

Differences in Microbiota Membership along the Gastrointestinal Tract of Piglets and their Differential Alterations Following an Early-Life Antibiotic Intervention

Chunlong Mu¹, Yuxiang Yang¹, Yong Su¹, Erwin G. Zoetendal^{1,2}, Weiyun Zhu^{1*}

¹Laboratory of Gastrointestinal Microbiology, College of Animal Science and Technology, Nanjing Agricultural University, Nanjing, China

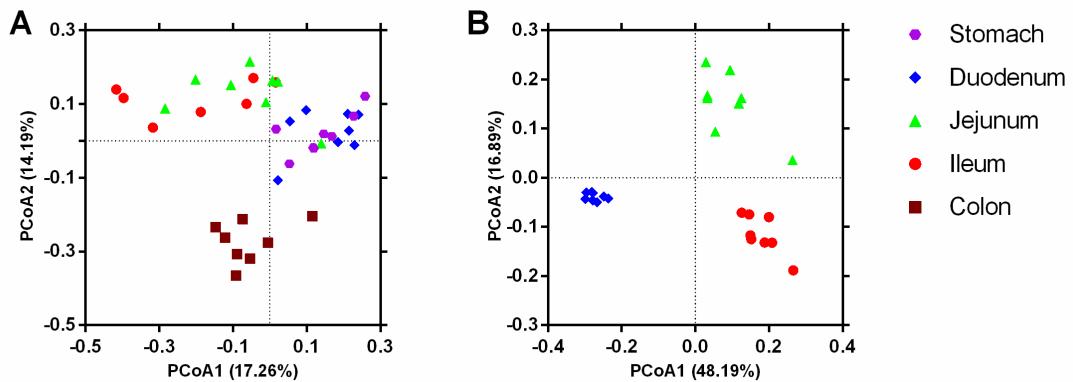
²Laboratory of Microbiology, Wageningen University, Wageningen, The Netherlands

Supplementary material

Summary

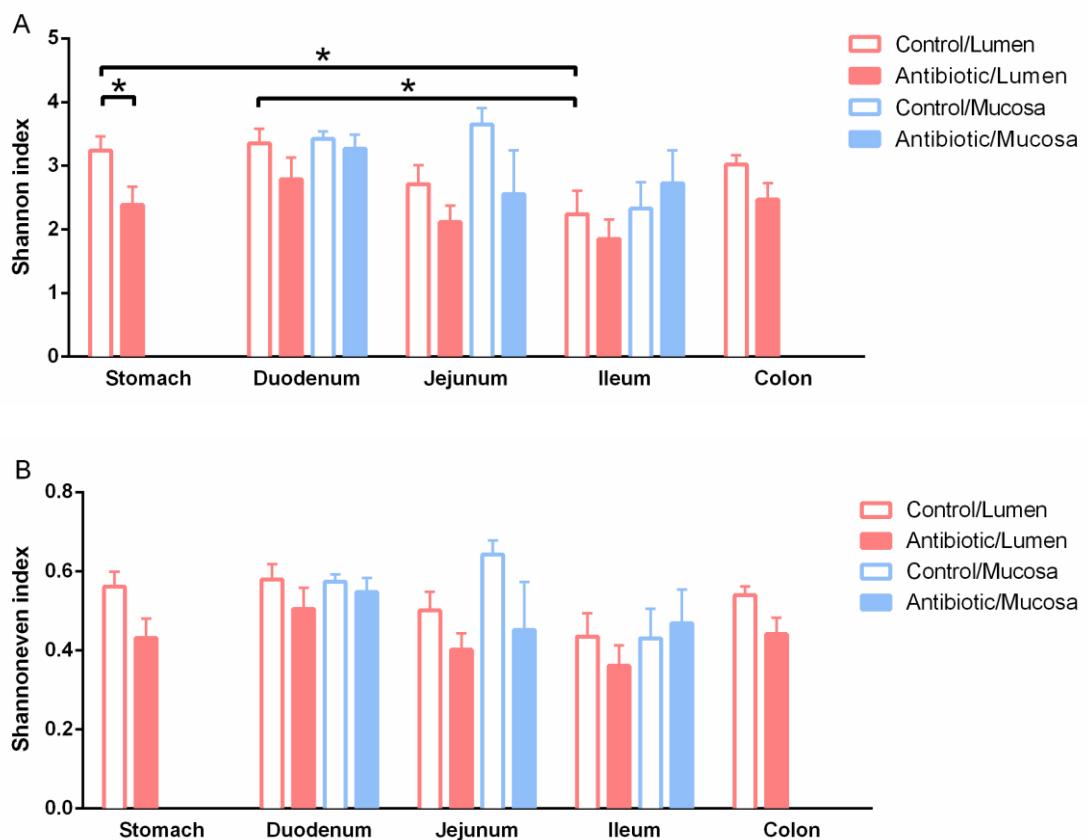
The supporting information includes five supplementary figures and nine supplementary tables.

Figure S1. Principle coordinate analysis plot



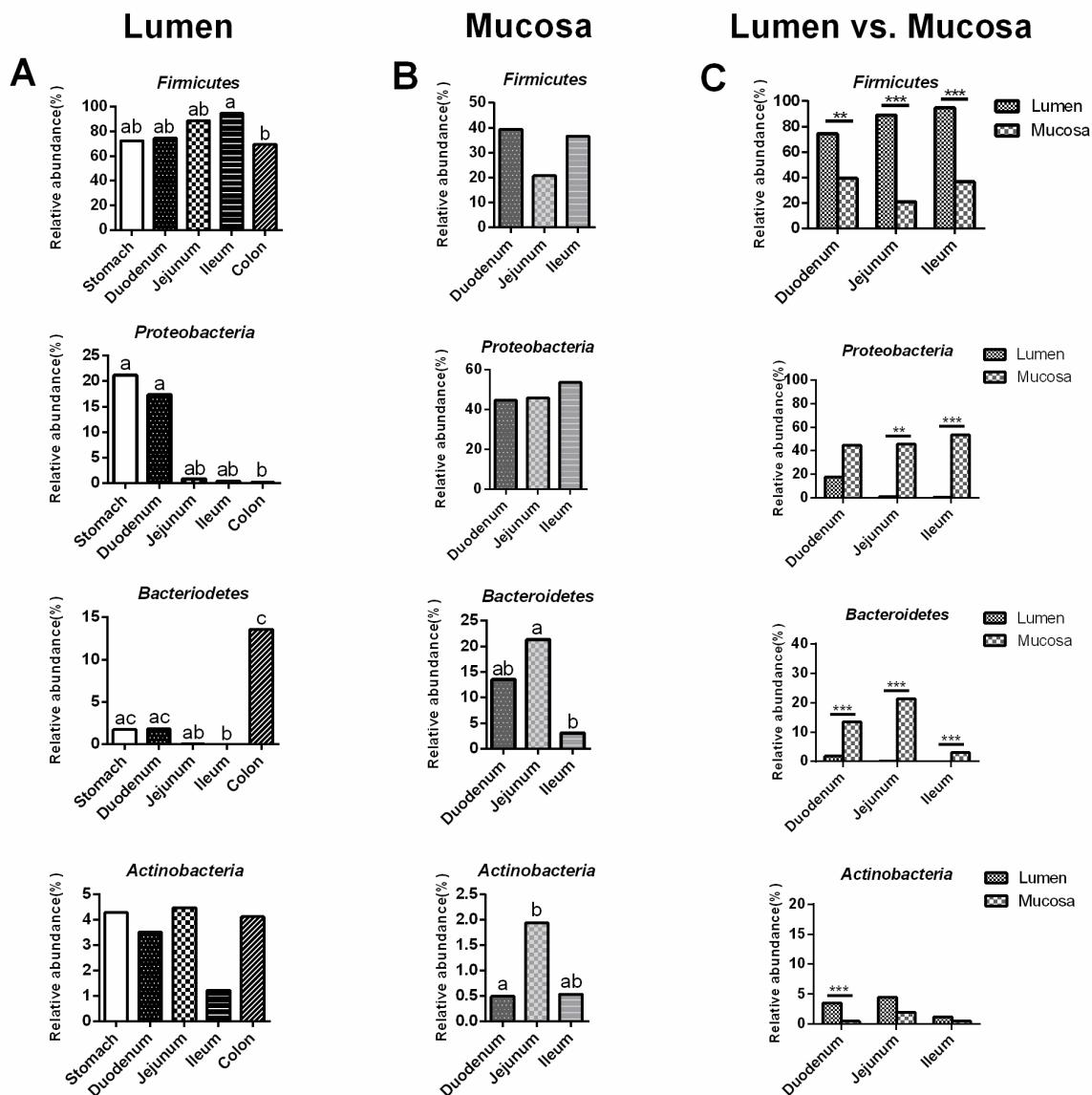
Principle coordinate analysis of all samples including luminal samples in control group (**A**) and mucosal samples in control group (**B**) by unweighted Unifrac distance.

Figure S2. Alpha-diversity index plot



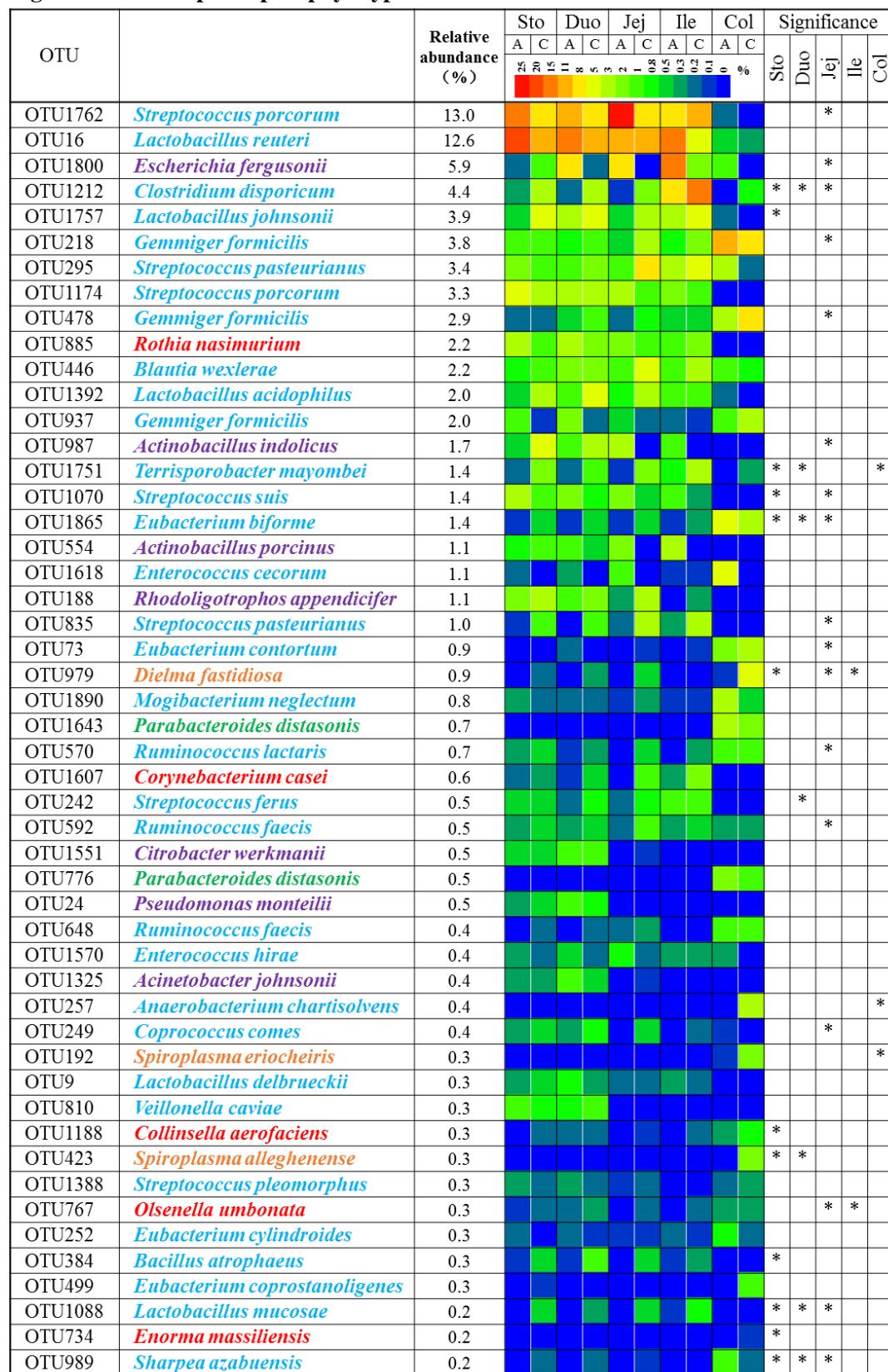
Shannon (A) and Shannoneven (B) index of gut microbiota across stomach and colon. Data were expressed as mean \pm SEM, $n = 8$ for each group. Asterisks indicate different from between groups (Student's *t*-test, $*P < 0.05$).

Figure S3. Relative abundance of dominant phylum



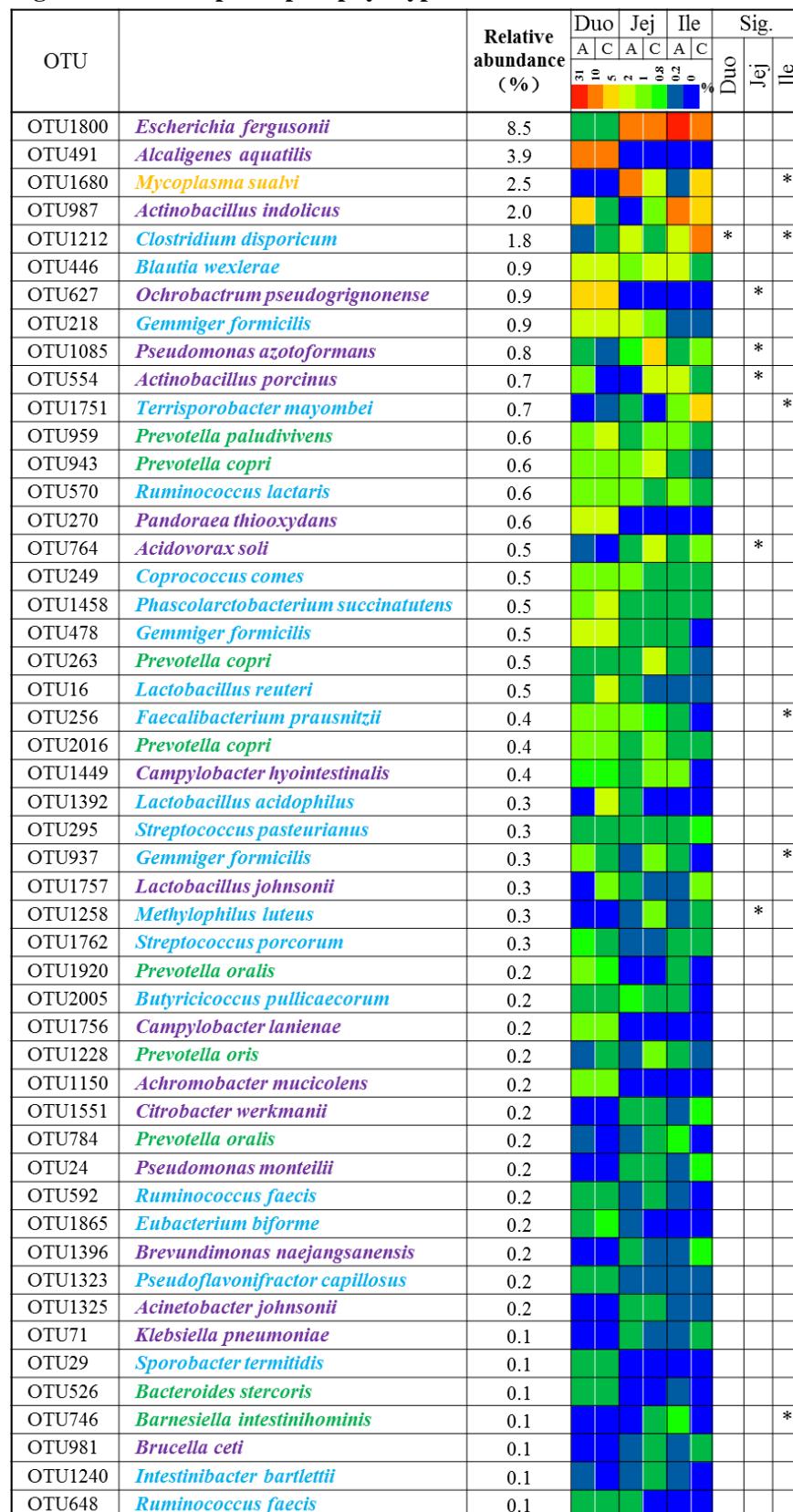
Relative abundance of dominant phylum in lumen (**A**), mucosa (**B**), and difference between lumen and mucosa (**C**). For (**A**) and (**B**), values are medians ($n = 8$). Different letters indicate significant difference based on non-parametric Kruskal–Wallis ANOVA. For (**C**), values are medians ($n = 8$). Asterisks indicate different in lumen compared with mucosa (Mann-Whitney U -test): ** $P < 0.01$; *** $P < 0.001$.

Figure S4. Heatmap of top 50 phylotypes in the lumen



Asterisks indicate significant difference between control and antibiotic treatment (Mann-Whitney U-test $P < 0.05$). The mean relative abundance was calculated using all the samples in control and antibiotic group. A, antibiotics; C, control; Col, colon; Duo, duodenum; Ile, ileum; Jej, jejunum; Sto, stomach.

Figure S5. Heatmap of top 50 phylotypes in the mucosa



Asterisks indicate significant difference between control and antibiotic treatment (Mann-Whitney *U*-test $P < 0.05$). The mean relative abundance was calculated using all the samples in control and antibiotic group. A, antibiotics; C, control; Col, colon; Duo, duodenum; Ile, ileum; Jej, jejunum; Sto, stomach.

Table S1. The composition and ingredient of experimental diets

Items	Content
Ingredients (%)	
corn	40
rice, broken	15
soybean meal, fermented	10
soybean meal, de-hulled	6
spray dried animal plasma	5
whey powder	7
fish meal	4
sugar	4.5
glucose	3
soybean oil	1.5
L-Lysine-HCl (98%)	0.3
L-Methionine	0.15
L-Threonine	0.2
L-Tryptophan	0.05
L-Isoleucine	0.05
L-Valine	0.05
Salt	0.3
Limestone	1.1
CaHPO ₄	0.8
Vitamin mixture ¹	0.2
Mineral mixture ²	0.8
Total	100
Nutrient composition, %	
Crude protein	20.2
Digestible energy (Mcal/kg)	3.4
Total calcium	0.85
Total phosphorus	0.7
Digestible Lys	1.45
Digestible Met+Cys	0.79
Digestible Thr	0.81
Digestible Trp	0.23
Digestible Ile	0.74
Digestible Leu	1.45
Digestible Val	0.89

¹ Vitamin mixture supplied the following per kg complete diet: vitamin A, 15,000 IU; vitamin D3, 3,000 IU; vitamin E, 150 mg; vitamin K3, 3 mg; vitamin B1, 3 mg; vitamin B2, 6 mg; vitamin B6, 5 mg; vitamin B12, 0.03 mg; niacin, 45 mg; vitamin C, 250 mg; calcium pantothenate, 9 mg; folic acid, 1 mg; biotin, 0.3 mg; choline chloride, 500 mg.

² Mineral mixture supplied the following per kg complete diet: Fe, 170 mg; Cu, 150 mg; I, 0.90 mg; Se, 0.2 mg; Zn, 150 mg; Mg, 68 mg; Mn, 80 mg; Co, 0.3 mg.

Table S2. Abundance of the dominant genus in the lumen of stomach, duodenum, jejunum, ileum and colon. The data of relative abundance (%) are expressed as median. Different letters indicate significant difference based on non-parametric Kruskal–Wallis ANOVA ($P < 0.05$), followed by multiple comparisons of mean ranks.

Items	Stomach	Duodenum	Jejunum	Ileum	Colon
<i>Streptococcus</i>	9.50a	10.64a	15.23a	17.56a	0.17b
<i>Lactobacillus</i>	12.88a	10.27a	11.90a	10.42ab	0.14b
<i>Subdoligranulum</i>	0.85a	1.57ab	2.72ab	0.51a	11.84b
<i>Escherichia-Shigella</i>	0.22a	0.16ab	0.01bc	0.03abc	0.01c
<i>Clostridium</i>	1.02a	0.88a	0.18a	0.39a	0.27a
<i>Actinobacillus</i>	1.01a	0.26ab	0.00b	0.00b	0.00b
<i>Blautia</i>	0.64ab	0.95ab	2.82a	0.90ab	0.11b
<i>Rothia</i>	0.91a	0.96a	0.53a	0.10ab	0.00b
<i>Enterococcus</i>	0.10a	0.17a	0.05ab	0.18a	0.00b
<i>Parabacteroides</i>	0.01ab	0.01ab	0.00a	0.00a	0.33b
<i>Prevotella</i>	0.36ac	0.18abc	0.00ab	0.00b	1.87c
<i>Mogibacterium</i>	0.08a	0.05a	0.19a	0.01a	0.11a
<i>Pseudomonas</i>	0.61a	0.45a	0.01ab	0.00b	0.00b
<i>Corynebacterium</i>	0.20a	0.20ab	0.15ab	0.27ab	0.02b
<i>Acinetobacter</i>	0.30a	0.37a	0.01ab	0.00b	0.00b
<i>Bacillus</i>	0.37a	0.76a	0.14ab	0.04ab	0.01b
<i>Ruminococcus</i>	0.05ab	0.14ab	0.04ab	0.03a	0.22b
<i>Sharpea</i>	0.05a	0.05a	0.04a	0.02a	0.00a
<i>Haemophilus</i>	0.21a	0.12a	0.00b	0.00b	0.00b
<i>Paracoccus</i>	0.02ac	0.07a	0.00bc	0.00bc	0.00b
<i>Stenotrophomonas</i>	0.14a	0.17a	0.01ab	0.00b	0.00b
<i>Rhizobium</i>	0.12a	0.10a	0.01ab	0.00ab	0.00b
<i>Faecalibacterium</i>	0.10a	0.07a	0.01ab	0.00b	0.02ab
<i>Solobacterium</i>	0.05a	0.03a	0.03a	0.02a	0.11a
<i>Sphingomonas</i>	0.03a	0.05a	0.07a	0.00a	0.00a
<i>Neisseria</i>	0.04a	0.02ab	0.00ab	0.00ab	0.00b
<i>RC10_gut_group</i>	0.00ab	0.01ab	0.00a	0.00a	0.09b
<i>Staphylococcus</i>	0.05a	0.07a	0.06a	0.00ab	0.00b
<i>Propionibacterium</i>	0.04a	0.03a	0.00b	0.00b	0.00b
<i>Treponema</i>	0.00a	0.00a	0.00a	0.00a	0.00a

Table S3. Abundance of the dominant genus in the mucosa of duodenum, jejunum, and ileum. The data of relative abundance (%) are expressed as median. Different letters indicate significant difference based on non-parametric Kruskal–Wallis ANOVA ($P < 0.05$), followed by multiple comparisons of mean ranks.

Items	Duodenum	Jejunum	Ileum
<i>Escherichia-Shigella</i>	0.24a	11.65b	16.12b
<i>Alcaligenes</i>	27.23a	0.01b	0.00b
<i>Prevotella</i>	8.03ab	9.58a	0.85b
<i>Clostridium</i>	0.56a	0.50a	9.41b
<i>Lactobacillus</i>	1.32a	0.30a	1.32a
<i>Actinobacillus</i>	0.31a	0.78a	0.44a
<i>Mycoplasma</i>	0.01a	0.63a	0.78a
<i>Pseudomonas</i>	0.29a	5.40b	1.45ab
<i>Subdoligranulum</i>	7.49a	2.43ab	0.38b
<i>Terrisporobacter</i>	0.21ab	0.06a	2.55b
<i>Blautia</i>	2.58a	1.13ab	0.30b
<i>Ruminococcus</i>	2.71a	1.56ab	0.31b
<i>Ochrobactrum</i>	6.84a	0.01b	0.00b
<i>Acidovorax</i>	0.04a	3.29b	0.52ab
<i>Streptococcus</i>	1.14a	1.11a	1.40a
<i>Leeia</i>	2.32a	0.02ab	0.00b
<i>Campylobacter</i>	0.18a	0.18a	0.02a
<i>Coprococcus</i>	1.66a	0.76ab	0.32b
<i>Phascolarctobacterium</i>	1.39a	0.31ab	0.03b
<i>Faecalibacterium</i>	1.52a	0.82ab	0.11b
<i>Methylophilus</i>	0.02a	1.31b	0.35ab
<i>Morganella</i>	0.49a	0.48a	0.01a
<i>Acinetobacter</i>	0.03a	1.08b	0.26ab
<i>Citrobacter</i>	0.03a	0.77b	0.23ab
<i>Anaerovibrio</i>	0.29a	0.49a	0.05a
<i>Achromobacter</i>	1.62a	0.02b	0.02b
<i>Butyricicoccus</i>	0.26a	0.49a	0.09a
<i>Parabacteroides</i>	0.65a	0.37ab	0.01b
<i>Bacteroides</i>	0.49a	0.20ab	0.05b
<i>Eubacterium</i>	0.69a	0.10b	0.08b

Table S4. Genus-level effects of antibiotic treatment (Ant) on the dominant bacteria in the lumen compared with the control group (Con). The data of relative abundance (%) are expressed as median. Asterisks indicate different from control group (Mann-Whitney U -test and a false discovery rate < 5%).

Table S5. Genus-level effects of antibiotic treatment (Ant) on the dominant bacteria in the mucosa compared with the control group (Con). The data of relative abundance (%) are expressed as median. Asterisks indicate different from control group (Mann-Whitney *U*-test and a false discovery rate < 5%).

Items	Duodenum		Jejunum		Ileum	
	Con	Ant	Con	Ant	Con	Ant
<i>Escherichia-Shigella</i>	0.499	0.708	16.998	29.238	29.506	36.973
<i>Alcaligenes</i>	24.841	26.519	0.013	0.020	0.039	0.030
<i>Prevotella</i>	8.846	8.551	12.525	5.243	2.542	6.691
<i>Actinobacillus</i>	0.693	9.392	4.026	0.135*	6.849	15.622
<i>Mycoplasma</i>	0.041	0.028	4.082	21.619	7.019	0.239*
<i>Phascolarctobacterium</i>	2.432	1.860	0.502	0.565	0.429	0.741
<i>Pseudomonas</i>	0.307	0.622	7.454	2.144	3.136	1.010
<i>Ruminococcus</i>	3.846	3.007	1.642	2.217	0.905	2.224
<i>Streptococcus</i>	1.504	1.978	1.041	1.429	2.175	1.536
<i>Blautia</i>	2.807	3.447	3.500	1.498	0.732	2.177
<i>Eubacterium</i>	0.877	0.846	0.134	0.389	0.167	0.130
<i>Faecalibacterium</i>	1.628	1.879	1.195	1.410	0.192	0.724*
<i>Lactobacillus</i>	9.245	0.998	0.804	2.024	1.651	0.713
<i>Clostridium</i>	0.803	0.374*	0.886	2.525	19.158	2.894*
<i>Coprococcus</i>	2.008	1.796	0.892	1.274	0.535	0.966
<i>Parabacteroides</i>	0.761	0.719	0.413	0.123	0.085	0.414
<i>Subdoligranulum</i>	6.936	6.940	2.659	3.278	0.467	1.622
<i>Achromobacter</i>	1.615	1.099	0.023	0.002	0.018	0.002
<i>Acidovorax</i>	0.048	0.201	4.569	0.704*	1.326	0.411
<i>Acinetobacter</i>	0.056	0.153	1.371	0.912	0.469	0.357
<i>Anaerovibrio</i>	0.428	0.279	0.499	0.189	0.648	0.619
<i>Bacteroides</i>	0.782	0.624	0.314	0.185	0.152	0.474
<i>Butyricicoccus</i>	0.600	0.637	0.644	1.016	0.153	0.824
<i>Campylobacter</i>	2.232	2.370	1.386	0.575	0.187	1.194
<i>Citrobacter</i>	0.039	0.039	0.713	0.649	1.005	0.339
<i>Leeia</i>	4.335	2.935	0.069	0.078	0.109	0.127
<i>Methylophilus</i>	0.029	0.087	1.994	0.411*	0.673	0.287
<i>Morganella</i>	0.489	0.365	1.418	0.219	0.225	0.442
<i>Ochrobactrum</i>	6.247	5.883	0.009	0.000*	0.009	0.001
<i>Terrisporobacter</i>	0.310	0.149	0.107	0.441	7.066	1.036*

Table S6. Phylotype-level effects of antibiotic treatment (Ant) on the dominant bacteria in the lumen compared with the control group (Con). The data of relative abundance (%) are expressed as median. Asterisks indicate different from control group (Mann-Whitney *U*-test and a false discovery rate < 5%).

Items	Taxonomy	Stomach		Duodenum		Jejunum		Ileum		Colon	
		Con	Ant	Con	Ant	Con	Ant	Con	Ant	Con	Ant
OTU1762	<i>g_Streptococcus</i>	5.704	16.246	3.180	6.534	4.315	22.425*	6.574	6.569	0.002	0.011
OTU16	<i>g_Lactobacillus</i>	5.114	15.276	4.513	7.166	7.480	1.980	4.281	10.690	0.198	0.037
OTU1800	<i>g_Escherichia-Shigella</i>	0.396	0.119	0.293	0.417	0.015	4.008*	0.057	0.718	0.012	0.038
OTU1212	<i>g_Clostridium</i>	1.713	0.127*	1.593	0.096*	0.246	0.042*	0.682	0.303	0.190	0.011
OTU1757	<i>g_Lactobacillus</i>	3.401	0.260*	3.564	1.468	1.854	0.278	6.198	0.597	0.005	0.004
OTU218	<i>g_Subdoligranulum</i>	1.164	0.464	1.899	0.900	2.924	0.379*	0.575	0.184	11.479	2.163
OTU295	<i>g_Streptococcus</i>	0.545	0.659	0.631	0.795	2.340	0.893	3.671	0.853	0.051	0.138
OTU1174	<i>g_Streptococcus</i>	2.555	5.422	3.063	3.605	1.246	4.318	0.500	0.882	0.002	0.003
OTU478	<i>g_Subdoligranulum</i>	0.270	0.153	0.655	0.361	0.858	0.228*	0.319	0.046	3.083	0.809
OTU885	<i>g_Rothia</i>	1.628	2.683	1.704	1.608	0.893	2.298	0.187	0.790	0.003	0.003
OTU446	<i>g_Blautia</i>	1.029	0.585	1.445	1.139	4.336	0.428	1.219	0.236	0.140	0.656
OTU1392	<i>g_Lactobacillus</i>	0.884	0.409	1.066	0.781	1.354	0.085	0.480	0.584	0.022	0.009
OTU937	<i>g_Subdoligranulum</i>	0.079	0.314	0.135	0.665	0.152	0.179	0.040	0.094	0.080	0.658
OTU987	<i>g_Actinobacillus</i>	1.176	0.716	0.275	0.649	0.000	0.308*	0.000	0.000	0.000	0.000
OTU1751	<i>f_Peptostreptococcaceae</i>	0.997	0.223*	0.655	0.114*	0.344	0.048	0.218	0.065	0.477	0.007*
OTU1070	<i>g_Streptococcus</i>	0.470	2.669*	0.854	1.265	0.861	1.551*	0.211	1.179	0.000	0.000
OTU1865	<i>f_Erysipelotrichaceae</i>	0.582	0.054*	0.586	0.110*	0.614	0.025*	0.274	0.016	1.380	0.839
OTU554	<i>g_Actinobacillus</i>	0.591	0.610	0.122	1.238	0.000	0.247	0.003	0.009	0.000	0.000
OTU1618	<i>g_Enterococcus</i>	0.016	0.029	0.053	0.033	0.014	0.026	0.040	0.029	0.003	0.010
OTU188	<i>p_Proteobacteria</i>	2.546	1.174	1.679	0.207	0.346	0.076	0.015	0.010	0.000	0.000
OTU835	<i>g_Streptococcus</i>	0.436	0.062	0.834	0.033	3.968	0.117*	0.458	0.156	0.000	0.001
OTU73	<i>f_Lachnospiraceae</i>	0.027	0.019	0.051	0.078	0.134	0.018*	0.058	0.010	1.216	0.816
OTU979	<i>p_Tenericutes</i>	0.143	0.001*	0.073	0.013	0.043	0.000*	0.037	0.000*	0.067	0.009
OTU1890	<i>g_Mogibacterium</i>	0.115	0.185	0.077	0.147	0.199	0.052	0.021	0.012	0.197	1.069
OTU1643	<i>g_Parabacteroides</i>	0.001	0.000	0.008	0.005	0.000	0.002	0.000	0.001	0.139	1.713
OTU570	<i>f_Lachnospiraceae</i>	0.144	0.168	0.152	0.052	0.049	0.012*	0.186	0.000	0.033	0.180
OTU1607	<i>g_Corynebacterium</i>	0.208	0.146	0.105	0.080	0.217	0.084	0.398	0.191	0.014	0.000
OTU242	<i>g_Streptococcus</i>	0.515	0.214	0.609	0.197*	0.915	0.180	0.437	0.057	0.004	0.010
OTU592	<i>f_Lachnospiraceae</i>	0.533	0.283	0.458	0.284	0.662	0.072*	0.216	0.062	0.068	0.291
OTU1551	<i>f_Enterobacteriaceae</i>	0.744	0.288	0.595	0.629	0.015	0.011	0.003	0.010	0.003	0.004
OTU776	<i>g_Parabacteroides</i>	0.000	0.000	0.002	0.004	0.000	0.000	0.000	0.000	0.163	0.461
OTU24	<i>g_Pseudomonas</i>	0.574	0.249	0.409	0.456	0.018	0.003	0.000	0.003	0.003	0.004
OTU648	<i>f_Lachnospiraceae</i>	0.078	0.033	0.030	0.013	0.029	0.009	0.039	0.016	0.009	0.033
OTU1570	<i>g_Enterococcus</i>	0.147	0.232	0.242	0.104	0.077	0.025	0.264	0.032	0.003	0.010
OTU1325	<i>g_Acinetobacter</i>	0.423	0.224	0.464	0.335	0.015	0.007	0.000	0.004	0.001	0.006
OTU257	<i>f_Ruminococcaceae</i>	0.003	0.004	0.011	0.018	0.021	0.002	0.003	0.005	0.603	0.014*
OTU249	<i>g_Coprococcus</i>	0.532	0.400	0.699	0.103	0.522	0.036*	0.252	0.031	0.043	0.031
OTU192	<i>p_Tenericutes</i>	0.001	0.001	0.014	0.000	0.000	0.000	0.000	0.000	0.695	0.053*
OTU9	<i>g_Lactobacillus</i>	0.213	0.177	0.189	0.155	0.171	0.083	0.094	0.027	0.001	0.004
OTU810	<i>g_Veillonella</i>	0.114	0.039	0.074	0.059	0.006	0.008	0.003	0.003	0.000	0.000
OTU1188	<i>g_Collinsella</i>	0.095	0.033*	0.237	0.110	0.046	0.017	0.045	0.005	0.339	0.278
OTU423	<i>p_Tenericutes</i>	0.030	0.000*	0.012	0.000*	0.002	0.000	0.000	0.000	0.010	0.014
OTU1388	<i>f_Erysipelotrichaceae</i>	0.090	0.176	0.106	0.155	0.104	0.058	0.024	0.014	0.062	0.138
OTU767	<i>g_Atopobium</i>	0.153	0.069	0.242	0.079	0.125	0.018*	0.125	0.005*	0.316	0.135
OTU252	<i>f_Erysipelotrichaceae</i>	0.046	0.178	0.062	0.188	0.027	0.084	0.055	0.009	0.114	0.119
OTU384	<i>g_Bacillus</i>	0.584	0.069*	1.180	0.035	0.231	0.017	0.064	0.010	0.006	0.008

OTU499	f_	Ruminococcaceae	0.043	0.003	0.024	0.007	0.017	0.004	0.006	0.000	0.005	0.014
OTU1088	g_	Lactobacillus	0.170	0.021*	0.073	0.011*	0.225	0.024*	0.489	0.027	0.000	0.004
OTU734	g_	Collinsella	0.005	0.000*	0.003	0.000	0.000	0.000	0.006	0.000	0.005	0.001
OTU989	g_	Sharpea	0.100	0.009*	0.094	0.011*	0.068	0.000*	0.025	0.001	0.005	0.034

Table S7. Phylotype-level effects of antibiotic treatment (Ant) on the dominant bacteria in the mucosa compared with the control group (Con). The data of relative abundance (%) are expressed as median. Asterisks indicate different from control group (Mann-Whitney *U*-test and a false discovery rate < 5%).

Items	Taxonomy	Duodenum		Jejunum		Ileum	
		Con	Ant	Con	Ant	Con	Ant
OTU1800	<i>g_Escherichia-Shigella</i>	0.229	0.165	8.985	9.109	15.242	25.732
OTU491	<i>g_Alcaligenes</i>	25.602	23.065	0.010	0.008	0.000	0.023
OTU1680	<i>g_Mycoplasma</i>	0.005	0.010	0.502	2.144	0.735	0.006*
OTU987	<i>g_Actinobacillus</i>	0.202	0.392	0.349	0.035	0.368	0.879
OTU1212	<i>g_Clostridium</i>	0.490	0.271*	0.458	1.371	8.079	0.867*
OTU446	<i>g_Blautia</i>	2.106	2.834	0.996	0.665	0.219	0.749
OTU627	<i>f_Brucellaceae</i>	6.439	4.766	0.006	0.000*	0.004	0.000
OTU218	<i>g_Subdoligranulu</i>	4.510	1.283	0.720	0.877	0.197	0.182
OTU1085	<i>g_Pseudomonas</i>	0.179	0.272	4.027	0.431*	0.874	0.416
OTU554	<i>g_Actinobacillus</i>	0.132	0.188	0.388	0.003*	0.053	0.415
OTU1751	<i>g_Terrisporobacter</i>	0.203	0.096	0.059	0.295	2.409	0.304*
OTU959	<i>g_Prevotella</i>	1.266	1.105	1.048	0.366	0.092	0.300
OTU943	<i>g_Prevotella</i>	1.216	0.928	1.466	0.267	0.078	0.318
OTU570	<i>g_Ruminococcus</i>	1.267	0.351	0.327	0.508	0.180	0.108
OTU270	<i>g_Leeia</i>	2.188	1.764	0.017	0.069	0.004	0.049
OTU764	<i>g_Acidovorax</i>	0.039	0.076	3.050	0.214*	0.498	0.266
OTU249	<i>g_Coprococcus</i>	1.379	1.128	0.356	0.314	0.256	0.195
OTU1458	<i>g_Phascolarctobacterium</i>	1.292	1.298	0.290	0.442	0.025	0.251
OTU478	<i>g_Subdoligranulum</i>	1.848	1.612	0.257	0.282	0.019	0.117
OTU263	<i>g_Prevotella</i>	0.630	0.316	1.145	0.200	0.108	0.420
OTU16	<i>g_Lactobacillus</i>	0.514	0.468	0.083	0.307	0.268	0.243
OTU256	<i>g_Faecalibacterium</i>	1.012	1.663	0.625	0.721	0.087	0.537*
OTU2016	<i>g_Prevotella</i>	1.355	0.795	0.429	0.236	0.197	0.405
OTU1449	<i>g_Campylobacter</i>	0.068	0.118	0.081	0.113	0.015	0.144
OTU1392	<i>g_Lactobacillus</i>	0.126	0.059	0.110	0.182	0.149	0.093
OTU295	<i>g_Streptococcus</i>	0.396	0.605	0.484	0.415	0.623	0.264
OTU937	<i>g_Subdoligranulum</i>	0.499	0.484	0.312	0.037	0.050	0.211*
OTU1757	<i>g_Lactobacillus</i>	0.589	0.082	0.069	0.576	0.445	0.151
OTU1258	<i>g_Methylophilus</i>	0.021	0.044	1.223	0.219	0.336	0.172*
OTU1762	<i>g_Streptococcus</i>	0.405	0.741	0.166	0.014	0.146	0.411
OTU1920	<i>g_Prevotella</i>	0.057	0.375	0.000	0.020	0.000	0.000
OTU2005	<i>g_Butyricicoccus</i>	0.172	0.162	0.413	0.214	0.076	0.351
OTU1756	<i>g_Campylobacter</i>	0.048	0.030	0.025	0.000	0.001	0.038
OTU1228	<i>g_Prevotella</i>	0.458	0.221	0.446	0.048	0.013	0.150
OTU1150	<i>g_Achromobacter</i>	1.536	1.035	0.021	0.002	0.017	0.002
OTU1551	<i>g_Citrobacter</i>	0.030	0.020	0.711	0.228	0.220	0.169
OTU784	<i>g_Prevotella</i>	0.115	0.113	0.204	0.059	0.005	0.019
OTU24	<i>g_Pseudomonas</i>	0.028	0.028	0.388	0.162	0.427	0.200
OTU592	<i>g_Ruminococcus</i>	0.447	0.412	0.193	0.047	0.091	0.076
OTU1865	<i>g_Eubacterium</i>	0.634	0.234	0.078	0.087	0.073	0.052
OTU1396	<i>g_Brevundimonas</i>	0.051	0.040	0.162	0.211	0.128	0.145
OTU1323	<i>f_Ruminococcaceae</i>	0.461	0.159	0.149	0.155	0.109	0.147
OTU1325	<i>g_Acinetobacter</i>	0.023	0.028	0.631	0.210	0.209	0.169
OTU71	<i>g_Klebsiella</i>	0.019	0.010	0.196	0.191	0.161	0.126
OTU29	<i>f_Ruminococcaceae</i>	0.321	0.284	0.065	0.058	0.035	0.084
OTU526	<i>g_Bacteroides</i>	0.313	0.235	0.091	0.091	0.021	0.078

OTU746	<i>g_Barnesiella</i>	0.044	0.054	0.404	0.019	0.000	0.282*
OTU981	<i>g_Brucella</i>	0.012	0.018	0.298	0.168	0.108	0.164
OTU1240	<i>f_Ruminococcaceae</i>	0.070	0.197	0.311	0.064	0.062	0.206
OTU648	<i>g_Ruminococcus</i>	0.289	0.064	0.015	0.104	0.037	0.007

Table S8. Distribution of the KEGG categories predicted to be represented in bacterial communities of lumen of duodenum, jejunum and ileum compared with the mucosa. The data from control group were used for analysis here. Data are presented as mean relative abundances. Asterisks indicate different in lumen compared with the mucosa in the corresponding location (false discovery rate < 5%).

Items	KEGG Pathways	Duodenum		Jejunum		Ileum	
		Mucosa	Lumen	Mucosa	Lumen	Mucosa	Lumen
Cell Motility	Bacterial motility proteins	1.292	0.895*	1.240	0.885	1.325	0.833
Cell Motility	Flagellar assembly	0.630	0.171*	0.542	0.068*	0.612	0.289
Membrane Transport	Bacterial secretion system	0.822	0.562*	0.738	0.477*	0.702	0.490*
Folding, Sorting and Degradation	Ubiquitin system	0.003	0.021*	0.011	0.022	0.009	0.011
Amino Acid Metabolism	Alanine, aspartate and glutamate metabolism	1.000	0.893*	0.885	0.923	0.768	0.947*
Amino Acid Metabolism	Arginine and proline metabolism	1.253	1.155*	1.117	1.280*	1.002	1.047
Amino Acid Metabolism	Glycine, serine and threonine metabolism	0.822	0.708*	0.810	0.688*	0.729	0.715
Amino Acid Metabolism	Lysine biosynthesis	0.784	0.665*	0.682	0.679	0.563	0.732*
Amino Acid Metabolism	Lysine degradation	0.248	0.183	0.301	0.123*	0.245	0.116*
Amino Acid Metabolism	Phenylalanine metabolism	0.218	0.186	0.267	0.173*	0.214	0.135
Amino Acid Metabolism	Phenylalanine, tyrosine and tryptophan biosynthesis	0.793	0.665*	0.718	0.767	0.567	0.642
Amino Acid Metabolism	Tryptophan metabolism	0.286	0.231	0.341	0.170	0.279	0.151*
Amino Acid Metabolism	Valine, leucine and isoleucine biosynthesis	0.791	0.698*	0.701	0.777	0.686	0.708
Amino Acid Metabolism	Valine, leucine and isoleucine degradation	0.420	0.325	0.477	0.220*	0.382	0.220*
Carbohydrate Metabolism	Butyrate metabolism	0.681	0.599	0.783	0.468*	0.713	0.594
Carbohydrate Metabolism	Citrate cycle	0.605	0.466*	0.617	0.379*	0.523	0.372*
Carbohydrate Metabolism	Propionate metabolism	0.665	0.543*	0.700	0.425*	0.615	0.521
Carbohydrate Metabolism	Starch and sucrose metabolism	0.742	0.898*	0.785	0.988*	0.842	1.061*
Enzyme Families	Peptidases	1.675	1.925*	1.668	2.042*	1.641	1.842*
Glycan Biosynthesis and Metabolism	Glycosyltransferases	0.322	0.487*	0.346	0.513*	0.287	0.392
Glycan Biosynthesis and Metabolism	Lipopolysaccharide biosynthesis	0.253	0.174	0.298	0.152*	0.208	0.075

Glycan Biosynthesis and Metabolism	N-Glycan biosynthesis	0.020	0.050*	0.016	0.069*	0.008	0.024
Metabolism of Other Amino Acids	beta-Alanine metabolism	0.298	0.233	0.345	0.212*	0.248	0.163
Metabolism of Other Amino Acids	Taurine and hypotaurine metabolism	0.095	0.115*	0.114	0.098*	0.128	0.129
Metabolism	Carbohydrate metabolism	0.146	0.214*	0.169	0.236*	0.167	0.185
Metabolism	Energy metabolism	0.732	0.603*	0.857	0.598*	0.752	0.614

Table S9. Effect of antibiotic treatment (Ant) on the KEGG orthology involved in carbohydrate-active enzyme predicted to be differentially represented (false discovery rate < 5%) in bacterial communities compared with control group (Con). Data are presented as mean relative abundances. L represents for Lumen and M for mucosa.

Gut location	KEGG_Description	KEGG orthology	Cazy family	Con	Ant
Stomach-L	sialidase-1 [EC:3.2.1.18]	K01186	GH1	0.012	0.041
	beta-glucosidase [EC:3.2.1.21]	K05350	GH1	0.044	0.067
	hyaluronate lyase [EC:4.2.2.1]	K01727	PL8	0.005	0.021
	sucrose phosphorylase [EC:2.4.1.7]	K00690	GH13	0.016	0.033
	endo-1,3(4)-beta-glucanase [EC:3.2.1.6]	K01180	GH9	6.45E-06	0
Duodenum-L	pectate disaccharide-lyase [EC:4.2.2.9]	K01731	PL1	4.61E-06	0
Jejunum-L	xylan 1,4-beta-xylosidase [EC:3.2.1.37]	K01198	GH1	0.000	0.002
	(1->4)-alpha-D-glucan	1-alpha-D-glucosylmutase	K06044	GH13	0.000
	[EC:5.4.99.15]				
	maltooligosyltrehalose trehalohydrolase [EC:3.2.1.141]	K01236	GH13	0.000	0.002
	alpha-N-acetylglucosaminidase [EC:3.2.1.50]	K01205	GH89	0.000	0.001
Ileum-M	trehalose-phosphatase [EC:3.1.3.12]	K01087	GT20	0.000	0.002
	alpha,alpha-trehalose-phosphate synthase (UDP-forming)	K00697	GT20	0.000	0.003
	[EC:2.4.1.15]				
	4-alpha-glucanotransferase [EC:2.4.1.25]	K00705	GH13	0.048	0.065
	levanase [EC:3.2.1.65]	K01212	GH32	0.003	0.011
Colon-L	alpha-L-fucosidase [EC:3.2.1.51]	K01206	GH29	0.014	0.034
	starch synthase [EC:2.4.1.21]	K00703	GT5	0.048	0.065
	levansucrase [EC:2.4.1.10]	K00692	GH32	4.73E-05	0.000
	beta-glucosidase [EC:3.2.1.21]	K01188	GH1	0.069	0.045
	endo-1,4-beta-xylanase [EC:3.2.1.8]	K01181	GH3	0.019	0.005
Ileum-L	endoglucanase [EC:3.2.1.4]	K01179	GH5	0.079	0.027
	cellulose 1,4-beta-celllobiosidase [EC:3.2.1.91]	K01225	GH5	0.000379	8.51E-05
	molybdenum transport protein [EC:2.4.2.-]	K03813	GH10	0.006	0.002
	alpha-N-arabinofuranosidase [EC:3.2.1.55]	K01209	GH13	0.049	0.024
	levansucrase [EC:2.4.1.10]	K00692	GH32	0.005	0.000128