

Note

Liquid chromatographic separation and behaviour of some triazines on styrene–divinylbenzene columns

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(First received December 23rd, 1988; revised manuscript received March 7th, 1989)

Derivatives of *s*-triazines are widely employed in agriculture as effective herbicide components for weed control in food crops, and their determination is therefore important. A common method for determining triazines is gas chromatography, but it is not suitable for some of the higher molecular weight compounds and those with polar groups. High-performance liquid chromatography (HPLC) seems an attractive alternative as triazines absorb strongly in the UV region. Both reversed-phase^{1–5} and normal-phase applications^{6–8} have been reported.

In the last decade, styrene–divinylbenzene resins have been applied in HPLC and in previous papers^{9–11} the preparation and characterization of polymeric columns were reported. The aim of this study was to investigate the elution behaviour of some *s*-triazines with different substituents at positions 2, 4 or 6 on PRP-1 and Porapak Q polymeric laboratory-prepared columns. The influence of the mobile phase pH on retention data and the relationships between capacity factors (k') and mobile phase composition (ϕ) were investigated using water–acetonitrile eluents. The results are discussed from the point of view of the application of such materials to the preconcentration and determination of *s*-triazines in water.

EXPERIMENTAL

The chromatographic measurements were carried out on a Spectra-Physics (San Jose, CA, U.S.A.) Model 8700 liquid chromatograph, equipped with a Uvidec-100 UV spectrophotometer (Jasco, Tokyo, Japan) set at 220 nm. This wavelength was determined experimentally to be the optimum for all triazines tested. Chromatograms were recorded on a Spectra-Physics SP 4270 integrator. Samples were introduced with a Rheodyne Model 7125 injector (Spectra-Physics), equipped with a 10- μ l sampling loop. HPLC-quality acetonitrile (Rudi Pont, Eurobase, Milan, Italy) and deionized, distilled water were filtered through a Millipore 0.45- μ m filter and degassed with a helium purge. The pH was adjusted with 0.01 *M* phosphoric acid and 0.01 *M* potassium dihydrogen- or sodium monohydrogenphosphate. The reported pH values are the pH of the solution before the addition of acetonitrile. Solvent mixtures are expressed as percent by volume.

The solutes (Table I) (Ciba-Geigy, Basle, Switzerland) were dissolved in

methanol. Their concentrations were about 1 mg/l and allowed suitable responses from the detector without overloading the column. PRP-1 and Porapak Q columns (150 × 4.6 mm I.D.) were prepared as described previously¹¹. Potassium nitrate was used as the unretained solute. All experiments were run in triplicate at a flow-rate of 1 ml/min.

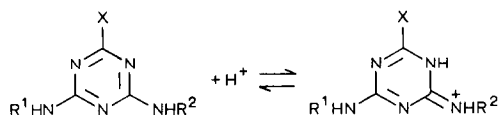
Enrichment experiments were carried out with a commercial PRP-1 cartridge containing about 100 mg of adsorbent. Similar Porapak Q cartridges were prepared in the laboratory. Before use, the traps were washed with 2 ml of acetonitrile and 2 ml of water. A vacuum was applied by a water pump and, after 250–1000 ml of aqueous sample had passed through, *s*-triazines were desorbed passing 1 ml of acetonitrile, which was collected in a glass vial.

The re-usability of the cartridge was investigated by making repeated pre-concentration runs and by restoring the adsorbents with 10 ml of acetonitrile.

RESULTS AND DISCUSSION

All *s*-triazines have similar structures and their physico-chemical properties are determined primarily by the substituent in the 2-position. Aminoalkyl groups in the 4- and 6-positions have a less marked effect. Some characteristics of the studied compounds are given in Table I.

s-Triazines are weak bases and they behave in aqueous solutions according to the equation



where X = Cl, OCH₃ or SCH₃ and R¹ and R² are alkyl groups. One would expect their retention to be affected by a change in the pH of the mobile phase. Consequently, the dependence of the capacity factors on mobile phase pH was evaluated in the pH range 2–11 and the results are shown in Figs. 1 and 2. The pK_a values of individual triazines indicate that the protonation of thiomethyl and methoxy derivatives occurs at pH < 4, while a pH < 2 is required for the protonation of chloro derivatives. Figs. 1 and

TABLE I
CHARACTERISTICS OF *s*-TRIAZINES STUDIES⁶

Compound	Substituent			pK _a	MW
	X	R ¹	R ²		
Propazine	Cl	CH(CH ₃) ₂	CH(CH ₃) ₂	1.85	229.7
Atrazine	Cl	C ₂ H ₅	CH(CH ₃) ₂	1.70	215.7
Simazine	Cl	C ₂ H ₅	C ₂ H ₅	1.90	201.5
Prometryne	SCH ₃	CH(CH ₃) ₂	CH(CH ₃) ₂	4.05	241.3
Prometone	OCH ₃	CH(CH ₃) ₂	CH(CH ₃) ₂	4.20	225.3

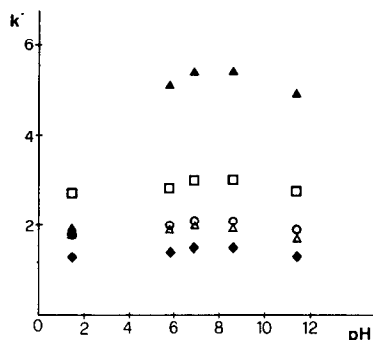


Fig. 1. Plot of k' vs. pH of the mobile phase on PRP-1 column. Mobile phase: acetonitrile-water (60:40). ◆ = Simazine; ○ = atrazine; □ = propazine; ▲ = prometryne; △ = prometone.

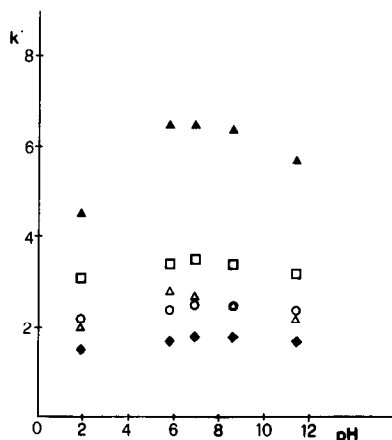


Fig. 2. Plot of k' vs. pH of the mobile phase on Porapak Q column. Mobile phase and symbols as in Fig. 1.

2 indicate that the retention of chlorotriazines is independent of the mobile phase pH, whereas the retention of prometone and prometryne increase with increase in pH up to about pH 7. The behaviour of the compounds studied is similar on the PRP-1 and Porapak Q phases, but the *s*-triazines are more retained on the latter phase. Further, the curvature for prometone is more pronounced on Porapak Q. A pH of 7 was selected as the optimum, as it gives both the highest retention and the best peak symmetry. In acidic mobile phases the peaks of more basic compounds exhibit noticeable tailing.

The relationships between $\log k'$ and acetonitrile volume fraction, ϕ , are shown in Figs. 3 and 4. Again, the retention behaviour of the compounds studied was the same on PRP-1 and Porapak Q phases. *s*-Triazines with the same substituent in positions 4 and 6 showed the retention order $\text{SCH}_3 > \text{Cl} > \text{OCH}_3$. Such a behaviour was observed by Jork and Roth¹ on a $\mu\text{Bondapak C}_{18}$ stationary phase with methanol-water as eluent, and Pacáková *et al.*⁵ observed that methoxy compounds were more retained than chloro compounds on a Separon SI C_{18} phase. Further, methoxy derivatives could not be separated from chloro derivatives on either of these stationary phases. However, on PRP-1 and Porapak Q polymeric phases, such a preparation is possible at all acetonitrile concentration.

With *s*-triazines with the same substituent in position 2, the retention on polymeric phases increases with increasing number of methylene groups on the amino substituents.

Figs. 3 and 4 show non-linear relationships between $\log k'$ and ϕ . Regression analysis showed that all data points can be fitted to a quadratic expression¹²:

$$\log k' = A\phi^2 + B\phi + C$$

The regression parameters and correlation coefficients are given in Table II.

Extrapolation to 0% organic modifier concentration yielded the logarithm of

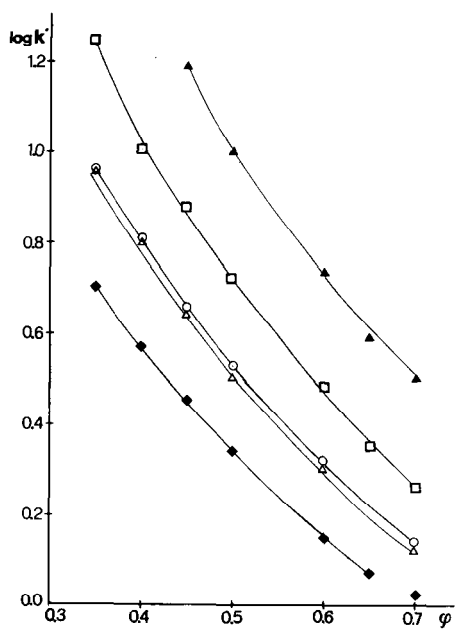


Fig. 3. Plot of $\log k'$ vs. ϕ on PRP-1 column. Mobile phase: acetonitrile-water (pH 7). Symbols as in Fig. 1.

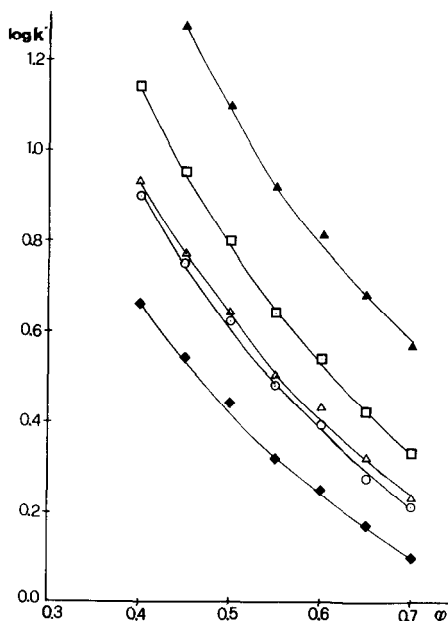


Fig. 4. Plot of $\log k'$ vs. ϕ on Porapak Q column. Mobile phase and symbols as in Fig. 1.

the capacity factor in pure water (k'_w). The high k'_w values suggest that PRP-1 and Porapak Q materials could be effective in extracting *s*-triazines from water. In fact, herbicides are usually present at concentrations below the detection limit (detector sensitivity), so that a preconcentration step is necessary. The suitability of PRP-1 and Porapak Q for enrichment from aqueous solutions was evaluated by sampling distilled water containing *s*-triazines at concentrations below the detection limit (signal-to-noise ratio = 3), *i.e.*, about 0.5 ng of the tested substance in the volume injected (0.01–0.05 ppm).

TABLE II

REGRESSION PARAMETERS OF THE RELATIONSHIP BETWEEN LOG k' AND ϕ

$$\text{Log } k' = A\phi^2 + B\phi + C.$$

Compound	PRP-1				Porapak Q			
	A	B	C	r^a	A	B	C	r^a
Propazine	3.2	-6.2	3.03	0.999	3.7	-6.7	3.20	0.999
Atrazine	2.7	-5.2	2.46	0.999	2.8	-5.4	2.62	0.999
Simazine	2.4	-4.4	1.96	0.999	2.1	-4.2	2.00	0.998
Prometryne	3.7	-7.0	3.60	0.998	3.9	-7.3	3.70	0.997
Prometone	3.4	-6.0	2.65	0.997	2.8	-5.3	2.60	0.997

^a r = Correlation coefficient.

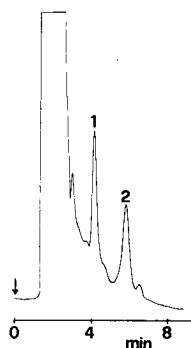


Fig. 5. Chromatogram obtained on sampling on a PRP-1 cartridge 200 ml of tap water containing 0.4 $\mu\text{g/l}$ of atrazine (peak 1) and 0.3 $\mu\text{g/l}$ of propazine (peak 2). Mobile phase: acetonitrile-water (60:40) (pH 7).

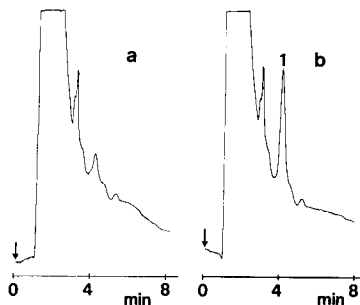


Fig. 6. Chromatograms obtained on sampling on a Porapak Q cartridge 100-ml aliquots of Po river water: (a) unspiked; (b) spiked with 0.8 $\mu\text{g/l}$ of atrazine (peak 1). Mobile phase as in Fig. 5.

Recoveries of herbicides were evaluated by comparing the areas of the peaks obtained for the sample and for a standard solution. The recoveries were 90–100% for all the *s*-triazines and they did not vary with increase in water sample volumes (up to 1000 ml) or increase in herbicide concentration (up to 1 mg/ml). Further, it was observed that after four extractions the adsorbent capacity of polymeric traps had not decreased significantly.

Figs. 5 and 6 show typical chromatograms obtained by this procedure for tap water and Po river water samples, both unspiked and spiked with atrazine and/or propazine.

The results obtained show that polymeric columns could be usefully employed in the analysis of *s*-triazines. Further, such materials could also make it possible to concentrate aqueous solutions of herbicides at parts per 10^{12} levels in a simple manner.

REFERENCES

- 1 H. Jork and B. Roth, *J. Chromatogr.*, 144 (1977) 39.
- 2 A. Di Corcia, M. Marchetti and R. Samperi, *J. Chromatogr.*, 405 (1987) 357.
- 3 N. M. J. Vermeulen, Z. Apostolides, D. J. J. Potgieter, P. C. Nel and N. S. H. Smit, *J. Chromatogr.*, 240 (1982) 247.
- 4 M. Popl, Z. Voznakova, V. Tatar and J. Strnadova, *J. Chromatogr. Sci.*, 21 (1983) 39.
- 5 V. Pacáková, K. Štulík and M. Příhoda, *J. Chromatogr.*, 442 (1988) 147.
- 6 E. Smolkova and V. Pacakova, *Chromatographia*, 11 (1978) 698.
- 7 P. Dufek and V. Pacáková, *J. Chromatogr.*, 187 (1980) 341.
- 8 P. Dufek, V. Pacáková and E. Tesařová, *J. Chromatogr.*, 191 (1980) 115.
- 9 S. Coppi, G. Blo and A. Betti, *J. Chromatogr.*, 388 (1987) 135.
- 10 S. Coppi, A. Betti and S. Caldari, *J. Chromatogr.*, 395 (1987) 159.
- 11 S. Coppi, A. Betti, C. Bigli, G. P. Cartoni and F. Coccioli, *J. Chromatogr.*, 442 (1988) 97.
- 12 P. J. Schoenmakers, H. A. H. Billiet and L. de Galan, *J. Chromatogr.*, 185 (1979) 179.