REVIEW 10.1111/1469-0691.12697

Treating infections caused by carbapenemase-producing *Enterobacteriaceae*

L. S. Tzouvelekis[1], A. Markogiannakis[2], E. Piperaki[1], M. Souli[3] and G. L. Daikos[4]

1) *Department of Microbiology, School of Medicine, University of Athens,* 2) *Department of Pharmacy, Laikon General Hospital,* 3) *Fourth Department of Medicine, and* 4) *First Department of Propaedeutic Medicine, University of Athens, Athens, Greece*

Abstract

Carbapenemase-producing *Enterobacteriaceae* (CPE) have spread worldwide, causing serious infections with increasing frequency. CPE are resistant to almost all available antibiotics, complicating therapy and limiting treatment options. Mortality rates associated with CPE infections are unacceptably high, indicating that the current therapeutic approaches are inadequate and must be revised. Here, we review 20 clinical studies (including those describing the largest cohorts of CPE-infected patients) that provided the necessary information regarding isolate and patient characteristics and treatment schemes, as well as a clear assessment of outcome. The data summarized here indicate that treatment with a single *in vitro* active agent resulted in mortality rates not significantly different from that observed in patients treated with no active therapy, whereas combination therapy with two or more *in vitro* active agents was superior to monotherapy, providing a clear survival benefit (mortality rate, 27.4% vs. 38.7%; $p < 0.001$). The lowest mortality rate (18.8%) was observed in patients treated with carbapenem-containing combinations.

Keywords: Antibiotic combinations, carbapenem, carbapenemase, *Enterobacteriaceae*, *Klebsiella pneumoniae*, treatment
Article published online: 29 May 2014
Clin Microbiol Infect 2014; **20**: 862–872

Corresponding author: G. L. Daikos, First Department of Propaedeutic Medicine, Medical School, University of Athens, Mikras Asias 75, Athens 115-26, Greece
E-mail: gdaikos@med.uoa.gr

Introduction

Carbapenemase-producing *Enterobacteriaceae* (CPE), which affect mostly seriously ill hospitalized patients, have been a cause of concern worldwide for more than a decade. However, many important treatment issues are still being debated [1–4]. The lack of randomized clinical trials has been mentioned on many occasions as one of the main factors hampering optimization of antibiotic treatment against CPE infections [3,5,6]. In fact, devising highly effective therapeutic regimens with the currently available antibiotics is probably not feasible, given that the vast majority of CPE isolates are resistant to the most clinically reliable antibiotic classes, β-lactams and aminoglycosides. Moreover, the emergence and increasing prevalence of CPE strains showing decreased susceptibility to colistin and/or tigecycline [7–9], two drugs that, despite their doubtful efficacy in various types of CPE infection, have become the first-line choices [1,10], further restrict the inherently limited therapeutic options. One might reasonably suppose that we are close to or even have already reached an impasse, and that completely new antimicrobials are urgently needed [11–13]. Then again, data from *in vitro* and *in vivo* experimental studies and, most importantly, the accumulation and systematic analysis of clinical observations regarding the efficacy of antibiotic regimens used in CPE infections, have provided opportunities to substantially improve therapeutic approaches with the currently available drugs [2–4,10,14]. Here, we have attempted to summarize the current situation as regards the treatment of CPE infections. Emphasis has been given to the most debatable issues, combination therapy and the role of carbapenems being among them.

Changing Profile of the CPE population

There have been clear indications of an ongoing geographical expansion of certain CPE strains. After the global dissemina-

tion of *Klebsiella pneumonia* carbapenemase (KPC) producers, it seems that New Delhi metallo-β-lactamase (NDM)-positive and, at an apparently higher rate, OXA-48-positive strains are spreading throughout the world. In addition, the bla_{KPC}, bla_{NDM} and bla_{OXA-48} gene variants are now encountered in a wide variety of distinct strains belonging to several enterobacterial species, although *K. pneumoniae* and *Escherichia coli* remain the predominant ones [1,15–17]. The bla_{VIM} and bla_{IMP} genes are also found in several enterobacterial species; nevertheless, these strains are largely confined to their original foci, i.e. the Mediterranean countries and the Far East, respectively. Of particular importance is the fact that CPE are no longer confined within the hospital environment. Apart from the early recognized spread in long-term-care facilities [18], CPE are currently found in the community [15,19] and in food-producing animals [20,21]. From a clinical point of view, the most important development concerns susceptibility to colistin and tigecycline. Not unexpectedly, CPE isolates showing decreased susceptibility and/or resistance to the latter drugs have occurred in high-prevalence settings where colistin and tigecycline are heavily used [22,23]. An additional negative development is the production of rRNA methyltransferases of the ArmA/RmtA family by most NDM producers, which precludes the use of all clinically available aminoglycosides [24,25]. To our knowledge, a systematic appraisal of the trends for carbapenem MICs of CPE populations has not been published. Our own 4-year records (2010–2013) from selected sentinel hospitals in Athens, Greece indicate increases in carbapenem MIC_{50} and MIC_{90} values in CPE (mostly KPC-positive *K. pneumoniae* isolates) (G. L. Daikos, unpublished data). On the other hand, a possible predominance of strains producing OXA-48 (far from being considered to be a 'positive' development) may decrease the respective values, as the latter enzyme and its derivatives show weaker activity against carbapenems than the other types of acquired carbapenemases. Also, a minority of OXA-48-positive enterobacteria do not co-produce extended-spectrum β-lactamases (ESBLs). Consequently, they are susceptible to newer-generation cephalosporins, which are not included in the substrate spectrum of OXA-48 [26].

CPE and Antibiotics in the Laboratory

In vitro studies

Owing to the multidrug-resistant nature of CPE, *in vitro* studies focus mainly on the search for combinations of antibiotics with synergistic activity, mostly using time-kill assays and, to a lesser extent, checkerboard techniques and the chemostat model. Apart from the application of different techniques, results (i.e. synergy, indifference, and antagonism) are strongly influenced by the MIC fraction of the tested antibiotic(s). Consequently, the findings of these studies are often not readily comparable, and are occasionally conflicting or difficult to interpret. On the other hand, a rather consistent finding in various time-kill studies is the synergism between carbapenems and aminoglycosides [27–29]. Also, a recent meta-analysis of relevant studies by Zusman *et al.* [30] showed that combinations of carbapenems, especially doripenem, with polymyxins often result in a synergistic effect against carbapenem-resistant *K. pneumoniae* isolates, whereas antagonism is rare. Needless to say, these *in vitro* interactions, regardless of how consistent and strong they may be, would not necessarily translate into a favourable clinical outcome.

Animal infection models

Despite the fact that assessing the efficacy of antibiotic regimens in animal experimental infections caused by CPE, especially with doses simulating human pharmacokinetics, may offer straightforward information, the number of relevant studies is relatively small, and the studies are mainly focused on carbapenems. Nevertheless, these studies provided indications that exploiting the pharmacokinetic/pharmacodynamic (PK/PD) features of these drugs can maximize their *in vivo* efficacy against VIM-producing and KPC-producing enterobacteria [1,2]. Indeed, inclusion of a carbapenem given at high doses and by prolonged infusion in combination regimens is gaining ground in clinical practice (discussed in the next section). In recent studies, doripenem was found to be efficacious against NDM-1-producing enterobacteria in the murine thigh infection model; notably, the drug showed remarkable activity against doripenem-resistant isolates [31,32]. Doripenem, in combination with amikacin, has also been found to be effective in a murine pneumonia model [29]. On the other hand, the activity of carbapenem was clearly inferior to that of ceftazidime against OXA-48-positive, ESBL-negative enterobacteria in both the murine thigh and peritonitis models, even when the MICs of the former drugs were within the susceptible range [33,34].

Antimicrobial Therapy

Ideally, antibiotic treatment schemes for CPE infections should be based on data obtained from randomized controlled trials (RCTs). Despite the decade-long—and apparently worsening—'carbapenemase problem', only two such trials, focusing on the evaluation of colistin vs. colistin–carbapenem combinations, are being conducted in Europe (AIDA, NCT01732250; http://clinicaltrials.gov/show/NCT01732250) and the USA (NCT01597973; http://clinicaltrials.gov/show/NCT01597973).

Given the scarcity of these technically and administratively demanding trials, therapeutic approaches in CPE infections are inevitably based on the accumulating clinical experience. It must be emphasized that even the most meticulous reviews of case reports, case series and observational studies cannot replace RCTs. Indeed, the wide heterogeneity of the published studies (different types of infections, different groups of patients, and the variety of treatment regimens, which, to make matters worse, have been evaluated with different methods and outcome definitions) causes serious difficulties in data compilation, and precludes the possibility of a rigorous meta-analysis. Notwithstanding these limitations, critical appraisal of the published clinical data is not only justifiable but also necessary in order to improve the care of CPE-infected patients. Indeed, in most relevant studies including large cohorts of patients with severe underlying conditions, the unacceptably high mortality rates associated with CPE infections (40% ± 10% are commonly reported) suggest that 'appropriate antibiotic therapy' is something of a euphemism, and that, in fact, our MIC-based approaches must be revised while the results of RCTs are awaited. It should be also mentioned that the design of the latter studies is largely guided by collective clinical experience.

The problem could be partly alleviated by the introduction of new drugs. Indeed, various compounds with potential anti-CPE activity have been developed by the industry (reviewed in [2,11,25,35,36]). From the publicly available information, however, it seems that only four new drugs—plazomicin (a sisomicin derivative that withstands the activity of all important aminoglycoside-modifying enzymes) [37], as well as the potent inhibitors of class A carbapenemases avibactam, MK-7655 (both diazabicyclooctane derivatives) [38,39] and the boronate RPX7009 [36]—are expected to be clinically available within the near future. It is therefore, evident that we shall continue to rely on the available antibiotics and must try to optimize their efficacy against CPE.

The increasing amount of clinical data and the systematic presentation of large cohorts of CPE-infected patients during recent years prompted us to revisit the relevant therapeutic issues. We performed a systematic review of the literature to identify studies reporting on CPE infections that contained adequate information regarding the *in vitro* susceptibilities of the infecting organisms to the antimicrobials used, the type of carbapenemase produced, the treatment schemes, and a clear assessment of the outcomes in terms of mortality rates. All studies available in MEDLINE evaluating the treatment and outcome of CPE-infected patients were considered (search terms: *Enterobacteriaceae*, *Klebsiella pneumoniae*, infection, bloodstream, bacteraemia (bacteremia), sepsis, carbapenem resistance, carbapenemase, KPC, metallo-β-lactamase, VIM, IMP, NDM, and OXA-48). Twenty studies that included data for more than ten patients and provided the necessary information were selected for review [40–59] (Table 1).

Although CPE have been established as important nosocomial pathogens for more than a decade in many parts of the world, there is a paucity of clinical data from countries that have been most affected, i.e. the Indian subcontinent, China, and Israel. Moreover, these studies have focused on *K. pneumoniae* producing mainly KPC or VIM, as clinical experience with other CPE is quite limited. A total of 907 patients infected with carbapenemase-producing *K. pneumoniae* were identified, 683 (75.3%) producing KPC-type enzymes, 188 (20.7%) producing VIM, and 36 (4.0%) producing OXA-48. The vast majority of these patients had serious infections: 339 had primary bacteraemia, 135 had bacteraemia related to intravascular catheters, and, of the remaining 433, 198 had pneumonia, 96 had urinary tract infections, 83 had intra-abdominal infections, and 56 had other infections. It is of note that most (approximately two-thirds) of the latter 433 patients had infections complicated with secondary bacteraemias. The affected patients were debilitated, with serious underlying diseases and comorbid conditions. A large proportion (70%) were intensive-care unit patients, but a significant number of patients were in surgical and medical wards. More worryingly, CPE have spread to solid organ transplant recipients and patients with haematological malignancies [60,61].

The efficacy of various antibiotic regimens used against CPE infections was assessed by compiling data from 889 patients. Among these patients, 441 (48.6%) received combination therapy (at least two drugs were active *in vitro* against the infecting organism), 346 (38.1%) received monotherapy (one drug was active *in vitro*), and 102 (11.3%) received inappropriate therapy (no drug was active *in vitro*). It should be noted that carbapenem susceptibility status considered as reported in the selected studies, in the majority of which the previous CLSI interpretive criteria were applied. Treatment with a single *in vitro* active agent, i.e. carbapenem, tigecycline, or colistin, resulted in unacceptably high mortality rates (40.1%, 41.1%, and 42.8%, respectively), similar to that observed in patients who received 'inappropriate' therapy (46.1%). The poor performance of monotherapy against CPE infections was more apparent in critically ill patients with severe sepsis, in patients with septic shock and in patients with rapidly fatal underlying disease, resulting in even higher mortality rates, ranging from 49% to 83.3%, according to the findings of a recent study [59].

In contrast, combination therapy provided a survival benefit and was superior to monotherapy (Fig. 1). By dividing the patients who received combination therapy into two groups

TABLE 1. Narrative summary of case series and observational studies (including more than ten patients) of carbapenemase-producing *Klebsiella pneumoniae*, with strain and patient characteristics, antibiotic treatment, and outcome

Reference	Country of publication, year	Enzyme type(s)	Imipenem/meropenem breakpoint used by the authors (mg/L)	No. of patients (type of infection)	Severity of underlying disease	Antibiotic regimen	No. of patients who survived/ died	Mortality rate, % (time of assessment)
40	Greece, 2007	VIM	≤4	28 BSIs (17 primary bacteraemias, 3 pneumonias, 2 urinary tract infections, 6 intravascular catheter-related)	APACHE II score (mean ± SD), 15.75 ± 5.15	CARB COL AMG CARB + AMG CARB + COL ATM + AMG No active drug	7/1 0/4 2/1 6/0 1/0 1/1 2/2	32.1 (14-day mortality)
41	Greece, 2008	VIM	≤4	15 BSIs (8 primary bacteraemias, 4 intravascular catheter infections, 1 pneumonia, 1 surgical site infection, 1 cholangitis) 2 VAP	APACHE II score (mean; range), 22; 10–33	COL TIG COL + AMG COL + DOX CARB + COL CARB + AMG + DOX	6/0 0/1 2/0 0/1 5/1 1/0	17.6 (in-hospital mortality)
42	Greece, 2009	VIM	≤4	67 BSIs (15 primary bacteraemias, 30 pneumonias, 4 urinary tract infections, 7 intravascular catheter infections, 4 soft tissue infections, 7 intra-abdominal infections)	Non-fatal:[a] 1 patient Ultimately fatal: 58 patients Rapidly fatal: 8 patients	CARB COL AMG CARB + AMG or COL No active drug	1/3 11/4 5/3 11/1 14/4	23.9 (14-day mortality)
43	USA, 2009	KPC	≤4	8 BSIs (4 primary bacteraemias, 2 urosepsis, 1 pneumonia, intravascular catheter infection), 4 pneumonias, 9 other infections	APACHE II score (mean; range) 21.1; 6–37	CARB TIG AMG CARB + TIG TIG + AMG No active drug	3/1 4/1 3/0 0/1 1/0 3/4	33.3 (in-hospital mortality)
44	Greece, 2009	KPC	≤4	9 pneumonias, 3 BSIs (2 primary bacteraemias, 1 intravascular catheter infection), 2 surgical site infections	Not reported	AMG COL + TIG COL + AMG COL + TIG + AMG No active drug	1/0 4/2 3/0 2/0 0/2	28.6 (14-day mortality)
45	Greece, 2010	KPC	≤4	14 BSIs (7 primary bacteraemias, 5 intravascular catheter infections, 1 pneumonia, 1 cholangitis), 2 surgical site infections, 1 urinary tract infection, 1 VAP	APACHE II score (mean; range), 18.4; 8–37	COL TIG AMG COL + AMG COL + TIG + AMG No active drug	6/5 1/0 1/1 1/0 1/0 1/1	38.9 (in-hospital mortality)
46	Greece, 2010	VIM, KPC	≤4	37 BSIs (primary source not reported)	APACHE II score (mean; range), 23; 4–36	COL COL + AMG	8/12 8/9	56.7 (in-hospital mortality)
47	Greece, 2011	KPC	≤4	53 BSIs (23 primary bacteremias, 12 catheter infections, 7 pneumonias, 6 urinary tract infections, 4 soft tissue infections, 1 central nervous system infection)	APACHE II score (mean ± SD), 21.1 ± 8.2	CARB COL TIG AMG COL + AMG CARB + AMG TIG + COL or AMG CARB + TIG CARB + TIG + COL TIG + COL + AMG No active drug	0/1 3/4 3/2 2/0 2/0 1/0 12/0 1/0 1/0 3/0 7/11	34.0 (in-hospital mortality)
48	Brazil, 2012	KPC	≤4	8 BSIs (4 primary bacteraemias, 2 pneumonias, 2 urinary tract infections), 2 urinary tract infections, 2 surgical site infections	Not reported	COL CARB COL + CARB TIG + CARB COL + TIG	2/1 0/2 1/2 1/0 3/0	41.7 (30-day mortality)
49	USA, 2012	KPC	≤4	36 BSIs (13 intravascular catheter infections, 10 pneumonias, 6 primary bacteraemias, 7 urinary tract infections)	APACHE II score (mean ± SD): monotherapy, 17.4 ± 6.65; combination therapy, 21.3 ± 8.69	COL CARB TIG AMG	3/4 2/1 1/4 2/0	38.2 (28-day mortality)

Table I (*Continued*)

Reference	Country of publication, year	Enzyme type(s)	Imipenem/meropenem breakpoint used by the authors (mg/L)	No. of patients (type of infection)	Severity of underlying disease	Antibiotic regimen	No. of patients who survived/ died	Mortality rate, % (time of assessment)
						CARB + COL	4/1	
						CARB + TIG	3/0	
						TIG + AMG	2/0	
						COL + TIG	1/0	
						COL + FQ	1/0	
						No active drug	2/3	
50	Italy, 2012	KPC	≤4	125 BSIs (75 primary bacteraemias, 28 pneumonias, 17 urinary tract infections, 13 intravascular catheter infections)	APACHE II score (mean ± SD): survivors, 24 ± 15; non-survivors, 40 ± 22	TIG	9/10	41.6 (30-day mortality)
						COL	11/11	
						AMG	9/4	
						COL + TIG	16/7	
						TIG + AMG	6/6	
						COL + AMG	1/6	
						CARB + COL or AMG	10/4	
						CARB + TIG + COL	14/2	
						CARB + AMG + TIG or COL	5/2	
51	Spain, 2012	OXA-48	≤2	13 surgical site infections, 8 urinary tract infections, 10 BSIs (6 primary bacteraemias, 4 intravascular catheter infections), 1 pneumonia	Charlson index (median; range), 5; 0–11	CARB	0/1	61.1 (in-hospital mortality)
						TIG	0/1	
						AMG	1/2	
						COL	1/0	
						CARB-containing combination	3/3	
						CARB-sparing combination	8/11	
						No active drug	1/4	
52	Italy, 2013	KPC	≤2	12 BSIs (7 primary bacteraemias, 5 VAPs) 11 VAP, 2 urinary tract infections, 1 peritonitis	Not reported	CARB-sparing combination (based on high dose of tigecycline, 100 mg every 12 h)	19/3	13.6 (30-day mortality)
53	Italy, 2013	KPC	≤2	30 BSIs (26 intra-abdominal infections, 4 surgical site infections)	APACHE II score (mean ± SD), 23.4 ± 1.7	TIG + COL (12 patients received a high dose of tigecycline, 100 mg every 12 h)	18/12	40.0 (30-day mortality)
54	USA, 2013	KPC	≤1	17 BSIs (8 intra-abdominal infections, 3 primary bacteraemias, 2 pneumonias, 2 urinary tract infections, 2 intravascular catheter infections)	APACHE II score (mean; range), 18; 4–26)	COL	2/0	17.6 (30-day mortality)
						AMG	4/0	
						CIP	1/0	
						CARB + COL	1/0	
						CARB + DOX	1/0	
						CARB + COL + AMG + TIG	1/0	
						No active drug	4/3	
55	Spain, 2014	VIM	≤4	16 BSIs (5 pneumonias, 5 urinary tract infections, 3 peritonitis, 1 meningitis, 2 intravascular catheter infections)	APACHE II score (mean; range), 16.7; 7–27	TIG	6/2	25.0 (30-day mortality)
						TIG + COL	5/2	
						TIG + COL + AMG	1/0	
56	Greece, 2014	VIM, KPC	≤1	69 BSIs (39 intravascular catheter infections, 30 primary bacteraemias), 35 VAPs, 13 urinary tract infections, 6 intra-abdominal infections, 4 surgical site infections	APACHE II score (range), 36–58	TIG	11/5	21.6 (14-day mortality)
						COL	20/6	
						AMG	17/5	
						FQ	Not reported	
						TIG + AMG	9/2	
						COL + AMG	15/2	
						COL + TIG	5/4	
						TIG + COL + AMG	4/0	
						CARB + COL + AMG + TIG	Not reported	
						No active drug	13/7	
57	USA, 2014	KPC	≤4	15 BSIs (7 urinary tract infections, 3 pneumonias, 3 intravascular catheter infections, 1 primary bacteraemia, 1 intra-abdominal infection)	APACHE II score (mean; range), 12.1; 2–22	CARB	0/1	33.3 (in-hospital mortality)
						AMG	2/0	
						TIG + AMG	0/1	
						COL + TIG	1/0	
						TIG + COL + AMG	1/1	
						TIG + COL + FOS	1/0	
						TIG + AMG + FOS	1/0	
						TIG + COL + FOS	1/0	
						CARB + COL + TIG	1/0	
						CARB + COL + TIG + AMG	1/0	

Table I (*Continued*)

Reference	Country of publication, year	Enzyme type(s)	Imipenem/meropenem breakpoint used by the authors (mg/L)	No. of patients (type of infection)	Severity of underlying disease	Antibiotic regimen	No. of patients who survived/ died	Mortality rate, % (time of assessment)
58	Greece, 2014	KPC	≤1	15 BSIs (5 primary bacteraemias, 4 VAPs, 2 intra-abdominal infections, 2 intravascular catheter infections, 1 urinary tract infection, 1 meningitis)	APACHE II score (mean ± SD), 20.5 ± 6.0	No active drug	1/2	40.0 (28-day mortality)
						TIG + FOS	2/0	
						COL + FOS	4/2	
						AMG + FOS	1/1	
						TIG + AMG + FOS	1/0	
						COL + AMG + FOS	0/1	
						TIG + COL + FOS	1/2	
59	Greece, 2014	VIM, KPC	≤8	205[b] BSIs (83 primary bacteraemias, 43 pneumonias, 19 urinary tract infections, 6 surgical site infections, 29 intra-abdominal infections, 22 intravascular catheter infections, 3 other infections)	Non fatal: 109 patients Ultimately fatal: 53 patients Rapidly fatal: 43 patients	TIG	16/11	36.4 (28-day mortality)
						COL	10/12	
						AMG	7/2	
						CARB	5/7	
						Other monotherapy	2/0	
						CARB-containing regimen	25/6	
						CARB-sparing regimen	50/22	
						No active drug	4/8	

BSI, bloodstream infection; CARB, carbapenem; COL, colistin; AMG, aminoglycoside; ATM, aztreonam; TIG, tigecycline; DOX, doxycycline; FQ, fluoroquinolone; FOS, fosfomycin; SD, standard deviation; VAP, ventilator-associated pneumonia.
[a]McCabe and Jackson classification [79].
[b]Eighteen patients who died within 48 h after onset of bacteraemia were not included in the outcome analysis.

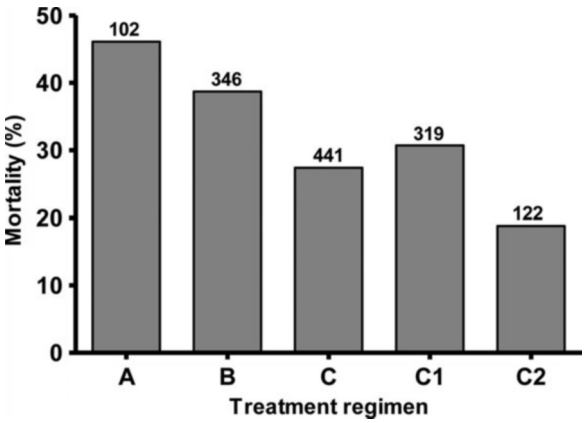

FIG. 1. Outcomes of patients infected with carbapenemase-producing *Klebsiella pneumoniae*, according to treatment regimen. Regimen A: inappropriate therapy (no drug was active *in vitro*). Regimen B: monotherapy (one drug was active *in vitro*). Regimen C: combination therapy (two or more drugs were active *in vitro*). Regimen C1: combination therapy with two or more *in vitro*-active drugs not including a carbapenem. Regimen C2: combination therapy with two or more *in vitro*-active drugs, one of which was a carbapenem. Regimen B vs. regimen A: p, not significant. Regimens C, C1 and C2 vs. regimen B: p 0.001, p 0.034, and p <0.0001, respectively. Numbers above columns indicate the number of patients.

on the basis of inclusion of a carbapenem in the treatment scheme, it was shown that the carbapenem-containing combinations resulted in significantly lower mortality rates (18.8%) than the carbapenem-sparing combinations (mortality rate, 30.7%) (Fig. 1).

To further assess the efficacy of different treatment schemes in various types of infection, we identified 414 patients (148 with primary bacteraemia, 125 with pneumonia, 41 with urinary tract infections, 53 with intravascular catheter-related bacteraemia, 41 with complicated intra-abdominal infections, and 45 with surgical site infections) for whom it was possible to extract information on treatment and outcomes per type of infection. As shown in Table 2, the lowest mortality rate (30.1%) was observed for urinary tract infections and the highest (38.5%) for primary bacteraemia. Monotherapy was associated with high mortality rates for all types of infection, whereas carbapenem-containing combinations appeared to be the most effective treatment regimens.

One should bear in mind that the above discussion is based on studies mainly reporting on infections caused by KPC-producing and VIM-producing isolates; only 36 patients infected with OXA-48 producers were included in the analysis. Similar data regarding infections caused by NDM-producing organisms are still limited. As with the other CPE, combinations including colistin and/or tigecycline are frequently employed in treating the respective infections [62]. However, the relatively low

TABLE 2. Outcome of 414 patients infected with carbapenemase-producing *Klebsiella pneumoniae* according to treatment regimen and type of infection

Treatment regimen	Primary bacteraemia No. of patients (mortality) (%)	Pneumonia[a] No. of patients (mortality) (%)	cIAI[b] No. of patients (mortality) (%)	Urinary tract infection[c] No. of patients (mortality) (%)	Surgical site infection[d] No. of patients (mortality) (%)
No active therapy	11 (54)	8 (50)	3 (33.3)	13 (38.5)	9 (44.4)
Monotherapy	56 (46.4)	45 (46.7)	3 (33.3)	18 (38.9)	22 (46.7)
Combination therapy	81 (34.6)	72 (29.1)	36 (30.6)	24 (20.8)	14 (28.6)
Carbapenem-sparing	59 (40.7)	54 (30.4)	35 (31.4)	14 (28.6)	8 (37.5)
Carbapenem-containing	22 (18.2)	18 (27.8)	1 (0.0)	10 (10)	6 (16.7)

[a]Eighty-eight patients were complicated with secondary bacteraemia.
[b]Complicated intra-abdominal infection; 40 patients had secondary bacteraemia.
[c]Thirty-eight patients had secondary bacteraemia.
[d]Twenty-two patients had secondary bacteremia.

carbapenem MICs in a significant proportion of OXA-48-producing *K. pneumoniae* should also allow treatment with carbapenem-containing combinations [51,63]. Furthermore, oximino-cephalosporins may be an option for OXA-48-positive *K. pneumoniae* not co-producing an ESBL [34].

It should be emphasized once again that the assessment of the available clinical data as attempted here lacks the characteristics of a vigorous meta-analysis, as it was not possible to measure and adjust for potential confounders, including patients' age and status, comorbitities, severity of sepsis, and time to initiation of appropriate antimicrobial treatment. Thus, we cannot exclude the possibility that our analysis, in some cases, might have resulted in biased associations between antimicrobial treatment and outcome. Nevertheless, given that the majority of patients infected with CPE are debilitated, with various underlying diseases, and that >80% of them had severe infections (bloodstream infections or pneumonia), it is unlikely that residual confounding could account, to an appreciable extent, for the significantly different failure rates between treatment groups. Indeed, in the largest two series that have adjusted for a number of potential confounders [50,59], among other factors, including severity of underlying disease, APACHE score, septic shock, and time to initiation of appropriate treatment, monotherapy was a predictor of death, whereas combination therapy provided a significant survival benefit that appeared to be more pronounced when a carbapenem was included in the regimen. These observations, along with the findings in the present analysis, may be taken as an indication that the superiority of combination therapy to monotherapy is, at least partly, driven by carbapenems and their potential additive or synergistic activity with aminoglycosides, colistin, or tigecycline. It must be pointed out, however, that the positive 'carbapenem effect' was clearly seen in infections where the carbapenem MIC of the responsible CPE strains was $\leq$ 8 mg/L, a limit that is in line with the EUCAST breakpoint and the respective PK/PD features of this group of β-lactams. Also, in the study of Tumbarello *et al.* [50], this effect was extended to strains with carbapenem MICs up to 16 mg/L, when the latter drugs were used in triple combinations. Although the number of such cases presented in the latter study was low, this remarkable observation deserves further attention.

Although there is a clear trend towards increasing resistance to colistin among CPE, this drug remains one of the most *in vitro*-active agents against these pathogens. It can be reasonably expected that its use, mostly in combination schemes, will continue. The inferior performance of colistin monotherapy against these infections may be explained, among other factors, by the delay in attaining an efficacious drug concentration and the suboptimal dosing of the drug. Kinetic studies in humans have shown that it takes at least 2 days for the drug to achieve steady state. An initial loading dose of 9 000 000 IU can overcome this delay [64]. Although this practice is now widely accepted by physicians and has a theoretical basis, no study so far has shown improvement in patient outcomes with application of this approach. Moreover, the current dosing schemes for colistin do not provide serum concentrations that would be sufficient for the treatment of infections caused by pathogens with MICs higher than 0.5 mg/L. As suggested by Garonzik *et al.* [65], in such cases the drug —if deemed to be potentially useful—should be used as part of combination schemes. As the complex PK/PD properties of the drug have only recently been elucidated [64–66], the optimal dosing of colistin remains to be determined. The commonly used schemes have been based primarily on data from animal infection models, indicating a correlation between exposure time and antibacterial activity [66]. Thus, searching for a more effective dosing scheme may be worth trying. The long half-life of colistin, as well as its concentration-dependent killing and adaptive resistance phenomena [67–69], favour the administration of this agent at higher dosages at longer intervals. However, the issues of nephrotoxicity and neurotoxicity should be priorities, if any effort to enhance colistin efficacy by changing the dosing schemes is to be made.

The results of tigecycline monotherapy were also poor. The relatively low clinical effectiveness of tigecycline in severe infections could be partly attributable to the PK/PD profile of the drug. Tigecycline shows mainly bacteriostatic activity against Gram-negative organisms, and the attainable drug concentrations at several anatomical sites are suboptimal. The serum concentrations achieved with the standard dosing regimen of the drug (50 mg twice daily) range from 0.6 mg/L to 0.9 mg/L, whereas those attained in the urine and in the epithelial lining fluid are several-fold lower [70–72]. The drug concentrations attainable with this standard dosing regimen, combined with this drug's MIC profile for contemporary CPE isolates, make it unlikely that tigecycline will cure CPE infections at anatomical sites where drug concentrations are suboptimal. By increasing the dose of tigecycline to 200 mg daily, it is possible to drive the PK/PD profile of the drug to acceptable exposures and improve patient outcomes [73]. Preliminary data obtained in critically ill patients with intra-abdominal infections caused by KPC-prodicing *K. pneumoniae* showed that 'high doses' of tigecycline (100 mg every 12 h) were associated with lower mortality rates than the conventional dosing scheme of the drug [53]. When we are faced with the daily challenge of managing critically ill patients with CPE infections, in the absence of alternative therapeutic options, it is inevitable that, on some occasions, the off-label 'high dose' of tigecycline will be used to optimize the therapeutic effectiveness of the drug. This approach, however, if adopted, should be practised with close monitoring for toxicity.

The number of CPE isolates showing resistance to almost all available agents is worryingly high in various settings [74]. Given that fosfomycin shows good *in vitro* activity against most CPE, this agent could be selected as salvage therapy in situations where therapeutic options are very limited [74,75]. Although the main indication for fosfomycin remains the treatment of lower urinary tract infections, some investigators have included this drug in various combination schemes to treat critically ill patients with CPE infections [58,76]. The available data, however, are too limited to allow a sound hypothesis regarding its efficacy. Also, the potential of fosfomycin to rapidly select resistant mutants during therapy is a matter for consideration [77].

A significant proportion of CPE, especially those producing KPC or VIM enzymes, show *in vitro* susceptibility to aminoglycosides (usually only to gentamicin or, to a lesser extent, amikacin) [50,59]. Taking mainly into account the extensive clinical experience with these antibiotics and their well-studied PK/PD characteristics, we have considered it reasonable to include them among the drugs that may be preferred in combination schemes [10]. According to the data reviewed here, treatment with an aminoglycoside alone is the most efficacious monotherapy, especially in urinary tract infections with or without secondary bacteraemia. It is also of note that therapeutic schemes including an aminoglycoside and a carbapenem appeared to be the most effective combinations (mortality rate, 11.1%; data not shown). Although the numbers of the respective cases were relatively low, this potential 'synergy', which is in line with *in vitro* and *in vivo* experimental data [27,29], may warrant further consideration.

It must be admitted that meta-analyses have produced conflicting results regarding the alleged superiority of combination therapy over monotherapy in infections caused by Gram-negative pathogens. As was pointed out in a recent review, the expectation of increasing therapeutic efficacy by exploiting the observed *in vitro* synergy between two antibiotics and of preventing the development of resistance during treatment are the main reasons for the preference of many clinicians to use antibiotic combination regimens; however, if we take into account only the data provided by RCTs, both notions are disputed [78]. We nevertheless believe that the analysis presented here provides clear indications in favour of the use of combination regimens for the treatment of CPE infections, particularly in severely ill patients. Moreover, it can be argued that most of the aforementioned RCTs used antibiotic regimens that included at least one reliable agent (usually a β-lactam), whereas, in the case of CPE infections, the baseline antimicrobials, namely colistin and tigecycline, are of doubtful efficacy.

Acknowledgements

We thank all our colleagues in hospitals throughout Greece for sharing valuable information.

Transparency Declaration

The authors declare no conflict of interest.

References

1. Tzouvelekis LS, Markogiannakis A, Psichogiou M, Tassios PT, Daikos GL. Carbapenemases in *Klebsiella pneumoniae* and other *Enterobacteriaceae*: an evolving crisis of global dimensions. *Clin Microbiol Rev* 2012; 25: 682–707.
2. Markogiannakis A, Tzouvelekis LS, Psichogiou M, Petinaki E, Daikos GL. Confronting carbapenemase-producing *Klebsiella pneumoniae*. *Future Microbiol* 2013; 8: 1147–1161.

3. Petrosillo N, Giannella M, Lewis R, Viale P. Treatment of carbapenem-resistant *Klebsiella pneumoniae*: the state of the art. *Expert Rev Anti Infect Ther* 2013; 11: 159–177.
4. Zavascki AP, Bulitta JB, Landersdorfer CB. Combination therapy for carbapenem-resistant Gram-negative bacteria. *Expert Rev Anti Infect Ther* 2013; 11: 1333–1353.
5. Hirsch EB, Tam VH. Detection and treatment options for *Klebsiella pneumonia* carbapenemases (KPCs): an emerging cause of multidrug-resistant infection. *J Antimicrob Chemother* 2010; 65: 1119–2225.
6. Kmeid JG, Youssef MM, Kanafani ZA, Kanj SS. Combination therapy for Gram-negative bacteria: what is the evidence? *Expert Rev Anti Infect Ther* 2013; 11: 1355–1362.
7. Capone A, Giannella M, Fortini D *et al.* High rate of colistin resistance among patients with carbapenem-resistant *Klebsiella pneumoniae* infection accounts for an excess of mortality. *Clin Microbiol Infect* 2013; 19: E23–E30.
8. Ah Y-M, Kim A-J, Lee J-Y. Colistin resistance in *Klebsiella pneumonia*. *Int J Antimicrob Agents* 2014; 44: 8–15.
9. Villa L, Feudi C, Fortini D, Garcia-Fernandez A, Carattoli A. Genomics of KPC-producing *Klebsiella pneumoniae* sequence type 512 clone highlights the role of RamR and ribosomal S10 protein mutations in conferring tigecycline resistance. *Antimicrob Agents Chemother* 2014; 58: 1707–1712.
10. Daikos GL, Markogiannakis A, Souli M, Tzouvelekis LS. Bloodstream infections caused by carbapenemase-producing *Klebsiella pneumoniae*: a clinical perspective. *Expert Rev Anti Infect Ther* 2012; 10: 1393–1404.
11. Patel G, Bonomo RA. Status report on carbapenemases: challenges and prospects. *Expert Rev Anti Infect Ther* 2011; 9: 555–570.
12. van Duin D, Kaye KS, Neuner EA, Bonomo RA. Carbapenem-resistant *Enterobacteriaceae*: a review of treatment and outcomes. *Diagn Microbiol Infect Dis* 2013; 75: 115–120.
13. Bassetti M, Merelli M, Temperoni C, Astilean A. New antibiotics for bad bugs: where are we? *Ann Clin Microbiol Antimicrob* 2013; 12: 22.
14. Tambyah PA, Hara GL, Daikos GL *et al.* Treatment of extensively drug-resistant Gram-negative infections in critically ill patients: outcome of a consensus meeting at the 13th Asia-Pacific Congress of Clinical Microbiology and Infection, October 2012. *J Global Antimicrob Resist* 2013; 1: 117–122.
15. Cantón R, Akóva M, Carmeli Y *et al.* Rapid evolution and spread of carbapenemases among *Enterobacteriaceae* in Europe. *Clin Microbiol Infect* 2012; 18: 413–431.
16. Nordmann P, Poirel L, Walsh TR, Livermore DM. The emerging NDM carbapenemases. *Trends Microbiol* 2011; 19: 588–595.
17. Munoz-Price LS, Poirel L, Bonomo RA *et al.* Clinical epidemiology of the global expansion of *Klebsiella pneumoniae* carbapenemases. *Lancet Infect Dis* 2013; 13: 785–796.
18. Urban C, Bradford PA, Tuckman M *et al.* Carbapenem-resistant *Escherichia coli* harboring *Klebsiella pneumoniae* carbapenemase β-lactamases associated with long-term care facilities. *Clin Infect Dis* 2008; 46: e127–e130.
19. Nordmann P, Poirel L, Toleman MA, Walsh TR. Does broad-spectrum β-lactam resistance due to NDM-1 herald the end of the antibiotic era for treatment of infections caused by Gram-negative bacteria? *J Antimicrob Chemother* 2011; 66: 689–692.
20. Fischer J, Rodríguez I, Schmoger S *et al. Salmonella enterica* subsp. *enterica* producing VIM-1 carbapenemase isolated from livestock farms. *J Antimicrob Chemother* 2013; 68: 478–480.
21. Fischer J, Rodríguez I, Schmoger S *et al. Escherichia coli* producing VIM-1 carbapenemase isolated on a pig farm. *J Antimicrob Chemother* 2012; 67: 1793–1795.
22. Giakkoupi P, Papagiannitsis CC, Miriagou V *et al.* An update of the evolving epidemic of bla_{KPC-2}-carrying *Klebsiella pneumoniae* in Greece (2009–10). *J Antimicrob Chemother* 2011; 66: 1510–1513.
23. Mammina C, Bonura C, Di Bernardo F *et al.* Ongoing spread of colistin-resistant *Klebsiella pneumoniae* in different wards of an acute general hospital, Italy, June to December 2011. *Euro Surveill* 2012; 17: pii: 20248.
24. Berçot B, Poirel L, Nordmann P. Updated multiplex polymerase chain reaction for detection of 16S rRNA methylases: high prevalence among NDM-1 producers. *Diagn Microbiol Infect Dis* 2011; 71: 442–445.
25. Wachino J, Arakawa Y. Exogenously acquired 16S rRNA methyltransferases found in aminoglycoside-resistant pathogenic Gram-negative bacteria: an update. *Drug Resist Updat* 2012; 15: 133–148.
26. Poirel L, Héritier C, Tolün V, Nordmann P. Emergence of oxacillinase-mediated resistance to imipenem in *Klebsiella pneumoniae*. *Antimicrob Agents Chemother* 2004; 48: 15–22.
27. Le J, McKee B, Srisupha-Olarn W, Burgess DS. *In vitro* activity of carbapenems alone and in combination with amikacin against KPC-producing *Klebsiella pneumoniae*. *J Clin Med Res* 2011; 19: 106–110.
28. Urban C, Mariano N, Rahal JJ. *In vitro* double and triple bactericidal activities of doripenem, polymyxin B, and rifampin against multidrug-resistant *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, and *Escherichia coli*. *Antimicrob Agents Chemother* 2010; 54: 2732–2734.
29. Hirsch EB, Guo B, Chang KT *et al.* Assessment of antimicrobial combinations for *Klebsiella pneumoniae* carbapenemase-producing *K. pneumoniae*. *J Infect Dis* 2013; 207: 786–793.
30. Zusman O, Avni T, Leibovici L *et al.* Systematic review and meta-analysis of *in vitro* synergy of polymyxins and carbapenems. *Antimicrob Agents Chemother* 2013; 57: 5104–5111.
31. Wiskirchen DE, Nordmann P, Crandon JL, Nicolau DP. Efficacy of humanized carbapenem exposures against New Delhi metallo-β-lactamase (NDM-1)-producing *Enterobacteriaceae* in a murine infection model. *Antimicrob Agents Chemother* 2013; 57: 3936–3940.
32. Wiskirchen DE, Nordmann P, Crandon JL, Nicolau DP. *In vivo* efficacy of human simulated regimens of carbapenems and comparator agents against NDM-1 producing *Enterobacteriaceae*. *Antimicrob Agents Chemother* 2014; 58: 1671–1677.
33. Wiskirchen DE, Nordmann P, Crandon JL, Nicolau DP. Efficacy of humanized carbapenem and ceftazidime regimens against *Enterobacteriaceae* producing the OXA-48 carbapenemase in a murine infection model. *Antimicrob Agents Chemother* 2014; 58: 1678–1683.
34. Mimoz O, Grégoire N, Poirel L, Marliat M, Couet W, Nordmann P. Broad-spectrum β-lactam antibiotics for treating experimental peritonitis in mice due to *Klebsiella pneumoniae* producing the carbapenemase OXA-48. *Antimicrob Agents Chemother* 2012; 56: 2759–2760.
35. Buynak JD. β-Lactamase inhibitors: a review of the patent literature (2010–2013). *Expert Opin Ther Pat* 2013; 23: 1469–1481.
36. Drawz SM, Papp-Wallace KM, Bonomo RA. New β-lactamase inhibitors: a therapeutic renaissance in an MDR world. *Antimicrob Agents Chemother* 2014; 58: 1835–1846.
37. Livermore DM, Mushtaq S, Warner M *et al.* Activity of aminoglycosides, including ACHN-490, against carbapenem-resistant *Enterobacteriaceae* isolates. *J Antimicrob Chemother* 2011; 66: 48–53.
38. Stachyra T, Levasseur P, Péchereau MC. *In vitro* activity of the β-lactamase inhibitor NXL104 against KPC-2 carbapenemase and *Enterobacteriaceae* expressing KPC carbapenemases. *J Antimicrob Chemother* 2009; 64: 326–329.
39. Livermore DM, Warner M, Mushtaq S. Activity of MK-7655 combined with imipenem against *Enterobacteriaceae* and *Pseudomonas aeruginosa*. *J Antimicrob Chemother* 2013; 68: 2286–2290.
40. Daikos GL, Karabinis A, Paramythiotou E *et al.* VIM-1-producing *Klebsiella pneumoniae* bloodstream infections: analysis of 28 cases. *Int J Antimicrob Agents* 2007; 29: 471–473.
41. Souli M, Kontopidou FV, Papadomichelakis E, Galani I, Armaganidis A, Giamarellou H. Clinical experience of serious infections caused by

Enterobacteriaceae producing VIM-1 metallo-β-lactamase in a Greek university hospital. *Clin Infect Dis* 2008; 46: 847–854.
42. Daikos GL, Petrikkos P, Psichogiou M *et al.* Prospective observational study of the impact of VIM-1 metallo-β-lactamase on the outcome of patients with *Klebsiella pneumoniae* bloodstream infections. *Antimicrob Agents Chemother* 2009; 53: 1868–1873.
43. Weisenberg SA, Morgan DJ, Espinal-Witter R, Larone DH. Clinical outcomes of patients with *Klebsiella pneumoniae* carbapenemase-producing *K. pneumoniae* after treatment with imipenem or meropenem. *Diagn Microbiol Infect Dis* 2009; 64: 233–235.
44. Maltezou HC, Giakkoupi P, Maragos A *et al.* Outbreak of infections due to KPC-2-producing *Klebsiella pneumoniae* in a hospital in Crete (Greece). *J Infect* 2009; 58: 213–219.
45. Souli M, Galani I, Antoniadou A *et al.* An outbreak of infection due to β-lactamase *Klebsiella pneumoniae* carbapenemase 2-producing *K. pneumoniae* in a Greek university hospital: molecular characterization, epidemiology, and outcomes. *Clin Infect Dis* 2010; 50: 364–373.
46. Mouloudi E, Protonotariou E, Zagorianou A *et al.* Bloodstream infections caused by metallo-β-lactamase *Klebsiella pneumoniae* carbapenemase-producing *K. pneumoniae* among intensive care unit patients in Greece: risk factors for infection and impact of type of resistance on outcomes. *Infect Control Hosp Epidemiol* 2010; 31: 1250–1256.
47. Zarkotou O, Pournaras S, Tselioti P *et al.* Predictors of mortality in patients with bloodstream infections caused by KPC-producing *Klebsiella pneumoniae* and impact of appropriate antimicrobial treatment. *Clin Microbiol Infect* 2011; 17: 1798–1803.
48. Bergamasco MD, Barroso-Barbarosa M, Oliveira Garcia D *et al.* Infection with *Klebsiella pneumoniae* carbapenemase (KPC)-producing *K. pneumoniae* in solid organ transplantation. *Transpl Infect Dis* 2012; 14: 198–205.
49. Qureshi ZA, Paterson DL, Potoski BA *et al.* Treatment outcome of bacteremia due to KPC-producing *Klebsiella pneumoniae*: superiority of combination antimicrobial regimens. *Antimicrobial Agents Chemother* 2012; 56: 2108–2113.
50. Tumbarello M, Viale P, Viscoli C *et al.* Predictors of mortality in bloodstream infections caused by *Klebsiella pneumoniae* carbapenemase-producing *K. pneumoniae*: importance of combination therapy. *Clin Infect Dis* 2012; 55: 943–950.
51. Navarro S-FC, Mora-Rillo M, Romero-Gómez MP *et al.* Bacteraemia due to OXA-48-carbapenemase-producing *Enterobacteriaceae*: a major clinical challenge. *Clin Microbiol Infect* 2013; 19: E72–E79.
52. Sbrana F, Malacarne P, Viaggi B *et al.* Carbapenem-sparing antibiotic regimens for infections caused by *Klebsiella pneumoniae* carbapenemase-producing *K. pneumoniae* in intensive care unit. *Clin Infect Dis* 2013; 56: 697–700.
53. Di Carlo P, Gulotta G, Casuccio A *et al.* KPC-3 *Klebsiella pneumoniae* ST258 clone infection in postoperative abdominal surgery patients in an intensive care setting: analysis of a case series of 30 patients. *BMC Anesthesiol* 2013; 13: 1–8.
54. Clancy CJ, Chen L, Shields RK *et al.* Epidemiology and molecular characterization of bacteremia due to carbapenem-resistant *Klebsiella pneumoniae* in transplant recipients. *Am J Transpl* 2013; 13: 2619–2633.
55. Balandin MB, Simón IF, García VP *et al.* Tigecycline therapy for infections due to carbapenemase-producing *Klebsiella pneumoniae* in critically ill patients. *Scand J Infect* 2014; 175: 175–180.
56. Kontopidou F, Giamarellou H, Katerelos P *et al.* Infections caused by carbapenem-resistant *Klebsiella pneumoniae* among patients in intensive care units in Greece: a multi-centre study on clinical outcome and therapeutic options. *Clin Microbiol Infect* 2014; 20: O117–O123.
57. McLaughlin MM, Advincula MR, Malczynski M, Barajas G, Qi C, Scheetz MH. Quantifying the clinical virulence of *Klebsiella pneumoniae* producing carbapenemase *Klebsiella pneumoniae* with a *Galleria mellonella* model and a pilot study to translate to patient outcomes. *BMC Infect Dis* 2014; 14: 1–10.
58. Pontikis K, Karaiskos I, Bastani S *et al.* Outcomes of critically ill intensive care unit patients treated with fosfomycin for infections due to pandrug-resistant and extensively drug-resistant carbapenemase-producing Gram-negative bacteria. *Int J Antimicrob Agents* 2014; 43: 52–59.
59. Daikos GL, Tsaousi S, Tzouvelekis LS *et al.* Carbapenemase-producing *Klebsiella pneumoniae* bloodstream infections: lowering mortality by antibiotic combination schemes and the role of carbapenems. *Antimicrob Agents Chemother* 2014; 58: 2322–2328.
60. Satlin MJ, Calfee DP, Chen L. Emergence of carbapenem-resistant *Enterobacteriaceae* as causes of bloodstream infections in patients with hematologic malignancies. *Leuk Lymphoma* 2013; 54: 799–806.
61. Satlin MJ, Jenkis SG, Walsh TJ. The global challenge of carbapenem-resistant *Enterobacteriaceae* in transplant recipients and patients with hematologic malignancies. *Clin Infect Dis* 2014; 58: 1274–1283.
62. Kumarasamy KK, Toleman MA, Walsh TR *et al.* Emergence of a new antibiotic resistance mechanism in India, Pakistan, and the UK: a molecular, biological, and epidemiological study. *Lancet Infect Dis* 2010; 10: 597–602.
63. Maherault A-C, Nordmann P, Therby A, Pangon B. Efficacy of imipenem for the treatment of bacteremia due to an OXA-48-producing *Klebsiella pneumoniae* isolate. *Clin Infect Dis* 2012; 54: 577–578.
64. Plachouras D, Karvanen M, Friberg LE *et al.* Population pharmacokinetic analysis of colistin methanesulfonate and colistin after intravenous administration in critically ill patients with infections caused by gram-negative bacteria. *Antimicrob Agents Chemother* 2009; 53: 3430–3436.
65. Garonzik SM, Li J, Thamlikitkul V *et al.* Population pharmacokinetics of colistin methanesulfonate and formed colistin in critically ill patients from a multicenter study provide dosing suggestions for various categories of patients. *Antimicrob Agents Chemother* 2011; 55: 3284–3294.
66. Dudhani RV, Turnidge JD, Nation RL, Li J. *f*AUC/MIC is the most predictive pharmacokinetic/pharmacodynamic index of colistin against *Acinetobacter baumanni* in murine thigh and lung infection models. *J Antimicrob Chemother* 2010; 65: 1984–1990.
67. Daikos GL, Skiada A, Pavleas J *et al.* Serum bactericidal activity of three different dosing regimens of colistin with implications for optimum clinical use. *J Chemother* 2010; 22: 175–178.
68. Skiada A, Markogiannakis A, Plachouras D, Daikos GL. Adaptive resistance to cationic compounds in *Pseudomonas aeruginosa*. *Int J Antimicrob Agents* 2011; 37: 187–193.
69. Fernández L, Gooderham WJ, Bains M, McPhee JB, Wiegand I, Hancock RE. Adaptive resistance to the 'last hope' antibiotics polymyxin B and colistin in *Pseudomonas aeruginosa* is mediated by the novel two-component regulatory system ParR–ParS. *Antimicrob Agents Chemother* 2010; 54: 3372–3382.
70. Agwuh KN, MacGowan A. Pharmacokinetics and pharmacodynamics of the tetracyclines including glycylcyclines. *J Antimicrob Chemother* 2006; 58: 256–265.
71. Burkhardt O, Rauch K, Kaever V, Hadem J, Kielstein JT, Welte T. Tigecycline possibly underdosed for the treatment of pneumonia: a pharmacokinetic viewpoint. *Int J Antimicrob Agents* 2009; 34: 101–102.
72. Peterson LR. A review of tigecycline—the first glycylcycline. *Int J Antimicrob Agents* 2008; 32(suppl 4): S215–S222.
73. Cunha BA. Pharmacokinetic considerations regarding tigecycline for multidrug-resistant (MDR) *Klebsiella pneumoniae* or MDR *Acinetobacter baumannii* urosepsis. *J Clin Microbiol* 2009; 47: 1613.
74. Endimiani A, Patel G, Hujer KM *et al.* *In vitro* activity of fosfomycin against *bla*KPC containing *Klebsiella pneumoniae* isolates, including those non susceptible to tigecycline and/or colistin. *Antimicrob Agents Chemother* 2010; 54: 526–529.
75. Falagas ME, Maraki S, Karageorgopoulos DE, Kastoris AC, Mavromanolakis E, Samonis G. Antimicrobial susceptibility of multidrug-

resistant (MDR) and extensively drug-resistant (XDR) *Enterobacteriaceae* isolates to fosfomycin. *Int J Antimicrob Agents* 2010; 35: 240–243.
76. Michalopoulos A, Virtzili S, Rafailidis P, Chalevelakis G, Damala M, Falagas ME. Intravenous fosfomycin for the treatment of nosocomial infections caused by carbapenem-resistant *Klebsiella pneumoniae* in critically ill patients: a prospective evaluation. *Clin Microbiol Infect* 2010; 16: 184–186.
77. Karageorgopoulos DE, Miriagou V, Tzouvelekis LS, Spyridopoulou K, Daikos GL. Emergence of resistance to fosfomycin used as adjunct therapy in KPC *Klebsiella pneumoniae* bacteraemia: report of three cases. *J Antimicrob Chemother* 2012; 67: 2777–2779.
78. Tamma PD, Cosgrove SE, Maragakis LL. Combination therapy for treatment of infections with gram-negative bacteria. *Clin Microbiol Rev* 2012; 25: 450–470.
79. McCabe MR, Jackson GG. Gram-negative bacteremia I. Etiology and ecology. *Arch Intern Med* 1962; 110: 847–855.