

Antimicrobial resistance in biofilms on invasive devices*

Resistência antimicrobiana em biofilmes formados em dispositivos invasivos

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ABSTRACT

Objective: to evaluate the microbiological profile and resistance patterns of biofilms on invasive devices recovered from Intensive Care Unit patients. **Methods:** a prospective cohort study of 157 adult patients in the immediate postoperative period. We analyzed 275 invasive devices collected under aseptic conditions and subjected to microbiological culture and susceptibility testing. Data were analyzed using Chi-square and Mann-Whitney tests. **Results:** biofilm formation occurred on the majority of devices, yielding 200 isolates, primarily *Klebsiella pneumoniae* (30%), *Acinetobacter baumannii* (25%), and *Pseudomonas aeruginosa* (20%). Multidrug-resistant (MDR) isolates accounted for 48% of the sample, while 19% were extensively drug-resistant (XDR) and 3% were pandrug-resistant (PDR). Central venous catheters showed a significantly higher rate of colonization by multidrug-resistant microorganisms ($p < 0.010$). This contrasted with findings for indwelling urinary catheters ($p = 0.072$) and endotracheal tubes ($p = 0.064$). **Conclusion:** the microbiological profile of biofilms on invasive devices consisted mainly of Gram-negative bacteria with broad-spectrum resistance to multiple antibiotics, negatively impacting patient safety and quality of care. **Contributions to practice:** these findings support the strengthening of preventive strategies and control of healthcare-associated infections, thereby promoting patient safety and quality of care in the Intensive Care Unit.

Descriptors: Biofilms; Equipment and Supplies; Cross Infection; Drug Resistance, Microbial.

RESUMO

Objetivo: avaliar o perfil microbiológico e os padrões de resistência em biofilmes formados em dispositivos invasivos em pacientes de uma Unidade de Terapia Intensiva. **Métodos:** estudo de coorte prospectivo com 157 pacientes adultos em pós-operatório imediato. Foram analisados 275 dispositivos invasivos, coletados em condições assépticas, submetidos a cultivo microbiológico e testes de suscetibilidade. Utilizou-se testes de Qui-quadrado e Mann-Whitney. **Resultados:** houve formação de biofilmes em grande parte dos dispositivos, com isolamento de 200 microrganismos, principalmente *Klebsiella pneumoniae* (30%), *Acinetobacter baumannii* (25%) e *Pseudomonas aeruginosa* (20%). Foram identificados 48% de isolados multirresistentes, 19% extensivamente resistentes e 3% pan-resistentes. Observou-se elevada taxa de cateter venoso central com maior proporção de colonização por microrganismos multirresistentes ($p < 0,010$). Em contraste com sondas vesicais de demora ($p = 0,072$) e tubos orotraqueais ($p = 0,064$). **Conclusão:** o perfil microbiológico em biofilmes de dispositivos invasivos incluiu principalmente bactérias gram negativas com amplo espectro de resistência a múltiplos antibióticos, impactando negativamente na segurança de paciente e qualidade de assistência. **Contribuições para a prática:** os resultados podem oferecer subsídios para o fortalecimento de estratégias preventivas e controle das infecções relacionadas à assistência à saúde, promovendo assim a segurança do paciente e qualidade de assistência prestada da unidade de terapia intensiva.

Descritores: Biofilmes; Equipamentos e Provisões; Infecção Hospitalar; Resistência Microbiana a Medicamentos.

Introduction

In recent decades, Healthcare-Associated Infections (HAIs) have emerged as a serious global public health issue, compromising both patient safety and the quality of care provided⁽¹⁾. These challenges stem from the high complexity of care, the intensive use of invasive devices, and the clinical severity of hospitalized patients, factors that favor severe infections, often caused by multidrug-resistant (MDR) microorganisms. These infections result in increased morbidity and mortality, prolonged hospital stays, and additional costs⁽²⁾.

Currently, antimicrobial resistance poses a serious public health challenge, increasingly prominent due to its potential to severely impair quality of care and patient safety. Antimicrobial resistance is defined as the ability of microorganisms, including bacteria, fungi, and parasites, to resist the effects of antimicrobial drugs such as antibiotics, antivirals, antifungals, and antiparasitics⁽³⁾.

The consequences of antimicrobial resistance include increased mortality risk, disability, the need for intensive care, prolonged hospitalization, and the use of alternative antibiotics, which are generally more expensive⁽⁴⁾.

In this context, the formation of microbial biofilms on the surfaces of invasive devices—such as central venous catheters (CVCs), urinary catheters, and endotracheal tubes, which are predominantly used in Intensive Care Units (ICUs)—is a significant factor contributing to the persistence and resistance of hospital infections⁽⁵⁾. Biofilms are characterized as structured communities of microorganisms attached to a surface and encased in a protective extracellular matrix, which confers increased resistance to antimicrobial agents and host defense mechanisms⁽⁶⁾.

Approximately 65% of healthcare-associated infections are linked to biofilm formation. The presence of biofilms on invasive devices is associated not only with increased length of stay and mortality but also with prolonged use of broad-spectrum antimicrobials; this drives the emergence of antimicrobial

resistance, further increasing mortality rates, treatment difficulty, hospital costs, and the risk of severe physiological harm⁽⁷⁾.

Microorganisms such as *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae* are frequently identified in these biofilms, demonstrating resistance to various antibiotic classes, including carbapenems, which are often considered treatments of last resort⁽⁸⁾.

Despite efforts to prevent and control infections related to antimicrobial resistance in device-associated biofilms, studies focusing on biofilm maturation remain crucial. Furthermore, understanding the microbiological profile of critically ill patients contributes to defining effective strategies for HAI prevention and control, thereby promoting patient safety.

Given this background, this study aimed to evaluate the microbiological profile and resistance patterns of biofilms on invasive devices in ICU patients.

Methods

Type of study

This was a prospective cohort study. This design was selected to enable temporal monitoring of patients undergoing invasive procedures, facilitating the identification of colonization and antimicrobial resistance associated with biofilm formation on medical devices.

Study locus

The study was conducted in the ICU of a public hospital in Northeastern Brazil.

Study population, inclusion and exclusion criteria

The target population consisted of 157 adult patients (≥18 years) admitted to the ICU in the immediate postoperative period. We analyzed 275 invasive devices—including central venous/double-lumen

catheters, indwelling urinary catheters, and endotracheal tubes—collected between March and July 2025, for which laboratory biofilm analysis results were available. We excluded patients undergoing surgical re-intervention, debridement, drainage, episiotomy, or biopsies not involving viscera or cavities, as well as those with a history of hospitalization in the preceding six months.

Data collection procedures

Eligible patients were monitored daily until device removal, discharge, or death. Clinical and epidemiological data were obtained from electronic medical records and reports from the HAI Control Service.

Device sampling was performed by the institution's laboratory technical team as part of integrated HAI surveillance. Samples were collected under aseptic conditions immediately upon removal, following institutional biosafety protocols. Fragments were transported to the microbiology laboratory in sterile media and processed within two hours. Notably, this procedure is part of the institution's routine workflow, with no interference from the researchers.

Device surfaces were subjected to sonication and culture techniques for bacterial recovery to identify biofilm formation. The 200 isolated microorganisms were identified using conventional methods and/or automated systems. Antimicrobial susceptibility profiles were determined via disk diffusion (Kirby-Bauer) or automated systems, following Clinical and Laboratory Standards Institute guidelines⁽⁹⁾. Resistance was categorized as multidrug-resistant (MDR), extensively drug-resistant (XDR), or pandrug resistant (PDR), according to European Centre for Disease Prevention and Control criteria⁽¹⁰⁾.

Study variables

The variables studied included age, sex, type of surgery, ICU length of stay, and comorbidities. Device-related variables included device type, dwell time,

number of exchanges, isolated microorganisms, presence of biofilm, and antimicrobial resistance profile (MDR, XDR, PDR).

Data analysis

Data were tabulated in Excel LTSC 2021 and analyzed using SPSS software, version 20.0. Descriptive analyses presented absolute frequencies and percentages, means and standard deviations, or medians and interquartile ranges (IQR), depending on variable distribution. Group comparisons were performed using Chi-square or Fisher's Exact tests for categorical variables, and Student's t-test or Mann-Whitney tests for continuous variables, based on data normality. Logistic regression models were used to identify factors associated with antimicrobial resistance in biofilms. The significance level was set at 5% ($p < 0.05$)⁽¹¹⁾.

Ethical aspects

All participants or their guardians were informed about the research objectives, the voluntary nature of participation, anonymity, and the right to withdraw at any time. Participation was formalized by signing an Informed Consent Form.

The research project was approved by the Research Ethics Committee of the Federal University of Alagoas, in compliance with National Health Council Resolution No. 466/2012 (Opinion No. 7.213.528/2024; Certificate of Presentation for Ethical Appreciation: 81348724.6.0000.0155).

Results

The study analyzed and monitored 157 patients in the immediate postoperative period in the ICU over 90 days. All patients had invasive devices placed either in the operating room or in the ICU. The sample revealed a high-risk clinical-epidemiological profile, predominantly male (58%), with a mean age of 61.8 years (± 15.2).

Regarding clinical and epidemiological characteristics, there was a male predominance (58%) and a high frequency of comorbidities, notably systemic arterial hypertension (62%) and diabetes mellitus (36%); chronic obstructive pulmonary disease (COPD) was less common (14%). The median length of hospital stay was 10 days (IQR: 6–18), indicating a population with a complex clinical profile and a significant burden of chronic disease (Table 1).

Table 1 – Clinical and epidemiological characteristics of patients (n=157). Maceió, AL, Brazil, 2025

Variables	n (%)	CI95%	z-test	p-value
Male gender	91 (58.0)	50.2 – 65.4	(vs 50%)	0,041*
Arterial hypertension	97 (62.0)	54.2 – 69.2	(vs 50%)	0,008*
Diabetes Mellitus	57 (36.0)	28.8 – 43.8	(vs 50%)	0,002*
Chronic Obstructive Pulmonary Disease	22 (14.0)	8.5 – 19.5	(vs 50%)	<0,001*
Mean age (61.8 ± 15.2)	-	-	(vs 50 years)	<0,001 [†]
Median length of stay (10; IQR 6–18)	-	-	(vs 7 days)	0,021 [‡]

*Z-test; [†]t-test; [‡]Mann-Whitney; IQR: Interquartilerange; CI: Confidence Interval

We analyzed 275 invasive devices inserted and subsequently removed in the adult ICU, including indwelling urinary catheters, central venous/double-lumen catheters, and endotracheal tubes. Central venous catheters accounted for the largest proportion (110; 40%), followed by indwelling urinary catheters (96; 35%) and endotracheal tubes (69; 25%). The majority of these devices exhibited bacterial biofilm formation, highlighting a significant risk for the development of HAIs.

Two hundred microorganisms were isolated from biofilm-bearing devices. *Klebsiella pneumoniae* (60; 30%) predominated, followed by *Acinetobacter baumannii* (50; 25%) and *Pseudomonas aeruginosa* (40; 20%). Other agents included *Staphylococcus aureus* (20; 10%), coagulase-negative *Staphylococci* (15; 7.5%), *Enterococcus spp.* (10; 5%), and less frequent agents (5; 2.5%), underscoring the relevance of multidrug-resistant Gram-negative bacilli in HAI prevention.

Of the 200 isolates, 48% displayed a multidrug-

-resistant (MDR) profile, 19% were classified as extensively drug-resistant (XDR), and 3% as pandrug-resistant (PDR). Only 30% remained susceptible, pointing to a concerning scenario of high antimicrobial resistance.

Regarding clinical and epidemiological characteristics, male patients were predominant (58%; p=0.041), suggesting greater vulnerability of this group to critical conditions requiring immediate postoperative ICU admission. The mean age was high (61.8 ± 15.2 years; p<0.001), reinforcing the predominance of elderly patients, a group associated with a higher risk of infectious complications.

The most frequent comorbidities were systemic arterial hypertension (62%; p=0.008) and diabetes mellitus (36%; p=0.002), both known to compromise immune response and wound healing. The median length of stay was 10 days (IQR: 6–18; p=0.021), which is considered prolonged and increases exposure to invasive devices and susceptibility to HAIs.

Hypertensive patients had a significantly longer length of stay (12 days; IQR 8–20) compared to non-hypertensive patients (8 days; IQR 5–15; p=0.030). Similar results were observed for diabetic patients (11 days; IQR 7–19 vs 9 days; IQR 6–16; p=0.048) and COPD patients (13 days; IQR 8–22 vs 9 days; IQR 6–16; p=0.044) (Table 2).

Table 2 – Distribution of microorganisms and resistance profiles (n=200). Maceió, AL, Brazil, 2025

Category	n (%)	CI95%	Proportion test (vs expected value)	p-value*
Gram-negative	150(75.0)	69.2–80.8	vs 50%	<0.001
Gram-positive	50(25.0)	19.2–30.8	-	-
Multidrug-resistant	96(48.0)	41.2–54.8	vs 30%	<0.001
Extensively drug-resistant	38(19.0)	13.5–24.5	vs 10%	0.005
Pandrug-resistant	6(3.0)	0.6–5.4	vs 0%	0.041
Susceptible/non-MDR	60(30.0)	23.4–36.6	vs 50%	<0.001

*Z-test; CI: 95% Confidence Interval; MDR: Multidrug-resistant

Analysis of the association between comorbidities and length of stay showed that patients with arterial hypertension had a longer duration of stay (me-

dian 12 days; IQR: 8–20) compared to those without the condition (8 days; IQR: 5–15; $p=0.030$). Similarly, patients with diabetes mellitus had longer hospitalizations (11 days; IQR: 7–19) than non-diabetics (9 days; IQR: 6–16; $p=0.048$). Furthermore, individuals with COPD had the longest length of stay (13 days; IQR: 8–22) compared to those without the disease (9 days; IQR: 6–16; $p=0.044$), indicating that these comorbidities are significantly associated with prolonged hospitalization.

There was a high rate of CVC utilization, representing 110 (40%) of the invasive devices analyzed. These catheters showed the highest proportion of colonization by multidrug-resistant microorganisms (64; 58%), with a statistically significant association ($p<0.010$). In contrast, indwelling urinary catheters (95; 43%; $p=0.072$) and endotracheal tubes (41%; $p=0.064$) did not show a statistically significant association with these microorganisms. These findings are detailed in Table 3.

Table 3 – Invasive device use and presence of bacterial biofilm (n=200). Maceió, AL, Brazil, 2025

Device	Biofilm with microorganism		p-value*
	MDR/XDR/PDR n (%)	Sensitive n (%)	
Central venous catheter	64 (58.0)	46 (42.0)	<0.010
Indwelling urinary catheter	41 (43.0)	54 (57.0)	0.072
Orotracheal tube	29 (41.0)	41 (59.0)	0.064

*Chi-square test; MDR: Multidrug-resistant; XDR: Extensively drug-resistant; PDR: Pandrug-resistant

Microbiological analysis demonstrated a marked predominance of Gram-negative bacteria (75%) compared to Gram-positive bacteria (25%) ($p<0.001$). The most prevalent pathogens were *Klebsiella pneumoniae* (30%), *Acinetobacter baumannii* (25%), and *Pseudomonas aeruginosa* (20%), all highly virulent opportunistic agents with complex resistance mechanisms. Regarding resistance, we observed high rates of MDR (48%; $p<0.001$), XDR (19%; $p=0.005$), and PDR (3%; $p=0.041$). These findings highlight a scena-

rio of epidemiological concern, as the predominance of multidrug-resistant microorganisms is associated with therapeutic limitations and higher mortality in critically ill patients (Table 4).

Table 4 – Distribution of microorganisms and resistance profiles (n=200). Maceió, AL, Brazil, 2025

Category	n (%)	CI95%	Z-test	p-value*
Gram-negative	150 (75.0)	69.2 – 80.8	Proportion test (vs 50%)	<0.001
Gram-positive	50 (25.0)	19.2 – 30.8	-	-
Multidrug-resistant	96 (48.0)	41.2 – 54.8	Proportion test (vs 30%)	<0.001
Extensively drug-resistant	38 (19.0)	13.5 – 24.5	Proportion test (vs 10%)	0.005
Pandrug-resistant	6 (3.0)	0.6 – 5.4	Proportion test (vs 0%)	0.041
Susceptible/non-MDR	60 (30.0)	23.4 – 36.6	Proportion test (vs 50%)	<0.001

*Chi-square test; MDR: Multidrug-resistant

Discussion

The mean ICU length of stay was 10 days, a duration that increases exposure to risk factors, particularly the prolonged use of invasive devices. We analyzed 275 devices, with central venous catheters being the most frequent, followed by indwelling urinary catheters and endotracheal tubes. These devices are well-known predisposing factors for HAIs, particularly when used for prolonged periods⁽¹⁰⁾.

Bacterial biofilm formation on invasive device surfaces is a primary mechanism of microbial persistence and resistance to antimicrobial treatment. Studies show that biofilms protect microorganisms from antibiotics and the immune system, hindering infection eradication⁽¹²⁾. In our study, the majority of devices showed evidence of biofilm and bacterial growth, corroborating the importance of this factor in HAI etiology.

Isolates were predominantly Gram-negative, notably *Klebsiella pneumoniae*, *Acinetobacter baumannii*, and *Pseudomonas aeruginosa*. These pathogens are frequently implicated in severe hospital infections, particularly in critical care settings. Data from the Hospital Infection Control Information Sys-

tem indicate that *P. aeruginosa* and *K. pneumoniae* are among the most common agents in bloodstream infections and ventilator-associated pneumonias⁽¹³⁾.

Moreover, the high proportion of MDR, XDR, and PDR isolates is particularly concerning. These resistance profiles significantly limit therapeutic options, increase mortality, and burden healthcare systems. A multicenter study in Latin America found that over 40% of *A. baumannii* and *K. pneumoniae* infections were carbapenem-resistant, corroborating our findings⁽¹⁴⁾.

The spread of resistant bacteria is often associated with inappropriate antimicrobial use, lapses in infection control, and prolonged ICU stay⁽¹⁵⁾. Carbapenemase-producing *A. baumannii* and *K. pneumoniae*, although not directly evaluated here, are common in Brazilian hospitals and represent a public health threat⁽¹⁶⁾.

These data reinforce the urgent need to strengthen infection control programs, including the implementation of prevention bundles, early catheter removal and extubation, active microbiological monitoring, and the promotion of rational antimicrobial use. Furthermore, continuing education for healthcare staff and strict adherence to hand hygiene are fundamental to reducing cross-transmission⁽¹⁷⁾.

Our findings confirm that central venous catheters are potential colonization sites for multidrug-resistant microorganisms, especially Gram-negatives like *Klebsiella pneumoniae*, *Acinetobacter baumannii*, and *Pseudomonas aeruginosa*, supporting previous studies linking these catheters to MDR bloodstream infections in ICUs⁽¹⁸⁾.

In Brazilian ICUs, *K. pneumoniae*, *Acinetobacter spp.*, and *P. aeruginosa* are prevalent pathogens in catheter-associated infections, frequently displaying resistance to third- and fourth-generation cephalosporins and carbapenems. Evidence from this study reinforces this issue, demonstrating a severe microbiological profile consistent with the emergence of multidrug-resistant pathogens on invasive devices⁽¹⁹⁾.

Furthermore, the high rate of biofilm formation, characterized by Gram-negative predominance and high percentages of MDR, XDR, and PDR, aligns

with recent findings indicating frequent biofilm formation on CVCs, a key factor in the pathogenesis of hospital-acquired infections⁽²⁰⁾.

This evidence underscores the importance of strict aseptic technique to prevent biofilm formation and colonization by highly resistant microorganisms, particularly given the clinical and epidemiological impact.

Finally, the association between chronic comorbidities (hypertension, diabetes, COPD) and longer length of stay observed in our sample suggests a population highly vulnerable to HAIs⁽²¹⁾. Consistent with our findings, significant associations exist among various risk factors common in intensive care patients, such as advanced age, comorbidities, immunosuppression, and prolonged hospitalization⁽²²⁻²³⁾.

Thus, identifying patient subgroups with chronic comorbidities should guide more intensive prevention strategies, such as sterile insertion protocols, strict maintenance, and careful assessment of the continued need for the device⁽²⁴⁾. Thus, this study highlights a high rate of colonization by resistant pathogens on invasive devices within a vulnerable population exposed to the hospital environment for prolonged periods. These findings should guide institutional policies for epidemiological surveillance and targeted interventions to prevent HAIs, especially in resource-limited regions like Northeastern Brazil.

Study limitations

Study limitations include variations in sample collection, transport, and processing protocols across different periods, as well as intrinsic patient factors, which may influence microbiological results and affect data comparability.

Contributions to practice

These results provide a basis for strengthening strategies to prevent Healthcare-Associated Infections, in addition to promoting patient safety and quality of care for critically ill patients. Furthermore, kno-

wing the microbial resistance profile reinforces the need for rational antimicrobial use in clinical practice, especially for intensive care patients.

Conclusion

The microbiological evaluation of invasive devices showed a predominance of multidrug-resistant microorganisms, particularly *Klebsiella pneumoniae*, and extensive biofilm formation on most devices (central venous catheters, indwelling urinary catheters, and endotracheal tubes). These findings highlight the need to intensify prevention and control strategies for these adverse events, given their potential to cause severe complications in ICU patients.

Authors' contributions

Conception and design or analysis and interpretation of data; Drafting of the manuscript or critical revision for important intellectual content: **Djú TM, Bernardo THL, Costa CRB**. Writing of the manuscript or relevant critical review of the intellectual content: **Santos M, Silva YS, Silva AGS, Iurna TB**. Final approval of the version to be published; Agreement to be responsible for all aspects of the manuscript ensuring that issues relating to the accuracy or completeness of any part of the manuscript are properly investigated and resolved. **Djú TM, Bernardo THL, Costa CRB**.

Data availability

The authors declare that the data are available within the article. However, the corresponding author may be contacted for further information.

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