SUPPORTING INFORMATION

Modulation of antimicrobial potency of human cathelicidin peptides against the ESKAPE pathogens and *in vivo* efficacy in a murine catheter-associated biofilm model

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Supporting Tables

Peptide	Hemolysis% at 220 µM	HPLC retention time (min)
17mF-W	7.1	10.535
17W2	8.0	10.452
17tF-W	12.1	11.108
17B-tF	30.2	11.621
17tF2	32.5	11.625
17BIPHE2	61.5	11.725

Table S1. Correlation between hemolysis and HPLC retention time of the designed LL-37 peptides¹

¹It is evident that there is a logarithmic correlation between hemolysis and peptide HPLC retention time. The longer the retention time, the more hemolytic the peptide.



Correlation between hemolysis and HPLC retention time

PeptideHEK293HaCaTTHP-117tF-W>25>25>12.517BIPHE2>25>25>12.5

Table S2. Toxic effects (LC50 in μ M) of 17tF-W and 17BIPHE2 on other human cell lines¹

¹Cytotoxicity was evaluated by the XTT assays as described [74].

Supporting Figures

Figure S1: Evidence for high purity of the peptides. All the peptides show a single major peak detected at 220 nm by UV. The retention time for each peak is labeled (see methods).

17tF-W



17mF-W



17W2



17B-tF



17tF2



17BIPHE2



Figure S2: The S. aureus USA300 killing kinetics of Group 2 peptides at 3.1 µM.



Figure S3: LL-37 based designer peptides to the action of proteases analyzed by SDS-PAGE.

(A) Peptide bands of 17BIPHE2 (~2 kD) after incubation with chymotrypsin (C), tryptin (T), *S. aureus* protease V8 (V), or fungal proteinase K (K) for 0 and 24 h. 17BIPHE2 was degraded only by tryptin (lane 5), M is protein marker. (B) SDS-PAGE of peptides 17tF2 and 17B-tF after 24 h digestion by the same four proteases in panel A. (C) SDS-PAGE of peptides 17W2 and 17BIPHE2 after 24 h digestion by the same four proteases in panel A at 37°C. In both panels (B) and (C), lanes 1,6: control peptide; 2,7: chymotrypsin; 3,8: Trypsin; 4,9: V8 protease; 5,10: proteinase K.





Ctr C T V K Ctr C T V K



Ctr C T V K Ctr C T V K

Fig. S4. Comparison of membrane permeation of *A. baumannii* by the LL-37 designer peptides at 3.1 μ M. Propidium iodide is used as an indicator dye. RFU = Relative Fluorescence Unit.



Fig. S5. Membrane permeation of *K. pneumoniae* by the LL-37 designer peptides at 3.1 μ M. Propidium iodide is used as an indicator dye. RFU = Relative Fluorescence Unit.

