

# Hot-spring inputs and climate drive dynamic shifts in archaeal communities in Lake Magadi, Kenya Rift Valley

Evan R. Collins<sup>1</sup>, Troy M. Ferland<sup>2</sup>, Isla S. Castañeda<sup>3</sup>, R. Bernhart Owen<sup>4</sup>, Tim K. Lowenstein<sup>5</sup>,  
†Andrew S. Cohen<sup>6</sup>, Robin W. Renaut<sup>7</sup>, Molly D. O’Beirne<sup>1</sup>, Josef P. Werne<sup>1</sup>

<sup>1</sup>Department of Geology and Environmental Science, University of Pittsburgh, Pittsburgh, PA, 15213, United States

<sup>2</sup>Lamont-Doherty Earth Observatory, Columbia University, Palisades, NY, 10964, United States

<sup>3</sup>Department of Earth, Geographic and Climate Sciences, University of Massachusetts Amherst, Amherst, MA, 01003, United States

<sup>4</sup>Department of Geography, Hong Kong Baptist University, Kowloon Tong, Hong Kong

<sup>5</sup>Department of Geological Sciences, State University of New York, Binghamton, NY 13902, United States

<sup>6</sup>Department of Geosciences, The University of Arizona, Tucson, AZ 85721, United States

<sup>7</sup>Department of Geological Sciences, University of Saskatchewan, Saskatoon, SK S7N 5E2, Canada

Correspondence to: Evan R. Collins (erc92@pitt.edu/ecollins452@gmail.com)

**Abstract.** The Methane Index (MI) is an organic geochemical index that uses isoprenoid glycerol dialkyl glycerol tetraethers (GDGTs) as a proxy for methane cycling. Here, we report results from sediments in core MAG14-2A that span almost 500 ka in Lake Magadi, Kenya. The deposits show abrupt shifts between high and low MI values through calcareous, tuffaceous and zeolitic silts. The MI “switches “off” (MI < 0.2) and “on” (MI > 0.5) through the core with bulk organic matter enriched in <sup>13</sup>C during “MI-off” periods (~ -18‰) in the upper part of the core, whereas <sup>13</sup>C is lower (-22 to -25‰) in lower parts of the sedimentary sequence. Sediments deposited when the MI switches “on” showed δ<sup>13</sup>C<sub>OM</sub> values as low as -89.4 ‰, but most were within the range of -28 to -30‰, which is consistent with contributions from methanogens rather than methanotrophs. Thus, the likely source of these high MI values in Lake Magadi is methanogenic archaea. Our results show that hydrothermal inputs of bicarbonate-rich waters into Lake Magadi combined with further evaporative concentration cause a shift in the dominant archaeal communities, alternating between two stable states.

## 1 Introduction

Life thrives in East African soda lakes and has been the subject of modern studies of both prokaryotic and eukaryotic organisms, but few have studied their sediments over geologic timescales (Schagerl, 2016 and

chapters therein). Soda lakes represent ~ 18,500 km<sup>2</sup> in East Africa (calculated from values in Melack  
and MacIntyre, 2016). When compared to the three largest African freshwater lakes (lakes Victoria,  
Tanganyika, and Malawi), these soda lakes account for ~ 13% of the total lake-surface area in East Africa.  
A survey of microbial isolate diversity in East African lakes found evidence for cyanobacterial and  
archaeal primary producers with both oxygenic and anoxygenic phototrophs among the microbial  
population (Grant and Jones, 2016). Unique aerobic and anaerobic heterotrophs that use a variety of  
electron donors, including sulfur, sulfate, nitrite, carbon dioxide, and methane, were also identified  
(Grant and Jones, 2016, and sources therein). Many thermophilic archaea and bacteria isolates were also  
observed near hot-spring outflows (Grant and Jones, 2016).

Saline alkaline (soda) lakes in the East African Rift often become stratified meromictic water bodies with  
a dense monimolimnion below a chemocline (Melack and MacIntyre, 2016). Oxygen rarely penetrates  
the monimolimnion waters, and as a result, anaerobic bacteria and archaea dominate the bottom waters  
and sediments. Remineralizing organic matter from the upper water column (mixolimnion) supports the  
microbes and ~~generation of the~~ anaerobic oxidation of methane (AOM). Methane-oxidizing microbes,  
specifically archaeal anaerobic methane-oxidizers (ANME), are coupled to sulfate-reducing bacteria in a  
microbial consortium (Boetius et al., 2000; Hinrichs and Boetius, 2002; Werne et al., 2004). ANME  
mediate methane levels in freshwater and soda lakes and in modern oceanic systems, and account for  
approximately 90% of methane consumed through AOM (Egger et al., 2018). Rates of methane  
consumption differ by environment and type of ANME, with global freshwater systems ranging from 1  
to 1 x 10<sup>5</sup> nmol methane L<sup>-1</sup> day<sup>-1</sup> consumed (Martinez-Cruz et al., 2018). Although soda lakes have been  
less studied, consumption rates as high as 1.6 x 10<sup>4</sup> nmol methane L<sup>-1</sup> day<sup>-1</sup> have been observed in  
freshwater Lake Kivu (Roland et al., 2018). Tracking AOM over geologic time periods is important  
because methane release from tropical wetlands was concomitant with the end of glacial conditions in  
Europe and is poorly constrained (DeMenocal et al., 2000; Riddell-Young et al., 2023). Additionally,  
large methane releases might have been partly responsible for the Permo-Triassic mass-extinction event  
(Bernier, 2002).

Over geologic time, it is possible to gauge periods of increased methane oxidation, as shown by Zhang et al. (2011) in oceanic systems by using a ratio of archaeal GDGT lipids (de Rosa et al., 1977; Langworthy, 1977). The ratio, as described by Zhang et al. (2011), is known as the methane index (MI), which uses GDGTs produced predominantly by Euryarchaeal ANME. The MI has been used to discern methanotrophy using the assumption that benthic methanotrophic Euryarchaeota preferentially produce GDGT-1, -2, and -3, and that GDGTs crenarchaeol (cren) and crenarchaeol' (cren') are thought to come from Thaumarchaeota and Crenarchaeota, which are part of the TACK superphylum, typically found in the upper water column (Sinninghe Damsté et al., 2002; Pitcher et al., 2009; Zhang et al., 2011). Currently, the newly suggested names in the Genome Taxonomy Database for Thaumarchaeota and Crenarchaeota are Nitrososphaerota and Thermoproteota, respectively (Oren and Garrity, 2021; Rinke et al., 2021), which are used in this paper.

~~Moreover,~~ Kim and Zhang (2023) have shown a qualitative and quantitative relationship between the MI and methanotrophy in deep time, namely from the late Oligocene to the early Miocene. Kim and Zhang (2023) showed that the MI is applicable to AOM, with other biomarkers co-occurring in high-MI intervals representative of not only the Group I consortium of anaerobic methanotrophs (ANME) that produce GDGTs, but also of Group 2 and Group 3 consortia (ANME-2 and ANME-3 respectively). Until now, no studies have directly applied the MI to sediments in African soda lakes despite evidence for AOM in modern soda lakes. Combined with MI values, other methane-related indices are used here to interpret methanogenesis and methanotrophy related to AOM. Previous studies have used GDGT-0 and GDGT-2 ratioed to the GDGT crenarchaeol value, which was originally thought to only be produced by mesophilic Thermoproteota (Blaga et al., 2009; Weijers et al., 2012). However, the optimum temperature for crenarchaeol production is closer to 40-45 °C (Zhang et al., 2006). Blaga et al. (2009) found that methanogens predominantly produced GDGT-0, whereas Weijers et al. (2011) showed that methanotrophic archaea predominantly produce GDGT-2. ~~However, the optimum temperature is closer to 40-45 °C (Zhang et al., 2006).~~

Lake Magadi (Kenya) is a sulfate-limited lake, and therefore, methanogenesis and methanotrophy may  
85 co-occur without suppression of the higher energy yield of sulfate reduction (Nijaguna, 2006; Sorokin et  
al., 2007; Deocampo and Renaut, 2016; Lameck et al., 2023). Here, we document evidence of methane  
cycling in Lake Magadi using archaeal isoprenoid GDGT lipid biomarkers. Environmental influences on  
archaeal community composition included precipitation/evaporation fluctuations and variations in  
hydrothermal activity, the latter often related to contemporary tectonics. This study leverages four  
90 methane-related indices: (1) the MI; (2) the %GDGT-0/crenarchaeol; (3) %GDGT-2/crenarchaeol; and  
(4) the ratio of isoprenoid GDGTs [2]/[3] (hereafter, [2]/[3]) to understand methane cycling in recent and  
ancient lacustrine sediments. Two distinct communities were found using a combination of the MI and  
ratios of GDGT-0 and GDGT-2, normalized to crenarchaeol. Intervals of high methanotrophy, as  
evidenced by MI and %GDGT-2/crenarchaeol, were related to an equally high proportion of  
95 methanogens, while in periods when crenarchaeol was dominant, the methane indices were low.

## 2 Materials and methods

### 2.1 Study locations and sampling

Modern Lake Magadi is a seasonally flooded, saline alkaline pan composed of bedded trona  
([Na<sub>3</sub>(CO<sub>3</sub>)(HCO<sub>3</sub>)·2H<sub>2</sub>O]) located in the southern Kenya Rift near the border with Tanzania (Baker,  
100 1958; Eugster, 1980). Its elevation is approximately 600–605 m above sea level (asl), and it has a  
maximum depth during the rainy season of a few decimeters to ~ 1 m (Fig. 1; Renaut and Owen, 2023).  
Although Lake Magadi is situated near the equator, it lies in a rain shadow. Consequently, today it has a  
large moisture deficit (2400 mm evaporation versus 500 mm precipitation annually: Damnati and Taieb,  
1995).

105 The modern alkaline lake is fed by ephemeral streams and alkaline hot springs (up to 86°C at adjacent  
Nasikie Engida), distributed along faults around the shoreline (Baker, 1958; Crane, 1981; Allen et al.,  
1989; Renaut and Owen, 2023). Former high-level shorelines are preserved as coarse clastic sediments  
and locally as stromatolites around the lake. These vary in age and record lakes of different depths during

110 the Quaternary. Outcrops of sediments relevant to this study and situated near Lake Magadi are the  
Oloronga Beds and the Green Beds. The chert-bearing Oloronga Beds in outcrop ~~have been dated~~were  
laid down between ~ 800 and 300 ka, with cores extending this back to 1 Ma (Owen et al., 2019). Green  
Beds outcrops include abundant chert and have been variably dated between 191 and 40 ka (Behr and  
Röhricht, 2000; Owen et al., 2019) with cores suggesting a range from 380 to 105 ka (Owen et al., 2019).  
115 ~~More recently, the~~The High Magadi Beds were deposited between ~25 and 9 ka (Fairhead et al., 1972;  
Goetz and Hillaire-Marcel, 1992; Williamson et al., 1993; Behr and Röhricht, 2000; Owen et al., 2019;  
Reinhardt et al., 2019). Calcrete commonly caps the Oloronga Beds (Eugster, 1980), but fluvial erosion  
locally scoured those sediments before the Green Beds were laid down (Renaut and Owen, 2023).

120 ~~More recently, the stratigraphic terminology has been revised by Owen et al. (2025). They continued to~~  
~~recognise the Oloronga Beds below Lake Magadi under the same name but raised it to formation status~~  
~~with dates of 1000–380 ka. In contrast, they renamed the Green Beds to the Oloika Formation (380–105~~  
~~ka) and reassigned the Evaporite Series to the Magadi Formation (105–0 ka) and the High Magadi Beds~~  
~~to the High Magadi Formation. Various higher lake levels have been suggested based on stromatolites~~  
125 ~~(Casanova and Hillaire-Marcel, 1987) but some details are uncertain. Although Lake Magadi is situated~~  
~~near the equator, it lies in a rain shadow. Consequently, today it has a large moisture deficit (2400 mm~~  
~~evaporation versus 500 mm precipitation annually; Damnati and Taieb, 1995).~~

Lake Magadi was cored as part of the *Hominin Sites and Paleolakes Drilling Project* (HSPDP) in June  
130 2014 to further our understanding of the paleoenvironments in the East African Rift Valley and to better  
contextualize hominin remains and artifacts, and to understand possible environmental influences on  
hominin evolution and migration (Cohen et al., 2016). A 197.4-meter core (MAG-14-2A) was drilled in  
the northern end of Lake Magadi (1°51'5.76" S 36°16'45.84" E; Owen et al., 2019). In total, 107.7 m of  
sediments were recovered, with an overall core recovery of 55.4% (Cohen et al., 2016). Here, we use the  
135 age model from Owen et al. (2019). The core ranges from the modern trona surface (0 ka) to the Magadi  
Trachyte basement, dated to ~ 1 Ma at the core site (Owen et al., 2019). Cores were sampled in 2016  
during the initial core description at the Continental Scientific Drilling Facility (CSD, formerly LacCore)

at the University of Minnesota, Minneapolis. Altogether, 61 samples, covering the period from 456 ka to 14.9 ka (Table S1), were collected and freeze-dried from dark brown to black silty clay intervals. Based on their color, these samples were expected to have a high total organic carbon that would yield the highest quantity of biomarkers for our study. The non-ideal sampling strategy is due to poor core recovery.

Over the past million years, Lake Magadi has varied from swampy fresh water bodies to a large fresh to mildly saline lake that was continuously fed by rivers and groundwater, to smaller hypersaline lakes bounded by the Magadi grabens that dried to trona pans partly fed by hot springs (Owen et al., 2019; Renaut and Owen, 2023). From 545 to 380 ka the Magadi catchment progressively changed to a more arid condition with the palaeolake marked by abundant calcareous, organic-rich sediments (Owen et al., 2019). Periodic freshwater inundation occurred from 380 to 105 ka into a highly saline, alkaline lake that accumulated minor calcite and magnesium-rich calcite at lake margins. Ash that fell into this waterbody reacted to form a variety of zeolites with anoxic, sulfate-rich bottom-water brines subjected to microbial sulfate reduction (Owen et al., 2019; Deocampo et al., 2022). The most recent phases of the lake (105 to 0 ka) were more evaporatively enriched, with abundant trona and minor nahcolite. Well-preserved diatoms in sediments deposited after ~500 ka suggest very high aqueous silica in the paleolake in order to explain the preservation of their frustules under highly alkaline conditions, which may reflect strong evaporative concentration of silica-rich hydrothermal inflows (Owen et al., 2019).

## **2.2 ~~Leaf-wax~~GDGT and bulk organic preparation and analysis**

### **2.2.1 Lipid extraction**

To obtain a total lipid extract (TLE), 61 samples from Lake Magadi were freeze dried and homogenized and ca. 5–10 g of sediment were ultrasonically extracted with 2:1 DCM:MeOH. The TLE for each sample was treated with activated copper shot to remove elemental sulfur. The TLEs were then separated into three fractions (apolar (AP), polar one (P1), and polar two (P2)) using activated alumina via short column chromatography. The AP fraction was eluted with 4 mL of 9:1 Hexane (Hex):DCM (v/v), the P1 fraction with 4 ml 1:1 DCM:MeOH, and the P2 fraction with 4 ml MeOH. The P1 fractions were dried down and

165 re-dissolved in 99:1 Hex:Isopropanol (IPA) (v/v) and filtered through a 0.45  $\mu\text{m}$  4 mm diameter PTFE  
filter prior to GDGT analysis.

### **2.2.2 Bulk organic $\delta^{13}\text{C}_{\text{OM}}$ analysis**

Samples were subsampled from the same intervals as organic biomarkers for bulk organic carbon isotope  
analysis. Powdered sediment samples were weighed in silver capsules and carbonates were removed by  
170 adding 5% HCl in four-hour increments. Samples were analyzed on a Costech Elemental Analyzer  
coupled to a ThermoFinnigan Delta V Plus isotope ratio monitoring mass spectrometer (IRMS). Samples  
are reported as per mil (‰) deviations from the Vienna Pee Dee Belemnite (VPDB) standard in  
conventional delta notation.

## **2.3 GDGT preparation and analysis**

### **2.3.1 GDGT analysis**

Polar samples from Lake Magadi were analyzed for core lipid isoprenoid glycerol dialkyl glycerol  
tetraethers (iso-GDGTs) at the University of Massachusetts Amherst on an Agilent 1260 series high  
performance liquid chromatograph (HPLC; Fig. S1) in tandem with an Agilent 6120 series single  
quadrupole mass selective detector (MSD). Compounds were ionized using atmospheric pressure  
180 chemical ionization (APCI). The columns used for GDGT separation were a pre-column guard followed  
by two ultra-high performance liquid chromatography (UHPLC) silica columns (BEH HILIC, 2.1x150  
mm, 1.7  $\mu\text{m}$ , Waters) connected in series and kept at 30 °C. Elution solvents followed Hopmans et al.  
(2016) using a flow rate of 0.2 mL min<sup>-1</sup>. Two solvent mixtures, hexane (A) and 9:1 Hex:IPA (B), were  
eluted isocratically for 25 minutes with 18% B, a linear gradient to 35% B in 25 minutes, a second linear  
185 gradient to 100% B in 30 minutes.

### **2.3.2 GDGT indices**

Several different ratios based on the relative abundance of different isoprenoid GDGTs have been  
developed to determine their source(s). The methane index (MI) is defined by Zhang et al. (2011) and is  
calculated as in Eq. (1):

190

$$MI = \frac{GDGT-1+GDGT-2+GDGT-3}{GDGT-1+GDGT-2+GDGT-3+Cren+Cren'} \quad (1)$$

MI values range between 0 and 1 with values > 0.5 considered to be derived from methanotrophic communities and values < 0.3 considered normal sedimentary conditions (Zhang et al., 2011). These  
 195 proposed ranges from Zhang et al. (2011) were derived from GDGTs found in marine sediments, so the cutoff values for methanotrophy may differ in lacustrine sediments, particularly those in saline, alkaline environments.

The ratio of GDGT-2 / crenarchaeol (%GDGT-2/cren) also indicates methanotrophy (values > 0.2),  
 200 specifically methanotrophy associated with sulfate-methane transition zones (Weijers et al., 2011). These values were normalized and converted to percentages so that the numbers produced could be contextualized with the other indices used (Eq. 2). As a result, %GDGT-2/cren contributions greater than 33% will be considered methanotrophic signals.

$$205 \quad \%GDGT-2/cren = \frac{[GDGT-2]}{[GDGT-2]+[Cren]} * 100 \quad (2)$$

Methanogenic inputs are calculated similarly to Eq. 2 above using GDGT-0 in place of GDGT-2. Blaga et al. (2009) found that values of GDGT-0 / (GDGT-0 + cren) > 2 are associated with methanogenic  
 210 archaeal communities in a study of freshwater lakes. Similarly, in a study of Eocene marine sediments, Inglis et al. (2015) normalized the equation and converted it to a percentage, a convention we follow (Eq. 3). They found that contributions from methanogens were indicated by values greater than 67%.

$$\%GDGT-0/cren = \frac{[GDGT-0]}{[GDGT-0]+[Cren]} * 100 \quad (3)$$

215 The GDGT-2 / GDGT-3 ([2]/[3]) index ~~was~~is used here to describe both mesophilic environments as well as environments with high MI values. Rattanasriampaipong et al. (2022) found that differences in



[2]/[3] are linked to distinct archaeal communities whereby low values of [2]/[3] (ca. 0.55) are observed in thermophilic cultures while elevated values are indicative of hot spring mats (ca. 1.00), shallow aerobic ammonia-oxidizing archaea (AOA; ca. 1.16), or archaea in suspended particulate matter (ca. 2.52). This is the same version described in Rattanasriampaipong et al. (2022) (Eq. 4).

$$[2]/[3] = [GDGT - 2]/[GDGT - 3] \tag{4}$$

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2.4 Bulk geochemistry

Bulk geochemical data and core descriptions from Owen et al. (2019, 2024) were used to interpret hot spring influences in the intervals of focus in the sediment core (designated as intervals 1, 3, and 5 in the results and discussion sections)~~intervals of focus (i.e., Intervals 1, 3, and 5)~~. They attributed rare earth element (REE) anomalies to increased lake alkalinity, which reflected increased evaporation and the development of highly saline, alkaline lakes and possibly increased hydrothermal/fluvial inflow ratios. All statistical analyses were performed using the GraphPad Prism 10<sup>®</sup> software (<https://www.graphpad.com/>). Only the necessary data to determine relationships between the bulk geochemistry of ~~rare earth elements~~REEs (~~REEs~~; La, Ce, Nd, Sm, Eu, Yb, Lu) and methane indices (MI, %GDGT-0/cren, %GDGT-2/cren, and [2]/[3]) were imported. The data was tested for normality via the built-in "Normality and Lognormality Tests" function in Graph Pad<sup>®</sup>. Tests yielded lognormal distributions of each dataset and found the data to be non-normally distributed. Prior to performing a principal component analysis (PCA) or correlation matrix, the data needed to be tested for normality. To determine whether the data were normally distributed, the built-in "Normality and Lognormality Tests" function in GraphPad<sup>®</sup> yielded lognormal distributions of each dataset. The data were found to be non-normally distributed. Additionally, pyrite cubes were visually assessed without the use of a microscope during the initial core description and during organic extractions, however, no smear slides were collected and assessed.

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### 2.4.1 Principal component analysis (PCA)

For the PCA, the imported data were analyzed using the built-in PCA function in GraphPad®. The data were standardized, which scaled the data to a mean of 0 and a standard deviation of 1. The principal components were selected based on their eigenvalues using the Kaiser Rule, which selects eigenvalues greater than 1.0. Principal components 1 and 2 explained 58.0% of the variance in the data.

### 2.4.2 Correlation matrix

The correlation matrix was performed using the built-in function in GraphPad®. Because the data were non-normally distributed, the nonparametric Spearman correlation was chosen over the Pearson correlation. An r value was computed for every pair of Y datasets using the default two-tailed option at a 95% confidence interval.

## 3 Results

### 3.1 GDGT lipid variability

Samples are split into six intervals (1-6) based partly on their fractional abundances of GDGT-0 and cren as well as their MI values: (1) 35.67 to 32.61 m; ca. 17.7 to 14.9 ka, (2) 67.81 to 43.51 m; ca. 129 to 38.9 ka (3) 86.06 to 70.78 m; ca. 197 to 149 ka, (4) 96.38 to 94.91 m; ca. 318 to 315 ka, (5) 104.10 to 103.16 m; ca. 324 to 323 ka, and (6) 130.21 to 119.64 m; ca. 456 to 391 ka (Table S1).

In each of the intervals of the core, ~~Table S1~~ Table S1 MI, %GDGT-0/cren, and %GDGT-2/cren values oscillate between high and low values, changing abruptly from one interval to the next (Table S1). The methanotrophic (%GDGT-2/cren) and the methanogenic (%GDGT-0/cren) indices track similarly to MI values; that is, when values of MI are high, so are the other two indices. It should be noted that there are some large gaps in sampling between intervals in the core due to our sampling regime (i.e. targeting intervals with high apparent organic matter based on darker silty matrix). Interval 1 is characterized by a higher proportion of cren and lower overall index values. The %GDGT-0/cren index averages 36.3 % ( $\pm 0.09$  %) in this interval while the %GDGT-2/cren index averages 10.6 % ( $\pm 0.05$  %; Fig. 2). The MI in

this interval is correspondingly low with an average of 0.25 ( $\pm 0.07$ ), well below the MI = 0.5 cutoff range for methanotroph-impacted communities. As such, this interval could be used for the [2]/[3] index; values averaged 2.1 ( $\pm 1.02$ ). Interval 2 has much higher values for each of these indices, where the average  
270 %GDGT-0/cren = 99.3 % ( $\pm 0$  %) and the average %GDGT-2/cren = 93.6 % ( $\pm 0.04$  %). MI values in Interval 2 are also high with an average of 0.96 ( $\pm 0.02$ ). Of note, there is a large gap where no measurements were taken from 43.55 to 46.68 ~~15~~ m (~9.7 kyr) as well as from 50.36 to 58.74 m (~32.5 kyr). Interval 3 averages for %GDGT-0/cren and %GDGT-2/cren are 54.3 ( $\pm 0.27$  %) and 20.4 % ( $\pm 0.26$  %), respectively. However, there is one anomalously high value at 77.32 m with %GDGT-0/cren and  
275 %GDGT-2/cren values at 99.6 and 93.8 % and an MI = 0.96. Excluding the high index value, the averages were lowered to 48.6 ( $\pm 0.22$  %) and 11.2 % ( $\pm 0.05$  %) for the %GDGT-0/cren and %GDGT-2/cren values and the MI average was lowered from 0.33 ( $\pm 0.23$ ) to 0.26 ( $\pm 0.06$ ). With the exclusion of 77.32 m, the [2]/[3] index averaged 1.5 ( $\pm 0.80$ ) in this interval, lower than Interval 1. Interval 4 is characterized by high index values, with a similarly abrupt shift from low values. Averages of the %GDGT-0/cren and  
280 %GDGT-2/cren are 98.1 ( $\pm 0.04$  %) and 88.2 % ( $\pm 7.41$  %) and an average MI of 0.92 ( $\pm 0.04$ ); these average index values are similarly high as compared to Interval 2. Interval 5 is a shift to lower overall index values with averages of %GDGT-0/cren and %GDGT-2/cren at 40.1 ( $\pm 0.17$  %) and 9.2 % ( $\pm 0.03$  %) and an average MI of 0.22 ( $\pm 0.05$ ). Finally, Interval 6 shows a period in the core with high index values throughout. Averages of %GDGT-0/cren, %GDGT-2/cren, and MI are 97.6 % ( $\pm 0.03$  %), 89.4 %  
285 ( $\pm 0.08$  %), and 0.95 ( $\pm 0.05$ ), respectively.

### 3.2 Bulk $\delta^{13}\text{C}_{\text{OM}}$ values

~~Table S1~~ Bulk  $\delta^{13}\text{C}_{\text{OM}}$  values follow a similar pattern to the indices described in section 3.1, that is the values oscillate between high and low values between intervals. Samples in Interval 1 ranged from -21.9 to -16.8 ‰ and had an average  $\delta^{13}\text{C}_{\text{OM}}$  value of -18.4 ‰ with respect to VPDB. Interval 2 samples had  
290 the most  $^{13}\text{C}$ -depleted values in all sampled intervals, ranging from -89.4 to -24.7 ‰ with an average of -35.1 ‰ and excluding the three outlier values (-48.1, -64.2, and -89.4 ‰), the Interval 2 average was -28.2 ‰. In Interval 3, the  $\delta^{13}\text{C}_{\text{OM}}$  had a narrower range from -24.4 to -21.4 ‰ and an average of -22.5 ‰. A lighter signal from Interval 4 yielded a narrow range of values from -27.0 to -25.4 ‰ averaging -26.0

‰. Interval 5 had slightly heavier values ranging from -25.0 to -18.1 ‰ with an average of -22.1 ‰.

295 Lastly, Interval 6 had depleted  $\delta^{13}\text{C}_{\text{org}}$  values similar to intervals 2 and 4, with a range of -28.2 to -22.1  
‰ and an average of -25.2 ‰. Analytical reproducibility of duplicate runs was better than  $\pm 0.15$  ‰  
VPDB.

### 3.3 Bulk geochemistry

Both a PCA and correlation matrix were performed using the MI, Ca/Na, %GDGT-0/cren, %GDGT-  
300 2/cren, and [2]/[3] to compare to the REEs La, Ce, Nd, Sm, Eu, Tb, Yb, and Lu (Fig. 3). Increased values  
of REEs are characteristic of sodic systems influenced by hydrothermal springs, ~~namely such as~~ Mono  
Lake in California and this system (Johannesson and Lyons, 1994; Owen et al., 2019). Additionally, the  
Ca/Na is a proxy for the proportion of hydrothermal inflow to Lake Magadi such that a higher Ca/Na  
indicates more freshwater input, while a lower Ca/Na indicates a higher proportion of hydrothermal water.

305

A PCA (Fig. 4a) and non-parametric Spearman correlation matrix (Fig. 4b) were performed to quantify  
the relationship between REEs, MI, Ca/Na and [2]/[3]. The PCA showed that Ca/Na loaded positively on  
PC1 and PC2 and each of the methane indices loaded positively on PC1 and negatively on PC2. The REE  
La loaded positively on PC2 and negatively on PC1 while the REEs Ce, Nd, Sm, Eu, Yb, and Lu loaded  
310 negatively on PCs 1 and 2. This indicates a negative relationship between the negatively loaded REEs  
and a high Ca/Na. Similarly, the correlation matrix of REEs and methane indices showed a negative  
relationship between each index and REE, except for the relationship of [2]/[3] and Nd ( $r=0.02$ ) and  
%GDGT-2/cren ( $r=-0.04$ ), which showed no observable linear trend. The REEs and methane indices did  
not load on the same PC axis showing that there was also not a nonlinear trend associated with the REEs  
315 and methane indices.

## 4 Discussion

### 4.1 Lake Magadi archaeal community shifts

The abrupt changes in isoprenoid GDGT-based indices in the sediment record of Lake Magadi indicate  
shifts in the archaeal communities present (Fig. 2). Shifts between two distinct communities were inferred

320 using a combination of the Methane Index (MI) and ratios of GDGT-0 and GDGT-2 normalized to  
crenarchaeol (Eqs. 2 and 3). We denote these shifts as either “MI-on periods”, characterized by  $MI > 0.5$   
during intervals 2, 4, and 6, and “MI-off periods”, characterized by  $MI < 0.5$  during intervals 1, 3, and 5.  
Oscillations between these two states are discussed in detail in the following sections.

#### 4.1.1 MI-on periods

325 In Lake Magadi, during the MI-on periods (Fig. 2; Intervals 2, 4, and 6), the MI is persistently greater  
than 0.83 and displays more  $^{13}\text{C}$ -depleted  $\delta^{13}\text{C}_{\text{OM}}$  values compared to MI-off periods, indicating periods  
of enhanced methane cycling. AOM is a likely mode of methane cycling in Interval 6 as well as parts of  
Interval 2 because SRB and AOM archaea live in a consortium together at the sulfate methane transition  
zone, or SMTZ (Boetius et al., 2000; Hinrichs and Boetius, 2002; Werne et al., 2004). Thus, in intervals  
330 of the Magadi core where a SMTZ is suspected, such as in parts of Interval 2 and most of Intervals 4 and  
6, there should be an increase in indices related to methanotrophy such as high MI and %GDGT-2/cren  
(Weijers et al., 2011). Additionally, whereas methanogens and methanotrophs appear to be present in a  
consortium based on both the methane indices as well as bulk  $\delta^{13}\text{C}_{\text{OM}}$ , the majority of the contributions  
are coming from methanogens, as seen in the ternary plot in Fig. 5. This may seem counter-intuitive as  
335 the MI has been typically used to describe samples exhibiting a high predilection towards methanotrophy,  
but a high MI value does not necessarily exclude methanogenesis and conversely neither does a low MI,  
rather the low MI value suggests a predominance of Thermoproteota over Euryarchaeota (Zhang et al.,  
2011). High %GDGT-0/cren and %GDGT-2/cren index values in Intervals 2, 4, and 6 (Fig. 2) show that  
methanogenesis is co-occurring with AOM. The [2]/[3] index is also useful in understanding the  
340 proportion of methanotrophs in sediments, even in intervals with high MI values like those discussed  
herein (Table S1; Fig. 2). Values of the GDGT [2]/[3] ratio track nearly identically to the MI values (Fig.  
2), indicating that the MI is influenced by GDGT-2, which is characteristic of methanotrophs (Pancost et  
al., 2001; Schouten et al., 2003; Zhang et al., 2011).

345 Typically, methanogenesis in sulfate-rich systems is suppressed in favor of sulfate reduction caused by  
competition for both  $\text{H}_2$  and organic substrates (Fazi et al., 2021; Sorokin et al., 2015). However, reports

of methanogenesis co-occurring with SRB have been noted when methanogens are using non-competitive substrates such as methanol, or when sulfate levels are low (Oremland et al., 1982; Giani et al., 1984; Hoehler et al., 2001; Bebout et al., 2004; Arp et al., 2008, 2012; Jahnke et al., 2008; Smith et al., 2008; Robertson et al., 2009). Furthermore, pyrite cubes are common and scattered through the intervals where high index values are observed, indicating that there was a substrate for SRB, though it may have been in low concentration (Table S1). Thus, the combined evidence of pyrite in intervals with high GDGT-based indices (e.g. MI, %GDGT-0/cren, and %GDGT-2/cren [2]/[3]) indicates the presence of a SMTZ that supports AOM with the co-occurrence of methanogenesis.

Looking at Figure 45, it appears that GDGT-0 is the dominant GDGT compared to GDGT-2 and cren, indicating that this interval is likely methanogen-dominant rather than ANME dominant. Interval 2 (Figs. 3-2 and 5) of the Magadi core appears to be more influenced by methanogenesis than AOM, resulting from a more prevalent as seen in a high %GDGT-0/cren signal accompanied by a high %GDGT-2/cren signal, high [2]/[3] ratios, and a more  $^{13}\text{C}$ -depleted bulk  $\delta^{13}\text{C}_{\text{OM}}$  signal (average = -35.1 ‰; median = -28.6 ‰). Values of bulk  $\delta^{13}\text{C}_{\text{OM}}$  are similarly  $^{13}\text{C}$ -depleted in AOM-dominant Euryarchaeotal systems ranging from active mud volcanoes (~-27 ‰; ANME-1), a Danish freshwater lake (average ~-29.7‰; ANME-2), and the Sea of Galilee (~-30 ‰; ANME-2) in Israel (Lee et al., 2018; Norði et al., 2013; Sivan et al., 2011). At points where the bulk  $\delta^{13}\text{C}_{\text{OM}}$  values are at their lowest (e.g., -89 ‰), they are accompanied by a lower %GDGT-2/cren at ca. 95 % and an elevated %GDGT-0/cren at > 99.5 %. This aligns with the literature as Summons et al. (1998) reported  $\delta^{13}\text{C}_{\text{OM}}$  values between -53.4 and -48.7 ‰ in the total lipid extract of methylotrophic methanogens using non-competitive substrates in anoxic hypersaline environments. Furthermore, as these waters are typically sulfate-limited, acetoclastic and/or hydrogenotrophic methanogenesis is likely dominant when evidence for SRB is lacking (i.e., pyrite). Zhuang et al. (2016) performed compound-specific isotope analysis on several archaeol compounds from the Orca Basin and found archaeol and hydroxyarchaeol using  $\text{H}_2$  or  $\text{CO}_2$  (diagnostic of methanogens and methanotrophs) were relatively depleted (ca. -80 to -60 ‰) compared to the bulk  $^{13}\text{C}_{\text{OM}}$  (ca. -22 ‰). Zhuang et al. (2016) concluded that acetoclastic and/or hydrogenotrophic methanogenesis was unlikely due to high  $\text{SO}_4^{2-}$  concentrations in the Orca Basin, which may be the case in Lake Magadi. In Interval 2,

375 there is no evidence of visible pyrite cubes and we did not have a priority at the time of sampling to check  
a thin section of each sample for smaller pyrite aggregates. This indicates that other Euryarchaeotal  
communities may have different forms of AOM occurring in the sediments. These other forms of AOM  
include nitrate/nitrite reduction and iron coupled to AOM (Zandt et al., 2018). This is further bolstered  
by the evidence outlined by Kim and Zhang (2023) that not only quantitatively linked AOM to high MI  
380 values, but also to non-Group I ANME Euryarchaea because other non-GDGT producing ANME (e.g.  
ANME-2 and ANME-3) were shown to co-exist with Group I ANME. In the intervals that are missing  
pyrite (i.e., most of Interval 2; see Table S1; Ferland, 2017), the pyrite may have either been too small to  
see with the naked eye or the excess H<sub>2</sub>S could have been incorporated into the kerogen by reacting with  
labile organic matter. From 59.40 to 58.80 m, values of the bulk  $\delta^{13}\text{C}_{\text{OM}}$  dip as low as -89.4 ‰ (Table S1;  
385 Fig. 2), which aligns well with methanogenic archaeal biomass (Norði et al., 2013). However, as discussed  
above there is likely acetoclastic and/or hydrogenotrophic methanogenesis co-occurring in these high  
index intervals and is likely the dominant process where sulfate-dependent AOM is absent, and the  
sulfate-dependent AOM is likely replaced by coupling to either nitrate/nitrite or iron reduction.

390 Samples in Interval 4 (Table S1) of the Magadi core have high index values but no evidence for sulfate-  
dependent methanotrophy except for high MI values. This interval is thus interpreted as being  
methanogenic rather than methanotrophic. The abundance of pyrite in the four samples with low MI  
values (Table S1; 104.10 to 103.16 m), indicates sulfate reduction not linked to AOM. This is not observed  
in any other level of the core and a hypothesized series of reactions is described below, which may be  
395 linked to an abundance of SRB, anaerobic ammonium oxidizing (anammox) bacteria, and  
Thermoproteota (AOA) in the overlying water column. Due to periodic influxes of freshwater in Magadi,  
in addition to a permanent meromixis present in almost all samples [in this study](#) post 380 ka, the water  
column would have been oxic in the upper portion and anoxic below the chemocline. Freshwater pulses  
would have also brought nutrients to the lake such as ammonia (NH<sub>4</sub><sup>+</sup>) and sulfate (SO<sub>4</sub><sup>2-</sup>). The oxic  
400 portion of the water column would have supported microaerophilic AOA that oxidize NH<sub>4</sub><sup>+</sup> to nitrite  
(NO<sub>2</sub><sup>-</sup>), which is then transported to the anoxic part of the water column (Straka et al., 2019). Here,  
anammox bacteria use excess NH<sub>4</sub><sup>+</sup> and NO<sub>2</sub><sup>-</sup> from the AOA and convert these to N<sub>2</sub>. Excess SO<sub>4</sub><sup>2-</sup> is

simultaneously being used by SRB, creating  $\text{HS}^-$  that is reacting with iron species in the sediments and being buried as pyrite. Ladderane lipids characteristic of marine annamox bacteria (Jetten et al., 2009) were not studied in Magadi sediments. However, there is both 16S rRNA and lipid evidence for the production of ladderanes in hot springs in the western United States suggesting that annamox bacteria can persist in hot spring environments (Jaeschke et al., 2009). Additionally, Kambura et al. (2016) found evidence for *Planctomycetes* in both microbial mat and water samples surrounding the hot springs of Lake Magadi, lending credence to the hypothesis of AOA persisting in Lake Magadi. Without other lines of evidence, however, these are hypothetical reactions for explaining excess pyrite in the sediments without accompanying MI values. Nonetheless, this explanation has merit because of the high relative abundance of both crenarchaeol and cren'.

In nearly all of Interval 6 (Table S1), there is evidence for a higher proportion of methanotrophic archaea from 128.74 to 119.64 m (increased %GDGT-2/cren and [2]/[3]) and methanogenesis in the intervals from 130.21 to 129.77 m (Table S1; higher %GDGT-0/cren compared to %GDGT-2/cren). Samples from 123.43 to 119.64 (Table S1) are of note because the [2]/[3] values are lower than the MI values whereas every other MI and [2]/[3] values align nearly 1:1. This is likely due to GDGT-2 not being the dominant control of the MI, and while both %GDGT-0/cren and %GDGT-2/cren are equally high, there may be other factors in the water column exporting GDGT-2 to the sediments, possibly from deep-dwelling Group I.1b Thermoproteota, although this is unlikely due to limitations of depth (Taylor et al., 2013). The 656 m paleoshoreline reported by Casanova (1986, 1987) and Casanova and Hillaire-Marcel (1987) would imply a maximum water depth of ~ 50 m during the Late Pleistocene (African Humid Period: AHP) based on present topography. However, sedimentary evidence for such a high paleoshoreline is not seen throughout the Magadi Basin. Earlier water depths are also unclear because accommodation space was changing as the axial rift developed with faulting and subsidence (Owen et al., 2024). This is not deep enough (> 1 km) to support Group I.1b Thermoproteota per the constraints outlined in Taylor et al. (2013).



#### 4.1.2 MI-off periods

430 In intervals characterized by low MI, %GDGT-0/cren and %GDGT-2/cren values (Fig. 2; MI-off intervals are odd numbers highlighted in blue), the  $\delta^{13}\text{C}_{\text{COM}}$  values are  $^{13}\text{C}$ -enriched relative to those intervals characterized by higher index values (Fig. 2; MI-on; Fig. 2). Since the methane cycling indices (%GDGT-0/cren and %GDGT-2/cren) are both predominantly influenced by the availability of crenarchaeol, MI-off periods are marked by increased production in crenarchaeol. Typically, crenarchaeol is produced in  
435 open ocean systems, freshwater lakes, and soils by the mesotrophic aerobic ammonium oxidizing phylum Nitrososphaerota. However, they can also be found in other environments, such as [in](#) hot spring mats ~~in~~ [made by](#) Thermoproteota (Pearson et al., 2004, 2008; Schouten et al., 2013). As Thermoproteota require oxygen to oxidize ammonium to nitrate, the increased presence of crenarchaeol in the MI-off intervals, therefore, suggests periods when conditions were more oxic, at least in the upper water column. The  
440 increase in crenarchaeol, as well as the low [2]/[3] index values, suggest that more Thermoproteota were present in Lake Magadi in those periods. As mentioned in Section 4.1.1, three groups of AOA are of interest for interpreting which archaeal groups are found in low index intervals of Lake Magadi. Averages of [2]/[3] from the global dataset in Rattanasriampaipong et al. (2022) are as follows: hot spring mats (avg. = 1.00), shallow AOA cultures (avg. = 1.16), and shallow core tops (avg. = 2.64). Placing these on  
445 a continuum, we can approximate the environment from [2]/[3] averages in Magadi, though it should be noted that the shallow AOA and shallow core-top values in Rattanasriampaipong et al. (2022) are based on marine core-tops, while the hot spring mats are based on terrestrial hot springs like those observed around Lake Magadi (i.e. pH > 6.5).

450 Interval 1 captures a transition from a more arid East Africa to a wetter period at the onset of the African Humid Period (AHP). During wetter periods, more allochthonous material is transported to the lake, which includes vegetal remains that impact the overall bulk  $\delta^{13}\text{C}_{\text{COM}}$  values. This allochthonous vegetation enriches the overall bulk  $\delta^{13}\text{C}_{\text{COM}}$  values more significantly than other intervals in the Magadi core. Average values of bulk  $\delta^{13}\text{C}_{\text{COM}}$  are -17.7 ‰ in Interval 1, which correspond to the  $\delta^{13}\text{C}_{\text{COM}}$  values of  
455 aquatic sedges mixed with a terrestrial signal of grassy woodland (Sikes, 1994; Reiffarth et al., 2016).

Pollen records in Lake Magadi indicate that a mixture of C4 grassy woodlands and C4 aquatic sedges were predominant in the landscape that surrounded Lake Magadi at this time (Muiruri et al., 2021). In agreement with the pollen record, the  $\delta^{13}\text{C}_{\text{OM}}$  values likely record a mixture of C4 grasses and C4 sedges. Similar  $\delta^{13}\text{C}$  values were reported in  $\text{C}_{21}$  to  $\text{C}_{33}$  n-alkanes in equatorial regions of Cameroon, ranging from -18.2 to -17.6‰ and recording the signals from C4 grasses and sedges (Garcin et al., 2014). Supporting the pollen record, the  $\delta^{13}\text{C}_{\text{OM}}$  values are likely reflecting  $\delta^{13}\text{C}$  values similar to those observed by Garcin et al. (2014) in equatorial regions of Cameroon. The bulk  $\delta^{13}\text{C}_{\text{OM}}$  is likely recording a mixture of C4 grasses and C4 sedges similar to  $\delta^{13}\text{C}$  values of  $\text{C}_{27}$  to  $\text{C}_{33}$  n-alkanes obtained from C4 grasses and sedges in Cameroon which ranged from -18.2 to -17.6 ‰ (Garcin et al., 2014). This all suggests that the bulk  $\delta^{13}\text{C}_{\text{OM}}$  signal is dominated by terrestrial biomass, unlike other sections of the core, and there does not appear to be a significant influence from the benthic microbial community (i.e., methane cyclers or SRB).

Values of the [2]/[3] index average 2.1 in Interval 1 with some values as high as 3.74 and 4.63 at 33.28 and 33.03 m, respectively (Table S1). The higher values are closer to what is captured from deep oceanic suspended particulate matter (SPM) and deep ocean core-tops below the pycnocline, though caution should be used when comparing lacustrine and oceanic sediments (Rattanasriampaipong et al., 2022). The increase in %GDGT-0/cren (50.6 and 54.3 %; Table S1) and the slightly increased MI values (0.37 and 0.41; Table S1) imply that these samples were deposited in a deeper lacustrine environment. Evidence for a deeper paleolake at ~40 feet (~17–18 m) above the modern lakeshore (Baker, 1958) is also observed in the High Magadi Formation (ca. 17.7 to 10.8 ka) indicating that there was fresh water flowing into the lake during the period of deposition in Interval 1, likely creating a fresher water cap on the meromictic Lake Magadi (Barker et al., 1991; Behr, 2002; Owen et al., 2019). However, excluding the high [2]/[3] index values in Interval 1, the average is 1.6, which is closer to the hot spring mats and shallow AOA cultures (Rattanasriampaipong et al., 2022). Likely, the higher [2]/[3] index values represent periods of increased methanogenesis occurring in the sediments, with AOA input from the upper water column likely induced by proportional increases in the amount of hydrothermal inflow to the lake (Section 4.2.1). Fig. 4 shows that the Ca/Na is anti-correlated with REEs in both the PCA and correlation matrix. Since the

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proportion of Ca/Na decreases when REEs increase, we can say that statistically, when it is drier (and  
 485 thus proportionately more hydrothermally influenced) the Ca/Na decreases, REE values increase, and the  
 methane indices are suppressed. In the periods of lower [2]/[3] values, the community is interpreted as  
 being dominated by AOA and thermophilic AOA cultures (i.e., Thermoproteota; Rattanasriampaipong et  
 al., 2022) and is further supported by high % cren and % cren'. Kumar et al. (2019) described similarly  
 low [2]/[3] values in the water column of Lake Malawi that are akin to values observed in Lake Magadi  
 490 in both Intervals 1 and 3. They found that values of a lower normalized [2] / [2+3], ranging from 0.55 to  
 0.59, in Lake Malawi were associated with the shallower Thermoproteota (Thaumarchaeota) Group I.1b.  
 This is compared to higher values of [2] / [2+3] in the deeper dwelling Thermoproteota Group I.1a, which  
 means that most samples in Interval 1 are likely sourced from Group I.1b (Kumar et al., 2019). Kumar et  
 al. (2019) concluded that Group I.1b Thermoproteota were contributing to the lower [2] / [2+3] values,  
 495 while the more deeply dwelling Group I.1a Thermoproteota were more prevalent in aphotic portions of  
 the water column (Kumar et al., 2019). The normalized [2] / [2+3] used by Kumar et al. (2019), with  
 values ranging from 0.55 to 0.65, approximates values of [2]/[3] in the 1.30 to 1.65 range as described in  
 this paper. More recently, Baxter et al. (2021) found that Thermoproteota I.1b are more prevalent in the  
 upper oxygenated portion of the water column within the photic zone as evidenced by a higher relative  
 500 abundance of crenarchaeol and lower relative abundance of GDGT-2. Thus, our interpretations of  
 thaumarchaeotal AOA in Lake Magadi sediments are consistent with data from Baxter et al. (2021) and  
 Kumar et al. (2019). This interpretation is consistent with Interval 1 being a period of proportionately  
 more freshwater and HCO<sub>3</sub><sup>-</sup>-rich hydrothermal input and a deeper lake overall, which would explain the  
 accompanying increase in crenarchaeol.

505 Interval 3 [2]/[3] averages are lower overall (Table S1; avg. = 5.4), with only one outlying high value (ca.  
 77.32 m at a value of 36.7). Excluding this high index value, the [2]/[3] average drops to 1.5, which is  
 closer to what is observed in shallow AOA cultures and hot spring mats. With most samples being closer  
 to unity (i.e., [2]/[3] = 1.0), it is likely that hot springs had a greater influence on the community  
 510 composition in these intervals. Samples that are closer to unity (70.78, 70.86, and 71.08 to 75.93 m) also  
 have a relatively <sup>13</sup>C-enriched δ<sup>13</sup>C<sub>COM</sub> values (avg. = -21.8‰) compared to samples with a higher [2]/[3].

~~(averaging 1.5 excluding the outlying value of 36.7). This average is closer to shallow Group I.1a Thermoproteota as described previously.~~ Average isotope values in Interval 3 are between oceanic hydrothermal vents (avg. = -19.0 ‰) and terrestrial alkaline hot spring systems such as the Bison Pool hot spring in Yellowstone National Park (avg. = -24.9 ‰) (Shah et al., 2008; Schubotz et al., 2013). Since elevated amounts of GDGT-2 (i.e., relative abundance > 45%) are associated with Euryarchaeota, and values in Intervals 1, 3, and 5 are much lower than 45% (Table S1), these intervals are likely dominated by Thermoproteotal AOA (Pancost et al., 2001; Turich et al., 2007; Taylor et al., 2013). Archaeal community composition in Intervals 1, 3, and 5 is independent of these external factors and is related to hydrothermal flows. This further supports the hot springs driving the lake archaeal community composition as there was less overall precipitation and the Thermoproteotal communities were more abundant during Intervals 1, 3, and 5.

Lastly, Interval 5, which only has 4 samples, has similarly low values of [2]/[3] (average = 1.4) like Intervals 1 and 3, which is likely indicative of Thermoproteotal AOA cultures. ~~The  $CPI_{alk}$  and  $CPI_{FA}$  averages were 4.6 and 5.0, indicating more terrestrial input. So, while these values are lower than Interval 3, and closer to the values in Interval 1, these still indicate a higher terrestrial input during this timeframe.~~

#### 4.2 The influence of hot spring/runoff ratios on the archaeal methane cycles

Hydrothermal fluids in the basin are rich in carbonate and bicarbonate as well as  $Na^+$  ions, inferred to be a result of the weathering and alteration of trachytic (silica-rich) basement rock (Jones et al., 1977; Allen et al., 1989), and mantle-derived  $CO_2$  discharged mainly along faults (Lee et al., 2017; Muirhead et al.).  $Ca^{2+}$  and  $Mg^{2+}$  are also very low (Deocampo and Renaut, 2016). Renaut and Owen (2023) note that hydrothermal waters become important contributors to lake recharge during arid phases when fluvial inflow declines. For example, Nasikie Engida, a small hypersaline lake northwest of Lake Magadi, is partly maintained today by hydrothermal inputs, with trona, nahcolite and zeolites accumulating during dry periods when there is little or no fluvial inflow (DeCort et al., 2019; Renaut et al., 2020; Renaut and Owen, 2023). Magadi Core MAG14-2A lacks evidence for complete desiccation (e.g., mudcracks, soils, calcrete) and the lake appears to have retained surface waters through multiple drought episodes during

the last million years (Owen et al., 2019). In contrast, separate lakes in the neighbouring Koora Basin  
540 (~10 km to the east), dried out many times, leading to soil formation in that basin (Owen et al., 2024).

Owen et al. (2019) also noted that during periods when highly saline, alkaline water dominated at Magadi,  
ash was zeolitized, REE patterns developed significant anomalies, and pyrite developed in anoxic/euxinic  
bottom waters of a meromictic lake. They also noted the excellent preservation of diatom opaline silica  
545 in highly alkaline lakes after about 540 ka, which suggests that very high levels of silica in lake water  
preserved their frustules from dissolution. Hydrothermal springs at Magadi today contain high silica, but  
their waters require further evaporation to achieve concentrations that would preserve diatom silica under  
very high pH conditions. High silica concentrations brought about by strong evaporation in a lake  
maintained by spring inflows (meteoric or hydrothermal) may partly explain the abundant chert through  
550 the Magadi sedimentary sequence.

Proportional increases in hot-spring water during periods of increased aridity would have favoured the  
development of highly saline, alkaline waters with significant impacts on archaeal communities. Samples  
in Interval 6 represent a drier period when the lake area and volume had shrunk, partly due to tectonic  
555 influences (Owen et al., 2024), and lake floor anoxia was prevalent (Owen et al., 2019). Between ~380  
and 105 ka (Fig. 2; Interval 5 through mid-Interval 2), the paleolake was frequently meromictic with a  
freshwater mixolimnion that supported freshwater planktonic diatoms while the saline monimolimnion  
and lake floor waters favoured alteration of ash to a variety of zeolites (e.g., erionite, phillipsite,  
clinoptilolite, analcime) (Owen et al., 2019). Similarly, from ~105 to 0 ka (Fig. 2; mid-Interval 2 through  
560 Interval 1) low Ca/Na, increased Br, and the abundant zeolite formation, indicates saline conditions.  
After about 80 ka, tectonic adjustments and increasing aridity led to desiccation in the Koora Basin  
suggesting that spring inflows were important in maintaining a hypersaline lake in the Magadi Basin, as  
they do today during dry seasons (Owen et al., 2019). Ca/Na ratios have steadily decreased over time,  
though not at a 1:1 rate, which aligns with methane index values and sudden increases of these indices.  
565 Fig. 4 shows that the Ca/Na and methane indices are statistically different from one another in both the  
correlation matrix and PCA plot. In Fig. 5b4b, Ca/Na is loaded positively on PC1 and PC2, while the

methane indices ([2]/[3], MI, %0/Cren, and %2/Cren) are loaded positively on PC1 and negatively on PC2. Furthermore, REE data also appear to reflect our MI-off and MI-on periods as the REEs are anticorrelated in the correlation matrix (Fig. 5a4b) and loaded on different PC axes (Fig. 5b4a).  
570 Consequently, we interpret the changes in salinity and alkalinity in the Magadi paleolakes as reflecting the impact of climate on spring/runoff ratios into the Magadi Basin, which in turn have exerted significant impact on the archaeal communities through the last million years. Samples in the low-MI intervals (ca. 32.61–35.67, 70.78–75.93, and 103.2–104.1 m) likely reflect proportionally increased spring/runoff ratios at Magadi caused by increased evaporation and decreased precipitation in the surrounding  
575 landscape.

## 5. Conclusions

Sediments in Lake Magadi track the environmentally driven changes in archaeal communities over the past ~ 456 ka. Using the MI to track the predominantly archaeal inputs at Lake Magadi, we have observed sudden and distinct shifts between mixed communities of Euryarchaeotal methanogens and  
580 methanotrophs transitioning to mesophilic AOA Thermoproteota communities and back again. This shift is driven, in part, by moisture balances in the East African Rift, with wetter conditions periodically causing freshwater floods into a saline lake to form a meromictic waterbody at Magadi, and with more archaea derived from the upper water column rather than the sediments, as evidenced by low MI, low [2]/[3], and relatively  $^{13}\text{C}$ -enriched bulk  $\delta^{13}\text{C}_{\text{OM}}$ . Methane indices were typically higher during periods of  
585 reduced hydrothermal activity, indicating more Euryarchaeal communities, whereas Thermoproteota communities thrived during periods of higher hydrothermal activity. This is a clear relationship between low MI values, spring/runoff ratios, lake salinity, alkalinity and the development of mesophilic Thermoproteota. This study is one of the first to examine methane cycling in a soda lake over geologic time and provides valuable insights into how variable these systems can be. Soda lakes are important  
590 ecosystems for methane cyclers and should be studied more closely so that we can improve understanding of global methane contributions in the past, and constrain sources in the future.

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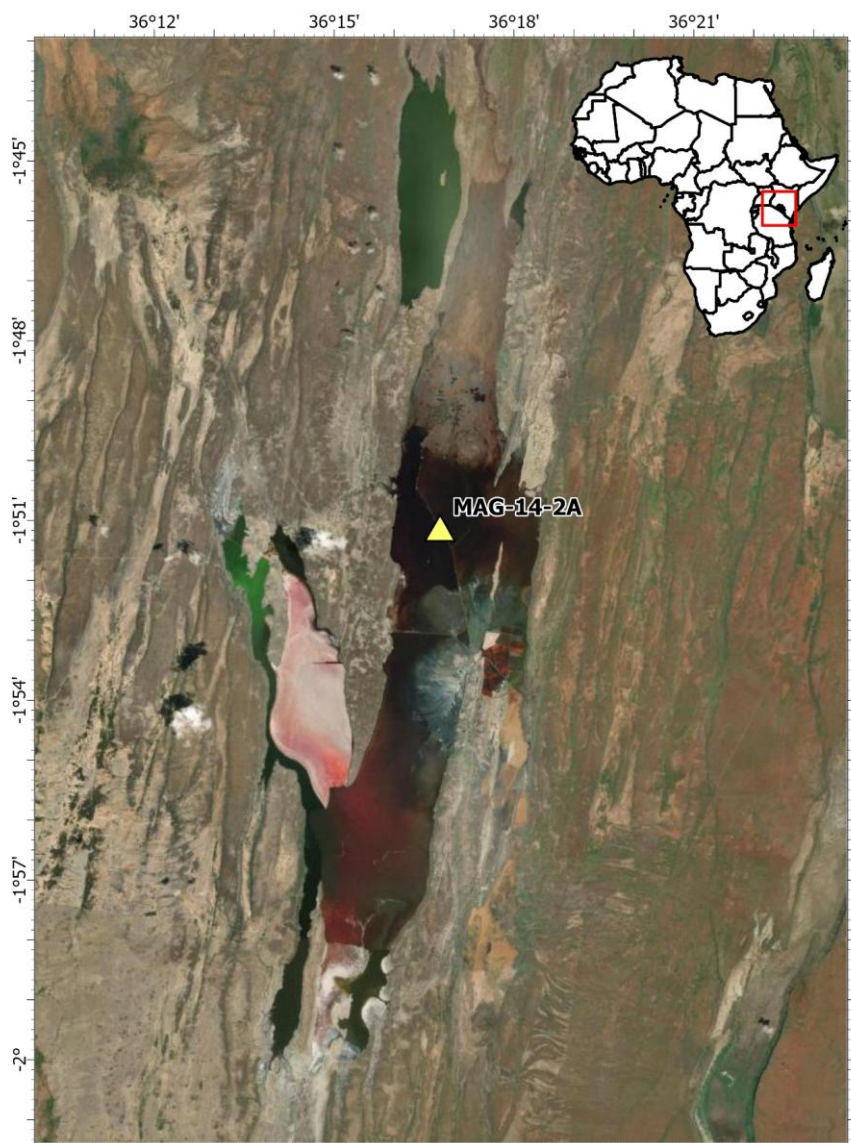
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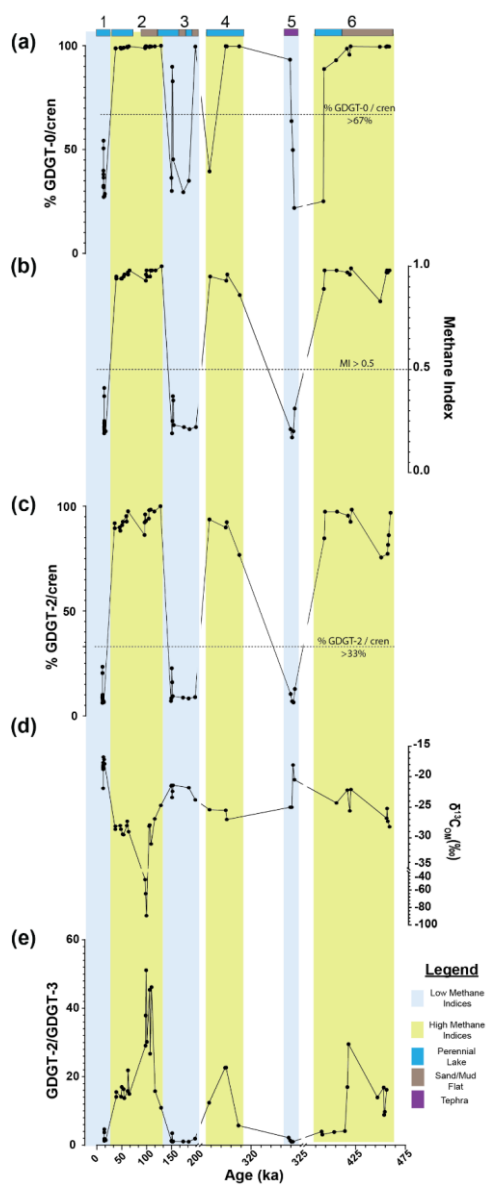
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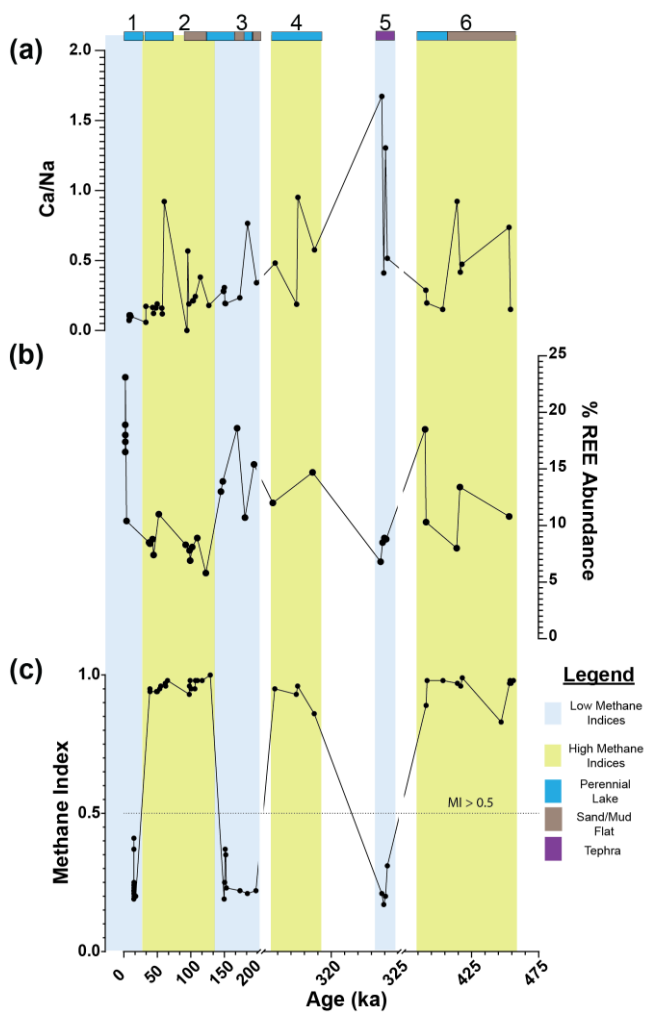


**Figure 1.** Map of the drilling location of MAG-14-2A (yellow triangle) in Lake Magadi for the Hominin Sites and Paleolakes Drilling Project (HSPDP).



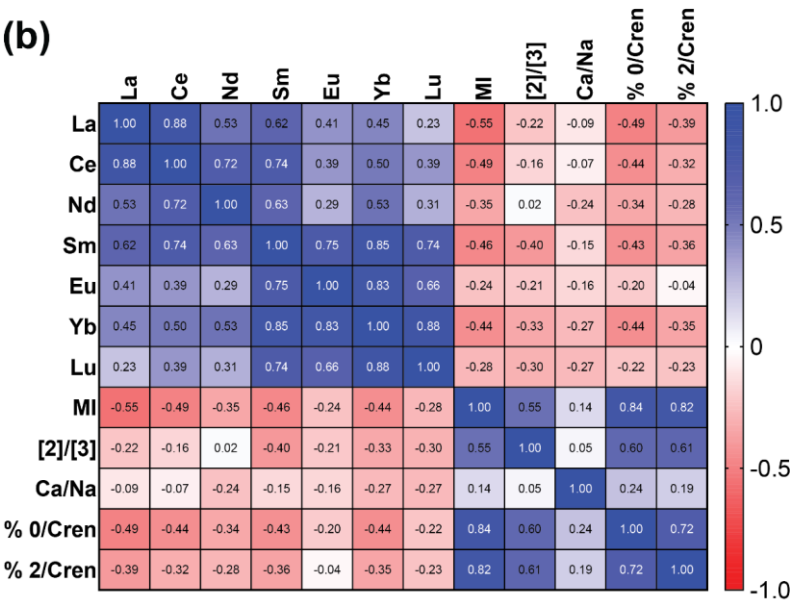
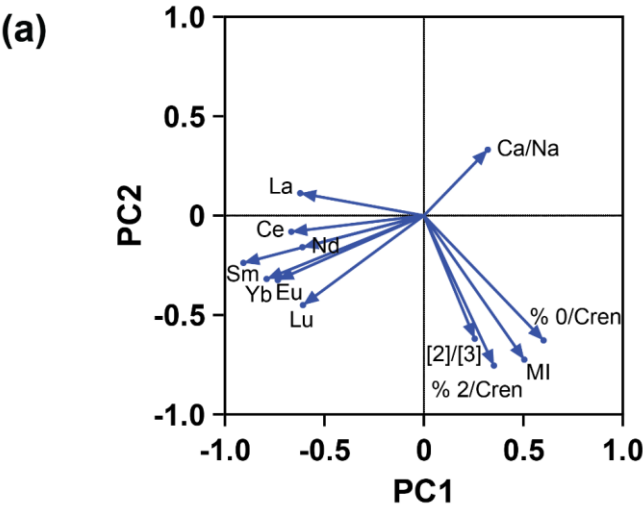


**Figure 2.** Downcore variations in Lake Magadi of the **a)** %  $\delta^{13}\text{C}_{\text{org}}$ , **b)** MI, **c)** %  $\delta^{13}\text{C}_{\text{org}}$ , **d)** bulk  $\delta^{13}\text{C}_{\text{org}}$ , and **e)** the GDGT-2/GDGT-3 ([2]/[3]) values from ca. 14.9 to 456 ka. Sections 1, 3, and 5 are low MI intervals outlined in blue, the high MI intervals in Sections 2, 4, and 6 are in yellow. Checkered patterns indicate periods of tuffaceous silt deposit, which align with the low MI intervals. Bands at the top of the graph indicate the inferred (via Renaut and Owen, 2023) lake levels and major inputs with dark blue indicating a perennial lake, brown indicating a sand or mud flat, and purple indicating tephra. Dotted lines on each section denote the cut-off points for methane related indices MI ( $>0.5$ ), %GDGT-2/cren ( $>33\%$ ), and %GDGT-0 / cren ( $>67\%$ ). See Section 2.3.2 for more details. Note the breaks in the X-axis scale.

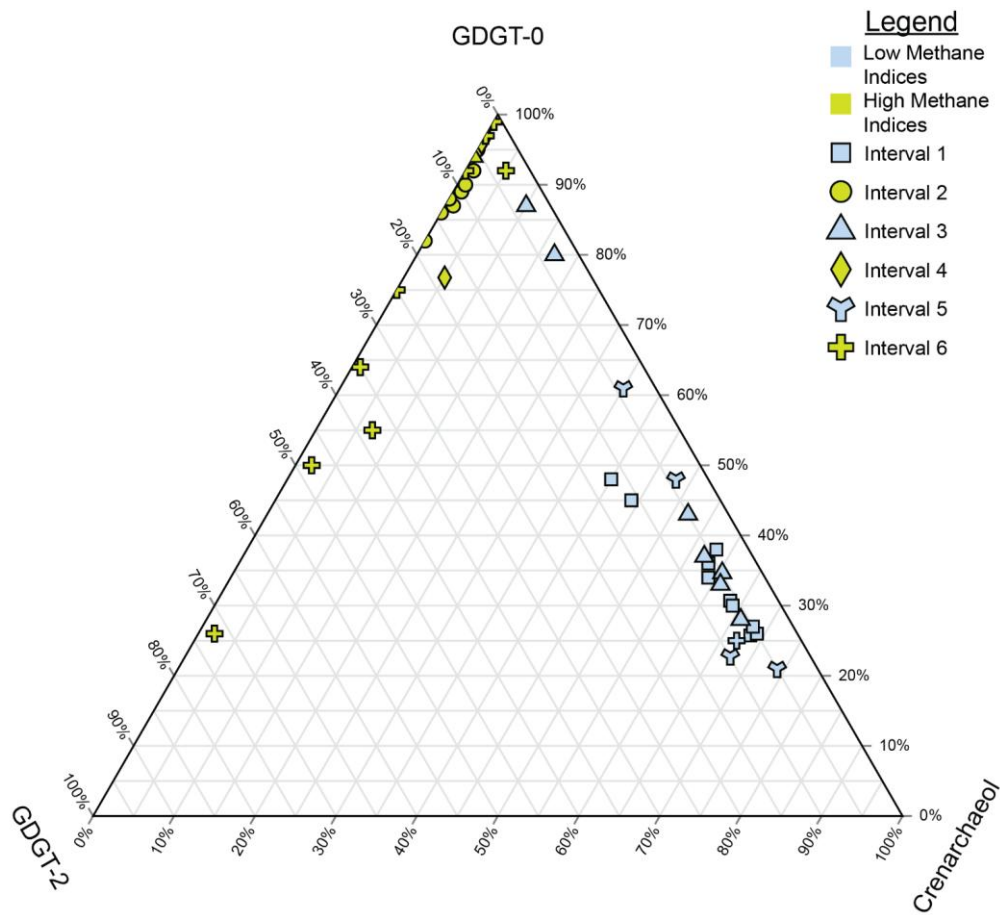


**Figure 3.** Downcore plot for Lake Magadi of **a)** Ca/Na, **b)** %-REE abundance, and **c)** MI. Values range from ca. 14.9 to 456 ka and Sections 1, 3, and 5 are outlined in blue reflecting a low MI interval, while high MI intervals are outlined in yellow. ~~The checkered pattern is indicative of periods of higher inferred hydrothermal flow.~~ Bands at the top of the graph indicate the inferred (via Renault and Owen, 2023) lake

levels and major inputs with dark blue indicating a perennial lake, brown indicating a sand or mud flat, and purple indicating tephra. The dotted line on the MI plot (c) denotes the cutoff point  $>-0.5$  for values significantly affected by methane cycling archaea. Note the breaks in the X-axis scale and in the Y-axis scale for the  $\delta^{13}\text{C}_{\text{OM}}$ . REE values are from Owen et al. (2019).



**Figure 4.** Both **a)** PCA and **b)** Spearman Correlation Matrix showing the relationship between methane related indices (MI and [2] / [3]) and REEs (La, Ce, Nd, Sm, Eu, Tb, Yb, and Lu) in the sampled intervals of the core. A negative relationship is seen between the methane indices and REEs as shown by opposing eigenvectors on the PCA (**a**) and negative r values on the correlation matrix (**b**). REE values are from Owen et al. (2019).



1025 **Figure 5.** Ternary plot of crenarchaeol, GDGT-0, and GDGT-2, which are used to calculate the methane  
indices. Samples are split by both their interval (denoted by their shape) and whether they are from a high  
MI (yellow) or low MI (blue) interval. Higher proportions of GDGT-0 indicate methanogenic inputs,  
higher GDGT-2 indicate methanotrophy, and higher crenarchaeol indicates more mesophilic conditions  
influenced by hot springs.

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