

Differences of gut microbiota in the freshwater blackworm (*Lumbriculus variegatus*: *Oligochaeta*) at two different habitat conditions

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Supplementary information:

- Table S1-S5

Table S1. The number of sequences of the crude and filtered read counts in samples that used in this study.

Samples	Crude read counts (No. of non-filtered seq.)	Filtered read counts (No. of quality-filter passed and singleton excluded seq.)
GP7A	2018	1764
GP7B	1020	899
GP7C	1119	1023
GP8A	906	827
GP8B	1977	1840
GP8C	1515	1432

Table S2. The values of alpha diversity metrics comparison between sampling sites GP7 and GP8. The asterisks indicate significant differences according to two-tailed unpaired *t*-test between sampling sites GP7 and GP8 ($p < 0.05$).

Alpha diversity metrics	GP7	GP8
Shannon diversity *	6.13	5.86
Faith's phylogenetic diversity *	12.28	10.40
Chao1 richness	246.89	207.12
Observed OTUs *	149.33	129.67

Table S3. The relative abundance (%) of phyla in 6 samples from sampling sites GP7 (GP7A-C) and GP8 (GP8A-C).

Phylum	GP7			GP8			<i>p-value</i>
	GP7A	GP7B	GP7C	GP8A	GP8B	GP8C	
Proteobacteria	63.14	73.14	67.25	60.98	63.14	68.04	0.177
<i>Alphaproteobacteria</i>	28.63	43.33	42.75	29.61	34.51	32.94	0.173
<i>Betaproteobacteriales (formerly Betaproteobacteria)*</i>	24.31	23.73	17.45	13.14	11.57	15.29	0.021
<i>Gammaproteobacteria (Except Betaproteobacteriales)**</i>	9.22	5.49	7.06	17.65	16.27	17.84	0.002
<i>Deltaproteobacteria</i>	0.98	0.59	0.00	0.59	0.78	1.96	0.163
Planctomycetes	9.61	3.53	8.04	5.49	9.41	6.67	0.478
Actinobacteria	6.67	7.65	3.73	3.73	4.12	2.35	0.072
Verrucomicrobia*	3.92	6.86	9.61	4.71	1.37	0.78	0.049
Firmicutes	3.92	3.14	4.90	1.57	6.27	2.94	0.406
Cyanobacteria	2.75	1.18	0.20	3.53	1.57	10.59	0.146
Fusobacteria	0.98	0.39	0.20	0.59	5.49	0.78	0.193
Patescibacteria	3.33	0.39	0.98	0.39	0.20	0.39	0.150
Chloroflexi*	0.78	0.59	0.78	0.39	0.39	0.59	0.024
Tenericutes	0.59	0.59	0.59	0.00	0.39	0.59	0.135
WPS-2	0.39	0.78	0.00	0.00	0.00	0.00	0.113
Bacteroidetes	0.39	0.00	0.39	0.00	0.20	0.20	0.219
Dependentiae	0.20	0.20	0.20	0.00	0.20	0.00	0.092
Acidobacteria	0.39	0.00	0.00	0.00	0.00	0.00	0.211
Gemmatimonadetes	0.00	0.00	0.00	0.00	0.39	0.00	0.211
Epsilonbacteraeota	0.00	0.00	0.00	0.00	0.20	0.00	0.211
Rokubacteria	0.00	0.00	0.00	0.00	0.20	0.00	0.211
Uncl. Bacteria	0.00	0.00	0.39	0.00	0.00	0.00	0.211
Unassigned	2.94	1.57	2.75	18.63	6.47	6.08	0.096

The asterisks indicate significant differences according to one-tailed unpaired t-test between sampling sites GP7 and GP8.

* ($p < 0.05$); ** ($p < 0.01$); *** ($p < 0.001$)

Table S4. The top 15 OTUs in 6 samples from sampling sites GP7 (GP7A-C) and GP8 (GP8A-C).

#OTU ID	GP7			GP8			Total Abundance (%)	Phylum	Class	Order	Family	Genus*
	GP7A	GP7B	GP7C	GP8A	GP8B	GP8C						
FTOM01000018.227.1697	6.86	8.04	33.53	2.94	2.16	5.88	9.90	Proteobacteria	Alphaproteobacteria	Rhodobacterales	Rhodobacteraceae	Rhodobacter
EF555461.1.1516	17.06	10.39	1.96	1.57	14.90	10.98	9.48	Proteobacteria	Gammaproteobacteria	Betaproteobacteriales	Burkholderiaceae	Achromobacter
GQ396821.1.1509	13.14	12.16	13.14	2.35	7.65	6.47	9.15	Proteobacteria	Gammaproteobacteria	Betaproteobacteriales	SC-I-84	uncultured bacterium
HQ178940.1.1470	9.22	3.14	5.69	8.82	12.16	13.73	8.79	Proteobacteria	Gammaproteobacteria	Xanthomonadales	Xanthomonadaceae	Arenimonas
denovo.CleanUp.ReferenceOTU306	0.00	0.00	0.00	0.20	41.96	0.00	7.03	Proteobacteria	Alphaproteobacteria	Rhizobiales	Hyphomicrobiaceae	Pedomicrobium
denovo.CleanUp.ReferenceOTU11	14.71	0.00	2.35	1.37	0.39	22.55	6.90	Proteobacteria	Alphaproteobacteria	Rhizobiales	Beijerinckiaceae	alphaI cluster
FJ592534.1.1396	16.47	1.76	0.00	4.31	0.00	8.04	5.10	Proteobacteria	Gammaproteobacteria	Betaproteobacteriales	Rhodocyclaceae	Candidatus Accumilibacter
DQ303296.1.1480	8.82	2.55	2.75	0.98	6.47	2.16	3.95	Proteobacteria	Alphaproteobacteria	Rhizobiales	Xanthobacteraceae	Bradyrhizobium
denovo.CleanUp.ReferenceOTU582	0.00	22.75	0.00	0.00	0.00	0.00	3.79	Proteobacteria	Alphaproteobacteria	Acetobacterales	Acetobacteraceae	
denovo.CleanUp.ReferenceOTU238	3.53	6.67	9.41	0.39	0.78	1.96	3.79	Proteobacteria	Alphaproteobacteria	Rhodobacterales	Rhodobacteraceae	
HQ114181.1.1456	2.16	1.18	1.76	3.53	9.61	3.92	3.69	Proteobacteria	Gammaproteobacteria	Cellvibrionales	Haliaceae	OM60(NOR5) clade
KC749107.1.1486	5.29	4.51	1.96	1.96	2.75	2.94	3.24	Proteobacteria	Alphaproteobacteria	Rhizobiales	Rhizobiales Incertae Sedis	uncultured
FPLK01000581.11.1482	6.47	2.94	3.53	1.18	4.12	0.59	3.14	Proteobacteria	Alphaproteobacteria	Rhizobiales	Rhizobiales Incertae Sedis	uncultured
EF438215.1.1399	0.00	0.00	0.00	1.76	0.00	15.29	2.84	Cyanobacteria	Oxyphotobacteria	Nostocales	Phormidiaceae	Tychonema CCAP 1459-11B
HG970980.1.1567	0.78	0.59	0.00	0.20	14.90	0.59	2.84	Fusobacteria	Fusobacteriia	Fusobacteriales	Fusobacteriaceae	Fusobacterium

Table S5. Comparison of shared or not-shared OTU numbers between sampling sites GP7 and GP8.

No of OTUs	GP7A	GP7B	GP7C	GP8A	GP8B	GP8C
All_shared	18	18	18	18	18	18
GP7_shared	32	32	32	15	19	11
GP8_shared	10	9	8	22	22	22
not_shared	119	75	77	68	84	72