

## **Supplemental Material**

# **Protein- and RNA-Enhanced Fermentation by Gut Microbiota of the Earthworm *Lumbricus terrestris***

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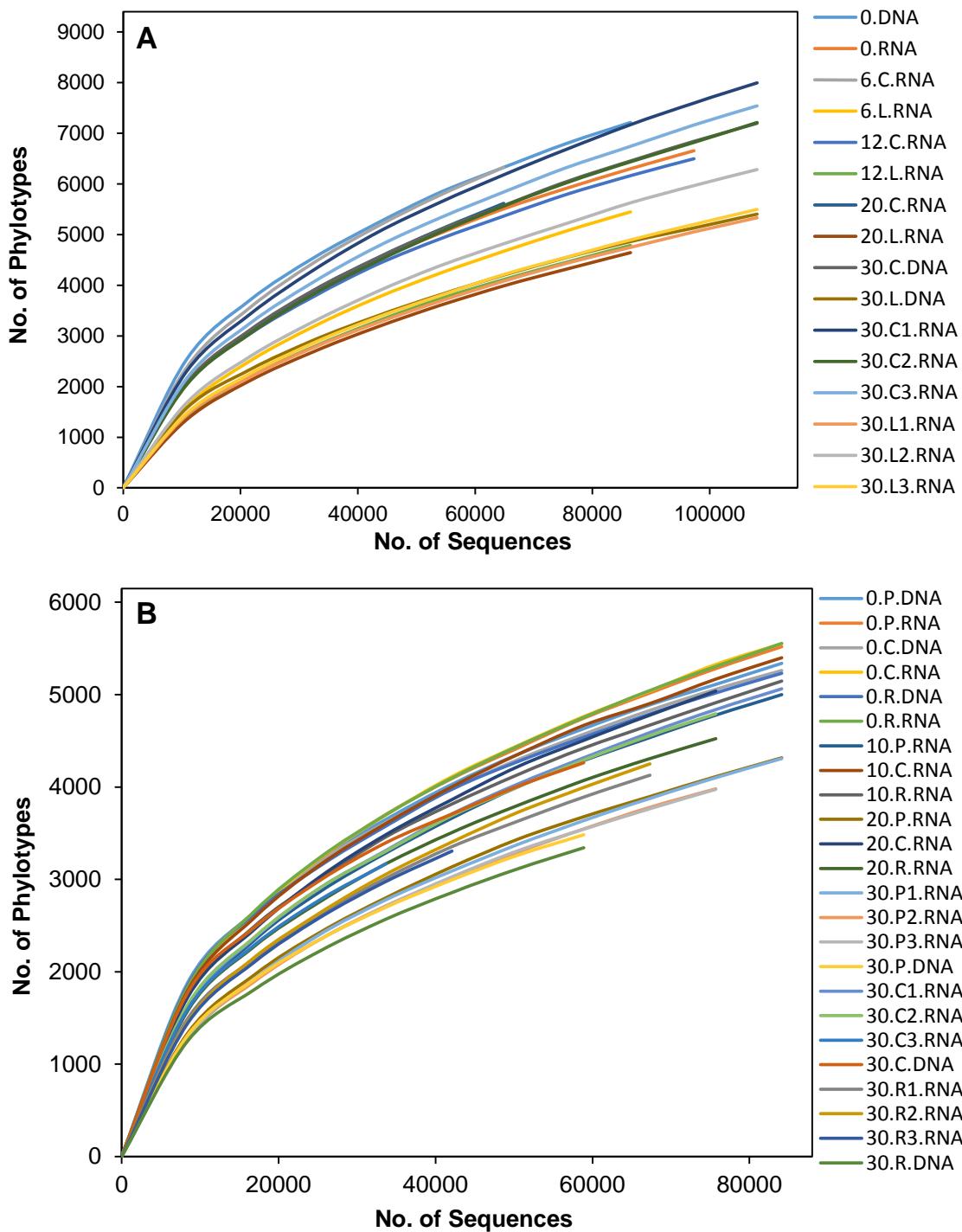
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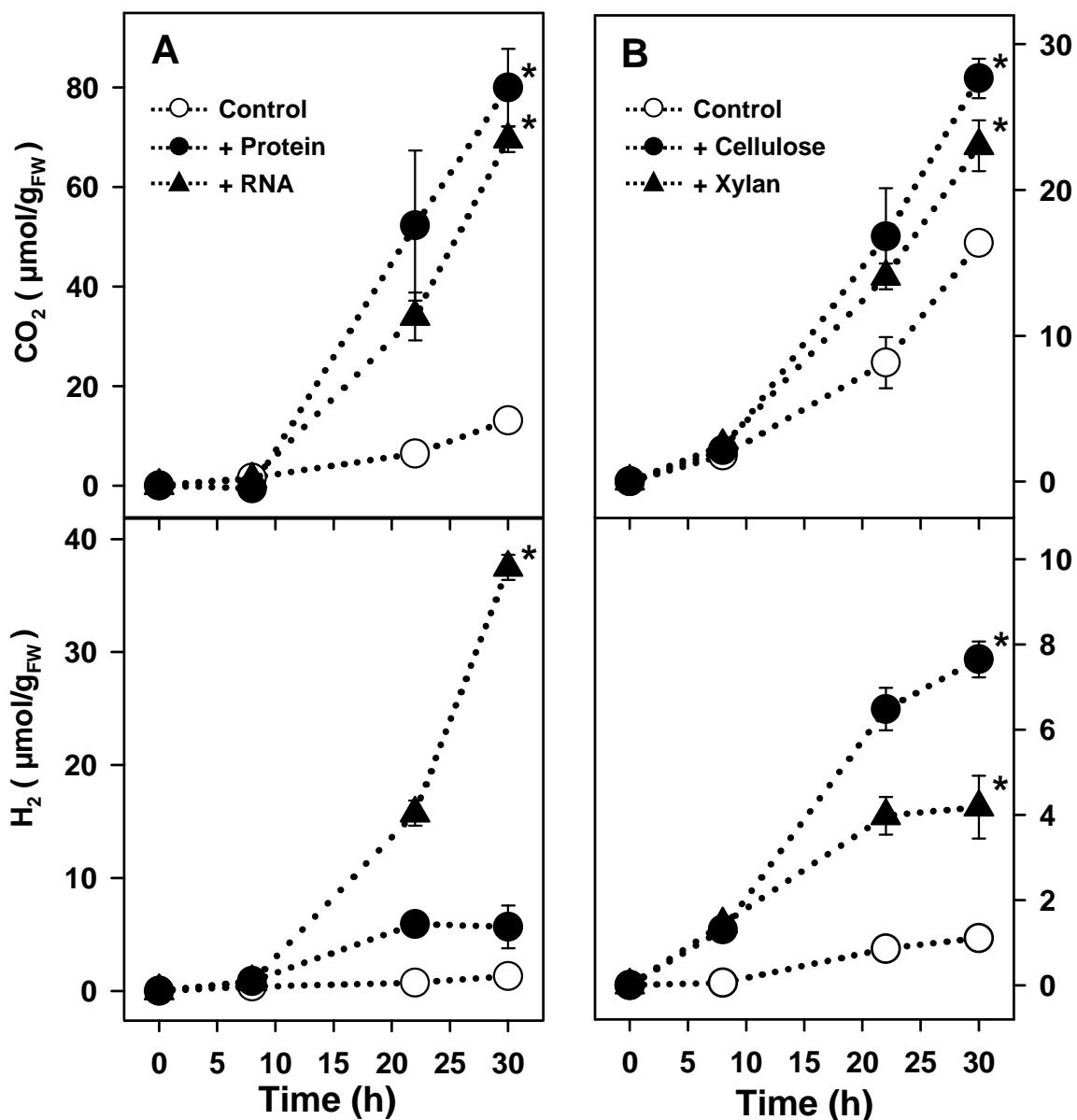
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**Figure S1.** Rarefaction analyses of bacterial 16S rRNA and 16S rRNA gene sequences obtained from (A) cell-lysate treatment and (B) protein and RNA treatments. Phylotypes were based on a 99% (panel A) and 97% (panel B) sequence similarity cutoff. Abbreviations: 0, 6, 10, 12, 20, 30 indicate the time of sampling in hours; C, unsupplemented control; L, lysate treatment; P, protein treatment; R, RNA treatment. For both panels, RNA or DNA samples of the three replicates were always pooled except for RNA samples at 30 hour, the identification numbers (e.g., C1) of which indicate the respective replicates.



**Figure S2.** Effect of biopolymers on the formation of H<sub>2</sub> and CO<sub>2</sub> in anoxic microcosms of *L. terrestris* gut contents. The amount of biopolymer-derived carbon added per microcosm (10 ml containing 1 g<sub>FW</sub> gut content) approximated 2.4 mmoles (corresponds to 54 mg protein, 81 mg RNA, 68 mg cellulose, or 63 mg xylan). Controls lacked supplemental biopolymers. Values are the arithmetic average of three replicate analyses, and error bars indicate the standard deviations. Some standard deviations are smaller than the size of the symbol and therefore not apparent. The asterisk (\*) indicates significant *P* values (*P* ≤ 0.05) of control vs. protein, RNA, cellulose, or xylan treatments at the end of incubation. *P* values were calculated by t-Test with unequal variances. FW, fresh weight.

**Table S1.** Effect of cell lysates from *S. cerevisiae* and *E. coli* on the formation of CO<sub>2</sub> and H<sub>2</sub> in anoxic microcosms of *L. terrestris* gut contents.

Treatment <sup>a</sup>	Replicate	Gaseous products ( $\mu\text{mol/g}_{\text{FW}}$ )					
		pH		CO <sub>2</sub>		H <sub>2</sub>	
		1	2	1	2	1	2
Control		6.8	6.9	18.6	21.6	2.2	2.4
<i>E. coli</i>		6.6	6.6	60.4	55.3	5.9	9.7
<i>S. cerevisiae</i>		6.6	6.6	63.8	63.6	14.6	15.7

<sup>a</sup>The amount of carbon derived from filter sterilized *S. cerevisiae* lysate (5.1% dry weight) and *E. coli* lysate (5.3% dry weight) added per microcosm (10 ml containing 1 g<sub>FW</sub> gut content) approximated 2.0 mmoles and 2.2 mmoles, respectively. Filter-sterilized lysate alone did not display any fermentation activity. Control lacked supplemental lysate. Earthworms from which gut contents were obtained were maintained at 15°C, and microcosms were incubated at 15°C for 44 hours. FW , fresh weight.

**Table S2.** Statistical *P* values of the products formed in (A) *S. cerevisiae* lysate, (B) protein, and (C) RNA treatments.

**(A) Lysate treatment (Figure 1)<sup>a</sup>**

Products																		
CO <sub>2</sub>		H <sub>2</sub>		Succinate		Lactate		Formate		Acetate		Propionate		Butyrate		Methylbutyrate		
C	L	C	L	C	L	C	L	C	L	C	L	C	L	C	L	C	L	
<b>Mean value<sup>b</sup></b>	30	149	0.9	33	4.1	58	1.3	1.5	6.2	48	29	262	7.7	14	2.0	13	10	96
<b>Variance</b>	479	1046	0.0	3.8	0.0	142	0.0	0.0	0.9	50	31	2656	3.2	0.2	0.1	0.1	1.6	88
<b>P value<sup>c</sup></b>	0.001		0.006		0.016		0.103		0.009		0.016		0.030		0.000		0.004	

**(B) Protein treatment (Figure 3)<sup>a</sup>**

CO <sub>2</sub>		H <sub>2</sub>		Succinate		Lactate		Formate		Acetate		Propionate		Butyrate		Methylbutyrate		
C	P	C	P	C	P	C	P	C	P	C	P	C	P	C	P	C	P	
<b>Mean value<sup>b</sup></b>	7.1	38	0.5	3.2	0.0	0.0	0.3	0.4	1.5	11	15	90	3.5	19	0.9	11	3.0	30
<b>Variance</b>	0.2	3.5	0.0	0.1	0.0	0.0	0.0	0.0	0.1	0.7	0.2	7.4	0.1	0.5	0.0	0.0	0.0	8.6
<b>P value<sup>c</sup></b>	0.001		0.001		-		0.502		0.016		0.010		0.000		0.009		0.004	

**(C) RNA treatment (Figure 3)<sup>a</sup>**

CO <sub>2</sub>		H <sub>2</sub>		Succinate		Lactate		Formate		Acetate		Propionate		Butyrate		Methylbutyrate		
C	R	C	R	C	R	C	R	C	R	C	R	C	R	C	R	C	R	
<b>Mean value<sup>b</sup></b>	7.1	76	0.5	48	0.0	13	0.3	6.1	2.8	26	15	59	3.5	3.7	0.9	0.4	3.0	2.1
<b>Variance</b>	0.2	39	0.0	9.6	0.0	0.1	0.0	0.0	0.1	0.4	0.2	4.8	0.1	0.0	0.0	0.0	0.0	0.4
<b>P value<sup>c</sup></b>	0.001		0.003		0.000		0.000		0.000		0.001		0.775		0.118		0.158	

<sup>a</sup>C, L, P, and R corresponds to unsupplemented control, lysate, protein, and RNA treatments.

<sup>b</sup>Mean value (*n* = 3) of the different fermentation products in  $\mu\text{mol/g}_{\text{FW}}$ . FW, fresh weight.

<sup>c</sup>*P* values were calculated by t-Test with unequal variances. Values are significant at *P* ≤ 0.05.

**Table S3.** Time-resolved alpha diversity of the microbial community in the *S. cerevisiae* lysate treatment.

Sampling time:	0 h		6 h		12 h		20 h	
Treatment: <sup>a</sup>	DNA	RNA	C.RNA	L.RNA	C.RNA	L.RNA	C.RNA	L.RNA
Number of sequences	90591	105034	70681	90615	104319	87844	70036	96059
Observed PTs <sup>b</sup> (normalized) <sup>c</sup>	7384 (6328)	6911 (5506)	6625 (6326)	5596 (4670)	6720 (5365)	4833 (4112)	5848 (5618)	4917 (3989)
Chao1 (normalized) <sup>c</sup>	11975 (10608)	11553 (9751)	11842 (11419)	10159 (8995)	11199 (9460)	8722 (7864)	10712 (10409)	9341 (7911)
Shannon (normalized) <sup>c</sup>	9.8 (9.7)	7.3 (7.3)	8.2 (8.1)	6.1 (6.0)	7.9 (7.9)	5.8 (5.8)	7.2 (7.2)	6.2 (6.2)
Sampling time:	30 h							
Treatment: <sup>a</sup>	C1.RNA	C2.RNA	C3.RNA	L1.RNA	L2.RNA	L3.RNA	C.DNA	L.DNA
Number of sequences	109807	147651	126676	114515	126281	142222	111359	122114
Observed PTs <sup>b</sup> (normalized) <sup>c</sup>	8086 (6179)	8411 (5566)	8150 (5841)	5509 (4083)	6825 (4822)	6332 (4214)	7323 (5573)	5745 (4200)
Chao1 (normalized) <sup>c</sup>	14324 (11675)	14417 (10764)	13660 (10822)	10514 (8101)	12345 (9390)	11492 (8483)	12972 (10395)	9945 (7761)
Shannon (normalized) <sup>c</sup>	8.3 (8.3)	7.6 (7.6)	7.7 (7.6)	6.6 (6.6)	7.2 (7.1)	6.7 (6.7)	8.3 (8.2)	7.2 (7.2)

<sup>a</sup>C and L corresponds to unsupplemented control and cell lysate treatments, respectively. RNA or DNA samples of the three replicates were always pooled except for RNA samples at 30 h (the numbers of each treatment at 30 h [e.g., C1] indicate the respective replicate). DNA, 16S rRNA genes; RNA, 16S rRNA.

<sup>b</sup>Phylotypes (PTs) were clustered based on a sequence similarity cut-off of 99%.

<sup>c</sup>The data sets were normalized to 64,864 sequences for comparison of amplicon libraries of different sizes.

**Table S4.** Statistical *P* values of abundant families that were stimulated by supplemental lysate, protein, or RNA.<sup>a</sup>

**(A) Lysate Treatment (Figure 2)**

Family	Treatment	Mean	Variance	<i>P</i> value <sup>b</sup>
<i>Aeromonadaceae</i>	Control	0.2	0.0	0.010
	Lysate	1.4	0.0	
<i>Clostridiaceae</i>	Control	1.3	0.0	0.007
	Lysate	11.9	2.4	
<i>Enterobacteriaceae</i>	Control	0.3	0.0	0.011
	Lysate	12.6	4.9	
<i>Lachnospiraceae</i>	Control	0.4	0.0	0.018
	Lysate	9.7	4.8	
<i>Peptostreptococcaceae</i>	Control	10.3	5.6	0.003
	Lysate	23.1	6.1	

**(B) Protein and RNA Treatments (Figure 4)**

Family	Treatment	Mean	Variance	<i>P</i> Value <sup>b</sup>
<i>Aeromonadaceae</i>	Control	0.2	0.0	0.005 (0.005)
	RNA	19.1	5.5	
	Protein	0.4	0.0	
<i>Clostridiaceae</i>	Control	1.5	0.2	0.000 (0.001)
	Protein	12.9	1.3	
	RNA	3.3	0.2	
<i>Fusobacteriaceae</i>	Control	1.0	0.3	0.029 (0.023)
	Protein	8.4	4.7	
	RNA	0.2	0.0	
<i>Peptostreptococcaceae</i>	Control	4.6	0.5	0.006 (0.005)
	Protein	22.5	5.8	
	RNA	2.9	0.4	

<sup>a</sup>Families were designated (a) abundant when a family had a relative abundance of  $\geq 5\%$  in at least one sampling period and (b) stimulated when the increase in relative abundance over time was more pronounced in at least one treatment (lysate, protein, or RNA) compared to the respective unsupplemented control.

<sup>b</sup>*P* values of control vs. lysate, protein, or RNA treatments were calculated from relative abundances at the end of the 30 h incubation by t-Test with unequal variances (parenthetical values indicate *P* values of protein vs. RNA treatments). Values are significant at *P*  $\leq 0.05$ .

**Table S5.** Fatty acid profiles of anoxic microcosms of *L. terrestris* gut contents supplemented with different biopolymers.

Treatment <sup>a</sup>	Time (h)	Products ( $\mu\text{mol/g}_{\text{FW}}$ ) <sup>b</sup>						
		Acetate	Succinate	Formate	Propionate	Butyrate	Methylbutyrate	Lactate
<b>(A) Protein and RNA</b>								
Control	0	2.2 ± 0.5	0.5 ± 0.0	3.6 ± 0.4	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.6 ± 0.1
	30	23 ± 2.8	0.0 ± 0.0	2.9 ± 0.8	4.5 ± 0.3	1.9 ± 0.1	5.3 ± 0.1	0.5 ± 0.1
Protein	0	2.7 ± 0.1	0.4 ± 0.1	2.7 ± 0.3	0.0 ± 0.0	0.9 ± 0.2	0.0 ± 0.0	1.2 ± 0.2
	30	46 ± 5.6*	0.0 ± 0.0	13 ± 9.2	22 ± 1.7*	25 ± 2.6*	53 ± 1.3*	5.8 ± 2.4
RNA	0	18 ± 1.4	4.9 ± 0.3	2.3 ± 0.7	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	4.9 ± 0.3
	30	87 ± 5.3*	19 ± 1.4*	39 ± 1.3*	8.0 ± 0.9*	3.2 ± 1.4	5.5 ± 1.2	12 ± 0.5*
<b>(B) Cellulose and Xylan</b>								
Control	0	1.2 ± 0.1	0.5 ± 0.1	3.2 ± 0.2	0.0 ± 0.0	0.0 ± 0.0	0.7 ± 0.3	0.3 ± 0.0
	30	25 ± 1.4	1.9 ± 0.2	2.5 ± 0.8	3.7 ± 0.1	0.6 ± 0.0	6.8 ± 0.2	0.8 ± 0.1
Cellulose	0	1.8 ± 0.2	0.4 ± 0.1	0.7 ± 0.3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.3 ± 0.1
	30	33 ± 7.0	3.1 ± 0.1*	2.8 ± 2.9	4.0 ± 0.2	0.7 ± 0.0	7.1 ± 0.3	2.0 ± 0.2*
Xylan	0	12 ± 0.7	0.6 ± 0.0	1.3 ± 0.4	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.4 ± 0.0
	30	45 ± 4.4*	2.7 ± 0.1*	3.0 ± 0.1	4.3 ± 0.1*	0.9 ± 0.0*	7.2 ± 0.3	1.6 ± 0.1*

<sup>a</sup>The amount of biopolymer-derived carbon added per microcosm (10 ml containing 1 g<sub>FW</sub> gut content) approximated 2.4 mmoles (corresponds to 54 mg protein, 81 mg RNA, 68 mg cellulose, or 63 mg xylan). Controls lacked supplemental biopolymers. Values are the arithmetic average of three replicate analyses (± standard derivation). Time resolved production of CO<sub>2</sub> and H<sub>2</sub> are shown in Supplemental Figure S2. FW, fresh weight.

<sup>b</sup>The asterisk (\*) indicates significant *P* values (*P* ≤ 0.05) of control vs. protein, RNA, cellulose, or xylan treatments at the end of incubation. *P* values were calculated by t-Test with unequal variances.

**Table S6.** Effect of different amounts of protein or RNA on the formation of CO<sub>2</sub> or H<sub>2</sub>, respectively, in anoxic microcosms of *L. terrestris* gut contents.

Treatment	Carbon (mmol) <sup>a</sup>	μmol/g <sub>FW</sub> <sup>b</sup>	P Value <sup>c</sup>
<b>Protein</b>		<b>CO<sub>2</sub></b>	
	0.0	10.3 ± 2.8	
	0.5	25.0 ± 2.0	0.002
	2.0	55.6 ± 5.1	0.000
<b>RNA</b>		<b>H<sub>2</sub></b>	
	0.0	0.8 ± 0.1	
	0.5	8.7 ± 1.8	0.002
	2.0	47.3 ± 9.3	0.001

<sup>a</sup>The amount of biopolymer-derived carbon added per 10 ml microcosm containing 1 g<sub>FW</sub> gut content (one mmole carbon corresponds to 22.5 mg protein and 33.8 mg RNA).

<sup>b</sup>Amounts of CO<sub>2</sub> and H<sub>2</sub> at the end of incubation (30 h). Values are the arithmetic average of three replicate analyses (± standard deviation). FW, fresh weight.

<sup>c</sup>P values were calculated by t-Test with different variances and are based on the difference between the unsupplemented control and the supplemented treatment. Values are significant at *P* ≤ 0.05.

**Table S7.** Fermentation profiles (A) and estimated recoveries (B) of anoxic microcosms of *L. terrestris* gut contents supplemented with casamino acids or ribose.

**(A) Fermentation Profile<sup>a</sup>**

Treatment	Time (h)	pH	Products ( $\mu\text{mol/g}_{\text{FW}}^{\text{b}}$ )									
			$\text{CO}_2$	$\text{H}_2$	Acetate	Ethanol	Succinate	Lactate	Formate	Propionate	Butyrate	Methylbutyrate
<b>Control</b>	0	$7.0 \pm 0.0$	$0.0 \pm 0.0$	$0.0 \pm 0.0$	$2.4 \pm 0.2$	$0.0 \pm 0.0$	$0.7 \pm 0.0$	$1.0 \pm 0.1$	$1.0 \pm 0.1$	$0.0 \pm 0.0$	$0.0 \pm 0.0$	$0.0 \pm 0.0$
	30	$7.0 \pm 0.0$	$7.5 \pm 1.0$	$0.7 \pm 0.1$	$16 \pm 0.4$	$0.0 \pm 0.0$	$0.4 \pm 0.1$	$0.8 \pm 0.1$	$0.7 \pm 0.1$	$3.0 \pm 0.1$	$1.3 \pm 0.1$	$1.7 \pm 0.1$
<b>Casamino acids<sup>c</sup></b>	0	$7.0 \pm 0.0$	$0.0 \pm 0.0$	$0.0 \pm 0.0$	$2.5 \pm 0.1$	$0.0 \pm 0.0$	$0.6 \pm 0.0$	$1.3 \pm 0.0$	$1.2 \pm 0.2$	$0.0 \pm 0.0$	$0.0 \pm 0.0$	$0.0 \pm 0.0$
	30	$6.9 \pm 0.0$	$33 \pm 2.8^*$	$3.8 \pm 0.2^*$	$90 \pm 3.4^*$	$0.0 \pm 0.0$	$2.6 \pm 0.7^*$	$1.4 \pm 0.3$	$3.2 \pm 0.9^*$	$16 \pm 1.1^*$	$8.7 \pm 0.4^*$	$14 \pm 1.5^*$
<b>Ribose<sup>d</sup></b>	0	$7.0 \pm 0.0$	$0.0 \pm 0.0$	$0.0 \pm 0.0$	$1.9 \pm 0.5$	$0.0 \pm 0.0$	$0.0 \pm 0.0$	$0.9 \pm 0.3$	$0.9 \pm 0.2$	$0.0 \pm 0.0$	$0.0 \pm 0.0$	$0.0 \pm 0.0$
	30	$6.8 \pm 0.0$	$12 \pm 1.0^*$	$3.9 \pm 0.8^*$	$32 \pm 3.1^*$	$19 \pm 2.3^*$	$1.9 \pm 0.4^*$	$1.3 \pm 0.1^*$	$7.8 \pm 0.4^*$	$3.1 \pm 0.2$	$1.5 \pm 0.2$	$2.0 \pm 0.2$

**(B) Recoveries (%)<sup>e</sup>**

		$\text{CO}_2$	$\text{H}_2$	Acetate	Ethanol	Succinate	Lactate	Formate	Propionate	Butyrate	Methylbutyrate	Total
<b>Casamino acids<sup>c</sup></b>	Carbon	2.1	na	12.3	-	0.8	0.0	0.2	3.2	2.5	5.0	<b>26.2</b>
	Reducing Equivalents	na	0.1	11.7	-	0.6	0.0	0.1	3.6	2.9	6.2	<b>25.3</b>
<b>Ribose<sup>d</sup></b>	Carbon	3.5	na	24.0	26.9	2.3	2.0	5.2	0.1	0.6	1.2	<b>65.8</b>
	Reducing Equivalents	na	1.2	24.0	40.3	2.0	2.0	2.6	0.2	0.8	1.5	<b>74.6</b>

<sup>a</sup>Controls lacked supplemental substrates. Values are the arithmetic average of three replicate analyses ( $\pm$  standard derivation). FW, fresh weight.

<sup>b</sup>The asterisk (\*) indicates significant *P* values ( $P \leq 0.05$ ) of control vs. casamino acids or ribose treatments at the end of incubation. *P* were calculated by t-Test with unequal variances.

<sup>c</sup>The amount of casamino acid-derived carbon added per microcosm (10 ml containing 1 g<sub>FW</sub> gut content) approximated 1.2 mmoles (corresponds to 25.2 mg casamino acids; based on the chemical formula of casein [CH<sub>1.95</sub>N<sub>0.25</sub>O<sub>0.49</sub>S<sub>0.007</sub>]<sub>n</sub>, yielding an average oxidation level of -0.214 for carbon).

<sup>d</sup>The amount of ribose per microcosm approximated 5 mM, of which 3 mM were consumed.

<sup>e</sup>Recoveries were calculated at the end of the 30 hour incubation and are based on the arithmetic average of three replicate analyses. -, no net increase of the product during the 30 h incubation. na, not applicable.

**Table S8.** Time-resolved alpha diversity of the microbial community in protein and RNA treatments.

Sampling Time	0 h						10 h			20 h			
	Treatment: <sup>a</sup>	C.DNA	P.DNA	R.DNA	C.RNA	P.RNA	R.RNA	C.RNA	P.RNA	R.RNA	C.RNA	P.RNA	R.RNA
Number of sequences		95270	98173	93122	96621	98821	101284	108667	118733	103052	80188	90359	82138
Observed PTs <sup>b</sup> (normalized) <sup>c</sup>		5527 (3478)	5660 (3572)	5444 (3404)	5886 (3463)	5898 (3517)	6004 (3504)	6008 (3572)	5780 (3392)	5578 (3353)	5175 (2910)	4452 (2522)	4689 (2720)
Chao1 (normalized) <sup>c</sup>		7925 (5967)	8092 (5999)	7908 (5797)	9270 (6431)	9151 (6383)	9723 (6339)	9382 (6094)	9177 (5734)	8510 (5744)	8668 (6121)	7092 (4988)	7542 (5481)
Shannon (normalized) <sup>c</sup>		9.3 (9.2)	9.3 (9.2)	9.3 (9.2)	8.0 (7.9)	8.3 (8.2)	8.3 (8.2)	8.4 (8.3)	7.6 (7.5)	8.2 (8.1)	8.0 (7.9)	6.6 (6.5)	7.6 (7.5)
Sampling Time	30 h												
	Treatment: <sup>a</sup>	C1.RNA	C2.RNA	C3.RNA	P1.RNA	P2.RNA	P3.RNA	R1.RNA	R2.RNA	R3.RNA	C.DNA	P.DNA	R.DNA
Number of sequences		84481	83599	35563	114393	80012	78166	73608	69987	48891	59211	61820	63663
Observed PTs <sup>b</sup> (normalized) <sup>c</sup>		5077 (2866)	5011 (2855)	3247 (1788)	4944 (2769)	4082 (2237)	4033 (2241)	4299 (2421)	4329 (2355)	3544 (1911)	4274 (2583)	3558 (1997)	3465 (1909)
Chao1 (normalized) <sup>c</sup>		8266 (5806)	8117 (5742)	5378 (5302)	8014 (5075)	6927 (4930)	6730 (4713)	7128 (5208)	7330 (5539)	6063 (5242)	6557 (5439)	5838 (4707)	5739 (4589)
Shannon (normalized) <sup>c</sup>		7.7 (7.6)	7.9 (7.8)	7.9 (7.9)	6.7 (6.6)	6.7 (6.6)	6.7 (6.6)	7.5 (7.4)	7.4 (7.3)	7.4 (7.3)	9.3 (9.3)	7.1 (7.0)	6.2 (6.2)

<sup>a</sup>C, P, and R corresponds to unsupplemented control, protein, and RNA treatments, respectively. RNA or DNA samples of the three replicates were always pooled except for RNA samples at 30 h (the numbers of each treatment at 30 h [e.g., C1] indicate the respective replicate). DNA, 16S rRNA genes; RNA, 16S rRNA.

<sup>b</sup>Phylotypes (PTs) were clustered based on a sequence similarity cut-off of 97%.

<sup>c</sup>The data sets were normalized to 33,658 sequences for comparison of amplicon libraries of different sizes.

**Table S9.** Instrumentation utilized for analyses of organic acids and gases.

Parameter	Chromatograph		
	Hewlett Packard 1090 Series II	Hp Hewlett 5890 Packard Series II	Schambeck SRI 8610C
<b>Detected compounds</b>	Organic acids	Hydrogen	Carbon dioxide
<b>Column</b>	Rezex ROA-Organic Acids (300 x 7,8 mm; Phenomenex, Torrance, CA, USA)	Molecular sieve13X, 2 m x 1/8" (Restek, Bellefonte, PA, USA)	Hayesep-D 2 m x 1/8" (SRI Instruments, Earl St. Torrance, CA, USA)
<b>Oven temperature</b>	60 °C	60 °C	80 °C
<b>Detector</b>	G1362A refractive index detector (RID)	Thermal conductivity detector (WLD)	Thermal conductivity detector (WLD)
<b>Detector temperature</b>	40 °C	175 °C	175 °C
<b>Flow rate</b>	0.8 ml/min	20 ml/min	20 ml/min
<b>Injections volume</b>	20 µl	0.1 ml	0.1 ml
<b>Software</b>	ChemStation (Agilent Technologies, Böb-lingen, Germany)	EuroChrom Software for Windows (Ver: Basic Edition V3. 05, Wissenschaftliche Gerätebau, Berlin, Germany)	Peak simple Software (Ver: 4.20, SRI Instruments)
<b>Mobile phase/ Carrier gas</b>	4 mM H3PO4	Argon	Helium

**Table S10.** Summary of families of the *S. cerevisiae* lysate treatment (the most abundant taxa are displayed in Figure 2).<sup>a</sup>

Phylum, Class, Family <sup>c</sup>	Sampling time:	0 h		6 h		12 h		20 h	
		Treatment <sup>b</sup> :	D	R	C.R.	L.R.	C.R.	L.R.	C.R.
<b>Actinobacteria,</b>									
<i>Acidimicrobia</i> ,									
<i>Acidimicrobiaceae</i> (8)		2.1	1.2	1.7	0.6	1.5	0.6	1.3	0.7
TM214 group (4)		1.0	0.3	0.4	0.2	0.6	0.2	0.3	0.2
Unassigned <i>Acidimicrobiales</i> (6)		1.0	0.4	0.6	0.3	0.8	0.2	0.5	0.2
<i>Actinobacteria</i> ,									
<i>Mycobacteriaceae</i> (2)		0.5	0.2	0.2	0.1	0.2	0.0	0.1	0.0
<i>Nakamurellaceae</i> (1)		0.3	0.3	0.3	0.1	0.4	0.1	0.2	0.1
<i>Cellulomonadaceae</i> (2)		0.1	0.5	0.6	0.3	0.7	0.2	0.5	0.2
<i>Intrasporangiaceae</i> (1)		0.4	0.3	0.5	0.1	0.6	0.1	0.4	0.1
<i>Microbacteriaceae</i> (5)		1.7	0.7	1.0	0.4	1.6	0.3	0.9	0.4
<i>Micrococcaceae</i> (1)		0.3	0.2	0.2	0.1	0.2	0.1	0.1	0.1
<i>Micromonosporaceae</i> (1)		0.1	0.1	0.1	0.0	0.1	0.0	0.1	0.0
<i>Nocardioidaceae</i> (7)		1.7	0.6	0.7	0.3	0.9	0.3	0.7	0.3
<i>Propionibacteriaceae</i> (3)		0.5	0.3	0.4	0.1	0.4	0.1	0.3	0.1
<i>Streptomycetaceae</i> (2)		0.6	0.4	0.3	0.1	0.4	0.1	0.2	0.1
Unassigned <i>Actinobacteria</i> (4)		2.2	1.0	1.1	0.4	1.1	0.4	0.8	0.4
<i>Thermoleophilia</i> ,									
<i>Gaiellaceae</i> (2)		0.8	0.2	0.2	0.1	0.3	0.1	0.1	0.1
Unassigned <i>Gaiellaceae</i> (5)		1.9	0.3	0.3	0.1	0.5	0.1	0.3	0.1
<i>Patulibacteraceae</i> (1)		0.1	0.2	0.2	0.1	0.2	0.1	0.1	0.1
<i>Solirubrobacteraceae</i> (2)		0.2	0.4	0.4	0.1	0.5	0.1	0.3	0.1
Unassigned <i>Solirubrobacteraceae</i> (3)		1.1	0.2	0.2	0.1	0.3	0.1	0.2	0.1
<i>Chloroflexi</i> ,									
Unassigned <i>Chloroflexi</i> (3)		1.5	0.5	0.5	0.2	0.7	0.2	0.4	0.3
<i>Firmicutes</i> ,									
<i>Bacilli</i> ,									
<i>Bacillaceae</i> (4)		1.5	2.0	1.3	1.9	1.6	0.4	1.0	0.5
<i>Paenibacillaceae</i> (1)		0.0	0.0	0.0	0.1	0.2	0.1	0.1	0.2
<i>Clostridia</i> ,									
<i>Clostridiaceae</i> (10) [CL8, CL10, CL18, CL15]		0.0	0.0	0.1	1.0	0.6	4.3	0.7	5.9
<i>Lachnospiraceae</i> (5) [CL6]		0.0	0.0	0.0	0.0	0.0	0.5	0.0	3.4
<i>Peptostreptococcaceae</i> (14) [CL2, CL5]		0.2	0.4	4.6	36.6	12.1	38.6	17.1	27.8
<i>Proteobacteria</i> ,									
<i>Alphaproteobacteria</i> ,									
<i>Bradyrhizobiaceae</i> (1)		1.2	1.8	1.7	0.6	1.2	0.4	1.0	0.4
<i>Hyphomicrobiaceae</i> (2)		0.2	0.2	0.3	0.1	0.3	0.1	0.2	0.1
<i>Methylobacteriaceae</i> (2)		0.2	0.3	0.2	0.1	0.3	0.1	0.2	0.1
<i>Phyllobacteriaceae</i> (1)		0.3	0.4	0.5	0.2	0.5	0.1	0.3	0.1

Sampling time:	0 h		6 h		12 h		20 h		
	Treatment <sup>b</sup> :	D	R	C.R	L.R	C.R	L.R	C.R	L.R
<b>Phylum, Class, Family<sup>c</sup></b>									
<i>Rhodobiaceae</i> (1)		1.4	0.6	0.7	0.3	0.7	0.2	0.6	0.2
<i>Xanthobacteraceae</i> (4)		2.5	1.1	1.3	0.4	1.6	0.4	1.2	0.4
<i>Acetobacteraceae</i> (1)		0.0	0.2	0.1	0.1	0.1	0.0	0.1	0.1
<i>Rhodobacteraceae</i> (3)		0.2	0.8	0.7	0.3	0.7	0.3	0.4	0.3
<i>Rhodospirillaceae</i> (1)		0.0	0.1	0.1	0.0	0.1	0.0	0.1	0.0
Unassigned <i>Rhodospirillales</i> (6)		0.0	0.8	1.2	1.2	0.5	1.6	0.4	1.1
<i>Deltaproteobacteria</i> ,									
<i>Nitrospinaceae</i> (3)		0.2	0.8	0.8	0.3	0.8	0.3	0.7	0.3
<i>Sorangiineae</i> (2)		0.1	0.3	0.3	0.1	0.4	0.1	0.2	0.1
Unassigned <i>Deltaproteobacteria</i> (2)		0.6	0.5	0.3	0.2	0.4	0.1	0.3	0.1
<i>Gammaproteobacteria</i> ,									
<i>Aeromonadaceae</i> (3) [CL7]		5.8	3.0	0.4	10.4	0.4	8.9	0.6	7.2
<i>Shewanellaceae</i> (1)		0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1
<i>Enterobacteriaceae</i> (7) [CL4]		0.6	0.4	0.2	3.5	0.3	8.6	0.6	14.6
<i>Planctomycetes</i> ,									
<i>Planctomycetacia</i> ,									
<i>Planctomycetaceae</i> (13)		0.8	1.1	1.6	0.8	1.3	0.8	2.0	0.8
<i>Tenericutes</i> ,									
<i>Mollicutes</i> ,									
<i>Mycoplasmataceae</i> (4)		6.0	36.8	26.8	14.0	23.8	10.4	27.1	12.9
<i>Verrucomicrobia</i> ,									
<i>Spartobacteria</i> ,									
<i>Xiphinematobacteraceae</i> (2)		3.3	4.2	6.5	2.5	3.2	2.5	5.1	2.5
Unassigned <i>Chthoniobacterales</i> (1)		1.1	0.2	0.2	0.1	0.1	0.1	0.1	0.1

Sampling time:	30 h								
	Treatment <sup>b</sup> :	D.C	D.L	C1.R	C2.R	C3.R	L1.R	L2.R	L3.R
<b>Phylum, Class, Family<sup>c</sup></b>									
<i>Actinobacteria</i> ,									
<i>Acidimicrobia</i> ,									
<i>Acidimicrobiaceae</i> (8)		1.4	1.3	2.0	1.9	1.5	0.7	1.2	0.9
TM214 group (4)		0.6	0.6	0.6	0.5	0.4	0.3	0.3	0.3
Unassigned <i>Acidimicrobiales</i> (6)		0.6	0.5	0.7	0.6	0.6	0.3	0.4	0.3
<i>Actinobacteria</i> ,									
<i>Mycobacteriaceae</i> (2)		0.2	0.3	0.2	0.1	0.2	0.1	0.1	0.1
<i>Nakamurellaceae</i> (1)		0.1	0.2	0.4	0.4	0.3	0.3	0.2	0.1
<i>Cellulomonadaceae</i> (2)		0.2	0.1	0.6	0.7	0.6	0.3	0.3	0.3
<i>Intrasporangiaceae</i> (1)		0.3	0.3	0.5	0.4	0.4	0.2	0.2	0.2

Phylum, Class, Family <sup>c</sup>	Sampling time: Treatment <sup>b:</sup>	30 h							
		D.C	D.L	C1.R	C2.R	C3.R	L1.R	L2.R	L3.R
<i>Microbacteriaceae</i> (5)		0.9	1.2	1.5	1.4	1.6	1.1	0.6	0.5
<i>Micrococcaceae</i> (1)		0.1	0.2	0.2	0.2	0.1	0.1	0.1	0.1
<i>Micromonosporaceae</i> (1)		0.1	0.1	0.1	0.1	0.1	0.0	0.0	0.0
<i>Nocardioidaceae</i> (7)		1.0	1.0	0.9	0.9	0.7	0.3	0.4	0.3
<i>Propionibacteriaceae</i> (3)		0.2	0.2	0.4	0.4	0.4	0.2	0.2	0.2
<i>Streptomycetaceae</i> (2)		0.3	0.4	0.3	0.3	0.4	0.3	0.2	0.2
Unassigned <i>Actinobacteria</i> (4)		1.0	1.0	1.0	0.9	0.7	0.4	0.5	0.4
<i>Thermoleophilia</i> ,									
<i>Gaiellaceae</i> (2)		0.4	0.4	0.2	0.2	0.2	0.1	0.1	0.1
Unassigned <i>Gaiellaceae</i> (5)		0.8	0.9	0.4	0.4	0.3	0.2	0.2	0.2
<i>Patulibacteraceae</i> (1)		0.1	0.2	0.1	0.1	0.2	0.2	0.1	0.1
<i>Solirubrobacteraceae</i> (2)		0.2	0.2	0.4	0.4	0.3	0.2	0.2	0.1
Unassigned <i>Solirubrobacteraceae</i> (3)		0.4	0.5	0.3	0.2	0.3	0.1	0.1	0.1
<i>Chloroflexi</i> ,									
Unassigned <i>Chloroflexi</i> (3)		0.9	0.8	0.6	0.6	0.5	0.3	0.4	0.3
<i>Firmicutes</i> ,									
<i>Bacilli</i> ,									
<i>Bacillaceae</i> (4)		3.3	1.3	1.5	1.4	1.8	1.1	0.8	0.7
<i>Paenibacillaceae</i> (1)		0.5	0.1	0.1	0.2	0.1	0.1	0.1	0.1
<i>Clostridia</i> ,									
<i>Clostridiaceae</i> (10) [CL8, CL10, CL18, CL15]		8.5	10.6	0.9	1.1	1.1	11.4	8.6	10.9
<i>Lachnospiraceae</i> (5) [CL6]		0.5	1.3	0.1	0.1	0.1	10.8	8.7	6.4
<i>Peptostreptococcaceae</i> (14) [CL2, CL5]		16.5	29.4	7.0	9.9	11.2	20.3	18.7	23.3
<i>Proteobacteria</i> ,									
<i>Alphaproteobacteria</i> ,									
<i>Bradyrhizobiaceae</i> (1)		0.5	0.6	0.9	0.6	0.8	0.3	0.4	0.5
<i>Hypomicrobiaceae</i> (2)		0.1	0.2	0.3	0.3	0.2	0.1	0.2	0.1
<i>Methyllobacteriaceae</i> (2)		0.1	0.1	0.2	0.2	0.2	0.1	0.1	0.1
<i>Phyllobacteriaceae</i> (1)		0.1	0.1	0.2	0.2	0.3	0.1	0.1	0.1
<i>Rhodobiaceae</i> (1)		0.7	0.8	0.7	0.6	0.7	0.3	0.5	0.3
<i>Xanthobacteraceae</i> (4)		1.0	1.1	1.4	1.6	1.1	0.8	0.6	0.5
<i>Acetobacteraceae</i> (1)		0.0	0.0	0.1	0.1	0.1	0.1	0.1	0.0
<i>Rhodobacteraceae</i> (3)		0.1	0.1	0.4	0.4	0.4	0.3	0.3	0.2
<i>Rhodospirillaceae</i> (1)		0.0	0.0	0.1	0.1	0.1	0.0	0.1	0.0
Unassigned <i>Rhodospirillales</i> (6)		0.5	0.4	0.2	1.5	1.4	0.7	0.6	0.7
<i>Deltaproteobacteria</i> ,									
<i>Nitrospinaceae</i> (3)		0.1	0.1	0.5	0.5	0.2	0.1	0.2	0.2
<i>Sorangiineae</i> (2)		0.1	0.1	0.4	0.5	0.3	0.2	0.2	0.1
Unassigned <i>Deltaproteobacteria</i> (2)		0.2	0.1	0.2	0.2	0.1	0.0	0.0	0.0
<i>Gammaproteobacteria</i> ,									
<i>Aeromonadaceae</i> (3) [CL7]		4.8	0.2	0.1	0.2	0.1	1.3	0.9	1.2

	Sampling time: Treatment <sup>b</sup> :	30 h							
		D.C	D.L	C1.R	C2.R	C3.R	L1.R	L2.R	L3.R
<b>Phylum, Class, Family<sup>c</sup></b>		<b>Relative Abundance (%)</b>							
<i>Shewanellaceae</i> (1)		2.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
<i>Enterobacteriaceae</i> (7) [CL4]		1.4	12.3	0.3	0.3	0.2	10.2	9.4	13.1
<b>Planctomycetes</b> ,									
<i>Planctomycetacia</i> ,									
<i>Planctomycetaceae</i> (13)		0.7	0.4	2.9	2.6	2.0	0.8	1.4	1.2
<b>Tenericutes</b> ,									
<i>Mollicutes</i> ,									
<i>Mycoplasmataceae</i> (4)		8.7	2.1	23.8	28.0	29.0	12.8	12.9	11.0
<b>Verrucomicrobia</b> ,									
<i>Spartobacteria</i> ,									
<i>Xiphinematobacteraceae</i> (2)		2.7	1.8	5.9	5.2	4.9	2.4	4.7	4.6
Unassigned <i>Chthoniobacterales</i> (1)		0.9	0.2	0.1	0.1	0.1	0.0	0.0	0.0

<sup>a</sup>Listed are families that had at least one phylotype with  $\geq 1,000$  reads.

<sup>b</sup>Abbreviations: D, 16S rRNA genes; R, 16S rRNA; C, control treatment; L, *S.cerevisiae* lysate treatment. The numbers of each treatment at 30 h (e.g., C1) indicate the respective replicates.

<sup>c</sup>The number of phylotypes with  $\geq 1,000$  reads are shown in parenthesis. Phylotypes that responded to lysate amendment are bold and in brackets (see Figure 5B for detailed relative abundancies of responsive phylotypes).

**Table S11.** Summary of families of the protein and RNA treatments (the most abundant taxa are displayed in Figure 4).<sup>a</sup>

Sampling time:	0 h						10 h			20 h		
	Treatment <sup>b</sup> :	p.D	p.R	C.D	C.R	r.D	r.R	p.R	C.R	r.R	p.R	C.R
Phylum, Class, Family <sup>c</sup>	Relative Abundance (%)											
<b>Actinobacteria,</b>												
Acidimicrobia,												
Acidimicrobiaceae (7)	1.2	0.8	1.2	1.0	1.2	1.0	0.7	1.1	1.0	0.5	1.1	0.7
Unassigned Acidimicrobiales (7)	1.3	0.8	1.3	0.7	1.3	0.8	0.7	1.0	0.8	0.5	0.9	0.7
Actinobacteria,												
Mycobacteriaceae (2)	0.4	0.1	0.4	0.1	0.4	0.1	0.0	0.0	0.0	0.0	0.1	0.0
Unassigned Corynebacteriales (1)	0.1	0.1	0.1	0.1	0.1	0.1	0.0	0.1	0.0	0.0	0.1	0.0
Cellulomonadaceae (1)	0.1	0.3	0.1	0.3	0.1	0.3	0.3	0.4	0.3	0.2	0.4	0.3
Intrasporangiaceae (1)	0.4	0.2	0.4	0.3	0.4	0.2	0.3	0.3	0.3	0.2	0.3	0.3
Microbacteriaceae (5)	1.0	1.3	1.0	1.4	0.9	1.3	0.9	1.3	0.8	0.6	0.9	0.6
Micrococcaceae (1)	0.2	0.1	0.2	0.1	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Micromonosporaceae (2)	0.3	0.2	0.3	0.2	0.3	0.2	0.2	0.3	0.2	0.1	0.2	0.2
Nakamurellaceae (1)	0.1	0.2	0.1	0.2	0.1	0.2	0.2	0.2	0.2	0.1	0.2	0.2
Nocardioidaceae (4)	0.8	0.4	0.8	0.4	0.8	0.4	0.3	0.5	0.4	0.2	0.4	0.3
Propionibacteriaceae (3)	1.0	0.4	0.9	0.4	0.9	0.5	0.3	0.5	0.4	0.3	0.5	0.3
Pseudonocardiaceae (3)	0.6	0.5	0.6	0.5	0.6	0.6	0.6	0.7	0.6	0.3	0.6	0.5
Streptomycetaceae (2)	0.5	0.4	0.5	0.4	0.4	0.3	0.3	0.3	0.3	0.2	0.3	0.3
DA023 group (13)	2.3	1.8	2.2	1.5	2.2	1.5	1.1	1.4	1.2	0.5	0.9	0.9
Unassigned Holophagae (1)	0.1	0.0	0.1	0.0	0.1	0.1	0.0	0.1	0.0	0.0	0.0	0.0
Thermoleophilia,												
Gaiellaceae (1)	0.5	0.1	0.5	0.1	0.5	0.2	0.2	0.2	0.2	0.1	0.2	0.2
Unassigned Gaiellales (8)	2.3	0.5	2.2	0.5	2.2	0.5	0.5	0.7	0.6	0.3	0.6	0.5
Patulibacteraceae (1)	0.1	0.1	0.2	0.1	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Solirubrobacteraceae (4)	0.8	1.0	0.9	1.0	0.9	1.0	0.8	1.2	0.9	0.5	1.1	0.8
Unassigned Solirubrobacterales (6)	1.2	0.3	1.3	0.3	1.3	0.3	0.2	0.3	0.3	0.2	0.3	0.2
Unassigned Actinobacteria (5)	1.6	0.6	1.7	0.6	1.6	0.7	0.6	0.9	0.7	0.4	0.7	0.6
<b>Chloroflexi,</b>												
Caldilineae,												
Caldilineaceae (1)	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Chloroflexia,												
Chloroflexaceae (1)	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.0	0.1	0.1
KD4-96 group (4)	2.6	0.9	2.7	0.9	2.8	0.9	0.7	1.0	0.7	0.5	0.7	0.6
Unassigned Thermomicrobia (2)	0.3	0.1	0.4	0.1	0.4	0.1	0.1	0.1	0.1	0.0	0.1	0.1
Unassigned Chlorofexi (3)	0.5	0.2	0.6	0.2	0.5	0.2	0.1	0.2	0.2	0.1	0.2	0.1
<b>Firmicutes</b>												
Bacilli												
Bacillaceae (6)	0.6	0.6	0.6	0.6	0.6	0.8	0.4	0.6	1.1	0.3	0.5	0.8
Clostridia												
Clostridiaceae (9) [PR7, PR12]	0.2	0.3	0.2	0.2	0.2	0.2	3.4	0.5	0.5	7.8	0.8	0.6

Sampling time:	0 h						10 h			20 h			
	Treatment <sup>b</sup> :	p.D	p.R	C.D	C.R	r.D	r.R	p.R	C.R	r.R	p.R	C.R	r.R
<b>Phylum, Class, Family<sup>c</sup></b>		<b>Relative Abundance (%)</b>											
<i>Lachnospiraceae</i> (1)		0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1
<i>Peptostreptococcaceae</i> (4) [PR2, PR8]		0.2	0.2	0.2	0.1	0.1	0.1	17.3	1.4	0.8	25.8	4.1	0.9
Unassigned <i>Clostridiales</i> (1)		0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.0
<b>Fusobacteria,</b>													
<i>Fusobacteria,</i>													
<i>Fusobacteriaceae</i> (1) [PR6]		0.0	0.0	0.1	0.0	0.0	0.0	0.5	0.1	0.2	4.0	0.8	0.1
<b>Nitrospinae,</b>													
<i>Nitrospina,</i>													
<i>Nitrospiraceae</i> (1)		0.0	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.0	0.0	0.0
Unassigned <i>Nitrospirales</i> (3)		0.5	0.3	0.5	0.3	0.5	0.3	0.2	0.3	0.3	0.2	0.3	0.4
<b>Planctomycetes</b>													
<i>Planctomycetacia,</i>													
<i>Planctomycetaceae</i> (64)		5.9	8.8	6.1	9.0	6.8	9.1	7.0	9.3	9.5	5.3	10.2	10.3
Unassigned <i>Phycisphaerae</i> (7)		1.2	1.0	1.0	0.8	1.3	0.9	0.6	0.7	1.0	0.3	0.7	0.8
Unassigned <i>Planctomycetales</i> (1)		0.1	0.1	0.1	0.1	0.1	0.1	0.0	0.0	0.1	0.0	0.1	0.0
<b>Proteobacteria,</b>													
<i>Alphaproteobacteria,</i>													
<i>Bradyrhizobiaceae</i> (1)		0.7	1.2	0.6	1.1	0.6	1.2	1.0	1.3	1.0	0.7	1.2	0.7
<i>Hyphomicrobiaceae</i> (2)		0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.1	0.2	0.1
<i>Methylbacteriaceae</i> (2)		0.2	0.2	0.2	0.2	0.2	0.3	0.2	0.3	0.2	0.2	0.3	0.2
<i>Phyllobacteriaceae</i> (1)		0.2	0.3	0.2	0.3	0.2	0.3	0.3	0.4	0.3	0.2	0.3	0.2
<i>Rhodobiaceae</i> (1)		1.1	0.4	1.0	0.4	1.0	0.4	0.3	0.4	0.3	0.2	0.4	0.2
<i>Xanthobacteraceae</i> (7)		2.4	1.4	2.4	1.2	2.2	1.4	1.1	1.5	1.2	0.8	1.5	0.9
Unassigned <i>Rhizobiales</i> (1)		0.1	0.1	0.0	0.1	0.1	0.1	0.1	0.1	0.1	0.0	0.1	0.0
<i>Rhodobacteraceae</i> (1)		0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
<i>Rhodospirillaceae</i> (7)		0.6	1.3	0.6	1.2	0.6	1.3	1.3	1.7	1.4	0.8	1.5	1.2
Unassigned <i>Rhodospirillales</i> (4)		0.4	0.6	0.5	0.5	0.5	0.5	0.6	0.6	0.6	0.3	0.6	0.5
<i>Betaproteobacteria,</i>													
<i>Alcaligenaceae</i> (1)		0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
<i>Comamonadaceae</i> (1)		0.0	0.1	0.0	0.1	0.0	0.1	0.1	0.1	0.1	0.1	0.1	0.1
<i>Deltaproteobacteria,</i>													
<i>Nitrospinaceae</i> (2)		0.1	0.7	0.1	0.6	0.1	0.6	0.6	0.6	0.6	0.3	0.5	0.5
<i>Cystobacteraceae</i> (1)		0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.2	0.0	0.1	0.1
<i>Nannocystineae</i> (1)		0.0	0.1	0.0	0.1	0.0	0.1	0.0	0.1	0.0	0.0	0.1	0.1
<i>Sorangiiineae</i> (1)		0.0	0.1	0.0	0.1	0.0	0.1	0.1	0.1	0.1	0.0	0.1	0.1
Unassigned <i>Deltaproteobacteria</i> (2)		0.5	0.4	0.5	0.4	0.5	0.4	0.3	0.5	0.4	0.3	0.4	0.3
<i>Gammaproteobacteria,</i>													
<i>Aeromonadaceae</i> (4) [PR3]		0.4	0.1	0.3	0.1	0.3	0.8	1.3	0.3	12.0	0.7	0.3	25.0
<i>Shewanellaceae</i> (1)		0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
<i>Enterobacteriaceae</i> (3) [PR33]		0.1	0.3	0.1	0.1	0.1	0.2	0.6	0.1	0.4	0.7	0.2	0.7
<i>Sinobacteraceae</i> (1)		0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.0	0.1	0.0

	Sampling time:		0 h						10 h			20 h		
	Treatment <sup>b</sup> :		p.D	p.R	C.D	C.R	r.D	r.R	p.R	C.R	r.R	p.R	C.R	r.R
Phylum, Class, Family <sup>c</sup>	Relative Abundance (%)													
<i>Tenericutes</i> ,														
<i>Mollicutes</i> ,														
<i>Mycoplasmataceae</i> (3)		13.2	28.8	13.0	32.3	11.0	28.1		20.3	25.6	20.6	20.0	28.1	15.3
Unassigned <i>Mollicutes</i> (2)		0.2	0.3	0.2	0.3	0.2	0.3		0.2	0.2	0.2	0.2	0.2	0.1
<i>Verrucomicrobia</i> ,														
<i>Spartobacteria</i> ,														
<i>Chthoniobacteraceae</i> (1)		0.1	0.2	0.2	0.2	0.2	0.2		0.1	0.1	0.1	0.1	0.1	0.0
<i>Xiphinematobacteraceae</i> (2)		4.5	5.0	4.9	4.4	5.5	5.3		5.0	5.6	4.2	3.7	4.6	2.8
Unassigned <i>Chthoniobacterales</i> (8)		3.7	0.7	4.3	0.8	4.6	0.8		0.5	0.6	0.5	0.3	0.5	0.4
<i>Verrucomicrobiae</i> ,														
<i>Verrucomicrobiaceae</i> (1)		0.2	0.1	0.1	0.1	0.2	0.1		0.0	0.1	0.0	0.0	0.0	0.0
Sampling time:		30 h												
Treatment <sup>b</sup> :		p.D	p1.R	p2.R	p3.R	C.D	C1.R	C2.R	C3.R	r.D	r1.R	r2.R	r3.R	
Phylum, Class, Family <sup>c</sup>	Relative Abundance (%) <sup>b</sup>													
<i>Actinobacteria</i> ,														
<i>Acidimicrobia</i> ,														
<i>Acidimicrobiaceae</i> (7)		0.6	0.6	0.6	0.6	1.2	1.0	1.0	0.8	0.6	0.8	0.7	1.1	
Unassigned <i>Acidimicrobiales</i> (7)		0.5	0.4	0.4	0.5	1.3	0.9	0.8	0.7	0.5	0.7	0.6	0.8	
<i>Actinobacteria</i> ,														
<i>Mycobacteriaceae</i> (2)		0.3	0.0	0.0	0.0	0.3	0.1	0.1	0.1	0.2	0.1	0.0	0.0	
Unassigned <i>Corynebacteriales</i> (1)		0.0	0.0	0.0	0.0	0.1	0.0	0.1	0.0	0.0	0.0	0.0	0.0	
<i>Cellulomonadaceae</i> (1)		0.1	0.2	0.2	0.2	0.3	0.4	0.4	0.3	0.1	0.3	0.3	0.5	
<i>Intrasporangiaceae</i> (1)		0.2	0.1	0.2	0.2	0.4	0.3	0.3	0.2	0.2	0.2	0.2	0.2	
<i>Microbacteriaceae</i> (5)		0.4	0.5	0.6	0.6	0.9	1.1	0.8	0.8	0.4	1.0	0.7	1.2	
<i>Micrococcaceae</i> (1)		0.1	0.1	0.0	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	
<i>Micromonosporaceae</i> (2)		0.1	0.1	0.1	0.2	0.3	0.2	0.3	0.2	0.1	0.2	0.2	0.2	
<i>Nakamurellaceae</i> (1)		0.1	0.1	0.1	0.1	0.1	0.1	0.2	0.2	0.0	0.2	0.1	0.2	
<i>Nocardioidaceae</i> (4)		0.3	0.2	0.2	0.2	0.7	0.3	0.5	0.3	0.3	0.3	0.3	0.4	
<i>Propionibacteriaceae</i> (3)		0.4	0.3	0.3	0.3	1.0	0.4	0.5	0.4	0.4	0.4	0.3	0.5	
<i>Pseudonocardiaceae</i> (3)		0.2	0.3	0.4	0.3	0.5	0.5	0.6	0.5	0.3	0.5	0.5	0.7	
<i>Streptomycetaceae</i> (2)		0.2	0.1	0.2	0.2	0.5	0.3	0.4	0.3	0.2	0.3	0.3	0.4	
DA023 group (13)		0.8	0.2	0.4	0.4	1.8	0.7	0.8	0.4	0.6	0.6	0.7	0.6	
Unassigned <i>Holophagae</i> (1)		0.1	0.0	0.0	0.0	0.2	0.1	0.0	0.0	0.1	0.0	0.0	0.1	
<i>Thermoleophilia</i> ,														
<i>Gaiellaceae</i> (1)		0.2	0.1	0.1	0.1	0.5	0.2	0.2	0.2	0.2	0.2	0.1	0.2	
Unassigned <i>Gaiellales</i> (8)		1.0	0.3	0.3	0.4	2.4	0.7	0.6	0.6	1.0	0.6	0.5	0.7	
<i>Patulibacteraceae</i> (1)		0.1	0.0	0.0	0.1	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.1	
<i>Solirubrobacteraceae</i> (4)		0.3	0.4	0.5	0.5	0.8	1.0	1.0	0.8	0.3	0.8	0.7	1.1	

	Sampling time:		30 h										
	Treatment <sup>b</sup> :		p.D	p1.R	p2.R	p3.R	C.D	C1.R	C2.R	C3.R	r.D	r1.R	r2.R
<b>Phylum, Class, Family<sup>c</sup></b>		<b>Relative Abundance (%)<sup>b</sup></b>											
Unassigned <i>Solirubrobacterales</i> (6)		0.5	0.2	0.2	0.2	1.1	0.3	0.3	0.3	0.5	0.2	0.2	0.3
Unassigned <i>Actinobacteria</i> (5)		0.7	0.4	0.4	0.4	1.6	0.7	0.8	0.7	0.7	0.6	0.5	0.8
<i>Chloroflexi</i> , <i>Caldilineae</i> ,		0.0	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.0	0.1	0.1	0.1
<i>Caldilineaceae</i> (1)		0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.1	0.1	0.0	0.0
<i>Chloroflexia</i> ,		1.1	0.4	0.4	0.5	2.9	0.9	0.7	0.6	1.1	0.7	0.7	0.9
<i>Chloroflexaceae</i> (1)		0.2	0.0	0.1	0.1	0.4	0.1	0.1	0.1	0.2	0.1	0.1	0.1
KD4-96 group (4)		0.2	0.1	0.1	0.1	0.6	0.2	0.2	0.1	0.2	0.1	0.2	0.2
Unassigned <i>Thermomicrobia</i> (2)		0.2	0.0	0.1	0.1	0.4	0.1	0.1	0.1	0.2	0.1	0.1	0.1
Unassigned <i>Chloroflexi</i> (3)		0.2	0.1	0.1	0.1	0.6	0.2	0.2	0.1	0.2	0.1	0.2	0.2
<b>Firmicutes</b>													
<i>Bacilli</i>													
<i>Bacillaceae</i> (6)		1.7	0.4	0.3	0.4	1.0	0.6	0.7	0.7	1.1	1.2	1.2	1.8
<i>Clostridia</i>		13.8	11.1	13.3	11.9	1.5	1.4	1.1	1.3	0.8	1.8	1.7	3.0
<i>Clostridiaceae</i> (9) [PR7, PR12]		0.3	0.3	0.2	0.2	0.2	0.1	0.1	0.1	0.7	1.1	1.1	2.2
<i>Lachnospiraceae</i> (1)		18.2	18.4	20.6	23.1	3.4	4.1	3.6	3.5	1.3	2.2	2.4	4.5
<i>Peptostreptococcaceae</i> (4) [PR2, PR8]		0.2	0.5	0.4	0.6	0.1	0.3	0.4	0.4	0.0	0.0	0.0	0.0
Unassigned <i>Clostridiales</i> (1)		11.5	10.5	7.9	6.3	1.4	0.6	1.6	0.5	0.6	0.2	0.2	0.2
<i>Fusobacteria</i> , <i>Fusobacteriia</i> ,		0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.1	0.1	0.1
<i>Fusobacteriaceae</i> (1) [PR6]		0.2	0.1	0.1	0.2	0.6	0.2	0.3	0.2	0.2	0.3	0.2	0.3
<i>Nitrospinae</i> , <i>Nitrospira</i> ,		3.2	5.7	5.6	6.1	6.6	9.2	9.5	6.4	3.4	8.1	8.3	10.6
<i>Nitrospiraceae</i> (1)		0.6	0.3	0.3	0.3	1.0	0.5	0.5	0.3	0.6	0.5	0.5	0.5
Unassigned <i>Nitrospirales</i> (3)		0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.0	0.0	0.0	0.0
<b>Planctomycetes</b>													
<i>Planctomycetacia</i> ,		0.4	0.3	0.3	0.3	1.0	0.5	0.5	0.3	0.6	0.5	0.5	0.5
<i>Planctomycetaceae</i> (64)		0.1	0.1	0.1	0.1	0.2	0.2	0.2	0.2	0.1	0.1	0.1	0.2
Unassigned <i>Phycisphaerae</i> (7)		0.1	0.1	0.1	0.1	0.2	0.2	0.3	0.2	0.1	0.2	0.2	0.2
Unassigned <i>Planctomycetales</i> (1)		0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.0	0.0	0.0	0.0
<b>Proteobacteria</b> ,													
<i>Alphaproteobacteria</i> ,		0.1	0.1	0.1	0.1	0.1	0.2	0.2	0.2	0.1	0.1	0.1	0.2
<i>Bradyrhizobiaceae</i> (1)		0.1	0.1	0.1	0.1	0.1	0.2	0.2	0.2	0.1	0.1	0.1	0.2
<i>Hyphomicrobiaceae</i> (2)		0.1	0.1	0.1	0.1	0.2	0.2	0.3	0.2	0.1	0.2	0.2	0.2
<i>Methylobacteriaceae</i> (2)		0.1	0.1	0.1	0.1	0.2	0.2	0.3	0.2	0.1	0.2	0.2	0.2
<i>Phyllobacteriaceae</i> (1)		0.1	0.2	0.2	0.2	0.2	0.3	0.3	0.2	0.1	0.2	0.2	0.3
<i>Rhodobiaceae</i> (1)		0.4	0.3	0.2	0.2	0.9	0.5	0.5	0.3	0.3	0.2	0.2	0.4
<i>Xanthobacteraceae</i> (7)		0.9	0.7	0.7	0.8	2.1	1.5	1.6	1.2	0.8	0.9	0.9	1.2
Unassigned <i>Rhizobiales</i> (1)		0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.1	0.0	0.1	0.1	0.1
<i>Rhodobacteraceae</i> (1)		0.0	0.1	0.1	0.0	0.1	0.1	0.1	0.1	0.0	0.1	0.1	0.1
<i>Rhodospirillaceae</i> (7)		0.3	0.8	0.8	0.8	0.6	1.2	1.4	1.1	0.3	1.1	1.0	1.6

	Sampling time:		30 h										
	Treatment <sup>b</sup> :		p.D	p1.R	p2.R	p3.R	C.D	C1.R	C2.R	C3.R	r.D	r1.R	r2.R
<b>Phylum, Class, Family<sup>c</sup></b>		<b>Relative Abundance (%)<sup>b</sup></b>											
Unassigned <i>Rhodospirillales</i> (4)		0.2	0.3	0.4	0.3	0.5	0.5	0.6	0.4	0.2	0.5	0.4	0.5
<i>Betaproteobacteria</i> ,													
<i>Alcaligenaceae</i> (1)		0.1	0.1	0.1	0.0	0.2	0.1	0.1	0.1	0.1	0.1	0.0	0.1
<i>Comamonadaceae</i> (1)		0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.0	0.1	0.1	0.1
<i>Deltaproteobacteria</i> ,													
<i>Nitrospinaceae</i> (2)		0.1	0.2	0.3	0.2	0.1	0.3	0.5	0.3	0.1	0.4	0.5	0.5
<i>Cystobacteraceae</i> (1)		0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.0	0.1	0.1	0.1
<i>Nannocystineae</i> (1)		0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.0	0.0	0.0	0.1
<i>Sorangiineae</i> (1)		0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.1	0.0	0.1
Unassigned <i>Deltaproteobacteria</i> (2)		0.2	0.2	0.2	0.2	0.4	0.3	0.3	0.3	0.2	0.3	0.3	0.4
<i>Gammaproteobacteria</i> ,													
<i>Aeromonadaceae</i> (4) [PR3]		4.4	0.3	0.4	0.3	2.2	0.1	0.3	0.1	44.5	17.9	20.7	22.2
<i>Shewanellaceae</i> (1)		0.7	0.0	0.0	0.0	1.1	0.0	0.0	0.0	0.5	0.2	0.2	0.2
<i>Enterobacteriaceae</i> (3) [PR33]		0.8	0.4	0.4	0.3	0.2	0.1	0.1	0.1	4.0	2.3	2.6	3.4
<i>Sinobacteraceae</i> (1)		0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.0	0.0	0.0	0.0
<i>Tenericutes</i> ,													
<i>Mollicutes</i> ,													
<i>Mycoplasmataceae</i> (3)		6.8	18.0	15.7	16.2	7.8	32.3	28.4	19.3	6.5	21.6	17.7	29.5
Unassigned <i>Mollicutes</i> (2)		0.1	0.2	0.2	0.2	0.1	0.3	0.3	0.2	0.1	0.2	0.2	0.3
<i>Verrucomicrobia</i> ,													
<i>Spartobacteria</i> ,													
<i>Chthoniobacteraceae</i> (1)		0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.1	0.0	0.0	0.1
<i>Xiphinematobacteraceae</i> (2)		2.8	4.6	4.5	3.7	4.3	4.4	5.2	3.9	3.0	3.0	5.1	6.4
Unassigned <i>Chthoniobacterales</i> (8)		2.1	0.3	0.3	0.2	3.4	0.6	0.6	0.3	1.8	0.4	0.3	0.5
<i>Verrucomicrobiae</i> ,													
<i>Verrucomicrobiaceae</i> (1)		0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0

<sup>a</sup>Listed are families that had at least one phylotype with  $\geq 1,000$  reads.

<sup>b</sup>Abbreviations: D, 16S rRNA genes; R, 16S rRNA; C, control treatment; r, RNA treatment; p, protein treatment. The numbers of each treatment at 30 h (e.g., C1) indicate the respective replicates.

<sup>c</sup>The number of phylotypes with  $\geq 1,000$  reads are shown in parenthesis. Phylotypes that responded to protein or RNA amendment are bold and in brackets (see Figure 5B for detailed relative abundancies of responsive phylotypes).