



Supplement of

Fractionation of stable carbon isotopes during acetate consumption by methanogenic and sulfidogenic microbial communities in rice paddy soils and lake sediments

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Legends of the supplemental figures

- Fig.S1: Acetate conversion to CH₄ and CO₂ in unbuffered suspensions of paddy soil from Vercelli (Italy) without additions (water control), with gypsum, and with CH₃F. The panels show the temporal change of (A) concentrations of acetate; (B) δ^{13} C of acetate; (C) partial pressures of CH₄ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CH₄; (E) partial pressures of CO₂ (1 ppmv = 10⁻⁶ bar); and (F) δ^{13} C of CO₂. Means ± SE, n = 2.
- Fig.S2: Mariotti plots of (A, B) acetate consumption and (C, D) CH₄ production in (A, C) the absence (control) and (B, D) the presence of gypsum (+ sulfate) in 4 replicates of unbuffered suspensions of paddy soil from Vercelli.
- Fig.S3: Acetate conversion to CH₄ and CO₂ in phosphate-buffered (pH 7) suspensions of paddy soil from Vercelli (Italy) without (H₂O) and with addition of acetate (acetate) and with acetate and CH₃F. The soil was preincubated in the absence of sulfate, but sulfate (gypsum) was for the experimental incubation. The panels show the temporal change of (A) concentrations of acetate; (B) δ^{13} C of acetate; (C) partial pressures of CH₄ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CH₄; (E) partial pressures of CO₂ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CO₂. Means ± SE, n = 3.
- Fig.S4: Balance of the produced CH₄ + TIC against acetate consumed in phosphate-buffered suspensions of paddy soil from Vercelli and IRRI, and of sediments from the NE and SW basin of Lake Fuchskuhle. The figures show individual replicates (n = 3) of the unamended control (methanogenic conditions); of the experiment plus gypsum (CaSO₄-1); of preincubation and experiment plus gypsum (CaSO₄-2). The diagonal line indicates stoichiometric conversion (disproportionation) of acetate to CH₄ + CO₂.
- Fig.S5: Acetate conversion to CH₄ and CO₂ in phosphate-buffered (pH 7) suspensions of paddy soil from IRRI (the Philippines) without addition of acetate (H₂O), with addition of acetate (acetate) and with addition of acetate and CH₃F (Acet + CH₃F). The soil was preincubated in the absence of sulfate, and sulfate (gypsum) was also not added for the experimental incubation. The panels show the temporal change of (A) concentrations of acetate; (B) δ^{13} C of acetate; (C) partial pressures of CH₄ (1 ppmv =

10⁻⁶ bar); (D) δ^{13} C of CH₄; (E) partial pressures of CO₂ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CO₂. Means ± SE, n = 3.

- Fig.S6: Acetate conversion to CH₄ and CO₂ in phosphate-buffered (pH 7) suspensions of paddy soil from IRRI (the Philippines) without addition of acetate (H₂O), with addition of acetate (acetate) and with addition of acetate and CH₃F (Acet + CH₃F). The soil was preincubated in the absence of sulfate, but sulfate (gypsum) was for the experimental incubation. The panels show the temporal change of (A) concentrations of acetate; (B) δ^{13} C of acetate; (C) partial pressures of CH₄ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CH₄; (E) partial pressures of CO₂ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CO₂. Means \pm SE, n = 3.
- Fig.S7: Acetate conversion to CH₄ and CO₂ in phosphate-buffered (pH 7) suspensions of paddy soil from IRRI (the Philippines) without addition of acetate (H₂O), with addition of acetate (acetate) and with addition of acetate and CH₃F (Acet + CH₃F). The soil was preincubated in the presence of sulfate, and sulfate (gypsum) was also added for the experimental incubation. The panels show the temporal change of (A) concentrations of acetate; (B) δ^{13} C of acetate; (C) partial pressures of CH₄ (1 ppmv = 10^{-6} bar); (D) δ^{13} C of CH₄; (E) partial pressures of CO₂ (1 ppmv = 10^{-6} bar); (D) δ^{13} C
- Fig.S8: Balance of the produced CH₄ + TIC against acetate consumed in phosphate-buffered suspensions of paddy soil from Vercelli and IRRI, and of sediments from the NE and SW basin of Lake Fuchskuhle. The figures show individual replicates (n = 3) of the unamended control (methanogenic conditions); of the experiment plus gypsum (CaSO₄-1); of preincubation and experiment plus gypsum (CaSO₄-2). The diagonal line indicates stoichiometric conversion (disproportionation) of acetate to CH₄ + CO₂. The figure is identical to Fig.S4, but background production of CH₄ and CO₂ in the water control without addition of acetate was substracted.
- Fig.S9: Mariotti plots of (A, B, E) acetate consumption and (C, D, F) CH₄ production in (A, C) the absence (no sulfate); (B, D) the presence of gypsum (with sulfate); and (E, F)

after preincubation with sulfate and in the presence of sulfate in 3 replicates of phosphate-buffered suspensions of paddy soil from Vercelli.

- Fig.S10: Mariotti plots of (A, B, E) acetate consumption and (C, D, F) CH₄ production in (A, C) the absence (no sulfate); (B, D) the presence of gypsum (with sulfate); and (E, F) after preincubation with sulfate and in the presence of sulfate in 3 replicates of phosphate-buffered suspensions of paddy soil from the IRRI.
- Fig.S11: Mariotti plots of acetate consumption in the presence of methyl fluoride (CH₃F) in (A, C) the presence of gypsum (with sulfate); and (B, D) after preincubation with sulfate and in the presence of sulfate, in 3 replicates of phosphate-buffered suspensions of paddy soil from (A, B) Vercelli and (C, D) the IRRI.
- Fig.S12: Acetate conversion to CH₄ and CO₂ in phosphate-buffered (pH 7) suspensions of lake sediment from the SW basin of Fuchskuhle without addition of acetate (H₂O), with addition of acetate (acetate) and with addition of acetate and CH₃F (Acet + CH₃F). The sediment was incubated in the absence of sulfate. The panels show the temporal change of (A) concentrations of acetate; (B) δ^{13} C of acetate; (C) partial pressures of CH₄ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CH₄; (E) partial pressures of CO₂ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CO₂. Means ± SE, n = 3.
- Fig.S13: Acetate conversion to CH₄ and CO₂ in phosphate-buffered (pH 7) suspensions of lake sediment from the SW basin of Fuchskuhle without addition of acetate (H₂O), with addition of acetate (acetate) and with addition of acetate and CH₃F (Acet + CH₃F). The sediment was incubated in the presence of sulfate, and sulfate (gypsum) was also not added for the experimental incubation. The panels show the temporal change of (A) concentrations of acetate; (B) δ^{13} C of acetate; (C) partial pressures of CH₄ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CH₄; (E) partial pressures of CO₂ (1 ppmv = 10⁻⁶ bar); (D) δ^{13} C of CO₂. Means ± SE, n = 3.
- Fig.S14: Mariotti plots of (A, B) acetate consumption and (C, D) CH₄ production in (A, C) the absence (no sulfate) and (B, D) the presence (both preincubation and experimental incubation) of gypsum (plus sulfate) in 3 replicates of buffered suspensions of lake sediment from the NE basin of Fuchskuhle.

- Fig.S15: Mariotti plots of (A, B) acetate consumption and (C, D) CH₄ production in (A, C) the absence (no sulfate) and (B, D) the presence (both preincubation and experimental incubation) of gypsum (plus sulfate) in 4 replicates of buffered suspensions of lake sediment from the SW basin of Fuchskuhle.
- Fig.S16: Mariotti plots of acetate consumption in the presence of methyl fluoride (CH₃F) after preincubation and in the presence of gypsum (with sulfate) in 3 replicates of phosphate-buffered suspensions of lake sediment from the (A) NE basin and (B) SW basin of Fuchskuhle.

Table S1: Epsilon values (negative values) determined using Mariotti plots of ¹³C-acetate or ¹³CH₄ in unbuffered or phosphate-buffer suspensions without and with addition of sulfate, also after preincubation with sulfate, and in the absence and presence of methyl fluoride

		ε									
						Preincubation				Preincuba	ation
Soil	$\delta^{13}Cused$	without sulfate		with sulfate		with sulfate		with sulfate + CH3F		with sulfate + CH3F	
		mean	SE	mean	SE	mean	SE	mean	SE	mean	SE
Vercelli-soil	acetate	17.2	0.93	19.5	0.34						
	CH4	18.9	2.03	14.1	1.96						
IRRI-soil	acetate	20.2	0.80	20.6	0.62						
	CH4	21.6	3.23	19.4	1.42						
Vercelli-buffer	acetate	17.7	0.53	21.1	1.27	21.4	0.20	21.8	2.47	24.3	0.54
	CH4	29.7	0.61	22.9	1.30	24.9	1.54				
IRRI-buffer	acetate	20.0	2.41	19.1	0.51	21.2	1.98	23.8	5.59	10.1	1.12
	CH4	37.2	6.86	28.9	3.68	32.8	6.82				
NE-buffer	acetate	18.9	1.01			19.7	1.41			26.3	3.28
	CH4	27.9	2.46								
SW-buffer	acetate	11.4	1.47			14.3	1.30			17.8	0.06
	CH4	27.4	3.20								-

Vercelli Soil slurry



Vercelli

control

+ sulfate



Vercelli with sulfate





IRRI without sulfate



IRRI with sulfate



IRRI with sulfate, with preincubation





Vercelli







IRRI



Incubation with sulfate + CH3F



Fuchskuhle – **SW** without sulfate



Fuchskuhle – **SW** with sulfate



Fuchskuhle – **NE**



Fuchskuhle – SW



Fuchskuhle Incubation with sulfate and with CH3F





В