Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Chromatography A, 985 (2003) 39–45
JOURNAL OF CHROMATOGRAPHY A
www.elsevier.com/locate/chroma

Determination of siloxane–water partition coefficients by capillary extraction–high-resolution gas chromatography Study of aromatic solvents

Luigi Nardi

ENEA-National Agency for New Technologies, Energy and Environment, C.R. Casaccia, Via Anguillarese 301, 00060 S.M. Galeria, Rome, Italy

Abstract

Partition coefficients of benzene, toluene, ethylbenzene and xylenes (BTEX), between crosslinked polydimethylsiloxane and water, were determined at room temperature by capillary extraction (a form of in-tube solid-phase microextraction, SPME) coupled to open tubular gas chromatography (in-tube SPME–high-resolution GC). A series of 7–9 repetitive extractions, performed on a 1-ml volume of diluted aqueous BTEX sample by the double-syringe squeeze method, gave exponential regression curves which fit very well with those predicted by partition theory. From the equations of the curves of relative FID response vs. extraction number, experimental K_d were easily calculated and the results compared with literature values. The whole measurement requires about 1 h from the start of the experiment to the final calculation of all BTEX partition coefficients. In-tube SPME resulted in a fast, clean, efficient, and cheaper alternative than the classic 1-cm, externally coated, SPME fiber-holder technique.

Keywords: Siloxane–water partition coefficients; Partition coefficients; Solid-phase microextraction; Benzene; Toluene; Ethylbenzene; Xylenes

1. Introduction

Partition coefficients of organic compounds are of remarkable importance because bioavailability as well as residual concentrations in atmosphere, water and soil strictly depends on partition properties [1,2].

When an ''apolar'' substance is selected as extracting phase vs. water, the partition coefficient (K_d) of an organic compound can be defined as its index of ''hydrophobicity''. Here, the principle ''like dissolves like'' just applies in order to clarify liquid–liquid distribution phenomena, micelle formation, passive membrane transport, fate of chemicals in the environment, etc. [2].

Among various liquid extractants *n*-octanol seems to have gained a special acceptance as reference extractant, and *n*-octanol–water partition coefficients ($K_{o/w}$) are summarized in various reviews [3,4] and other references [5–9]. Due to the presence of a hydrophobic molecule and a hydrophilic end group, *n*-octanol seems preferable as extractant to study structure–activity relationships [2] within living organisms.

Other hydrophobic compounds give K_d values which generally correlate with $K_{o/w}$, therefore the selection of the extractant and that of the method to measure hydrophobicity of substances (i.e. K_d) *rela-*

E-mail address: luigi.nardi@casaccia.enea.it (L. Nardi).

0021-9673/02/$ – see front matter
PII: S0021-9673(02)01457-7

tive to each other depend on practical convenience. Because of this plethora of data, $K_{o/w}$ values reported by different authors do not agree as would be expected for the same substance. In fact, experimental determination of K_d is not a trivial task, being subjected to subtle variables as, for example, adsorptive losses during measurements and/or headspace interferences.

1.1. Alternatives to K_d measurement

Hydrophobicity might be evaluated following various approaches [10]. One of the oldest methods to measure K_d is the conventional ''shake-flask'' procedure [11] useful for molecules with K_d less than $\sim 10^4$. Even for this conceptually easy method an experienced user is necessary to avoid pitfalls. The slow-stirring method [12] may be helpful for more hydrophobic compounds; another one uses RP-HPLC [13–15] and is useful for K_d values less than $\sim 10^6$ in order to avoid very long elution times.

Recently, solid-phase microextraction (SPME) was also applied to measure K_d [16–20] because correlations are expected between $K_{o/w}$ and fiber–water partition coefficients, at least for those ''solid'' coatings acting as a liquid (polydimethylsiloxane (PDMS), polyacrylate). For these phases useful equations (see below) derived from the classical partition law can be applied with success.

When an aqueous sample of volume V_w is equilibrated with a volume V_{sil} of extraction phase, continually renewed each time, the following formula can be applied:

$$K_d = [(C_i/C_{i+1})_w - 1] \cdot V_w/V_{sil} \text{ (isothermal)} \quad (1)$$

where C_i and C_{i+1} are the analyte equilibrium concentrations in the aqueous phase for two consecutive extractions performed on the same (unchanged) sample aliquot. The integer value ''*i*'' indicates how many *identical* batch equilibrium extractions have been carried out. Of course, C_i is always greater than C_{i+1}, and in theory their ratio is a constant. It will be seen below that this accords well with the empirical exponential depletion curve:

$$C(i) = a \cdot e^{-bi} \quad (2)$$

Though depletion experiments [21] may be performed with SPME with standard 1-cm externally coated fibers [22] they may be profitably carried out by capillary extraction–high-resolution (HR) GC (in-tube SPME coupled to capillary GC) [23–26,28]. Recently, in fact, the author has proved that the hyphenation of the microextraction step with GC separation may be performed without any heated injector, just replacing the SPME fibers or stir bar sorptive extraction (SBSE) systems with short tracts of HRGC columns carrying at their ends *press-fits* (=''capillary extractors''). With its press-fit coupling capability, any capillary extractor may easily perform in-tube SPME extraction, and then be joined like a precolumn to the separation column, so taking up the dual role of the SPME system during the extraction step, and that of ''on-column'' injector liner during the (isothermal or temperature programmed) GC elution.

This brief work focuses on the experimental determination of PDMS–water partition coefficients using the capillary extraction–HRGC approach.

2. Materials and methods

2.1. Benzene, toluene, ethylbenzene and xylene (BTEX) standard sample

Benzene (Rudi Pont, purity >99.5%), toluene (redistilled, high purity, single peak by GC analysis), ethylbenzene (Fluka, >99%), *ortho*-, *meta*-, and *para*-xylenes (from Aldrich, purities >99%,) were used as primary substances. A BTEX stock standard mixture (six compounds) was prepared by mixing 2.00 ml of each solvent, and was stored at room temperature (RT) in a screw-closed glass bottle. A diluted BTEX solution (146 ppbv, part-per-billion, v/v) was made by adding 1 μl of the undiluted BTEX stock mix to 1.14 l of Milli-Q water (Millipore, Bedford, MA, USA). Cosolvents were not used, and solubility limits were not exceeded.

2.2. Extractors preparation

The extractors used are pieces of an ordinary ''apolar'' open tubular column made using persilylated glass capillary as support. Glass capillaries (0.474 mm I.D., ~0.9 mm O.D.) were drawn by

means of a laboratory-made glass-drawing-machine. After glass-drawing, they were leached, rinsed, dehydrated, persilylated, and statically coated with PDMS gum PS255 (a Petrarch Systems polydimethylsiloxane with ~1% vinyl groups) according to known (and valuable) recipes [27]. Resulting coatings were immobilized by dicumyl peroxide crosslinking, and finally they were conditioned to 330 °C for not less than 40 min. Extractor lengths were in the range 70–75 cm, and hold-up volumes were between 117 and 133 μl determining the volume of the coated extracting phase.

2.3. Measurement of V_{sil}

The volume V_{sil} of PDMS for each extractor was accurately known since they were cut from a single PS255 HRGC column of 0.474 mm I.D. statically coated by the author. Capillary extractors were cut so that their hold-up volumes are exactly known by weighing (see below). Phase-volumes V_{sil} were in the 0.48 to 0.55 μl range for extractor lengths of about 70–75 cm. From Eq. (1), it is clear that V_{sil} must be known with the greatest accuracy, since it might hide the biggest relative error. Let $C_{coat.sol}$ be the coating solution concentration (by % v/v), used to prepare capillary extractors [$C_{coat.sol}$ = (volume of stationary phase per unit volume of coating solution) × 100]. The phase ratio, β, of an extractor is the ratio of its (after coating) internal gas hold-up volume, V_g, versus V_{sil}:

$$\beta = V_g / V_{sil} \quad (3)$$

If film thickness is very low in comparison with column I.D., V_g is nearly identical to the geometrical void-volume of the uncoated capillary support, so we can determine the phase-ratio directly from the coating solution concentration:

$$\beta = 100 / C_{coat.\ sol} \quad (4)$$

(note that this is an approximate equation, though it is fully valid in the present work).

Rearranging Eqs. (3) and (4), we obtain:

$$V_{sil} = (\text{extractor hold-up volume}) \cdot C_{coat.\ sol} / 100 \quad (5)$$

The accuracy of V_{sil} depends mainly on the exactitude of $C_{coat.\ sol}$. Usually, with normal care, accuracy on V_{sil} is better than 1%.

2.4. Measurement of V_w

Two PTFE-tipped glass syringes (1 ml full scale, SGE Cod. 1MA6-RN-GT) were weighed dry by means of an analytical balance, and then they were rinsed several times with sample to get rid of any air bubble. One syringe was filled with 1.00 ml of sample and finally both syringes were again weighed. This way, considering that 1 mg of sample was the equivalent of 1 μl, the exact volume V_w of sample (volume in the barrel + hold-ups of both syringes) was known with an error less than ±1 μl.

2.5. GC system and analytical method

GC analyses were carried out by means of a Perkin-Elmer 8500 gas chromatograph, provided with a flame ionization detection (FID) system (200 °C) and a programmed temperature vaporizer (PTV) injector that remained unheated all the time. Carrier was helium at 10 p.s.i.g (pressure regulated; 1 p.s.i. = 6894.76 Pa). The laboratory-made glass capillary analytical column was 3 m × 0.16 mm I.D., 0.3 μm film thickness (polydimethylsiloxane gum, crosslinked). GC injection was performed connecting the ''charged'' (after extraction) capillary extractor as a precolumn, through the same PTFE unions used to connect syringes during the extraction step. To avoid carryover, analytes were desorbed from extractor into GC for 3 min, under room temperature carrier flow. During this time they were focused quantitatively at the entrance of a laboratory-made-cryofocusing device [23] that utilized a 0.32 mm I.D. transfer-line dipped into liquid nitrogen. Fast heating of the focusing transfer-line up to 200 °C realized the BTEX injection. Chromatograms were acquired and integrated by peak area, through Borwin software from Jasco (Como, Italy). Calculation and data reduction were partly performed with Microsoft Excel software.

2.6. Capillary extraction set-up and K_d determination

The simple experimental set-up is drawn in Fig. 1.

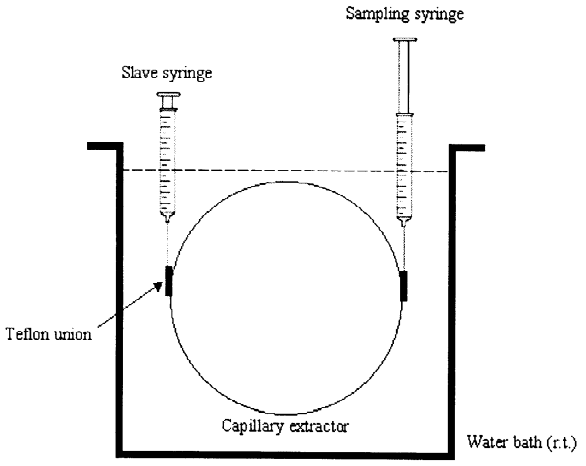

Fig. 1. Set-up for isothermal capillary extraction.

Two 1-ml syringes were sustained with clamps over an isothermal water bath. The capillary extractor ends were joined to syringe needles by a couple of finger-tight PTFE unions (~2 cm long) easily prepared from a piece of PTFE tube of ~0.4 mm I.D.×~1 mm O.D. These unions *must not leak* during fast sample ''squeeze'' to maintain V_w accuracy. Initially one syringe contained the aqueous sample, whereas the second one acted as a slave during the first sample extraction. The process was repeated in the opposite direction. Twenty cycles were enough to reach equilibrium extraction within 20 s (this was checked with a separate set of extractions: from 15–17 cycles and more, BTEX peak sizes remained unchanged within experimental errors). After equilibration the empty syringe was detached, and the residual aqueous sample was removed from the extractor by sucking it carefully within the barrel of the attached syringe. At this stage the liquid–air meniscus should not move back faster than 3–4 cm/s to avoid BTEX losses [28].

3. Results and discussion

Polydimethylsiloxane is widely used in solid-phase microextraction techniques (SPME, SBSE, capillary extraction, etc.): it is ''inert'', easy to coat, and to crosslink. It is also fully stable in water at

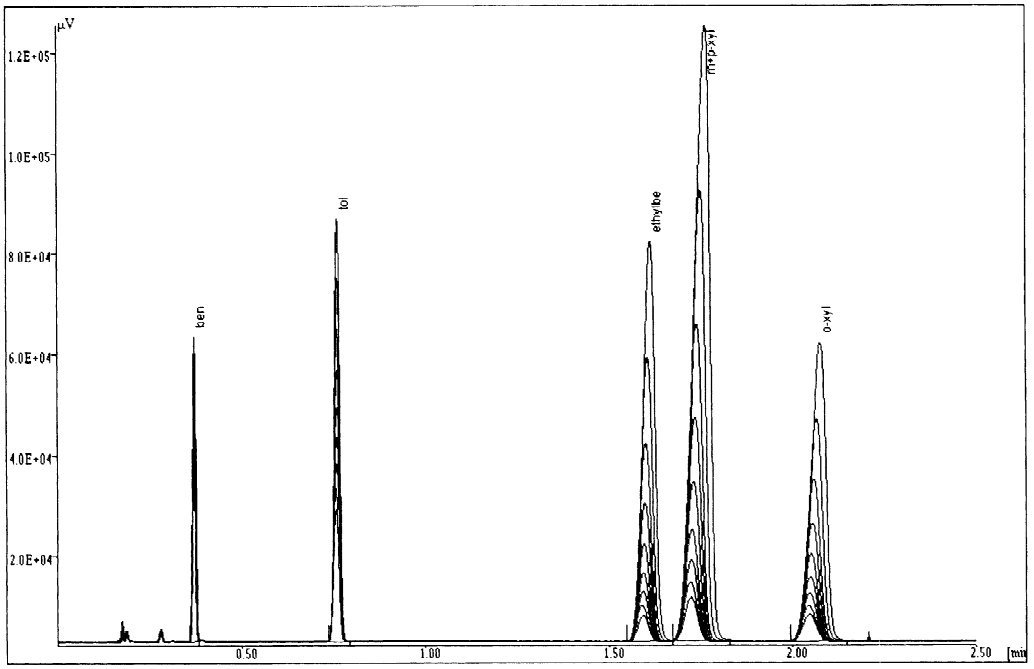

Fig. 2. Chromatograms of consecutive capillary extractions carried out on the same 1-ml sample aliquot. Depletion of successive in-tube extractions is clearly visible.

room as well as higher temperatures. Moreover, it is neither wetted nor swollen by water or diluted aqueous samples of hydrophobic compounds.

A series of 7–9 reiterated capillary extractions gave depletion curves from which exponential regression lines (and so K_d) might be easily calculated.

After an initial extraction with 1 ml sample, the same sample (still within the sampling syringe from the previous extraction) was extracted again with the same neat extractor (cleaned of previous BTEX analytes by preceding GC injection step). During each extraction, depletion of analytes was evident (Fig. 2) because the extractor length and its internal diameter (I.D.) were just selected to make this effect quite visible.

The experimental depletion curves derived from the actual peak sizes (Fig. 2) are plotted in Fig. 3, and those of a duplicate determination are reported in Fig. 4.

The precision of exponential regression lines passing through experimental points is impressive (r^2 correlation coefficients were always better than 0.998) considering that these points were *single* determinations. The repeatability of the depletion measurement as a whole is also remarkable.

From Eqs. (1) and (2), it may be derived that the ratio C_i/C_{i+1} for consecutive batch extractions at equilibrium is:

$$C_i/C_{i+1} = e^b \quad (6)$$

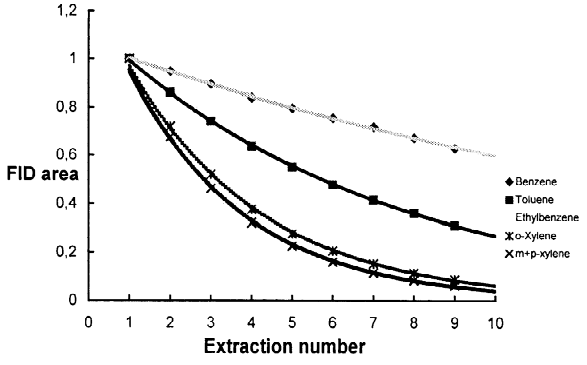

Fig. 3. Experimental depletion curves (exponential regressions) of BTEX analytes. Normalized FID area responses are reported vs. extraction number. Ethylbenzene and $m+p$-xylene curves overlay each other.

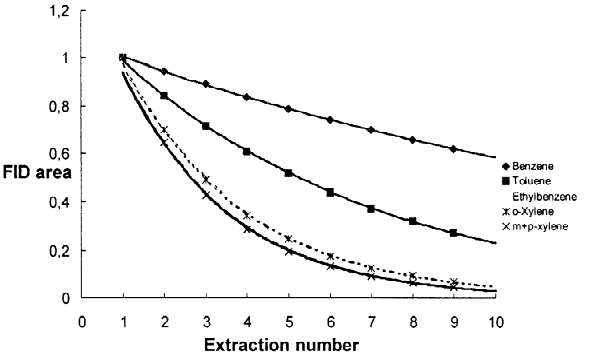

Fig. 4. Duplicate trial (to be compared with Fig. 3) of an independent capillary extraction experiment (determination no. 2 of Table 2).

where ''*b*'' is the constant that appears in Eq. (2) and is also the value that we, experimentally, are looking for to determine K_d.

3.1. Example of BTEX K_d calculation

As noted above, experimental points of depletion curves (Fig. 3) show excellent curve fits. The exponential regression fittings gave ''*b*'' values and quadratic correlation coefficients reported in Table 1. Then, from Eq. (1) we can calculate all BTEX K_d, reported in Table 2 as ''determination no. 1''.

As both V_w and V_{sil} can be measured with rather high accuracy (see Sections 2.3 and 2.4), and since precision of capillary extraction–HRGC is excellent (Figs. 3 and 4 are a clear demonstration) the proposed method of K_d measurement is remarkably simple and particularly accurate, at least to determine K_d between PDMS and water. Incidentally, it was noted that experimental regression lines of ethylbenzene and $m+p$-xylenes were always overlaying,

Table 1
Calculation example (single determination)

Analyte	Exponential regression curve $C(i) = a \cdot e^{-bi}$	r^2 correlation coefficient
Benzene	$y = 1.0598e^{-0.057x}$	0.9988
Toluene	$y = 1.1457e^{-0.1448x}$	0.9997
Ethylbenzene	$y = 1.351e^{-0.3516x}$	0.9986
$m+p$-Xylenes	$y = 1.3328e^{-0.3472x}$	0.9981
o-Xylene	$y = 1.3113e^{-0.3058x}$	0.9991

Table 2
Partition coefficients, K_d, for BTEX, measured from a set of three independent determinations

Determination	K_d					Temperature (°C)	V_w (μl)	V_{sil} (μl)	Hold-up volume (μl)
	Benzene	Toluene	Ethylbenzene	$m+p$-Xylenes	o-Xylene				
1	121	322	872	859	740	22±1	998	0.4823	117
2	146	349	953	943	794	21±1	998	0.4823	117
3	114	320	861	852	732	23±1	1003	0.5482	133
Average K_d ($n=3$)	127	330	895	885	755	22			
RSD (%)	13	4.9	5.6	5.7	4.5				

Table 3
Logarithmic values of K_d measured from the data of Table 1

Determination	Log K_d					Temperature (°C)
	Benzene	Toluene	Ethylbenzene	$m+p$-Xylenes	o-Xylene	
1	2.08	2.51	2.94	2.93	2.86	22±1
2	2.16	2.54	2.98	2.97	2.90	21±1
3	2.06	2.51	2.94	2.93	2.86	23±1
Average log K_d ($n=3$)	2.10	2.52	2.952	2.946	2.88	22
RSD (%)	2.67	0.83	0.81	0.83	0.67	
Literature values [29]	1.94	2.36	2.733	2.737[a]	2.69	RT

[a] Value refers to m-xylene only.

evidently because the K_d of these compounds are almost equal.

Repeatabilities of K_d are quantitatively shown in Tables 2 and 3, and in Figs. 3 and 4 which demonstrate very clearly the high precision achievable by capillary extraction.

Apart from K_d results, Table 2 also reports major parameters involved in the experiments. Table 3 reports logarithmic values of K_d, obtained with the present method, together with some literature data [29].

4. Conclusions

PDMS is an advantageous extraction phase to use with in-tube SPME–HRGC. Capillary extraction, just as SPME and SBSE, works on the partition law principle ($K_d = C_{sil}/C_w$) but is free from errors that easily affect both SPME and SBSE, and from unwanted system peaks typical of SBSE.

The proposed method for K_d determination is remarkably fast [28]. In this work, extractions, GC analyses, and K_d calculations required altogether just 1 h.

Repeatabilities of experimental points are notable, which is somehow a warranty if accuracy is also of concern. Moreover, errors that are ubiquitous to other experimental set-ups (due to headspaces, to losses of analytes outside SPME fibers, linked to vial walls and/or stirring devices adsorption, etc.) are here elegantly overcome by the sound concept of capillary extraction, joined to its fine practicability.

Capillary extractors are cheaply made, just trimming common apolar HRGC columns into pieces. Each extractor shows unpaired inertness toward adsorption losses and an inherent shielding toward cross-contaminations. Last, but not least, capillary extractors allow the mild ''on-column'' injection of 100% of extracted amounts, making in-tube SPME–HRGC a sensitive technique (limit of detection in the 0.01–1 ppb range) even with FID.

As in other branches of analytical science, capillary extraction requires some practice to be profitably employed by the users, but this technique is quite complementary to classic SPME or SBSE. So the effort may be very worthwhile.

References

[1] C. Tanford, The Hydrophobic Effect. Formation of Micelles and Biological Membranes, 2nd ed., Wiley–Interscience, New York, 1980.
[2] T. Braumann, J. Chromatogr. A 373 (1986) 191.
[3] J. Sangster, J. Phys. Chem. Data 18 (1989) 1111.
[4] A. Noble, J. Chromatogr. A 642 (1993) 3.
[5] W.I. Lyman, W.F. Reehl, D.H. Rosenblatt, Handbook of Chemical Property Estimation Methods—Environmental Behaviour of Organic Compounds, American Chemical Society, Washington, DC, 1990.
[6] D. Mackay, W.Y. Shiu, K.C. Ma, Illustrated Handbook of Physical–Chemical Properties and Environmental Fate for Organic Chemicals, Lewis Publishers, Boca Raton, FL, 1992.
[7] D.R. Lide (Ed.), 78th ed, Handbook of Chemistry and Physics, CRC Press, Boca Raton, FL, 1997.
[8] A. Leo, MedChem Database, Daylight Chemical Information Systems, Irvine, CA, 1997.
[9] J.A. Beauman, P.H. Howard, Physprop Database, Syracuse Research, Syracuse, NY, 1995.
[10] A. Paschke, P. Popp, Estimation of hydrophobicity of organic compounds, in: J. Pawliszyn (Ed.), Application of Solid Phase Microextraction, RSC Chromatography Monographs, Cambridge, UK, 1999, p. 140.
[11] OECD Guideline for Testing of Chemicals No. 107, Partition Coefficient (*n*-Octanol/Water), Shake Flask Method, OECD, Paris, 1995.
[12] A. Paschke, P. Popp, Fresenius J. Anal. Chem. 360 (1998) 52.
[13] J.G. Dorsey, M.G. Khaledi, J. Chromatogr. A 656 (1993) 485.
[14] OECD Guideline for Testing of Chemicals No. 117, Partition Coefficient (*n*-Octanol/Water), HPLC Method, OECD, Paris, 1989.
[15] P.A. Martos, A. Saraullo, J. Pawliszyn, Anal. Chem. 69 (1997) 402.
[16] J. Poerschmann, Z. Zhang, F.D. Kopinke, J. Pawliszyn, Anal. Chem. 69 (1997) 597.
[17] W.H.J. Vaes, U. Urrestarazu Ramos, K.C.H.M. Legierse, J.L.M. Hermens, The use of SPME to measure free concentrations and phospholipid/water and protein/water partition coefficients, in: J. Pawliszyn (Ed.), Application of Solid Phase Microextraction, RSC Chromatography Monographs, Cambridge, UK, 1999, p. 129.
[18] A. Paschke, P. Popp, Estimation of hydrophobicity of organic compounds, in: J. Pawliszyn (Ed.), Application of Solid Phase Microextraction, RSC Chromatography Monographs, Cambridge, UK, 1999, p. 140.
[19] J.R. Dean, W.R. Tomlinson, V. Makovskaya, R. Cummings, M. Hetheridge, M. Comber, Anal. Chem. 68 (1996) 130.
[20] W.H.J. Vaes, C. Hamwijk, E.U. Ramos, H.J.M. Verhaar, J.L.M. Hermens, Anal. Chem. 68 (1996) 4458.
[21] L. Urruty, M. Montury, J. Chromatogr. Sci. 37 (1999) 277.
[22] I. Valor, M. Pérez, C. Cortada, D. Apraiz, J.C. Moltó, G. Font, J. Sep. Sci. 24 (2001) 39.
[23] L. Nardi, A new high-performance cryofocalizer-injector for in-tube SPME- and headspace-HRGC applications, in: P. Sandra (Ed.), Proceedings of the 25th International Symposium on Capillary Chromatography, Riva del Garda, Italy, May 13–17, 2002, On CD-ROM.
[24] L. Nardi, In-tube SPME for trace determination of organic compounds in water by HRGC, in: P. Sandra (Ed.), 23rd International Symposium on Capillary Chromatography, Riva del Garda, Italy, June 5–10, 2000, On CD-ROM.
[25] L. Nardi, Am. Lab. 34 (2002) 30.
[26] L. Nardi, Italian patent RM2000A000301, 31st May, 2000.
[27] K. Grob, Making and Manipulating Capillary Columns for Gas Chromatography, Hüthig, Heidelberg, 1986.
[28] L. Nardi, J. Chromatogr. Sci., submitted for publication.
[29] A. Paschke, P. Popp, Estimation of hydrophobicity of organic compounds, in: J. Pawliszyn (Ed.), Application of Solid Phase Microextraction, RSC Chromatography Monographs, Cambridge, UK, 1999, p. 150, Data from Table 4.