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ASSESSMENT OF MICROBIAL CONTAMINATION OF INANIMATE OBJECTS IN CLINICAL ENVIRONMENT

By

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Abstract

Patient-care equipment and inanimate objects contaminated with bacteria and fungi are a persistent problem in countries like Nigeria, and remain overlooked. This study aimed to elucidate the magnitude of contaminations, diversity, and antimicrobial-susceptibility patterns of bacterial isolates from selected wards of State Hospital in Ota, Ogun State, Nigeria. Samples were collected using a sterile swab moistened with sterile normal saline and inoculated into bacteriological media and identified by biochemical characterization, followed by antimicrobial-susceptibility tests of the bacterial isolates using agar diffusion method. Of the 63 bacterial isolates from the inanimate objects and items of patient-care equipment examined, Gram-positive bacteria were 52 representing 82 % while Gram-negative bacteria were 11 representing 18 %. Among the Gram-positive bacteria isolated, Staphylococcus spp were predominant followed by Bacillus spp and Micrococcus spp. whereas common Gram-negative counterparts were Pseudomonas spp., Enterobacter spp., Neisseria spp. and Klebsiella spp. The fungal isolates were commonly Aspergillus spp., Penicillium spp. and Cuvulleria spp. Antibiograms of S. aureus and Bacillus species showed high resistance against penicillin (Augmentin). Isolates of Neisseria spp., Enterobacter spp and Pseudomonas showed 80 % resistance to Imipenem and ampicillin, whereas those of Klebsiella spp. displayed high resistance against ampicillin and nalidixic acid. Overall prevalence of multidrug-resistant bacteria was observed especially for augmentin both in Gram-positive and Gram-negative bacterial isolates. Therefore based on the obtained results, a stringent infection-vigilance program comprising routine sampling from equipment and inanimate objects combined with antimicrobial-resistance surveillance and decontamination efforts must be instituted promptly to cub the prevalence of hospital acquired infection.

Keywords: *Antibiogram, Antibiotics, Inanimate objects, Clinical Environment, Bacteria*

1.0 Introduction

Microbial contamination of inanimate objects in clinical environments is a widespread and persistent problem that poses a serious threat to patient health and safety in healthcare settings across the globe. Hospital-acquired infections (HAIs) are a major public health concern, impacting millions of patients each year and leading to substantial morbidity, mortality, and increased healthcare costs (Christenson et al., 2021). The World Health Organization (WHO) estimates that HAIs affect approximately 10 % of patients in developed countries, with the prevalence reaching alarming rates of up to 40 % in developing countries (WHO, 2021).

Inanimate surfaces within hospital environments are significant contributors to the emergence and transmission of hospital-acquired infections (HAIs) (Abourrichm, et al., 2025). HAIs are infections that develop in patients during the course of receiving medical care in a hospital, which were not present or incubating at the time of admission. These infections can arise during hospitalization or even after discharge (Leistner et al., 2023). In healthcare settings, inanimate surfaces—such as medical equipment, furniture, and door handles—are recognized as potential reservoirs for pathogens that can facilitate cross-contamination and trigger outbreaks (Jernigan *et al.*, 2017; Hassan et al., 2020). Bacteria possess a remarkable ability to colonize a wide range of

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surfaces, and numerous studies have demonstrated that they can survive for weeks on materials commonly used in hospital environments, including stainless steel and polymers (Hassan et al., 2020; Schmidt *et al.*, 2021; Abourrichm, et al., 2025).

The longer a hospital pathogen persists on a surface, the greater the likelihood it will be transmitted to vulnerable patients or healthcare professionals (Schmidt *et al.*, 2021). In the United States alone, hospital-acquired infections are responsible for an estimated 1.7 million infections and approximately 99,000 deaths annually, with associated healthcare costs ranging from USD 28 to 34 billion (Christenson *et al.*, 2021). Similarly, in Nigeria, the burden of hospital- acquired infections is considerable, with studies reporting prevalence rates ranging from 10 % to 30 % in various healthcare settings (Olowe, 2018). As a result, there is increasing recognition of the importance of cleaning and disinfecting environmental surfaces as a vital component of infection prevention and control programs (Jaouhar et al., 2020). This has led to a growing interest in novel technologies that can effectively disinfect healthcare surfaces. In recent years, various methods have been explored to enhance surface cleaning and disinfection efforts (Leistner et al., 2023). A critical review of literature, particularly in inpatient settings, reveals that non-critical hospital equipment and frequently used electronic devices—such as keyboards and wall-mounted phones—are often colonized by both pathogenic and non-pathogenic bacterial species (Abourrichm, et al., 2025; Schmidt *et al.*, 2021). Furthermore, contact with contaminated surfaces using clean or gloved hands can result in the transfer of organisms, which can subsequently be spread to other surfaces or individuals (Hassan et al., 2020).

Several pathogenic organisms are known to inhabit inanimate surfaces within healthcare environments. These include bacteria such as *Staphylococcus aureus*, *Escherichia coli*, and *Acinetobacter baumannii*, which are associated with a range of infections including skin infections, pneumonia, urinary tract infections, and bloodstream infections (Gebrezihier *et al.*, 2026; Bulwadda *et al.*, 2023; Zahornacký, *et al.*, 2022). Fungal pathogens such as *Candida albicans* and *Aspergillus* spp. are also commonly found on hospital surfaces and can cause respiratory and bloodstream infections, particularly in immunocompromised individuals. In addition to pathogenic organisms, non-pathogenic bacteria such as certain strains of *Lactobacillus* have also been identified on inanimate hospital objects (Saka et al., 2017).

Several factors contribute to the colonization and persistence of microorganisms on these surfaces. Surface characteristics, such as roughness or porosity, can promote microbial adhesion and survival. For example, organisms like *Staphylococcus aureus* and *Candida albicans* have been shown to persist for extended periods on rough or porous materials (Jabłońska-Trypuć et al., 2022; Yamaguchi *et al.*, 2011). Environmental conditions such as humidity and moisture also play a critical role. Microorganisms like *Pseudomonas aeruginosa* (Mahalingam *et al.*, 2021) and *Candida albicans* (Xu *et al.*, 2015) thrive in such conditions, further contributing to the risk of surface contamination and infection transmission. Despite significant advances in infection prevention and control (IPC) practices over the past few decades, microbial contamination of inanimate objects within healthcare environments remains an ongoing and complex issue. Furthermore, the challenge of controlling microbial contamination is compounded by the constant influx of patients, the high number of procedures and treatments performed, and the complexity of healthcare environments, where the need for constant sanitization and disinfection often competes with limited resources (Mahalingam *et al.*, 2021). Hence, the rise in healthcare-associated infections, particularly those caused by antimicrobial-resistant pathogens, continues to put immense pressure on healthcare systems worldwide. Therefore, the main objective of this study was to isolate and characterize microorganisms from inanimate objects around hospital environments.

2.0 MATERIALS AND METHODS

2.1 Sampling site and collection:

Samples were collected from the Ogun State Hospital, Ota Ogun State, Nigeria. Sterile swabs moistened by dipping into a sterile normal saline were used to collect microbial samples from various surfaces within the hospital environment which include the pediatric section, labor wards, general reception and dental wards. The surfaces sampled (approx. 10 cm × 10 cm) included frequently touched areas like chair arms, door handles, railings, stethoscopes and table surfaces. The swabbing was done by rubbing and rotating sterile swab stick that was dipped in sterile saline on the surfaces. After collection, each swab was immediately transferred into a sterile tube, labeled with location, time, and date, and transported to the laboratory within 2 hours using aseptic handling throughout.

2.2 Isolation and Identification of Bacterial Isolates:

Sample specimens collected were promptly inoculated onto nutrient agar, mannitol salt agar and MacConkey agar plates by rolling and rotating them on the surface. The inoculated plates were then labeled, inverted and incubated at 37 °C. Pure isolates were obtained by picking single pure colonies based on their morphological characteristics on the plates and were stored on sterile Mac Cartney bottles prior to identification and antibiotic sensitivity test.

Pure bacterial isolates were identified using API 20E and 20A test kits, which include 20 dehydrated biochemical substrates. A pure bacterial colony was suspended in sterile 0.85% saline to form a homogenous solution. Approximately 0.5 mL of this suspension was added to each chamber. Some wells were overlaid with mineral oil to create anaerobic conditions. The strips were incubated at 37°C for 18–24 hours. Color changes were either spontaneous or required reagent addition (e.g., for indole or nitrate tests). Results were recorded, scored, and analyzed against the API database to generate a unique 7-digit profile for species identification. The oxidase test result, forming the 21st parameter, was included to complete the profile.

Hemolytic activity of bacterial isolates was assessed by culturing them on blood agar plates containing 5 % blood. A

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clear, transparent zone around colonies indicated β-hemolysis (complete red blood cell lysis), a greenish or brownish discoloration was noted as α-hemolysis (partial lysis), and the absence of any visible change in the agar was interpreted as γ-hemolysis (no lysis). This test was used to further characterize the bacterial isolates, particularly in distinguishing pathogenic *Staphylococcus aureus*, which typically exhibits β-hemolysis.

3.2.8 Antibiotic Susceptibility Testing

Antibiotic susceptibility of pure bacterial isolates was assessed using the disc diffusion method on Mueller-Hinton agar as described by Obi et al. (2026). A bacterial suspension was evenly spread across the agar surface using a sterile swab. After allowing the plate to dry slightly, antibiotic discs specific to Gram-positive and Gram-negative bacteria were placed using sterile forceps. The plates were incubated at 37°C for 24 hours. Zones of inhibition were measured in millimeters (mm) to determine susceptibility. Gram-positive discs include; Cefazolin (30µg), Ofloxacin (5µg), Ciprofloxacin (5µg), Linezolid (30µg), Cefuroxime (30µg), Gentamicin (10µg), Tobramycin (10µg) while Gram-negative discs was made of Ofloxacin (5µg), Meropenem (10µg), Cefazolin (30µg), Cefuroxime (30µg), Ciprofloxacin (5µg) rounding the colonies.

RESULTS AND DISCUSSION

Microbial Distribution on Inanimate Objects Surfaces and Identification of Bacterial and Fungal Isolates

A total of 63 bacteria and six (6) fungi were isolated from different inanimate objects from the clinic. Out of the 63 bacterial isolates, 52 were Gram positive (82 %) and 11 were gram negative (18 %) (Fig.1). Fungal isolates identified microscopically include *Rhizopus sp*, *Penicillium sp*, *cuvularia sp*, and *Aspergillus sp*, which are known opportunistic pathogens in immune-compromised individuals (Table 1) while among gram-positive isolates the most frequently isolated species were *S. aureus* (22.1%), *Bacillus* species (36.1%), *Micrococcus*, *Lactobacillus* etc. On the hand, *Klebsiella* spp (18.4%), *E. coli* (14.1 %), *Neisseria* species, *Enterobacter*, *Pseudomonas* species predominantly *P. aeruginosa* were the most common gram-negative bacteria. Microscopic examination of the fungal isolates showed that three (3) of the six isolates had septate hyphae and conidia while the other three had non-septate phyhae (Plate 1).

This study explored the microbial contamination of inanimate surfaces within a hospital environment. Contaminated inanimate objects and patient-care equipment are proven sources of infections, and the bacteria can spread throughout hospital wards in an epidemic fashion. Among the eight stations/wards studied, the objects in the reception showed the highest degree of bacterial contamination, followed by the pediatric ward. Similar result was reported by Birru et al. (2021). In their report they examined three hospital wards and found that surgical and the pediatric ward had the highest microbial population. These findings confirm that surfaces such as chairs, beds, sinks, and tables serve as reservoirs for both bacterial and fungal pathogens, some of which are resistant to commonly used antibiotics. In all eight wards, patient-care equipment and inanimate objects directly or indirectly associated with patients were heavily contaminated with diverse species of bacterial pathogens. Of the isolates, Gram-positive cocci were the most dominant, and the extent of contamination was comparable to a number of studies reported from Ethiopia and Nigeria (Weldegebreal et al., 2019; Gelaw et al., 2014; Maryam et al., 2014). In their reports, they observed that Gram-positive bacteria, coagulase-negative staphylococci (CoNS; 52.2%) were predominant, followed by *Staphylococcus aureus* (47.7%), whereas common Gram-negative counterparts were *Acinetobacter* spp. (28.5%) and *Klebsiella* spp. (23.8%). In this study, the most frequently isolated bacteria were *Staphylococcus aureus, Escherichia coli, Klebsiella pneumoniae, and Pseudomonas aeruginosa* which are in agreement with their findings.

Isolation of *Staphylococcus* spp. from patient-care equipment and inanimate objects of all eight wards indicated their ubiquitous nature. This can be related to the fact that *Staphylococcus* spp. is members of the microbiome of both healthy as well as sick individuals. They can be dispersed widely through direct contact with contaminated inanimate objects or medical equipment and even by transient carriage on the hands of health-care workers. The results of this study also proved that Gram-negative bacilli colonized the surface of many inanimate objects and patient-care equipment in all three wards. These organisms are well-documented in the literature as major contributors to nosocomial infections (Gebrezihier et al., 2026; Bulwadda et al., 2023; Zahornacký, et al., 2022; Ahmed et al., 2019). Their presence on surfaces frequently touched by healthcare workers, patients, and visitors presents a direct threat to infection control in hospital settings. Notably, fungal isolates, such as *Candida albicans* and *Aspergillus spp*., though less frequently discussed in environmental monitoring, were also identified. These pathogens pose significant risks, particularly to immunocompromised patients, and their presence suggests a need to expand environmental surveillance beyond bacteria.

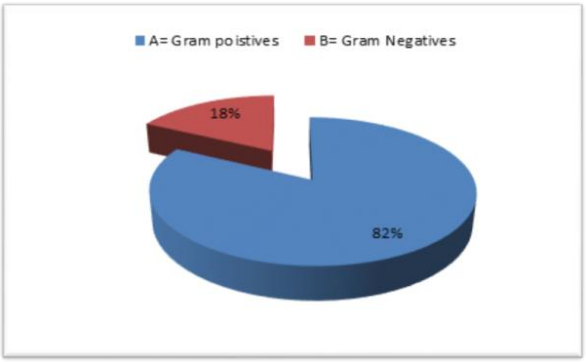


Figure 1: Distribution of isolated bacteria based on Gram reaction

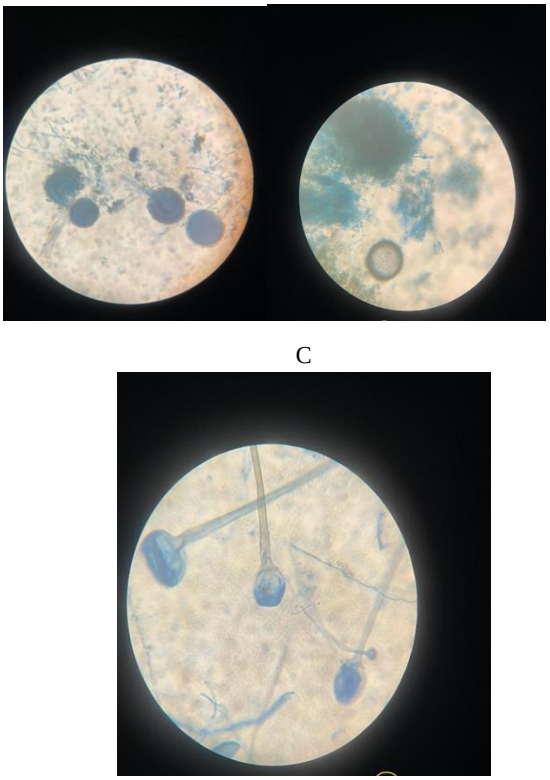
Table 1: Fungal isolates and their microscopic appearance

S/N	Fungal isolates	Microscopic appearance	Presumed organism
1	FWB	non-septate hyphae and conidia	*Aspergillus* sp.

2	LWB4	septate conidia and hyphae	*Cuvularia sp.*
3	LC1	septate conidia and hyphae	*Cuvularia*
4	LWBI	septate hyphae presence of phillide	*Penicillium* sp.
5	LWB2	non-septate hyphae and conidia	*Aspergillus* sp.
6	GWB	non-septate conidia and hyphae	*Rhizopus* sp.

Legend= FWB - female ward bed**;** LWB - labour ward bed**;** LCI - lab chair**;** LWB1- labour ward bed; LWB2 - labour ward bed 2**;** GWB – gynecology ward bed

A B



C

Plate 1: Microscopic appearance of the fungal isolates A= *Aspergillus sp.*; B=*Penicillum sp.*; C =*Rhizopus sp.*

Antibiotic Susceptibility Pattern of bacterial isolates

Results of the antibiotic susceptibility test are shown in figures 2 and 3. Out of the 52 Gram positive bacterial isolates, 42 of them were resistance to Augmentin while 33 were resistance to Carbapenem and Cephalosporins (Figure 2). Meanwhile, the bacterial isolates were highly susceptible to Chloramphenicol, Azithromycin, Vancomycin and Gentamicin. Similarly, results obtained with the antibiotic Gram-negative discs are shown Figure 3. Ten out of the 11 isolated Gram negative bacteria showed resistance to Imipenem, Amoxicillin while 9 showed resistance to Co-trimoxazole, Nalidixic acid. On the other hand, Gram negative bacterial isolates were susceptible to Gentamicin, Ofloxacin, Ciprofloxacin and Nitrofurantoin. *Staphylococcus* sp. and *Bacillus* sp. isolated from this study showed multiple resistance to 11 out of the 12 antibiotic discs tested while *Bacillus* species were resistance to 10 of the antibiotics discs for Gram positive bacterial isolates. Also, results obtained from the susceptibility test for Gram negative organisms showed that the isolates were resistant to multiple antibiotic discs (Figure 3).

The antimicrobial susceptibility test results showed high resistance to cephalosporins (e.g., *Cefuroxime* and *Cefazolin*), moderate resistance to fluoroquinolones like *Ciprofloxacin*, and retained susceptibility to newer or less commonly used antibiotics such as Linezolid, Meropenem, and Gentamicin. These patterns raise serious concerns, especially in resource-constrained healthcare systems where access to second-line drugs may be limited (Gebrezihier *et al.*, 2026; Khan et al., 2025). We also found that the most predominant Gram-positive isolate showed resistance to both penicillin and trimethoprim–sulfamethoxazole to a greater extent than the others. Similar phenomena have been observed in a number of studies reported from various regions of Nigeria (Mbanga *et al.*, 2018; Maryam *et al.*, 2014; Fagade *et al.*, 2010). Antimicrobial-susceptibility tests in this study revealed that bacterial isolates were resistant to multiple antibiotics, indicating a high risk of nosocomial outbreaks due to drug-resistant bacteria. Similar observation was reported by Gebrezihier *et al.* (2026) in their study carried out in Ethiopia. Therefore, stringent infection-prevention and control programs comprising routine sampling from patient-care equipment and inanimate objects among wards must be implemented, along with antimicrobial-resistance surveillance and decontamination efforts. Overall, the study highlights the importance of routine cleaning and disinfection of hospital surfaces, regular monitoring of microbial contamination, and strict adherence to infection prevention protocols.

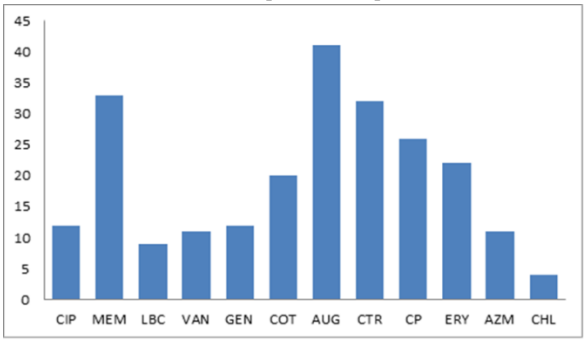


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Figure 2: Antibiotic resistant pattern of Gram positive bacterial isolates

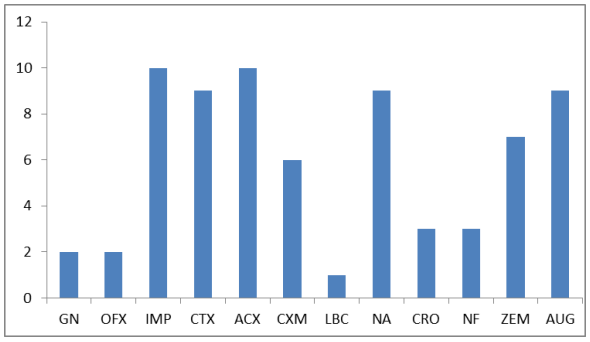

Figure 3: Antibiotic resistant pattern of Gram negative bacterial isolates

CONCLUSION

This study demonstrates that hospital surfaces, although seemingly clean, are often reservoirs for pathogenic microorganisms, including drug-resistant strains. This reality poses a silent but significant threat to patients and healthcare workers alike. The isolation of multidrug-resistant bacteria like *Pseudomonas aeruginosa* and *E. coli* from inanimate objects suggests that environmental hygiene alone is not enough. Instead, a more integrated approach is required, one that combines regular surveillance, rigorous disinfection, responsible antibiotic use, and staff education. In simpler terms, cleaning must go beyond what the eyes can see, and infection control must adapt to the evolving nature of microbial resistance.

Ethical Approval: This study did not involve human participants or animal experimentation and therefore did not require formal ethical approval. All procedures involving data handling and analysis were conducted responsibly and in accordance with standard academic and institutional guidelines.

Declaration of Conflict of Interest: The authors declare that no known conflict of interest or personal relationships that could have influence the work reported in this paper.

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