The oxygen isotope equilibrium fractionation between sulfite species and water

Inigo A. Müller a,b,*, Benjamin Brunner a,c, Christian Breuer b,d, Max Coleman e,f, Wolfgang Bach b,d

a Biogeochemistry Department, Max Planck Institute for Marine Microbiology, Celsiusstrasse 1, 28359 Bremen, Germany
b MARUM-Center for Marine Environmental Sciences, University of Bremen, Leobener Strasse, 28359 Bremen, Germany
c Center for Geomicrobiology, Department of Biosciences, Aarhus University, Ny Munkegade 114-116, Aarhus 8000C, Denmark
d Department of Geosciences, University of Bremen, Klagenfurter Strasse, 28359 Bremen, Germany
e Planetary Surface Instruments Group, NASA Jet Propulsion Laboratory, California Institute of Technology, 4800 Oak Grove Drive, Pasadena, CA 91109, USA
f NASA Astrobiology Institute, USA

Received 13 September 2012; accepted in revised form 17 June 2013; available online 12 July 2013

Abstract

Sulfite is an important sulfoxy intermediate in oxidative and reductive sulfur cycling in the marine and terrestrial environment. Different aqueous sulfite species exist, such as dissolved sulfur dioxide (SO₂), bisulfite (HSO₃⁻), pyrosulfite (S₂O₅²⁻) and sulfite sensu stricto (SO₃²⁻), whereas their relative abundance in solution depends on the concentration and the pH. Conversion of one species into another is rapid and involves in many cases incorporation of oxygen from, or release of oxygen to, water (e.g. SO₂ + H₂O ↔ HSO₃⁻ + H⁺), resulting in rapid oxygen isotope exchange between sulfite species and water. Consequently, the oxygen isotope composition of sulfite is strongly influenced by the oxygen isotope composition of water. Since sulfate does not exchange oxygen isotopes with water under most earth surface conditions, it can preserve the sulfite oxygen isotope signature that it inherits via oxidative and reductive sulfur cycling. Therefore, interpretation of δ¹⁸O SO₄²⁻ values strongly hinges on the oxygen isotope equilibrium fractionation between sulfite and water which is poorly constrained. This is in large part due to technical difficulties in extraction of sulfite from solution for oxygen isotope analysis.

To overcome these challenges, anoxic isotope equilibration experiments were performed with dissolved sodium sulfite in solutions with distinct oxygen isotope signatures. Sulfite was precipitated using two different agents, barium chloride and silver nitrate. The experiments were performed at 22 °C and varying pH of 1.5, 6.3, 6.6, and 9.7 to investigate how changes in sulfite speciation affect the oxygen isotope equilibrium fractionation between sulfite and water.

From the experiments at pH 1.5 where SO₂ is the dominant sulfite species, a rough estimate of 37.0‰ was determined for the oxygen isotope equilibrium fractionation factor between aqueous SO₂ and water (ε⁰EQSO₂→H₂O). The oxygen isotope equilibrium fractionation between the aqueous phases is much larger than the known oxygen isotope equilibrium fractionation between gaseous SO₂ and water vapor, probably because of a stronger association with water molecules. At pH values of 6.3–9.7 a more firm estimate for the oxygen isotope equilibrium fractionation between HSO₃⁻, SO₃²⁻ and water (ε⁰EQSO₃²⁻→H₂O) of 15.2 ± 0.7‰ was obtained.

Our results provide new insights into the oxygen isotope fractionation during reductive and oxidative sulfur cycling. They demonstrate that isotope exchange between sulfite and water during dissimilatory sulfate reduction (DSR) alone is too small to be responsible for the apparent oxygen isotope equilibrium fractionation between sulfate and water mediated by DSR. Our
1. INTRODUCTION

1.1. The role of sulfite in shaping the oxygen isotope composition of sulfate

Due to its reactivity, sulfite is not abundant in the environment. It is released into the environment as SO$_2$ by magmatic processes such as the degassing of SO$_2$ from hydrothermal systems at the seafloor or from volcanically active terrestrial environments where it can undergo disproportionation to sulfate and sulfide or elemental sulfur (Kusakabe et al., 2000; Butterfield et al., 2011). An important anthropogenic source of SO$_2$ released into the atmosphere is the burning of fossil fuels (e.g. coal, oil, gas, wood and gasoline), which is one of the causes for acid rain (Holt et al., 1981; Zhao et al., 1988; Quinn, 1989; Pham et al., 1996).

Despite its scarceness in the environment, sulfite is assumed to be an important sulfury intermediate in reductive and oxidative sulfur cycling. It is an intermediate in dissimilatory sulfate reduction (DSR), where sulfate is used as electron acceptor in the oxidation of organic matter (Peck, 1962; Peck and Stulberg, 1962; Mizutani and Rafter, 1973; Fritz et al., 2002; Brunner and Bernasconi, 2005; Brunner et al., 2005, 2012; Turchyn et al., 2010). Sulfite is also thought to be an important intermediate during the oxidation of reduced sulfur compounds (e.g. H$_2$S, HS$^-$, FeS$_2$ and FeS) and elemental S which exist in high abundance under reducing conditions in marine sediments at the seafloor or in ore deposits (Schippers et al., 1996). These reduced sulfur compounds can be oxidized by microorganisms or abiotically where molecular oxygen (O$_2$) and/or ferric iron (Fe$^{3+}$) act as oxidants. Oxidative and reductive sulfur cycling leaves imprints in the oxygen isotope composition of sulfate, an ion that does not spontaneously exchange oxygen isotopes with water unless exposed to extremely high temperatures and/or extremely low pH (Lloyd, 1968; Chiba and Sakai, 1985). Therefore, the oxygen isotope composition of sulfate has been used to investigate sulfur oxidation (e.g. Lloyd, 1967, 1968; Böttcher et al., 2001, 2005; Balci et al., 2007; Brunner et al., 2008; Kohl and Bao, 2011; Balci et al., 2012; Brabec et al., 2012), DSR (Mizutani and Rafter, 1973; Fritz et al., 1989; Wortmann et al., 2007) as well as past and present reductive and oxidative sulfur cycling on Earth (Turchyn and Schrag, 2004; Bottrell and Newton, 2006; Bottrell et al., 2009; Pirlet et al., 2010; Riedinger et al., 2010). Unlike sulfite, sulfate easily exchanges its oxygen isotopes with ambient water, and due to its importance as intermediate in oxidative and reductive sulfur cycling, it is very likely that the isotope effects associated with this isotope exchange process are pivotal in shaping the isotopic signature of sulfate. So far, the equilibrium isotope effect for oxygen isotope exchange between sulfate and water has not been determined accurately, only a preliminary estimate exists ($\sim$11.5‰ at 23 °C, Brunner et al., 2006).

1.2. Species-dependent oxygen isotope exchange between sulfite and water

In aqueous solutions, sulfite exists in the form of different sulfur species which have in common that the sulfur atom has an oxidation state of +IV. Simon and Waldmann (1955, 1956) investigated the characteristics of sulfite species in aqueous solutions (reaction 1, 2 and 3) and their dependence on the pH, as well as the total concentration of sulfite species. They observed that SO$_2$ in solution forms bisulfite ions (H$\text{SO}_3^-$, reaction 1) with sulfurous acid (H$_2$SO$_3$) as a hypothetical intermediate. However, H$_2$SO$_3$ as species so far has not been detected in aqueous solutions (Sülzle et al., 1988). Simon and Waldmann (1955, 1956) observed that bisulfite forms sulfite sensu stricto (SO$_3^{2-}$) and another proton via reaction 2 and described a reaction where two H$\text{SO}_3^-$ ions react to form pyrosulfite (S$_2$O$_5^{2-}$) and water (Eq. 3). All abbreviations in the text are listed in Table 1 with a short explanation.

$$\text{SO}_2 + \text{H}_2\text{O} \leftrightarrow (\text{H}_2\text{SO}_3) \leftrightarrow \text{H}\text{SO}_3^- + \text{H}^+ \quad (1)$$

$$\text{H}\text{SO}_3^- \leftrightarrow \text{SO}_3^{2-} + \text{H}^+ \quad (2)$$

$$2\text{H}\text{SO}_3^- \leftrightarrow \text{S}_2\text{O}_5^{2-} + \text{H}_2\text{O} \quad (3)$$

By now, the reactions between sulfite species have been studied in more detail, and the kinetics of oxygen exchange between water and sulfite species are fairly well constrained (Eigen et al., 1961; Betts and Libich, 1970; Betts and Voss, 1970; Connick et al., 1982; Horner and Connick, 1986, 2003). Betts and Voss (1970) determined the kinetics of the sulfite water exchange (Eq. 1) with $^{18}$O as stable isotope tracer and by mass spectrometry. Horner and Connick (1986, 2003) evaluated the equilibrium constants for the isomerization of the bisulfite ion (Eq. 4) as well as the kinetics between the isomers of bisulfite, of pyrosulfite (S$_2$O$_5^{2-}$) and water (Eq. 3) by studying the characteristics of these sulfite species in solution with $^{18}$O nuclear magnetic resonance spectroscopy.

$$\text{HSO}_3^- \leftrightarrow \text{SO}_3\text{H}^- \quad (4)$$

The relative distribution of the bisulfite isomers as a function of pH range is currently not well constrained. Horner and Connick (2003) observed that the SO$_2$H$^-$ reacts more rapid with H$^+$ via Eq. (1) with decreasing pH, whereas exchange between the two isomers via Eq. (4) increases with pH, which is possibly related to the presence of SO$_3^{2-}$ as an intermediate at higher pH. Given the equilibrium constants (e.g. from Horner and Connick, 2003) and total sulfite concentration it is possible to quantitatively predict the sulfite species distribution in an aqueous solution as a function of pH. Fig. 1 depicts such a distribution for a total sulfite concentration of 0.02 M – the concentration chosen for the...
experiments in our study – and is based on equilibrium constants for Eqs. (1–3) ($Q_1 = 1.37 \text{ m}, Q_2 = 6.34 \text{ m}, Q_3 = 0.082 \text{ m}$). The differences between molal (m) and molar (M) units is considered to be unimportant compared to uncertainties in the rate constant values (Horner and Connick, 2003), the constant for the water dissociation ($pQ_{\text{H}_2\text{O}} = 13.79$), and on the equilibrium constant for the dissociation of sodium sulfite ion ($\text{NaSO}_3^- = \text{Na}^+ + \text{SO}_3^{2-}$, log $K = 0.83$, from Visual MINTEQ database 2008).

In Eqs. (1 and 3) $\text{SO}_2$ and $\text{S}_2\text{O}_5^{2-}$ react with water during the conversion to $\text{HSO}_3^-$. Oxygen isotope exchange between sulfite species and water occurs during these reactions, with the consequence that oxygen isotope exchange is fastest at extremely low pH where $\text{SO}_2$ is the most prominent sulfite species (Holt et al., 1981). The oxygen isotope exchange between sulfite species and water is still rapid at circum neutral pH conditions but slows down tremendously at pH above 10, until it ceases at pH 12.7 where only $\text{SO}_3^{2-}$ is in solution (see Betts and Voss, 1970; Horner and Connick, 2003).

1.3. Precipitation of sulfite salts for subsequent oxygen isotope analysis and determination of isotope fractionation between sulfite and water

Even though the kinetic behavior of sulfite species in aqueous solutions has been explored in detail, reliable...
information on the oxygen isotope equilibrium fractionation between sulfite species and water is lacking. One reason for the lack of this data is the technical difficulty of extracting sulfite species from solution for oxygen isotope analysis. The experimental challenge arises from the fact that typically more than one sulfite species coexist in solution (Fig. 1) causing extraction techniques such as precipitation of sulfite as barium sulfite or silver sulfite to induce conversion of all sulfite species, (i.e. SO$_2$ and S$_2$O$_5^{2-}$/CO$_3^{2-}$) to SO$_3^{2-}$/CO$_3^{2-}$, via Eqs. (1–4). The oxygen isotope signature of the precipitated sulfite salt cannot be directly attributed to an equilibrium isotope fractionation between a certain sulfite species and water for two reasons: First, each sulfite species is expected to possess an individual oxygen isotope equilibrium fractionation which could vary for the different sulfite species, and second, the wholesale conversion of all sulfite species to SO$_3^{2-}$/CO$_3^{2-}$ is likely to induce incorporation of additional oxygen from water and kinetic oxygen isotope effects. Furthermore, the precipitated sulfite salts such as BaSO$_3$ can be strongly hygroscopic, resulting in uptake of water vapor from the air, which typically introduces oxygen with a much lighter isotopic composition than the isotope composition of the sample.

To overcome these challenges and to determine the effective oxygen isotope equilibrium fractionation between different sulfite species and water, replicate oxygen isotope equilibration experiments were performed in water with three distinct isotope compositions of which sulfite was collected as two different sulfite salts (BaSO$_3$ and Ag$_2$SO$_3$). For each sulfite salt, four aliquots were precipitated by addition of isotopically distinct precipitation solutions (barium chloride and silver chloride solution, respectively). This approach allows disentangling isotope effects caused by the precipitation process from the actual equilibrium isotope effects. The comparison between the results obtained from different sulfite salts helped assessing the reliability of different sulfite precipitation strategies.

2. METHODS

To determine the oxygen isotope equilibrium fractionation between dissolved sulfite species and water three prerequisites must be fulfilled: the experimental/technical challenges related to the collection of samples for isotope analysis must be identified, experimental approaches to overcome these challenges must be established, and an isotope mass balance that considers all relevant factors affecting the oxygen isotope composition of the precipitates in our experiments must be developed.

2.1. Experimental challenges and approaches on how to cope with them

Isotope effects during the conversion of sulfite species into sulfite salt described schematically in Fig. 2 have a major impact on the experimental results. Here, we summarize the resulting experimental challenges.

The precipitation of sulfite species in the presence of cations such as Ba$^{2+}$ or Ag$^+$ induces the conversion of all sulfite species to SO$_3^{2-}$ prior to the precipitation as BaSO$_3$ or Ag$_2$SO$_3$, respectively. This precipitation procedure causes kinetic oxygen isotope effects during conversion of SO$_2$ and S$_2$O$_5^{2-}$ (Eqs. 1 and 3) into HSO$_3$/CO$_3$H as well as the incorporation of water oxygen during these reactions. The H$_2$O molecules involved in the two reactions might also undergo kinetic oxygen isotope effects (Fig. 2: left side).

The isotope effects during the precipitation of SO$_3^{2-}$ as BaSO$_3$ or Ag$_2$SO$_3$ might be mainly due to kinetic isotope effects, but we cannot exclude oxygen isotope equilibrium fractionation between the solid and dissolved sulfite phase.
The precipitation of sulfite salts might also trap minor amounts of water molecules within the salt structure ("lattice water"; see Walton and Walden, 1946a, 1946b; Neagley and Rochester, 1988) and sulfite salts might be hygroscopic, where they attract water vapor with entirely distinct oxygen isotope composition (Fig. 2: right side). A further challenge is the ongoing rapid oxygen exchange between sulfite species and water during the precipitation procedure, which would lead to a non-quantitative conversion of the oxygen isotope composition of certain sulfite species into the sulfite salt.

To cope with the experimental challenges we performed the equilibration experiments and the precipitation procedure under specific conditions that enabled us to evaluate the quality of our oxygen isotope results and to determine the oxygen isotope equilibrium fractionation between sulfite and water.

The experiments were carried out under anoxic conditions to prevent oxidation of sulfite to sulfate. Performing the experiments in isotopically distinct experimental waters (H$_2$O$_{exp}$) allowed us to assess if there are external (e.g. water vapor due to the hygroscopic nature of sulfite salts) or internal contaminants (e.g. sulfate from sulfite oxidation or sulfate in the original starting material).

To limit the number of sulfite species to consider in the isotope mass balance, we chose a total concentration of sulfite of 0.02 M combined with two different pH settings: very low pH (pH 1.5, SO$_3^{2-}$ and HSO$_3^{-}$ dominate) and slightly acidic to basic solutions (pH 6.3–9.7, HSO$_3^{-}$ and/or SO$_3^{2-}$ dominate). This concentration is higher than in most natural environments, which has the advantage that enough sample material is available for the assessment of isotope effects related to the oxygen isotope exchange between sulfite species and water, while the sulfite species S$_2$O$_5^{2-}$ is present in negligible amounts, allowing us to ignore this sulfite species in our isotope mass balance approach (Fig. 1). An additional simplification of the isotope mass balance calculation was based on the consideration that the molecular structure of the two bisulfite isomers (HSO$_3^{-}$, SO$_3^{2-}$) and SO$_3^{2-}$ is similar, thus also similar equilibrium isotope effects relative to the water can be expected. Therefore, we treated the oxygen equilibrium isotope effects for these three species as identical. This simplification ignores potential oxygen isotope effects between acid/conjugate-base pairs, which can be considerable. For example, in aqueous solution at 25 °C, bicarbonate (here denoted as CO$_3^{2-}$ to emphasize that the proton interacts with oxygen from bicarbonate) is enriched in $^{18}$O relative to the carbonate ion (CO$_3^{2-}$) by approximately 6‰ (Beck et al., 2005).

Whereas the isotope effect between the SO$_3^{2-}$ isomer and SO$_3^{2-}$ might be as large as what has been reported for CO$_3^{2-}$, H$^+$ and CO$_3^{2-}$ the oxygen isotope effect between the HSO$_3^{-}$ isomer and SO$_3^{2-}$ is expected to be smaller because the proton does not directly interact with the oxygen atoms of sulfite.

To prevent oxygen exchange between sulfite species and water during the precipitation of sulfite salts we shifted the pH to strongly basic conditions, thereby inducing rapid conversion of all sulfite species to SO$_3^{2-}$, which does not exchange oxygen with water. This procedure is only applicable for sulfite precipitation with BaSO$_3$ and not with Ag$_2$SO$_3$, which would precipitate as AgOH under strongly basic conditions. Using precipitation solutions (H$_2$O$_{prec}$) with different $^{18}$O$_{H_2O}$ values allowed for the quantification of the amount of oxygen incorporated from water during pH-shifting and subsequent sulfite salt precipitation. High extent of incorporation of oxygen from the precipitation solution into the sulfite salts formed indicated ongoing oxygen isotope exchange during the precipitation process, and thus provided information on the reliability of the calculated estimates for equilibrium isotope fractionation. We precipitated sulfite species as different sulfite salts (BaSO$_3$, Ag$_2$SO$_3$) to assess the consequences of different precipitation kinetics and solubilities on the observed
isotope effects (i.e. rapid and quantitative precipitation vs. sluggish and non-quantitative precipitation) and to assess if there is a benefit in obtaining a sulfite salt less hygroscopic than the other.

2.2. Equilibrium experiments

2.2.1. 18O labeled experimental solutions

For the equilibration experiments, water with three different oxygen isotope compositions (\(\delta^{18}O_{\text{H}_{2}O_{\text{exp}}}\) of \(-7.7\%_{\text{o}}, 28.1\%_{\text{o}}\), and \(64.6\%_{\text{o}}\)) were prepared by mixing de-ionized water (18 MΩ, abbreviated as MQ) with appropriate amounts of water consisting of 98% \(^{18}\)O (NUKEM GmbH). This range of isotope values was chosen to obtain a sufficient spread in isotope compositions from which reliable results could be obtained.

2.2.2. Preparation of sulfite equilibration experiments

Glass flasks (Duran glass bottles, volume 290 ml) were filled with 250 ml of experimental solutions (\(\text{H}_2\text{O}_{\text{exp}}\)), leaving \(~40\) ml headspace upon sealing with a rubber stopper. To assure anoxic conditions, the solutions in the flasks were purged with oxygen-free nitrogen gas (N\(_2\)) for 30 min. Meanwhile 0.63 g sodium sulfite (\(\text{Na}_2\text{SO}_3\)) from Fluka, \(M_R = 126.04\) g mol\(^{-1}\) was weighed into 1.5 ml plastic microtubes from which it was added to the experimental flasks. To prevent O\(_2\) contamination, this transfer was made inside a plastic glove bag fully purged twice with a N\(_2\) atmosphere (Fig. 3a). The experimental solutions were again purged for 5 min with N\(_2\) gas prior to addition of the \(\text{Na}_2\text{SO}_3\) powder, after which the flasks were immediately sealed with rubber stoppers.

2.2.3. Adjusting the pH of the equilibrium experiments

The experiments at pH 1.5 were adjusted with hydrochloric acid by injecting 2 ml 6 M HCl through the rubber stopper of the experimental flasks inside the glove bag (Fig. 3a). This avoids loss of gaseous SO\(_2\) that is produced at this low pH (Fig. 1). The experiments at pH 6.3 and 6.6 were performed in 40 mM acetate buffer solution prepared by adding 0.94 g sodium acetate trihydrate (\(M_R = 136.1\) g mol\(^{-1}\)) and 222 µl acetic acid (\(M_R = 60.05\) g mol\(^{-1}\)) to the experimental solutions. Upon shaking the sealed experiment flasks the \(\text{Na}_2\text{SO}_3\) dissolved immediately. The experiments at pH 9.7 were prepared simply by adding \(\text{Na}_2\text{SO}_3\) into the solutions. The pH was measured with a pH electrode (pH-Meter 766 Calimatic from Knick) with accuracy of less than 0.09 pH units.

2.2.4. Experiment duration

The experiments at pH 1.5 were equilibrated for 1 day, the experiments at near neutral pH for 2 days and experiments at pH 9.7 were equilibrated for 3 days to ensure that the oxygen of the sulfite species was in equilibrium with the surrounding water. This is more than sufficient time for sulfite species to reach oxygen exchange equilibrium with the water as the study by Betts and Voss (1970) revealed half-lives of 25.3 h and 1.3 min for oxygen exchange at pH 10.5 and pH 8.9, respectively. At pH below 8.2 the oxygen exchange is even more rapid so that it was not possible to determine the half-life (Betts and Voss, 1970). Over the time of equilibration, the experiment flasks were kept inside the glove bag under a N\(_2\) atmosphere.

2.3. Precipitation of sulfite salts with isotopically distinct precipitation solutions and different cations (\(\text{Ag}^{+}\), \(\text{Ba}^{2+}\))

2.3.1. 18O labeled solutions for precipitation

Precipitation of sulfite salts with solutions of distinct \(\delta^{18}O_{\text{H}_{2}O_{\text{exp}}}\) values enabled us to calculate the amount of water derived oxygen incorporated during precipitation. Water with four different oxygen isotope compositions (\(\delta^{18}O_{\text{H}_{2}O_{\text{exp}}}\) of \(-7.7\%_{\text{o}}, 48\%_{\text{o}}, 162\%_{\text{o}}\), and \(212\%_{\text{o}}\)) was prepared by mixing MQ water with appropriate amounts of water consisting of 98% \(^{18}\)O.

2.3.2. Added solutions containing \(\text{Ag}^{+}\) or \(\text{Ba}^{2+}\)

Three sets of solutions to be added were prepared. For experiments at pH 6.3 and 6.6, solutions were prepared by dissolving AgNO\(_3\) (\(M_R = 169.87\) g mol\(^{-1}\), AppliChem) in the four isotopically distinct solutions to a final concentration of 70 mM. For experiments at pH 6.3 and 9.7, the solutions were prepared by dissolving BaCl\(_2\) 2H\(_2\)O (\(M_R = 244.26\) g mol\(^{-1}\), Sigma Aldrich) in the isotopically distinct solutions to a concentration of 50 mM. For experiments at pH 1.5 and 6.6, the solutions were prepared by dissolving BaCl\(_2\) + 2H\(_2\)O and sodium hydroxide (NaOH, \(M_R = 39.9\) g mol\(^{-1}\)) in the isotopically distinct solutions to a concentration of 50 and 100 mM, respectively. The addition of NaOH induces rapid conversion of all aqueous sulfite species to SO\(_{3}^{2-}\), and efficiently stops further oxygen isotope exchange between sulfite and water. This approach is feasible with barium, but not applicable to silver, because silver would precipitate as silver hydroxide (AgOH) and silver oxide (Ag\(_2\)O) under alkaline conditions (Biedermann and Sillén, 1960). A study by Krekeler and Cypionka (1995) showed that sulfite is not abiotically oxidized in presence of nitrate under neutral pH, however, this possibility cannot be fully excluded for acid conditions. All solutions were purged with N\(_2\), sealed with rubber stoppers and placed inside the glove bag together with the experimental solutions.

2.3.3. Precipitation of sulfite salts

Sulfite rapidly oxidizes in the presence of air, with fastest rates in seawater occurring at pH 6.5 (Zhang and Millero, 1991). Therefore, the precipitation of sulfite salts was carried out in the N\(_2\)-flushed glove bag. Aliquots (5 ml) of the four isotopically distinct solutions (\(\text{H}_2\text{O}_{\text{add}}\)) containing the \(\text{Ag}^{+}\) or \(\text{Ba}^{2+}\), respectively, were distributed into plastic centrifuge tubes (Sarstedt, 15 ml), after which 5 ml of the experimental solution was pipetted into each (Fig. 3b). Thereby, 5 ml of an experimental solution (\(\delta^{18}O_{\text{H}_{2}O_{\text{exp}}}\)) was mixed with 5 ml of the cation containing solution (\(\delta^{18}O_{\text{H}_{2}O_{\text{add}}}\), resulting in 10 ml of a precipitation solution (\(\text{H}_2\text{O}_{\text{prec}}\)). The isotope composition of this precipitation solution (\(\delta^{18}O_{\text{H}_{2}O_{\text{prec}}}\)) was calculated from the known amounts of the former two constituents (actual mol amounts of isotopologues in 5 ml of \(\text{H}_2\text{O}_{\text{add}}\) and in 5 ml of \(\text{H}_2\text{O}_{\text{add}}\)) and their measured isotope compositions (\(\delta^{18}O_{\text{H}_{2}O_{\text{exp}}}\) and
The oxygen isotope composition of experimental water and of the precipitation solution can strongly influence the measured $\delta^{18}O_{\text{SO}_3^2/\text{CO}_3}$ value.

### 2.3.4. Collection of sulfite salts

The precipitated salts (described above) were collected in capped centrifuge tubes and then centrifuged at 2000 rpm for 5 min. Subsequently the supernatant was decanted and 5 ml of acetone was added to the remaining salt precipitate (after Betts and Libich, 1970) followed by vigorous mixing and subsequent centrifugation. The acetone supernatant was decanted and the acetone rinse procedure was repeated once more, before the precipitates were oven-dried for 2 days at 50°C. Initially, a few selected samples were alternatively freeze-dried. Because both methods yielded identical results all precipitates were subsequently dried in the oven.

### 2.3.5. Trapping of laboratory water vapor

Water vapor is a potential contaminant in hygroscopic sulfite precipitates. To determine the oxygen isotope composition of water vapor in our laboratory, we collected ice that accumulated on a cooling coil ($T = -27°C$) in a closed vessel. To accumulate enough ice for isotope measurements we had to open the lid of the vessel several times so that fresh water vapor could enter and subsequently condense during closed phases.

### 2.4. Stable isotope measurements

The oxygen isotope composition of the precipitates was measured by a Finnigan DELTA plus continuous flow isotope ratio mass spectrometer (IRMS) coupled to a TC/EA. In brief, the precipitates were weighed (BaSO$_3$ between 0.3 and 0.4 mg and Ag$_2$SO$_3$ between 0.6 and 0.8 mg) into silver cups which were loaded into an auto sampler coupled to a thermochemical reduction device (TC/EA Finnigan). In the TC/EA the samples were reduced at 1450°C in the presence of carbon to CO gas, which was carried through a gas chromatography column before analysis by IRMS. The isotope results are reported in the standard $\delta$-notation where the isotope ratio ($R = ^{18}O/^{16}O$) of the sample is compared to the ratio of the Vienna Standard Mean Ocean Water (VSMOW), i.e. $\delta^{18}O = (R_{\text{sample}}/R_{\text{VSMOW}} - 1) \times 1000$.


\[ \text{Prec}_{\text{oxygen}} = 3 \cdot [\text{SO}_2] + 3 \cdot [\text{HSO}_3^-] + 3 \cdot [\text{SO}_3^{2-}] + 3 \cdot [\text{SO}_4^{2-}] + [\text{H}_2\text{O}_{\text{prec}} \text{ incorp}] + [\text{cont}] \]  

(6)

In Eq. (6) the amount of \( \text{SO}_2 \) is multiplied by a factor of 3 because \( \text{SO}_2 \) is converted to \( \text{HSO}_3^- \) via Eq. (1) prior to the precipitation of the sulfite salt, with the third oxygen atom being contributed from the precipitation solution. This is accounted for in the isotope mass balance for sulfite precipitates by assuming that the oxygen from \( \text{SO}_2 \) retains its isotope composition (i.e. \( 2/3 \delta^{18} \text{O}_{\text{SO}_2} \)) and that one third of the contributed oxygen comes from the precipitation solution (\( \delta^{18} \text{O}_{\text{H}_2\text{O}_{\text{prec}}} \)) including a potential oxygen isotope fractionation for the reaction of water–oxygen (\( \delta^{18} \text{O}_{\text{H}_2\text{O}_{\text{prec}} = -0.3 \%o} \)).

\[ \text{Prec}_{\text{oxygen}} \cdot \delta^{18} \text{O}_{\text{prec}} = 3 \cdot [\text{SO}_2] \cdot \left( \frac{2}{3} \cdot \delta^{18} \text{O}_{\text{SO}_2} + \frac{1}{3} \cdot \left( \delta^{18} \text{O}_{\text{H}_2\text{O}_{\text{prec}} = -0.3 \%o} + \delta^{18} \text{O}_{\text{H}_2\text{O}_{\text{prec}} = -0.3 \%o} \right) \right) 
+ 3 \cdot [\text{HSO}_3^-] \cdot \delta^{18} \text{O}_{\text{HSO}_3^-} 
+ 3 \cdot [\text{SO}_3^{2-}] \cdot \delta^{18} \text{O}_{\text{SO}_3^{2-}} 
+ [\text{H}_2\text{O}_{\text{prec}} \text{ incorp}] \cdot \delta^{18} \text{O}_{\text{H}_2\text{O}_{\text{prec}}} 
+ [\text{cont}] \cdot \delta^{18} \text{O}_{\text{cont}} \]  

(7)

where \( \delta^{18} \text{O}_{\text{prec}} \) refers to the oxygen isotope composition of the precipitate, \( \text{H}_2\text{O}_{\text{prec}} \) designates water molecules from the precipitation solution that are incorporated in the sulfite salt during precipitation and \( \delta^{18} \text{O}_{\text{cont}} \) to the oxygen isotope composition of the contaminant.

This isotope mass balance presented in Eq. (7) is based on the assumption that there is either no isotope effect for conversion of one sulfite species to another (e.g. no isotope discrimination between remaining \( \text{SO}_2 \) and \( \text{SO}_3^- \) that is converted to \( \text{SO}_4^{2-} \)), or that there is a quantitative conversion of all sulfite species without simultaneous oxygen isotope exchange with water. If simultaneous oxygen isotope exchange with water occurs during the conversion of sulfite into the sulfite precipitate a quantitative conversion of all sulfite species would not suffice to justify the above assumption. In this case, estimates for equilibrium isotope fractionations based on Eq. (7) have to be considered with caution, as they might be biased by ignoring kinetic isotope effects resulting from the transformation of one sulfite species into another.

Eq. (7) can be further simplified. In contrast to the distinct structure of the \( \text{SO}_2 \) molecule the structure of the molecules of both bisulfite isomers (\( \text{HSO}_3^- \), \( \text{SO}_3^{2-} \)) and \( \text{SO}_4^{2-} \) are similar. We expect the oxygen isotope fractionation with water to be similar for these three sulfite species, and consequently assign the same value to the isotope composition of the bisulfite isomers and \( \text{SO}_3^{2-} \) (\( \delta^{18} \text{O}_{\text{SO}_3^{2-}} \)) which are in oxygen isotope equilibrium with water. This simplification neglects the fact that oxygen isotope fractionation between
acid/conjugate-base pairs can be considerable, for example, in aqueous solution at 25 °C, bicarbonate is enriched in $\text{^{18}O}$ relative to the carbonate ion ($\text{CO}_3^{2-}$) by approximately $6\%$ (Beck et al., 2005). Furthermore, we consider that oxygen isotope exchange may happen during the precipitation and thus introduce quantities for sulfate species that are in original condition (e.g. $[\text{SO}_2\text{orig}]$) and quantities for sulfate species that have exchanged isotopes during the precipitation procedure (e.g. $[\text{SO}_2\text{exchanged}]$):

$$[\text{Prec}_{\text{oxygen}}] \cdot \delta^{18}\text{O}_{\text{prec}} = 3 \cdot [\text{SO}_2\text{orig}] \cdot \left( \frac{2}{3} \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) \right) + \frac{1}{3} \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) + \frac{1}{3} \cdot \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) + 3 \cdot \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) + 3 \cdot \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) + \left( [\text{SO}_2\text{exchanged}] + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) + \left( [\text{cont}] \cdot \delta^{18}\text{O}_{\text{cont}} \right) \right)$$

(8)

In Eq. (8), $\delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}}$ represent the oxygen isotope equilibrium fractionation between dissolved $\text{SO}_2$ and water, and between $\text{SO}_2^{2-}$ and water, respectively. We use {} to indicate that all terms within these parentheses possess the same property, which is defined as subscript right of the parenthesis, e.g. {}$\text{orig}$. Division of Eq. (8) by $[\text{Prec}_{\text{oxygen}}]$ rewritten in a simplified form results in:

$$\delta^{18}\text{O}_{\text{prec}} = a_1 \cdot \left( 2 \cdot \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) \right) + \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) + a_2 \cdot \left( 2 \cdot \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) \right) + \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) + 3 \cdot b_1 \cdot \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) + 3 \cdot b_2 \cdot \left( \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} \right) + c \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + d \cdot \delta^{18}\text{O}_{\text{cont}}$$

with:

$$a_1 = \frac{[\text{SO}_2\text{orig}]}{[\text{Prec}_{\text{oxygen}}]}, \quad a_2 = \frac{[\text{SO}_2\text{exchanged}]}{[\text{Prec}_{\text{oxygen}}]},$$

$$b_1 = \frac{[[\text{HSO}_4^-] + [\text{SO}_2\text{H}^+] + [\text{SO}_2^{2-}]]_{\text{orig}}}{[\text{Prec}_{\text{oxygen}}]}$$

$$b_2 = \frac{[[\text{HSO}_4^-] + [\text{SO}_2\text{H}^+] + [\text{SO}_2^{2-}]]_{\text{exchanged}}}{[\text{Prec}_{\text{oxygen}}]}$$

$$c = \frac{[\text{H}_2\text{O}_{\text{prec incorp}}]}{[\text{Prec}_{\text{oxygen}}]}$$

$$d = \frac{[\text{cont}]}{[\text{Prec}_{\text{oxygen}}]}$$

$$1 = 3 \cdot (a_1 + a_2) + 3 \cdot (b_1 + b_2) + c + d$$

(9)

The expression $1 = 3(a_1 + a_2) + 3(b_1 + b_2) + c + d$ results from the division of Eq. (8) by $[\text{Prec}_{\text{oxygen}}]$ which normalizes all oxygen contributions to the total oxygen of the precipitate (Prec$_{\text{oxygen}}$). We can rearrange Eq. (9) to:

$$\delta^{18}\text{O}_{\text{prec}} = (2 \cdot a_1 + 3 \cdot b_1) \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + (a_1 + 3 \cdot a_2) + 3 \cdot b_2 + c \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + 2 \cdot (a_1 + a_2) \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + 3 \cdot (b_1 + b_2) \cdot \delta^{18}\text{O}_{\text{cont}}$$

(10)

By combination of all terms related to the experimental solution and the precipitation solution, respectively, we can rewrite Eq. (10) as:

$$\delta^{18}\text{O}_{\text{prec}} = k \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + m \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + 2 \cdot (a_1 + a_2) \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + 3 \cdot (b_1 + b_2) \cdot \delta^{18}\text{O}_{\text{cont}}$$

with:

$$k = 2 \cdot a_1 + 3 \cdot b_1, \quad m = a_1 + 3 \cdot a_2 + 3 \cdot b_2 + c$$

(11)

The value of $m$ is a measure of how much water oxygen was incorporated into the sulfite salt during the precipitation procedure (via sulfite species conversion, oxygen reequilibration or incorporation of lattice water; see Fig. 2), whereas the value of $k$ is a measure of how much water oxygen in the sulfite salt originates from the experimental solution (oxygen of sulfite species in equilibrium with the experimental solution). The values of $\delta^{18}\text{O}_{\text{prec}}, \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}}$ and $\delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}}$ can be experimentally accessed. Consequently, $m$ can be determined for each individual experiment where the isotope composition of experimental water (\$^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}}$) is constant and the isotope composition of the precipitation solution (\$^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}}$) is varied, via graphical determination of the slope of the regression line in a \$^{18}\text{O}_{\text{prec}}$ vs. \$^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}}$ plot (for an example, see Fig. 4a). In such a plot the four precipitated sulfite salts with distinct oxygen isotope compositions were prior to precipitation in equilibrium with the same experimental solution as dissolved sulfite species. Once the value of $m$ is determined by the above procedure, Eq. (11) can be rearranged to:

$$\delta^{18}\text{O}_{\text{prec}} = m \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} = k \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + 2 \cdot (a_1 + a_2) \cdot \delta^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} + 3 \cdot (b_1 + b_2) \cdot \delta^{18}\text{O}_{\text{cont}}$$

(12)

Thus, the value of $k$ for an experiment at one specific pH can be determined by the slope of a regression line in a \$^{18}\text{O}_{\text{prec}}$ vs. \$^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}}$ plot, whereas the intercept of the regression line at \$^{18}\text{O}_{\text{H}_2\text{O}_{\text{prec}}} = 0$ corresponds to the constant term in Eq. (12). This term is composed of the isotope effects, the isotope composition of the contaminant and their relative contributions (Fig. 4b). Small experimental differences between the three isotopically distinct experimental solutions of one specific pH can produce slightly distinct values for $m$ which require the determination of the water oxygen contribution in the sulfite salt from the experimental solution $k$ as described in Eq. (12). Alternatively, $k$, $m$ and the value for the
intercept can also be directly determined by performing a multiple linear regression analysis, which has the disadvantage that slight differences in the values for $m$ cannot be taken into account in the determination of the value for the intercept and $k$. Therefore, we consider using a multiple linear regression analysis for this study only a second choice, although it is undoubtedly a more elegant approach than using two consecutive regressions (two-step linear regression).

If experiment solution and precipitation solution were the only oxygen sources available (i.e., no contaminant, $d = 0$), the sum of $k$ and $m$ would always be 1. Consequently, the value of $d$ can be determined:

$$d = 1 - (k + m) \quad (13)$$

The knowledge of the values of $\delta^{18}O_{\text{prec}}$, $\delta^{18}O_{\text{H}_{2}O_{\text{exp}}}$, $\delta^{18}O_{\text{H}_{2}O_{\text{cont}}}$, $m$, $k$, and $d$ is insufficient to determine the remaining unknown parameters in Eq. (12) (i.e., $a_1$, $a_2$, $b_1$, $b_2$, $e_1$,$O_{\text{SO}_{2}^{-}}$,$e_{2}$,$O_{\text{SO}_{2}^{-}}$,$e_{3}$,$O_{\text{H}_{2}O_{\text{SO}_{2}^{-}}}$,$e_{4}$,$O_{\text{SO}_{2}^{-}}$,$e_{5}$,$O_{\text{H}_{2}O_{\text{SO}_{2}^{-}}}$, $\delta^{18}O_{\text{cont}}$), but we can further simplify the isotope mass balance by performing experiments at specific pH (exclusion of certain sulfite species) or by applying precipitation techniques that minimize the effect of simultaneous oxygen isotope exchange with ambient water.

### 3. RESULTS

All the measured isotope values ($\delta^{18}O_{\text{H}_{2}O_{\text{prec}}}$, $\delta^{18}O_{\text{H}_{2}O_{\text{exp}}}$, and $\delta^{18}O_{\text{prec}}$) from the experiments that were performed under different pH conditions and with or without shifting the pH during precipitation are reported in Table 2. The values of the oxygen isotope compositions of the experimental solution, the precipitation solution, and the precipitated sulfite salt were further used to calculate the relative oxygen source contribution (e.g., $m$, $k$, $d$) in the sulfite salts for all individual experiments after Eq. (11). In the following, we report and discuss the values obtained via the two-step regression, which slightly deviate from the values one would obtain by using a multiple linear regression analysis (Table 3). The reported standard deviations are based on simplified error estimates obtained by linear regression analysis calculated for 95% confidence limits.

#### 3.1. Relative oxygen source contribution into the sulfite salts of the individual experiments (Table 3)

We first compared values of $m$ for experiments that were carried out at a pH of 6.3 (no SO$_2$ species expected, Fig 1), where sulfite salts were precipitated with different solutions containing either Ba$^{2+}$ or Ag$^+$. The experiments using Ag$^+$ as the precipitation agent yielded a relative incorporation of oxygen from the precipitation solution of $m_{\text{Ag}^+} = 0.48 \pm 0.04$; a value much smaller than the experiments where sulfite was precipitated with Ba$^{2+}$ ($m_{\text{Ba}^{2+}} = 0.90 \pm 0.01$). From the $m$-values we could calculate the relative oxygen contributions of the experimental solution (Eq. 12) with $k_{\text{Ag}^+} = 0.58 \pm 0.04$ and $k_{\text{Ba}^{2+}} = 0.01 \pm 0.01$, and with the relation $1 = m + k + d$ we obtained a relative oxygen contribution from the contaminant of $d_{\text{Ag}^+} = -0.07 \pm 0.06$ and $d_{\text{Ba}^{2+}} = 0.09 \pm 0.02 (9\%$ contaminant), respectively. The slightly negative value for $d$ (sum of $k$ and $m$ close to 1) shows that, unlike for BaSO$_4$, there is no contaminant in Ag$_2$SO$_4$.

The experiments at pH 6.6 where we precipitated the sulfite species with Ba$^{2+}$ solutions while shifting the pH towards strongly basic conditions (NaOH addition) revealed a higher oxygen contribution from the experimental solution ($k_{\text{Ba}^{2+}} = 0.86 \pm 0.00$) than experiments where sulfite species were precipitated with Ag$^+$ solutions ($k_{\text{Ag}^+} = 0.57 \pm 0.03$). The relative oxygen contribution of the precipitation solution is close to zero in experiments precipitated with Ba$^{2+}$ ($m_{\text{Ba}^{2+}} = 0.03 \pm 0.01$), whereas the one for experiments precipitated with Ag$^+$ is similar to the experiment at pH 6.3 ($m_{\text{Ag}^+} = 0.43 \pm 0.04$). The small $m$-value for experiments precipitated with Ba$^{2+}$ while shifting the pH indicates that the oxygen exchange was efficiently stopped and we calculated an oxygen contribution from the contaminant of $d_{\text{Ba}^{2+}} = 0.11 \pm 0.00$, which is similar to the experiment at pH 6.3 where we did not shift the pH during precipitation.
The experiment at pH 9.7 where mainly SO\textsubscript{3}\textsuperscript{2−} prevails in solution were precipitated by adding Ba\textsuperscript{2+} without shifting the pH and the results (m\textsubscript{Ba\textsuperscript{2+}} = 0.04 ± 0.01, k\textsubscript{Ba\textsuperscript{2+}} = 0.77 ± 0.01 and d\textsubscript{Ba\textsuperscript{2+}} = 0.19 ± 0.01) were similar to the results of the experiments at pH 6.6 where we shifted the pH by addition of NaOH, but incorporated higher amounts of the contaminant source.

The experiments performed at pH 1.5 were precipitated by addition of NaOH and using Ba\textsuperscript{2+} as precipitation agent. The graphically determined values for k and the averaged m (Fig. 4) are 0.45 ± 0.01 and 0.40 ± 0.02, respectively, resulting in an estimate for d of 0.15 ± 0.02, which is comparable to our other experiments with BaSO\textsubscript{3} as precipitate. The value for m is much higher than the expected amount of oxygen from the conversion of SO\textsubscript{2} to HSO\textsubscript{3}−, which indicates that in the presence of SO\textsubscript{2} the pH shift by addition of NaOH during the precipitation cannot impede ongoing oxygen exchange between sulfite species and water.

4. DISCUSSION

In the first part of this section we discuss the appropriateness of different sulfite conversion/precipitation techniques. The second and third part of the section are devoted to the calculation of estimates for e\textsubscript{SO\textsubscript{2}/H\textsubscript{2}O} and e\textsubscript{SO\textsubscript{2}/H\textsubscript{2}O}, whereas the fourth part discusses implications of the newly determined oxygen isotope equilibrium fractionation factors on stable isotope signatures observed during active sulfur cycling.

4.1. Comparison between different precipitation agents (Ba\textsuperscript{2+} and Ag\textsuperscript{+}) and differences between precipitation at constant pH and with pH shift (with NaOH)

For our isotope mass balance approach it is of major interest to know how much oxygen isotope exchange occurs between sulfite species and water during the precipitation of sulfite salts. From the observation that experiments using

---

Table 2

Oxygen isotope data of sulfite–water equilibration experiments performed under different pH conditions.

<table>
<thead>
<tr>
<th>Experiment description</th>
<th>pH</th>
<th>δ\textsuperscript{18}O H\textsubscript{2}O\textsubscript{exp} (‰)</th>
<th>δ\textsuperscript{18}O H\textsubscript{2}O\textsubscript{prec} (‰)</th>
<th>δ\textsuperscript{18}O precipitate (‰)</th>
<th>Experiment description</th>
<th>pH</th>
<th>δ\textsuperscript{18}O H\textsubscript{2}O\textsubscript{exp} (‰)</th>
<th>δ\textsuperscript{18}O H\textsubscript{2}O\textsubscript{prec} (‰)</th>
<th>δ\textsuperscript{18}O precipitate (‰)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(1) Oxygen isotope values of experiments precipitated as BaSO\textsubscript{3} at constant pH (at 295°K)</td>
<td>6.31</td>
<td>−7.7</td>
<td>−0.5</td>
<td>0.5</td>
<td>(4) Oxygen isotope values of experiments precipitated as Ag\textsubscript{2}SO\textsubscript{3} at constant pH (at 295°K)</td>
<td>6.31</td>
<td>−7.7</td>
<td>−0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>(2) Oxygen isotope values of experiments precipitated as BaSO\textsubscript{3} with pH shift (at 295°K)</td>
<td>6.70</td>
<td>−7.7</td>
<td>3.1</td>
<td>0.1</td>
<td>(5) Oxygen isotope values of experiments precipitated as Ag\textsubscript{2}SO\textsubscript{3} at constant pH (at 295°K)</td>
<td>6.70</td>
<td>−7.7</td>
<td>3.1</td>
<td>0.1</td>
</tr>
<tr>
<td>(3) Oxygen isotope values of experiments precipitated as BaSO\textsubscript{3} at constant pH (at 295°K)</td>
<td>9.76</td>
<td>−7.7</td>
<td>1.8</td>
<td>0.5</td>
<td>(6) Oxygen isotope values of experiments precipitated as BaSO\textsubscript{3} with pH shift (at 295°K)</td>
<td>9.76</td>
<td>−7.7</td>
<td>1.8</td>
<td>0.5</td>
</tr>
</tbody>
</table>

---
Ag\(^+\) as precipitation agent yielded smaller oxygen contributions from the precipitation solution than using Ba\(^{2+}\); it is evident that much more oxygen isotope exchange between sulfite and water occurs during the precipitation of sulfite as Ag\(_2\)SO\(_3\) than during the precipitation of sulfite as Ag\(_2\)O (Fig. 5). This difference may be attributed to the higher solubility of BaSO\(_3\) compared to Ag\(_2\)SO\(_3\) (BaSO\(_3\): \(K_{sp} = 5.0 \times 10^{-10}\); Ag\(_2\)SO\(_3\): \(K_{sp} = 1.5 \times 10^{-14}\), at 25 °C from Lide, 1998), which might result in a more sluggish precipitation of BaSO\(_3\) than Ag\(_2\)SO\(_3\). Precipitation of sulfite species with Ba\(^{2+}\) without shifting the pH is thus not suitable for the determination of the oxygen isotope equilibrium fractionation between sulfite and water. However, the alternative, where BaSO\(_3\) was precipitated after shifting the pH of the sulfite solution to higher values, efficiently stopped the oxygen isotope exchange between sulfite and water. Unfortunately, this procedure cannot be used with silver solutions as silver hydroxide (AgOH) and silver oxide (AgO) would precipitate in alkaline solutions (Biedermann and Sillén, 1960). The comparison of the obtained \(m\) values from the precipitation of sulfite species at pH 6.6 (no SO\(_2\) species expected, Fig 1) shows that there is much less oxygen incorporation from the precipitation solution when the pH is shifted to strongly basic conditions and subsequent precipitation as BaSO\(_3\) salt compared to the direct precipitation with Ag\(^+\) (Fig. 6a). We hypothesize that the observed 3% oxygen contribution from the precipitation solution in the BaSO\(_3\) salt may represent entrained water in the barium precipitate rather than actual isotope exchange between sulfite and water during sulfite precipitation at high pH. Such lattice water was observed for barium sulfate, where a group of three water molecules can substitute for one molecule of barium sulfate and/or hydroxyl groups are occluded in the structure of the salt (Walton and Walden, 1946a, 1946b; Neagle and Rochester, 1988). Still, the small value for \(m\) demonstrates that the approach to precipitate sulfite by the combined use of NaOH/Ba\(^{2+}\), is a reliable procedure that immediately stops isotope exchange between sulfite species at circum-neutral to high pH values, and should allow for an accurate determination.

Table 3
Relative oxygen source contribution into the sulfite precipitates with two-step regression.

<table>
<thead>
<tr>
<th>Experiment</th>
<th>(H_2O_{prec}) oxygen contribution (m^*)</th>
<th>(H_2O_{exp}) oxygen contribution (k)</th>
<th>Contaminant oxygen contribution (d)</th>
<th>(\delta^{18}O) with (\delta^{18}O_{cont} = -28.7 \pm 3.6%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1) Ba(^{2+}), pH 6.3</td>
<td>0.90 ± 0.01</td>
<td>0.01 ± 0.01</td>
<td>0.09 ± 0.02</td>
<td>n.d.</td>
</tr>
<tr>
<td>2) Ba(^{2+}), pH 6.6, with pH shift</td>
<td>0.03 ± 0.01</td>
<td>0.86 ± 0.00</td>
<td>0.11 ± 0.00</td>
<td>15.2 ± 0.7%</td>
</tr>
<tr>
<td>3) Ba(^{2+}), pH 9.7</td>
<td>0.04 ± 0.01</td>
<td>0.77 ± 0.01</td>
<td>0.19 ± 0.01</td>
<td>15.2 ± 0.7%</td>
</tr>
<tr>
<td>4) Ag(^+), pH 6.3</td>
<td>0.48 ± 0.04</td>
<td>0.58 ± 0.04</td>
<td>−0.07 ± 0.06</td>
<td>10.9 ± 0.9%</td>
</tr>
<tr>
<td>5) Ag(^+), pH 6.6</td>
<td>0.43 ± 0.04</td>
<td>0.57 ± 0.03</td>
<td>−0.00 ± 0.05</td>
<td>10.9 ± 0.9%</td>
</tr>
<tr>
<td>6) Ba(^{2+}), pH 1.5, with pH shift</td>
<td>0.40 ± 0.02</td>
<td>0.45 ± 0.01</td>
<td>0.13 ± 0.02</td>
<td>n.d.</td>
</tr>
</tbody>
</table>

Relative oxygen source contribution determined with multiple regressions.

<table>
<thead>
<tr>
<th>Exp.</th>
<th>(m)</th>
<th>(k)</th>
<th>(d)</th>
<th>Intercept</th>
<th>(\delta^{18}O_{cont} = -28.7 \pm 3.6%)</th>
<th>(\delta^{18}O_{cont} = -27.5 \pm 0.3%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1)</td>
<td>0.90 ± 0.00</td>
<td>0.00 ± 0.01</td>
<td>0.11 ± 0.01</td>
<td>6.50 ± 0.40</td>
<td>n.d.</td>
<td></td>
</tr>
<tr>
<td>2)</td>
<td>0.03 ± 0.00</td>
<td>0.87 ± 0.01</td>
<td>0.10 ± 0.01</td>
<td>9.89 ± 0.26</td>
<td>14.7 ± 0.6%</td>
<td>14.5 ± 0.4%</td>
</tr>
<tr>
<td>3)</td>
<td>0.04 ± 0.01</td>
<td>0.79 ± 0.01</td>
<td>0.17 ± 0.02</td>
<td>5.95 ± 0.75</td>
<td>13.8 ± 1.4%</td>
<td>13.5 ± 1.2%</td>
</tr>
<tr>
<td>4)</td>
<td>0.48 ± 0.01</td>
<td>0.55 ± 0.02</td>
<td>−0.03 ± 0.02</td>
<td>10.92 ± 0.89</td>
<td>10.9 ± 0.9% (no contaminant)</td>
<td>10.9 ± 0.9% (no contaminant)</td>
</tr>
<tr>
<td>5)</td>
<td>0.43 ± 0.01</td>
<td>0.59 ± 0.02</td>
<td>−0.02 ± 0.02</td>
<td>11.11 ± 1.05</td>
<td>11.1 ± 1.1% (no contaminant)</td>
<td>11.1 ± 1.1% (no contaminant)</td>
</tr>
<tr>
<td>6)</td>
<td>0.40 ± 0.01</td>
<td>0.48 ± 0.01</td>
<td>0.12 ± 0.02</td>
<td>10.77 ± 0.74</td>
<td>n.d.</td>
<td></td>
</tr>
</tbody>
</table>

* \(m\)-value is the average of the 3 \(m\) values calculated for each of the 3 isotopically distinct \(H_2O_{exp}\) of the experiments with same pH. n.d.: not determined.
of the oxygen isotope equilibrium fractionation between sulfite species and the experimental solution. This is further supported by the comparison of the k values of the experiments at pH 6.6, which is the amount of water oxygen contribution in the sulfite salt derived from the experimental solution. The k value for experiments where sulfite species were precipitated with Ba\(^{2+}\) while shifting the pH indicated that approximately 86% of the oxygen in the sulfite salt is derived from the experimental solution, whereas only 57% of the oxygen in the silver precipitate is derived from the experimental solution (Fig. 6b). Using Ba\(^{2+}\) as precipitation agent with a pH shift provides the best results as it immediately terminates the oxygen exchange between sulfite species and water and thus the true oxygen isotope equilibrium fractionation between sulfite and water is retained in the BaSO\(_3\).

### 4.2. Oxygen isotope equilibrium fractionation between sulfite and water in barium sulfite precipitates BaSO\(_3\)

Experiments at pH 6.6 and 9.7 were chosen to determine \(\varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ}\) because under these conditions SO\(_2\) does not exist in solution and the parameters \(a_1\) and \(a_2\) in Eq. (11) become zero. It can be further assumed that the contribution of the precipitation solution is solely caused by entrainment of water into the salt crystal, and not due to isotope exchange between sulfite and water. This assumption is based on the observation that oxygen isotopic exchange between sulfite and water is slow at pH 9.7 and because NaOH was added to the experiment carried out at pH 6.6 to shift the pH to a high value during the precipitation of BaSO\(_3\). Consequently the parameter \(b_2\) becomes zero and \(c\) equals \(m\). Eq. (11) can thus be rewritten as:

\[
\begin{align*}
\delta^{18}O_{\text{prec}} &= 3 \cdot b_1 \cdot \left(\delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} + \varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ}\right) + c \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} \\
&\quad + d \cdot \delta^{18}O_{\text{cont}} \\
&= k \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} + m \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} + k \cdot \varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ} \\
&\quad + (1 - k - m) \cdot \delta^{18}O_{\text{cont}}
\end{align*}
\]

with:

\[
k = 3 \cdot b_1, \quad m = c, \quad d = 1 - (k + m) \quad (14)
\]

In the case that \(k\) would be equal to one and all oxygen in the precipitate would be derived from the experimental solutions the contribution of \(\delta^{18}O_{\text{cont}}\) would become zero and the constant equal to the equilibrium fractionation \(\varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ}\). In experiments at pH 6.6 and pH 9.7 \(k\) is less than one and at the same time slightly different (Table 3). Therefore, we can determine the values for \(\varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ}\) and \(\delta^{18}O_{\text{cont}}\) by comparing the results of both experiments graphically with each other. For the comparison, we rearrange Eq. (14) for the two experimental pH solutions, where for each pH all terms with the exception of the equilibrium fractionation and \(\delta^{18}O_{\text{cont}}\) are assumed to be constant:

\[
\begin{align*}
\left\{ \delta^{18}O_{\text{prec}} - k \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} - m \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} \right\}_{pH\,6.6} &\equiv \varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ} + \frac{\left\{1 - (k - m)\right\}_{pH\,6.6}}{\left\{k\right\}_{pH\,6.6}} \cdot \delta^{18}O_{\text{cont}} \\
&\equiv \varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ} + \frac{\left\{1 - (k - m)\right\}_{pH\,6.6}}{\left\{k\right\}_{pH\,6.6}} \cdot \delta^{18}O_{\text{cont}}
\end{align*}
\]

and

\[
\begin{align*}
\left\{ \delta^{18}O_{\text{prec}} - k \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} - m \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} \right\}_{pH\,9.7} &\equiv \varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ} + \frac{\left\{1 - (k - m)\right\}_{pH\,9.7}}{\left\{k\right\}_{pH\,9.7}} \cdot \delta^{18}O_{\text{cont}} \\
&\equiv \varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ} + \frac{\left\{1 - (k - m)\right\}_{pH\,9.7}}{\left\{k\right\}_{pH\,9.7}} \cdot \delta^{18}O_{\text{cont}} \quad (15)
\end{align*}
\]

By plotting the results of the three isotopically distinct experimental solutions of two pH experiments on the same plot, we can draw a regression line through all the points and extrapolate the line to the intercept with the \(y\) axis where \(k\) is equal to one and as consequence \(d = 0\). Thus, the values of the two unknowns can be determined graphically where \(\varepsilon_{SO_3^{2-} \rightarrow H_2O}^{EQ}\) corresponds to the intercept of the regression line with the \(y\) axis and \(\delta^{18}O_{\text{cont}}\) corresponds to
the slope of the regression line (Fig. 7). The value for the oxygen isotope equilibrium fractionation between sulfite and water is \( \delta^{18}O_{SO_3^{--}H_2O} = 15.2 \pm 0.7\%_o \) and the value of the oxygen isotope composition of the contaminant is \( \delta^{18}O_{cont} = -28.7 \pm 3.6\%_o \) (see Fig. 7). As shown below, the oxygen isotope composition of the contaminant is almost identical to values of water vapor measured in the laboratory. We note that without the additional credibility lent by this finding, our estimate for the isotope composition of the contaminant and \( \delta^{18}O_{SO_3^{--}H_2O} \) would hinge on an extrapolation from two clusters of data. Future experimental approaches that either minimize the contribution of the contaminant (i.e., addition of data sets to improve reliability of extrapolation of regression) or independently determine the isotope composition of the contaminant (e.g., by controlling isotope composition of water vapor in laboratory) are needed to better constrain the estimate for \( \delta^{18}O_{SO_3^{--}H_2O} \).

4.2.1. Isotope composition of water vapor in the laboratory

The oxygen isotope composition of the collected water (\( \delta^{18}O_{vapor} \)) was \(-27.5 \pm 0.3\%_o \) which is close to the value obtained for the contaminant in our BaSO₄ precipitates (\( \delta^{18}O_{cont} = -28.7 \pm 3.6\%_o \)) and underlines that water vapor is likely the major contaminant during the post-precipitation steps in the experimental procedure. Using the estimate for the isotope composition of \(-28.7 \pm 3.6\%_o \) for the contaminant for the determination of \( \delta^{18}O_{SO_3^{--}H_2O} \) with a multiple linear regression for the experiment at pH 6.6 and pH 9.7, we find values of 14.7 \pm 0.6\%_o and 13.8 \pm 1.4\%_o. Using an estimate of \(-27.5 \pm 0.3\%_o \) for the contaminant, one obtains slightly smaller values of 14.5 \pm 0.4\%_o and 13.5 \pm 1.2\%_o for the experiment at pH 6.6 and 9.7, respectively (Table 3). The uncertainties derived with the multiple linear regression approach are smaller than the uncertainties from the two-step linear regression approach via Fig. 7, as the multiple regression approach does not consider slight deviations in the \( m \) values determined for the three distinct experimental solutions at the same pH. In the two-step regression approach to determine the equilibrium values we accounted for these slight deviations of \( m \) with the consequence that we obtain slightly higher error ranges, however, the values should be more trustworthy. Table 3 displays the values of \( m, k, d \) and \( c \) from the two-step regression approach in the upper half and in the lower half the same parameters determined via a multiple regression approach. Additionally, we displayed the calculated intercept and the \( c \) values corrected for the contaminant using the \( \delta^{18}O \) derived in Fig. 7 and the \( \delta^{18}O \) measured for the vapor, respectively. The deviations between the two approaches highlight the need for additional experimental work for a higher accuracy in the estimate of the value for the oxygen isotope equilibrium fractionation between sulfite and water.

4.2.2. Comparison of the oxygen isotope equilibrium fractionations between sulfite and water determined from silver sulfite precipitates

The experiments at pH 6.3 and 6.6 where sulfite was precipitated as Ag₂SO₃ through the addition of AgNO₃ can be used to obtain another estimate for \( \delta^{18}O_{SO_3^{--}H_2O} \) which is independent of the precipitation technique for BaSO₄. The estimate for \( \delta^{18}O_{SO_3^{--}H_2O} \) based on Ag₂SO₃ precipitates has to be considered less reliable than the values obtained by shifting the pH with NaOH and precipitation as BaSO₄, as ongoing oxygen isotope exchange between sulfite and water during the precipitation of Ag₂SO₃ can obscure kinetic isotope effects (i.e., preferred precipitation of isotopically light/heavy sulfite species). Still, the value for \( \delta^{18}O_{SO_3^{--}H_2O} \) by the Ag₂SO₃ method should not deviate strongly from the value obtained with BaSO₄ because a large kinetic isotope effect during the precipitation of Ag₂SO₃ is not expected (i.e., no chemical bonds are broken). We can simplify Eq. (11) by assuming that no SO₂ exists at pH 6.3 and 6.6 (Fig. 1) and by considering the entrainment of precipitation water into Ag₂SO₃ precipitates minor compared to the portion of the oxygen in Ag₂SO₃ that was contributed by oxygen isotope exchange between sulfite and water during the precipitation (i.e., \( c = 0 \)):

\[
\delta^{18}O_{prec} = 3 \cdot b_1 \left( \delta^{18}O_{H_2O_{exp}} + \delta^{18}O_{SO_3^{--}H_2O} \right) + 3 \cdot b_2 \left( \delta^{18}O_{H_2O_{exp}} + \delta^{18}O_{SO_3^{--}H_2O} \right) + d \cdot \delta^{18}O_{cont}
\]

\[
= k \cdot \delta^{18}O_{H_2O_{exp}} + m \cdot \delta^{18}O_{H_2O_{exp}} + \left( k + m \right) \cdot \delta^{18}O_{SO_3^{--}H_2O} + d \cdot \delta^{18}O_{cont}
\]

with:

\[
k = 3 \cdot b_1
\]

\[
m = 3 \cdot b_2
\]

\[
d = 1 - \left( k + m \right)
\]

The small values for \( d \) (sum of \( k \) and \( m \) close to 1) show that, unlike for BaSO₄, there is no contaminant in Ag₂SO₃. This allows us to set \( d = 0 \) in Eq. (16):

\[
\delta^{18}O_{prec} = m \cdot \delta^{18}O_{H_2O_{exp}} = k \cdot \delta^{18}O_{H_2O_{exp}} + \delta^{18}O_{SO_3^{--}H_2O}
\]

Eq. (17) can be used to graphically determine the value for \( \delta^{18}O_{SO_3^{--}H_2O} \) of 10.9 \pm 0.9\%_o (Fig. 8). This value obtained from Ag₂SO₃ precipitates is approximately 4.3\%_o lighter.
Fig. 8. Graphical determination of the oxygen isotope equilibrium fractionation between sulfite and water from AgSO₃ precipitates from experiments at pH 6.3 and 6.6. In case of the experiments with silver, there is no contamination with water vapor; therefore, the oxygen isotope equilibrium fractionation between sulfite and water can be determined graphically after Eq. (17) where δ¹⁸O_H₂O = 0.

than the value from the BaSO₃ (15.2 ± 0.7‰). This lighter value is likely due to normal kinetic isotope effects (isotopically light sulfite species are preferred) during precipitation of sulfite which are obscured by the ongoing oxygen exchange of the dissolved sulfite with the precipitation solution. Analogous kinetic oxygen isotope fractionation effects have been reported for the rapid precipitation of carbonate minerals (Kim et al., 2006). Another explanation for the smaller equilibrium fractionation could be co-precipitation of AgNO₃ within the Ag₂SO₃. However, this is rather improbable as nitrate would leave a signal in the contaminant term of our isotope mass balance approach because direct oxygen exchange between NO₃⁻ and water is extremely slow (Hall and Alexander, 1940; Bunton et al., 1952; Gamsjäger and Murmann, 1983), but we did not observe a contamination source in our silver precipitates. Furthermore, no nitrogen peaks (N₂) were detected during the isotope analyses by continuous flow mass spectrometry where carbon monoxide and N₂ are separated by gas chromatography. This observation rules out significant contamination with nitrate.

4.3. Determination of the oxygen isotope equilibrium fractionation between dissolved SO₂ and water from experiments at pH 1.5

In the experiments at pH 1.5 the amounts of HSO₃⁻ and SO₂ in solution are approximately identical (Fig. 1). For the precipitation of BaSO₃, NaOH was used to rapidly shift the pH of the experimental solution to higher values where no oxygen isotope exchange between sulfite and water occurs. From the experiments carried out at pH 6.6, it is known that this approach is likely effective for sulfite species such as HSO₃⁻, SO₃²⁻ and SO₂(H₂)³⁻. From this we deduce that our precipitation technique will suppress isotope exchange between precipitation water and those species (i.e. h₂ = 0), however we cannot infer that this is also the case for SO₂. Therefore, Eq. (11) can only be slightly simplified:

\[ \delta^{18}O_{\text{prec}} = k \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} + m \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{res}}} + 2 \cdot (a_1 + a_2) \cdot \epsilon_{\text{SO}_3^{3-} \rightarrow \text{H}_2\text{O}} + (a_1 + a_2) \cdot \epsilon_{\text{H}_2\text{O} \rightarrow \text{SO}_3^{3-}} + 3 \cdot b_1 \]

\[ \epsilon_{\text{SO}_3^{3-} \rightarrow \text{H}_2\text{O}} + d \cdot \delta^{18}O_{\text{cont}} \]

with:

\[ k = 2 \cdot a_1 + 3 \cdot b_1 \]

\[ m = a_1 + 3 \cdot a_2 + c \]

\[ d = 1 - (k - m) \]

The graphically determined values for k and the averaged m (Fig. 4) are 0.45 ± 0.01 and 0.40 ± 0.02, respectively, resulting in an estimate for d of 0.15 (15% contaminant), which is comparable to our other experiments with BaSO₃ as precipitate. In order to solve Eq. (18), the results from the experiments a pH 6.6 and pH 9.7 can be used as estimates (i.e. c = 0.03, \( \delta^{18}O_{\text{cont}} = -28.7 ± 3.6\)‰, \( \epsilon_{\text{SO}_3^{3-} \rightarrow \text{H}_2\text{O}} = 15.2 ± 0.7\)‰). We can explore the ratio between HSO₃⁻ and SO₂ for the case that the entire SO₂ pool exchanged its oxygen with water during the precipitation, i.e. where \( a_1 = 0 \). The equations for k and m from above are simplified (\( k = 3 \cdot b_1 \) and \( m = 3 \cdot a_2 + c \)) and the ratio between HSO₃⁻ and SO₂ becomes:

\[ \frac{k}{m-c} = \frac{0.45}{0.37} \]

This ratio (55% HSO₃⁻ to 45% SO₂ when normalized to 100%) corresponds to a pH of 1.3 in the pH dependent species distribution of sulfite species (Fig. 1), which is close to the measured pH in the experimental solution of 1.5. If we were to choose a slightly higher ratio between HSO₃⁻ and SO₂, that is closer to the predicted ratio at pH 1.5 (Fig. 1) the value for \( a_1 \) would become negative, which is impossible. Choosing a lower ratio between HSO₃⁻ and SO₂ would yield values for \( a_1 \) that are larger than zero, but would require a pH lower than 1.3, thus even further away from the measured pH of 1.5. We attribute the difference between the pH estimate derived from Eq. (19) and the actually measured pH to uncertainties in the equilibrium constants used in the sulfite species distribution. These uncertainties in the equilibrium constants result in a large uncertainty for the ratio between HSO₃⁻ and SO₂ because concentration gradients are very steep in this pH range (Fig. 1). Therefore, it can be concluded that almost the entire pool of SO₂ exchanged oxygen isotopes with the solution during the precipitation of BaSO₃, confirming that isotope exchange at very low pH via Eq. (1) is extremely rapid and implying that it cannot be immediately stopped by addition of NaOH. Assuming that \( a_1 = 0 \) (i.e. \( b_1 = k/3 \), \( a_2 = (m-c)/3 \)), the unknown parameters in Eq. (18) can be separated:

\[ \frac{\delta^{18}O_{\text{prec}} - m \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} - k \cdot \epsilon_{\text{SO}_3^{3-} \rightarrow \text{H}_2\text{O}} - d \cdot \delta^{18}O_{\text{cont}}}{(m-c)/3} = \]

\[ = \frac{3 \cdot k}{m-c} \cdot \delta^{18}O_{\text{H}_2\text{O}_{\text{exp}}} + 2 \cdot \epsilon_{\text{SO}_3^{3-} \rightarrow \text{H}_2\text{O}} + \epsilon_{\text{H}_2\text{O} \rightarrow \text{SO}_3^{3-}} \]
The value of \(2 \cdot \frac{\Delta^{18}O_{SO_2-H_2O}}{\Delta^{18}O_{SO_2-HSO_3^-}}\) can be determined graphically \((74.0 \pm 1.7 \text{‰}, \text{Fig. 9})\). We cannot disentangle the potential kinetic isotope fractionation that oxygen undergoes, which is incorporated from the precipitation solution during the conversion of SO\(_2\) to HSO\(_3^-\) \((\varepsilon_{SO_2:SO_3} = \varepsilon_{SO_2:HSO_3^-})\), from the equilibrium isotope fractionation between SO\(_2\) and H\(_2\)O \((\varepsilon_{SO_2:H_2O})\). Furthermore, we have to acknowledge that – analogous to the precipitation experiments with silver nitrate – a kinetic isotope effect could be imposed on the oxygen isotope composition of SO\(_2\) that is being converted to HSO\(_3^-\) \((\varepsilon_{SO_2:SO_3} = \varepsilon_{SO_2:HSO_3^-})\) that cannot be excluded from our isotope mass balance. To obtain at least a rough estimate of \(\varepsilon_{SO_2:H_2O}\) it can be assumed that these kinetic isotope fractionations are small compared to the equilibrium isotope fractionation and can be set equal to zero. With these assumptions, the calculated oxygen isotope equilibrium fractionation between dissolved SO\(_2\) and water is approximately 37.0‰. Taking into account the uncertainties that come along with the unknown kinetic isotope effect for the conversion of SO\(_2\) to HSO\(_3^-\), the rough estimate of \(\varepsilon_{SO_2:H_2O}\) is displayed without an error estimate. We can compare this value for \(\frac{\Delta^{18}O_{SO_2-H_2O}}{\Delta^{18}O_{SO_2-HSO_3^-}}\) to the study on oxygen isotope fractionation on the oxygen exchange between gaseous SO\(_2\) and water vapor by Holt et al. (1983), who observed an isotope fractionation of approximately 24‰ at 22 °C. Our value for the oxygen isotope equilibrium fractionation between aqueous SO\(_2\) and water is approximately 13‰ higher than the equilibrium fractionation for gaseous SO\(_2\) and water vapor. This difference between the liquid phase equilibrium fractionation and gas phase equilibrium fractionation is very large. For a comparison, at equilibrium at 25 °C, aqueous carbon dioxide (CO\(_2\)) is enriched in \(^{18}O\) by approximately 1‰ compared to gaseous CO\(_2\) (calculated from Bottinga, 1968 and Brenninkmeijer et al., 1983), and aqueous ammonia (NH\(_3\)) is enriched in \(^{15}N\) by approximately 7.2‰ to 15.3‰ compared to gaseous NH\(_3\) (calculated from and Scaln, 1958; Hanschmann, 1981 and Urey, 1947; for a review see Li et al., 2012). The apparent difference between the \(\frac{\Delta^{18}O_{SO_2-H_2O}}{\Delta^{18}O_{SO_2-HSO_3^-}}\) for gaseous and aqueous phase even exceeds the difference of liquid gas fractionation for the oxygen isotope composition of water with approximately 9.5‰ at 22 °C (Horita and Wesolowski, 1994). If we were to use smaller values for the [HSO\(_3^-\)]/[SO\(_3^2-\)] ratio (which would imply a pH-dependent species distribution that differs strongly from our calculations, see Fig. 1), we would obtain only slightly smaller values for \(\frac{\Delta^{18}O_{SO_2-H_2O}}{\Delta^{18}O_{SO_2-HSO_3^-}}\) that would not resolve the discrepancy between our findings and the value determined by Holt et al. (1983). Ignoring the large uncertainties involved, it might be possible that a part of this discrepancy is due to a higher stability of aqueous SO\(_2\) compared to gaseous SO\(_2\) as there is a strong association of hydrated SO\(_2\) with a high abundance of water molecules, forming stronger hydrogen bonds than hydrated CO\(_2\) (Moin et al., 2011) or it can be attributed to the aforementioned kinetic isotope effects. However, in the case of the latter, one needs to consider that inverse kinetic isotope effects (preferred use of H\(_2\)O and SO\(_3^2-\) enriched in \(^{18}O\)) would need to be postulated to explain the difference between our findings and the results by Holt et al. (1983).

### 4.4. Implications of the obtained oxygen isotope equilibrium fractionation values

Oxygen isotope exchange between sulfate and water is often considered to have a major impact on the oxygen isotope signatures of sulfate affected by oxidative and reductive sulfur cycling. We were able to determine the oxygen isotope equilibrium fractionation between sulfate (HSO\(_3^-\), SO\(_3^2-\), NaSO\(_3^-\)) and water of \(\frac{\Delta^{18}O_{SO_3^-:H_2O}}{\Delta^{18}O_{SO_2:HSO_3^-}} = 15.2 \pm 0.7\text{‰}\) and also to provide a rough estimate for the oxygen isotope fractionation between dissolved sulfate dioxide and water \(\varepsilon_{SO_2:HSO_3^-:H_2O} = 37.0\text{‰}\). These values are of importance for a better understanding of the oxygen isotope signature in residual sulfate during dissimilatory sulfate reduction (DSR) as well as for the understanding of the oxygen isotope signatures in sulfate produced during oxidative processes.

During DSR, where sulfate is reduced via intermediates such as adenosine 5′-phosphosulfate (APS) and sulfite to sulfide, the oxygen isotope composition in the residual sulfate is dependent on the oxygen isotope composition of the surrounding water (Mizutani and Rafter, 1973) despite the fact that sulfate does not directly exchange its oxygen with the water (Lloyd, 1967; Chiba and Sakai, 1985). This observation is generally explained as a result of DSR-mediated equilibrium oxygen isotope fractionation between residual sulfate and the water driving the oxygen isotope composition of sulfate towards a constant value of approximately 23‰ (Zeebe, 2010) to 26‰ (Fritz et al., 1989) at 22 °C. This mechanism was intensively investigated both by experimental studies as well as modeling approaches (Fritz et al., 1989; Brunner and Bernasconi, 2005; Brunner et al., 2005, 2012; Blake et al., 2006; Wortmann et al., 2007; Turchyn et al., 2010). It is assumed that the oxygen isotope
fractionation observed during DSR results from reversible enzymatic steps during sulfate reduction, enabling the oxidation of sulfite intermediates that rapidly exchange oxygen isotopes with ambient water back to sulfate. Sulfite is considered to be an important intermediate in DSR and is known to rapidly exchange oxygen isotopes with water, unlike the more oxidized sulfuryl anions in DSR, such as APS or cell-internal sulfate (Brunner et al., 2012; Kohl et al., 2012). The value of 15.2% obtained for $^{18}O_{SO_2}$ is much smaller than the estimates for DSR-mediated SO$_3$ equilibrium between sulfate and water. Oxygen isotope exchange between sulfite and water alone therefore cannot be responsible for the oxygen isotope fractionation during DSR and highlights the importance of the isotope effects related to the back oxidation of sulfite to APS, which involves the incorporation of oxygen from adenosine monophosphate (AMP) (Peck, 1962; Peck and Stulberg, 1962; Fritz et al., 2002; Wortmann et al., 2007; Brunner et al., 2012).

However, our finding that there may be a much higher oxygen isotope equilibrium fractionation between SO$_2$ and water of $^{18}O_{SO_2} ightarrow H_2O = 37.0$% adds a slight twist to these considerations, as it highlights that different sulfite species may have strongly different isotope equilibrium values. In this respect, S$_2$O$_5^{2-}$ is of interest, because, unlike SO$_3$, it can exist at circum-neutral conditions that are physiologically relevant. As S$_2$O$_5^{2-}$ existed only in extremely small quantities in our experiments, we could not determine an equilibrium isotope fractionation for this species. In biological systems, low abundance of a species does not imply its unimportance, and it is interesting to note that S$_2$O$_5^{2-}$ has been considered as a potential alternative to SO$_3^{2-}$ in DSR (Woolfolk, 1962).

Our results will also be useful for the study of oxidative sulfur cycling of reduced sulfur compounds, which operate depending on environmental conditions, such as pH, over various abiotic and biochemical pathways with a multitude of distinct sulfoxy intermediates. Many of these pathways have in common that in a chain of oxidation steps sulfite is the final sulfoxy intermediate before the oxidation to the final product sulfate. If sulfite is the final sulfoxy intermediate during the oxidation of reduced sulfur compounds, rapid oxygen exchange between sulfite species and water should impact the oxygen isotope composition of the produced sulfate. To fully express oxygen isotope equilibrium fractionations with respect to the water, sulfite species require enough time for oxygen exchange which is further dependent on the oxygen rate of sulfate to sulfite. Studies on the oxidation of reduced sulfur compounds explained larger oxygen isotope offsets between the produced sulfate and water with the presence of sulfite as final sulfoxy intermediate and smaller offsets being produced from other oxidation pathways involving distinct sulfoxy intermediates (e.g. Hubbard et al., 2009; Kohl and Bao, 2011; Balci et al., 2012; Heidel et al., 2013). These studies show that further research has to be performed on the mechanisms of sulfite oxidation as kinetic isotope effects might play a significant role on the final oxygen isotope composition of the sulfate (Müller et al., 2013). Knowledge of the effective value of $^{18}O_{SO_3} ightarrow H_2O$ and having a rough estimate of $^{18}O_{S_2O_5}^{2-}$ will enable a more quantitative interpretation of the oxygen isotope signature of sulfate produced from oxidative sulfur cycling.

**5. CONCLUSIONS**

We performed experiments with dissolved sulfite species to determine the oxygen isotope equilibrium fractionation factor between dissolved sulfite and water. The experimentally most feasible way to determine the oxygen isotope equilibrium values was to add NaOH which shifts the pH to values higher than 12, thereby overcoming the challenges of ongoing oxygen isotope exchange between sulfite and water during the precipitation of sulfate salts. This procedure, however, comes at a price; precipitated BaSO$_4$ salts are strongly hygroscopic and the effects of the contamination through uptake of water vapor have to be resolved.
by mathematical treatments and corrections after data generation. Silver sulfite salts do not show this hygroscopic behavior, but silver nitrate solutions cannot be used at high pH because of co-precipitation of AgOH with Ag$_2$SO$_3$. The methods tested and applied in our research are transferable to the determination of the oxygen isotope equilibrium fractionation between water and oxyanions other than sulfite, where similar challenges as for the precipitation of sulfite for oxygen isotope analysis exist, e.g. nitrite, or selenite.

We determined a rough estimate for the oxygen isotope equilibrium fractionation between SO$_3$ and water at low pH of $\epsilon_{SO_3-H_2O} = 37.0\%$ and a more firm estimate for the oxygen isotope equilibrium between SO$_3^{2-}$ and water at circum neutral pH of $\epsilon_{SO_3^{2-},H_2O} = 15.2 \pm 0.7\%$. With increasing pH, HSO$_3^{-}$ and SO$_2^{4-}$ become more abundant. As SO$_3$ disappears from the sulfite system, the overall oxygen exchange between water and sulfite is still rapid, but not as rapid as in the presence of SO$_2$. At very high pH ($>12$) only SO$_2^{4-}$ and some Na$_2$SO$_3$ will be present in solution, as consequence oxygen isotope exchange between sulfite and water is extremely slow. Under these conditions, oxygen isotope disequilibrium between sulfite and water will be retained (Fig. 10).

ACKNOWLEDGMENTS

The authors thank T.G. Ferdelman and M.M.M. Kuypers for supporting this study, T. Max for crucial help with the mass spectrometer and assistance in the laboratory. We thank S.M. Bernasconi for the isotopic analysis and support as well as A.V. Turchyn for inspiring discussions and valuable advice. This project would not have been possible without the financial support provided by MARUM (Center for Marine Environmental Sciences) and the Max Planck Society. We would like to acknowledge K.W. Mandernack, B.A. Wing, the anonymous reviewer and the associate editor E.A. Schauble for their detailed, insightful comments and suggestions that greatly helped to improve our manuscript. The contribution of M.C. was carried out at the Jet Propulsion Laboratory (JPL), California Institute of Technology, under contract with the National Aeronautics and Space Administration (NASA), with support from the NASA Astrobiology Institute (NAI-WARC).

REFERENCES


*Associate editor:* Edwin Schauble