

Supplementary Material A - Figures and Tables for:

***Limosilactobacillus reuteri*-derived tripeptide SKL exerts antibacterial activity against systemic and wound infections via dual membrane-killing and anti-virulence mechanisms**

Qi Lu ^{a, b}, Di Wang ^a, Haojie Jing ^a, Haijian Wang ^{a, b}, Na Li ^a, Baikui Wang ^{a, c}, Longhai Yu ^a, Yifei Feng ^{a, b}, Liyuan Mo ^a, Kun Zhou ^a, Min Yue ^{a, c}, Yan Li ^{a, b*}

a MOA Key Laboratory of Animal Virology & Zhejiang Provincial Engineering Research Center of Animal Biological Products, Zhejiang University College of Animal Sciences, Hangzhou, Zhejiang 310058, China

b Hainan Institute of Zhejiang University, Sanya, Hainan 572025, China

c Key Laboratory of Systems Health Science of Zhejiang Province, School of Life Science, Hangzhou Institute for Advanced Study, University of Chinese Academy of Sciences, Hangzhou, Zhejiang 310024, China

*Corresponding author: Yan Li, yanli3@zju.edu.cn

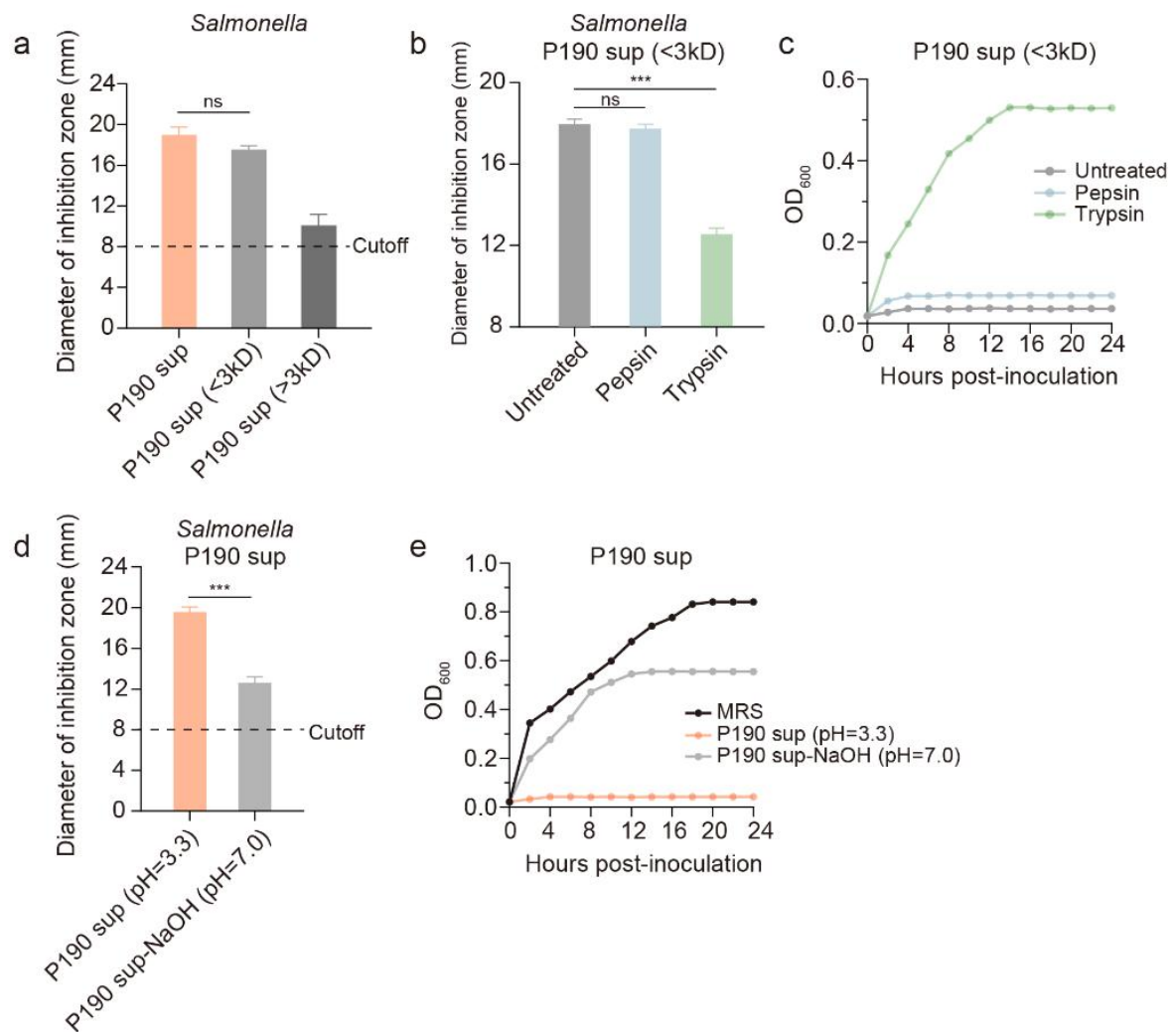


Figure S1. The antibacterial activity of *Limosilactobacillus reuteri* P190 supernatant resides in a small peptide fraction (<3 kDa) that is sensitive to protease digestion, and neutralization of organic acids partially reduces its activity. (a) Antimicrobial activity of *L. reuteri* P190 cell-free supernatant (P190 sup) and its <3 kDa and >3 kDa molecular weight fractions, against *S. Typhimurium* SL1344, determined by agar well diffusion assay. The dashed line indicates the threshold for detectable activity. (b) Inhibition zone diameters of the <3 kDa fraction of P190 sup, either untreated or pre-digested with pepsin or trypsin, against *S. Typhimurium* SL1344, determined by agar well diffusion assay. (c) Growth inhibition of *S. Typhimurium* SL1344 by the <3 kDa fraction of P190 sup, either untreated or pre-digested with pepsin or trypsin, as monitored by OD₆₀₀ measurements over 24 hours. (d) Antimicrobial activity of *L. reuteri* P190 cell-free supernatant (P190 sup) at its native pH (3.3) and following neutralization to pH 7.0 with NaOH, determined by agar well diffusion assay against *S. Typhimurium* SL1344. The dashed line indicates the threshold for detectable activity. (e) Growth inhibition of *S. Typhimurium* SL1344

by MRS medium, P190 sup (pH 3.3), and P190 sup neutralized to pH 7.0 with NaOH, monitored by OD₆₀₀ over 24 hours. Data are shown as the mean \pm SD. Statistical significance was assessed by one-way ANOVA. *** $p < 0.001$.

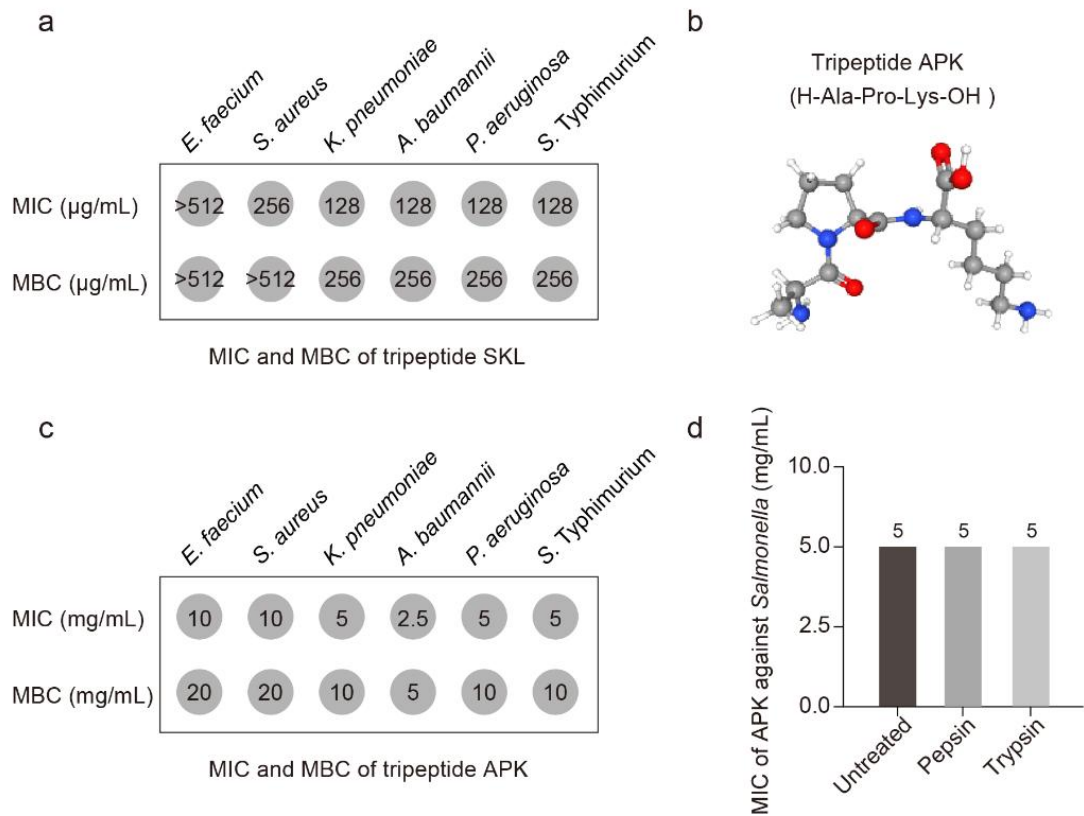


Figure S2. Antimicrobial activities of SKL and APK. The antimicrobial spectra of SKL and APK were assessed against a panel of clinically relevant pathogens, including *E. faecium*, *S. aureus*, *K. pneumoniae*, *A. baumannii*, *P. aeruginosa*, and *S. Typhimurium*. (a) Minimum inhibitory concentrations (MIC) and minimum bactericidal concentration (MBC) of SKL ($\mu\text{g/mL}$) against the indicated strains. (b) Chemical structure of APK (Ala-Pro-Lys). (c) MIC and MBC of APK (mg/mL) against the same pathogen panel. (d) MIC of APK against *S. Typhimurium* SL1344, determined either without treatment or after pre-digestion with pepsin or trypsin.

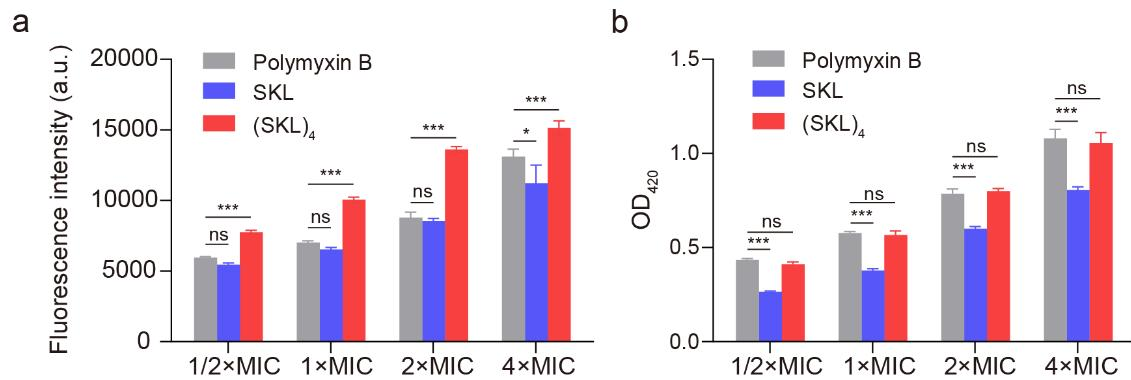


Figure S3. The membrane-disruptive effect of (SKL)₄ is comparable to that of polymyxin B against *Salmonella Typhimurium* SL1344. (a) Outer membrane permeability of *S. Typhimurium* SL1344 treated with polymyxin B, SKL, or (SKL)₄ at increasing concentrations (1/2x, 1x, 2x, 4x MIC), assessed by NPN fluorescence uptake assay. (b) Inner membrane permeability of *S. Typhimurium* SL1344 under the same treatment conditions, measured by monitoring the hydrolysis of ONPG (OD₄₂₀). Data are shown as the mean ± SD. Statistical significance was determined by two-way ANOVA. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; ns, not significant.

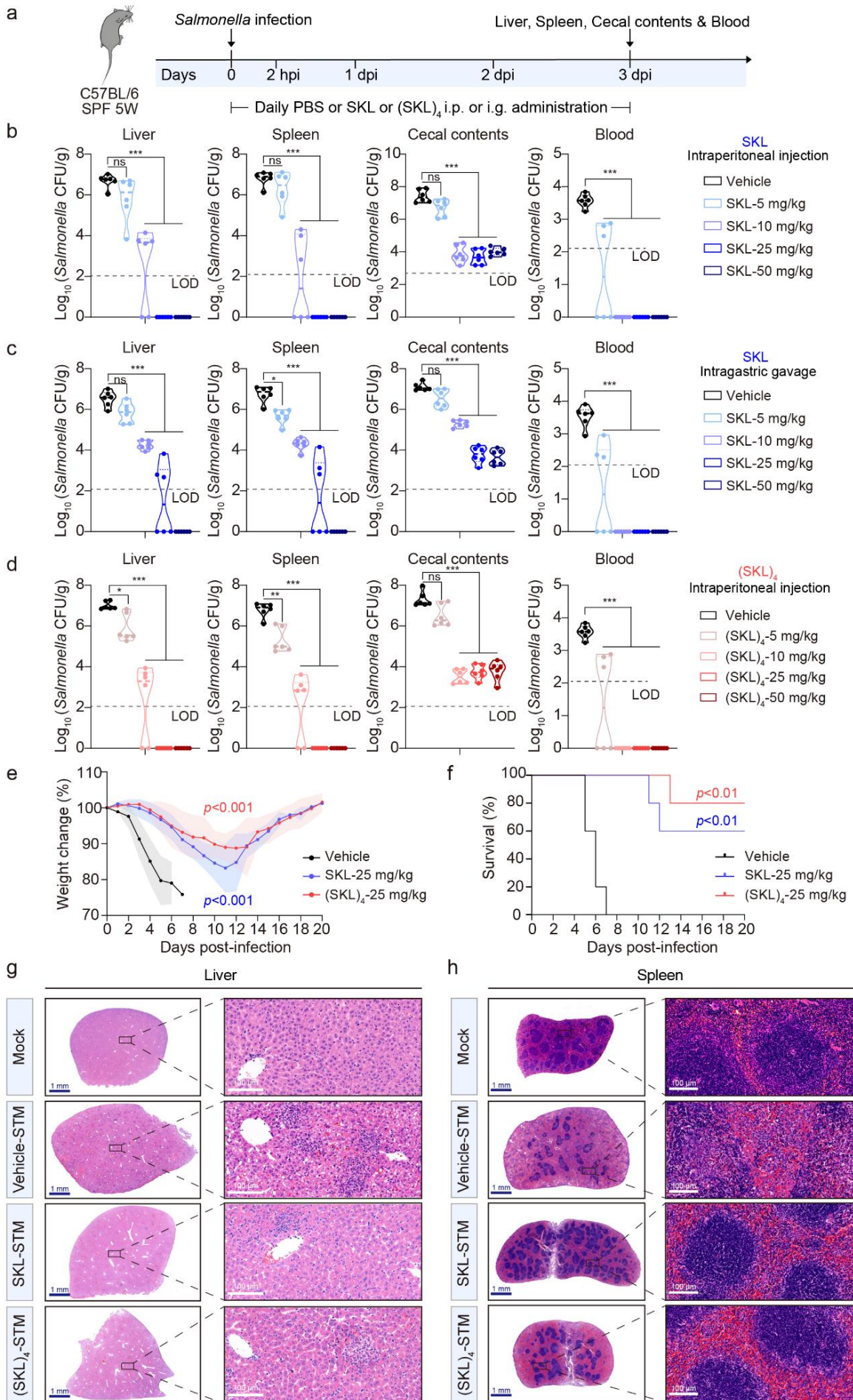


Figure S4. Dose optimization of SKL and (SKL)₄ demonstrates potent therapeutic efficacy against systemic *Salmonella* infection in immunocompetent mice via both intraperitoneal injection and oral administration. (a) Schematic experimental timeline for dose optimization in C57BL/6 mice. (b-d) Bacterial loads (CFU) in liver, spleen, cecal contents, and blood of mice treated with PBS vehicle or escalating doses (5, 10, 25, 50 mg/kg) of SKL via i.p. injection (b), SKL via intragastric gavage (c), or (SKL)₄ via i.p. injection (d) (n = 6 per group). The dashed line indicates the limit of detection (LOD). (e, f) Body weight changes (e) and survival rates (f) of mice infected with *S. Typhimurium* SL1344 and treated with PBS vehicle, SKL (25 mg/kg, i.p.), or (SKL)₄ (25 mg/kg, i.p.) (n = 5 per group). (g, h) Histological examination of H&E-stained liver (g) and spleen (h) sections from the mice. Mock group (uninfected, untreated) served as the healthy control. The vehicle-STM, SKL-STM and (SKL)₄-STM groups were infected with *S. Typhimurium* SL1344 and subsequently treated with PBS vehicle, SKL or (SKL)₄, respectively. Data are presented as the mean ± SD. Statistical significance for bacterial burdens and body weight changes was assessed using two-way ANOVA, whereas survival curves were analyzed using the log-rank (Mantel-Cox) test. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; ns, not significant.

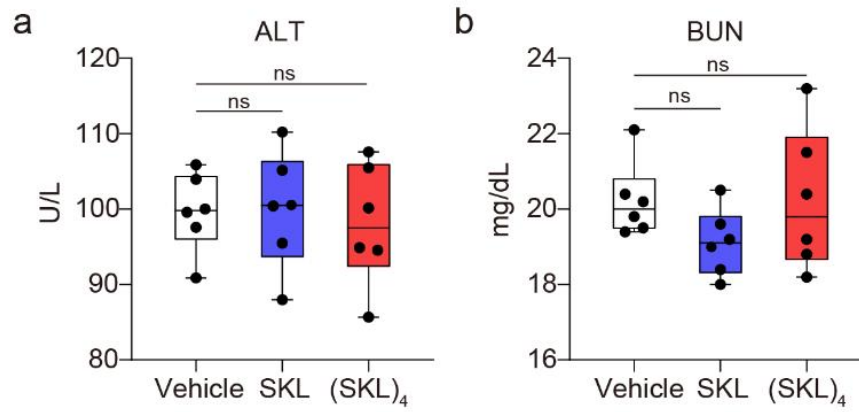


Figure S5. SKL and (SKL)₄ exhibit minimal hepatotoxicity and nephrotoxicity in C57BL/6 mice. (a) Serum alanine aminotransferase (ALT) levels (U/L) in mice treated with PBS vehicle, polymyxin B, SKL, or (SKL)₄ (n = 6 per group). (b) Blood urea nitrogen (BUN) levels (mg/dL) in mice treated with PBS vehicle, polymyxin B, SKL, or (SKL)₄ (n = 6 per group) under the same conditions. Data are shown as the mean ± SD. Statistical significance was determined by one-way ANOVA. ns, not significant.

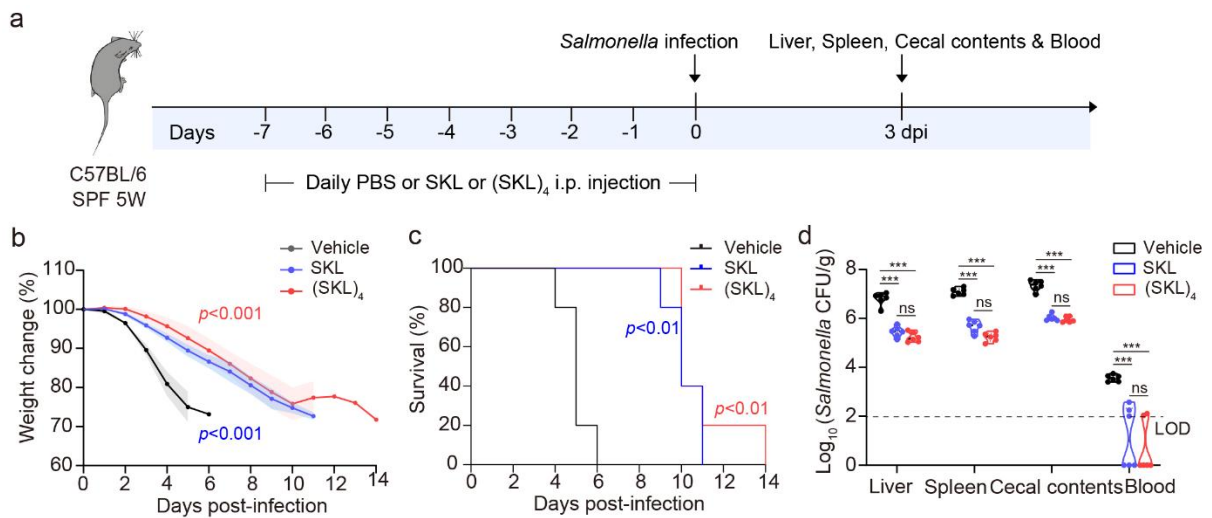


Figure S6. Prophylactic administration of SKL and (SKL)₄ provides protection against systemic *Salmonella* infection in mice. (a) Schematic experimental timeline for prophylactic administration. (b, c) Body weight changes (b) and Survival rates (c) of infected mice following pretreatment with PBS vehicle, SKL, or (SKL)₄ (n = 5 per group). (d) Bacterial loads (CFU) of *S. Typhimurium* SL1344 in liver, spleen, cecal contents, and blood at 3 dpi (n = 6 per group). The dashed line indicates the limit of detection (LOD). Data are shown as the mean ± SD. Statistical significance for body weight and bacterial loads was assessed by two-way ANOVA, whereas survival curves were analyzed using the log-rank (Mantel-Cox) test. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; ns, not significant.

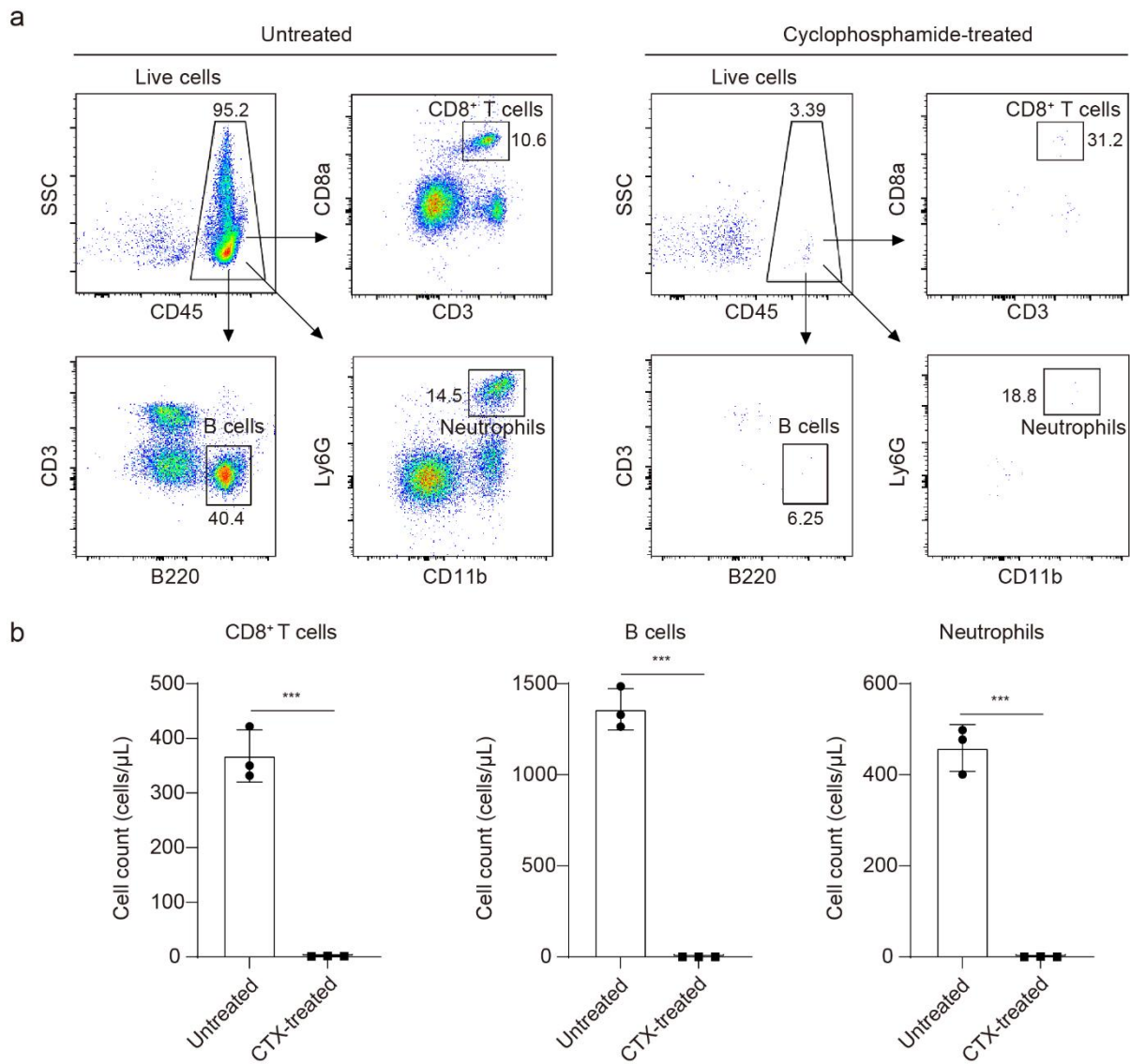


Figure S7. Cyclophosphamide induces depletion of immune cells in the peripheral blood of C57BL/6 mice. (a) Representative flow cytometry plots illustrating the gating strategy for identifying T cells, B cells, and neutrophils. (b) Quantification of immune cells. The data are shown as the means \pm SD ($n = 3$ per group). Significance is measured by the Welch t -test. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

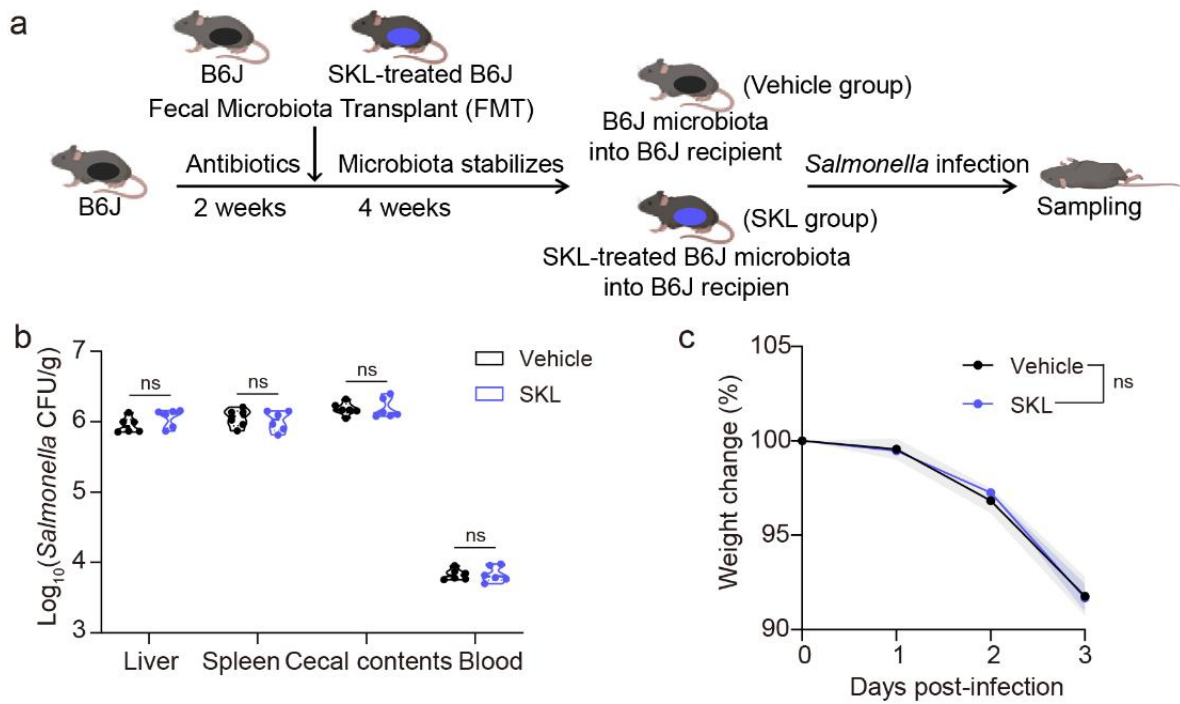


Figure S8. Fecal microbiota transplantation confirms that the *in vivo* efficacy of SKL is independent of the gut microbiota. (a) Schematic of the experimental design for fecal microbiota transplantation. (b) Bacterial loads (CFU) of *S. Typhimurium* SL1344 in liver, spleen, cecal contents, and blood of recipient mice that received fecal transplants from vehicle- or SKL-treated donors, assessed at 3 dpi (n = 6 per group). (c) Body weight changes (%) of recipient mice that received fecal transplants from vehicle- or SKL-treated donor microbiota following *S. Typhimurium* SL1344 infection (n = 10 per group). Data are shown as the mean \pm SD. Statistical significance was assessed by two-way ANOVA. ns, not significant.

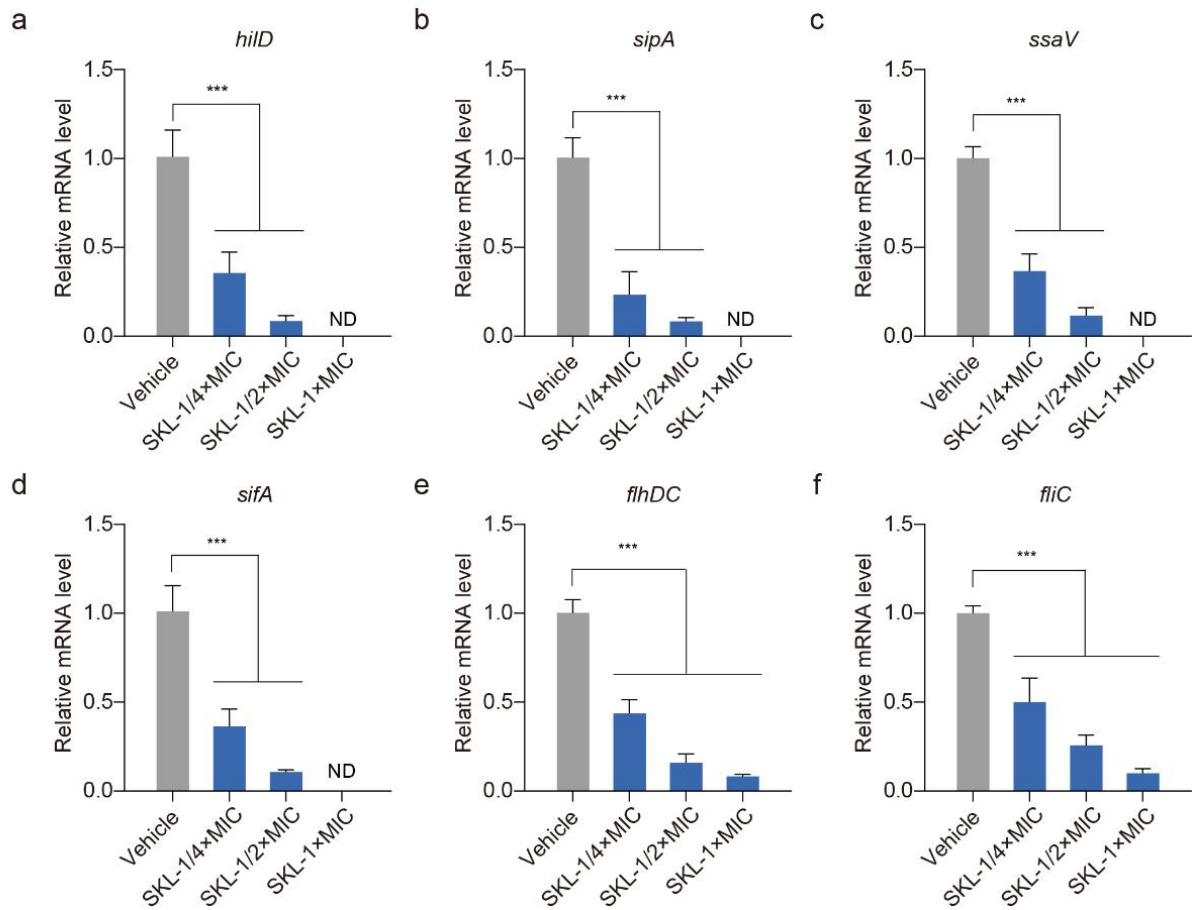


Figure S9. Dose-dependent suppression of *Salmonella* virulence gene expression by SKL. (a–f) qRT-PCR analysis of *hilD* (a), *sipA* (b), *ssaV* (c), *sifA* (d), *flhDC* (e), and *fliC* (f) mRNA levels in *S. Typhimurium* SL1344 treated with PBS vehicle, 1/4×MIC, 1/2×MIC, or 1×MIC SKL. Data are presented as mean ± SD (n = 6 per group). Statistical significance was assessed by one-way ANOVA. * p < 0.05; ** p < 0.01; *** p < 0.001; ND; undetectable.

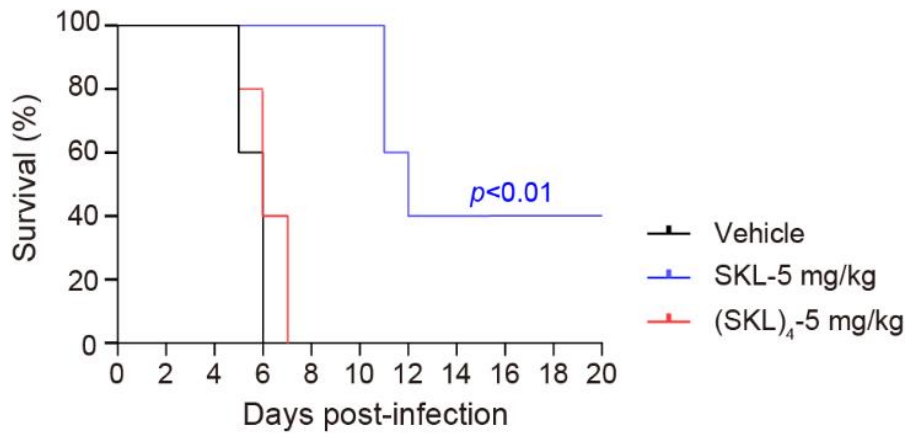


Figure S10. SKL improves the survival of *Salmonella*-infected mice at subinhibitory doses. Survival rates of mice treated with PBS vehicle, SKL (5 mg/kg, i.p.), or (SKL)₄ (5 mg/kg, i.p.) following *S. Typhimurium* SL1344 infection (n = 5 per group). Survival curves were analyzed using the log-rank (Mantel-Cox) test.

Supplemental Table S1: Antibacterial activity of 325 lactic acid bacterial (LAB) strains against Gram-negative *Salmonella* Typhimurium SL1344 and Gram-positive *Staphylococcus aureus* CMCC26003

Strain Code	Species Identification	Inhibition Zone Diameter (mm) against S. Typhimurium SL1344	Inhibition Zone Diameter (mm) against S. aureus CMCC26003
P001	<i>Enterococcus faecium</i>	8	8
P002	<i>Enterococcus faecium</i>	8	8
P003	<i>Enterococcus faecium</i>	8	8
P004	<i>Enterococcus faecium</i>	8	8
P005	<i>Pediococcus acidilactici</i>	9.2	8
P006	<i>Pediococcus acidilactici</i>	10.3	8
P007	<i>Pediococcus acidilactici</i>	8	8
P008	<i>Pediococcus acidilactici</i>	9.9	8
P009	<i>Pediococcus acidilactici</i>	8	8
P010	<i>Pediococcus acidilactici</i>	11.4	9.1
P011	<i>Enterococcus faecium</i>	8	8
P012	<i>Enterococcus faecium</i>	10.2	8
P013	<i>Enterococcus faecium</i>	8	8
P014	<i>Enterococcus faecium</i>	8	8
P015	<i>Enterococcus faecium</i>	8	8
P016	<i>Enterococcus faecium</i>	8	8
P017	<i>Pediococcus</i>	8	8

	<i>acidilactici</i>		
P018	<i>Pediococcus acidilactici</i>	10.6	9.2
P019	<i>Enterococcus faecium</i>	8	8
P020	<i>Pediococcus acidilactici</i>	8	8
P021	<i>Enterococcus faecium</i>	8	8
P022	<i>Lactobacillus paracasei</i>	11.2	9.2
P023	<i>Lactobacillus paracasei</i>	12.5	10.2
P024	<i>Lactobacillus paracasei</i>	8	8
P025	<i>Lactobacillus plantarum</i>	12.8	10.4
P026	<i>Lactobacillus paracasei</i>	8	8
P027	<i>Lactobacillus plantarum</i>	8	8
P028	<i>Lactobacillus paracasei</i>	13.2	11.8
P029	<i>Lactobacillus acidophilus</i>	14.5	12.4
P030	<i>Lactobacillus acidophilus</i>	8	8
P031	<i>Lactobacillus acidophilus</i>	8	8
P032	<i>Lactobacillus acidophilus</i>	8	8
P033	<i>Lactobacillus acidophilus</i>	11.4	8
P034	<i>Lactobacillus paracasei</i>	8	8
P035	<i>Lactobacillus paracasei</i>	8	8
P036	<i>Lactobacillus paracasei</i>	14.7	12.9
P037	<i>Lactobacillus paracasei</i>	13.5	11.24
P038	<i>Lactobacillus</i>	8	8

	<i>paracasei</i>		
P039	<i>Lactobacillus paracasei</i>	8	8
P040	<i>Lactobacillus paracasei</i>	12.4	10.1
P041	<i>Lactobacillus paracasei</i>	12	10
P042	<i>Lactobacillus plantarum</i>	8	8
P043	<i>Lactobacillus paracasei</i>	8	8
P044	<i>Lactobacillus rhamnosus</i>	15.8	13.7
P045	<i>Lactobacillus rhamnosus</i>	8	8
P046	<i>Lactobacillus rhamnosus</i>	8	8
P047	<i>Lactobacillus rhamnosus</i>	17.2	14.1
P048	<i>Lactobacillus rhamnosus</i>	8	8
P049	<i>Lactobacillus rhamnosus</i>	10.2	8
P050	<i>Lactobacillus rhamnosus</i>	11.6	8
P051	<i>Lactobacillus plantarum</i>	8	8
P052	<i>Enterococcus faecium</i>	8	8
P053	<i>Lactobacillus plantarum</i>	8	8
P054	<i>Lactobacillus rhamnosus</i>	9.2	8
P055	<i>Lactobacillus rhamnosus</i>	8	8
P056	<i>Lactobacillus rhamnosus</i>	9.7	8
P057	<i>Lactobacillus rhamnosus</i>	8	8
P058	<i>Lactobacillus rhamnosus</i>	8	8
P059	<i>Lactobacillus</i>	8	8

	<i>rhamnosus</i>		
P060	<i>Lactobacillus rhamnosus</i>	8	8
P061	<i>Lactobacillus rhamnosus</i>	8	8
P062	<i>Lactobacillus rhamnosus</i>	8	8
P063	<i>Lactobacillus rhamnosus</i>	8	8
P064	<i>Enterococcus faecium</i>	8	8
P065	<i>Enterococcus faecalis</i>	8	8
P066	<i>Enterococcus faecalis</i>	8	8
P067	<i>Enterococcus faecium</i>	8	8
P068	<i>Enterococcus faecium</i>	8	8
P069	<i>Enterococcus faecium</i>	8	8
P070	<i>Enterococcus faecalis</i>	8	8
P071	<i>Enterococcus faecalis</i>	8	8
P072	<i>Limosilactobacillus reuteri</i>	12.5	12.2
P073	<i>Enterococcus faecium</i>	8	8
P074	<i>Enterococcus faecium</i>	8	8
P075	<i>Enterococcus faecium</i>	8	8
P076	<i>Lactobacillus johnsonii</i>	8	8
P077	<i>Enterococcus faecium</i>	8	8
P078	<i>Enterococcus faecalis</i>	8	8
P079	<i>Enterococcus faecium</i>	8	8
P080	<i>Enterococcus</i>	8	8

	<i>faecalis</i>		
P081	<i>Limosilactobacillus reuteri</i>	13.5	12.4
P082	<i>Enterococcus faecium</i>	8	8
P083	<i>Enterococcus faecalis</i>	8	8
P084	<i>Limosilactobacillus reuteri</i>	11	8.8
P085	<i>Enterococcus faecium</i>	8	8
P086	<i>Enterococcus faecalis</i>	8	8
P087	<i>Lactobacillus salivarius</i>	8	8
P088	<i>Limosilactobacillus reuteri</i>	11.5	8.8
P089	<i>Enterococcus faecium</i>	8	8
P090	<i>Lactobacillus mucosae</i>	8	8
P091	<i>Lactobacillus johnsonii</i>	8	8
P092	<i>Lactobacillus johnsonii</i>	8	8
P093	<i>Lactobacillus vaginalis</i>	8	8
P094	<i>Lactobacillus crispatus</i>	15.1	14.5
P095	<i>Limosilactobacillus reuteri</i>	11.8	9.2
P096	<i>Enterococcus faecium</i>	8	8
P097	<i>Lactobacillus delbrueckii</i>	8	8
P098	<i>Limosilactobacillus reuteri</i>	13.2	11.3
P099	<i>Lactobacillus mucosae</i>	8	8
P100	<i>Enterococcus faecalis</i>	8	8
P101	<i>Enterococcus</i>	8	8

	<i>faecium</i>		
P102	<i>Limosilactobacillus reuteri</i>	12.7	11.2
P103	<i>Lactobacillus salivarius</i>	8	8
P104	<i>Lactobacillus vaginalis</i>	8	8
P105	<i>Lactobacillus mucosae</i>	8	8
P106	<i>Lactobacillus mucosae</i>	8	8
P107	<i>Lactobacillus mucosae</i>	8	8
P108	<i>Limosilactobacillus reuteri</i>	11	10.7
P109	<i>Lactobacillus mucosae</i>	8	8
P110	<i>Lactobacillus vaginalis</i>	8	8
P111	<i>Lactobacillus mucosae</i>	8	8
P112	<i>Limosilactobacillus reuteri</i>	12.5	11.1
P113	<i>Limosilactobacillus reuteri</i>	12.2	10.8
P114	<i>Enterococcus faecalis</i>	8	8
P115	<i>Lactobacillus mucosae</i>	8	8
P116	<i>Lactobacillus salivarius</i>	13.9	11.1
P117	<i>Lactobacillus mucosae</i>	8	8
P118	<i>Lactobacillus mucosae</i>	11.4	8
P119	<i>Lactobacillus mucosae</i>	10	8
P120	<i>Enterococcus faecium</i>	8	8
P121	<i>Enterococcus faecalis</i>	8	8
P122	<i>Enterococcus</i>	8	8

	<i>faecalis</i>		
P123	<i>Enterococcus faecium</i>	8	8
P124	<i>Enterococcus faecium</i>	8	8
P125	<i>Enterococcus faecalis</i>	8	8
P126	<i>Limosilactobacillus reuteri</i>	9.2	8
P127	<i>Enterococcus faecium</i>	8	8
P128	<i>Enterococcus faecalis</i>	8	8
P129	<i>Enterococcus faecium</i>	8	8
P130	<i>Enterococcus faecalis</i>	8	8
P131	<i>Enterococcus faecalis</i>	8	8
P132	<i>Enterococcus faecium</i>	8	8
P133	<i>Enterococcus faecalis</i>	8	8
P134	<i>Enterococcus faecalis</i>	8	8
P135	<i>Enterococcus faecium</i>	8	8
P136	<i>Enterococcus faecium</i>	8	8
P137	<i>Enterococcus faecium</i>	8	8
P138	<i>Enterococcus faecium</i>	8	8
P139	<i>Enterococcus faecium</i>	8	8
P140	<i>Enterococcus gallinarum</i>	8	8
P141	<i>Enterococcus faecium</i>	8	8
P142	<i>Enterococcus hirae</i>	8	8
P143	<i>Enterococcus gallinarum</i>	10.8	8

P144	<i>Enterococcus gallinarum</i>	8	8
P145	<i>Enterococcus faecium</i>	8	8
P146	<i>Enterococcus gallinarum</i>	8	8
P147	<i>Enterococcus faecium</i>	8	8
P148	<i>Enterococcus gallinarum</i>	8	8
P149	<i>Enterococcus faecium</i>	8	8
P150	<i>Enterococcus faecium</i>	10.4	8
P151	<i>Enterococcus gallinarum</i>	8	8
P152	<i>Enterococcus hirae</i>	8	8
P153	<i>Enterococcus faecalis</i>	8	8
P154	<i>Lactobacillus crispatus</i>	13.1	11
P155	<i>Weissella minor</i>	11.5	8
P156	<i>Limosilactobacillus reuteri</i>	11.8	9.4
P157	<i>Weissella minor</i>	10.5	8
P158	<i>Lactobacillus vaginalis</i>	8	8
P159	<i>Lactobacillus crispatus</i>	13.7	11.4
P160	<i>Lactobacillus salivarius</i>	14.6	13.4
P161	<i>Weissella minor</i>	8	8
P162	<i>Enterococcus faecium</i>	8	8
P163	<i>Lactobacillus crispatus</i>	8	8
P164	<i>Weissella minor</i>	10	8
P165	<i>Lactobacillus vaginalis</i>	8	8
P166	<i>Lactobacillus agilis</i>	14	12.1
P167	<i>Lactobacillus</i>	14	12.3

	<i>crispatus</i>		
P168	<i>Lactobacillus johnsonii</i>	10.9	8
P169	<i>Weissella minor</i>	11.8	8
P170	<i>Weissella minor</i>	11.7	8
P171	<i>Limosilactobacillus reuteri</i>	17.1	15
P172	<i>Limosilactobacillus reuteri</i>	15.8	12.4
P173	<i>Lactobacillus johnsonii</i>	8	8
P174	<i>Lactobacillus crispatus</i>	15.5	13.4
P175	<i>Weissella minor</i>	11.6	8
P176	<i>Weissella minor</i>	12.9	8
P177	<i>Weissella minor</i>	12.9	8
P178	<i>Weissella minor</i>	11.3	8
P179	<i>Enterococcus faecalis</i>	9.3	8
P180	<i>Lactobacillus salivarius</i>	11.7	8
P181	<i>Lactobacillus salivarius</i>	13.5	11.6
P182	<i>Lactobacillus salivarius</i>	12.8	9.6
P183	<i>Enterococcus faecalis</i>	8	8
P184	<i>Lactobacillus salivarius</i>	13	11.8
P185	<i>Enterococcus faecalis</i>	8	8
P186	<i>Enterococcus faecalis</i>	8	8
P187	<i>Enterococcus faecalis</i>	8	8
P188	<i>Lactobacillus salivarius</i>	12.2	10.3
P189	<i>Lactobacillus salivarius</i>	12.3	10
P190	<i>Limosilactobacillus reuteri</i>	20.2	15.6

P191	<i>Lactobacillus salivarius</i>	11.7	8
P192	<i>Lactobacillus salivarius</i>	12	10.2
P193	<i>Weissella minor</i>	9.9	8
P194	<i>Lactobacillus salivarius</i>	11.5	8
P195	<i>Limosilactobacillus reuteri</i>	16.2	12.8
P196	<i>Enterococcus faecalis</i>	8	8
P197	<i>Lactobacillus salivarius</i>	11.8	8
P198	<i>Lactobacillus crispatus</i>	11.8	8
P199	<i>Lactobacillus crispatus</i>	11.9	8
P200	<i>Pediococcus pentosaceus</i>	11.3	8
P201	<i>Lactobacillus agilis</i>	12.5	9.2
P202	<i>Pediococcus pentosaceus</i>	11	8
P203	<i>Lactobacillus salivarius</i>	14.8	11.4
P204	<i>Lactobacillus agilis</i>	13.8	10.1
P205	<i>Lactobacillus salivarius</i>	15.5	13.4
P206	<i>Lactobacillus agilis</i>	14.1	11.7
P207	<i>Pediococcus pentosaceus</i>	12.8	9.8
P208	<i>Lactobacillus agilis</i>	15.3	12.72
P209	<i>Lactobacillus salivarius</i>	15.8	13.24
P210	<i>Limosilactobacillus reuteri</i>	14.8	12.7
P211	<i>Limosilactobacillus reuteri</i>	12.5	10.1
P212	<i>Lactobacillus johnsonii</i>	8	8
P213	<i>Lactobacillus salivarius</i>	18.6	15.4

P214	<i>Lactobacillus plantarum</i>	18.6	16.2
P215	<i>Enterococcus faecium</i>	13.6	10
P216	<i>Enterococcus faecium</i>	13	10.1
P217	<i>Enterococcus faecium</i>	13.5	10
P218	<i>Enterococcus faecalis</i>	12.2	9.2
P219	<i>Enterococcus faecalis</i>	12	8
P220	<i>Enterococcus faecalis</i>	10.6	8
P221	<i>Enterococcus faecalis</i>	11.4	9.2
P222	<i>Enterococcus faecalis</i>	11.6	9.4
P223	<i>Enterococcus faecium</i>	13	11
P224	<i>Enterococcus faecium</i>	8	8
P225	<i>Enterococcus faecalis</i>	11	8
P226	<i>Enterococcus faecalis</i>	11.7	9.4
P227	<i>Enterococcus faecium</i>	10.6	8
P228	<i>Enterococcus faecium</i>	11.1	9.6
P229	<i>Enterococcus faecium</i>	8	8
P230	<i>Enterococcus faecium</i>	8	8
P231	<i>Enterococcus faecium</i>	8	8
P232	<i>Enterococcus faecium</i>	8	8
P233	<i>Enterococcus faecalis</i>	8	8
P234	<i>Enterococcus faecalis</i>	8	8

P235	<i>Enterococcus faecium</i>	8	8
P236	<i>Enterococcus faecium</i>	8	8
P237	<i>Pediococcus pentosaceus</i>	13.7	11.4
P238	<i>Enterococcus faecium</i>	8	8
P239	<i>Enterococcus faecalis</i>	8	8
P240	<i>Pediococcus pentosaceus</i>	17.1	14.2
P241	<i>Pediococcus pentosaceus</i>	12.7	9.8
P242	<i>Enterococcus faecalis</i>	8	8
P243	<i>Pediococcus pentosaceus</i>	12.9	8.9
P244	<i>Enterococcus faecium</i>	12	8
P245	<i>Enterococcus faecium</i>	8	8
P246	<i>Enterococcus faecalis</i>	8	8
P247	<i>Lactobacillus salivarius</i>	17.5	14.6
P248	<i>Lactobacillus salivarius</i>	17.9	13.8
P249	<i>Enterococcus faecalis</i>	8	8
P250	<i>Lactobacillus agilis</i>	17.8	16
P251	<i>Lactobacillus salivarius</i>	19.8	16.8
P252	<i>Enterococcus faecalis</i>	8	8
P253	<i>Lactobacillus agilis</i>	16	13.8
P254	<i>Lactobacillus salivarius</i>	19.9	17.1
P255	<i>Enterococcus faecalis</i>	8	8
P256	<i>Lactobacillus salivarius</i>	19.6	16.3

P257	<i>Lactobacillus agilis</i>	15.1	12.4
P258	<i>Lactobacillus agilis</i>	15.5	12.4
P259	<i>Lactobacillus agilis</i>	15.3	12.5
P260	<i>Lactobacillus salivarius</i>	19.6	16.4
P261	<i>Lactobacillus mucosae</i>	8	8
P262	<i>Enterococcus faecalis</i>	12.2	8
P263	<i>Enterococcus faecalis</i>	8	8
P264	<i>Lactobacillus mucosae</i>	8	8
P265	<i>Lactobacillus salivarius</i>	19	15.3
P266	<i>Enterococcus faecium</i>	8	8
P267	<i>Lactobacillus johnsonii</i>	8	8
P268	<i>Limosilactobacillus reuteri</i>	16.4	13.4
P269	<i>Lactobacillus johnsonii</i>	8	8
P270	<i>Limosilactobacillus reuteri</i>	17.1	12.8
P271	<i>Limosilactobacillus reuteri</i>	17.2	13.8
P272	<i>Lactobacillus johnsonii</i>	8	8
P273	<i>Limosilactobacillus reuteri</i>	17.3	15.1
P274	<i>Lactobacillus johnsonii</i>	8	8
P275	<i>Lactobacillus delbrueckii</i>	8	8
P276	<i>Lactobacillus salivarius</i>	18.5	15.7
P277	<i>Lactobacillus johnsonii</i>	8	8
P278	<i>Limosilactobacillus reuteri</i>	12.1	10.1
P279	<i>Limosilactobacillus</i>	17.1	14.2

	<i>reuteri</i>		
P280	<i>Lactobacillus vaginalis</i>	8	8
P281	<i>Lactobacillus vaginalis</i>	8	8
P282	<i>Lactobacillus crispatus</i>	16	13.2
P283	<i>Enterococcus faecalis</i>	8	8
P284	<i>Lactobacillus salivarius</i>	19.3	16.7
P285	<i>Lactobacillus salivarius</i>	17.9	14.3
P286	<i>Enterococcus faecalis</i>	8	8
P287	<i>Enterococcus faecalis</i>	8	8
P288	<i>Lactobacillus salivarius</i>	19.9	16.2
P289	<i>Limosilactobacillus reuteri</i>	13.5	10.9
P290	<i>Enterococcus faecalis</i>	8	8
P291	<i>Lactobacillus salivarius</i>	18.6	14.5
P292	<i>Lactobacillus salivarius</i>	18.8	13.8
P293	<i>Enterococcus faecalis</i>	8	8
P294	<i>Lactobacillus salivarius</i>	19.4	16.7
P295	<i>Lactobacillus salivarius</i>	18.4	15.4
P296	<i>Lactobacillus salivarius</i>	17.9	14
P297	<i>Lactobacillus johnsonii</i>	8	8
P298	<i>Pediococcus pentosaceus</i>	8	8
P299	<i>Enterococcus faecalis</i>	8	8
P300	<i>Limosilactobacillus</i>	13.8	11.6

	<i>reuteri</i>		
P301	<i>Lactobacillus plantarum</i>	13.5	12.1
P302	<i>Lactobacillus salivarius</i>	18.4	16.2
P303	<i>Enterococcus faecium</i>	8	8
P304	<i>Enterococcus faecalis</i>	8	8
P305	<i>Lactobacillus crispatus</i>	15.4	12.5
P306	<i>Lactobacillus crispatus</i>	15.5	12.4
P307	<i>Lactobacillus johnsonii</i>	8	8
P308	<i>Lactobacillus salivarius</i>	17.4	15.2
P309	<i>Enterococcus faecium</i>	8	8
P310	<i>Enterococcus faecium</i>	11.4	8
P311	<i>Enterococcus faecalis</i>	8	8
P312	<i>Enterococcus faecalis</i>	8	8
P313	<i>Enterococcus faecium</i>	8	8
P314	<i>Enterococcus faecium</i>	8	8
P315	<i>Enterococcus faecium</i>	8	8
P316	<i>Enterococcus faecium</i>	8	8
P317	<i>Enterococcus faecalis</i>	8	8
P318	<i>Enterococcus faecium</i>	8	8
P319	<i>Enterococcus faecium</i>	8	8
P320	<i>Enterococcus faecalis</i>	8	8
P321	<i>Pediococcus</i>	8	8

	<i>pentosaceus</i>		
P322	<i>Pediococcus pentosaceus</i>	13	10.1
P323	<i>Enterococcus faecalis</i>	8	8
P324	<i>Enterococcus faecium</i>	8	8
P325	<i>Enterococcus faecium</i>	8	8

Notes: ① The antibacterial activity was determined by the agar well diffusion assay using Oxford cups (inner diameter = 8 mm). ② Inhibition zone diameters were measured to the nearest 0.1 mm

Supplemental Table S3. Primer Sequences Used in Quantitative Real-Time Polymerase Chain Reaction

Primers	Forward/Reverse	Sequences (5' to 3')
<i>16S rRNA</i>	Forward	CAGCCACACTGGAAGTGA
	Reverse	GTGCTTCTTCTGCGGGTAAC
<i>qseC</i>	Forward	AATTGACGCAACGTCTCAGCC
	Reverse	ACGGCCTCAAATCCACCTTCC
<i>phoP</i>	Forward	ACGGCGTTTGAATACACCAT
	Reverse	TGACATCGTGCGGATACTGG
<i>lrp</i>	Forward	TCGATCGTAACATTCTTAAT
	Reverse	TGACGCATCCAGATAATGCG
<i>cpxA</i>	Forward	GCTGCTGAAGAAGGTGAAGG
	Reverse	TTCTGCTGCTGTTGCTGTTG
<i>hilD</i>	Forward	GAGATACCGACGCAACGACT
	Reverse	TTGGTTTGCTGCTCGTTTGG
<i>sipA</i>	Forward	TTCGACTAACAGCAGCA
	Reverse	CCGTCGTACCGGCTTTATTA
<i>ssaV</i>	Forward	AGTCGCAATGCGTTCATGGTTAG
	Reverse	TTCTTCATTGTCCGCCAACTC
<i>sifA</i>	Forward	TTAAAAAGTGAAATCCTTACCAACTCC
	Reverse	C
<i>flhDC</i>	Forward	TCCGCTTTTGCTTTGCCAGTAGAA
	Reverse	GTCAAACCGGAAATGACAAACTAA
<i>fliC</i>	Forward	ACCCTGCCGCAGATGGT
	Reverse	CGCGAAAGACGATGCGGC
	Forward	CAGAAATCTGCGCGCGAGACAT
	Reverse	