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Original Article

Antibiotic Resistance Trends in ESKAPE Pathogens Isolated at a Health Practice and Research Hospital: A Five-Year Retrospective Study

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Abstract

Introduction: Antimicrobial resistance remains a global threat with increasing morbidity and mortality rates. The aim of this study was to identify the antimicrobial resistance trends among ESKAPE pathogens (*Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa,* and *Enterobacter spp.*) isolated from clinical samples at a Health Practice and Research Hospital over five years.

Methodology: Microbiological diagnosis utilized classical culture methods and automated systems. Antimicrobial susceptibility analysis was conducted using BD Phoenix, adhering to European Committee on Antimicrobial Susceptibility Testing (EUCAST) standards.

Results: A total of 15,272 clinical strains of ESKAPE pathogens were identified in this study. The most frequently isolated pathogens among ESKAPE were *K. pneumoniae* (3.938, 27.79%), *Acinetobacter baumannii* (3,013, 19.73%) and *Enterococcus faecium* (2,966, 19.24%). Bacterial strains were isolated predominantly from urine (3,263, 21.37%), followed by blood cultures (3,099, 20.29%). ESKAPE pathogens were most commonly found in internal intensive care units (4,758, 31.16%), followed by surgical intensive care units (4,000, 26.19%). Reduced resistance rates were observed for most antibiotics against *Enterococcus faecium* and *Staphylococcus aureus.* The vancomycin resistance rate for *Enterococcus faecium* was 18.48%, and the methicillin resistance rate for *Staphylococcus aureus* was 44.87%. A concerning trend of increasing antimicrobial resistance was noted in *Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa,* and *Enterobacter spp.*

Conclusions: The alarming rise in antimicrobial resistance among *Pseudomonas aeruginosa, Acinetobacter baumannii*, and *Klebsiella pneumoniae* is a significant concern. The high rates of antimicrobial resistance observed in ESKAPE pathogens underscore the urgent need for improvement in antimicrobial stewardship and infection prevention and control programs.

Key words: Antibiotic resistance; ESKAPE; hospital.

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Introduction

Antimicrobial resistance (AMR) is estimated to contribute to approximately 700,000 deaths worldwide annually. Failure to address AMR is projected to lead to staggering costs of up to 100 trillion USD and 10 million deaths each year by 2050. The most significant impact is anticipated in Asia and Africa, with an estimated 4.7 and 4.2 million deaths, respectively [1].

Quick and accurate diagnostic techniques for identifying AMR genes and bacterial infections in clinical settings are lacking, leading to unnecessary use of broad-spectrum antibiotics [2]. The abuse and overuse of antimicrobial agents are major contributors to the development of AMR [3]. Among multi-antibiotic resistant bacteria, "ESKAPE pathogens" have a profound impact on healthcare-associated infections [4]. The term ESKAPE, coined by Rice in 2008, reflects the ability of these bacteria to "escape" the effects of different antibiotics [5].

The acronym ESKAPE encompasses *Enterococcus faecium (E. faecium), Staphylococcus aureus (S. aureus), Klebsiella pneumoniae (K. pneumoniae), Acinetobacter baumannii (A. baumannii), Pseudomonas aeruginosa (P. aeruginosa), and*

Enterobacter spp., all of which show increasing multidrug resistance and high virulence. These pathogens are major contributors to nosocomial infections due to their elevated antimicrobial resistance levels [6]. ESKAPE bacteria are not only significant for causing nosocomial infections but also serve as models for understanding pathogenesis, transmission, and resistance [5]. In February 2017, the World Health Organization (WHO) identified ESKAPE pathogens as having "priority status" on the list of pathogens urgently requiring the development of new antimicrobials, guiding research and development related to new antibiotics [7].

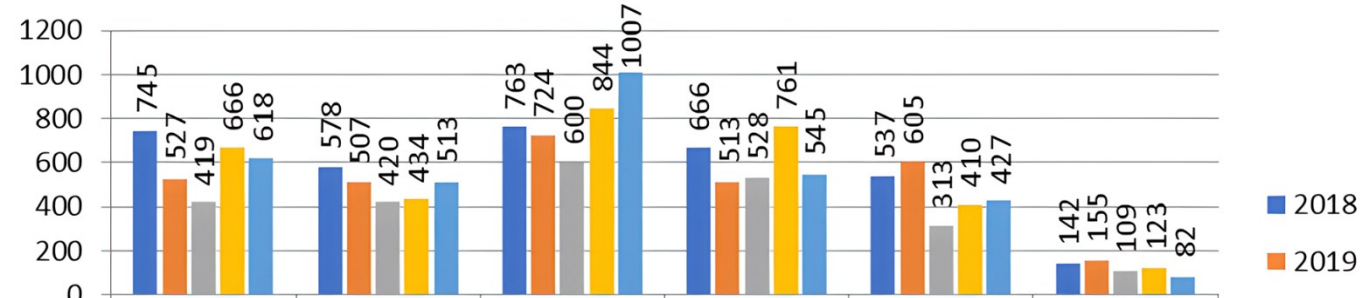
Achieving effective empiric therapy necessitates site-specific surveillance studies and the use of antibiograms as a control measure to diminish the incidence of infections caused by ESKAPE pathogens [8]. The objective of this study was to examine ESKAPE pathogens and the prevailing antibiotic resistance patterns isolated from Kahramanmaraş Sütçü Imam University Health Practice and Research hospital during the period January 2018 to December 2022.

Methodology

The study included 15,272 ESKAPE strains isolated from diverse samples of hospitalized, emergency, and outpatient clinic patients between January 2018 and December 2022. These strains were obtained from the Medical Microbiology Laboratory of Kahramanmaraş Sütçfigureü Imam University Health Practice and Research Hospital, in Turkey. On arrival at the laboratory, the samples were cultured on 5% sheep blood and eosin methylene blue (EMB) agar media, followed by incubation at 37 °C for 24-48 hours. Gram-positive bacteria were identified using colony morphology, Gram staining, catalase and coagulase tests, and the Phoenix TM 100 automated identification system (BD Phoenix System, Beckton Dickinson, USA). Gram-negative bacteria were identified through conventional methods, including carbohydrate and citrate utilization, urease production, oxidase test, and the Phoenix TM 100 automated identification system. Antibiotic susceptibilities of all identified strains were determined with the Phoenix TM 100 automated identification system, following EUCAST (European Committee on Antimicrobial Susceptibility Testing) limit values.

Strains that recurred in the study data were excluded. In instances of the same isolate regrowing from the same patient, only the first isolate was included in the study, in accordance with the cumulative antibiogram data rules. First isolate refers to the first microbial isolate of a particular species obtained from clinical samples of a patient during the analyzed periods (separately for the years 2018, 2019, 2020, 2021 and 2022). Electronic data collection was performed from the records of the laboratory information system corresponding to clinically relevant samples positive for ESKAPE pathogens.

Figure 1. Trends in the number of ESKAPE pathogens isolated from clinical samples from 2018 to 2022.



	E. faecium	S. aureus	K. pneumoniae	A. baumannii	P. aeruginosa	Enterobacter spp.
2018	745	578	763	666	537	142
2019	527	507	724	513	605	155
2020	419	420	600	528	313	109
2021	666	434	844	761	410	123
2022	618	513	1007	545	427	82

Statistical analysis

Data analysis was conducted using IBM SPSS version 22 (IBM SPSS for Windows version 22, IBM Corporation, Armonk, New York, United States) and R 3.3.2 software. Data parameters were expressed as percentages (%) and counts (n). The Chi-Square test and Fisher's Exact test were employed to examine the differences between qualitative variables and frequency distributions among groups. Statistical significance was defined as $p < 0.05$.

Ethical approval

Approval for this study was granted by the Ethics Committee of Kahramanmaraş Sütçü Imam University Faculty of Medicine (Session no: 2023/17, Decision no: 02).

Results

Throughout the study period, 61,406 individual bacterial strains were recovered from clinical samples, with 15,272 (24.87%) identified as ESKAPE pathogens. Of these, 7,922 (51.87%) were isolated from male patients, and 7,350 (48.13%) from female patients.

Among the 15,272 ESKAPE clinical strains, *K. pneumoniae* was the most frequently isolated pathogen (3,938, 27.79%), followed by *A. baumannii* (3,013, 19.73%), *E. faecium* (2,966, 19.24%), *S. aureus* (2,452, 16.05%), *P. aeruginosa* (2,292, 15.00%), and *Enterobacter spp.* (611, 4.00%). In comparison to 2018, there was an increasing trend in the isolation rate of only *K. pneumoniae* in 2022, while a decreasing trend was observed in the isolation rate of the other four pathogens. The trends in the number of ESKAPE pathogens isolated from clinical samples from 2018 to 2022 are illustrated in Figure 1.

ESKAPE pathogens were most frequently isolated from urine cultures (3,263, 21.37%), followed by blood (3,099, 20.29%), wound (2,614, 17.12%), and tracheal aspirate cultures (2,458, 16.09%). The distribution of sample types from which ESKAPE pathogens were isolated is shown in Figure 2.

Figure 2. Types of clinical samples from which ESKAPE pathogens were isolated.

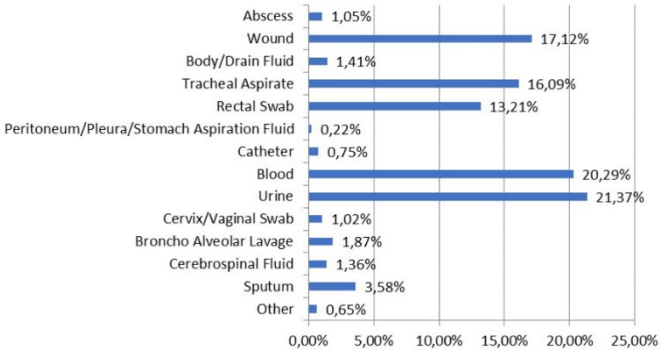
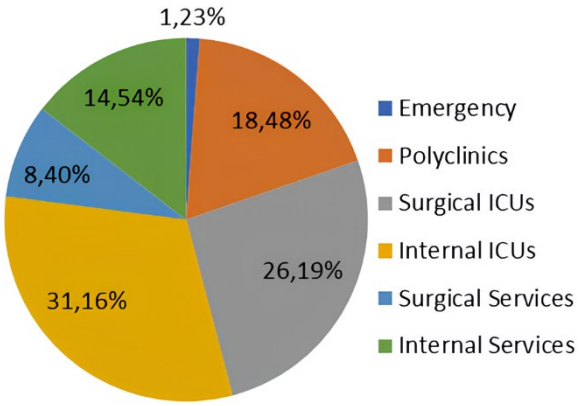


Figure 3. Distribution of ESKAPE pathogens according to clinics.



ESKAPE pathogens were most frequently isolated from internal ICUs (4,758, 31.16%), followed by surgical ICUs (4,000, 26.19%), polyclinics (2,823, 18.48%), and internal wards (2,221, 14.54%). ESKAPE pathogens were also identified in the emergency department (at least 1.23%) and surgical wards (8.40%). The ICU ESKAPE isolation rate was found to be statistically significant ($p < 0.001$). The distribution of ESKAPE pathogens across clinical specialties is presented in Figure 3.

E. faecium displayed high resistance to ampicillin (93.77%) but was highly sensitive to linezolid (97.48%). The susceptibility rates were lower for gentamicin high level (24.15%) and streptomycin high

Table 1. Distribution of antibiotic resistance rates of *Enterococcus faecium* according to years.

ANTIBIOTICS	2018	2019	2020	2021	2022	TOTAL	*p/x²
	n/R (%)	n/R (%)	n/R (%)	n/R (%)	n/R (%)	R (%)	
Ampicillin	740/691 (93.38)	524/500 (95.42)	405/384 (94.81)	658/614 (93.31)	594/546 (91.92)	2735 (93.77)	0.307/1,041
Gentamicin High Level (synergy)	738/535 (72.49)	527/382 (72.49)	408/327 (80.15)	663/539 (81.30)	618/450 (72.82)	2233 (75.85)	0.894/0,017
Linezolid	745/25 (3.36)	531/18 (3.39)	410/14 (3.41)	665/14 (2.11)	618/2 (0.32)	73 (2.52)	< 0.001*/15,993
Streptomycin High Level (synergy)	740/429 (57.97)	527/331 (62.81)	408/315 (77.21)	663/463 (69.83)	618/389 (62.94)	1927 (66.15)	0.062/3,475
Teicoplanin	743/489 (65.81)	523/275 (52.58)	410/255 (62.20)	666/475 (71.32)	616/374 (60.71)	1868 (62.52)	0.052/3,779
Vancomycin	745/133 (17.85)	527/77 (14.61)	410/109 (26.59)	666/137 (20.57)	618/90 (14.56)	546 (18.84)	0.102/2,670

*Comparison of antibiotic resistance between 2018-2022. Chi-Square test; Fisher exact test; a: 0.05; *distributional difference is statistically significant; R: Resistant.

level (33.85%). In comparison to 2018, by 2022, ampicillin resistance decreased by 1.46%, linezolid resistance decreased by 3.04% ($p < 0.001$), teicoplanin resistance decreased by 5.1%, and vancomycin resistance decreased by 3.29%, while streptomycin high-level resistance increased by 4.97%. The gentamicin high-level antibiotic resistance rate remained relatively stable (Table 1).

In this study, penicillin (98.01%) showed the highest resistance among antibiotics in *S. aureus*, while vancomycin resistance was not detected. Very low antibiotic resistance was observed in daptomycin (0.59%), linezolid (0.56%), and teicoplanin (1.68%). In addition, low rates of antibiotic resistance were found in amikacin, levofloxacin, ciprofloxacin, and trimethoprim/sulfamethoxazole. Oxacillin resistance was noted at 44.87%. In comparison to 2018, by 2022, there were decreased resistance rates in *S. aureus* for all antibiotics except penicillin and trimethoprim/sulfamethoxazole. The increase in resistance in penicillin (from 97.07% to 99.59%, $p < 0.002$) and trimethoprim/sulfamethoxazole (from 0.19% to 3.54%, $p < 0.001$) was statistically significant. Conversely, the decrease in resistance to clindamycin, erythromycin, fusidic acid, gentamicin, teicoplanin, and tetracycline was statistically significant ($p < 0.005$) (Table 2).

Between 2018 and 2022, cefuroxime (82.83%), ceftazidime (76.75%), and cefepime (75.88%) exhibited the highest resistance rates in *K. pneumoniae*, while imipenem (24.41%) was the most sensitive antibiotic. Increasing antimicrobial resistance trends were observed in *K. pneumoniae* against all tested antibiotics. Over the years, resistance rates in all antibiotics displayed a significant upward trend. Specifically, within carbapenems, ertapenem resistance increased from 36.97% in 2018 to 58.81% in 2022, imipenem resistance rose from 12.63% to 37.90% ($p < 0.001$), and meropenem resistance increased from 20.77% to 43.07% ($p < 0.001$). Resistance rates of cephalosporins cefepime, ceftazidime, cefuroxime, and ceftriaxone were 67.50%, 64.48%, 79.48%, and 70.08% in 2018, increasing to 84.36%, 85.82%, 88.40%, and 72.99% in 2022. The increase in resistance

Table 2. Distribution of antibiotic resistance rates of *Staphylococcus aureus* by years.

ANTIBIOTICS	2018	2019	2020	2021	2022	TOTAL	*p/x^2
	n/R (%)	n/R (%)	n/R (%)	n/R (%)	n/R (%)	R (%)	
Amikacin	-	415/10 (2.41)	420/10 (2.38)	433/19 (4.39)	512/9 (1.76)	48 (2.74)	-
Clindamycin	561/171 (30.48)	503/153 (30.42)	420/150 (35.71)	434/88 (20.28)	512/10 (21.48)	672 (27.67)	< 0.001*/11,209
Daptomycin	554/3 (0.54)	503/4 (0.80)	420/3 (0.71)	433/3 (0.69)	511/1 (0.20)	14 (0.59)	0.357/0,849
Erythromycin	571/179 (31.35)	505/155 (30.69)	420/157 (37.38)	434/92 (21.20)	513/121 (23.59)	704 (28.84)	0.004*/8,133
Fusidic Acid (STAFINE)	576/72 (12.50)	504/39 (7.74)	420/43 (10.24)	433/39 (9.01)	511/52 (10.18)	245 (9.93)	< 0.001*/102,682
Gentamicin	558/71 (12.72)	507/54 (10.65)	420/47 (11.19)	433/57 (13.16)	512/20 (3.91)	249 (10.33)	< 0.001*/27,336
Levofloxacin	574/48 (8.36)	507/19 (3.75)	420/9 (2.14)	434/26 (5.99)	512/31 (6.05)	133 (5.26)	0.143/2,136
Linezolid	568/5 (0.88)	505/3 (0.59)	420/1 (0.24)	433/3 (0.69)	512/2 (0.39)	14 (0.56)	0.316/1,002
Oxacillin	563/288 (51.15)	503/187 (37.18)	420/194 (46.19)	433/172 (39.72)	513/257 (50.10)	1098 (44.87)	0.729/0,119
Penicillin	546/530 (97.07)	481/473 (98.34)	389/384 (98.71)	411396 (96.35)	485/483 (99.59)	2266 (98.01)	< 0.002*/9,493
Ciprofloxacin	556/53 (9.53)	502/19 (3.78)	420/10 (2.38)	434/26 (5.99)	509/33 (6.48)	141 (5.63)	0.068/3,328
Teicoplanin	576/13 (2.26)	503/11 (2.19)	420/9 (2.14)	432/7 (1.62)	511/1 (0.20)	41 (1.68)	0.002*/9,049
Tetracycline	576/178 (30.90)	505/153 (30.30)	420/139 (33.10)	433/82 (18.94)	511/128 (25.05)	680 (27.66)	0.032*/4,587
Trimethoprim/sulfamethoxazole	530/1 (0.19)	502/24 (4.78)	420/10 (2.38)	434/23 (5.30)	509/18 (3.54)	76 (6.95)	< 0.001*/16,208
Vancomycin	576/0 (0.00)	502/0 (0.00)	420/0 (0.00)	432/0 (0.00)	509/0 (0.00)	0 (0.00)	-

Comparison of antibiotic resistance between 2018-2022. Chi-Square test; Fisher exact test; a:0.05; distributional difference is statistically significant; R: Resistant.

Table 3. Distribution of antibiotic resistance rates of *Klebsiella pneumoniae* by years.

ANTIBIOTICS	2018	2019	2020	2021	2022	TOTAL	*p/x^2
	n/R (%)	n/R (%)	n/R (%)	n/R (%)	n/R (%)	R (%)	
Ceftriaxone	762/534 (70.08)	726/491 (67.63)	599/419 (69.95)	844/585 (69.21)	1007/735 (72.99)	2764 (69.97)	0.178/1,812
Amoxicillin Clavunate	738/512 (69.38)	700/447 (63.86)	573/373 (65.10)	799/541 (67.71)	957/697 (72.83)	2570 (67.78)	0.118/2,432
Ertapenem	760/281 (36.97)	716/335 (46.79)	593/296 (49.92)	839/430 (51.25)	1005/591 (58.81)	1933 (48.75)	0.072/3,228
Gentamicin	763/316 (41.42)	728/216 (29.67)	600/202 (33.67)	844/345 (0.88)	1007/438 (43.50)	1517 (37.82)	0.380/0,768
Imipenem	760/96 (12.63)	720/89 (12.36)	597/152 (25.46)	825/278 (33.70)	1000/379 (37.90)	994 (24.41)	< 0.001*/139,921
Meropenem	756/157 (20.77)	713/253 (35.48)	590/211 (35.76)	844/322 (38.15)	1003/432 (43.07)	1375 (34.64)	< 0.001*/96,280
Piperacillin/Tazobactam	762/395 (51.84)	722/386 (53.46)	599/317 (52.92)	842/471 (55.94)	1005/642 (63.88)	2211 (55.60)	< 0.001*/25,926
Cefepim	760/513 (67.50)	498/361 (72.49)	350/275 (78.57)	498/381 (76.51)	614/518 (84.36)	2048 (75.88)	< 0.001*/51,567
Ceftazidime	763/492 (64.48)	651/469 (72.04)	522/414 (79.31)	715/587 (82.10)	846/726 (85.82)	2688 (76.75)	< 0.001*/99,252
Cefuroxime	385/306 (79.48)	405/338 (83.46)	350/289 (82.57)	496/398 (80.24)	612/541 (88.40)	1872 (82.83)	< 0.001*/14,705
Ciprofloxacin	738/361 (48.92)	723/419 (57.95)	593/331 (55.82)	840/542 (64.52)	1001/667 (66.63)	2320 (58.77)	< 0.001*/42,808
Trimethoprim/sulfamethoxazole	763/412 (54.00)	724/390 (53.87)	597/341 (57.12)	844/542 (64.22)	1006/676 (67.20)	2361 (59.28)	< 0.001*/31,929

*Comparison of antibiotic resistance between 2018-2022. Chi-Square test; Fisher exact test; a: 0.05; * distributional difference is statistically significant; R: Resistant.

in all other tested antibiotics, except ceftriaxone, amoxicillin clavulanate, ertapenem, and gentamicin, was statistically significant ($p < 0.005$) (Table 3).

A. baumannii exhibited high resistance to all tested antibiotics. In comparison to 2018, there was an upward trend in the resistance rates of all antibiotics in 2022. The most significant increase was observed in trimethoprim/sulfamethoxazole (from 55.81% in 2018 to 81.80% in 2022). The rise in resistance to amikacin, meropenem, and trimethoprim/sulfamethoxazole was statistically significant ($p < 0.005$) (Table 4).

Resistance rates in tested antibiotics, except levofloxacin (55.81%), were below 50% in *P. aeruginosa*. The most susceptible antibiotic was amikacin, with a resistance rate of 8.33%. In comparison to 2018, there was an increase in the resistance rates of all tested antibiotics in *P. aeruginosa* in 2022, except for piperacillin/tazobactam. However, when comparing 2021 and 2022, a decrease in resistance rates was observed in 2022 for some antibiotics that were high in 2021 (imipenem from 57.14% to 47.75%, meropenem from 44.63% to 36.07%, ceftazidime from 36.17% to 31.38%, ciprofloxacin from 46.58% to 41.87%). The increase in resistance to imipenem, cefepime, and ciprofloxacin was statistically significant ($p < 0.005$) (Table 5).

In *Enterobacter spp.,* the most resistant antibiotic was ampicillin with a 100% resistance rate for all five years, followed by amoxicillin clavulanate with a resistance rate of 98.68%. The most sensitive

Table 4. Distribution of antibiotic resistance rates of *Acinetobacter baumannii* by years.

ANTIBIOTICS	2018	2019	2020	2021	2022	TOTAL	*p/x^2
	n/R (%)	n/R (%)	n/R (%)	n/R (%)	n/R (%)	R (%)	
Amikacin	667/577 (86.50)	513/453 (88.30)	528/500 (94.69)	757/717 (94.71)	545/516 (94.67)	2763 (91.77)	< 0.001*/22,621
Gentamicin	665/627 (94.28)	512/471 (91.99)	528/503 (95.26)	761/733 (96.32)	545/519 (95.22)	2853 (94.61)	0.465/0,532
Imipenem	666/626 (93.99)	513/461 (89.86)	528/500 (94.70)	755/725 (96.02)	545/521 (95.59)	2833 (94.03)	0.157/1,996
Levofloxacin	-	145/121 (83.44)	522/496 (95.01)	757/725 (95.77)	544/522 (95.95)	1864 (92.54)	-
Meropenem	667/624 (93.55)	508/465 (91.53)	525/499 (95.04)	761/726 (95.40)	544/521 (95.77)	2835 (94.26)	0.062*/3,459
Ciprofloxacin	645/609 (94.41)	504/459 (91.07)	528/515 (97.53)	746/717 (96.11)	542/520 (95.94)	2820 (95.01)	0.225/1,468
Trimethoprim/Sulfamethoxazole	666/360 (55.81)	510/405 (79.41)	528/384 (72.72)	761/651 (85.54)	544/445 (81.80)	2245 (75.06)	< 0.001*/103,526

Comparison of antibiotic resistance between 2018-2022. Chi-Square test; Fisher exact test; a:0.05; distributional difference is statistically significant; R: Resistant.

Table 5. Distribution of antibiotic resistance rates of *Pseudomonas aeruginosa* by years.

ANTIBIOTICS	2018	2019	2020	2021	2022	TOTAL	*p/x^2
	n/R (%)	n/R (%)	n/R (%)	n/R (%)	n/R (%)	R (%)	
Amikacin	537/49 (9.12)	605/50 (8.26)	312/22 (7.05)	409/33 (8.07)	425/39 (9.18)	193 (8.33)	0.977/0,001
Gentamicin	535/128 (23.93)	597/101 (16.92)	311/55 (17.68)	-	-	284 (19.51)	-
Levofloxacin	-	443/256 (57.79)	311/151 (48.55)	400/228 (57.00)	424/254 (59.91)	889 (55.81)	-
Imipenem	537/211 (39.29)	605/257 (42.48)	313/123 (39.30)	406/232 (57.14)	423/202 (47.75)	1025 (45.19)	0.008*/6,911
Meropenem	532/167 (31.39)	568/204 (35.92)	310/99 (31.94)	410/183 (44.63)	427/154 (36.07)	807 (35.99)	0.127/2,324
Piperacillin/Tazobactam	533/197 (36.96)	580/190 (32.76)	313/79 (25.24)	409/116 (28.36)	427/150 (35.13)	732 (31.69)	0.557/0,344
Cefepim	521/190 (36.47)	494/181 (6.64)	250/87 (34.80)	348149 (42.82)	332/159 (47.89)	766 (39.72)	< 0.001*/10,945
Ceftazidime	530/159 (30.00)	586/192 (32.76)	310/105 (33.87)	412/149 (36.17)	427/134 (31.38)	739 (32.83)	0.644/0,212
Ciprofloxacin	499/167 (33.47)	564/273 (48.40)	303/134 (44.22)	395/184 (46.58)	418/175 (41.87)	933 (42.90)	0.008*/6,861

Comparison of antibiotic resistance between 2018-2022. Chi-Square test; Fisher exact test; a:0.05; distributional difference is statistically significant; R: Resistant.

Table 6. Distribution of antibiotic resistance rates of *Enterobacter spp.* by years.

ANTIBIOTICS	2018	2019	2020	2021	2022	TOTAL	*p/x^2
	n/R (%)	n/R (%)	n/R (%)	n/R (%)	n/R (%)	R (%)	
Amikacin	142/1 (0.70)	155/6 (3.87)	106/6 (5.66)	122/2 (1.64)	82/5 (6.10)	20 (3.59)	0.016*/5,800
Amoxicillin Clavunate	139/135 (97.12)	151/148 (98.01)	107/106 (99.07)	123/122 (99.19)	81/81 (100)	592 (98.68)	0.123/2,374
Ampicillin	142/142 (100)	154/154 (100)	109/109 (100)	123/123 (100)	82/82 (100)	610 (100)	-
Ceftriaxone	142/45 (33.69)	154/55 (35.71)	109/56 (51.38)	121/42 (34.71)	82/39 (47.56)	237 (40.61)	0.018*/5,586
Ertapenem	142/29 (20.42)	138/30 (21.73)	107/39 (36.45)	116/37 (31.90)	82/30 (36.59)	165 (29.42)	0.008*/6,999
Gentamicin	142/23 (16.19)	155/19 (12.26)	109/14 (12.84)	122/12 (9.84)	82/6 (7.32)	74 (11.69)	0.056/3,637
Imipenem	142/6 (4.23)	148/11 (7.43)	107/13 (12.15)	119/9 (7.56)	82/7 (8.54)	46 (7.98)	0.183/1,767
Levofloxacin	-	120/10 (8.33)	107/21 (19.63)	122/14 (11.48)	82/9 (10.98)	54 (12.61)	-
Meropenem	142/5 (3.52)	145/11 (7.58)	103/12 (11.65)	122/10 (8.20)	82/8 (9.76)	46 (8.14)	0.054/3,696
Piperacillin/Tazobactam	141/27 (19.01)	152/42 (27.63)	108/42 (38.89)	121/27 (22.31)	82/27 (32.93)	165 (28.15)	0.020*/5,363
Cefepim	142/31 (21.83)	111/31 (27.92)	75/33 (44.00)	80/18 (22.50)	56/10 (17.86)	123 (26.82)	0.534/0,386
Ceftazidime	142/33 (23.24)	131/55 (41.98)	100/51 (51.00)	98/43 (43.88)	65/35 (53.85)	217 (42.79)	< 0.001*/18,935
Ciprofloxacin	140/26 (18.57)	154/24 (15.58)	106/21 (19.81)	121/13 (10.74)	82/9 (10.98)	93 (15.13)	0.133/2,246
Trimethoprim/Sulfamethoxazole	142/27 (19.01)	154/20 (12.99)	109/17 (15.60)	122/15 (12.30)	82/10 (12.20)	89 (14.42)	0.185/1,752

Comparison of antibiotic resistance between 2018-2022. Chi-Square test; Fisher exact test; a:0.05; distributional difference is statistically significant; R: Resistant.

antibiotics were amikacin with a resistance rate of 3.59%, imipenem with a resistance rate of 7.98%, and meropenem with a resistance rate of 8.14%. Antibiotic resistance rates, which increased from 2018 to 2020, tended to decrease in 2021, and started to increase again in 2022. In comparison to 2018, *Enterobacter spp.* Showed increased resistance rates to all tested antibiotics in 2022, except for gentamicin, cefepime, ciprofloxacin, and trimethoprim/sulfamethoxazole. The increase in resistance in amikacin, ceftriaxone, ertapenem, piperacillin/tazobactam, and ceftazidime was statistically significant ($p < 0.005$) (Table 6).

Discussion

The results of this study showed that the most frequently isolated bacteria among the 15,272 ESKAPE clinical strains were *K. pneumoniae* (3,938, 27.79%), *A. baumannii* (3,013, 19.73%), and *E. faecium* (2,966, 19.24%), respectively (Figure 1). A study by Orosz *et al.* [9] reported similar findings with *K. pneumoniae, P. aeruginosa*, and *E. faecium* being the most frequently isolated bacteria, partially aligning with the current study. However, in a similar study by Ramsamy *et al.* [10], the order differed, with *S. aureus* (38.0%) being the most frequently isolated pathogen, followed by *K. pneumoniae* (22.2%) and *P. aeruginosa* (17.4%). Masoud *et al.* [4] found *P. aeruginosa* (25%) and *S. aureus* (17.1%) as the most frequently isolated, while Arbune *et al.* [11] reported *E. coli* (38.26%) and *S. aureus* (26%) from clinical samples. The variability in the dominant pathogens isolated has been attributed to factors such as immunosuppression status, comorbidity, and geographical location [12].

In this study, ESKAPE pathogens were most frequently isolated from urine cultures (3,263, 21.37%), followed by blood culture samples (3,099, 20.29%) (Figure 2). Similar studies, such as those by Orosz *et al.* and Benko *et al.* [3,9], also reported ESKAPE pathogens being most frequently isolated from urine cultures, followed by blood cultures. However, Arbune *et al.* [11] isolated ESKAPE pathogens from urine (45.6%) and wound cultures (35.9%), while Ramsamy *et al.* [10] found ESKAPE pathogens in blood (16.38%) followed by urine (13.94%) culture samples.

Data from the Centers for Disease Control and Prevention showed that the six ESKAPE pathogens are responsible for two-thirds of all healthcare-associated infections. Unfortunately, ESKAPE pathogens are steadily increasing in hospitals and are becoming increasingly resistant to many antimicrobial agents [5]. Antibiotic resistance is a global problem, limiting treatment options and increasing patient mortality and morbidity [13]. In this study, *E. faecium*, an ESKAPE pathogen, exhibited high resistance to ampicillin (93.77%), while the linezolid resistance rate was very low (2.52%). Although the resistance rate of vancomycin was 18.84%, a higher resistance rate was found for teicoplanin (62.52%) (Table 1). Ramsamy *et al.* [10] reported that all *E. faecium* strains were sensitive to vancomycin and linezolid. Benkő *et al.* [3] found resistance to be 21.2% for teicoplanin and 33.3% for vancomycin, with no linezolid resistance reported in 2020.

In the current study, when comparing 2018 with 2022, there was a decrease of 1.46% in ampicillin resistance, 3.04% in linezolid resistance ($p < 0.001$), 5.1% in teicoplanin resistance, and 3.29% in vancomycin resistance in *E. faecium*, while there was a 4.97% increase in streptomycin high-level resistance. The gentamicin high-level resistance rate remained almost the same (Table 1). Interestingly, a study on Enterococci in Italy reported opposite results. In the Italian study, when comparing 2015 to 2019, there was an increase in resistance rates for ampicillin (from 81.6% to 87.5%), linezolid (from 0% to 0.6%), teicoplanin (from 3.7% to 6.7%), and vancomycin (from 3.7% to 6.1%). However, a decrease was reported in the rate of resistance to streptomycin high level (from 74% to 64.5%) and gentamicin high level (from 60% to 54.2%) [14].

The 44.87% rate of methicillin resistance observed in *S. aureus* in the current study was close to the 46% resistance percentage (Table 2) determined by Perovic *et al.* [15]. MRSA rates have been reported to exceed 20% in all WHO regions and 80% in some countries [16]. The prevalence of MRSA in Africa varies from 12% in Tunisia to 82% in Egypt [17]. The National Health Laboratory Service (NHLS) public sector susceptibility data presented by Crowther-Gibson *et al.* [18] (January–December 2009) showed varying rates of MRSA.

In this study, the antibiotic to which *S. aureus* was most resistant was penicillin (98.01%), while vancomycin resistance was not found. Resistance rates were very low for daptomycin (0.59%), linezolid (0.56%), and teicoplanin (1.68%) (Table 2). In contrast, Arbune *et al.* [11] detected higher vancomycin (14.3%) and teicoplanin (12.2%) resistance rates but lower linezolid resistance (0.3%). The most resistant antibiotic among those tested was reported to be erythromycin (resistance rate 67.1%). Benkő *et al.* [3] did not detect resistance to linezolid, vancomycin, and teicoplanin, but found erythromycin to be the most resistant antibiotic (resistance rate 22%). In the current

study, compared to 2018, decreasing trends in antimicrobial resistance were observed in *S. aureus* against all antibiotics except penicillin and trimethoprim/sulfamethoxazole in 2022. Lugito *et al.* [19], in a similar study conducted between 2019 and 2022, found a decrease in all antibiotic resistance rates tested in *S. aureus*, consistent with the current study results. In studies conducted between 2011 and 2015, Ramsamy *et al.* [10] reported decreasing resistance rates in *S. aureus* strains in various antibiotic classes such as gentamicin, clindamycin, ciprofloxacin, erythromycin, and rifampicin, which is also consistent with the current study. It was reported that no resistance was observed to vancomycin, teicoplanin, and linezolid.

In the current study, *K. pneumoniae* was the most common Gram-negative bacteria isolated from the ESKAPE group (27.79%) (Figure 1). According to the 2015 global surveillance report by the Healthcare-associated Infections Surveillance Network and the United States National Healthcare Safety Network, 8.7% and 9.9% of all hospital-acquired infections were reported to be *K. pneumoniae* [20]. The steady rise in third-generation cephalosporin and amoxicillin-clavulanate antimicrobial resistance, indicative of inhibitor resistance and ESBL development in *K. pneumoniae*, is a matter of concern in this study. The WHO reported that the resistance to third-generation cephalosporins observed in *K. pneumoniae* is more than 30% worldwide and more than 60% in some countries [16].

In this study, the amoxicillin-clavulanate resistance rate increased from 69.38% in 2018 to 72.83% in 2022. Similarly, the resistance rates for cefepime, ceftazidime, cefuroxime, and ceftriaxone, which were 67.50%, 64.48%, 79.48%, and 70.08% in 2018, respectively, increased to 84.36%, 85.82%, 88.40%, and 72.99% in 2022 (Table 3). A study conducted in India reported a similar increase in resistance to the tested antibiotics in *K. pneumoniae* between 2018 and 2022 [21]. Lin *et al.* [22] also reported an increase in antibiotic resistance rates in *K. pneumoniae*. In 2006, the resistance rates for imipenem, cefazolin, gentamicin, tobramycin, ciprofloxacin, and ceftazidime were 2.33%, 27.91%, 16.28%, 13.95%, 18.60%, and 9.30%, respectively, while in 2020, they increased to 12.83%, 40.82%, 21.57%, 25.07%, 44.61%, and 17.78%, and a decrease was reported in piperacillin-tazobactam resistance from 13.95% to 13.70%. Lugito *et al.* [19] reported that resistance rates of *K. pneumoniae* against aminoglycoside, trimethoprim-sulfamethoxazole, penicillin-beta lactamase inhibitor, 3rd generation cephalosporin, quinolone, meropenem, and ertapenem peaked from 2019 to 2020 reaching over 50.0%, then decreased in 2021. The resistance rate was seen to be at its lowest level in 2022, ranging from 5% to 20%. The increase in carbapenem resistance in *K. pneumonia*e is also a major medical concern [23]. In the current study, an increase in resistance rates to carbapenem group antibiotics was observed in *K. pneumoniae* over 5 years. Ertapenem resistance increased from 36.97% in 2018 to 58.81% in 2022, imipenem resistance increased from 12.63% in 2018 to 37.90% in 2022, and meropenem resistance increased from 20.77% in 2018 to 43.07% in 2022. These carbapenem resistance rates are lower (Table 3) compared to a study conducted by Scaglione *et al*. [24] (74.9%), but higher than those reported by El-Kady *et al.* [25] (8.9%).

A. baumannii is among the most challenging pathogens among ESKAPE pathogens, capable of rapidly developing antibiotic resistance [26]. Resistance is increasingly emerging against nearly all routinely prescribed antimicrobial agents, including aminoglycosides, fluoroquinolones, and broad-spectrum β-lactams. Most strains exhibit resistance to cephalosporin-class antimicrobials, and resistance to carbapenems is becoming more frequent [27]. Once considered the primary treatment, carbapenems are no longer effective in controlling infections caused by this organism. The most significant consequence of infection with carbapenem-resistant *A. baumannii* is the need to resort to "last-resort" antibiotics such as colistin, polymyxin B, or tigecycline [28]. In this study, resistance rates of over 90% were observed for all tested antibiotics in *A. baumannii*, except for trimethoprim/sulfamethoxazole (75.06%). Compared to the rates in 2018, resistance to all antibiotics increased in 2022 (Table 4). This finding suggests that these antibiotics are not effectively used for the treatment of *A. baumannii* infections, and there are limited treatment options outside of last-resort antibiotics. In contrast to the current study, Lugito *et al.* [19], reported that with the exception of tigecycline, the resistance pattern of *A. baumannii* was above 50.0%, peaking from 2019 to 2020, then decreasing in 2021, and reaching 0.0% in most antimicrobials by 2022. Sannathimmappa *et al.* [29]. reported a decrease in antibiotic resistance rates over the years in *A. baumannii*, except for amikacin and tigecycline. Scaglione *et al.* [24] found lower resistance rate to carbapenems from 2015 to 2019 (74.4%) compared to their studies from 2010 to 2014 (100%).

P. aeruginosa is an opportunistic pathogen mainly causing nosocomial infections, which often *exhibits* high rates of antibiotic resistance [30,31]. In this study, an increase in resistance rates for all tested antibiotics in *P. aeruginosa*, except for piperacillin/tazobactam, was observed in 2022 compared to 2018. However, when comparisons were made between 2021 and 2022, some antibiotics that had high resistance rates in 2021 showed a decrease in resistance rates in 2022. Among the antibiotics with a high resistance rate in 2021 that decreased in 2022, imipenem decreased from 57.14% to 47.75%, meropenem from 44.63% to 36.07%, ceftazidime from 36.17% to 31.38%, and ciprofloxacin decreased from 46.58% to 41.87%. While the resistance levels found in *P. aeruginosa* in this study, ranging from 8.33% to 55.81%, (Table 5) are close to the resistance rates reported by Arabestani *et al.* [32], ranging from 9.6% to 61.29%, they are higher than the rates in the study conducted by Ramsamy *et al.* [10] (6.30% to 17.00%). In the current study, the most effective antibiotic against *P. aeruginosa* was amikacin (8.33%) (Table 5). Lari *et al.* [33] reported high resistance to all antibiotics except for colistin. Farhan *et al.* [34] found that imipenem was the most effective antibiotic. In a study by Lugito *et al.* [19], *P. aeruginosa* was reported to exhibit very high resistance rates to ceftriaxone, ertapenem, and tigecycline (88.9% - 100.0%), while the resistance rates to aminoglycosides, quinolones, imipenem, and meropenem had decreased from 2019 to 2021 and reached 0.0% in 2022. In contrast to the current study, Ramsamy *et al.* [10] observed decreasing trends in resistance rates for ceftazidime (from 17% to 13%, $p = 0.004$), piperacillin-tazobactam (from 27% to 21%, $p < 0.001$), meropenem (from 18% to 10%, $p < 0.001$), ciprofloxacin (from 22% to 18%, $p = 0.002$), and amikacin (from 10% to 8%, $p < 0.001$) in *P. aeruginosa* between 2013 and 2015.

Among ESKAPE pathogens, ampicillin resistance remained at 100% for five years in *Enterobacter* spp., (Table 6), which was isolated the least (4.00%) (Figure 1). The amoxicillin-clavulanate resistance rate was 97.12% in 2018 and gradually increased over the years, reaching 100% in 2022. The ceftazidime resistance rate increased from 23.24% in 2018 to 53.85% in 2022, and ceftriaxone increased from 33.69% to 47.56%. Compared to 2018, a decrease in gentamicin, cefepime, ciprofloxacin, and trimethoprim/sulfamethoxazole resistance rates in Enterobacter spp. was detected in 2022 (Table 6). Consistent with these findings, Intra *et al.* [35] reported decreased resistance to fluoroquinolones and aminoglycosides. Lugito *et al.* [19] found that the resistance rates of *Enterobacter spp*. against ceftazidime increased dramatically from 2019 to 2020 (from 0.0% to 60.0%), reaching 66.7% in 2021 and 2022. It was also reported that the rates of resistance to ceftriaxone and trimethoprim/sulfamethoxazole increased dramatically from 2019 to 2020, then decreased, and resistance to ciprofloxacin increased in 2020 and 2021, and then decreased in 2022. In a 4-year study by Lugito *et al*. [19], the resistance rate to levofloxacin, amikacin, meropenem, and fosfomycin was reported to be 0.0%.

In this study, ESKAPE pathogens were most frequently isolated in intensive care units (57.35%) at a statistically significant rate ($p < 0.001$) (Figure 3). This result is consistent with many studies reporting that ESKAPE pathogens are most frequently isolated from intensive care units [36,37]. This may be due to the advanced age of patients in intensive care units, immune suppression, prolonged hospitalization, intensive antibiotic treatments, and more invasive procedures [38].

Conclusions

According to the results of this study, the decrease in the resistance rates of most antibiotics tested against *E. faecium* and *S. aureus*, the absence of vancomycin resistance in *S. aureus*, and the low resistance rates in daptomycin (0.59%), linezolid (0.56%), and teicoplanin (1.68%) are encouraging. However, the increasing antimicrobial resistance trends of *K. pneumoniae, A. baumannii,* and *P. aeruginosa* against almost all antibiotics are alarming. In *Enterobacter spp*., antibiotic resistance, which increased from 2018 to 2020, tended to decrease in 2021, while resistance rates began to increase again in 2022, and an increase in resistance was detected in most antibiotics tested in 2022. Carbapenem resistance remains above 90% in *A. baumannii*, ranges from 24.41% to 48.75% in *K. pneumoniae*, and from 35.99% to 45.19% in *P. aeruginosa*, but resistance is increasing over the years. Unless the necessary precautions are taken, resistance will continue to increase, and the number of antibiotics used in treatment will decrease. The high rates of antimicrobial resistance observed in ESKAPE pathogens indicate the need to improve antimicrobial management and infection prevention and control programs. According to these results, surveillance studies should be carried out at regular intervals and data on sensitivity and resistance should be collected and strategies should be developed to control antibiotic resistance and ensure the use of more appropriate antibiotics. Healthcare personnel should be trained and hand hygiene should be emphasized. According to the

antibiogram results, rational antibiotic use should be ensured and unnecessary use of antibiotics should be prevented. There is also a need for further studies on this subject.

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Authors' contributions

Study concept and design: Orhan, Aral; data collection: Küçük, Kirişci; analysis and interpretation: Orhan, Altun; drafting of the manuscript: Orhan, Altun, Küçük; Critical revision of the text for important intellectual content: Aral, Kirişci; statistical analysis: Doğaner; Study supervision: Aral, Kirişci

References

1. WHO (2017) Thailand: national strategic plan on antimicrobial resistance 2017-2021. Available: https://www.who.int/publications/m/item/thailand-national-strategic-plan-on-antimicrobial-resistance-2017-2021. Accessed: 15 June 2023.
2. De Oliveira DMP, Forde BM, Kidd TJ, Harris PNA, Schembri MA, Beatson SA, Paterson DL, Walker MJ (2020) Antimicrobial resistance in ESKAPE pathogens. Clin Microbiol Rev 33: e00181-19. doi: 10.1128/CMR.00181-19.
3. Benkő R, Gajdács M, Matuz M, Bodó G, Lázár A, Hajdú E, Papfalvi E, Hannauer P, Péter Erdélyi P, Pető Z (2020) Prevalence and antibiotic resistance of ESKAPE pathogens isolated in the emergency department of a tertiary care teaching hospital in Hungary: a 5-year retrospective survey. Antibiotics 9: 624. doi: 10.3390/antibiotics9090624.
4. Masoud SS, Kovacevich A, Gangji R, Nyawale H, Nyange M, Ntukula A (2022) Extent and resistance patterns of ESKAPE pathogens isolated in pus swabs from hospitalized patients. Can J Infect Dis Med Microbiol 2022. doi: 10.1155/2022/3511306.
5. Rice LB (2008) Federal funding for the study of antimicrobial resistance in nosocomial pathogens: No ESKAPE. J Infect Dis 197: 1079-1081. doi: 10.1086/533452.
6. Mulani MS, Kamble EE, Kumkar SN, Tawre MS, Pardesi KR (2019) Emerging strategies to combat ESKAPE pathogens in the era of antimicrobial resistance: a review. Front Microbiol 10: 539. doi: 10.3389/fmicb.2019.00539.
7. WHO (2017) WHO publishes list of bacteria for which new antibiotics are urgently needed. Available: https://www.who.int/news/item/27-02-2017-who-publishes-list-of-bacteria-for-which-new-antibiotics-are-urgently-needed. Accessed: 30 May 2023.
8. Ramphal R (2005) Importance of adequate initial antimicrobial therapy. Chemotherapy 51: 171–176. doi: 10.1159/000086574.
9. Orosz L, Lengyel G, Ánosi N, Lakatos L, Burián K (2022) Changes in resistance pattern of ESKAPE pathogens between 2010 and 2020 in the clinical center of University of Szeged, Hungary. Acta Microbiol Immunol Hung 69: 27-34. doi: 10.1556/030.2022.01640.
10. Ramsamy Y, Essack SY, Sartorius B, Patel M, Mlisana KP (2018) Antibiotic resistance trends of ESKAPE pathogens in Kwazulu-Natal, South Africa: a five-year retrospective analysis. Afr J Lab Med 7: 887. doi: 10.4102/ajlm.v7i2.887.
11. Arbune M, Gurau G, Niculet E, Iancu AV, Lupasteanu G, Fotea S, Vasile MC, Tatu AL (2021) Prevalence of antibiotic resistance of ESKAPE pathogens over five years in an infectious diseases hospital from South-East of Romania. Infect Drug Resist 14: 2369-2378. doi: 10.2147/IDR.S312231.
12. Mawalla B, Mshana SE, Chalya PL, Imirzalioglu C, Mahalu W (2011) Predictors of surgical site infections among patients undergoing major surgery at Bugando Medical Centre in Northwerstern Tanzania. BMC Surgery 11: 21.
13. Armin S, Fallah F, Karimi A, Shirvani F, Azimi L, Tehrani NA, Abdollahi N, Mobasseri P, Rajabnejad M, Ghanaiee RM, Afatemi SMH, Fahimzad SA, Karami N, Tajbakhsh M, Ghandchi G, Tabatabaei SR (2023) Prevalence and antimicrobial resistance patterns in ESKAPE pathogens in Iran. Arch Pediatr Infect Dis 11: e129629. doi: 10.5812/pedinfect-129629.
14. Boccella M, Santella B, Pagliano P, De Filippis A, Casolaro V, Galdiero M, Borrelli A, Capunzo M, Boccia G, Franci G (2021) Prevalence and antimicrobial resistance of *Enterococcus* species: a retrospective cohort study in Italy. Antibiotics 10: 1552. doi: 10.3390/antibiotics10121552.
15. Perovic O, Iyaloo S, Kularatne R, Lowman W, Bosman N, Wadula J, Seetharam S, Duse A, Mbelle N, Bamford C, Dawood H, Mahabeer Y, Bhola P, Abrahams S, Moodley AS (2015) Prevalence and trends of *Staphylococcus aureus* bacteraemia in hospitalized patients in South Africa, 2010 to 2012: Laboratory-based surveillance mapping of antimicrobial resistance and molecular epidemiology. PLoS One 10: e0145429. doi: 10.1371/journal.pone.0145429.
16. World Health Organization (2014) Antimicrobial resistance: global report on surveillance. Available: https://iris.who.int/bitstream/handle/10665/112642/9789241564748_eng.pdf?sequence=1. Accessed: 12 August 2023
17. Falagas ME, Karageorgopoulos DE, Leptidis J, Korbila IP (2013) MRSA in Africa: filling the global map of antimicrobial resistance. PloS One 8: e68024. doi:10.1371/journal.pone.0068024.
18. Crowther-Gibson P, Govender N, Lewis DA, Bamford C, Brink A, von Gottberg A, Klugman K, du Plessis M, Fali A, Harris B, Keddy K, Botha M (2011) Part IV. Human infections and antibiotic resistance. S Afr Med J 101: 567-78.
19. Lugito NPH, Cucunawangsih C, Wiwing V, Suryadinata N (2023) Prevalence of antimicrobial resistance of ESKAPE pathogens before and during pandemic COVID-19 pandemic in a university affiliated hospital in Tangerang, Indonesia. Open J Med Microbiol 13: 146-158. doi: 10.4236/ojmm.2023.132013.
20. Perez F, Villegas MV (2015) The role of surveillance systems in confronting the global crisis of antibiotic-resistant bacteria. Curr Opin Infect Dis 28: 375–383. doi: 10.1097/QCO.0000000000000182.
21. Sharma A, Thakur A, Thakur N, Kumar V, Chauhan A, Bhardwaj N (2023) Changing trend in the antibiotic resistance pattern of *Klebsiella pneumonia* isolated from endotracheal aspirate samples of ICU patients of a tertiary care hospital in north India. Cureus 15: e36317. doi: 10.7759/cureus.36317.
22. Lin Z, Yu J, Liu S, Zhu M (2022) Prevalence and antibiotic resistance of *Klebsiella pneumoniae* in a tertiary hospital in Hangzhou, China, 2006–2020. J Int Med Res 50: 03000605221079761. doi: 10.1177/03000605221079761.

23. Madni O, Amoako DG, Abia ALK, Rout J, Essack SY (2021) Genomic investigation of carbapenem-resistant *Klebsiella pneumonia* colonization in an intensive care unit in South Africa. Genes 12: 951. doi: 10.3390/genes12070951.
24. Scaglione V, Reale M, Davoli C, Mazzitelli M, Serapide F, Lionello R, Gamba VL, Fusco P, Bruni A, Procopio D, Garofalo E, Longhini F, Marascio N, Peronace C, Giancotti A, Gallo L, Matera G, Liberto MC, Cesana BM, Costa C, Trecarihi EM, Quirino A, Torti C (2022) Prevalence of antibiotic resistance over time in a third-level university hospital. Microb Drug Resist 28: 425-435. doi: 10.1089/mdr.2021.0109
25. El-Kady R, Karoma S, Atrouni AA (2022) Multidrug-resistant Gram-negative ESKAPE pathogens from a tertiary-care hospital: prevalence and risk factors. Egypt J Med Microbiol 31: 135-142. doi: 10.21608/EJMM.2022.256008.
26. Xie R, Zhang XD, Zhao Q, Peng B, Zheng J (2018) Analysis of global prevalence of antibiotic resistance in *Acinetobacter baumannii* infections disclosed a faster increase in OECD countries. Emerg Microbes Infect 7: 31. doi: 10.1038/s41426-018-0038-9.
27. Sohail M, Rashid A, Aslam B, Waseem M, Shahid M, Akram M, Khurshid M, Rasool MH (2016) Antimicrobial susceptibility of *Acinetobacter* clinical isolates and emerging antibiogram trends for nosocomial infection management. Rev Soc Bras Med Trop 49: 300-304. doi: 10.1590/0037-8682-0111-2016.
28. Nath H, Barkataki D (2016) Prevalence of ESBL and MBL producing *Acinetobacter* isolates in clinical specimens in tertiary care hospital, Assam, India. Int J Curr Microbiol App Sci 5: 515-522. doi: 10.20546/ijcmas.2016.511.060.
29. Sannathimmappa MB, Nambiar V, Aravindakshan R (2021) Antibiotic resistance pattern of *Acinetobacter baumannii* strains: a retrospective study from Oman. Saudi Journal Med Med Sci 9: 256-260. doi: 10.4103/sjmms.sjmms_855_20.
30. Horna G, Quezada K, Ramos S, Mosqueda N, Rubio M, Guerra H, Ruiz J (2019) Specific type IV pili groups in clinical isolates of *Pseudomonas aeruginosa*. Int Microbiol 22: 131-141. doi: 10.1007/s10123-018-00035-3.
31. Li F, Chen D, Li L, Liang D, Wang F, Zhang B (2020) Analysis of metallo-β-lactamases, *oprD* mutation, and multidrug resistance of β-lactam antibiotic-resistant strains of *Pseudomonas aeruginosa* isolated from Southern China. Curr Microbiol 77: 3264-3269. doi: 10.1007/s00284-020-02148-3.
32. Arabestani MR, Rajabpour M, Mashouf RY, Alikhani MY, Mousavi SM (2015) Expression of efflux pump MexAB-OprM and OprD of *Pseudomonas aeruginosa* strains isolated from clinical samples using qRT-PCR. Arch Iran Med 18: 102-108.
33. Lari AR, Azimi L, Soroush S, Taherikalani M (2015) Low prevalence of metallo-β-Lactamase in *Pseudomonas aeruginosa* isolated from a tertiary burn care center in Tehran. Int J Immunopathol Pharmacol 28: 384-389. doi: 10.1177/0394632015578343.
34. Farhan SM, Ibrahim RA, Mahran KM, Hetta HF, El-Baky RMA (2019) Antimicrobial resistance pattern and molecular genetic distribution of metallo-βlactamases producing *Pseudomonas aeruginosa* isolated from hospitals in Minia, Egypt. Infect Drug Resist 12: 2125-2133. doi: 10.2147/IDR.S198373.
35. Intra J, Carcione D, Sala RM, Siracusa C, Brambilla P, Leoni V (2023) Antimicrobial resistance patterns of *Enterobacter cloacae* and *Klebsiella aerogenes* strains isolated from clinical specimens: a twenty-year surveillance study. Antibiotic*s* 12: 775. doi: 10.3390/antibiotics12040775.
36. Flores-Paredes W, Luque N, Albornoz R, Rojas N, Espinoza M, Pons MJ, Ruiz J (2021) Evolution of antimicrobial resistance levels of ESKAPE microorganisms in a Peruvian IV-level hospital. J Infect Chemother 53: 449-462. doi: 10.3947/ic.2021.0015.
37. Llaca-Díaz JM, Mendoza-Olazarán S, Camacho-Ortiz A, Flores S, Garza-González E (2012) One-year surveillance of ESKAPE pathogens in an intensive care unit of Monterrey, Mexico. Chemotherapy 58: 475-481. doi: 10.1159/000346352.
38. Ture Z, Güner R, Alp E (2022) Antimicrobial stewardship in the intensive care unit. J Intensive Med 3: 244-253. doi: 10.1016/j.jointm.2022.10.001.

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