

Supplementary Data

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Grape seed proanthocyanidin extract ameliorates dextran sulfate sodium-induced colitis through intestinal barrier improvement, oxidative stress reduction, and inflammatory cytokines and gut microbiota modulation

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Table S1 Disease activity index (DAI) scoring system

DAI score	Weight loss (%)	Stool consistency	Occult/gross bleeding
0	None	Normal	Normal
1	1-5		
2	6-10	Loose stools	Hemoccult positive
3	11-18		
4	>18	Diarrhea	Gross bleeding

Table S2 Pathological scores system

Score	Inflammation	Mucosal damage	Crypt damage	Range of lesions(%)
0	None	None	None	0
1	Mild	Mucous layer	1/3	1-25
2	Moderate	submucosa	2/3	26-50
3	Severe	Muscularis and serosa	100%	51-75
4	-	-	100%+epithelium loss	76-100

Table S3 Oligonucleotide primers used for RT-qPCR

Gene	5'-3' Primer sequence
TNF- α	F: ATGAGAAGTTCCCAAATGGC R: CTCCACTTGGTGGTTGCTA
IL-1 β	F: TGCCACCTTTGACAGTGATG R: AAGGTCCACGGAAAGACAC
IL-10	F: GGACCAGCTGGACAACATACTGCTA R: CCGATAAGGCTTGGCAACCCAAGT
NLRP3	F: TGGAAGATTACCCGCCGAGAA R: TCCCAGCAAACCCATCCACTCTTC
Caspase-1	F: TGAAAGACAAGCCAAGGTGATC R: CAATGAAAAGTGAGCCCCTGAC
ASC	F: CATTTGTCTTGGCTGGTGGTCT R: CGGACACGGACAGGATTGACA
ZO-1	F: TCTTCCATCATTCGCTGTGT R: TCTGAAACCATCAAGTCCACA
Occludin	F: TCACTTTCTGCAGGTGACT R: GGGAACGTGGCCGATATAATG
Claudin	F: ATGCAAAGATGTTTGCACAG R: TACAAATTCCCATTGCAGCCC
β -actin	F: TGTCCACCTTCCAGCAGATGT R: AGCTCAGTAACAGTCCGCCTAGA

Table S4 Significantly different phyla altered by the treatment of GSPE in mice

Species name	Relative abundance					
	Control-Mean(%)	Control-SD(%)	DSS-Mean(%)	DSS-SD(%)	GSPE-Mean(%)	GSPE-SD(%)
p_Firmicutes	67.41	2.548	37.41	5.41	40.6	5.88
p_Bacteroidetes	20.24	3.322	42.11	6.32	30.37	4.53
p_Proteobacteria	7.757	2.392	8.942	17.1	12.62	10.12
p_Actinobacteria	2.575	1.035	5.249	3.081	4.045	3.942
p_Epsilonbacteraeota	0.7992	0.9475	0.6497	1.018	5.867	8.902
p_Patescibacteria	0.5947	0.5286	4.748	6.573	0.8259	1.109
p_Verrucomicrobia	0.3568	0.4053	0.01834	0.02322	5.104	1.13

Data are expressed as mean ± standard deviation, n=5.

Table S5 Significantly different families altered by the treatment of GSPE in mice

Species name	Relative abundance					
	Control-Mean(%)	Control-SD(%)	DSS-Mean(%)	DSS-SD(%)	GSPE-Mean(%)	GSPE-SD(%)
f_Muribaculaceae	12.7	4.186	34.38	2.21	14.61	1.787
f_Staphylococcaceae	36.25	21.59	0.443	0.9334	5.566	12.43
f_Erysipelotrichaceae	9.505	8.707	13.7	6.701	16.21	11.22
f_Lactobacillaceae	3.655	4.261	12.7	7.597	3.717	4.668
f_Lachnospiraceae	5.317	8.45	3.189	2.876	5.625	6.364
f_Burkholderiaceae	1.983	2.727	3.484	5.931	8.539	6.946
f_Prevotellaceae	3.211	2.796	3.321	2.395	5.329	4.339
f_Ruminococcaceae	3.572	0.308	2.62	0.232	5.051	0.497
f_Bacteroidaceae	2.345	2.553	2.496	4.181	6.224	6.711
f_Enterobacteriaceae	2.299	3.506	4.45	9.905	2.748	3.668
f_Helicobacteraceae	0.7992	0.9475	0.6497	1.018	5.867	8.902
f_Peptostreptococcaceae	3.923	3.068	0.4291	0.7438	1.821	1.046
f_Saccharimonadaceae	0.5947	0.5286	4.748	6.573	0.8259	1.109
f_Akkermansiaceae	0.3568	0.4053	0.01834	0.02322	5.104	10.13
f_Rikenellaceae	1.319	0.9512	0.9938	0.453	2.117	1.554
f_Bifidobacteriaceae	0.6503	0.8399	1.53	2.944	1.935	2.028
f_Enterococcaceae	2.267	4.242	1.304	2.813	5.104	0.13
f_Atopobiaceae	0.2618	0.4611	1.782	0.5845	1.37	1.968
f_Veillonellaceae	0.7792	1.722	2.05	4.58	0.2718	0.5661
f_Desulfovibrionaceae	0.777	1.103	0.9727	1.321	1.13	0.8294
f_Eggerthellaceae	0.4697	0.1344	1.925	1.592	0.3668	0.2966
f_Moraxellaceae	2.091	2.449	0.001112	0.002486	0.03168	0.06622
f_Marinifilaceae	0.2462	0.4501	0.2446	0.2504	1.239	1.798

Data are expressed as mean ± standard deviation, n=5.

Table S6 Significantly different genera altered by the treatment of GSPE in mice

Species name	Relative abundance					
	Control-Mean(%)	Control-SD(%)	DSS-Mean(%)	DSS-SD(%)	GSPE-Mean(%)	GSPE-SD(%)
g_norank_f_Muribaculaceae	12.68	4.185	34.05	20.87	14.61	8.786
g_Staphylococcus	33.81	20.01	0.3613	0.7739	5.116	11.43
g_Lactobacillus	3.655	0.426	12.7	0.759	3.717	0.4668
g_Dubosiella	5.158	0.5327	9.103	1.89	4.109	0.4311
g_Ileibacterium	2.647	3.847	2.71	5.384	9.286	9.105
g_Parasutterella	1.684	2.875	3.462	5.937	8.476	7.004
g_Bacteroides	2.345	2.553	2.496	4.181	6.224	6.711
g_Escherichia-Shigella	2.294	3.505	4.437	9.909	2.745	3.67
g_Helicobacter	0.7992	0.9475	0.6497	1.018	5.867	8.902
g_Lachnospiraceae_NK4A136_group	3.992	7.554	1.082	1.45	1.685	2.231
g_Romboutsia	3.923	3.068	0.4291	0.7438	1.821	1.046
g_Candidatus_Saccharimonas	0.5947	0.5286	4.748	6.573	0.8259	1.109
g_Prevotellaceae_UCG-001	1.855	2.167	1.671	1.481	2.26	1.744
g_Akkermansia	0.3568	0.4053	0.01834	0.02322	5.104	1.013
g_unclassified_f_Prevotellaceae	0.8782	1.034	1.246	1.923	2.587	2.65
g_Turicibacter	0.9754	0.5342	1.282	2.12	1.869	2.669
g_Bifidobacterium	0.6503	0.8399	1.53	2.944	1.935	2.028
g_Enterococcus	2.267	4.242	1.303	2.811	0.5213	0.6562
g_Ruminococcaceae_UCG-014	1.542	2.517	1.304	1.443	1.189	1.927
g_Coriobacteriaceae_UCG-002	0.2618	0.4611	1.782	0.5845	1.37	1.968
g_Veillonella	0.7792	0.022	2.049	0.458	0.2712	0.056
g_[Eubacterium]_fissicatena_group	0.05669	0.08198	0.7003	1.403	1.976	3.667
g_Jeotgalicoccus	2.185	1.592	0.08059	0.1603	0.4191	0.9355
g_unclassified_f_Lachnospiraceae	0.3818	0.5913	0.8693	0.7995	1.094	1.991
g_Alistipes	0.6853	0.7585	0.438	0.4871	1.063	0.9597
g_Psychrobacter	2.09	2.45	0	0	0.02946	0.06587
g_norank_f_Desulfovibrionaceae	0.4969	0.8733	0.6119	1.139	1.009	0.8207
g_Enterorhabdus	0.3135	0.1	1.307	1.134	0.2496	0.2249
g_Odoribacter	0.2379	0.4467	0.2234	0.2274	1.233	1.801
g_unclassified_f_Ruminococcaceae	0.3946	0.4502	0.4235	0.4915	0.6925	0.4877
g_Ruminiclostridium_5	0.3741	0.5224	0.04002	0.04208	1.029	1.423
g_norank_f_Muribaculaceae	12.68	4.185	34.05	20.87	14.61	8.786
g_Staphylococcus	33.81	20.01	0.3613	0.7739	5.116	11.43

Data are expressed as mean ± standard deviation, n=5.