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Journal of Chromatography A, 1055 (2004) 159–168
JOURNAL OF CHROMATOGRAPHY A
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Practical aspects of splitless injection of semivolatile compounds in fast gas chromatography

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Received 2 February 2004; received in revised form 15 July 2004; accepted 15 July 2004

Abstract

Possibilities and practical aspects of implementation of splitless injection of larger volumes for fast GC purposes utilizing narrow-bore column, hydrogen as carrier gas, fast temperature programming under programmed flow conditions and commercial instrumentation were searched. As a model sample semivolatile compounds of a broad range of volatility and polarity (7 *n*-alkanes and 19 pesticides) were chosen. Peak shapes, peak broadening and peak areas and its repeatability were evaluated under various experimental set-ups (liner/injection technique combinations). Various factors, such as liner design, injection technique, retention gap length, compound volatility and polarity, the solvent used, initial oven temperature influenced compound focusation and/or maximal injection volume. Combination of analytical column (CP-Sil 13 CB 25 m long, 0.15 mm i.d., film thickness 0.4 μm) with normal-bore retention gap (1 m long, 0.32 mm i.d.) allowed maximal injection volume 8 μl for 4 mm i.d. liner used without any peak distortion when solvent recondensation in the retention gap was employed.

Keywords: Trace analysis; Fast gas chromatography; Splitless injection; Large volume injection; *n*-Alkanes; Pesticides

1. Introduction

There is revived interest in the development and implementation of methods of faster GC. The recent papers [1,2] summarise the advantages of faster GC analysis, general approaches to faster GC method development and to practical aspects of fast gas chromatography with the utilization of open tubular capillary columns with the stress on trace analysis [2]. There is a number of ways to take the advantage of the improved speed of analysis by faster GC. Numerous options exist for pushing the speed of capillary gas chromatography (CGC) analysis faster. According to the classification of types of faster GC analyses, analysis time of fast GC is in minutes range and the usual value of peak width at half height is 0.2–3 s. Thanks to the same and in some case even higher separation efficiency compared to conventional CGC [3–5], the use of fast GC is advantageous for routine analysis and can be typically obtained from columns with an inner diameter of 100 μm [4–10]. Commercial instrumentation of a novel generation is suitable both for conventional and fast GC [2].

To avoid peak width broadening the injection system has to satisfy the required input band width. Any extra-column contribution to band broadening defeats the efficiency preferred by options for faster GC [1]. Splitting injection techniques offer narrow input bands, but only very small sample quantities are introduced onto a column, and/or most of the sample is split to vent. They require low volume injection, which negatively influences the minimum detectable concentration, C_{min}, ($C_{min} = Q_{min}/V_{inj}$, where Q_{min} denotes the minimum detectable amount for a mass sensitive or a concentration for

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0021-9673/$ – see front matter © 2004 Published by Elsevier B.V.
doi:10.1016/j.chroma.2004.07.058

concentration sensitive detectors, V_{inj} is the sample volume introduced onto a column). Due to a low injection volume, the minimum detectable concentration is far too high for many practical applications. To improve the minimum detectable concentration, larger sample volumes have to be injected utilizing non-splitting injection techniques.

Owing to the focussing effects, splitless and on-column injection, and the programmable temperature vaporizer (PTV) have been successfully combined with fast CGC. It needs, however, optimization of various experimental parameters. van Ysacker et al. [11] explored non-splitting injection techniques.

On-column injection is one of the most suitable injection modes for fast GC applications in the field of trace analysis. Besides offering the possibility of injecting larger sample volumes, it eliminates the discrimination of high boiling analytes and offers mild injection conditions for reactive compounds. For a narrow-bore columns a few micro-litres should be considered as large volumes. Usual volumes for fast GC when using narrow-bore (e.g. i.d. 100 μm) analytical columns are ca. 0.1 μl. Our recent publications [8,9] presents configuration (a retention gap (0.32 mm i.d. 1–5 m) coupled to a narrow-bore analytical column (0.1 mm i.d. 5 m)) that allows introduction of 40–80-fold larger sample volumes without any distortion of peak shapes compared to "usual" fast GC set-ups using narrow-bore columns. Focussing effects depend in compound volatility and various experimental parameters. However, there is a limitation of on-column injection analyzing very polar compounds with regard to a retention gap inertness [12]. Analysis of real-life samples might lead to problems with tolerance of the GC system to co-injected matrix components [13], but simple matrices such as water, wine, even cleaned extracts of plant matrices are supposed to be suitable to analyze less polar compounds.

Combination of PTV (with solvent vent mode) with fast CGC with narrow-bore column (i.d. 100 μm) allows even larger sample volume introduction, resulting in excellent LODs; [7,10]. There might be problems with losses of some compounds due to a liquid rinsing or flooding the liner and depression of adsorption in the PTV [7], and/or with thermolabile compounds deposition [10]. Time elapsed for solvent evaporation and sample transfer step are relatively long compared to GC separation time.

Splitless injection has been the most frequently utilized for applications in environmental analysis with conventional GC [14] and also fast CGC [4–7]. Introduction of volumes 0.25 μl [8] up to 1 μl [5] without any peak distortion was observed with the column i.d. 100 μm. The aim of this paper was the translation of splitless injection in fast GC in theory to its real use in practice: to study the feasibility of splitless injection combined with fast GC with larger injection volumes of non-polar and polar semivolatile compounds of a wide range of volatility; to search the influence of various practical factors on peak broadening and precision of analytical results. For this work a column of 0.15 mm i.d. was chosen instead of 0.1 mm. This diameter can be used in majority of GC instruments and offers more flexibility with respect to flow, loadability and operation.

Table 1
List of pesticides and their chemical classes according to their elution order used for experiments

No.	Pesticide	Chemical class
1	Simazine	Triazine
2	Diazinon	Organophosphorus
3	Terbuthylazine	Triazine
4	Dimethoate	Organophosphorus
5	Pyrimethanil	Anilinopyrimidine
6	Chlorpyrifos-methyl	Organophosphorus
7	Fenitrothion	Organophosphorus
8	Chlorpyrifos	Organophosphorus
9	Cyprodinil	Anilinopyrimidine
10	Penconazole	Triazole
11	Captan	Phthalimide
12	Methidathion	Organophosphorus
13	Kresoxim-methyl	Oximinoacetate
14	Myclobutanil	Triazole
15	Tebuconazole	Triazole
16	Phosalone	Organophosphorus
17	Bitertanol	Triazole
18	Cypermethrin	Pyrethroid
19	Etofenprox	Non-ester pyrethroid

No. represents elution order.

2. Experimental

2.1. Chemicals

Standards of pesticides were obtained from different sources and were of purity >95%, list of pesticides used is given in Table 1. Stock solution of pesticides was prepared in Suprasolv toluene (Merck, Darmstad, Germany) with approximate concentration 0.5 mg ml^{-1}. Solution of simazine was prepared separately in Suprasolv acetone (Merck, Darmstad, Germany). Solutions were diluted with selected solvent: ethyl acetate, *n*-hexane and toluene (Suprasolv, Merck, Darmstad, Germany) to get the final test solutions (1.25–10 ng μl^{-1}). Stock solution of *n*-alkanes C_{10}, C_{12}, C_{14}, C_{16}, C_{18}, C_{22}, C_{26} (Fluka, Buchs, Switzerland) was prepared in *n*-hexane at approximate concentration 1 mg ml^{-1}. Standards were weighted on Sartorius Analytic MC1 scales (Sartorius, Götingen, Germany).

2.2. Instrumental

GC measurements were performed on a HP 6890 gas chromatograph (Hewlett-Packard, Avondale, PA, USA) equipped with a split/splitless injector (Agilent BTO septa), an autosampler HP 7683 and a flame ionization detector (FID) operated at 320 °C with rate of data acquisition 50 Hz. Chromatographic column CP-Sil 13 CB 25 m long, i.d. 0.15 mm, film thickness 0.4 μm was obtained from Varian (Middelburg, The Netherlands). A non-polar deactivated retention gap i.d. 0.32 mm (Supelco, Bellefonte, USA) was connected with the chromatographic column with a press-fit connec-

tor 0.32–0.1 mm (Agilent Technologies, Switzerland) and sealed with a polyimide resin (Supelco, Bellefonte, USA). As a carrier gas hydrogen (purity 99.99%) was used (Linde Technoplyn, Bratislava, Slovak Republic); electronic pressure control was employed. Splitless injector was operated at 250 °C for *n*-hexane injection, at 260 °C for ethyl acetate injections and at 300 °C for toluene injections. Single tapered liner i.d. 4 and 2 mm i.d. were utilized (Agilent Technologies, Switzerland). For injection 10 μl syringe with 23–26 s/42 hp point style needle was used. GC separation was under temperature programmed conditions: initial temperature 80 °C 1 min, 60 °C min^{-1} to 290 °C, hold time 5.5 min. Carrier gas flow programming was used: 2.3 ml min^{-1} hold 5.5 min, 2 ml min^{-2} to 3.4 ml min^{-1}.

3. Results and discussion

For the study of various phenomena a model mixture of semivolatile non-polar and polar compounds of a broad range of volatilities was selected: *n*-alkanes (*n*-C_{10}–C_{26}) and pesticides (belonging to different chemical classes with different chemical properties and polarities); the list of selected pesticides according to their elution order is presented in Table 1. For the comparison of behaviour of pesticides with different polarities and study of discrimination of high boiling compounds also *n*-alkanes were present in the mixture. For fast GC separation a narrow-bore column with hydrogen as a carrier gas with a fast temperature programming and a flow programming was used. To increase column capacity and/or carrier gas flow through the column, i.d. 0.15 mm was chosen with semipolar stationary phase CP-Sil 13 CB (86% dimethyl 14% phenyl siloxane).

3.1. Splitless time

For performing splitless injection in fast GC, mechanisms increasing vapours transfer rates from the injector to the column must be employed. First of all liners with small internal diameter were proposed [11]. The column temperature set more than some 30 °C below (pressure corrected) boiling point of the solvent is permitting solvent recondensation in the column what creates vacuum resulting in sucking of vapours from the inlet into the column [11,15,16]. Other possibility to increase the transfer is to increase the column flow by means of increased column head pressure.

In our preliminary experiments we have found that septum head of split/splitless injector is sufficiently sealing the injector only up to the pressure approximately 345 kPa (above atmospheric conditions), which was chosen for further experiments. At higher pressures noticeable hissing of the carrier gas occurred during pulling a syringe needle from the inlet resulting in the loss of the sample vapours and subsequent irreproducible errors of measured peak areas.

Splitless time necessary for adequate transfer of sample vapours was examined by evaluation of the obtained peak areas of *n*-alkanes and pesticides at different splitless times in the range of 0.25–2 min. Because of our interest in the hot splitless large volume injection, single tapered liner (i.d. 4 mm) with internal volume 900 μl was chosen. Injections were performed by an autosampler and the inlet temperature was 250 °C. Two different volumes of *n*-alkanes and pesticides solution in *n*-hexane 1 and 5 μl were injected with five replicates. Analytical column was connected to the normal-bore retention gap 0.32 mm i.d. 1 m long with a glass press-fit connector. During the splitless period the oven temperature was held at 80 °C, what is 45 °C below boiling point of *n*-hexane in the injector (bp at injector pressure is 125 °C calculated according to literature [17]). The time optimum for transport of vapours of multicomponent mixture components also with regards to injected volume to the column is approximately 1 min. The most important fact that can be derived from obtained results is that sample transfer rate is sufficiently high even for wide-bore liner. This fact can be explained by the utilization of normal-bore (0.32 mm) retention gap what strongly improved the transfer rate due to an increasing vapour sucking process and a significantly decreased restriction to flow when compared 0.15 and 0.1 mm i.d. narrow-bore columns. Relative standard deviations (R.S.D.) of peak areas are generally decreased with increasing splitless time.

3.2. Injection volume—influence of liner, retention gap and injection technique

In fast gas chromatography volumes only up to 1 μl were injected with splitless injector without any peak distortion [4]. The maximum sample volume which can be injected is determined by the requirement that the sample vapours must be stored in the vaporizing chamber while they are being transferred into the column [18]. The volume of vapours is dependent on conditions in the injector such as pressure and temperature together with molecular weight and density of the used solvent. Fact that significantly higher pressures of carrier gas are used in fast GC opens possibility to inject larger volume of solution since the higher pressure compress the emerged volume of vapour similarly as in pulsed splitless injection [19]. As we have used the solvent recondensation in the retention gap maximal injection volume is limited also by the ability of the retention gap to retain the flooded zone without entering the analytical column to avoid analytes peak shapes distortion [8,9]. For the determination of maximal injection volume the impact of various parameters was evaluated.

Serious problem affecting peak areas reproducibility in the hot splitless injection is discrimination of higher boiling compounds in the needle during injection. According to Grob Jr. discrimination of semivolatile compounds can be reduced by hot needle injection technique [20]. Other possibility of the sample loss is explosive evaporation of droplets formatted of liquid streaming from the tip of the needle in the case of fast cold needle injection. Droplets falls below the column installation level in the inlet and are evaporated by explosions

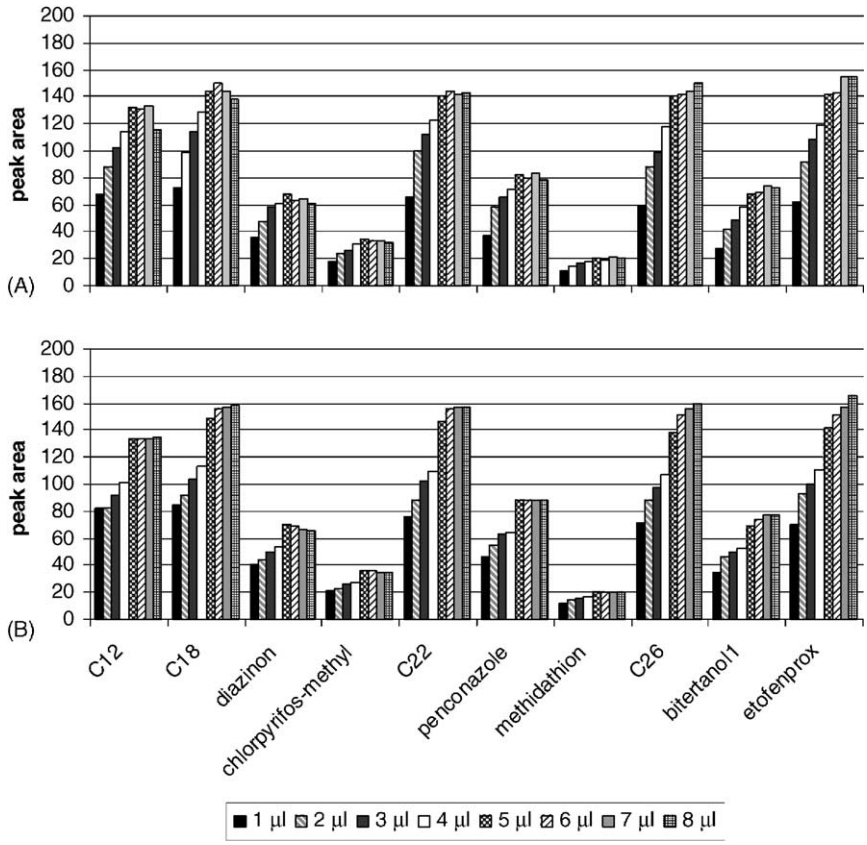

Fig. 1. Graph of the dependence of average peak areas (n = 8) for different injection volumes (1–8 μl) at constant amount per compound injected (10 ng in *n*-hexane) for 4 mm i.d. single tapered liner and different injection techniques; (A) manual cold needle injection, (B) manual hot needle injection.

causing fast transport of mixture of vapours and liquid on the outer side of the inlet liner and their subsequent loss by the split vent [21].

From the point of the injection into the hot splitless injector, the first parameter expected to influence the chromatographic results is the maximal volume of vapours that can be retained in the liner. Two liners with different i.d. and internal volume were tested: 4 mm i.d. liner (900 μl) and 2 mm i.d. liner (250 μl). In Table 2 injection volumes filling the capacity of given liners by vapours by 75 and 100% are shown (inlet pressure 345 kPa and temperature 250 °C) for different solvents calculated by Agilent FlowCalc 2.0 [22]. Our experiments with liners were carried out using analytical column connected to the retention gap (1 m long, 0.32 mm i.d.) with identical inlet pressure and temperature. For injection different volumes (1–8 μl) of solution of *n*-alkanes and pesticides in *n*-hexane were searched. To avoid the influence of the injection amount of compounds on the peak shape and peak broadening, the concentration of compounds was different and the injected amount for all compounds was approximately 10 ng for all injected volumes.

Three injection techniques were tested with regard to peak area value and its repeatability:

- Manual cold needle solvent flush technique.
- Manual hot needle solvent flush technique.
- Fast autosampler injection.

For all injection techniques an increase of peak areas was observed with an increase of the injected volume (while keeping the injected amount/mass per compound constant). We suppose it is caused by the improved vaporisation and sample transfer processes.

Table 2
Comparison of capacity of liners at given conditions and different solvents

Solvent	% of filled liner volume by vapours of injected volume (μl), t = 250 °C, P = 345 kPa			
	4 mm i.d. single tapered liner, internal volume 900 μl		2 mm i.d. liner, internal volume 250 μl	
	75%	100%	75%	100%
n-Hexane	9.1	>10	2.5	3.3
Toluene	7.3	9.8	2	2.7
Ethyl acetate	6.8	9	1.9	2.5

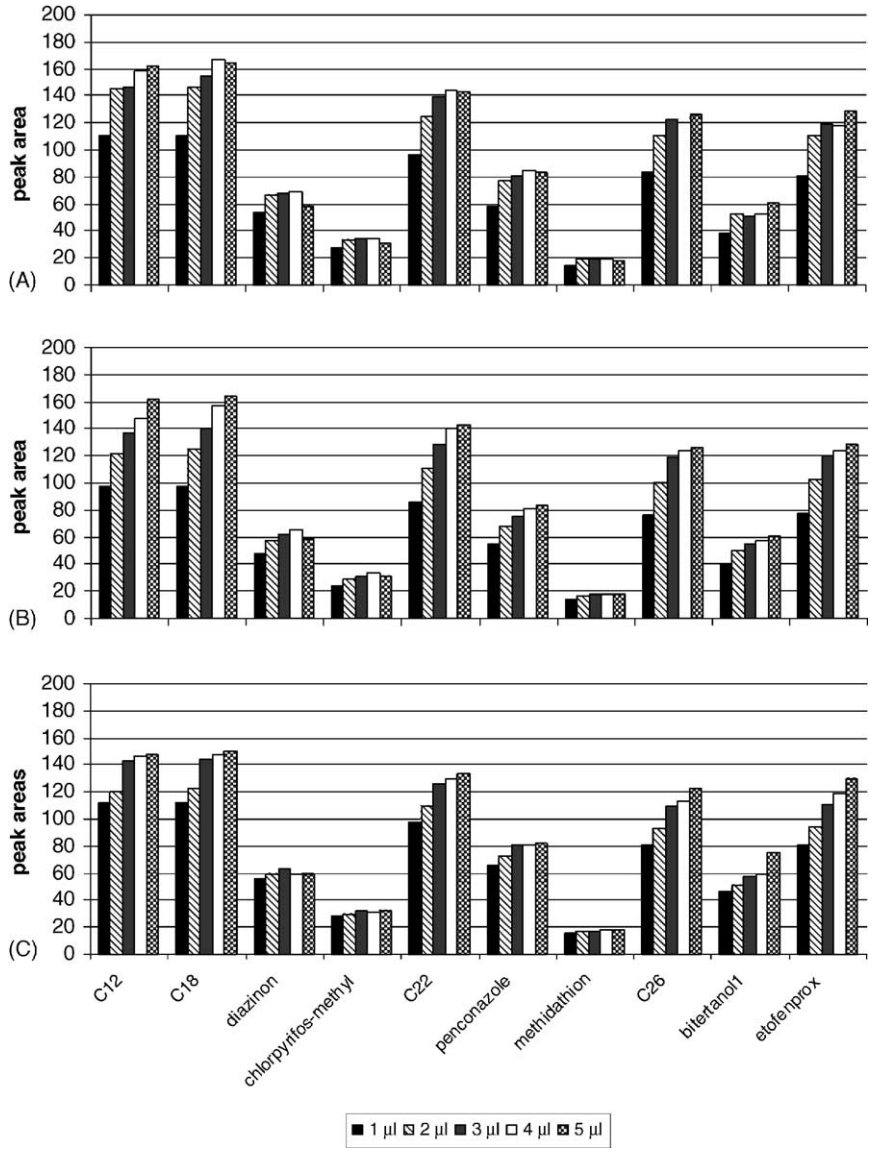

Fig. 2. Graph of the dependence of average peak areas (n = 8) for different injection volumes (1–5 μl) at constant amount per compound injected (10 ng in n-hexane) for 2 mm i.d. liner and different injection techniques; (A) manual cold needle injection, (B) manual hot needle injection, (C) autosampler injection.

Average peak areas (n = 8) of selected compounds are presented in Figs. 1A and 2A for both liners. For 4 mm i.d. liner maximal volume that could be injected was 8 μl (Fig. 1A); for 2 mm i.d. liner 5 μl (Fig. 2A), in both cases higher volume caused peak splitting. Chromatograms of maximal volumes are presented in Fig. 3A and B. The difference in the maximal volumes is supposed to be the consequence of liner capacity and different dilution of vapours in the liner by the carrier gas and the retention gap length. In the narrower 2 mm i.d. liner vapours were less diluted by the carrier gas than in the 4 mm i.d. liner, therefore created vacuum was stronger and the transport of vapours was faster. Subsequently also the recondensation process was finished in the shorter time, so lower portion of transported solvent was eliminated by carrier gas flow rate and larger portion remained in the retention gap causing retention gap overloading. For the 4 mm i.d. liner more diluted vapour enters the retention gap. Now only partial recondensation occurs resulting in an increased liquid capacity. Criteria for determination of liner capacity or liner overflow are peak areas. For the 4 mm i.d. liner peak areas for 8 μl injection volume are lower than for 7 μl injection volume for compounds with volatility up to the n-alkane C_{26}. For the less volatile compounds peak areas increased. This might be caused by the loss of analytes by diffusion to the septum purge vent, less volatile compounds exhibit lower diffusion coefficients and thus their diffusion to the septum purge vent is lower. According to Table 2, maximal capacity of 2 mm i.d. liner is expected to be 3.3 μl for n-hexane, but peak areas have shown only insignificant decease for 5 μl injection. Predicted capacity of 4 mm i.d. liner is >10 μl but decrease in peak areas pointing out on sample loss occurs already for 7 μl injection. Since 2 mm i.d. liner provides

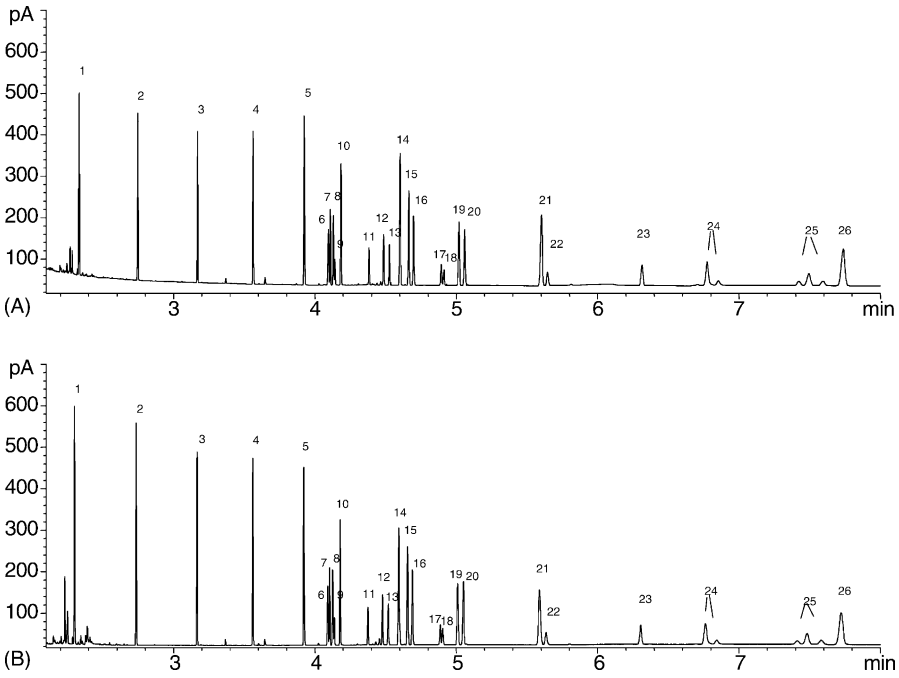

Fig. 3. Chromatogram of pesticides and *n*-alkanes at maximum injection volume, column CP-Sil 13 CB (25 m × 0.15 mm × 0.4 μm), retention gap 1 m × 0.32 mm, temperature program 80 °C hold 1 min, gradient 65 °C min^{-1} to 290 °C, programmed flow 2.3 ml min^{-1} hold 5.5 min, ramp 2 ml min^{-2} to 3.4 ml min^{-1}, splitless injection at 250 °C, purge time 1 min, injected amount 10 ng, FID operated at 320 °C. (A) 4 mm i.d. single tapered liner, injection volume 8 μl; (B) 2 mm i.d. liner, injection volume 5 μl. Elution order: 1—C_{10}, 2—C_{12}, 3—C_{14}, 4—C_{16}, 5—C_{18}, 6—simazin, 7—diazinon, 8—terbuthylazine, 9—dimethoate, 10—pyrimethanil, 11—chlorpyrifos-methyl, 12—fenitrothion, 13—chlorpyrifos, 14—C_{22}, 15—cyprodinyl, 16—penconazole, 17—captan, 18—methidathion, 19—kresoxim-methyl, 20—myclobutanil, 21—C_{26}, 22—tebuconazole, 23—phosalone, 24—bitertanol, 25—cypermethrin, 26—etofenprox.

faster sample transfer due to solvent recondensation in the retention gap its capacity is increased compared to prediction (Table 2) [24].

Hot needle technique provides evaporation by means of a thermospray where aerosol is created and subsequent evaporation from little droplets is faster [23]. As presented in Fig. 1B, peak areas of selected compounds are slightly higher for the hot needle injection technique compared to cold one. Increase of peak areas in the case of 4 mm i.d. liner is more distinct for lower injection volumes than for larger volumes. Higher injection volumes might cause needle to cool down with subsequent formation of streaming liquid. In the case of 2 mm i.d. (Fig. 2A: cold needle; 2B: hot needle) liner almost no differences in peak areas are observed for both injection techniques. In 2 mm i.d. liner transport of heat to the syringe needle and injected liquid is probably higher and thus evaporation is faster.

When peak areas obtained by injection into different liners by different injection techniques are compared, following conclusions can be done:

- Response of more volatile non-polar compounds (*n*-alkanes C_{10}–C_{18}) is highest for 2 mm i.d. liner and is not affected by injection technique.
- Response of more volatile pesticides is not significantly affected by liner type and injection technique.
- Response of semivolatile *n*-alkanes (C_{22} and C_{26}) and less volatile pesticides (eluted after tebuconazole) is highest for 4 mm i.d. liner and hot needle technique, for 2 mm i.d. liner injection technique does not affect their response.

The difference in peak areas among combinations of liner/injection technique are relatively small (<8%), what has not significant effect in real sample analysis, except of volatile compounds (the range of volatility equivalent to *n*-alkanes C_{10}–C_{18}), which provide about 20% higher response utilizing 2 mm i.d. liner when equal injection volumes are compared (results are not affected by the used injection technique).

With autosampler measurements 2 mm i.d. liner was used. Fast autosampler injection is considered as cold needle type of injection. Dependence of peak areas on injected volume is presented in Fig. 2C. Maximal injection volume for autosampler HP 7683 is limited by the ability to use only half of the syringe volume; since 10 μl syringe was used maximal injection volume was 5 μl. As presented in Fig. 3C peak areas are very similar to those obtained by manual injection.

The R.S.D. values were found to be the function of injection technique, injection volume and liner i.d. Hot needle technique provided R.S.D. values of average peak areas in the range of 3–11%, while cold needle technique provided values of R.S.D. in the range of 5–15%, both for 4 mm i.d. liner. Application of the hot needle technique decreased R.S.D. values of average peak areas for 2 mm i.d. liner from the range of 4 to 13% for cold needle to values generally less than 10%. Values of R.S.D. are only slightly dependent on compound

volatility. Repeatability of the autosampler injection was the best and was found to be in the range of R.S.D. 2–7% for the more volatile compounds and 5–10% for the less volatile compounds. When different solvents are compared, the best repeatability of autosampler injection was obtained with ethyl acetate with R.S.D.s ranging from 2 to 7% for all compounds.

3.3. Injection volume—influence of solvent

Since in pesticide residue analysis usually toluene and ethyl acetate are used as injection solvents also influence of the solvent properties on vaporization of sample are evaluated in this work. Autosampler and 4 mm i.d. single tapered liner were used. Temperature of injector was set to 300 °C for toluene and 260 °C for ethyl acetate to keep the same temperature difference between inlet temperature and bp of solvent (pressure corrected) as for the case of *n*-hexane. With regards to ability of retention gap to retain the same maximal volume of the recondensed solvent as for the *n*-hexane, also initial oven temperature was changed to 135 °C for toluene and 90 °C for ethyl acetate as discussed later. In both cases, 1 m long retention gap with i.d. 0.32 mm was utilized. Injection volumes were in the range of 1–5 μl with eight replicates.

In Fig. 4A the obtained peak areas of selected compounds in toluene solution are represented. Response of volatile *n*-alkanes is slightly lower when compared to *n*-hexane, however, the response of high boiling compounds is not affected. Significantly different results were obtained for ethyl acetate solutions (Fig. 4B). Peak areas in all cases, but mainly for lower injection volumes, polar compound and less volatile compounds increased by approximately 10–50% when compared to *n*-hexane or toluene solutions. As bp of ethyl acetate is only by 10 °C higher than bp of *n*-hexane, therefore, such differences of responses are supposed to be caused by the higher polarity of ethyl acetate over *n*-hexane or toluene.

3.4. Focusation—influence of liner, retention gap length, solvent and temperature

To avoid the peak width broadening the injection system has to satisfy the required input band width. In splitless injection the input band width is limited by splitless time. Therefore, focusation effects must be employed to reconcentrate analytes into narrow band that does not significantly decrease efficiency of separation.

For evaluation of focusation effects peak width at half heights were compared in the dependence of injected volume of *n*-hexane solutions for different liner diameters (4 and 2 mm) and different retention gap lengths. Peak widths at half heights of less volatile compounds eluting after C_{16} were con-

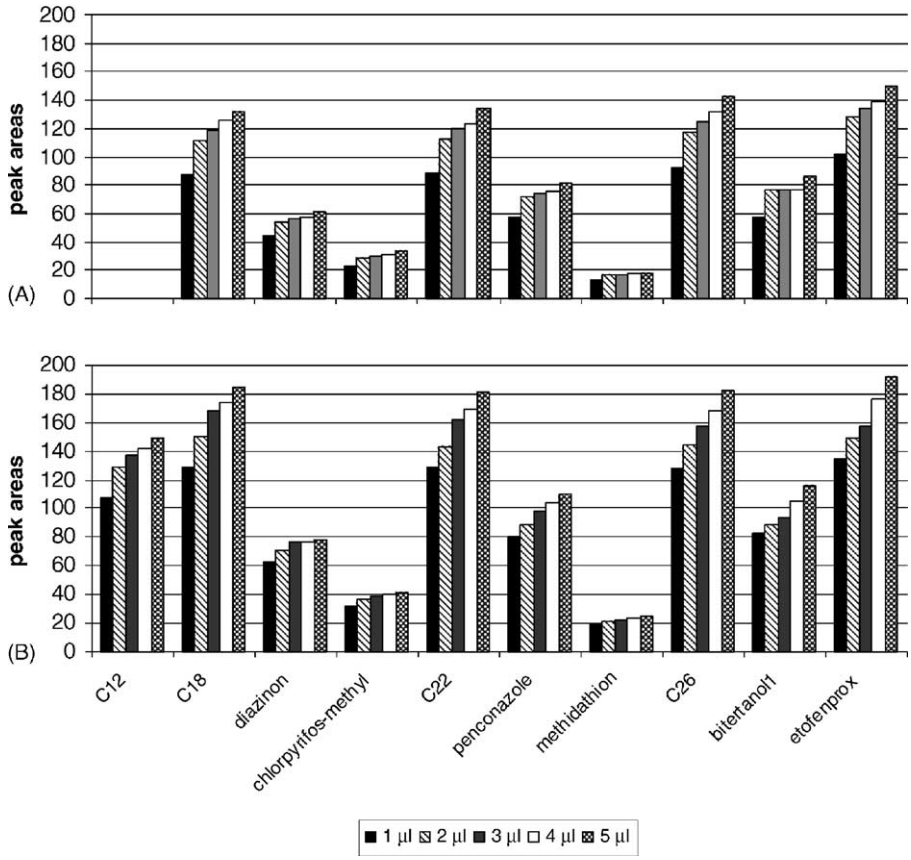

Fig. 4. Graph of the dependence of average peak areas (n = 8) for different injection volumes (1–5 μl) at constant amount per compound injected (10 ng) with autosampler for different solvents: (A) toluene, (B) ethyl acetate.

stant and not dependent on injected volume or retention gap length, there was a phase-ratio focussing. However, there was a difference in maximal injection volume. Half meter long retention gap retained maximal injection volume 4 μl while 1 m long retention gap retained maximal injection volume 8 μl without any peak distortion. An interesting observation was found in the dependence of peak widths when different liners are compared. As illustrated in Figs. 5A and 6A, peak width of more volatile compounds (*n*-C_{10}–C_{16}) is decreasing with injection volume up to approximately 5 μl for 4 mm i.d. liner. This is explained by trapping and reconcentration of these compounds by the solvent effect [9]. More comparable peak widths are obtained for 2 mm i.d. liner and injection of volume ≥3 μl (Fig. 6A). In the wider liner vapours of injected sample are more diluted and thus lower amount of solvent recondeses in the retention gap and subsequently solvent effect is less efficient. For higher boiling compounds solvent effect is no longer important and focusation by stationary phase ratio is sufficient.

As explained above the capacity of retention gap towards maximal retained volume depends on the amount of accumulated solvent during solvent recondensation. Similar experiment was carried out using toluene solutions of *n*-alkanes and pesticides. Different volumes of solutions were injected at different oven temperatures ranging from 80 to 150 °C, 4 mm liner employed. The obtained peak widths for the maximal injection volumes without peak distortion are presented in Fig. 5B. For the lowest oven temperature 80 °C overloading of the retention gap occurred at injection volume >2 μl. The increased oven temperature improved the tolerance of the retention gap towards higher volumes. At oven temperature 135 °C, which is 37 °C below the bp of toluene (bp of toluene is 172 °C, corrected to the injector pressure [17]) injection of 8 μl was possible (*n*-C_{10} and C_{12} were no longer separated from the solvent peak). At the highest oven temperature tested (150 °C) peaks of compounds up to the elution temperature of cyprodinyl are significantly broadened as the stationary phase ratio focusation effect was no longer sufficient. In Fig. 6B the dependence of peak widths on injected volume of toluene solution of *n*-alkanes and pesticides for selected compounds are presented for the oven temperature 135 °C. Solvent effect significantly influenced peak width of

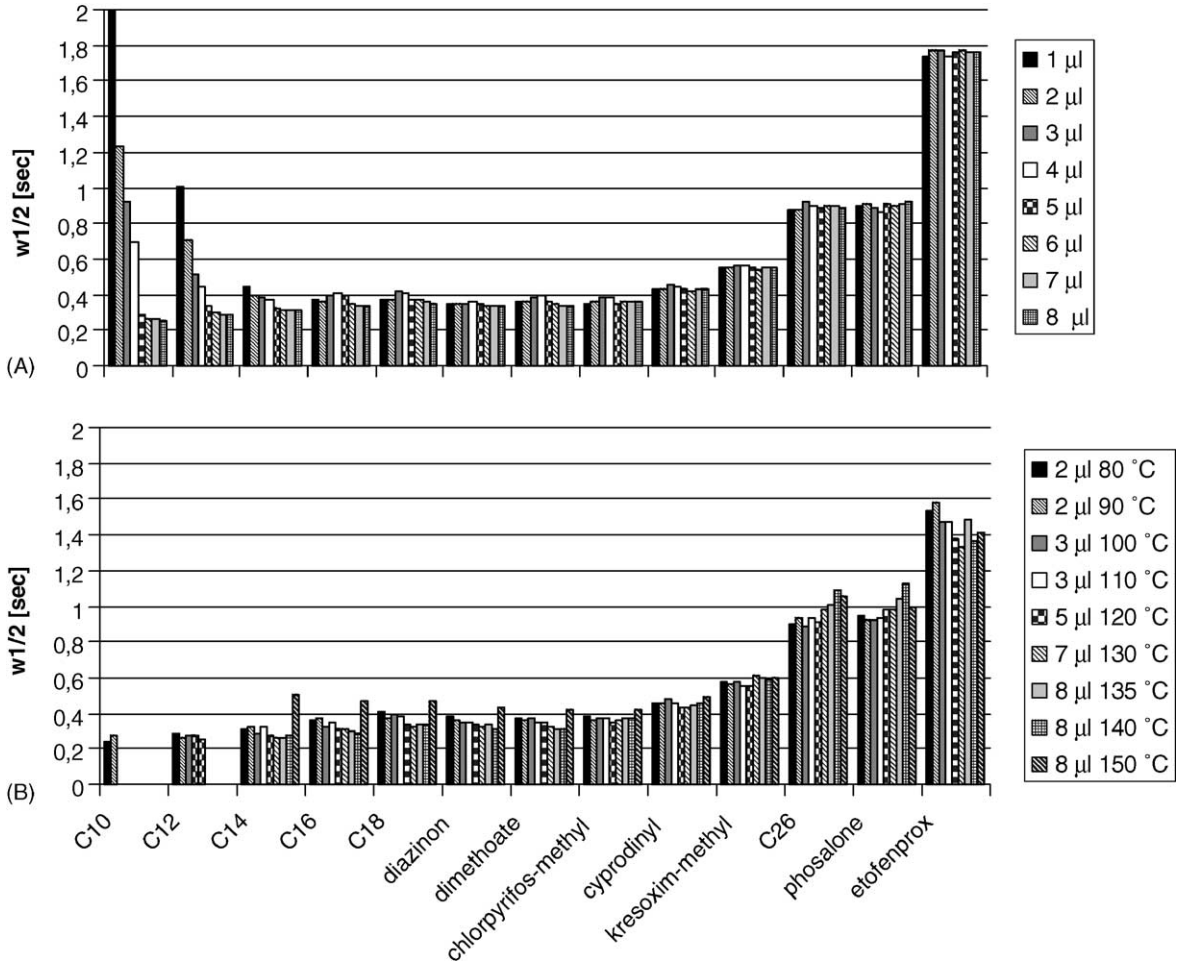

Fig. 5. Graph of the dependence of average peak widths at half heights for different volumes injected at constant amount per compound (10 ng) for different solvents and liners: (A) solvent *n*-hexane, manual hot needle injection ($n = 8$), 4 mm i.d. liner; (B) solvent toluene, manual hot needle injection, 4 mm i.d. liner, maximal volumes injected at different initial oven temperature ($n = 2$).

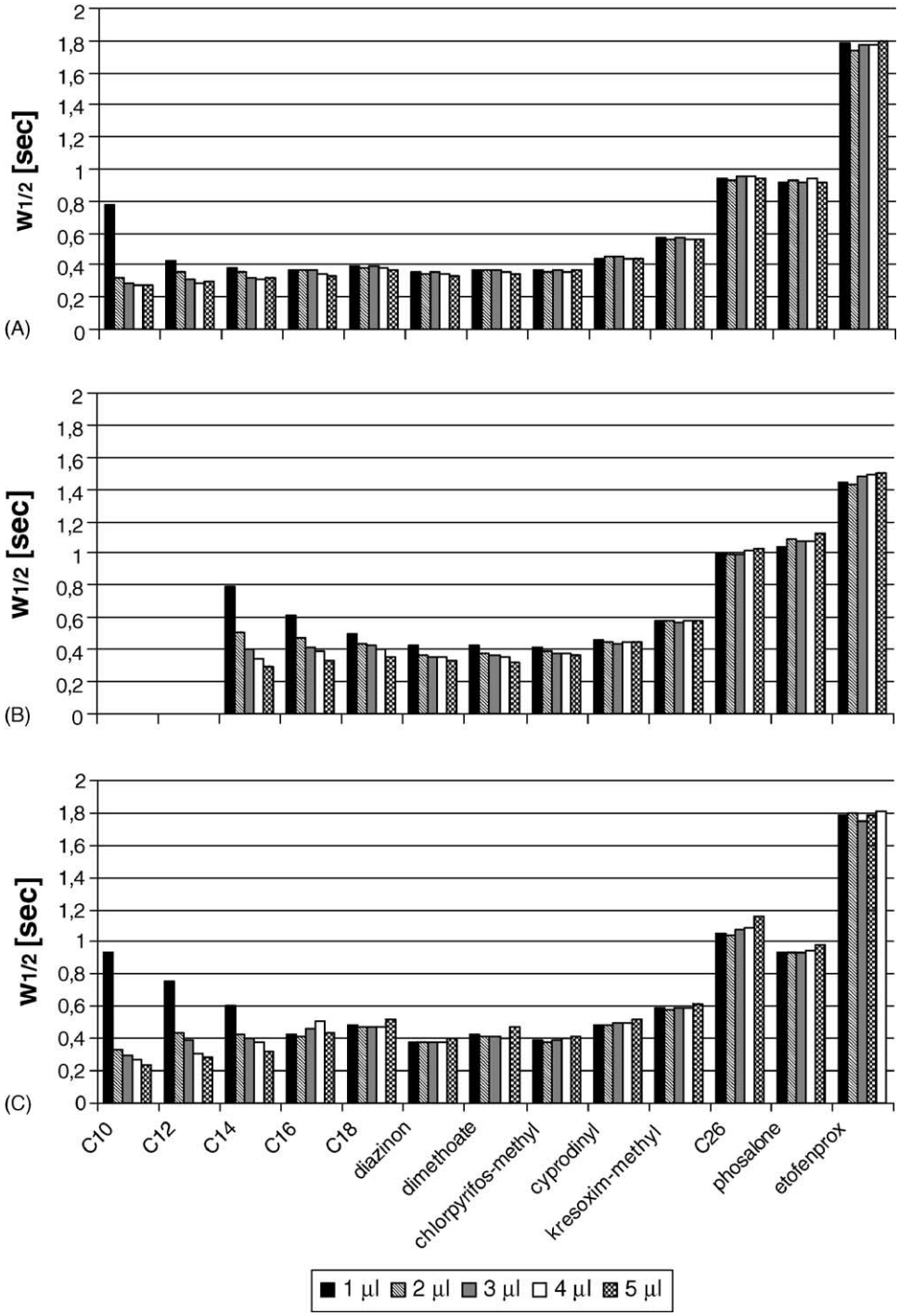

Fig. 6. Graph of the dependence of average peak widths at half heights for different volumes injected by autosampler at constant amount per compound (10 ng) for different solvents (n = 8) and liners: (A) solvent *n*-hexane, 2 mm i.d. liner; (B) solvent toluene, 4 mm i.d. liner; (C) solvent ethyl acetate, 4 mm i.d. liner.

analytes of volatility up to *n*-C_{18}, higher boiling compounds are less affected.

Similar set of experiments was carried out with ethyl acetate. Oven temperature was set to 90 °C what is 40 °C below corrected bp of ethyl acetate (130 °C [17]) and retention gap tolerated injection volume of 8 μl (manual injection) similarly as for toluene and *n*-hexane. The obtained peak widths with autosampler injection are presented in Fig. 6C. Peak widths are increased for all compounds when compared to toluene and *n*-hexane solutions, significantly for the less polar compounds. Peak shapes and peak symmetries were not affected.

It is important to give notice to the potential change of sample transfer rate from the injector to the retention gap

when the initial oven temperature is changed and necessity to adjust the splitless time.

Repeatability of measured peak widths expressed as R.S.D. was generally better than 8% in the entire study.

4. Conclusions

Possibilities of increasing injection volumes in fast GC with standard splitless injector without any peak distortion were searched utilizing narrow-bore analytical column coupled with normal-bore retention gap. The influence of the injection volume, type of liner, retention gap length, type of solvent and oven temperature on peak areas, peak shape and peak broadening of analytes of a broad range of volatility and polarity was investigated. The model mixture contained non-polar compounds *n*-alkanes and pesticides belonging to different chemical classes.

The use of splitless injection in fast GC is limited mainly by the time necessary to transport vapours of a sample to the analytical column. Employment of solvent recondensation in the retention gap increased transport rate of vapours sufficiently even for 4 mm i.d. liner with purge time within 1 min. The most important parameter influencing maximal injection volume was found to be the retention gap length; it must be capable to retain whole volume of recondensed solvent. However, there exists a certain possibility to "tune" its capacity by changing the oven temperature. Liner influences dilution of sample vapours by carrier gas. Single tapered liner with 4 mm i.d. in combination with 1 m long retention gap allowed injection up to 8 μl while direct liner with 2 mm i.d. allowed injection up to 5 μl under the same pressure and temperature conditions. For injection of samples in different solvents initial oven temperature and temperature of the injector should be adjusted. Peak areas increase with increasing injection volume at constant injection amount per compound was observed. Utilization of cold needle or hot needle injection technique did not significantly affect peak areas. Hot needle injection technique provided significantly better peak areas repeatability that was comparable with autosampler injection and was found to be in the range of 3–11% R.S.D. Peak shapes and peak broadening were not affected by injection volume for semivolatile compounds; more volatile compound requiring solvent effect for efficient focusation exhibits decrease of peak width with increasing volume injected.

For performing splitless injection in the fast GC also "slower gases" (e.g. helium) are expected to be utilized as carrier gas, but for the same column i.d. helium requires much higher inlet pressure than hydrogen. Utilization of 0.15 mm i.d. instead of 0.1 mm i.d. column offers more flexibility with respect to flow and the sample transfer process what results in easier operation and compatibility with GC instruments.

How low in concentration levels can we go using this technique and/or practical limitations with real samples is the subject of our next paper [25].

Acknowledgements

The authors gratefully acknowledge support of a part of this research within the framework of the Slovak Grant Agency (Vega, Project No. 1/9126/02) and NATO Project No. SfP 977 983.

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