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Note

Improved chromatographic procedure for determination of 9-(N-methylaminomethyl)anthracene isocyanate derivatives by high-performance liquid chromatography

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Diisocyanates are used extensively in the manufacture of polyurethane products and the determination of these hazardous chemicals in air has been the subject of several studies[1]. With the introduction of high-performance liquid chromatography (HPLC), highly sensitive and selective analytical methods for diisocyanates have been developed.

We recently reported a sampling technique for the determination of diisocyanates in air[2–4]. In this chemisorption method, the diisocyanates are trapped on Amberlite XAD-2 coated with 9-(N-methylaminomethyl)anthracene and the urea derivatives formed are desorbed and determined by HPLC. The method is based on a wet sampling procedure developed by Sangö and Zimerson[5]. In this original chromatographic determination, the excess of reagent is eluted with the solvent front by using acetonitrile–water–triethylamine as the eluent. When more reagent is used as, for example, in the chemisorption technique[2–4], or when the reagent is partly decomposed[5], the reagent peak becomes broad and the urea derivatives elute pn the tail of the reagent peak.

We now report an improved method for the HPLC determination of the urea derivatives.

EXPERIMENTAL

Chemicals

Acetonitrile (Rathburn Chemicals, HPLC grade), toluene (May & Baker, analytical purity), triethylamine (Merck) and N,N-dimethylformamide (DMF) (Mallinckrodt, analytical purity) were used without further purification. 1,6-Hexamethylene diisocyanate (HDI) (Fluka, puriss) and 4,4′-diphenylmethane diisocyanate (MDI) (Merck Schuchard, zur Synthese) were vacuum distilled before use. Water was purified in a Millipore Milli R/Q water purifier. 9-(N-Methylaminomethyl)anthracene hydrochloride was purchased from Sangö and Zimerson[5]. Reagent solution (1.07 mg/ml in toluene) and solutions of the urea derivatives of HDI (4.0 μg/ml in DMF) and of MDI (9.2 μg/ml in DMF) were prepared as previously described[2–4].

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Chromatographic determination

The chromatograhic runs were performed with a Waters Assoc. HPLC instrument, consisting of a WISP 710 A autosampler, a 6000 A pump, an M 440 ultraviolet absorbance detector and an M 730 data module. The instrument was further equipped with a Waters Assoc. radial compression separation system with a C_{18} reversed-phase Radial-Pak A column (100 × 5 mm I.D., 10 μm, Waters Assoc.). A second type of column tested was Nucleosil C_{18} (150 × 4.6 mm I.D., 5 μm, Macherey, Nagel & Co.).

The wash eluent, mobile phase A, consisted of 200 ml of water containing 2.5% of triethylamine, with the pH adjusted to 3.0 with phosphoric acid. The water was diluted to 1 l with acetonitrile. The chromatographic eluent, mobile phase B, consisted of 200 ml of water diluted to 1.0 l with acetonitrile. The detector was operated at 254 nm. The injection volume was 15 μl.

Calculations

The column efficiency was calculated from $N = 5.54\,(t_R/w_{1/2})^2$ where N is the number of theoretical plates, t_R is the retention time of the retained urea derivative and $w_{1/2}$ is the peak width at half-height. The column capacity factor, k', was calculated from $(t_R - t_0)/t_0$, where t_0, the hold-up time for an unretained solute, was determined by the first detector signal deflection from the baseline[6].

Procedure

A new Waters Assoc. C_{18} Radial-Pak A column was washed with mobile phase A for 2 h using a flow-rate of 1.5 ml/min. The column was equilibrated with mobile phase B. The sample was injected and k' and N were determined. A standard solution of urea derivative was then injected repeatedly until breakthrough of the reagent occurred. Each sample was chromatographed for 10 min and a flow-rate of 1.5 ml was used. The reagent was washed out of the column within 1 h using mobile phase A. The column was again equilibrated with the chromatographic eluent and k' and N were determined.

RESULTS AND DISCUSSION

Typical chromatograms for the urea derivative eluted with mobile phase A (the old method) and mobile phase B (the improved method) are shown in Fig. 1. When using triethylamine (mobile phase A), the separation between the reagent amine and urea derivative is not always easily attained, especially when larger amounts of reagents are used[2–4], or when the reagent is partly decomposed[5]. With mobile phase B, these separation difficulties are circumvented as the reagent and most of its decomposition products are retained on the column. For the detection of early eluting peaks, for example, the phenylisocyanate derivative, this method is advantageous as there is no reagent peak that can obscure the derivatives. Shorter analysis times can be obtained with higher flow-rates of the mobile phase.

The affinity of the reagent for the column permits 35–50 sample injections before breakthrough of excess of reagent. Two different new Waters Assoc. C_{18} columns were used in this experiment. The retention of the urea derivative was slightly changed during the procedure and at the end of the injections had increased by 5

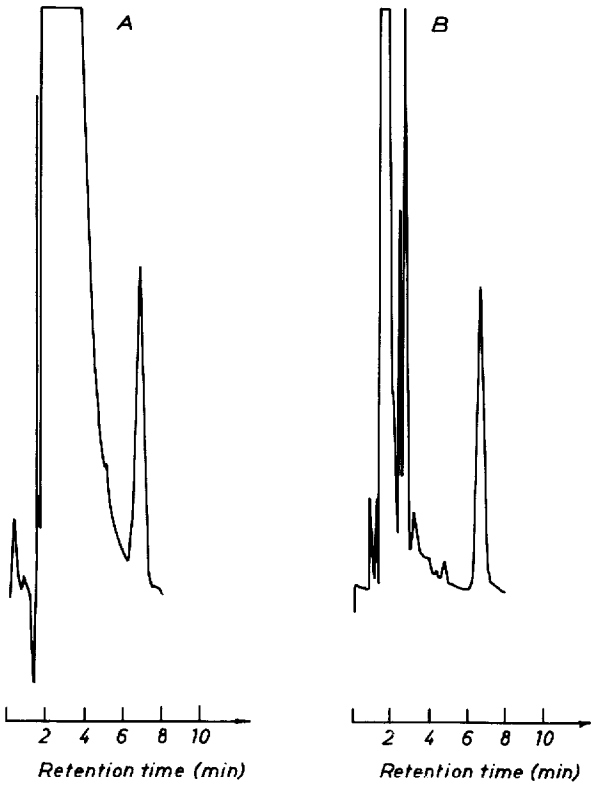

Fig. 1. Chromatogram of MDI–9-(N-methylaminomethyl)anthracene derivative. The concentration corresponds to 0.10 μg/ml of MDI and the concentration of the reagent is 0.50 mg/ml. Volume injected, 15 μl. (A) Mobile phase A (acetonitrile–water–triethylamine), flow-rate 0.8 ml/min; (B) mobile phase B (acetonitrile–water), flow-rate 0.9 ml/min.

sec. The column is easily regenerated by washing with mobile phase A for 1 h. As can be seen from Table I, the procedure did not deteriorate the column. The capacity factor of the column used for the MDI derivative was increased after the first 35 injections. On repeating the procedure, no further change in k' was observed.

Commercial C_{18} reversed-phase columns differ in their behaviour[5–7]. The chromatographic properties are known to be influenced by hydrophobic interactions and by the number of remaining "uncapped" silanol groups. Severe tailing and extremely

TABLE I

COLUMN CAPACITY FACTOR (k') AND NUMBER OF THEORETICAL PLATES (N) OF A NEW WATERS ASSOC. C_{18} COLUMN AND OF THE COLUMN AFTER TREATMENT

Urea derivative	*New column*		*No. of injections*	*Used column*	
	k'	N		k'	N
MDI	7.9	1602	35	9.2	1694
HDI	9.0	1684	50	9.0	1684

long retention times of amine solutes can be explained by silanol–amine interactions. By using alkylammonium compounds in the mobile phase (as mobile phase A), the silanol groups can be blocked and the interactions between amine and silanol groups minimized. However, in columns with a large number of "uncapped" silanol groups it is possible to take advantage of the amine–silanol interactions. Using a Waters Assoc. Radial-Pak A column and a mobile phase without blocking agent, the amine reagent can be retained on the column for several hours.

To investigate whether this chromatographic behaviour was general for commercial C_{18} columns, a Nucleosil C_{18} column was subjected to the treatment described above. However, the amine reagent was eluted within 30 min, which means that the procedure described here is not applicable to this column.

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