

A novel device for capturing breath samples for solvent analysis

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Abstract

We have developed a novel breath sampling device suitable for capturing a portion of end-tidal air. This breath sample is then transferred onto a Perkin Elmer automated thermal desorption (ATD) sampling tube which is subsequently analysed by ATD-gas chromatography-mass spectrometry (GCMS). The breath sampler has been evaluated in the laboratory, in brief field trials and in human volunteer studies. The method is sensitive with a typical detection limit of 1 nmol/l and reproducible with an overall coefficient of variation between 5% and 15% for collection and analysis of breath samples from volunteers. The field trials used the sampler to assess exposure to solvents in several industries including the shoe manufacturing industry, the inks and coatings industry and at dry cleaning establishments. The sampler was found easy to use and reliable. Solvents detected include ethyl acetate (6.4–25.5 nmol/l), propan-2-ol (3.4–39.3 nmol/l), 2-butanone (0–6.6 nmol/l) and tetrachloroethene (0–557 nmol/l). The breath sampler was also used to monitor the elimination of solvents in breath from human volunteers after exposure chamber studies. More than 500 breath samples have been analysed from 24 volunteers in exposures to 10 different solvents (toluene, trimethyl benzene, tetrachloroethene, tetrahydrofuran, acetone, propan-2-ol, xylene, 2-butanone, 1-methoxy-2-propanol and *n*-hexane). The breath sampler allowed the rapid and non-invasive collection of data on elimination of solvents. Published by Elsevier Science B.V.

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1. Introduction

Volatile organic chemicals are used extensively in industry and may be absorbed through the skin as well as by inhalation. In these instances, simply

monitoring air levels alone may not be sufficient to assess exposure and biological monitoring can be an important tool in contributing to the assessment of occupational exposure.

For some solvents, biological monitoring methods involve the analysis of solvent in blood. However, the collection of blood samples requires skilled medical staff and is not always welcomed by the workers. The analysis of solvents in breath

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has been proposed as a non-invasive alternative to blood sampling. It is based on the principle that there is an almost instantaneous equilibrium established between solvents in the pulmonary blood supply and in the air of the lung and therefore, analysis of solvents in breath should provide a good indicator of the levels of solvents in blood.

The use of breath analysis for routine biological monitoring has been slow to develop, probably because the techniques available for breath collection and analysis are found to be unsuitable for occupational exposure assessment. A variety of different techniques and devices have previously been used for breath sampling in industry, including glass pipettes, aluminium tubes, syringes, gas sampling bags and others (Wilson, 1986). However, many of these breath sampling processes are impracticable for widespread use, either because of the size of breath samples collected, such as with the large gas sampling bags, the fragility of the sampling device, for example glass syringes, or the size of equipment. Of the breath analysis techniques available, very few have been employed in the area of occupational exposure. Droz et al. (1988) used gas sampling bags and colorimetric detector tubes to investigate levels of tetrachloroethene in two dry cleaning shops and one aluminium can manufacturing plant. Campbell et al. (1985) and Drummond et al. (1988) used a portable mass spectrometer fitted with a Haldane Priestley sampling tube to measure benzene in the breath of workers in the coke oven industry and carbon disulphide in the textile industry. Peragio et al. (1994) used a portable exhaled air sampler consisting of a 1-m aluminium Haldane–Priestley tube connected to a motorised syringe which they used to measure concentrations of *n*-hexane and toluene in the shoe manufacturing industry. To further promote the use of breath analysis, a method of sampling is required which collects the end-tidal air reproducibly, minimises solvent losses, does not contaminate the sample, is inexpensive and easy to use. This paper describes the validation of a novel breath sampler for collecting alveolar breath for solvent analysis.

2. Methods and materials

2.1. Sampler design and principle of operation

The design and operation of the breath sampler is based on a variation of the Haldane–Priestley tube principle whereby the final portion of breath is held within a tube, then sampled. Fig. 1 shows a photograph of the breath sampler and Fig. 2 shows a cross sectional diagram through the breath sampler. The breath sampler consists of an aluminium outer casing narrowed at one end to form a mouth piece and at the other end to form an adapter which accepts Perkin Elmer automated thermal desorption tubes. Inside the sampler casing is a collapsible sampling bag which is attached to a moveable brass ring which is guided down the length of the casing by two open channels. At either end are two non-return valves one of which is also attached to the brass ring. The volunteer or worker breathes through the sampler until the final portion of breath has been exhaled and is trapped within the sampler. The volume of the captured sample is 85 ml. An absorption tube is then attached to the end of the sampler and by moving the brass ring and the non-return valve down the casing, the captured breath is forced out of the sampling bag and is transferred on to the absorption tube. The absorption tube is then sealed and stored until analysis by GCMS.

2.2. Analytical methodology

Samples were quantified against calibrations generated using solvents in methanol spiked onto absorption tubes. Analysis of the tube is performed by ATD-GCMS. The solvents are desorbed from the tubes at 150°C for 7 min onto the cold trap set at 2°C. Maintaining the cold trap at this temperature limits the problem of water vapour icing up the trap whilst still trapping a large number of solvents. The trap is then heated to 250°C for 10 min to desorb the solvents. The volatiles are swept onto the GC column (25 m × 0.32 mm 5 µm CP–SIL 8CB capillary column,



Fig. 1. Photograph of the breath sampler.

Chrompack, London). The GC oven is held initially at 60°C for 2 min, then increased to 180°C at a rate of 15°C/min and then ramped at 30°C/min until 260°C where this is held for 0.5

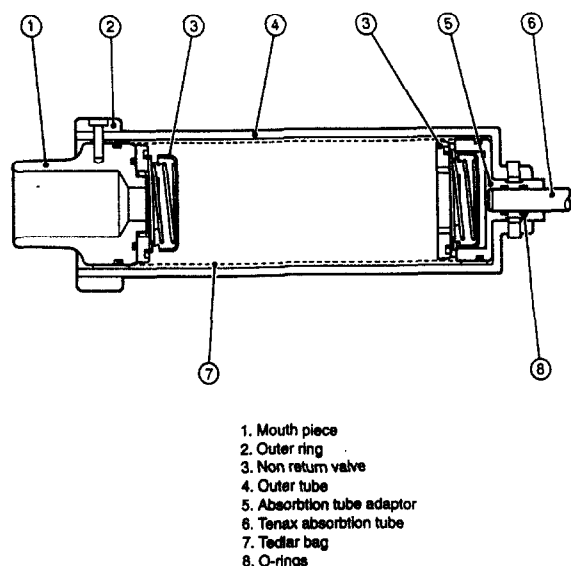


Fig. 2. Cross sectional diagram through the breath sampler.

min. The mass spectrometer is operated in full scan mode (mass 30–200) to allow identification.

The limit for this calibration system is 1 nmol/l of solvent based on three times background interference for toluene, xylene, tetrachloroethene and trichloroethene. Intra-assay variation of spiked tubes was investigated with five commonly used solvents. The results were 4.6% for toluene, 2.3% for trichloroethene, 4.4% for tetrachloroethene, 5.09% for trimethylbenzene and 8.2% for xylene. Inter-assay variation was studied during a series of exposure chamber studies for trimethylbenzene exposure. Inter-assay variation was 9.6%. The method is linear up to at least 1755 nmol/l in breath (2-butanone).

2.3. Validation of the breath sampler

The breath sampler system was validated in the laboratory and tested in workplace studies and in several controlled atmosphere human volunteer studies with a range of typical solvents.

2.4. Laboratory based studies

Prior to the evaluation of the breath sampling system in field and exposure chamber studies, a number of parameters were investigated in the laboratory. The possibility of carry-over of solvent within the sampler and the reproducibility of the sampler were investigated.

A volunteer was exposed briefly to toluene at 50 ppm and was asked to collect a breath sample using the sampler. The captured breath was then transferred onto a sampling tube for analysis. The breath sampler was returned to its ready-to-use position, a process which draws in air from the surrounding environment. The sample of air in the breath sampler was then transferred to a sampling tube to monitor the quantity of toluene contamination left inside the sampler as carry-over from the breath sample. This process was repeated again. All samples were subsequently analysed. The concentration of toluene in the first breath sample was 20 nmol/l. The quantity of toluene carried over to the second tube was 2.9 nmol/l and the amount of toluene found in final wash was less than the detection limit of 1 nmol/l. This demonstrated that carry-over of solvents could occur but that a simple filling and expulsion of air through the sampler cleans it and removes carry-over.

2.5. Field studies

The breath sampling system was used in field studies to investigate exposure to solvents in the workplace and to assess the acceptability of the breath sampler by workers. The breath sampler was used to assess exposure at a shoe manufacturers, at an inks and coatings factory and at a dry-cleaning establishment. At the shoe factory, four workers who were exposed to an adhesive containing toluene and 2-butanone were studied. Blood, breath and urine samples were collected towards the end of the working day. No exposure monitoring was undertaken. At the inks and coating factory, post-shift breath samplers were collected from the workers ($n = 11$) who were exposed to a mixture of solvents. At the dry-cleaning establishment breath samples were collected

from two subjects throughout the working day to investigate the continuous background exposure to tetrachloroethene and also to monitor breath levels immediately after a short burst of high exposure. Continuous ambient monitoring of the establishment showed background levels of 7–8 ppm of tetrachloroethene.

2.6. Controlled atmosphere studies

The breath sampler was used to monitor breath solvent levels in a number of different controlled atmosphere studies. Subjects were exposed, at different times and with differing numbers of volunteers to a range of solvents at the current UK occupational exposure limit for between 2 and 12 h. For the studies involving toluene, tetrachloroethene, acetone, propan-2-ol, 1-methoxy-2-propanol and *n*-hexane, a single volunteer was exposed to each solvent for 2 h. These studies were spread out over 1 year. For the propan-2-ol exposure, the metabolite acetone was measured in breath. The tetrahydrofuran and 2-butanone breath samples were collected from a study investigating dermal absorption of vapour. Five volunteers took part in this study. The study on trimethylbenzene was an investigation into an appropriate biological monitoring method using four subjects and the xylene controlled atmosphere study involved exposing four volunteers to xylene over a simulated 12-h shift system. Breath samples using the novel breath sampler were collected at varying time points during and post-exposure and enabled the reproducibility of the sampler and the rate of elimination of the solvents from breath post-exposure to be studied. All samples were analysed by ATD-GCMS within 2 weeks of collection.

3. Results

3.1. Field studies

Positive results were found in all the industries studied. Breath levels measured in the shoe manufacturing industry ($n = 4$) were 180–274 nmol butanone/l, 215–1926 nmol acetone/l and 2–4 nmol toluene/l. Levels of solvents in breath de-

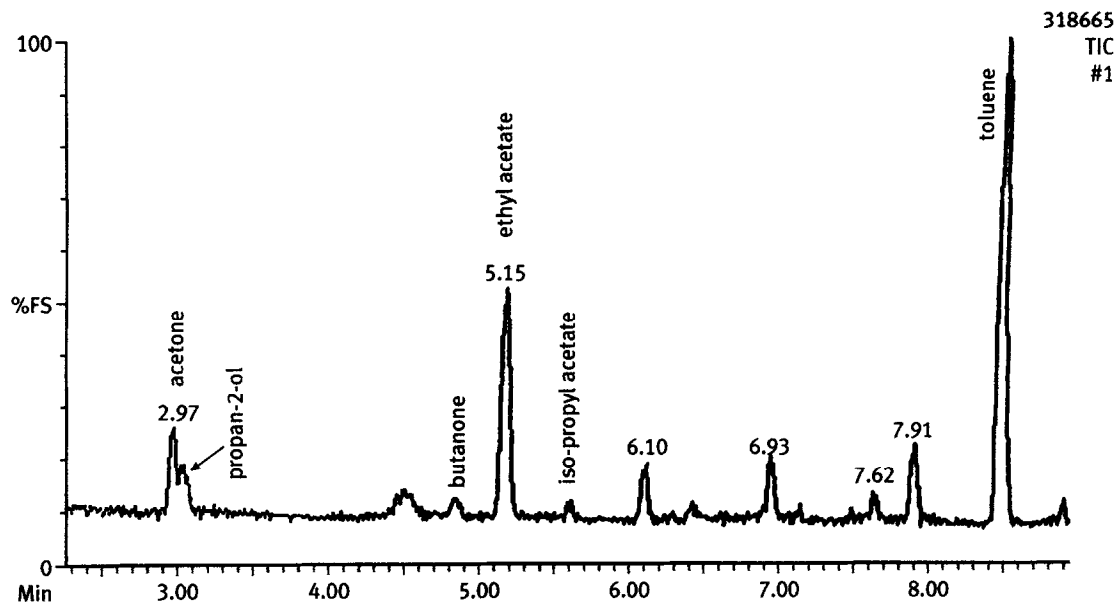


Fig. 3. Total Ion Chromatogram of breath sample from a worker exposed to a mixture of solvents in an inks and coating factory.

ected in workers in the inks and coatings industry ($n = 11$) included ethyl acetate (6.4–25.5 nmol/l), propan-2-ol (3.4–39.3 nmol/l), 2-butanone (0–6.6 nmol/l), isopropyl acetate (0–3.8 nmol/l) and toluene (2–20 nmol/l). Fig. 3 shows a chromatogram from a breath sample of a worker exposed to solvents in the inks and coatings fac-

tory. In the dry-cleaning establishment breath levels of tetrachloroethene ranged from 96 to 556.7 nmol/l ($n = 23$). The higher results coincided with the opening and emptying of the dry cleaning machine.

The breath sampler was found to be acceptable to all workers and volunteers who used it. Work-

Table 1

Reproducibility of results of breath analysis from multiple samples collected with the novel breath sampler during and after exposure to toluene, xylene, trimethylbenzene and tetrachloroethene

	Toluene in samples samples collected in quick succession on three occasions after exposure of 1 volunteer to 50 ppm for < 1 h				Xylene in samples samples collected from 4 volunteers at approx. hourly intervals during exposure to 50 ppm to 50 ppm				Trimethylbenzene in samples collected from 3 volunteers at approx. hourly intervals during			Tetrachloroethene in samples collected in quick succession on two occasions after exposure of 1 volunteer to 50 ppm for 2 h	
				V1	V2	V3	V4	V1	V2	V3			
Mean breath solvent concentration (nmol/l)	53.2	15.8	12.8	128.0	198.0	194.0	251.0	127.0	132.0	105.0	32.0	16.0	
Coefficient of variation (%)	8.0	6.6	8.0	12.0	14.0	15.0	12.0	6.0	15.0	9.0	2.8	5.0	
Number of breath samples (<i>n</i>)	5.0	5.0	4.0	10.0	10.0	8.0	10.0	4.0	4.0	4.0	5.0	7.0	

Table 2

Range of breath solvents and concentrations determined from exposure chamber studies

Solvent	Number of volunteers	Chamber conc. (ppm)	Duration of exposure (h)	Maximum concentration of solvent in breath immediately after leaving the chamber (nmol/l)
Toluene	1	50	2	126
Trimethyl benzene	3	25	4	161
Tetrahydrofuran	5	100	4	300
Tetrachloroethene	1	50	2	82
Acetone from exposure to acetone and propan-2-ol	1	400	2	240
Xylene	4	50	12	120
2-butanone	4	200	2	1755
1-methoxy-2-propanol	1	100	2	233
<i>n</i> -Hexane	1	20	2	761

ers were asked to breathe as normally as possible through the sampler and were encouraged to keep breathing through the system to ensure that the alveolar air was sampled. To make the system as near to normal breathing as possible and to prevent the problem of variable length of time for breath-holding, workers were instructed not to hold their breath before using the sampler. There were no incidences of non-cooperation with regards to this non-invasive form of sampling. Medical staff using the sampler also commented that it was easy to use.

3.2. Controlled atmosphere studies

The breath sampler has also been used to monitor excretion of solvents from human volunteers after controlled exposure chamber studies. More than 500 samples have been analysed from 24 volunteers in exposure to nine different solvents. Table 1 shows the overall reproducibility of the sampler and ATD method for four solvents at various concentrations. The coefficients of variation for sample collection and analysis were between 2.8% and 8% when samples were collected in quick succession from a single volunteer. When multiple samples were collected from volunteers over an extended period as in the xylene and trimethylbenzene studies, the coefficient of variations were slightly larger and ranged from 6% to 15%. This probably reflects both variations in the way subjects exhaled into the

sampler and also changing inhalation concentrations. The results for xylene show that the variation in breath concentrations due to sample collection and analysis (up to 15%) are less than the inter-individual variation in mean breath concentrations (range 128–251 nmol/l).

Table 2 summarises the conditions for each exposure chamber study and reports the maximum breath solvent levels measured at the end of exposure in each case. However, the rapid elimination curves from volunteers exposed to tetrachloroethene, xylene and 2-butanone (Fig. 4) show that sampling in this initial phase may be impractical in the first few minutes. Study of the toxicokinetics of the solvent may identify a time point where more reproducible breath sampling may be undertaken.

4. Discussion

A novel breath sampler has been developed which is simple in design, portable, robust, easy to use and uses laboratory equipment which is generally readily available.

The validation of the breath sampler in workplace studies has clearly shown that it is of practical use in the field. At the inks and coatings factory, five different solvents were determined in the breath of workers. At the dry cleaning establishment, the levels of breath tetrachloroethene found in the workers agreed with that of other studies (Lauwerys et al., 1983; Solet

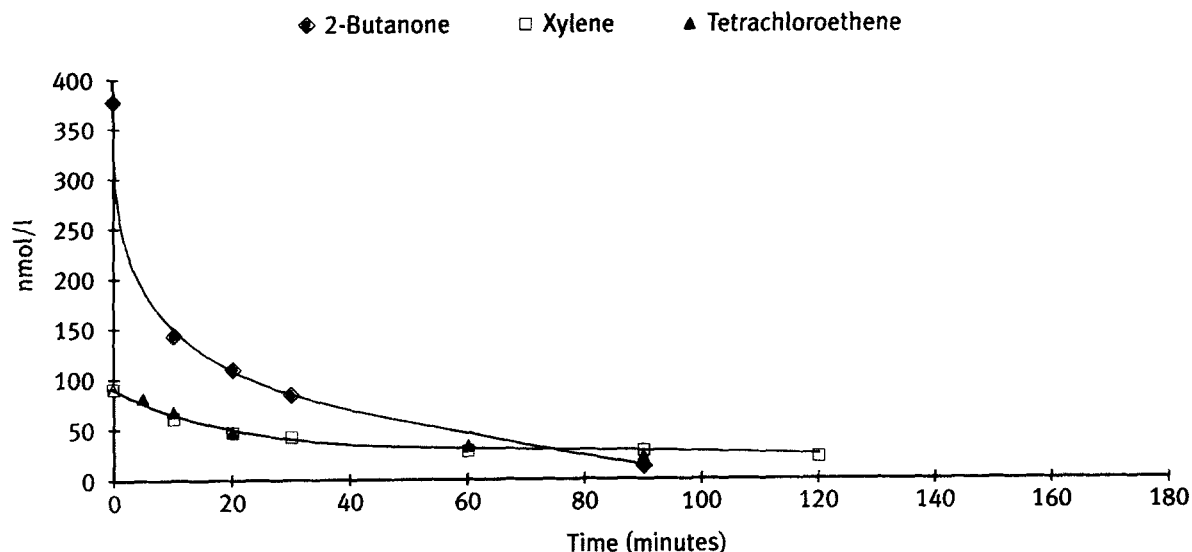


Fig. 4. Elimination of solvents from breath of volunteers after exposure to 2-butanone (200 ppm for 2 h), xylene (50 ppm for 8 h), tetrachloroethene (50 ppm for 2 h), in controlled exposure chamber studies.

et al., 1990). This study demonstrated the flexibility of the breath sampler, and the potential of such a non-invasive sampling technique to collect multiple samples to highlight particular tasks where exposure is highest.

Analysis of breath samples from human volunteer studies has shown that the sampler can be used for a range of solvents and that breath solvents levels can be determined during exposure and post-exposure allowing the toxicokinetics of solvents to be elucidated and sampling strategies for breath sampling to be identified.

In conclusion, the breath sampler has been proven to be easy to use, was found to be acceptable to workers and is of practical use in the field of occupational exposure to solvents.

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