Simultaneous determination of chlorinated ethenes and ethene in groundwater using headspace solid phase microextraction with gas chromatography

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Abstract

Widespread contamination of groundwater by chlorinated ethenes and their biological dechlorination products necessitates reliable monitoring of liquid matrices; current methods approved by the U.S. Environmental Protection Agency (EPA) require a minimum of 5 mL of sample volume and cannot detect all daughter products simultaneously. Here we report on the simultaneous detection of six chlorinated ethenes and ethene itself using a liquid sample volume of 1 mL by concentrating the compounds onto an 85-µm Carboxen polydimethyl silane (PDMS) solid phase microextraction (SPME) fiber in 5 minutes and subsequent chromatographic analysis in 14.5 minutes. Linear increases in signal response were obtained over 3 orders of magnitude (~0.05 to ~50 µM) for simultaneous analysis with R^2 values of $\geq$ 0.99. Method detection limits (1.3-6 µg/L) were at or below the EPA's maximum contaminant levels. Matrix spike studies with groundwater and mineral medium showed recovery rates between 79-108%. The method's utility was demonstrated in small-scale, sediment-column microcosms assessing the bioremediation potential of chlorinated ethene-contaminated groundwater. Owing to its low sample volume requirements, good sensitivity, and broad target analyte range, the method is suitable for routine compliance monitoring and particularly attractive for interpreting bench-scale feasibility studies commonly performed during the remedial design stage of groundwater cleanup projects.

Keywords: solid phase microextraction, reductive dechlorination, chlorinated ethenes, ethene, autosampler

Introduction

The chlorinated ethenes perchloroethene (PCE) and trichloroethene (TCE), once widely used in industry as metal degreasing agents and as solvents in dry cleaning, are classified as probable and known human carcinogens, respectively [1]. These compounds are widespread contaminants of groundwater, as are their various products of chemical, biological and physical transformation, including the three isomers of dichloroethene (*cis*-1,2-, *trans*-1,2-, and 1,1-DCE), and vinyl chloride (VC) a known human carcinogen [2]. Anaerobic reductive dechlorination of these chlorinated ethenes involves the stepwise replacement of each chlorine atom with a hydrogen atom to the end product ethene.

In order to carry out a mass balance to monitor the desirable anaerobic, reductive process of microbiological dechlorination of these compounds, it is critical to have a method capable of simultaneous detection of the parental chlorinated ethenes, their lower chlorinated intermediates, and the non-chlorinated end product, ethene. The U.S. Environmental Protection Agency (EPA) maximum contaminant levels (MCLs) for these compounds are all in the low µg/L range. In order to obtain sufficiently low method detection limits (MDLs) with headspace injection, the minimum liquid volume using U.S. EPA Method 5021A is 10 mL. U.S. EPA Method 624 involves the sample concentration approach called purge and trap, reducing the liquid volume requirement to 5 mL [3]. Whereas this volume is only a fraction of the volume of a standard U.S. EPA Volatile Organic Analysis (VOA) 40-mL vial, it still represents a challenge in

commonly performed remediation feasibility studies that require the repeated sampling and analysis of laboratory-scale reactors and microcosms [4-8]. This analytical challenge is common to the long established groundwater remediation industry and to the emerging field of bioenergy production using contaminated process water.

Various studies describe the use of solid phase microextraction as a means for concentrating analytes from sample headspace (HS SPME) followed by gas chromatography (GC) flanked by detection using either a flame ionization detector (FID), electron capture detector, or mass spectrometer for the analysis of PCE, TCE, DCE, or a combination of these [9-12] None of these methods, however, include analysis of vinyl chloride and ethene. In addition, most of these methods use liquid volumes of 5 mL or greater and require below room-temperatures for the extraction. A detailed review of HS SPME is in Zhang and Pawliszyn [13], and a comparison between HS SPME versus purge and trap is in Lara-Gonzalo et al. [14].

Song et al. [15], Wymore et al. [16], and Chung et al. [7] note the use of HS SPME GC-FID for analysis of TCE, *cis*-DCE, VC, and ethene from groundwater samples, but none describe the method in detail. Song et al. [15] state that the analyses were carried out with a 75-µm carboxen-PDMS fiber, but a full description of the method was not published. Wymore et al. [16] validated their method by sending various samples to off-site laboratories that used U.S. EPA methods and receiving blind samples from an independent vendor to analyze on-site; they too do not further describe their method. Chung et al. [7] provide the greatest detail on their method, e.g. fiber type, adsorption time, column, temperature profile, and note that the SPME was carried out manually. Their calibration approach, however, was conducted in 160-mL serum bottles with 100 mL of liquid, whereas the samples were analyzed in 2.5 mL vials with 2 mL of liquid. It may be possible to scale the analyses for different liquid and headspace volumes, but

this would impinge on achieving the desired low detection limits. Thus, the method requires further validation.

This study describes the first HS SPME GC-FID method able to simultaneously detect PCE, TCE, *cis*-, *trans*-, and 1,1-DCE, vinyl chloride, and ethene; the method was tested in synthetic and actual groundwater. Furthermore, this method is automated and uses a sample volume of only 1 mL, which is ideal for laboratory settings. It has recently been applied successfully by Ziv-El et al. [8] who studied a lab-scale membrane biofilm reactor (MBfR) whose application is reductive dechlorination of TCE in groundwater. In this study we also present results where this method was used in a feasibility study to assess bioremediation using a sediment column, flow-through microcosm.

Experimental

Sample preparation

1-mL liquid samples were analyzed in 2-mL crimp top vials with a magnetic cap and silicon/PTFE septa (MicroSOLV Eatontown, NJ). The samples were vortexed inverted for 1 minute to promote rapid mass transfer of the chemicals into the vial headspace.

Analysis

The samples were processed with an AOC-5000 autosampler as follows (Shimadzu, Columbia, MD). The vials were heated and vortexed in an agitator oven at 30˚C for 1 minute, followed by a 5-minute adsorption period by solid-phase microextraction, using an 85-µm Carboxen-PDMS fiber (Supelco, Bellefonte, PA), followed by 5 minutes of desorption into a gas

chromatograph equipped with a flame ionization detector (Shimadzu GC-2010, Columbia, MD). The compounds were separated with an Rt-QSPLOT column of 30 m length, 0.32 mm ID, and 10 µm film thickness (Restek, Bellefonte, PA). Helium was the carrier gas, flowing at a rate of 1.85 mL/min. To optimize the practical range of the method, a split ratio of 10 was selected which offered a good balance between low MDLs and a broad linear dynamic range for all analytes. The injection port was held at 240˚C and the temperature profile was 110˚C for 1 minute, a ramp of 50˚C/minute to 200˚C, and another ramp of 20˚C/minute to 220˚C and held for 3 minutes, for a total analysis time of 9.15 minutes. The FID detector temperature was 240°C and the composition of the flame was (He:H_2:zero-grade air) 35:49:490 mL/min.

Calibration curves, limits of detection, and linearity

The calibrations were carried out with neat PCE (Sigma Aldrich, St. Louis, MO), TCE (Sigma Aldrich, St. Louis, MO), *cis*-, *trans*-, and 1,1-DCE (Supelco, Bellefonte, PA), gaseous VC at 99.5% (Fluka, Milwaukee, WI), gaseous ethene at 99.5% (Matheson Tri-gas, Basking Ridge, NJ).

For PCE, TCE, *cis*-DCE, *trans*-1,2-DCE, and 1,1-DCE a stock solution, described in Table S1a of the Supplementary Information, was prepared in 245-mL serum bottles containing a Teflon-lined stirring bar and filled with deionized water, leaving no headspace, capped with a polytetrafluoroethylene (PTFE)-lined stopper and crimped. The solution was stirred at room temperature for four hours before use. A second stock solution was prepared by a two orders of magnitude dilution into the same serum bottle setup described above. Details of the VC and ethene stock concentrations are provided in Table S1b. The concentrations were prepared in

120-mL serum bottles holding a PTFE-lined stirring bar, capped with a PTFE-lined stopper, and crimped. The mixtures were stirred for 4 hours before use.

Varying volumes of the liquid stock solutions were added to the 2-mL sampling vials with a gas-tight syringe containing deionized water to provide a final volume of 1 mL; the final concentrations and preparation details are in Table S1c. The gaseous stock solutions were then added as listed in Table S1d. Before adding the stock solutions of liquid and gas, the volume equivalent to that being added was removed from the vial headspace so as to minimize chemical losses due to over-pressurization.

The method detection limits (MDLs) were determined as described previously [17]. Seven blank samples were analyzed and the signal mean and standard deviation were determined. The MDL was the lowest concentration analyzed for which the signal for seven samples was always larger than three standard deviations above the mean of the blanks.

The limits of linearity were determined as the concentration range for which the coefficient of determination (R^2) was greater than 0.99. A seven point calibration was then carried out in triplicate.

Recoveries of Arizona groundwater and anaerobic mineral medium

The recoveries of the compounds (n = 4) were tested in Arizona groundwater and in anaerobic mineral media [18], typical of that used for anaerobic reductive dechlorination feasibility studies, with spiked samples containing target analytes at concentrations between 9-36 µM. The spiked concentration values for each in µM and mg/L are in Table I.

Maximum holding time for abiotic samples

The maximum time that samples can be stored, was defined as the time for which the compound recoveries remained in the range of 90 to 110%. Triplicate samples were tested by spiking deionized water with target analytes to final concentrations of 10-30 µM and subsequent analysis after temporal storage in two conditions: upright in the autosampler at room temperature and inverted at 4˚C, to minimize losses through the cap.

Sediment column microcosm study

The utility of the HS-SPME-GC-FID method was assessed in a bench-scale feasibility study design typical of the groundwater remediation industry. A sediment column, flow-through microcosm was inoculated with the mixed-microbial consortium and fed with groundwater containing TCE at a concentration ~50 µg/L. The influent-feed cycle was 56 µL for 90 seconds followed by a 240 second pause, resulting in an effective feed-rate of 0.91 mL/hour. Influent and effluent samples were taken periodically with a gas-tight syringe by sampling 0.2 mL and diluting in 0.8 mL of deionized water, and analyzed as described above.

Results and Discussion

All seven compounds – PCE, TCE, *cis*-DCE, *trans*-DCE, 1,1-DCE, VC, and ethene – could be separated and analyzed simultaneously, as seen in the chromatogram in Figure 1. The adsorption and desorption times, injector and detector temperatures, and column temperature profile were optimized with extensive screening to provide maximum signal response and ensure no carry-over of analytes. The MDLs for all the compounds,Table II, were at or below the U.S. EPA maximum contaminant levels (MCL), except for VC where the MDL was slightly (0.5

µg/L) above the MCL. Page and Lacrois [19] demonstrated that lower detection limits can be achieved with a fiber coated with carbonxen/PDMS as opposed to PDMS alone. Furthermore, analyzing TCE in municipal sewage, Wejnerowska and Gaca [20] report a four times lower detection limit using an ECD detector (0.005 µg/L) compared to an FID detector (6 µg/L), and this may improve the detection limits for the method described in this study. However, such modifications are non-essential for feasibility studies [4-6] and would be beneficial mostly if the method is applied to environmental monitoring for compliance purposes. For remediation feasibility studies, replacement of the FID with a halogen-responsive ECD is counter-productive, as it makes impossible the simultaneous detection of the fully dechlorinated product ethene along with its chlorinated parental compounds. While mass spectrometric detection can add crucial information, FID is much more broadly available and less expensive to perform.

Using extensive screening we found that the linear range of this method extended across three orders of magnitude for all seven compounds when monitored jointly (Table II) and could be extended further for at least one order of magnitude when analytes were assayed individually. As a comparison, Poli et al. [11] reported a linear range of four orders of magnitude for PCE and TCE analysis in urine with an MS detector. Fabbi et al. [21] observed linearity across three orders of magnitude in concentration for olive oil samples. Wejnerowska and Gaca [20] achieved linearity across a single order of magnitude using an FID detector and twice that when using an ECD detector.

Analyte recoveries in Arizona groundwater and mineral medium, reported in Table I as the percent recovery compared to spiked deionized water, were minimally sensitive to the aqueous sample matrix assayed (Table I). The recoveries of PCE, TCE, and three DCE isomers were 99-108% for groundwater and 94-100% for anaerobic mineral medium; these recoveries

were similar to those reported by Wu et al. [22] who analyzed industrial wastewater samples. The recoveries for VC and ethene were lower: ~90% in groundwater and ~80% in the mineral medium, respectively. Other studies have analyzed samples from a diverse group of liquids including vegetable oil [19], olive oil [23], municipal sewage [20], urine [11], and rat blood [24]. No studies have reported on method development for these compounds in groundwater or mineral medium, and the studies referenced above did not assess compound recoveries.

We further studied the impact of sample storage for vials kept upright at room temperature in the autosampler and inverted at 4˚C (Figure S1), and calculated the corresponding maximum holding times, Table III. Samples were stable for up to 1 hour in the autosampler and at least 47 hours at 4˚C. Other studies using autosamplers [7, 8, 23] do not report holding times. Use of a chilled autosampler could potentially extend the here reported sample holding times but this equipment is not widely available in typical lab settings.

Finally, we applied the method to the analysis of a lab-scale sediment flow-through column experiment, operated in triplicate, to examine the time course of biological reductive dechlorination of TCE to ethene. In Figure 2B are the results for a representative column. One measurement was taken for each sediment column at each time point and trends were similar across the columns. The method was successful in tracking reduction of TCE to *cis*-DCE, VC, and ethene over an 80-day span with starting concentrations in the low µg/L range. The sum of the products at any point (i.e., the mass balance) fluctuated by at most 40%. These fluctuations are indicative of sorption phenomena taking place in the experimental setup.

Based on the results obtained, this method appears best suited for laboratory feasibility studies, such as small-scale sediment columns or water treatment technologoies [7, 8], where sample volume is limited and samples taken are processed immediately to obtain an optimal

mass balance. The method also may serve for compliance monitoring when sample volumes are limited and prevent the use of conventional EPA standard methods.

Conclusions

A rapid, simple, and replicable method was developed for the simultaneous detection of chlorinated ethenes and ethene quantification with a linear range across at least three orders of magnitude and detection at or below the EPA MCLs. This method is advantageous for analysis of biological reductive dechlorination where detection of ethene is essential to detect and monitor the desired outcome. Additionally, the small (1 mL) required sample volume and its ability to simultaneously track chloroethene conversion and ethene evolution make it particularly well suited for bench-scale experiments where the reactor volume is often small, as in the sediment column, flow-through microcosm presented here and in Ziv-El et al. [8].

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Table I. Recovery rates obtained for different matrices (*n*=4).

Table II. Compound retention times, limits of linearity for the simultaneous detection of all analytes each containing seven calibration points measured in triplicates, and method detection limits determined with seven replicates.

Table III. Maximum storage, defined as recovery between 90-110%, for triplicate samples in deionized water with a concentration in the high range of the calibration curve. Samples were held upright in the autosampler or inverted at 4˚C.

Figure 1. Example chromatogram in DI water for (A) upper linearity values (Table 1) and (B) near the MDLs (Table 1), and in (C) spiked groundwater near the upper linearity values.

Figure 2. Application of HS-SPME-GC-FID method. A schematic of the multi-channel lab-scale sediment flow-through columns used in this study is in panel (A). In panel (B) are the effluent reductive dechlorination products from a column inoculated with the mixed-microbial consortium DehaloR^2 [8] and fed synthetic groundwater spiked with TCE. The columns were operated in triplicate and these results are from a representative column.