Tetrahedron 64 (2008) 5036–5043

Contents lists available at ScienceDirect

Tetrahedron

journal homepage: www.elsevier.com/locate/tet

Stereoselective synthesis of hydroxylated β-aminocyclohexanecarboxylic acids

Loránd Kiss [a], Enikő Forró [a], Tamás A. Martinek [a], Gábor Bernáth [a,b], Norbert De Kimpe [c], Ferenc Fülöp [a,b,*]

[a] Institute of Pharmaceutical Chemistry, University of Szeged, Eötvös u. 6, H-6720 Szeged, Hungary
[b] Research Group for Stereochemistry, Hungarian Academy of Sciences, University of Szeged, Eötvös u. 6, H-6720 Szeged, Hungary
[c] Department of Organic Chemistry, Faculty of Bioscience Engineering, Ghent University, Coupure Links 653, B-9000 Ghent, Belgium

ARTICLE INFO

Article history:
Received 30 January 2008
Received in revised form 4 March 2008
Accepted 19 March 2008
Available online 22 March 2008

Keywords:
β-Lactams
β-Amino acids
Epoxidation
Stereoselective synthesis

ABSTRACT

A simple synthetic approach has been developed for the regio- and diastereoselective synthesis of hydroxylated 2-aminocyclohexanecarboxylic acid stereoisomers from 1,4-cyclohexadiene by the reductive opening of appropriate epoxide intermediates derived from the corresponding bicyclic β-lactams. This method has been extended to the synthesis of these hydroxylated β-amino acids in enantiomerically pure form.

1. Introduction

In recent years, conformationally constrained alicyclic β-amino acids have gained great interest as a consequence of their pharmacological potential. These compounds are found in a large number of natural products, β-lactams or antibiotics (e.g., cispentacin). They are also important building blocks for the synthesis of peptide oligomers.[1] Gellman et al. recently reported the incorporation of 3-methoxy- or 3-phenoxy-substituted *trans*-2-aminocyclopentanecarboxylic acid residues into short 12-helical β-peptides.[2a] The presence of a polar side-chain in the peptide oligomers not only exerts a great influence on the formation of their secondary structure, but can also have an enormous effect on their biological activity in an amphiphilic structure.[2] Apart from this, hydroxylated derivatives (taxol, bestatin, and related compounds) are of considerable interest, all having promising biological properties as potential therapeutic agents.[3] Among the alicyclic hydroxy-β-amino acids, the natural oryzoxymycin exhibits moderate activity against *Xanthomonas oryzae*.[4] Whilst a number of methods have been developed for the diastereo- and enantioselective preparation of cyclic β-amino acids, only a few examples are available for the synthesis of hydroxyl-substituted β-aminocyclohexanecarboxylic acids.[3e,5] One short approach for the synthesis of hydroxy-functionalized β-aminocyclohexanecarboxylic acids is the base-induced fragmentation of β-amino esters with an oxanorbornene or oxanorbornane skeleton.[6] Methyl 2-benzyloxycarbonylamino-5-hydroxy-3-cyclohexenecarboxylate has been used as a starting compound for the incorporation of an extra amino group onto the cyclohexane skeleton.[7] The introduction of a hydroxy group onto the cyclohexane ring has also been accomplished stereoselectively from *cis*- and *trans*-2-aminocyclohexenecarboxylic acids by iodolactonization or via the corresponding oxazine derivatives.[8] Our research group recently developed a new method for the hydroxylation of *trans*-2-aminocyclohexenecarboxylic acid by functionalization of the olefinic bond via an epoxidation reaction.[9] Our present aim was to utilize the above procedure involving the stereodirected epoxidation reaction for the synthesis of different hydroxylated 2-aminocyclohexanecarboxylic acids. The β-lactam **2** derived from 1,4-cyclohexadiene offered an excellent possibility for the introduction of a hydroxy function onto the cyclohexane ring by epoxidation of the olefinic bond.

2. Results and discussion

1,4-Cyclohexadiene (**1**) was first transformed into ethyl *cis*-2-benzyloxycarbonylamino-4-cyclohexenecarboxylate (**3a**) via β-lactam derivative **2**.[11] Chlorosulfonyl isocyanate (CSI) addition to **1** afforded lactam **2**, which was subsequently subjected to lactam ring opening and N-protection reactions forming ester **3a**. In the next step of the strategy, β-amino ester **3a** was submitted to epoxidation of the olefinic bond with *m*-CPBA in CH_2Cl_2 (Scheme 1). Epoxidation of a mono-*N*-protected aminoalkene (carbamate or amide) with peracids is known to give a high degree of 'cis selectivity',[10]

* Corresponding author. Tel.: +36 62 545564; fax: +36 62 545705.
E-mail address: fulop@pharm.u-szeged.hu (F. Fülöp).

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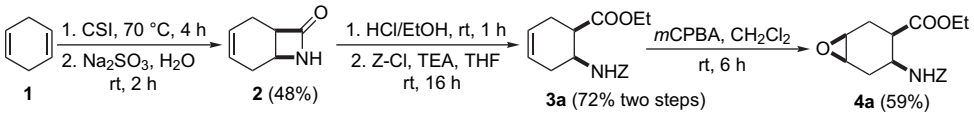
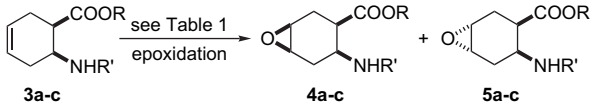
doi:10.1016/j.tet.2008.03.068



Scheme 1. Formation of epoxy amino ester **4a**.

presumably via a hydrogen-bonding interaction of the amide and the peracid in the transition state of the reaction.

The epoxidation of different amino esters or acids with Boc or Z protection at the nitrogen has been performed (Scheme 2) under the conditions presented in Table 1.



Scheme 2. Epoxidation of *cis*-β-amino esters **3a–c**; **a**: R=Et, R′=Z; **b**: R=Bn, R′=Z; **c**: R=Et, R′=Boc.

Table 1
Epoxidation reaction of *cis*-β-amino esters **3a–c**

Entry	R	R′	Alkene	Reaction conditions	Yield (%)	Epoxide
1	Et	Z	**3a**	*m*-CPBA, CH_2Cl_2	59	**4a**
2	Et	Z	**3a**	Peracetic acid	51	**4a**
3	Et	Z	**3a**	Dimethyldioxirane	64	**4a**
4	Et	Z	**3a**	*m*-CPBA, [BMIM][PF_6]	49	**4a**
5	Bn	Z	**3b**	*m*-CPBA, CH_2Cl_2	65	**4b**
6	Et	BOC	**3c**	*m*-CPBA, CH_2Cl_2	57	**4c**
7	Et	BOC	**3c**	Dimethyldioxirane	58	**4c**

The epoxidations were performed in non-polar or polar solvents (e.g., CH_2Cl_2, MeCN or the ionic liquid 1-butyl-3-methylimidazolium hexafluorophosphate), using different oxidizing agents, such as *m*-chloroperbenzoic acid, peracetic acid or dimethyldioxirane. In all cases, the reaction resulted diastereoselectively in *cis*-epoxides **4a–c** as single diastereoisomers, in moderate yields.

The stereoselectivity of the epoxidation reactions was determined from the spectroscopic and GC analyses of the crude products. The stereochemical analysis is presented for epoxide **4a** and the structural assignment can be transferred to **4b** and **4c**. First, the conformation of the cyclohexane ring was determined. The non-overlapping H-6 signal at 2.35 ppm exhibited a scalar coupling of 7.8 Hz together with a weak NOE interaction with H-1 at 2.58 ppm, which indicated their trans-diaxial (*anti*-periplanar) orientation. Due to considerable overlap, the HSQC spectrum was utilized to estimate the coupling constants around H-3; it revealed that the highest ^3J(H-3,H-2) was approximately 5.5 Hz, suggesting an equatorial position for H-2. A strong NOE crosspeak was observed in DMSO between the NH at 6.23 ppm and H-6_{ax} at 2.35 ppm. These findings supported a twist conformation with an axial substituent at position 2 and an equatorial substituent at position 1 (Fig. 1). However, such a conformation can accommodate both cis and trans arrangements of the epoxy ring with an axial oxygen. The structure refinement at the HF/3-21 G level for the two

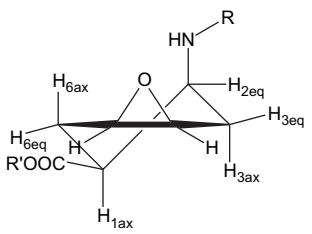


Figure 1. Proposed stereostructure of epoxides **4a–c**.

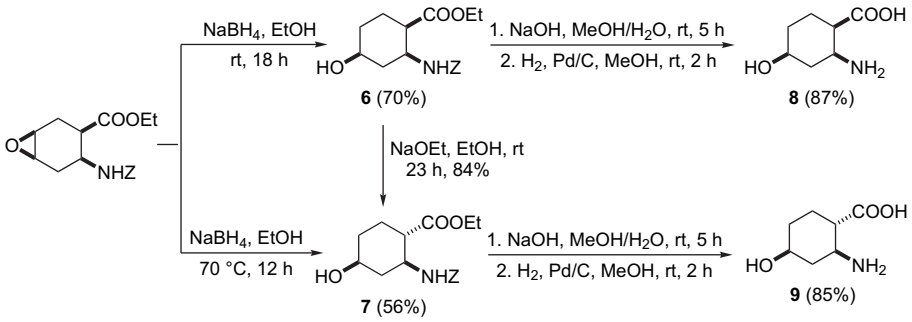
epoxy ring orientations revealed that the equatorial H-5 exhibits different distances from H-6_{ax}. For the trans diastereomer, the distance between H-5 and H-6_{ax} is 2.8 Å, while 2.4 Å is predicted for cis geometry. Due to the inverse sixth-power dependence of the NOE intensity on distance, the difference on the NOE intensity scale is approximately a factor of two, which is not difficult to measure. As an internal reference, the intensity of the H-2_{eq} H-1_{ax} NOESY crosspeak was used (d=2.5 Å). The measured integral value for the H-5 H-6_{ax} crosspeak was 0.38 relative to the H-2_{eq} H-1_{ax} signal, which corresponds to a distance of 2.9 Å. These findings strongly support the *cis*-epoxy arrangement.

The introduction of a hydroxy group onto the cyclohexane skeleton of the 2-aminocyclohexanecarboxylic acid moiety was based on the reductive opening of the oxirane ring of epoxide **4a** (Scheme 3). Opening of the oxirane ring in **4a** using $NaBH_4$ in EtOH at room temperature for 18 h proceeded regioselectively, affording only 4-hydroxy derivative **6** in 70% yield. When opening of the oxirane ring of **4a** was effected at 70 °C for 12 h, isomerization at C-1 was observed, resulting in only hydroxy derivative **7** (Scheme 3). Isomerization also occurred when hydroxy derivative **6** was stirred in EtOH at room temperature in the presence of 1.2 equiv of NaOEt for 23 h, giving compound **7**. The base-mediated partial isomerization of *cis*-epoxy amino ester **4a** could be accomplished in refluxing EtOH in the presence of 3 equiv of K_2CO_3 for 3 days, furnishing a mixture of epoxides **4d**/**4a** in a ratio of 3:1 (Scheme 4).

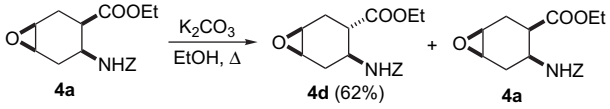
The opposite regioselectivity observed on the reductive opening of epoxides **4a** and **4d** can be analyzed on the basis of their conformational arrangement depicted in Figure 2. The formation of 4-hydroxy-substituted *cis*-amino ester **6** from **4a** or 5-hydroxy substituted *trans*-amino ester **10** from **4d**[9] probably involves hydride attack from the sterically less hindered side (6-H_{eq} and 3-H_{eq}, *approach* **a** for **4a** and *approach* **b** for **4d** in Fig. 2) of the cyclohexane skeleton. Since the opening of the oxirane ring by hydride occurs faster than the isomerization at C-1, it is concluded that the same approach is valid for the formation of hydroxy derivative **7**.

Alkaline hydrolysis of the ester group in compounds **6** and **7** and deprotection of the amino groups by catalytic hydrogenation afforded 2-amino-4-hydroxycyclohexanecarboxylic acids **8** and **9** (Scheme 3).

For 2-amino-4-hydroxy ester **6**, position 4 for the hydroxy group is proven unequivocally from the COSY spectrum. Unfortunately, broadened signals were obtained, possibly due to the conformational flexibility of **6**, which prevented a complete conformational assignment by NMR spectroscopy. Nevertheless, a weak NOE interaction was detected between H-4 and H-2, indicating the cis arrangement of the hydroxy group relative to the protected amino group. For 2-amino-4-hydroxy ester **7**, the COSY crosspeak pattern indicated that the hydroxy group occupies position 4. The conformation of the cyclohexane scaffold is chair with scalar coupling constant values above 12 Hz for ^3J(H-1_{ax},H-2_{ax}), ^3J(H-2_{ax},H-3_{ax}), and ^3J(H-1_{ax},H-6_{ax}), pointing to a trans-diequatorial arrangement of the protected amino and carboxylic groups. The rigid chair conformation is corroborated by the mutual NOEs observed between H-2_{ax}, H-4_{ax}, and H-6_{ax}. The NOEs to H-4_{ax} and the large coupling between H-3_{ax} and H-4_{ax} prove the equatorial orientation of the hydroxy group. For 2-amino-4-hydroxy acid **9**, the COSY spectrum indicates that the hydroxy group is in position 4. The conformation is chair with scalar couplings of ^3J(H-1_{ax},H-2_{ax})=11 Hz and



Scheme 3. Formation of 4-hydroxylated amino esters **6** and **7** and amino acids **8** and **9**.



Scheme 4. Isomerization of the epoxy amino ester **4a** to **4d**.

^3J(H-1$_{ax}$,H-6$_{ax}$)=12 Hz, pointing to a trans-diequatorial arrangement of the amino and carboxylic groups. H-4 exhibits two large couplings (around 12 Hz), which proves the equatorial position of the hydroxy group cis to the amino group. The all-equatorial orientation of the substituents is supported by the NOE interactions observed in the H-2$_{ax}$–H-4$_{ax}$–H-6$_{ax}$ triangle. For 2-amino-4-hydroxy acid **8**, the connectivity pattern proves position 4 for the hydroxy group. The highest scalar coupling around H-1 is estimated as less than 4 Hz, showing its equatorial arrangement, while H-2 is axial because ^3J(H-2$_{ax}$,H-3$_{ax}$)=10.6 Hz. The cis orientation of the hydroxy group relative to the amino substituent is strongly supported by a strong NOE interaction between H-2$_{ax}$ and H-4$_{ax}$. The conformation of the cyclohexane scaffold cannot be determined, due to the unclear spatial arrangement around H-5 and H-6. The most likely explanation is a conformational equilibrium between twist and chair forms.

The method presented above indicated that the presence of a mono-protected amino group (carbamate) on the cycloalkene skeleton always gave rise to cis selectivity during epoxidation, which resulted, after reductive opening of the oxirane ring, in a hydroxy derivative with the hydroxy functionality on the cyclohexane skeleton cis relative to the protected amino group in the C-2 carbamate.

It was expected that introduction of a hydroxy group trans relative to the amino group in C-2 on the cyclohexanecarboxylic amino ester moiety would be possible without changing the reaction conditions or the oxidizing agent, but starting from the Boc-protected lactam **11**. In lactam **11**, the *N*-substituted imide moiety would not exert the cis stereodirecting effect observed earlier in the transition state of the epoxidation reaction. For this reason Boc-protected lactam **11** was treated with *m*-chloroperbenzoic acid at 0 °C for 5 h (Scheme 5). In this case, due to the presence of the bulky Boc group, *trans*-epoxide **12** was formed exclusively (yield 67%). Interestingly, when the reaction was performed at room temperature for 12 h, a mixture of *trans*- and *cis*-epoxides **12** and **12a** was detected in a 2:1 ratio (Scheme 5). The COSY connectivity pattern accounts for the formation of the epoxy ring in the indicated position for both **12** and **12a**. The scalar couplings do not readily allow the stereochemical assignment. Nevertheless, quantitative analysis of the NOESY crosspeak intensities of H-2–H-3$_{eq}$ and H-2–H-3$_{eq}$ revealed intensity ratios of 1.26 and 1.89 for **12** and **12a**, respectively. The ab initio modeling of the rigid ring systems led to calculated NOE intensities of 1.28 and 1.99 for the trans and cis orientations of the epoxy ring relative to the lactam ring, respectively. The stereochemical assignment (**12**: *trans*, **12a**: *cis*) is supported by the significant downfield shifts ($\Delta\delta$ ca. +0.35 ppm) observed for H-3$_{ax}$ and H-6$_{ax}$ in **12**, caused by the shielding effect of the vicinal epoxy oxygen in the axial position. The latter ring opening of **12** unequivocally proved its stereochemistry.

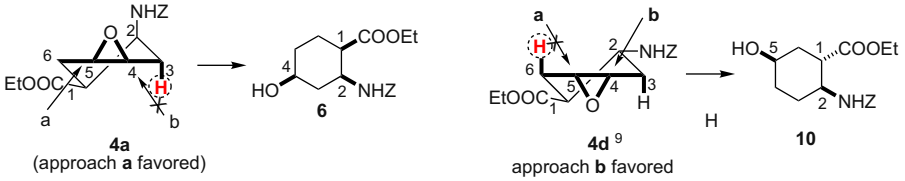
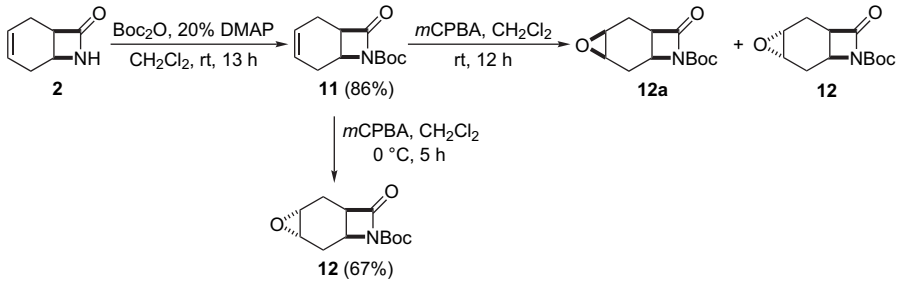
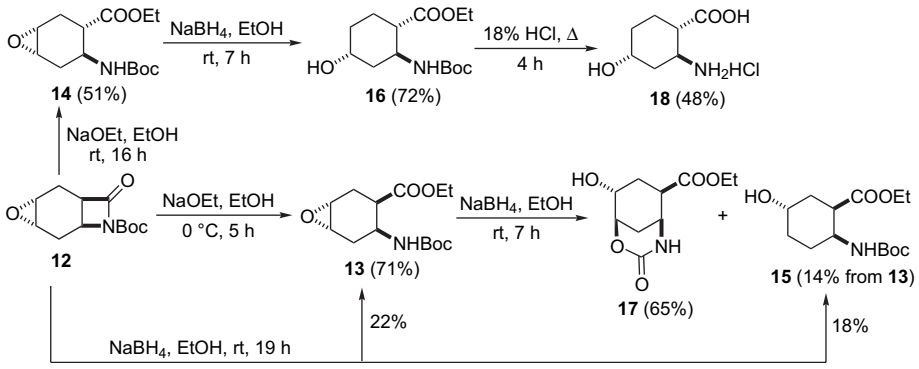


Figure 2. Reductive opening of the oxirane ring in *cis*-amino ester **4a** and *trans*-amino ester **4d**.



Scheme 5. Epoxidation of the Boc-protected β-lactam **11**.



Scheme 6. Formation of hydroxylated amino esters **15** and **16** and amino acid **18**.

Opening of the lactam ring in **12** in the presence of 1.2 equiv of NaOEt in EtOH at room temperature for 16 h led to isomerization at C-1, giving epoxy ester **14** in 51% yield (Scheme 6). Isomerization at C-1 was avoided by carrying out the reaction at 0 °C for 5 h, resulting in epoxy ester **13** (71% yield). Subsequently, reductive opening of the oxirane ring of **14** with $NaBH_4$ in EtOH at room temperature for 7 h afforded 4-hydroxylated amino ester **16** regioselectively in 72% yield. Reductive oxirane ring opening for epoxide **13** was performed under similar conditions as for **14** but the corresponding hydroxylated amino ester **15** was formed only in low yield (18%). The main product of this reaction was oxazinone derivative **17**, formed in good yield, which probably involves attack of the carbonyl oxygen at C-4 of the oxirane ring (Scheme 6). For **13**, the conformation of the cyclohexane scaffold is the same as that for **4a**. A small coupling is measured between H-1_{ax} and H-2_{eq} (3.1 Hz), while a large coupling (9.2 Hz) and a weak NOE are observed between H-1_{ax} and H-6_{ax}. The NOE intensities for H-5_{eq}–H-6_{eq} and H-5_{eq}–H-6_{ax} are the same, which indicates that H-5_{eq} bisects the dihedral. H-4 exhibits a larger NOE toward H-3_{eq} than H-3_{ax}. These observations are in good accord with the trans orientation of the epoxy ring relative to the protected amino group.

For 2-*N*-Boc-amino-4-hydroxy ester **16**, the COSY connectivity pattern unequivocally proves position 4 for the hydroxyl group. The trans-diequatorial arrangement of the protected amino and carboxyl groups is indicated by the large vicinal coupling (11.5 Hz) between the trans-diaxial H-1_{ax} and H-2_{ax}. The scalar couplings and the NOE interactions H-1_{ax}–H-3_{ax} and H-1_{ax}–H-5_{ax} show that the cyclohexane ring attains a stable chair conformation. Only one large vicinal coupling (>11 Hz) was observed around H-3_{ax}, strongly supporting the axial orientation of the hydroxyl group, which is possible only with a trans arrangement relative to the protected amino group.

The regioselectivity of the oxirane ring opening in **13** and **14** can be modeled analogously to that presented for epoxides **4a** and **4d**.

The stereochemistry of the opening of the epoxide in **13** is governed by the axial arrangement of H-6_{ax}, which does not favor attack according to *approach* **a** (Fig. 3). With the favored *approach* **b**, formation of 5-hydroxy-substituted ester **15** was observed (Fig. 3). In the case of epoxide **14**, hydride attack at position 5 is favored from the less hindered face (6-H_{eq}, *approach* **a** in Fig. 3), furnishing hydroxylated ester **16**.

According to this modeling, the formation of oxazinone derivative **17** can be readily understood. While the NHBoc and ester groups in *trans*-amino epoxide **14** are presumably equatorially oriented, in *cis*-derivative **13** the axial NHBoc (through its carbonyl oxygen) attacks the oxirane ring at C-4, forming the bicyclic **17** (Fig. 4).

It was expected that formation of oxazinone derivative **17** could be avoided by opening of the oxirane ring in lactam **12**. For this reason lactam **12** was treated with $NaBH_4$ in EtOH at room temperature (Scheme 6). After stirring for 19 h at room temperature, the formation of epoxide **13** (22%) and 5-hydroxylated ester **15** (18%) was observed, together with a large amount of unreacted material. Simultaneous hydrolysis of the ester and deprotection of the amino group of **18** by treatment with 18% HCl under reflux for 4 h led to (*r*-1,*t*-2,*c*-4)-2-amino-4-hydroxycyclohexanecarboxylic acid hydrochloride **18** (Scheme 6). Unfortunately, not only was hydroxylated ester **15** formed in only low yield (Scheme 6), but its hydrolysis and deprotection under the same conditions as for **16** failed, instead giving a complex mixture, probably involving

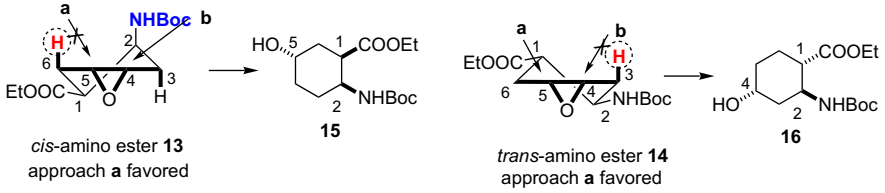


Figure 3. Reductive opening of the oxirane ring in *cis*-amino ester **13** and *trans*-amino ester **14**.

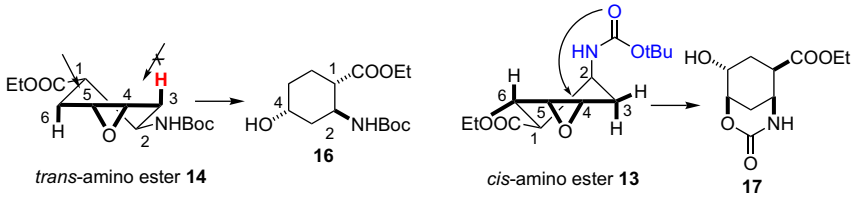
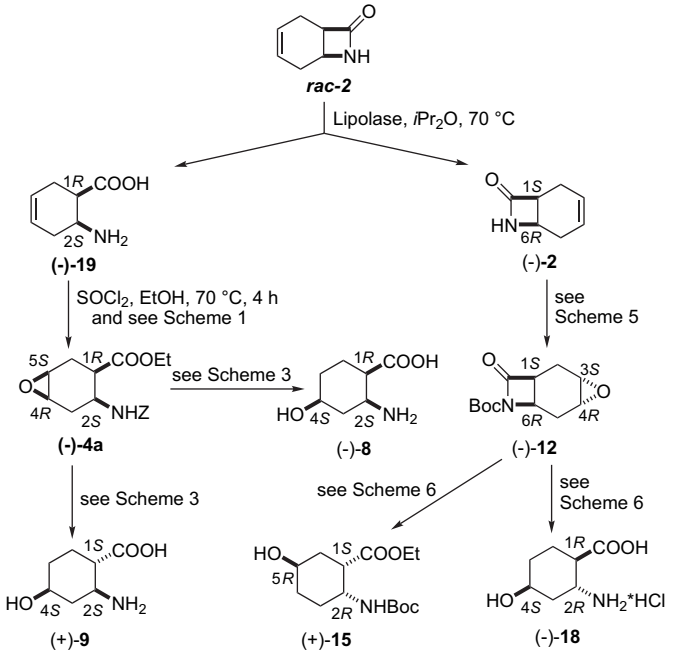


Figure 4. Reductive opening of the oxirane ring of *trans*-amino ester **14** and opening of the oxirane ring mediated by the Boc group in *cis*-amino ester **13**.

different elimination reactions. For **15**, the scalar coupling pattern indicates that the hydroxyl group occupies position 5. The highest coupling constant is estimated as <5 Hz around H-1, showing its equatorial position, while a single large coupling is measured for H-2 (9.4 Hz), pointing to its axial orientation. These findings prove the cis arrangement of the protected amino and carboxylic groups. ^3J(H-5_{ax},H-6_{ax})=9.6 Hz suggests an equatorial orientation for the hydroxyl group, which supports trans stereochemistry relative to the protected amino group. For **18**, the COSY spectrum unequivocally proves position 4 for the hydroxyl group. The scalar couplings point to a stable chair conformation with a trans-diequatorial orientation of the amino and carboxyl groups (^3J(H-1_{ax},H-2_{ax})=11.5 Hz). No large coupling was observed for H-4, while both H-3_{ax} and H-5_{ax} exhibit two large couplings (>12 Hz), which proves the axial orientation for the hydroxyl group (trans to the amino group).

Epoxidation and reductive oxirane ring opening were also performed for enantiomerically enriched substances (Scheme 7). The gram-scale resolution of (±)-**2** was performed with H_2O (0.5 equiv) in the presence of Lipolase [lipase B from *Candida antarctica*, produced by submerged fermentation of a genetically modified *Aspergillus oryzae* microorganism and adsorbed on a macroporous resin, was from Sigma–Aldrich (Catalog no. L4777); 30 mg mL^{-1}] in ^{i}Pr$_2$O at 65 °C, using a slightly modified literature procedure.[13] The enantioselective (E>200) ring cleavage resulted in (−)-**19** (ee=99%, yield=44%) and (−)-**2** (ee=95%, yield=46%), which could be easily separated.



Scheme 7. Synthesis of the corresponding chiral hydroxylated compounds **8**, **9**, **15**, and **18**.

3. Conclusion

In summary, a novel and simple approach to hydroxy-functionalized β-aminocyclohexanecarboxylic acid derivatives has been investigated, starting from the readily available 1,4-cyclohexadiene, based on diastereoselective epoxidation reactions with opposite diastereoselectivity and hydroxylation involving regioselective opening of the oxirane ring. Racemic and enantiomerically enriched β-amino-hydroxycarboxylic acids could be synthesized by this route.

4. Experimental

4.1. General

Melting points were determined with a Kofler apparatus. NMR spectra were recorded on a Bruker DRX 400 spectrometer. Chemical shifts are given in parts per million relative to TMS as internal standard with $CDCl_3$ or D_2O or DMSO as solvents. The reagents and solvents were used as received from the supplier. Optical rotations were measured with a Perkin–Elmer 341 polarimeter.

Ethyl *cis*-2-(*tert*-butoxycarbonylamino)-4-cyclohexenecarboxylate **3c** was prepared according to a known procedure.[8a] Gram-scale resolution of 7-azabicyclo[4.2.0]oct-3-en-8-one, (±)-**2**: crystalline racemic **2** (6 g, 48.8 mmol) was dissolved in ^{i}Pr$_2$O (180 mL). Lipolase (5.4 g, 30 mg mL^{-1}) and H_2O (0.43 mL, 24.4 mmol) were added and the mixture was shaken in an incubator shaker at 65 °C for 51 h. The reaction was stopped by filtering off the enzyme at 49% conversion. The solvent was evaporated off and the residue (−)-**2** crystallized out [2.76 g, 46%; $[\alpha]_D^{25}$ −26.8 (*c* 0.4, $CHCl_3$); mp 150–153 °C (recrystallized from ^{i}Pr$_2$O); ee 95%]. The enzyme was filtered off and washed with distilled H_2O (3×30 mL), and the H_2O was evaporated off in vacuo, yielding crystalline β-amino acid (−)-**19** [2.92 g, 44%; $[\alpha]_D^{25}$ −38.6 (*c* 0.33, H_2O); mp 232–234 °C [recrystallized from H_2O/Me_2CO]; ee 99%]. The ^{1}H NMR data for (−)-**2** and (−)-**19** are similar to those in the literature.[13]

4.1.1. Ethyl cis-2-(benzyloxycarbonylamino)-4-cyclohexenecarboxylate (*3a*)

A solution of lactam **2**[11] (6 g, 48.8 mmol) was treated with HCl/EtOH (25 mL) at 0 °C for 30 min. The mixture was treated with Et_2O (100 mL), the crystals formed were filtered off, and the crude product was crystallized from EtOH/Et_2O (1:1) to give ethyl *cis*-2-amino-4-cyclohexenecarboxylate hydrochloride.

Yield: 8.4 g, 84%; a white solid; mp 93–95 °C. ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.28 (t, J=7.1 Hz, 3H, CH_3), 2.40–2.73 (m, 4H, CH_2), 3.21–3.24 (m, 1H, H-1), 3.82–3.87 (m, 1H, H-2), 4.20–4.26 (m, 2H, OCH_2), 5.59–5.64 (m, 1H, H-5), 5.72–5.76 (m, 1H, H-4), 8.52 (br s, 3H, N–H). Anal. Calcd for $C_9H_{16}ClNO_2$ (205.7): C, 52.56; H, 7.84; N, 6.81. Found: C, 52.91; H, 7.46; N, 6.49.

To a solution of ethyl 2-amino-4-cyclohexenecarboxylate hydrochloride (6 g, 29.23 mmol) and Et_3N (9 mL, 89 mmol) in THF (160 mL), benzyl chloroformate (7.5 g, 44 mmol) was added at 0 °C. After stirring for 16 h, the mixture was taken up in EtOAc (200 mL), washed with H_2O (3×100 mL), dried (Na_2SO_4), concentrated under reduced pressure, and the residue was crystallized from *n*-hexane.

Yield: 7.6 g, 86%; a white solid; mp 59–60 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.24 (t, J=7.10 Hz, 3H, CH_3), 2.18–2.23 (m, 1H, CH_2), 2.31–2.40 (m, 2H, CH_2), 2.49–2.54 (m, 1H, CH_2), 2.80–2.81 (m, 1H, H-1), 4.12–4.16 (m, 2H, OCH_2), 4.25–4.28 (m, 1H, H-2), 5.08 (s, 2H, OCH_2), 5.41–5.45 (br s 1H, N–H), 5.58–5.62 (m, 1H, H-5), 5.64–5.68 (m, 1H, H-4), 7.28–7.36 (m, 5H, Ar–H). IR (KBr): ν_{max} 3346, 2972, 2926, 1726, 1716, 1539. Anal. Calcd for $C_{17}H_{21}O_4N$ (303.3): C, 67.31; H, 6.98; N, 4.62. Found: C, 67.01; H, 6.59; N, 4.22.

4.1.2. Benzyl cis-2-(benzyloxycarbonylamino)-4-cyclohexenecarboxylate (*3b*)

To a solution of *cis*-2-amino-4-cyclohexenecarboxylic acid[12] (11 g, 78 mmol) in 2 M NaOH (45 mL), benzyl chloroformate (13.5 mL, 78 mmol) and 2 M NaOH (45 mL) were added simultaneously dropwise at −10 °C over a period of 30 min. The mixture was stirred at 0 °C for 6 h. HCl (10%) was then added until pH 3 was attained. The mixture was extracted with EtOAc (3×70 mL) and the organic layer was washed with H_2O, dried (Na_2SO_4), and concentrated, giving a white solid with mp 135–136 °C (*n*-hexane). This compound (8 g, 29.1 mmol) was dissolved in THF (130 mL), and DBU (6.6 mL, 43.6 mmol) and benzyl bromide (4.8 mL, 40 mmol)

were then added. The mixture was stirred under reflux for 2 h and 10% HCl was added dropwise until neutral pH was attained. It was next diluted with EtOAc (250 mL), washed with brine, dried (Na_2SO_4), concentrated under reduced pressure, and the residue was crystallized from *n*-hexane.

Yield: 21.6 g, 76%; a white solid; mp 69–70 °C. ^{1}H NMR (400 MHz, $CDCl_3$,): δ 2.14–2.21 (m, 1H, CH_2), 2.33–2.40 (m, 2H, CH_2), 2.51–2.59 (m, 1H, CH_2), 2.86–2.90 (m, 1H, H-1), 4.28–4.31 (m, 1H, H-2), 5.02–5.16 (m, 4H, OCH_2), 5.39 (br s, 1H, N–H), 5.58–5.63 (m, 1H, H-5), 5.65–5.70 (m, 1H, H-4), 7.28–7.38 (m, 10H, Ar–H). IR (KBr): ν_{max} 3347, 3033, 2922, 1710, 1683, 1532. Anal. Calcd for $C_{22}H_{23}NO_4$ (365.4): C, 72.31; H, 6.34; N, 3.83. Found: C, 72.80; H, 6.57; N, 3.70.

4.2. General procedure for epoxidation of amino esters 3a, 3b, and 3c or β-lactam 13 with *m*-chloroperbenzoic acid

To a solution of amino ester **3a**, **3b**, **3c** or β-lactam **13** (36 mmol) in CH_2Cl_2 (200 mL), *m*-CPBA (43 mmol) was added at 0 °C. After stirring for 5 h, further CH_2Cl_2 (150 mL) was added and the mixture was washed with saturated aqueous $NaHCO_3$ (3×150 mL). The organic layer was dried (Na_2SO_4) and concentrated under reduced pressure. The crude product was chromatographed over silica gel (*n*-hexane/EtOAc 3:1).

4.2.1. Ethyl (1R,2S*,4R*,5S*)-2-(benzyloxycarbonylamino)-4,5-epoxycyclohexanecarboxylate (**4a**)*

Yield: 6.8 g, 59%; a white solid; mp 64–65 °C. ^{1}H NMR (400 MHz, DMSO): δ 1.15 (t, *J*=7.1 Hz, 3H, CH_3), 2.03–2.19 (m, 3H, H3, $H6_{eq}$), 2.35 (dd, *J*=7.8, 15.7 Hz, 1H, $H6_{ax}$), 2.58 (td, *J*=7.0, 3.2 Hz, 1H, H1), 3.14–3.19 (m, 2H, H4, H5), 3.88–3.96 (m, 1H, H2), 4.01 (q, *J*=7.1 Hz, 2H, CH_2), 4.99 (s, 2H, OCH_2), 6.23 (d, *J*=9.0 Hz, 1H, NH), 7.25–7.38 (m, 5H, Ar). ^{13}C NMR (100 MHz, DMSO): δ 14.2, 23.9, 28.4, 39.7, 46.2, 51.2, 51.9, 60.4, 65.7, 128.0, 128.7, 129.2, 137.8, 155.4, 172.8. IR (KBr): ν_{max} 3418, 2993, 2952, 1733, 1726, 1511. Anal. Calcd for $C_{17}H_{21}NO_5$ (319.3): C, 63.94; H, 6.63; N, 4.39. Found: C, 63.57; H, 6.36; N, 4.01.

4.2.2. Benzyl (1R,2S*,4R*,5S*)-2-(benzyloxycarbonylamino)-4,5-epoxycyclohexanecarboxylate (**4b**)*

Yield: 8.9 g, 65%; a white solid; mp 69–70 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 2.11 (ddd, *J*=3.2, 5.9, 15.6 Hz, 1H, $H6_{eq}$), 2.16–2.27 (m, 2H, H3), 2.53 (td, *J*=6.6, 3.1 Hz 1H, H1), 2.65 (dd, *J*=7.6, 15.6 Hz, 1H, $H6_{ax}$), 3.16–3.22 (m, 2H, H4, H5), 4.11–4.19 (m, 1H, H2), 5.00–5.17 (m, 4H, 2×OCH_2), 5.77 (d, *J*=9.9 Hz, 1H, NH), 7.27–7.40 (m, 10H, Ar). ^{13}C NMR (100 MHz, $CDCl_3$): δ 25.1, 29.7, 41.1, 46.7, 51.5, 52.2, 67.3, 67.4, 128.66, 128.69, 128.9, 129.0, 129.1, 129.2, 136.5, 137.2, 156.3, 173.2. IR (KBr): ν_{max} 3436, 3007, 2947, 1724, 1719, 1507. Anal. Calcd for $C_{22}H_{23}NO_5$ (381.4): C, 69.28; H, 6.08; N, 3.67. Found: C, 68.93; H, 5.79; N, 3.32.

4.2.3. Ethyl (1R,2S*,4R*,5S*)-2-(tert-butoxycarbonylamino)-4,5-epoxycyclohexanecarboxylate (**4c**)*

Yield: 5.8 g, 57%; a white solid; mp 58–60 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.27 (t, *J*=7.10 Hz, 3H, CH_3), 1.41 (s, 9H, ^{t}Bu), 2.09 (ddd, *J*=3.2, 6.2, 15.7 Hz, 1H, H6), 2.15–2.20 (m, 2H, H3), 2.46 (ddd, *J*=3.2, 6.3, 7.4 Hz, 1H, H1), 2.62 (dd, *J*=7.5, 15.7 Hz, 1H, CH_2), 3.17–3.20 (m, 2H, H4, H5), 4.01–4.10 (m, 1H, H2), 4.14 (q, *J*=7.10 Hz, 2H, OCH_2), 5.50 (d, *J*=9.8 Hz, 1H, NH). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.8, 25.3, 29.0 (3C), 29.8, 41.1, 46.0, 51.6, 52.3, 61.4, 79.8, 155.8, 173.5. IR (KBr): ν_{max} 3433, 2979, 1723, 1720, 1499. Anal. Calcd for $C_{14}H_{23}NO_5$ (285.3): C, 58.93; H, 8.12; N, 4.91. Found: C, 58.63; H, 7.83; N, 4.72.

4.2.4. Ethyl (1S,2S*,4R*,5S*)-2-(benzyloxycarbonylamino)-4,5-epoxycyclohexanecarboxylate (**4d**)*

Yield: 7.1 g, 62%; a white solid; mp 55–58 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.22 (t, *J*=7.10 Hz, 3H, CH_3), 1.90–1.97 (m, 1H, CH_2), 2.09–2.32 (m, 3H, CH_2), 2.62–2.69 (m, 1H, H-1), 3.16–3.27 (m, 2H, H-4 and H-5), 4.07–4.16 (m, 3H, H-2 and OCH_2), 5.07 (s, 2H, OCH_2), 5.40 (br s, 1H, NH), 7.30–7.34 (m, 5H, Ar–H). ^{13}C NMR ($CDCl_3$): 20.6, 24.6, 29.2, 41.8, 47.2, 51.8, 52.2, 61.6, 67.3, 128.7, 129.1, 137.1, 156.0, 173.6. IR (KBr): ν_{max} 3312, 2982, 2917, 1721, 1687, 1544. Anal. Calcd for $C_{17}H_{21}NO_5$ (319.3): C, 63.94; H, 6.63; N, 4.39. Found: C, 63.66; H, 6.31; N, 4.18.

4.3. General procedure for reductive opening of epoxides 4a, 14, and 21

To a solution of epoxide (3.2 mmol) in EtOH (15 mL), $NaBH_4$ (6.3 mmol) was added in several portions. The mixture was stirred at the temperature and for the time indicated, saturated NH_4Cl/H_2O (1 mL) was then added, and most of the solvent was evaporated off. The residue was taken up in EtOAc (25 mL), washed with H_2O, dried (Na_2SO_4), and concentrated. The crude oily product was chromatographed over silica gel (*n*-hexane/EtOAc 1:2).

4.3.1. Ethyl (1R,2S*,4S*)-2-(benzyloxycarbonylamino)-4-hydroxycyclohexanecarboxylate (**6**)*

Yield: 720 mg, 70%; a white solid; mp 50–53 °C. ^{1}H NMR (400 MHz, DMSO): δ 1.14 (t, *J*=7.2 Hz, 3H, CH_3), 1.41–1.63 (m, 3H, H3, H6), 1.69–1.80 (m, 2H, H5), 1.85–1.95 (m, 1H, H6), 2.68–2.75 (m, 1H, H1), 3.63–3.72 (m, 1H, H4), 3.77–3.89 (m, 1H, H2), 4.00 (q, *J*=7.2 Hz, 1H, CH_2), 4.97–5.20 (m, 2H, OCH_2), 6.89 (d, *J*=8.4 Hz, 1H, NH), 7.27–7.39 (m, 5H, Ar). ^{13}C NMR (100 MHz, DMSO): δ 14.8, 21.8, 31.5, 37.3, 44.1, 48.3, 60.5, 66.1, 67.0, 128.5, 128.6, 129.1, 138.0, 155.9, 173.5. IR (KBr): ν_{max} 3396, 2941, 1724, 1716, 1513. Anal. Calcd for $C_{17}H_{23}NO_5$ (321.4): C, 63.54; H, 7.21; N, 4.36. Found: C, 63.13; H, 7.01; N, 4.02.

4.3.2. Ethyl (1S,2S*,4S*)-2-(benzyloxycarbonylamino)-4-hydroxycyclohexanecarboxylate (**7**)*

Yield: 575 mg, 56%; a white solid; mp 52–55 °C. ^{1}H NMR (400 MHz, DMSO): δ 1.01–1.07 (m, 1H, $H5_{ax}$), 1.09 (t, *J*=7.1 Hz, 3H, CH_3), 1.19 (q, *J*=11.8 Hz, 1H, $H3_{ax}$), 1.38 (dq, *J*=4.0, 13.2 Hz, 1H, $H6_{ax}$), 1.71–1.85 (m, 2H, $H5_{eq}$, $H6_{eq}$), 1.88–1.98 (m, 1H, $H3_{eq}$), 2.19 (dt, *J*=3.4, 11.6 Hz, 1H, H1), 3.37–3.49 (m, 1H, H4), 3.57 (dq, *J*=3.9, 10.8 Hz, 1H, H2), 3.97 (q, *J*=7.1 Hz, 2H, OCH_2), 4.71 (d, *J*=4.5 Hz, 1H, OH), 4.97 (br s, 2H, OCH_2), 7.25–7.38 (m, 6H, NH, Ar). ^{13}C NMR (100 MHz, DMSO): δ 14.8, 26.4, 34.3, 42.5, 48.9, 50.7, 60.6, 65.9, 67.9, 128.4, 128.6, 129.2, 138.1, 156.0, 174.4. IR (KBr): ν_{max} 3339, 2938, 1722, 1712, 1544. Anal. Calcd for $C_{17}H_{23}NO_5$ (321.4): C, 63.55; H, 7.21; N, 4.36. Found: C, 63.19; H, 7.10; N, 4.08.

4.4. General procedure for alkaline hydrolysis and deprotection of esters 6 and 7

To a solution of amino ester **6** or **7** (1.92 g, 6 mmol) in MeOH (40 mL), NaOH (0.72 g, 18 mmol) in H_2O (12 mL) was added. After stirring for 5 h, 10% HCl was added at 0 °C until pH 5, after which the mixture was extracted with $CHCl_3$ (3×70 mL). The combined organic layers were dried (Na_2SO_4) and concentrated, and the products were used without further purification.

A mixture of a solution of benzyloxycarbonylamino acid obtained above (440 mg, 1.5 mmol) in MeOH (15 mL) and 10% Pd/C (80 mg) was stirred under H_2 at atmospheric pressure for 2 h. The Pd/C was filtered off and the filtrate was concentrated under reduced pressure. The residue was crystallized from MeOH/Et_2O (1:1).

4.4.1. (1R,2S*,4S*)-2-Amino-4-hydroxycyclohexanecarboxylic acid (**8**)*

Yield: 207 mg, 87%; white crystals; mp 222–225 °C. ^{1}H NMR (400 MHz, DMSO): δ 1.14–1.28 (m, 2H, $H5_{eq}$, $H6_{ax}$), 1.41 (q, *J*=10.8 Hz, 1H, $H3_{ax}$), 1.50–1.58 (m, 1H, $H5_{ax}$), 1.90 (d, *J*=11.8 Hz, 1H,

$H3_{eq}$), 2.05–2.15 (m, 1H, $H6_{eq}$), 2.18–2.23 (m, 1H, H1), 3.13 (td, J=4.0, 10.3 Hz, H2), 3.43–3.52 (m, 1H, H4). ^{13}C NMR (100 MHz, DMSO): δ 23.6, 31.9, 37.3, 41.6, 48.9, 67.2, 176.0. IR (KBr): ν_{max} 3132, 3117, 1598, 1455. Anal. Calcd for $C_7H_{13}NO_3$ (159.2): C, 52.82; H, 8.23; N, 8.80. Found: C, 52.44; H, 8.02; N, 8.48.

4.4.2. (1S,2S*,4S*)-2-Amino-4-hydroxycyclohexanecarboxylic acid (**9**)*

Yield: 203 mg, 85%; white crystals; mp 256–258 °C. ^1H NMR (400 MHz, D_2O): δ 1.22–1.48 (m, 3H, $H3_{ax}$, $H5_{ax}$, $H6_{ax}$), 2.00 (d, J=11.7 Hz, 1H, $H5_{eq}$), 2.10–2.22 (m, 2H, H1, $H6_{eq}$), 2.24–2.31 (m, 1H, $H3_{eq}$), 3.30 (dt, J=3.9, 11.5 Hz, 1H, H2), 3.66–3.74 (m, 1H, H4). ^{13}C NMR (100 MHz, D_2O): δ 26.4, 33.5, 37.9, 47.8, 50.7, 67.9, 180.3. IR (KBr): ν_{max} 3146, 2935, 1584, 1519. Anal. Calcd for $C_7H_{13}NO_3$ (159.2): C, 52.82; H, 8.23; N, 8.80. Found: C, 52.47; H, 8.03; N, 8.55.

*4.4.3. 7-(tert-Butoxycarbonyl)azabicyclo[4.2.0]oct-3-en-8-one (**11**)*

A solution of β-lactam **2** (6 g, 48.8 mmol), 4-dimethylaminopyridine (DMAP) (1.19 g, 9.7 mmol), and Boc_2O (15.5 g, 73.2 mmol) in CH_2Cl_2 (100 mL) was stirred at room temperature for 13 h. The mixture was then diluted with CH_2Cl_2 (140 mL) and washed with H_2O (3×150 mL). The organic layer was dried (Na_2SO_4) and the residue was purified by column chromatography (silica gel, *n*-hexane/EtOAc 2:1).

Yield: 9.3 g, 86%; a white solid; mp 63–64 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 1.52 (s, 9H, CH_3), 2.12–2.18 (m, 2H, CH_2), 2.45–2.53 (m, 1H, CH_2), 2.75–2.83 (m, 1H, CH_2), 3.36–3.41 (m, 1H, H-1), 4.23–4.28 (m, 1H, H-2), 5.75–5.79 (m, 1H, CH), 5.84–5.90 (m, 1H, CH). IR (KBr): ν_{max} 1798, 1720, 1343. Anal. Calcd for $C_{12}H_{17}NO_3$ (223.3): C, 64.55; H, 7.67; N, 6.27. Found: C, 64.19; H, 8.01; N, 6.01.

4.4.4. (1R,3S*,4R*,6S*)-3,4-Epoxy-7-tert-butoxycarbonyl-azabicyclo[4.2.0]octan-8-one (**12a**)*

Yield: 5.7 g, 67%; a white solid; mp 105–106 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 1.53 (s, 9H, ^{t}Bu), 2.03–2.13 (m, 2H, $H3_{eq}$, $H6_{eq}$), 2.63 (dd, J=3.3, 16.0 Hz, 1H, $H6_{ax}$), 2.94 (dd, J=3.2, 16.7 Hz, 1H, $H3_{ax}$), 3.13 (t, J=7.1 Hz, 1H, H1), 3.19 (t, J=3.8 Hz, 1H, H4), 3.23 (t, J=3.7 Hz, 1H, H5), 4.02 (t, J=6.4 Hz, 1H, H2). ^{13}C NMR (100 MHz, $CDCl_3$): δ 21.6, 22.7, 28.8 (3C), 43.6, 48.2, 50.7, 51.3, 83.4, 148.5, 168.2. IR (KBr): ν_{max} 2977, 1804, 1701, 1335. Anal. Calcd for $C_{12}H_{17}NO_4$ (239.3): C, 60.24; H, 7.16; N, 5.85. Found: C, 59.89; H, 6.90; N, 5.55.

4.4.5. (1R,3R*,4S*,6S*)-3,4-Epoxy-7-tert-butoxycarbonyl-azabicyclo[4.2.0]octan-8-one (**12**)*

Yield: 2.6 g, 30%; a white solid; mp 106–108 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 1.52 (s, 9H, ^{t}Bu), 2.14 (ddd, J=1.5, 6.8, 15.3 Hz, 1H, $H3_{eq}$), 2.19–2.35 (m, 2H, $H6_{ax}$, $H6_{eq}$), 2.60 (ddd, J=4.0, 7.0, 15.3 Hz, 1H, $H3_{ax}$), 3.19 (dt, J=1.6, 4.0 Hz, 1H, H4), 3.22–3.30 (m, 2H, H5, H1), 4.00 (q, J=6.8 Hz, 1H, H2). ^{13}C NMR (100 MHz, $CDCl_3$): δ 21.3, 26.6, 28.9 (3C), 44.1, 47.9, 49.1, 49.7, 84.0, 148.5, 168.0. IR (KBr): ν_{max} 2973, 1810, 1705, 1303. Anal. Calcd for $C_{12}H_{17}NO_4$ (239.3): C, 60.24; H, 7.16; N, 5.85. Found: C, 59.87; H, 6.97; N, 5.62.

4.5. General procedure for ring opening reaction of lactam 12

To a solution of 2-azetidinone **12** (2.4 g, 10 mmol) in anhydrous EtOH (50 mL), NaOEt (1.2 equiv) was added and the mixture was stirred for the time and at the temperature indicated. It was then diluted with EtOAc (100 mL), washed with H_2O, dried (Na_2SO_4), and concentrated. The crude oily product was chromatographed over silica gel (*n*-hexane/EtOAc 2:1).

4.5.1. Ethyl (1R,2S*,4S*,5R*)-2-(tert-butoxycarbonylamino)-4,5-epoxycyclohexanecarboxylate (**13**)*

Yield: 2 g, 71%; a white solid; mp 96–98 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 1.20 (t, J=7.1 Hz, 3H, CH_3), 1.37 (s, 9H, ^{t}Bu), 1.87 (td, J=4.7, 15.5 Hz, 1H, $H3_{eq}$), 1.94 (dd, J=5.5, 15.7 Hz, 1H, $H6_{eq}$), 2.02 (dd, J=5.8, 15.5 Hz, 1H, $H3_{ax}$), 2.22 (ddd, J=2.6, 8.9, 15.7 Hz, 1H, $H6_{ax}$), 2.51–2.57 (m, 1H, H1), 3.10 (t, J=3.7 Hz, 1H, H4), 3.13–3.16 (m, 1H, H5), 3.81–3.88 (m, 1H, H2), 3.99–4.11 (m, 4H, 2×OCH_2), 6.88 (d, J=8.0 Hz, 1H, NH). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.2, 22.9, 28.5 (3C), 30.0, 39.4, 44.3, 50.6, 51.5, 60.4, 60.7, 153.4, 172.7. IR (KBr): ν_{max} 3270, 2925, 1737, 1680. Anal. Calcd for $C_{14}H_{23}NO_5$ (285.3): C, 58.93; H, 8.12; N, 4.91. Found: C, 58.57; H, 7.88; N, 4.60.

4.5.2. Ethyl (1S,2S*,4S*,5R*)-2-(tert-butoxycarbonylamino)-4,5-epoxycyclohexanecarboxylate (**14**)*

Yield: 1.45 g, 51%; a white solid; mp 90–91 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 1.23 (t, 3H, CH_3), 1.41 (s, 9H, CH_3), 2.22–2.54 (m, 5H, H-1, CH_2), 3.13–3.21 (m, 2H, H-4, H-5), 3.82–3.85 (m, 1H, H-2), 4.13–4.16 (m, 2H, OCH_2), 4.65 (br s, 1H, NH). IR (KBr): ν_{max} 3363, 2990, 1728, 1681. Anal. Calcd for $C_{14}H_{23}NO_5$ (285.3): C, 58.93; H, 8.12; N, 4.91. Found: C, 58.62; H, 8.01; N, 4.63.

4.5.3. Ethyl (1R,2S*,5S*)-2-(tert-butoxycarbonylamino)-4-hydroxycyclohexanecarboxylate (**15**)*

Yield: 560 mg, 18%; a white solid; mp 101–103 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 1.28 (t, J=7.1 Hz, 3H, CH_3), 1.39–1.45 (m, 10H, $H4_{ax}$, ^{t}Bu), 1.64 (ddd, J=4.5, 9.5, 13.8 Hz, 1H, $H6_{ax}$), 1.74–1.88 (m, 2H, H3), 1.88–1.96 (m, 1H, $H4_{eq}$), 2.25 (td, J=4.3, 13.8 Hz, 1H, $H6_{eq}$), 2.98 (q, J=4.6 Hz, 1H, H1), 3.68–3.92 (m, 2H, H2, $H5_{ax}$), 4.16 (q, J=7.2 Hz, 2H, OCH_2), 5.3 (br s, 1H, NH). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.9, 27.5, 29.1 (3C), 33.3, 35.8, 49.5, 61.3, 67.0, 156.0, 170.1. IR (KBr): ν_{max} 3364, 3207, 2973, 1724, 1684, 1519. Anal. Calcd for $C_{14}H_{25}NO_5$ (287.4): C, 58.52; H, 8.77; N, 4.87. Found: C, 58.29; H, 8.46; N, 4.51.

4.5.4. Ethyl (1S,2S*,4R*)-2-(tert-butoxycarbonylamino)-4-hydroxycyclohexanecarboxylate (**16**)*

Yield: 410 mg, 72%; a white solid; mp 120–121 °C. ^1H NMR (400 MHz, DMSO): δ 1.16 (t, J=7.1 Hz, 3H, CH_3), 1.24–1.41 (m, 11H, ^{t}Bu, $H3_{ax}$, $H5_{ax}$), 1.49–1.58 (m, 2H, $H5_{eq}$, $H6_{eq}$), 1.69 (td, J=4.0, 13.5 Hz, 1H, $H3_{eq}$), 1.83 (dq, J=4.3, 12.9 Hz, 1H, $H6_{eq}$), 2.23 (dt, J=3.6, 11.2 Hz, 1H, H1), 3.83–3.94 (m, 2H, H2, H4), 3.95–4.10 (m, 2H, OCH_2), 4.48 (d, J=2.9 Hz, 1H, OH), 6.66 (d, J=9.6 Hz, 1H, NH). ^{13}C NMR (100 MHz, DMSO): δ 14.4, 22.9, 28.7 (3C), 31.1, 39.3, 46.2, 49.2, 60.0, 64.5, 77.6, 152.9, 173.9. IR (KBr): ν_{max} 3528, 3354, 2986, 1708, 1686. Anal. Calcd for $C_{14}H_{25}NO_5$ (287.4): C, 58.52; H, 8.77; N, 4.87. Found: C, 58.20; H, 8.42; N, 4.54.

4.5.5. (1S,2S*,4R*)-2-Amino-4-hydroxycyclohexanecarboxylic acid hydrochloride (**18**)*

Ester **16** (786 mg, 1.5 mmol) in 18% HCl solution (6 mL) was heated under reflux for 4 h. The reaction mixture was next concentrated under reduced pressure and crystallized from EtOH/Et_2O (1:1).

Yield: 140 mg, 48%; white crystals; mp 199–202 °C. ^1H NMR (400 MHz, D_2O): δ 1.53–1.77 (m, 3H, $H3_{ax}$, $H5_{ax}$, $H6_{ax}$), 1.82 (qd, J=2.8, 13.6 Hz, 1H, $H5_{eq}$), 1.96 (dd, J=4.1, 12.0 Hz, 1H, $H6_{eq}$), 2.11 (d, J=13.6 Hz, 1H, $H3_{eq}$), 2.26 (dt, J=3.8, 11.3 Hz, 1H, H1), 3.54 (dt, J=4.0, 12.0 Hz, 1H, H2), 4.19–4.24 (m, 1H, $H4_{eq}$). ^{13}C NMR (100 MHz, D_2O): δ 23.4, 30.9, 35.7, 48.4 (2C), 65.6, 180.6. IR (KBr): ν_{max} 3323, 2932, 1716, 1503. Anal. Calcd for $C_7H_{14}ClNO_3$ (195.6): C, 42.97; H, 7.21; N, 7.16. Found: C, 42.63; H, 7.01; N, 6.82.

4.5.6. Ethyl (1R,5S*,6R*,8R*)-8-hydroxy-3-oxo-2-oxa-4-azabicyclo[3.3.1]nonane-6-carboxylate (**17**)*

Yield: 268 mg, 65%; a white solid; mp 138–141 °C ^1H NMR (400 MHz, DMSO): δ 1.20 (t, J=7.1 Hz, 3H, CH_3), 1.65–1.80 (m, 3H, H3, $H6_{ax}$, $H6_{eq}$), 2.12 (d, J=13.2 Hz, 1H, H3), 2.76 (ddd, J=1.3, 5.0, 12.2 Hz, 1H, H1), 3.73–3.78 (m, 1H, H2), 3.87–3.92 (m, 1H, H5), 4.00–4.14 (m, 2H, OCH_2), 4.21–4.27 (m, 1H, H4), 5.21 (d, J=3.8 Hz,

1H, OH), 7.41 (d, J=4.5 Hz, 1H, NH). ^{13}C NMR (100 MHz, DMSO): δ 14.8, 24.5, 26.7, 42.8, 47.2, 61.1, 66.5, 74.8, 153.3, 173.2. IR (KBr): ν_{max} 3270, 2957, 1737, 1678. Anal. Calcd for $C_{10}H_{15}NO_5$ (229.2): C, 52.40; H, 6.60; N, 6.11. Found: C, 52.04; H, 6.26; N, 5.89.

4.6. Synthesis of the enantiomers

For preparation of the optically active compounds, the same procedures were used as for the racemic substances. The NMR spectra of the enantiomers were identical with those of the corresponding racemic compounds.

*4.6.1. Ethyl 1R,2S-2-(benzyloxycarbonylamino)-4-cyclohexenecarboxylate [(−)-**3a**]*
Yield: 82%; mp 58–60 °C; $[\alpha]_D^{25}$ −12.5 (c 0.57, $CHCl_3$).

*4.6.2. Ethyl 1R,2S,4R,5S-2-(benzyloxycarbonylamino)-4,5-epoxycyclohexanecarboxylate [(−)-**4a**]*
Yield: 51%; mp 62–64 °C; $[\alpha]_D^{25}$ −7.1 (c 1.72, $CHCl_3$).

*4.6.3. Ethyl 1R,2S,4S-2-(benzyloxycarbonylamino)-4-hydroxycyclohexanecarboxylate [(−)-**6**]*
Yield: 63%; mp 52–54 °C; $[\alpha]_D^{25}$ −12.5 (c 0.53, $CHCl_3$).

*4.6.4. Ethyl 1S,2S,4S-2-(benzyloxycarbonylamino)-4-hydroxycyclohexanecarboxylate [(+)-**7**]*
Yield: 49%; mp 51–53 °C; $[\alpha]_D^{25}$ +6.8 (c 0.41, $CHCl_3$).

*4.6.5. 1R,2S,4S-2-Amino-4-hydroxycyclohexanecarboxylic acid [(−)-**8**]*
Yield: 87%; mp 220–224 °C; $[\alpha]_D^{25}$ −20.5 (c 0.5, H_2O); ee=96%.

*4.6.6. 1S,2S,4S-2-Amino-4-hydroxycyclohexanecarboxylic acid [(+)-**9**]*
Yield: 78%; mp 254–257 °C; $[\alpha]_D^{25}$ +18.5 (c 1.2, H_2O); ee=97%.

*4.6.7. 1S,6R-7-Benzyloxycarbonylazabicyclo[4.2.0]oct-3-en-8-one [(−)-**11**]*
Yield: 77%; mp 62–65 °C; $[\alpha]_D^{25}$ −81 (c 1.05, $CHCl_3$).

*4.6.8. 1S,3S,4R,6R-3,4-Epoxy-7-benzyloxycarbonyl-azabicyclo[4.2.0]octan-8-one [(−)-**12**]*
Yield: 61%; mp 104–107 °C; $[\alpha]_D^{25}$ −34.5 (c 0.75, $CHCl_3$).

*4.6.9. Ethyl 1S,2R,4R,5S-2-(tert-butoxycarbonylamino)-4,5-epoxycyclohexanecarboxylate [(+)-**13**]*
Yield: 66%; mp 95–97 °C; $[\alpha]_D^{25}$ +19.5 (c 0.55, $CHCl_3$); ee=93%.

*4.6.10. Ethyl 1R,2R,4R,5S-2-(tert-butoxycarbonylamino)-4,5-epoxycyclohexanecarboxylate [(−)-**14**]*
Yield: 44%; mp 90–92 °C; $[\alpha]_D^{25}$ −17.4 (c 0.75, $CHCl_3$); ee=92–93%.

*4.6.11. Ethyl 1S,2R,5R-2-(tert-butoxycarbonylamino)-4-hydroxycyclohexanecarboxylate [(+)-**15**]*
Yield: 16%; mp 101–103 °C; $[\alpha]_D^{25}$ +14.5 (c 0.5, $CHCl_3$).

*4.6.12. Ethyl 1R,2R,4S-2-(tert-butoxycarbonylamino)-4-hydroxycyclohexanecarboxylate [(−)-**16**]*
Yield: 62%; mp 119–121 °C; $[\alpha]_D^{25}$ −10.8 (c 0.33, $CHCl_3$).

*4.6.13. 1R,2R,4S-2-Amino-4-hydroxycyclohexanecarboxylic acid hydrochloride [(−)-**18**]*
Yield: 41%; mp 197–200 °C; $[\alpha]_D^{25}$ −13 (c 0.3, H_2O); ee=92%.

Acknowledgements

We are grateful to the Hungarian Research Foundation (OTKA No. F67970 and T049407) and the National Research and Development Office, Hungary (GVOP-311-2004-05-0255/3.0) for financial support.

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