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Editorial OMICS International

Multi-Drug Resistant Gram-Negative Bacteria: Antibiotic-Resistance and New Treatment Strategies

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Abstract

In this editorial, we treat the multi-drug-resistance of microorganisms such as *Klebsiella pneumonia* (*Kp*) and *Acinetobacter baumanii* and the issues concerning the management of these infections. Diseases caused by carbapenemase-resistant *Kp* (CR-*Kp*) represent an emerging threat worldwide due to high mortality rate and limited therapeutic options. Consequently innovative therapies have been suggested for their treatment. Colistin- based combinations are considered the milestone of the therapy for CR-*Kp*. They include meropenem+colistin, meropenem +colistin+tigecycline, the double carbapenem+colistin, tigecycline+colistin, colistin+gentamicin and even colistin +vancomycin. However, colistin use might be limited by its potential nephrotoxicity and resistance. Other antibiotic combinations concern the tigecycline with gentamicin, fosfomycin with aminoglycoside and ertapenem with meropenem.

Thus, the double carbapenem-regimen might be considered as a suitable therapy in those subjects in whom previous antimicrobial combinations failed. New antibiotics such as ceftazidime-avibactam effective on CR-*Kp* and ceftolozane-tazobactam active against XDR (Extensively Drug Resistant) *Pseudomonas aeruginosa* are now being used in many countries. The mortality results to be lower in patients treated with antibiotic combinations than in those who underwent monotherapy. Efforts should be made by the clinicians in order to limit the widespread of these resistant microorganisms all over the world. Encouraging new solutions as bacteriophage therapy or biocides currently does not seem the right choice.

Keywords: Multidrug-resistance; *Klebsiella pneumoniae*; *Acinetobacter baumannii*; Innovative therapies; Colistin; Antibiotics combinations; Mortality rate

Editorial

Infections caused by multidrug-resistant (MDR) Gram-negative bacteria such as *Acinetobacter baumannii* and *Klebsiella pneumoniae* constitute an important issue for establishing a correct and appropriate therapy in patients suffering from diseases deriving from these microorganisms, both in Intensive Care Units (ICUs) and in non-ICU settings. Carbapenemase-producing *klebsiella pneumoniae* (CP-*Kp*) is the most common microorganism involved in nosocomial infections over the last years. Outbreaks due to KPC (*K. pneumoniae*-carbapenemase) have been detected in many countries around the world; indeed these infections have become endemic other than in Europe also in United States, Israel, and China [1].

KPC is an important mechanism of resistance for an increasingly wide range of Gram-negative bacteria and is no longer limited to *K. pneumoniae* being present also in *E. coli.* The spread of carbapenem-resistant *Enterobacteriaceae* (CRE) has become a major challenge worldwide, with a high impact on both morbidity and mortality in human as well as in livestock. Infections due to CP-*Kp* are associated with a high mortality rate [2,3].

In addition to resistance to carbapenems, CRE have recently developed resistance to colistin, the cornerstone of combination therapy against these microorganisms, as well as to fluoroquinolones and aminoglycosides [4,5]. Consequently the proper management of these difficult-to-treat bacteria could be very important especially in some settings such as ICUs, cardiology and transplant wards where these infections play a crucial role.

There is evidence that combination therapies are more effective than monotherapy against CRE; however, the best regimen including two or more drugs has not been found yet [6]. In this challenging scenario, unconventional therapies have been proposed over the last years including colistin-based associations such as the double carbapenem regimen with colistin, colistin in combination with tigecycline, colistin plus gentamicin, colistin plus meropenem with or without tigecycline, even colistin plus vancomycin as well as combinations without colistin such as fosfomycin plus an aminoglycoside, tigecycline with gentamicin and meropenem plus ertapenem [2,7]. Several regimens of triple therapy are under consideration during the last years [8].

Anyway a double-carbapenem regimen has been shown to be effective and safe [9]. The use of the double carpanem therapy (i.e. different combinations of two carbapenems including ertapenem together with one of other carbapenems such as imipenem, meropenem or doripenem) has been found very useful showing a marked synergism between the two antimicrobial agents and a strong bactericidal effect compared with the single components [10]. In these combinations ertapenem plays a crucial role because it acts as a suicide inhibitor against the KPCs allowing other carbapenems to enter into the cells deprived of carbapenemases and to exert easily their antibacterial activity. Following this, at Academic Hospital of Sapienza University of Rome (Italy), we evaluated through antibiotic kill studies

the *in vitro* synergistic activity of meropenem plus ertapenem against pandrug-resistant *Klebsiella* isolated from 14 patients with CP-*Kp* infections who were successfully treated with a double-carbapenem therapy (ertapenem and meropenem). These carbapenems alone showed very high MIC values (between 128 and 256 mcg/ml) through macrodilution method in cation–adjusted Muller-Hinton broth. However their association induced both clinical (defervescence in 48 h) and microbiological (absence of growth in culture media after 48 h of therapy) responses. In *in vitro* studies, combination treatment exhibited higher bacterial killing than a single antibiotic, even in the presence of high carbapenem MICs. This report suggests that the double carbapenem regimen might be considered a promising option in CP-*Kp* infections, especially in patients for whom colistin treatment is inappropriate due to resistance or toxicity [11,12].

In any case, the addition of colistin seems to strongly increase the antibacterial activity of carbapenem-associations even against colistin-resistant bacteria [13].

A case of bloodstream infection due to pandrug-resistant *K. pneumoniae* has been reported to be successfully treated with an innovative regimen comprising a combination of colistin plus double carbapenem (ertapenem and meropenem at sub-MIC values). *In vitro* analysis showed the synergistic and bactericidal effect of this regimen [8].

However the role of colistin in the setting of colistin-resistant strains is still debated. Although considered as part of antimicrobial combinations against CR-*Kp,* colistin use might be limited by its potential nephrotoxicity and resistance; thus as stated before, colistin-free unconventional approaches such as the double-carbapenem regimen, have been recently proposed among innovative approaches as a valid alternative therapeutic option in severe infections.

The antibiotic associations are resulted to be appropriate also for *Acinetobacter baumannii* that does not produce KPC but oxacillinase. Therapeutic options for this microorganism are severely limited by the emergence of strains resistant to most antibiotics including carbapenems. In fact an increasing incidence of carbapenem-resistance has recently been reported among multidrug-resistant *Acinetobacter* due also to the decreased membrane permeability towards antibiotics and to the expression of active efflux pumps that excrete drugs. Yet this pathogen is intrinsically resistant to ertapenem so that the double carbapenem treatment is not suitable in this case. Consequently the treatment of these infections represents a real challenge. As a matter of facts, it has been reported that the association of colistin with vancomycin and meropenem results to be effective in pediatric patients from ICU with systemic infections by this MDR microorganism [14].

The rationale of using colistin plus vancomycin is based on the hypothesis that colistin increases the permeability of the outer membrane, thereby enhancing the antibacterial activity of large sized hydrophobic molecules, such as vancomycin, which are normally excluded by the Gram-negative outer membrane. Thus, the membrane-perturbing properties of colistin could allow vancomycin to reach its periplasmic target at inhibitory concentrations [14,15]. However clinical experience due to the use of these two nephrotoxic antibiotics is still poor and it should be more carefully evaluated.

Hence it would be interesting to examine these actual innovative therapies against MDR Gram-negative bacteria in order to estimate their activity related to different pathologies so that suitable treatments might be suggested. It has been reported that in monotherapy treated individuals, the mortality results to be higher than in patients who received combination treatments [7]. In severe infections such as those due to MDR bacteria, the patient's outcome and the mortality rate are often related to the commonly administered antibiotic treatment regimens.

With regard to patients who received combination treatments, mortality varies depending on the kind of the association. For instance in 51 patients receiving the tigecycline-colistin combination the mortality ranged from 0 to 30% [7,16], in 15 patients treated with tigecycline plus gentamicin this value was included between 0-50% [2,7], in 25 individuals undergone treatments with a carbapenem-colistin combination it ranged from 0-67% [17] and in 30 patients receiving colistin plus gentamicin it was from 40 to 61% [2,18]. Regarding patients who were treated with monotherapy, the mortality resulted to be higher (i.e. ranging from 7 to 80% for the gentamicin treatment) [2,16,19].

Anyway it can be deduced that the double or triple therapy may be considered a valid option for clinical cure and microbiological eradication in individuals with serious diseases.

The availability of new drugs against CRE seems to be encouraging for clinicians facing critically ill patients with systemic infections due to carbapenem-resistant *K. pneumoniae* and *A. baumannii;* however, certain data are still pending. New antibiotics such as ceftazidime-avibactam effective on KPC (*Klebsiella pneumoniae*-carpapenemase) and ceftolozane-tazobactam active against XDR (Extensively Drug Resistant) *Pseudomonas aeruginosa* are now being used in many countries [20-22]. These recently approved drugs have become available for MDR bacteria even for those microorganisms resistant to treatment regimens described so far. For instance ceftolozane-tazobactam (c/t) was recently approved for the cure of complicated intra-abdominal infections or urinary tract infections. Notably c/t demonstrated activity against many MDR isolates of *Pseudomonas aeruginosa* including carbapenem-resistant strains that do not produce a carbapenemase [20].

Newly discovered molecular mechanisms explain how bacteria manage to survive on antibiotic treatment and cause chronic and recurrent infections. This is often due to a physiological state called persistence where the bacteria are tolerant to multiple antibiotics. Bacterial cells may then switch into persistence which can lead to a dormant phase in a kind of hibernation. Afterwards the microorganisms can resuscitate to cause relapsing infections at any time after the treatment is abandoned. About this, it would be very interesting to study the molecular methods that underlie this issue [23].

Conclusions and Further Perspectives

The study of MDR *Enterobacteriaceae,* with particular emphasis on carbapenem-resistant *Klebsiella pneumoniae* and *Acinetobacter baumannii* should be focused on both phenotypic and genotypic pattern of antibiotic resistance and on new and active therapeutic regimens for a better understanding of the mechanism of antibiotic resistance and for suggesting more effective antimicrobial treatments. It has been reported that for infections due to KPC-producing *K. pneumoniae*, treatment failure was more common among patients who were treated with monotherapy than among those undergone antibiotic combinations [24]. The use of colistin in monotherapy could be a challenge even for individuals in whom this drug is effective due to its low plasma concentration thereby implying an increase in the

daily dose of the antibiotic with possible consequences due to colistin-induced nephrotoxicity. Innovative therapies are then advocated. The double carbapenem regimen alone or in combination with colistin even in patients yielding a colistin-resistant microorganism, the associations of tygecycline with colistin, fosfomycin with an aminoglycoside, a carbapenem or the colistin with an aminoglycoside and even colistin plus vancomycin, have been referred as antibiotic combinations effectively administered to patients infected with carbapenemase-producing *Enterobacteriaceae* [25]. Synergy studies *in vitro* correlated to clinical effectiveness, are a potential benefit arising from the use of antibiotic combinations. These associations might also prevent the occurrence of resistant strains during therapy and result in lower mortality in critically ill patients with *Klebsiella*-KPC infections [26]. The mortality rate results to be higher in a critical care setting (above 60%) than in non-ICU patients (below 50%) for the same antibiotic treatment either combination therapy or monotherapy [6].

The most common genotypic patterns of carbapenemases-producing *Klebsiella spp* such as KPC, metallo-β-lactamase and oxacillinase in various geographical areas, should be carefully taken into account.

Among the KPCs, the KPC2 and KPC3 are currently the most widespread variants whereas in the mid-90s only KPC1 was detected. The most frequent KPCs result to be the sequence types ST258 and ST512 belonging to the group KPC3 [27].

New emerging multi-resistant microorganisms other than *K. pneumoniae* and *A. baumannii* (i.e. *Enterobacter spp, E. coli, Proteus spp, Ps. aeruginosa*) are appearing in the current scenario worsening the present situation. Molecular investigations should be then conducted in order to identify the involved genes.

In conclusion, the multi-resistance is becoming an increasingly problem in the last decade that requires a drastic intervention for limiting the further widespread of resistant bacteria. New recently approved drugs such as ceftazidime-avibactam effective on KPC and ceftolozane-tazobactam effective on XDR *Ps. aeruginosa,* can be considered as a rescue treatment for these infections.

Alternatively new therapeutic programs other than antibiotics could be possibly taken into account such as some anti-bacterial experimental treatments taking advantage of medicinal oils, antibodies, common biocides [28], killing factors, drugs with antibacterial activity different from antibiotics and even phage therapy [29]. In the latter case a specific bacteriophage against a particular microorganism (i.e. MDR *Klebsiella pneumoniae* or *Acinetobacter baumannii*) could be used in order to lyse and kill the bacterium infected by the phage. Unlike the antibiotics, in this situation no resistance occurs. The limit of phage therapy, inherent in the bacteriophage nature, lies in a narrow spectrum of action that on one hand ensures high specificity and the absence of side-effects, on the other hand needs the absolute knowledge of the infection etiologic agent. The process that is on the basis of phage therapy, is complex requiring an efficient laboratory for isolating a strictly specific pathogen bacteriophage against the bacterium responsible of the infection. Phage therapy firstly employed by the microbiologist Felix-d'Herelle, was successfully adopted in clinical practice in eastern countries, but it has been completely abandoned by western countries [29].

All in all, it would be therefore highly recommended for clinicians to make every effort to limit the widespread of the MDR bacteria all over the world in order to avoid that the carbapenemases can become a "every day" as well as the ESBL (extended spectrum β-lactamases).

Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

References

1. Nordmann P, Cuzon G, Naas T (2009) The real threat of *Klebsiella pneumoniae* carbapenemase-producing bacteria. Lancet Infect Dis 9: 228-236.
2. Tumbarello M, Viale P, Viscoli C, Trecarichi EM, Tumietto F, et al. (2012) Predictors of mortality in bloodstream infections caused by *Klebsiella pneumoniae* carbapenemase-producing *K. pneumoniae*: importance of combination therapy. Clin Infect Dis 55: 943-950.
3. Capone A, Giannella M, Fortini D, Giordano A, Meledandri M, et al. (2013) High rate of colistin resistance among patients with carbapenem-resistant *Klebsiella pneumoniae* infection accounts, an excess of mortality. Clin Microbiol Infect 19: E23-30.
4. Falagas ME, Kasiakou SK (2005) Colistin: the revival of polymyxins for the management of multidrug-resistant Gram-negative bacterial infections. Clin Infect Dis 40: 1333-1341.
5. Alexander BT, Marschall J, Tibbetts RJ, Neuner EA, Dunne WM, et al. (2012) Treatment and clinical outcomes of urinary tract infections caused by KPC-producing *Enterobacteriaceae* in a retrospective cohort. Clin Ther 34: 1314-1323.
6. Falagas ME, Lourida P, Poulikakos P, Rafailidis PI, Tansarli GS, et al. (2014) Antibiotic Treatment of Infections Due to Carbapenem-Resistant *Enterobacteriaceae*: Systematic Evaluation of the available Evidence. Antimicrob Agents Chemother 58: 654-663.
7. Falagas ME, Zarkotou O, Pournaras S, Tselioti P, Dragoumanos V, et al. (2011) Predictors of mortality in patients with bloodstream infections caused by KPC producing *Klebsiella pneumoniae* and impact of appropriate antimicrobial treatment. Clin Microbiol Infect 17: 1798–1803.
8. Oliva A, Mascellino MT, Cipolla A, D'Abramo A, De Rosa A, et al. (2015) Therapeutic strategy for pandrug-resistant *Klebsiella pneumoniae* severe infections: short-course treatment with colistin increases the in vivo and in vitro activity of double carbapenem regimen. Int J Infect Dis 33: 132-134.
9. Giamarellou H, Galani L, Baziaka F, Karaiskos I (2013) Effectiveness of a double carbapenem regimen for infections in humans due to carbapenemase producing pandrug-resistant *Klebsiella pneumoniae*. Antimicrob Agents Chemother 57: 2388-2390.
10. Oliva A, D'Abramo A, D'Agostino C, Iannetta M, Mascellino MT, et al. (2014) Synergistic activity and effectiveness of a double-carbapenem regimen in pandrug-resistant *Klebsiella pneumoniae* bloodstream infections. J Antimicrob Chemother 69: 1718-1720.
11. Oliva A, Gizzi F, Mascellino MT, Cipolla A, D'Abramo A, et al. (2015) Bactericidal and synergistic activity of double-carbapenem regimen for infections caused by carbapenemase-producing *Klebsiella pneumoniae*. Clin Microbiol Infect 22: 147-153.
12. Bulik CC, Nicolau DP (2011) Double-carbapenem therapy for carbapenemase-producing *Klebsiella pneumoniae*. Antimicrob Agents Chemother 55: 3002e4.
13. Oliva A, Scorzolini L, Castaldi D, Gizzi F, De Angelis M, et al. (2017) Double-carbapenem regimen, alone or in combination with colistin, in the treatment of infections caused by carbapenem-resistant *Klebsiella pneumoniae* (CR-Kp). J Infect 74: 103-106.
14. Ceccarelli G, Oliva A, D'Ettorre G, D'Abramo A, Caresta E, et al. (2015) The role of vancomycin in addition with colistin and meropenem against colistin-sensitive multidrug resistant *Acinetobacter baumannii* causing severe infections in a Paediatric Intensive Care Unit. BMC Infectious Diseases 15: 393.

15. Gordon NC, Png K, Wareham DW (2010) Potent synergy and sustained bactericidal activity of a vancomycin–colistin combination versus multidrug resistant strains of *Acinetobacter baumannii*. Antimicrob Agents Chemother 54: 5316–5322.
16. Capone A, Giannella M, Fortini D, Giordano A, Meledandri M, et al. (2013) High rate of colistin resistance among patients with carbapenem-resistant *Klebsiella pneumoniae* infection accounts for an excess of mortality. Clin Microbiol Infect 19: 23-30.
17. Qureshi ZA, Paterson DL, Potoski BA, Kilayko MC, Sandovsky G, et al. (2012) Treatment outcome of bacteraemia due to KPC-producing *Klebsiella pneumoniae*: superiority of combination antimicrobial regimens. Antimicrob Agents Chemother 56: 2108-2113.
18. Mouloudi E, Protonotariou E, Zagorianou A, Iosifidis E, Karapanagiotou A, et al. (2010) Bloodstream infections caused by beta-lactamase *Klebsiella pneumoniae* carbapenemase-producing among intensive care unit patients in Greece: risk factors for infection and impact of type of resistance on outcomes. Infect Control Hosp Epidemiol 31: 1250–1256.
19. Alexander BT, Marschall J, Tibbetts RJ, Neuner EA, Dunne WM, et al. (2012) Treatment and clinical outcomes of urinary tract infections caused by KPC-producing *Enterobacteriaceae* in a retrospective cohort. Clin Ther 34: 1314–1323.
20. Munita JM, Aitken SL, Miller WR, Perez F, Rosa R, et al. (2017) Multicenter Evaluation of Ceftolozane/Tazobactam for Serious Infections Caused by Carbapenem-Resistant *Pseudomonas aeruginosa*. Clin Infect Dis.
21. Alidjanov JF, Fritzenwanker M, Hoffman I, Wagenlehner FM (2017) Ceftazidime-avibactam: novel antimicrobial combination for the treatment of complicated urinary tract infections. Future Microbiol.
22. Marshall S, Hujer AM, Rojas LJ, Papp-Wallace KM, Humphries RM, et al. (2017) Can Ceftazidime-Avibactam and Aztreonam Overcome β-Lactam Resistance Conferred by Metallo-β-Lactamases in *Enterobacteriaceae*?. Antimicrob Agents Chemother 61.
23. Wood TK, Knabel SJ, Kwan BW (2013) Bacterial persister cell formation and dormancy. Appl Eviron Microbiology 79: 7116-7121.
24. Lee GC, Burgess DS (2012) Treatment of *Klebsiella pneumoniae* carbapenemase (KPC) infections: a review of published case series and case reports. Ann Clin Microbiol Antimicrob 11: 32-38.
25. Falagas ME, Karageorgopoulos DE, Nordmann P (2011) Therapeutic options for infections with *Enterobacteriaceae* producing carbapenem-hydrolyzing enzymes. Future Microbiol 6: 653–666.
26. Souli M, Galani I, Boukovalas S, Gourgoulis MG, Chryssouli Z, et al. (2011) *In vitro* interactions of antimicrobial combinations with fosfomycin against KPC-2-producing *Klebsiella pneumoniae* and protection of resistance development. Antimicrob Agents Chemother 55: 2395-2397.
27. Nordmann P, Cuzon G, Naas T (2009) The real threat of *Klebsiella pneumoniae* carbapenemase-producing bacteria. Lancet Infect Dis 9: 228-236.
28. Vijayakumar R, Saleh Al-Aboody M, Al-Fonaisan MK, Alturaiki W, Mickymaray S, et al. (2016) Determination of minimum inhibitory concentrations of common biocides to multidrug-resistant Gram-negative bacteria. Appl Med Res 2: 56-62.
29. Wright A, Hawkins CH, Anggard EE, Harper DR (2009) A controlled clinical trial of a therapeutic bacteriophage preparation in chronic otitis due to antibiotic-resistant *Pseudomonas aeruginosa;* a preliminary report of efficacy. Clin Otolaryngol 34: 349-357.
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