

Data report: carbon content and isotopic composition of basalts and sediments in North Pond, Expedition 336¹

Kasumi Sakata,^{2,3} Hikaru Yabuta,² Minoru Ikehara,⁴ and Tadashi Kondo²

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Abstract

The uppermost about 500 m of basaltic ocean crust is permeable, and fluid flow is focused in specific areas at the contacts of lava flows or in brecciated zones. In these areas, seawater oxidizes young basaltic crust (younger than 10 Ma), and this interaction affects the microbial ecosystems. Iron cycling, both oxidation and reduction of iron, supports metabolic activity in basalts; however, the microorganisms responsible for Fe oxidation of basalts are not clear. In this study, carbon isotopic analyses of basalts and sediments at North Pond, the western flank of the mid-Atlantic Ridge, were conducted to understand the origin and formation of carbon compounds in relation to possible microbial activity in basaltic crust. Total carbon (TC) contents range approximately from 6 to 11 wt% for whole-sediment samples. Depth profiles of the carbon isotopic compositions ($\delta^{13}\text{C}$ -TC) for sediments (approximately -0.04‰ to $+1.93\text{‰}$) are similar to those of TC. TC (approximately 0.01–0.37 wt%) and total organic carbon (TOC) (approximately 0.01–0.03 wt%) contents for basalts are almost constant with depth, whereas sediment breccias and carbonates contain more carbon than basalts (approximately 3.56–11.9 wt%). The value of $\delta^{13}\text{C}$ -TC for basalts ranges approximately from -21.8‰ to $+2.69\text{‰}$. Sediment breccias and carbonates have larger $\delta^{13}\text{C}$ -TC values, approximately from -18.6‰ to $+2.82\text{‰}$. The value of $\delta^{13}\text{C}$ -TOC for hard rocks is lower at greater depths. The value of $\delta^{13}\text{C}$ -kerogen is slightly smaller than that of $\delta^{13}\text{C}$ -TOC.

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²Department of Earth and Space Science, Graduate School of Science, Osaka University, 1-1 Machikaneyama, Toyonaka, Osaka 560-0043, Japan. Correspondence author. ksakata@ess.sci.osaka-u.ac.jp

³Present address: Toyonaka High School, 2-5-12 Uenonishi, Toyonaka, Osaka 560-0011, Japan.

⁴Center for Advanced Marine Core Research, Kochi University, Nankoku 783-8502, Japan.

Introduction

The majority of habitable environments on the Earth are dark biosphere separated from sunlight. About one-third of all biomass and about three-fourths of all prokaryotic cells on the Earth are in the sediments buried in the oceanic subsurface (Whitman et al., 1998). Kallmeyer et al. (2012) report that total cell abundance in subseafloor sediments (2.9×10^{29}) is about 92% less than the previous estimated values by Whitman et al. (1998) (35.5×10^{29}). During this decade, basaltic ocean crust has been focused on as a new deep submarine biosphere (Bach and Edwards, 2003). To date, Juan de Fuca Ridge in the eastern Pacific has been a primary study site for basaltic ocean crust because water-rock reactions in basaltic crusts can be clearly observed there (e.g., Wheat et al.,



2000; Fisher et al., 2003). In the Juan de Fuca basaltic oceanic crust, iron cycling, both oxidation and reduction, supports metabolic activity in basalts; however, the microorganisms responsible for Fe oxidation in basalts are not identified (Orcutt et al., 2011). From September to November 2011, Integrated Ocean Drilling Program (IODP) Expedition 336 explored the deep biosphere at North Pond, on the western flank of the Mid-Atlantic Ridge ([Expedition 336 summary](#) [Expedition 336 Scientists, 2012]). The North Pond sedimentary environment is oligotrophic and low temperature (10°–15°C) and allows fast seawater circulation (2–3 m/h) (Langseth et al., 1984). Therefore, the site is a good target for comparison to Juan de Fuca Ridge, which has higher temperatures (~60°C) and slower seawater circulation (approximately 1400–1500 kg/day) (Wheat et al., 2003). However, North Pond sediment contains low organic carbon contents (<0.3 wt%) (Ferdelman et al., 2011), which makes life detection at this site challenging. In this study, we analyze organic carbon and carbon isotopes of basalts and sediments collected from North Pond to understand the origin and formation of carbon compounds in relation to possible microbial activity in the basaltic crust.

Methods

Preparation of samples for elemental and isotopic analysis

HCl, HF, and methanol were obtained from Wako Pure Chemical Industries. Procedures for preparing samples are shown in Figure [F1](#). Sediment samples were dried at 45°C. Basalt samples were trimmed by at least 5.0 mm to prevent surface contamination. The trimmed, inner basalt samples were ground into powder. Carbonates and sediment breccias were prepared for analysis in the same way as the basalts. To remove carbonates, 10 g of powdered sample was washed with 6 M HCl at least 20 times, until the yellow iron contamination in the supernatant solution was clear. A group of these washed samples was then dried at around 70°C. Recovered acid-insoluble matter was washed with deionized water (“milli-Q” = 18.2 MΩ) until the pH of the supernatant solution was neutral. Finally, the neutralized insoluble matter was washed with methanol and dried at around 70°C. Sediment breccias and carbonates were collected between basaltic layers at 163.04 and 163.70 meters below seafloor (mbsf) in Hole U1382A and from 117.15, 145.93, and 154.04 mbsf in Hole U1383C. These samples were prepared the same way as the basalt samples. To extract kerogen, splits of the 6 M HCl-washed basalt samples were further washed with 1 M HCl/9 M HF. HCl/HF demineraliza-

tion treatment is a conventional technique to extract kerogen (Durand and Nicaise, 1980). The acid-insoluble residue was rinsed with milli-Q water and methanol and then dried at around 70°C.

All samples were analyzed twice by elemental analyzer (Flash EA 1112; Thermo Fisher Scientific K.K.)/isotope ratio mass spectrometer (Delta Plus Advantage; Thermo Fisher Scientific K.K.) (EA/IRMS) at Kochi Core Center, Japan. We analyzed about 10 mg of sediment samples and 50 mg of basalt samples to measure carbon content and $\delta^{13}\text{C}$ of total carbon (TC). Additionally, we analyzed about 50–100 mg of the organic (6 M HCl treated) and kerogen (1 M HCl/9 M HF treated) basalt samples to measure carbon contents and $\delta^{13}\text{C}$. All samples were analyzed in Sn capsules. Elemental and isotopic compositions were calibrated relative to sulfanilamide and L-alanine, respectively. We also analyzed only Sn cups for blank analysis. Carbon was not detected in only Sn cups. The analytical error for determination of carbon contents was within 0.005 wt%. The precision of $\delta^{13}\text{C}$ was better than 0.1‰.

Results

Sediments

TC contents range from 6 to ~11 wt% for whole-sediment samples. Depth profiles of TC were similar at the three sites (Holes U1382B, U1383D, and U1384A) (Table [T1](#); Fig. [F2](#)). These values were consistent with TC data for North Pond sediments reported by Bode (1979). From 0 to ~40 mbsf, TC increases with depth, with maximum values at approximately 20–40 mbsf. From 40 to ~90 mbsf, TC decreases with depth. $\delta^{13}\text{C}$ -TC ranges from -0.04‰ to about +1.93‰ for whole-sediment samples (Table [T1](#); Fig. [F2](#)). Both $\delta^{13}\text{C}$ -TC values and TC contents change at a certain depth (Table [T1](#); Fig. [F2](#)).

Basalts

TC (approximately 0.01–0.37 wt%) and total organic carbon (TOC) (approximately 0.01–0.03 wt%) values of basalts are almost constant over the depth interval analyzed (Table [T2](#); Fig. [F3](#)), whereas sediment breccias and carbonates contain more carbon than basalts (approximately 3.56–11.9 wt%). The carbon isotopic composition of TC ($\delta^{13}\text{C}$ -TC) of basalt samples ranges approximately from -21.8‰ to +2.69‰. Sediment breccias and carbonates have slightly higher $\delta^{13}\text{C}$ -TC, at approximately -18.6‰ to +2.82‰. The carbon isotopic compositions of TOC ($\delta^{13}\text{C}$ -TOC) of the corresponding samples show constant values (approximately -25‰) over the depth interval analyzed. The carbon isotopic compositions of kerogen

($\delta^{13}\text{C}$ -kerogen) have slightly lower values (approximately -30.4‰ to -27.6‰) than those of $\delta^{13}\text{C}$ -TOC (Table T2). In HCl/HF treatment, it is likely that the F⁻ ion forms fluoride as a secondary product (Cody et al., 2005), which might cause errors between $\delta^{13}\text{C}$ -TOC and $\delta^{13}\text{C}$ -kerogen. Our data are roughly comparable with $\delta^{13}\text{C}$ -TOC for the Juan de Fuca basalts (approximately -34.6‰ to -21.6‰) (Lever et al., 2013). However, the carbon isotopic compositions of basalts probably show mass-dependent isotopic fractionation because the TC and TOC of basalts were low.

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Figure F1. Analytical procedures for (A) sediments and (B) hard rock samples. EA/IRMS = elemental analyzer/isotope ratio mass spectrometer.

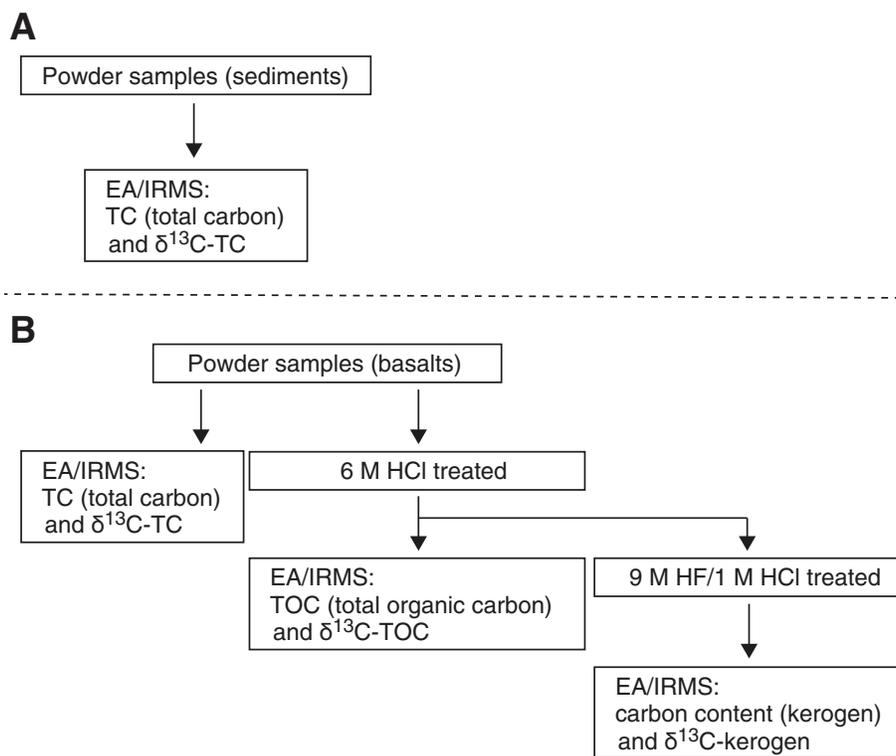


Figure F2. Total carbon (TC) content and $\delta^{13}\text{C}$ -TC of sediments, Holes U1382B, U1383D, and U1384A. We analyzed each sample twice. Plots = average values. Error bars = ranges between minimum and maximum values. VPDB = Vienna Pee Dee belemnite.

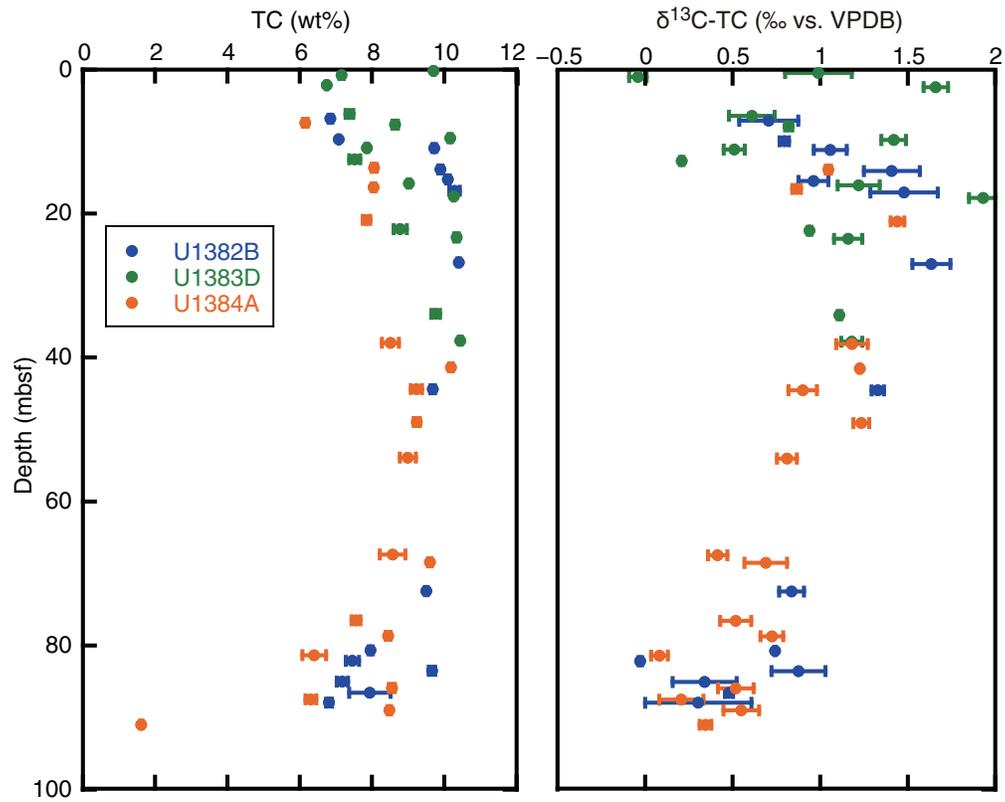


Figure F3. Depth-related profiles, Holes U1382A and U1383C. **A.** Total carbon (TC) content. **B.** Total organic carbon (TOC) content. **C.** $\delta^{13}\text{C}$ -TC. **D.** $\delta^{13}\text{C}$ -TOC of hard rocks (basalts, carbonates, and sedimentary breccias). We analyzed each sample twice. Plots = average values. Error bars = ranges between minimum and maximum values. VPDB = Vienna Pee Dee belemnite.

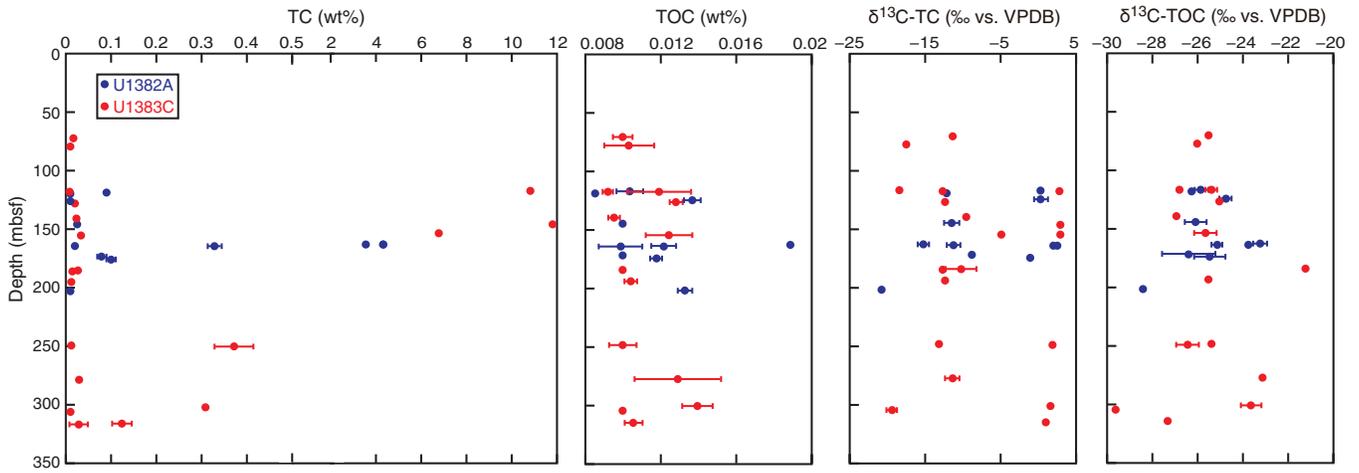


Table T1. Total carbon (TC) contents and carbon isotopic values ($\delta^{13}\text{C}$ [bulk]) of sediment samples, Holes U1382B, U1383D, and U1384A.

Core, section, interval (cm)	Depth (mbsf)	TC (wt%)	$\delta^{13}\text{C}$ -TC (‰ vs. VPDB)
336-U1382B-			
2H-2, 73–78	6.83	6.85 (± 0.05)	0.71 (± 0.17)
2H-4, 61–66	9.71	7.08 (± 0.00)	0.80 (± 0.03)
2H-5, 31–37	10.91	9.72 (± 0.05)	1.06 (± 0.09)
2H-7, 28–33	13.88	9.89 (± 0.06)	1.41 (± 0.16)
3H-1, 22–27	15.25	10.10 (± 0.05)	0.96 (± 0.09)
3H-2, 33–37	16.85	10.28 (± 0.15)	1.48 (± 0.19)
4H-3, 42–47	26.80	10.40 (± 0.04)	1.64 (± 0.11)
6H-2, 55–60	44.42	9.68 (± 0.06)	1.33 (± 0.04)
9H-1, 42–47	72.45	9.50 (± 0.04)	0.84 (± 0.07)
9H-7, 28–33	80.69	7.96 (± 0.06)	0.74 (± 0.00)
9H-CC2, 4–28	82.14	7.46 (± 0.18)	-0.03 (± 0.01)
10H-3, 19–23	83.52	9.66 (± 0.09)	0.88 (± 0.15)
10H-4, 19–24	85.03	7.18 (± 0.15)	0.34 (± 0.18)
10H-5, 23–28	86.57	7.94 (± 0.57)	0.48 (± 0.02)
10H-6, 10–15	87.94	6.81 (± 0.08)	0.30 (± 0.30)
336-U1383D-			
1H-1, 15–21	0.18	9.70 (± 0.02)	0.99 (± 0.19)
1H-2, 11–16	0.77	7.16 (± 0.06)	-0.04 (± 0.05)
1H-3, 11–17	2.18	6.75 (± 0.02)	1.66 (± 0.07)
2H-2, 18–23	6.16	7.38 (± 0.11)	0.61 (± 0.13)
2H-3, 16–21	7.64	8.64 (± 0.08)	0.82 (± 0.02)
2H-4, 55–60	9.53	10.16 (± 0.04)	1.42 (± 0.07)
2H-5, 40–45	10.88	7.86 (± 0.04)	0.51 (± 0.06)
2H-6, 49–54	12.47	7.52 (± 0.15)	0.21 (± 0.01)
3H-2, 35–40	15.85	9.02 (± 0.04)	1.22 (± 0.12)
3H-3, 63–68	17.63	10.26 (± 0.02)	1.93 (± 0.08)
3H-6, 67–72	22.17	8.78 (± 0.18)	0.94 (± 0.01)
3H-7, 32–37	23.32	10.34 (± 0.06)	1.16 (± 0.08)
5H-1, 31–37	33.94	9.76 (± 0.12)	1.11 (± 0.01)
5H-4, 22–27	37.65	10.44 (± 0.04)	1.18 (± 0.06)
336-U1384A-			
2H-7, 89–94	13.66	8.06 (± 0.07)	1.05 (± 0.02)
3H-3, 36–41	16.37	8.05 (± 0.05)	0.87 (± 0.03)
3H-6, 37–42	20.88	7.85 (± 0.10)	1.44 (± 0.04)
5H-5, 19–25	37.96	8.51 (± 0.24)	1.18 (± 0.09)
5H-7, 60–67	41.38	10.18 (± 0.05)	1.23 (± 0.00)
6H-2, 26–31	44.39	9.23 (± 0.16)	0.90 (± 0.08)
6H-5, 34–40	48.97	9.24 (± 0.08)	1.24 (± 0.04)
7H-2, 59–64	53.90	8.99 (± 0.22)	0.81 (± 0.06)
8H-5, 67–72	67.37	8.57 (± 0.36)	0.42 (± 0.06)
8H-6, 25–30	68.45	9.60 (± 0.05)	0.69 (± 0.12)
9H-5, 37–42	76.51	7.56 (± 0.12)	0.52 (± 0.09)
9H-7, 11–16	78.67	8.44 (± 0.06)	0.73 (± 0.07)
10H-2, 13–18	81.36	6.40 (± 0.33)	0.08 (± 0.05)
10H-5, 18–23	85.91	8.55 (± 0.09)	0.52 (± 0.10)
10H-6, 27–31	87.49	6.31 (± 0.15)	0.21 (± 0.13)
10H-7, 27–32	89.00	8.48 (± 0.05)	0.55 (± 0.10)
11H-2, 46–51	90.99	1.62 (± 0.04)	0.35 (± 0.03)

VPDB = Vienna Peedee belemnite.

Table T2. Total carbon (TC) contents, total organic carbon (TOC) contents, and carbon isotopic values ($\delta^{13}\text{C}$ -TC and $\delta^{13}\text{C}$ -TOC) of basalt, sediment, and sediment breccia samples and carbon contents and carbon isotopic values ($\delta^{13}\text{C}$ -kerogen) of kerogen samples extracted from basalt samples, Holes U1382A and U1383C.

Core, section, interval (cm)	Lithologic character	Depth (mbsf)	TC (wt%)	$\delta^{13}\text{C}$ -TC (‰ vs. VPDB)	TOC (wt%)	$\delta^{13}\text{C}$ -TOC (‰ vs. VPDB)	Carbon (wt%)	$\delta^{13}\text{C}$ -kerogen (‰ vs. VPDB)
336-U1382A-								
3R-3, 89–93	Basalt	117.10	0.09 (± 0.00)	0.12 (± 0.11)	0.01 (± 0.00)	-25.90 (± 0.34)	—	—
3R-4, 99–103	Basalt	118.61	0.01 (± 0.00)	-12.27 (± 0.31)	0.01 (± 0.00)	-26.28 (± 0.13)	—	—
4R-2, 14–17	Basalt	124.50	0.01 (± 0.00)	0.22 (± 0.88)	0.01 (± 0.00)	-24.77 (± 0.26)	31.72 (± 8.52)	-27.63 (± 0.66)
6R-3, 30–33	Basalt	144.54	0.03 (± 0.01)	-11.62 (± 0.95)	0.01 (± 0.00)	-26.10 (± 0.48)	30.61	-29.87
8R-2, 13–15	Basalt	162.73	0.02 (± 0.00)	-15.38 (± 0.71)	0.02 (± 0.00)	-23.23 (± 0.29)	—	—
8R-3, 10–1	Sediment	163.04	4.33 (± 0.01)	-11.37 (± 0.87)	—	—	—	—
8R-4, 34–36	Basalt	163.70	0.33 (± 0.02)	1.84 (± 0.30)	0.01 (± 0.00)	-25.16 (± 0.24)	—	—
8R-4, 34–36	Sediment	163.70	3.56 (± 0.16)	2.38 (± 0.10)	—	—	—	—
9R-1, 76–79	Basalt	171.68	0.08 (± 0.01)	-8.91 (± 0.01)	0.01 (± 0.00)	-26.42 (± 1.17)	—	—
9R-3, 21–24	Basalt	174.02	0.10 (± 0.01)	-1.18 (± 0.27)	0.01 (± 0.00)	-25.48 (± 0.69)	8.04 (± 2.33)	-30.44 (± 0.26)
12R-2, 44–47	Basalt	201.55	0.01 (± 0.00)	-20.94 (± 0.07)	0.01 (± 0.00)	-28.45 (± 0.17)	—	—
336-U1383C-								
2R-1, 126–130	Basalt	70.79	0.02 (± 0.00)	-11.57 (± 0.04)	0.01 (± 0.00)	-25.53 (± 0.13)	—	—
3R-1, 110–113	Basalt	77.72	0.01 (± 0.00)	-17.65 (± 0.35)	0.01 (± 0.00)	-26.05 (± 0.13)	—	—
7R, 235–237	Basalt	116.80	0.01 (± 0.00)	-18.59 (± 0.36)	—	—	—	—
7R-2, 70–72	Carbonate	117.15	10.86 (± 0.04)	2.69 (± 0.20)	0.01 (± 0.00)	-25.39 (± 0.26)	—	—
7R-2, 85–88	Basalt	117.31	0.01 (± 0.00)	-12.91 (± 0.07)	0.01 (± 0.00)	-26.82 (± 0.03)	—	—
8R-2, 59–62	Basalt	126.57	0.02 (± 0.00)	-12.56 (± 0.04)	0.01 (± 0.00)	-25.07 (± 0.06)	—	—
9R-4, 72–75	Basalt	139.38	0.02 (± 0.00)	-9.63 (± 0.12)	0.01 (± 0.00)	-26.98 (± 0.14)	—	—
10R-2, 65–67	Carbonate	145.93	11.89 (± 0.01)	2.76 (± 0.11)	—	—	—	—
11R-1, 52–55	Basalt	154.04	0.03 (± 0.00)	-5.01 (± 0.45)	0.01 (± 0.00)	-25.67 (± 0.50)	—	—
11R-1, 52–55	Sediment	154.04	6.81 (± 0.01)	2.82 (± 0.00)	—	—	—	—
14R-2, 11–13	Basalt	183.65	0.03 (± 0.00)	-10.41 (± 2.02)	0.01 (± 0.00)	-21.24 (± 0.11)	—	—
14R-2, 67–69	Basalt	184.21	0.02 (± 0.00)	-12.79 (± 0.19)	—	—	—	—
16R-1, 68–70	Basalt	193.49	0.01 (± 0.00)	-12.57 (± 0.42)	0.01 (± 0.00)	-25.55 (± 0.15)	2.86 (± 0.19)	-29.72 (± 0.46)
23R-1, 48–51	Basalt	247.49	0.01 (± 0.00)	-13.29 (± 0.23)	0.01 (± 0.00)	-25.41 (± 0.10)	—	—
23R-2, 12–15	Basalt	248.48	0.37 (± 0.04)	1.79 (± 0.15)	0.01 (± 0.00)	-26.46 (± 0.50)	—	—
26R-1, 127–130	Basalt	276.68	0.03 (± 0.00)	-11.53 (± 0.92)	0.01 (± 0.00)	-23.15 (± 0.08)	—	—
29R-1, 94–97	Basalt	300.15	0.31 (± 0.00)	1.45 (± 0.24)	0.01 (± 0.00)	-23.65 (± 0.44)	—	—
30R-1, 18–20	Basalt	303.89	0.01 (± 0.00)	-19.57 (± 0.67)	0.01 (± 0.00)	-29.66 (± 0.04)	21.21 (± 0.39)	-29.42 (± 0.94)
31R-2, 49–51	Basalt	314.00	0.13 (± 0.02)	0.88 (± 0.43)	0.01 (± 0.00)	-27.36 (± 0.12)	—	—

Carbon contents of kerogen were determined relative to fractions treated with HCl/HF. VPDB = Vienna Peedee belemnite.