Risk Factors Affecting Patterns of Antibiotic Resistance and Treatment Efficacy in Extreme Drug Resistance in Intensive Care Unit-Acquired *Klebsiella Pneumoniae* Infections: A 5-Year Analysis

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Background: We investigated the factors affecting antibiotic resistance in the intensive care unit (ICU)-related hospital-acquired infections caused by *Klebsiella pneumoniae* (KP-HAI) and the effects of antibiotics used for high-level antibiotic resistance on patient survival.

Material/Methods: This retrospective study was performed at the adult ICU of Bezmialem Vakif University Hospital. Patients who were followed up between 01 January 2012 and 31 May 2017 were evaluated. Each KP strain was categorized according to resistance patterns and analyzed. The efficiency of antibiotic therapy for highly-resistant KP-HAI was determined by patients' lifespans.

Results: We evaluated 208 patients. With the prior use of carbapenem, antibiotics against resistant Gram-positives, and tigecycline, it was observed that the resistance rate of the infectious agents had a significant increase. As the resistance category increases, a significant decrease was seen in the survival time. We observed that if the treatment combination included trimethoprim-sulfamethoxazole, the survival time became significantly longer, and tigecycline-carbapenem-colistin and tigecycline-carbapenem combination patients showed significantly shorter survival times.

Conclusions: When the resistance increases, delays will occur in starting suitable and effective antibiotic treatment, with increased sepsis frequency and higher mortality rates. Trimethoprim-sulfamethoxazole can be an efficient alternative to extend survival time in trimethoprim-sulfamethoxazole-susceptible KP infections that have extensive drug resistance.

MeSH Keywords: Cross Infection • Intensive Care Units • *Klebsiella Pneumoniae* • Trimethoprim-Sulfamethoxazole Combination

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Background

Hospital-acquired infections (HAIs) are the major cause of morbidity and mortality in intensive care units (ICUs) [1]. Invasive procedures such as endotracheal intubation and central venous catheterization, comorbidities such as diabetes and chronic pulmonary disease, acute conditions such as surgery and trauma, and treatment-associated factors such as antibiotic use and blood transfusion have been described to be among the main predisposing factors that cause increased infection rates in ICUs [2]. Enterobacteriaceae is one of the most important causes of HAIs. *Klebsiella pneumoniae* (KP) is the most common microorganism causing HAI among Enterobacteriaceae, which can lead to outbreaks [3]. Increasing antimicrobial drug resistance of KP in the last few decades has been a problem worldwide [4]. Due to increased resistance in the carbapenems group, combination therapy with several susceptible antibiotics has become compulsory [5]. One of these is colistin, which was discontinued for systemic use due to its adverse effects, but has returned to use as there are no other alternatives [6]. Today, because of excessive usage, high resistance rates to this antibiotic have been reported from some countries [5–8], including Turkey [4,9,10]. On the other hand, there is also an increase in the resistance to another effective antibiotic, tigecycline [11,12]. Treatment options dramatically declined because of increased antibiotic resistance to colistin and tigecycline, as well as the absence of alternative antibiotics such as Fosfomycin and ceftazidime-avibactam in Turkey. Furthermore, there are very few publications in the literature about antimicrobial treatment against KP infections with a high-resistance profile.

In this study, we investigated the factors affecting antibiotic resistance in ICU-related KP-HAI and the effects of antibiotics used in the presence of high-level antibiotic resistance on patient survival.

Material and Methods

Study design

This retrospective study was performed at the adult ICU of Bezmialem Vakif University Medical Faculty Hospital. Patients who were followed up between 01 January 2012 and 31 May 2017 were evaluated. KP-HAIs are categorized according to their antibiotic resistance profiles [13]. We searched the literature and determined possible risk factors [12,14–19]. To determine the risk factors that affect antibiotic resistance, the attributes of the cases, grouped based on their resistance category, were compared with each other. In order to display the efficiency of the antibiotics given to the patients with limited treatment options, the efficiency of the antibiotic(s) used in

the cases with high-level antibiotic resistance strain infection were compared based on the patients' lifespans.

Study setting

Bezmialem Vakif University Medical Faculty Hospital has 550 beds and serves approximately 70 000 hospitalized patients annually in İstanbul, Turkey. We have 54 adult ICU beds in 5 different ICUs. The Anesthesia and Reanimation ICU I is a mixed medical-surgical ICU with 12 beds. The Respiratory ICU is a mixed medical-surgical ICU with 12 beds (it was converted to a mixed medical-surgical ICU as the Anesthesia and Reanimation ICU II in March 2013). The Neurology ICU is a mixed medical-surgical ICU with 9 beds. The Cardiovascular Surgery ICU is a surgical ICU with 9 beds. The Coronary ICU is a medical ICU with 12 beds. The annual adult patient count is approximately 3000 who are admitted to ICU.

Inclusion and exclusion criteria

ICU-related KP-HAIs ≥18 years old cases whose duration of hospitalization was > 48 hours were included in the study. Only the first infection was taken into consideration of the patients who developed KP-HAI several times (both device-related and the other HAIs). We excluded patients who did not meet the inclusion criteria, those whose information was not available, and those who could not be followed up because they were referred to another ICU.

Definitions

Glasgow coma scale (GCS) and APACHE II: GCS scores at the time of the ICU admission and APACHE II scores at the time of infection diagnosis were taken into consideration.

Transfusion: Blood/blood product transfusions were noted only if they were administered within 5 days before the diagnosis of infection.

Suitable/Unsuitable antibiotic treatment: Any of the antibiotics given to the patient in the first 48 hours were presumed as "suitable antibiotic treatment" if it was sensitive and "unsuitable antibiotic treatment" if it was resistant, according to the antibiogram of the microorganism.

Sepsis and septic shock: The diagnosis of sepsis and septic shock were made as per the manual [20].

Antimicrobial resistance category: Each KP strain was categorized according to the antibiogram results as "non-multi drug-resistant" (non-MDR) if it was non-susceptible to less than 3 antibiotic groups, "multi-drug-resistant" (MDR) if it was non-susceptible to 3 or more antibiotic groups but susceptible to

at least 2 groups, "extensively drug-resistant" (XDR) if it was only susceptible to 1 or 2 antibiotic groups, and "pan-drug-resistant" (PDR) if it was non-susceptible to all antibiotics in all antimicrobial categories [13]. Since several required antibiotics have not been tested in the antibiotic susceptibility tests, the terms XDR and PDR should be interpreted as 'possibly XDR' and 'possibly PDR'. Because there were only a small number of PDR strains, the categories of XDR and PDR were combined and analyzed together (XDR-PDR).

Antibiotics and doses used for treatment: The dose of tigecycline was 2×50 mg intravenous (IV) after a 100 mg loading dose, colistin was 3×150 mg IV maintenance after a 300 mg IV loading dose, meropenem was 3×1 gr IV, imipenem was 4×500 mg IV, piperacillin-tazobactam 3×4.5 gr IV, ceftriaxone 2×1 gr IV, gentamicin 1×1–3 mg/kg IV, ciprofloxacin 2×400 mg IV and levofloxacin 1×750 mg IV, trimethoprim-sulfamethoxazole' was (dosing is based on the trimethoprim component) 15–20 mg/kg/day IV with dividing the dose into 3 or 4 parts. Antibiotic doses (except tigecycline and ceftriaxone) of the patients with high creatinine levels were adjusted in accordance with their creatinine clearances.

Data collection

HAIs were diagnosed by the infection control team according to the criteria of "Centers for Disease Control and Prevention (CDC)" during daily active surveillance [21,22]. The infection control team is composed of an infection control nurse and an infectious disease and clinical microbiology specialist (also responsible for infection control). In Turkey, all data on HAIs in ICU patients are entered into an online database of the Ministry of Health and we can access this data any time. Demographic data, cause of hospitalization, comorbidities, probable risk factors for HAIs (e.g., surgical drain, dialysis/hemodiafiltration, urinary catheter, colostomy, mechanic ventilation, central venous catheter, tracheotomy, total parenteral nutrition, transfusion), invasive procedures, antibiotics used before and after infection (antibiotic use among ICU follow-up was considered), antibiotic initiation time and antimicrobial sensitivity data were obtained from the database of the Ministry of Health. Sepsis, development of septic shock (during ICU follow up), GCS, and APACHE II scores data were obtained from patient files.

Microbiology

Hemoculture bottles (Becton-Dickinson, USA) were placed in a BACTEC FX instrument (Becton-Dickinson, USA) which was set up for 6 days of the incubation program. Signal-positive hemoculture samples and all other samples sent for culture (urine, endotracheal aspirate/sputum, abscess and other biologic samples such as cerebrospinal fluid, tissue, and ascites) were examined with Gram's strain and eosin methylene blue agar (Salubris, Turkey), respectively, and then passaged to chocolate agar (Salubris, Turkey). After the media were incubated for 18–24 hours at 37°C, the reproduced bacteria were identified by conventional and automated systems (VITEK® 2 Compact (BioMérieux, France). Identification of the strains with VITEK 2 were also confirmed with VITEK® MALDI-TOF (BioMérieux, France). Antimicrobial sensitivity results were assessed with an automated system (VITEK-2, BioMérieux, France) as per the criteria of "Clinical and Laboratory Standards Institute" [23,24]. Minimum inhibitory concentration values found for colistin were also estimated in accordance with breakpoint test values of the European Committee on Antimicrobial Susceptibility Testing.

Statistics

Descriptive statistics are presented as mean, standard deviation, median and interquartile range, and percentage. Normality was assessed by use of the Kolmogorov-Smirnov test. Kruskal-Wallis analysis was used to compare the averages of quantitative attributes in 3 groups and post hoc Dunn tests for multiple comparisons. Categorical variables were compared by chi-square test. When mortality was considered as a dependent variable, we used the function of Cox proportional hazards to assess the effects of treatments with the multivariate statistical method. Survival rates were calculated with Kaplan-Meier method and the analyses were made using IBM SPSS 20.0. The significance level (p-value) was accepted as 0.05 in all tests.

Ethics statement

The Ethics Committee of Bezmialem Vakif University approved the study (Ethics Committee Approval: 22/06/2017-11378).

Results

Study population

We included 211 patients admitted to the ICU from 01 January 2012 to 31 May 2017. Three patients were excluded from the study because they were discharged to be followed up in different hospitals.

We included 208 KP-HAI patients (63% male and 27% female). The average values were: median age was 67.5 years (interquartile range, 59.3-78.0 years), median GCS was 9.0 (interquartile range, 6.0–12.0), median APACHE II score was 17.5 (interquartile range, 13.3–22.0), median duration of ICU hospitalization before the infection was 15.0 days (interquartile range, 9.0–31.0 days), and median duration of ICU hospitalization after infection was 16.5 days (interquartile range, 6.0–34.8 days). Throughout their follow-up in hospital, the crude mortality rate was 70% (n=146).

Table 1. Antibiotic resistance rates of *Klebsiella pneumoniae*.

Antibiotic	n (Sensitive)	n (Resistance)	Total	% (Resistance)
Tigecycline	140	59	199	29.6
Colistin	114	85	199	42.7
Trimethoprim-sulfamethoxazole	119	88	207	42.5
Gentamicin	93	115	208	55.3
Amikacin	93	114	207	55.1
Meropenem	91	117	208	56.3
Imipenem	86	122	208	58.7
Levofloxacin	56	152	208	73.1
Ciprofloxacin	57	151	208	72.6
Piperacillin-tazobactam	35	173	208	83.2
Cefoperazone-sulbactam	33	168	201	83.6
Cefepime	18	189	207	91.3
Ceftazidime	19	189	208	90.9

Distribution of infections and antimicrobial resistance

The distribution of infections was: 64.9% pneumonia, 21.2% central line-associated bloodstream infection (CLABSI), 4.8% bloodstream infection (BSI) proved by the laboratory, 3.8% urinary system infection (USI), and 6.3% other infections (skin soft tissue and surgical site infections). The secondary BSI rate was 16.8%. Twenty-five percent of infections (52) were determined to be polymicrobial.

The minimum resistance rate was against tigecycline (29.6%), followed by colistin (42.7%) and trimethoprim-sulfamethoxazole (42.5%). Resistance against meropenem was 56.3%. Other resistance rates are given in Table 1.

Patient rates according to non-MDR, MDR, XDR, PDR, and XDR-PDR resistance categories were determined respectively as follows: 20.2%, 40.4%, 36.5%, 2.9%, and 39.4%.

Effect of patient characteristics (risk factors) on resistance categories

When non-MDR, MDR, and XDR-PDR groups were compared with each other by univariate analysis; the APACHE II scores of MDR and XDR-PDR groups were higher (p=0.013), such as the frequency of septic shock (p=0.018).

Patients who were hospitalized for pneumonia and those who had been using quinolone (ciprofloxacin and levofloxacin) or 3rd generation cephalosporins (ceftriaxone, cefotaxime, and ceftazidime) before infection had higher MDR and XDR-PDR profiles (p=0.011, p=0.003, p=0.028, and, respectively).

XDR-PDR category patients had a shorter duration of hospitalization in the ICU after the diagnosis of infection (p=0.017).

MDR is found more frequently in patients who developed CLABSI (p=0.035), had chronic obstructive pulmonary disease (p=0.046), and who were hospitalized in the ICU for respiratory insufficiency (p=0.040). With the increasing antibiotic resistance of microorganisms, a significant increase was observed in the risk of developing sepsis originating from KP-HAIs (p=0.009). With the prior use of carbapenems (meropenem and imipenem), antibiotics against resistant Gram-positives (vancomycin, teicoplanin, linezolid, and daptomycin) and tigecycline, it was observed that resistance rates of an infectious agent significant increased (p=0.001, p=0.001, and p≤0.001, respectively) (Table 2). Throughout the duration of hospitalization after infection, the average survival time of non-MDR, MDR, and XDR-PDR groups were 69.5±12.3, 48.4±6.4, and 31.5±5.7 days, respectively. As the resistance category worsens, a significant decrease is seen in the survival time (see Figure 1).

Resistance and treatment attributes of XDR-PDR strains

The lowest resistance rates in this category were for tigecycline (36.6%), followed by trimethoprim-sulfamethoxazole (47.6%), colistin (76.8%), gentamicin (84.1%), and amikacin (90.2%). Carbapenems and all other antimicrobials (quinolones, third-generation cephalosporins, cefepime, piperacillin-tazobactam, and cefoperazone-sulbactam) were found to be non-susceptible.

As we analyzed antibiotic treatments used by patients in the XDR-PDR resistance category, we saw that the combination of double antibiotics was the most-prescribed treatment (55.0%), followed by triple combinations (23.2%) and single-antibiotic

Table 2. Comparison of antibiotic resistance profiles and patient attributes.

Variable	non-MDR (n: 42) n (%)	MDR (n: 84) n (%)	XDR-PDR (n: 82) n (%)	p
Gender (male)	27 (64.3)	52 (61.9)	52 (63.4)	0.961
Age (median [IQR]) (years)	66.5 [59.8–78.0]	70.0 [58.5–78.8]	67.0 [59.0–79.0]	0.800
GCS (first hospitalization day in ICU) (median [IQR])	10.0 [7.8–12]	8.0 [5.0–11.0]	9.0 [6.0–12.0]	0.083
APACHE II score (day of culture was taken) (median [IQR])	15.0 [12.0–19.3]*	17.0 [13.0–22.8]	19.0 [15.0–19.0]	**0.013**
Hospital stay prior to HAI (median [IQR]) (days)	19.0 [8.5–28.5]	16.0 [10.0–33.0]	24.0 [10.8–41.3]	0.156
ICU stay prior to HAI (median [IQR]) (days)	13.5 [7.0–25.3]	14.0 [9.3–27.8]	18.0 [8.8–34.5]	0.292
Hospital stay after infection (median [IQR]) (days)	24.5 [13.3–47.0]	20.5 [10.0–42.8]	14.0 [3.8–29.0]*	**0.003**
ICU stay after infection (median [IQR]) (days)	20.5 [9.8–35.8]	18.5 [7.3–42.8]	14.0 [3.8–27.0]*	**0.017**
Initiation of effective therapy (within 48 h)	30 (71.4)*	46 (54.8)*	37 (45.1)*	**0.021**
General category				
Elective surgery	8 (19.0)	16 (19.0)	10 (12.2)	0.426
Emergent surgery	6 (14.3)	14 (16.7)	20 (24.4)	0.298
Medical	28 (66.7)	50 (59.5)	51 (62.2)	0.738
Trauma	6 (14.3)	9 (10.7)	5 (6.1)	0.311
Underlying disease				
Diabetes	13 (31.0)	24 (28.6)	22 (26.8)	0.889
Hypertension	24 (57.1)	39 (46.4)	47 (57.3)	0.308
Chronic obstructive pulmonary disease	6 (14.3)	24 (28.6)*	12 (14.6)	**0.046**
Cancer	10 (23.8)	24 (28.6)	23 (50.0)	0.841
Renal insufficiency	4 (9.5)	20 (23.8)	22 (26.8)	0.080
Cardiac insufficiency	9 (21.4)	18 (21.4)	15 (18.3)	0.859
Coronary artery disease	9 (21.4)	12 (14.3)	21 (25.6)	0.187
Neurological disorder	13 (31.0)	32 (38.1)	25 (30.5)	0.536
ICU hospitalization reason				
Renal insufficiency	4 (9.5)	12 (14.3)	8 (9.8)	0.594
Cancer	6 (14.3)	14 (16.7)	7 (8.5)	0.285
General body trauma	6 (14.3)	8 (9.5)	3 (3.7)	0.104
Respiratory insufficiency	19 (45.2)	53 (63.1)*	37 (45.1)	**0.040**
Acute abdomen	0 (0)	5 (6.0)	5 (6.1)	n/applicable
Neurological disorder	13 (31.0)	31 (36.9)	34 (41.5)	0.514
Cardiac insufficiency	5 (11.9)	9 (10.7)	8 (9.8)	0.933
Pneumonia	3 (7.1)*	26 (31.0)	19 (23.2)	0.011
Sepsis	0 (0)	5 (6.0)	7 (8.5)	n/applicable
Other infections	2 (4.8)	6 (7.1)	10 (12.2)	0.309
Hospital infections				
Pneumonia	30 (71.4)	51 (60.7)	54 (65.9)	0.481
CVC-Blood Stream Infection	5 (11.9)	25 (29.8)*	14 (17.1)	**0.035**

Table 2 continued. Comparison of antibiotic resistance profiles and patient attributes.

Variable	non-MDR (n: 42)		MDR (n: 84)		XDR-PDR (n: 82)		p
	n (%)		n (%)		n (%)		
Laboratory-Proven Bacteremia	2	(4.8)	1	(1.2)	7	(8.5)	n/applicable
Secondary Bacteremia	8	(19.0)	11	(13.1)	16	(19.5)	0.495
Urinary System Infection	3	(7.1)	2	(2.4)	3	(3.7)	n/applicable
Other Infections	2	(4.8)	2	(2.4)	3	(3.7)	n/applicable
Polymicrobial	12	(28.6)	23	(27.4)	17	(20.7)	0.513
Sepsis	10	(23.8)*	34	(40.5)*	43	(52.4)*	**0.009**
Septic Shock	4	(9.5)*	23	(27.4)	27	(32.9)	**0.018**
Risk factors							
Drain	10	(23.8)	37	(44.0)	33	(40.2)	0.081
Dialysis/Hemodiafiltration	5	(11.9)	15	(17.9)	14	(17.1)	0.678
Urinary catheter	41	(97.6)	83	(98.8)	79	(96.3)	n/applicable
Colostomy	2	(4.8)	4	(4.8)	5	(6.1)	n/applicable
Mechanic ventilation	38	(90.5)	83	(98.8)	79	(96.3)	n/applicable
CVC	38	(90.5)	79	(94.0)	77	(93.9)	0.721
Tracheotomy	9	(21.4)	32	(38.1)	33	(40.2)	0.096
TPN	15	(35.7)	32	(38.1)	26	(31.7)	0.687
Transfusion	23	(54.8)	53	(63.1)	45	(54.9)	0.496
Antibiotic use before infection							
Quinolone	3	(7.1)*	24	(28.6)	29	(35.4)	**0.003**
Ampicillin-sulbactam	14	(33.3)	22	(26.2)	14	(17.1)	0.112
Cefazolin	4	(9.5)	18	(21.4)	12	(14.6)	0.203
3rd generation cephalosporin	7	(16.7)*	33	(39.3)	31	(37.8)	**0.028**
Carbapenem	13	(31.0)*	38	(45.2)*	54	(65.9)*	**0.001**
Piperacillin-tazobactam	6	(14.3)	23	(27.4)	27	(32.9)	0.085
Cefoperazone-sulbactam	9	(21.4)	28	(33.3)	31	(37.8)	0.182
Effective antibiotics against resistant Gram-positives	11	(26.2)*	31	(36.9)*	48	(58.5)*	**0.001**
Colistin (Intravenous)	6	(14.3)	17	(20.2)	24	(29.3)	0.134
Colistin (Inhaler)	8	(19.0)	17	(20.2)	28	(34.1)	0.068
Tigecycline	8	(19.0)*	24	(28.6)*	44	(53.7)*	**<0.001**
Metronidazole	1	(2.4)	10	(11.9)	8	(9.8)	0.210
Antifungal	3	(7.1)	11	(13.1)	15	(18.3)	0.227
Macrolide	3	(7.1)	14	(16.7)	15	(18.3)	0.243
Trimethoprim-sulfamethoxazole	2	(4.8)	8	(9.5)	14	(17.1)	0.096
Aminoglycoside	1	(2.4)	7	(8.3)	5	(6.1)	0.428

* p≤0.05. Non-MDR – non-multidrug-resistant; MDR – multidrug-resistant; XDR – extensively drug-resistant; PDR – pandrug-resistant; IQR – interquartile range, GCS – Glasgow Coma Scale; APACHE II score – Acute Physiology and chronic Health Enquiry II score; ICU – Intensive Care Unit; HAI – hospital-acquired infection; CVC – central venous catheter; TPN – total parenteral nutrition. Effective antibiotics against resistant Gram-positives; vancomycin, teicoplanin, linezolid, and daptomycin.
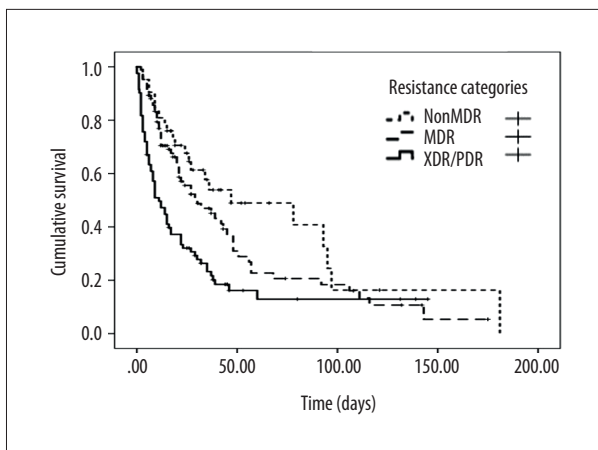
Figure 1. The relation between antibiotic resistance categories and cumulative survival.

treatment (18.3%). Antibiotic applications in combination treatments and their frequency rates were tigecycline (71%), carbapenems (46%), colistin (33%), and trimethoprim-sulfa-methoxazole (26%).

In regard to the double combinations of tigecycline, the observations showed that it was combined with trimethoprim-sulfa-methoxazole, carbapenems, and colistin, and their rates were tigecycline plus trimethoprim-sulfamethoxazole: 23.2%, tigecycline plus carbapenems: 20.7%, and tigecycline plus colistin: 9.8%. Secondary to tigecycline, carbapenems was the most frequent component used with colistin (carbapenems plus colistin: 7.3%). The combination of Tigecycline plus carbapenems plus colistin was the most-used triple treatment (11.0%), and tigecycline, colistin, and trimethoprim-sulfamethoxazole were not used alone in treatment.

When we looked at the significant effect of XDR-PDR on Cox regression analysis of the relationship between patient survival and antibiotic(s) initiated after infection, we found that patients receiving the combination of tigecycline plus trimethoprim-sulfamethoxazole had longer survival times (p=0.018, *HR*: 0.709), whereas patients receiving combinations of tigecycline plus carbapenems plus colistin and tigecycline plus carbapenems had significantly shorter survival times (p=0.027, *HR*: 1.401and p=0.49, *HR*: 1.259, respectively) (Table 3).

Regarding the significant effect of XDR-PDR on Cox regression analysis for one-to-one effects of antibiotics on patient survival, we observed that if the treatment combination included trimethoprim-sulfamethoxazole, the survival time became significantly longer (p=0.005, *HR*: 0.460) (Table 4).

Table 3. Effect of Antibiotic combinations on patient survival (Cox regression with enter method).

| | | HR | 95.0% CI for HR | |
			Lower	Upper
		p	Lower	Upper
XDR-PDR	**<0.001**	1.485	1.244	1.773
TGC-CP-CS	**0.027**	1.401	1.039	1.890
TGC-CP	**0.049**	1.259	1.001	1.583
TGC-CS	0.326	1.149	0.871	1.516
TGC-TMPS	**0.018**	0.709	0.532	0.943
CP-CS	0.720	0.938	0.659	1.334

Model Chi-square=33.83, p≤0.001. Variables included to Cox analyze: non-MDR – non-multiple drug resistance; MDR– multiple drug resistance; XDR-PDR – extensive drug resistance/pan-drug resistance; TGC-CP-CS – Tigecycline, Carbapenem, and Colistin; TGC-CP – Tigecycline and Carbapenem; TGC-CS – Tigecycline and Colistin; TGC-TMPS – Tigecycline and Trimethoprim-sulfamethoxazole); CP-CS – Carbapenem and Colistin).

Table 4. One-to-one effect of antibiotic groups on patient survival (Cox regression with enter method).

| | | HR | 95.0% CI for HR | |
			Lower	Upper
		p	Lower	Upper
XDR-PDR	**<0.001**	1.481	1.238	1.772
Combinations with TMPS	**0.005**	0.460	0.267	0.794
Combinations with TGC	0.196	1.267	0.885	1.814
Combinations with CP/ single	0.370	1.171	0.829	1.655
Combinations with CS	0.432	1.148	0.814	1.618

Model Chi-square=29.78, p≤0.001. XDR-PDR – extensive drug resistance/pan-drug resistance; TMPS – Trimethoprim-sulfamethoxazole; TGC – Tigecycline; CP – Meropenem); CS – Colistin.

Discussion

In this study, high resistance rates were found even in potent antimicrobials like carbapenems, colistin, and tigecycline. As the resistance against the antibiotics increased, a shortening in average survival time was observed.

Some antibiotics, which were used before infection (quinolones, third-generation cephalosporins, carbapenems, antibiotics

effective against resistant Gram-positives and tigecycline), were observed to increase the resistance.

Among patients in the XDR-PDR category, it was observed that patients using the combination of tigecycline plus trimethoprim-sulfamethoxazole had significantly longer survival times and the main effect of the combination was originating from trimethoprim-sulfamethoxazole, but survival time of patients using tigecycline plus carbapenems plus colistin and tigecycline plus carbapenems was significantly shorter.

In various studies, a linear relationship was reported between bacterial resistance and delay in initiating effective antibiotic treatment [25–27]. Development of sepsis and accordingly high mortality rates are expected results of the delay in starting effective treatment. In addition, many studies showed that antibiotic resistance also had an independent relationship with high mortality rates [6,12,18]. In our study, compatible with the literature, a linear significant relation was found with the delay in initiating effective antibiotic, sepsis, septic shock, and mortality as the resistance category was worsened.

HAIs lengthen the duration of hospitalization. Also, when we investigated whether there was a variance in ICU hospitalization duration according to different resistance categories, we found that the XDR-PDR category patients had significantly shorter ICU hospitalization duration than patients with other resistance categories. This situation is explained by the shortness of the average survival time in XDR-PDR resistance category patients.

Many studies found that prior prolonged, incomplete, or excessive usage of colistin had an influence on the selection of colistin-resistant strains [14,17,18,28]. In our study, it was impossible to make one-to-one comparisons because colistin-resistant and -sensitive strains were not compared, but when a comparison was made between resistance categories, such a relationship was observed.

We found that MDR and XDR-PDR frequency was significantly higher than the non-MDR frequency in the patients who previously used quinolone and third-generation cephalosporins. These findings are compatible with the literature [19,29]. Several studies show a relationship between prior carbapenem usage and increased resistance [16,30]. In our study, a significant linear relationship between carbapenem use and resistance categories was determined, not only between non-MDR and other resistance categories, but there was also a significant difference between MDR and XDR-PDR categories as well. In previous studies, the relationship was not revealed between previous tigecycline use and development of resistance, but in our study, a linear significant relation was determined between tigecycline use and resistance categories as

well as carbapenem use. In our ICU, *Acinetobacter baumannii* is the most common agent that causes HAIs, and its infections lead to excessive tigecycline use. It was concluded that this excessive tigecycline use may have caused such a result.

In some studies, glycopeptide use was found to be related to the development of resistance by univariate analysis, but this relationship was not significant in multivariate analysis [16,19]. In our study, a significant linear relationship between using effective antibiotics against resistant Gram-positives and resistance categories was determined by univariate analysis, but it was impossible to determine if there was an independent effect because multivariate analysis was not done.

There are only a small number of studies that widely investigated the treatment of resistant infections. Recently, Muri et al. found that trimethoprim-sulfamethoxazole was effective in monotherapy of non-ICU patients with carbapenem-resistant but trimethoprim-sulfamethoxazole-sensitive infections [31]. Vidaillac et al. reported the synergistic effect of colistin plus trimethoprim-trimethoprim-sulfamethoxazole [32].

In various studies, synergistic effects of colistin, tigecycline, and rifampicin combinations were mentioned in the treatment of KPC-producing KP strains [33,34]. Meredith et al. revealed that the doripenem and colistin combination has a synergistic effect [35]. In our study, the most preferred antibiotherapy against XDR strains is tigecycline, including combinations, perhaps because the origin of this preference was that the lowest rate of resistance in XDR strains was found to be tigecycline. Instead of a single regimen, tigecycline was preferred in combination treatment because it is bacteriostatic, and its use is not recommended in nosocomial pneumonia because it does not diffuse enough into the urine to reach its effective dose. Nevertheless, many studies showed the positive effect of colistin and meropenem in combination, and tigecycline was strongly preferred with this combination. In Turkey, which is an endemic country for tuberculosis, rifampicin, a major anti-tuberculous drug, was not preferred in combinations. Trimethoprim-sulfamethoxazole was used in combination treatments because it has the second-highest sensitivity (after tigecycline) against XDR strains. Due to its nephrotoxic effects, trimethoprim-sulfamethoxazole was not preferred for use together with other nephrotoxic agents like colistin. Trimethoprim-sulfamethoxazole was combined with tigecycline in all patients using it. As seen in Table 3, combination treatments with tigecycline and trimethoprim-sulfamethoxazole were found to be related with long-term survival, in contrast to tigecycline plus carbapenems and tigecycline plus carbapenems plus colistin combinations, which are associated with shorter patient survival. All XDR strains were found to be resistant to carbapenems (100%). In this regard, using carbapenem with an agent like tigecycline, which is bacteriostatic, has

a negative effect on patient survival because it cannot reach a sufficient concentration in the blood, it cannot diffuse enough into the urine to reach its effective dose, and its use is not recommended in nosocomial pneumonia. When we look at the characteristics of the patients that were initiated with the tigecycline plus carbapenems plus colistin combination, this combination can be seen as a salvage treatment because of the higher preference rate in PDR KP infections (preferred in 4/6 of PDR cases) and in difficult cases, which have higher APACHE II scores (averagely 19.4±4.9). Accordingly, the survival time of these cases was shorter. Although we did not use tigecycline, trimethoprim-sulfamethoxazole and colistin alone, a comparison of survival times according to one-to-one antimicrobials shows that treatments including trimethoprim-sulfamethoxazole are associated with longer survival times (Table 4). Our observations did not show any extending effect of other antimicrobials on survival. However, trimethoprim-sulfamethoxazole was used only in combination with tigecycline. This situation indicates 2 possibilities: either trimethoprim-sulfamethoxazole causes this effect on its own, as several studies suggest [31], or the combination of trimethoprim-sulfamethoxazole with tigecycline creates a synergistic effect and extended lifespan.

This study has several certain limitations. First, the retrospective design of the study should be acknowledged as a natural limitation, but we think that our data are reliable because identification of HAIs was done prospectively with daily active surveillance and the data were entered into the database rapidly. In addition, the results of antibiotic susceptibility testing were for the most part complete. In a few HAIs, the susceptibility of some antibiotics could not be studied because the proper kit was not available in the microbiology laboratory, but this was rare and probably did not affect the results. Antimicrobial resistance can differ among regions and hospitals. Our study was conducted in a single center, so our antimicrobial susceptibility results may not reflect the situation at the regional level.

However, as our hospital is one of the biggest reference hospitals in this region, and the ICU bed count of our hospital is greater than the mean ICU counts of all hospitals in Turkey, we believe that our results are valuable. Another limitation of our study is that we could not study specific antibiotic resistance genes and enzymes with molecular or genetic tests and we could not investigate clonal dissemination of isolates because of the high cost.

Our literature search revealed very few studies examining the efficacy of trimethoprim-sulfamethoxazole in XDR-KP infections. In addition, among the investigations about combination treatments for resistant KP infections, studies on combinations with trimethoprim-sulfamethoxazole are limited. Multicenter and prospective investigations are needed to better assess the efficacy of trimethoprim-sulfamethoxazole on single or combination treatments in XDR-KP infections.

Conclusions

Using broad-spectrum antibiotics is generating even more resistant strains, especially in critical-care units like ICUs. As long as the resistance increases, delays will occur in starting suitable and efficient antibiotic treatment, and sepsis frequency and mortality rates will become higher. Our results show that trimethoprim-sulfamethoxazole can be used in patients with trimethoprim-sulfamethoxazole-susceptible XDR-KP infection for whom there is no alternative therapy that is more effective than these drugs. Rational antibiotic use and strict adjustment to the infection control measures will become more and more important until we can find new antibiotics.

Conflict of interest

None.

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