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# Resolution of *N*-protected *cis*- and *trans*-3-aminocyclohexanols via lipase-catalyzed enantioselective acylation in organic media

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**Abstract**—The enzymatic acylation of *N*-protected *cis*-and *trans*-1,3-aminocyclohexanols using lipases in organic solvents is described. By modifying certain reaction parameters such as the solvent, the lipase and the *N*-protecting group, it is possible to achieve high enantioselectivities and to obtain enantiomerically pure 3-aminocyclohexanols. The influence of the *N*-protecting group and the conformation of the substrate on the reaction rate was also studied.


## 1. Introduction

Possessing two functional groups, 1,3-aminoalcohols are valuable intermediates in organic synthesis. They have found use as chiral auxiliaries and ligands in asymmetric synthesis[1] and are also structural features of many biologically active compounds.[2] For these reasons, the development of new synthetic routes towards 1,3-aminoalcohols in enantiopure form is of great importance.[3]

In the past few years, biocatalysis has emerged as powerful means for the preparation of pure enantiomers either through enzymatic resolution processes or biocatalytic desymmetrization of *meso*-compounds.[4] The majority of the enzyme-catalyzed reactions involve the use of hydrolases. Lipases in particular have proven to be excellent biocatalysts for the resolution of alcohols and amines.[5]

Recently, the chemoenzymatic preparation of mono and bicyclic *cis*- and *trans*-1,3-aminoalcohols has been reported.[6] Depending on the substrates and the reaction conditions, good enantioselectivities were obtained in enzymatic acylation, hydrolysis and alcoholysis processes, including a procedure published very recently for the enzymatic hydrolysis of *trans*-1,3-aminocyclohexanol.[7]

As part of our research on the chemoenzymatic synthesis of cyclic aminoalcohols using lipases,[8] we have focused on the resolution of *cis*- and *trans*-3-aminocyclohexanols either in their free or *N*-protected form. The two functional groups present in these molecules offer two different methods for their biocatalytic preparation. The biocatalytic acylation of the amine group in the free 3-aminocyclohexanol can be achieved using nonactivated esters as the acyl donor. On the other hand, by protecting the amine group, the enzymatic resolution can be carried out on the alcohol moiety.

## 2. Results and discussion

First, we studied the enzymatic *N*-acetylation of unprotected (±)-*cis*-3-aminocyclohexanol, (±)-*cis*-**1**, which was prepared from 2-cyclohexenol by a slight modification of a method described by Sammes and Thetford.[9] Ethyl acetate was used as the acyl donor, since activated esters, such as vinyl acetate, react with the amino group even in the absence of the biocatalyst. Several commercially available lipases were tested under different reaction conditions. However, no product formation could be seen even after prolonged reaction times.

In view of these results, we focused on the resolution of the *N*-protected derivative (±)-*cis*-3-phthalimidocyclohexanol (±)-*cis*-**2a**, an intermediate in the synthesis of (±)-*cis*-**1**, carrying out the enzymatic resolution at the alcohol moiety (Scheme 1). Our initial experiments were

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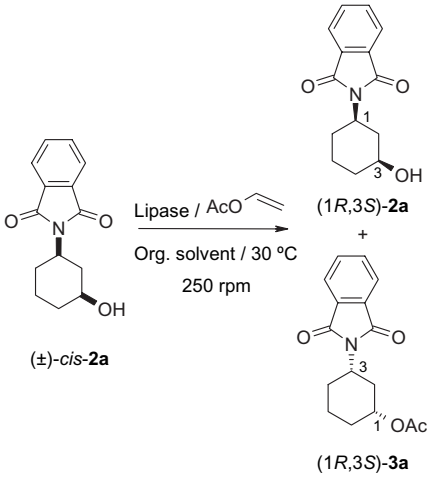



**Scheme 1.** Enzymatic acylation of (±)-*cis*-**2a** in organic solvents.

designed to find the most suitable lipase for catalyzing the acetylation of (±)-*cis*-**2a** using vinyl acetate in $^{t}$BuOMe. As shown in Table 1, all lipases tested, except for PPL, yielded good enantioselectivities, showing similar $E$ values.[10] Lipase B from *Candida antarctica* (CAL-B) exhibited a high reaction rate, giving a conversion of 44% after 1 h (entry 1). The reaction catalyzed by lipase A from *C. antarctica* (CAL-A) was slightly slower (entry 2), while with the two varieties of *Pseudomonas cepacea*, very long reaction times were needed to reach low conversions, especially in the case of the nonimmobilized form of PSL (entries 3 and 4).

When examining the influence of the solvent on the enantioselectivity of the acylation of alcohol (±)-*cis*-**2a** using CAL-B and vinyl acetate, THF turned out to be the best solvent (entry 6). After 3 h, 49% conversion was achieved with an excellent enantioselectivity ($E$ >200). Toluene and vinyl acetate both gave high enantioselectivities and high reaction rates, while 1,4-dioxane showed a decrease in the reaction rate. Very low enantioselectivities were achieved in the resolutions carried out in cyclohexane and $^{i}Pr_2O$.

Next, different lipases were used in THF for the resolution of (±)-*cis*-**2a**. Surprisingly, with CAL-A no reaction was observed even after long reaction times. The highest enantiomeric ratio was recorded for the resolution of (±)-*cis*-**2a** (entry 12) using PSL-C, although the reaction rate was lower than with CAL-B.

After the successful resolution of the *cis*-aminoalcohol, we turned our attention to the *trans* diastereomer (±)-*trans*-**2a**. The substrate was prepared by Mitsunobu reaction followed by hydrolysis of the ester.[11] To our surprise, none of the lipases tested catalyzed the acetylation under various reaction conditions.

In order to complete our studies on the enzymatic acetylation of 1,3-aminoalcohols and to examine the influence of the *N*-protecting group, we decided to prepare the less rigid (±)-*trans N*-benzyloxycarbonylcyclohexanol (±)-*trans*-**2b** from racemic aminoalcohol *trans*-(±)-**2a** by removal of the phthalimide group with hydrazine followed by introduction of a Cbz group. CAL-B and PSL-C were used in THF with vinyl acetate (Table 2). Contrary to the previous finding, lipase B from *C. antarctica* exhibited high activity and yielded 44% conversion in 3 h with an excellent enantioselectivity ($E$ >200) (enrty 1). On the other hand, poor enantioselectivity and low reaction rate were obtained with PSL-C (entry 2). Different behaviour in the reaction rate between diastereomers in the enzymatic transesterification has been previously reported by Tanikaga et al.[12] and by us.[13] In accordance with Tanikaga's reports, we also observed no or very low activity in the transesterification of *cis*-2- and *trans*-3-substituted cyclohexanols with an axial hydroxyl group, which might influence its binding to the active site in the enzyme giving no acylated products. In (±)-*trans*-**2a** the bulky and rigid phthalimide moiety occupies the equatorial position forcing the hydroxyl group into an axial orientation. In the case of the more flexible Cbz group the energy difference between the two conformers of the cyclohexane ring with the hydroxyl group in the axial or equatorial position is sufficiently smaller to allow the

**Table 1.** Lipase-catalyzed acetylation of (±)-*cis*-**2a**[a]

| Entry | Lipase | Solvent | Time (h) | $c$ (%)[b] | Ee (%)[c] (1*R*,3*S*)-**3a** | Ee (%)[c] (1*R*,3*S*)-**2a** | $E$[d] |
|---|---|---|---|---|---|---|---|
| 1 | CAL-B | $^{t}$BuOMe | 1 | 44 | 92 | 73 | 52 |
| 2 | CAL-A | $^{t}$BuOMe | 3 | 18 | 94 | 18 | 40 |
| 3 | PSL-C | $^{t}$BuOMe | 72 | 9 | 96 | 10 | 54 |
| 4 | PSL | $^{t}$BuOMe | 192 | 9 | 96 | 9 | 53 |
| 5 | PPL | $^{t}$BuOMe | 192 | 5 | 90 | 5 | 19 |
| 6 | CAL-B | THF | 3 | 49 | 99 | 95 | >200 |
| 7 | CAL-B | Toluene | 1 | 34 | 96 | 50 | 95 |
| 8 | CAL-B | Vinyl acetate[e] | 3 | 52 | 91 | 99 | 102 |
| 9 | CAL-B | 1,4-Dioxane | 17 | 45 | 92 | 73 | 52 |
| 10 | CAL-B | $^{i}Pr_2O$ | 17 | 29 | 52 | 22 | 4 |
| 11 | CAL-B | Cyclohexane | 17 | 46 | 29 | 24 | 2 |
| 12 | PSL-C | THF | 3 | 37 | >99 | 58 | >200 |

[a] Reactions were carried with 3 equiv of vinyl acetate at 30 °C.
[b] Conversion, $c = ee_S/(ee_S + ee_P)$.
[c] Determined by HPLC.
[d] Enantiomeric ratio, $E = \ln[(1-c)(1+ee_P)/\ln[(1-c)(1-ee_P)]$.
[e] Vinyl acetate was used as the solvent and as the acylating agent.

**Table 2.** Lipase-catalyzed acetylation of the *N*-benzyloxycarbonyl derivatives (±)-*cis*/*trans*-**2b**[a]

| Entry | Substrate | Lipase | Time (h) | *c* (%)[b] | $Ee_P$ (%)[c] | $Ee_S$ (%)[c] | *E* |
|---|---|---|---|---|---|---|---|
| 1 | (±)-*trans*-**2b** | CAL-B | 3 | 44 | 98 | 78 | >200 |
| 2 | (±)-*trans*-**2b** | PSL-C | 30 | 17 | 91 | 17 | 25 |
| 3 | (±)-*cis*-**2b** | CAL-B | 3 | 22 | >99 | 28 | >200 |
| 4 | (±)-*cis*-**2b** | PSL-C | 2 | 21 | >99 | 27 | >200 |

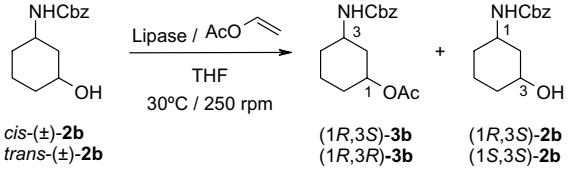
[a] Reactions were carried with 3 equiv of vinyl acetate at 30 °C.
[b] Conversion, $c = ee_S/(ee_S + ee_P)$.
[c] Determined by HPLC.

hydroxyl group to adopt the required equatorial position more easily and by this being able to react.

Next, we carried out the enzymatic acetylation of (±)-*cis*-**2b**, under the same conditions (Scheme 2). Entries 3 and 4 in Table 2 show the results obtained. With this substrate, both CAL-B and PSL-C displayed a slight drop in the reaction rate compared with the phthalimide derivative and with the Cbz-protected *trans* diastereoisomer. The enantioselectivity of both reactions was again very high (*E* >200).

In order to analyze the economic efficiency of these biocatalytic methods, we investigated the feasibility of reusing the immobilized lipase PSL-C. Even after six cycles the enantioselectivity remained very high (*E* >200), giving an excellent resolution in all the cycles (Table 3). A moderate decrease in the enzyme activity was seen during the sixth run, that is, the conversion was half in comparison to the fresh enzyme.

The absolute configuration of the biocatalytically prepared (1*R*,3*S*)-**2a**,**b** and (1*R*,3*S*)-**3b** has been tentatively assigned on the basis of two convergent criteria (a) the enantiotopic preference displayed by CAL-B in the transesterification of secondary alcohols following in all cases the Kazlauskas's rule;[14] (b) confirming the absolute configuration of the derivative (1*R*,3*R*)-**1** by comparison of its specific rotation sign with the literature data.[7]




*cis*-(±)-**2b**
*trans*-(±)-**2b**
(1*R*,3*S*)-**3b**
(1*R*,3*R*)-**3b**
(1*R*,3*S*)-**2b**
(1*S*,3*S*)-**2b**

**Scheme 2.** Lipase-catalyzed esterification of (±)-*cis*/*trans*-**2b**.

**Table 3.** Recycling of the lipase PSL-C in the enzymatic acylation of *cis*-**2a**[a]

| Cycle no | $Ee_S$ (%) | $Ee_P$ (%) | *c* (%) | *E* |
|---|---|---|---|---|
| 1 | 84.6 | >99 | 46 | >200 |
| 2 | 74.6 | >99 | 43 | >200 |
| 3 | 58.9 | >99 | 38 | >200 |
| 4 | 46.3 | >99 | 32 | >200 |
| 5 | 38.9 | >99 | 28 | >200 |
| 6 | 28.7 | >99 | 23 | >200 |

[a] PSL-C catalyzed esterification of with 10 equiv of vinyl acetate in THF at 30 °C and 5 h reaction.

## 3. Conclusion

In conclusion, we have developed a new and efficient route to enantiomerically pure *cis*- and *trans*-1,3-aminoalcohols via enzymatic transesterification using CAL-B and PSL-C as the biocatalysts in THF. This allows a direct access to all four stereoisomers of these interesting building blocks in high enantiomeric excesses.

## 4. Experimental

### 4.1. General

Lipase B of *C. antarctica* (CAL-B, 7300 U/g) was a gift from Novo Nordisk Co. Lipases from *Pseudomonas cepacea* PSL (>10 kU/g) and PSL-C (1019 U/g) were purchased from Amano Co. Lipase A from *C. antarctica* is commercially available as CHIRAZYME L-5 (CAL-A, 1 kU/g) from Roche Molecular Biochemicals. Lipase from porcine pancreas (PPL, 46 U/mg) was acquired from Sigma. Melting points were taken using a Gallenkamp apparatus and are uncorrected. Optical rotations were measured using a Perkin–Elmer 241 polarimeter. IR spectra were recorded on a Perkin–Elmer 1720-X FT spectrophotometer. $^1$H- and $^{13}$C NMR spectra were measured on a Bruker AC 300 ($^1$H, 300.13 MHz and $^{13}$C, 75.4 MHz) or a Bruker DPX-300 ($^1$H, 300.13 MHz and $^{13}$C, 75.4 MHz) using residual solvents peaks as the internal standard. Mass spectra were recorded on a Hewlett Packard 1100 Series. Flash column chromatography was performed using Merck silica gel 60 (230–400 mesh). The enantiomeric excesses were determined by chiral HPLC analysis on a Hewlett Packard or a Shimadzu LC liquid chromatograph. Two well resolved peaks were obtained for all racemic compounds (1.0 mg in 4 mL mobile phase; 20 μL sample).

**4.1.1. Typical procedure for the enzymatic acylation of the (±)-*N*-protected 3-aminocyclohexanols, (±)-*cis*/*trans*-2a and b.** To a mixture of the corresponding racemic alcohol (±)-*cis*/*trans*-**2a** and **b** (0.4 mmol) and CAL-B (50 mg), the organic solvent (8 mL) and vinyl acetate (103.2 mg, 3 equiv) were added under nitrogen atmosphere at room temperature. The resulting mixture

was shaken at 30 °C and 250 rpm. The progress of the reaction was monitored by TLC. As work-up, the enzyme was filtered off, washed with dichloromethane and the solvent evaporated under reduced pressure. The products were further purified by flash column chromatography of the residue (eluent: *n*-hexane/ethyl acetate 1:1 for *N*-phthalimide derivatives, *n*-hexane/diethyl ether 3:7 for the *N*-Cbz derivatives) to afford compounds (+)- or (−)-*cis*/*trans*-**3a** and **b** and the corresponding enantiomer of the remaining substrate *cis*/*trans*-**2a** and **b**.

**4.1.2. (1*R*,3*R*)-3-Aminocyclohexanol, (1*R*,3*R*)-1.**[7] $Na_2CO_3$ (14 mg, 0.14 mmol) was added to a solution of (1*R*,3*R*)-**3b** (40 mg, 0.14 mmol) in methanol (3 mL) and stirred at room temperature until disappearance of the starting material. Water was added (3 mL) and the mixture was extracted with dichloromethane (3 × 3 mL). The organic phases were dried ($Na_2SO_4$) and evaporated to afford (1*R*,3*R*)-**2b** (34 mg, 99%), which was used without further purification in the deprotection step. To a solution of (1*R*,3*R*)-**2b** (34 mg, 0.14 mmol) in methanol (3.8 mL) was added formic acid (0.134 mL) and Pd/C (19 mg). The mixture was stirred until the disappearance of the starting material (overnight). Filtration through Celite, washing the filter cake with methanol and evaporation afforded (1*R*,3*R*)-**1** (12 mg, 74%) as a pure white solid. $[\alpha]_D^{20} = -9.1$ (*c* 0.8, MeOH) 98% ee. $^1$H NMR $\delta$ 1.54–2.26 (m, 8H), 3.51 (m, 1H), 4.34 (br s, 1H), 8.75 (br s, 1H) ppm. $^{13}$C NMR $\delta$ 19.5, 31.5, 32.4, 38.0, 47.4, 66.2 ppm.

**4.1.3. (±)-*cis*-2-(3-Hydroxycyclohexyl)isoindole-1,3-dione, (±)-*cis*-2a.**[9] A slight modification of a reported method[9] was employed. NBS (1.8 g, 10.06 mmol) was added to a stirred solution of 2-cyclohex-2-enyl-3a,7a-dihydroisoindole-1,3-dione (1.7 g, 7.5 mmol) in chloroform (54 mL) and water (0.2 mL, 11.11 mmol) and the mixture was stirred in the darkness at room temperature for 4 h, after which was washed with 1 M solution of thiosulfate (70 mL) and the aqueous phase was re-extracted with dichloromethane (50 mL). The combined organic phases were dried ($Na_2SO_4$), evaporated and further purified by flash column chromatography (eluent: *n*-hexane/diethyl ether 1:1), to give the corresponding orthoamide (1.98 g, 87%), which was used in the dehalogenation step (avoiding the hydrolysis of the acetal) to afford the final phthalimide (±)-*cis*-**2a** (1.04 g, 73%) according to the literature procedure.[9]

**4.1.3.1. 2-((1*R*,3*S*)-3-Hydroxycyclohexyl)isoindole-1,3-dione, (1*R*,3*S*)-2a.** White solid. Yield 50%. $[\alpha]_D^{20} = +41.9$ (*c* 0.6, $CHCl_3$) 95% ee. HPLC conditions: Column Chiralcel OD, hexane/propan-2-ol, 95:5, 0.6 mL/min, 25 °C, $t_R$ (1*R*,3*S*) 38.9 min and $t_R$ (1*S*,2*R*) 43.5 min. $^1$H NMR $\delta$ 1.22–1.50 (m, 2H), 2.31–2.55 (m, 7H), 3.69 (m, 1H), 4.15 (tt, $J = 3.9$ and 12.3 Hz, 1H), 7.69 (dd, $J = 2.9$ and 5.6 Hz, 2H), 7.82 (dd, $J = 3.1$ and 5.0 Hz, 2H) ppm. $^{13}$C NMR $\delta$ 22.3, 28.5, 34.5, 38.7, 48.5, 69.9, 123.1, 131.9, 133.8, 168.2 ppm. ESIMS *m*/*z* 268.0 [(M+Na)$^+$, 100], [(M+1)$^+$, 20]. IR 3385, 1763, 1704 cm$^{-1}$.

**4.1.4. (±)-*trans*-2-(3-Hydroxycyclohexyl)isoindole-1,3-dione, (±)-*trans*-2a.** Chloroacetic acid (246.4 mg, 2.6 mmol) and $PPh_3$ (681.9 mg, 2.6 mmol) were added to a stirred solution of (±)-*cis*-**2a** (580 mg, 2.37 mmol) in dried THF (30 mL) at 0 °C under nitrogen and left to stir until complete dissolution of the solids. Then, DEAD (0.454 mL, 2.6 mmol) was added dropwise waiting until the disappearance of the yellow colour of the solution before adding the next drop. After stirring for 1.5 h at 0 °C, the reaction was concentrated and purified by flash column chromatography (eluent: *n*-hexane/diethyl ether 1:1) to afford the corresponding chloroacetate in 71% yield. The product was dissolved in methanol (13.5 mL) and $Na_2CO_3$ (172 mg, 1.67 mmol) was added and left to stir for 2 h at room temperature. The solvent was removed, the crude was dissolved in water (10 mL) and extracted with dichloromethane (3 × 10 mL), the organic phases were dried ($Na_2SO_4$) and evaporated. The residue was again dissolved in dichloromethane (10 mL) and filtered through silica affording a clear solution that was evaporated to give (±)-*trans*-**2a** (406.4 mg, 1.66 mmol 99%). White solid. $^1$H NMR $\delta$ 1.41–1.98 (m, 7H), 2.19 (dq, $J = 4.2$ and 12.3 Hz, 1H), 2.51 (dt, $J = 2.5$ and 13.1 Hz, 1H), 4.32 (m, 1H), 4.61 (tt, $J = 3.7$ and 15.5 Hz, 1H), 7.69 (dd, $J = 3.1$ and 5.2 Hz, 2H), 7.81 (dd, $J = 3.1$ and 5.4 Hz, 2H) ppm. $^{13}$C NMR $\delta$ 19.6, 29.6, 31.7, 36.0, 45.4, 65.7, 122.7, 132.1, 134.0, 168.0 ppm. ESIMS *m*/*z* 268.0 [(M+Na)$^+$, 100]. IR 3451, 1703, 1462 cm$^{-1}$.

**4.1.5. (±)-*cis*-(3-Hydroxycyclohexyl)carbamic acid benzyl ester, (±)-*cis*-2b.** Hydrazine hydrate (42 mg, 0.8 mmol) was added to a solution of (±)-*cis*-**2a** (200 mg, 0.8 mmol) in methanol (10 mL) and the solution was heated at reflux overnight. The reaction was cooled to room temperature and concentrated. The residue was filtered off and washed with chloroform (10 mL) and dichloromethane (20 mL). The organic phases were evaporated to give the free aminoalcohol as a white solid, which was protected without any further purification. This solid was dissolved in water (3.5 mL) and cooled to 0 °C, to which $Na_2CO_3$ (163.5 mg, 1.6 mmol) and CbzCl (0.288 mL, 1.7 mmol) were added. After stirring overnight at room temperature, the reaction was extracted with dichloromethane (3 × 10 mL), concentrated and purified by flash column chromatography (eluent: *n*-hexane/diethyl ether, 1:1) giving (±)-*cis*-**2b** as a white solid (161 mg, 81%).

**4.1.5.1. ((1*R*,3*S*)-3-Hydroxycyclohexyl)carbamic acid benzyl ester, (1*R*,3*S*)-2b.** White solid. Melting point 95–97 °C. Yield 70%. $[\alpha]_D^{20} = -11.0$ (*c* 0.52, $CHCl_3$) 28% ee. HPLC conditions: Column Chiralcel OD hexane/ethanol, 90:10, 0.8 mL/min, 40 °C, $t_R$ (1*R*,3*S*) 17.6 min and $t_R$ (1*S*,3*R*) 10.1 min. $^1$H NMR $\delta$ 1.10–1.4 (m, 4H), 1.50–1.95 (m, 4H), 2.13–2.17 (br d, $J = 11.6$ Hz, 1H), 3.61 (m,

1H), 5.08 (br s, 2H), 7.36 (m, 5H) ppm. $^{13}$C NMR $\delta$ 20.3, 31.9, 34.2, 41.0, 48.0, 66.5, 68.7, 128.1, 128.1, 128.5, 136.5, 155.5 ppm. ESIMS $m/z$ 272 [(M+Na)$^+$, 100], 288 [(M+K)$^+$, 10], 250 [(M+1)$^+$, 15]. IR 3412, 3338, 1693 cm$^{-1}$.

**4.1.6. (±)-*trans*-(3-Hydroxycyclohexyl)carbamic acid benzyl ester, (±)-*trans*-2b.** The compound (±)-*trans*-**2b** was prepared from (±)-*trans*-**2a** following the same method applied for the preparation of (±)-*cis*-**2b**. Yield 64% from (±)-*trans*-**2a**.

**4.1.6.1. ((1*S*,3*S*)-3-Hydroxycyclohexyl)carbamic acid benzyl ester, (1*S*,3*S*)-2b.** White solid. Melting point 118–119 °C. Yield 51%. $[\alpha]_D^{20} = +4.7$ ($c$ 0.5, $CHCl_3$) 78% ee. HPLC conditions: Column Chiralcel OD hexane/ethanol, 90:10, 0.8 mL/min, 35 °C, $t_R$ (1*R*,3*S*) 11.1 min and $t_R$ (1*S*,3*R*) 14.7 min. $^1$H NMR $\delta$ 1.40–2.10 (m, 8H), 3.70 (br d, $J = 3.2$ Hz, 1H), 4.01 (m, 1H), 4.20 (m, 1H), 5.17 (m, 2H), 6.30 (br s, 1H), 7.49–7.51 (m, 5H) ppm. $^{13}$C NMR $\delta$ 18.9, 31.5, 32.0, 39.0, 45.7, 45.8, 65.8, 66.3, 127.6, 127.8, 128.2, 136.2, 156.0 ppm. ESIMS $m/z$ 272 [(M+Na)$^+$, 100], 288 [(M+K)$^+$, 5], 250 [(M+1)$^+$, 5]. IR 3332, 1689, 1540 cm$^{-1}$.

**4.1.7. Acetic acid (1*R*,3*S*)-3-(1,3-dioxo-1,3-dihydroisoindol-2-yl)cyclohexyl ester, (1*R*,3*S*)-3a.** White solid. Melting point 95–96 °C. Yield 45%. $[\alpha]_D^{20} = +28.2$ ($c$ 0.5, $CHCl_3$) >99% ee. HPLC conditions: Column Chiralcel OD hexane/propan-2-ol, 95:5, 0.6 mL/min, 20 °C, $t_R$ (1*R*,3*S*) 14.0 min and $t_R$ (1*S*,3*R*) 16.0 min. $^1$H NMR $\delta$ 1.33–1.41 (m, H), 1.64–2.20 (m, 8H), 2.32 (q, $J = 11.6$ Hz, 4.17 (tt, $J = 12.5$ and 3.9 Hz, 1H), 4.72 (m, 1H), 7.66 (dd, $J = 2.7$ and 5.2 Hz, 2H), 7.77 (dd, $J = 3.1$ and 5.1 Hz, 2H) ppm. $^{13}$C NMR $\delta$ 21.3, 22.1, 28.6, 30.6, 34.7, 48.1, 71.7, 123.1, 131.8, 133.9, 168.1, 170.4 ppm. ESIMS $m/z$ 310 [(M+Na)$^+$, 100], 326 [(M+K)$^+$, 5], 288 [(M+1)$^+$, 10]. IR 1770, 1722, 1704 cm$^{-1}$.

**4.1.8. Acetic acid (1*R*,3*S*)-3-benzyloxycarbonylamino cyclohexyl ester, (1*R*,3*S*)-3b.** White solid. Melting point 113–115 °C. Yield 20%. $[\alpha]_D^{20} = +9.1$ ($c$ 0.45, $CHCl_3$) >99% ee. HPLC conditions: Column Chiralcel OD hexane/ethanol, 100:0 to 95:5 in 80 min, 0.8 mL/min, 40 °C, $t_R$ (1*R*,3*S*) 48.7 min and $t_R$ (1*S*,3*R*) 50.3 min. $^1$H NMR $\delta$ 1.10–1.50 (m, 5H), 1.72–1.98 (m, 3H), 2.03 (s, 3H), 2.19–2.23 (m, 1H), 3.63 (m, 1H), 4.78 (m, 2H), 5.08 (br s, 2H), 7.35 (m, 5H) ppm. $^{13}$C NMR $\delta$ 20.7, 21.3, 30.7, 38.0, 66.6, 71.0, 128.1, 136.5, 155.4, 170.2 ppm. ESIMS $m/z$ 314 [(M+Na)$^+$, 100], 330 [(M+K)$^+$, 5], 292 [(M+1)$^+$, 5]. IR 3320, 1729, 1682 cm$^{-1}$.

**4.1.9. Acetic acid (1*R*,3*R*)-3-benzyloxycarbonylamino cyclohexyl ester, (1*R*,3*R*)-3b.** White solid. Melting point 120–125 °C. Yield 40%. $[\alpha]_D^{20} = -9.6$ ($c$ 0.13, $CHCl_3$), 98% ee. As the title compound could not be resolved it was hydrolyzed and analyzed as (1*S*,3*S*)-**2b**. $^1$H NMR $\delta$ 1.20–1.39 (m, 1), 1.40–1.78 (m, 6H), 1.90–2.17 (m, 5H), 3.91 (m, 1H), 4.60 (m, 1H), 5.11 (m, 3H), 7.36 (m, 5H) ppm. $^{13}$C NMR $\delta$ 19.7, 21.2, 29.6, 32.4, 36.9, 46.4, 66.7, 69.6, 128.5, 125.5, 136.7, 170.3, 178.6 ppm. ESIMS $m/z$ 314 [(M+Na)$^+$, 100], 315. IR 3336, 1731, 1532 cm$^{-1}$.

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