Journal of Inclusion Phenomena and Molecular Recognition in Chemistry **10**: 69–78, 1991.

Complexes of β-Cyclodextrin with Chloronitrobenzenes and with Solvents in Water + Organic Solvent Mixtures

J. TARASZEWSKA
Institute of Physical Chemistry, Polish Academy of Sciences, 01-224 Warsaw, Poland

(Received 29 January 1990; in final form: 2 July 1990)

Abstract. Inclusion complexes of chloronitrobenzenes with β-cyclodextrin in aqueous 0.1 M phosphate buffer solutions containing various concentrations of ethanol, dimethylsulfoxide, dimethylformamide, acetone and acetonitrile have been studied by a polarographic method. The diffusion coefficients and the stability constants of the corresponding complexes have been determined. Using an equation derived by us which takes account of the change in the cyclodextrin concentration due to the simultaneous complexation of the solvent, both stability constants have been calculated. The influence of solvent on the stability constant of chloronitrobenzenes is discussed

Key words. Cyclodextrin, stability constants, chloronitrobenzenes, organic solvents.

1. Introduction

Cyclodextrins (cycloamyloses) are known to form inclusion complexes with a variety of substances in the solid state as well as in solution.

For a long time it was generally believed that cyclodextrins (CD) form inclusion complexes only in pure aqueous solutions [1, 2] mainly because attempts to induce precipitation of CD adducts from organic solvents failed.

The first successful work concerning the influence of solvent on the formation of inclusion complexes with CD was that of Siegel and Breslow [3] who determined the stability constants of several organic species with β-CD in dimethylformamide (DMF), dimethylsulfoxide (DMSO) and in H_2O + DMSO mixtures. On the basis of this paper, as well as from data of other authors [4–6], one can conclude that: (1) the solvent plays a key role in the complexation process; (2) the complexation in non aqueous and water + organic solvent mixtures is weaker than in pure aqueous solutions.

However, recently Nelson *et al.* [7] have reported that the stability constants of pyrene complexes with β- and γ-CD in the presence of 10% *t*-butanol were an order of magnitude greater than the values obtained in pure aqueous solutions.

Molecules of organic solvents can also be complexed by CD, as has been found for alcohols [8–10]. Complexation of other solvents with α-CD has also been reported [11].

The aim of this work was to study the complexation of isomeric chloronitrobenzenes (ClNB) by β-CD in aqueous solutions of 0.1 M phosphate buffer containing various concentrations of such solvents as ethanol (EtOH), acetonitrile (ACN), acetone (AC), DMF and DMSO.

Investigation of the interactions with CD of a second substrate featuring widely varying chemical and structural properties may offer a useful insight into the binding characteristics of CD.

These studies may also be useful from a practical point of view because the presence of an organic solvent is often inevitable in the complexation of drugs which are very poorly soluble in pure water.

The complexation of isomeric ClNB by α- and β-CD in pure aqueous solutions has been studied by us previously [12] using a polarographic method.

2. Experimental

The reduction of isomeric ClNB at the Hg electrode from H_2O + organic solvent mixtures containing 0.1 M phosphate buffer (pH = 7) in the absence and in the presence of increasing concentrations of β-CD was studied by a polarographic method. All experiments were carried out in a three-electrode system at 25 ± 0.5°C. The counter electrode was a Pt cylinder. A 1 M NaCl aqueous calomel electrode (NCE) was used as the reference electrode. It was connected to the electrode cell via an intermediate vessel filled with the solution under investigation. Solutions were deaerated by flushing with pure and dried argon presaturated with the solution under investigation; during measurements the gas was passed over the solution. Since ClNB is very volatile, the time of deaeration was strictly controlled and two wash bottles with the solution under investigation were placed upstream of the measuring cell itself.

The polarographic curves were recorded using a measuring system constructed from an EP-20A potentiostat, an EG-20 function generator (both produced by ELPAN, Poland) and an XY/t-102 recorder (ZDEMP, Poland).

β-CD (Chinoin, Hungary) was used as received. Thermogravimetric analysis showed that it contained 11 wt.% H_2O. The *o*- *m*- and *p*-isomers of ClNB as well as Na_2HPO_4 and KH_2PO_4, used for the preparation of the phosphate buffer, were p. a. grade reagents (POCh, Poland). The concentration of ClNB in all experiments was 1.25×10^{-4} M and the concentration of β-CD was varied from about 3 mM to more than 14 mM. The concentration of CD was always in great excess compared to that of the electroactive substrate, in order that equilibrium should be achieved in the whole diffusion layer. Organic solvents were dried and purified for electrochemical use [13]. The purity of solvents was checked by gas chromatography. The amount of the organic contaminants was less than 0.2%. Triply distilled H_2O was used for preparation of the solutions.

3. Results

Electroreduction of isomeric ClNB at the Hg electrode in H_2O + organic solvent mixtures studied is similar to that in pure aqueous solutions. It gives a single, well-defined irreversible diffusion wave at a potential of about -0.4 V, which corresponds to a four-electron reduction of ClNB to chlorophenylhydroxylamine.

In order to determine the stability constants of ClNB with β-CD we applied a polarographic method based on the determination of diffusion coefficients of the

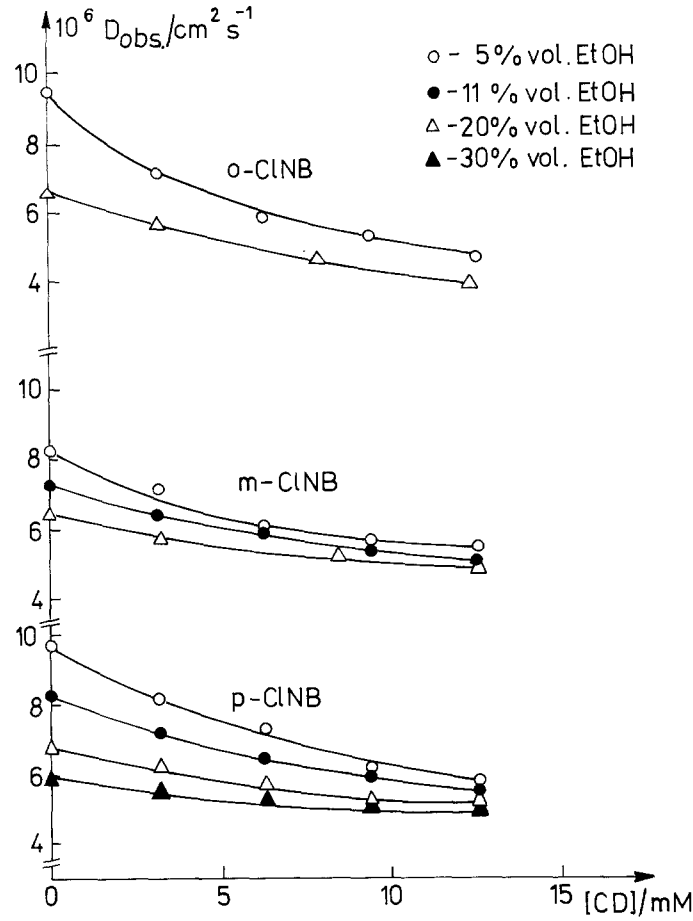

Fig. 1. Changes in the observed diffusion coefficients D_{obs} of isomeric ClNB in H_2O + EtOH mixtures of 0.1 M phosphate buffer vs. β-CD concentration.

free substrate D_f and of the observed diffusion coefficients D_{obs} in the presence of increasing concentrations of β-CD. All diffusion coefficients were calculated from the polarographic limiting instantaneous diffusion currents using the Ilkovic equation corrected for spherical diffusion by Koutecky [14]. The changes in D_{obs} vs. β-CD concentration for isomeric ClNB in H_2O + EtOH mixtures are shown in Figure 1, and for o − ClNB in H_2O + DMSO, H_2O + DMF, H_2O + AC and H_2O + ACN in Figure 2.

The method by which the stability constants K_s were calculated has been described by Osa *et al.* [15] and by us [12] previously. When a complex of 1 : 1 stoichiometry is formed, as has been assumed for the complex of ClBN with β-CD on the basis of the data of Harata [16] for other disubstituted benzenes, the values of K_s and D_c can be calculated by a linear regression analysis using

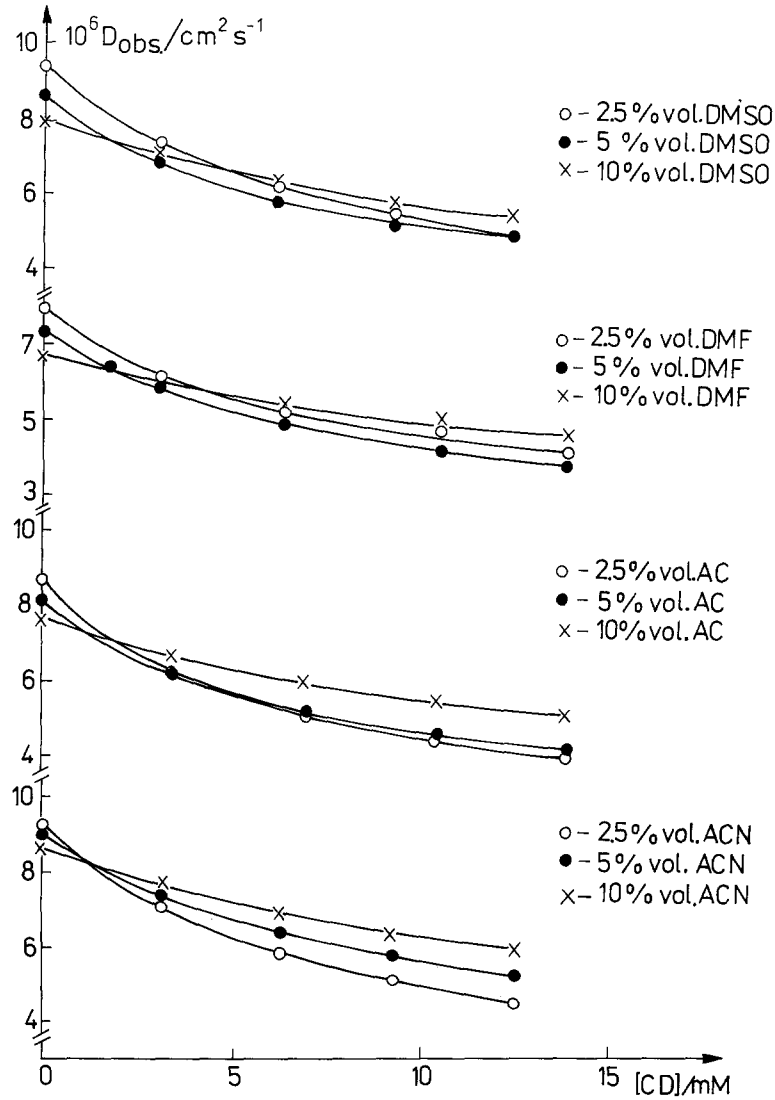

Fig. 2. Changes in the observed diffusion coefficients D_{obs} of *o*-ClNB in H_2O + DMSO, H_2O + DMF, H_2O + AC and H_2O + ACN mixtures of 0.1 M phosphate buffer vs. β-CD concentration.

Equation (1)

$$D_{obs} = \frac{(D_f - D_{obs})}{K_s \times C_{CD}} + D_c \tag{1}$$

The values of K_s, D_c and D_f can be also calculated by a nonlinear iterative minimization of the sum of squares of the deviation from the true line

$$\sum_{i=1}^{N} (D_i^{obs} - D_i^{calc})^2 \tag{2}$$

where N is the number of points in one set of data and

$$D_i^{\text{calc}} = \frac{D_f^* + D_c^* K_s^* C_{CD}}{1 + K_s^* C_{CD}} \tag{3}$$

where K_s^*, D_c^* and D_f^* are the assumed values.

The stability constants K_s of isomeric ClNB with β-CD in H_2O + EtOH mixtures containing 0.1 M phosphate buffer are presented in Figure 3. The stability constants K_s of *o*-ClNB with β-CD in H_2O + EtOH, H_2O + DMSO, H_2O + DMF, H_2O + AC and H_2O + ACN mixtures in 0.1 M phosphate buffer are shown in Figure 4.

The results presented show that increasing the concentration of the organic solvent in the mixture results in the complexation of ClNB by β-CD becoming weaker. The values of K_s for isomeric ClNB with β-CD obtained in H_2O + 20% vol. EtOH mixtures are in good agreement with those reported by Żukowski *et al.* [17] determined by reversed-phase high-performance liquid chromatography.

The calculated values of D_c for *o*-ClNB with β-CD are in the limits 1.8–2.5 × 10^{-6} cm^2 s^{-1} depending on the composition of the mixture.

4. Discussion

In the derivation of Equations (1) and (3) it was assumed that the concentration of free CD is equal to its analytical concentration.

This assumption, which is valid in the case of one guest molecule species present in solution at a much lower concentration than the concentration of CD, may not be true in the presence of a second guest species which can form weak complexes with CD but which is present in solution at a much higher concentration than the concentration of the first guest.

Therefore: (1) taking account of two independent equilibria due to the complexation of guest 1 (ClNB) and guest 2 (solvent) with CD and (2) assuming that the concentration of CD connected with guest 1 is negligible in comparison with the concentration of CD bound to guest 2, we derived [18] the following equation for D_{obs}.

$$D_{obs} = \frac{K_s^{(1)}\{(D_c + D_f)[1 + K_s^{(2)}(C_2 - C_{CD})] - 2D_c K_s^{(1)} C_{CD} + (D_f - D_c) \times \sqrt{(1 + C_{CD} + K_s^{(2)})^2 + C_2 K_s^{(2)}[2 + K_s^{(2)}(C_2 - 2C_{CD})]}\} - 2D_f K_s^{(2)}}{2[(C_{CD} K_s^{(1)} + 1)(K_s^{(1)} - K_s^{(2)}) + K_s^{(1)} K_s^{(2)} C_2]} \tag{4}$$

where C_1, C_2, C_{CD} are the analytical concentrations of guest 1, guest 2, and CD, respectively; D_f and D_c are the diffusion coefficients of free guest 1 and the complex of guest 1 with CD, respectively; and $K_s^{(1)}$ and $K_s^{(2)}$ denote the stability constants of guest 1 and guest 2 with CD, respectively.

Using Equation (4) the stability constants of ClNB with β-CD and the stability constants of solvents with β-CD have been calculated by a computer program. The results of the calculations are reported in Tables I and II.

The results presented show that in all the H_2O + organic solvent mixtures studied the value of K_s of ClNB with β-CD does not change with the composition of the mixture. The decrease of K_s presented in Figures 3 and 4 was due to the fact that the change in the concentration of free CD with increasing content of the organic solvent in the mixture was not taken into account.

Table I. Stability constants of *o*-ClNB with β-CD in H_2O + organic solvent mixtures

solvent	$K_s(M^{-1})$
H_2O + EtOH	227 ± 20
H_2O + DMSO	227 ± 20
H_2O + DMF	227 ± 20
H_2O + AC	350 ± 50
H_2O + ACN	500 + 50

Table II. Stability constants of organic solvents with β-CD

solvent	$K_s(M^{-1})$
EtOH	0.5 ± 0.2
DMSO	1.3 ± 0.2
DMF	2.2 ± 0.3
AC	5.0 ± 0.5
ACN	6.0 + 0.5

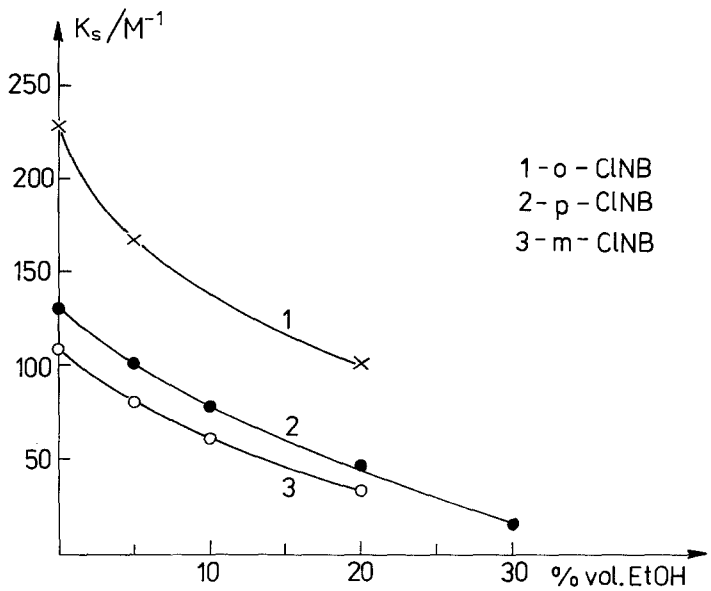

Fig. 3. Changes in the stability constants K_s of isomeric ClNB with β-CD vs. EtOH content in the mixture.

The complexes of solvents with β-CD are weak. The value of K_s for the EtOH complex is in good agreement to that reported in the literature [8] determined by a spectrophotometric technique. The comparison of the values of K_s for DMSO and ACN with the corresponding values for the complexes with α-CD [11] shows that the stability of the complex of β-CD with DMSO is stronger and with ACN is similar to that with α-CD, respectively.

However, the complexes of the studied solvents with α- and β-CD are weak, neglecting their formation may be responsible for the erroneous calculation of other constants. In particular, a solvent such as ACN has been employed as a solvating agent in many experiments designed to monitor the catalytic efficiency of CD in ester hydrolysis reactions [15, 20]. Being unaware of the ACN complexation the calculated catalytic rate enhancements were no doubt underestimated.

There are some data in the literature [21–23] concerning the crystal structures of α-CD complexes with DMSO and DMF as well as the β-CD complex with EtOH.

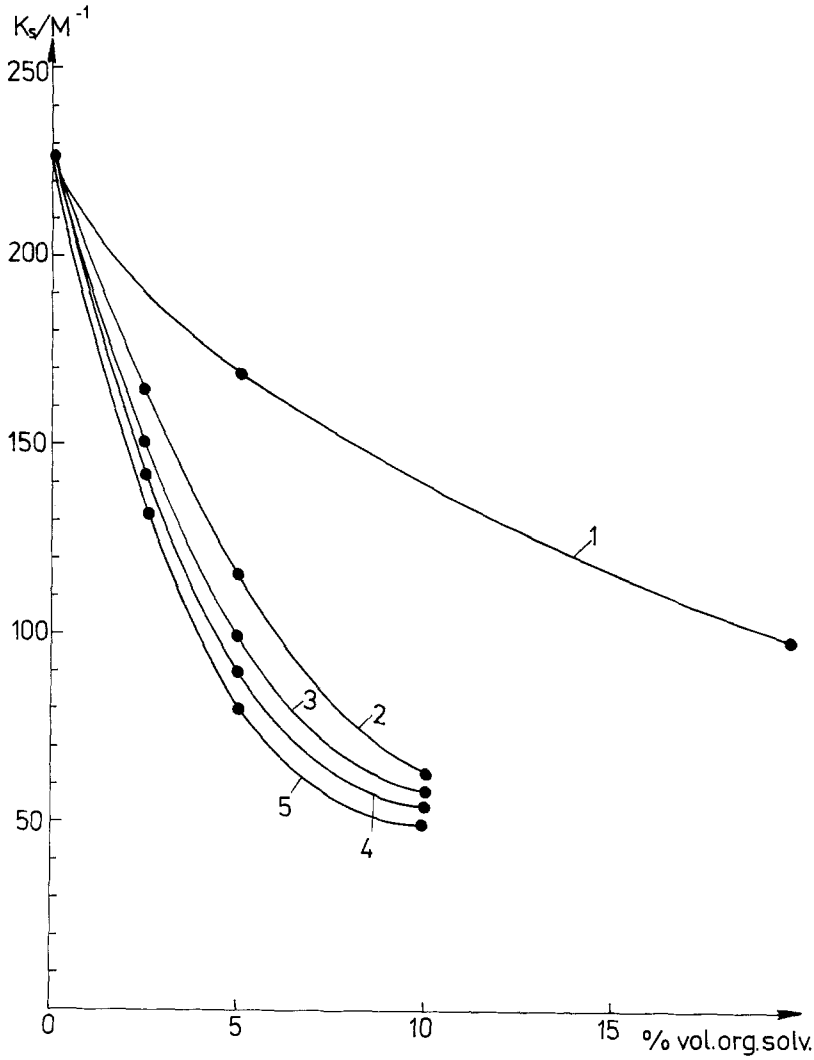

Fig. 4. Changes in the stability constants K_s of *o*-ClNB with β-CD vs. organic solvent content in the mixture; Curves: (1) in H_2O + EtOH, (2) in H_2O + DMSO, (3) in H_2O + DMF, (4) in H_2O + AC, (5) in H_2O + ACN.

The X-ray study of α-CD crystals obtained from a DMSO–methanol solution has shown that the α-CD molecule includes DMSO and methanol molecules simultaneously. The DMSO molecule is located on the secondary hydroxyl side in the α-CD cavity. The molecule of DMF is also incorporated in the cavity of α-CD. The DMF molecule is so small that the vacant place is occupied by H_2O. Three H_2O molecules are located near the guest molecule, while the other two water molecules fill the intermolecular space. The main forces responsible for the complexation are van der Waals interactions. In the case of the EtOH complex with β-CD three H_2O molecules and one EtOH molecule are included within the β-CD cavity. One can suppose that a similar picture exists in the solution.

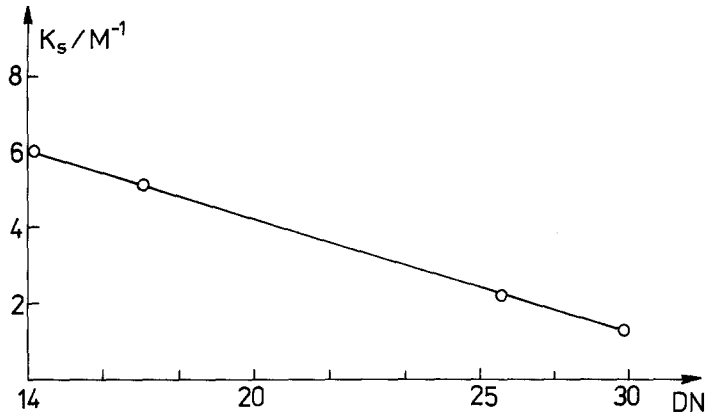

Fig. 5. Dependence of the stability constants K_s of complexes of β-CD with aprotic solvents on the donor number (DN) of the solvent.

The fact that the K_s values of solvents with β-CD, similar to the case of complexes with α-CD, are independent of the solvent concentration in the mixture is particularly significant. It implies that the bonding mechanism cannot involve H_2O molecules in any significant way. This means that suggested mechanisms such as the release of H_2O from the CD cavity and the hydrophobicity of CD cannot play an important role here. We found a linear dependence of the stability constants of complexes of β-CD with aprotic solvents on the donor number (DN) of the solvent (Figure 5).

As has been shown in Table I the K_s values for the complex of o-ClNB with β-CD in H_2O + EtOH, H_2O + DMSO and H_2O + DMF mixtures were independent of the composition of the mixture and equal to the value in pure aqueous solution. However, in H_2O + AC and H_2O + ACN an increase in the K_s values was observed.

In order to clarify this result we took account of the difference in the structure of the pure solvents as well as the difference in the thermodynamic properties of the solvent mixtures. Bastiansen and Viervoll [24] comparing the structure of AC and DMSO by the electron diffraction sector method found a planar structure for the AC molecule and a pyramidal structure for the DMSO molecule. The thermodynamic properties of mixtures of these solvents with H_2O also vary from H_2O + AC showing large positive deviations from ideality to H_2O + DMSO showing large negative deviations from ideality. In H_2O + DMSO and H_2O + DMF mixtures strong mutual interactions between the molecules of both solvents [25–28] also exist.

In view of the above considerations one can assume that there are some differences in the complexation of AC and probably of ACN in comparison with the complexation of DMSO and DMF by CD. In the presence of AC and ACN the part of the interior of the β-CD cavity in which o-ClNB is located can be more hydrophobic than in the presence of DMSO and DMF and this can be the reason for the higher K_s values of o-ClNB with β-CD in these solvents. However, to get a better insight into the complexation process further studies, especially with substi-

tuted CDs could be very useful. Such studies are the subject of further investigation by us.

5. Conclusions

On the basis of this work one can draw the following conclusions: (1) the presence of the organic solvent up to 20% vol. in the case of EtOH and up to 10% vol. in the case of DMSO, DMF, AC and ACN does not cause any decrease in the K_s value of ClNB with β-CD. The decrease of K_s was due to the fact that the change in the concentration of free CD with the increase in the content of the organic solvent in the mixture was not taken into account. The presence of the organic solvent can also cause an increase of K_s of the other substrate. (2) The polarographic method is a convenient tool for the determination of the stability constants of inclusion complexes of solvents with CD. (3) The stability constants of the solvents studied with β-CD are weak and similar to the case of α-CD.

Acknowledgement

I wish to express my gratitude to Mr. J. Śledzik for his help in the experimental part of this work. This work has been carried out within the framework of Research Project WPR-6/III of the Polish Academy of Sciences.

References

1. J. L. Lach and Ting-Fong Chin: *J. Pharm. Sci.* **53**, 69 (1964).
2. D. W. Griffiths and M. L. Bender: *Adv. Catal.* **23**, 209 (1973).
3. D. Siegel and R. Breslow: *J. Am. Chem. Soc.* **97**, 6869 (1975).
4. W. V. Gerasimowicz and J. F. Wójcik: *Bioorg. Chem.* **11**, 420 (1982).
5. T. Matsue, U. Akiba, K. Suzufuji and T. Osa: *Denki Kagaku*, **53**, 508 (1985) (in English).
6. A. Harada and S. Takahashi: *Chem. Lett.* 2089 (1984).
7. G. Nelson, G. Patonay and J. M. Warner: *J. Incl. Phenom.* **6**, 277 (1988).
8. Y. Matsui and K. Mochida: *Bull. Chem. Soc. Jpn.* **52**, 2808 (1979).
9. A. Buvari, J. Szejtli and L. Barcza: *J. Incl. Phenom.* **1**, 151 (1983).
10. H. Fujiwara, H. Arakawa, S. Murata and Y. Sasaki: *Bull. Chem. Soc. Jpn.* **60**, 3891 (1987).
11. R. I. Gelb, L. M. Schwartz, M. Radeos, R. E. Edmonds and D. A. Laufer: *J. Am. Chem. Soc.* **104**, 6183 (1982).
12. J. Taraszewska and A. K. Piasecki: *J. Electroanal. Chem.* **226**, 137 (1987).
13. C. K. Mann in A. J. Bard (Ed). *Electroanalytical Chemistry*, Vol. 3, Marcel Dekker, New York. p. 57, 1969.
14. J. Koutecky: *Czech. J. Phys.* **2**, 50 (1953).
15. T. Osa, T. Matsue and M. Fujihira: *Heterocycles* **6**, 1833 (1977).
16. K. Harata: *Bioorg. Chem.* **10**, 255 (1981).
17. J. Żukowski, D. Sybilska and J. Jurczak: *Anal. Chem.* **57**, 2215 (1985).
18. J. Taraszewska and A. K. Piasecki: *Proc. 4th Intern. Symp. Cyclodextrins*, Kluwer Academic Publishers Group, p. 247 (1988).
19. M. Komiyama and M. L. Bender: *J. Am. Chem. Soc.* **100**, 2259 (1978).
20. Y. Kimura and M. L. Bender: *Bioorg. Chem.* **4**, 237 (1975).
21. K. Harata: *Bull. Chem. Soc. Jpn.* **51**, 1644 (1978).
22. K. Harata: *Bull. Chem. Soc. Jpn.* **52**, 2451 (1979).

23. R. Tokuoka, M. Abe, T. Fujiwara, K. Tomita and W. Saenger: *Chem. Lett.* 491 (1980).
24. O. Bastiansen and H. Viervoll: *Acta Chem. Scand.* **2**, 702 (1948).
25. J. M. G. Cowie and P. M. Toporowski: *Can. J. Chem.* **39**, 2240 (1961).
26. G. Ebert and J. Wendorf: *Ber. Bunsenges. Phys. Chem.* **74**, 1071 (1970).
27. E. S. Verstakov, P. S. Yastremskii, Yu. M. Kessler, V. V. Goncharov and V. V. Kokovin: *Zh. Strukt. Khim.* **21**, 91 (1980) C. A. **94**, 91329u (1981).
28. A. I. Mishustin and Yu. M. Kessler: *Zh. Strukt. Khim.* **15**, 205 (1974). C. A. **81**, 30412a (1974).