Quantification of volatile alkylated selenium and sulfur in complex aqueous media using solid-phase microextraction

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Revised for publication in *Journal of Chromatography A*

This document is the accepted manuscript version of the following article:
Vriens, B., Mathis, M., Winkel, L. H. E., & Berg, M. (2015). Quantification of
volatile-alkylated selenium and sulfur in complex aqueous media using solid-phase
microextraction. *Journal of Chromatography A*, 1407, 11-20. 1
<https://doi.org/10.1016/j.chroma.2015.06.054>

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Abstract

Biologically produced volatile, alkylated Se and S compounds play an important role in the global biogeochemical Se and S cycles, are important constituents of odorous industrial emissions, and contribute to (off-)flavors in food and beverages. This study presents a fully automated direct-immersion solid-phase microextraction (DI-SPME) method coupled with capillary gas chromatography-mass spectrometry (GC/MS) for the simultaneous quantification of 10 volatile alkylated Se and S compounds in complex aqueous media. Instrumental parameters of the SPME procedure were optimized to yield extraction efficiencies of up to 96% from complex aqueous matrices. The effects of sample matrix composition and analyte transformation during sample storage were critically assessed. With the use of internal standards and procedural calibrations, the DI-SPME-GC/MS method allows for trace-level quantification of volatile Se and S compounds in the ng/L range (e.g., down to 30 ng/L dimethyl sulfide and 75 ng/L dimethyl selenide). The applicability and robustness of the presented method demonstrate that the method may be used to quantify volatile Se and S compounds in complex aqueous samples such as industrial effluents or food and beverage samples.

16 **Highlights**

- 17 • Extraction of volatile alkylated Se and S compounds from complex aqueous media
- 18 • Simultaneous analysis of 10 volatile alkylated Se and S compounds in the ng/L range
- 19 • Thorough evaluation of sample matrix effects
- 20 • Assessment of potential analyte transformations during sample storage
- 21 • Application of method on (incubated) raw wastewater and natural surface water

22 **Keywords**

23 Selenium, Sulfur, SPME, GC/MS, Methylation

1. Introduction

The chemical elements selenium (Se) and sulfur (S) have similar chemical properties and are essential to many organisms [1,2]. In the natural environment, the distribution and bioavailability of these elements is significantly affected by a process called bio-alkylation: the biological production of volatile alkylated Se and S compounds that may be emitted to the atmosphere [3,4]. The alkylation of Se and S is performed by a wide range of organisms (bacteria, fungi, algae, plants, animals, and even humans [5]) and leads to the formation of biogenic volatiles (e.g., dimethyl sulfide (DMS), dimethyl disulfide (DMDS), and Se analogues dimethyl selenide (DMSe), and dimethyl diselenide (DMDS₂) [3,4,6]) that have been identified in soils, fresh waters, oceans, and air [4,7]. However, quantitative field measurements of volatile alkylated Se and S species are relatively scarce, which causes large uncertainties in global atmospheric budget estimates [8-10]. Volatile S species (and possibly Se species as well) can contribute to malodor of industrial emissions (e.g., at wastewater treatment facilities [11,12]) and to (off-)flavors of food (e.g., fruit [13] and cheese [14,15]) and beverages (e.g., wine [16-18] and milk [19]). It is thus important that volatile alkylated Se and S species can be accurately quantified in a wide variety of sample matrices.

Because volatile alkylated S and (particularly) Se species are typically present in low concentrations in the aqueous phase in the natural environment (ng/L range), their speciation analysis is usually preceded by a preconcentration procedure. Although several methods for the preconcentration and speciation analysis of volatile S compounds are available [20,21], preconcentration and speciation methods for Se have focused on the major aqueous Se species, i.e., selenate and selenite [22,23]. Dissolved volatile alkylated Se species have received less attention than anionic Se compounds, even though their presence in aqueous samples may

compromise Se quantification by ICP/MS [24]. Hyphenated analytical procedures such as purge-and-trap combined with cryotrapping [25], solid-phase extraction with preconcentration columns and cartridges [23], liquid-phase (micro) extraction [26,27], and solid-phase (micro) extraction [20,23] have been deployed to preconcentrate volatile Se and S species from aqueous samples. Compared to other preconcentration procedures, solid-phase microextraction (SPME) may be preferred because it is relatively fast, solvent-free and requires only small sample volumes [28].

Solid-phase microextraction has previously been used for the analysis of alkylated S species [29-33], volatile Se species [34-42], or both S and Se species [43,44]. In addition, SPME has been employed in combination with derivation techniques for aqueous inorganic Se speciation analysis [45-49]. The majority of SPME methods for volatile Se and S species have been conducted in headspace extraction mode (e.g., for the analysis of volatile compounds in beverages [43] and human specimens [34,47], or in controlled laboratory incubation experiments [44]) where the SPME fiber is exposed to (relatively) clean gaseous sample headspace. Solid-phase microextraction of volatile alkylated Se and S compounds is rarely performed directly in the aqueous phase, especially in complex and/or turbid samples. However, many volatile alkylated Se and S species are highly soluble in water and are therefore preferably extracted from the aqueous phase: the Henry constants of volatile Se and S species (defined as the ratio of aqueous-phase concentrations over gas-phase concentrations at equilibrium, see Table S1 in the Supplementary Material) illustrate that, at equilibrium, their concentrations in the aqueous phase greatly exceed those in the gas phase.

Here, we present a direct-immersion SPME (DI-SPME) procedure for the direct *in situ* extraction of 10 (semi)-volatile alkylated Se and S compounds from complex aqueous samples. For analyte separation and quantification, the SPME procedure is coupled with GC/MS. We

70 systematically evaluated and optimized the instrumental- and sample parameters of the SPME
71 procedure (e.g., fiber coating type, extraction parameters, as well as sample salinity, pH, and
72 organic matter content) and assessed potential analyte transformations during prolonged sample
73 storage. We applied the method for the quantification of volatile alkylated Se and S analytes in
74 aqueous samples from a peat bog and in raw wastewater. The high degree of repeatability of
75 quantification, as well as the low method detection limits, demonstrates that the method may be
76 applied to a variety of complex and even turbid aqueous samples.

2. Material and methods

2.1. Chemicals and reagents

The following analytes were purchased from Sigma Aldrich (Buchs, Switzerland) and used without further purification: dimethyl sulfide (DMS, $\geq 99\%$), dimethyl disulfide (DMDS, $\geq 99\%$), diethyl sulfide (DES, 98%), diethyl disulfide (DEDS, $\geq 99\%$), ethylmethyl sulfide (EMS, 96%), dipropyl sulfide (DPS, 97%), methyl phenyl sulfide (MPS, $\geq 99\%$), and ethane thiol (ESH, 97%). Dimethyl selenide (DMSe, 99%) and dimethyl diselenide (DMDS₂, 96%) were purchased from Alfa Aesar, Zurich, Switzerland. Dipropyl ether (DPE, $\geq 99\%$), deuterated dimethyl sulfide (DMS-d₆, 99 atom-% D), deuterated p-xylene (XYL-d₁₀, 99 atom-% D), deuterated toluene (TOL-d₈, 99.96 atom-% D) (all Sigma Aldrich, Buchs, Switzerland) and ¹³C-labeled methyl-tert-butyl ether (MTBE, ≥ 99 atom-% ¹³C) (CDN isotopes Canada) were used as internal standards. Selected chemical properties of the analytes and internal standards are listed in the Supplementary Material Table S1. The following chemicals were used for optimization of the SPME procedure: sodium chloride (NaCl, $\geq 99.0\%$), sodium hydroxide (NaOH, $\geq 99.0\%$), a humic acid (CAS 1415-93-6), monosodium phosphate and disodium phosphate ($\geq 98\%$) (all obtained from Sigma Aldrich, Buchs, Switzerland), and HPLC-grade methanol (Alfa Aesar, Zürich, Switzerland). Sodium selenite ($> 98.0\%$) was obtained from Sigma-Aldrich, Switzerland. The sample pH was adjusted with NaOH or ultrapure nitric acid (HNO₃, Carl Roth GmbH, Karlsruhe, Germany). All chemicals were of analytical grade or higher.

2.2. Preparation of standards and samples

All glassware, vials, and syringes were cleaned and rinsed with diluted HNO₃, ultrapure water, and methanol before use. Headspace-free stock solutions of individual analyte standards were

prepared using 10 μ L and 100 μ L gas-tight micro-syringes (Hamilton, Bonaduz, Switzerland) in ultrapure HPLC-grade methanol, hexadecane, or undecane ($\geq 99\%$, Sigma-Aldrich, Buchs, Switzerland) in amber glass crimp vials (2 mL or 10 mL, BGB Analytics, Boeckten, Switzerland) with silicone-PTFE septa (BGB Analytics, Boeckten, Switzerland). From these stock solutions, headspace-free working solutions of individual or combined analytes were freshly prepared in methanol, undecane, hexadecane (for calibration of on-column injections), or ultrapure water (for calibration of SPME analyses, 18 M Ω , Thermo Fisher, NANOpure, Reinach, Switzerland). In order to account for variability in GC/MS sensitivity and SPME-fiber wearing, a mixture of internal standards (~ 1 μ g/L DPE, MTBE, DMS- d_6 , TOL- d_8 , and p-XYL- d_{10}) in methanol was added to all standards and samples measured with the combined DI-SPME-GC/MS method. The internal standards were injected into the crimped vials directly before (< 1 min) the start of extraction. All solutions were stored at 4°C in the dark until use.

2.3. GC/MS analysis

Separation and quantification of analytes was performed using a GC/MS system (Thermo Scientific DSQ II with Trace GC Ultra Gas Chromatograph, Thermo Fisher, Switzerland), equipped with a Stabilwax polyethylene glycol column (60 m, 0.32 mm ID, 1 μ m coating) (BGB Analytics, Boeckten, Switzerland). For optimization of the GC/MS procedure, standards were injected directly into the GC/MS via a cold splitless on-column injector port with He as the carrier gas at a constant injector head pressure of 100 kPa. The optimized temperature program of the GC consisted of a 4 min hold time at 40°C, heating with 7°C/min to 140°C, heating at 25°C/min to 200°C, followed by a 20 min hold time at 200°C. With this program, the separation of 30 investigated compounds was achieved within 40 min (Figure 1). The retention of the

investigated compounds was inversely correlated with the analytes' volatility (Supplementary Material Figure S1).

The GC was connected to the MS through a heated transfer line (deactivated TSP-FS tubing, 530 μm ID, 660 μm OD, BGB Analytics, Boeckten, Switzerland) at 220°C. The MS was operated in the positive ion electron impact mode with a source temperature of 250°C. The MS was tuned on a daily basis. Obtained chromatograms were processed and integrated using the Xcalibur software (Thermo Scientific, Switzerland). Eluted analytes were identified by their specific masses (using species-specific target ions and qualifier ions, see Supplementary Material Table S2), either in a selective ion mode (SIM) or in a scanning mode (SCAN). The SCAN mass range was set to 50–250 amu in order to capture the mass fragments of interest for the identification of the target analytes, and to guarantee sufficient sensitivity by omitting redundant masses: particularly masses below 50 amu suffer from minor interferences caused by bleeding of alkyl-groups from the glycol- and siloxane phases of the GC column and SPME fiber, respectively. Calibrations of on-column injections of standards were based on a 3 point plus blank linear fit over at least two orders of magnitude in the ng/L concentration range. An overview of the investigated target analytes, their retention factors, the linear calibration ranges, and the calibration regressions of the on-column injections are given in Supplementary Material Table S2.

2.4. DI-SPME extraction

Analyte preconcentration with DI-SPME was conducted using a CombiPal autosampler system (CTC, Zwingen, Switzerland). For DI-SPME, the GC/MS system was equipped with a splitless injector that contained a Merlin Microseal septum (Merlin Instruments), a SPME liner of

0.75 mm ID (Supelco, Bellefonte, PA, USA), and a deactivated guard column (0.5 m × 0.53 mm, BGB Analytics, Boeckten, Switzerland). Helium was used as carrier gas at a constant 100 kPa pressure. Prior to use, a preconditioning of both the SPME fiber (1 h at 260°C) and the GC column (1 h at 200°C) took place. The optimization of the DI-SPME procedure took place with headspace-free solutions of analytes in ultrapure water (18 MΩ, Thermo Fisher, NANOpure, Reinach, Switzerland), which were prepared from stock solutions as described above. Different SPME fiber coatings were evaluated (purchased from Supelco, Bellefonte, PA, USA) and both instrumental parameters (extraction time 10 sec–100 min, extraction temperature 35–80°C, desorption time 30 sec–10 min, and desorption temperature 120–310°C) and sample parameters (methanol content 0–15%, pH 3.5–8, ionic strength 0–4 M NaCl, and organic matter content 0–50 g/L humic acid) were optimized. Other instrumental settings (vial agitation during extraction: 500 rpm, vial penetration for extraction: 22 mm and injector penetration for desorption: 45 mm) were adopted from the manufacturers' recommendations and not further optimized.

2.5. Method application

The final DI-SPME-GC/MS method was used to quantify volatile S and Se compounds in raw wastewater from the aerobic and anaerobic sludge reactors of a communal wastewater treatment facility in Dübendorf, Switzerland, and in natural surface water from an ombrotrophic peat bog (Gola di Lago, Switzerland) [50]. Headspace-free samples were collected in triplicate in 10 mL amber glass vials that were immediately sealed with silicone-PTFE septa. A first aliquot of these samples (referred to as 'original aliquots') was analyzed within 12 h of collection (samples stored at 4°C in the dark) to quantify the natural concentrations of volatile S and Se compounds. A second aliquot (referred to as 'incubated aliquots') was spiked with 100 µL of a 100 mM sodium selenite solution to yield a final Se concentrations of ~1mM, placed in a Multitron incubator

166 (Infors-HT, Bottmingen, Switzerland) in the dark at 25°C, and analyzed after 48 h. Before
167 analysis, the DI-SPME-GC/MS method was calibrated (on a daily basis) using aqueous standards
168 with known amounts of analytes and internal standards that were added as described above. The
169 accuracy of the method was verified by spiking measured samples with known amounts of
170 analytes, according to the method of standard addition.

3. Results and discussion

3.1. Optimization of instrumental parameters

An overview of the obtained optimal instrumental settings for the DI-SPME procedure is provided in Table 1. Out of eight investigated SPME fiber coatings, the 75 μm CAR/PDMS phase delivered the highest extraction efficiencies for most of the investigated compounds (Figure 2), which concurs with previous studies [32,35].

Efficient extraction of analytes with a boiling point below 50°C (e.g., DMS) was achieved with extraction temperatures up to 50°C (Figure 3A). However, volatile analytes showed a decreased response with extraction temperatures exceeding 50°C, which is potentially due to outgassing of volatile analytes through the septum puncture during extraction [43]. The response of analytes with intermediate boiling points (e.g., EMS and DMSe, between 50–80°C) was not much affected by an increasing extraction temperature, whereas the responses of compounds with a boiling point above 80°C (e.g., DMDSe and DEDS) increased significantly with higher extraction temperatures (Figure 3A). A compromise extraction temperature of 50°C was chosen.

The responses of all investigated compounds increased with longer extraction times (Figure 3B). Although equilibrium between the fiber and the sample was probably not completely attained within the investigated timespan, an extraction time of 50 min was chosen to avoid excessive extraction time. The optimal instrumental settings for extraction reported in this study (Table 1) indicate that a longer extraction at higher temperatures is necessary for direct-immersion extraction of volatile Se and S compounds compared to headspace-SPME (HS-SPME) [30,31,33,34,36,38,43,44].

Increasing analyte extraction efficiencies and decreasing peak tailing factors were observed with increasing desorption temperatures (Figure 3C). The observed tendency of the analyte responses (particularly that of DMS) to plateau after desorption temperatures higher than 250°C indicate that near complete desorption (equilibration) was reached (Figure 3C). However, higher desorption temperatures also increased the background noise and thereby potentially deteriorated the detection limits: “bleeding” of polysiloxanes from the SPME-fiber, septum, and/or column at desorption temperatures > 285°C induced a 50-fold increase in the background noise at certain analyte target masses (data not shown). Therefore, a desorption temperature of 260°C was chosen as a compromise between high responses and minimal column bleeding and peak tailing factors.

The analyte responses consistently increased with increasing desorption times up to 4 min and leveled off thereafter (Figure 3D). A desorption time of 5 min was chosen to ensure complete transfer of the analytes from the SPME fiber to the GC column and thereby prevent carry-over to subsequent samples. The transformation of analytes during extraction or desorption, e.g., oxidation of DMS to DMSO or of ESH to DEDS [17,51] was not observed at the investigated extraction and desorption temperatures.

3.2. Optimization of sample parameters

A variation in sample pH between 3.5 and 8 only marginally affected the SPME extraction efficiency (Figure 4A). This can be explained by the fact that the investigated substances are not or only weakly deprotonated ($pK_a > 30$) and because the CAR/PDMS fiber coating is relatively inert to changes in acidity [28]. A slight decrease in the extraction efficiency was observed with an increasing organic matter (OM) concentration in the samples (Figure 4B), which may be the result of analyte binding to OM or sorption of OM to the fiber coating and thus competition with the analytes [52,53]. The observed reduction in extraction of volatile Se and S species with

increasing levels of OM corroborates studies where proteins decreased the extraction of DMDS [35]. Because the typical concentration range of OM in natural surface waters is 0.1–10 mg/L (and up to 50 mg/L in peat waters) [54], and because volatiles may bind to OM (e.g., DMSe to OM-rich soils [55]), it is important to consider the OM content of an (environmental) sample and to mimic the OM content in calibration standards, if feasible.

Increasing salt concentrations in the samples deteriorated the extraction efficiencies of the investigated analytes (Figure 4C). In addition, higher salt contents (>1.5 M) worsened the reproducibility and caused salt-crystallization around the septa puncture, in the liner and in the pre-column. This precipitation of salt provoked shifts in the analyte retention times and caused corrosion of the fiber, thereby shortening the fiber lifetime. Although the addition of salt may enhance the efficiency of headspace extraction of volatile analytes [28,35], salt addition was considered not beneficial for the direct-immersion extraction of the investigated analytes.

Decreasing extraction efficiencies were also observed with increasing sample methanol content (Figure 4D). The response of more apolar compounds (e.g., p-XYL-d₁₀ and TOL-d₈ with dipole moments < 0.5 D) decreased more rapidly with increasing methanol content compared to the response of more polar compounds (e.g., DES and DMSe with dipole moments ~1.5 D). Although methanol is unlikely to occupy sites on the CAR/PDMS-fiber coating because of its hydrophilicity (its octanol-water partition coefficient (logK_{ow}) is -0.69), it may act as a co-solvent for analytes and thus reduce analyte extraction efficiency [28]. It has been demonstrated that the presence of a competing solvent (e.g., ethanol) deteriorates the extraction of volatile Se and S species from the headspace overlying aqueous samples [43], although this effect may be less pronounced at solvent concentrations below 5–19% [32]. These observations agree with our observation (Figure 4D) that increased methanol levels reduce the extraction efficiency of

volatile Se and S species. Because some of the investigated analytes and internal standards (e.g., DMDSe, TOL-d₈, and XYL-d₁₀) are poorly soluble in water, stock solutions needed to be prepared in organic solvents. Therefore, these stock solutions were prepared at high concentrations (>50 mg/L) to minimize the amount of organic solvent to <0.1% in all calibration standards and samples.

3.3. Stability of analytes in stock solutions

Recurrent analysis of stored stock solutions that contained multiple analytes indicated that the concentrations of the analytes with S-S and Se-Se bonds decreased over the course of days and that additional compounds were formed. In a stock solution containing DMDSe and DMDS, a third peak emerged after four days of storage (Figure 5A). The corresponding mass spectrum indicates that the mixed Se-S compound dimethyl selenyl sulfide (DMSeS) was formed, which suggests the following chalcogenide exchange reaction to occur [56,57]:

In a stock solution containing both DMDSe and ESH, two additional peaks emerged after eight days of storage (Figure 5B). Retention times and mass spectra indicated that these peaks most likely belonged to DEDS and ethylmethyl selenyl sulfide (ESSeM). The formation of DEDS may be caused by the oxidation of ethane thiol [51] via:

The formation of ESSeM may be caused by a subsequent chalcogenide exchange between DEDS and DMDSe via:

259 The abovementioned oxidation and chalcogenide exchange reactions occurred exclusively in
 260 concentrated stock solutions in methanol and were not observed in the diluted aqueous working
 261 solutions. Although more investigation is necessary to constrain the exact mechanisms and
 262 kinetics of transformation under the storage conditions used in this study, chalcogenide exchange
 263 reactions between volatile Se and S compounds and oxidation reactions of thiols have been
 264 previously documented [44,57-59] and may lead to degradation of standards and generation of
 265 mixed Se-S compounds. Therefore, the daily preparation of fresh stock solutions is necessary to
 266 guarantee that analyte transformation reactions do not affect the calibration.

267 *3.4. Assessment of the method performance*

268 *3.4.1. Analytical figures of merit*

269 The performance of the final DI-SPME-GC/MS method was evaluated by analysis of pure
 270 water samples and by application to natural water and wastewaters. The extraction efficiencies of
 271 analytes in the DI-SPME method were calculated from responses of standards in ultrapure water,
 272 which were quantified from the calibrations made with on-column injection of standards
 273 dissolved in organic solvents. The analyte extraction efficiencies were variable and reached up to
 274 96% for DMSe (Table 2). The enrichment factors and distribution constants of analytes between
 275 the fiber and the sample (K_{fs}) in the final DI-SPME method were determined from the ratio of
 276 analyte concentrations in the fiber coating (determined by calibration with direct on-column
 277 injections) and the analyte concentration in pure water standards. The enrichment factors
 278 (between 59 for DPS and 1074 for DMSe) and $\log K_{fs}$ values (between 1.8 for DPS and MPS and
 279 3.0 for DMSe and EMS, see Table 2) were high but also variable: this variability is due to the

different properties of the investigated compounds (i.e., different affinities to the CAR/PDMS fiber). In general, hydrophobic compounds (high $\log K_{ow}$ values) and compounds with a high boiling point had lower extraction efficiencies and thus lower $\log K_{fs}$ values (see Figure S2 in the Supplementary Material).

The limits of detection for the final DI-SPME-GC/MS method are given by i) the instrumental detection limit (IDL, blank equivalent concentration + $3 \times \sigma$ of blank using an on-column injected calibration), which reflects the lowest amount that the instrumentation can reliably detect and thus the sensitivity of the GC/MS, ii) the limit of detection (LOD, equivalent to the IDL but with incorporation of the DI-SPME efficiency and calculated in relation to the sample volume), and iii) by the method detection limit (MDL, $3 \times \sigma$ of a sample near the detection limit), which is based on replicates of the entire DI-SPME-GC/MS procedure and thus also includes any errors caused during sample preparation. These figures of merit are summarized for all the investigated compounds in Table 2.

The IDL values ranged between 2.5 pg (DEDS) and 43 pg (DMSe); LOD values were between 4 ng/L (DMS) and 47 ng/L (DES). The overall MDL values in ultrapure water (using internal standards in a consistent solvent content) varied between 30 ng/L (DMS) and 240 ng/L (MPS). Calibration curves indicated that most analyte responses were linear between their MDLs and 100 $\mu\text{g/L}$, depending on the analyte (Figure S3 in Supplementary Material). Additional tests showed that the fiber reached saturation for most analytes at concentrations higher than 1 mg/L, corresponding to the absolute amount of 10 μg analyte (equivalent of ~ 10 wt-% of the fiber coating, data not shown).

The obtained detection limits in this study are compared to other SPME-based methods in Supplementary Material Table S3. They are in the same range as most headspace-SPME methods

for volatile S and Se studies [33,35,43] but are up to 70 times improved for some volatile Se and S species compared to other DI-SPME methods [32,40]. Since compromises were made in the optimization process, improved quantification limits may be achieved when single analytes are investigated. The coupling of SPME with GC-ICP/MS may lead to improved sensitivity for Se [44,60], but the sensitivity for S is not expected to improve due to plasma-generated interferences on S masses. Other analytical methods for volatile S and Se species (e.g., solid-phase [58] and liquid phase-extraction [27]) occasionally report better detection limits, but in turn often require solvents for extraction, a larger sample volume, or (multiple) drying and/or derivation steps.

3.4.2. Application to natural water and wastewater samples

The applicability and robustness of the presented DI-SPME-GC/MS method were tested through analyte quantification in original and incubated peat bog water and wastewaters. In both the original peat bog water and wastewaters, volatile alkylated S species were detected: DMS was the most abundant species (up to 262 ng/L), and DMDS was present at concentrations up to 80 ng/L (see Table 3). Volatile alkylated Se species were not detected in the original peat bog water or wastewater samples. In contrast, the incubated aqueous samples (supplemented with inorganic selenite) contained significant amounts of volatile alkylated Se after 48 h of incubation: up to 1810 ng/L DMSe and 450 ng/L DMDS were detected. Furthermore, incubation of the samples led to increased concentrations of volatile alkylated S species: up to 903 ng/L DMS and 144 ng/L DMDS. Both original and incubated samples were spiked with known amounts of analyte according to the method of standard addition. The consistently high recoveries of the spikes (between 77% – 104%, see Table 3) illustrate that the analytes were reliably quantified in the investigated samples.

Although earlier studies have indicated that biological alkylation of S and Se may lead to the formation of ethylated and mixed methyl-ethylated species [61], no higher-alkylated volatile

species (i.e., ESH, DEDS, DES, EMS, DPS, or MPS) were detected in either the original or the incubated wastewaters and peat bog water. In addition to the species listed in Table 3, the mixed S-Se species DMSeS was identified in the incubated wastewaters and peat bog water. Using a linear interpolation of the calibration curves of DMDS and DMDS_e, the species DMSeS was estimated to be present at up to 130 ng/L and 200 ng/L in the incubated wastewater and peat bog water, respectively. Although a calibration with a DMSeS standard (synthesis described in [56]) would be required to reliably quantify the concentration of DMSeS, these quantification estimates suggest that DMSeS is an important natural alkylation product. This observation of DMSeS being a major biogenic Se species agrees with observations of DMSeS in natural environments [7,62], but it should be noted that DMSeS in our experiments may also have originated from the abiotic chalcogenide exchange reaction between DMDS_e and DMDS during incubation (Figure 5A).

3.5. Headspace extraction versus direct-immersion extraction

The figures of merit presented in this study demonstrate that DI-based SPME yields detection limits and repeatability comparable to HS-SPME based methods (Table S3). However, there are important advantages of DI-SPME over HS-SPME.

For example, HS-SPME procedures (e.g., for the analysis of volatile Se and S compounds in wine [17,43]) often show decreasing extraction efficiency with increasing solvent content: HS-SPME methods therefore rely on a consistent solvent content in both the calibrations and samples or on the addition of salt (which forces the equilibrium of dissolved volatiles to the gaseous phase but adds an additional sample treatment step). Here we showed that the content of organic solvent can also be a critical parameter in DI-SPME methods. However, our study illustrates that, even in the case of turbid aqueous samples, additional sample preparation steps (e.g., filtration, sample dilution [17], addition of salt, or addition of chemical suppressors to eliminate interferences [16]) are not required to achieve high sensitivity and selectivity with DI-SPME.

351 An often-mentioned advantage of HS-SPME over DI-SPME may be the increased fiber
352 lifetime in the headspace mode, and several techniques were proposed to improve DI-based
353 extraction of analytes from heavily contaminated waters (e.g., membranes to reduce the
354 interference of OM [26]). However, the presented method indicates that the SPME-fiber lifetime
355 is mainly affected by the number of vial- and GC septum-punctures and general mechanical
356 wearing through agitation and not by the presence of turbidity in a sample. In this regard, direct
357 immersion of the SPME-fiber may even add an additional stirring function and enhances
358 homogenization and thus formation of equilibrium. Further advantages of DI-SPME over HS-
359 SPME include a higher amount of soluble analytes in the aqueous phase, a reduced loss of
360 headspace gas (and thus analyte) through the septum puncture, particularly at elevated extraction
361 temperatures, and the fact that biological alkylation occurs in the water phase: this places
362 extraction close to the source of analyte production, which is particularly important when the
363 produced volatile species are highly reactive [21,22].

4. Conclusions

The optimization of the SPME parameters conducted in this study illustrates that sample parameters (in particular the solvent- and organic matter content) are at least as important as instrumental parameters for the performance of the SPME procedure. Aqueous samples coming from natural waters and industrial effluents may potentially contain a range of matrix components (e.g., solvents, organic matter) that can lead to interferences during analysis. However, it is unrealistic to exactly reproduce the matrices of natural- and industrial samples in calibration standards as these matrices are often unknown and may vary with time. The high recoveries of the analyte-spikes added to our field samples (Table 3) show that with conventional instrumentation, the use of internal standards, and with a careful sample-parameter evaluation, sample matrix effects do not impede a sensitive quantification of volatile S and Se compounds in complex aqueous samples. The high degree of repeatability of analyte quantification in wastewaters and peat bog water (Table 3) illustrates that the presented method is robust, reproducible, and sensitive enough for the trace-level quantification of volatile Se and S species in complex samples. Therefore, the presented method is potentially also suitable for the quantification of volatile S and Se compounds in a variety of sample matrices such as algal slurries, industrial effluents, biological specimens, and food and beverage samples.

Supplementary Material

Supplementary Figures S1–S3 and Tables S1–S3.

Acknowledgements

We acknowledge C. Stengel for general experimental assistance and A. Castrischer and J. Bolotin for assistance with the SPME-GC/MS analyses. We thank B. Sinnet for sample collection at Eawag's wastewater treatment facility. P. Poggiati and M. Sulmoni from the Department of Environment (Ticino, Switzerland) are acknowledged for permission to collect the field samples at Gola di Lago.

Figures

Figure 1.

393 **Figure 2.**

394

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396 **Figure 3.**

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398 **Figure 4.**

401 **Figure 5.**

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403

404 **Tables**

405 **Table 1.** Optimized instrumental settings for DI-SPME of the investigated analytes

Parameter	Setting
SPME fiber	75 μ m Carboxen/PDMS
Extraction time	50 min
Extraction temperature	50°C
Desorption time	5 min
Desorption temperature	260°C

406

407 **Table 2.** Analytical figures of merit

Analyte	IDL (pg)	Internal standard	Extraction efficiency (%)	Enrichment factor (%)	LogK _{fs}	LOD (ng/L)	MDL (ng/L)
DMS	6	DMS-d ₆	77 ± 5	806	2.9 ± 0.0	4	30
DMSe	43	DMS-d ₆	96 ± 8	1074	3.0 ± 0.1	24	75
EMS	11	DMS-d ₆	86 ± 2	976	3.0 ± 0.1	5	35
ESH	15	DMS-d ₆	78 ± 7	850	2.9 ± 0.0	10	80
DES	42	DPE, MTBE	44 ± 1	485	2.7 ± 0.0	47	190
DMDS	12	DPE, MTBE	22 ± 2	178	2.3 ± 0.0	28	35
DMDS _e	13	DPE, MTBE	24 ± 4	223	2.3 ± 0.1	27	36
DEDS	3	XYL-d ₁₀ , TOL-d ₈	17 ± 4	123	2.1 ± 0.1	7	51
DPS	3	XYL-d ₁₀ , TOL-d ₈	11 ± 0	59	1.8 ± 0.1	14	225
MPS	5	XYL-d ₁₀ , TOL-d ₈	12 ± 1	67	1.8 ± 0.0	17	240

408 Instrumental detection limit (IDL), extraction efficiency, enrichment factor, fiber-sample
 409 distribution coefficient (K_{fs}), limit of detection (LOD), and method detection limit (MDL) of the
 410 investigated analytes. For the method characterization, all target analytes were measured on their
 411 target ion mass (Supplementary Material Table S2) in SIM mode in 10 mL vials in ultrapure
 412 water. Standard deviations from triplicate replication.

Table 3. Application of the presented method to complex aqueous samples

Peat bog water	Original aliquot			Incubated aliquot		
	Measured concentration (ng/L)	Spiked concentration (ng/L)	Recovery (%)	Measured concentration (ng/L)	Spiked concentration (ng/L)	Recovery (%)
DMS	170 ± 40	850	104 ± 9	903 ± 32	850	100 ± 10
DMS _e	30 ± 20*	1410	83 ± 11	1810 ± 480	1410	92 ± 6
EMS	< LOD	-	-	< LOD	-	-
ESH	< LOD	-	-	12 ± 10*	-	-
DES	< LOD	-	-	< LOD	-	-
DMDS	80 ± 42	1060	101 ± 15	144 ± 31	1060	94 ± 7
DMDS _e	< LOD	1980	92 ± 4	375 ± 91	1980	103 ± 9
DEDS	< LOD	-	-	< LOD	-	-
DPS	< LOD	-	-	< LOD	-	-
MPS	< LOD	-	-	< LOD	-	-
Wastewater aerobic sludge	Original aliquot			Incubated aliquot		
	Measured concentration (ng/L)	Spiked concentration (ng/L)	Recovery (%)	Measured concentration (ng/L)	Spiked concentration (ng/L)	Recovery (%)
DMS	190 ± 100	-	-	601 ± 98	-	-
DMS _e	< LOD	-	-	320 ± 230	-	-
EMS	< LOD	840	95 ± 5	< LOD	840	93 ± 7
ESH	13 ± 12*	860	98 ± 4	15 ± 10*	860	102 ± 11
DES	< LOD	840	85 ± 15	< LOD	840	89 ± 5
DMDS	34 ± 18*	1060	105 ± 8	83 ± 1	1060	104 ± 9
DMDS _e	< LOD	-	-	98 ± 56	-	-
DEDS	< LOD	980	83 ± 10	< LOD	980	106 ± 13
DPS	< LOD	-	-	< LOD	-	-
MPS	< LOD	-	-	< LOD	-	-
Wastewater anaerobic sludge	Original aliquot			Incubated aliquot		
	Measured concentration (ng/L)	Spiked concentration (ng/L)	Recovery (%)	Measured concentration (ng/L)	Spiked concentration (ng/L)	Recovery (%)
DMS	262 ± 94	850	91 ± 6	750 ± 28	850	104 ± 13
DMS _e	< LOD	1410	86 ± 3	1500 ± 100	1410	90 ± 3
EMS	< LOD	-	-	< LOD	-	-
ESH	17 ± 10*	-	-	19 ± 7*	-	-
DES	< LOD	840	89 ± 9	< LOD	840	103 ± 8
DMDS	52 ± 19	-	-	102 ± 36	-	-
DMDS _e	< LOD	1980	94 ± 6	450 ± 61	1980	95 ± 4
DEDS	< LOD	-	-	< LOD	-	-
DPS	< LOD	840	85 ± 6	< LOD	840	77 ± 20
MPS	< LOD	1120	94 ± 5	< LOD	1120	86 ± 9

414 Measured concentrations, spiked concentrations, and spike recoveries of the investigated analytes
 415 in the original (left) and incubated (right) aliquots of peat bog water and wastewaters. < LOD;
 416 below detection limit, - ; not determined, * below MDL but above LOD (see LOD and MDL
 417 values in Table 2). Indicated standard deviations from analysis of triplicate samples.

References

- [1] A. Fernandez-Martinez, L. Charlet, Selenium environmental cycling and bioavailability: A structural chemist point of view. *Rev. Environ. Sci. Biotechnol.* 8 (2009) 81-110.
- [2] L.A. Wessjohann, A. Schneider, M. Abbas, W. Brandt, Selenium in chemistry and biochemistry in comparison to sulfur. *Biol. Chem.* 388 (2007) 997-1006.
- [3] R. Bentley, T.G. Chasteen, Environmental vocs—formation and degradation of dimethyl sulfide, methanethiol and related materials. *Chemosphere* 55 (2004) 291-317.
- [4] L. Winkel, B. Vriens, J.G. Jones, L. Schneider, E.A.H. Pilon-Smits, G.S. Bañuelos, Selenium cycling across soil-plant-atmosphere interfaces: a critical review. *Nutrients* 7 (2015) 4199-4239.
- [5] J.S. Thayer, Review: Biological methylation of less-studied elements. *Appl. Organomet. Chem.* 16 (2002) 677-691.
- [6] T.G. Chasteen, R. Bentley, Biomethylation of selenium and tellurium: Microorganisms and plants. *Chem. Rev.* 103 (2003) 1-25.
- [7] D. Amouroux, P.S. Liss, E. Tessier, M. Hamren-Larsson, O.F.X. Donard, Role of oceans as biogenic sources of selenium. *Earth. Planet. Sci. Lett.* 189 (2001) 277-283.
- [8] A.J. Kettle, T.S. Rhee, M. von Hobe, A. Poulton, J. Aiken, M.O. Andreae, Assessing the flux of different volatile sulfur gases from the ocean to the atmosphere. *J. Geophys. Res. Atmos.* 106 (2001) 12193-12209.
- [9] S.F. Watts, The mass budgets of carbonyl sulfide, dimethyl sulfide, carbon disulfide and hydrogen sulfide. *Atmos. Environ.* 34 (2000) 761-779.
- [10] H. Wen, J. Carignan, Reviews on atmospheric selenium: Emissions, speciation and fate. *Atmos. Environ.* 41 (2007) 7151-7165.
- [11] F. Dincer, A. Muezzinoglu, Odor-causing volatile organic compounds in wastewater treatment plant units and sludge management areas. *J. Env. Sc. Health A.* 43 (2008) 1569-1574.

- [12] Y. Sheng, F. Chen, X. Wang, G. Sheng, J. Fu, Odorous Volatile Organic Sulfides in Wastewater Treatment Plants in Guangzhou, China. *Water Environ. Res.* 80 (2008) 324-330.
- [13] H. Weenen, W.E. Koolhaas, A. Apriyantono, Sulfur-Containing Volatiles of Durian Fruits (*Durio zibethinus* Murr.). *J. Agric. Food. Chem.* 44 (1996) 3291-3293.
- [14] H.M. Burbank, M.C. Qian, Volatile sulfur compounds in Cheddar cheese determined by headspace solid-phase microextraction and gas chromatograph-pulsed flame photometric detection. *J. Chromatogr. A* 1066 (2005) 149-157.
- [15] P. Molimard, H.E. Spinnler, Review: Compounds Involved in the Flavor of Surface Mold-Ripened Cheeses: Origins and Properties. *J. Dairy Sci.* 79 (1996) 169-184.
- [16] Y. Fang, M.C. Qian, Sensitive quantification of sulfur compounds in wine by headspace solid-phase microextraction technique. *J. Chromatogr. A* 1080 (2005) 177-185.
- [17] R. López, A.C. Lapeña, J. Cacho, V. Ferreira, Quantitative determination of wine highly volatile sulfur compounds by using automated headspace solid-phase microextraction and gas chromatography-pulsed flame photometric detection: Critical study and optimization of a new procedure. *J. Chromatogr. A* 1143 (2007) 8-15.
- [18] M. Mestres, O. Busto, J. Guasch, Analysis of organic sulfur compounds in wine aroma. *J. Chromatogr. A* 881 (2000) 569-581.
- [19] P.A. Vazquez-Landaverde, J.A. Torres, M.C. Qian, Quantification of Trace Volatile Sulfur Compounds in Milk by Solid-Phase Microextraction and Gas Chromatography-Pulsed Flame Photometric Detection. *J. Dairy Sci.* 89 (2006) 2919-2927.
- [20] S.K. Pandey, K.-H. Kim, A Review of Methods for the Determination of Reduced Sulfur Compounds (RSCs) in Air. *Environ. Sci. Technol.* 43 (2009) 3020-3029.
- [21] W. Wardencki, Problems with the determination of environmental sulphur compounds by gas chromatography. *J. Chromatogr. A* 793 (1998) 1-19.

- [22] X. Dauchy, M. Potingautier, A. Astruc, M. Astruc, Analytical methods for the speciation of selenium compounds - A review. *Fresenius J. Anal. Chem.* 348 (1994) 792-805.
- [23] C. Herrero Latorre, J. Barciela García, S. García Martín, R.M. Peña Crecente, Solid phase extraction for the speciation and preconcentration of inorganic selenium in water samples: A review. *Anal. Chim. Acta* 804 (2013) 37-49.
- [24] D. Juresa, D. Kuehnelt, K.A. Francesconi, Consequences of Vapor Enhancement on Selenium Speciation Analysis by HPLC/ICPMS. *Anal. Chem.* 78 (2006) 8569-8574.
- [25] C. Pecheyran, D. Amouroux, O.F.X. Donard, Field determination of volatile selenium species at ultra trace levels in environmental waters by on-line purging, cryofocusing and detection by atomic fluorescence spectroscopy. *J. Anal. At. Spectrom.* 13 (1998) 615-621.
- [26] E. Ghasemi, M. Sillanpaa, N.M. Najafi, Headspace hollow fiber protected liquid-phase microextraction combined with gas chromatography-mass spectroscopy for speciation and determination of volatile organic compounds of selenium in environmental and biological samples. *J. Chromatogr. A* 1218 (2011) 380-386.
- [27] M. Hashemi, R.A. Montes, T.G. Chasteen, Determination of volatile organochalcogens using liquid phase microextraction and gas chromatography with fluorine-induced chemiluminescence detection. *Environ. Biotechnol.* 8 (2012) 55-62.
- [28] Z. Mester, Sturgeon, R., Pawliszyn, J., Solid phase microextraction as a tool for trace element speciation. *Spectrochim. Acta B* 56 (2001) 233-260.
- [29] C. Haberhauer-Troyer, E. Rosenberg, M. Grasserbauer, Evaluation of solid-phase microextraction for sampling of volatile organic sulfur compounds in air for subsequent gas chromatographic analysis with atomic emission detection. *J. Chromatogr. A* 848 (1999) 305-315.
- [30] F. Lestremay, V. Desauziers, J.C. Roux, J.L. Fanlo, Development of a quantification method for the analysis of malodorous sulphur compounds in gaseous industrial effluents

by solid-phase microextraction and gas chromatography-pulsed flame photometric detection. *J. Chromatogr. A* 999 (2003) 71-80.

[31] K.C. Li, D. Shooter, Analysis of sulfur-containing compounds in ambient air using solid-phase microextraction and gas chromatography with pulsed flame photometric detection. *Int. J. Environ. Anal. Chem.* 84 (2004) 749-760.

[32] M. Mestres, M.P. Martí, O. Busto, J. Guasch, Analysis of low-volatility organic sulphur compounds in wines by solid-phase microextraction and gas chromatography. *J. Chromatogr. A* 881 (2000) 583-590.

[33] A.T. Nielsen, S. Jonsson, Quantification of volatile sulfur compounds in complex gaseous matrices by solid-phase microextraction. *J. Chromatogr. A* 963 (2002) 57-64.

[34] M. Bueno, F. Pannier, Quantitative analysis of volatile selenium metabolites in normal urine by headspace solid phase microextraction gas chromatography-inductively coupled plasma mass spectrometry. *Talanta* 78 (2009) 759-763.

[35] N. Campillo, R. Peñalver, M. Hernández-Córdoba, Determination of dimethylselenide and dimethyldiselenide in milk and milk by-products by solid-phase microextraction and gas chromatography with atomic emission detection. *Talanta* 80 (2010) 1856-1861.

[36] C. Dietz, J. Sanz Landaluze, P. Ximenez-Embun, Y. Madrid-Albarran, C. Camara, SPME-multicapillary GC coupled to different detection systems and applied to volatile organo-selenium speciation in yeast. *J. Anal. At. Spectrom.* 19 (2004) 260-266.

[37] C. Dietz, T. Pérez-Corona, Y. Madrid-Albarrán, C. Cámara, J. Anal. At. Spectrom. SPME for on-line volatile organo-selenium speciation. *J. Anal. At. Spectrom.* 18 (2003) 467-473.

[38] C. Dietz, J. Sanz Landaluze, P. Ximénez-Embún, Y. Madrid-Albarrán, C. Cámara, Volatile organo-selenium speciation in biological matter by solid phase microextraction-moderate temperature multicapillary gas chromatography with microwave induced plasma atomic emission spectrometry detection. *Anal. Chim. Acta* 501 (2003) 157-167.

- [39] K.M. Kubachka, J. Meija, D.L. LeDuc, N. Terry, J.A. Caruso, Selenium Volatiles as Proxy to the Metabolic Pathways of Selenium in Genetically Modified *Brassica juncea*. *Environ. Sci. Technol.* 41 (2007) 1863-1869.
- [40] M. Lenz, E.D.van Hullebusch, F. Farges, S. Nikitenko, P.F.X. Corvini, P.N.L. Lens, Combined Speciation Analysis by X-ray Absorption Near-Edge Structure Spectroscopy, Ion Chromatography, and Solid-Phase Microextraction Gas Chromatography Mass Spectrometry To Evaluate Biotreatment of Concentrated Selenium Wastewaters. *Environ. Sci. Technol.* 45 (2011) 1067-1073.
- [41] J. Meija, J.M. Bryson, A.P. Vonderheide, M. Montes-Bayón, J.A. Caruso, Studies of Selenium-Containing Volatiles in Roasted Coffee. *J. Agric. Food. Chem.* 51 (2003) 5116-5122.
- [42] J. Sanz Landaluze, C. Dietz, Y. Madrid, C. Cámara, Volatile organoselenium monitoring in production and gastric digestion processes of selenized yeast by solid-phase microextraction-multicapillary gas chromatography coupled microwave-induced plasma atomic emission spectrometry. *Appl. Organomet. Chem.* 18 (2004) 606-613.
- [43] N. Campillo, R. Peñalver, I. López-García, M. Hernández-Córdoba, Headspace solid-phase microextraction for the determination of volatile organic sulphur and selenium compounds in beers, wines and spirits using gas chromatography and atomic emission detection. *J. Chromatogr. A* 1216 (2009) 6735-6740.
- [44] J. Meija, M. Montes-Bayón, D.L. LeDuc, N. Terry, J.A. Caruso, Simultaneous Monitoring of Volatile Selenium and Sulfur Species from Se Accumulating Plants (Wild Type and Genetically Modified) by GC/MS and GC/ICPMS Using Solid-Phase Microextraction for Sample Introduction. *Anal. Chem.* 74 (2002) 5837-5844.
- [45] N. Campillo, R. Penalver, M. Hernandez-Cordoba, C. Perez-Sirvent, M.J. Martinez-Sanchez, Comparison of two derivatizing agents for the simultaneous determination of selenite and organoselenium species by gas chromatography and atomic emission detection

after preconcentration using solid-phase microextraction. *J. Chromatogr. A* 1165 (2007) 191-199.

[46] E. Dimitrakakis, C. Haberhauer-Troyer, Y. Abe, M. Ochsenkühn-Petropoulou, E. Rosenberg, Solid-phase microextraction–capillary gas chromatography combined with microwave-induced plasma atomic-emission spectrometry for selenite determination. *Anal. Bioanal. Chem.* 379 (2004) 842-848.

[47] D.C. Kapsimali, G.A. Zachariadis, Headspace and direct immersion solid phase microextraction procedures for selenite determination in urine, saliva and milk by gas chromatography mass spectrometry. *J. Chromatogr. B* 877 (2009) 3210-3214.

[48] D.C. Kapsimali, G.A. Zachariadis, Comparison of tetraethylborate and tetraphenylborate for selenite determination in human urine by gas chromatography mass spectrometry, after headspace solid phase microextraction. *Talanta* 80 (2010) 1311-1317.

[49] P. Shahdousti, N. Alizadeh, Headspace-solid phase microextraction of selenium(IV) from human blood and water samples using polypyrrole film and analysis with ion mobility spectrometry. *Anal. Chim. Acta* 684 (2011) 67-71.

[50] B. Vriens, M. Lenz, L. Charlet, M. Berg, L. Winkel, Natural wetland emissions of methylated trace elements. *Nature Communications* 5 (2014) 3035.

[51] F. Lestremat, F.A.T. Andersson, V. Desauziers, Investigation of artefact formation during analysis of volatile sulphur compounds using solid phase microextraction (SPME). *Chromatographia* 59 (2004) 607-613.

[52] F.S. Higashikawa, M.L. Cayuela, A. Roig, C.A. Silva, M.A. Sánchez-Monedero, Matrix effect on the performance of headspace solid phase microextraction method for the analysis of target volatile organic compounds (VOCs) in environmental samples. *Chemosphere* 93 (2013) 2311-2318.

- [53] A.L. Simplicio, L. Vilas Boas, Validation of a solid-phase microextraction method for the determination of organophosphorus pesticides in fruits and fruit juice. *J. Chromatogr. A* 833 (1999) 35-42.
- [54] E.M. Thurman, Amount of Organic Carbon in Natural Waters, in: *Organic Geochemistry of Natural Waters*, Springer Netherlands, 1985, pp. 7-65.
- [55] R. Zieve, P.J. Peterson, Sorption of dimethylselenide by soils. *Soil Boil. Biochem.* 17 (1985) 105-107.
- [56] T.G. Chasteen, Confusion between dimethyl selenenyl sulfide and dimethyl selenone released by bacteria. *Appl. Organomet. Chem.* 7 (1993) 335-342.
- [57] J. Meija, J.A. Caruso, Selenium and Sulfur Trichalcogenides from the Chalcogenide Exchange Reaction. *Inorg. Chem.* 43 (2004) 7486-7492.
- [58] J.L. Gomez-Ariza, I. Giraldez, E. Morales, J.A. Pozas, Use of solid phase extraction for speciation of selenium compounds in aqueous environmental samples. *Analyst* 124 (1999) 75-78.
- [59] M. Klein, H. Preud'homme, M. Bueno, F. Pannier, Study of volatile selenium metabolites stability in normal urine: effects of sample handling and storage conditions. *J. Anal. At. Spectrom.* 26 (2011) 602-607.
- [60] J. Duan, X. Li, C. Yu, B. Hu, Headspace stir bar sorptive extraction combined with GC-ICP-MS for the speciation of dimethylselenide and dimethyldiselenide in biological samples. *J. Anal. At. Spectrom.* 24 (2009) 297-303.
- [61] P. Pinel-Raffaitin, C. Pécheyran, D. Amouroux, New volatile selenium and tellurium species in fermentation gases produced by composting duck manure. *Atmos. Environ.* 42 (2008) 7786-7794.
- [62] E. Tessier, D. Amouroux, G. Abril, E. Lemaire, O.X. Donard, Formation and volatilisation of alkyl-iodides and -selenides in macrotidal estuaries. *Biogeochemistry* 59 (2002) 183-206.

Figure Captions

Figure 1.

Chromatographic separation of volatile Se and S compounds using the GC/MS method. The relative intensity of the 30 compounds (corresponding numbers listed below, abbreviations in Table 2) was obtained by normalizing the chromatogram to the highest response obtained with a composite standard solution. Compounds indicated with an asterisk were used as analytes or internal standards in this study; their retention factors and details of calibration are given in Table 2.

Figure 2.

Evaluation of different SPME fiber coatings. The responses of analytes (normalized to the highest response obtained for individually measured analytes) were compared after extraction with different SPME-fiber coatings (all other parameters were kept constant, see Table 1). Error bars indicate standard deviations of analysis of triplicate samples.

Figure 3.

Optimization of instrumental SPME parameters. Plotted are the responses of selected compounds (normalized to the highest response obtained within each optimization) and the corresponding trends against A) the investigated extraction temperature range, B) the investigated extraction time range, C) the investigated desorption temperature range, and D) the investigated desorption time range. All other parameters were kept constant during each optimization. The legend applies to all panels, the trend lines serve illustrative purposes only, and the error bars indicate standard deviations of analysis of triplicate samples. The peak tailing factor is plotted (frame C, right vertical axis) against the investigated desorption temperature range after calculation of peak asymmetry (using the front-width and tail-width of peaks at 5% of its

maximum height) for p-XYL-d₁₀, DMS, and ESH (error bars in this case indicate the standard deviation over these three compounds).

Figure 4.

Optimization of SPME sample parameters. Responses of selected compounds (normalized to the highest response obtained within each optimization) and the corresponding trend lines are plotted against A) sample pH (adapted using 10 mM phosphate buffer), B) sample organic matter content, C) sample salt content, and D) sample methanol content. All other sample parameters were kept constant (Table 1) during each optimization. The legend applies to all panels, the trend lines serve illustrative purposes only, and the error bars indicate standard deviations of triplicate samples.

Figure 5.

Transformation of analytes during prolonged storage of stock solutions. The solid lines (green peaks) are composite chromatograms of the characteristic masses of DMDS and DMDSe (panel A) and of the characteristic masses of ESH and DMDSe (panel B) in fresh stock solutions. The dashed lines (red peaks) are composite chromatograms of the characteristic masses of the observed exchange products after storing a combined stock solution for four (panel A) and eight days (panel B). The mass spectra of the transformation products are shown in the yellow frames.