

Studies on the Behavior of Some Phosphorus-Contained Calix[*n*]arenes in Reversed-Phase High-Performance Liquid Chromatography

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Abstract

Reversed-phase high-performance liquid chromatography is applied to studies of the chromatographic characteristics of calix[4]-, calix[6]-, and calix[8]arenes functionalized at the lower rim of the macrocycle by phosphoryl groups. Acetonitrile–water is used as the mobile phase, LiChrosorb 18 is the stationary phase, and ultraviolet detection is at 254 nm. The influence of the size and conformation of the calix[*n*]arene macrocyclic skeleton and the nature of substituents at the lower and upper rims on chromatographic characteristics of calix[*n*]arenes are determined. Retention times and capacity factors of calix[*n*]arenes are determined. It is found that the retention times of calix[*n*]arenes are determined mainly by the presence of hydrophobic *tert*-butyl groups at the upper rim of the macrocyclic ring.

Introduction

Calix[*n*]arenes are macrocyclic molecules made up of phenol units linked via methylene groups (1,2). Due to their cup-shaped architecture formed by ordered, oriented, substituted benzene rings, calix[*n*]arenes can include small guest molecules of an appropriate size and configuration (Figure 1) in the cavity and retain them through different physical interactions (van der Waals forces, CH- π and solvophobic interactions, hydrogen bonds, etc.). The findings of Gutsche (1), Ungaro (3), Shinkai (4), Reinhoudt (5), and Bohmer (6) et al. suggested that calixarenes could be regarded as a third generation of supramolecular hosts (7) after cyclodextrins (8) and crown ethers (9). The binding properties of calixarenes may be improved by the introduction of different functional groups (1) including phosphorus-contained groups at the lower or upper rim of the macrocyclic skeleton (10–18). These groups can increase the cavity size and serve as the additional complexing center for guest molecules.

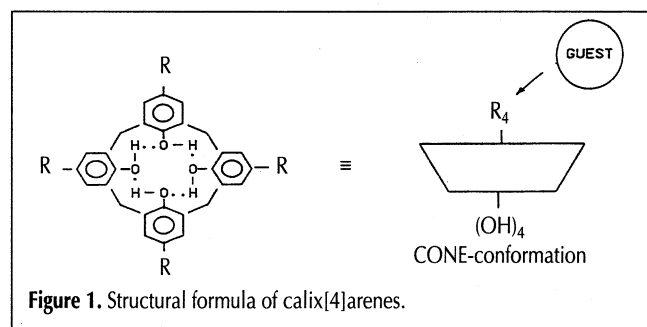
In this study, we reported the investigation of the behavior of

calix[4]-, calix[6]-, and calix[8]arenes functionalized at the lower rim of the macrocycle with phosphoryl groups in reversed-phase high-performance liquid chromatography (HPLC). Knowledge of their behavior would be useful in analyzing their quality in manufacturing as well as in developing methods to modify the sorbent surface in the design of new chromatographic materials.

Experimental

Reagents

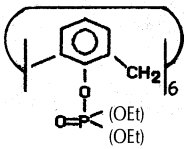
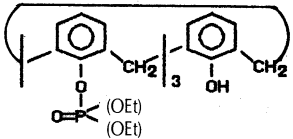
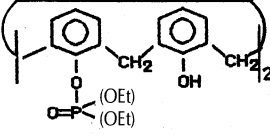
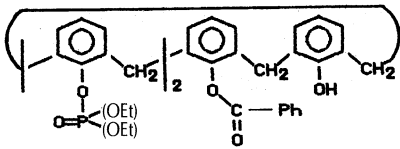
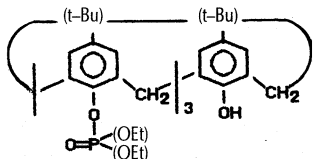
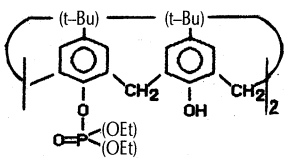
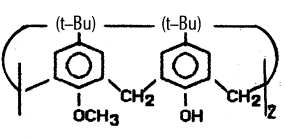
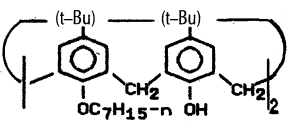
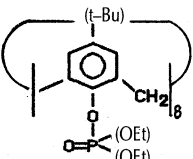
All solvents were of analytical grade and were used without purification. Compound 1 was prepared according to the methods described by Markovsky, Kalchenko, Visotsky, Simonov, Dvorkin, Jatchenko, and Lipkowski (Synthesis and structure of hexa[diethoxy-phosphoryloxy]calix[6]arene. *Supramolecular Chemistry*, in press). Compounds 2, 4, and 5 were prepared as described in reference 19, and compounds 3, 6, 7, and 9 were prepared as described in references 20, 21, 22, and 13, respectively. Compound 8 was prepared as follows: 0.5 g (0.694 mmol) of *tert*-butylcalix[4]arene, toluene complex, and 0.96 g (6.94 mmol) of K_2CO_3 were added to a solution of 0.358 g (2.08 mmol) of *n*-heptyl bromide in 50 mL of acetonitrile and refluxed for 20 h. Then the salt was filtered, and the solution was evaporated to dryness at low pressure. Recrystallization from a methanol–water mixture (80%, 10 mL) gave 0.33 g (52%) of white powder; the melting point was 132–134°C.



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Table I. Retention Time (t_R), Capacity Factor (k'), and Peak-Width-to-Height Ratio (S/h) of Calixarenes

Compound no.	Formula	t_R	k'	S/h
1		6.57	2.29	0.04
2		6.67	2.34	0.04
3		7.33	2.67	0.02
4		13.33	5.67	0.02
5		52.22	25.11	3.67
6		57.33	27.67	6.37
7		84.33	41.17	11.42
8		101.67	49.84	20.0
9			not eluted	

Elemental analysis provided the following results: C 82.46%; H 9.98%; calculated for $C_{58}H_{84}O_4$ C 82.41%; H 10.02%. The nuclear magnetic resonance (1H NMR) spectrum was as follows: $[(CD_3)_2C=O, HMDS, 300\text{ Mhz}]$, δ , ppm: 0.87 [t, $^3J_{HH} = 4.8\text{ Hz}$, 6H, $CH_3(CH_2)_6O$], 0.98 and 1.18 [s, 36 H, $C(CH_3)_3$]; 1.3–1.5 [m, 12 H, $CH_3(CH_2)_3O$], 1.73 [q, $^3J_{HH} = 5\text{ Hz}$, HH, $CH_3(CH_2)_3CH_2CH_2CH_2O$], 2.03 [q, $^3J_{HH} = 5.1\text{ Hz}$, 4H, $CH_3(CH_2)_4CH_2CH_2O$], 3.37 and 4.26 [dd, $^2J_{HH} = 12.6\text{ Hz}$, 8H, $ArCH_2Ar$], 3.94 [t, $^3J_{HH} = 6.6\text{ Hz}$, 4H, $CH_3(CH_2)_5CH_2O$], 7.08 and 7.12 (s, 8H, CH_{arom}), 8.5 (s, 2H, OH).

HPLC analysis

Analysis conditions for the calixarenes were as follows. A type 333 HPLC unit (Institute of Physical Chemistry, Polish Academy of Sciences, Warsaw, Poland) equipped with a 10^{-4} -mL detection cell was used. The column (250×1 -mm i.d.) was packed with LiChrosorb RP 18 (Merck, Darmstadt, Germany) by the viscosity method. The mobile phase consisted of MeCN– H_2O in a ratio of 86:14 (v/v). Ultraviolet (UV) detection was used at 254 nm. The temperature was 26°C , and the flow rate was $0.04\text{ mL}\cdot\text{min}^{-1}$.

Results and Discussion

The objects of this investigation were a number of calix[n]arenes ($n = 4, 6, \text{ and } 8$) possessing two, three, four, six, and eight phosphoryl groups and two alkyl groups at the lower rim of the macrocycle (compounds 1–9 in Table I). According to NMR and x-ray data, compounds 2–8 existed in stereochemically rigid cone (1) conformations in which all benzene rings are oriented upward and all phosphoryl (or alkyl) groups are oriented downward from the main plane of the molecule formed by four methylene links in the macrocyclic skeleton. The conformation of compound 1 (calix[6]arene hexaphosphate) is more complicated. According to the x-ray structural analysis data (L.N. Markovsky, V.I. Kalchenko, M.A. Visotsky, J.A. Simonov, A.A. Dvorkin, A.V. Jatchenko, and J. Lipkowski. Synthesis and structure of hexa[diethoxy-phosphoryloxy] calix[6]arene. *Supramolecular Chemistry*, in press), the molecule of compound 1 exists in the centrosymmetrical flattened 1,2,3-alternate (1) conformation. The diametrically opposite benzene rings are parallel. Their dihedral angles with the main plane of the macrocycle are 61.7° , 38.7° , and 108.4° . In this conformation, four phosphoryl groups are connected with benzene rings and are oriented away from the cycle; two other groups are inside it, blocking the macrocyclic cavity from both sides. In contrast to conformationally rigid calix[4]arenes, hexaphosphorylated calix[6]arene molecules are flexible. In solution, free internal rotation of aromatic rings around $Ar-CH_2-Ar$ bonds is observed. The conformation of compound 9 (*tert*-butyl calix[8]arene octaphosphate) is unknown in detail. According to 1H NMR spectral data (13), fast rotation of aromatic rings through the macrocyclic annulus takes place. The retention times and capacity factors of compounds 1–9 were determined and are presented in Table I in increasing order. Compound 1, which possessed alternatively three upward-

oriented and three downward-oriented benzene rings where the molecular cavity is occupied by two ethoxy groups, had the shortest retention time. A similar retention time manifested in di- and tri-diethoxyphosphoryl calix[4]arenes (compounds 2 and 3). This finding demonstrates that polar diethoxyphosphoryl groups at the lower rim play an insignificant role in the molecular adsorption on the sorbent surface covered by octadecyl groups.

Adsorption increased as a result of the exchange of one phosphoryl group of compound 2 to the benzoyl group. The retention time of compound 4 (diphosphorylbenzoyl calix[4]arene) was twice as high as that of compound 2 (triphosphoryl calix[4]arene). This result indicates the significant role of $CH-\pi$ interaction between aromatic rings of the benzoyl group and octadecyl chains of the sorbent surface in the adsorption process. Among the investigated substituents, hydrophobic *tert*-butyl groups at the upper rim of the macrocycle mainly influenced the interaction forces.

Thus, the introduction of four *tert*-butyl groups in compound 2 influenced hydrophobic interactions with the sorbent surface and resulted in increased retention time from 6.67 to 52.22 min (see compound 5 in Table I). It is interesting that introduction at the lower rim of two hydrophilic dihydroxyphosphoryl groups (compound 6) or two hydrophobic alkyl groups (compounds 7 and 8) instead of the diethoxyphosphoryl groups of compound 5 led to increased adsorption. Perhaps the strong protonodative dihydroxyphosphoryl groups formed hydrogen bonds with oxygen atoms of free and octadecylated groups of LiChrosorb RP 18 matrices. This suggestion was confirmed by the strong peak broadening of compound 6 (the peak-width-to-height ratio is 6.37, see Table I). The increased retention time of compounds 7 and 8 could be explained by the additional participation of two preorganized methyl or heptyl groups in hydrophobic interactions with LiChrosorb RP 18 octadecyl chains. This interaction was stronger because the alkyl chain was longer. *Tert*-butyl calix[8]arene octaphosphate (compound 9) retention was the strongest among the investigated substances. Compound 9 was adsorbed irreversibly with LiChrosorb RP 18, and peak 9 was not observed on the chromatogram, even after 120 min. This finding demonstrates the potential of using calixarenes for the modification of alkylated silicagels and the production of new sorbents.

Conclusion

The sorption of calix[n]arenes ($n = 4, 6, \text{ and } 8$) functionalized at the lower rim of the macrocycle with phosphoryl groups by LiChrosorb RP 18 under reversed-phase HPLC conditions was determined mainly by the presence of *tert*-butyl groups at the upper rim due to their hydrophobic interactions with octadecyl groups on the sorbent surface. Polar phosphoryl groups played an insignificant role in the adsorption process.

Acknowledgments

This work is the result of a joint cooperation between the Institute of Physical Chemistry at the Polish Academy of

Sciences and the Institute of Organic Chemistry at the National Academy of Sciences of Ukraine. M.A. Visotsky thanks the International Soros Education Program for grant PSU053020. L.N. Markovsky and V.I. Kalchenko thank INTAS for partial financial support of this work through grant 94/1914.

References

1. C.D. Gutsche. *Calixarenes*, Royal Society of Chemistry, Cambridge, MA 1989.
2. *Calixarenes: A Versatile Class of Macrocyclic Compounds*, J. Vicens, V. Bohmer, Eds., Kluwer, Dordrecht, 1991.
3. R. Ungaro, A. Casnati, F. Ugozzoli, A. Pochini, J.-E. Dozol, C. Hill, and H. Roquette. 1,3-Dialkoxycalix[4]arenecrowns-6 in 1,3-alternate conformation: Cesium-selective ligands that exploit cation-arene interactions. *Angew. Chem., Int. Ed. Engl.* **33**: 1506–509 (1994).
4. S. Shinkai. Calixarenes: The third generation of supramolecules. *Tetrahedron* **49**: 8933–68 (1993).
5. P. Timmerman, W. Verboom, F.-C.I.M. van Veggel, J.P.M. van Duyhoven, and D.N. Reinhoudt. A novel type of stereoisomerism in calix[4]arene-based carceplexes. *Angew. Chem., Int. Ed. Engl.* **33**: 2345–48 (1994).
6. V. Bohmer. Calixarenes. Macrocycles with (almost) unlimited possibilities. *Angew. Chem., Int. Ed. Engl.* **34**: 713–45 (1995).
7. J.M. Lehn. Supramolecular chemistry: Receptors, catalysts, and carriers. *Science* **227**: 849–56 (1985).
8. G. Wenz. Cyclodextrins as building blocks for supramolecular structures and functional units. *Angew. Chem., Int. Ed. Engl.* **33**: 803–22 (1994).
9. J. An, J.S. Bradshaw, and R.M. Izatt. Macropolycyclic polyethers (cages) and related compounds. *Chem. Rev.* **92**: 543–72 (1992).
10. M. Almi, A. Arduini, A. Casnati, A. Pochini, and R. Ungaro. Chloromethylation of calixarenes and synthesis of new water soluble macrocyclic hosts. *Tetrahedron* **45**: 2177–82 (1989).
11. T. Arimura, T. Nagasaki, S. Shinkai, and T. Matsuda. Host-guest properties of new water soluble calixarenes derived from *p*-(chloromethyl)calixarenes. *J. Org. Chem.* **54**: 3766–68 (1989).
12. C. Floriani, D. Jacoby, A. Chiest-Vilia, and C. Guastini. Aggregation of metal ions with functionalized calixarenes: Copper (I) chloride complex. *Angew. Chem.* **101**: 1430–31 (1989).
13. Z. Goren and S.-E. Biali. Multiple reductive cleavage of calixarene diethyl phosphate esters: A route to [1ⁿ] metacyclophanes. *J. Chem. Soc. Perkin Trans. 1*: 1484–87 (1990).
14. F. Grunszpan, Z. Goren, and S.-E. Biali. Partially OH depleted calix[4]arenes. *J. Org. Chem.* **56**: 532–36 (1991).
15. J. Ting, W. Verboom, L.C. Groenen, J.-D. van Zoom, and D.N. Reinhoudt. Selectivity dextroxylylated calix[4]arenes and 1,3-dithiocalix[4]arenes; novel classes of calix[4]arenes. *J. Chem. Soc. Chem. Commun.* 1432–33 (1990).
16. R.G. Janssen, W. Verboom, S. Harkema, G.J. Hummel, D.N. Reinhoudt, A. Pochini, R. Ungaro, P. Prados, and J. Mendoza. Selective phosphorylation of *p*-tert-butylcalix[6]arene. *J. Chem. Soc. Chem. Commun.* 506–508 (1993).
17. L.N. Markovsky, V.I. Kalchenko, and N.A. Parchomenko. *O*-Phosphorylated *p*-tert-butylcalixarenes. *Zh. Obshch. Khim.* **60**: 2811–12 (1990).
18. V.I. Kalchenko, L.I. Atamas, V.V. Pirozhenko, L.N. Markovsky. Phosphorylation of tetramethyl ether 5,11,17,23-tetrabromocalix[4]arene. *Zh. Obshch. Khim.* **62**: 2623–24 (1992).
19. L.N. Markovsky, M.A. Visotsky, V.I. Kalchenko, V.V. Pirozhenko, J. Lipkowski, and J.A. Simonov. Phosphorotropic rearrangement in synthesis of asymmetrically substituted calix[4]arenes. *J. Chem. Soc. Chem. Commun.* 69–71 (1996).
20. V.I. Kalchenko, J. Lipkowski, J.A. Simonov, M.A. Visotsky, K. Suwinska, A.A. Dvorkin, V.V. Pirozhenko, I.F. Tsimbal, and L.N. Markovsky. Stereochemistry of diphosphorylated calix[4]arenes. *Zh. Obshch. Khim.* **65**: 1311–20 (1995).
21. V.I. Kalchenko, M.A. Visotsky, V.V. Pirozhenko, A.N. Shivanyuk, and L.N. Markovsky. Dihydroxyphosphorylated derivatives of calix[4]arenes. *Zh. Obshch. Khim.* **64**: 1560–61 (1994).
22. P.S. Dijkstra, A.I. Brinvuk, K.-E. Bugge, D.N. Reinhoudt, S. Harkema, R. Ungaro, F. Ugozzoli, and E. Ghidini. Kinetically stable complexes of alkali cations with rigidified calix[4]arenes: Synthesis, x-ray structures and complexation of calixarenes and calixpherands. *J. Am. Chem. Soc.* **111**: 7567–75 (1989).

Manuscript accepted July 31, 1996.