# A simple and reliable method reducing sulfate to sulfide for multiple sulfur isotope analysis

Lei Geng, Joel Savarino, Clara A Savarino, Nicolas Caillon, Pierre Cartigny,
Shohei Hattori, Sakiko Ishino, | Naohiro Yoshida

**RESEARCH ARTICLE**

WILEY Rapid Communications in Mass Spectrometry

# A simple and reliable method reducing sulfate to sulfide for multiple sulfur isotope analysis

Lei Geng[1] | Joel Savarino[1] | Clara A. Savarino[1,2] | Nicolas Caillon[1] | Pierre Cartigny[3] | Shohei Hattori[4] | Sakiko Ishino[4] | Naohiro Yoshida[4,5]

[1] Univ. Grenoble Alpes, CNRS, IRD, Institut des Géosciences de l'Environnement, IGE, 38000 Grenoble, France

[2] School of Chemistry, Cardiff University, Cardiff CF10 3AT, UK

[3] Laboratoire de Géochimie des Isotopes Stables, Institut de Physique du Globe de Paris, Sorbonne Paris Cité, Univ. Paris Diderot, UMR 7154 CNRS, 75005 Paris, France

[4] School of Materials and Chemical Technology, Tokyo Institute of Technology, 226-8502 Yokohama, Japan

[5] Earth-Life Science Institute, Tokyo Institute of Technology, 152-8551 Tokyo, Japan

**Correspondence**
J. Savarino, Univ. Grenoble Alpes, CNRS, IRD, Institut des Géosciences de l'Environnement, IGE, 38000 Grenoble, France.
Email: joel.savarino@cnrs.fr

L. Geng, Anhui Province Key Laboratory of Polar Environment and Global Change, School of Earth and Space Sciences, University of Science and Technology of China, Hefei, Anhui 230026, China.
Email: genglei@ustc.edu.cn

**Present Address**
Lei Geng, Anhui Province Key Laboratory of Polar Environment and Global Change, School of Earth and Space Sciences, University of Science and Technology of China, Hefei, Anhui 230026, China.

**Rationale:** Precise analysis of four sulfur isotopes of sulfate in geological and environmental samples provides the means to extract unique information in wide geological contexts. Reduction of sulfate to sulfide is the first step to access such information. The conventional reduction method suffers from a cumbersome distillation system, long reaction time and large volume of the reducing solution. We present a new and simple method enabling the process of multiple samples at one time with a much reduced volume of reducing solution.

**Methods:** One mL of reducing solution made of HI and $NaH_2PO_2$ was added to a septum glass tube with dry sulfate. The tube was heated at 124°C and the produced $H_2S$ was purged with inert gas (He or $N_2$) through gas-washing tubes and then collected by NaOH solution. The collected $H_2S$ was converted into $Ag_2S$ by adding $AgNO_3$ solution and the co-precipitated $Ag_2O$ was removed by adding a few drops of concentrated $HNO_3$.

**Results:** Within 2–3 h, a 100% yield was observed for samples with 0.2–2.5 µmol $Na_2SO_4$. The reduction rate was much slower for $BaSO_4$ and a complete reduction was not observed. International sulfur reference materials, NBS-127, SO-5 and SO-6, were processed with this method, and the measured against accepted $\delta^{34}S$ values yielded a linear regression line which had a slope of 0.99 ± 0.01 and a $R^2$ value of 0.998.

**Conclusions:** The new methodology is easy to handle and allows us to process multiple samples at a time. It has also demonstrated good reproducibility in terms of $H_2S$ yield and for further isotope analysis. It is thus a good alternative to the conventional manual method, especially when processing samples with limited amount of sulfate available.

# 1 | INTRODUCTION

Stable sulfur isotopes have been widely used to trace a range of biogeochemical processes.[1] The discovery in 2000 of the mass-independent isotopic fractionations of sulfur isotopes (S-MIF) in sulfate and sulfide in Archean rocks[2] showed the potential of the S-MIF signals for tracking the oxygenation of the atmosphere 2.4 Gy ago,[3] and the geochemical evolution of Mars.[4] The S-MIF signals in ice-core sulfate have also been observed and demonstrated to be useful for tracking the sulfur cycle in today's stratosphere and they serve as a unique proxy of large volcanic eruptions that inject sulfur into the stratosphere and thus have global climate impacts.[5-8] Multiple sulfur isotope compositions can also help to constrain the oceanic sulfur cycle (e.g., [9,10]).

To access the S-MIF signals, precise analysis of the four sulfur isotopes ($^{32}S$, $^{33}S$, $^{34}S$ and $^{36}S$) is necessary. The isotopic results are expressed as $\delta^{3x}S = {}^{3x}R_{sample}/{}^{3x}R_{CDT} - 1$, where x = 3, 4, and 6, and the δ values are calculated using the CDT standard. The S-MIF values are then defined by:

$$\Delta^{33}S = \delta^{33}S - \left[\left(\delta^{34}S + 1\right)^{0.515} - 1\right]$$

$$\Delta^{36}S = \delta^{33}S - \left[\left(\delta^{34}S + 1\right)^{1.90} - 1\right]$$

The isotopic analysis is conventionally performed by reducing sulfate ($SO_4^{2-}$) to hydrogen sulfide ($H_2S$), converting $H_2S$ into silver sulfide ($Ag_2S$), and fluorinating $Ag_2S$ to sulfur hexafluoride ($SF_6$) for isotopic composition analysis by isotope ratio mass spectrometry (IRMS).[2,6,11,12] The reduction from $SO_4^{2-}$ to $H_2S$ is mainly achieved by two different reducing agents: tin(II) ($Sn^{2+}$) solutions and hydroiodic acid (HI)/hypophosphorous acid ($H_3PO_2$) mixtures.[13-15] The $Sn^{2+}$ solution is mainly applied to solid samples (e.g., minerals) with an optimum reaction temperature between 280 and 300°C, and the HI reducing solution can be applied to aqueous samples at 100–125°C.[14] Currently, the most widely used reducing method in sulfur isotope geochemistry follows the reducing agent recipe (500 mL concentrated HI, 816 mL concentrated HCl, and 245 mL 50% $H_3PO_2$) of Thode et al.,[16] and uses a distillation apparatus similar to that described in Forrest and Newman.[17]

In the reducing solution of Thode et al,[16] high concentrations of HI seem to be the most important component of the reducing agent for complete sulfate reduction, and the presence of $H_3PO_2$ or $NaH_2PO_2$ increases the reduction speed by maintaining a high hydroiodic acid to iodine ratio which is one of the factors favoring the reduction.[14,18] HCl is only of secondary importance and its presence is suggested to increase the acidity and volume, and reduce the use of relatively expensive HI.[13,19] However, Gustafsson[20] found the presence of water to be detrimental for the reduction because water tends to dilute and thus lower the concentration of HI, and at lower HI concentration, side products (viz, $SO_2$ and elemental S) will be formed.[18] In this regard, mixing 50% $H_3PO_2$ and concentrated HCl with the concentrated HI may have drawbacks for the reduction efficiency, because both $H_3PO_2$ (50%) and concentrated HCl (37%) contain more than 50% water by weight. To avoid additional water in the reducing solution, the $H_3PO_2$ can be replaced with dry $NaH_2PO_2$ salt, and HCl can be omitted. Gustafsson[20] and Davis and Lindstrom[18] have used a reducing solution containing only HI (57%) and $NaH_2PO_2$ salt, and found a good reduction yield. In particular, Davis and Lindstrom[18] found that the optimum composition of the reducing solution for complete and fast sulfate reduction is 0.13 g $NaH_2PO_2$ in 1 mL HI (57%). In these studies, aqueous sulfate samples were processed and a cumbersome distillation apparatus was used.

In summary, it seems that the best composition of the reducing solution would be a mixture of 0.13 g $NaH_2PO_2$ in 1 mL HI (57%), and the amount of water in the reduction experiment should be limited. The latter requirement suggests dry sulfate samples are a better choice as the starting material. Typically, barium sulfate ($BaSO_4$) is the preferred sulfate form for the four-sulfur isotopes analysis because it is the natural form found in major geological samples or can be readily prepared from natural samples containing soluble sulfate (e.g., sea water) by precipitation with excess $BaCl_2$ solution. $BaSO_4$ has very low solubility (≈ 0.02 mg/L at 20°C) and this may inhibit the reaction efficiency and speed, especially when the volume of the reducing solution is small. Alternatively, soluble sulfate in natural samples can also be extracted and purified by other methods such as using an ion-exchange resin[21] and this can yield dry $Na_2SO_4$ by evaporating the eluent. We thus conducted tests with both $BaSO_4$ and $Na_2SO_4$ to explore the reaction efficiency of the reduction process with respect to different sulfate forms. In this report, we present a series of experiments where we used a reducing solution comprising $NaH_2PO_2$ and HI (57% by weight) to process dry sulfate samples (both $Na_2SO_4$ and $BaSO_4$). To avoid the cumbersome distillation apparatus, we tested a simple flow system with only sealed glass tubes connected by PEEK tubes and explored the possibility of processing multiple samples at one time. The reproducibility for $H_2S$ yield and for further sulfur isotope analysis is reported.

# 2 | EXPERIMENTAL

## 2.1 | Reagents

The new reducing solution was made of 100 mL concentrated hydriodic acid (HI, 57% by weight) and 13 g sodium hypophosphite ($NaH_2PO_2$). The reducing solution was prepared in a fume hood, where 100 mL HI and 13 g $NaH_2PO_2$ were added to a flask. The flask was placed on a hot plate magnetic stirrer and a magnetic stir bar was used to mix the liquid and the salt. Because HI is easily oxidized by $O_2$, helium (He) or another inert gas stream (e.g., $N_2$) was introduced by a PEEK tube into the flask to purge the mixture. While purging with He, the hot plate temperature was set at 130°C. The flask was heated at 130°C for at least 1 h to reduce any sulfur compounds into $H_2S$ (that was flushed away from the reagents) and to reduce traces of $I_2$ (in the form of $I_3^-$) into $I^-$ by $NaH_2PO_2$. The solution started with a deep color ($I_3^-$) and became colorless with time. After being heated for 1 h, the solution was allowed to cool down under the He stream and then stored in a sealed brown bottle. The reducing solution may become oxidized over time; this is indicated by a light yellow color, which may become deeper depending on the degree of oxidation.

Different from what can be found in the literature, in this study we used sodium hydroxide (NaOH, 0.1 M) as the trapping solution to
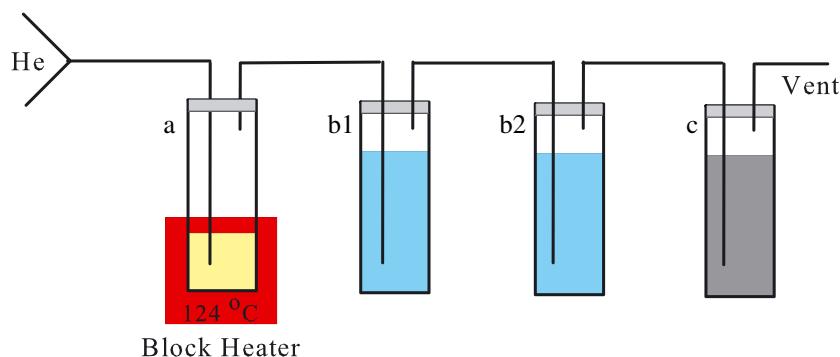
**FIGURE 1** Sketch of the reduction train. a: block heater and the reduction tube; b1 & b2: gas washing tubes; c: $H_2S$ collection tube [Color figure can be viewed at wileyonlinelibrary.com]

collect the reduction product $H_2S$. Conventional trapping solutions, cadmium (or zinc) acetate ($Cd(CH_3CO_2)_2$, 0.1 M) and/or silver nitrate ($AgNO_3$, 0.01 M) were also investigated, and the results were compared with that from the NaOH trapping solution. As detailed below, using NaOH as the trapping solution allows direct quantification of the sulfur concentration by UV absorption spectroscopy, which is faster and more reliable than gravimetric techniques.

## 2.2 | Apparatus

The reduction train is sketched in Figure 1. The main parts of the apparatus are four 15-mL glass tubes each with a nitrile/PTFE septum and a block heater. Reaction tube 'a', two gas washing tube 'b1' and 'b2' and the collection tube 'c' were connected with PEEK tubes (1/16" ID) directly through the septum. Alternatively, a drying cartridge filled with potassium perchlorate ($KClO_4$) and a cryogenic trap (whose internal temperature can be controlled between −200°C and −80°C) can be placed between the trap 'b2' and the collection tube to test the possibility of using pure $H_2S$ as the working gas for isotope analysis. The drying cartridge and the cryogenic trap allow us to purify $H_2S$ without any loss. The dry sulfate sample (i.e. $Na_2SO_4$) and 1 mL reducing solution were introduced into glass tube 'a', which was placed on a block heater and purged with a He flow for 20 minutes before turning the heater to a temperature of 124°C. The purge before the heating stage is necessary to remove traces of $I_2$, especially when the reducing solution has a light yellow appearance over time due to slight oxidation.

The He gas was supplied from a tank. In practice, we used a home-made flow distributor to distribute the He gas to eight reaction train flows, as shown in Figure 2. Each flow was then guided to an individual reduction train, and the flow rate (~2 mL/min) of each reduction train was controlled by a micro-flow meter (ref: P-446, IDEX Health & Science, Sainte-Foy-La-Grande, France). In this way, multiple samples can be processed simultaneously.

## 2.3 | Testing samples

We used the above-mentioned reducing solution and apparatus to process dry sulfate samples in the forms of barium sulfate ($BaSO_4$) and sodium sulfate ($Na_2SO_4$). The $Na_2SO_4$ samples were prepared from a 1 mM $Na_2SO_4$ solution (0.142 g $Na_2SO_4$ in 1 L Milli-Q water; Millipore SAS, Molsheim, France), and then the desired volume of the $Na_2SO_4$ solution (e.g., 0.2 or 0.5 mL, equivalent to 0.2 or 0.5 μmol $SO_4^{2-}$) was added to a pre-cleaned reaction tube. The reaction tube

was allowed to completely dry in a 100°C oven, and the sample was then stored for later use.

In order to prepare the $BaSO_4$ samples, the desired volume (e.g., 0.2 or 0.5 mL) of the 1 mM $Na_2SO_4$ solution was added to the reaction tube, and a drop of 1 M HCl solution was then added to remove any carbonate in the solution. After storage overnight, a drop of 0.1 M $BaCl_2$ solution was added to the reaction tube to precipitate $BaSO_4$. After the $BaSO_4$ had precipitated, the samples were divided into two sets, which were then processed differently. One set of $BaSO_4$ samples was dried completely in an oven at 100°C, so the dry samples contain $BaSO_4$, $BaCl_2$ and NaCl. We termed this set of samples $BaSO_4$-EB ($BaSO_4$ with excess $BaCl_2$). The other set of $BaSO_4$ samples was centrifuged and the supernatant was removed. The remained solids were rinsed with Milli-Q water and then separated from the rinsing water by centrifuging. This step was repeated three times before the sample was placed in the oven to dry. This set of samples was termed P-$BaSO_4$ (pure $BaSO_4$).

In addition, international reference materials (in the form of $BaSO_4$), IAEA-SO-5 ($\delta^{34}S$ = (0.5 ± 0.2) ‰), IAEA-SO-6 ($\delta^{34}S$ = (−34.1 ± 0.2) ‰) and NBS-127 ($\delta^{34}S$ = (20.3 ± 0.4) ‰) were prepared by weighing ~ 0.5 mg of the $BaSO_4$ standards into reaction tubes. After reduction, these samples were further converted into $SF_6$ for isotope analysis. We note that the reference materials were not weighed precisely because of the capability of our balance (0.1 mg precision).
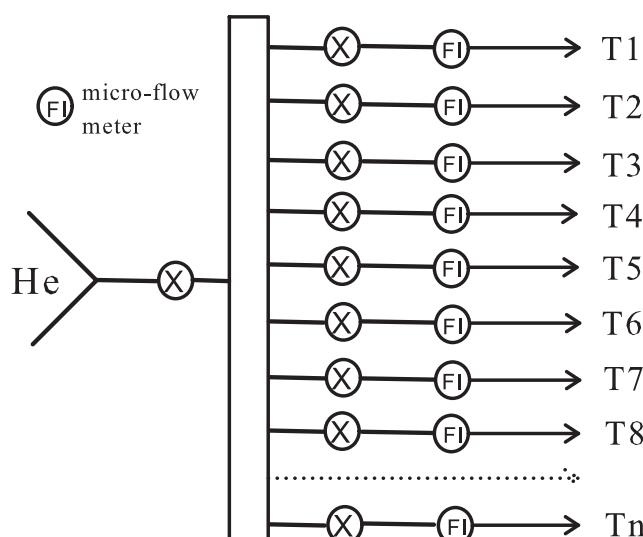


**FIGURE 2** Sketch of the flow system containing multiple reaction trains. "T1…Tn" indicate the reduction trains assembled

However, the purpose of processing these samples is to test potential sulfur isotope fractionation during the reduction, rather than to assess the reduction yield (which can be assessed from the samples made from drying $Na_2SO_4$ solution with accurate measurement of sulfur content, or precipitating $BaSO_4$ from the same $Na_2SO_4$ solution).

## 2.4 | Quantification

The yield of the reduction from sulfate ($SO_4^{2-}$) to sulfide ($S^{2-}$) can be directly assessed by determining the quantity of $H_2S$ collected in the NaOH trapping solution. Hydrogen sulfide ($H_2S$) solution is known to absorb UV light with a peak absorbance at 230 nm.[22,23] Guenther et al[22] have shown that in alkaline solutions with pH >8, $H_2S$ is present nearly 100% in the form of the bisulfide ion ($HS^-$), and they found that at pH ~8, UV determination of $HS^-$ yields are accurate because precise estimates of total sulfide concentration in the solution can be achieved. Thus, with NaOH as the trapping solution, the yield of the reduction can be directly assessed by measuring $HS^-$ in the solution with optical methods. In comparison, the conventional trapping solution (cadmium acetate or silver nitrate) collects $H_2S$ as a precipitate, which makes it difficult to directly quantify the reduction yield.

In this study, we used a UV spectrophotometer (model 6850; Jenway, Stone, UK) to determine the concentration of $H_2S$ in the NaOH trapping solution. The calibration standards were made by mixing sodium sulfide nonahydrate ($Na_2S \cdot 9H_2O$, >99.99% purity; Sigma-Aldrich, St Louis, MO, USA) with 0.1 M NaOH solution. A few crystals of $Na_2S \cdot 9H_2O$ were quickly rinsed on Kimwipes® disposable wipers to remove surface oxidation products, dried and weighed directly. A stock solution of 0.01 M $HS^-$ was made by mixing 0.0125 g of pre-cleaned $Na_2S \cdot 9H_2O$ in 5 mL 0.1 M NaOH solution. A set of working standards, 0.0 μM, 20 μM, 50 μM and 100 μM, was then made by diluting 0, 0.02, 0.05 and 0.1 mL of the stock solution into the required volume of 0.1 M NaOH to obtain a 10-mL standard solution. The stock solution should be stored in a sealed brown bottle and flushed with He before storage, since sulfide is easily oxidized by $O_2$ once in contact with air. Even when the stock solution was flushed before storage, we noticed significant loss of sulfide after 2–3 days. Guenther et al[22] made the stock solution in a glass aspirator bottle purged with $N_2$, and stated that the solution should be stable for about 1–2 weeks. In practice, we prepared a fresh stock solution once every 2 days, and working standards every day.

## 2.5 | Procedure

Prior to the reduction, all glassware, caps, septum and PEEK tubes were cleaned with Milli-Q water. The PEEK tubes have to be flushed to ensure that there is no water left inside them; otherwise the water will block the flow of the carrier gas in the reduction line.

In a fume hood, 1 mL of reducing solution was added to a pre-prepared reaction tube to a known amount of dry sulfate. In the reaction tube, the reducing solution was purged with He for 20 min at room temperature to remove any $I_2$ and $O_2$. The gas washing tubes ('b1' and 'b2' in Figure 1) and the collection tube ('c' in Figure 1) were prepared by adding 12 mL Milli-Q water and 12 mL 0.1 M NaOH, respectively. After the reducing solution had been purged for 20 min,

the reduction train was assembled (Figure 1) and the reaction tube was placed in the block heater and heated at 124°C. At lower temperatures the reduction speed will be slow, while if the temperature is too high, an excessive amount of phosphine ($PH_3$) will be produced from the decomposition of $NaH_2PO_2$.[14] For the alternative setup, the drying agent was in-line with the cryogenic system, and the latter was set at −200°C to trap the reaction products. When the reaction was over, the temperature of the cryogenic trap was raised to −120°C when the produced $H_2S$ was released and trapped in the collection tube.

The collection tube was removed from the reduction train after the reaction was complete. The concentration of $H_2S$ in the trapping solution was first measured by UV spectrophotometry as described in section 2.4, in order to assess the yield. Then 1 mL of 0.01 M $AgNO_3$ was added to the collection solution to precipitate $Ag_2S$ and $Ag_2O$. After gentle shaking, a few drops of concentrated $HNO_3$ (68%) were added to the suspension. Following thorough shaking, the $Ag_2O$ dissolved and only $Ag_2S$ remained in the solid phase. The tube was allowed to settle, and a plastic laboratory dropper was used to remove the supernatant. The solid was then rinsed three times with Milli-Q water, transferred to an aluminum boat and dried prior to fluorination.

## 2.6 | Isotope analysis

To explore potential sulfur isotope fractionation during the reduction, we processed the international sulfate reference materials IAEA-SO-5, IAEA-SO-6 and NBS-127, following the procedure mentioned in section 2.5. The reference materials were weighed, and approximately 0.5 mg was added to the reaction tube. The reaction was stopped after ~5 h.

After being converted into $Ag_2S$ as described in section 2.5, the reference materials were transported to the Stable Isotope Geochemistry Laboratory at IPG-Paris (Institut de Physique du Globe, Paris, France) for sulfur isotope analysis. At IPG, the samples were dried, transferred to an aluminum boat and then weighed. Due to the small quantity (~0.3 mg $Ag_2S$ or less) of the sample, we found it is very difficult to transfer the dry $Ag_2S$ from the collection tube to the Al boat. As an alternative, we transferred the solid together with a small amount of water from the tube to the Al boat, and then dried the samples. Under these circumstances we found that, after drying, the inside wall of the Al boat became light-brown in color, and the mass of the dried Al boat plus the sample exceeded the sum of the sample and the Al boat, indicating the gain of extra mass during the drying process. This is probably due to the development of a thin layer of $Al_2O_3$ on the Al metal surface when Al contacts with water at the drying temperature (70°C). This is consistent with the observation that, after drying an Al boat with Milli-Q water at 70°C, a brown layer was formed on the inner surface of the Al boat and the mass of the Al boat was increased. Nevertheless, the fluorination yields and the sulfur isotopic analysis results suggested this influences neither the fluorination procedure nor the isotopic composition.

The dried $Ag_2S$ samples were fluorinated in nickel bombs under approximately 37 kPa of fluorine gas ($F_2$) at 250°C overnight. The evolved $SF_6$ was purified cryogenically and then by gas chromatography. Because of the small amount of samples (<0.5 mg $Ag_2S$), a microvolume cold finger of an isotope ratio mass spectrometer (MAT 253; Thermo

Scientific, Bremen, Germany) working in dual-inlet mode was used to concentrate the sample gas for isotope analysis.[24] The analytical uncertainty (1σ) for the instrument was 0.25‰ for $\delta^{34}S$ values, 0.010‰ for $\Delta^{33}S$ and 0.062‰ for $\Delta^{36}S$ obtained by replicate analysis (N = 4) of IAEA-S-1 over a period of 4 weeks (once a week) when the processed sulfate standards were also measured for sulfur isotopic composition.

# 3 | RESULTS AND DISCUSSION

## 3.1 | H₂S collection agents

The reduction product, $H_2S$, has to be collected and converted into $Ag_2S$ before fluorination for isotope analysis. As mentioned above, $Cd(CH_3CO_2)_2$[16,17] and $AgNO_3$[13] have both been shown to be able to efficiently trap $H_2S$ by forming CdS and $Ag_2S$ precipitates, respectively. The CdS precipitate is further converted into $Ag_2S$ by adding $AgNO_3$ solution.[16,17]

The conventional reducing solution commonly contains phosphorous acid ($H_3PO_3$) or hypophosphorous acid ($H_3PO_2$),[13] and phosphine ($PH_3$) is produced when the reducing solution is heated.[18] Once $PH_3$ comes in contact with $AgNO_3$, it reduces $Ag^+$ to $Ag^0$ and this leads to excess precipitate in addition to $Ag_2S$.[17] To prevent this, Thode et al[16] and Forrest et al[17] used $Cd(CH_3CO_2)_2$ as the trapping solution. In particular, Forrest et al[17] flushed the $Cd(CH_3CO_2)_2$ solution with $N_2$ for 15 min after the CdS precipitated and prior to adding $AgNO_3$. This step was found to effectively remove $PH_3$ and thus no excess precipitate formed. However, Arnold et al[13] found that when using $AgNO_3$ as the trapping solution, the excess Ag precipitate in the trap is not detrimental to the final isotope analysis of the sulfur content after fluorination. Because of this, $AgNO_3$ appears to be the better reagent for the collection of $H_2S$, given the environmentally toxic nature of $Cd^{2+}$.

In this study, we first employed 0.01 M $AgNO_3$ as the trapping solution. However, we observed spuriously high precipitates in the trap as soon as the reducing solution was heated, and the trapping solution turned completely dark in a few minutes, even when there was no sulfate added to the reducing solution. At the same time, we noticed an apparent silver mirror on the inside wall of the collection tube, indicating reduction of $Ag^+$ to $Ag^0$. This severe reduction of the $AgNO_3$ solution is probably due to the high production of $PH_3$ from our reducing solution. Different from the conventional reducing solution, our reducing solution used $NaH_2PO_2$ instead of $H_3PO_2/H_3PO_3$. $NaH_2PO_2$ starts to decompose and produce $PH_3$ at 90°C, while $H_3PO_3$ effectively decomposes to yield $PH_3$ at 200°C. Therefore, at the temperature of the reduction experiment (i.e., 124°C), our reducing solution was presumably producing much more $PH_3$ than the conventional reducing solution. To remove the excess precipitate other than $Ag_2S$ caused by $PH_3$, we used 1 M $HNO_3$ followed by 1 M $NH_4OH$ to wash the precipitate formed in the $AgNO_3$ trapping solution. Only part of the precipitate was removed after these treatments and there was still more precipitate than expected. Thus, $AgNO_3$ is not a good choice as the trapping solution, as least for our reducing solution.

To avoid the reduction of $Ag^+$ by $PH_3$, we next tested 0.1 M $Cd(CH_3CO_2)_2$ as the trapping solution and following the strategies described in Forrest et al.[17] Despite this, excess precipitation was still frequently observed after $AgNO_3$ was added to the trapping solution for conversion of CdS into $Ag_2S$. In particular, we noticed that during the collection of $H_2S$, yellow material was accumulating at the wall directly above the surface of the $Cd(CH_3CO_2)_2$ solution, indicating the formation of CdS. However, at the same time, the entire $Cd(CH_3CO_2)_2$ solution became light brown and the brown color became deeper with increasing trapping time. When $AgNO_3$ was added after the collection, the trapping solution turned dark with extensive precipitate at the same time. Obviously, there were still interferences between the trapping solution and/or $AgNO_3$ with the volatile product(s) of the reducing solution. Similarly, excess precipitate remained after washing with 1 M $HNO_3$ and 1 M $NH_4OH$. This, together with the toxic nature of $Cd^{2+}$, made us decide to abandon $Cd(CH_3CO_2)_2$ as the trapping solution in our system.

Instead, we used 0.1 M NaOH as the trapping solution to collect $H_2S$. At a pH of 13, the trapped $H_2S$ mainly existed in the form of $HS^-$, as shown in Figure 3A. Since the NaOH trapping solution was purged with He, under this condition the dissolved $O_2$ concentration was very low and thus the trapped sulfide was stable. The use of NaOH as the trapping solution has two advantages: (1) the trapped $H_2S$ can be precisely quantified in real-time using UV spectrophotometry, as described in section 2.4, and thus the progress toward to complete reduction of a sulfate sample can be monitored; and (2) no reaction occurs between $PH_3$ and $AgNO_3$ thus avoiding the production of excessive mass interference.

After sample collection, 1 mL 0.01 M $AgNO_3$ was added to the trap to produce $Ag_2S$. AgOH was produced at the same time, and this quickly changed to $Ag_2O$. The suspension was allowed to settle for 10–20 min after thorough shaking, and a few drops of 68% $HNO_3$ were then added to acidify the trapping solution. $Ag_2O$ was readily dissolved in the acidified solution and only $Ag_2S$ remains.

## 3.2 | H₂S yield

In the 0.1 M NaOH trapping solution, sulfide was mainly present in the form of $HS^-$ (Figure 3A). Figure 3B shows the typical absorbance spectra of two $Na_2S$ working standards (in 0.1 M NaOH matrix) and two NaOH trapping solutions after 2 h collection of $H_2S$ and, as expected, the absorbance spectra peak was at ~230 nm, consistent with that from Guenther et al.[22] Figure 3C shows the plot of the average of the calibration curve over 3 days of analyzing working standards.

As described in section 2.3, three different sulfate samples were processed using our system, $Na_2SO_4$, $BaSO_4$-EB and P-$BaSO_4$, and the time-resolved $H_2S$ yields from these three materials are plotted in Figure 4. The real-time production of $H_2S$ was monitored by UV determination of $HS^-$ in the trapping solution every 15–20 min. Once the produced $H_2S$ reached the amount expected from the starting sulfate, or no longer increased with time, the block heater was turned off and the reduction train was flushed with He for a further 20 min after the reaction tube had cooled to room temperature.

In general, $Na_2SO_4$ was reduced faster than P-$BaSO_4$, and much faster than $BaSO_4$-EB. Regardless of the quantity of the starting sulfate, after 1 h of reduction an average $H_2S$ yield of 85.7 ± 10.3% was reached when $Na_2SO_4$ was the starting material. In comparison,
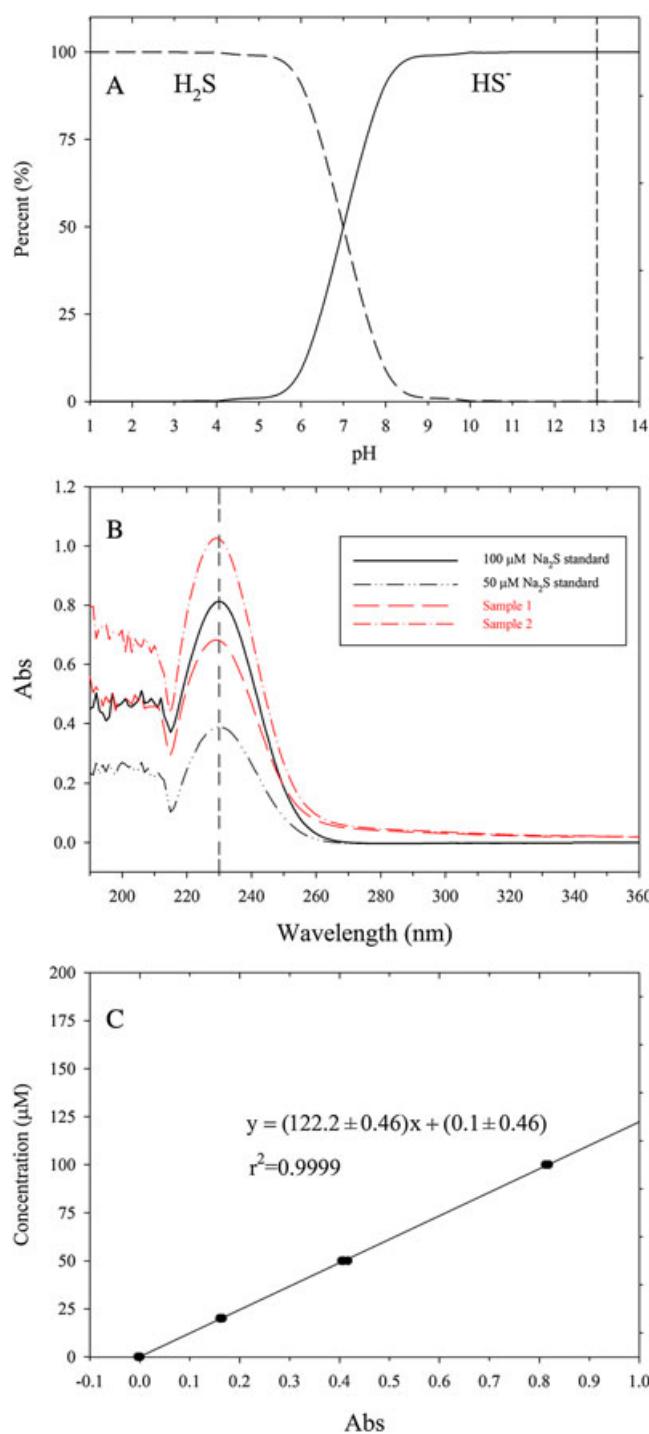
**FIGURE 3** A) Percentages of $H_2S$ and $HS^-$ in solutions with different pH calculated with $pK_{a1}$ of 7.0 and $pK_{a2}$ of 19, where the vertical dashed line indicates the pH of the trapping solution used in this study. B) Absorbance spectra of $Na_2S$ working standards and trapping solutions after 1 h collection, where the vertical dashed line indicates the absorbance peak of 230 nm; C) A 3-day averaged calibration curve for $H_2S$ quantification [Color figure can be viewed at wileyonlinelibrary.com]

the $H_2S$ yield after 1 h of reduction was 63.9 ± 2.1% for $BaSO_4$-EB and only 18.5 ± 0.04% for P-$BaSO_4$. After 2 h, a 99.5 ± 3.7% yield was reached for $Na_2SO_4$, indicating the completion of the reduction. However, after 2 h, it appeared that no more $H_2S$ was produced for $BaSO_4$-EB and P-$BaSO_4$, and the yield remained at 80.4 ± 0.75% for
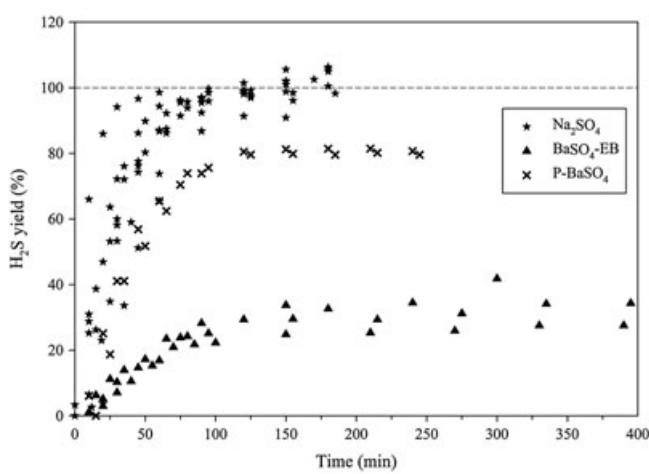


**FIGURE 4** Time-resolved yields of $H_2S$ from the reduction of dry $Na_2SO_4$, $BaSO_4$-EB ($BaSO_4$ with excess $Ba^{2+}$) and P-$BaSO_4$ (pure $BaSO_4$)

$BaSO_4$-EB and 28.5 ± 0.09% for P-$BaSO_4$ after 4 or 5 h. For two of the $BaSO_4$-EB samples, we let the reaction continue overnight, and the yield increased from 41.7% and 34.5% at 5 h to 58.3% and 86.5%, respectively.

The final yields (yield after stopping the reaction) of $Na_2SO_4$, $BaSO_4$-EB and P-$BaSO_4$ sample with different quantities of sulfate are plotted in Figure 5. Overall, $Na_2SO_4$ was often 100% reduced within 2 h regardless of the starting quantity, even when the drying agent and the cryogenic trap were put in-line, while a 100% yield for $BaSO_4$-EB and P-$BaSO_4$ was never observed even after overnight heating.

The different apparent reaction speeds and yields of $H_2S$ between $Na_2SO_4$, $BaSO_4$-EB and P-$BaSO_4$ and the reducing solution probably reflect the effect of the sulfate salt solubility. $Na_2SO_4$ is soluble in water, while $BaSO_4$ has a very low solubility of 0.01 μmol/mL in water at 20°C and less than 0.02 μmol/mL at ~120°C.[25] Given the small volume of the reducing solution used (1 mL), there would be less than 2% of the added $BaSO_4$ (if 1 μmol is added) dissolved. Our observations clearly point to the sulfate ion ($SO_4^{2-}$) or sulfuric acid
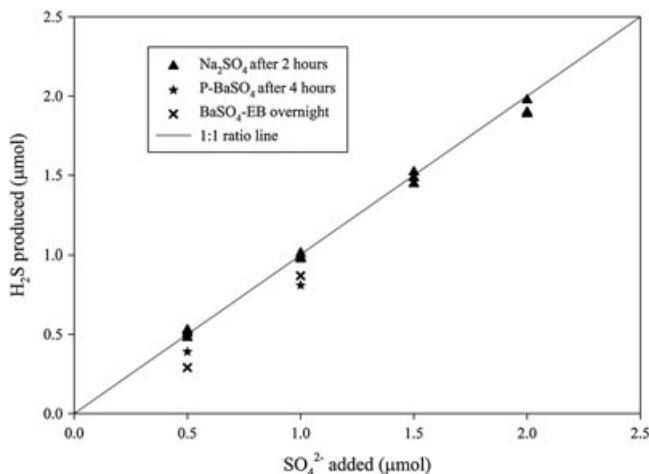


**FIGURE 5** Yields of $H_2S$ from the reduction of $Na_2SO_4$, $BaSO_4$-EB ($BaSO_4$ with excess $Ba^{2+}$) and P-$BaSO_4$ (pure $BaSO_4$) at different sulfate quantities at the time that the reaction is stopped

**TABLE 1** Fluorination yields and measured isotopic ratios of the sulfate standards processed with this system

| Standards | Ag$_2$S (mg) | SF$_6$ yield (%) | Δ$^{33}$S values vs CDT (‰) | | δ$^{34}$S values vs CDT (‰) | | Accepted δ$^{34}$S[a] values vs CDT (‰) |
|---|---|---|---|---|---|---|---|
| NBS-127 | 0.20 | 101.7 | 0.015 | 0.025 ± 0.010 | 19.8 | 21.6 ± 1.3 | 20.3 ± 0.5 |
| | 0.10 | 105.3 | 0.018 | | 22.4 | | |
| | 0.08 | 93.7 | 0.033 | | 22.8 | | |
| | 0.12 | 98.2 | 0.034 | | 21.4 | | |
| IAEA-SO-5 | 0.51 | 104.6 | 0.063 | 0.097 ± 0.071 | 0.7 | 0.7 ± 0.2 | 0.5 ± 0.5 |
| | 0.52 | 101.6 | 0.052 | | 0.7 | | |
| | 0.82 | 26.1 | 0.203 | | 0.8 | | |
| | 0.21 | 99.3 | 0.067 | | 0.5 | | |
| IAEA-SO-6 | 0.41 | 113.5 | 0.077 | 0.086 ± 0.020 | -34.0 | -33.5 ± 0.6 | -34.1 ± 0.5 |
| | 0.46 | 106.9 | 0.065 | | -33.9 | | |
| | 0.13 | 102.0 | 0.110 | | -32.9 | | |
| | 0.15 | 84.6 | 0.090 | | -32.9 | | |

The values of Δ$^{36}$S are not reported as when the samples were measured the mass spectrometer had a high background of mass 131 (15 to 50 mV) and thus the Δ$^{36}$S data were discarded. The initial masses of the BaSO$_4$ standards were only approximately weighed, and the mass(es) in Ag$_2$S form were obtained according to the measured H$_2$S production after ~5 h of reduction.

[a]Accepted values are taken from Halas and Szaran.[28]

(H$_2$SO$_4$) as the reactive species with the reducing acids, i.e. the sulfate salt has to be dissolved first in order to produce H$_2$S. This explains why the BaSO$_4$ samples reacted so slowly with the reducing solution relative to Na$_2$SO$_4$. In addition, if there is an excess of Ba$^{2+}$ ions in the solution (due to the dissolution of the excess BaCl$_2$ used to precipitate BaSO$_4$ from Na$_2$SO$_4$), this will inhibit the dissolution of BaSO$_4$ as the dissociation equilibrium of BaSO$_4$ will be pushed to the BaSO$_4$ side, following Le Chatelier's principle. This probably explains why the reducing reaction with P-BaSO$_4$ was faster than that with BaSO$_4$-EB. To confirm the effect of excess Ba$^{2+}$ ions on the reduction of BaSO$_4$, we prepared a few BaSO$_4$ samples with considerably more Ba$^{2+}$ by adding 1 mL of 0.1 M BaCl$_2$ to 1 mL of 1 mM Na$_2$SO$_4$ solution. These samples were then directly dried without removing the supernatant from the precipitate. For these samples, after the reduction started, we measure the trapping solution every hour for 7 h, and no H$_2$S was detected at any time.

Therefore, the solubility of the sulfate salt largely affects the reduction speed and the overall yield. We thus recommend extracting and converting sulfate in natural samples into Na$_2$SO$_4$ whenever possible when applying our reducing solution. The extraction of sulfate can be conducted using the IC method described in Geng et al[26] or the anion-exchange resin method described in Le Gendre et al.[21] If the use of BaSO$_4$ is unavoidable, excess Ba$^{2+}$ should be removed after precipitating BaSO$_4$ with BaCl$_2$, while increasing the volume of the reducing solution (e.g., using 10 mL instead of 1 mL) and/or the reaction time may improve the yield.

## 3.3 | Isotope analysis of the standard materials

Since the overall goal of reducing sulfate to sulfide is to perform the four-sulfur isotopes analysis, we processed three different barium sulfate standards, IAEA-SO-5, IAEA-SO-6 and NBS-127, which were equivalent to P-BaSO$_4$ samples. Unfortunately, there are no international standards in sodium sulfate form and thus a strict comparison of the isotopic precision of the reduction step for the two chemical forms is impossible. Even a simple comparison of the salt from an identical sulfate batch is not possible, as BaSO$_4$ reduction will never reach full

decomposition. The fluorination yields from Ag$_2$S to SF$_6$ and sulfur isotopic compositions measured from these standards are listed in Table 1. The fluorination yield range is from 84.6 to 113.5% with an average of 101 ± 7.5%, except for one standard with a low yield of 26.1%. Regardless of the fluorination yield, the measured isotopic ratios of all the processed sulfate standards are statistically consistent with their accepted values, including the one with relatively low yield (26.1%). The measured δ$^{34}$S(‰)$_{VCDT}$ values of all standards with different quantities of sulfur (0.34–2 μmol in SF$_6$) versus their accepted δ$^{34}$S(‰)$_{VCDT}$ values are plotted in Figure 6. A least-squares linear regression gives a slope of (0.99 ± 0.01), suggesting good reproducibility and the conservation of sulfur isotopic composition during the reduction of sulfate to sulfide using our reducing system, despite the reduction yields of these standard materials not being 100%. This is not a surprise. In fact, if any sulfur isotope fractionation occurs during the reduction, it would be between the solid BaSO$_4$ and the dissolved HSO$_4^-$ (the form of SO$_4^{2-}$ in concentrated acid solution), but not in the step(s) from SO$_4^{2-}$ to H$_2$S because the dissolved part is

$$y = (0.99 \pm 0.014)x$$
$$R^2 = 0.998$$
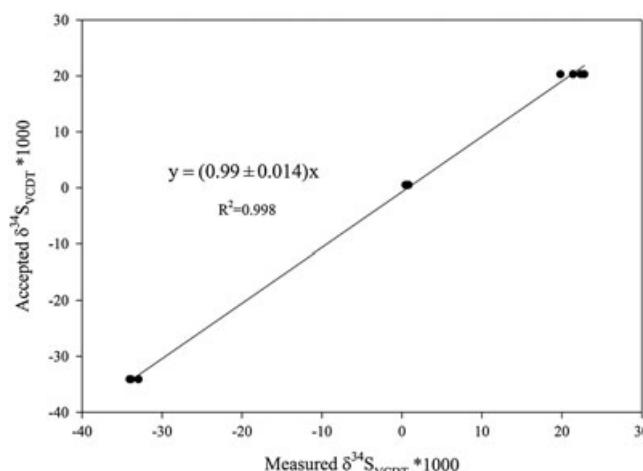
**FIGURE 6** Measured versus accepted δ$^{34}$S (‰)$_{VCDT}$ values of IAEA-SO-5, IAEA-SO-6 and NBS-127. The reduction of these sulfate standards to H$_2$S were conducted using the protocol described in this study

100% converted into H$_2$S. Kusakabe and Robinson[27] found that the sulfur isotope fractionation between solid BaSO$_4$ and the dissolved HSO$_4^-$ in the BaSO$_4$–HSO$_4^-$–H$_2$O system is very small (less than 0.4‰ in the temperature range from 110 to 350°C), which could explain why the solubility effect seems to not affect the isotopic measurements.

For these standards, we also reported the $\Delta^{33}$S values and they are all not distinct from what can be expected from mass-dependent fractionation. However, we did not include the $\Delta^{36}$S values as, when these standards were measured, the mass spectrometer had a high and variable background at $m/z$ 131 up to 50 mV which caused the $\delta^{36}$S values to drift and made them unreliable.

## 4 | CONCLUSIONS

We present a simple and reliable reducing method modified from the literature for the conversion of sulfate into sulfide for four-sulfur isotopes analysis. This system is simple to set up, easy to replace and cheap to acquire and is made from sealed test tubes and PEEK flow lines (metal part, e.g. needle, in contact with the hot reducing solution is not allowed). This method uses a reducing solution made of 100 mL 57% HI and 13 g NaH$_2$PO$_2$, and a very small amount (1 mL) of reducing solution was demonstrated to be able to completely reduce a soluble sulfate salt (0.5–2.5 µmol) to sulfide within 2 h, thus minimizing the use of relatively expensive HI. In practice, nothing prohibits the recycling of the used reducing solution by adding a few mg of NaH$_2$PO$_2$ in order to reduce I$_2$ back to I$^-$ in a boiling flask if the used solution turns brown.[14] In addition, the reduction train avoids the use of a distillation apparatus, and multiple reduction trains can be operated at a time, making it easier to process multiple samples simultaneously. The use of NaOH as the trapping solution allows the assessment of reduction yield directly from UV determination of HS$^-$ in the trapping solution.

This new approach was demonstrated to produce H$_2$S very rapidly with a 100% recovery when soluble sulfate salt was used (e.g., Na$_2$SO$_4$), as opposed to BaSO$_4$ for which the kinetic was slow and conversion never reached 100% even after overnight reaction. However, despite the relatively low reduction yield for BaSO$_4$, there was no significant isotope fractionation effect induced by the reduction. As it is the dissolved part of the sulfate salt that reacts with the reducing solution, this method is most suitable for natural samples containing soluble sulfate (e.g., aerosol, snow and ice core), which can be extracted (e.g., by the resin method) and converted into Na$_2$SO$_4$. The use of the barite precipitate method for sulfate extraction and purification is not recommended as the salt solubility inhibits the reduction speed and yield. If BaSO$_4$ is the main form of sulfate (e.g., barite), increasing the volume of the reducing solution and/or the reaction time may improve the H$_2$S yield although there is no guarantee of a complete conversion. While poor conversion and fluorination yields do not seem to introduce isotope fractionations, poor yield reduces the sensitivity of the method to sample sizes above a few micromoles of sulfate and it may also have consequence on the mass-dependent slopes between the sulfur isotope ratios as the $^{33}$S/$^{32}$S ratios of the international standards have never been calibrated.

## ORCID

*Lei Geng* http://orcid.org/0000-0003-2175-2538
*Joel Savarino* http://orcid.org/0000-0002-6708-9623

## REFERENCES

1. Canfield DE. Biogeochemistry of sulfur isotopes. *Rev Mineral Geochem*. 2001;43(1):607-636. https://doi.org/10.2138/gsrmg.43.1.607

2. Farquhar J, Bao HM, Thiemens M. Atmospheric influence of Earth's earliest sulfur cycle. *Science*. 2000;289(5480):756-758. https://doi.org/10.1126/science.289.5480.756

3. Feulner G. The Faint Young Sun Problem. *Rev Geophys*. 2012;50(2):375-393. https://doi.org/10.1029/2011rg000375

4. Farquhar J, Savarino J, Jackson TL, Thiemens MH. Evidence of atmospheric sulphur in the martian regolith from sulphur isotopes in meteorites. *Nature*. 2000;404(6773):50-52. https://doi.org/10.1038/35003517

5. Cole-Dai J, Ferris D, Lanciki A, et al. Cold decade (AD 1810-1819) caused by Tambora (1815) and another (1809) stratospheric volcanic eruption. *Geophys Res Lett*. 2009;36(22):703-708. https://doi.org/10.1029/2009gl040882

6. Savarino J, Romero A, Cole-Dai J, Bekki S, Thiemens MH. UV induced mass-independent sulfur isotope fractionation in stratospheric volcanic sulfate. *Geophys Res Lett*. 2003;30(21):2131-2141. https://doi.org/10.1029/2003gl018134

7. Baroni M, Thiemens MH, Delmas RJ, Savarino J. Mass-independent sulfur isotopic compositions in stratospheric volcanic eruptions. *Science*. 2007;315(5808):84-87. https://doi.org/10.1126/science.1131754

8. Lanciki A, Cole-Dai J, Thiemens MH, Savarino J. Sulfur isotope evidence of little or no stratospheric impact by the 1783 Laki volcanic eruption. *Geophys Res Lett*. 2012;39(1):1806-1810. https://doi.org/10.1029/2011gl050075

9. Wu N, Farquhar J, Strauss H, Kim ST, Canfield DE. Evaluating the S-isotope fractionation associated with Phanerozoic pyrite burial. *Geochim Cosmochim Acta*. 2010;74(7):2053-2071. https://doi.org/10.1016/j.gca.2009.12.012

10. Sansjofre P, Cartigny P, Trindade RIF, et al. Multiple sulfur isotope evidence for massive oceanic sulfate depletion in the aftermath of Snowball Earth. *Nat Commun*. 2016;7:12192-12199. https://doi.org/10.1038/ncomms12192

11. Ono S, Wing B, Rumble D, Farquhar J. High precision analysis of all four stable isotopes of sulfur ($^{32}$S, $^{33}$S, $^{34}$S and $^{36}$S) at nanomole levels using a laser fluorination isotope-ratio-monitoring gas chromatography–mass spectrometry. *Chem Geol*. 2006;225(1–2):30-39. https://doi.org/10.1016/j.atmosenv.2014.02.001

12. Ueno Y, Aoyama S, Endo Y, Matsu'ura F, Foriel J. Rapid quadruple sulfur isotope analysis at the sub-micromole level by a flash heating with

CoF$_3$. *Chem Geol*. 2015;419:29-35. https://doi.org/10.1016/j.chemgeo.2015.10.032

13. Arnold GL, Brunner B, Müller IA, Røy H. Modern applications for a total sulfur reduction distillation method – what's old is new again. *Geochem Trans*. 2014;15(1):4-15. https://doi.org/10.1186/1467-4866-15-4

14. van Grondelle MC, van de Craats F, van der Laarse JD. Microcoulometric determination of total inorganic sulphur in water by a hydroiodic acid reduction method. *Anal Chim Acta*. 1977;92(2):267-275. https://doi.org/10.1016/S0003-2670(01)93500-7

15. Toshiyasu K, Tomoo T, Yooko Y, Ikuko K. Tin (II)-strong phosphoric acid. A new reagent for the determination of sulfate by reduction to hydrogen sulfide. *Bull Chem Soc Jpn*. 1955;28(9):641-644. https://doi.org/10.1246/bcsj.28.641

16. Thode HG, Monster J, Dunford HB. Sulphur isotope geochemistry. *Geochim Cosmochim Acta*. 1961;25(3):159-174. https://doi.org/10.1016/0016-7037(61)90074-6

17. Forrest J, Newman L. Silver-110 microgram sulfate analysis for the short time resolution of ambient levels of sulfur aerosol. *Anal Chem*. 1977;49(11):1579-1584. https://doi.org/10.1021/ac50019a030

18. Davis JB, Lindstrom F. Spectrophotometric microdetermination of sulfate. *Anal Chem*. 1972;44(3):524-532. https://doi.org/10.1021/ac60311a018

19. Norwitz G. Spectrophotometric determination of sulphate in propellents and nitrocellulose. *Analyst*. 1971;96(1144):494-501. https://doi.org/10.1039/an9719600494

20. Gustafsson L. Determination of ultramicro amounts of sulphate as methylene blue –I: The colour reaction. *Talanta*. 1960;4(4):227-235. https://doi.org/10.1016/0039-9140(60)80126-9

21. Le Gendre E, Martin E, Villemant B, Cartigny P, Assayag N. A simple and reliable anion-exchange resin method for sulfate extraction and purification suitable for multiple O- and S-isotope measurements. *Rapid Commun Mass Spectrom*. 2017;31(1):137-144. https://doi.org/10.1002/rcm.7771

22. Guenther EA, Johnson KS, Coale KH. Direct ultraviolet spectrophotometric determination of total sulfide and iodide in natural waters. *Anal Chem*. 2001;73(14):3481-3487. https://doi.org/10.1021/ac0013812

23. Williams RJ. Determination of inorganic anions by ion chromatography with ultraviolet absorbance detection. *Anal Chem*. 1983;55(6):851-854. https://doi.org/10.1021/ac00257a010

24. Yang DA, Landais G, Assayag N, Widory D, Cartigny P. Improved analysis of micro- and nanomole-scale sulfur multi-isotope compositions by gas source isotope ratio mass spectrometry. *Rapid Commun Mass Spectrom*. 2016;30(7):897-907. https://doi.org/10.1002/rcm.7513

25. Blount CW. Barite solubilities and thermodynamic quantities up to 300 °C and 1400 bars. *Am Mineral*. 1977;62:9-10.

26. Geng L, Schauer AJ, Kunasek SA, et al. Analysis of oxygen-17 excess of nitrate and sulfate at sub-micromole levels using the pyrolysis method. *Rapid Commun Mass Spectrom*. 2013;27(21):2411-2419. https://doi.org/10.1002/rcm.6703

27. Kusakabe M, Robinson BW. Oxygen and sulfur isotope equilibria in the BaSO$_4$-HSO$_4^-$-H$_2$O system from 110 to 350°C and applications. *Geochim Cosmochim Acta*. 1977;41(8):1033-1040. https://doi.org/10.1016/0016-7037(77)90098-9

28. Halas S, Szaran J. Improved thermal decomposition of sulfates to SO$_2$ and mass spectrometric determination of $\delta^{34}$S of IAEA SO-5, IAEA SO-6 and NBS-127 sulfate standards. *Rapid Commun Mass Spectrom*. 2001;15(17):1618-1620. https://doi.org/10.1002/rcm.416