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ELECTRON CAPTURE GAS CHROMATOGRAPHY WITH SPLITLESS INJECTION ON ISOTHERMALLY OPERATED WIDE-BORE GLASS CAPILLARY COLUMNS

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SUMMARY

The injection of large volumes (up to 50 μl) of diluted samples into a wide-bore isothermally operated glass capillary column has been studied.

A series of *n*-alkanes (C_{11}–C_{16}) have been evaluated as sample solvents by measurement of the resolution between the pesticides *p,p'*-DDD and *o,p'*-DDT. Variations in retention time for the solutes injected in different solvents and in different volumes of the same solvent have been measured. The effect on resolution of changing the injector port temperature has been studied at a constant column temperature of 210°.

A double-injection technique utilizing a combination of a high-boiling (C_{15}) and a low-boiling (C_6) *n*-alkane solvent is described.

More than 500 injections with sample volumes of ⩾5 μl have been made on a 0.77 mm I.D. SE-30 column without significant deterioration of its performance.

INTRODUCTION

The splitless injection technique allows injection of large volumes of diluted samples on capillary columns[1]. Most work in this area has so far been done with flame ionization detection and temperature programming of the column. The electron capture detector (ECD) is extremely sensitive to temperature changes, and for this reason it was desirable to apply the splitless injection technique to isothermal analysis.

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To achieve a high column efficiency and minimum quenching of the ECD the choice of solvent is critical. The splitless isothermal technique has been utilized by Buser in the analysis of polychlorinated dibenzo-*p*-dioxines[2,3] with *n*-tetradecane as solvent at column temperatures between 205° and 225°. Buser used a high injector port temperature (350°) in combination with injection periods of 20 sec, resulting in 90–95% transfer of sample on to the column[2]. In the present study it was possible to keep the split valve closed, giving 100% transfer with an injector port temperature only slightly above that of the column.

The influence of solvent volatility and injection volume on resolution has been studied, using C_{11}–C_{16} *n*-alkanes as solvents.

A double solvent injection technique, comprising initial injection of a high-boiling *n*-alkane followed by the sample in a low-boiling solvent, is described.

Wide-bore capillary columns with helium as carrier gas were chosen. They have a large sample capacity and permit a high volume flow of gas. Hence the construction of "heart-cutting" systems will be facilitated.

EXPERIMENTAL

Apparatus

A Varian 1400 gas chromatograph was fitted with an injector for split/splitless operation (Ultrasep, Turku, Finland). It was maintained at 250° unless otherwise stated.

The SE-30 glass capillary column (10 m × 0.77 mm I.D., film thickness 0.48 μm) was purchased from Ultrasep. It was operated isothermally at 210° throughout this study. Helium was used as carrier gas, with an inlet pressure of 0.2 bar giving a flow-rate of 4.7 ml/min.

The ^{63}Ni ECD of d.c. type was operated at 275°, and 40 ml/min of nitrogen was added through the hydrogen inlet in the detector base. The electrometer setting was either 4×10^{-10} or 8×10^{-10}A.

Chemicals

n-Hexane and *n*-heptane were of analytical grade quality (Merck, Darmstadt, G.F.R.). C_{11}–C_{16} *n*-alkanes (>99% pure) were obtained from Fluka (Buchs, Switzerland) and were used as received. Pesticides were gifts from the Swedish Agricultural Laboratory (Uppsala, Sweden).

A stock solution of the pesticides in *n*-hexane was prepared. Test solutions of *ca.* 1.2×10^{-8} *M* of each pesticide were prepared by evaporation of an appropriate volume of the stock solution and redissolving in the *n*-alkane to be used.

Injections

Usually 5 μl were injected into the gas chromatograph, corresponding to *ca.* 20 pg of each pesticide.

Splitless injection was performed without septum purge flow and without flushing of the injector port throughout this study. The septum purge and split line valves were kept closed in order to assure a constant flow of carrier gas through the column.

The double solvent injection technique was performed by first rapidly intro-

ducing a high-boiling *n*-alkane (*ca.* 1 sec). About 5 sec later the sample dissolved in *n*-hexane was injected with a separate syringe (5 μl in 3–4 sec).

RESULTS AND DISCUSSION

The isothermal splitless injection technique gives retention time variation for the solutes with different solvents and injection volumes. Therefore, calculation of plate numbers is misleading, and column performance was evaluated by measuring the resolution of the two partly overlapping peaks (*p,p'*-DDD and *o,p'*-DDT) according to the formula:

$$R_S = \frac{2\Delta V_R}{w_1 + w_2}$$

where ΔV_R is the distance between peak maxima and w_1 and w_2 are the peak widths at base.

The injector purge delay time is a critical factor when splitless injection is used for quantitative work. Alkane solvents in conjunction with flame ionization detection give broad solvent peaks, and therefore purge delay times of less than *ca.* 1 min must be used. This usually means that solutes of high molecular weight are partially lost through the split line[4]. With the combination of electron capture detection and alkane solvents a closed split valve can be used; thus solute losses are avoided. Furthermore, retention times can be measured accurately because the flow of the carrier gas is constant throughout the analysis.

Choice of solvent

Condensation of the solvent in the first part of the column is of crucial importance when the splitless injection technique is used. Thus, the choice of solvent will depend on the column temperature. Table I shows that the solvent should not be more volatile than *n*-tetradecane at a column temperature of 210°, which is in agreement with the recommendation given by Buser[2]. The use of C_{10} and lower *n*-alkanes led to severe peak distortion and resolution could not be calculated. *n*-Alkanes gave a very low ECD response, and large amounts could be injected without excessively broad solvent fronts appearing although the split valve was kept closed.

TABLE I

RESOLUTION OF *p,p'*-DDD AND *o,p'*-DDT INJECTED IN 5.0 μl OF DIFFERENT SOLVENTS AT A COLUMN TEMPERATURE OF 210°

Injector port temperature, 250°.

n-Alkane solvent	*Boiling point (°C)*	*R_s (mean values)*	*Relative standard deviation (%) (n = 10)*
C_{11}	196	0.88	4.87
C_{12}	216	1.05	5.38
C_{13}	235	1.16	2.47
C_{14}	253	1.22	1.55
C_{15}	271	1.21	4.31
C_{16}	287	1.21	4.93

Solvent volume

The influence of solvent volume on resolution is shown in Fig. 1. Optimum resolution is obtained with injected volumes of 2–4 μl. Too a small solvent volume leads to a sharp decrease in resolution, showing that the solvent in itself is indispensable (*cf.* ref. 5).

The possibility of injecting large volumes of solvent on the wide-bore capillary is also illustrated in Fig. 1. No drastic decrease in resolution was observed even with an injection volume of 100 μl. However, the broad solvent front and impurities in the solvent prevented accurate calculation of the resolution in this case.

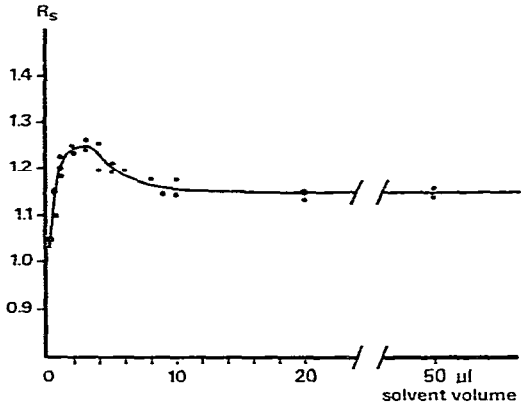


Fig. 1. Resolution of *p,p'*-DDD and *o,p'*-DDT as a function of solvent (*n*-pentadecane) volume.

Droplet and lens formation was clearly observed in the first part of the column with injection volumes of ≥5 μl of *n*-tetradecane or *n*-pentadecane. Nevertheless, the column did not significantly deteriorate during this study, which comprised more than 500 injections.

Injector port temperature

For heat-sensitive samples it is important to use a low injector port temperature. Therefore, the influence of injector port temperature on resolution was studied. The injector port temperature need be only slightly above that of the column (Fig. 2), the split valve being closed to ensure a quantitative transfer of the solutes into the column.

Retention time variation

The retention time for a solute is a function of both solvent volatility (Fig. 3) and solvent volume (Fig. 4). The increase in condensation with the heavier alkane solvents is accompanied by an increase in resolution (Table I), indicating that the condensed amount of solvent with C_{11}–C_{13} is too small to take full advantage of the solvent effect. Fig. 3 indicates a parallel shift to longer retention times for all solutes when going from C_{13} to C_{16} as solvent.

The increase in retention time with larger injection volumes is shown in Fig. 4, which indicates a linear relationship between 10 and 100 μl. This effect must be kept in mind, especially when utilizing the technique for qualitative purposes.

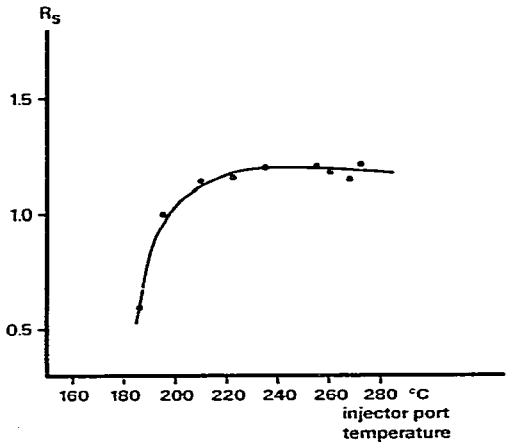
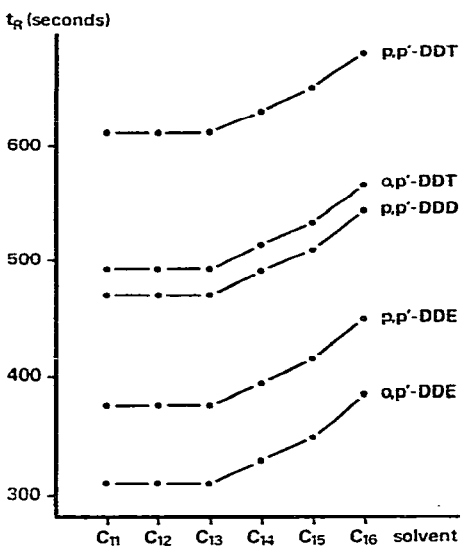


Fig. 2. Resolution as a function of injector port temperature. *p,p'*-DDD and *o,p'*-DDT were injected in 5.0 μl of *n*-pentadecane. Column temperature: 210°.

Double solvent injection

The usefulness of the present technique is hampered by the very limited choice of sample solvent. This drawback was overcome by injection of a suitable high-boiling alkane as a "barrier", followed by injection of the sample dissolved in a volatile solvent, which if it were injected alone would lead to poor resolution (Fig. 5A). The resolution of *p,p'*-DDD and *o,p'*-DDT obtained with this double solvent injection (Fig. 5B) is comparable with that obtained with a split injection (Fig. 5C). The influence of the amount of C_{15} *n*-alkane injected prior to 5 μl of *n*-hexane is illustrated in Fig. 6. The volume of the C_{15} *n*-alkane "barrier" should not be less than 5 μl if the resolution is to be maintained.

The resolution decreases with increasing time delay between injections (Fig. 7).



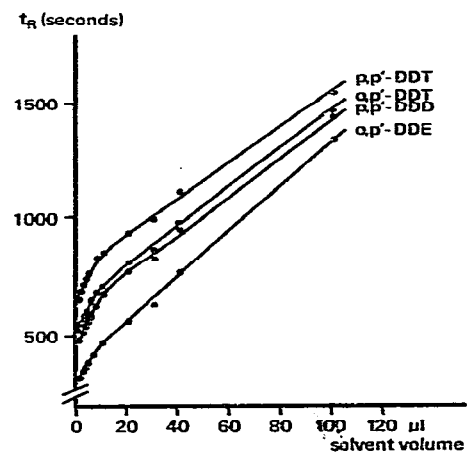


Fig. 3. Variation in retention time for a pesticide mixture injected in different solvents at a column temperature of 210°.

Fig. 4. Variation in retention time for a pesticide mixture injected in different volumes of *n*-pentadecane at a column temperature of 210°.

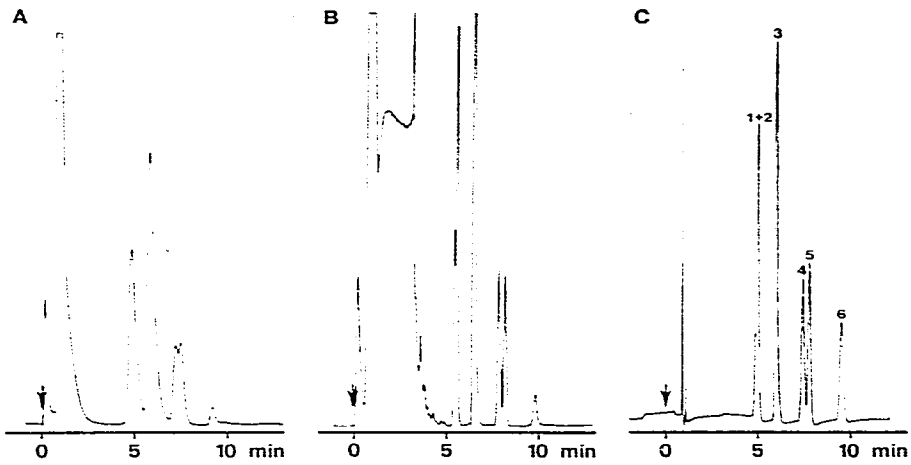
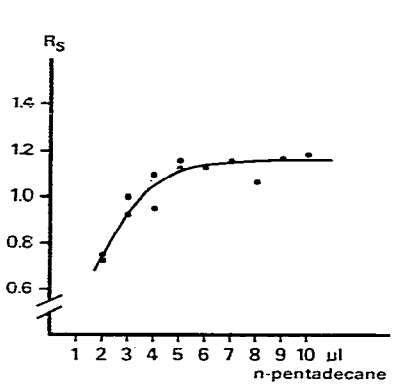


Fig. 5. Chromatograms of the pesticide mixture dissolved in *n*-hexane. A, Splitless injection of 5.0 μl of *n*-hexane solution. B, Double solvent injection. 5.0 μl of *n*-pentadecane injected prior to 5.0 μl of the *n*-hexane pesticide solution. Time between injections was *ca.* 5 sec. C, Split injection of 0.2 μl with a split ratio of 1:15. 1, *p,p'*-DDMU; 2, *o,p'*-DDE; 3, *p,p'*-DDE; 4, *p,p'*-DDD; 5, *o,p'*-DDT; 6, *p,p'*-DDT.



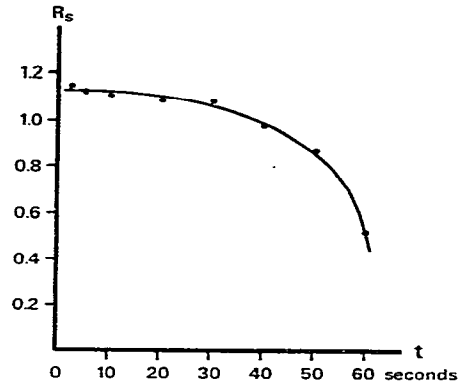


Fig. 6. Resolution as a function of the amount of *n*-pentadecane injected prior to 5.0 μl of *n*-hexane containing *p,p'*-DDD and *o,p'*-DDT.

Fig. 7. Resolution as a function of time elapsed between injection of 5 μl of *n*-pentadecane prior to the solutes in 5 μl of *n*-hexane.

To take advantage of the double solvent injection technique the delay should not exceed *ca.* 30 sec. A delay of 3–4 sec is easily achieved after some practice.

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