Journal of Microbiology, Immunology and Infection 55 (2022) 1219—1228

Available online at www.sciencedirect.com

ScienceDirect

journal homepage: www.e-jmii.com

Original Article

Antimicrobial treatment of monomicrobial phenotypic carbapenem-resistant *Klebsiella pneumoniae* bacteremia: Two are better than one

Wen-Chia Tsai [a], Ling-Shan Syue [a,b], Wen-Chien Ko [a,b,c], Ching-Lung Lo [a,b,*], Nan-Yao Lee [a,b,c,]**

[a] *Division of Infectious Diseases, Department of Internal Medicine and Tainan, Taiwan*
[b] *Center for Infection Control, National Cheng Kung University Hospital, Tainan, Taiwan*
[c] *Department of Medicine, College of Medicine, National Cheng Kung University, Tainan, Taiwan*

Received 25 April 2021; received in revised form 25 August 2021; accepted 4 September 2021
Available online 1 October 2021

KEYWORDS
Carbapenem-resistant;
Klebsiella pneumoniae;
Phenotype;
Bacteremia;
Combination therapy;
MICs

Abstract *Backgrounds:* Infections caused by carbapenem-resistant *Klebsiella pneumoniae* (CRKP) are emerging worldwide. The optimal treatment for CRKP infections is challenging for clinicians because therapeutic agents are greatly limited.
Material and methods: A retrospective study of CRKP monomicrobial bacteremia was conducted at a medical center between 2010 and 2016. The use of at least one or more drugs with *in vitro* activity against the blood isolates was defined as appropriate combination therapy. The logistic regression model and propensity score analysis was used to assess clinical effects of therapeutic strategies. The 30-day crude mortality was the primary end point.
Results: Two hundred and three patients were eligible and the 30-day mortality rate was 37.9% (77 patients). As compared with monotherapy, empirical (11.6 vs. 57.3%, $p < .001$) or definitive (26.5% vs. 48.6%, $p = .001$) combination antibiotic therapy showed a lower 30-day mortality rate independently. The propensity score analysis showed that those receiving combination therapy had less clinical ($p \leq .001$) or microbiological failure ($p = .003$) and a lower 30-day mortality rate ($p < .001$). Among various regimens of definitive therapy, the 30-day mortality rate was the lowest among patients with appropriate combination therapy 23.6%, ($p < .001$; by log rank test). The primary outcome was similar in those with definitive carbapenem-

* Corresponding author. Department of Internal Medicine, National Cheng Kung University Hospital, No. 138, Sheng Li Road, 704, Tainan, Taiwan. Fax: +886 6 2752038.
** Corresponding author. Department of Internal Medicine, National Cheng Kung University Hospital, No. 138, Sheng Li Road, 704, Tainan, Taiwan. Fax: +886 6 2752038.
E-mail addresses: raycigol@gmail.com (C.-L. Lo), nanyao@mail.ncku.edu.tw (N.-Y. Lee).

https://doi.org/10.1016/j.jmii.2021.09.002

containing and carbapenem-sparing combination regimens (p = .81). The presence or absence of carbapenemase production did not affect the mortality rate (p = .26).
Conclusion: Combination therapy, regardless of carbapenem-containing or carbapenem-sparing regimens, was associated with a favorable outcome.

Introduction

Multidrug resistance among Enterobacterales is a growing public health crisis that makes many healthcare-associated infections difficult to treat with current antibiotics.[1–3] The decreasing activity of third-generation cephalosporin and fluoroquinolones has led to explore the use of carbapenem for common healthcare-associated infections, extending pressure for the emergence of carbapenem-resistant Enterobacterales (CRE), particularly *Klebsiella pneumoniae*.[3,4]

Over the past decades, carbapenem-resistant *K. pneumoniae* (CRKP) related to the overexpression of carbapenem-hydrolyzing beta-lactamases, such as *K. pneumoniae* carbapenemase (KPC), has emerged as a global threat.[5–7] These pathogens often carry antibiotic-resistant genes in addition to beta-lactamases.[6,7] Bloodstream infections (BSIs) caused by carbapenem-resistant isolates are associated with higher mortality rates (40%—70%) compared to carbapenem-susceptible isolates (20%—30%) respectively.[7–10]

The therapeutic options for patients infected by CRKP were based on case reports, case series, retrospective and observational studies, but the optimal options are presently unknown.[9,11–15] Previous retrospective studies demonstrated patients receiving combination therapy were more likely to survive than those with monotherapy, regardless of carbapenemase production.[16–20] However, a little percentage of carbapenemase-producing isolates may be retained phenotypic susceptibility with lower minimum inhibitory concentrations (MICs) of carbapenems, in despite of molecular confirmation of the carriage of carbapenemase genes.[21] Conflict of resistance gene and phenotypic susceptibility which raises the question of clinical application of carbapenems.

New generation of anti-CRE agents (such as ceftazidime-avibactam, cefiderocol, meropenem-vaborbactam, imipenem-relebactam, plazomicin, and eravacycline) have shown to improve outcomes for CRE infections,[22–26] but most of such agents are not usually available worldwide. Two of our previous studies focusing on the BSIs with non-carbapenemase producing CRKP[20] or cefepime-susceptible CRKP[27] showed the protective effect of combination therapy. This study aimed to assess the clinical outcome, predictors of mortality, and emphasized therapeutic strategy with current available agents for CRKP bacteremia in the real world, regardless of the mechanism and phenotype of carbapenem resistance.

Materials and methods

Study population and data collection

The microbiology database at National Cheng Kung University Hospital (NCKUH) in southern Taiwan was reviewed for the cases of *K. pneumoniae* bacteremia between August 2010 and August 2016. If a patient experienced more than one bacteremic episode, only the first episode was included. The study was approved by the NCKUH Institutional Review Board (ER-100-182). Adults (age, $\geq$ 18 years) were included with fulfilling the following criteria: bacteremia in combination with sepsis and parenteral antibiotic treatment administrated for at least 48 h before the end of antibiotic therapy or death, and with sufficient doses as recommended by the Clinical and Laboratory Standards Institute (CLSI).[28] Patients with polymicrobial bacteremia were excluded.

The empirical therapy group included the patients who received combination therapy or monotherapy, of which the first dose was administered within the first 24 h after blood cultures had been taken. Empirical antimicrobial therapy was defined as administered within 72 h after bacteremia onset, and that administered afterward as definitive therapy. Also, definitive antimicrobial therapy was defined as antibiotic that was continued or initiated on the day that the drug susceptibility test results were reported, and that was started no later than 5 days after the index positive blood sample for culture had been drawn. Appropriate therapy was indicated when the pathogen was *in vitro* susceptible to one of the prescribed drugs. The prescription of antibiotic agents, indication, and dosage would be approved by the antimicrobial stewardship team in the study hospital.

Microbiology and antimicrobial susceptibilities

We used the VITEK 2 system (bioMérieux, Marcy l'Etoile, France) for species identification and antibiotic susceptibility testing. MICs of antibiotics were determined by the broth microdilution method and interpreted according to the CLSI breakpoints,[28] but MICs of colistin and tigecycline were interpreted by the breakpoints established by the European Committee on Antimicrobial Susceptibility Testing.[29] Carbapenem resistance was indicated as *in vitro* resistance to at least one of ertapenem, imipenem, or meropenem.

Molecular detection of ESBL, AmpC, and carbapenemase genes

We performed multiplex polymerase chain (PCR) with whole genomic DNA extracted by the QIAamp DNA minikit and a QIAcube instrument (Qiagen, Valencia, CA) from overnight colonies grown on agar (Remel, Lenexa, KS)[30] to detect AmpC (bla_{CMY}, bla_{DHA}, bla_{FOX}, bla_{MOX}, bla_{ACC}, bla_{MIR}, and bla_{ACT}), ESBL (bla_{TEM}, bla_{SHV}, and bla_{CTX-M}), and carbapenemase (bla_{KPC}, bla_{VIM}, bla_{IMP}, bla_{NDM}, and bla_{OXA}) genes.

Clinical evaluation and outcomes

Demographic data was retrieved using standard record form from medical charts. Bacteremia was defined as the isolation of the organisms in at least one blood culture with a compatible clinical syndrome. Patients receiving antimicrobial therapy for more than 48 h were enrolled for the assessment of outcome. The primary outcome measure was crude 30-day mortality, and secondary outcomes included total hospital length of stay (LOS) after bacteremia onset, clinical and microbiological failure during hospitalization. Immunosuppression was defined as the following; the receipt of corticosteroid (at least 10 mg prednisolone or an equivalent daily dosage) for more than two weeks, or of antineoplastic chemotherapy or antirejection medication within four weeks before the onset of bacteremia. The severity of underlying illness was stratified as being rapidly fatal, ultimately fatal, or nonfatal.[31] The severity of bacteremia was graded using the Pitt bacteremia score on the day of bacteremia onset.[32] Clinical failure was defined as the following: for at least five days, initial antimicrobial therapy failed to resolve sepsis symptoms or signs, or a fatal outcome ensued. The detection of *K. pneumoniae* blood isolate after antimicrobial therapy for at least 72 h was regarded as microbiological failure.

Statistical analyses

Data were analyzed using the SPSS software for Windows, version 20.0. Continuous variables were expressed as mean values ± standard deviations (SDs) and compared by the Mann—Whitney U test or Student *t*-test. Categorical variables were expressed as the percentages of total numbers of patients and compared by the Fisher exact test or χ^2 test. Independent predictors for 30-day mortality were identified using logistic regression analysis. Variables with a *P* value of 0.1 or less in the univariate analysis were included in the multiple conditional logistic regression analysis, including the following items: age, pneumonia, Pitt bacteremia score (≥4 points), rapidly fatal underlying disease, colistin-base therapy, carbapenemase-producing isolates, appropriate definitive therapy, combinational definitive therapy. Kaplan—Meier survival curves were compared by the log-rank test and a Cox proportional hazard model was using for the survival analysis, adjusted for confounding variables. A *P* value less than 0.05 was considered statistically significant, and all tests were two-tailed.

Because of the differences in baseline characteristics of the retrospective design, the propensity score matching method was applied to minimize the differences between monotherapy and combination therapy groups. Propensity scores were calculated through a multivariate logistic regression model in which the dependent variable was a binary indicator of combination therapy or monotherapy. The covariates to generate the propensity score included age, gender (male), comorbidity conditions (hepatic cirrhosis, diabetes mellitus, end-stage renal disease under dialysis, congestive heart failure with an ejection fraction less than 45%, chronic obstructive pulmonary disease or immunocompromising conditions), Pitt bacteremia score, intensive care unit stay on Day 1 of bacteremia, and source of bacteremia. The 1:1 nearest neighbor matching without replacement was performed with a caliper width of 0.20. Standardized mean biases were tested to ensure balance after propensity score matching between the monotherapy and combination therapy groups.

Results

Patient population

In the study period, overall 293 patients had experienced CRKP bacteremia and 203 met the inclusion criteria for the analyses of microbiological and clinical outcome (Fig. 1). Most (201, 99.0%) of bacteremic episodes were hospital-onset. Males constituted for 63.5% (129 patients) and all had one or more co-morbidity. The median age was 71 (interquartile range [IQR]: 58—80) years. The median duration of hospitalization before the onset of bacteremia was 15 days (IQR: 5—41 days). Vascular catheter-related infections (66 patients, 32.5%) and pneumonia (49, 24.1%) were the major sources of bacteremia, followed by primary (47, 23.2%), urinary tract infections (29, 14.3%), skin-soft tissue infections (19, 9.4%) and intra-abdominal infections (6, 3.0%).

Antimicrobial susceptibilities, measured by MIC values, of these CRKP isolates were shown in Table 1. The vast majority (92.6%) were susceptible to colistin. The susceptible rate for ertapenem, imipenem, and meropenem was 9.4%, 60.6%, and 67.5%, respectively. Of note, 147 (72.4%) isolates showed discordant susceptibilities, *i.e.*, an isolate was resistant to one carbapenem but susceptible to the others. 108 (53.2%) isolates were ertapenem-resistant and meropenem/imipenem-susceptible, 19 (9.4%) imipenem-resistant and meropenem/ertapenem-susceptible, 16 (7.9%) meropenem/ertapenem-resistant and imipenem-susceptible, and 11 (5.4%) imipenem/ertapenem-resistant and meropenem-susceptible. In this study, the genes encoding three ESBLs (CTX-M, SHV, and TEM), two AmpC beta-lactamases (CMY and DHA), and three carbapenemases (KPC, IMP, and OXA) were found in CRKP blood isolates. The number of isolates of KPC, IMP, and OXA-48 like beta-lactamase was 1, 42, and 9, respectively.

Meropenem MICs of 203 isolates ranged from 0.25 to ≥32 μg/mL; MIC50 and MIC_{90} were 0.5 and 16 μg/mL respectively. Patients were stratified into four categories by meropenem MIC value: highly susceptible (<0.5 μg/mL), susceptible (0.5—1 μg/mL), intermediate to resistant (2—8 μg/mL), and highly resistant (>8 μg/mL). 30-day ($p < .001$) and in-hospital ($p < .001$) mortality rate

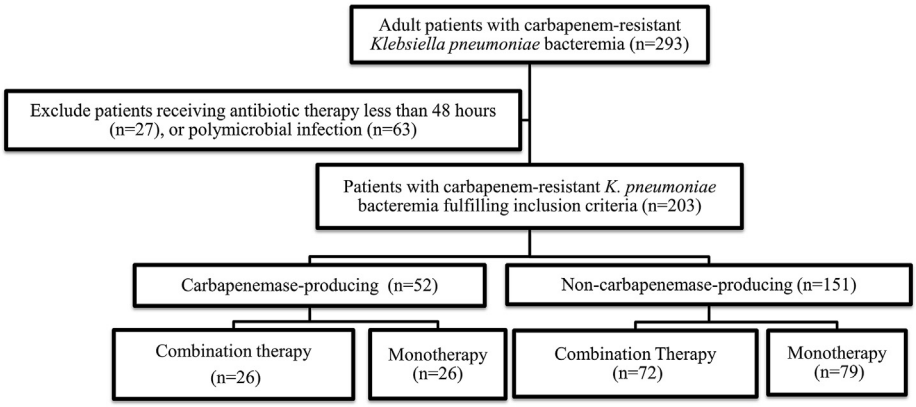

Figure 1. Flow diagram of study.

Table 1 *In vitro* susceptibilities and minimum inhibitory concentrations (MICs) of 203 blood isolates of carbapenem-resistant *Klebsiella pneumoniae*.

Antimicrobial agents	MIC (μg/mL)		Numbers of susceptible isolates (%)
	MIC_{50}/MIC_{90}	Range	
Ertapenem	2/32	0.5 - > 32	19 (9.4)
Imipenem	1/16	0.25 - > 32	123 (60.6)
Meropenem	0.5/16	0.25 - > 32	137 (67.5)
Amikacin	4/64	1 - > 64	144 (70.9)
Cefotaxime	32/128	1 - > 128	2 (0.98)
Cefepime[a]	8/32	0.5 - > 32	129 (63.5)
Piperacilin-tazobactam	32/128	4 - > 128	44 (21.7)
Ciprofloxacin	8/32	0.06 - > 16	9 (4.4)
Colistin[b]	0.5/1	0.125—32	188 (92.6)
Tigecycline[b]	0.5/3	0.25—8	106 (52.2)

[a] Cefepime-susceptible isolates include the susceptible and suceptible-dose- dependent categories, according to the CLSI criteria.
[b] *In vitro* susceptibilities and MIC are interpreted according to the breakpoints recommended by the EUCAST in 2021, and otherwise by the CLSI criteria.

increased as meropenem MIC increased (linear-by-linear association).

One hundred patients died during hospitalization, and resulting in an in-hospital mortality rate of 49.2%, and 30-day crude mortality rate 37.9% (77). In a multivariate logistic regression analysis, 30-day mortality was independently associated with a critical illness (Pitt bacteremia score ≥4 points at bacteremia onset, adjusted odds ratio [aOR]: 8.04; 95% confidence interval [CI]: 3.69—17.53; $P < .001$), pneumonia (aOR 5.48; 95% CI 2.34—12.84; $p < .001$), a rapidly fatal underlying disease (aOR 3.74; 95% CI 1.24—11.31; $p = .02$), appropriate definitive antimicrobial therapy (aOR 0.11; 95% CI 0.03—0.42; $p = .001$), and combination regimens (aOR 0.19; 95% CI 0.08—0.42; $p < .001$; Table 2).

The odds ratio of 30-day mortality of individuals infected by susceptible isolates (MIC 0.5—1 μg/mL) was 10.35 (95% CI 2.47—43.4; $p < .001$), intermediate to resistant isolates (2—8 μg/mL) 11.31 (95% CI 2.61—48.92; $p = .001$), and highly resistant isolates (>8 μg/mL) 14.1 (95% CI 3.22—61.51; $p = .001$), compared with those infected by highly susceptible isolates (<0.5 μg/mL) as the reference in the Cox regression model after adjustment of confounding variables.

Empirical antibiotic therapy

According to the study criteria, there were 88 patients receiving appropriate empirical therapy and 177 appropriate definitive therapy (Fig. 1). Patients with empirical combination therapy showed a lower 30-day mortality rate (10/86, 11.6% *vs.* 67/117, 57.3%; $p < .001$) or clinical failure rate (9/86, 10.5% *vs.* 59/117, 50.4%; $p < .001$) than those with empirical monotherapy, regardless of appropriateness, and more often received appropriate therapy (54/86, 62.8% *vs.* 34/117, 29.1%; $p < .001$). In the colistin-containing group, the mortality was lower than that of the colistin-sparing group, though the difference is not statistically significant.

Among patients who received carbapenem-containing empirical therapy, the 30-day mortality rate increased from 0% in the causative isolates with meropenem MIC of <0.5 μg/mL to 64.3% with MIC of >8 μg/mL ($p < .001$, linear-by-linear association). Similarly, among patients who

Table 2 Multivariate logistic regression analysis of risk factors of 30-day crude mortality among 203 adults with monomicrobial carbapenem-resistant *Klebsiella pneumoniae* bacteremia.

Variables	Survivors (n = 126)	Non-survivors (n = 77)	Univariate analysis		Multivariate analysis	
			OR (95% CI)	*P* values	OR (95% CI)	*P* values
Age; median (IQR), years	70 (55–77)	73 (63–82)	–	0.03	1.01 (0.98–1.03)	0.672
Pneumonia	18 (14.3)	31 (40.3)	4.03 (2.06–7.95)	<0.001	5.48 (2.34–12.84)	<0.001
Pitt bacteremia score ≥4 points	10 (7.9)	16 (20.8)	3.04 (1.30–7.11)	0.01	8.04 (3.69–17.53)	<0.001
Rapidly fatal underlying disease	4 (4.5)	12 (20.7)	5.48 (1.67–17.96)	0.005	3.74 (1.24–11.31)	0.02
Appropriate definitive therapy	119 (94.45)	57 (74.0)	0.21 (0.10–0.48)	<0.001	0.11 (0.03–0.42)	0.001
Combinational definitive therapy	73 (53.9)	26 (32.5)	0.35 (0.19–0.63)	0.001	0.19 (0.08–0.42)	<0.001

Data are given as number (percentage) unless otherwise specified. Ellipses indicate not available.
OR indicates odds ratio, CI confidence interval, and SD standard deviation.

received carbapenem-containing definitive therapy, the mortality rate raised from 3.2% in the isolates with meropenem MIC of <0.5 μg/mL to 84.6% with MIC of >8 μg/mL ($p < .001$, linear-by-linear association).

Definitive antibiotic therapy

The characteristics of patients with definitive monotherapy (n = 105) or combination therapy (n = 98) were summarized at Table 3. There were no significant differences in terms of age, sex, comorbidity, source of bacteremia, or disease severity. The 30-day mortality rate was higher in those with monotherapy than those with combination therapy (51/105, 48.6% *vs.* 26/98, 26.5%; $p = .001$, stander deviation [SD] = −0.22). Also, there was a higher clinical failure rate (48/105, 45.7% *vs.* 20/98, 20.4%; $p < .001$, SD = −0.25), microbiological failure rate (24/105, 22.9% *vs.* 6/98, 6.1%; $p = .001$, SD = −0.17), and longer hospital stay (median, 20 days *vs.* 17 days; $p = .003$, SD = 1.17) among definitive monotherapy than combination therapy. The Kaplan–Meier survival analysis also favored combination therapy ($p < .001$).

In the propensity score match analysis, 92 pairs of patients with monotherapy and those with combination therapy could be matched based on the basis of the propensity score with the ratio of 1:1. The standardized mean differences of co-variables were >10% before matching and became <10% after matching. After adjustment for confounding factors, combination therapy remained to be significantly associated with less clinical (18.5% *vs.* 43.7%, $p < .001$, SD = −0.25) and microbiological (6.5% *vs.* 22.8%, $p = .003$, SD = −0.17) failure, and 30-day mortality (23.9% *vs.* 50.0%, $p < .001$, SD = −0.22) than those received monotherapy.

Within various regimens for definitive therapy (appendix A), the 30-day mortality rate was the lowest in patients with appropriate (including one active agent *in vitro* at least) combinations therapy (21/89, 23.6%), followed by appropriate monotherapy (36/88, 40.9%), inappropriate combination therapy (5/9, 55.6%), and inappropriate monotherapy (15/17, 88.2%), as revealed by the Kaplan–Meier survival analysis in which most favored appropriate combination therapy ($p < .001$, by log rank test; Fig. 2). The odds ratio of 30-day mortality of adults with appropriate combinations therapy was 0.09 (95% CI 0.04–0.12; $p < .001$), appropriate monotherapy 0.27 (95% CI, 0.14–0.54; $p < .001$), inappropriate combinations therapy 0.33 (95% CI, 0.12–0.49; $p = .038$), as compared with inappropriate monotherapy in the Cox regression model after adjustment of confounding variables. In the subgroup analysis with the Cox regression model after adjustment of three confounding variables (*i.e.*, a critical illness, pneumonia, and a rapidly fatal underlying disease), the odds ratio of 30-day mortality of those treated by combination therapy with two active agents was 0.06 (95% CI: 0.008–0.42; $p = .005$), combination therapy with one active agent 0.31 (95% CI: 0.13–0.52; $p < .001$) as compared with that of appropriate monotherapy. Of 89 patients definitively treated by appropriate combination therapy, the 30-day mortality rate was lower among those with two active agents than one active agent (1/19, 5.3% *vs.* 20/70, 28.6%; $p = .036$).

Among those with definitive therapy, either carbapenem-containing (19/68, 27.9% *vs.* 29/67, 48.3%; $p = .028$) or carbapenem-sparing (7/30, 23.3% *vs.* 22/45, 48.9%; $p = .03$) combination therapy fared better than monotherapy (Fig. 3). The 30-day mortality rate of those with carbapenem-containing regimens was similar to that of carbapenem-sparing among those with definitive combination therapy, (19/68, 27.9% *vs.* 7/30, 23.3%; $p = .81$). Of note, among those with definitive carbapenem-containing combination therapy, the 30-day mortality rate raised from 0% in the causative isolates with meropenem MIC <0.5 μg/mL to 77.8% in those of MIC >8 μg/mL ($p < .001$, linear-by-linear association, Fig. 4), and was lower than that among adults with monotherapy in corresponding MIC categories. In other words, carbapenem-containing combination therapy showed a better prognosis than carbapenem monotherapy for the isolates of MIC 0.5–8 μg/mL.

Discussion

The potential synergistic or additive effects of combinations of certain antimicrobials has been explored for the management of infections caused by multidrug-resistant organisms.[33] The current study aimed at the patients with BSIs caused by CRKP with different mechanisms of resistance and demonstrated patients received monotherapy had higher mortality, worse clinical and microbiological

Table 3 Characteristics of 203 adults with monomicrobial carbapenem-resistant *Klebsiella pneumoniae* bacteremia appropriately and definitively treated.

Characteristics	Crude analysis			Propensity score matched analysis		
	Monotherapy group, n = 105	Combination therapy group, n = 98	Standard deviation	Monotherapy group, n = 92	Combination therapy group, n = 92	Standard deviation
Age, median (IQR), year	71 (56.5–80.0)	71 (60.8–78.3)	0.19	71 (54–79)	70 (58–78)	0.09
Gender, male	66 (62.9)	63 (64.3)	0.01	59 (64.1)	59 (64.1)	0.01
Length of hospital before bacteremia, median (IQR), day	20 (4.5–44.0)	14 (4.75–35.0)	−2.18	21 (4–44)	14 (4–34)	−2.2
Comorbidity						
Diabetes mellitus	63 (60.0)	60 (61.2)	0.2	55 (59.8)	56 (60.9)	0.1
Chronic kidney disease	36 (34.3)	32 (32.7)	−0.1	32 (34.8)	31 (33.7)	−0.01
Malignancy	33 (31.4)	27 (27.6)	−0.4	30 (32.6)	24 (26.1)	0.08
Liver cirrhosis	19 (18.3)	10 (10.2)	−0.08	15 (16.3)	8 (8.7)	0.07
Severity of underlying disease (McCabe classification)						
Rapidly fatal	12 (11.4)	14 (14.3)	0.02	12 (13.0)	10 (10.9)	0.02
Pitt bacteremia score, ≥4 points	48 (45.7)	47 (48.4)	0.03	42 (45.7)	42 (45.7)	0.001
Source of bacteremia						
Vascular catheter-related infection	31 (29.5)	35 (31.7)	0.06	29 (31.5)	35 (38.0)	0.06
Primary bacteremia	27 (25.7)	20 (20.4)	−0.5	23 (25.0)	20 (21.7)	0.05
Intra-abdominal infection	4 (3.8)	2 (2.0)	−0.2	3 (3.3)	2 (3.2)	−0.02
Pneumonia	21 (20.0)	28 (28.6)	0.9	21 (22.8)	22 (23.9)	0.01
Skin and soft-tissue infection	9 (8.6)	10 (10.2)	0.02	9 (9.8)	10 (10.9)	0.02
Urosepsis	17 (16.2)	12 (12.2)	−0.04	11 (12.0)	12 (13.0)	−0.04
Hospital stay of survivors, median (IQR), day	20 (13.5–29.5)	17 (12.0–22.0)	1.17	16 (10–19)	15 (10–18)	0.12
Carbapenemase producing isolates	26 (24.8)	26 (26.5)	0.2	26 (28.3)	26 (28.3)	0.02
Appropriate definitive therapy	88 (83.8)	89 (90.8)	0.07	81 (88.0)	84 (91.3)	0.07
Clinical failure	48 (45.7)	20 (20.4)	−0.25	43 (43.7)	17 (18.5)	−0.25
Microbiological failure	24 (22.9)	6 (6.1)	−0.17	21 (22.8)	6 (6.5)	−0.17
30-day mortality	51 (48.6)	26 (26.5)	−0.22	46 (50,0)	22 (23.9)	−0.22
Crude mortality	56 (53.3)	43 (43.9)	−0.1	50 (54.3)	38 (41.3)	−0.1

Data are given as numbers (percentages), unless otherwise specified.
SD indicates standard deviation; IQR, interquartile range, OAA other active agent.

outcomes than combination therapy regardless empirical or definitive therapy. Moreover, the risk of mortality increased when meropenem MIC of the causative isolates raised.

The resistance of carbapenems of our CRKP isolates was major focus on ertapenem other than imipenem or meropenem those was similar to the previous surveillance studies.[34,35] The mechanisms of carbapenem resistance in *K. pneumoniae* are variable, which may involve with or without hyper-production of AmpC beta-lactamases or ESBLs, production of carbapenemases, modification in the outer membrane permeability, up-regulation of efflux pumps, or in combination.[5,35,36] It raises clinical concerns in the determination of precise resistance mechanisms, which would not be necessary in a MIC-based therapeutic approach.[10,37–39] The perform confirmatory tests for KPC or other carbapenemases was not a routine practice based on current breakpoint met the criteria of the probability of pharmacodynamic target attainment,[10] unless for epidemiological or infection control purposes.[28] Furthermore, the commercial tools of carbapenemases detection are not world widely available, especially in our country. Our result showed that appropriate combination therapy is adequate for clinical management whether carbapenemases detection is done or not.

Although CPKP isolates are defined as being resistant to one of the carbapenems, some have relatively low carbapenem MICs, raising the question of their therapeutic potential against CPKP infections.[40] Most of our patients with carbapenem therapy experiencing clinical failure were infected by the isolates with a higher meropenem MIC

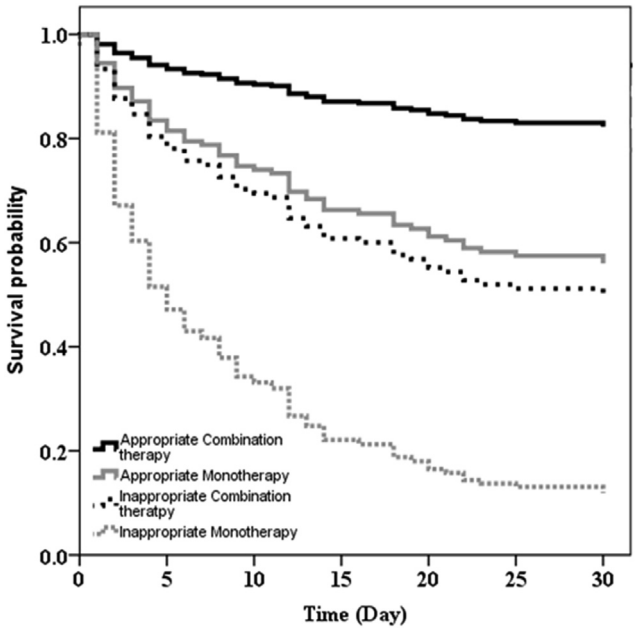

Figure 2. Survival analysis curves according to different definitive therapeutic approaches. Footnote: Inappropriate monotherapy (n = 17; gray dot line); inappropriate combination therapy (n = 9; black dot line); appropriate monotherapy (n = 88; gray solid line); appropriate combination therapy (n = 89; black solid line) ($p < .001$ by log rank test).

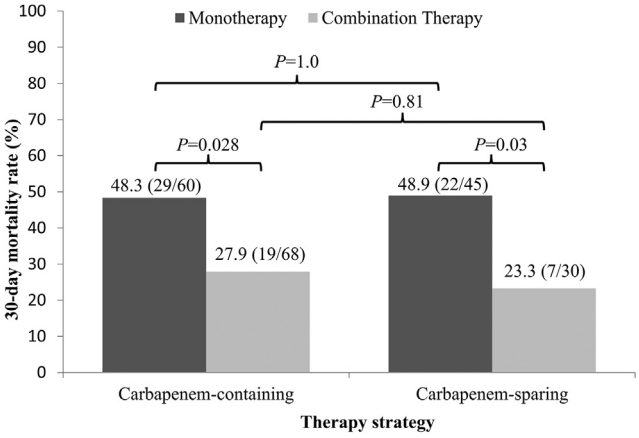

Figure 3. The 30-day mortality rates stratified by definitive carbapenem-containing or -sparing monotherapy or combination therapy.

(>0.5 μg/mL). Our works and other studies assessing patient outcomes based upon MICs suggest the need to reassess current susceptibility breakpoints for carbapenems.[10,21]

Colistin showed excellent *in vitro* activity in our study. Though the mortality among those with colistin monotherapy was high-which as previous reports,[33,41] the mortality rate of the patients treated by colistin-containing combination therapy is lower than that of those treated by colistin-sparing regimens, although not statistically significant. Previous data also demonstrated better clinical outcome of colistin-containing combination therapy than colistin monotherapy in multidrug-resistant *K. pneumoniae*-associated hospital-acquired pneumonia and ventilator-associated pneumonia.[42] According to the recent CLSI criteria, the colistin breakpoint of Enterobacterales is changed from "susceptible" to "intermediate" when the MIC is no more than 2 μg/mL. The revision of breakpoint interpretation may account for that colistin is not suitable as monotherapy; however, it may be beneficial when used with another effective agent. Currently, no randomized controlled trials comparing combination therapy with monotherapy for patients with CRE infections are available.[33] Nonetheless, the superiority of combination therapy (at least two agents *in vitro* active against the causative pathogen) over monotherapy (one *in vitro* active agent) was supported by many data, in terms of patient survival for critical illness caused by CRE infections.[17,43]

The 30-days crude mortality was independently associated with presence of rapid fatal underlying disease, bacteremic pneumonia, critical illness, and appropriateness of antibiotic therapy those were in accordance with previous researches.[17,18,27,44,45] The last factor, the only modifiable one, was the appropriateness of antibiotic therapy. Our analysis may be unique from previous studies in that the survival impact of empirical and definitive antimicrobial agents was correlated with meropenem MICs. The previous studies show carbapenem—base combination therapy with aminoglycoside, colistin, high-dose tigecycline, or fosfomycin were conferred better clinical outcomes for those infected by the carbapenem-resistant isolate.[46,47] Based on the meropenem MIC category, carbapenem-based combination therapy with another *in vitro* active agent provides survival benefit, even within the susceptible category of meropenem (0.5—1 μg/mL). However, limited data are evaluating for the suitable breakpoints of carbapenem-based combinations to predict a better outcome.[10] Our data demonstrated those infected by the isolates of meropenem MIC ≤1 μg/mL and treated by meropenem-containing combination therapy fared well as carbapenem-sparing therapy.

There were several limitations in our work. First, most patients were elderly, and had more than one comorbidity which increased the risk of all-cause mortality. Nevertheless, the multivariable regression analysis correcting for source of infection, age, comorbidities, and disease severity showed no significant impact among different therapeutic strategy. Second, the CRKP isolates were collected through phenotypic MIC values, and absence of carbapenemases confirm test. However, our study purposed to investigate the amendable variables correlating with the outcome of phenotypic CRKP bacteremia. Our study demonstrated clinical evidence that promoted the MIC-based therapeutic approach, regardless of resistance mechanisms.[39] Among the types of carbapenemases, KPC is not the major one in our study, which may affect the applicability of our study result in the KPC-prevailing regions.[48] Due to the limited number of carbapenemase-producing isolates, it was difficult to reveal the impact of different carbapenemases on clinical outcome. Finally, the study was focus on the hospitalization period, the 30-days crude mortality assessment was the endpoint of the study, and the long-term outcome remained undefined.

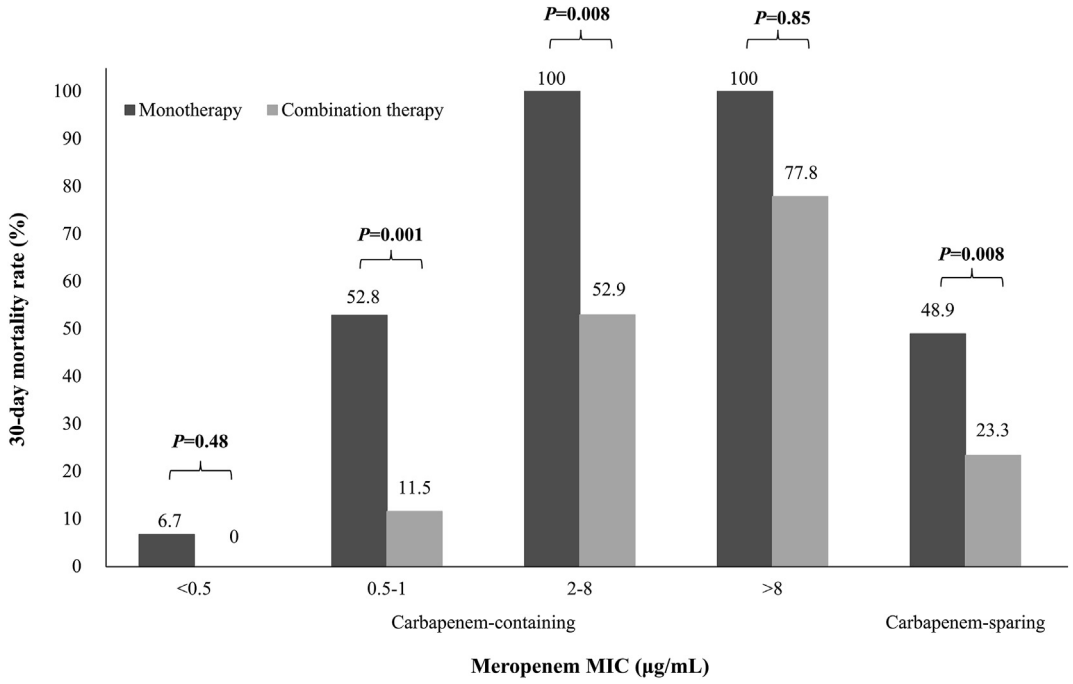

Figure 4. The 30-day mortality rates stratified by meropenem MICs and therapeutic strategies.

Despite the above limitations, this study includes a large patient and can provide useful information to optimize therapeutic approaches for the treatment.

Conclusion

Our data illustrate unfavorable outcomes in patients with phenotypic CRKP bacteremia as meropenem MICs raise. Carbapenem-containing combination therapy was preferred for the causative isolates with a meropenem MIC of <8 μg/mL. Combination therapy regardless of carbapenem-based or carbapenem-sparing with two or more effective drugs, will be more effective than monotherapy whether carbapenemases present or not.

Financial support

This study was supported by the grants from the Ministry of Science and Technology, Taiwan (MOST 109-2327-B-006-005-) and National Cheng Kung University Hospital, Tainan, Taiwan (NCKUH-11002055).

Transparency declarations

None to declare.

Author contributions

W.C.T, N.Y.L and W.C.K. conceived the study. W.C.T, L.C.L and S.L.S provided data collection, statistical and analytic support. W.C.T and N.Y.L performed the laboratory work. W.C.T, N.Y.L and W.C.K analyzed the data. W.C.T prepared the manuscript. All authors reviewed and edited the manuscript.

Declaration of competing interest

All authors: no conflicts.

References

1. Peleg AY, Hooper DC. Hospital-acquired Infections due to gram-negative bacteria. *N Engl J Med* 2010;**362**:1804—13.
2. Munoz-Price LS, Poirel L, Bonomo RA, Schwaber MJ, Daikos GL, Cormican M, et al. Clinical epidemiology of the global expansion of *Klebsiella pneumoniae* carbapenemases. *Lancet Infect Dis* 2013;**13**:785—96.
3. Girometti N, Lewis RE, Giannella M, Ambretti S, Bartoletti M, Tedeschi S, et al. *Klebsiella pneumoniae* bloodstream infection: epidemiology and impact of inappropriate empirical therapy. *Medicine (Baltim)* 2014;**93**:298—309.
4. Paterson DL. Resistance in gram-negative bacteria: Enterobacteriaceae. *Am J Med* 2006;**119**:S20—8.
5. Schwaber MJ, Carmeli Y. Carbapenem-resistant Enterobacteriaceae: a potential threat. *J Am Med Assoc* 2008;**300**:2911.
6. Nordmann P, Cuzon G, Naas T. The real threat of *Klebsiella pneumoniae* carbapenemase-producing bacteria. *Lancet Infect Dis* 2009;**9**:228—36.
7. Gomez Simmonds A, Nelson B, Eiras DP, Loo A, Jenkins SG, Whittier S, et al. Combination regimens for treatment of carbapenem-resistant *Klebsiella pneumoniae* bloodstream infections. *Antimicrob Agents Chemother* 2016;**60**:3601—7.
8. Schwaber MJ, Carmeli Y. Mortality and delay in effective therapy associated with extended-spectrum beta-lactamase production in Enterobacteriaceae bacteraemia: a systematic review and meta-analysis. *J Antimicrob Chemother* 2007;**60**:913—20.

9. Ben-David D, Kordevani R, Keller N, Tal I, Marzel A, Gal Mor O, et al. Outcome of carbapenem resistant *Klebsiella pneumoniae* bloodstream infections. *Clin Microbiol Infect* 2012;**18**:54—60.
10. Patel TS, Nagel JL. Clinical outcomes of Enterobacteriaceae infections stratified by carbapenem MICs. *J Clin Microbiol* 2015;**53**:201—5.
11. 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—404.
12. Falagas ME, Lourida P, Poulikakos P, Rafailidis PI, Tansarli GS. Antibiotic treatment of infections due to carbapenem-resistant Enterobacteriaceae: systematic evaluation of the available evidence. *Antimicrob Agents Chemother* 2014;**58**:654—63.
13. Tang HJ, Lai CC, Chen CC, Zhang CC, Weng TC, Chiu YH, et al. Colistin-sparing regimens against *Klebsiella pneumoniae* carbapenemase-producing *K. pneumoniae* isolates: combination of tigecycline or doxycycline and gentamicin or amikacin. *J Microbiol Immunol Infect* 2019;**52**:273—81.
14. Li J, Bi W, Dong G, Zhang Y, Wu Q, Dong T, et al. The new perspective of old antibiotic: *In vitro* antibacterial activity of TMP-SMZ against *Klebsiella pneumoniae*. *J Microbiol Immunol Infect* 2020;**53**:757—65.
15. Yang TY, Lu PL, Tseng SP. Update on fosfomycin-modified genes in Enterobacteriaceae. *J Microbiol Immunol Infect* 2019;**52**:9—21.
16. Qureshi ZA, Paterson DL, Potoski BA, Kilayko MC, Sandovsky G, Sordillo E, et al. Treatment outcome of bacteremia due to KPC-producing *Klebsiella pneumoniae*: superiority of combination antimicrobial regimens. *Antimicrob Agents Chemother* 2012;**56**:2108—13.
17. Zarkotou O, Pournaras S, Tselioti P, Dragoumanos V, Pitiriga V, Ranellou K, 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—803.
18. Tumbarello M, Viale P, Viscoli C, Trecarichi EM, Tumietto F, Marchese A, 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—50.
19. Tumbarello M, Trecarichi EM, De Rosa FG, Giannella M, Giacobbe DR, Bassetti M, et al. Infections caused by KPC-producing *Klebsiella pneumoniae*: differences in therapy and mortality in a multicentre study. *J Antimicrob Chemother* 2015;**70**:2133—43.
20. Lee NY, Tsai CS, Syue LS, Chen PL, Li CW, Li MC, et al. Treatment outcome of bacteremia due to non-carbapenemase-producing carbapenem-resistant *Klebsiella pneumoniae* bacteremia: role of carbapenem combination therapy. *Clin Therapeut* 2020;**42**:e33—44.
21. Esterly JS, Wagner J, McLaughlin MM, Postelnick MJ, Qi C, Scheetz MH. Evaluation of clinical outcomes in patients with bloodstream infections due to Gram-negative bacteria according to carbapenem MIC stratification. *Antimicrob Agents Chemother* 2012;**56**:4885—90.
22. Tumbarello M, Losito AR, Giamarellou H. Optimizing therapy in carbapenem-resistant Enterobacteriaceae infections. *Curr Opin Infect Dis* 2018;**31**:566—77.
23. Jean SS, Gould IM, Lee WS, Hsueh PR, International Society of Antimicrobial Chemotherapy (ISAC). New drugs for multidrug-resistant gram-negative organisms: time for stewardship. *Drugs* 2019;**79**:705—14.
24. Motsch J, Murta de Oliveira C, Stus V, Köksal I, Lyulko O, Boucher HW, et al. RESTORE-IMI 1: a multicenter, randomized, double-blind trial comparing efficacy and safety of imipenem/relebactam vs colistin plus imipenem in patients with imipenem-nonsusceptible bacterial infections. *Clin Infect Dis* 2020;**70**:1799—808.
25. Karaiskos I, Lagou S, Pontikis K, Rapti V, Poulakou G. The 'old' and the 'new' antibiotics for MDR gram-negative pathogens: for whom, when, and how. *Front Public Health* 2019;**7**:151.
26. Sheu CC, Chang YT, Lin SY, Chen YH, Hsueh PR. Infections caused by carbapenem-resistant Enterobacteriaceae: an update on therapeutic options. *Front Microbiol* 2019;**10**:80.
27. Lee NY, Lo CL, Chen PL, Syue LS, Li CW, Li MC, et al. Clinical impact of cefepime breakpoint in patients with carbapenem-resistant *Klebsiella pneumoniae* bacteraemia. *Int J Antimicrob Agents* 2021;**57**:106250.
28. Clinical and Laboratory Standards Institute (CLSI). *Performance standards for antimicrobial susceptibility testing*. 31th ed. Wayne, PA: CLSI; 2021. CLSI supplement M100.
29. The European Committee on Antimicrobial Susceptibility Testing. *Breakpoint tables for interpretation of MICs and zone diameters*. Version 11.0, http://www.eucast.org; 2021.
30. Dallenne C, Da Costa A, Decré D, Favier C, Arlet G. Development of a set of multiplex PCR assays for the detection of genes encoding important beta-lactamases in Enterobacteriaceae. *J Antimicrob Chemother* 2010;**65**:490—5.
31. McCabe WR, Jackson GG. Gram-negative bacteremia: I. etiology and ecology. *Arch Intern Med* 1962;**110**:847—55.
32. Chow JW, Yu VL. Combination antibiotic therapy versus monotherapy for gram-negative bacteraemia: a commentary. *Int J Antimicrob Agents* 1999;**11**:7—12.
33. Rodríguez-Baño J, Gutiérrez-Gutiérrez B, Machuca I, Pascual A. Treatment of infections caused by extended-spectrum-beta-lactamase-, AmpC-, and carbapenemase-producing Enterobacteriaceae. *Clin Microbiol Rev* 2018;**31**. https://doi.org/10.1128/CMR.00079-17.
34. Endimiani A, Perez F, Bajaksouzian S, Windau AR, Good CE, Choudhary Y, et al. Evaluation of updated interpretative criteria for categorizing *Klebsiella pneumoniae* with reduced carbapenem susceptibility. *J Clin Microbiol* 2010;**48**:4417—25.
35. Wang JT, Wu UI, Lauderdale TLY, Chen MC, Li SY, Hsu LY, et al. Carbapenem-nonsusceptible enterobacteriaceae in Taiwan. *PLoS One* 2015;**10**:e0121668.
36. 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—77.
37. Lee NY, Lee CC, Huang WH, Tsui KC, Hsueh PR, Ko WC. Cefepime therapy for monomicrobial bacteremia caused by cefepime-susceptible extended-spectrum beta-lactamase-producing Enterobacteriaceae: MIC matters. *Clin Infect Dis* 2013;**56**:488—95.
38. Lee NY, Lee CC, Huang WH, Tsui KC, Hsueh PR, Ko WC. Carbapenem therapy for bacteremia due to extended-spectrum-β-lactamase-producing *Escherichia coli* or *Klebsiella pneumoniae*: implications of ertapenem susceptibility. *Antimicrob Agents Chemother* 2012;**56**:2888—93.
39. Kahlmeter G. Breakpoints for intravenously used cephalosporins in Enterobacteriaceae–EUCAST and CLSI breakpoints. *Clin Microbiol Infect* 2008;**14**(Suppl 1):169—74.
40. Daikos GL, Markogiannakis A. Carbapenemase-producing *Klebsiella pneumoniae*: (when) might we still consider treating with carbapenems? *Clin Microbiol Infect* 2011;**17**:1135—41.
41. Paul M, Carmeli Y, Durante-Mangoni E, Mouton JW, Tacconelli E, Theuretzbacher U, et al. Combination therapy for carbapenem-resistant gram-negative bacteria. *J Antimicrob Chemother* 2014;**69**:2305—9.
42. Abdelsalam MFA, Abdalla MS, El-Abhar HSE. Prospective, comparative clinical study between high-dose colistin monotherapy and colistin-meropenem combination therapy for treatment of hospital-acquired pneumonia and ventilator-associated pneumonia caused by multidrug-resistant *Klebsiella pneumoniae*. *J Glob Antimicrob Resist* 2018;**15**:127—35.

43. Doi Y, Paterson DL. Carbapenemase-producing enterobacteriaceae. *Semin Respir Crit Care Med* 2015;**36**:74—84.
44. Schwaber MJ, Klarfeld-Lidji S, Navon-Venezia S, Schwartz D, Leavitt A, Carmeli Y. Predictors of carbapenem-resistant *Klebsiella pneumoniae* acquisition among hospitalized adults and effect of acquisition on mortality. *Antimicrob Agents Chemother* 2008;**52**:1028—33.
45. Gupta N, Limbago BM, Patel JB, Kallen AJ. Carbapenem-resistant Enterobacteriaceae: epidemiology and prevention. *Clin Infect Dis* 2011;**53**:60—7.
46. Rafailidis PI, Falagas ME. Options for treating carbapenem-resistant Enterobacteriaceae. *Curr Opin Infect Dis* 2014;**27**:479—83.
47. Ku YH, Chen CC, Lee MF, Chuang YC, Tang HJ, Yu WL. Comparison of synergism between colistin, fosfomycin and tigecycline against extended-spectrum β-lactamase-producing *Klebsiella pneumoniae* isolates or with carbapenem resistance. *J Microbiol Immunol Infect* 2017;**50**:931—9.
48. Lee YL, Lu MC, Shao PL, Lu PL, Chen YH, Cheng SH, et al. Nationwide surveillance of antimicrobial resistance among clinically important gram-negative bacteria, with an emphasis on carbapenems and colistin: results from the Surveillance of Multicenter Antimicrobial Resistance in Taiwan (SMART) in 2018. *Int J Antimicrob Agents* 2019;**54**:318—28.

Appendix A. Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.jmii.2021.09.002.