Supplementary Material

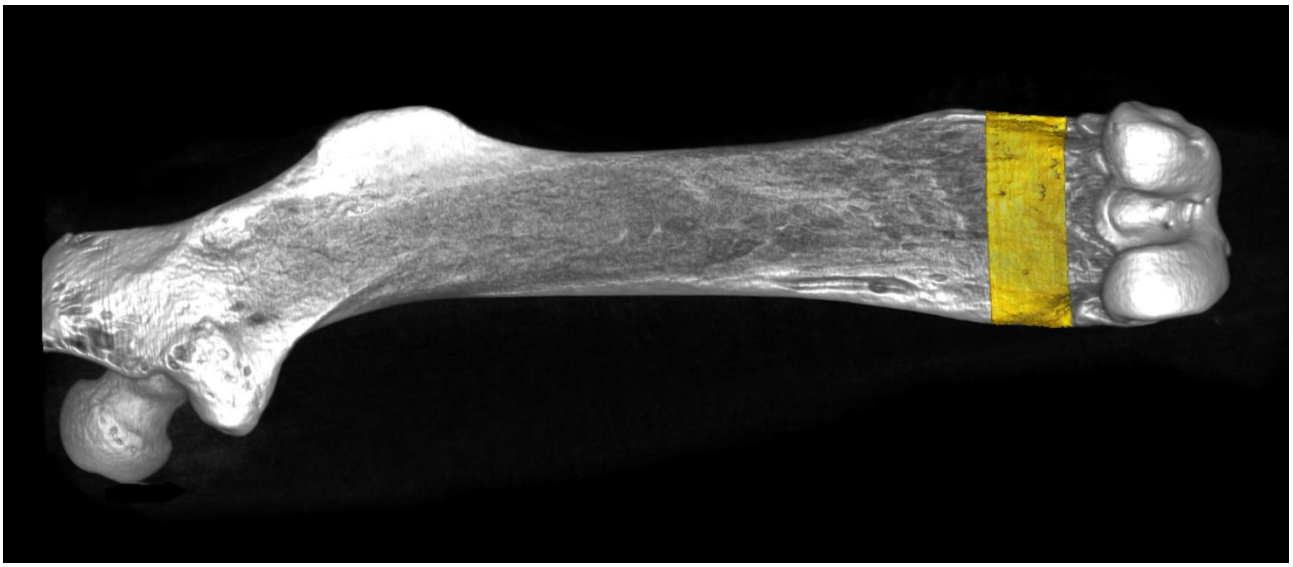

Figure S1. Micro-CT image showing the ROI of the femur in the coronal plane. The yellow zone represents the ROI of the femur used for bone microarchitecture assessment. ROI, region of interest; micro-CT, micro-computed tomography.

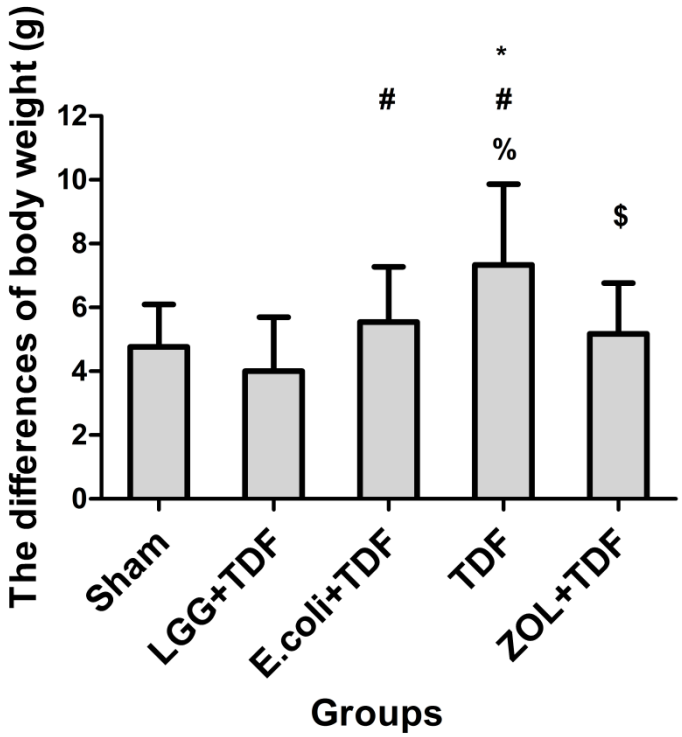

Figure S2. Differences of body weight between baseline and 8 weeks. N = 10–14 mice per group. All data were normally distributed according to the Shapiro-Wilk normality test and analyzed using 2-way ANOVA and post hoc tests applying the LSD correction for multiple comparisons.[*] $p < 0.05$ compared with the Sham group; [#] $p < 0.05$ compared with the LGG+TDF group; [%] $p < 0.05$ compared with the *E. coli*+TDF group; [$] $p < 0.05$ compared with the TDF group.

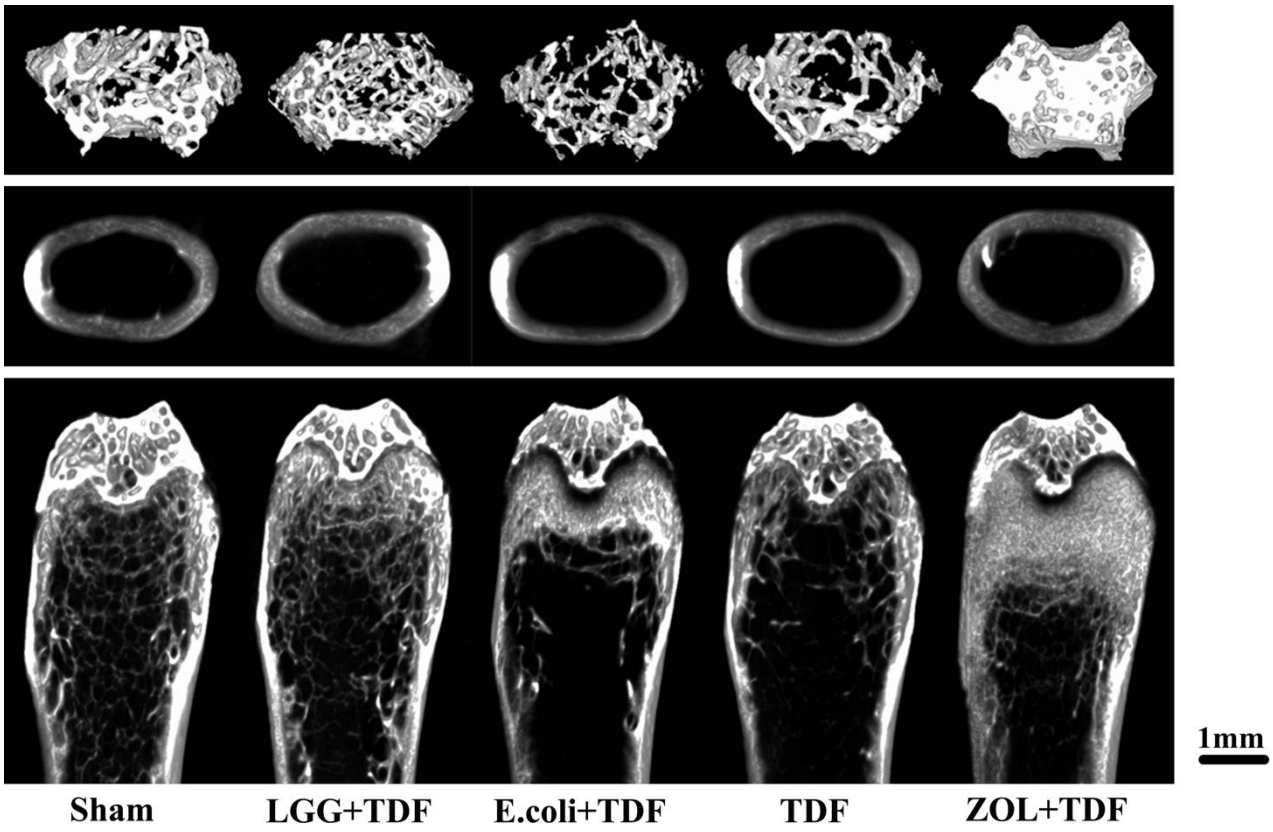

Figure S3. Representative images of the comparison of the bone microarchitecture reconstructed using micro-CT at 4 weeks. Trabecular bone in BM in horizontal plane, cortical bone in horizontal plane, and femur in coronal plane were exhibited. BM, bone marrow; micro-computed tomography.

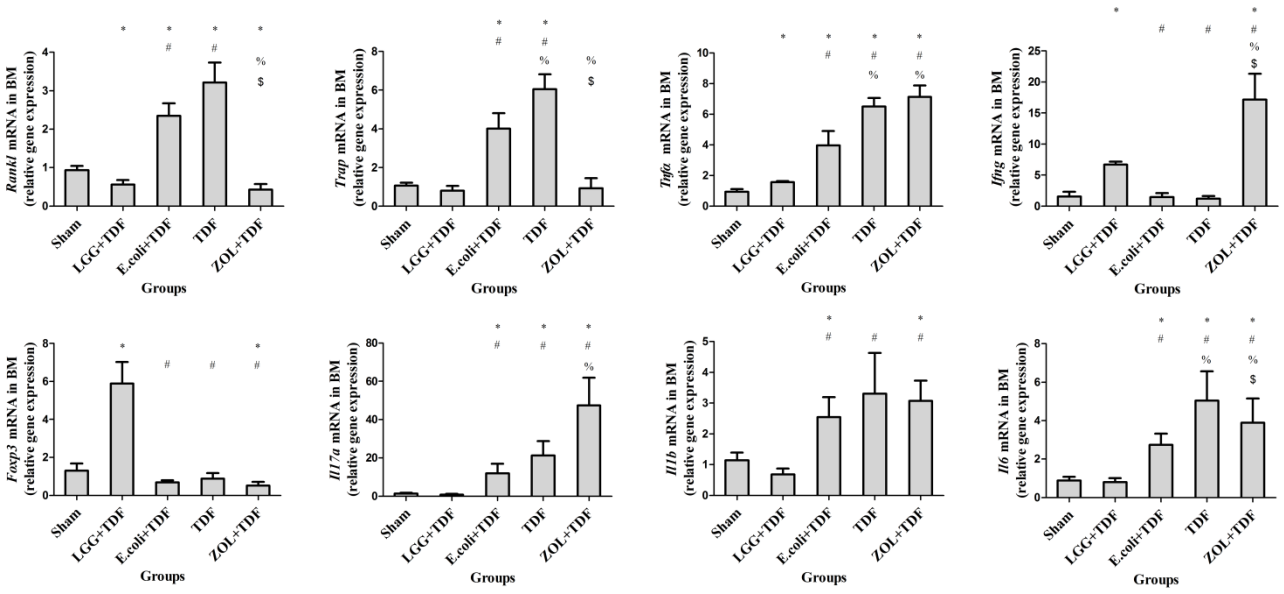

Figure S4. Analysis of the transcript levels of osteoclast markers and proinflammatory cytokines in BM using qPCR. The mRNA levels of *Rankl*, *Trap*, *Tnfα*, *Ifng*, *Foxp3*, *Il17a*, *Il1b*, and *Il6* were detected from BM-derived cells. N = 6–7 mice per group in all panels. All data were normally distributed according to the Shapiro-Wilk normality test and analyzed using 2-way ANOVA and post hoc tests applying the LSD correction for multiple comparisons.[*] $p < 0.05$ compared with the Sham group; [#] $p < 0.05$ compared with the LGG+TDF group; [%] $p < 0.05$ compared with the *E. coli*+TDF group; [$] $p < 0.05$ compared with the TDF group. *Trap*, tartrate-resistant acid phosphatase; *Foxp3*, forkhead box P3.

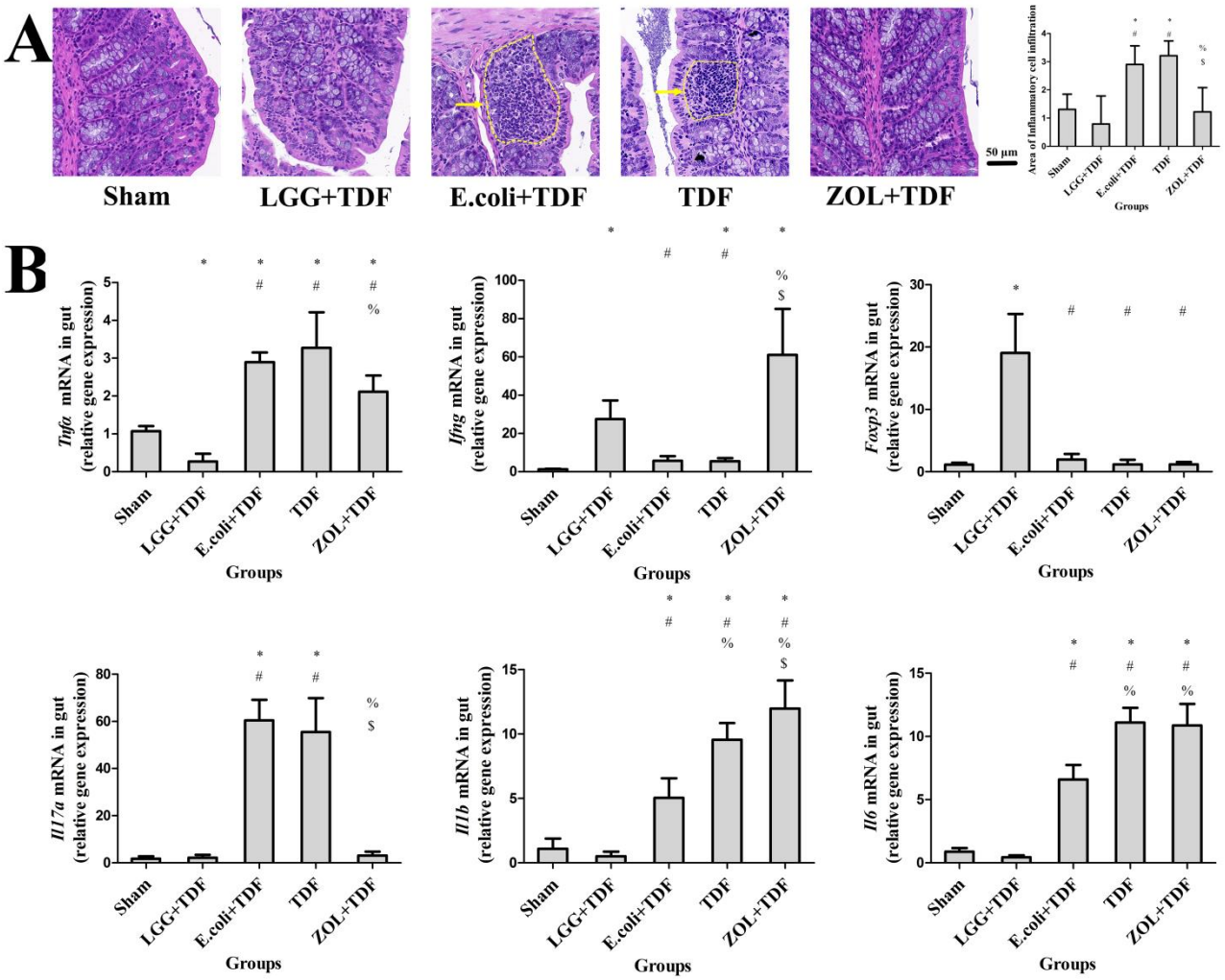

Figure S5. Inflammatory cell infiltration and proinflammatory cytokines in the gut. (**A**) Representative images and quantification of the area of inflammatory cell infiltration in intestinal mucosa from intestinal slices stained usin hematoxylin and eosin (H&E) in the five groups (20.0×). N = 6 mice per group in the panel. Yellow dotted lines represent the zone of inflammatory cell infiltration instead of the intestinal gland in intestinal mucosa. (**B**) Quantitative real-time PCR (qPCR) analysis measuring transcript levels of proinflammatory cytokines in the gut. The mRNA levels of *Tnfα*, *Ifng*, *Foxp3*, *Il17a*, *Il1b*, and *Il6* were detected from the intestinal tissue of the five groups. N = 6 mice per group in all panels. All data were normally distributed according to the Shapiro-Wilk normality test and analyzed using 2-way ANOVA and post hoc tests applying the LSD correction for multiple comparisons.* $p < 0.05$ compared with the Sham group; $^{\#}$ $p < 0.05$ compared with the LGG+TDF group; $^{\%}$ $p < 0.05$ compared with the *E. coli*+TDF group; $^{\$}$ $p < 0.05$

compared with the TDF group. *Foxp3*, forkhead box P3; *Tnfα*, tumor necrosis factor alpha; *Ifng*, interferon gamma; *Il17a*, interleukin 17a; *Il1b*, intereukin 1; *Il6*, interleukin 6.

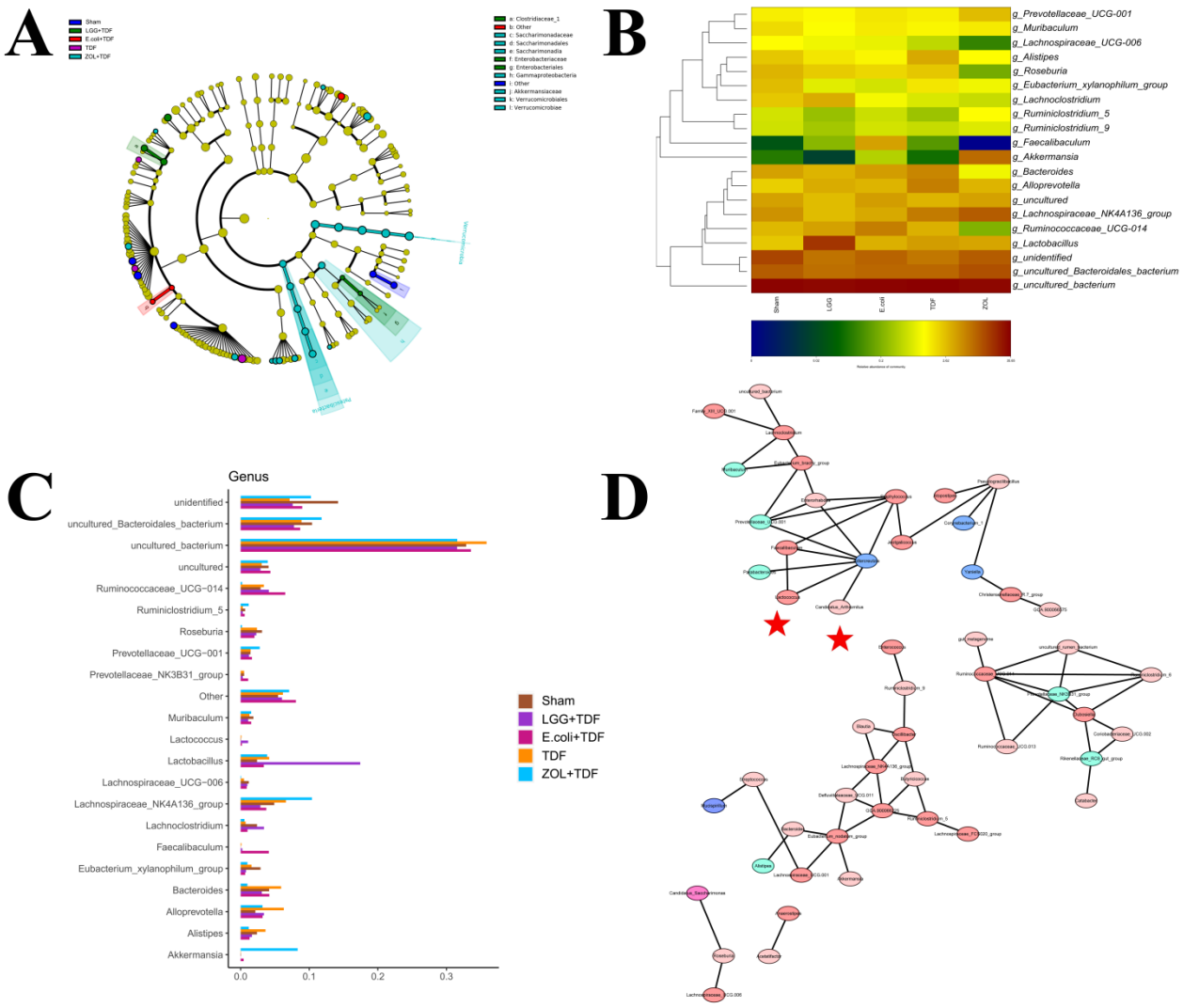

Figure S6. Comparison of the intestinal community structure in the five groups at 8 weeks. (**A**) Cladogram for the taxonomic representation of the significant differences among the five groups. (**B**) A heatmap based on taxonomy and species components analysis at the genus level. (**C**) Distribution of the predominant bacteria at the genus level. (**D**) A network diagram showing the associations of the dominant genera. A red star represents that *Lactococcus* is positive linked with *Candidatus_Arthromitus* in the highly abundant species of the LGG group. LGG, *Lactobacillus*

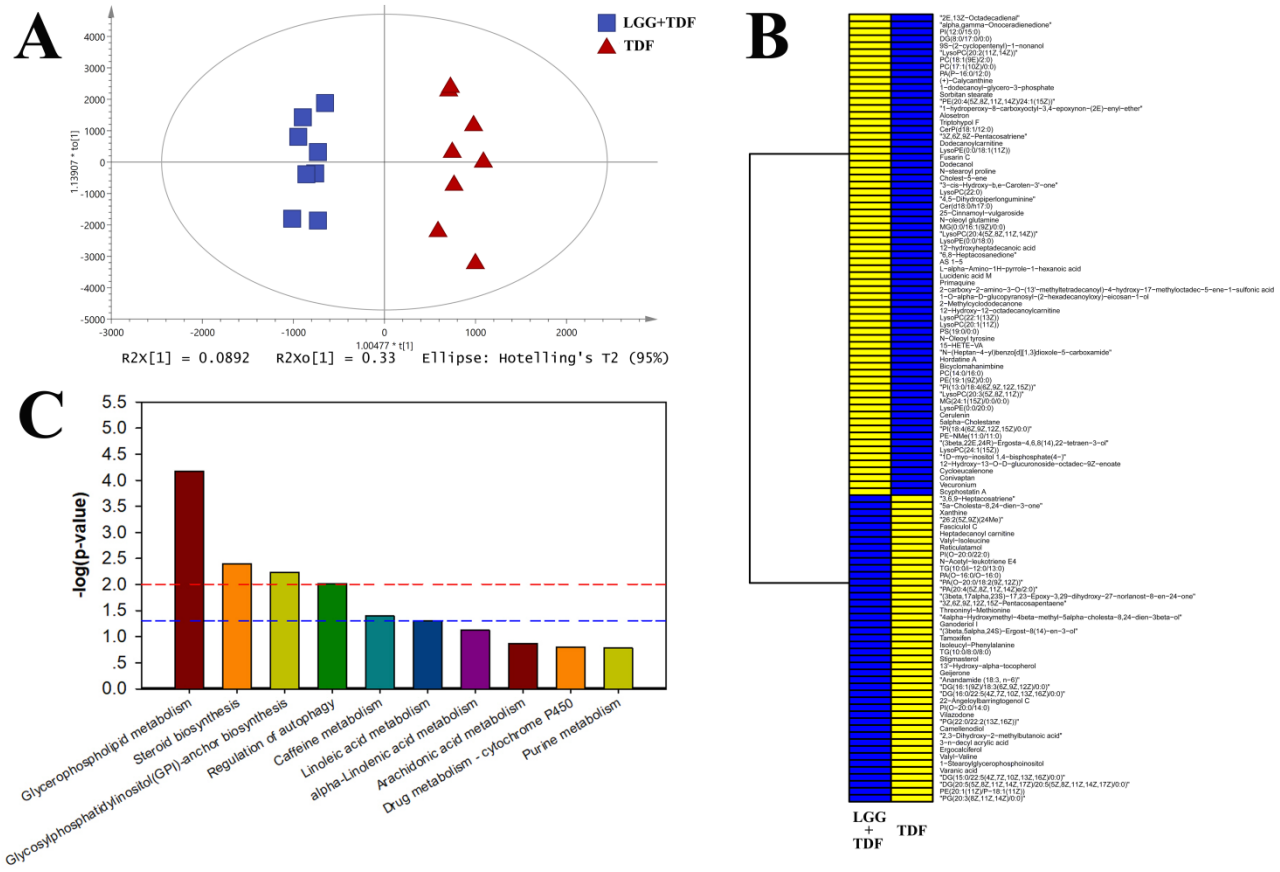

Figure S7. LGG reconstructed the intestinal metabolites composition. (**A**) OPLS-DA scores plot of subjects from the LGG and TDF groups based on the feces spectral data of UPLC–Q-TOF/MS. (**B**) Heatmap showing 116 differentially expressed metabolites based on a VIP > 1.0 between the LGG and TDF groups. The yellow box represents relatively highly expressed metabolites between two groups and the blue box represents relatively lowly expressed metabolites between two groups. (**C**) The representative metabolic pathways screened through the KEGG database. The upper red dashed line represents significantly different metabolic pathways between two groups ($p < 0.01$). The upper blue dashed line represents significantly different metabolic pathways between two groups ($p < 0.05$). LGG, *Lactobacillus rhamnosus GG*; OPLS-DA, orthogonal partial least-squares-discriminant analysis; TDF, tenofovir disoproxil fumarate; UPLC–Q-TOF/MS, ultra performance liquid chromatography–quadrupole time-of-flight mass spectrometry; VIP, Variable importance in the projection; KEGG, Kyoto Encyclopedia of Genes and Genomes.

Table S1. Sequences of the primers used for quantitative real-time reverse transcription PCR (qRT-PCR).

	Forward primer (5′ to 3′)	**Reverse primer (5′-3′)**
Rankl	GCAGCATCGCTCTGTTCCTGTA	CCTGCAGGAGTCAGGTAGTGTGTC
Trap	CAGCAGCCAAGGAGGACTAC	ACATAGCCCACACCGTTCTC
Tnfα	AACTCCAGGCGGTGCCTAT	TGCCACAAGCAGGAATGAGA
Ifng	GGTCCAGCGCCAAGCAT	GCTGGATTCCGGCAACAG
Foxp3	GGCCCTTCTCCAGGACAGA	GCTGATCATGGCTGGGTTGT
Il17a	TCAGCGTGTCCAAACACTGAG	CGCCAAGGGAGTTAAAGACTT
Il1b	GCCCATCCTCTGTGACTCAT	AGGCCACAGGTATTTTGTCG
Il6	CCATCCAGTTGCCTTCTTGG	TTTCTGCAAGTGCATCATCG
Cldn2	TCTCAGCCCTGTTTTCTTTGG	GGCGAGCAGGAAAAGCAA
Cldn3	TCATCACGGCGCAGATCA	CTCTGCACCACGCAGTTCA
Cldn15	GGCGGCATCTGTGTCTTCTC	TGGTGGCTGGTTCCTCCTT

Foxp3, forkhead box P3; *Tnfα*, tumor necrosis factor alpha; *Ifng*, interferon gamma; *Il17a*, interleukin 17a; *Il1b*, intereukin 1; *Il6*, interleukin 6; *Rankl*, receptor activator of nuclear factor kappa-B ligand; *Trap*, tartrate-resistant acidic phosphatase; *Cldn*2, claudin 2; *Cldn*3, claudin 3; *Cldn*15, claudin 15.

Table S2. BMD and bone histomorphometry for the centrum and mandible in the five groups at 8 weeks (n=10-12)

Groups	Sham	LGG+TDF	*E. coli*+TDF	TDF	ZOL+TDF
Centrum:					
Ma.BMD (mg/cm^3)	656.821 ± 36.164	739.518 ± 30.606*	582.242 ± 17.532$^{*,\#}$	605.48 ± 29.492$^{*,\#}$	754.64 ± 47.419$^{*,\%,\$}$
BV/TV (%)	34.11 ± 2.855	39.269 ± 3.876*	26.067 ± 1.503$^{*,\#}$	27.746 ± 1.254$^{*,\#}$	36.568 ± 3.136$^{\%,\$}$
BS/BV (mm^{-1})	41.204 ± 2.781	37.282 ± 3.538	46.644 ± 3.368$^{*,\#}$	44.505 ± 0.981$^{\#}$	41.572 ± 3.273$^{\#,\%}$
Tb.Th (mm)	0.049 ± 0.003	0.054 ± 0.005*	0.043 ± 0.003$^{*,\#}$	0.045 ± 0.001$^{\#}$	0.048 ± 0.004$^{\#,\%}$
Tb.N (mm^{-1})	7.001 ± 0.309	7.269 ± 0.115	6.066 ± 0.329$^{*,\#}$	6.17 ± 0.153$^{*,\#}$	7.448 ± 0.112$^{\%,\$}$
Tb.Sp (mm)	0.094 ± 0.007	0.084 ± 0.006*	0.122 ± 0.008$^{*,\#}$	0.117 ± 0.005$^{*,\#}$	0.084 ± 0.006$^{*,\%,\$}$
Mandible:					
BMD in alveolar bone (mg/cm^3)	1366.035 ± 72.504	1404.075 ± 29.549	1158.191 ± 48.43$^{*,\#}$	1240.548 ± 81.715$^{*,\#}$	1563.073 ± 81.857$^{*,\#,\%,\$}$
BMD in mandibular angle (mg/cm^3)	2557.638 ± 54.444	2558.554 ± 36.681	2369.966 ± 100.499$^{*,\#}$	2259.621 ± 53.824$^{*,\#,\%}$	2524.011 ± 27.551$^{\%,\$}$

Parameters on BMD and bone microarchitecture were measured in the L4 centrum and mandible by micro-CT at 8 weeks. Data are expressed as mean ± SD. All data were normally distributed according to the Shapiro-Wilk normality test and analyzed using 2-way ANOVA and post hoc tests applying the LSD correction for multiple comparisons.* $p < 0.05$ compared with the Sham group; $^{\#}$ $p < 0.05$ compared with the LGG+TDF group; $^{\%}$ $p < 0.05$ compared with the *E. coli*+TDF group; $^{\$}$ $p < 0.05$ compared with the TDF group.

Ma.BMD, bone mineral density in bone marrow; BV/TV, bone volume/total volume; BS/BV, bone surface area/bone volume; Tb.Th, trabecular thickness; Tb.N, trabecular number; Tb.Sp, trabecular separation.

Table S3. BMD and bone histomorphometry for femurs in the five groups at 4 weeks (n =10–12)

Groups	Sham	LGG+TDF	*E. coli*+TDF	TDF	ZOL+TDF
Ma.BMD (mg/cm^3)	737.255 ± 31.907	769.977 ± 62.472	502.017 ± 16.411$^{*,\#}$	533.662 ± 24.35$^{*,\#}$	1371.32 ± 51.994$^{*,\#,\%,\$}$
Ma.BMC (mg)	1.735 ± 0.191	1.784 ± 0.206	1.179 ± 0.153$^{*,\#}$	1.305 ± 0.11$^{*,\#}$	3.671 ± 0.249$^{*,\#,\%,\$}$
%Ct.Ar (%)	26.506 ± 1.598	26.967 ± 2.598	22.077 ± 3.152$^{*,\#}$	20.942 ± 1.607$^{*,\#}$	29.388 ± 2.139$^{\%,\$}$
BV/TV (%)	33.538 ± 4.407	35.95 ± 3.075	13.921 ± 1.327$^{*,\#}$	18.375 ± 2.588$^{*,\#}$	95.363 ± 2.651$^{*,\#,\%,\$}$
BS/BV (mm-1)	41.175 ± 6.562	35.907 ± 5.455	57.65 ± 4.618$^{*,\#}$	52.534 ± 5.19$^{*,\#}$	9.581 ± 1.53$^{*,\#,\%,\$}$
Tb.Th (mm)	0.055 ± 0.011	0.053 ± 0.007	0.033 ± 0.002	0.038 ± 0.002	0.208 ± 0.03$^{*,\#,\%,\$}$
Tb.N (mm-1)	6.852 ± 0.404	6.773 ± 0.426	4.222 ± 0.34$^{*,\#}$	4.87 ± 0.258$^{*,\#,\%}$	4.719 ± 0.6$^{*,\#}$
Tb.Sp (mm)	0.093 ± 0.01	0.092 ± 0.006	0.208 ± 0.017$^{*,\#}$	0.165 ± 0.013$^{*,\#,\%}$	0.01 ± 0.004$^{*,\#,\%,\$}$

Parameters on BMD and bone microarchitecture were measured in the femur by micro-CT at 4 weeks. Data are expressed as mean ± SD. All data were normally distributed according to the Shapiro-Wilk normality test and analyzed using 2-way ANOVA and post hoc tests applying the LSD correction for multiple comparisons.* $p < 0.05$ compared with the Sham group; $^{\#}$ $p < 0.05$ compared with the LGG+TDF group; $^{\%}$ $p < 0.05$ compared with the *E. coli*+TDF group; $^{\$}$ $p < 0.05$ compared with the TDF group.

Ma.BMD, bone mineral density in bone marrow; Ma.BMC, bone mineral content in bone marrow; %Ct.Ar, cortical bone area/total bone area; BV/TV, bone volume/total volume; BS/BV, bone surface area/bone volume; Tb.Th, trabecular thickness; Tb.N, trabecular number; Tb.Sp, trabecular separation.

Table S4. Alpha diversity indices for feces of mice in the five groups at 8 weeks at 97% identity (n = 4)

Groups	Sham	LGG+TDF	E.coli+TDF	TDF	ZOL+TDF
Chao1	382.09 ± 13.387	339.845 ± 21.323*	388.843 ± 21.987$^{\#}$	386.928 ± 15.062$^{\#}$	265.002 ± 6.797$^{*,\#,\%,\$}$
goods_coverage	0.99817 ± 0.0001	0.99807 ± 0.00029	0.99808 ± 0.00021	0.9981 ± 0.00042	0.99876 ± 0.00008$^{*,\#,\%,\$}$
observed_species	342.625 ± 13.877	294.75 ± 25.261*	340.7 ± 14.703$^{\#}$	346.5 ± 22.456$^{\#}$	239.425 ± 6.439$^{*,\%,\$}$
Shannon	6.139 ± 0.335	5.272 ± 0.735*	5.846 ± 0.214	6.027 ± 0.434$^{\#}$	5.582 ± 0.12

Data are expressed as the mean ± SD. All data were normally distributed according to the Shapiro-Wilk normality test and analyzed using 2-way ANOVA and post hoc tests applying the LSD correction for multiple comparisons.* $p < 0.05$ compared with the Sham group; $^{\#}$ $p < 0.05$ compared with the LGG+TDF group; $^{\%}$ $p < 0.05$ compared with the *E. coli*+TDF group; $^{\$}$ $p < 0.05$ compared with the TDF group.