Journal of Chromatography, 349 (1985) 167–172
Elsevier Science Publishers B.V., Amsterdam — Printed in The Netherlands

CHROMSYMP. 656

USE OF CYCLODEXTRINS IN CHROMATOGRAPHY FOR SELECTIVE SEPARATIONS, PRE-CONCENTRATION AND PREPARATION OF DEFINED MIXTURES

S. KRÝSL and E. SMOLKOVÁ-KEULEMANSOVÁ*
Department of Analytical Chemistry, Charles University, Albertov 2030, 128 40 Prague 2 (Czechoslovakia)

SUMMARY

Interactions of cyclodextrins or cyclodextrin polymers with substances in the gaseous or liquid phase were studied under stationary and dynamic conditions. The results were utilized for chromatographic purposes, selective separations, pre-concentration and the preparation of well defined gaseous mixtures or solutions with low contents of the test substances. Positional isomers of benzene derivatives were separated by gas–solid chromatography. The inclusion complexes with substances in the gaseous state were obtained under stationary conditions and were used for the storage and preparation of mixtures with low contents of toluene. The formation of the cyclodextrin complexes was followed in the liquid–solid system under dynamic conditions. The gel capacities and the equilibrium constants for the inclusion complexes obtained from the breakthrough curves permit conclusions to be drawn concerning the use of cyclodextrins for selective pre-concentration.

INTRODUCTION

The inclusion properties of cyclodextrins (CDs), *i.e.*, their ability to interact selectively with substances of various types on the basis of the shape and size of their molecules, provide extensive application possibilities in chromatography[1,2]. Selective interactions have been found in both liquid and gas chromatography. In liquid chromatography, cyclodextrins have been used as stationary phases in the form of gels, cyclodextrin–epichlorohydrin[3–6] or polyurethane[7,8] resins or as a polymer formed by polymerization of cyclodextrin with ethylene glycol–di(epoxypropyl) ether[9,10]. Chemically bonded phases with cyclodextrins have also been described[11–16]. Cyclodextrins have also been used as selective components of mobile phases[1,17–19]. In gas chromatography, the interactions of solid cyclodextrins[20], including O-methylated cyclodextrins[21,22] with gaseous components have been used in gas–solid chromatography, and cyclodextrins have been used as components of stationary phases in gas–liquid chromatography[23,24].

The formation of well defined inclusion complexes makes it possible to use cyclodextrins not only in selective separations of gaseous substances, but also in the storage of substances and the preparation of gaseous mixtures or solutions containing

trace amounts of a given component. Cyclodextrin polymers permit the selective concentration of substances from solutions, followed by their chromatographic determination. In continuation of our previous research, these application possibilities were studied.

EXPERIMENTAL

α- or β-CD (ICN Pharmaceuticals, NY, U.S.A.) was deposited on the inert support, Chromosorb W (60–80 mesh), from a dimethylformamide solution. The weight ratio was 3 and 1.5 times that for the complete coverage for α- and β-CD, respectively. The solvent was evaporated at 110°C and a pressure of 13.3 kPa in a vacuum evaporator, and the preparation was dried under identical conditions for at least 10 h. The coverage was determined gravimetrically, after dissolution of a known weight of CD. The α- or β-CD polymer (CDP), prepared by cross-linking of CD with ethylene glycol–di(epoxypropyl) ether in a poly(vinylacetate) medium, was kindly provided by Professor Szejtli (Chinoin, Budapest). The α-CDP contained 45% α-CD, with a grain size of 0.09–0.123 mm and a swollen capacity of 5.0 ml/g of sorbent. The content of β-CD in β-CDP was 49%, the grain size 0.09–0.125 mm and the swollen capacity 45 ml/g. The swollen gels were packed in 50 × 4 mm columns.

The chromatographic measurements were carried out on a CHROM-5 instrument (Laboratorní Přístroje, Prague, Czechoslovakia), equipped with a flame-ionization detector and a TZ 4221 recorder, or on a Packard 428 chromatograph (Packard-Becker, Delft, The Netherlands).

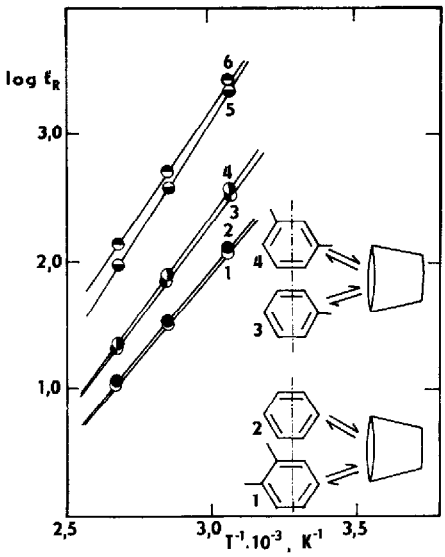
RESULTS AND DISCUSSION

Separation of positional isomers

It has been demonstrated that the selectivity of the separation process based on inclusion can also be used when a solid cyclodextrin interacts with a gaseous substance[25]. This fact has been used for the separation of positional isomers, especially in the benzene series.

The dimensions of the benzene molecule are close to those of the α-CD cavity. Therefore, substituents in various positions on the benzene ring strongly affect this interaction. The character of the interaction is demonstrated in Fig. 1, depicting the temperature dependence of the logarithms of the retention times of benzene, toluene, ethylbenzene and *o*-, *m*- and *p*-xylene; the orientation of the molecules during interaction is also indicated. A comparison of these dependences also indicates a common character of the interactions of benzene and *o*-xylene, toluene and *m*-xylene and ethylbenzene and *p*-xylene, which can be explained by the same orientation of the molecules during the interaction, with only part of the molecule penetrating into the cavity. As follows from the log t'_R values, the stability of the intermediate complexes increases in the series benzene, *o*-xylene < toluene, *m*-xylene < ethylbenzene, *p*-xylene.

A practical consequence of this specific interaction is the separation of *o*-, *m*- and *p*-xylene (see Fig. 2) at 100°C on a stationary phase containing 3.5% of α-CD deposited on Chromosorb W.



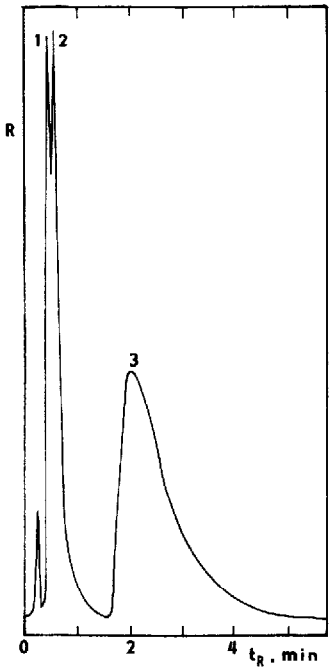


Fig. 1. Temperature dependence of the reduced retention times of aromatics and graphical representation of a possible configuration in the inclusion interaction with the α-CD cavity. 1, *o*-Xylene; 2, benzene; 3, toluene; 4, *m*-xylene; 5, ethylbenzene; 6, *p*-xylene.

Fig. 2. Separation of the xylene isomers on an α-CD phase. Column temperature, 100°C; flow-rate (N_2), 22 ml/min. 1, *o*-Xylene; 2, *m*-xylene; 3, *p*-xylene.

Use of complexes formed by inclusion of substances from the gaseous phase

By exposure of sorbents coated with α- or β-CD to media with a known pressure of the vapour of a substance (usually the saturated vapour at a given temperature), sufficiently stable complexes can be reproducibly prepared that dissociate in aqueous solution or at an elevated temperature and release the original component, the guest. The time dependence of the stability of these inclusion complexes is shown in Fig. 3 for α- and β-CD with toluene. When the complex was stored in a stoppered bottle at 4°C, the stability remained virtually unchanged for over 1 month. The same complex was less stable at 25°C, and the toluene content decreased by *ca.* 15% during the same period. On transferring the complex to an aqueous solution the toluene content decreased to about 25% of the initial value in 1 month.

It follows that inclusion from the gaseous phase permits the simple preparation of inclusion complexes that can be stored for long periods without appreciable decomposition. These complexes can be utilized in many ways. For example, standard mixtures of substances can be prepared for trace gas chromatographic analysis. In view of their low equilibrium constants, complexes with low contents of test components can be obtained that yield aqueous solutions with a known, trace content of a substance that is normally sparingly soluble in water. For example, saturation of a sorbent with 3.5% of α-CD by toluene vapour at 25°C permits the preparation of

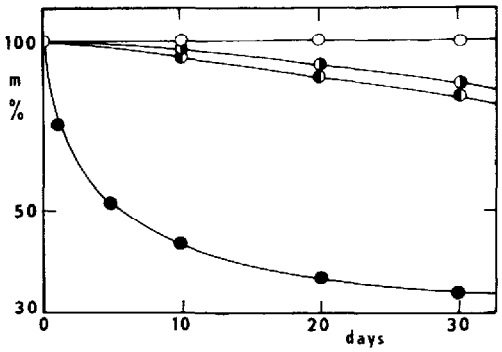


Fig. 3. Time dependence of the stability of the inclusion complexes of α- and β-CD with toluene. m = Relative toluene content in the complex: ○, β-CD–toluene stored at 4°C; ◑, the same complex stored at 25°C; ◐, α-CD–toluene at 25°C; ●, β-CD–toluene dissolved in water at 25°C.

a phase that, on dissolution of 100 mg in 100 ml of water, yields a solution with a toluene concentration of 15 $\mu g\ l^{-1}$, with a 5% relative precision.

Another possibility is the use of thermal decomposition of the inclusion complex. A solid phase is then injected into the injection port. Temperatures of 120–150°C are required to release the volatile component quantitatively. The complexes formed by inclusion from the gaseous phase can thus be used to advantage for calibration of the detection devices at very low concentrations. The relative standard deviation of these methods of injection is 5–10%. A drawback is the possible thermal degradation of the cyclodextrin itself at higher temperatures, as the degradation products may affect the chromatographic packing. The new calibration method can be used in the chromatographic determination of trace pollutants in the atmosphere.

Use of complexes formed by inclusion from solutions

The inclusion properties of CDPs were studied by using selected phenolic compounds as models. The measurements were carried out dynamically with a CDP

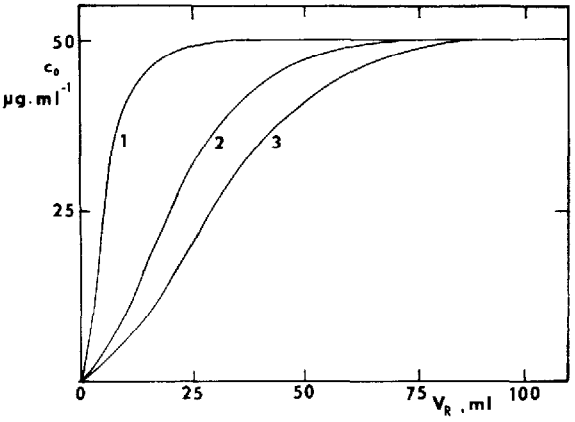


Fig. 4. Dependence of the outlet concentration of nitrophenols on the elution volume for α-CDP using a 50 × 4 mm column, pH = 7.00, I = 0.05. 1, *o*-Nitrophenol; 2, *m*-nitrophenol; 3, *p*-nitrophenol.

TABLE I

CAPACITIES (Q') AND EQUILIBRIUM CONSTANTS (K_{eq}) FOR α-CDP and β-CDP GELS

K_{eq} is defined as the dissociation constant of the inclusion complex (see ref. 25)

Guest	*α-CDP*		*β-CDP*	
	Q' ($mg\ g^{-1}$)	K_{eq} (mol^{-1})	Q' ($mg\ g^{-1}$)	K_{eq} (mol^{-1})
Phenol	–	–	1.49	34.4
o-Cresol	1.66	30.1	2.04	47.3
m-Cresol	2.25	41.6	2.06	47.8
p-Cresol	2.78	50.5	2.88	67.4
o-Nitrophenol	1.81	35.7	2.53	58.6
			(1.06)**	(48.7)**
m-Nitrophenol	6.43	131	4.97	118
	(6.53)*	(133)*	)2.08)**	(96.2)**
p-Nitrophenol	8.85	184	6.74	162
	(9.50)*	(198)*	(2.82)**	(131)**
α-Naphthol	6.75	138	20.3	549
p-Chlorophenol	16.1	357	10.4	260
3,4-Xylenol	–	–	5.08	121

* For pH 3.5.

** For $c_0 = 25\ mg\ l^{-1}$.

column and a mobile phase containing a potential guest as the saturating component. Fig. 4 shows, as an example, the outlet concentrations of *o*-, *m*- and *p*-nitrophenol as a function of the elution volume. From the experimental breakthrough curves the capacities of α- and β-CDP and the equilibrium constants of the complexes formed were found and are given in Table I for phenol and some of its derivatives. It can be seen that the α-CDP capacity increases for phenols substituted in the *para*-position; with β-CDP considerable increase in the capacity for 1-naphthol is observed.

These findings can be used for the selective trapping of some trace substances in waters[26]. For these purposes, a simple apparatus was used, where the flow-rate through a CDP column was controlled by the gas overpressure at the sample surface. The inclusion complexes formed were decomposed by an increase in temperature (*ca.* 170°C) and analysed gas chromatographically, or they were transferred in solution and determined spectrophotometrically. When the trapping efficiencies were determined for model solutions of trace substances, it was found that β-CDP had an 89% efficiency for 1-naphthol and only 34% for phenol. It follows from the literature on ion-exchanger sorbents[27,28], *e.g.*, XAD-2 (1-naphthol, 91%; phenol, 40%), that the efficiency of cyclodextrin polymers is comparable to that of ion exchangers. Moreover, both CDPs exhibit selective properties that are useful, especially in analyses of multi-component pollutant mixtures in water. The use of β-CDP may be of practical significance for the selective trapping of voluminous molecules, *e.g.*, some pesticides and polyaromatics.

CONCLUSION

Inclusion, as a specific phenomenon in the interaction of a host in the gaseous or liquid phase with a solid or dissolved cyclodextrin, offers many possibilities for application in gas and liquid chromatography:

(1) gas chromatography with a solid phase containing cyclodextrin permits some specific separations, especially of positional isomers;

(2) inclusion complexes of cyclodextrins with a known stoichiometry can be used in gas chromatography for the preparation of defined gaseous mixtures or aqueous solutions of trace substances;

(3) polymeric cyclodextrins can be used for the trapping of certain trace substances in waters.

REFERENCES

1 W. L. Hinze, *Sep. Purif. Methods*, 10 (1981) 159.
2 E. Smolková-Keulemansová, *J. Chromatogr.*, 251 (1982) 17.
3 J. Solms and R. H. Egli, *Helv. Chim. Acta*, 48 (1965) 1225.
4 Societe des Produits Nestlé, *Neth. Pat. Appl.*, 6 505 361, 1964.
5 N. Wiedenhof, *Stärke*, 21 (1969) 163.
6 J. L. Hoffman, *Anal. Biochem.*, 33 (1970) 299.
7 Y. Mizobuchi, M. Tanaka and T. Shono, *J. Chromatogr.*, 194 (1980) 153.
8 Y. Mizobuchi, M. Tanaka and T. Shono, *J. Chromatogr.*, 208 (1981) 35.
9 B. Zsadon, M. Szilasi, F. Tödös, K. H. Otta, É. Fenyvesi and J. Szejtli, *Acta Chim. Acad. Sci. Hung.*, 100 (1979) 265.
10 B. Zsadon, M. Szilasi, F. Tödös, É. Fenyvesi and J. Szejtle, *Stärke*, 31 (1979) 11.
11 M. Tanaka, Y. Mizobuchi, T. Sonoda and T. Shono, *Anal. Lett.*, 14 (1981) 281.
12 M. Tanaka, Y. Kawaguchi, M. Nakae, Y. Mizobuchi and T. Shono, *J. Chromatogr.*, 299 (1984) 341.
13 M. Tanaka, Y. Kawaguchi, T. Niinae and T. Shono, *J. Chromatogr.*, 314 (1984) 193.
14 M. Tanaka, Y. Kawaguchi, T. Shono, M. Uebori and Y. Kuge, *J. Chromatogr.*, 301 (1984) 345.
15 D. W. Armstrong, W. DeMond, A. Alak, W. L. Hinze, T. E. Riehl and K. H. Bui, *Anal. Chem.*, 57 (1985) 234.
16 W. L. Hinze, T. E. Riehl, D. W. Armstrong, W. DeMond, A. Alak and T. Ward, *Anal. Chem.*, 57 (1985) 237.
17 D. Sybilska, J. Lipkowski and J. Wójcikowski, *J. Chromatogr.*, 253 (1982) 95.
18 T. Sakai, Y. Niinuma, S. Yanagihara and K. Ushio, *J. Chromatogr.*, 276 (1983) 182.
19 Y. Nobuhara, S. Hirano and Y. Nakanishi, *J. Chromatogr.*, 258 (1983) 276.
20 E. Smolková-Keulemansová, H. Králová, S. Krýsl and L. Feltl, *J. Chromatogr.*, 241 (1982) 3.
21 J. Mráz, L. Feltl and E. Smolková-Keulemansová, *J. Chromatogr.*, 286 (1984) 17.
22 M. Tanaka, S. Kawano and T. Shono, *Fresenius' Z. Anal. Chem.*, 316 (1983) 54.
23 D. Sybilska and T. Kościelski, *J. Chromatogr.*, 261 (1983) 357.
24 T. Kościelski, D. Sybilska, L. Feltl and E. Smolková-Keulemansová, *J. Chromatogr.*, 286 (1984) 23.
25 S. Krýsl, *Ph.D. Thesis*, Charles University, Prague, 1984.
26 S. Krýsl, *Method of Trapping of Volatile Substances, Czech. Pat. Appl.*, PV 05254-84, 1984.
27 R. C. Chang and I. S. Fritz, *Talanta*, 25 (1978) 659.
28 G. A. Junk, J. J. Richard, M. D. Grieser, D. Witiak, M. D. Arguello, R. Vick, H. I. Svec, J. Fritz and G. V. Calder, *J. Chromatogr.*, 99 (1974) 745.