Supplementary material and method for supplementary figure S4.

Sampling was carried out during a research cruise (64PE424) onboard the R/V Pelagia from July - August 2017. The cruise followed a North to South transect in the North Atlantic. The two samples shown here are from a northern station (61.71°, -20.49°) and a southern station (30.66°, -14.25°). McLane in situ pumps (McLane Laboratories Inc., Falmouth) fitted with pre-ashed 0.7 µm x 142 mm, GF/F filters (Pall Corporation, Port Washington, NY) were used to collect suspended particulate matter (SPM) from the water column. Between 400 and 500 L was filtered at 200 m depth at each station using a cut-off at a pre-programmed pressure threshold. The filters were stored at −80 °C until extraction. Filters were freeze-dried and extracted and further treated as described in Materials and Methods in the main text. The extracts were analyzed in two ways, first using the identical method as described in the main text. In addition, analysis was performed using a reversed phase method based on Wörmer et al. (2013) with some modifications. Identical eluents, gradient and column conditions were used but the analysis was performed on the UHPLC-Q Exactive system described here. HESI settings were as follows: sheath gas (N2) pressure 40 (arbitrary units), auxiliary gas (N2) pressure 10 (arbitrary units), auxiliary gas (N2) T 50 ˚C, sweep gas (N2) pressure 5 (arbitrary units), spray voltage 4.5 kV (positive ion ESI), capillary temperature 300 °C, S-Lens 70 V. IPLs were analyzed with a mass range of m/z 400 to 2000 and data dependent approach as described in the main text. Peak integration and area corrections based on the PAF internal standard were performed as described in main text.

Supplementary figure legends

Fig. S1. Structures of isoprenoid core-GDGTs discussed in the text (left) and the most commonly detected IPL types (right).

Fig. S2. HPLC-ESI/MS Extracted ion current (EIC) of *m/z* 1643.4549 within 3 ppm mass accuracy representing the ammoniated molecule of DH-GDGT-0 in subsurface sediment (10-12 cm) at 1306 mbsl. The chromatogram shows three separate peaks representing three different isomers (the peak at 19.39 min is the signal of the second isotopologue of GDGT-1). MS^2 mass spectra of each of the three isomers are shown in panel a-c in order of retention time. Panel a shows a single fragment ion at *m/z* 1302.3280. Panel b shows an additional fragment ion at m/z 1284.3152. Panel c shows a main fragmentation product at *m/z* 1464.3739. The fragmentation product at *m/z* 1302.3280 shown in panel a represents the core lipid of GDGT-0 resulting from the loss of 2 hexose moieties from DH-GDGT-0, with 1 hexose moiety attached to either side of the molecule. The additional

fragment ion at *m/z* 1284 shown in panel b represents the loss of an additional water from the GDGT-0 core lipid, which can only occur if a free alcohol moiety is present and thus both hexose moieties are postulated to be attached to one side of the GDGT-0 molecule. Panel C shows the loss of a single hexose from the *m/z* 1643 parent, but no further fragmentation. suggesting that this isomer is in fact the IPL-type HCP GDGT, where the hexose is positioned at the opposite side from the cyclopentanetetraol moiety. If the hexose moiety would have been bound to the cyclopentanetetraol moiety, as suggested by Sturt et al., 2004, an additional loss of water from the MH-GDGT-0 would have been expected, similar to what was observed in the fragmentation of DH-GDGT-0 shown in panel b.

Fig. S3. Summed EICs of monoisotope plus first isotopologue in diverse Arabian Sea sediments of a) MH-GDGTs (e.g. MH-GDGT-0; $[M+NH_4]^+$, *m/z* 1481.4020 plus 1482.4054, respectively), b) DH-GDGTs (e.g. DH-GDGT-0; $[M+NH_4]^+$, *m/z* 1643.4549 plus 1644.4582, respectively) and c) HPH-GDGTs (e.g. HPH-GDGT-0; $[M+NH_4]^+$, *m/z* 1723.4212 plus 1724.4246, respectively) within 3 ppm mass accuracy. The used internal standard, 1-O-hexadecyl-2-acetoyl-sn-glycero-3-phosphocholine (PAF; not shown), elutes at 31.4 minutes.

Fig. S4. Peak area per liter for six IPL-GDGTs (GDGT-0 and crenarchaeol with MH (monohexose), DH (dihexose) and HPH (hexose phosphohexose) headgroups) in two suspended particulate matter (SPM) samples from the North Atlantic (A = Northern station, B = Southern station) analysed using a normal phase (blue bars) and reversed phase HPLC/MS method (orange bars). Numbers in graph indicate the ratio between peak areas measured with normal phase vs. reversed phase method. N.D. = not detected.

Supplementary figures and tables

Fig. S1.

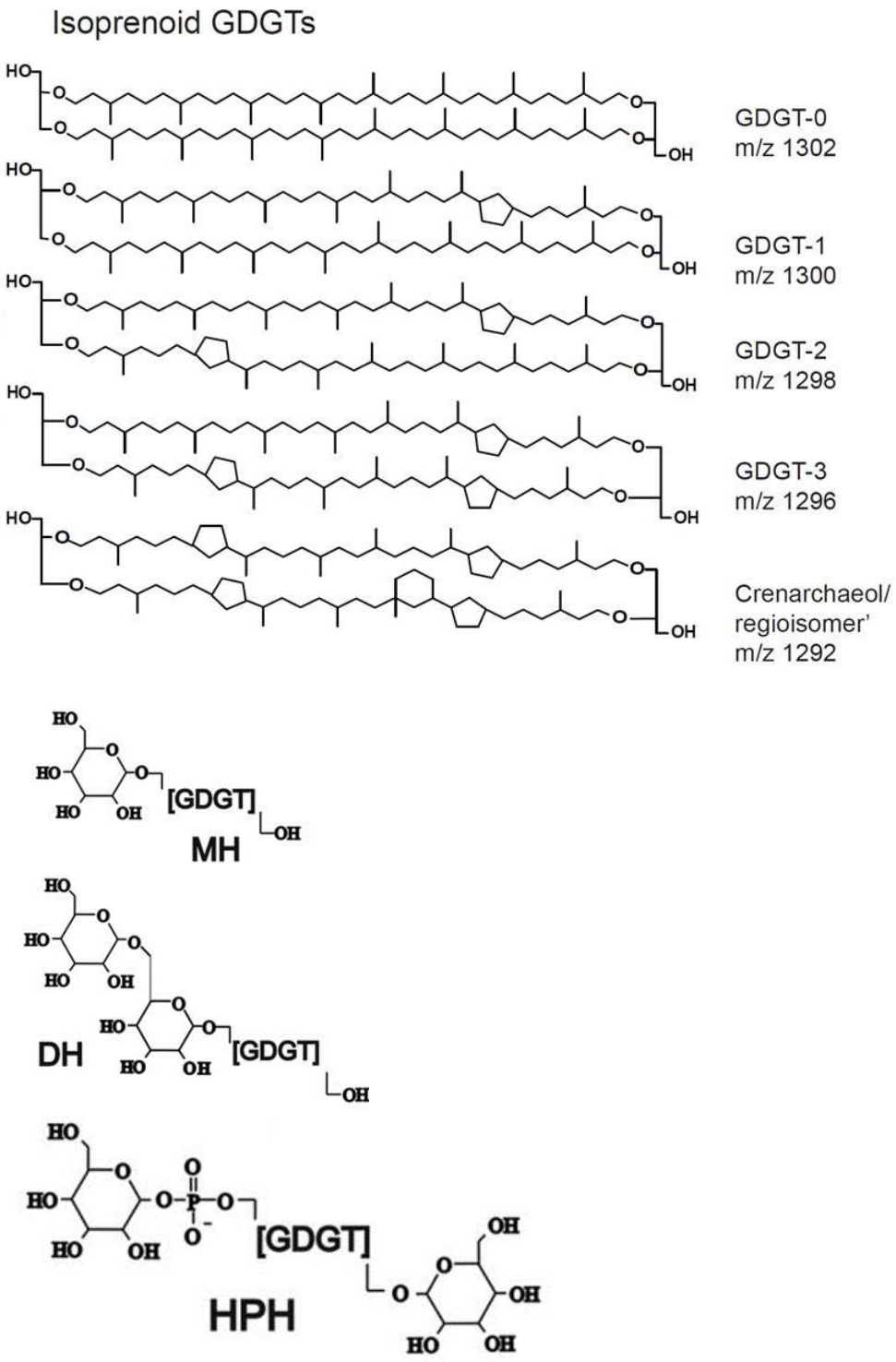


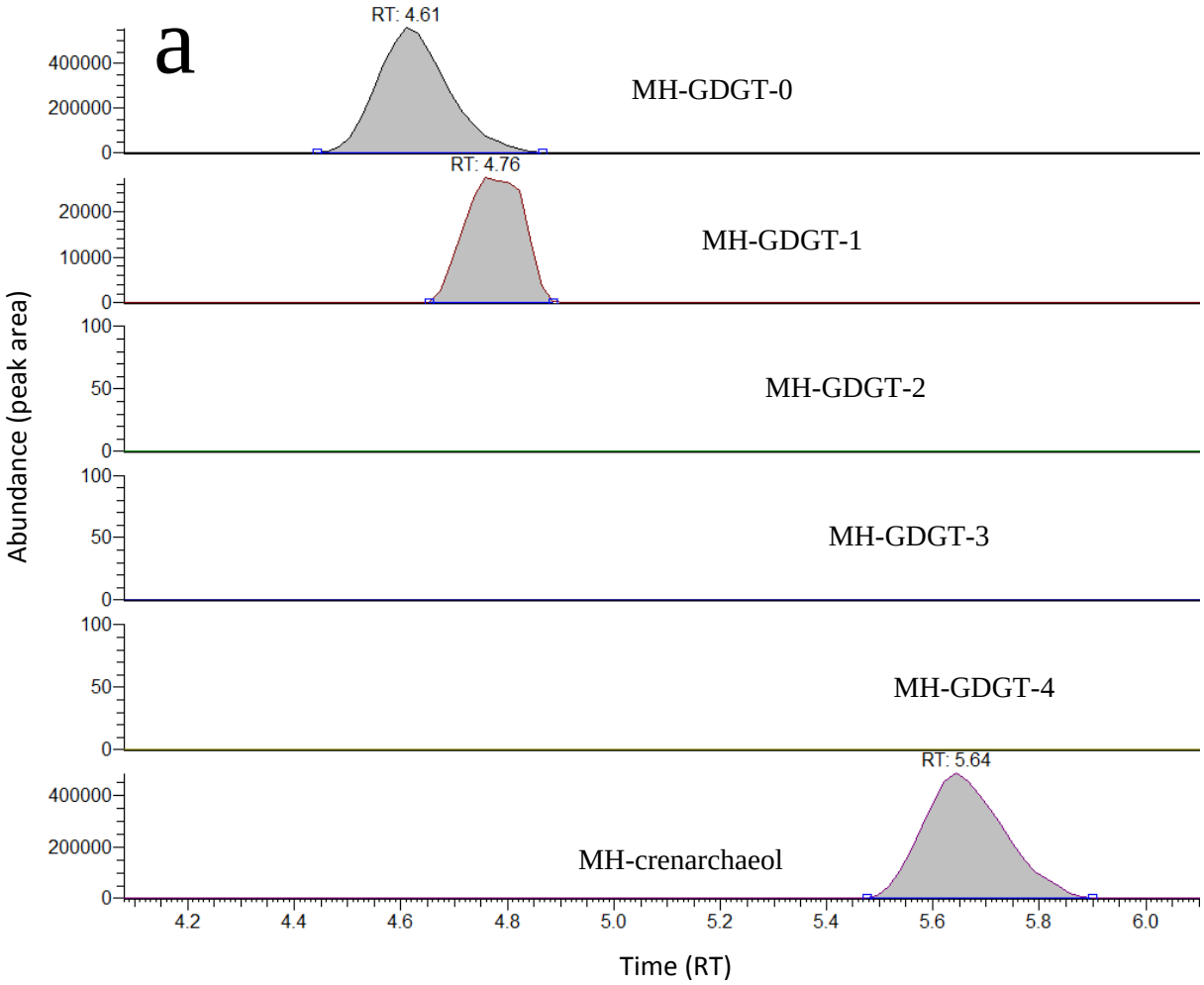
Table S1. PCR primers, efficiency and R^2 of the QPCR assays

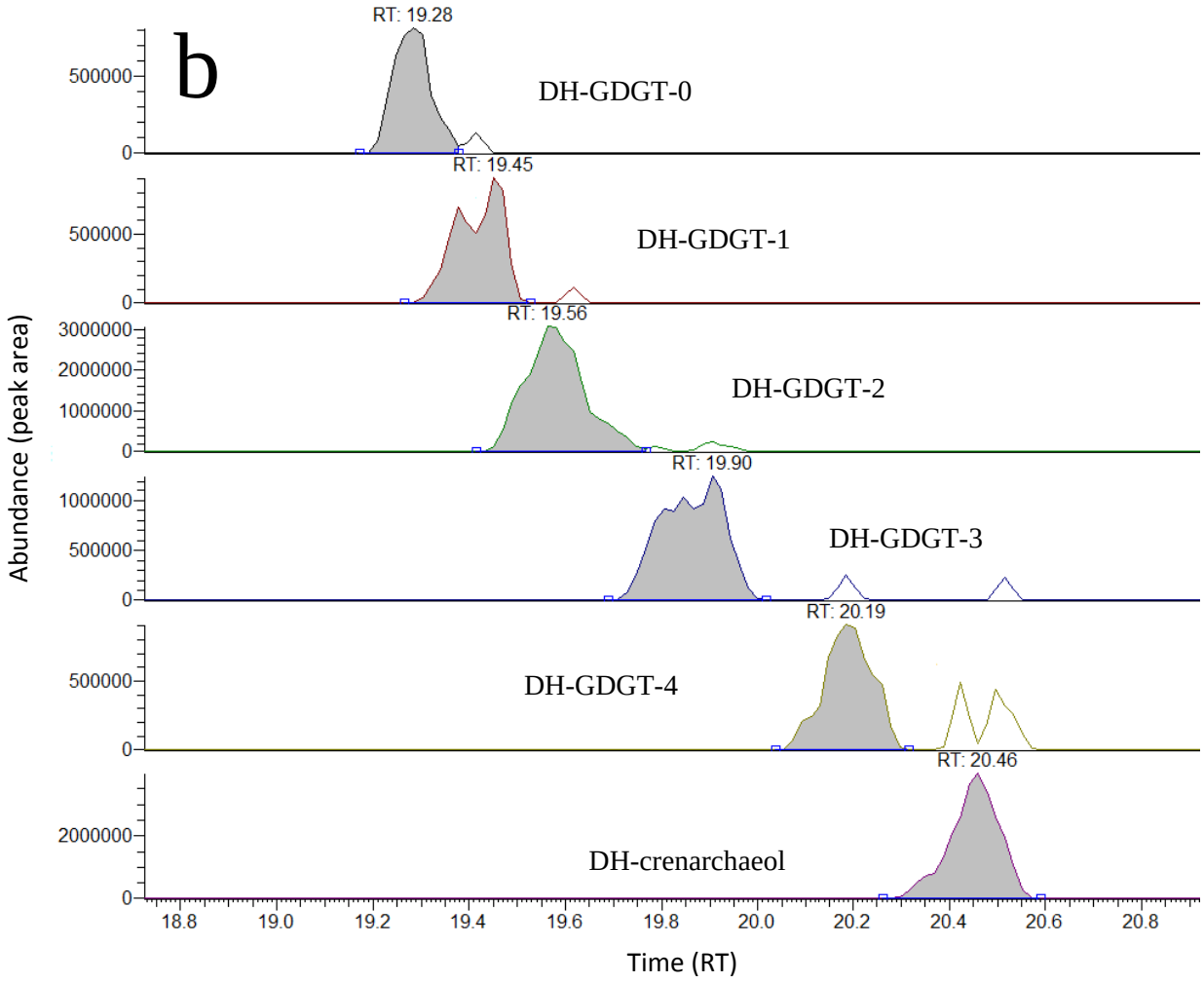
QPCR (gene)	Primers	Ref	Efficiency	R^2
16S rRNA	Parch519F/ARC915R	Pitcher et al., 2011	99.8	0.96
16S rRNA	Parch519F/ARC915R	Pitcher et al., 2011	87.5	1.00
16S rRNA	Parch519F/ARC915R	Pitcher et al., 2011	90.0	0.99
*Amo*A	CrenAmoAQ-F/CrenAmoAModR	Pitcher et al., 2011	97.5	1.00
*Amo*A	CrenAmoAQ-F/CrenAmoAModR	Pitcher et al., 2011	78.5	1.00

Fig. S2.

EIC of m/z 1643.4549 +/- 3 ppm
RT: 19.22
a
19.39
RT: 20.14
b
RT: 20.97
c
Relative abundance
Time (RT)
ms² of m/z 1643
1643
1302
1284
1464
m/z
m/z 1302
GDGT-0
m/z 1284
m/z 1464

Fig. S3


a
RT: 4.61
MH-GDGT-0
RT: 4.76
MH-GDGT-1
MH-GDGT-2
MH-GDGT-3
MH-GDGT-4
RT: 5.64
MH-crenarchaeol
Abundance (peak area)
Time (RT)
400000
200000
0
20000
10000
100
50
4.2
4.4
4.6
4.8
5.0
5.2
5.4
5.6
5.8
6.0


b
RT: 19.28
DH-GDGT-0
RT: 19.45
DH-GDGT-1
RT: 19.56
DH-GDGT-2
RT: 19.90
DH-GDGT-3
RT: 20.19
DH-GDGT-4
RT: 20.46
DH-crenarchaeol
Abundance (peak area)
Time (RT)
18.8
19.0
19.2
19.4
19.6
19.8
20.0
20.2
20.4
20.6
20.8

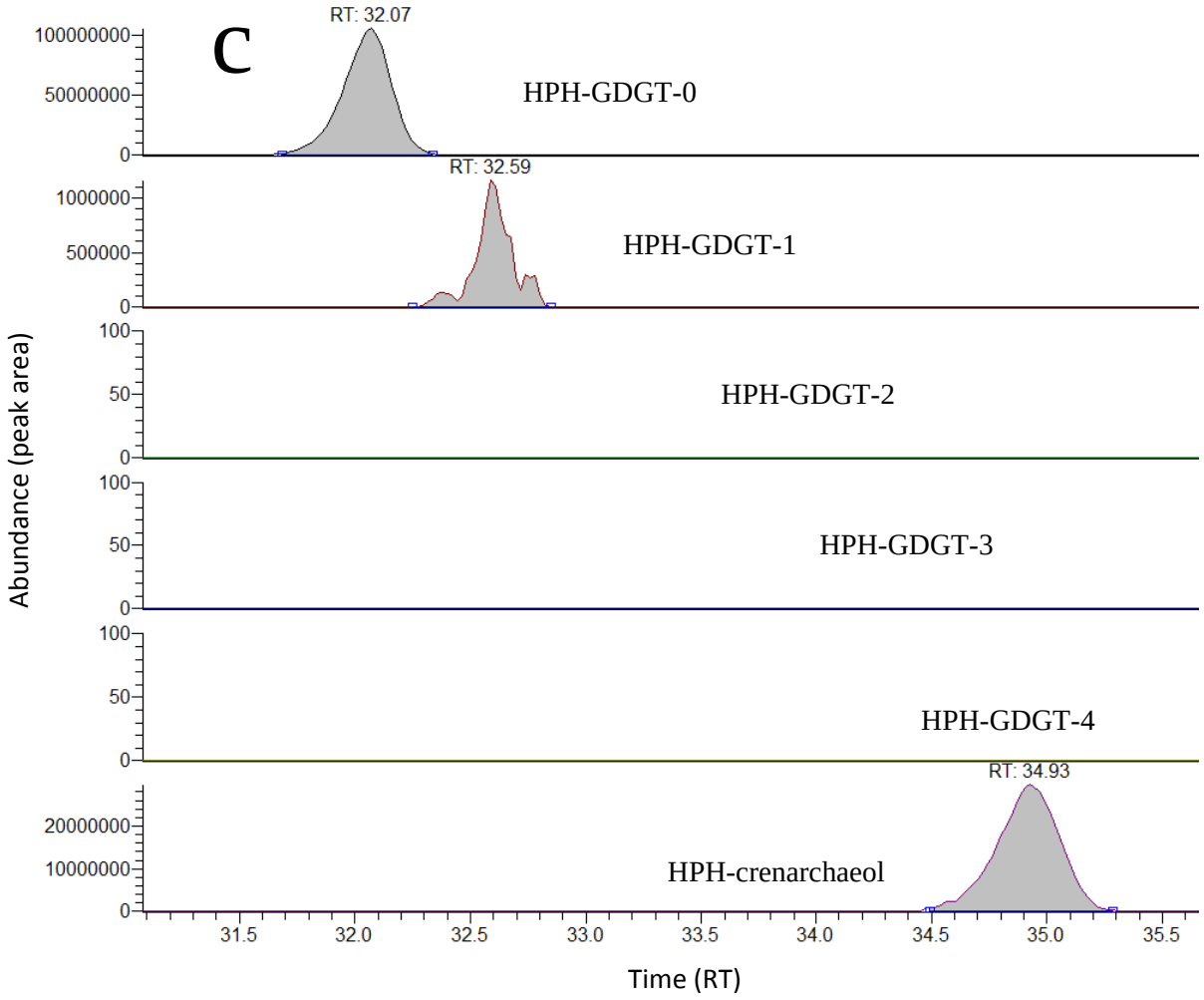
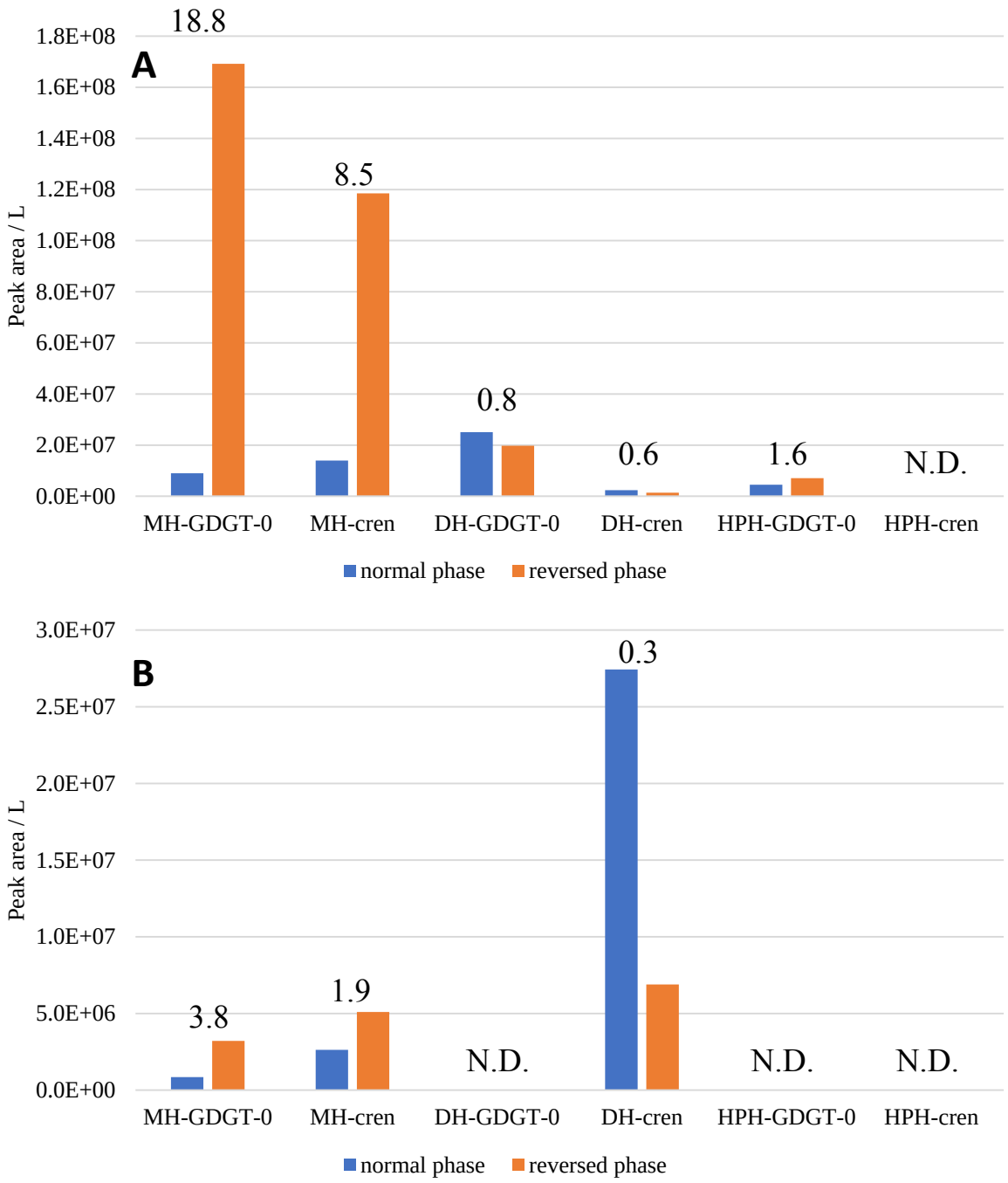

C
RT: 32.07
HPH-GDGT-0
RT: 32.59
HPH-GDGT-1
HPH-GDGT-2
HPH-GDGT-3
HPH-GDGT-4
RT: 34.93
HPH-crenarchaeol
Abundance (peak area)
Time (RT)

Fig. S4


A
18.8
8.5
0.8
0.6
1.6
N.D.
1.8E+08
1.6E+08
1.4E+08
1.2E+08
1.0E+08
8.0E+07
6.0E+07
4.0E+07
2.0E+07
0.0E+00
Peak area / L
MH-GDGT-0
MH-cren
DH-GDGT-0
DH-cren
HPH-GDGT-0
HPH-cren
normal phase
reversed phase
B
3.8
1.9
N.D.
0.3
N.D.
N.D.
3.0E+07
2.5E+07
2.0E+07
1.5E+07
1.0E+07
5.0E+06
0.0E+00
Peak area / L
MH-GDGT-0
MH-cren
DH-GDGT-0
DH-cren
HPH-GDGT-0
HPH-cren
normal phase
reversed phase