, methyl red test, V-P test, citrate test, urease test, decarboxylases test, triple sugar iron agar test. On the other hand, Gram-positive bacteria were identified based on Gram reaction, hemolytic pattern, coagulase test [7].
- Antibiotic susceptibility testing was performed using the Kirby-Bauer disk diffusion method according to CLSI guidelines[8].

RESULTS :

From total 230 samples collected from various high touch surface areas of all critical units processed, 81 samples (35.2 %) were culture positive and showed growth. Among these 81 culture positives, gram positive cocci predominanted over gram negative bacilli. Gram positive cocci were 49 (60.5%) and gram negative bacilli were 32 (39.5 %).

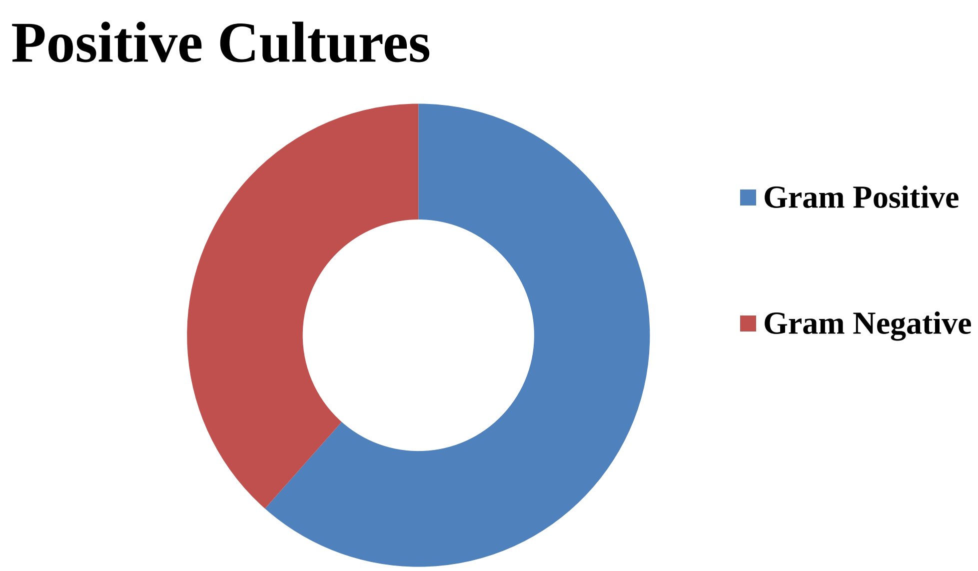

Table: 1

Gram positive cocci		
Organism	**No.of isolates**	**%**
CONS	34	41.9 %
S.aureus	15	18.5 %
Total	49	**60.5** %

Among49 isolates of gram positive cocci, CONS were 34 (41.9 %) and Staphylococcus aureus (S.aureus) isolates were 15 (18.5 %)

Table :2

Gram negative bacilli		
Organism	**No.of isolates**	%
Acinetobacter	9	11.0 %
Klebsiella	9	11.0 %
Pseudomonas	6	7.4 %
Bacillus sps	5	6.1 %
Enterobacter	2	2.5 %
Citrobacter	1	1.23%
Total	32	**39.5** %

Among 32 isolates of gram negative bacilli, *Acinetobacte*r were 9 (11%), followed by *Klebsiella* 9 (11 %), *Pseudomonas* 6 (7.4 %), *Bacillus* species 5 (6.1%), *Enterobacter* 2 (2.5%) and *Citrobacter* was 1 (1.23%).

Table :3 - Proportion of bacterial isolates from various high touch areas :

S.No	**Sampling Site**	**Organisms Isolated**
1	Bed Side Rail	S.aureus(2), CONS(6), Klebsiella (2)
2	Side tables	S.aureus(3), CONS(4), Acinetobacter (2),Citrobacter (1)
3	Trays	CONS (2), Acinetobacter (1), Bacillus spp (1)
4	Trolleys	S.aureus (2), CONS (2)
5	Door knobs/handles	S.aureus (3), CONS(6), Acinetobacter(3), Pseudomonas (2)
6	Light switches	Klebsiella (2), CONS (3), Enterobacter (2)

7	Washing sinks	CONS (5), Klebsiella (3), Pseudomonas (4),Bacillus (4)
8	IV Stands	CONS (2), Acinetobacter (2)
9	BP Apparatus	CONS (2), Klebsiella (2),S.aureus (3)
10	Stethoscopes	CONS(2), S.aureus (1), Acinetobacter (1)

From 230 swabs taken different high touch areas, the proportion of *CONS* isolated was 6-from bed side rail, 4 from side tables, 2 from trays, 2 from trolleys, 6 from door handles, 3 from light switches, 5 from washing sinks, 2 each from IV stands, BP apparatus and stethoscopes. *Staphylococcus aureus* isolated was 2 from bed side rail, 3 from side tables, 2 from trolleys, 3 from door knobs/handles, 5 from washing sinks, 2 from BP apparatus and 1 from stethoscopes. *Acinetobacter* isolated was 2 from side tables, 1 from tray, 3 from door knobs, 2 from IV stands, and 1 from stethoscope. *Klebsiella* isolated was 2 from bed side rail, 2 from light switches, 3 from washing sinks and 2 from BP apparatus. *Pseudomonas* isolated was 2 from door knobs and 4 from washing sinks. *Bacillus spp* isolated was 4 from light switches and 1 from tray. 2 isolates of **Enterobacte**r from light switches, 1 isolate of *Citrobacter* from side table.

Table :4 - Proportion of bacterial isolates from different ICU wards

S.No	Bacteria	SICU	MICU	NICU	PICU	Post Operative	CICU
1	CONS	8	6	2	4	10	4
2	S.aureus	5	2	0	0	6	2
3	Acinetobacter	3	1	0	1	2	2
4	Klebsiella	3	2	1	0	3	0
5	Pseudo	2	1	0	0	2	1
6	Bacillus spp	0	2	0	0	1	2
7	Enterobacter spp	0	2	0	0	0	0
8	Citrobacter	1	0	0	0	0	0

In the present study, the highest bacterial isolates were detected from surgical wards- post operative (24) and SICU (22) followed by MICU (16), CCIU (11), PICU (5) and NICU (3).
The frequency of Gram-positive bacteria was 16 in post operative ward, 13 in SICU, 8 in MICU, 6 in CICU, 4 in PICU, 2 in NICU. The predominant Gram-positive bacteria was *CONS* (34) followed by *S.aureus* (14). On the other hand, the proportion of the Gram-negative bacteria was 9 in SICU, 8 in MICU, 8 in post operative ward, 5 in CICU, 1 each from NICU and PICU. The predominant Gram-negative bacteria isolated were *Acinetobacter* (9), followed by *Klebsiella* (9), *Pseudomonas* (6), *Bacillus spp* (5), *Enterobacter* (2), *Citrobacter* (1).

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Colour coding in antibiogram

colour	Sensitivity % value	indicates
Green	> 80%	Drugs that may be reasonable choices for empirical therapy
Yellow	60-80%	Drugs that might be selected for empirical therapy in specific circumstances such as OPD setting, stable patient, etc…
Red	<60%	Drugs that may not be of reasonable choices for empirical therapy
Gray	Data not available	Includes : Intrinsic resistance, no breakpoint available, Clinically ineffective, drugs not tested and not routinely used.

Antimicrobial susceptibility profile of gram-negative bacteria isolates

Organism	Number of isolates	FIRST LINE DRUGS						2ND LINE					RESERVED	
		Amoxiclav	Ampicillin sulbactum	Ceftazidime	Ceftriaxone	Levofloxacin	Tetracycline	Gentamycin	Cotrimoxazole	Piperacillin tazobzctum	Cefperazone sulbactum	Ceftazidime- avibactum	Meropenem	Minocycline
Acenitobacter	9	IR	48%	11%	12%	25%	40%	31%	68%	40%	96%	47%	37%	41%
Klebsiella	7	IR	31%	26%	19%	37%	52%	64%	53%	55%	68%	57%	68%	58%
Pseudomonas	5	IR	21%	41%	IR	51%	49% (IR)	29% (IR)	73%	70%	74%	53%	57%	80%
Enterobacter	2													
Citrobacter	1													

Klebsiella is <60% sensitive to all the first line antibiotics like amoxiclav, ampicillin-sulbactum, 1st to 3rd generation cephalosporins, fluoroquinolones like levofloxacin; <60% sensitive to 2nd line antibiotics like cotrimoxazole, Piperacillin-tazobactum and ceftazidime avibactum. It is 60-80% sensitive for 2nd line antibiotics like gentamicin, cefperazone- sulbactum, and also 60-80% sensitive to reserved drugs like carbapenems, and Minocycline. *Acinetobacter* is <60% sensitive to all the first line antibiotics like amoxiclav, ampicillin-sulbactum, 1st to 3rd generation cephalosporins, levofloxacin, cotrimoxazole and tetracycline; <60% sensitive to 2nd line antibiotics like piperacillin-tazobactum, ceftazidime- avibactum, cefperazone -sulbactum and reserved group meropenem. 60-80% sensitive to 2nd line antibiotic aminoglycosides – gentamicin and minocycline and 96% sensitive to tobramycin. *Pseudomonas* is <60% sensitive to all the first line antibiotics like amoxiclav, ampicillin-sulbactum, 1st to 3rd generation cephalosporins, levofloxacin, cotrimoxazole and tetracycline; <60% sensitive to 2nd line antibiotics ceftazidime- avibactum, cefperazone -sulbactum and and reserved group minocycline. 60-80% sensitive to 2nd line antibiotics – gentamicin and piperacillin-tazobactum and tobramycin and 80% sensitive to carbapenems

Antimicrobial susceptibility profile of gram-positive isolates

organism	No.of isolates	Ampicillin	Amoxiclav	Azithromycin	Ceftriaxone	Cefotaxime	Ciprofloxacin	Clindamycin	Cefoxitin	Gentamycin	levofloxacin	High level gentamycin	Co-trimoxazole	Doxycycline	Vancomycin	Linezolid	Minocycline	Teicoplanin
		FIRST LINE DRUGS											2ND LINE			RESERVED		
S.aureus	484	21 %	58 %	28 %	35 %	31 %	25 %	58 %	16 %	70 %	30 %	-	70%	80 %	83%	95%	85 %	86%
CONS	141	-	52 %	36 %	42 %	36 %	25 %	54 %	12 %	74 %	-	-	-	78 %	66%	97%	52 %	-

Staphylococcus aureus is <60% sensitive to all the first line antibiotics like: ampicillin, amoxiclav, azithromycin, 1st to 3rd generation cephalosporins, levofloxacin;

60-80% sensitive to first line antibiotics like gentamycin and co-trimoxazole.

> 80 % sensitive to 1st line antibiotics Doxycycline and 2nd line antibiotics Vancomycin and Linezolid and reserved drugs minocycline and Teicoplanin. In *CONS*, susceptibility pattern is similar to S.aureus except doxycycline which is less sensitive i.e 60-80%.

DISCUSSION

In hospitals, high-touch surfaces can harbour pathogens responsible for nosocomial infections, leading to a greater risk of contamination among at-risk patients[10,11]

In our study, the overall culture positivity was 35.2% which is similar to the study conducted by Hammuel C, Jatau ED, Whong et al (2014)[12] and Getachew H, Derbie A, Mekonnen D (2018)[13]where the culture positivity was 39.4% and 46.3% respectively.

In other studies done by Mbanga J, Sibanda A, Rubayah S et al in 2018[14], and Lalami AEO, Touijer H, Ettayebi M eta al in 2016[15] , the culture positivity rate was high accounting for 86.2 % and 96.3% respectively which might be probably because of differences in hand hygiene, frequency of surfaces decontamination, the use of antiseptics, and disinfection techniques.

In the present study, isolation of gram positive bacteria (60.5%) predominated over gram negative bacteria (39.5%) which is correlating with the study of Tajeddin E, Rashidan M, Razaghi M et al[16] and Darge A, Kahsay AG, Hailekiros H et al[17], where the predominance of gram positive bacteria was 60.7% and 68.4% respectively. This predominance of gram positive bacteria may be because of transmission from the skin and nasal cavities of healthcare personnel and patients. However, studies conducted by Mbanga J, Sibanda A, Rubayah S [14] and Lalami AEO, Touijer H, Ettayebi M [15] reported Gram-negative bacteria as the predominant isolates. These variations may be attributed to differences in the study period, hospital settings, and the presence of patients already colonized or infected in the ward.
Among different wards, the highest numbers of bacteria were recovered from the surgical ward – SICU (22 isoaltes) and post-operative ward (24 isolates), followed by other medical ICU wards. Similar findings were found in a study done by Firesbhat, A., Tigabu, A., Tegene, B. et al[18] and Getachew H, Derbie A, Mekonnen D et al[13]. It may be due to high and unrestricted human trafficking.
In our study, the highest bacterial isolates were recovered from washing sinks(16) and door knobs/handles.(14), followed by side tables (11). But in the study by Getachew H, Derbie A, Mekonnen D et al[13] and Bakkali M, Hmid K, Kari K, Zouhdi M et al[19], most of the isolates were recovered from the bedside tables and bedsheets. This variation may be due to differences in hospital cleaning protocols, frequency of surface contact, and environmental factors. Moreover, sinks and door handles are high-touch surfaces often exposed to moisture, making them potential reservoirs for bacterial contamination. In contrast, in other settings, bedside tables and bedsheets, which are in prolonged contact with patients, cross-contamination from a patient's flora, health care workers' hands, and contamination during the washing process may serve as primary sources of bacterial transmission.
In our study, *CONS* was the most frequently isolated bacteria, 34 (41.9%) followed by *S. aureus*- 15 (18.5%), which correlates with the study of Firesbhat, A., Tigabu, A., Tegene, B. et al[18] *S.aureus* constitutes the normal human skin and mucous membranes flora and they are regularly shed onto the hospital environment by patients and medical personnel[20].
Majority of the isolates of *CONS* showed susceptibility to doxycycline (78 %), clindamycin (74%), gentamicin (54 %), and this is in align with a study conducted by Getachew H, Derbie A, Mekonnen D et al[13] and Endalafer N et al[21] . On the other hand, majority of *S. aureus* isolates were susceptible to doxycycline (80 %), gentamicin (70%), and cotrimoxazole (70 %). This finding correlates with the study of Getachew H, Derbie A, Mekonnen D et al[13] and Endalafer N et al[21] . Most of the *klebsiella* isolates showed susceptibility to cefaperazone-sulbactum (68 %), gentamicin (64%), but in the study of Firesbhat, A., Tigabu, A., Tegene, B. *et al.* most of the *Klebsiella* isolates showed susceptibility to amikacin (90%), 70% each to gentamicin, cotrimoxazole, cefepime, imipenem, and 60% to ceftazidime. This variation may be due to differences in geographical location, local antibiotic prescribing practices, infection control measures, sample size, sources of isolates, and the time period during which the studies were conducted. Additionally, the emergence of resistant clones and the presence of resistance genes such as ESBLs or carbapenemases in different populations could contribute to the observed differences in susceptibility patterns.
In our study, *pseudomonas* and *acinetobacter* were resistant to most of the antibiotics except for cotrimoxazole and cefoperazone-sulbactum which coincides with the study of Getachew H, Derbie A, Mekonnen D et al[13].

CONCLUSIONS :

In the present study, various bacterial isolates were identified from different high touch areas of ICUs, potentially predisposing patients to healthcare-associated infections. *Coagulase-negative Staphylococci* (CoNS), *Staphylococcus aureus*; *Klebsiella and Acinetobacter* were the most frequently isolated bacteria from high-touch surfaces. These findings underscore the need for regular and rigorous sanitation and disinfection protocols, along with continuous surveillance and monitoring of bacterial contaminants and their antimicrobial resistance patterns on contact surfaces. Implementing such practices is essential to minimize the risk of cross-contamination and prevent the spread of resistant pathogens in healthcare settings.

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