

# Supporting Information for

## **Linkages between mineralogy, fluid chemistry and microbial communities within hydrothermal chimneys from the Endeavour Segment, Juan de Fuca Ridge**

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### **Introduction**

This supporting information provides data tables for Mössbauer spectroscopy, microbial community composition and characteristics, microbial diversity index analyses, and most-probable-number of viable cell estimates.

Table S1. Mössbauer parameters at 295 K and 4 K

Parameter	Species	Bastille		Dante		Hot Harold	
Temp (K)		295	4	295	4	295	4†
Isomer Shift (mm/s)						0.40	
Quadrupole Splitting (mm/s)						0.25	
Width (mm/s)	unknown					0.35	
Area (% of total area)						25	
Isomer Shift (mm/s)		0.31	0.41	0.32	0.40	0.35	0.44
Quadrupole Splitting (mm/s)	sulfide	0.61	0.60	0.60	0.61	0.61	0.65
Width (mm/s)		0.39	0.90	0.42	0.40	0.28	0.45*
Area (% of total area)		71	63	78	47	34	-
Isomer Shift (mm/s)						0.41	
Quadrupole Splitting (mm/s)	unknown					1.08	
Width (mm/s)						0.36	
Area (% of total area)						38	
Isomer Shift (mm/s)						1.48	
Quadrupole Splitting (mm/s)	unknown					2.12	
Width (mm/s)						0.23*	
Area (% of total area)						4	
Isomer Shift (mm/s)				0.53	-0.14		
Quadrupole Splitting (mm/s)				-0.01	0.40		
Field (T)	unknown			27.9	22.5		
Width (mm/s)				1.13	0.82		
Area (% of total area)				22	16		
Isomer Shift (mm/s)		0.24	0.37				
Quadrupole Splitting (mm/s)		0.01	0.02				
Field (T)	chalcopyrite	35.6	37.3				
Width (mm/s)		0.34	0.82				
Area (% of total area)		29	37				
Isomer Shift (mm/s)						1.08	
Quadrupole Splitting (mm/s)						-0.53	
Field (T)	unknown					33.4	
Width (mm/s)						0.60*	
Area (% of total area)						15	
Isomer Shift (mm/s)						0.78	
Quadrupole Splitting (mm/s)						-0.18	
Field (T)	unknown					43.0*	
Width (mm/s)						0.60*	
Area (% of total area)						5	
Isomer Shift (mm/s)						0.49	
Quadrupole Splitting (mm/s)						0.12	
Field (T)	unknown					45.5	
Width (mm/s)						0.32	
Area (% of total area)						5	
Isomer Shift (mm/s)						0.50	
Quadrupole Splitting (mm/s)	unknown					-0.09	
Field (T)						49.5	
Width (mm/s)						0.54	

Area (% of total area)	12					
$\chi^2$	542.0	1245.1	777.0	1078.8	448.6	-
$\chi^2_{\text{norm}}$	1.0	2.4	1.5	2.1	0.9	-

\*Indicates parameter held constant to obtain fit

‡Only the central doublet was fit for the 4K Hot Harold spectrum. Other distributions were too poorly resolved to obtain an accurate fit. Relative area and  $\chi^2$  values are not reported because the fit is incomplete.

§The parameters for these phases can be found in Bishop *et al.* [2014], Dyar *et al.* [2006 and 2013], Lane *et al.* [2015], and Pitman *et al.* [2014].

Table S2. Distribution of representative bacterial and archaeal 16S rRNA gene phylotypes in the interior of the actively venting hydrothermal chimneys

	T range (°C)	pH range	Carbon sources	O <sub>2</sub> preference	Phototroph	% identity	Number of clones			Ref.
							Bastille	Dante	Hot Harold	
<b>Bacteria</b>										
<i>Alphaproteobacteria</i>										
<i>Methylobacterium tardum</i>	15-35	N	AU	AE	No	99	0	1	0	21
<i>Rhodospseudomonas julia</i>	25-35	6	FAU	FAN	Yes	93	5	0	0	22
<i>Thalassobacter stenotrophicus</i>	13-37	N	H	AE	No*	94	1	0	0	27
Other <i>Alphaproteobacteria</i>							0	4	1	
<i>Betaproteobacteria</i>										
<i>Acidovorax temperans</i>	37	N	H	AE	No	99	0	1	0	55
<i>Delftia acidovorans</i>	30	N	H	AE	No	99	0	10	0	54
<i>Herbaspirillum huttiense</i> subsp. <i>putei</i>	25-37	N	H	AE	No	99	0	3	0	9
<i>Ralstonia pickettii</i>	35	N	H	FAN	No	99-100	0	5	0	36
<i>Roseateles depolymerans/aquatilis</i>	5-43	5-8	H	AE	No*	90-99	0	19	25	49
Other <i>Betaproteobacteria</i>							0	5	3	
<i>Gammaproteobacteria</i>										
<i>Marinobacter lutaoensis</i>	25-50	5-9	H	AE	No	90	0	1	1	43
<i>Thioalkalivibrio denitrificans/nitratis</i>	up to 47	7.5-10.7	AU	FAN	No	88-92	0	1	3	46
<i>Enterbacter pyrinus</i>	27-36	N	H	FAN	No	97	0	1	0	8
<i>Kangiella koreensis</i>	4-43	5.5-8	H	FAN	No	90-91	2	0	0	59
<i>Alcanivorax hongdengensis</i>	10-42	4-10	H	AE	No	90	0	0	1	56
<i>Endozoicomonas elysicola</i>	4-37	N	H	AE	No	91	0	0	1	23
<i>Acinetobacter junii/calcoaceticus/johnsonii</i>	15-37	N	H	AE	No	97-98	0	6	0	5
<i>Pseudomonas fulva/hibiscicola/panacis</i>	4-37	N	H	AE	No	98-99	0	3	2	51
<i>Leucothrix mucor</i>	15-35	N	H	AE	No	90-91	2	0	1	15
<i>Sedimenticola selenatireducens</i>	28	N	H	AN	No	93	1	0	0	32
<i>Stenotrophomonas maltophila</i>	35	N	H	AE	No	98	0	2	0	33
Other <i>Gammaproteobacteria</i>							0	4	0	

\*Produces bacteriochlorophyll *a*

	T range (°C)	pH range	Carbon sources	O <sub>2</sub> preference	Phototroph	% identity	Number of clones			Ref.
							Bastille	Dante	Hot Harold	
<i>Deltaproteobacteria</i>										
<i>Desulfobulbus mediterraneus</i>	10-30	6.3-8.0	H	AN	No	87-99	15	0	1	38
<i>Desulfobacterium indolicum/anilini</i>	30	7.2	H	AN	No	84-91	8	0	0	3, 40
<i>Desulfocapsa sulfexigens</i>	5-35	6-8.2	AU	AN	No	91	1	0	0	13
<i>Desulfococcus multivorans</i>	35	N	H	AN	No	87	1	0	0	-
<i>Geothermobacter ehrlichii</i>	35-65	5-8	H	AN	No	80-85	3	0	0	20
<i>Desulfuromonas svalbardensis</i>	-2-20	6.5-7.5	H	AN	No	86	1	0	0	52
<i>Desulfonatronum lacustre</i>	37-40	8-10	H	AN	No	81	2	1	0	34
<i>Desulfomonile limimaris</i>	37	N	H	AN	No	92	1	0	0	48
Other <i>Deltaproteobacteria</i>							1	0	0	
<i>Epsilonproteobacteria</i>										
<i>Sulfurovum lithotrophicum</i>	10-40	5-9	AU	FAN	No	90-99	2	1	1	17
<i>Nitratiruptor tergaricus</i>	40-57	5.4-6.9	AU	FAN	No	92-94	0	0	2	31
<i>Nitratifactor salsuginis</i>	28-40	5.6-7.6	AU	FAN	No	90-96	1	1	4	31
<i>Sulfurospirillum halorespirans</i>	25-30	N	H	AN	No	90	1	0	0	26
<i>Hydrogenimonas thermophila</i>	35-65	4.9-7.2	AU	FAN	No	89-91	1	2	0	50
<i>Helicobacter pullorum</i>	37-42	N	H	MAE	No	87	4	0	0	47
<i>Sulfurimonas autotrophica/paralvinellae</i>	10-40	4.5-9	AU	AE	No	88-95	0	1	12	16
<i>Caminibacter profundus</i>	45-65	6.5-7.4	AU	AN	No	93	0	0	1	29
Other <i>Epsilonproteobacteria</i>							1	0	0	
<i>Actinobacteria</i>										
<i>Aquificae</i>										
<i>Desulfurobacterium pacificum</i>	55-85	5.5-7.5	AU	AN	No	96	0	0	2	25
CFB Group										
<i>Alkaliflexus imshenetskii</i>	28-35	7.5-10.2	H	AN	No	85	1	0	0	60
<i>Chlorobium</i> sp.	25	N	AU	AN	Yes	80-83	2	0	0	30
<i>Salinimicrobium catena</i>	15-42	6.5-9	H	AE	No	84-85	2	0	0	58
Other CFB Group							0	0	1	
<i>Chloroflexi</i>										
<i>Anaerolinea thermophila</i>	50-60	6-8	H	AN	No	82	2	0	0	41
<i>Levilinea saccharolytica</i>	25-50	6-7.2	H	AN	No	90	1	0	0	57
<i>Cyanobacteria</i>										
<i>Prochlorococcus marinus</i>			AU	AE	Yes	89	0	1	0	7

	T range (°C)	pH range	Carbon sources	O <sub>2</sub> preference	Phototroph	% identity	Number of clones			Ref.
							Bastille	Dante	Hot Harold	
<i>Deinococcus-Thermus</i>										
<i>Oceanithermus desulfurans</i>	40-68	5.5-8.4	FAU	MAE	No	99	1	0	0	28
<i>Firmicutes</i>										
<i>Clostridium tepidiprofundum</i>	22-60	4-8.5	H	AN	No	80	2	0	0	44
<i>Planctomycetes</i>										
<i>Pirellula baltica/staleyii</i>	28-30	N	H	AE	No	86-87	3	0	0	39
<b>Archaea</b>										
<i>Thermococci</i>										
<i>Thermococcus onnurineus/nautili</i>	63-90	5-9	H	AN	No	86-87	11	0	0	2
<i>Methanococci</i>										
<i>Methanocaldococcus infernus</i>	55-91	5.3-7	AU	AN	No	88	3	0	0	19
<i>Archaeoglobi</i>										
<i>Ferroglobus placidus</i>	65-95	6-8.5	AU	AN	No	80	1	0	0	14
<i>Geoglobus acetivorans</i>	50-85	5-7.5	FAU	AN	No	79	1	0	0	45
<i>Other Euryarchaeota</i>										
<i>Aciduliprofundum boonei</i>	55-75	3.3-5.8	H	AN	No	86	1	0	0	37
<i>Thermoprotei</i>										
<i>Vulcanisaeta distributa</i>	70-92	3.5-5.6	H	AN	No	78-93	3	0	0	18
<i>Hyperthermus butylicus</i>	85-108	7	H	AN	No	92	1	0	0	61
<i>Korarchaeota</i>										
<i>Korarchaeum cryptofilum</i>	85	6.5	H	AN	No	94	2	0	0	12

Abbreviations: N, neutrophile; AE, aerobe; AN, anaerobe; AU, autotroph; FAN, facultative anaerobe; H, heterotroph; MAE, microaerophile.

Table S3. Diversity indices for the Bastille, Dante and Hot Harold hydrothermal chimneys used in this study

	Bastille	Dante	Hot Harold
<b><math>\alpha</math> Diversity</b>			
Chao1 <sup>a</sup>	15.13	18.34	11.20
Shannon index <sup>b</sup>	3.75	3.57	2.84
Observed Species	15.0	16.2	11.0
<b><math>\beta</math> Diversity</b>			
UNiFRac			
Bastille	0.0	0.7336	0.6684
Dante		0.0	0.3715
Hot Harold			0.0
Weighted UNiFRac			
Bastille	0.0	0.5926	0.5634
Dante		0.0	0.1963
Hot Harold			0.0

<sup>a</sup>Chao1 richness estimator [*Chao*, 1987]

<sup>b</sup>Shannon diversity index [*Shannon*, 1948]

Table S4. Most-probable-cell estimates of various hyperthermophilic anaerobes per gram of hydrothermal mineral material. Enrichments were incubated at 90°C

Sample	Most-probable-number estimates		
	Iron reducers	Methanogens <sup>a</sup>	Heterotrophs <sup>a</sup>
Bastille	108	34	3,470
Dante	13	4	511
Hot Harold	106	ND	1,061

<sup>a</sup>Results previously reported in *Ver Eecke et al.* [2012]

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