

Supporting Information

for

Isotopically labeled sulfur compounds and synthetic selenium and tellurium analogues to study sulfur metabolism in marine bacteria

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Tables with strains, primers and the full results of the headspace analyses

Table S1: Wild type and mutant strains investigated in this study.

Strain	Genotype	Locus Tag ^a	Source
<i>Phaeobacter gallaeciensis</i>			
DSM 17395	wild type		[1]
CZ01	<i>dmdA::kan</i>	PGA1_262p01830	This work
WP45	<i>cysI::EZTn5</i>	PGA1_c20760	[1]
WP73	<i>sat/cysC::EZTn5</i>	PGA1_c24800	[1]
262 kb mutant	DSM 17395 Δ 262 kb		[2]
<i>Ruegeria pomeroyi</i>			
DSS-3 (= DSM 15171 ^T)	wild type		[3]
J471	<i>dmdA::pBIO1870</i>	SPO1913	[4]
J474	<i>dddQ::pBIO1869</i>	SPO1596	[5]
<i>Roseobacter denitrificans</i>			
OCh 114 (= DSM 7001 ^T)	wild type		[6]
<i>Oceanibulbus indolifex</i>			
HEL-45 (= DSM 14862 ^T)	wild type		[7]
<i>Dinoroseobacter shibae</i>			
DFL-12 (= DSM 16493 ^T)	wild type		[8]
<i>Labrenzia alexandrii</i>			
DFL-11 (= DSM 17067 ^T)	wild type		[9]
<i>Hoeflea phototrophica</i>			
DFL-43 (= DSM 17068 ^T)	wild type		[10]
<i>Roseovarius mucosus</i>			
DFL-24 (= DSM 17069 ^T)	wild type		[11]

^a Locus tags of the targeted genes.

Table S2: Oligonucleotide primers used in this study.

Primer	Sequence
Cl1f	ATTGGA ACTGGCAAGTGCGGC
Cl1r	AACTTCCATCGTGACTATGTCAGC
nptII-f	CCTTTAGTGAGGGTTAATTGCGC
nptII-r	GGATGAATGTCAGCTACTGG

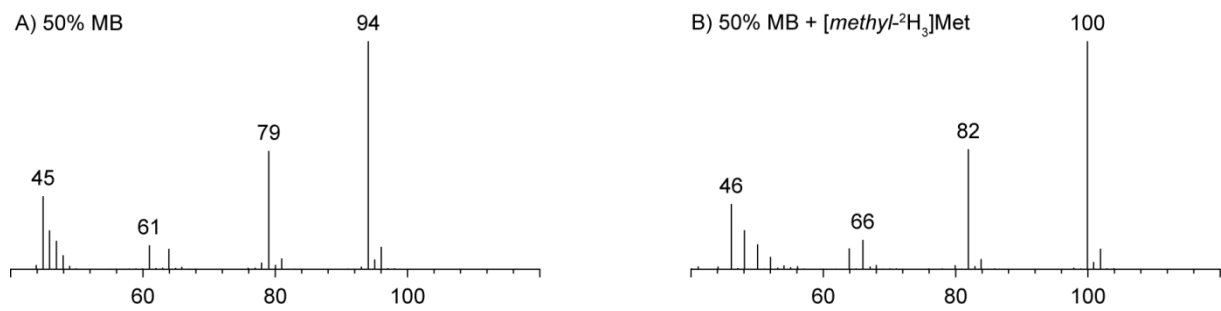


Figure S1: Incorporation of $[methyl-^2H_3]$ methionine into dimethyl disulfide (1) by *Phaeobacter gallaeciensis*.

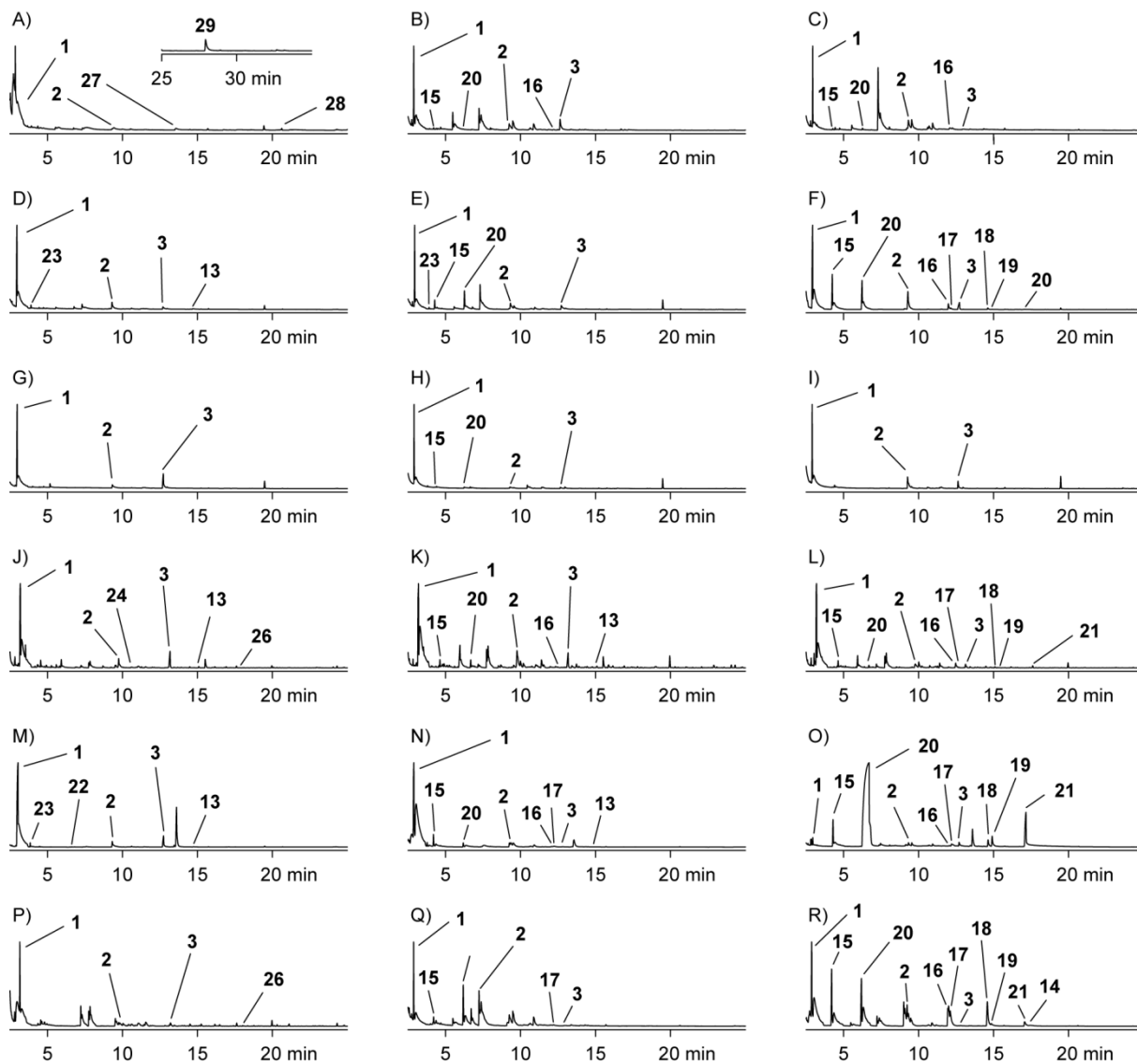


Figure S2: Chromatograms of headspace extracts from *R. denitrificans* DSM 7001 grown on A) 50% MB2216, B) 50% MB2216 + 1 mmol/l Na_2SeO_4 , C) 50% MB2216 + 1 mmol/l Na_2SeO_3 , from *O. indolifex* DSM 14862 grown on D) 50% MB2216, E) 50% MB2216 + 1 mmol/l Na_2SeO_4 , F) 50% MB2216 + 1 mmol/l Na_2SeO_3 , from *D. shibae* DSM 16493 grown on G) 50% MB2216, H) 50% MB2216 + 1 mmol/l Na_2SeO_4 , I) 50% MB2216 + 1 mmol/l Na_2SeO_3 , from *L. alexandrii* DFL-11 grown on J) 50% MB2216, K) 50% MB2216 + 1 mmol/l Na_2SeO_4 , L) 50% MB2216 + 1 mmol/l Na_2SeO_3 , from *H. phototrophica* DFL-43 grown on M) 50% MB2216, N) 50% MB2216 + 1 mmol/l Na_2SeO_4 , O) 50% MB2216 + 1 mmol/l Na_2SeO_3 , and from *R. mucosus* DFL-24 grown on P) 50% MB2216, Q) 50% MB2216 + 1 mmol/l Na_2SeO_4 , R) 50% MB2216 + 1 mmol/l Na_2SeO_3 .

Table S3: Feeding of selenate and selenite to marine bacteria.

Compound ^a	<i>p</i>	DSM 17395			DSM 7001			DSM 14862			DSM 16493			DFL-11			DFL-43			DFL-24		
		50% MB2216	+1 mmol/l Na ₂ SeO ₄	+1 mmol/l Na ₂ SeO ₃	50% MB2216	+1 mmol/l Na ₂ SeO ₄	+1 mmol/l Na ₂ SeO ₃	50% MB2216	+1 mmol/l Na ₂ SeO ₄	+1 mmol/l Na ₂ SeO ₃	50% MB2216	+1 mmol/l Na ₂ SeO ₄	+1 mmol/l Na ₂ SeO ₃	50% MB2216	+1 mmol/l Na ₂ SeO ₄	+1 mmol/l Na ₂ SeO ₃	50% MB2216	+1 mmol/l Na ₂ SeO ₄	+1 mmol/l Na ₂ SeO ₃	50% MB2216	+1 mmol/l Na ₂ SeO ₄	+1 mmol/l Na ₂ SeO ₃
Dimethyl disulfide (1)	768	xxx	xxx	xxx	xxx	xxx	xxx	xxx	xx	xxx	xxx	xxx	xxx	xxx	xxx	xxx	xxx	x	xxx	xxx	xxx	
S-Methyl propanethioate (23)	802						x	x								x						
Dimethyl selenyl sulfide (15)	814		xxx	xxx		x	x		x	xxx		x		x	x		xx	xx		xx	xxx	
Methylpyrazine	825				x	x	x						x	x	x	x				x		
2-(Methoxymethyl)furan	827					x	x							x	x		x	x		x	x	
Furfural	835					x							x	x	x							
2-Furanmethanol	860		xx		xx	xxx	xx						xx	xx	xx					xx	x	
Dimethyl diselenide (20)	884		xx	xx		x	x		xx	xxx		xx		x	x		xx	xxx		xxx	xxx	
Bis(methylthio)methane (22)	885															x						
Cyclohexanone	896				x	x							x	x		x	x		xxx	xxx	x	
2,5-Dimethylpyrazine	911		xxx		xx	xxx	xxx						x	x	x	x	x	x	xx	xx	xx	
2-Acetylfuran	913				xx	xxx	xx						xx	xx	xx	x	xx	x	xxx	xxx	xx	
Benzaldehyde	961					x	x			x			x		x			x	xx	x	xx	
Dimethyl trisulfide (2)	967	x	xx	xxx	xx	xx	xx	xx	x	xxx	xx	xx	xxx	xx	xx	xx	xx	xx	x	xx	xx	xx
4-Methylthio-2-butanone (24)	990												x									
2-Acetylthiazole	1016					x	x						x	x	x	x	x		xx	x	x	
Benzyl alcohol	1033												x	x	x							
Bis(methylthio)selenide (16)	1045		x	x		x	x			xx			x	x		x	x				xx	
Methyl methylseleno disulfide (17)	1051		x	x						xx			x		x		x	x		x	xx	
S-Methyl methanethiosulfonate (3)	1063	xx	xx	x		xx	x	xx	xx	x	xx	xx	xx	xx	x	xx	x	x	x	x	x	
Acetophenone	1065														x				x			
1,2,4-Trithiolane (27)	1088				x																	
1-Undecene	1091															xxx	xx	xx				
Methyl benzoate	1093												x									
Nonanal	1103					x							x		x				x		x	
2-Phenylethanol	1111												x	x	x							
Methyl methylthio diselenide (18)	1119		x							x					x			x			xx	

Methyl (methylthio)methyl disulfide (13)	1123				x			x	x		x	x		
Phenylacetone	1127							x		x			x	
Bis(methylseleno)sulfide (19)	1127						x			x			x	x
Methyl nicotinate	1137							xx	x	x				
Ethyl benzoate	1170	xx	x			xx	x							
Dimethyl triselenide (21)	1203									x		xx		x
Decanal	1203			x	x			x		x			x	
Dimethyl tetrasulfide (14)	1213		x											x
Benzothiazol (26)	1220							x					x	
Ethyl phenylacetate	1244									x			x	
1,2,4,5-Tetrathiane (28)	1350				x									
Lenthionine (29)	1624				xxx									

^a Known contaminants such as plasticizers and compounds arising from the agar medium are not mentioned. Relative amounts of compounds were given as follows: x: 0-2%, xx: 2-8%, xxx: >8% of total area in GC. ^b Linear retention index on a HP-5MS fused silicic capillary GC column determined from a homologous series of *n*-alkanes.

References

1. Thole, S.; Kalhoefer, D.; Voget, S.; Berger, M.; Engelhardt, T.; Liesegang, H.; Wollherr, A.; Kjelleberg, S.; Daniel, R.; Simon, M.; Thomas, T.; Brinkhoff, T. *ISME J.* **2012**, *6*, 2229–2244.
2. Petersen, J.; Brinkmann, H.; Berger, M.; Brinkhoff, T.; Päucker, O.; Pradella, S. *Mol Biol Evol.* **2011**, *28*, 1229–1240.
3. González, J. M.; Covert, J. S.; Whitman, W. B.; Henriksen, J. R.; Mayer, F.; Scharf, B.; Schmitt, R.; Buchan, A.; Fuhrman, J. A.; Keine, R. P.; Moran, M. A. *Int. J. Syst. Evol. Microbiol.* **2003**, *53*, 1261–1269.
4. Todd, J. D.; Curson, A. R. J.; Sullivan, M. J.; Kirkwood, M.; Johnston, A. W. B. *PLOS ONE* **2012**, *7*, e35947.
5. Todd, J. D.; Curson, A. R. J.; Kirkwood, M.; Sullivan, M. J.; Green, R. T.; Johnston, A. W. B. *Environ. Microbiol.* **2011**, *13*, 427–438.
6. Shiba, T. *Syst. Appl. Microbiol.* **1991**, *14*, 140–145.
7. Wagner-Döbler, I.; Rheims, H.; Felske, A.; El-Ghezal, A.; Flade-Schröder, D.; Laatsch, H.; Lang, S.; Pukall, R.; Tindall, B. J. *Int. J. Syst. Evol. Microbiol.* **2004**, *54*, 1177–1184.
8. Biebl, H.; Allgaier, M.; Tindall, B. J.; Koblizek, M.; Lünsdorf, H.; Pukall, R.; Wagner-Döbler, I. *Int. J. Syst. Evol. Microbiol.* **2005**, *55*, 1089–1096.
9. Biebl, H.; Pukall, R.; Lünsdorf, H.; Schulz S.; Allgaier, M.; Tindal, B. J.; Wagner-Döbler, I. *Int. J. Syst. Evol. Microbiol.* **2007**, *57*, 1095–1107.
10. Biebl, H.; Tindall, B. J.; Lünsdorf, H.; Allgaier, M.; Wagner-Döbler, I. *Int. J. Syst. Evol. Microbiol.* **2006**, *56*, 821–826.
11. Biebl, H.; Allgaier, M.; Lünsdorf, H.; Pukall, R.; Tindall, B. J.; Wagner-Döbler, I. *Int. J. Syst. Evol. Microbiol.* **2005**, *55*, 2377–2383.