

Aquifer environment selects for microbial species cohorts in sediment and groundwater

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SUPPLEMENTAL MATERIALS

Table S1: Normalized abundance values for the 133 organisms across the 36 metagenomic datasets, with phylum designation for each organism appended. (Separate Excel workbook.)

Table S2: Nucleotide divergence between *rpS3* genes and ribosomal protein encoding scaffolds for organisms identified in both the CSP-I_5m_4 and RBG_5m samples sharing >85% pairwise identity.

Table S3: Geochemical conditions determined from the metagenomic sampling sites.

Figure S1: Concatenated ribosomal protein tree for taxonomic placement of RBG_5m and CSP-I_5m_4 organisms.

Figure S2: Experimental schema for the work reported in this study.

Table S2: Nucleotide divergence between *rpS3* genes and ribosomal protein encoding scaffolds for organisms identified in both the CSP-I_5m_4 and RBG_5m samples sharing >85% pairwise identity.

RBG scaffold names	CSP scaffold names	<i>rpS3</i> % ID	Ribosomal protein scaffold % ID	bp of scaffold overlap
RBG16c_scaffold_21741	CSP2_16ft4sub_bin1_scaffold_25	100	97.93	14,707
RBG16c_scaffold_26	Rifle_16ft_4_minimus_37633	100	99.97	11,989
RBG16c_scaffold_26	Rifle_16ft_4_minimus_24053	100	97.96	11,989
RBG16c_scaffold_2975	Rifle_16ft_4_minimus_38035	100	98.4	28,502
RBG16c_scaffold_2975	Rifle_16ft_4_minimus_24026	100	99.45	18,862
RBG16c_scaffold_4006	Rifle_16ft_4_minimus_1461	100	99.93	5,868
RBG16c_scaffold_43787	Rifle_16ft_4_minimus_2958	100	98.56	10,369
RBG16c_scaffold_613	Rifle_16ft_4_minimus_184	100	99.89	40,742
RBG16c_scaffold_613	Rifle_16ft_4_minimus_36741	100	99.68	4,695
RBG16c_scaffold_966	Rifle_16ft_4_minimus_15147	100	100	5,641
RBG16c_scaffold_5334	Rifle_16ft_4_minimus_3054	99.9	99.65	9,016
RBG16c_scaffold_5757	Rifle_16ft_4_minimus_23719	99.89	99.51	40,080
RBG16c_scaffold_5757	Rifle_16ft_4_minimus_37664	99.89	99.82	56,902
RBG9_49	Rifle_16ft_4_minimus_38099	99.85	99.83	31,132
RBG16c_scaffold_12982	CSP2_16ft4sub_bin8_scaffold_14	99.7	99.85	8,905
RBG16c_scaffold_11510	Rifle_16ft_4_minimus_38982	99.69	99.58	19,991
RBG16c_scaffold_21377	Rifle_16ft_4_minimus_5175	99.55	98.45	2,131
RBG16c_scaffold_21377	Rifle_16ft_4_minimus_39832	99.4	99	8,445
RBG16c_scaffold_17034	Rifle_16ft_4_minimus_24570	98.45	96.64	8,674
RBG16c_scaffold_17034	Rifle_16ft_4_minimus_4190	98.4	96.48	8,021
RBG16c_scaffold_14114	Rifle_16ft_4_minimus_30550	97.78	96.5	3,452
RBG16c_scaffold_2413	Rifle_16ft_4_minimus_15241	97.5	96.03	7,510
RBG16c_scaffold_6059	Rifle_16ft_4_minimus_21028	97.45	99.89	5,496
RBG16c_scaffold_70150	Rifle_16ft_4_minimus_5046	97.14	99.33	13,174
RBG16c_scaffold_105097	Rifle_16ft_4_minimus_30456	96.68	91.05	4,515
RBG16c_scaffold_105097	Rifle_16ft_4_minimus_1560	92.2	85.94	13,125
RBG16c_scaffold_8450	Rifle_16ft_4_minimus_37862	91.16	87.76	23,391
RBG16c_scaffold_291	Rifle_16ft_4_minimus_20697	88.59	85.9	5,440
RBG16c_scaffold_15232	Rifle_16ft_4_minimus_1560	87.55	74.24	15,960
RBG16c_scaffold_2679	Rifle_16ft_4_minimus_23719	87.18	78.9	34,358
RBG16c_scaffold_2679	Rifle_16ft_4_minimus_37664	87.18	78.9	38,357
RBG16c_scaffold_15232	Rifle_16ft_4_minimus_30456	87.15	74.88	5,398
RBG16c_scaffold_10561	Rifle_16ft_4_minimus_30456	86.38	79.9	5,429
RBG16c_scaffold_5757	Rifle_16ft_4_minimus_34155	86.31	75.35	9,361
RBG16c_scaffold_388	Rifle_16ft_4_minimus_10062	86.28	78.53	14,536

continued

RBG scaffold names	CSP scaffold names	<i>rpS3</i> % ID	Ribosomal protein scaffold % ID	bp of scaffold overlap
RBG16c_scaffold_5757	Rifle_16ft_4_minimus_309	86.14	76.52	29,871
RBG16c_scaffold_936	Rifle_16ft_4_minimus_9892	85.88	83.21	13,017
RBG16c_scaffold_5757	Rifle_16ft_4_minimus_39	85.84	73.35	36,639
RBG16c_scaffold_5930	Rifle_16ft_4_minimus_34155	85.66	72.66	9,707
RBG16c_scaffold_5757	Rifle_16ft_4_minimus_37884	85.64	76.25	24,428
RBG16c_scaffold_5757	Rifle_16ft_4_minimus_25120	85.64	77.28	13,816
RBG16c_scaffold_10561	Rifle_16ft_4_minimus_1560	85.61	78.17	14,329
RBG16c_scaffold_39454	Rifle_16ft_4_minimus_30456	85.52	77.85	5,478
RBG16c_scaffold_2576	Rifle_16ft_4_minimus_1560	85.39	73.5	14,327
RBG16c_scaffold_23587	Rifle_16ft_4_minimus_2010	85.3	84.04	9,646
RBG16c_scaffold_388	Rifle_16ft_4_minimus_37664	85	72.33	12,367
RBG16c_scaffold_388	Rifle_16ft_4_minimus_23719	85	71.28	20,710

Table S3: Geochemical conditions determined from the metagenomic sampling sites. All geochemical analyses are based on groundwater sampling, where the closest available date of sampling to when the metagenomic samples were taken is reported. Geochemical species measurements are in mg/L except where otherwise indicated, and were determined as described previously (phosphate, N'Guessan *et al.*, 2010; nitrogen species, Mouser *et al.*, 2009; others, Williams *et al.*, 2011). EC = electrical conductance, DO = dissolved oxygen, ND = not determined.

DATASET	CSP-I	CSP-II/CSP_GW	RBG	AAC1	GW2011
Metagenomic sampling date	07/20/2011	03/28/2013 (sed), 06/03/2013 & 07/07/2013 (GW)	07/2007	07/2010	07/2011
Geochemical sampling date	09/12/2011	06/12/2012	07/18/2008	07/30/2010	07/11/2011
Well name	FP-101	FP-101	D-04	CD-01	CD-01
pH	7.11	7.56	ND	ND	7.25
EC (uS/cm)	2579	3250	ND	ND	2639
DO	0.784	0.329	ND	ND	ND
Fe²⁺	0.01	0.25	3.13	ND	1.68
Cl⁻	203.13	ND	163.4	182.9	188.2
NO₃⁻	6.4	4.3	0.19	ND	ND
NO₂⁻	0.144	0.137	0.02	ND	ND
NH₄⁺	0.03	0.00	1.95	ND	ND
PO₄³⁻	0.16	0.15	0.11	ND	ND
SO₄²⁻	816.5	ND	838.6	782.9	816.5

REFERENCES

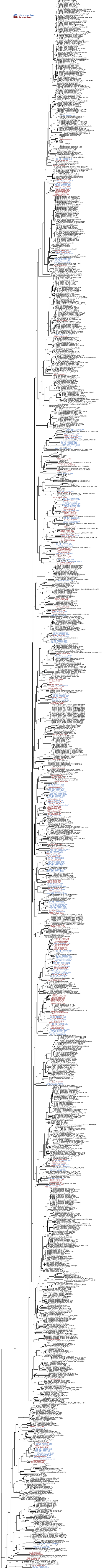
Mouser PJ, N'Guessan AL, Elifantz H, Holmes DE, Williams KH, Wilkins MJ, *et al.* (2009). Influence of heterogeneous ammonium availability on bacterial community structure and the expression of nitrogen fixation and ammonium transporter genes during in situ bioremediation of uranium-contaminated groundwater. *Environ Sci Technol* **43**:4386–92.

N'Guessan AL, Elifantz H, Nevin KP, Mouser PJ, Methé B, Woodard TL, *et al.* (2010). Molecular analysis of phosphate limitation in Geobacteraceae during the bioremediation of a uranium-contaminated aquifer. *ISME J* **4**:253–66.

Williams KH, Long PE, Davis JA, Wilkins MJ, N'Guessan AL, Steefel CI, *et al.* (2011). Acetate availability and its influence on sustainable bioremediation of Uranium-contaminated groundwater. *Geomicrobiol J* **28**:519–539.

Figure S1 (next page): Concatenated ribosomal protein tree including CSP-I_5m_4 (blue) and RBG_5m (red) organisms. Ribosomal proteins utilized were RpL2, 3, 4, 5, 6, 14, 15, 16, 18, 22, 24 and RpS3, 8, 10, 17, 19. For a scaffold to be included, a minimum of 8 of the 16 genes was required. Each individual gene set was aligned using MUSCLE version 3.8.31 and then manually curated to remove end gaps and ambiguously aligned regions. The curated alignments were concatenated to form a 16-gene, 1,319 taxa, 2,304-position alignment. A maximum likelihood phylogeny for the concatenated alignment was conducted using RAxML under the LG+ α + γ model of evolution and with 100 bootstrap replicates. Bootstrap values greater than 50 are displayed on the tree nodes.

CSP_15m_4 organisms



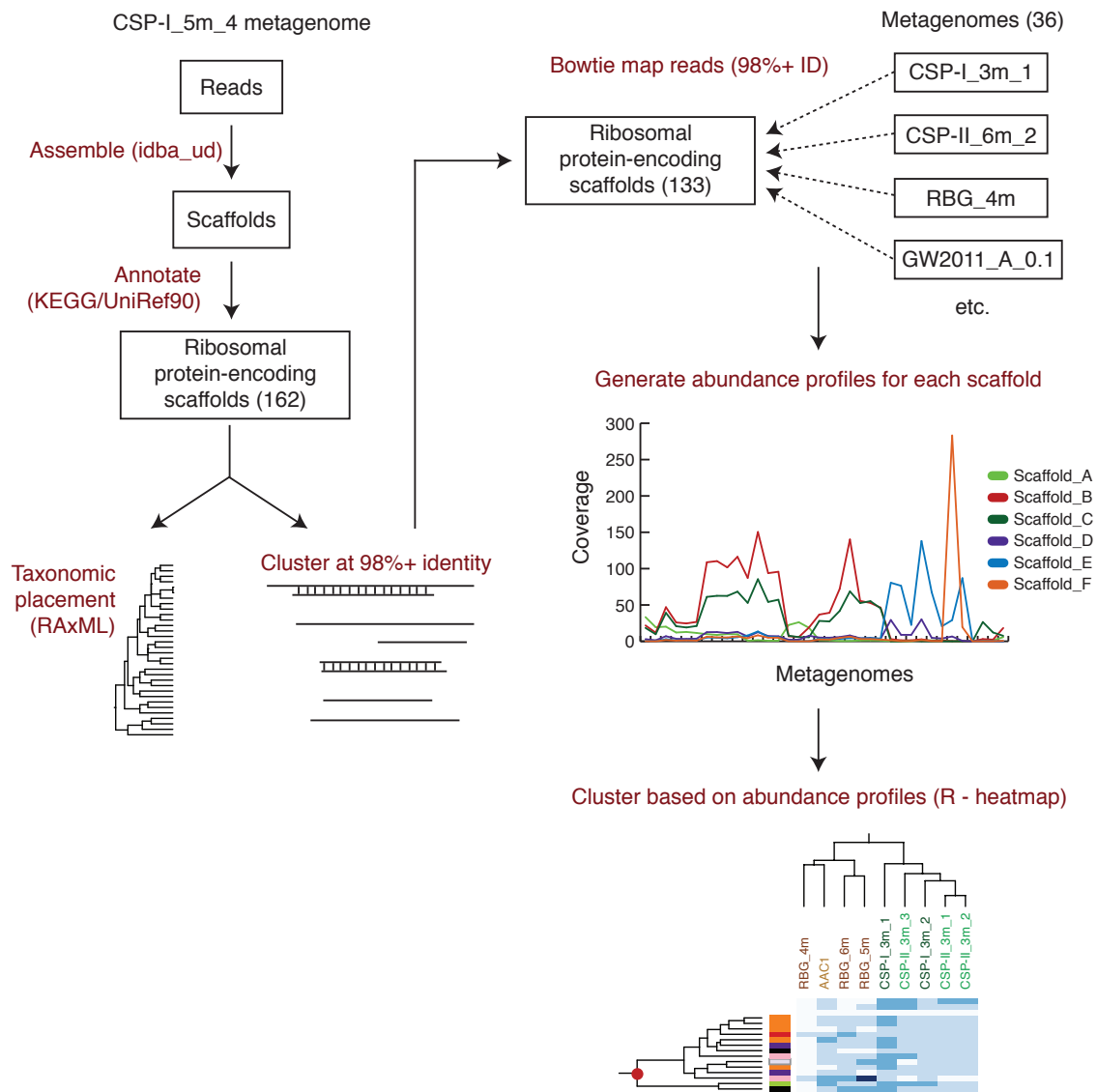


Figure S2: Experimental schema for the work reported in this study.