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REVIEW ARTICLE

Combination antibiotic therapy for multidrug-resistant Gram-negative bacteria

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Abstract

Combination antibiotic therapy for Gram-negative sepsis is controversial. The present review provides a brief summary of the existing knowledge on combination therapy for severe infections with multidrug-resistant *Pseudomonas* spp., *Acinetobacter* spp., and *Enterobacteriaceae.* Empirical combination antibiotic therapy is recommended for severe sepsis and septic shock to reduce mortality related to inappropriate antibiotic treatment. Because definitive combination therapy has not been proven superior to monotherapy in meta-analyses, it is generally advised to de-escalate antibiotic therapy when the antibiotic susceptibility profile is known, although it cannot be excluded that some subgroups of patients might still benefit from continued combination therapy. Definitive combination therapy is recommended for carbapenemase-producing *Enterobacteriaceae* and should also be considered for severe infections with *Pseudomonas* and *Acinetobacter* spp. when beta-lactams cannot be used. Because resistance to broad-spectrum beta-lactams is increasing in Gram-negative bacteria and because no new antibiotics are expected to become available in the near future, the antibacterial potential of combination therapy should be further explored. *In vitro* data suggest that combinations can be effective even if the bacteria are resistant to the individual antibiotics, although existing evidence is insufficient to support the choice of combinations and explain the synergistic effects observed. *In vitro* models can be used to screen for effective combinations that can later be validated in animal or clinical studies. Further, in the absence of clinical evidence, *in vitro* data might be useful in supporting therapeutic decisions for severe infections with multidrug-resistant Gram-negative bacteria.

Key words: *Acinetobacter, combination therapy, enterobacteriaceae, in vitro, multidrug-resistant, pseudomonas, sepsis, synergy*

Introduction

Combination antibiotic therapy is frequently used to treat severe Gram-negative infections but is controversial and debatable. Potential achievements with combinations as compared with monotherapy include a broader antibacterial spectrum, synergistic effects, and reduced risk for emerging resistance during therapy. In the absence of evidence-based treatment options, combinations are increasingly employed to enhance the antibacterial effects of available drugs against multidrug-resistant strains. However, excessive use of combinations should be avoided because it might be associated with increased risk for toxicity, superinfections, selection of resistant strains, and higher costs.

The aim of the present review is to present and discuss existing knowledge on combination therapy for severe infections with Gram-negative bacteria, as well as to examine the potential use of antibiotic combinations and *in vitro* studies to manage the growing threat of multidrug-resistant *Pseudomonas* spp., *Acinetobacter* spp., and *Enterobacteriaceae*.

Empirical combination therapy for Gram-negative sepsis

The results of published clinical studies and meta-analyses on combination therapy for Gram-negative sepsis are diverse and contradictory (1–5). In a review article, combination therapy was associated with reduced mortality only in the subgroup of

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Pseudomonas aeruginosa bacteraemia (1). In another review combination therapy was superior to monotherapy for severely ill patients, particularly those in septic shock (2). According to a recent Cochrane review, the addition of an aminoglycoside to a broad-spectrum beta-lactam does not reduce the overall mortality in patients with Gram-negative sepsis, but is associated with an increased risk for adverse events and is therefore discouraged (3).

The conflicting results might be explained by variations between studies with regard to patient characteristics, severity of infections, infection sites, causative bacteria, and antibiotic treatment. Delayed appropriate antibiotic therapy is known to be strongly associated with increased mortality in patients with septic shock (6), and broad-spectrum combination therapy will increase the probability for appropriate therapy as compared with single antibiotics (4,6–9). Therefore, empirical combination therapy is recommended for severe sepsis and septic shock with Gram-negative bacteria, particularly for neutropenic patients and patients at high risk of being infected with multidrug-resistant strains (10). The optimal choice of antibiotics depends on the local resistance epidemiology as well as individual risk factors for resistance, including recent antibiotic use, hospitalization, and previous colonization or infection with resistant strains (5).

Definitive combination therapy for Gram-negative sepsis

Definitive combination therapy including two antibiotics to which the bacteria are susceptible has been suggested to improve clinical outcome as compared with monotherapy for critically ill or neutropenic patients and severe infections with *Pseudomonas* spp. (11,12). Further, it has been argued that combinations should be used to prevent emergence of resistance during therapy (13). However, recent meta-analyses conclude that the existing clinical evidence is insufficient to support the use of definitive combination therapy for these reasons and that combination therapy is associated with an increased risk for ototoxicity, nephrotoxicity, bacterial superinfections, and selection of resistant strains (3,5). It has therefore been recommended to de-escalate antibiotic treatment to the most appropriate single agent as soon as the antibiotic susceptibility profile of the causative pathogen is known (5,10).

However, the non-inferiority with monotherapy reported in these meta-analyses refers to treatment with a broad-spectrum beta-lactam (3,5) and might not be valid for severe Gram-negative infections when these antibiotics cannot be used due to resistance or intolerance. For example, clinical studies strongly suggest that combination therapy is superior to monotherapy for carbapenemase-producing *Enterobacteriaceae*, even when the isolated bacteria are susceptible *in vitro* to the individual drugs (14–16). Monotherapy with an aminoglycoside is equally effective as beta-lactam antibiotics for urinary tract infections, but not for other infections, severe sepsis, or septic shock (17). Tigecycline is associated with higher mortality rates than carbapenems for severe Gram-negative infections, especially for hospital-acquired pneumonia, and has been questioned because of a bacteriostatic effect and reports of emerging resistance and breakthrough bacteraemia during therapy (18–22). Based on existing clinical data and a high risk for resistance development when used alone, monotherapy is not recommended for colistin or parenteral fosfomycin (14,15,21,23–25).

Thus, when beta-lactams are not suitable, prolonged or definitive combination therapy might be warranted for severe Gram-negative infections to improve the insufficient clinical efficacy of available treatment options.

Suggested antibiotic combinations

Clinical data to support the choice of antibiotic combinations are sparse and conflicting. Outcome might be difficult to assess for the severely ill patients included in these studies because of frequent changes in antibiotic therapy, co-morbidity and high all-cause mortality. Moreover, the results for specific combinations might differ between studies because of differences in patient material, infections, antibiotics used, dosage regimens, treatment durations, and strain-dependent factors.

Combination therapy for suspected Gram-negative sepsis and severe infections with *Pseudomonas* spp. typically includes a broad-spectrum beta-lactam and an aminoglycoside or a fluoroquinolone. However, colistin combinations are increasingly used as a last-resort treatment for multidrug-resistant strains (1,2,5,7–10,21). Combinations that include an aminoglycoside, ampicillin/sulbactam, a carbapenem, colistin, or rifampin have been successful against multidrug-resistant *Acinetobacter* spp. (26–29). Colistin–tigecycline and other combinations including an aminoglycoside, a carbapenem, colistin, fosfomycin, rifampin, or tigecycline have been advocated for carbapenemase-producing *Enterobacteriaceae* (14,16,21,30,31). Based on retrospective analysis, it has been recommended to use combinations including a carbapenem for these bacteria if the carbapenem minimum inhibitory concentration (MIC) is ≤4 mg/L (30).

Combinations effective in vitro

In vitro, antibiotic combinations are usually evaluated with the checkerboard method or by time-kill experiments using static antibiotic concentrations. According to standard definitions, synergy depicts an enhanced antibacterial effect with the combination after 24 hours as compared with the effects of the individual antibiotics. The results from published *in vitro* studies are conflicting, which may be due to differences in methods, antibiotic concentrations, bacterial inocula, and strain-dependent factors. However, in many of these studies antibiotic combinations have demonstrated synergistic or bactericidal effects against bacteria that have been resistant to the individual drugs.

For example, synergistic effects have been demonstrated for double and triple antibiotic combinations including an aminoglycoside, an anti-pseudomonal beta-lactam, colistin, a fluoroquinolone, a macrolide, or rifampin against multidrug-resistant *Pseudomonas* spp. (32–36). Double and triple antibiotic combinations including an aminoglycoside, ampicillin/sulbactam, a carbapenem, colistin, rifampin, tigecycline, or vancomycin have been effective against multidrug-resistant *Acinetobacter* spp. (35,37–40). For carbapenemase-producing *Enterobacteriaceae*, double and triple antibiotic combinations that include an aminoglycoside, aztreonam, a carbapenem, colistin, rifampin, tigecycline, or fosfomycin have demonstrated synergistic or bactericidal effects *in vitro* (25,35,41–44).

Mechanisms of synergy

The mechanisms of synergy are often not fully understood, but plausible explanations exist for some antibiotics. Colistin, which is frequently a component of effective combinations, increases the permeability of other antibiotics through the bacterial outer membrane by a detergent mechanism (45). This mechanism can counteract acquired resistance mediated by decreased antibiotic permeability (e.g. porin loss), and will also enable antibiotics that are not traditionally considered treatment options for Gram-negative bacteria to exert their actions. For instance, the addition of rifampin to colistin and meropenem/doripenem has resulted in synergistic effects *in vitro* against multidrug-resistant *Pseudomonas* spp., *Acinetobacter* spp., and carbapenemase-producing *Enterobacteriaceae* and has been reported as successful treatment in case reports (35,44,46,47). Synergy has sometimes been demonstrated for combination therapy that comprises several beta-lactams. For example, ertapenem–doripenem has been used against carbapenemase-producing *Klebsiella pneumoniae* (48,49). In these combinations synergy is probably achieved because beta-lactams, when hydrolysed, act as competitive beta-lactamase inhibitors (50).

Discussion and conclusions

Empirical combination antibiotic therapy is recommended for severe sepsis and septic shock caused by Gram-negative bacteria to reduce mortality related to inappropriate antibiotic treatment. Definitive combination therapy has not been proven superior to monotherapy with a broad-spectrum beta-lactam for patients with Gram-negative sepsis but is associated with an increased risk for toxicity and bacterial superinfections. However, the performed meta-analyses might have been insufficiently powered to detect benefits of definitive combination therapy in certain subgroups (e.g. critically ill or neutropenic patents and *Pseudomonas aeruginosa* bacteraemia). Definitive combination therapy is advocated for carbapenemase-producing *Enterobacteriaceae* and should also be considered for *Pseudomonas* and *Acinetobacter* spp. in situations in which beta-lactam monotherapy cannot be used because alternative antibiotics alone are often insufficient for severe infections.

Because resistance to carbapenems and other broad-spectrum beta-lactams is increasing, and because there is a lack of new antibiotics, it is urgent to explore the potential of combination therapy to enhance the antibacterial effects of available drugs. Clinical data to support the choice of combinations are insufficient. *In vitro* data suggest that combination therapy can be effective even if the bacteria are resistant to the individual drugs, but the results vary greatly between studies. A better appreciation of the mechanisms of synergy would facilitate the understanding of results obtained and help to predict the effects of other antibiotic combinations. For example, colistin is more likely to overcome impermeability than changes in the target molecule, and ertapenem can act as a competitive carbapenemase inhibitor in the periplasmic space only if the antibiotic molecules can penetrate the bacterial outer membrane.

For several reasons, the clinical relevance of *in vitro* findings is uncertain. However, *in vitro* models can be used to perform a large-scale screening for synergistic combinations to be further explored in animal studies and prospective clinical studies. In addition, in situations where there are no evidence-based treatment options, *in vitro* data can be useful to support therapeutic decisions for severe infections with multidrug-resistant Gram-negative bacteria.

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