

High diversity of *Rhodobacterales* in the subarctic North Atlantic Ocean and gene transfer agent protein expression in isolated strains

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Supplement 1. Supplementary information: Tables S1 and S2; Figs. S1, S2 and S3

Table S1. Accession numbers for sequences used in analyses

Species/strain/sequence	Major capsid protein (GTA <i>g5</i>)	16S rDNA
<i>Brucella abortus</i>	YP_221349	
<i>Dinoroseobacter shibae</i> DFL 12	YP_001533511	NC_009952
<i>Loktanella vestfoldensis</i> SKA53	ZP_01004657	AAMS00000000
<i>Oceanibulbus indolifex</i> HEL-45	ZP_02154026	AJ550939
<i>Oceanicola granulosus</i> HTCC2516	ZP_01156865	AY424896
<i>Oceanicola batsensis</i> HTCC2597	AY424898	ZP_01001458
<i>Jannaschia</i> sp. CCS1	CP000264	ABD54552
<i>Phaeobacter gallaeciensis</i> BS107	ZP_02144071	NR_027609.1
<i>Roseobacter denitrificans</i> OCh 114	YP_683229	M59063
<i>Roseobacter litoralis</i> Och 149	ZP_02138791	ABIG01000001
<i>Roseobacter</i> sp. SK209-2-6	ZP_01755050	AAYC01000001.1
<i>Roseobacter</i> sp. AzwK-3b	ZP_01902399	DQ223017.1
<i>Loktanella</i> sp. CCS2	ZP_01751692	AAYB00000000
<i>Roseobacter</i> sp. MED193	ZP_01057324	AANB00000000
<i>Roseovarius nubinihibens</i> ISM	ZP_00959192	AALY00000000
<i>Roseovarius</i> sp. 217	ZP_01036483	AAMV00000000
<i>Roseovarius</i> sp. HTCC2601	ZP_01445424	AATQ00000000
<i>Roseovarius</i> sp. TM1035	ZP_01880957	AAMV00000000
<i>Sagittula stellata</i> E-37	AAYA00000000	AAYA00000000
<i>Ruegeria pomeroyi</i> DSS-3	YP_167486	CP000031
<i>Ruegeria</i> sp. TM1040	YP_613056	CP000377
<i>Sulfitobacter</i> sp. NAS-14.1	ZP_00963032	AALZ01000006.1
<i>Sulfitobacter</i> sp. EE-36	ZP_00955637	AF007254
<i>Phaeobacter</i> sp. Y4I	YP_002695122	NW_002475507
<i>Octadecabacter antarcticus</i> 307	YP_002676353	U14583
<i>Roseobacter</i> sp. GAI101	YP_002698783	AF007257
<i>Rhodobacter sphaeroides</i> 2.4.1	YP_001167284	CP000143
<i>Rhodobacter capsulatus</i>	AF181080	D16427
<i>Paracoccus denitrificans</i> PD1222	YP_916678	CP000489
<i>Citricella</i> sp. SE45	ACNW01000090	AF388308
Rhodobacterales bacterium HTCC2150	ZP_01741236	AAXZ00000000
DMLB-w4.2	GQ433713	GQ433708
DMLB-w6	GQ433711	GQ433707
DMLB-w8	GQ433712	GQ433706
DMLB-w12.1	GQ433715	GQ433710
DMLB-w13	GQ433714	GQ433709
CB1005	FJ869049	FJ869041
CB1006	FJ869050	FJ869042

Supplement 1 (continued)

Table S1 (continued)

Species/strain/sequence	Major capsid protein (GTA <i>g5</i>)	16S rDNA
CB1023	FJ869052	FJ869044
CB1024	FJ869051	FJ869043
CB1040	FJ869053	FJ869045
CB1049	FJ869054	FJ869046
CB1051	FJ869055	FJ869047
CB1079	FJ869056	FJ869048
CBg501	EU929030	
CBg502	EU929031	
CBg503	EU929032	
CBg504	EU929033	
CBg505	EU929034	
CBg507	EU929036	
CBg508	EU929037	
CBg509	EU929038	
CBg510	EU929039	
CBg512	EU929041	
CBg513	EU929042	
CBg514	EU929043	
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CBg518	EU929047	
CBg519	EU929048	
CBg520	EU929049	
CBg521	EU929050	
CBg522	EU929051	
CBg523	EU929052	
CBg524	EU929053	
CBg525	EU929054	
CBg526	EU929055	
<i>Escherichia coli</i> phage HK97	AF069529	

Table S2. Estimation of gene transfer agent (GTA) capsid protein size based on genome sequence information and western blots

Species	Predicted precursor capsid protein size (kDa)	Predicted mature capsid protein size (kDa)	Estimated size on western blot (kDa)
<i>Rhodobacter capsulatus</i> SB1003	42.2	31.4	32
<i>Ruegeria pomeroyi</i> DSS-3	42.0	31.3	34.5

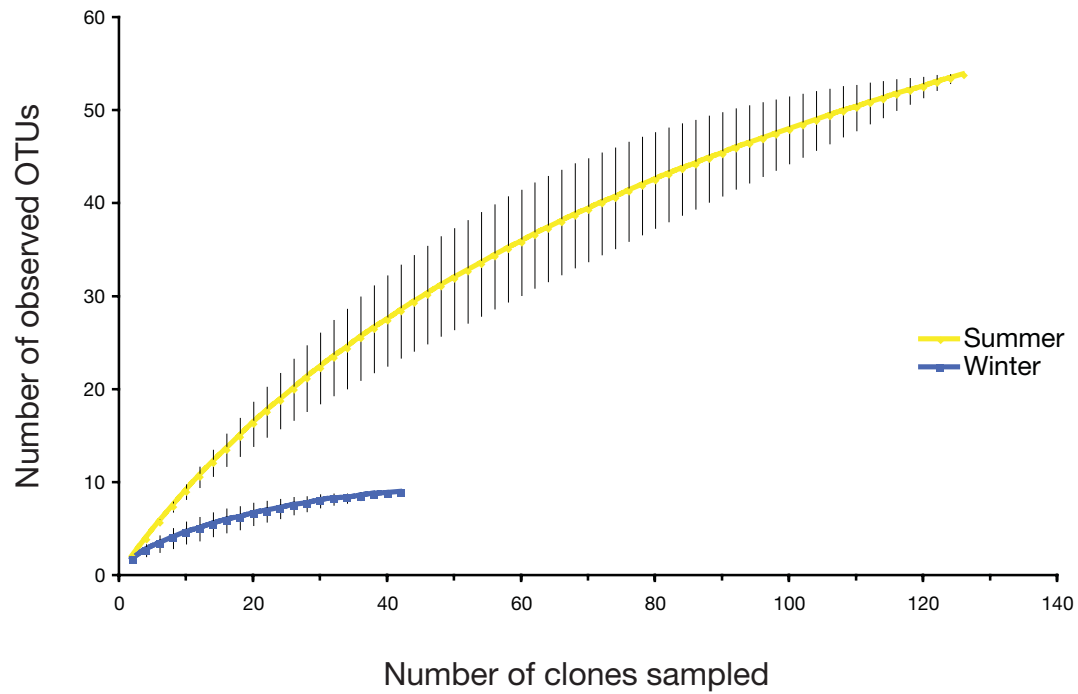


Fig. S1. Rarefaction analyses of clone library sequence discovery. Summer sample is in yellow and winter in blue. Error bars show the 95% confidence intervals for the expected number of operational taxonomic units (OTUs). Analyses were conducted using Analytical Rarefaction v1.4 (www.uga.edu/strata/software/Software.html)

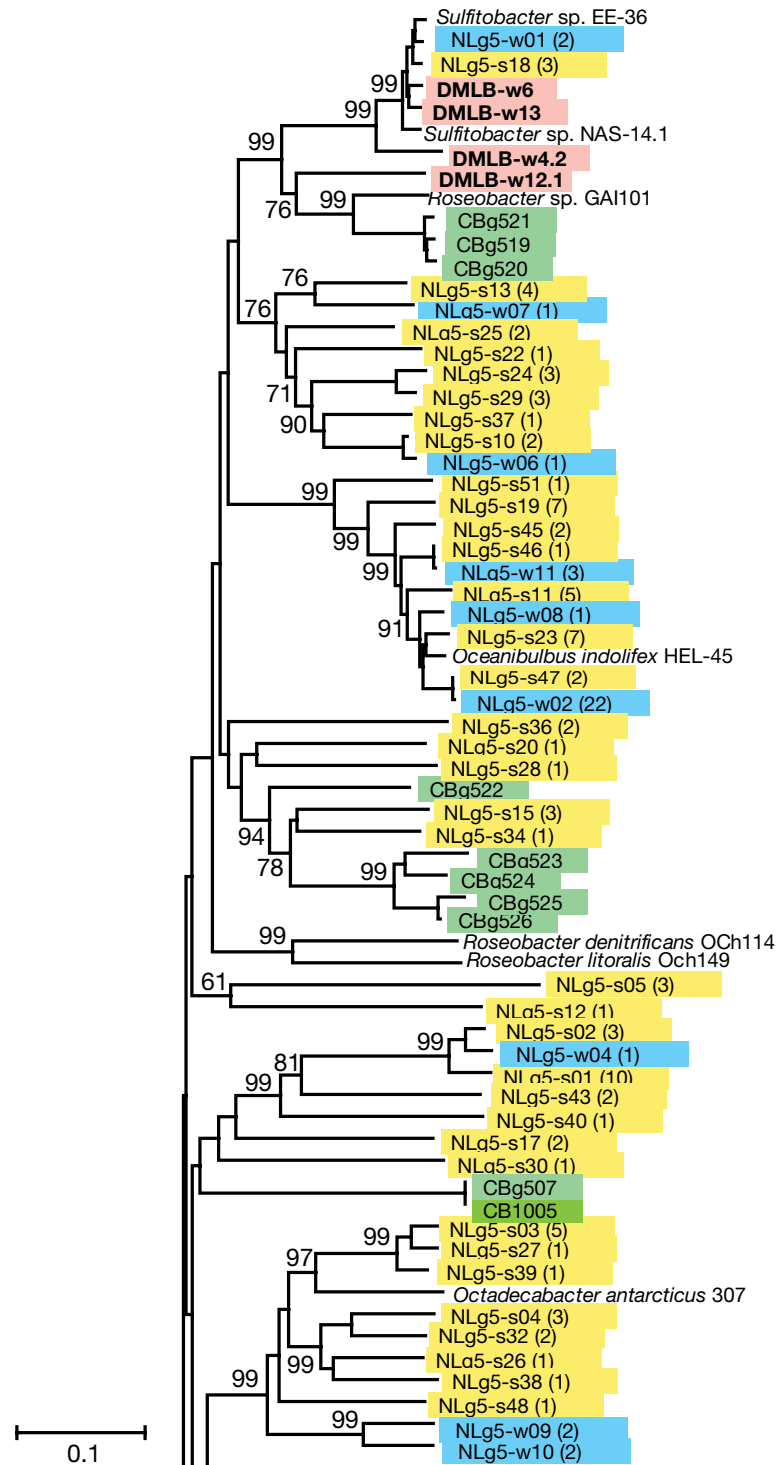


Fig. S2. Phylogenetic tree of gene transfer agent (GTA) capsid gene sequences. This is the same tree as presented in Fig. 1 in the main article, with all operational taxonomic units (OTUs) shown as individual branches. Sequences amplified from the western North Atlantic microbial communities in this study are represented by yellow boxes for sequences from July and blue boxes for sequences from December. Isolates identified in this study (DMLB-w4.2, DMLB-w6, DMLB-w8, DMLB-w12, and DMLB-w13) are highlighted in pink. Dark green boxes represent sequences from clones and light green boxes represent strains from Chesapeake Bay. Numbers in brackets beside the NL sequence identifiers represent the number of clones that fall in each OTU. The neighbor-joining tree is based on the aligned nucleotide sequences corresponding to the *g5* amplicon (747 positions including gaps). Bootstrap values (percentages based on 10 000 replicates) are shown for some branches. Scale bar indicates the number of substitutions per site. The *g5* sequence from *Brucella abortus* was used as the outgroup for the tree (removed for simplicity). Accession numbers for previously published sequences are in Table S1

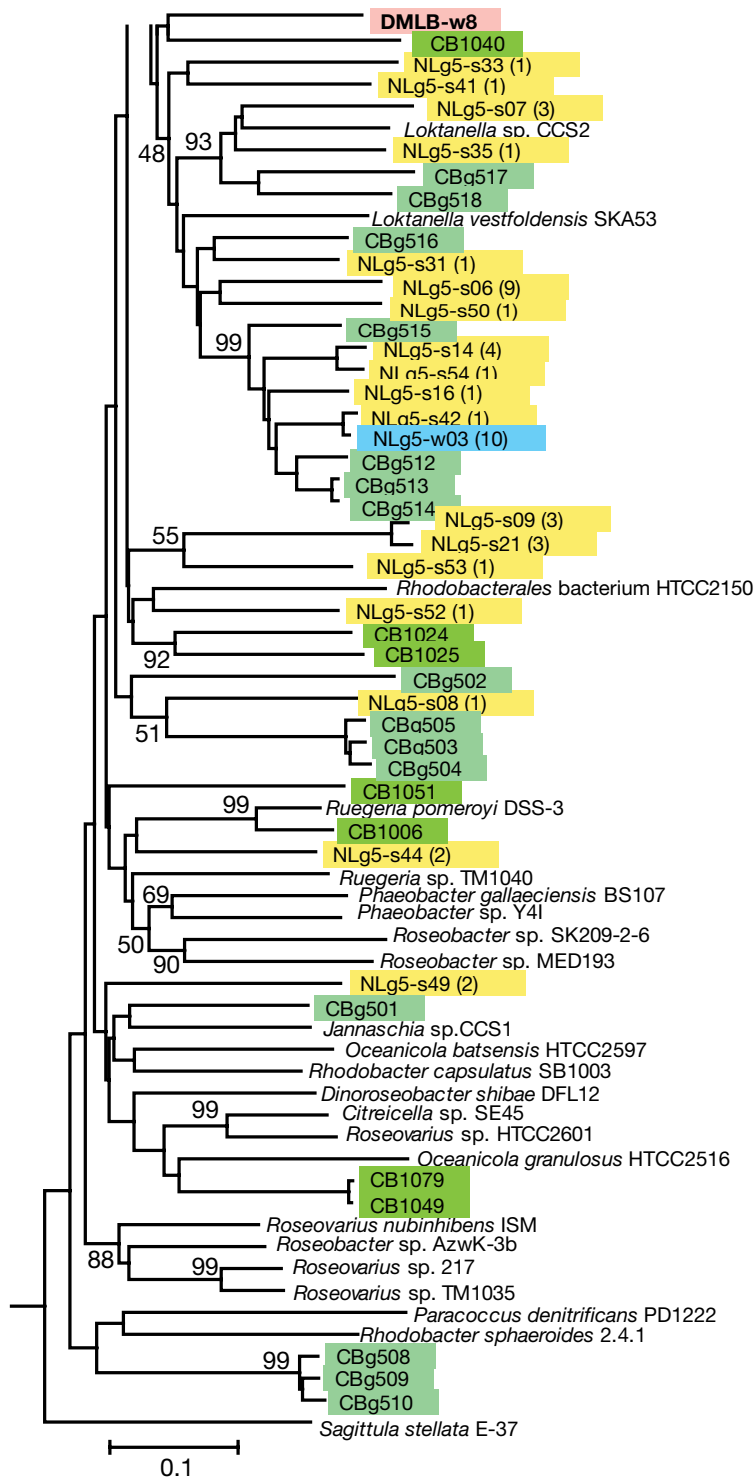


Fig. S2 (continued)

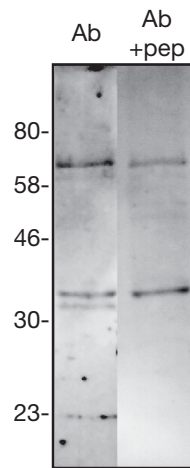


Fig. S3. Peptide neutralization of anti-GTA capsid protein antibodies to identify specific protein detection. A side-by-side comparison of western blot results with cells from strain DMLB-w6 is shown. The membrane was incubated with the anti-GTA capsid protein antibody (Ab) and the pre-neutralized antibody (Ab+pep)