Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Chromatography A, 1011 (2003) 125–134
JOURNAL OF CHROMATOGRAPHY A
www.elsevier.com/locate/chroma

Application of solid-phase microextraction and gas chromatography–mass spectrometry to the determination of volatile organic compounds in end-exhaled breath samples ☆

C. Prado*, P. Marín, J.F. Periago
Instituto de Seguridad y Salud Laboral, Apartado 35, E-30120 El Palmar, Región de Murcia, Spain

Received 7 March 2003; received in revised form 27 May 2003; accepted 17 June 2003

Abstract

Analysis of exhaled air is of particular interest as an indicator of health as well as a tool for the diagnosis of diseases. It is also a very attractive procedure for the biological control of the exposition to hazardous solvents. This kind of analysis presents numerous advantages over other methods, the most important being that it is not an invasive procedure and, therefore, it is well accepted and can be applied to a wide range of compounds. Furthermore, the analysis is simplified since the matrix is less complex that in the case of blood or urine. In spite of these obvious advantages and the good results obtained, analysis of exhaled air is not in daily use, probably due to the fact that there are no normalized systems of sampling, thus making the interpretation of the results difficult. In this paper, a method for the determination of tetrachloroethylene in exhaled air using solid-phase microextraction is presented. This method, which can be applied to other volatile organic compounds, was developed with special emphasis of end-exhaled breath sampling. The sample is collected in a glass tube whose ends are closed once the exhalation is finished. The tube has an orifice sealed with a septum through which the fiber is inserted. Then, the fiber is desorbed in the injector of a gas chromatograph and the analysis is accomplished using mass spectrometry for the identification and quantification of the components. The proposed system avoids the need of complex sampling equipment and allows analysis of the alveolar fraction. Additionally, the system is economical and easy to handle, thus facilitating the development of normalized methods and its routine use in field studies.

Keywords: End-exhaled air analysis; Biological monitoring; Volatile organic compounds

1. Introduction

In recent years there has been increased interest in the determination of the constituents of exhaled breath as disease markers in medicine and as exposure markers in occupational toxicology [1–4].

Various kinds of breath samples could be used, including mixed expired air and end expired air. Mixed expired air can be used qualitatively, in order to know the existence of a contaminant exposition. Additionally, the increased concentration of one or more components in exhaled air can be directly related to a disease.

The measurement of volatile compounds in breath

☆Presented at the 2nd Meeting of the Spanish Society of Chromatography and Related Techniques, Barcelona, 26–29 November 2002.

*Corresponding author. Fax: +34-68-365-501.
E-mail address: celiaa.prado@carm.es (C. Prado).

0021-9673/03/$ – see front matter
doi:10.1016/S0021-9673(03)01103-8

was considered a good procedure to estimate the internal dose of a toxicant by biological monitoring, since both the theoretical bases and the relevant toxicokinetic data have been already established. However, since breath is not a homogeneous sample, the portion of breath sample which reflects the blood solvent concentration must be identified and collected when breath is going to be used for biological monitoring. End-exhaled air sampling can be obtained by a non-invasive way and allows for valid estimates of partial solvent pressure in arterial blood, given the existing equilibrium between alveolar gas and arterial blood [5,6]. Nevertheless, the use of breath analysis has been decreasing since it was incorporated into the biological exposure index (BEI) of the American Conference of Governmental Industrial Hygienists (ACGIH). Actually it is not routinely used for occupational exposure monitoring. In fact, of 42 biological exposure indices for 22 chemical substances proposed by the ACGIH in 1990, 11 of them were for exhaled air [7]. Actually, 58 BEIs have been proposed for 38 substances, but only two of them were adopted for end-exhaled air [8]. As it has been pointed out, this limited use of breath analysis was attributed to a lack of both basis for data interpretation and methodology [9].

An extensive review of the procedures, applications and limitations of breath sampling was recently presented in the literature [10]. It was pointed out that the development of methods involving collection devices with adsorbent tubes could be considered as an important advance to promote the use of breath analysis in field use. In this way, a system was designed to collect and trap the end-expired air, which consisted of a modified Haldane–Priestley tube. The design allows to concentrate aliquots of exhaled air from one or more exhalations to an adsorbent tube packed with active carbon for chemical desorption [11], or with an adequate sorbent for thermal desorption [12,13]. The system was suitable for monitoring *n*-hexane, toluene, styrene and isoflurane in breath. The good correlation obtained between personal exposure and the level of the biomarker allows biological exposure limits to be calculated for environmental limits [14–16].

Solid phase microextraction (SPME), in which a fused-silica fiber coated with a stationary phase is used, has been applied to the analysis of environmental contaminant compounds in water, soil matrixes and, more recently, for the analysis of drug compounds in biological matrixes [17–20]. SPME has also been applied to the determination of some constituents, as acetone, ethanol and isoprene, in human breath [21–23].

In this work, the applicability of SPME to accomplish the biological monitoring of organic compounds in end-exhaled air has been studied. We have focused the study on tetrachloroethylene as a model compound—a solvent used in dry-cleaning shops and in metal cleaning operations. The capacity of humans to metabolize tetrachloroethylene is limited. It is known that only 1–3% of the amount of tetrachloroethylene absorbed is metabolized to trichloroacetic acid by humans, the remaining being excreted unchanged in exhaled air. This fact turns tetrachloroethylene into a good model of respiratory elimination. This compound is also interesting because is one of the compounds that have a biological limit value.

2. Experimental

2.1. Studies in controlled atmosphere

The determination of the calibration curve includes the preparation of standard gas mixtures of organic vapour in air. This requires the generation of controlled atmospheres having different concentrations of the pollutant. The system used to generate the standard atmosphere is illustrated in Fig. 1 was previously described [24]. It consists in a dynamic method based on a syringe pump (Harvard Apparatus, Holliston, MA, USA) that provides a constant flow-rate of tetrachloroethylene (Merck, Darmstadt, Germany) by the movement of a syringe plunger. The content of the 50-μl syringe (Hamilton, Reno, NV, USA) is injected through a septum to the air line and evaporated with clean compressed air.

The atmospheres were checked by active samplers consisting of activated charcoal tubes connected to the atmosphere by means of a sampling pump. In all cases the concentration measured by the activated charcoal tubes agreed to the theoretical concentration calculated from airflow and syringe speed within 10% [25].

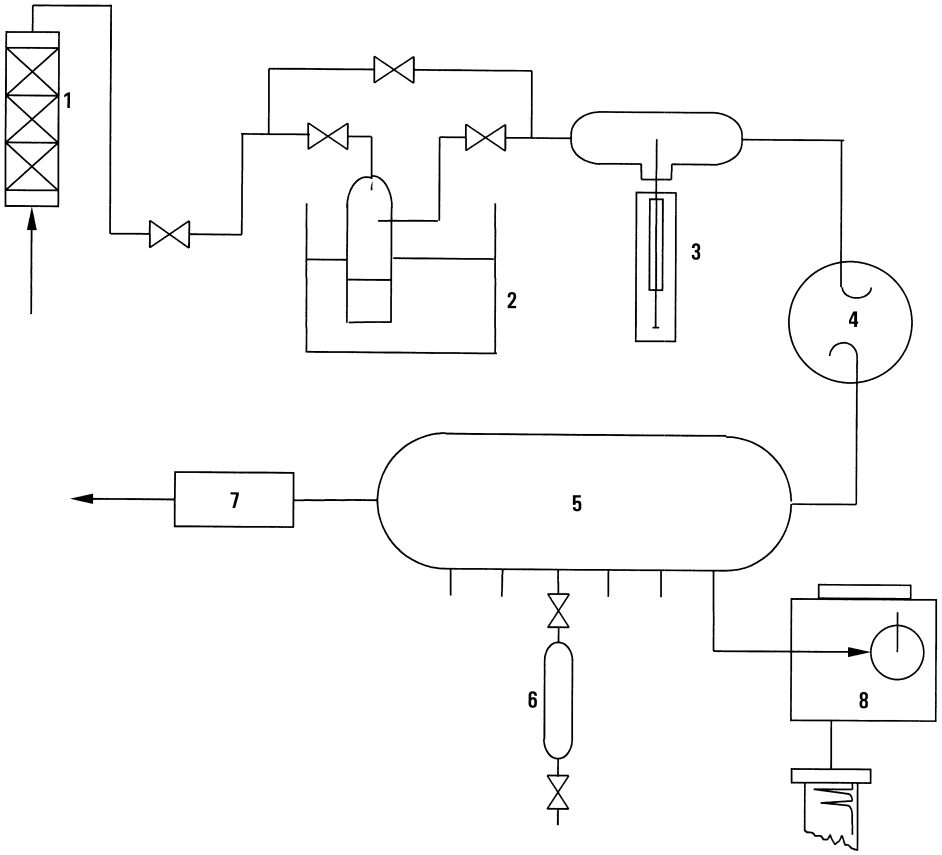

Fig. 1. Scheme of the system used to generate the controlled atmosphere. 1 = Filter; 2 = humidifier; 3 = automatic injector; 4 = mixing chamber; 5 = sampling chamber; 6 = exhaled air sampling device; 7 = humidity sensor; 8 = gas chromatograph.

The humidity of the atmosphere could be adjusted by introducing water bubbles into the incoming airflow. Relative humidity of 98–100% was used for all experiments, since it is the expected value in the exhaled breath.

The device to collect breath samples consists of a 125-ml glass bulb fitted with valves on both ends. One of the ends allows a disposable mouthpiece to be used. This glass tube was coupled to the atmosphere through a valve. This scheme allows to simulate the concentrations of tetrachloroethylene in human breath.

The atmosphere was stable through all experiments and the coefficient of variation, determined by on-line chromatography, was lower than 4%. The experiments were carried out at different concentration levels of tetrachloroethylene, ranging between 2 and 35 mg/m^3. All experiments were done at ambient temperature.

Preparation of standard atmospheres and sampling were realized in a room separated from the analytical laboratory. For every experiment, the glass tube was connected to the atmosphere and the contaminated air stream was passed through the tube for 1 min. Then the tube was closed and analyzed. Then, clean air was passed through the tube in order to be conditioned for a new sampling. In a control assay, a blank sample was collected and analysed in the same way.

2.2. Collection of human exhaled breath

Samples of exhaled air were obtained by having the subject taken two or three deep breaths, inhaling

and holding their breath for 10–15 s, then exhaling into the glass tube with the valves in the open position. At the end of an exhalation the two valves are closed, trapping an aliquot of end-expired (alveolar) breath in the tube. Exhaled breath samples were collected in an area separated from the site of exposure.

2.3. Solid phase microextraction method and GC–MS analysis

SPME devices and 100-μm bonded polydimethylsiloxane fiber assemblies were purchased from Supelco (Bellefonte, PA, USA). The fiber was conditioned in a GC injection port at 250 °C for 1 h to remove fiber contaminants.

The SPME fiber was introduced into the glass tube through a silicone septum and exposed for 1 min. Care was taken to ensure that always the same length of the septum-piercing needle passed through the septum. After extraction, the fiber was withdrawn into the needle, pulled out from the tube and injected into the GC.

A HP 6890Plus gas chromatograph (Hewlett-Packard, Palo Alto, CA, USA) coupled to a HP5973 mass detector was used for all experiments. The gas chromatograph was equipped with split–splitless injector and a crosslinked methyl silicone capillary column measuring 50 m×0.2 mm I.D. with a phase thickness of 1 μm (HP-1MS, Hewlett-Packard).

The SPME fiber was exposed into the injection port, that uses a Merlin (Merlin, Half Moon Bay, CA, USA) microseal instead a standard septum, for 2 min at 200 °C. This treatment provided a complete desorption of tetrachloroethylene, that was checked with a second desorption. The column temperature was hold at 35 °C for 2 min and increased at 15 °C/min up to 160 °C. The flow-rate of the helium carrier gas was settled constant to 1.2 ml/min. Injection was made in the splitless mode and the splitter was opened after 2 min. A silanized narrow-bore injector liner (0.75 mm I.D., Supelco) was installed.

The mass spectrometer, with the ion source maintained at 230 °C, was used in the full scan mode. The scan range was from m/z 20 to m/z 250 at 5.98 scan/s. The tetrachloroethylene retention time was 9.5 min. Standard autotunes with perfluorotributylamine (PFTBA) were carried out on a daily basis as well as a blank desorption of the fiber to ensure that there was not any peaks eluting at the same time as tetrachloroethylene.

2.4. Comparison to other method of end-exhaled air sampling, sorbent concentration and thermal desorption analysis

Samples of controlled atmospheres were also taken with a previously developed device as reference method for sampling end-exhaled breath. The system has already been described [11,12,16] and it consists of an aluminium tube modified to concentrate aliquots of end-exhaled (alveolar) air from one or more exhalations on to a thermal desorption tube (Perkin-Elmer, Beaconsfield, UK). A servomotor driven gas syringe (50 ml) draws a sample from the aluminium tube and then dispenses it on to an adsorbent tube packed with Tenax TA (SKC, PA, USA). The sampler was coupled to the atmosphere system through a valve. This scheme allows to simulate the concentrations of this compound in human breath. The experiments were carried out at two different concentration levels of tetrachloroethylene, 3.9 and 9.7 mg/m^3, and 98–100% of relative humidity. The total volume passed through the adsorbent tube was 500 ml.

The adsorbent tubes were desorbed with a Perkin-Elmer Model ATD 50 thermal desorption system, directly connected to a Perkin-Elmer Model 8700 gas chromatograph by a heated transfer line. The operating conditions are summarized in Table 1.

3. Results and discussion

Table 2 shows the results of the repeatability study on the determination of tetrachloroethylene concentration in the simulated exhaled air. Replicate measurements were performed by extracting the tetrachloroethylene in six glass bulbs obtained from the same controlled atmosphere. Three different concentrations were checked. The repeatability was evaluated by calculating the mean, standard deviation, and relative standard deviation (RSD) of the observed values. RSD values for within-day measurement were satisfactory and lower than 7%. It must be stressed that these RSD values include not

Table 1
Experimental conditions for thermal desorption and gas chromatography (FID = flame ionization detection)

Parameters	
Thermal desorption parameters	
Carrier gas	N_2 (68.95 kPa, 10 p.s.i)
Desorption	7 min
Oven temperature	230 °C
Cold trap	
Lower temperature	−30 °C
Upper temperature	240 °C
Heated transfer line temperature	120 °C
Gas chromatographic parameters	
Column	FFAP (25 m × 0.2 mm × 0.3 μm)
Oven temperature	Isothermal, 90 °C
Detector temperature	280 °C
Detection	FID

only the precision of the sampling and analytical method but also the homogeneity of the atmosphere generated.

Table 3 shows the results from inter-day precision. Each value was obtained from a different controlled atmosphere that was generated in 10 different days, so the concentrations were only approximately the same. RSD value, that also includes the inter-day precision of the atmosphere generation, are similar to those found for methods involving both SPME [21,22,26] and sorbent tubes to trap analytes in exhaled breath [11,12], thus reflecting that precision for such samples is reliable enough to provide representative values.

There are no standard reference materials available for human breath. However, the test atmospheres generated in this work meets the requirements for a calibration gas mixture [25] and the concentration of each experiment can be considered as ''true concentration''. The calibration curve was calculated with the results obtained from the analysis of the simulated end-exhaled breath samples taken from the controlled atmosphere, as standard calibration, over the concentration range from 2 to 33.5 mg/m^3. The area of the peaks in the total ion chromatogram has

Table 2
Repeatability (as RSD) of the determination of tetrachloroethylene at different concentrations levels

Tetrachloroethylene concentration (mg/m^3)	Peak area (TIC)	RSD (%)
2.2	636 611	6.2
	619 817	
	578 476	
	622 122	
	638 811	
	700 116	
3.9	1 168 335	7.0
	1 070 886	
	1 041 117	
	1 146 982	
	1 266 787	
	1 168 335	
8.3	2 816 327	4.9
	2 827 183	
	3 127 662	
	2 743 660	
	2 941 583	
	2 793 675	

RSD, relative standard deviation (%); TIC: total ion chromatogram.

Table 3
Inter-day precision of the determination of tetrachloroethylene at different days

Tetrachloroethylene concentration (mg/m^3)	Peak area (TIC)	RSD (%)
3.9	1 168 335	12.2
4.0	1 364 800	
3.9	1 016 095	
4.1	1 042 721	
3.9	938 258	
3.9	1 187 659	
3.9	1 019 908	
3.8	1 176 931	

RSD, relative standard deviation (%); TIC, total ion chromatogram.

been represented versus the tetrachloroethylene atmosphere concentrations. The concentration range studied was the expected for occupationally exposed subjects. Every level has been measured at a different day, and represents the arithmetic mean of at least three replicate determinations, i.e. three different samples of the same controlled atmosphere. The curve shows good response linearity over the studied range. Linear correlation was as follows: $y = -965\,837 + 469\,622x$ (x = concentration (mg/m^3) of tetrachloroethylene, y = peak area, $r = 0.995$). The method detection limit was determined by analyzing test samples and multiplying the standard deviation of the mean by the appropriate Student's t-value for 99% confidence level with the appropriate degrees of freedom. The calculated value was 0.3 mg/m^3 for eight replicates.

Under the sampling and analytical conditions described here, tetrachloroethylene in exhaled air can be quantified at concentrations about 40 times lower than the biological exposure index proposed, for end-exhaled breath, by the ACGIH for this compound [8]. So, the developed method permits both to monitor subjects who are exposed to low concentrations of tetrachloroethylene and to detect the accumulation of this chemical in fatty tissues by measuring the compound in exhaled breath some time after exposure has occurred [27]. Single ion monitoring (SIM) can also be used to quantify the compound, thus notably improving the analytical results. The SIM acquisition can give, for example, a 20-fold increase in sensitivity [28].

It must be pointed out that the use of mass spectrometry can be necessary in order to identify exhaled breath constituents for medical diagnostics or in occupational exposure to complex mixtures of compounds that can interact toxicologically. Nevertheless, the procedure can be simplified by using a FID detector in routine monitoring of occupational exposure to a compound. As an example, Fig. 2 shows a chromatogram obtained for tetrachloroethylene analysis, corresponding to the lowest concentration studied 1.9 mg/m^3, using flame ionization detection (FID).

For purposes of comparison, samples of the same atmospheres were analyzed for tetrachloroethylene

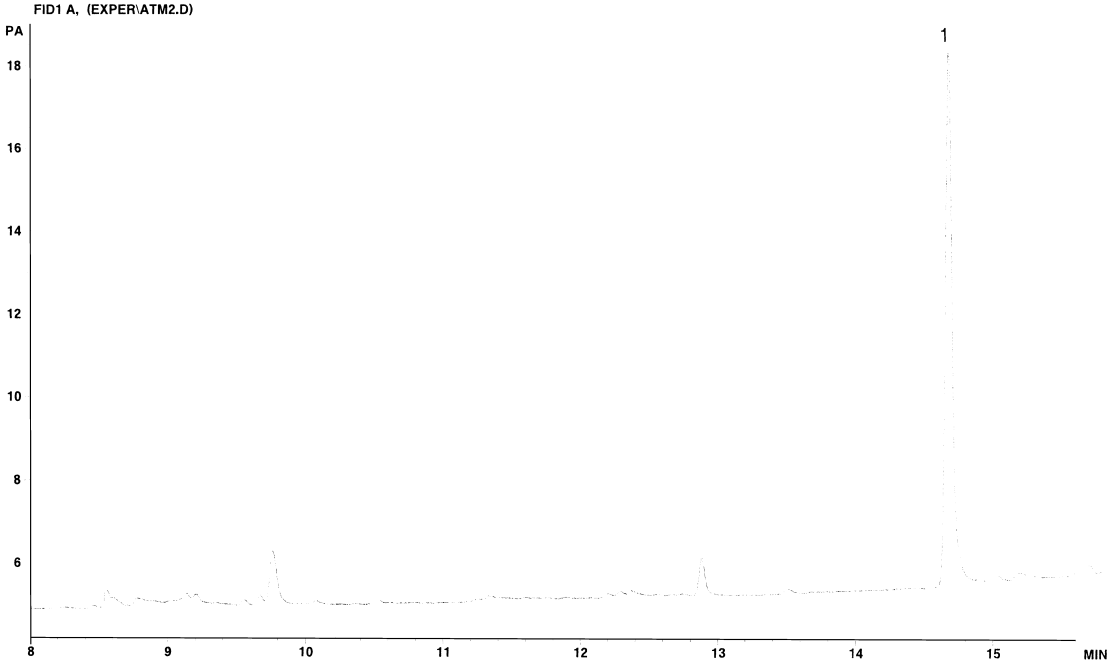

Fig. 2. Gas chromatogram obtained from the analysis of a sample bulb by SPME–GC–FID (1.9 mg/m^3 controlled atmosphere of tetrachloroethylene). (1) Tetrachloroethylene.

Table 4
Results of method comparison and overall uncertainty for three concentration level

Concentration level (mg/m^3)	Measured concentration (mg/m^3)				Overall uncertainty[a] (%)	
	SPME		Tenax tube		SPME	Tenax tube
1.93	2.33	2.04	1.97	2.22		
	2.18	2.04	2.23	2.12	22.5	20.7
	2.03	2.14	2.12	2.00		
3.85	3.68	4.56	4.05	3.76		
	3.87	3.59	4.11	3.88	21.8	10.6
	3.64	3.47	3.64	3.98		
9.69	10.51	9.95	9.39	9.71		
	10.58	8.38	9.33	9.35	16.7	5.4
	10.12	9.55	9.47	9.46		

[a] $\frac{|\bar{x} - x_{true}| + 2s}{x_{true}} \cdot 100$, where $\bar{x}$ is the mean value of the repeated measurements, x_{true} is the true atmosphere concentration, s is the standard deviation of measurements.

using both SPME and adsorbent concentration method, previously described in Section 2.4. In field studies, this last method has been found suitable for monitoring solvents in end-exhaled breath [11,12,14–16]. Table 4 shows the results obtained by the two methods of sampling and analysis of exhaled breath air at three different tetrachloroethylene concentration level. On these results an analysis of variance has been carried out, for each concentration, in order to determine whether there are significant differences with respect to the two sampling and analyzing methods. Table 5 shows that no differences are found to be significant at 5% level at any of the tested tetrachloroethylene concentrations between the results obtained by means the developed procedure and the obtained by the comparison method.

Results of overall uncertainty of the measuring procedures are also showed in Table 4. Overall uncertainty is expressed, on a relative basis, by a combination of bias and precision [30]. Results for the proposed method and the comparison method are similar. The overall uncertainty was also similar to the NIOSH method for tetrachloroethylene in exhaled breath by a portable GC [31].

The sampling and SPME method, as described

Table 5
Analysis of experimental data: analysis of variance

Source of variation	Sum of squares	Degrees of freedom	Mean square	F-ratio	Significance level
$C = 1.93$ mg/m^3					
Between groups	8.33×10^{-4}	1	8.33×10^{-4}	0.07	0.803^{NS}
Within groups	0.127133	10	0.0127133		
Total	0.127967	11			
$C = 3.85$ mg/m^3					
Between groups	0.0310083	1	0.0310083	0.33	0.5776^{NS}
Within groups	0.936017	10	0.0936017		
Total	0.967025	11			
$C = 9.69$ mg/m^3					
Between groups	0.472033	1	0.472033	1.39	0.2659^{NS}
Within groups	3.39837	10	0.339837		
Total	3.8704	11			

NS, not significant at 5% level.

here, was applied to tetrachloroethylene determination from exhaled breath samples of an occupationally exposed subject in a cleaning shop. Unlike other alveolar breath collection devices, the worker does not need special training to provide end-expired air samples [4,10,29] and even the sample can be collected by the worker himself [32]. Fig. 3 illustrates the chromatograms obtained with samples collected before and after a typical exposure on the third day of the week. The time required to perform the complete sampling procedure and analysis described is less than 11 min, which is lower than that required using S_2C or thermal desorption methods.

Table 6 shows the resulting alveolar concentrations. It is remarkable the presence of tetrachloroethylene in the sample of exhaled air taken 16 h after exposure. Tetrachloroethylene tends to accumulate in fatty tissues. Hence, in repeated daily exposure there is a progressive accumulation of the solvent in the organism. This fact could explain the relatively high level of this sample.

Table 6 also shows the results obtained after three consecutive extractions from the same sample. The results show a good concordance. This feature is of a particular interest as it offers the advantage of repeated analysis of a sample, that is not shared by

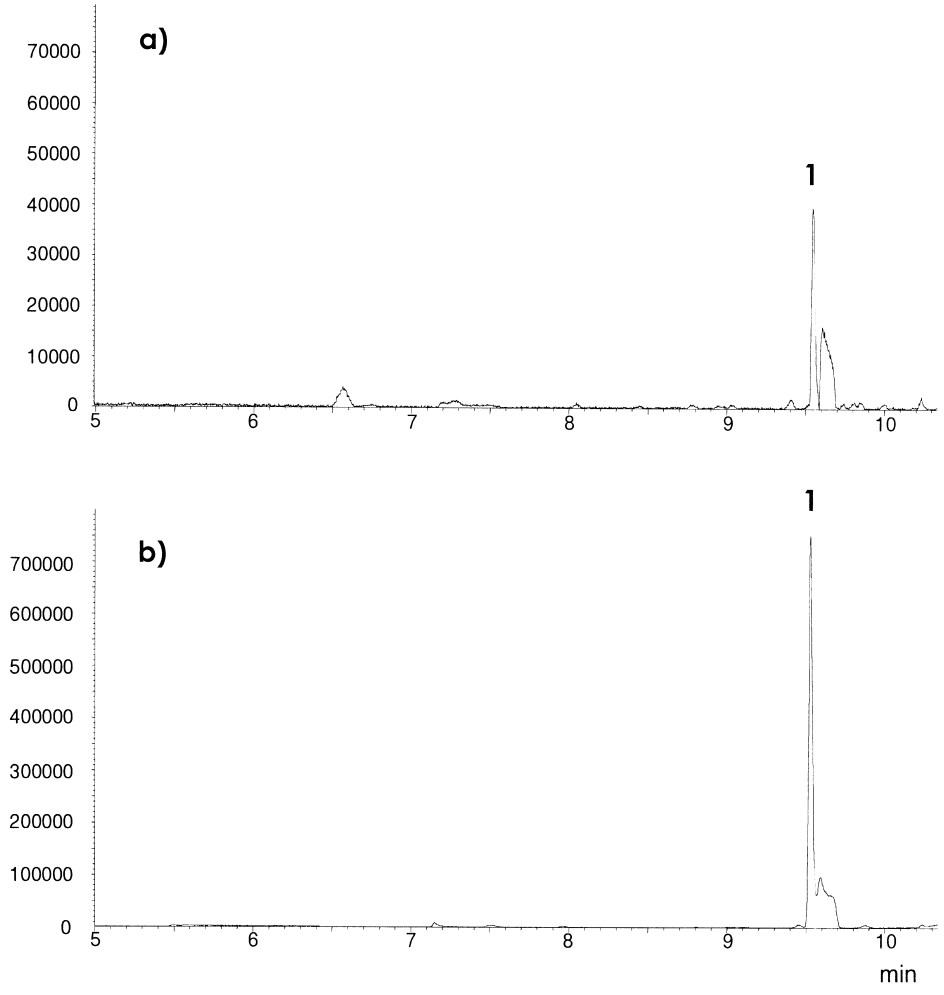

Fig. 3. Total ion chromatogram obtained from the analysis of exhaled breath of a person exposed to tetrachloroethylene. (a) Collected 16 h after exposure, (b) after exposure. (1) Tetrachloroethylene.

Table 6
Results of tetrachloroethylene concentration in end-exhaled air from an occupationally exposed subject

	Concentration (mg/m^3)
Tetrachloroethylene in end-exhaled air before exposure	3.4
Tetrachloroethylene in end-exhaled air after exposure	25.8
2nd Extraction	26.0
3rd Extraction	27.1

the thermal desorption method of analysis, that uses a sorbent to trap contaminants, because in this case all the sample is analyzed. Additionally, this feature would allow to detect and quantify other compounds of interest using different fiber coating depending on the polarity of the analytes to be extracted. Future research needs to concentrate on this topic.

4. Conclusions

SPME can be very useful for the biological monitoring of organic compounds in exhaled breath of occupationally exposed people. The method proposed in this study combines the sampling of alveolar breath using a glass tube with SPME, thus, allowing the quantification of volatile organic compounds in exhaled breath.

The extraction of tetrachloroethylene in exhaled breath samples shows good repeatability and the calibration curve also shows good linearity. The analytical procedure described here is simple, fast and highly reproducible. The proposed method could also be applied to other volatile organic compounds. Obviously, a calibration curve must be obtained for every compound to be checked.

The advantage the method offers when compared to others aimed to the same purpose, including the method based on sorbent concentration and thermal desorption, is to simplify the capture procedure and analysis. The proposed method avoids the need of complex sampling equipment for the alveolar breath fraction, and of additional instrumentation for sample desorption. Thus, the proposed method provides a practical tool for biological control of occupational exposure for industrial hygiene studies.

References

[1] M. Phillips, J. Herrera, S. Krishnan, M. Zain, J. Greeberg, R.N. Cataneo, J. Chromatogr. B. 729 (1999) 75.
[2] J.K. Schubert, K.H. Spittler, G. Braun, K. Geiger, J. Guttmann, J. Appl. Physiol. 90 (2001) 486.
[3] W.H. Cheng, W.J. Lee, J. Lab. Clin. Med. 133 (1999) 218.
[4] A.B. Lindstrom, J.D. Pleil, Biomarkers 7 (2002) 189.
[5] G.R. Kelman, Br. J. Ind. Med. 39 (1982) 259.
[6] H.K. Wilson, Scan. J. Environ. Health 12 (1986) 174.
[7] American Conference of Governmental Industrial Hygienists, Threshold limit values (TLVs) for chemical substances and physical agents and biological exposure indices (BEIs), ACGIH, Cincinati, OH, 1990.
[8] American Conference of Governmental Industrial Hygienists, Threshold limit values (TLVs) for chemical substances and physical agents and biological exposure indices (BEIs), ACGIH, Cincinati, OH, 2000.
[9] P.O. Droz, M.P. Guillemin, J. Occup. Med. 28 (1986) 593.
[10] H.K. Wilson, A.C. Monster, Occup. Environ. Med. 56 (1999) 753.
[11] J.F. Periago, A. Luna, A. Morente, A. Zambudio, J. Appl. Toxicol. 12 (1992) 91.
[12] J.F. Periago, C. Prado, I. Ibarra, J. Tortosa, J. Chromatogr. A 657 (1993) 147.
[13] C. Prado, J.F. Periago, A. Sepúlveda-Escribano, J. Chromatogr. A 719 (1996) 87.
[14] J.F. Periago, A. Cardona, D. Marhuenda, J. Roel, M. Villanueva, J. Martí, A. Luna, Int. Arch. Occup. Environ. Health 65 (1993) 275.
[15] J.F. Periago, A. Morente, M. Villanueva, A. Luna, J. Appl. Toxicol. 14 (1994) 63.
[16] C. Prado, J.A. Tortosa, I. Ibarra, A. Luna, J.F. Periago, J. Appl. Toxicol. 17 (1997) 179.
[17] Z. Zhang, J. Pawliszyn, Anal. Chem. 65 (1993) 1843.
[18] W.M. Coleman III, J. Chromatogr. Sci. 35 (1997) 245.
[19] H.L. Lord, J. Pawliszyn, Anal. Chem. 69 (1997) 3899.
[20] X.P. Lee, T. Kumazawa, K. Sato, J. Chromatogr. Sci. 35 (1997) 302.
[21] C. Grote, J. Pawliszyn, Anal. Chem. 69 (1997) 587.
[22] R. Hyŝpler, S. Crhová, J. Gasparič, Z. Zadák, M. Čížková, V. Balasová, J. Chromatogr. B 739 (2000) 183.
[23] M. Guidotti, B. Onorati, E. Lucarelli, G. Blasi, G. Ravaioli, Am. Clin. Lab. 20 (2001) 23.
[24] C. Prado, I. Ibarra, J.F. Periago, J. Chromatogr. A 778 (1997) 255.
[25] European Commission, Adopted European Standard. Workplace atmospheres: Diffusive samplers for the determination of gases and vapours. Requirements and test methods, EN838, EC, Brussels, 1995.
[26] X.P. Lee, T. Kumazawa, K. Sato, H. Seno, A. Ishii, O. Suzuki, Chromatrographia 47 (1998) 593.

[27] P.O. Droz, M.M. Wu, W.G. Cumberland, M. Berode, Br. J. Ind. Med. 46 (1989) 447.
[28] F.L. Cardinali, D.L. Ashley, J.V. Wooten, J.M. McCraw, S.W. Lemire, J. Chromatogr. Sci. 38 (2000) 49.
[29] R.A. Glaser, J.E. Arnold, S.A. Shulman, Am. Ind. Hyg. Assoc. J. 51 (1990) 139.
[30] European Commission, Adopted European Standard. Workplace atmospheres: General requirements for the performance of procedures for the measurement of chemical agents, EN482, EC, Brussels, 1994.
[31] National Institute of Occupational Safety and Health, Manual of Analytical Methods (NMAM), 4th ed, NIOSH, Washington, DC, 1998.
[32] P. Egeghi, L. Nylander-French, K.K. Gwin, I. Hertz-Picciotto, S.M. Rappaport, Ann. Occup. Hyg. 46 (2002) 489.