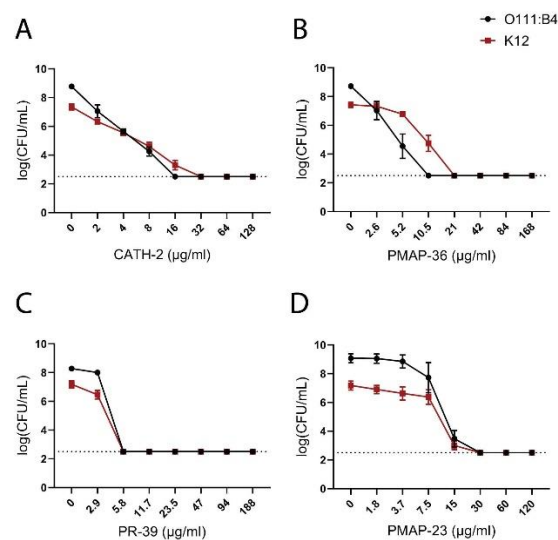


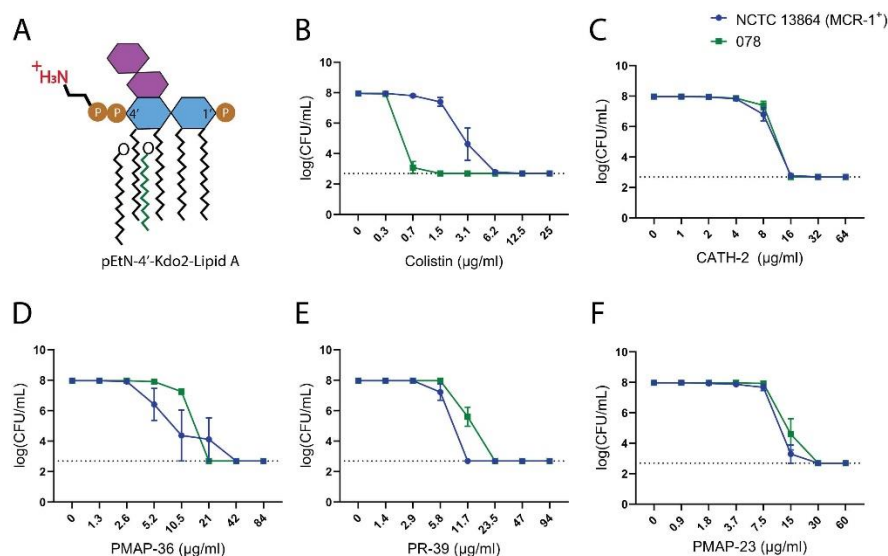
## Supplementary data

# Effects of *Escherichia. coli* LPS structure on antibacterial and anti-endotoxin activities of host defense peptides

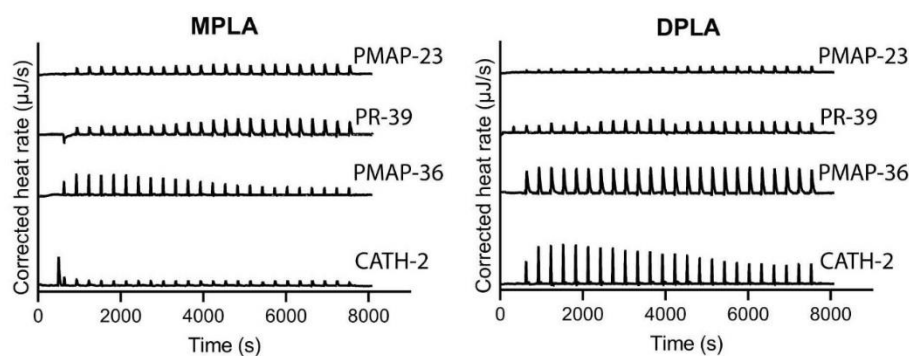
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**Figure S1.** Determination of MBC values (μg/ml) of HDPs against *E. coli* strains (O111:B4 or K-12). MBC values of CATH-2 (A), PMAP-36 (B), PR-39 (C), and PMAP-23 (D) were determined by colony count assay. Surviving bacterial colonies were detected after incubation with HDPs for 3 h in MHB. Shown are mean ± SEM of three independent experiments, dashed line shows the detection limit of the assay.



**Figure S2.** Determination of MBC values ( $\mu\text{g/ml}$ ) of HDPs against MCR-1<sup>+</sup> *E. coli*. **(A)** MCR-1<sup>+</sup> bacteria have a Phosphoethanolamine (pEtN) group attached to 1' or 4'-Phosphate of Lipid A in the LPS. MBC values of Colistin **(B)**, CATH-2 **(C)**, PMAP-36 **(D)**, PR-39 **(E)** and PMAP-23 **(F)** for *E. coli* strains (NCTC 13864 or 078) were determined by colony count assay. Surviving bacterial colonies were detected after incubation with HDPs for 3 h in MHB. Shown are mean  $\pm$  SEM of three independent experiments, dashed line shows the detection limit of the assay.



**Figure S3.** ITC spectra of the interaction between Lipid A species and peptides. 200  $\mu\text{M}$  of CATH-2, PMAP-36, PR-39 or PMAP-23 was titrated into *E. coli* Lipid A (MPLA or DPLA) solution (25  $\mu\text{M}$ ) and heat rates were recorded. Shown is a representative of two measurements.

**Table S1.** Dissociation constants and stoichiometry calculated using ITC data for interactions between *E. coli* LPS or Lipid A and HDPs. Data was fitted using an independent model, except for PMAP-36 and *E. coli* O111 LPS where a multiple sites model was used. n.d. = not defined which indicates no binding occurred. Shown is an average of two experiments.

	<i>E. coli</i> O111		<i>E. coli</i> K-12		MPLA		DPLA	
	Kd	n	Kd	n	Kd	n	Kd	n
CATH-2	2.55E-06	0.66	1.15E-06	0.35	n.d.	n.d.	1.02E-06	1.5
PMAP-36	2.50E-08	0.11	n.d.	n.d.	2.09E-06	1.3	n.d.	n.d.
	6.54E-08	1.56						
PR-39	3.68E-07	0.34	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
PMAP-23	1.83E-06	1.18	3.35E-07	1.16	n.d.	n.d.	n.d.	n.d.