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Original Article

Microorganisms grown in urine cultures and antimicrobial resistance patterns: A randomised retrospective analysis from a tertiary hospital

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Abstract

Introduction: Patients in intensive care units (ICU) are 5-7 times more susceptible to infection than other groups, which increases the prevalence of hospital-acquired infections and associated sepsis accounting for 60% of deaths. Gram-negative bacteria are the most common source of urinary tract infections that cause morbidity, mortality, and sepsis in the ICU. The aim of this study is to detect the most commonly grown microorganisms and antibiotic resistance in urine cultures in the intensive care units of our tertiary city hospital, which has more than 20% of the ICU beds in Bursa By this way, we suppose that we will contribute to surveillance studies in our province, our country.

Methodology: Patients who were admitted to Bursa City Hospital adult ICU for various reasons between 15.07.2019 and 31.01.2021 and had growth in urine cultures were retrospectively screened. The urine culture result, growing microorganism, antibiotic used, and resistance status were recorded and analyzes were performed according to hospital data.

Results: Gram-negative growth was observed in 85.6% (n = 7707), gram-positive growth in 11.6% (n = 1045), and candida fungus growth in 2.8% (n = 249). Resistance to at least one antibiotic in urine cultures was observed for *Acinetobacter* (71.8), *Klebsiella* (51%), *Proteus* (47.95%), *Pseudomonas* (33%), *E. coli* (31%) and *Enterococci* (26.75%), respectively.

Discussion: Developing a health system leads to longer life expectancy, longer intensive care treatment, and more frequent interventional procedures. In terms of being a resource for empirical treatments, early initiation of empirical treatments to control the urinary tract infection disrupts the patient's hemodynamics and increases mortality and morbidity.

Key words: Urine culture; antibiotic resistance; intensive care unit.

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Introduction

Intensive care units (ICU) are specialized units of hospitals where high-level medical treatment, mechanical ventilation support, hemodynamic imaging, nutritional support, specialty drugs are used, and advanced antibiotics are applied [1]. It is referred to as the epicenter of infections due to the increased risk of infection and increased hospitalization of comorbid patients in these specialized units where such important services are provided and frequent use of invasive procedures that cross body barriers [2].

It is estimated that there are more than 150 million urinary tract infections per year worldwide. It has been reported that the annual number of urinary tract cases reaching a doctor in the USA is 8 million. Urinary tract infections, which are important causes of morbidity and mortality in all genders, are the most common of all bacterial infections in women [3-7].

Gram-negative bacteria are the most common source of urinary tract infections that cause morbidity, mortality, and sepsis in the ICU [8]. *E. coli, Enterobacter* sps, *Klebsiella* sps, *Acinetobacter* sps, *Pseudomonas aeruginosa* are the most common urinary infection agents [8,9]. The presence of treatment-resistant strains such as *E. coli* and *Klebsiella* sps among these microorganisms makes antibiogram tests more important to determine antibiotic susceptibility [10].

American Society of Infectious Diseases (IDSA) recommends clinical, hospital, local, regional, and national surveillance studies for the attending physicians, to obtain information about the local antibiotic resistance rates against the infectious agents in the hospitals where they work and to make the empirical treatment of the pathogens causing urological infections appropriate [7]. It has been shown in studies worldwide that the activity of Sulfometaxozole (STX) against *E. coli* urinary samples varies according to geographical location and this has made it more crucial to conduct surveillance studies at the hospital scale [11,12].

The aim of this study is to detect the most commonly grown microorganisms and antibiotic resistance in urine cultures in the intensive care units of our tertiary city hospital, which has more than 20% of the ICU beds in Bursa [13]. In this way, we suppose that we will contribute to surveillance studies in our province, our country, and perhaps globally.

Methodology

Patients who were admitted to Bursa City Hospital (BCH) adult ICU for various reasons between 15.07.2019 and 31.01.2021 and had growth in urine cultures were randomly and retrospectively screened. The patients' age, gender, laboratory data (CPR, WBC, PLT, BUN, creatine, urinary leukocyte, urine pH), urine culture result, growing microorganism, antibiotic used and resistance status were recorded and analyzes were performed according to hospital data. Those whose data could not be reached, patients under 18 years of age and pregnant women were excluded from the study. Local ethics committee approval for the study was obtained from the BCH Clinical Research Ethics Committee.

Statistical analysis

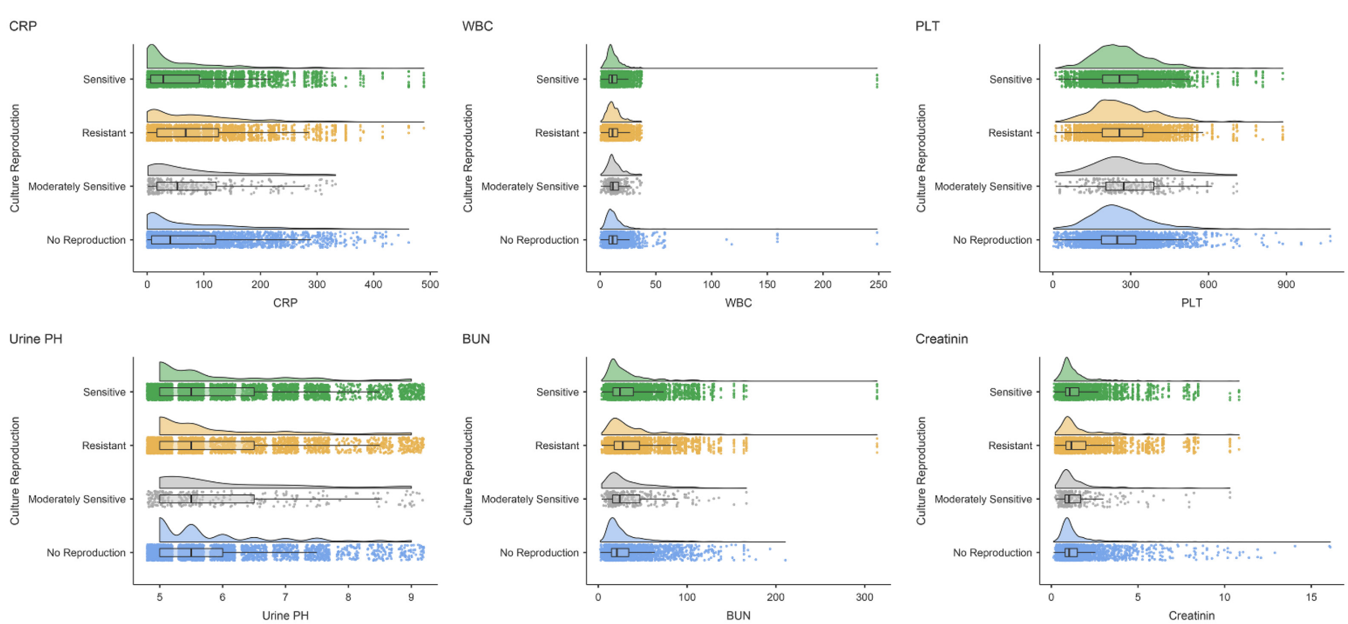
The findings of the study are evaluated using ‘The Jamovi project (2021), Jamovi (Version 2.0.0) [Computer Software]. Descriptive statistical methods (mean, standard deviation, median, frequency, percentage, minimum, maximum) were used while evaluating the study data. The Mann-Whitney U test was used for comparisons between two groups of quantitative variables without normal distribution. Pearson chi-square test and Fisher Exact test were used to compare qualitative data. One-way ANOVA test and the Kruskal-Wallis test were used to investigate whether there was a significant difference between more than two independent groups according to the arithmetic mean. Post-hoc Tukey and Tamhane tests were used to investigate which groups caused the significant difference between the groups. Pearson test was used for parametric variables and the Spearman test was used for nonparametric variables during correlation analysis. Statistically, $p < 0.05$ was considered significant at the 95% confidence interval.

Results

Data from 19,562 patients, who were hospitalized in the BCH adult ICU during the date range specified in the study plan and who were also examined and treated for UTI during their treatment, were collected. Within this patient population, 12,560 patients were included in the study, 6,924 of these patients were excluded from the study due to deficiencies in the file data and 78 of them were pregnant.

In terms of the gender distribution of the patients included in the study, 47% (n = 5,904) of the patients were male and 53% (n = 6,656) were female. Of the continuous variables, mean age value was 69.6 ± 14.6, mean CRP was 74.6 ± 84.6, mean WBC was 12.6 ± 9.35, mean PLT was 270 ± 119, mean BUN was 33.5 ±

Figure 1. Culture Reproduction Distributions of Continuous Variables.



27.9, mean creatine was 1.54 ± 1.39, mean urinary leukocytes was 148 ± 706, mean urine pH was 5.87 ± 1.1. Figure 1 shows distribution details (mean, SD, box plot, and value accumulation curve) of CRP, WBC, PLT, BUN, creatine, urinary leukocytes, and urinary PH values, which are the continuous variables in our study, according to the culture growth types.

Regarding the urine culture results of our patients, while in 71.7% (n = 9,001) of the patients, growth was detected, in 28.3% (n = 3,559) growth was not detected. The highest growth in urine cultures was observed for *Escherichia coli* (28.3%) (n = 3550), *Klebsiella* spp (14.9%) (n = 1,867) secondly, *Acinetobacter* spp (8.2%) (n = 1,033) as the third. While 79.7% (n = 5,303) of female patients had growth in urine culture, this rate was 62.6% (n = 3,698) in males. A statistically significant difference was found between men and women in terms of whether there was a growth in urine culture ($p < 0.001$) (Table 1).

When the microorganisms with at least one antibiotic resistance were evaluated, it was observed that there were 3,566 resistant, 5,133 susceptible, and 302 intermediate-sensitive microorganisms. When antibiotic resistance was examined in all patients, it was found that 30.4% (n = 1,797) in men, 50.1% (n = 3336) in women were infected with susceptible bacteria, while resistant bacteria were 29.6% (n = 1,747) in men and 27.3% (n = 1,819) in women. A statistically significant difference was found between the genders in terms of antibiotic resistance ($p < 0.001$) (Table 2).

Regarding the resistance distribution of microorganisms, with at least one antibiotic resistance, grown in urine cultures, there are 40.9% (n = 5,133) susceptible microorganisms, and *E. coli* ranks first with 19% (n = 2,387). Resistant microorganisms take their place in the distribution with 28.4% (n = 3,566) and *E. coli* is in the first place in this group with 8.7% (n = 1,095). *Candida albicans* were found to be 1.2% (n = 151) susceptible and 0.1% (n = 10) resistant to the treatment. During the assessment of the distributions of resistance to drugs, cephalosporin ranked first with 17% (n = 606), of the most common resistance. Colistin, on the other hand, ranked first among sensitive drugs with 8.8% (n = 453).

It was observed that 7,408 of the patients had a urinary catheter inserted, and a significant difference was found in terms of drug resistance and growth of resistant microorganisms with the patients who did not have a urinary catheter ($p < 0.001$, $p < 0.001$).

A significant difference was found in terms of drug resistance and growth of resistant microorganisms in patients hospitalized for more than 7 days in terms of the number of intensive care unit stays ($p < 0.001$, $p < 0.001$).

Within the isolates in our study, gram-negative growth was observed in 85.6% (n = 7707), gram-positive growth in 11.6% (n = 1045), and candida fungus growth in 2.8% (n = 249).

In our study, the distribution of microorganism species was as follows; *E. coli* in 39.4% (n = 3550), *Klebsiella* in 20.7% (n = 1867), *Acinetobacter* in 11.5% (n = 1033), *Pseudomonas* in 7.6% (n = 981). Among the gram-positive agents, the most frequent growth detected was *Enterococci* in 7.7% (n = 692).

Table 1. Microorganism Resistance Distributions.

Microorganism	Sensitive	Resistant	Intermediate Sensitive	No proliferation	Total	Incidence rate
Alcaligenes faecalis ssp faecalis	3	8	0	0	11	0.122208643
C. albicans	151	10	2	0	163	1.810909899
C. krusei	10	2	0	0	12	0.13331852
C. tropicalis	72	1	1	0	74	0.822130874
Citrobacter freundii	51	6	1	0	58	0.644372847
Elizabethkingia meningoseptica	1	13	1	0	15	0.16664815
Escherichia coli	2387	1095	68	0	3550	39.44006222
Morganella morganii ssp morganii	5	7	1	0	13	0.144428397
Proteus mirabilis	87	94	15	0	196	2.177535829
Providencia stuartii	9	14	3	0	26	0.288856794
Pseudomonas aeruginosa	398	229	54	0	681	7.565826019
Stenotrophomonas maltophilia	3	0	0	0	3	0.03332963
Acinetobacter	207	781	45	0	1033	11.47650261
Enterobacter	100	70	8	0	178	1.977558049
Enterococci	456	185	51	0	692	7.688034663
Klebsiella	873	946	48	0	1867	20.74213976
Serratia	54	21	1	0	76	0.844350628
Staphylococci	222	79	3	0	304	3.377402511
Streptococci	44	5	0	0	49	0.544383957

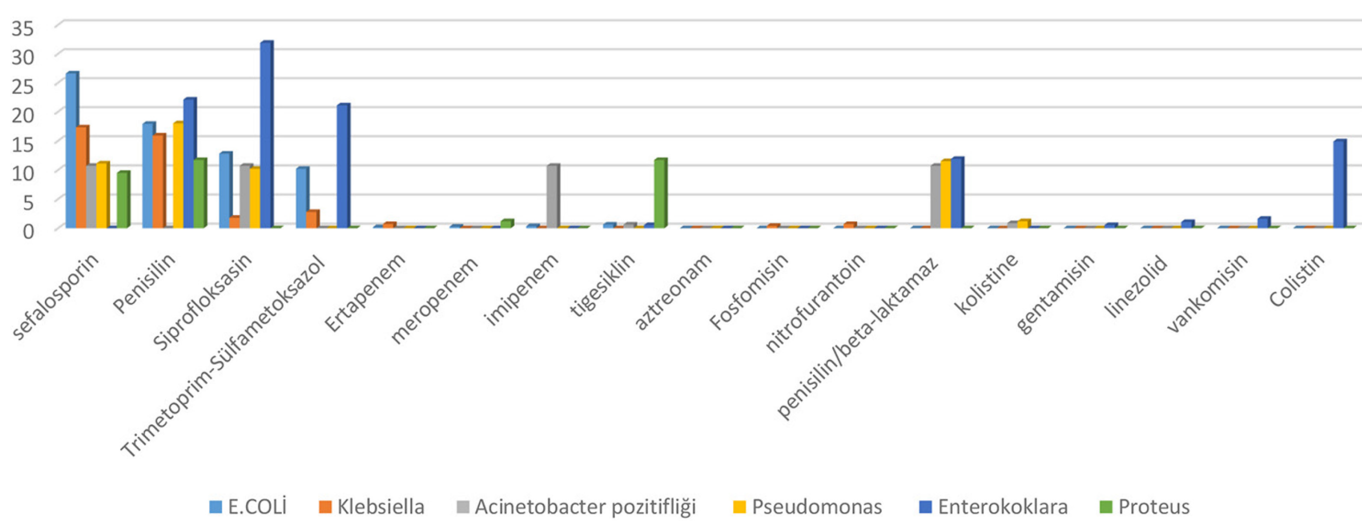
$\chi^2 = 14284$; $p < 0.001$.

Table 2. Antibiotic Resistance Distributions.

Antibiotic name *	Sensitive	Resistant	Intermediate Sensitive	No proliferation	Total
Colistin	453	51	1	0	505
Tigecycline	414	48	64	0	526
Tobramycin	283	217	0	0	500
Imipenem	343	177	28	0	548
Cephalosporin	438	606	37	0	1081
Ciprofloxacin	232	418	20	0	670
Penicillin-beta-lactamase	358	340	9	0	707
Gentamicin	373	202	2	0	577
Amikacin	413	122	25	0	560
Trimethoprim/Sulfamethoxazole	270	307	50	0	627
Nitrofurantoin	53	7	0	0	60
Fosfomycin	73	14	1	0	88
Meropenem	258	134	19	0	411
Ertapenem	50	9	0	0	59
Penicillin	243	522	10	0	775
Aztreonam	195	203	13	0	411
Vancomycin	124	3	0	0	127
Teicoplanin	101	7	0	0	108
Linezolid	121	3	0	0	124
Fluconazole	39	5	1	0	45
Amphotericin B	43	3	0	0	46
Micafungin	43	2	0	0	45
Voriconazole	44	1	1	0	46
Caspofungin	44	1	0	0	45
Fusidic Acid	6	6	0	0	12
Tetracycline	6	10	0	0	16
Daptomycin	13	1	0	0	14
Levofloxacin	79	144	20	0	243
Flucytosine	20	1	1	0	22
Mupirocin	1	0	0	0	1
Azithromycin	0	2	0	0	2
Total	5133	3566	302	3559	12560
Gender **	**Sensitive**	**Resistant**	**Intermediate Sensitive**	**No proliferation**	**Total**
Male	1797	1747	154	2206	5904
Female	3336	1819	148	1353	6656
Total	5133	3566	302	3559	12560

* $\chi^2 = 15083$, $p < 0.001$; ** $\chi^2 = 625$, $p < 0.001$.

Figure 2. Microorganism Antibiotic Resistance Distributions.



Resistance to at least one antibiotic in urine cultures was observed for *Acinetobacter* (71.8), *Klebsiella* (51%), *Proteus* (47.95%), *Pseudomonas* (33%), *E. coli* (31%) and *Enterococci* (26.75%), respectively.

In our cultures with *E. coli* growth, Cephalosporin was the antibiotic with the highest resistance (26.6%) for *E. coli*, followed by Penicillin (17.9%), Ciprofloxacin (12.78%), Trimethoprim-Sulfamethoxazole (10.2%), and the lowest resistance was for Ertapenem (0.17%), Meropenem (0.26%), Imipenem (0.35%) and Tigecycline (0.62%) (Figure 2).

Regarding the resistance for *Klebsiella*, the highest resistance was observed for Cephalosporin (17.3%), Penicillin (15.9%) and its derivatives (10%), and Aztreonam (6.38%). The least resistance for *Klebsiella* was detected in Fosfomycin (0.4%), Ertapenem (0.71%), and Nitrofurantoin (0.71%) (Figure 2).

In our cultures with *Acinetobacter* growth, the most common antibiotic resistance was determined for Penicillin/Beta-lactamase (10.71%), Cephalosporin (10.71%), Ciprofloxacin (10.71%) and Imipenem (10.71%). The least resistance for *Acinetobacter* was observed against Tigecycline (0.62%) and Colistin (0.87%) (Figure 2).

Considering the resistance rate for *Pseudomonas*, Penicillin was the antibiotic with the highest resistance (18%) followed by Penicillin/Beta-lactamase (11.5%), Cephalosporins (11.1%), Ciprofloxacin (10.2%), and the lowest resistance was seen against Colistin (1.2%) (Figure 2).

Ciprofloxacin was found to be the antibiotic with the highest resistance for *Enterococci* (31.9%), followed by Penicillin (22.1%), Trimethoprim/sulfamethoxazole (21.1%), Penicillin/Beta-lactamase (11.9%), and the lowest resistance was for Gentamicin (0.54%), Tigecycline (0.54%), Linezolid (1.08%) and Vancomycin (1.62%). Furthermore, we found that Colistin was the antibiotic with the highest resistance (14.9%) for Proteus, in our study, followed by penicillin (11.7%), tigecycline (11.7%), cephalosporins (9.5%), and the lowest resistance was found against meropenem (1.2%) (Figure 2).

Discussion

UTI has been reported to be one of the most common intensive care-acquired infections in the world, accounting for 40% of nosocomial infections in ICU, and 80% of these infections are related to urinary instrumentation [14,15]. During the 3-year period of the study, 34% (N = 9001) of 26,445 patients were hospitalized in the ICU, and 58.9% (n = 7,402) of these infections were in the cases with urinary catheters. Currently, urinary system infection was found to be the most common cause among intensive care infections in our study, and the most common cause was urinary catheterization, which is consistent with the literature [16,17].

Either in outpatient or inpatient follow-ups, UTI is more prevalent in women with [17,18]. In our study, the percentage of urinary culture positivity in ICU is 58% (n = 5,329) in females and 42% (n = 3,865) in males, which has been known for a long time related to the shortness of the urethra in females [19]. Due to this anatomical difference, avoiding urinary catheterization in women as much as possible and paying attention to general hygiene rules may be a step to prevent urinary tract infections.

In our study, we found a significant relationship between prolonged hospitalization and the possibility of developing positivity in urinary culture. It has been reported that the length of stay in the intensive care unit is a risk factor for the development of many infections, which has been confirmed in the literature previously [20]. Prolonged hospitalization may cause an increased number of interventional procedures, longer antibiotic use during empirical treatments, and more antibiotic use may lead to increased antibiotic resistance.

A single agent has grown in 100% of our patients with positive urine culture, and evidence from the world shows that single agent growth is dominant in urine cultures with growth [21]. Our culture, in which more than one microorganism grows, is not found in isolates and this supports the previous publications suggesting urinary tract infection is of a mono-microorganism nature [22].

We found Gram-negative bacteria as the most common bacterial group in our breeding cultures. The fact that Gram-negative bacteria is the most common cause of urinary tract infections was previously reported by Mazzariol *et al.* [23]. In the isolates in our study, gram-negative growth was 85.6% (n = 7707), gram-positive growth was 11.6% (n = 1045), *Candida* fungus growth was observed in 2.8% (n = 249). In a previous study, it was reported that in cultures with positive urinary tract infection, the responsible agents were Gram-negative in 47.6%, Gram-positive in 25.6%, and *Candida* in 26.8% of the cultures [10]. We think that the presence of a Gram-negative growth higher than in the literature could be due to the difference in the general flora of our hospital. The increased Gram-negative growth in our study and the literature might be due to the fact that this type of microorganism settles and reproduce more frequently and rapidly in the urinary tract. Considering the proximity of the gram-negative bacteria group, which loves the intestinal flora, to the urinary system, planning medical interventional procedures may contribute to the prevention of such infections.

Among 9,001 cultures where the microorganism grew, the *Enterobacter* family was the most common factor. As a well-established finding in the literature, the most common cause of urinary tract infections that have been reported previously is *Enterobacter* family [24,25]. In our study, the distribution of this family of microorganisms was as follows; *E. coli* in 39.4% (n = 3,550), *Klebsiella* in 20.7% (n = 1,867), *Acinetobacter* in 11.5% (n = 1,033), *Pseudomonas* in 7.6% (n = 981). Among the Gram-positive growths, the most frequent growth was *Enterococci* at 7.7% (n = 692). In a former study, the frequency of *E. coli* was 56.6%, *Klebsiella* 9.4%, *Proteus* 9.4%, *Enterobacteria* 3.8%, *Pseudomonas* 5.8%, and *Acinetobacter* 0%, determined [24]. In another study, *E. coli* growth was found in 46.1%, *Klebsiella* in 13.5%, *Proteus* in 7.8%, *Enterobacteria* in 3.6%, *Pseudomonas* in 15.5% and *Acinetobacter* in 1% [25]. We attribute the fact that the most common factor is *E. coli* in many studies and the remaining ranking differs between studies, to the populations included in the studies being selected from groups with different characteristics and variations of flora in clinics, hospitals, provinces, and the countries.

In our cultures with *E. coli* growth, resistance to at least one antibiotic was observed at a rate of 31%. Cephalosporin was the antibiotic with the highest resistance (26.6%) for *E. coli*, in our study, followed by Penicillin (17.9%), Ciprofloxacin (12.78%), Trimethoprim-Sulfamethoxazole (10.2%), and the lowest resistance was for Ertapenem (0.17%), Meropenem (0.26%), Imipenem (2%). 0.35) and Tigecycline (0.62%). In a study on the antibiotic resistance of isolates obtained from urine cultures in Turkey, for *E. coli*, resistance was found to be 17.53% for Penicillin/Beta-lactamase derivatives, 47% for Co-trimoxazole. 31% for Ciprofloxacin, 5.9% for Amikacin, and 17.5% for Gentamicin, 1% for Imipenem, and with 0% resistance was found for Meropenem being the most sensitive antibiotic [26]. In many studies in our country, Imipenem and Meropenem were found to be the most sensitive antibiotics for *E. coli* [26,27].

Resistance to at least one antibiotic was observed in 51% of cultures with *Klebsiella* microorganism. The highest resistance was observed for Cephalosporin (17.3%), Penicillin (15.9%), derivatives (10%), Aztreonam (6.38%). The least resistance for *Klebsiella* was seen in Fosfomycin (0.4%), Ertapenem (0.71%), and Nitrofurantoin (0.71%). In studies conducted on isolates containing Klebsiella, Penicillin/Beta-lactamase resistance was found to be 6.6%-15%, Cephalosporin resistance was 9.9-29.7%, Imipenem resistance was found to be 1.7-3%. The most sensitive antibiotic was imipenem (98%), Meropenem (99%), and it was comparable to our study [26,28].

Acinetobacter growth was observed in 11.5% of our cultures. In a study conducted in Europe, the rate of *Acinetobacter* growth was reported to be 2.6% [29], and in a study conducted in Spain it was reported to be 6.7% [30]. In a recent study conducted in Turkey, it was reported that it was the most common isolate in infections in ICU [31]. In our cultures with *Acinetobacter* positivity, the rate of resistance to at least one antibiotic was 75.8%, and the most common antibiotic resistance was determined for Penicillin/Beta-lactamase (10.71%), Cephalosporin (10.71%), Ciprofloxacin (10.71%) and Imipenem (10.71%). The least resistance for *Acinetobacter* was observed against Tigecycline (0.62%) and Colistin (0.87%). Uzun *et al.* reported that the most sensitive antibiotics to *Acinetobacter* species were Colistin (100%), Tigecycline (97%), Netilmicin (66.7%), and Tobramycin (70.7%); Imipenem resistance was 86.5%, Meropenem resistance was 94.5%, and Amikacin resistance was 89.3% [31]. *Acinetobacter* with an increasing incidence seems to be more problematic with antibiotic resistance from year to year. In a previous study conducted in our country, the antibiotics with the highest resistance reported for *Acinetobacter* species were Cephalosporins and Ciprofloxacin. The antibiotic most sensitive has been reported to be Colistin [32]. It is pleasing that our finding is compatible with the literature, with the increasing frequency of *Acinetobacter*, it is still sensitive to Colistin.

In our cultures with *Pseudomonas* growth, resistance to at least one antibiotic was observed at a rate of 33%. Penicillin was found to be the antibiotic with the highest resistance (18%) for *Pseudomonas* in our study, followed by Penicillin/Beta-lactamase (11.5%), Cephalosporins (11.1%), Ciprofloxacin (10.2%), and the lowest resistance was seen against Colistin (1.2%). In a 5-year surveillance study, Tigecycline, Imipenem, and Cephalosporins were found to be the most resistant antibiotics for *Pseudomonas*. In the same study, Colistin was reported to be the most sensitive antibiotic for *Pseudomonas* [32].

In our cultures with enterococcal growth, resistance to at least one antibiotic was observed at a rate of

26.75%. In our study, Ciprofloxacin was found to be the antibiotic with the highest resistance for *Enterococci* (31.9%), followed by Penicillin (22.1%), Trimethoprim/Sulfamethoxazole (21.1%), Penicillin/Beta-lactamase (11.9%), and the lowest resistance was for Gentamicin (0.54%), Tigecycline (0.54%), Linezolid (1.08%) and Vancomycin (1.62%). In another study, *Enterococci* were also reported as the most common resistance to Tetracycline, while Vancomycin and Teicoplanin were reported as the most sensitive antibiotics in the same study [33].

In our cultures with *Proteus*, resistance to at least one antibiotic was observed at a rate of 47.95%. Colistin was found as the antibiotic with the highest resistance (14.9%) for *Proteus*, in our study, followed by Penicillin (11.7%), Tigecycline (11.7%), Cephalosporins (9.5%), and the lowest resistance was found against Meropenem (1.2%). In a resistance study conducted for *Proteus* species, the highest resistance was found against Colistin, Ampicillin, Trimethoprim/Sulfamethoxazole, and the lowest resistance was reported to be against Meropenem in the same study [34].

Candida was grown in 2.8% (n = 249) of our cultures. When we look at the subclasses of *Candida* proliferation, *Candida albicans* was of 65.4% (n = 163), *Candida crusei* 29.8% (n = 74) and *Candida tropicalis* 4.8% (n = 12). In a study conducted in Turkey, it was reported that *Candida* growth was 26.8% [10]. In a study in which cultures were examined in another urinary tract infection group, *Candida albicans* growth (19.7%) was reported more obviously [35]. Resistance to any antifungal for *Candida* was 5.6%(n = 13). 77% (n = 10) of those that showed resistance was *Candida albicans* subtype. Fungal growth was far below reported in the literature. This may be related to measures against *Candida* reproduction and spread.

Conclusions

Developing health system leads to longer life expectancy, more comorbidities, longer intensive care treatment, more frequent interventional procedures and more complications. In terms of being a resource for empirical treatments, early initiation of empirical treatments to control the urinary tract infection disrupting the patient's hemodynamics and increasing mortality and morbidity, which is one of the most feared courses in intensive care and accurate determination of the hospital's flora and the right choice of antibiotics will save lives.

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