plasmid in *C. werkmanii* from various sources will offer more insights into the potential role of *mcr-8.1* in the dissemination of colistin resistance.

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Transparency declarations

We declare no conflicts of interest.

Data availability

The complete nucleotide sequence of AHM21C2521I, pHNAH212521-MCR-8 and pHNAH212521-NDM-1 has been deposited in GenBank under accession numbers CP144503, CP144504 and CP144505, respectively.

Supplementary data

Figures S1 and S2 and Tables S1 and S2 are available as Supplementary data at *The Journal of Antimicrobial Chemotherapy* Online.

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Beyond the FIC index: the extended information from fractional inhibitory concentrations (FICs)

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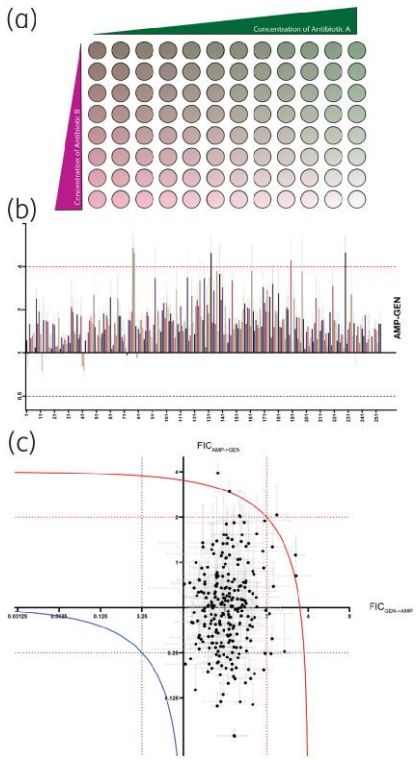

Figure 1. Gentamycin (GEN) and ampicillin (AMP) checkerboard assays and new data based on the FIC index. (a) A schematic representation of a checkerboard assay indicating the concentrations present of both antibiotics in the wells of the microtitre plate. (b) Data previously published on the FICi index of the interaction between AMP and GEN in a collection of 254 clinical *E. coli* isolates.[5] (c) the same experiments as (b) but reanalysed and illustrated as separate FIC values. The dotted lines indicate the limits of synergy and antagonism in the FIC representation (blue for synergy red for antagonism). The continuous lines represent the FICi limits of synergy and antagonism similarly (FICi = 0.5 for synergy and FICi = 4 for antagonism). To allow for intuitive reading of the FIC figure, values are represented on $\log_2$ axes starting at 0.5. This way, values to the left of the axes origin represent positive effects (synergistic) and to the right negative effects (antagonistic). Grey lines are standard deviations based on at least $n = 3$ for every data point.

Various assays, such as the commonly used checkerboard assay, quantify antibiotic interactions by calculating the fractional inhibitory concentration index (FICi).

$$FICi = \frac{C_A}{MIC_A} + \frac{C_B}{MIC_B}$$

where MIC_A and MIC_B are the minimum inhibitory concentrations of antibiotics A and B, and C_A and C_B are the concentrations inhibiting bacterial growth in combination.

According to Loewe's additivity model, in an additive interaction the FICi equation simplifies to 1. Deviations from FICi = 1 indicate interactions: FICi > 1 shows negative interaction, while FICi < 1 indicates a positive one. Practical limits define synergy as FICi ≤ 0.5 and antagonism as FICi ≥ 4.

The FICi value, however, has limitations. It is uncommon that antibiotics interact in a balanced reciprocal way as Loewe's model predicts, and there are many examples of established antibiotic interactions where the mechanism behind them is directional.[1] The combination of streptomycin with penicillin is synergistic in *Escherichia coli* due to penicillin damaging the cell membrane and causing increased uptake of streptomycin.[2] However, streptomycin has no effect on the action of penicillin. Similarly, the combination of streptomycin and cefotaxime is synergistic in *Enterobacter cloacae* due to streptomycin inhibiting a β-lactamase that impedes the effect of cefotaxime.[3] For antagonisms, the combination of colistin with vancomycin is prominent, with colistin suppressing the effects of vancomycin while having no effect from it.[4] All these interactions can be quantified with a FICi score, but lack information on which compound is the effector and which is the affected, respectively. Taking these considerations into account and the fact that the FIC index is an addition of two concentration ratios, $\frac{C_A}{MIC_A}$ for antibiotic A and $\frac{C_B}{MIC_B}$ for antibiotic B it is clear that some information is lost when adding the two ratios.

However, information on the directionality and nature of the interaction can be recovered if we consider the individual FICs as separate metrics:

$$FIC_{B\to A} = \left(\frac{C_A}{MIC_A}\right)$$

$$FIC_{A\to B} = \left(\frac{C_B}{MIC_B}\right)$$

The FICi index becomes

$$FIC_i = FIC_{B\to A} + FIC_{A\to B}$$

$FIC_{A\to B}$ is then a ratio between the inhibitory concentration of drug B alone (MIC_B) and the inhibitory concentration of drug B in combination with A (C_B). The ratio of these two can be interpreted as a metric of how the addition of drug A has affected the killing of B, showing whether A promotes or inhibits the action of B.

Each of the two fractional inhibitory concentrations (FIC) contributes equally to the sum that is the FIC index. Thus, a FICi = 1 could potentially be the result of many different pairs of $FIC_{B\to A}$ *and* $FIC_{A\to B}$. For instance, a scenario where $FIC_{B\to A} = 0.5$ *and* $FIC_{A\to B} = 0.5$ adds up to the same FICi = 1 as a scenario where $FIC_{B\to A} = 0.1$ *and* $FIC_{A\to B} = 0.9$, even though the nature of the interaction is clearly different.

Instead of adding the two FIC for a single FICi value the same information can be represented in a two-axis plot where $FIC_{B\to A}$ is on the *x*-axis and $FIC_{A\to B}$ on the *y*-axis. Our traditional limits of synergy and antagonism can be adapted to that representation assuming a completely bilateral interaction where $FIC_{B\to A} = FIC_{A\to B}$. Then a synergistic FICi < 0.5 translates to $FIC_{B\to A} = FIC_{A\to B} = 0.25$ and an antagonistic FICi > 4 translates to $FIC_{B\to A} = FIC_{A\to B} = 2$. This yields the following limits of the unilateral interactions:

$FIC_{A\rightarrow B} \leq 0.25$: *Compound A promotes the action ofcompound B*
$0.25 < FIC_{A\rightarrow B} < 2$: *Compounds A and B are additive/independent*
$FIC_{A\rightarrow B} = 0.5$ *is the threshold of additivity/independence*
$FIC_{A\rightarrow B} \geq 2$: *Compound A inhibits the action of compound B*

Applying this analysis to an already published dataset of ampicillin and gentamicin interactions in *E. coli* isolates[5] shows a more complete picture then the original analysis (Figure 1). While the original FICi metric (Figure 1b) suggested additive interactions, individual FIC values reveal a range of effects (Figure 1c), Specifically, gentamicin consistently inhibits the efficacy of ampicillin to varying degrees ($FIC_{GEN\rightarrow AMP}$, in Figure 1c), and ampicillin has a variable impact on the efficacy of gentamicin ($FIC_{AMP\rightarrow GEN}$, in Figure 1c), which was previously masked by the FICi metric. We suggest that this easy analysis will improve our understanding of how antibiotics interact.

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Evaluation of a novel lateral flow immunochromatographic assay for the rapid detection of KPC, NDM, IMP, VIM and OXA-48 carbapenemases in Gram-negatives

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The dissemination of carbapenemase-producing (CP) Gram-negatives is a matter of great clinical concern given the major role of these pathogens as causes of nosocomial infections. The production of carbapenemases and ESBLs has become a signature for difficult-to-treat infections (DTIs) because the spectrum of available therapies is drastically reduced.[1] The rapid and effective detection of antibiotic-resistant bacteria is a critical step for antibiotic stewardship and infection control.[1,2] Despite technological improvements, the identification of pathogenic bacteria, as well as the detection of antibiotic resistance, remains complex and time-consuming, with time to results often above 24–48 h or associated with very high costs, such as multiplex PCRs.[3]

The most widespread carbapenemases in Enterobacterales belong to Ambler class A (mostly KPC enzymes), MBLs (Ambler class B) of NDM, VIM and IMP type, and carbapenem-hydrolysing Ambler class D enzymes of OXA-48 type.[3,4] Phenotypic approaches such as antimicrobial susceptibility, even though cheap, require bacterial growth (>24 h), and are often difficult