Original Article

# Antimicrobial Susceptibility Profiles of Microorganisms Isolated from Patients Treated in Intensive Care Units Before and During the COVID-19 Pandemic: A Single-center Retrospective Study

## Türkiye'de COVID-19 Pandemisi Öncesi ve Sırasında Yoğun Bakım Ünitesinde Tedavi Edilen Hastalardan İzole Edilen Mikroorganizmaların Antimikrobiyal Duyarlılık Profilleri: Tek Merkezli Retrospektif Bir Çalışma

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## ABSTRACT

**Objective:** The global coronavirus disease-2019 (COVID-19) pandemic has increased public health challenges, especially in intensive care units (ICU) where COVID-19 patients are at increased risk of infectious complications. This study aimed to identify, compare, and evaluate antimicrobial susceptibility profiles of microbial isolates from ICU patients of a tertiary hospital in Türkiye before and during the COVID-19 pandemic.

**Methods:** In this retrospective analysis, we analyzed data from 1462 patients who were admitted to the ICU of Sivas Cumhuriyet University Faculty of Medicine Application and Research Hospital between January 2018 and December 2022. In this analysis, demographic and clinical variables including age, gender, and antimicrobial susceptibility test results, as well as the annual distribution and antimicrobial resistance profiles of clinical isolates were determined.

**Results:** Among the 1687 sputum, 1396 urine, and 1307 blood cultures analyzed, there was a significant increase in sputum cultures during the pandemic (21.94%; p=0.012). The proportion of gram-negative bacteria was high in all cultures. *Pseudomonas aeruginosa*

## ÖZ

**Amaç:** Küresel koronavirüs hastalığı-2019 (COVID-19) pandemisi, özellikle COVID-19 hastalarının enfeksiyöz komplikasyon riskini artırdığı için yoğun bakım ünitelerindeki halk sağlığı sorunlarını da artırmıştır. Bu çalışmanın amacı, COVID-19 pandemisi öncesinde ve pandemi sırasında Türkiye'deki üçüncü basamak bir hastanenin yoğun bakım ünitesindeki (YBÜ) hastalarından izole edilen mikrobiyal izolatların antimikrobiyal duyarlılık profillerinin değerlendirilmesidir.

**Yöntemler:** Bu retrospektif çalışmada, Ocak 2018 ile Aralık 2022 tarihleri arasında Sivas Cumhuriyet Üniversitesi Tıp Fakültesi Uygulama ve Araştırma Hastanesi YBÜ'ye kabul edilen 1462 hastaya ait veriler analiz edilmiştir. Bu amaçla hastalara ait yaş, cinsiyet ve antimikrobiyal duyarlılık testi sonuçlarını içeren demografik ve klinik değişkenlerin yanı sıra klinik izolatların yıllık dağılımı ve antimikrobiyal direnç profilleri belirlenmiştir.

**Bulgular:** Analiz edilen 1687 balgam, 1396 idrar ve 1307 kan kültürü arasında, pandemi sırasında balgam kültürlerinde önemli bir artış olmuştur (%21,94; p=0,012). Gram-negatif bakterilerin oranı tüm kültürlerde yüksek tespit edilmiştir. Balgam kültürlerinde

(*P. aeruginosa*) was common in sputum cultures, *Escherichia coli (E. coli)* in urine cultures, and coagulase-negative staphylococci (CoNS) in blood cultures. Gram-negative bacteria gradually increased in all cultures from 2018 to 2022. There was a decrease in gram-positive bacteria. In general, antibiotic resistance of *P. aeruginosa* and *E. coli* isolates increased before the pandemic but decreased during the pandemic.

**Conclusion:** Our study shows that infection profiles before and during the pandemic are different from each other. Continuous monitoring of resistance patterns will contribute to developing infection control strategies to prevent the development of antimicrobial resistance.

**Keywords:** Antimicrobial resistance, intensive care unit, COVID-19, antimicrobial susceptibility profile

*Pseudomonas aeruginosa (P. aeruginosa)*, idrar kültürlerinde *Escherichia coli (E. coli)* ve kan kültürlerinde koagülaz-negatif Stafilokoklar yaygın bulunmuştur. Gram-negatif bakteriler 2018'den 2022'ye kadar tüm kültürlerde kademeli olarak artmıştır. Gram-pozitif bakterilerde ise azalma görülmüştür. Genel olarak, *P. aeruginosa* ve *E. coli* izolatlarının antibiyotik direnci pandemi öncesinde artmış ancak pandemi sırasında azalmıştır.

**Sonuç:** Çalışmamız pandemi öncesi ve pandemi sırasındaki enfeksiyon profillerinin birbirinden farklı olduğunu göstermektedir. Direnç paternlerinin sürekli izlenmesi, antimikrobiyal direnç gelişimini önlemek için enfeksiyon kontrol stratejilerinin geliştirilmesine katkıda bulunacaktır.

**Anahtar Kelimeler:** Antimikrobiyal direnç, yoğun bakım ünitesi, COVID-19, antimikrobiyal duyarlılık profili

## Introduction

Antimicrobial resistance is an important public health problem that is increasing daily (1). Uncontrolled use of antimicrobial agents in the treatment of infectious diseases is one of the leading factors causing the development of antimicrobial resistance (2). Nosocomial infections cause serious mortality and morbidity by prolonging the hospital stay of patients (3). Infections in intensive care units (ICUs) account for 25% of nosocomial infections (4). Patients hospitalized in ICUs are usually immunocompromised individuals who have undergone interventional procedures and receive broad-spectrum antibiotic treatment (2). Intensive use of antibiotics in ICUs is one of the leading factors in the emergence and development of antimicrobial resistance. *Acinetobacter baumannii (A. baumannii), Pseudomonas aeruginosa (P. aeruginosa), Klebsiella pneumoniae (K. pneumoniae),* methicillin-resistant *Staphylococcus aureus (S. aureus), Escherichia coli (E. coli),* Vancomycin-Resistant *Enterococcus, Serratia marcescens, Enterobacter cloacae (E. cloacae)* are among the microorganisms causing nosocomial infections (4,5). Antimicrobial resistance profiles of the microorganisms causing these infections vary regionally (4). Identifying these microorganisms and determining their antimicrobial susceptibility profiles are essential for clinicians to control nosocomial infections and determine infectious disease treatment plans (6). With the emergence of the coronavirus disease-2019 (COVID-19) pandemic, researchers have warned of the risks of inappropriate antibiotic use and the risks that may occur, based on experience from past outbreaks (7). Especially the low immunity of patients treated in ICUs and the effect of invasive interventions applied to these patients cause an increased risk of infectious diseases in these patients. This leads to more antimicrobial agents (2). Monitoring and evaluating the use of antimicrobial agents in critical situations such as pandemics, where infectious diseases are widespread throughout society, is essential for the continuation of public health. As a result of research conducted in different countries, it is stated that the use of antimicrobial agents has increased during the COVID-19 pandemic (8). In this study, it was aimed to retrospectively investigate the microorganisms isolated from the samples sent to the microbiology laboratory and antimicrobial susceptibility profiles of patients who were being treated in

the ICUs of Sivas Cumhuriyet University Faculty of Medicine Application and Research Hospital before and during the COVID-19 pandemic.

## Methods

### Study Design and Setting

In this study, we retrospectively analyzed the reports of patients treated in the ICU of Sivas Cumhuriyet University Faculty of Medicine Application and Research Hospital. The study was conducted to determine the resistance profiles of microorganisms isolated from sputum, urine, and blood cultures. Sivas Cumhuriyet University Faculty of Medicine Application and Research Hospital is a tertiary care teaching hospital with 1050 beds. We surveyed samples, demographic characteristics, and results of culture tests of patients treated in the ICU of the hospital between January 2018 and December 2022. In Türkiye, the period before this date was considered pre-pandemic since the pandemic was declared on March 11, 2020.

### Inclusion Criteria

This study included isolates from sputum, urine, and blood culture samples. Two blood cultures with the same results were included in the study. The first samples from recurrent patient samples were accepted into the study, and the others were excluded. In addition, samples considered to be contaminated were excluded.

### Bacterial Identification, and Antimicrobial Susceptibility Tests

Clinical isolates in the samples sent to the microbiology laboratory were identified by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) using Microflex LT MALDI-TOF MS (Bruker Daltonics, Germany) according to the manufacturer's operating procedures. Antimicrobial susceptibility tests of the identified bacterial isolates were analyzed using Phoenix 100® (Becton Dickinson, USA), and yeast isolates were analyzed using Micronaut (Bruker Daltonics, Germany) test kits according to the manufacturer's procedures. Antimicrobial susceptibilities of the isolates were evaluated according to the European Committee on Antimicrobial Susceptibility Testing criteria (9).

## Data Collection

Demographic information such as age, and gender, and clinical information such as antimicrobial susceptibility test results were collected from the electronic medical records of the hospital. This study was conducted in compliance with the Helsinki Declaration and informed consent was obtained from all participants before starting the study.

## Ethical Principles of the Research

Before initiating the study, ethical permission for the research was obtained from the Sivas Cumhuriyet University Ethical Committee of Non-invasive Clinical Research (decision no: 2023-10/03, date: 19.10.2023).

## Statistical Analysis

Statistical analyses were conducted using GraphPad Prism (version 8) software package. Descriptive analyses were presented using frequencies and percentages. Changes in antimicrobial resistance before and during the pandemic were analyzed using $x^2$ or Fisher's exact test. The significance level was accepted at a $p < 0.05$.

# Results

## Demographics of the Study Population

As presented in Table 1, the study population was nearly equally distributed by sex and predominantly elderly. A total of 4405 microbiological culture results were included in the study (38.43% sputum, 32.8% urine, 29.77% blood). The sputum culture rate was higher in the pre-pandemic period (16.49%) than in pandemic period (21.94 %) (p=0.012).

## Clinical Isolates

Gram-negative bacteria were the most frequently isolated microorganisms from sputum, urine, and blood cultures in the ICU. Gram-positive bacteria were more common in blood cultures (46.31%) than in sputum, and urine cultures. During the pandemic period, the proportion of gram-negative bacteria grown in sputum, urine, and blood cultures increased (77.26%, 75%, and 56.52%, respectively) (Table 2, Figure 1).

As shown in Figure 1 and Table 2, the dominant pathogens varied by specimen: *P. aeruginosa, S. aureus* and *K. pneumoniae* were common in sputum; *E. coli, K. pneumoniae* and *Enterococcus faecium (E. faecium)* in urine; and CoNS, *K. pneumoniae* and *S. aureus* in blood cultures.

## Antimicrobial Resistance in Bacteria

### Antimicrobial Resistance Profile of Gram-positive Bacteria

Antimicrobial resistance status of gram-positive bacteria commonly grown in sputum, urine, and blood cultures were analyzed. Overall, *S. aureus* isolates grown in sputum, urine and blood cultures were susceptible to vancomycin, teicoplanin, tigecycline and trimethoprim-sulfamethoxazole, but showed high resistance to penicillin (99.2% sputum; 100% urine; 99.0% blood cultures) and ampicillin (99.2% sputum; 100% urine; 99.1% blood cultures) (Figure 2).

*E. faecium* grown in urine culture showed increased resistance to teicoplanin (0.0% pre-pandemic; 14.3% pandemic; p=0.007) and decreased resistance to amoxicillin-clavulanate (97.9% pre-pandemic; 84.9% pandemic; p=0.032) during the pandemic (Figure 2).

Resistance to trimethoprim-sulfamethoxazole (9.8% pre-pandemic; 32.3% pandemic; p<0.001) and teicoplanin (3.9% pre-pandemic; 11.4% pandemic; p=0.031) increased, while resistance to clindamycin (69.6% pre-pandemic; 54.4% pandemic; p=0.008) decreased in CoNS grown in blood cultures. A decrease in amoxicillin-clavulanate (12.9% pre-pandemic; 0.0% pandemic; p=0.032) resistance was also observed in *Enterococcus faecalis (E. faecalis)* grown in blood cultures (Figure 2).

### Antimicrobial Resistance of Gram-negative Bacteria

Antimicrobial resistance profiles of the most frequently grown six gram-negative bacteria in sputum, urine, and blood cultures are shown in Figure 2. During the pandemic, *P. aeruginosa* in sputum cultures showed a decrease in cephalosporin resistance (42.3% pre-pandemic; 27.6% pandemic; p<0.001) and an increase in gentamicin resistance (16.7% pre-pandemic; 47.6% pandemic; p<0.001). *K. pneumoniae* grown in sputum cultures showed a decrease in levofloxacin resistance (83.3% pre-pandemic; 60.6% pandemic; p<0.001). *E. coli* grown in sputum cultures showed decreasing resistance to β-lactam/adjuvant antibiotics (69.4% pre-pandemic; 44.1% pandemic; p<0.001) and ciprofloxacin (70.8% pre-pandemic; 50.5% pandemic; p=0.013). During the pandemic period, *E. cloacae* isolates were resistant to carbapenems (2.8% pre-pandemic; 39.4% pandemic; p<0.001) and ceftazidime (16.7% pre-pandemic; 61.9% pandemic; p=0.027), while *Proteus mirabilis* (*P. mirabilis)* isolates had increased resistance to cefuroxime (0.0% pre-pandemic; 19.05% pandemic; p=0.018) and gentamicin (25.9% pre-pandemic; 54.8% pandemic; p=0.034).

*E. coli* grown in urine cultures were resistant to trimethoprim-sulfamethoxazole (56.9% pre-pandemic; 43.6% pandemic; p=0.008), β-lactam/adjuvant (43.4% pre-pandemic; 28.7% pandemic; p<0.001), ampicillin (83.9% pre-pandemic; 71.1% pandemic; p=0.002) and ciprofloxacin (55.4% pre-pandemic; 44.1% pandemic; p=0.025) significantly decreased during the pandemic period. However, resistance to ceftazidime (44.3% pre-pandemic; 88.9% pandemic; p<0.001) increased. Ceftazidime (75.2% pre-pandemic; 98.5% pandemic; p<0.001) resistance of *K. pneumoniae* grown in urine cultures increased, while levofloxacin (90.9% pre-pandemic; 61.5% pandemic; p<0.001) resistance decreased. Piperacillin/tazobactam (62.7% pre-pandemic; 3.4% pandemic; p<0.001) and ceftazidime (47.1% pre-pandemic; 12.7% pandemic; p<0.001) resistance of *P. aeruginosa* isolates grown in urine cultures decreased during the pandemic period. Trimethoprim-sulfamethoxazole (57.8% pre-pandemic; 81.8% pandemic; p=0.025) resistance of *P. mirabilis* isolates increased.

In blood cultures, piperacillin/tazobactam (47.1% pre-pandemic; 37.7% pandemic; p=0.001) and ceftazidime (37.7% pre-pandemic; 19.0% pandemic; p=0.002) resistance of *P. aeruginosa* decreased, while tigecycline (8.3% pre-pandemic; 33.3% pandemic; p<0.001) resistance of *E. coli* increased.

## Table 1. Characteristics of patients hospitalized in ICU

| Characteristics | | Pre-pandemic (%) | During-pandemic (%) | Total (%) | p-value |
|---|---|---|---|---|---|
| Total n (%) | | 566 (38.71) | 896 (61.29) | 1462 | |
| Gender n (%) | Female | 287 (19.63) | 415 (28.39) | 702 (48.02) | 0.107 |
| | Male | 279 (19.08) | 481 (32.90) | 760 (51.98) | |
| Age group n (%) | ≤44 | 37 (2.53) | 94 (6.43) | 131 (8.96) | 0.089 |
| | 45-64 | 120 (8.21) | 197 (13.47) | 316 (21.68) | |
| | 65-74 | 139 (9.51) | 220 (15.05) | 362 (24.56) | |
| | 75-84 | 168 (11.49) | 246 (16.83) | 414 (28.32) | |
| | ≥85 | 102 (6.98) | 139 (9.51) | 242 (16.48) | |
| | Median (IQR) | 74 (20) | 72 (21) | 72 (20) | |
| Culture type n (%) | Sputum culture | 724 (16.49) | 963 (21.94) | 1687 (38.43) | 0.012* |
| | Urine culture | 660 (15.03) | 736 (16.77) | 1396 (32.80) | |
| | Blood culture | 624 (14.21) | 683 (15.56) | 1307 (29.77) | |
| | Total cultures | 2008 (45.74) | 2382 (54.26) | 4390 | |

The $x^2$ test was used to analyze the data. *: $p < 0.05$
ICU: Intensive care units, IQR: Interquartile range

## Table 2. Distribution of clinical isolates according to samples

| Pathogens | Sputum (%) | | Urine (%) | | Blood (%) | |
|---|---|---|---|---|---|---|
| | Pre-pandemic (n=724) | During-pandemic (n=963) | Pre-pandemic (n=663) | During-pandemic (n=748) | Pre-pandemic (n=624) | During-pandemic (n=683) |
| **Gram-positive bacteria** | **26.93** | **22.74** | **17.27** | **19.57** | **46.31** | **40.56** |
| *Staphylococcus aureus* | 13.95 | 17.76 | 0.76 | 1.90 | 8.01 | 9.52 |
| CoNS | 0.83 | 0.10 | 1.82 | 2.45 | 25.80 | 18.30 |
| *Enterococcus faecalis* | 0.41 | 0.10 | 6.21 | 5.84 | 4.97 | 6.00 |
| *Enterococcus faecium* | 0.41 | 0.10 | 7.58 | 7.74 | 1.12 | 3.22 |
| *Streptococcus pneumoniae* | 0.97 | 1.97 | 0.00 | 0.00 | 0.80 | 0.44 |
| *Corynebacterium striatum* | 8.98 | 1.87 | 0.61 | 0.27 | 3.53 | 1.76 |
| Others | 1.38 | 0.83 | 0.30 | 1.36 | 2.08 | 1.32 |
| **Gram-negative bacteria** | **72.93** | **77.26** | **69.39** | **75.00** | **50.96** | **56.52** |
| *Pseudomonas aeruginosa* | 19.06 | 16.10 | 7.42 | 9.51 | 8.49 | 8.78 |
| *Acinetobacter baumannii* | 11.19 | 14.75 | 4.39 | 4.21 | 6.09 | 7.61 |
| *Klebsiella pneumoniae* | 12.15 | 14.33 | 13.94 | 18.34 | 10.90 | 12.30 |
| *Escherichia coli* | 9.94 | 9.66 | 26.52 | 30.03 | 8.49 | 7.03 |
| *Stenotrophomonas maltophilia* | 3.73 | 4.15 | 0.15 | 0.14 | 3.53 | 5.86 |
| *Enterobacter cloacae* | 1.66 | 2.28 | 1.06 | 1.36 | 1.76 | 2.49 |
| *Proteus mirabilis* | 3.73 | 4.36 | 6.97 | 5.98 | 1.28 | 1.90 |
| Others | 11.46 | 11.63 | 8.94 | 5.43 | 10.42 | 10.54 |
| **Fungi** | **0.14** | **0.00** | **13.33** | **5.43** | **2.72** | **2.93** |
| *Candida albicans* | 0.14 | 0.00 | 7.42 | 2.72 | 1.12 | 0.88 |
| Others | 0.00 | 0.00 | 5.91 | 2.72 | 1.60 | 2.05 |

Bold values mean total percentages of gram-positive, gram-negative, and fungi
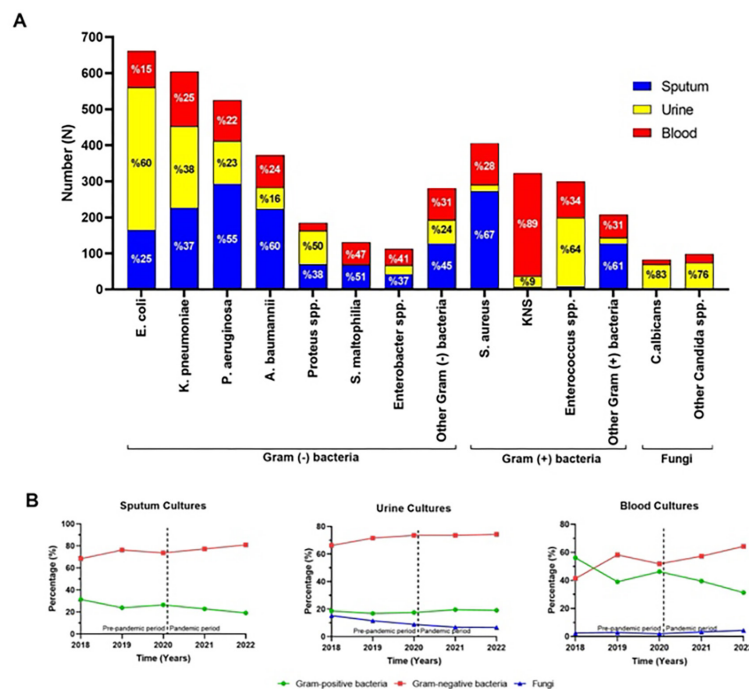CoNS: Coagulase-negative staphylococci

**Figure 1.** Clinical isolate types in ICU

(A) The composition of all pathogens. (B) Distribution of pathogens before and during the pandemic (2018-2022).

*ICU: Intensive care unit*

**Annually Antimicrobial Resistance Distribution of Clinical Isolates**

As shown in Figure 3, the annual variation in antibiotic resistance rates of *P. aeruginosa*, *E. coli*, and *K. pneumoniae* isolates frequently grown in sputum, urine, and blood cultures were analyzed.

In all types of cultures,−β-lactam/adjuvant antibiotic resistance in *P. aeruginosa* isolates across all cultures decreased after 2019. Resistance to fluoroquinolones steadily increased before the pandemic but showed a continuous decrease during the pandemic period. Nevertheless, fluoroquinolones resistance rates in sputum and urine cultures in 2022 were higher than those in 2018. Carbapenem resistance in isolates from sputum cultures increased before the pandemic but remained stable during the pandemic period. The highest carbapenem resistance in isolates from blood cultures was observed in 2021. Cephalosporin resistance in isolates from sputum cultures decreased from 48.42% in 2019 to 24.78% in 2022, while the resistance rate in urine cultures decreased to 8% in 2022. Cephalosporin resistance rate in blood cultures decreased from 51.72% in 2019 to 17.5% in 2022. The resistance of *P. aeruginosa* isolates to aminoglycosides increased from 2018 to 2020, but after 2020; it decreased again to the resistance rates observed in 2018 (Figure 3).

While the antifolate, cephalosporins, and aminoglycosides resistance of *E. coli* isolates grown in urine and blood cultures increased before the pandemic, it decreased to its lowest rate in 2021 and then rose again in 2022. Specifically, antifolate resistance decreased from 60% to 36% in urine isolates and

from 66.6% to 29.4% in blood isolates. Similarly, cephalosporin resistance dropped from 56.8% to 48.6% in urine and from 57.1% to 23.6% in blood samples. For aminoglycosides, resistance fell from 12% to 6.4% in urine cultures and from 25% to 5.5% in blood cultures. Antifolate resistance rate of *E. coli* isolates in sputum cultures was the highest (67%) in 2019 and decreased to the lowest level (35%) in 2022. Additionally, the rate of cephalosporin resistance in sputum cultures decreased from 57.7% in 2020 to 23.5% in 2022. There was no significant change in aminoglycoside resistance. While the β-lactam/adjuvant, and fluoroquinolone resistance rates of *E. coli* isolates growing in all cultures were highest in the pre-pandemic period, they decreased during the pandemic. Resistance rates of *E. coli* isolates to carbapenems were the lowest compared to other antimicrobials. In 2019, carbapenem resistance (22.6%) increased in sputum isolates (Figure 3).

There was no significant change in the antifolate resistance of *K. pneumoniae* isolates grown in sputum cultures. In urine and blood cultures, antifolate resistance increased in a fluctuating manner. The distribution of β-lactam/adjuvant antibiotic resistance in *K. pneumoniae* isolates grown in all culture types showed a similar pattern. Fluoroquinolone resistance distribution was similar across all culture types. The highest fluoroquinolone resistance was observed in isolates obtained from urine cultures, with a rate of 91.1% in 2019. Carbapenem resistance of isolates grown in sputum, urine, and blood cultures increased gradually over the years. In 2022, the resistance rates were 62.8%, 50%, and 56.4%, respectively. Cephalosporin resistance rates decreased during the pandemic period; however, an increase started to
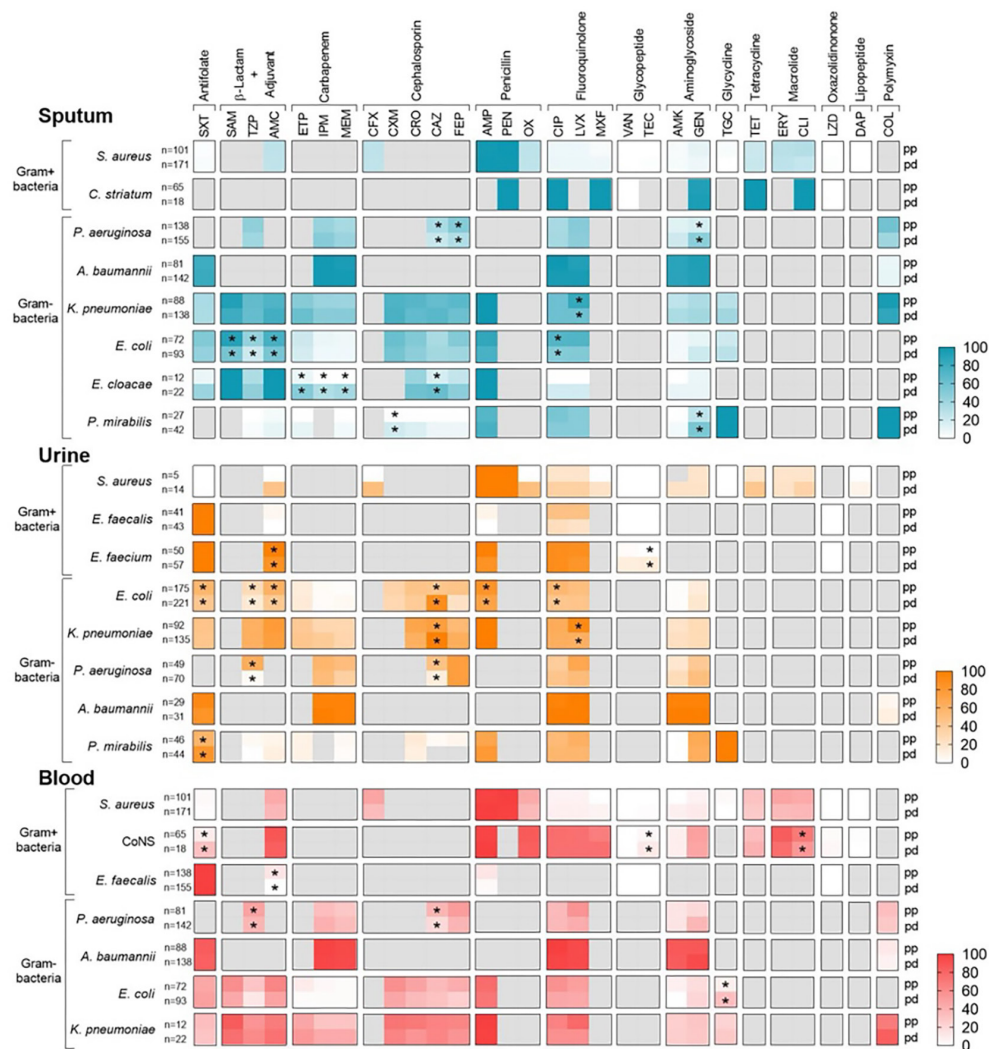
**Figure 2.** Antimicrobial resistance profile of gram-positive, and gram-negative bacteria grown in sputum, urine, and blood cultures

In the heatmap, the color of each cell shows the percentages of resistant strains grouped according to the antimicrobial classification indicated in the columns. Gray cells indicate that the bacteria were not tested with the antimicrobial listed in the column. Pre-pandemic period (pp) and pandemic period (pd) were compared in each bacterial group. Asterisked cells indicate isolates with a significantly different resistance profile to the indicated antimicrobial during the pandemic compared to the pre-pandemic period (analyzed by chi-square and Fisher's exact test)

occur again in 2022. The aminoglycoside resistance rates of *K. pneumoniae* isolates grown in sputum, urine, and blood cultures were 9.3%, 13.9%, and 7.7%, respectively, in 2018. However, in 2022, the resistance rates had increased to 48.9%, 36.1%, and 42.3%, respectively (Figure 3).

**Antimicrobial Resistance in Fungi**

Antimicrobial resistance of *Candida albicans (C. albicans)* and other Candida isolated from urine and blood cultures were analyzed. *C. tropicalis*, *C. glabrata*, *C. parapsilosis*, and *C. krusei* were included among another Candida. Although the resistance rates of *C. albicans* isolates grown in urine cultures to azoles (43.8% pre-pandemic; 37.5% pandemic; p=0.341), echinocandins (17.7% pre-pandemic; 10.0% pandemic; p=0.432), and amphotericin B (2.0% pre-pandemic; 0.0%

pandemic; p>0.999) decreased during the pandemic period, there was no significant change (Figure 4). On the other hand, the resistance rates of *C. albicans* isolates grown in blood culture to azoles (32.14% pre-pandemic; 70.8% pandemic; p=0.012) and echinocandins (7.7% pre-pandemic; 25.0% pandemic; p=0.321) increased during the pandemic period. All blood culture isolates were susceptible to amphotericin B (Figure 4).

## Discussion

Antibiotic resistance poses a significant global public health threat and leads to increased mortality, hospitalizations, and prolonged hospital stays due to infections caused by resistant bacteria (1). The emergence of the COVID-19 pandemic has put great pressure on healthcare systems and led to a significant increase in empirical antibiotic treatments, especially among
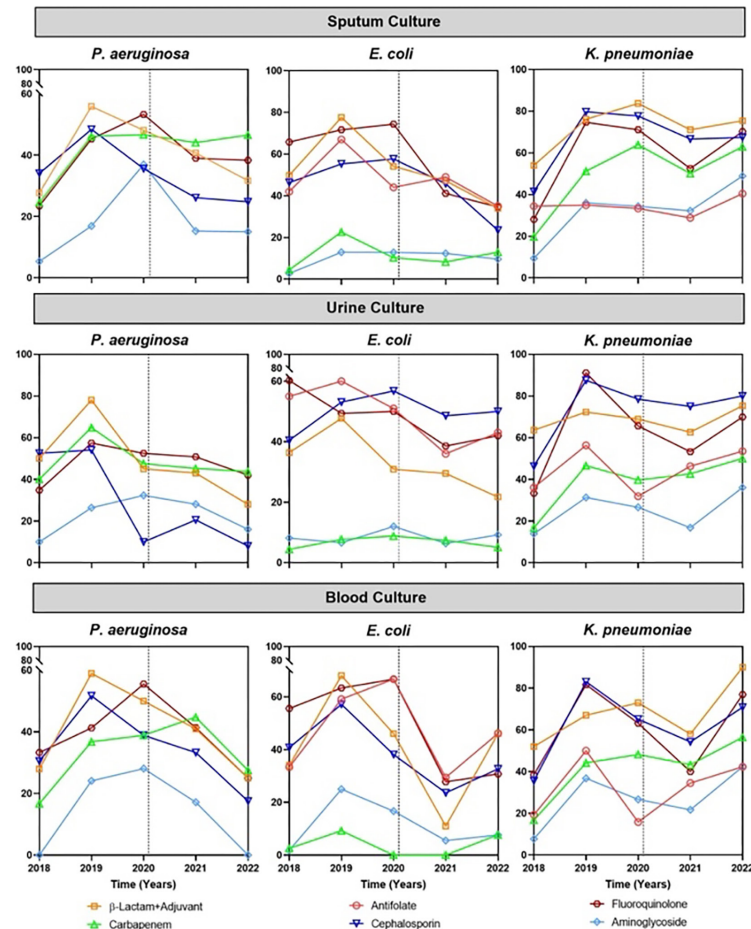
**Figure 3.** Annual change in resistance to antimicrobial classes of some of the most common isolates grown in sputum, urine, and blood cultures

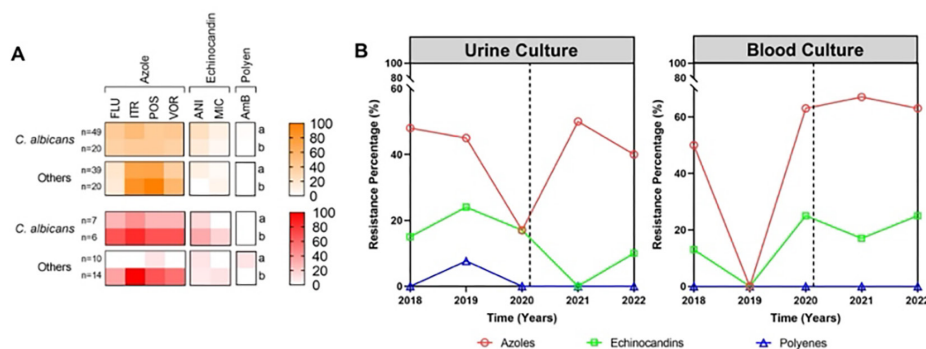*The dashed line indicates the beginning of the pandemic period*



**Figure 4.** (A) Antimicrobial resistance profile of fungi grown in urine and blood cultures. (B) Annual change in antimicrobial resistance of C. albicans grown in urine and blood cultures

Antimicrobial resistance profile of fungi grown in urine and blood cultures, and annual change in antimicrobial resistance of *C. albicans*. In the heatmap, the color of each cell shows the percentages of resistant strains grouped according to the antimicrobial classification indicated in the columns. Orange color represents urine cultures, and red color represents blood cultures. Pre-pandemic period (pp) and pandemic period (pd) were compared in each bacterial group. Time-antimicrobial resistance graphs show the annual change in antimicrobial resistance of *C. albicans* grown in urine and blood cultures. The dashed line indicates the beginning of the pandemic period

intensive-care patients (10). Despite this increase in antibiotic use, our study did not observe an overall significant increase in resistance development during the pandemic, consistent with findings in Taiwan however, a notable exception was noted in *K. pneumoniae* showing an increase in resistance during this period, which is consistent with findings from a separate study (10,11).

Our study revealed a significant increase in the number of sputum cultures during the pandemic, possibly attributable to the increased frequency of respiratory sampling required by the pandemic. Gram-negative bacteria were isolated more frequently than gram-positive bacteria in all cultures, which is consistent with previous studies conducted in ICU patients (12,13). Consistent with previous studies, *P. aeruginosa* was found to be the most frequently isolated bacterium from sputum samples in our study (14,15). Urinary tract infections constitute an important part of nosocomial infections in ICU settings (16). In our study, *E. coli* and *K. pneumoniae* were the most frequently isolated bacteria in urine cultures to previous findings (17,18). Bloodstream infections constitute an important burden on human health and the most frequently isolated organism in our study was CoNS, followed by *S. aureus* and *P. aeruginosa*; this is consistent with similar studies showing that gram-positive bacteria are predominant in blood cultures (19,20).

Regarding antimicrobial resistance patterns, our findings revealed that *S. aureus* isolates were susceptible to trimethoprim-sulfamethoxazole, vancomycin, teicoplanin, and tigecycline and showed marked resistance to ampicillin which was also shown in previous studies (12). In contrast, we observed a significant increase in trimethoprim-sulfamethoxazole and teicoplanin resistance and a significant decrease in clindamycin resistance rate among CoNS isolated from blood cultures. Teicoplanin resistance also increased in a post-COVID-19 study, consistent with our findings (11).

Similar to the findings of a study conducted in Romania, a decrease in penicillin resistance among *Enterococcus species* was observed in our study. Furthermore, consistent with the study conducted in Colombia, an increase in vancomycin and teicoplanin resistance was recorded among *E. faecium* isolates (11,21).

The resistance rates of *A. baumannii* isolates to antifolates, carbapenems, fluoroquinolones, and aminoglycosides were found to be quite high in other studies (22). Specifically, no significant change in antimicrobial resistance was observed among *A. baumannii* isolates in our study.

Regarding *P. aeruginosa* isolates, our study revealed a significant decrease in piperacillin-tazobactam and ceftazidime resistance in isolates obtained from blood and urine cultures. However, in sputum culture isolates, resistance to ceftazidime and cefepime decreased significantly, while gentamicin resistance showed a significant increase. These findings are consistent with another study covering the years 2016-2020, which observed a decrease in resistance rates in many antibiotic groups, except amikacin, in *P. aeruginosa* strains during the pandemic period (23).

In terms of *E. coli* isolates our study showed a significant decrease in resistance to antifolate, β-lactam/adjuvant antibiotics,

ciprofloxacin, and ampicillin during the pandemic period. However, resistance to ceftazidime increased. Similarly, as reported in other studies on *K. pneumoniae* isolates; a high level of ampicillin resistance was found in our study (17,18). In particular, *K. pneumoniae* exhibited a high resistance rate against many commonly used antimicrobials. While ceftazidime resistance increased in our study, levofloxacin resistance decreased significantly. Unlike our study, there is another study showing an increase in levofloxacin resistance (11).

Regarding fungal infections, *C. albicans* was the most frequently isolated yeast fungus in our study, which is consistent with previous studies showing its prevalence, especially in ICU patients (24). *C. albicans* isolates obtained from urine cultures generally showed decreased antimicrobial resistance during the pandemic, while those obtained from blood cultures showed increased resistance.

Despite the valuable data provided by our study, some limitations should be noted, including small sample sizes for some isolates and limitations in generalizing the findings due to the single-center nature of the study. Nevertheless, our study highlights the importance of comprehensive research on antimicrobial resistance patterns both before and during pandemics and provides valuable information to address this global health challenge.

## Conclusion

While most of the microorganisms analyzed (*S. aureus*, *C. striatum*, *A. baumannii*, *E. faecalis*) showed no significant change in resistance patterns before and during the pandemic, *P. aeruginosa* and *E. coli* isolates showed a downward trend, while *K. pneumoniae* showed an increase in resistance after 2022. This is thought to be due to the irrational use of antibiotics. In addition to the development of strategies for the control of infectious diseases, it is necessary to raise awareness among healthcare professionals and the public on the rational use of antibiotics. Furthermore, further research is needed to elucidate the development of antimicrobial resistance.

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