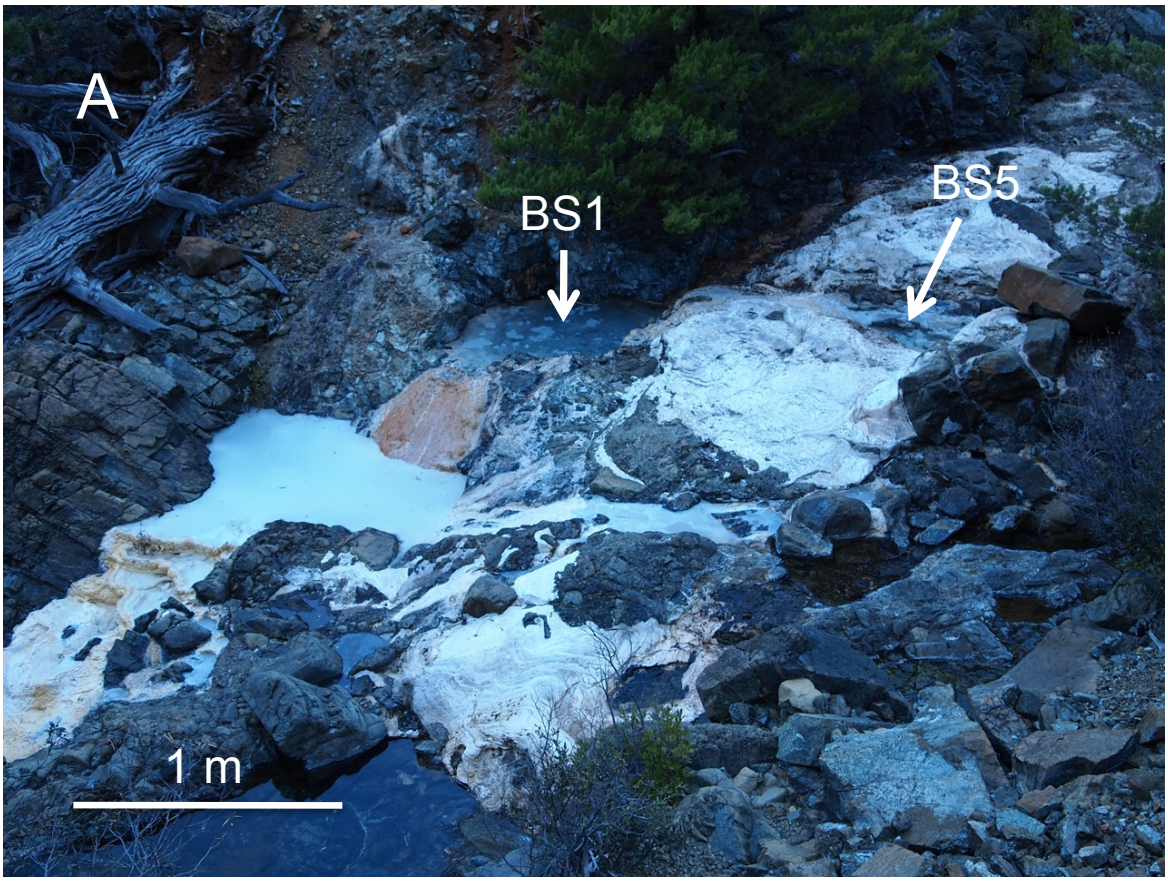
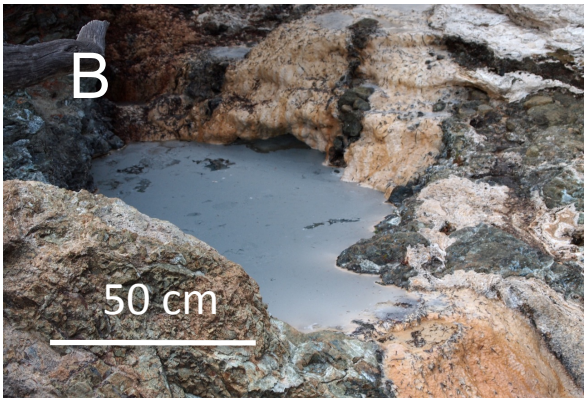
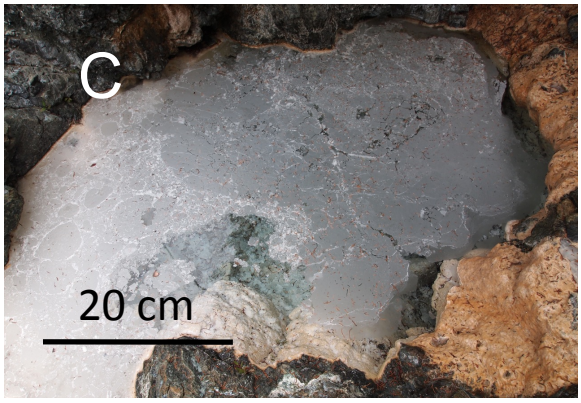
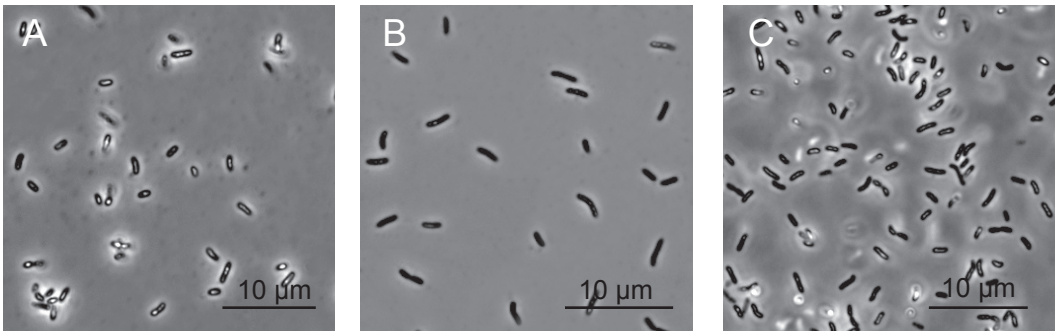


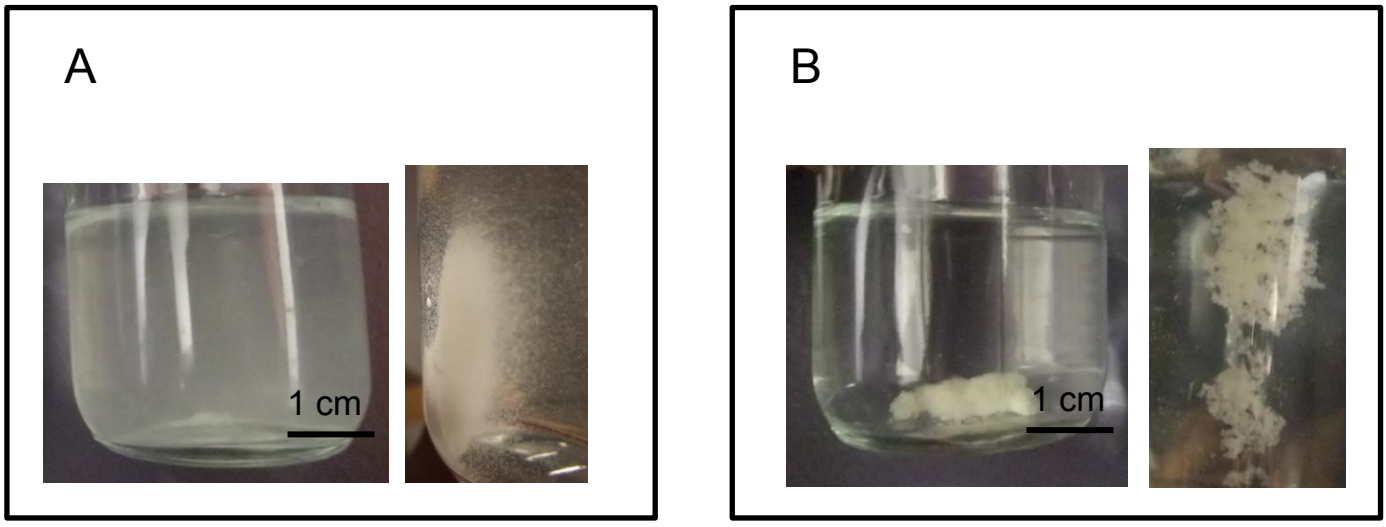




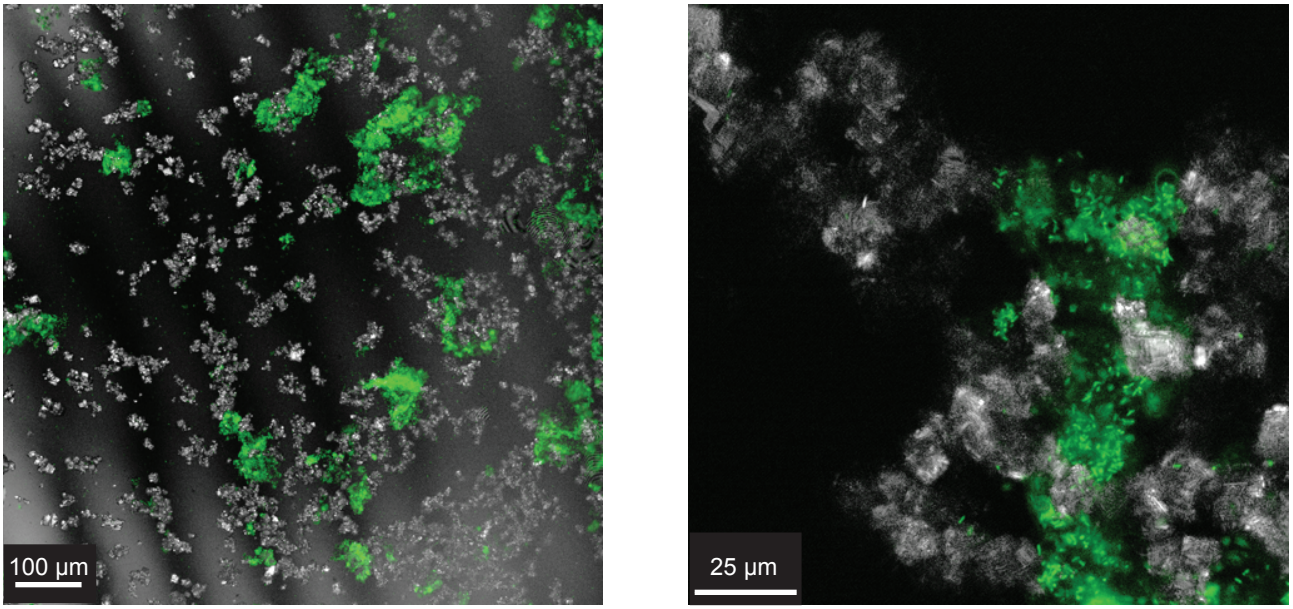
Supplementary Figure 1: Spring fluid was collected at Barnes springs (BS) for the microbial isolation.
A; BS complex, B; BS1 and C; BS5. The surface of springs is covered with calcite skin. Due to the gas bubbling from the bottom, part of the calcite at the surface was broken in both springs.



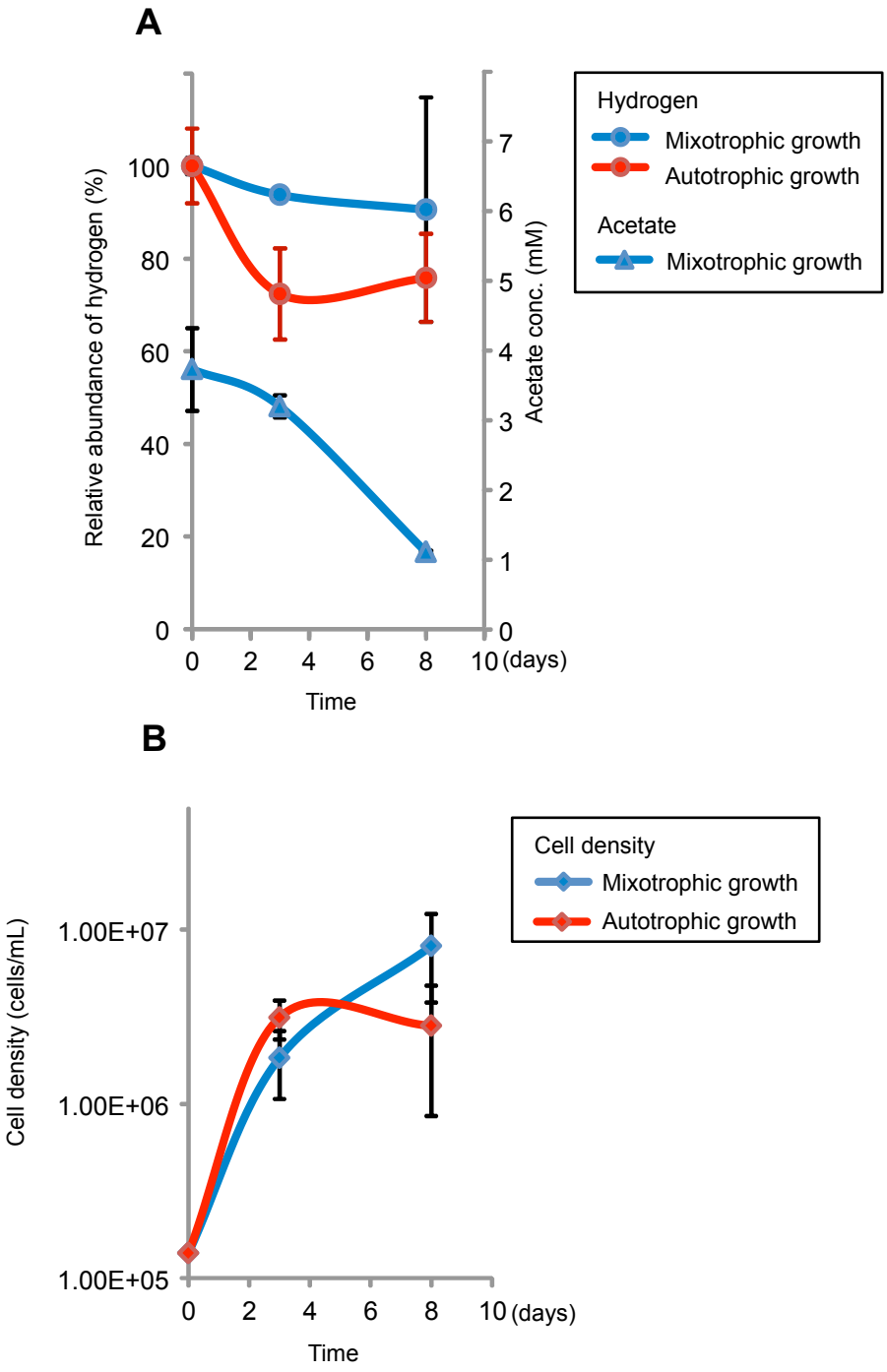
Supplementary Figure 2 Phase contrast images of isolated strains A) A1, B) B1 and C) H1



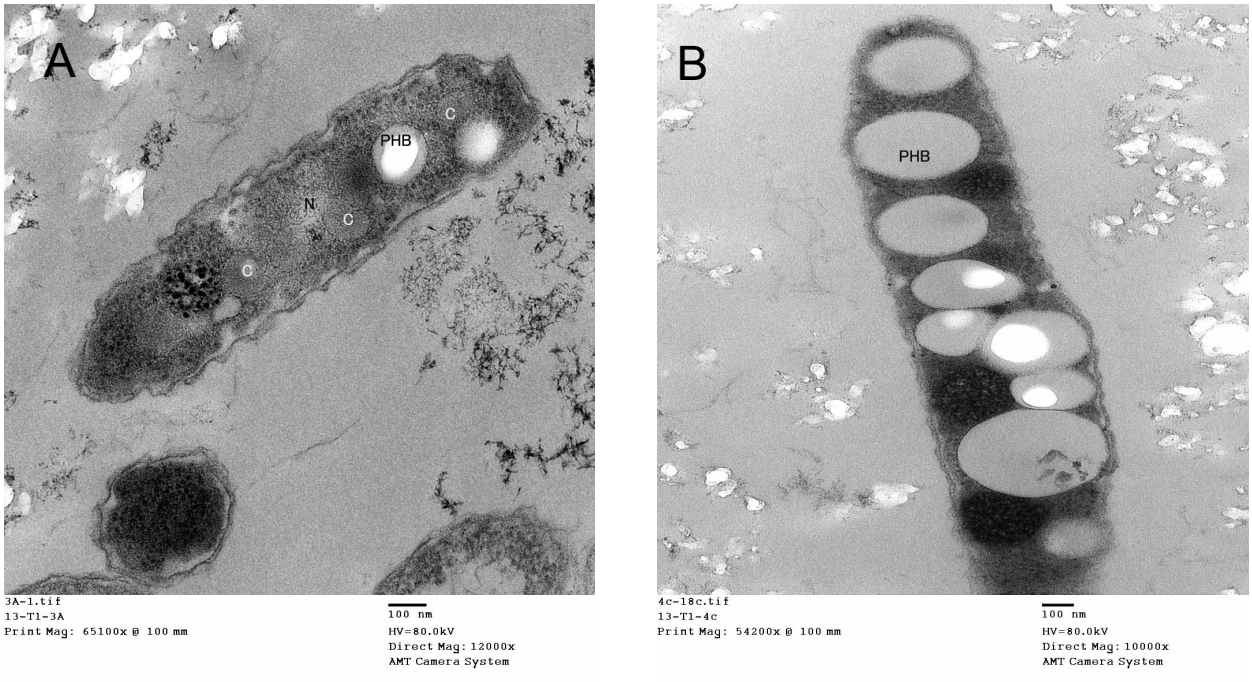
Supplementary Figure 3 Two-week cultivation of strain A1 in the liquid medium
A) no cell control, B) strain A1



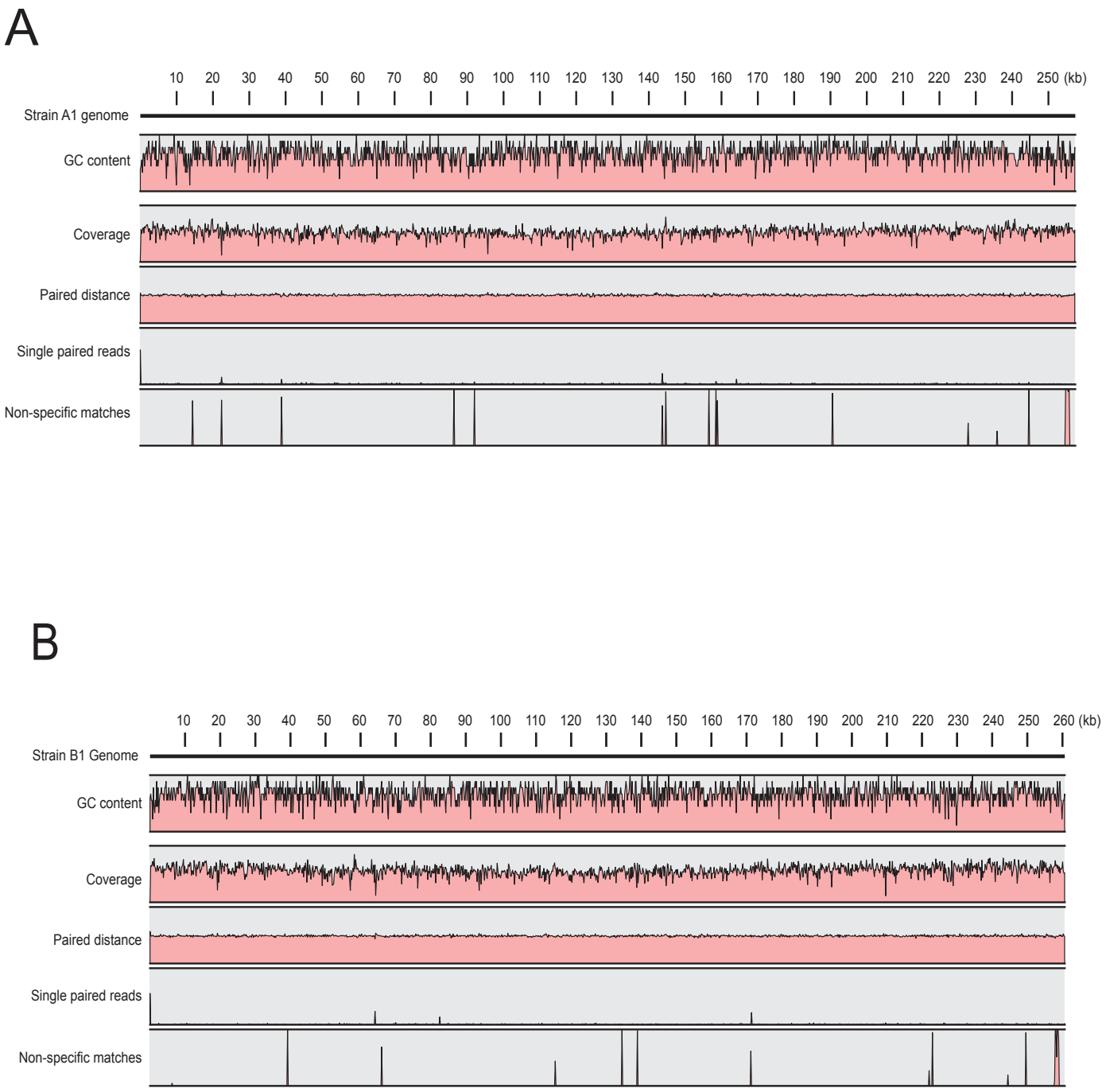
Supplementary Figure 4 Confocal microscopic observations of the cell aggregates on calcium carbonate with different magnifications. Strain A1 was grown autotrophically for three days. Cells were stained with SYBR Green I (Green). Calcium carbonate precipitate was also observed (White).



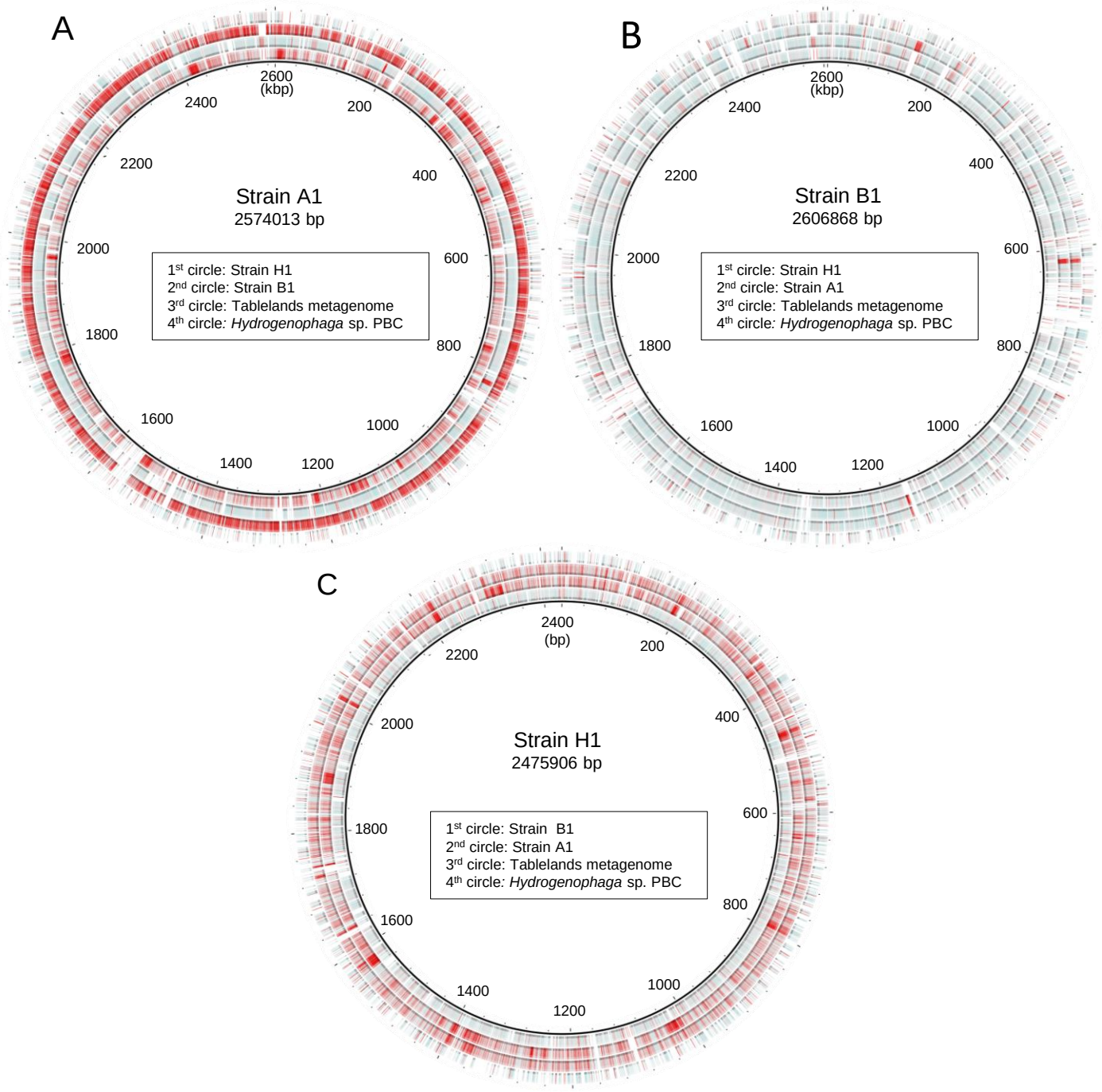
Supplementary Figure 5; Growth curves of strain A1 (n=3). A) Acetate (triangle) and hydrogen (circle) consumption and B) cell densities (triangle). Samples were collected at 0, 3 and 8 days after cell innoculation. To count cell numbers, the samples were shaken vigorously. Thus the cells were detouched from calcite and the autotrophic growth for strain A1 was stopped. Bars indicate standard deviation.



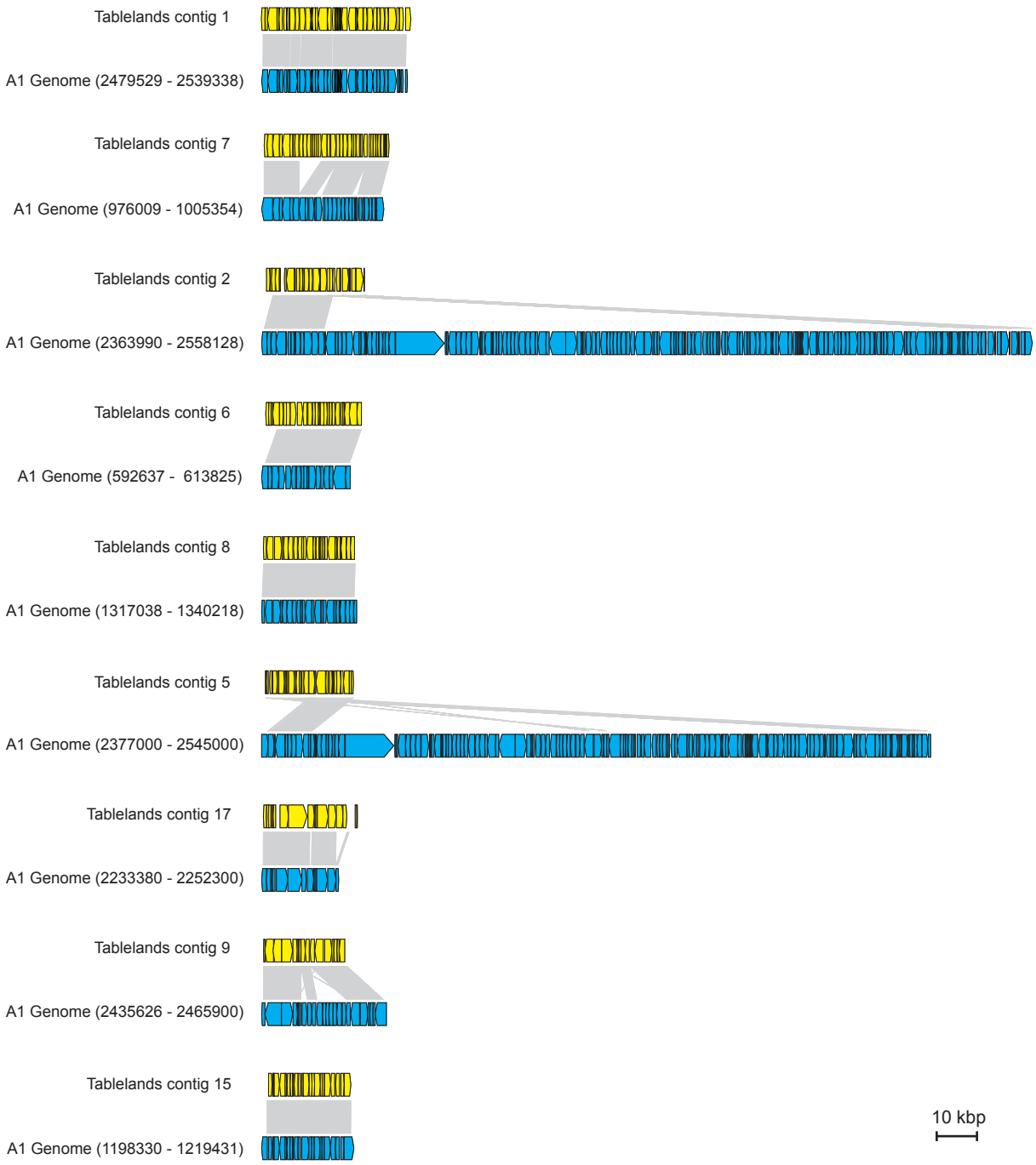
Supplementary Figure 6 Thin-section TEM observations of strain A1. The cells were fixed after five-days cultivations. A) Autotrophic growth, B) Heterotrophic growth. C; Carboxysome, N; Nucleoid, PHB; Polyhydroxybutyrate



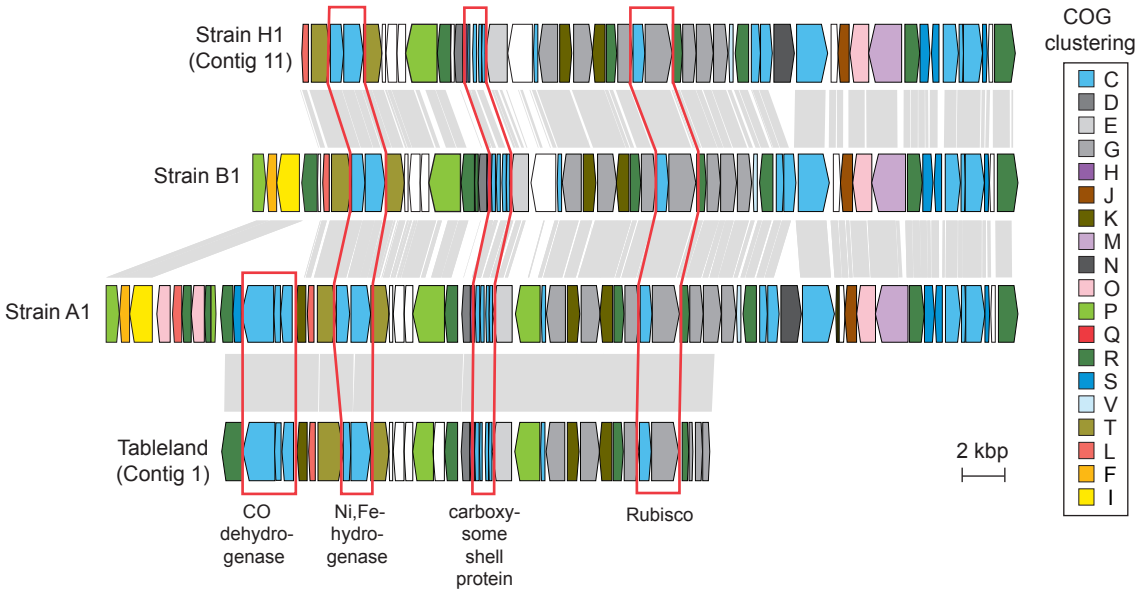
Supplementary Figure 7 Raw reads mapping to the gap-closed genome sequences of strain A1 (A) and B1 (B)



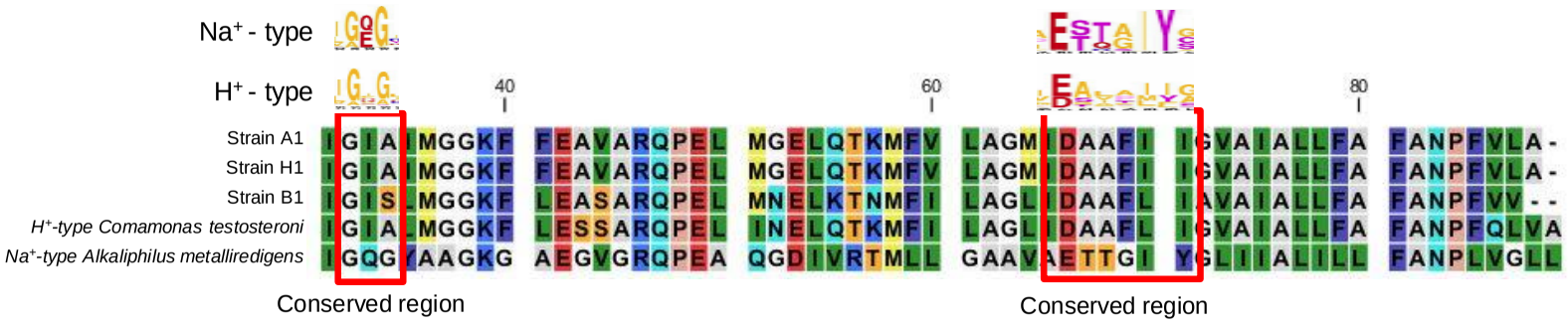
Supplementary Figure 8 Genome-wide comparisons of strain A1 (A), B1 (B) and H1 (C). The inner scales designate the coordinates (in kbp). The first to fourth circles show the blastn analysis of strains A1, B1 and H1 against each genome or metagenome as indicated in the figure. The lower (gray) and upper (red) identity threshold in the blastn analysis is >90 or >95 respectively.



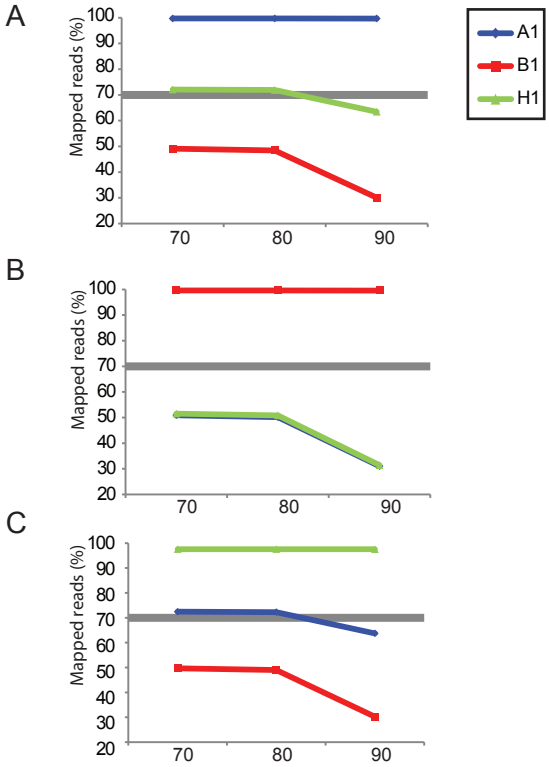
Supplementary Figure 9 Blastn comparisons of the long contigs from the Tablelands[1] (yellow) with the genome sequence of strain A1 (Blue)



Supplementary Figure 10 Comparisons of predicted gene clusters encoding carbon fixation related proteins



Supplementary Figure 11 Comparison of cation binding site in F_0F_1-ATPase. The conserved regions were reported by Mulkidjanian *et al.*[2]



Supplementary Figure 12 Mapped percent of raw reads to each genome. Raw reads from strain A1, B1 and H1 with average read length ~100 bp were mapped to the (draft) genome sequences of strains A1 (A), B1 (B) and H1 (C). The length cut off is 95 %. The numbers shown in the bottom of each graph indicate the similarity cut-off value (e.g. 70 means 70% similarity in 95% length.).

Supplementary Table 1 Geochemical properties of the studied terrestrial serpentinizing sites and other alkaline soil or ground water

	Present*										Absent*		
	USA		Portugal	Canada	Finland			Japan	USA	South Africa	USA	Norway	Mid-Atrantic
	The Cedars		**Cabeço de Vide**	**Table lands**	**Outokumpu**			**Mizunami**	**Chicago**	**Kalahari Shield**	**The Cedars**	**Lake Ophiolite**	**Lost City**
Component	***BS1***	***BS5***	***AC3***	***WHC2b***	***100 m deep***	***1000 m deep***	***1500 m deep***	***MIU (Sep, 2005)***	***LC4***	***Evander, EV818 DRLW***	***GPS1***	***Gw3***	***30° N*** [e]
pH	11.5	11.6	11.4	12.3		8-9		8.2	12.5	8.0	11.9	8.8	9-9.8
E_h (mV)	-250	-550	-383	-552	-	-	-	-308	-45	-	-656	-	-
DO (mM)	<dl	<dl	<dl	-	-	-	-	-	<0.12	<dl	<dl	-	-
N_2 (% by vol)	49.6	53.6	-	-	33	40	38	-	-	-	36.6	-	-
H_2 (% by vol)	39.2	34	-	-	0.024	<0.003	0.02	-	-	-	50.9	-	-
CH_4 (% by vol)	6.5	5.3	-	-	58	49	48	-	-	-	7.4	-	-
$N_{2\,(aq)}$ mM	-	-	-	-	-	-	-	-	-	-	-	-	-
$H_{2\,(aq)}$ mM	-	-	-	1.04	-	-	-	-	-	-	-	-	0.25-0.43
$CH_{2\,(aq)}$ mM	-	-	-	0.02	-	-	-	-	-	-	-	-	0.13-0.28
Na^+ (mM)	2.0	1.98	2.26	-	57.42	76.56	57.85	22.7	2.2	18.90	14.69	0.54	479-485
K^+ (mM)	0.038	0.03	0.12	-	0.76	0.59	1.64	0.041	1.2	-	0.13	0.012	-
Ca^{2+} (mM)	-	1.17	0.58	1.12	38.29	55.55	65.81	17.2	27	13.70	0.94	0.031	21.0-23.3
Mg^{2+} (mM)	-	0.036	<dl	0.03	0.77	0.71	3.79	0.07	<0.001	-	0.004	0.42	9-19
PO_4^{3-} (mM)	<dl	<dl	-	-	-	-	-	<0.105	-	-	<dl	0.00063	-
HCO_3^- (mM)	<dl	<dl	-	-	-	-	-	-	-	-	<dl	-	-
Cl^- (mM)	1.7	1.49	1.48	9.59	128.34	184.75	198.57	52.6	2.1	40.00	8.73	0.54	546-549
NO_3^- (mM)	<dl	<dl	<dl	-	-	-	-	<0.005	-	0.56	<dl	0.019	-
SO_4^{2-} (mM)	<dl	0.001	0.03	-	1.58[§]	1.67[§]	1.65[§]	0.014	0.055	3.08	<dl	0.038	5.9-12.9
TIC (mM)	-	0.07	0.11	0.37	-	-	-	-	-	0.275 (DIC)	0.035	0.792 (DIC)	-
DOC (mM)	-	0.02	-	0.08	-	-	-	0.13	-	-	0.17	0.00135 (TOC)	-
Ca/Na ratio	NA	0.59	0.26	NA	0.67	0.73	1.14	0.76	12.27	0.72	0.06	0.06	~0.05
occurrence of serpentinization	+	+	+	+	-	-	+	-	-	-	+	+	+
Reference	This study	3, 4	5, 6	7	8	8	8	9	10	11	3,4	12	13

-; not available, <dl; below the detection lmit, * Alkaline environments where strains A1, B1 and H1 related phylotypes are present or absent, §Total S concentration

Supplementary Table 2 Percent similarities of 16S rRNA gene sequence between strains A1, B1 and H1, and type strains in the three closest genera

		1	2	3	4	5	6
Strain A1	1	100	-	-	-	-	-
Strain B1	2	97.7	100	-	-	-	-
Strain H1	3	98.95	98.75	100	-	-	-
Malikia granosa DSM 15619^T	4	95.56	96.51	96.58	100	-	-
Hydrogenophaga flava DSM 619^T	5	94.96	96.12	95.98	96.64	100	-
Macromonas bipunctata IAM 14880^T	6	94.07	94.72	94.65	96.5	95.61	100

Supplementary Table 3 Substrate utilization for strains A1, B1 and H1

		A1	B1	H1
Autotrophic growth				
Electron donor and Carbon source	***Electron Acceptor***			
H_2 + $CaCO_3$	Oxygen	++	-	++
H_2 + $CaCO_3$	Thiosulfate	-	NT	NT
H_2 + $CaCO_3$	Nitrate	NT	-	++
Thiosulfate + $CaCO_3$	Oxygen	++	-	NT
Heterotrophic growth				
Electron donor and carbon source	***Electron Acceptor***			
Formate + $CaCO_3$	Oxygen	+	+	+
Acetate + $CaCO_3$	Oxygen	+	+++	++
Propionate + $CaCO_3$	Oxygen	-	-	NT
Butyrate + $CaCO_3$	Oxygen	+	+++	+
Lactate + $CaCO_3$	Oxygen	+	+++	++
Pyruvate + $CaCO_3$	Oxygen	+	+++	NT
Glucose + $CaCO_3$	Oxygen	-	+++	++
Glutamate + $CaCO_3$	Oxygen	-	-	NT
Glycerol + $CaCO_3$	Oxygen	-	-	NT
Cyclohexane + $CaCO_3$	Oxygen	+	±	-
Electron donor and/or carbon source				
Formate + H_2 + $CaCO_3$	Oxygen	++	+	++
Acetate + H_2 + $CaCO_3$	Oxygen	+++	+++	+++
Butyrate + H_2 + $CaCO_3$	Oxygen	+++	+++	+
Lactate + H_2 + $CaCO_3$	Oxygen	+++	+++	+++
Pyruvate + H_2 + $CaCO_3$	Oxygen	+++	+++	NT
Glucose + H_2 + $CaCO_3$	Oxygen	+	+++	+++
Cyclohexane + H_2 + $CaCO_3$	Oxygen	+++	±	-
Electron Acceptor				
Acetate + H_2 + $CaCO_3$	Oxygen	+++	+++	+++
Acetate + H_2 + $CaCO_3$	Nitrate	-	+++	+++
Acetate + H_2 + $CaCO_3$	Sulfate	-	-	-
Acetate + H_2 + $CaCO_3$	Thiosulfate	+++	-	-
Acetate + H_2 + $CaCO_3$	Fumarate	-	-	-
Acetate + H_2 + $CaCO_3$	Iron	-	-	-
Fermentation				
Glucose		-	+++	+++
Cyclohexane		-	-	NT

+; positive, - Negative, NT; not tested

Supplementary Table 4 Statstics for assembled genomes of strains A1, B1 and H1

Sample	Word Length	Contig Count*	Contig Count Over_1kbp	N50 Length	Max Contig	Total Contig Length	Total Reads	Reads assembled
A1	53	36	25	226,682	471,724	2,561,383	3,857,324	3,829,752
B1	53	22	20	267,175	516,238	2,611,506	4,385,866	4,346,338
H1	53	96	70	87,939	198,940	2,511,299	4,927,302	4,836,043

*Length cut off > 300 bp

Supplementary Table 5 Primer sequences for the qPCR analyses

Targeted Genes	Forward (5' - 3')	Reverse (5' - 3')
SRAA-0853	CGTCGCCAACTGGTTCTT	CCGTACCACCACTGAATCA
SRAA-747	CCGAGCACAAGACCTACA	GAATGGGCACCGAGATGTT
SRAA-1136	CCACCACGCAGAACAATCAA	CAGCTCAAACAGGGGTCGTA
SRAA-2301	CGCCTTTTTGGAGCAGCA	GAGGAAAAAGCGCAGGTC
SRAA-2294	CCCGCATCAAGAAGATCAAC	GGCATCCAGTGCAGCTTTA
SRAA-2317	AGCGCCGGTGTCAAAGAATA	GTGTCGAGCGGGATGTAA

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