Chiral Recognition of Zolmitriptan by Modified Cyclodextrins

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The two pyrrolidinylidenesulfamido-modified β-cyclodextrins (β-CDs) **3** and **4** were prepared and studied for chiral discrimination of the enantiomers (*R*)- and (*S*)-**1** of zolmitriptan. The pyrrolidinylidenesulfamido spacer improved the chiral discrimination and binding abilities of these modified cyclodextrins. The hosts **3** and **4** showed higher selectivity for (*S*)-**1**. The association constants (*Table*) and enantioselectivity factors were calculated for the complexes of (*R*)- and (*S*)-**1** with the β-CDs **2**–**4**. The formation of host·guest complexes was confirmed by ^{1}H-NMR studies.

Introduction. – Zolmitriptan (=4-{{3-[2-(dimethylamino)ethyl]-1*H*-indol-5-yl}methyl}oxazolidin-2-one) is a recently approved drug for the acute treatment of migraine and related vascular headaches [1]. It is observed that with (*S*)-zolmitriptan ((*S*)-**1**; *Fig. 1*) being used clinically, the presence of the (*R*)-antipode (*R*)-**1**, even as an impurity, may alter the clinical profile of (*S*)-**1** [2]. All over the world, drug-regulatory authorities increasingly tend to insist on the marketing of single-enantiomer drugs, especially due to the deleterious effects shown by the other enantiomer. Therefore, we made an attempt in the present investigation for the chiral discrimination of zolmitriptan by means of modified cyclodextrins utilizing fluorescence as the signaling tool.

Cyclodextrins (CD) are cyclic oligosaccharides consisting of six (α-CD), seven (β-CD), and eight (γ-CD) 1,4-linked α-D-glucopyranose units. CDs and modified CDs have attracted much attention as aqueous-based hosts for the study of chiral recognition of biological substrates [3]. These CDs act as host molecules by forming inclusion complexes with a wide variety of guests [4]. Inclusion-complex formation occurs as a result of interaction of the hydrophobic cavity of CD with the hydrophobic portion of the guest. Each CD exists as a single enantiomer, and the interaction with a racemic guest may lead to the formation of diastereoisomeric complexes of different thermodynamic stability. The CDs may discriminate the enantiomers of the same guest. However, one can expect an increased chiral discrimination by modification of one of the OH groups of the CDs. Such a modification may induce substantial changes in the asymmetry of the CD resulting in additional and more specific interactions between the

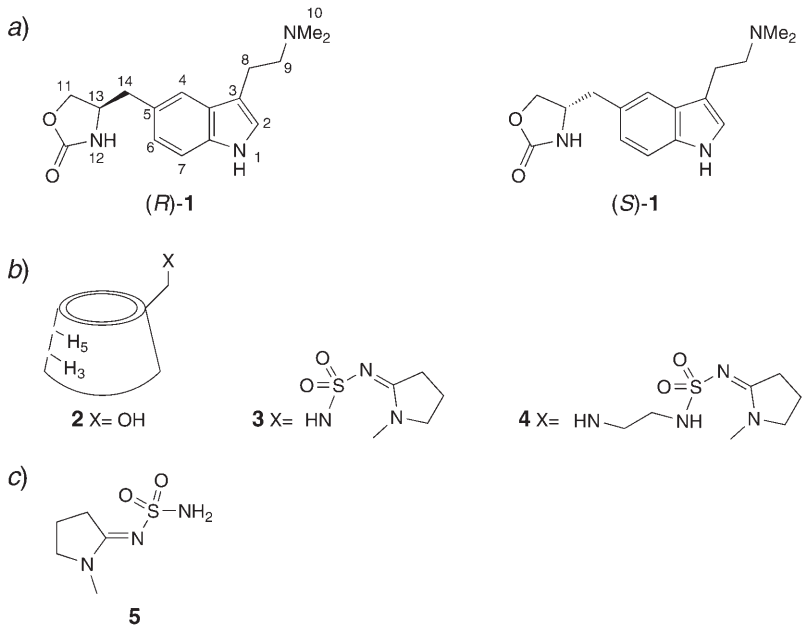
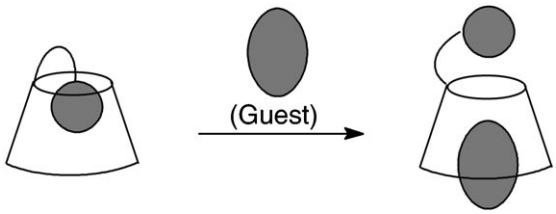


Fig. 1. a) *(*R*)- and (*S*)-Zolmitriptan* ((*R*)- and (*S*)-**1**; arbitrary atom numbering), b) *native β-cyclodextrin* (**2**) *and modified cyclodextrins* **3** *and* **4**, *and* c) *free spacer* **5**

chiral area of the guest and the asymmetry of the host by restricting the geometry of binding leading to greater enantioselectivity [5]. Various modified CDs have been studied for chiral discrimination of the enantiomers of a variety of guests [6]. *Ueno* and co-workers have prepared many kinds of modified cyclodextrins as chemosensors for molecular recognition explaining the mode of self-inclusion and the interaction in the complex formation [7]. These modified CDs, which may also be called artificial receptors, have an appended moiety which sticks down into the hydrophobic cavity of the CD and an 'induced fit' conformational change occurs on inclusion of the guest. The guest displaces the appended moiety from inside to the outside of the cyclodextrin cavity (*Scheme*), a phenomenon that can be studied by observing the change in fluorescence.

Scheme. *Exchange Phenomenon at the CD of the Spacer with the Guest*



In this paper, we wish to report our results on the enantiomer discrimination by the two β-CD derivatives mono{6-deoxy-6-{{[(1-methylpyrrolidin-2-ylidene)amino]sulfonyl}amino}}-β-cyclodextrin (**3**), and mono{6-deoxy-6-{{2-{{[(1-methylpyrrolidin-2-ylidene)amino]sulfonyl}amino}ethyl}amino}}-β-cyclodextrin (**4**) (*Fig. 1*). These two derivatives and native β-cyclodextrin (β-CD; **2**) were selected to investigate their inclusion-complexation behavior with the guest drug (*RS*)-zolmitriptan (**1**) by fluorescence spectroscopy. Previously we have reported **3** as a chemosensor for the molecular recognition of some organic guests [8].

Results and Discussion. – The fluorescence-spectral titrations were performed in 0.1M tetraborate buffer (pH 7.2) at 25°, and the complex association constants (K_s) and enantioselectivity factors (α) for different complexes were calculated. Complexation of the guest with the host were also established by a ^{1}H-NMR study.

In the titration experiments by means of fluorescence spectroscopy, the fluorescence intensity of the guest **1** gradually decreased upon the addition of various concentrations of the hosts **2**, **3**, or **4**. The quenching in fluorescence intensity of the guest was maximum with the host **4** followed by **3** and **2**. *Fig. 2* shows the fluorescence spectra of the guests (*R*)- and (*S*)-**1** in 0.1M tetraborate buffer (pH 7.2) containing various concentrations of **3** and **4**. The hosts **3** and **4** discriminated the enantiomers of the guest **1** by a different quenching of their fluorescence intensity. Thus, the decrease in fluorescence intensity in the presence of the hosts **3** and **4** was higher in the case of (*S*)-**1** than of (*R*)-**1**. Host **2** did not show any selectivity in the fluorescence quenching.

The association constants (K_s) of these host·guest complexes were calculated (*Table*) from the variation of the fluorescence intensities by using the *Benesi–Hildebrand* equation [9]. For a 1:1 complex [guest]·[host], the following expression can be written: $H + G \rightleftharpoons H \cdot G$. The association constant of the complex (K_s) is given by *Eqn. 1*, where [H], [G] and [H·G] are equilibrium concentrations. The association-constant value for the inclusion complex can be determined by the typical double-reciprocal plots of *Eqn. 2*, where F_0 is the fluorescence intensity of the guest alone, F the fluorescence intensity of the complex, and F_{inf} the fluorescence intensity when all the guest molecules are essentially complexed with the host. *Eqn. 2* assumes that only a 1:1 complex is formed. This assumption can be readily tested by using a reciprocal plot (*Fig. 3*) of $\Delta F/F_0$ *vs.* 1/[H].

$$K_s = \frac{[H \cdot G]}{[H][G]} \tag{1}$$

$$1/(F_0 - F) = 1/(F_0 - F_{inf})[H]K_s + 1/(F_0 - F_{inf}) \tag{2}$$

The variation in the association constants for each of the enantiomers of the guest indicates the formation of binary complexes of different stability with the hosts. Host **2** had not discriminated the enantiomers of the drug, but by modifying **2** to **3** and **4**, a good chiral discrimination was observed, and it could be explained by calculating the enantioselectivity factor α [6g] (*Eqn. 3*), were F_S is the fluorescence intensity of the

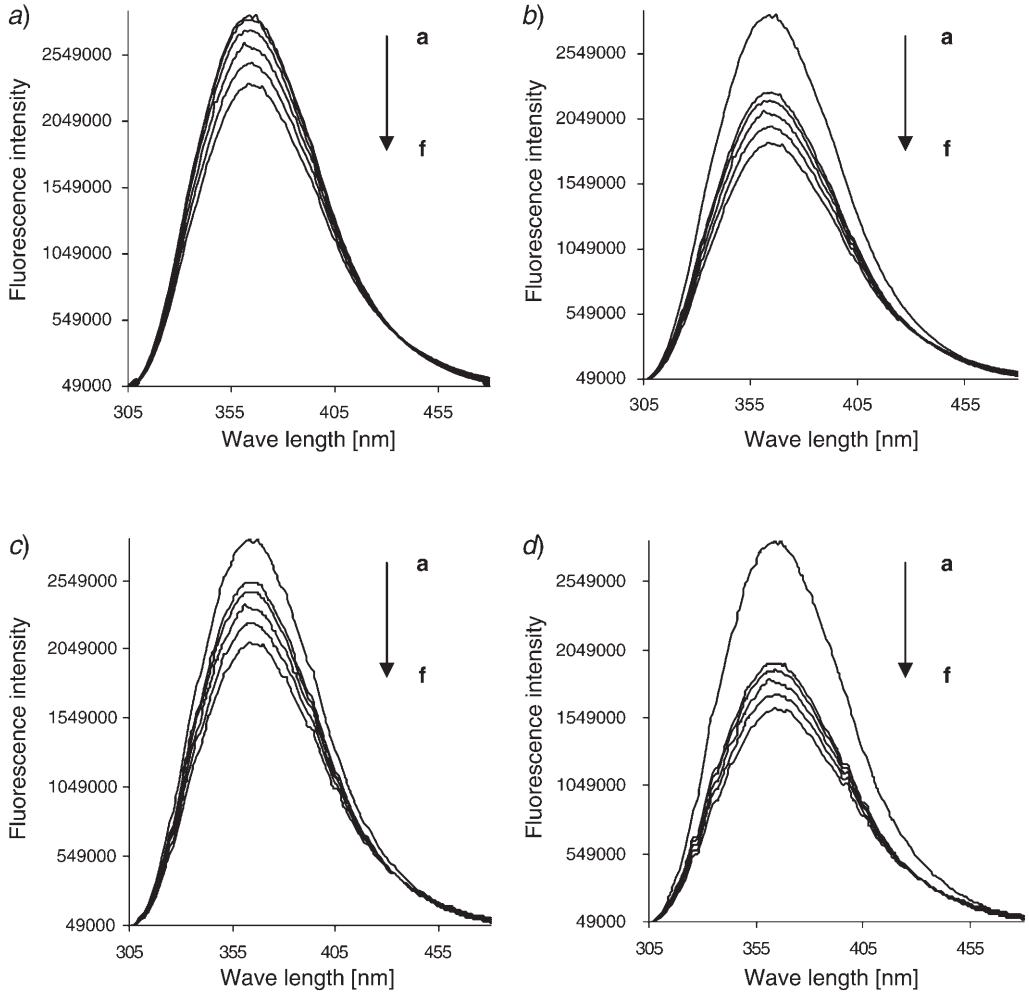


Fig. 2. *Fluorescence spectra of* a) (R)-**1** *and* b) (S)-**1** *with various concentrations of* **3**, *and of* c) (R)-**1** *and* d) (S)-**1** *with various concentrations of* **4**. The concentrations of **3** and **4** increased from 0 to 0.1, 0.2, 0.5, 1.0, and 1.5 mM (from *a* to *f*), and the concentration of (*R*)-**1** and (*S*)-**1** was $1 \cdot 10^{-4}$ M.

Table 1. *Association Constants* K_s [M^{-1}] *of the Drug · Host Complexes*

Host	(*R*)-**1**	(*S*)-**1**
2	$1.0 \cdot 10^4 \pm 2 \cdot 10^2$	$1.0 \cdot 10^4 \pm 2 \cdot 10^2$
3	$1.2 \cdot 10^3 \pm 4 \cdot 10$	$2.0 \cdot 10^4 \pm 3 \cdot 10^2$
4	$1.1 \cdot 10^4 \pm 2 \cdot 10^2$	$4.5 \cdot 10^4 \pm 4 \cdot 10^2$

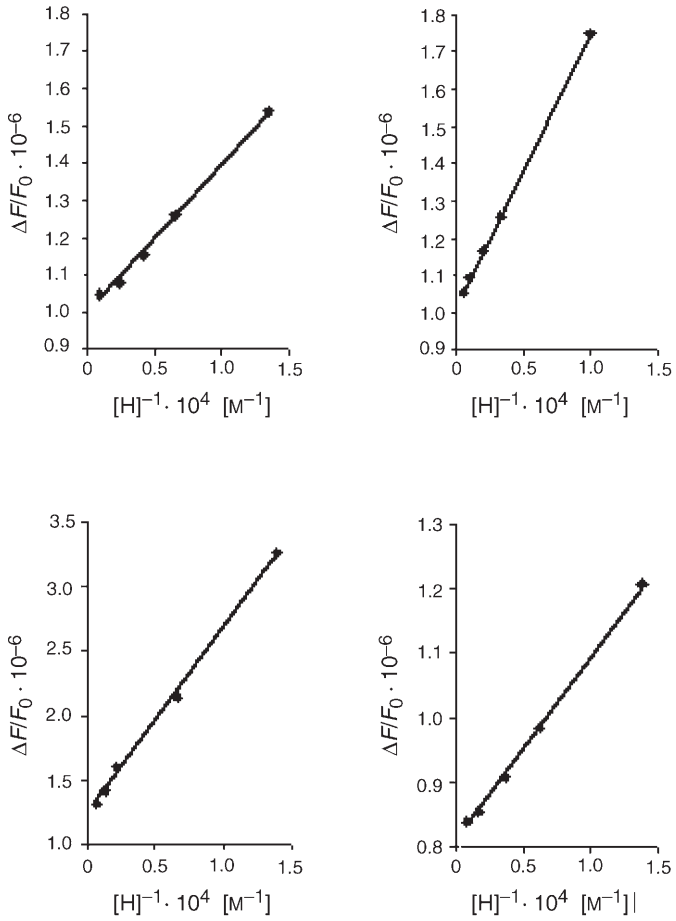


Fig. 3. *Estimation of association constants for* a) (R)-**1** *and* b) (S)-**1** *with* **3**, *and for* c) (R)-**1** *and* d) (S)-**1** *with* **4**. The plot is based on the fluorescence-intensity changes at 366 nm with a 1:1 model: $\Delta F/F_0$ *vs.* $[H]^{-1}$ ($R = 0.999$).

(S)-**1** · host complex and F_R the fluorescence intensity of the (R)-**1** · host complex. The value of α was 2.87 for the guest with **3** and 2.04 for the guest with **4**.

$$\alpha = \Delta F_S/\Delta F_R, \text{ with } \Delta F_S = F_0 - F_S \text{ and } \Delta F_R = F_S - F_R \quad (3)$$

The inclusion phenomena of the guest into the cyclodextrin cavity was studied with the host **3** as a representative example by means of ^{1}H-NMR spectroscopy. NMR Spectroscopy is one of the most important techniques used for the characterization of inclusion complexes. The formation of an inclusion complex results in chemical-shift changes of the CD-host and the guest protons [6e][10]. The inclusion of the spacer into the β-CD cavity and its movement to the outside environment after inclusion of the guest into the CD cavity can be easily monitored by the ^{1}H-NMR chemical-shift

changes of the protons of β-CD (**2**), the host **3**, and the free spacer **5**. In the case of the host **3**, the inclusion of the spacer into the cavity resulted in a shift of the signals of the glucose H–C(3) ($\Delta\delta = -0.016$) and H–C(5) ($\Delta\delta = 0.007$) as compared to H–C(3) and H–C(5) of **2**, and also by the upfield shift of pyrrolidinylidene-proton signals of the host **3** ($\Delta\delta = -0.020$ to -0.053) as compared to those of the free spacer **5**. Thus, the downfield shift of the pyrrolidinylidene protons of the spacer ($\Delta\delta =$ up to 0.047) of the host **3** upon inclusion of the guest into the cavity established the movement of the spacer from the hydrophobic to hydrophilic environment.

The mode of inclusion of the guest into the cavity of host **3** can be seen from *Figs. 4* and *5*. The ^{1}H-NMR spectra of guest **1** and of the 1:1 complexes of (*S*)-**1**·**3** and (*R*)-**1**·**3** are shown in *Fig. 4*. The glucose H–C(3) signal of the complexes (*R*)-**1**·**3** ($\Delta\delta = -0.014$) and (*S*)-**1**·**3** ($\Delta\delta = -0.010$) and the H–C(5) signal of (*R*)-**1**·**3** ($\Delta\delta = -0.016$) and (*S*)-**1**·**3** ($\Delta\delta = -0.005$) shifted upfield as compared to those of **3** indicating the formation of a complex of the guest with **3**. Decoupling of the OH protons of the host was observed in the complex showing the existence of fast proton exchange, probably associated with the alteration of the H-bonding network due to the interaction with the guest. The changes in the ^{1}H-NMR chemical shifts of the guest zolmitriptan (**1**) were also observed in the complex (*Fig. 5*). In (*R*)-**1**·**3**, all the protons of zolmitriptan were shifted upfield indicating inclusion of the total moiety in the cyclodextrin cavity, whereas in the case of (*S*)-**1**·**3**, the protons of CH_2N (=CH_2(9); $\Delta\delta = 0.028$) and Me_2N (=2 Me(10); $\Delta\delta = 0.027$) were shifted downfield. Thus, this part of the guest (*S*)-**1** probably was not included into the cavity and interacted with the

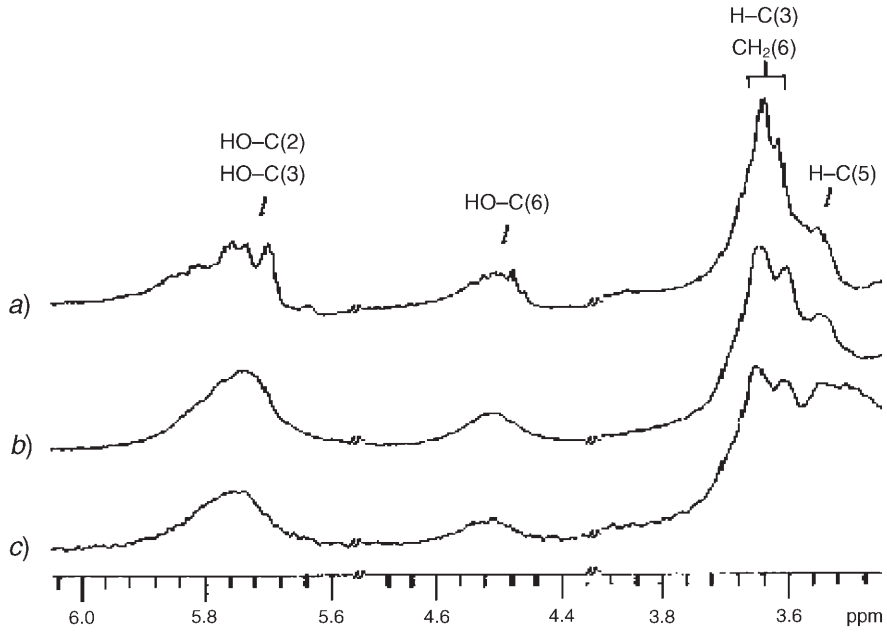


Fig. 4. *^{1}H-NMR Spectrum* (200 MHz, (D_6)DMSO, 25°) *of* a) *host* **3**, b) *1:1 complex (*S*)-***1***·***3***, and* c) *1:1 complex (*R*)-***1***·***3*** (glucose protons)

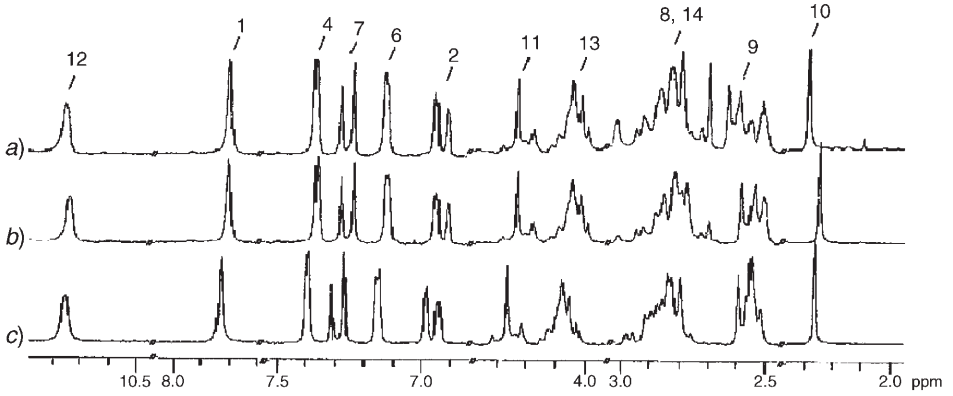


Fig. 5. *^{1}H-NMR Spectrum* (200 MHz, (D_6)DMSO, 25°) *of* a) *1:1 complex (*S*)-***1*·***3***, b) *1:1 complex (*R*)-***1*·***3***, and* c) *guest* **1** (see *Fig. 1* for atom numbering of **1**)

OH protons of the wider rim of the host **3**, causing the downfield shift of these protons. The remaining protons of the guest, however, were shifted upfield, indicating their inclusion in the host cavity. These shifts in the ^{1}H-NMR spectra indicated complex formation between zolmitriptan and the host **3**. Different changes in the chemical shifts for (*R*)-**1**·**3** and (*S*)-**1**·**3** established that the mode of inclusion of the two guest enantiomers (*R*)- and (*S*)-**1** into the host cavity is different.

Summary. The modified β-CDs **3** and **4** showed enantioselectivity in their interactions with the chiral guests of zolmitriptan, host **3** exhibiting more enantioselectivity than **4**. The methodology to determine the enantioselectivities is based on the enantiomer discrimination in complex formation, which differs from most of the existing methods which depend upon kinetic discrimination. The described methodology may find utility in analytical procedures where fast screening methods are required and may also lead to the development of a new type of enantioselective catalysts in organic synthesis.

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Experimental Part

General. Chlorosulfonyl isocyanate (=sulfuryl chloride isocyanate) and ethylenediamine (=ethane-1,2-diamine; *Aldrich*) were used as received without further purification. β-Cyclodextrin (*Sigma Aldrich*) was recrystallized from H_2O and dried *in vacuo* at 60°. The 1-methylpyrrolidin-2-one (*S. D. Fine*) was distilled and dried before use. All solns. were prepared with high-purity H_2O. Stock solns. of guest ($1 \cdot 10^{-4}$ mol l^{-1}) and host ($1 \cdot 10^{-4}$ mol l^{-1}) were prepared by dissolving the appropriate amounts of the compounds in H_2O. CC = Column chromatography. TLC: precoated silica gel plates (*60 F_{254}*, 0.2 mm layer; *E. Merk*). M.p.: *Fischer–Johns* melting-point apparatus; uncorrected. UV Spectra: *Jasco* spectrophotometer; λ_{max} in nm. Fluorescence spectra: *Fluorolog* instrument; 1×0.2 cm quartz cell; zolmitriptan (UV, λ_{max} 283 nm) was excited at 293 nm. ^{1}H-NMR Spectra: *Gemini* 200-MHz spectrometer; (D_6)DMSO at 25°; δ in ppm, J in Hz. HR-MS: *Applied-Biosystems* mass spectrometer; in m/z.

Mono{6-deoxy-6-{{2-{{[(1-methylpyrrolidin-2-ylidene)amino]sulfonyl}amino}ethyl}amino}}-β-cyclodextrin (**4**). Mono{6-[(2-aminoethyl)amino]-6-deoxy}-β-cyclodextrin [11] (1 mmol) and *N*-(1-methylpyrrolidin-2-ylidene)sulfamoyl chloride [12] (1.1 mmol) were dissolved in 1-methylpyrrolidin-2-one (20 ml), and the mixture was stirred at r.t. for 24 h. The mixture was poured into excess acetone (300 ml), and the precipitate was filtered and washed with acetone. The precipitate was dissolved in H_2O, the soln. filtered and lyophilized, and the residue purified by CC (*RP*^{18} silica gel): 1.12 g (83.7%) of **4**. Light yellow powder. M.p. 185–187°. UV/VIS (tetraborate buffer): 267 nm. ^{1}H-NMR (D_2O): 2.08–2.23 (*m*, 4 H); 2.84–2.96 (*m*, 4 H); 3.03 (*s*, 3 H); 3.16 (*d*, *J* = 7.5, 2 H); 3.5–3.76 (*m*, 21 H); 3.8–4.0 (*m*, 21 H); 5.07 (*s*, 7 H). ESI-HR-MS: 1337.4664 ($[M+H]^+$, $C_{49}H_{85}N_4O_{36}S^+$; calc. 1337.4612).

1:1 Complexes (*S*)-**1** · **3** *and* (*R*)-**1** · **3** *for* ^{1}*H-NMR Studies.* Host **3** (1 mmol) was dissolved in distilled H_2O (10 ml), then the (*S*)- or (*R*)-**1** (1 mmol) dissolved in H_2O (3 ml) was added slowly, and stirring at 50° was continued for 3 h. Then the mixture was lyophilized to remove H_2O and to isolate the complex.

REFERENCES

[1] G. R. Martin, *N. Adv. Headache Res.* **1994**, *4*, 3; G. R. Martin, A. D. Robertson, S. J. MacLennan, D. J. Prentice, V. J. Barrett, J. Buckingham, A. C. Honey, H. Giles, *Br. J. Pharmacol.* **1997**, *121*, 157.

[2] R. Yates, K. Niarn, R. Dixon, J. V. Kemp, A. L. Dane, *J. Clin. Pharmacol.* **2002**, *42*, 1244; A. D. Oldman, L. A. Smith, H. J. McQuay, R. A. Moore, *Pain* **2002**, *97*, 247; L. Yu, T. Yao, S. Ni, S. Zeng, *Biomed. Chromatogr.* **2005**, *19*, 191.

[3] A. Ueno, T. Kuwabara, A. Nakamura, F. Toda, *Nature (London, U.K.)* **1992**, *356*, 136; K. Hamasaki, H. Ikeda, A. Nakamura, A. Ueno, F. Toda, I. Suzuki, T. Osa, *J. Am. Chem. Soc.* **1993**, *115*, 5035; M. Rekharsky, Y. Inoue, *J. Am. Chem. Soc.* **2000**, *122*, 4418; G. Hembury, M. Rekharsky, A. Nakamura, Y. Inoue, *Org. Lett.* **2000**, *2*, 3257.

[4] M. L. Bender, M. Komiyama, 'Cyclodextrin Chemistry', Springer-Verlag, New York, 1978; J. Szejtly, 'Cyclodextrins and Their Inclusion Complexes', Akademiai Kiado, Budapest, 1982; R. J. Clarke, J. H. Coates, S. F. Lincoln, *Adv. Carbohydr. Chem. Biochem.* **1989**, *46*, 205.

[5] C. J. Easton, S. F. Lincoln, *Chem. Soc. Rev.* **1996**, *25*, 163.

[6] a) S. E. Brown, J. H. Coates, P. A. Duckworth, S. F. Lincoln, C. J. Easton, B. L. May, *J. Chem. Soc., Faraday Trans.* **1993**, *89*, 1035; b) I. Tabushi, Y. Kuroda, T. Mizutani, *J. Am. Chem. Soc.***1986**, *108*, 4514; c) S. E. Brown, J. H. Coates, C. J. Easton, S. J. Van Eyk, S. F. Lincoln, B. L. May, M. A. Stile, C. B. Whalland, M. L. Williams, *J. Chem. Soc., Chem. Commun.* **1994**, 47; d) R. Corradini, A. Dossena, G. Implellizzeri, G. Maccarrone, R. Marchelli, E. Rizzarelli, G. Sartor, G. Vecchio, *J. Am. Chem. Soc.* **1994**, *116*, 10267; e) S.-Y. Jia, Y.-Q. Hao, L.-N. Li, K. Chen, Y. Wu, J. Liu, L. Wu, Y.-H. Ding, *Chem. Lett.* **2005**, *34*, 1248; f) H. Bakirci, W. M. Nau, *J. Photochem. Photobiol. A: Chem.* **2005**, *173*, 340; g) S. Pagliari, R. Corradini, G. Galaverna, S. Sforza, A. Dossena, M. Montalti, L. Prodi, N. Zaccheroni, R. Marchelli, *Chem.–Eur. J.* **2004**, *10*, 2758.

[7] H. Ikeda, Q. Li, A. Ueno, *Bio. Med. Chem. Lett.* **2006**, *16*, 5420; M. A. Hossain, H. Mihara, A. Ueno, *J. Am. Chem. Soc.* **2003**, *125*, 11178; M. A. Hossain, K. Takahashi, H. Mihara, A. Ueno, *J. Inclusion Phenom. Macrocyclic Chem.* **2002**, *43*, 271; T. Tanabe, K. Touma, K. Hamasaki, A. Ueno, *Anal. Chem.* **2001**, *73*, 1877.

[8] L. R. Reddy, M. A. Reddy, N. Bhanumathi, K. R. Rao, *J. Chem. Res., Synop.* **2000**, 494.

[9] K. A. Connors, 'Binding Constants. The Measurements of Molecular Complex Stability', Wiley, New York, 1987; H. A. Benesi, J. H. Hildebrand, *J. Am. Chem. Soc.* **1949**, *71*, 2703; C. Yang, L. Liu, T.-W. Mu, Q.-X. Guo, *Anal. Sci.* **2000**, *16*, 537.

[10] H. J. Schneider, F. Hacket, V. Rudiger, H. Ikeda, *Chem. Rev.* **1998**, *98*, 1755; P. V. Demarco, A. L. Thakkar, *J. Chem. Soc., Chem. Commun.* **1970**, 2; H. Yamamura, Y. Yamada, R. Miyagi, K. Kano, S. Araki, M. Kawai, *J. Inclusion Phenom. Macrocyclic Chem.* **2003**, *45*, 211.

[11] Y. Takenaka, M. Higashi, N. Yoshida, *J. Chem. Soc., Perkin Trans. 2* **2002**, 615.

[12] K. R. Rao, Y. V. D. Nageswar, T. N. Srinivasan, P. B. Sattur, *Synth. Commun.* **1988**, *18*, 877.

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