

Supplementary Figures

Early immune innate hallmarks and microbiome changes across the gut during *Escherichia coli* O157:H7 infection in cattle

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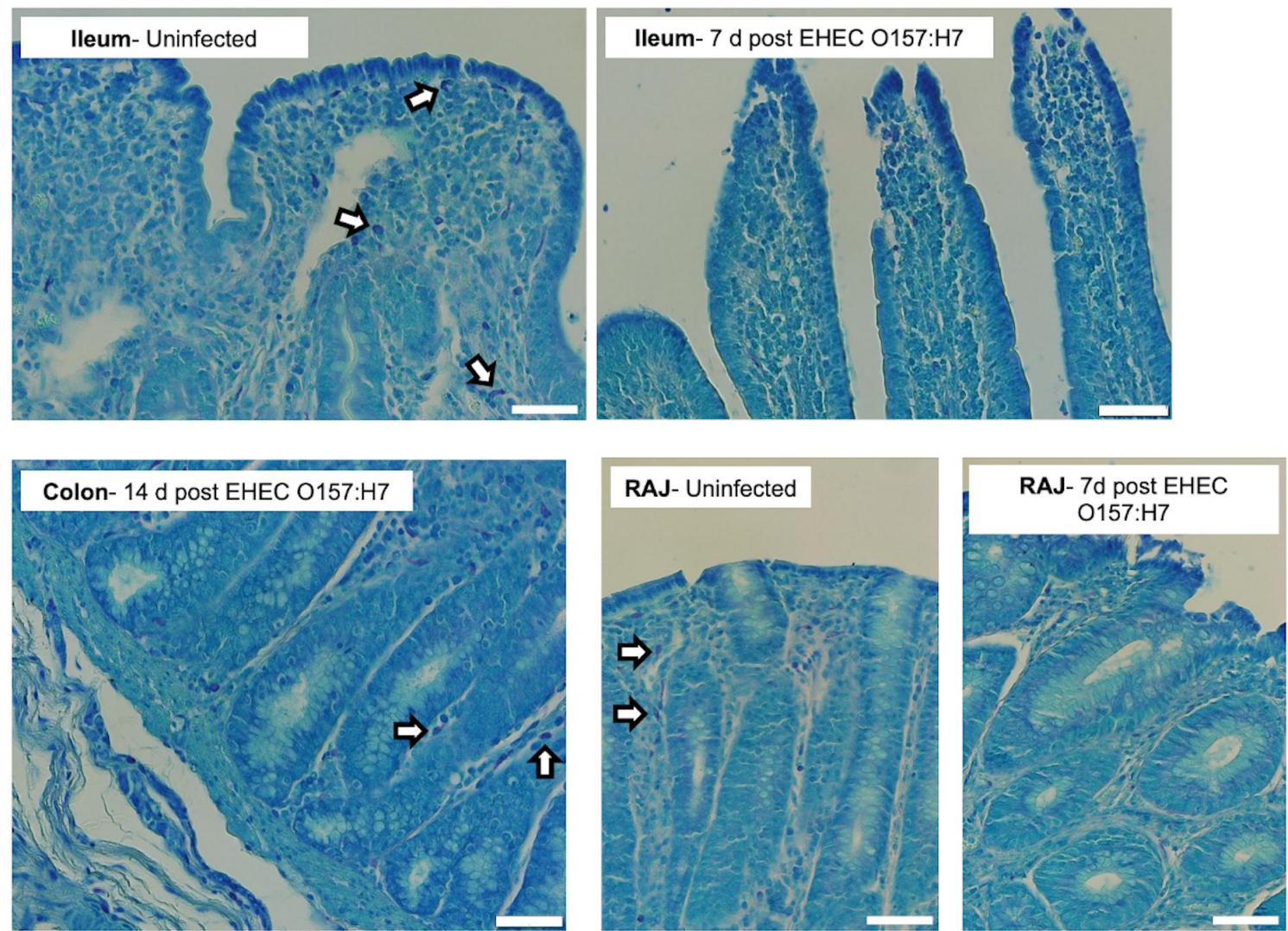
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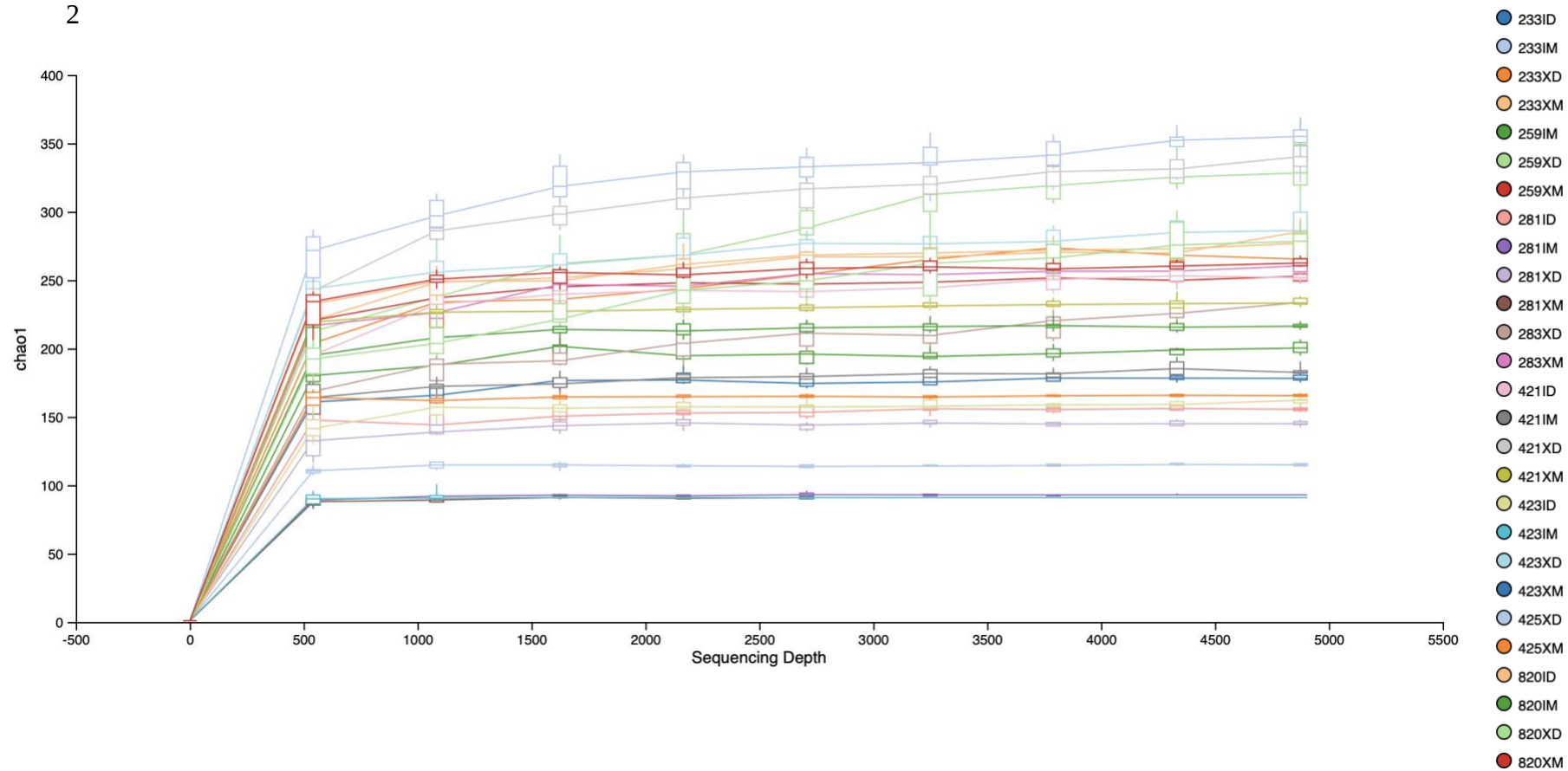
Supplementary Figure 1. Representative histologic images of mast cells producing acidic polysaccharides components (purple) in ileum, colon and rectal anal junction (RAJ) of calves challenged by EHEC O157:H7 (7 d and 14 d post-challenge). Histological detection by toluidin blue staining. Bar = 50 μ m.

Sup Fig 1

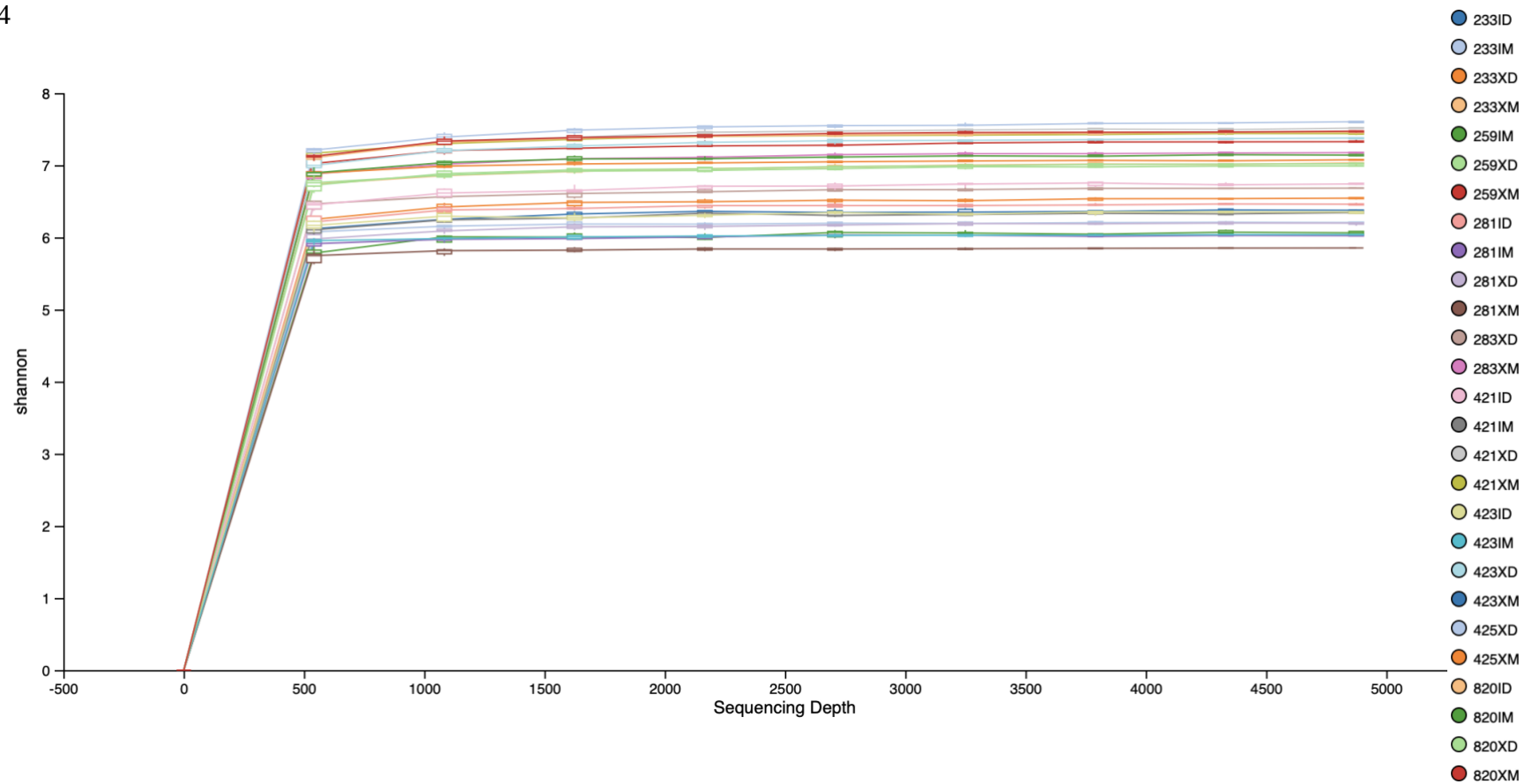


1 **Supplementary Figure 2. Chao and Shannon index rarefaction**

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Supplementary Figure 3

Table Taxonomic Analysis

ileum mucosa						
Phylum	Family	Genus	Relative abundance (%)		SEM	P-value
			Uninfected	Infected		
Actinobacteria			7.55	6.74	2.63	0.857
	Corynebacteriaceae	Corynebacterium 1	0.56	2.62	0.29	0.854
			0.49	2.62	0.94	0.582
	Micrococcaceae		0.54		0.19	0.359
	Propionibacteriaceae	Cutibacterium	1.95	3.50	0.97	0.857
			1.93	3.48	0.96	0.857
	Atopobiaceae	Olsenella	2.94		1.08	0.558
2.48				0.97	0.558	
Bacteroidetes			9.07	11.20	3.49	1
	Bacteroidaceae	Bacteroides	2.13		0.75	0.372
			2.11		0.75	0.372
	Marinifilaceae	Odoribacter	0.35		0.22	1
			0.34		0.16	0.845
	Muribaculaceae	uncultured bacterium	2.12	2.08	0.97	1
			1.67		0.85	0.721
	Prevotellaceae	Prevotella 1	2.65	4.12	1.08	0.475
			0.12	1.32	0.47	0.359
		Prevotella 9	0.30		0.19	1
		Prevotellaceae NK3B31 group	0.60		0.22	0.558
		Prevotellaceae UCG-003	0.38	0.31	0.16	1
		uncultured		0.19	0.06	0.582
		Unclassified		0.13	0.07	0.854
	Rikenellaceae	Rikenellaceae RC9 gut group	0.85	0.89	0.34	1
			0.44	0.88	0.24	0.582
	Tannerellaceae	Parabacteroides	0.66		0.23	0.359
			0.65		0.22	0.359
Cyanobacteria			2.97	9.30	2.60	0.229
	Unclassified	Unclassified	0.42		0.20	0.845
			0.42		0.20	0.845
	Eimeria praecox	Eimeria praecox	2.47	8.16	2.36	0.23
			2.47	8.16	2.36	0.23
Epsilonbacteraeota			0.37	1.13	0.34	0.359
	Campylobacteraceae	Campylobacter	0.37	1.13	0.34	0.36
			0.36	1.13	0.34	0.359
Firmicutes			65.40	62.00	5.82	1
	Christensenellaceae	Christensenellaceae R-7 group	1.04		0.53	0.845
			1.03		0.52	0.845
	Clostridiaceae 1	Candidatus Arthromitus	15.05	31.38	10.19	0.629
			13.74	28.22	10.55	0.857
			1.11	3.02	1.10	0.582
	Clostridiales vadinBB60 group		0.08		0.06	1
	Family XIII	Family XIII AD3011 group	1.87	1.15	0.65	1
				0.55	0.15	0.359
		Mogibacterium	1.00	0.26	0.49	1
		[Eubacterium] brachy group	0.31		0.14	0.558
		[Eubacterium] nodatum group	0.25		0.11	0.845
		Unclassified	0.25		0.08	0.359
	Lachnospiraceae	Butyrivibrio	14.64	8.17	3.28	0.4
			0.55		0.27	0.845
		Lachnospiraceae NK3A20 group		0.59	0.26	0.435
		Lachnospiraceae UCG-002		0.40	0.15	0.123
		Roseburia	0.11		0.10	1
		Syntrophococcus	2.08		0.98	0.582
		Unclassified	8.04	4.25	1.89	0.629
	Peptostreptococcaceae	Romboutsia	9.35	3.91	3.48	0.721
			8.76	3.23	3.23	0.854
			Unclassified	0.53		0.35
	Ruminococcaceae	Candidatus Soleaferrea	17.77	11.29	3.81	0.629
			0.37		0.14	0.27
			0.54		0.16	0.582
			0.24		0.08	0.119

	Ruminococcaceae UCG-002	0.12		0.15	1
	Ruminococcaceae UCG-004		0.45	0.14	0.123
	Ruminococcaceae UCG-005	5.90	2.34	2.10	0.854
	Ruminococcaceae UCG-009	0.14		0.06	0.27
	Ruminococcaceae UCG-010	2.49		0.98	0.845
	Ruminococcaceae UCG-013	0.23	0.35	0.11	0.854
	Ruminococcaceae UCG-014	0.46		0.21	0.845
	Ruminococcus 2	0.12	0.50	0.14	0.359
	[Eubacterium] coprostanoligenes group	5.35	1.92	1.41	0.229
	Unclassified	0.67	2.06	0.54	0.359
Erysipelotrichaceae		2.83	4.92	1.95	0.857
	Erysipelotrichaceae UCG-004	0.20		0.08	0.27
	Erysipelotrichaceae UCG-009	0.29	0.15	0.10	0.854
	Turicibacter	0.96		1.66	0.854
	uncultured	0.17		0.07	0.27
Proteobacteria		13.40	8.71	4.76	0.629
	Beijerinckiaceae	0.72		0.25	0.119
	Rhodobacteraceae	0.29		0.16	0.845
	Succinivibrio		1.09	0.45	0.435
	Succinivibrionaceae	0.13	1.09	0.44	0.854
	Burkholderiaceae	0.80		0.42	0.854
	Sutterella	0.74		0.41	1
	Neisseriaceae	1.14		0.50	0.27
	Enterobacteriaceae	9.91	6.36	4.74	0.857
	Escherichia-Shigella	9.84	6.32	4.70	0.629
	Moraxellaceae	0.23		0.08	0.582
Tenericutes		0.82	0.34	0.33	0.854

ileum digesta						
Phylum	Family	Genus	Relative abundance (%)		SEM	P-value
			Uninfected	Infected		
Actinobacteria			2.47		0.97	1
	Atopobiaceae		2.14		0.84	1
		Olsenella	1.69		0.77	1
Bacteroidetes			4.49		4.73	1
	Bacteroidaceae		1.00		0.68	1
		Bacteroides	1.00		0.68	1
	Muribaculaceae		0.51		1.12	1
	Prevotellaceae		1.91		1.38	1
Cyanobacteria			1.07	0.68	0.33	0.80
Firmicutes			77.29	70.73	7.28	0.37
	Christensenellaceae		0.89		1.06	0.8
	Clostridiaceae 1		10.13	4.50	4.79	1
		Candidatus Arthromitus	0.53	0.80	0.22	0.8
		Clostridium sensu stricto 1	9.60	3.71	4.85	1
	Deffluviitaleaceae		0.17		0.07	0.333
		Deffluviitaleaceae UCG-011	0.17		0.07	0.333
	Family XIII		1.76	2.77	0.73	0.8
		Mogibacterium	1.04		0.67	1
		[Eubacterium] brachy group		0.26	0.05	0.2
		[Eubacterium] nodatum group	0.48	0.42	0.23	1
	Lachnospiraceae		13.86	13.88	2.32	0.8
		Howardella	0.64		0.34	1
		Lachnospiraceae NK3A20 group	0.23		0.41	1
		Syntrophococcus	2.92		1.15	0.4
		[Ruminococcus] gauvreauii group	1.76		0.84	0.4
		Unclassified	5.82	8.29	2.04	0.8
	Peptostreptococcaceae		19.31	7.94	6.22	0.8
		Romboutsia	17.51	6.67	5.67	0.8
		Unclassified	1.40	1.25	0.47	1
	Ruminococcaceae		22.57	26.96	4.62	0.8
		Fournierella	0.15		0.06	0.333
		Ruminococcaceae UCG-005	6.02		3.54	1
		Ruminococcaceae UCG-013	0.27	0.27	0.08	1
		Ruminococcaceae UCG-014	0.67	0.38	0.24	0.8
		Ruminococcus 2		3.61	1.10	0.139
		[Eubacterium] coprostanoligenes group	9.81	7.65	2.67	0.8
		Unclassified	0.51	2.75	0.71	0.2
	Unclassified			0.52	0.17	0.4
		Unclassified		0.52	0.17	0.4
	Erysipelotrichaceae		8.27	11.05	3.84	1
		Erysipelotrichaceae UCG-004	0.13		0.05	0.333
		Solobacterium	0.36		0.20	1
		Turicibacter	6.08	9.99	3.59	0.8
Patescibacteria			0.53		0.19	0.554
	Saccharimonadaceae		0.48		0.19	0.554
Proteobacteria			13.01	13.36	4.87	0.80
	Burkholderiaceae		1.11		1.00	1
	Enterobacteriaceae		11.77	9.22	4.96	0.8
		Escherichia-Shigella	11.74	9.22	4.94	0.8

			Rectum mucosa			
Phylum	Family	Genus	Relative abundance (%)		SEM	P-value
			Uninfected	Infected		
Actinobacteria			7.89		3.34	0.359
Bacteroidetes			17.75	29.59	3.75	0.229
	Bacteroidaceae		4.31	6.90	1.46	0.629
		Bacteroides	4.32	6.90	1.46	0.629
	Marinifilaceae		0.80	0.48	0.23	0.721
		Odoribacter	0.76	0.29	0.20	0.475
	Muribaculaceae		3.00	5.77	1.23	0.229
		uncultured Porphyromonadaceae bacterium		0.56	0.17	0.629
		uncultured bacterium	2.32	5.04	1.13	0.4
	Prevotellaceae		6.16	9.51	1.49	0.4
		Alloprevotella	0.62	2.48	0.73	0.4
		Prevotella 1	0.16	0.56	0.13	0.229
		Prevotella 2	1.47	1.46	0.43	0.629
		Prevotella 9	0.46	1.00	0.21	0.229
		Prevotellaceae NK3B31 group	1.36	1.13	0.38	1
		Prevotellaceae UCG-003	0.79	1.06	0.31	0.4
		uncultured	0.30	0.95	0.30	0.475
		Unclassified		0.78	0.22	0.372
	Rikenellaceae		1.97	2.03	0.48	1
		Alistipes	0.86	0.94	0.20	0.857
		Rikenellaceae RC9 gut group	1.11	1.09	0.30	0.857
	Tannerellaceae		0.92	2.06	0.35	0.229
		Parabacteroides	0.92	2.06	0.35	0.229
	Flavobacteriaceae			0.15	0.06	0.558
Cyanobacteria			1.51	0.71	0.60	0.854
	uncultured bacterium			0.52	0.20	0.558
		uncultured bacterium		0.52	0.20	0.558
Firmicutes			63.63	63.03	3.24	1
	Streptococcaceae		6.98		2.95	0.242
	Christensenellaceae		2.11	3.67	0.75	0.4
		Christensenellaceae R-7 group	2.11	3.67	0.75	0.4
	Clostridiaceae 1		0.20	0.16	0.06	1
		Clostridium sensu stricto 1	0.20	0.16	0.06	1
	Clostridiales vadinBB60 group			0.38	0.12	0.582
		uncultured bacterium		0.38	0.12	0.582
	Defluviitaleaceae		4.38		1.84	0.242
		Defluviitaleaceae UCG-011	4.39		1.84	0.242
	Family XIII		0.34	0.60	0.22	1
		Family XIII AD3011 group	0.29	0.17	0.08	0.475
	Lachnospiraceae		15.20	18.87	3.14	0.857
		Agathobacter		0.48	0.22	0.119
		Blautia	0.17	0.16	0.07	0.854
		Butyrivibrio	0.51	0.54	0.25	1
		Coprococcus 3	0.29		0.10	0.435
		Lachnoclostridium 10		0.45	0.18	0.558
		Lachnospiraceae AC2044 group		0.53	0.18	0.359
		Lachnospiraceae NK4A136 group		0.31	0.11	0.119
		Lachnospiraceae UCG-010		0.81	0.29	0.854
		Roseburia	0.44		0.13	0.123
		Tyzzzeria 4		0.58	0.20	0.359
		Unclassified	12.77	13.80	2.86	0.857
	Peptostreptococcaceae		0.27		0.08	0.123
	Ruminococcaceae		27.79	37.02	6.13	0.629
		Candidatus Soleaferrea		0.41	0.22	0.582
		Fournierella	0.24		0.08	0.696
		Negativibacillus	0.53	1.55	0.30	0.057
		Oscillibacter	0.27	0.48	0.08	0.229
		Ruminiclostridium 6		0.37	0.14	0.559
		Ruminiclostridium 9	0.18	0.32	0.09	0.475
		Ruminococcaceae UCG-002	0.38	0.84	0.29	0.854
		Ruminococcaceae UCG-004		0.10	0.04	0.558
		Ruminococcaceae UCG-005	12.76	19.23	4.44	0.629
		Ruminococcaceae UCG-009	0.41	0.42	0.22	0.854
		Ruminococcaceae UCG-010	5.46	5.00	1.10	0.857
		Ruminococcaceae UCG-013	0.57		0.21	0.721

		Ruminococcaceae UCG-014	1.33	1.02	0.46	0.857
		[Eubacterium] coprostanoligenes group	3.32	4.01	0.88	1
		uncultured		0.43	0.11	0.212
		Unclassified	1.48	2.16	0.39	1
	Unclassified			0.46	0.14	0.629
		Unclassified		0.46	0.14	0.629
	Erysipelotrichaceae		0.59	1.80	0.39	0.114
		Erysipelotrichaceae UCG-004		1.03	0.33	0.057
		Solobacterium	0.10		0.03	0.123
		[Anaerorhabdus] furcosa group		0.24	0.15	1
		uncultured		0.15	0.07	0.845
Proteobacteria			5.61	4.76	1.53	1
	uncultured		0.30	0.41	0.16	0.857
		gut metagenome	0.30	0.25	0.11	0.857
	Succinivibrionaceae		0.20	1.91	0.99	0.721
		Succinivibrio	0.20	1.91	0.99	0.721
	Burkholderiaceae		1.26	2.20	0.45	0.4
		Sutterella	1.26	2.20	0.45	0.4
Spirochaetes			0.20	0.36	0.12	1
	Spirochaetaceae		0.20	0.35	0.12	1
		Treponema 2	0.16	0.35	0.13	0.854
Tenericutes			0.98	1.51	0.29	0.629
	gut metagenome			0.94	0.38	0.854
		gut metagenome		0.94	0.38	0.854

Rectum digesta						
Phylum	Family	Genus	Relative abundance (%)		SEM	P-value
			Uninfected	Infected		
Bacteroidetes			19.3625	24.28	3.450129	0.886
	Bacteroidaceae		3.894159087	4.162689279	1.168233	1
		Bacteroides	3.894159087	4.162689279	1.168233	1
	Marinifilaceae		0.5839289	0.289527819	0.132333	0.245
		Odoribacter	0.506959236	0.175038492	0.129741	0.183
	Muribaculaceae		6.097750964	7.34193672	1.865475	1
		uncultured Porphyromonadaceae bacterium		0.847737035	0.363911	0.869
		uncultured bacterium	5.075845228	6.298822757	1.68914	0.886
		Unclassified	0.39492396		0.14902	1
	Prevotellaceae		4.489654792	8.608008956	1.722329	0.343
		Alloprevotella	0.57574133	2.98765317	1.072122	0.486
		Prevotella 1	0.121013681	0.22594707	0.049194	0.686
		Prevotella 2	0.797714226	1.605788794	0.508177	0.561
		Prevotella 9		0.405067207	0.110095	0.309
		Prevotellaceae NK3B31 group	0.502239996	0.589478549	0.161243	1
		Prevotellaceae UCG-003	1.84704742	1.413356832	0.398998	0.886
		uncultured	0.10854826	0.509077939	0.149316	0.2
		Unclassified	0.29782454	0.871639397	0.210865	0.343
	Rikenellaceae		2.463403822	1.720361427	0.521793	0.686
		Alistipes	1.158601847	0.448787953	0.25538	0.245
		Rikenellaceae RC9 gut group	1.304801976	1.271573474	0.338506	1
	Tannerellaceae		1.426802248	1.163602302	0.33654	1
		Parabacteroides	1.426802248	1.163602302	0.33654	1
	uncultured		0.199577758		0.092372	0.869
		uncultured bacterium	0.199577758		0.092372	0.869
Cyanobacteria			0.26	0.1725	0.040309	0.191
	uncultured bacterium		0.193770917	0.116372695	0.051437	0.46
		uncultured bacterium	0.193770917	0.116372695	0.051437	0.46
Firmicutes			73.185	71.905	2.862713	1
	Christensenellaceae		3.408515617	5.498796682	1.381911	0.686
		Christensenellaceae R-7 group	3.408515617	5.498796682	1.381911	0.686
	Clostridiaceae 1		1.108854581	0.137724677	0.42041	0.3
		Clostridium sensu stricto 1	0.742603158	0.137724677	0.259015	0.3
	Clostridiales vadinBB60 group		0.255282496	0.171686723	0.080597	0.882
		uncultured bacterium		0.171686723	0.059756	0.657
	Family XIII		1.176845262	0.619286081	0.277664	0.343
		Family XIII AD3011 group	0.563702009		0.188198	0.46
		[Eubacterium] brachy group	0.052607606		0.020521	0.869
		[Eubacterium] nodatum group	0.350542769		0.116714	0.309
	Lachnospiraceae		16.71442335	19.49568441	2.148406	0.486
		Agathobacter		0.225429389	0.074943	0.408
		Blautia		0.127756612	0.049983	0.186
		Butyrivibrio	0.220754488	0.479259745	0.162142	0.645
		Coprococcus 3	0.08692902		0.149556	0.869
		Dorea		0.172791913	0.063723	0.356
		Lachnoclostridium 10		0.247648854	0.104289	0.62
		Lachnospiraceae AC2044 group		1.251794331	0.370364	0.124
		Lachnospiraceae NK4A136 group		0.573031143	0.226004	0.62
		Lachnospiraceae UCG-010	0.585205461	1.573944426	0.47894	0.686
		Roseburia	0.510669448		0.14049	0.124
		Tyzzerella 4	0.149274329	1.221485477	0.433991	0.645
		Unclassified	11.66683027	12.9276814	1.488806	0.886
	Ruminococcaceae		38.60713954	44.16735124	5.519006	0.686
		Candidatus Soleaferrea	1.230231649	0.768702996	0.421596	0.561
		Fournierella	0.766322427		0.239188	0.3
		Negativibacillus	1.142086918	2.641065332	0.6959	0.343
		Oscillibacter	0.196799371	0.449403083	0.086096	0.191
		Ruminiclostridium 9	0.121353701		0.031758	0.3
		Ruminococcaceae UCG-002	0.919138776	0.843622108	0.270282	1
		Ruminococcaceae UCG-005	16.96592825	23.58553038	4.74098	0.686
		Ruminococcaceae UCG-008	0.083490732	0.178974148	0.069585	1
		Ruminococcaceae UCG-009	0.472121251	0.425165639	0.176254	1
		Ruminococcaceae UCG-010	4.269824524	3.386350483	0.990932	1
		Ruminococcaceae UCG-013	0.824161586	0.454162068	0.185123	0.486
		Ruminococcaceae UCG-014	1.3364725	0.552344705	0.49924	1

		[Eubacterium] coprostanoligenes group	6.979429084	6.938152552	1.221397	0.886
		uncultured	0.16195072	0.275584322	0.071989	0.657
		Unclassified	2.849390188	2.927313848	0.674835	0.886
	Unclassified		0.481825325	0.523724887	0.142696	0.686
		Unclassified	0.481825325	0.523724887	0.142696	0.686
	Erysipelotrichaceae		1.693215173	0.967331981	0.521912	0.686
		Erysipelotrichaceae UCG-004	0.100692938		0.258805	0.657
		[Anaerorhabdus] furcosa group	0.174207962	0.197024277	0.088066	1
		uncultured	0.181525854		0.041874	0.183
Proteobacteria			4.3925	2.565	0.695656	0.486
	uncultured		1.068360058	0.096739235	0.336474	0.183
		gut metagenome	1.005999691	0.082932801	0.32713	0.183
	Succinivibrionaceae		0.095163342	0.429308257	0.136253	0.645
		Succinivibrio		0.429308257	0.139653	0.408
	Burkholderiaceae		1.765172647	1.706221189	0.514899	1
		Sutterella	1.757369088	1.706221189	0.516654	1
	Enterobacteriaceae			0.332526906	0.715607	0.869
		Escherichia-Shigella		0.332526906	0.715607	0.869
Tenericutes			1.2	0.9125	0.416889	1
	gut metagenome		1.074163549	0.755097109	0.449454	0.878
		gut metagenome	1.074163549	0.755097109	0.449454	0.878