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Reversal of carbapenemase-producing *Klebsiella pneumoniae* epidemiology from bla_{KPC}- to bla_{VIM}-harbouring isolates in a Greek ICU after introduction of ceftazidime/avibactam

Matthaios Papadimitriou-Olivgeris[1]†, Christina Bartzavali[2], Anastasia Lambropoulou[2], Anastasia Solomou[3], Ekaterini Tsiata[4], Evangelos D. Anastassiou[2], Fotini Fligou[3], Markos Marangos[1], Iris Spiliopoulou[2] and Myrto Christofidou[2]*

[1]Division of Infectious Diseases, Department of Internal Medicine, School of Medicine, University of Patras, 26504, Rion-Patras, Greece; [2]Department of Microbiology, School of Medicine, University of Patras, 26504, Rion-Patras, Greece; [3]Anesthesiology and Critical Care Medicine, School of Medicine, University of Patras, 26504, Rion-Patras, Greece; [4]Department of Pharmacy, University General Hospital of Patras, 26504, Rion-Patras, Greece

*Corresponding author. Tel: +30 2610 997632; Fax: +30 2610 994922; E-mail: christof@upatras.gr
†Present address: Department of Infectious Diseases, University Hospital of Lausanne, Lausanne, Switzerland.

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Objectives: Our aim was to determine the epidemiology of bloodstream infections (BSIs) by carbapenemase-producing *Klebsiella pneumoniae* (CP-Kp) after the introduction of ceftazidime/avibactam in January 2018 among ICU patients.

Patients and methods: All patients hospitalized at the ICU of the University General Hospital of Patras, Greece with CP-Kp BSI during 2015–18 were included. MICs of meropenem, fosfomycin, tigecycline and ceftazidime/avibactam (only for isolates from 2018) were determined by Etest, whereas for colistin, the broth microdilution method was applied. All isolates were tested by PCR for the presence of bla_{KPC}, bla_{VIM}, bla_{NDM} and bla_{OXA-48} genes.

Results: Among 170 BSIs due to CP-Kp (2015–18), 132 (78%) were caused by isolates carrying bla_{KPC} (4 ceftazidime/avibactam-resistant), 17 bla_{VIM} (10%), 16 bla_{NDM} (9%) and 5 carrying both bla_{KPC} and bla_{VIM} (3%). From 2015 to 2017 (125 BSIs), KPC-producing strains (110; 88%) predominated, followed by NDM-producing strains (15; 12%), whereas no VIM-producing strain was isolated. Among the 45 BSIs in 2018, 22 (49%) were due to isolates carrying bla_{KPC} (4 ceftazidime/avibactam resistant), followed by 17 (38%) carrying bla_{VIM}, 5 (11%) carrying both bla_{KPC} and bla_{VIM}, and 1 isolate carrying bla_{NDM} (2%). MBLs were more frequent in 2018 compared with 2015–17 (51% versus 12%; $P < 0.001$). Multivariate analysis found that prior administration of ceftazidime/avibactam ($P=0.014$; OR 16.7, 95% CI 1.8–158.6) was independently associated with the development of BSI due to ceftazidime/avibactam-resistant isolates.

Conclusions: Widespread ceftazidime/avibactam use may lead to a change in the palette of carbapenemases by replacing KPC with MBL-producing isolates.

Introduction

Ceftazidime/avibactam is a novel β-lactam/β-lactamase inhibitor combination that inactivates class A (KPC) and class C β-lactamases, whereas it has no activity against MBLs, such as VIM or NDM.[1] Owing to its superiority over colistin in the guided treatment of carbapenem-resistant Enterobacteriaceae[2] and the low level of resistance among bla_{KPC}-carrying isolates,[3] ceftazidime/avibactam became an important addition to the depleted armamentarium in the Greek setting, where KPC is the predominant carbapenemase among *Klebsiella pneumoniae*.[3,4]

The aims of the present study were to evaluate the changes in carbapenemase epidemiology in an ICU after the introduction of ceftazidime/avibactam and to identify risk factors for bloodstream infection (BSI) due to carbapenemase-producing ceftazidime/avibactam-resistant *K. pneumoniae* (CZA-R).

Materials and methods

This is a retrospective study carried out at the general ICU (13 beds) of the University General Hospital of Patras, Greece, during a 4 year period (2015–18). Patients with at least one positive blood culture for

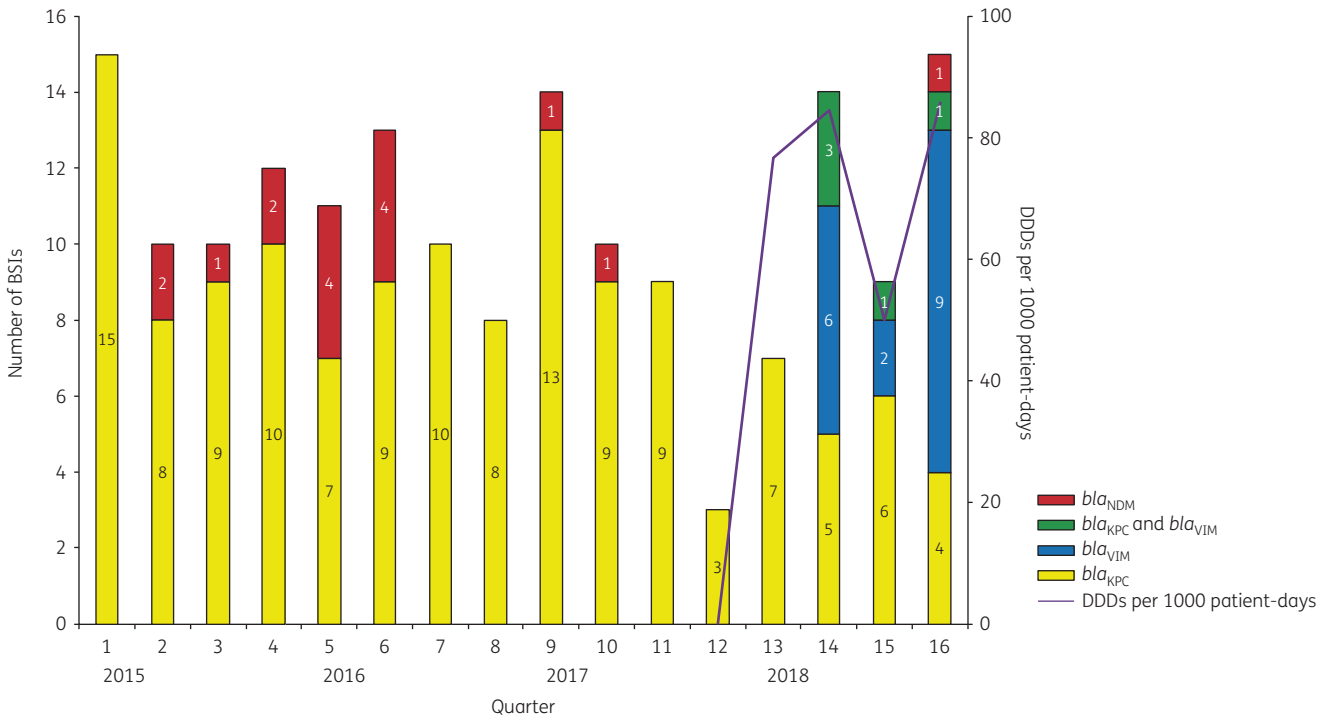

Figure 1. Quarterly distribution of carbapenemase genes among CP-Kp BSIs and ceftazidime/avibactam consumption (DDDs per 1000 patient-days). This figure appears in colour in the online version of *JAC* and in black and white in the print version of *JAC*.

carbapenemase-producing *K. pneumoniae* (CP-Kp) were included in the study. Multiple episodes of bacteraemia from the same patient were included if a duration of at least 2 months occurred between two episodes. Ceftazidime/avibactam was available from January 2018 and its consumption in the ICU (data from Department of Pharmacy) was calculated using the DDD per 1000 patient-days, as described by the WHO Anatomical Therapeutic Chemical (ATC)/DDD. From January to July, ceftazidime/avibactam was used as definite and empirical treatment, but from July onwards, after the first signs of a changing epidemiology, empirical treatment was limited and was mainly used as guided therapy. The study was approved by the Hospital Ethics Committee (No. 858).

K. pneumoniae strains isolated from blood cultures deriving from ICU patients were identified using the Vitek 2 Advanced Expert System (bioMerieux, Marcy-l'Étoile, France). MICs of meropenem, fosfomycin, tigecycline and ceftazidime/avibactam (only for isolates from 2018) were determined by Etest (bioMerieux), whereas for colistin, the broth microdilution method was applied.[5] All isolates were tested by PCR for the presence of bla_{KPC}, bla_{VIM}, bla_{NDM} and bla_{OXA-48} genes.[6]

We compared the rate of MBL-producing isolates before (2015–17) and after the introduction of ceftazidime/avibactam (2018). In order to identify the risk factors for the development of BSI due to CZA-R isolates in 2018, CZA-R BSIs (cases) were compared with BSIs due to ceftazidime/avibactam-susceptible *K. pneumoniae* (CZA-S) isolates (controls). Data (epidemiological data, comorbidities, antimicrobial administration and ICU procedures) were obtained from patients' chart reviews and the ICU computerized database (Criticus™, University of Patras, Greece).

SPSS version 23.0 (SPSS, Chicago, IL, USA) was used for data analysis. Categorical variables were analysed by using the Fisher exact test and continuous variables with Mann–Whitney *U*-test, as appropriate. Backward stepwise multiple logistic regression analysis used all the variables from the univariate analysis with a $P < 0.1$. $P < 0.05$ was considered statistically significant.

Results and discussion

During the study period (2015–18), 170 BSIs due to CP-Kp were observed among 155 patients (15 patients had a second bacteraemia during the same or subsequent hospitalization); 132 (78%) were caused by isolates carrying bla_{KPC} (4 CZA-R), 17 bla_{VIM} (10%), 16 bla_{NDM} (9%) and 5 carrying both bla_{KPC} and bla_{VIM} (3%) (Figure 1). From 2015 to 2017 (125 BSIs), KPC-producing strains (110; 88%) predominated, followed by NDM-producing strains (15; 12%), whereas no VIM-producing strain was isolated. Among the 45 BSIs in 2018, 22 (49%) were due to isolates carrying bla_{KPC} (4 CZA-R), followed by 17 (38%) carrying bla_{VIM}, 5 (11%) carrying both bla_{KPC} and bla_{VIM}, and 1 isolate carrying bla_{NDM} (2%). MBLs were more frequent in 2018 compared with 2015–17 (51% versus 12%; $P < 0.001$). Overall colistin and tigecycline resistance was 46% and 23%, respectively. The overall consumption of ceftazidime/avibactam was 74.2 DDD per 1000 patient-days.

Results of the univariate analysis for development of BSI due to CZA-R isolates are shown in Table 1. Multivariate analysis found that prior administration of ceftazidime/avibactam (P=0.014; OR 16.7, 95% CI 1.8–158.6) was independently associated with the development of such infections.

In the present study, after the introduction of ceftazidime/avibactam in the ICU, an increase of ceftazidime/avibactam resistance was noted due to an increased incidence of MBL-producing isolates.

In a European, as well as in a Greek study, >98% of MBL-negative isolates were ceftazidime/avibactam susceptible, but these data were collected before its widespread use.[3,7] As

Table 1. Univariate analysis of risk factors for CZA-R BSI during 2018

Characteristic	CZA-S BSI (*n*=18)	CZA-R BSI (*n*=27)	*P*
Days at risk[a]	45.8 ± 64.6	53.5 ± 70.7	0.372
Demographics			
age (years)	60.2 ± 15.5	50.4 ± 14.6	0.041
male gender	14 (77.8)	21 (77.8)	1.000
Chronic diseases			
diabetes mellitus	2 (11.1)	2 (11.1)	1.000
COPD	0 (0.0)	4 (14.8)	0.138
chronic heart failure	0 (0.0)	1 (3.7)	1.000
chronic renal failure	0 (0.0)	3 (11.1)	0.264
malignancy	1 (5.6)	1 (3.7)	1.000
immunosuppression	0 (0.0)	1 (3.7)	1.000
obesity (BMI $\geq$30 kg/m^2)	7 (38.9)	8 (29.6)	0.538
Charlson Comorbidity Index	2.6 ± 1.8	2.6 ± 3.3	0.368
Admission data			
APACHE II score upon admission	19.3 ± 5.3	16.2 ± 5.4	0.128
prior surgery	6 (33.3)	8 (29.6)	1.000
Prior antibiotic administration[b]			
penicillin/β-lactamase inhibitors	11 (61.1)	24 (88.9)	0.064
third- and fourth-generation cephalosporins	4 (22.2)	13 (48.1)	0.118
ceftazidime/avibactam	1 (5.6)	13 (48.1)	0.003
carbapenems	13 (72.2)	19 (70.4)	1.000
quinolones	3 (16.7)	10 (37.0)	0.188
colistin	12 (66.7)	20 (74.1)	0.739
aminoglycosides	6 (33.3)	5 (18.5)	0.304
fosfomycin	2 (11.1)	2 (7.4)	1.000
tigecycline	11 (61.1)	17 (63.0)	1.000
glycopeptides	15 (83.3)	20 (74.1)	0.716
linezolid	10 (55.6)	20 (74.1)	0.218
daptomycin	4 (22.2)	10 (37.0)	0.343
number of antibiotics administered	5.3 ± 2.9	6.6 ± 2.8	0.136
antifungal administration	11 (61.1)	21 (77.8)	0.317
ICU procedures			
corticosteroid administration	12 (66.7)	17 (63.0)	1.000
parenteral nutrition	10 (55.6)	12 (44.4)	0.550
enteral nutrition	12 (66.7)	22 (81.5)	0.304
Microbiological data			
resistance of bacteraemic isolate			
meropenem	18 (100)	27 (100)	–
colistin	12 (66.7)	8 (29.6)	0.031
tigecycline	4 (22.2)	9 (33.3)	0.514
gentamicin	18 (100)	25 (92.6)	0.509
fosfomycin	8 (44.4)	11 (40.7)	1.000
primary bacteraemia	7 (38.9)	16 (59.3)	0.231

Data are *n* (%) of patients or mean ± SD.
[a]Length of stay until BSI development.
[b]Administration on the last 30 days prior to BSI onset.

previously shown, ceftazidime/avibactam resistance among KPC-producing isolates could arise during treatment.[2,8] In our setting, it remains low (18%). The two published mechanisms were plasmid-borne bla_{KPC-3} mutations,[9] or porin deficiency combined with high ceftazidime hydrolysis.[10]

The main problem were the MBL-producing isolates, which are *de facto* resistant to ceftazidime/avibactam, especially VIM-producing isolates which were replaced in our hospital during 2009 by KPC-producing strains, and since then accounted for <4% of CP-Kp BSIs.[11] This abrupt change in carbapenemase epidemiology

renders the empirical use of ceftazidime/avibactam problematic, while it remains a safe and efficacious option as a definitive therapy after the acquisition of antimicrobial susceptibility results. This is another setback in the fight against CP-Kp, since resistance to other last-line antibiotics (colistin, tigecycline and gentamicin) is already high, and a combination treatment, which was associated with higher survival, cannot be assured.[3,4,11,12]

To the best of our knowledge, this is the first study to identify risk factors for the development of BSI due to CZA-R. The only risk factor found was the prior administration of ceftazidime/avibactam, which we hypothesize disturbed the enteric flora, creating a susceptible substrate for colonization by resistant strains. A previous study identified renal replacement therapy as a risk factor for ceftazidime/avibactam resistance development during ceftazidime/avibactam treatment among patients with microbiological failure.[13] In our study, the four patients with CZA-R KPC-producing *K. pneumoniae* received ceftazidime/avibactam the month prior to BSI onset, while this was true for 9 out of 23 BSIs due to MBL-producing isolates. Since half of patients with CZA-R BSI did not receive ceftazidime/avibactam, this suggests that cross-transmission of such isolates might have played an important role in their dissemination in our setting.

The study has some limitations. It is a retrospective study with a small number of patients with CP-Kp bacteraemia. No active surveillance of rectal colonization was implemented during the study period in order to ascertain the hypothesis that CZA-R strains were transmitted from colonized patients in nearby beds via the staff or arise from susceptible isolates after exposure to ceftazidime/avibactam.

Ceftazidime/avibactam remains an active antibiotic against KPC-producing *K. pneumoniae*, but may lead to a change in the palette of carbapenemases by replacing KPC- with MBL-producing isolates. The results of our study highlight the need to optimize the appropriate and judicious use of ceftazidime/avibactam to minimize the consequences associated with antibiotic-resistant organisms.

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Transparency declarations

None to declare.

References

1 Sternbach N, Leibovici Weissman Y *et al*. Efficacy and safety of ceftazidime/avibactam: a systematic review and meta-analysis. *J Antimicrob Chemother* 2018; **73**: 2021–9.

2 van Duin D, Lok JJ, Earley M *et al*. Colistin versus ceftazidime–avibactam in the treatment of infections due to carbapenem-resistant Enterobacteriaceae. *Clin Infect Dis* 2018; **66**: 163–71.

3 Galani I, Karaiskos I, Karantani I *et al*. Epidemiology and resistance phenotypes of carbapenemase-producing *Klebsiella pneumoniae* in Greece, 2014 to 2016. *Euro Surveill* 2018; doi:10.2807/1560-7917.

4 Papadimitriou-Olivgeris M, Bartzavali C, Spyropoulou A *et al*. Molecular epidemiology and risk factors for colistin- or tigecycline-resistant carbapenemase-producing *Klebsiella pneumoniae* bloodstream infection in critically ill patients during a 7-year period. *Diagn Microbiol Infect Dis* 2018; **92**: 235–40.

5 EUCAST. *Breakpoint Tables for Interpretation of MICs and Zone DiametersVersion 8.1*. 2018. http://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Breakpoint_tables/v_8.1_Breakpoint_Tables.pdf.

6 Queenan AM, Bush K. Carbapenemases: the versatile β-lactamases. *Clin Microbiol Rev* 2007; **20**: 440–58, table of contents.

7 Kazmierczak KM, de Jonge BLM, Stone GG *et al*. In vitro activity of ceftazidime/avibactam against isolates of Enterobacteriaceae collected in European countries: INFORM global surveillance 2012–15. *J Antimicrob Chemother* 2018; **73**: 2782–8.

8 Shields RK, Potoski BA, Haidar G *et al*. Clinical outcomes, drug toxicity, and emergence of ceftazidime–avibactam resistance among patients treated for carbapenem-resistant Enterobacteriaceae infections. *Clin Infect Dis* 2016; **63**: 1615–8.

9 Shields RK, Chen L, Cheng S *et al*. Emergence of ceftazidime–avibactam resistance due to plasmid-borne bla_{KPC-3} mutations during treatment of carbapenem-resistant *Klebsiella pneumoniae* infections. *Antimicrob Agents Chemother* 2017; **61**: e02097–16.

10 Shen Z, Ding B, Ye M *et al*. High ceftazidime hydrolysis activity and porin OmpK35 deficiency contribute to the decreased susceptibility to ceftazidime/avibactam in KPC-producing *Klebsiella pneumoniae*. *J Antimicrob Chemother* 2017; **72**: 1930–6.

11 Spyropoulou A, Papadimitriou-Olivgeris M, Bartzavali C *et al*. A ten-year surveillance study of carbapenemase-producing *Klebsiella pneumoniae* in a tertiary care Greek university hospital: predominance of KPC- over VIM- or NDM-producing isolates. *J Med Microbiol* 2016; **65**: 240–6.

12 Bassetti M, Giacobbe DR, Giamarellou H *et al*. Management of KPC-producing *Klebsiella pneumoniae* infections. *Clin Microbiol Infect* 2018; **24**: 133–44.

13 Shields RK, Nguyen MH, Chen L *et al*. Pneumonia and renal replacement therapy are risk factors for ceftazidime–avibactam treatment failures and resistance among patients with carbapenem-resistant Enterobacteriaceae infections. *Antimicrob Agents Chemother* 2018; **62**: e02497–17.