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Synthesis of new diaza-18-crown-6 ethers derived from *trans*-(*R*,*R*)-1,2-diaminocyclohexane and investigation of their enantiomeric discrimination ability with amino acid ester salts

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ABSTRACT

The synthesis of four diaza-18-crown-6 ethers with C_2-symmetry derived from *trans*-(*R*,*R*)-1,2-diaminocyclohexane bearing methyl, phenyl and phenoxymethyl moeities attached to a stereogenic centre on the crown ring were achieved. Enantiomeric discrimination of these macrocycles against amino acid methyl ester salts was examined by ^{1}H NMR titration method. They exhibit strong binding ability and some of them show a very high enantioselectivity towards amino acid esters, corresponding to 5.37 kJ/mol of binding energy difference in $CDCl_3$ at 25 °C. Computational modelling showed parallel results with experimental calculations, thus providing a detailed understanding of molecular recognition mode and binding sites between the hosts and the guests.

1. Introduction

Designing synthetic receptors that are capable of discriminating between enantiomers have occupied a special place[1] in gaining a basic knowledge of molecular recognition, separation of enantiomers for synthetic[2] and analytical[3] applications. The ability of a molecule to discriminate between an enantiomer pair is based on its capacity to form a molecular complex preferentially with one of the enantiomers by non-covalent interactions, such as hydrogen bonding, electrostatics and hydrophobics.[4]

Optically active polyazamacrocycles are important compounds in organic,[5] supramolecular,[6] medicinal[7] and bioorganic[8] chemistry. Additionally, these structures have shown extraordinary properties for the molecular recognition of cationic[9] and anionic species[10] and have been widely used as chiral solvating agents (CSAs) for the fast and accurate determination of enantiomeric excesses in combination with NMR spectroscopy.[11] Among the described non-racemic chiral polyazamacrocycles, those bearing *trans*-1,2-diaminocyclohexane as the chiral receptor are especially useful due to the structural peculiarities of this moiety. The six-membered ring of the molecule with a *trans*-1,2-substitution also renders a C_2-symmetry and a rigid scaffold with a very precisely defined disposition of the two amino groups.[12] However, the syntheses of these systems are not trivial, due to the low yields usually associated with the key macrocyclization step.

Amino acids and their derivatives are chiral organic molecules involved in a wide variety of biological processes. The study of the enantiomeric recognition of these compounds is of particular significance for understanding the interactions between biological molecules and design of asymmetric catalytic systems, new pharmaceutical agents,[13] and separation materials.[3a] Chiral crown ethers with C_2-symmetry are characterized by their relatively simple recognition mechanism and usually show higher enantioselectivity than those with C_1- and D_1-symmetry.[1] In particular, macrocycles with C_2-symmetry have been extensively studied because of their structural architecture.[14] It has been reported that macrocycles synthesized from chiral 1,2-diaminocyclohexane have been used as molecular receptors for peptides[15] and in enantiomeric recognition of amino acids.[16] The fields of application of this molecule range from the preparation of chiral catalysts for asymmetric synthesis and the synthesis of supramolecular receptors or the chiral stationary phases for separation science.[17] Systematic investigation on the synthesis of macromolecules from this versatile amine should lead to fruitful results.

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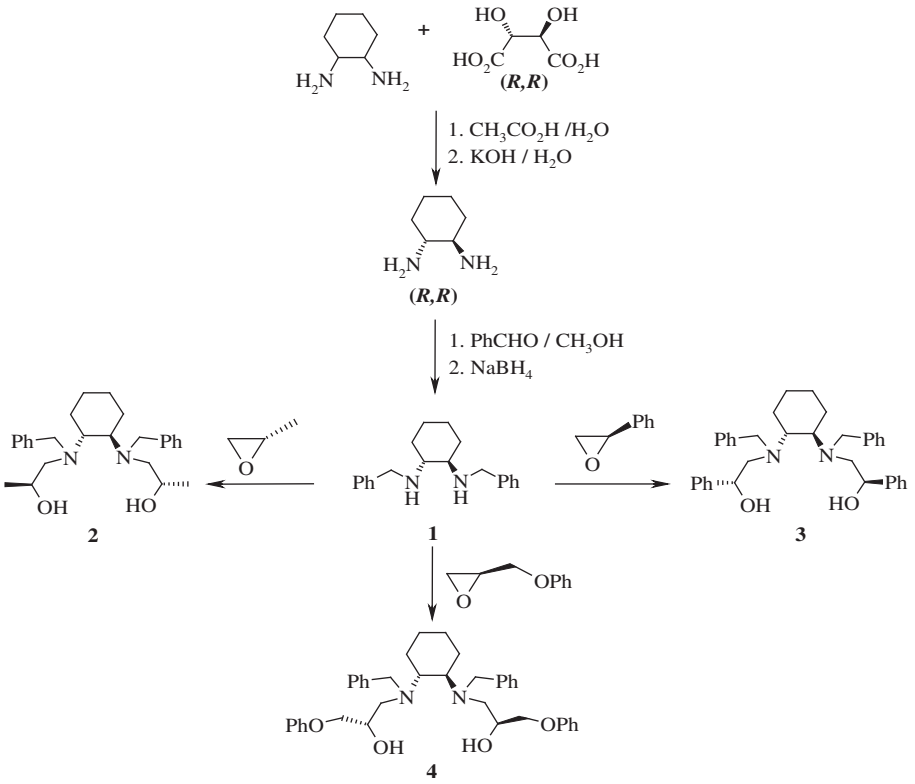
We herein report on the results of the synthesis of C_2-symmetric chiral diaza-18-crown-6 ether derivatives **5**–**8** and their molecular recognition properties towards various salts of amino acid esters. Therefore, C_2-symmetric chiral amino alcohols **2**–**4** were synthesized from chiral 1,2-diaminocyclohexane **1** and chiral terminal epoxides by regioselective ring opening reaction in order to incorporate the additionally stereogenic centres to the macroring (Scheme 1). This chirality built into the macrocycles offers additional interesting features enabling the macrocycle to be used as a potential receptor for molecular recognition. Chiral diaza-18-crown-6 derivative macrocycles **5**–**8** were synthesized by ring closing reaction of chiral amino alcohols **2**–**4** and appropriate ditosylates (Scheme 2). Their complexation ability and molecular recognition properties towards enantiomers of alanine, phenylalanine and valine methyl ester hydrochlorides were investigated by ^{1}H NMR titration method. Molecular dynamic calculations were also performed for the complexes of the host **8** with enantiomers of phenylalanine methyl ester salts to the detailed binding modes of these complexes.



Scheme 1. Synthesis of chiral amino alcohols **2**–**4**.

2. Results

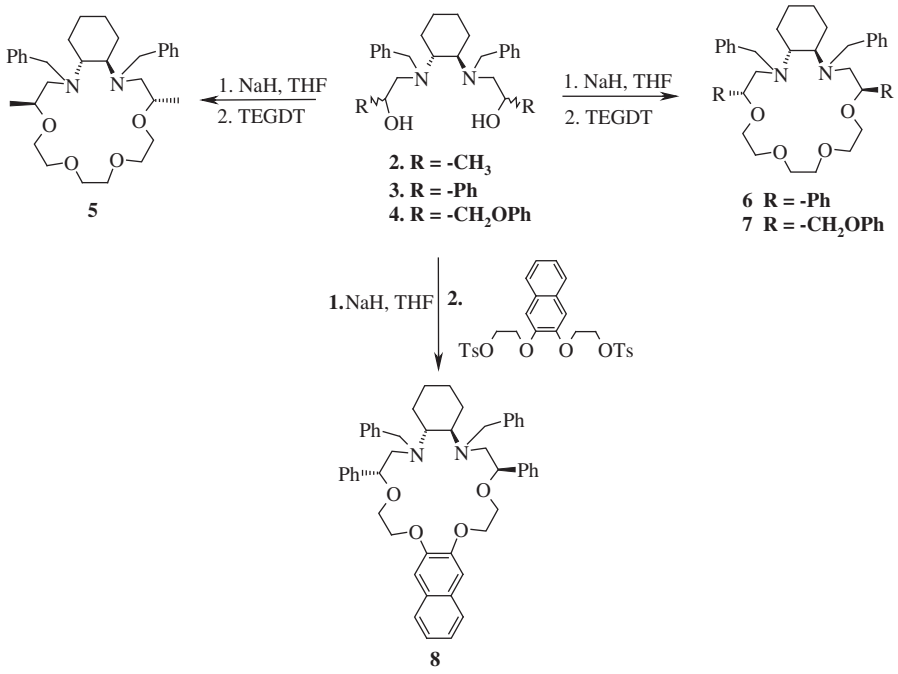
2.1. Synthesis

In the present study, four macrocycles involving chiral 1,2-diaminocyclohexane unit and having stereogenic centres bearing methyl, phenyl, phenoxymethyl moieties were synthesized. Chiral 1,2-diaminocyclohexane was chosen as a starting material because of its ease of synthesis in sufficiently large quantities combined with a unique molecular architecture, which allows further sophisticated structures. Thus, (*R*,*R*)-*trans*-1,2-diaminocyclohexane, (*S*)-propylene oxide, (*R*)-styrene oxide and (*S*)-glycidyl phenyl ether were used as chiral sources. Chiral amine **1** was prepared by treating enantiomerically pure *trans*-(*R*,*R*)-1,2-diaminocyclohexane obtained from *cis*-/*trans* isomeric mixture by classical resolution technique,[18] and benzaldehyde followed by $NaBH_4$ reduction of the intermediate imine.[19] Chiral amino alcohols **2**–**4** were synthesized by the regioselective ring opening reaction of (*S*)-propylene oxide, (*R*)-styrene oxide and (*S*)-glycidyl phenyl ether with chiral amine **1** in 99%, 75% and 82.5% yields, respectively (Scheme 1). Amine **1** exhibited a preference for nucleophilic attack at the terminal or achiral carbon of epoxides with a S_N2 reaction to give the corresponding chiral β-amino alcohols **2**–**4** with retention of configuration at stereogenic centre as reported previously.[20,21] Therefore, this route provides an easy and practical method for preparing chiral ligands incorporating with different alkyl or aryl substituents. Diaza-18-crown-6 derivatives **5**–**7** were synthesized under high dilution technique by ring closing reaction of chiral amino alcohols **2**–**4** and triethylenglycol ditosylate in moderate yields (Scheme 2). Macrocyclic **8** was also obtained by reacting chiral amino alcohols **3** with ditosylate bearing a naphtho unit, which may create an additional binding site, possibly providing π–π stacking interactions and enabling specific recognition for amino acid derivatives with aromatic side chains.[22,23]

2.2. Determination of binding constant by ^{1}H NMR titration

In order to determine the stoichiometry of the complexes, a Job plot experiment was performed. A typical plot for the complex of **5** with L-alanine methyl ester salt is illustrated in Fig. 1. The concentration of the complex approaches maximum when the molar fraction of [R]/[R]+[L] is 0.5, suggesting that host **5** forms a 1:1 complex with L-alanine methyl ester hydrochlorides. The ^{1}H NMR titrations were used to obtain binding constant between hosts and guests. For each complex, a set of 9 samples containing constant host concentration (0.833 mM) and increasing concentration of the guest ranging from 0.417 to 4.54 mM was prepared and their ^{1}H NMR spectrum was recorded.

The chemical shifts in the α-hydrogen of each guest in the ^{1}H NMR spectrum versus increasing the guest concentration were



Scheme 2. Synthesis of chiral diaza-18-crown-6 ethers **5**–**8**.

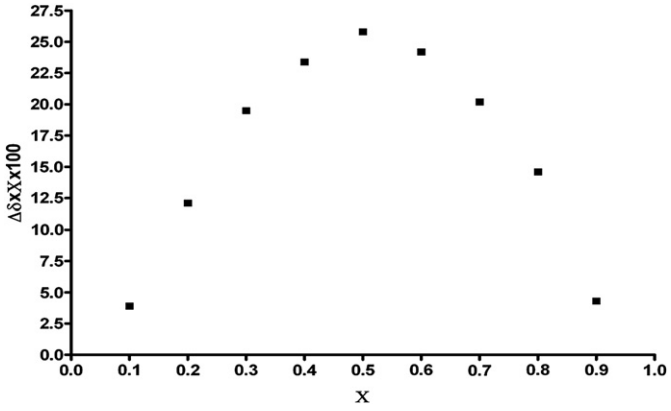


Fig. 1. Job plots of macrocycle **5** with L-alanine methyl ester salt. [X=molar fraction of guest, Δδ=chemical shift change of α-proton of guest].

determined. The changes in the chemical shifts of the α-hydrogen of D- and L-phenylalanine methyl ester salts upon complexation with the host **5** are demonstrated in Fig. 2. These changes were fitted to Equation 3 derived from Equation 1 in order to calculate the dissociation constant within 95% of confidence (see Table 1). A representation of the data fitted to Equation 4 for the changes in the chemical shifts of the α-hydrogen of D- and L-alanine methyl ester salts complexed with the host **5** are plotted in Fig. 3.

$$\text{Host} + \text{Guest} \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} \text{Host}\cdot\text{Guest} \tag{1}$$

$$K_{\text{diss}} = \frac{k_{-1}}{k_1} = \frac{[\text{Host}][\text{Guest}]}{[\text{Host}\cdot\text{Guest}]} \tag{2}$$

$$[\text{Guest}]_{\text{tot}} = [\text{Guest}]_{\text{free}} + [\text{Guest}]_{\text{complex}} \tag{3}$$

$$\delta_{\text{obs}} = \frac{(\delta_0 K_{\text{diss}} + \delta_{\max}[\text{Guest}]_{\text{tot}})}{(K_{\text{diss}} + [\text{Guest}]_{\text{tot}})} \tag{4}$$

where K_{diss} is the dissociation constant between a host and a guest, δ_0 is the chemical shift in specific hydrogen of a guest in the absence of a host, δ_{obs} is the observed chemical shift in the same hydrogen in the presence of a host and $\delta_{\max}$ is the maximum chemical shift achieved in the presence of a host.

2.3. Computational modelling

Root Mean Square Displacement (RMSD) changes during the molecular dynamic simulation performed for a period of 20 ns at 700 K for the host **8** are presented in Supplementary data. The conformation with the largest population obtained from cluster analyses for the host **8** is illustrated in Fig. 4. RMSD changes during the molecular dynamic simulation performed for a period of 20 ns at 300 K for the complexes of the host **8** with D- and L-phenylalanine salts are displayed in Supplementary data. The conformation with the largest population obtained from cluster analyses for the complexes of the hosts with D- and L-phenylalanine salts are illustrated in Fig. 5.

3. Discussion

^{1}H NMR spectra (400 MHz) of both chiral amino alcohols **2**–**4** and macrocycles **5**–**8** show considerable line broadening compared to the compound **1** almost for all the peaks, probably indicating a higher conformational flexibility and intermolecular mobility as reported previously.[14,24] It is known that the symmetry and rigidity of cyclohexane ring display very characteristic ^{1}H NMR spectra with clear axial and equatorial positions for all the protons within the ring.[12] However, no significant change was observed in ^{1}H NMR spectrum at 233 K in $CDCl_3$ as shown in Fig. 6 for chiral amino alcohol **3** which is in agreement with those reported previously.[25,26]

Systems having strong interactions or good chiral recognition allow for further investigation of their structural properties. The choice of the 18-membered macrocycles is generally based on the

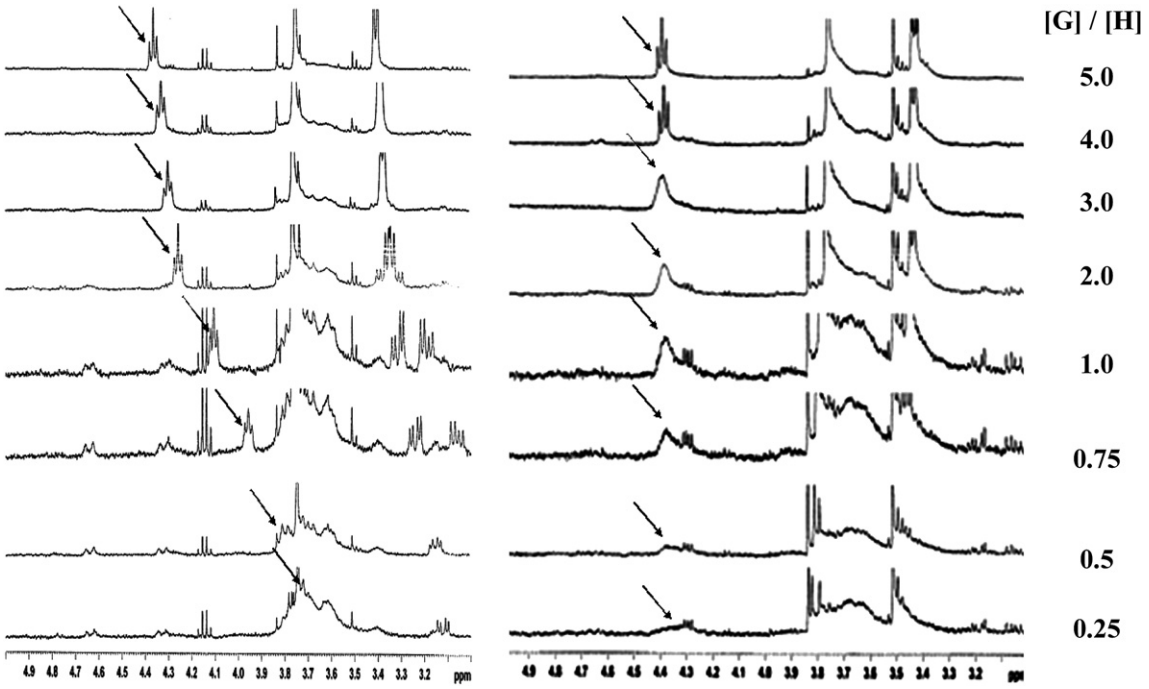


Fig. 2. Changes in ^{1}H NMR chemical shifts in the α-hydrogen of methyl ester salts of L-phenylalanine (left) and D-phenylalanine salt (right) on their concentration in the presence of the macrocycle **5** (0.833 mM) in $CDCl_3$ at 25 °C.

fact that they form more stable complexes with ammonium cations in solution than other crown ethers with larger or smaller rings. The insertion of 1,2-diaminocyclohexane subunit also provides a favourable effect in their interaction with ammonium salts: first, $N^+H\cdots N$ hydrogen bond interaction are much more stable than $N^+H\cdots O$ bonds.[27] Second, cyclic 1,2-diaminocyclohexane has unique structural properties, which make it very useful for the induction of a chiral environment (chiral ligands).[12] To detail the interactions in the complex formation is a very difficult task and involves sophisticated use of various approaches. The use of NMR spectroscopic methods to study such complexation phenomena is one of the most useful techniques available to chemists. This method includes the observation of changes in the chemical shift occurred in one site of either the host or the guest. One of the disadvantages of this technique is the requirement for the higher concentration of the species being followed, which may lead to self aggregation and therefore misleading to changes in chemical shifts.

Table 1
Binding constants (K), enantioselectivities K_L/K_D, free-energy changes ($-\Delta G_o$), and $\Delta\Delta G_o$ calculated from ΔG_o for complexation of guests with chiral hosts **5**–**8** in $CDCl_3$ at 25 °C

Host[a]	Guest[b]	K (M^{-1})	K_L/K_D	$-\Delta G_o$ (kJ mol^{-1})	$\Delta\Delta G_o$ (kJ mol^{-1})
5	L-AlaOMe·HCl	2060±47	1.92	18.90±0.12	1.61
	D-AlaOMe·HCl	1074±61		17.29±0.28	
	L-PheOMe·HCl	18,832±1224	8.64	24.38±0.32	5.34
	D-PheOMe·HCl	2179±61		19.04±0.14	
	L-ValOMe·HCl	196±7	2.39	13.07±0.17	2.15
	D-ValOMe·HCl	82±4		10.92±0.24	
6	L-AlaOMe·HCl	327±22	0.24	14.34±0.33	−3.53
	D-AlaOMe·HCl	1355±58		17.87±0.21	
	L-PheOMe·HCl	378±24	0.22	14.70±0.32	−3.72
	D-PheOMe·HCl	1694±88		18.42±0.26	
	L-ValOMe·HCl	540±25	0.78	15.58±0.23	−0.60
	D-ValOMe·HCl	688±44		16.19±0.36	
7	L-AlaOMe·HCl	561±28	0.67	15.68±0.25	−0.98
	D-AlaOMe·HCl	831±21		16.66±0.13	
	L-PheOMe·HCl	791,766±10,293	1.06	33.65±0.06	0.15
	D-PheOMe·HCl	745,712±20,880		33.50±0.14	
	L-ValOMe·HCl	1858±19	0.54	18.65±0.05	−1.53
	D-ValOMe·HCl	3451±68		20.18±0.10	
8	L-AlaOMe·HCl	833±36	1.43	16.67±0.21	0.91
	D-AlaOMe·HCl	580±30		15.76±0.26	
	L-PheOMe·HCl	1302±73	0.15	17.77±0.35	4.75
	D-PheOMe·HCl	194±12		13.02±0.30	
	L-ValOMe·HCl	1994±44	2.09	18.82±0.28	1.83
	D-ValOMe·HCl	952±30		16.99±0.16	

$\Delta\Delta G_o = \Delta G_o(L) - \Delta G_o(D)$.
[a] Concentration of macrocycle **5**–**8**=8.33×10^{-4} mol dm^{-3}.
[b] AlaOMe·HCl: alanine methyl ester hydrochloride, PheOMe·HCl: phenylalanine methyl ester hydrochloride, ValOMe·HCl: valine methyl ester hydrochloride.

The data in Table 1 indicate that macrocycle **5** possessing a methyl side arm attached to the stereogenic centre on the crown ring prefers to bind L enantiomers of all the guests. It is expected that the side arms with small sizes like methyl would not have a significant influence on the discrimination ability of a crown ring along with its flexibility. However, the introduction of the *trans*-diaminocyclohexane unit into the crown ring may slow down the conformational changes in the ring as well as provide chiral binding sites for the guests and hence this would be the main driving force for the discrimination of the enantiomers as illustrated in Fig. 7. Thus, the substituent on the crown ring will not be the primary contributors to the enantiomeric recognition. In fact, the data in Table 1 for the complexes of macrocycle **5** with amino acid salts pairs fit this assumption since the host had a significant ability to discriminate between the enantiomers of phenylalanine methyl ester salts compared to those of alanine and valine. This may be attributed to the favourable π−π interactions between the benzyl group of L guest and the benzyl group of the host since it the anti (methyl=↑, benzyl=↓), anti (benzyl=↑, methyl=↓) orientation of the methyl and the *N*-benzyl groups on the crown ring is expected (Fig. 7). It is known that this kind of stacking plays crucial roles in chemical and biological recognition, which is detailed in an excellent review by Diederich and co-workers.[28] This assumption is based on the postulation that the α-hydrogen of the guests will have an average location facing opposite to the cyclohexane unit since this part of the crown ring is thought to undergo major conformational changes and therefore it will impose rather more steric sites to interact with the guests.

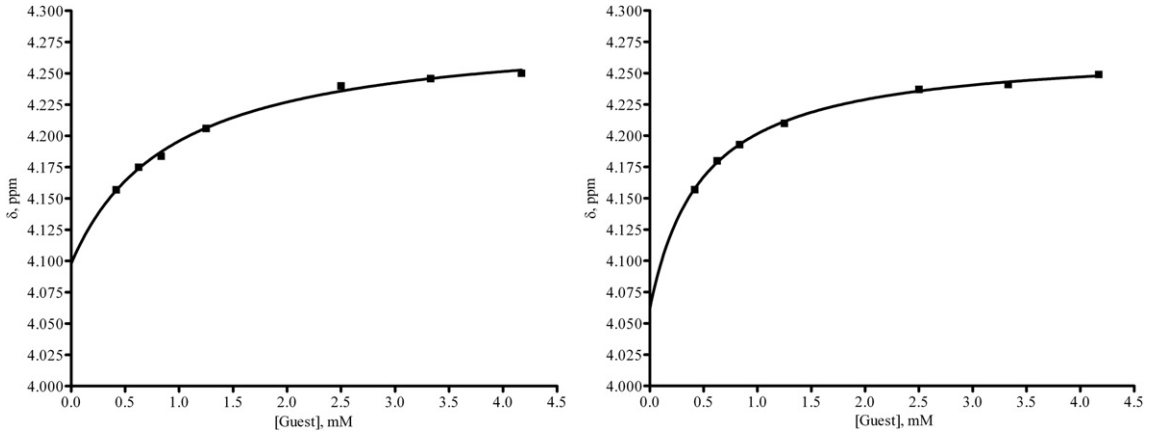


Fig. 3. Chemical shifts in the α-hydrogen of methyl ester salts of D-alanine (left) and L-alanine (right) on their concentration in the presence of the macrocycle **5** (0.833 mM) in $CDCl_3$ at 25 °C.

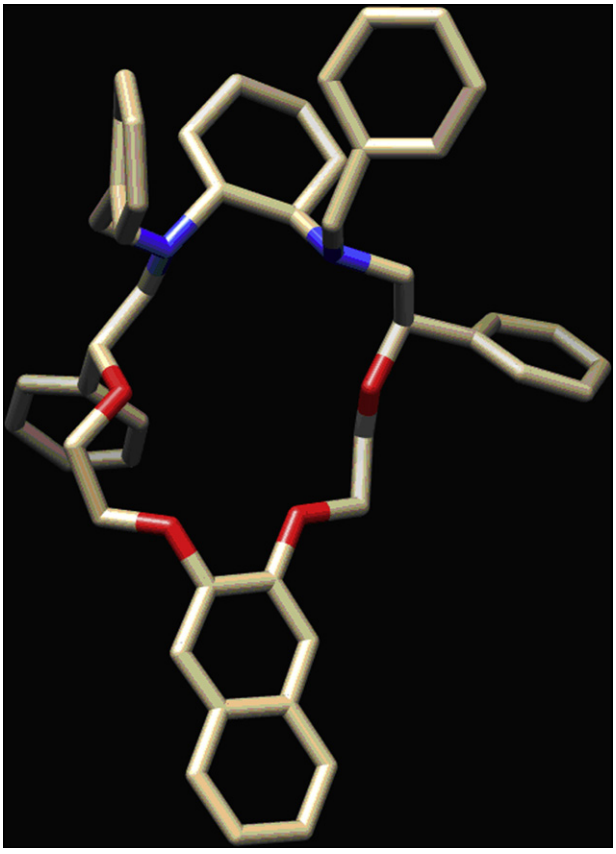


Fig. 4. A conformation of the macrocycle **8** with the largest population corresponding to one of a lower energy conformer obtained from MD calculations.

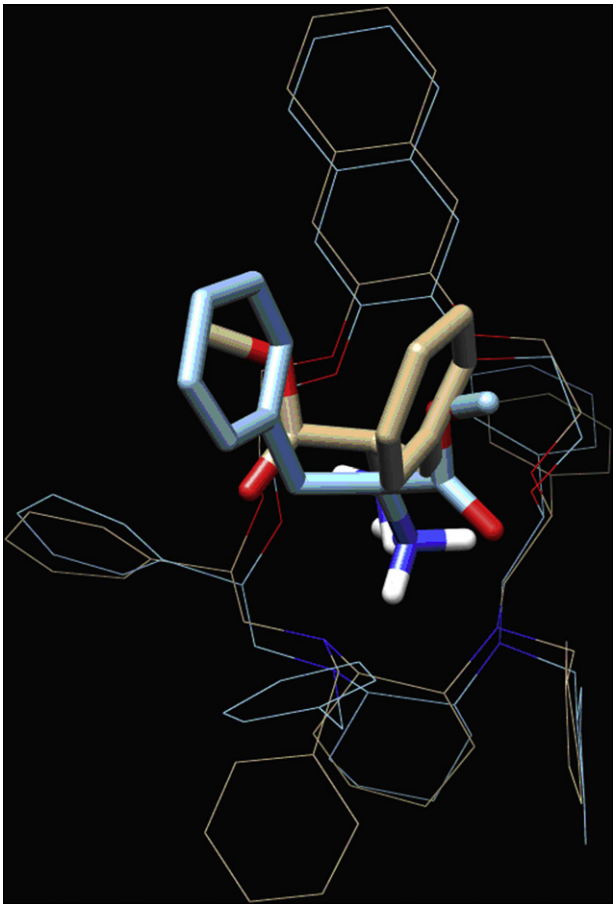


Fig. 5. The superimposed structure of the complexes of the macrocycle **8** with D- and L-phenylalanine methyl ester salts. Each complex corresponds to a conformation with largest populations extracted from MD calculations.

In the case of macrocycle **6**, the methyl group attached to the stereogenic centre is replaced with the phenyl and the absolute configuration is inverted. Therefore, one should expect that the host **6** will have an opposite discrimination affect against the guests compared to the host **5**. In fact it was observed that the host **6** prefers to form more stable complexes with D-enantiomers and the discrimination factor is still significantly larger for the phenylalanine salts (Table 1). However, one may wonder why the L enantiomer of phenylalanine salt is not preferably bound to the host **6** if the assumption of the attractive π–π interactions between the guest and the host is favoured. This may be attributed to the changes in the orientation of the groups in the host **6**, namely the phenyl and benzyl. In this case, the anti (phenyl=↓, benzyl=↑), anti (benzyl=↓, phenyl=↑) orientation is excepted (Figs. 8 and 9) and therefore, the benzyl group in D-enantiomer will have a favourable π–π interactions with the *N*-benzyl group of the host with *syn* alignment as occurred between the L enantiomer of phenylalanine salt and the host **5**. It was found that the host **6** shows a quite similar discrimination factor against enantiomers of alanine methyl ester salts. It is quite possible that a favourable interaction takes place between the methyl, the side group in D-alanine ester salt and the benzyl in the host, which is regarded as CH/π interactions. It is also a common non-covalent interaction in chemical and biological

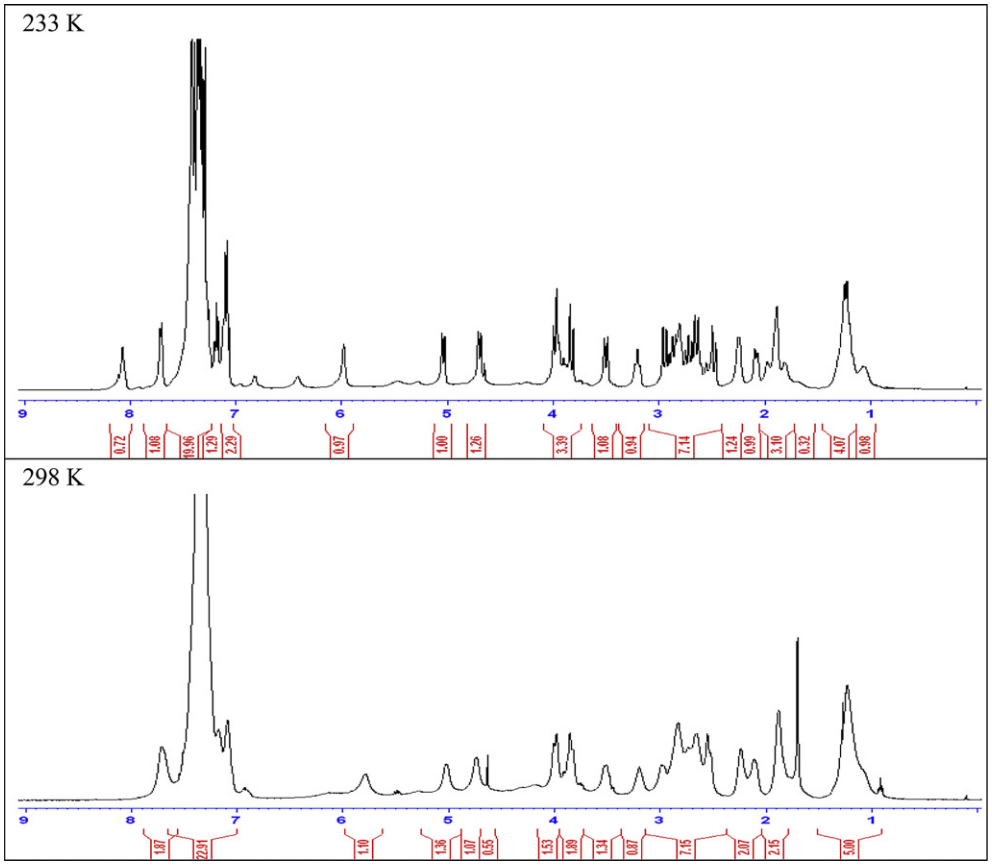


Fig. 6. ^{1}H NMR spectra of amino alcohol **3** in $CDCl_3$ at 298 and 233 K.

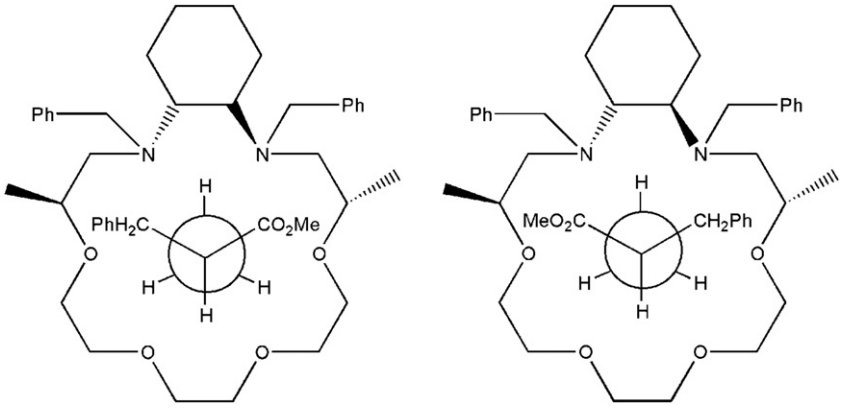


Fig. 7. A representative interaction of the macrocycle **5** with D- and L-phenylalanine ester salts.

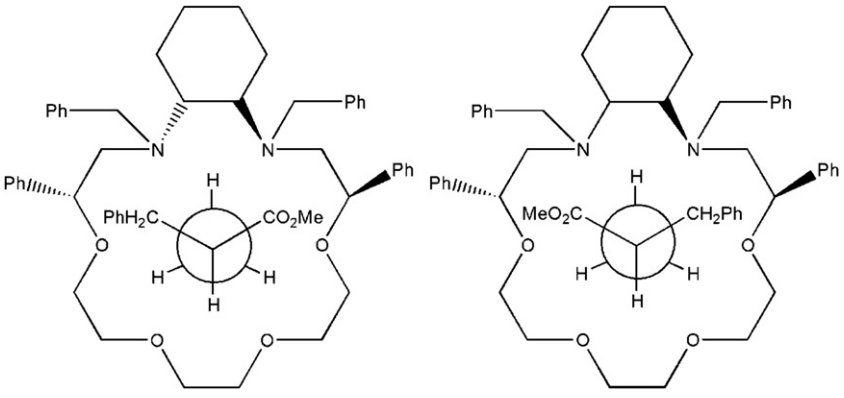


Fig. 8. A representative interaction of the macrocycle **6** with D- and L-phenylalanine ester salts.

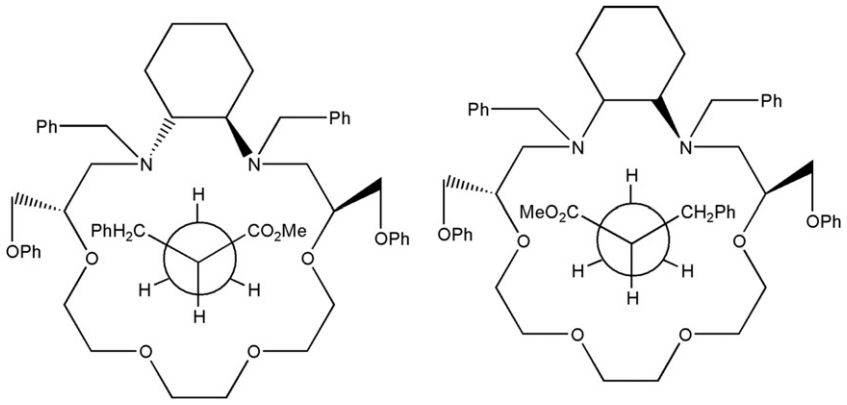


Fig. 9. A representative interaction of the macrocycle **7** with D- and L-phenylalanine ester salts.

recognition, with particular interest in determining the tertiary structure of proteins.[29]

As to the host **7**, a similar action was achieved in for the discrimination between enantiomers of alanine and valine salts while a reverse effect is found for that of phenylalanine salts compared to the host **6**. It is likely that $-CH_2OPh$ group will have more conformational flexibility compared to the phenyl group in the host **6** and probably larger pocket site. So the isopropyl group will be fitted to the pocket and therefore it will be better differentiated by this pocket.

The introduction of the naphthyl unit into the crown ring resulted in producing the host **8**, which prefers to bind enantiomers' L configuration, an opposite effect found by the host **6**, although both have the same absolute configuration at their stereogenic centres. It is likely that the naphthyl group causes conformational changes in the crown ring and hence in the alignment of the groups on the ring and possibly forming a pseudo-chiral binding site with a hydrophobic nature. Thus, the site arms of all the amino acid ester salts with L configuration will be sandwiched between the naphthyl unit and the phenyl group (Fig. 10). The larger discrimination factor found for the salts of phenylalanine methyl ester may be associated with the more pronounced interactions between the side arm of this guest, namely the benzyl group, and the naphthyl and phenyl groups in the host via $\pi-\pi$ stacking in a manner of sandwiching.

Computational calculations indicated indeed that the host **8** forms more stable complex with L enantiomer of phenylalanine salt. The superimposed conformers with the larger populations obtained from cluster analysis showed that in fact the benzyl group of L enantiomer has a location to interact with the naphthyl and the phenyl groups in the host as postulated above (Fig. 10).

4. Conclusion

Four chiral diaza-18-crown-6 ether derivatives with C_2-symmetry were prepared and their complexation and discrimination ability against alanine, valine and phenylalanine methyl ester salts were studied by ^1H NMR titration method. They showed strong affinity and different complementarities for various amino acid ester salts, and exhibit excellent enantiodiscriminating abilities towards some of these guests. It was observed that changes in the substituents attached to chiral centre in the crown ring as well as insertion of the naphthyl unit into the ring influence both binding and enantiodiscrimination ability of macrocyclic compounds against amino acid salts pairs. Thus the information gained in this study provides significant results, which may be used in the understanding of the molecular recognition in the biological processes.

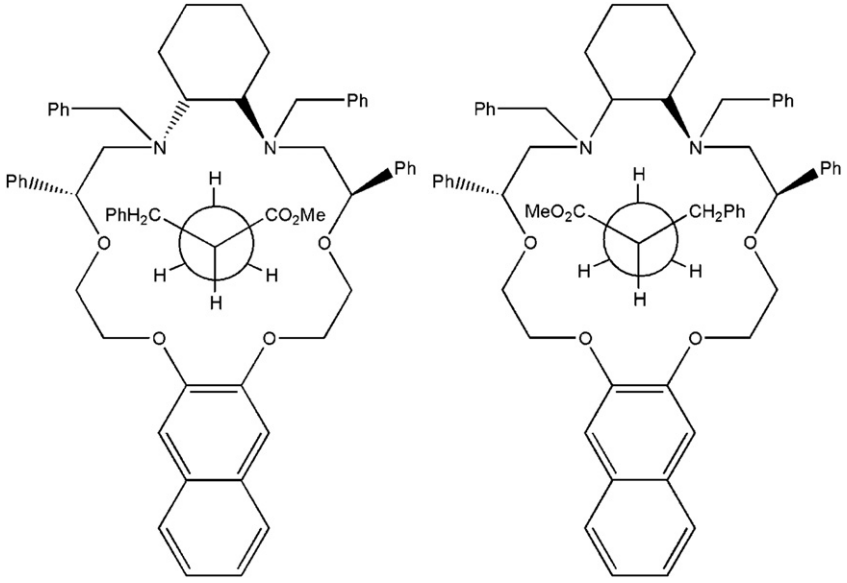


Fig. 10. A representative interaction of the macrocycle **8** with D- and L-phenylalanine ester salts.

5. Experimental

5.1. General information

All chemicals were reagent grade unless otherwise specified. (*R*)-styrene oxide, (*S*)-propylene oxide and (*S*)-glycidyl phenyl ether were purchased from Sigma–Aldrich. Silica Gel 60 (Merck, 0.040–0.063 mm) and silica gel/TLC-cards (F_{254}) were used for flash column chromatography and TLC. THF was dried (on sodium benzophenone ketyl) and distilled prior to use. All reactions were carried out under N_2 atmosphere with a dry solvent of anhydrous conditions, unless otherwise noted. Melting points were determined with GALLENKAMP Model apparatus with open capillaries. Optical rotations were taken on a Perkin Elmer 341 model polarimeter. IR spectra were recorded on Mattson 1000 ATI Unicam FT-IR spectrometer. ^{1}H NMR (400 MHz) and ^{13}C NMR (100 MHz) spectra were recorded on Bruker Avance-400 MHz high performance digital FT-NMR spectrometer, with tetramethylsilane as the internal standard solutions in deuteriochloroform. Elemental analyses were obtained with CARLO-ERBA Model 1108 instrument.

5.2. NMR host–guest titration

The host compound was dissolved in an appropriate amount of solvent ($CDCl_3$) and the resulting solution evenly distributed among eight NMR tubes. The guest compound was also dissolved in the appropriate amount of solvent and added in increasing amount to the NMR tubes, so that the solutions with the following relative amounts (equiv) of guest versus host compound (concentration was 8.33×10^{-4} M) were obtained 0.25, 0.50, 0.75, 1.0, 2.0, 3.0, 4.0, 5.0. The chemical shift change of α-proton of guests shifted to the downfield by the increasing the ratio of guests as shown in Fig. 3. The dissociation constants (*K*) between each host and guest were calculated from the change in δ values against increasing guest concentration in the presence of constant host concentration. by non-linear least-squares fitting method[30] using GraphPad Pris 4 pocket programme.

5.3. Evaluation of the stoichiometry of the host–guest complex (Job plots)

The stoichiometry of host–guest complexes were determined according to Job's method of continuous variations.[31] Stock solutions of hosts (5 mM) and guests (5 mM) were prepared in $CDCl_3$. These solutions were distributed among ten NMR tubes in such a way that the molar fractions *X* of host and guest in the resulting solutions increased (or decreased) from 0.1 to 1.0 (and vice versa). The compellation-induced shifts ($\Delta\delta$) were multiplied by *X* and plotted against *X* itself (Job plots). A representative example of macrocycle **5** with L-alanine methyl ester hydrochloride can be seen from Fig. 2.

5.4. Synthesis

5.4.1. (R,R)-1,2-Diaminocyclohexane. (*R*,*R*)-1,2-Diaminocyclohexane was isolated from isomeric *cis*- and *trans*-1,2-diaminocyclohexane according to the method reported.[18] (*R*,*R*)-1,2-Diaminocyclohexane L-tartrate: yield: 39%; $[\alpha]_D^{20}$ +11.6 (*c* 1, H_2O) (lit.[18] +11.6 (*c* 1, H_2O)); (*R*,*R*)-1,2-diaminocyclohexane: yield: 95%, $[\alpha]_D^{20}$ −19.8 (*c* 1, 1 M HCl), [lit.[18] −20 (*c* 1, 1 M HCl).

*5.4.2. (R,R)-N,N′-Dibenzyl-1,2-diaminocyclohexane (**1**).* This compound was synthesized by a previous method.[19] Yield: 2.60 g, 99%; $[\alpha]_D^{20}$ −79.7 (*c* 2.5, $CHCl_3$) (lit.[19] −80 (*c* 2.5, $CHCl_3$)); δ_H (400 MHz, $CDCl_3$) 7.50–7.20 (10H, m), 3.98 (2H, d, *J* 13.2 Hz), 3.75 (2H, d, *J* 13.2 Hz), 2.40–2.30 (2H, m), 2.24 (2H, d, *J*=13.2 Hz), 2.15 (2H, bs), 1.85–1.81 (2H, m), 1.36–1.30 (2H, m), 1.15–1.1 (2H, m); δ_C (100 MHz, $CDCl_3$) 141.0, 128.4, 128.2, 126.8, 60.9, 50.9, 31.6, 25.1.

*5.4.3. N,N′-Dibenzyl-N,N′-di[(S)-2-hydroxypropyl]-(1R,2R)-diaminocyclohexane (**2**).* (*S*)-Propylene oxide (118 mg, 2.0 mmol) was added to a solution of (*R*,*R*)-*N*,*N*′-dibenzyl-1,2-diaminocyclohexane (**1**) (250 mg, 0.82 mmol) in methanol (3 mL) and stirred at 40 °C for 24 h and 50 °C for 24 h. Solvent evaporated to give pure of compound **2** (340 mg, 99%) as a viscous colourless oil; [found: C, 76.16; H, 9.39; N, 6.76. $C_{26}H_{38}N_2O_2$ requires C, 76.06; H, 9.33; N, 6.82%]; $[\alpha]_D^{20}$ −38.3 (*c* 1, $CHCl_3$); ν_{max} (liquid film) 3384, 3031, 2923, 2858, 1953, 1600, 1452, 1296, 1388, 1336, 1253, 1132, 1060, 964, 746 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 7.36–7.15 (10H, m), 5.92 (2H, bs), 3.74 (2H, d, *J* 12.8 Hz), 3.49–3.40 (4H, m), 2.70–2.60 (4H, m), 2.40–2.34 (2H, m), 2.00–1.90 (2H, m), 1.80–1.60 (2H, m), 1.20–0.9 (10H, m); δ_C (100 MHz, $CDCl_3$) 139.8, 129.6, 128.2, 127.2, 65.9, 64.3, 59.8, 56.9, 26.0, 25.0, 21.78.

*5.4.4. (1R,2R)-N,N′-Dibenzyl-bis[(R)-2-hydroxy(2-phenyl)ethyl]-1,2-diaminocyclohexane (**3**).* (*R*)-Styrene oxide (1.63 g, 13.6 mmol) was added to a solution of (*R*,*R*)-*N*,*N*′-dibenzyl-1,2-diaminocyclohexane (2 g, 6.8 mmol) in methanol (3 mL) and stirred at 40, 50 and 60 °C, 24 h for each temperature. Solvent was evaporated and then unreacted epoxide and amine was removed by kugelrohr distillation apparatus. Crude product was purified by crystallization from *n*-hexane to give **3** (2.72 g, 75%) as a white powder, mp 202–203 °C; [found: C, 80.25; H, 7.46; N, 5.30. $C_{36}H_{42}N_2O_2$ requires C, 80.89; H, 7.86; N, 5.24%]; $[\alpha]_D^{20}$ −147.6 (*c* 1, $CHCl_3$); ν_{max} (KBr) 3429, 3242, 3063, 3030, 2928, 2851, 1490, 1458, 1336, 1240, 1195, 1105, 1066, 1022, 758, 714 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 7.71–7.07 (20H, m), 5.78 (2H, bs, OH), 5.02 (1H, bs), 4.73 (1H, bs), 4.0–3.50 (4H, m), 3.19–2.55 (6H, m), 2.23–1.69 (4H, m), 1.28–1.23 (4H, m). δ_C (100 MHz, $CDCl_3$) 143.2, 138.4, 130.6, 130.1, 128.4, 127.3, 126.0, 72.3, 69.0, 58.4, 57.5, 25.7, 24.2.

*5.4.5. N,N′-Dibenzyl-N,N′-di[(S)-3-phenoxy-2-hydroxypropyl]-(1R,2R)-1,2-diaminocyclo-hexane (**4**).* (*S*)-Phenyl glycidyl ether (1.53 g, 10.2 mmol) was added to a solution of (*R*,*R*)-*N*,*N*′-dibenzyl-1,2-diaminocylohexane (**1**) (1.5 g, 5.1 mmol) in methanol (4 mL) at 60 °C for 24 h. Solvent was evaporated then the crude product was purified by crystallization from *n*-hexane–ethanol to give **4** (2.5 g, 82.5%) as a white solid, mp 83–85 °C; $[\alpha]_D^{20}$ −53 (*c* 1, EtOH); [found: C: 76.52, H: 7.52, N: 4.91. $C_{38}H_{46}N_2O_4$ requires C: 76.74, H: 7.80, N: 4.71%]; ν_{max} (KBr) 3360, 3169, 3060, 3028, 2929, 2855, 1600, 1496, 1455, 1373, 1302, 1274, 1120, 1080, 1036, 935, 884, 813, 754, 693 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 7.45–7.05 (10H, m), 5.6 (2H, bs, OH), 4.36–3.79 (4H, m), 3–2.68 (3H, m), 2.31–1.92 (3H, m), 1.26 (2H, bs); δ_C (100 MHz, $CDCl_3$) 158.8, 138.3, 129.9, 129.5, 128.9, 128.4, 120.9, 114.7, 70.5, 67.1, 58.7, 56.3, 52.2, 25.7, 24.3.

*5.4.6. N,N′-Dibenzyl–(2R,9R)-dimethyl-(5S, 6S)-(5, 6)-cyclohexenyl-4,7-diaza-1,10,13,16-tetraoxaoctadecane (**5**).* To a suspension of 325 mg (12.2 mmol, 90% in mineral oil) of NaH in 50 mL of the dry THF at 0 °C was added a solution of *N*,*N*′-dibenzyl-*N*,*N*′-di[(*S*)-2-hydroxypropyl]-(1*R*,2*R*)-diaminocyclohexane (**3**) (1.0 g, 2.44 mmol) in 50 mL of THF. The reaction mixture was refluxed for 2 h. After cooling to 0 °C a solution of tri(ethylene glycol)di(*p*-toluenesulfonate) 1.12 g (2.44 mmol) in 75 mL of THF was slowly added. The suspension was refluxed for 96 h. The solvent was evaporated after adding 50 mL of water to the residue. The mixture was extracted with CH_2Cl_2 (3×25 mL) and the combined organic layers were washed with water (30 mL), dried on $MgSO_4$ and the solvent was evaporated in vacuo to give the crude product, which was purified by column chromatography on silica gel using (85:10:5 PE/EtOAc/TEA) as an eluent to give compound **5** (0.67 g, 31.6%) as a viscous colourless oil; [found: C: 54.20, H: 8.72, N: 5.31. $C_{32}H_{46}N_2O_4$ requires C: 54.40, H:

8.81, N: 5.36%]; R_f (85/10/5 PE/EtOAc/TEA, on silica gel TLC) 0.6; $[\alpha]_D^{20}$ −40.2 (*c* 1, $CHCl_3$); ν_{max} (liquid film) 3060, 3024, 2901, 2241, 1950, 1739, 1704, 1618, 1493, 1452, 1371, 1279, 1107, 960, 828, 737, 700, 647, 470 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 7.33–7.18 (5H, m), 3.77–3.43 (9H, m), 2.87 (1H, dd, *J* 8.4, 5.2 Hz), 2.77 (1H, m), 2.51 (1H, m), 2.04 (1H, d, 11.2 Hz), 1.71 (1H, d, 7.6 Hz), 1.29–1.06 (5H, m); δ_C (100 MHz, $CDCl_3$) 141.5, 129.0, 127.9, 126.4, 76.1, 71.1, 71.0, 67.7, 27.3, 26.2, 18.7.

*5.4.7. N,N′-Dibenzyl-(2R,9R)-2,9-diphenyl-(5R,6R)-5,6-cyclohexenyl-4,7-diaza-1,10,13,16-tetraoxaoctadecane (**6**).* This compound was prepared as described above for compound **5** starting from NaH (373 mg, 14 mmol, 90% in mineral oil), chiral amino alcohol **3** (1.5 g 2.8 mmol) and TEGDT (1.29 g, 2.8 mmol). The crude product was purified by column chromatography on silica gel using 85:10:5 PE/EtOAc/TEA as an eluent to give **6** (1.27 g, 45.5%) as a viscous colourless oil; [found: C: 77.32, H: 7.89, N: 4.15. $C_{42}H_{52}N_2O_4$ requires C: 77.46, H: 8.02, N: 4.32%]; R_f (85:10:5 PE/EtOAc/TEA, on silica gel TLC) 0.65; $[\alpha]_D^{20}$ −39 (*c* 1, $CHCl_3$); ν_{max} (liquid film) 3082, 3025, 2930, 2859, 2363, 1738, 1616, 1493, 1451, 1372, 1243, 1109, 734, 700 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 7.36–7.24 (20H, m), 4.633 (2H, bs), 4.04–3.38 (18H, m), 2.75–2.66 (4H, m), 1.554 (4H, m), 0.94–0.92 (4H, m); δ_C (100 MHz, $CDCl_3$) 142.1, 141.6, 128.4, 128.2, 128.0, 127.5, 127.3, 126.1, 81.3, 72.2, 71.1, 70.9, 68.1, 60.7, 56.7, 28.3, 26.0.

*5.4.8. N,N′-Dibenzyl-(2R,9R)-2,9-diphenoxymethyl-(5R,6R)-5,6-cyclohexenyl-4,7-diaza-1,10,13,16-tetraoxaoctadecane (**7**).* Macrocycle **7** was prepared as usual manner by using NaH (342.6 mg 12.85 mmol 90% in mineral oil), chiral amino alcohol **4** (1.5 g, 1.57 mmol) and TEGDT (1.18 g, 1.57 mmol). Crude product was purified by column chromatography on silica gel using 85:10:5 PE/EtOAc/TEA as an eluent to give **7** (620 mg, 34%) as a viscous colourless oil; [found: C: 67.32, H: 7.89, N: 4.15. $C_{44}H_{56}N_2O_4$ requires C: 67.69, H: 7.90, N: 3.95%]; R_f (85:10:5 PE/EtOAc/TEA, on silica gel TLC) 0.60; $[\alpha]_D^{22}$ +8.5 (*c* 1, $CHCl_3$); ν_{max} (liquid film) 3060, 3009, 2927, 2858, 1599, 1494, 1452, 1350, 1295, 1244, 1114, 1039, 970, 753, 694 cm^{-1}. δ_H (400 MHz, $CDCl_3$) 7.53–7.00 (m, 10H), 4.22–3.74 (m, 11H), 3.20 (bs, 1H), 2.86–2.78 (m, 2H), 1.94–1.71 (m, 2H), 1.37–1.29 (m, 2H). δ_C (100 MHz, $CDCl_3$) 159.1, 141.4, 129.5, 128.6, 128.2, 126.4, 120.8, 114.7, 71.5, 71.2, 70.1, 69.5, 61.4, 56.0, 51.2, 29.1, 26.2.

*5.4.9. N,N′-Dibenzyl-(5R,6R)-5,6-cyclohexenyl-(2R,9R)-2,9-diphenyl-14,15-naphtho-4,7-diaza-1,10,13,16-tetraoxaoctadecane (**8**).* In a manner similar to that described for the preparation of **5** by using NaH (150 mg, 5.6 mmol 90% in mineral oil), chiral amino alcohol **3** (0.6 g, 1.12 mmol) and 2,3-bis[2-(*p*-tolylsulphonyl)ethoxy]naphthalene (625 mg 1.12 mmol). Crude product was purified by column chromatography on silica gel using 85:10:5 PE/EtOAc/TEA as an eluent to give **8** (0.4 g, 32.65%) as a viscous yellow oil; [found: C: 80.05, H: 7.12, N: 3.60. $C_{50}H_{56}N_2O_4$ requires C: 80.21, H: 7.46, N: 3.74%]; R_f (85:10:5 PE/EtOAc/TEA, on silica gel TLC) 0.63; $[\alpha]_D^{20}$ −22.6 (*c* 1, $CHCl_3$); ν_{max} (liquid film) 3388, 3061, 3015, 2931, 2856, 1627, 1601, 1508, 1486, 1452, 1362, 1259, 1216, 1175, 1116, 1053, 1026, 950, 851, 757, 702, 667, 469 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 7.77–7.25 (13H, m), 4.70 (1H, s), 4.54–4.34 (2H, bd), 3.86–3.80 (3H, bd), 3.48 (1H, bs), 2.83–2.71 (2H, m), 1.75–1.68 (2H, m), 1.39 (1H, m), 1.00–0.98 (2H, m); δ_C (100 MHz, $CDCl_3$) 149.8, 142.1, 141.7, 129.6, 129.3, 128.3, 128.1, 127.9, 127.4, 127.2, 127.1, 126.8, 126.5, 80.7, 68.2, 66.8, 65.4, 62.4, 56.7, 27.4, 26.1.

5.5. Computational calculations

AMBER (v9)[32] suite of programmes were used for all molecular dynamic calculations. The hosts and ligands were designed by GaussView 3.09,[33] followed by optimization with Gaussain 03[34] using semi-empirical AM1 method. AM1-Bcc (Austian model with Bond and charge correction)[35] atomic partial charges for the host were determined by antechamber module of AMBER (v9) package and the General AMBER Force Field (GAFF)[36] was adopted in simulation because it handles small organic molecules. For guests parameters were adopted from ff99SB libary.[37]

The host molecule was minimized with a total of 5000 steps, 2500 of steepest descent followed by 2500 of conjugate gradient (maxcyc-ncyc), using a nonbonded cut off of 999 Å and a generalized Born solvent model (igb=0). The system was then heated from 0 K to 700 K at eight steps for a period of 200 ps and it was further simulated at 700 K for a period of 20,000 ps (igb=0). A cluster analysis was performed with 167 intervals out of 500,000 frames to obtain a conformer with a larger population to represent the lower energy conformer. A potassium ion was inserted into the crown cavity of this conformer to achieve a preassociation conformation in order to accommodate guests. Each ligand was manually placed on the surface of the crown ring so that maximum contact points between ammonium and crown donors are achieved. The complexes were minimized with a total of 5000 steps, 2500 of steepest descent, followed by 2500 of conjugate gradient, using a nonbonded cut off of 999 Å and a generalized Born solvent model (igb=0). The system was then heated from 0 K to 700 K at eight steps for a period of 200 ps and it was further simulated at 700 K for a period of 20,000 ps (igb=0). A cluster analysis was performed with 67 intervals out of 20,000 frames and the coordinates of the structure with the largest population was recorded and this was minimized followed by cooling from 700 K to 300 K at eight steps for a period 200 ps and it was further computed at 300 K for a period of 20,000 ps. Molecular dynamic coordinates were recorded with 1.0 ps intervals. A cluster analysis was used to obtain the conformer with the largest population for each complex.

Energy changes and root-mean-square displacement (RMSD) analysis for the hosts and complexes were carried out on the trajectories by the ptraj module of AMBER (v9). 3D structures were displayed using by Chimera (UCSF)[38] and potential energy and RMSD graphics are shown by GraphPad Pris 4 pocket programme.

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Supplementary data

Supplementary data related to this article can be found online at http://dx.doi.org/10.1016/j.tet.2012.10.020.

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