Compound-Specific Chlorine Isotope Analysis: A Comparison of Gas Chromatography/Isotope Ratio Mass Spectrometry and Gas Chromatography/Quadrupole Mass Spectrometry Methods in an Interlaboratory Study

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S *Supporting Information*

ABSTRACT: Chlorine isotope analysis of chlorinated hydrocarbons like trichloroethylene (TCE) is of emerging demand because these species are important environmental pollutants. Continuous flow analysis of noncombusted TCE molecules, either by gas chromatography/isotope ratio mass spectrometry (GC/IRMS) or by GC/quadrupole mass spectrometry (GC/qMS), was recently brought forward as innovative analytical solution. Despite early implementations, a benchmark for routine applications has been missing. This study systematically compared the performance of GC/qMS versus GC/IRMS in six laboratories involving eight different instruments (GC/IRMS, Isoprime and Thermo MAT-253; GC/qMS, Agilent 5973N, two Agilent 5975C, two Thermo DSQII, and one Thermo DSQI). Calibrations of $^{37}Cl/^{35}Cl$ instrument data against the international SMOC scale (Standard Mean Ocean Chloride) deviated between instruments and over time. Therefore, at least two calibration standards are required to obtain true differences between samples. Amount dependency of $\delta^{37}Cl$ was pronounced for some instruments, but could be eliminated by corrections, or by adjusting amplitudes of standards and samples. Precision decreased in the order GC/IRMS ($1\sigma \approx 0.1‰$), to GC/qMS ($1\sigma \approx 0.2-0.5‰$ for Agilent GC/qMS and $1\sigma \approx 0.2-0.9‰$ for Thermo GC/qMS). Nonetheless, $\delta^{37}Cl$ values between laboratories showed good agreement when the same external standards were used. These results lend confidence to the methods and may serve as a benchmark for future applications.

Over the past 15 years, the study of stable isotopes in environmental pollutants has increased enormously, triggered by the development of continuous flow analytical techniques for isotopes such as $^{13}C/^{12}C$, $^{15}N/^{14}N$, $^{2}H/^{1}H$. Target compounds are isolated from interfering sample components through chromatographic separation, either by gas or by liquid chromatography (GC or LC). Then, they are combusted (or pyrolyzed) online to simple analyte gases (e.g., CO_2, N_2, H_2) which are directly transferred in the continuous helium carrier stream to an isotope-ratio mass spectrometer (IRMS). Following this scheme, suitable analytical techniques for measurement of carbon, nitrogen, and hydrogen isotopes are by now quite well established for an increasing number of environmental pollutants.

It has been demonstrated that such compound-specific isotope analysis is particularly insightful when isotopes are analyzed for two or more elements in environmental contaminants because of enhanced isotopic information. Multiple isotope analysis may provide unique mechanistic insight into contaminant degradation pathways,[1−8] and may also distinguish between different contamination sources.[9,10] However, such multielement isotope information has not been available for many priority pollutants, most prominently chlorinated ethylenes. For these prevalent organic groundwater contaminants online isotope analysis has until recently been limited to carbon raising the need for compound specific chlorine isotope analysis.

Continuous flow chlorine isotope analysis has been prevented by the necessity of converting compounds into analytes such as methyl chloride[11] or cesium chloride[12] which cannot be generated online in a carrier gas flow. Offline analysis, on the other hand, is carried out on bulk samples, which requires tedious separation of compounds from interfering components.

Therefore, previous chlorine isotope studies have focused mainly on pure compounds, either on the determination of chlorine isotope composition of chlorinated ethylenes of different manufactures[10,13−16] or on the measurement of chlorine isotope fractionation during evaporation of pure chlorinated ethylenes.[17,18] Studies involving mixtures of chlorinated ethylenes, either from field studies or from laboratory degradation experiments, have been limited.[19−25]

Recently, innovative approaches were brought forward for online isotope analysis of chlorinated ethylenes. The first approach[26] is unique for GC/IRMS because it does not include a combustion/pyrolysis step, as is typically the case for isotope analysis of C, N, and H. Instead, intact noncombusted chlorinated ethylene molecules are directly transferred within the He carrier stream into the IRMS ion source. There, they are ionized and fragmented, and selected isotopologue ions, or isotopologue ion fragments, are recorded simultaneously in dedicated collector cup configurations. In order to correct for instrument drifts, values are measured in comparison to reference/monitoring peaks which are introduced via a dual inlet system and consist of the same target analyte [e.g., trichloroethylene (TCE) pulses when TCE is measured]. Resultant machine delta values are converted into delta values relative to the international SMOC (Standard Mean Ocean Chloride) standard[27] by external calibration with independently characterized secondary standards, again of the same target analyte (i.e., a TCE standard calibrated to SMOC when TCE is measured).

An alternative approach[24] has been brought forward by Sakaguchi-Söder et al. with gas chromatography coupled to conventional quadrupole mass spectrometry (GC/qMS). Also, here analysis is conducted on noncombusted molecules. In contrast to the IRMS method, however, compound-specific standards were not available so that isotope ratios were calculated from ion multiplet intensities of molecular and fragment ions.[24] A theoretical justification for both approaches (GC/IRMS and GC/qMS) was subsequently given by Elsner and Hunkeler.[28] Very recently, the GC/qMS approach was further tested and modified by Aeppli et al.[30] In contrast to Sakaguchi-Söder et al.,[24] Aeppli et al.[30] considered only molecular ions for their calculations, and they performed a calibration with external standards to obtain δ values on the SMOC scale.

These two new analytical concepts are promising, and each has its specific advantages: Whereas the GC/IRMS provides high precision for a narrow range of compounds, GC/qMS instruments are shown to be not as precise, yet universal with respect to target analytes. Although both approaches have been spearheaded to a point where compound-specific values on the SMOC scale can be obtained, a systematic comparison between the performances of the two techniques has not yet been carried out. In particular, to ensure that the results derived by both concepts and in different laboratories are indeed reliable, the following fundamental aspects remain to be investigated experimentally.

Precision. To date, the precision of the two different methods (GC/IRMS versus GC/qMS) has not yet been systematically compared with the same substance on different instruments and in different laboratories to obtain a representative performance overview.

Amount Dependency. Stable isotope measurements are known to show an amount dependency. On the one hand, precision deteriorates with smaller sample amounts when approaching the shot-noise limit of the detector.[31,32] On the other hand, space-charge effects in the ion source of the IRMS may cause a nonlinear ionization efficiency at higher molecule abundance. These effects must be corrected to obtain accurate values. Alternatively, a strict standard bracketing strategy may be adopted where standard peak amplitudes are in the same range as the analyte peaks (within 20% difference[30]). These observations demand a comprehensive survey for the case of continuous flow chlorine isotope analysis.

Referencing and Standardization. Ideally, peaks of the target analyte are introduced at the beginning and at the end of each gas chromatographic run in order to correct for instrument drift (monitoring/"reference" gas). This option is implemented in IRMS, but typically not in qMS instruments. In addition, when using either GC/IRMS or GC/qMS, samples should be bracketed by external standards which have been characterized relative to SMOC by an independent method beforehand. Online chlorine isotope analysis conducts the measurements on noncombusted target molecules. Therefore, monitoring gas and standards must have the same molecular structure as the target compound.

For both GC/IRMS and GC/qMS, calibrations with two external compound-specific standards may be difficult to implement in routine practice, however, so that it could be an attractive shortcut to use just one external standard. Occasionally, the absolute scale is not even of importance, for example in laboratory experiments where kinetic isotope effects are determined from the relative difference between samples. Are external standards even required in such a case? A systematic investigation is needed to explore how calibration is best accomplished for online chlorine isotope analysis and whether standard bracketing is necessary.

Accuracy[34]. Even if a two-point calibration is performed it remains to be investigated whether the same result is indeed obtained when the same standards are used (a) with the same method but different instruments in different laboratories and (b) with different methods (GC/IRMS versus GC/qMS).

Therefore, in this study the performance of the two chlorine isotope techniques was examined by analyzing TCE standards in six different laboratories by eight different instruments. Six GC/qMS instruments were used: one Agilent 5973N, two Agilent 5975C, one Thermo DSQI, and two Thermo DSQII quadrupole mass selective detectors (in Darmstadt, Tübingen, Neuchâtel, Duisburg-Essen, Neuchâtel, and Duisburg-Essen, respectively). The other two laboratories used GC/IRMS instrumentation: one (Waterloo) an Isoprime Limited instrument (previously known as Micromass, U.K.) and the other (München) a Thermo Fisher Finnigan MAT 253 device. Both of them have a dedicated detector cup configuration to acquire the mass-to-charge ratios m/z of 95 and 97 (single dechlorinated fragment ion of TCE).

The first major objective of this work was to test for the precision and the quantification limit of the method. To this end, an aliquot of pure TCE was sent out to the participating laboratories, and they were asked to perform quintuplicate analyses of increasingly smaller amounts to span a range of concentrations typical of their method. Also, it was our aim to test the effect of amount dependency, that is, to compare nonlinearity effects of different instruments, and to test whether they can be eliminated by proper protocols.

The second major objective was to evaluate how instrument data should be calibrated to the international SMOC scale. It was our aim to examine how much the GC/qMS "raw" R ratios changed over time, how values improved after calibration with

external standards, and whether none, one, or two external standards are required in routine applications.

The third major objective was, finally, to evaluate the accuracy of results obtained in different laboratories, as well as the total uncertainty of the measurement. To this end, five pure unknown TCE samples were sent out along with two pure TCE standards (EIL-1, $\delta^{37}Cl$ = +3.05‰; EIL-2, $\delta^{37}Cl$ = −2.70‰) that had been characterized beforehand at the University of Waterloo. Laboratories were asked to analyze these samples relative to their in-house standards (monitoring gas peaks for GC/IRMS, external standards for GC/qMS) and to report their instrument data (machine δ values for GC/IRMS, calculated "raw" R ratios for GC/qMS).

This round robin test differs from a proficiency test in two aspects. (1) Absolute trueness of $\delta^{37}Cl$ values was not a primary objective. Although the TCE calibration standards (EIL-1 and EIL-2) are calibrated to SMOC at the University of Waterloo using secondary TCE standards (see Experimental Section), we did not attempt to characterize associated measurement errors. Instead, our focus was on internal consistency of measurements following a study design of Brand and Coplen:[35] it was tested whether the same results were obtained in different laboratories when the same standards were used as anchors for calibration, irrespective of their absolute trueness on the SMOC scale. (2) This study had the objective to test different methods, rather than ranking different laboratories. We therefore did not suppress information exchange between participating laboratories, but rather catalyzed a consistent measurement approach (Table 1). For this reason, six out of eight instrument setups in this study used headspace analysis, since this injection technique is known to be associated with minimal carbon isotope fractionation effects.[33] For the same reason (to obtain a "best case" result as a benchmark for further studies) a reductionist approach was chosen where samples contained pure TCE rather than complex substance mixtures.

EXPERIMENTAL SECTION

Chemicals. TCE was purchased from different manufacturers. The source and purity of the different products are summarized in Table S1 (Supporting Information). The pure TCE products were divided into 1.8 mL glass vials using a 50 mL glass syringe, to ensure homogeneity. Vials were filled without headspace and sealed immediately with Teflon lined caps, to avoid volatilization. These vials were shipped to all participating laboratories. Neither evaporation from closed vials nor decomposition was observed during the time of this interlaboratory test. The secondary standards EIL-1 and EIL-2 were calibrated to the SMOC scale (n = 15) against defined standards using a GC/IRMS in Waterloo[26] where the calibration at equal peak amplitudes so that no correction for amount dependency was necessary. The standard deviation replicate of measurements of the TCE standards were 0.07‰ and 0.11‰, respectively (n = 15). Standards were kept in 2.0 mL vials sealed with Teflon/silicone (PTFE/silicone) septa caps and stored in the dark. Analysis of the stored standards along a period of two years indicated no shift in their isotopic composition.

Chlorine Isotope Measurements. Chlorine isotope composition was measured in each laboratory either by GC/qMS or GC/IRMS. The following instruments were used (detailed analytical methods are provided in the Supporting Information, as well as briefly summarized in Table 1).

Tübingen. An Agilent 7890A GC coupled to an Agilent 5975C quadrupole mass selective detector (Santa Clara, CA) was used for measurements. Headspace injections were performed using an automatic multipurpose sampler (Gerstel, Mülheim an der Ruhr, Germany). This instrument setup will be referred in the following text as qMS-Agilent-1.

Darmstadt. A 6890N GC coupled to an Agilent 5973N quadrupole mass selective detector (Santa Clara, CA) was used. Samples were preconcentrated by a purge and trap system PTA 3000 from IMT (Moosbach, Germany). This instrument setup will be referred to in the following text as qMS-Agilent-2.

Neuchâtel. A Thermo Trace GC−DSQII MS (Thermo Fisher Scientific, Waltham, MA) was used for measurements. Headspace injections were performed using a CombiPal autosampler (CTC Analytics, Zwingen, Switzerland). This instrument will be referred to in the following text as qMS-Thermo-1.

Additionally, an Agilent 7890A GC coupled to an Agilent 5975C quadrupole mass selective detector (Santa Clara, CA) was tested in Neuchâtel. Headspace injections were performed using a CombiPal autosampler (CTC Analytics, Zwingen, Switzerland). This instrument will be referred to in the following text as qMS-Agilent-3.

Waterloo. An Agilent 6890 GC coupled to a continuous flow-IRMS (IsoPrime, Micromass, currently elementar) was used for measurements. Injections were carried out using a CombiPal (CTC Analytics, Zwingen, Switzerland) solid-phase microextraction (SPME) autosampler. This instrument will be referred to in the following text as IRMS-Isoprime.

Duisburg-Essen. A Thermo Trace GC−DSQII MS (Thermo Fisher Scientific, Waltham, MA) and a Thermo Trace GC−DSQI MS (Thermo Fisher Scientific, Waltham, MA) were used for measurements. Both instruments were equipped with a CombiPal autosampler (CTC Analytics, Zwingen, Switzerland) for headspace injections. These instruments will be referred to in the following text as qMS-Thermo-2 and qMS-Thermo-3, respectively.

München. A Trace GC (Thermo Fisher Scientific, Milan, Italy) directly coupled to a Finnigan MAT 253 IRMS (Thermo Fisher Scientific, Bremen, Germany) was used for measurements. Headspace injections where carried out using a Concept autosampler (PAS Technologies, Magdala, Germany). This instrument will be referred to in the following text as IRMS-Thermo.

Each laboratory was allowed to analyze the samples according to their optimized methods with regard to recorded masses, peak integration, dwell time, etc.

Calculations, GC/qMS. Raw isotope ratios were calculated from the ion abundance of the mass spectrum. According to Sakaguchi-Söder et al.,[24] chlorine isotope ratios of the target compounds can be calculated as a weighted average from isotopologue ion multiplets according to the general expression

$$R_X = a \times R_m + b \times R_1 + c \times R_2 \ldots + n \times R_{n-2} \qquad (1)$$

where $a, b, ..., n$ are the normalized intensities of the various peak groups and where R is the isotope ratio derived from the respective peak group. Using this basic concept, Sakaguchi-Söder proposed a series of mathematical equations to determine chlorine isotope ratios for six chlorinated ethenes, three chlorinated ethanes, atrazine, and two dioxin congeners.[29] For the

Table 1. Technical Parameters and Summary of Main Results Obtained on the Different Instruments

	GC/IRMS	GC/IRMS	GC/qMS	GC/qMS	GC/qMS	GC/qMS	GC/qMS	GC/qMS
instrument code	IRMS-Thermo	IRMS-Isoprime	qMS-Agilent-1	qMS-Agilent-2	qMS-Agilent-3	qMS-Thermo-1	qMS-Thermo-2	qMS-Thermo-3
laboratory	München	Waterloo	Tübingen	Darmstadt	Neuchâtel	Neuchâtel	Duisburg-Essen	Duisburg-Essen
instrument manufacturer	Thermo	IsoPrime	Agilent	Agilent	Agilent	Thermo	Thermo	Thermo
qMS or IRMS	Finnigan MAT 253	IsoPrime, Micromass	Agilent 5975C	Agilent 5973N	Agilent 5975C	DSQII	DSQII	DSQI
m/z	95, 97	95, 97	60, 62, 95, 97, 130, 132	60, 62, 95, 97, 130, 132	130, 132	60, 62, 95, 97, 130, 132	60, 62, 95, 97, 130, 132	60, 62, 95, 97, 130, 132
dwell time	∞	∞	30 ms	100 ms	50 ms	30 ms	10 ms	10 ms
GC	Thermo Trace GC	Agilent 6890	Agilent 7890A	Agilent 6890N	Agilent 7890A	Thermo Trace GC	Thermo Trace GC	Thermo Trace GC
flow	1.4 mL min⁻¹	1.8 mL min⁻¹	1.0 mL min⁻¹	1.0–2.0 mL min⁻¹	1.2 mL min⁻¹	1.5 mL min⁻¹	1.0 mL min⁻¹	1.0 mL min⁻¹
split	1:15	splitless	1:10–1:100	splitless	1:10	1:10	1:10	1:10
column	DB-5 (30 m × 0.25 mm × 0.25 μm, Agilent J&W) + a wax column (60 m × 0.25 mm ×0.50 μm, Supelcowax 10)	DB-5 (60 m × 0.320 mm × 1 μm, Agilent J&W)	RTX-VMS (60 m × 0.25 mm × 1.4 μm, Restek)	DB-624 (60 m × 0.25 mm × 1.4 μm, Agilent J&W)	DB-5 (30 m × 0.25 mm × 0.25 μm, Agilent J&W)	DB-VRX (60 m × 0.25 mm × 1.4 μm, Agilent J&W)	RTX-VMS capillary column (60 m × 0.32 mm, 1.8 μm, Restek)	StabilwaxDA (60 m × 0.32 mm, 1 μm, Restek)
injection technique	automated HS	SPME	automated HS	purge and trap	automated HS	automated HS	automated HS	automated HS
peak integration software	Isodat version 2.5	MassLynx Inorganic version 4017	ChemStation software, version E.01.01.355; RTE integrator	ChemStation version D.02.00.237; ChemStation Integrator	ChemStation version E.02.01.1177	Excalibur version 1.4	Excalibur version 1.4	Excalibur version 1.4
typical precision (1σ)	<0.1‰ᵃ	0.09 ± 0.03‰ᵇ	0.18–0.50‰ᵃ; 0.26 ± 0.05‰ᵇ	0.15–0.57‰ᵃ; 0.27 ± 0.06‰ᵇ	0.16–0.49‰ᵃ; 0.37 ± 0.11‰ᵇ	0.37–0.92‰ᵃ; 0.71 ± 0.11‰ᵇ	0.35–0.84‰ᵃ,ᵈ; 0.35‰ ± 0.33ᵇ,ᵉ	0.63 ± 0.09‰ᵇ,ᵈ; 0.30 ± 0.11‰ᵇ,ᵉ
detection limit	5.3 ng for 1σ<0.15‰	not determined	20 ng for 1σ<0.5‰	10 ng for 1σ<0.5‰	>36 ng for 1σ<0.5‰	12.5 ng for 1σ<1.0‰	0.4 ng for 1σ<0.85‰ᶜ	not determined
mass dependency	1‰ over 0.8–53 ng	high (not determined in this study)	2‰ over 0.4–40 ng	6‰ over 15–1023 ng	<1‰ over 0.75–75 ng	8‰ over 12.5–800 ng	20‰ over 1.0–200 ngᶜ	not determined
two-point calibration curve slope	1.08 ± 0.02		1.20 ± 0.05	0.72 ± 0.03	1.09 ± 0.06	1.24 ± 0.22	0.91 ± 0.14ᶜ; 1.35 ± 0.22ᵈ	1.31 ± 0.12ᶜ; 1.33 ± 0.10ᵈ
av uncertainty of calibrated SMOC values ± standard deviation for replicate measurement of different TCE compoundsᵉ	0.040 ± 0.003‰ ($n = 10$, $m = 20$)		0.11 ± 0.02‰ ($n = 10$, $m = 20$)	0.08 ± 0.01‰ ($n = 10$, $m = 20$)	0.10 ± 0.01‰ ($n = 10$, $m = 20$)	0.47 ± 0.22‰ ($n = 5$, $m = 10$)	0.53 ± 0.13‰ᶜ and 0.53 ± 0.10‰ᵈ ($n = 5$, $m = 10$)	0.39 ± 0.02‰ᶜ and 0.21 ± 0.03‰ᵈ ($n = 5$, $m = 10$)
range of z-scoresᶠ	0.50–1.05		0.06–1.58	0.08–2.37	0.26–0.79	0.09–1.23	0.13–1.44ᶜ; 0.58–13.73ᵈ	0.13–0.69ᶜ; 0.68–3.45ᵈ

ᵃ Values obtained in the linearity test of Figure 1 (TCE repeatedly injected at varying amount). ᵇ Values obtained in the accuracy test of Figure 3b (TCE repeatedly injected at same amount). ᶜ Default peak integration settings. ᵈ Modified peak integration settings. ᵉ Values obtained in the accuracy test of Figure 3c. ᶠ Absolute value of z-score, calculated for accuracy test.

specific case of TCE this gives

$$R_{TCE} = \frac{I_{130}}{I_{130} + I_{95} + I_{60}} \times \left(\frac{1}{3} \cdot \frac{I_{132}}{I_{130}}\right)$$
$$+ \frac{I_{95}}{I_{130} + I_{95} + I_{60}} \times \left(\frac{1}{2} \cdot \frac{I_{97}}{I_{95}}\right)$$
$$+ \frac{I_{60}}{I_{130} + I_{95} + I_{60}} \times \left(\frac{I_{62}}{I_{60}}\right)$$
$$= \frac{1}{3} \cdot \frac{I_{132}}{I_{130} + I_{95} + I_{60}} + \frac{1}{2} \cdot \frac{I_{97}}{I_{130} + I_{95} + I_{60}}$$
$$+ \frac{I_{62}}{I_{130} + I_{95} + I_{60}} \tag{2}$$

where I_{132} and I_{130} are peak intensities of the most abundant peaks in the molecular ion group, I_{97} and I_{95} in the group of the single dechlorinated fragment ion, and I_{62} and I_{60} in the group of the double dechlorinated fragment. The binomial coefficients account for the fact that ^{37}Cl can sit in either of three positions in the molecular ion, in either of two positions in the single dechlorinated fragment, and in only one position in the double dechlorinated fragment.

Alternatively, as validated theoretically by Elsner and Hunkeler,[28] isotope ratios should be directly attainable from any pair of isotopologues, either of the parent ion, or of any fragment ion. For the specific case of TCE this gives

$$R_{TCE} = \frac{1}{3} \cdot \frac{I_{132}}{I_{130}} = \frac{1}{2} \cdot \frac{I_{97}}{I_{95}} = \frac{I_{62}}{I_{60}} \tag{3}$$

R_{TCE} ratios calculated according to eqs 2 or 3 are subsequently converted to an internal δ scale by referencing against an external laboratory standard, R_{std}, measurements of which bracketed those of the samples:

$$\delta = [(R_{sample}/R_{std}) - 1] \times 1000 \tag{4}$$

Choice of Ions. Consequently, isotope values may be derived from GC/qMS measurements in different ways. Calculations may consider only molecular ions,[30] also fragment ions (eqs 1 and 2), only the most abundant two peaks[30] (eqs 1 and 2), or also the less abundant peaks of a given multiplet. In our study, laboratories were allowed to analyze the samples according to their optimized methods. The different calculation approaches were recently compared experimentally.[36] It was found that considering only molecular ions resulted in less precise raw isotope ratios than considering the two most abundant ions of each fragment group (for TCE: 60, 62, 95, 97, 130, 132). On the other hand, including all nine ions of TCE resulted in less precise raw isotope ratios again, since analyzing too many ions conflicted with maintaining a sufficient dwell time and scan rate. Although significant, the magnitude of the differences reported in ref 36 suggests that different calculation schemes are not a likely reason for the systematic differences observed in this study.

Peak Integration Parameters. Most participating laboratories relied on default peak integration settings of their software. In one case (qMS-Thermo-2 and qMS-Thermo-3 in Duisburg-Essen) evaluation was performed in two ways: (1) with default peak parameters; (2) with parameters such that the peak start and end points were as similar as possible for each fragment ion. (A defined adjustment proved difficult, since the software

"Excalibur" treated every ion as an independent compound.) Unless mentioned otherwise, results of the optimized evaluation method 2 are presented.

Calculations, GC/IRMS. In the GC/IRMS method, target peaks were automatically evaluated against monitoring peaks that were introduced through a dual inlet system during each sample run using the respective software (Isodat 2.0, Thermo Electron Corporation, and MassLynx Inorganic, V. 4017, GV Instruments, currently Isoprime Limited, U.K., in München and Waterloo, respectively).

External Calibration. For both analytical techniques (GC/qMS and GC/IRMS), these "raw" δ values were subsequently converted to the international SMOC scale using two previously characterized TCE standards, EIL-1 and EIL-2, which are 5.75‰ apart on the $\delta^{37}Cl_{SMOC}$ scale. Internal values were transformed according to a two-point linear regression trend line. The slope's error of the calibration curve, as it appears in the following sections, was calculated as 95% confidence interval (slope's standard error multiplied by the student t for $\alpha = 0.05$). The performance of the different laboratories in terms of accuracy was tested by z-score,[40] defined as $z = (x - X)/\sigma$, where x is the average value obtained in the respective laboratory, X is the assigned value (or "consensus value"), and σ is the standard deviation of the measured value in the laboratory. The assigned value, X, was set as the results obtained by IRMS-Isoprime from Waterloo, following the long experience that has been gained in this laboratory over the years. The uncertainty of SMOC values derived from multiple measurements of the same sample and following the uncertainty of the calibration curve was calculated as

$$S_m = \left(\frac{S_r}{|M|}\right) \sqrt{\frac{1}{m} + \frac{1}{n} + \frac{(y_m - y_{avg})^2}{M^2 \sum (x_i - x_{avg})^2}} \tag{5}$$

where S_m is the standard deviation of a reported x value derived from its associated instrument value y through calibration, S_r is the standard deviation of the calibration's regression, M is the slope of the calibration curve, m is the number of $x-y$ pairs of the calibration curve, n is the number of replicate measurements of the unknown, y_m is the measured y value to be calibrated, y_{avg} is the average y value of the calibration curve, x_i is the concentration of the standards, and x_{avg} is the average concentration of the standards.

The S_m value can be further used to calculate the 95% confidence interval, Δ, of the value on the SMOC scale by student's t as

$$\Delta = t \frac{S_m}{\sqrt{n}} \tag{6}$$

RESULTS AND DISCUSSION

Precision and Amount Dependency ("Linearity"). To test for the precision and linearity of the different analytical approaches, two aspects were addressed: (1) How precise are replicate measurements at a given injected amount? (2) How precise are measurements at a range of varying injected amounts? Figure 1 presents these two aspects for replicate TCE measurements over a wide range of amounts. Typically, 1σ ($n = 5$, unless specified otherwise) values obtained for replicate injections of the same amount of TCE were higher on GC/qMS instruments than on GC/IRMS. One standard deviation (1σ) of GC/qMS
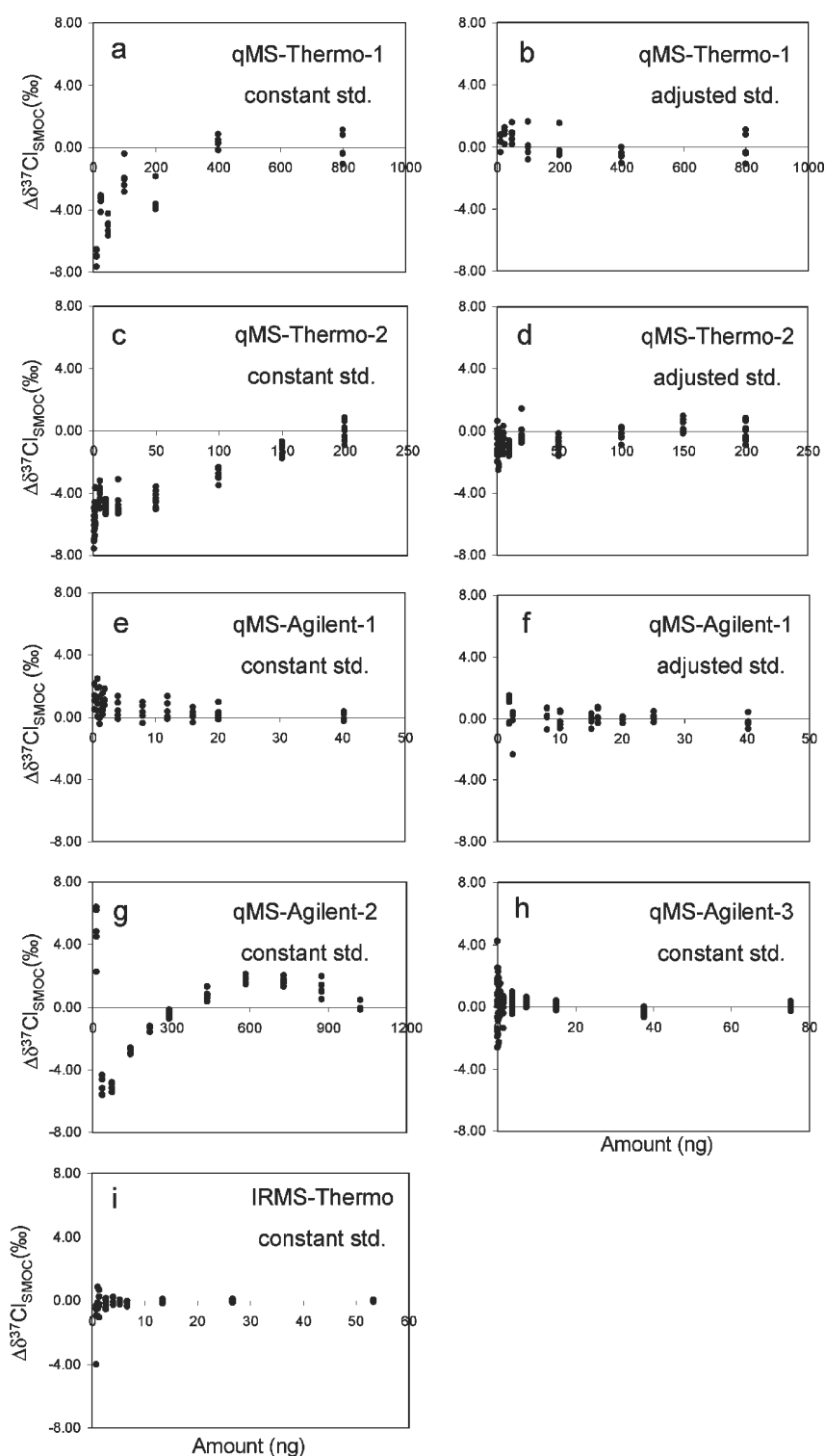
Figure 1. Measured δ^{37}Cl depending on the on-column amount of TCE. Values are presented as the difference from the average of the replicates in the highest on-column amount. (a) qMS-Thermo-1 (automated headspace injections, amplitude of standards along the sequence was held constant). (b) qMS-Thermo-1 (automated headspace injections, standards were adapted to the amplitude of target peaks). (c) qMS-Thermo-2 (automated headspace injections, amplitude of standards along the sequence was held constant). (d) qMS-Thermo-2 (automated headspace injections, standards were adapted to the amplitude of target peaks). (e) qMS-Agilent-1 (automated headspace injections, amplitude of standards along the sequence was held constant). (f) qMS-Agilent-1 (automated headspace injections, standards were adapted to the amplitude of target peaks). (g) qMS-Agilent-2, (purge-and-trap injection, amplitude of standards along the sequence was held constant). (h) qMS-Agilent-3 (automated headspace injections, amplitude of standards along the sequence was held constant). (i) IRMS-Thermo, automated headspace injections where reference peaks were introduced at the beginning and end of each GC run (amount was held constant).

instruments was in the following range (on-column amounts and corresponding figure panels are given in brackets): 0.18−0.50‰ on the qMS-Agilent-1 (0.4−40 ng; Figure 1e), 0.15−0.57‰ on the qMS-Agilent-2 (37−1000 ng; Figure 1g), and 0.16−0.49‰ on the qMS-Agilent-3 (0.75−75 ng; Figure 1h; n = 15 for each amount). On the Thermo instruments, lower precisions were observed with typical 1σ in the range 0.37−0.92‰ on the qMS-Thermo-1 (13−800 ng; Figure 1a), and 0.35−0.84‰ on the qMS-Thermo-2 (0.4−200 ng; Figure 1c; up to n = 10 for each amount). In GC/IRMS measurements, in contrast, much higher precisions were observed, with 1σ = 0.06‰ at 53 ng on the IRMS-Thermo (Figure 1i). A similar precision was obtained earlier on the IRMS-Isoprime,[26] but was not evaluated again in this study. At 1.1 ng TCE the value for 1σ was still 0.6‰ at the IRMS-Thermo, representing the lower limit for precise chlorine isotope analysis (Figure 1i). The better performance of GC/IRMS likely reflects the ability to simultaneously acquire different ions in a dedicated cup configuration, as well as to introduce monitoring gas peaks during each gas chromatographic run.

Amount Dependency. The second aspect of precision is the "linearity": How precise are measurements in a range of varying injected amounts (i.e., are the measured values amount dependent)? Here, our study clearly shows that amount dependency ("non-linearity") effects can be very substantial, and where they occur appropriate correction is mandatory, such as brought forward by Shoukar-Stash et al., in 2006.[26] Surprisingly, similar analytical concepts did not necessarily entail an identical pattern of amount dependency. For example, on the IRMS-Thermo linearity was significantly better than on the IRMS-Isoprime.[26] Even instruments of similar construction such as qMS-Thermo-1 and 2, or qMS-Agilent-1, 2, and 3 gave different amount dependency trends. It should be noted, however, that results from different instruments do not necessarily reflect identical ranges of injected amounts; hence, the data evaluation must be performed individually by each laboratory for the analyzed concentration range.

Importantly, our results demonstrate that a proper standardization scheme, as brought forward by Aeppli et al.,[30] may eliminate the need for an amount dependency correction. Here, samples are bracketed by known external standards along the sequence of measurements. Our data clearly shows that amount dependency effects are pronounced if standards have a constant amplitude whereas amplitudes of the samples vary (Figure 1e,a,c for qMS-Agilent-1, qMS-Thermo-1, and qMS-Thermo-2, respectively). In contrast, they become negligible if the amount of the standard is adapted to the sample (Figures 1f, 1b, and 1d for qMS-Agilent-1, qMS-Thermo-1, and qMS-Thermo-2, respectively). This strategy of adjusting samples and standards to the same concentration is clearly advantageous when samples with known concentrations are analyzed. Alternatively, good linearity is desirable (e.g., as observed on the IRMS-Thermo, or on the qMS-Agilent-3). If linearity is not as good, however, and if sample concentrations are not known beforehand, an amount dependency correction may have to be performed on the basis of amount dependency trends that are defined by running additional standards with differing amounts along the sequence.

Besides processes in the ion source, also the injection technique may be of importance since isotope discrimination during injection might contribute to the amount dependency effects. To test this hypothesis a comparison of manual headspace injection versus purge-and-trap was performed at the qMS-Agilent-2, and a comparison between automated headspace injections versus SPME injections was performed at the qMS-Agilent-1 (Figure S1 in the Supporting Information). The comparison shows that the injection technique alone cannot explain the strong amount dependency on the qMS-Agilent-2. It remains to be investigated whether there is potential for improvement by a specific tuning for enhanced linearity as is well-known from GC/IRMS applications.[37]

Calibration of Data to the SMOC δ Scale. Calibration by external standards (1) corrects for instability of the instrument; (2) projects instrument values on the international SMOC scale through correction for an offset; (3) corrects for distortion relative to the SMOC scale (e.g., if two standards that are 5‰ apart differ by only 4‰ on the instrument scale).

Instability of the Instrument. The variability of "raw" instrument values R calculated from mass spectra according to eq 2 was investigated on the qMS-Agilent-1 instrument (Figure 2a). Figure 2a shows that large differences in the R values were obtained at different dates and with different filaments (differences of 0.004 corresponding to 12‰ on the δ scale; eq 4). However, these differences could be corrected when standards were analyzed in the same sequence (Figure 2b). Thus, temporal variations in the instrument largely cancel out when standards are exposed to the same conditions as the samples. These results emphasize the need for standardization in GC/qMS when determination of absolute values is of concern or when a set of samples is not analyzed in the same sequence: Although in theory calculated R values are directly related to the $^{35}Cl/^{37}Cl$ ratio of the bulk material[24] (see eq 2), our results show that these R values are influenced by instrumental factors and require proper correction.

Slopes of Two-Point Calibrations. To obtain absolute values on the international SMOC scale, a two-point calibration was performed in each of the participating laboratories using the EIL-1 and EIL-2 standards. Although such two-point isotopic calibrations are well established in dual inlet-IRMS and EA-IRMS measurements (see, e.g., refs 35,38), they are rare in compound specific isotope analysis by GC/IRMS. Figure 2c presents the resultant calibration curves of the different laboratories. The comparison reveals that curves were different between laboratories, giving slopes for GC/qMS instruments of 1.20 ± 0.05 (qMS-Agilent-1), 0.72 ± 0.03 (qMS-Agilent-2), 1.09 ± 0.06 (qMS-Agilent-2), 1.24 ± 0.22 (qMS-Thermo-1), 0.91 ± 0.14 (qMS-Thermo-2 with default peak integration), 1.35 ± 0.22 (qMS-Thermo-2 after peak reintegration), 1.31 ± 0.12 (qMS-Thermo-3 with default peak integration), and 1.33 ± 0.10 (qMS-Thermo-3 after peak reintegration), and for GC/IRMS of 1.08 ± 0.02 (IRMS-Thermo). The given uncertainty corresponds to 95% confidence intervals that were calculated from the slope's standard error multiplied by the appropriate student t factor for α = 0.05.

Calibration slopes did not only differ between instruments, but also may change over time. Whereas calibration slopes of the IRMS-Thermo ranged between 0.98 ± 0.04 and 1.09 ± 0.04 in five different measurement series within a three month period, slopes of the qMS-Agilent-1 in Tübingen ranged between 1.14 ± 0.12 and 1.25 ± 0.10 in nine different measurement series within a similar time period. A longer period of observation was recorded on the IRMS-Isoprime in Waterloo over the years 2008−2010, where slopes of almost 150 distinct measurement sequences ranged from 0.90 to 1.23 (Figure 2d).

Since finding and characterizing two sufficiently different standards for every new target compound is difficult and labor intensive, at the beginning of this paper we raised the question
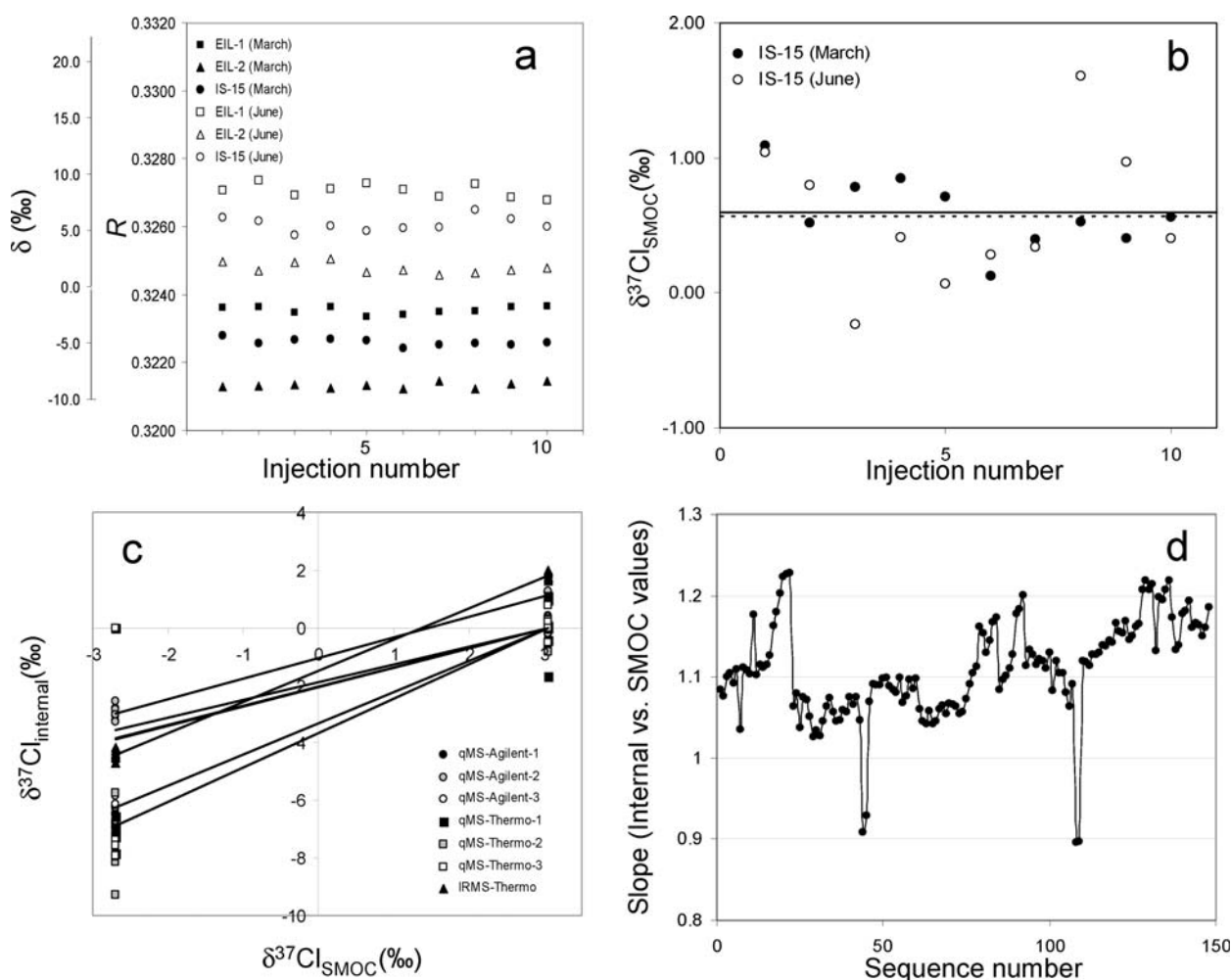
Figure 2. (a) Nominal R and corresponding δ values calculated according to eq 2 for three different types of TCE using data from qMS-Agilent-1 analysis in March and June 2010 with different filaments (constant amount injected). The R and δ values are referred to a relative scale. (b) The corresponding $\delta^{37}Cl_{smoc}$ (‰) values of the TCE product IS-15, obtained from the same qMS-Agilent-1 measurements after external calibration with EIL-1 and EIL-2. Average values for the measurements of March and of June are indicated as solid and dashed lines, respectively. (c) Calibration curves for converting the internal values to the SMOC scale in different laboratories. (d) Slopes of calibration curves measured by IRMS-Isoprime for almost 150 distinct measurement sequences during the years 2008−2010.

whether calibration with two standards is necessary if the primary focus is on differences in isotope ratios rather than on absolute numbers, and if all samples are analyzed on the same day. Would calculation of isotope ratios from isotopologue ions (e.g., according to eq 3) still give useful, internally consistent data sets, even if the numbers were not correct on an absolute scale? Our results strongly discourage such a practice. The calibration slopes clearly document a significant, nonsystematic distortion of instrument scales relative to the SMOC scale. Therefore, even *differences* between isotope values (e.g., changes in isotope ratios relative to an initial value or to an undefined internal standard) would be inaccurate without proper calibration, and would, for example, give wrong fractionation factors in a degradation experiment.

Accuracy Test. The need for a multipoint calibration is further supported by the results of the accuracy test, as shown in Figure 3. Five TCE products of different producers were analyzed in the different laboratories, using either one or two TCE standards for calibration to the SMOC scale (Figure 3; the figure's raw data is provided in the Supporting Information,

Table S2). When results were projected on the SMOC scale using one standard only, large differences were obtained (Figure 3a). In contrast, when using two calibration anchor points, dramatically better agreement was accomplished between laboratories (Figure 3b,c). Excellent agreement was observed between the two laboratories that used GC/IRMS, where values of all five products were identical within 0.2‰. Good agreement was observed with GC/qMS values where more significant differences were observed, yet generally in the range of 1σ ($n = 10$).

Accuracy and Total Uncertainty. For defining the accuracy and total uncertainty of the measurements, different strategies may be taken: (1) When asking what the uncertainty of an *average* value on the SMOC scale is, based on n replicates and a calibration curve with a defined uncertainty, eqs 5 and 6 should be applied. (2) When asking whether a single measurement belongs to, or differs from, an *average* value of n replicates, the standard deviation of the replicates should be considered, with 2σ indicating the 95% confidence interval. (3) When asking
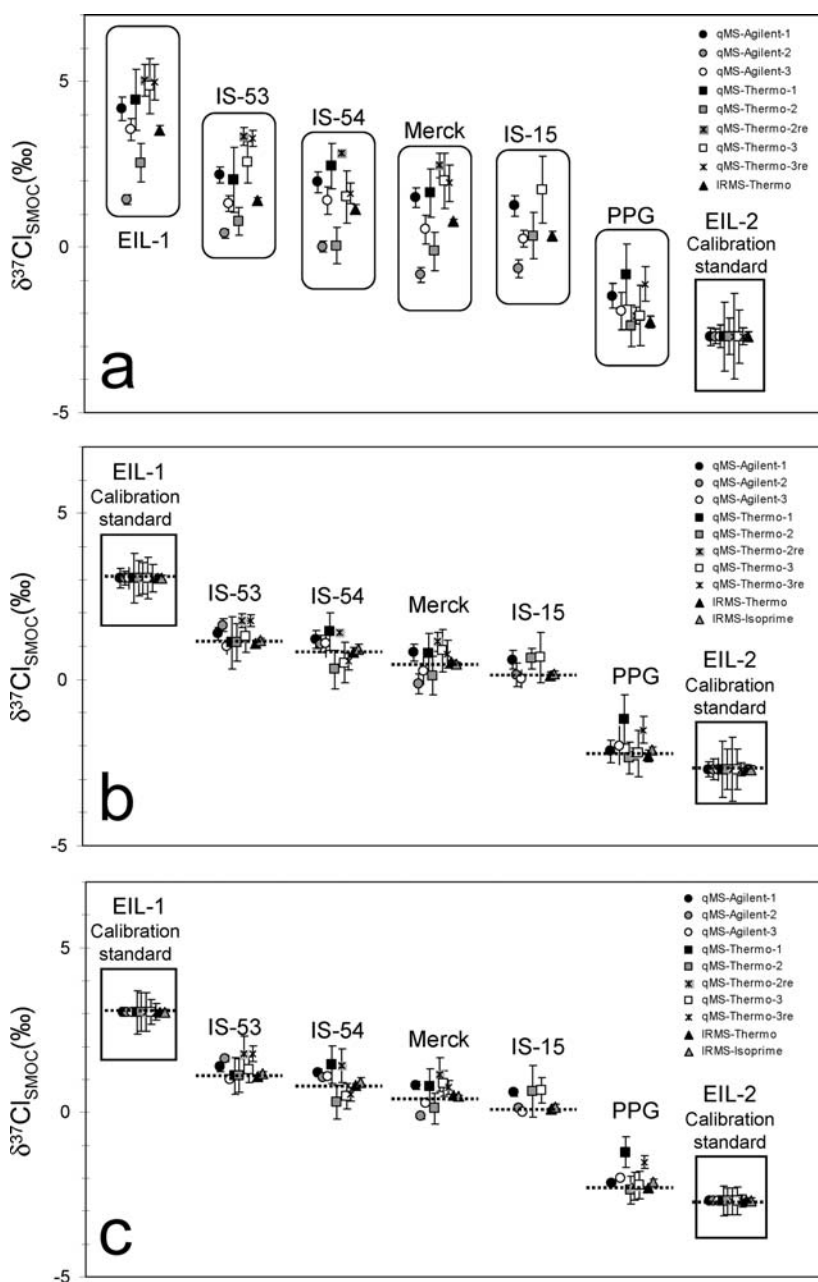
Figure 3. Measured $\delta^{37}Cl$ of 5 different TCE products in different laboratories. (a) Calibration to the SMOC scale was carried out using a single standard; error bars indicate 1σ on the internal instrument scale (y axis of Figure 2c). (b) Calibration to the SMOC scale was carried out using two standards; error bars indicate 1σ converted to the SMOC scale (x axis of Figure 2c, relevant for the z-score). (c) Calibration to the SMOC scale was carried out using two standards; error bars indicate the total uncertainty of each data point (95% confidence interval) calculated according to eqs 5–6 (error that would be reported in an interlaboratory exercise). Horizontal dashed lines represent the consensus value (i.e., the measurements on the IRMS-Isoprime in Waterloo).

whether an average value from a single laboratory is similar to a consensus value, z-scores may be calculated.

In the first case, average SMOC values are associated with (a) the uncertainty of n replicate measurements and (b) the uncertainty of the calibration curve which converts instrument values on the SMOC scale (eqs 5 and 6). Both uncertainties depend on the number of measurements, that is, n (the measurements of the unknown) and m (the number of data points in the calibration). The average uncertainty of the calibrated SMOC values are in the range 0.08–0.11‰ for the Agilent qMS instruments, 0.21–0.47‰ for the Thermo qMS instruments (note that $n = 5$, $m = 10$), and

0.04‰ for GC/IRMS-Thermo instrument ($n = 10$, $m = 20$). Values for each individual instrument are provided in Table 1.

Alternatively, in the second case (i.e., for differentiating between samples that are run in a single sequence and are therefore associated with the same uncertainty of the same calibration curve), one may consider the standard deviation of replicate injections to differentiate between samples. For 95% confidence intervals in this case, at least two standard deviations should be allowed. Therefore, if two samples, e.g., at a field site, are to be different on the 95% confidence level, they should differ at least by four standard deviations. With the data summarized in

Table 1, this amounts to about 2‰ for the Agilent GC/qMS instruments, up to 4‰ for the Thermo GC/qMS instruments, and about 0.4‰ for the GC/IRMS instruments (Table 1), depending on the number of replicate measurements. Such ranges would be in line with current guidelines for carbon isotope analysis which recommend as a rule of thumb that samples be 2‰ apart when the total uncertainty of the measurement is 0.5‰.[39]

In the third case, the data of this interlaboratory comparison allows a rough estimate of the accuracy of the different methods relative to a consensus value. If the IRMS-Isoprime results are taken as the consensus values of the unknown samples,[40] and if the averages measured by the other instruments are compared to these consensus values, the most extreme difference between each instrument and the consensus value may provide an error estimate of the instrument. Doing so, the following intervals were obtained for the GC/qMS instruments: 0.43‰ (qMS-Agilent-1), 0.60‰ (qMS-Agilent-2), 0.21‰ (qMS-Agilent-3), 0.92‰ (qMS-Thermo-1), 0.50‰ and 0.62‰ (qMS-Thermo-2, with default and modified peak integration settings, respectively), and 0.61‰ and 0.66‰ (qMS-Thermo-3, with default and modified peak integration settings, respectively). For the GC/IRMS instrument, the interval was 0.17‰ (IRMS-Thermo). In most cases, the calculated z-scores[40] for these differences were lower than 2.0 (Table 1), indicating that the specified precision agrees with deviations from the consensus value. Exceptional are IS-15 in qMS-Agilent-2, with a z-score of 2.37, and various measurements of qMS-Thermo-2 and qMS-Thermo-3 after changing its peak integration settings. Whereas the >2.0 value by qMS-Agilent-2 is exceptional for this instrument, and may stem from inadvertent sample handling, the values of the two qMS-Thermo instruments indicate a systematic bias in the estimated precision. Thus, although the change in the peak integration setting improved the precision of the qMS-Thermo-2 and qMS-Thermo-3 measurements, the accuracy test indicates that the stated precision underestimated the true uncertainty. Overall, however, deviations in the accuracy tests were in good agreement with the precision of the respective instruments indicating the absence of additional systematic errors.

Finally, we caution that in reality errors may be greater than reported in this study because samples are typically not pure and are not determined in 10-fold replicate. Also, our set of five unknown samples is small, and larger deviations may occur if a greater set of samples is analyzed. Finally, calibration in all laboratories was conducted with the same secondary standards eliminating a possible systematic bias from different standard sets.

■ CONCLUSIONS

Chlorine isotope methods are at their initial stages of application, and it is therefore crucial to systematically evaluate instrumental behavior and to define proper protocols of analytical and standardization schemes for future routine applications. This study investigated precision and accuracy, as well as different schemes of standardization, for continuous flow GC/IRMS and GC/qMS methods. Different instruments were found to present individual amount dependency trends and variable calibration slopes against the SMOC scale. The different amount dependency trends could be eliminated by standard bracketing when the amplitude of the external standard was adapted to that of the respective sample, consistent with Aeppli et al.[30] The different calibration slopes against the SMOC scale, on the other hand, emphasized the need for a careful standardization and calibration scheme in each laboratory and on each measurement day.

In particular, we found that it is mandatory to include a minimum of two compound-specific calibration standards with defined $\delta^{37}Cl_{SMOC}$ in any sequence of samples in order to obtain true differences between samples and to convert internal values to the international SMOC scale. In practice, this may be a challenge since for chlorine isotope analysis a universal standard is not as easily obtainable as, e.g., for carbon, where CO_2 can be used for all analyzed compounds.

Our study also gives the first comprehensive comparison of the precision and the total uncertainty associated with the analytical techniques. Our results suggest that isotope values can be confidently assessed to be different, if they differ at least by 0.4‰ in GC/IRMS measurements (either Isoprime or Thermo MAT-253), by 2‰ in most GC/qMS instruments measurements, and up to 4‰ in GC/qMS measurements on one of the Thermo instruments tested in this study.

On the other hand, despite lower precision, GC/qMS applications have considerable advantages over GC/IRMS. First, whereas we are aware of only two GC/IRMS instruments worldwide (München and Waterloo) that are configured to analyze $\delta^{37}Cl$ in TCE directly, without prior offline separation and conversion to methyl chloride or CsCl, a vast number of laboratories are equipped with GC/qMS instruments. Second, whereas GC/IRMS instruments are limited to analysis of C_2 chlorinated compounds, GC/qMS can be applied for $\delta^{37}Cl$ analysis in a variety of compounds, as well as in specific fragments of such compounds. This enables the introduction of chlorine isotope analysis for new compounds of interest.

Finally, our study demonstrated good to excellent agreement of $\delta^{37}Cl$ values measured with GC/IRMS and GC/qMS in different laboratories/instruments if the same external standards were used. This general agreement is very encouraging and lends confidence to both methods. The results of this interlaboratory test may, therefore, serve as a benchmark for future applications of compound-specific chlorine isotope analysis.

■ ASSOCIATED CONTENT

ⓢ Supporting Information. Methods of chlorine isotope measurements, Table S1 listing the TCE products used in this study, Table S2 summarizing $\delta^{37}Cl$ values of TCE products measured at different laboratories, and Figure S1 showing measured $\delta^{37}Cl$ in GC/qMS-Darmstadt and GC/qMS-Tübingen depending on the amount on-column using different injection methods. This material is available free of charge via the Internet at http://pubs.acs.org.

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■ REFERENCES

(1) Elsner, M.; McKelvie, J.; Couloume, G. L.; Sherwood Lollar, B. *Environ. Sci. Technol.* **2007**, *41*, 5693–5700.

(2) Elsner, M. *J. Environ. Monit.* **2010**, *12*, 2005–2031.

(3) Kuder, T.; Wilson, J. T.; Kaiser, P.; Kolhatkar, R.; Philp, P.; Allen, J. *Environ. Sci. Technol.* **2005**, *39*, 213–220.

(4) Meyer, A. H.; Penning, H.; Elsner, M. *Environ. Sci. Technol.* **2009**, *43*, 8079–8085.

(5) Penning, H.; Sørensen, S. R.; Meyer, A. H.; Aamand, J.; Elsner, M. *Environ. Sci. Technol.* **2010**, *44*, 2372–2378.

(6) Zwank, L.; Berg, M.; Elsner, M.; Schmidt, T. C.; Schwarzenbach, R. P.; Haderlein, S. B. *Environ. Sci. Technol.* **2005**, *39*, 1018–1029.

(7) Hofstetter, T. B.; Bernasconi, S. M.; Schwarzenbach, R. P. *Environ. Sci. Technol.* **2008**, *42*, 7737–7743.

(8) Vogt, C.; Cyrus, E.; Herklotz, I.; Herrmann, S.; Bahr, A.; Richnow, H. H.; Fischer, A. *Environ. Sci. Technol.* **2008**, *42*, 7793–7800.

(9) Schmidt, T. C.; Zwank, L.; Elsner, M.; Berg, M.; Meckenstock, R. U.; Haderlein, S. B. *Anal. Bioanal. Chem.* **2004**, *378*, 283–300.

(10) Shouakar-Stash, O.; Drimmie, R. J.; Frape, S. K. *J. Contam. Hydrol.* **2003**, *60*, 211–228.

(11) Holt, B. D.; Sturchio, N. C.; Abrajano, T. A.; Heraty, L. J. *Anal. Chem.* **1997**, *69*, 2727–2733.

(12) Holmstrand, H.; Andersson, P.; Gustafsson, O. *Anal. Chem.* **2004**, *76*, 2336–2342.

(13) Beneteau, K. M.; Aravena, R.; Frape, S. K. *Org. Geochem.* **1999**, *30*, 739–753.

(14) Drenzek, N. J.; Tarr, C. H.; Eglinton, T. I.; Heraty, L. J.; Sturchio, N. C.; Shiner, V. J.; Reddy, C. M. *Org. Geochem.* **2002**, *33*, 437–444.

(15) Jendrzejewski, N.; Eggenkamp, H. G. H.; Coleman, M. L. *Appl. Geochem.* **2001**, *16*, 1021–1031.

(16) Van Warmerdam, E. M.; Frape, S. K.; Drimmie, R. J.; Flatt, H.; Cherry, J. A. *Appl. Geochem.* **1995**, *10*, 547–552.

(17) Huang, L.; Sturchio, N. C.; Abrajano, T.; Heraty, L. J.; Holt, B. D. *Org. Geochem.* **1999**, *30*, 777–785.

(18) Poulson, S. R.; Drever, J. I. *Environ. Sci. Technol.* **1999**, *33*, 3689–3694.

(19) Abe, Y.; Aravena, R.; Zopfi, J.; Shouakar-Stash, O.; Cox, E.; Roberts, J. D.; Hunkeler, D. *Environ. Sci. Technol.* **2009**, *43*, 101–107.

(20) Hofstetter, T. B.; Reddy, C. M.; Heraty, L. J.; Berg, M.; Sturchio, N. C. *Environ. Sci. Technol.* **2007**, *41*, 4662–4668.

(21) Holmstrand, H.; Mandalakis, M.; Zencak, Z.; Andersson, P.; Gustafsson, O. *Chemosphere* **2007**, *69*, 1533–1539.

(22) Numata, M.; Nakamura, N.; Koshikawa, H.; Terashima, Y. *Environ. Sci. Technol.* **2002**, *36*, 4389–4394.

(23) Reddy, C. M.; Xu, L.; Drenzek, N. J.; Sturchio, N. C.; Heraty, L. J.; Kimblin, C.; Butler, A. *J. Am. Chem. Soc.* **2002**, *124*, 14526–14527.

(24) Sakaguchi-Söder, K.; Jager, J.; Grund, H.; Matthäus, F.; Schüth, C. *Rapid Commun. Mass Spectrom.* **2007**, *21*, 3077–3084.

(25) Sturchio, N. C.; Clausen, J. L.; Heraty, L. J.; Huang, L.; Holt, B. D.; Abrajano, T. A. *Environ. Sci. Technol.* **1998**, *32*, 3037–3042.

(26) Shouakar-Stash, O.; Drimmie, R. J.; Zhang, M.; Frape, S. K. *Appl. Geochem.* **2006**, *21*, 766–781.

(27) Kaufmann, R.; Long, A.; Bentley, H.; Davis, S. *Nature* **1984**, *309*, 338–340.

(28) Elsner, M.; Hunkeler, D. *Anal. Chem.* **2008**, *80*, 4731–4740.

(29) Sakaguchi-Söder, K. A New Method for Compound-Specific Stable Chlorine Isotope Analysis: Basics and Application. Ph.D. Thesis, Technische Universität, Darmstadt, Germany, 2010.

(30) Aeppli, C.; Holmstrand, H.; Andersson, P.; Gustafsson, O. *Anal. Chem.* **2010**, *82*, 420–426.

(31) Merritt, D. A.; Freeman, K. H.; Ricci, M. P.; Studley, S. A.; Hayes, J. M. *Anal. Chem.* **1995**, *67*, 2461–2473.

(32) Sherwood Lollar, B.; Hirschorn, S. K.; Chartrand, M. M. G.; Lacrampe-Couloume, G. *Anal. Chem.* **2007**, *79*, 3469–3475.

(33) Slater, G. F.; Dempster, H.; Sherwood Lollar, B.; Ahad, J. *Environ. Sci. Technol.* **1999**, *33*, 190–194.

(34) International Standardization Organization. *Accuracy (Trueness and Precision) of Measurement Methods and Results, Part 1: General Principles and Definitions. ISO 5. 5725−1*; Geneva, 1994.

(35) Brand, W. A.; Coplen, T. B. *J. Anal. Chem.* **2001**, *370*, 358–362.

(36) Jin, B.; Laskov, C.; Rolle, M.; Haderlein, S. B. *Environ. Sci. Technol.* **2011**, *45*, 5279–5286.

(37) Brand, W. A. In *Handbook of Stable Isotope Analytical Techniques*; de Groot, P. A., Ed.; Elsevier: Amsterdam, 2004; Vol. I, pp 835−858.

(38) Schimmelmann, A.; Albertino, A.; Sauer, P. E.; Qi, H.; Molinie, R.; Mesnard, F. *Rapid Commun. Mass Spectrom.* **2009**, *23*, 3513–3521.

(39) Hunkeler, D.; Meckenstock, R. U. ; Sherwood Lollar, B.; Schmidt, T. C.; Wilson, J. T. *A Guide for Assessing Biodegradation and Source Identification of Organic Ground Water Contaminants using Compound Specific Isotope Analysis (CSIA)*; U.S. EPA: OK, 2008.

(40) International Standardization Organization. *Statistical Methods for Use in Proficiency Testing by Interlaboratory Comparisons; Text in German and English. DIN ISO 13528*; Berlin, 2009.