Surface Methane Concentrations Along the Mid-Atlantic Bight Driven by Aerobic Subsurface Production Rather Than Seafloor Gas Seeps

Mihai Leonte1, Carolyn D. Ruppel1, Angel Ruiz-Angulo2, and John D. Kessler1,3

1Department of Earth and Environmental Sciences, University of Rochester, Rochester, NY, USA, 2U.S. Geological Survey, Woods Hole, MA, USA, 3Icelandic Meteorological Office, Reykjavik, Iceland

Abstract  Relatively minor amounts of methane, a potent greenhouse gas, are currently emitted from the oceans to the atmosphere, but such methane emissions have been hypothesized to increase as oceans warm. Here, we investigate the source, distribution, and fate of methane released from the upper continental slope of the U.S. Mid-Atlantic Bight, where hundreds of gas seeps have been discovered between the shelf break and ~1,600 m water depth. Using physical, chemical, and isotopic analyses, we identify two main sources of methane in the water column: seafloor gas seeps and in situ aerobic methane oxidation, the dominant methane sink in this region, with the average fraction of methane oxidized being 50%. Due to methane oxidation in the deeper water column, below 200 m depth, surface concentrations of methane are influenced more by methane sources found near the surface (0–10 m depth) and in the subsurface (10–200 m depth), rather than seafloor emissions at greater depths.

Plain Language Summary  Methane is a greenhouse gas that is second only to carbon dioxide in its contribution toward modern global warming. Here we investigate the mechanisms responsible for adding and removing methane from ocean waters along the Mid-Atlantic Bight, since future climate change is suspected to increase the release and/or production of methane into the oceans. Our study shows that methane was released at the seafloor from gas seeps in the form of bubbles and produced in surface and subsurface waters. Regardless of the source, methane was observed to be actively removed from these ocean waters through microbial oxidation, helping to limit atmospheric emission, especially for methane originating at greater depths.

1. Introduction

The increase in radiative forcing due to rising methane (CH₄) concentrations during the Anthropocene is second only to that of carbon dioxide (CO₂), making CH₄ a driver of ongoing and future climate change (Ciais et al., 2013). Although the global annual emission of CH₄ from the oceans to the atmosphere has been estimated to be relatively small (6–12 Tg; Weber et al., 2019), future ocean warming has the potential to increase the input of benthic CH₄ to the water column and possibly the atmosphere (Hovland et al., 1993; Ruppel & Kessler, 2017; Saunois et al., 2016).

Methane sources into seawater include diffusive and ebullitive emissions of microbially derived and thermogenically derived CH₄ from the seafloor as well as aerobic in situ production in the oceanic water column (Reeburgh, 2007). Active gas seeps are responsible for high CH₄ fluxes in waters where bubbles are emitted from the seafloor and through the water column traversed by the bubble plume (Heeschen et al., 2003; Leonte et al., 2018; McGinnis et al., 2006; Römer et al., 2012). The vertical distance that CH₄ can be transported via gas bubbles depends on several factors, including seafloor depth and the bubble diameter at the seafloor (McGinnis et al., 2006; Rehder et al., 2009; Wang et al., 2016). However, only in rare cases is CH₄ emitted from the seafloor in gas bubbles likely to reach the sea-air interface (Ruppel & Kessler, 2017). Instead, CH₄ diffuses out of bubbles and dissolves into the water column relatively quickly, being replaced by other gases inside the bubble (Leonte et al., 2018; McGinnis et al., 2006; Rehder et al., 2009; Wang et al., 2016). Further
vertical transport of the dissolved CH$_4$ occurs at significantly slower rates than those associated with buoyant bubble rise.

Apart from seafloor sediments, CH$_4$ is also produced in the water column through aerobic methanogenesis. Several studies investigating dissolved CH$_4$ in open ocean environments have noted a subsurface CH$_4$ concentration maximum between 100 and 200 m depths in the water column (e.g., Holmes et al., 2000). However, since methanogenesis has traditionally been considered a strictly anaerobic process, the apparent production of CH$_4$ in oxygenated subsurface waters was termed the “Ocean Methane Paradox” (Holmes et al., 2000; Kiene, 1991; Reeburgh, 2007). Previous studies have examined the fecal pellets formed by zooplankton or other marine organisms as suitable microenvironments where anaerobic conditions, and thus methanogenesis, might occur (Nihous & Masutani, 2006; Oremland, 1979). More recent studies have identified several pathways for aerobic CH$_4$ production to occur including the cycling of dimethylsulfoniopropionate and the degradation of phosphonates commonly found as part of dissolved organic matter (Damm et al., 2008; Karl et al., 2008; Repeta et al., 2016). Regardless of the source, CH$_4$ concentrations at any depth may be decreased by mixing with lower concentration waters and by microbial oxidation (Leonte et al., 2017; Mau et al., 2012). Since these sink terms appear ubiquitous throughout the water column (e.g., Mau et al., 2012), sources of CH$_4$ at shallower water depths have a greater potential to contribute CH$_4$ to the atmosphere.

Here, our objectives were to quantify the fate of seafloor-released CH$_4$ and determine the relative contribution of seafloor gas seeps versus in situ aerobic methanogenesis to sea surface CH$_4$ concentrations. To investigate these objectives, we collected water samples for CH$_4$ concentration and stable carbon isotopic analysis along the U.S. Mid-Atlantic Bight (MAB) onboard the R/V Hugh R. Sharp from 25 August to 5 September 2017. The combination of dissolved CH$_4$ concentration and stable isotope measurements has been shown to constrain CH$_4$ production pathways (e.g., Whiticar, 1999) and the extent of CH$_4$ oxidation (e.g., Leonte et al., 2017, 2018), and similar analyses were conducted here. This region was chosen due to the recent discovery of over 300 seafloor methane seeps which originate at water depths from 53 to 1,612 m (Skarke et al., 2014). Some of the seeps lie at or within a few hundred meters shallower than the landward limit of gas hydrate stability (~575 m water depth; Brothers et al., 2013; Skarke et al., 2014). Gas hydrate is a solid, ice-like structure formed by water molecules which can trap CH$_4$ and other gases when sufficiently low temperatures, high pressures, and high CH$_4$ concentrations are found in ocean sediments. Due to their sensitivity to temperature perturbations, upper continental slope gas hydrates are considered susceptible to degradation and CH$_4$ release as ocean waters warm (Berndt et al., 2014; Kvenvolden & Rogers, 2005; Ruppel, 2011; Ruppel & Kessler, 2017). In total, we surveyed 25 sampling stations on seven downslope sampling transects along a 230 km corridor of the upper continental slope between northern Virginia and Cape Hatteras (35.524°–37.539°N) to assess the extent of CH$_4$ oxidation and the distribution of CH$_4$ sources in the water column. Here, we commonly refer to three depth horizons based on the depth where samples were collected: the near surface (0–10 m), the subsurface (10–200 m), and the deeper water column (200 m to seafloor). Since our study area was along the continental slope, the seafloor depth varies among stations.

2. Materials and Methods

2.1. Water Sample Collection and Preparation

Water samples for this study were collected along seven transects on the MAB upper continental slope during a cruise aboard the R/V Hugh R. Sharp (Figure 1). The transect locations were chosen based on the distribution of CH$_4$ seeps delineated by Skarke et al. (2014) and new water column imaging data collected during this research cruise using the U.S. Geological Survey’s Simrad EK80 broadband fisheries echosounder with a 38 kHz transducer (Baldwin et al., 2020). At each station on the sampling transects, water was collected at up to 12 distinct water depths using Niskin bottles arrayed on a conductivity-temperature-depth (CTD) carousel. Temperature, salinity, and density were recorded using a SeaBird Electronics 911 plus CTD for each station, and plots of these data can be found in the supporting information. Three to five stations spanning 136 to 1,025 m water depth comprised each of the transects except for 6 and 7, which consisted of a single station each. In this study, we refer to sample locations as TNSM, where N is the number of the transect and M is the station number. The EK80 imaged the position of the CTD apparatus in the water column during deployments, and CTD casts were designed to intersect or avoid acoustically detectable benthic CH$_4$ plumes, depending on the goal of each measurement.
Using 1/4" Tygon tubing, water samples were transferred from Niskin bottles to 250 ml glass vials (Wheaton) for CH₄ concentration analysis and 500 ml glass vials for δ¹³C-CH₄ isotopic analysis. Vials were filled from the bottom, and the flow rate of water was controlled to ensure that bubbles did not form within the vial during filling. Water was allowed to overflow in order to flush the vials with at least two vial volumes of water before capping with a butyl rubber stopper and aluminum crimp cap (Wheaton). Immediately after capping, a 10 ml ultrahigh purity nitrogen headspace was introduced into each vial via displacement. Two syringes were used simultaneously, one to inject nitrogen and the other to withdraw an equal volume of water. Mercuric chloride (HgCl₂) was then added to each vial to halt further biological activity and preserve the samples until analysis. A gastight syringe (Hamilton) was used to add 250 and 500 μl of a supersaturated HgCl₂ solution to each CH₄ concentration vial and each isotopic ratio vial, respectively. Vials filled for CH₄ concentration analysis were stored in a temperature-controlled incubator set to 6 °C to allow the headspace to equilibrate at near-bottom water temperature for at least 12 hr before analysis in the shipboard laboratory. Vials were placed sideways inside the incubator to prevent the headspace gas from escaping through the rubber stopper. Samples meant for isotopic ratio analysis were stored upside down in custom built sample boxes until analysis on shore.

2.2. Methane Concentration and Stable Carbon Isotopic Ratio Analysis
Methane concentration analyses were performed on board the R/V Hugh R. Sharp using an Agilent 6850 gas chromatograph equipped with a flame ionization detector. From each CH₄ concentration vial, 8 ml of headspace gas was drawn out using a syringe, while 8 ml of CH₄-free water was injected back in to maintain
atmospheric pressure. For each analysis, 4 ml of gas was used to flush and fill a 50 µl sample loop. This allowed the headspace gas of each vial to be analyzed in duplicate. By switching a two-way Valco valve, the nitrogen carrier gas transferred the contents from the sample loop onto a Gas-Pro capillary column (Length = 15 m, i.d. = 0.32 mm, Agilent). The GC was kept isothermal at 40 °C giving CH₄ a retention time of 0.80 min. Peak areas recorded by the FID were converted to CH₄ concentrations using a four-point calibration generated from a nitrogen blank (AirGas) and CH₄ standards with concentrations of 1.01, 10.1, and 100 ppm (AirLiquide). For certain samples where ethane, propane, and butane were analyzed in addition to CH₄, the GC temperature was set to 40 °C for 2.75 min before the temperature was increased at 40 °C min⁻¹ to 70 °C, where it was held for an additional 1.5 min. The retention times for ethane, propane, and butane were 1.14, 2.23, and 4.35 min, respectively. Since our samples showed no trace of ethane, propane, or butane, the dissolved concentrations of these gases were not calculated. For CH₄, dissolved concentrations were calculated by considering the CH₄ concentration measured from the headspace, the volumes of the headspace and water in each sample vial, the temperature and salinity of the water sample, and CH₄ solubility (Wiesenburg & Guinasso, 1979). The average standard deviation of duplicate analyses was 2.5% of the dissolved CH₄ concentrations calculated.

Analyses of stable carbon CH₄ isotopic values were carried out at the Woods Hole Isotope Laboratories following the same procedures described in Leonte et al. (2017). Briefly, headspace gas was injected into a sample loop and preconcentrated before being transferred onto a capillary column using a helium carrier gas. The gas chromatograph, Agilent 6890, was heated from −40 to 180 °C in a stepwise fashion to achieve gas separation. Sample CH₄ was oxidized to CO₂ using a Finnigan GCCIII combustion interface before isotopic ratio analysis on a Thermo Finnigan Delta± XL. On average, the 1σ standard deviation of each measurements was 0.1‰.

2.3. Water Current Velocity Measurements

Water current velocity was measured by the U.S. Geological Survey using an RDI Workhorse 300 kHz Acoustic Doppler Current Profiler (ADCP) in a deepwater housing which was mounted inside the CTD carousel looking downward. The ADCP was lowered into the water using the CTD carousel, and data were recorded during most CTD deployments and downloaded following each cast. The lowered ADCP (LADCP) data were processed with the Lamont Doherty Earth Observatory software package, version IX_13 (Thurnherr, 2011), using bottom tracking, GPS (from CTD time series), and data from the hull mounted RDI Workhorse 300 kHz ADCP to constrain the inversion model. LADCP processing yielded north and east velocity components that were used to calculate the direction and speed of ocean currents (Ruiz-Angulo et al., 2020). Current velocities were determined at 10 m depth intervals between 10 m below the sea surface and the seafloor for all stations and transects except for T4S1, the single station that comprises Transect 7 and the stations on Transect 1. A discussion of the water current velocity data and their relationship to CH₄ transport can be found in the supporting information. For this discussion we relied on the LADCP data which spanned the entire water column, rather than the hull mounted ADCP which only measured current velocities to ~100 m water depth.

2.4. Fraction of Methane Oxidized

In a gas seep environment, after CH₄ dissolves into a parcel of water, its concentration will decrease due to mixing or microbial oxidation (Mau et al., 2012). However, only microbial oxidation will concurrently shift δ¹³C-CH₄ isotopic measurements toward heavier values due to a fractionation effect (Whiticar, 1999). By comparing changes in CH₄ concentration with δ¹³C-CH₄ isotopic values, the amount of CH₄ removed from the water column through oxidation can be calculated. This approach was used by Leonte et al. (2017) using equation (1) to calculate f_c or the fraction of CH₄ emitted to the water column that has been removed through microbial oxidation.

\[
f_c = 1 - \left( \frac{\delta R}{\delta R_0 + 1000} \right) ^{\frac{\alpha_c}{\alpha_c}}
\]

The fraction of CH₄ oxidized, f_c, is calculated based on the difference in δ¹³C-CH₄ isotopic values between the time CH₄ is initially dissolved into the water column, δR₀, and a later time when a water sample is
collected, $\delta R$, along with the fractionation factor, $\alpha_c$, which is a constant characterizing the magnitude of isotopic fractionation as CH$_4$ oxidation progresses. If the source of CH$_4$ to the water column is a gas seep, $\delta R_0$ represents the isotopic value of CH$_4$ when it dissolves from the gas into the liquid phase. If in situ methanogenesis is the CH$_4$ source, $\delta R_0$ is the isotopic value of CH$_4$ when it is initially generated by microbes but before fractionation through microbial oxidation. This model assumes that only oxidation affects $\delta^{13}$C-CH$_4$ isotopic values within a water parcel as it is carried by ocean currents. A fractionation factor of $\alpha_c = 1.0115$ was determined by Leonte et al. (2017) in Hudson Canyon, which also lies along the USAM, and was used for the calculations presented here since that study and this current one consider similar geographical and biogeochemical environments. In the Hudson Canyon, the dominant water column CH$_4$ sink was microbial oxidation as opposed to mixing with neighboring waters, fitting the conditions of a closed system, and allowing for $\alpha_c$ to be calculated based on CH$_4$ concentration and $\delta^{13}$C-CH$_4$ measurements (Leonte et al., 2017).

3. Results

3.1. Gas Bubble Identification

The Simrad EK80 echosounder was used to detect water column gas bubbles and to determine whether the CTD carousel was within a bubble plume during water sampling (Baldwin et al., 2020). This allowed us to determine if the water samples collected were directly influenced by gas bubble emissions and thus constrain the isotopic signature of CH$_4$ emitted from gas seeps. Figure 2 shows images generated by the echosounder as water samples were collected at T1S2 and T4S3, highlighting strong near-seafloor water column backscatter associated with gas bubbles at T1S2 and no such signal at T4S3. The echosounder data also revealed strong water column backscatter for T1S1 through T1S4 and less intense backscatter at T2S3 and T5S2. It should be noted that the echosounder record collapses onto a flat display all detections within a conical ensonification area beneath the EK80 transducer, and the ensonification cone is larger for greater water depths. In some cases, the CTD carousel appeared to be within a backscatter cloud associated with a bubble plume, yet elevated CH$_4$ was not detected in the water samples. For example, the echosounder imaged strong near-seafloor backscatter at T2S2, but the water samples collected within the apparent bubble flare had CH$_4$ concentrations below 4 nM. This apparent contradiction could reflect the highly localized nature of fluctuations in CH$_4$ concentrations near the seafloor. Alternately, the enhanced backscatter imaged by the echosounder could represent bioscatters like krill instead of CH$_4$ bubbles.

3.2. Methane Concentration and Isotopic Distributions

Dissolved CH$_4$ concentrations along the MAB varied based on depth and location and spanned several orders of magnitude from 1.3 nM to 2.1 $\mu$M (Leonte et al., 2020). Nearly all samples were supersaturated with CH$_4$ compared to the atmosphere; average CH$_4$ concentrations measured near the surface were 4.6 $\pm$ 0.2 nM which is slightly greater compared to atmospheric equilibrium (~2.1 nM). Samples considered “near the surface” were those collected at a depth of 10 m or shallower since water density was usually uniform over this depth horizon. At most stations, CH$_4$ concentrations were lower at the surface than at greater depths, but the highest CH$_4$ concentrations were not necessarily measured at the seafloor. For example, at Transect 2 the highest CH$_4$ concentrations were consistently measured at 400 m depth, which was up to several hundred meters shallower than the seafloor based on the station (Figure 3a).

Stable isotopic values of $\delta^{13}$C-CH$_4$ were variable within the water column, ranging from $-70.2\%e$ to $-14.9\%e$ (Leonte et al., 2020). Similar to the CH$_4$ concentration measurements, $\delta^{13}$C-CH$_4$ isotopic values did not have a straightforward relationship with depth, which was likely due to the different CH$_4$ sources and sinks active in the water column. Lighter, or more negative, $\delta^{13}$C-CH$_4$ isotopic values were measured in the subsurface where aerobic in situ CH$_4$ production was likely or at stations located close to seafloor gas seeps which also contributed CH$_4$ to the water column. The lightest isotopic values were measured directly overlying gas seeps. At stations farther than 2.5 km from gas seeps, isotope distributions were highly heterogeneous with large gradients in isotopic values over relatively short depth intervals. Near the surface (0–10 m), $\delta^{13}$C-CH$_4$ isotopic values were close to atmospheric values at $-47\%e$, suggesting near equilibrium between the sea surface and the atmosphere (Schaefer et al., 2016).
4. Discussion
4.1. Methane Sources

Along the MAB, CH$_4$ sources in the water column include seafloor gas seeps, in situ aerobic production, and the atmosphere. At most sampling stations, we observed at least one CH$_4$ concentration maxima occurring roughly between 100 and 200 m depths (Figure 3b). This feature is consistent with previous studies presenting vertical CH$_4$ concentration profiles in open ocean environments (Holmes et al., 2000; Reeburgh, 2007). While gas seeps have been identified along the MAB originating at depths as shallow as 53 m, this subsurface CH$_4$ maximum persisted even where seeps were not present at these depths (e.g., Transects 4, 6, and 7). The

Figure 2. Acoustic echosounder images recorded at two sampling stations. The x axis represents time, while the colored dots represent acoustic backscatter within the water column recorded by the echosounder. This may include suspended particles, gas bubbles, or the Niskin bottle carousel (red line). The top panel shows the image recorded at Transect 1, Station 2, CTD Cast 1 (T1S2) where intense backscatter near the seafloor is indicative of gas bubbles, while the bottom panel was recorded at Transect 4, Station 3, CTD Cast 1 (T4S3) where sparse backscatter suggests that no bubbles were present.
Figure 3. Interpolated CH₄ concentration and δ¹³C-CH₄ measurements for two sampling transects along the MAB. The station numbers within Transect 2 (a) and Transect 4 (b) are indicated, and black dots show where specific water samples were collected. Note the different scale ranges for the different concentration and isotope plots. Measured data were interpolated using Ocean Data View. Although Transects 2 and 4 are only 30 km apart, they show different CH₄ concentration and δ¹³C-CH₄ distributions.
nearest gas seep was identified at a distance of 4.4, 8.3, and 2.5 km away from Transects 4, 6, and 7, respectively, based on data collected by Baldwin et al. (2020) and Skarke et al. (2014). At these transects, CH₄ concentrations measured in the subsurface were not only elevated above background levels but also consistently larger than measurements from the deeper water column. For example, Figure 3b shows an interpolation of CH₄ concentration and δ¹³C-CH₄ isotope measurements from Transect 4. The highest CH₄ concentrations were centered on 100 m for all stations, and although a cluster of seeps was identified to the south of Transect 4, these seeps originate at depths of 211–494 m (Skarke et al., 2014), well below the observed CH₄ maximum.

Figure 4 shows a plot of δ¹³C-CH₄ isotopic values against CH₄ concentrations for samples collected at depths shallower than 200 m. This plot differentiates between samples that were collected near known gas seeps (Transects 1, 2, 3, and 5) and samples that were not (Transects 4, 6, and 7). Transects identified as being near gas seeps were within 2.5 km of a gas seep identified in the data collected here, Baldwin et al. (2020), or reported in Skarke et al. (2014). Where gas seeps were present, CH₄ concentrations were larger, ranging from 17 to 139 nM, while samples without gas seep influence had concentrations ranging from 3 to 55 nM. For transects without nearby gas seeps, CH₄ concentrations decrease below 200 m depth, suggesting that aerobic methanogenesis is not active below this depth or that the sink of CH₄ oxidation is outcompeting this CH₄ source. However, for transects within 2.5 km of identified seafloor gas seeps, elevated CH₄ concentrations were observed at multiple depths throughout the water column. For example, multiple clusters of gas seeps have been identified close to Transect 1 where high CH₄ concentrations were recorded near the seafloor at multiple stations. This suggests that below 200 m depth, gas seeps are the dominant source of CH₄ to the water column.

In order to constrain the isotopic signature of CH₄ produced through aerobic methanogenesis in subsurface waters, we examined δ¹³C-CH₄ measurements of samples collected in the subsurface, at depths shallower than 200 m, that were not influenced by nearby gas seeps (Figure 4). Within this subset of data, the sample with the highest CH₄ concentration also had the lightest δ¹³C-CH₄ value, −65.1‰, which we assume represents the isotopic signature of CH₄ produced aerobically in this region.

In order to determine whether seafloor gas seeps directly influence CH₄ concentrations at the surface, it is important to constrain the vertical distance over which gas bubbles provide a source of CH₄ to the water column. This quantity depends largely on the seafloor depth of bubble release, the chemical composition inside and outside the bubble, and the diameter of gas bubbles when emitted at the seafloor. The Texas A&M Oilspill Calculator was designed to calculate the flux of CH₄ to the water column during bubble ascent (Leonte et al., 2018; McGinnis et al., 2006; Socolofsky et al., 2015; Wang et al., 2016). To date, the only measurements of gas bubble diameter along the MAB were reported by Skarke et al. (2014); using ROV video footage, bubble diameters of 2.3–3.2 and 3.1–4.2 mm were measured at two distinct gas seeps. Based on these measurements, the Texas A&M Oilspill Calculator model shows that bubbles originating at the seafloor with diameters of 2, 3 and 4 mm lose over 90% of their starting CH₄ as they ascend to 55, 105, and 150 m above the seafloor, respectively. This means that gas seeps only have the potential to directly increase water column CH₄ concentrations over these depth horizons and that gas seeps that originate at seafloor depths greater than 150 m are unlikely to increase surface CH₄ concentrations via bubble transport. Once gas bubble CH₄ dissolves into the water column, it is carried by water currents along isopycral surfaces. Given the close relationship between water density and depth among our transects (Figure S3), a dissolved CH₄ plume derived from a gas seep should remain at a similar depth while being transported by ocean currents. As seep derived CH₄ is transported, CH₄ sinks such as microbial oxidation and dilution with background waters actively reduce CH₄ concentrations (Mau et al., 2012). Since deeper waters have a longer transit time to reach the surface, it is more likely that CH₄ emitted from shallower seeps will influence sea surface CH₄ concentrations.

Measurements of δ¹³C-CH₄ isotopes have previously been used to identify the mechanism responsible for generating CH₄ (Whiticar, 1999). Both microbial and thermogenic processes can convert organic matter to CH₄, but these two pathways produce CH₄ with divergent isotopic signatures. Generally speaking, microbial production tends to generate CH₄ with isotopic values between −80‰ and −50‰, while thermogenic production leads to CH₄ isotopic values between −50‰ and −20‰ (Whiticar, 1999). The data measured here
varied through these microbial and thermogenic ranges, but most of these samples were heavily influenced by aerobic CH4 oxidation in the water column, which shifts the $\delta^{13}$C-CH4 toward heavier (apparently thermogenic) values. Thus, in order to determine if CH4 was produced microbially or thermogenically, we had to identify $\delta^{13}$C-CH4 values that were least likely to be influenced by oxidation (labeled here as $\delta R_0$). Estimates of the isotopic signature of gas seep CH4 are more robust if they are based on $\delta^{13}$C-CH4 measurements from samples collected on top of, or close to, gas seeps with a high bubble abundance. In these environments, the high flux of CH4 would overwhelm any potential shift in isotopic values due to mixing with nearby waters or oxidation. Stations at Transect 1 showed the greatest abundance of bubbles identified by the echosounder, and $\delta^{13}$C-CH4 isotopic values measured on samples collected near this seafloor source ranged from $-70.2^{\circ}$ to $-62.0^{\circ}$. Among these, the sample with the lightest $\delta^{13}$C-CH4 value, $-70.2^{\circ}$, and highest CH4 concentration, 2.1 $\mu$M, was collected at Transect 1, Station 3. Due to these conditions, $\delta R_0$ is likely close to $-70.2^{\circ}$ for the gas seeps located near Transect 1.

At Transect 2 the highest CH4 concentrations were all measured at a depth of 400 m for Stations 3, 4, and 5. Although our echosounder readings did not detect a large abundance of water column bubbles at these stations, previous studies have identified a number of gas seeps near Transect 2, which could act as the dominant source of CH4 at 400 m depth (Skarke et al., 2014). A Keeling plot was constructed to estimate the $\delta^{13}$C-CH4 of CH4 emitted from gas seeps near Transect 2 (Figure 5). A Keeling plot can be used to estimate the isotopic signature of a given source term by calculating the linear least squares regression when plotting measurements of $\delta^{13}$C-CH4 versus the inverse of CH4 concentration (Keeling, 1958). The y intercept represents the $\delta^{13}$C-CH4 signature when the concentration of CH4 from this source is infinite compared to the background. For our calculation, we used a geometric mean regression, rather than a least squares regression, to take into account errors in both the x and y variables used in the Keeling plot (Pataki et al., 2003). This approach suggests that the $\delta^{13}$C-CH4 of CH4 emitted from gas seeps near Transect 2 was $-53.4^{\circ}$. Although certain measurements from Transect 2 outside of the concentration plume at 400 m had lighter $\delta^{13}$C-CH4 isotopic values than the samples used to construct the Keeling plot, they also had lower CH4 concentrations, suggesting that CH4 was derived from a different source compared to the CH4 plume identified at 400 m. The $\delta R_0$ value of $-53.4^{\circ}$ implies that seeps in this region emit CH4 with a heavier $\delta^{13}$C-CH4 value than the seeps identified at Transect 1. However, based on the Geometric Mean regression calculation, this $\delta R_0$ value had an uncertainty of 7.2$^{\circ}$ due to the low sample size ($n = 3$) and variability of the data (Figure 5).

Given the available data presented here, the value of $\delta^{13}$C-CH4 emitted from seafloor seeps along the MAB, $\delta R_0$, was assumed to be within the range of $-70.2^{\circ}$ and $-53.4^{\circ}$.
Based on the isotopic values of CH4 emitted from gas seeps determined here (−70.2‰ to −53.4‰), this CH4 is likely produced through microbial processes. This is consistent with a previous study investigating the δ13C signature of autogenic carbonates formed near gas seeps along the MAB (Prouty et al., 2016). In the subsurface, where water depth was shallower than 200 m, measurements of δ13C-CH4 ranged between −65.4‰ and −42.1‰, suggesting that the CH4 source in the subsurface is also of microbial origin. Although some samples collected shallower than 200 m depth have δ13C-CH4 signatures heavier than the typical microbial production cutoff of −50‰, it is likely that when this CH4 was produced, the δ13C-CH4 values were within the microbial range and were later altered through microbial oxidation to a heavier value, as is evidenced by the concurrent shifts to lower dissolved concentrations (Figure 4). Thermogenic production, unlike microbial production, would also lead to the formation of longer chained hydrocarbons such as ethane, propane, and butane. Several water samples collected at Transect 5 were analyzed for these compounds, and all were below detection limits.

4.2. Methane Oxidation

Microbial oxidation of CH4 readily occurs in oxygenated waters, making this process an important sink of oceanic CH4 (Chan et al., 2019; Reeburgh, 2007). Previous work in the Hudson Canyon showed that CH4 emitted from seafloor seeps is quickly and quantitatively oxidized (Leonte et al., 2017). However, the unique topography and water circulation patterns in Hudson Canyon likely helped enhance the rate of CH4 oxidation by trapping CH4 and methanotroph-laden waters within the canyon walls. Similarly, samples collected here in other regions of the MAB have a wide range of δ13C-CH4 values, driven partially by varying isotopic signatures of CH4 sources (see previous section) and mostly by the microbial oxidation of CH4. At nearly all transects, δ13C-CH4 isotopic values heavier than −30‰ have been measured, which is significantly heavier than the δ13C-CH4 of CH4 entering the water column from gas seeps (−70.2‰ to −53.4‰) or produced by aerobic methanogenesis (−65.1‰). Samples with δ13C-CH4 isotopic values heavier than −53.4‰ have likely been influenced by microbial oxidation since this process progressively enriches the CH4 pool within a parcel of water with the heavier isotopologue, 13CH4. For deep water column samples, collected below 200 m, where gas seeps are the dominant CH4 source, the fraction of CH4 oxidized, f, was calculated using equation (1) based on δ13C-CH4 measurements of CH4 entering the water column, δR0 (−70.2‰ to −53.4‰) and δ13C-CH4 measurements of water samples influenced by oxidation, δR. Since we determined a range of values for δR0, the resulting calculations of f, using equation (1) also produced a range of values. We calculated the maximum value of f when δR0 was set to −70.2‰ and the minimum value of f when δR0 was set to −53.4‰ (Leonte et al., 2020). For δR measurements lighter than
-53.4‰, the minimum \( f_c \) value calculated was negative and thus set to 0. This situation implies that at a minimum, no oxidation had occurred at the time of sampling after seep-derived CH\(_4\) dissolved into the water column.

Calculated \( f_c \) values show that CH\(_4\) oxidation is an active process which has affected many of these samples to a large degree. Of the 75 samples collected from the deeper water column, below 200 m, and measured for CH\(_4\) isotopic ratios, 7 had a minimum \( f_c \) value greater than 0.9 and 28, 47, and 54 samples had minimum \( f_c \) values greater than 0.7, 0.5 and 0.3, respectively (Leonte et al., 2020). On the other hand, maximum \( f_c \) values suggest that 47 samples have over 90% of CH\(_4\) removed through oxidation. The distribution of \( \delta R \) and \( f_c \) values was also correlated to the presence or absence of nearby gas seeps. Figure 6 shows a plot of the minimum CH\(_4\) fraction oxidized, \( f_c \), against \( \delta^{13}C-\text{CH}_4 \) isotopic values of samples collected from the deeper water column. Transects 1, 2, and 3, located close to gas seep clusters, showed an abundance of samples with \( f_c \) values as low as 0 due to significant CH\(_4\) inputs from nearby seeps. However, samples from these transects also had a number of samples with \( f_c \) values close to 1 indicating a strong microbial oxidation signal despite the influence of gas seeps. For Transects 4, 5, 6, and 7, \( f_c \) values had a narrower range that was also shifted toward larger values of \( f_c \) due to a lack of active CH\(_4\) inputs.

Water column data collected within the subsurface also indicate a strong microbial oxidation sink. Figure 4 shows a plot of CH\(_4\) concentration and \( \delta^{13}C-\text{CH}_4 \) measurements consistent with the expected trend produced through microbial oxidation: Lower CH\(_4\) concentrations correlate with heavier \( \delta^{13}C-\text{CH}_4 \) values as oxidation progresses. As mentioned above, dividing the shallow water column samples based on the influence of nearby gas seeps results in different patterns regarding CH\(_4\) concentration and \( \delta^{13}C-\text{CH}_4 \) measurements. Since \( \delta^{13}C-\text{CH}_4 \) values for samples affected by gas seeps within the subsurface (-65.4‰ to -54.8‰) are within the same range as CH\(_4\) emitted from gas seeps within the deeper water column (-70.2‰ to -53.4‰), it is possible that this isotopic variation is entirely driven by the differences in CH\(_4\) isotopes emitted from gas seeps. When calculating the fraction oxidized using a \( \delta R_0 \) value range of -70.2‰ to -53.4‰, the minimum \( f_c \) value for these samples was 0 while the maximum \( f_c \) value ranged from 0.37 to 0.91 (Leonte et al., 2020). For samples in the subsurface that were collected farther than 2.5 km from the nearest gas seep, the sample with the highest CH\(_4\) concentration also had the lightest isotopic signature (-65.1‰), which we assume represents the isotopic signature for in situ methanogenesis. Assuming \( \delta R_0 = -65.1\% \), \( f_c \) was calculated based on equation (1). Values of \( f_c \) ranged from 0 to 0.88, implying a strong microbial oxidation sink (Leonte et al., 2020). However, equation (1) assumes that no additional CH\(_4\) is added to a water parcel after the initial start of the oxidation reaction. Since in situ methanogenesis likely provides a semicontinuous flux of CH\(_4\) to the water column, the quantity of CH\(_4\) oxidized is likely underestimated using equation (1). Despite the CH\(_4\) maximum observed between 100 and 200 m water depths, dissolved CH\(_4\) concentrations quickly decreased to values near atmospheric equilibrium closer to the sea surface. This suggests that CH\(_4\) oxidation is a thorough and near-complete removal mechanism of dissolved CH\(_4\) despite widespread gas seepage and aerobic methanogenesis along the MAB.

5. Conclusions

When assessing the contribution of CH\(_4\) to surface waters from various sources along the MAB, depth was found to be a key variable. Within the subsurface between 100 and 200 m depths, in situ production was responsible for elevating CH\(_4\) concentrations above background at nearly all stations, with shallow gas seeps providing an additional CH\(_4\) source at these depths. In the deeper water column, depths greater than 200 m, gas seeps appeared to be the dominant source of CH\(_4\). Measurements of \( \delta^{13}C-\text{CH}_4 \) suggest that the CH\(_4\)
emitted from the sampled gas seeps is produced through microbial rather than thermogenic processes; however, more seep gas sampling on the MAB continental slope will be needed to confirm the variation and spatial distribution of this CH₄ source. In this study, dissolved δ¹³C-CH₄ measurements were used to constrain the fraction of CH₄ oxidized in the water column, f_c. Despite variations in the CH₄ isotopes at the point of release into the water column, estimates of f_c still display substantial CH₄ oxidation.

As water currents carry waters laden with CH₄, dilution and microbial oxidation act to reduce CH₄ concentrations, sometimes down to background levels. The determination of f_c using equation (1) is based on a closed system Rayleigh model. This model, which takes a closed system Lagrangian approach, assumes that only microbial oxidation is responsible for reducing CH₄ concentration and altering δ¹³C-CH₄ measurements. As the water parcel travels away from the seep, oxidation progressively reduces the concentration of CH₄ and shifts δ¹³C-CH₄ toward heavier values. This approach was found to be well suited to Hudson Canyon (Leonte et al., 2017), which could be treated roughly as a closed system. Along the MAB, seeps are found in clusters and there is more interaction with water on the shelf, the lower continental slope, and other parts of the upper continental slope. Thus, CH₄ can be introduced into or diluted as water parcels move through the area. When investigated in Hudson Canyon, a closed system Rayleigh model was deemed most appropriate even though CH₄ concentrations and δ¹³C-CH₄ were periodically reset to starting values. Adopting the closed system Rayleigh model along the part of the MAB studied here means that we cannot fully resolve all the open system processes affecting CH₄ in the water column, but the main drawback with the closed system model is underestimating (rather than overestimating) CH₄ oxidation. The magnitude of f_c is proportional to the difference between δR₀ and δR, and as oxidation progresses, δR becomes heavier, or more positive, thus increasing the difference between δR₀ and δR. Additional seep input would introduce lighter, or more negative, CH₄, thus decreasing the difference between δR₀ and δR and causing the value of f_c to underestimate the true amount of oxidation. Thus, the closed system approach is likely a conservative approach to assessing CH₄ oxidation.

At water depths shallower than 200 m, both gas seeps and in situ production act as sources to the water column. The δ¹³C-CH₄ measurements suggest that in situ production generates CH₄ with an isotopic value of −65.1‰, and the relationship between δ¹³C-CH₄ and CH₄ concentrations (Figure 4) shows that oxidation is readily occurring in the subsurface. The combination of dilution and microbial oxidation readily decreased CH₄ concentrations close to background levels at water depths shallower than 100 m, although surface CH₄ concentrations were slightly higher than equilibrium with the atmosphere. This suggests a small flux of CH₄ from the ocean to the atmosphere, despite much higher CH₄ concentrations at depth. Overall, CH₄ sinks readily reduce CH₄ concentrations within the water column and prevent CH₄ from reaching the atmosphere despite significant CH₄ sources at depth.

References


