

1 **Supplementary Material**

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3 **Table S1.** Percent of total organic compounds peak area detected in 500 °C and 700 °C

4 APL by GC-MS. Compounds sorted in increasing order of the molar mass.

Compounds	Molar mass (g/mol)	% of total organic compound peak area detected	
		Non-ozonated 500 °C APL	Non-ozonated 700 °C APL
Acetonitrile	41.05	2.4	14.74
Propanenitrile	55.08	0.7	2.59
Acetamide	59.07	17.0	19.06
Acetic acid	60.06	15.1	-
Pyrrrole	67.09	0.5	3.49
1H-Imidazole	68.08	-	1.25
2-Propenamide	71.08	-	0.77
Propanamide	73.09	3.2	2.53
Acetamide, N-methyl-	73.09	1.6	1.32
Propionic acid	74.08	1.7	-
1-Butanol	74.12	1.3	0.8
Pyridine	79.10	1.2	3.93
1,3-Diazine	80.09	-	0.57
1H-Pyrazole, 3-methyl-	82.10	0.6	-
1H-Imidazole, 4-methyl-	82.10	-	1.9
trans-Crotonamide	85.10	-	0.71
2-Pyrrolidinone	85.11	3.0	2.52
Acetamide, N,N-dimethyl-	87.12	0.8	0.56
Propanoic acid, 2-methyl-	88.11	1.0	-
Glycerin	92.09	1.3	-
Pyridine, 2-methyl-	93.13	-	1.65
Pyridine, 3-methyl-	93.13	-	0.86
2-Aminopyridine	94.11	0.5	-
3-Aminopyridine	94.11	2.0	4.92
Phenol	94.11	-	2.08
Pyrazine, methyl-	94.11	-	1.65
2-Aminopyridine	94.11	-	0.75
Pyrimidine, 2-methyl-	94.11	-	0.72
2(1H)-Pyridinone	95.10	0.5	1.42
3-Pyridinol	95.10	3.7	4
1H-Imidazole, 2,4-dimethyl-	96.13	-	1.44
3-Dimethylaminoacrylonitrile	96.13	-	0.57
2-Furanmethanol	98.10	-	1.33
2,5-Pyrrolidinedione	99.09	1.6	1.66
2-Piperidinone	99.13	1.2	-
Butanamide, 3-methyl-	101.2	1.0	0.59
Phenol, 4-methyl-	108.1	0.8	1.31
2-Pyridinamine, 5-methyl-	108.1	-	1.03
Pyrimidine, 4,6-dimethyl-	108.1	-	0.95
4-Pyridinemethanol	109.1	3.5	-
3-Pyridinol, 2-methyl-	109.1	0.6	1.03
3-Pyridinol, 6-methyl-	109.1	-	2.91
2-Amino-4-methylpyrimidine	109.1	-	0.44
2(1H)-Pyridinone, 3-methyl-	109.1	-	0.6
Pyrrrole-2-carboxamide	110.1	0.8	-
2(1H)Pyrimidinone, 1-methyl-	110.1	0.5	-
3,4-Dimethyl-3-pyrrolin-2-one	111.1	0.5	-
2,5-Pyrrolidinedione, 1-methyl-	113.1	0.5	1
2,4-Imidazolidinedione, 5-methyl-	114.1	0.6	-
Pentanoic acid, 3-methyl-	116.2	2.1	-

Pentanamide, 5-hydroxy-	117.2	0.6	-
Indolizine	117.2	-	0.57
Pyrazine, 2-ethyl-3-methyl-	122.2	-	0.69
2(1H)-Pyridinone, 3,6-dimethyl-	123.2	-	0.79
1-Propanone, 1-(2-furanyl)-	124.1	0.5	-
5,6-Dihydro-6-methyluracil	128.1	1.1	1.46
5,5-dimethyl-2,4-imidazolidinedione	128.1	6.4	1.78
Oxazolidine, 2-ethyl-2,3-dimethyl-	129.2	0.7	-
DL-Alanine, N-acetyl-	131.1	0.7	0.55
Quinoline, 5,6,7,8-tetrahydro-	133.2	-	0.89
Acetamide, N-3-pyridinyl-	136.2	1.7	0.76
dl-5-Ethyl-5-methyl-2,4-	142.2	2.9	0.87
5-Isopropyl-2,4-imidazolidinedione	142.2	2.4	-
6-Amino-1-methylpurine	150.2	1.0	0.6
Acetamide, N-(4-aminophenyl)-	150.2	0.6	-
Formamide, N,N-dibutyl-	157.3	1.9	-
Uric acid	168.1	1.6	-
Phosphonic acid, (p-hydroxyphenyl)-	174.1	1.0	-
D-Allose	180.2	0.9	-
Pyrrolo[1,2-a]pyrazine-1,4-dione, hexahydro-3-(2-methylpropyl)-	210.3	1.0	-
5,10-Diethoxy-2,3,7,8-tetrahydro-1H,6H-dipyrrolo[1,2-a;1',2'-d]pyrazine	250.3	1.2	-
Piperazine, 1-[(2,4-dichlorobenzoyl)methyl]-4-methyl-	287.2	-	0.49
Pyrimidine-2,4,6-trione, 1-cyclohexyl-5-[(2-piperazin-1-yl-ethylamino)methylene]-	349.4	-	0.48
Deoxyspergualin	387.5	-	0.42

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8 **Table S2.** Average reads and average number of microbial zOTUs observed per digester

	MB1	MB2	IB1	IB2
Total reads	332,218	314,665	355,290	367,660
Average ± Standard deviation read per digester	27,685 ± 677	26,222 ± 813	29,607 ± 683	30,638 ± 855
Observed zOTU	899 ± 51	928 ± 69	525 ± 56	355 ± 21

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- 11 **Table S3.** Average and standard deviation (Avg ± std) of observed OTU, Shannon Index
 12 (H) and Chao1 value per triplicate digesters

Sample	MB1		MB2		IB1		IB2	
	Shannon	Chao1	Shannon	Chao1	Shannon	Chao1	Shannon	Chao1
Inocula	1062	4.7	1067	4.4	528	4.3	423	3.6
Control	1028	4.4	1069	4.6	553	3.4	386	3.3
Digester fed non-ozonated APL	1156	4.5	1156	5.0	697	3.7	432	3.3
Digester fed ozonated APL	1176	4.5	1196	5.1	736	3.6	428	3.4

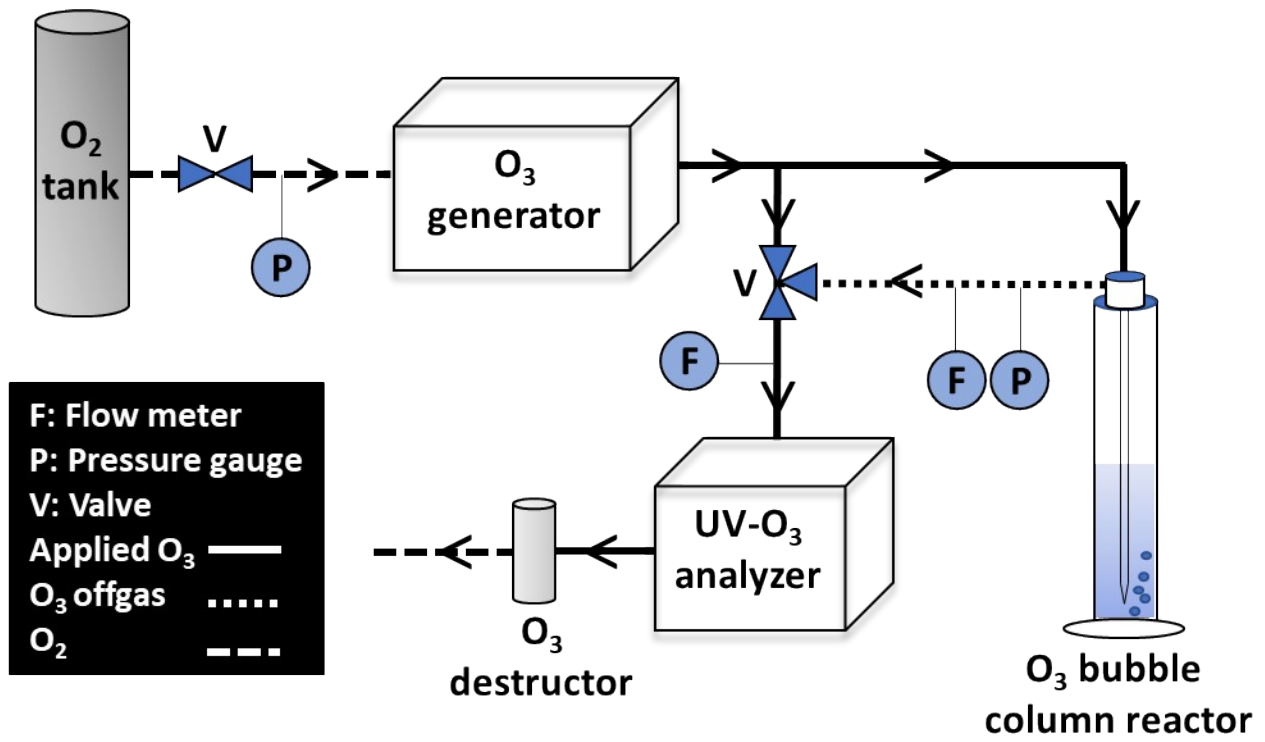
14 **Table S4.** Functions carried out by the identified bacterial taxa described in Section
 15 3.6.2

OTU #	OTU Name	Taxonomic Level	Function	Reference
7	Sphingobacteriaceae	Family	Involved in hydrolysis of phenolics and polycyclic and monocyclic aromatic hydrocarbons	(1)
38	Anaerolineaceae	Family	Utilizes carbohydrates and proteinaceous carbon sources and produces hydrogen as a fermentation byproducts	(2)
5	Clostridicaea	Family	Involved in syntrophic acetate oxidation (SAO) along with hydrogenotrophic methanogens and contributes to methane formation in high-ammonia processes	(3,4)
15	<i>Synergistes</i>	Genus	Involved in syntrophic acetate oxidation (SAO) along with hydrogenotrophic methanogens and contributes to methane formation in high-ammonia processes	(4,5)
14	<i>Cytophaga</i>	Genus	Degrades biopolymers such as xylan and cellulose	(6)
3 and 24	<i>Bellilinea</i>	Genus	Found in methanogenic propionate-degrading consortia	(7)
41	<i>Symbiobacterium</i>	Genus	Moderately anaerobic, thermophilic and chemo-organotrophic bacteria involved in SAO	(8)
59	<i>Acetovibrio</i>	Genus	Acetogenic bacteria degrading cellulose and cellobiose	(9,10)
105	<i>Sedimentibacter</i>	Genus	Strictly anaerobic bacteria producing acetate and butyrate by utilizing complex substrates including alpha-keto acids and amino acids while carbohydrates are not utilized	(11)

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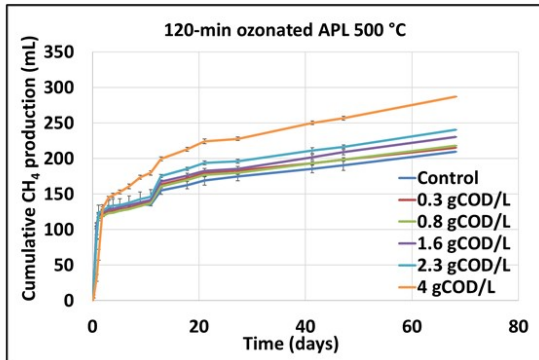
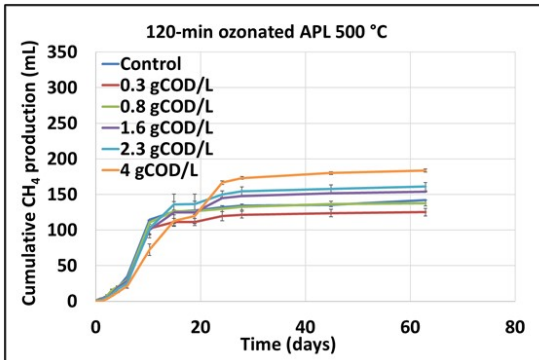
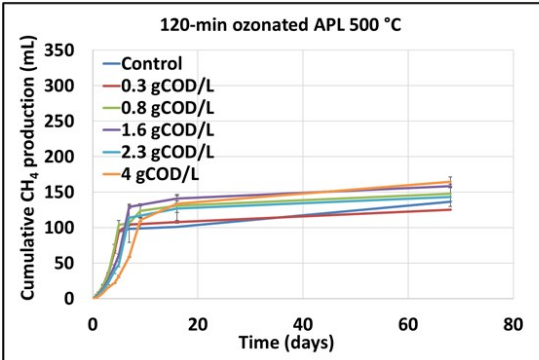
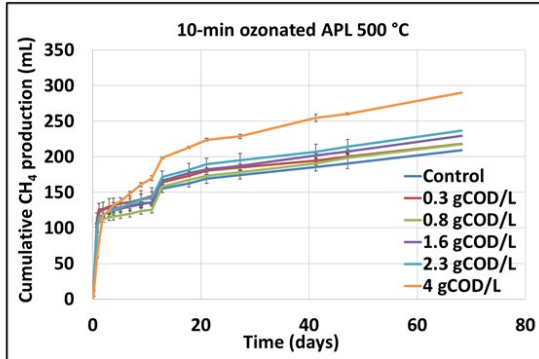
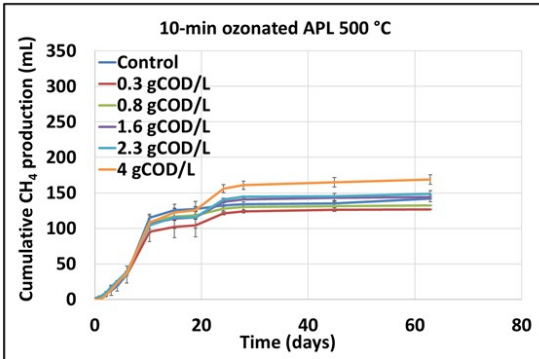
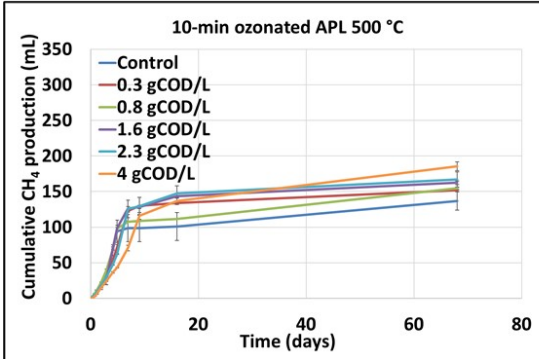
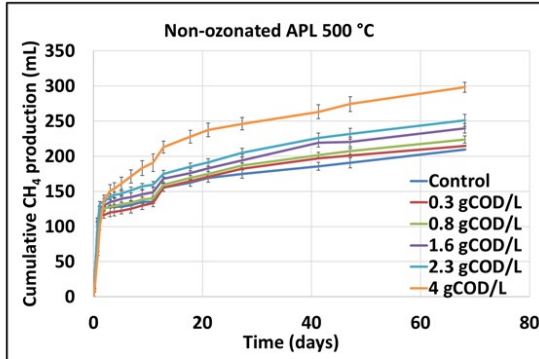
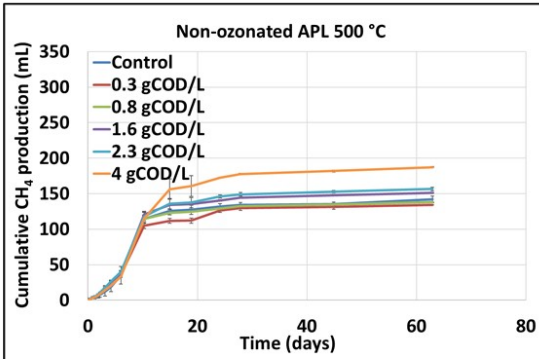
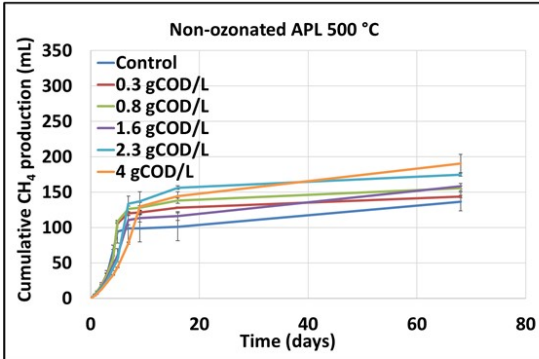
20 **Figure S1.** Pre-ozonation Schematic.

A

Municipal biomass no. 1

Municipal biomass no. 2

Industrial biomass no. 1



B

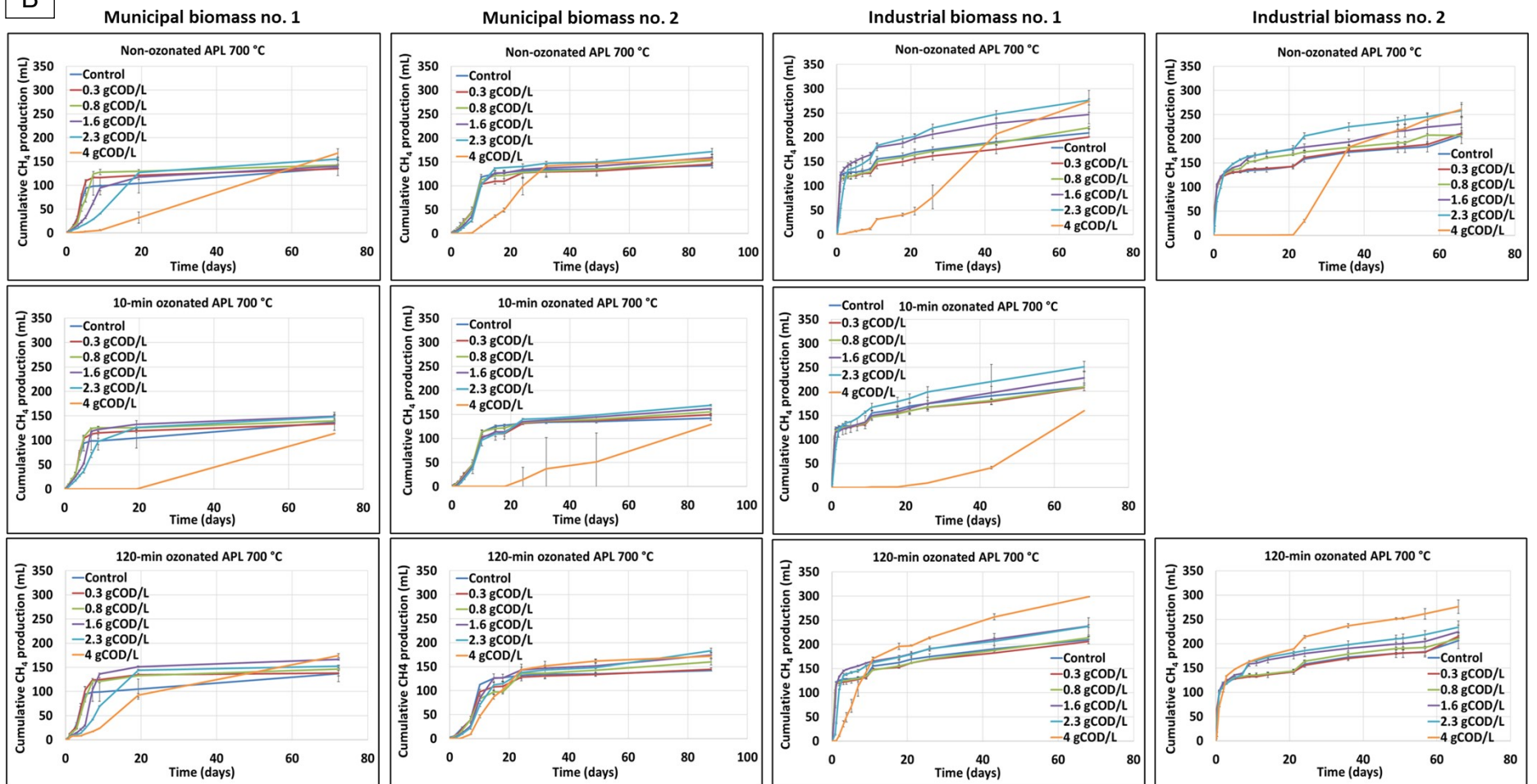


Figure S2. Cumulative methane production from ATA on non-ozonated, 10-min ozonated and 2-h ozonated APL using different inocula at different APL COD concentrations. Average values and standard deviations are from triplicates bottles. (A) 500 °C APL, (B) 700 °C APL.

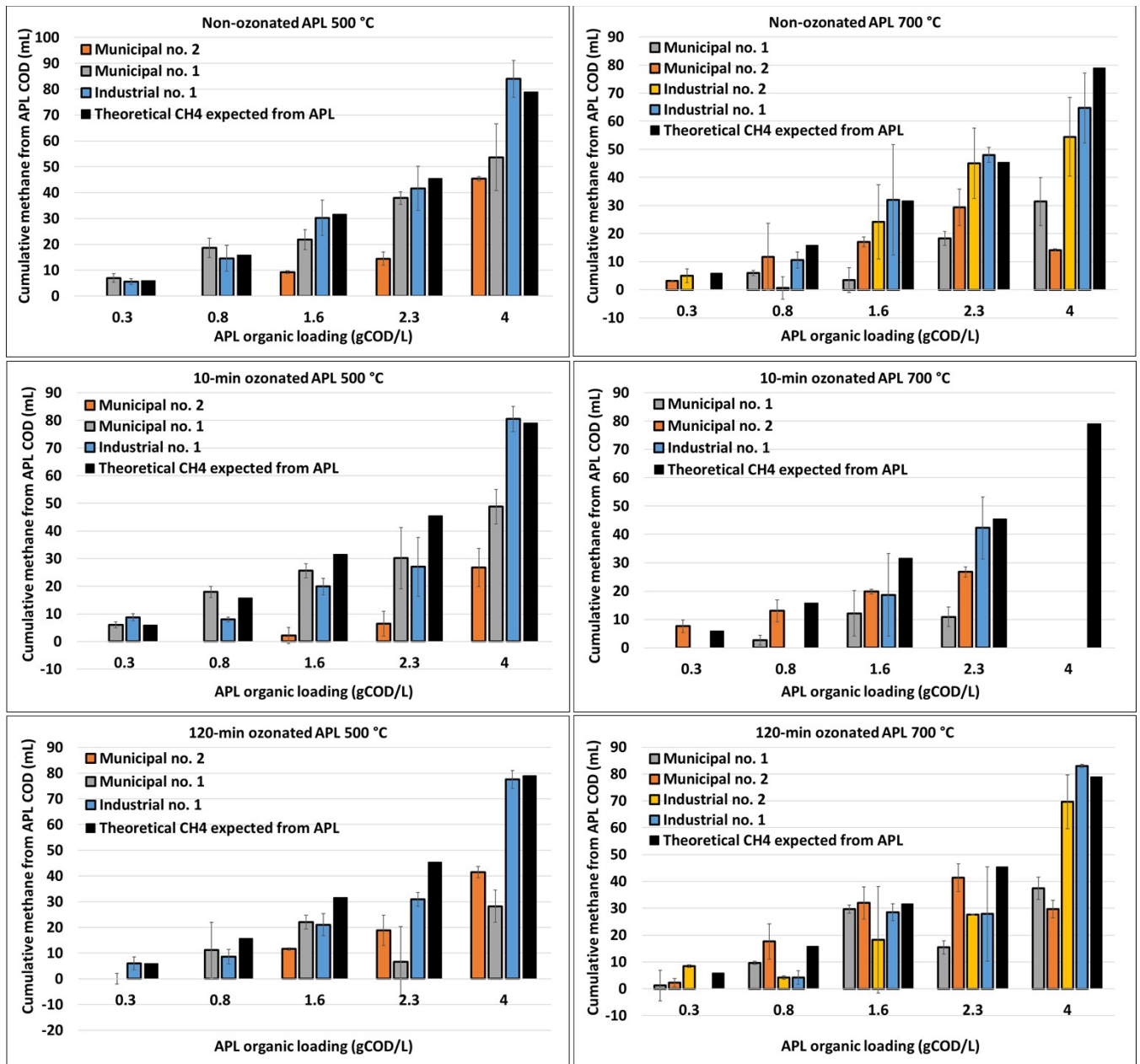


Figure S3. Final cumulative methane production from ATAs on non-ozonated, 10-min ozonated and 2-h ozonated APL produced at 500 °C and 700 °C at different concentrations employing different inocula. The black bars show the theoretical stoichiometric methane production (395 mL CH₄ produced per gCOD removed at 35 °C and 1 atm) for comparison to the actual methane produced from APL COD at each concentration. Error bars represent standard deviation from triplicate bottles. Some error bars are small and not visible.

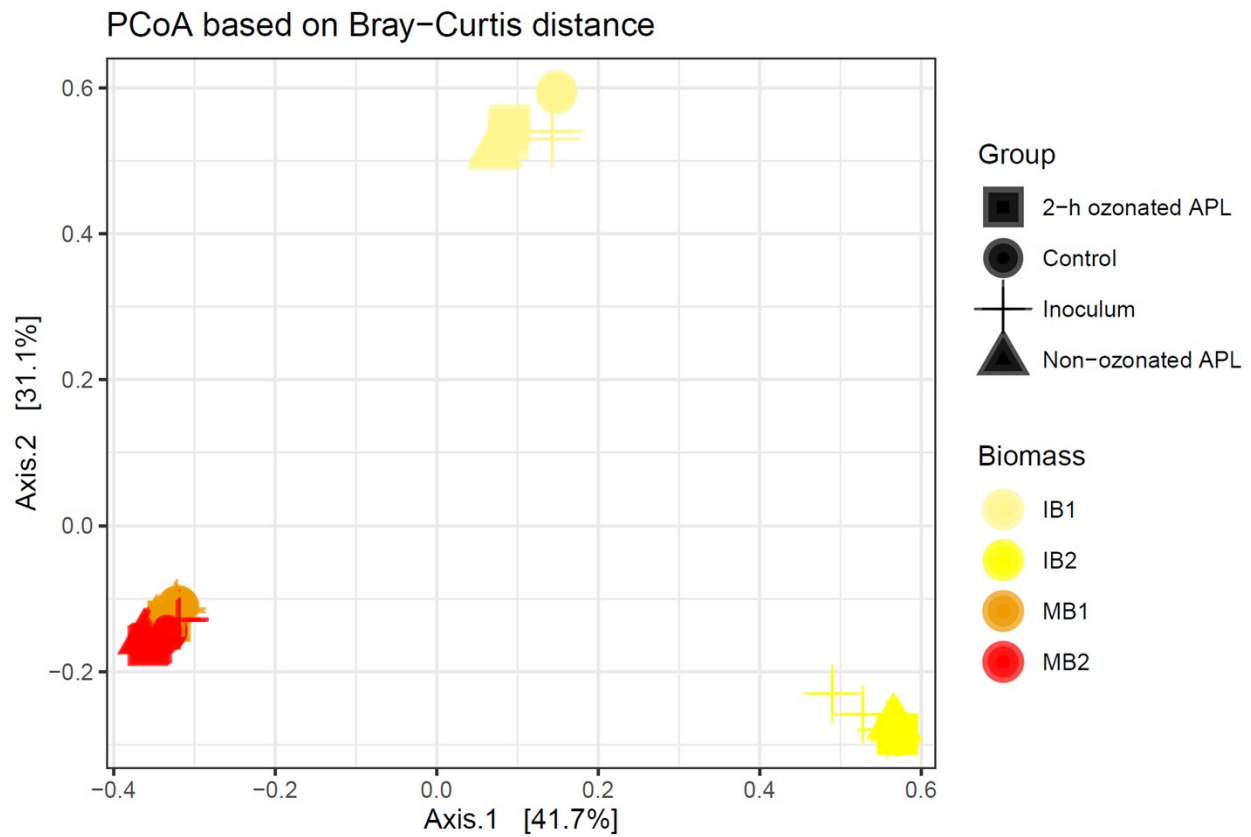


Figure S4. PCoA analysis of all four MB1, MB2, IB1 and IB2 biomass. MB1 and MB2 communities clustered closer, exhibiting some similarities whereas IB1 and IB2 communities are clustered further apart demonstrating different communities.

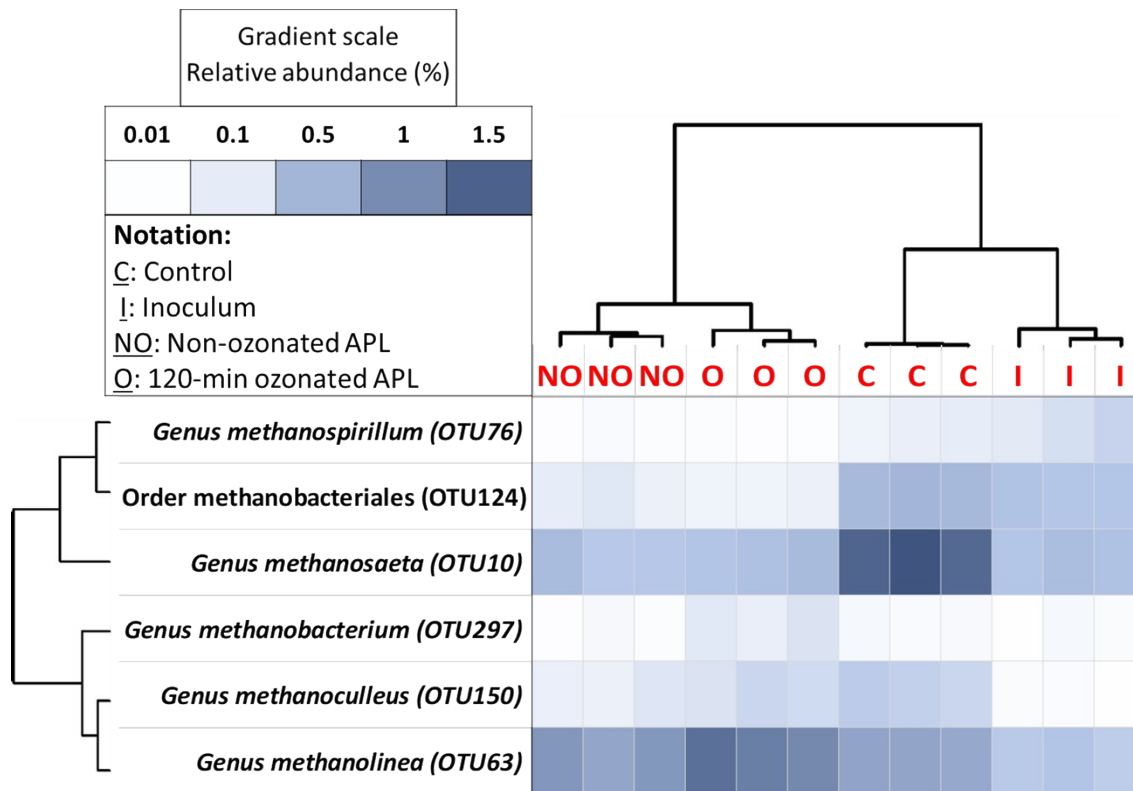


Figure S5. Dual hierarchical clustering of the 6 most abundant archaeal OTUs in MB2. These OTUs represent $86 \pm 5.2\%$ of total archaeal community relative abundance in all MB2-inoculated bottles.

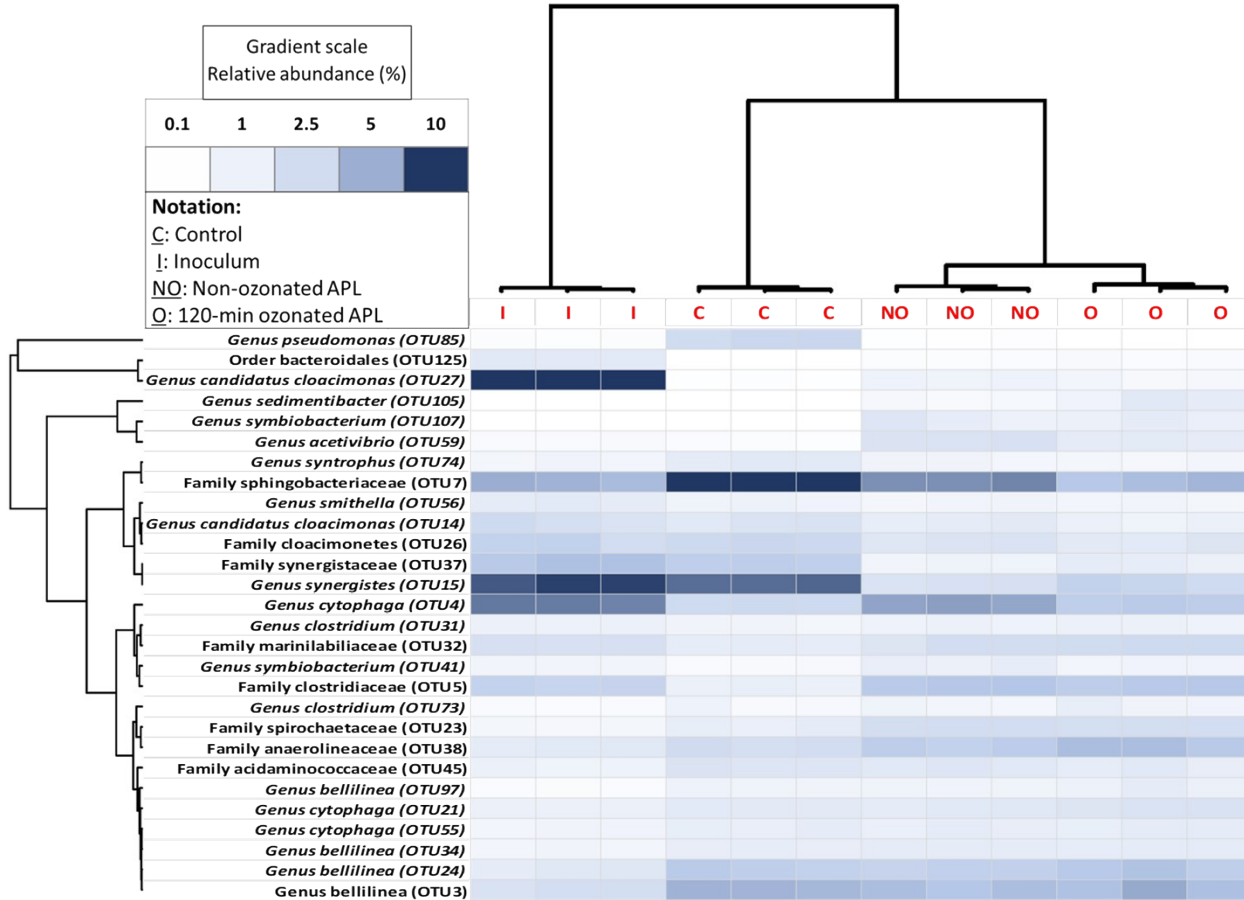


Figure S6. Dual hierarchical clustering of the 28 most abundant bacterial OTUs in MB2. These OTUs represent $58 \pm 5\%$ of the total MB2 bacterial community abundance in all MB2-inoculated bottles.

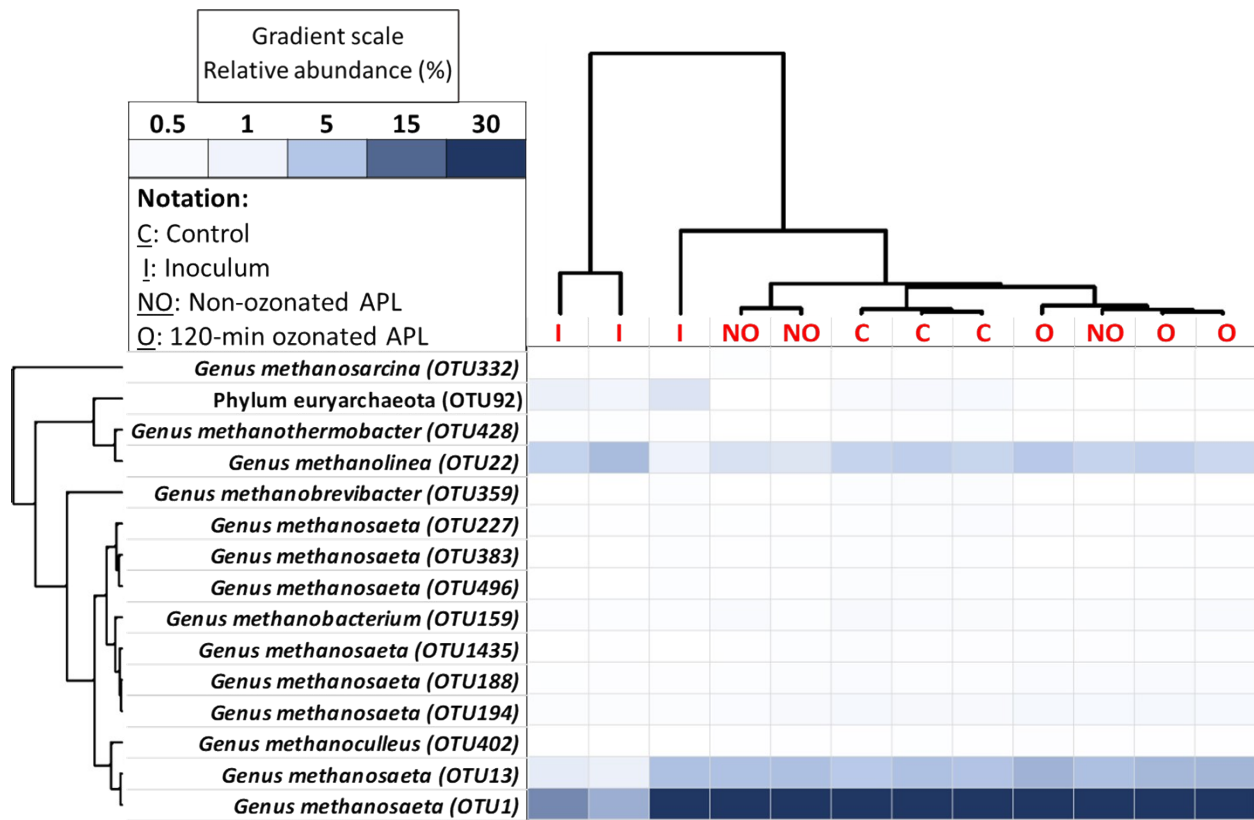


Figure S7. Dual hierarchical clustering of the 15 most abundant archaeal OTUs in IB2. These OTUs represent $99 \pm 0.3\%$ of total archaeal community relative abundance in all IB2-inoculated bottles.

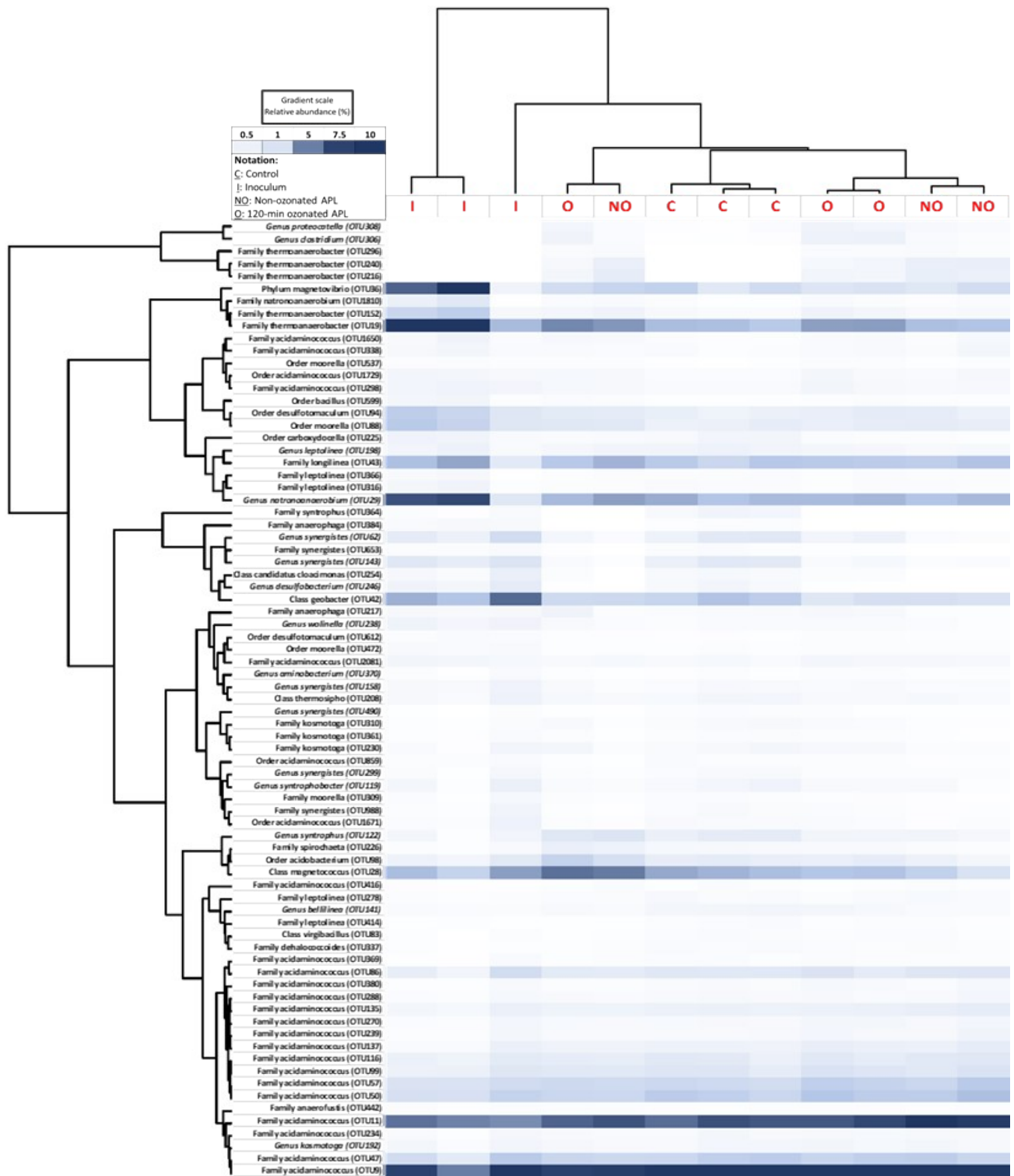


Figure S8. Dual hierarchical clustering of the 77 most abundant bacterial OTUs in IB2. These OTUs represent $89 \pm 2\%$ of the total IB2 bacterial community abundance in all IB2-inoculated bottles.

References

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