Cover sheet for SI

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Complete perchlorate reduction using methane as the sole electron donor and carbon source

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Table S1. Equations applied for the CH_4-permeation calculations:

$$P_{m-lf} = \frac{\alpha P_0 + \beta P_{hs}}{\alpha + \beta}$$

$$Q\frac{P_{hs}}{H}k_2 = \left(P_0 - \left(\frac{\alpha P_0 + \beta P_{hs}}{\alpha + \beta}\right)\right)\pi(d_m - Z_m)L_m n_m k_1$$

in which $\alpha = k_1 K_m Z_{lf} H(d_m - Z_m)$, $\beta = k_2 D_{lf} Z_m (d_m + Z_{lf})$

$P_{m\text{-}lf}$	Methane pressure at the interface of membrane and liquid film (bar)
P_0	Methane pressure in the hollow-fiber lumen (bar)
P_{hs}	Methane pressure in the headspace (bar)
Q	Water flow rate in the serum bottle (7.2×10^{-4} m^3/d)
H	Henry's law constant of CH_4 (0.7512 m^3 bar/mol)
D_{lf}	CH_4-diffusion coefficient in water (1.9×10^{-4} m^2/d)[1]
k_1	Coefficient that converts CH_4 from volume to mass (1g/0.00154 m^3 @ standard temperature and pressure)
k_2	Coefficient that converts CH_4 from moles to mass (16g/mol)
d_m	Hollow-fiber out diameter (2.8×10^{-4} m)
Z_m	Membrane thickness (5.0×10^{-5} m)
L_m	Hollow-fiber length (m)
n_m	Number of hollow fibers (32)

Table S2. Experimental parameters for the CH_4-permeation test

Parameter	Temperature (K)	Pressure (bar)	Flow rate (m^3/d)	Fiber Length (m)	Fiber Numbers	Fiber O.D. (μm)	Fiber thickness (μm)	Km[a]
Composite fiber	298	1.00	7.2×10^{-4}	0.05	32	2.8×10^{-4}	0.5×10^{-4}	1.03×10^{-7}

a: units are m^3 CH_4 @ standard temperature and pressure - m membrane thickness/m^2 hollow fiber surface area - d – bar.

Table S3. The influent and effluent concentrations of electron acceptors for each stage

	NO_3^--N		NO_2^--N		ClO_4^-	
stages	Influent (mg N/L)	Effluent (mg N/L)	Influent (mg N/L)	Effluent (mg N/L)	Influent (mg/L)	Effluent (mg/L)
1	NA	0	1.69±0.006	0.027±0.021	1.32±0.09	1.01±0.14
2	NA	0	NA	0	1.01±0.008	0.17±0.11
3	1.21±0.09	0.000±0.000	NA	0	1.02±0.017	0.011±0.006
4	11.3±0.40	3.51±1.25	NA	0	1.04±0.052	1.02±0.069
5	4.49±0.04	0.023±0.029	NA	0	1.0±0.003	0.006±0.007
6	NA	0	NA	0	5.43±0.017	0.057±0.097
7	NA	0	5.22±0.13	0.000±0.000	5.07±0.096	2.54±0.40

Table S4. Primers and PCR conditions for tested genes

PCR Program	Primers	Sequence	Target Gene	Ref	Slope	Efficiency
95°C 1 min (95°C 5 sec 60°C 31 sec 72°C 20 sec)×40 72°C 1 min	320F 598R	5’-GCGCCCACCACTACATGTAYGGNCC-3’ 5’-GGTGGTCGCCGTACCARTCRAA-3’	*pcrA*	2	-.3.106	1.10
94°C 2 min (94°C 30 sec 58°C 20 sec 72°C 60 sec)×40 72°C 10 min	M2f M2r	5'-TAYGTSGGGCAGGARAAACTG-3' 5'-CGTAGA AGA AGCTGGTGCTGTT-3'	*narG*	3	-3.144	1.08
95°C 2 min (94°C 30 sec 60°C 60 sec 72°C 60 sec)×40 72°C 5 min	cd3af R3cd	5’-GTSAACGTSAAGGARACSGG-3’ 5’-GASTTCGGRTGSGTCTTGA-3’	*nirS*	4	-.3.419	0.995
94°C 10 min (94°C 30 sec 58°C 300 sec 72°C 60 sec)×40 72°C 1 min	Mlas rev	5'-GGTGGTGTMGGDTTCACMCARTA-3' 5'-CGTTCATBGCGTAGTTVGGRTAGT-3'	*mcrA*	5	-3.221	1.04
95°C 10 min (95°C 60 sec 60°C 60 sec 72°C 60 sec)×40 72°C 5 min	A189F MB661R	5'-GGNGACTGGGACTTCTGG-3' 5'-CCGGMGCAACGTCYTTACC-3'	*pMMO*	6	-3.383	0.98
95°C 2 min (95°C 10 sec 56°C 20 sec 68°C 20 sec)×40 72°C 1 min	16SF 16SR	5'-GTGSTGCAYGGYTGTCGTCA-3' 5'-ACGTCRTCCMCACCTTCCTC-3'	*16S rDNA* for bacteria	7	-3.215	1.05
94°C 10 min (94°C 30 sec 58°C 20 sec 72°C 60 sec)×40 72°C 1 min	ARC787F ARC1059R	5'-ATTAGATACCCSBGTAGTCC-3' 5'-GCCATGCACC WCCTCT-3'	*16S rDNA* for archaea	8	-3.630	0.89

Table S5. Pearson Correlation Matrix

		A16S	B16S	mcrA	MMO	pcrA	narG	nirS	CH_4 flux	NO_3^- flux	NO_2^- flux	ClO_4^- flux
A16S	Pearson Correlation	1.000	0.135	-0.323	0.579	0.983**	0.210	0.919**	0.304	-0.169	0.797	0.439
	Sig. 2-tailed		0.799	0.532	0.229	0.000	0.689	0.010	0.558	0.748	0.058	0.384
B16S	Pearson Correlation	0.135	1.000	0.835*	0.861*	0.137	0.903*	0.484	0.277	0.326	-0.145	-0.173
	Sig. 2-tailed	0.799		0.039	0.028	0.796	0.014	0.331	0.595	0.529	0.784	0.743
mcrA	Pearson Correlation	-0.323	0.835*	1.000	0.549	-0.325	0.834*	0.020	0.282	0.554	-0.542	-0.487
	Sig. 2-tailed	0.532	0.039		0.259	0.530	0.039	0.970	0.588	0.254	0.267	0.327
pMMO	Pearson Correlation	0.579	0.861*	0.549	1.000	0.599	0.863*	0.843*	0.416	0.209	0.329	-0.090
	Sig. 2-tailed	0.229	0.028	0.259		0.209	0.027	0.035	0.411	0.691	0.524	0.865
pcrA	Pearson Correlation	0.983**	0.137	-0.325	0.599	1.000	0.195	0.933**	0.251	-0.243	0.880*	0.339
	Sig. 2-tailed	0.000	0.796	0.530	0.209		0.711	0.007	0.632	0.642	0.021	0.511
narG	Pearson Correlation	0.210	0.903*	0.834*	0.863*	0.195	1.000	0.508	0.624	0.625	-0.109	-0.360
	Sig. 2-tailed	0.689	0.014	0.039	0.027	0.711		0.303	0.186	0.185	0.837	0.483
nirS	Pearson Correlation	0.919**	0.484	0.020	0.843*	0.933**	0.508	1.000	0.341	-0.078	0.721	0.230
	Sig. 2-tailed	0.010	0.331	0.970	0.035	0.007	0.303		0.508	0.883	0.106	0.661
CH_4 flux	Pearson Correlation	0.304	0.277	0.282	0.416	0.251	0.624	0.341	1.000	0.864*	0.075	-0.401
	Sig. 2-tailed	0.558	0.595	0.588	0.411	0.632	0.186	0.508		0.027	0.888	0.431
NO_3^- flux	Pearson Correlation	-0.169	0.326	0.554	0.209	-0.243	0.625	-0.078	0.864*	1.000	-0.428	-0.522
	Sig. 2-tailed	0.748	0.529	0.254	0.691	0.642	0.185	0.883	0.027			
NO_2^- flux	Pearson Correlation	0.797	-0.145	-0.542	0.329	0.880*	-0.109	0.721	0.075	-0.428	1.000	0.139
	Sig. 2-tailed	0.058	0.784	0.267	0.524	0.021	0.837	0.106	0.888	0.397		
ClO_4^- flux	Pearson Correlation	0.439	-0.173	-0.487	-0.090	0.339	-0.360	0.230	-0.401	-0.522	0.139	1.000
	Sig. 2-tailed	0.384	0.743	0.327	0.865	0.511	0.483	0.661	0.431	0.288	0.793	

*. Correlation is significant at the 0.05 level; ** Correlation is significant at the 0.01 level (2-tailed).

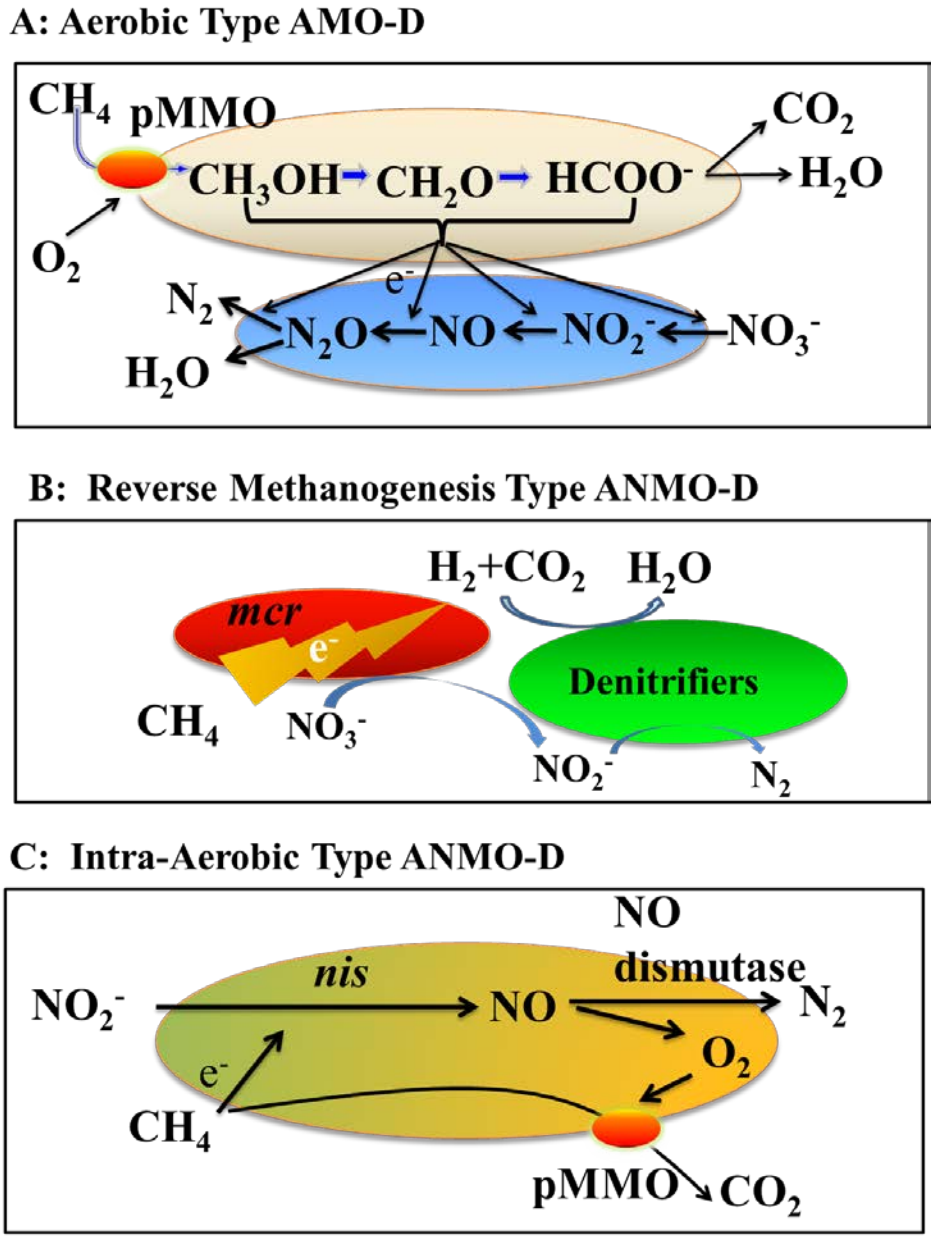

Figure S1. The proposed AMO-D and ANMO-D pathways for CH_4 oxidation coupled to NO_3^-/NO_2^- reduction. A: AMO-D requires two microorganisms: aerobic methanotrophs oxidize methane and produce organic compounds, which are further used by denitrifiers to reduce NO_3^- to N_2. B: Reverse-methanogenesis-type ANMO-D requires two microorganisms: Archaea reduce NO_3^- to NO_2^- and produces H_2 via reverse methanogenesis, and a denitrifier that oxidizes the H_2 to drive NO_2^- respiration to N_2. C: Intra-aerobic-type ANMO-D is carried out by one bacterium, which dismutates NO to form N_2 and O_2, with the O_2 used as a co-substrate for methane mono-oxygenation by the same bacterium.

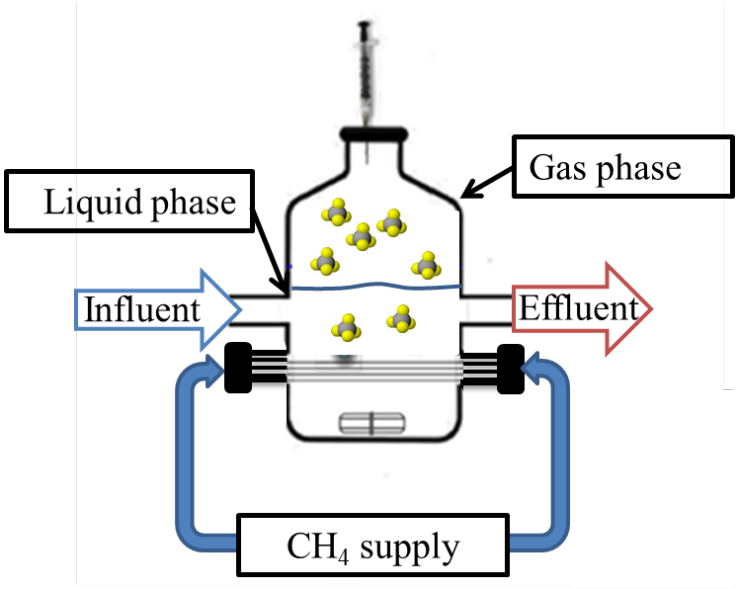

Figure S2. Schematic of the set up for the CH_4-permeation experiment (This figure is our own work and is substantially modified from the original figure in Tang et al. (2012)).[9]

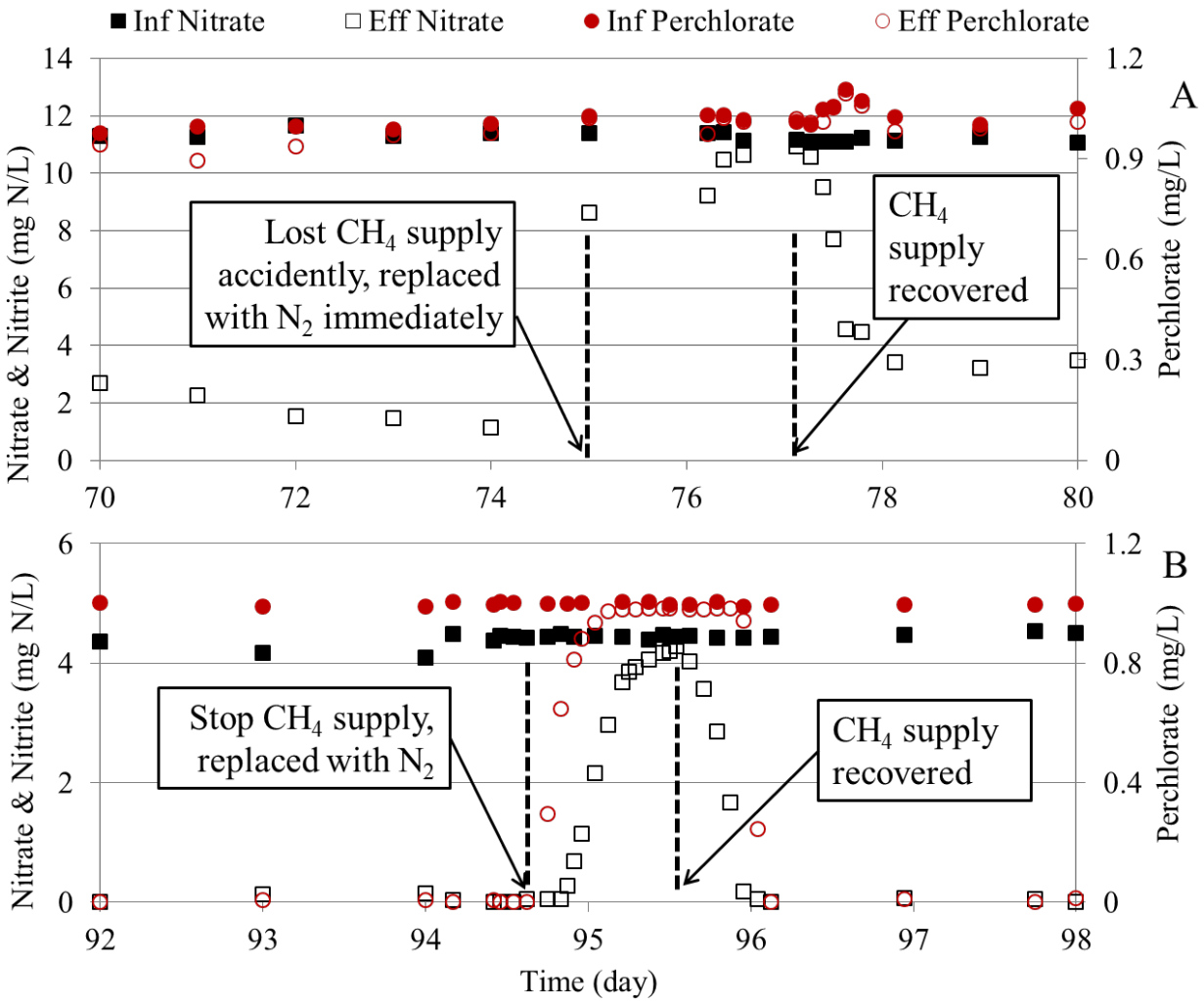

Figure S3. The NO_3^- and ClO_4^- concentrations in the MBfR influent and effluent in Stage 4 (A) and Stage 5 (B), when CH_4 supply was stopped and recovered.

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