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CHARACTERIZATION OF HOSPITAL ENVIRONMENTAL MICROFLORA AND THEIR ROLE IN HEALTHCARE-ASSOCIATED INFECTIONS: A REVIEW

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ABSTRACT

Healthcare-associated infections (HAIs) remain a leading contributor to patient morbidity, mortality, and healthcare costs worldwide, affecting an estimated 5% of hospitalized patients. Hospital environmental surfaces — bed rails, door handles, ICU equipment, operation tables, floors, and medical instruments — act as reservoirs that harbor and transmit pathogenic microorganisms. This review synthesizes a dissertation study conducted at a tertiary care hospital that characterized environmental microflora and evaluated their association with HAIs. Using a cross-sectional observational design, 150 environmental swabs were cultured, yielding a 73.3% culture-positivity rate; bacterial isolates (72.7%) outnumbered fungal isolates (27.3%), with Staphylococcus aureus (37.5%) and Candida species (50% of fungal isolates) predominant. Comparison of 110 environmental isolates against 120 clinical HAI isolates (urinary tract infection, surgical site infection, bloodstream infection, and ventilator-associated pneumonia) revealed substantial organism overlap — up to 90% concordance for coagulase-negative staphylococci — and a statistically significant association between environmental contamination and HAI occurrence (70.6% concordance, $\chi^2 = 12.5$, $p < 0.05$). Bed rails and ICU equipment showed the strongest site-specific correlation with HAI cases. These findings, discussed against the wider hospital-microbiome and infection-control literature, reinforce that environmental surfaces are not passive bystanders but active contributors to nosocomial pathogen transmission, and they underscore the continued need for rigorous environmental surveillance, enhanced disinfection protocols, and antimicrobial

stewardship in hospital settings.

KEYWORDS: Hospital Environmental Microflora; Healthcare-Associated Infections; Nosocomial Pathogens; Environmental Surveillance; Infection Control.

1. INTRODUCTION

Healthcare-associated infections (HAIs) are infections acquired during hospitalization or medical treatment that were neither present nor incubating at the time of admission. They represent one of the most persistent challenges to patient safety worldwide, with point-prevalence surveys estimating that approximately 5% of hospitalized patients acquire at least one HAI during their stay. Beyond their direct toll on morbidity and mortality, HAIs impose substantial and largely preventable costs on healthcare systems.

A growing body of evidence identifies the hospital built environment itself — not only direct patient-to-patient or patient-to-healthcare-worker contact — as a significant reservoir of pathogenic microorganisms. Surfaces, equipment, air, and water within hospitals host a diverse microbial community that includes both benign environmental organisms and opportunistic pathogens capable of surviving for extended periods outside a host. Understanding the composition of this hospital environmental microflora, and its relationship to clinically confirmed HAIs, is therefore central to designing effective infection prevention and control strategies.

This review synthesizes a dissertation study that characterized environmental microflora across multiple hospital departments and directly compared environmental isolates with isolates recovered from patients diagnosed with suspected HAIs. The aim is to situate the study's findings within the broader literature on hospital microbiomes, surface contamination, and infection control, and to draw out their practical implications for healthcare facilities.

2. Sources, Reservoirs, and Persistence of Hospital Environmental Microflora

Microbial contamination of the hospital environment originates from several converging sources. Patients themselves shed organisms through respiratory secretions, skin, wounds, and body fluids; healthcare workers can act as vectors via contaminated hands, clothing, and equipment; and high-touch inanimate surfaces — bed rails, door handles, tables, chairs, and shared medical devices — accumulate and retain pathogens that are subsequently transferred through contact. Invasive devices such as catheters, ventilators, and intravenous lines add a further route by which environmental organisms gain direct access to normally sterile body sites.

A defining feature of many nosocomial pathogens is their capacity to persist on dry inanimate surfaces for extended periods — in some cases, months. Organisms such as methicillin-resistant Staphylococcus aureus (MRSA), vancomycin-resistant enterococci (VRE), Clostridium difficile, Acinetobacter baumannii, and Pseudomonas aeruginosa are well documented in the literature for this environmental resilience, which allows them to function as ongoing reservoirs for transmission long after the index patient has been discharged. Consistent with this literature, certain hospital zones — patient beds and bed rails, bathrooms and sinks, medical equipment, staff resting areas, and even mobile phones and protective clothing — have been repeatedly identified as contamination hotspots warranting targeted surveillance and cleaning.

3. Antimicrobial Resistance in Hospital Microflora

Perhaps the most consequential feature of hospital environmental microflora is the frequent presence of antimicrobial-resistant organisms. Multidrug-resistant organisms — including MRSA, carbapenem-resistant Klebsiella pneumoniae, Acinetobacter baumannii, and extended-spectrum beta-lactamase (ESBL)-producing Enterobacteriaceae — have been documented circulating among patients, healthcare workers, and environmental surfaces, with resistance genes capable of horizontal transfer within these reservoirs. The clinical consequence is that infections originating from environmental exposure are disproportionately likely to be difficult to treat, reinforcing the importance of environmental surveillance as an early-warning system for resistant organisms rather than a purely academic exercise.

4. Study Design and Methods

The reviewed study was conducted as a cross-sectional observational investigation across multiple hospital departments, including general wards, the Intensive Care Unit (ICU), the operation theatre, the outpatient department, and the laboratory section, over a two-month period. A total of 150 environmental swab samples were collected from six high-contact sites — bed rails, door handles, ICU equipment, operation tables, floors, and medical instruments — using sterile cotton swabs moistened with normal saline and a standardized 5 cm × 5 cm sampling area.

Samples were cultured on Nutrient Agar, Blood Agar, MacConkey Agar, and Sabouraud Dextrose Agar, with bacterial cultures incubated at 37°C for 24–48 hours and fungal cultures at 25–28°C for 3–5 days. Isolates were identified through colony morphology, Gram staining,

standard biochemical tests (catalase, coagulase, and oxidase for Gram-positive organisms; indole, citrate utilization, urease, and triple sugar iron testing for Gram-negative organisms), and, for fungi, Lactophenol Cotton Blue staining. In a linked analytical component, the study also collected 100–150 clinical samples (blood, urine, wound swabs, and respiratory specimens) from patients with suspected HAIs, processed through the same identification pipeline, to allow direct comparison between environmental and clinical isolates and statistical testing (chi-square) of the association between environmental contamination and HAI occurrence.

5. Key Findings

5.1 Overall Microbial Contamination

Of the 150 environmental samples collected, 110 (73.3%) yielded positive microbial growth. Among these positive isolates, bacteria predominated (72.7%) over fungi (27.3%). Site-wise positivity was highest at bed rails (83.3%) and ICU equipment (75%), and lowest at medical instruments (64%), a pattern broadly consistent with these sites' frequency of hand contact and proximity to patients.

Sampling Site	Positive Samples (n)	Positivity Rate (%)
Bed rails	25	83.3
ICU equipment	15	75.0
Floors	22	73.3
Door handles	18	72.0
Operation tables	14	70.0
Medical instruments	16	64.0

5.2 Distribution of Isolates

Among the 80 bacterial isolates, Staphylococcus aureus was the single most frequent organism (37.5%), followed by coagulase-negative staphylococci (22.5%), Escherichia coli (12.5%),

Klebsiella pneumoniae (10%), Pseudomonas aeruginosa (8.75%), Acinetobacter species (6.25%), and Bacillus species (2.5%). Gram-positive organisms accounted for 62.5% of bacterial isolates overall. Among the 30 fungal isolates, Candida species were most common (50%), followed by Aspergillus (26.7%), Penicillium (16.7%), and Cladosporium (6.6%).

5.3 Environmental–Clinical Isolate Comparison

Among 120 clinical HAI cases, urinary tract infection was the most frequent presentation (33.3%), followed by surgical site infection (25%), bloodstream infection (20.8%), and ventilator-associated pneumonia (20.8%). When the organisms recovered from these clinical

cases were compared with the 110 environmental isolates, substantial concordance emerged for several species: coagulase-negative staphylococci matched at 90%, Staphylococcus aureus at 85.7%, and Candida species at 66.7%, with somewhat lower but still notable concordance for the Gram-negative organisms (50–58.3%).

Organism	Clinical Isolates (n)	Environmental Isolates (n)	Match (%)
Coagulase-negative staphylococci	20	18	90.0
Staphylococcus aureus	35	30	85.7
Candida spp.	10	15	66.7
Pseudomonas aeruginosa	12	7	58.3
Escherichia coli	18	10	55.5
Klebsiella pneumoniae	15	8	53.3
Acinetobacter spp.	10	5	50.0

Statistically, 85 patients had documented exposure to contaminated surfaces, of whom 60 harbored isolates matching the corresponding environmental sample — a 70.6% association, confirmed as statistically significant by chi-square testing ($\chi^2 = 12.5$, $p < 0.05$). Site-specific correlation with HAI cases was highest for ICU equipment (86.7%) and bed rails (80%), and lowest for medical instruments (62.5%), broadly mirroring the site-wise contamination pattern observed in the environmental-only analysis.

6. DISCUSSION

The reviewed study's overall culture-positivity rate (73.3%) and the predominance of bacterial over fungal isolates are consistent with the broader hospital-microbiome literature, which has repeatedly identified high-touch inanimate surfaces as active microbial reservoirs rather than incidental contact points. The prominence of Staphylococcus aureus aligns with a well-established body of work implicating this organism in pneumonia, bloodstream infections, and surgical site infections; its frequent co-occurrence with coagulase-negative staphylococci — typically regarded as normal skin flora but capable of opportunistic infection in patients with indwelling devices — mirrors findings from ICU-focused surface-contamination studies such as those examining Gram-negative bacterial burden and Clostridium difficile transmission in intensive care settings.

The recovery of Gram-negative organisms such as Pseudomonas aeruginosa, Klebsiella pneumoniae, and Acinetobacter species is particularly noteworthy given their well-documented association with severe, often antimicrobial-resistant infections and their capacity for prolonged environmental survival, a pattern previously described for Acinetobacter species surviving on dry surfaces under varying humidity conditions. The

fungal findings — Candida and Aspergillus predominating — likewise echo prior work identifying these genera as recurrent contaminants of critical care environments, with particular relevance for immunocompromised patients.

The study's most clinically significant contribution is its direct, paired comparison of environmental and clinical isolates, which found substantial organism-level concordance (up to 90% for coagulase-negative staphylococci) and a statistically significant association between environmental exposure and HAI occurrence. This finding is consistent with, and adds site-specific, quantified support to, the broader theoretical model in which contaminated surfaces act as an intermediate link in a transmission chain — from patient or environmental source, to surface, to healthcare worker hands, to the next patient — a model well described in prior surface-contamination and hospital-microbiome research. The particularly strong correlation observed for ICU equipment and bed rails is consistent with these being high-acuity, high-touch zones where the consequences of contamination are most severe.

The study's finding that cleaning and disinfection measurably reduced microbial load, while some organisms nonetheless persisted afterward, reflects a tension long noted in the infection-control literature: routine cleaning protocols reduce but do not eliminate environmental bioburden, particularly for organisms capable of biofilm formation or unusual environmental resilience. This reinforces calls in the literature for enhanced disinfection technologies (e.g., ultraviolet or hydrogen peroxide vapor systems) and continuous, rather than one-time, environmental surveillance.

7. Strengths and Limitations

7.1 Strengths

- Paired sampling design linking environmental and clinical isolates within the same institutional setting, enabling direct, quantified comparison rather than relying on separately published literature for context.
- Broad sampling across multiple high-risk hospital departments (ICU, operation theatre, general wards, OPD), capturing site-specific variation in contamination and its clinical correlation.
- Use of standard, internationally recognized microbiological identification methods (culture, Gram stain, biochemical panels, LPCB staining for fungi), supporting comparability with other institutional studies.
- Statistical testing (chi-square) of the environmental–HAI association, moving beyond descriptive concordance to a formally significant result ($p < 0.05$).

7.2 Limitations

- The study is single-center and cross-sectional, limiting generalizability to other facilities and precluding causal or temporal conclusions about directionality of transmission (environment-to-patient versus patient-to-environment).
- Organism identification relied on phenotypic methods (culture, biochemical tests) rather than molecular typing (e.g., pulsed-field gel electrophoresis or whole-genome sequencing); as such, "matches" between clinical and environmental isolates reflect species-level concordance rather than confirmed strain-level transmission.
- The two-month study duration is relatively short, and seasonal or temporal variation in hospital microbiota — documented elsewhere in the literature — could not be captured.
- Antimicrobial susceptibility testing of the isolates does not appear to have been performed or reported, which limits the study's ability to directly characterize the resistance profile of the environmental reservoir it describes.

8. Implications for Infection Control Practice

These findings carry direct implications for hospital infection prevention programs. First, the disproportionate contamination and HAI-correlation observed at bed rails and ICU equipment support prioritizing these sites for high-frequency cleaning, dedicated disinfection protocols, and possibly point-of-care rapid testing during outbreak investigations. Second, the persistence of organisms after cleaning underscores that environmental hygiene should be treated as a continuous surveillance activity rather than a periodic audit, with routine microbiological monitoring used to verify — not merely assume — disinfection efficacy. Third, given the well-documented capacity of hospital environmental organisms to harbor antimicrobial resistance, integrating environmental surveillance data with antimicrobial stewardship programs would allow facilities to detect emerging resistant reservoirs before they translate into clinical outbreaks. Finally, targeted staff training on hand hygiene and equipment decontamination — particularly in high-acuity units such as the ICU and operation theatre — remains a foundational, low-cost intervention supported by this study's site-specific findings.

9. CONCLUSION

This review has synthesized a dissertation study that characterized hospital environmental microflora and quantitatively linked it to healthcare-associated infections. The study found substantial microbial contamination of hospital surfaces (73.3% culture positivity), a

bacterial-dominant flora led by Staphylococcus aureus, and a statistically significant, site-specific correlation between environmental contamination and HAI occurrence. These findings reinforce a growing consensus in the literature that the hospital built environment functions as an active reservoir and transmission route for nosocomial pathogens, not merely a passive backdrop to patient care. Sustained investment in environmental surveillance, evidence-based disinfection protocols, and antimicrobial stewardship — particularly targeted at high-contact, high-acuity sites such as bed rails and ICU equipment — remains essential to reducing the burden of healthcare-associated infections and safeguarding patient safety.

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