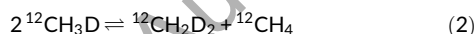


LETTER TO THE EDITOR

Method for calibrating methane clumped isotope measurements via catalytic equilibration of methane isotopologues on γ -alumina

Measurements of the precise abundances of $^{13}\text{CH}_3\text{D}$ and $^{12}\text{CH}_2\text{D}_2$ – methane isotopologues carrying two simultaneous substitutions of rare isotopes – were first reported five years ago.¹ Since then, the volume of data on these species has grown enormously. At least 23 papers have reported data on one or both of these isotopologues (supporting information). Measurements have been made using two isotope ratio mass spectrometers – the ThermoFinnigan MAT 253 Ultra² and the Nu Instruments Panorama³ – and one infrared spectrometer – the Aerodyne Research laser trace gas monitor.⁴ Methods were developed to determine apparent equilibrium temperatures stored in populations of methane molecules collected from the environment, a technique known as clumped isotope thermometry (see Eiler^{5,6} for a history of the field).

Clumped isotope thermometry of methane is based on the natural tendency for ^{13}C and/or one or more deuterium (D) atoms to co-occur (or “clump”) in the same molecule at abundances slightly greater than random (or “stochastic”). The deviation from being stochastic is greater at lower temperatures. Statistical mechanical calculations yield predictions of the equilibrium constants (K_{eq} s) of the reactions:



Relationships describing the theoretical dependence of the K_{eq} s on temperature have been presented by a number of workers (supporting information). Modeled values of K_{eq} for $^{13}\text{CH}_3\text{D}$ (Equation (1)) generally agree within 0.3‰ at geologically relevant temperatures.

The apparent equilibrium temperatures of samples are determined by comparison of the measured reaction quotients (Q) of Equations (1) and (2) with their equilibrium constants calculated for different temperatures (T). In isotope geochemistry, the degree to which a sample is clumped is described using a capital-delta symbol (Δ -notation):

$$\Delta = \frac{Q}{K_{\infty}} - 1 \quad (3)$$

where K_{∞} is a statistical factor that corresponds to a hypothetical sample in which isotopes are distributed randomly across all possible methane species: K_{∞} is 1 for Equation (1), and $6/16$ for Equation (2). It is equivalent to the value taken by K_{eq} at infinite temperature.

With a number of laboratories gearing up to make methane clumping measurements, this may be an opportune time (i) to propose a generally accepted method of generating equilibrated methane samples for the purposes of temperature calibration; and (ii) to perform round-robins (interlaboratory comparisons) of methane isotopic measurements (including δD values and the clumped parameters) to address differences of scaling, linearity, and anchoring between labs and instruments. This contribution tackles the first of these problems by describing a simple method for equilibrating methane isotopologues at low temperature. The latter effort is under way and will be documented elsewhere.

Many different catalysts have been shown to promote D/H exchange amongst methane isotopologues (see Sattler⁷ for a detailed review of the history of catalyst-assisted hydrogen exchange in alkanes). Previous studies in which methane clumped isotopes were equilibrated used borosilicate (Pyrex) or quartz “break-seal” tubes loaded with a silica/alumina-supported nickel (Ni) or alumina-supported platinum (Pt) catalyst and an adsorbent (silica gel or zeolite molecular sieve) that were evacuated under heat, charged with several milliliters SATP of methane, sealed off with a natural gas torch, and then heated at temperatures $\geq 150^\circ\text{C}$ for durations ranging from days to weeks.^{1,4,8} This process rearranges the carbon-hydrogen (C–H) bonds in CH_4 and redistributes hydrogen isotopes (H and D) amongst the molecules of CH_4 according to their thermodynamic tendencies. Methane isotopologue measurements on all three instruments were calibrated against suites of CH_4 samples treated in this way.

We wish to share a different method – tested in our laboratories – that uses a catalyst capable of exchanging the C–H bonds of methane under near-ambient conditions. It avoids the hazards associated with open flames and unshielded furnaces. The catalyst that we use is a form of alumina, $\gamma\text{-Al}_2\text{O}_3$, prepared from thermal treatment of boehmite ($\gamma\text{-AlO}(\text{OH})$). Gamma-alumina has exceptional catalytic ability for activating carbon-hydrogen bonds in hydrocarbons across a wide range of temperatures.^{9–13}

A temperature-controlled reaction vessel was built to conduct equilibration experiments (supporting information). It consists of an

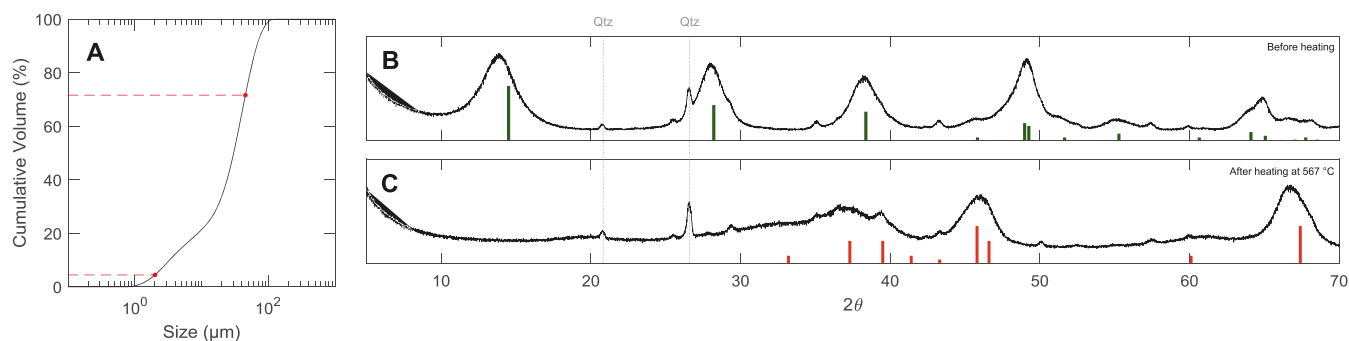


FIGURE 1 Characterization of catalyst powder. A, Particle size analysis, determined with a laser particle size analyzer (Mastersizer 3000; Malvern Panalytical, Malvern, UK), average of eight analyses. Results indicate that particles $<2\ \mu\text{m}$ comprise 4.5% of the catalyst volume, and particles $<45\ \mu\text{m}$ are 72% of the volume (red lines). Powder X-ray diffraction spectrum of PURAL® SB catalyst: B, before and C, after heating at 567°C for 18 h under flowing helium. The before spectrum is consistent with reported boehmite spectra (green sticks), and the after spectrum is consistent with the spectrum of γ -alumina (orange sticks).^{18,19} Dotted lines represent peak positions of quartz (from quartz wool). Spectra were recorded with a D5000 diffractometer (Bruker, Karlsruhe, Germany) [Color figure can be viewed at wileyonlinelibrary.com]

approximately 20-inch length of $\frac{1}{2}$ " (o.d.) stainless steel tubing flanked by two high-pressure, high-temperature valves (rated to 118 bar @ 648°C). Catalyst powder (described below) was packed into steel sleeves using pre-combusted quartz wool as a filler. The catalyst sleeves were then inserted into the steel reactor tubing. Ceramic spacers were used to keep the catalyst sleeves centered. The reaction vessel was placed inside a tube furnace (Lindberg 55070-S; Lindberg, a unit of General Signal, Watertown, WI, USA), and the ends of the tube furnace were plugged with kaolin wool. The temperature was monitored by several K-type thermocouples (Omega Engineering, Norwalk, CT, USA) attached to the outside of the stainless-steel tube and embedded in the furnace insulation. The furnace temperature was controlled via a single-loop PID controller. The measurement error is estimated to be $\pm 2^\circ\text{C}$ across the temperature range studied.

Samples of boehmite (PURAL® SB) were obtained from Sasol. PURAL™ is a high-purity alumina hydrate produced in Sasol's Brunsbüttel, Germany facility (technical data, Sasol company website). PURAL SB is a loose white powder with an average particle size of $\sim 30\ \mu\text{m}$ (Figure 1A). A minor but not insignificant fraction of the particles is less than $2\ \mu\text{m}$ in size; hence, an appropriate mesh frit was installed on the ends of the reactor tube to protect vacuum components from airborne particulate matter.

Activation of the catalyst involves converting the boehmite into γ -alumina. This can be accomplished by baking the catalyst overnight (18 h) at 567°C either under dynamic vacuum ($<10\ \text{mTorr}$) or under a helium stream flowing at $>30\ \text{scm}^3/\text{min}$. This temperature was previously determined by John et al to be optimal for γ -alumina activation.¹² We verified that the phase transition from boehmite to γ -alumina was complete by using powder X-ray diffraction (XRD) spectroscopy (Figures 1B and 1C).^{*} We find that catalyst activity is sensitive to pretreatment temperature. Catalyst treated at 700°C had

only one-third the activity of catalyst treated at 567°C for D/H exchange (data not shown).

Once conditioned, the catalyst was stored under overpressure ($>1.3\ \text{bar}$ absolute) of helium to prevent leakage of air and water vapor into the reaction chamber. The catalyst need not be reconditioned unless exposed to significant amounts of air or other poisons. We estimate the number of active sites on the γ -alumina catalyst to be $17\ \mu\text{mol}$ sites per gram, which is close to that measured by previous studies.⁹ This was accomplished by treating the catalyst with excess deuterium (D_2) and then back-titrating with normal CH_4 to measure the quantity of CH_3D produced.

A sample of methane enriched in $^{13}\text{CH}_3\text{D}$ was obtained from Prof. Shuhei Ono at MIT (Cambridge, MA, USA). This sample has a $\Delta^{13}\text{CH}_3\text{D}$ value of approximately +4000‰ (relative to stochastic) as measured at MIT (S. Ono, personal communication). We blended this sample 1:100 with a commercially sourced pure methane gas ("RG", 99.999%; Air Liquide, Radnor, PA, USA) used as our in-house working gas. The blended mixture has a $\Delta^{13}\text{CH}_3\text{D}$ value of approximately +41‰ relative to RG (supporting information). This mixture was the starting gas in our experiments.

To begin an experiment, the reactor was first brought to the target temperature (25 or 100°C) and evacuated. The starting gas was then introduced at a pressure of 0.7–1.5 bar absolute. Samples were withdrawn at designated times and frozen using liquid nitrogen (-196°C) into custom 10-cm^3 stainless-steel cylinders (Hoke, Spartanburg, SC, USA; nos. 2HSY10 and 7155F2Y) containing $\sim 0.5\ \text{g}$ silica gel (Sigma-Aldrich, St Louis, MO, USA; catalog no. 214426). Prior to use, cylinders were heated with a heat gun (Master, Racine, WI, USA) for several minutes under dynamic vacuum to remove adsorbed water, CO_2 , and other contaminants.

Samples were analyzed using a tunable infrared laser direct-absorption spectrometer (TILDAS; Aerodyne Research Inc., Billerica, MA, USA) following the method of Ono et al.⁴ In brief, a sample of CH_4 is compared with a reference sample of pure methane (here RG) in alternating fashion. The relative abundances of $^{12}\text{CH}_4$, $^{13}\text{CH}_4$, $^{12}\text{CH}_3\text{D}$, and $^{13}\text{CH}_3\text{D}$ are quantified using a spectroscopic model

^{*}It is important to determine the XRD pattern to verify the purity of the γ -alumina, as other phases have different catalytic properties. For example, alumina containing aluminum oxide chloride hydroxide [$\text{Al}_{45}\text{O}_{45}(\text{OH})_{45}\text{Cl}$] was found to be inactive under the same experimental conditions, and θ -alumina was significantly less active for D/H exchange catalysis.

(Voigt profile) fit to the absorption data. Pressure is controlled by a custom-designed dual-bellows gas-inlet system. This instrument performs similarly to the MIT instrument as described in Ono et al.⁴ The one-second Allan deviation is ~1.0‰ for the ratio $^{13}\text{CH}_3\text{D}/^{12}\text{CH}_4$.

Tabulated isotopologue-abundance data on the experimental samples can be found in the supporting information. The values of $\delta^{13}\text{C}$ and δD remained relatively constant during both 25 and 100°C experiments, and the values of $\Delta^{13}\text{CH}_3\text{D}$ decreased with time. The rate of change slowed asymptotically in both experiments.

The time-series experiments can be modeled by the following rate law:

$$\Delta_t = F \cdot \Delta_0 + (1 - F) \cdot \Delta_{\text{eq}}, \text{ where } F = e^{-kt} \quad (4)$$

Here, t is the time since the beginning of the experiment, and k is the pseudo-first-order decay constant for a given quantity of methane over a given quantity of catalyst [dimensions of $(\text{time})^{-1}$]. The decay constant is related to the half-exchange timescale (HET) by the relation:

$$k = \ln 2 / \text{HET} \quad (5)$$

The HET is the time over which clumped isotope values have moved halfway towards their values at equilibrium. Curves fitted to the experimental data are shown in Figure 2.

The decay constant k is a function of temperature and the quantities of CH_4 and catalyst. We isolate the effect of temperature by defining the methane- and catalyst-independent rate constant k° as follows:

$$k^\circ = k \cdot n_{\text{methane}} / m_{\text{catalyst}} \quad (6)$$

Here, k° has dimensions of $(\text{quantity } \text{CH}_4) (\text{time})^{-1} (\text{quantity catalyst})^{-1}$, n_{methane} is the number of moles of methane present, and m_{catalyst} is the mass of the catalyst (in grams). Fitted kinetic constants for each experiment are listed in Table 1.

Extrapolation of measured k° values to higher and lower temperatures yields the curve shown in Figure 3A. The extrapolation suggests that experiments conducted at temperatures of $>200^\circ\text{C}$ under the conditions that we tested (~ 2 g catalyst, ~ 1 mmol CH_4 , activation at 567°C) will fully equilibrate (>5 half-exchange times) in less than 15 min. It also suggests that experiments conducted at temperatures as low as 0°C may fully equilibrate in approximately 1 week. The reaction rate can be scaled up by increasing the amount of catalyst loaded. The apparent activation energy of 28 kJ/mol is similar to the values of 24 kJ/mol and 31 kJ/mol determined by previous workers using CH_4/CD_4 mixtures.^{9,13}

Figure 3B depicts the best fit of the experimental asymptotes to the theoretical curve from Young et al.⁸ The curve has been fitted to the data by sliding it in the vertical direction. This yields a 2.82‰ difference between RG and stochastic reference frames and implies a 172°C temperature for RG. Figure 3C shows the same data plotted in the stochastic reference frame. Also shown are previously-published data for samples of CH_4 driven to equilibrium over supported metal catalysts.

In most labs, samples of CH_4 with varying δD values are generated routinely and analyzed to derive correction factors for the instrument. In generating these heated gas lines, previous investigators have noted that the δD value can change significantly (by $>100\%$) after heating.^{1,4} Such changes have been attributed to hydrogen exchange of CH_4 with hydroxyl groups on glass tube surfaces or methyl groups on catalyst surfaces. Bulk isotopic shifts do not affect the calibration as long as the isotopologues have reached equilibrium. They are, however, an annoyance as they introduce an element of unpredictability in the δD values comprising the points on the heated gas line. The method that we use does not appear to

FIGURE 2 Demonstration of successful low-temperature equilibration of CH_4 isotopologues at natural abundance levels, starting with a sample enriched by 41‰ in $^{13}\text{CH}_3\text{D}$. Experiments conducted at A, 100°C and B, 25°C . Error bars (95% confidence interval) are smaller than the symbols. Data expressed relative to reference gas (RG). Curves represent minimizations of the sum of squared residuals to Equation (4). The red and green curves asymptote at +0.92‰ and +3.15‰, respectively [Color figure can be viewed at wileyonlinelibrary.com]

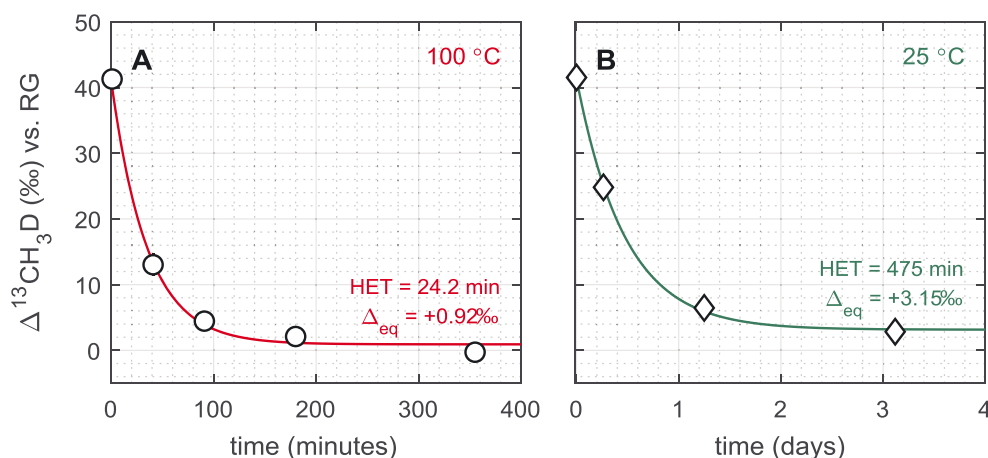


TABLE 1 Exchange kinetics determined under different conditions

T (°C)	$\gamma\text{-Al}_2\text{O}_3$ (g)	CH_4 (mmol)	HET (min)	k° ($\text{mmol min}^{-1} \text{g}^{-1}$)
25	2.1	1.08	475	7.5×10^{-4}
100	2.1	0.52	24.2	7.2×10^{-3}

HET, half-exchange time.

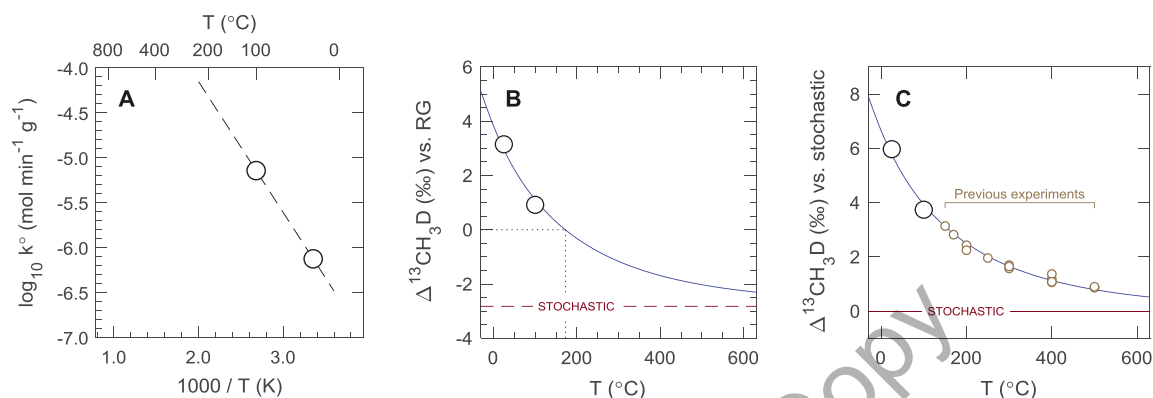


FIGURE 3 A, Temperature-dependence of the equilibration rate k° . The slope of this Arrhenius plot implies an activation energy of 28 kJ/mol. B, Experimental results compared with theoretical prediction of temperature-dependence of $\Delta^{13}\text{CH}_3\text{D}$ at equilibrium (see text). The y-axis is equivalent to $\Delta_{\text{sam vs. sto}} - \Delta_{\text{RG vs. sto}}$. C, Experimental results from this study (large circles), translated into the stochastic reference frame, compared against catalytically-equilibrated methane gases from previous studies^{1,4,8,20} (small circles). A linear function²¹ was used to convert Δ_{18} data reported in Stolper et al.¹ into their implied equilibrated $\Delta^{13}\text{CH}_3\text{D}$ values [Color figure can be viewed at wileyonlinelibrary.com]

induce any obvious shifts in $\delta^{13}\text{C}$ or δD values. In addition, the deuterium content of the CH_4 can be readily adjusted. Deuteriomethane is available commercially. Deuterium-depleted methane can be synthesized from aluminum carbide and light water⁴ or by using methanogen cultures.¹⁴ Alternatively, normal CH_4 can be equilibrated with deuterium-depleted H_2 created through electrolysis of light water¹⁵ or equilibration of light water with normal H_2 over hydrophobic catalyst.^{16,17}

Prior studies indicate that the method described above can equilibrate $^{12}\text{CH}_2\text{D}_2$ in methane on timescales similar to those observed here.⁹ Exchange of deuterium with other small alkanes has also been tested.¹⁰ Hence, γ -alumina could enable temperature calibration of several other clumped- or position-specific isotope geothermometers. The catalyst material is stable and active for exchange at temperatures at least as high as 400°C.¹³

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of this article.

Supporting Information for:

Method for calibrating methane clumped isotope measurements via catalytic equilibration of methane isotopologues on γ -alumina

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The following papers reported the abundance of $^{13}\text{CH}_3\text{D}$ and $^{12}\text{CH}_2\text{D}_2$ in natural settings or in experiments conducted without severe enrichment in deuterium.

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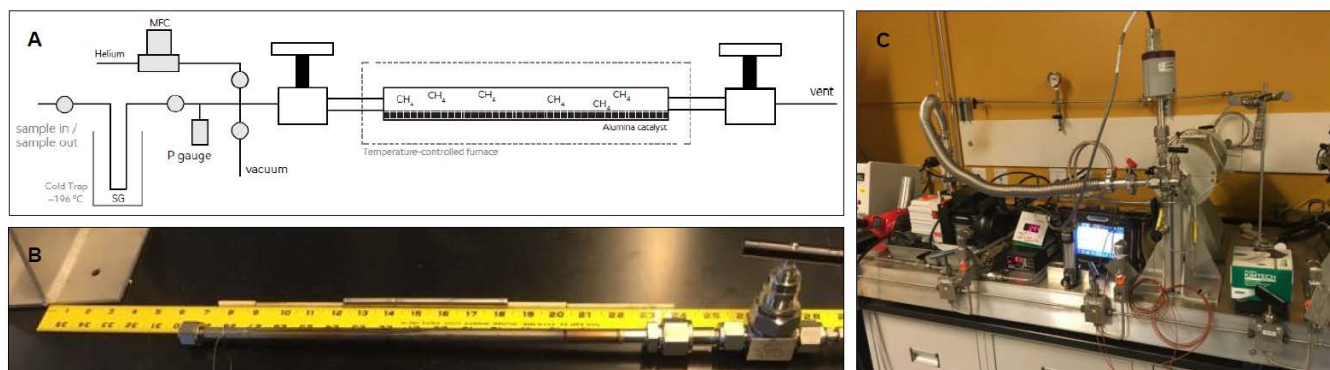
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Supporting Fig. 1. Experimental apparatus for equilibration of CH_4 gases over catalyst at URC. **(A)** Cartoon of the design. **(B)** Photo of contents and layout of inner sleeves containing packed catalyst. White cylindrical objects are ceramic sticks used as spacers. **(C)** Photo of experimental setup.

Table S1

Carbon and hydrogen isotope ratios and clumped isotopologue abundances of methane in equilibration experiments.

	TP	Time (min)	Isotope/Isotopologue Ratios vs RG (‰)		
			$\delta^{13}\text{C}$ values	δD values	$\Delta^{13}\text{CH}_3\text{D}$
25 °C	0	0	-1.31	0.07	41.52 ± 0.89
	1	380	-1.24	0.56	24.81 ± 0.74
	2	1800	-1.15	0.51	6.46 ± 1.50
	3	4490	-1.21	0.43	2.84 ± 0.60
100 °C	0	0	-1.06	-0.12	41.29 ± 0.86
	1	41	-0.96	0.81	13.03 ± 1.70
	2	91	-0.61	0.55	4.45 ± 0.76
	3	180	-1.53	-0.04	2.11 ± 1.23
	4	355	-1.52	-0.08	-0.28 ± 0.46

All ratios are reported with respect to an arbitrary reference frame (RG), notionally that of a commercially-sourced methane sample (reference gas). Analytical uncertainties for $\delta^{13}\text{C}$ and δD values are both $\pm 0.2\text{‰}$ (95% confidence intervals). Uncertainties listed for $\Delta^{13}\text{CH}_3\text{D}$ are 95% confidence intervals; the last digit (hundredths place) is not significant. Note that error bars for $\Delta^{13}\text{CH}_3\text{D}$ data are relatively large ($\sim 1\text{‰}$) because sample sizes were small and acquisition time was limited. Longer counting would have yielded better errors.