
Supplementary information

Testing physiologically relevant conditions in minimal inhibitory concentration assays

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Supplementary Table S1. Minimal inhibitory concentrations of antibiotics and antimicrobial peptides against representative strains of ESKAPE pathogens in various media reflecting lab or *in vivo* conditions. Host mimicking conditions include Mueller Hinton broth (MHB), Tissue culture media, media with human serum, synthetic cystic fibrosis sputum medium (SCFM) with and without the addition of mucin and lung surfactant. Values are given as µg/mL for the following antibiotics: Azithromycin (AZM), erythromycin (ERM), clarithromycin (CLR), ciprofloxacin (CIP), trimethoprim (TMP), sulfamethoxazole (SMZ), Ampicillin (AMP); Daptomycin (DAP), Vancomycin (VAN), and Minocycline (MINO). Bolded values indicate MICs with more that 2-fold decreases in physiologically relevant conditions. – indicates no data was obtained.

Antibiotic	MHB	Tissue Culture Medium ^a	Human Serum-containing medium ^b	SCFM	SCFM + 0.4% mucin	Lung surfactant
<i>Pseudomonas aeruginosa</i> (PA01)						
AZM ^{1,2}	128	2-8	2	32	4	-
ERM ^{1,2,3}	64-128	64	8-64	>256	32	256
CLR ¹	128	64	64	64	64	-
CIP ^{3,*}	<0.25-0.5	<0.25	<0.25-0.5	0.25	1	4
TMP [*]	64	128	64	32	64	-
MINO ³	16	-	>256	-	-	32
AMP ^{3,*}	128-256	128	64-128	256	256	256
<i>Klebsiella pneumoniae</i> (KPLN649)						
AZM [*]	8	2	1	256	2	-
ERM [*]	256	128	128	>128	16	-
CLR [*]	256	128	64	128	8	-
CIP [*]	1	0.5	0.5	1	1	-
TMP [*]	>256	>256	>256	>256	>256	-
AMP [*]	128	128	128	128	-	-
<i>Acinetobacter baumannii</i> (Ab5075/ 98-37-09)						
AZM ^{2,*}	8-32	0.5-1	0.5	256	2	-
ERM ^{2,*}	32	1-4	2	256	32	-
CLR ^{2,*}	32-64	0.5-32	4-32	256	64	-
CIP ^{2,*}	16	32	32	1	64	-
TMP [*]	256	256	256	256	256	-
AMP ³	8	8	8	-	-	0.5
<i>Enterobacter cloacae</i> (218R1)						
AZM [*]	4	1	< 0.5	256	4	-
ERM [*]	128	32	8	256	32	-
CLR [*]	256	256	64	>256	64	-
CIP [*]	<0.5	<0.25	<0.25	0.5	<0.25	-
TMP [*]	<0.5	2	<0.5	2	32	-
<i>Enterococcus faecium</i> #1–1 (ATCC 29212)						
AZM [*]	4	< 0.5	< 0.5	64	2	-
ERM [*]	16	1	< 0.5	16	32	-
CLR [*]	16	2,4	1	128	32	-

CIP*	<0.5	1	2	1	32	-
TMP*	4	<0.5	<0.5	<0.5	32	-
<i>Staphylococcus aureus</i> (ATCC 25923)						
AZM*	1	<0.25	<0.25	1	-	-
ERM*	8	0.5	4	1	-	-
CLR*	1	<0.5	<0.5	1	-	-
CIP*	<0.25	<0.25	<0.25	<0.25	-	-
VAN*	1.5	<0.25	<0.25	1.5	-	-
TMP*	1	<0.5	<0.5	1	-	-
SMZ*	>256	>256	128->256	>256	-	-
AMP*	1	<0.25	<0.25-2	<0.25	-	-

^a Tissue culture media refers to RPMI-1640 with 5% MHB¹ or 5% LB² added.

^b Human serum refers to RPMI-1640+5% MHB +20% human serum^{1,*} or 100% human serum³.

* Unpublished data from Hancock lab

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2. Lin, L. *et al.* Azithromycin synergizes with cationic antimicrobial peptides to exert bactericidal and therapeutic activity against highly multidrug-resistant Gram-negative bacterial pathogens. *EBioMedicine* **2**, 690–698 (2015).
3. Colquhoun, J. M., Wozniak, R. A. F. & Dunman, P. M. Clinically relevant growth conditions alter *Acinetobacter baumannii* antibiotic susceptibility and promote identification of novel antibacterial agents. *PLOS ONE* **10**, e0143033 (2015).