Predominance of hexamethylated 6-methyl branched glycerol dialkyl glycerol tetraethers in the Mariana Trench: source and environmental implication

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Abstract. Branched glycerol dialkyl glycerol tetraethers (brGDGTs) are useful molecular indicators for organic carbon (OC) sources and the paleoenvironment. Their application in marine environments, however, is complicated because of a mixed terrestrial and marine source. Here, we examined brGDGTs in sediments from the Mariana Trench, the deepest ocean without significant terrestrial influence. Our result shows a strong predominance of hexamethylated 6-methyl brGDGT (IIIa’) (73.40 ± 2.39 % of total brGDGTs) and an absence of 5-methyl brGDGTs, different from previously reported soils and marine sediments that comprised both 5-methyl and 6-methyl brGDGTs. This unique feature, combined with high δ13COC (−19.82 ± 0.25 %), low OC/TN ratio (6.72 ± 0.84), low branched and isoprenoid tetraether (BIT) index (0.03 ± 0.01), and high acyclic hexa-/pentamethylated brGDGT ratio (7.13 ± 0.98), support that brGDGTs in the Mariana Trench sediments are autochthonous rather than terrestrial products. The compiling of literature data shows that the enhanced fractional abundance of hexamethylated 6-methyl brGDGTs is a common phenomenon in continental margins when the marine influence was intensified. The cross plot of acyclic hexa-/pentamethylated brGDGT ratio and fractional abundance of brGDGT IIIa’ provide a novel approach to distinguish terrestrial and marine-derived brGDGTs.

1 Introduction

Glycerol dialkyl glycerol tetraethers (GDGTs), including isoprenoidal GDGTs (iGDGTs) and branched GDGTs (brGDGTs), are widely distributed biomarkers in terrestrial and marine settings (De Rosa and Gambacorta, 1988; Sinninghe Damsté et al., 2000). iGDGTs containing an isoprenoid carbon skeleton are predominantly synthesized by archaea belonging to the phylum Thaumarchaeota (Sinninghe Damsté et al., 2002; Schouten et al., 2008; Knappy et al., 2011; Zeng et al., 2019). Unlike iGDGTs, brGDGTs consisting of four to six methyl groups and zero to two cyclopentane moieties are synthesized by some bacteria including, but not limited to, Acidobacteria (Sinninghe Damsté et al., 2011). These bacteria are able to alter the degree of methylation and cyclization of brGDGTs with changing ambient environmental conditions (Weijers et al., 2007b). A survey for global soils reveals that the cyclization of branched tetraethers (CBT) correlates with soil pH, while the methylation of branched tetraethers (MBT) is dependent on mean annual air temperature (MAT) and to a lesser extent on soil pH (Weijers et al., 2007b; De Jonge et al., 2014a), leading to the development of brGDGT-based MBT–CBT proxies for pH and MAT. BrGDGTs are generally more abundant in peats and soils than marine sediments, and they decrease from coastal to distal marine sediments (Hopmans et al., 2004; Schouten et al., 2013). Thus, brGDGTs and iGDGTs were thought to be biomarkers for terrestrial (particularly soil) and marine organic matter, respectively. This source differ-
ence led to the development of the branched vs. isoprenoid tetraether (BIT) index for estimation of terrestrial (soil) OC in marine sediments (Hopmans et al., 2004).

BrGDGT-derived proxies such as BIT, MBT, and CBT have been used to assess OC sources (Herfort et al., 2006; Kim et al., 2006; Loomis et al., 2011; Wu et al., 2013), soil pH, and MAT in diverse environments (Weijers et al., 2007a; Sinninghe Damsté et al., 2008; Peterse et al., 2012; Yang et al., 2014). However, one weakness of these proxies is the source uncertainty. Although brGDGTs were assumed to be specific for soil and peat bacteria (Hopmans et al., 2004; Weijers et al., 2007a, b), different compositions of brGDGTs among rivers (Zhang et al., 2012; Zell et al., 2013, 2014a), lakes (Sinninghe Damsté et al., 2009; Tierney and Russell, 2009; Loomis et al., 2011; Buckles et al., 2014), marine waters (Liu et al., 2014; Xie et al., 2014; Zell et al., 2014b), and sediments (Peterse et al., 2009; Zhu et al., 2011; Xiao et al., 2016) suggest multiple sources. Besides temperature and pH, oxygen (Qin et al., 2015) and moisture (Dang et al., 2016a) can also influence the composition of GDGTs. For example, in a Swiss lake (Lake Lugano), the vertical pattern of brGDGTs and bacterial 16S rRNA gene data suggested that brGDGTs were synthesized by multiple groups of bacteria thriving under contrasting redox regimes (Weber et al., 2018).

Weijers et al. (2007b) detected nine brGDGT isomers in peat and soils and assigned them to 5-methyl brGDGTs. De Jonge et al. (2013) developed a new chromatographic method using two silica columns and found that the brGDGTs previously identified as 5-methyl brGDGTs were actually mixtures of 5-methyl and 6-methyl isomers. As a result, the number of brGDGTs increased from 9 to 15, which was further expanded after the identification of 7-methyl brGDGTs (Ding et al., 2016). The analytical improvement has opened a window for the redefinition and recalibration of brGDGT-derived proxies (De Jonge et al., 2014a; Xiao et al., 2015). Adopting the new chromatographic method, several studies have provided clues of in situ production of brGDGTs in rivers (De Jonge et al., 2014b, 2015), lakes (Weber et al., 2015, 2018), and marine sediments (De Jonge et al., 2016; Sinninghe Damsté, 2016). However, rivers, lakes, and marginal seas are usually subject to terrestrial influence, making it difficult to distinguish allochthonous terrestrial and autochthonous aquatic contributions to the brGDGT pool.

Here, we choose the Challenger Deep, Mariana Trench, to study brGDGTs in marine settings. This deepest trench (11000 m) is remote from any landmass and has no significant terrestrial influence (Jamieson, 2015). Our goals are (1) to determine the composition and concentration of brGDGTs in the Mariana Trench sediments and constrain their source and (2) to characterize in situ production of brGDGTs in marine sediments and assess their environmental implication.

2 Material and methods

2.1 Study area and samples

The Mariana Trench is formed as the subduction of the Pacific plate beneath the eastern edge of the Philippine Sea plate. It has a total length of ca. 2500 km and a mean width of 70 km (Fryer, 1996). The Challenger Deep is located on the southern rim of the Mariana Trench and has a water depth of 11000 m. The Mariana Trench is overlain by extremely oligotrophic waters with annual primary production of 59 g C m⁻² yr⁻¹ (Jamieson, 2015). However, sediments in the Challenger Deep were found to support elevated microbial activity compared to adjacent abyssal plains (Glud et al., 2013). This characteristic has been attributed to unique V-shaped geometry, intense seismic activity, and high-frequency fluid dynamics within the trench that promote lateral transport of sediments and organic matter from shallow regions into the trench bottom (Jamieson, 2015; Xu et al., 2018).

During an expedition aboard RV Zhang Jian (December 2016 to February 2017), a sediment core (MT1, 11 cm long) was retrieved from the Challenger Deep using an autonomous 11000 m rated lander (Fig. 1). The core was immediately stored at −20 °C in a dark room until transported to the laboratory in Shanghai (China). The core was sliced at 1–2 cm intervals. All sediment samples (n = 10) were freeze-dried at −40 °C and homogenized by steel spatulas.

Besides the Mariana Trench sediments, a soil sample (Soil-1) from China grassland was analyzed. This soil sample was used for comparison of brGDGTs between soil and trench sediments.

2.2 Lipid extraction and GDGT analyses

Sediment samples (0.5–2 g) were mixed with an internal standard C₄₆ GDGT (Huguet et al., 2006) and 15 mL of dichloromethane/methanol (3:1, v/v). After being ultrasonically extracted for 15 min, the extracts were centrifuged (3000 rpm, 5 min), and the supernatants were decanted into clean flasks. This extraction was repeated three times. The combined extracts were concentrated by a rotary evaporator and further blown down to dryness under mild nitrogen streams. The total lipid extract was dissolved in hexane/isopropanol (99:1, v/v) and filtered through a 0.45 µm PTFE filter. An Agilent ultrahigh-performance liquid chromatography – atmospheric pressure chemical ionization – triple quadruple mass spectrometry system (UHPLC–APCI–MS) was used for analysis of GDGTs. The separation of 5- and 6-methyl brGDGTs was achieved with two silica liquid chromatography (LC) columns in sequence (150 mm × 2.1 mm; 1.9 µm, Thermo Finnigan; USA). The detailed instrumental parameters were described by Hopmans et al. (2016). The protonated ions were m/z 1050, 1048, 1046, 1036, 1034, 1032, 1022, 1020, and 1018 for
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brGDGTs; 1302, 1300, 1298, 1296, and 1292 for iGDGTs; and 744 for C46 GDGT. Since the response factors of GDGTs were not determined due to the lack of authentic standard, we did not calculate the absolute concentration of GDGTs. Instead, we reported the relative concentration based on peak areas (pa) of respective GDGTs normalized to total GDGTs.

2.3 GDGT-derived parameters

The BIT index, ratio of acyclic hexa- to pentamethylated brGDGTs, and weighted average number of cyclopentane moieties for the tetramethylated brGDGTs (#rings\textsubscript{tetra}) were calculated according to the definitions of Hopmans et al. (2004), Xiao et al. (2016), and Sinninghe Damsté (2016), respectively.

\[
\text{BIT} = \frac{(Ia + IIa + IIIa + IIa' + IIIa')/(Ia + IIa + IIIa + IIa' + IIIa' + Cren)}{(IIa + Ila + IIIa + IIa' + IIIa')/(IIa + Ila')}
\]

\[
\sum \text{IIa} / \sum \text{Ila} = (IIIa + IIIa')/(IIa + Ila')
\]

\[
#\text{rings}_{\text{tetra}} = (Ib + 2 \cdot Ic)/(Ia + Ib + Ic)
\]

The roman numbers denote relative abundance of GDGTs that were depicted in Fig. 2.

2.4 Bulk geochemical analysis

About 1–2 g of each sediment sample was treated with 1 N HCl to remove carbonates, rinsed with ultrapure water, and then freeze-dried. After having homogenized with an agate mortar and pestle, approximately 35–40 mg of de-carbonated sediments was analyzed using a model 100 isotope ratio mass spectrometer (Isoprime Corporation, Cheadle, UK) and a Vario ISO TOPE cube elemental analyzer (Elementar Analysensysteme GmbH, Hanau, Germany). All isotopic data were reported in \( \delta \) notation relative to Vienna Pee Dee Belemnite (VPDB). The intra-lab standard for normalizing stable isotopic composition of OC (\( \delta^{13} \text{C}_{\text{OC}} \)) was USG24 (graphite, \(-16.05\%e\)) (IAEA, Vienna, Austria). The average standard deviation of each measurement, determined by replicate analyses of two samples, was \( \pm 0.004 \text{ wt }% \) for organic carbon (OC) content, \( \pm 0.031 \text{ wt }% \) for total nitrogen (TN) content, and \( \pm 0.03 \%e \) for \( \delta^{13} \text{C}_{\text{OC}} \).

2.5 Literature data compilation

The dataset is composed of 2031 samples, including 634 soil samples, 473 peat samples, 88 river samples, 410 lake samples, and 426 marine samples (Fig. 1). The sample information was listed in the Supplement. The soil samples are from globally distributed soils (De Jonge et al., 2014a; Ding et al., 2015; Xiao et al., 2015; Yang et al., 2015; Lei et al., 2016; Wang et al., 2016, 2018, 2019; Li et al., 2018; Zang et al., 2018). The peat samples are from 96 different peatlands around the world (Naafs et al., 2017). The river samples are from the Danube River (Freymond et al., 2016), the Yenisei River (De Jonge et al., 2015), and the Tagus River (Warden et al., 2016). The lake samples are from east African lakes (Russell et al., 2018), Chinese lakes (Dang et al., 2016b, 2018; Li et al., 2017), Lake St Front (Martin et al., 2019), Lake Lugano, and other lakes in the European Alps (Weber et al., 2018). The marine samples are from Atlantic Ocean (Warden et al., 2016), the Kara Sea (De Jonge et al., 2015, 2016), the Berar River delta (Sinninghe Damsté, 2016), the Ceará Rise (Soelen et al., 2017), the North Sea (Dearing Crampton-Flood et al., 2018), and the Mariana Trench (this study). The criteria for citing the literature data is that 5- and 6-methyl brGDGTs should be separated and quantified. It is noted that...
Figure 2. Chemical structures of brGDGTs and crenarchaeol.

Two studies (Weber et al., 2018; Martin et al., 2019) have analyzed 5-, 6-, and 7-methyl brGDGTs, but due to very limited reports for 7-methyl brGDGTs they are not included in our literature dataset.

2.6 Statistical analysis

The SPSS package 22 (IBM, USA) was used for statistical analyses including Pearson correlation coefficient (r) and one-way analysis of variance (ANOVA). The significance level was set at \( p < 0.05 \).

3 Results

3.1 Bulk geochemical parameters

The OC content, TN content, molar ratio of OC to TN content (OC/TN), and \( \delta^{13}C_{OC} \) are summarized in Table 1. The OC and TN contents of sediments varied between 0.26 % and 0.31 % with an average of 0.28 ± 0.01 % (mean ± SD; same hereafter) and between 0.04 % and 0.06 % (0.05 ± 0.01 %), respectively. The OC/TN ratio and \( \delta^{13}C_{OC} \) ranged from 5.62 to 8.34 (6.72 ± 0.84) and −19.47 % to −20.27 % (−19.82 ± 0.25 %), respectively. Both the \( \delta^{13}C_{OC} \) and OC/TN ratio were comparable to previously reported levels for the southern Mariana Trench rim and slope (\( \delta^{13}C_{OC} \) −20.48 ± 0.88 %; OC/TN, 7.00 ± 1.76) (Luo et al., 2017).

<table>
<thead>
<tr>
<th>Sample ID</th>
<th>Depth (cm)</th>
<th>OC (wt %)</th>
<th>TN (wt %)</th>
<th>OC/TN (mol/mol)</th>
<th>( \delta^{13}C_{OC} ) (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MT1</td>
<td>0–2</td>
<td>0.31</td>
<td>0.05</td>
<td>6.52</td>
<td>−20.02</td>
</tr>
<tr>
<td>MT2.5</td>
<td>2–3</td>
<td>0.27</td>
<td>0.05</td>
<td>6.05</td>
<td>−19.66</td>
</tr>
<tr>
<td>MT3.5</td>
<td>3–4</td>
<td>0.29</td>
<td>0.05</td>
<td>6.85</td>
<td>−19.83</td>
</tr>
<tr>
<td>MT4.5</td>
<td>4–5</td>
<td>0.27</td>
<td>0.05</td>
<td>5.78</td>
<td>−19.84</td>
</tr>
<tr>
<td>MT5.5</td>
<td>5–6</td>
<td>0.29</td>
<td>0.06</td>
<td>6.13</td>
<td>−19.94</td>
</tr>
<tr>
<td>MT6.5</td>
<td>6–7</td>
<td>0.30</td>
<td>0.06</td>
<td>5.62</td>
<td>−19.47</td>
</tr>
<tr>
<td>MT7.5</td>
<td>7–8</td>
<td>0.27</td>
<td>0.04</td>
<td>7.27</td>
<td>−19.54</td>
</tr>
<tr>
<td>MT8.5</td>
<td>8–9</td>
<td>0.29</td>
<td>0.05</td>
<td>6.93</td>
<td>−19.82</td>
</tr>
<tr>
<td>MT9.5</td>
<td>9–10</td>
<td>0.28</td>
<td>0.04</td>
<td>7.74</td>
<td>−20.09</td>
</tr>
<tr>
<td>MT10.5</td>
<td>10–11</td>
<td>0.26</td>
<td>0.04</td>
<td>8.34</td>
<td>−20.27</td>
</tr>
</tbody>
</table>
3.2 Composition and fractional abundance of GDGTs

The fractional abundance of iGDGTs and brGDGTs is summarized in Table 2. iGDGTs were the dominant components, accounting for 96.8 % to 98.6 % of total GDGTs in Mariana Trench sediments. The proportion of brGDGTs was substantially lower than that of iGDGTs, ranging from 1.4 % to 3.2 %. For all sediment samples, the BIT index remained at a low level (0.03 ± 0.01).

With improved chromatographic performance, 5- and 6-methyl brGDGTs were completely separated (Fig. 3a, b). Interestingly, the mass chromatograms of the Mariana Trench sediment (MT-4) only showed a single peak for acyclic penta- (m/z 1036; Fig. 3c) and hexamethylated (m/z 1050; Fig. 3d) brGDGTs. This feature is different from most previous studies showing two or more peaks (i.e., 5-, 6-, and even 7-methyl brGDGTs) occurred in soils, lake, and marine sediments (e.g., De Jonge et al., 2013; Xiao et al., 2015; Ding et al., 2016). In order to determine the structure of brGDGTs, we compared the mass spectra of brGDGTs between MT-4 and Soil-1. The soil sample (Soil-1) has been reported to contain both 5-methyl brGDGTs (major component) and 6-methyl brGDGTs (minor component) (Xiao et al., 2015), and its IIIa/IIa’ and Ila/Ila’ ratios are 12.5 and 8.2, respectively (Fig. 3a, b). For the mixed sample of Soil-1 (soil) and MT-4 (Mariana Trench), the mass spectrum showed two peaks for m/z 1050 (hexamethylated brGDGTs) as well as for m/z 1036 (pentamethylated brGDGTs) (Fig. 3e, f). The comparison of retention time among Soil-1, MT-4, and the mixed sample (Soil-1 + MT-4) revealed that the peaks of m/z 1050 and 1036 in MT-4 were pentamethylated 6-methyl brGDGTs (Ila’) and hexamethylated 6-methyl brGDGTs (IIIa’), respectively, eluting after 5-methyl brGDGTs (Fig. 3). This structural assignment was corroborated by an intermediate level of 5-methyl/6-methyl brGDGT ratio in the mixed sample (1.4 for m/z 1050 and 7.4 for m/z 1036) compared to that in Soil-1 (12.5 and 8.2, respectively) and MT-4 (0 for both) (Fig. 3).

In the sediment core of the Mariana Trench, the brGDGTs were constantly dominated by 6-methyl isomers (82.25 %–86.91 %). The fractional abundance of 5-methyl brGDGTs, however, was too low to be quantified. BrGDGT IIIa’ was the dominant compound (73.40 ± 2.39 % of total brGDGTs), followed by brGDGT Ia (12.46 ± 1.14 %) and brGDGT IIa’ (10.45 ± 1.20 %). The proportion of cyclic compounds (brGDGT Ib, Ic, Iib’) was only 3.69 ± 0.75 %, resulting in a low level of #rings=[67] (0.26 ± 0.04). The classification based on the number of methyl groups showed the dominance of hexamethylated brGDGTs (73.53 ± 2.56 %) over tetramethylated (15.43 ± 1.53 %) and pentamethylated (11.04 ± 1.49 %) brGDGTs.

4 Discussion

4.1 In situ production of 6-methyl brGDGTs in the Mariana Trench

To our knowledge, there are only two studies reporting GDGTs in the Mariana Trench. Guan et al. (2019) investigated iGDGT distribution in the surface sediments (4900–7068 m depth) from the southern Mariana Trench, while Ta et al. (2019) analyzed iGDGTs and brGDGTs in two sediment cores (ca. 5400 m depth) in the subduction plate of the Mariana Trench. However, neither of these studies separated the 5- and 6-methyl brGDGTs. Our improved chromatographic method demonstrated the strong predominance of 6-methyl brGDGTs and the absence of 5-methyl brGDGTs in the Mariana Trench sediments. In order to decipher the mechanism of producing such unique compositions of brGDGTs, the source assessment is needed.

Multiple lines of evidence (i.e., δ13COC, OC/TN ratio, and biomarkers) support an in situ production of brGDGTs in the Mariana Trench. The δ13COC and OC/TN ratio are widely used indicators to distinguish terrestrial vs. marine OC. Generally, marine algae and bacteria are protein-rich and have a OC/TN ratio of 4 to 10, whereas vascular land plants are cellulose and lignin-rich and have a OC/TN ratio of 20 or
between marine sediments and atmospheric dust in the equatorial west African coast, and their great difference suggested an in situ production rather than dust input for brGDGTs in the marine sediments. Here, we compared the brGDGT compositions between the Mariana Trench sediments and terrestrial samples reported in literature (Fig. 5). Relative to the Mariana Trench sediments (brGDGT Ia 12.46 ± 1.14%, 5-methyl brGDGTs ~0, brGDGT IIa' 73.40 ± 2.39%), those terrestrial samples had significantly higher proportions of brGDGT Ia (soil 37.52 ± 25.91%, peat 59.40 ± 21.19%, river 15.38 ± 2.97%) and 5-methyl brGDGTs (soil 23.56 ± 14.83%, peat 34.04 ± 19.18%, river 33.25 ± 8.51%) but lower proportions of brGDGT IIa' (soil 4.89 ± 4.82%, peat 4.86 ± 4.68%, river 11.68 ± 4.40%) ($p < 0.005$) (Fig. 5). In addition, those terrestrial samples are globally distributed, many of which are from the inner Asian continent, a major source area for dust in the North Pacific (Husar et al., 2001). Thus, brGDGTs in the Mariana Trench sediments are unlikely derived from atmospheric dusts.

Low BIT index of the Mariana Trench sediments (0.03 ± 0.01; Fig. 6) is similar to distal marine sediments (an average of 0.04) (Schouten et al., 2013; Weijers et al., 2014), again suggesting insignificant terrestrial inputs in the Mariana Trench. By compilation of globally distributed 1354 soils and 589 marine sediments, Xiao et al. (2016) proposed the $(\text{IIa} + \text{IIa}')/(\text{IIa} + \text{IIa}')$ ratio as an indicator for the source of brGDGTs, which was < 0.59 in 90% of soils and 0.59–0.92 and > 0.92 in marine sediments with and without significant terrestrial inputs, respectively. For the Mariana Trench sediments, the $(\text{IIa} + \text{IIa}')/(\text{IIa} + \text{IIa}')$ ratio varied between 5.68 and 8.33 (7.13 ± 0.98) (Fig. 6). Such a high $(\text{IIa} + \text{IIa}')/(\text{IIa} + \text{IIa}')$ ratio suggests a predominant marine source for brGDGTs in the Mariana Trench sediments.

Overall, the bulk geochemical parameters, the composition of brGDGTs, and the BIT index unanimously support the in situ product rather than terrestrial input for brGDGTs in Mariana Trench sediments.

Table 2. Fractional abundance of brGDGTs and iGDGTs in the Mariana Trench sediments.

<table>
<thead>
<tr>
<th>Sample ID</th>
<th>Ia (%)</th>
<th>Ib (%)</th>
<th>Ic (%)</th>
<th>IIa (%)</th>
<th>IIa' (%)</th>
<th>IIb (%)</th>
<th>IIb' (%)</th>
<th>IIC (%)</th>
<th>IIIa (%)</th>
<th>IIIa' (%)</th>
<th>IIIb (%)</th>
<th>IIIb' (%)</th>
<th>IIIc (%)</th>
<th>brGDGTs (%)</th>
<th>iGDGTs (%)</th>
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<tbody>
<tr>
<td>MT1</td>
<td>13.6</td>
<td>2.7</td>
<td>1.5</td>
<td>10.4</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>71.8</td>
<td>0.0</td>
</tr>
<tr>
<td>MT2.5</td>
<td>13.5</td>
<td>2.4</td>
<td>1.6</td>
<td>12.1</td>
<td>0.0</td>
<td>1.3</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>69.0</td>
<td>0.0</td>
</tr>
<tr>
<td>MT3.5</td>
<td>11.1</td>
<td>1.4</td>
<td>0.6</td>
<td>9.5</td>
<td>0.0</td>
<td>0.6</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
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<td>9.2</td>
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<td>0.8</td>
<td>0.0</td>
<td>0.0</td>
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<td>0.0</td>
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<td>9.1</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
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<td>11.3</td>
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<td>1.2</td>
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<td>0.0</td>
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<td>0.0</td>
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<td>MT8.5</td>
<td>13.0</td>
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<td>1.1</td>
<td>12.7</td>
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4.2 High proportion of brGDGT IIIa’ as a common phenomenon in marine environments

Not only the Mariana Trench but also continental margins showed relatively high proportions of hexamethylated 6-methyl brGDGTs in sediments. Dearing Crampton-Flood et al. (2018) investigated brGDGTs and bulk properties of organic matter in a sediment core from the North Sea Basin. The OC content, $\delta^{13}$C$_{OC}$ value, BIT, and #rings$_{tetra}$ index indicated a transition from the predominant marine OC in the Pliocene to the predominant terrestrial OC in the Pleistocene. Correspondingly, the proportion of brGDGT IIIa’ was significantly higher in the Pliocene (8.06 ± 1.92 %) than the Pleistocene (5.16 ± 0.83 %) and exhibited a significant correlation with $\delta^{13}$C$_{OC}$ ($R^2 = 0.68, p < 0.001$) and the BIT index ($R^2 = 0.46, p < 0.001$) (Fig. 7a, b, c). Similar to the North Sea Basin, the proportion of GDGT IIIa’ in the Kara Sea also showed a significant correlation with $\delta^{13}$C$_{OC}$ ($R^2 = 0.34; p < 0.001$) and the BIT index ($R^2 = 0.50; p < 0.001$) in a 303 cm sediment core covering at least the past 1.33 × 10$^3$ years (Fig. 7d, e, f) (De Jonge et al., 2016). These results suggest that brGDGTs synthesized by marine organisms comprise higher fractional abundance of hexamethylated 6-methyl brGDGTs.

Besides temporal variations in sediment cores, the fractional abundance of 6-methyl brGDGTs also changed from land to sea in modern samples. Warden et al. (2016) analyzed brGDGTs along a transect from the Tagus River to the deep ocean off the Portuguese margin. Along this transect, the BIT index significantly decreased from 0.9 to < 0.1, reflecting an increase in marine contribution to the sedimentary OC pool (Fig. 7h). Meanwhile, the proportion of brGDGT IIIa’ substantially increased from 11.07 ± 2.62 % to 29.31 ± 6.45 %, and brGDGT IIIa’ became the dominant compound of brGDGTs (Fig. 7g). In surface sediments of the Berau River delta, the #rings$_{tetra}$ index, an indicator of the
source of brGDGTs, showed a marked increase from the river mouth (0.22) to the shelf break (0.83) (Sinninghe Damsté, 2016), while the proportion of brGDGT IIIa’ increased seawards, presenting similar distribution patterns as the $\delta^{13}$C$_{OC}$ and BIT index (Fig. 7i, j, k).

In sum, the studies for the Kara Sea (De Jonge et al., 2016), the North Sea Basin (Dearing Crampton-Flood et al., 2018), the Tagus River basin (Warden et al., 2016), and the Berau River delta (Sinninghe Damsté, 2016) show enhanced fractional abundance of 6-methyl brGDGTs (particularly IIIa’) as marine influence was intensified. These findings, along with the strong predominance of brGDGT IIIa’ in the Mariana Trench sediments, suggest that the high proportion of brGDGT IIIa’ in total brGDGTs is a common phenomenon in marine environments where in situ production of brGDGTs is significant.
4.3 Potential mechanisms to produce high fractional abundance of brGDGT IIIa′

A survey of brGDGTs in globally distributed soils suggested that brGDGT-producing microbes could adjust their membrane lipid compositions in response to changing environmental conditions, reflected by the increase in cyclization degree of brGDGTs and the shift from 5- to 6-methyl group with increasing pH and decreasing methylation of brGDGTs with temperature (Weijers et al., 2007b; De Jonge et al., 2014a; Ding et al., 2015; Xiao et al., 2015). This adaption mechanism may be extrapolated to marine brGDGT-producing organisms. In the Mariana Trench, in situ production yielded brGDGTs with a strong predominance of brGDGT IIIa′ (> 69 %), low proportion of cyclopentane-containing brGDGTs (< 10 %), and low level of the #rings$_{stra}$ index (< 0.32). These characters seem in contrast to the previous result that the fractional abundance of cyclopentane-containing brGDGTs is positively correlated with pH (Sinninghe Damsté, 2016). This difference can be explained by two reasons. First, the isomerization of brGDGTs is more efficient in response to changing pH compared to the cyclization of brGDGTs (Ding et al., 2015). Based on the global soil dataset, the soil pH presents stronger correlations with the isomerization of branched tetaether index (IBT; Xiao et al., 2015) than with the #rings$_{stra}$ index as well as the cyclization index (CBT$_{Sme}$) (De Jonge et al., 2014a). Meanwhile, global soils with pH > 8 (n = 58) are characterized by higher fractional abundance of 6-methyl brGDGTs (68.22 ± 10.41 %) than the cyclopentane-containing brGDGTs (16.69 ± 9.43 %). Thus, weakly alkaline sediment and seawater (pH ~ 8.0) may be important factors for producing more 6-methyl hexamethylated brGDGTs in the Mariana Trench. The second explanation is the effect of low temperature. Marine microbes tend to produce more hexamethylated brGDGTs at low temperature (Sinninghe Damsté, 2016), thus reducing the proportion of cyclic tetramethylated and pentamethylated brGDGTs. The ternary diagram, plotted with fractional abundance of tera-, penta-, and hexamethylated brGDGTs (Fig. 8), shows that brGDGTs in the Mariana Trench sediments comprise high fractional abundance of hexamethylated brGDGTs (73.53 ± 2.56 %). Given these facts, we propose that low temperature and high pH in deep-sea environments are responsible for the production of brGDGTs with a high degree of methylation and predominance of 6-methyl brGDGTs, especially brGDGT IIIa′.

In situ production of brGDGTs may take place in the water column, sediments, or both. Sinninghe Damsté (2016) suggested that in situ production of brGDGTs was a widespread phenomenon in shelf sediments that was especially pronounced at 50–300 m depths. The extended dataset (63–5521 m depths) showed a large variability for the degree of cyclization of brGDGTs (Weijers et al., 2014), suggesting that the brGDGTs are mainly produced in sediments where the pH of porewater is more variable than that of overlying seawaters. However, in the Mariana Trench sediments, the degree of cyclization fell in a narrow range (0.26 to 0.32). As a result, both water column and sediments are possibly where brGDGTs are produced.

The bottom of the Mariana Trench is an extreme environment, characterized by high hydrostatic pressure (> 100 MPa), low temperature (ca. 2 °C), and darkness (Jamieson, 2015). In addition, the surface waters in the Mariana Trench region are extremely oligotrophic, leading to a low sinking flux of particulate organic matter to the seafloor. Under such extreme conditions, unique microbes may have evolved, such as proliferation of hydrocarbon-degrading bacteria (Liu et al., 2019). These deep-sea microbes may have different responses to changing ambient temperature and pH than their shallow-dwelling counterparts, and they thus produce brGDGTs with different compositions. Investigations of microbial community and intact polar lipids are needed for understanding the source and environmental implication of brGDGTs in the Mariana Trench and other marine settings.

4.4 Deciphering brGDGT provenance in marine sediments

There are increasing concerns about the robustness of brGDGT-based proxies. Deciphering the provenance of brGDGTs is prerequisite for the application of brGDGT-based proxies. In continental margins, intense land–sea interaction occurs, resulting in the complex composition and sources of brGDGTs (De Jonge et al., 2016; Sinninghe Damsté, 2016; Dearing Crampton-Flood et al., 2018). Our study highlights that marine in situ production of brGDGTs tends to exhibit higher fractional abundance of brGDGT IIIa′ relative to terrestrial brGDGTs. However, there is an overlap for the fractional abundance of brGDGT IIIa′ between
Figure 9. Scatterplots of the \((\text{IIIa} + \text{IIIa}')/(\text{IIa} + \text{IIa}')\) ratio versus the proportion of brGDGT \text{IIIa}' in globally distributed soils and marine sediments. The solid, dashed, and dotted lines denote the linear fit, 95\% confidence band, and 95\% prediction band of concatenated data, respectively. The number of samples, slope, \(R^2\), and \(p\) values of calibration are for the global distributed soils, marine sediments, and Mariana Trench sediments.

The unique composition of brGDGTs in the Mariana Trench has significant implications for the brGDGT-based proxies. As a setting remote from landmass, the Mariana Trench provides an opportunity to distinguish marine in situ production from a terrestrial origin of brGDGTs that usually muddles the interpretation of shelf sediments. However, it is unclear what the similarity and differences are for brGDGT-producing microbes and their response to environmental factors between the Mariana Trench and continental shelf. In addition, the weight contribution to the brGDGT pool from sediments and the water column remains elusive. Since factors such as nutrients, particle loading, bacterial community, and oceanographic parameters (e.g., oxygenation, salinity, currents) vary significantly between the shelf and trench as well as among different hadal trenches, the brGDGT-producing microbes are likely different. Therefore, investigation of brGDGTs in multiple hadal trenches and shallow marine regions is needed to decipher their source and environmental control, which are beneficial for accurate application of the brGDGT-based proxies such as MBT, CBT, and BIT.

5 Conclusions

Our study represents the first investigation for 5-methyl and 6-methyl brGDGTs in the hadal trench, enabling us to draw four conclusions.

The Mariana Trench sediments are characterized by the strong predominance of 6-methyl brGDGTs (84.57 ± 1.53\% of total brGDGTs), especially brGDGT IIIa' (73.40 ± 2.39\%), whereas 5-methyl brGDGTs are below the detection limit. This unique feature has never been reported and attributed to the combined effect of insignificant terrestrial influence, alkaline seawater, and low subsurface temperature in the Mariana Trench.

High brGDGT \((\text{IIIa} + \text{IIIa}')/(\text{IIa} + \text{IIa}')\) ratios (7.13 ± 0.98), enriched \(\delta^{13}\text{C}_{\text{OC}}\) signatures (−19.82 ± 0.25\%), low OC/TN ratios (6.72 ± 0.84), low BIT index (0.03 ± 0.01), high abundance of 6-methyl brGDGTs, and absence of 5-methyl brGDGTs support an in situ production of brGDGTs in the Mariana Trench sediments.

BrGDGTs in sediments from the Mariana Trench and continental margins comprise a higher proportion of hexamethylated 6-methyl brGDGTs with intensified marine influence. The slope of fractional abundance of brGDGT IIIa' and the \((\text{IIIa} + \text{IIIa}')/(\text{IIa} + \text{IIa}')\) index can be used to deci-
pher terrestrial and marine provenance of brGDGTs. Since in situ production of the predominant hexamethylated 6-methyl brGDGTs influences the robustness of brGDGT-based proxies, this study provides a new way to estimate brGDGT sources and holds some promise in reducing uncertainty of brGDGT-based paleoenvironmental proxies.

The uniqueness of the Mariana Trench in that it is remote from any landmass allows us to distinguish marine in situ production from terrestrial origin of brGDGTs. However, it is unclear how comparable this unique site is to shallow marine settings and other hadal trenches. Therefore, comparison studies of brGDGTs for different hadal trenches as well as between hadal and non-hadal sites are recommended.

**Data availability.** Data have been made available through figshare: https://doi.org/10.6084/m9.figshare.9896120.v1 (Xiao et al., 2019).

**Supplement.** The supplement related to this article is available online at: https://doi.org/10.5194/bg-17-2135-2020-supplement.

**Author contributions.** WX and YX developed the study design. Fieldwork for this study was conducted by YX. WX, YW, YL, XZ, and LS conducted the experiments and data analyses. All authors contributed to data interpretation. WX and YX compiled and processed all presented data and wrote the manuscript.

**Competing interests.** The authors declare that they have no conflict of interest.

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