A comparison of isotope ratio mass spectrometry and cavity ring-down spectroscopy techniques for isotope analysis of fluid inclusion water

Stefan de Graaf\textsuperscript{1} | Hubert B. Vonhof\textsuperscript{1} | Therese Weissbach\textsuperscript{2} | Jasper A. Wassenburg\textsuperscript{1} | Elan J. Levy\textsuperscript{1} | Tobias Kluge\textsuperscript{2} | Gerald H. Haug\textsuperscript{1}

\textsuperscript{1}Climate Geochemistry Department, Max Planck Institute for Chemistry, Mainz, Germany
\textsuperscript{2}Ruprecht-Karls University Heidelberg, Institute of Environmental Physics, Heidelberg, Germany

**Rationale:** Online oxygen ($\delta^{18}$O) and hydrogen ($\delta^{2}$H) isotope analysis of fluid inclusion water entrapped in minerals is widely applied in paleo-fluid studies. In the state of the art of fluid inclusion isotope research, however, there is a scarcity of reported inter-technique comparisons to account for possible analytical offsets. Along with improving analytical precisions and sample size limitations, interlaboratory comparisons can lead to a more robust application of fluid inclusion isotope records.

**Methods:** Mineral samples—including speleothem, travertine, and vein material—were analyzed on two newly setup systems for fluid inclusion isotope analysis to provide an inter-platform comparison. One setup uses a crusher unit connected online to a continuous-flow pyrolysis furnace and an isotope ratio mass spectrometry (IRMS) instrument. In the other setup, a crusher unit is lined up with a cavity ring-down spectroscopy (CRDS) system, and water samples are analyzed on a continuous standard water background to achieve precisions on water injections better than 0.1\(\,^{\circ}\) for $\delta^{18}$O values and 0.4\(\,^{\circ}\) for $\delta^{2}$H values for amounts down to 0.2\(\,\mu\)L.

**Results:** Fluid inclusion isotope analyses on the IRMS setup have an average 1\(\sigma\) reproducibility of 0.4\(\,^{\circ}\) and 2.0\(\,^{\circ}\) for $\delta^{18}$O and $\delta^{2}$H values, respectively. The CRDS setup has a better 1\(\sigma\) reproducibility (0.3\(\,^{\circ}\) for $\delta^{18}$O values and 1.1\(\,^{\circ}\) for $\delta^{2}$H values) and also a more rapid sample throughput (<30 min per sample). Fluid inclusion isotope analyses are reproducible at these uncertainties for water amounts down to 0.1\(\,\mu\)L on both setups. Fluid inclusion isotope data show no systematic offsets between the setups.

**Conclusions:** The close match in fluid inclusion isotope results between the two setups demonstrates the high accuracy of the presented continuous-flow techniques for fluid inclusion isotope analysis. Ideally, experiments such as the one presented in this study will lead to further interlaboratory comparison efforts and the selection of suitable reference materials for fluid inclusion isotopes studies.
1 INTRODUCTION

Fluid inclusions in minerals constitute a unique archive of paleo-fluids in the geological record. For this reason, numerous techniques have been developed for analyzing fluid inclusions over the years for the study of paleo-fluid systems.1,2 Within the wide array of techniques, oxygen (δ18O) and hydrogen (δD) isotope analysis of fluid inclusion water is of particular interest, as it may provide direct information on the origin and type of mineral-forming paleo-fluids. Indirect reconstructions of paleo-fluid δ18O values are commonly performed using the δ18O values of calcite or quartz but are subject to uncertainties related to nonequilibrium effects during calcite precipitation3,4 and general uncertainty concerning the temperature of the fluid. Direct isotope analysis of fluid inclusions thus significantly improves the reconstruction of paleo-fluid composition and is equally applicable to non-oxygen-bearing host minerals.

In speleothem research, fluid inclusion isotope records have been used in combination with U-Th dating to reconstruct rainfall δ18O values and associated climate change through time.5–12 Fluid inclusion isotope data of halite deposits have been used to reconstruct the isotope history of ocean water.13,14 When applied to vein-type deposits, fluid inclusion isotope data can be used to reconstruct basin-scale fluid flow circulation in the subsurface, which mainly finds an interest in ore geology,15–18 petroleum geology,19,20 and petrology.21

Isotope analysis of fluid inclusions is not straightforward, as inclusions are only microns to hundreds of microns in size. As a consequence, bulk analytical techniques have mostly been developed, in which the integral volume of a large number of fluid inclusions within a mineral sample is released and subsequently analyzed. The first techniques emerged in the 1970s and relied on water release through thermal decrepitation in an off-line preparation device and subsequent analysis on a dual-inlet mass spectrometer.22–25 In general, only hydrogen isotope ratios are usable, due to considerable uncertainties associated with measuring oxygen isotope ratios with these techniques.26–28 Thermal decrepitation techniques have been applied mainly on vein-type deposits because fluids involved in the deposition of hydrothermal (ore) minerals are typically characterized by large isotope variations.15,16,29–31

In the 2000s, fundamentally different techniques for fluid inclusion isotope analysis were developed that allow for analysis using continuous-flow isotope ratio mass spectrometry (CF-IRMS).32,33 Within these setups, the spectrometer is connected online to a mechanical crusher unit that is maintained at a relatively low temperature (120°C–130°C). A pyrolysis reactor within the line converts released water vapor into hydrogen and carbon monoxide gas, which are subsequently separated in a gas chromatographic column before analysis by IRMS. Reliable data for both oxygen and hydrogen isotope ratios can be acquired using IRMS techniques for sub-microliter amounts of fluid inclusion water, as demonstrated in various studies on speleothems.5,7,34 Furthermore, the technique has successfully been applied on hydrothermal vein mineralization to gain insight into subsurface fluid flow dynamics.18–20,35 As a more cost-efficient alternative, fluid inclusion isotope ratios may also be measured on an IRMS instrument using an off-line extraction method.36

Since the beginning of the 2010s, new online setups have been developed using cavity ring-down spectroscopy (CRDS), which was already an established technique for the accurate isotope analysis of water samples.37,38 An advantage of laser absorption spectrometry is its ability to measure δ18O and δD values of water vapor without the need to first split the water molecules into separate oxygen and hydrogen species. Laser spectroscopy techniques developed for fluid inclusion isotope analysis use crusher units similar to IRMS setups and run on a dry nitrogen carrier gas,39 on a “wet” carrier gas containing a constant standard water background,40,41 or under vacuum.21

For continuous-flow IRMS and CRDS setups, reported precisions for fluid inclusion δ18O analysis of mineral samples (δ18Ofi values) are usually around 0.5‰, whereas precisions between 1.5‰ and 2‰ are reported for fluid inclusion δD analysis (δDfi values). Such precisions can be achieved for water yields as low as 0.15–0.2 μL10,32 Mineral samples, however, may contain considerably less fluid inclusion water per gram—the typical upper capacity limit of most commonly used crushers. Lowering the minimum sample size required for fluid inclusion isotope analysis would, therefore, enable data acquisition of mineral samples with low water contents. Furthermore, increasing sample throughput rates—currently more than an hour per sample in most reported setups—would be helpful to increase data density in speleothem time series and facilitate more thorough replication in fluid inclusion isotope data sets.

Another point of attention in the state of the art of fluid inclusion isotope research is the scarcity of reported inter-technique comparisons to account for possible analytical offsets between laboratories. For instance, in Arlenzo et al.,39 a comparison of three speleothem samples shows that isotope ratios from a CRDS setup are lower by 0.7‰ for δ18Ofi values and 2.5‰ for δDfi values compared with an IRMS technique. Meckler et al.9 observed offsets toward lower δ18Ofi values of fluid inclusion water in speleothem samples in an IRMS technique than in a CRDS technique, whereas the δDfi values do reproduce. Systematic offsets of approximately 1‰ in δ18Ofi values were found in a speleothem sample set by Wainer et al.,7 comparing two IRMS techniques. Especially in speleothem-based paleoclimate research, where isotope variations can be small,42,43 such offsets in δ18O values may impact climate interpretations.

The previously cited speleothem studies demonstrate that interlaboratory differences may exist for fluid inclusion isotope analysis. For a more robust application of fluid inclusion isotope techniques in paleo-fluid studies, the field, therefore, needs more thorough interlaboratory comparison experiments, ideally based on reliable fluid inclusion-bearing standard materials available to all laboratories that use the techniques. This is particularly valid for ongoing discussion on potential diagenetic exchange of oxygen between fluid inclusion water and host calcite in speleothems.44,45 As long as the interlaboratory reproducibility of particularly δ18Ofi data is underdefined, it remains difficult to distinguish between diagenetic effects and possible analytical artifacts.
In this contribution, we present an extensive comparison study of two commonly used continuous-flow setups for online fluid inclusion isotope analysis, both available at the Max Planck Institute for Chemistry in Mainz, Germany. One setup uses a crusher unit and cryogenic trap connected to a pyrolysis furnace and an IRMS instrument (cf. Vonhof et al33). The second technique couples a crusher unit to a CRDS instrument and runs on humidified nitrogen carrier gas (cf. Affolter et al40). Several fluid inclusion-bearing mineral samples, including speleothem, travertine, and vein material, were run on both setups. In doing so, we can present a robust data comparison between the two main analytical techniques currently used in the field.

2 | ISOPOE RATIO MASS SPECTROMETRY SETUP

2.1 | Design of the line

The setup for fluid inclusion isotope analysis using an IRMS system is similar to that first presented by Vonhof et al33. It consists of (a) a preparation unit, (b) a continuous-flow pyrolysis furnace (high-temperature conversion elemental analyzer [TC-EA]; Thermo Scientific, Bremen, Germany), and (c) a continuous-flow isotope ratio mass spectrometer (Thermo Scientific Delta V). The three units are connected with 1/16 in. stainless steel capillaries (Figure 1). The flow rates of helium carrier gas through the line are maintained at 95 mL/min by the use of mass flow controllers.

The preparation unit consists of a crusher placed in an oven at 120°C. Water released within the crusher is vaporized and transported by He carrier gas to the TC-EA. A cryogenic trap is positioned in the line between the crusher and the TC-EA to collect water that is being released. The cryogenic trap consists of a 60 cm coiled 1/16 in. stainless steel capillaries, which is located outside the oven so that it can be cooled down by submersion into an ethanol-liquid nitrogen sludge at ~100 to ~90°C. At these temperatures, water vapor is effectively trapped, whereas contaminant species in the carrier gas and common inclusion gas phases such as CO2 or CH4 are flushed through. The cryogenic trap can be heated to ~150°C with a flash heater to generate a water pulse short enough to be isotopically analyzed. In the TC-EA pyrolysis reactor, water vapor is transformed into H2 and CO gas by reaction with glassy carbon at 1400°C.46 A gas chromatographic column at 65°C separates the H2 and CO gases before entry into the mass spectrometer. The gas flow into the mass spectrometer is controlled by a ConFlo IV Universal continuous-flow interface (Thermo Scientific). In the mass spectrometer, a rapid magnet peak jump between the analysis of H2 and CO gases allows for the acquisition of both hydrogen and oxygen isotope ratios from a single water sample (Figure 2).

Within the preparation unit, a bypass (through port D in Figure 1) maintains a continuous flow of He through the TC-EA reactor tube into the mass spectrometer when opening the crusher to load a sample. The design allows for flushing atmosphere from the crusher before starting the analysis.33 During the analysis, background intensities in the mass spectrometer are generally below 100 mV for m/z 28, 29, and 30 (CO), below 200 mV for m/z 2 (H2), and in the range of 40–80 mV for m/z 40 (Ar). For comparison, a water sample of 0.2 μL produces peak intensities of approximately 13 000 mV for m/z 29 and 6000 mV for m/z 2 (Figure 2).

The in-house built crusher (Figure 3) closely resembles the design presented by Plessen and Lüders.47 The body of the crusher is composed of stainless steel and is fitted with an aluminum bronze...
piston with a convex stainless steel tip that can be lowered onto the sample with a rotating movement. The bottom of the stainless steel cup (internal diameter: 22 mm; internal depth: 12.4–14.8 mm) that holds the sample matches the curvature of the piston, thereby achieving high crushing efficiency. For carbonate samples, typically a fine-grained residue with fragments <0.1 mm in size is left after crushing. Up to approximately 0.7 cm³ of material can be loaded at a time. O-rings consisting of fluorocarbon encapsulated in fluorinated ethylene propylene (FEP/FPM; Dichtelemente arcus GmbH, Seevetal, Germany) are used to create gas-tight seals between the different components of the crusher. A stainless steel septum port (septum type: 11 mm Thermogreen LB-2; Sigma-Aldrich, St Louis, MO, USA) enables standard water injections into the crusher unit. By injecting directly into the crusher chamber, evaporation of sample water occurs at the same spot for both standard water injections and sample crushes.

### 2.2 Analytical performance

Standard waters were analyzed by injection into the septum port of the crusher to monitor the stability and precision of the setup. A 0.5-μL Medical SGE syringe (model 0.5BNR-5/0.63; part number: 000300; Trajan Scientific, Ringwood, Australia) was used for standard water injections. Standard waters of variable isotope composition were used in this study (Table 1), all calibrated against international standards. Isotope ratios throughout this paper are reported on the VSMOW scale. A series of standard water injections in the size range of 0.1–0.5 μL into the crusher on the IRMS setup display a standard deviation better than 0.1‰ for δ¹⁸O values and 1.0‰ for δ²H values. The analytical uncertainty on fluid inclusion isotope ratios, however, is considerably higher, as the analysis of mineral samples involves a more complex analytical...
the linearity is calculated using Equations 1 and 2 for needed to reach stable values. For the series of standards in Table 2, four consecutive measurements of the same standard water are become evident when analyzing isotopically different standard waters standard water.

The setup exhibits both a memory effect and linearity, which size effects in the direct comparison of the sample with the water yield of the sample crush. This is to minimize possible standard water injections following the sample crush are tailored to water injections and sample crushes (usually 4 min). Volumes of bracketing standard water measurements that are run directly before and after the crush. Cold-trap times are kept equal for both standard water injections and sample crushes (usually 4 min). Volumes of standard water injections following the sample crush are tailored to the water yield of the sample crush. This is to minimize possible size effects in the direct comparison of the sample with the standard water.

The setup exhibits both a memory effect and linearity, which become evident when analyzing isotopically different standard waters (Table 2). Due to the sample-to-sample memory effect, about three to four consecutive measurements of the same standard water are needed to reach stable values. For the series of standards in Table 2, the linearity is calculated using Equations 1 and 2 for $\delta^2H$ and $\delta^{18}O$ values, respectively, with terms in the numerator referring to true isotope ratios of the standard waters. The memory effect is determined in Equations 3 and 4 for $\delta^2H$ and $\delta^{18}O$ values, respectively. Uppercase letters in Equations 1–4 refer to superscripts in Table 2.

| Standard      | $\delta^{18}O$ value % vs VSMOW | $\delta^2H$ value % vs VSMOW |
|---------------|---------------------------------|-----------------------------|
| Turkana       | 6.96                            | 45.81                       |
| VSMOW         | 0.00                            | 0.00                        |
| Mainz-Kona mix| −5.95                           | −43.37                      |
| Mainz tap     | −8.70                           | −62.96                      |
| Grimsel       | −9.33                           | −84.73                      |
| Chamonix      | −13.68                          | −101.59                     |
| Huagapo       | −14.11                          | −108.58                     |
| SLAP          | −55.50                          | −427.51                     |

SLAP, Standard Light Antarctic Precipitation; VSMOW, Vienna Standard Mean Ocean Water.

Prior to analysis of a mineral sample, a standard water is repetitively measured to condition the analytical line. The mineral sample in the crushing cell is crushed and measured as soon as the mass spectrometer is recording stable values for the injected standard water. Direct calibration of fluid inclusion isotope ratios is performed by comparing recorded isotope ratios of the sample crush with bracketing standard water measurements that are run directly before and after the crush. Cold-trap times are kept equal for both standard water injections and sample crushes (usually 4 min). Volumes of standard water injections following the sample crush are tailored to the water yield of the sample crush. This is to minimize possible size effects in the direct comparison of the sample with the standard water.

Analytical procedure and data calculation

When switching to another standard water, isotope ratios display a memory effect, stabilizing again after three to four injections. Area $H_2$ and area $CO$ are measures of the peak size (i.e., water amount). Superscript uppercase letters refer to numbers used in Equations 1–4. IRMS, isotope ratio mass spectrometry.

Equations 5 and 6 are used for the final calculation of true sample crush $\delta^2H_{\text{crush}}$ and $\delta^{18}O_{\text{crush}}$ values, respectively. The equations include a correction for both memory effect and linearity. The subscript "crush" refers to the isotope results of the sample crush. The subscript "STD" refers to a standard water measurement of the approximately same amount as the water yield of the crush, and the subscript "true" to the true isotope ratios of this standard water:

$$\delta^2H_{\text{crush}} = (\delta^2H_{\text{crush}} - \delta^2H_{\text{STD}}) \cdot m_{\text{STD}} \cdot l_{\text{crush}} + \delta^2H_{\text{STD true}}.$$  (5)

$$\delta^{18}O_{\text{crush}} = (\delta^{18}O_{\text{crush}} - \delta^{18}O_{\text{STD}}) \cdot m_{\text{STD}} \cdot l_{\text{crush}} + \delta^{18}O_{\text{STD true}}.$$  (6)

Calibrations presented in Table 2 are performed on a weekly basis to account for drift in the memory effect and linearity. Typically, $l_{\text{crush}}$
and $l_O$ are between 0.98 and 1.13, while $m_H$ and $m_O$ are around 1.16 and 1.08, respectively. To minimize uncertainties related to the correction of these effects, a standard water that is expected to be isotopically close to the fluid inclusion water of the sample is used during analysis.

2.4 Fluid inclusion isotope results

The analytical performance of the IRMS setup was determined by running a set of mineral samples following the protocol outlined earlier. Sample material includes a fluorite vein, travertine material, and speleothems from tropical, temperate, and high-altitude settings (Table 3). The sample crushes liberated 0.08–0.44 μL of fluid inclusion water. Fluid inclusion isotope data of the speleothem and travertine samples together cover a wide isotope range along the global meteoric water line (GMWL48; Figure 4), with each sample exhibiting relatively homogeneous isotope ratios (Table 4). The average 1σ reproducibility of these samples is 0.4% for $\delta^{18}O_{\text{fi}}$ values and 2.0‰ for $\delta^{2}H_{\text{fi}}$ values. The fluorite vein sample FL-HZ has the highest fluid inclusion isotope ratios and plots to the right of the GMWL.

| Name     | Location          | Type                  | Age          | Drip water $\delta^{18}O$ value | Drip water $\delta^{2}H$ value |
|----------|-------------------|-----------------------|--------------|---------------------------------|-------------------------------|
| Huagapo-7| Andes Mts. (Peru) | Stalagmite (calcite)  | Late Holocene| $-14.11$                       | $-108.58$                     |
| HV-SA-11 | Saudi Arabia      | Stalagmite (calcite)  | Pleistocene  |                                 |                               |
| Scladina | Ardennes (Belgium)| Stalagmite (calcite)  | Early Holocene| $-6.5$                         | $-42.4$                       |
| Semproniano | Central Italy   | Fissure ridge travertine (calcite) | Pleistocene |                                 |                               |
| La Vièrge | Rodrigues (Mauritius) | Stalagmite (calcite) | Late Holocene| $-3.3$                         | $-14.9$                       |
| FL-HZ    | Harz Mts. (Germany)| Fluorite vein         | Mesozoic     |                                 |                               |

$^a$Only measured on the cavity ring-down spectroscopy setup.

FIGURE 4 Fluid inclusion $\delta^{18}O_{\text{fi}}$ and $\delta^{2}H_{\text{fi}}$ values of mineral crushes on the isotope ratio mass spectrometry setup, with the speleothem and travertine samples plotting along the global meteoric water line (GMWL). The fluorite sample FL-HZ did not form in meteoric water and therefore plots off the GMWL. [Color figure can be viewed at wileyonlinelibrary.com]
values of $-9.0\%$ (1σ of 0.09%) and $\delta^{2}H$ values of $-65.1\%$ (1σ of 0.33%).

The crusher used in this setup is the same as in the IRMS setup (Figure 3). Between the homogenization volume and the crusher device, a backflow preventer (Swagelok, Solon, OH, USA; SS-2C-1/3) is installed to prevent sample water vapor backflow into the purge capillary upon crushing. A 7 μm pore size filter (Swagelok SS-2TF-7) at the outflow of the crusher ensures that mineral particles that are released during crushing do not disperse downstream. The last element in the preparation unit is a stainless steel volume of 40 cm³ (Swagelok 304L-HDF2-40) that serves to smooth out the entry of sample water into the CRDS instrument over a wider time interval, which facilitates precise data analysis and calculations.

### 3.2 | Data processing

Water can be released in the crusher either through sample crushing or through water injection in the septum port. Upon water liberation in the crusher, peaks are generated on top of the stable water background. The Picarro L2140-i analyzer records values every 0.7 s for water content and $\delta^{18}O$ and $\delta^{2}H$ values. Raw data files are processed in a Python script to compute isotope ratios of the sample peaks, (b) weighted average isotope ratios of the sample peaks, and (c) water background concentration and isotope ratios.

In the calculations, the background is subtracted from the sample peaks to acquire isotope ratios of the samples. For the background values of each peak, an average is taken over a 1 min time interval before and a 3 min interval after the peak to account for any drift in the backgrounds. Sample peaks are characterized by a sharp increase followed by a more gradual decrease toward background levels. The script analyzes the slope of the water vapor concentration to determine the peak starts and ends (Figure 6). Peak starts are defined when the slope exceeds 10 ppmv/s, and peak ends are subsequently set when the down-going slope exceeds $-0.5$ ppmv/s. Parameter values in the calculations were empirically established aiming for the highest precision on standard water measurements. The total analytical time required for the analysis of a single water sample is 8–10 min depending on the water amount.

### 3.3 | Analytical performance

The performance of the CRDS setup was monitored by measuring standard waters (Table 1), which were injected directly into the crusher with a 0.5 μL micro-syringe (SGE Analytical Science—model BNR-5/0.63). A total of 355 standard water measurements

| TABLE 4 | Fluid inclusion isotope results on the IRMS setup |
|----------|--------------------------------------------------|
| Weight (g) | Water yield (μL) | $\delta^{18}O$ value (%) vs VSMOW | $\delta^{2}H$ value (%) vs VSMOW |
|---------------|-------------------|-----------------|-----------------|
| **Huagapo-7 (stalagmite)** | | | |
| 0.29 | 0.25 | −16.6 | −122.9 |
| 0.28 | 0.30 | −16.9 | −120.7 |
| 0.36 | 0.38 | −16.6 | −128.2 |
| 0.28 | 0.34 | −16.7 | −121.4 |
| | SD: 0.17 | SD: 3.38 |
| **Scladina (stalagmite)** | | | |
| 0.44 | 0.12 | −6.9 | −46.7 |
| 0.39 | 0.26 | −6.9 | −50.2 |
| 0.45 | 0.25 | −5.9 | −48.2 |
| 0.46 | 0.26 | −7.4 | −50.0 |
| | SD: 0.64 | SD: 1.67 |
| **La Vierge (stalagmite)** | | | |
| 0.80 | 0.20 | −2.5 | −12.2 |
| 0.76 | 0.14 | −3.1 | −10.4 |
| 0.82 | 0.08 | −2.2 | −7.3 |
| 0.86 | 0.13 | −2.8 | −11.8 |
| | SD: 0.41 | SD: 2.22 |
| **Semproniano (travertine)** | | | |
| 0.29 | 0.44 | −4.6 | −33.9 |
| 0.27 | 0.24 | −4.2 | −32.2 |
| 0.36 | 0.31 | −4.9 | −34.1 |
| 0.37 | 0.32 | −4.8 | −33.4 |
| | SD: 0.32 | SD: 0.85 |
| **FL-HZ (fluorite vein)** | | | |
| 0.53 | 0.40 | 2.7 | −5.3 |
| 0.49 | 0.29 | 3.3 | 0.8 |
| 0.46 | 0.26 | 2.9 | −0.1 |
| | SD: 0.28 | SD: 3.28 |

Water yields are estimated by comparison with bracketing standard water injections. IRMS, isotope ratio mass spectrometry; SD, standard deviation.
demonstrates that isotope analyses on the CRDS setup need to be corrected for linearity and sample size effects. Memory effects in this setup are absent (Table 5). The linearity of the instrument was established through injections of different standard waters of known isotope composition (Figure 7).

Injected amounts display a linear relation to integrated peak areas of the water vapor concentration (Figure 8), allowing for accurate monitoring of size effects. Size effects exist for both $\delta^{18}$O and $\delta^{2}$H values, becoming especially evident for water amounts below 0.1 μL (Figure 9). The size effect on $\delta^{18}$O values largely follows a logarithmic decrease; the steepness of the trend slightly decreases with increasing isotope ratios of the standard water (Figure S1, supporting information). The size effect on $\delta^{2}$H values depends more strongly on the isotope difference between the sample and the background. Toward lower water amounts, $\delta^{2}$H values tend to move toward the background values. Being isotopically similar to the background water, Mainz tap standard water consequently lacks a size effect on $\delta^{2}$H values. It is noteworthy that because a 0.5-μL micro-syringe was used, the standard water measurements for the amounts above 0.5 μL represent two rapid consecutive injections. Although this leads to a slightly different peak shape, it does not affect data quality.

A 7-point running standard deviation over the standard water measurements indicates standard deviations below 0.2‰ and 0.7‰ for $\delta^{18}$O and $\delta^{2}$H values, respectively, at amounts above 0.1 μL (Figure 9). For standard water injections above 0.2 μL, the standard deviations drop even further, generally remaining below 0.1‰ for $\delta^{18}$O values and 0.4‰ for $\delta^{2}$H values. Precisions are independent of the isotope ratios of the standard water for the isotope range covered in Figure 9. The data of standard water injections were collected over a month, without any detectable drift in the linearity and size effects.

3.4 Fluid inclusion isotope results

For isotope analysis of fluid inclusion water, mineral samples are crushed to analyze the liberated water. Opening and closing the crusher to load a sample leads to instability in the water background, which requires a 10–15 min waiting time for restabilization (Figure 6). During the stabilization period, atmospheric moisture and water adsorbed onto the sample are flushed out of the system. Because the subsequent crush and isotope analysis take another 10 min, the total time to load and analyze one mineral sample is 25–30 min.

First, a size effect correction is applied to isotope ratios of sample crushes using the size-dependent logarithmic trends shown in Figure 9. The size effect correction takes into account the dependence of the logarithmic trends on water isotope ratios, which is especially evident for $\delta^{2}$H values. Second, sample crushes are corrected for linearity as established from standard water injections (Figure 7). The corrections are implemented in the Python script. Precise calculations behind the corrections are explained in more detail in the supporting information (S1).

Fluid inclusion isotope data were collected on the CRDS setup for the same samples as were analyzed on the IRMS setup (Table 3). Water yields of the crushes range from 0.11 to 0.70 μL, which is similar to the water yields on the IRMS setup. The data of the speleothem and travertine samples fall close to the GMWL and have
an average 1σ reproducibility of 0.3‰ for $\delta^{18}$O$_{fi}$ values and 1.1‰ for $\delta^2$H$_{fi}$ values, excluding the speleothem sample HV-SA-11, which shows more variable isotope signatures over a $\delta^{18}$O$_{fi}$ range of $-11.6‰$ to $-8.7‰$ along the GMWL (Figure 10; Table 6). The fluorite vein sample exhibits the highest isotope ratios and has a standard deviation of 0.3‰ for $\delta^{18}$O$_{fi}$ values and 2.1‰ for $\delta^2$H$_{fi}$ values.

### 4 | DISCUSSION

#### 4.1 | Fluid inclusion isotope results

Fluid inclusion isotope data were acquired on the CRDS and IRMS analytical lines using the same mineral samples, and thus a direct comparison can be made between the setups. Isotope ratios of the samples span 20‰ for $\delta^{18}$O$_{fi}$ values and 130‰ for $\delta^2$H$_{fi}$ values across a range relevant for most naturally occurring water.

On both analytical setups, the fluid inclusion isotope signatures of the speleothem and travertine samples plot close to the GMWL.
which is in accordance with the expected meteoric signature of the mineral-forming fluids of these samples. Fluid inclusion isotope data of the La Vierge sample from the island of Rodrigues in the Indian Ocean are similar to present-day rainfall in the region and lie close to unpublished drip water isotope data (Table 3). The Huagapo-7 sample formed during the Late Holocene at an elevation of 3850 m above sea level in the Andes Mountains, where present-day rainfall has comparatively low isotope ratios (Table 3). The Holocene Scladina stalagmite sample from The Meuse valley in Belgium also closely matches present-day drip water (Table 3). The higher isotope variability of sample HV-SA-11 from Saudi Arabia is probably related to internal heterogeneities in the fluid inclusion water, as the subsamples were not taken from a single growth layer of this stalagmite. The water content of the HV-SA-11 subsamples is also considerably more variable than that of the other samples (Table 6). We have no drip water of the now-dry cave, but the $\delta^{18}O$ range of −11.6 to −8.7‰ is comparable with that of present-day drip water in other caves on the Arabian Peninsula.

4.2 Inter-technique comparison

A direct comparison of fluid inclusion isotope data acquired on both setups shows no systematic offsets for either $\delta^{18}O$ or $\delta^2H$ values (Figure 11). For the five samples run on both setups, the average inter-platform difference is only 0.16‰ for $\delta^{18}O$ values and 0.78‰ for $\delta^2H$ values (Table 7), which is well within the analytical uncertainties. The data plot on a 1:1 line, demonstrating that both continuous-flow techniques for online isotope analysis of fluid inclusion water are able to produce accurate data. Minor instabilities that may appear in the background levels during analysis on the CRDS setup (Figure 6) seem not to affect data accuracy, as the calculation protocol takes into account drift in background values.

Another comparison experiment that was recently performed between a CRDS setup at Heidelberg University and the IRMS setup in Mainz shows an equally good inter-platform reproducibility. The CRDS setup in Heidelberg is similar to that in Mainz and differs only in the use of a single oven, a different crushing mechanism and a larger volume after the crusher of 400 cm$^3$. The comparison study of Weissbach also used material from the Scladina and Huagapo stalagmites. However, it is difficult to make a direct comparison with our study, as the samples were taken from a different part of the stalagmites. Nonetheless, the average values of the Huagapo stalagmite (−16.9‰ for $\delta^{18}O$ values and −124.4‰ for $\delta^2H$ values) are strikingly similar to those that we determined for the Huagapo sample (Table 7).
Regarding analytical uncertainties on the carbonate sample crushes \((n = 16\) and \(n = 38\) for IRMS and CRDS setup, respectively), both systems in Mainz have a similar reproducibility for δ\(^{18}\)O values at around 0.3–0.4‰ (1σ). For δ\(^{2}\)H values, the CRDS setup performs better with a 1σ uncertainty of 1.1‰ compared with 2.0‰ on the IRMS setup. For both setups, it is evident that sample crushes show higher standard deviations than standard water injections, which on the CRDS analyzer have standard deviations below 0.2‰ for δ\(^{18}\)O and 0.7‰ for δ\(^{2}\)H values, respectively. The larger uncertainty for crushed samples could be due to internal heterogeneities in their fluid inclusion content. Alternatively, the process of crushing may cause a variety of minor interferences that add to the uncertainty of the isotope analysis. For a better constraint on precisions of sample crushes, the field needs widely available reference material; this would also facilitate future interlaboratory comparison efforts. In any case, the precisions achievable on both setups in the experiments presented here are sufficient to discern relatively subtle climate signals characteristic of speleothem records.

4.3 | Pushing the limit: Analysis of water amounts < 0.1 μL

Analyses are reproducible on both setups for amounts reaching down to 0.1 μL. For sample material with low fluid inclusion densities, optimizing the analytical performance at low water yields is important, as it may bring samples in reach that were previously not suitable for fluid inclusion isotope analysis. The relatively good performance of the CRDS setup in Mainz for low water yields can be explained by...
| Weight (g) | Water yield (μL) | $\delta^{18}$O value (% vs VSMOW) | $\delta^{2}$H value (% vs VSMOW) |
|-----------|-----------------|----------------------------------|---------------------------------|
| Huagapo-7 (stalagmite) | | | |
| 0.42 | 0.39 | −16.8 | −124.3 |
| 0.56 | 0.70 | −16.9 | −124.9 |
| 0.49 | 0.38 | −16.5 | −125.2 |
| 0.27 | 0.21 | −16.3 | −124.8 |
| 0.28 | 0.22 | −16.8 | −126.2 |
| 0.22 | 0.21 | −17.3 | −128.7 |
| 0.38 | 0.37 | −16.7 | −126.3 |
| 0.39 | 0.33 | −16.5 | −125.6 |
| 0.32 | 0.37 | −16.6 | −125.4 |
| 0.35 | 0.28 | −16.9 | −125.2 |
| | | SD: 0.28 | SD: 1.23 |
| HV-SA-11 (stalagmite) | | | |
| 0.67 | 0.67 | −9.8 | −73.0 |
| 0.36 | 0.52 | −8.7 | −65.1 |
| 0.31 | 0.16 | −9.0 | −62.2 |
| 0.31 | 0.20 | −11.6 | −83.5 |
| 0.37 | 0.27 | −11.2 | −84.1 |
| 0.42 | 0.23 | −10.6 | −77.4 |
| 0.66 | 0.18 | −9.8 | −65.3 |
| 0.59 | 0.27 | −11.3 | −81.3 |
| | | SD: 0.31 | SD: 1.06 |
| Scladina (stalagmite) | | | |
| 0.63 | 0.24 | −6.7 | −47.3 |
| 0.47 | 0.22 | −6.9 | −46.2 |
| 0.38 | 0.12 | −6.7 | −48.3 |
| 0.47 | 0.21 | −6.5 | −46.8 |
| 0.64 | 0.15 | −7.2 | −48.3 |
| 0.69 | 0.27 | −6.8 | −46.8 |
| 0.69 | 0.19 | −7.2 | −48.6 |
| 0.72 | 0.30 | −6.2 | −45.5 |
| 0.50 | 0.42 | −7.0 | −46.6 |
| | | SD: 0.37 | SD: 1.63 |
| La Vierge (stalagmite) | | | |
| 0.73 | 0.23 | −2.6 | −11.6 |
| 0.78 | 0.24 | −2.8 | −12.4 |
| 0.62 | 0.11 | −2.5 | −10.3 |
| 1.09 | 0.16 | −3.2 | −13.9 |
| 0.88 | 0.14 | −2.8 | −8.6 |
| 0.92 | 0.14 | −3.1 | −11.2 |
| 0.73 | 0.22 | −2.1 | −11.8 |
| 0.74 | 0.24 | −3.1 | −12.9 |
| | | SD: 0.37 | SD: 1.63 |
| Semproniano (travertine) | | | |
| 0.60 | 0.40 | −5.6 | −33.9 |
| 0.86 | 0.50 | −5.0 | −35.0 |
| 0.50 | 0.44 | −4.8 | −35.0 |
1. The Picarro analyzer used in this study (L2140-i) provides data at a high frequency (every 0.7 s), compared with a 2 s time period in Dassié et al\(^41\) (L2120-i) and a 12 s time period in Affolter et al\(^40\) (L1102-i).

2. The good performance for small sample sizes could be related to the improved temperature control of the parts sensitive to temperature drops (i.e., mixing volume and T-piece connector into which background water is supplied). Because these parts are heated in a separate oven in our setup, the backgrounds are less disturbed during the crushing procedure. Background disturbances have an increasingly negative effect as samples become smaller.

3. The extensive series of standard water isotope analyses on the CRDS setup show that size effects are systematic for both δ\(^{18}\)O and δ\(^{2}\)H values and can thus be corrected for (Figure S2, supporting information). The success of these size corrections is also demonstrated by the independence of acquired isotope ratios of the water yields of the sample crushes, which range from 0.11 to 0.70 μL (Table 6).

Whereas the more complex analytical protocol on the IRMS setup impairs reliable data calculation for sample crushes yielding water amounts below 0.1 μL, the systematic behavior of the size effect on the CRDS setup potentially opens up the possibility of analysis of such small amounts. Even standard water injections down to 0.03 μL on the CRDS setup follow the size-dependent trends, albeit at higher uncertainties, with 1σ uncertainties running up to 0.5‰ for δ\(^{18}\)O values and 1.4‰ for δ\(^{2}\)H values (Figure 9). Nonetheless, these uncertainties would still be sufficient for studies on basinal fluid systems, which are generally characterized by large isotope variations, commonly exceeding 10‰ in δ\(^{18}\)O values and even more in δ\(^{2}\)H values.\(^54\) Further detailed testing would be needed to explore the applicability of the CRDS setup for samples with water yields under the 0.1 μL threshold. This could include determining the optimal concentration of background water vapor, which was set to a default of 14,000 ppmv in all our experiments.

**TABLE 6** (Continued)

| Weight (g) | Water yield (μL) | δ\(^{18}\)O value (‰ vs VSMOW) | δ\(^{2}\)H value (‰ vs VSMOW) |
|------------|------------------|-------------------------------|-------------------------------|
| 0.88       | 0.66             | −4.8                          | −34.4                         |
| 0.37       | 0.34             | −4.6                          | −33.7                         |
| 0.30       | 0.28             | −4.8                          | −34.4                         |
| 0.35       | 0.33             | −4.8                          | −34.8                         |
| 0.35       | 0.35             | −4.9                          | −32.5                         |
| 0.32       | 0.33             | −5.6                          | −34.1                         |
| 0.44       | 0.48             | −4.8                          | −34.0                         |
| 0.36       | 0.24             | −4.8                          | −33.2                         |
|            |                  | SD: 0.35                      | SD: 0.76                      |

FL-HZ (fluorite vein)

| Weight (g) | Water yield (μL) | δ\(^{18}\)O value (‰ vs VSMOW) | δ\(^{2}\)H value (‰ vs VSMOW) |
|------------|------------------|-------------------------------|-------------------------------|
| 0.53       | 0.17             | 3.1                           | 0.1                           |
| 0.43       | 0.19             | 2.9                           | −2.3                          |
| 0.48       | 0.26             | 2.4                           | −5.4                          |
| 0.49       | 0.28             | 2.6                           | −2.6                          |
| 0.44       | 0.30             | 2.4                           | −4.2                          |
|            |                  | SD: 0.30                      | SD: 2.09                      |

Standard deviations for HV-SA-11 are not given as this sample has an isotopically heterogeneous fluid inclusion content.

CRDS, cavity ring-down spectroscopy; SD, standard deviation.

**FIGURE 11** Comparison of fluid inclusion isotope data acquired on the cavity ring-down spectroscopy and isotope ratio mass spectrometry setups for δ\(^{2}\)Hfi (left) and δ\(^{18}\)Ofi (right) values.

The data of Huagapo-7, Scladina, La Vièrge, Semproniano, and FL-HZ are included along with 1σ error bars. The data are reproducible between the two setups and show no systematic offsets for either δ\(^{2}\)Hfi or δ\(^{18}\)Ofi values.
4.4 | IRMS versus CRDS techniques

Although the size limitation and δ18Oi precision on both setups are similar, an important advantage of the CRDS setup is that the background water constantly going through the system removes sample-to-sample memory effects, which are known to occur in these analyzers when running them on a dry carrier gas. Although the memory effect in the IRMS setup can be monitored and corrected for as presented in this study, it remains a calculation step that adds to the final analytical uncertainty. A second advantage of the absence of memory effects is that the full procedure for analyzing a mineral sample on the CRDS setup takes less than 30 min because standard water bracketing is not necessary. That is considerably less time than the 1.5–2 h needed on the IRMS setup, where standard water bracketing is indispensable. This currently means that up to 16 samples can be analyzed in an 8 h working day on the CRDS system, in comparison with five samples on the IRMS system.

The potential downside of the use of a water background in the CRDS system is the inherent sensitivity to slight pressure changes in the line. Therefore, the crusher design is of key importance for a functional CRDS setup. Several tests with more powerful compressed air crushers resulted in instabilities in the water background, which compromised the analytical performance. Our experiments showed that in addition to the Potsdam crusher type used in this experiment (Figure 3), an Amsterdam crusher type (using a piston driven by compressed air) can yield reproducible results, but only if the crushing action is performed in a single stroke, as opposed to multiple repetitive strokes for a finer crush residue. The IRMS setup is less sensitive to the crusher type owing to the dry carrier gas and cryogenic trapping procedure and may, therefore, be preferred if repetitive crushing is required to acquire higher water yields.

A second potential issue on the CRDS setup could be the release of contaminant compounds during the crushing procedure. For instance, volatile organic compounds can negatively affect isotope analysis on CRDS instruments. Furthermore, inclusions in minerals forming in hydrothermal settings may contain gas phases such as CO2, CH4, or H2S. In the IRMS setup, such gas phases are flushed through during cold-trapping and do not interfere with the measurement, but this is not the case in the procedure followed for the CRDS analyzer. The reproducibility between the setups as presented in this study indicates no influence of such interferences for the samples used here. Nevertheless, the application of the CRDS setup for analyzing organic-rich samples and samples containing multiphase fluid inclusions may require cautious monitoring of such potential interferences.

5 | CONCLUSIONS

Two continuous-flow setups for online analysis of fluid inclusion δ18O and δ2H values are compared. The first setup uses a crushing cell with cold-trap that is connected to a TC-EA pyrolysis furnace and an IRMS instrument, and the second consists of a crushing cell coupled to a CRDS analyzer and runs on a moisturized carrier gas. For the CRDS system, the use of two separate ovens and a large mixing volume limits water background variations to a minimum (<20 ppmv) and therewith improves the analytical precision. Based on standard water injections, precisions on the IRMS setup are better than 0.1‰ for δ18O values and 1.0‰ for δ2H values, but significant memory effects must be corrected for. The CRDS setup has water injection precisions generally better than 0.1‰ for δ18O values and 0.4‰ for δ2H values (1σ) for amounts down to 0.2 μL and displays no memory effects.

We revised calculation protocols for both setups taking into account linearity and sample size effects and—for the IRMS setup—memory effects. Mineral samples of variable fluid inclusion isotope composition were crushed on both setups. For fluid inclusion water yields down to 0.1 μL, the δ18Oi values of the samples are reproducible at 0.3–0.4‰ (1σ) on both techniques. The CRDS setup outperforms the IRMS setup for δ2H values (1σ precision of 1.1‰ vs. 2.0‰). Another clear advantage of the CRDS technique is that due to the lack of sample-to-sample memory effects, it has a considerably more rapid sample throughput (<30 min per sample crush) than the IRMS technique. Furthermore, the CRDS setup holds the potential to achieve high precisions at amounts even below 0.1 μL without compromising the rapid sample throughput.

The δ18Oi and δ2H values of both the IRMS and the CRDS techniques correspond to the expected values of the samples. The inter-technique comparison demonstrates that there are no significant analytical offsets between the setups, which is in line with earlier observations of Meckler et al. for δ2H values. This shows that the IRMS and the CRDS techniques are both able to produce accurate fluid inclusion isotope data. Ideally, the experiments presented here...
ACKNOWLEDGEMENTS

Michael Welling and Florian Rubach are gratefully acknowledged for providing invaluable technical support around the design and assembly of the setups. The authors thank four anonymous reviewers for their comments and suggestions which helped to improve the manuscript.

REFERENCES

1. Roedder E. Fluid Inclusions. Mineralogical Society of America: Reviews in Mineralogy v. 12. 1984. https://doi.org/10.1555/9781510508271
2. Roedder E. Fluid inclusion analysis — Prologue and epilogue. Geochim Cosmochim Acta. 1990;54(3):495-507. https://doi.org/10.1016/0016-7037(90)90347-N
3. Daëron M, Drysdale RN, Peral M, et al. Most earth-surface calcites precipitate out of isotopic equilibrium. Nat Commun. 2019;10(1):1-7. https://doi.org/10.1038/s41467-019-08336-5
4. Watkins JM, Hunt JD, Ryerson FJ, DePaolo DJ. The influence of temperature, pH, and growth rate on the $\delta^{18}O$ composition of inorganically precipitated calcite. Earth Planet Sci Lett. 2014;404:332-343. https://doi.org/10.1016/j.epsl.2014.07.036
5. Flettman D, Burns SJ, Neff U, Mangini A, Matter A. Changing moisture sources over the last 330,000 years in northern Oman from fluid-inclusion evidence in speleothems. Quat Res. 2003;60(2):223-232. https://doi.org/10.1016/S0033-5894(03)00086-3
6. Van Breukelen MR, Vonhof HB, Hellstrom JC, Wester WCG, Kroon D. Fossil dripwater in stalagmites reveals Holocene temperature and rainfall variation in Amazonia. Earth Planet Sci Lett. 2008;275(1-2):54-60. https://doi.org/10.1016/j.epsl.2008.07.060
7. Wainer K, Genty D, Blamart D, et al. Speleothem record of the last 180 ka in Villars cave (SW France): Investigation of a large $\delta^{18}O$ shift between MIS6 and MIS5. Quaternary Sci Rev. 2011;301(2-3):130-146. https://doi.org/10.1016/j.quascirev.2010.07.004
8. Avienzo MM, Swart PK, Pourmand A, et al. Bahamian speleothem temperature reveals decrease associated with Heinrich stadials. Earth Planet Sci Lett. 2015;430:377-386. https://doi.org/10.1016/j.epsl.2015.08.035
9. Meckler AN, Affolter S, Dublysnyk YV, et al. Glacial–interglacial temperature change in the tropical West Pacific: A comparison of stalagmite-based paleo-thermometers. Quaternary Sci Rev. 2015;127:90-116. https://doi.org/10.1016/j.quascirev.2015.06.015
10. Uemura R, Nakamoto M, Asami R, et al. Precise oxygen and hydrogen isotope determination in nanoliter quantities of speleothem inclusion water by cavity ring-down spectroscopic techniques. Geochim Cosmochim Acta. 2016;172:159-176. https://doi.org/10.1016/j.gca.2015.09.017
11. Affolter S, Häuselmann A, Flettman D, Edwards RL, Cheng H, Leuenberger M. Central Europe temperature constrained by speleothem fluid inclusion water isotopes over the past 14,000 years. Sci Adv. 2019;5(6):1-9, eaav3809. https://doi.org/10.1126/sciadv.aav3809
12. Demény A, Kern Z, Németh A, et al. North Atlantic influences on climate conditions in east-Central Europe in the late Holocene reflected by flowstone compositions. Quat Int. 2019;512:99-112. https://doi.org/10.1016/j.quaint.2019.02.014
13. Knauth LP, Beeunas MA. Isotope geochemistry of fluid inclusions in Permian halite with implications for the isotopic history of ocean water and the origin of saline formation waters. Geochim Cosmochim Acta. 1986;50(3):419-433. https://doi.org/10.1016/0016-7037(86)90195-X
14. Lécuyer C, Fourel F, Blaney M, Brand U, Fralick P. $\delta^2$H of water from fluid inclusions in Proterozoic halite: Evidence for a deuterium-depleted hydrosphere? Chem Geol. 2020;541:1-11. https://doi.org/10.1016/j.chemgeo.2020.119583
15. Baatarsoogt B, Schwinn G, Wagner T, Taubald H, Beitter T, Markl G. Contrasting paleofluid systems in the continental basement: A fluid inclusion and stable isotope study of hydrothermal vein mineralization, Schwarzwald district, Germany. Geofluids. 2007;7(2):123-147. https://doi.org/10.1111/j.1468-8123.2007.00169.x
16. Wilkinson JJ. A review of fluid inclusion constraints on mineralization in the Irish ore field and implications for the genesis of sediment-hosted Zn-Pb deposits. Econ Geol. 2010;105(2):417-442. https://doi.org/10.2113/gsecongeo.105.2.417
17. Richard A, Boulvais P, Mercadier J, et al. From evaporated seawater to uranium-mineralizing brines: Isotopic and trace element study of quartz–dolomite veins in the Athabasca system. Geochim Cosmochim Acta. 2013;113:38-59. https://doi.org/10.1016/j.gca.2013.03.009
18. De Graaf S, Lüders V, Banks DA, et al. Fluid evolution and ore deposition in the Harz Mountains revisited: Isotope and crush-leach analyses of fluid inclusions. Miner Deposita. 2020;55(1):47-62. https://doi.org/10.1007/s00126-019-00880-w
19. De Graaf S, Nooitgedacht CW, Le Goff J, Van der Lubbe JHJL, Vonhof HB, Reijmer JJG. Fluid-flow evolution in the Albanide fold-thrust belt: Insights from hydrogen and oxygen isotope ratios of fluid inclusions. AAPG Bull. 2019;103(10):2421-2445. https://doi.org/10.1306/20151918034
20. De Graaf S, Reijmer JJG, Bertotti GV, et al. Fracturing and calcite cementation controlling fluid flow in the shallow-water carbonates of the Jandaíra formation, Brazil. Mar Petrol Geol. 2017;80:382-393. https://doi.org/10.1016/j.marpetgeo.2016.12.014
21. Czuppon G, Ramsay RR, Özgenc I, et al. Stable (H, O, C) and noble-gas (He and Ar) isotopic compositions from calcite and fluorite in the Speewah dome, Kimberley region, Western Australia: Implications for the conditions of crystallization and evidence for the influence of crustal-mantle fluid mixing. Mineral Petrol. 2014;108(6):759-775. https://doi.org/10.1007/s10017-014-0333-7
22. Coleman ML, Shepherd TJ, Durham JJ, Rouse JE, Moore GR. Reduction of water with zinc for hydrogen isotope analysis. Anal Chem. 1982;54(6):992-995. https://doi.org/10.1021/ac00243a035
23. Ohba T, Matsuo S. Precise determination of hydrogen and oxygen isotope ratios of water in fluid inclusions of quartz and halite. Geochim J. 1988;22(2):55-68. https://doi.org/10.2343/geochemj.22.55
24. Ohmoto H, Rye RO. Hydrogen and oxygen isotopic compositions of fluid inclusions in the Kuroko deposits, Japan. Econ Geol. 1974;69(6):947-953. https://doi.org/10.2113/gsecongeo.69.6.947
25. Schwarz HP, Harmon RS, Thompson P, Ford DC. Stable isotope studies of fluid inclusions in speleothems and their paleoclimatic significance. Geochim Cosmochim Acta. 1976;40(6):657-665. https://doi.org/10.1016/0016-7037(76)90111-3
26. Dennis PF, Rowe PJ, Atkinson TC. The recovery and isotopic measurement of water from fluid inclusions in speleothems. Geochim Cosmochim Acta. 2001;65(6):871-884. https://doi.org/10.1016/S0016-7037(00)00576-7
27. Simon K. Does $\delta^2$D from fluid inclusion in quartz reflect the original hydrothermal fluid? Chem Geol. 2001;177(3-4):483-495. https://doi.org/10.1016/S0009-2541(00)00417-4
28. Verheyden S, Genty D, Cattani O, Van Breukelen M. Characterization of fluid inclusions in speleothems: Heating experiments and isotopic

Orcid

Stefan de Graaf https://orcid.org/0000-0003-1054-1049
Hubert B. Vonhof https://orcid.org/0000-0002-0897-8244
42. Lachniet MS. Climatic and environmental controls on speleothem oxygen-isotope values. Quaternary Sci Rev. 2009;28(5–6):412-432. https://doi.org/10.1016/j.quascirev.2008.10.021

43. McDermott F. Palaeo-climate reconstruction from stable isotope variations in speleothems: A review. Quaternary Sci Rev. 2004;23(7–8):901-918. https://doi.org/10.1016/j.quascirev.2003.06.021

44. Demény A, Czuppon G, Kern Z, et al. Recrystallization-induced oxygen isotope changes in inclusion-hosted water of speleothems – Paleoclimatological implications. Quat Int. 2016;415:25-32. https://doi.org/10.1016/j.quaint.2015.11.137

45. Uemura R, Kina Y, Shen C-C, Omine K. Experimental evaluation of oxygen isotopic exchange between inclusion water and host calcite in speleothems. Clim Past. 2020;16(1):17-27. https://doi.org/10.5194/cp-16-17-2020

46. Sharp ZD, Atudorei V, Durakiewicz T. A rapid method for determination of hydrogen and oxygen isotope ratios from water and hydrous minerals. Chem Geol. 2001;178(1–4):197-210. https://doi.org/10.1016/S0009-2541(01)00262-5

47. Plessen B, Lüders V. Simultaneous measurements of gas isotopic compositions of fluid inclusion gases (N₂, CH₄, CO₂) using continuous-flow isotope ratio mass spectrometry. Rapid Commun Mass Spectrom. 2012;26(9):1157-1161. https://doi.org/10.1002/rcm.6201

48. Craig H. Isotopic variations in meteoric waters. Science. 1961;133(3465):1702-1703. https://doi.org/10.1126/science.133.3465.1702

49. Weissbach T. Spectroscopic isotope ratio analysis on speleothem fluid inclusions - analytics and paleoclimatic case studies. PhD Thesis. Heidelberg University; 2020.

50. Li H, Cheng H, Sinha A, et al. Hydro-climatic variability in the southwestern Indian Ocean between 6000 and 3000 years ago. Clim Past. 2018;14(12):1881-1891. https://doi.org/10.5194/cp-14-1881-2018

51. Fleitmann D, Matter A. The speleothem record of climate variability in southern Arabia. C R Geosci. 2009;341(8–9):633-642. https://doi.org/10.1016/j.crte.2009.01.006

52. Gonfiantini R, Panichi C, Tongiorgi E. Isotopic disequilibrium in travertine deposition. Earth Planet Sci Lett. 1968;5:55-58. https://doi.org/10.1016/0012-821X(68)90012-3

53. Kele S, Breitenbach SFM, Capezzuoli E, et al. Temperature dependence of oxygen- and clumped isotope fractionation in carbonates: A study of travertines and tufas in the 6-95°C temperature range. Geochim Cosmochim Acta. 2015;168:172-192. https://doi.org/10.1016/j.gca.2015.06.032

54. Taylor HP. Oxygen and hydrogen isotope relationships in hydrothermal mineral deposits. In: Barnes HL, ed. Geochemistry of Hydrothermal Ore Deposits. 3rd ed. New York: John Wiley & Sons, Inc.; 1997:229-302.

55. West AG, Goldsmith GR, Brooks PD, Dawson TE. Discrepancies between isotope ratio infrared spectroscopy and isotope ratio mass spectrometry for the stable isotope analysis of plant and soil waters. Rapid Commun Mass Spectrom. 2010;24(14):1948-1954. https://doi.org/10.1002/rcm.4597

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