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4 **Clumped isotope effects during OH and Cl oxidation of methane**

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ABSTRACT

A series of experiments were carried out to determine the clumped ($^{13}\text{CH}_3\text{D}$) methane kinetic isotope effects during oxidation of methane by OH and Cl radicals, the major sink reactions for atmospheric methane. Experiments were performed in a 100 L quartz photochemical reactor, in which OH was produced from the reaction of $\text{O}(^1\text{D})$ (from O_3 photolysis) with H_2O , and Cl was from photolysis of Cl_2 . Samples were taken from the reaction cell and analyzed for methane ($^{12}\text{CH}_4$, $^{12}\text{CH}_3\text{D}$, $^{13}\text{CH}_4$, $^{13}\text{CH}_3\text{D}$) isotopologue ratios using tunable infrared differential laser absorption spectroscopy. Measured kinetic isotope effects for singly substituted species were consistent with previous experimental studies. For doubly substituted methane, $^{13}\text{CH}_3\text{D}$, the observed kinetic isotope effects closely follow the product of the kinetic isotope effects for the ^{13}C and deuterium substituted species (i.e., $^{13,2}\text{KIE} = ^{13}\text{KIE} \times ^2\text{KIE}$). The deviation from this relationship is $0.3\text{‰} \pm 1.2\text{‰}$ and $3.5\text{‰} \pm 0.7\text{‰}$ for OH and Cl oxidation, respectively. This is consistent with model calculations performed using quantum chemistry and transition state theory. The OH and Cl reactions enrich the residual methane in the clumped isotopologue in open system reactions. In a closed system, however, this effect is overtaken by the large D/H isotope effect, which causes the residual methane to become anti-clumped relative to the initial methane. Based on these results, we demonstrate that oxidation of methane by OH, the predominant oxidant for tropospheric methane, will only have a minor ($\sim 0.3\text{‰}$) impact on the clumped isotope signature ($\Delta^{13}\text{CH}_3\text{D}$, measured as a deviation from a stochastic distribution of isotopes) of tropospheric methane. This paper shows that $\Delta^{13}\text{CH}_3\text{D}$ will provide constraints on

- 45 methane source strengths, and predicts that $\Delta^{12}\text{CH}_2\text{D}_2$ can provide information on
- 46 methane sink strengths.

1. INTRODUCTION

Atmospheric methane is the second most important long-lived greenhouse gas behind carbon dioxide, and is a much stronger greenhouse gas than CO₂ on a molecule-per-molecule basis (e.g. Lelieveld et al., 1998). Atmospheric methane has more than doubled from preindustrial levels below 800 parts per billion by volume (ppbv) (Craig and Chou, 1982; Khalil and Rasmussen, 1982; Rasmussen and Khalil, 1984) to current atmospheric concentrations of over 1800 ppbv. Methane also plays a central role in the chemistry of the troposphere and stratosphere (Harnung and Johnson, 2012). In the troposphere, methane oxidation moderates hydroxyl radical concentrations (OH) and is the main photochemical precursor for carbon monoxide (CO), in addition to forming formaldehyde (CH₂O) and ozone (O₃). In the stratosphere, oxidation of methane results in the formation of stratospheric water vapor (e.g. Rosenlof et al., 2001) and has an impact on stratospheric HO_x (OH and HO₂), ClO_x, and ozone concentrations.

The dominant sink of tropospheric methane is oxidation by OH, which is responsible for about 90% of methane oxidation. Due to the strong temperature dependence of the CH₄+OH reaction, as well as the global distribution of OH radicals, methane oxidation by OH occurs predominantly in the lower tropical troposphere (e.g. Fung et al., 1991; Dlugokencky et al., 2011) during high sunlight conditions. As a result, the majority of methane is oxidized by OH at temperatures around 298 K. Minor oxidation pathways include oxidation by Cl in the marine boundary layer (Allen et al., 2007) and uptake by soil microbes. In the stratosphere, oxidation by OH, Cl, and O(¹D) are all sinks of methane. Natural sources of methane include wetlands, termites, and oceans.

Anthropogenic methane sources include fossil fuels, ruminant agriculture, rice agriculture, and biomass burning. The budgets of different sources and the balance of sources and sinks, both on a regional and a global level, still have high uncertainties (Dlugokencky et al., 2011; Kirschke et al., 2013).

One contentious issue in methane biogeochemistry is the cause of decadal variability in methane mixing ratios. Methane concentrations were rising steadily from the industrial revolution until the 1980's (Stauffer et al., 1985; Etheridge et al., 1992; Dlugokencky et al., 1994), when methane growth rates began to decline (Steele et al., 1992; Dlugokencky et al., 1994) and eventually methane concentrations stabilized (Dlugokencky et al., 2003). However, starting around 2007, methane growth rates have increased again (Rigby et al., 2008; Bousquet et al., 2011). A number of theories have been proposed to explain this variability in methane growth rates, including changes in microbial sources (Mikaloff Fletcher et al., 2004; Bousquet et al., 2006; Kai et al., 2011), changes in fossil fuel emissions (Aydin et al., 2011; Simpson et al., 2012), or changes in the OH sink (Bekki et al., 1994). Reducing the uncertainties in methane biogeochemistry is key to understanding and constraining the global methane cycle and predicting future changes in the methane cycle in a changing world.

High-precision measurements of global, hemispheric, and regional methane mixing ratios and bulk isotope ratios ($\delta^{13}\text{C}$ and δD) have provided significant insight into the seasonal, decadal, and interhemispheric variability in methane concentrations (e.g. Quay et al., 1999; Lassey et al., 2000; Miller et al., 2002). In particular, measurements and modeling of $\delta^{13}\text{C}$ values of atmospheric methane, as well as the interhemispheric variability in $\delta^{13}\text{C}$ and its changes over time, have provided additional constraints on the

global methane cycle (Allan et al., 2007). Seasonal covariation between methane concentration and isotope ratios ($\delta^{13}\text{C}$ and δD) suggest that seasonal changes in methane are due to changes in both source and sink concentrations over the course of the year (Bergamaschi et al., 2000; Allan et al., 2001). Despite these efforts, the methane budget has significant uncertainties (Fung et al., 1991; Dlugokencky et al., 2011).

Most stable isotope work on methane to date has focused on the measurement of individual isotope ratios ($\delta^{13}\text{C}$, δD) rather than isotopologues ($\delta^{13}\text{CH}_4$, $\delta^{12}\text{CH}_3\text{D}$), generally by conversion of methane to CO_2 and H_2 and subsequent analysis by isotope ratio mass spectrometry (e.g. Miller et al., 2002). Recent developments in near-infrared and mid-infrared laser spectroscopy instruments have allowed direct measurement of intact methane isotopologues (i.e. $\delta^{13}\text{CH}_4$, $\delta^{12}\text{CH}_3\text{D}$), both with preconcentration (e.g. Eyer et al., 2016) and at atmospheric abundance (e.g. Santoni et al., 2012). High-precision and high temporal resolution measurements of atmospheric methane (e.g. Röckmann et al., 2016), once widely deployed, promise modest reductions in uncertainties for different methane sources and sinks (Rigby et al., 2012).

Alongside technological improvements in the measurement of traditional (singly-substituted) isotopologues of methane, recent advances in isotope ratio mass spectrometry (Eiler et al., 2013; Stolper et al., 2014) and quantitative infrared laser spectroscopy (Ono et al., 2014; Wang et al., 2015) are facilitating the ability to measure the rare doubly-substituted methane isotopologues (i.e. $^{13}\text{CH}_3\text{D}$, and $^{12}\text{CH}_2\text{D}_2$) at natural abundance. To first order, the abundance of these “clumped” (doubly-substituted) isotopologues follow a stochastic distribution of isotopologues (i.e. $[^{12}\text{CH}_4][^{13}\text{CH}_3\text{D}] =$

[¹³CH₄][¹²CH₃D]]. However, small (per mil level) deviations from this stochastic distribution are observed due to thermodynamic (Ma et al., 2008; Stolper et al., 2014; Liu and Liu, 2016), kinetic (Wang et al., 2015), photolytic (Schmidt et al., 2013; Schmidt and Johnson, 2015), and combinatorial (Yeung, 2016) effects. Stolper et al. (2014) hypothesize that there could be significant clumped kinetic isotope effects during the photochemical methane sink reactions. If this were the case, doubly-substituted isotopologues of methane could be used as tracers for atmospheric sink reactions.

Kinetic isotope effects have been measured experimentally for both OH and Cl oxidation of methane for ¹³CH₃D (Joelsson et al., 2014, Joelsson et al., 2015). The uncertainties in the measurements, however, are too large to resolve per mil level deviations from a stochastic distribution during these sink reactions. One recent study (Joelsson et al. 2015) suggests that the impact of OH oxidation of atmospheric methane was $\Delta^{13}\text{CH}_3\text{D} = 20\text{‰} \pm 20\text{‰}$ (measured as a deviation from a stochastic distribution of isotopes). The uncertainty is larger than the range of measured $\Delta^{13}\text{CH}_3\text{D}$ values of methane sources (-4‰ to 6‰, Wang et al., 2015).

In this study, a series of methane oxidation experiments were carried out using both OH and Cl. At several points during the experiment, residual methane was sampled and measured for abundance of isotopologues of methane (¹²CH₄, ¹³CH₄, ¹²CH₃D, ¹³CH₃D) by using tunable mid-infrared laser differential absorption spectroscopy (TILDAS, Ono et al., 2014). Experimental results are compared with a model constructed using quantum chemistry and transition state theory. We will discuss our results using a simple box model (Tans, 1997) to provide a first estimate the $\Delta^{13}\text{CH}_3\text{D}$ value of tropospheric methane, as well as its seasonal variability.

2. METHODS

2.1. Terminology and notation

Isotopologue ratios (R values) express the ratio of a particular minor isotopologue ($^{13}\text{CH}_4$, $^{12}\text{CH}_3\text{D}$, $^{13}\text{CH}_3\text{D}$) to the major isotopologue ($^{12}\text{CH}_4$). For example,

$$^{13}R = \frac{[^{13}\text{CH}_4]}{[^{12}\text{CH}_4]} \quad \text{Eq. 1}$$

Prefix superscripts are used to refer to specific isotopologues, with ^{13}R referring to the $^{13}\text{CH}_4 / ^{12}\text{CH}_4$ ratio, 2R referring to the $^{12}\text{CH}_3\text{D} / ^{12}\text{CH}_4$ ratio, and $^{13,2}R$ referring to the $^{13}\text{CH}_3\text{D} / ^{12}\text{CH}_4$ ratio. Isotopologue ratios are measured relative to a reference gas (AL-1, Ono et al., 2014), and standardized to the starting isotopologue composition of the methane used in the experiments. Results are presented in isotopologue delta notation relative to initial (starting) methane isotopologue composition:

$$\delta^{13}\text{CH}_4 = \frac{^{13}R_{\text{sample}}}{^{13}R_{\text{initial}}} - 1 \quad \text{Eq. 2}$$

Note that for deuterated species, there are symmetry factors (e.g., $\text{D}/\text{H} = 1/4$ $\text{CH}_3\text{D}/\text{CH}_4$), which cancel in delta notation. At natural isotopic abundances, isotopologue ratios are approximately equal to isotope ratios, i.e. $\delta^{13}\text{CH}_4 \approx \delta^{13}\text{C}$ and $\delta^{12}\text{CH}_3\text{D} \approx \delta\text{D}$.

When analyzing the doubly-substituted isotopologue $^{13}\text{CH}_3\text{D}$, we are also concerned with the deviation from a stochastic distribution of isotopes (Stolper et al. 2014). We define this quantity as:

$$\Delta^{13}\text{CH}_3\text{D} = \frac{[^{13}\text{CH}_3\text{D}][^{12}\text{CH}_4]}{[^{12}\text{CH}_3\text{D}][^{13}\text{CH}_4]} - 1 \quad \text{Eq. 3}$$

or (Ono et al., 2014)

$$\Delta^{13}\text{CH}_3\text{D} = \ln \frac{[^{13}\text{CH}_3\text{D}][^{12}\text{CH}_4]}{[^{12}\text{CH}_3\text{D}][^{13}\text{CH}_4]} \quad \text{Eq. 4}$$

Because $\Delta^{13}\text{CH}_3\text{D}$ values are close to 1, the differences between the two definitions is small. A first-order Taylor expansion shows that the differences in calculated $\Delta^{13}\text{CH}_3\text{D}$ values from the two definitions are on the order of $\Delta^{13}\text{CH}_3\text{D}^2/2$ and can be generally ignored (Liu and Liu, 2016). The definition from Eq. 4 is used for the remainder of this paper.

In regard to our experimental results, we are concerned with the $\Delta^{13}\text{CH}_3\text{D}$ value relative to the initial (starting) methane, whose $\Delta^{13}\text{CH}_3\text{D}$ value is not zero. Therefore, with regard to our experimental results, we define $\Delta^{13}\text{CH}_3\text{D}$ as:

$$\Delta^{13}\text{CH}_3\text{D} = \ln \left(\frac{(\delta^{13}\text{CH}_3\text{D}+1)}{(\delta^{12}\text{CH}_3\text{D}+1) \cdot (\delta^{13}\text{CH}_4+1)} \right) \quad \text{Eq. 5}$$

Where the δ values are defined relative to the initial (starting) methane.

2.2. CH₄ + OH Experiments

A total of five CH₄ + OH experiments were carried out (OH-1 to OH-5), with two to three samples (plus initial methane) collected for each experiment at different degrees of oxidation. Oxidation experiments were carried out in a 100 L quartz photochemical reaction cell at the Copenhagen Center for Atmospheric Research at the University of Copenhagen (Nilsson et al., 2009). The reaction cell is coupled to a Bruker IFS 66v/S Fourier transform infrared spectrometer with multipass optics and a liquid nitrogen cooled InSb detector. The cell is located in a temperature-controlled housing to keep temperature stable at 298 K (± 1 K) during the experiments, and is surrounded by a series of 16 UV-C Hg lamps (used to photolyze the O₃).

The reaction cell was evacuated by a turbo pump prior to experiments. Degassed (using several freeze-pump-thaw cycles) water vapor was expanded into the cell up to a pressure of approximately 20 mbar. Following this, approximately 100 mL STP (standard temperature and pressure) of methane (at natural isotopic abundances) was expanded and flushed into the 100 L reaction cell in helium carrier gas. Nitrous oxide (N_2O) (0.5 ml STP to 1 ml STP) was also added as a tracer of oxidation by $\text{O}(^1\text{D})$, as N_2O can be oxidized by $\text{O}(^1\text{D})$ but not by OH. After adding the desired amount of methane and nitrous oxide, the cell was pressurized to 930 mbar with helium. A split of the gas mixture was sampled (as described below) to provide an initial isotopic composition. Following sampling, several FTIR spectra were recorded, with measurements continuing until multiple repeated measurements gave consistent values. Once repeated measurements were consistent with each other, we assumed that the gas in the reaction cell was well mixed and that the measured concentrations were representative of the average concentration within the cell. These spectra provided the starting concentration of methane within the reaction cell.

In our experiments, methane oxidation by $\text{O}(^1\text{D})$ can compete with the $\text{CH}_4 + \text{OH}$ reaction. Following Cantrell et al. (1990), we kept the H_2O to CH_4 ratio as high as possible (> 100) and added O_3 in small increments to maximize oxidation by OH versus $\text{O}(^1\text{D})$. Ozone was added to the cell by expanding it into a 1 L expansion volume and then flushing it into the cell with helium. Approximately 6 ml to 12 ml STP of O_3 was added at a time. Following each addition of O_3 , the cell was irradiated with the UV-C Hg lamps for around 180 seconds to photolyze the ozone and form $\text{O}(^1\text{D})$. $\text{O}(^1\text{D})$ reacts with the H_2O in the cell to produce two OH radicals, which subsequently oxidize the

methane. Several (around 5 to 15) additions of ozone were made, irradiating the cell between additions. Ozone had to be added and photolyzed in small amounts to maintain the low O_3/H_2O ratio.

During sampling, approximately 13 ml STP (550 μ mol) of methane was sampled from the cell. The gas within the cell was drawn through a needle valve and a liquid nitrogen trap (to remove ozone and carbon dioxide), and then methane was trapped on silica gel at -196°C . During the sampling of methane, oxygen (O_2) was trapped along with methane. This was removed during the sample purification step (Section 2.4).

Multiple FTIR spectra were taken before and after each sampling and oxidation step to quantify the amount of methane oxidized or sampled. The quantification of methane concentrations by FTIR was used to calculate the fraction of methane oxidized (i.e. f values), and used to determine when to collect samples.

2.3. CH_4 + Cl Experiments

Chlorine oxidation experiments were carried out in a similar way to the CH_4 + OH experiments described above. The same 100 L quartz photochemical reaction cell was used (Nilsson et al. 2009). Methane (200 mL STP) and chlorine gas (variable amounts) were separately expanded into a 1 L expansion volume, where the pressure was measured with a manometer. The gas was then flushed into the 100 L reaction cell with the helium carrier gas. After adding the desired amount of methane and chlorine, the cell was pressurized to 400 mbar with helium. Initial methane concentrations were measured by FTIR.

The gas mixture was irradiated with the UV-A lamps positioned around the reaction cell, causing Cl_2 photolysis and subsequent oxidation of methane by chlorine atoms. Following irradiation, the residual concentration of methane in the cell was measured by FTIR. If insufficient methane was oxidized, additional chlorine was added to the cell and irradiation was repeated. Our goal was 15% to 20% oxidation at each step. Once a sufficient amount of oxidation had occurred, methane was sampled from the cell. For the CI experiments, repeated FTIR measurements did not give consistent results or converge to a particular value within the timescale of our experiments. Because of this, , we were unable to obtain precise f value measurements (i.e. fraction of methane oxidized) for calculating precise kinetic isotope effects (α values). However, we were able to determine the relative relationships among isotope effects from TILDAS measurements.

2.4. Sample purification and TILDAS analysis

Following collection, samples were sealed in gas tight storage flasks or flame sealed in glass tubes and transported to the Stable Isotope Geobiology Laboratory at the Massachusetts Institute of Technology (MIT). At MIT, the samples were purified using the preparatory gas chromatography method described by Wang et al. (2015). Sampled gases were concentrated on a trap filled with silica gel, and then another trap filled with activated charcoal, at $-196\text{ }^\circ\text{C}$. The activated charcoal trap was heated to $100\text{ }^\circ\text{C}$ in a flow of helium to introduce the methane sample into a 1.5 m column packed with Carboxen and held at $30\text{ }^\circ\text{C}$. Following the elution of the N_2 and O_2 peaks, the CH_4 peak was trapped on a silica gel trap at $-196\text{ }^\circ\text{C}$. A thermal conductivity detector (TCD)

was used to time the elution of the gases. Following the trapping of the methane, residual helium was pumped away and the purified methane was transferred to a separate inlet line for the tunable infrared laser differential absorption spectroscopy (TILDAS) instrument. Purified methane was analyzed for relative concentrations of $^{12}\text{CH}_4$, $^{12}\text{CH}_3\text{D}$, $^{13}\text{CH}_4$, and $^{13}\text{CH}_3\text{D}$ using TILDAS, as described previously (Ono et al., 2014).

Sample purity was assessed by comparing spectroscopically-measured methane mixing ratios to those calculated based on sample pressure and volume within the cell. Based on this comparison, methane had a consistent purity of 99% or higher, with the major contaminant expected to be trace amounts of nitrogen or helium that leak through during the GC purification step. Due to the high specificity of the rovibrational methane absorption lines measured by TILDAS, trace impurities of nitrogen and helium will not significantly affect the measured isotope ratios. The major effect will come from uncertainties in the spectral line broadening parameters in different gas matrices (i.e. line broadening from pure methane versus nitrogen versus helium). Given the higher than 99% purity in the methane, the spectral line broadening will be dominated by self-broadening due to methane, and the less than 1% impurities will have an insignificant effect.

2.5. Error propagation

The amount of methane within the reaction cell at various steps during the oxidation reaction was quantified using FTIR. The mean and standard deviation for multiple FTIR measurements were used to calculate the mean and standard deviation

for the f values (fraction of initial methane remaining) at different sampling steps. The mean and standard deviation of the isotope ratio measurements is calculated from multiple ($N = 8$ to $N = 10$) sample versus reference cycles for analysis of each methane sample. Uncertainty in the initial isotopic composition of the methane within the cell, as well as possible fractionation during sampling, is propagated in the reported uncertainty in the measurements. The ordinary least squares regression method was used to calculate the best estimate for the kinetic isotope effects as well as the standard errors and 95% confidence intervals. Errors in the isotopologue ratios (δ values) for each sample are correlated, due to common terms in the calculation of the reported isotopic signatures. Errors in the f values and δ values are not correlated because they are measured independently by different techniques.

3. RESULTS

3.1 Results from control experiment

A control experiment was performed to test for isotope fractionation during sample collection. Approximately 100 ml (STP) of methane was put into the cell and it was filled to 930 mbar total pressure with helium. Three samples were then sequentially collected using the same method used to sample the experiments. The isotope ratios from these three control samples are presented in Table 1. Measured $\delta^{13}\text{CH}_4$ and $\delta^{13}\text{CH}_3\text{D}$ ratios both differed by up to 1.1 ‰, which is larger than the analytical uncertainty in the $\delta^{13}\text{CH}_4$ and $\delta^{13}\text{CH}_3\text{D}$ measurements. This suggests that there was some degree of fractionation in $^{13}\text{CH}_4$ and $^{13}\text{CH}_3\text{D}$ during sample collection. $\delta^{12}\text{CH}_3\text{D}$ ratios agree to within 0.2 ‰, which is within the precision of the measurement. The uncertainty in the initial isotopic composition and the inconsistent ^{13}C fractionation during sample collection is the largest uncertainty in measured $\delta^{13}\text{CH}_4$ and $\delta^{13}\text{CH}_3\text{D}$ ratios and the error is propagated in all the following calculations. Only 2 of the 7 initial methane values measured (Control-1-b and Control-1-c, Table 1) had unusual $\delta^{13}\text{CH}_4$ values compared to the other five. However, because we had no justification for excluding these outliers, we included them, and the corresponding uncertainty they caused, in all the analyses.

3.2. Results from OH oxidation experiments

Results from OH oxidation experiments are reported in Table 1. Kinetic isotope effects (KIE) can be calculated from the slope of a linear regression of $\ln(R)$ versus $\ln(f)$, where $\ln()$ is the natural log function. These were plotted for the three isotopologue ratios measured ($^{12}\text{CH}_3\text{D}/^{12}\text{CH}_4$, $^{13}\text{CH}_4/^{12}\text{CH}_4$, $^{13}\text{CH}_3\text{D}/^{12}\text{CH}_4$). The slope of this

regression represents our best estimate for the value of α^{-1} , where α is the inverse of
 the kinetic isotope effect ($k_{\text{light}}/k_{\text{heavy}}$). The sum of the square of the residuals for the fit
 was computed and used to calculate the uncertainty in the slope. A two-tailed t test was
 used with the uncertainty in the slope to calculate a 95% confidence interval for the
 calculated isotope effects. The calculation gives the kinetic isotope effects $k_{\text{OH}}/{}^2k_{\text{OH}} =$
 ${}^2\text{KIE}_{\text{OH}} = 1.3344 \pm 0.0182$, $k_{\text{OH}}/{}^{13}k_{\text{OH}} = {}^{13}\text{KIE}_{\text{OH}} = 1.0061 \pm 0.0012$, and $k_{\text{OH}}/{}^{13,2}k_{\text{OH}} =$
 ${}^{13,2}\text{KIE}_{\text{OH}} = 1.3429 \pm 0.0192$, where k_{OH} is the rate constant for the ${}^{12}\text{CH}_4 + \text{OH}$ reaction
 and ${}^2k_{\text{OH}}$, ${}^{13}k_{\text{OH}}$, and ${}^{13,2}k_{\text{OH}}$ are the rate constants for the reactions of ${}^{12}\text{CH}_3\text{D}$, ${}^{13}\text{CH}_4$,
 and ${}^{13}\text{CH}_3\text{D}$, respectively, with OH. We can also look at the inverse of the kinetic
 isotope effects, which are the isotopic fractionation factors ($\alpha = 1/\text{KIE}$). This gives
 values of ${}^2\alpha_{\text{OH}} = {}^2k_{\text{OH}}/k_{\text{OH}} = 0.7494 \pm 0.0103$, ${}^{13}\alpha_{\text{OH}} = {}^{13}k_{\text{OH}}/k_{\text{OH}} = 0.9939 \pm 0.0012$, and
 ${}^{13,2}\alpha_{\text{OH}} = {}^{13,2}k_{\text{OH}}/k_{\text{OH}} = 0.7447 \pm 0.0108$.

We are interested in calculating the relationship between the kinetic isotope
 effects (e.g. Wang et al., 2016), which we define here as ${}^{13,2}\gamma_{\text{OH}} = (k \cdot {}^{13,2}k) / ({}^2k \cdot {}^{13}k) =$
 ${}^{13,2}\alpha_{\text{OH}} / ({}^2\alpha_{\text{OH}} \cdot {}^{13}\alpha_{\text{OH}})$. Because errors for KIEs are highly correlated, errors for ${}^{13,2}\gamma_{\text{OH}}$
 values were propagated by a Monte Carlo simulation ($N=10000$ to 100000). This yields
 the best estimate of ${}^{13,2}\gamma_{\text{OH}} = 0.9997 \pm 0.0012$ (2σ). The uncertainty in the gamma
 value (1.2 % at the 2σ level) is smaller than the uncertainty in the individual
 fractionation factors due to the highly correlated nature of the fractionation factors. This
 is a factor of 20 improvement over the previous measurement of this value (${}^{13,2}\gamma_{\text{OH}} =$
 0.98 ± 0.02 , Joelsson et al., 2015).

3.3. Results from Cl oxidation experiments

Isotopic results from Cl oxidation experiments are presented in Table 1. Relationships between the measured isotope ratios (R values) for $\delta^{12}\text{CH}_3\text{D}$ versus $\delta^{13}\text{CH}_4$ and $\delta^{13}\text{CH}_3\text{D}$ versus $\delta^{13}\text{CH}_4$ are shown in Figure 2. As stated above, we were unable to obtain precise f value measurements by FTIR for the Cl oxidation experiments. However, we were able to determine the relative kinetic isotope effects, and use literature values for the kinetic isotope effects to calculate f values. Isotope ratios (2R and $^{13,2}R$) are shown relative to the ^{13}R value because absolute measurements of the kinetic isotope effects for $^{13}\text{CH}_4/^{12}\text{CH}_4$ ($^{13}\text{KIE}_{\text{Cl}} = 1.0621 \pm 0.0001$, 2σ ; Tyler et al. 2000) are higher precision than measurements of $^2\text{KIE}_{\text{Cl}}$ (1.474 ± 0.020 , 2σ , Tyler et al. 2000) and $^{13,2}\text{KIE}_{\text{Cl}}$ (1.60 ± 0.04 , Joelsson et al. 2014). The slope of $\ln(^2R_{\text{sample}}/{}^2R_{\text{initial}}) / \ln(^{13}R_{\text{sample}}/{}^{13}R_{\text{initial}}) = (^2\alpha - 1) / (^{13}\alpha - 1) = ^2\varepsilon / ^{13}\varepsilon$. Here $\varepsilon = \alpha - 1$ is the isotopic fractionation. Our results show that this relationship equals 5.5614 ± 0.0608 (95% confidence interval). A similar relationship for $^{13,2}R$ and ^{13}R gives a $^{13,2}\varepsilon / ^{13}\varepsilon$ value of 6.274 ± 0.064 (95% confidence interval). If we assume the Tyler et al. (2000) value of $^{13}\text{KIE}_{\text{Cl}} = 1.0621 \pm 0.0001$, this give us a $^2\text{KIE}_{\text{Cl}}$ value of 1.482 ± 0.009 and a $^{13,2}\text{KIE}_{\text{Cl}}$ value of 1.579 ± 0.011 . Expressed as isotopic fractionation factors (α values), this gives $^2\alpha_{\text{Cl}} = 0.675 \pm 0.004$, $^{13}\alpha_{\text{Cl}} = 0.9415 \pm 0.0001$, and $^{13,2}\alpha_{\text{Cl}} = 0.633 \pm 0.004$.

This is consistent with some previous measurements of $^2\text{KIE}_{\text{Cl}}$ (1.474 ± 0.020 , Tyler et al. 2000) and $^{13,2}\text{KIE}_{\text{Cl}}$ (1.60 ± 0.04 , Joelsson et al. 2014). Using the Crowley et al. (1999) and Saueressig et al. (1995) value of $^{13}\text{KIE}_{\text{Cl}} = 1.066 \pm 0.002$ gives a $^2\text{KIE}_{\text{Cl}}$ value of 1.558 ± 0.033 and a $^{13,2}\text{KIE}_{\text{Cl}}$ value of 1.635 ± 0.042 . We choose to use the Tyler et al. (2000) results for $^{13}\text{KIE}_{\text{Cl}}$ because they are an order of magnitude more precise than the Crowley et al. (1999) and Saueressig et al. (1995) results.

356 Assuming the Tyler et al. (2000) value for $^{13}\text{KIE}_{\text{Cl}}$, we can use our present results to
357 calculate $^{13,2}\gamma_{\text{Cl}} = ^{13,2}\alpha_{\text{Cl}} / (^2\alpha_{\text{Cl}} * ^{13}\alpha_{\text{Cl}})$. This gives a value of $^{13,2}\gamma_{\text{Cl}} = 0.9965 \pm 0.0007$.
358 Thus, unlike the $^{13,2}\gamma_{\text{OH}}$ value, which is within an uncertainty of 1, there appears to be an
359 inherent $^{13}\text{CH}_3\text{D}$ clumped isotope effect from Cl oxidation of CH_4 . The magnitude of this
360 effect is approximately $-3.5 \text{ ‰} \pm 0.7 \text{ ‰}$. Possible sources of this clumped effect will be
361 discussed in Section 4.2.

4. DISCUSSION

4.1 Importance of $^{13,2}\gamma$ in predicting the clumped isotopic signature of kinetic reactions

The advantage of the present study over previous studies (Joelsson et al., 2014; Joelsson et al., 2015) is the simultaneous measurement of four different isotopologues of methane ($^{12}\text{CH}_4$, $^{13}\text{CH}_4$, $^{12}\text{CH}_3\text{D}$, $^{13}\text{CH}_3\text{D}$) to obtain precise relationships between the kinetic isotope effects for different isotopologues, and thus a precise value for $^{13,2}\gamma_{\text{Cl}}$ and $^{13,2}\gamma_{\text{OH}}$. This value is a measure of the inherent clumped isotope effect during the kinetic reaction, and is important to predicting the effect of the methane sink reactions on the $\Delta^{13}\text{CH}_3\text{D}$ values of atmospheric methane.

There are two end-member scenarios to consider, methane oxidation in an open system and methane oxidation in a closed system. In an open system at steady state with a single irreversible sink reaction, the $^{13,2}\gamma$ parameter is related to the offset between the $\Delta^{13}\text{CH}_3\text{D}$ of the source methane and that of the methane in the system. In particular,

$$\Delta^{13}\text{CH}_3\text{D} = (\Delta^{13}\text{CH}_3\text{D})_{\text{SOURCE}} - \ln(^{13,2}\gamma) \quad \text{Eq. 6}$$

A derivation of this relationship can be found in the Appendix. Eq. 6 indicates that the clumped isotopic composition of a methane reservoir at steady state with a single source and a single sink is equal to the $\Delta^{13}\text{CH}_3\text{D}$ value of the source, plus an offset due to the inherent clumping during the sink reaction. To the first order, tropospheric methane can be modeled as an open system near steady state, with a single dominant sink reaction (OH oxidation). As we show here, OH oxidation has a $^{13,2}\gamma$ value of 0.9997 ± 0.0012 , which suggest that, to the first order, the $\Delta^{13}\text{CH}_3\text{D}$ of tropospheric

methane will be about $0.3\text{ ‰} \pm 1.2\text{ ‰}$ heavier (more clumped) than the combined $\Delta^{13}\text{CH}_3\text{D}$ of the methane sources. This suggests that the $\Delta^{13}\text{CH}_3\text{D}$ of tropospheric methane reflects the $\Delta^{13}\text{CH}_3\text{D}$ of the methane source signatures, with only a minor influence from the sink reaction. This makes $\Delta^{13}\text{CH}_3\text{D}$ a useful tracer for constraining the relative contribution of different methane sources (Joelsson et al., 2015).

In a closed system, the $\Delta^{13}\text{CH}_3\text{D}$ of the residual methane is related to the $\Delta^{13}\text{CH}_3\text{D}$ of the initial methane ($\Delta^{13}\text{CH}_3\text{D}_0$) plus an offset that is a function of the isotopic fractionation factors ($^{13}\alpha$, $^2\alpha$), the fraction of initial methane remaining (f), and the $^{13,2}\gamma$ parameter. This can be written as:

$$\Delta^{13}\text{CH}_3\text{D} = \Delta^{13}\text{CH}_3\text{D}_0 + (^{13}\varepsilon \cdot ^2\varepsilon) \cdot \ln(f) + (^{13,2}\gamma - 1) \cdot (^{13}\alpha \cdot ^2\alpha) \cdot \ln(f) \quad \text{Eq. 7}$$

A full derivation of this equation is given in the Appendix. First, consider the case where $^{13,2}\gamma = 1$ (i.e. there is no inherent clumped kinetic isotope effect), then the $\Delta^{13}\text{CH}_3\text{D}$ value simplifies to

$$\Delta^{13}\text{CH}_3\text{D} \approx \Delta^{13}\text{CH}_3\text{D}_0 + (^{13}\varepsilon \cdot ^2\varepsilon) \cdot \ln(f) \quad \text{Eq. 8}$$

In most normal kinetic processes, both $^{13}\varepsilon$ and $^2\varepsilon$ are negative (i.e., both ^{13}C and D have normal kinetic isotope effects). This means that the $(^{13}\varepsilon \cdot ^2\varepsilon) \cdot \ln(f)$ term will be negative and the $\Delta^{13}\text{CH}_3\text{D}$ of the residual methane will become more negative (relative to the initial $\Delta^{13}\text{CH}_3\text{D}$) as more methane is oxidized as observed in our experiments (Table 1).

As we will show below, $^{13,2}\gamma$ is expected to be slightly less than 1 for bond breaking reactions (i.e., reactions that consume methane). In this case, the third term ($^{13,2}\gamma$) will cause methane to be more clumped than in the “stochastic” ($^{13,2}\gamma = 1$) scenario. Measured $\Delta^{13}\text{CH}_3\text{D}$ values for the OH and Cl oxidation experiments are shown as a function of f in Figure 3. In both cases, all the data show negative $\Delta^{13}\text{CH}_3\text{D}$

values (relative to starting methane) for $f < 1$, as predicted from the closed system isotope effect. However, in both cases, the measured $\Delta^{13}\text{CH}_3\text{D}$ values are higher (more clumped) than predicted from a purely “stochastic” process with no inherent clumped isotope effect ($^{13,2}\gamma = 1$), suggesting that $^{13,2}\gamma < 1$.

4.2 Inherent clumped isotope effects during kinetic reactions

4.2.1 Clumped isotope effects during kinetic reactions – review of the theory

Transition state theory can provide insight into directions, magnitudes, and mechanisms for clumped isotope fractionation during kinetic reactions (Eiler, 2007), and has been used previously to understand the kinetic isotope effects in the carbonate clumped-isotope system (Guo et al., 2009). According to transition state theory, the observed kinetic isotope effects (including clumped kinetic isotope effects) depend largely upon how tightly bound the transition state is relative to the reactants, which is related to the vibrations frequencies (often described in wavenumbers, cm^{-1}). Tables 2 and 3 show vibrational frequencies calculated using the unrestricted MP2 method (Møller and Plesset, 1934) and the aug-cc-pVQZ basis sets (Dunning Jr., 1989; Woon and Dunning Jr., 1993), as calculated in Joelsson et al. (2014, 2015). The calculations were carried out using the Gaussian09 program package (Frisch et al., 2009). In cases where multiple configurations are possible (e.g. $\text{HO-H-}^{12}\text{CH}_2\text{D}$), the average value of the three configurations is given in the table. The calculated transition state geometries are shown in Figure 4. Additional details of the calculations are available in Joelsson et al. (2014, 2015).

4.2.2 Zero point energy and the kinetic clumped isotope effects

The effect of zero-point energy on clumping during methane oxidation is illustrated by comparing the zero point energy shifts (ΔZPE) from the ^{12}C - ^{13}C substitution for $^x\text{CH}_4$ and $^x\text{CH}_3\text{D}$ (Figure 5). D/H substitution for methane (i.e., $^{12}\text{CH}_4$ to $^{12}\text{CH}_3\text{D}$) produces a large ΔZPE of 648.74 cm^{-1} ; that is, $^{12}\text{CH}_3\text{D}$ is about 648.74 cm^{-1} more stable than $^{12}\text{CH}_4$. $^{13}\text{C}/^{12}\text{C}$ substitution of $^x\text{CH}_4$, causes a ΔZPE of 29.76 cm^{-1} , whereas $^{13}\text{C}/^{12}\text{C}$ substitution of the $^x\text{CH}_3\text{D}$ species causes a ΔZPE of 31.85 cm^{-1} . Thus, the difference between the $^{13}\text{C}/^{12}\text{C}$ isotope effect in $^x\text{CH}_3\text{D}$ and $^x\text{CH}_4$ ($\Delta\Delta ZPE$) of 2.09 cm^{-1} is the zero-point energy contribution to the inherent clumped effect of the methane.

If we consider the H-O-H- CH_3 transition state of the $\text{CH}_4 + \text{OH}$ reaction, we observe that ^{13}C - ^{12}C substitution causes ΔZPE of 31.36 cm^{-1} , whereas the corresponding ΔZPE for the H-O-H- CH_2D transition state (i.e. the transition state of the H-abstraction reaction) is 33.34 cm^{-1} . This yields a $\Delta\Delta ZPE$ for the transition state of 1.98 cm^{-1} for the H-abstraction reaction. Because $\Delta\Delta ZPE$ of the reactant (2.09 cm^{-1}) is largely conserved in the transition state (1.98 cm^{-1}) for the H-abstraction reaction (secondary isotope effect), a very minor kinetic clumped effect is expected.

In contrast, the ΔZPE of $^{13}\text{C}/^{12}\text{C}$ substitution of the H-O-D- $^x\text{CH}_3$ transition state (corresponding to the D-abstraction reaction) is 31.90 cm^{-1} , causing a $\Delta\Delta ZPE$ of only 0.54 cm^{-1} . Therefore, a significant kinetic clumped isotope effect is expected for the D-abstraction reaction (primary D isotope effect).

The clumped isotope effect for methane largely comes from the $\Delta\Delta ZPE$ of the ^{13}C -D stretching vibration of methane (Table 2). In the transition state for the D-abstraction

reaction, this stretching vibration becomes unbound, effectively removing the interaction between ^{13}C and D, the origin of the clumped effect. The weakening of the ^{13}C -D bond in the transition state offsets some of energy gain from clumping. As a result, the ^{13}C -D bond does not have as strong of a stabilizing effect in the more weakly bound transition state than in the more strongly bound reactants.

4.2.3 Using Transition State Theory to Calculate $^{13,2}\gamma$ Values

As illustrated in Section 4.2.2, we predict a large difference in the inherent clumped isotope effect during the D-abstraction (primary) versus H-abstraction (secondary) reactions. Unfortunately, it is difficult to experimentally separate the clumped effect due to primary versus secondary effects without measuring the products. Transition state theory allows us calculate $^{13,2}\gamma$, and examine the clumped isotope effect from the $\text{CH}_4 + \text{OH}$ reactions. To examine the influence of different factors on the $^{13,2}\gamma_{\text{OH}}$ value, we look at four factors – the imaginary frequency ratio term (imfreq), the tunneling term (tunnel), the inherent clumping of the transition state (Qts), and the inherent clumping of the reactant (Qreact) (Bigeleisen and Wolfsberg, 1958; Lasaga and Gibbs, 1991). These terms are

$$\text{imfreq} = \left(\frac{^{13,2}\nu^\ddagger * \nu^\ddagger}{^{13}\nu^\ddagger * ^2\nu^\ddagger} \right) \quad \text{Eq. 9}$$

$$\text{tunnel} = \left(\frac{^{13,2}\Gamma_{\text{tunn}}^\ddagger * \Gamma_{\text{tunn}}^\ddagger}{^{13}\Gamma_{\text{tunn}}^\ddagger * ^2\Gamma_{\text{tunn}}^\ddagger} \right) = \left(\frac{\left(\left(1 + \frac{1}{24} * \left[\frac{h^{13,2}\nu^\ddagger}{k_B T} \right]^2 \right) * \left(1 + \frac{1}{24} * \left[\frac{h\nu^\ddagger}{k_B T} \right]^2 \right) \right)}{\left(\left(1 + \frac{1}{24} * \left[\frac{h^{13}\nu^\ddagger}{k_B T} \right]^2 \right) * \left(1 + \frac{1}{24} * \left[\frac{h^2\nu^\ddagger}{k_B T} \right]^2 \right) \right)} \right) \quad \text{Eq. 10}$$

471 Qts =

472
$$\left(\frac{\left[\prod_{i=1}^{14} \frac{h^{13,2} \nu_i^\ddagger}{k_B T} / \left(1 - \exp\left(-\frac{h^{13,2} \nu_i^\ddagger}{k_B T} \right) \right) \right] \left[\prod_{i=1}^{14} \frac{h \nu_i^\ddagger}{k_B T} / \left(1 - \exp\left(-\frac{h \nu_i^\ddagger}{k_B T} \right) \right) \right]}{\left[\prod_{i=1}^{14} \frac{h^{13} \nu_i^\ddagger}{k_B T} / \left(1 - \exp\left(-\frac{h^{13} \nu_i^\ddagger}{k_B T} \right) \right) \right] \left[\prod_{i=1}^{14} \frac{h^2 \nu_i^\ddagger}{k_B T} / \left(1 - \exp\left(-\frac{h^2 \nu_i^\ddagger}{k_B T} \right) \right) \right]} \right) * \exp \left(-\frac{h}{2k_B T} (\sum_{i=1}^{14} {}^{13,2} \nu_i^\ddagger + \right.$$

473
$$\left. \sum_{i=1}^{14} \nu_i^\ddagger - \sum_{i=1}^{14} {}^{13} \nu_i^\ddagger - \sum_{i=1}^{14} {}^2 \nu_i^\ddagger) \right)$$
 Eq, 11

474

475 Qreact =

476
$$\left(\frac{\left[\prod_{i=1}^9 \frac{h^{13,2} \nu_i^R}{k_B T} / \left(1 - \exp\left(-\frac{h^{13,2} \nu_i^R}{k_B T} \right) \right) \right] \left[\prod_{i=1}^9 \frac{h \nu_i^R}{k_B T} / \left(1 - \exp\left(-\frac{h \nu_i^R}{k_B T} \right) \right) \right]}{\left[\prod_{i=1}^9 \frac{h^{13} \nu_i^R}{k_B T} / \left(1 - \exp\left(-\frac{h^{13} \nu_i^R}{k_B T} \right) \right) \right] \left[\prod_{i=1}^9 \frac{h^2 \nu_i^R}{k_B T} / \left(1 - \exp\left(-\frac{h^2 \nu_i^R}{k_B T} \right) \right) \right]} \right) * \exp \left(-\frac{h}{2k_B T} (\sum_{i=1}^9 {}^{13,2} \nu_i^R + \right.$$

477
$$\left. \sum_{i=1}^9 \nu_i^R - \sum_{i=1}^9 {}^{13} \nu_i^R - \sum_{i=1}^9 {}^2 \nu_i^R) \right)$$
 Eq. 12

478 Here, $\nu^\ddagger$ is the imaginary frequency along the reaction coordinate, $\nu_i^\ddagger$ is the i -th (real)

479 vibrational frequency of the transition state, ν_i^R is the i -th vibrational frequency of the

480 reactant (CH₄). The prefix superscript ^{13,2} refers to the ¹³CH₃D isotopologue, ¹³ refers to

481 the ¹³CH₄ isotopologue, ² refers to the ¹²CH₃D isotopologue, and no prefix superscript

482 refers to the ¹²CH₄ isotopologue. Transition state theory states that:

483
$${}^{13,2} \gamma = \frac{{}^{13,2} k^* k}{{}^{13} k^* {}^2 k} = (\text{imfreq}) * (\text{tunnel}) * (Qts) / (Qreact)$$
 Eq, 13

484 Taking the natural logarithm of both terms allows us to separate the four terms and

485 examine the effect of different factors on the inherent clumped isotope effect.

486
$$\ln ({}^{13,2} \gamma) = \ln(\text{imfreq}) + \ln(\text{tunnel}) + \ln(Qts) - \ln(Qreact)$$
 Eq. 14

Calculations were carried out separately for each of the four factors, for primary (D-abstraction), secondary (H-abstraction), and total isotope effects, for a range of temperatures. Results are shown in Figure 6. Several important observations can be made. One is that the tunneling and imaginary frequency factors do not significantly affect the $^{13,2}\gamma_{\text{OH}}$ value within the Wigner tunneling approximation. Rather, as described above, the clumping effect comes from the difference $\Delta\Delta\text{ZPE}$ of the transition state (Qts) and the reactants (Qreact). This difference is largest for primary isotope effects (D-abstraction reactions) and is larger at lower temperatures than at higher temperatures. As the temperature increases, the clumped isotope effect disappears, largely due to the reduction of the inherent clumped signature of the reactants. The Wigner correction for tunneling (equation 10) assumes a parabolic potential at the transition state (Wigner, 1932; Bell, 1959). This tunneling correction may not be accurate at lower temperatures, and thus the clumped effect from tunneling might be larger than predicted here. A more accurate potential energy surface would be required to make a more precise estimate of the clumped effect due to tunneling.

Although the primary isotope effect (D-abstraction) is expected to produce a significant clumped isotopologue fractionation, we observe only a small clumped effect. This is because the majority of methane is oxidized through the H-abstraction channel, which is an order of magnitude faster (for both Cl and OH oxidation) than the D-abstraction channel. According to the estimated rates for the $\text{CH}_4 + \text{OH}$ reaction (Table 5), 96.6% of the total CH_4 loss for the mono-deuterated species ($^{12}\text{CH}_3\text{D}$ and $^{13}\text{CH}_3\text{D}$) is due to H-abstraction, with only 3.4% being due to D-abstraction.

4.2.4 Vibrational mode-specific clumping effects

The Qts and Qreact terms in the transition state expression can be further decomposed to analyze the effect of individual vibrational motions on the inherent clumped effects in the transition state and the reactants. In particular, we can look at the “clumped” contribution from a single normal vibrational mode, i , in either the reactant or the transition state:

$$\gamma_i = \frac{\left[\frac{h^{13,2}\nu_i}{k_B T} / \left(1 - \exp\left(-\frac{h^{13,2}\nu_i}{k_B T}\right) \right) \right] \left[\frac{h\nu_i}{k_B T} / \left(1 - \exp\left(-\frac{h\nu_i}{k_B T}\right) \right) \right]}{\left[\frac{h^{13}\nu_i}{k_B T} / \left(1 - \exp\left(-\frac{h^{13}\nu_i}{k_B T}\right) \right) \right] \left[\frac{h^2\nu_i}{k_B T} / \left(1 - \exp\left(-\frac{h^2\nu_i}{k_B T}\right) \right) \right]} \\ * \exp\left(-\frac{h}{2k_B T} ({}^{13,2}\nu_i + \nu_i - {}^{13}\nu_i - {}^2\nu_i)\right) \quad \text{Eq. 15}$$

In Eq. 15, ν_i represents the vibrational frequency of the i -th vibration in either the reactant (ν_i^R) or the transition state ($\nu_i^\ddagger$). Values of $\ln(\gamma_i)$ at 298 K are tabulated in Table 2 for the reactant (ground-state methane) and Table 3 for the CH₄+OH transition state.

As illustrated in Table 2, the inherent clumped isotope effect of the CH₄ reactant is due primarily to the symmetric stretching vibration (3072 cm⁻¹), with contributions from the twisting motion (1584 cm⁻¹). The bending motion (1350 cm⁻¹) causes a small anti-clumping effect, and the asymmetric stretch (3210 cm⁻¹) has little effect. Because H-D isotopic substitution changes the symmetry of the molecule, the C-H asymmetric stretch is only doubly degenerate for the mono-deuterated species, with the third C-H asymmetric stretch being replaced by a C-D stretching motion (around 2300 cm⁻¹). This change causes a small anti-clumping effect.

For $^{12}\text{CH}_4$ and $^{13}\text{CH}_4$, the carbon atom is located at the center of mass of the molecule. For this type of system, the frequency of the symmetric stretch vibration will be completely insensitive to the mass of the central atom (Rosenthal, 1934). For $^{12}\text{CH}_3\text{D}$ and $^{13}\text{CH}_3\text{D}$, the carbon atom is no longer located at the center of mass, and its position will change during the symmetric stretch vibration. As a result, the frequency of this vibrational motion will depend on the mass of the carbon atom (see Eqs. 26 and 27 of Rosenthal, 1934). This is the cause of the large inherent clumping effect of the C-H stretching vibration. In essence, the breaking of the symmetry of the molecule with deuterium isotopic substitution causes subsequent carbon isotopic substitution to have a larger impact on the CH_3D system than the CH_4 system. This is shown schematically in Figure 7. A similar argument can explain the inherent clumping effect of the HCH twist vibration. In both cases, the C atom is stationary during the vibration in the CH_4 species, but moves with the vibration in the CH_3D species. Because of this, carbon isotopic substitution does not affect the energy of the vibration in CH_4 but lowers the energy (i.e. stabilizes the molecule) in the monodeuterated (CH_3D) species.

For the other vibrational modes of CH_4 , there is an anti-clumping effect. In particular, the HCH bend vibrations and the replacement of third C-H asymmetric stretch with a C-D stretch both cause anti-clumping effects. Although ^{13}C substitution results in a net stabilization for both the CH_4 and CH_3D , the stabilization effect is smaller for the CH_3D species than for the CH_4 . This is interesting in comparison to CO_2 (Wang et al., 2004), where all four vibrational modes contribute to a positive clumping effect for $\Delta^{18}\text{O}^{13}\text{C}^{16}\text{O}$ (approximately equal to Δ_{47}) and where the C-O asymmetric stretch is the dominant contributor to clumping.

A similar pattern is observed in the transition state as in the reactants (Table 4). The “new” vibrational modes (the O-H stretch and CHO bend and stretch vibrations) do not contribute significantly to the clumping effects, with the dominant clumping effect for the H-abstraction reaction coming from the vibrational motion analogous to the C-H symmetric stretching motion and the HCH twisting motion. The C-H symmetric stretching motion has no contribution to clumping in the D-abstraction reaction. This is likely because the symmetry of the stretching motion is preserved during the D-abstraction reaction (i.e. 3 “equivalent” hydrogens) but destroyed in the H-abstraction reaction (2 hydrogens and 1 deuterium). As for the reactants, it is the preservation or destruction of local symmetry in the transition state that causes the C-H symmetric stretch to have no clumping effect for the D-abstraction reaction but a large clumping effect for the H-abstraction reaction.

4.2.5 The CH₄+Cl reaction

The analyses performed above (4.2.3 and 4.2.4) is repeated for the CH₄+Cl reaction using the transition-state frequencies reported in Joelsson et al. (2014). Results are tabulated in Table 4. Similar results are observed for the H-abstraction channel, with the C-H “symmetric” stretch causing a large clumped effect in the transition state (as in the reactants), and the transition state and reactants having very similar clumped effects. The D-abstraction reaction appears anti-clumped in the transition state, causing the apparent clumped effect during the primary (D-abstraction) reaction to be larger than the inherent clumped effect of the reactant methane. In this case, the contribution from the imaginary frequency term (D-transfer mode) is non-negligible and contributes about 1/3 of the observed anti-clumping effect. The other 2/3

come from the HCH bending vibration. As with the CH₄+OH reaction, the D-abstraction channel is significantly slower than the H-abstraction channel, so the large primary clumped effect is largely not observed. Our calculations show that transition state theory predicts a clumped effect that is an order of magnitude smaller than the observed value. There is also a large discrepancy in the ¹³KIE_{Cl} values. These discrepancies are probably due to an underestimate of the tunneling effect (Corchado et al., 2000). It is possible that a higher level of theory or a more accurate (multi-dimensional) tunneling correction could resolve some of these discrepancies (Roberto-Neto et al., 1998; Corchado et al., 2000). We attribute the discrepancies to inadequacies in the Wigner tunneling approximation for the CH₄+Cl reaction, which has been previously demonstrated (Roberto-Neto et al., 1998; Corchado et al., 2000). Higher level tunneling corrections are necessary to fully assess the impact of tunneling on the clumped kinetic isotope effects, for both the CH₄+Cl reaction as well as and the CH₄+OH reaction at lower temperatures.

4.2.6 Clumped isotope effects for ¹²CH₂D₂

Recent advances in technology (Young et al., 2016) allow precise measurements of $\Delta^{12}\text{CH}_2\text{D}_2 = \ln([^{12}\text{CH}_2\text{D}_2] * [^{12}\text{CH}_4] / [^{12}\text{CH}_3\text{D}]^2)$ at natural abundances. Kinetic isotope effects for ¹²CH₂D₂ have been measured in the past using isotopically-enriched samples (e.g. Feilberg et al., 2005), although higher-precision measurements will likely be required for the interpretation of atmospheric isotope ratios. Although ¹²CH₂D₂ was not measured in the present study, the transition state structure calculated here (Figure 4) were used to compute vibrational frequencies for ¹²CH₂D₂ and the relevant transition states (Table 7). Using the same level of theory and assumptions used above for

$^{13}\text{CH}_3\text{D}$, kinetic isotope effects for the reaction of $^{12}\text{CH}_2\text{D}_2$ with OH and Cl were calculated (Table 5). Due to the different symmetries of the three relevant species ($^{12}\text{CH}_2\text{D}_2$, $^{12}\text{CH}_3\text{D}$, and $^{12}\text{CH}_4$), we were unable to provide a mode-specific analysis as performed in Sections 4.2.4 and 4.2.5. However, we were able to calculate clumped isotope effects, which are reported in Table 6.

Although the $^{12}\text{CH}_2\text{D}_2$ kinetic isotope effect for OH oxidation has been measured previously (Gierczak et al., 1997), the large uncertainties in that study make comparison to our calculations difficult. Assuming the calculations are accurate, OH oxidation will have a significant (-73.4‰) effect on the $\Delta^{12}\text{CH}_2\text{D}_2$ value of atmospheric methane. Therefore, whereas $\Delta^{13}\text{CH}_3\text{D}$ of atmospheric methane will primarily provide constraints on the source strengths, $\Delta^{12}\text{CH}_2\text{D}_2$ can provide information on sink reactions as well.

For the Cl reaction, our calculated kinetic isotope effect for $^{12}\text{CH}_2\text{D}_2$ (2.184) is smaller than that measured by Feilberg et al. (2005)'s relative rate experiments (2.45 ± 0.05). This is similar to the discrepancy observed in the $^{12}\text{CH}_3\text{D}$ and $^{13}\text{CH}_3\text{D}$, which we attribute to the inadequacy of the Wigner tunneling correction in describing the tunneling contribution to the kinetic isotope effect for the $\text{CH}_4 + \text{Cl}$ reaction.

4.3 Predicting the $\Delta^{13}\text{CH}_3\text{D}$ of Tropospheric Methane

To obtain an estimate for the $\Delta^{13}\text{CH}_3\text{D}$ of tropospheric methane, we consider the simple two box (Northern Hemisphere and Southern Hemisphere) model of Tans (1997) using the input parameters from Table S3 of Kai et al. (2011). This model was chosen for its simplicity (a two-box atmospheric model) and its success in explaining changes in

methane concentrations, isotope ratios, and interhemispheric differences in concentrations and isotope ratios during the late twentieth century (Kai et al., 2011). Kai et al. (2011) have 9 methane sources and provide estimated source input fluxes for both hemispheres and estimated isotopic signatures ($\delta^{13}\text{CH}_4$ and $\delta^{12}\text{CH}_3\text{D}$) of the sources. We assigned $\Delta^{13}\text{CH}_3\text{D}$ values to the different sources based on the measured $\Delta^{13}\text{CH}_3\text{D}$ values of different sources from Wang et al. (2015), as shown in Table 8. The seven sources given by Kai et al. (2011) are tundra, lakes, swamps and marshes, agriculture, landfills, fossil fuels, biomass burning, and gas hydrates.

For the lake input, we assumed a $\Delta^{13}\text{CH}_3\text{D}$ of 3.0 ‰, based on the measured values from Upper Mystic Lake from Wang et al. (2015) (Table 8). For the swamp input, we chose a value of 0.7 ‰ based on measurements from Cedar Swamp in Massachusetts, USA (Wang et al. 2015). We assumed both the agricultural input and the termite input had $\Delta^{13}\text{CH}_3\text{D}$ values of 1.0 ‰, which is the approximate measured value for methane from bovine rumen (“Sally” from Wang et al. 2015). We assume that fossil fuels and biomass burning have an approximately thermogenic signature of around 3.0 ‰, on the basis of measurements from the Marcellus and Utica formations from Stolper et al. (2014) and Wang et al. (2015). For gas hydrates, we assume $\Delta^{13}\text{CH}_3\text{D}$ values of around 6.0 ‰, based on data from the Northern Cascadia Margin in Wang et al. (2015). For tundra, we assume a value of 3.0 ‰. $\Delta^{13}\text{CH}_3\text{D}$ values have not been measured for tundra, landfills, biomass burning, or termites. The results from the model are relatively insensitive to the chosen values of $\Delta^{13}\text{CH}_3\text{D}$ for the gas hydrates, tundra, and termite sources, due to their relatively small contribution (approximately 10 Tg yr⁻¹, 10 Tg yr⁻¹, and 20 Tg yr⁻¹, respectively), to the total input (approximately 550 Tg

yr⁻¹). Biomass burning, because it is a high temperature process, is assumed to be approximately in equilibrium, and thus the $\Delta^{13}\text{CH}_3\text{D}$ value will depend upon the temperature. A value of 3.0 ‰ was chosen for biomass burning, which corresponds to a temperature of around 150°C. Higher temperature would produce $\Delta^{13}\text{CH}_3\text{D}$ values close to 0 ‰, so we also tested biomass burning $\Delta^{13}\text{CH}_3\text{D}$ values of 0.0 ‰ to investigate the sensitivity to this input parameter. We assumed that landfills produce predominantly biogenic methane with $\Delta^{13}\text{CH}_3\text{D}$ values around 1.0 ‰, similar to values measured from the bovine rumen samples (Wang et al., 2015).

The sinks of tropospheric methane include OH oxidation, soil uptake, and stratospheric uptake. We assume relative sink strengths from Mikaloff Fletcher et al. (2004), and isotopic fractionation factors for OH oxidation from the present study and for soil uptake ($^2\alpha_{\text{soil}}$ and $^{13}\alpha_{\text{soil}}$) from Snover and Quay (1990). We assumed a $^{13,2}\alpha_{\text{soil}}$ value for soil uptake by assuming that $^{13,2}\gamma_{\text{soil}} = 1$ for biological oxidation of methane. For the present study, we do not include stratospheric exchange and include only OH oxidation and soil uptake. We assumed a relative contribution of soil uptake of about 5.6%, compared to 94.4% of methane removal from OH oxidation. We assumed an interhemispheric exchange lifetime of 1 year and a methane removal lifetime of 8.7 years. Our calculations gave us a total sink $^2\alpha$ value of 0.7592, $^{13}\alpha$ value of 0.9933, and a $^{13,2}\alpha$ value of 0.7538.

Our model predicts a $\Delta^{13}\text{CH}_3\text{D}$ value of around 3.1 ‰ for the northern hemisphere and around 3.0 ‰ for the southern hemisphere. If we assume the $\Delta^{13}\text{CH}_3\text{D}$ value of biomass burning is 0.0 ‰, we calculate a tropospheric $\Delta^{13}\text{CH}_3\text{D}$ value of 2.9 ‰ for the northern hemisphere and 2.8 ‰ for the southern hemisphere. The calculated

0.1 ‰ interhemispheric difference in $\Delta^{13}\text{CH}_3\text{D}$ is on the order of magnitude of the precision of the current measurements. Note that the calculation above assumes a steady state and does not account for the seasonal cycle of methane concentrations and isotopes, which could increase the difference in $\Delta^{13}\text{CH}_3\text{D}$ between the two hemispheres. It also does not account for the return flux of methane impacted by stratospheric fractionation, which could have a significant effect on the clumped signature of tropospheric methane.

For a first-order estimate of seasonal effects, we assume that seasonal changes in the rate of OH oxidation account for the majority of the seasonal changes in methane concentrations and isotopic signatures. Given that, we can make the approximation that:

$$\delta = \delta_0 - \varepsilon \frac{\Delta[\text{CH}_4]}{[\text{CH}_4]} \quad \text{Eq. 16}$$

If we assume a starting $[\text{CH}_4] = 1800 \text{ ppm}$, $\delta^{13}\text{C} = -48 \text{ ‰}$, $\delta^2\text{H} = -86 \text{ ‰}$, and $\Delta^{13}\text{CH}_3\text{D} = 3.0 \text{ ‰}$, then a seasonal cycle of 30 ppb causes a $\Delta^{13}\text{CH}_3\text{D}$ shift of around 0.2 ‰, ranging from 2.9‰ to 3.1‰. This first-order approximation suggests that seasonal variations in the $\Delta^{13}\text{CH}_3\text{D}$ value of atmospheric methane should be visible at a 0.1 ‰ level of precision. Examining seasonal variations in methane concentrations, $\delta^{13}\text{C}$, δD , and $\Delta^{13}\text{CH}_3\text{D}$ may help to constrain the source of seasonal variability and what portion can be attributed to changes in the sink reaction (OH oxidation) versus changes in various sources.

4.5 Measurement of multiply-substituted isotopologues in atmospheric methane

To our knowledge, the only measurements of multiply-substituted isotopologues of methane in the atmosphere have been those of Mroz et al. (1989). Mroz et al. (1989) measured $^{12}\text{CD}_4$ (or $^{13}\text{CHD}_3$) concentrations over 500 times larger than predicted from a statistical distribution of isotopes. Mroz et al. (1989) acknowledge the possibility that the reason for such large enrichments could be related to the anthropogenic release of $^{12}\text{CD}_4$ for use as an atmospheric tracers (e.g. Cowan et al., 1976, Fowler and Berr, 1983, Alei et al., 1987). CD_4 has also been used as a tracer in other settings, such as geochemical experiments (e.g. Stalker et al., 2009). It is also possible that the measurement was an analytical error (Stolper et al., 2014), and unless it can be reproduced independently it is difficult to rule out this possibility. Modeling (Kaye and Jackman, 1990) suggests a chemical source for this anomalous enrichment is unlikely, leaving an anthropogenic source (i.e. tracer releases) or an analytical error as the most likely explanations.

In contrast to tracers like $^{12}\text{CD}_4$ (or $^{13}\text{CD}_4$), $^{12}\text{CH}_2\text{D}_2$ and $^{13}\text{CH}_3\text{D}$ do not have a long history of anthropogenic release into the environment. They are also significantly more abundant than $^{12}\text{CH}_4$, so environmental releases will likely have a minor effect on the atmospheric concentration of these isotopologues. Therefore, measurement of tropospheric $^{12}\text{CH}_2\text{D}_2$ and $^{13}\text{CH}_3\text{D}$ will likely provide information about both atmospheric sources ($^{12}\text{CH}_2\text{D}_2$ and $^{13}\text{CH}_3\text{D}$) and sinks ($^{12}\text{CH}_2\text{D}_2$ but not $^{13}\text{CH}_3\text{D}$). The analytical techniques exist to measure these isotopologues at natural abundance (Ono et al., 2014; Stolper et al., 2014; Young et al., 2016), particularly with methane preconcentration and purification. Improved understanding of the chemistry of these doubly-substituted isotopologues, as well as the development of techniques to measure

709 them in the atmosphere, will improve our understanding of the global biogeochemical
710 cycle of methane and its changes over time.

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5. CONCLUSIONS

This study reports the clumped kinetic isotope effects for oxidation of $^{13}\text{CH}_3\text{D}$ by OH and Cl. It is demonstrated that the effect of OH oxidation on the clumped isotopic composition of atmospheric methane (in an open system) is relatively minor, on the order of $0.3\text{‰} \pm 1.2\text{‰}$. This is an order of magnitude reduction in uncertainty over previous measurements (Joelsson et al., 2015). Cl oxidation has a larger effect on the clumped isotopic signature of atmospheric methane ($3.5\text{‰} \pm 0.7\text{‰}$), but is a relatively minor sink pathway in the troposphere ($< 5\%$), and thus will not affect the average global $\Delta^{13}\text{CH}_3\text{D}$ value of tropospheric methane significantly. It might be important when considering stratospheric methane or local effects, however.

Quantum chemistry and transition state theory are used to argue that gas phase oxidation reactions of methane should result in the residual methane being more clumped than expected, due to the relative weakness of the ^{13}C -D bond in the transition state producing less clumped effect than the reactants. This is particularly true for deuterium extraction reactions; hydrogen extraction reactions, in contrast, are expected to produce a very small clumped kinetic isotope effect. A simple box model is used to predict that the $\Delta^{13}\text{CH}_3\text{D}$ of atmospheric methane is around 3.0‰ , with a 0.1‰ interhemispheric difference and a seasonal cycle on the order of 0.2‰ . Based on the results of both the experimental and theoretical calculations, we predict that measurements of atmospheric $\Delta^{13}\text{CH}_3\text{D}$ will provide insight into atmospheric source signatures, whereas $\Delta^{12}\text{CH}_2\text{D}_2$ will provide valuable information about atmospheric sink processes. Combining measurements of $\Delta^{13}\text{CH}_3\text{D}$ and $\Delta^{12}\text{CH}_2\text{D}_2$ with more routine measurements of $\delta^{13}\text{C}$ and δD will improve our

understanding of atmospheric methane cycling and methane biogeochemistry. This work opens the door for the interpretation of future measurements of the $\Delta^{13}\text{CH}_3\text{D}$ and $\Delta^{12}\text{CH}_2\text{D}_2$ of atmospheric methane.

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8. TABLES

Table 1: Fraction of initial methane remaining (f values) and isotopic results ($\delta^{12}\text{CH}_3\text{D}$, $\delta^{13}\text{CH}_4$, $\Delta^{13}\text{CH}_3\text{D}$) relative to initial methane isotopic composition for all samples analyzed. 95% confidence intervals for $\delta^{12}\text{CH}_3\text{D}$ and $\delta^{13}\text{CH}_4$ are better than 0.3‰. 95% confidence intervals for $\Delta^{13}\text{CH}_3\text{D}$ values are reported in the table. The majority of the uncertainty in $\delta^{12}\text{CH}_3\text{D}$ and $\delta^{13}\text{CH}_4$ is due to uncertainty in the initial methane isotopic composition; analytical uncertainty for individual samples was better than 0.1‰ for both $\delta^{12}\text{CH}_3\text{D}$ and $\delta^{13}\text{CH}_4$.

	f	$\sigma(f)$	$\delta^{12}\text{CH}_3\text{D}$ (‰)	$\delta^{13}\text{CH}_4$ (‰)	$\Delta^{13}\text{CH}_3\text{D}$ (‰)	Unc (‰)
Control-1-a	1.000	0.000	-0.2	0.3	-0.09	0.35
Control-1-b	1.000	0.000	-0.4	-0.8	0.14	0.17
Control-1-c	1.000	0.000	-0.3	-0.5	0.07	0.15
OH-1-0	1.000	0.000	-0.1	0.3	0.02	0.29
OH-1-1	0.806	0.005	65.3	2.2	-0.34	0.26
OH-1-2	0.750	0.010	90.5	2.6	-0.58	0.22
OH-1-3	0.522	0.071	159.3	9.3	-1.25	0.49
OH-2-0	1.000	0.000	1.1	0.3	-0.06	0.19
OH-2-1			82.2	2.5	-0.33	0.19
OH-2-2			128.6	3.7	-0.53	0.17
OH-3-0	1.000	0.000	0.0	0.2	-0.21	0.67
OH-3-1	0.770	0.005	74.2	2.3	-0.33	0.15
OH-3-3	0.461	0.005	217.6	5.5	-1.13	0.17
OH-4-1	0.840	0.002	52.4	1.8	-0.22	0.22
OH-4-3	0.526	0.002	175.7	4.8	-0.78	0.32
OH-5-0	1.000	0.000	-0.1	0.3	0.12	0.33
OH-5-1	0.819	0.003	59.2	2.0	-0.30	0.29
OH-5-2	0.623	0.004	129.9	3.5	-0.70	0.63
OH-5-3	0.489	0.002	196.6	4.2	-0.82	0.28
Cl-2-1	0.562	*	202.7	34.1	-9.44	0.46
Cl-3-1	0.799	*	71.6	13.0	-3.68	0.20
Cl-3-2	0.627	*	160.9	27.5	-7.69	0.40
Cl-3-3	0.446	*	296.3	48.2	-13.24	0.27
Cl-4-1	0.796	*	72.8	13.3	-3.81	0.17
Cl-4-2	0.666	*	134.8	23.9	-6.65	0.35
Cl-4-3	0.508	*	241.0	40.3	-11.37	0.19

* f values for Cl oxidation experiments were calculated from $\delta^{13}\text{CH}_4$ values assuming a $^{13}\text{KIE}_{\text{Cl}}$ of 1.0621 from Tyler et al. (2000).

Table 2: Normal mode vibrational frequencies for different isotopologues of CH₄, as well as contribution of each normal mode to ^{13,2}γ_{OH} value at 298 K. Vibrational frequencies from Joelsson et al. (2014, 2015). γ_i is defined in Eq. 15.

	Mode Description	¹² CH ₄ (cm ⁻¹)	¹³ CH ₄ (cm ⁻¹)	¹² CH ₃ D (cm ⁻¹)	¹³ CH ₃ D (cm ⁻¹)	1000 ln(γ _i)
1	HCH Bend	1350.08	1341.71	1193.16	1184.99	-1.08
2	HCH Bend	1350.08	1341.71	1193.16	1184.99	-1.08
3	HCH Bend	1350.09	1341.72	1344.39	1337.16	-1.92
4	HCH Twist	1584.76	1584.76	1520.79	1519.84	1.68
5	HCH Twist	1584.76	1584.76	1520.80	1519.84	1.68
6	C-H Stretch (Sym)	3072.73	3072.73	3113.43	3109.55	8.13
7	C-H Stretch (Asym)	3210.84	3199.37	3210.59	3199.09	0.07
8	C-H Stretch (Asym) *C-H (or D) Stretch	3210.84	3199.37	3210.59	3199.09	0.07
9	(Asym)	3210.84	3199.38	2320.64	2309.31	-1.65
	ZPE / Total	9962.51	9932.75	9313.77	9281.93	5.91

* Mode 9 becomes C-D stretch in the monodeuterated (C_{3v}) species.

Table 3: Normal mode vibrational frequencies for different isotopologues of the H-O-H-CH₃ transition state, as well as contribution of each normal mode to $^{13,2}\gamma_{\text{OH}}$ value at 298 K for both primary (D-abstraction, γ_i^{D}) and secondary (H-abstraction, γ_i^{H}) isotope effects. Vibrational frequencies from Joelsson et al. (2015). Mode descriptions are approximate and are based on corresponding modes of reactants (CH₄, OH) and descriptions given by Lasaga and Gibbs (1991). γ_i is defined in Eq. 15. Given vibrational frequencies for transition states (HO-H-¹²CH₂D and HO-H-¹³CH₂D) are an average of the vibrational frequencies for the 3 different possible configurations.

	(Approx) Mode Description	HO-H- ¹² CH ₃ (cm ⁻¹)	HO-H- ¹³ CH ₃ (cm ⁻¹)	HO-D- ¹² CH ₃ (cm ⁻¹)	HO-D- ¹³ CH ₃ (cm ⁻¹)	HO-H- ¹² CH ₂ D (cm ⁻¹)	HO-H- ¹³ CH ₂ D (cm ⁻¹)	1000 ln(γ_i^{D})	1000 ln(γ_i^{H})
<i>i</i>	Reaction Coordinate	-1734.32	-1734.28	-1316.36	-1316.31	-1723.50	-1723.46	-0.01	-0.01
1	Dihedral Torsion	7.10	7.10	7.06	7.06	6.88	6.88	0.00	0.00
2	CHO Bend	314.54	314.29	269.42	269.39	278.38	277.89	-0.13	0.11
3	CHO Bend	323.01	322.12	287.61	287.09	317.36	316.65	-0.26	-0.12
4	CHO Stretch	794.51	782.40	776.26	763.34	767.85	757.29	0.75	-2.23
5	CHO Stretch	921.92	916.94	910.67	906.77	914.12	907.76	-1.53	1.86
6	HCH Bend	1215.07	1209.61	1034.46	1029.21	1126.04	1119.90	-1.00	0.74
7	HCH Bend	1305.43	1301.29	1142.44	1138.53	1206.94	1201.81	-0.77	1.35
8	HCH Bend	1379.95	1374.27	1270.62	1265.42	1288.72	1286.22	-1.10	-5.50
9	HCH Twist	1486.84	1484.97	1474.18	1471.50	1408.03	1405.33	1.39	1.36
10	HCH Twist	1502.10	1501.40	1475.04	1472.32	1462.95	1461.39	3.50	1.49
11	CH Symm. Stretch	3114.59	3112.50	3114.51	3112.43	3166.99	3160.51	0.00	9.22
12	CH Asymm. Stretch	3250.81	3238.57	3250.66	3238.41	3252.07	3239.76	0.04	0.14
13	CH Asymm. Stretch	3253.67	3241.36	3253.47	3241.14	2343.81	2332.10	0.04	-2.66
14	OH Stretch	3807.53	3807.53	3807.46	3807.46	3807.52	3807.52	0.00	0.00
	Tunneling							-0.02	-0.01
	ZPE / Total	11338.52	11307.16	11036.93	11005.03	10673.84	10640.50	0.92	5.75

Table 4: Normal mode vibrational frequencies for different isotopologues of the Cl-H-CH₃ transition state, as well as contribution of each normal mode to $^{13,2}\gamma_{\text{Cl}}$ value at 298 K for both primary (D-abstraction, γ_i^{D}) and secondary (H-abstraction, γ_i^{H}) isotope effects. Vibrational frequencies from Joelsson et al. (2014). Mode descriptions are approximate and are based on corresponding modes of reactants (CH₄, OH) and descriptions given by Lasaga and Gibbs (1991). γ_i is defined in Eq. 15. Given vibrational frequencies for transition states (Cl-H-¹²CH₂D and Cl-H-¹³CH₂D) are an average of the vibrational frequencies for the 3 different possible configurations.

	Mode	Cl-H- ¹² CH ₃	Cl-H- ¹³ CH ₃	Cl-D- ¹² CH ₃	Cl-D- ¹³ CH ₃	Cl-H- ¹² CH ₂ D	Cl-H- ¹³ CH ₂ D	1000 ln(γ_i^{D})	1000 ln(γ_i^{H})
Imfreq	H transfer	-1146.03	-1138.79	-874.84	-868.14	-1140.34	-1132.86	-1.35	-0.25
1	CHCl bnd	364.90	364.73	311.27	311.27	306.50	306.21	-0.11	0.05
2	CHCl bnd	364.92	364.75	311.28	311.28	363.30	363.07	-0.11	0.04
	CHCl str /								
3	bnd	507.45	498.97	500.03	491.82	496.27	488.89	-0.33	-1.10
4	CHCl str	974.16	971.90	817.55	814.93	906.54	903.96	0.15	0.32
5	CHCl str	974.16	971.91	817.55	814.93	971.66	968.85	0.15	0.80
6	HCH bnd	1229.82	1221.33	1167.96	1160.91	1172.00	1163.06	-2.58	0.41
7	HCH bnd	1447.25	1442.99	1446.97	1442.64	1222.42	1217.89	0.11	-0.09
8	HCH bnd	1447.25	1442.99	1446.97	1442.64	1434.51	1430.68	0.11	-0.78
9*	CH sym. str	3127.36	3126.40	3127.13	3126.19	3194.70	3188.20	-0.03	11.65
10	CH asym str	3299.24	3286.07	3299.17	3285.99	2367.08	2355.66	0.01	-5.07
11	CH asym str	3299.24	3286.07	3299.17	3285.99	3299.13	3285.93	0.01	0.05
Tunnel								0.55	-0.24
ZPE/									
Total		8517.87	8489.05	8272.53	8244.29	7867.05	7836.20	-3.42	5.79

* Frequencies 9 and 10 were swapped for the Cl-H-¹²CH₂D and Cl-H-¹³CH₂D to preserve the C-H symmetric stretch as frequency 9

1033 **Table 5:** Relative rates for OH oxidation of different isotopologues of CH₄ by different
 1034 pathways (H-abstraction versus D-abstraction).

Reaction	Relative Rate (298 K)
$^{12}\text{CH}_4 + \text{OH} \rightarrow ^{12}\text{CH}_3 + \text{H}_2\text{O}$	1.000
$^{13}\text{CH}_4 + \text{OH} \rightarrow ^{13}\text{CH}_3 + \text{H}_2\text{O}$	0.994
D/H primary isotope effects (D-abstraction)	
$^{12}\text{CH}_3\text{D} + \text{OH} \rightarrow ^{12}\text{CH}_3 + \text{HDO}$	0.026
$^{13}\text{CH}_3\text{D} + \text{OH} \rightarrow ^{13}\text{CH}_3 + \text{HDO}$	0.026
D/H secondary isotope effects (H-abstraction)	
$^{12}\text{CH}_3\text{D} + \text{OH} \rightarrow ^{12}\text{CH}_2\text{D} + \text{H}_2\text{O}$	0.731
$^{13}\text{CH}_3\text{D} + \text{OH} \rightarrow ^{13}\text{CH}_2\text{D} + \text{H}_2\text{O}$	0.727
Total H or D abstraction fractionation factor	
$^{12}\text{CH}_3\text{D} + \text{OH} \rightarrow \text{products}$	0.757
$^{13}\text{CH}_3\text{D} + \text{OH} \rightarrow \text{products}$	0.752

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Table 6: Comparison between calculated Kinetic Isotope Effects for the CH₄+OH reaction and CH₄+Cl reaction with the measured Kinetic Isotope Effects from the present study.

Parameter	Calculated (TST)	Measured
¹³ KIE _{OH}	1.0060	1.0061 ± 0.0012
² KIE _{OH}	1.3208	1.3344 ± 0.0182
^{13,2} KIE _{OH}	1.3292	1.3429 ± 0.0192
^{2,2} KIE _{OH}	1.9066	
^{13,2} γ _{OH}	0.9997	0.9997 ± 0.0012
^{2,2} γ _{OH}	0.9266	
¹³ KIE _{Cl}	1.0280	*1.0621 ± 0.0001
² KIE _{Cl}	1.410	*1.482 ± 0.009
^{13,2} KIE _{Cl}	1.450	*1.579 ± 0.011
^{2,2} KIE _{Cl}	2.184	
^{13,2} γ _{Cl}	0.9996	*0.9965 ± 0.0007
^{2,2} γ _{Cl}	0.9633	

* Measured values for kinetic isotope effects for the CH₄+Cl reactions assume a ¹³KIE_{Cl} value of 1.0621 ± 0.0001 (Tyler et al., 2000) and the relationships observed in our experiments.

1044 **Table 7:** Normal mode vibrational frequencies for $^{12}\text{CH}_2\text{D}_2$ and the $^{12}\text{CH}_2\text{D}_2+(\text{OH},\text{Cl})$
 1045 transition states. Given vibrational frequencies for transition states are an average of
 1046 the vibrational frequencies for the 3 different possible configurations.

Mode	$^{12}\text{CH}_2\text{D}_2$	HO-H- $^{12}\text{CHD}_2$	HO-D- $^{12}\text{CH}_2\text{D}$	Cl-H- $^{12}\text{CHD}_2$	Cl-D- $^{12}\text{CH}_2\text{D}$
Imfreq (<i>i</i>)		-1712.06	-1307.65	-1134.19	-871.89
1	1058.51	6.74	6.84	288.33	274.96
2	1120.10	263.80	249.67	328.02	309.96
3	1269.94	294.35	278.65	480.14	486.12
4	1372.62	745.50	751.30	894.69	729.50
5	1481.88	909.26	891.04	924.34	815.60
6	2268.64	1045.88	969.45	1052.53	1108.04
7	2377.23	1086.52	1092.39	1136.54	1221.26
8	3149.04	1217.04	1204.62	1314.42	1433.89
9	3210.33	1320.84	1276.61	2291.06	2366.87
10		1394.92	1450.13	2453.95	3194.55
11		2281.37	2343.74	3250.50	3299.06
12		2413.17	3166.88		
13		3211.94	3251.89		
14		3807.52	3807.46		
ZPE	8654.15	9999.42	10370.33	7207.26	7619.91

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1049 **Table 8:** Input parameters for model of $\Delta^{13}\text{CH}_3\text{D}$ of atmospheric methane. All numbers
 1050 are from Kai et al. (2011), except for the $\Delta^{13}\text{CH}_3\text{D}$ of different sources, which are
 1051 estimated based on Wang et al. (2015).

Source	Source Input (Tg yr ⁻¹)	$\delta^{13}\text{C}$ (‰)	δD (‰)	$\Delta^{13}\text{CH}_3\text{D}$ (‰)	% from NH	% from SH
Tundra	10	-65	-340	3.0	98	2
Lakes (bogs, fens)	40	-60	-320	3.0	85	15
Swamps and Marshes	100	-60	-320	0.7	40	60
Agriculture (rice, livestock)	199	-69	-310	1.0	84	16
Landfills	44	-52	-290	3.0	86	14
Fossil Fuels	76	-38	-160	3.0	87	13
Biomass Burning	35	-23	-225	3.0	53	47
Termites	20	-58	-345	1.0	55	45
Gas Hydrates	10	-52	-205	6.0	98	2
Total	534	-57	-284	2.8	74	26

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9. FIGURES

Figure 1: Results from OH oxidation experiments, showing $\ln(R/R_{\text{initial}})$ versus $\ln(f)$ for different isotopologues ($^{12}\text{CH}_3\text{D}$, $^{13}\text{CH}_4$, $^{13}\text{CH}_3\text{D}$). The slope of the regression line is $\alpha - 1$, where $\alpha = 1/\text{KIE}$.

Figure 2: Results from Cl oxidation experiments, showing $\ln(R/R_{\text{initial}})$ versus $\ln(^{13}R/^{13}R_{\text{initial}})$ for $^{12}\text{CH}_3\text{D}$ and $^{13}\text{CH}_3\text{D}$. The slope of the regression line is $(^x\alpha - 1)/(^{13}\alpha - 1)$, where $\alpha = 1/\text{KIE}$ and $x=^2$ or 13,2 .

Figure 3: Illustration of the effect of different γ values on the $\Delta^{13}\text{CH}_3\text{D}$ for the OH and Cl oxidation experiments, using the kinetic isotope effects discussed in the results section.

1069 **Figure 4:** Calculated transition state geometries for HO-H-CH₃ and Cl-H-CH₃.

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Figure 5: Illustration of the effect of different γ values on the $\Delta^{13}\text{CH}_3\text{D}$ for the OH and Cl oxidation experiments, using the kinetic isotope effects discussed in the results section.

1076 **Figure 6:** Effect of different factors on the calculated $^{13,2}\gamma$ value, as a function of
1077 temperature.

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Figure 7: Effect of isotopic substitution on the C-H symmetric stretching motion of CH₄. In the ^xCH₄ (T_d) molecule, carbon is at the center of mass and does not move during the vibration. As a result, carbon isotopic substitution does not affect the energy of the vibration. In the ^xCH₃D (C_{3v}) molecule, however, the center of mass is shifted away from the carbon atom, so the carbon atom moves during the vibration. Because of this, the energy of the vibration is lower with carbon isotopic substitution. This is the dominant source of the clumped effect for methane

10. Appendix

10.1 Derivation of Open System Isotope Effect

To illustrate the effects of an open system at steady state on $\Delta^{13}\text{CH}_3\text{D}$, we will consider a simple box model with various sources of methane and a single sink reaction. A scenario where this would be useful would be obtaining a first-order estimate of the $\Delta^{13}\text{CH}_3\text{D}$ of atmospheric methane. To follow this scenario, we will consider OH oxidation to be the only sink of methane. In reality, OH oxidation accounts for almost 90% of the tropospheric methane sink (with soil uptake and stratospheric exchange being minor sinks), so this is a good first approximation for a simple atmospheric scenario. The change in $^{12}\text{CH}_4$ over time can be written:

$$\frac{d[^{12}\text{CH}_4]}{dt} = \sum_i F_i - k[\text{OH}][^{12}\text{CH}_4]$$

Where the summation i is over all the sources of methane, F_i is the volume-weighted flux ($\text{molecules cm}^{-3} \text{ s}^{-1}$) of $^{12}\text{CH}_4$ into the box (i.e. atmosphere) from source i , and k is the rate constant for the $^{12}\text{CH}_4 + \text{OH}$ reaction. The concentration and fluxes of $^{12}\text{CH}_4$ are approximately equal to the concentration and fluxes of total methane, as $^{12}\text{CH}_4$ accounts for over 98% of all methane at natural abundances.

Assuming that the concentration of methane is at steady state, we get:

$$\frac{d[^{12}\text{CH}_4]}{dt} = 0$$

And therefore:

$$[^{12}\text{CH}_4] = \frac{\sum_i F_i}{k[\text{OH}]}$$

Let $F = \sum_i F_i$ be the total source flux of methane to the atmosphere, then the fractional contribution from each source can be denoted as $f_i = F_i/F$.

1109 The mass balance for the isotopes can be written as:

$$\frac{d[^{13}\text{CH}_4]}{dt} = \sum_i {}^{13}R_i * F_i - {}^{13}\alpha \cdot k \cdot [\text{OH}][^{13}\text{CH}_4]$$

$$\frac{d[^{12}\text{CH}_3\text{D}]}{dt} = \sum_i {}^{12}R_i * F_i - {}^{12}\alpha \cdot k \cdot [\text{OH}][^{12}\text{CH}_3\text{D}]$$

$$\frac{d[^{13}\text{CH}_3\text{D}]}{dt} = \sum_i {}^{13,2}R_i * F_i - {}^{13,2}\alpha \cdot k \cdot [\text{OH}][^{13}\text{CH}_3\text{D}]$$

1110 In these equations, ${}^{13}R_i$ is the $^{13}\text{CH}_4/^{12}\text{CH}_4$ ratio of the source i , ${}^{12}R_i$ is the $^{12}\text{CH}_3\text{D}/^{12}\text{CH}_4$
 1111 ratio of source i , and ${}^{13,2}R_i$ is the $^{13}\text{CH}_3\text{D}/^{12}\text{CH}_4$ ratio of the source i . Similarly, ${}^{13}\alpha$, ${}^{12}\alpha$,
 1112 and ${}^{13,2}\alpha$ are the kinetic isotope effects (k/k^*) for $^{13}\text{CH}_4$, $^{12}\text{CH}_3\text{D}$, and $^{13}\text{CH}_3\text{D}$,
 1113 respectively.

1114 Assuming that the isotopic composition of methane is also at steady state, we
 1115 get:

$$\frac{d[^{13}\text{CH}_4]}{dt} = 0$$

$$\frac{d[^{12}\text{CH}_3\text{D}]}{dt} = 0$$

$$\frac{d[^{13}\text{CH}_3\text{D}]}{dt} = 0$$

1116 And therefore:

$$[^{13}\text{CH}_4] = \frac{\sum_i {}^{13}R_i F_i}{{}^{13}\alpha \cdot k \cdot [\text{OH}]}$$

$$[^{12}\text{CH}_3\text{D}] = \frac{\sum_i {}^{12}R_i F_i}{{}^{12}\alpha \cdot k \cdot [\text{OH}]}$$

$$[^{13}\text{CH}_3\text{D}] = \frac{\sum_i {}^{13,2}R_i F_i}{{}^{13,2}\alpha \cdot k \cdot [\text{OH}]}$$

1117 Because we measure isotope ratios rather than absolute abundances, it is useful to look
 1118 at the $^{13}\text{CH}_4/^{12}\text{CH}_4$ ratio. Given the steady state approximations, this gives us:

$$\frac{[^{13}\text{CH}_4]}{[^{12}\text{CH}_4]} = \frac{\sum_i ^{13}R_i f_i}{^{13}\alpha}$$

$$\frac{[^{12}\text{CH}_3\text{D}]}{[^{12}\text{CH}_4]} = \frac{\sum_i ^2R_i f_i}{^2\alpha}$$

$$\frac{[^{13}\text{CH}_3\text{D}]}{[^{12}\text{CH}_4]} = \frac{\sum_i ^{13,2}R_i f_i}{^{13,2}\alpha}$$

1119 Now consider the value of $\Delta^{13}\text{CH}_3\text{D}$:

$$\Delta^{13}\text{CH}_3\text{D} = \ln \left(\frac{\frac{[^{13}\text{CH}_3\text{D}]}{[^{12}\text{CH}_4]}}{\frac{[^{13}\text{CH}_4]}{[^{12}\text{CH}_4]} * \frac{[^{12}\text{CH}_3\text{D}]}{[^{12}\text{CH}_4]}} \right) = \ln \left(\frac{(\sum_i ^{13,2}R_i f_i)}{(\sum_i ^{13}R_i f_i)(\sum_i ^2R_i f_i)} * \frac{^{13}\alpha * ^2\alpha}{^{13,2}\alpha} \right)$$

1120 Now if we let:

$$^{13,2}\gamma = \frac{^{13,2}\alpha}{^{13}\alpha * ^2\alpha}$$

1121 Then we can simplify to:

$$\Delta^{13}\text{CH}_3\text{D} = \ln \left(\frac{(\sum_i ^{13,2}R_i f_i)}{^{13,2}\gamma (\sum_i ^{13}R_i f_i)(\sum_i ^2R_i f_i)} \right)$$

1122 This means that, if $^{13,2}\gamma = 1$, then the clumped isotopic signature of methane in the box
 1123 (i.e. $\Delta^{13}\text{CH}_3\text{D}$) is independent of the isotope effects during the oxidation of methane and
 1124 depends only on the isotopic composition (i.e. $\delta^{13}\text{CH}_4$, $\delta^{12}\text{CH}_3\text{D}$, $\delta^{13}\text{CH}_3\text{D}$) of the
 1125 methane sources scaled by their fractional contribution to the total source. In the case
 1126 that $^{13,2}\gamma \neq 1$, the $^{13,2}\gamma$ factor contributes approximately a constant offset of $-\ln(^{13,2}\gamma)$. If
 1127 we assume that $^{13,2}\gamma_{\text{OH}} = 0.9997 \pm 0.0012$, then the $^{13,2}\gamma$ factor means that the steady

1128 state $\Delta^{13}\text{CH}_3\text{D}$ value would be $0.3\text{‰} \pm 1.2\text{‰}$ higher than predicted purely based on the
1129 source signatures.

1130 Let us simplify the scenario further and consider only a single source of methane.

1131 In this case:

$$\Delta^{13}\text{CH}_3\text{D} = \ln\left(\frac{{}^{13,2}R}{{}^{13,2}\gamma * {}^{13}R * {}^2R}\right)$$

1132 Now, since $\ln({}^{13,2}R/{}^{13}R * {}^2R) = \Delta^{13}\text{CH}_3\text{D}_{\text{SOURCE}}$, we can simplify this to:

$$\Delta^{13}\text{CH}_3\text{D} = (\Delta^{13}\text{CH}_3\text{D})_{\text{SOURCE}} - \ln({}^{13,2}\gamma)$$

1133 So the clumped isotopic composition of a methane reservoir at steady state with a
1134 single source and a single sink is equal to the $\Delta^{13}\text{CH}_3\text{D}$ value of the source, plus an
1135 offset due to the inherent clumping during the sink reaction. For ${}^{13,2}\gamma < 1$, as expected
1136 for most normal kinetic reactions, $\ln({}^{13,2}\gamma)$ will be negative, so the resulting $\Delta^{13}\text{CH}_3\text{D}$ of
1137 the reservoir will be higher (i.e. more clumped) than that of the source. Although this
1138 effect is small for OH oxidation of CH_4 , ($0.3\text{‰} \pm 1.2\text{‰}$), it can be significant for cases
1139 like Cl oxidation of CH_4 ($3.5\text{‰} \pm 0.7\text{‰}$).

1140 10.2 Derivation of Closed System Kinetic Isotope Effect

1141 In this section, we will discuss the effect of an irreversible reaction (such as OH
1142 or Cl oxidation) on the $\Delta^{13}\text{CH}_3\text{D}$ value of methane in a closed system, such as our
1143 photochemical reaction cell. Assume the case where CH_4 of a known isotopic
1144 composition is initially added to the system. Let ${}^2\bar{\delta}_0$, ${}^{13}\bar{\delta}_0$, and ${}^{13,2}\bar{\delta}_0$ be the isotope ratios
1145 of the initial ${}^{12}\text{CH}_3\text{D}$, ${}^{13}\text{CH}_4$, ${}^{13}\text{CH}_3\text{D}$, all relative to ${}^{12}\text{CH}_4$, on a V-PDB (for carbon) / V-
1146 SMOW (for hydrogen) hybrid scale.

1147 We are interested in the “clumped” isotopic signature of methane ($\Delta^{13}\text{CH}_3\text{D}$) in
1148 the gas as a function of the fraction of initial gas remaining, f . Using traditional Rayleigh

1149 fractionation equations (Johnson et al., 2002), we can calculate the isotopic signature
 1150 for each minor isotopologue in the system. This gives us:

$$(^2\delta + 1) = (^2\delta_0 + 1)f^{2\varepsilon}$$

$$(^{13}\delta + 1) = (^{13}\delta_0 + 1)f^{13\varepsilon}$$

$$(^{13,2}\delta + 1) = (^{13,2}\delta_0 + 1)f^{13,2\varepsilon}$$

1151 Here, $^x\varepsilon = ^x\alpha - 1$ is the isotopic fractionation (see Coplen, 2011).

1152

1153 Note that these are equations to calculate the δ value of the remaining methane in the
 1154 system (i.e. remaining reactant), not the product. The clumped isotopic signature of the
 1155 residual methane can be written as:

$$\Delta^{13}\text{CH}_3\text{D} = \ln\left(\frac{(^{13,2}\delta + 1)}{(^{13}\delta + 1) * (^2\delta + 1)}\right) = \ln\left(\frac{(^{13,2}\delta_0 + 1)}{(^{13}\delta_0 + 1) * (^2\delta_0 + 1)} * \frac{f^{13,2\varepsilon}}{f^{13\varepsilon} * f^{2\varepsilon}}\right)$$

$$\Delta^{13}\text{CH}_3\text{D} = \ln\left(\frac{(^{13,2}\delta_0 + 1)}{(^{13}\delta_0 + 1) * (^2\delta_0 + 1)}\right) - \ln\left(\frac{f^{13,2\varepsilon}}{f^{13\varepsilon} * f^{2\varepsilon}}\right)$$

1156 This gives

$$\Delta^{13}\text{CH}_3\text{D} \approx \Delta^{13}\text{CH}_3\text{D}_0 + \ln(f) * (^{13,2}\varepsilon - ^{13}\varepsilon - ^2\varepsilon)$$

1157 Since,

$$^{13,2}\varepsilon - ^{13}\varepsilon - ^2\varepsilon = (^{13,2}\alpha - 1) - (^{13}\alpha - 1) - (^2\alpha - 1)$$

$$^{13,2}\varepsilon - ^{13}\varepsilon - ^2\varepsilon = ^{13,2}\gamma \cdot ^{13}\alpha \cdot ^2\alpha - ^{13}\alpha - ^2\alpha + 1$$

1158 Which can be factored and simplified to:

$$^{13,2}\varepsilon - ^{13}\varepsilon - ^2\varepsilon = ^{13}\varepsilon * ^2\varepsilon + (^{13,2}\gamma - 1) \cdot ^{13}\alpha \cdot ^2\alpha$$

1159

1160 This gives us:

$$\Delta^{13}\text{CH}_3\text{D} = \Delta^{13}\text{CH}_3\text{D}_0 + ({}^{13}\varepsilon \cdot {}^2\varepsilon) * \ln(f) + ({}^{13,2}\gamma - 1) \cdot {}^{13}\alpha \cdot {}^2\alpha \cdot \ln(f)$$