

## Note

### Separation and determination of aliphatic alcohols by high-performance liquid chromatography with UV detection

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Aliphatic alcohols can usually be separated by gas-liquid chromatography (GLC) either underivatized or as different ethers and esters<sup>1-5</sup>. The thermal lability of some of these derivatives and incomplete derivatization can, however, cause problems in quantitative analysis.

Silica can be used as a stationary phase to separate underivatized aliphatic alcohols by high-performance liquid chromatography (HPLC), but the low UV absorptivity of these compounds makes the detection of small amounts difficult. To overcome this problem, the extinction coefficient can be improved by making a derivative of the alcohol, e.g., with 3,5-dinitrobenzoyl chloride (DNBC)<sup>6</sup>, prior to HPLC separation. For complete derivatization the reaction mixture must be heated for about 1 h, with simultaneous liberation of hydrochloric acid.

The aim of this study was to develop a sensitive method for the determination of residual 2-ethylhexanol in a thermally labile and acid- and base-degradable polymer. Sample degradation resulted in liberation of more 2-ethylhexanol. These features excluded both GLC and the DNBC derivatization method in HPLC.

Phenyl isocyanate was found to be a rapid and quantitative derivatizing agent for 2-ethylhexanol and other aliphatic alcohols. No catalysts (thermal or chemical) are needed. The general reaction is shown in Fig. 1.

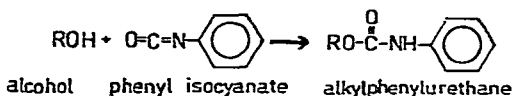


Fig. 1. General reaction of an alcohol and phenyl isocyanate.

## EXPERIMENTAL

### Apparatus

A Hupe & Busch 1010A liquid chromatograph equipped with a Hewlett-Packard 1030B variable-wavelength UV detector, a Hewlett-Packard 3352 Lab Data System for processing of the chromatograms (all from Hewlett-Packard, Karlsruhe, G.F.R.), a Goerz Servogor 541 recorder (Goerz Electro, Vienna, Austria) and a balanced-density, self-packed Sil 60-D10 C<sub>18</sub> reversed-phase column (250 × 4 mm I.D., packing material from Macherey, Nagel & Co., Düren, G.F.R.) was used for

chromatographic separation of the products. Injections were made by a pressure-tight 10- $\mu$ l SGE syringe (Scientific Glass, North Melbourne, Australia). A Perkin-Elmer 402 scanning spectrophotometer (Perkin-Elmer, Beaconsfield, Great Britain), a Perkin-Elmer 621 infrared (IR) spectrophotometer (Perkin-Elmer, Überlingen, G.F.R.), a Jeol FX-60 Fourier-transform  $^1\text{H}$  and  $^{13}\text{C}$  nuclear magnetic resonance (NMR) spectrometer (Jeol, Tokyo, Japan) and a Büchi melting-point apparatus (Büchi, Flawil, Switzerland), together with the liquid chromatograph, were used to check the purity of standard alkylphenylurethanes.

#### *Reagents*

The eluent was a mixture of acetonitrile (ACN; Merck, Darmstadt, G.F.R.; zur Rückstandanalyse) and deionized water. Phenyl isocyanate, N,N-dimethylformamide (DMF), methanol, propanol-1 and -2 and butanol-1 and -2 and 2-methylpropanol-1 (isobutanol) (Merck) were of analytical-reagent grade. Pentanol-1 and -3 and nonanol-1 were of synthetic grade. Triethylamine (Merck) was redistilled before use.

#### *Standards*

O-Alkyl-N-phenylurethane standards were prepared by mixing each alcohol with phenyl isocyanate in a molar ratio of 1:1.1. After cooling the reaction mixture, the white precipitate was collected by filtration and recrystallized from light petroleum (b.p. 443–533 °K.) until the melting point corresponded to the values in the literature<sup>7</sup>. The purity of the products was checked by  $^{13}\text{C}$  NMR and IR spectroscopy, melting point determinations and liquid chromatography.

#### *Sample derivatization procedure*

Samples and standards were dissolved in DMF because it was the best solvent for the polymer samples. Other solvents, such as acetonitrile and dioxane, could also be used. A 1-ml volume of sample solution containing 0.5–10 mg of the alcohol was pipetted into a glass-stoppered test-tube, then 0.5 ml of phenyl isocyanate was added and the tube was shaken vigorously and allowed to stand for 15 min. A 0.5-ml volume of methanol (or, if methanol was to be determined, some other alcohol) was added in order to destroy the excess of phenyl isocyanate. Before injecting the sample, an appropriate amount of internal standard, usually another alkylphenylurethane, was added.

#### *Liquid chromatography*

Derivatized samples and standards (1–10  $\mu$ l) were injected into the chromatographic system with a syringe. The Sil 60-D10  $\text{C}_{18}$  column was in a thermostated oven at 323 °K.

O-(2-Ethylhexyl)-N-phenylurethane [with the corresponding *n*-nonyl compound as internal standard (ISTD)] was determined isocratically ( $\text{H}_2\text{O}$ -ACN = 30:70). For the separation of mixtures of alkylphenylurethanes, different gradients were used. The peaks were monitored at 230 nm.

#### *Peak identification and quantitation*

The chromatograms were processed both qualitatively and quantitatively with

the Lab Data System with an ISTD program for quantitation. The peak height measurement technique gave results that were in good agreement with those obtained with the Lab Data System.

## RESULTS AND DISCUSSION

### *Amount of phenyl isocyanate, effect of temperature, catalysts and time on the derivatization reaction*

At least a 5-fold molar excess of phenyl isocyanate was used to ensure that all residual alcohols were derivatized in the polymer samples, which could contain other isocyanate-consuming compounds also. The effects of catalysts, temperature and time were also checked. It is generally known that tertiary amines catalyse urethane-forming reactions. The addition of 10 mg of triethylamine to the reaction mixture, however, resulted in the formation of two peaks (Fig. 2). Peak No. 2 is probably an allophanate (Fig. 3). After this behaviour was observed, no catalyst was used.

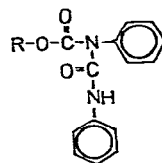
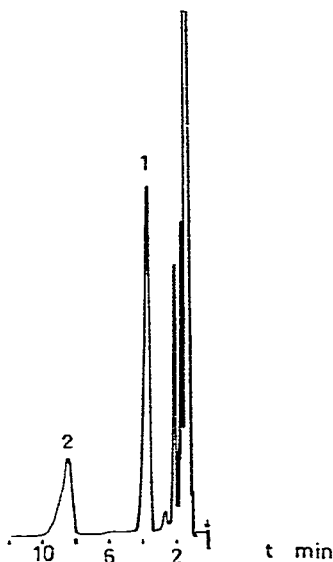


Fig. 2. Chromatogram of 2-ethylhexanol derivatized with phenyl isocyanate in the presence of triethylamine. Peaks: 1 = 2-ethylhexylphenylurethane; 2 = unknown.

Fig. 3. Allophanate of an alcohol.

No significant difference in the reaction velocity at 296° and 323° K was found. Because the polymer samples were unstable at elevated temperature, derivatization at room temperature was preferred.

For pure standards (*ca.* 1% in DMF) the reaction was completed in 5 min. The polymer samples were, however, kept at room temperature for 15 min before destroying the excess of phenyl isocyanate. This was due to the high polymer concentration (10%, w/v) in the samples.

### *Destruction of the excess of phenyl isocyanate*

Because phenyl isocyanate reacts vigorously with water, which is one of the constituents of the eluent, the excess of the reagent was destroyed with an alcohol (amines can also be used). The alcohol or amine must not affect the peaks being determined.

### *Stability of alkyl phenylurethanes*

The stability of the alkyl phenylurethanes was followed for 24 h and no changes in the chromatograms were noticed.

### *Detection*

Alkyl phenylurethanes have two absorption maxima in the UV region, one at 200 nm and the other at 230 nm (in ethanol, Fig. 4). The latter was chosen for the detection wavelength because of solvent absorption at 200 nm. It is possible to use a fixed wavelength detector at 254 nm also, but compared with 230 nm the sensitivity is reduced by a factor of six. The molar extinction coefficient for common alkylphenylurethanes at 230 nm is about  $17,000 \text{ l mole}^{-1} \text{ cm}^{-1}$ ; this allows detection down to the 1–10-ng level.

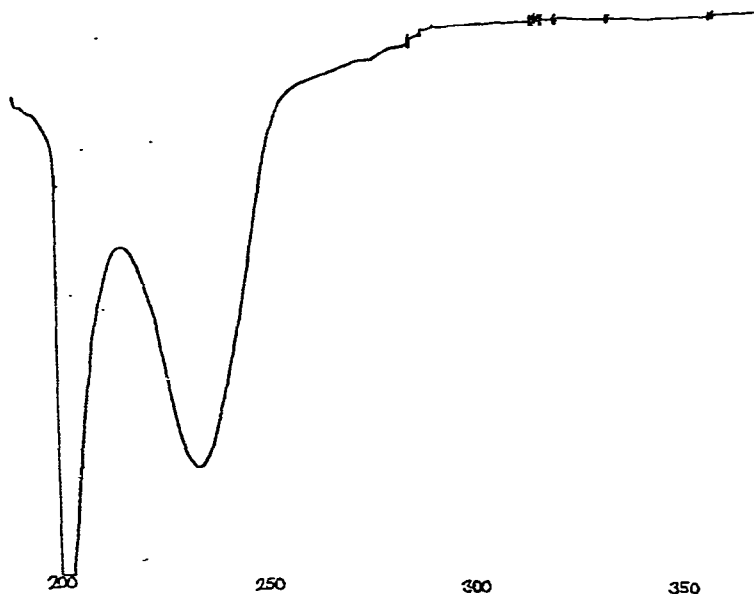


Fig. 4. UV scan of nonylphenylurethane.

### *Linearity*

The most suitable concentration range of the alcohols for the determinations was 0.1–10 mg/ml. The linearity in this range is shown in Fig. 5. Derivatization was carried out separately for each concentration and the corresponding pure crystalline urethane was used as a standard. The reaction was found to be  $\geq 98\%$  quantitative.

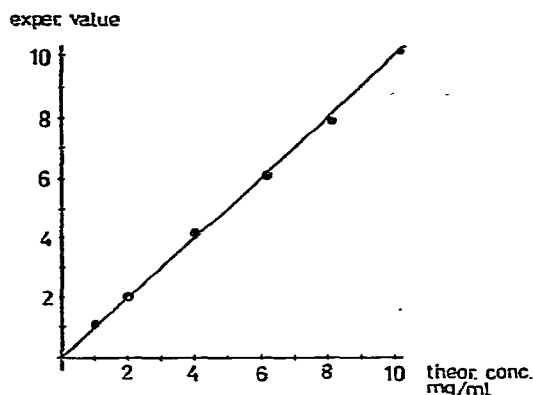


Fig. 5. Linearity curve for 2-ethylhexylphenylurethane.

### Applications

The method described here has been used for determination of residual 2-ethylhexanol in polymer samples (Fig. 6). The method was shown to work well and can be used for any alcohol provided that the corresponding urethane can be eluted.

The method also works with moist samples, but then more phenyl isocyanate must be added because each water molecule consumes two phenyl isocyanate molecules. As a test for the method, a mixture of common short-chain aliphatic alcohols was derivatized and the components were separated as shown in Fig. 7.

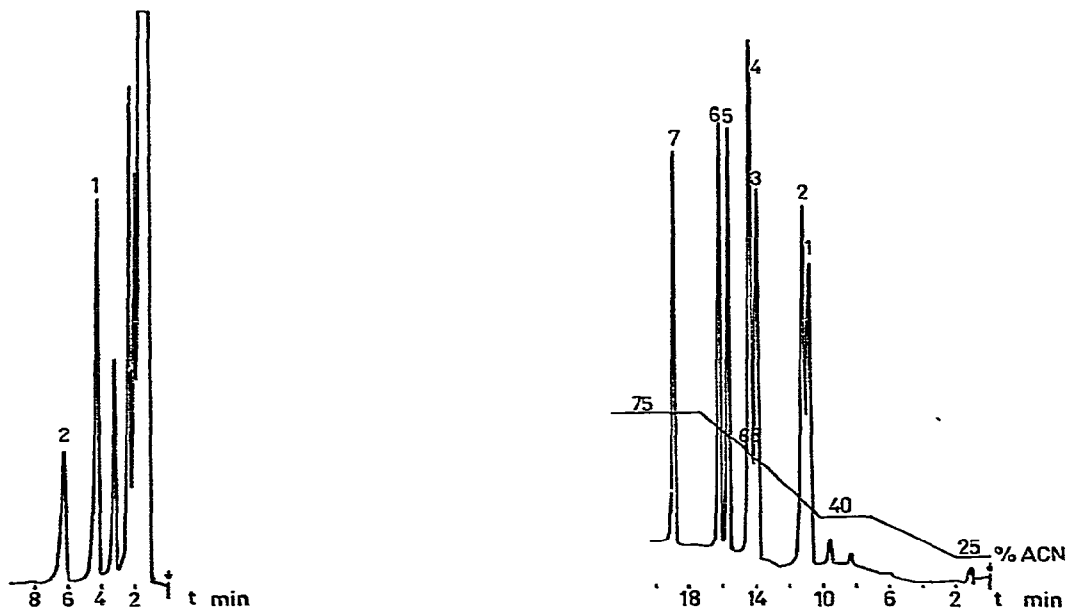


Fig. 6. Typical chromatogram of 2-ethylhexylphenylurethane in a polymer sample. Column, SIL 60-D10 C<sub>18</sub>, 250 × 4 mm I.D.; eluent, H<sub>2</sub>O-ACN (30:70); UV detection at 230 nm, a.u.f.s. = 1. Peaks: 1 = 2-ethylhexylphenylurethane; 2 = nonylphenylurethane (ISTD).

Fig. 7. Separation of alkyphenylurethanes. Column and detection as in Fig. 6. The peaks correspond to the following alcohols (about 1 μg of each); 1 = propanol-2; 2 = propanol-1; 3 = isobutanol; 4 = butanol-1 and -2; 5 = pentanol-3; 6 = pentanol-1; 7 = heptanol-1.

## ACKNOWLEDGEMENT

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## REFERENCES

- 1 A. E. Pierce, *Silylation of Organic Compounds*, Pierce, Rockford, Ill., 1968, pp. 72-159.
- 2 J. K. Haken, *J. Gas Chromatogr.*, 1 (1963) 30.
- 3 R. J. Argauer, *Anal. Chem.*, 40 (1968) 122.
- 4 T. Walle and H. Ehrsson, *Acta Pharm. Suecica*, 7 (1970) 389.
- 5 F. K. Kawahara, *Anal. Chem.*, 40 (1968) 1009.
- 6 J. F. Lawrence and R. W. Frei, *Chemical Derivatization in Liquid Chromatography*, Elsevier, Amsterdam, Oxford, New York, 1976, p. 151.
- 7 A. I. Vogel, *Elementary Practical Organic Chemistry, Part 2, Qualitative Organic Analysis*, William Clowes & Sons, London, 2nd ed., 1966, p. 320.