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Polymyxin monotherapy or in combination against carbapenem-resistant bacteria: systematic review and meta-analysis

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Objectives: The objective of this study was to summarize available data on polymyxin-based combination therapy or monotherapy for carbapenem-resistant Gram-negative bacteria.

Methods: This is a systematic review. We included observational studies and randomized controlled trials (RCTs) comparing polymyxin monotherapy versus polymyxin-based combination therapy in adult patients with infections caused by carbapenem-resistant or carbapenemase-producing Gram-negative bacteria. Only named antibiotic regimens were included. The primary outcome was 30 day mortality. Unadjusted OR (uOR) and adjusted OR where available with 95% CI were pooled in random-effects meta-analyses.

Results: Twenty-two studies including 28 comparisons were included. Polymyxin monotherapy was associated with a uOR of 1.58 (95% CI = 1.03–2.42) for mortality compared with polymyxin/carbapenem combination therapy (seven observational studies, 537 patients), without heterogeneity. Subgrouping studies to serious and critical risk of bias resulted in uORs of 0.94 (95% CI = 0.42–2.09) and 1.94 (95% CI = 1.17–3.23), respectively. Mortality was significantly higher with polymyxin monotherapy compared with combination therapy with tigecycline, aminoglycosides or fosfomycin (potentially double-coverage regimens): uOR of 1.57 (95% CI = 1.06–2.32) overall (10 observational studies and 1 RCT, 585 patients, no heterogeneity) and uOR of 2.09 (95% CI = 1.21–3.6) for *Klebsiella pneumoniae* bacteraemia (7 observational studies, 285 patients, no heterogeneity); very low quality evidence. Two RCTs and one observational study assessing rifampicin/colistin combination therapy for *Acinetobacter baumannii* infections showed no difference in mortality compared with colistin monotherapy; moderate quality evidence.

Conclusions: The significant association observed in observational studies between polymyxin monotherapy and mortality cannot be taken as proof of combination therapy effects due to the low quality of the evidence. The only three RCTs to date show no effect of rifampicin/colistin or fosfomycin/colistin on mortality for *Acinetobacter* infections.

Introduction

The alarming rise of MDR Gram-negative bacteria (GNB) has required the reintroduction of the polymyxins, whose use was limited up until the past decade.[1] For carbapenem-resistant (CR) GNB, the polymyxins (colistin and polymyxin B) are sometimes the only therapeutic option,[2] with variable susceptibility also to tigecycline and aminoglycosides. Use of polymyxins has its difficulties, including poorer efficacy than β-lactams,[3] nephrotoxicity with high dosing[4] and development of resistance during therapy.[5] In order to improve outcomes, polymyxins are sometimes used in combination with other antibiotics,[6] whether they have *in vitro* activity against the bacteria or not. Combination therapy might prevent resistance development, achieve higher rates of success and allow lower doses or shorter treatment periods,[7] albeit with higher costs, potential side effects and a possible opposite effect on resistance development than that predicted by *in vitro* studies.[8] Polymyxins exert a detergent-like effect on lipid structures, thereby disrupting the bacterial cell wall. The proposed mechanism for synergy is that cell wall disruption will allow penetration of other antibiotics and disrupt antimicrobial resistance mechanisms such as efflux pumps.[9,10]

In vitro data support synergy for specific polymyxin-based combinations. Carbapenems demonstrated better synergy with polymyxins on *Acinetobacter baumannii* (AB) than on other bacteria and better synergy with meropenem than with imipenem.[11]

However, *in vitro* synergy studies are limited in their ability to predict clinical effects and clinical evidence is sparse. The main limitation of existing clinical studies assessing combination therapy for CRGNB relates to their assessment of combination therapy as a concept, rather than the assessment of specific combinations of antibiotics.[12] Current studies convey a general message of 'the more drugs, the better', a practice that cannot be implemented and might be ecologically unwise.

In this systematic review and meta-analysis, we sought to examine the effectiveness of polymyxin-based combination versus monotherapy, by antibiotic types and bacterial species.

Methods

We included all clinical studies [whether retrospective, prospective or randomized controlled trials (RCTs)] comparing intravenous polymyxin (colistin or polymyxin B) monotherapy versus any polymyxin-based combination therapy in adult patients with documented infection caused by polymyxin-susceptible, CR or carbapenemase-producing GNB, provided that the study reported on outcomes for a specific polymyxin and a specific combination regimen (named antibiotics). If more than one comparison was reported, we included all reported comparisons. No language or year restrictions were applied. We did not include studies using inhaled polymyxins. Only studies reporting on more than five patients per treatment group were included.

We searched PubMed, the Cochrane library, references of all included studies and narrative or systematic reviews on the topic.[13–15]

In the databases, we used the following search string: '(colisti* OR polymyxin) AND (enterobacteriaceae OR klebsiella OR acinetobacter OR e. coli OR pseudomonas) AND [in PubMed] (random* OR prospective OR retrospective OR cohort OR observational OR blind)'. We estimated that this search string will be relatively sensitive. The last search was run on 10 April 2016. Each study was screened and reviewed for eligibility independently by two authors. In case of missing data for an eligible study, an attempt was made to contact the study authors for clarification.

Outcomes

The primary outcome was 30 day all-cause mortality and if not reported at day 30 we extracted and documented the closest timepoint. Both crude outcome rates and adjusted effect estimates were extracted for mortality. Secondary outcomes included clinical and microbiological failure rates, length of hospital stay, superinfections, development of resistance to polymyxins or the combination antibiotic and occurrence of adverse events including *Clostridium difficile* infection, nephrotoxicity and others.

Data were extracted independently by two of the authors using a predefined data extraction form and then compared for verification. In the event of a dispute, a third author acted as referee. From individual studies, we sought to extract patient demographics, formulation and dosage including loading for polymyxins and the combination antibiotic, clinical data regarding the infection including source, place of acquisition sepsis presentation and severity, types and resistance profile of the bacteria.

We documented the study design and for all studies assigned the risk of bias for six domains of potential bias, addressing the outcome of mortality. The domains were taken from the Cochrane Risk of Bias Assessment Tool for Non-Randomized Studies of Interventions (ACROBAT-NRSI, currently renamed to ROBINS-I[16]). We addressed bias due to confounding (adjustment to risk factors other than the treatment regimen in observational studies), selection of participants (no inherent selection when assigning patients to monotherapy or combination therapy), classification of the intervention (protocol definitions of mono- and combination therapies regarding start in relation to infection onset and minimal duration), departure from intended intervention (information on adherence to the monotherapy and combination therapy regimens in relation to the group assignment), bias due to missing data (loss of patients for outcome analysis) and bias in selection of the reported results (reporting results for all predefined outcomes). The seventh domain (bias in measurement of the outcome) was not relevant. Each domain was assigned a low, moderate, serious (including serious and critical together) or unknown (no information) risk of bias. Quality of the evidence was ranked based on the risk of bias according to the GRADE classification.[17] Inclusion of mixed sources of infection and polymicrobial infections lowered the quality of the evidence in 'other considerations'. We assessed the effect of the quality of the evidence through subgroup analysis.

Data were compiled separately for each specific comparison and bacteria: *Klebsiella pneumoniae* (KP), AB and *Pseudomonas aeruginosa* (PA). As we anticipated a difference between the effects of combination with *in vitro* active antibiotics versus non-active ones, we planned a subgroup analysis of covering combinations (in which both drugs in the combination were active *in vitro*) for mortality. Lastly, we divided all studies into three subgroups according to infection types—bloodstream infections (BSI), ventilator-associated pneumonia (VAP) or hospital-acquired pneumonia (HAP) infections, and studies reporting on mixed types of infections. We computed OR with 95% CI and pooled these in the meta-analysis. A random-effects model with the restricted maximum likelihood estimation method was used for the analysis, as we assumed clinical heterogeneity between included studies. When comparing subgroups, we used mixed-effects analysis. I^2 and τ^2 statistics were used to report heterogeneity. We used R[18] with the Metafor[19] package for statistical analysis.

Results

The search yielded 494 references, of which 39 potentially eligible studies were identified for full-text review, resulting in 22 studies fulfilling the inclusion criteria (Figure 1) with 28 comparisons. Overall, nine studies assessed tigecycline,[20–27,45] seven studies assessed carbapenems,[21,22,28–32] three studies assessed rifampicin,[33–35] three studies assessed aminoglycosides,[21,25,36] three studies assessed sulbactam,[30,37,38] two studies assessed vancomycin,[22,39] one study assessed piperacillin/tazobactam[29] and one study assessed intravenous fosfomycin.[40] Five studies used polymyxin B while all others used colistin formulations. Only three studies used a colistin loading dose. The main study characteristics are reported in Table 1.

Half (11/22) of the studies reported on predominantly (>80%) ICU patients. The mean or median age ranged from 51 to 77 years. Nine studies permitted the inclusion of polymicrobial infections. Overall, 10 comparisons included BSI only, 13 included various types of infections and 5 were limited to HAP or VAP.

Three were RCTs and all others were retrospective observational studies including from 7–138 patients per treatment group (studies with fewer than 6 patients excluded). In the observational studies, an adjusted analysis for mortality was reported for only three comparisons between monotherapy and combination therapy. Since each involved a different comparison, these could not be pooled. All results presented, except for colistin/rifampicin, are based on unadjusted comparisons between groups. Only the RCTs were classified as low risk for selection of participants and confounding. All studies were at high risk of intervention measurement bias. Risk of bias assessment is presented in Figures 2 and 3. As data on length of hospital stay, superinfections, development of resistance and occurrence of adverse events including *C. difficile* infection, nephrotoxicity and others were missing, we do not report on these outcomes.

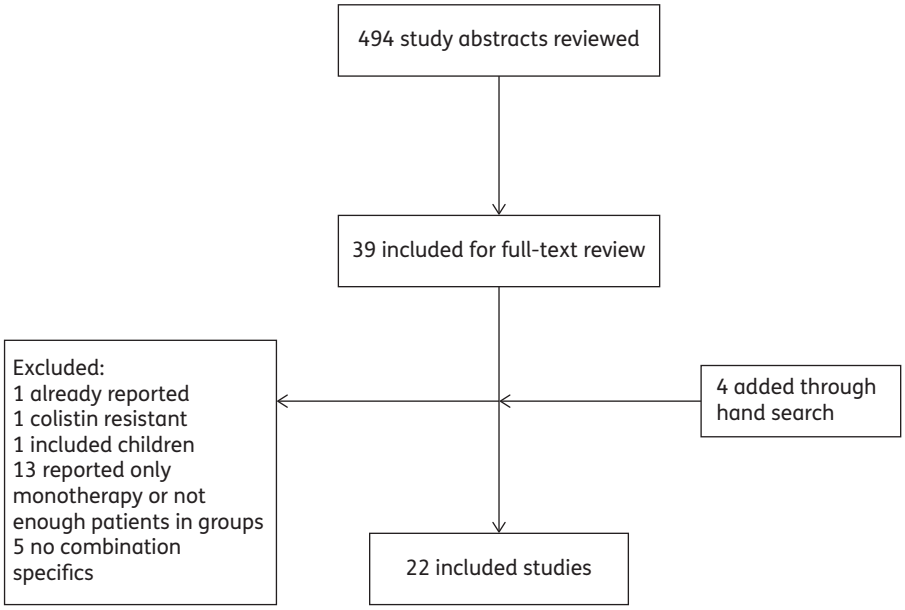

Figure 1. Study flow.

Polymyxin/carbapenem combinations

Seven observational studies reported on mortality with polymyxin monotherapy versus polymyxin/carbapenem combinations totalling 537 patients and yielding an OR of 1.58 (95% CI = 1.03–2.42, I^2 = 0%) in favour of combination therapy (Figure 4). Overall, the quality of the evidence was very low, but when subgrouping studies to serious and critical risk of bias considering all domains, the ORs were 0.94 (95% CI = 0.42–2.09, two studies) and 1.94 (95% CI = 1.17–3.23, five studies), respectively. There was no significant difference between ORs for studies assessing only *Acinetobacter* sp. and those assessing a mix of bacteria. Imipenem and meropenem doses were inconsistent; however, no study specifically used high-dose meropenem. The polymyxins assessed were polymyxin B (two studies) and colistin (four studies). Although studies included the baseline definitions for having carbapenem resistance, the presence of carbapenemases, their type and MIC range were largely lacking. Specifically, only one study[21] reported mortality by carbapenem MIC, finding an association between carbapenem MIC and mortality with combination therapy.

Three studies (369 patients) reported on clinical failure, with no difference between study groups [OR of 0.95 (95% CI = 0.52–1.73)]. Microbiological failure was reported in two studies (188 patients), with a statistically non-significant difference favouring combination therapy [OR of 0.63 (95% CI = 0.35–1.15)].

Colistin/rifampicin combination

Three studies assessed colistin/rifampicin combination therapy, with two RCTs[33,34] contributing 73% of the weight. All three studies included patients with AB infections. Mortality was assessed in a total of 284 patients and produced an OR of 1.46 (95% CI = 0.66–3.24, I^2 = 38%; moderate quality evidence). These studies also assessed microbiological failure, with an advantage to combination therapy [OR of 1.96 (95% CI = 1.19–3.22), I^2 = 0%; high-quality evidence]. One study examined clinical failure with 13/22 (59%) patients in monotherapy and 10/21 (48%) in combination therapy (*P* = not significant in the original study).

Potentially covering combination therapy

In this analysis, we included combination regimens of polymyxin with an aminoglycoside, tigecycline or fosfomycin. Although the studies did not provide complete susceptibility information for the combined antibiotic, bacteria were usually susceptible to these antibiotics. For the analysis of mortality, tigecycline combinations were examined in eight comparisons, aminoglycoside in three and both in addition to polymyxins were examined in two comparisons in a total of 10 studies. In addition, a single-centre RCT from Thailand examined the use of intravenous fosfomycin in combination with colistin. Together, these studies included 585 patients and yielded an OR of 1.57 (95% CI = 1.06–2.32, I^2 = 0%; Figure 5) in favour of potentially double-coverage combination therapy with an overall very low quality of evidence. The RCT showed no significant difference for 28 day mortality. When subgrouped according to risk of bias, the low-to-serious risk group (five studies) had an OR of 1.76 (95% CI = 0.98–3.18) while the critical risk group (six studies) had an OR of 1.42 (95% CI = 0.83–2.44). Restricting the analysis to KP BSI alone resulted in an OR of 2.09 (95% CI = 1.21–3.6, I^2 = 0%) (seven studies, 285 patients; Figure 6) with low-quality evidence (upgraded from very low due to a large effect and no heterogeneity).

Other combinations (Table 2)

In three studies examining polymyxin/sulbactam combination therapy, there was no significant difference for mortality (with significant heterogeneity) and failure (without heterogeneity). Monotherapy was associated with microbiological failure [OR of 2.47 (95% CI 1.33–4.59)]. In two studies reporting on colistin/glycopeptide combinations, there was no difference in all three

Table 1. Characteristics of included studies

Author	Publication year	Study years	Location	Study type	Bacteria	Carbapenemase	Carbapenem MIC (mg/L)	Polymicrobial	Setting	Infection type	Polymyxin	Combination with	Number of patients
Falagas	2010	2000–07	Greece	Ret	AB CRE PA	NA	NA	NA	mostly ICU	any	colistin	carbapenem	104
Petrosillo	2014	2010–11	Italy, Turkey	Ret	AB KP PA	NA	NA	yes	ICU	BSI VAP UTI SSTI IAI	colistin	vancomycin	103
												tigecycline	73
												carbapenem	82
Mouloudi	2010	2007–08	Greece	nested CC	KP	blaVIM blaKPC blaSHV blaTEM blaCTX-M	>64	NA	ICU	BSI	colistin	aminoglycoside	36
Nguyen	2010	2004–08	USA	Ret	KP	KPC-2 KPC-3	>8	no	mix	BSI	polymyxin B	tigecycline	22
Zarkotou	2011	2008–10	Greece	Ret	KP	blaKPC	NA	no	mostly ICU	BSI	polymyxin B	tigecycline	16
Ku	2012	2009	USA	Ret	AB CRE	blaKPC	NA	yes	mix	BSI HAP UTI SSTI	colistin	tigecycline	90
Simsek	2012	2008–11	Turkey	Ret	AB	NA	NA	NA	mix	BSI VAP SSTI	colistin	rifampicin	23
Tumbarello	2012	2010–11	Italy	Ret	KP	blaKPC2 blaKPC3	NA	NA	mix	BSI	colistin	tigecycline	45
												tigecycline + meropenem	38
Tumbarello	2012	2010–11	Italy	Ret	KP	NA	NA	NA	mix	BSI	colistin	tigecycline + meropenem	38
Durante-Mangoni	2013	2008–11	Italy	RCT	AB	blaOXA-51-like	≥16	yes	ICU	HAP VAP BSI IAI	colistin	rifampicin	209
Aydemir	2013	2011–12	Turkey	RCT	AB	NA	NA	no	ICU	VAP	colistin	rifampicin	43
Garnacho-Montero	2013	2008–11	Spain	Ret	AB	NA	NA	no	ICU	VAP BSI	colistin	vancomycin	57
Batirel	2014	2009–12	Turkey	Ret	AB	NA	NA	yes	mix	BSI	colistin	carbapenem	138
												sulbactam	105
Chuang	2014	2009–10	Taiwan	Ret	AB	NA	NA	no	ICU	HAP VAP	colistin	carbapenem	119
Kalin	2014	2011	Turkey	Ret	AB	NA	NA	NA	ICU	VAP	colistin	sulbactam	82
Daikos	2014	2009–10	Greece	Ret	KP	KPC-2 VIM-1	>8	NA	mix	BSI	colistin	tigecycline	33
												aminoglycoside	33
												both	39
Kontopidou	2014	2009–10	Greece	Ret/Pro	KP	KPC VIM	NA	NA	ICU	VAP BSI UTI SSTI IAI	colistin	aminoglycoside	43
												tigecycline	35
												both	30
Lopez-Cortes	2014	2010	Spain	Ret	AB	NA	NA	yes	mix	HAP UTI BSI IAI SSI	colistin	tigecycline	55
Crusio	2014	2009–10	USA	Pro	KP AB PA	NA	NA	no	mostly ICU	BSI	polymyxin B	carbapenem	56
Sirijatuphat	2014	2010–11	Thailand	RCT	AB	NA	NA	yes	mix	HAP VAP UTI BSI IAI SSTI CNS	colistin	fosfomycin	82
Yilmaz	2015	2011–13	Turkey	Ret	AB	NA	NA	NA	ICU	VAP	colistin	carbapenem	50
												sulbactam	37
Rigatto	2015	2013–14	Brazil	Ret	AB PA	NA	>32	yes	ICU	BSI HAP VAP UTI IAI	polymyxin B	carbapenem	92
Gomez-Simmonds	2016	2006–13	USA	Ret	KP	blaKPC	>16	no	ICU	BSI	polymyxin B	tigecycline and aminoglycoside	39

Ret, retrospective; Pro, prospective; CRE, carbapenem-resistant Enterobacteriaceae; SSI, surgical site infection; SSTI, skin and soft tissue; NA, not available; CC, case-control; IAI, intrabdominal infection; UTI, urinary tract infection.

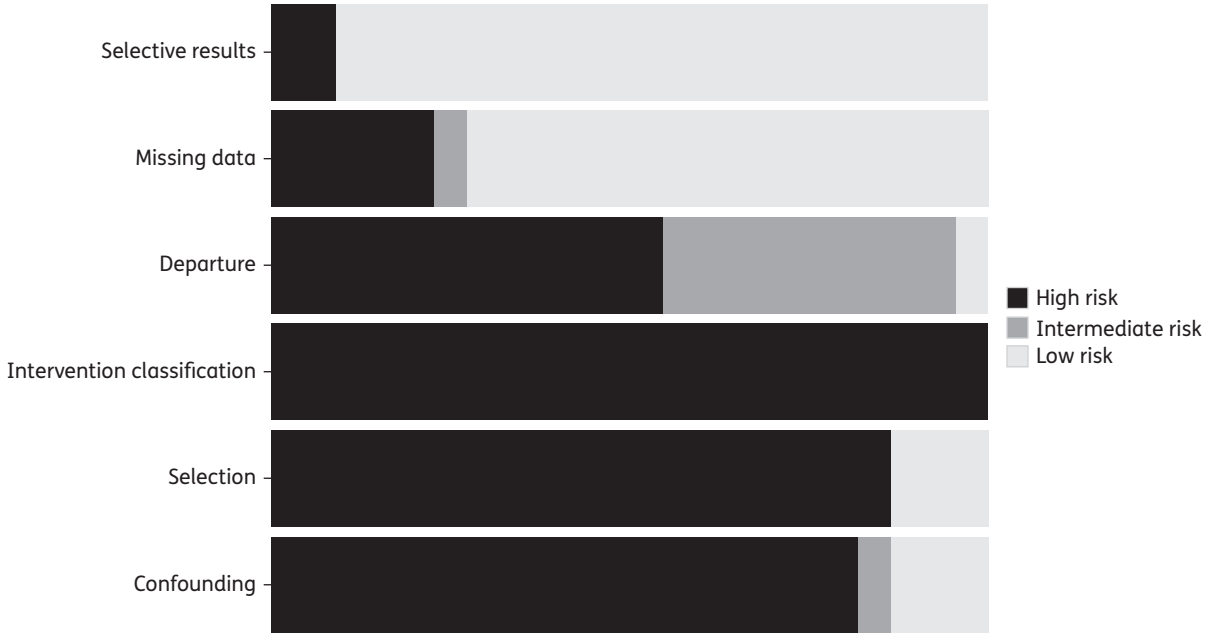

Figure 2. Risk of bias per domain.

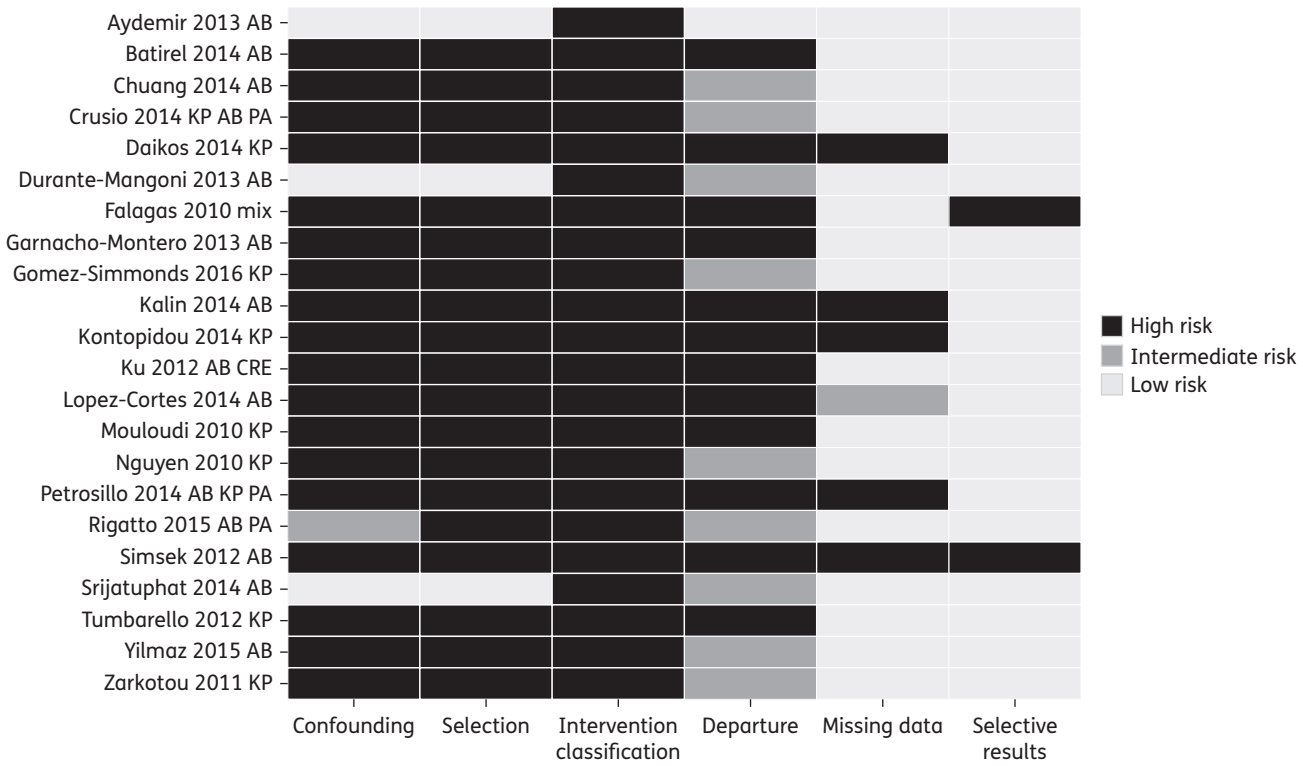

Figure 3. Risk of bias per study and domain. CRE, carbapenem-resistant Enterobacteriaceae.

outcomes. A study assessing piperacillin/tazobactam combination therapy reported only on clinical failure with no difference between groups.

Type of infection (monotherapy versus any combination)

Polymyxin monotherapy was associated with an OR of 2.23 (95% CI = 1.51 – 3.3) for mortality in BSI. For HAP/VAP [OR of 0.69 (95%

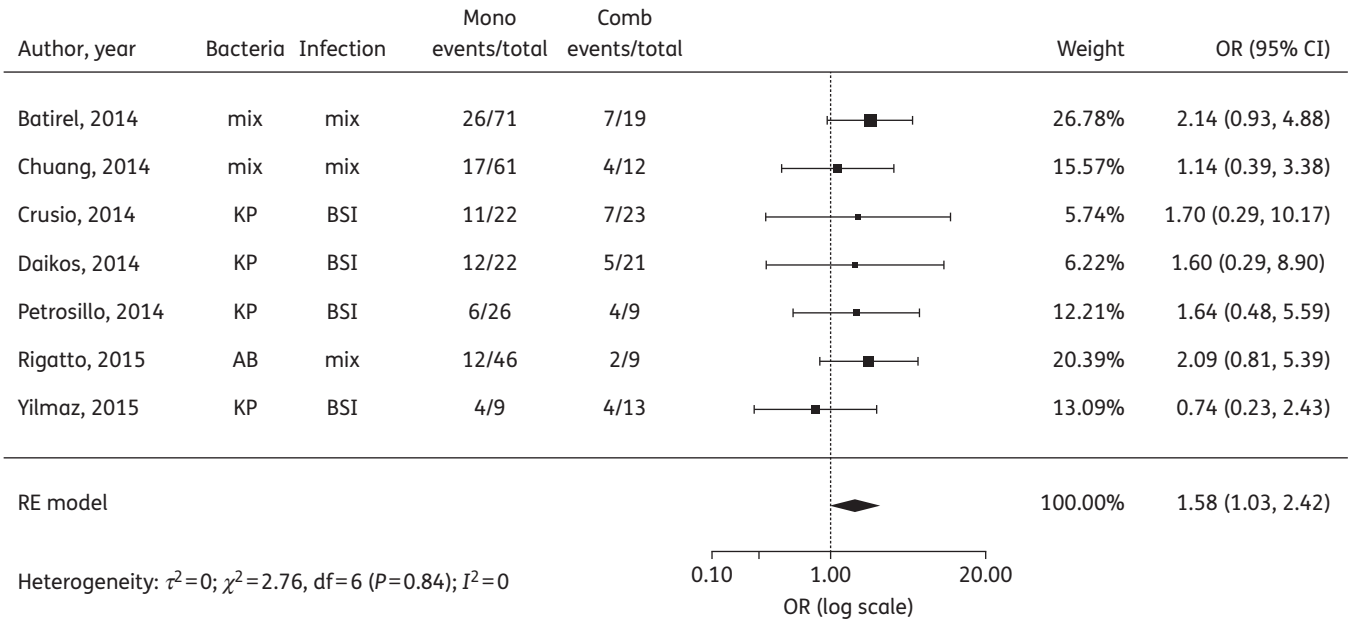

Figure 4. Polymyxin monotherapy versus combination with carbapenems, all-cause mortality. RE, random effects.

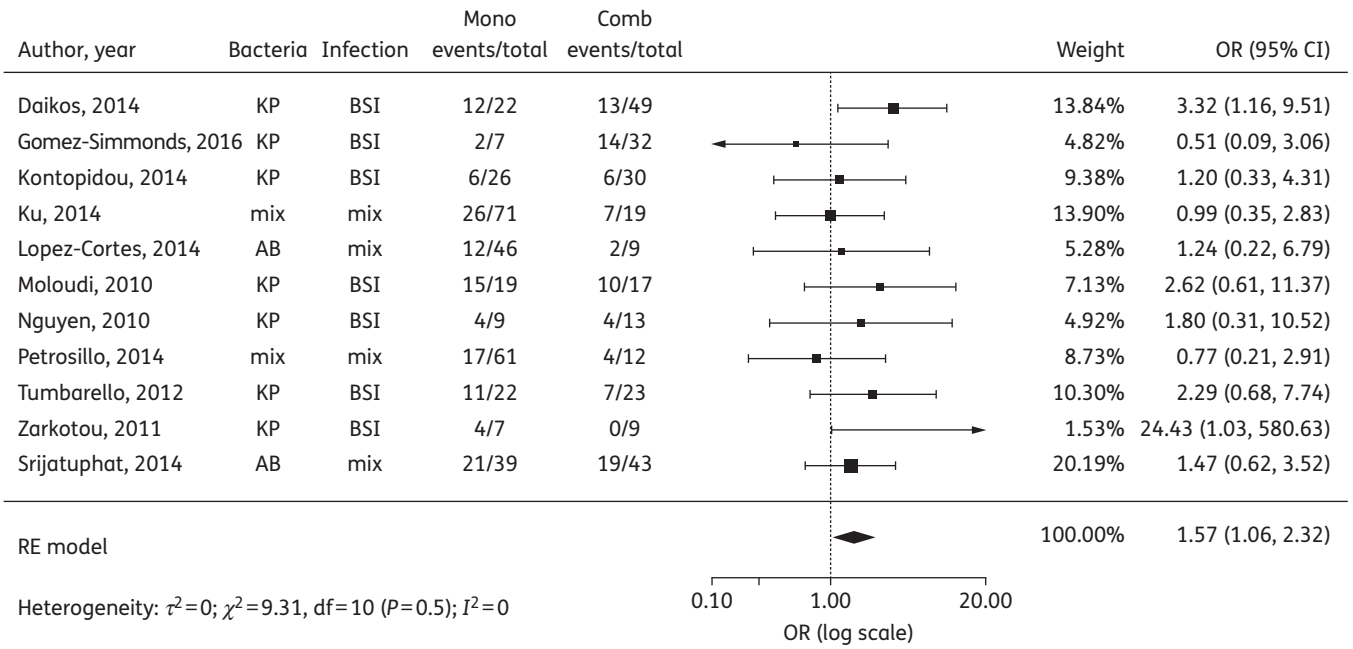

Figure 5. Polymyxin monotherapy versus combination with tigecycline, aminoglycoside or fosfomycin, all-cause mortality.

CI = 0.39–1.24)] and mixed infections [OR of 1.25 (95% CI = 0.87–1.80)], there was no significant mortality difference between monotherapy and combination therapy (Figure 7).

Discussion

We aimed to compile the knowledge to date on combination therapy for carbapenemase-producing, and preferably CRGNB, by type of combination therapy, type of bacterium and MIC of the carbapenems, recognizing that these factors underlie heterogeneity in response to the antibiotic regimen used. Of 28 studies reporting on polymyxin monotherapy versus polymyxin combination therapy for carbapenemase-producing GNB, only 23 reported mortality for named combinations. Of them, only 3 were RCTs and out of 18 observational studies, only 3 reported an adjusted OR for mortality with named combination regimens. The latter based the adjusted analysis on 61–136 patients included and 20–58 patients with outcomes per study.

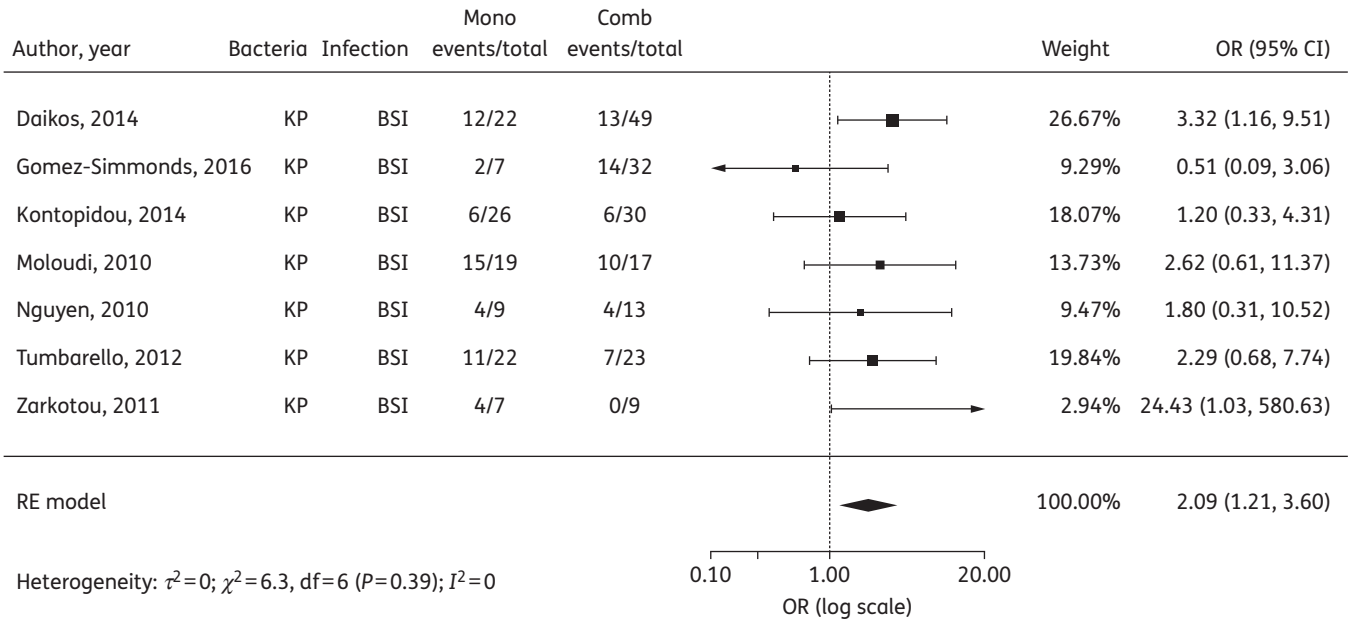

Figure 6. Polymyxin monotherapy versus combination with tigecycline or aminoglycoside in KP BSI, all-cause mortality.

The only evidence that we could compile from RCTs is that colistin/rifampicin combination therapy may have no clinical advantage over colistin monotherapy, but may result in better microbiological cure. The single RCT examining intravenous fosfomycin also did not show a survival advantage over the combination. Observational studies showed an unadjusted association between polymyxin monotherapy and mortality or clinical failure in comparison with polymyxin/carbapenem combination therapy, similar for different bacteria but addressing a mix of phenotypically carbapenem-susceptible and CR bacteria. A stronger, but still unadjusted association was observed for combination therapy of polymyxin with aminoglycosides, tigecycline or fosfomycin (double- to triple-coverage therapy) and survival, especially for KP bacteraemia. Except for the RCTs examining colistin/rifampicin and intravenous fosfomycin combination therapy, all results are based on very low quality evidence originating from selection bias, unattended confounding and with no control over treatment regimens assigned to patients. Subgroup analysis showed exaggeration of the association between polymyxin/carbapenem combinations and survival in studies with critical risk of bias.

An important risk of bias measure in observational studies provides confidence that patients indeed received the studied interventions (classification bias). Risk of bias was high for all studies in this domain; most studies did not provide explicit definitions for the treatment groups and we know from clinical practice how frequently treatments are modified along the course of an infection among patients with CRGNB infections. Furthermore, selection bias inherent to observational studies depends on the topic addressed and the likelihood of association between the intervention assessed and outcome.[41] We believe that the interventions of polymyxin monotherapy versus combination therapy are high on the selection bias plausibility range. We expect *a priori* that combination therapy would be given to patients with polymicrobial infections in which we do not know if the CR bacterium is the causative pathogen or to patients with better prognosis in locations where combination therapy is believed to be better than polymyxin monotherapy.[12] Importantly, none of the studies examining carbapenem combination therapy, except for one, reported carbapenem MICs. Carbapenems might be partially effective against CR bacteria in the low range of resistant MICs.[21] Thus, bias directs results to a survival advantage with combination therapy.

The main weakness of this study stems from the fact that the above traits were missing from the studies included in this systematic review.[42] We compiled unadjusted ORs from observational studies due to lack of adjusted data. Some studies did not restrict inclusion to specific types of infection and we show the importance of this factor. Studies assessing colistin mostly did not use a loading dose. The strength of systematic reviews and meta-analyses inevitably relies on the studies comprising them.[13–15] By nature, this study cannot provide better methodology than the referenced studies. Our conclusions are in conflict with the results presented—all observational analyses favour combination therapy and we argue that this is no proof for combinations' effects given the high risk of bias and discourage combination therapy. Two findings in our analysis supporting a possibly true effect might prove us wrong. The first is the lack of heterogeneity in all comparisons. The second is that grouping studies by the level of certainty in the CRGNB as the pathogen causing the infection (subgroup analysis by type of infection), demonstrated an advantage to combination therapy in CRGNB BSI, where we are certain of the pathogen, while the direction was opposite for VAP/HAP.

The optimal methodology to compare monotherapy versus combination therapy is to examine *in vitro* plausible combinations in an RCT. Two such trials are underway[43,44] that might shed some light on combination with carbapenems. However, recognizing the clinical and operational difficulties in conducting such a trial, high-quality observational studies still have a role. A good observational study should: examine specific interventions; define the

Table 2. Outcomes of other combinations

						Mortality		Clinical failure		Microbiological failure	
Author	Year	Bacteria	Polymyxin	Combination	Infection	monotherapy	combination therapy	monotherapy	combination therapy	monotherapy	combination therapy
Falagas	2010	AB PA	colistin	piperacillin/tazobactam	mix			6/35	6/16		
Batirel	2014	AB	colistin	sulbactam	BSI	26/36	32/69				
Kalin	2014	AB	colistin	sulbactam	VAP	27/47	27/35			13/47	5/35
Yilmaz	2015	AB	colistin	sulbactam	VAP	7/17	14/20	4/17	9/20	8/17	8/20
Garnacho-Montero	2013	AB	colistin	vancomycin	VAP BSI	14/28	14/29	7/28	10/29	8/23	11/29
Petrosillo	2014	mix	colistin	vancomycin	mix	17/61	14/42				

Figure 7. Polymyxin monotherapy versus combination therapy, all-cause mortality by infection type.

interventions by applying clear rules as to the minimal requirement for a patient to qualify as 'monotherapy' or 'combination therapy'; and attempt to control and report on important clinical variables such as restricting inclusion to monomicrobial infections and the appropriateness of empirical antibiotic therapy. An observational analysis can be done only if there are enough patients fulfilling the definitions defined for the interventions (addressing dosing, minimal duration and a relevant time window in relation to infection). Furthermore, enough patients are needed to allow for adjustment for important confounders with methods such as propensity score matching and/or controlling for covariates (e.g. regression).[42]

In summary, studies to date show an unadjusted association between combination therapy consisting of polymyxins with carbapenems or polymyxins with tigecycline and/or aminoglycosides and survival. Removing studies at critical risk of bias showed no association between carbapenem combination therapy and survival. No evidence exists to prove cause and effect for these comparisons. The only RCTs to date showed no mortality benefit for rifampicin or fosfomycin in combination with colistin for AB infections. No solid evidence exists for combination therapy against KP infections. The addition of carbapenems, tigecycline and aminoglycosides to polymyxins in the treatment of CRGNB cannot be recommended. Unnecessary use of carbapenems might fuel the epidemic of CRGNB in endemic settings. Well-conducted observational studies and RCTs are urgently needed.[43,44]

Acknowledgements

We would like to dedicate this manuscript to the memory of our dear colleague Sergey Altunin who passed away after completion of data extraction and start of the analysis.

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Transparency declarations

None to declare.

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